Age workshop for black sea bass (*Centropristis striata*)

August 18-20, 2009

SC Department of Natural Resources

Marine Resources Research Institute

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Participants:

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NCDMF	Chris Stewart
FLFWC	Janet Tunnell
SCDNR/MARMAP	Joseph Ballenger, Byron White, Paulette Mikell, Jessica Stephen, Sarah Goldman, Marcel Reichert, Bryan Danson, Gregg Swanson
GADNR	Eric Robillard
SEDAR	Dale Theiling

Introduction:

Black sea bass (BSB) from the US South Atlantic will undergo an update assessment in 2010 that will include data through 2009. South Carolina Department of Natural Resources (SCDNR/MARMAP) has primarily provided age data for previous assessments. National Marine Fisheries Service (NMFS) Beaufort Laboratory and North Carolina Division of Marine Fisheries (NCDMF) will also be providing BSB age data for the update assessment. Georgia Department of Natural Resources (GADNR) and Florida Fish and Wildlife Commission (FLFWC) will begin aging BSB for future stock assessments, but will not provide data for the update assessment in 2010. With the increase in age data providers and a change in SCDNR/MARMAP personnel responsible for aging BSB, the consistency in age analysis between labs becomes jeopardized. This workshop will address this issue as well as provide improved merged data sets for the assessment. Topics discussed during this age workshop were methodology for preparing samples for aging, interpretation of the otolith macro-structure, and conversion of increment counts to calendar ages. We will exchange otoliths for inter-lab calibration, will calculate APE and will produce an aging error matrix for the assessment model. This report will serve as a working paper for the 2010 SEDAR updates.

Methodology:

Because the sagittal otoliths of BSB are relatively thin and show a distinct alternating pattern of translucent and opaque zones, SCDNR/MARMAP, NMFS and NCDMF age BSB by viewing whole otoliths immersed in water (Figure 1). FLFWC sections all otoliths, but may convert to reading otoliths whole. As the fish gets older, the otolith thickens and becomes more opaque, thus making the increments on the margin more difficult to count (Mercer 1978). Participants of this workshop have agreed to verify increment counts using sections with those otoliths having six or more increments. Also discussed was the preferred lighting for viewing the otolith with the microscope. The choice of light source, transmitted or reflected, did have some effect on the appearance of the core area. NCDMF, NMFS, and some from SCDNR/MARMAP use reflected light while reading. Participants agreed to consider using both reflected and transmitted lighting while viewing the structure.

In order to assign each fish to the correct year class, one must record the amount of translucent material at the edge, or margin, of the otolith along with the increment count. The three labs currently aging BSB are using the same edge codes implemented by SCDNR/MARMAP and used in age studies of other reef fish:

1 Opaque zone on the otolith edge

2 Small translucent zone on otolith edge equivalent to <30% of the previous translucent zone

3 Moderate translucent zone on otolith edge equivalent to 30-60% of the previous translucent zone

4 Wide translucent zone on otolith edge equivalent to >60% of the previous translucent zone

If any amount of an opaque zone appears on the edge of the otolith, the reader will count it as an increment. The frequency plot of edge types by month has been supplied by SCDNR/MARMAP based on the age data they have already collected (n =21,005; Figure 2). Opaque zones began forming as early as January, with the most prevalent month of opaque zone formation occurring in March. By April, translucent zones were forming once again, though a large portion of otoliths still possessed opaque zones on the edge (edge type 1). By May, significant translucent zones were forming along the otolith edge. Thus, based on these observations, increment counts were converted to calendar age by advancing the increment count by one if the specimen was collected between January 1 and April 30 (the month of increment formation) and having a wide translucent zone (an edge type of 3 or 4). Ages for the remaining specimens were equal to the original increment counts.

To determine the theoretical growth of BSB based on observed size-at-age data, the calendar age for a fish will be converted to a fractional age based on peak spawning (April), a theoretical birth date (April 1), and the month of capture of that fish. The observed size-at-fractional age will be used in the theoretical growth model.

For any given otolith, if a reader deemed it un-ageable, the reader did not assign an age to the specimen and the reader gave it an A quality. In further inter-lab calibrations, we excluded these specimens.

Inter-Lab Calibration:

NCDMF provided whole (n = 205) and sectioned (n = 141) BSB otoliths to NMFS prior to this age workshop. The two primary NMFS BSB age readers examined the samples and recorded increment count and edge type for each sample. NMFS personnel brought this set of otoliths to the meeting, and workshop participants chose 100 whole otoliths from the set for the primary age readers from the other labs to read during this meeting. From this initial sample, an average percent error (APE) of 7.46% was calculated among the seven readers (two from NMFS, two from SCDNR/MARMAP, one from FLFWC, one from GADNR, and one from NCDMF) participating in the workshop (Table 1). Within-lab, the APE was 2.62% and 8.38% for SCDNR/MARMAP and NMFS, respectively.

Prior to the workshop, Chris Stewart (NCDMF) was the only reader with experience aging BSB. Assignment of the first increment was the largest controversy; the group will investigate a definitive way of assigning the first increment. One reader said he would only count the first opaque zone closest to the core if there were clear separation of the core to that first opaque zone by a translucent zone. Bryan Danson of SCDNR introduced the topic of using an acceptable range of radial measurement from the core to the first increment as a guide to accepting or rejecting an opaque zone as the first increment. He indicated a former SCDNR/MARMAP researcher had established the following criteria for counting the first opaque zone:

<0.87mm – the mark (opaque zone) is not an increment

0.87 – 1.30 mm – the mark (opaque zone) becomes a judgment call

>1.30 – the mark (opaque zone) is an increment

Subsequently, images of whole BSB otoliths were projected on a screen, from which increment placement and agreement was discussed. We then took a measurement from the core to the first increment with the ocular micrometer. These preliminary measurements were similar to those established by the former SCDNR/MARMAP researcher.

An additional method of defining the first increment includes measuring from the core to edge (margin) of young of the year (y-o-y) BSB otoliths (Wenner et al. 1986). Once we have calculated the marginal measurements of y-o-y BSB, they will be compared to the measurements taken from the first increment to the core on the adult fish.

Beyond the concern with the interpretation of the first annulus, several readers pointed out that there appears to be a check mark on some of the otoliths. They appeared as very thin, faint or incomplete opaque zones that occurred primarily between the third and fourth increment or the second and third increment. This check mark coincides with the timing of transition of BSB from females to males (McGovern et.al. 2002). If we knew the sex of the fish, we could look for a correlation between the occurrence of the check mark and the sex of the fish. Researchers need to investigate this issue further.

Eric Robillard of GADNR brought up the issue of age readings from whole versus sectioned otoliths. Because of this issue with red porgy and some other coastal fish species, the group felt that we should address this question by first reading a set of otoliths whole, then sectioning them and reading them again. For long-term analysis, we suggested the use of samples that have pairs of otoliths so the section and whole otolith could be maintained together for future calibration work. When selecting which otolith of the pair to section, the group decided to alternate sectioning the right then the left of each pair to avoid any potential bias.

After the conclusion of the workshop, we circulated two additional calibration sets among the labs participating in the workshop. The first, a set of 100 otoliths provided by SCDNR/MARMAP, was a set of randomly selected otoliths from the MARMAP survey for whole otolith to sectioned otolith comparison as well as inter-laboratory calibration of age readings. The second, a NMFS randomly selected 100 whole otoliths from all fisheries and all states, was used in the inter-laboratory calibration. This brought the total number of otoliths used in the final inter-laboratory calibration to 300 otoliths (100 each from NCDMF, SCDNR/MARMAP, and NMFS) and 100 otoliths used in the investigation of aging precision between whole and sectioned otoliths.

For the final inter-calibration study amongst laboratories and the whole versus sectioned otolith aging comparison study, SCDNR/MARMAP added an additional two age readers, as they will be aging fish for the upcoming SEDAR BSB update in 2010. These two readers aged the 100 whole otoliths from NMFS and the 100 whole and sectioned otoliths from SCDNR/MARMAP, meaning they aged 200 individuals used in the inter-laboratory calibration study and all individuals used in the whole versus sectioned otolith aging study.

When the additional 200 otoliths are added to the 100 otoliths aged at the workshop for the inter-laboratory calibration study, the APE decreases slightly overall to 7.38% (Table 1). Within-lab, the APE was 4.07% and 6.81% for SCDNR/MARMAP and NMFS, respectively.

We further investigated the bias and precision of age readings among individual readers using a parametric paired t-test and a simple linear regression (Table 2,

Figure 3 and

Figure 4). The resulting slopes of the linear regressions we also tested to detect systematic differences between readers. However, these tests are ineffective at detecting differences when one reader underages an otolith at one end of the age range and then overages at the other end compared to another reader (Campana et al. 19955). In addition, these tests cannot detect non-linear differences among readers when the differences are centered about the 1:1 line, thus we used bias plots (

Figure 4) to visually ascertain the potential for this problem among readers (Campana et al. 1995). Given the inherent limitations associated with paired t-tests and simple linear regressions, we also used Bowker's symmetry test to detect bias among readers. In previous research, Hoenig et al. (1995) note that Bowker's symmetry test is a more robust statistical test for detecting biases. To determine the level of precision between readers, three measures of precision were calculated: mean coefficient of variation (CV), percent agreement, and APE (Campana et al. 1995).

In general, the bias tests indicated that bias was relatively small among most readers compared (

Table 2,

Figure **3** and

Figure **4**). Overall, out of the 36 possible pairwise comparisons among the nine readers, 29 passed at least one statistical test trying to detect bias (

Table 2). While no pairwise comparison passed all three statistical tests, if we exclude the linear regression test, as it seemed to indicate biases more often than the other two tests, 19 of 36 possible pairwise comparisons passed both the pair t-test and Bowker's symmetry test. The most significant problem appears to be that NMFS Reader 2 is systematically underaging BSB compared to other readers (Figure 3 and 4). Of the seven pairwise comparisons that failed all statistical tests of bias, NMFS Reader 2 was involved in five (

Table 2).

With regards to precision between readers, mean CV ranged from 3.7-10.6%, percent agreement ranged from 54.6%-84.0%, and APE ranged from 2.7-7.5% (Table 2**).** Based on the group decided acceptable APE of 5% or less, assuming no bias between the readings, 15 of 36 pairwise comparisons passed. Further, if you consider an APE of 6.5% or less acceptable, 32 of 36 pairwise comparisons passed. When problems arose, most often they were due to the underaging of BSB by NMFS Reader 2 compared to the other readers (

Table 2).

Sectioned vs. Whole Otolith Study

Based on the results of the whole otolith versus sectioned otolith comparison study, it appears that in general the mean APE is reduced when BSB are aged via sectioned otoliths (Table 3). On average, there is approximately a 50% reduction in mean APE between the two aging methods. However, the average difference in ages between BSB aged whole and aged via sections is only approximately 0.12 years (Table 3). Further, even at the maximum age that can possibly be aged whole (6 years, based on criteria determined during this workshop), the average difference in ages between the two methods would still be less than 0.5 years.

In addition, the bias tests within a reader comparing the two aging methodologies indicate that the bias is relatively small (Table 4, Figure 5 and Figure 6). Overall, for the nine different readers, two passed all bias tests, three passed two tests, and all passed at least one (Table 4). Further, upon visual inspection of Figure 5, the only general problem appears to be a slight overaging of BSB at young ages via sectioned otoliths when compared to whole otoliths. This problem likely arises because of the difficulty of determining the first annulus in sectioned otoliths. Further inspection of the bias plots (Figure 6) also indicated no significant biases between aging methods within a given reader.

Precision between aging methods within a reader indicated that mean CV ranged from 4.6-12.5%, percent agreement ranged from 60.0-82.1%, and APE ranged from 3.3-8.8% (Table 4). APE was less than 6% for all readers except one. The high APE for South Carolina Reader 2 appears to be due to a problem with overaging sectioned BSB otoliths compared to whole otolith age, especially for fish three years of age and younger, indicating a difference in core interpretation between the aging methodologies for this reader.

Follow-up Work:

MARMAP's existing BSB age data (n ≈ 18,000) includes increment counts only. As these fish were aged prior to the implementation of the edge codes now used, MARMAP will review those specimens collected from January through April to establish the edge type.

In an effort to resolve the inconsistencies in assigning the first increment, NMFS will begin collecting y-o-y BSB in Bogue Sound, NC by use of traps and hook and line. During October and November, SEAMAP trawls will retain y-o-y BSB. A researcher at Belle Baruch Institute of the University of South Carolina has started collecting y-o-y BSB in the Winyah Bay estuary, which we may be able to analyze.

Long-term goal includes a formal compilation of workshop reports and reference collections.

Literature Cited:

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Mercer L.P. 1978. The reproductive biology and population dynamics of black sea bass, Centropristis striata. The College of William and Mary, College of Marine Science, PhD dissertation.

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Wenner C.A, W.A. Roumillat, and C.W. Waltz. 1986. Contributions to the life history of black sea bass, *Centropristis striata*, off the southeastern United States. Fishery Bulletin 84: 723-741.

Tables:

Table 1: Average percent errors (APE) for the seven primary age readers who participated in the aging workshop for three different inter-laboratory calibration sets (NCDMF, SCDNR/MARMAP, and NMFS) read by all. Two readers from both SCDNR/MARMAP and NMFS participated, thus intra-lab APE for these laboratories are also reported.

Set	Overall APE	SCDNR/MARMAP APE	NMFS APE
All combined	7.38%	4.07%	6.80%
NCDMF	7.46%	2.62%	8.38%
MARMAP	8.08%*	8.39%*	4.20%
NMFS	7.16%*	4.09%*	7.91%

*Indicates the two additional SCDNR/MARMAP age readers were included in the APE calculation.

Table 2: Results of various tests and indicators used to determine the accuracy and precision of age readings between nine different primary BSB age readers from SCDNR/MARMAP (4), NMFS (2), NCDMF (1), GADNR (1), and FLFWC (1).

		Paired t-Test	Linear Regression	Symmetry Test			
Reader 1	Reader 2	p-value	p-value	p-value	CV	% Agree.	APE
SC-1	FL	0.3541	<0.0001*	0.4363	4.34%	80.28%	3.07%
SC-1	GA	0.0289*	<0.0001*	0.5902	5.25%	77.54%	3.71%
SC-1	SC-2	0.0346*	0.0053*	0.0588	5.75%	72.45%	4.07%
SC-1	NMFS-1	0.0623	<0.0001*	0.1860	6.86%	68.66%	4.85%
SC-1	NMFS-2	<0.0001*	<0.0001*	<0.0001*	9.45%	56.80%	6.68%*
SC-1	NC	0.0109*	<0.0001*	0.1179	5.13%	77.03%	3.63%
SC-1	SC-3	0.6338	<0.0001*	0.0969	5.06%	78.97%	3.58%
SC-1	SC-4	0.1858	<0.0001*	0.1857	6.70%	70.05%	4.74%
FL	GA	0.2389	0.0011*	0.6646	5.16%	77.30%	5.32%*
FL	SC-2	0.3337	0.0503	0.0139*	5.87%	73.85%	4.15%
FL	NMFS-1	0.4361	<0.0001*	0.5430	7.19%	69.06%	5.08%*
FL	NMFS-2	<0.0001*	<0.0001*	<0.0001*	9.91%	56.34%	7.01%*
FL	NC	0.0536	0.0017*	0.1821	4.57%	80.14%	3.23%
FL	SC-3	0.2938	0.0008*	0.1572	7.54%	70.90%	5.33%*
FL	SC-4	0.2706	0.0004*	0.5606	7.01%	70.53%	4.96%
GA	SC-2	0.8723	0.0035*	0.2356	7.43%	67.61%	5.25%*
GA	NMFS-1	0.8230	<0.0001*	0.4732	8.88%	62.37%	6.28%*
GA	NMFS-2	<0.0001*	<0.0001*	<0.0001*	8.75%	61.97%	6.19%*
GA	NC	0.1661	<0.0001*	0.6698	3.74%	84.03%	2.65%
GA	SC-3	0.0007*	0.4041	0.0482*	5.91%	74.48%	4.18%
GA	SC-4	0.0016*	0.0025*	0.0140*	7.97%	67.88%	5.64%*
SC-2	NMFS-1	0.7303	<0.0001*	0.1283	7.88%	65.85%	5.58%*
SC-2	NMFS-2	<0.0001*	<0.0001*	<0.0001*	10.61%	54.61%	7.50%*
SC-2	NC	0.8621	<0.0001*	0.0645	6.40%	72.30%	4.52%
SC-2	SC-3	0.0046*	<0.0001*	0.0245*	6.79%	73.98%	4.80%
SC-2	SC-4	0.0592	<0.0001*	0.1044	5.89%	78.06%	4.16%
NMFS-1	NMFS-2	<0.0001*	<0.0001*	<0.0001*	9.62%	58.48%	6.80%*
NMFS-1	NC	0.7412	<0.0001*	0.4338	7.44%	67.72%	5.26%*
NMFS-1	SC-3	0.4057	<0.0001*	0.0944	7.35%	68.78%	5.20%*
NMFS-1	SC-4	0.4809	<0.0001*	0.4121	8.00%	65.26%	5.66%*
NMFS-2	NC	<0.0001*	0.0898	<0.0001*	8.70%	60.34%	6.15%*
NMFS-2	SC-3	0.0010*	0.0686	0.0943	7.32%	69.43%	5.17%*
NMFS-2	SC-4	0.0007*	0.0070*	0.0563	8.45%	64.62%	5.97%*
NC	SC-3	<0.0001*	0.8099	0.0048*	6.62%	73.10%	4.68%
NC	SC-4	0.0002*	0.1163	0.0025*	7.80%	69.19%	5.52%*
SC-3	SC-4	0.3718	<0.0001*	0.4285	6.14%	75.25%	4.83%*

*Indicates p-value is <0.05 or APE is >5%.

Table 3: Mean APE calculated for whole otolith and sectioned otolith readings of the SCDNR/MARMAP reference set (n=100) provided for whole otolith versus sectioned otolith comparisons. Nine readers from five agencies (1 from FLFWC, 1 from GADNR, 4 from SCDNR/MARMAP, 1 from NCDMF, and 2 from NMFS) aged all otoliths. We only included fish that were aged by all readers by both methods in the analysis

Group	N	Whole	Sectioned	Avg. Diff. (Years)	% Reduction
Overall APE	93	7.97%	3.94%	-0.12	50.56%
SCDNR/MARMAP APE	97	8.15%	3.89%	-0.16	52.27%
NMFS APE	96	3.90%	2.71%	-0.12	30.51%

Table 4: Results of various tests and indicators used to determine the accuracy and precision of age readings between whole otolith and sectioned otolith readings. Nine different primary BSB age readers from SCDNR/MARMAP (4), NMFS (2), NCDMF (1), GADNR (1), and FLFWC (1) participated.

	Paired t-test	Linear Regression	Symmetry Test			
Reader	p-value	p-value	p-value	CV	% Agree	APE
South Carolina R1	0.5743	0.0986	0.2053	7.90%	71.13%	5.59%*
Florida	0.6825	<0.0001*	0.6419	7.63%	77.08%	5.39%*
Georgia	0.8361	0.0028*	0.4151	4.63%	82.11%	3.28%
South Carolina R2	0.0085*	<0.0001*	0.2135	12.46%	60.00%	8.81%*
NMFS Reader 1	0.0001*	0.2949	0.0023*	8.47%	69.79%	5.99%*
NMFS Reader 2	0.7870	<0.0001*	0.3950	7.34%	73.74%	5.19%*
North Carolina	0.0024*	0.2188	0.0120*	5.86%	80.00%	4.14%
South Carolina R3	0.0116*	0.2135	0.2231	5.14%	78.79%	3.63%
South Carolina R4	0.1156	0.0002*	0.5511	8.48%	72.00%	5.99%*

*Indicates p-value is <0.05 or APE is >5%.

Figures:

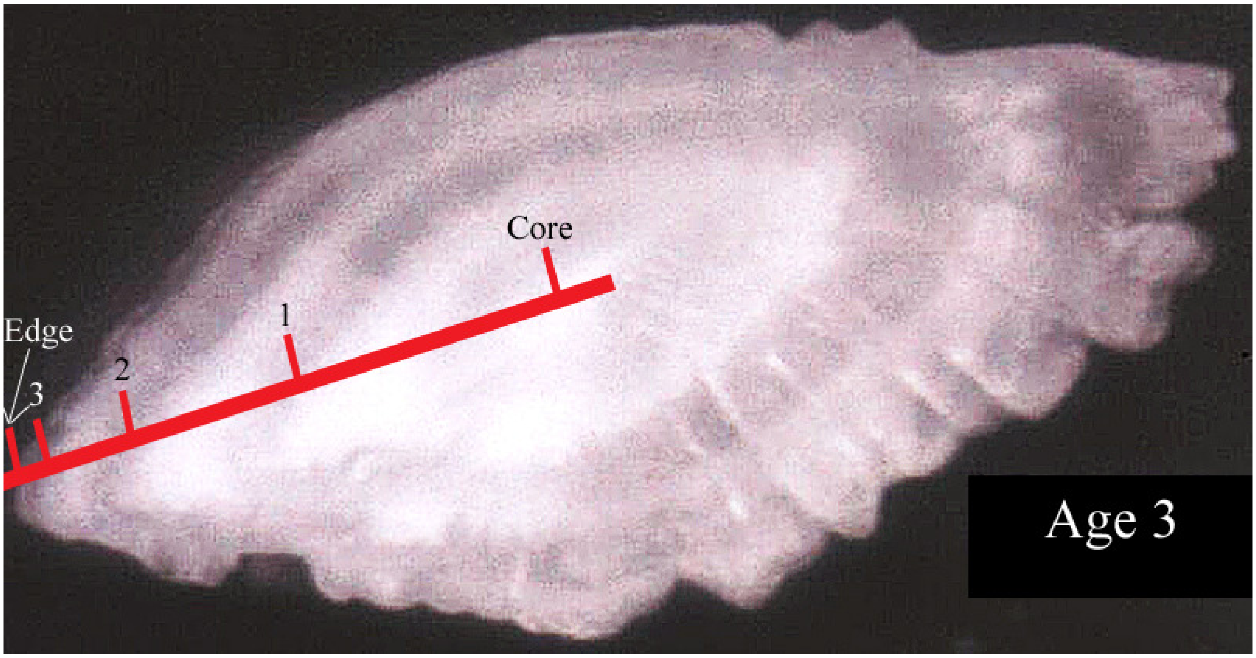


Figure 1: Whole BSB otolith aged under reflected light. Picture is of a three-year-old specimen.

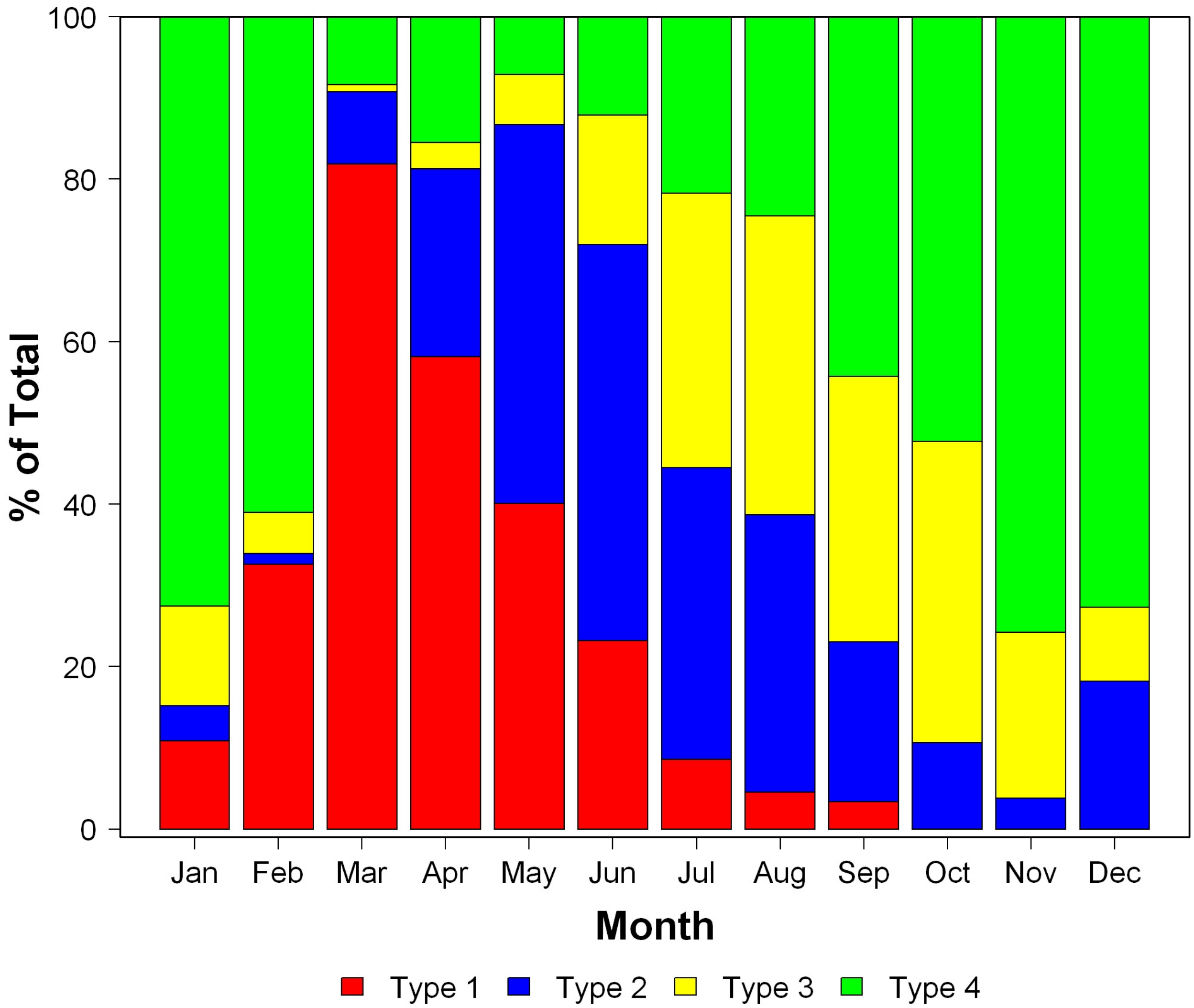
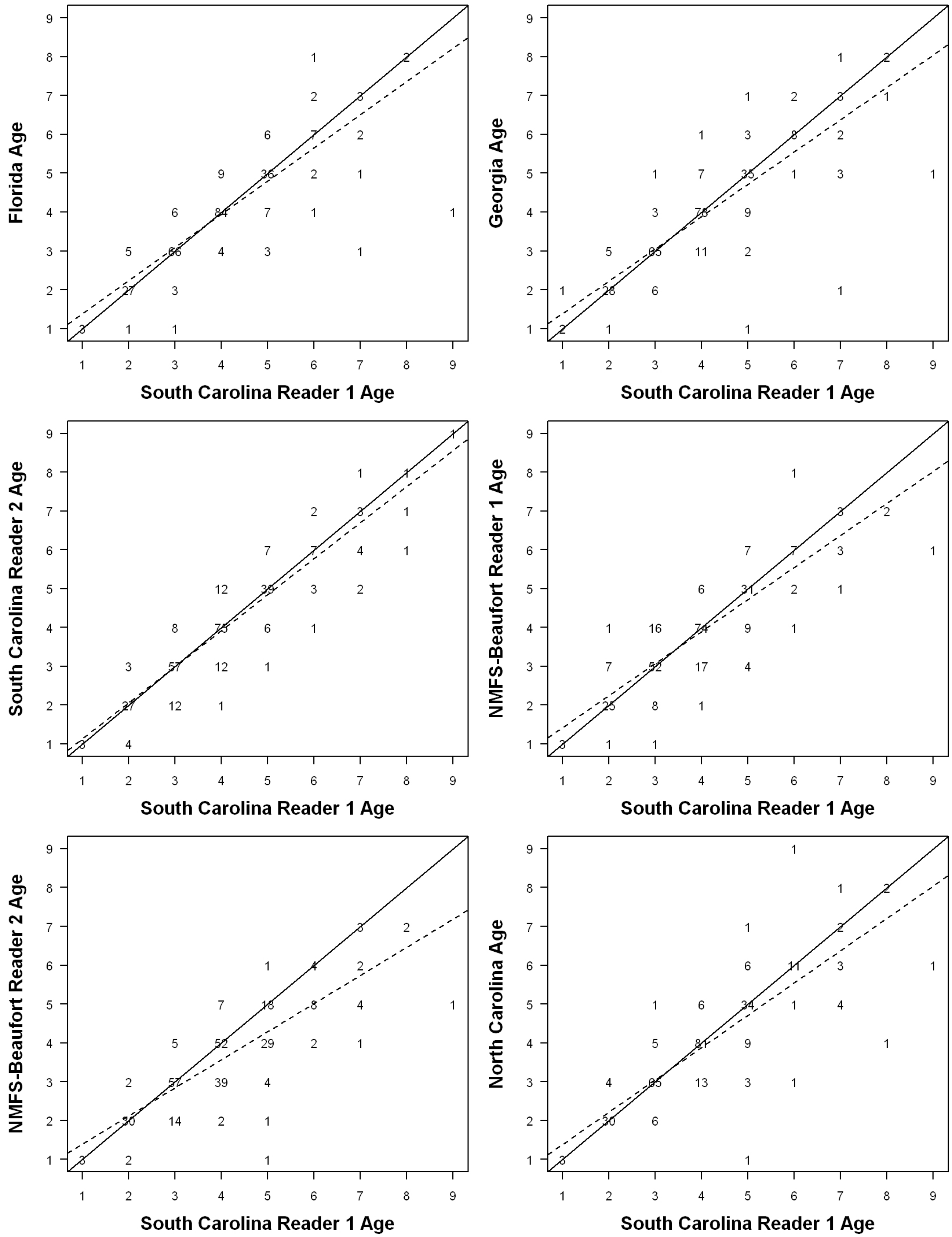
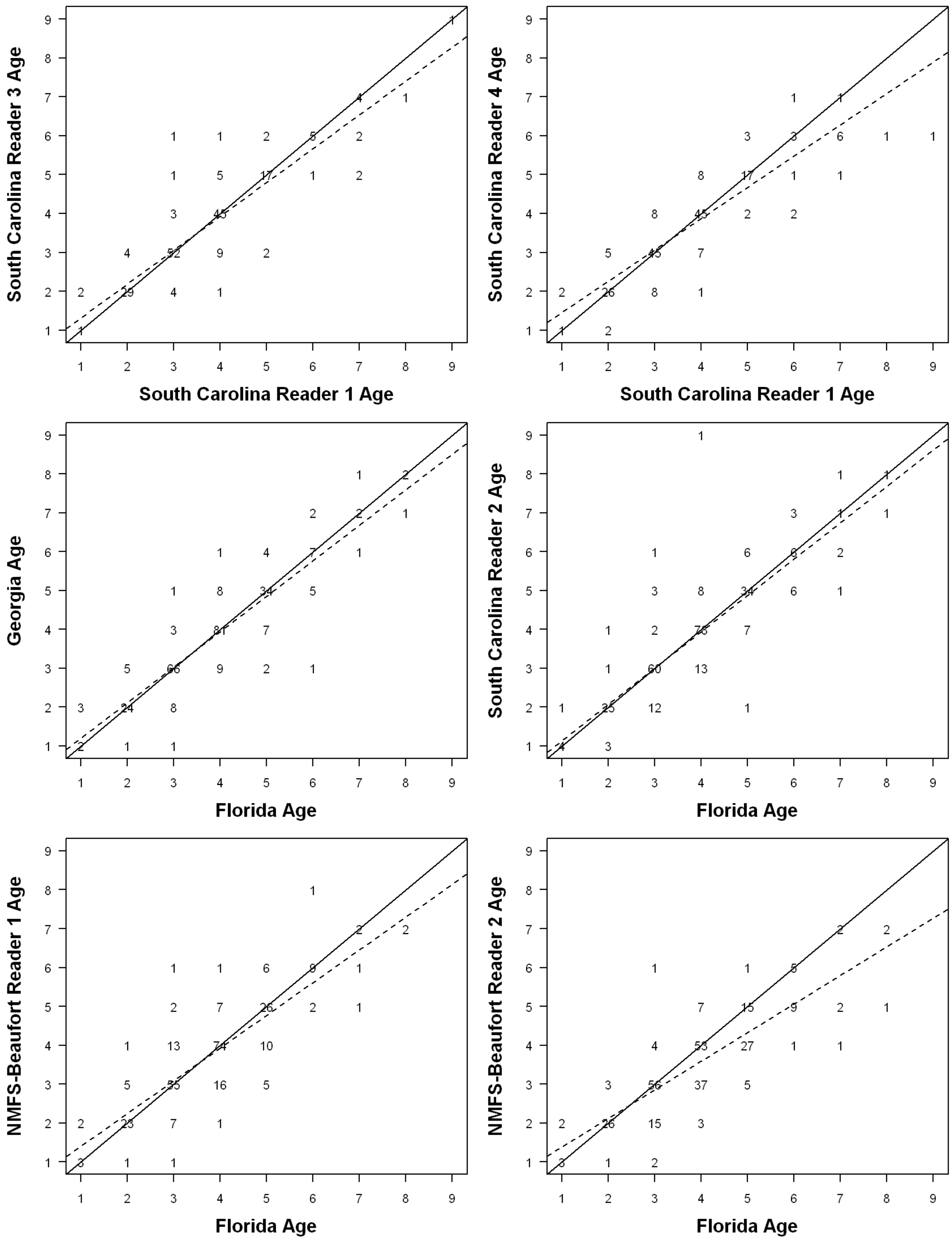


Figure 2: Black sea bass edge type distribution by month for individuals aged (n=21,005) by SCDNR/MARMAP.

Figure 3: Plots of linear regressions among individual primary age readers of BSB for upcoming SEDAR BSB update. Numbers represent the number of fish assigned that combination of ages by the readers. Solid line indicates perfect agreement (1:1 line) between readers. Dashed line indicates the calculated linear regression between age readings of the different readers.

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South Carolina Reader 1 Age
South Carolina Reader 4 Age
South Carolina Reader 1 Age
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South Carolina Reader 2 Age
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Florida Age
NMFS-Beaufort Reader 2 Age
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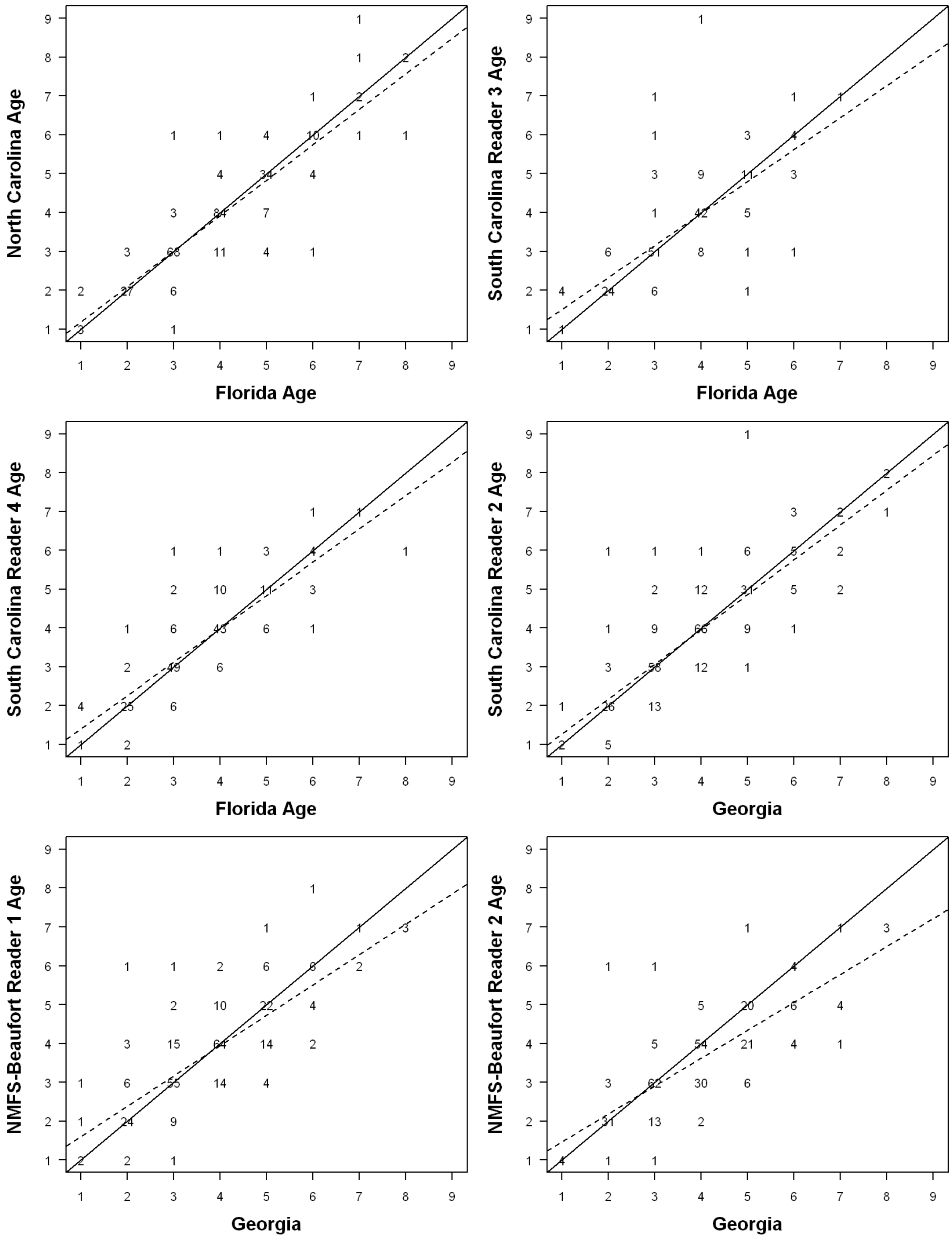
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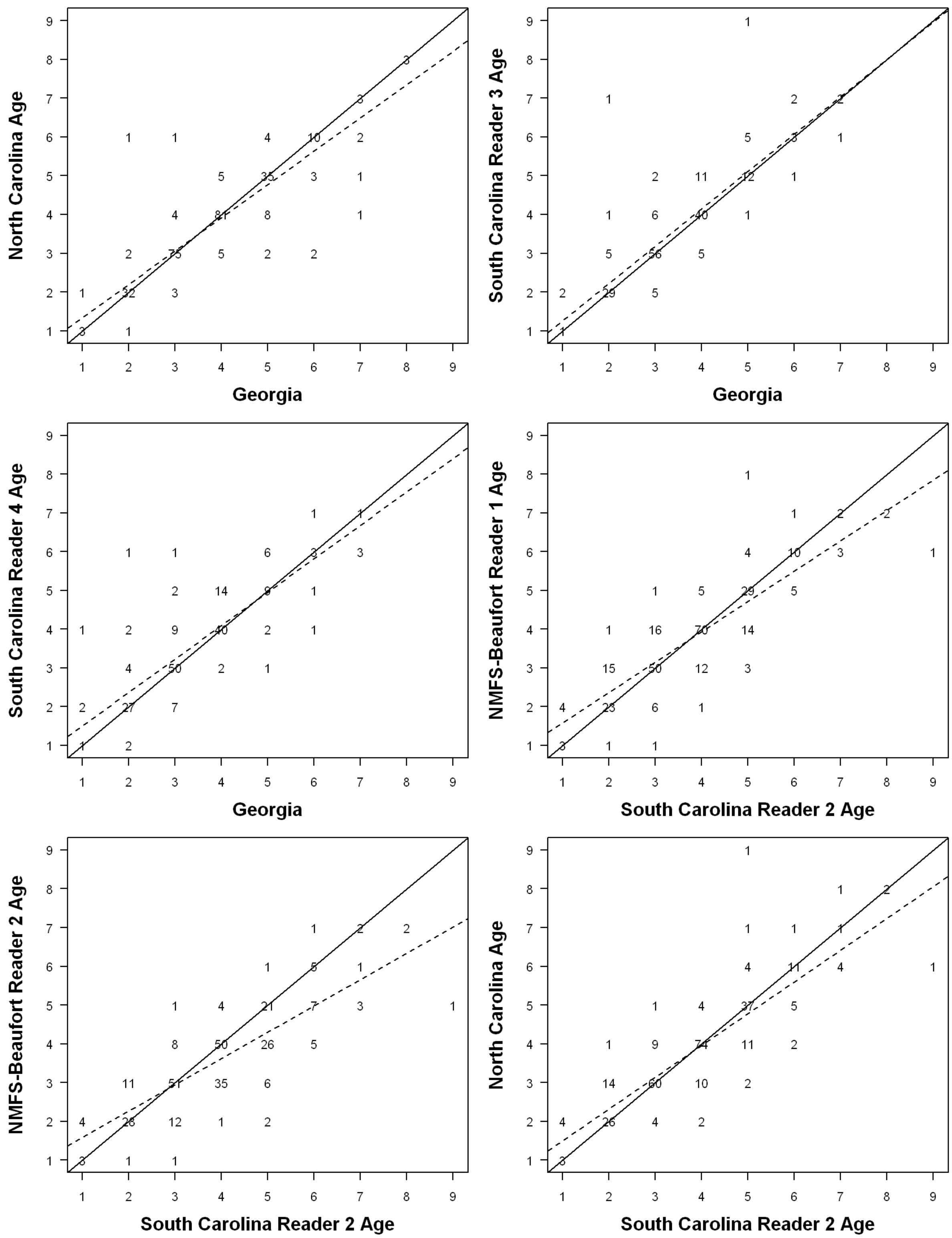
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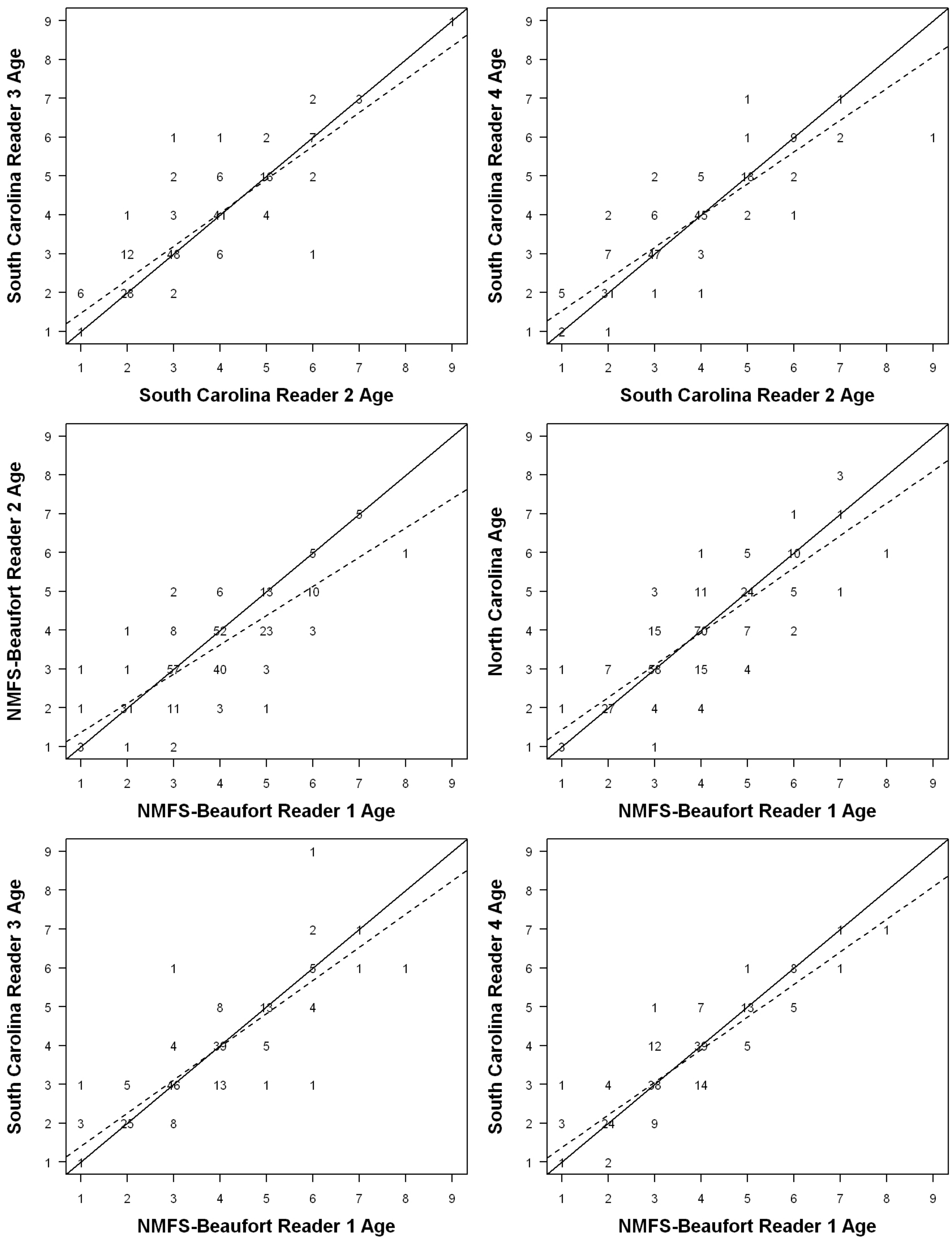
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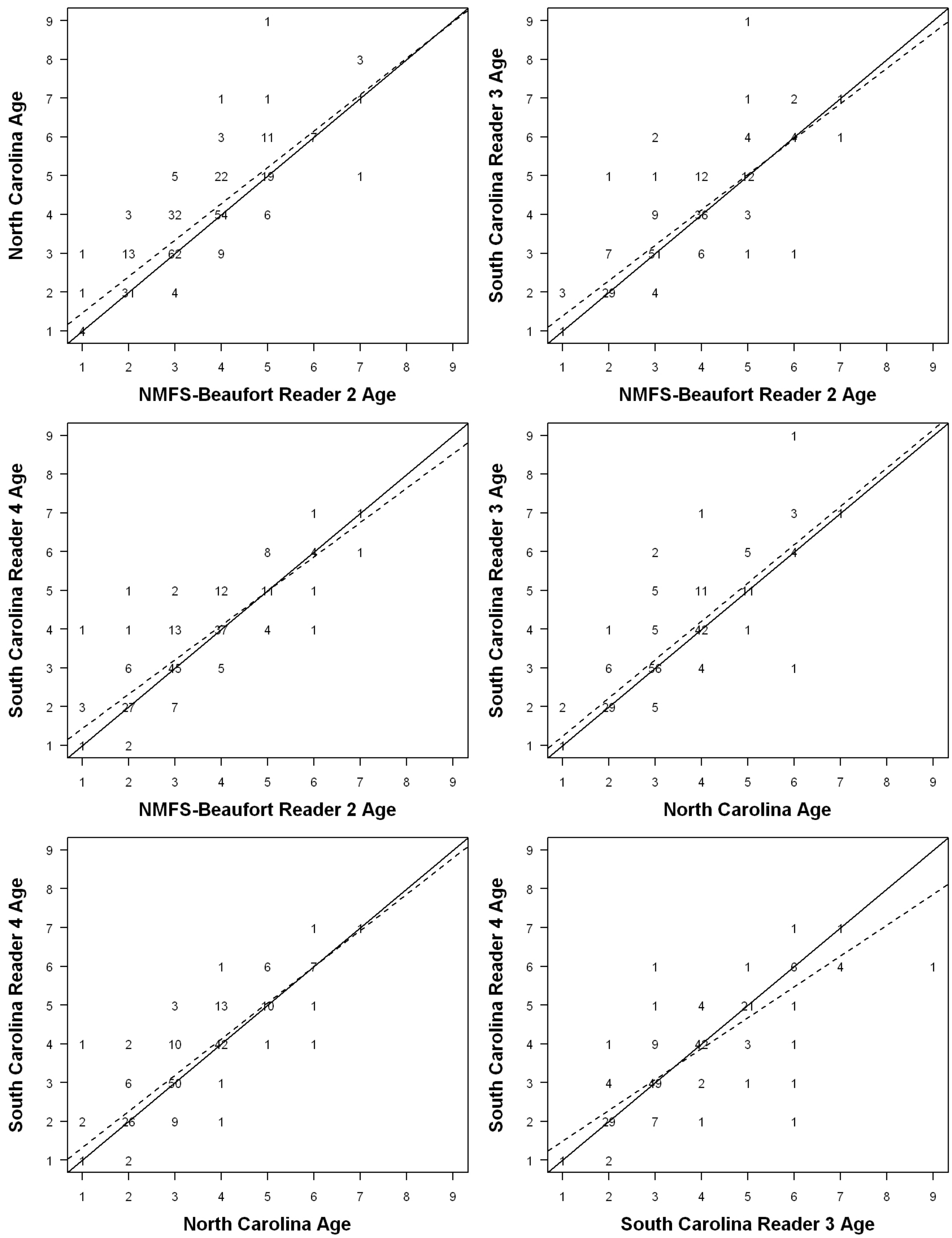
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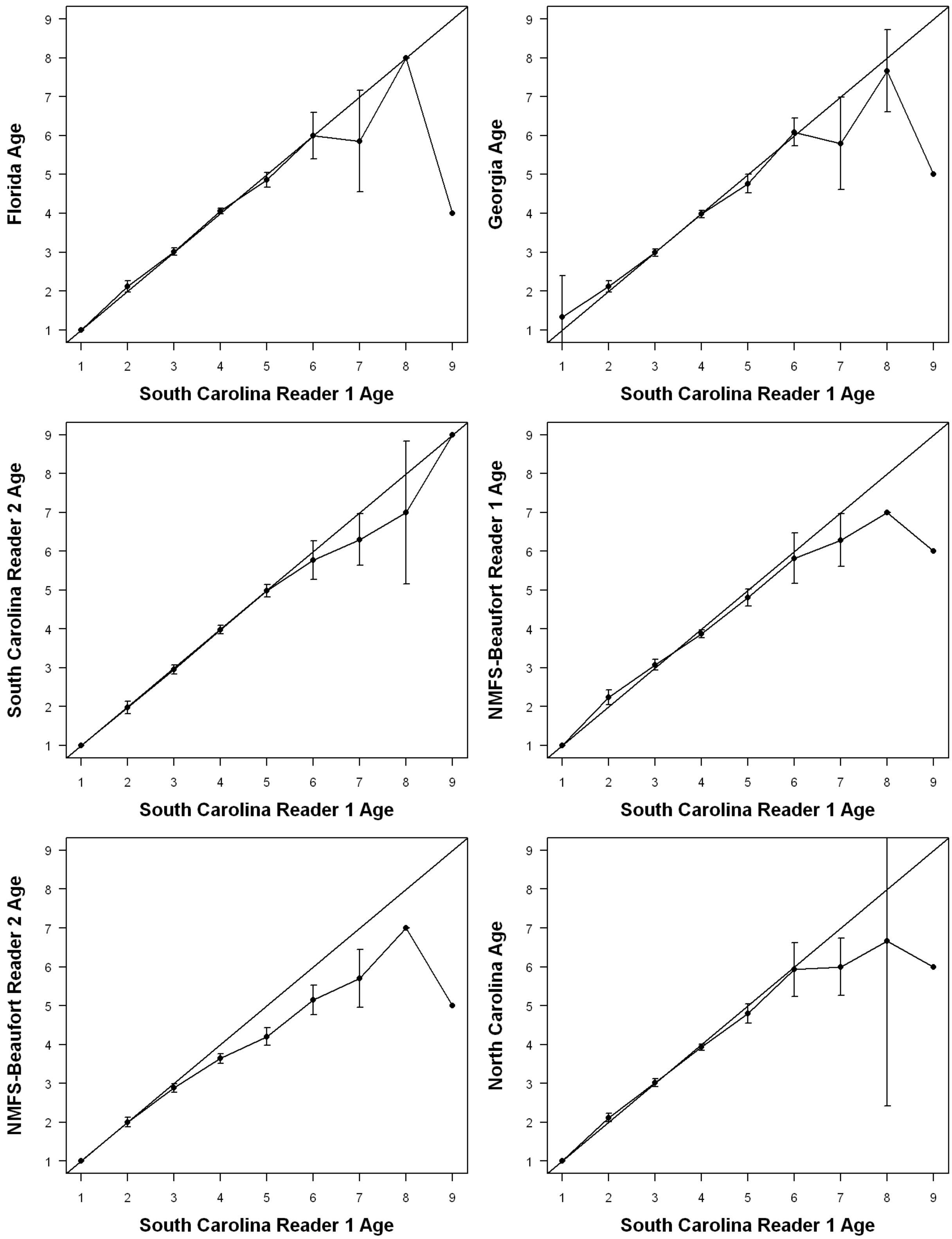
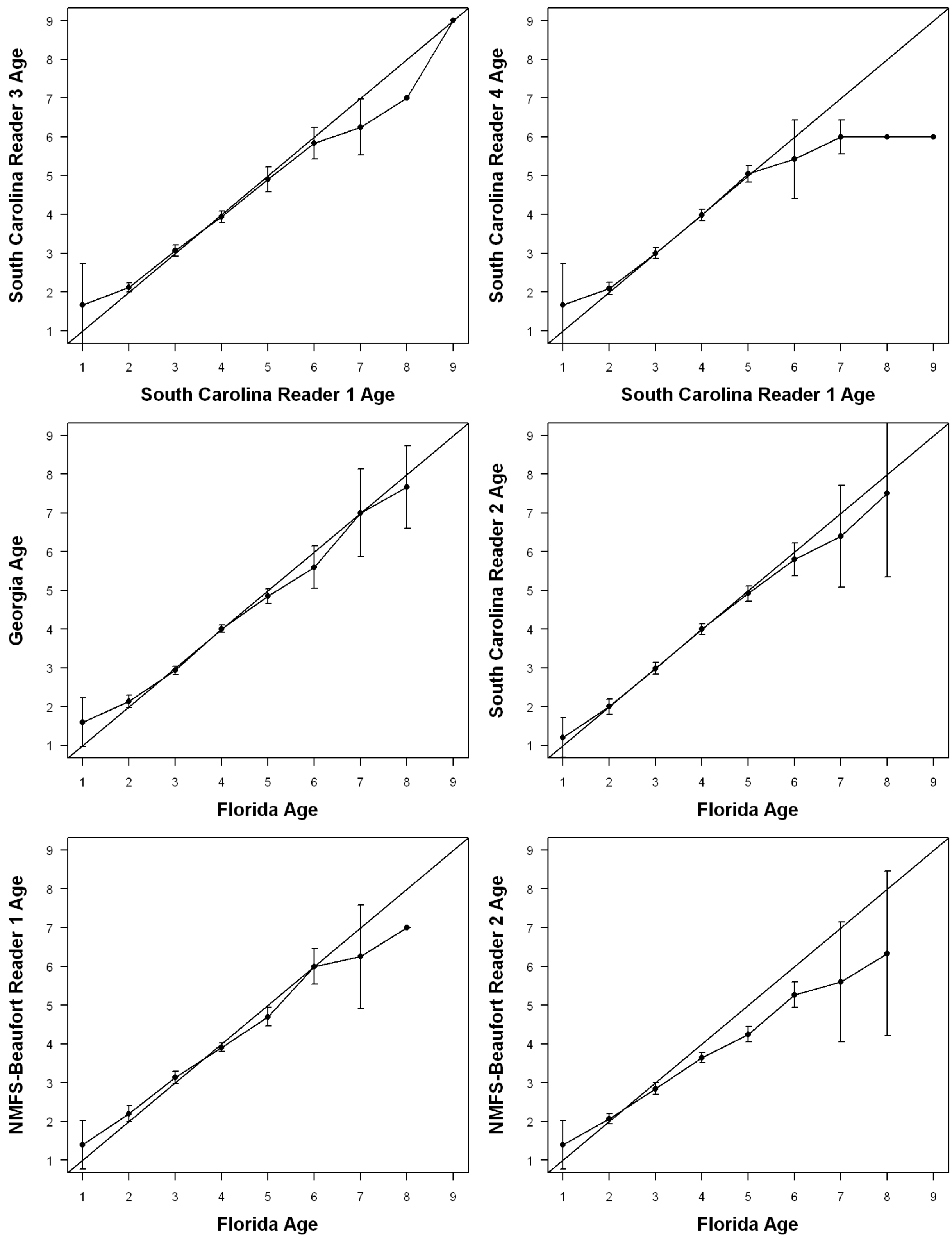
South Carolina Reader 3 Age

Figure 4: Bias plots of all pairwise age comparisons among the nine readers. Each error bar represents the 95% confidence interval about the mean age assigned by one ager (y-axis) for all fish assigned a given age by the second reader (x-axis).

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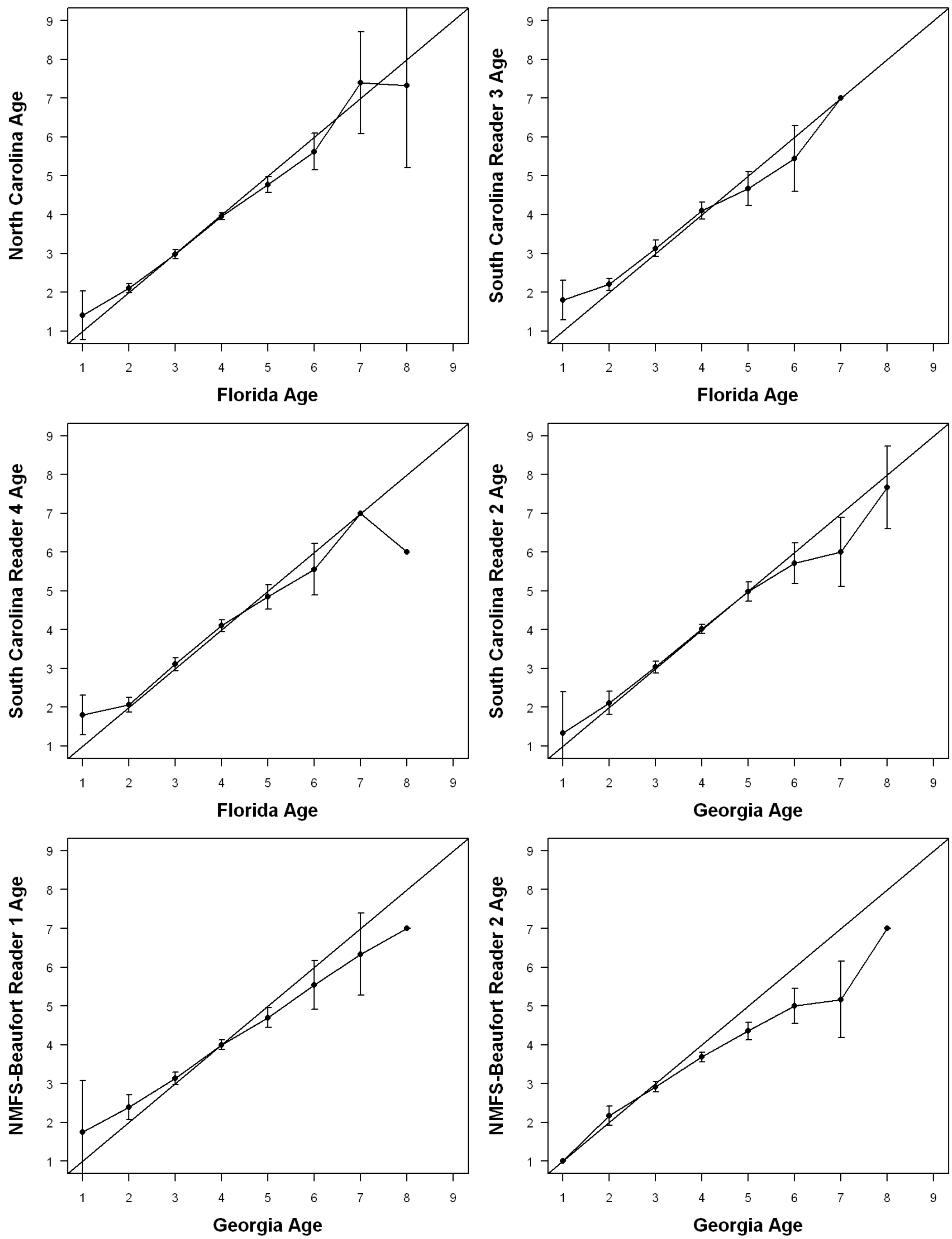
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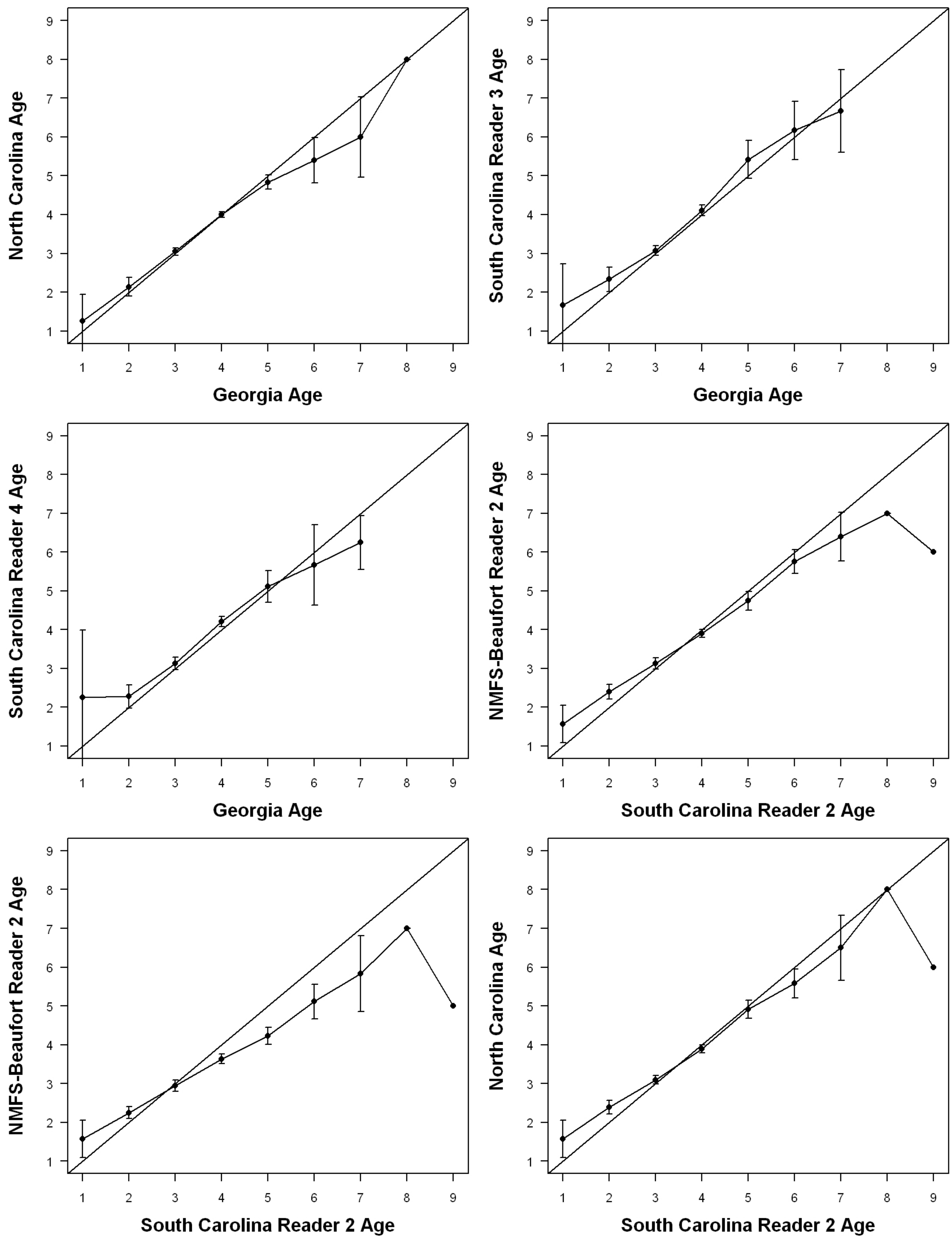
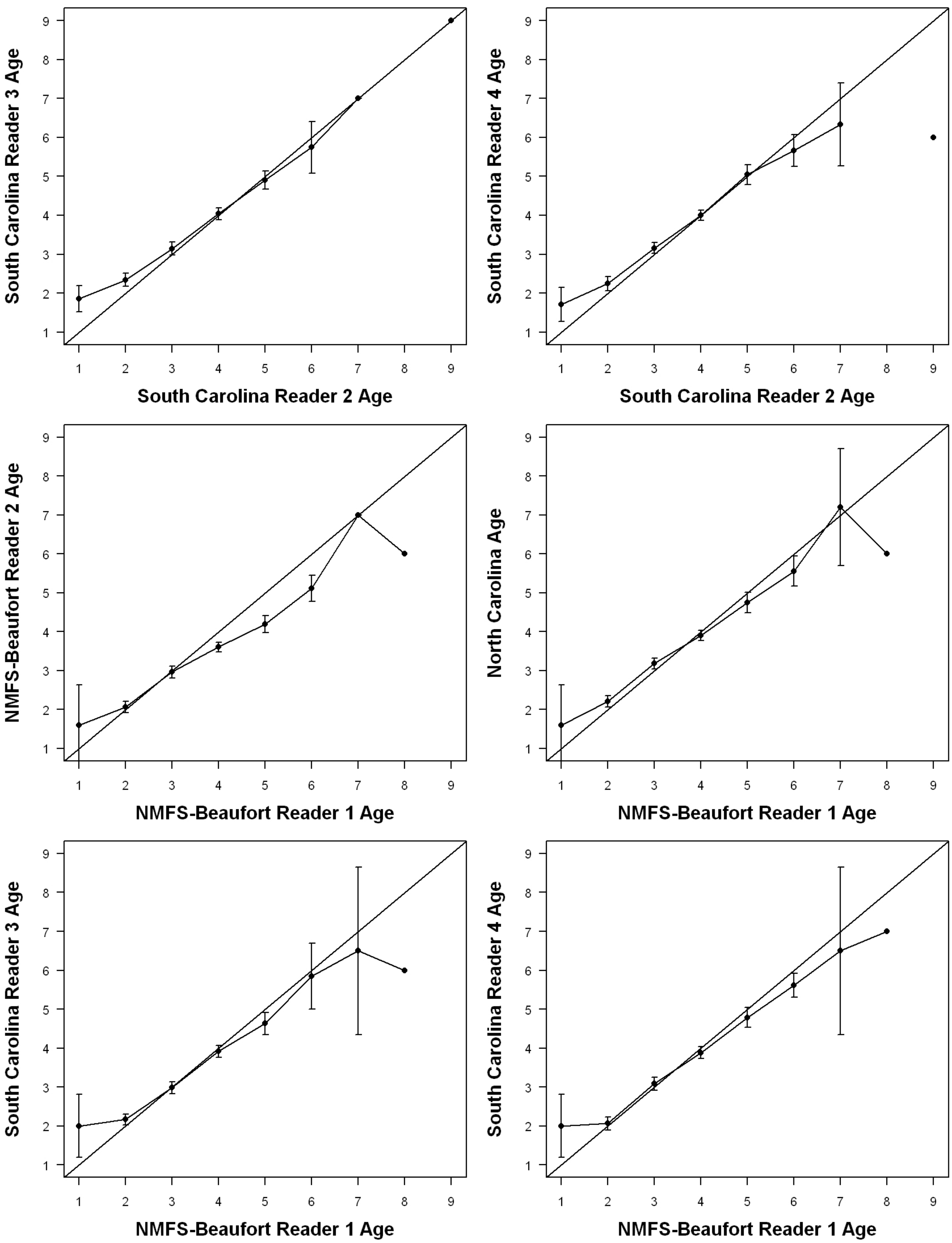
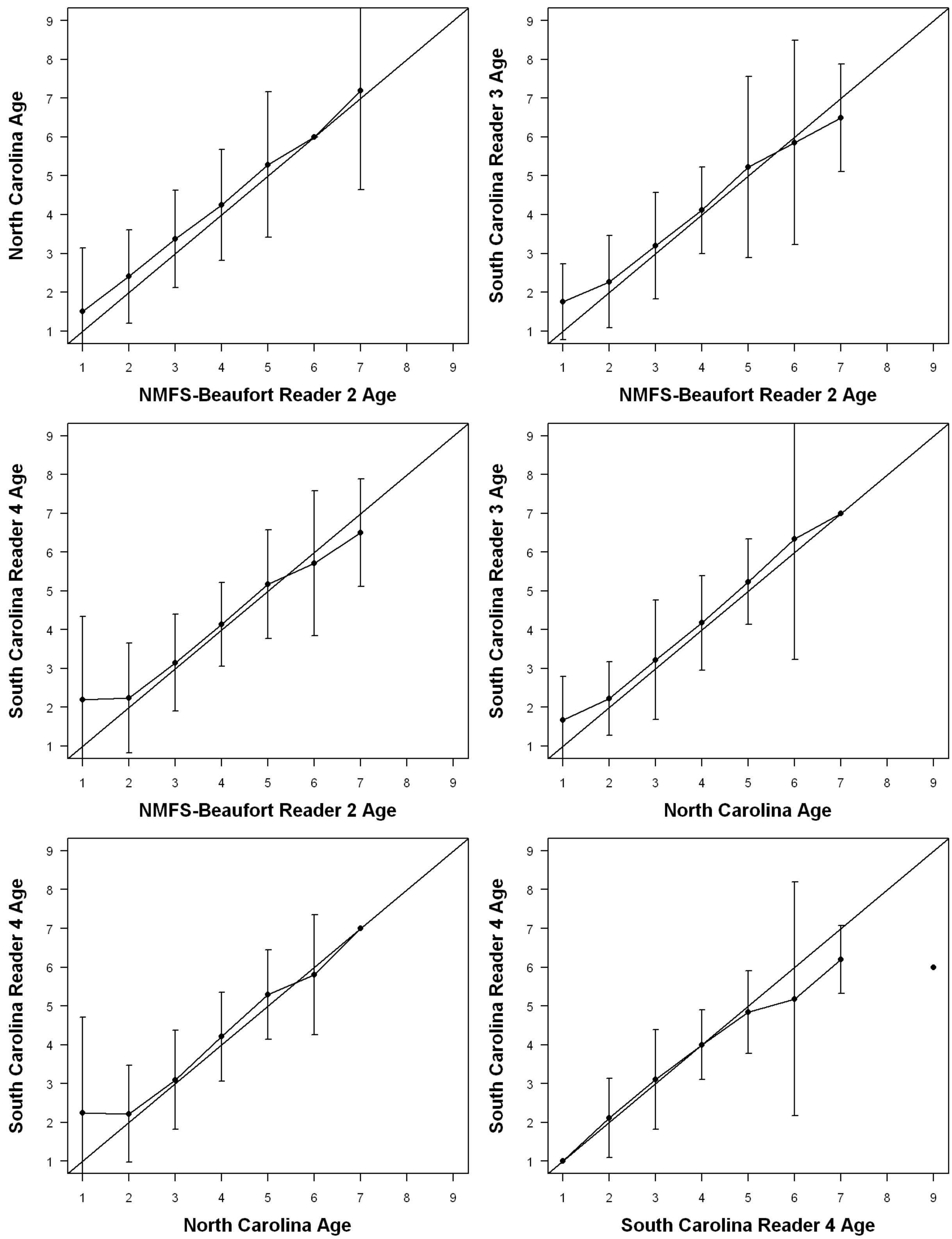
NMFS-Beaufort Reader 2 Age
Georgia Age

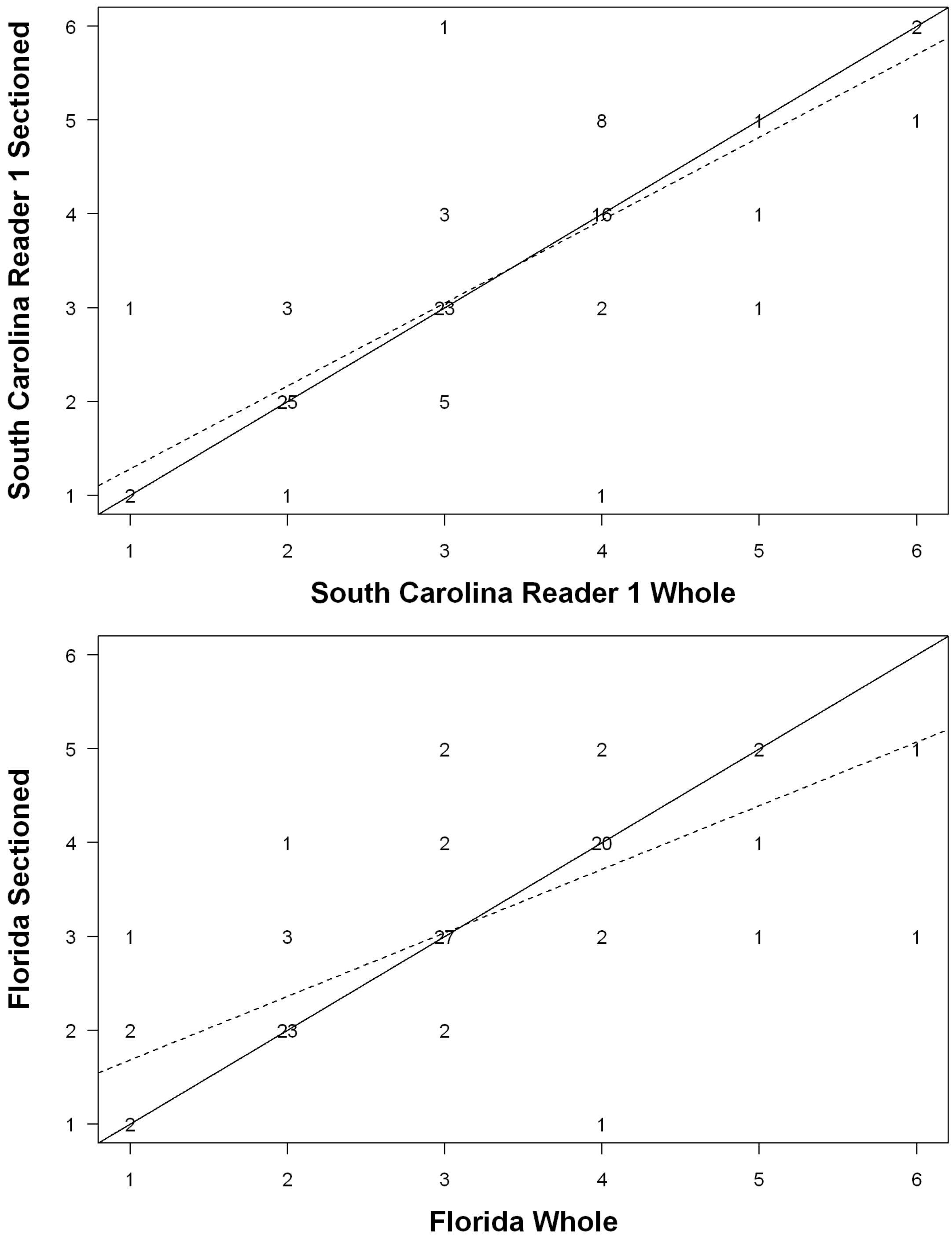

North Carolina Age
Georgia Age
South Carolina Reader 3 Age
Georgia Age
South Carolina Reader 4 Age
Georgia Age
NMFS-Beaufort Reader 2 Age
South Carolina Reader 2 Age
NMFS-Beaufort Reader 2 Age
South Carolina Reader 2 Age
North Carolina Age
South Carolina Reader 2 Age

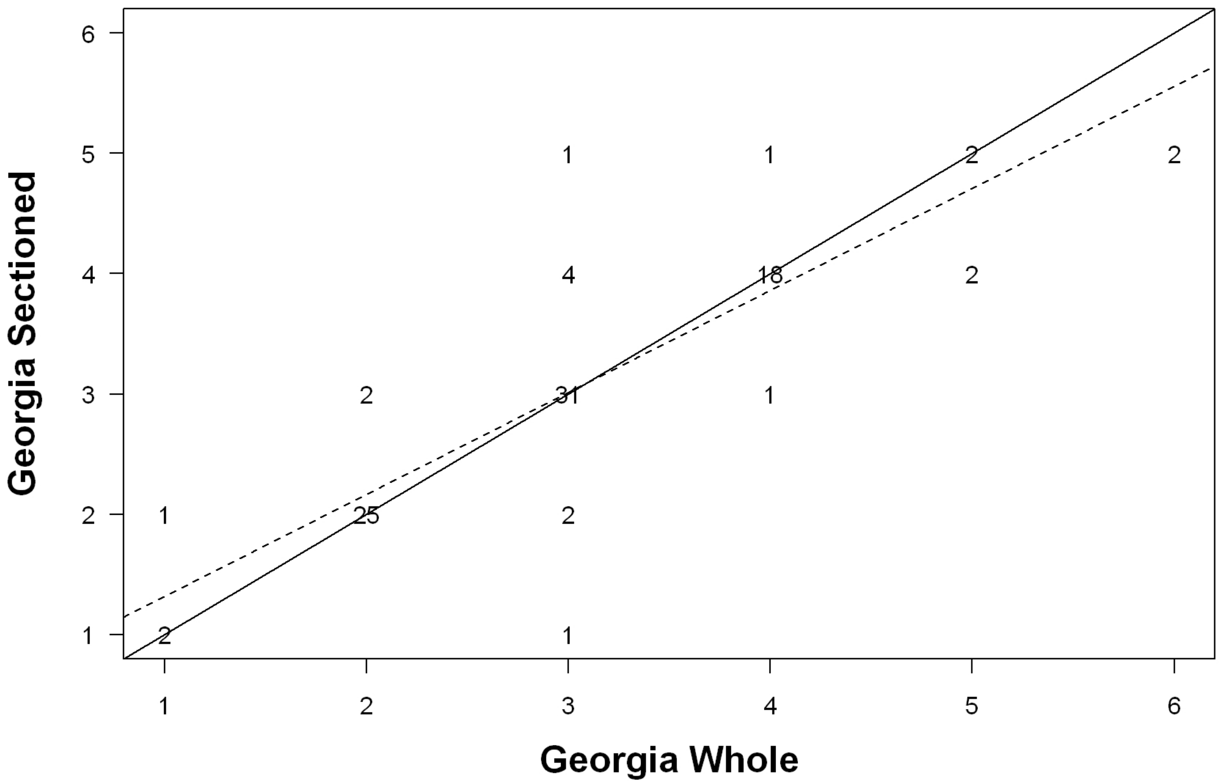

South Carolina Reader 3 Age
South Carolina Reader 2 Age
South Carolina Reader 4 Age
South Carolina Reader 2 Age
NMFS-Beaufort Reader 2 Age
NMFS-Beaufort Reader 1 Age
North Carolina Age
NMFS-Beaufort Reader 1 Age
South Carolina Reader 3 Age
NMFS-Beaufort Reader 1 Age
South Carolina Reader 4 Age
NMFS-Beaufort Reader 1 Age

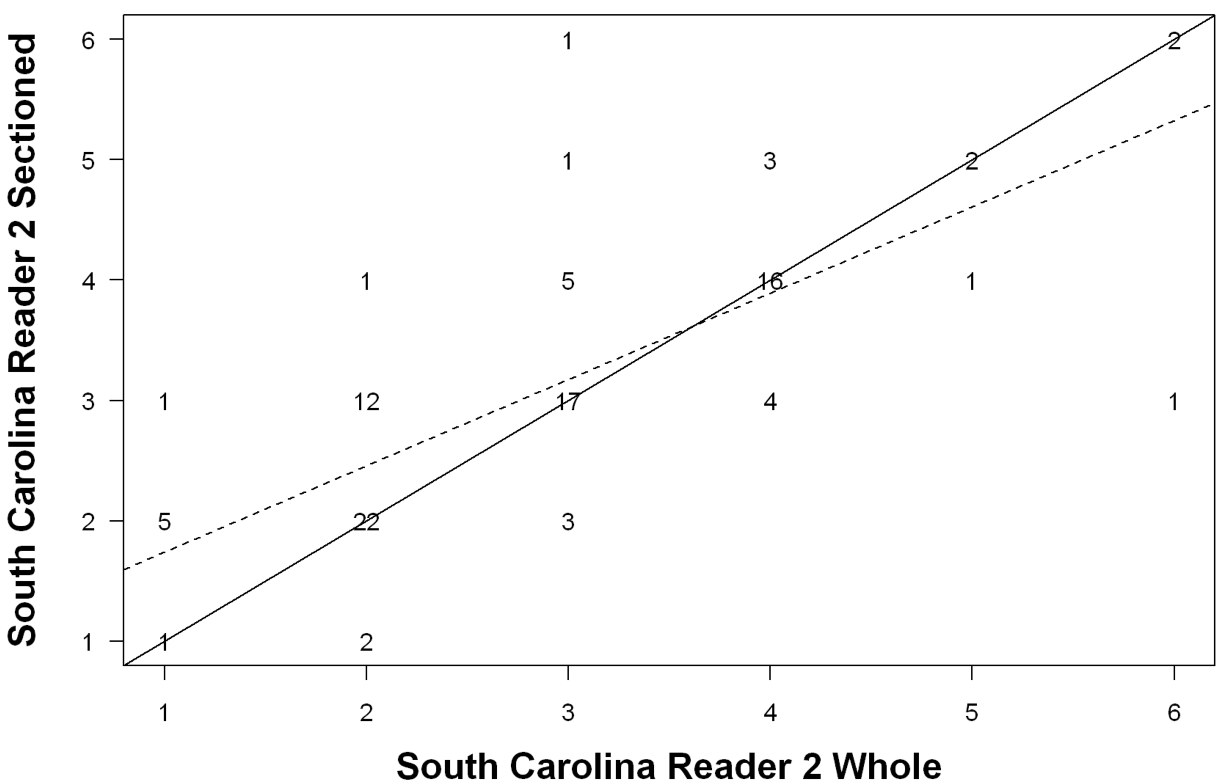


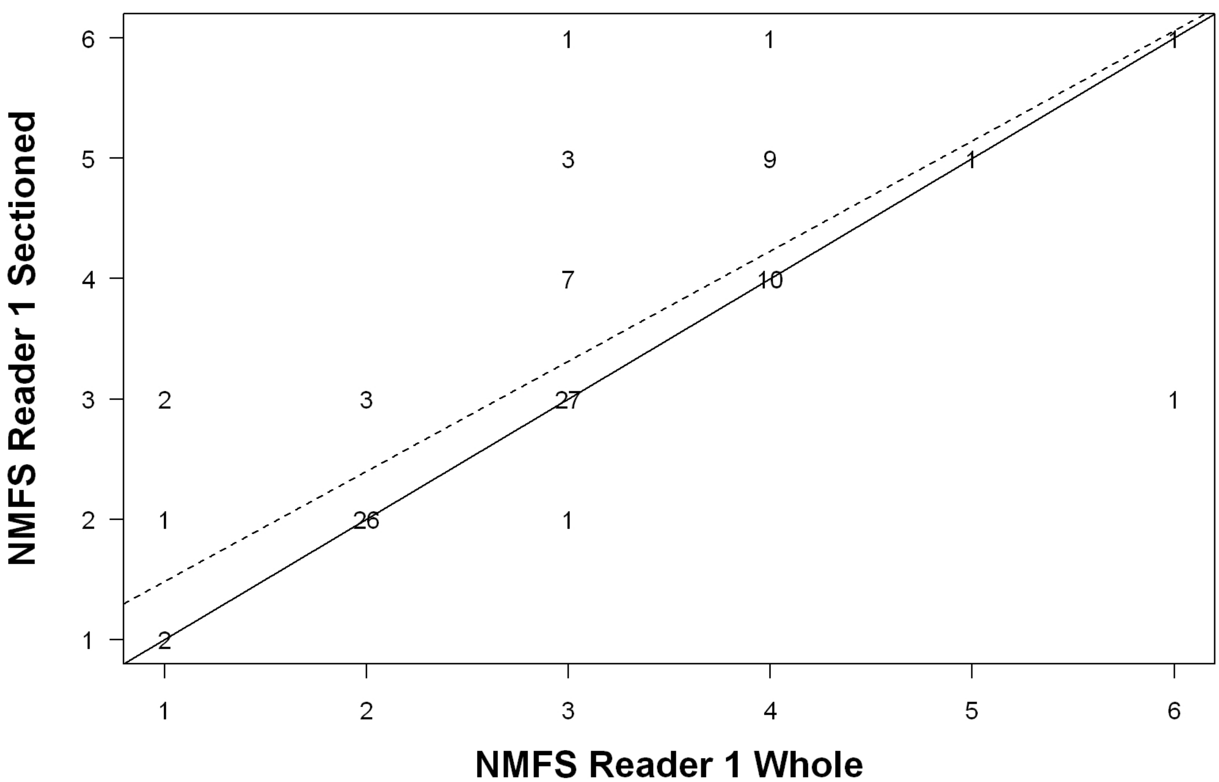
North Carolina Age
NMFS-Beaufort Reader 2 Age
South Carolina Reader 3 Age
NMFS-Beaufort Reader 2 Age
South Carolina Reader 4 Age
NMFS-Beaufort Reader 2 Age
South Carolina Reader 3 Age
North Carolina Age
South Carolina Reader 4 Age
North Carolina Age
South Carolina Reader 4 Age
South Carolina Reader 4 Age

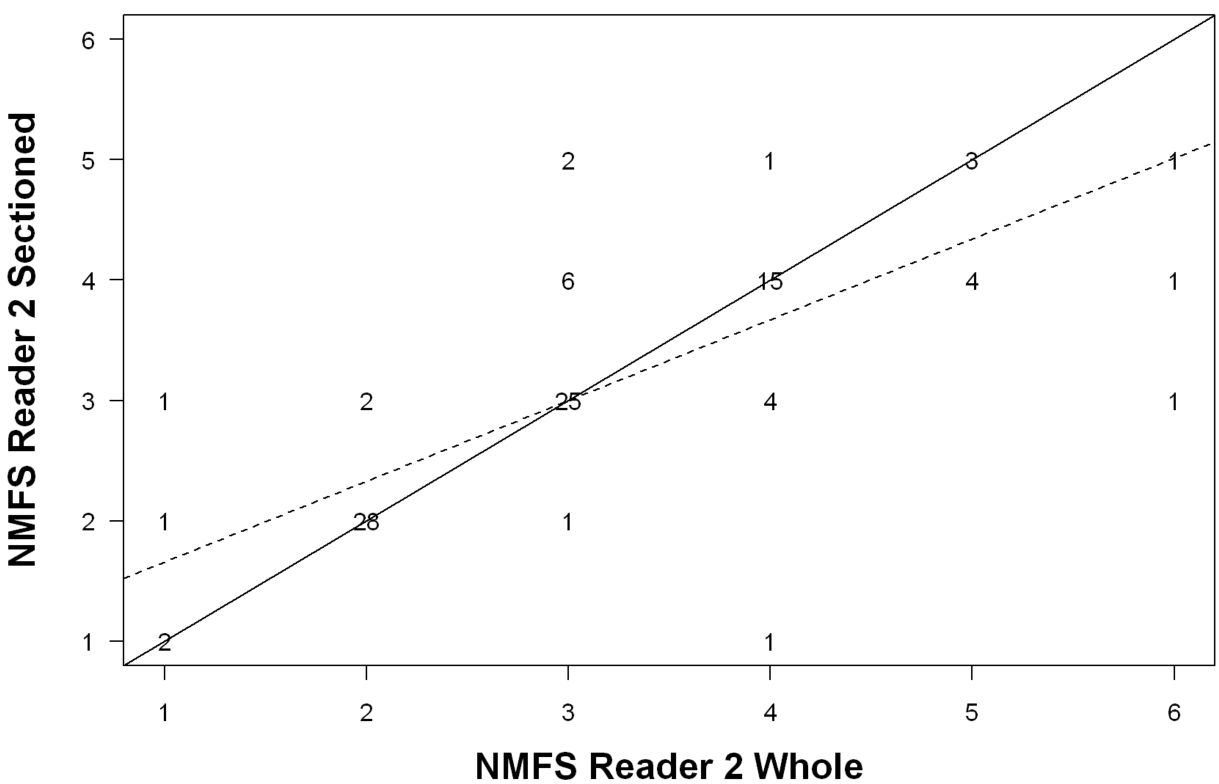
Figure 5: Plots of linear regressions of whole vs. sectioned otolith age readings for BSB primary age readers for upcoming SEDAR BSB update. Numbers represent the number of fish assigned that combination of ages by the different aging methods. Solid line indicates perfect agreement (1:1 line) between aging methods. Dashed line indicates the calculated linear regression between age readings of the different methods.

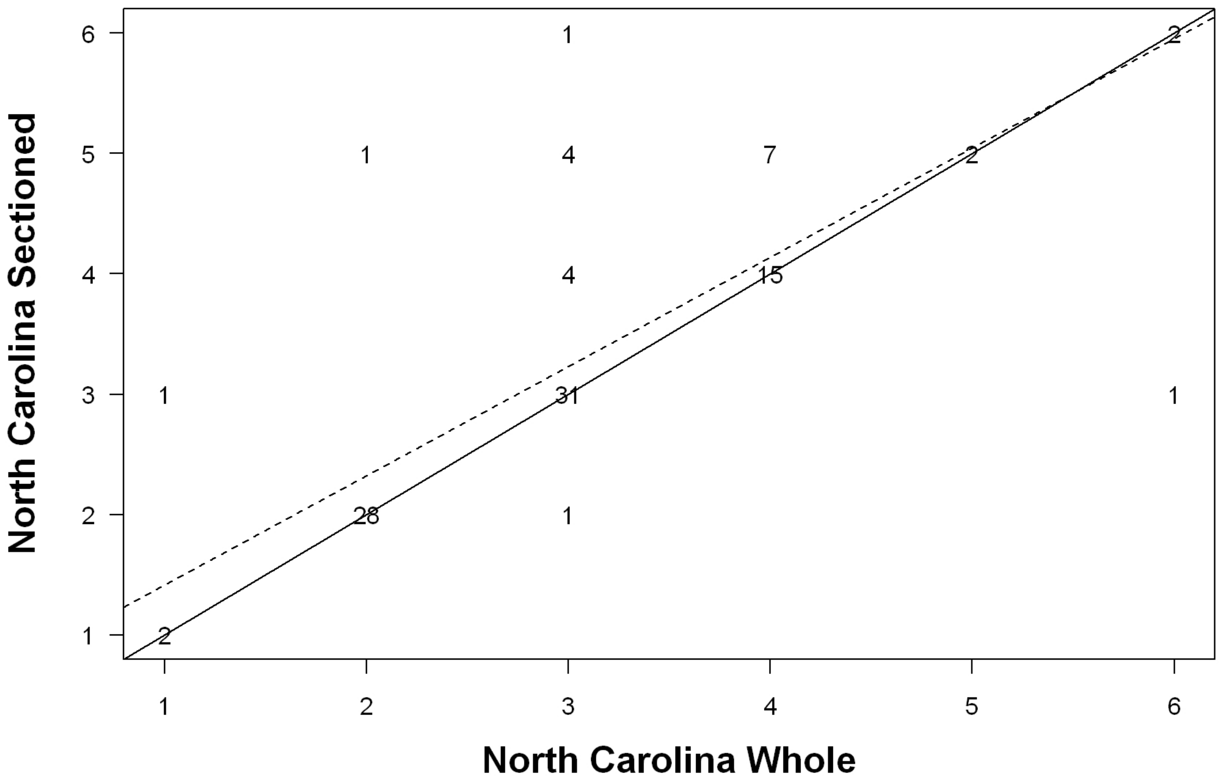


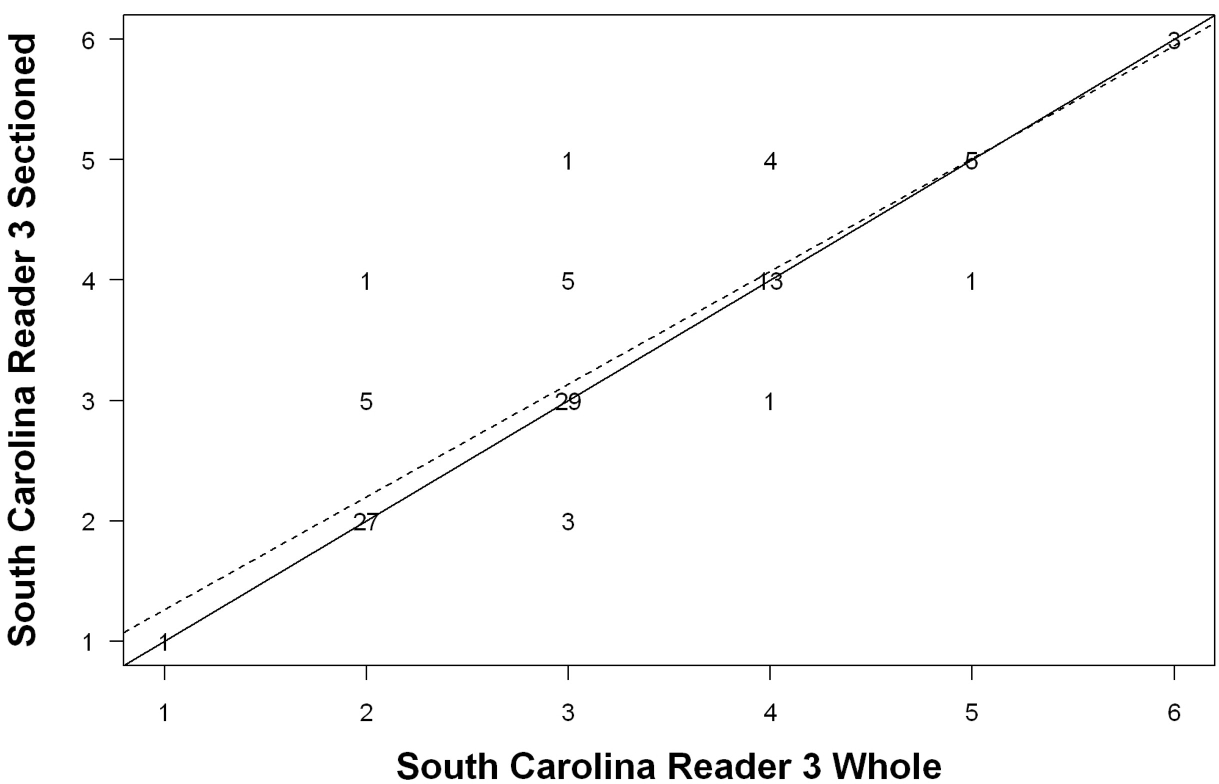

Georgia Sectioned
Georgia Whole
1
2
3
4
5
6
1
1
2
4
18
2
2
31
1
1
25
2
2
1
2
2


South Carolina Reader 2 Sectioned
South Carolina Reader 2 Whole
1
2
3
4
5
6
1
2
1
3
2
1
5
16
1
1
12
17
4
1
5
22
3
1
2


NMFS Reader 1 Sectioned
NMFS Reader 1 Whole
1
2
3
4
5
6
1 1 1
3 9 1
7 10
2 3 27 1
1 26 1
2


NMFS Reader 2 Sectioned
NMFS Reader 2 Whole
1
2
3
4
5
6
2 1 3 1
6 15 4 1
1 2 25 4 1
1 28 1
2 1


North Carolina Sectioned
North Carolina Whole
1 2 3 4 5 6
1 2 3 4 5 6
1
2
1
4
7
2
4
15
1
31
1
28
1
2


South Carolina Reader 3 Sectioned
South Carolina Reader 3 Whole
1 2 3 4 5 6
1 2 3 4 5 6
3
1
4
5
1
5
13
1
5
29
1
27
3
1

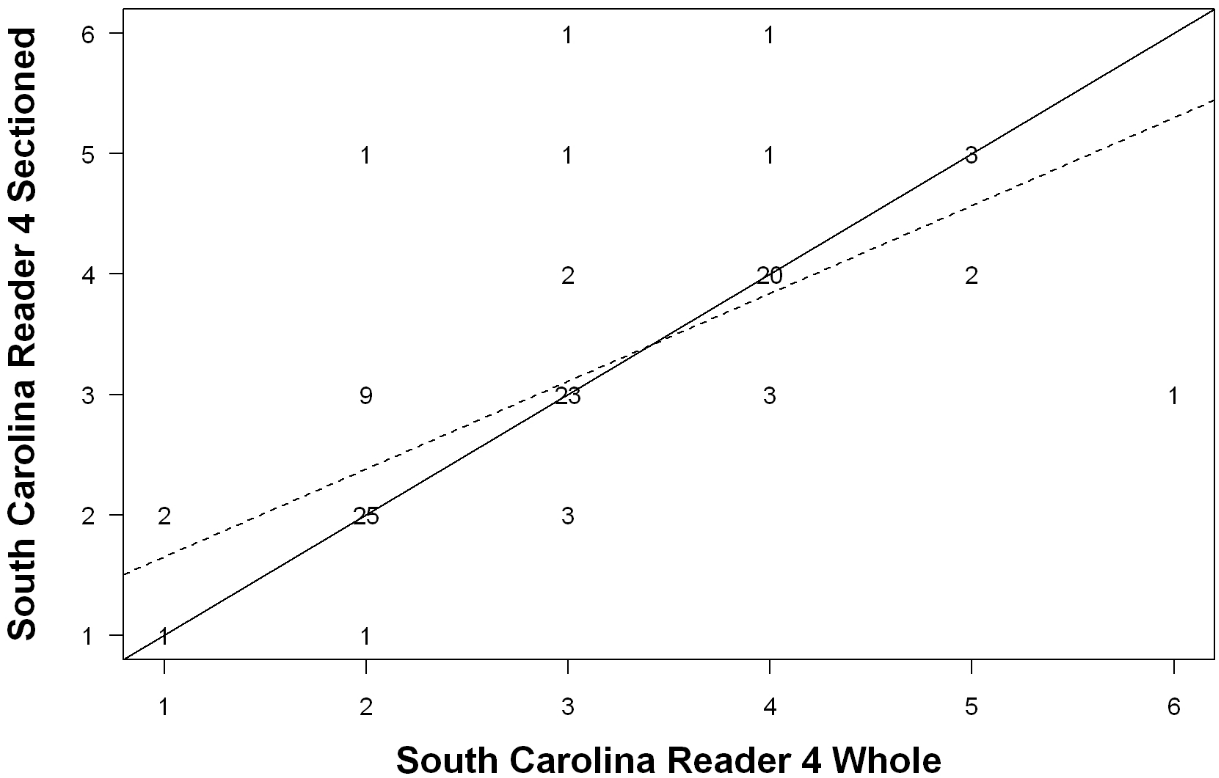
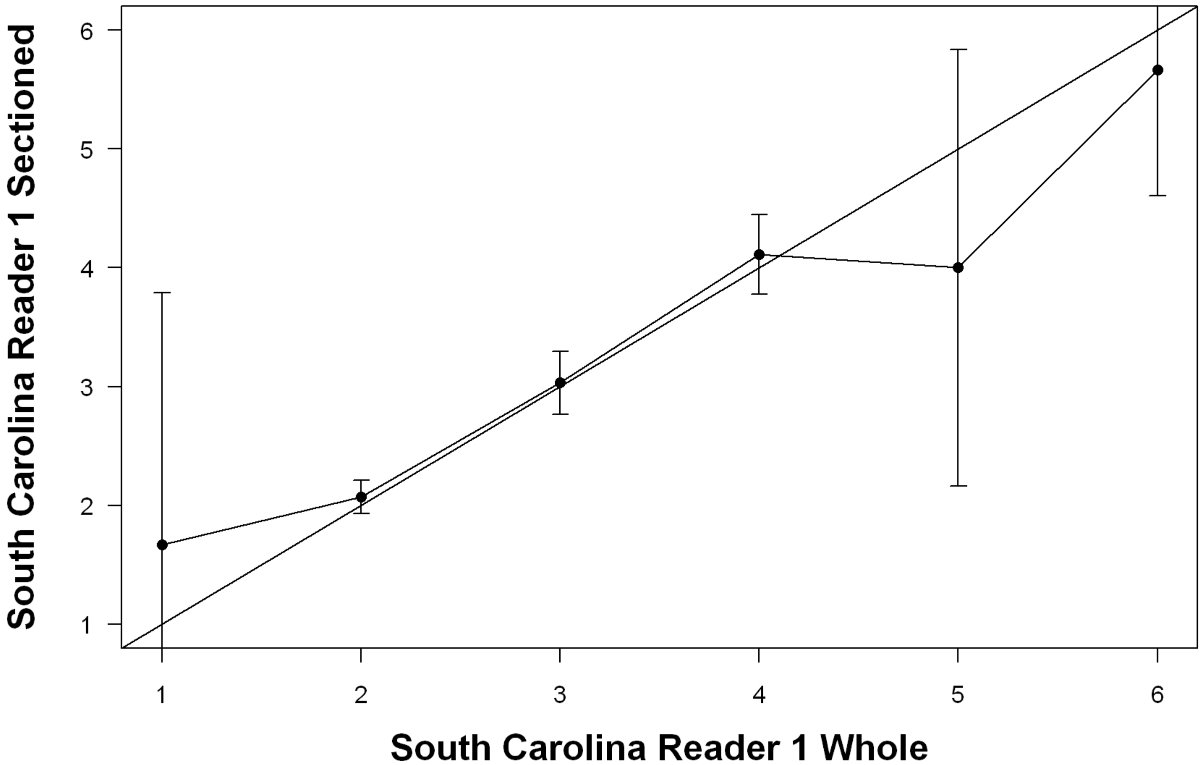
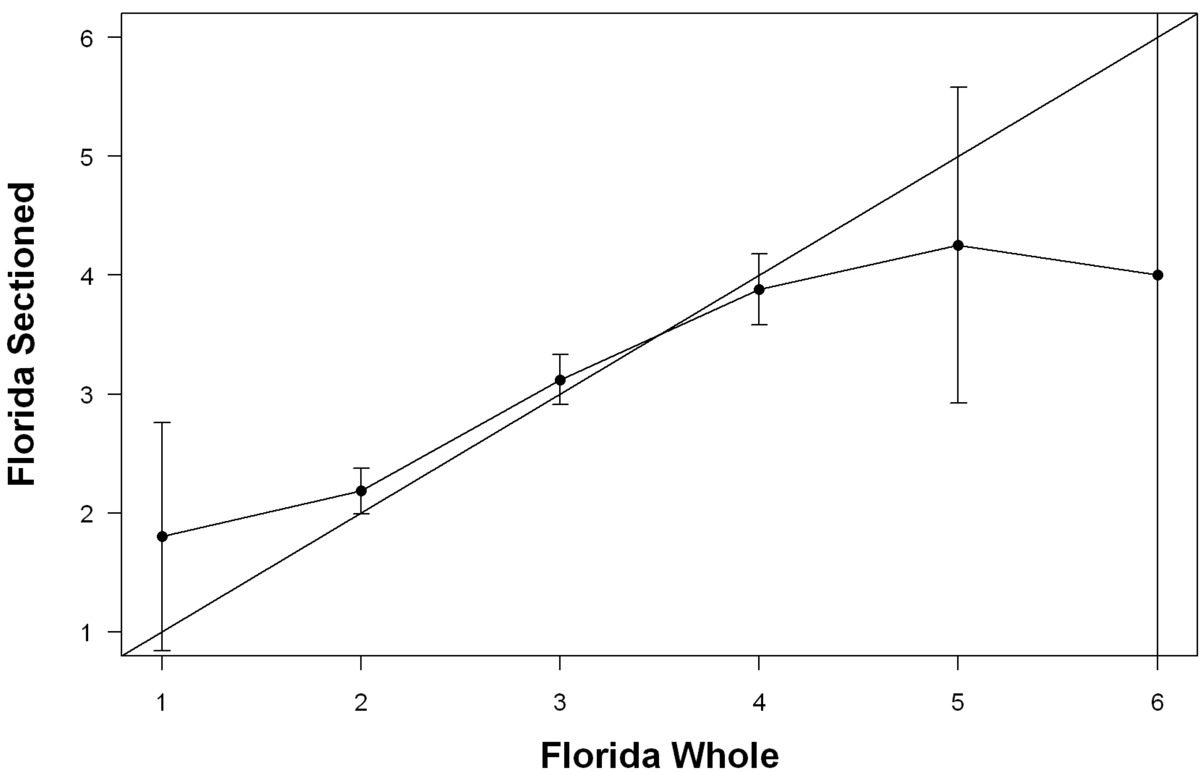
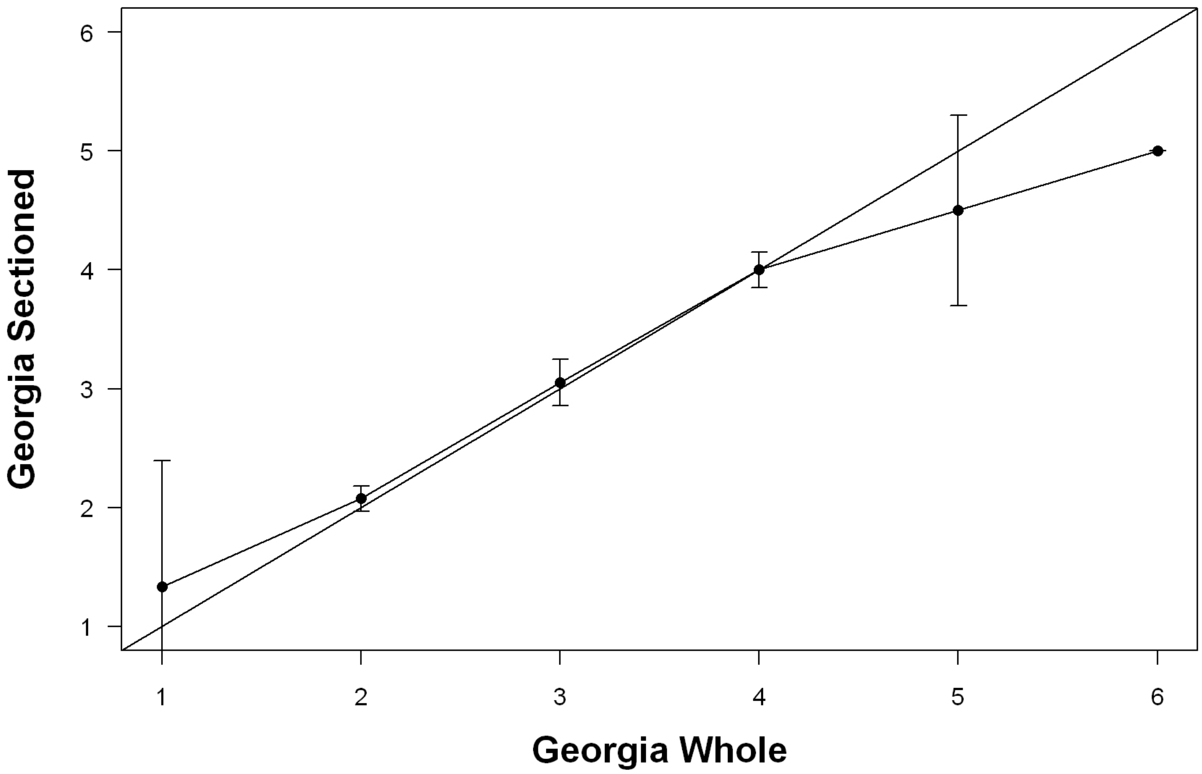
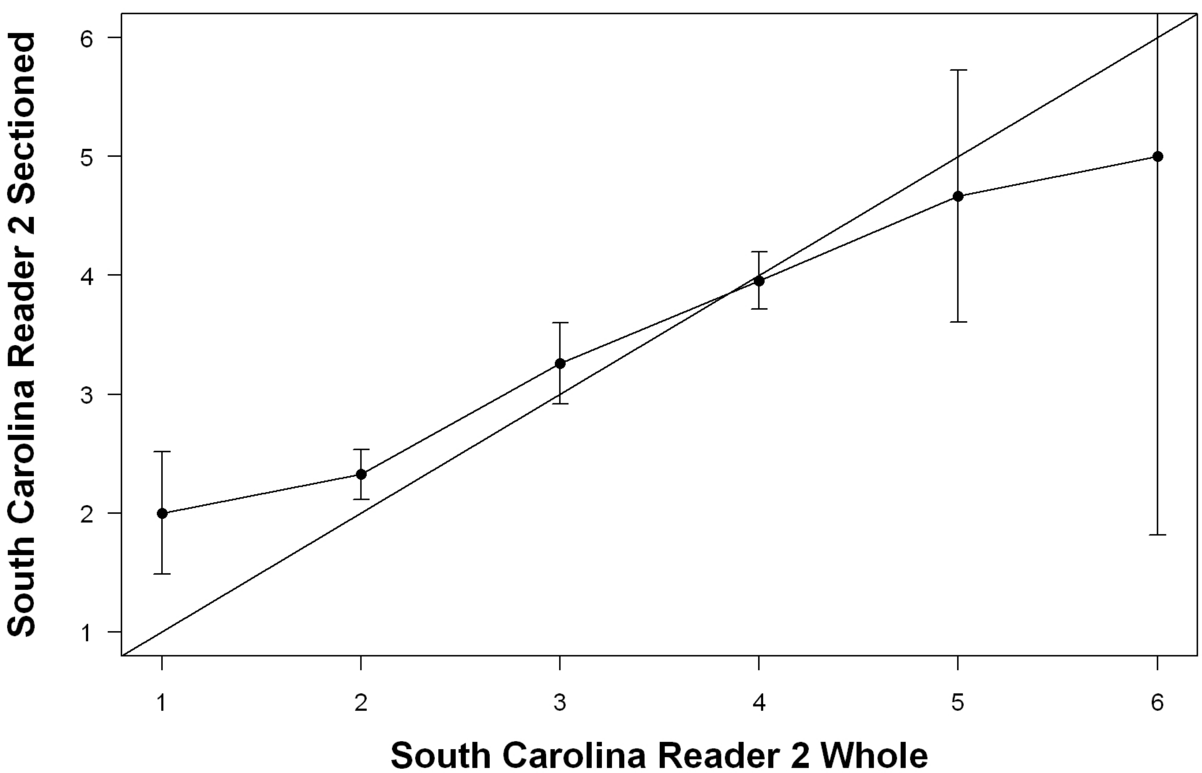

South Carolina Reader 4 Sectioned
South Carolina Reader 4 Whole
1
2
3
4
5
6
1
1
1
1
1
1
3
2
20
2
9
23
3
1
2
25
3
1
1

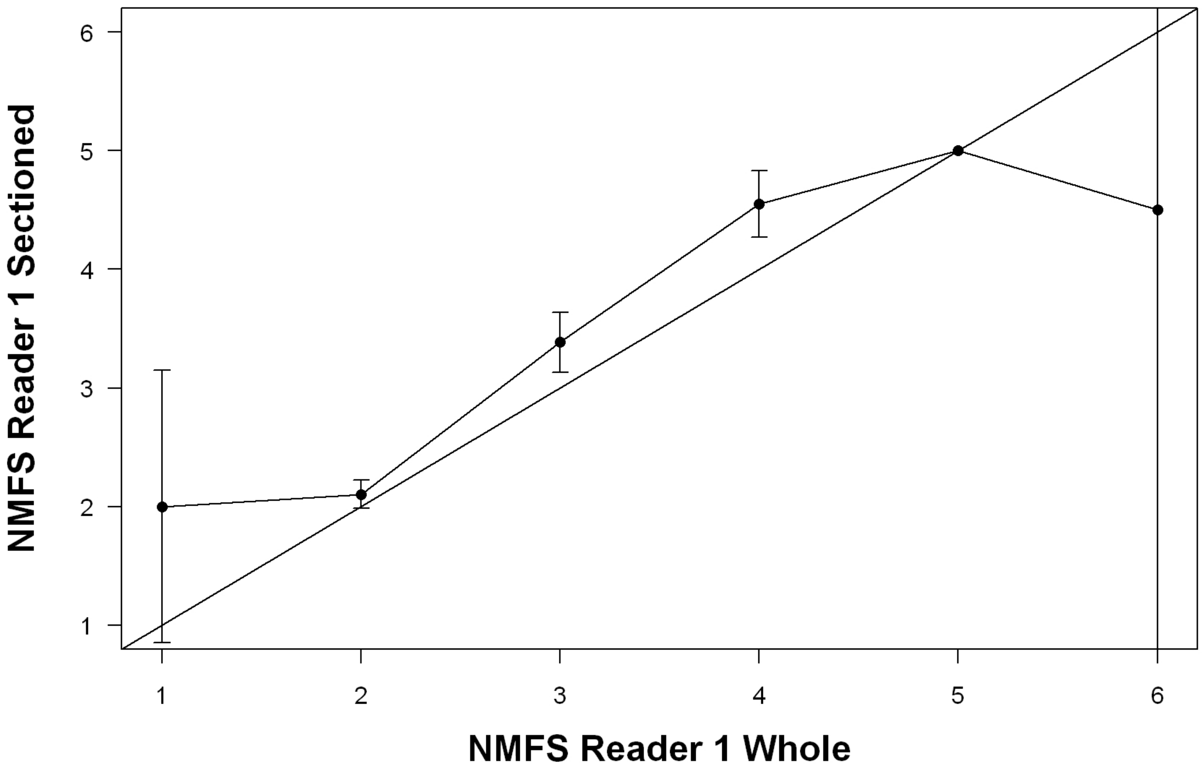
Figure 6: Bias plots of all pairwise whole otolith vs. sectioned otolith age. Each error bar represents the 95% confidence interval about the mean age assigned via sectioned otolith (y-axis) for all fish assigned a given age by whole otolith (x-axis).

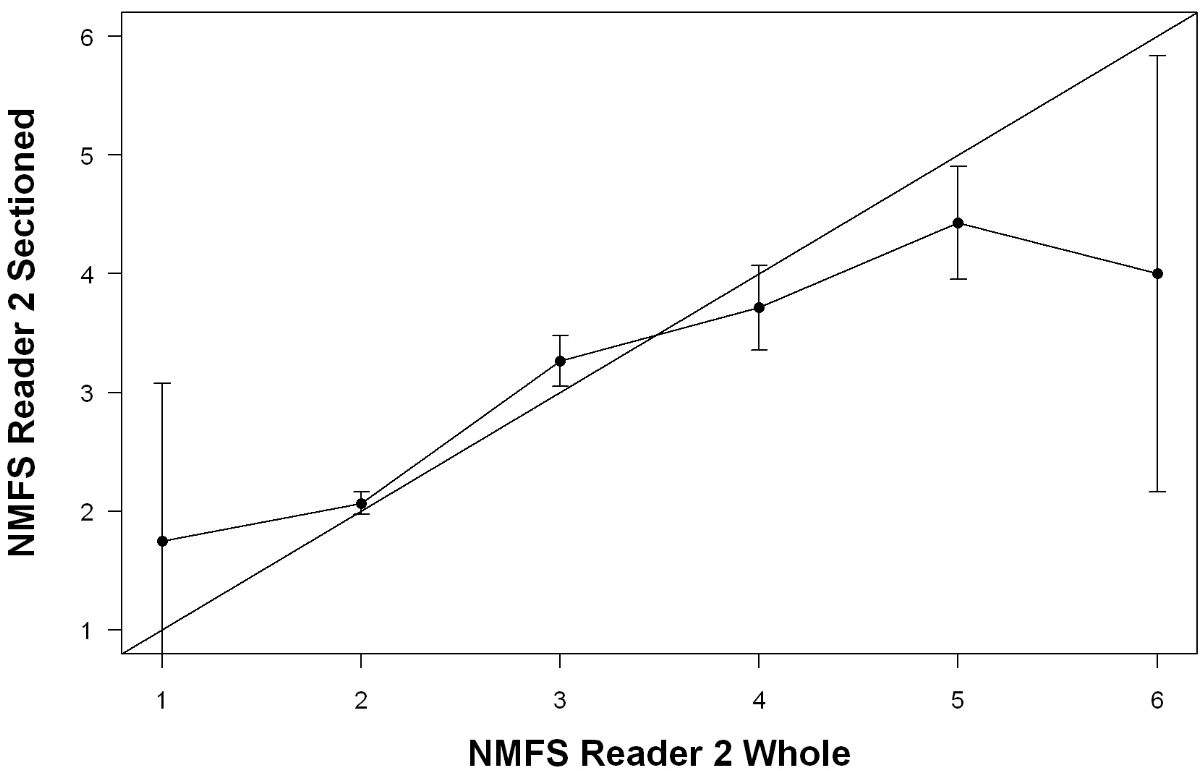


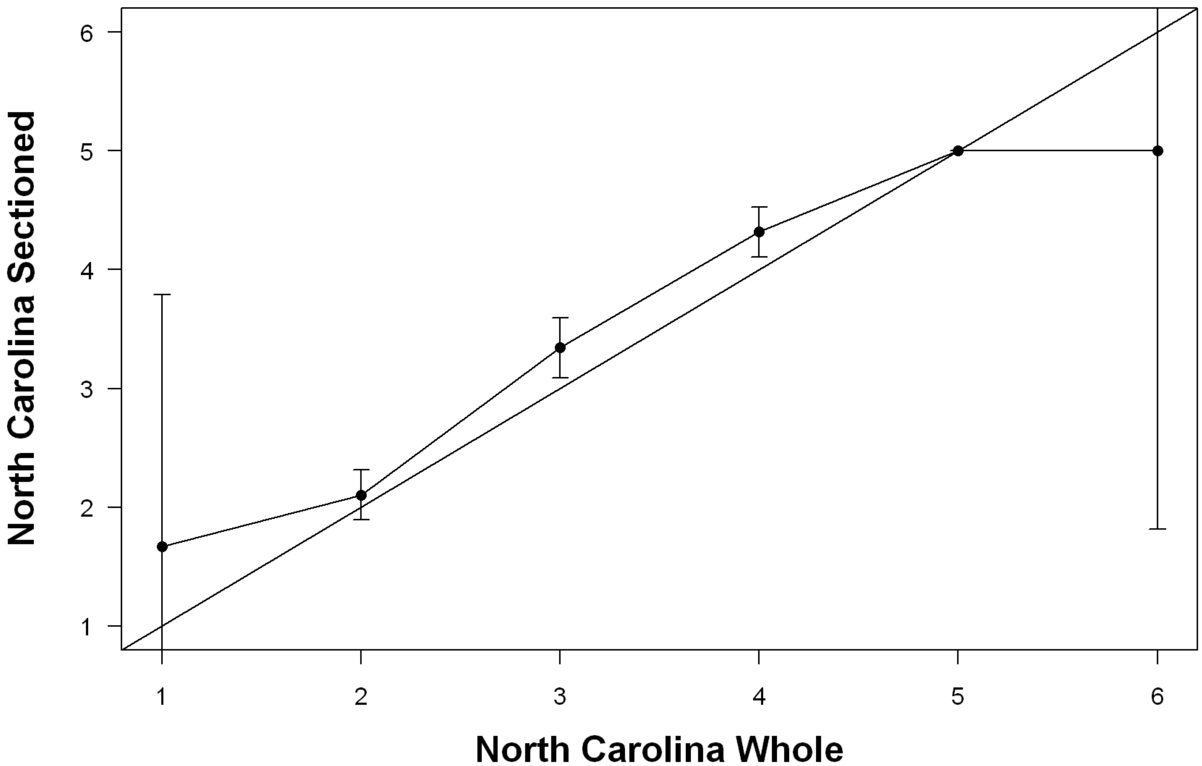


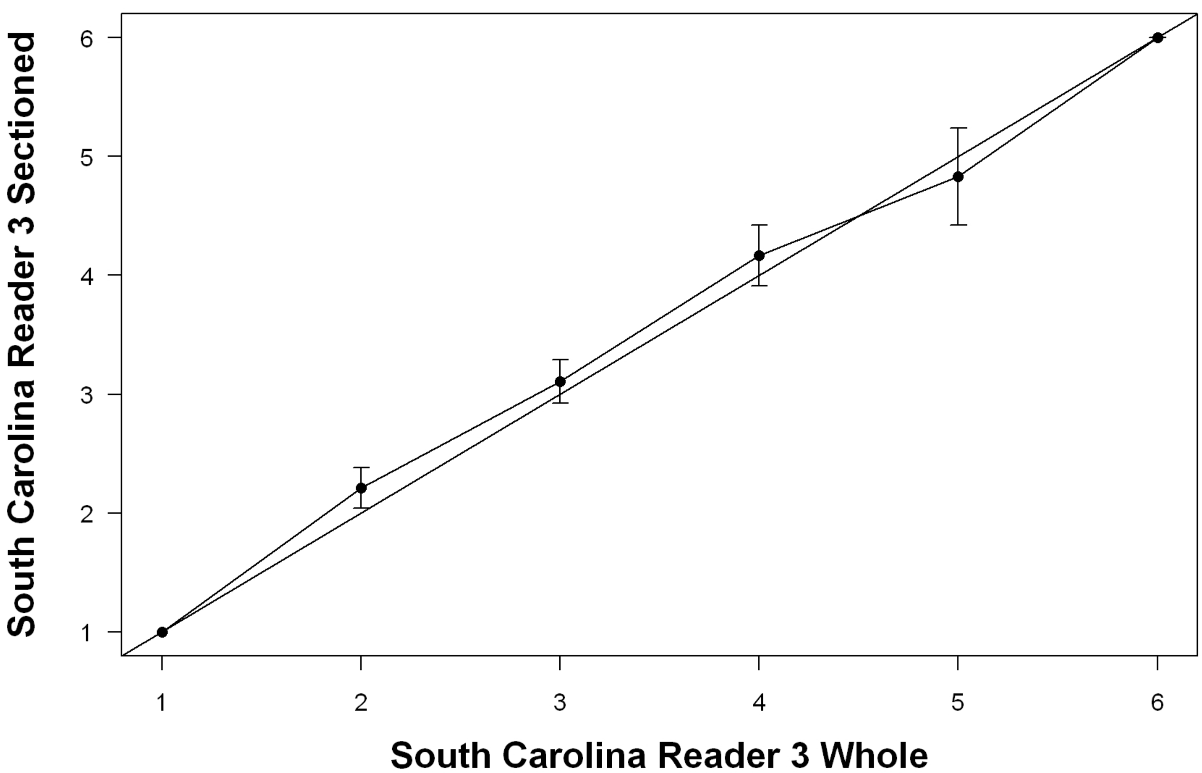

Georgia Sectioned
Georgia Whole
1
2
3
4
5
6

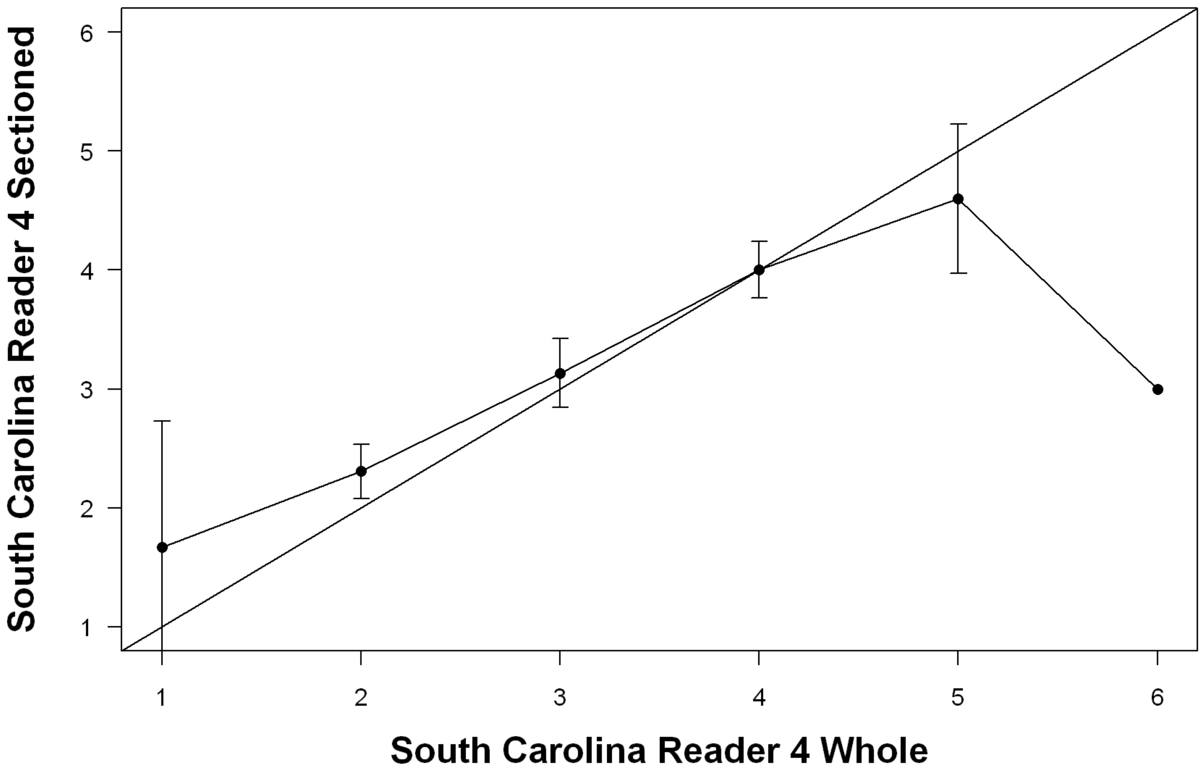

South Carolina Reader 2 Sectioned
South Carolina Reader 2 Whole
1
2
3
4
5
6


6
5
4
3
2
1
1
2
3
4
5
6
NMFS Reader 1 Sectioned
NMFS Reader 1 Whole


6
5
4
3
2
1
1
2
3
4
5
6
NMFS Reader 2 Sectioned
NMFS Reader 2 Whole


North Carolina Sectioned
North Carolina Whole
1
2
3
4
5
6


South Carolina Reader 3 Sectioned
South Carolina Reader 3 Whole
1
2
3
4
5
6


South Carolina Reader 4 Sectioned
6
5
4
3
2
1
1
2
3
4
5
6
South Carolina Reader 4 Whole