TITLE

Regional variation in mercury bioaccumulation among NW Atlantic Golden (*Lopholatilus chamaeleonticeps*) and Blueline (*Caulolatilus microps*) Tilefish

RUNNING TITLE

THg bioaccumulation among NW Atlantic Tilefish

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KEY WORDS

body size, continental shelf, Malacathidae, resource partitioning, sex, nitrogen stable isotopes

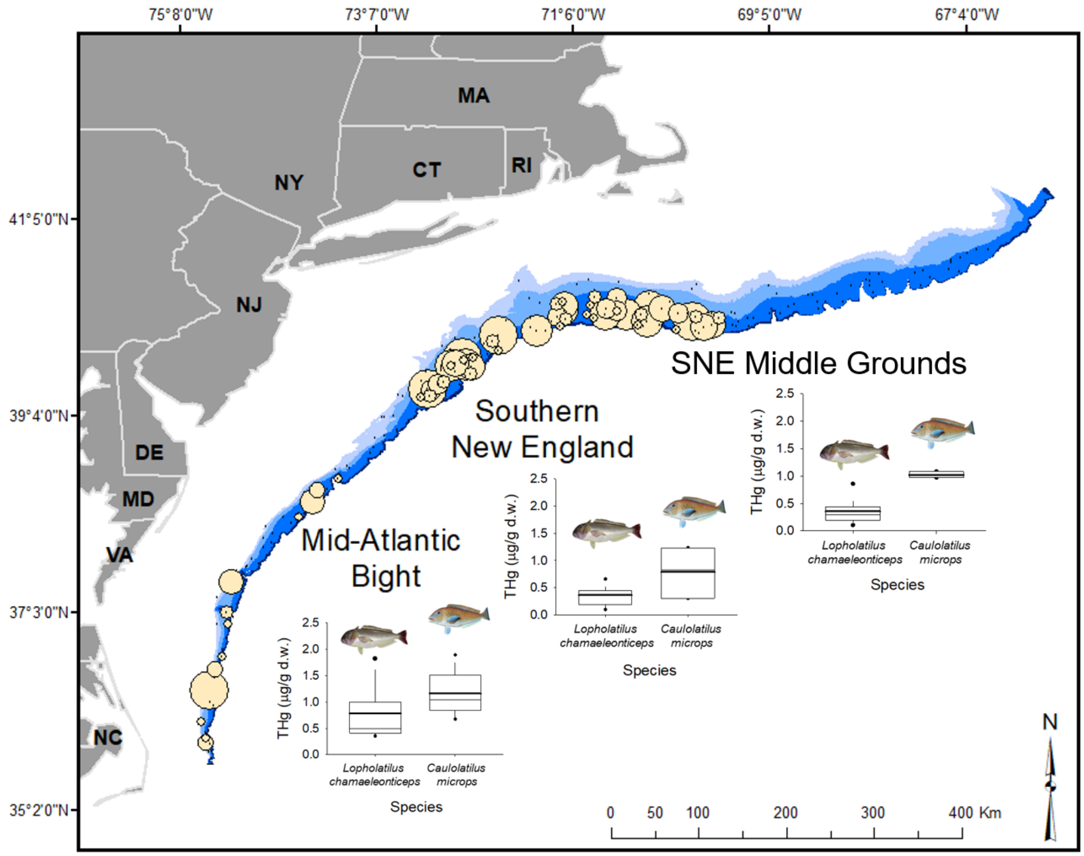
HIGHLIGHTS

- Total mercury (THg) concentrations were examined in Tilefishes in the NW Atlantic.
- Intra- and inter-specific THg bioaccumulation patterns relate to body size and region.
- THg concentrations were low with <2% fish exceeding restrictive USEPA guidelines.
- THg in NW Atlantic fish was lower than reported for Gulf of Mexico conspecifics.
- Findings inform guidance toward regional THg contamination of high-value fisheries.

ABSTRACT

Mercury (Hg) concentrations in fishes from the NW Atlantic Ocean pose concern due to the importance of this region to U.S. fisheries harvest. In this study, total Hg (THg) concentrations and nitrogen stable isotope ($\delta^{15}N$) values were quantified in muscle tissues sampled from Golden (*Lopholatilus chamaeleonticeps)* and Blueline (*Caulolatilus microps*) Tilefish collected during a fishery-independent survey conducted in the NW Atlantic to compare bioaccumulation patterns between these species. Total Hg concentrations averaged (± SD) 0.4 ± 0.4 µg/g dry weight (d.w.) for *L. chamaeleonticeps* and 1.1 ± 0.7 µg/g d.w. for *C. microps* with < 2% of all sampled fish, those > 70cm fork length, exceeding the most restrictive USEPA regulatory guidelines for human consumption (THg > 0.46 ug/g w.w.), when converted to wet weight concentrations. The THg concentrations reported here for individuals from the NW Atlantic stock are comparable to those reported for similarly sized individuals collected from the SW Atlantic stock but notably lower than those reported for Gulf of Mexico *L. chamaeleonticeps*, indicating different Hg exposure and assimilation kinetics for fish from the NW Atlantic, and highlights the broad geographic variability of Hg bioaccumulation among Tilefish stocks. *Caulolatilus microps* had higher $\delta^{15}N$ values relative to *L. chamaeleonticeps* and a pattern of decreasing THg concentrations was also present from south to north across the study range. It is concluded that this trophic difference and spatial pattern in Tilefish THg concentrations emphasizes the habitat and resource partitioning mechanisms described for these sympatric species that permits their coexistence in the continental shelf environment. Importantly, regional variability in THg concentrations accentuate the possible roles of fine-scale biotic and abiotic processes that can act to regulate Hg bioaccumulation among individuals and species.

GRAPHICAL ABSTRACT



Caption: Regional THg (µg/g d.w.) content in muscle tissue of two Tilefish species from the NW Atlantic (images: https://www.fishwatch.gov/profiles/Tilefish). Colored circles represent catch with larger circles indicating greater catch.

DATA DEPOSITION

Data are available from the Dryad Digital Repository: https://doi.org/10.5061/dryad.qnk98sfcs (Olin et al. 2020) and (Roose et al. Submitted).

INTRODUCTION

Habitat selection and foraging behaviors contribute to mercury (Hg) bioaccumulation in marine top predator and invertebrate species (Choy et al. 2009). Among North American marine commercial fishes, Hg concentrations in Atlantic King Mackerel (*Scomberomorus cavalla*), Swordfish (*Xiphias gladius*) and Gulf of Mexico (GOM) Golden Tilefish (*Lopholatilus chamaeleonticeps*) are sufficiently high to restrict consumption for sensitive population groups. Mercury concentrations quantified in Atlantic King Mackerel and Swordfish are primarily associated with their carnivorous diets and roles as top predators in marine food webs (Cai et al., 2007; Choy et al., 2009). In contrast, *L. chamaeleonticeps* and Blueline (*Caulolatilus microps*) Tilefish are demersal fishes that inhabit hummocky terrain along continental shelf-edge and slope environments at depths ranging from 80–500 m (Able et al., 1982; Twitchell et al., 1985; Pierdomencio et al., 2015). Tilefish diets also generally consist of lower trophic level invertebrates including crustaceans, annelids, mollusks, echinoderms, and fishes (Freeman and Turner, 1977; Ross, 1982; Steimle et al.,1999). Such contrasts in Tilefish habitat and diet preferences relative to pelagic top predators predict that Hg concentrations in these species should be lower based on Hg bioaccumulation kinetics and the phenomenon of food web biomagnification (Morel et al., 1998). However, recent studies have demonstrated substantial spatial variability in Hg concentrations among Tilefish across relatively broad geographic scales.

Deep-water oceanic ecosystems represent a major sink of global Hg emissions (Mason and Sheu, 2002; Driscoll et al., 2013). However, Hg concentrations can vary markedly within and among ocean basins and this variation has been observed in Hg bioaccumulation trends in marine species (Aston et al., 1972; Lamborg et al., 2014). Evidence for regional differences in Hg levels exists within a species (Adams and McMichael, 2007; Harris et al., 2012; Sinkus et al., 2017). Differences in fish muscle tissue Hg concentrations between the GOM and SW Atlantic have been documented for several species (Adams and Onorato, 2005; Adams and McMichael, 2007). The GOM represents a Hg bioaccumulation hotspot for *L. chamaeleonticeps* with concentrations measured in fish collected from this region frequently exceeding regulatory agency consumption advisories (Perrot et al., 2019). Perrot et al. (2019) provided evidence for the role of Mississippi River sediment loads in mitigating Hg bioaccumulation in GOM *L. chamaeleonticeps* collected within 50 km of the river mouth. However, *L. chamaeleonticeps* collected at greater distances (> 100 km) from the river mouth were significantly higher in Hg contamination with < 5% of collected individuals being below consumption threshold advisories (Perrot et al., 2019). In contrast, Hg concentrations measured in *L. chamaeleonticeps* collected from the SW Atlantic are sufficiently low to reduce the extent of consumption advisories recommended for fish collected from this region (White et al., 2020). Such patterns suggest that the anthropogenic deposition and sediment contamination are primary drivers of Hg bioaccumulation. However, ecological and biological characteristics of fish populations can contribute to the highly variable nature of Hg bioaccumulation that is observed among populations.

Fish growth is an important modulator of pollutant bioaccumulation with faster growing individuals typically having greater capacities for growth dilution as a mechanism to modulate Hg bioaccumulation relative to those exhibiting slower growth (Simoneau et al., 2005). Both *L. chamaeleonticeps* and *C. microps* are long-lived and sexually dimorphic, with males attaining larger body size at age relative to females (Harris et al., 2004; Palmer et al., 2004; Ross and Huntsman, 1982; Turner et al., 1983; Lombardi-Carlson et al., 2015). Growth patterns within these Tilefish populations have exhibited temporal variability as a response to fisheries harvesting practices (Harris et al., 2004; Palmer et al., 2004). For example, median size at maturity declined significantly in *L. chamaeleonticeps* between 1978–1982 following overfishing in the NW Atlantic, though age at maturity has largely since rebounded (McBride et al., 2013). Similarly, age distributions of *C. microps* population in the SW Atlantic did not differ significantly between 1980-1987 and 1996-1998 (Harris et al., 2004). However, lengths at age for fish collected in these two periods declined substantially with fishes collected during 1996–1998 being significantly smaller relative to similarly aged conspecifics collected in 1980–1987 (Harris et al., 2004). Median ages of GOM *L. chamaeleonticeps* harvested from 2001–2009 also varied significantly among these collection years with fish caught early in this time series being significantly older and larger than more recent collections (Lombardi et al., 2010). Such spatial and temporal contrasts in fish age and growth patterns have been demonstrated to influence Hg bioaccumulation among fish populations and remain important when comparing among species and populations (Simoneau et al., 2005; Li et al., 2018).

In the NW Atlantic, Tilefish demonstrate patchy distributions with propensities for high site fidelity linked to thermal and sediment preferences for burrow construction (Able et al.,1982, 1987; Grimes et al., 1986; McBride et al., 2013; Nitschke and Miller, 2016). Studies evaluating *L. chamaeleonticeps* and *C. microps* diets suggest opportunism, feeding on benthic-associated prey, typified by crustaceans, annelids, mollusks and echinoderms, with increased incorporation of larger prey, such as fishes and decapods, as individuals grow and mature (Freeman and Turner, 1977; Ross, 1982; Steimle et al., 1999). Olin et al. (2020) found evidence to support ontogenetic diet shifts based on nitrogen stable isotope values and the use of regional resource pools linked to depth preferences among species inhabiting continental shelf-edge environments. The consumption of benthic-associated prey and changes across ontogeny coupled with proximity to contaminated sediments are known pathways of exposure to environmental pollutants such as Hg and polycyclic aromatic hydrocarbons (Snyder et al., 2020; Perrot et al., 2019; White et al., 2020). Differences in the extent of sediment contamination and use of regional resource pools has the potential to contribute to the broader geographic patterns demonstrated for Tilefish Hg bioaccumulation among populations (Mason and Sheu, 2002;

Driscoll et al., 2013). However, specific factors contributing to the regional and fine scale spatial patterns of pollutant bioaccumulation within Tilefish populations are less well resolved.

In this study, *L. chamaeleonticeps* and *C. microps* were collected from the NW Atlantic to evaluate inter- and intra-specific differences in Hg bioaccumulation across a broad regional scale. The objectives were to i) quantify potential differences in Hg bioaccumulation between species; ii) evaluate sex-specific ontogenetic relationships in Hg bioaccumulation within these species and; iii) determine the extent of spatial variability in Tilefish Hg concentrations to gain a better understanding of ecological and biological factors that can influence Hg bioaccumulation by these species.

MATERIALS AND METHODS

Sample collection

Muscle samples were collected from *L. chamaeleonticeps* (*n* = 484) and *C. microps* (*n* = 64) sampled from a depth range of 75–310 m across shelf waters of the NW Atlantic Ocean, from the southern flank of Georges Bank, Cape Cod, Massachusetts to the Mid-Atlantic Bight, Cape Hatteras, North Carolina during a fishery-independent survey using a stratified random design conducted in July and August 2017 (see Frisk et al., 2018; Olin et al., 2020). Detailed survey methods are reported in Frisk et al., (2018). Briefly, Tilefish were captured using bottom-set longlines consisting of a one-nautical mile steel cable mainline equipped with 150 evenly spaced gangions baited with squid (*Illex* spp.). Biological data collected at the time of capture included fork length (cm), body mass (kg) and sex (via examination of gonads upon dissection, when feasible). Muscle samples (1–2 g) were excised from within the edible portion of the dorsal muscle filet at the time of collection, transferred into cryotubes and subsequently stored at -20ºC. For each longline deployment, depth was recorded, and regional locations were classified as Southern New England Middle Grounds (SNMG), Southern New England (SNE), and Mid-Atlantic Bight (MAB) based on latitude and longitude.

Tissue preparation and analysis

Prior to mercury and stable isotope analysis, muscle samples were placed in a dried tared weigh boat and weighed (± 0.01 g wet wt.) then oven-dried at 60°C until a consistent weight was achieved (~ 48 hrs). Dried samples were re-weighed to determine tissue moisture content (%) and then ground into a fine powder using a glass mortar and pestle. Ground muscle tissue was weighed into (0.02–0.03 g) precleaned nickel boats for total mercury analysis (THg) using a Milestone Direct Mercury Analyzer-80 (DMA-80) instrument. For quality control, a certified reference (DORM-4, National Research Council of Canada) sample was included with every ten tissue samples. Recovery (average ± SD) of the certified reference was 98.4 ± 14.0%. For comparison with literature-based results, dry weight (d.w) THg concentrations (µg/g) were converted to wet weight (w.w.) concentrations as indicated in equation 1:

$$C_{ww} = C_{dw} \cdot \left[\frac{100 - M}{100}\right]$$

where C_{ww} represents the predicted w.w. THg concentration, C_{dw} is the reported d.w. THg concentration and *M* represents the sample moisture content (%; USEPA, 2011). All glassware and utensils used to prepare samples were pre-washed in a 10% HNO_3 acid bath followed by rinsing with distilled water. Nickel boats for Hg analysis were pre-washed with soap and water followed by thorough rinsing with distilled water and subsequent combustion at 650°C for 1 hr.

To assess for differences in species' trophic positions that can influence Hg bioaccumulation (Atwell et al. 1998), all samples were processed for the stable isotope of nitrogen ($\delta^{15}N$). Briefly, between 0.48–0.58 µg of ground muscle tissue was weighed into 8 x 5 mm tin capsules and relative abundances of nitrogen ($^{15}N/^{14}N$) were determined on a Thermo Finnigan Delta V Plus mass spectrometer (Thermo Finnigan, San Jose, California, USA) coupled with an elemental analyzer (Costech, Valencia, California, USA). The results are expressed in standard delta notation (δ), defined as parts per thousand (‰) as indicated in equation 2:

$$\delta = [(R_{Sample}/R_{Standard}) - 1] \times 10^3$$

where R is the ratio of heavy to light isotope in the sample and standard, respectively (Coplen, 2011). For quality control, reference samples (acetanilide, Bass protein) were included with every ten tissue samples and the standard deviations ranged from 0.11–0.12‰ for $\delta^{15}N$ (see Olin et al., 2020 for full details).

Data analyses

All statistical analyses were performed in R (version 3.4.1, R Development Core Team, 2018) within the RStudio interface (version 1.0.136, R Studio Team, 2018). The level of significance (α) was set at 0.05. Data normality and homoscedasticity were assessed prior to statistical analysis through visual inspection of probability plots and through Shapiro-Wilk and Levene tests. For fork length, body mass and THg concentrations (d.w. and w.w.), $\log_{10}$-transformation was used to meet assumptions of normality and stabilize variance. For $\delta^{15}N$ results, no transformations were necessary.

Analysis of variance (ANOVA) was used to complete all pairwise comparisons, with body mass and fork length covariates included where appropriate. Linear or non-linear regressions were used to describe the relationships between body mass or fork length with Hg concentrations between species and sexes with coefficients of determination (R^2) used to determine the best fit of linear and non-linear regression formats to the data. Regression analysis was also used to estimate the relationships between THg concentrations and the stable isotope of nitrogen ($\delta^{15}N$). Analysis of covariance (ANCOVA) was used to evaluate for significant differences between linear regressions. Statistical comparison of non-linear regressions followed the residual sums of squares method outlined by Chen et al., (1992) with the probability calculator utility of SYSTAT 11 (SYSTAT 2004) used to estimate statistical p-values.

Regional patterns in Tilefish THg bioaccumulation among the SNMG, SNE and MAB were examined by spatially interpolating THg concentrations of both species using the empirical Bayesian kriging (Pilz and Spöck, 2008) routine in ArcGIS (version 10.4.3). Bayesian kriging approaches generate semi-variograms using sample subsets and more accurately interpolate and calculate error estimates for datasets with small sample sizes (Ceriani et al., 2014). Such approaches are useful to examine distinct spatial patterns occurring in any measured environmental and/or biological parameter (McMahon et al., 2013; Ceriani et al., 2014; Olin et al., 2020). Due to differences in length and mass among fishes sampled from each region, THg concentration data were size standardized for inclusion in the geospatial analysis. For each station location within a region, a point estimate representing the average size standardized THg concentration for all individuals sampled from a single longline set was included in the geospatial model. The resulting spatial contour maps were used to characterize regional patterns of THg bioaccumulation based on fine scale variation of THg values across station locations.

RESULTS

A total of 548 individuals including 484 *L. chamaeleonticeps* and 64 *C. microps* (Table 1) were evaluated in this study. All *L. chamaeleonticeps* were caught from depths > 90 m with *C. microps* caught from depths between 75–132 m (Table 1). For *L. chamaeleonticeps*, the female: male ratio was 45:55 with 48 individuals being of unknown sex. For *C. microps*, the female: male ratio was 55:45. For both species, males were consistently of greater length and mass relative to females (Table 1) but these differences were only significant for *C. microps*, (fork length: ANOVA; $F_{1,62} = 20.3$; $p < 0.001$; body mass: ANOVA; $F_{1,62} = 6.5$; $p < 0.013$). *L.*

chamaeleonticeps of unknown sex were shorter (ANOVA; $F_{2,482} = 17.7$, $p < 0 .001$) and lighter (ANOVA; $F_{2,482} = 100.8$, $p < 0.001$) relative to males (Table 1). However, in comparison to females, *L. chamaeleonticeps* of unknown sex were similar in length ($p = 0.223$), but of lower mass ($p < 0.001$; Table 1).

Dry weight THg concentrations ranged from 0.02 to 6.5 μg/g in *L. chamaeleonticeps* and from 0.1 to 4.4 μg/g in *C. microps* (Table 1). Average THg concentrations for *L. chamaeleonticeps* (mean ± SD; 0.4 ± 0.4 μg/g d.w.) were lower relative to average concentrations measured in *C. microps* (1.1 ± 0.7 μg/g d.w.). No significant differences in THg concentrations were evident between sexes in *L. chamaeleonticeps* (ANOVA: $F_{2,479} = 2.6$; $p = 0.141$) or *C. microps* (ANOVA; $F_{1,61} = 3.8$; $p = 0.055$).

Estimated wet weight THg concentrations ranged from 0.01–1.4 μg/g among sampled fishes with the highest average concentration determined for *C. microps* (mean ± SD; 0.3 ± 0.2 μg/g; Table 1). *L. chamaeleonticeps* of unknown sex had the lowest average THg concentration and comparison of THg concentrations against the U.S. Environmental Protection Agency's (USEPA) regulatory criteria indicated that only 1.6% of all sampled fish exceeded the most restrictive consumption threshold ('Choices to Avoid'; THg > 0.46 ug/g) based on w.w. concentrations (USEPA, 2020). Among *L. chamaeleonticeps*, 93.0% of the sampled fish had THg concentrations that categorized them in the USEPA's least restrictive group for human consumption ('Best Choice'; THg < 0.15 ug/g w.w.; USEPA, 2000). For *C. microps*, 51.6 % of sampled fish had THg concentrations that placed them into the second most restrictive consumption group (THg range 0.23–0.46 ug/g w.w.) suggesting one meal per week.

Linear regression analyses showed significant positive relationships between THg and fork length (Figure 1A) and body mass for both species (Figure 1B). No significant differences were determined in the relationships between THg and fork length (ANCOVA: $F_{1,544}$ = 0.10, p = 0.747) or body mass (ANCOVA: $F_{1,544}$ = 0.46, p = 0.496) between the species. There were significant differences in the relationships between THg and fork length between male and female *L. chamaeleonticeps* ($F_{3,478}$ = 9.30, p < 0.001; Figure 2A) but not significant between THg and body mass (ANCOVA: $F_{1,478}$ = 0.09, p = 0.906; Figure 2B). No significant difference was evident between the non-linear regressions describing the relationship between THg and fork length for *C. microps* ($F_{3,58}$ = 2.28, p = 0.106; Figure 2C). However, the linear regressions describing the relationships between THg and body mass did differ between male and female *C. microps* (ANCOVA: $F_{1,60}$ = 12.95, p = 0.001; Figure 2D). Significant relationships between THg and δ^{15}N were estimated for both species (Figure 3). These relationships did not differ between sexes in either species.

Regional differences in THg concentrations were observed for *L. chamaeleonticeps* (ANOVA; $F_{2,481}$ = 40.9, p < 0 .001; Figure 4A). Specifically, THg concentrations were significantly higher in individuals sampled from the MAB (mean ± SD; 1.1 ± 0.6 µg/g d.w.) compared to SNE (0.4 ± 0.2 µg/g d.w.) and SNMG (0.3 ± 0.5 µg/g d.w.; Figure 4A) regions. Regional differences in THg concentrations were observed for *C. microps*, though these differences were not statistically significant (ANOVA; $F_{1,62}$ = 1.8; p = 0.167; Figure 4B). Empirical Bayesian kriging illustrated distinct spatial patterns in THg concentrations for both species. Specifically, a north-south latitudinal gradient was observed for THg concentrations for both species (Figure 5A, B), with lower concentrations for individuals sampled from SNMG and SNE regions compared to the MAB (Figure 5A, B).

DISCUSSION

The results of this study contribute to the growing body of evidence surrounding regional variability in Hg bioaccumulation by *L. chamaeleonticeps* and *C. microps*. The THg concentrations reported here for individuals from the NW Atlantic stock are comparable to those reported for similarly sized individuals collected from the SW Atlantic stock (Sinkus et al., 2017; White et al., 2020) but notably lower than those reported for GOM *L. chamaeleonticeps* (Karimi et al., 2012; Fitzgerald and Gohlke, 2014; Perrot et al., 2019) indicating the lower bioaccumulation potential for fish from the NW Atlantic. However, even within the localized range encompassed from Cape Hatteras to Georges Bank in the current study, there was significant variability in the extent of THg bioaccumulation quantified within and among these non-migratory Tilefish species. Importantly, such regional variability observed in these sedentary species helps accentuate the possible roles of fine-scale biotic and abiotic processes that can act to regulate Hg bioaccumulation among individuals and species.

Mercury concentrations in Tilefishes from this study demonstrated a general decline from south to north with fish collected from the MAB region having higher concentrations relative to fish from SNE and SNMG regions, even following size-standardization. Perrott et al. (2019) also demonstrated regional variability in Hg bioaccumulation among GOM *L. chamaeleonticeps* with individuals collected proximate to the mouth of the Mississippi River generally exhibiting reduced Hg bioaccumulation relative to fishes collected from northeastern regions of the GOM. This pattern was attributed to sediment deposition from the Mississippi River that could reduce Hg bioavailability to the base of the GOM food-web influenced by this tributary (Perrott et al., 2019). Similar to the Mississippi River, the Hudson River in New York is responsible for large inputs of freshwater and terrestrial material (inorganic and organic dissolved/particulate matter). These Hudson River inputs account for 93% of Hg delivered to the proximate coastal environment (Balcom et al., 2008) and have the potential to influence Hg dynamics of the continental slope and shelf communities similar to the Mississippi River. Hollweg et al. (2010) demonstrated that the shelf and slope sediments consistent with Tilefish habitat are important areas of methylmercury (MeHg) production in the NW Atlantic. The extent of MeHg production rates in this region, however, are similar to those reported for regions on the SNE continental shelf (Hammerschmidt and Fitzgerald, 2006) indicating that the latitudinal gradient of Hg bioaccumulation observed for *L. chamaeleonticeps* and *C. microps* in this study cannot be solely attributed to differences in sediment contamination or potential point source contributions. Future research focused on identifying Hg source contributions to these species using emerging techniques such as Hg isotopes (*e.g.,* Perrot et al., 2019) could prove valuable for delineating mechanisms that contribute to the fine-scale variability in Hg bioaccumulation among these stocks.

Mercury bioaccumulation and biomagnification in aquatic species and food-webs represents a combination of biotic methylation and demethylation processes coupled with assimilation by primary consumers and subsequent trophic transfer (Morel et al., 1998). Both *L. chamaeleonticeps* and *C. microps* in the NW Atlantic display similar ecological niche characteristics (Olin et al., 2020); for example, these species exhibit shifts in prey preference throughout ontogeny, as well as high dietary similarity (Ross, 1982; Steimle et al., 1999). Olin et al. (2020) concluded that body size- and depth-specific patterns of resource use by *L. chamaeleonticeps* and *C. microps* are mechanisms that may help reduce competition between these sympatric species. For example, *C. microps* generally occupy slightly higher trophic positions (estimated using $\delta^{15}N$) relative to *L. chamaeleonticeps* across the sampling region encompassed in the current study (Olin et al., 2020). The availability of prey resources may also contribute to the latitudinal pattern in Hg concentrations observed in the current study. Grimes et al. (1986) observed potential fish prey (*Anthias* spp; *Helicolenus dactylopterus*; *Sebastes* spp; *Laemonema* spp.) near Tilefish burrows with greater frequency than crustaceans in the MAB region relative to more northerly regions. In marine food-webs, fish species typically occupy higher trophic positions and exhibit greater degrees of Hg contamination relative to invertebrate prey (Atwell et al., 1998). Both *L. chamaeleonticeps* and *C. microps* collected from the MAB region had higher trophic positions relative to conspecifics caught within the SNE and SNMG regions, with *C. microps* also generally having a higher trophic position relative to *L. chamaeleonticeps* (Olin et al., 2020). Thus, regional increases in fish consumption by *L. chamaeleonticeps* and *C. microps* in the MAB region relative to the SNE and SNMG regions could contribute to the latitudinal pattern in Hg concentrations observed in this study. Further, potential resource partitioning of benthic invertebrate and fish resources between *L. chamaeleonticeps* and *C. microps* could also contribute to the general trend of higher Hg concentrations in *C. microps*, especially if this species consumes proportionally more fish prey relative to *L. chamaeleonticeps.*

Mercury concentrations quantified in *L. chamaeleonticeps* and *C. microps* for this study were strongly correlated with fish length, mass and trophic position. Sinkus et al. (2017) also demonstrated significant size- and age-related Hg bioaccumulation for *C. microps* caught from the SW Atlantic with a similar relationship between Hg and body mass also described by Perrot et al. (2019) for GOM *L. chamaeleonticeps*. The concentrations of bioaccumulative pollutants such as Hg in fish tissues are a function of dietary uptake, animal growth rates, and the capacity for whole body elimination (Sijm et al., 1992). Under steady-state bioaccumulation kinetics, the competing processes of pollutant uptake and elimination become balanced within the animal's lifetime and no further age-related increases in pollutant concentration are realized once steady-state is achieved (Mackay and Paterson, 1981). This contrasts the non-steady state condition under which pollutant uptake rate exceeds that of whole-body elimination and pollutant concentrations continue to increase in the animal over its lifespan. The ontogenetic patterns of Hg bioaccumulation for *L. chamaeleonticeps* and *C microps* described in this study and those of Sinkus et al. (2017) and Perrot et al. (2019) are consistent with non-steady state bioaccumulation. In comparison, White et al. (2020) did not observe any ontogenetic relationships for Hg bioaccumulation among *L. chamaeleonticeps* sampled from the same SW

Atlantic region as Sinkus et al. (2017). The absence of any relationships between Hg concentrations and fish total length, mass or age as indicated by White et al. (2020) is representative of the steady-state condition. However, *L. chamaeleonticeps* collected by White et al. (2020) were predominantly > 50 cm fork length which generally excludes smaller juveniles for which rapid growth rates can serve to dilute the Hg mass assimilated from the diet to a much greater extent than in larger, older and more slowly growing individuals (Sijm et al., 1992). Subsequently, predictive relationships between pollutant concentrations and fish size derived in the absence of such smaller individuals will exclude the low degree of bioaccumulation typical of young rapidly growing individuals (Paterson et al., 2006). Despite this consideration, that White et al. (2020) did not observe any relationships between Hg concentrations and fish size or age for the range of individual *L. chamaeleonticeps* included in their study (~50–95 cm) continues to demonstrate that the collected individuals had achieved the steady-state condition. This pattern may emphasize the role of resource partitioning (Olin et al., 2020) in regulating bioaccumulation between *L. chamaeleonticeps* and *C microps* in the SW Atlantic. For example, diets of juvenile *L. chamaeleonticeps* ranging from 21–50 cm fork length are dominated (> 90% by volume) by echinoderm and arthropod prey (Steimle et al., 1999) with crustaceans generally representing the preferred prey, regardless of *L. chamaeleonticeps* age and size (Freeman and Turner, 1977). In contrast, Ross (1982), estimated that fish represents approximately 32% of the *C. microps* diet by volume for fishes collected from coastal South Carolina waters. For *C. microps*, diets potentially consisting of greater proportions of fish prey relative to invertebrates will represent a greater extent of dietary Hg uptake in comparison to *L. chamaeleonticeps* given the generally higher Hg concentrations achieved by marine fish species relative to invertebrate prey (Atwell et al.,1998). Such arguments remain speculative in the absence of regional gut contents data for *L. chamaeleonticeps* and *C. microps* but emphasize the need for additional information on the biotic parameters that can contribute to the regional variability in Hg bioaccumulation among Tilefish stocks.

Like the results of Sinkus et al. (2017) and White et al (2020), no significant differences in average Hg concentration were observed between male and female *L. chamaeleonticeps* and *C. microps* in our study. However, significant differences between males and females was determined for the regression describing the relationship between THg concentrations and fork length in *L. chamaeleonticeps*. In contrast, for *C. microps* the relationship between THg concentrations and body mass differed significantly between males and females which may be a factor of the limited sample size for the species in this study. Both *L. chamaeleonticeps* and *C. microps* exhibit dimorphic growth with males achieving larger size at age relative to females (Turner et al., 1983; Schmidtke, 2017). Previous research suggests that *C. microps* typically reach reproductive maturity by approximately 50 cm (Ross and Merriner, 1983; Harris et al., 2004) with dimorphism being contributed to by the increased allocation to reproductive rather than somatic growth in females (Schmidtke, 2017). Mercury offloading into eggs is not a major pathway of Hg elimination by female fishes with egg concentrations typically being much lower relative to muscle tissues (Frank et al.,1978; Niimi, 1983; Johnston et al., 2001). This characteristic, in combination with lower growth rates relative to males, describes a reduced capacity of females for somatic growth dilution of assimilated dietary Hg. It must also be noted that a smaller size distribution of *C. microps* was collected for the current study relative to *L. chamaeleonticeps*. Specifically, the absence of smaller (< 40 cm) juvenile and larger (> 80 cm) adult *C. microps* in our dataset generally excludes the fastest and slowest growing individuals, respectively, and subsequently the maximal and minimal capacities for growth dilution that can regulate the allometry of persistent pollutant bioaccumulation (Sijm and Van der Linde, 1995; Paterson et al., 2006). Inclusions of a broad range of sizes in future studies would help to resolve potential differences in Hg bioaccumulation between male and female *C. microps*.

Conclusion

Due to its nature as a global environmental pollutant, the bioaccumulation of Hg by fish species is unavoidable and with marine fisheries contributing more than two thirds to the global fish catch (FAO, 2008) consumption guidelines represent a necessary precaution to minimize human exposure risks. As a colloquialism and environmentally safe and sustainable seafood choice, Tilefish bear the stigma of the GOM *L. chamaeleonticeps* stock that regularly exceeds Hg regulatory thresholds for safe human consumption (Perrott et al., 2019). Of the fishes collected for this study, < 2% exceeded the USEPA's most restrictive guideline for human consumption due to Hg contamination (THg concentrations > 0.46 ug/g w.w.; USEPA, 2000) and were within expected ranges reported in previous studies of individuals from the NW Atlantic (*e.g.*, Karimi et al., 2012; White et al., 2020). This result emphasizes the conclusion of White et al. (2020) in that regional regulatory management of Hg contamination guidelines among Tilefish stocks would prove valuable for increasing public awareness. It is important to note that the majority of *L. chamaeleonticeps* individuals included in this study were predominantly between 30–50 cm and likely consisting of individuals 5 years old and younger (Palmer et al., 2004; Lombardi and Andrews, 2015). In contrast, fish that exceeded the 0.46 μg/g zero consumption guideline were all ≥ 72 cm. These larger individuals are likely >10–15 years old, thereby having a greater lifetime of Hg exposure and assimilation from the diet relative to smaller, younger fish. Landings data reported for *L. chamaeleonticeps* from 2002–2019 (Northeast Fisheries Science Center, 2014; Nitschke, 2020) indicate that that dominant size class in the fishery range from 45–65 cm, with low proportions of individuals >70 cm entering the market. The results of our study do demonstrate that a small proportion of individuals from the NW Atlantic *L. chamaeleonticeps* stock can still meet or exceed the recommended Hg guideline for human consumption. However, the population demographics of this fishery help to limit this risk due to the lower representation of such larger individuals in the commercial fishery. From this perspective, recent efforts to increase public awareness of such seafood provenance are anticipated to prove valuable for consumers in making safe seafood choices (e.g., www.seafoodwatch.org). Of interest, *C. microps* demonstrated a higher degree of Hg contamination relative to *L. chamaeleonticeps* with approximately 52% of individuals collected for this study exceeding the second most restrictive regulatory guideline of one meal per week (THg range 0.23–0.46 ug/g w.w.). Although these fish were of larger size relative to *L. chamaeleonticeps* sampled here, this difference in Hg bioaccumulation remained following size-standardization, emphasizing the roles of fish age and growth that must be accounted for when comparing Hg bioaccumulation among species and populations (Simoneau et al., 2005). However, this observed difference in Hg contamination between *L. chamaeleonticeps* and *C. microps* also underscores the potentially important role that ecology, specifically habitat and resource partitioning (Olin et al., 2020), may play in regulating Hg bioaccumulation between these sympatric species.

AUTHOR CONTRIBUTIONS

HR, JAO and GP contributed to conception and design of the study. HR conducted laboratory analysis. HR, JAO and GP performed the statistical analysis. HR wrote the first draft of the manuscript. All authors contributed to manuscript writing and approved the submitted version.

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CONFLICT OF INTEREST STATEMENT

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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TABLES

Table 1. Fork length and body mass of NW Atlantic Tilefish species with associated THg concentrations (dry and wet weight) and moisture content of muscle tissues separated by sex. Data are mean ± 1 SD and range.

Species	Sex	*n*	Fork Length (cm)	Body Mass (kg)	THg (µg/g d.w.)	THg (µg/g w.w.)	Moisture (%)	Depth (m)
Lopholatilus chamaeleonticeps	F	196	43.8 ± 7.7 (29.0–101.0)	1.2 ± 1.2 (0.3–13.0)	0.4 ± 0.4 (0.05–3.7)	0.09 ± 0.09 (0.01–0.9)	75.8 ± 3.8 (62.7–86.5)	136.5 ± 34.7 (93.2–289.0)
	M	240	48.0 ± 11.3 (28.0–110.0)	1.7 ± 2.0 (0.1–22.1)	0.4 ± 0.5 (0.06–6.5)	0.1 ± 0.1 (0.01–1.4)	75.1 ± 3.8 (60.0–82.3)	138.7 ± 36.8 (93.2–292.6)
	UNK	48	41.5 ± 4.4 (29.0–49.0)	0.9 ± 0.3 (0.3–1.8)	0.3 ± 0.1 (0.02–0.7)	0.06 ± 0.03 (0.01–0.2)	75.7 ± 3.1 (68.5–82.1)	141.4 ± 31.9 (104.2–267.0)
Caulolatilus microps	F	35	55.7 ± 8.0 (46.0–80.0)	2.5 ± 1.5 (1.2–7.8)	1.1 ± 0.8 (0.1–4.4)	0.3 ± 0.2 (0.03–1.1)	73.7 ± 7.3 (42.0–79.1)	94.4 ± 16.0 (74.9–131.7)
	M	29	66.8 ± 11.0 (38.0–83.0)	4.1 ± 2.1 (0.3–8.5)	1.2 ± 0.5 (0.2–2.4)	0.3 ± 0.2 (0.06–0.9)	74.0 ± 8.1 (50.0–80.0)	104.1 ± 16.8 (74.9–125.4)

FIGURES:

Figure 1. Relationships between THg (µg/g d.w.) and (A) fork length (cm) and (B) body mass (kg) for *L. chamaeleonticeps* (○; top) and *C. microps* (●; bottom regression statistics) from the NW Atlantic. Solid and dashed lines in both panels represent least squares regression lines for *L. chamaeleonticeps* and *C. microps*, respectively.

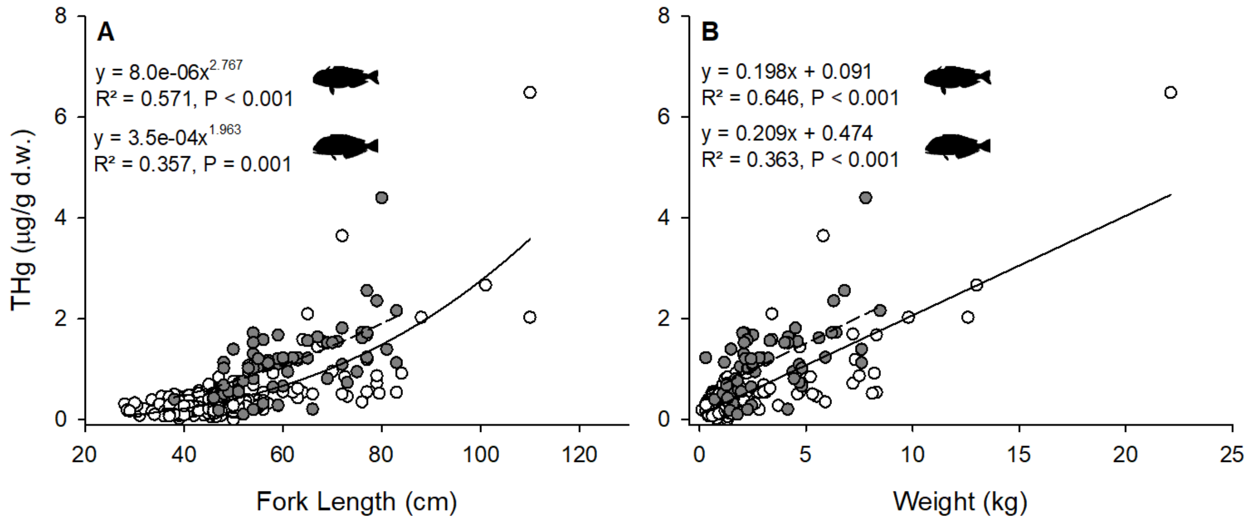


Figure 2. Relationships between THg (µg/g d.w.) and fork length (cm) or body mass (kg) for (A, B) *L. chamaeleonticeps* and (C, D) *C. microps* from the NW Atlantic. Open (○) and shaded (●) symbols indicate females and males, respectively. Solid and dashed lines in all panels represent least squares regressions for females and males, respectively.

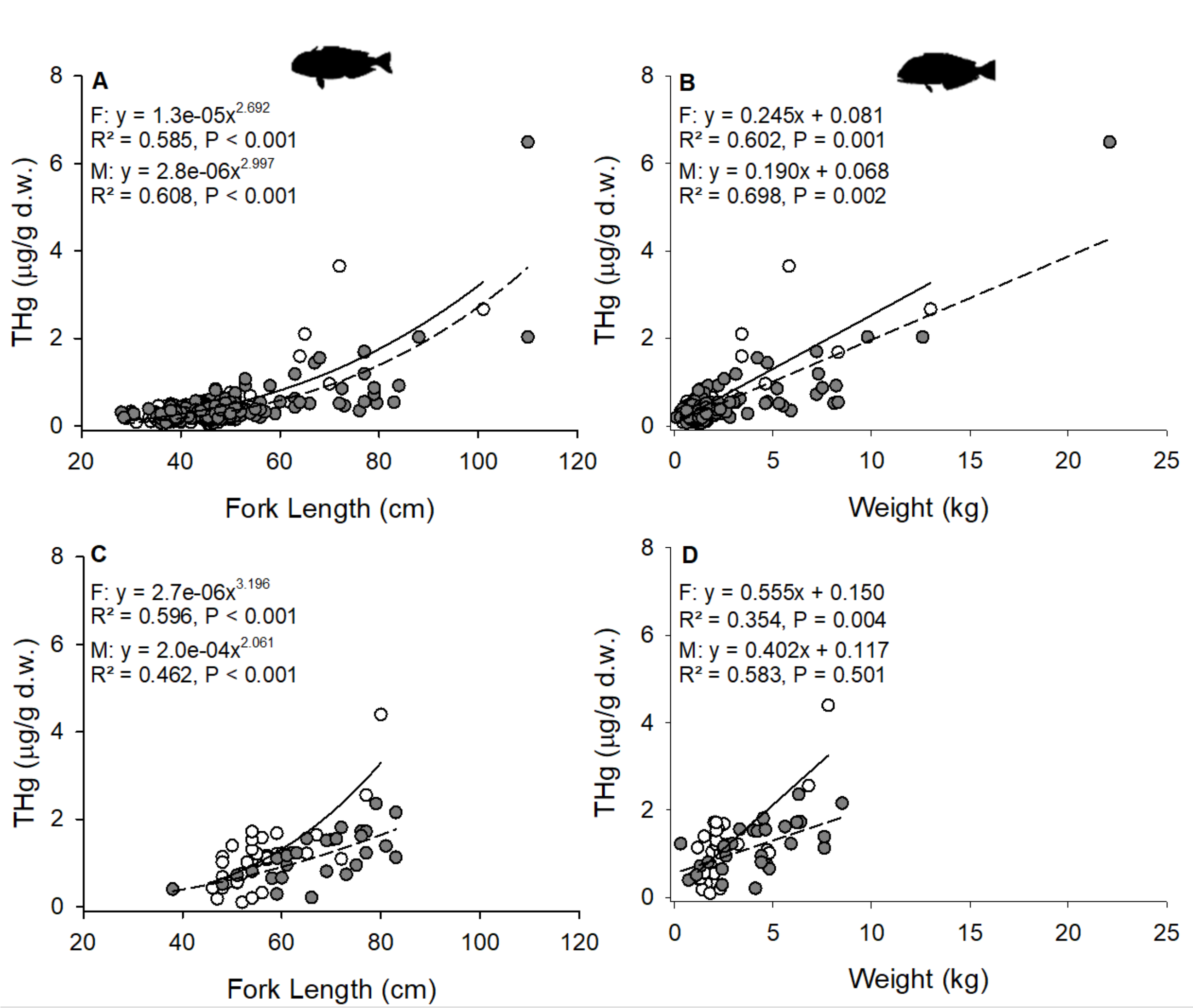


Figure 3. Relationships between THg concentration (µg/g d.w.) and nitrogen stable isotope values ($\delta^{15}N$; ‰) for *L. chamaeleonticeps* (○; top regression statistics) and *C. microps* (●; bottom regression statistics) from the NW Atlantic.

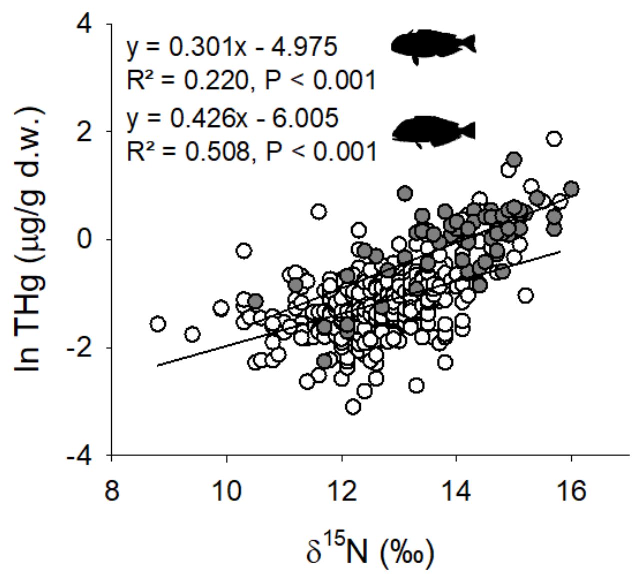
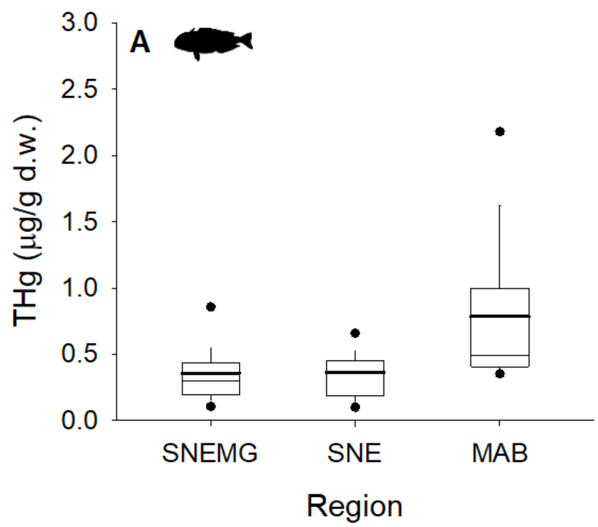


Figure 4. Size-standardized THg concentrations for (A) *L. chamaeleonticeps* and (B) *C. microps* collected from Southern New England Middle Grounds (SNMG), Southern New England (SNE) and Mid-Atlantic Bight (MAB) regions of the NW Atlantic. Individual box plots provide mean (thick line), median (thin line), 25th and 75th percentiles (box), and the 5th and 95th percentiles (●).



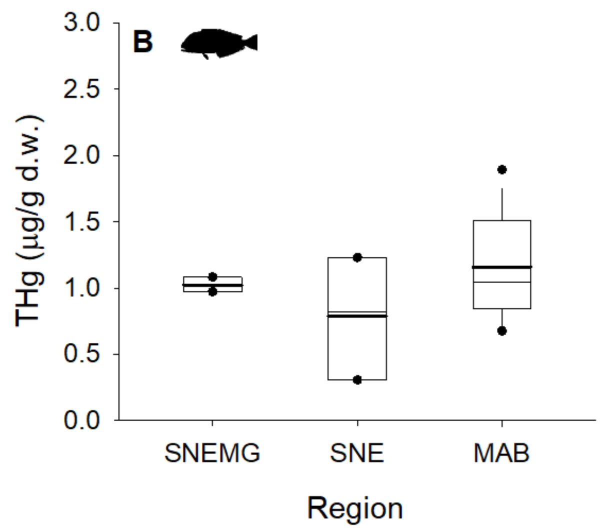


Figure 5. Spatial interpolation of size standardized THg concentrations (ug/g d.w.) in NW Atlantic (A) *L. chamaeleonticeps* and (B) *C. microps* using empirical Bayesian kriging. MAB, SNE and SNMG abbreviations indicate Mid-Atlantic Bight, Southern New England and Southern New England Middle Grounds, respectively.