VIRGINIA WATER RESOURCES RESEARCH CENTER

Development of Aquatic Life Use Assessment Protocols for Class VII Waters in Virginia

2017 Report of the Academic Advisory Committee for Virginia Department of Environmental Quality

SPECIAL REPORT

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Virginia Water Resources Research Center (MC 0444)
210 Cheatham Hall, Virginia Tech
310 West Campus Drive
Blacksburg, VA 24061
(540) 231-5624
FAX: (540) 231-6673
E-mail: water@vt.edu

Stephen Schoenholtz, Director

DEVELOPMENT OF
AQUATIC LIFE USE ASSESSMENT PROTOCOLS
FOR
CLASS VII WATERS IN VIRGINIA

2017 Report of the Academic Advisory Committee for Virginia Department of Environmental Quality

by:

Andrew L. Garey
Chesapeake Bay Tributary Project Coordinator
Virginia Department of Environmental Quality

Greg C. Garman
Member, Academic Advisory Committee
Associate Professor, Department of Biology
Director, VCU Rice Rivers Center
Virginia Commonwealth University

Leonard A. Smock
Member, Academic Advisory Committee
Professor Emeritus, Department of Biology
Virginia Commonwealth University

Edited by:
Jane L. Walker

Publication of the
Virginia Water Resources Research Center
210 Cheatham Hall, Virginia Tech
310 West Campus Drive
Blacksburg, VA 24061

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Members of the 2017 Academic Advisory Committee to the Virginia Department of Environmental Quality

Stephen H. Schoenholtz, Chair
Virginia Water Resources Research Center / Department of Forest Resources and Environmental Conservation
Virginia Tech

E. Fred Benfield
Department of Biology
Virginia Tech

Paul Bukaveckas
Department of Biology / Rice Rivers Center / Center for Environmental Studies
Virginia Commonwealth University

Gregory C. Garman
Department of Biology / Center for Environmental Studies
Virginia Commonwealth University

Carl Hershner
Department of Biology / Center for Coastal Resources Management
Virginia Institute of Marine Science
College of William and Mary

Wu-Seng Lung
Department of Civil and Environmental Engineering
University of Virginia

Kevin J. McGuire
Virginia Water Resources Research Center / Department of Forest Resources and Environmental Conservation
Virginia Tech

Daniel McLaughlin
Virginia Water Resources Research Center / Department of Forest Resources and Environmental Conservation
Virginia Tech

Durelle T. Scott IV
Department of Biological Systems Engineering
Virginia Tech

Leonard A. Shabman
Resources for the Future

Eric P. Smith
Department of Statistics
Virginia Tech

Leonard A. Smock
Department of Biology
Virginia Commonwealth University

Meredith Steele
Department of Crop and Soil Environmental Sciences
Virginia Tech

Kurt Stephenson
Department of Agricultural and Applied Economics
Virginia Tech

Jane L. Walker
Virginia Water Resources Research Center
Virginia Tech

Kang Xia
Department of Crop and Soil Environmental Sciences
Virginia Tech

Carl E. Zipper
Department of Crop and Soil Environmental Sciences
Virginia Tech

Introduction

This report details the progress made in fiscal year (FY) 2017 by the Academic Advisory Committee (AAC) toward the development of a biological assessment index for Class VII swamp waters in the Coastal Plain physiographic province of Virginia. The goal of this work was to provide a biological assessment index that can be used by the Virginia Department of Environmental Quality (DEQ) for monitoring that informs the agency's semi-annual 305(b)/303(d) report to the U.S. Environmental Protection Agency (EPA). Through previous work, the AAC has developed and validated a working index using fish assemblage data to evaluate anthropogenic impairment of Class VII waters in Virginia's Chowan River Basin. In addition, that work provided preliminary evidence that macroinvertebrate assemblages in the region may have utility for assessing water quality impairment.

Based on a consensus among AAC members and DEQ staff, past efforts have focused on Class VII sites within Virginia's Chowan River Basin, which drains to Albemarle Sound in North Carolina. Because the initial development phase has been successfully completed for that basin, it was determined that FY 2017 efforts focus on collecting data, data analysis, and index development for Class VII sites outside the Chowan Basin. Thus, this report details efforts during FY 2017 on Class VII waters that are within Virginia's Chesapeake Bay watershed. The specific tasks addressed during FY 2017 and reported here are as follows:

Task 1: Identify gaps in the existing database for Class VII waters within Virginia's Chesapeake Bay watershed to complete the remaining tasks.

Task 2: Fill data gaps with a combination of data mining and targeted field work.

> Subtask 2.1: Data Mining and Site Reconnaissance. Query available datasets (VCU INSTAR and DEQ EDAS) to determine potential sites to be included in a Chesapeake Bay watershed database, including sites for which data already exist and candidate sites for sampling to provide new data. Develop land cover data for all sites using GIS. Reconnoiter the new sites to determine their suitability for inclusion in the database prior to field sampling.
>
> Subtask 2.2: Compile Existing Data. Compile a working database for sites to be used to develop a biological assessment index for Chesapeake Bay watershed Class VII waters.
>
> Subtask 2.3: Sample Class VII sites in the Chesapeake Bay watershed to provide data for the index development. Give priority to sites that are preliminarily classified as either reference or altered sites.

Task 3: Analyze the data available after 2017 field sampling, focused on developing a preliminary biological assessment index for Class VII waters in the Chesapeake Bay watershed.

Methods

Study Sites

Seventeen swamp sites within the Chesapeake Bay watershed in the Coastal Plain of Virginia were included in this investigation (Table 1, Figure 1). Sites were selected based on review of electronic online map resources and, if available, past field notes that indicated sites were likely swamp waters. Priority was given to sites that appeared to be surrounded by either heavily developed or heavily forested watersheds in order to assess the effectiveness of the newly developed biological assessment index for detecting anthropogenic effects. At four of the sites, archival fish data from the VCU INSTAR database (http://gis.vcu.edu/instar/) and habitat and water chemistry data from the AAC's FY 2013 investigation (Garman *et al*. 2013) were used. Archival data included one collection from 2001 and three from 2007. At the remaining 13 sites, new fish, habitat, and water chemistry data were collected between November 17, 2016 and April 14, 2017. The complete fish assemblage dataset used in this study is included in Appendix A.

Data on benthic macroinvertebrates were available for two of the four sites for which archived data in the VCU Instar database were available. Benthic macroinvertebrates were collected at each of the 13 sites sampled during 2016-2017. Together, these sites add 15 macroinvertebrate collections to the database for this project. Identification of benthic macroinvertebrates is ongoing, and data from these samples are not presented in this report.

Water Chemistry

A YSI multimeter was used to record surface water pH, dissolved oxygen (DO), temperature, and specific conductance (SpC) at each sampling site. At the 13 sites visited in 2016-2017, water samples were collected for analysis of total nitrogen (TN) and total phosphorus (TP).

Habitat Variables

The eight-metric Blackwater Habitat Protocol (BHP; Garey *et al*. 2014) was used to evaluate habitat conditions at each site. The BHP is a rapid, field-based method to identify swamp systems using characteristics such as channel and floodplain geomorphology, hydrology, and vegetation. Habitat data were used to determine if relationships occur between natural habitat variability and fish assemblage metrics. Existence of relationships could help determine which metrics might be confounded by such natural variability.

Fish Collections

Fish collections were made by experienced ichthyologists (S. McIninch and D. Hopler, VCU) following EPA Protocols (Barbour *et al*. 1999). At each site, a single pass was made using a Smith-Root Model LR-24 direct-current backpack electrofisher. The sampling area at each site encompassed 100 meters along the main channel of each system, as well as several sweeps in backwater habitat adjacent to the channel.

Watershed Land Cover

All geospatial analyses were conducted using ARCMAP, Version 10.1. Watersheds were delineated using 10-m digital elevation models downloaded from the United States Geological

Survey National Elevation Dataset (http://ned.usgs.gov/). Watershed land cover data for the archival sites were downloaded from the 2011 National Land Cover Database (NLCD; http://www.mrlc.gov/nlcd2011.php), and land cover data for new sites were downloaded from the 2011-2015 Virginia Geographic Information Network Database (VGIN; http://vgin.maps.arcgis.com). Land cover data were clipped to the watershed boundaries and quantified for each watershed.

The original classifications employed by the NLCD and VGIN were aggregated to reflect four basic classifications: *agriculture, developed, forest,* and *other* (additional details of this process available upon request). After the reclassification, the percentages of the total land cover area within each class were calculated for each watershed. The forest class was used as the overall gradient of anthropogenic influence within each watershed. Sites were preliminarily classified as *reference* if their watersheds consisted of >70% forest, *intermediate* for watersheds consisting of 50-70% forest, or *altered* for those consisting of <50% forest (Garey *et al*. 2015, 2016). Based solely on the percentage of forest within the watershed, seven sites would have been classified as reference, five would have been intermediate, and five sites would have been classified as altered (Table 1).

Fish Index Development

A total of 41 candidate fish assemblage metrics were calculated to develop a fish-based, multimetric index of biotic integrity (IBI) for Chesapeake Bay watershed swamp sites. Metrics were selected to include fish assemblage abundance, evenness, richness, and diversity, as well as feeding, habitat use, spawning, individual condition, and pollution tolerance. Ecological information on fish species was derived from Jenkins and Burkhead (1994) and other published sources. Final decisions regarding the traits of each species were based on the best professional judgments of VCU fish biologists (Dr. Steve McIninch and Dr. Greg Garman). Ecological information used in this investigation is included in Appendix B.

The response of each metric to stress was determined based on simple linear correlations between percent forest land cover and raw metric values (*i.e.*, an increase or decrease in metric values with increasing watershed disturbance). Metrics were then scaled as described by Blocksom (2003), such that maximum and minimum values for each metric were set at the 97.5 and 2.5 percentiles of the entire dataset, respectively. The percent comparability of each raw metric value to the endpoints was used as the final metric score. Final scores were between 0 and 100, with higher scores indicating greater ecological integrity. Metrics with raw values that decreased with stress were scored using the equation:

$$(Raw\ Value - Floor)/(Ceiling - Floor)$$

where *Raw Value* represents the raw metric value, *Floor* represents the 2.5 percentile, and *Ceiling* represents the 97.5 percentile. Metrics with raw values that increased with stress were scored using the equation:

$$(Ceiling - Raw\ Value)/(Ceiling - Floor)$$

The final IBI model was constructed by selecting the metric score combination (arithmetic mean of metric scores) from the set of 41 initial candidate metrics that yielded the highest correlation with the percentage of forest cover within each watershed. To effectively achieve this result, a code script was developed using R, version 3.1 (R Core Team 2014), following the algorithm presented by Schoolmaster *et al*. (2013). Briefly, this algorithm allows for the selection of the most effective subset from a set of *n* metrics without evaluating all possible combinations, which is prohibitively inefficient.

<u>Fish Index Evaluation</u>

Sites were assigned to site-condition categories (*altered*, *intermediate*, or *reference*; Table 1) based on the reference filters previously agreed upon by the AAC (Table 2) with one exception. The pH thresholds derived from highly tannic and acidic swamp sites in the Chowan Basin may not be appropriate for Chesapeake Bay watershed swamp sites. Thus, the requirements for pH as derived in the Chowan Basin were not used. For a site to be assigned to the reference class, all criteria for which data exist (except pH) have to be satisfied, whereas stressed sites can be designated as altered based on attainment of only one of the listed criteria.

Fish IBI scores were plotted and compared to their pre-assigned condition categories to determine the degree of separation between stressed and reference sites. In addition, the simple linear correlation of the IBI scores against forest land cover was calculated to indicate the overall effectiveness of the index.

<u>Water Chemistry Quality Assurance/Quality Control Evaluation</u>

On May 1, 2017, the field sampling crew was audited to assure compliance with DEQ standard operating procedures by Mr. James Beckley, DEQ Water Quality Monitoring Quality Assurance Officer. Because of limited personnel availability, the audit was conducted at a stream site in Richmond, Va. (Horse Swamp Creek), rather than at one of the sites included in the dataset presented here. Duplicate samples for TN and TP were obtained from Horse Swamp Creek and Dark Swamp to evaluate sampling precision for these parameters (per DEQ protocol, TN and TP results shown for Dark Swamp are from the first replicates obtained).

<u>Results</u>

<u>Water Chemistry</u>

Mean specific conductance (118 µS/cm, std. dev = 82 µS/cm) and pH (6.75, std. dev = 0.68) were higher in Chesapeake Bay watershed swamp sites from this investigation compared to Chowan Basin swamp sites previously investigated (SpC mean = 97 µS/cm, std. dev = 101 µS/cm; pH mean = 5.60, std. dev = 0.88). The overall difference between SpC at Chesapeake Bay and Chowan sites was not significant (Student's T-test, $p = 0.49$). However, the mean pH for Chesapeake Bay sites was significantly higher than that of Chowan sites (Student's T-test, $p<0.001$).

No site in the Chesapeake Bay watershed study exceeded the established threshold of 350 µS/cm used to indicate stressed conditions in the Chowan Basin. Elevated SpC (> 250 µS/cm) occurred at three sites identified as altered based on forest cover: Dark Swamp, France Swamp, and Grays Creek (Table 3). The Beaverdam Creek site, which would have been considered as a reference site from the land-cover data, was classified as an intermediate site for exceeding 150 µS/cm.

The pH criteria established for reference classification in the Chowan Basin would have prevented three additional sites from being classified as reference (Canal Swamp, Cohoke Mill Creek, and Timber Branch Swamp; Table 3). These three sites exhibited near-neutral pH (7.2-7.4) and were well above the previously-selected reference filter cutoff of 6.5. By relaxing the pH requirement, a total of six reference sites were assigned (Table 1). Lodge Creek had an observed pH (8.24) that exceeded the stressed threshold established in the Chowan Basin of 7.5; adherence to this threshold would have changed the site condition rating from intermediate to altered. Because of the overall difference in pH observed between Chesapeake Bay and Chowan Basin sites, the pH filters were not used to re-classify sites.

The observed nitrogen data did not affect any of the initial classifications assigned by forest cover (Table 3). The TN concentrations exceeded the reference threshold of 1.5 mg/L at only three sites (Lodge Creek, Oldham Run, and White Oak Run) and exceeded the stressed threshold of 3.0 mg/L only at Oldham Run.

Total phosphorus was detected at all sites at which samples were taken; however, in most cases, it was not possible to determine whether the Chowan Basin reference threshold for TP was exceeded because the parameter quantification limit (PQL) of the laboratory is 0.1 mg/L and the current reference threshold is 0.05 mg/L. For the eight sites where TP was detected but below the PQL (*i.e.*, TP listed as <0.1 mg/L in Table 3), we conclude the TP concentration was between the method detection limit of 0.02 mg/L and the PQL of 0.1 mg/L. At the unnamed tributary to Piscataway Creek and at White Oak Run, TP was 0.2 mg/L, which exceeded the altered threshold of 0.1 mg/L. The observed TP concentration caused the unnamed tributary to Piscataway Creek to be classed as an altered site. In all other cases, the observed TP concentrations did not change the site condition category assignments that would have resulted from using only land cover.

<u>Water Chemistry Quality Assurance/Quality Control Evaluation</u>

The field audit results and memorandum from Mr. James Beckley concerning the audit are included in Appendices C and D. Mr. Beckley concluded that water chemistry measurement and water sample collection were conducted in general accordance with DEQ standard operating procedures, but he did make several suggestions for changes to ensure data integrity. These changes included modifying the hand placement on sample bottles during filling, and discarding of rinse water downstream of the sampling point to help prevent contamination. Mr. Beckley concluded that these sample-collection issues were minor and were unlikely to affect data integrity. This conclusion is supported by the results of the replicate TN and TP analysis. The TN concentrations were 0.90 and 0.83 mg/L in the two replicates collected at Dark Swamp and 1.14 and 1.24 mg/L in the two replicates collected at Horse Swamp Creek. All replicate TP concentrations were below the PQL of 0.1 mg/L. At both sites, the relative percent difference (RPD; the difference in measurements divided by mean of measurements) was 8% for TN and below the quantification limit for TP. Therefore, sampling precision was greater than the DEQ required minimum sampling precision of 10% RPD.

<u>Fish IBI</u>

The best-performing fish index, which included 12 metrics (Table 4), exhibited a strong statistically significant relationship with percent forest land cover (r^2= 0.87, $p<0.001$). The proportion of individuals that were introduced species increased with forest land cover in the dataset, which is contrary to what is expected. After further examination, it was determined that this effect was driven largely by a single site: an unnamed tributary to Diascund Creek. This site was located approximately 4 km (2.5 mi) above the Diascund Reservoir, which is regularly stocked for recreational fishing. Although watershed land cover draining to the site was 87% forested, the fish assemblage was dominated by bluegill sunfish, which comprised 62% of fish in the sample (103 of 166 individuals). Bluegill are non-native to these swamp waters and are regularly stocked in most reservoirs for recreational fishing and as forage for other species. Based on this finding and the potential for a confounding effect of nearby impoundments on the detection of impairment, the proportion of introduced individuals was eliminated from the recommended final index.

The final recommended 11-metric index exhibited a strong relationship with watershed forest land cover (r^2= 0.80, $p<0.001$), and there was no overlap between scores of *a priori*-assigned reference and altered sites (Figure 2). The index included metrics that encompass several important aspects of ecological integrity, including stress tolerance (2 metrics), ecological habit (4 metrics), individual condition (1 metric), community evenness (3 metrics), and diversity and whether species were native to the Chesapeake Bay watershed (1 metric; Table 4).

The 11-metric index included several metrics that may be considered theoretically redundant, most importantly, the three measures of community evenness and the two metrics pertaining to vegetation spawners. Therefore, a second index was generated that removed theoretically redundant items so that only the metric with the strongest relationship to watershed forest land cover was included. The resulting index included Simpson Evenness (all species) and percentage of vegetation spawners but not the other two community evenness metrics nor the number of vegetation spawners. This change produced a simpler 8-metric index that explained the majority of the variation among sites attributable to forest land cover (r^2= 0.54, $p<0.001$; Figure 3) with no overlap between altered and reference sites.

Neither the 11-metric nor the 8-metric IBI was significantly related with the total BHP habitat score, any of the eight BHP metrics, or total watershed land area (liner regression for continuous variables, ANOVA for categorical variables, all p-values > 0.10). This result indicates that the indices show no strong evidence of being confounded by natural environmental variability among swamp sites.

Discussion

This investigation provides a new addition in our understanding of swamp waters in Virginia. Reports by the AAC in previous years detailed the development of a decision-support tool for biologists to rapidly identify likely Class VII waters (the BHP), as well as the development of a working index of biotic integrity for Chowan Basin swamps (the Blackwater Condition Index, BCI). The results presented here show strong evidence that it is also possible to evaluate anthropogenic influence on Chesapeake Bay watershed swamp systems by evaluating fish assemblages. Given the small number of sites included in this investigation, however, these results should be considered preliminary, most especially because the indices developed here were not validated with an independent set of study sites.

Recommendations

Based on the results presented here and the progress made thus far toward the development of criteria for aquatic life use in swamps, we make the following recommendations for future discussion and investigation:

1) Implementation plan for Blackwater Condition Index (index for Chowan Basin sites only). We recommend a plan be developed that details additional testing and validation required for the BCI, and a time-specific plan for finalizing the BCI for regular use in DEQ water quality monitoring and assessment. Development of this plan should begin with the AAC but should subsequently include input from DEQ and EPA biologists. It is important that the implementation plan includes clear guidance for assessors and other agency staff on the recommended proper use of the BCI for regulatory assessment.

2) Further development of IBI's for swamp sites in the Chesapeake Bay watershed. Additional site investigations are needed to refine and validate the results presented here. In addition, the reference filters used to pre-classify Chesapeake Bay watershed swamps into site condition classes should be revisited. Several sites with predominantly forested watersheds and no apparent causes of impairment exhibited pH values above the Chowan River Basin reference threshold of 6.5. Further investigation is needed to determine whether such observations should be considered naturally occurring and whether the reference filter values should be amended. Furthermore, the presence and effects of impoundments on IBI performance should be evaluated for the swamp sites included in this project. Sites directly below man-made impoundments have been excluded from this and previous investigations. However, results presented here indicate that impoundments within watersheds adjacent to or below study sites might have confounding effects, most especially on fish assemblages given the ability of fish to exhibit long-distance dispersal.

3) Further development of macroinvertebrate-based indices for assessment of swamp waters. Preliminary results from 11 Chowan Basin study sites indicated that the macroinvertebrate assemblages were more strongly related to watershed land cover alteration than fish assemblages. Time and resources have prevented a more thorough evaluation of the potential for

using macroinvertebrates for assessment of swamp waters. However, collection of macroinvertebrate samples at swamp sites is ongoing. A more comprehensive evaluation of the effectiveness of macroinvertebrate-based indices in swamp waters should be conducted once these data are developed.

Literature Cited

Barbour, M.T., J. Gerritsen, B.D. Snyder, and J.B. Stribling. 1999. *Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers*. USEPA, Washington, D.C.

Blocksom, K.A. 2003. A performance comparison of metric scoring methods for a multimetric index for Mid-Atlantic Highlands streams. *Environmental Management* **31**: 0670-0682.

Garman, G.C., L.A. Smock, A.L. Garey, and S.P. McIninch. 2013. *Development of Aquatic Life Use Assessment Protocols for Class VII Waters in Virginia: 2013 Report of the Academic Advisory Committee for Virginia Department of Environmental Quality.* SR53-2013. November 2013. Virginia Water Resources Research Center, Blacksburg, VA.

Garey, A.L., G.C. Garman, and L.A. Smock. 2014. *Development of Aquatic Life Use Assessment Protocols for Class VII Waters in Virginia: 2014 Report of the Academic Advisory Committee for Virginia Department of Environmental Quality*. SR55-2014. October 2014. Virginia Water Resources Research Center, Blacksburg, VA.

Garey, A.L., G.C. Garman, and L.A. Smock. 2015. *Development of Aquatic Life Use Assessment Protocols for Class VII Waters in Virginia: 2015 Report of the Academic Advisory Committee for Virginia Department of Environmental Quality*. SR56-2015. June 2015. Virginia Water Resources Research Center, Blacksburg, VA.

Garey, A.L., G.C. Garman, and L.A. Smock. 2016. *Development of Aquatic Life Use Assessment Protocols for Class VII Waters in Virginia: 2016 Report of the Academic Advisory Committee for Virginia Department of Environmental Quality*. SR58-2016. May 2016. Virginia Water Resources Research Center, Blacksburg, VA.

Jenkins, R.E. and N.M. Burkhead. 1994. *Freshwater Fishes of Virginia*. American Fisheries Society, Bethesda, MD.

Schoolmaster, D.R., Jr., J.B. Grace, E.W. Schweiger, G.R. Guntenspergen, B.R. Mitchell, K.M. Miller, and A.M. Little. 2013. An algorithmic and information-theoretic approach to multimetric index construction. *Ecological Indicators* **26**: 14-23. DOI: 10.1016/j.ecolind.2012.10.016.

Table 1: Information on the 17 swamp sites included in the FY 2017 investigation.

Site	River Basin	Date sampled	Lat	Long	Developed (%)	Forest (%)	Agriculture (%)	Site Condition Rating
Beaverdam Creek	James	3/24/2017	37.49	-76.91	2.2%	90.8%	1.6%	Int
Canal Swamp	Potomac	4/12/2017	38.17	-76.91	0.9%	85.5%	11.0%	Ref
Cohoke Mill Creek	York	4/14/2017	37.63	-76.96	1.3%	83.6%	11.6%	Ref
Dark Swamp	James	11/17/2016	37.16	-76.85	5.6%	47.3%	13.1%	Alt
Dragon Run	Ches. Bay	11/9/2007	37.82	-76.91	1.4%	50.7%	31.7%	Int
France Swamp	York	11/18/2016	37.42	-76.78	28.0%	48.5%	17.5%	Alt
Goldenvale Creek	Rappahannock	4/12/2017	38.17	-77.22	1.6%	88.2%	4.8%	Ref
Grays Creek	James	8/16/2007	37.17	-76.87	4.8%	49.8%	3.2%	Alt
Lodge Creek	Potomac	4/3/2017	37.97	-76.54	2.7%	54.9%	32.0%	Int
Mapisco Creek	James	7/31/2007	37.32	-77.02	4.3%	61.2%	20.7%	Int
Oldham Run	Potomac	4/3/2017	38.03	-76.67	1.5%	43.9%	48.6%	Alt
Rumley Marsh	James	3/24/2017	37.51	-77.04	2.8%	87.0%	5.3%	Ref
Timber Branch Swamp	Ches. Bay	3/29/2017	37.69	-76.78	0.4%	93.8%	5.2%	Ref
UT to Diascund Creek	James	3/24/2017	37.48	-76.97	3.4%	87.3%	5.4%	Ref
UT to Piscataway Creek	Rappahannock	4/14/2017	37.87	-77.00	2.1%	68.9%	13.1%	Alt
White Marsh	Ches. Bay	11/7/2001	37.80	-76.80	7.5%	56.0%	32.5%	Int
White Oak Run	Rappahannock	4/12/2017	38.31	-77.39	7.7%	40.6%	31.5%	Alt

Developed, forest, and agriculture show the proportion of watershed land area comprised of each land cover class. Alt, Int, and Ref = altered, intermediate, and reference, respectively. Ches. Bay = swamp is not within a major river basin. UT = unnamed tributary.

Table 2: Reference filters determining site-impairment status; filters are subject to change based on data availability.

Parameter	Reference Threshold	Stressed Threshold
Physicochemistry		
Specific Conductance	<150 µS/cm	>350 µS/cm
Total Nitrogen	<1.5 mg/L	>3 mg/L
Total Phosphorus	<0.05 mg/L	>0.1 mg/L
pH	<6.5	>7.5
Other	No other measured parameters indicate site should be 303d listed	Other chemical stressors present that are likely to affect community
Land Cover		
GIS Land Use/Land Cover	>70 percent forested land cover in watershed	<50 percent forested land cover
Intact Riparian Vegetation	>50 m from both banks	<10 m, either bank, or <25 m from both banks
General Site Characteristics		
Point Sources/Others	No NPDES sites within watershed	NA*
Site Reconnaissance Land Use/Land Cover	No extensive development in the watershed that is likely to impact the system	NA*
Visible System Impairment	No visible signs of direct alteration to the water body (*e.g.*, dams, weirs, levees, artificial channelization)	NA*

NPDES = National Pollutant Discharge Elimination System
NA* = Not applicable; these filters are not typically used to designate systems as stressed.

Table 3: Water chemistry data collected at the 17 swamp sites.

Site	pH	DO	SpC	Temp	TN	TP
Beaverdam Creek	7.00	7.09	165	17.62	0.28	<0.1
Canal Swamp	7.23	11.58	63	23.49	0.59	<0.1
Cohoke Mill Creek	7.41	7.78	115	17.42	0.49	<0.1
Dark Swamp	6.93	10.56	252	9.98	0.9	<0.1
Dragon Run	6.22	8.01	61.9	8.68	NA	NA
France Swamp	6.74	8.54	290	12.13	NA	NA
Goldenvale Creek	6.49	8.43	55	17.96	0.35	<0.1
Grays Creek	7.09	8.6	286	15.29	NA	NA
Lodge Creek	8.24	10.07	101	14.23	1.6	<0.1
Mapisco Creek	6.18	8.7	61	21.13	NA	NA
Oldham Run	6.94	9.73	104	15.25	3.76	BDL
Rumley Marsh	5.64	11.35	33	9.14	0.2	<0.1
Timber Branch Swamp	7.4	8.38	73	20.98	0.45	BDL
UT Diascund Creek	6.35	10.68	85	12.49	0.35	<0.1
UT to Piscataway Creek	6.76	8.7	65	18.94	0.95	0.2
White Marsh	5.39	11.9	73.1	10.32	NA	NA
White Oak Run	6.82	6.41	124	19.45	1.83	0.2

DO = dissolved oxygen in mg/L; SpC = specific conductance in µS/cm; Temp = water temperature in ºC; TN and TP = total nitrogen and total phosphorus, respectively, in mg/L. Site names in bold indicate reference conditions based solely on forest cover. Underlined site names indicate altered conditions based solely on forest cover. Shaded values are above the respective Chowan Basin reference threshold (pH = 6.5; SpC = 150 µS/cm; TN = 1.5 mg/L; TP = 0.05 mg/L). Boxed-in values are above the respective Chowan Basin altered threshold (pH = 7.5; TN = 3 mg/L; TP = 0.1 mg/L). BDL= below the detection limit of 0.02 mg/L. <0.1 indicates the presence of TP, but the concentration is below that which the lab can reliably quantify.

Table 4: Fish metrics selected for inclusion in the preliminary Index of Biotic Integrity.

Metric*	Metrics type	Explanation	Response to Stress†
Sensitive species (%)	Stress tolerance	Percentage of individuals that are stress-sensitive	Increase
Tolerant species (%)	Stress tolerance	Percentage of individuals that are stress-tolerant	Decrease
Vegetation spawners (%)	Ecological habit	Percentage of individuals that spawn on aquatic vegetation	Decrease
Vegetation spawners (n)	Ecological habit	Number of individuals that spawn on aquatic vegetation	Decrease
Facultative air breathers (n)	Ecological habit	Number of individuals that can utilize atmospheric oxygen	Decrease
Invertivores (%)	Ecological habit	Percentage of individuals that are invertivores	Decrease
Anomalies (n)	Individual condition	Number of individuals with deformities, lesions or tumors	Increase
Simpson evenness (natives)	Community evenness	Simpson Diversity Index divided by richness (native species only)	Decrease
Simpson evenness (all species)	Community evenness	Simpson Diversity Index divided by richness (all species)	Decrease
Pielou evenness (natives)	Community evenness	Shannon Diversity Index divided by richness (native species only)	Decrease
Shannon diversity (introduced)	Diversity AND nativeness	Shannon Diversity Index (non-native species only)	Increase
~~Introduced species (%)~~	~~Nativeness~~	~~Percentage of individuals that are non-native~~	~~Decrease~~

Bold metrics were selected for inclusion in the simplified 8-metric IBI. Strikethrough indicates that the metric was eliminated due to potential confounding effects of impoundment influence.

* % = proportion or percentage of individuals; n = number of species.

† Increase / Decrease = response of raw metric values to increasing watershed land cover disturbance.

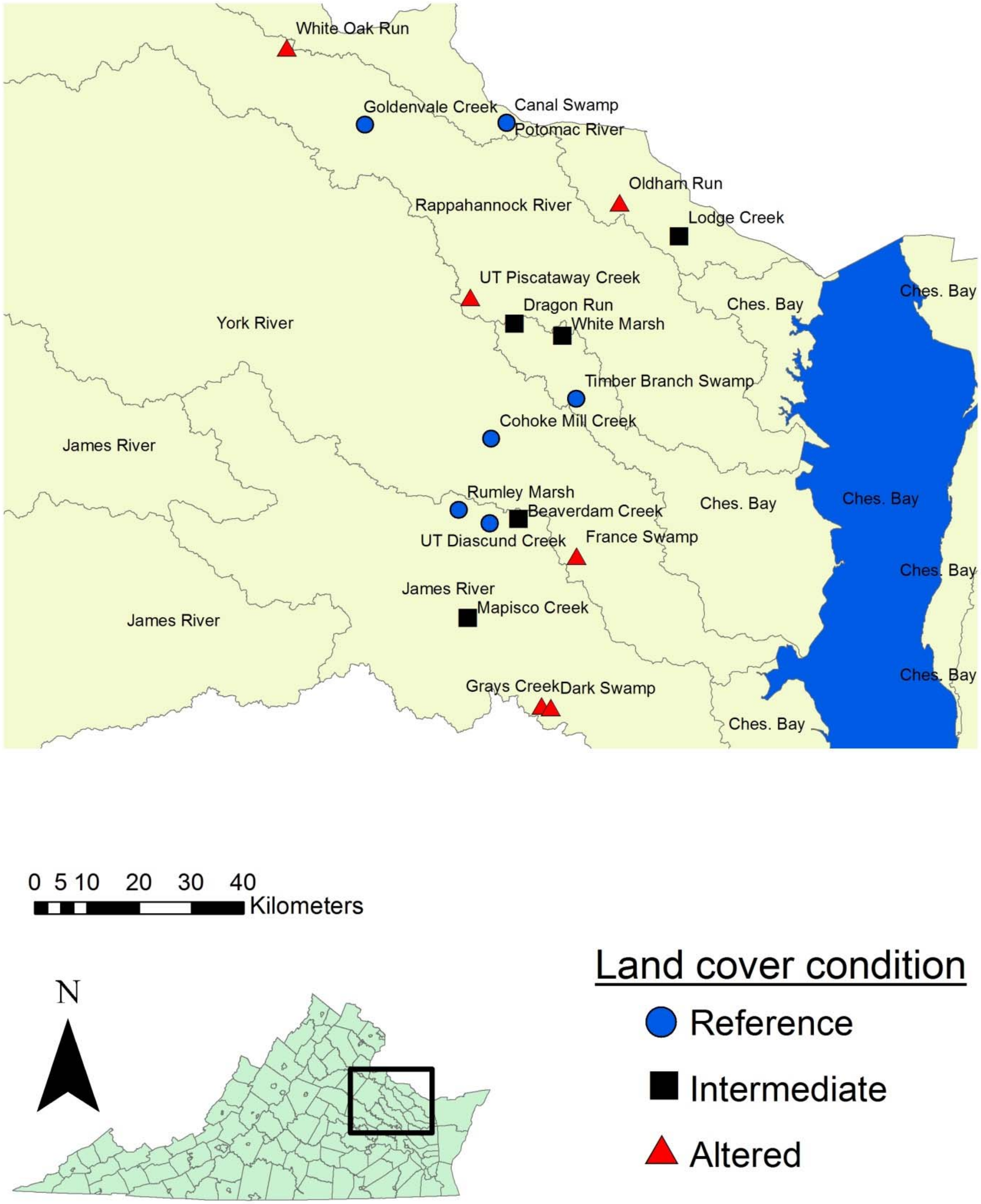

Figure 1: Location of 17 swamp sites included in this investigation. Boundaries indicate major river basins surrounding each site.

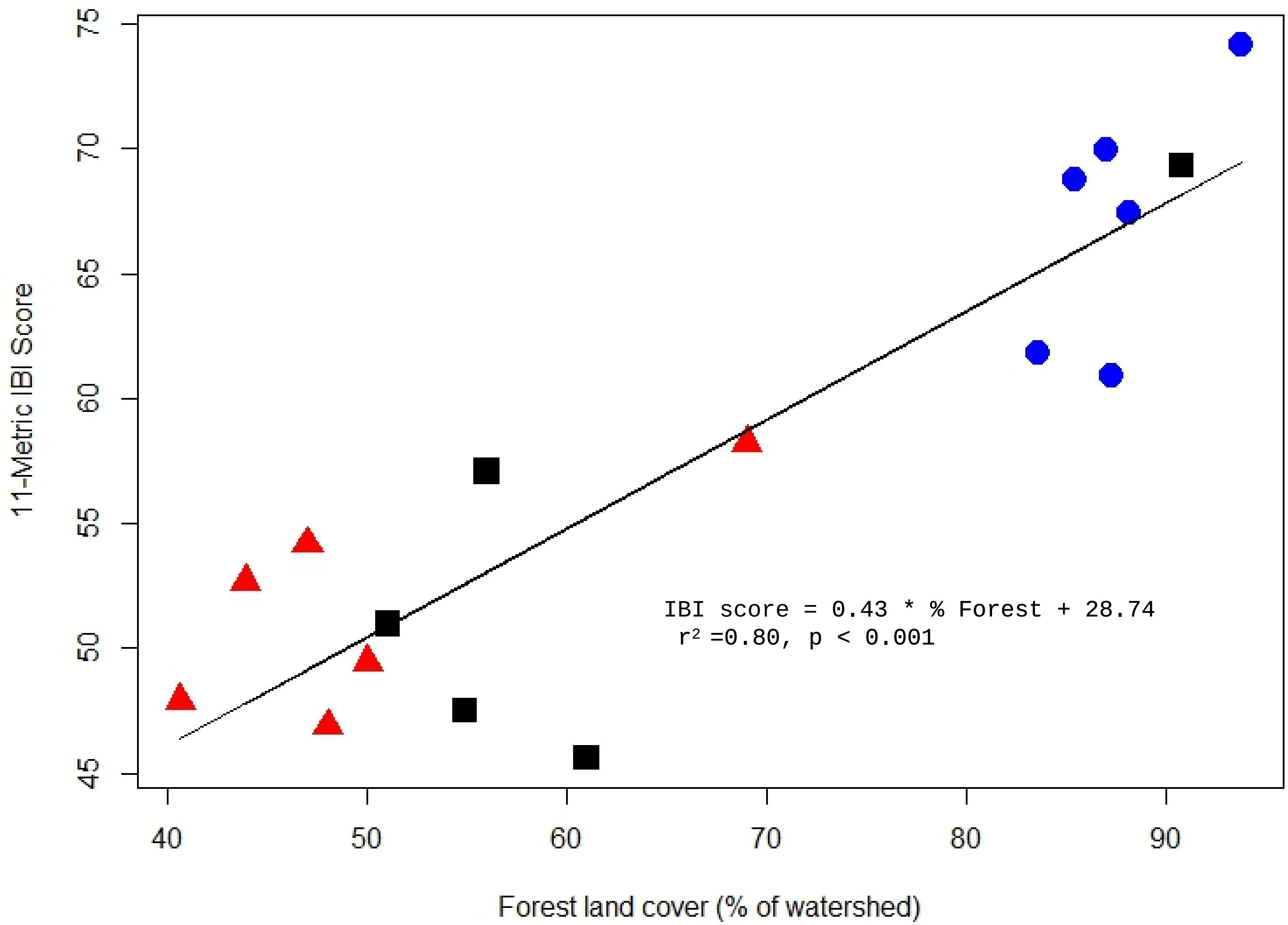

Figure 2: Correlation of 11-metric IBI with percent forest land cover. Red triangles = altered sites, black squares = intermediate sites, blue circles = reference sites.

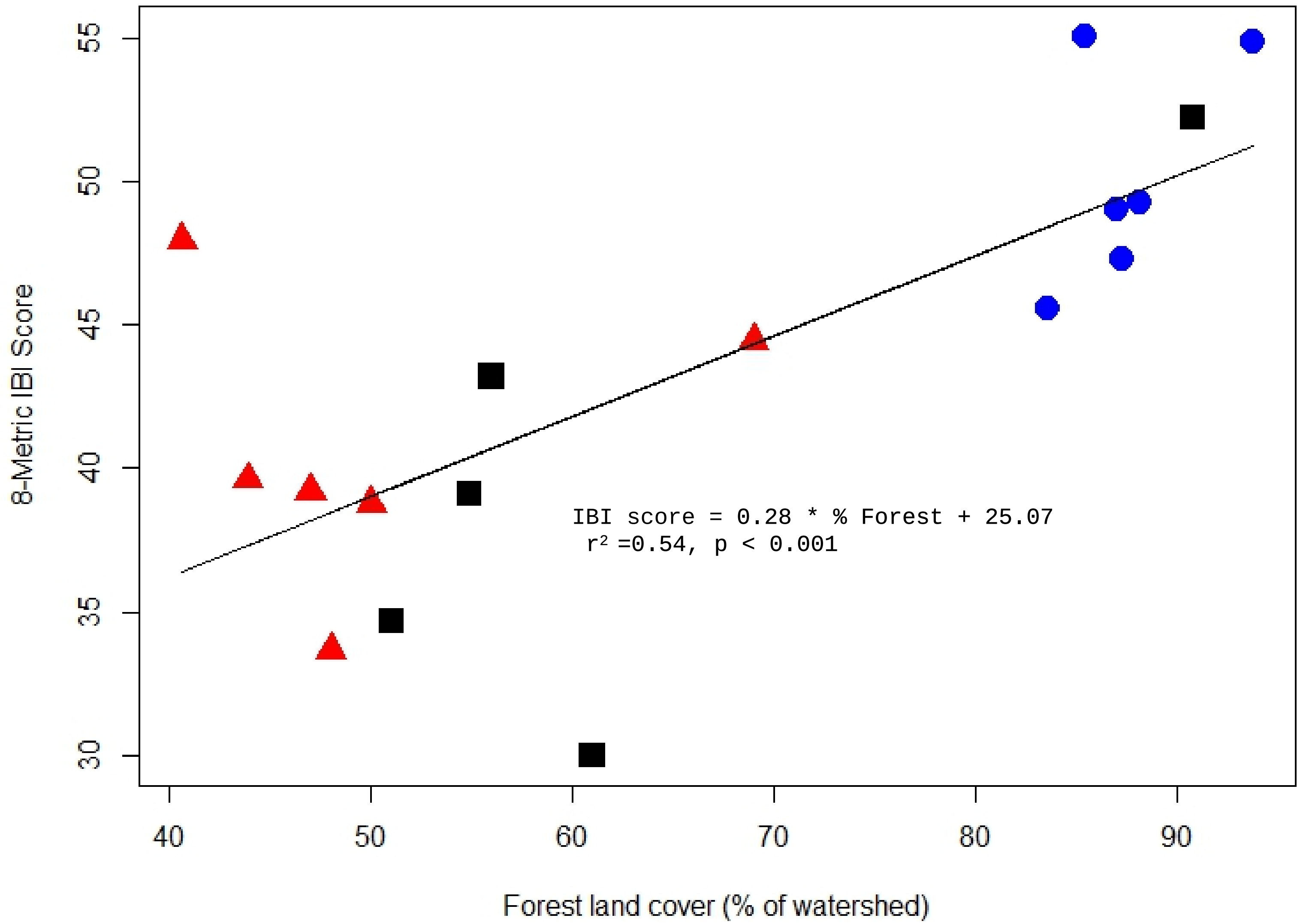

Figure 3: Correlation of 8-metric IBI with percent forest land cover. Red triangles = altered sites, black squares = intermediate sites, blue circles = reference sites.

Appendix A: Fish Data for the 17 Study Sites

Numbers represent number of individuals collected at each site. UT= unnamed tributary.

Common name	Genus and species	Beaverdam Creek	Canal Swamp	Cohoke Mill Creek	Dark Swamp	Dragon Run
American eel	*Anguilla rostrata*	1	1	5	9	6
American pickerel	*Esox americanus*	0	0	0	0	2
Banded killifish	*Fundulus diaphanus*	0	0	0	1	0
Banded sunfish	*Enneacanthus obesus*	9	25	12	6	10
Bluegill	*Lepomis macrochirus*	0	0	10	16	7
Bluespotted sunfish	*Enneacanthus gloriosus*	7	0	18	35	0
Bowfin	*Amia calva*	0	0	0	0	0
Brown bullhead	*Ameiurus nebulosus*	0	6	0	0	0
Eastern mudminnow	*Umbra pygmaea*	0	35	1	0	0
Eastern silvery minnow	*Hybognathus regius*	0	0	0	0	0
Flier	*Centrarchus macropterus*	2	0	0	0	0
Golden shiner	*Notemigonus crysoleucas*	0	5	0	0	24
Largemouth bass	*Micropterous salmoides*	0	0	0	2	2
Margined madtom	*Noturus insignis*	0	0	0	0	0
Mosquitofish	*Gambusia holbrooki*	0	3	0	0	0
Mud sunfish	*Acantharchus pomotis*	0	0	0	0	0
Pirate perch	*Aphredoderus sayanus*	12	0	7	0	0
Pumpkinseed	*Lepomis gibbosus*	1	0	13	8	2
Redbreast sunfish	*Lepomis auritus*	0	0	0	7	0
Redear sunfish	*Lepomis microlophus*	0	0	1	0	0
Redfin pickerel	*Esox niger*	1	0	1	0	0
Swamp darter	*Etheostoma fusiforme*	0	0	0	0	0
Tadpole madtom	*Noturus gyrinus*	0	0	1	0	0
Tessellated darter	*Etheostoma olmstedi*	0	0	0	5	0
Warmouth	*Lepomis gulosus*	0	0	0	0	2
Yellow bullhead	*Ameiurus natalis*	0	0	0	1	3

Appendix A continued.

Common name	Genus and species	France Swamp	Goldenvale Creek	Grays Creek	Lodge Creek	Mapisco Creek
American eel	*Anguilla rostrata*	23	6	50	117	12
American pickerel	*Esox americanus*	0	0	0	0	0
Banded killifish	*Fundulus diaphanus*	1	0	0	0	0
Banded sunfish	*Enneacanthus obesus*	10	0	2	7	0
Bluegill	*Lepomis macrochirus*	7	4	2	27	9
Bluespotted sunfish	*Enneacanthus gloriosus*	7	2	3	35	0
Bowfin	*Amia calva*	0	0	0	0	0
Brown bullhead	*Ameiurus nebulosus*	0	0	0	3	1
Eastern mudminnow	*Umbra pygmaea*	0	0	0	1	0
Eastern silvery minnow	*Hybognathus regius*	0	0	0	0	1
Flier	*Centrarchus macropterus*	0	0	0	0	2
Golden shiner	*Notemigonus crysoleucas*	1	0	1	1	0
Largemouth bass	*Micropterous salmoides*	1	0	0	0	2
Margined madtom	*Noturus insignis*	0	0	0	0	0
Mosquitofish	*Gambusia holbrooki*	6	0	0	1	1
Mud sunfish	*Acantharchus pomotis*	0	1	0	0	0
Pirate perch	*Aphredoderus sayanus*	8	0	2	0	1
Pumpkinseed	*Lepomis gibbosus*	31	0	3	0	5
Redbreast sunfish	*Lepomis auritus*	36	0	36	0	0
Redear sunfish	*Lepomis microlophus*	0	0	0	0	0
Redfin pickerel	*Esox niger*	0	0	1	1	1
Swamp darter	*Etheostoma fusiforme*	0	0	0	0	1
Tadpole madtom	*Noturus gyrinus*	0	0	0	1	1
Tessellated darter	*Etheostoma olmstedi*	15	0	6	8	0
Warmouth	*Lepomis gulosus*	0	0	1	0	0
Yellow bullhead	*Ameiurus natalis*	2	1	0	0	1

Appendix A continued.

Common name	Genus and species	Oldham Run	Rumley Marsh	Timber Branch Swamp	UT Diascund Creek	UT Piscataway Creek
American eel	*Anguilla rostrata*	36	0	1	3	2
American pickerel	*Esox americanus*	0	0	2	0	4
Banded killifish	*Fundulus diaphanus*	0	0	0	0	0
Banded sunfish	*Enneacanthus obesus*	17	8	12	8	1
Bluegill	*Lepomis macrochirus*	0	0	0	103	0
Bluespotted sunfish	*Enneacanthus gloriosus*	0	4	8	3	1
Bowfin	*Amia calva*	0	0	1	0	0
Brown bullhead	*Ameiurus nebulosus*	0	0	0	0	0
Eastern mudminnow	*Umbra pygmaea*	15	0	1	2	2
Eastern silvery minnow	*Hybognathus regius*	0	0	0	0	0
Flier	*Centrarchus macropterus*	0	0	0	0	0
Golden shiner	*Notemigonus crysoleucas*	6	0	2	0	1
Largemouth bass	*Micropterous salmoides*	0	0	0	8	0
Margined madtom	*Noturus insignis*	0	0	0	1	0
Mosquitofish	*Gambusia holbrooki*	0	0	0	1	0
Mud sunfish	*Acantharchus pomotis*	0	3	0	0	0
Pirate perch	*Aphredoderus sayanus*	13	11	4	21	1
Pumpkinseed	*Lepomis gibbosus*	0	0	2	0	0
Redbreast sunfish	*Lepomis auritus*	0	0	0	0	7
Redear sunfish	*Lepomis microlophus*	0	0	0	1	0
Redfin pickerel	*Esox niger*	0	4	1	2	0
Swamp darter	*Etheostoma fusiforme*	0	0	0	0	0
Tadpole madtom	*Noturus gyrinus*	10	0	0	0	7
Tessellated darter	*Etheostoma olmstedi*	1	2	0	9	3
Warmouth	*Lepomis gulosus*	0	0	0	0	0
Yellow bullhead	*Ameiurus natalis*	3	0	0	4	1

Appendix A continued.

Common name	Genus and species	White Marsh	White Oak Run
American eel	*Anguilla rostrata*	1	0
American pickerel	*Esox americanus*	2	0
Banded killifish	*Fundulus diaphanus*	0	0
Banded sunfish	*Enneacanthus obesus*	2	0
Bluegill	*Lepomis macrochirus*	7	0
Bluespotted sunfish	*Enneacanthus gloriosus*	0	0
Bowfin	*Amia calva*	0	0
Brown bullhead	*Ameiurus nebulosus*	0	0
Eastern mudminnow	*Umbra pygmaea*	35	1
Eastern silvery minnow	*Hybognathus regius*	0	0
Flier	*Centrarchus macropterus*	0	0
Golden shiner	*Notemigonus crysoleucas*	14	0
Largemouth bass	*Micropterous salmoides*	0	0
Margined madtom	*Noturus insignis*	0	0
Mosquitofish	*Gambusia holbrooki*	1	0
Mud sunfish	*Acantharchus pomotis*	0	0
Pirate perch	*Aphredoderus sayanus*	24	0
Pumpkinseed	*Lepomis gibbosus*	0	0
Redbreast sunfish	*Lepomis auritus*	0	0
Redear sunfish	*Lepomis microlophus*	0	0
Redfin pickerel	*Esox niger*	0	0
Swamp darter	*Etheostoma fusiforme*	0	0
Tadpole madtom	*Noturus gyrinus*	2	0
Tessellated darter	*Etheostoma olmstedi*	0	0
Warmouth	*Lepomis gulosus*	0	0
Yellow bullhead	*Ameiurus natalis*	0	0

Appendix B: Ecological Information

Numbers indicate whether species is correctly described by each ecological state. 1: yes, 0: no.

Common name	Genus and species	Omnivore	Vegetation spawner	Vegetation specialist	Nester	Invertivore	Native	Piscivore
American eel	*Anguilla rostrata*	1	0	0	0	1	1	0
American pickerel	*Esox americanus*	0	1	1	0	0	1	1
Banded killifish	*Fundulus diaphanus*	0	1	0	0	1	1	0
Banded sunfish	*Enneacanthus obesus*	0	0	0	0	1	1	0
Bluegill	*Lepomis macrochirus*	0	0	0	1	1	0	0
Bluespotted sunfish	*Enneacanthus gloriosus*	0	0	0	1	1	1	0
Bowfin	*Amia calva*	0	1	0	1	0	1	1
Brown bullhead	*Ameiurus nebulosus*	1	0	0	1	0	1	0
Eastern mudminnow	*Umbra pygmaea*	0	0	0	1	1	1	0
Eastern silvery minnow	*Hybognathus regius*	1	0	0	0	0	1	0
Flier	*Centrarchus macropterus*	0	0	0	1	1	1	0
Golden shiner	*Notemigonus crysoleucas*	0	0	0	0	0	1	0
Largemouth bass	*Micropterous salmoides*	0	0	0	1	0	0	1
Margined madtom	*Noturus insignis*	0	0	0	1	1	1	0
Mosquitofish	*Gambusia holbrooki*	0	0	0	0	1	1	0
Mud sunfish	*Acantharchus pomotis*	0	0	0	1	1	1	0
Pirate perch	*Aphredoderus sayanus*	0	0	0	0	1	1	0
Pumpkinseed	*Lepomis gibbosus*	0	0	0	1	1	1	0
Redbreast sunfish	*Lepomis auritus*	0	0	0	1	1	1	0
Redear sunfish	*Lepomis microlophus*	0	0	0	1	1	0	0
Redfin pickerel	*Esox niger*	0	1	1	0	0	1	1
Swamp darter	*Etheostoma fusiforme*	0	1	1	0	1	1	0
Tadpole madtom	*Noturus gyrinus*	0	0	0	1	1	1	0
Tessellated darter	*Etheostoma olmstedi*	0	0	0	1	1	1	0
Warmouth	*Lepomis gulosus*	0	0	1	0	1	1	1
Yellow bullhead	*Ameiurus natalis*	1	0	0	1	1	1	0

Appendix B continued.

Common name	Genus and species	Swamp specialist	Opportunistic colonizer	Facultative air breather	Structure-oriented	Pollution-tolerant	Pollution-sensitive
American eel	*Anguilla rostrata*	0	0	1	0	0	0
American pickerel	*Esox americanus*	1	0	0	0	0	0
Banded killifish	*Fundulus diaphanus*	0	1	0	0	0	0
Banded sunfish	*Enneacanthus obesus*	0	1	0	0	1	0
Bluegill	*Lepomis macrochirus*	0	1	0	0	1	0
Bluespotted sunfish	*Enneacanthus gloriosus*	0	1	0	0	0	0
Bowfin	*Amia calva*	1	0	1	0	0	0
Brown bullhead	*Ameiurus nebulosus*	0	1	0	0	1	0
Eastern mudminnow	*Umbra pygmaea*	0	1	1	1	1	0
Eastern silvery minnow	*Hybognathus regius*	0	1	0	0	0	0
Flier	*Centrarchus macropterus*	1	0	0	0	0	0
Golden shiner	*Notemigonus crysoleucas*	0	1	0	0	1	0
Largemouth bass	*Micropterous salmoides*	0	1	0	0	1	0
Margined madtom	*Noturus insignis*	0	0	0	1	0	0
Mosquitofish	*Gambusia holbrooki*	0	1	0	0	0	0
Mud sunfish	*Acantharchus pomotis*	0	0	0	0	0	0
Pirate perch	*Aphredoderus sayanus*	0	0	0	1	1	0
Pumpkinseed	*Lepomis gibbosus*	0	0	0	0	0	0
Redbreast sunfish	*Lepomis auritus*	0	0	0	0	0	0
Redear sunfish	*Lepomis microlophus*	0	1	0	0	1	0
Redfin pickerel	*Esox niger*	0	0	0	1	0	0
Swamp darter	*Etheostoma fusiforme*	1	0	0	0	0	1
Tadpole madtom	*Noturus gyrinus*	1	0	0	1	0	1
Tessellated darter	*Etheostoma olmstedi*	0	0	0	1	1	0
Warmouth	*Lepomis gulosus*	0	0	0	1	0	0
Yellow bullhead	*Ameiurus natalis*	0	0	0	1	0	0

Appendix C: Water Chemistry Quality Assurance/Quality Control Field Audit Form

PREPARATION FOR SAMPLE RUN		Y/N/NA	COMMENTS
1	Stainless Steel sample bucket clean with no rust or scratches?	N	Plastic bucket used to transport water for field radings. Acceptable.
2	Sample bottles free of dirt, stains, or other visible contaminants?	Y	
3	Any other sampling equipment used during the audit visibly clean and in good working order?	Y	
4	Temperature bottles and bacteria mesh bags present in the sample cooler(s)?	N	Samples frozen prior to delivery to lab.
5	Sample cooler(s) filled with sufficient ice for the sample run?	Y	
6	Sample tags and forms printed and contain the necessary information?	Y	
Issues that should be addressed: No issues.			
PROBE CALIBRATION		Y/N/NA	COMMENTS
1	Is the probe barometer within 10 mmHg of the laboratory barometer or National Weather Service readings?	Y	Cal checked at VCU lab during Feb 17 lab audit
2	Is the conductivity solution freshly prepared (within 1 month) and free of algae and other foreign materials?	Y	
3	Is the calibrated conductivity probe reading within 1.0% of the conductivity standard?	Y	Calibrations done weekly
5	DO probe calibrated to within 0.2 mg/L of theoretical levels? (0.1 mg/L for optical DO probes)	Y	Calibrations done weekly
6	Is the pH probe calibrated with 4, 7, 10 buffers or the two closest buffers which should bracket expected field values?	Y	Calibrations done weekly
7	Is the calibrated pH probe reading within 0.2 S.U. of each buffer value?	Y	Calibrations done weekly
8	Is pH 4 buffer used to protect the probe during transport to and from the field? If not, is non-lab grade water used which will not submerge the pH probe?	Y	tap water used.
Issues that should be addressed: No isses.			

BRIDGE, WADING,AND STREAMBANK SAMPLING		Y/N/NA	COMMENTS
1	Bridge sampling, do field staff wear reflective garments?	NA	Bridge sampling not done.
2	If sampling on a bridge, does it occur on the upstream side?	NA	Bridge sampling not done.
3	If wading, does field staff wade upstream to the sample site to avoid disturbing sediment at the site?	N	Wading downstream was necessary. Efforts taken to minimize sediment distrubance.
4	Field sonde deployed (DO, pH, etc.) before collecting samples?	N	Sample collected for testing back at truck.
5	If collecting samples with a bucket, is it rinsed with sample water prior to collecting the sample?	Y	
6	Samples collected at the Thalweg with no disturbance of bottom sediment?	Y	
7	Bacteria samples collected before other samples at the site?	NA	
8	Bacteria bottles filled according to SOP procedure? (No rinsing, filled to or above marked line, sterile technique.)	NA	
9	Sample bottles (except bacteria) rinsed with sample water prior to filling?	Y	TP sample was prefilled with preservative but TN bottle was rinsed.
10	Sample water well mixed prior to pouring into sample bottles?	NA	Directly collected in sample bottle
11	Is an air space left in sample bottles used for routine sampling (bacteria, nutrients, TSS, etc.)?	Y	
12	Preservatives (example: sulfuric acid) added to the samples which require preservation?	N	TP bottles prefilled with acid.
13	For chlorophyll a, is the sample filtered using gentle pressure (syringe) or at 12 mmHg (vacuum pump)?	NA	
14	If the sample water has a pH < 7.0, 1ml of magnesium carbonate added just prior to completing filtration?	NA	
15	Chlorophyll filters wrapped in foil following SOP procedures?	NA	
16	Sample tags completed and attached to the correct sample bottle/filters?	Y	Done back at truck. TP bottle was marked.

17	**Sample bottles placed into the cooler and covered up to the bottleneck with ice?**	**Y**	
18	**Chlorophyll filters placed on top of the ice in a waterproof bag with the bag opening hanging out of the closed lid?**	**NA**	
Issues that should be addressed: Discard all rinses downstream or away from sampling point to avoid sampling rinses. Adjust hand placment so it is downstream of flow to minimize contamination potental. While 15 minuites is allowed to bring a sample in bucket back to the truck for field readings, collect field readings in-situ wherever possible such as from nearest bridge or access point. Attach probe guard to sonde when deploying to minimize risk of potental damage to sensors.			
QUALITY ASSURANCE		Y/N/NA	COMMENTS
1	**Sonde kept in the passenger compartment of the vehicle when traveling to and from the field?**	**Y**	
2	**Mid day DO probe check reads within 95 to 105% saturation? Check done with wet towel or wetted storage cup.**	**NA**	**Only one sample site tested**
3	**Did field staff collect Equipment Blank (EB) samples (bacteria samples are exempt)?**	**NA**	**EB samples not part of this study.**
4	**Prior to collecting EB samples, was sample equipment and bottles rinsed with lab grade water simulating field rinsing?**	**NA**	
5	**EB bottles filled with fresh lab grade water after it has gone through the appropriate field equipment?**	**NA**	
6	**Were EB bottles properly labeled with sample tags using a sample depth of 0.0 and EB for sample type?**	**NA**	
7	**EB samples handled in the same manner as routine samples?**	**NA**	
8	**Did field staff collect split samples?**	**Y**	
9	**Were split samples collected at the same site(s) as the EB samples and after EB collection?**	**NA**	
10	**Split samples collected after field rinsing the equipment and bottles used sample water?**	**Y**	
11	**Bacteria split samples collected by suspending or immersing the bottles in the water simultaneously?**	**NA**	

12	**Split sample containers filled promptly to prevent settling?**	**Y**	
12a	**If settling occurs, is sample water mixed using a stainless steel spoon or by swirling the contents of the container?**	**NA**	
13	**Split sample tags recorded properly using S1, S2, etc.**	**Y**	
14	**Sample tags for split samples placed on the correct bottles?**	**Y**	
15	**Split samples handled in the same manner as routine samples?**	**Y**	
Issues that should be addressed: No issues.			
	PROBE END OF DAY CHECK	**Y/N/NA**	**COMMENTS**
1	**Sonde is allowed to adjust to room temperature before performing the end of day calibration checks?**	**NA**	**EOD check was not observed but given sample collected at one station, EOD check not critical.**
2	**Is conductivity readings within 5% for conductivity standards <1000 uS/cm (10% for standards >1000 uS/cm)?**	**NA**	
3	**Is the DO probe reading within 0.5 mg/L of theoretical levels?**	**NA**	
4	**Is the pH probe reading within 0.2 S.U. of each buffer value (7.00 and 4.00 and/or 10.00)?**	**NA**	
Issues that should be addressed: No issues.			
	SHIPPING SAMPLES TO THE LABORATORY	**Y/N/NA**	**COMMENTS**
1	**Field parameter data/ QC sample information entered correctly into CEDS?**	**NA**	
2	**Samples packed for shipment to laboratory according to SOP?**	**Y**	**Not observed but discussed. Samples are frozen prior to delviery to DCLS.**
3	**Sampling equipment cleaned according to the SOP?**	**Y**	**Not observed but discussed. Equipment appears to be clean and in good working order.**
Issues that should be addressed: No issues.			

Appendix D: Memorandum from Mr. James Beckley Concerning the Chemistry Quality Assurance / Quality Control Field Audit

MEMORANDUM

Virginia Department of Environmental Quality

Water Quality Monitoring and Assessment

629 East Main Street	**11th Floor**
Post Office Box 1105	**(804) 698-4025 phone**
Richmond, Virginia 23218	**(804) 698-4032 fax**

SUBJECT: VCU Class VII Aquatic Life Use Survey field audit

TO: David A. Hopler Jr.

COPY: Dr. Andrew Garey

FROM: James Beckley

DATE: 5/2/2017

On May 1, I conducted a field audit of water chemistry and field parameter sampling as part of Virginia Commonwealth University (VCU) contracted study to develop an aquatic life use assessment protocols for Virginia Class VII swamp waters. The audit occurred at Horse Swamp Creek in Henrico County. Below are my findings.

1. Water samples were discarded upstream of the sampling point allowing the possibility of rinsed water to reenter the sample bottle. Recommend discarding water downstream or well away from the sampling location to avoid potential contamination.

2. Hand placement allowed water to wash over the hand prior to entering the bottle resulting in potential contamination. Avoid allowing water from washing over ungloved hands before entering the sample bottle. Use of a small sampling wand can eliminate this issue entirely.

3. Site water was collected in a bucket to test field parameters back at the vehicle due to the bulky multiprobe used in the study. Samples generally were collected and tested within the 15 minute holding time allowed in applicable EPA and Standard Methods. However, there is considerable risk of changing readings using this sampling protocol. Recommend collecting field probe readings in-situ using the instrument. The study does allow sampling from the nearest bridge or access point to avoid needing to carry the sonde to the actual sampling location.

4. Observed the sonde was placed in the sample bucket without the probe guard to minimize the volume of water needed in the bucket. Sonde guard should be installed prior to collecting readings to avoid damaging the sensors due to resting on the bottom of the bucket or stream.

Overall the issues describe above are relatively minor given the total nitrogen and total phosphorus parameters are less sensitive to rinse and hand placement contamination and methods allow 15 minute holding time for field readings. That said, altering sampling protocol to minimize contamination sources is highly recommended to ensure good data quality. Regarding the practice of collecting a sample by bucket for field readings and not using a probe guard may result in altered readings. Deploying the sonde in-situ at the nearest available location with attached probe guard will ensure good data quality.

The enclosed field audit sheets contain additional details of the audit findings.