

# REGIONAL VARIATION IN MERCURY AND STABLE ISOTOPES OF RED SNAPPER (*LUTJANUS CAMPECHANUS*) IN THE NORTHERN GULF OF MEXICO, USA

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Abstract—The presence of total mercury (Hg) in fish tissue and the potential associated health risks has become a global concern in marine ecosystems. Few studies have examined basin-scale variation in Hg accumulation in marine ecosystems, and determining if Hg concentrations in fish tissue vary across marine ecosystems is a key monitoring question. The present study evaluated Hg concentrations in red snapper (*Lutjanus campechanus*) tissue across three regions of the northern Gulf of Mexico (Alabama, Louisiana, and Texas, USA) and between two habitat types (oil and gas platforms and nonplatforms) within each region. Nitrogen ( $\delta^{15}$ N), carbon ( $\delta^{13}$ C), and sulfur ( $\delta^{34}$ S) stable isotopes were used to investigate ecological differences that may affect Hg concentrations among regions and between habitats. Mercury concentrations in red snapper tissue were positively correlated with fish total length. Regional differences in Hg concentrations were significant, with fish collected from Alabama having the highest concentrations and fish collected from Louisiana having the lowest. No significant difference existed in Hg concentrations. While  $\delta^{15}$ N did not differ significantly among the three regions, Texas red snapper were more enriched in  $\delta^{34}$ S and depleted in  $\delta^{13}$ C compared with Alabama and Louisiana red snapper. Although the majority of red snapper collected in the present study had Hg concentrations below safe consumption guidelines, regional differences suggest that spatially explicit monitoring programs may be important for basin-wide assessments. Environ. Toxicol. Chem. 2013;32:434–441. © 2012 SETAC

Keywords-Mercury Stable isotope Red snapper Gulf of Mexico

# INTRODUCTION

Concern over high total mercury (Hg) concentrations in fish in the United States has led to 15 states issuing marine fish consumption advisories. As a result, nearly 65% of the U.S. coastline is under an Hg advisory, including the entire Gulf of Mexico [1]. Despite the importance of marine fish in human diets and high levels of Hg observed by monitoring agencies [2], much of the research that has been conducted on aquatic Hg cycling has occurred in freshwater ecosystems [3]. Studies of Hg bioaccumulation in marine ecosystems often examine fish collected from limited geographic areas [4–7]. How Hg concentrations in fish vary within ocean basins has not been well studied. Assessing spatial variation in Hg concentrations across marine ecosystems is critical for the design of Hg-monitoring programs and could provide insight into the source of Hg in fish tissues, an area of ongoing research [reviewed in 8].

Inorganic Hg deposition and the biogeochemical processes that influence the conversion of inorganic Hg to the more toxic, accumulative methyl form can vary across large marine ecosystems, leading to spatial variation in Hg contamination of fish [9,10]. The dominant inputs of inorganic Hg to the Gulf are direct atmospheric deposition and riverine sources [11]. This includes the Mississippi and Atchafalaya Rivers, which drain 40% of the conterminous United States and are major contributors of inorganic Hg to areas directly off the Louisiana coast [9]. Additional regional sources that could influence Hg concentrations are the approximately 4,000 offshore oil and gas platforms that reside in the Gulf, with a majority located off the coast of Louisiana. Oil and gas production activities have been shown to enhance inorganic Hg concentrations in sediments around platforms [11], but the mechanisms responsible for this pattern are unexplained.

Sulfate-reducing bacteria convert inorganic Hg in marine sediments to methylmercury (MeHg) [12], which can accumulate in food chains when it is absorbed by primary producers, resulting in fish being exposed to MeHg primarily through the diet [13]. Prey preferences, ontogenetic shifts in habitat, migration, and seasonal movements are factors that complicate the determination of Hg bioaccumulation in marine fish. Therefore, studying species with some degree of site fidelity may help to assess the linkages between Hg levels in fish tissue and MeHg bioavailability in marine ecosystems [4].

Red snapper (*Lutjanus campechanus*) is a commercially important reef-associated fish in the Gulf, with an estimated site fidelity range of 25 to 60% per year [14–16], and a low-level consumer that feeds mostly in the benthos [17,18]. Typical of reef-associated fish, red snapper tend to aggregate near structured environments on the sea floor, including oil and gas platforms [19,20]. Thus, red snapper may be a good indicator species for assessing patterns of MeHg bioavailability and bioaccumulation in offshore environments (as indicated [4,6]), in addition to evaluating potential influences of Hg from oil and gas development.

The primary objective of the present study was to determine if spatial differences in Hg concentrations of red snapper tissue

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occur across three regions of the northern Gulf (Alabama, Louisiana, and Texas, USA). As a secondary objective, red snapper were collected from oil and gas platforms, along with other habitat types, within each region to establish if association with platforms affects red snapper Hg accumulation in marine environments. Stable isotope analysis of nitrogen ( $\delta^{15}$ N), carbon ( $\delta^{13}$ C), and sulfur ( $\delta^{34}$ S) was coupled with Hg analysis to evaluate the bioaccumulation of Hg in fish tissue [21,22] and to examine if possible differences in feeding ecology, fluvial influences, and habitat use may affect Hg concentrations of red snapper collected from different regions.

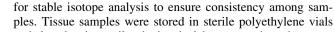
#### METHODS

## Sample collection

Red snapper were opportunistically sampled from recreational landings off the coasts of Port Aransas and Galveston, Texas, USA (n = 369); Port Fourchon, Louisiana, USA (n = 355); and Dauphin Island, Alabama, USA (n = 270), during the summers of 2007 and 2008 (Fig. 1). Fish were collected from two habitat types within each region: oil and gas platforms (both standing and toppled) and nonplatform habitats (natural bottom, cement blocks, and wrecks). Fish collected within 50 m of oil and gas platforms were categorized as platform. Stratified random sampling was used to select a subsample of 50 red snapper tissue samples analyzed for stable isotope ratios from each region and year (n = 300). These 50 samples were also divided by habitat, consisting of 25 collected from platform habitats and 25 from nonplatform habitats.

In the field, fish total lengths (TLs) were measured to the nearest millimeter. However, because samples were collected dockside from recreationally harvested fish, TL measurements could not be obtained for 287 individuals (29% of all individuals sampled). As part of a concurrent study, otoliths were removed from all fish collected. Estimated fish length was calculated based on power relationships between TL and otolith weight in milligrams [23]. In red snapper TL was strongly correlated with otolith weight (n=204;  $y=16.487x^{0.530}$ ,  $r^2=0.947$ ), and this relationship was used to approximate TL of the individuals that were not directly measured in the field.

Tissue samples for Hg and stable isotope analyses were removed in the field using a clean stainless steel scalpel. Epaxial



and placed on ice until arrival to the laboratory, where they were stored at  $-80^{\circ}$ C. In the laboratory, tissue samples were dried in a drying oven (model DX 600; Yamato) at 60°C for 24 h, and then individual samples were homogenized with a ball-mill grinder. Ground muscle tissue was stored in clean glass scintillation vials until Hg or stable isotope analysis.

muscle tissue was sampled from the anterior portion of the fillet

from both sides of the fish. The left tissue sample was des-

ignated for Hg analysis, while the right tissue sample was used

To determine if a relationship existed between Hg concentrations in red snapper tissues and Hg concentrations in sediment collected near platforms, a total of 112 sediment samples were collected from 38 platforms in July 2007 and May 2008 within the Grand Isle, South Timbalier, and Ship Shoal mineral leasing areas (Louisiana highlighted area, Fig. 1). Sediment samples were collected within 50 m of the platform using a Teflon-coated ponar grab. A plastic scoop was used to collect sediment samples from the center of the ponar grab sample, to minimize contamination risks. Samples were placed in WhirlPak bags and stored on ice until arrival to the laboratory, where they were stored at  $-80^{\circ}$ C. In the laboratory, all sediment samples were dried in a drying oven at 105°C for 24 h. To homogenize each individual sample, the dry sediment was pulverized with a clean agate mortar and pestle. Each pulverized sediment sample was stored in a clean scintillation vial until Hg analysis was performed.

#### Mercury analysis

Ninety-seven percent of the total Hg in red snapper tissue is MeHg [4,8]; thus, total Hg was analyzed as a proxy for MeHg. Total Hg analysis was performed on all red snapper tissue samples (n = 994) and sediment samples (n = 112) with a direct Hg analyzer (DMA-80; Milestone) using thermal decomposition, gold amalgamation, and atomic absorption spectrometry [24]. A detailed description of Hg analysis, including a description of standards used, was provided in a previous study [6]. Quality assurance included certified reference and duplicate samples. Reference samples (MESS-3, PACS-2, or DORM-2 [National Research Council of Canada]) were analyzed approximately every 10 samples, and the mean recovery was 97.7 ± 4.6% (mean ± standard deviation, n = 150). Duplicate samples were analyzed approximately every 20 samples, and the mean relative percent difference was 9.21 ± 7.67% (n = 64).

# Stable isotope analysis

Stable isotope analysis of nitrogen ( $\delta^{15}N$ ), carbon ( $\delta^{13}C$ ), and sulfur ( $\delta^{34}$ S) was coupled with Hg analysis to evaluate bioaccumulation of Hg in fish tissue [21,22,25] and to investigate possible feeding ecology and fluvial input differences that could alter Hg concentrations of red snapper collected among three sample regions. For stable isotope analysis, approximately 5 mg of dry ground tissue samples were placed in a tin boat with 10 mg of precombusted vanadium pentoxide ( $V_2O_5$ ). The isotopic composition of  $\delta^{15}$ N,  $\delta^{13}$ C, and  $\delta^{34}$ S was analyzed with a Finnigan MAT DeltaPlus continuous-flow stable isotope mass spectrometer (Thermo Fisher Scientific) attached to a Carlo Erba elemental analyzer at Louisiana State University following the batch analysis methods and standards used by Fry [26]. Replicate isotope analyses of N, C, and S generally agreed by 0.2‰ or better. Isotopic ratios are reported relative to the standards atmospheric  $N_2$  for  $\delta^{15}N$ , Vienna PeeDee belemnite for  $\delta^{13}$ C, and Vienna Canyon Diablo troilite for  $\delta^{34}$ S using the

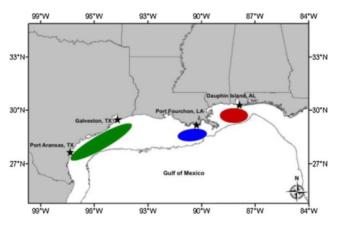


Fig. 1. Sampling regions along the continental shelf of the northern Gulf of Mexico where red snapper, *Lutjanus campechanus*, were collected during the summers of 2007 and 2008. The 200-m depth contour is present to represent the continental shelf edge. [Color figure can be seen in the online version of this article, available at wileyonlinelibrary.com.]

### formula

 $\delta_{\text{sample}}(\%_0) = (R_{\text{sample}}/R_{\text{standard}} - 1) \times 1,000$ 

where R represents the isotopic ratio. Although 300 samples were chosen for this analysis, due to inadequate sample material only 288 samples were used to determine stable isotope ratios of red snapper tissue.

## Statistical analysis

Normality was tested using a probability plot of the residuals versus the expected values, and the homogeneity of variances was tested with residual plots. To meet parametric assumptions, Hg concentrations were log-transformed prior to statistical analysis. A general linear model was used to examine the main effects of year, region, habitat, and TL on Hg concentrations of red snapper tissue. No significant year (p = 0.813) differences were detected in the general linear model; therefore, year was combined in the models. Fixed-effects analysis of covariance models were computed to test for differences by region and habitat in red snapper Hg concentration (n = 994), and TL was included as the covariate to correct for size-related differences in red snapper (as described in Wells et al. [6]). Tukey's honestly significant difference test was used to detect a posteriori differences among means [27]. Linear regressions were used to evaluate the relationship of red snapper Hg concentrations and stable isotope ratios to TL for each region. To assess regional and habitat variation in Hg concentrations and

stable isotope ratios of red snapper (n = 288), a two-factor multiple analysis of covariance was used, with Hg,  $\delta^{15}$ N,  $\delta^{13}$ C, and  $\delta^{34}$ S as the dependent variables [28]. Independent variables included region and habitat, with TL as the covariate. Following the significant multiple analysis of covariance model (Table 1), univariate analysis of covariance models were used to identify individual dependent variable responses. Reported values and standard errors are based upon least square means. A linear regression analysis was also used to determine if a relationship existed between Hg concentrations in Louisiana red snapper tissue and sediment samples collected from the same platforms. A total of 104 fish tissue samples collected from 15 different platforms were analyzed. To correct for TL, red snapper tissue Hg concentrations were lengthdetrended by subtracting the common within-group linear slope from the observed concentration.

Quadratic discriminant function analysis was used to test the ability of Hg concentrations and stable isotope ratios of red snapper tissues to distinguish region and habitat of capture. Jackknife cross-validated classifications were used to quantify classification success to respective regions and regions by habitat. Given differences in fish size among regions and between habitats, quadratic discriminant function analysis models were based on both original and length-corrected residuals on TL of Hg,  $\delta^{15}$ N,  $\delta^{13}$ C, and  $\delta^{34}$ S [29]. Differences between quadratic discriminant function analysis models using original and length-corrected values were small (<2% overall model)

Table 1. Results of analysis of covariance (ANCOVA) and multivariate analysis of covariance (MANCOVA) to test for differences in total Hg concentrations and stable isotope ratios of red snapper, *Lutjanus campechanus*, collected among three regions and between two habitat types in the Gulf of Mexico during the summers of 2007 and 2008

Model	Independent variables	F	df (factor, model error)	р
logHg ANCOVA ( $n = 993$ )	Region	126	2,983	< 0.001
	Habitat	1.29	1,983	0.256
	Region <sup>*</sup> habitat	15.9	2,983	< 0.001
	TL	205	1,983	< 0.001
	Region*TL	37.5	2,983	< 0.001
	Habitat*TL	8.53	1,983	0.004
MANCOVA $(n = 288)$	Region	56.9	8,552	< 0.001
	Habitat	6.46	4,275	< 0.001
	Region*habitat	3.04	8,552	0.002
	TL	31.9	4,275	< 0.001
	Region <sup>*</sup> TL	6.10	8,552	< 0.001
	Habitat*TL	4.04	4,275	0.003
logHg ANCOVA ( $n = 288$ )	Region	57.1	2,278	< 0.001
	Habitat	2.98	1,278	0.085
	Region*habitat	7.21	2,278	0.009
	TL	60.1	1,278	< 0.001
	Region *TL	16.0	2,278	< 0.001
	Habitat*TL	12.1	1,278	0.006
$\delta^{15}$ N ANCOVA ( <i>n</i> = 288)	Region	1.89	2,278	0.154
o 1112.000 11 (tr _200)	Habitat	16.1	1,278	< 0.001
	Region*habitat	0.16	2,278	0.855
	TL	51.7	1,278	< 0.001
	Region*TL	3.65	2,278	0.027
	Habitat <sup>*</sup> TL	0.05	1,278	0.830
$\delta^{13}$ C ANCOVA ( $n = 288$ )	Region	176	2,278	< 0.001
0 C M (0 - 200)	Habitat	13.2	1,278	0.003
	Region*habitat	2.01	2,278	0.136
	TL	1.17	1,278	0.280
	Region *TL	5.33	2,278	0.005
	Habitat <sup>*</sup> TL	1.31	1,278	0.254
$\delta^{34}$ S ANCOVA ( <i>n</i> = 288)	Region	22.9	2,278	< 0.001
	Habitat	0.07	1,278	0.791
	Region*habitat	2.76	2,278	0.065
	TL	12.7	1,278	0.003
	Region <sup>*</sup> TL	3.90	2,278	0.021
	Habitat <sup>*</sup> TL	3.22	1,278	0.074

classification success); thus, the original models were used. A canonical discriminant analysis was used to visualize differences in regional and habitat Hg concentrations and stable isotope ratios of red snapper tissues. All analyses were performed using the Statistical Analysis System [30] and JMP statistical software [31] with a significance level of  $\alpha = 0.05$ .

### RESULTS

Red snapper Hg concentrations varied significantly among the three regions sampled in the northern Gulf, whereas only  $\delta^{13}C$  and  $\delta^{34}S$  ratios varied significantly among regions (Table 1). The highest concentrations of Hg were observed in fish collected from Alabama (587  $\pm$  12.5 ng/g dry wt), followed by Texas ( $504 \pm 12.1 \text{ ng/g}$  dry wt), and then Louisiana  $(349 \pm 13.3 \text{ ng/g dry wt})$ . Nitrogen ratios in red snapper tissue samples did not differ significantly (p = 0.154) among the three regions sampled. Alabama and Louisiana red snapper tissue samples did not differ significantly in  $\delta^{13}C$  and  $\delta^{34}\bar{S}$  but were significantly enriched in  $\delta^{13}C$  (p < 0.001) and depleted in  $\delta^{34}$ S (p < 0.001) compared with Texas red snapper samples (Fig. 2). Relationships between total Hg concentrations and  $\delta^{15}$ N in red snapper from Alabama, Louisiana, and Texas were positively correlated with TL (Fig. 3). No significant relationship existed between  $\delta^{13}C$  and TL for red snapper, whereas  $\delta^{34}$ S and TL were negatively correlated (Table 1).

Total Hg concentrations in red snapper tissue samples did not differ significantly between habitat types (Table 1), with mean values of  $490 \pm 12.5$  ng/g dry weight for nonplatform habitats and  $470 \pm 8.4$  ng/g dry weight for platform habitats. While the habitat and region interaction was significant (Table 1), red snapper Hg concentration patterns between habitats within regions were not consistent. No significant difference existed between habitats in Alabama (p = 0.062). Louisiana platform habitats had higher Hg concentrations than nonplatform habitats (p = 0.003), and nonplatform habitats in Texas had higher Hg concentrations than platform habitats (p = 0.017). For this reason, habitat types were combined for Hg and TL correlation analyses. In addition, no significant relationship existed between Hg concentrations of Louisiana red snapper tissue samples and platform sediment ( $r^2 = 0.24$ , p = 0.064,  $F_{1,14} = 4.11$ ).

As with total Hg concentrations, it was difficult to distinguish a clear pattern in stable isotope ratios between platform and nonplatform habitats. Overall, a significant enrichment in  $\delta^{15}$ N and  $\delta^{13}$ C was observed at platform habitats relative to nonplatform habitats, with no difference observed in  $\delta^{34}$ S (Table 1). However, only Louisiana red snapper collected from platforms were significantly enriched in  $\delta^{15}$ N relative to fish collected from nonplatform habitats (p = 0.021), and only Texas red snapper collected at platforms were significantly enriched in  $\delta^{13}$ C compared to fish collected at nonplatform habitats (p = 0.026). No significant differences existed between habitats within regions for  $\delta^{34}$ S ratios (Table 1, Fig. 2).

Mean jackknifed classification accuracies of the quadratic discriminant function analysis models based on total Hg and stable isotope ratios of red snapper tissue samples were more successful at correctly classifying collection region than habitat

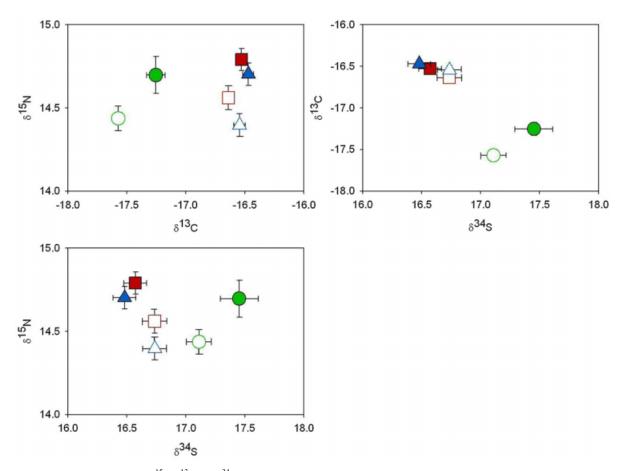


Fig. 2. Comparison of regional means of  $\delta^{15}$ N,  $\delta^{13}$ C, and  $\delta^{34}$ S of red snapper, *Lutjanus campechanus* (n = 288), collected from platform (closed) and nonplatform (open) habitats among Alabama (square), Louisiana (triangle), and Texas (circle) during the summers of 2007 and 2008. [Color figure can be seen in the online version of this article, available at wileyonlinelibrary.com.]

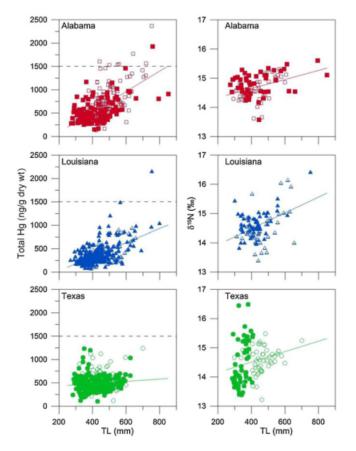


Fig. 3. Left column scatterplots represent relationships between total Hg and total length (Alabama, n = 269,  $y = 2.09^{*}TL - 307$ ,  $r^{2} = 0.32$ , p < 0.001; Louisiana, n = 355,  $y = 1.50^{\circ}$ TL – 272,  $r^2 = 0.30$ , p < 0.001; Texas, n = 369,  $y=0.25^{*}TL - 383$ ,  $r^{2}=0.02$ , p=0.016), and right column scatterplots represent relationships between  $\delta^{15}N$  and TL (Alabama, n = 93, y = $0.001^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.002$ ; Louisiana,  $n = 98, y = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.003^{*}TL - 14.1, r^{2} = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.003^{*}TL - 14.1, r^{2} = 0.003^{*}TL - 14.1, r^{2} = 0.14, p = 0.003^{*}TL - 14.1, r^{2} = 0.14, r^{2} = 0.003^{*}TL - 14.1, r^{2} = 0.003^{*}TL$ 13.2,  $r^2 = 0.23$ , p < 0.001; Texas, n = 97,  $y = 0.002^*$ TL - 13.6,  $r^2 = 0.09$ , p < 0.003) of red snapper, Lutjanus campechanus, collected during the summers of 2007 and 2008. Linear fit lines are through all points to signify regional differences. Fish collected from platform (closed) and nonplatform (open) habitats are illustrated to show concentrations did not differ between habitats. Dashed lines represent the U.S. Environmental Protection Agency's guideline for safe fish consumption. Analyses were performed on log-transformed data, and raw data are presented in the graphs. [Color figure can be seen in the online version of this article, available at wileyonlinelibrary.com.]

type within regions. Regional differences yielded an overall classification success of 76% (Table 2), with the majority of misclassifications occurring between Alabama and Louisiana. Adding habitat diminished overall classification success to 54% (Table 3), which is not surprising given the lack of a consistent pattern in Hg concentrations or stable isotope ratios between habitat types. The low classification success between habitats was primarily due to misclassifications within regions. The canonical discriminant analysis plot further illustrates the separation of regions and overlap of habitats within regions (Fig. 4).

#### DISCUSSION

Red snapper Hg concentrations were dependent on the region of capture within the Gulf, with fish collected off the coast of Alabama having the highest concentrations of Hg, followed by Texas, and then Louisiana. Five percent of fish collected off the coast of Alabama exceeded U.S. Environmental Protection Agency guidelines of 1,500 ng/g dry weight

Table 2. Jackknife cross-validation classification success (%) of red snapper, *Lutjanus campechanus*, to three regions in the Gulf of Mexico; percentages were estimated from quadratic discriminant function analysis of total Hg concentrations and stable isotope ratios

Region	% Correct	
Alabama	73	
Louisiana	67	
Texas	87	
Total	76	

for safe consumption, whereas no fish collected off the coast of Texas and <1% of fish collected off the coast of Louisiana exceeded these guidelines (Fig. 3). While a majority of Alabama samples were collected from fishing rodeos, which resulted in the average size of fish collected from Alabama being larger than the average size of fish collected from other regions, Alabama red snapper still had higher concentrations of Hg even when equivalent size classes (i.e., 400–600 mm) were considered.

Regional differences in Hg availability may explain observed differences in red snapper Hg concentrations among regions. According to previous studies, direct atmospheric deposition is the primary source of Hg into the Gulf ecosystem, with little influence from riverine inputs [9,32]. Therefore, regional differences in red snapper Hg concentrations would not be expected to be caused by differences in the amount of Hg deposition from major rivers emptying into the northern Gulf. Conversely, the Mississippi River discharge creates a seasonally large hypoxic zone that extends from the Mississippi River delta to the upper Texas coast, with isolated regions off Galveston and Freeport, Texas [33]. One characteristic of the Gulf hypoxic zone is the release of hydrogen sulfide from sediments [33], which can inhibit MeHg production [34,35]. As a result, spatial variability observed in Hg concentrations may not be a direct result of riverine inputs of Hg; rather, the increased sulfide levels associated with hypoxic conditions may yield reduced Hg concentrations in fish from Louisiana and Texas compared to Alabama.

Alternatively, the observed regional differences in Hg concentration may be due to demographic characteristics of red snapper. Mercury concentrations can increase as a function of fish age and size due to trophic magnification through the food web and increased activity costs linked with mating and foraging [13,36]. Generally, faster-growing fish have lower Hg concentrations than slower-growing fish at specific lengths, depending on consumption rates and activity costs [36,37]. Fischer et al. [38] observed Texas red snapper to be significantly smaller in mass-at-age compared to Alabama and Louisiana red snapper. Additionally, female red snapper collected off the

Table 3. Jackknife cross-validation classification success (%) of red snapper, *Lutjanus campechanus*, between two habitat types within three regions in the Gulf of Mexico; percentages were estimated from quadratic discriminant function analysis of total Hg concentrations and stable isotope ratios

Region*habitat	% Correct	
AL platform	48	
AL nonplatform	56	
LA platform	45	
LA nonplatform	22	
TX platform	74	
TX nonplatform	78	
Total	54	

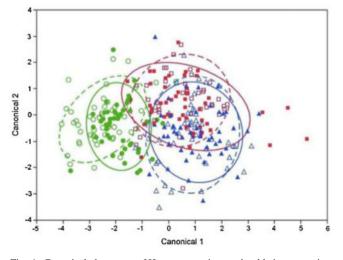


Fig. 4. Canonical plot scores of Hg concentrations and stable isotope ratios of red snapper, *Lutjanus campechanus*, collected from platform (closed) and nonplatform (open) habitats among Alabama (square), Louisiana (triangle), and Texas (circle) during the summers of 2007 and 2008. Ellipses indicate 95% confidence levels. [Color figure can be seen in the online version of this article, available at wileyonlinelibrary.com.]

coast of Alabama attained maturation at smaller sizes and younger ages than females collected off the coast of Louisiana [39]. Based on these studies, Hg concentrations in fish collected from Texas would be expected to be highest due to regional variability in growth rates; however, earlier maturation of Alabama red snapper may increase their activity costs, resulting in the observed higher Hg concentration levels.

Examination of  $\delta^{15}$ N,  $\delta^{13}$ C, and  $\delta^{34}$ S allowed for the evaluation of whether variation in Hg concentrations could be caused by diet and trophic position differences of red snapper among regions. In the present study, a positive linear relationship existed between Hg concentrations and trophic position ( $\delta^{15}$ N) with TL, suggesting the potential for bioaccumulation of Hg with increase in size and trophic position [40–42], as has been found in previous studies [6,10]. The average  $\delta^{15}$ N values of phytoplankton, the dominant basal resource for red snapper (S.T. Daigle, 2011, Master's thesis, Louisiana State University, Baton Rouge, LA, USA), can vary among sample regions from 3 to 10‰ (Table 4) [18,44,45]. However, these ratios were not significantly different in red snapper tissue collected from the three different regions, making interpretation of this result difficult. Alabama and Louisiana red snapper were more

Table 4. Mean nitrogen ( $\delta^{15}$ N) and carbon ( $\delta^{13}$ C) stable isotope ratios of primary producers collected among three regions of the Gulf of Mexico during previous studies

Producer	Region	$\delta^{15}N$	$\delta^{13}C$
Phytoplankton	AL/MS LA	$5.9^{\rm a}, 9.9^{\rm b}$ $2.7^{\rm c}, 5.1^{\rm d}$	$-22.7^{a}, -21.8^{b}$ $-22.1^{c}, -20.9^{d}$
ВМА	TX AL	7 <sup>e</sup> 7.2 <sup>a</sup>	$-21^{e}$ -19.9 <sup>a</sup>
2	LA		_
	1	_	

<sup>a</sup>Wells et al. [18].

<sup>b</sup> Moncreiff and Sullivan [43].

<sup>d</sup> Daigle (S.T. Daigle, 2011, Master's thesis, Louisiana State University, Baton Rouge, LA, USA).

<sup>e</sup>Rooker et al. [44].

BMA = benthic microalgae.

enriched in  $\delta^{13}$ C and depleted in  $\delta^{34}$ S compared with Texas red snapper. The average  $\delta^{13}$ C of phytoplankton does not vary among the sample regions (approximately -21%; Table 4) [18,44,45; S.T. Daigle, 2011, Master's thesis, Louisiana State University, Baton Rouge, LA, USA]; hence, regional differences in fish tissue may reflect variation in diet. Benthic feeders tend to be more enriched in  $\delta^{13}C$  and depleted in  $\delta^{34}S$  than pelagic feeders [46,47]. For instance, benthic algae is more enriched in  $\delta^{13}$ C compared with marine phytoplankton (-17 and -21%, respectively), whereas benthic plants and microalgae absorb sulfides depleted in  $\delta^{34}S$  compared with sulfate of seawater [47]. To examine the benthic contribution to red snapper, Wells et al. [18] analyzed benthic microalgae collected off the coast of Alabama and found it had an average  $\delta^{13}C$  value of -19.9‰. Furthermore, Thomas and Cahoon [46] showed that sulfur was vitally important for separating benthic and pelagic food sources for five different fish species. While results from the present study may suggest that Texas red snapper have a more pelagic diet than Alabama and Louisiana red snapper, consistent  $\delta^{15}$ N ratios would imply that red snapper are feeding at the same trophic level across all regions. Explicitly interpreting red snapper feeding ecology with stable isotopes is complicated by the possibility that baseline isotopic ratios may differ among regions. Additional dietary analysis is needed before definitive conclusions can be made about regional differences in red snapper feeding ecology and if these differences contribute to variations in Hg concentrations.

Habitat type was not a significant factor affecting Hg concentrations in red snapper throughout the northern Gulf. Furthermore, Hg concentrations in Louisiana red snapper tissue samples collected from platforms were not significantly correlated with Hg levels in platform sediments. Relatively few pristine natural reefs and hard-bottom substrate occur off the coast of Louisiana due to the abundance of platforms in the area. One concern is that oil production activity increases Hg to the surrounding area because drilling fluids containing barite, used in the process can elevate the Hg concentration in sediments around platforms. Recent studies showed a decrease of total Hg and MeHg in sediment samples with increasing distance from platforms [11,48]. Dietary studies have revealed that red snapper prefer sand- and mud-associated prey, with only a small percentage of their diet consisting of reef-associated species [17,18], such as those inhabiting platforms. Although red snapper aggregate near platforms, they tend to periodically move away from platforms, possibly for foraging purposes [49; M. McDonough, 2009, Master's thesis, Louisiana State University, Baton Rouge, LA, USA]. These movement patterns, along with different habitat types in close proximity to each other, may explain the negligible difference in habitat Hg concentrations and the low classification success by habitats in the present study.

The majority of red snapper sampled in the present study pose limited risk to consumers, with fish from all three regions having lower average concentrations of Hg than most other Gulf species examined in previous studies [50]. The discriminant analysis indicated that regional classification success was higher than habitat classification success. This further confirms that region of capture has more influence over Hg concentrations and that platforms most likely are not a major contributor of Hg to red snapper in the northern Gulf. If other fish species with higher overall levels of Hg contamination also exhibit regional variation in Hg, then these differences could be important when setting advisories and determining risk. Future studies should determine if other species exhibit spatial varia-

<sup>&</sup>lt;sup>c</sup> Wells and Rooker [45].

tion in Hg concentration and further explore the potential mechanisms responsible for this pattern.

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