Spatial and temporal distribution of grouper larvae (Serranidae: Epinephelinae: Epinephelini) in the Gulf of Mexico and Straits of Florida

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Abstract—Little is known about the seasonality and distribution of grouper larvae (Serranidae: Epinephelini) in the Gulf of Mexico and Atlantic Ocean off the coast of the southeast United States. Grouper larvae were collected from a transect across the Straits of Florida in 2003 and 2004 and during the Southeast Area Monitoring and Assessment Program spring and fall surveys from 1982 through 2005. Analysis of these larval data provided information on location and timing of spawning, larval distribution patterns, and interannual occurrence for a group of species not easily studied as adults. Our analyses indicated that shelf-edge habitat is important for spawning of many species of grouper—some species for which data were not previously available. Spawning for some species may occur year-round, but two peak seasons are evident: late winter and late summer through early fall. Interannual variability in the use of three important subregions by species or groups of species was partially explained by environmental factors (surface temperature, surface salinity, and water depth). A shift in species dominance over the last three decades from spring-spawned species (most of the commercial species) to fall-spawned species also was documented. The results of these analyses expand our understanding of the basic distribution and spawning patterns of northwest Atlantic grouper species and indicate a need for further examination of the changing population structure of individual species and species dominance in the region.

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Adult grouper (Serranidae: Epinephelini) are commercially and recreationally important species that are highly susceptible to overfishing (Coleman et al., 1996), largely due to their spawning behavior and slow growth (Manooch, 1987; Shapiro, 1987; Coleman et al., 1996). Many species of groupers aggregate at consistent locations and time of year for spawning (Nemeth et al., 2007; Starr et al., 2007), and these aggregations are often targeted by fishermen (Burton et al., 2005). Fishing pressure on adult grouper and changes to habitat at all life-history stages of grouper have made evident the need for more effective fisheries management strategies. Most research on Gulf of Mexico grouper focuses on single species (e.g., Brule et al., 1999, 2003), over a very limited area (e.g., single spawning aggregations: Nemeth, 2005; off the coast of a single state or county: Coleman et al., 1996), or over short temporal durations (e.g., Eggleston, 1995).

Plankton surveys provide a reliable source of fishery-independent data for fishery management purposes and grouper larvae are routinely collected in these surveys (Houde, 1982; Marancik et al., 2005; Hernandez et al., 2010). Ichthyoplankton surveys provide data on seasonal (Hernandez et al., 2010), spatial (Ditty et al., 2004), and environmental characteristics associated with spawning (Richardson et al., 2009), all of which are particularly useful for species that are rare, elusive, or endangered as adults. For example, data on abundance and habitat use for early life stages have been directly integrated into fisheries management of bluefin tuna (*Thunnus thynnus*) through stock assessment calculations (Scott et al., 1993). Spatial and temporal distribution and frequency of collection of larvae reflect changes in the juvenile and adult population structure (Richardson et al., 2010), which, coupled with climate models, may provide a means of forecasting the abundance and distribution of future populations (Hare et al., 2010).

Recent examination of larval grouper morphological characters from the most comprehensive collections available in the U.S. southeast region resulted in more precise taxon identi-

fication than had been previously attainable (Marancik et al., 2010). With these newly identified larvae, our purpose here is to describe the spatial and seasonal distribution patterns of 15 species (whose larvae could be identified) and four multispecies groups (whose larvae could not be identified to species but which share similar physical attributes). Specifically, our objectives were to describe 1) the spawning season; 2) locations of spawning; 3) environmental factors associated with larval grouper habitat; and 4) decadal-scale variability in the distribution and habitat use of grouper larvae. Our work is focused on grouper larvae from the Straits of Florida and the northern Gulf of Mexico.

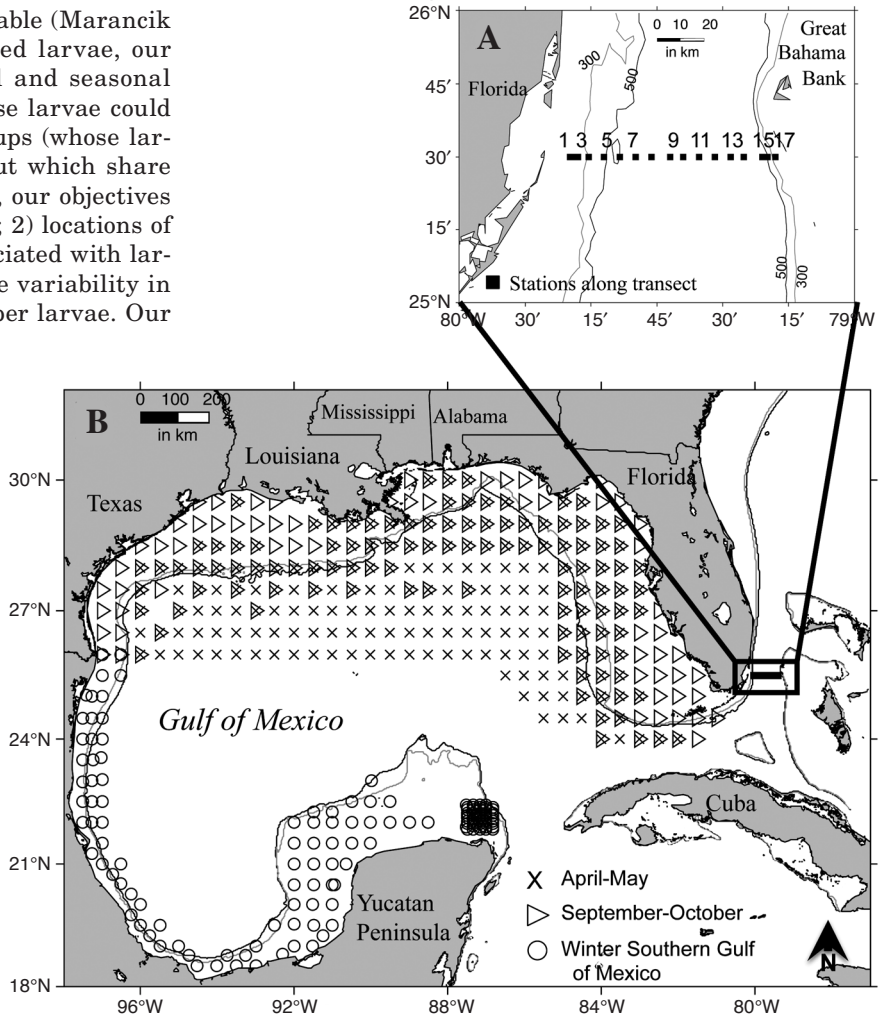

Figure 1

Map of sampling regions showing (**A**) an expanded view of the transect stations across the Straits of Florida, and (**B**) the northern Gulf of Mexico Southeast Area Monitoring and Assessment Program (SEAMAP) sampling stations and east Florida shelf transect. SEAMAP stations are coded by season: X=April–May, triangles=September–October, and circles=winter southern Gulf of Mexico sampling. The 100-m and 200-m isobaths are also shown.

Materials and methods

Collections

Samples were collected as part of two separate sampling programs: one across the Straits of Florida and one in the northern Gulf of Mexico.

Straits of Florida A 17-station transect crossing the Straits of Florida at 25.5°N was sampled as part of a larval billfish (Istiophoridae and Xiphiidae) project conducted by researchers at the University of Miami, Rosenstiel School of Marine and Atmospheric Science. Samples were collected at the beginning of each month from January 2003 through December 2004 (Fig. 1A). The transect extended from the Florida shelf break to the Great Bahama Bank. The three easternmost and three westernmost stations were approximately 2 km apart; the remaining stations were approximately 5.5 km apart. Samples were collected with an asymmetrical MOCNESS (multiple opening-closing net and environmental sensing system) consisting of a 4-m^2 frame with 1000-µm mesh nets and a 1-m^2 frame with 150-µm mesh nets (Guigand et al., 2005). The MOCNESS sampled in 25-m depth bins from 0–50 m at the westernmost station or 0–100 m at deeper stations. Surface waters (0–0.5 m) were sampled with a combined neuston net composed of a 1×2 m mouth with 1000-µm mesh net and a 1×0.5-m mouth with 150-µm mesh net. Samples were collected between sunrise and sunset, and the entire transect was generally sampled within a 48-hour period. At least 16 of the 17 stations were successfully sampled on all but three cruises; weather limited sampling in December 2003, January 2004, and November 2004. Samples were immediately preserved in 95% ethanol and, after 2–5 days, were transferred to 70% ethanol for long-term storage. Llopiz and Cowen (2008) and Richardson et al. (2010) provide further details of the Straits of Florida sampling survey.

In the laboratory, all larval fish were removed from all neuston samples, samples collected in 2003 with both the 1-m^2 and 4-m^2 MOCNESS, and samples collected in 2004 from only the 4-m^2 MOCNESS. Genetic sequencing of the cytochrome oxidase subunit I gene (as in Richardson et al., 2007) was used to identify a subset (approximately 40%) of the Straits of Florida grouper larvae to species (Marancik et al., 2010). The remaining larvae were either identified to species or grouped with morphologically similar species according to physical attributes (Marancik et al., 2010). Body length and developmental stage were recorded for each fish. Developmental stage refers to the upward (dorsal) flexion of the notochord tip (urostyle) concurrent with caudal

fin base and principal ray formation (Moser, 1996): the preflexion stage occurs when the notochord is straight; the flexion stage occurs when the notochord is obviously flexed and caudal rays are forming; and the postflexion stage occurs when the notochord tip is aligned vertically with the caudal base plate (hypural) elements. Owing to a single grouper larva collected in neuston samples (1 individual in 383 neuston stations), only MOCNESS samples were used in analyses.

Gulf of Mexico Grouper larvae were collected from the Southeast Area Monitoring and Assessment Program (SEAMAP) resource surveys conducted in the United States territorial waters of the Gulf of Mexico by the National Marine Fisheries Service (NMFS) Southeast Fisheries Science Center. All SEAMAP plankton samples included in our analyses were collected from 1982 through 2005 with either a bongo net consisting of a 61-cm frame and 335-µm mesh nets towed obliquely from 2–5 m off the bottom or to a maximum depth of 200 m, or with a neuston net with 1×2 m frame and a 950-µm mesh net towed at the surface. Samples were collected throughout the day and night depending on when the ship reached each station. Environmental data consistently collected over the entire SEAMAP time series were surface temperature, surface salinity, and water depth and therefore these were the only environmental variables considered in analyses. Plankton samples were initially fixed in either 5–10% unbuffered formalin (the majority of samples) or 95% ethanol. Formalin-fixed samples were transferred to 95% ethanol after 48 hours, and samples initially fixed in ethanol were transferred to fresh 95% ethanol after 24 to 36 hours. All fish larvae were removed from samples, identified to the lowest taxonomic level possible, and measured at the Sea Fisheries Institute, Plankton Sorting and Identification Center in Szczecin, Poland. Grouper larvae were further identified on the basis of morphological characters (Marancik et al., 2010).

Plankton collections were made in all months of the year during the 23 years of SEAMAP surveys included in our analyses. The greatest effort was conducted in May (2419 neuston and 1529 bongo samples) and September (2167 neuston and 1904 bongo samples); the least effort occurred in February (40 neuston and 41 bongo samples) and March (50 neuston and 178 bongo samples; Table 1). The most complete sampling coverage of the continental shelf of the northern Gulf of Mexico began in 1986 and continues to the present. Unfortunately, the months of November through March, likely the peak spawning season for many grouper species (Hood and Schlieder, 1992; Coleman et al., 1996; Nemeth et al., 2007), were rarely and inconsistently sampled during SEAMAP. Grouper larvae have been re-examined and identified from collections through 2005; therefore only data from SEAMAP surveys from 1986 through 2005 were statistically analyzed.

The most temporally and spatially consistent sampling effort was conducted during two dedicated SEAMAP plankton surveys: the spring and fall surveys (Fig. 1B). Within these two annual surveys, sampling coverage was fairly consistent from 1986 through 2005. The percentage of stations sampled gulf-wide, roughly representing the area covered, ranged from 28.7% to 54.0% (mean=45.1%) in the spring and from 26.1% to 76.7% (mean=61.3%) in the fall, and the targeted survey area was usually represented over its entire north–south and east–west extent (Table 2; Lyczkowski-Shultz and Hanisko, 2007; Muhling et al., 2010). The most consistent sampling occurred in April–May and September–October except for three years during which sampling began late (spring 2003, spring 2004, fall 2005) and one year which finished early (fall 1997).

The spring and fall surveys targeted different bathymetric zones with overlap at the shelf edge. During the spring plankton survey, conducted in April and May (1982–present), stations were sampled from the shelf edge to the United States Exclusive Economic Zone (EEZ) within a 0.5°×0.5° (56-km) grid. The second dedicated plankton survey, called the "fall plankton survey," was conducted from late-August through October (1986–present) from the coast to the continental shelf edge (10–200 m water depth) and from south Texas to south Florida. Additional specimens and data came from plankton sampling conducted by the National Marine Fisheries Service (NMFS) Southeast Fisheries Science Center during SEAMAP summer and fall trawl surveys, winter plankton surveys, squid-butterfish surveys, Alabama summer and fall plankton surveys, and the fall pelagic fish survey in the Gulf of Mexico (Table 3; see Lyczkowski-Shultz and Hanisko, 2007, for details).

Analyses

Seasonal and spatial occurrence The spatial consistency and monthly frequency of sampling in the Straits of Florida makes these data the best suited for determining the seasonality of larval grouper occurrence and, in turn, presumed seasonality of spawning. Only specimens identified to species were used in analyses. We used quotient analysis to define potential and peak season of occurrence and cross-transect distribution, using the Straits of Florida data. With this analysis, the ratio of the proportion of larval occurrence to the proportion of observations was determined within environmental (spatial or temporal) bins in order to discover when or where larvae were collected with higher (or lower) frequency than would be expected if larvae were evenly distributed. Quotient values >1 indicate a relatively higher occurrence of larvae (based on the number of observations) than expected, whereas values <1 indicate lower than expected occurrence (van der Lingen et al., 2001). Significance of the quotient values (above or below the null of 1) was determined by a bootstrapping technique similar to that used in Bernal et al. (2007). Quotient analysis is relatively robust for data sets containing many zero values, allowing analysis of the complete data set and cross-transect relationships despite the rarity of grouper larvae in collections. Analyses were conducted

Table 1

Percent frequency of occurrence (%FO) of total larvae and number of total larvae and preflexion larvae collected, size range, and monthly occurrence of specimens of each grouper species or species group collected during Southeast Area Monitoring and Assessment Program (SEAMAP) surveys from 1982 through 2005 (Marancik et al., 2010). The "◯" symbol denotes the total seasonal occurrence of any larvae; the "+" symbol denotes presence of preflexion larvae collected. Species complexes are listed as subdivisions of the species-groups (small spinelets+standard pigment, and long curved spinelets+standard pigment) were included with the species group in analyses (e.g., 50 individuals were included in the long curved spinelets+standard pigment group).

	%FO		*n*		Size	Month collected											
Species or group	Bongo	Neuston	Total	Preflexion	range (mm)	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
Cephalopholis cruentata	0.29	0.09	44	27	2.2–6.4				+	+	◯	◯	+	+	+		
Cephalopholis fulva	0.01	0.02	3	1	4.2–4.8				◯	◯				+			
Gonioplectrus hispanus	0.03	0.03	5		2.9–10.6								◯	◯			
Hyporthodus mystacinus	0.01	0.01	2		5.3–6.0					◯					◯		
Paranthias furcifer	0.06	0.01	12		4.7–10.9				◯	◯				◯			
Epinephelus striatus/ Mycteroperca venenosa	0.01	0.02	3	3	2.3–2.6									◯	◯		
Small epinephelini + standard pigment	1.02	0.04	162	162	1.2–4.8	+	+		+	+	+	+		+	+	+	
Small spinelets + standard pigment	0.9	0.41	199	121	2.2–6.5		+		+	+	+			+	+	+	
Epinephelus adscensionis/ striatus	0.01	0.01	2		6.6–8.0					◯							
E. morio/drummondhayi/ guttatus	0.01	0.01	2		5.5–6.5					◯							
A. afer/E. adscensionis/ D. inermis	0.06	0.00	5		6.2–8.1	◯			◯	◯							
Long curved spinelets + standard pigment	0.24	0.05	26	4	3.0–5.6				+	+				◯	◯		
E. itajara/Hyporthodus flavolimbatus/ niveatus/nigritus	0.15	0.01	24		4.9–24.3					◯			◯	◯	◯	◯	
Mycteroperca spp.																	
Small: Dorsal–ventral tail pigment	0.13	0.02	12	11	2.0–4.7				+	+	+			+		+	
Medium: *E. itajara/Mycteroperca*	0.11	0.07	15	4	2.7–10.1		◯		+	+	◯			◯	+	+	
Large: *Mycteroperca* spp.	0.14	0.05	19		5.5–19.5	◯	◯		◯	◯	◯						
Long straight spinlelets	0.04	0.00	3	1	3.3–4.4		+			◯				◯			
Specimens with broken spines	0.04	0.02	6	4	2.6–5.0					+				+			
Total number of larvae collected in bongos (neuston)			544			5(0)	6(2)	0(0)	20(13)	160(44)	6(1)	3(0)	5(0)	147(31)	71(19)	10(1)	0(0)
% FO of larvae collected in bongo (neuston)	3.29	0.89				3.8(0)	12.2(5.0)	0(0)	2.8(1.2)	5.2(1.4)	0.8(0.1)	0.4(0)	1.4(0)	4.4(0.9)	5.3(1.5)	1.5(0.2)	0(0)
Total number of plankton samples collected: bongo			7848			133	41	178	639	1529	767	671	360	1904	830	547	249
Neuston			9102			162	40	50	978	2419	835	635	339	2167	795	482	200

Table 2

Mean percent sampling coverage (% Coverage; number of grid cells sampled/number of grid cells in subregion×100) and range of latitudes and longitudes sampled in each Gulf of Mexico subregion (a–e, see Fig. 2) during the spring or fall Southeast Area Monitoring and Assessment Program (SEAMAP) plankton surveys over the time intervals 1986–95 and 1996–2005.

	a: Texas-Mexico shelf. to 90 deg. longitude			b: Texas-Louisiana shelf			c: Mississippi–Alabama–north Florida shelf			d: west Florida shelf			e: offshore southwest Florida to 90 deg. Longitude		
Region	% Coverage	Range latitude	Range longitude	% Coverage	Range latitude	Range longitude	% Coverage	Range latitude	Range longitude	% Coverage	Range latitude	Range longitude	% Coverage	Range latitude	Range longitude
Spring															
1986–1995	42.4	26.0–27.4	94.7–90.6	37.7	28.0–28.3	94.0–90.5	47.8	28.0–29.8	89.9–85.0	27.8	24.4–27.2	84.5–83.6	53.2	24.5–27.4	90.0–85.0
1996–2005	54.8	26.0–27.5	95.9–90.7	50.0	28.0	95.9–90.5	47.1	28.0–29.9	90.0–85.1	22.0	24.1–26.0	84.5–83.6	58.7	24.5–27.5	90.0–85.0
Fall															
1986–1995	44.5	26.0–27.5	97.0–95.9	96.4	28.0–29.5	96.5–90.5	59.8	28.0–30.5	90.0–85.0	58.7	25.3–29.8	84.5–81.8	1.4	27.5	85.0
1996–2005	66.5	26.0–27.5	97.0–92.8	86.0	28.0–29.5	96.5–90.5	58.0	28.1–30.4	90.0–85.1	45.7	25.7–29.6	84.5–82.2	5.7	26.0–27.5	85.0

by using functions written for MATLAB (for Mac, vers. R2010a; The MathWorks Inc., Natick, MA).

Before formal analyses of larval grouper distributions, steps were taken to control for inconsistencies in sampling effort over the long time-scale of SEAMAP sampling. Each station sampled during SEAMAP was assigned to a cell within a 0.5°×0.5° resolution grid encompassing the northern Gulf of Mexico (23–30°N latitude, and 81–98°W longitude; Fig. 1B). If more than one station was sampled within a grid cell during a single month of any year, the mean value of each environmental variable was taken. This procedure provided a sampling regime that was consistent over time and facilitated comparisons between environmental and larval fish data. Owing to the uneven spatial sampling effort among seasons and the low total abundance of grouper in Gulf of Mexico samples, 1) larvae were standardized to presence or absence within each grid cell for each month of each year sampled, 2) no size-specific analyses were conducted, 3) larvae collected from bongo and neuston samples were combined, and 4) statistical analyses were limited to samples collected during spring (April–May) and fall (September–October) from 1986 through 2005.

Influence of environmental factors and change over time Interannual variability in Gulf of Mexico regional larval grouper habitat use was examined by using generalized additive models (GAMs), a regression technique used to fit nonlinear relationships. Seasonal mean surface temperature, mean surface salinity, mean water depth, and year for subregions of the Gulf of Mexico were modeled to predict interannual variability in percent frequency of occurrence (%FO; Hastie and Tibshirani, 1990; Wood, 2006). The northern Gulf of Mexico (north of 23°N) was divided into subregions (labeled a–e in Fig. 2) that reflected the presence of grouper larvae and orientation of the coastline in relation to bathymetry. Within each subregion, %FO was calculated as the number of grid cells in which any grouper were present divided by the number of grid cells sampled during spring (April–May; 1986–2005) or fall (September–October; 1986–2005) surveys. GAMs are most effective for data sets with few zeros (years sampled, but no grouper collected); therefore GAMs were generated only for subregions and seasons (i.e., spring or fall) during which grouper were collected in at least 60% of the years being analyzed. Models of data collected during spring surveys were limited to depths <900 m to reduce the number of grid cells included in analyses owing to the near absence of grouper larvae at depths >900 m. With these restrictions, only 3 of the 5 subregions (Fig. 2, subregions b–d) contained enough data on which to base a model. Data from both bongo and neuston net samples were combined in order to include as many larval grouper data as possible. GAMs generated for bongo data provided similar, but weaker, results; therefore the combined data are presented. The full model used to explain %FO within subregion (r) and season (s) was the following:

Table 3

Gulf of Mexico plankton sampling data from 1982 through 2005 by month and Southeast Area Monitoring and Assessment Program (SEAMAP) survey type showing the number and occurrence (number of stations with grouper present, "Occ. grouper") of grouper larvae in neuston, bongo, and nonstandard sampling gear, and ranges in latitude and longitude of the sampling surveys. WP=winter plankton surveys, **=sampling outside of established SEAMAP surveys, SP=spring plankton surveys, SQ=squid-butterfish surveys, AS=Alabama summer plankton surveys, SG=summer groundfish trawl surveys, FP=fall plankton surveys, AF=Alabama fall plankton surveys, FG=fall groundfish trawl surveys, and FS=fall pelagic fish surveys.

		Neuston			Bongo			Non-standard gear		Latitude		Longitude	
Month	Survey type	No. grouper	Occ. grouper	No. samples	No. grouper	Occ. grouper	No. samples	No. grouper	No. samples	South	North	West	East
January	WP	0	0	162	5	5	133	0	5	18.50	30.00	–97.50	–87.00
February	**	na	0	0	0	0	5	na	0	29.92	30.10	–88.68	–88.40
February	WP	2	2	40	6	5	36	0	3	22.00	29.51	–95.88	–87.83
March	**	0	0	13	0	0	130	na	0	28.18	30.20	–91.50	–86.50
March	SP	0	0	37	0	0	48	na	0	28.46	29.19	–91.50	–89.48
April	**	0	0	5	na	0	0	na	0	29.00	29.01	–88.83	–88.83
April	SP	13	12	973	20	18	639	na	0	23.15	30.01	–96.00	–83.20
May	**	3	3	84	na	0	0	na	0	28.97	29.09	–88.85	–88.14
May	SP	40	30	2321	160	80	1513	0	42	23.50	30.01	–96.04	–82.00
May	SQ	1	1	14	0	0	16	na	0	28.55	29.53	–90.08	–87.93
June	**	0	0	72	0	0	50	na	0	25.00	30.00	–89.50	–83.00
June	AS	0	0	4	0	0	2	na	0	30.00	30.23	–88.31	–87.50
June	SG	0	0	510	2	2	568	na	0	26.00	30.22	–97.20	–86.00
June	SP	1	1	249	4	4	147	na	0	24.50	30.00	–96.00	–83.50
July	**	0	0	21	0	0	22	na	0	25.00	28.82	–88.48	–82.00
July	SG	0	0	611	3	3	646	na	0	26.00	30.05	–97.04	–87.00
July	SQ	0	0	3	0	0	3	na	0	28.00	28.03	–84.88	–84.21
August	**	0	0	13	0	0	13	na	0	27.53	29.16	–91.27	–87.63
August	FP	0	0	258	4	4	278	na	0	24.00	30.25	–97.25	–82.00
August	SQ	0	0	68	1	1	69	na	0	26.49	30.07	–96.81	–85.03
September	AF	0	0	134	0	0	6	na	0	30.00	30.32	–88.17	–87.97
September	FP	31	19	2033	147	84	1898	na	0	24.00	31.50	–97.25	–79.67
October	**	0	0	38	2	2	41	na	0	28.48	29.82	–90.73	–84.99
October	AF	0	0	18	na	0	0	na	0	30.12	30.28	–88.13	–87.98
October	FG	3	3	454	20	15	488	na	0	25.99	30.18	–97.22	–85.01
October	FP	16	9	276	44	23	290	na	0	24.00	30.01	–91.50	–81.49
October	FS	0	0	9	5	4	11	na	0	27.06	29.12	–96.06	–86.09
November	**	0	0	32	1	1	32	na	0	26.49	30.08	–86.08	–82.49
November	FG	1	1	436	7	5	504	na	0	26.02	30.17	–96.98	–84.45
November	FS	0	0	11	2	2	11	na	0	25.03	29.16	–89.02	–84.01
November	WP	0	0	3	0	0	3	na	0	29.00	29.00	–91.50	–90.50
December	FG	0	0	4	0	0	7	na	0	28.50	29.00	–91.50	–90.50
December	WP	0	0	196	0	0	242	0	9	25.00	30.00	–96.00	–85.00

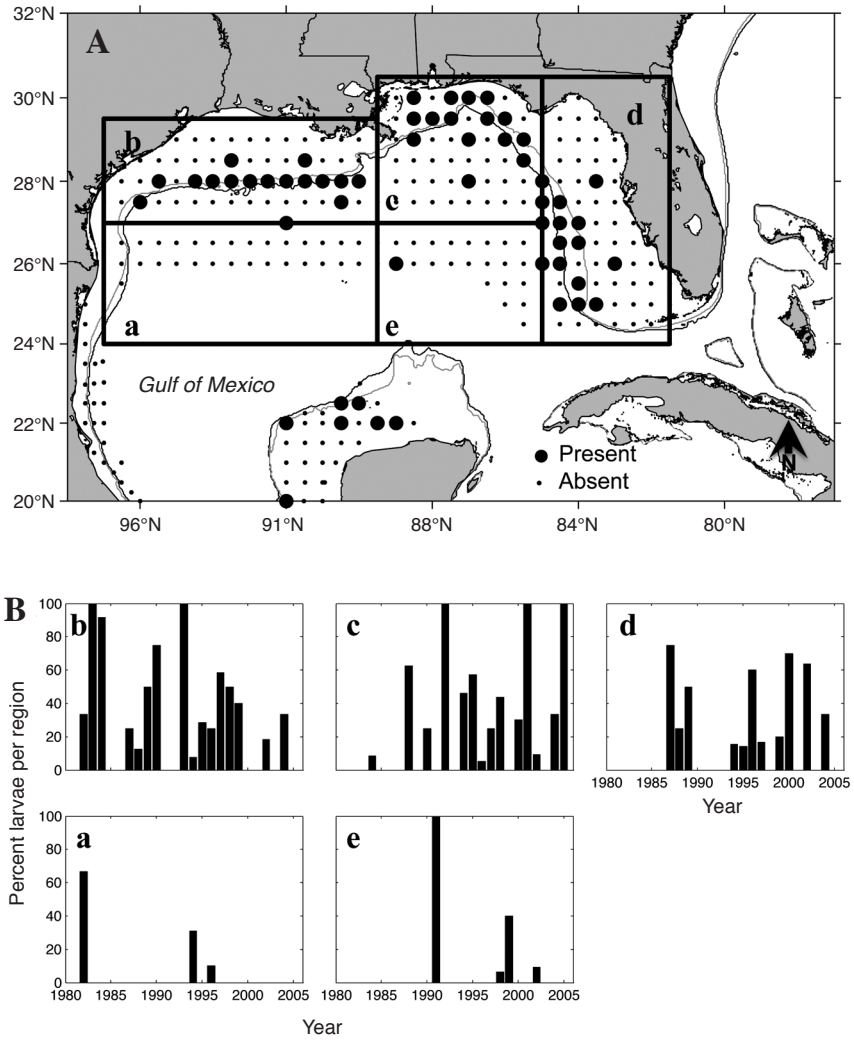

Figure 2

Spatial distribution of the most recently spawned (least developed) grouper larvae with standard pigment collected during Southeast Area Monitoring and Assessment Program plankton surveys, 1982–2005. (**A**) The five subregions (a–e) in the northern Gulf of Mexico based on the presence of small larvae and orientation of the coastline in relation to bathymetry. The southern Gulf of Mexico was sampled only during one year and was not included in analyses. (**B**) Bar graph of the percentage of recently spawned grouper larvae collected in each region by year. Lowercase letters in each subpanel correspond to the subregion letters in panel A: a) Mexico–Texas shelf to 90°W longitude, b) Texas, Louisiana shelf, c) Mississippi–Alabama–north Florida shelf, d) west Florida shelf, and e) open water east of 90°W longitude.

$$y_{r,s} = a_{r,s} + g_1(year_{r,s}) + g_2(stemp_{r,s}) + g_3(ssal_{r,s}) + g_4(wdep_{r,s}) + e_{r,s},$$

where a = the subregion by season intercept;
g = the nonparametric smoothing function for each term;
stemp, *ssal*, and *wdep* = the mean surface temperature, mean surface salinity, and mean water depth for each subregion by season for each year, respectively; and
e = a normally distributed random error term with a mean of zero and finite variance.

The model was run with all combinations of covariates to find the best subset of covariates (best fit) required to explain %FO for each subregion by season. Two evaluation techniques were used to select the best model. The generalized cross-validation (GCV) score is a measure of the predictive squared error of the model (Wood, 2006). Akaike's information criterion with a

low-sample-size bias-correction term (AICc; Burnham and Anderson, 2002) evaluates the trade-off between the number of covariates in a model and the likelihood of the model accurately predicting new data (Akaike, 1973)—therefore reducing the chances of a model with redundant covariates appearing to better explain the data. The best model was indicated by the lowest value of each evaluation score, and in all cases, both techniques yielded the same results (data not shown). AIC_c scores were also used to calculate a relative likelihood of each model being the best model (Burnham and Anderson, 2002). GAMs were created with the MGCV library (vers. 1.6-1) in R software (for Mac, vers. 2.11.0; R Development Core Team, 2008).

Results

Grouper larvae were generally collected in low numbers during both the fine-scale sampling in the Straits of Florida and the broad-scale sampling in the Gulf of Mexico. A total of 665 individuals (521 individuals identified to species) were collected in 384 stations (both MOCNESS frame sizes and all depths combined) from the Straits of Florida. A total of 544 individuals were collected in 16,950 samples from the Gulf of Mexico (433 individuals in 7848 bongo samples; 111 individuals in 9102 neuston net samples).

Seasonal occurrence

Grouper larvae were collected during all months of sampling in the Straits of Florida (Table 4) and in all months except March and December in the Gulf of Mexico (Table 1). Most Straits of Florida larvae, specifically preflexion larvae whose presence indicate recent spawning, occurred during February through May. A second, less diverse and less numerous group of larvae was present from July through October (Table 4). The high apparent abundance of larvae in February was due to a single collection of >150 specimens of preflexion *Epinephelus guttatus* (red hind) at one station. The lowest occurrence and species richness (number of species captured) of larvae were observed during the months of January, July, August, and December.

Spatial occurrence

Straits of Florida Larvae were distributed in two distinct assemblages across the Straits of Florida: an eastern assemblage and a western assemblage (Fig. 3). Five species occurred significantly more frequently within the eastern 10 km of the transect (Fig. 3, A–E). For most of these species, the pattern was the same for all developmental stages. *Cephalopholis cruentatus* (graysby), one of the more abundant species, was collected across the transect across the straits, but was collected most frequently on the eastern side at the preflexion stage, whereas flexion and postflexion stage larvae were collected across the transect, occurring at no stations significantly more or less frequently (Fig. 3, F–G). Four species were collected significantly more frequently on the western side of the transect (Fig. 3, H–K). The remaining six species were not collected at high enough frequencies to analyze statistically (<5 specimens) or were collected evenly across the transect (*Paranthias furcifer*, Fig. 3L).

Gulf of Mexico Distribution patterns of grouper larvae of all sizes were categorized into five subregions of occurrence in the northern Gulf of Mexico (Fig. 4). Small (1.3–4.3 mm NL) preflexion larvae without prominent dorsal and pelvic spines are indicative of recent spawning and were collected in 18 of 23 years of SEAMAP surveys. Repeated occurrence of these earliest stage larvae gave evidence of three of the subregions as areas of spawning activity: the Texas–Louisiana Shelf west of the Mississippi River (TX–LA; north of 27.5°N and west of 90°W; Fig. 2 subregion b), the north-central Gulf shelf off the coasts of Mississippi, Alabama, and northern Florida (MS–AL–nFL; north of 27.5°N and between 90° and 85°W; Fig. 2 subregion c), and the west Florida shelf (wFL; north of 23° N and between 85–81°W; Fig. 2 subregion d). These three subregions accounted for the vast majority of grouper larvae collected (n=314) and were the only subregions containing early-stage larvae. Later-stage larvae were also collected farther offshore in two additional subregions (Fig. 4, A–C and F).

Sampling was conducted off the coast of the Yucatan Peninsula in the southern Gulf of Mexico during January and February 1990. Although grouper larvae were collected (n=14), the data were not included in our analyses because of the timing (winter) and infrequency (a single year) of sampling in the area.

Graysby were the most abundant group of larvae collected in the Gulf of Mexico that could be identified to species. Graysby larvae occurred during both spring and fall surveys. Most specimens were collected during July–October and were distributed primarily along the west Florida shelf (n=35). A few larvae were collected on or near the Louisiana shelf and Mississippi–Alabama–north Florida shelf during the fall survey (n=2) and in deep offshore waters off southwestern Florida during the spring survey (n=5; Fig. 4A).

Small *Mycteroperca* spp. larvae (i.e., specimens with dorsal-ventral tail pigment; Table 1) were collected during April–June and September and November in all three presumptive spawning subregions of the northern Gulf identified in this study (Fig. 4D; spring: all three, fall: TX–LA, MS–AL–nFL). These specimens were primarily collected along the shelf break. A similar spatial and temporal distribution pattern was observed for several slightly larger larvae identified as either *E. itajara* or *Mycteroperca* spp. based on the presence of pigment on the cleithral symphysis, standard tail pigment, and broad-based, long and curved spinelets (Marancik et al., 2010). These specimens also were collected during April–June and September–November along the shelf break of all three presumed spawning subregions (spring: all three, fall: TX–LA and wFL; Fig. 4E, Table

Table 4

Percent frequency of occurrence (%FO) of total larvae, total number of larvae and preflexion larvae collected, assemblage location (assigned based on quotient analysis), and monthly occurrence of larvae of each species of grouper collected along the Straits of Florida transect (see Fig. 1A). The order in which the species are listed reflects month of first capture. The "○" symbol indicates months with only later stage larvae collected. The "+" symbol indicates when preflexion larvae were collected. The "△" symbol indicates when preflexion larvae were collected significantly more frequently than would be expected if larvae were evenly dispersed (based on quotient analysis).

			No. of larvae			Month collected											
Species	Common name	%FO	Total	Preflexion	Assemblage	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
Mycteroperca bonaci	Black grouper	1.82	8	2	West	△			○	○	○					○	△
Cephalopholis fulva	Coney	1.56	27	13	East		△	△	○			○					
Epinephelus guttatus	Red hind	1.30	199	183	East		△	△									
Epinephelus striatus	Nassau grouper	1.04	11	6	East		△	○									
Mycteroperca venenosa	Yellowfin grouper	1.30	20	6	East		+	△	+								
Paranthias furcifer	Atlantic creolefish	1.56	13	8			○	△	○								
Mycteroperca phenax	Scamp	1.04	4	2				△	△								
Epinephelus morio	Red grouper	1.82	9	2	West			○		△							
Hyporthodus nigritus	Warsaw grouper	0.26	1						○								
Hyporthodus niveatus	Snowy grouper	1.82	7	6	West					+				△	△		
Epinephelus drummondhayi	Speckled hind	0.52	2							○					○		
Cephalopholis cruentata	Graysby	9.64	141	17	East						+	△	△	○	○	+	
Alphestes afer	Mutton hamlet	0.26	1								○						
Hyporthodus flavolimbatus	Yellowedge grouper	4.95	73	6	West								○	○	△	○	
Hyporthodus mystacinus	Misty grouper	0.78	4											○	○		
Gonioplectrus hispanus	Spanish flag	0.26	1												○		
	Number of species		16	11		1	5	7	6	4	3	2	2	4	6	3	1

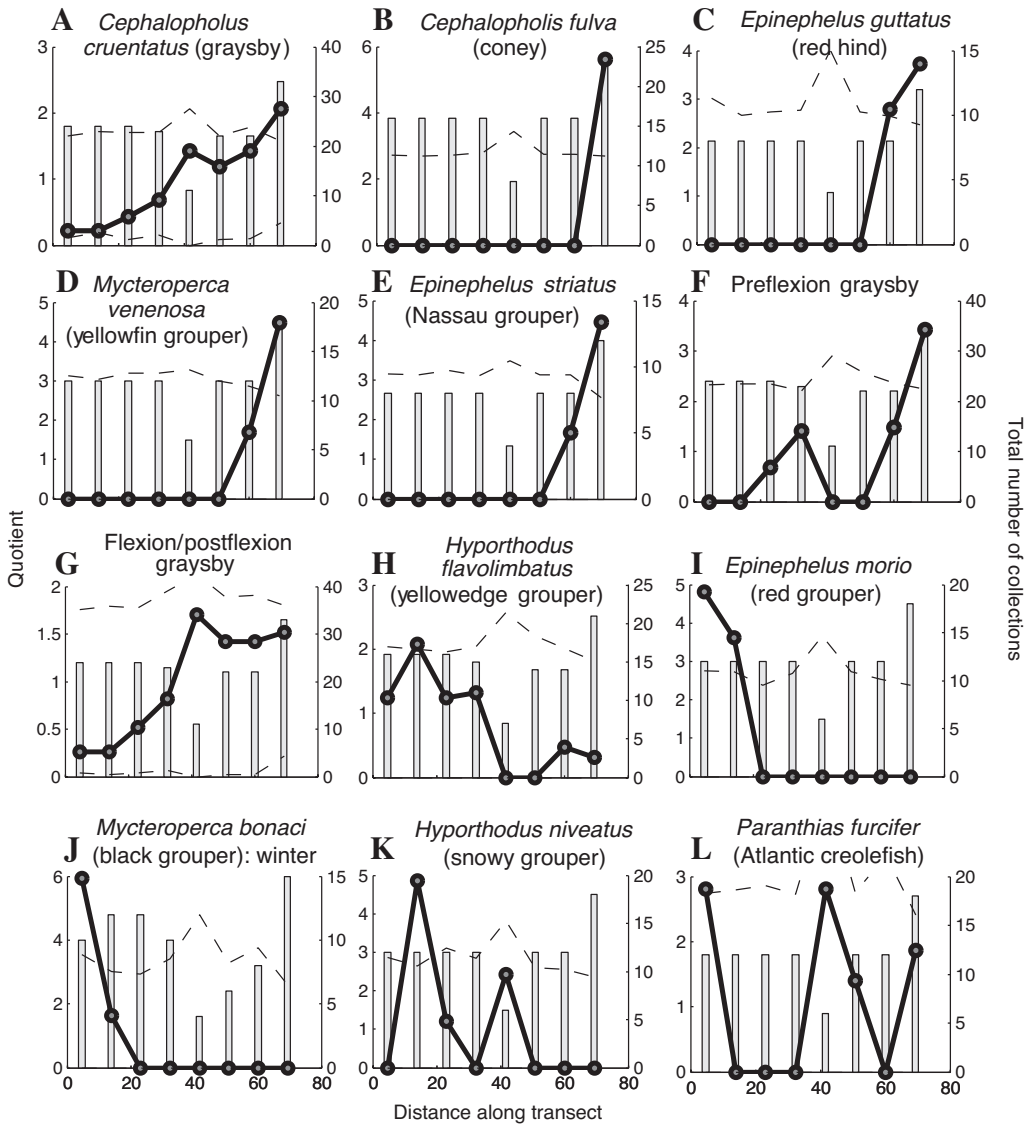

Figure 3

Quotients of larval grouper (proportion of larvae divided by proportion of collections) and total number of collections within 9.25-km bins across the Straits of Florida transect. Quotient plots for members of the eastern Straits of Florida larval fish assemblage by species (**A–G**). Graysby (*Cephalopholis cruentata*) is divided into (F) preflexion and (G) flexion or postflexion larvae. Quotient plots for members of the western Straits of Florida larval fish assemblage (**H–K**) and for a species collected evenly across the transect (**L**). The solid line represents the quotient of larval occurrence, and the dashed lines are the upper and lower confidence intervals for the null hypothesis (i.e., even distribution across the transect). The *x*-axis spans the length of the transect from west to east. The bars represent the number of samples collected in 9.25-km bins. Only months in which the species occurred (Table 4) were included in analyses.

1). The largest *Mycteroperca* spp. specimens, identified by anal-fin ray counts >10 (Smith, 1971), were collected in the northern Gulf of Mexico only during April–June and primarily on the TX–LA shelf (Fig. 4F; Table 1). In addition, two larger *Mycteroperca* spp. larvae were collected in January and February off the Yucatan Peninsula in the southern Gulf of Mexico.

Like the *Mycteroperca* spp., members of two multispecies groups (larvae with small spinelets, and those with long, curved spinelets) were collected in April–June and September–November in all three identified Gulf of Mexico spawning subregions (Fig. 4, B–C). Specimens of the group of species with standard tail pigment and long and curved spinelets collected in

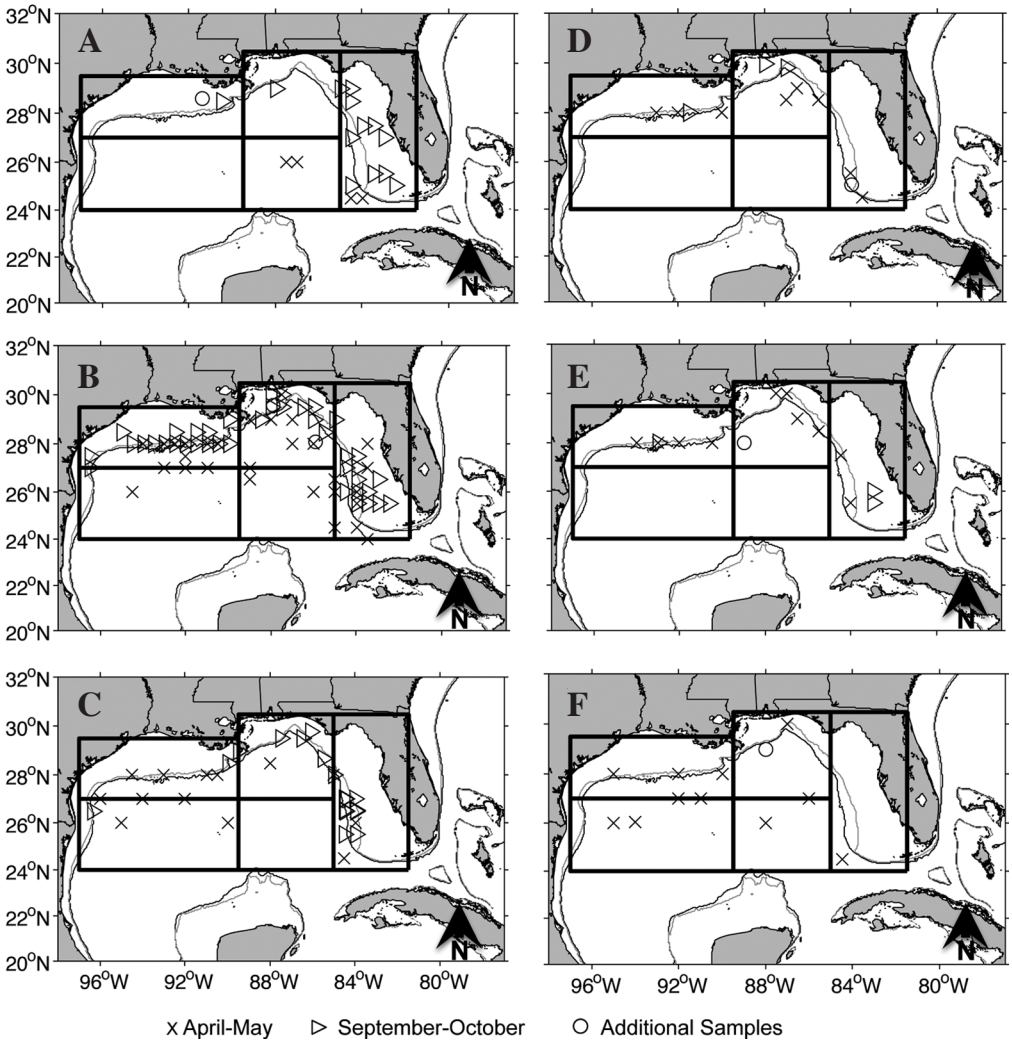

Figure 4

Spatial and seasonal distributions of species and morphologically discrete groups of species with standard pigment (Marancik et al., 2010) collected during Southeast Area Monitoring and Assessment Program Gulf of Mexico surveys 1982–2005. (**A**) *Cephalopholis cruentata* (graysby); (**B**) specimens with small spinelets; (**C**) specimens with long and curved spinelets; (**D**) small *Mycteroperca* with the dorsal-ventral tail pigment pattern; (**E**) mid-size specimens of *Epinephelus itajara* or *Mycteroperca* spp., and (**F**) large specimens of *Mycteroperca* spp. X=specimens collected in spring (April–May), triangles=specimens collected in fall (September–October), circles=specimens collected outside spring and fall surveys. Boxes denote the five subregions of the northern Gulf of Mexico used for analyses.

the spring survey were mostly collected on and near the shelf break off the coasts of Texas, Louisiana, Mississippi, and Alabama (TX–LA and MS–AL–nFL). Specimens collected during the fall survey, however, occurred only on the wFL shelf (Fig. 4C). Members of this species group may include postflexion *Epinephelus drummondhayi, E. itajara, Hyporthodus flavolimbatus, H. nigritus, H. niveatus,* members of the *Mycteroperca* genus that lack cleithral symphysis pigment, or a combination of these species (Marancik et al., 2010).

Four species (*C. fulva*, *H. mystacinus*, *Gonioplectrus hispanus*, and *Paranthias furcifer*) and a two-species complex (either *E. striatus* or *M. venenosa*) also were collected in SEAMAP survey samples, but in very low numbers (Table 1). These larvae occurred on the TX–LA shelf and offshore of the wFL shelf. There were too few larvae to define seasonal patterns.

Influence of environmental factors

The generalized additive models used to evaluate the influence of select physical variables measured during SEAMAP plankton surveys in the Gulf of

Table 5

Results of best generalized additive models (GAMs) predicting percent frequency of occurrence for each subregion and season. TX–LA=Texas–Louisiana shelf, MS–AL–nFL=Mississippi–Alabama–northern Florida Shelf, and wFL=west Florida Shelf. ****P*-value <0.001, *** *P*-value <.01 ** *P*-value < 0.05 and *P*-value <0.1. Covariates that were not included in the model are marked with "na." The percentage of the deviance from the null model explained by this model is labeled "% Dev. exp." Generalized cross-validation (GCV) and Akaike's information criterion (AIC_c) were used to determine the best model from all combinations of covariates (year, surface temperature, surface salinity, and water depth). r^2=the adjusted coefficient of determination. AIC_c weight is a measure of the likelihood that the model is the best representation of the data (values range from 0 to 1).

Region	Season	Intercept	Year	Surface temperature	Surface salinity	Water depth	r^2	% Dev. exp.	GCV	AICc	AICc weight
TX-LA	Fall	****2.719	na	*1.0	na	na	0.0958	14.3	3.7348	84.89725	0.162
MS-AL-nFL	Fall	****6.0257	**1.0	na	na	na	0.244	28.4	21.453	119.8604	0.383
wFL	Fall	****6.5939	***2.920	**2.436	***2.195	***2.076	0.874	94.5	12.247	87.30072	0.998
MS-AL-nFL	Spring	****3.7240	****2.942	na	**2.141	**1.0	0.764	84	5.6079	89.9197	0.523

Mexico did reasonably well in predicting presence or absence of grouper in a subregion for a year under a given set of environmental conditions. The percent deviance from the null model explained by the best of these models ranged from 14.3% to 94.5%. The TX–LA shelf models examined explained very little of the deviance in the data, and several models fit the data almost equally (low AIC_c weights; Table 5). Thus, grouper occurrence in this subregion is not well explained by any combination of surface temperature, surface salinity, water depth, or year. The west Florida shelf model based on the fall survey data and the Mississippi–Alabama–north Florida shelf model based on the spring survey data were the most successful in predicting the occurrence of grouper larvae, describing 94.5% and 84% of the deviances, respectively. The significant covariates in each subregion by season GAM revealed changes in frequency of occurrence over time and regionally specific influences of water depth, surface salinity, and surface temperature (Table 5). Annual frequencies of grouper collections were sufficient for generating GAMs for the three subregions characterized by the presence of the smallest larvae for the fall season (Fig. 2, subregions b–d). The only spring data set with larvae collected in enough years to warrant modeling was the MS–AL–nFL shelf subregion. Models of Gulf subregions east of 90° W longitude (wFL shelf and MS–AL–nFL shelf) were positively correlated with year, with higher occurrence since the early to mid 1990s (Fig. 5). The west Florida shelf model was also significantly influenced by mean surface temperature (>29°C), mean surface salinity (>35.5), and water depth (>129 m) (Table 5, Fig. 5). The occurrence of grouper larvae in the gulf west of 90° W longitude (TX–LA shelf) was significantly influenced by mean surface temperature (>28°C; Table 5, Fig. 5), although this relationship was weak. The occurrence of grouper larvae in the north central Gulf (MS–AL–nFL) increased from 1990 to 2000, but was highest after 1995, in midrange surface salinities (34–35), and in mean water depth <350 m (Table 5, Fig. 6).

Change in occurrence over time

The Gulf of Mexico subregion by season GAMs revealed a change in grouper occurrence over the SEAMAP survey time series, with %FO highest after the mid 1990s. This shift was evident in the patterns of occurrence of the more abundant grouper species and species groups (Fig. 7). Before 1995, grouper occurrences were higher in the spring than in the fall. Since 1995, higher occurrences have been observed in the fall than in the spring. No *Mycteroperca* spp. (three size groups combined) were collected in the fall before 1995, but since 1995, these larvae have occurred in fall survey samples. Similarly, larval graysby were rarely collected before 1995 (occurring in 2 of the 10 years between 1986 and 1995), but they have become more common in samples during recent

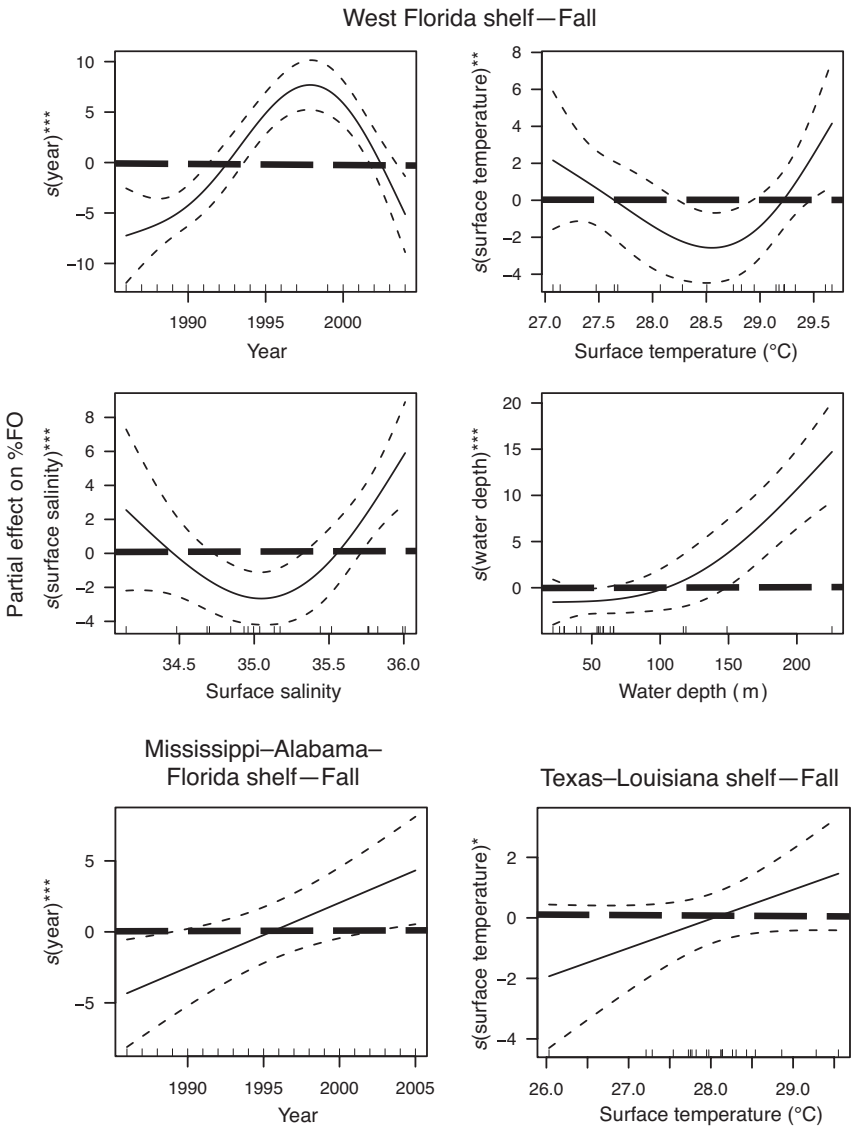

Figure 5

Partial effects [*s*(covariate); *y*-axis] of the generalized additive model (GAM) for each covariate (*x*-axis) used in the best subregion by season model of percent frequency of occurrence (%FO) of grouper larvae (all taxa combined) collected during Southeast Area Monitoring and Assessment Program fall sampling (September–October, 1986–2005) in the three subregions analyzed. Modeled partial effects (solid lines) and 95% confidence intervals (dotted lines) are shown for each included covariate. The dark dashed line marks no effect. Percent frequency of occurrence was higher for partial effects greater than 0, and lower for values less than 0. Vertical dashes along the *x*-axis denote actual sample values for the covariate. ****$P<0.001$, ***$P<0.01$, **$P< 0.05$.

decades (7 of the 10 years between 1996 and 2005) and are often collected in multiple months within a year. Graysby larvae have also become a significant percentage of the total catch of grouper larvae collected in the recent decade: 3–33% (mean=18.7%) before 1995; 5–100% (mean=40.3%) after 1995. A comparison of survey coverage for the two periods (1986–95, 1996–2005) revealed comparable sampling effort during spring and fall surveys. Compared with percent coverage during the period since 1995, the percent coverage during fall surveys before 1995 was similar or slightly higher, whereas in spring the percentages were similar or slightly lower before 1995. Therefore, differences in sampling effort did not

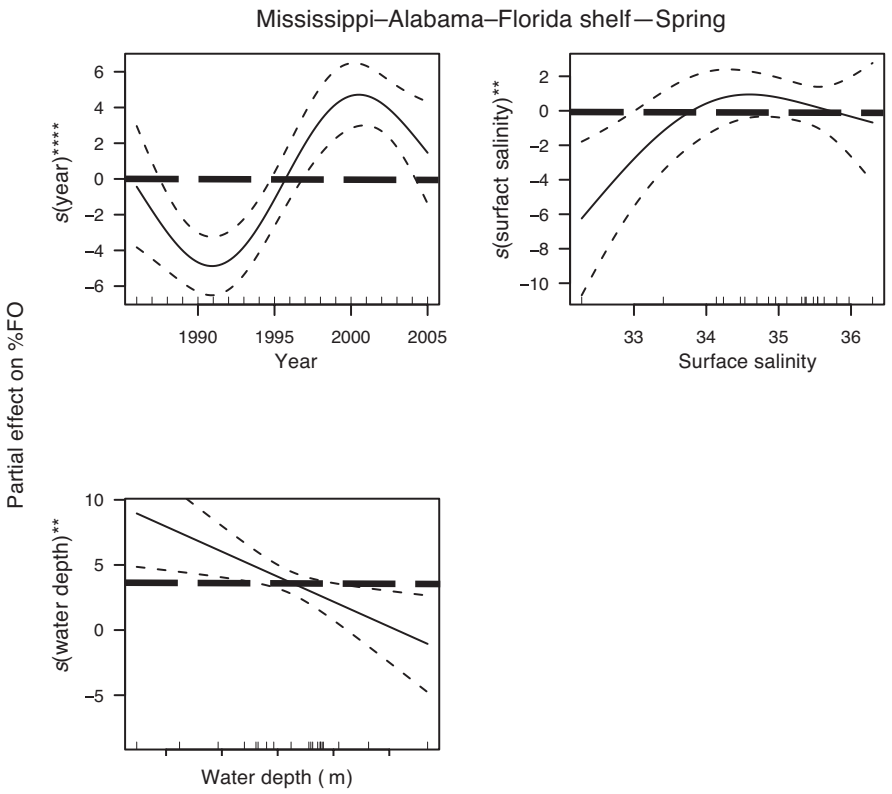

Figure 6

Partial effects [s(covariate); *y*-axis] of the generalized additive model (GAM) for each covariate (*x*-axis) used in the best subregion by season GAM of percent frequency of occurrence (%FO) of grouper larvae (all taxa combined) collected during Southeast Area Monitoring and Assessment Program spring sampling (April–May, 1986–2005) in the Mississippi–Alabama–northern Florida subregion. Modeled partial effects (solid lines) and 95% confidence intervals (dotted lines) are shown for each included covariate. The dark dashed line marks no effect. Percent frequency of occurrence was higher for partial effects greater than 0, and lower for values less than 0. Vertical dashes along the *x*-axis denote actual sample values for the covariate. ****$P<0.001$, ***$P<0.01$, **$P< 0.05$.

likely cause the observed increase in fall-spawned grouper larvae in the years since 1995.

Discussion

Seasonal occurrence

Larval grouper seasonality, as defined by the occurrence of larvae in collections from the Straits of Florida, relates directly to spawning season. Spawning likely occurs approximately one month (average pelagic larval duration <45 days; Colin et al., 1997; Lindemann et al., 2000; Fitzhugh et al., 2005) before occurrence of postflexion-stage larvae, and within two weeks of the occurrence of preflexion-stage larvae (Glamuzina et al., 2000; Leu et al., 2005). Because larvae were generally collected at the beginning of each month during collections in the Straits of Florida, actual spawning could have occurred in the month before collection. Although larvae were collected year-round, most larvae were collected during early February and March (Table 4)—a period that would correspond with a January through March spawning season. This is generally considered the primary spawning season of most northwest Atlantic groupers (Collins et al., 1998; Johnson et al., 1998; Brule et al., 2003; Nemeth et al., 2007; Starr et al., 2007). A second period of high larval species richness was observed during early September and October, indicative of spawning from August to October (Table 4; Bullock et al., 1996; Sadovy and Eklund, 1999; Richards et al., 2005).

Analysis of larval seasonal occurrence indicated longer spawning seasons than those identified in studies of adult groupers. Graysby are considered fall spawners throughout their range (Richards et al., 2005), and most graysby larvae were collected during July–October in shallow shelf waters on the west Florida Shelf (mean depth of 49.2 m vs. >60 m for all other taxa). However,

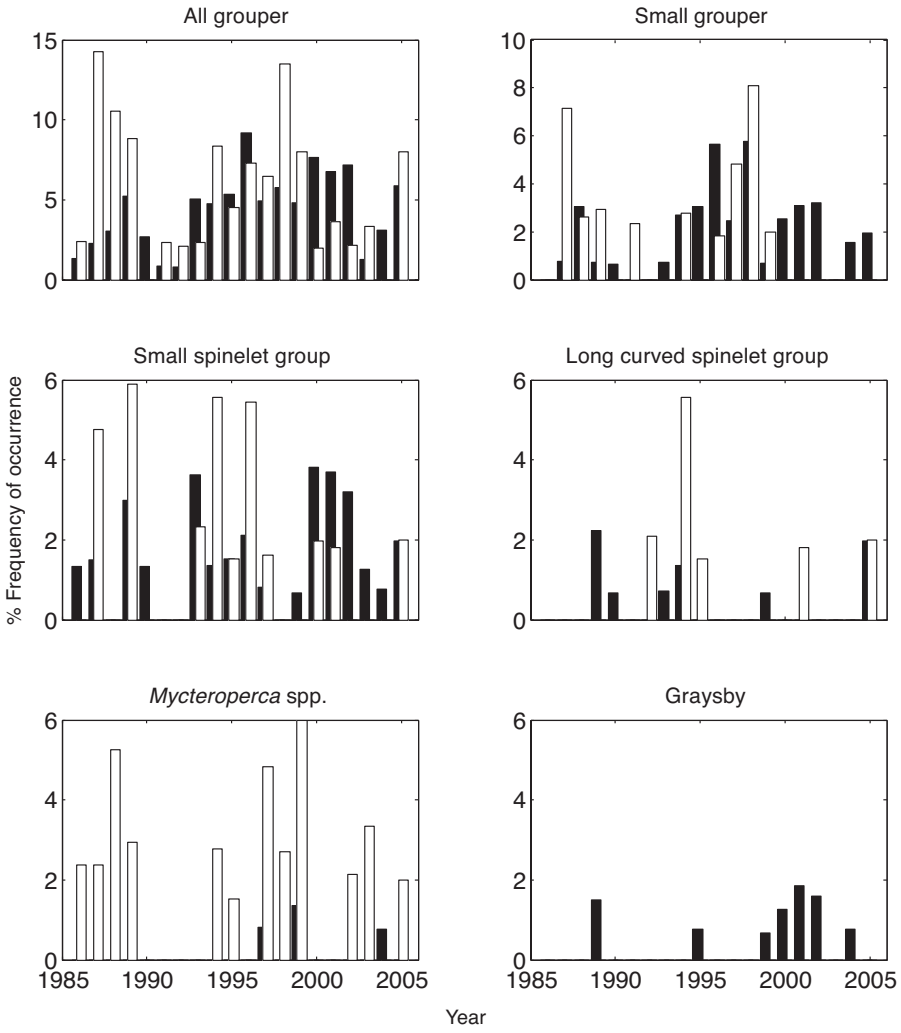

Figure 7

Interannual patterns in percent frequency of occurrence of the six most abundant taxa of grouper larvae collected in the northern Gulf of Mexico during Southeast Area Monitoring and Assessment Program spring (white bars) and fall (black bars) surveys. Spring data was constrained to stations at depths <900 m.

a few specimens were collected during April and May in deep offshore Gulf of Mexico waters (Fig. 4A). These larvae were morphologically identical to the larvae collected on the shelf; therefore misidentification is unlikely. Unlike the fall-spawned graysby, the spring-spawned graysby were collected in neuston nets (surface <0.5 m) and at stations with water temperatures warmer than surrounding stations (data not shown), indicating an association with Loop Current water that is transported north from the Caribbean Sea into the eastern Gulf of Mexico. These specimens may have been spawned locally and entrained in a Loop Current eddy or may have originated south of the study area (Campeche Bank or Caribbean Sea) and been carried north. Either way, these larvae represent an expanded spawning season (April–October) not previously recorded in the literature.

Similarly, spawning season determined through observation of adult red grouper in the Gulf of Mexico was limited to January–March (Johnson et al., 1998, Brule et al., 1999). A significant number of red grouper larvae from the Straits of Florida sampling were captured in May, indicating a spawning season extending from January to May. Burgos et al. (2007) collected spawning females from mid February to mid June in North and South Carolina—a period coinciding with the timing of our collection of larvae.

Spatial occurrence

Grouper larvae, in general, have a narrow distribution pattern regardless of water properties such as temperature and salinity. Grouper larvae were collected along

the shelf break throughout much of the northern Gulf of Mexico from Texas to southern Florida. Similarly, most of the Straits of Florida larvae were collected from stations closest to the coasts of Florida and the Bahamas (Fig. 3). A similar affinity for shelf edge habitat has been observed among adult grouper (Koenig et al., 1996; Brule et al., 1999; Sadovy and Eklund, 1999; Brule et al., 2003), and most spawning occurs inshore of or along the shelf break (Collins et al., 1998; Brule et al., 2003; Nemeth et al., 2007). Further, a higher specimen-to-sample ratio was observed in the Straits of Florida (665 individuals in 384 MOCNESS stations) than that from the Gulf of Mexico (544 individuals in 16950 bongo and neuston stations). Sampling gear (MOCNESS vs. bongo), sampling strategy (discrete depth vs. oblique), and location of sampling all contributed to the wide differences in the numbers of grouper larvae collected during the two sampling programs. The MOCNESS sampled more water per tow than the bongo nets, and proportionately more of the sampling occurred at depths likely to contain grouper larvae (<50 m). In addition, more of the Straits of Florida (including the area upstream from the sampling area) includes shelf edge habitat than the basin-wide sampling area of the Gulf of Mexico. This was especially the case during the spring SEAMAP survey (season of highest grouper occurrence) when the target sampling area is deep offshore water within the Gulf of Mexico. Thus, a higher percentage of grouper habitat (subsurface waters over shelf edge) was sampled along the transect through the narrow Straits of Florida than in the broad SEAMAP survey area within the Gulf of Mexico and likely accounted for many of the differences in catch rates between the two sampling programs.

Analysis of the larval data supported the conclusion that most Gulf of Mexico grouper species depend on shelf-edge habitat for spawning. Juveniles of many of these species move inshore to coastal and estuarine nursery habitats (Eggleston, 1995; Ross and Moser, 1995; Lindemann et al., 2000; Fitzhugh et al., 2005). However in the Straits of Florida, flexion and postflexion larval graysby were collected farther offshore than were preflexion larvae of the species (Fig. 3). At least two scenarios could explain this pattern in distribution. The offshore flexion and postflexion larvae collected in the straits could have been carried by the Florida Current into the sampling area from spawning sites as far away as the Gulf of Mexico or Caribbean Sea. Transport from upstream spawning locations explains the high diversity of grouper species collected in the area and is corroborated by genetic analysis of Gulf of Mexico and southeast United States populations (Zatcoff et al., 2004; Cushman et al., 2009). The fate of larvae carried away from coastal and estuarine habitat in the Loop–Florida–Gulf Stream currents is variable (Hare and Walsh, 2007; Richardson et al., 2009). Some individuals arrive at suitable habitat along the U.S. east coast far from spawning sites (e.g., bluefish [*Pomatomus saltatrix*]; Hare and Cowen, 1996), but many are carried too far north for survival (e.g., gray snapper [*Lutjanus griseus*]; Denit and Sponaugle, 2004) or never reach the coast (Hare and Walsh, 2007). Similarly, these later-stage larvae may have been advected offshore from nearby spawning sites and rely on regularly occurring oceanic events (e.g., gyres and meanders: Porch, 1998; frontal eddies: Sponaugle et al., 2005) or periodic events (e.g., wind storms: Shenker et al., 1993) to move them onshore toward nursery habitat. This second scenario would provide for some degree of self-recruitment. These scenarios may apply to other species of grouper; however, most species were collected too infrequently or in too narrow a size range to detect differences in distribution patterns between early life history stages. Further research is needed to determine the most likely processes driving the distribution patterns observed among Straits of Florida grouper larvae. The results of such an analysis, the identification of recruitment pathways and survival rates, would have major implications for the management of populations spawning in the area.

Specimens identified as either *E. itajara* or *Mycteroperca* spp. were collected during spring (majority) and fall surveys (Fig. 4E). The fall contingent was collected on the southwest Florida and Louisiana shelves and represents evidence of fall-spawning *Mycteroperca* spp. or a previously undocumented spawning location for *E. itajara*. Most species of *Mycteroperca* are known to spawn in the winter and spring months in the Gulf of Mexico and Caribbean (Hood and Schlieder, 1992; Bullock and Murphy, 1994; Brule et al., 2003; Fitzhugh et al., 2005), and there were no large *Mycteroperca* spp. larvae collected in the fall survey to confirm a fall spawning population (Fig. 4F). However, the spawning seasons of many species of *Mycteroperca* are unknown, and at least one species (*M. bonaci*) is believed to spawn year-round (Brule et al., 2003), and therefore fall-spawned *Mycteroperca* spp. are possible. *E. itajara* are known to spawn in fall (Sadovy and Eklund, 1999). Although they are believed to occur throughout the coastal Gulf of Mexico (Heemstra and Randall, 1993), no *E. itajara* spawning sites have been recorded in the northwestern Gulf of Mexico (Sadovy and Eklund, 1999). These specimens could indicate an undocumented spawning site for *E. itajara* in the northwestern Gulf of Mexico, but targeted sampling in the area and molecular identification of larvae would be needed to verify and locate a new spawning site. Genetic confirmation of a northwest Gulf of Mexico population may be possible because Brazilian, Belizian, and Florida populations of *E. itajara* are genetically highly separated (Craig et al., 2009).

Influence of environmental factors

Interannual variability in the occurrence of grouper larvae was influenced by hydrography. The variables involved and the extent of that involvement varied by subregion and season (Figs. 5 and 6). Surface temperature and salinity were significant factors in the fall west Florida shelf model, which together with year and water depth, explained over 90% of the deviance in the data. Surface salinity was also significant in

the Spring MS–AL–nFL subregion model and, along with year and water depth, explained over 80% of the deviance. These two models describe the importance of shelf-edge habitat. Low occurrence of grouper larvae in SEAMAP collections made it difficult to analyze fine spatial (within subregion) or temporal (within year) scale interactions with environment. Targeted sampling within subregions would be needed to better describe the relationship between the physical environment and larval occurrence.

Change in occurrence over time

The data presented here represent the best existing data set for examining long-term trends of larval grouper abundance in the southeast United States. We attempted to control for inconsistencies in sampling, but the results from this study cannot be fully separated from sampling bias, consistently low catches, nonspecies-level identifications, and missed peak spawning season for many commercially relevant species. However, our results provide evidence of a shift in grouper species composition toward fall-spawning populations over the SEAMAP time frame (Figs. 5–7). Spring-spawned larvae dominated collections before 1995, but in the more recent decade, fall-spawned larvae have come to dominate or have gone from nonexistent to present in larval collections. The relative increase in occurrence of fall-spawned larvae was best illustrated by the rapid rise in the number of larval graysby collected. In addition, a clear increase in the collection of fall-spawned members of a morphologically indistinguishable group of species (with small spinelets and standard tail pigment), including several commercially important species (namely, small *H. flavolimbatus* [<4.5 mm BL], *E. itajara, H. niveatus,* and possibly preflexion *Mycteroperca* spp. lacking pigment at the cleithral symphysis), was also observed (Fig. 7).

A shift in larval occurrence could result from a shift in abundance at the adult population level (e.g., changes in population size or spawning stock biomass), changes in the survivability of larvae (e.g., changes to maternal condition, fecundity, food availability, environmental regime, etc.), or a change in distribution. There is some evidence of changes in adult grouper population dynamics. For example, graysby larvae were one of the most abundant grouper species in our Straits of Florida collections and have become common in SEAMAP collections since 1995. Similarly, the occurrence of adult graysby increased off the coast of North Carolina between 1975 and 1992 (Parker and Dixon, 1998). Similar data on the abundance of adult graysby from the Florida Shelf is limited, but adult graysby are one of only three species of grouper that were not being overfished in the Florida Keys before 1996 (Ault et al., 1998) and were a dominant species on Florida Keys reefs in the early 1990s (Sluka et al., 1998). In addition, a decline in the abundance of larger grouper since the early 1990s (Bohnsack et al., 1994) could result in an increase in the abundance of smaller grouper species, like graysby (Sluka et al., 1998; Chiappone et al., 2000). However, increases in larval occurrence could also be the result of a shift in adult habitat use without an increase in population size. In the southern Caribbean, a significant shift in graysby distribution to deeper habitat coinciding with a reduction in coral cover has been observed (Nagelkerken et al., 2005). A similar shift in adult distribution on the west Florida shelf could explain the increase in larval occurrence observed in our study. Further examination of the potential causes of a shift in species dominance at the adult level and additional targeted investigations into larval survivability are needed to corroborate our findings of a shift in dominance in the northern Gulf of Mexico. However, the larval data here indicate that shifts in grouper abundance and species composition occurred over the last three decades.

Conclusions

Analysis of larval grouper distribution patterns provided a means of independently corroborating location and seasonality of spawning, but also allowed us to identify new patterns in grouper distribution and species composition in the Straits of Florida and northern Gulf of Mexico. The timing of larval occurrence, and thus the timing of spawning, for most species fell into one of two seasons, confirming what was already documented on spawning season for many species. However, two species, *Cephalopholis cruentatus* (graysby) and *Epinephelus morio* (red grouper), were collected during longer seasons than previously reported. Grouper larvae were collected in three distinct subregions of the Gulf of Mexico and along the shelf edge in both the gulf and Straits of Florida. Analysis of larval occurrence by subregional mean water depth, surface temperature, and surface salinity further corroborated the importance of shelf-edge habitat, particularly on the west Florida shelf in fall and the Mississippi–Alabama–north Florida shelf in spring. The species composition of grouper larvae in the Gulf of Mexico may have changed over the course of SEAMAP sampling. The frequency of occurrence of fall-spawned species has increased in relation to spring-spawned species since 1995. In the Straits of Florida, preflexion graysby were collected along the shelf edge, but flexion and postflexion larvae of the species were collected farther offshore. The distribution of later-stage graysby larvae could be evidence of processes directing self-recruitment or loss to the population. These data provided a first-time look at larval grouper distribution patterns over a large spatial and time scale and provided evidence of several topics needing further research.

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