Using Probabilistic Monitoring Data to Validate the Non-Coastal Virginia Stream Condition Index

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1.0 EXECUTIVE SUMMARY

Biological monitoring of streams and rivers is an integral component of the water quality monitoring program in the Commonwealth of Virginia. Biological monitoring allows the Virginia Department of Environmental Quality (VDEQ) to assess ecological condition. Biological surveys are used to answer the questions of whether these waterbodies support survival and reproduction of desirable aquatic species and if the waterbodies meet their designated aquatic life uses.

In 2000, the United States Environmental Protection Agency (USEPA) contracted Tetra Tech to develop a multimetric macroinvertebrate index for the Commonwealth of Virginia. This index contains eight core metrics that when calculated into one number is known as the Virginia Stream Condition Index (VSCI) (Tetra Tech, 2003). Tetra Tech developed the VSCI using Virginia's existing biomonitoring database (data collected 1994-2001). This dataset contained a significant amount of upstream (reference) control sites for use with the Rapid Bioassessment Protocols (USEPA, 1999). Reference sites in the Central Appalachian Ecoregion, Piedmont Ecoregions and headwater streams were limited. This report focuses on validating the VSCI and proposing assessment recommendations.

Using a new, independent probabilistic monitoring program (ProbMon) database (number of stations =350, 2001-2004), VDEQ has validated the VSCI using a spatially diverse (ecoregionally and stream size) data set free of psuedoreplication. These probabilistic data sets have allowed VDEQ to narrow data gaps and test the proposed VSCI against many classification variables, which include season, stream size, ecoregion, bioregion, river basin, regional office, and sampling technique. VDEQ also reviewed the recommended best standard values for the eight core metrics. These metrics include EPT taxa, Total taxa, % Ephemeroptera, % Plecoptera + Trichoptera (-Hydropsychidae), % Chironomidae, % Top 2 Dominant Taxa, Modified Family Biotic Index (MFBI), and % Scrapers. For a detailed explanation of multimetric index development, metric definitions, and an overview of bioassessment, please review the document 'A Stream Condition Index for Virginia Non-Coastal Streams' at http://www.deq.virginia.gov/watermonitoring/pdf/vastrmcon.pdf.

Reviewing the probabilistic biological data has confirmed that the VSCI works well to discriminate between sites with acceptable water quality and habitat versus sites with degraded water quality and habitat. Potential seasonal, ecoregion, bioregion, basin size, and sampling method patterns were found in Nonmetric Multidimensional Scaling ordination results. However, Mean Similarity statistical results indicated that these patterns have low classification strength. These patterns were further tested for environmental significance using Principal Components Analysis (PCA) to evaluate metric clustering by classification category. PCA failed to display any clustering of metrics by classification. Box-and-whisker plots of the metrics and the VSCI were used to graphically determine any impacts of these classifications on the reference stations. Individual metric differences were noted by classification category. However, differences in individual core metrics did not affect the overall VSCI score. The median values for the VSCI in the box-and-whisker plots were similar and the interquartile ranges of the VSCI scores in all of the graphs were above 60. Current results do not support calibrating the VSCI by season, sampling method, bioregion, or basin size. Recommending new best standard values for calculating the VSCI is not warranted at this time, but will be reviewed periodically as new data becomes available. Results of calibrating the best standard values by bioregion and/or season are predictable. Mountain stream VSCI scores are slightly lower and Piedmont VSCI scores are slightly higher. The same overall percentage of streams would be designated impaired because the calibrated reference sites adjust in the same manner; subsequently, lowering or raising the assessment cutoff.

Aquatic Life Use (ALU) tiers established using the Virginia SCI

VDEQ considers VSCI scores above the 10th percentile of reference distribution to be similar to the bulk of reference sites and not impaired. Sites with VSCI scores below the 10th percentile of reference distribution are not similar to most natural streams and considered degraded. Other states with rigorously tested bioassessment methods use similar percentile of reference distribution methods to determine biological integrity (Ohio; Yoder and Rankin 1995, Florida; Barbour et al. 1996).

The 10th percentile from the probabilistic data set was 58.5 and the 10th percentile from Tetra Tech's analysis of targeted data was 61.3. The average 10th percentile cutoff from both data sets is 59.9. To keep the assessment cutoff simple, the impairment threshold was rounded to 60.

The precision of the VSCI was estimated to be +/- 7.9 scoring units on a 100 point scale. This precision was calculated using data from stations with replicate samples that were collected on the same day.

Aquatic life use tiers were established above and below the impairment threshold (Tables E-1 and 12) based on the average 50th percentile scores from the TetraTech reference dataset and the ProbMon reference dataset (upper tier); and the TetraTech stressed dataset and ProbMon stressed dataset (lower tier) (Table 13). The aquatic life use tiers that can be discerned using the Virginia SCI is shown in Figure 29. Integrated report assessment decisions and methodology for aquatic life use using the VSCI will be found in VDEQ's Water Quality Assessment Guidance Manual.

Table E-1. Virginia SCI scores and associated aquatic life use (ALU) tiers.

VSCI Score	ALU Tiers
$\leq$ 42	Severe Stress
43 - 59	Stress
60 - 72	Good
$\geq$ 73	Excellent

2.0 INTRODUCTION

In 2000, the United States Environmental Protection Agency (USEPA) contracted Tetra Tech to develop a multimetric macroinvertebrate index for the Commonwealth of Virginia. This index contains eight core metrics that are collectively known as the Virginia Stream Condition Index (VSCI) (Tetra Tech, 2003). Tetra Tech developed the VSCI using Virginia's existing biomonitoring database, which contained a significant amount upstream control sites for use with the Rapid Bioassessment Protocols (USEPA, 1999). Reference sites in the Central Appalachian Ecoregion, Piedmont Ecoregions and headwater streams were limited. This report focuses on validating the VSCI (confirming that VSCI works as documented in the Tetra Tech report) and proposing assessment recommendations.

Using a new, independent probabilistic monitoring program (ProbMon) database (number of stations =350, 2001-2004), VDEQ has validated the VSCI using a spatially diverse (ecoregionally and stream size) data set free of psuedoreplication. Virginia also used data from West Virginia's probabilistic program that met the reference filter criteria to evaluate additional reference sites located in the Central Appalachian Ecoregion. These probabilistic data sets have allowed VDEQ to narrow data gaps and to evaluate the proposed VSCI by classification variables such as season, stream size, ecoregion, bioregion, river basin, regional office, and sampling technique. Classification variable refers to grouping similar sampling stations. For example, grouping all the sites in one bioregions or all the sites sampled in one season. VDEQ also reviewed the recommended best standard values for the eight core metrics. These metrics include EPT taxa, Total taxa, % Ephemoeroptera, % Plecoptera + Trichoptera (-Hydropsychidae), % Chironomidae, % Top 2 Dominant Taxa, Modified Family Biotic Index (MFBI), and % Scrapers. For a detailed explanation of multimetric index development, metric definitions, and an overview of bioassessment, please review 'A Stream Condition Index for Virginia Non-Coastal Streams' http://www.deq.virginia.gov/watermonitoring/pdf/vastrmcon.pdf.

Probabilistic monitoring is the monitoring of stations selected using a probabilistic criterion. These monitoring stations are generated by a computer program that randomly chooses monitoring sites on rivers and streams throughout Virginia. Probabilistic monitoring sampling points were generated by the United States Environmental Protection Agency using a sampling design similar to USEPA's Environmental Mapping and Assessment Program (EMAP). VDEQ samples 50 to 60 randomly selected stations per year throughout the Commonwealth for a variety of chemical, biological, and habitat parameters. ProbMon sites are monitored in both spring and fall and include streams and rivers of various sizes and different geographic regions. For more information please see: www.deq.virginia.gov/probmon.

Reference and stress filters (Table 1-3) were developed to screen large amounts of data and to define the least disturbed condition and stressed conditions found in Virginia streams. The reference and stress filters use data from land cover, water quality, and habitat surveys. The habitat results have unitless metrics and are based on EPA's RBP habitat assessment protocol which uses visual observations. The values used to screen for reference condition were from a variety of sources (Miltner 1998, USEPA 1999, USEPA 2000, Dodd 2000, Ohio EPA 1999, Boward 1999, Carle 2005, Wang 2003) and VDEQ data analysis (Tetra Tech 2003 and VDEQ 2005). Best Professional Judgment (BPJ) was used to eliminate candidate sites that agency biologists knew were not reference (n=8) or stress (n=30) condition from their site specific watershed knowledge.

Table 1. List of Piedmont Bioregion reference filters.

Piedmont Reference Filter	
% Urban	< 5%
Total Nitrogen	< 1.5 mg/L
Total Phosphorus	< 0.05 mg/L
Specific Conductance (uS/cm)	< 250
Dissolved Oxygen	> 6 mg/L
pH	> 6 or < 9
Channel Alteration	> 11
Epifaunal Substrate/Cover	> 11
Riparian Vegetative Zone	> 11
Total Habitat	> 140

Table 2. List of Mountain Bioregion reference filters.

Mountain Reference Filter	
% Urban	< 5%
Total Nitrogen	< 1.5 mg/L
Total Phosphorus	< 0.05 mg/L
Specific Conductance (uS/cm)	< 250
Dissolved Oxygen	> 6 mg/L
pH	> 6 or < 9
Channel Alteration	> 11
Epifaunal Substrate/Cover	> 11
Embeddedness	> 11
Riparian Vegetative Zone	> 11
Total Habitat	> 140

Table 3. List of stress filters all ecoregions.

Stress Filter	
% Urban	> 10%
Total Nitrogen	> 3 mg/L
Total Phosphorus	> 0.1 mg/L
Specific Conductance (uS/cm)	> 500
Riparian Vegetative Zone	< 6
Total Habitat	< 120

VDEQ identified 60 (sample n=104) new reference sites and 33 (sample n=64) stressed sites from the probabilistic data sets for this validation study. The breakdown of the reference and stressed sites by season and ecoregion is found in Tables 5 and 6. A map of the stations by ecoregion is found in Figure 1.

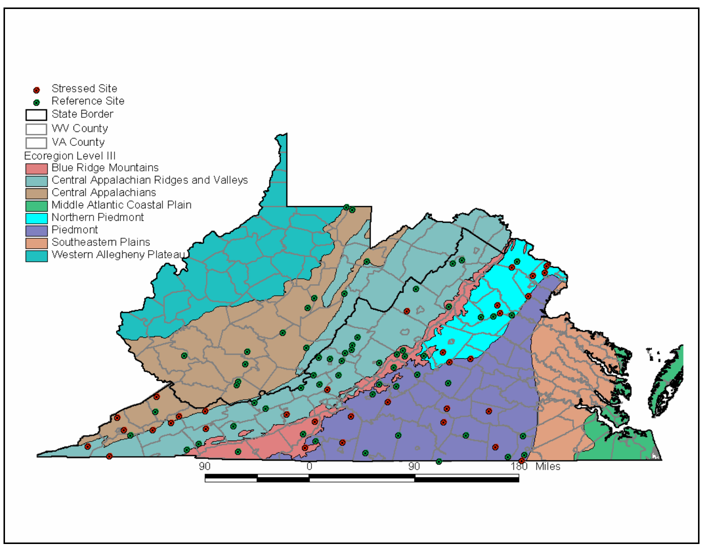
Table 4. Filter results for reference sites by ecoregion and season.

Reference Sites (Number of samples =104)			
	Mountain	Piedmont	West Virginia Mountain
Spring	29	18	9
Fall	28	15	5
Total	57	33	14

Table 5. Filter results stresses sites by ecoregion and season.

Stressed Sites (Number of samples = 64)		
	Mountain	Piedmont
Spring	15	18
Fall	15	16
Total	30	34

Figure 1. Map of reference (n=60) and stressed (n=33) stations. Stations from West Virginia Department of Environmental Protection were included due to limited data in Virginia's section of the Central Appalachian Ecoregion.



3.0 METHODS

The data used in this study were collected according to VDEQ standard operating procedures set forth by the Ambient Monitoring Program http://www.deq.virginia.gov/watermonitoring/pdf/wqmsop.pdf, Biological Monitoring Program Quality Assurance Project Plan for Wadeable Streams and Rivers (VDEQ 2006) and Probabilistic Monitoring Program http://www.deq.virginia.gov/probmon/pdf/report1.pdf.

Probabilistic data is stored in a Microsoft Access database. Land cover, nutrient, and field data tables were queried using the reference and stress filters described in the introduction. Stations categorized by reference and stress query conditions were reviewed and verified by regional biologists. The final data sets of reference and stress stations were compiled in Microsoft Access tables for use in PC-ORD Version 4 (McCune and Medford 1999), MeanSim (Van Sickle 1997), SYSTAT11, SAS, R (R Development Team 2006), and ArcView 3.2.

The reference taxa data were graphically reviewed using Nonmetric Multidimensional Scaling (NMS) ordination and statistically explored for differences using the Mean Similarity (MeanSim) program. Significant classification differences would result in recommending recalibration of the VSCI for assessment purposes. These classification differences were evaluated for environmental significance using Principal Components Analysis (PCA) and box-and-whisker plots. Best Standard Values (BSV) from the targeted Ecological Data Application System (EDAS) database and the probabilistic database were compared. Reference and stress box-and-whisker plots were generated to recommend an assessment value and to assess the accuracy of the VSCI (the percent of correctly assessed stations). Precision of the VSCI was tested using replicate stations. Cumulative distribution functions were generated for the entire non-coastal area of Virginia to understand the variability in the population associated with the core metrics and VSCI.

Using PC-ORD, NMS was used to graphically evaluate patterns in the reference community data by bioregion, ecoregion, season, stream order, basin size, VDEQ regional office (including West Virginia data), collection method, and river basin. Two bioregions were defined in Virginia, one bioregion is all of the mountain ecoregions (Blue Ridge Mountain, Central Appalachian Ridge and Valley, and Central Appalachian) and the other bioregion is all of the piedmont ecoregions (Piedmont and Northern Piedmont). NMS ordinations were used to graphically explore the reference community data to find patterns. The NMS graphs presented in this report were generated by Log (X+1) transforming the reference taxa. Rare taxa were not excluded from the analysis.

Graphical patterns were further explored using the MeanSim analysis program. MeanSim software and documentation is provided free of charge on EPA's website http://www.epa.gov/naaujydh/pages/models/dendro/meansim6.htm. MeanSim analysis is based on a matrix of pairwise similarities for all possible pairs of objects (Van Sickle 1997). VDEQ used a Bray-Curtis similarity input matrix for all MeanSim analysis. MeanSim analysis was performed by bioregion, ecoregion, season, basin size, and collection method. The program outputs Within Group (W) similarity and Between Group (B) similarity. Classification strength (CS) was estimated by subtracting B from W (CS=W-B). M is a method of exploring the between classification variability and within class variability. M was calculated by dividing B by W (M=B/W). If the 'no class structure' hypothesis is true, then M should yield a value close to 1. The MeanSim program outputs a p-value based on 10,000 permutations of the input matrix. Information provided from MeanSim is used to evaluate the strength of classification categories. It is important to determine the environmental significance of MeanSim results. Environmental significance occurs when a classification variable significantly affects one or more of the core metrics in such a way that the distribution of VSCI reference scores is significantly changed.

Environmental significance was tested using Principle Components Analysis (PCA) and box-and-whisker plots. Using the eight core metrics from reference stations, PCA plots were produced in PC-ORD and evaluated by season, bioregion, bioregion + season, ecoregion, basin size, and sampling method. Richness metrics were log transformed and the proportion metrics were arcsine transformed in the input matrix. PCA graphs were evaluated to identify clusters of metrics by classification categories. Box-and-whisker plots of the eight core metrics and the VSCI from reference stations were graphically evaluated by season, bioregion, bioregion + season, ecoregion, basin size, and sampling method to determine if these classifications impacted individual core metric values and final VSCI scores.

Cumulative distribution function (CDF) plots were generated using the R program. The CDF estimates the probability that a variable is less than or equal to some value. This function is most useful when displayed graphically. The analyst is able to determine the likelihood that a variable would be less than a particular threshold. It can also provide the probability that a variable would be above a threshold or if it would be within a certain range. For probabilistic data used in this validation, these probabilities apply to non-tidal streams found in the mountain and piedmont ecoregions. Detailed information on CDF curve generation can be found at the following EPA website: http://www.epa.gov/nheerl/arm/analysispages/monitanalysisinfo.htm.

CDF plots were created from the probabilistic data set to generate best standard values for each recommended core metric value and the VSCI. These probabilistic best standard values were compared to the recommended best standard values from Tetra Tech's report, which used targeted stations found in EDAS.

Box-and-whisker plots for reference and stressed sites and their corresponding percentile values were generated using the SYSTAT11 program. Percentiles were used to determine assessment values for this data set. VSCI accuracy (the percent of correctly assessed stations) was determined using this information. VSCI precision was calculated using replicate stations. Precision was calculated by calculating the root mean square error (RMSE), which estimates sampling error associated with a method. The VSCI precision was estimated by determining the 90% confidence interval (1.645 X RSME) (Maxted 2000). The last measure of variability was the signal-to-noise ratio (S/N) which estimated the variance among all sites to the replicate sites

(Kauffman 1999). Larger S/N ratio indicates lower relative variability. The S/N ratio was calculated in SAS. Maps were produced using ArcView 3.2.

4.0 DATA RESULTS

NMS and MeanSim results are presented first, followed by PCA and box-and-whisker plots which evaluate environmental significance of select classification categories. Next, best standard value assessment results from CDF curves for the core metrics and the VSCI are shown. Finally, box-and-whisker plots with corresponding reference site percentiles are presented.

4.1 NMS ordination results

NMS graphs were produced by running PC-ORD NMS 'slow and thorough setting' on Autopilot Mode with a Log (X+1) transformation to produce the following tables and graphs. The final stress (Table 6) was 17.49 and accounted for 80.6% of the variation (Table 7). Final stress is an important measure of confidence in the ordination results. Typically a final stress less than 20 indicates the ordination is providing reasonable results.

Table 6. Stress in relation to dimensionality (number of axes).

	Stress in real data 40 run(s)			Stress in randomized data Monte Carlo test, 50 run			
Axes	Minimum	Mean	Maximum	Minimum	Mean	Maximum	p
1	37.530	44.893	57.177	49.390	52.635	57.175	0.0196
2	22.982	23.868	41.250	31.892	32.835	34.425	0.0196
3	17.493	17.935	18.413	23.715	24.461	25.676	0.0196
4	14.247	14.341	14.886	18.938	19.598	20.492	0.0196
5	11.981	12.061	12.511	15.661	16.295	16.851	0.0196
6	10.235	10.334	10.783	13.297	13.854	14.328	0.0196

p = proportion of randomized runs with stress < or = observed stress

Table 7. Variation explained by axis (r-squared).

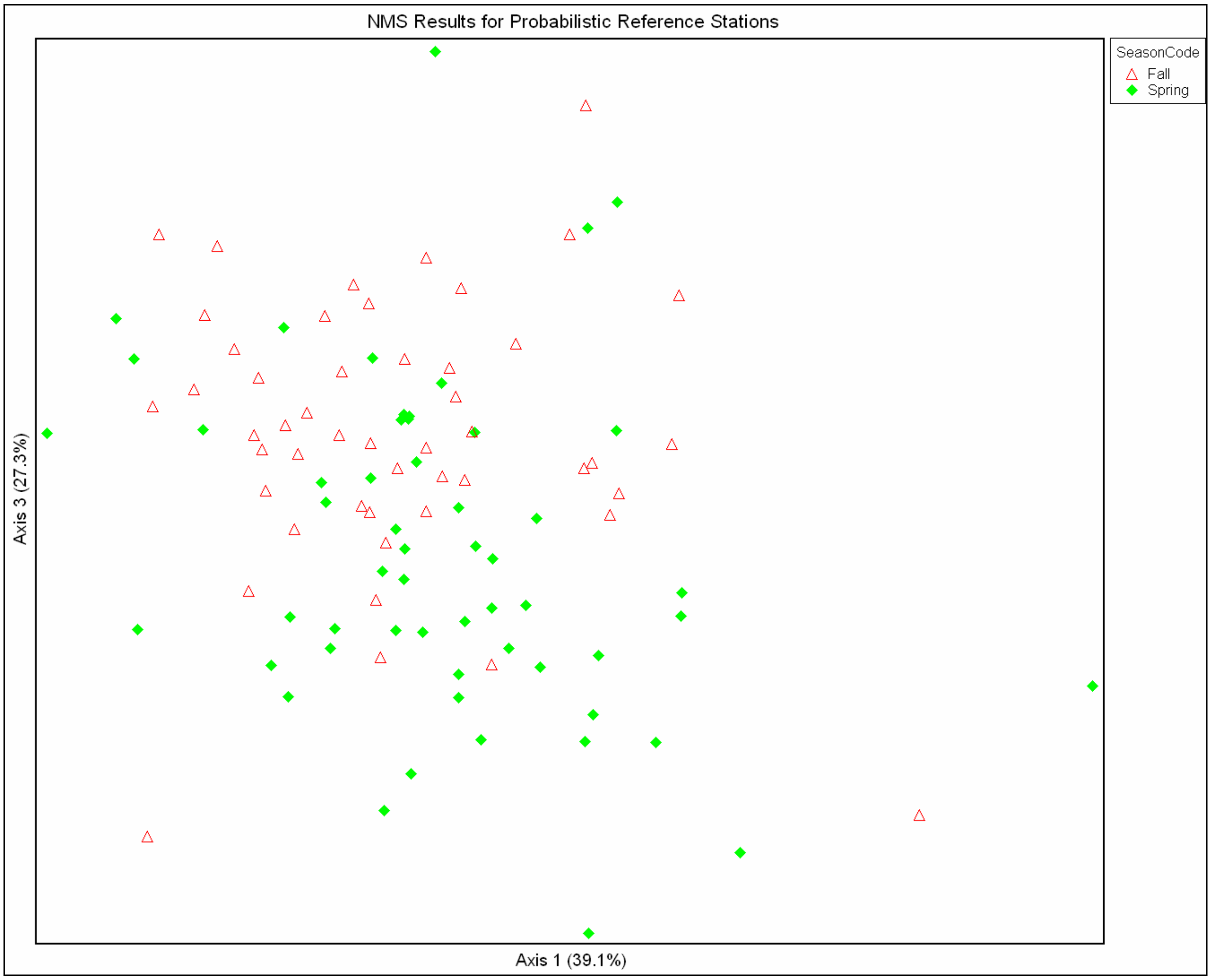
Axis	Increment	Cumulative
1	.391	.391
2	.143	.533
3	.273	.806

Figures 2-9 contain the NMS ordination results. Only graphs of axis 1 and 3 are presented in this report since axis 1 and 3 explain the greatest amount of variation (Table 7). The points on each of these graphs represent individual reference sites.

Season (Figure 2), ecoregion (Figure 3), bioregion (Figure 4), bioregion and season (Figure 5), basin size (Figure 6), and sampling method (Figure 8) contain potential clustering of reference sites and were further evaluated for statistical and environmental significance with Mean

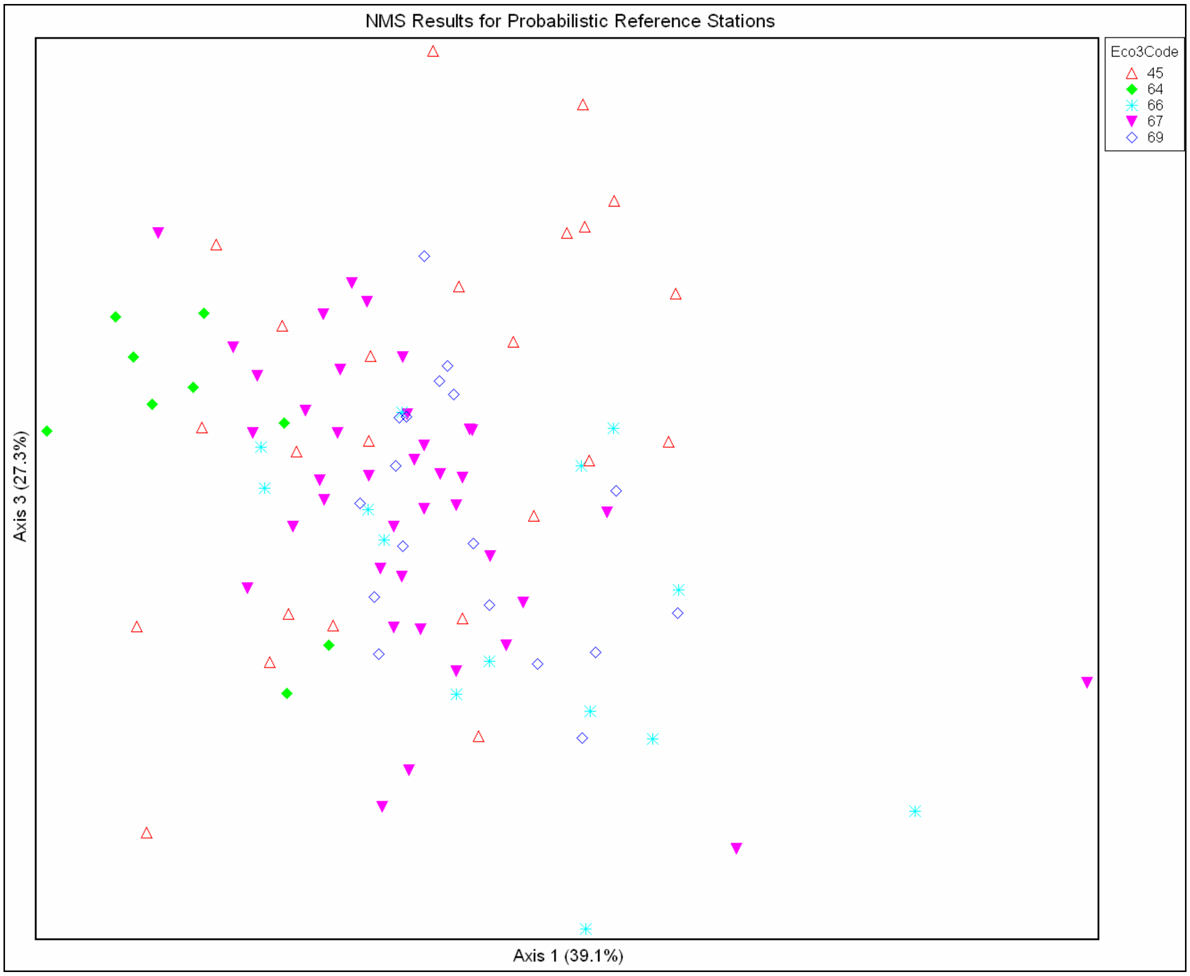
Similarity (Table 8, Appendix A), Principal Components Analysis (Figures 10-15), and box-and-whisker plots (Figures 16-21). The VDEQ regional office (Figure 7) and river basin (Figure 9) ordinations do not contain any potential patterns.

Figure 2. NMS results by season (n=104).



The open red triangles represent reference stations sampled in the fall season and the solid green diamonds represent reference stations sampled in the spring season. A slight clustering of stations was noted in the spring data with the majority of stations being in the lower left.

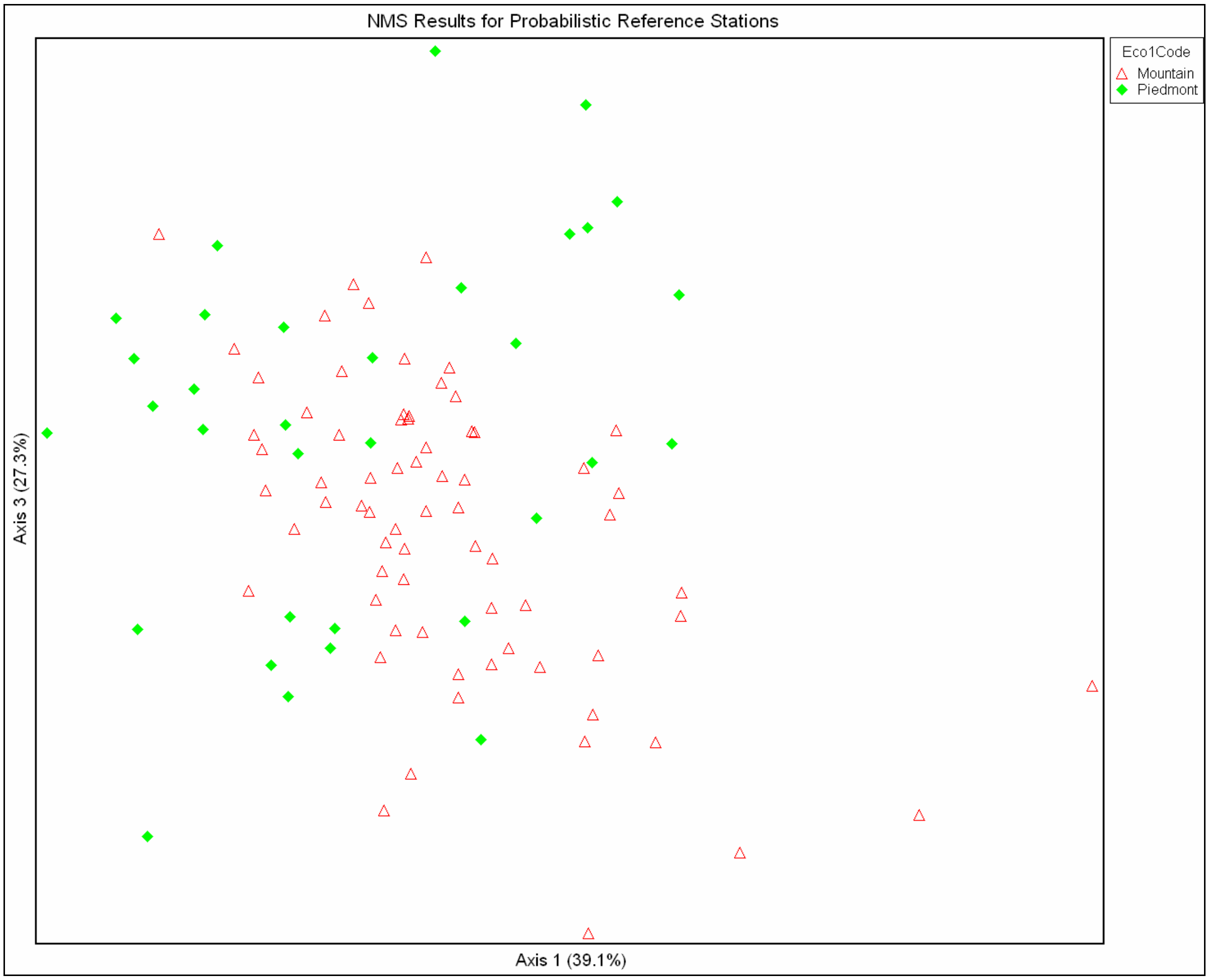
Figure 3. NMS results by ecoregion (n=104).



The open red triangles represent reference stations sampled in ecoregion 45 (Piedmont), the solid green diamonds represent reference stations sampled in ecoregion 64 (Northern Piedmont), the blue asterisk represent reference stations sampled in ecoregion 66 (Blue Ridge Mountains), the upside down solid pink triangles represent reference stations sampled in ecoregion 67 (Central Appalachian Ridge and Valley), and the open blue diamonds represent stations sampled in ecoregion 69 (Central Appalachians).

Clustering was observed with two separate groups of Piedmont stations with one small group in the upper right and another in the lower left. Examination of individual stations determined that these groups were separated due to stream slope (i.e., low versus high gradient streams within the same ecoregion. Slight grouping and separation of stations was also observed within the Central Appalachian Ridge and Valley stations. This separation was also due to stream slope with the larger, low gradient valley streams separating from the smaller high gradient streams. Therefore, it appears that benthic communities from streams with high gradient were possibly different from those in streams with low gradient.

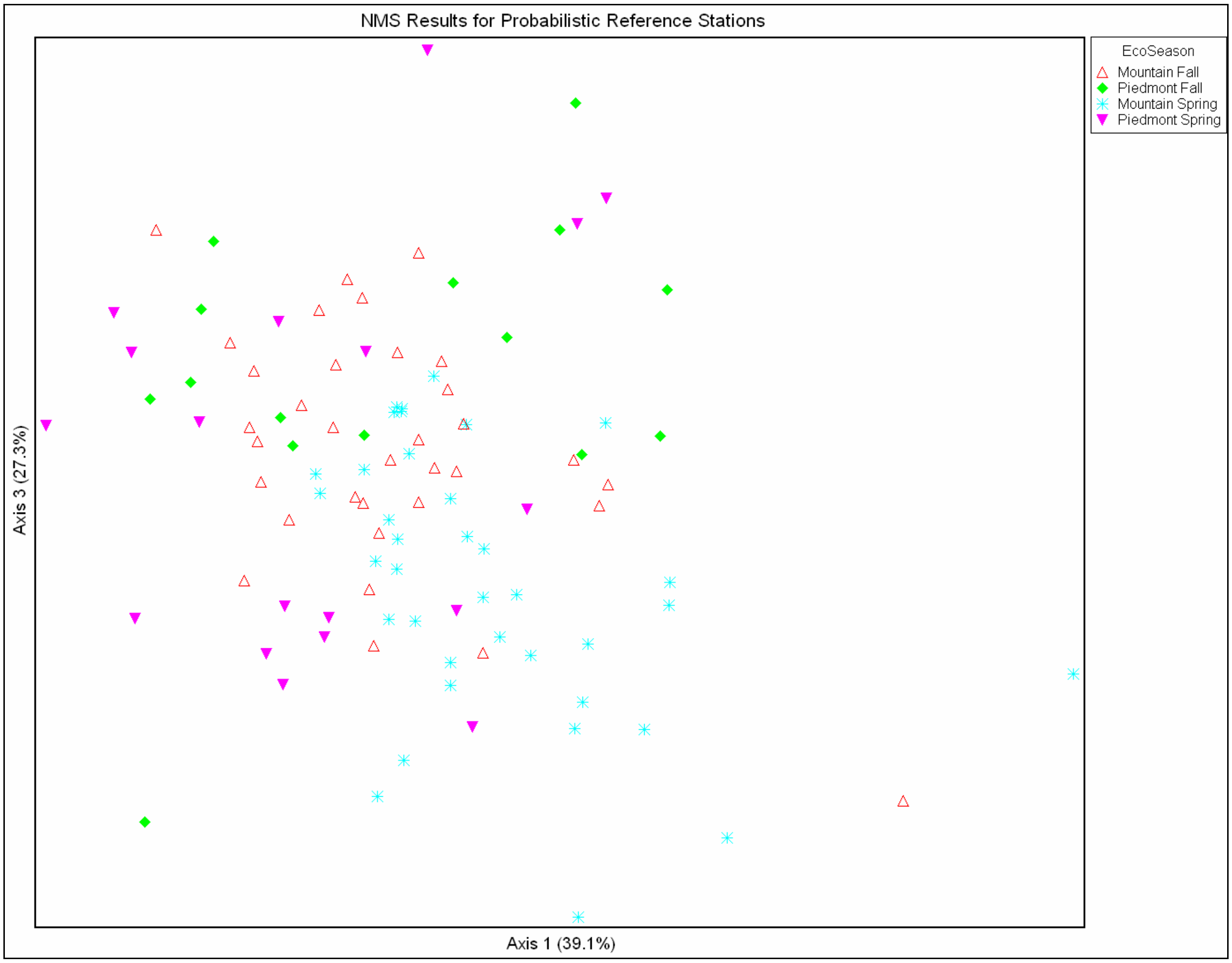
Figure 4. NMS results by bioregion (n=104).



The open red triangles represent reference stations sampled in the Mountain Bioregion and the solid green diamonds represent reference stations sampled in the Piedmont Bioregion.

Clustering was observed with separate groups of the Piedmont Bioregion. One group is located in the top center of the chart and two other groups appear along the left side in the upper left and lower left of the chart. This separation could also be from slight differences in the benthic communities due to stream gradient at the stations.

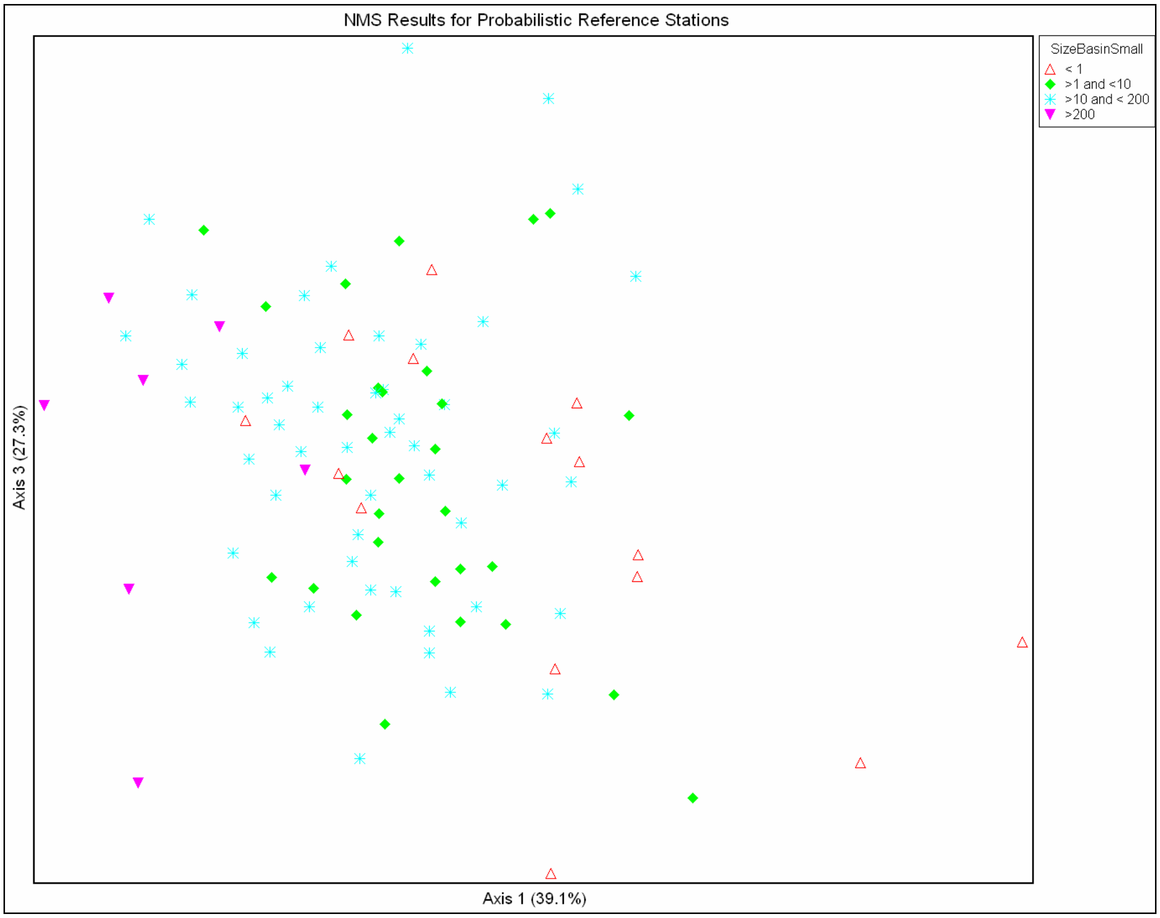
Figure 5. NMS results by bioregion and season (n=104).



The open red triangles represent reference stations sampled during the fall season in the Mountain Bioregion, the solid green diamonds represent reference stations sampled during the fall season in the Piedmont Bioregion, the blue asterisk represent reference stations sampled during the spring season in the Mountain Bioregion, and the solid upside down triangles represent reference stations sampled during the spring season in the Piedmont Bioregion.

Clustering appears to occur with some of the Piedmont fall data grouped in the top center and upper left of the chart. A group of the Piedmont spring data is clustered in the upper left and another group is in the lower left. The Mountain spring and fall data appear to mostly overlap in the center of the chart.

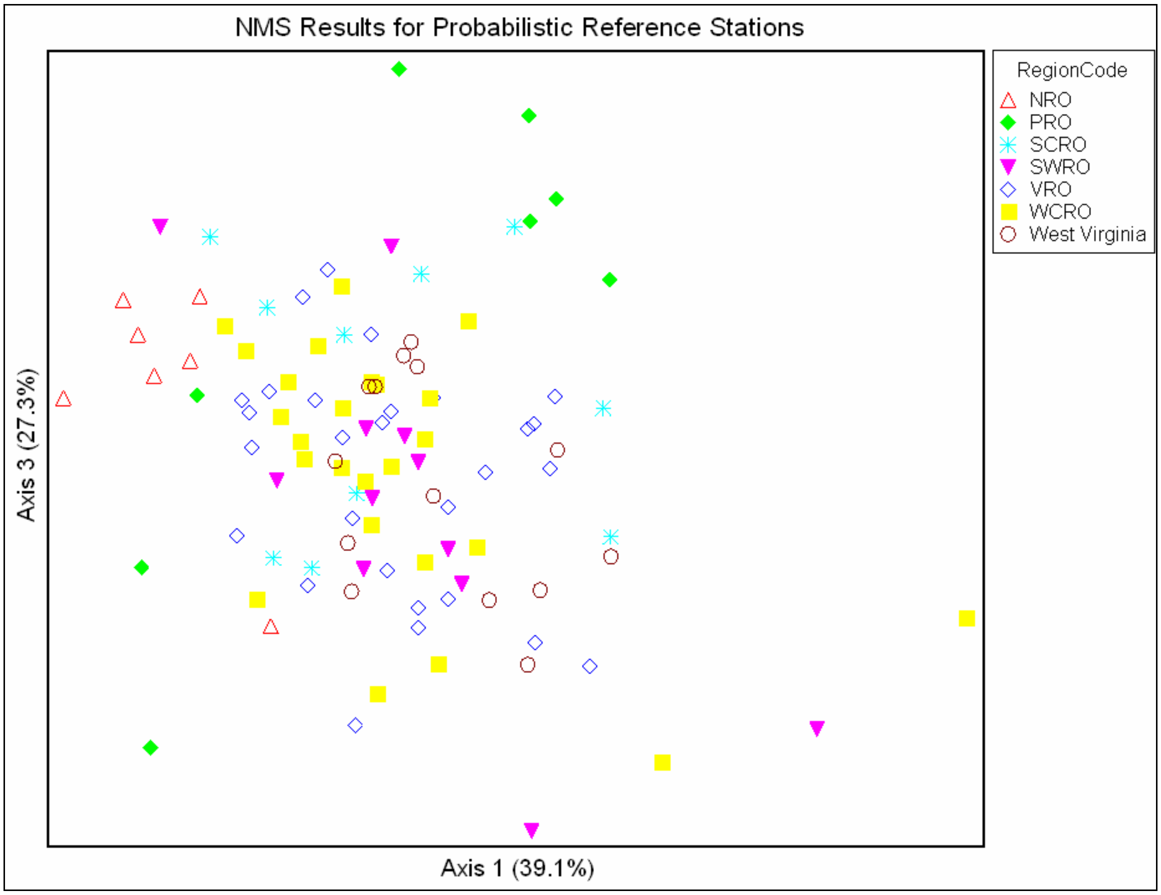
Figure 6. NMS results by basin size (n=104). Units are in square miles.



The open red triangles represent reference stations with a watershed size less than 1 square mile, the solid green diamonds represent reference stations with a watershed size between 1 square mile and 10 square miles, the blue asterisk represent reference stations with a watershed size between 10 square miles and 200 square miles, and the solid upside down triangles represent reference stations with a watershed size greater than 200 square miles.

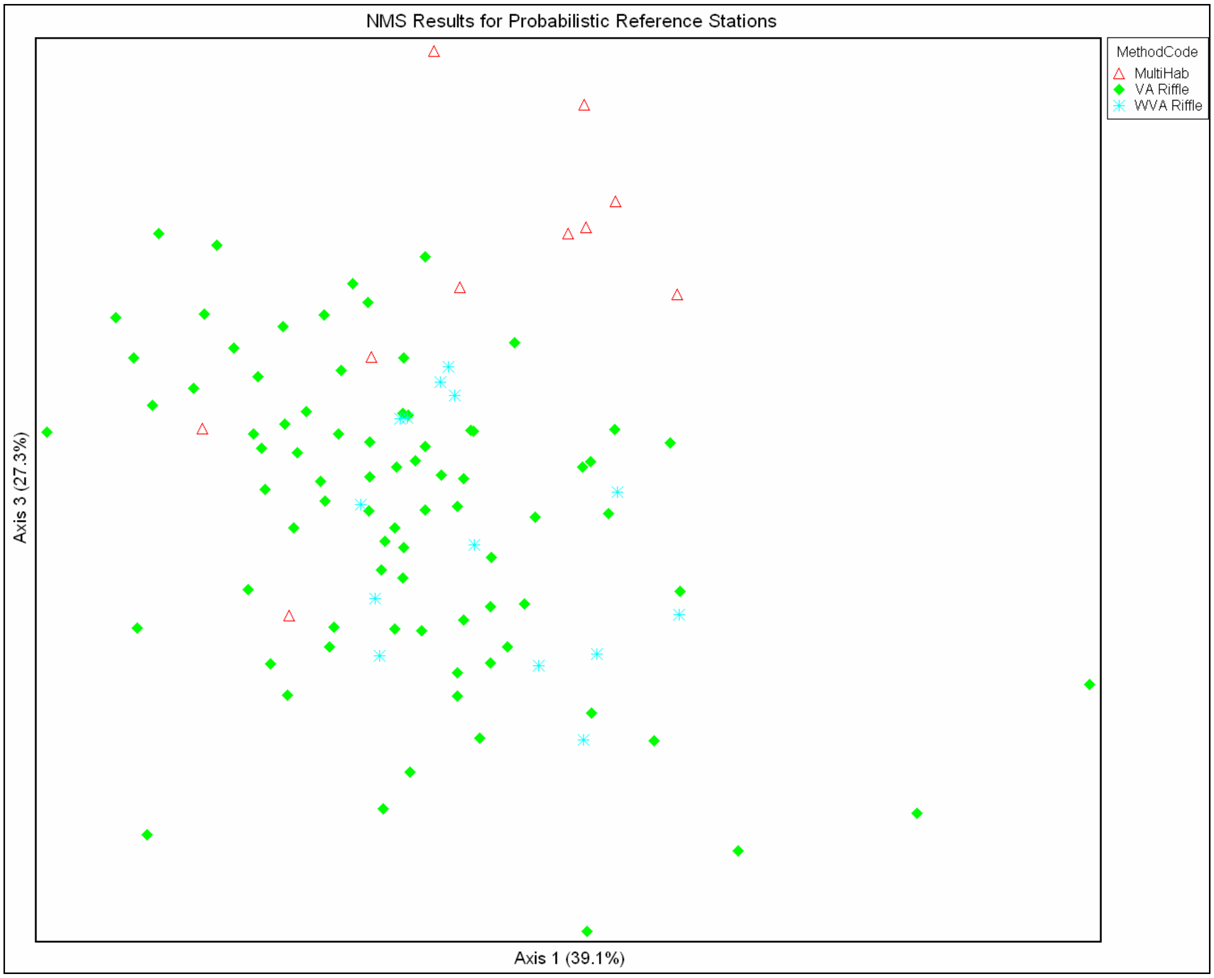
Benthic community data from basins that were < 1 square mile appears to be clustered around the center of the chart and that from basins > 200 square miles is clustered to the left of the chart. The communities occurring in small to medium-sized streams appear to mostly overlap. Overall, it appears that the benthic communities collected at stations in very small and large basins (i.e., low and high order stream) are different from those at small to medium-sized streams.

Figure 7. NMS results by regional office (including West Virginia data) (n=104).



The open red triangles represent reference stations sampled in VDEQ's Northern Region Office (NRO), the solid green diamonds represent reference stations sampled in VDEQ's Piedmont Region Office (PRO), the blue asterisk represent reference stations sampled in VDEQ's South Central Region Office (SCRO), the upside down solid pink triangles represent reference stations sampled in VDEQ's Southwest Region Office (SWRO), the open blue diamonds represent stations sampled in VDEQ's Valley Region Office (VRO), the solid yellow squares represent stations sampled in VDEQ's West Central Region Office (WCRO), and the open red circles represent stations sampled in West Virginia's probabilistic sampling that met VDEQ reference filters.

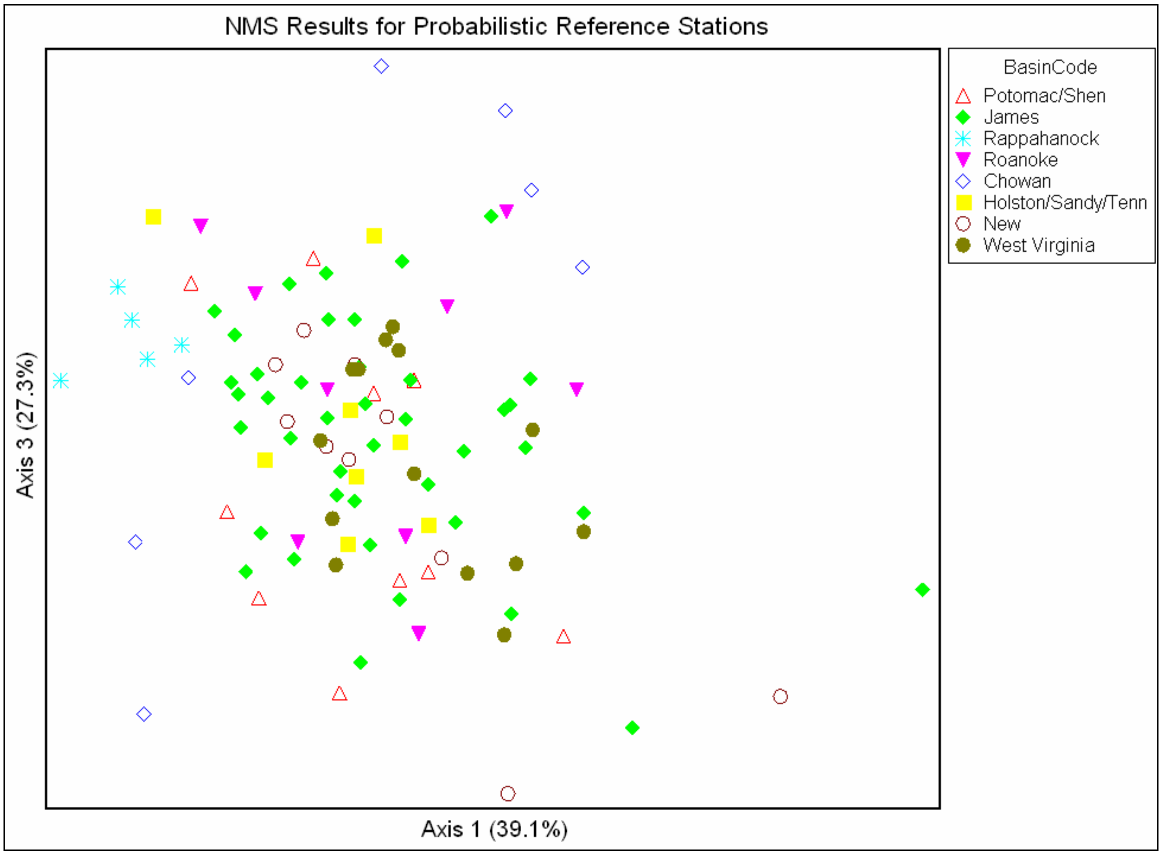
Figure 8. NMS results by sampling method (n=104).



The open red triangles represent reference stations sampled using VDEQ's multi-habitat method, the solid green diamonds represent reference stations sampled using VDEQ's riffle method, and the blue asterisk represent reference stations sampled using West Virginia's riffle method.

Most of the stations where multi-habitat methods were used are clustered in the top center of the chart; whereas, many of the Virginia riffle stations overlap with the West Virginia riffle stations.

Figure 9. NMS results by river basin (n=104).



The open red triangles represent reference stations sampled in the Potomac and Shenandoah river basins, the solid green diamonds represent reference stations sampled in the James river basin, the blue asterisk represent reference stations sampled in the Rappahannock river basin, the upside down solid pink triangles represent reference stations sampled in the Roanoke river basin, the open blue diamonds represent stations sampled in the Chowan river basin, the solid yellow squares represent stations sampled in the Holston, Big Sandy, and Tennessee river basins, the open red circles represent stations sampled in the New river basin, and the solid brown circles represent stations sampled in West Virginia (includes the Cheat, New, Greenbrier, Coal, and Elk river basins).

4.2 MeanSim results

NMS ordination results showed that classification variables in Figures 2-4, 6 and 8 contained potential clustering of reference station communities. Therefore, Mean Similarity analysis was used to further evaluate the results for statistical significance. According to Van Sickle (2000), classification strengths greater than 10 may indicate the need to recalibrate the index.

MeanSim results indicated that the classification strength of the classification variables was low and no variable exceeding 4.4 (Table 8). Thus, overall within group similarity was not different from between group similarity. Combining potential classification categories such as season and bioregion did not improve the classification strength. MeanSim results do not warrant recalibrating the VSCI for use in different seasons, ecoregions, or basin sizes. However, these classifications were tested for environmental significance to determine if any variable affected the core metrics and VSCI (Figures 10-19 and Sect. 4.3).

Percent similarity can be graphically evaluated using a mean similarity dendrogram (Van Sickle 1997). Appendix A contains mean similarity dendrogram graphs and description of the results for all of the categories in Table 8.

Table 8. MeanSim analysis results.

Bray Curtis Similarity Matrix							
	N (ref sites)	N (Groups)	Within Group (W)	Between Group (B)	CS (W-B)	M (B/W)	p-value
Season	104	2	35.9%	31.7%	4.3%	0.88	0.0001
Basin Size	104	4	35.1%	32.7%	2.3%	0.93	0.0002
Ecoregion (III)	104	5	36.4%	32.9%	3.5%	0.91	0.0001
Bioregion	104	2	34.5%	32.2%	2.3%	0.93	0.0001
Bioregion and Season	104	4	36.8%	32.4%	4.4%	0.88	0.0001
Collection Method	104	3	35.1%	32.6%	2.5%	0.93	0.0033

4.3 Environmental significance results

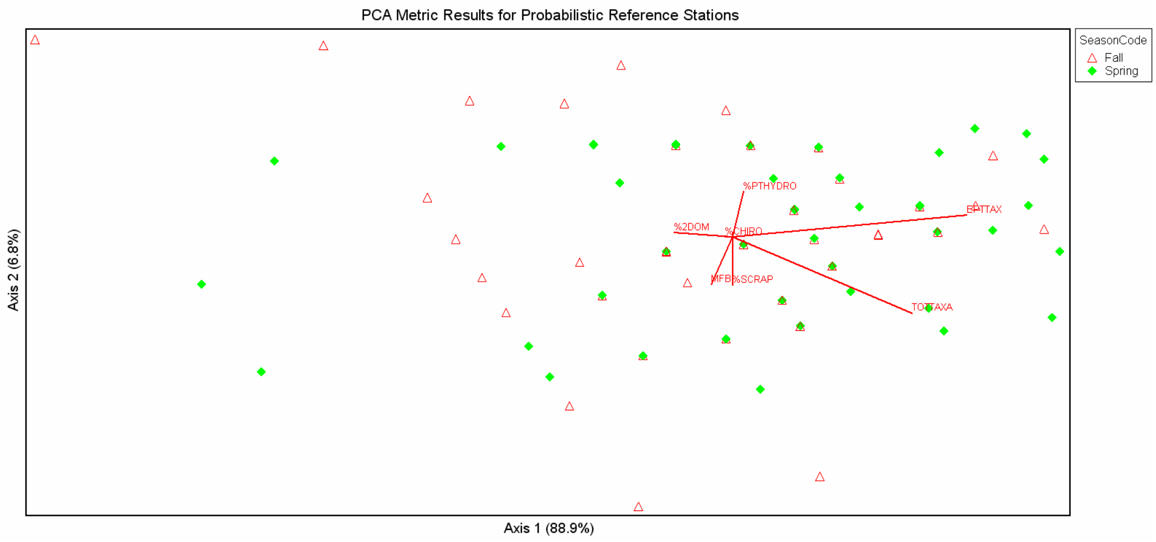
Environmental significance was evaluated based on the results in section 4.2. Environmental significance occurs when a classification variable significantly affects one or more of the core metrics in such a way that the distribution of VSCI reference scores is significantly changed. Potential seasonal, ecoregion, bioregion, bioregion + season, basin size, and sampling method patterns were found in NMS ordination results (Section 4.2). MeanSim statistical results indicated that these patterns had low classification strength. Principal Components Analysis (PCA) was used to determine if classification variables affected the core biological metrics in an environmentally significant fashion.

PCA results were generated using a variance-covariance matrix centered by parameters and graphs were produced to evaluate metric clustering by classification variable (Figures 10-15). The richness metrics were log transformed and the proportion metrics were arcsine transformed in the input matrix. The PCA graphs were evaluated to identify clusters of metrics by classification variable. Axis 1 and 2 accounted for approximately 95.7% of the variation. The metric vectors represent Pearson correlations of a least 0.02 with Axis 1 or 2. None of the stations formed distinct patterns in Figures 10-15 and the metric vectors did not cluster by classification variable.

Systat 11 was used to create box plots. The box outlines are the 25th to 75th percentiles (this is known as the interquartile range). The solid line inside the box is the median value of the data. The whisker ends are 1.5 x the interquartile range. The '*' on the box plots denotes values greater than the 1.5 x the interquartile range (an outlier) but less than 3 x the interquartile range. The 'o' represents values > 3.0 x interquaritle range (an extreme outlier).

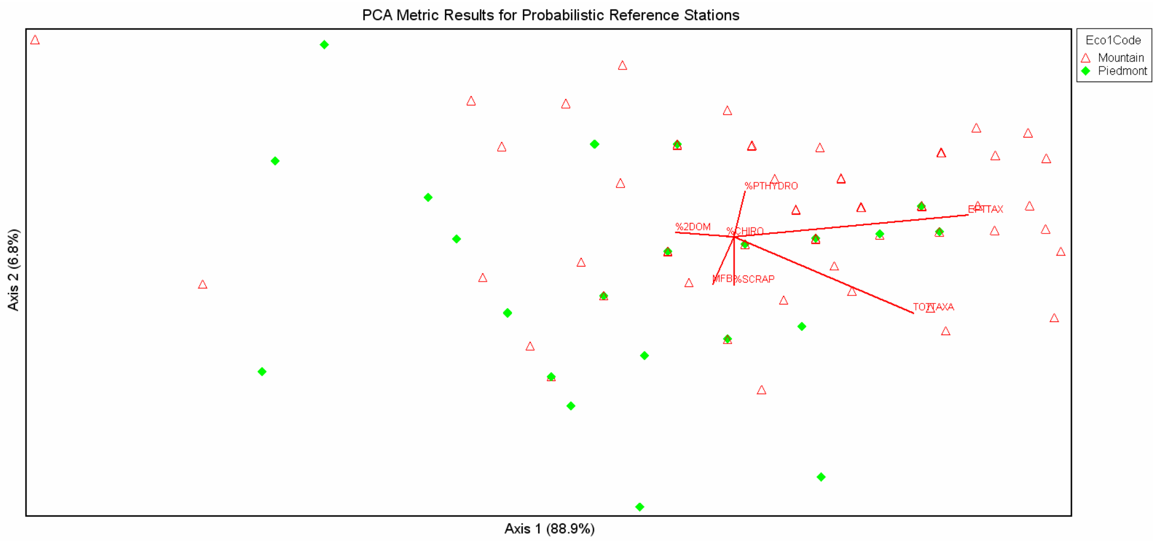
Box-and-whisker plots of the eight core metrics and VSCI score were used to graphically determine significant variation on the reference stations by classification variable (Figures 16 - 21). Metric and VSCI box-and-whisker plots were generated by season, bioregion, bioregion + season, ecoregion, basin size, and sampling method. Individual metrics were different by classification category. Season (Figure 16) had little difference in individual metrics and the VSCI except for % Scrapers, which were higher in the fall and % Chironomidae, which were higher in the spring. In the graphs for bioregion (Figure 17), ecoregion (Figure 18), and sampling method (Figure 21), the EPT taxa and MFBI metrics were different. The EPT taxa were higher in the Mountain Bioregion compared to the Piedmont Bioregion. It was noted that reference Central Appalachian stations have VSCI scores similar to other mountain ecoregions. EPT numbers were higher in reference sites sampled with the riffle method versus the sites sampled with the multi-habitat method. Reference stations with a watershed less than 1 square mile (Figure 20) have a higher percentage % Plecoptera + Trichoptera (- Hydropsychidae) as expected. However, the differences in individual core metrics do not appear to affect the final VSCI score. The median values for the VSCI in the graphs were similar and the interquartile ranges of the VSCI scores in all of the box-and-whisker plots were above 60.

Figure 10. PCA metric results by season (n=104).



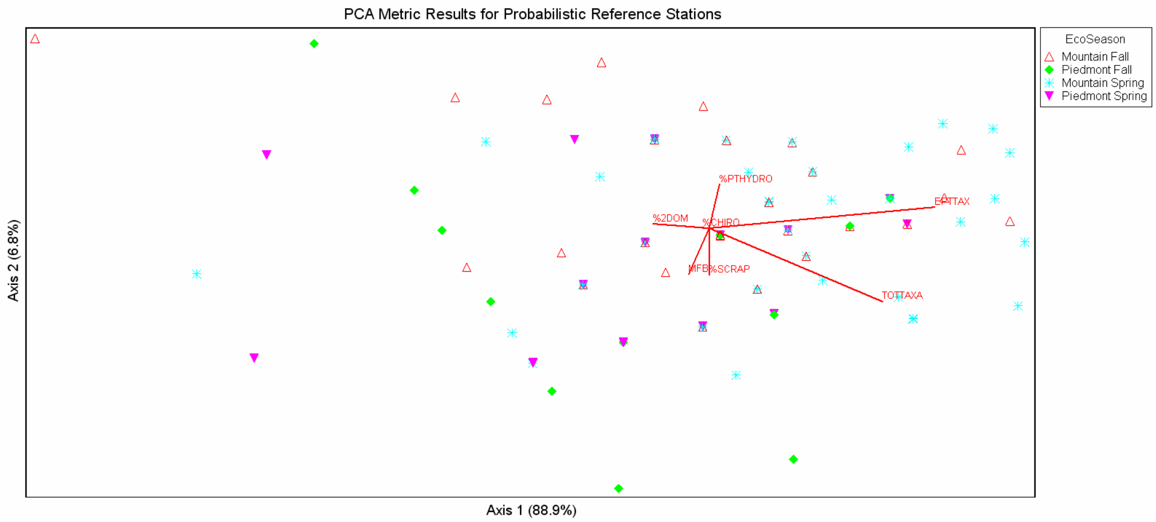
The open red triangles represent reference stations sampled in the fall season and the solid green diamonds represent reference stations sampled in the spring season. Vectors represent the core metrics. Fall and spring samples are spread out and overlap with no distinct pattern.

Figure 11. PCA metric results by bioregion (n=104).



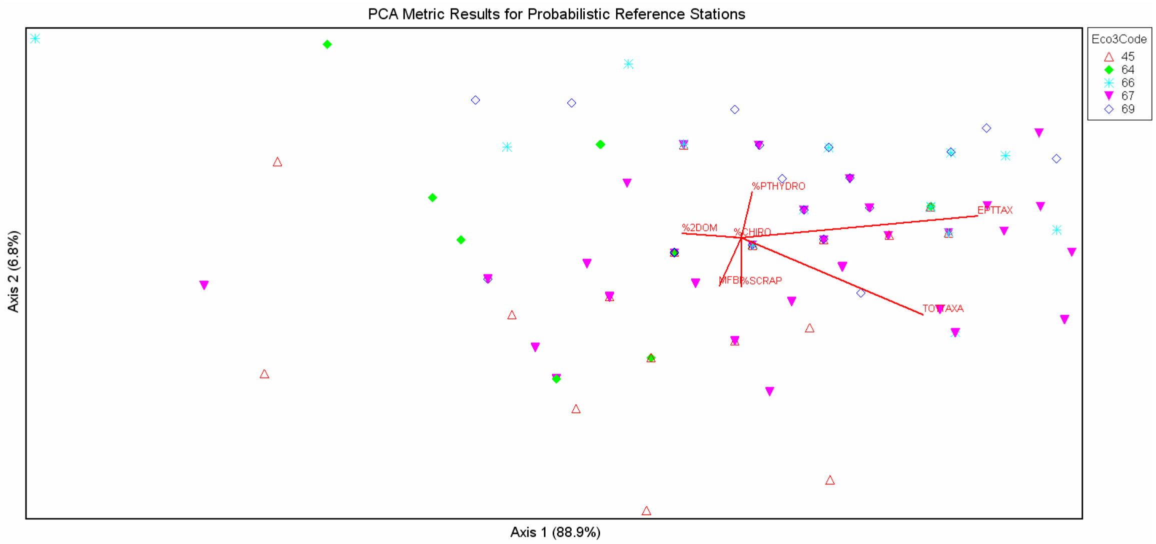
The open red triangles represent reference stations sampled in the Mountain Bioregion and the solid green diamonds represent reference stations sampled in the Piedmont Bioregion. Vectors represent the core metrics. There does not appear to be any distinct patterns in the two different bioregions.

Figure 12. PCA metric results by bioregion and season (n=104).



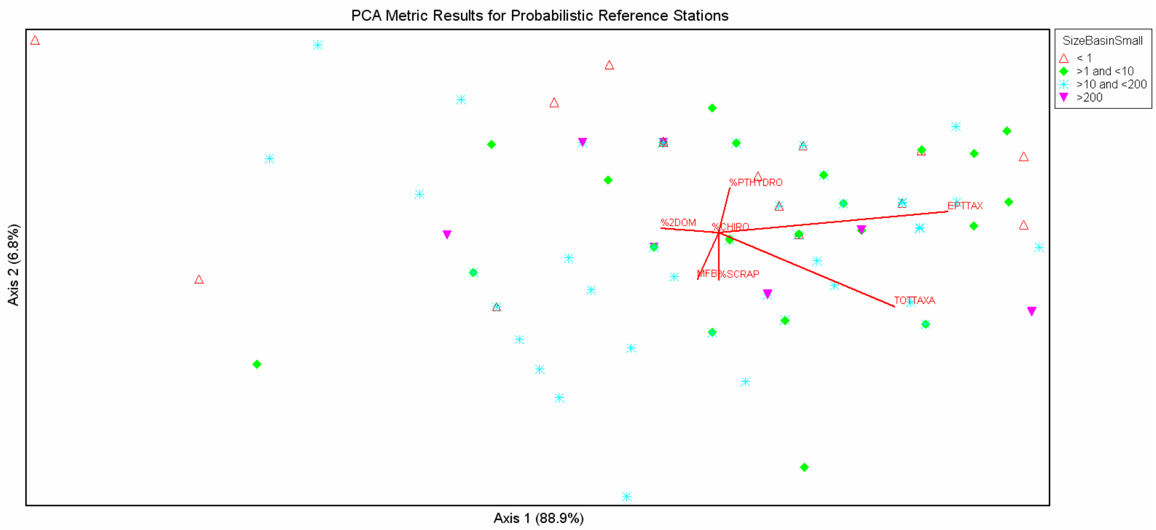
The open red triangles represent reference stations sampled during the fall season in the mountain bioregion, the solid green diamonds represent reference stations sampled during the fall season in the piedmont bioregion, the blue asterisk represent reference stations sampled during the spring season in the mountain bioregion, and the solid upside down triangles represent reference stations sampled during the spring season in the piedmont bioregion. Vectors represent the core metrics. There does not appear to be any distinct patterns in the four different combinations of bioregion and season.

Figure 13. PCA metric results by ecoregion (n=104).



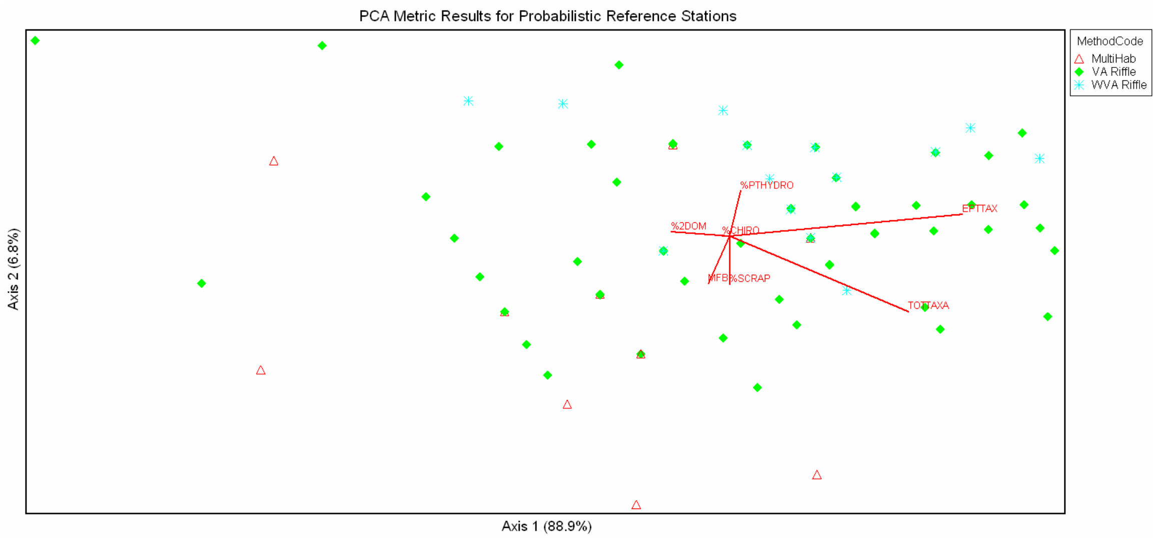
The open red triangles represent reference stations sampled in ecoregion 45 (Piedmont), the solid green diamonds represent reference stations sampled in ecoregion 64 (Northern Piedmont), the blue asterisk represent reference stations sampled in ecoregion 66 (Blue Ridge Mountains), the upside down solid pink triangles represent reference stations sampled in ecoregion 67 (Central Appalachian Ridge and Valley), and the open blue diamonds represent stations sampled in ecoregion 69 (Central Appalachian). Vectors represent the core metrics. There does not appear to be any distinct patterns in the five different ecoregions.

Figure 14. PCA metric results by basin size (n=104).



The open red triangles represent reference stations with a watershed size less than 1 square mile, the solid green diamonds represent reference stations with a watershed size between 1 square mile and 10 square miles, the blue asterisk represent reference stations with a watershed size between 10 square miles and 200 square miles, and the solid upside down triangles represent reference stations with a watershed size greater than 200 square miles. Vectors represent the core metrics. There does not appear to be any distinct patterns in the four different basin sizes.

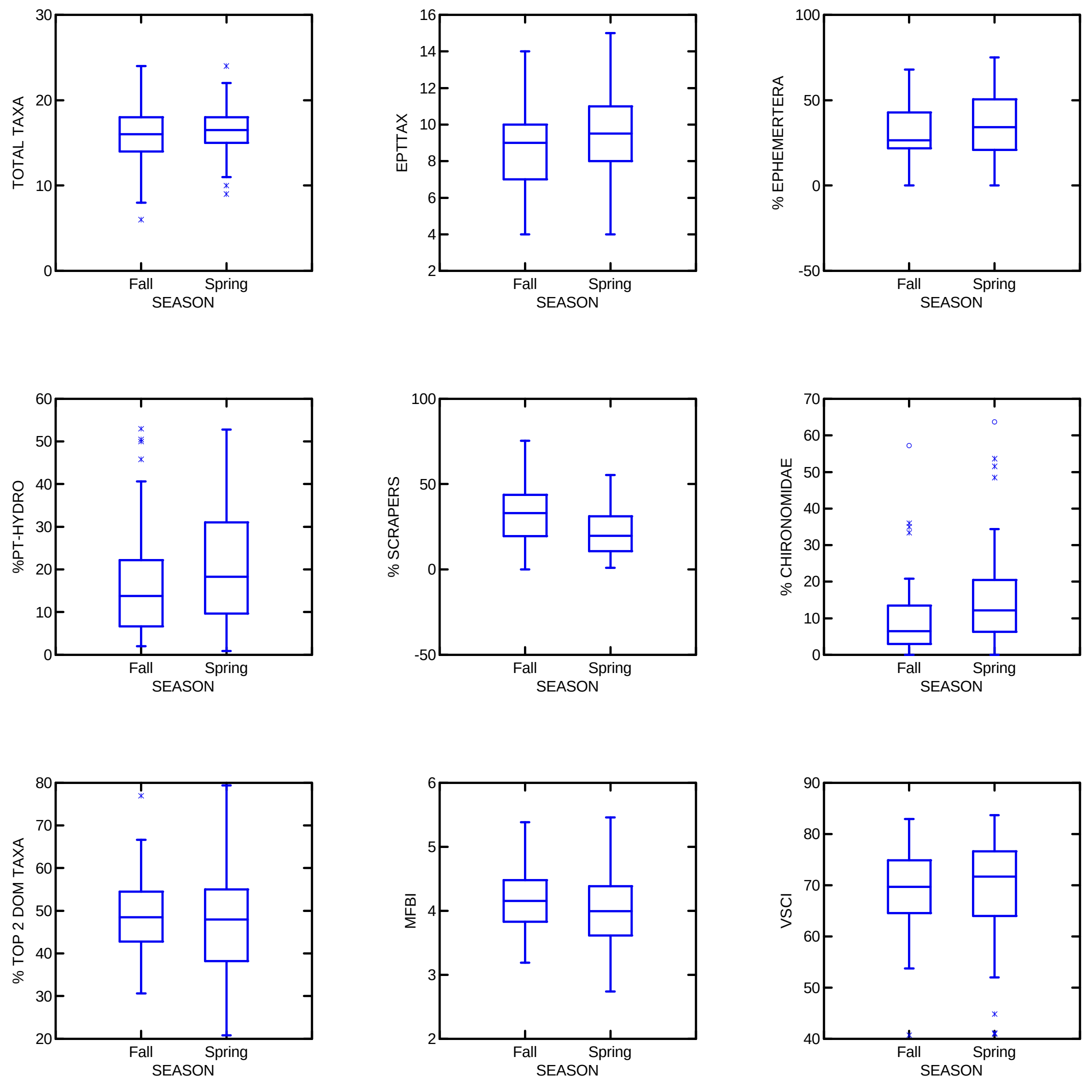
Figure 15. PCA metric results by sampling method (n=104).



The open red triangles represent reference stations sampled using VDEQ's multi-habitat method, the solid green diamonds represent reference stations sampled using VDEQ's riffle method, and the blue asterisk represent reference stations sampled using West Virginia's riffle method.

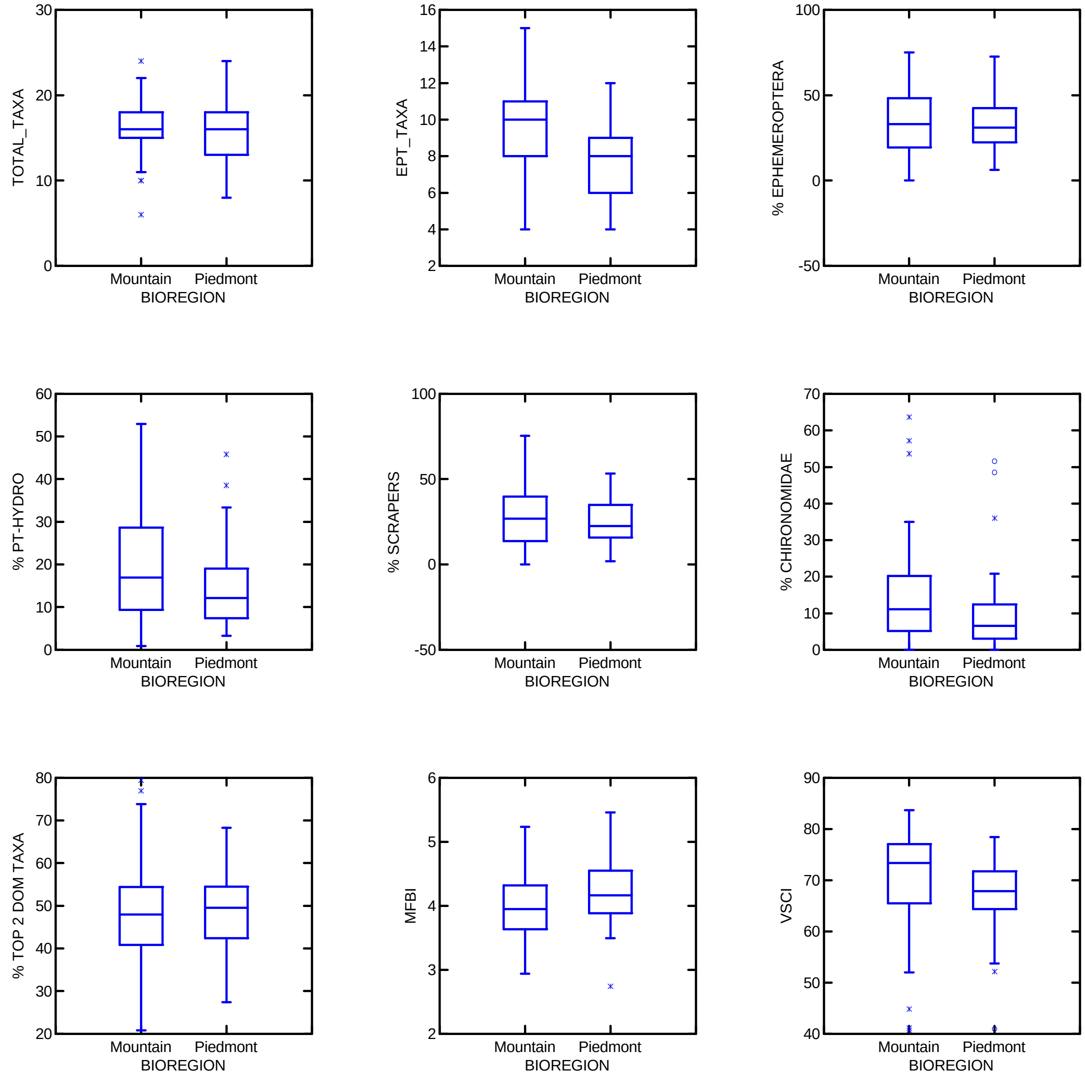
Vectors represent the core metrics. There does not appear to be any distinct patterns in the three different sampling methods.

Figure 16. Box-and-whisker plot of core metrics and VSCI by season.



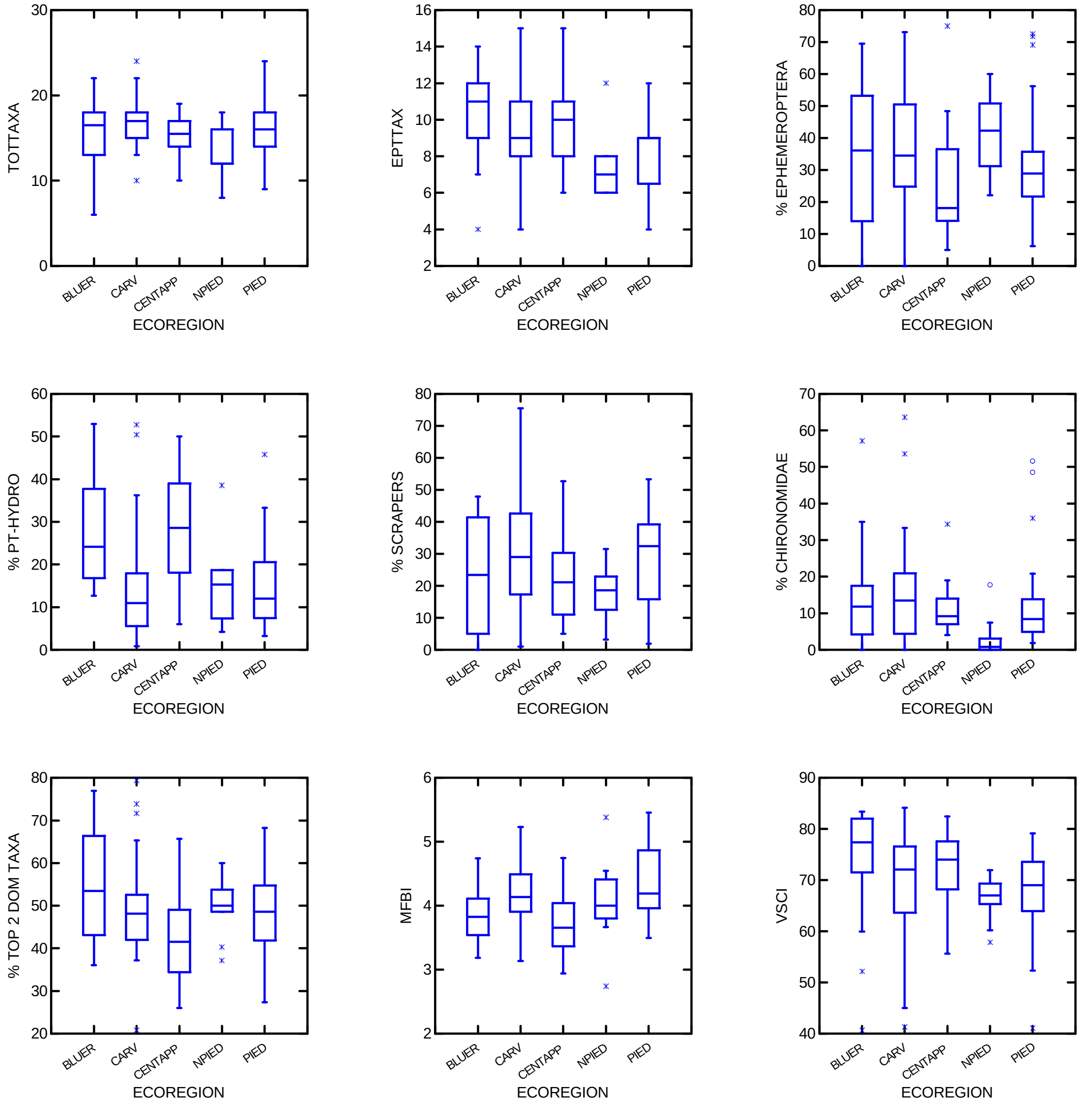
Spring sampling (n=56) occured from March to June and fall sampling (n=48) occured from August to November.

Figure 17. Box-and-whisker plot of core metrics and VSCI by bioregion.



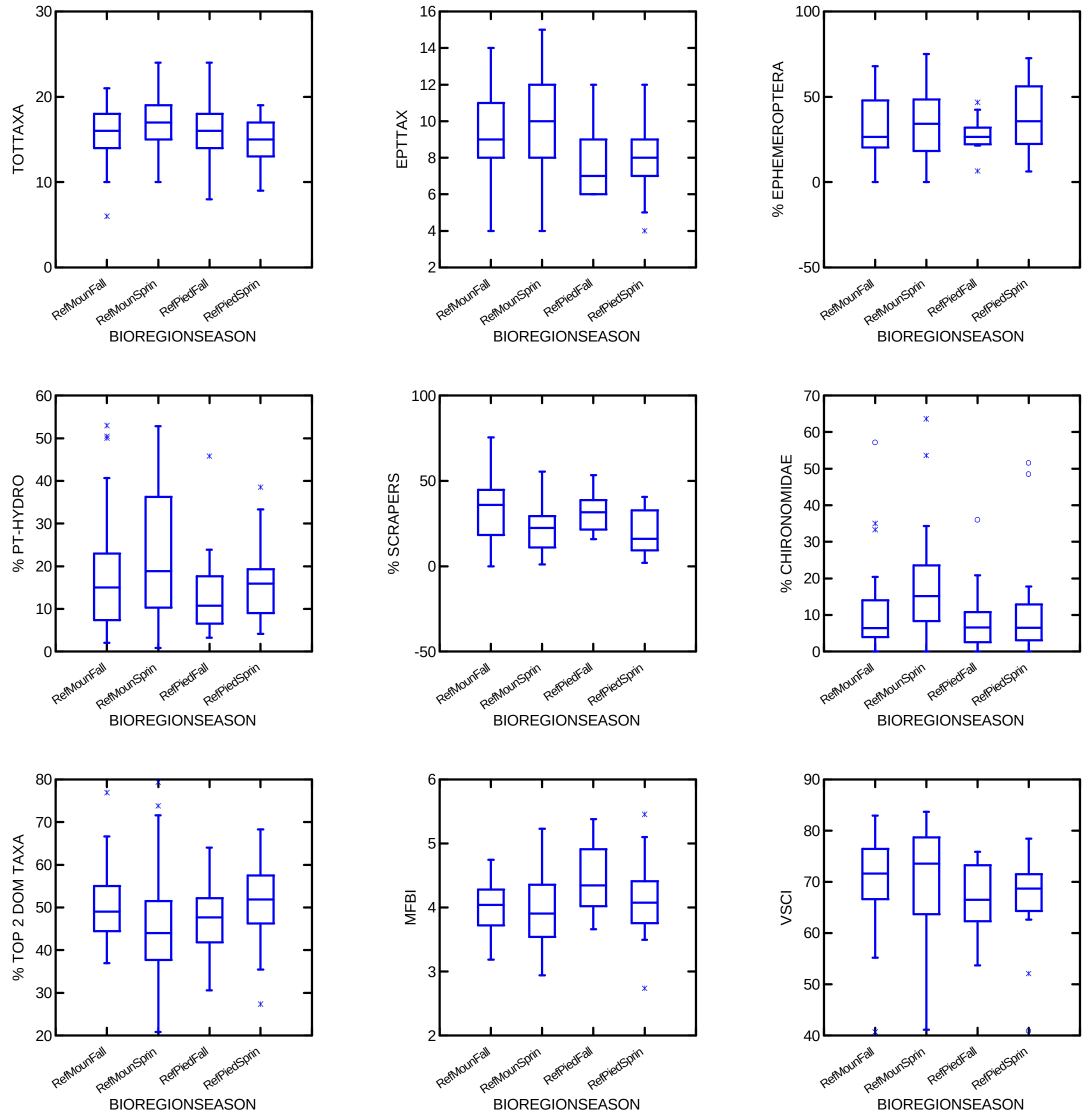
The Mountain Bioregion (n=71) consists of the Blue Ridge Mountain, Central Appalachian, and Central Appalachian Ridge and Valley Ecoregions. The Piedmont Bioregion (n=33) consists of the Northern Piedmont and Piedmont Ecoregions.

Figure 18. Box-and-whisker plot of core metrics and VSCI by ecoregion.



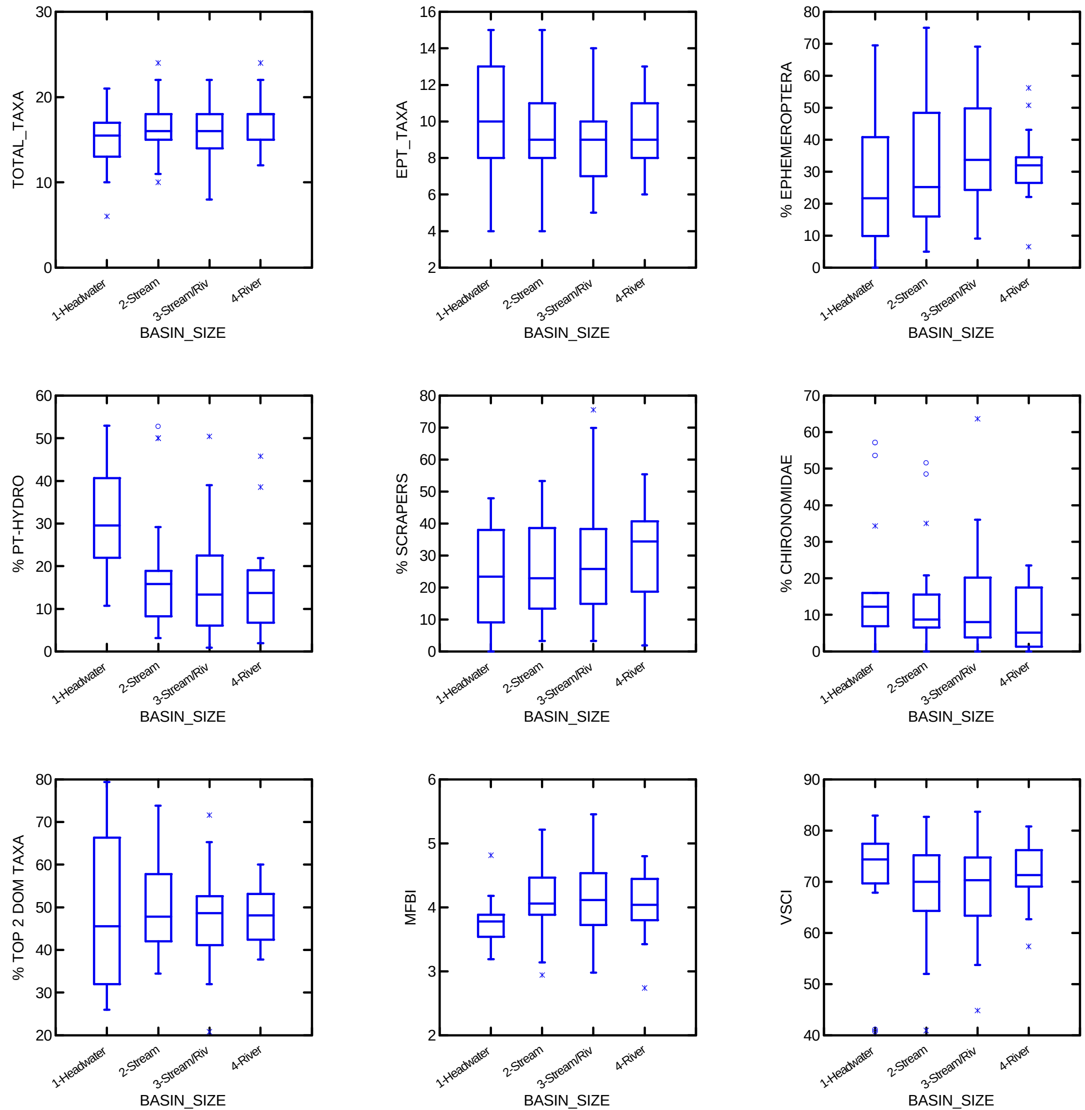
The Mountain Bioregion consists of the Blue Ridge Mountain (n=14), Central Appalachian (n=18), and Central Appalachian Ridge and Valley (n=39) Ecoregions. The Piedmont Bioregion consists of the Northern Piedmont (n=9) and Piedmont Ecoregions (n=24).

Figure 19. Box-and-whisker plot of core metrics and VSCI by bioregion and season.



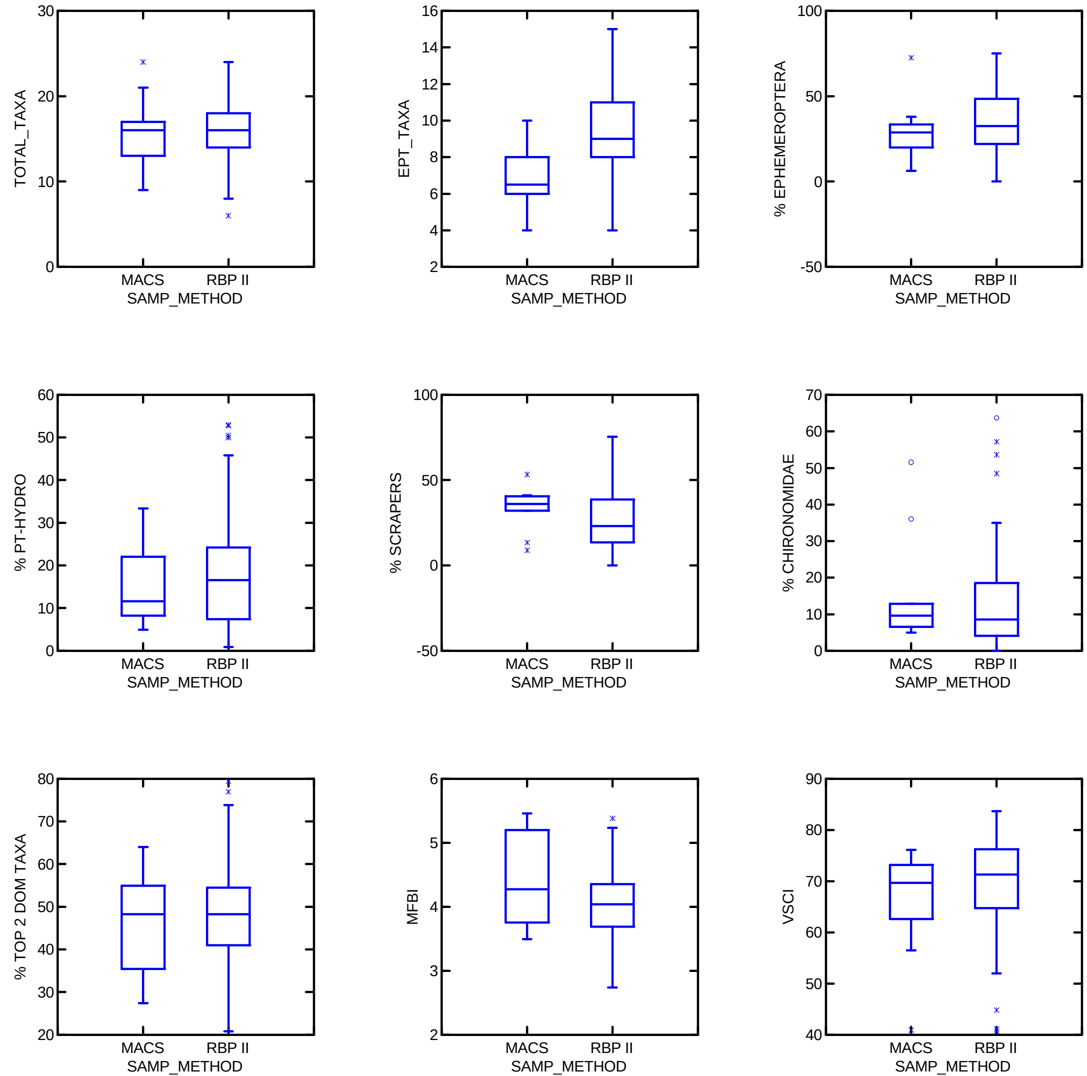
Spring sampling occured from March to June and fall sampling occured from August to November. The Mountain Bioregion consists of the Blue Ridge Mountain, Central Appalachian, and Central Appalachian Ridge and Valley Ecoregions. The Piedmont Bioregion consists of the Northern Piedmont and Piedmont Ecoregions. This plot shows Mountain Bioregion spring (n=29) and fall (n=29) samples, and Piedmont Bioregion spring (n=18), and fall (n=15) samples.

Figure 20. Box-and-whisker plot of core metrics and VSCI by basin size.



Headwater streams (n=14) were defined as a watershed with less than 1 square mile, small streams (n=29) were defined as stations with a watershed size between 1 square mile and 10 square miles. Stations with a watershed size between 10 square miles and 200 square miles were defined as large streams/small rivers (n=54), and stations with a watershed size greater than 200 square miles were defined as rivers (n=7). These delineations were based on descriptions by Ohio EPA (Ohio EPA 1999).

Figure 21. Box-and-whisker plot of core metrics and VSCI by sampling method.



The multi-habitat sampling method (n=10) is abbreviated using MACS and the riffle sample method (n=94) is abbreviated using RBP II.

4.4 Best Standard Value Analysis Results

CDF curves of the eight core metrics were used to generate statewide best standard values (BSV). See Appendixes B-D for complete data output of the core metrics. Average BSV recommended by Tetra Tech along with the average probabilistic BSV are in Table 9. CDF BSV results by individual season and bioregion are in Table 10.

VSCI scores were calculated with bioregion and season specific BSV and average BSV to determine if there were any significant advantages in using the average best standard value method (Figures 22 to 24) versus the recalibrated BSV method (Figures 25 to 28).

The probabilistic values were generated for the entire population in the non-coastal region of Virginia. In six of the eight core metrics, the probabilistic BSV compared favorably with Tetra Tech's recommended best standard values. VDEQ's probabilistic data set is not large enough to recommend better BSV for % Plecoptera + Trichoptera (- Hydropsychidae) and % Ephemeroptera. VDEQ will review those two metrics when more data is available (see Appendix B for detailed output).

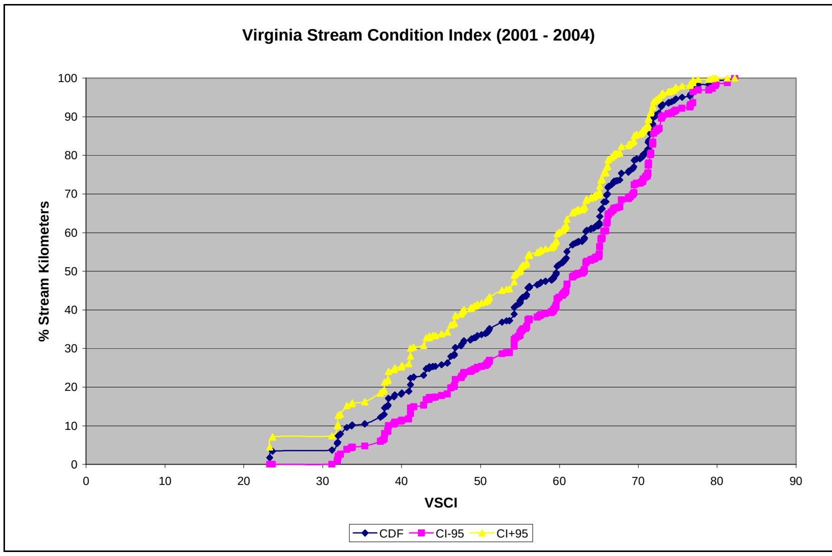
Table 9. Best standard value (BSV) comparison.

Metric	TetraTech BSV	ProbMon BSV
Total Taxa (95Pct)	22.0	18.9
EPT Taxa (95Pct)	11.0	11.7
% Ephem (95Pct)	61.3	47.7
% PT- Hydro (95Pct)	35.6	56.2
% Scrapers (95Pct)	51.6	46.7
% Chiro (5 Pct)	0.0	1.6
% 2 Dom (5 Pct)	30.8	32.8
HBI (5 Pct)	3.2	2.9

Table 10. Probabilistic BSV by season and ecoregion.

Metric	Mountain Spring BSV	Mountain Fall BSV	Piedmont Spring BSV	Piedmont Fall BSV
Total Taxa (95Pct)	20.4	19.5	19.0	19.1
EPT Taxa (95Pct)	13.7	11.3	9.6	8.9
% Ephem (95Pct)	69.0	53.5	53.4	45.1
% PT- Hydro (95Pct)	70.7	53.6	30.8	25.4
% Scrapers (95Pct)	43.8	77.0	35.4	45.2
% Chiro (5 Pct)	1.0	0.9	0.0	0.0
% 2 Dom (5 Pct)	36.5	32.4	25.3	29.9
HBI (5 Pct)	2.5	3.0	3.7	3.6

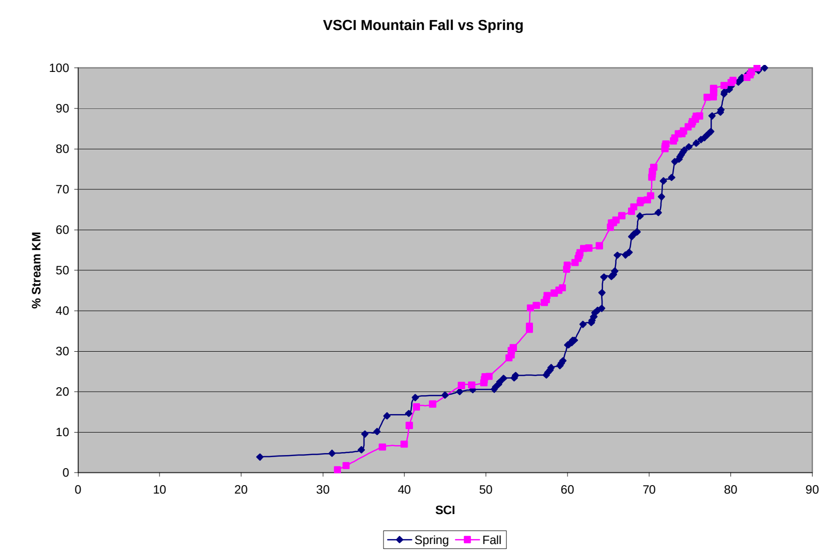
Figure 22. CDF curve of Non-coastal VSCI scores generated using average BSV (n=187).



Season	Value	N	VSCI	CI-95%	CI+95%
All	1Pct	1	23.3	23.3	31.2
All	5Pct	3	31.7	23.3	32.6
All	10Pct	9	33.7	31.4	38.3
All	20Pct	24	41.0	37.8	46.0
All	25Pct	30	43.5	38.3	49.5
All	30Pct	38	46.8	41.1	54.0
All	40Pct	58	54.3	47.8	59.3
All	50Pct	85	59.6	54.9	63.2
All	60Pct	106	63.3	60.0	65.9
All	70Pct	125	66.1	65.1	69.4
All	75Pct	134	67.8	65.9	71.2
All	80Pct	147	70.6	67.5	71.7
All	90Pct	161	72.1	71.6	74.4
All	95Pct	174	75.4	72.8	76.9
All	99Pct	183	79.7	76.9	82.2

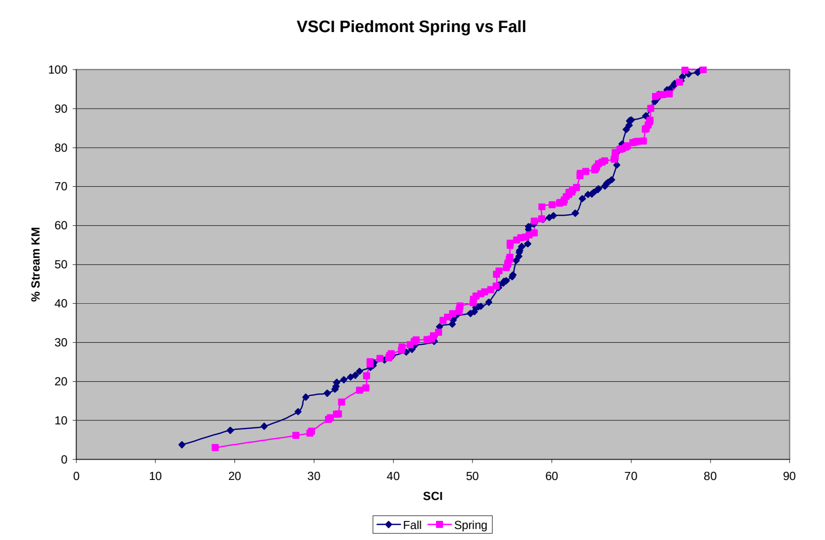
The CDF curve contains data from all non-coastal stations sampled by the probabilistic monitoring program from 2001-2004.

Figure 23. Average BSV spring (n=81) and fall (n=78) Mountain Bioregion CDF and percentiles



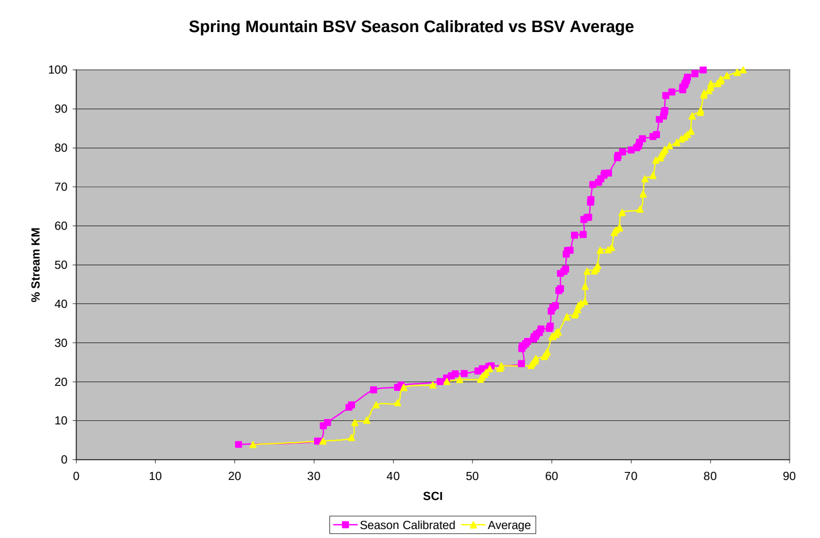
Season	Value	N	VSCI	CI-95%	CI+95%
Spring	5Pct	2	32.1	22.3	37.0
Spring	10Pct	4	36.3	22.3	43.0
Spring	25Pct	20	57.7	37.8	61.8
Spring	50Pct	43	65.8	62.4	68.8
Spring	75Pct	55	73.0	70.8	77.6
Spring	90Pct	70	78.8	76.8	81.2
Spring	95Pct	73	79.9	79.0	82.4
Season	Value	N	VSCI	CI-95%	CI+95%
Fall	5Pct	2	36.0	33.7	40.0
Fall	10Pct	4	40.4	33.8	43.7
Fall	25Pct	13	51.0	40.8	55.4
Fall	50Pct	27	59.9	55.3	66.8
Fall	75Pct	52	70.5	67.9	74.8
Fall	90Pct	67	76.6	73.1	81.9
Fall	95Pct	71	78.0	76.9	82.4

Figure 24. Average BSV spring (n=103) and fall (n=88) Piedmont Bioregion CDF and percentiles



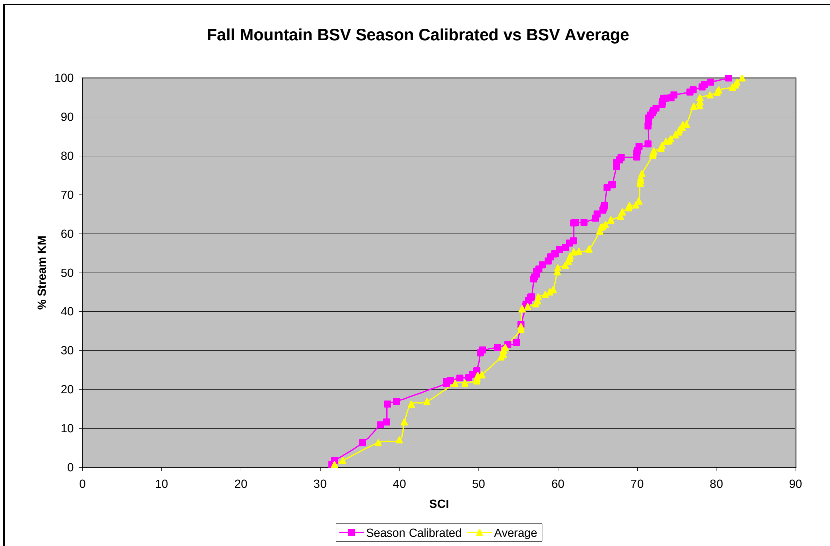
Season	Value	N	VSCI	CI-95%	CI+95%
Spring	5Pct	1	24.0	17.5	31.4
Spring	10Pct	4	31.6	19.0	34.8
Spring	25Pct	13	37.1	33.4	46.4
Spring	50Pct	43	54.4	48.4	57.8
Spring	75Pct	74	65.7	59.3	72.0
Spring	90Pct	94	72.5	69.9	76.6
Spring	95Pct	99	75.4	72.4	79.1
Season	Value	N	VSCI	CI-95%	CI+95%
Fall	5Pct	1	15.4	13.3	26.8
Fall	10Pct	3	25.4	13.3	32.4
Fall	25Pct	16	38.0	28.5	47.6
Fall	50Pct	39	55.4	50.0	60.0
Fall	75Pct	64	68.1	63.1	69.8
Fall	90Pct	74	72.4	69.1	76.3
Fall	95Pct	81	74.9	72.9	76.8

Figure 25. Spring Mountain Bioregion BSV comparison VSCI CDF and percentiles



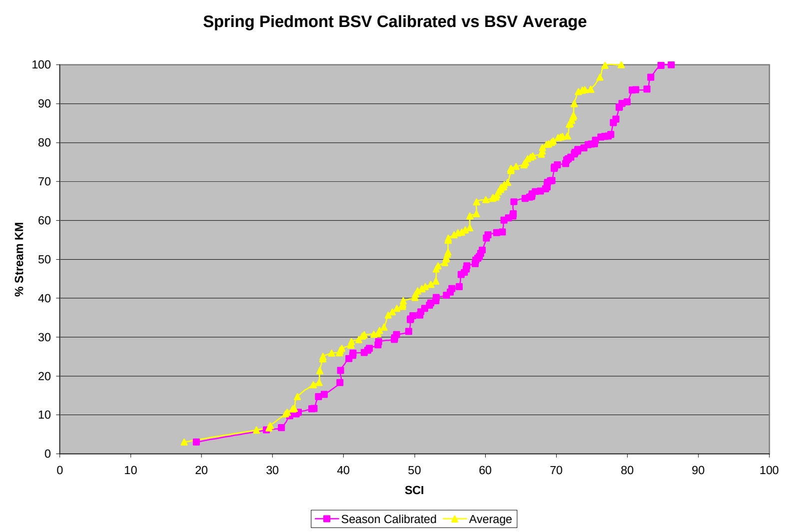
Average	Value	N	VSCI	CI-95%	CI+95%
Average	5Pct	2	32.1	22.3	37.0
Average	10Pct	4	36.3	22.3	43.0
Average	25Pct	20	57.7	37.8	61.8
Average	50Pct	43	65.8	62.4	68.8
Average	75Pct	55	73.0	70.8	77.6
Average	90Pct	70	78.8	76.8	81.2
Average	95Pct	73	79.9	79.0	82.4
Calibrated	**Value**	**N**	**VSCI**	**CI-95%**	**CI+95%**
Calibrated	5Pct	2	30.5	20.5	33.0
Calibrated	10Pct	4	32.0	20.5	40.8
Calibrated	25Pct	19	56.2	34.6	59.9
Calibrated	50Pct	40	61.8	59.9	64.7
Calibrated	75Pct	56	67.6	64.8	73.5
Calibrated	90Pct	70	74.3	72.6	77.0
Calibrated	95Pct	73	76.5	74.3	77.6

Figure 26. Fall Mountain Bioregion BSV comparison VSCI CDF and percentiles



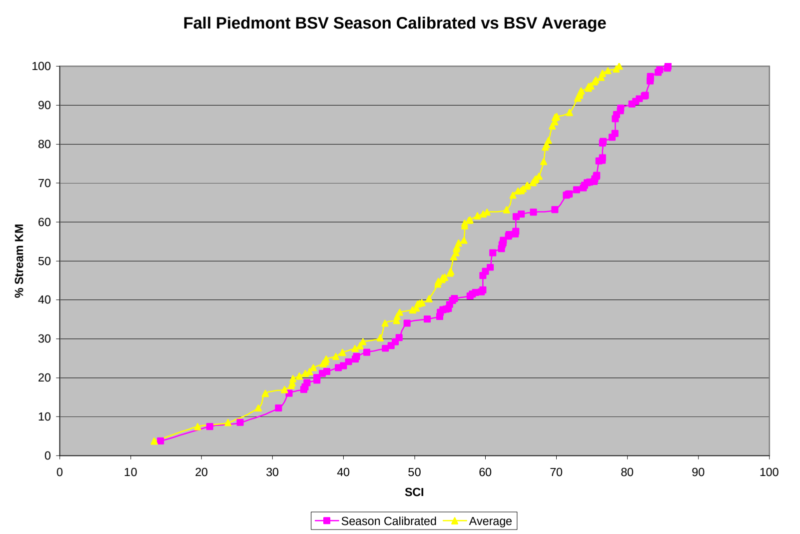
Average	Value	N	VSCI	CI-95%	CI+95%
Average	5Pct	2	36.0	33.7	40.0
Average	10Pct	4	40.4	33.9	43.7
Average	25Pct	13	51.0	40.8	55.4
Average	50Pct	27	59.9	55.3	66.7
Average	75Pct	52	70.5	67.9	74.8
Average	90Pct	67	76.6	73.1	81.9
Average	95Pct	71	78.0	76.9	82.4
Calibrated	**Value**	**N**	**VSCI**	**CI-95%**	**CI+95%**
Calibrated	5Pct	2	34.3	32.5	35.9
Calibrated	10Pct	3	37.1	32.5	40.1
Calibrated	25Pct	14	49.8	38.4	55.4
Calibrated	50Pct	28	57.3	55.3	64.3
Calibrated	75Pct	50	67.1	64.8	71.4
Calibrated	90Pct	62	71.5	71.4	74.4
Calibrated	95Pct	71	74.3	72.0	78.5

Figure 27. Spring Piedmont Bioregion BSV comparison VSCI CDF and percentiles



Average	Value	N	VSCI	CI-95%	CI+95%
Average	5Pct	1	24.0	17.5	31.4
Average	10Pct	4	31.6	19.0	34.8
Average	25Pct	13	37.1	33.4	46.4
Average	50Pct	43	54.4	48.4	57.8
Average	75Pct	74	65.7	59.5	71.9
Average	90Pct	94	72.5	70.0	76.6
Average	95Pct	99	75.4	72.4	79.1
Calibrated	**Value**	**N**	**VSCI**	**CI-95%**	**CI+95%**
Calibrated	5Pct	1	25.5	19.3	32.4
Calibrated	10Pct	4	32.8	19.3	39.3
Calibrated	25Pct	13	41.1	36.4	50.8
Calibrated	50Pct	44	58.8	52.4	63.9
Calibrated	75Pct	74	71.4	64.7	78.0
Calibrated	90Pct	94	79.2	77.6	83.8
Calibrated	95Pct	99	83.0	78.7	86.2

Figure 28. Fall Piedmont Bioregion BSV comparison VSCI CDF and percentiles



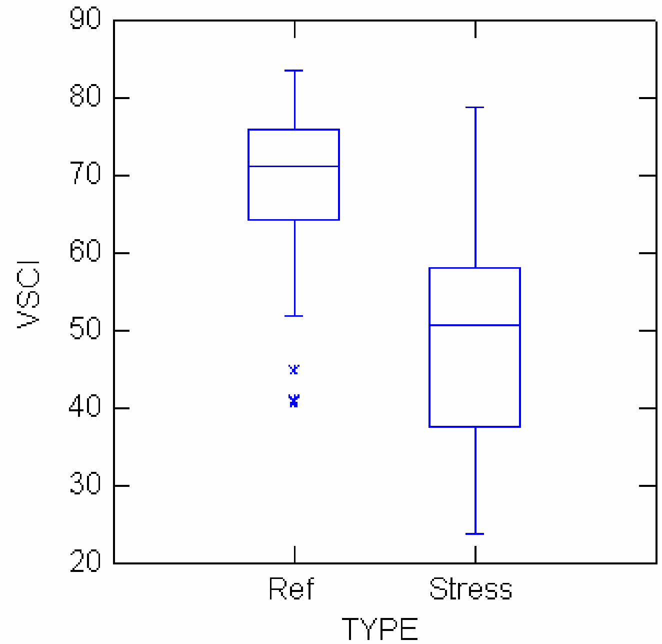
Average	Value	N	VSCI	CI-95%	CI+95%
Average	5Pct	1	15.4	13.3	26.8
Average	10Pct	3	25.4	13.3	32.4
Average	25Pct	16	38.0	28.5	47.6
Average	50Pct	39	55.4	50.0	60.0
Average	75Pct	64	68.1	63.1	69.8
Average	90Pct	74	72.4	69.1	76.3
Average	95Pct	81	74.9	72.9	76.7
Calibrated	**Value**	**N**	**VSCI**	**CI-95%**	**CI+95%**
Calibrated	5Pct	1	16.6	14.2	29.4
Calibrated	10Pct	3	27.6	14.2	34.6
Calibrated	25Pct	16	41.7	31.6	53.6
Calibrated	50Pct	40	60.8	54.8	65.7
Calibrated	75Pct	65	75.9	69.9	78.3
Calibrated	90Pct	76	80.2	78.2	83.2
Calibrated	95Pct	81	83.0	78.9	85.7

Using ecoregionally and seasonally calibrated BSV lowers overall Mountain Bioregion scores in both seasons. The same calibration raises overall Piedmont Bioregion scores in both seasons. This result is expected as average BSV tend to be slightly reduced for Mountain streams and slightly increased for Piedmont streams.

4.5 VSCI Discrimination Ability and Reference Percentiles.

The VSCI works well to discriminate between sites with acceptable water quality and,habitat versus sites with degraded water quality and habitat. The 10th percentile from the probabilistic data set was 58.5, which is a few points lower than the 10th percentile of 61.3 reported by Tetra Tech (2003) from the targeted database (Figure 29).

Figure 29. Box-and-whiskers all reference (n = 104) versus all stress (n = 64) stations.



Reference Percentile	VSCI
1%	40.8
5%	52.1
10%	58.5
20%	62.8
25%	64.3
30%	66.4
40%	68.5
50%	71.3
60%	73.3
70%	74.8
75%	76.0
80%	76.9
90%	79.6
95%	81.7
99%	83.3

4.6 VSCI Variability

Virginia's probabilistic program replicated 5% (n = 16) of the non-coastal sampling effort. Descriptive statistics, precision calculations, and signal-to-noise ratios from these replicate stations by season are found in Table 11. VSCI scores are relatively stable and the overall precision of the index is good (precision is calculated at the 90% confidence interval). A certain amount of variability is associated with biological indexes. This variation is due to natural variation in hydrology, water chemistry, and habitat throughout the commonwealth of Virginia. The VSCI appears to be less variable in the fall than in the spring sampling season. The precision is better in the fall and the signal-to-noise ratio is much higher. However, many factors are affecting this randomly replicate data set and more data is necessary to confirm this trend. It appears that higher quality sites were replicated in the fall which may explain the lower variability associated with the fall sampling season in this data set.

Table 11. Descriptive statistics, precision (90% CI), and signal-to-noise results from replicate samples.

Category	Mean (Field Samples)	Mean (Duplicate Samples)	S.D. (Field Samples)	S.D. (Duplicate Samples)	Precision	Signal-to-Noise
All Seasons	62.7	63.4	12.1	12.2	7.9	1.9
All Fall	70.7	71.9	4.9	6.0	4.7	24.6
All Spring	54.6	55.0	11.9	11.0	10.1	4.2

5.0 CONCLUSIONS AND RECOMMENDATIONS

After reviewing the probabilistic biological data it has been confirmed that the VSCI works well to discriminate between sites with acceptable water quality and habitat versus sites with degraded water quality and habitat. Potential seasonal, ecoregion, bioregion, basin size, and sampling method patterns were found in NMS ordination results. However, MeanSim statistical results indicated that these patterns had low classification strength. These patterns were further tested for environmental significance using PCA to evaluate metric clustering by classification variable. The PCA failed to display any clustering of metrics by classification variable. Box-and-whisker plots of the metrics and the VSCI were used to visually determine any classification influence on the reference stations. Individual metric differences were noted by classification variable. Season showed little difference in individual metrics and the VSCI except for % Scrapers (higher in the fall) and % Chironomidae (higher in the spring). In plots classified by bioregion and sampling method it was noted that EPT taxa and MFBI metrics were different. The EPT taxa were higher at the Mountain Bioregion compared to the Piedmont Bioregion. EPT Taxa were higher at reference sites sampled using the riffle method versus sites sampled with the multi-habitat method. Reference stations with a watershed less than 1 square mile had a higher percentage of Plecoptera + Trichoptera (- Hydropsychidae). However, differences in individual core metrics were balanced out by the VSCI. The median values for the VSCI in the box-and-whisker plots were similar and the interquartile ranges of the VSCI scores in all of the graphs were above 60.

Current data analysis results do not support calibrating the VSCI by season, sampling method, bioregion, or basin size. Additionally, recalibrating the VSCI by one of these classification schemes would lower confidence in the assessment screening value by lowering the number of reference sites available in these categories. Recommending new best standard values for calculating the VSCI is not necessary as this point and but this idea will be revisited in the future when more ProbMon data is available. Results of calibrating the best standard values by bioregion and/or season are predictable. Mountain stream VSCI scores are slightly depressed and Piedmont VSCI scores are slightly higher when calibrated by bioregion and/or season. The overall percentage of streams designated as impacted would not change because the calibrated reference sites adjust in the same manner thereby lowering or raising the assessment cutoff. Percent Plecoptera + Trichoptera (- Hydropsychidae) best standard value will be further reviewed when confidence intervals from probabilistic data yield more precise estimates.

Aquatic Life Use (ALU) tiers established using the Virginia SCI

VDEQ considers VSCI scores above the 10^{th} percentile of reference distribution to be similar to the bulk of reference sites and not impaired. Sites with VSCI scores below the 10^{th} percentile of reference distribution are not similar to most natural streams and considered degraded. Several states use similar percentile of reference distribution methods to determine biological integrity (Ohio; Yoder and Rankin 1995, Florida; Barbour et al. 1996).

The 10th percentile from the probabilistic data set was 58.5 and the 10th percentile from Tetra Tech's analysis of targeted data was 61.3. The average 10th percentile cutoff from both data sets is 59.9. To keep the assessment cutoff simple, the impairment threshold was rounded to 60.

The precision of the VSCI was estimated to be +/- 7.9 scoring units on a 100 point scale. This precision was calculated using data from replicate stations that were collected on the same day.

Aquatic life use tiers were established above and below the impairment threshold (Table 12) based on the average 50th percentile scores from the Tetra Tech reference dataset and the ProbMon reference dataset (upper tier); and the Tetra Tech stressed dataset and ProbMon stressed dataset (lower tier) (Table 13). The aquatic life use tiers that can be discerned using the Virginia SCI are shown in Figure 30. Integrated report assessment decisions and methodology for aquatic life use using the VSCI will be found in VDEQ's Water Quality Assessment Guidance Manual.

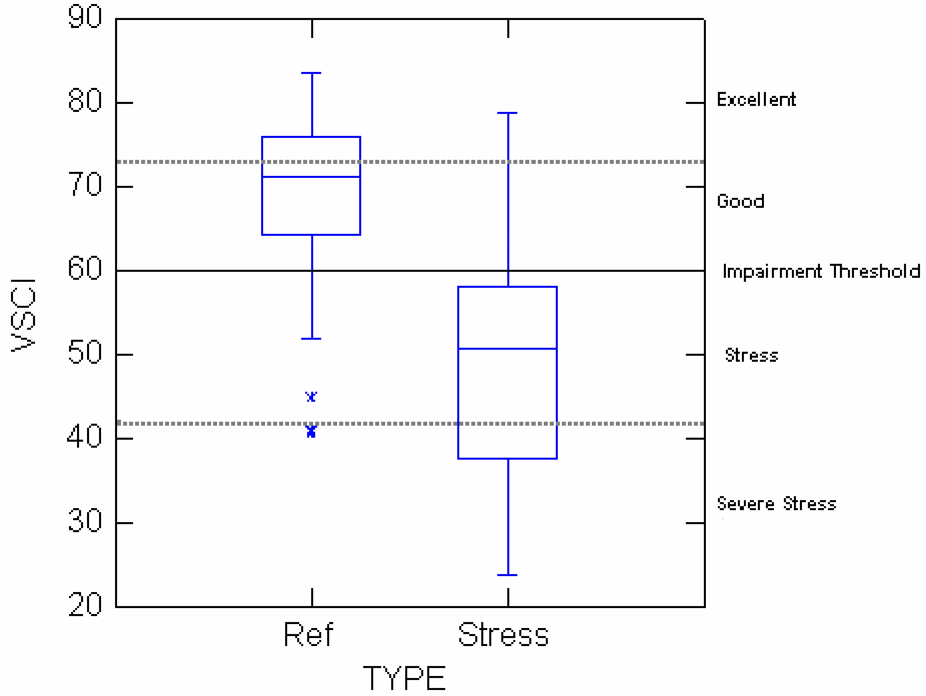
Table 12. Virginia SCI scores and associated aquatic life use (ALU) tiers.

VSCI Score	ALU Tiers
≤ 42	Severe Stress
43 - 59	Stress
60 - 72	Good
≥ 73	Excellent

Table 13. Upper and lower ALU tier determination.

	50th Percentile		50th Percentile
Target Reference	75	Target Stress	35
ProbMon Reference	71	ProbMon Stress	50
Average	**73**	**Average**	**42**

Figure 30. Aquatic life use tiers established for the VSCI. The solid line represents the average 10th percentile from the ProbMon and Tetra Tech data sets.



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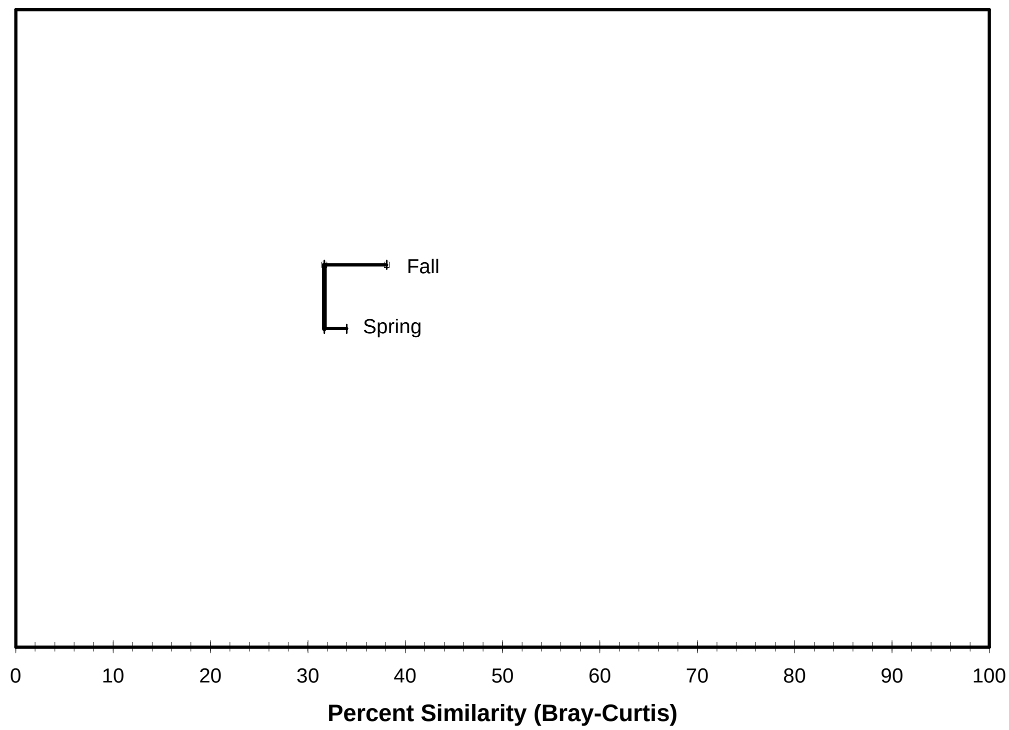
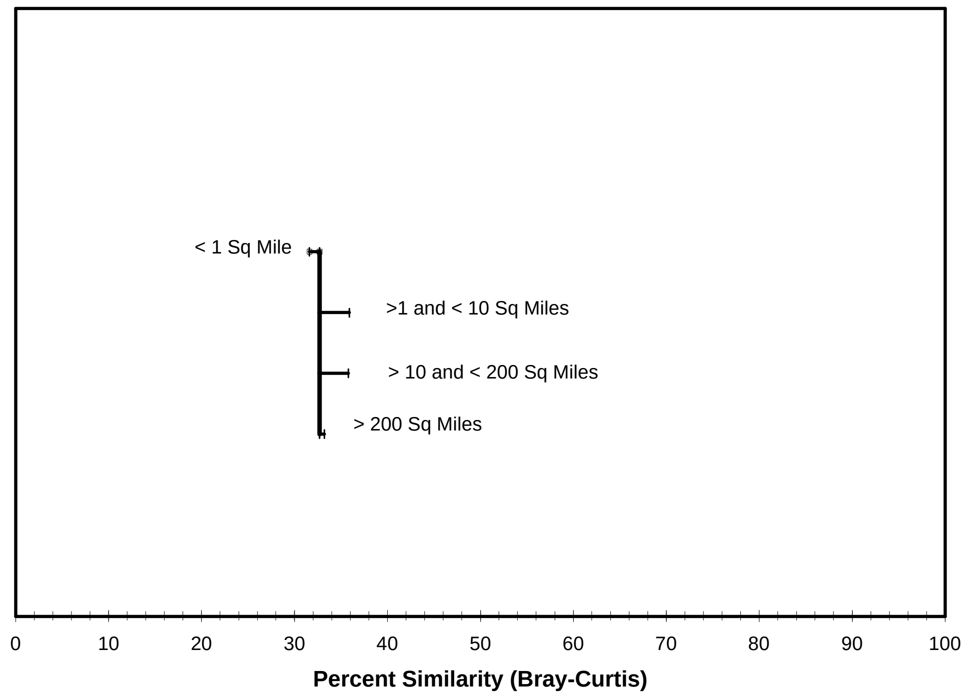
Appendix A. Mean Similarity Dendrogram Results.

Figure 31. Season dendrogram.



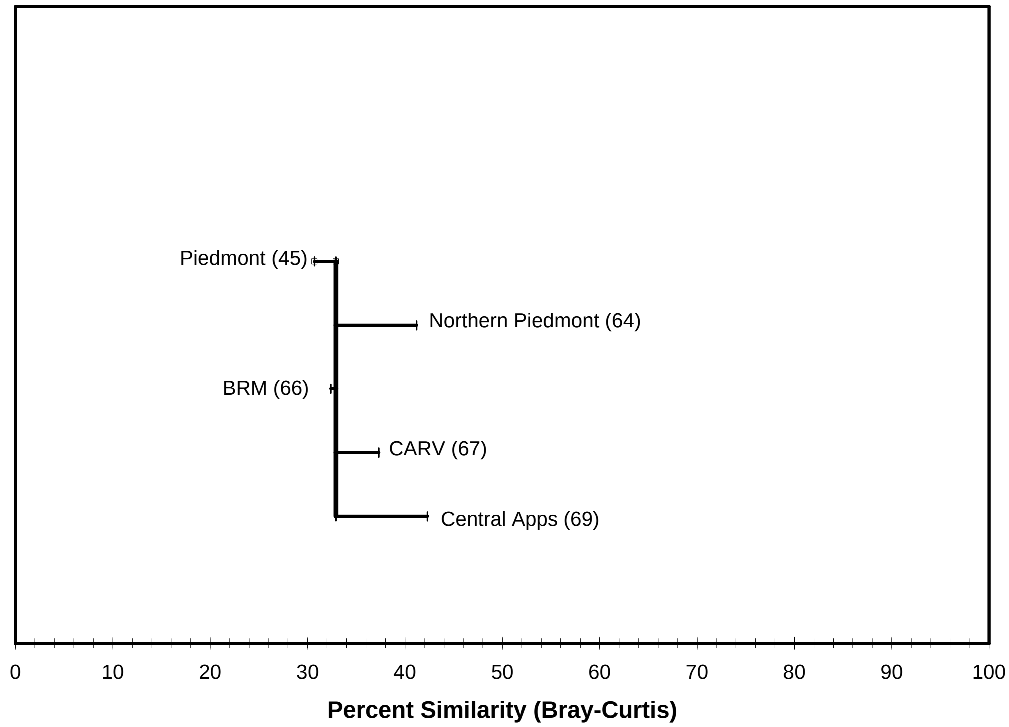
Analysis of reference community data with MeanSim resulted in low (4.3%) classification strength between the spring and fall sampling periods.

Figure 32. Basin size dendrogram.



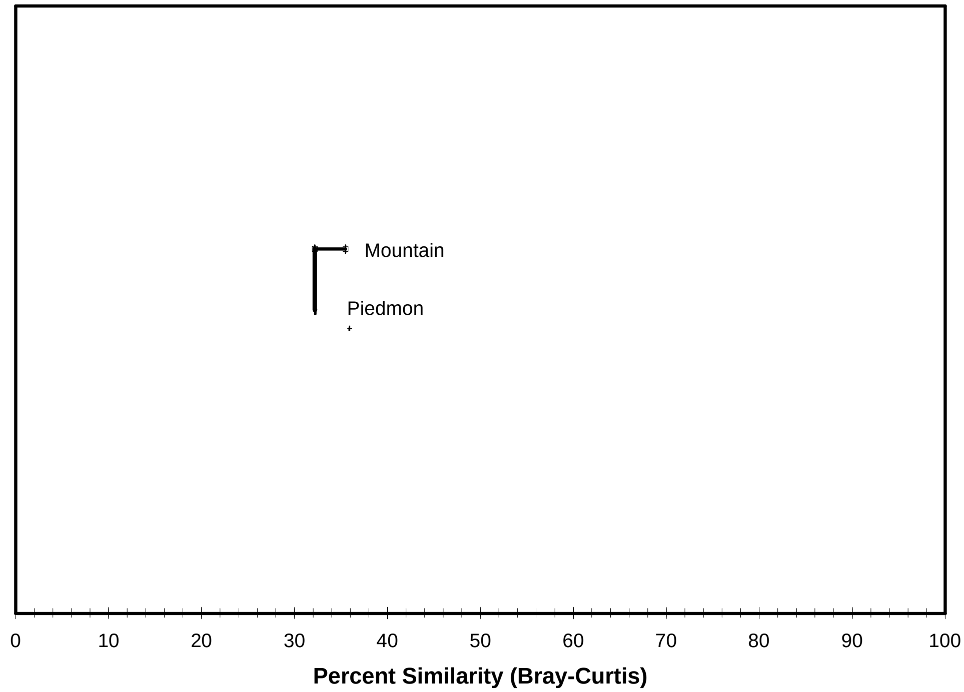
Analysis of reference community data with MeanSim resulted in low (2.3%) classification strength among the four different basin sizes.

Figure 33. Level III Ecoregion dendrogram.



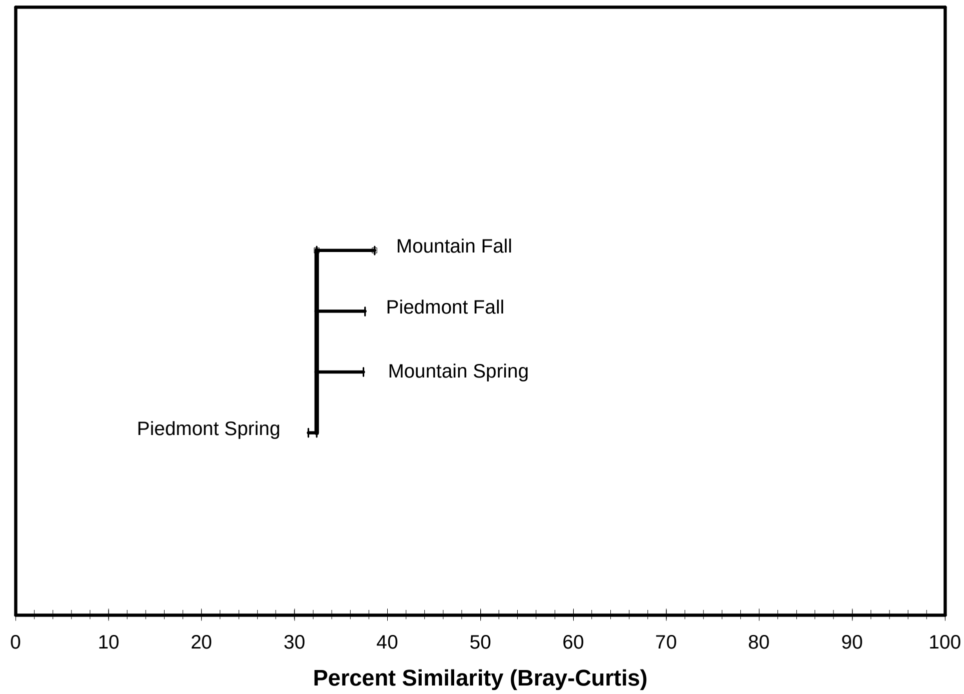
Analysis of reference community data with MeanSim resulted in low (3.5%) classification strength among the five different Level III Ecoregions.

Figure 34. Bioregion dendrogram.



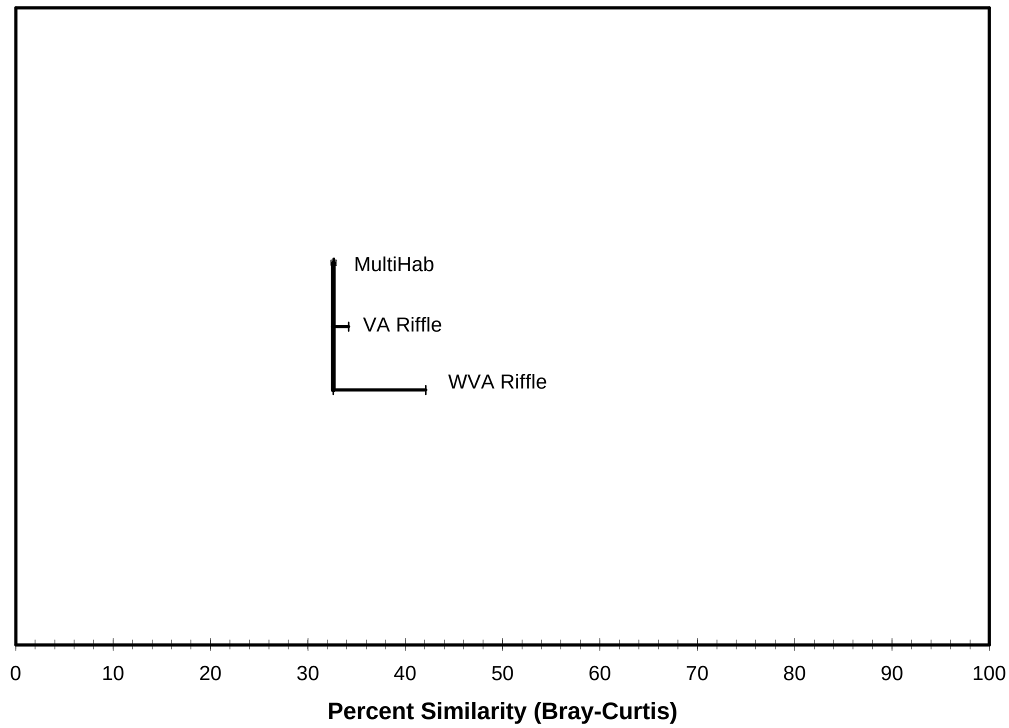
Analysis of reference community data with MeanSim resulted in low (2.3%) classification strength between the Mountain and Piedmont Bioregions.

Figure 35. Bioregion and season dendrogram.



Analysis of reference community data with MeanSim resulted in low (4.4%) classification strength among the different bioregion-season combinations.

Figure 36. Sampling method dendrogram.



Analysis of reference community data with MeanSim resulted in low (2.5%) classification strength among the three different sampling methods.

Appendix B. Average Non-Coastal Core Metric CDF Outputs.

Season	Value	N	Total Taxa	CI-95%	CI+95%
All	5Pct	2	5.4	2.0	8.2
All	10Pct	9	8.5	5.2	9.4
All	25Pct	27	10.9	9.6	11.8
All	50Pct	68	13.7	12.9	14.4
All	75Pct	131	16.0	15.7	17.1
All	90Pct	163	18.2	17.6	18.9
All	95Pct	170	18.9	18.3	21.1
Season	**Value**	**N**	**EPT Taxa**	**CI-95%**	**CI+95%**
All	5Pct	5	1.2	0.0	2.0
All	10Pct	11	2.1	1.1	3.0
All	25Pct	29	3.8	3.4	4.2
All	50Pct	75	6.5	6.0	6.9
All	75Pct	130	8.2	7.7	8.8
All	90Pct	159	9.9	9.1	11.8
All	95Pct	176	11.7	10.1	12.4
Season	**Value**	**N**	**%Ephemeroptera**	**CI-95%**	**CI+95%**
All	5Pct	9	0.0	0.0	1.6
All	10Pct	9	0.4	0.0	3.5
All	25Pct	34	8.5	4.4	13.4
All	50Pct	79	19.7	16.7	21.3
All	75Pct	123	29.3	24.7	33.5
All	90Pct	157	41.5	38.4	45.9
All	95Pct	170	47.7	42.3	52.9
Season	**Value**	**N**	**%PT-Hydrophychidae**	**CI-95%**	**CI+95%**
All	5Pct	8	0.2	0.0	1.0
All	10Pct	19	1.1	0.2	2.0
All	25Pct	58	5.4	2.6	6.9
All	50Pct	115	13.5	9.6	16.7
All	75Pct	150	21.6	19.5	30.0
All	90Pct	179	39.0	30.1	59.1
All	95Pct	184	56.2	39.7	80.3
Season	**Value**	**N**	**%Scrapers**	**CI-95%**	**CI+95%**
All	5Pct	5	0.0	0.0	0.7
All	10Pct	5	0.4	0.0	1.1
All	25Pct	22	7.3	3.3	10.3
All	50Pct	73	18.6	13.9	21.3
All	75Pct	123	30.6	26.4	31.8
All	90Pct	152	37.8	34.9	42.8
All	95Pct	171	46.7	40.5	55.6
Season	**Value**	**N**	**%Chironomidae**	**CI-95%**	**CI+95%**
All	5Pct	9	1.6	0.0	3.3
All	10Pct	24	3.7	1.6	6.2
All	25Pct	59	9.6	6.6	10.9
All	50Pct	115	20.2	14.3	23.8
All	75Pct	154	33.2	27.6	38.8
All	90Pct	172	42.7	39.2	61.1
All	95Pct	184	59.3	43.4	80.1
Season	**Value**	**N**	**% Top 2 Dominant Taxa**	**CI-95%**	**CI+95%**
All	5Pct	6	32.8	21.7	36.1
All	10Pct	18	37.7	32.5	42.4
All	25Pct	53	46.4	44.0	47.6
All	50Pct	102	53.3	51.0	55.7
All	75Pct	153	63.9	59.0	67.5
All	90Pct	179	78.9	68.8	84.0
All	95Pct	184	83.6	79.1	98.3
Season	**Value**	**N**	**HBI (Family Biotic Index**	**CI-95%**	**CI+95%**
All	5Pct	3	2.9	2.5	3.5
All	10Pct	11	3.6	2.9	3.8
All	25Pct	39	4.0	3.8	4.2
All	50Pct	92	4.5	4.3	4.6
All	75Pct	144	5.2	4.9	5.3
All	90Pct	170	5.7	5.5	6.2
All	95Pct	179	6.1	5.7	6.7

Appendix C. Mountain Bioregion Core Metric CDF Outputs by Season.

Season	Value	N	Total Taxa	CI-95%	CI+95%	Season	Value	N	Total Taxa	CI-95%	CI+95%
Spring	5Pct	1	2.6	2.0	6.4	Fall	5Pct	1	5.8	5.5	6.1
Spring	10Pct	4	6.2	2.0	9.3	Fall	10Pct	3	6.9	5.4	8.4
Spring	25Pct	11	10.7	7.2	12.1	Fall	25Pct	13	10.1	7.4	11.9
Spring	50Pct	29	13.5	12.2	15.3	Fall	50Pct	31	13.1	11.9	13.9
Spring	75Pct	49	16.7	15.4	18.3	Fall	75Pct	46	15.1	14.1	16.3
Spring	90Pct	63	18.9	17.7	21.2	Fall	90Pct	69	18.1	15.8	20.2
Spring	95Pct	71	20.4	19.3	21.6	Fall	95Pct	74	19.5	17.5	21.0
Season	**Value**	**N**	**EPT Taxa**	**CI-95%**	**CI+95%**	**Season**	**Value**	**N**	**EPT Taxa**	**CI-95%**	**CI+95%**
Spring	5Pct	2	1.1	1.0	2.8	Fall	5Pct	4	2.3	1.2	2.8
Spring	10Pct	3	2.5	1.0	4.3	Fall	10Pct	8	3.0	1.5	3.5
Spring	25Pct	17	5.7	3.5	7.2	Fall	25Pct	8	4.0	3.4	5.1
Spring	50Pct	42	8.1	7.3	8.7	Fall	50Pct	27	6.4	5.4	7.5
Spring	75Pct	51	9.7	8.8	11.3	Fall	75Pct	51	8.7	7.5	10.4
Spring	90Pct	74	12.2	10.4	15.6	Fall	90Pct	65	10.6	9.4	16.0
Spring	95Pct	77	13.7	11.9	16.0	Fall	95Pct	71	11.3	10.7	13.8
Season	**Value**	**N**	**%Ephemeroptera**	**CI-95%**	**CI+95%**	**Season**	**Value**	**N**	**%Ephemeroptera**	**CI-95%**	**CI+95%**
Spring	5Pct	4	0.0	0.0	9.1	Fall	5Pct	5	0.0	0.0	2.1
Spring	10Pct	4	0.0	0.0	9.4	Fall	10Pct	5	0.0	0.0	5.4
Spring	25Pct	14	11.0	2.1	20.8	Fall	25Pct	15	8.0	0.0	10.8
Spring	50Pct	37	29.4	20.4	36.9	Fall	50Pct	27	17.0	10.8	21.2
Spring	75Pct	62	42.0	39.0	48.7	Fall	75Pct	51	31.6	21.6	47.3
Spring	90Pct	76	61.1	43.8	71.5	Fall	90Pct	69	52.8	34.2	59.4
Spring	95Pct	78	69.0	56.6	73.1	Fall	95Pct	70	53.5	48.1	67.9
Season	**Value**	**N**	**%PT-Hydrophychidae**	**CI-95%**	**CI+95%**	**Season**	**Value**	**N**	**%PT-Hydrophychidae**	**CI-95%**	**CI+95%**
Spring	5Pct	4	0.2	0.0	0.9	Fall	5Pct	11	0.0	0.0	1.7
Spring	10Pct	13	2.0	0.0	4.0	Fall	10Pct	11	0.0	0.0	2.1
Spring	25Pct	38	10.3	3.9	12.6	Fall	25Pct	27	3.6	1.8	7.0
Spring	50Pct	56	16.9	13.0	26.0	Fall	50Pct	48	11.6	7.1	19.4
Spring	75Pct	69	30.8	25.9	52.0	Fall	75Pct	65	23.5	18.0	40.0
Spring	90Pct	78	60.9	36.4	83.6	Fall	90Pct	73	40.4	24.2	63.5
Spring	95Pct	79	70.7	50.9	84.6	Fall	95Pct	76	53.6	38.6	64.5
Season	**Value**	**N**	**%Scrapers**	**CI-95%**	**CI+95%**	**Season**	**Value**	**N**	**%Scrapers**	**CI-95%**	**CI+95%**
Spring	5Pct	2	0.0	0.0	0.9	Fall	5Pct	2	0.0	0.0	6.0
Spring	10Pct	2	0.5	0.0	1.0	Fall	10Pct	2	4.0	0.0	10.0
Spring	25Pct	7	3.3	0.8	10.5	Fall	25Pct	11	15.1	5.7	20.5
Spring	50Pct	24	14.1	10.1	21.7	Fall	50Pct	25	25.4	20.2	37.0
Spring	75Pct	44	25.4	22.2	31.8	Fall	75Pct	49	44.9	37.0	52.2
Spring	90Pct	62	37.2	31.8	43.7	Fall	90Pct	67	64.6	51.5	78.9
Spring	95Pct	71	43.8	39.1	50.6	Fall	95Pct	75	77.0	54.4	86.3
Season	**Value**	**N**	**%Chironomidae**	**CI-95%**	**CI+95%**	**Season**	**Value**	**N**	**%Chironomidae**	**CI-95%**	**CI+95%**
Spring	5Pct	2	1.0	0.0	4.5	Fall	5Pct	3	0.9	0.8	0.9
Spring	10Pct	4	2.6	0.0	6.3	Fall	10Pct	7	1.1	0.6	3.0
Spring	25Pct	23	10.4	5.7	12.4	Fall	25Pct	25	3.9	2.3	5.1
Spring	50Pct	42	17.8	13.1	21.9	Fall	50Pct	47	7.0	5.2	23.1
Spring	75Pct	60	23.8	22.0	35.5	Fall	75Pct	71	33.3	20.6	42.0
Spring	90Pct	75	42.7	31.4	81.0	Fall	90Pct	75	42.1	38.9	57.1
Spring	95Pct	78	62.4	38.9	88.8	Fall	95Pct	76	42.3	41.1	57.1
Season	**Value**	**N**	**% Top 2 Dominant Taxa**	**CI-95%**	**CI+95%**	**Season**	**Value**	**N**	**% Top 2 Dominant Taxa**	**CI-95%**	**CI+95%**
Spring	5Pct	5	36.5	32.6	37.4	Fall	5Pct	1	32.4	30.8	39.8
Spring	10Pct	10	37.8	35.5	40.9	Fall	10Pct	3	38.4	30.8	42.8
Spring	25Pct	26	45.7	39.6	47.6	Fall	25Pct	16	44.6	40.4	51.1
Spring	50Pct	43	49.9	47.7	54.7	Fall	50Pct	45	56.3	51.0	64.9
Spring	75Pct	64	59.3	54.7	78.4	Fall	75Pct	65	69.2	63.2	79.0
Spring	90Pct	77	79.0	71.5	100.0	Fall	90Pct	73	79.5	76.2	93.8
Spring	95Pct	79	92.8	78.3	100.0	Fall	95Pct	75	89.1	78.7	93.8
Season	**Value**	**N**	**HBI (Family Biotic Index**	**CI-95%**	**CI+95%**	**Season**	**Value**	**N**	**HBI (Family Biotic Index**	**CI-95%**	**CI+95%**
Spring	5Pct	2	2.5	2.3	2.8	Fall	5Pct	1	3.0	3.0	3.2
Spring	10Pct	3	2.7	2.4	3.4	Fall	10Pct	8	3.6	2.9	3.8
Spring	25Pct	11	3.6	3.0	3.8	Fall	25Pct	20	3.9	3.8	4.1
Spring	50Pct	25	3.9	3.8	4.3	Fall	50Pct	39	4.3	4.1	4.5
Spring	75Pct	60	4.7	4.3	5.1	Fall	75Pct	62	4.8	4.5	5.0
Spring	90Pct	72	5.2	4.8	5.8	Fall	90Pct	71	5.1	4.9	5.7
Spring	95Pct	78	5.6	5.2	6.1	Fall	95Pct	75	5.2	5.0	5.7

Appendix D. Piedmont Bioregion Core Metric CDF Outputs by Season.

Season	Value	N	Total Taxa	CI-95%	CI+95%	Season	Value	N	Total Taxa	CI-95%	CI+95%
Spring	5Pct	2	7.3	5.0	8.6	Fall	5Pct	2	6.1	6.0	8.7
Spring	10Pct	5	8.6	5.0	9.8	Fall	10Pct	4	7.8	6.0	9.6
Spring	25Pct	12	10.8	9.5	11.6	Fall	25Pct	13	10.6	9.4	12.4
Spring	50Pct	47	13.1	11.9	14.2	Fall	50Pct	34	14.6	13.0	15.5
Spring	75Pct	76	16.5	15.3	17.5	Fall	75Pct	54	17.0	15.7	17.7
Spring	90Pct	90	18.2	17.1	20.3	Fall	90Pct	64	17.9	17.2	24.0
Spring	95Pct	90	19.0	17.8	22.0	Fall	95Pct	79	19.1	18.0	20.9
Season	**Value**	**N**	**EPT Taxa**	**CI-95%**	**CI+95%**	**Season**	**Value**	**N**	**EPT Taxa**	**CI-95%**	**CI+95%**
Spring	5Pct	2	0.8	0.0	1.5	Fall	5Pct	3	0.0	0.0	1.0
Spring	10Pct	5	1.5	0.5	2.0	Fall	10Pct	3	0.8	0.0	1.8
Spring	25Pct	20	3.1	2.1	3.6	Fall	25Pct	13	2.8	1.6	4.0
Spring	50Pct	45	5.0	3.9	6.3	Fall	50Pct	30	4.7	4.2	5.4
Spring	75Pct	69	7.4	6.5	8.3	Fall	75Pct	56	6.8	5.7	7.8
Spring	90Pct	94	9.1	7.8	9.9	Fall	90Pct	72	8.3	7.5	11.3
Spring	95Pct	94	9.6	8.5	12.0	Fall	95Pct	72	8.9	8.0	14.0
Season	**Value**	**N**	**%Ephemeroptera**	**CI-95%**	**CI+95%**	**Season**	**Value**	**N**	**%Ephemeroptera**	**CI-95%**	**CI+95%**
Spring	5Pct	8	0.0	0.0	1.0	Fall	5Pct	10	0.0	0.0	1.2
Spring	10Pct	8	0.0	0.0	1.5	Fall	10Pct	10	0.0	0.0	2.6
Spring	25Pct	19	6.1	1.0	9.3	Fall	25Pct	16	3.3	1.7	6.1
Spring	50Pct	43	17.8	10.8	19.2	Fall	50Pct	43	18.4	6.7	22.4
Spring	75Pct	60	23.0	19.8	34.9	Fall	75Pct	61	26.3	23.3	33.8
Spring	90Pct	83	38.5	34.7	54.9	Fall	90Pct	77	37.3	33.0	45.4
Spring	95Pct	93	53.4	40.2	59.2	Fall	95Pct	83	45.1	35.8	62.2
Season	**Value**	**N**	**%PT-Hydrophychidae**	**CI-95%**	**CI+95%**	**Season**	**Value**	**N**	**%PT-Hydrophychidae**	**CI-95%**	**CI+95%**
Spring	5Pct	10	0.0	0.0	1.1	Fall	5Pct	14	0.0	0.0	0.0
Spring	10Pct	10	0.0	0.0	1.5	Fall	10Pct	14	0.0	0.0	0.3
Spring	25Pct	22	2.9	1.3	6.5	Fall	25Pct	16	0.8	0.0	2.2
Spring	50Pct	53	11.5	6.9	15.5	Fall	50Pct	41	6.5	2.5	10.6
Spring	75Pct	78	19.8	15.8	22.8	Fall	75Pct	63	13.2	10.8	21.7
Spring	90Pct	92	26.1	22.6	40.1	Fall	90Pct	76	22.2	18.7	30.1
Spring	95Pct	96	30.8	23.9	49.4	Fall	95Pct	79	25.4	22.6	41.9
Season	**Value**	**N**	**%Scrapers**	**CI-95%**	**CI+95%**	**Season**	**Value**	**N**	**%Scrapers**	**CI-95%**	**CI+95%**
Spring	5Pct	3	0.0	0.0	1.5	Fall	5Pct	5	0.0	0.0	1.3
Spring	10Pct	3	0.2	0.0	3.9	Fall	10Pct	5	0.0	0.0	1.7
Spring	25Pct	20	6.3	3.3	6.9	Fall	25Pct	15	4.8	1.3	12.0
Spring	50Pct	40	11.2	7.9	18.8	Fall	50Pct	37	16.9	12.3	24.4
Spring	75Pct	75	23.9	19.5	29.6	Fall	75Pct	62	29.3	26.1	35.9
Spring	90Pct	90	31.7	27.1	39.4	Fall	90Pct	79	41.9	34.6	48.0
Spring	95Pct	94	35.4	31.6	76.4	Fall	95Pct	82	45.2	41.9	52.3
Season	**Value**	**N**	**%Chironomidae**	**CI-95%**	**CI+95%**	**Season**	**Value**	**N**	**%Chironomidae**	**CI-95%**	**CI+95%**
Spring	5Pct	9	0.0	0.0	2.1	Fall	5Pct	8	0.0	0.0	2.2
Spring	10Pct	10	0.8	0.0	4.7	Fall	10Pct	12	2.6	0.9	4.1
Spring	25Pct	28	6.8	4.5	10.8	Fall	25Pct	30	8.3	4.4	9.5
Spring	50Pct	55	24.3	13.7	31.2	Fall	50Pct	51	15.9	12.1	17.6
Spring	75Pct	79	37.0	33.7	50.5	Fall	75Pct	72	34.2	17.7	65.9
Spring	90Pct	97	60.5	44.3	80.3	Fall	90Pct	85	72.9	45.9	78.2
Spring	95Pct	100	64.1	59.8	90.1	Fall	95Pct	86	74.4	67.5	78.6
Season	**Value**	**N**	**% Top 2 Dominant Taxa**	**CI-95%**	**CI+95%**	**Season**	**Value**	**N**	**% Top 2 Dominant Taxa**	**CI-95%**	**CI+95%**
Spring	5Pct	1	25.3	21.7	35.6	Fall	5Pct	4	29.9	25.5	31.3
Spring	10Pct	9	35.8	22.4	38.4	Fall	10Pct	10	33.1	29.6	35.2
Spring	25Pct	26	43.9	38.3	46.6	Fall	25Pct	20	39.4	35.1	47.0
Spring	50Pct	53	53.3	47.8	57.8	Fall	50Pct	55	54.5	47.1	58.8
Spring	75Pct	78	63.2	58.3	74.4	Fall	75Pct	72	64.2	58.9	75.8
Spring	90Pct	95	80.4	71.7	87.8	Fall	90Pct	83	78.3	73.6	92.9
Spring	95Pct	96	81.7	77.9	93.1	Fall	95Pct	85	85.1	76.6	92.9
Season	**Value**	**N**	**HBI (Family Biotic Index**	**CI-95%**	**CI+95%**	**Season**	**Value**	**N**	**HBI (Family Biotic Index**	**CI-95%**	**CI+95%**
Spring	5Pct	8	3.7	3.7	3.8	Fall	5Pct	2	3.6	3.4	3.9
Spring	10Pct	11	3.8	3.7	4.1	Fall	10Pct	6	3.9	3.4	4.1
Spring	25Pct	26	4.2	4.1	4.4	Fall	25Pct	21	4.3	3.9	4.6
Spring	50Pct	55	4.8	4.6	5.1	Fall	50Pct	44	5.0	4.6	5.3
Spring	75Pct	83	5.6	5.2	5.8	Fall	75Pct	70	5.9	5.4	6.2
Spring	90Pct	93	6.1	5.8	7.3	Fall	90Pct	81	6.3	6.1	6.8
Spring	95Pct	97	6.4	6.1	7.8	Fall	95Pct	84	6.7	6.3	7.9

Appendix E. Reference station information.

Ref Station	Size	Region	Eco1	Eco3	Basin	LatDD	LongDD	CollMeth	CollDate	Gradient	ALTER	BANKS	BANKVEG	COVER	EMBED	FLOW	RIFFLES	RIPVEG	SEDIMENT	VELOCITY	POOLSUB	POOLVAR	SINUOSITY	TotHabSc	Ave_Temp	Ave_DO	Ave_pH	Ave_Cond
1ANOG000.91f	95.96	NRO	Piedmont	64	Potomac	39.0446	-77.6598	RPB II	8/31/2004	High	20	14	19	17	9	20	11	19	8	18	-1	-1	-1	155	14.28	10.27	7.04	149.00
1ANOG000.91s	95.96	NRO	Piedmont	64	Potomac	39.0446	-77.6598	RBP II	5/6/2004	High	20	14	19	17	9	20	11	19	8	18	-1	-1	-1	155	14.28	10.27	7.04	149.00
1BCDR010.21f	117.52	VRO	Mountain	67	Shenadoah	39.0613	-78.3460	RBP II	11/4/2002	High	19	14	20	18	15	19	13	20	16	19	-1	-1	-1	173	10.70	10.95	7.70	131.00
1BCDR010.21s	117.52	VRO	Mountain	67	Shenadoah	39.0613	-78.3460	RBP II	5/6/2002	High	19	14	20	18	15	19	13	20	16	19	-1	-1	-1	173	10.70	10.95	7.70	131.00
1BCDR027.54f	30.64	VRO	Mountain	67	Shenandoah	39.0217	-78.4598	RPB II	10/13/2004	High	19	17	16	17	11	19	19	19	13	19	-1	-1	-1	169	12.80	9.80	6.40	102.00
1BCDR027.54s	30.64	VRO	Mountain	67	Shenandoah	39.0217	-78.4598	RBP II	4/19/2004	High	19	17	16	17	11	19	19	19	13	19	-1	-1	-1	169	12.80	9.80	6.40	102.00
1BNFS102.55f	75.32	VRO	Mountain	67	Shenadoah	38.7032	-78.9206	RBP II	10/24/2002	High	19	12	17	19	18	19	19	14	16	17	-1	-1	-1	170	15.15	10.10	7.50	104.00
1BNFS102.55s	75.32	VRO	Mountain	67	Shenadoah	38.7032	-78.9206	RBP II	5/9/2002	High	19	12	17	19	18	19	19	14	16	17	-1	-1	-1	170	15.15	10.10	7.50	104.00
1BNKW001.97f	6.07	VRO	Mountain	66	Shenandoah	38.4832	-78.5161	RPB II	10/15/2003	High	19	15	20	19	17	20	19	19	20	10	-1	-1	-1	178	9.95	10.25	7.25	50.50
1BNKW001.97s	6.07	VRO	Mountain	66	Shenandoah	38.4832	-78.5161	RBP II	4/8/2003	High	19	15	20	19	17	20	19	19	20	10	-1	-1	-1	178	9.95	10.25	7.25	50.50
2-BNF003.52f	0.71	SCRO	Mountain	66	James	37.7188	-79.2018	RBP II	10/22/2001	High	20	20	18	20	16	20	20	18	15	19	-1	-1	-1	186	9.69	11.05	7.46	13.75
2-BNF003.52s	0.71	SCRO	Mountain	66	James	37.7188	-79.2018	RBP II	4/3/2001	High	20	20	18	20	16	20	20	18	15	19	-1	-1	-1	186	9.69	11.05	7.46	13.75
2-COO002.35f	0.70	SCRO	Piedmont	45	James	37.5195	-78.5234	MACS	11/1/2001	High	20	16	16	18	20	16	18	20	19	19	-1	-1	-1	182	9.20	9.11	6.36	62.00
2-COO002.35s	0.70	SCRO	Piedmont	45	James	37.5195	-78.5234	MACS	4/11/2001	High	20	16	16	18	20	16	18	20	19	19	-1	-1	-1	182	9.20	9.11	6.36	62.00
2-CWP023.28f	49.97	VRO	Mountain	67	James	37.9383	-79.7211	RBP II	10/22/2001	High	20	16	15	19	18	15	10	11	18	20	-1	-1	-1	162	14.50	10.80	8.05	137.00
2-CWP023.28s	49.97	VRO	Mountain	67	James	37.9383	-79.7211	RBP II	5/15/2001	High	20	16	15	19	18	15	10	11	18	20	-1	-1	-1	162	14.50	10.80	8.05	137.00
2-CWP053.78f	28.88	VRO	Mountain	67	James	38.0998	-79.6498	RBP II	10/11/2001	High	20	20	20	18	20	20	13	20	20	20	-1	-1	-1	191	13.10	10.85	8.10	135.50
2-CWP053.78s	28.88	VRO	Mountain	67	James	38.0998	-79.6498	RBP II	5/30/2001	High	20	20	20	18	20	20	13	20	20	20	-1	-1	-1	191	13.10	10.85	8.10	135.50
2-DCK003.94f	1.70	WCRO	Mountain	67	James	37.4633	-80.3483	RPB II	8/16/2004	High	20	20	20	20	19	20	20	20	15	15	-1	-1	-1	189	14.79	9.40	6.16	0.19
2-DCK003.94s	1.70	WCRO	Mountain	67	James	37.4633	-80.3483	RBP II	6/1/2004	High	20	20	20	20	19	20	20	20	15	15	-1	-1	-1	189	14.79	9.40	6.16	0.19
2-HAZ006.34f	5.59	SCRO	Piedmont	45	James	37.4798	-79.1712	MACS	10/22/2001	Low	20	12	14	15	-1	15	-1	20	10	-1	17	10	13	146	19.00	9.14	7.76	80.60
2-HAZ006.34s	5.59	SCRO	Piedmont	45	James	37.4798	-79.1712	MACS	5/10/2001	Low	20	12	14	15	-1	15	-1	20	10	-1	17	10	13	146	19.00	9.14	7.76	80.60
2-JKS028.69f	434.28	WCRO	Mountain	67	James	37.8227	-79.9894	RPB II	11/1/2004	High	20	17	18	19	20	20	20	12	19	20	-1	-1	-1	185	11.76	10.99	8.02	159.00
2-JKS028.69s	434.28	WCRO	Mountain	67	James	37.8227	-79.9894	RBP II	5/13/2004	High	20	17	18	19	20	20	20	12	19	20	-1	-1	-1	185	11.76	10.99	8.02	159.00
2-JOB001.02f	13.09	WCRO	Mountain	67	James	37.5030	-80.1150	RBP II	10/9/2001	High	20	18	17	19	17	19	20	12	15	18	-1	-1	-1	175	9.75	10.83	8.03	100.75
2-JOB001.02s	13.09	WCRO	Mountain	67	James	37.5030	-80.1150	RBP II	4/20/2001	High	20	18	17	19	17	19	20	12	15	18	-1	-1	-1	175	9.75	10.83	8.03	100.75
2-LIJ003.06f	0.24	VRO	Mountain	66	James	37.8849	-79.1589	RPB II	8/26/2004	High	14	18	19	14	11	13	20	13	17	12	-1	-1	-1	151	5.80	11.10	7.60	146.00
2-LIJ003.06s	0.24	VRO	Mountain	66	James	37.8849	-79.1589	RBP II	4/1/2004	High	14	18	19	14	11	13	20	13	17	12	-1	-1	-1	151	5.80	11.10	7.60	146.00
2-MIW003.45f	15.76	VRO	Mountain	67	James	37.9966	-79.7119	RPB II	8/25/2004	High	20	18	20	16	17	18	16	20	15	18	-1	-1	-1	178	10.10	11.20	6.10	54.00
2-MIW003.45s	15.76	VRO	Mountain	67	James	37.9966	-79.7119	RBP II	4/28/2004	High	20	18	20	16	17	18	16	20	15	18	-1	-1	-1	178	10.10	11.20	6.10	54.00
2-OGL005.53f	1.69	WCRO	Mountain	67	James	37.8399	-80.1225	RBP II	10/9/2001	High	20	17	18	15	20	14	20	14	15	15	-1	-1	-1	168	13.95	10.14	8.24	73.85
2-OGL005.53s	1.69	WCRO	Mountain	67	James	37.8399	-80.1225	RBP II	5/1/2001	High	20	17	18	15	20	14	20	14	15	15	-1	-1	-1	168	13.95	10.14	8.24	73.85
2-PTR005.13f	8.16	WCRO	Mountain	67	James	37.6226	-79.8901	RPB II	10/8/2003	High	20	12	18	20	20	20	20	20	10	20	-1	-1	-1	180	13.73	8.16	6.44	31.55
2-PTR005.13s	8.16	WCRO	Mountain	67	James	37.6226	-79.8901	RBPII	4/3/2003	High	20	12	18	20	20	20	20	20	10	20	-1	-1	-1	180	13.73	8.16	6.44	31.55
2-RED003.65f	16.68	WCRO	Piedmont	45	James	37.5089	-79.3835	RPB II	9/14/2004	High	20	19	20	19	16	20	20	13	8	20	-1	-1	-1	175	10.30	12.37	6.45	34.30
2-RED003.65s	16.68	WCRO	Piedmont	45	James	37.5089	-79.3835	RBP II	4/28/2004	High	20	19	20	19	16	20	20	13	8	20	-1	-1	-1	175	10.30	12.37	6.45	34.30
2-RKF026.13f	94.46	VRO	Piedmont	64	James	37.8670	-78.8220	RPB II	9/16/2004	High	20	13	18	14	11	17	13	18	7	17	-1	-1	-1	148	17.80	9.90	6.00	56.00
2-RKF026.13s	94.46	VRO	Piedmont	64	James	37.8670	-78.8220	RBP II	5/12/2004	High	20	13	18	14	11	17	13	18	7	17	-1	-1	-1	148	17.80	9.90	6.00	56.00
2-SMR004.80f	0.41	VRO	Mountain	66	James	37.9349	-79.0880	RBP II	10/17/2001	High	20	18	20	20	19	17	20	20	19	19	-1	-1	-1	192	10.55	10.00	6.50	15.50
2-SMR004.80s	0.41	VRO	Mountain	66	James	37.9349	-79.0880	RBP II	5/29/2001	High	20	18	20	20	19	17	20	20	19	19	-1	-1	-1	192	10.55	10.00	6.50	15.50
2-STH000.50f	117.83	VRO	Mountain	67	James	37.7730	-79.3781	RBP II	10/25/2002	High	20	20	20	18	16	20	18	14	17	15	-1	-1	-1	178	13.00	10.25	7.85	232.00
2-STH000.50s	117.83	VRO	Mountain	67	James	37.7730	-79.3781	RBP II	5/14/2002	High	20	20	20	18	16	20	18	14	17	15	-1	-1	-1	178	13.00	10.25	7.85	232.00
2-STV000.48f	4.52	WCRO	Mountain	67	James	37.6205	-80.1868	RPB II	9/13/2004	High	15	19	19	17	13	20	20	14	15	17	-1	-1	-1	169	6.80	11.11	6.75	31.20
2-STV000.48s	4.52	WCRO	Mountain	67	James	37.6205	-80.1868	RBP II	4/15/2004	High	15	19	19	17	13	20	20	14	15	17	-1	-1	-1	169	6.80	11.11	6.75	31.20
2-TYE008.77f	197.39	VRO	Piedmont	45	James	37.6332	-78.9033	RPB II	11/4/2004	High	19	19	19	17	11	19	18	19	17	18	-1	-1	-1	176	14.70	10.90	6.00	32.00
2-TYE008.77s	197.39	VRO	Piedmont	45	James	37.6332	-78.9033	RBP II	5/6/2004	High	19	19	19	17	11	19	18	19	17	18	-1	-1	-1	176	14.70	10.90	6.00	32.00
2-TYS000.85f	15.13	VRO	Mountain	66	James	37.8561	-79.0585	RBP II	10/15/2002	High	19	20	20	19	13	17	19	18	19	18	-1	-1	-1	182	10.50	10.20	7.10	17.00
2-TYS000.85s	15.13	VRO	Mountain	66	James	37.8561	-79.0585	RBP II	5/21/2002	High	19	20	20	19	13	17	19	18	19	18	-1	-1	-1	182	10.50	10.20	7.10	17.00
2-WLN006.90f	17.55	VRO	Mountain	67	James	37.8974	-79.8037	RBP II	10/29/2002	High	19	17	20	19	18	18	19	20	19	18	-1	-1	-1	187	14.05	9.60	6.90	35.00
2-WLN006.90s	17.55	VRO	Mountain	67	James	37.8974	-79.8037	RBP II	5/15/2002	High	19	17	20	19	18	18	19	20	19	18	-1	-1	-1	187	14.05	9.60	6.90	35.00
2-XUF000.55s	0.22	WCRO	Mountain	67	James	37.7998	-79.8486	RBP II	4/17/2002	High	20	18	18	17	20	12	20	20	15	15	-1	-1	-1	175	11.41	9.52	6.41	45.60
3-RAP008.71f	656.12	NRO	Piedmont	64	Rappahannock	38.3716	-77.7243	RBP II	10/10/2001	High	20	16	18	18	16	16	16	18	15	20	-1	-1	-1	173	18.61	9.23	8.10	71.00
3-RAP008.71s	656.12	NRO	Piedmont	64	Rappahannock	38.3716	-77.7243	RBP II	4/24/2001	High	20	16	18	18	16	16	16	18	15	20	-1	-1	-1	173	18.61	9.23	8.10	71.00
3-RAP028.98s	499.03	NRO	Piedmont	64	Rappahannock	38.3562	-77.9557	RBP II	5/13/2004	High	20	13	15	17	16	20	13	20	17	19	-1	-1	-1	170	23.55	8.54	7.48	70.60

Appendix E. Reference station information continued.

Ref Station	Size	Region	Eco1	Eco3	Basin	LatDD	LongDD	CollMeth	CollDate	Gradient	ALTER	BANKS	BANKVEG	COVER	EMBED	FLOW	RIFFLES	RIPVEG	SEDIMENT	VELOCITY	POOLSUB	POOLVAR	SINUOSITY	TotHabSc	Ave_Temp	Ave_DO	Ave_pH	Ave_Cond
3-ROB005.42f	151.35	NRO	Piedmont	64	Rappahannock	38.3508	-78.1143	RBP II	10/18/2001	High	20	18	18	18	17	18	18	18	17	19	-1	-1	-1	181	14.90	11.15	7.25	73.50
3-ROB005.42s	151.35	NRO	Piedmont	64	Rappahannock	38.3508	-78.1143	RBP II	4/10/2001	High	20	18	18	18	17	18	18	18	17	19	-1	-1	-1	181	14.90	11.15	7.25	73.50
4ABEE001.20s	5.56	PRO	Piedmont	45	Roanoke	36.5434	-78.6323	MACS	5/7/2002	High	20	16	18	14	13	10	9	20	12	15	-1	-1	-1	147	17.48	8.47	6.85	92.50
4ABOR033.22f	53.49	WCRO	Piedmont	45	Roanoke	37.3851	-79.4631	RPB II	11/24/2003	High	17	15	16	14	13	20	20	20	5	20	-1	-1	-1	160	7.80	11.41	7.80	53.10
3-RAP028.98s	499.03	NRO	Piedmont	64	Rappahannock	38.3562	-77.9557	RBP II	5/13/2004	High	20	13	15	17	16	20	13	20	17	19	-1	-1	-1	170	23.55	8.54	7.48	70.60
3-ROB005.42f	151.35	NRO	Piedmont	64	Rappahannock	38.3508	-78.1143	RBP II	10/18/2001	High	20	18	18	18	17	18	18	18	17	19	-1	-1	-1	181	14.90	11.15	7.25	73.50
3-ROB005.42s	151.35	NRO	Piedmont	64	Rappahannock	38.3508	-78.1143	RBP II	4/10/2001	High	20	18	18	18	17	18	18	18	17	19	-1	-1	-1	181	14.90	11.15	7.25	73.50
4ABEE001.20s	5.56	PRO	Piedmont	45	Roanoke	36.5434	-78.6323	MACS	5/7/2002	High	20	16	18	14	13	10	9	20	12	15	-1	-1	-1	147	17.48	8.47	6.85	92.50
4ABOR033.22f	53.49	WCRO	Piedmont	45	Roanoke	37.3851	-79.4631	RPB II	11/24/2003	High	17	15	16	14	13	20	20	20	5	20	-1	-1	-1	160	7.80	11.41	7.80	53.10
4ABOR033.22s	53.49	WCRO	Piedmont	45	Roanoke	37.3851	-79.4631	RBPII	3/11/2003	High	17	15	16	14	13	20	20	20	5	20	-1	-1	-1	160	7.80	11.41	7.80	53.10
4AEKH003.18f	2.41	SCRO	Piedmont	45	Roanoke	36.8662	-79.1402	RBP II	10/30/2001	Low	20	11	20	19	-1	14	-1	20	10	-1	13	10	15	152	10.75	10.00	7.44	80.00
4AEKH003.18s	2.41	SCRO	Piedmont	45	Roanoke	36.8662	-79.1402	RBP II	5/15/2001	Low	20	11	20	19	-1	14	-1	20	10	-1	13	10	15	152	10.75	10.00	7.44	80.00
4ALBT003.07f	2.44	WCRO	Piedmont	45	Roanoke	36.7923	-80.1653	RPB II	9/20/2004	High	19	10	10	16	11	20	20	17	5	15	-1	-1	-1	143	10.36	10.46	7.19	39.46
4ALBT003.07s	2.44	WCRO	Piedmont	45	Roanoke	36.7923	-80.1653	RBP II	4/15/2004	High	19	10	10	16	11	20	20	17	5	15	-1	-1	-1	143	10.36	10.46	7.19	39.46
4ASRV012.19f	10.26	SCRO	Piedmont	45	Roanoke	36.6480	-79.5516	RBP II	10/30/2001	Low	20	9	9	16	-1	18	-1	19	10	-1	13	15	17	146	14.95	9.53	7.67	60.55
4ASRV012.19s	10.26	SCRO	Piedmont	45	Roanoke	36.6480	-79.5516	RBP II	6/4/2001	Low	20	9	9	16	-1	18	-1	19	10	-1	13	15	17	146	14.95	9.53	7.67	60.55
5AFON024.32f	93.66	PRO	Piedmont	45	Chowan	36.6201	-77.5758	MACS	11/6/2003	Low	20	14	12	11	-1	18	-1	18	10	-1	8	13	10	134	15.12	9.21	6.49	46.20
5AFON024.32s	93.66	PRO	Piedmont	45	Chowan	36.6201	-77.5758	MACS	4/28/2003	Low	20	14	12	11	-1	18	-1	18	10	-1	8	13	10	134	15.12	9.21	6.49	46.20
5ANMR007.11s	143.01	PRO	Piedmont	45	Chowan	36.8702	-78.2983	MACS	5/6/2002	High	20	12	14	15	5	12	14	20	10	15	-1	-1	-1	137	14.68	9.84	7.04	63.00
5ANTW093.62f	510.57	PRO	Piedmont	45	Chowan	36.8594	-77.5871	RPB II	11/18/2004	High	20	19	20	14	13	20	12	19	12	17	-1	-1	-1	166	20.45	7.99	6.84	76.00
5ANTW093.62s	510.57	PRO	Piedmont	45	Chowan	36.8594	-77.5871	RBP II	4/26/2004	High	20	19	20	14	13	20	12	19	12	17	-1	-1	-1	166	20.45	7.99	6.84	76.00
5ARSK003.66f	25.99	PRO	Piedmont	45	Chowan	36.5961	-77.7728	MACS	10/4/2004	Low	20	14	14	14	-1	17	-1	19	10	-1	11	16	12	147	16.04	7.73	6.35	70.00
5ARSK003.66s	25.99	PRO	Piedmont	45	Chowan	36.5961	-77.7728	MACS	4/29/2004	Low	20	14	14	14	-1	17	-1	19	10	-1	11	16	12	147	16.04	7.73	6.35	70.00
6AFOX001.69f	3.10	SRO	Mountain	69	Big Sandy	37.1592	-82.1662	RPB II	10/26/2004	High	18	13	16	17	16	17	16	16	12	11	-1	-1	-1	152	11.30	10.30	7.60	178.00
6AFOX001.69s	3.10	SRO	Mountain	69	Big Sandy	37.1592	-82.1662	RBP II	5/5/2004	High	18	13	16	17	16	17	16	16	12	11	-1	-1	-1	152	11.30	10.30	7.60	178.00
6BLSR004.78f	6.00	SRO	Mountain	69	Tennessee	36.8721	-82.4734	RPB II	10/6/2004	High	19	16	18	19	17	16	16	18	17	11	-1	-1	-1	167	15.26	8.99	6.64	25.00
6BLSR004.78s	6.00	SRO	Mountain	69	Tennessee	36.8721	-82.4734	RBP II	5/10/2004	High	19	16	18	19	17	16	16	18	17	11	-1	-1	-1	167	15.26	8.99	6.64	25.00
6CSFH084.73f	123.39	SRO	Mountain	67	Holston	36.6916	-81.7718	RBP II	11/6/2002	High	18	11	14	17	15	17	13	12	17	19	-1	-1	-1	153	13.24	10.90	8.10	196.50
6CSFH084.73s	123.39	SRO	Mountain	67	Holston	36.6916	-81.7718	RBP II	5/7/2002	High	18	11	14	17	15	17	13	12	17	19	-1	-1	-1	153	13.24	10.90	8.10	196.50
6CSFH098.10f	74.72	SRO	Mountain	67	Holston	36.7653	-81.6213	RPB II	10/27/2004	High	18	12	11	18	17	18	10	11	16	17	-1	-1	-1	148	14.30	9.60	7.90	161.00
6CSFH098.10s	74.72	SRO	Mountain	67	Holston	36.7653	-81.6213	RBP II	5/11/2004	High	18	12	11	18	17	18	10	11	16	17	-1	-1	-1	148	14.30	9.60	7.90	161.00
9-DDD006.61f	6.41	WCRO	Mountain	66	New	36.8854	-80.3187	RPB II	10/9/2003	High	20	12	17	17	13	20	20	18	12	17	-1	-1	-1	166	12.35	10.24	7.36	62.15
9-DDD006.61s	6.41	WCRO	Mountain	66	New	36.8854	-80.3187	RBPII	3/6/2003	High	20	12	17	17	13	20	20	18	12	17	-1	-1	-1	166	12.35	10.24	7.36	62.15
9-SFK002.81f	15.98	SRO	Mountain	67	New	36.9848	-81.1875	RPB II	8/18/2004	High	19	14	18	19	18	19	16	18	16	17	-1	-1	-1	174	8.28	10.76	6.58	61.00
9-SFK002.81s	15.98	SRO	Mountain	67	New	36.9848	-81.1875	RBP II	4/28/2004	High	19	14	18	19	18	19	16	18	16	17	-1	-1	-1	174	8.28	10.76	6.58	61.00
9-WFC010.66f	207.11	WCRO	Mountain	67	New	37.2789	-80.9254	RBP II	10/11/2001	High	20	20	20	19	18	18	18	16	15	20	-1	-1	-1	184	14.25	9.68	8.14	200.60
9-WFC010.66s	207.11	WCRO	Mountain	67	New	37.2789	-80.9254	RBP II	5/8/2001	High	20	20	20	19	18	18	18	16	15	20	-1	-1	-1	184	14.25	9.68	8.14	200.60
9-WLK024.17f	192.95	WCRO	Mountain	67	New	37.2025	-80.7858	RBP II	10/11/2001	High	20	20	19	15	20	20	20	20	20	20	-1	-1	-1	194	17.25	9.82	8.30	238.05
9-WLK024.17s	192.95	WCRO	Mountain	67	New	37.2025	-80.7858	RBP II	6/6/2001	High	20	20	19	15	20	20	20	20	20	20	-1	-1	-1	194	17.25	9.82	8.30	238.05
9-XDP000.65f	0.20	SRO	Mountain	66	New	36.6665	-81.1332	RPB II	9/25/2003	High	19	17	20	15	16	17	15	20	16	10	-1	-1	-1	165	15.64	9.18	7.39	103.20
9-XDP000.65s	0.20	SRO	Mountain	66	New	36.6665	-81.1332	RBP II	4/17/2003	High	19	17	20	15	16	17	15	20	16	10	-1	-1	-1	165	15.64	9.18	7.39	103.20