

Abstract—A total of 1006 king mackerel (*Scomberomorus cavalla*) representing 20 discrete samples collected between 1996 and 1998 along the east (Atlantic) and west (Gulf) coasts of Florida and the Florida Keys were assayed for allelic variation at seven nuclear-encoded microsatellites. No significant deviations from Hardy-Weinberg equilibrium expectations were found for six of the microsatellites, and genotypes at all microsatellites were independent. Allele distributions at each microsatellite were independent of sex and age of individuals. Homogeneity tests of spatial distributions of alleles at the microsatellites revealed two weakly divergent “genetic” subpopulations or stocks of king mackerel in Florida waters—one along the Atlantic coast and one along the Gulf coast. Homogeneity tests of allele distributions when samples were pooled along seasonal (temporal) boundaries, consistent with the temporal boundaries used currently for stock assessment and allocation of the king mackerel resource, were nonsignificant. The degree of genetic divergence between the two “genetic” stocks was small: on average, only 0.19% of the total genetic variance across all samples assayed occurred between the two regions. Cluster analysis, assignment tests, and spatial autocorrelation analysis did not generate patterns that were consistent with either geographic or spatial-temporal boundaries. King mackerel sampled from the Florida Keys could not be assigned unequivocally to either “genetic” stock. The genetic data were not consistent with current spatial-temporal boundaries employed in stock assessment and allocation of the king mackerel resource. The genetic differences between king mackerel in the Atlantic versus those in the Gulf most likely stem from reduced gene flow (migration) between the Atlantic and Gulf in relation to gene flow (migration) along the Atlantic and Gulf coasts of peninsular Florida. This difference is consistent with findings for other marine fishes where data indicate that the southern Florida peninsula serves (or has served) as a biogeographic boundary.

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Population structure of king mackerel (*Scomberomorus cavalla*) around peninsular Florida, as revealed by microsatellite DNA*

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The king mackerel (*Scomberomorus cavalla*) is a coastal pelagic fish distributed in the western Atlantic Ocean from Massachusetts (USA) to Rio de Janeiro (Brazil) and in the Gulf of Mexico and Caribbean Sea (Rivas, 1951; Collette and Nauen, 1983). Recreational and commercial catches of king mackerel in U.S. waters are substantial (Manooch, 1979; MSAP¹; Legault et al.²) and the species is critical to the southeastern Atlantic coast (hereafter “Atlantic”) and northern Gulf of Mexico (hereafter “Gulf”) charter-boat industries. Management of the king mackerel resource in U.S. waters is under the jurisdiction of two regional fishery management councils, and current management planning (GMFMC³) is based on a two migratory unit (stock) hypothesis: one stock (Atlantic migratory unit) occurs in the U.S. southeastern Atlantic, the other (Gulf of Mexico migratory unit) occurs in the Gulf. Separation of the two stocks has been based primarily on mark-and-recapture studies carried out prior to 1984 and on differences in growth rate (MSAP¹; Williams and Godcharles⁴). Based on the mark-and-recapture studies, it was hypothesized that the two stocks mixed extensively during

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¹ MSAP (Mackerel Stock Assessment Panel). 1994. Contribution report of the mackerel stock assessment panel. Contribution report MIA-93/94-42, 27 p. Southeast Fishery Science Center, National Marine Fisheries Service, 75 Virginia Beach Dr., Miami, FL 33149.

² Legault, C. M., M. Ortiz, G. Scott, N. Cummings, and P. Phares. 2000. Stock assessment analyses on Gulf of Mexico king mackerel. Contribution report SFD-99/00-83, 48 p. Southeast Fishery Science Center, National Marine Fisheries Service, 75 Virginia Beach Dr., Miami, FL 33149.

³ GMFMC (Gulf of Mexico Fishery Management Council). 1996. Amendment 8 to the fishery management plan for coastal migratory pelagic resources in the Gulf of Mexico and South Atlantic includes environmental assessment, regulatory impact review, and initial regulatory flexibility analysis. Gulf of Mexico Fishery Management Council, 3018 U.S. Hwy, 301 North, Suite 1000, Tampa, FL 33619-2266.

⁴ Williams, R. O., and M. F. Godcharles. 1984. Completion report, king mackerel tagging and stock assessment. Project rep. 2-341-R, plus figures and tables., 45 p. FL Dept. Nat. Res., Florida Marine Research Institute, 100 8th Ave. SE, St. Petersburg, FL 33701.

the winter months along the southeast coast of Florida. For purposes of stock assessment and resource allocation (Sutter et al., 1991; GMFMC⁵), the boundary between the two stocks was specified as the Volusia/Flagler county line (east coast of Florida) in winter (1 November–31 March) and the Monroe/Collier county line (west coast of Florida) in summer (1 April–31 October). Pragmatically, this means that king mackerel caught south of the Volusia/Flagler county line (including the Florida Keys) between 1 November and 31 March belong to Gulf stock, whereas fish caught south of the Monroe/Collier county line between 1 April and 31 October belong to Atlantic stock.

Data from additional mark-capture (Fable et al., 1987; Sutter et al., 1991; Schaefer and Fable, 1994; Fable⁶), growth rate (DeVries and Grimes, 1997), otolith shape (DeVries et al., 2002), and temporal-geographic sampling studies (Collins and Stender, 1987; Trent et al., 1983; Grimes et al., 1990) are consistent with the hypothesis that king mackerel in the Atlantic differ from those in the Gulf. In addition, on the basis of allozyme evidence (Johnson et al., 1993) and studies of early life history (Grimes et al., 1990; Grimes et al.⁷), DeVries and Grimes (1997) suggested that there might be two distinct stocks of king mackerel in the northern Gulf. Johnson et al. (1993) found a high frequency of the *PEPA-2a* allele of the nuclear-encoded dipeptidase (*PEPA-2*) locus among king mackerel from the western and northwestern Gulf, whereas a high frequency of the *PEPA-2b* allele occurred among king mackerel from the Atlantic and northeastern Gulf. Johnson et al. (1993) hypothesized that the two (putative) Gulf stocks mixed to varying degrees in the northern Gulf. Considering all the data acquired to date, DeVries and Grimes (1997) suggested there may be three stocks of king mackerel in U.S. waters: one in the Atlantic, one in the eastern Gulf, and one in the western Gulf.

The allozyme data of Johnson et al. (1993) did not distinguish king mackerel in the eastern Gulf from those in the Atlantic, and to that extent, argued against the hypothesis that king mackerel in the Atlantic and Gulf represented two distinct stocks. Gold et al. (1997), however, assayed variation in restriction sites of mitochondrial (mt)DNA among king mackerel collected from 13 localities along the U.S. Atlantic

coast and northern Gulf and found significant (but weak) heterogeneity only in comparisons of pooled mtDNA haplotypes from Atlantic localities with pooled haplotypes from Gulf localities. Thus, the mtDNA data did not support the hypothesis that two genetically identifiable stocks of king mackerel occur in the northern Gulf, but rather were consistent with the hypothesis that separate stocks of king mackerel may exist in the Atlantic and in the Gulf. Estimates of F_{ST} , a measure of population subdivision, between king mackerel in the Atlantic and Gulf were small, indicating that mixing between Atlantic and Gulf king mackerel occurs.

Gold et al. (1997) also examined spatial variation in frequencies of the two alleles at *PEPA-2*. Results were essentially the same as those reported by Johnson et al. (1993): high frequency of the *PEPA-2a* allele among king mackerel in the western Gulf and high frequency of the *PEPA-2b* allele in the eastern Gulf and Atlantic. Tests of independence of *PEPA-2* genotypes with age and sex of individual fish, however, revealed significant nonrandom associations among Gulf fish of *PEPA-2a* homozygous genotypes with males and of *PEPA-2b* homozygous genotypes with females. Moreover, among fish sampled from the Atlantic, there was a highly significant decrease in the frequency of *PEPA-2b* alleles with increasing fish age. The same trend was found among fish sampled from the Gulf, but to a lesser extent. Tests of independence of sex versus age, and of mtDNA variation versus sex or age, were nonsignificant. These findings strongly indicated that the use of *PEPA-2* genotypes to distinguish stocks of king mackerel is compromised and that the hypothesis of eastern and western stocks of king mackerel in the Gulf needs to be re-evaluated.

Finally, Broughton et al. (2002) surveyed allelic variation at five nuclear-encoded microsatellites among a subset of the samples of king mackerel studied by Gold et al. (1997). Tests of homogeneity in allele distribution at the five microsatellites indicated that samples from Port Aransas, Texas (western Gulf), and Gulfport, Mississippi (central Gulf), differed from each other and from the remaining samples (including two samples from the Atlantic, one from the Florida Keys, one from the eastern Gulf, one from the western Gulf, and one from Veracruz, Mexico). No significant differences in allele frequencies at any microsatellite were found between samples representing geographic extremes, and no significant geographic patterns were found when samples were combined into regional groupings reflecting current hypotheses of king mackerel stock structure in U.S. waters.

Of concern to management of the king mackerel resource in U.S. waters is the degree of mixing between the presumed stocks in the Atlantic and Gulf. Analysis of mark-and-recapture data collected from 1985 to 1993 (MSAP¹) indicated that roughly 3.0% of fish tagged in the Atlantic were recaptured in the Gulf, whereas 6.4% tagged in the Gulf were recovered in the Atlantic. More liberal estimates (SFC⁸) of recaptures (generated when utilizing summer

⁵ GMFMC (Gulf of Mexico Fishery Management Council). 1984. Final amendment 1, fishery management plan and environmental impact statement for coastal migratory pelagic resources (mackerels) in the Gulf of Mexico and South Atlantic region. Gulf of Mexico Fishery Management Council, 3018 U.S. Hwy, 301 North, Suite 1000, Tampa, FL 33619-2266.

⁶ Fable, W. A., Jr. 1988. Stock identification of king mackerel based on mark-recapture. Unpubl. manuscript from meeting on stock identification of king mackerel in the Gulf of Mexico. Southeast Fish. Sci. Center Contribution Report 2-18-88, 24 p. Southeast Fisheries Science Center, 3500 Delwood Beach Road, Panama City, FL 32408.

⁷ Grimes, C. B., J. H. Finucane, and L. A. Collins. 1988. Distribution and occurrence of young king mackerel, *Scomberomorus cavalla*, in the Gulf of Mexico. Unpubl. manuscript from meeting on stock identification of king mackerel in the Gulf of Mexico. Panama City Lab. Contribution Report 2-18-88, 31 p. NMFS SE Fish. Cntr., 3500 Delwood Beach Road Panama City, FL 32408.

⁸ SFC (Southeastern Fisheries Center). 1992. Preliminary analysis of southeastern U.S. king mackerel mark-recapture data: 1985–1993. Contribution report MIA-93/94-36, 19 p. Southeast Fisheries Science Center, National Marine Fisheries Service, 75 Virginia Beach Dr., Miami, FL 33149.

and winter seasons in the “mixing” zone) suggested that 2.6–30.9% of recaptured tagged-fish in the Atlantic were returned as Gulf fish and 1.5–13.6% of recaptures tagged in the Gulf were returned as Atlantic fish. These mixing rates, however, were questioned (Jones et al.⁹) because virtual population analysis (VPA) based estimates of fishing mortality for the directed king mackerel fisheries in the Gulf and Atlantic corresponded to annual exploitation rates of 0.30 and 0.11, respectively, whereas exploitation rates calculated from the 1985–93 (uncorrected) tag returns ranged from 0.027 to 0.033 (Gulf) and from 0.036 to 0.045 (Atlantic). The difference between the two estimates of exploitation rates implied either that the true exploitation rate was overestimated by VPA or underestimated by uncorrected tag-return data, leading to the conclusion (Jones et al.⁹) that little confidence should be placed in reported mixing rates based on mark-and-recapture data.

The goals of this project were to use nuclear-encoded microsatellites to define more rigorously the spatial-temporal limits of the two stocks (if separate stocks exist) and to estimate the proportions of both stocks in the mixing zone. The issues of spatial-temporal limits and mixing of the two (presumed) stocks are important in relation to assessing and allocating the king mackerel resource, particularly during the winter season. For example, mark-recapture data (MSAP¹) indicated that ~20% of fish tagged in the mixing zone in southeastern Florida moved into the Gulf. If this means that only ~20% of winter catches from the east coast of Florida are Gulf stock, as opposed to 100% under the current management plan, the allowable biological catch (ABC) for the Gulf stock would decrease significantly (MSAP¹). Because the Gulf stock of king mackerel currently is considered overfished (Legault et al.²) reductions in ABC of the Gulf stock could have significant economic impact.

The choice to employ microsatellites for the project was straightforward. Briefly, microsatellites are rapidly evolving, short stretches of DNA composed of di-, tri-, and tetranucleotide arrays that are abundant, highly polymorphic, and inherited in a codominant fashion (Weber, 1990; Wright, 1993; Wright and Bentzen, 1994). Because allele frequencies at microsatellites are generally consistent with equilibrium expectations of diploid, Mendelian loci and because identification of individual microsatellites is by polymerase-chain-reaction (PCR) amplification, both of which remove most of the problems associated with homology of alleles, microsatellites have proven to be useful genetic markers of population structure in numerous taxa, including fishes (Angers and Bernatchez, 1998; Ruzzante et al., 1996; O’Connell et al., 1998; Nielsen et al., 1999). In addition, new alleles at microsatellite loci appear to arise rapidly (Schug et al., 1998), generating high allelic diversity important for statistical power in exact tests and

other tests of allele-distribution homogeneity (Estoup et al., 1998; Ross et al., 1999).

Materials and methods

A total of 20 samples of king mackerel were procured between 1996 and 1998 from 11 different offshore sites (Table 1, Fig. 1). The sample from Panama City was obtained from charter boat catches, and the samples from Sarasota and Jacksonville were obtained from tournaments. The remaining samples were obtained from commercial catches. Tissue samples (heart and muscle) were removed from each fish, frozen in liquid nitrogen, transported to College Station, and stored at -80°C . Sex of individuals was recorded for all samples, except for the March 1997 sample from the Florida Keys. Approximate ages of individuals from all samples except for the July 1998 sample from Jacksonville, the March 1997 sample from the Florida Keys, and the April 1997 sample from Sarasota, were determined by otolith-increment analysis by following methods outlined in DeVries and Grimes (1997).

Initially, we planned to deploy the five microsatellites developed in a prior study (Broughton et al., 2002). Two of these (*Sca-8* and *Sca-47*), however, had proven difficult to amplify consistently in the prior study and therefore were omitted from our study. A third microsatellite, *Sca-30*, developed by Broughton et al. (2002), also was omitted because of difficulties with consistent amplification and because allele distributions at *Sca-30* were highly leptokurtic (Broughton et al., 2002). A total of five new microsatellites was then developed from the microsatellite-enriched genomic library generated by Broughton et al. (2002). Candidate microsatellites were sequenced from either or both ends by using standard M13 sequencing primers and an Applied Biosystems (Perkin Elmer) 377 automated DNA sequencer. The OLIGO software package (National Biosciences, Inc., 1992) was used to identify primers from regions flanking microsatellites. Primers were designed according to preset criteria that included product length, internal stability, proportion GC content, and primer T_m difference. PCR amplifications were performed under a variety of experimental conditions to optimize procedures that produced high yields of target sequence and minimized additional fragments (“stutter” bands). Experimental tractability (reproducibility, consistency, range in allele size, frequency of “stutter” bands [if present], and microsatellite polymorphism) of PCR-amplified microsatellites were evaluated by screening a panel of king mackerel samples available in the laboratory. PCR primer sequences, the length (in base pairs) of the cloned allele, and the annealing temperature in PCR amplification for the seven microsatellites used in the project are given in Appendix Table 1. Two of these, *Sca-37* and *Sca-44*, were developed previously by Broughton et al. (2002).

For assay of individual fish, genomic DNA was isolated from frozen tissues as described in Gold and Richardson (1991). Genotypes at the seven microsatellites were determined by PCR amplification and gel electrophoresis. Prior

⁹ Jones, C. M., M. E. Chittenden, and J. R. Gold. 1994. Report to the mackerel stock identification working group. Unpubl. document of meeting held 8 Sep 94 to 9 Sep 94 at Panama City Lab., SE Fish. Sci. Cent. 7 p. Southeast Fisheries Science Center, National Marine Fisheries Service, 3500 Delwood Beach Road, Panama City, FL 32408.

Table 1

Localities, acronyms, dates of collection, and number of individuals (by sex) of king mackerel (*Scomberomorus cavalla*) sampled from the east and west coasts of Florida and the Florida Keys.

Sample locality	Acronym	Date of capture	Number of individuals			Migratory group ¹
			Female	Male	Total	
Atlantic Ocean (east coast)						
Jacksonville, FL	JCK ¹	Jul 1996	48	0	48	Atlantic
Jacksonville, FL	JCK ²	Jul 1998	28	3	31	Atlantic
New Smyrna Beach, FL	NSB	Jul 1996	41	9	50	Atlantic
Cape Canaveral, FL	CCN	Dec 1998	24	26	50	Gulf
Sebastian, FL	SEB ¹	Mar 1997	29	21	50	Gulf
Sebastian, FL	SEB ²	Mar 1998	24	26	50	Gulf
Sebastian, FL	SEB ³	Dec 1998	35	15	50	Gulf
Ft. Pierce, FL	FTP	Apr 1996	31	25	56	Atlantic
West Palm Beach, FL	WPB	May 1998	29	25	54	Atlantic
Florida Keys						
Key West, FL	KEY ¹	Mar 1996	41	10	51	Gulf
Key West, FL	KEY ²	Mar 1997	—	—	29	Gulf
Key West, FL	KEY ³	Jan 1999	29	19	48	Gulf
Gulf of Mexico (west coast)						
Marco Island, FL	MCI	Apr 1996	29	26	55	Atlantic
Boca Grande, FL	BCG	Apr 1996	31	4	35	Gulf
Sarasota, FL	SAR ¹	Apr 1996	39	5	44	Gulf
Sarasota, FL	SAR ²	Nov 1996	60	0	60	Gulf
Sarasota, FL	SAR ³	Apr 1997	55	2	57	Gulf
Sarasota, FL	SAR ⁴	Apr 1998	68	2	70	Gulf
Sarasota, FL	SAR ⁵	Nov 1998	62	6	68	Gulf
Panama City, FL	PCY	Oct 1996	25	25	50	Gulf

¹ Classification as Atlantic or Gulf migratory group based on the two migratory unit hypothesis where boundaries change seasonally. See text for further details.

to amplification, one of the primers was kinase-labeled with $\gamma^{32}\text{P}$ -ATP by T4 polynucleotide kinase (30 min, 37°C). PCR reactions contained approximately 5 ng of genomic DNA, 0.1 units of *Taq* DNA polymerase, 0.5 μM of each primer, 800 μM dNTPs, 1–2 mM MgCl_2 , 1X *Taq* buffer at pH 9.0 (Promega, Corp., Madison, WI), and sterile deionized water in a total volume of 10 μL . Thermal cycling was carried out in 96-well plates as follows: denaturation (45 sec, 95°C), annealing (30 sec, temperature as in Appendix Table 1), and polymerization (30 sec, 72°C) for 30 cycles. Aliquots (3 μL) of each PCR reaction were electrophoresed in 6% denaturing polyacrylamide (“sequencing”) gels. Gels were dried and exposed to x-ray film. Alleles at individual microsatellites were scored as number of repeats by comparison to the cloned (and sequenced) allele. Genotypes at each microsatellite for each individual were scored and entered into a database.

Initial statistical analysis involved generation of allele frequencies and (direct-count) heterozygosity values, and significance testing of genotypic proportions in relation

to those expected under conditions of Hardy-Weinberg equilibrium. Significance testing of Hardy-Weinberg equilibrium proportions involved exact tests performed using Markov-chain randomization (Guo and Thompson, 1992); probability (*P*) values for tests at each microsatellite within each sample were estimated by permutation (bootstrapping) with 1000 resamplings (Manly, 1991). Significance levels for simultaneous tests were adjusted with the sequential Bonferroni approach (Rice, 1989). Tests of genotypic equilibrium at pairs of microsatellites were carried out as a surrogate to assess whether any microsatellites were genetically linked. Probability values for (exact) tests of genotypic equilibrium were generated by 1000 resamplings, and significance levels for simultaneous tests were adjusted with the sequential Bonferroni approach. Allele frequencies and heterozygosity values were obtained by using BIOSYS-1.7 (Swofford and Selander, 1981), and tests of Hardy-Weinberg and genotypic equilibrium employed the package GENEPOP (Raymond and Rousset, 1995). Exact tests also were used to test independence

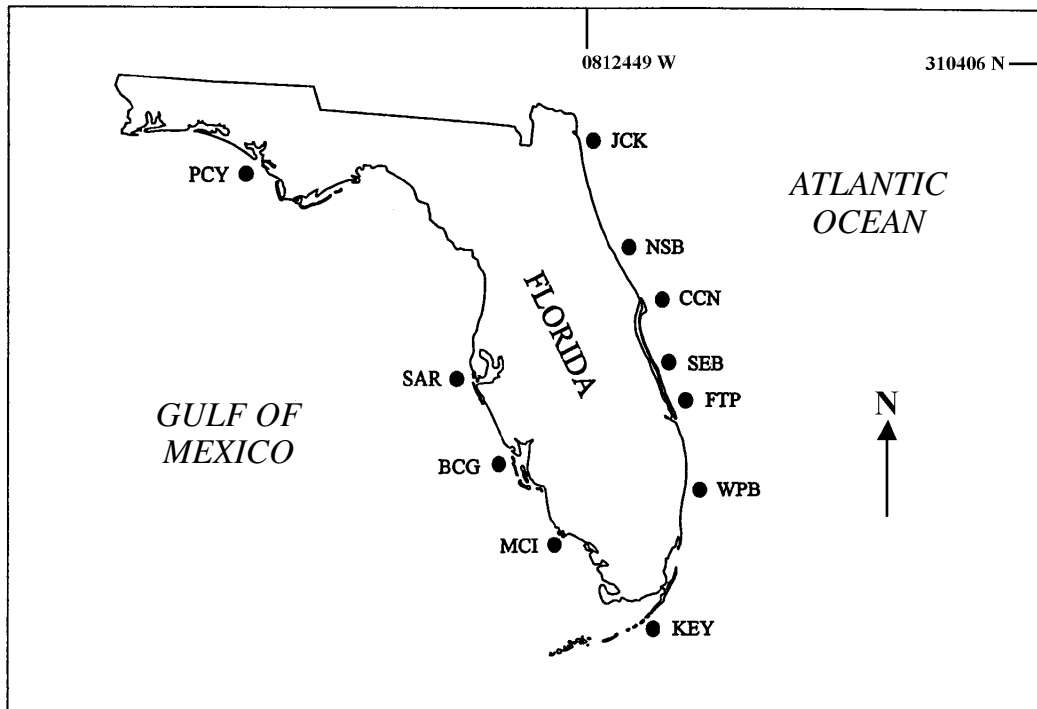


Figure 1

Sampling localities for king mackerel examined in the present study. Acronyms for sample localities are defined in Table 1.

of the distribution of genotypes at each microsatellite with the sex and age of individuals. Initial tests involved each of the 20 samples separately. We then pooled individuals sampled at Atlantic localities (nine samples), in the Florida Keys (three samples), at Gulf localities (eight samples), and over all localities (20 samples) in order to increase cell sizes in individual tests. Probability (P) values for these tests of independence were estimated by permutation (1000 resamplings) and significance levels for simultaneous tests were adjusted with the sequential Bonferroni approach.

Tests of genetic homogeneity among samples included exact tests, as implemented in GENEPOP, the Monte Carlo procedure of Roff and Bentzen (1989), as implemented in the restriction enzyme analysis package of McElroy et al. (1992), and the analysis of molecular variance (AMOVA) of Excoffier et al. (1992). Significance of tests of genetic homogeneity employed permutation with 1000 resamplings per individual comparison, and significance levels for simultaneous tests were adjusted by using the sequential Bonferroni approach. Tests of genetic homogeneity were carried out separately for each of the seven microsatellites. Individual tests were carried out 1) among all 20 samples, 2) among samples (nine) from Atlantic localities, 3) among samples (three) from the Florida Keys, and 4) among samples (eight) from Gulf localities. Analysis of molecular variance (AMOVA) was employed to generate estimates of (genetic) variance components and Φ statistics for the same comparisons. Φ statistics are a set of hierar-

chical F -statistic analogs that consider evolutionary distance among alleles. Significance of Φ statistics employed permutation (1000 resamplings).

Exact tests, the Monte Carlo procedure of Roff and Bentzen, and AMOVA also were used to assess genetic homogeneity 1) between samples from the Atlantic versus those from the Gulf (excluding samples from the Florida Keys), and 2) among samples from the Atlantic, the Florida Keys, and the Gulf. This design was chosen *a priori*, in part because it was geographically logical, in part because the southern Florida peninsula apparently serves (or has served) as a biogeographic boundary for a number of marine species (Aulsebrook, 1992; Gold and Richardson, 1998). The hierarchical capability of AMOVA also permitted a test of homogeneity among samples within the three regional groupings. We also carried out homogeneity testing to examine the temporal stock boundaries currently used in management planning for the king mackerel resource. Each of the 20 samples of king mackerel was designated as either Atlantic or Gulf stock according to the time of year during which they were sampled. Six of the samples thus were designated Atlantic stock and 14 of the samples were designated Gulf stock (Table 1). Genetic homogeneity was then tested between the Atlantic and Gulf "stocks," by using exact tests and the Monte Carlo procedure of Roff and Bentzen.

Clustering of genetic distances, spatial autocorrelation analysis, and assignment tests also were employed to assess temporal and spatial variation of microsatellites.

Table 2

Summary of variation in seven microsatellites among king mackerel (*Scomberomorus cavalla*) sampled from the east and west coasts of Florida and the Florida Keys.

Microsatellite	Repeat sequence	No. of alleles	Average heterozygosity \pm SE	P_{HW}^1
<i>Sca-14</i>	(CA) ₆ TA (CA) ₁₃	5	0.474 \pm 0.016	0/20
<i>Sca-23</i>	(CA) ₄ AAC (AG) ₁₂	24	0.803 \pm 0.105	4/20 ²
<i>Sca-37</i>	(TG) ₈ AG (TG) ₄ AG (TG) ₄	9	0.509 \pm 0.014	0/20
<i>Sca-44</i>	(CTCG) ₂ CTAT (CTGT) ₅	8	0.677 \pm 0.014	0/20
<i>Sca-49</i>	(TG) ₁₇	15	0.656 \pm 0.018	0/20
<i>Sca-61</i>	(CA) ₆ TGTA (CA) ₈	6	0.311 \pm 0.018	0/20
<i>Sca-65</i>	(TG) ₁₃	24	0.798 \pm 0.016	0/20

¹ Proportion of samples where $P < 0.05$, after Bonferroni correction.

² Probability values for three samples (SEB², KEY¹, and SAR⁴) were 0.000. The probability value for SAR¹ was 0.006 (adjusted α was 0.003).

Cavalli-Sforza's chord distance (Cavalli-Sforza and Edwards, 1967), as implemented in the GENEDIST program in version 3.4 of the phylogenetic inference package (PHYLIP) of Felsenstein (1992), was used to estimate the degree of genetic divergence or similarity between pairs of samples; neighbor joining (Saitou and Nei, 1987), from the NEIGHBOR program in PHYLIP, was used to cluster the resulting genetic distance matrix. A consensus of 500 neighbor-joining topologies was constructed by using the CONSENSE program in PHYLIP. Spatial autocorrelation analysis was carried out to determine whether allele distributions at each microsatellite at any given sample locality were independent of allele distributions in adjacent localities. Briefly, autocorrelation coefficients (Moran's I values), generated as a function of geographic distance between pairs of sample localities, were used to summarize patterns of geographic variation of allele frequencies at each microsatellite. Positive autocorrelations between adjacent localities, with decreasing autocorrelation as geographic distance between localities increases, are generally interpreted as an isolation-by-distance effect (Sokal and Oden, 1978a). We employed the spatial autocorrelation analysis program (SAAP) of Wartenberg (1989) and followed procedures outlined in Sokal and Oden (1978a, 1978b). "Noise" was minimized by including only alleles that occurred 20 or more times in the data set. The first of two SAAP runs employed equal geographic distances between each of five distance classes; the second employed equal numbers of pairwise comparisons in each distance class. Finally, assignment tests (Paetkau et al., 1995, 1997) were used to "assign" individuals within each of the 20 samples to one of two regional (spatial) groupings, Atlantic or Gulf. The two groupings were employed largely as a result of homogeneity tests of allele distributions, where existence of the two spatial groupings was weakly supported. Assignment tests have a number of uses (Waser and Strobeck, 1998): in this case we were interested in the proportion of individuals within a sample that could be assigned to each regional group, in relation to the locality of the sample and the season in which it was procured. Assignment tests were carried out employ-

ing the "assignment calculator" software available at <http://www.biology.ualberta.ca/jbruzusto/Doh.html>.

Results and discussion

Allele frequencies at the seven microsatellites in each of the 20 samples are given in Appendix Tables 2 and 3; number of individuals assayed, heterozygosity (direct count) values, and probability of conformance to expected Hardy-Weinberg proportions per microsatellite per individual sample are given in Appendix Tables 4 and 5. Summary statistics are given in Table 2 and include 1) repeat sequence of the cloned allele, 2) number of alleles detected, 3) average (direct count) heterozygosity (\pm SE) observed among samples, and 4) results of tests of conformance of observed genotype proportions to expectations of Hardy-Weinberg equilibrium. Cloned alleles at the seven microsatellites included simple (*Sca-49*, *Sca-65*) and complex (*Sca-14*, *Sca-23*, *Sca-37*, *Sca-61*) dinucleotide repeats and one complex tetranucleotide repeat (*Sca-44*). All dinucleotide microsatellites included CA (or complementary TG) repeats, with the number of alleles per microsatellite ranging from five (*Sca-14*) to twenty-four (*Sca-23* and *Sca-65*). Direct count heterozygosity, averaged over the twenty samples, ranged from 0.311 \pm 0.018 (*Sca-61*) to 0.803 \pm 0.105 (*Sca-23*). These results indicate that the seven microsatellites assayed in king mackerel are typical of microsatellites found in other vertebrate organisms, including fishes (e.g. DeWoody and Avise, 2000; Turner et al., 1998; Gold et al., 2001).

After sequential Bonferroni correction (Rice, 1989), genotype proportions at six of the microsatellites in all twenty samples did not deviate significantly from proportions expected under Hardy-Weinberg equilibrium. Genotype proportions at *Sca-23* among three of the samples (SEB², KEY¹, and SAR⁴) differed significantly ($P=0.000$) from Hardy-Weinberg equilibrium expectations, and at a fourth sample (SAR¹), the probability value of 0.006 was very close to the Bonferroni adjusted alpha of 0.003 (Appendix

Table 3

Probability of genotype equilibrium (pairwise comparisons) among seven microsatellite loci in king mackerel (*Scomberomorus cavalla*) sampled from the east and west coasts of Florida and the Florida Keys. Corrected α (for initial test) = 0.002.

Microsatellite	<i>Sca-14</i>	<i>Sca-23</i>	<i>Sca-37</i>	<i>Sca-44</i>	<i>Sca-49</i>	<i>Sca-61</i>	<i>Sca-65</i>
<i>Sca-14</i>	—	0.389	0.684	0.900	0.665	0.977	0.906
<i>Sca-23</i>		—	0.022	1.000	0.805	0.603	0.194
<i>Sca-37</i>			—	0.724	0.876	0.910	0.430
<i>Sca-44</i>				—	0.328	0.000	0.020
<i>Sca-49</i>					—	0.295	0.998
<i>Sca-61</i>						—	0.873
<i>Sca-65</i>							—

Table 4

Results of tests for spatial homogeneity in allele distribution of seven microsatellites among king mackerel (*Scomberomorus cavalla*) sampled from the east and west coasts of Florida and the Florida Keys. P_{EXACT} = probability based on Fisher's exact tests, with 1000 permutations. P_{RB} = probability based on 1000 bootstrapped replicates (after Roff and Bentzen 1989).

Test group	All samples ¹		Atlantic localities ²		Florida Keys ³		Gulf localities ⁴	
	P_{EXACT}	P_{RB}	P_{EXACT}	P_{RB}	P_{EXACT}	P_{RB}	P_{EXACT}	P_{RB}
Microsatellite								
<i>Sca-14</i>	0.465	0.447	0.845	0.826	0.510	0.503	0.697	0.668
<i>Sca-23</i>	0.026	0.106	0.250	0.416	0.076	0.068	0.175	0.140
<i>Sca-37</i>	0.431	0.388	0.592	0.498	0.038	0.024	0.592	0.619
<i>Sca-44</i>	0.084	0.073	0.558	0.508	0.111	0.161	0.187	0.136
<i>Sca-49</i>	0.230	0.472	0.487	0.746	0.591	0.529	0.112	0.112
<i>Sca-61</i>	0.278	0.428	0.531	0.563	0.065	0.084	0.273	0.296
<i>Sca-65</i>	0.611	0.588	0.457	0.611	0.910	0.957	0.411	0.169

¹ "All" includes all twenty samples.

² Atlantic includes JKV¹–JKV², NSB, CCN, SEB¹–SEB³, FTP, and WPB (nine samples total). For explanation of these geographic abbreviations see Table 1.

³ Keys include KEY¹–KEY³ (three samples total).

⁴ Gulf includes MCI, BCG, SAR¹–SAR⁵, and PCY (seven samples total).

Tables 4 and 5). F_{IS} values (after Weir and Cockerham, 1984) for these four samples were all positive, indicating a deficit of heterozygotes and the possible presence of a null allele. However, probability values for tests of Hardy-Weinberg equilibrium at *Sca-23* among the remaining 16 samples were nonsignificant and averaged (\pm SE) 0.385 \pm 0.076. Although the possibility of a null allele at *Sca-23* cannot be dismissed unequivocally, genotypes at *Sca-23* appear overall to be distributed in accordance with Hardy-Weinberg equilibrium expectation. Finally, tests of genotypic equilibrium between pairs of microsatellites (samples pooled) yielded only one significant value (*Sca-44* \times *Sca-61*) following Bonferroni correction (Table 3). Probability values of tests involving these two microsatellites, i.e. *Sca-44* \times *Sca-61*, carried out within each of the 20 samples were all nonsignificant and averaged (\pm SE) 0.338

\pm 0.067. Of the remaining (pairwise) tests carried out within samples (420 tests in all), only four significant probability values were obtained: *Sca-37* and *Sca-44* (CCN), *Sca-23* \times *Sca-44* (PCY), *Sca-37* \times *Sca-49* (BCG), and *Sca-23* \times *Sca-44* (SEB²). These results indicate that genotypes at the seven microsatellites are independent and hence are not linked genetically.

Tests for independence of allele distributions at each of the seven microsatellites versus both sex and age of individuals were carried out 1) within each of the 20 samples, and 2) among individuals (pooled) sampled from the Atlantic, the Florida Keys, the Gulf, and overall. In tests for independence with sex, eight significant probability values ($P < 0.05$) were found prior to Bonferroni correction for simultaneous tests (data available from first author). None of these, however, were significant after Bonferroni correc-

Table 5

Results of tests for spatial homogeneity in allele distribution of seven microsatellites between and among pooled samples of king mackerel (*Scomberomorus cavalla*) from the east and west coasts of Florida and the Florida Keys. P_{EXACT} = probability based on Fisher's exact tests, with 1000 permutations. P_{RB} = probability based on 1000 bootstrapped replicates (after Roff and Bentzen 1989). Φ_{CT} = estimate of population subdivision based on AMOVA; P is the probability that Φ_{CT} differs significantly from zero (5000 permutations).

Microsatellite	Comparison of Atlantic ¹ with Gulf ²				Comparison of Atlantic and Keys ³ and Gulf			
	P_{EXACT}	P_{RB}	Φ_{CT}	P	P_{EXACT}	P_{RB}	Φ_{CT}	P
<i>Sca-14</i>	0.009	0.006	0.005	0.010	0.020	0.029	0.004	0.031
<i>Sca-23</i>	0.010	0.007	0.001	0.125	0.026	0.081	0.000	0.256
<i>Sca-37</i>	0.426	0.443	-0.001	0.588	0.009	0.001	-0.001	0.794
<i>Sca-44</i>	0.060	0.056	0.004	0.023	0.008	0.004	0.003	0.024
<i>Sca-49</i>	0.506	0.491	-0.001	0.912	0.580	0.535	-0.001	0.707
<i>Sca-61</i>	0.391	0.382	0.001	0.169	0.520	0.689	-0.001	0.398
<i>Sca-65</i>	0.625	0.645	0.004	0.556	0.775	0.508	0.004	0.145

¹ Atlantic includes JKV¹-JKV², NSB, CCN, SEB¹ - SEB³, FTP, and WPB (nine samples total). For explanation of these geographic abbreviation, see Table 1.

² Gulf includes MCI, BCG, SAR¹-SAR⁵, and PCY (seven samples total).

³ Keys includes KEY¹-KEY³ (three samples total).

tion. In addition, one would expect that eight of 162 tests would be significant by chance alone at $\alpha = 0.05$. Finally, only one of the significant probability values occurred in a pooled comparison (*Sca-49* in the test of individuals from the Atlantic), where larger sample sizes were expected to increase robustness of tests of independence. In tests for independence of allele distributions with the age (year class) of individuals, eight significant probability values ($P < 0.05$) were found prior to Bonferroni correction for simultaneous tests (data available from first author), two of which (*Sca-23* in SAR⁴ and *Sca-61* in SAR⁴) were significant after Bonferroni correction with the use of eight simultaneous tests per microsatellite to estimate adjusted α levels. In both instances, nonindependence appeared to stem from an elevated incidence of specific alleles: *Sca-23**19 occurred at a frequency of 36% in the 1989 year class, as opposed to other year classes where its frequency ranged from 0% to 15%; and similarly, *Sca61**12 occurred at a frequency of 25–30% in the 1986 and 1989 year classes, as opposed to a frequency of 0–10% in the other year classes. We suspect these are anomalous instances that do not reflect an age-related effect, in part because allele distributions at *Sca-23* and *Sca-61* were independent of year class in all other samples, and in part because allele distributions at all microsatellites were independent of year class in pooled comparisons, where larger sample sizes should increase robustness of tests of independence. We concluded that allelic variation at the seven microsatellites essentially is independent of variation in both sex and age (year class).

Spatial homogeneity in allele distributions at each microsatellite was tested 1) over all 20 samples, 2) among samples from the Atlantic, 3) among samples from the

Florida Keys, and 4) among samples from the Gulf. Only three significant probability values were found prior to Bonferroni correction: the exact test at *Sca-23* in the comparison over all 20 samples, and both the exact test and the Roff-Bentzen procedure at *Sca-37* in the comparison among samples from the Florida Keys (Table 4). None of the probability values were significant after Bonferroni correction. Tests for spatial homogeneity 1) between pooled samples from the Atlantic versus pooled samples from the Gulf, and 2) among pooled samples from the Atlantic, Florida Keys, and Gulf, indicated that all three regional groupings differed genetically from one another (Table 5). For the comparison Atlantic versus Gulf, significant heterogeneity prior to Bonferroni correction was found at *Sca-14* (all three statistical approaches), *Sca-23* (exact test and the Roff-Bentzen procedure), and *Sca-44* (for the probability that $\Phi_{CT} > 0$). Probability values for *Sca-14* and *Sca-23* were marginal in relation to the (initial) Bonferroni adjusted α of 0.007, whereas the probability that $\Phi_{CT} > 0$ at *Sca-44* was nonsignificant after Bonferroni correction (Table 5). For the comparison of Atlantic, Florida Keys, and Gulf samples, significant heterogeneity was found at *Sca-14* (all three statistical approaches) and *Sca-23* (exact test only) before but not after Bonferroni correction; heterogeneity at *Sca-37* and *Sca-44* in the same comparison was significant both before and after Bonferroni correction in at least one of the three statistical approaches (Table 5). Frequency differences at *Sca-14*, *Sca-23*, and *Sca-44* among the three regional groupings are shown in Table 6 and indicate that small differences in frequency of several alleles at each microsatellite appear to account for observed heterogeneity among the pooled sample comparisons.

Table 6

Allele frequencies at *Sca 14* and *Sca 23* for king mackerel (*Scomberomorus cavalla*) from the Atlantic, Florida Keys, and Gulf. Allele numbers represent the size in base pairs of the fragment amplified.

Microsatellite (allele)	Atlantic	Florida Keys	Gulf
<i>Sca-14</i>			
91	0.017	0.004	0.006
93	0.053	0.058	0.034
95	0.670	0.667	0.735
97	0.233	0.240	0.200
99	0.029	0.031	0.027
<i>Sca-23</i>			
138	0.063	0.054	0.052
140	0.028	0.050	0.028
142	0.221	0.250	0.241
144	0.012	0.008	0.005
146	0.131	0.092	0.121
148	0.026	0.008	0.015
150	0.146	0.108	0.132
152	0.185	0.177	0.149
154	0.008	0.011	0.015
156	0.100	0.111	0.109
158	0.011	0.011	0.011
160	0.009	0.011	0.012
162	0.001	0.000	0.001
164	0.015	0.027	0.016
166	0.002	0.004	0.009
168	0.002	0.004	0.000
170	0.009	0.019	0.018
172	0.017	0.019	0.039
174	0.009	0.015	0.010
176	0.002	0.008	0.009
178	0.002	0.008	0.001
180	0.001	0.004	0.002
182	0.000	0.000	0.002
184	0.002	0.000	0.000
<i>Sca44</i>			
153	0.004	0.015	0.009
157	0.084	0.046	0.090
161	0.030	0.019	0.041
165	0.366	0.308	0.295
169	0.398	0.465	0.438
173	0.104	0.131	0.118
177	0.012	0.011	0.010
181	0.000	0.004	0.000

In general, results of the three approaches to homogeneity testing were fairly consistent, with one notable exception. At *Sca-37*, probability values from the exact test and the Roff-Bentzen procedure were 0.009 and 0.001 in the comparison of Atlantic, Florida Keys, and Gulf samples, respectively, whereas the probability that Φ_{CT} differed from zero was 0.794 (Table 5). We examined

this discrepancy further by carrying out “V” tests of homogeneity (DeSalle et al. 1987) for each allele at *Sca-37*. Significant heterogeneity ($P < 0.05$) was found only at *Sca-37*12*: this allele was found only in the March 1997 sample from the Florida Keys (KEY²), where it occurred at a frequency of 6.9% (Appendix Tables 2 and 3). Because there were far fewer alleles at *Sca-37* sampled from the Florida Keys (258) than from either the Atlantic (884) or Gulf (872), the disproportionate frequency of this allele within the Florida Keys likely accounts for the significance encountered in the exact test and the Roff-Bentzen procedure. Given the absence of this allele in two of the three samples from the Florida Keys, we do not believe the significant heterogeneity detected at *Sca-37* is meaningful biologically.

Although homogeneity testing of pooled samples indicated that samples from the Atlantic differed from samples from the Gulf at *Sca-14* and *Sca-23*, and that samples from the Florida Keys differed from the other two at *Sca-44*, the allele-frequency differences were small and accounted for only a fraction of the overall genetic variance. Results of AMOVA for the comparison of Atlantic with Gulf samples revealed that on average 99.74% of the total genetic variance at the seven microsatellites occurred within samples, as compared to only 0.19% between regions. For the comparison of Atlantic, Florida Keys, and Gulf samples, 99.78% of the genetic variance on average occurred within samples, whereas only 0.11% occurred among regions. For both comparisons, the proportion of the variation among samples within regions accounted for the remainder of the genetic variance, and for both comparisons, this proportion was small and statistically nonsignificant.

Finally, homogeneity tests were used to examine the temporal stock boundaries currently used in management of the king mackerel resource by classifying each of the 20 samples as either Atlantic or Gulf stock (Table 1). No significant heterogeneity at any of the seven microsatellites was found, providing no genetic evidence for existence of temporal boundaries dividing Atlantic and Gulf migratory units (stocks).

Neighbor joining of Cavalli-Sforza’s chord distances between pairs of samples yielded little evidence of geographic structure among the 20 samples. With few exceptions, samples from the same or geographically proximate localities did not cluster together, and most nodes in the topology (available from the first author) were supported by well less than 50% of bootstrap proportions. Spatial autocorrelation (SAAP) analysis also indicated the absence of a relationship between allele frequency and geographic distance. Initially, SAAP analysis employed both equal geographic distances between each of five distance classes and equal numbers of pairwise comparisons in each distance class. Analysis involving equal geographic distances between distance classes generated an uneven number of pairwise comparisons among distance classes, i.e. 18, 14, 15, 5, and 3 pairwise comparisons in distance classes 1–5, respectively, resulting in a high variance in Moran’s I values among alleles in distance classes 4 and 5. Accordingly, the analysis was restricted to equal numbers of pairwise

comparisons (eleven) in each distance class. A total of 50 alleles (five at *Sca-14*, sixteen at *Sca-23*, four at *Sca-37*, six at *Sca-44*, five at *Sca-49*, three at *Sca-61*, and eleven at *Sca-65*) was tested, resulting in 250 Moran's I values. Only 10 significant ($P < 0.05$) Moran's I values were generated: one at *Sca-14* (positive in the third distance class); seven at *Sca-23* (two positive in the second distance class, four positive in the third distance class, and one negative in the fifth distance class); two at *Sca-37* (one negative in the fourth distance class and one positive in the fifth distance class), and one at *Sca-65* (negative in the fifth distance class). No significant Moran's I values were found at *Sca-44*, *Sca-49*, and *Sca-61*. Only one of the "significant" Moran's I values (a positive value for *Sca-23**22 in the third distance class) remained significant after Bonferroni correction. The general absence of spatial autocorrelation indicates that gene flow in king mackerel is consistent with expectations of an island model (*sensu* Wright, 1943) of population structure, meaning that there is roughly an equal probability of gene flow between any of the 20 sample localities.

Assignment tests generally were concordant with other analyses of king mackerel microsatellites in that fish from the Atlantic appeared to be weakly divergent genetically from fish in the Gulf. Each of the 20 samples included high proportions of both Atlantic and Gulf fish, and there appeared to be no strong geographic pattern in proportion of fish assigned to either Atlantic or Gulf groups (Table 7). On average, samples from the Atlantic contained more "Atlantic" fish (54%), whereas samples from the Gulf contained more "Gulf" fish (51.2%). The three samples from the Florida Keys, on average, contained more "Atlantic" fish (53.3%), but this finding is misleading because the proportion of "Atlantic" fish in the three samples from the Florida Keys ranged from 37.2% to 64.0% (Table 7). In addition, the estimated proportions of Atlantic versus Gulf fish in samples from the Florida Keys were not consistent with what might be predicted based on the time of sampling and the spatial-temporal boundaries used currently in king mackerel stock assessment. The KEY¹ and KEY² samples were obtained in March, a time when both would be considered Gulf stock, yet close to the temporal boundary (1 April) when they would be considered Atlantic stock. The estimated proportion of Gulf fish in these two samples was 62.8% (KEY¹) and 41.4% (KEY²). Alternatively, the KEY³ sample was obtained in January when king mackerel in the Florida Keys are considered Gulf stock. The estimated proportion of Gulf fish in the KEY³ sample was 36.0%.

Synopsis and conclusions

Genetic data obtained in our study are compatible with the hypothesis that two, weakly differentiated "genetic" subpopulations of king mackerel exist in waters off Florida and that considerable, perhaps extensive, mixing occurs between them. King mackerel sampled from the Florida Keys cannot be assigned unequivocally to either "genetic" stock; all collections tested appeared to be mix-

Table 7

Assignment (as percentage) of individuals from 20 samples of king mackerel (*Scomberomorus cavalla*) from the east and west coast of Florida and the Florida Keys to either "Atlantic" or "Gulf" group.

Sample	Assigned group	
	Atlantic	Gulf
Atlantic Ocean		
JCK ¹	51.8	48.2
JCK ²	62.5	37.5
NSB	58.0	42.0
CCN	46.0	54.0
SEB ¹	58.0	42.0
SEB ²	62.0	38.0
SEB ³	52.0	48.0
FTP	53.6	46.4
WPB	41.8	58.2
Avg.	54.0	46.0
Florida Keys		
KEY ¹	37.2	62.8
KEY ²	58.6	41.4
KEY ³	64.0	36.0
Avg.	53.3	46.7
Gulf of Mexico		
MCI	49.1	50.9
BCG	45.7	54.3
SAR ¹	46.8	53.2
SAR ²	53.3	46.7
SAR ³	53.6	46.4
SAR ⁴	41.1	58.6
SAR ⁵	54.3	45.7
PCY	46.0	54.0
Avg.	48.8	51.2

tures, with approximately equal proportions of fish from the two "genetic" stocks. These results are not consistent with the current spatial-temporal boundaries employed in stock assessment and allocation of the king mackerel resource. Results are consistent with the hypothesis that considerable gene flow occurs among all of the localities sampled, and that differences in gene flow likely do not arise as a function of geographic distance. Similar findings were obtained by Gold et al. (1997) in their study of variation in king mackerel mitochondrial DNA. The genetic differences between king mackerel in the Atlantic versus those in the Gulf most likely stem from reduced gene flow (migration) between the Atlantic and Gulf in relation to gene flow (migration) along the Atlantic and Gulf coasts of peninsular Florida. This is consistent with the notion based on studies of other marine fishes (Avise et al., 1992; Gold and Richardson, 1998) that the southern Florida peninsula serves (or has served) as a biogeographic boundary.

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Appendix Table 1Microsatellites employed for king mackerel (*Scomberomorus cavalla*).

Microsatellite	Primer sequence (5' → 3') (forward and reverse, respectively)	Length (base pairs)	Annealing temperature (°C)
<i>Sca-14</i>	ATT CCC CAA ACA ATA CAC AC AGT GGA CGA CCC ATT CTA C	93	56
<i>Sca-23</i>	AGC CCT CTT ACA ATC TGC TAC CC AAA CCT TTA AGG CCT CAA GTA AAG	146	58
<i>Sca-37</i>	GCG CCG TGA CTT TTT ATT GCT C CAA CAA TTA GTC GCA GCC CTA G	154	58
<i>Sca-44</i>	ATG GCC AAA TGG CAC ATA ATC A GGG CAG CTC CAT GGG TCT GAG T	169	58
<i>Sca-49</i>	AGA TGT GAC AAC AGT GGG ATG GCA GCA GTA ATA AAG	157	56
<i>Sca-61</i>	GGT ACT GTC GGG AGA ATG AGA T TGA ATT TTA TAT GGA GGG TCT G	228	56
<i>Sca-65</i>	AGC TGC TGC CAT GAT TTG TT TCC TCC ACT GCC CCT TTC TT	129	52

Appendix Table 2

Allele frequencies at microsatellites in king mackerel (*Scomberomorus cavalla*). Legend to samples: JKV¹ = Jacksonville, Florida (July 1996); JKV² = Jacksonville, Florida (July, 1998); NSB = New Smyrna Beach, Florida (July 1996); CCN = Cape Canaveral, Florida (December 1998); SEB¹ = Sebastian, Florida (March 1997); SEB² = Sebastian, Florida (March 1998); SEB³ = Sebastian, Florida (December 1998); FTP = Ft. Pierce, Florida (April 1996); WPB = West Palm Beach, Florida (May 1998); KEY¹ = Key West, Florida (March 1996).

Microsatellite (allele) ¹	Sample									
	JKV ¹	JKV ²	NSB	CCN	SEB ¹	SEB ²	SEB ³	FTP	WPB	KEY ¹
<i>Sca-14</i>										
91	0.000	0.031	0.040	0.030	0.010	0.030	0.000	0.009	0.009	0.010
93	0.046	0.063	0.070	0.040	0.041	0.070	0.050	0.063	0.037	0.030
95	0.741	0.719	0.650	0.630	0.735	0.590	0.670	0.643	0.667	0.680
97	0.185	0.188	0.220	0.250	0.184	0.280	0.250	0.259	0.259	0.260
99	0.028	0.000	0.020	0.050	0.031	0.030	0.030	0.027	0.028	0.020
<i>Sca-23</i>										
138	0.046	0.016	0.060	0.080	0.090	0.030	0.060	0.091	0.073	0.029
140	0.019	0.032	0.030	0.050	0.020	0.000	0.060	0.027	0.018	0.039
142	0.231	0.210	0.120	0.250	0.230	0.230	0.210	0.236	0.264	0.265
144	0.009	0.016	0.000	0.020	0.020	0.000	0.010	0.018	0.018	0.000
146	0.102	0.129	0.140	0.100	0.140	0.170	0.140	0.127	0.136	0.147
148	0.019	0.048	0.030	0.000	0.060	0.010	0.010	0.036	0.027	0.020
150	0.176	0.161	0.140	0.180	0.120	0.190	0.100	0.155	0.100	0.088
152	0.241	0.177	0.180	0.120	0.180	0.280	0.150	0.145	0.191	0.098
154	0.009	0.016	0.000	0.010	0.010	0.000	0.000	0.018	0.009	0.020
156	0.111	0.113	0.130	0.110	0.060	0.050	0.140	0.064	0.091	0.118
158	0.019	0.016	0.020	0.020	0.000	0.000	0.010	0.000	0.018	0.020
160	0.000	0.016	0.020	0.000	0.010	0.010	0.030	0.000	0.000	0.000
162	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.009	0.000
164	0.009	0.016	0.030	0.010	0.000	0.030	0.020	0.009	0.009	0.039
166	0.000	0.000	0.010	0.000	0.000	0.000	0.000	0.000	0.009	0.000
168	0.000	0.000	0.000	0.000	0.000	0.000	0.010	0.009	0.000	0.010
170	0.009	0.016	0.010	0.020	0.000	0.000	0.010	0.018	0.000	0.039
172	0.000	0.016	0.060	0.010	0.030	0.000	0.010	0.018	0.009	0.029
174	0.000	0.000	0.020	0.000	0.010	0.000	0.010	0.018	0.018	0.010
176	0.000	0.000	0.000	0.010	0.010	0.000	0.000	0.000	0.000	0.010
178	0.000	0.000	0.000	0.000	0.000	0.000	0.020	0.000	0.000	0.010
180	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.009	0.000	0.010
182	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
184	0.000	0.000	0.000	0.010	0.010	0.000	0.000	0.000	0.000	0.000
<i>Sca-37</i>										
138	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
142	0.000	0.031	0.030	0.000	0.000	0.010	0.010	0.000	0.000	0.020
144	0.667	0.688	0.600	0.694	0.720	0.630	0.540	0.623	0.630	0.637
146	0.009	0.016	0.040	0.010	0.030	0.010	0.030	0.019	0.019	0.000
148	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
150	0.000	0.000	0.010	0.000	0.000	0.000	0.000	0.000	0.000	0.000
152	0.009	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
154	0.296	0.266	0.320	0.286	0.240	0.330	0.400	0.330	0.286	0.333
156	0.019	0.000	0.000	0.010	0.010	0.020	0.020	0.028	0.010	0.010
<i>Sca44</i>										
153	0.000	0.000	0.000	0.010	0.000	0.000	0.010	0.018	0.000	0.020
157	0.066	0.078	0.110	0.120	0.060	0.080	0.090	0.098	0.056	0.069
161	0.038	0.047	0.030	0.020	0.030	0.040	0.010	0.027	0.037	0.020
165	0.387	0.469	0.390	0.300	0.390	0.310	0.290	0.420	0.370	0.275
169	0.396	0.281	0.380	0.420	0.410	0.470	0.500	0.357	0.333	0.471
173	0.104	0.109	0.090	0.110	0.100	0.090	0.070	0.071	0.194	0.147
177	0.009	0.016	0.000	0.020	0.010	0.010	0.030	0.009	0.009	0.000
181	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000

continued

Appendix Table 2 (continued)

Microsatellite (allele) ¹	Sample									
	JKV ¹	JKV ²	NSB	CCN	SEB ¹	SEB ²	SEB ³	FTP	WPB	KEY ¹
<i>Sca-49</i>										
139	0.009	0.000	0.000	0.000	0.000	0.000	0.010	0.009	0.000	0.000
141	0.046	0.109	0.060	0.000	0.043	0.020	0.020	0.036	0.037	0.067
143	0.009	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.011
145	0.546	0.547	0.490	0.590	0.435	0.690	0.580	0.518	0.481	0.533
147	0.167	0.203	0.150	0.130	0.207	0.100	0.170	0.188	0.167	0.178
149	0.019	0.031	0.060	0.030	0.033	0.030	0.030	0.018	0.046	0.000
151	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
153	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
155	0.009	0.000	0.010	0.000	0.000	0.000	0.000	0.000	0.000	0.000
157	0.046	0.031	0.090	0.070	0.076	0.060	0.080	0.063	0.093	0.089
159	0.130	0.063	0.130	0.170	0.196	0.090	0.090	0.161	0.148	0.111
161	0.009	0.016	0.000	0.010	0.011	0.010	0.010	0.009	0.009	0.011
163	0.000	0.000	0.010	0.000	0.000	0.000	0.000	0.000	0.009	0.000
165	0.009	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.009	0.000
167	0.000	0.000	0.000	0.000	0.000	0.000	0.010	0.000	0.000	0.000
<i>Sca 61</i>										
208	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
210	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.010
224	0.083	0.078	0.100	0.041	0.071	0.102	0.130	0.074	0.046	0.088
226	0.139	0.063	0.050	0.112	0.061	0.071	0.090	0.065	0.083	0.137
228	0.769	0.859	0.850	0.847	0.857	0.827	0.780	0.861	0.861	0.765
230	0.009	0.000	0.000	0.000	0.010	0.000	0.000	0.000	0.009	0.000
<i>Sca 65</i>										
117	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.010
123	0.380	0.422	0.360	0.430	0.440	0.350	0.360	0.384	0.464	0.438
125	0.028	0.047	0.040	0.050	0.030	0.080	0.040	0.063	0.036	0.063
127	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.009	0.000	0.010
129	0.065	0.016	0.130	0.060	0.080	0.090	0.130	0.036	0.036	0.073
131	0.037	0.094	0.030	0.030	0.020	0.000	0.030	0.018	0.000	0.042
133	0.000	0.000	0.000	0.000	0.020	0.000	0.000	0.009	0.009	0.000
135	0.241	0.219	0.200	0.170	0.150	0.280	0.200	0.223	0.209	0.219
137	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.009	0.000
139	0.000	0.000	0.000	0.010	0.010	0.010	0.000	0.018	0.018	0.000
141	0.000	0.000	0.000	0.000	0.000	0.010	0.000	0.000	0.000	0.000
143	0.009	0.000	0.000	0.010	0.020	0.010	0.000	0.009	0.018	0.010
147	0.093	0.047	0.150	0.080	0.080	0.070	0.150	0.080	0.109	0.063
149	0.028	0.016	0.010	0.020	0.020	0.010	0.010	0.018	0.009	0.021
151	0.009	0.016	0.020	0.040	0.030	0.030	0.020	0.045	0.018	0.000
153	0.046	0.016	0.020	0.000	0.040	0.020	0.020	0.045	0.027	0.031
155	0.037	0.031	0.020	0.050	0.010	0.020	0.020	0.036	0.027	0.010
157	0.019	0.016	0.010	0.030	0.020	0.020	0.010	0.009	0.000	0.000
159	0.000	0.016	0.000	0.020	0.010	0.000	0.000	0.000	0.000	0.000
161	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
163	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.010
165	0.000	0.016	0.000	0.000	0.010	0.000	0.000	0.000	0.000	0.000
167	0.009	0.031	0.010	0.000	0.010	0.000	0.010	0.000	0.000	0.000
171	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000

¹ Allele number represents size in base pairs of the fragment amplified.

Appendix Table 3

Allele frequencies at microsatellites in king mackerel (*Scomberomorus cavalla*). Legend to samples: KEY² = Key West, Florida (March 1997); KEY³ = Key West, Florida (January 1999); MCI = Marco Island, Florida (April 1996); BCG = Boca Grande, Florida (April 1996); SAR¹ = Treasure Island, Florida (April 1996); SAR² = Treasure Island, Florida (November 1996); SAR³ = Treasure Island, Florida (April 1997); SAR⁴ = Treasure Island, Florida (April 1998); SAR⁵ = Treasure Island, Florida (November 1998); PCY = Panama City, Florida (October 1996).

Microsatellite (allele) ¹	Sample									
	KEY ²	KEY ³	MCI	BCG	SAR ¹	SAR ²	SAR ³	SAR ⁴	SAR ⁵	PCY
<i>Sca-14</i>										
91	0.000	0.000	0.009	0.014	0.000	0.009	0.009	0.000	0.007	0.000
93	0.069	0.080	0.036	0.057	0.033	0.018	0.063	0.021	0.036	0.020
95	0.724	0.620	0.745	0.771	0.685	0.764	0.741	0.771	0.714	0.680
97	0.190	0.250	0.173	0.143	0.250	0.200	0.170	0.186	0.221	0.230
99	0.017	0.050	0.036	0.014	0.033	0.009	0.018	0.021	0.021	0.070
<i>Sca-23</i>										
138	0.086	0.060	0.018	0.057	0.096	0.033	0.027	0.036	0.096	0.060
140	0.017	0.080	0.027	0.029	0.032	0.017	0.027	0.043	0.029	0.020
142	0.224	0.250	0.309	0.257	0.266	0.217	0.200	0.239	0.250	0.200
144	0.017	0.010	0.000	0.000	0.000	0.000	0.018	0.000	0.000	0.020
146	0.069	0.050	0.145	0.171	0.064	0.183	0.118	0.109	0.103	0.080
148	0.000	0.000	0.018	0.000	0.032	0.008	0.009	0.029	0.007	0.010
150	0.121	0.120	0.155	0.043	0.160	0.108	0.173	0.130	0.132	0.130
152	0.207	0.240	0.136	0.157	0.117	0.142	0.164	0.174	0.176	0.110
154	0.000	0.010	0.000	0.029	0.011	0.017	0.009	0.007	0.022	0.030
156	0.086	0.120	0.045	0.157	0.117	0.117	0.127	0.109	0.096	0.130
158	0.017	0.000	0.000	0.000	0.021	0.008	0.027	0.007	0.000	0.030
160	0.052	0.000	0.027	0.014	0.000	0.033	0.000	0.014	0.000	0.010
162	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.007	0.000	0.000
164	0.034	0.010	0.000	0.029	0.000	0.033	0.018	0.014	0.022	0.010
166	0.017	0.000	0.009	0.014	0.000	0.000	0.009	0.007	0.000	0.040
168	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
170	0.000	0.010	0.027	0.014	0.011	0.025	0.009	0.014	0.015	0.030
172	0.017	0.010	0.036	0.014	0.064	0.025	0.036	0.036	0.037	0.060
174	0.017	0.020	0.018	0.000	0.000	0.017	0.009	0.007	0.015	0.010
176	0.000	0.010	0.027	0.014	0.000	0.000	0.009	0.007	0.000	0.020
178	0.017	0.000	0.000	0.000	0.011	0.000	0.000	0.000	0.000	0.000
180	0.000	0.000	0.000	0.000	0.000	0.017	0.000	0.000	0.000	0.000
182	0.000	0.000	0.000	0.000	0.000	0.000	0.009	0.007	0.000	0.000
184	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
<i>Sca-37</i>										
138	0.069	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
142	0.034	0.000	0.009	0.000	0.000	0.017	0.009	0.007	0.000	0.023
144	0.603	0.653	0.627	0.700	0.628	0.642	0.679	0.600	0.579	0.593
146	0.000	0.010	0.018	0.043	0.021	0.025	0.036	0.007	0.007	0.000
148	0.000	0.000	0.000	0.000	0.000	0.009	0.000	0.000	0.000	0.000
150	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
152	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
154	0.276	0.327	0.327	0.257	0.319	0.292	0.250	0.343	0.379	0.349
156	0.017	0.010	0.018	0.000	0.032	0.017	0.027	0.043	0.036	0.035
<i>Sca-44</i>										
153	0.034	0.000	0.009	0.014	0.000	0.017	0.000	0.029	0.000	0.000
157	0.069	0.010	0.056	0.100	0.096	0.133	0.089	0.114	0.072	0.050
161	0.052	0.000	0.037	0.014	0.032	0.008	0.098	0.036	0.051	0.040
165	0.276	0.360	0.259	0.314	0.255	0.300	0.268	0.293	0.355	0.300
169	0.431	0.480	0.546	0.443	0.436	0.408	0.455	0.393	0.413	0.430
173	0.121	0.120	0.093	0.100	0.170	0.133	0.080	0.129	0.087	0.160
177	0.017	0.020	0.000	0.014	0.011	0.000	0.009	0.007	0.022	0.020
181	0.000	0.010	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000

continued

Appendix Table 3 (continued)

Microsatellite (allele) ¹	Sample									
	KEY ²	KEY ³	MCI	BCG	SAR ¹	SAR ²	SAR ³	SAR ⁴	SAR ⁵	PCY
<i>Sca-49</i>										
139	0.000	0.000	0.009	0.000	0.011	0.010	0.009	0.000	0.000	0.000
141	0.069	0.070	0.055	0.043	0.054	0.031	0.054	0.043	0.036	0.020
143	0.000	0.000	0.000	0.000	0.022	0.000	0.000	0.007	0.000	0.000
145	0.621	0.580	0.464	0.457	0.576	0.594	0.455	0.614	0.529	0.560
147	0.121	0.100	0.218	0.200	0.076	0.156	0.152	0.114	0.200	0.180
149	0.017	0.040	0.009	0.029	0.011	0.010	0.027	0.014	0.014	0.060
151	0.000	0.000	0.009	0.014	0.000	0.000	0.000	0.007	0.000	0.000
153	0.000	0.000	0.000	0.000	0.011	0.000	0.000	0.000	0.000	0.000
155	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
157	0.052	0.080	0.100	0.129	0.076	0.063	0.089	0.021	0.071	0.050
159	0.086	0.130	0.118	0.129	0.141	0.135	0.196	0.171	0.143	0.130
161	0.034	0.000	0.018	0.000	0.000	0.000	0.009	0.007	0.000	0.000
163	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
165	0.000	0.000	0.000	0.000	0.022	0.000	0.009	0.000	0.000	0.000
167	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.007	0.000
<i>Sca-61</i>										
208	0.000	0.000	0.000	0.000	0.000	0.009	0.000	0.000	0.000	0.000
210	0.000	0.000	0.000	0.000	0.011	0.000	0.000	0.000	0.000	0.000
224	0.052	0.130	0.073	0.086	0.054	0.138	0.127	0.107	0.100	0.070
226	0.017	0.100	0.055	0.143	0.098	0.078	0.100	0.114	0.121	0.070
228	0.931	0.760	0.873	0.757	0.837	0.776	0.755	0.779	0.771	0.860
230	0.999	0.010	0.000	0.014	0.000	0.000	0.018	0.000	0.007	0.000
<i>Sca-65</i>										
117	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.010
123	0.483	0.460	0.364	0.357	0.436	0.446	0.375	0.357	0.386	0.340
125	0.017	0.030	0.036	0.100	0.064	0.045	0.071	0.086	0.043	0.060
127	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
129	0.052	0.080	0.055	0.057	0.032	0.054	0.063	0.057	0.021	0.060
131	0.000	0.020	0.027	0.000	0.064	0.036	0.018	0.071	0.029	0.020
133	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
135	0.207	0.160	0.273	0.300	0.245	0.179	0.205	0.193	0.243	0.190
137	0.000	0.000	0.000	0.000	0.000	0.009	0.000	0.000	0.000	0.010
139	0.000	0.010	0.000	0.000	0.000	0.009	0.009	0.007	0.007	0.000
141	0.000	0.000	0.018	0.000	0.000	0.000	0.000	0.000	0.000	0.010
143	0.017	0.010	0.000	0.014	0.000	0.036	0.009	0.000	0.007	0.000
147	0.103	0.110	0.073	0.029	0.053	0.080	0.080	0.100	0.114	0.090
149	0.034	0.020	0.036	0.057	0.021	0.000	0.018	0.014	0.029	0.020
151	0.017	0.040	0.055	0.000	0.000	0.009	0.018	0.014	0.021	0.040
153	0.034	0.030	0.018	0.029	0.021	0.036	0.054	0.029	0.029	0.090
155	0.017	0.020	0.018	0.014	0.032	0.027	0.045	0.021	0.029	0.050
157	0.017	0.010	0.018	0.014	0.011	0.018	0.027	0.014	0.029	0.010
159	0.000	0.000	0.000	0.014	0.000	0.000	0.009	0.014	0.000	0.000
161	0.000	0.000	0.009	0.000	0.000	0.000	0.000	0.007	0.000	0.000
163	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
165	0.000	0.000	0.000	0.000	0.000	0.018	0.000	0.000	0.000	0.000
167	0.000	0.000	0.000	0.014	0.021	0.000	0.000	0.014	0.007	0.000
171	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.007	0.000

¹ Allele number represents size in base pairs of the fragment amplified.

Appendix Table 4

Summary statistics for each of seven microsatellites among samples of king mackerel (*Scomberomorus cavalla*): n = number of individuals assayed; H_{DC} = direct-count heterozygosity; and P_{HW} = probability that genotypes conform to expectations of Hardy-Weinberg equilibrium. Legends to sample localities are given in Appendix Table 2.

Microsatellite	JKV ¹	JKV ²	NSB	CCN	SEB ¹	SEB ²	SEB ³	FTP	WPB	KEY ¹
<i>Sca-14</i>										
n	54	32	50	50	49	50	50	56	54	50
H_{DC}	0.481	0.375	0.460	0.500	0.408	0.600	0.420	0.571	0.537	0.520
P_{HW}	0.710	0.297	0.141	0.354	0.333	0.431	0.440	0.203	0.070	0.103
<i>Sca-23</i>										
n	54	31	50	50	50	50	50	55	55	51
H_{DC}	0.833	0.806	0.780	0.840	0.820	0.600	0.800	0.855	0.800	0.765
P_{HW}	0.571	0.094	0.121	0.618	0.654	0.000	0.141	0.085	0.483	0.000
<i>Sca-37</i>										
n	54	32	50	49	50	50	50	53	54	51
H_{DC}	0.500	0.531	0.540	0.449	0.380	0.520	0.560	0.585	0.463	0.608
P_{HW}	0.039	0.306	0.903	0.193	0.607	1.000	0.784	0.291	0.293	0.069
<i>Sca-44</i>										
n	53	32	50	50	50	50	50	56	54	51
H_{DC}	0.604	0.719	0.720	0.680	0.720	0.620	0.660	0.643	0.722	0.569
P_{HW}	0.584	0.965	0.911	0.518	0.583	0.618	0.719	0.875	0.095	0.095
<i>Sca-49</i>										
n	54	32	50	49	46	50	50	56	54	45
H_{DC}	0.611	0.656	0.820	0.680	0.739	0.540	0.620	0.696	0.704	0.600
P_{HW}	0.722	0.543	0.871	0.509	0.290	0.753	0.313	0.958	0.943	0.571
<i>Sca-61</i>										
n	54	32	50	49	49	49	50	54	54	51
H_{DC}	0.407	0.281	0.280	0.306	0.224	0.306	0.360	0.204	0.278	0.392
P_{HW}	1.000	1.000	0.709	1.000	0.152	0.306	0.703	0.093	1.000	0.851
<i>Sca-65</i>										
n	54	32	50	50	50	50	50	56	55	48
H_{DC}	0.870	0.813	0.900	0.900	0.820	0.800	0.780	0.839	0.727	0.729
P_{HW}	0.975	0.388	0.647	0.985	0.777	0.242	0.917	0.978	0.479	0.838

Appendix Table 5

Summary statistics for each of seven microsatellites among samples of king mackerel (*Scomberomorus cavalla*): n = number of individuals assayed; H_{DC} = direct-count heterozygosity; and P_{HW} = probability that genotypes conform to expectations of Hardy-Weinberg equilibrium. Legends to sample localities are in Appendix Table 2.

Microsatellite (allele)	KEY ²	KEY ³	MCI	BCG	SAR ¹	SAR ²	SAR ³	SAR ⁴	SAR ⁵	PCY
<i>Sca-14</i>										
n	29	50	55	35	46	55	56	70	70	50
H_{DC}	0.517	0.500	0.473	0.371	0.543	0.345	0.429	0.414	0.500	0.520
P_{HW}	0.760	0.143	0.246	0.042	0.562	0.403	0.690	0.540	0.824	0.336
<i>Sca-23</i>										
n	29	50	55	35	47	60	55	69	68	50
H_{DC}	0.897	0.760	0.745	0.886	0.723	0.800	0.855	0.783	0.838	0.880
P_{HW}	0.416	0.106	0.032	0.947	0.006	0.039	0.846	0.000	0.647	0.365
<i>Sca-37</i>										
n	29	50	55	35	47	60	55	69	68	50
H_{DC}	0.552	0.469	0.491	0.486	0.447	0.617	0.446	0.571	0.543	0.419
P_{HW}	0.092	0.647	0.384	0.851	0.079	0.431	0.817	0.499	0.785	0.235
<i>Sca-44</i>										
n	29	50	54	35	47	60	56	70	70	50
H_{DC}	0.828	0.560	0.611	0.686	0.723	0.633	0.696	0.714	0.739	0.700
P_{HW}	0.686	0.219	0.953	0.722	0.353	0.495	0.335	0.813	0.132	0.050
<i>Sca-49</i>										
n	29	50	55	35	46	48	56	70	70	50
H_{DC}	0.586	0.580	0.727	0.771	0.630	0.625	0.714	0.500	0.686	0.640
P_{HW}	0.667	0.055	0.679	0.153	0.098	0.806	0.303	0.246	0.847	0.856
<i>Sca-61</i>										
n	29	50	55	35	46	58	55	70	70	50
H_{DC}	0.138	0.480	0.218	0.343	0.326	0.379	0.327	0.314	0.371	0.280
P_{HW}	1.000	0.486	0.390	0.073	1.000	0.348	0.016	0.105	0.364	1.000
<i>Sca-65</i>										
n	29	50	55	35	47	56	56	70	70	50
H_{DC}	0.690	0.800	0.727	0.743	0.681	0.696	0.839	0.843	0.886	0.880
P_{HW}	0.581	0.656	0.317	0.615	0.250	0.425	0.526	0.879	0.183	0.942