# Use of Scale Morphology for Discriminating Wild Stocks of Atlantic Striped Bass

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Abstract.—We used scale morphology to discriminate wild Atlantic striped bass Morone saxatilis from the Hudson River, Chesapeake Bay, and the Roanoke River. Morphological features used were perimeter shape as described by Fourier analysis, rectangularity, width and spacing of the first 10 circuli, and patterns of partitioning of the scale interior. Performance of discriminant functions was evaluated by examining the percent correct classification of known-origin samples (precision) and the bias in resulting stock composition estimates (accuracy). Correct classification rates ranged from 57% to 84%, varying with the number of stocks and year-classes included. Lowest rates were for three stocks with multiple year-classes; highest rates were for single-year-class analysis of the most geographically distant stocks. Despite some poor classification rates, bias and variance in stock composition estimates were low in four of five discrimination problems, suggesting that corrections based on the classification matrix could be applied with some confidence to unknown samples. However, we used bootstrapping to evaluate bias stability; further evaluation of independent samples would be useful.

Three major anadromous stocks of Atlantic striped bass Morone saxatilis occur along the east coast of the United States, spawning in the Hudson River, Chesapeake Bay, and the Roanoke River (Berggren and Lieberman 1978). Another major spawning stock once occurred in the Delaware River, but its abundance has been low for most of this century (Chittenden 1971; Rago et al. 1993). During late spring, summer, and fall, subadult and adult striped bass are distributed in coastal waters from North Carolina to Canada (Merriman 1941; Chapoton and Sykes 1961; Boreman and Lewis 1987), where they have supported important fisheries since colonial times. The Hudson River and Chesapeake Bay stocks appear to be more highly migratory than the Roanoke River population (Merriman 1941; Vladykov and Wallace 1952; Hassler et al. 1981; Boreman and Lewis 1987), and probably make up the bulk of the coastal stock. Striped bass stocks exhibit pronounced differences in maturation rates (Specker et al. 1987; Hoff et al. 1988; Olsen and Rulifson 1992) and recruitment levels (Boreman and Austin 1985; Richards and Deuel 1987), so stock composition is an important consideration in management of this species.

Although striped bass have been the subject of numerous stock identification studies in the past (Waldman et al. 1988), the need for information on stock composition has increased in recent years as managers have attempted to tailor regulations to rebuild some stocks and maintain others (ASMFC 1981, 1989, 1990; USDOC 1993). Important fishery and population monitoring programs are conducted in coastal areas where striped bass stocks mix (Richards and Deuel 1987; Rago et al. 1993); however, without stock discrimination these programs cannot provide population-specific estimates of vital rates. Previous studies of southern New England striped bass fisheries have shown both temporal and spatial variation in stock composition (Fabrizio 1987; Van Winkle et al. 1988; Wirgin et al. 1993), and stock composition monitoring concurrent with other biological sampling is needed

The purpose of our study was to determine whether the Hudson River, Chesapeake Bay, and Roanoke River stocks of Atlantic striped bass can be discriminated on the basis of scale morphology. Scales are routinely collected for age determination of striped bass, so sampling for stock identification would entail no further effort if scale morphology could be used. In a previous study of striped bass scales (Richards and Esteves 1997, this issue), Fourier shape descriptors, rectangularity, and some aspects of internal structure showed stock-specific differences that might form the basis for discriminating these stocks.

## Methods

Morphological variables considered in this study were Fourier coefficients (describing perimeter shape), rectangularity (a ratio shape descrip-

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| -<br>Variable         | Hudson River versus Chesapeake Bay<br>versus Roanoke River |   | Hudson River versus<br>Chesapeake Bay |                       | Hudson River<br>versus  |
|-----------------------|--|---|---------------------------------------|-----------------------|-------------------------|
|                       | All ages   | Age 2   | All ages                              | Age 2                 | Roanoke River,<br>age 2 |
| Fourier coefficients  |  |   |                                       |                       |                         |
| Amplitude coefficient | 7, 10, 11, 12  | 1, 3, 5   | 3, 5–11                               | 4, 5, 9, 11           | 5, 8, 11                |
| Phase coefficient     | 3, 10  | 2, 3, 4, 7, 9   | 2, 8, 9, 11, 12                       | 7, 9, 12              | 2, 3, 4, 7              |
| Ratio Shape           | Rectangularity   | Rectangularity  |                                       | Rectangularity        | Rectangularity          |
| Circulus<br>patterns  |  | Variance of widths 1-5<br>Variance of spaces 6-10<br>Average of widths 6-10 |                                       | Average of widths 1-5 |                         |
| Angles                | Left angle   | Left, right angles  | Right angle;<br>asymmetry             |                       | Left, right angle       |

TABLE 1.—Scale morphology variables included in discriminant analyses of striped bass from three stocks.

tor), circulus deposition patterns, and angular measurements of the scales' interior. The scales were from striped bass collected during the spawning season on spawning grounds in the Hudson River, Chesapeake Bay, and Roanoke River, Descriptions of sampling and data collection methods are provided by Richards and Esteves (1997). A subset of variables was selected for use in discriminant analysis. Variables were included in discriminant analyses if they could be corrected for variation with size according to the methods of Reist (1986), if they showed significant stock-of-origin effects in a multivariate analysis of variance (MANOVA), and if they were not highly correlated with other included variables (r < 0.70) (Bliss 1970). We eliminated size-corrected variables that showed significant year-class effects because we did not wish to discriminate on the basis of age structure differences among stocks.

We performed five sets of discriminant analyses: (1) a three-stock analysis (Hudson River, Chesapeake Bay, and Roanoke River) of all year-classes; (2) a three-stock analysis of only age-2 fish; (3) a Chesapeake Bay versus Hudson River analysis of all year-classes; (4) a Chesapeake Bay versus Hudson River analysis of age-2 fish; and (5) a Hudson River versus Roanoke River analysis of age-2 fish. We varied the number of stocks examined because several studies have had limited success in distinguishing Roanoke River and Chesapeake Bay stocks (Grove et al. 1976; Fabrizio 1987; Margraf and Riley 1993). Examining only a single yearclass allowed us to include variables that showed significant year-class effects or that could not be corrected for size effects. Age-2 samples (1988 year-class) provided the largest sample size across all stocks.

We tested the data sets for equality of covariance matrices. We used linear discriminant analysis if covariance matrices were equal and quadratic discriminant analysis if covariance matrices were unequal. Prior probabilities of group membership were assumed equal rather than proportional to sample size. We estimated the classification matrix by using jackknifing. Analyses were conducted using Statistical Analysis System version 6.03 (SAS Institute 1988).

Performance of the discriminant functions was evaluated via two measures: (1) the proportion of samples correctly classified to stock of origin; and (2) bias in stock composition estimates. We used the "misallocation statistic" (c) of Prager and Fabrizio (1990) as an index of bias in the stock composition estimates. The statistic varies between 0 (no bias) and 1 (maximum possible bias). Proportionate stock membership was estimated as the sum over all fish of the predicted probabilities of group membership. We developed confidence intervals (CIs) for the misallocation statistic and for stock composition estimates by bootstrapping (Efron 1982). The CIs included 95% of the outcomes of 400 bootstrap replicates.

#### Results

Three-Stock Discrimination, All Ages

Eight variables were selected for discrimination of three stocks with all year-classes (Table 1): six Fourier coefficients (four amplitude and two phase), rectangularity, and one angle measurement (left). With use of linear discriminant analysis, the overall correct classification rate for the three stocks was 56.6% (Table 2). The highest correct classification rate was for the Roanoke River stock

TABLE 2.—Classification matrices from five sets of discriminant analysis of known-origin samples of striped bass. Numbers with asterisks represent samples classified to their correct stock.

|                | Assigned stock |                |                    |                |                  |       |     |
|----------------|----------------|----------------|--------------------|----------------|------------------|-------|-----|
| Source stock   | Chesapeake Bay |                | Hudson River       |                | Roanoke River    |       | =   |
|                | Number         | %              | Number             | %              | Number           | %     | N   |
| _              |                | Three stoc     | ks, all ages (56.6 | % correct over | ali)             |       |     |
| Chesapeake Bay | 98*            | 53.3*          | 47                 | 25.5           | 39               | 21.2  | 184 |
| Hudson River   | 29             | 24.8           | 66*                | 56.4*          | 22               | 18.8  | 117 |
| Roanoke River  | 7              | 15.6           | 6                  | 13.3           | 32*              | 71.1* | 45  |
| Total          | 134            |                | 119                |                | 93               |       | 346 |
|                |                | Three sto      | cks, age 2 (60.5%  | correct overa  | II)              |       |     |
| Chesapeake Bay | 28*            | 56.0*          | 9                  | 18.0           | 13               | 26.0  | 50  |
| Hudson River   | 10             | 32.3           | 20*                | 64.5*          | 1                | 3.2   | 31  |
| Roanoke River  | 9              | 27.3           | 3                  | 9.1            | 21*              | 63.6* | 33  |
| Total          | 47             |                | 32                 |                | 35               |       | 114 |
|                | Chesape        | ake Bay versus | Hudson River, a    | ll ages (71.8% | correct overall) |       |     |
| Chesapeake Bay | 139*           | 75.5*          | 45                 | 24.5           |                  |       | 184 |
| Hudson River   | 40             | 34.2           | 77*                | 65.8*          |                  |       | 117 |
| Total          | 179            |                | 122                |                |                  |       | 301 |
|                | Chesap         | eake Bay versu | s Hudson River,    | age 2 (75.3% d | correct overall) |       |     |
| Chesapeake Bay | 38*            | 76.0*          | 12                 | 24,0           |                  |       | 50  |
| Hudson River   | 8              | 25.8           | 23*                | 74.2*          |                  |       | 31  |
| Total          | 46             |                |                    | 35             |                  |       | 81  |
|                | Hudso          | n River versus | Roanoke River, a   | ige 2 (84.4% c | orrect overall)  |       |     |
| Hudson River   |                |                | 27*                | 87.1*          | 4                | 12.9  | 31  |
| Roanoke River  |                |                | 6                  | 18.2           | 27*              | 81.8* | 33  |
| Total          |                |                | 33                 |                | 31               |       | 64  |

(71.1%); only 56.4% of Hudson River and 53.3% of Chesapeake Bay fish were correctly identified. The discriminant function performed better in terms of bias in stock composition estimates (c = 0.03; 95% CI = 0.01-0.06). The contributions of the Hudson River and Roanoke River stocks were overestimated by 4% and 6%, respectively, and the Chesapeake Bay stock was underestimated by 10% (Table 3).

# Three-Stock Discrimination, Age 2

Using only age-2 fish allowed us to include three Fourier coefficients that showed significant year-class effects and it eliminated the need to correct most variables for size effects. Other changes in the variable set occurred as a result of reevaluating the selection criteria on this smaller data set (Table 1). The final variable set for discrimination included three amplitude and five phase Fourier coefficients, rectangularity, two angle measurements, and three circulus pattern variables (variance in the width of the first five circuli, variance in the intercirculus spacing of circuli 6 through 10, and average width of circuli 6 through 10).

The overall correct classification rate based on

linear discriminant analysis was 60.5%. The rate for the Chesapeake Bay (56.0%) was lower than for the Hudson River (64.5%) and the Roanoke River (63.6%) (Table 2). Bias in stock composition estimates (c=0.13; 95% CI = 0.02-0.28) was higher than for the three-stock problem involving all age-classes. The proportional contribution of the Chesapeake Bay stock was overestimated by 18%, and the Hudson River and Roanoke River contributions were underestimated by 7% and 11%, respectively (Table 3).

# Chesapeake Bay versus Hudson River, All Ages

Fifteen variables were used in the Chesapeake Bay versus Hudson River, multi-age-class analysis: eight amplitude and five phase descriptors, one angle measurement, and angular asymmetry (Table 1). The overall correct classification rate by quadratic discriminant analysis was 71.8% (Hudson River, 65.8%; Chesapeake Bay, 75.5%; Table 2). Bias in the stock composition estimates was low (c = 0.01; 95% CI = 0-0.03) and only 1% of each stock was misallocated (Table 3).

TABLE 3.—Stock composition estimates (95% confidence limits are in parentheses) from five sets of discriminant analyses of striped bass from three stocks:  $P_j$  = true proportion in stock j;  $\hat{P}_j$  = estimated proportion in stock j based on predicted probabilities of stock membership;  $\Delta$  = difference between actual and estimated proportions; c = misal-location statistic, ranging from 0 to 1. Statistics were calculated from 400 bootstrap replicates.

| Stock and                   |                      | ersus Hudson River Chesapeake Bay versus<br>noke River Hudson River |                     |                      |
|-----------------------------|----------------------|---|---------------------|----------------------|
| statistic                   | All ages             | Age 2   | All ages            | Age 2                |
|                             |                      | Proportionate stock member  | ership              |                      |
| Chesapeake Bay              |                      |   |                     |                      |
| $P_{i}$                     | 0.53 (0.48, 0.58)    | 0.44 (0.35, 0.53)   | 0.61 (0.54, 0.66)   | 0.62 (0.51, 0.73)    |
| $oldsymbol{P_j}{\hat{P_j}}$ | 0.43 (0.39, 0.47)    | 0.62 (0.39, 0.80)   | 0.60 (0.55, 0.64)   | 0.64 (0.52, 0.78)    |
| Δ                           | -0.10 (-0.09, -0.11) | 0.18 (0.04, 0.27)   | -0.01 (0.01, -0.02) | 0.02 (0.01, 0.05)    |
| Hudson River                |                      |   |                     |                      |
| $P_{j}$                     | 0.34 (0.29, 0.39)    | 0.27 (0.20, 0.36)   | 0.39 (0.33, 0.46)   | 0.38 (0.27, 0.49)    |
| $\hat{P_j}$                 | 0.38 (0.34, 0.42)    | 0.20 (0.05, 0.38)   | 0.40 (0.36, 0.45)   | 0.36 (0.22, 0.48)    |
| Δ                           | 0.04 (0.05, 0.03)    | -0.07 (-0.15, -0.02)  | 0.01 (0.03, -0.01)  | -0.02 (-0.05, -0.01) |
| Roanoke River               |                      |   |                     |                      |
| $P_{j}$                     | 0.13 (0.09, 0.16)    | 0.29 (0.21, 0.38)   |                     |                      |
| $\hat{P}_{j}$               | 0.19 (0.14, 0.24)    | 0.18 (0.07, 0.34)   |                     |                      |
| Δ                           | 0.06 (0.05, 0.08)    | -0.11 (-0.14, -0.04)  |                     |                      |
|                             |                      | Misallocation   |                     |                      |
| Misallocation               |                      |   |                     |                      |
| statistic (c)               | 0.03 (0.01, 0.06)    | 0.13 (0.02, 0.28)   | 0.01 (0.0, 0.03)    | 0.03 (0.0, 0.12)     |

## Chesapeake Bay versus Hudson River, Age 2

Nine variables were included in the single-yearclass analysis of the Chesapeake Bay and Hudson River stocks: four amplitude and three phase coefficients, rectangularity, and average width of the first five circuli (Table 1). Quadratic discriminant analysis resulted in correct classification of 76.0% of Chesapeake Bay and 74.2% of Hudson River samples (75.3% overall; Table 2). Bias in the resulting stock composition estimates was low (c = 0.03; 95% CI = 0-0.12), and 2% of the samples were misallocated between stocks (Table 3).

# Hudson River versus Roanoke River, Age 2

We used 10 variables in the analysis of Hudson River versus Roanoke River, including three amplitude coefficients, four phase coefficients, rectangularity, and right and left angles (Table 1). By quadratic discriminant analysis, 87.1% of the Hudson River and 81.8% of the Roanoke River fish were correctly identified, for an overall correct rate of 84.4% (Table 2). The stock composition estimates had low bias (c = 0.04; 95% CI = 0-0.14) and 3% of the samples were misallocated between the stocks (Table 3).

#### Discussion

The precision of discriminant analysis based on striped bass scale morphology varied widely

among comparisons. The two analyses that included all three stocks had correct rates averaging only 57% and 61%. Although better than random, such rates are too low for three-stock discrimination to be practical for management. Exclusion of the Roanoke River stock added about 15% to correct classification rates, bringing them into the range of other stock identification studies of Chesapeake Bay and Hudson River striped bass (Waldman et al. 1988). The best overall rate (84%) was achieved for the comparison of Hudson River with Roanoke River fish, which involved the most geographically distant stocks considered.

Although most stock identification studies focus on correct classification rates, the adequacy of a stock identification method depends also on the stock composition estimates produced. If correct classification rates are low, but misclassifications are balanced, the estimate of stock composition may be accurate even though individual fish are not always classified accurately. This point is well illustrated by our results for the Hudson-Chesapeake analysis of age-2 fish. The overall correct classification rate was only 75.3%; however, the estimates of stock composition were very close to the true values (Table 3). Because stock composition estimates are the ultimate goal of discrimination, the distribution of misallocations is a key issue (Prager and Fabrizio 1990). Stock compo-

TABLE 3.—Extended.

| Stock and                                | Hudson River versus     |  |  |
|--|-------------------------|--|--|
| statistic                                | Roanoke River, age 2    |  |  |
| Proport                                  | ionate stock membership |  |  |
| Chesapeake Bay                           |                         |  |  |
| $P_{j}$                                  |                         |  |  |
| $\hat{P}_j$ $\hat{P}_j$                  |                         |  |  |
| Δ  |                         |  |  |
| ludson River                             |                         |  |  |
| $P_{j}$                                  | 0.48 (0.37, 0.60)       |  |  |
| $oldsymbol{P_j}{\hat{oldsymbol{eta}_j}}$ | 0.51 (0.34, 0.66)       |  |  |
| Δ  | 0.03 (0.03, 0.06)       |  |  |
| oanoke River                             |                         |  |  |
| $P_{j}$                                  | 0.52 (0.40, 0.63)       |  |  |
| $\hat{P}_j$ $\hat{P}_j$                  | 0.49 (0.34, 0.66)       |  |  |
| Δ  | -0.03 (-0.06, 0.03)     |  |  |
|  | Misallocation           |  |  |
| 1isallocation                            |                         |  |  |
| statistic (c)                            | 0.04 (0.0, 0.14)        |  |  |

sition estimates resulting from our discriminant analyses were quite accurate. In four of five problem sets, misallocation statistics (c) were 0.04 or lower, and most estimated stock compositions differed very little from true compositions.

Even more important than the observed pattern of misallocations, however, is potential variability in those patterns. Bias in stock composition estimates can be corrected by using a classification matrix for known-origin samples (e.g., by following methods outlined by Van Winkle et al. 1988). However, fish in an unknown sample will necessarily differ from those used to develop the discriminant function and its correction. Thus an important issue is consistency of bias from sample to sample. Consistency presumably depends on the representativeness of the known-origin samples; if the sample is adequate, variability in the bias estimates should be low. We attempted to address this issue for our samples by bootstrapping confidence intervals for the stock composition estimates and misallocation statistics. The confidence limits for the estimates were quite narrow, suggesting that bias correction could be applied with some assurance. In four of the five problem sets, confidence limits for the estimates showed a spread similar to the true (bootstrap) proportions, suggesting that the pattern of misallocations was fairly stable. These results are undoubtedly somewhat optimistic, however, because bootstrap replicates do not represent completely independent samples.

Based on results of other stock identification studies that incorporated scale shape analysis (Jarvis et al. 1978; Casselman et al. 1981; DePontual and Prouzet 1987; Riley and Carline 1982; Margraf and Riley 1993), we expected to see higher rates of correct classification for Atlantic striped bass. However, rates reported in some previous studies may be optimistic. For example, Jarvis et al. (1978) reported 80-100% correct classification of walleyes Stizostedion vitreum from two eastern Lake Erie stocks. Correct classification rates were estimated by resubstitution (i.e., by using the same samples used to estimate the discriminant function), which is known to bias correct classification rates upward (Lachenbruch and Mickey 1968). For comparison with Jarvis et al., we estimated a discriminant function for our age-2 Chesapeake Bay and Hudson River samples using the first 12 harmonics of the Fourier series and compared correct rates obtained by resubstitution and jackknifing. With jackknifing, the overall correct classification rate was 65%; with resubstitution, the correct rate rose to 100%.

Two other studies of striped bass have achieved moderate success in discriminating stocks according to scale morphology. Margraf and Riley (1993) used Fourier analysis to discriminate Hudson River and Chesapeake Bay stocks with 80% correct classification. Ross and Pickard (1990) correctly classified 73% of wild and hatchery Pacific striped bass using Fourier coefficients, and 90% when circulus features were added to the analysis. Our results from using multiple scale features were similar to these, although circulus features were not as powerful in our analysis as in Ross and Pickard's. Greater differences in circulus patterns might be expected between wild and hatchery stocks than among wild stocks.

Why scale morphology does not provide a more distinct stock-specific signal for wild striped bass is not clear. If striped bass stocks return to natal estuarine areas to spawn, a high degree of genetic isolation is possible. However, mitochondrial DNA studies of Atlantic striped bass show unusually low levels of base sequence heterogeneity (Wirgin et al. 1990, 1993), suggesting that little genetic differentiation has occurred. Environmental effects may be more important in producing the subtle differences in scale morphology that allow some discrimination. We found significant year-class effects in 28% of our size-corrected vari-

ables, suggesting that environmental factors have a substantial influence.

In a study of otoliths from Atlantic cod Gadus morhua, Campana and Casselman (1993) concluded that variation in otolith shape was due primarily to differences in growth rates among stocks. If striped bass scale shape is similarly influenced by growth rate, it is not clear whether one can expect stock discrimination to be successful, because the evidence is contradictory regarding the degree and direction of growth rate differences among Atlantic striped bass stocks (Setzler et al. 1980; Coutant 1985; ASMFC 1990; Conover 1990).

Although discrimination based on scale morphology did not consistently provide high correct classification rates for wild Atlantic striped bass, it provided accurate estimates of stock composition in four of the five analyses we conducted. Thus the utility of scale morphology hinges on the stability of bias in stock composition estimates (i.e., on how reliably corrections can be made based on the classification matrix). Our results suggest the bias may be quite stable; however, this should be more stringently tested on independent samples of known-origin fish, because the bootstrapping technique we used is probably optimistic.

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