Journal of Fish Biology (1994) **44**, 567–590

Allozyme differentiation within and between red drum (*Sciaenops ocellatus*) from the Gulf of Mexico and Atlantic Ocean

J. R. GOLD*, T. L. KING†, L. R. RICHARDSON*, D. A. BOHLMEYER* AND G. C. MATLOCK†

**Department of Wildlife and Fisheries Sciences, Texas A&M University, College Station, Texas 77843 and †Texas Parks and Wildlife Department, 4200 Smith School Road, Austin, Texas 78744 U.S.A.*

(*Received 28 September 1992, Accepted 28 April 1993*)

Nine polymorphic nuclear-gene (allozyme) loci were surveyed among 491 red drum (*Sciaenops ocellatus*) sampled in 1988 and 1989 from nearshore localities in the northern Gulf of Mexico (Gulf) and the Atlantic coast of the southeastern United States (Atlantic). Data were combined with those from a previous study to generate a data set of 762 individuals representing 11 sample localities in the Gulf and 175 individuals representing five sample localities in the Atlantic. The combined data set included individuals from the 1986 and 1987 year classes and permitted rigorous testing of both temporal and spatial genetic heterogeneity. Average heterozygosity-per-locus values (estimated using 33 assumed monomorphic loci) were 0·048 (Gulf red drum) and 0·046 (Atlantic red drum). Tests of heterogeneity in allele frequencies between year classes at individual localities and across regions (Gulf and Atlantic) were non-significant. Tests of spatial (geographic) heterogeneity indicated that red drum are weakly subdivided: genetically-differentiated subpopulations occur in the northern Gulf and along the south-eastern Atlantic coast. Genetic data were consistent with the hypothesis that red drum within the Gulf and along the Atlantic coast comprise single subpopulations. Genetic differences between Gulf and Atlantic red drum seem likely to stem from historical or recent interactions between dispersal and impediments to gene flow.

Key words: red drum; genetic differentiation; Gulf of Mexico; Atlantic Ocean.

I. INTRODUCTION

Red drum (*Sciaenops ocellatus* L.) is an important recreational species in the northern Gulf of Mexico (Gulf) and along the Atlantic coast (Atlantic) of the south-eastern United States (Matlock, 1984; Swingle, 1987). Historically, red drum supported an important commercial fishery (Swingle, 1987), and considerable effort has been spent domesticating the species for aquaculture (Chamberlain *et al.*, 1987). A major question in management of red drum is whether discrete subpopulations (stocks) exist within the Gulf or between the Gulf and Atlantic (McIlwain *et al.*, 1986). A second question is whether declines in red drum abundance (Matlock, 1984; Reagan, 1985) have affected the genetic diversity of the species.

Information on red drum population structure derived from non-genetic data is contradictory. Tagging studies indicate that movement of juveniles among nearshore localities (bays and estuaries) is limited (Adkins *et al.*, 1979; Matlock & Weaver, 1979; Osburn *et al.*, 1982). These and other studies (Matlock, 1987*a*) have led to the suggestion that two, perhaps three spatially-isolated red drum

0022–1112/94/040567+24 $08.00/0

subpopulations exist along the Gulf and Atlantic coasts. Information on red drum biology and life-history, alternatively, suggests that dispersal could be extensive at any of several life-stages (Overstreet, 1983; Mercer, 1984; Ross *et al.*, 1987; Lyczkowski-Schultz *et al.*, 1988).

Previous genetic studies have indicated that red drum may be subdivided within the Gulf and between the Gulf and Atlantic. Ramsey & Wakeman (1987) found significant heterogeneity in allele frequencies at a locus for glucose phosphate isomerase among red drum from the Gulf, and Bohlmeyer & Gold (1991) found highly significant heterogeneity in allele frequencies at a locus for adenosine deaminase between red drum from the Gulf and the Atlantic and among red drum from the Gulf. Estimated mean F_{ST} values (the standardized variance of allele frequencies among samples) in both studies, however, were approximately 0·019, indicating high gene flow among geographic samples. Gold & Richardson (1991) studied variation in mitochondrial (mt)DNA among most of the red drum examined by Bohlmeyer & Gold (1991) and found significant heterogeneity in the frequencies of four mtDNA haplotypes between pooled samples from the Gulf and pooled samples from the Atlantic. No mtDNA heterogeneity was found among samples from the Gulf or among samples from the Atlantic.

The purposes of this study were to further test the hypothesis that red drum are spatially subdivided, and to test for homogeneity in allele frequencies between samples from different year classes. Data on allelic variation at nine polymorphic allozyme loci among 491 red drum are presented and are combined with data from Bohlmeyer & Gold (1991) for analysis. The combined data set included 762 individuals from the Gulf and 175 individuals from the Atlantic and consisted of individuals from the 1986 and 1987 year classes. Combination of the two data sets permitted more rigorous testing of spatial homogeneity because of increased sample sizes at individual localities, as well as testing of temporal homogeneity (between year classes) at individual localities and across regions.

II. MATERIALS AND METHODS

Muscle and liver tissues were removed from 456 red drum from the Gulf and 35 red drum from the Atlantic, placed in liquid nitrogen for transport to the laboratory, and stored at −80° C. Fish were procured in 1988 and 1989 using a variety of methods. Collection localities are shown in Fig. 1 and number of individuals taken at each locality by year class are given in Table I. Total sample sizes in Table I include material from Bohlmeyer & Gold (1991). Ages of all but yearling (age zero) individuals (specimens <300 mm total length) were determined from annuli on otoliths using methods in Bumguardner (1991). The first annulus was considered to have formed 14–15 months after hatching (Bumguardner, 1991).

Individuals were surveyed for allelic variation at the nine presumptive nuclear-gene loci found to be polymorphic by Bohlmeyer & Gold (1991). The loci (number of alleles) were: *ACP-2** (acid phosphatase, three alleles); *ADA** (adenosine deaminase, 13 alleles); *ADH** (alcohol dehydrogenase, four alleles); *sAAT-1** (aspartate aminotransferase, four alleles); *EST-1** (esterase, two alleles); *GPI-B** (glucose phosphate isomerase, three alleles); *PEPB** (peptidase B, three alleles); *PEPD** (peptidase D, four alleles); and *PEPS** (peptidase S, three alleles). Techniques for vertical starch-gel electrophoresis of proteins, recipes for grinding and running buffers, starch composition of gels, protein staining, and

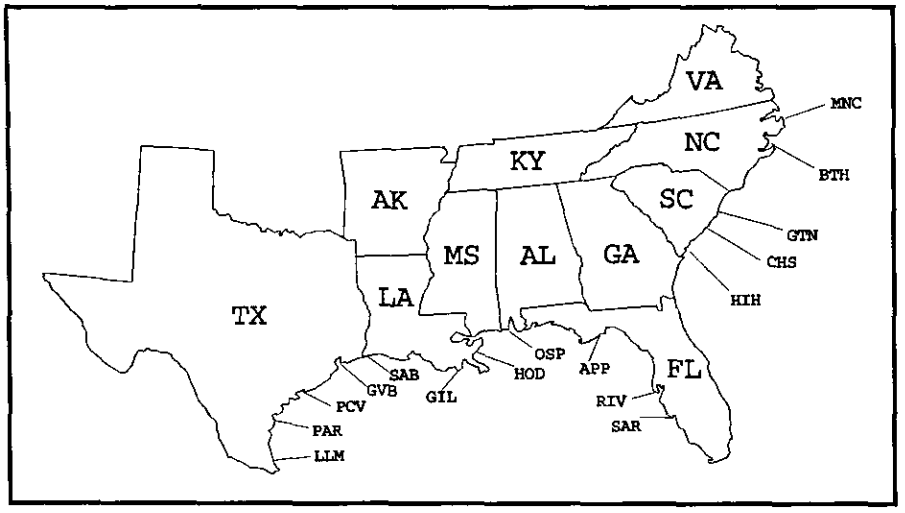

FIG. 1. Map of nearshore localities for red drum samples. Acronyms refer to localities defined in Table I.

interpretation of banding patterns of the presumptive loci may be found in Bohlmeyer (1989) and Bohlmeyer & Gold (1991). Designation of allelic variants was based on relative mobility to the most common allele, which was designated as Allele *100*.

Tests of Hardy–Weinberg equilibrium expectations, Wright's F_{ST} statistic (the standardized variance of allele frequencies among samples), Nei's (1978) unbiased genetic distance ($\bar{D}$) indices, and total gene diversity (partitioned among localities and among samples within localities) were carried out using BIOSYS-1 (Swofford & Selander, 1981). Deviations from Hardy–Weinberg equilibrium expectations were tested using pooled genotypes and the χ^2 statistic with 1 dof. Significance testing of allele-frequency heterogeneity among sampling localities (spatial) and between year classes (temporal) was accomplished using (i) *G* tests (Sokal & Rohlf, 1969) of contingency tables of allele counts and the BIOM-PC program package of Rohlf (1983), and (ii) *V* tests (DeSalle *et al.*, 1987) of arcsin, square-root transformed allele frequencies. Significance levels for all multiple tests performed simultaneously were adjusted after Cooper (1968).

Mean F_{ST} values were computed as the arithmetic average of F_{ST} values over the nine polymorphic loci. Estimates of gene flow (N_em, the effective number of migrants per generation) were calculated using Wright's (1943) island model, where $F_{ST}=1/(4N_em+1)$. Average levels of gene flow among geographic sample localities were estimated graphically using Slatkin's (1981) qualitative method. Cluster analysis of Nei's unbiased genetic distance ($\bar{D}$) values was carried out using UPGMA (unweighted pair-group method using arithmetic averages) after Sneath & Sokal (1973). The computer program used in UPGMA cluster analysis provides standard errors for phenogram nodes according to equations in Nei *et al.* (1985). Spatial autocorrelation analysis of frequencies of common alleles was carried out using the Spatial Autocorrelation Analysis Program (SAAP) of Wartenberg (1989) following recommendations in Sokal & Oden (1978*a*,*b*). Geographic distances between pairs of localities were estimated by SAAP (Wartenberg, 1989) using input latitudes and longitudes for each locality.

III. RESULTS

Allele frequencies at the nine polymorphic loci among all red drum surveyed to date are summarized by sample locality, year class, and region in Appendix Tables I–III. The mean number of alleles over all polymorphic loci was 4·33.

TABLE I. Collection localities and number of red drum collected

Locality	Year class	No. of individuals	
		This study	Total*
Gulf of Mexico	1986	158	464
	1987	298	298
Sarasota Bay, FL (SAR)	1986	37	86
	1987	24	24
Riviera Bay, FL (RIV)	1986	0	24
	1987	45	45
Apalachicola Bay, FL (APP)	1986	6	30
	1987	38	38
Biloxi Bay, MS (OSP)	1986	28	78
	1987	34	34
Black Bay, LA (HOD)	1986	0	50
Grand Isle, LA (GIL)	1986	24	74
	1987	47	47
Sabine Pass, TX (SAB)	1986	25	25
	1987	18	18
West Bay, TX (GVB)	1986	0	32
	1987	36	36
Pass Cavallo, TX (PCV)	1986	12	12
	1987	17	17
Redfish Bay, TX (PAR)	1986	8	35
	1987	21	21
Lower Laguna Madre, TX (LLM)	1986	18	18
	1987	18	18
Atlantic Ocean:	1986	0	140
	1987	35	35
Oregon Inlet, NC (MNC)	1986	0	15
Pamlico River, NC (BTH)	1986	0	23
	1987	4	4
North Inlet, SC (GTN)	1986	0	18
Charleston Bay, SC (CHS)	1986	0	34
	1987	31	31
Calibogue Sound, SC (HIH)	1986	0	50

Location acronyms as used in text, tables, and figures are given below each locality.

*Includes material studied by Bohlmeyer & Gold (1991).

New alleles were detected at *ACP-2** (Allele *125*), *ADA** (Alleles *113* and *118*), and *PEPD** (Allele *75*).

χ^2 tests for deviations from expected (Hardy–Weinberg) equilibrium genotypic frequencies were carried out (i) by locality within year class, (ii) by locality with year classes pooled, (iii) by year class with localities pooled, and (iv) by region. When corrected for multiple tests, significant deviations ($P<0{\cdot}05$) from

TABLE II. Mean heterozygosity per locus (direct-count estimate) for red drum samples from the Gulf of Mexico

Locality	Year class	Heterozygosity ± S.E.
Sarasota Bay, FL (SAR)	1986	0·238 ± 0·073
	1987	0·204 ± 0·085
Riviera Bay, FL (RIV)	1986	0·224 ± 0·075
	1987	0·198 ± 0·085
Apalachicola Bay, FL (APP)	1986	0·233 ± 0·085
	1987	0·235 ± 0·070
Biloxi Bay, MS (OSP)	1986	0·226 ± 0·073
	1987	0·258 ± 0·074
Black Bay, LA (HOD)	1986	0·219 ± 0·074
Grand Isle, LA (GIL)	1986	0·222 ± 0·088
	1987	0·241 ± 0·082
Sabine Pass, TX (SAB)	1986	0·213 ± 0·084
	1987	0·247 ± 0·083
West Bay, TX (GVB)	1986	0·226 ± 0·085
	1987	0·204 ± 0·074
Pass Cavallo, TX (PCV)	1986	0·222 ± 0·083
	1987	0·190 ± 0·067
Redfish Bay, TX (PAR)	1986	0·232 ± 0·084
	1987	0·201 ± 0·077
Lower Laguna Madre, TX (LLM)	1986	0·235 ± 0·088
	1987	0·222 ± 0·080
1986 Year class		0·227 ± 0·078
1987 Year class		0·221 ± 0·075
Gulf of Mexico		0·226 ± 0·076*
Southeastern Atlantic		0·213 ± 0·074†

*Estimated average heterozygosity over 42 loci = 0·048 ± 0·021.
†Estimated average heterozygosity over 42 loci = 0·046 ± 0·020.

equilibrium expectations were found only at *sAAT-1** in the 1986 year class and the 1986 and 1987 year classes (pooled) at West Bay, TX (GVB), and at *ADH** in the 1986 year class at North Inlet, SC (GTN). The deviations at *sAAT-1** appear to be due to two rare homozygotes and one rare heterozygote for Allele *90* found in the 1986 year class; the deviation at *ADH** was not significant (P=0·255) when all genotypic classes were used to calculate χ^2 values (Bohlmeyer & Gold, 1991).

Observed heterozygosity-per-locus values, based on direct-count estimates and averaged over all polymorphic loci, are shown in Table II for (i) all Gulf sample localities by year class, (ii) the 1986 and 1987 year classes in the Gulf, and (iii) by region (i.e., Gulf and Atlantic). Values for Atlantic samples (not shown) were essentially identical to values for Gulf samples. None of the heterozygosity-per-locus values, either among Gulf or Atlantic sample localities by year class, between year classes from the Gulf, or between samples from the Gulf and samples from the Atlantic, differed significantly at the 5% level. Assuming the 33 monomorphic loci found by Bohlmeyer & Gold (1991) are monomorphic in all individuals surveyed, average heterozygosity (H) values (± S.E.) of red

TABLE III. Results of tests for spatial heterogeneity in allele frequencies at nine polymorphic loci among samples of red drum

Test group	No. of samples	No. of *G* tests	No. of significant *G* tests	F_{ST}	N_em
Gulf of Mexico:					
1986 Year class	11	9	0	0·015	>10
1987 Year class	10	9	1*	0·022	>10
1986+1987 Year classes	11	9	1**	0·010	>10
Gulf v. Atlantic:					
1986+1987 Year classes	16	9	2***	0·014	>10
Pooled	2	9	4****	0·003	>10

PEPD, $P \simeq 0{\cdot}005$.
***ADA**, $P \simeq 0{\cdot}03$†.
****ADA**, $P \simeq 0{\cdot}007$†; *EST-1**, $P \simeq 0{\cdot}025$†.
*****ADA**, $P < 0{\cdot}001$; *sAAT-1**, $P \simeq 0{\cdot}005$; *ADH**, $P \simeq 0{\cdot}025$†; *EST-1**, $P \simeq 0{\cdot}03$†.
†Non-significant ($P < 0{\cdot}05$) when corrected for multiple tests.

drum in the Gulf and in the Atlantic are $0{\cdot}048 \pm 0{\cdot}021$ and $0{\cdot}046 \pm 0{\cdot}020$, respectively.

G tests for temporal heterogeneity in allele frequencies were carried out (i) between samples from the 1986 and 1987 year classes at each locality in the Gulf, and (ii) between 1986 and 1987 year classes in the Gulf pooled over all localities. When corrected for multiple tests, significant heterogeneity was found only at *ADA** ($P \simeq 0{\cdot}004$ and $P < 0{\cdot}001$) between the 1986 and 1987 year class samples at Riviera Bay, FL (RIV) and between the 1986 and 1987 year classes (pooled), respectively. Additional *G* tests were conducted after pooling low frequency *ADA** alleles (i.e., Alleles *150*, *130*, *118*, *113*, *90*, *85*, *78*, and *75* pooled). Significant heterogeneity was again found between the 1986 and 1987 year class samples at RIV ($P < 0{\cdot}01$) and between the 1986 and 1987 year classes ($P < 0{\cdot}001$). *V* tests of arcsin square-root transformed allele frequencies indicated that the heterogeneity between year classes at RIV and between the 1986 and 1987 Gulf year classes was due primarily to an elevated frequency of *ADA*110* in the 1986 RIV sample. In the Atlantic, no heterogeneity in allele frequency was detected at any locus between the 1986 and 1987 year class samples from Charleston Bay, SC (CHS).

G tests for spatial heterogeneity in allele frequencies were carried out (i) among Gulf sample localities within each year class, (ii) among Gulf sample localities with year classes pooled, (iii) among Gulf and Atlantic sample localities with year classes pooled, and (iv) between pooled samples (1986 and 1987 year classes) from the Gulf *v.* those from the Atlantic. When corrected for multiple tests, significant heterogeneity (Table III) was found only at *PEPD** ($P \simeq 0{\cdot}005$) among samples from the 1987 year class in the Gulf, and at *ADA** ($P < 0{\cdot}001$) and *sAAT-1** ($P \simeq 0{\cdot}005$) between pooled samples from the Gulf *v.* pooled samples from the Atlantic. Additional *G* tests were carried out at *PEPD** among Gulf samples from the 1987 year class and at *ADA** and *sAAT-1** between pooled samples from the Gulf *v.* those from the Atlantic after pooling low frequency

TABLE IV. Results of hierarchical analysis of subdivision, Gulf of Mexico

Comparison	Variance component	Percent of variance
Year class within locality	0·00965	30·82
Year class within region	0·01101	35·15
Year class within total	0·00930	29·69
Locality within region	0·00136	4·34
Locality within total	−0·00036	0·00
Region within total	−0·00172	0·00

Locality=individual sample localities (10 total).
Region=East (SAR, RIV, APP), Central (OSP, GIL); West (SAB, GVB, PCV, PAR, LLM).

alleles: *PEPD** (Alleles *155*, *85* and *75* pooled), *ADA** (Alleles *150*, *130*, *118*, *113*, *90*, *85*, *78*, *75* and *65* pooled), and *sAAT-1** (Alleles *120* and *90* pooled). Significant heterogeneity was again found at *PEPD** ($P \simeq 0{\cdot}015$) among the 1987 Gulf year class, and at *ADA** ($P < 0{\cdot}001$) and *sAAT-1** ($P \simeq 0{\cdot}016$) between pooled Gulf *v.* pooled Atlantic samples. The heterogeneity at *PEPD** in the 1987 year class from the Gulf is clearly due to an elevated frequency of Allele *115* in the sample from Bioloxi Bay, MS (OSP, Appendix, Table II) and is not indicative of a general trend of spatial genetic differentiation among samples from the Gulf. The heterogeneity at *ADA** and *sAAT-1** between pooled samples from the Gulf and pooled samples from the Atlantic appears to be due to cumulative frequency differences at several alleles at both loci (Appendix Table III). Bohlmeyer & Gold (1991) found significant heterogeneity at *ACP-2** among five geographic samples from the Atlantic. For reasons discussed in Bohlmeyer & Gold (1991), the heterogeneity at *ACP-2** among Atlantic samples was presumed to stem from sampling error. Estimates of F_{ST} and $N_e m$ for geographic (spatial) samples are also shown in Table III. Mean F_{ST} values (averaged over all loci) ranged from 0·003 in the pooled Gulf *v.* Atlantic comparison to 0·022 in the comparison among Gulf localities in the 1987 year class. $N_e m$ values, derived from mean F_{ST} values using the island model of Wright (1943), were greater than 10 for all spatial comparisons (Table III).

Hierarchical gene-diversity analysis was used to partition genetic diversity among samples from the 1986 and 1987 year classes in the Gulf. Individual year classes comprised the lowest level in the analysis and were partitioned into individual sample localities and three geographic regions: eastern Gulf [Sarasota Bay, FL (SAR), Riviera Bay, FL (RIV) and Apalachicola Bay, FL (APP)]; central Gulf [Bioloxi Bay, MS (OSP) and Grand Isle, LA (GIL)]; and western Gulf [Sabine Pass, TX (SAB), West Bay, TX (GVB), Pass Cavallo, TX (PCV), Redfish Bay, TX (PAR), and Lower Laguna Madre, TX (LLM)]. Approximately 96% of the total gene diversity occurred among year classes within locality, region, and total; whereas only 4% of the total gene diversity occurred among localities within regions (Table IV). None of the total sampling variance

TABLE V. Results of hierarchical subdivision, Gulf of Mexico *v.* Atlantic

Comparison	Variance component	Percent of variance
Locality within region	0·00477	30·04
Locality within total	0·00794	50·00
Region within total	0·00317	19·96

Locality=individual sample localities (16 total).
Region=Gulf of Mexico (11 localities) and Atlantic (five localities).

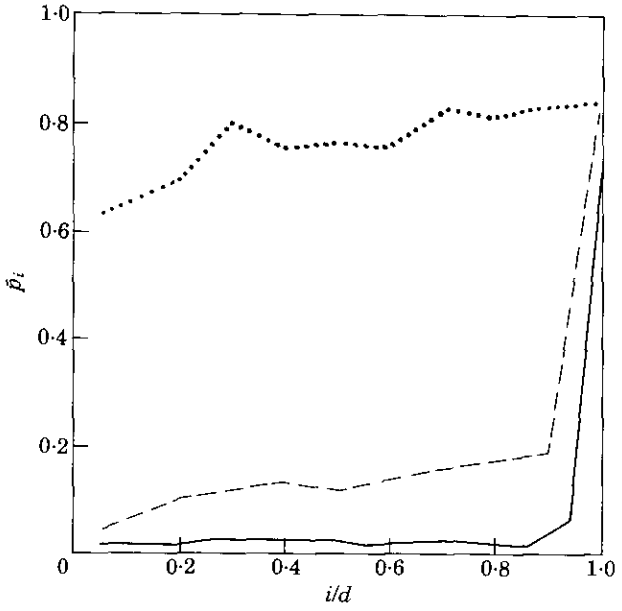

FIG. 2. Qualitative profile of gene flow, ——. Values were obtained by plotting conditional-average allele frequencies ($\bar{p}_i$) against occupancy rate (i/d). Also shown are theoretical profiles (after Slatkin, 1981) when $N_e m$ (effective number of migrants per generation) equals 1·250, – – – (high gene flow) and 0·025, · · · · (low gene flow).

was explained by among locality or among region effects, i.e., variation among localities and regions primarily reflects sampling variances.

Hierarchical gene-diversity analysis was also used to partition genetic diversity among pooled samples from the 1986 and 1987 year classes in the Gulf and Atlantic. Individual sample localities (11 in the Gulf and five in the Atlantic) comprised the lowest level and were partitioned into two regions (Gulf and Atlantic). Nearly 80% of the total gene diversity occurred among localities, whereas nearly 20% of the total gene diversity occurred between regions (Table V). These results corroborate the tests of allele frequency heterogeneity: red drum in the Gulf are differentiated genetically from red drum in the Atlantic.

Slatkin's (1981) qualitative method was used to assess gene flow among sample localities (both Gulf and Atlantic) representing the 1986 and 1987 year classes

separately and combined. The graph shown in Fig. 2 was derived from conditional average allele frequencies in all Gulf and Atlantic sample localities (1986 and 1987 year classes combined) and was identical to graphs derived in the same way using the 1986 and 1987 year classes separately. The graph is typical of species with high gene flow. Slatkin's (1985) quantitative method of estimating N_em could not be used as no private alleles (*sensu* Slatkin, 1985) were found at any locality. The geographic distribution of rare alleles (defined here as alleles occurring in less than 2% of all individuals examined) is shown in Table VI. All rare alleles, even those (*ACP-2*125*, *ADA*150*, *ADA*113*, *ADA*78*, *GPI-B*110* and *PEPS*95*) which occur at very low frequency, were found at non-neighbouring localities, and in some cases (*ACP-2*125*, *ADA*150*, *ADA*113* and *PEPS*95*), were found at widely separated localities. These data also suggest high levels of gene flow among geographic localities.

To explore further the possibility of regional subdivision within the Gulf, samples from both the 1986 and 1987 year classes were pooled by locality and localities were pooled into the three regions described above (eastern Gulf, central Gulf, and western Gulf). The 1986 year class sample from Black Bay, LA (HOD) was included in the central Gulf. Allele frequencies among the three regions were tested for heterogeneity using *G* tests. Significant heterogeneity was detected at two loci: *ADA** ($P<0{\cdot}001$) and *PEPD** ($P\simeq0{\cdot}006$). Arcsin square-root transformed frequencies of Alleles *125*, *115*, *110*, and *100* at *ADA* and Alleles *115* and *100* at *PEPD** were then tested for heterogeneity among the three regions using *V* tests. When corrected for multiple tests, significant heterogeneity among regions was found for *ADA*110* ($P\simeq0{\cdot}001$) and *PEPD*115* ($P\simeq0{\cdot}004$). The possibility that significant heterogeneity could be detected by chance alone was tested by randomly assigning the 11 geographic localities into 10 random groupings of three, three and five localities, thus mimicking the original groupings in terms of number of localities per grouping. Allele frequencies at *ADA** and *PEPD** in each of the 10 random groupings (designated RG 1–10) were then tested for heterogeneity using *G* tests. *V* tests were used to test for heterogeneity of arcsin square-root transformed frequencies of Alleles *125*, *115*, *110* and *100* at *ADA** and Alleles *115* and *100* at *PEPD**. Significant heterogeneity ($P<0{\cdot}05$) in *G* tests was found in four of 10 random groupings at *ADA** and at nine of 10 random groupings at *PEPD** (Table VII). When corrected for multiple tests, significant heterogeneity in *V*-tests was found at either Allele *115* or *110* at *ADA** in three of 10 random groupings and at Allele *115* at *PEPD** in five of 10 random groupings (Table VII). These results indicate that the heterogeneity detected among the original eastern-central-western groupings could be due to chance alone rather than geographic subdivision.

Matrices of Nei's unbiased genetic distance indices were generated (i) among all Gulf and Atlantic sample localities by year class, and (ii) among all Gulf and Atlantic sample localities with year classes at each locality pooled. In UPGMA-derived phenograms of both genetic distance matrices, standard errors of the most distant nodes were greater than the distance between the first and last nodes, effectively collapsing all nodes and demonstrating that all samples of red drum (by year class or with year classes combined) are not strongly differentiated genetically. The UPGMA phenogram derived from the genetic distance matrix of all Gulf and Atlantic sample localities (year classes pooled) is shown in Fig. 3.

TABLE VI. Geographic distribution of 17 rare† allozyme alleles among 16 geographic samples of red drum. Numbers represent total numbers of each allele found at a locality

Locus allele	Atlantic samples					Gulf samples										
	MNC	BTH	GTN	CHS	HIH	SAR	RIV	APP	OSP	HOD	GIL	SAB	GVB	PCV	PAR	LLM
*ACP-2*125*	—	—	—	—	—	1	—	—	—	—	—	1	—	—	—	—
*ADA*150*	—	1	—	—	—	—	—	—	—	—	2	—	—	—	—	—
*ADA*118*	1	—	—	1	—	1	1	1	—	—	1	—	—	—	—	—
*ADA*113*	—	—	—	1	—	—	—	1	—	—	2	—	—	—	—	—
*ADA*90*	—	—	—	—	1	2	1	2	5	3	8	2	1	—	1	1
*ADA*85*	—	—	—	1	—	4	7	1	4	2	4	1	1	—	1	1
*ADA*78*	—	—	—	—	—	—	—	—	—	—	1	—	1	—	—	—
*ADA*65*	—	—	—	—	—	1	—	1	2	—	3	—	2	—	—	—
ADH-20*	—	1	—	3	2	2	—	1	—	—	—	1	1	—	—	1
*sAAT-1*120*	1	1	—	3	1	—	—	—	—	—	1	1	—	—	—	1
*sAAT-1*90*	—	—	1	2	—	2	—	3	—	—	2	1	1	—	—	—
GPI-B-110*	—	—	—	1	—	2	—	—	—	1	—	—	—	—	—	—
*PEPB*115*	1	—	—	—	1	4	3	4	2	1	1	—	—	1	1	1
*PEPB*85*	—	—	—	—	2	2	—	—	1	—	—	—	2	—	—	1
*PEPD*115*	—	1	—	1	1	1	—	—	10	1	2	—	1	—	1	1
*PEPD*75*	—	—	—	1	—	—	—	—	2	—	—	—	—	—	—	—
*PEPS*95*	—	—	—	—	—	—	2	—	—	—	1	—	—	1	—	—

†Rare alleles are defined here as alleles occurring at a frequency of less than 2% over all samples.

TABLE VII. Results of tests of geographic subdivision in allele frequencies at two polymorphic loci among red drum in the Gulf of Mexico

Test group/ Locus†	Results of *G* tests (*P* values)	No. of alleles tested	No. of significant *V* tests
ECW/*ADA**	<0·001	4	3§
RG1/*ADA**	>0·050	4	0
RG2/*ADA**	>0·050	4	1‡
RG3/*ADA**	>0·050	4	1‡
RG4/*ADA**	<0·001	4	2‡
RG5/*ADA**	≃0·015	4	1
RG6/*ADA**	≃0·030	4	2§
RG7/*ADA**	>0·050	4	2§
RG8/*ADA**	>0·050	4	1‡
RG9/*ADA**	≃0·020	4	1‡
RG10/*ADA**	>0·050	4	1‡
ECW/*PEPD**	≃0·006	2	2§
RG1/*PEPD**	≃0·008	2	2‡
RG2/*PEPD**	>0·050	2	0
RG3/*PEPD**	≃0·020	2	1‡
RG4/*PEPD**	≃0·020	2	1‡
RG5/*PEPD**	≃0·015	2	1‡
RG6/*PEPD**	≃0·008	2	1
RG7/*PEPD**	≃0·045	2	1
RG8/*PEPD**	≃0·020	2	1
RG9/*PEPD**	≃0·018	2	1
RG10/*PEPD**	≃0·038	2	2‖

†ECW=East (SAR, RIV, APP), Central (OSP, HOD, GIL); West (SAB, GVB, PCV, PAR, LLM); RG1–10=random groupings of three, three, and five samples.

‡Non-significant ($P>0·05$) when corrected for multiple tests.

§One of three tests significant ($P<0·05$) when corrected for multiple tests.

‖One of two tests significant ($P<0·05$) when corrected for multiple tests.

Spatial autocorrelation analyses were used to determine whether allele frequencies at any Gulf locality (year classes pooled) were independent of allele frequencies at neighbouring localities. Only alleles whose frequency was greater than 4% in any 1986 or 1987 Gulf year class sample were used in order to minimize noise. Alleles used in autocorrelation analysis were: Alleles *115* and *110* at *ACP-2**, Alleles *125*, *115*, *110*, and *100* at *ADA**, Alleles *−100* and *−75* at *ADH**, Alleles *110* and *100* at *sAAT-1**, Allele *100* at *EST-1**, Allele *−100* at *GPI-B**, Allele *100* at *PEPB**, Alleles *100* and *85* at *PEPD**, and Allele *100* at *PEPS**. SAAP runs using five distance classes generated 80 Moran's I values (16 alleles × five distance classes). When equal numbers of pairwise comparisons were used, 10 significant ($P<0·05$) values were obtained; one significant (positive) value was found in the second distance class; two significant (negative) values were found in both the third and fourth distance classes; and five significant

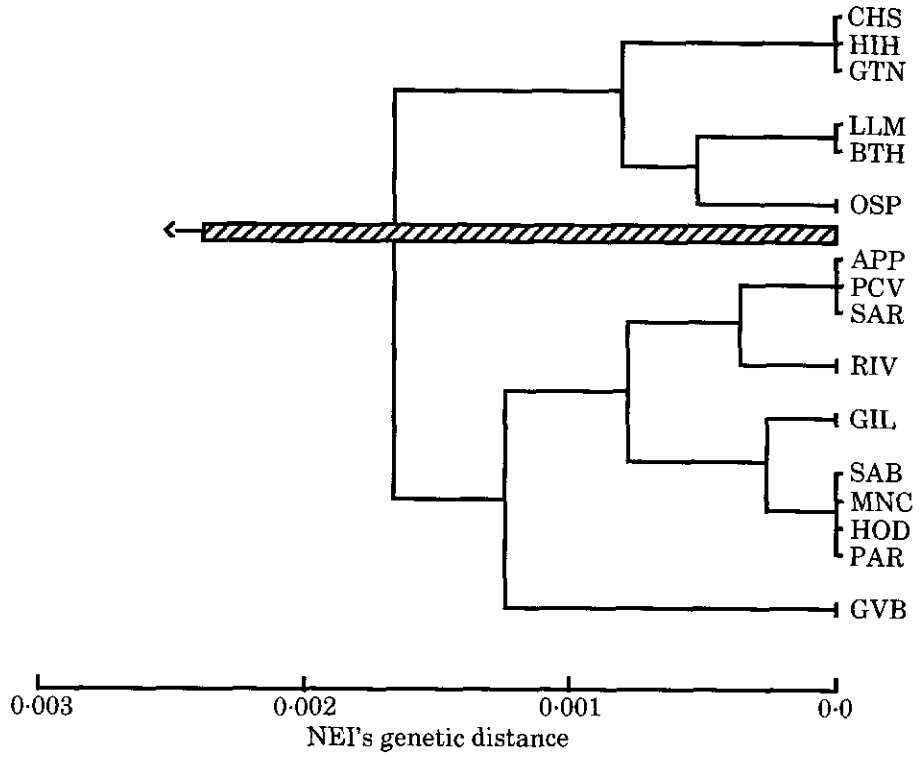

FIG. 3. UPGMA-derived phenogram of Nei's unbiased genetic distances. Operational units are all Gulf and Atlantic sample localities (1986 and 1987 year classes pooled). Hatched bar is the standard error of the last node in the phenogram.

(three positive and two negative) values were found in the last distance class. When equal geographic distances were used, six significant ($P<0{\cdot}05$) values were obtained: one significant (negative) value was found in the first distance class; one significant (negative) value was found in the third distance class; one significant (positive) value was found in the fourth distance class; and three (one positive and two negative) values were found in the last distance class. These findings indicate that allele frequencies at any Gulf locality are essentially independent of allele frequencies at neighbouring or distant localities. Graphical representations of these results are shown in Fig. 4, where mean (± 2 S.E.) Moran's I values, averaged over all 16 alleles, are plotted by distance class. In both SAAP runs [i.e., using equal frequencies, Fig. 4(a), or using equal distances, Fig. 4(b)], mean Moran's I values are essentially equivalent in both the first and last distance classes and are not significantly different from zero.

IV. DISCUSSION

Estimates of genome-wide variation based on nuclear-gene (allozyme) loci indicate that red drum in both the Gulf and Atlantic have levels of genetic variation typical of many vertebrate species. Ramsey & Wakeman (1987) examined 537 (primarily Gulf) red drum for 40 presumptive loci and found an average heterozygosity of 0·029. Average heterozygosity values of 0·048 (Gulf

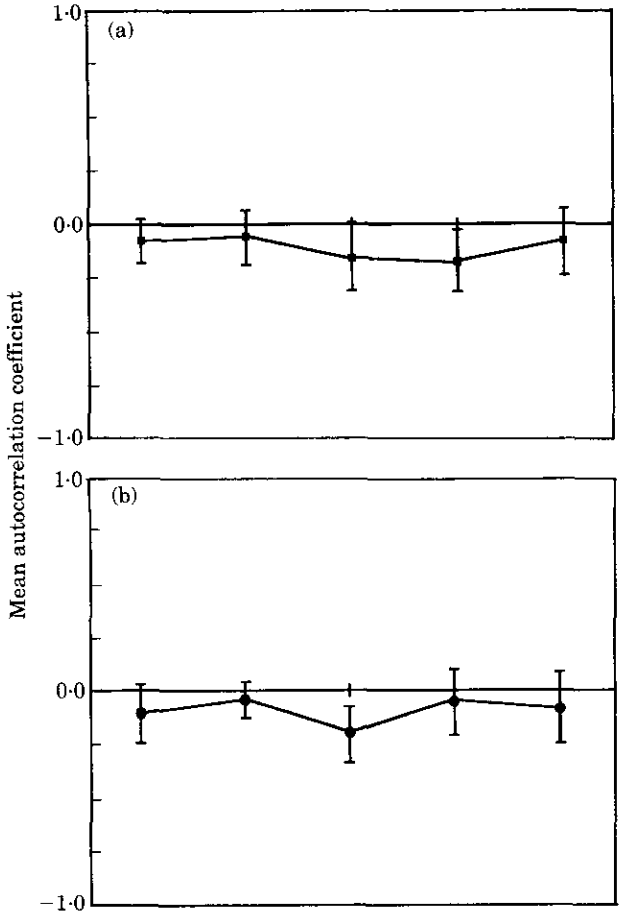

FIG. 4. Correlograms based on frequencies of 16 common alleles at nine polymorphic loci among 11 geographic samples of red drum from the Gulf. Abscissa: distance classes 1–5 (left to right). Ordinate: mean autocorrelation coefficients (Moran's *I* values) for each distance class. (a) Equal frequencies/distance class; (b) equal distances between distance classes.

red drum) and 0·046 (Atlantic red drum) found in this study for the 1986 and 1987 year classes (combined) do not differ significantly (Student's *t* tests, $P>0·05$) from the value reported by Ramsey & Wakeman (1987), and agree closely with values reported in a broad variety of vertebrates (Nevo, 1978), including other marine fishes (Winans, 1980; Smith & Fujio, 1982; Shaklee & Samollow, 1984; Milton & Shaklee, 1987). These findings suggest that the declines in red drum abundance (Matlock, 1984; Reagan, 1985) have probably not affected genetic diversity in the species.

Ramsey & Wakeman (1987) detected heterogeneity at a locus for glucose phosphate isomerase. The largest difference in allele frequencies, however, occurred between adjacent sample localities and no simple pattern of geographic differentiation was evident. Bohlmeyer & Gold (1991) found significant heterogeneity at the *ADA** locus among samples from the Gulf and between pooled samples from the Gulf *v.* pooled samples from the Atlantic. The heterogeneity

at *ADA** among samples from the Gulf appeared to stem from frequency differences at three rare *ADA** alleles (Alleles *110*, *115*, and *125*), two of which (Alleles *110* and *125*) appeared to exhibit a weak, east–west cline. Bohlmeyer & Gold (1991) noted, however, that sampling distributions given by the null hypothesis do not necessarily approximate a χ^2 distribution when alleles are rare (Chakraborty & Leimar, 1987), and moreover, that heterogeneity tests using single alleles at the same locus were not independent. As a consequence, they reached the conservative conclusion that the only unequivocal heterogeneity observed was that between Gulf and Atlantic red drum.

Results of the present study indicate the effective absence of both temporal and spatial genetic differentiation among red drum from the 1986 and 1987 year classes in the Gulf. Genotypes at nearly all loci within year classes and across the Gulf were in Hardy–Weinberg equilibrium, tests of temporal heterogeneity revealed a significant difference only in the frequency of one rare allele (*ADA*115*), and tests of spatial heterogeneity revealed a significant difference only in the frequency of one rare allele (*PEPD*115*) in the 1987 year class. In addition, F_{ST} (and N_em) values and Slatkin's (1981) qualitative graphic analysis indicated high levels of gene flow among all Gulf localities, there were no spatial groupings of rare alleles, and hierarchical analysis demonstrated that 96% of the genetic diversity was contained within individual year class samples. Finally, cluster analysis indicated that all Gulf samples were not strongly differentiated genetically and there was no significant spatial autocorrelation of allele frequencies. Slight, regionally-based genetic differentiation at two loci (*ADA** and *PEPD**) was suggested in heterogeneity tests involving pooled samples from three regions (eastern-central-western) within the Gulf. A bootstrap analysis, however, indicated that the heterogeneity detected could be due to chance alone rather than geographic subdivision. Given the above, the heterogeneity at *ADA** among the 1986 Gulf year class reported by Bohlmeyer & Gold (1991) appears to have been due to insufficient sampling or sampling error.

Red drum in the Gulf appear to be differentiated genetically from red drum in the Atlantic. Heterogeneity tests revealed significant differences in allele frequencies at two loci (*ADA** and *sAAT-1**) between pooled samples from the Gulf *v.* pooled samples from the Atlantic, and hierarchical gene-diversity analysis indicated that almost 20% of the genetic diversity was contained within regions (Gulf and Atlantic). Cluster analysis, however, indicated that red drum at individual sampling localities in the Gulf and Atlantic were not strongly differentiated genetically, and F_{ST} (and N_em) values, Slatkin's qualitative graphic analysis, and the absence of spatial groupings of rare alleles indicated that levels of gene flow between red drum in the Gulf and Atlantic are relatively high. Heterogeneity in allele frequencies, however, is not necessarily inconsistent with high levels of gene flow, small values of F can be associated with genetic differentiation, and genetic divergence can occur even with substantial gene exchange (Wright, 1969; Allendorf & Phelps, 1981).

Concordance of both nuclear-gene (allozyme) and mtDNA data sets (Gold & Richardson, 1991; this paper) strongly suggests the genetic discontinuity between Gulf and Atlantic red drum is real and not an artifact or accident of sampling. This finding is somewhat curious given several aspects of red drum biology which should facilitate dispersal and minimize spatial differentiation, and the much

greater geographic distance between sample localities in the Gulf (Sarasota Bay, FL *v.* the Lower Laguna Madre, TX) as compared to that between the easternmost sample locality in the Gulf (Sarasota Bay, FL) and the southernmost sample locality in the Atlantic (Calibogue Sound, SC). Red drum are long-lived, relatively strong swimmers that are capable of spawning at multiple localities throughout their lifetime (Matlock, 1987*b*). Moreover, at least some adult red drum form large schools offshore and are capable of extensive, long-distance migrations (Overstreet, 1983; Swingle *et al.*, 1984; Matlock, 1987*b*; Murphy & Taylor, 1990). In other marine fishes with similar life-histories, genetic divergence among discrete geographic localities is typically low and most genetic variation occurs within localities (Winans, 1980; Graves *et al.*, 1984; Gyllensten, 1985; Avise *et al.*, 1987).

The genetic difference between Gulf and Atlantic red drum likely stems from historical or recent interactions between dispersal and impediments to gene flow (Gold & Richardson, 1991). Possible impediments could be biogeographic and/or ecological in form. There are identifiable biogeographic provinces that separate Gulf from Atlantic marine fauna (Avise *et al.*, 1987), and several marine and freshwater species appear to be comprised of regional subpopulations in Gulf *v.* Atlantic coast areas (Avise, 1992). Alternatively, there are marine fish species found in nearshore areas of both the Gulf and Atlantic coast that exhibit little or no genetic discontinuity (Avise *et al.*, 1987). Another possibility might be that offshore currents utilized by red drum are not conducive to movement between the Gulf and Atlantic. Recent data on patterns of offshore surface currents in the western Atlantic (E. Waddell, pers. commun.) suggest that dispersal of red drum from the Atlantic into the Gulf would be less likely than the reverse. One final possibility might be the absence of suitable nearshore or other habitat for red drum along the southeastern (Atlantic) coast of Florida. Relatively few red drum are found from Fort Pierce, FL, southward to the northern part of the Florida Keys, a region that includes the Biscayne Bay system where red drum have not been observed for over 50 years (R. Taylor, pers. commun.). North of Biscayne Bay, red drum are common in the Mosquito Lagoon region near Cape Canaveral. Red drum samples from the Mosquito Lagoon region are now being surveyed in our laboratory for both allozyme and mtDNA phenotypes.

Direct and indirect assistance in procuring red drum specimens was provided by individuals from the following agencies or research organizations: North Carolina Division of Marine Fisheries, University of South Carolina, South Carolina Wildlife, Florida Department of Natural Resources, Gulf Coast Research Laboratory, Louisiana State University, Coastal Fisheries Branch of the Texas Parks and Wildlife Department, and the National Marine Fisheries Service. Their assistance is gratefully acknowledged. Special thanks are extended to C. Amemiya, C. Bailey, R. Cheramie, F. Deal, L. McEachron, T. McIlwain, M. Murphy, D. Roberts, B. Roumillat, and C. Wilson either for providing no-cost lodging during field trips or for arranging field details. We also thank R. Colura and B. Bumguardner for carrying out age determinations for otoliths, I. Blandon, G. Ramos, and E. Young for assistance in the laboratory, T. Dowling for providing the computer software used for conducting V tests and UPGMA clustering, and E. Waddell for sharing unpublished data on current patterns in the western Atlantic. Work was supported by the Sea Grant College Program (grants NA85AA-D-SG128 and NA89AA-D-SG139), the MARFIN Program of the U.S. Department of Commerce (grants NA89-WC-H-MF025 and NA90AA-H-MF107), the Coastal Fisheries Branch of

the Texas Parks and Wildlife Department, and the Texas Agricultural Experiment Station (Project H-6703). The views expressed in the paper are those of the authors and do not necessarily reflect the views of the National Oceanic and Atmospheric Administration or any of its sub-agencies. This paper represents number VII in the series 'Genetic studies in marine fishes' and Contribution Number 27 of the Center for Biosystematics and Biodiversity at Texas A&M University.

References

Adkins, G., Tarver, J., Bowman, P. & Savoie, B. (1979). A study of commercial finfish in coastal Louisiana. Baton Rouge, LA: *Technical Bulletin of the Louisiana Department of Wildlife and Fisheries*, **July,** 1–92.

Allendorf, F. W. & Phelps, S. R. (1981). Use of allelic frequencies to describe population structure. *Canadian Journal of Fisheries and Aquatic Sciences* **38,** 1507–1514.

Avise, J. C. (1992). Molecular population structure and the biogeographic history of a regional fauna: a case history with lessons for conservation biology. *Oikos* **63,** 62–76.

Avise, J. C., Arnold, J., Ball, R. M., Bermingham, E., Lamb, T., Neigel, J. E., Reeb, C. A. & Saunders, N. C. (1987). Intra-specific phylogeography: the mitochondrial DNA bridge between population genetics and systematics. *Annual Review of Ecology and Systematics* **18,** 489–522.

Bohlmeyer, D. A. (1989). A protein electrophoretic analysis of population structure in the red drum (*Sciaenops ocellatus*). MS thesis. College Station, Texas: Texas A&M University.

Bohlmeyer, D. A. & Gold, J. R. (1991). Genetic studies in marine fishes. II. A protein electrophoretic analysis of population structure in the red drum *Sciaenops ocellatus*. *Marine Biology* **108,** 197–206.

Bumguardner, B. W. (1991). Marking subadult red drums with oxytetracycline. *Transactions of the American Fisheries Society* **120,** 537–540.

Chakraborty, R. & Leimar, O. (1987). Genetic variation within a subdivided population. In *Population Genetics and Fishery Management* (Ryman, N. & Utter, F., eds), pp. 89–120. Seattle, WA: University of Washington Press.

Chamberlain, G. W., Miget, R. J. & Haby, M. G., Eds (1987). *Manual on Red Drum Aquaculture*. College Station, TX: Texas Agriculture Extension Service and Sea Grant College Program, Texas A&M University.

Cooper, D. W. (1968). The significance level in multiple tests made simultaneously. *Heredity* **23,** 614–617.

DeSalle, R., Templeton, A., Mori, I., Pletscher, S. & Johnston, J. S. (1987). Temporal and spatial heterogeneity of mtDNA polymorphisms in natural populations of *Drosophila mercatorum*. *Genetics* **116,** 215–233.

Gold, J. R. & Richardson, L. R. (1991). Genetic studies in marine fishes. IV. An analysis of population structure in the red drum (*Sciaenops ocellatus*) using mitochondrial DNA. *Fisheries Research* **12,** 213–241.

Graves, J. E., Ferris, S. D. & Dizon, A. E. (1984). Close genetic similarity of Atlantic and Pacific skipjack tuna (*Katsuwonus pelamis*) demonstrated with restriction endonuclease analysis of mitochondrial DNA. *Marine Biology* **79,** 315–319.

Gyllensten, U. (1985). The genetic structure of fish: differences in the intraspecific distribution of biochemical genetic variation between marine, anadromous, and freshwater species. *Journal of Fish Biology* **26,** 691–699.

Lyczkowski-Schultz, J., Steen, JR., J. P. & Comyns, B. H. (1988). Early life history of red drum (*Sciaenops ocellatus*) in the northcentral Gulf of Mexico. Final Report, Mississippi-Alabama Sea Grant Consortium, Project No. R/LR-12. Ocean Springs, Mississippi.

Matlock, G. C. (1984). A basis for the development of a management plan for red drum in Texas. Ph.D. dissertation, College Station, TX: Texas A&M University.

Matlock, G. C. (1987*a*). Maximum total length and age of red drum off Texas. *Northeast Gulf Science* **9,** 49–52.

Matlock, G. C. (1987*b*). The life history of the red drum. In *Manual on Red Drum Aquaculture* (Chamberlain, G. W., Miget, R. J. & Haby, M. G., eds), pp. 1–47. College Station, TX: Texas Agricultural Extension Service and Sea Grant College Program, Texas A&M University.

Matlock, G. C. & Weaver, J. E. (1979). *Fish Tagging in Texas Bays during November 1975–September 1976.* Austin, TX: Texas Parks and Wildlife Department, Coastal Fisheries Branch, Management Data Series No. 134.

McIlwain, T., McEachron, L., Murphy, M. D., Nelson, W. R., Shepard, J., Van Hoose, M., Condrey, R., Bane, N. & Becker, R. E. (1986). *State–Federal Cooperative Program for Red Drum Research in the Gulf of Mexico.* Ocean Springs, MS: Gulf States Marine Fisheries Commission.

Mercer, L. (1984). *A Biological and Fisheries Profile of Red Drum,* Sciaenops ocellatus. Special Scientific Report, North Carolina Department of Natural Resources and Community Development, Morehead City, NC: Division of Marine Fisheries **41,** 1–89.

Milton, D. A. & Shaklee, J. B. (1987). Biochemical genetics and population structure of blue grenadier, *Macruronus novaezelandiae* (Hector) (Pisces: Merluccidae), from Australian waters. *Australian Journal of Freshwater Research* **38,** 727–742.

Murphy, M. D. & Taylor, R. G. (1990). Reproduction, growth, and mortality of red drum, *Sciaenops ocellatus,* in Florida. *Fishery Bulletin* **88,** 531–542.

Nei, M. (1978). Estimation of average heterozygosity and genetic distance from a small number of individuals. *Genetics* **89,** 583–590.

Nei, M., Stephens, J. C. & Saitou, N. (1985). Methods for computing the standard errors of branching points in an evolutionary tree and their application to molecular data from humans and apes. *Molecular Biology and Evolution* **2,** 66–85.

Nevo, E. (1978). Genetic variation in natural populations: patterns and theory. *Theoretical Population Biology* **13,** 121–177.

Osburn, H. R., Matlock, G. C. & Green, A. W. (1982). Red drum (*Sciaenops ocellatus*) movement in Texas bays. *Contributions in Marine Science* **25,** 85–97.

Overstreet, R. M. (1983). Aspects of the biology of the red drum, *Sciaenops ocellatus,* in Mississippi. *Gulf Research Reports* (supplement) **1,** 45–68.

Ramsey, P. R. & Wakeman, J. M. (1987). Population structure of *Sciaenops ocellatus* and *Cynoscion nebulosus* (Pisces: Sciaenidae): biochemical variation, genetic subdivision and dispersal. *Copeia* **1987,** 682–695.

Reagan, R. E. (1985). Species profiles: life histories and environmental requirements of coastal fishes and invertebrates (Gulf of Mexico)—red drum. *U.S. Fish and Wildlife Service Biological Report* **82,** (11.36), 1–16 (U.S. Army Corps Engineers, Reference Number TR EL-82-4). Washington, WA: U.S. Fish & Wildlife Service.

Rohlf, F. J. (1983). *BIOM-PC: A Package of Statistical Programs to Accompany the Text Biometry.* San Francisco, CA: W. H. Freeman & Company.

Ross, J. L., Stephens, T. M., McKenna, S. A. & Burns, B. L. (1987). *Red Drum, Sciaenops ocellatus, Tagging in North Carolina Waters.* Report of the North Carolina Department of Natural Resources and Community Development, Morehead City, NC: Division of Marine Fisheries, 1–44.

Shaklee, J. B. & Samollow, P. B. (1984). Genetic variation and population structure in a deepwater snapper, *Pristipomoides filamentosus,* in the Hawaiian archipelago. *Fishery Bulletin* **82,** 703–713.

Slatkin, M. (1981). Estimating levels of gene flow in natural populations. *Genetics* **95,** 323–335.

Slatkin, M. (1985). Rare alleles as indiators of gene flow. *Evolution* **39,** 53–65.

Smith, P. J. & Fujio, Y. (1982). Genetic variation in marine teleosts: high variability in habitat specialists and low variability in habitat generalists. *Marine Biology* **69,** 7–20.

Sneath, P. H. A. & Sokal, R. R. (1973). *Numerical Taxonomy.* San Francisco, CA: W. H. Freeman & Company.

Sokal, R. R. & Oden, N. L. (1978*a*). Spatial autocorrelation in biology. I. Methodology. *Biological Journal of the Linnean Society* **10,** 199–228.

Sokal, R. R. & Oden, N. L. (1978*b*). Spatial autocorrelation in biology. 2. Some biological implications and four applications of evolutionary and ecological interest. *Biological Journal of the Linnean Society* **10,** 229–249.

Sokal, R. R. & Rohlf, F. J. (1969). *Biometry.* San Francisco, CA: W. H. Freeman & Company.

Swingle, W. E. (1987). Status of the commercial and recreational fishery. In *Manual on Red Drum Aquaculture* (Chamberlain, G. W., Miget, R. J. & Haby, M. G., eds), pp. 46–49. College Station, TX: Texas Agricultural Extension Service and Sea Grant College Program, Texas A&M University.

Swingle, W., Leary, T., Davis, D., Blomo, V., Tatum, W., Murphy, M., Taylor, R., Adkins, G., McIlwain, T. & Matlock, G. (1984). *Fishery Profile of Red Drum.* Ocean Springs, MS: Gulf States Marine Fisheries Commission.

Swofford, D. L. & Selander, R. B. (1981). BIOSYS-1: a FORTRAN program for the comprehensive analysis of electrophoretic data in population genetics and systematics. *Journal of Heredity* **72,** 281–283.

Wartenberg, D. (1989). *SAAP: A Spatial Autocorrelation Analysis Program.* Piscataway, NJ: Department of Environmental and Community Medicine, Robert Wood Johnson Medical School, University of Medicine and Dentistry of New Jersey.

Winans, G. A. (1980). Geographic variation in the milkfish *Chanos chanos.* I. Biochemical evidence. *Evolution* **34,** 558–574.

Wright, S. (1943). Isolation by distance. *Genetics* **28,** 114–138.

Wright, S. (1969). *Evolution and the Genetics of Populations,* Vol. II. *The Theory of Gene Frequencies.* Chicago, IL: University of Chicago Press.

APPENDIX I. Allele frequencies at nine polymorphic loci among sample localities from the 1986 year class, Gulf of Mexico and Atlantic Ocean

Locus allele	Atlantic samples					Gulf samples									
	MNC	BTH	GTN	CHS	HIH	SAR	RIV	APP	OSP	HOD	GIL	SAB	GVB	PCV	PAR	LLM
*ACP-2**																
125	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000
115	0·167	0·000	0·083	0·088	0·050	0·100	0·083	0·133	0·079	0·133	0·101	0·080	0·094	0·125	0·129	0·028
100	0·833	1·000	0·917	0·912	0·950	0·900	0·917	0·867	0·921	0·867	0·899	0·920	0·906	0·875	0·871	0·972
(*n*)	(15)	(23)	(18)	(34)	(50)	(85)	(24)	(30)	(76)	(45)	(74)	(25)	(32)	(12)	(35)	(18)
*ADA**																
150	0·000	0·022	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·007	0·000	0·000	0·000	0·000	0·000
130	0·000	0·000	0·056	0·059	0·040	0·047	0·042	0·017	0·051	0·030	0·047	0·020	0·016	0·125	0·029	0·028
125	0·400	0·370	0·360	0·324	0·330	0·284	0·229	0·417	0·346	0·410	0·304	0·480	0·421	0·375	0·357	0·500
118	0·000	0·000	0·000	0·000	0·000	0·006	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000
115	0·000	0·000	0·000	0·044	0·000	0·047	0·063	0·033	0·096	0·040	0·149	0·080	0·156	0·042	0·114	0·000
113	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·007	0·000	0·000	0·000	0·000	0·000
110	0·067	0·109	0·056	0·044	0·090	0·093	0·125	0·033	0·051	0·140	0·068	0·000	0·063	0·042	0·029	0·056
100	0·500	0·477	0·500	0·485	0·500	0·482	0·354	0·433	0·378	0·310	0·350	0·320	0·297	0·416	0·429	0·332
90	0·000	0·000	0·000	0·000	0·010	0·012	0·021	0·017	0·013	0·030	0·027	0·040	0·016	0·000	0·014	0·028
85	0·000	0·000	0·000	0·015	0·000	0·017	0·083	0·000	0·026	0·020	0·007	0·000	0·000	0·000	0·014	0·028
78	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·007	0·000	0·000	0·000	0·000	0·000
75	0·033	0·022	0·028	0·029	0·030	0·006	0·083	0·033	0·026	0·020	0·020	0·060	0·000	0·000	0·014	0·028
65	0·000	0·000	0·000	0·000	0·000	0·006	0·000	0·017	0·013	0·000	0·007	0·000	0·031	0·000	0·000	0·000
(*n*)	(15)	(23)	(18)	(34)	(50)	(86)	(24)	(30)	(78)	(50)	(74)	(25)	(32)	(12)	(35)	(18)
*ADH**																
− 100	0·600	0·630	0·666	0·529	0·550	0·500	0·354	0·550	0·555	0·500	0·527	0·420	0·453	0·375	0·515	0·527
− 75	0·400	0·327	0·278	0·441	0·410	0·459	0·604	0·433	0·411	0·480	0·426	0·500	0·516	0·583	0·414	0·417
− 50	0·000	0·043	0·000	0·015	0·020	0·035	0·042	0·017	0·034	0·020	0·047	0·060	0·031	0·042	0·071	0·056
− 20	0·000	0·000	0·056	0·015	0·020	0·006	0·000	0·000	0·000	0·000	0·000	0·020	0·000	0·000	0·000	0·000
(*n*)	(15)	(23)	(18)	(34)	(50)	(85)	(24)	(30)	(73)	(50)	(74)	(25)	(32)	(12)	(35)	(18)

Continued overleaf

APPENDIX I. *Continued*

Locus allele	Atlantic samples					Gulf samples										
	MNC	BTH	GTN	CHS	HIH	SAR	RIV	APP	OSP	HOD	GIL	SAB	GVB	PCV	PAR	LLM
*sAAT-1**																
120	0·033	0·000	0·000	0·029	0·010	0·000	0·000	0·000	0·000	0·000	0·007	0·000	0·000	0·000	0·000	0·000
110	0·033	0·065	0·139	0·162	0·150	0·157	0·091	0·150	0·121	0·110	0·074	0·100	0·100	0·042	0·129	0·028
100	0·934	0·935	0·833	0·794	0·840	0·837	0·909	0·817	0·879	0·890	0·905	0·900	0·883	0·958	0·871	0·972
90	0·000	0·000	0·028	0·015	0·000	0·006	0·000	0·033	0·000	0·000	0·014	0·000	0·017	0·000	0·000	0·000
(*n*)	(15)	(23)	(18)	(34)	(50)	(83)	(22)	(30)	(70)	(50)	(74)	(25)	(30)	(12)	(35)	(18)
*EST-1**																
100	0·900	0·957	0·917	0·838	0·920	0·890	0·937	0·917	0·949	0·950	0·939	0·940	0·906	1·000	0·971	0·944
95	0·100	0·043	0·083	0·162	0·080	0·110	0·063	0·083	0·051	0·050	0·061	0·060	0·094	0·000	0·029	0·056
(*n*)	(15)	(23)	(18)	(34)	(50)	(86)	(24)	(30)	(78)	(50)	(74)	(25)	(32)	(12)	(35)	(18)
*GPI-B**																
− 110	0·000	0·000	0·000	0·000	0·000	0·006	0·000	0·000	0·000	0·011	0·000	0·000	0·000	0·000	0·000	0·000
− 100	0·967	0·978	1·000	0·971	0·960	0·959	0·958	1·000	0·949	0·967	0·986	0·940	0·937	1·000	0·943	0·944
− 50	0·033	0·022	0·000	0·029	0·040	0·035	0·042	0·000	0·051	0·022	0·014	0·060	0·063	0·000	0·057	0·056
(*n*)	(15)	(23)	(18)	(34)	(50)	(86)	(24)	(30)	(78)	(45)	(74)	(25)	(32)	(12)	(35)	(18)
*PEPB**																
115	0·033	0·000	0·000	0·000	0·010	0·017	0·042	0·000	0·013	0·011	0·000	0·000	0·000	0·042	0·014	0·028
100	0·967	1·000	1·000	1·000	0·980	0·971	0·958	1·000	0·981	0·989	1·000	1·000	1·000	0·958	0·986	0·972
85	0·000	0·000	0·000	0·000	0·010	0·012	0·000	0·000	0·006	0·000	0·000	0·000	0·000	0·000	0·000	0·000
(*n*)	(15)	(23)	(18)	(34)	(50)	(86)	(24)	(30)	(78)	(45)	(74)	(25)	(32)	(12)	(35)	(18)
*PEPD**																
115	0·000	0·022	0·000	0·015	0·010	0·006	0·000	0·000	0·026	0·011	0·014	0·000	0·000	0·000	0·014	0·028
100	0·967	0·956	1·000	0·956	0·980	0·971	0·958	0·967	0·948	0·922	0·952	1·000	0·937	0·917	0·943	0·916
85	0·033	0·022	0·000	0·029	0·010	0·023	0·042	0·033	0·013	0·067	0·034	0·000	0·063	0·083	0·043	0·056
75	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·000	0·013	0·000	0·000	0·000	0·000	0·000	0·000	0·000
(*n*)	(15)	(23)	(18)	(34)	(50)	(86)	(24)	(30)	(76)	(45)	(74)	(25)	(32)	(12)	(35)	(18)
*PEPS**																
105	0·000	0·043	0·000	0·029	0·030	0·041	0·021	0·017	0·072	0·011	0·027	0·020	0·047	0·083	0·014	0·083
100	1·000	0·957	1·000	0·971	0·970	0·959	0·958	0·983	0·928	0·989	0·966	0·980	0·953	0·917	0·986	0·917
95	0·000	0·000	0·000	0·000	0·000	0·000	0·021	0·000	0·000	0·000	0·007	0·000	0·000	0·000	0·000	0·000
(*n*)	(15)	(23)	(18)	(34)	(50)	(86)	(24)	(30)	(76)	(45)	(74)	(25)	(32)	(12)	(35)	(18)

APPENDIX II. Allele frequencies at nine polymorphic loci among sample localities from the 1987 year class, Gulf of Mexico and Atlantic Ocean

Locus allele	Atlantic samples		Gulf samples										
	BTH	CHS	SAR	RIV	APP	OSP	HOD	GIL	SAB	GVB	PCV	PAR	LLM
*ACP-2**													
125	0·000	0·000	0·021	0·000	0·000	0·000	—	0·000	0·028	0·000	0·000	0·000	0·000
115	0·000	0·048	0·104	0·033	0·079	0·061	—	0·138	0·139	0·083	0·059	0·143	0·083
100	1·000	0·952	0·875	0·967	0·921	0·939	—	0·862	0·833	0·917	0·941	0·857	0·917
(*n*)	(4)	(31)	(24)	(45)	(38)	(33)		(47)	(18)	(36)	(17)	(21)	(18)
*ADA**													
150	0·000	0·000	0·000	0·000	0·000	0·000	—	0·011	0·000	0·000	0·000	0·000	0·000
130	0·125	0·000	0·042	0·033	0·026	0·015	—	0·011	0·028	0·028	0·059	0·048	0·028
125	0·375	0·468	0·312	0·279	0·396	0·353	—	0·350	0·361	0·306	0·324	0·405	0·361
118	0·000	0·016	0·000	0·011	0·013	0·000	—	0·011	0·000	0·000	0·000	0·000	0·000
115	0·125	0·094	0·188	0·133	0·079	0·118	—	0·127	0·083	0·167	0·029	0·119	0·028
113	0·000	0·016	0·000	0·000	0·013	0·000	—	0·011	0·000	0·000	0·000	0·000	0·000
110	0·000	0·000	0·000	0·011	0·066	0·015	—	0·011	0·000	0·000	0·029	0·024	0·056
100	0·375	0·375	0·416	0·500	0·368	0·426	—	0·329	0·500	0·471	0·559	0·356	0·527
90	0·000	0·000	0·000	0·000	0·013	0·044	—	0·043	0·000	0·000	0·000	0·000	0·000
85	0·000	0·000	0·021	0·033	0·013	0·000	—	0·032	0·028	0·014	0·000	0·000	0·000
78	0·000	0·000	0·000	0·000	0·000	0·000	—	0·000	0·000	0·014	0·000	0·000	0·000
75	0·000	0·031	0·021	0·000	0·013	0·029	—	0·043	0·000	0·000	0·000	0·048	0·000
65	0·000	0·000	0·000	0·000	0·000	0·000	—	0·021	0·000	0·000	0·000	0·000	0·000
(*n*)	(4)	(32)	(24)	(45)	(38)	(34)		(47)	(18)	(36)	(17)	(21)	(18)
*ADH**													
− 100	0·500	0·516	0·479	0·600	0·500	0·633	—	0·553	0·639	0·403	0·529	0·595	0·527
− 75	0·250	0·436	0·479	0·378	0·461	0·338	—	0·447	0·333	0·555	0·471	0·381	0·417
− 50	0·125	0·016	0·021	0·022	0·026	0·029	—	0·000	0·028	0·028	0·000	0·024	0·028
− 20	0·125	0·032	0·021	0·000	0·013	0·000	—	0·000	0·000	0·014	0·000	0·000	0·028
(*n*)	(4)	(31)	(24)	(45)	(38)	(34)		(47)	(18)	(36)	(17)	(21)	(18)

Continued overleaf

APPENDIX II. *Continued*

Locus allele	Atlantic samples		Gulf samples										
	BTH	CHS	SAR	RIV	APP	OSP	HOD	GIL	SAB	GVB	PCV	PAR	LLM
*sAAT-1**													
120	0·125	0·016	0·000	0·000	0·000	0·000	—	0·000	0·028	0·000	0·000	0·000	0·028
110	0·000	0·113	0·104	0·122	0·132	0·100	—	0·106	0·111	0·056	0·206	0·071	0·028
100	0·875	0·855	0·875	0·878	0·855	0·900	—	0·894	0·833	0·944	0·794	0·929	0·944
90	0·000	0·016	0·021	0·000	0·013	0·000	—	0·000	0·028	0·000	0·000	0·000	0·000
(*n*)	(4)	(31)	(24)	(45)	(38)	(5)		(47)	(18)	(36)	(17)	(21)	(18)
*EST-1**													
100	0·875	0·875	0·958	0·933	0·878	0·941	—	0·883	0·917	0·931	0·971	1·000	0·833
95	0·125	0·125	0·042	0·067	0·122	0·059	—	0·117	0·083	0·069	0·029	0·000	0·167
(*n*)	(4)	(32)	(24)	(45)	(37)	(34)		(47)	(18)	(36)	(17)	(21)	(18)
*GPI-B**													
− 110	0·000	0·016	0·021	0·000	0·000	0·000	—	0·000	0·000	0·000	0·000	0·000	0·000
− 100	1·000	0·968	0·979	1·000	0·974	0·897	—	0·979	0·972	0·958	0·971	0·905	0·972
− 50	0·000	0·016	0·000	0·000	0·026	0·103	—	0·021	0·028	0·042	0·029	0·095	0·028
(*n*)	(4)	(32)	(24)	(45)	(38)	(34)		(47)	(18)	(36)	(17)	(21)	(18)
*PEPB**													
115	0·000	0·000	0·021	0·011	0·053	0·000	—	0·011	0·000	0·000	0·000	0·000	0·000
100	1·000	1·000	0·979	0·989	0·947	1·000	—	0·989	1·000	0·972	1·000	1·000	0·972
85	0·000	0·000	0·000	0·000	0·000	0·000	—	0·000	0·000	0·028	0·000	0·000	0·028
(*n*)	(4)	(32)	(24)	(45)	(38)	(34)		(47)	(18)	(36)	(17)	(21)	(18)
*PEPD**													
115	0·000	0·000	0·000	0·000	0·000	0·088	—	0·000	0·000	0·014	0·000	0·000	0·000
100	1·000	0·953	1·000	0·978	0·934	0·853	—	0·936	0·972	0·958	0·941	0·929	1·000
85	0·000	0·031	0·000	0·022	0·066	0·059	—	0·064	0·028	0·028	0·059	0·071	0·000
75	0·000	0·016	0·000	0·000	0·000	0·000	—	0·000	0·000	0·000	0·000	0·000	0·000
(*n*)	(4)	(32)	(24)	(45)	(38)	(34)		(47)	(18)	(36)	(17)	(21)	(18)
*PEPS**													
105	0·000	0·016	0·021	0·056	0·066	0·074	—	0·011	0·028	0·014	0·029	0·024	0·028
100	1·000	0·984	0·979	0·944	0·934	0·926	—	0·989	0·972	0·986	0·942	0·976	0·972
95	0·000	0·000	0·000	0·000	0·000	0·000	—	0·000	0·000	0·000	0·029	0·000	0·000
(*n*)	(4)	(32)	(24)	(45)	(38)	(34)		(47)	(18)	(36)	(17)	(21)	(18)

APPENDIX III. Allele frequencies at nine polymorphic loci in two year classes in the Gulf of Mexico and in the Gulf of Mexico and the Atlantic Ocean

Locus allele	Year class (Gulf)		Gulf	Atlantic
	1986	1987		
*ACP-2**				
125	0·000	0·003	0·001	0·000
115	0·100	0·089	0·091	0·063
100	0·900	0·908	0·908	0·937
(*n*)	(456)	(297)	(947)	(175)
*ADA**				
150	0·001	0·002	0·001	0·003
130	0·040	0·029	0·035	0·028
125	0·353	0·341	0·351	0·372
118	0·001	0·005	0·003	0·003
115	0·083	0·116	0·090	0·028
113	0·001	0·003	0·002	0·003
110	0·070	0·020	0·046	0·060
100	0·383	0·432	0·410	0·469
90	0·019	0·013	0·018	0·003
85	0·017	0·017	0·017	0·003
78	0·001	0·002	0·002	0·000
75	0·023	0·017	0·020	0·028
65	0·008	0·003	0·005	0·000
(*n*)	(464)	(298)	(956)	(176)
*ADH**				
−100	0·500	0·543	0·516	0·566
−75	0·459	0·430	0·444	0·391
−50	0·039	0·020	0·035	0·020
−20	0·002	0·007	0·005	0·023
(*n*)	(458)	(298)	(950)	(175)
*sAAT-1**				
120	0·001	0·004	0·002	0·017
110	0·111	0·104	0·105	0·120
100	0·881	0·886	0·886	0·854
90	0·007	0·006	0·007	0·009
(*n*)	(449)	(269)	(912)	(175)
*EST-1**				
100	0·933	0·921	0·928	0·898
95	0·067	0·079	0·072	0·102
(*n*)	(464)	(297)	(955)	(176)
*GPI-B**				
−110	0·002	0·002	0·002	0·003
−100	0·962	0·963	0·960	0·971
−50	0·036	0·035	0·038	0·026
(*n*)	(459)	(298)	(951)	(176)
*PEPB**				
115	0·012	0·012	0·011	0·006
100	0·985	0·983	0·982	0·991
85	0·003	0·005	0·007	0·003
(*n*)	(459)	(298)	(951)	(176)

Continued overleaf

APPENDIX III. *Continued*

Locus allele	Year class (Gulf)		Gulf	Atlantic
	1986	1987		
*PEPD**				
115	0·011	0·012	0·011	0·009
100	0·952	0·946	0·945	0·968
85	0·035	0·042	0·043	0·020
75	0·002	0·000	0·001	0·003
(*n*)	(457)	(298)	(949)	(176)
*PEPS**				
105	0·038	0·037	0·036	0·023
100	0·960	0·961	0·961	0·977
95	0·002	0·002	0·003	0·000
(*n*)	(457)	(298)	(949)	(176)