

MBSS ROUND 4 SAMPLE DESIGN: ISSUES AND RECOMMENDATIONS

Prepared for

State of Maryland
Department of Natural Resources
580 Taylor Avenue
Tawes State Office Building
Annapolis, MD 21401

Prepared by

Mark Southerland, Lisa Methratta, and Ginny Rogers
Versar, Inc.
Ecological Sciences & Applications
9200 Rumsey Road, Suite 100
Columbia, MD 21045

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1 INTRODUCTION

The Maryland Department of Natural Resources (DNR) is preparing to conduct Round 4 of the Maryland Biological Stream Survey (MBSS). The MBSS is a stratified-random, probabilistic survey that provides essential information on the ecological status of Maryland streams for the state's natural resource decision makers.

Round 4 of MBSS is scheduled to begin in 2014. Maryland DNR's primary goal for Round 4 of the MBSS is to document changes in stream conditions over time. Round 4 will also provide status information at the basin and statewide scales. Owing to budget limitations, obtaining stream condition information at finer scales (e.g., Maryland 8-digit watersheds combined as Primary Sampling Units or PSUs) is not feasible. The level of effort available for this effort is approximately 100 sites per year, so a target of 340 sites sampled over 3 to 5 years was used for planning purposes.

2 DESIGN ISSUES

Given the sampling effort available and the primary goal of detecting changes in stream conditions over time (trends), the following issues were evaluated:

- Whether to use all or some repeat sites from previous rounds
- Which rounds to select repeat sites from (stream maps and designs vary among rounds)
- Which stream map to use
- How to select sites for good geographic coverage
- What is the ability to detect change in IBIs and is it acceptable

2.1 REPEAT SITE SAMPLING

Sampling the same sites previously sampled in earlier rounds will reduce the variation among sites (compared to new random sites) and will provide the greatest ability to detect trends. The downside of sampling only repeat sites is that any change in streams that differs from this population of sites will not be captured. The choice of Option 2 below depends on accepting the assumption that overall change will not differ significantly from the change in the population of resampled sites.

Option 1: Sample a combination of fixed (repeat) and random sites (partial replacement sampling) with an allocation of 50% repeat and 50% new sites

Option 2: Sample only (100%) repeat sites

2.2 INTER-ROUND SAMPLING

Repeat sampling allows a comparison of change over the time period that each site is re-sampled. Since no sites in Rounds 1, 2, and 3 were re-sampled, R4 will be the first time that repeat sampling will occur. Sampling sites from R1 in R4 will give the longest time period over which change could have occurred. The choice of option below depends on the importance of evaluating change for small streams and optimal geographic extent (see issues below).

Option 1: Sample repeat sites from R2 which uses the current random design and greatest coverage of stream types and geography

Option 2: Sample all repeat sites from R1 (thereby excluding small streams and full geographic coverage as discussed below)

2.3 STREAM MAP

The stream maps from which sample sites were selected differed among rounds, so re-sampling is limited to the stream map previously used. R1 used the 1:250,000-scale map, while R2 and R3 used the 1:100,000-scale map. Therefore, re-sampling sites from R1 in R4 will exclude the small streams not on the 1:250,000-scale map, so that they will not be characterized in the trends analysis. Appendix A illustrates the differences in IBIs obtained when the small streams not found on the 1:250,000-scale map are removed from the R2 data. While the differences are small at the statewide and ecoregion scale, larger differences are apparent in some basins. Nonetheless, smaller streams consistently score lower than larger streams, so R1 ratings are artificially high when trying to characterize all Maryland streams on the 1:100,000-scale map.

Option 1: Re-sample from R2 and be able to assess trends including small streams not on 1:250,000-scale map, but the time period of comparison will be limited to 13 years

Option 2: Re-sample from R1 and only assess trends for larger streams on 1:250,000-scale map, but be able to compare over the longer 18 years

2.4 GEOGRAPHIC COVERAGE

The evenness of the geographic coverage differed among rounds according to their sampling designs, so re-sampling is limited to the locations of sites previously sampled. The design of R1 was stratified on basin and stream order, so not all PSUs were sampled. Re-sampling R1 will mean that not all PSUs will have R4 sites. R2 has the most complete geographic coverage with 10 sites in each PSU.

Option 1: Re-sample from R2 for the best geographic coverage and retain the current sampling design, but limit the time period of comparison to 13 years

Option 2: Re-sample from R1 and exclude some PSUs, but be able to compare over the longer 18 years

2.5 ABILITY TO DETECT TRENDS

Appendix B provides the power analysis used to explore different sampling scenarios with varying levels of error, statistical power, and sample size. Random sites from R1 of MBSS were used in the analysis at the statewide, ecoregion, basin, and county levels of spatial resolution. Specifically, we determined the change in the BIBI score that could be detected given 1,000 total survey sites (equivalent to R1 and R2 sampling intensity) and 340 total survey sites (about 20 per basin).

Option 1: Re-Sample at an intensity comparable to R1 and R2 or 1,000 sites in R4, which would provide the ability to detect a change in BIBI (at 80% probability) of 0.10 statewide and 0.26-0.40 for basins

Option 2: Re-sample at the feasible intensity of 340 sites in R4, which would provide the ability to detect a change in BIBI (at 80% probability) of 0.19 statewide and 0.50-0.80 for basins

3 RECOMMENDATIONS

The MBSS has now completed three statewide surveys that provide useful baselines for assessing trends compared to the Round 4 sampling that will begin in 2014. Summary estimates of stream condition at the statewide and basin scales will be possible between each of these rounds and statewide R4 results. To maximize the utility of R4 for trends detection, we recommend re-sampling sites from previous rounds as the basis for the R4 design. The following consolidates solutions to each of the design issues into two options, although other permutations are possible:

Option 1: Re-sample 170 (50%) of sites in R2 allocated 2 to each PSU and sample 170 new random sites allocated 2 to each PSU (total of 4 sites in each PSU). These sites would include all streams on the 1:100,000-scale map and use the design stratified on PSU. The design would be able to detect a change of about 0.2 BIBI scores, or 20% of a condition class at the statewide level.

Option 2: Re-sample 340 of sites in R1 with no new random sites, allocated 20 to each basin and evenly spread among the stream orders (1, 2, and 3) on the 1:250,000-scale map. Some PSUs would not be sampled and the smaller streams occurring only on the 1:100,000-scale map would not be sampled. The design would be able to detect a change of about 0.2 BIBI scores, or 20% of a condition class at the statewide level. The trends analysis would only apply to streams on the 1:250,000-scale map.

APPENDIX A DIFFERENCE IN IBI SCORES RESULTING FROM DIFFERENT MAP SCALES

A1. INTRODUCTION

If the MBSS Round 4 design includes re-sampling sites from Round 1, only sites on the 1:250,000 sampling frame will be available and smaller streams on the 1:100,000-scale maps used for Rounds 2 and 3 will not be sampled. This analysis investigates the hypothesis of whether these smaller streams have lower indices of biotic integrity (and perhaps suffer greater degradation). A significant difference in stream condition estimates indicates that trends using this design cannot be extrapolated to the smaller, excluded streams.

The smallest streams from Round 1 scored lower BIBI and FIBI values (Figure A1). This pattern has persisted throughout later rounds of MBSS which were able to examine the biological status of more small streams (Figures A2 and A3). The concern is that when scores are aggregated to estimate statewide, ecoregion, and basin level averages, i.e., we may determine that an entire basin ranks as fair when, in fact if we had included more small streams in the survey, we may have determined the rank to be poor (Table A1).

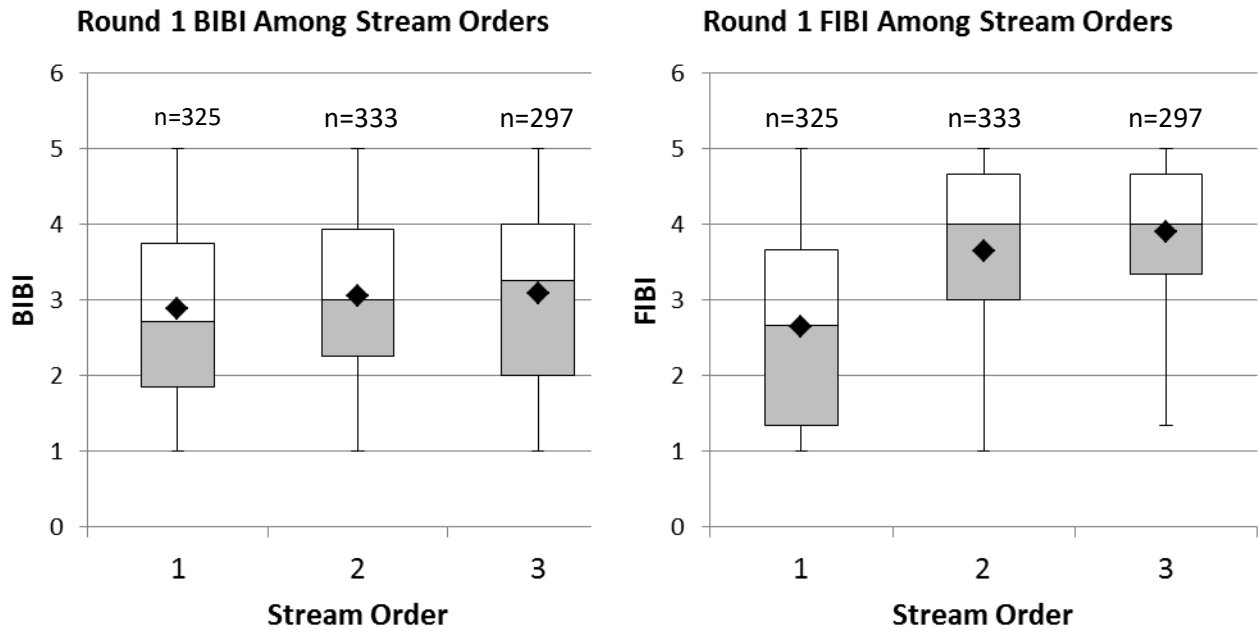


Figure A1. Statewide biological indicator scores from Round 1 of MBSS for which the sites were selected using a 1:100,000 sampling frame. Left Panel: Statewide BIBI by stream order for Round 1. Right Panel: Statewide FIBI by stream order for Round 1. Smaller streams (i.e., smaller order streams) are in somewhat worse condition.

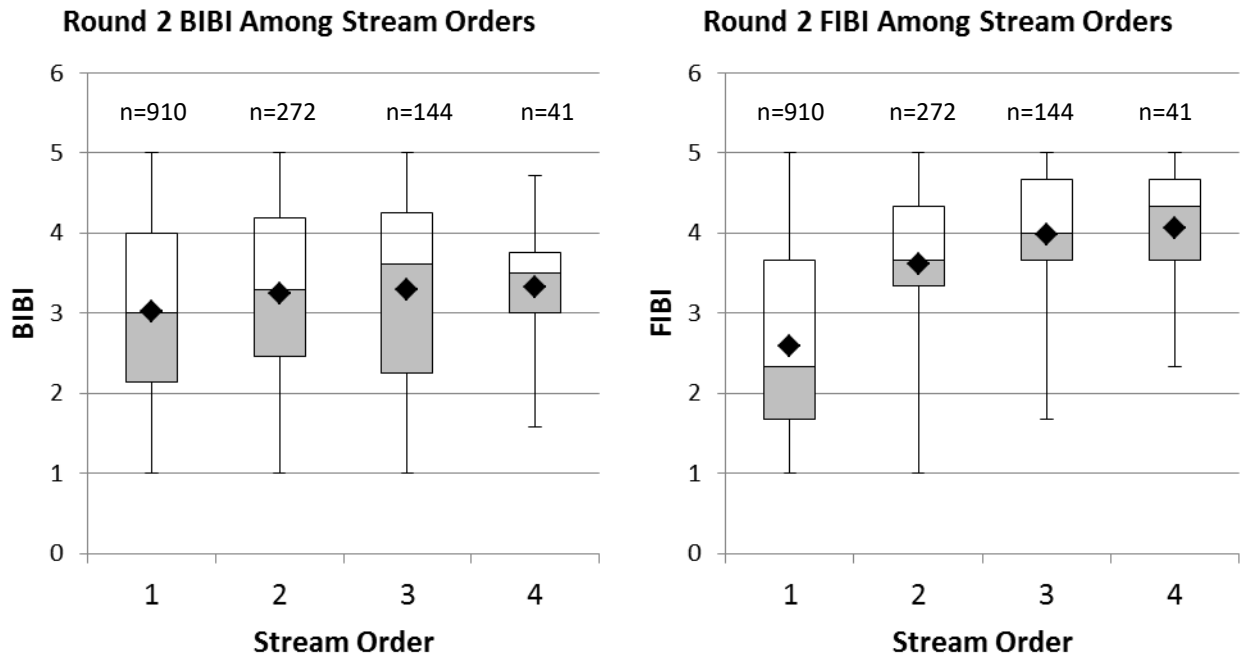


Figure A2. Statewide biological indicator scores from Round 2 of MBSS for which the sites were selected using a 1:250,000 sampling frame. Left Panel: Statewide BIBI by stream order for Round 1. Right Panel: Statewide FIBI by stream order for Round 1. Smaller streams (i.e., smaller order streams) are in somewhat worse condition.

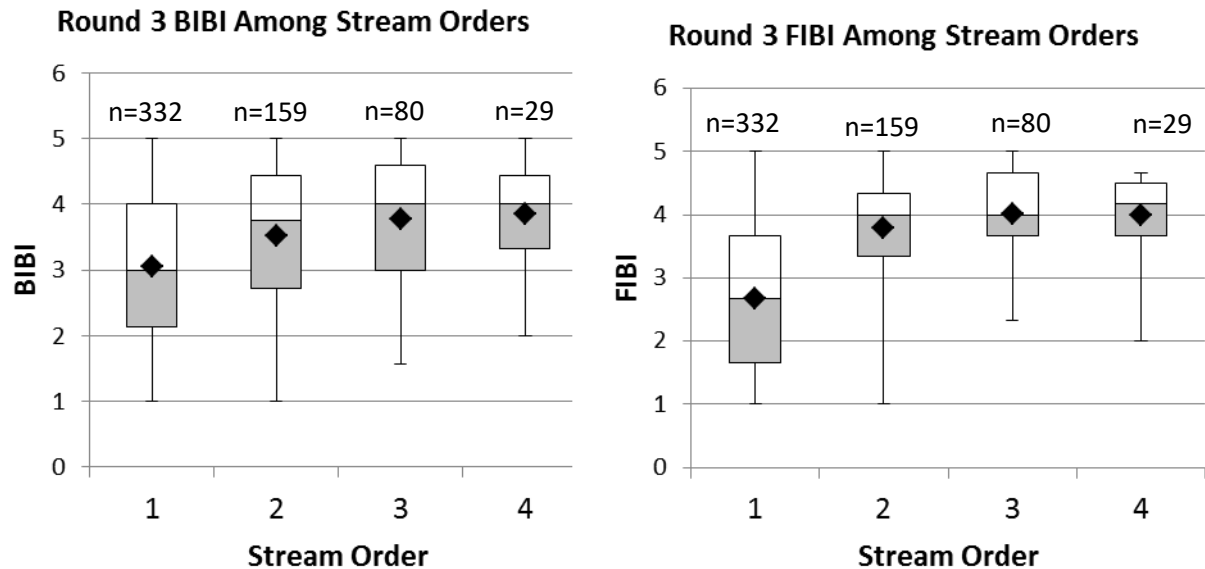


Figure A3. Statewide biological indicator scores from Round 3 of MBSS for which the sites were selected using a 1:250,000 sampling frame. Left Panel: Statewide BIBI by stream order for Round 1. Right Panel: Statewide FIBI by stream order for Round 1. Smaller streams (i.e., smaller order streams) are in somewhat worse condition.

Table A1. Ratings associated with BIBI and FIBI scores from the MBSS			
Very Poor	Poor	Fair	Good
1 to 2	>2 to 3	>3 to 4	>4 to 5

A2. METHODS

In this report, we address the concerns related to resampling Round 1 streams for Round 4 of the MBSS that (1) by resampling only streams from Round 1, we are not sufficiently sampling small streams, and (2) if we are under-representing the small streams in our assessment, then we may be biasing our interpretation of the biological status of Maryland streams. To address these concerns, we compared the BIBI and FIBI scores from all Round 2 sites to those Round 2 sites that could have been selected using the 1:250,000-scale map. We make these comparisons at the statewide, ecoregion, and basin scales. We considered

- The percentage of sites from the full set of Round 2 sites that could be selected using the 1:250,000-scale map
- The magnitude and direction of difference between mean index scores for biological integrity calculated using all of Round 2 sites and just the subset that could have been selected using the 1:250,000-scale map
- Whether the changes in biological index scores led to a different rating

A3. RESULTS

A3.1 STATEWIDE

From a statewide perspective, 84% of the Round 2 sites used to calculate the BIBI could have been selected using the 1:250,000-scale map and 86% of the Round 2 sites used to calculate the FIBI could have been selected using the 1:250,000-scale map (Table A2). The subset of Round 2 sites that were on the 1:250,000-scale map had slightly a greater mean BIBI score and mean FIBI score than when calculated using all Round 2 sites (Figure A4, Table A3). The statewide rating for FIBI changed from Fair to Good using sites on the 1:250,000-scale map.

Table A2. The number and percentage of sites from MBSS Round 2 at the statewide level that could have been selected using the 1:250,000-scale map. The total number of sites for BIBI and FIBI are different because fish are not sampled at all MBSS sites.

State	BIBI			FIBI		
	All R2 Sites	R2 Sites on the 1:250,000-Map	% of R2 Sites on 1:250,000-Map	All R2 Sites	R2 Sites on the 1:250,000-Map	% of R2 Sites on 1:250,000-Map
Maryland	1067	893	84%	966	826	86%

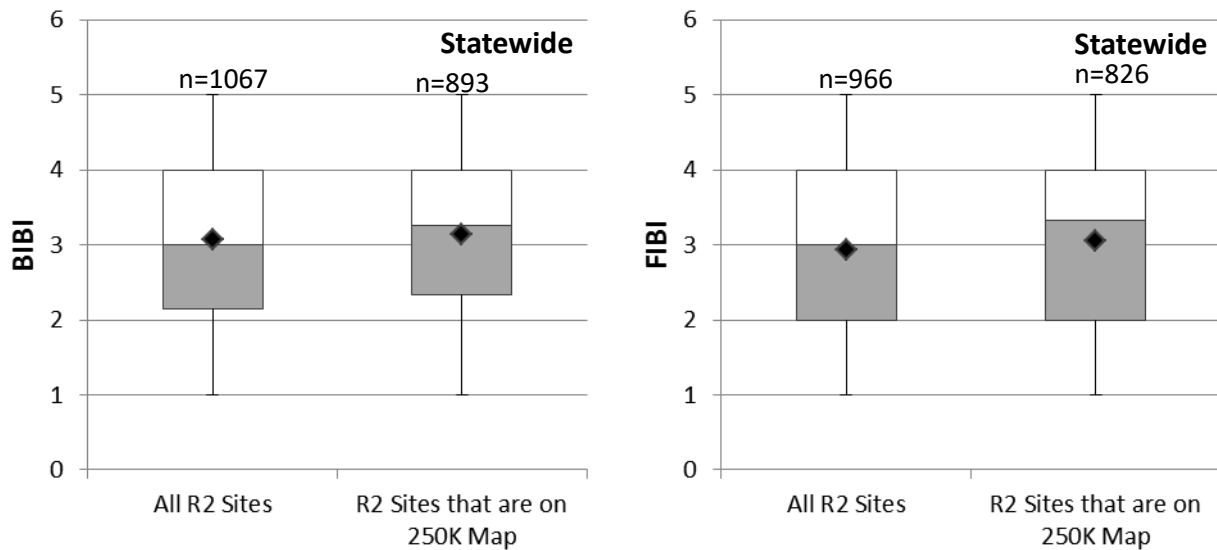


Figure A4. Comparison of statewide biological indices for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

Table A3. The difference in mean index scores (BIBI and FIBI) at the statewide level when all Round 2 sites are used compared to using only those Round 2 sites that could have been selected using the 1:250,000-scale map. The number of MBSS Round 2 sites for BIBI and FIBI are different because fish are not sampled at all MBSS sites.

State	BIBI				FIBI			
	All R2 Sites	R2 Sites on the 1:250,000-Map	Difference	BIBI Rating Changed	All R2 Sites	R2 Sites on the 1:250,000-Map	Difference	FIBI Rating Changed
Maryland	3.07	3.14	0.07	No	2.93	3.05	0.12	Yes

A3.2 ECOREGIONS

For the BIBI at the ecoregion scale, Highlands would retain the most Round 2 sites (94%) if Round 2 sites were selected using the 1:250,000-scale map, while East Piedmont and Coastal Plain would retain 88% and 74% respectively (Table A4). Both BIBI and FIBI were somewhat higher when calculated using the subset of Round 2 sites that could be selected using the 1:250,000-compared to when they were calculated using all Round 2 sites. This pattern was evident for the Highlands, East Piedmont, and Coastal Plain ecoregions (Figures A5, A6, A7, Table A5). Using only the 1:250,000-sites from Round 2 changed the FIBI rating for Coastal Plain from Poor to Fair.

Table A4. The number and percentage of sites from MBSS Round 2 at the ecoregion level that could have been selected using the 1:250,000-scale map. The total number of sites for BIBI and FIBI are different because fish are not sampled at all MBSS sites.

Ecoregion	BIBI			FIBI		
	All R2 Sites	R2 Sites on the 1:250,000-Map	% of R2 Sites on 1:250,000-Map	All R2 Sites	R2 Sites on the 1:250,000-Map	% of R2 Sites on 1:250,000-Map
Highland	315	296	94%	288	273	95%
East Piedmont	275	243	88%	271	241	89%
Coastal Plain	476	353	74%	406	311	77%

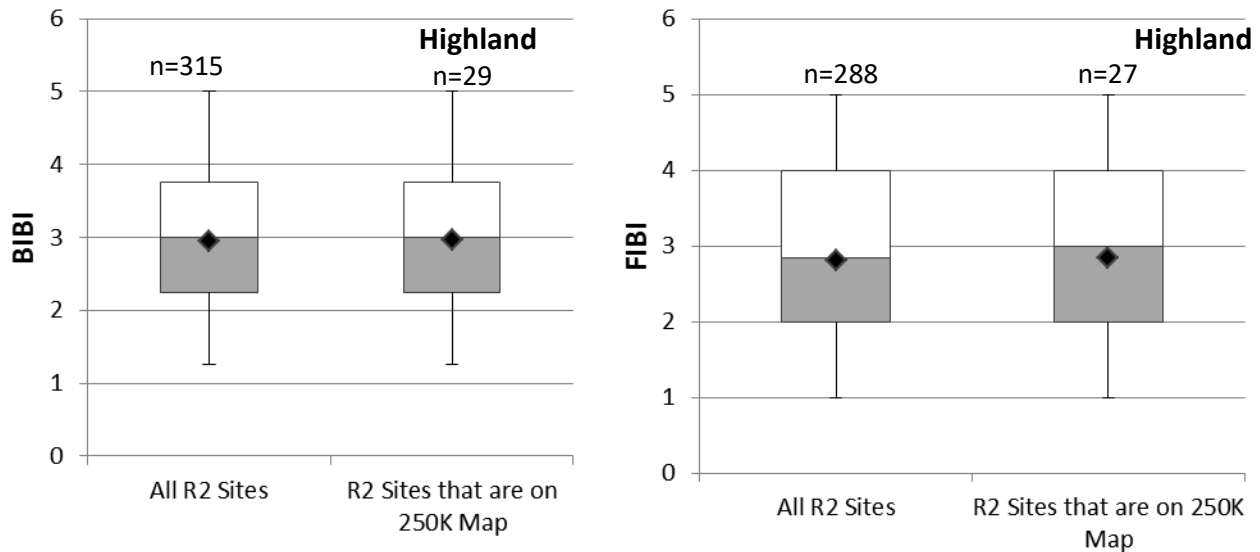


Figure A5. Comparison of biological indices in the Highlands ecoregion for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

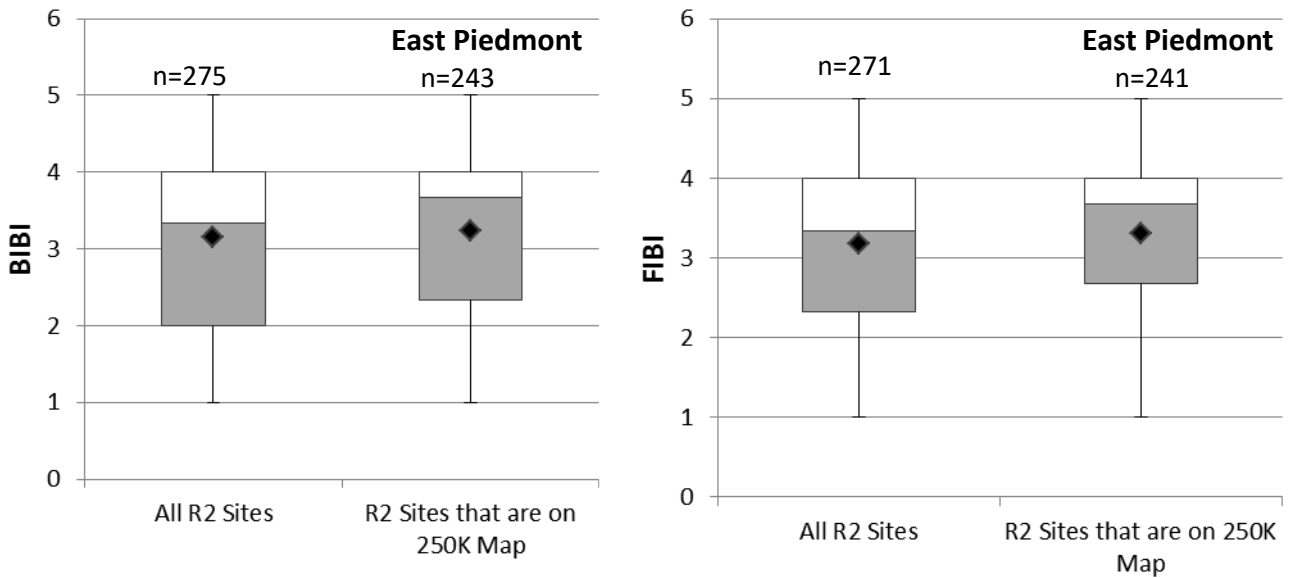


Figure A6. Comparison of biological indices in the East Piedmont ecoregion for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

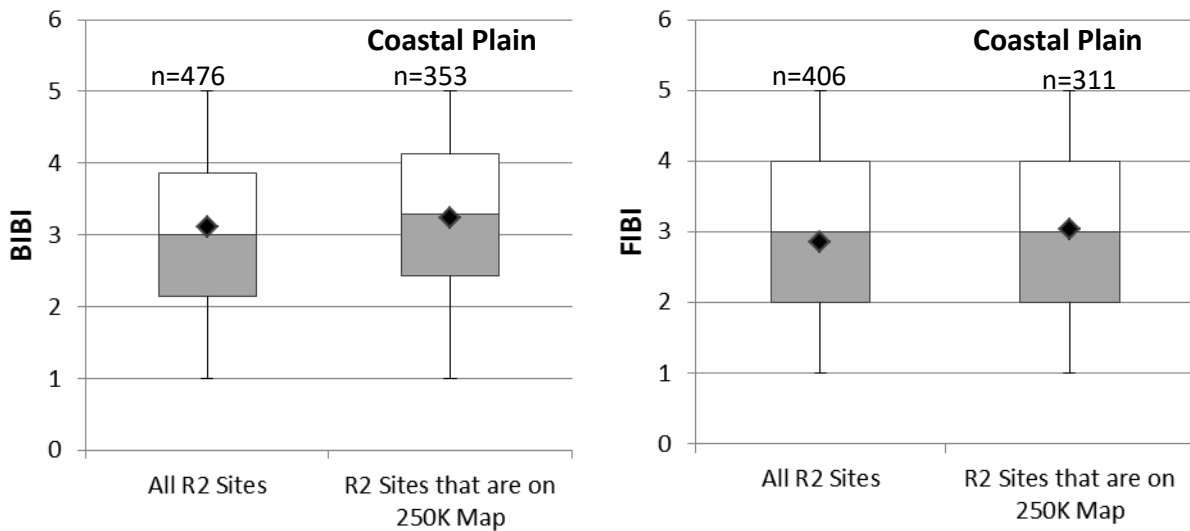


Figure A7. Comparison of biological indices in the Coastal Plain ecoregion for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

Table A5. The difference in mean index scores (BIBI and FIBI) at the ecoregion level when all Round 2 sites are used compared to using only those Round 2 sites that could have been selected using the 1:250,000-scale map. The number of MBSS Round 2 sites for BIBI and FIBI are different because fish are not sampled at all MBSS sites.								
Ecoregion	BIBI				FIBI			
	All R2 Sites	R2 Sites on the 1:250,000-Map	Difference	BIBI Rating Changed	All R2 Sites	R2 Sites on the 1:250,000-Map	Difference	FIBI Rating Changed
Highlands	2.95	2.96	0.01		2.81	2.85	0.03	
East Piedmont	3.16	3.24	0.08		3.17	3.30	0.13	
Coastal Plain	3.11	3.23	0.12		2.86	3.03	0.18	*

A3.3 BASINS

The percentage of sites from the total number sampled during Round 2 that could have been selected using the 1:250,000-map ranged from 44% to 97% for BIBI and from 33% to 97% for FIBI (Table A6). Both BIBI and FIBI scores were somewhat higher when calculated using only the Round 2 sites that could have been selected using the 1:250,000-scale map (Table A6). There was 1 basin for BIBI (Pocomoke) and seven basins for FIBI (Choptank, Lower Potomac, Nanticoke, Ocean Coastal, Patapsco, Washington Metro Potomac, and Patuxent) that were rated as Poor using all of the Round 2 sites but Fair when using only the Round 2 sites on the 1:250,000-scale map (Table A7, Figures A8-A25).

Table A6. The number and percentage of sites from MBSS Round 2 at the basin level that could have been selected using the 1:250,000-scale map. The total number of sites for BIBI and FIBI are different because fish are not sampled at all MBSS sites.						
Basin	BIBI			FIBI		
	All R2 Sites	R2 Sites on the 1:250,000-Map	% of R2 Sites on 1:250,000-Map	All R2 Sites	R2 Sites on the 1:250,000-Map	% of R2 Sites on 1:250,000-Map
BU	30	27	90%	26	23	88%
CK	34	16	47%	29	12	41%
CR	60	51	85%	55	46	84%
EL	29	23	79%	28	23	82%
GU	57	48	84%	54	47	87%
LP	116	92	79%	104	84	81%
MP	82	77	94%	75	70	93%
NO	69	67	97%	68	66	97%
NW	49	29	59%	34	23	68%
OC	9	4	44%	6	2	33%
PC	43	27	63%	25	19	76%
PP	107	94	88%	104	92	88%

Basin	BIBI			FIBI		
	All R2 Sites	R2 Sites on the 1:250,000-Map	% of R2 Sites on 1:250,000-Map	All R2 Sites	R2 Sites on the 1:250,000-Map	% of R2 Sites on 1:250,000-Map
PW	72	63	88%	70	62	89%
PX	95	85	89%	93	83	89%
SQ	48	42	88%	48	42	88%
UP	101	95	94%	84	81	96%
WC	30	22	73%	27	20	74%
YG	36	31	86%	36	31	86%

Basin	BIBI				FIBI				
	All R2 Sites	R2 Sites on the 1:250,000-Map	Difference	BIBI Rating Changed	Basin	All R2 Sites	R2 Sites on the 1:250,000-Map	Difference	FIBI Rating Changed
BU	2.10	2.15	0.05		BU	3.22	3.19	-0.03	
CK	3.24	3.77	0.52		CK	2.86	3.42	0.56	*
CR	3.33	3.41	0.08		CR	3.44	3.57	0.12	
EL	3.28	3.44	0.15		EL	3.59	3.81	0.22	
GU	3.39	3.61	0.22		GU	3.03	3.28	0.25	
LP	3.66	3.73	0.07		LP	2.96	3.19	0.23	*
MP	2.60	2.61	0.01		MP	2.94	3.00	0.05	
NO	3.38	3.35	-0.03		NO	2.68	2.70	0.02	
NW	2.77	2.88	0.11		NW	2.75	3.15	0.40	*
OC	2.65	2.71	0.06		OC	2.84	3.17	0.33	*
PC	2.89	3.18	0.29	*	PC	3.23	3.42	0.19	
PP	2.71	2.78	0.07		PP	2.93	3.05	0.11	*
PW	2.45	2.45	-0.002		PW	2.93	3.05	0.12	*
PX	3.22	3.28	0.07		PX	2.97	3.01	0.04	*
SQ	3.79	3.79	-0.01		SQ	3.15	3.28	0.13	
UP	2.98	2.99	0.01		UP	2.68	2.67	-0.01	
WC	3.22	3.45	0.24		WC	1.85	1.87	0.01	
YG	3.10	3.19	0.09		YG	2.65	2.79	0.14	

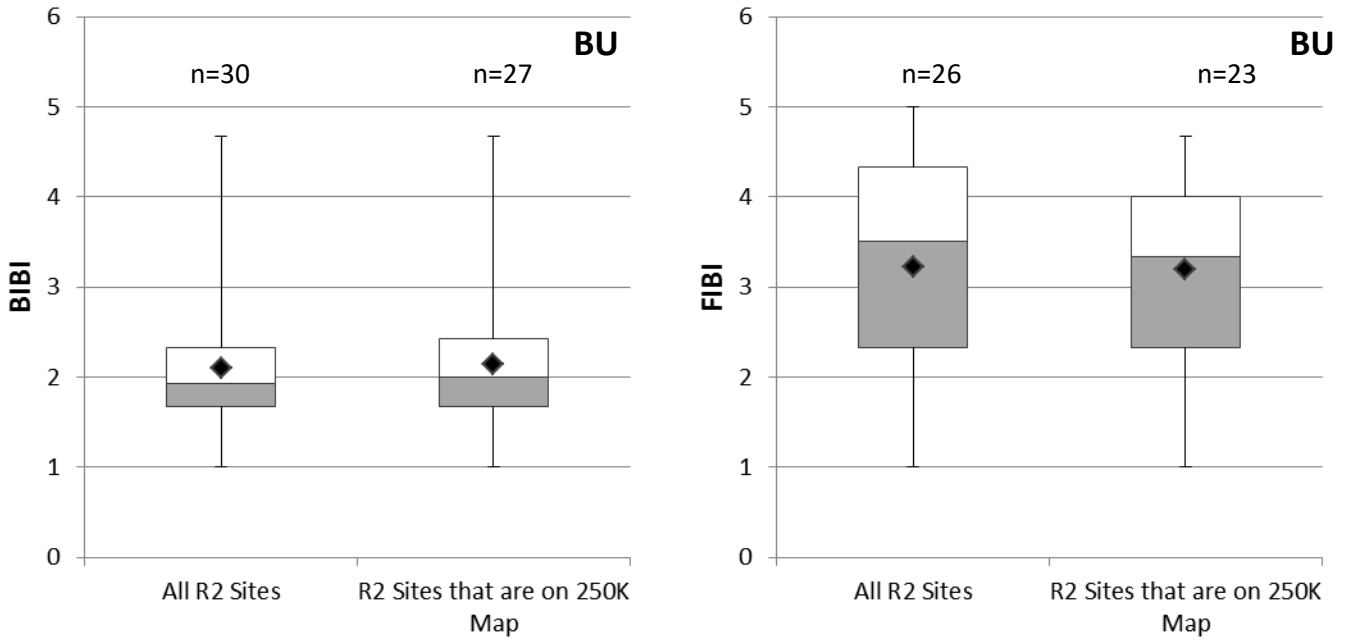


Figure A8. Comparison of biological indices in the Bush River basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

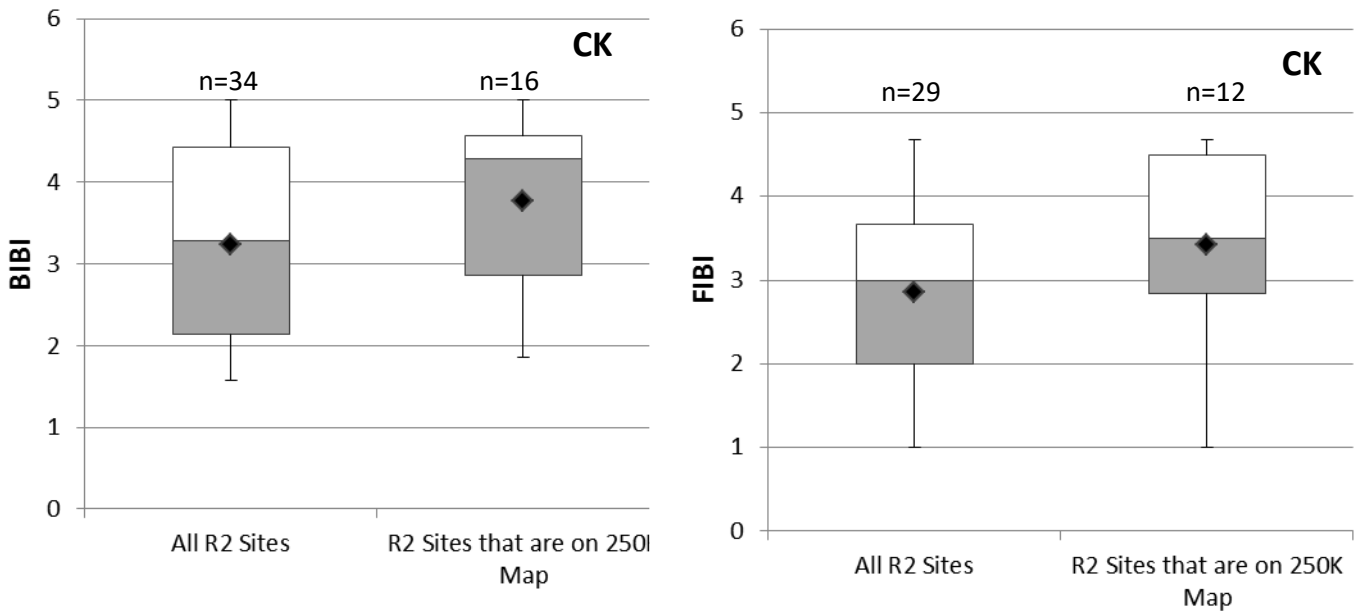


Figure A9. Comparison of biological indices in the Choptank River basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

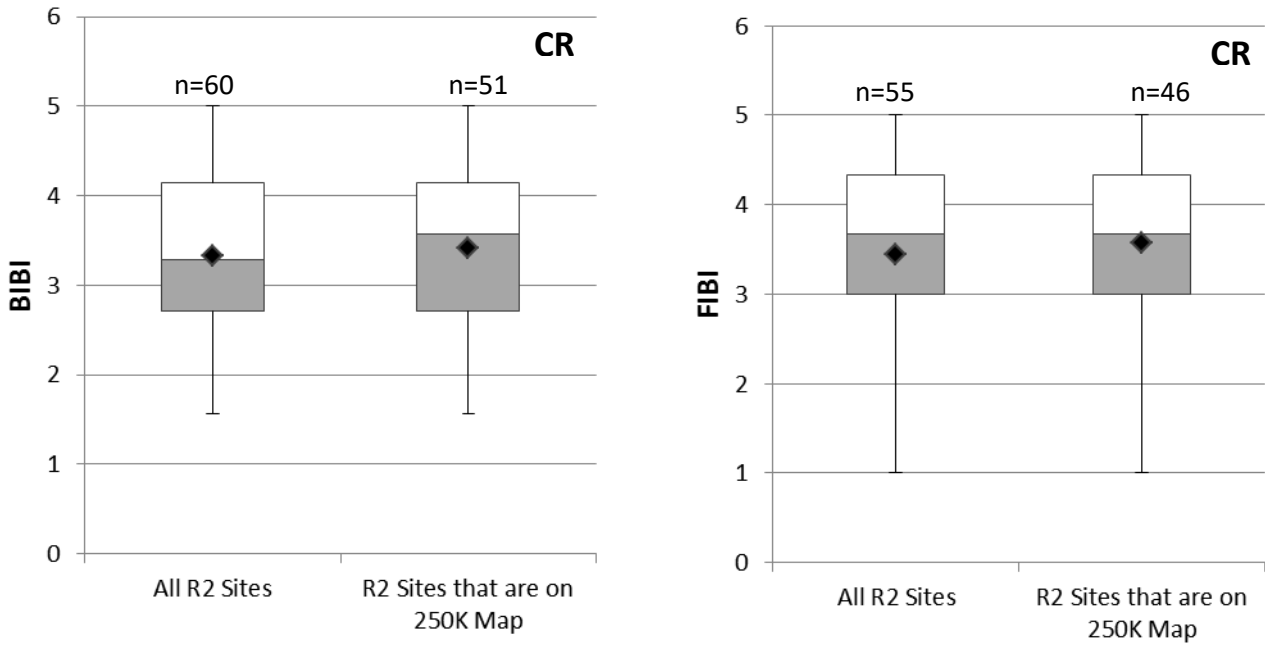


Figure A10. Comparison of biological indices in the Chester River basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

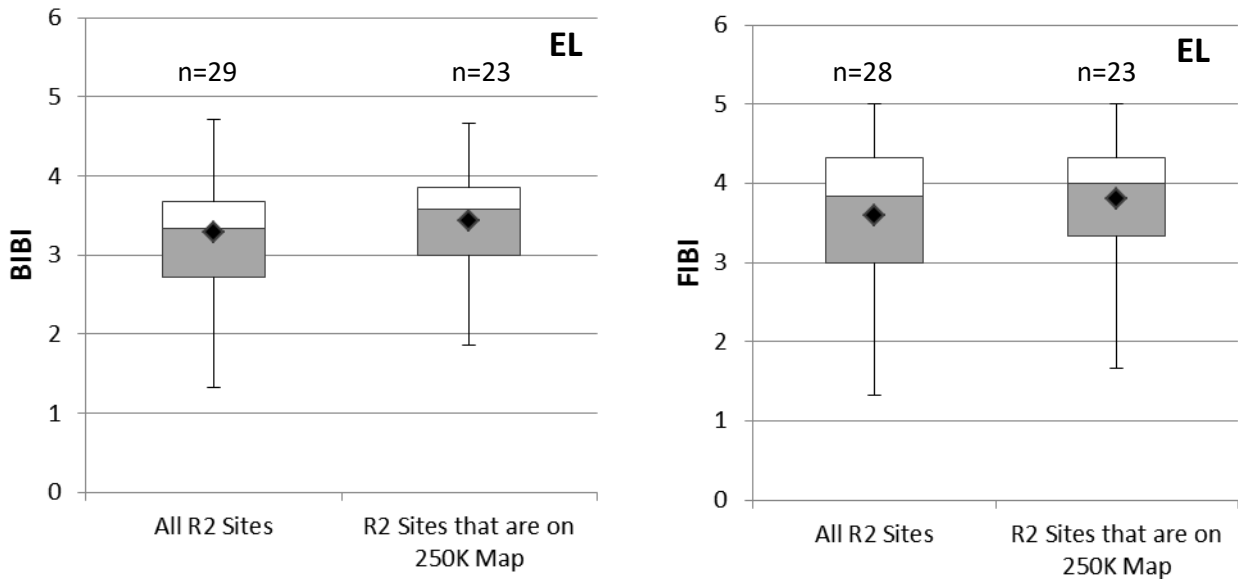


Figure A11. Comparison of biological indices in the Elk River basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

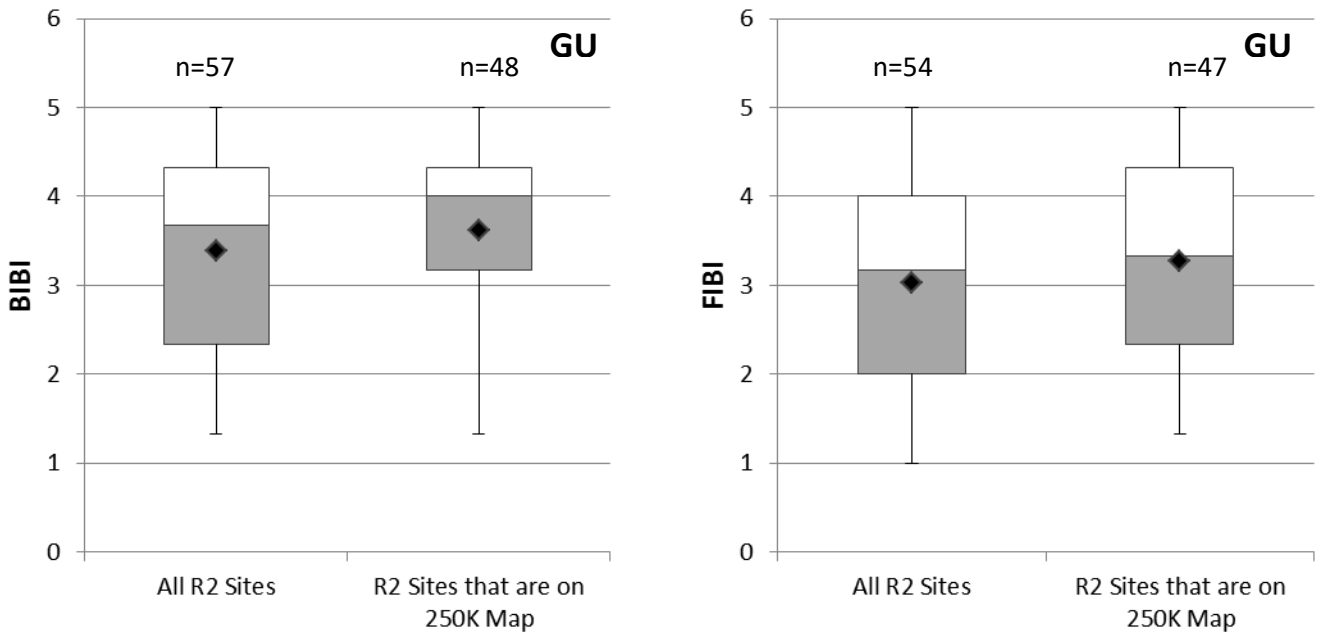


Figure A12. Comparison of biological indices in the Gunpowder River basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

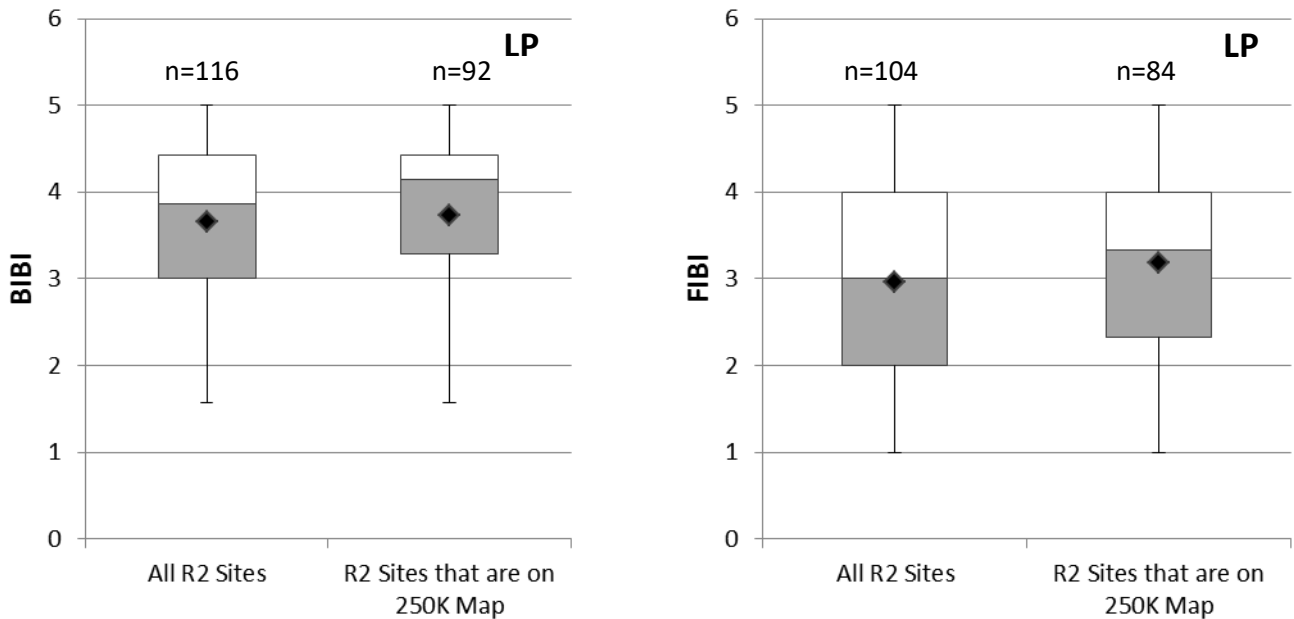


Figure A13. Comparison of biological indices in the Lower Potomac River basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

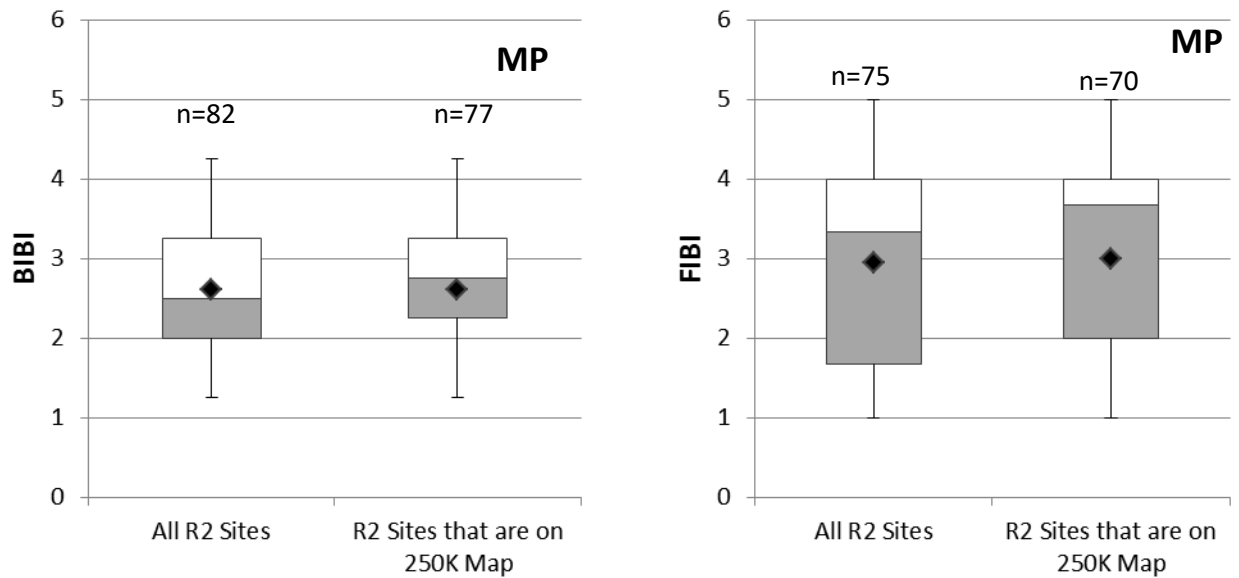


Figure A14. Comparison of biological indices in the Middle Potomac River basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

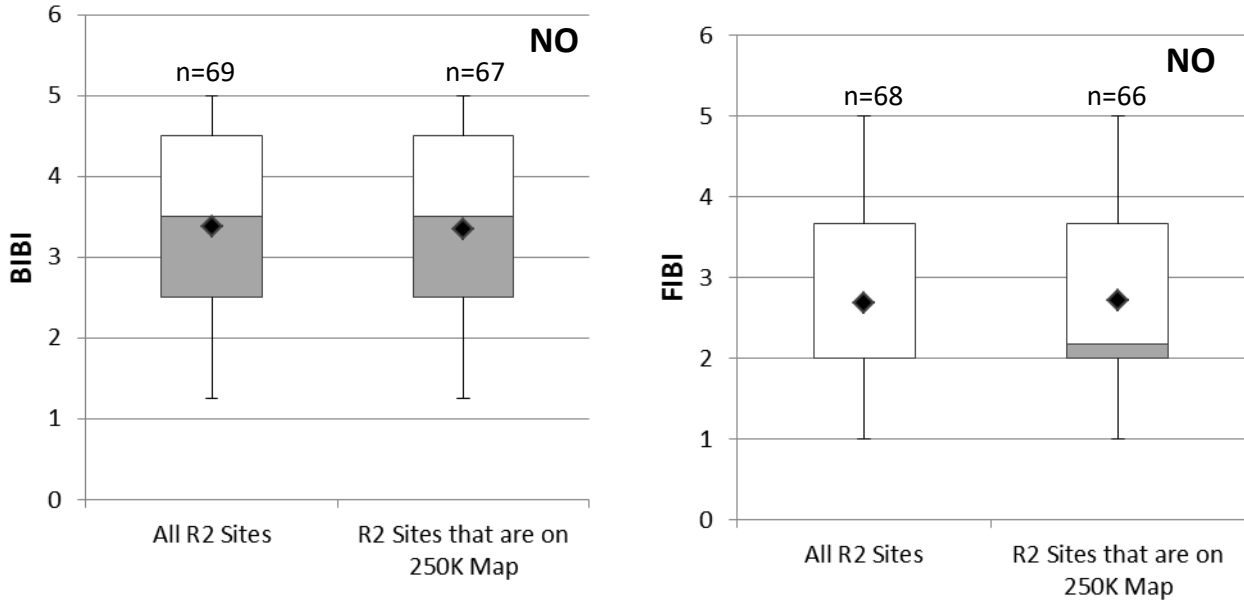


Figure A15. Comparison of biological indices in the North Branch Potomac River basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

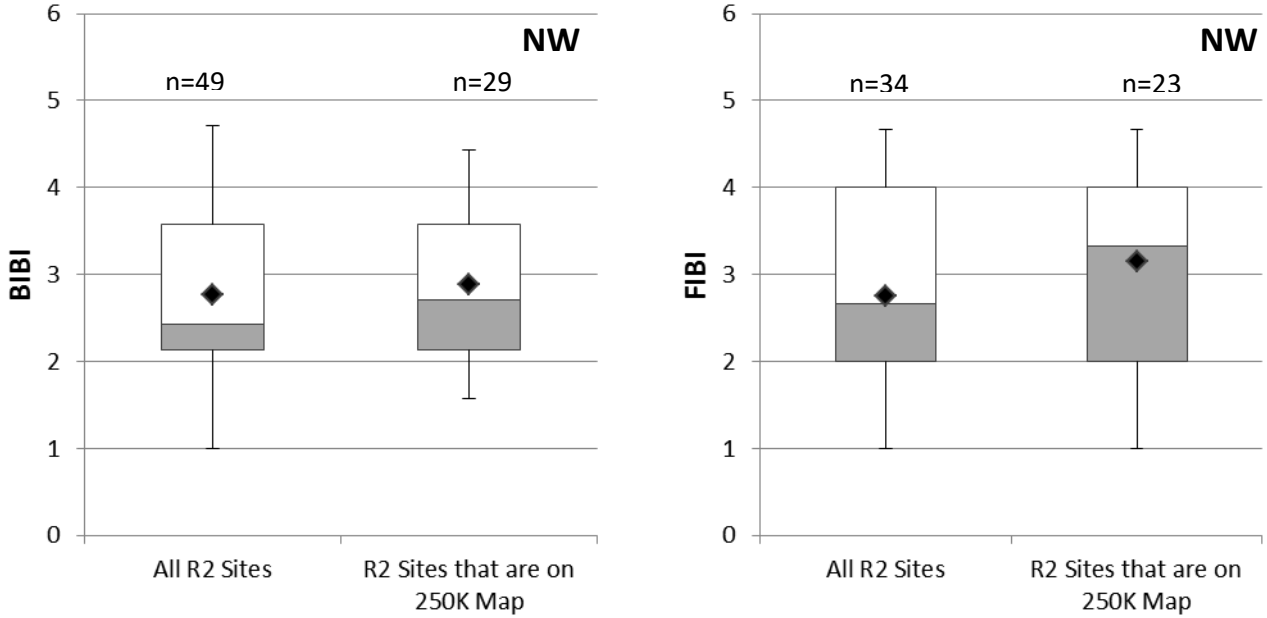


Figure A16. Comparison of biological indices in the Nanticoke River basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

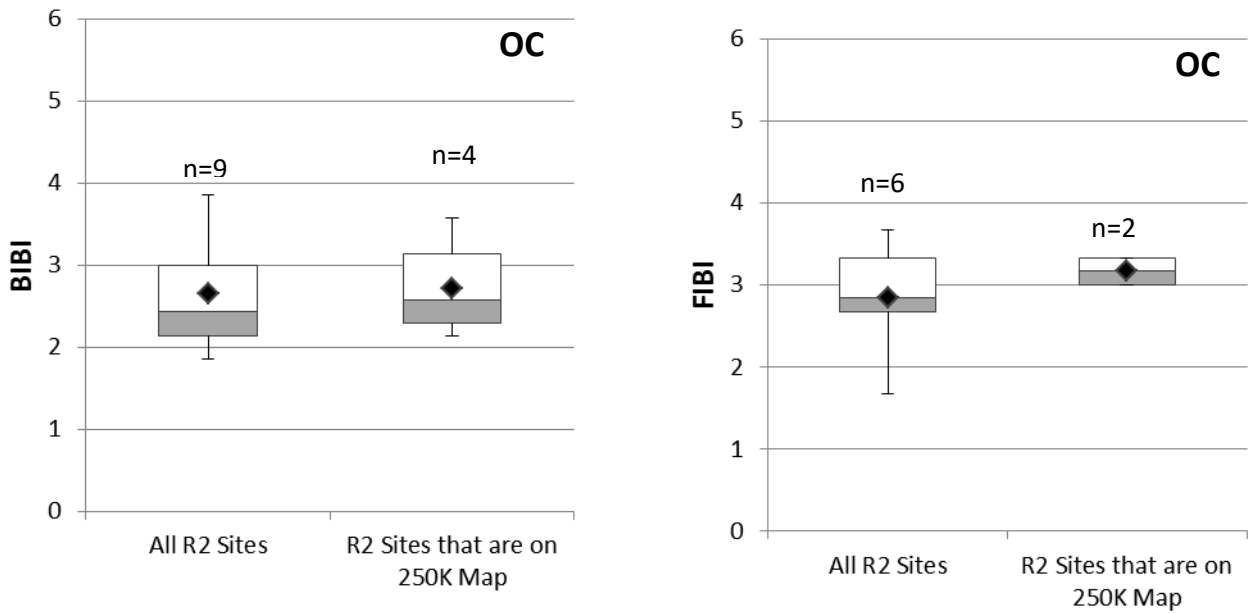


Figure A17. Comparison of biological indices in the Ocean Coastal basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

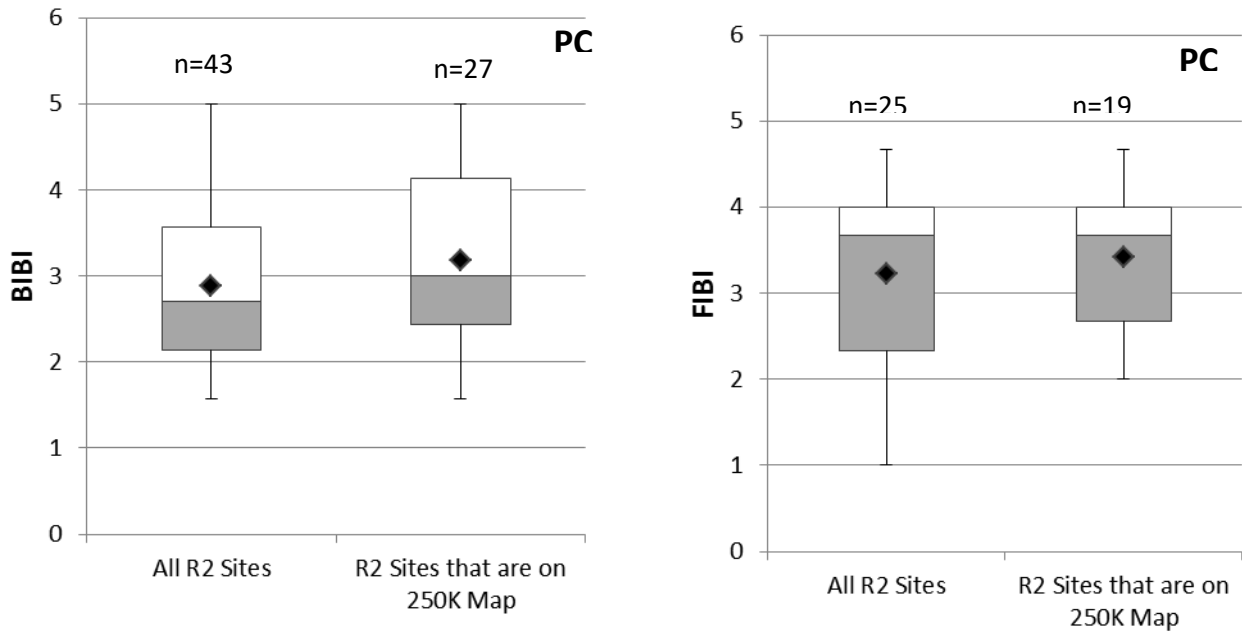


Figure A18. Comparison of biological indices in the Pocomoke River basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

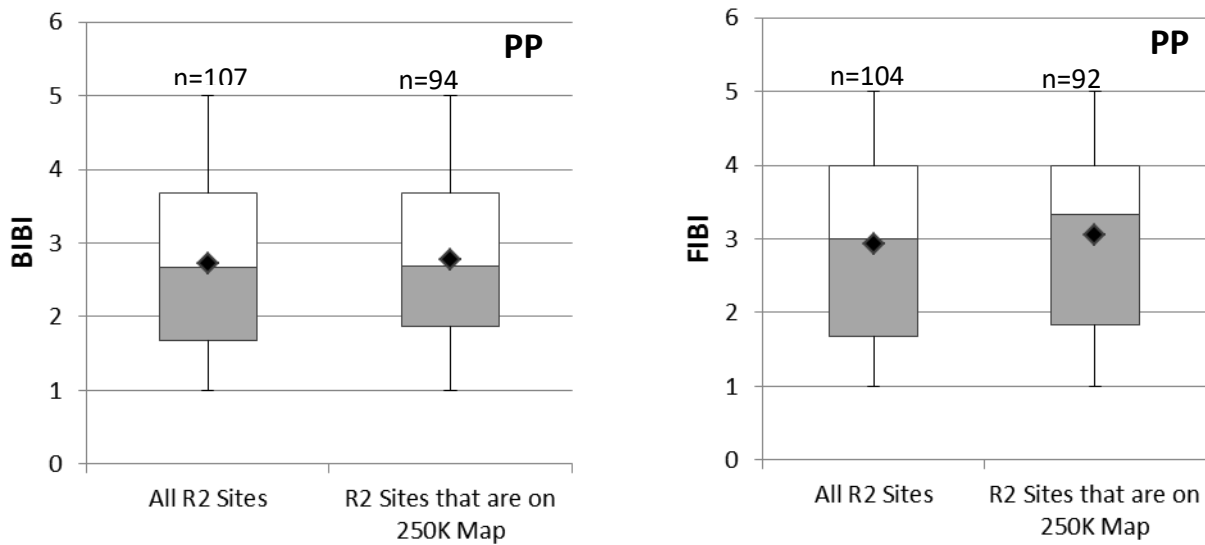


Figure A19. Comparison of biological indices in the Patapsco River basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

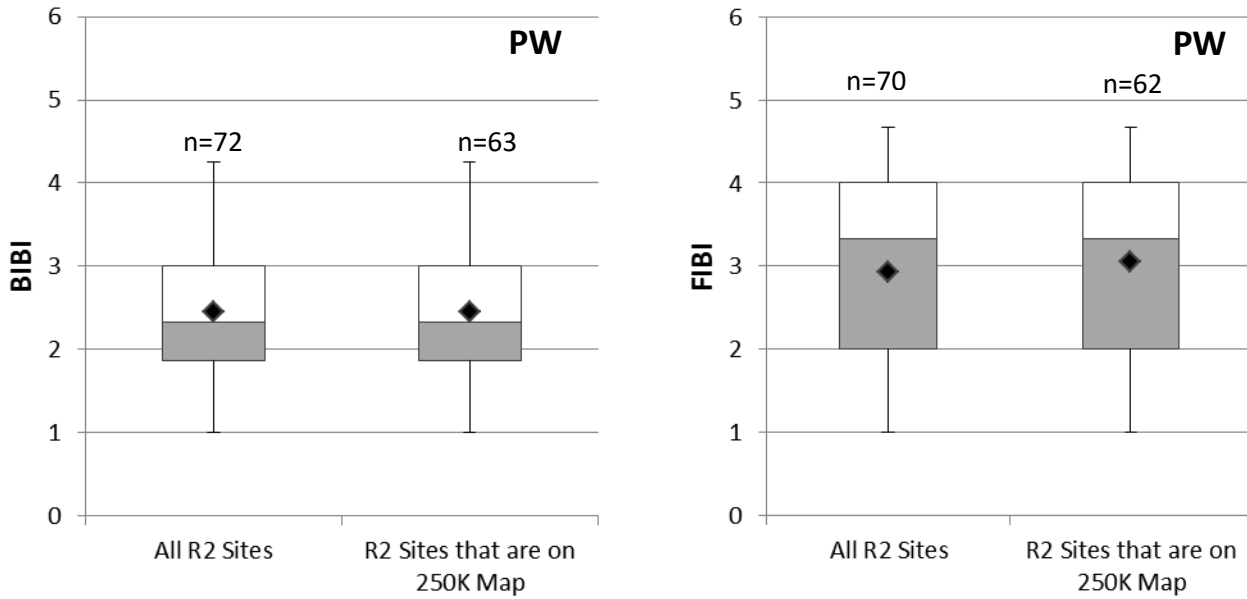


Figure A20. Comparison of biological indices in the Washington Metro Potomac River basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

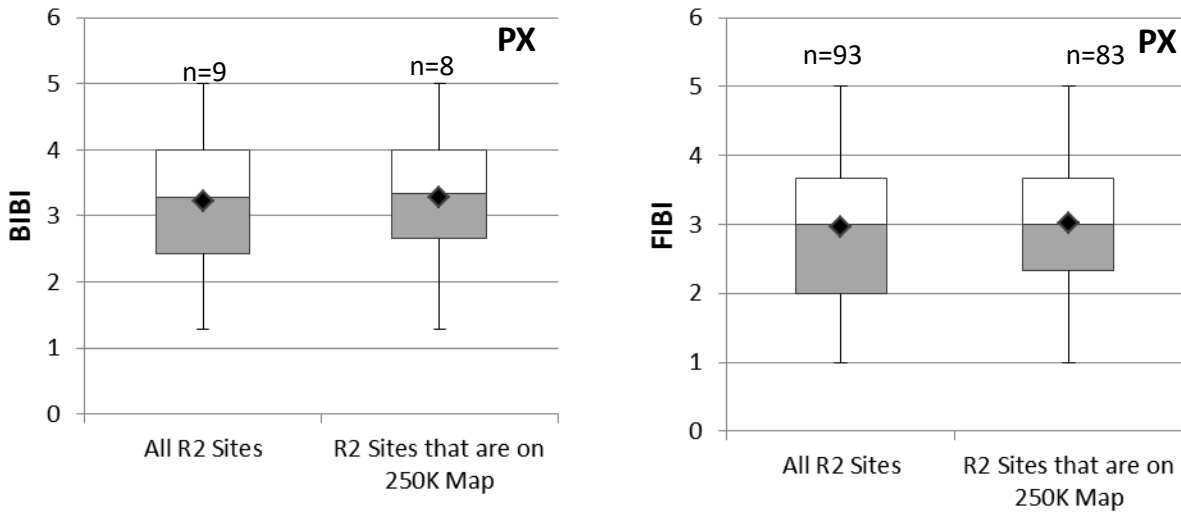


Figure A21. Comparison of biological indices in the Patuxent River basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

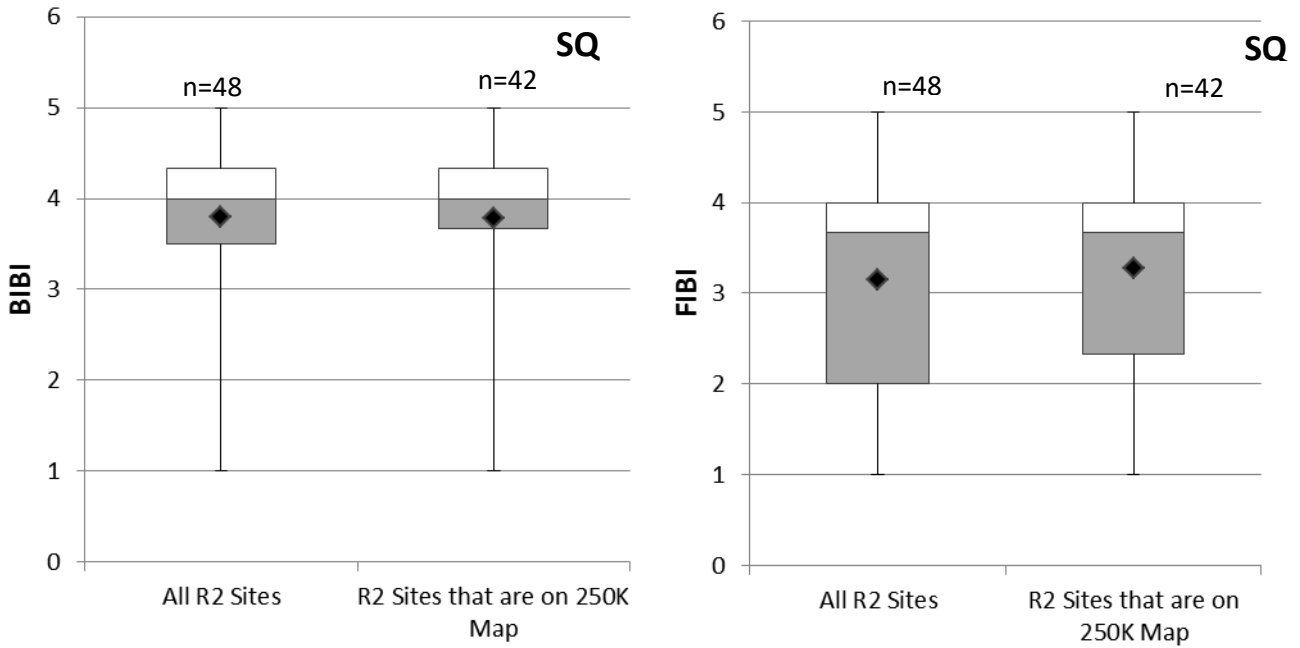


Figure A22. Comparison of biological indices in the Susquehanna River basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

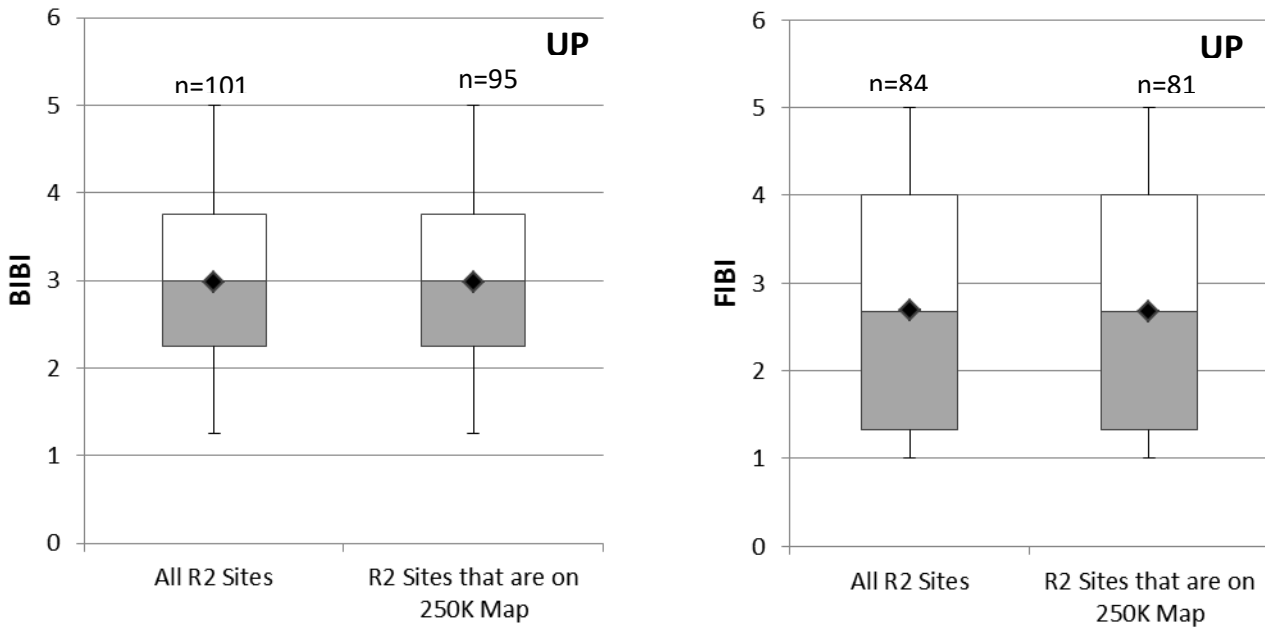


Figure A23. Comparison of biological indices in the Upper Potomac River basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

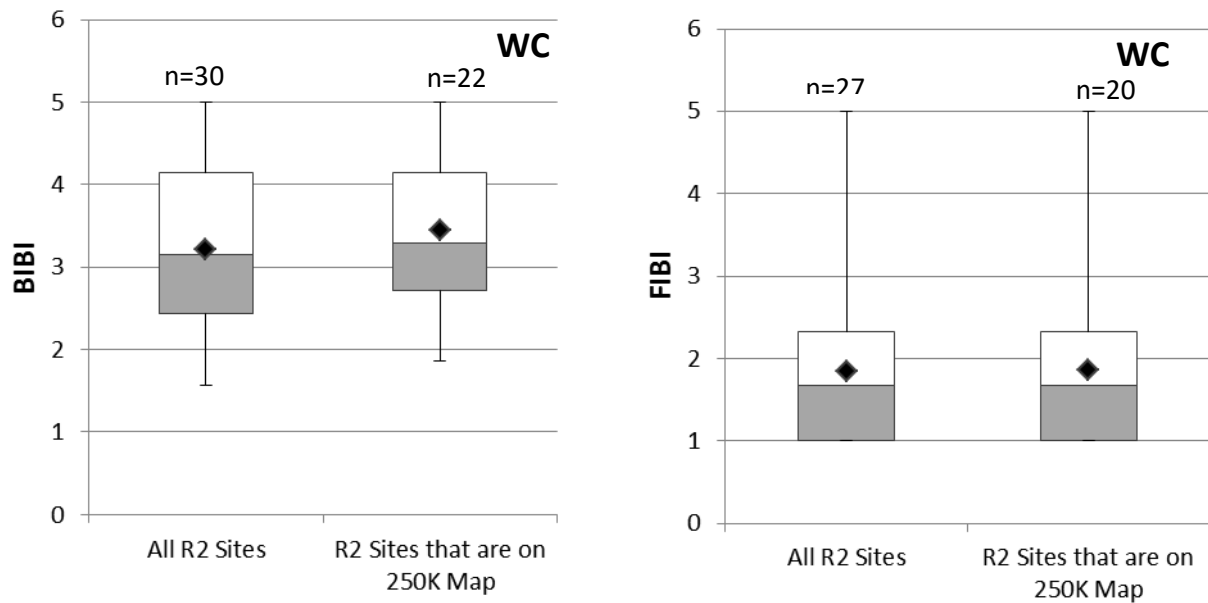


Figure A24. Comparison of biological indices in the West Chesapeake Bay basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

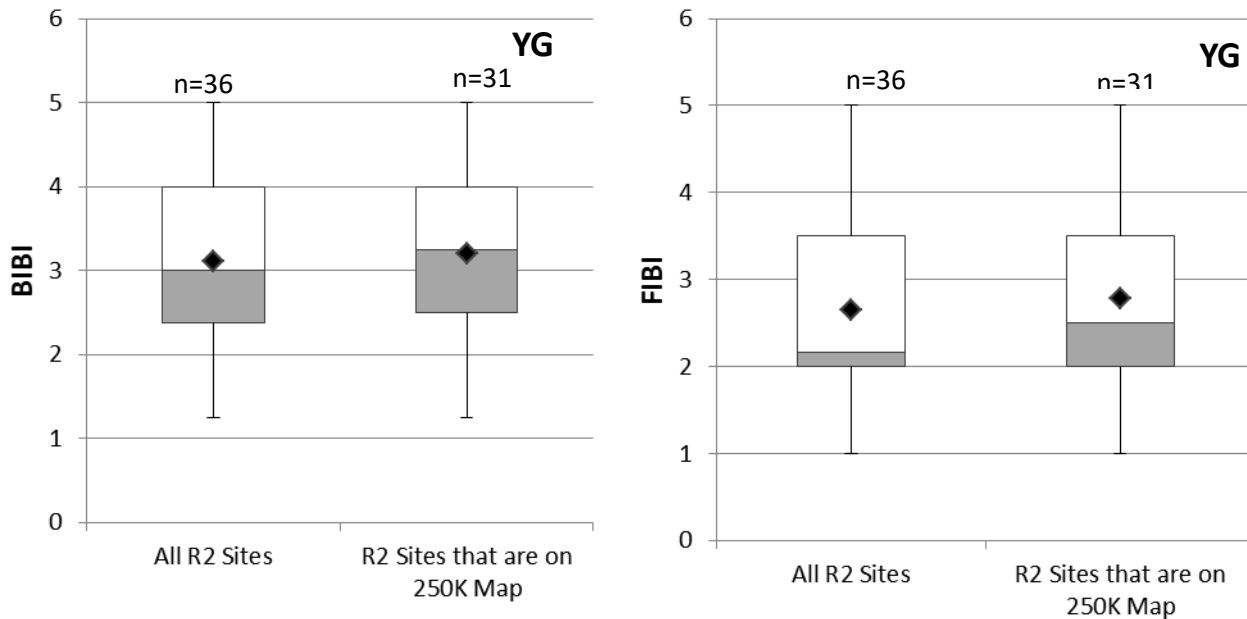


Figure A25. Comparison of biological indices in the Youghiogheny basin for all MBSS Round 2 sites vs. MBSS Round 2 sites that occur on the 1:250,000-scale map. Left Panel: BIBI. Right Panel: FIBI.

APPENDIX B POWER ANALYSIS OF SAMPLING SCENARIOS FOR MBSS ROUND 4

B1. METHODS

Power analysis was used to explore various sampling scenarios that involved different levels of error, statistical power, and sample size. Random sites from Round 1 of MBSS were used in the analysis. This was the most intensively sampled round and therefore was expected to have the greatest level of precision of the three rounds. Analyses were conducted for the statewide, ecoregion, basin, and county levels of spatial resolution and results for each are reported in separate sections.

Power analysis was conducted for the B-IBI in three ways to determine:

- The sample size required to distinguish a difference in the BIBI score from the 3.0 pass/fail threshold (Table 1) given a statistical power ranging from 0.1 to 1.0. The mean BIBI and standard deviation for each spatial unit from Round 1 MBSS were used in the analysis.
- The sample size required to distinguish a change in the BIBI score of ± 1.0 given a statistical power ranging from 0.1 to 1.0. The mean BIBI and standard deviation for each spatial unit from Round 1 MBSS were used in the analysis.
- The change in the BIBI score that could be detected given 1000 total survey sites (equivalent to Round 1 MBSS sampling intensity), 500 total survey sites (half of the Round 1 sampling intensity), or 170 total survey sites (about 10 per basin). The standard deviation in BIBI from Round 1 MBSS was used in the analysis. The total sample sizes considered (1000, 500, 170 sites) were divided evenly among the spatial units for each analysis, i.e.:
 - 1 State: 1000, 500, 170 sites
 - 3 Ecoregions: 330, 170 sites per ecoregion
 - 17 Basins: 60, 30, 10 sites per basin
 - 23 Counties plus Baltimore City: 40, 20, 10 sites per county

Power analysis was conducted for ANC (acid neutralizing capacity) in two ways to determine:

- The sample size required to distinguish a difference in the ANC value from the 200 $\mu\text{eq/L}$ threshold for acid sensitivity/non-sensitivity (Table 2), given the mean and standard deviation from R1 of sampling.
- The change in ANC that could be detected given 1000 total survey sites (equivalent to Round 1 MBSS sampling intensity), 500 total survey sites (half of the Round 1 sampling intensity), or 10 survey sites per spatial unit (e.g., county, basin). The standard deviations from Round 1 MBSS were used in the analysis. The total sample

sizes considered (1000, 500, 170 sites) were divided evenly among the spatial units for each analysis, i.e.:

- 1 State: 1000, 500, 170 sites
- 3 Ecoregions: 330, 170 sites per ecoregion
- 17 Basins: 60, 30, 10 sites per basin
- 23 Counties plus Baltimore City: 40, 20, 10 sites per county

Statistical power ≥ 0.8 was considered sufficient to be able to distinguish the mean B-IBI or ANC from the threshold values. Significance was assessed at $p = 0.05$. Power analysis was conducted at the statewide, ecoregion, basin, and county levels for both B-IBI and ANC. All analyses were carried out using SAS version 9.3.

Results from each analysis are presented in graphs from which the sampling effort necessary to attain the desired level of statistical power can be deduced for each level of spatial resolution. Actual means, standard deviations, and sample sizes for BIBI and ANC from Round 1 are reported in the Appendix for comparison with the output from the power analyses.

Value	Result
< 3	Fail
< = 3	Pass

< 0	Acidic
$\leq 0-50$	Highly Sensitive
$\leq 50-200$	Sensitive
≥ 200	Not Sensitive

B2. RESULTS OF BENTHIC INDEX OF BIOTIC INTEGRITY (BIBI) SAMPLE SIZE ASSESSMENT

B2.1 Statewide

State	Mean	StdDev	SE	RSE (%)	N
Maryland	3.00	1.09	0.04	1.18	954

B2.1.1 Detecting a 1.0 Change in BIBI at the State Level

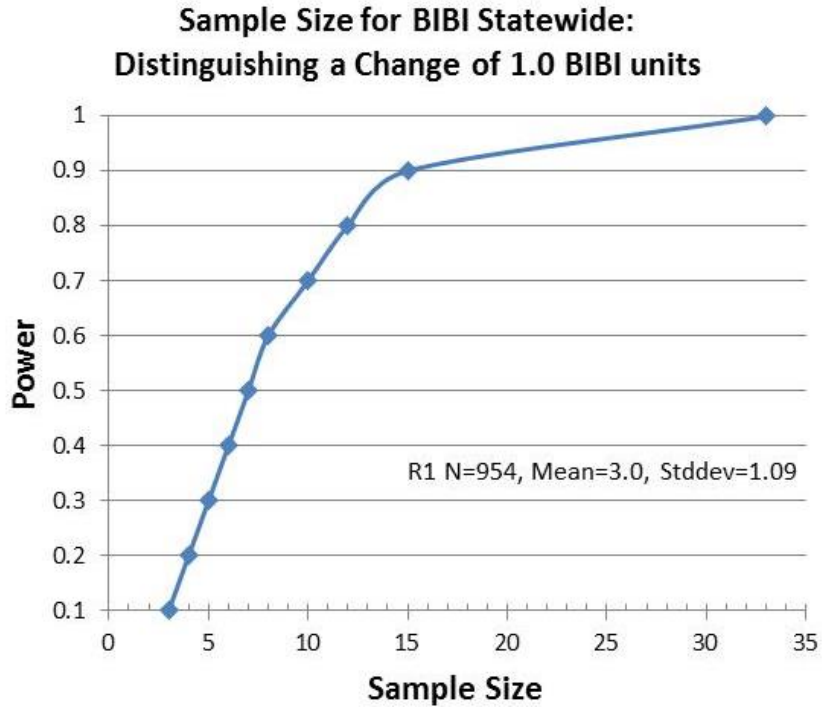


Figure B1. Sample size required with across a range of statistical power (0.1 to 1.0) to detect a change of 1.0 in BIBI units at the statewide level, given the mean and standard deviation from Round 1 of MBSS.

B2.1.2 Detecting a Difference from 3.0 in BIBI at the State Level

There is no sample size that would allow us to distinguish the statewide BIBI estimate from 3.0 because the statewide BIBI estimate in Round 1 MBSS was 3.0 with a standard deviation of 1.09.

B2.1.3 Detectable Change in BIBI at the State Level Using Three Potential Sample Sizes

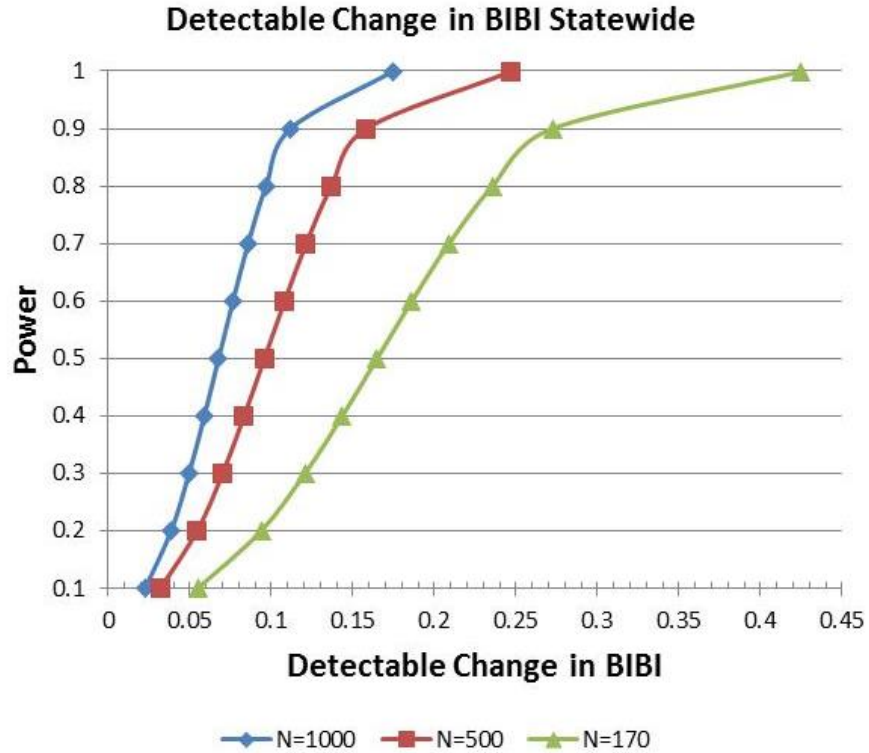


Figure B2. The change in the BIBI score that could be detected at the statewide level given a range of statistical power (0.1 to 1.0), and given the mean and standard deviation from Round 1 of sampling, if a sample size of 1000, 500, or 170 were used.

B2.2 Ecoregion

Table B4. Ecoregion BIBI summary statistics from Round 1 MBSS					
Ecoregion	Mean	StdDev	SE	RSE (%)	N
COASTAL	3.20	1.16	0.07	2.04	316
EPIEDMONT	3.08	1.09	0.06	2.10	284
HIGHLAND	2.77	0.98	0.05	1.88	354

B2.2.1 Detecting a 1.0 Change in BIBI at the Ecoregion Level

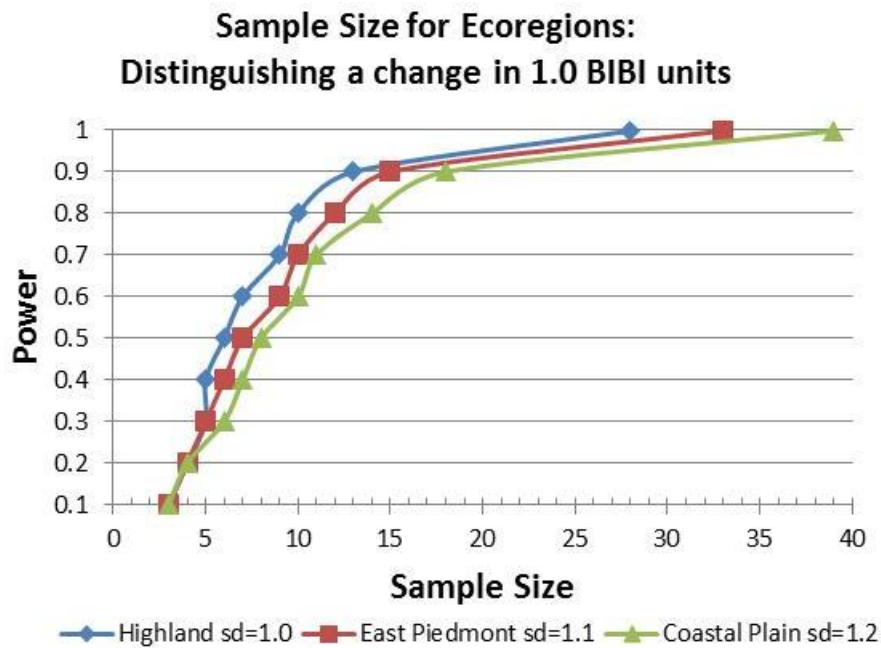


Figure B3. Sample size required to detect a change of 1.0 in BIBI units at the ecoregion level, given the means and standard deviations for each ecoregion from Round 1 of MBSS.

B2.2.2 Detecting a Difference from 3.0 in BIBI at the Ecoregion Level

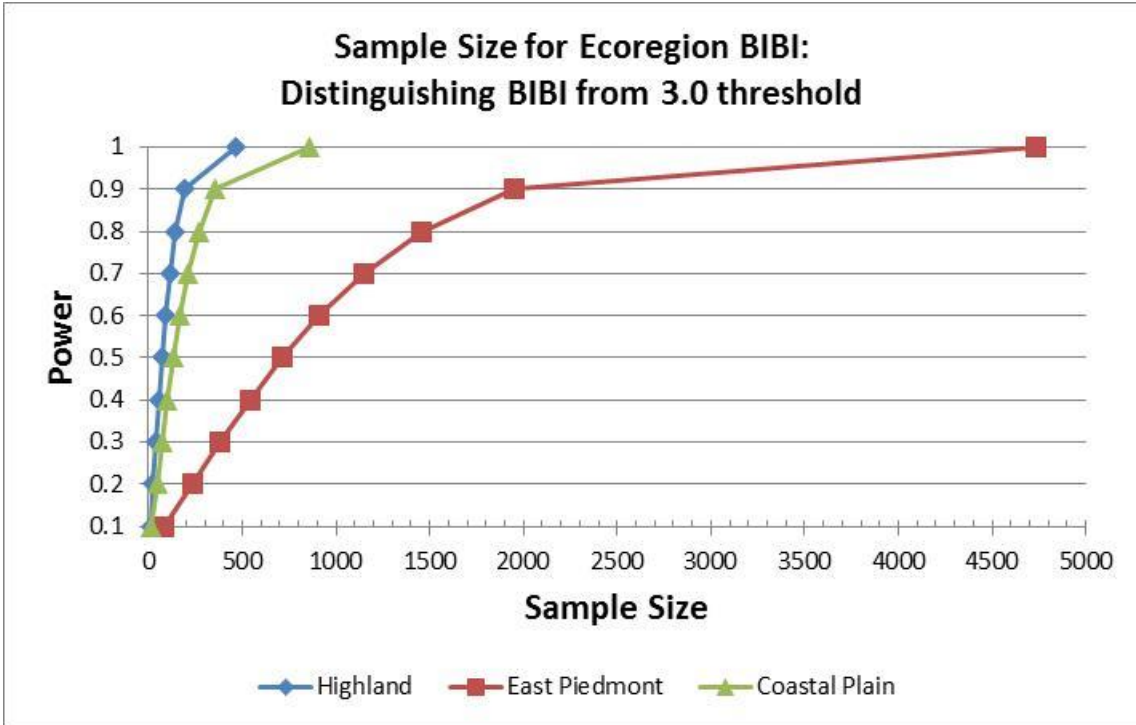


Figure B4. Sample size required to detect a significant difference from 3.0 in BIBI units at the ecoregion level, given the means and standard deviations for each ecoregion from Round 1 of MBSS.

B2.2.3 Detectable Change in BIBI at the Ecoregion Level Using Two Potential Sample Sizes

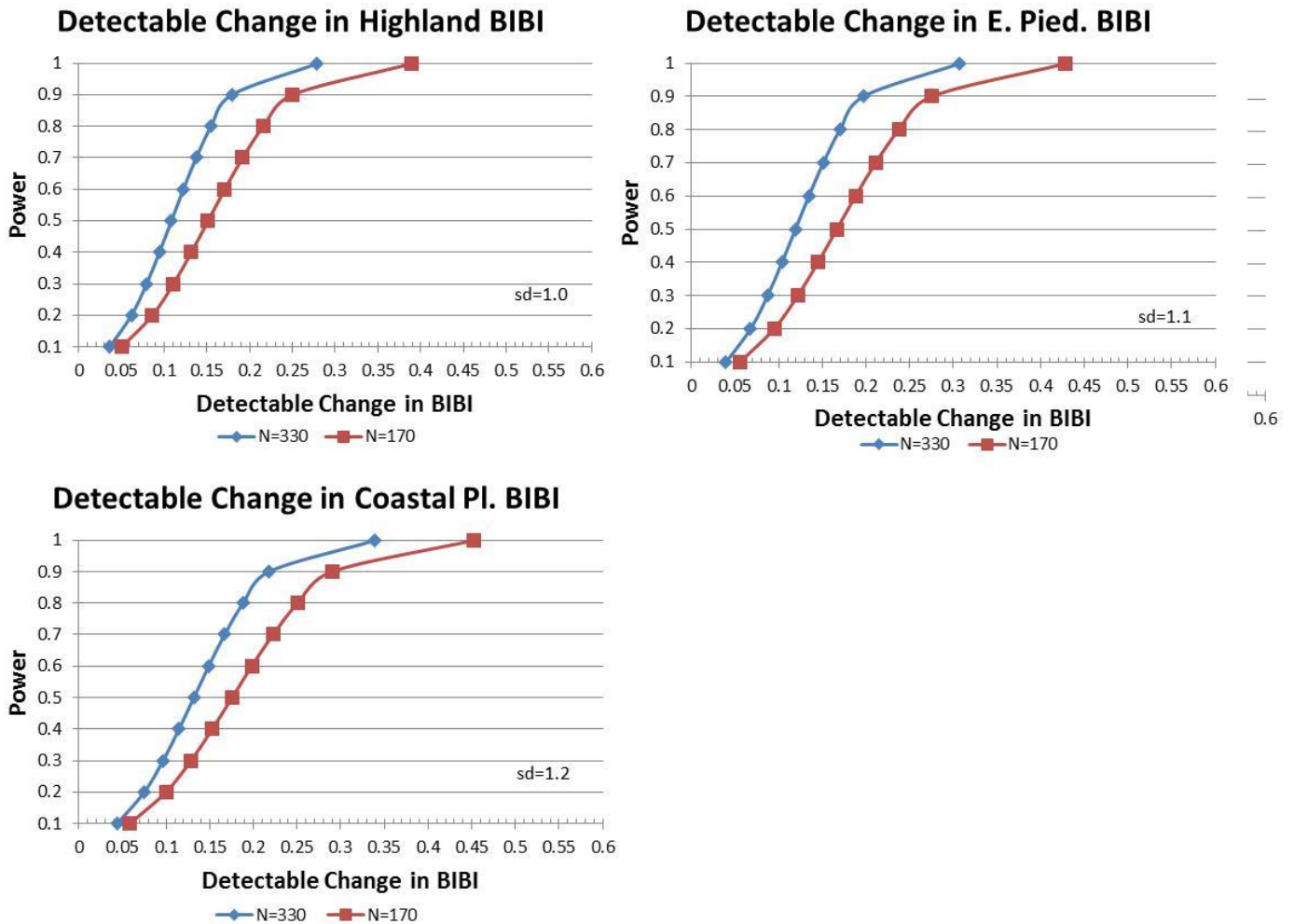


Figure B5. The detectable change in BIBI per ecoregion given the mean and standard deviation from Round 1 of MBSS, power=0.1 to 1.0 and sample size = 330 or 170.

B2.3 Basins

Table B5. Basin BIBI summary statistics from Round 1 MBSS					
Basin	Mean	StdDev	SE	RSE (%)	N
BU	2.30	0.94	0.21	9.08	20
CK	3.09	1.09	0.16	5.20	46
CR	3.77	0.94	0.15	3.85	42
EL	3.46	0.91	0.21	6.20	18
GU	3.86	0.75	0.11	2.89	45
LP	3.93	1.04	0.14	3.61	54
MP	2.13	0.67	0.06	3.00	109
NO	3.13	0.98	0.12	3.97	62
NW	3.38	1.10	0.26	7.64	18
PC	2.79	0.97	0.17	6.06	33
PP	2.82	1.09	0.10	3.39	129
PW	2.25	0.96	0.11	5.07	71
PX	3.18	1.14	0.13	3.96	82
SQ	3.57	0.76	0.13	3.51	37
UP	3.04	0.96	0.12	3.85	68
WC	2.64	0.97	0.16	6.23	35
YG	3.27	0.86	0.09	2.86	85

B2.3.1 Detecting a 1.0 Change in BIBI at the Basin Level

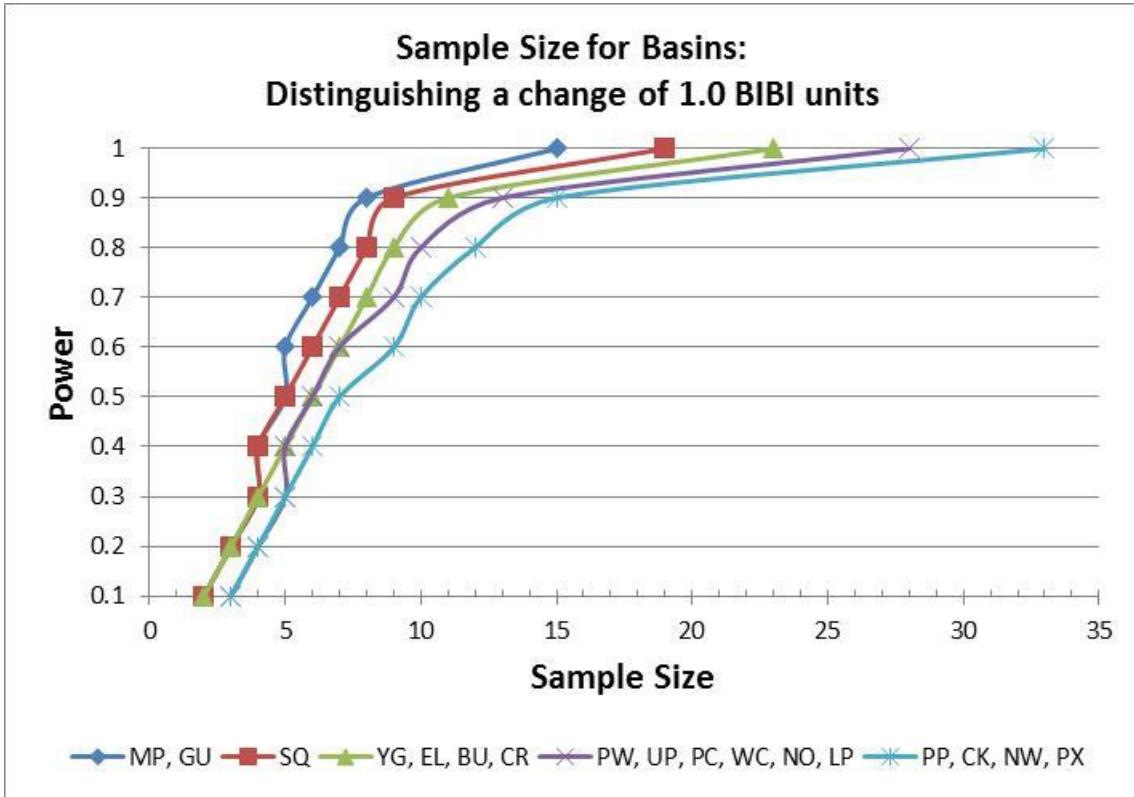


Figure B6. Sample size required to detect a change of 1.0 in BIBI units at the basin level, given the means and standard deviations for each basin from Round 1 of MBSS. Lines indicate results for groups of basins that had similar standard deviations.

B2.3.2 Detecting a Difference from 3.0 in BIBI at the Basin Level

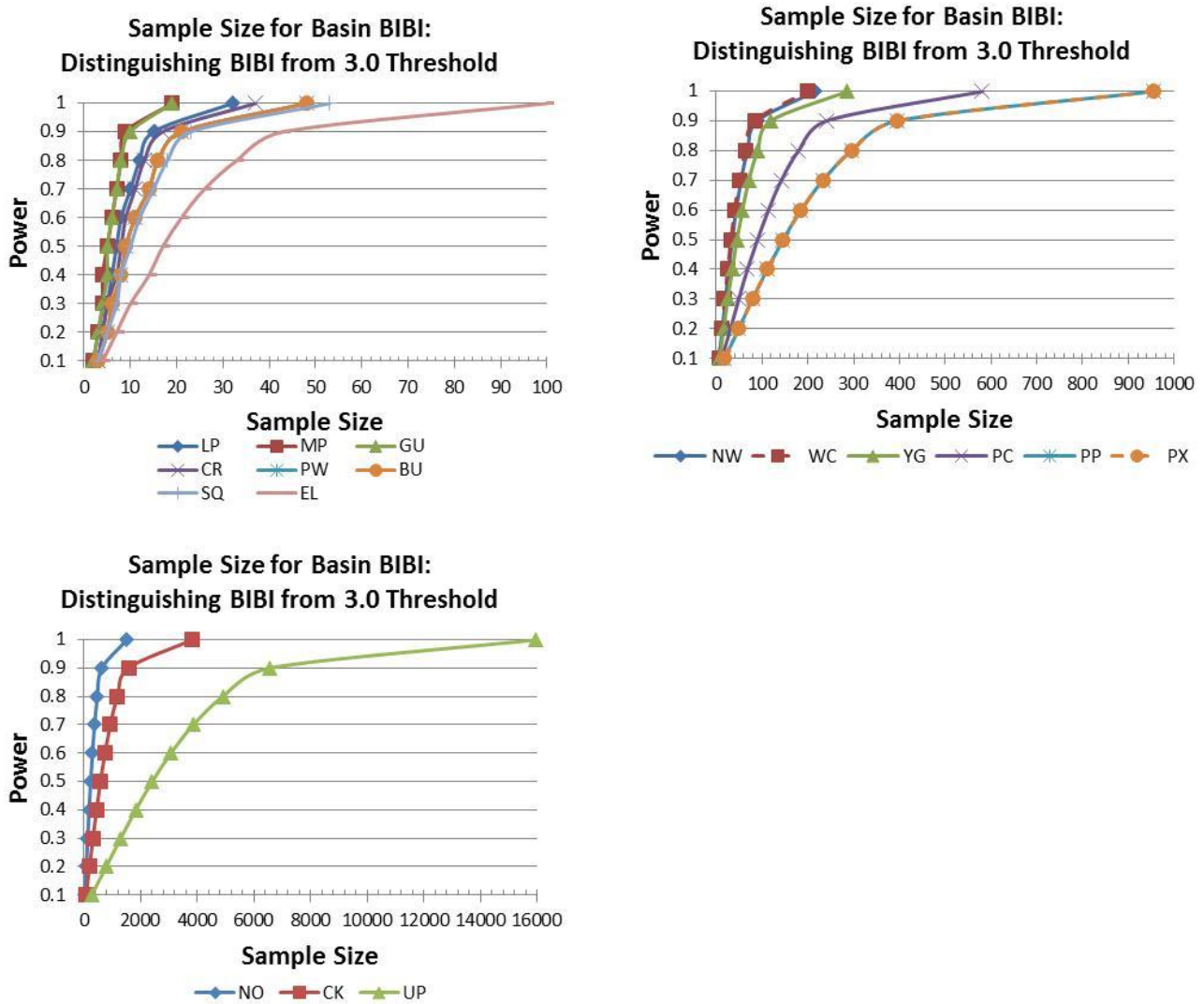


Figure B7. Sample size required to detect a significant difference from 3.0 BIBI pass/fail threshold at the basin level, given the means and standard deviations for each basin from Round 1 of MBSS. Individual panels depict basins with similar sample size requirements.

B2.3.3 Detectable Change in BIBI at the Basin Level Using Three Potential Sample Sizes

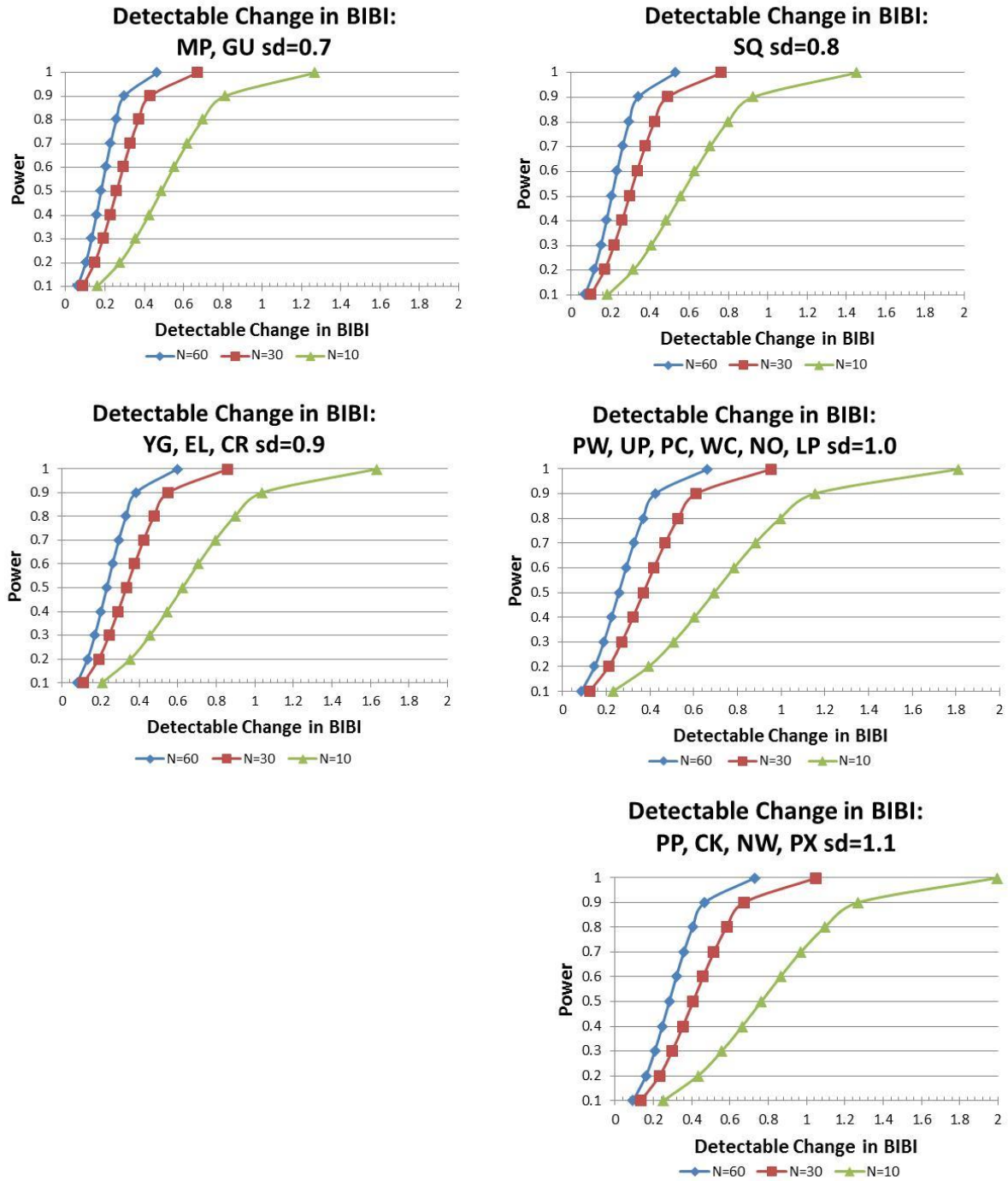


Figure B8. The detectable change in BIBI per basin given the mean and standard deviation from Round 1 of MBSS, power = 0.1 to 1.0 and sample size = 60, 30, 10 per basin. Individual panels represent basins with the same standard deviations in BIBI from Round 1 of MBSS and therefore have the same results from the power analysis.

B2.4 County

Table B6. County BIBI summary statistics from Round 1 MBSS					
County	Mean	Stdev	SE	RSE (%)	N
AA	2.66	1.12	0.16	6.00	49
AL	3.18	0.99	0.12	3.90	64
BA	3.26	1.26	0.14	4.37	78
BC	1.79	0.52	0.15	8.42	12
CA	2.82	1.07	0.27	9.47	16
CE	3.38	0.88	0.16	4.85	29
CH	4.15	0.85	0.13	3.19	41
CN	2.94	1.11	0.23	7.85	23
CR	2.82	0.93	0.10	3.54	88
DO	3.57	1.07	0.48	13.39	5
FR	2.10	0.65	0.08	3.65	72
GA	3.29	0.87	0.08	2.45	115
HA	3.14	1.06	0.16	5.03	45
HO	3.01	1.08	0.17	5.60	41
KE	2.88	0.87	0.33	11.44	7
MO	2.38	0.99	0.12	5.15	65
PG	2.79	1.05	0.16	5.66	44
QA	3.72	0.97	0.15	3.94	44
SM	4.00	0.97	0.24	6.05	16
SO	2.37	0.51	0.23	9.64	5
TA	3.48	1.14	0.29	8.20	16
WA	2.63	0.87	0.14	5.32	39
WI	3.01	1.05	0.18	6.09	33
WO	2.86	1.11	0.45	15.86	6

B2.4.1 Detecting a 1.0 Change in BIBI at the County Level

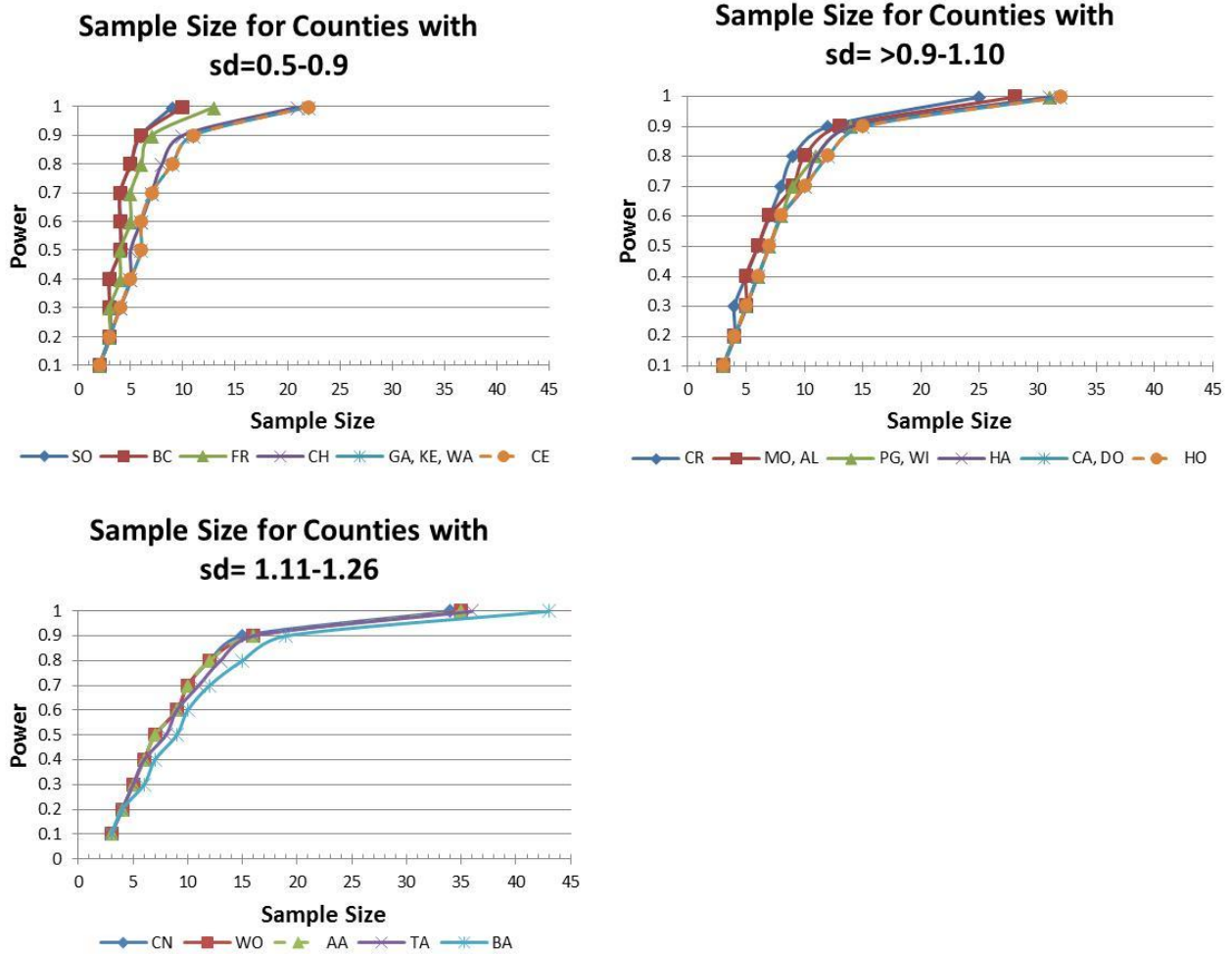


Figure B9. Sample size required to detect a change of 1.0 in BIBI units at the county level, given the means and standard deviations for each county from Round 1 of MBSS. Individual panels represent counties with similar standard errors from Round 1 MBSS. Lines represent specific counties.

B2.4.2 Detecting a Difference from 3.0 in BIBI at the County Level

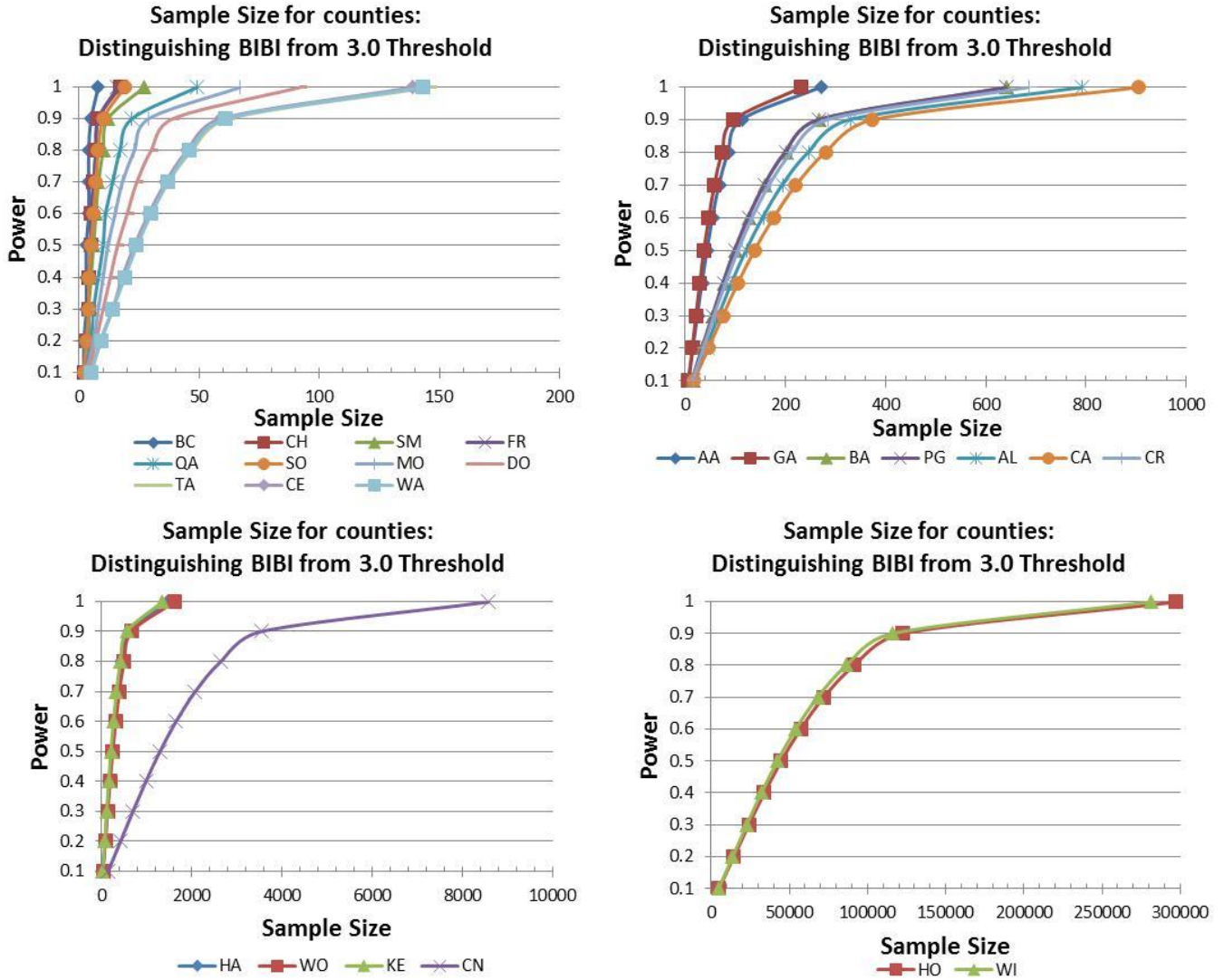


Figure B10. Sample size required to detect a significant difference from the 3.0 BIBI threshold at the county level, given the means and standard deviations for each county from Round 1 of MBSS. Individual panels represent counties with similar sample size requirements. Lines indicate specific counties.

B2.4.3 Detectable change in BIBI at the county level using three potential sample sizes

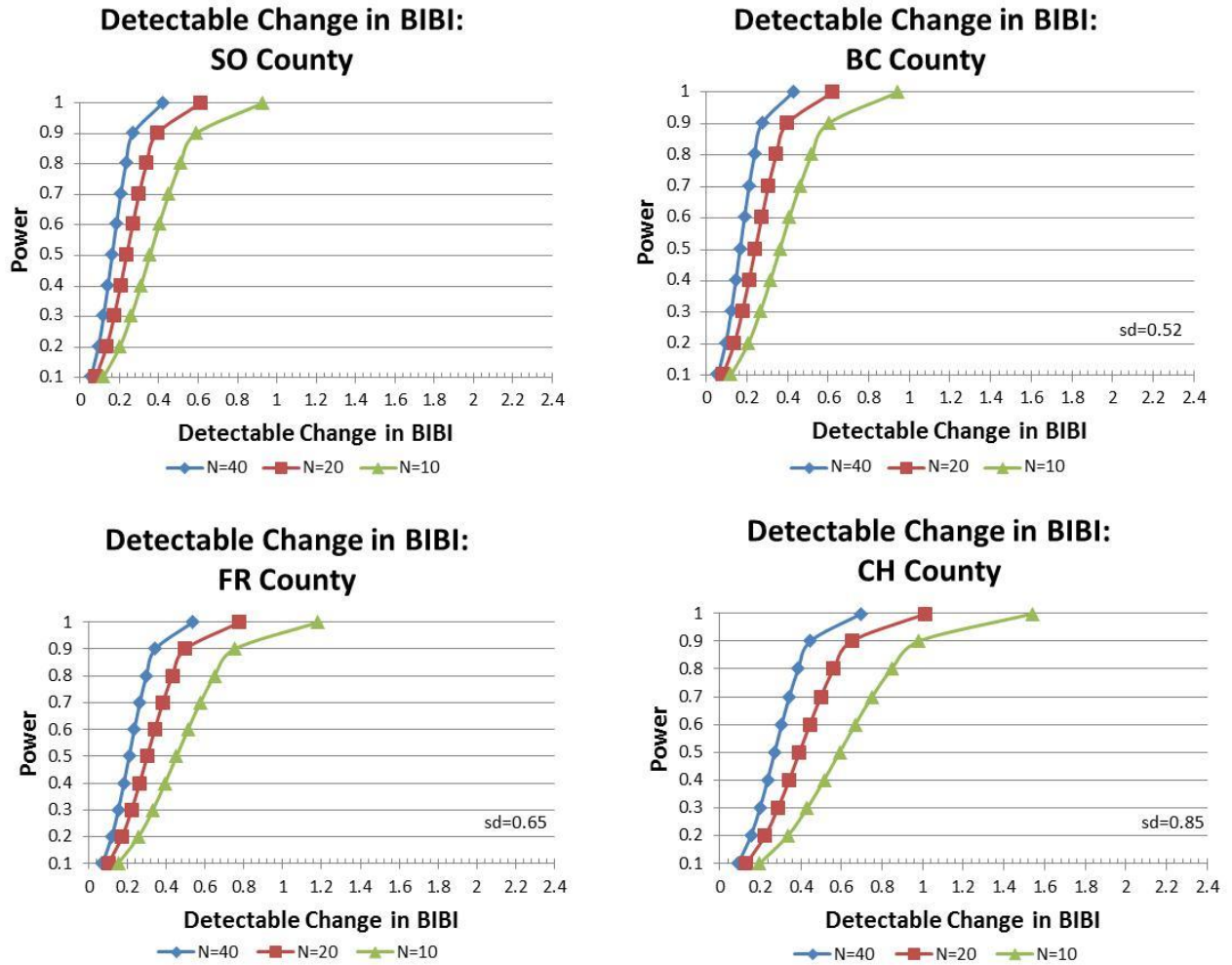


Figure B11. The detectable change in BIBI per county given the mean and standard deviation from Round 1 of MBSS, power=0.1 to 1.0 and sample size =40 (blue line), 20 (red line), 10 per basin (green line). Individual panels represent counties with the same standard deviations in BIBI from Round 1 of MBSS.

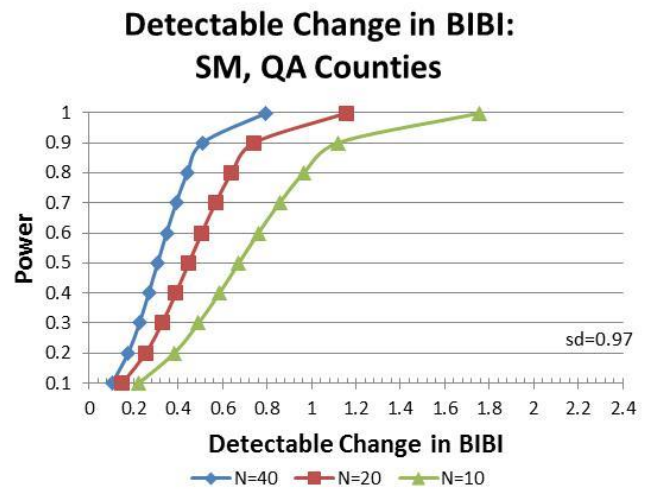
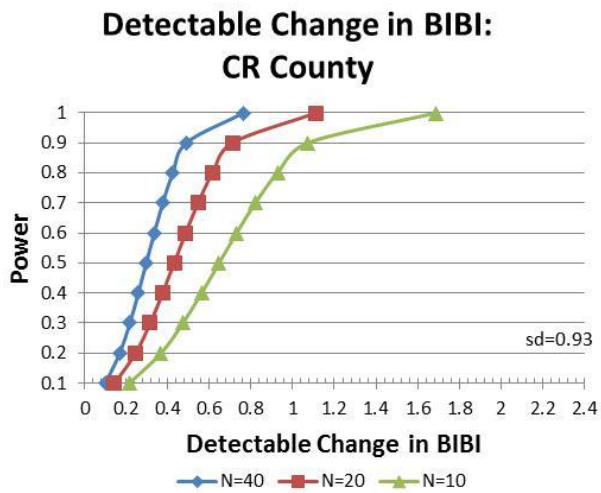
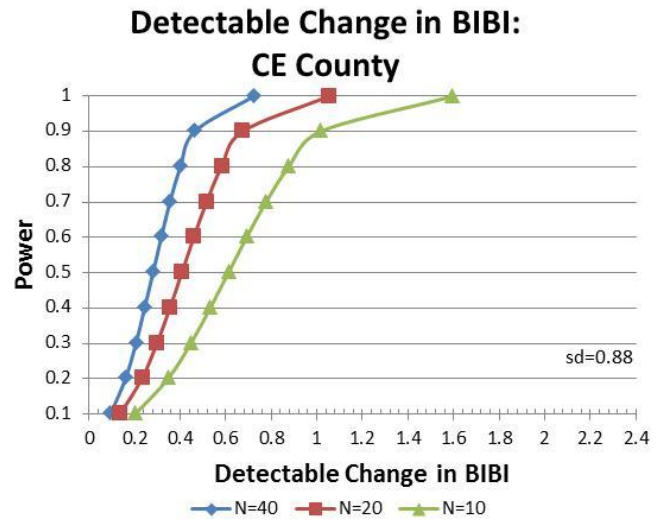
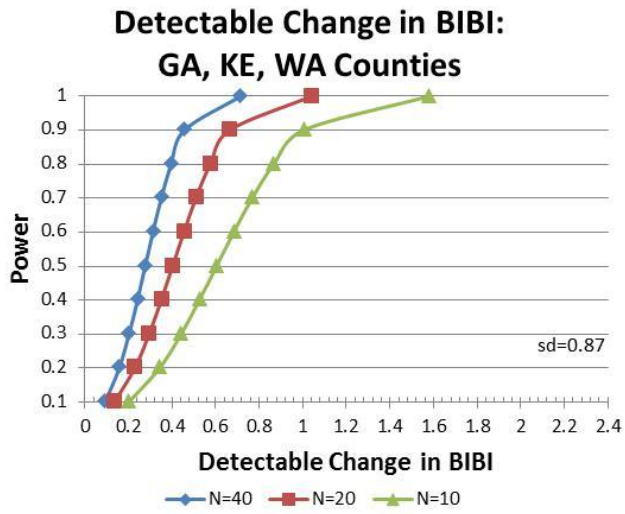
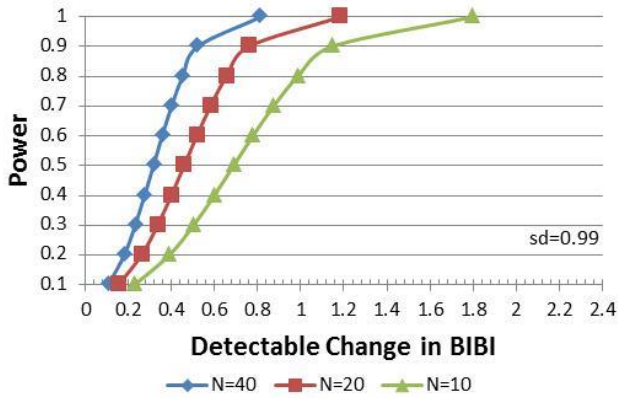
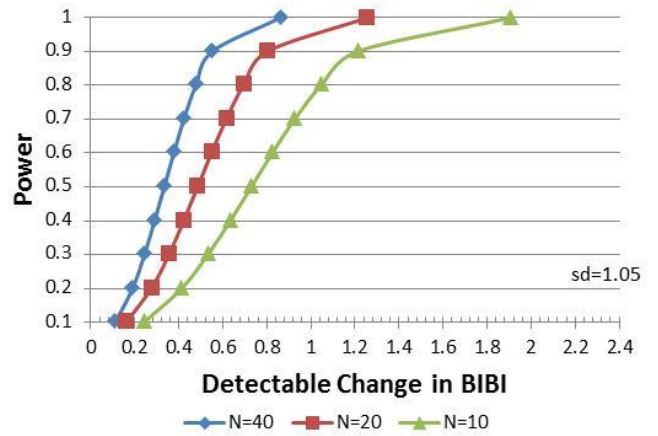


Figure B11. Continued.

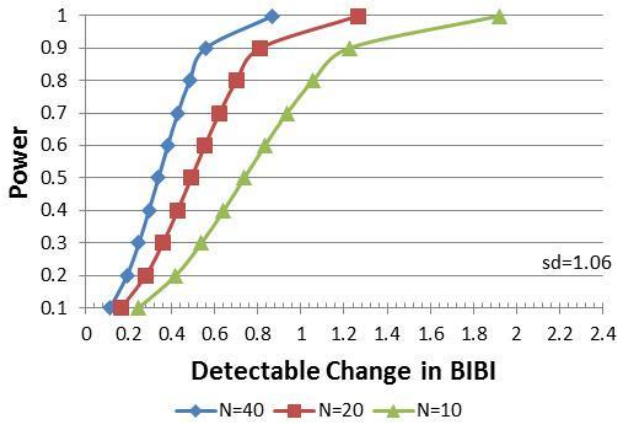
**Detectable Change in BIBI:
MO, AL Counties**



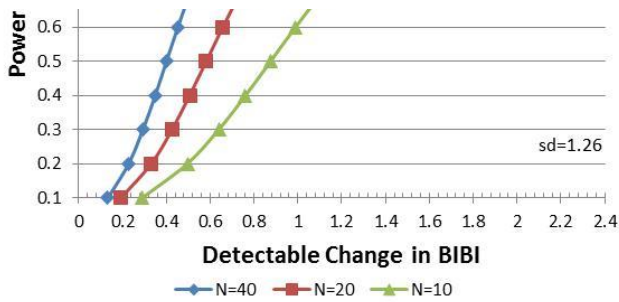
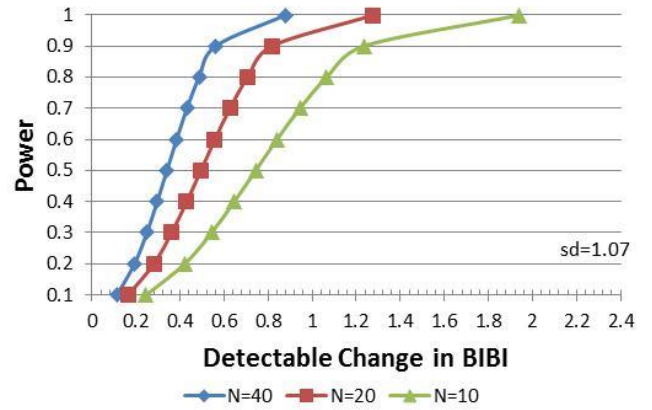
**Detectable Change in BIBI:
PG, WI Counties**



**Detectable Change in BIBI:
HA County**



**Detectable Change in BIBI:
CA, DO Counties**



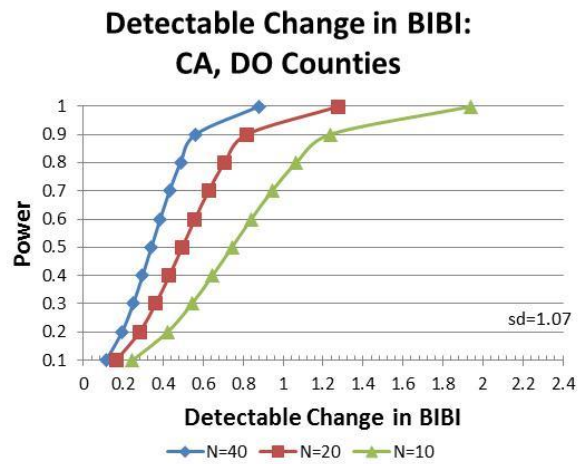
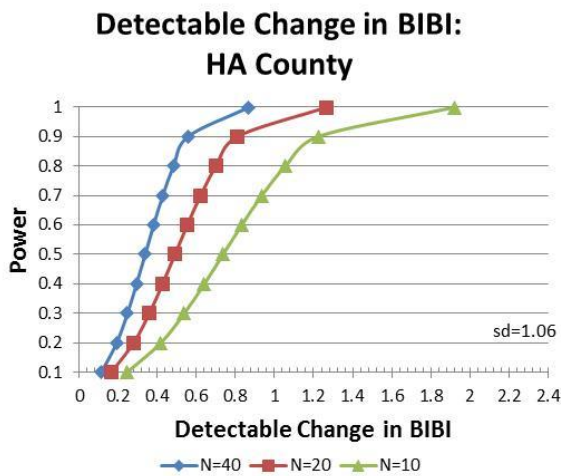
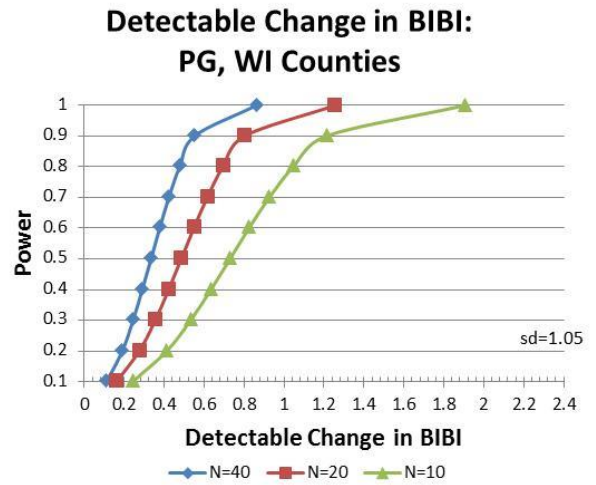
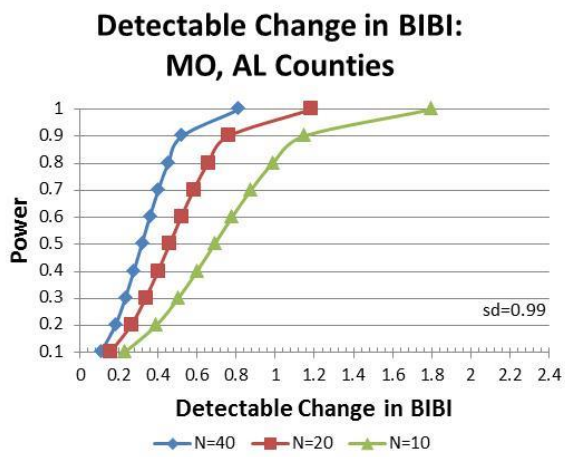


Figure B11. Continued

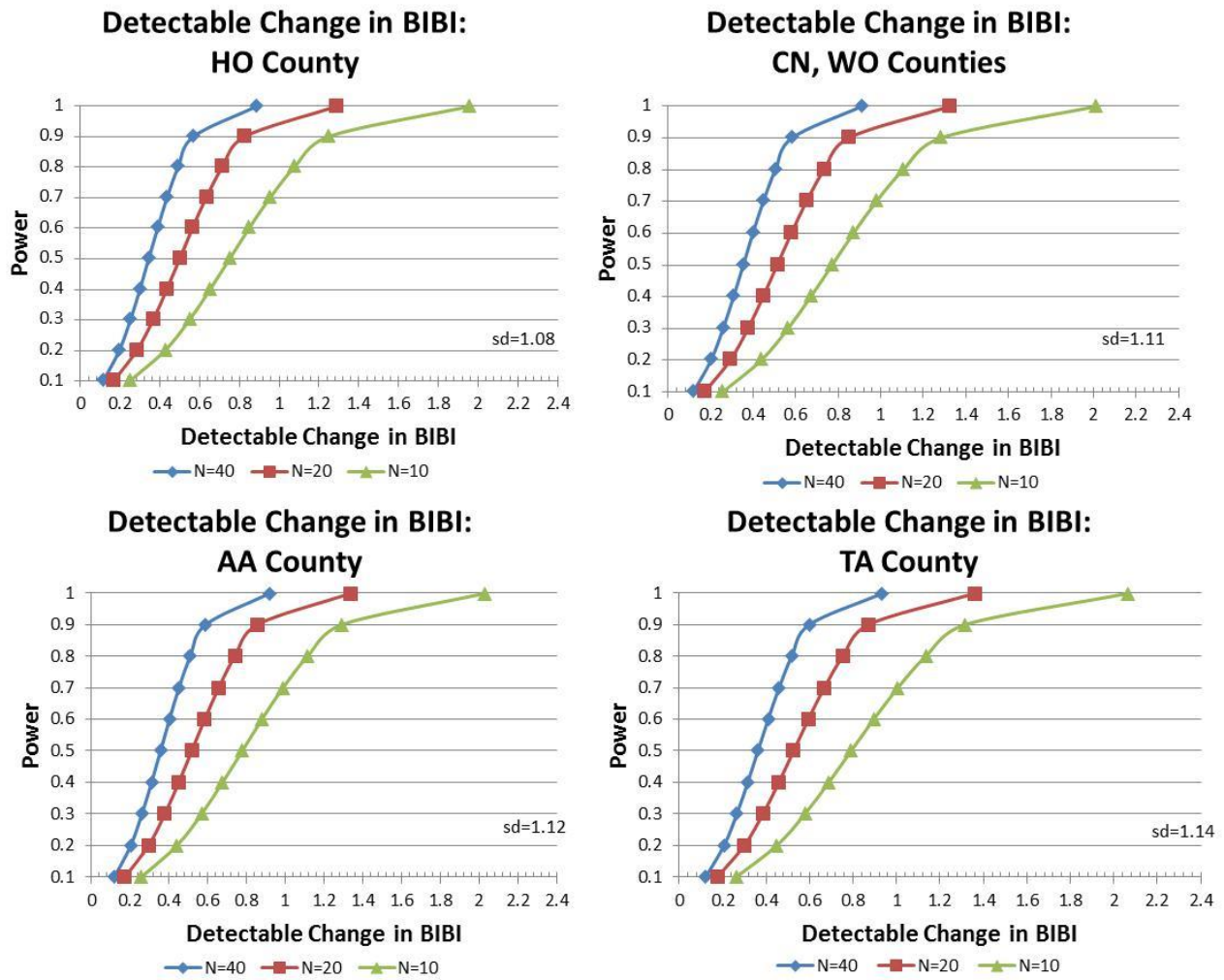


Figure B11. Continued.

B3. RESULTS OF ACID NEUTRALIZING CAPACITY (ANC) SAMPLE SIZE ASSESSMENT

B3.1 Statewide

Table B7. Statewide ANC summary statistics from Round 1 MBSS					
State	Mean	StdDev	SE	RSE (%)	n
Maryland	589	726	23.51	3.99	954

B3.1.1 Detecting a Difference from 200 $\mu\text{eq/L}$ at the State Level

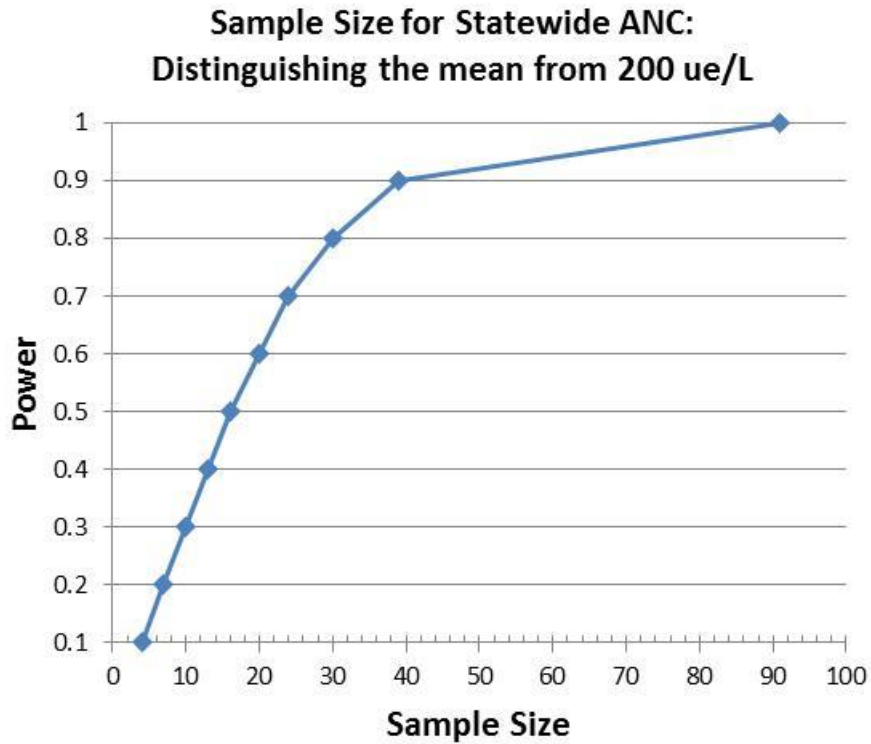


Figure B12. Sample size required across a range of statistical power (0.1 to 1.0) to detect significant difference from 200 $\mu\text{eq/L}$ ANC at the statewide level, given the mean and standard deviation from Round 1 of MBSS.

B3.1.2 Detectable Change in ANC at the State Level Using Three Potential Sample Sizes

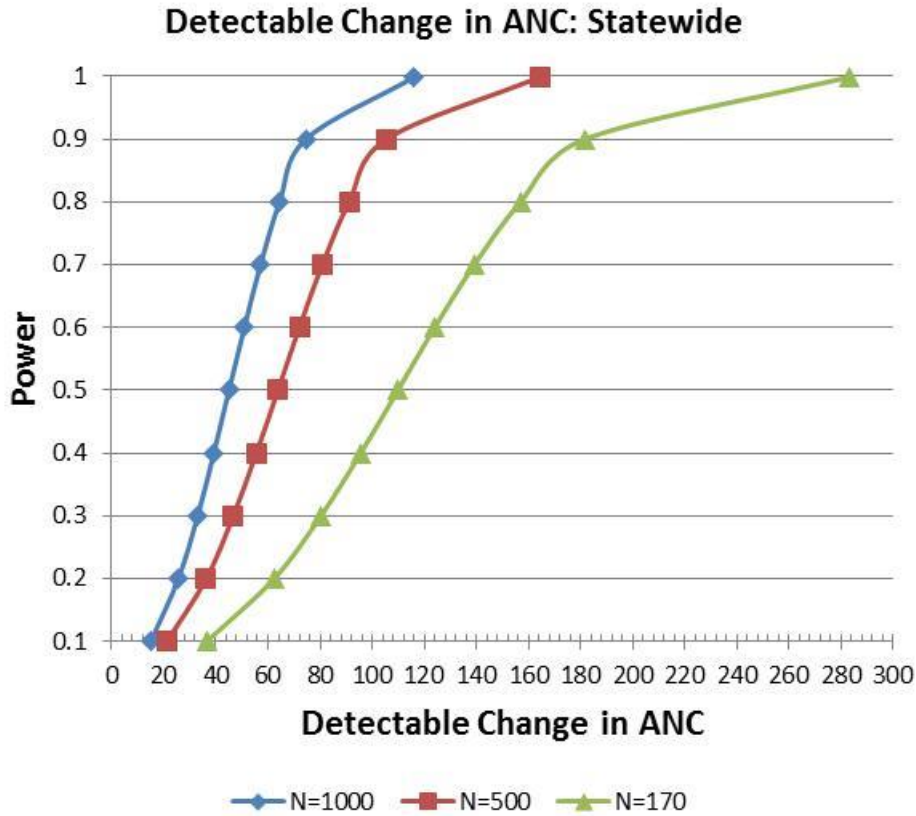


Figure B 13. The change in the ANC (µeq/L) that could be detected at the statewide level given a range of statistical power (0.1 to 1.0), and given the mean and standard deviation from Round 1 of sampling, if a sample size of 1000, 500, or 170 were used.

B3.2 Ecoregion

Table B8. Ecoregion ANC summary statistics from Round 1 MBSS					
Ecoregion	Mean	StdDev	SE	RSE (%)	N
COASTAL	397.35	396.43	22.30	5.61	316
EPIEDMONT	670.36	497.65	29.58	4.41	283
HIGHLAND	694.70	1017.09	54.06	7.78	354

B3.2.1 Detecting a Difference from 200 $\mu\text{eq/L}$ at the Ecoregion Level

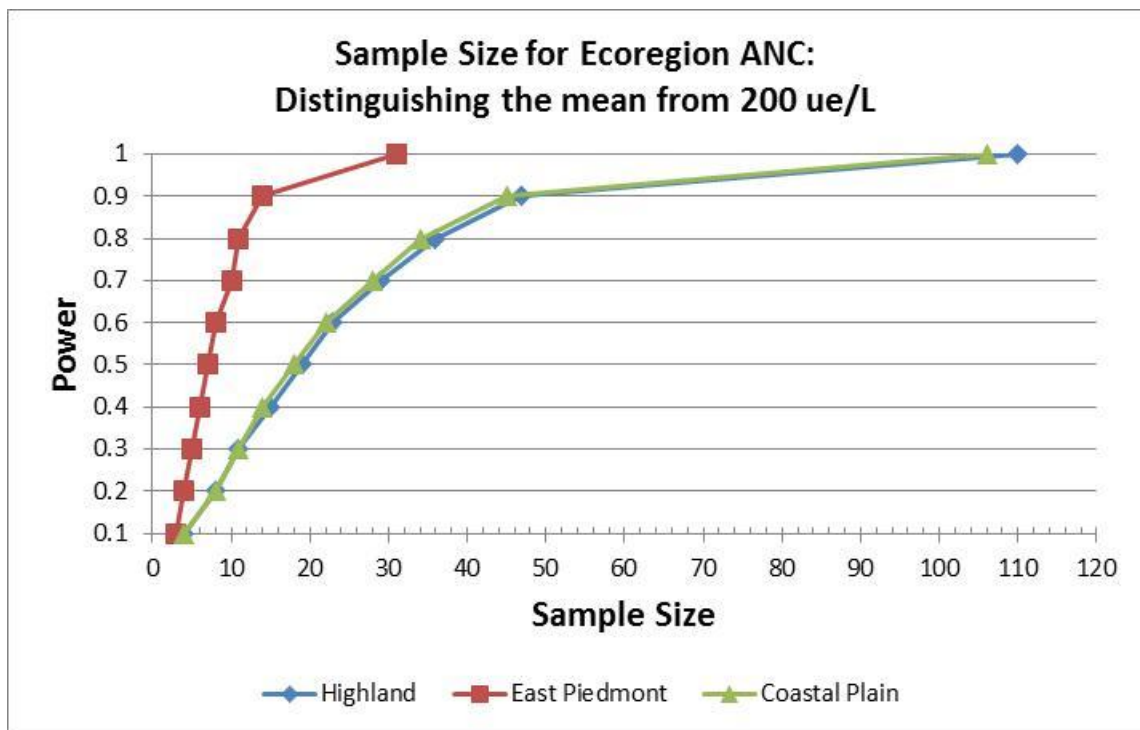


Figure B14. Sample size required to detect a significant difference from 200 $\mu\text{eq/L}$ ANC at the ecoregion level, given the means and standard deviations for each ecoregion from Round 1 of MBSS.

B3.2.2 Detectable Change in ANC at the Ecoregion Level Using Two Potential Sample Sizes

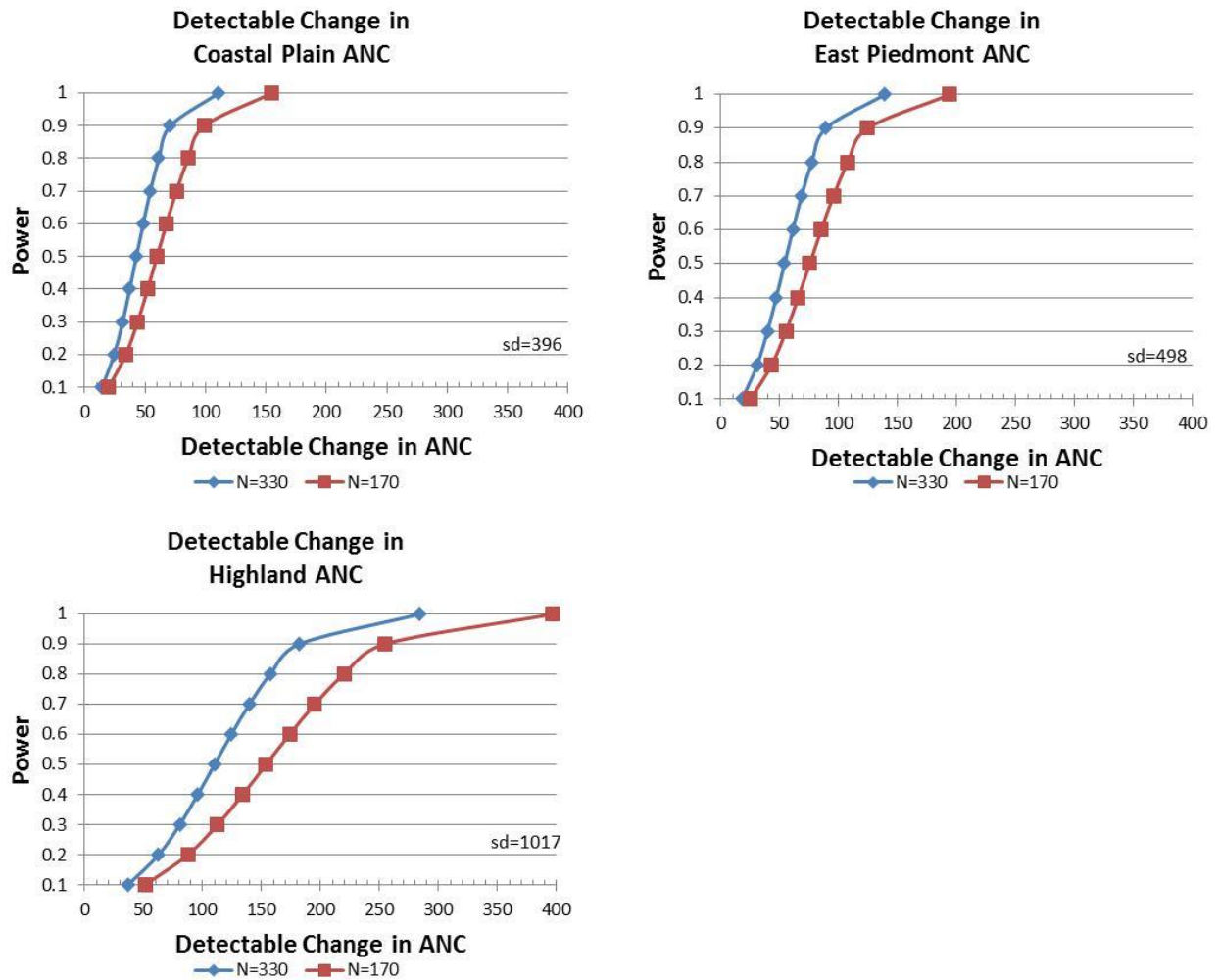


Figure B15. The detectable change in ANC ($\mu\text{eq/L}$) per ecoregion given the mean and standard deviation from Round 1 of MBSS, power=0.1 to 1.0 and sample size =330 (blue line) or 170 (red line).

B3.3 Basins

Table B 9. Basin ANC summary statistics from Round 1 MBSS					
Basin	Mean	StdDev	SE	RSE (%)	N
BU	569.2	235.9	52.74	9.27	20
CK	260.5	222.5	32.81	12.59	46
CR	575.7	366.4	56.53	9.82	42
EL	461.9	178.5	42.07	9.11	18
GU	526.5	392.1	58.45	11.10	45
LP	181.0	300.5	40.90	22.60	54
MP	868.0	672.4	64.40	7.42	109
NO	457.6	763.1	96.92	21.18	62
NW	235.3	111.4	26.25	11.16	18
PC	164.2	90.4	15.74	9.58	33
PP	841.2	620.3	54.83	6.52	128
PW	730.0	434.0	51.51	7.06	71
PX	525.3	436.8	48.24	9.18	82
SQ	469.4	215.2	35.37	7.54	37
UP	1272.8	1835.0	222.52	17.48	68
WC	430.2	225.6	38.14	8.86	35
YG	208.0	158.4	17.18	8.26	85

B3.3.1 Detecting a difference from 200 ueq/L at the basin level

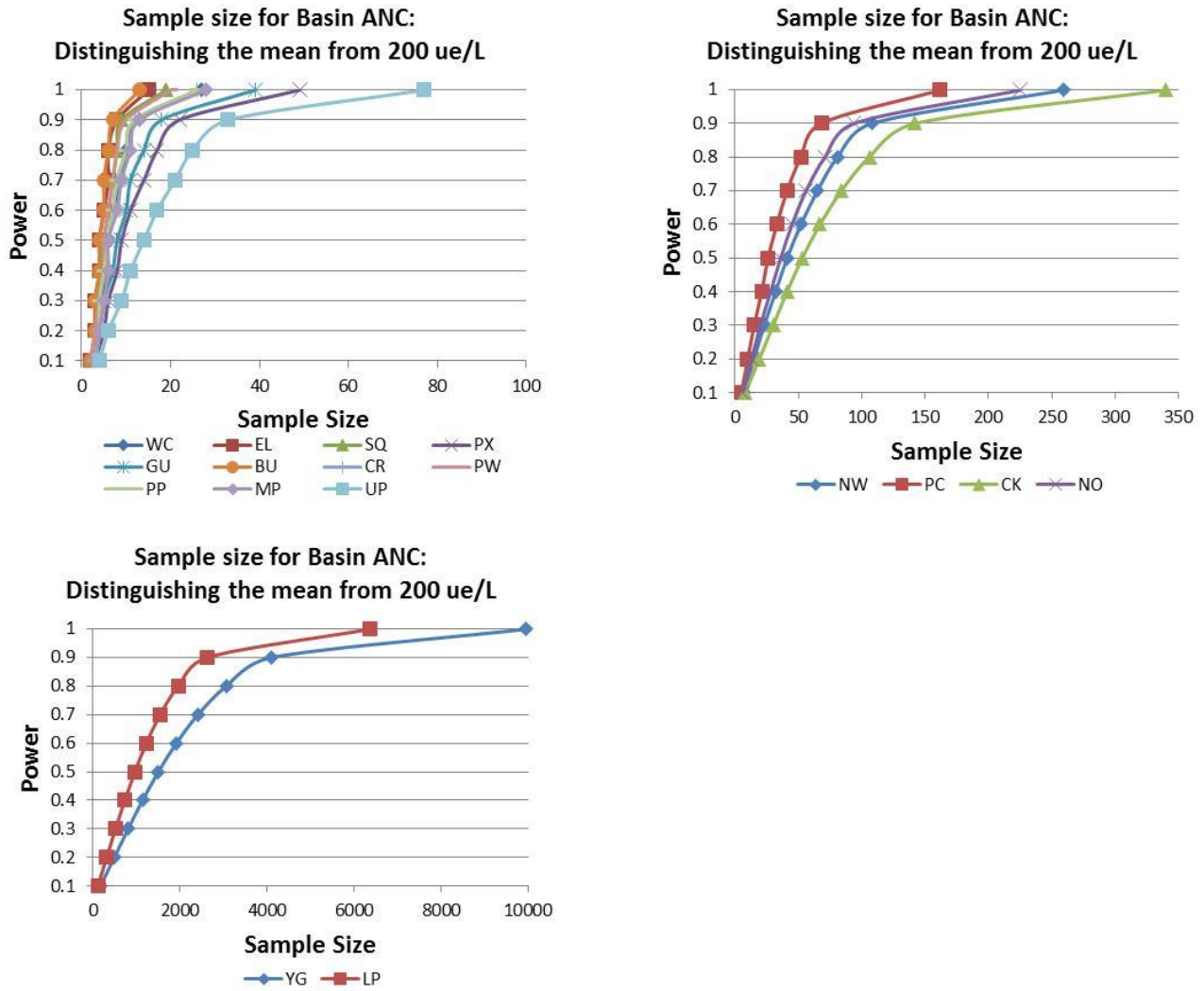


Figure B16. Sample size required to detect a significant difference from 200 ueq/L ANC at the basin level, given the means and standard deviations for each basin from Round 1 of MBSS. Individual panels depict basins with similar ranges of required sample size. Lines indicate specific basins.

B3.3.2 Detectable Change in ANC at the Basin Level Using Three Potential Sample Sizes

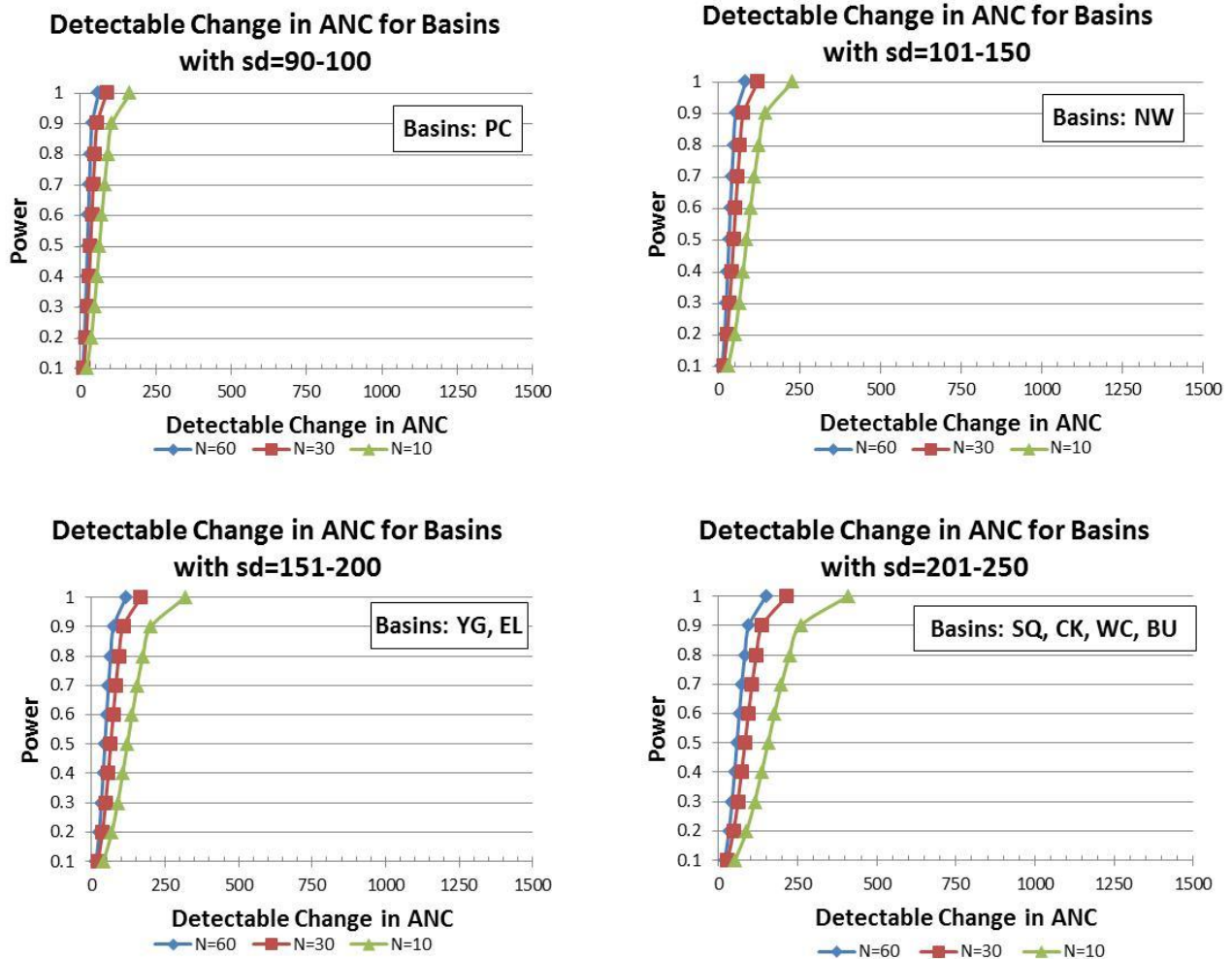


Figure B17. The detectable change in ANC ($\mu\text{eq/L}$) per basin given the mean and standard deviation from Round 1 of MBSS, power=0.1 to 1.0 and sample size =60 (blue line), 30 (red line), 10 per basin (green line). Individual panels represent basins with similar standard deviations in ANC from Round 1 of MBSS.

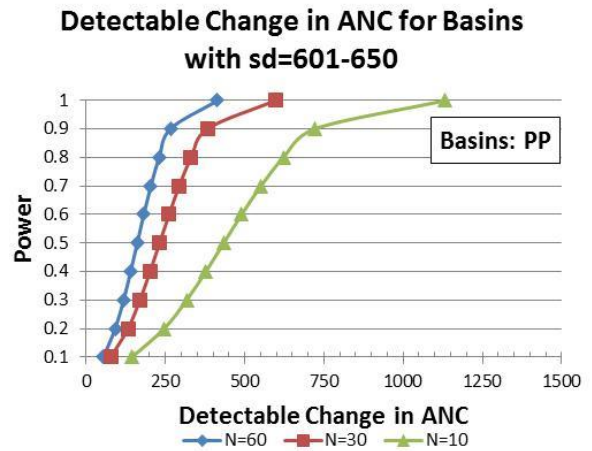
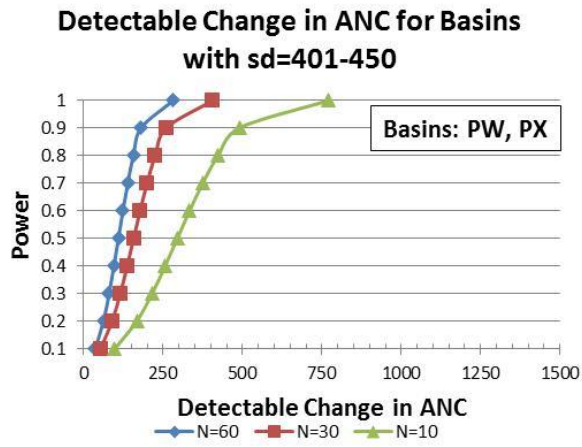
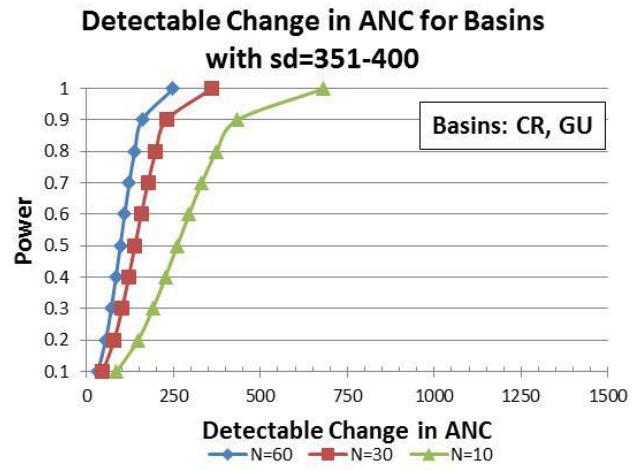
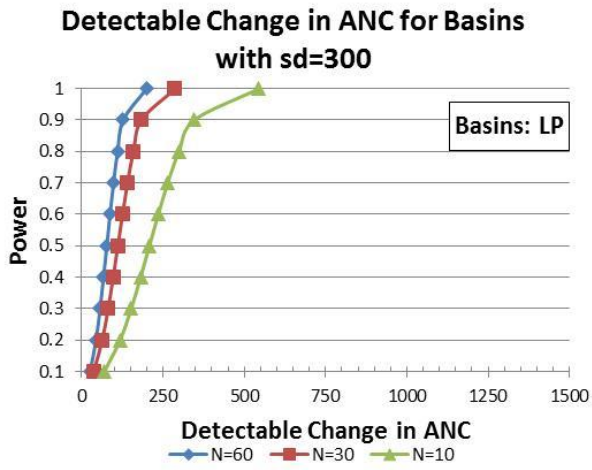
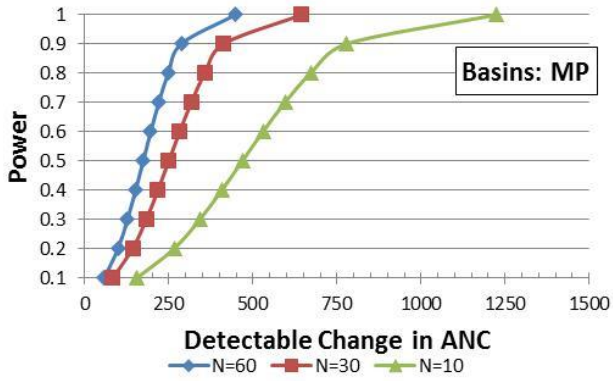
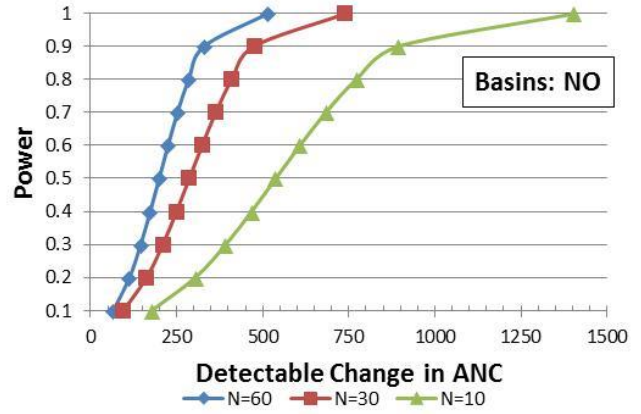


Figure B17. Continued.

Detectable Change in ANC for Basins with sd=651-700



Detectable Change in ANC for Basins with sd=751-800



Detectable Change in ANC for Basins with sd=1835

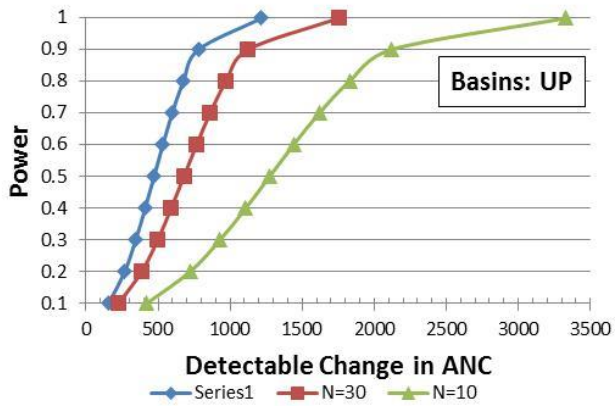


Figure B17. Continued.

B3.4 Counties

Table B10. County ANC summary statistics from Round 1 MBSS					
County	Mean	Stdev	SE	RSE	N
AA	442	275	39.2	8.9	49
AL	537	876	109.5	20.4	64
BA	868	650	73.6	8.5	78
BC	1560	725	209.2	13.4	12
CA	518	200	50.1	9.7	16
CE	544	215	40.0	7.4	29
CH	106	67	10.4	9.9	41
CN	184	76	15.9	8.6	23
CR	683	524	55.8	8.2	88
DO	164	48	21.5	13.1	5
FR	835	680	80.1	9.6	72
GA	190	167	15.6	8.2	115
HA	464	213	31.8	6.9	45
HO	691	403	62.9	9.1	41
KE	328	272	103.0	31.4	7
MO	669	440	54.6	8.2	65
PG	573	489	73.8	12.9	44
QA	548	360	54.3	9.9	44
SM	424	575	143.7	33.9	16
SO	156	177	79.3	51.0	5
TA	372	342	85.5	23.0	16
WA	1998	2058	329.6	16.5	39
WI	207	90	15.6	7.5	33
WO	131	140	57.1	43.7	6

B3.4.1 Detecting a Difference from 200 $\mu\text{eq/L}$ at the County Level

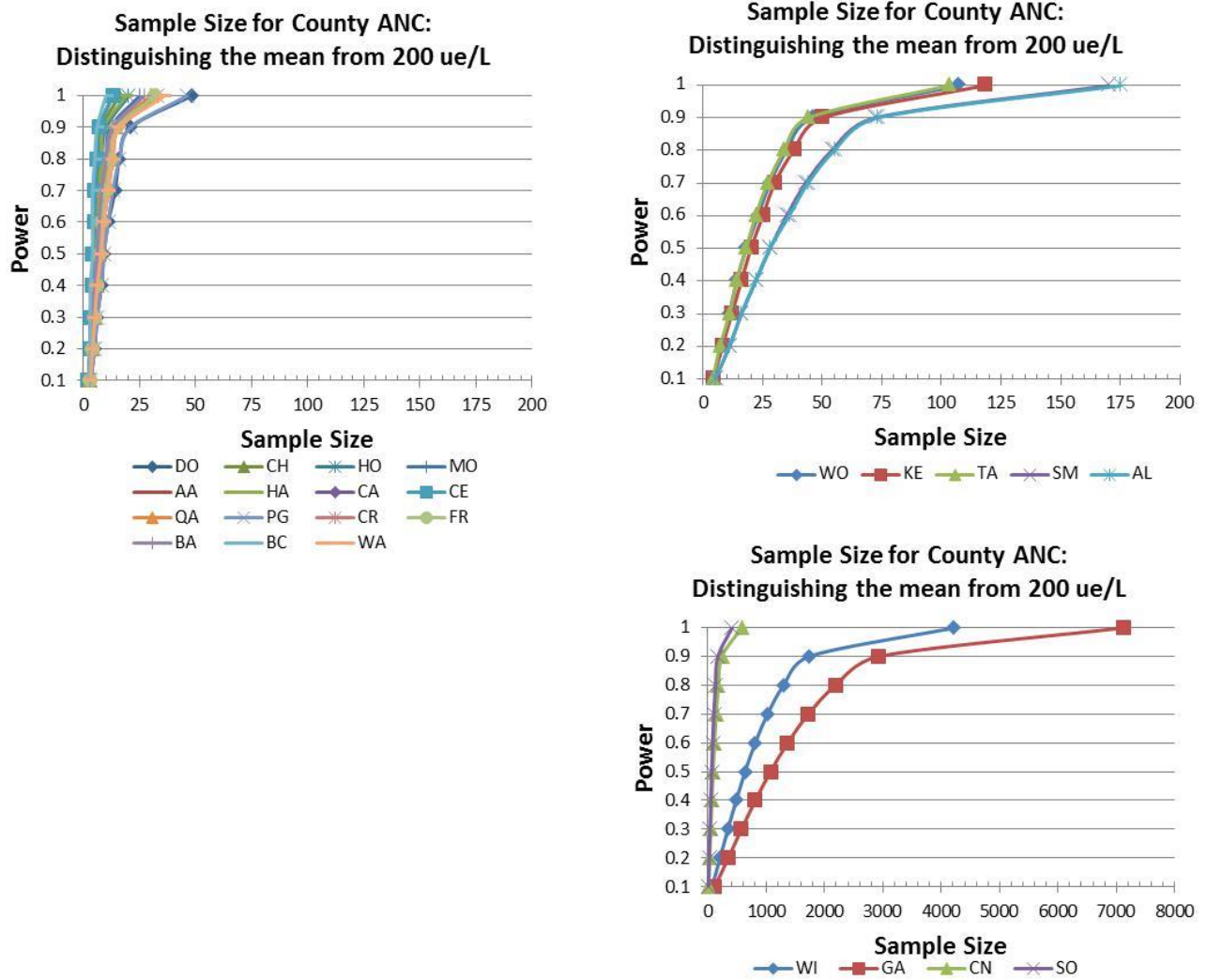


Figure B18. Sample size required to detect a significant difference from 200 $\mu\text{eq/L}$ ANC at the county level, given the means and standard deviations for each county from Round 1 of MBSS. Individual panels depict counties with similar ranges of required sample size. Lines indicate specific counties.

B3.4.2 Detectable Change in ANC at the County Level Using Three Potential Sample Sizes

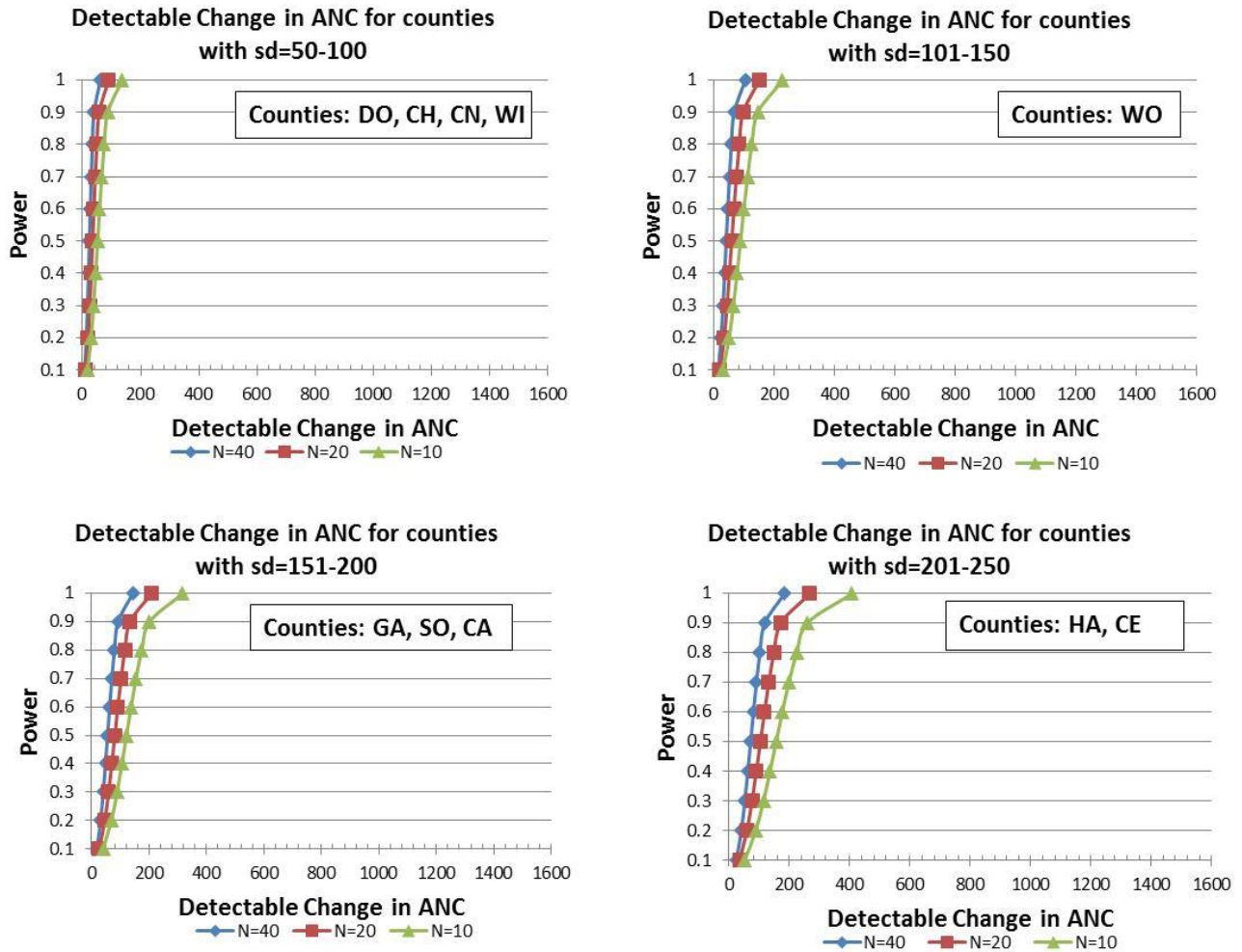


Figure B19. The detectable change in ANC ($\mu\text{eq/L}$) per county given the mean and standard deviation from Round 1 of MBSS, power = 0.1 to 1.0 and sample size = 40 (blue line), 20 (red line), 10 per basin (green line). Individual panels represent counties with the similar standard deviations in ANC from Round 1 of MBSS. Boxes indicate counties within the range for each panel.

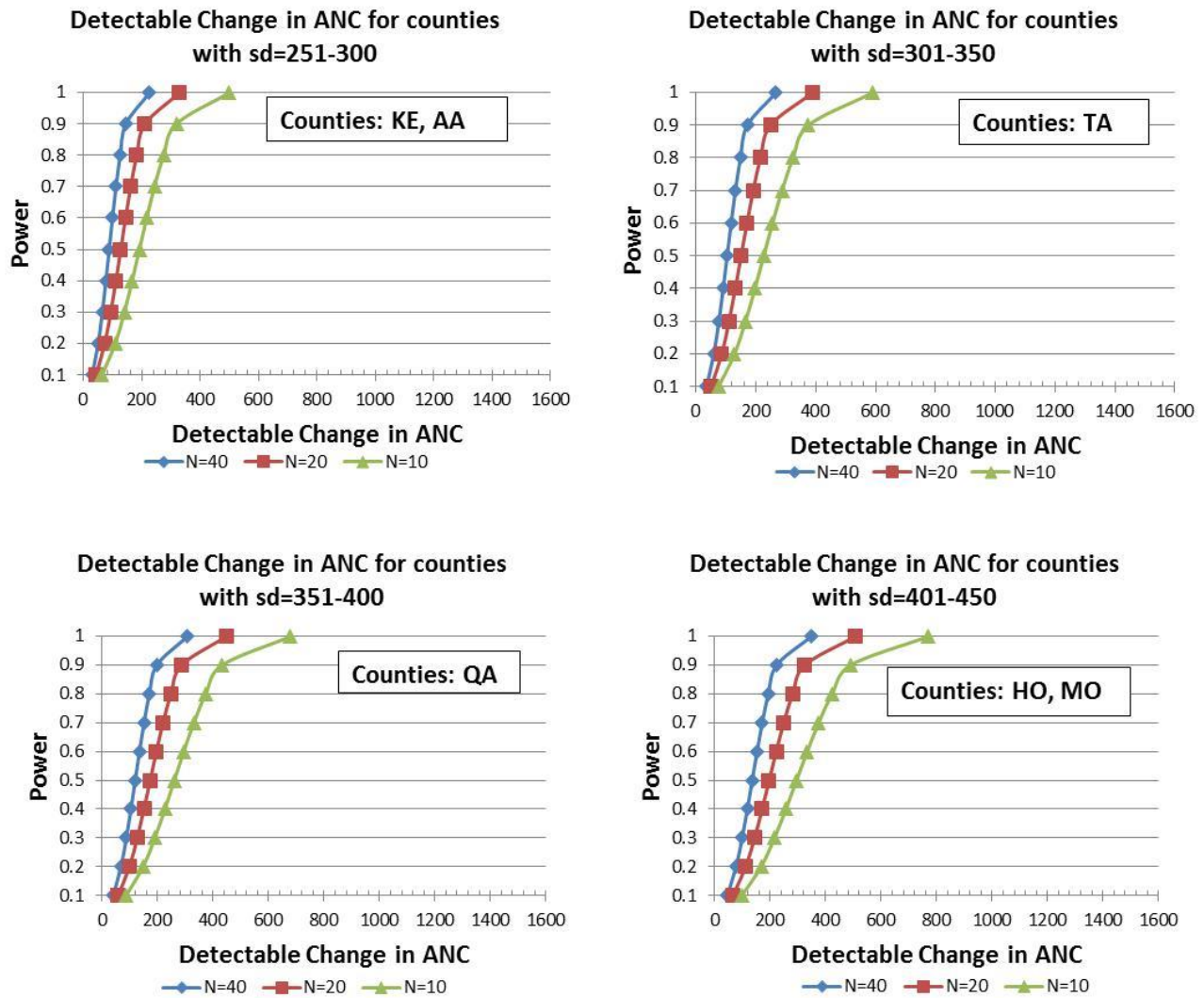


Figure B19. Continued.

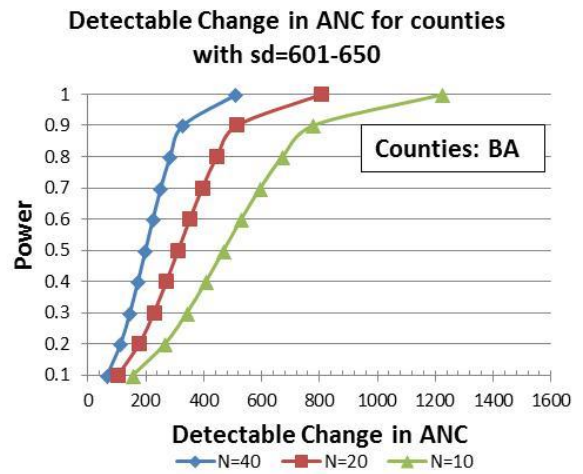
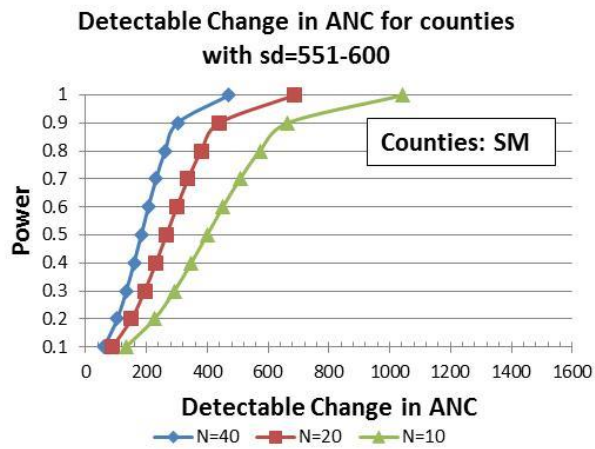
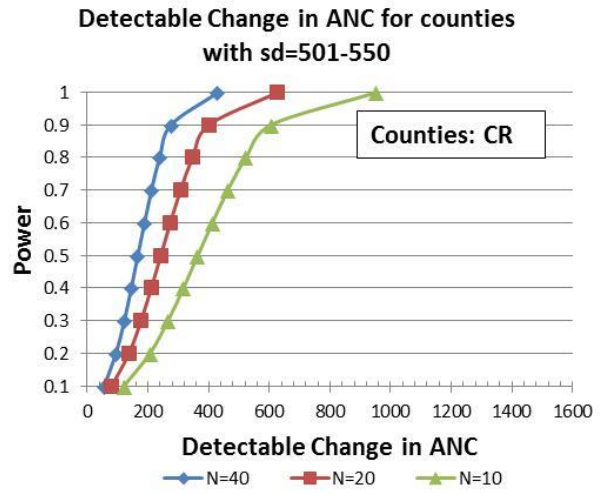
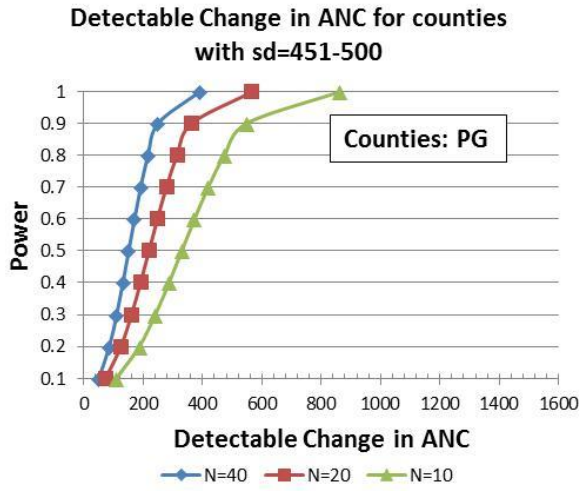


Figure B19. Continued.

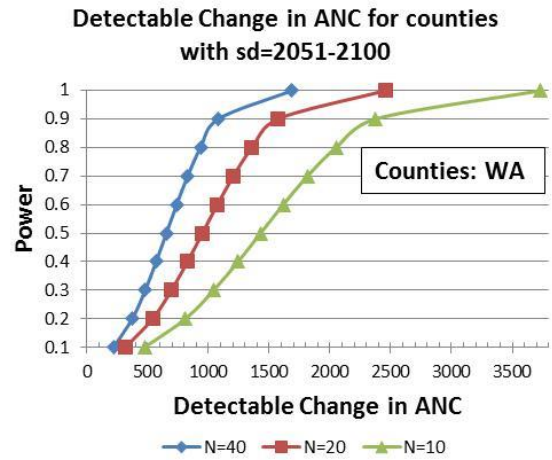
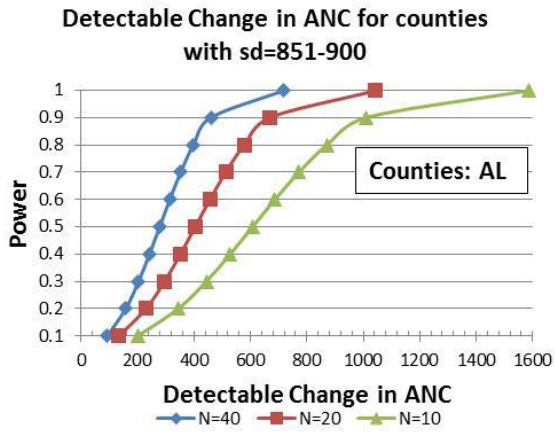
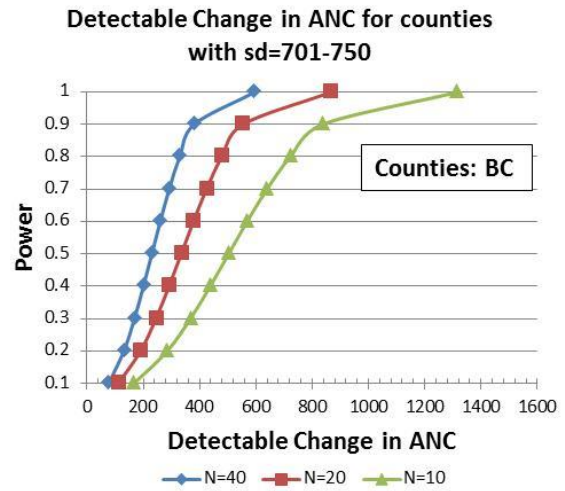
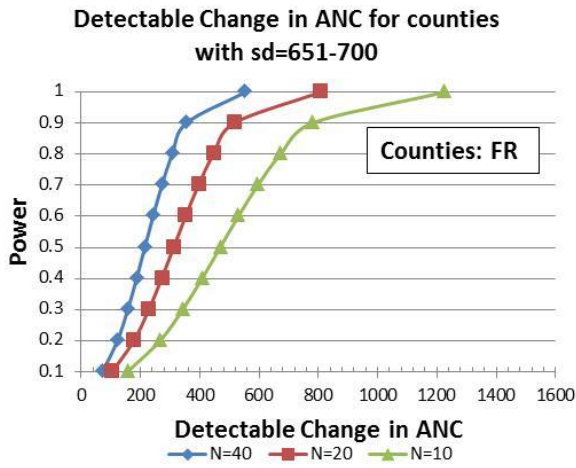


Figure B19. Continued