



Offshore wind farm effects on flounder and gadid dietary habits and condition on the northeastern US coast

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ABSTRACT: The first offshore wind farm in North America, Block Island Wind Farm (BIWF), was built on a pilot scale (five 6 MW turbines) approximately 5 km southeast of Block Island, Rhode Island. Potential effects of the BIWF on dietary habits were examined for fish collected in a trawl survey conducted monthly over 7 years (October 2012 to September 2019). Stomach content analysis was conducted on 3457 flounder, gadids, and black sea bass collected near BIWF and in 2 reference areas during baseline, construction, and operation time periods. Other trophic metrics such as fish condition, stomach fullness, and % empty stomachs were examined for an effect of wind farm operation. Temporal variation in trophic metrics was more common than spatial differences, with no consistent indication of an effect of wind farm operation across metrics or species. Prey accumulation curves indicated that diets were adequately characterized with sample sizes of approximately 40 stomachs for most time period by area combinations. Diet composition of hakes and flounder in all areas included a greater proportion of mysids and amphipods during the wind farm operation time period. Summer flounder and winter flounder condition was lower during the operation time period in all areas. Inclusion of mussels and associated epifauna (mysids) in fish diets following turbine installation indicate fish forage on the colonized turbines. Although substantial changes to fish diets were not evident at BIWF, cumulative trophic effects of larger wind farms should be examined as the offshore wind industry expands on the northeastern US coastline.

KEY WORDS: Fish stomach contents · Trophic interactions · Flounder · Hake · Prey accumulation curve

1. INTRODUCTION

Offshore wind development on the northeastern US coast has been accelerating since the first North American offshore wind farm (OSW), Block Island Wind Farm (BIWF), went into operation in October 2016. The Bureau of Ocean Energy Management (BOEM) projects 2000 turbines will be constructed in 15 active lease areas along the eastern US coast by 2030 (BOEM 2020). Given this rapid expansion of

OSWs, it is important to understand how the introduction of turbines, sometimes with scour protection around foundations and cables, not only affects fish abundance and distribution, but also their trophic interactions. A common change to fish distributions associated with OSWs is an artificial reef effect (Andersson et al. 2009, Winter et al. 2010, Leonhard et al. 2011, van Deurs et al. 2012, Reubens et al. 2013a, Degraer et al. 2020, HDR 2020, Wilber et al. in press), that is more pronounced when wind farms are

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constructed in soft bottom areas, thus introducing hard structure and associated colonizing communities to the area, as occurred at BIWF over a mostly sandy substrate (ten Brink & Dalton 2018, HDR 2020). Because artificial reefs can result in dietary shifts in fish (Sanchez-Jerez et al. 2002, Szedlmayer & Lee 2004, Page et al. 2007), potential changes to fish diets near BIWF are a pertinent research topic, given the importance of predator–prey relationships in influencing abundances and distributions in both populations (Link et al. 2012).

BIWF is located approximately 5 km southeast of Block Island, Rhode Island and was built on a pilot scale consisting of five 6 MW wind turbine generators. Within a year of installation, the turbines were colonized by blue mussels *Mytilus edulis* upon which barnacles *Balanus* spp. had settled (HDR 2020). As observed at European wind farms (Maar et al. 2009, Coates et al. 2014, Lefaible et al. 2019), the filter-feeding community deposited fecal pellets beneath and adjacent to the turbines, creating organically enriched sediments (HDR 2020). Sediment particle size also decreased in close proximity to the turbine foundations extending to 30 m distance in association with organic enrichment and an altered macrofaunal community. The installation of the turbine foundations, therefore, initiated the transition of benthic habitat from a sandy substrate dominated by polychaetes to organically enriched sediments with mussels and associated macrofauna, such as crabs, mysids, hydroids, anemones, and sea stars (HDR 2020).

The artificial reef effects at European offshore wind farms and at BIWF are well documented; however, the effects of the fouling communities and surrounding changes to the benthic communities on higher trophic levels are less well understood. Pouting *Trisopterus luscus* that aggregate at Belgian wind farms, consume epibionts on turbine foundations that include amphipods, porcelain crabs, and mussels (Reubens et al. 2011). Other European studies have documented an increase in prey availability by a factor of 50 (Lindeboom et al. 2011) and differences in diet composition between wind farm and reference areas (Reubens et al. 2013b). Potential shifts in trophic relationships caused by wind farms on the northeastern US coast are unknown.

In this context, resource managers and stakeholders highly dependent on commercial and recreational fishing in the area were concerned about the potential effects of the construction and operation of BIWF on demersal fish, including changes to prey composition, availability, and fish condition. In particular, recreational fishermen requested analysis of

stomach contents for flounders and gadids to assess potential impacts on their prey as a component of a comprehensive demersal trawl survey that spanned baseline, construction, and operation time periods (INSPIRE Environmental 2021a, Wilber et al. in press). Examination of flounder and gadids provides information on potential impacts on diets of benthic and benthopelagic fish species. Adult winter flounder *Pseudopleuronectes americanus* diets are varied and include amphipods, annelids, cnidarians, and mollusks (Stehlik & Meise 2000, Malek et al. 2016), whereas summer flounder *Paralichthys dentatus* (Buchheister & Latour 2011) and gadids (Bowman 1984, Garrison & Link 2000) feed higher in the water column and include fish in their diets. Flounder, Atlantic cod, hake, and black sea bass *Centropristis striata* have been identified as priority species to serve as indicators in assessments of impacts of offshore wind development (Malek et al. 2014, Petruny-Parker et al. 2015, Guida et al. 2017). The before, after, control, impact (BACI) study design compared the temporal variation in dietary habits, condition, and stomach fullness in an area near the wind farm to 2 reference areas for Atlantic cod *Gadus morhua*, red hake *Urophycis chuss*, silver hake *Merluccius bilinearis*, spotted hake *U. regia*, summer flounder, and winter flounder. Additionally, adaptive monitoring was conducted by adding black sea bass to the study protocol for the final year of sampling (October 2018 to September 2019) in response to concerns from the fishing community about potential predation on juvenile American lobster *Homarus americanus*, especially near the wind farm where the introduced structure may attract both species. Inclusion of cod provides information on dietary habits for a unique biological stock that has data gaps concerning its life history in comparison to other stocks (McBride & Smebdol 2005).

Stomach content analysis provides an indication of what fish have been eating within approximately 10 h of being collected (Hyslop 1980), whereas fish condition reflects prey assimilation and energy expenditure that accrues over a longer time period. Condition indices reflect an individual's stored energy and can be assessed directly through biochemical methods or through morphometric approaches that are easier and more widely used, although a surrogate of physiological condition (McPherson et al. 2011). Prey accumulation curves created for these species are a valuable resource for informing the sampling design and intensity for future monitoring studies of wind farms being developed on the northeastern US coast.

2. MATERIALS AND METHODS

2.1. Study area

A demersal trawl survey was conducted monthly over 7 years (October 2012 to September 2019) near BIWF. Three study areas were identified; the Area of Potential Effect (APE), and 2 nearby reference areas with similar habitat characteristics (Fig. 1). Tow lines in the APE were over bottom depths that ranged from 24 to 30 m and sediments comprised primarily of silty sand, sand, and gravel. The Reference Area South (REFS) was located south of Block Island and west of the APE, separated from the APE by a ridge of glacial moraine (till, boulders, and cobbles), with tow lines stationed at bottom depths from 18 to 37 m over primarily sand and gravel. Tow lines at the Reference Area East (REFE) were at bottom depths ranging from 24 to 40 m over primarily silty sand and silt. Each month 2 replicate tows were randomly selected

in each area from 6 pre-determined tow lines that were accessible for trawling with occasional disruptions from fixed fishing gear (e.g. gill nets and lobster trawls), resulting in up to 6 tows per month. Trawling was conducted using a 412 × 12 cm, 4-seam whiting trawl with 168 cm Thyburon otter doors and a 2.54 cm knotless cod end liner to ensure retention of the smaller fish. This gear is consistent with that used by the Northeast Area Monitoring and Assessment Program (NEAMAP) surveys to improve regional comparability of catch results. Fish collected in each tow were identified, weighed, measured to the nearest mm as total length (TL) for those without a forked tail, and enumerated. For each tow, a target of 5 stomachs from each of flounders and gadids (10 stomachs total maximum) were excised and preserved in 10% neutral-buffered formalin in Whirl-Pack bags. Fish chosen for dissection were selected randomly from each tow. If less than 5 of either flounders or gadids were present in a sample, all the stom-

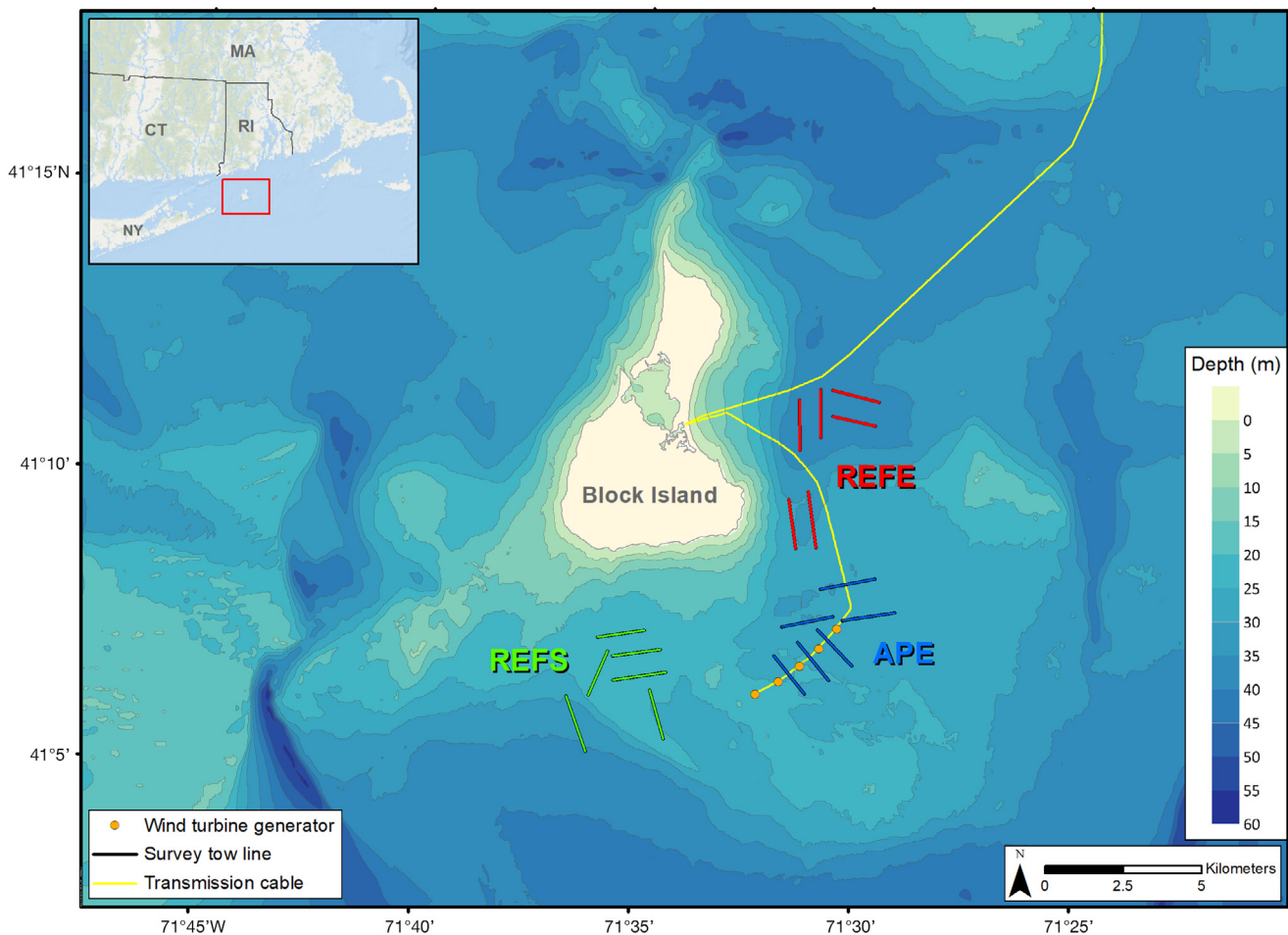


Fig. 1. Block Island Wind Farm (BIWF) demersal fish trawl study area with the locations of the 6 fixed trawl lines in each survey area; the Area of Potential Effect (APE), Reference Area South (REFS), and Reference Area East (REFE)

achs of the less numerous group were collected and stomachs from the other group were collected to make up the maximum of 10 stomachs. Empty stomachs were not collected, but noted on data sheets as dissected from either a flounder or a gadid, thus providing proportion of empty stomachs for a subset of the catch of these aggregate taxa by station. Those individuals not selected for dissection were returned to the water. In the laboratory, full stomach weights were recorded and all prey items were identified to the lowest practical identification level (LPIL) using the methods described by Weiss (1995), counted, and weighed (individually for larger-bodied prey like shrimps and anchovies, and at an aggregate level for smaller prey such as mysid shrimps and copepods).

2.2. Data analysis

Seasons were defined for the trawl survey based on monthly groupings with similar bottom water temperature variation. Fall includes October through December, winter extends from January through April, spring includes May through July, and summer includes August and September. Time periods were defined as: baseline (October 2012 through September 2014), construction phase I (July 2015 through October 2015), construction phase II (April 2016 through September 2016), and operation (October 2016 through September 2019). In the analyses for each impact phase the months for the 'baseline' period varied, i.e. October to September (2 full years) for baseline versus operation; July to October for baseline versus construction phase I; April to September for baseline versus construction phase II. Multiple disturbances occurred during each phase of construction, but broadly described: construction phase I includes wind turbine foundation construction and installation, e.g. pile driving, and construction phase II includes cable installation and turbine assembly, e.g. blade installation, jet plow trenching. Sample sizes were insufficient to test for differences in prey composition, condition, or stomach fullness between construction phases and the baseline time period, however, the prey identified during the years when construction occurred are tabulated and presented in the Supplement at www.int-res.com/articles/suppl/m683p123_suppl.pdf.

Multivariate statistical analyses on diet composition (i.e. ANOSIM, permutational multivariate analysis of variance [PERMANOVA], SIMPER, and the prey accumulation curves) were conducted using the PRIMER Version 7.0.1 computer program (Clarke et al. 2014). Linear models of stomach fullness and fish

condition were developed and evaluated in R 3.6.3 (R Core Team 2020). Results are presented with graphical representation of the data, the statistics calculated from the data, size of the effect (e.g. correlation coefficients, ANOSIM R values, and GLM coefficients), degrees of freedom, and p-values.

2.2.1. Prey composition

Monthly diet data for Atlantic cod (Table S1), red hake (Table S2), silver hake (Table S3), spotted hake (Table S4), summer flounder (Table S5), and winter flounder (Table S6) were obtained from stomach contents for all 7 years of sampling. Crabs (*Cancer* sp.) were commonly encountered in the diet of black sea bass (Table S7), and Pearson's linear correlation was used to compare black sea bass TL to the carapace width (CW) of crabs that were consumed whole. Haddock *Melanogrammus aeglefinus* (Table S8), windowpane *Scophthalmus aquosus* (Table S9), and yellowtail flounder *Pleuronectes ferruginea* (Table S10) were collected too infrequently to analyze stomach content data statistically. Prior to analyses, prey were aggregated into higher taxonomic levels, typically order or class, to prevent false distinctions in their functional value. One prey category, 'other crustacea', includes less-related taxa (e.g. mysids, stomatopods, and barnacles) that were not members of the decapod, amphipod, or isopod prey categories, as well as hermit crabs, a decapod that inhabits gastropod shells. Consistent with protocols used on regional fishery independent surveys such as NEAMAP, mean proportional weight contributions were evaluated as the dietary metric, increasing the comparability of the analysis with ongoing data collection efforts. Prey composition was calculated separately for each predator species as the mean proportional contribution (W_k) of each prey item (Buckel et al. 1999, Bonzek et al. 2009) by month and area, where:

$$\%W_k = \frac{\sum_{i=1}^n M_i q_{ik}}{\sum_{i=1}^n M_i} \times 100 \quad (1)$$

$$q_{ik} = \frac{w_{ik}}{w_i}$$

and where n is the total number of trawls that collected the predator fish species of interest, M_i is the sample size (counts) of that predator species in trawl i , w_i is the total weight of all prey items in the stomachs of all fish analyzed from trawl i , and w_{ik} is the total weight of prey type k in these stomachs.

Mean proportional contributions were calculated using only trawls in which at least 4 stomachs of the predator species were collected to avoid a potential bias from outlier individuals in trawls with smaller sample sizes. Potential seasonal differences in prey composition were explored for each predator species with multivariate techniques, i.e. ANOSIM and SIMPER using samples collected during the baseline and operation time periods. ANOSIM and PERMANOVA are non-parametric simulation and permutation tests that do not require assumptions of normality for the underlying data. Seasons with substantial dietary overlap were grouped in subsequent analyses, as defined by ANOSIM $R < 0.25$ (Creque & Czesny 2012). The mean proportional contributions of each prey item were square-root transformed prior to creating Bray-Curtis similarity matrices to increase the contribution of less common prey items to the analyses.

PERMANOVA (Anderson et al. 2008) were conducted to determine whether the taxonomic composition of prey assemblages differed by the BACI contrast or by either the time period (baseline vs. operation) or area (APE, REFE, REFS) main effects, followed by SIMPER analysis to identify which prey items were most responsible for observed differences. For black sea bass, an ANOSIM test was used to examine potential differences in diet composition among areas for 2019, the single year of sampling available for this species. For each of the other predator species, a PERMANOVA test was conducted on replicate monthly weighted prey contributions using the seasonal grouping of similar diet composition (described above). Because of the unbalanced distributions of individual hake species and the high degree of dietary overlap (Link et al. 2012), diets of red hake, silver hake, and spotted hake were examined collectively. These tests used the monthly mean proportional contribution (W_k) of each prey item for each area as the dependent variable. Data were square-root transformed to increase the contribution of the less abundant taxa to the analyses prior to creating Bray-Curtis similarity matrices. Ontogenetic shifts in diet were not considered to be a potential confounding factor in BACI analyses because of the similarity in fish sizes for those fish processed for dietary analyses (Fig. 2). For instance, the vast majority of Atlantic cod and black sea bass examined in this study were adults (>30 and >19 cm TL, respectively) and the shift to nearly exclusive piscivory

in hakes occurs at >35 cm TL (Bowman 1984), which is larger than most hake examined (Fig. 2). Large (>37.5 cm TL) summer flounder transition from a predominantly crustacean diet to one dominated by fish (Buchheister & Latour 2011) and similarly, large (>30 cm TL) winter flounder have a varied yet similar diet (Stehlik & Meise 2000). Larger sample sizes of winter flounder allowed separate PERMANOVA tests of diet composition in the fall and spring. Winter flounder have been reported to reduce (Levings 1974) or cease feeding during the winter spawning season (Van Guelpen & Davis 1979, Martell & McClelland 1994, Fairchild 2017). The spring is a critical recovery time period in which foraging resumes after spawning and depleted energy reserves are restored (Burton 1994, Wilber et al. 2013).

2.2.2. Stomach fullness and empty stomachs

A stomach fullness index (FI) was calculated as the difference between full and empty stomach weights, the total weight of food (FW), expressed as a percentage of the total fish weight according to a formula defined by Hureau (1969) as cited by Ouakka et al. (2017).

$$FI = FW / \text{fish weight} \times 100 \quad (2)$$

The fullness indices are continuous proportions (a continuous variable bounded by 0 and 1), so the statistical significance of the temporal, spatial, and interaction effects were estimated for each species using generalized linear models (GLMs) with a beta distribution and a logit link. A beta-logit regression

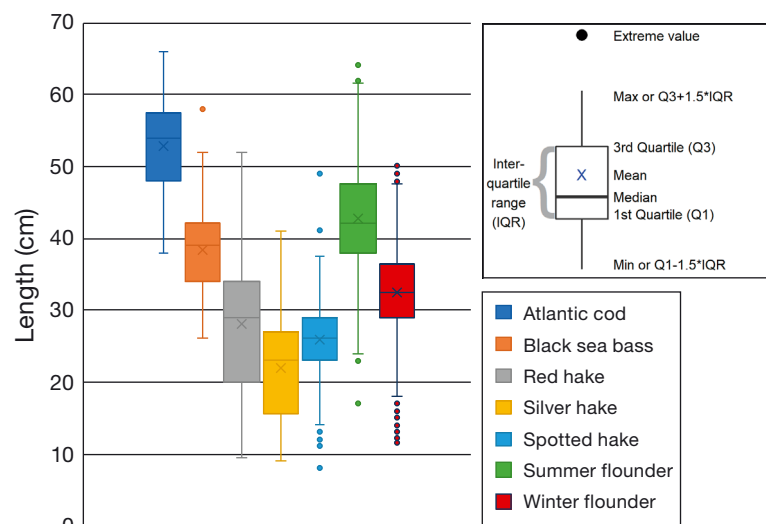


Fig. 2. Length distributions of fish used in dietary analyses and prey accumulation curves

was used to statistically model stomach fullness data as a function of time period, area, and an interaction term for each reference area. Diagnostic plots for the model residuals confirmed homogeneity of variances and lack of influential outliers. Stomach fullness data for Atlantic cod, red hake, and spotted hake were insufficient or too unbalanced among time period by area combinations to permit statistical analyses. Stomach fullness data were sufficient for silver hake (fall, winter, and spring), summer flounder (spring, summer, and fall), and winter flounder (fall). The percentage of empty stomachs was calculated by dividing the number of flounder and gadids with empty stomachs by the respective total number dissected for each area during each sampling period. The mean percentage of empty stomachs was plotted for season by area combinations across the 7 year study period to examine seasonal and interannual trends.

2.2.3. Prey accumulation curves

Prey accumulation curves were created to determine the minimum number of stomachs needed to characterize prey composition at the aggregate taxonomic level for the time period by area combinations. The aggregated prey level was used because creating these curves with LPIL data could be misleading given that the same prey item may be recorded multiple ways due to its condition at the time of identification. For instance, *Cancer irroratus*, *Cancer* sp., and Decapoda are possible LPILs for the same prey, which creates false diversity. Additionally, multivariate analyses of prey composition between baseline and operation time periods were conducted at the aggregate prey level, therefore it is appropriate to assess how well diets were characterized at this level. Prey accumulation curves were created for each combination of time period \times area for hakes, summer flounder, and winter flounder. Prey accumulation curves were created by area for black sea bass for the final year of sampling. Due to lower sample sizes, the Atlantic cod prey accumulation curve was created using all data collected across all time periods and areas.

The cumulative number of unique prey items was plotted against the cumulative number of stomachs examined. The point at which the curve reached an asymptote, i.e. new prey items were not added with the examination of more stomachs, indicates the minimum stomach sample size needed to characterize the prey assemblage (Cortés 1997, Chipps & Garvey 2006, Ramirez-Lunz et al. 2008). For all species, data were aggregated across all seasons prior to analyses

because seasonal dissimilarities in prey composition at the aggregate prey level were minor. A potential influence on the prey accumulation curves of ontogenetic changes in diets was not examined because of the similarity in fish sizes for those fish processed for dietary analyses (Fig. 2). Samples (stomach contents of individual fish) were randomized over 9999 permutations to produce an averaged curve for each predator species for each time period by area combination.

2.2.4. Fish condition

Fish condition indices were calculated as the residuals from the \log_{10} - \log_{10} regressions of mass (kg) to length (cm) (Le Cren 1951, Jakob et al. 1996) and were calculated separately for each species with sufficient data (i.e. at least 10 individuals in each area for each time period). Each regression analysis was conducted using all fish examined during the baseline and operation time periods, excluding seasons with unbalanced sample sizes across areas and time periods. Individual fish condition, therefore, is relative to that of other individuals of the same species collected during these time periods across seasons. Separate 2-factor ANOVA tests were used to test for differences in the condition of silver hake, summer flounder, and winter flounder by the area and time period factors. Residual diagnostic plots were used to verify the ANOVA assumptions, including normality of the residuals to validate interpretation of the p-values, an absence of influential outliers, and equality of variances across groups. The extent to which the residuals deviated from these ANOVA assumptions, and the influence of any deviations, are discussed in Section 3. Summer flounder tests included data from the spring, summer, and fall, whereas silver hake tests included data from fall, winter, and spring. The winter flounder test was conducted using only the fish collected in the fall to avoid potential confounding effects of variation related to winter spawning and subsequent recovery in the spring. A box plot of female winter flounder condition was created to examine seasonal and spatial variation that may reflect spawning activity.

3. RESULTS

The stomachs of 3457 fish were analyzed to determine the diets of Atlantic cod ($n = 98$), black sea bass ($n = 110$), red hake ($n = 168$), silver hake ($n = 624$), spotted hake ($n = 392$), summer flounder ($n = 435$),

and winter flounder (n = 1630). A complete list of stomach contents identified to LPIL and the frequency of occurrence by year of each prey item for each species is provided in the Supplement.

3.1. Prey composition

Atlantic cod diets were dominated by fish and crabs (Fig. 3). Black sea bass diets were dominated by decapods (crabs), polychaetes, and other crustacea (e.g. mysids and hermit crabs), while American lobster was not identified in any stomach contents. Crabs (*Cancer* sp.) that were consumed whole by black sea bass averaged 31 mm CW (range = 5 to 62 mm CW, n = 29) and fish size had a strong positive correlation with crab size (Pearson's $r = 0.63$, $df = 27$, $p < 0.001$). Red hake diets were comprised primarily of other crustacea (mysids), amphipods (Gammariidae and Aoridae), decapods (shrimp and crabs), and fish. Silver hake diets included primarily fish, other crustacea (mysids), amphipods (Ampeliscidae, Gammaridae, and Aoridae), and decapods (shrimp). Spotted hake diets were comprised primarily by amphipods, other crustacea (mysids), fish, and decapods (shrimp and crabs). Summer flounder diets were comprised primarily by fish, other crustacea, and decapods (crabs). Winter flounder had the most diverse diet of any fish examined (Fig. 3), with approximately 60% of the prey composition comprised of amphipods and polychaete worms. There were no

strong seasonal differences in prey composition at the aggregate taxon level for any species examined (all ANOSIM R values < 0.2).

Atlantic cod were not collected in sufficient quantities per trawl to permit comparisons of weighted prey contributions among areas, time periods, or seasons. For instance, only 7 of the 21 trawls that captured Atlantic cod collected 4 or more cod. Black sea bass diets (sampled in year 7 only) were strongly similar among areas (ANOSIM $R = 0.02$, $df = 2$ and 12 , $p = 0.540$). The diets of red hake, silver hake, and spotted hake were similar for each time period by area combination (Fig. 4) and when analyzed together, had mild spatial-temporal similarities by the BACI interaction term (PERMANOVA, $F = 1.75$, $df = 2$ and 167 , $p = 0.114$) and strong similarities across areas (PERMANOVA, $F = 0.60$, $df = 2$ and 167 , $p = 0.713$). Across the 3 areas, hake diets differed strongly between time periods (PERMANOVA, $F = 20.14$, $df = 1$ and 167 , $p < 0.001$), with higher contributions of other crustacea (primarily mysids) and amphipods during the operation time period and higher contributions of fish and decapods during the baseline time period (SIMPER). Summer flounder diet composition had strong similarities by the BACI interaction term (PERMANOVA, $F = 0.72$, $df = 2$ and 93 , $p = 0.661$) and among areas (PERMANOVA, $F = 0.69$, $df = 2$ and 93 , $p = 0.682$), with less similarity between time periods (PERMANOVA, $F = 1.60$, $df = 1$ and 93 , $p = 0.163$; Fig. 4).

For winter flounder sampled in the fall, the BACI contrast indicated strong spatial-temporal dietary similarities (PERMANOVA, $F = 0.80$, $df = 2$ and 38 , $p = 0.684$), and mild temporal differences across all areas (PERMANOVA, $F = 2.46$, $df = 1$ and 38 , $p = 0.016$), reflecting a higher baseline contribution of amphipods and sea anemones and a higher operation time period contribution of other crustacea (mysids) and annelid worms (SIMPER). In the spring, winter flounder prey assemblage composition differed mildly by time period (PERMANOVA, $F = 2.42$, $df = 1$ and 47 , $p = 0.037$) and more strongly by area (PERMANOVA, $F = 2.89$, $df = 2$ and 47 , $p = 0.006$), with some spatial-temporal similarities by the BACI contrast ($F = 1.42$, $df = 2$ and 47 , $p = 0.161$; Fig. 4). Similar to the fall, the contribution of amphipods was higher and the annelid worm contribution was lower during the baseline time period (SIMPER). Other taxa con-

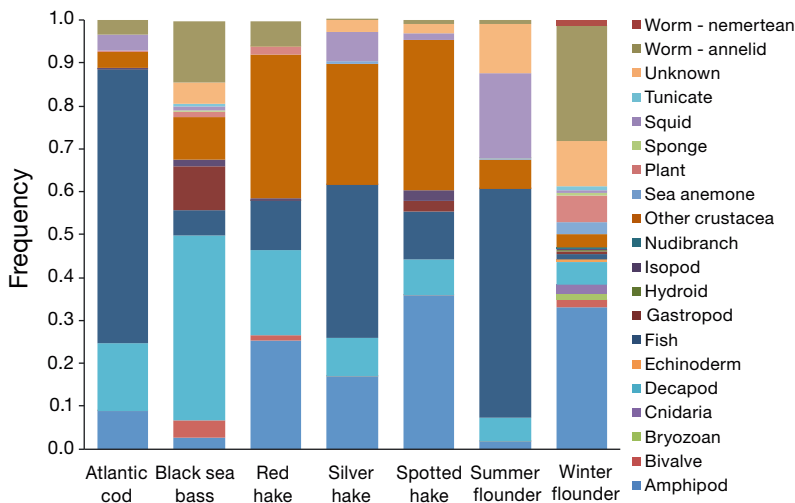


Fig. 3. Relative frequencies of the overall (7 years) diet composition based on weighted prey contributions for Atlantic cod *Gadus morhua*, black sea bass *Centropristis striata*, red hake *Urophycis chuss*, silver hake *Merluccius bilinearis*, spotted hake *U. regia*, summer flounder *Paralichthys dentatus*, and winter flounder *Pseudopleuronectes americanus*. Black sea bass were collected in the final year of sampling only. All dietary data are provided in the Supplement

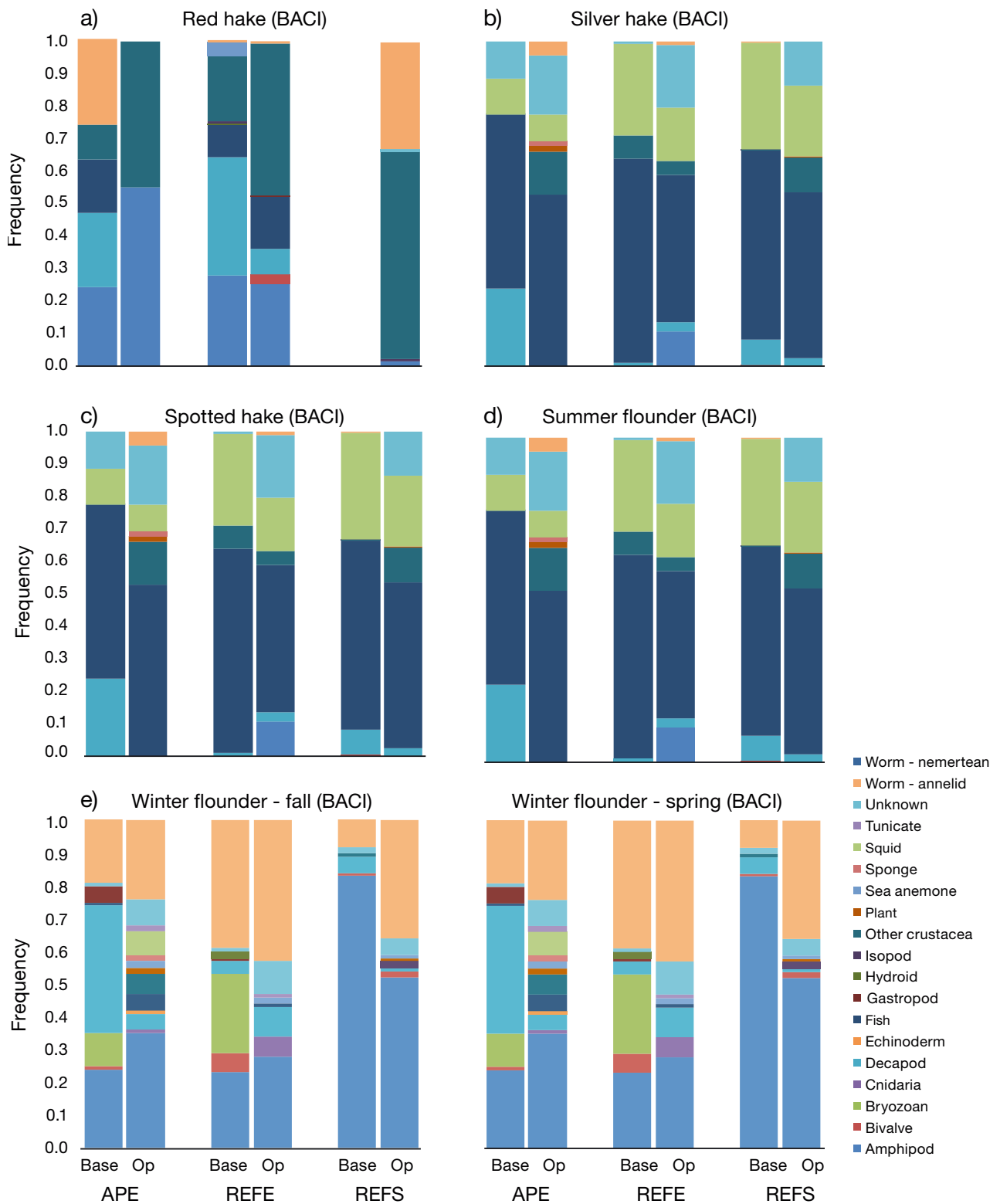


Fig. 4. Relative frequencies of the overall diet composition based on weighted prey contributions for (a) red hake *Urophycis chuss*, (b) silver hake *Merluccius bilinearis*, (c) spotted hake *U. regia*, (d) summer flounder *Paralichthys dentatus*, and (e) winter flounder *Pseudopleuronectes americanus* in fall and (f) spring for baseline and operation (Op) time periods. APE: Area of Potential Effect; REFS: Reference Area South; REFE: Reference Area East; BACI: before, after, control, impact

tributing to the time period differences were higher decapods and bryozoans during the baseline time period. The area difference reflects dissimilar prey assemblages at REFS compared to APE (PERMANOVA pairwise $t = 2.03$, $df = 1$ and 47 , $p = 0.004$) and REFE (PERMANOVA pairwise $t = 2.01$, $df = 1$ and 47 , $p = 0.007$). Prey assemblage differences among areas included higher contributions of annelids at REFE, amphipods at REFS, and decapods at APE (SIMPER).

3.2. Stomach fullness and empty stomachs

Silver hake stomach fullness at APE had a very small spatial-temporal interaction term for the REFE (GLM coefficient = -0.09 , $Z = -0.40$, residual $df = 508$, $p = 0.69$) and REFS (GLM coefficient = -0.02 , $Z = -0.08$, residual $df = 508$, $p = 0.94$). Likewise, summer flounder stomach fullness at APE had a very small spatial-temporal interaction term for the REFE (GLM coefficient = 0.04 , $Z = 0.18$, residual $df = 317$, $p = 0.86$) and REFS (GLM coefficient = 0.03 , $Z = 0.13$, residual $df = 317$, $p = 0.90$). Winter flounder stomach fullness for fish collected in the fall had a moderate spatial-temporal interaction term for REFE (GLM coefficient = 0.42 , $Z = 2.27$, residual $df = 423$, $p = 0.023$), with a decrease occurring at APE between time periods and a slight increase at REFE (Fig. 5).

Seasonal variation in the percent of empty flounder stomachs was evident, with higher percentages in the winter and lower percentages in the spring (Fig. 6). Although the empty stomachs were tallied onboard at the flounder taxon level, winter flounder accounted for 79% of the flounder stomachs examined; therefore, it is likely that the temporal fluctuations in empty stomachs are more influenced by this species. Seasonal fluctuations in gadid empty stomach percentages were not as evident. Empty stomach percentages did not appear to vary temporally in ways that differed among areas for either flounder or gadids (Fig. 6).

3.3. Prey accumulation curves

Prey accumulation curves for black sea bass reached an asymptote at APE by a sample size of approximately 30 stomachs (all collected in the 2018–2019 sampling), whereas in both refer-

ence areas, the asymptote had not been reached at this level of sampling intensity (Fig. 7). Larger sample sizes were needed before hake prey accumulation curves approached an asymptote for time period by area combinations, and the diets of hake collected at REFS during the baseline time period were not well characterized by the 46 stomachs analyzed (Fig. 7). Summer flounder prey accumulation curves reached an asymptote by 40 stomachs for some time

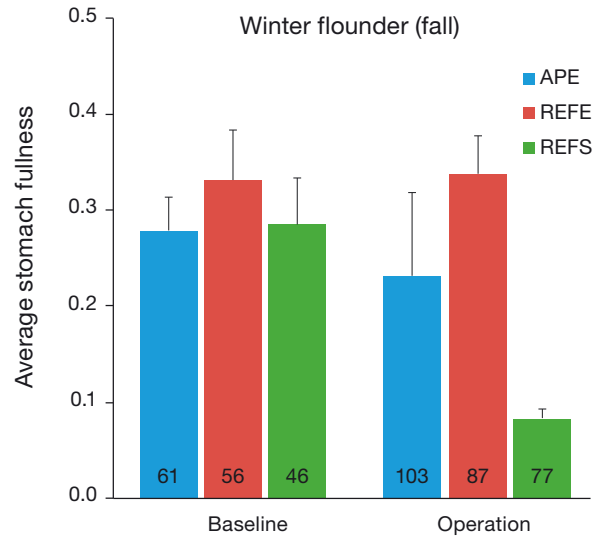


Fig. 5. Average stomach fullness indices (+SE) by time period and area for winter flounder *Pseudopleuronectes americanus*. Sample sizes are given at the bottom of each bar. APE: Area of Potential Effect; REFS: Reference Area South; REFE: Reference Area East

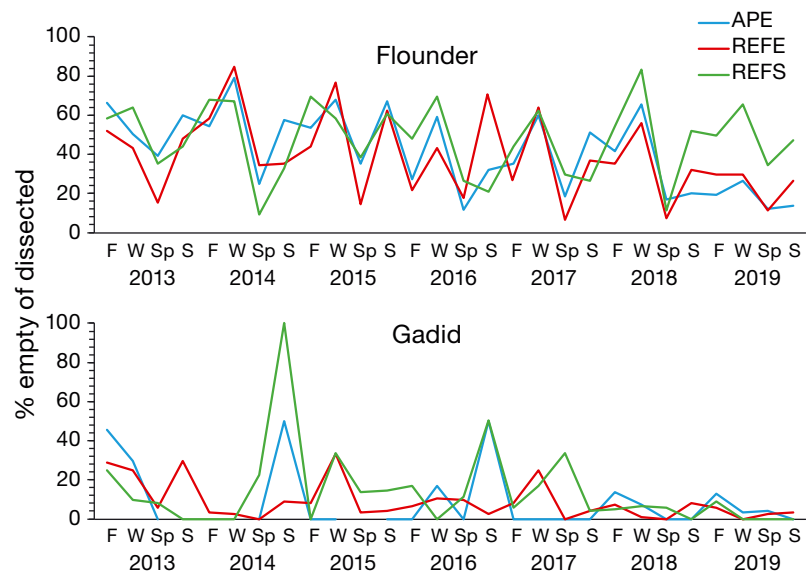


Fig. 6. Mean percentage of empty stomachs of the total number of stomachs dissected for (a) flounder and (b) gadids. Labels on the x-axis represent sampling seasons. F: fall; W: winter; Sp: spring; S: summer; APE: Area of Potential Effect; REFS: Reference Area South; REFE: Reference Area East

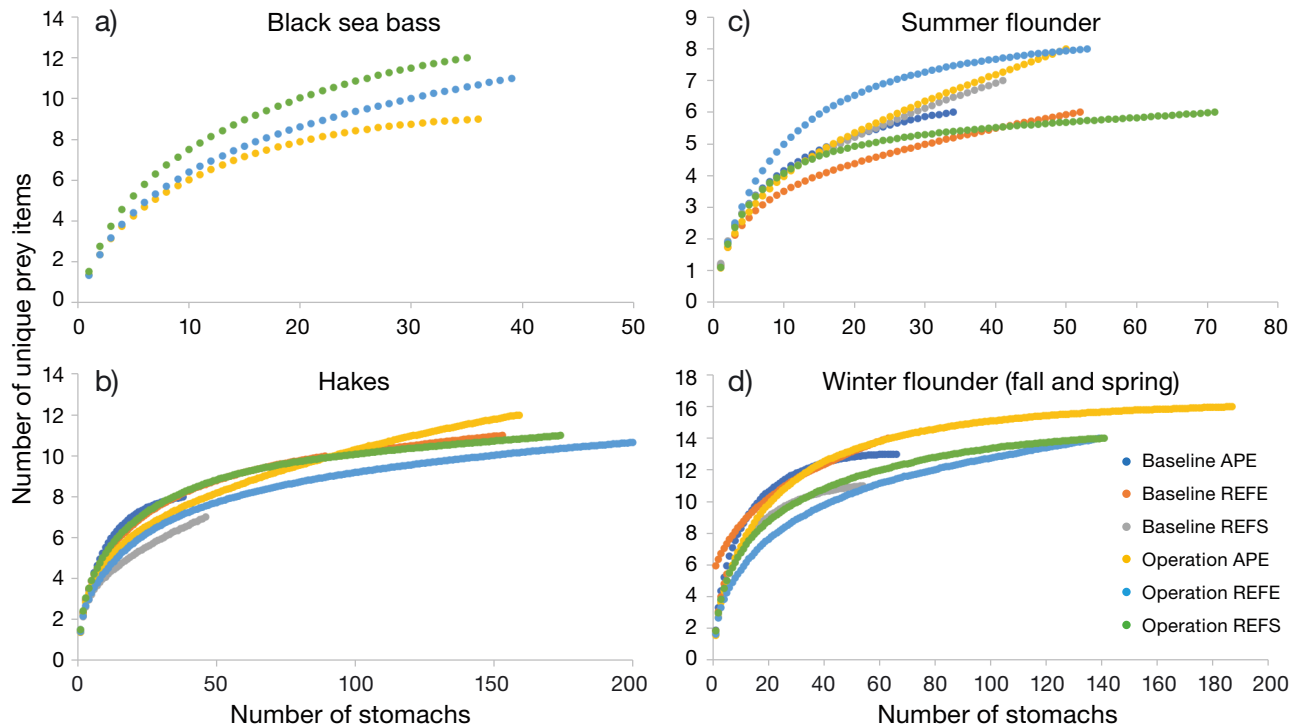


Fig. 7. Prey accumulation curves for (a) black sea bass *Centropristis striata*, (b) hakes, (c) summer flounder *Paralichthys dentatus*, and (d) winter flounder *Pseudopleuronectes americanus* calculated for each time period by area combination. APE: Area of Potential Effect; REFS: Reference Area South; REFE: Reference Area East

period by area combinations, whereas the diets of summer flounder collected in other time period by area combinations were not fully characterized (e.g. baseline REFS and operation APE). Prey accumulation curves for winter flounder fall and spring diets approached an asymptote by 40 samples for all time period by area combinations, with the exception of the REFE area during the baseline and the operation time periods (Fig. 7).

3.4. Fish condition

Residual diagnostics indicated that the ANOVA assumptions of approximate normality, equality of variances, and lack of outliers were met for silver hake, while summer flounder and winter flounder data sets had some deviations. The residuals from the summer flounder and winter flounder models were largely symmetric but with higher kurtosis than expected under normality with values near 15 (indicated by narrow interquartile range and more values outside the boxes in Fig. 8). These deviations are not overly problematic for the ANOVA and the p-values may be considered approximate (e.g. Knief & Forstmeier 2021).

Silver hake condition decreased in the REFS and REFE between the baseline and operation time periods, while increasing slightly at APE (Fig. 8), resulting in a moderate spatial-temporal interaction ($F = 4.16$, $df = 2$ and 509 , $p = 0.016$). Summer flounder average condition was lower during the operation time period across all areas (Fig. 8; $F = 17.44$, $df = 1$ and 318 , $p < 0.001$), with no detectable differences among areas ($F = 1.14$, $df = 2$ and 318 , $p = 0.32$) or by the interaction term ($F = 0.34$, $df = 1$ and 318 , $p = 0.72$). The condition of winter flounder collected in the fall was lower during the operation time period across all areas ($F = 11.27$, $df = 1$ and 424 , $p < 0.001$; Fig. 8), with no detectable spatial differences ($F = 1.43$, $df = 2$ and 424 , $p = 0.24$) or spatial-temporal interaction ($F = 0.08$, $df = 1$ and 424 , $p = 0.92$). Female winter flounder condition decreased in all areas in the winter, increasing to pre-spawning (fall) levels in the spring (Fig. 9).

4. DISCUSSION

The multiple metrics analyzed to assess fish trophic interactions near BIWF did not indicate a consistent effect of wind farm operation, either positive or nega-

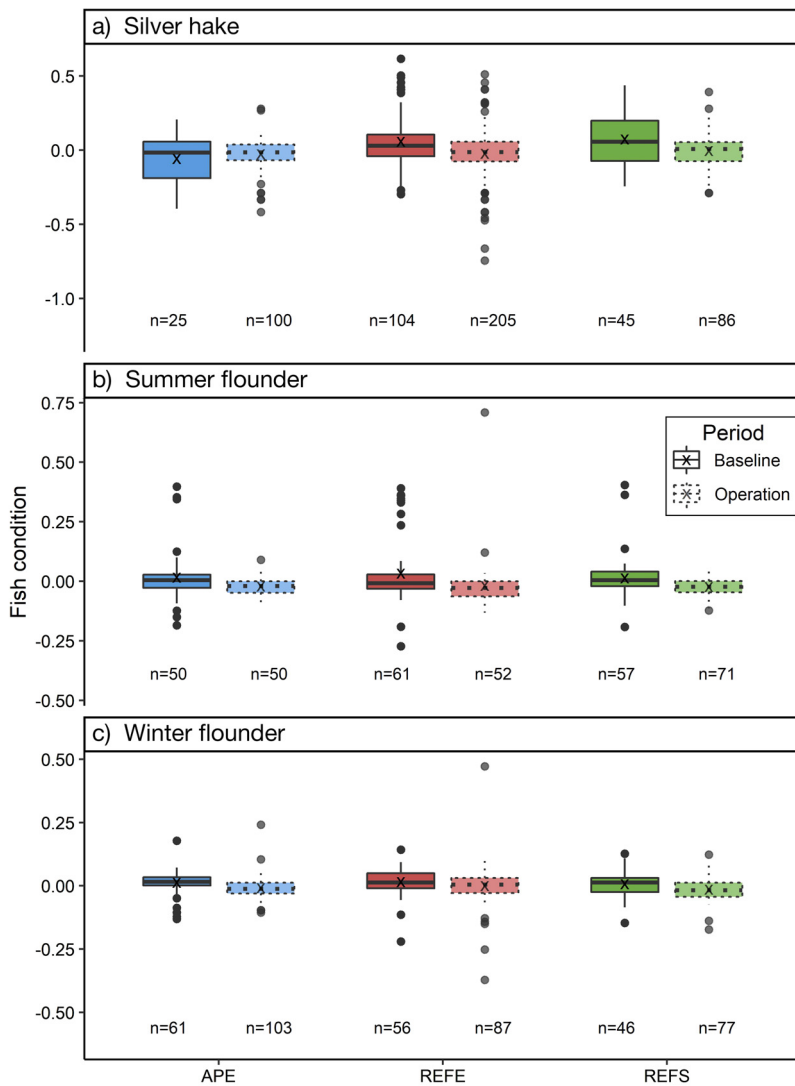


Fig. 8. Condition of (a) silver hake *Merluccius bilinearis*, (b) summer flounder *Paralichthys dentatus*, and (c) winter flounder *Pseudopleuronectes americanus* (fall) collected during the baseline and operation time periods. Box plot parameters as in Fig. 2

For instance, silver hake condition increased slightly at the wind farm between the baseline and operation time periods while decreasing at the reference areas, which is consistent with a positive wind farm effect. However, an accompanying change in silver hake diet composition only near the wind farm was not evident, indicating that temporal dietary changes for hakes occurred in all areas and spatial similarities were strong. Because fish condition reflects the net balance of energy consumed and expended, it is possible that better foraging efficiency near the wind farm where prey may be more concentrated may have contributed to higher silver hake condition (sensu Schwartzbach et al. 2020). Summer

flounder and winter flounder condition, however, was lower during the operation time period—a change that occurred across all 3 areas. There was no accompanying temporal change in diet composition for summer flounder, whereas winter flounder diets (fall and spring) shifted to a greater proportion of polychaetes and smaller proportion of amphipods during the operation time period across areas. Temporal variation in trophic metrics was more common than spatial variation, but a linkage among metrics, e.g. low stomach fullness coupled with low condition and a change in diet, was not evident for any species examined. Morphometric condition indices can be latent, acting as surrogate indicators of fish physiological status (McPherson et al. 2011); therefore, validating condition with a biochemical marker in future monitoring studies should be considered. For instance, variation in morphometric condition would be better understood if this metric was additionally correlated with other factors such as lipid stores, protein content, or stable isotope analysis (Davidson & Marshall 2010, McPherson et al. 2011, Mavraki et al. 2020). Given the high mobility of these fish, it is likely that large-scale impacts to prey resources would be necessary to elicit a consistent directional change in trophic metrics. Temporal changes to hake and winter flounder diets occurred in all areas, which may reflect regional variation in the relative availability of prey with associated variation in nutritional quality. The similarity in summer and winter flounder condition between the BIWF and reference areas is consistent with no differences in Atlantic cod and pouting condition between wind farm and reference areas in the North Sea, thus suggesting the wind farm did not provide inferior habitat (Reubens et al. 2014). Stomach fullness for winter flounder collected in the fall exhibited the only outcome consistent with a detrimental effect of BIWF operation, with a decrease in fullness between baseline and operation time periods at APE and an increase in fullness at REFE. However, even this result is equivocal because stomach fullness also decreased at REFS between time periods.

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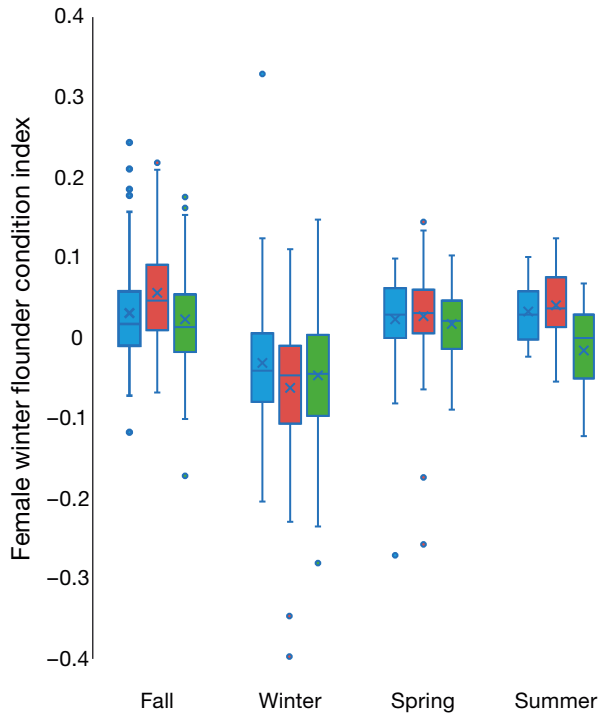


Fig. 9. Condition index for female winter flounder *Pseudopleuronectes americanus* from the Area of Potential Effect (APE, ■), Reference Area East (REFE, ■), and Reference Area South (REFS, ■) for the fall (October through December), winter (January through April), spring (May through July), and summer (August and September) for all 7 years of sampling. Box plot parameters as in Fig. 2

Although dietary shifts in prey composition attributable to BIWF operation were not evident, new prey items associated with the epifaunal community on wind turbine foundations occurred in the diets of some species across all areas following wind turbine installation. Blue mussels colonized the BIWF turbine structures and settled on the seafloor up to at least 90 m away from the turbine foundations (HDR 2020). Mussels were collected in low numbers in the BIWF demersal trawl survey prior to wind farm operation, with consistently higher collections at APE thereafter (INSPIRE Environmental 2021a). Blue mussels are a common colonizer of steel foundations of European wind turbines (Wilhelmsson & Malm 2008, Maar et al. 2009, Bouma & Lengkeek 2012), affecting the community composition of nearby hard bottom habitat. It appears a similar dispersal of blue mussels to adjacent areas occurred near BIWF, with collections at REFS and REFE by the final year of the survey (INSPIRE Environmental 2021a). Colonization of hard structures can increase biodiversity in an area and provide new prey and nursery resources (Sheehan et al. 2020). Recreational spearfishers at BIWF have noted that the

mussels covering the BIWF turbine foundations attract a diverse assemblage of foraging fish (ten Brink & Dalton 2018). Only 2 individual winter flounder had blue mussels in their stomach contents prior to turbine installation, whereas thereafter, mussels were found in the diets of Atlantic cod, haddock, red hake, and silver hake, and increased in frequency for winter flounder (see Supplement). Winter flounder are likely feeding on epibionts that colonized the turbine structures and the associated biotic assemblages. Additionally, the number of fish, particularly hake, that consumed mysids increased during the construction and operation years. This increase in mysid prey occurred for fish collected in all areas and may be related to the spread of mussel patches, which are associated with the mysid *Heteromysis formosa* near the wind farm (HDR 2020).

Diet composition observed for fish in this study was similar to other regional dietary fish studies. For instance, black sea bass, summer flounder, and winter flounder diet compositions were similar to those observed in a study that collected fish by multiple methods throughout Rhode Island state waters (Angell 2019). The diet of black sea bass shifts towards piscivory with ontogeny (Byron & Link 2010), with amphipods and mysids primarily consumed by juveniles, and crabs and fish increasingly consumed as black sea bass mature (Drohan et al. 2007). Black sea bass in the BIWF survey were adults and their stomach contents were dominated by crabs. Juvenile crabs (*Cancer irroratus*) also dominated black sea bass diets in New Jersey waters both at a reef that was fully colonized with epifauna and at a newer, uncolonized reef, suggesting they used the reefs as shelter, not foraging habitat (Steimle & Figley 1996). Lobsters were not identified in any stomach contents, which suggests lobsters were either not common in areas where black sea bass were foraging or were not present at the appropriate size class, since black sea bass feed opportunistically, consuming prey in proportion to their relative availability (Byron & Link 2010). Crabs that were consumed whole by black sea bass overlapped in size with the smallest lobsters collected in a ventless trap survey conducted at BIWF (INSPIRE Environmental 2021b). The designation of silver hake as a piscivore by Malek et al. (2016) agrees with the silver hake diet observed in this study, which was comprised primarily of fish and shrimp. Similarly, Malek et al. (2016) described winter flounder as a benthivore, which was confirmed by a diverse diet at BIWF that was comprised primarily of amphipods and polychaetes. Coupling stomach content analyses, which document short-term con-

sumption habits, with stable isotope analysis, which provides better information of assimilated prey items (Mavraki et al. 2021) may be useful in examining trophic effects in future OSW studies.

Prey accumulation curves, although not always included in fish diet studies, are essential to understanding the extent to which the sampling effort has adequately characterized the fish diet, thus affecting the reliability of conclusions. This study includes relatively large sample sizes of fish stomachs, which provide important information on using diet composition when analyzing environmental impacts associated with offshore wind development. For instance, the diet of winter flounder was best characterized in this study, perhaps because sample sizes were highest for this species and because this species was sampled consistently among areas. Asymptotes in prey accumulation curves were not reached for all time periods by area combinations for all predator species, but in cases where an asymptote was reached, a minimum of approximately 40 stomachs was needed to characterize fish diets within time period by area combinations. These prey accumulation curves can be used in the planning of future OSW monitoring efforts on the northeastern US coast. For instance, concentrating sampling effort on fewer species that represent different trophic categories (e.g. benthivore and piscivore), may be considered since sample sizes strongly influence the ability to fully characterize diets for most species. Likewise, focusing diet composition studies on species that are likely to be sampled within the study area in suitable numbers throughout most of the year will help yield representative sample sizes to accurately characterize diet.

The winter decline in female winter flounder condition and increase in percentage of flounder empty stomachs are consistent with nearshore spawning activity. Inshore winter flounder populations were once thought to spawn exclusively in estuaries and shallow embayments; however, more recently, nearshore coastal spawning has been observed in New Jersey (Wuenschel et al. 2009) and the Gulf of Maine (DeCelles & Cadrin 2010, Fairchild et al. 2013, Fairchild 2017), and the BIWF condition and empty stomach data suggest nearshore spawning activity in Rhode Island. Because winter flounder spawning success is dependent on restoring depleted energy reserves immediately following the spawning season, successful foraging in the spring and early summer is critical. Differences among areas in spring diet composition for winter flounder occurred during the baseline and operation time periods, probably reflecting spatial variation in prey availability, with no

indication of an adverse effect of BIWF operation on stomach fullness or condition.

The trophic impacts of invertebrate colonization of wind turbines and associated infrastructure varies among fish species based on their dietary habits. Wind turbines in Europe serve as foraging habitat for some fish species that feed on amphipods and decapods, such as Atlantic cod, pouting, and sculpin *Myoxocephalus scorpioides* (e.g. Reubens et al. 2011, 2014, Mavraki et al. 2021); however, some pelagic species, such as horse mackerel *Trachurus trachurus*, occur in close proximity to the turbines and continue to prey predominantly on zooplankton (Mavraki et al. 2021). Additionally, the trophic impact of OSWs on fish communities is affected by the surrounding habitat, i.e. whether the introduced hard structure and associated colonizing community are novel within a predominantly soft bottom area, as is common in the North Sea and many planned projects in the USA. Alternatively, the colonized community may be an extension of surrounding hard bottom habitat, which is a feature of several lease areas on the northeastern US coast and occurred adjacent to the southernmost wind turbine at BIWF (Guarinello & Carey 2020).

Results from BIWF indicate the diets of benthic and benthopelagic predators were influenced by the introduction of hard structure to the area, as evidenced by the higher incidence of blue mussels and mysids in stomach contents. Their overall diet composition characterized at an aggregate taxonomic level, however, did not differ from reference areas, indicating that the quality of foraging habitat near the wind farm was similar. Although the introduction of BIWF to the nearshore environment in Rhode Island did not substantially affect trophic interactions for flounder and gadids in the area, understanding the extent to which larger wind farms on the northeastern US coast may affect fish trophic relationships requires further assessments of diet composition and other trophic metrics.

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