

**CROSS-TROPHIC-LEVEL DYNAMICS IN AQUATIC ECOSYSTEMS AND THEIR
APPLICATION ACROSS ECOLOGICAL CONTEXTS**

By

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An Abstract of the Dissertation Presented
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Cross-trophic-level dynamics represent biotic interactions between organisms in a food web that span two (e.g., predator-prey interactions) or more (e.g., bottom-up or top-down indirect interactions) trophic levels. These dynamics are fundamental to understanding a variety of animal attributes across ecological contexts, including life-history traits, population limitation, and resource management. A classic example illustrates how declines in sea otter (*Enhydra lutris*) populations had negative indirect effects on giant kelp (*Macrocystis pyrifera*) populations due to reduced top-down regulation of a primary kelp predator, purple sea urchin (*Strongylocentrotus purpuratus*). The need for management approaches that incorporate this food-web connectivity is evident in the increasing adoption of ecosystem-based management approaches in place of the traditional single-species management paradigm, particularly in aquatic ecosystems. In this dissertation, I apply a unified theme—cross-trophic-level interactions in aquatic ecosystems—across ecosystem types, vertebrate classes, and animal attributes to examine both general ecological principles and applied resource management. In Chapter 1, I examine the role of a primary prey taxon in shaping a life-history trade-off in a polymorphic

amphibian population. The results of this chapter uncover sex-specific functional responses and, more broadly, highlight the importance of landscape heterogeneity in maintaining intrapopulation phenotypic diversity. In Chapter 2, I use metabarcoding of fecal DNA to describe the overwinter diet of a shorebird population that has recently experienced population declines. As the first diet study to utilize molecular approaches with this species, I provide multiple competing explanations for my novel results and illustrate the challenges of integrating initial molecular diet data into a species' known diet history based on traditional approaches. In Chapters 3* and 4, I conduct a landscape-scale Before-After Control-Impact experiment to assess the effects of commercial harvest on a basal resource (seaweed) at a whole-bed scale (Chapter 3), and whether bottom-up effects were detectable in high trophic-level consumers (birds; Chapter 4). My results indicate that the effects of commercial rockweed harvest on two common bed characteristics (biomass and height) are lesser at the scale of the rockweed bed than at smaller spatial scales that do not capture the spatial heterogeneity of harvest impacts. My findings in Chapter 4 suggest weak bottom-up indirect interactions between commercial rockweed harvest and coastal birds, and they raise questions about the use of the focal habitat relative to other habitat types in coastal birds' home ranges. Overall, my dissertation examines cross-trophic-level interactions across a diverse set of systems and illustrates how these food-web linkages can inform a variety of outcomes from life-history trade-offs to conservation planning.

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DEDICATION

For my parents, Tim and Alex. The path I have taken to get here may not have always been very clear, but thank you for encouraging me to forge ahead in the direction that most excited me.

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Chapter 1

The Role of a Primary Prey Taxon in Balancing a Life-history Trade-off

1.1. Chapter Abstract

Polymorphisms are a common source of phenotypic diversity in animal populations. This diversity is often maintained over time by trade-offs that alternatively favor morphs over space or time. In a polyphenic population of amphibians (Arizona tiger salamander; *Ambystoma mavortium nebulosum*), two morphs—paedomorphs and metamorphs—exhibit a trade-off between life-history strategy and dispersal capability. Paedomorphs remain in their natal pond and first breed at an earlier age, while metamorphs disperse between ponds of varying hydroperiods and live longer. This polyphenism is maintained in part by sex-specific reproductive advantages within each morph, with paedomorphic males achieving higher condition-weighted reproductive opportunities and metamorphic females achieving higher condition-weighted egg production. In this study, we examined the role of a high-quality, primary prey taxon (fairy shrimp)—which is accessible to the dispersal-capable morph only—in balancing the sex-specific component of this life-history trade-off. We found that both metamorph sexes required high fairy shrimp consumption to achieve the highest body condition. However, the composition of female diets was influenced much more strongly by the density of fairy shrimp biomass in ponds. These sex-specific functional responses to fairy shrimp not only provide evidence for the importance of a primary prey taxon in realizing a life-history trade-off, but they also suggest differential top-down structuring of nonpermanent pond food webs based on the adult sex ratio of metamorphs. More broadly, our findings highlight how prey resources can shape landscape heterogeneity, and how in turn this heterogeneity can maintain phenotypic trade-offs within populations.

1.2. Introduction

Polymorphisms are a common source of intrapopulation phenotypic variation that allow populations to achieve higher average fitness in heterogeneous environments than monomorphic populations (West-Eberhard 1989, Nijhout 2003, Roulin 2004). The production of different morphs within a population can result exclusively from variation in genotype (i.e., genetic polymorphism; Hedrick 1986, Roulin 2004), exclusively from environmental variation (i.e., environmental polyphenism; West-Eberhard 1989, Reed et al. 2010), or from a combination of genetic and environmental variation (Roff 1986, Matsumura 1996). Among all types of polymorphisms, there are frequently trade-offs (e.g., physiological, behavioral) between morphs that can result in different life-history strategies to maximize each morph's fitness (Zera and Denno 1997, Roulin 2004, Lackey et al. 2019). For instance, the two plumage morphs of the white-throated sparrow (*Zonotrichia albicollis*) differ in their reproductive behavior: white-striped males are promiscuous and invest in securing multiple mates at the expense of parental care, while tan-striped males are monogamous and invest more heavily in parental care (Tuttle et al. 2016). There are a number of hypothesized mechanisms for how species maintain stable polymorphisms through time (Roulin 2004). For example, there can be fitness trade-offs in which natural selection differentially favors morphs across environmental gradients (Frank and Slatkin 1990, Skúlason and Smith 1995) or morph ratios (i.e., frequency-dependent selection; Losey et al. 1997, Subramaniam & Rausher 2000).

Across a range of polymorphic taxa, fitness trade-offs occur between dispersal capability and life-history traits (Denno et al. 1980, Zera and Mole 1994, Lackey et al. 2019). In some wing-dimorphic insects, the energy used to develop wings and flight muscles precludes simultaneous investment in reproduction, and leads to delayed age at first reproduction (Zera

1984). As a result, dispersal polymorphisms can impose early-age fitness disadvantages on the more mobile morph that forgoes reproductive opportunities in favor of increased mobility. However, if dispersal-capable individuals live long enough, they may be able to recoup the initial penalties imposed by dispersal capability and achieve similar or greater lifetime fitness to that of dispersal-limited individuals (Lackey et al. 2019). For example, the dispersing morph could selectively colonize patches with rich, ephemeral food resources that contribute toward equalizing mean survival (e.g., via increased energy stores) or fecundity (e.g., via larger body size) relative to the dispersal-limited morph (Denno et al. 1980, Cook and Denno 1994, Denoël et al. 2007).

For dispersal polymorphisms to be maintained, there must be available fitness benefits for individuals that invest in mobility at the expense of short-term fitness, or presumably natural selection would favor monomorphic, dispersal-limited species. In environments where patch quality is always suitable in core patches and predictably oscillates between better and worse quality in ephemeral satellite patches, a dispersing morph can take advantage of seasonal pulses of high-quality resources and escape when conditions deteriorate (Solbreck 1978). In Atlantic coastal saltmarshes, the flightless morph of a delphacid planthopper inhabits high-marsh grasses year-round and oviposits at an earlier age than the volant morph. However, the volant morph colonizes nutritious patches of streamside grasses in the summer and achieves higher fecundity in these patches than adults that remain in the high marsh (Denno et al. 1980, 1986). There are several qualities of ephemeral patches—such as higher quality food resources (Whiteman et al. 1994, Denoël et al. 2007) or lower conspecific densities (Denno et al. 1986, Whiteman et al. 1994)—that could allow the more mobile morph to recoup initial fitness deficits through increased longevity or clutch size relative to the dispersal-limited morph.

In this study, we examine the diet of the dispersing morph in a polyphenic amphibian population to understand how food resources in ephemeral patches contribute toward a life-history trade-off. Larvae in facultatively paedomorphic populations of Arizona tiger salamanders (*Ambystoma mavortium nebulosum*) select between two discrete ontogenetic pathways based on environmental cues. Paedomorphs retain larval traits, such as gills, and remain in their natal pond as sexually mature adults, while metamorphs undergo metamorphosis, overwinter in the terrestrial environment, and disperse over land. Metamorphs are older at first reproduction on average, but they also have a longer average lifespan than paedomorphs (Lackey et al. 2019). The population at the Mexican Cut Nature Preserve (MCNP; 39.02°N, 107.06°W) in the Elk Mountains (Colorado, USA) occurs in a high-elevation (3640 m) wetland complex comprised of over fifty ponds that range from ephemeral to permanent hydroperiods (see a map of the ponds in Wissinger et al. 2003). Only metamorphs are able to move between ponds, which allows the morph to exploit resources in nonpermanent aquatic habitats that are not available to paedomorphs. Previous research suggests that the rewards are high: the caloric content of prey in metamorph guts was over three times higher in individuals captured in nonpermanent versus permanent ponds (Whiteman et al. 1994). Furthermore, this caloric difference is frequently driven by access to one nonpermanent-pond specialist prey taxon, the Colorado fairy shrimp (*Branchinecta coloradensis*; hereafter fairy shrimp; Whiteman et al. 1994, Denoël et al. 2007).

Despite this established morph-prey linkage, it remains unclear how this primary prey taxon interacts with the life-history trade-offs between the two salamander morphs. In this study, we evaluated several components of a predator-prey relationship to test the hypothesis that consumption of a high-quality ephemeral prey item (fairy shrimp) drives fitness gains accessible to dispersers (metamorphs) only. First, we quantified prey availability by assessing

spatiotemporal variation in fairy shrimp biomass across ponds and years. Second, we examined the taxonomic composition and biomass of metamorph diets in nonpermanent ponds and assessed how diets varied across a gradient of fairy shrimp density both with field observations and a microcosm experiment. Last, we examined the relationship between fairy shrimp consumption and body condition, a validated fitness proxy in this population (see Figure B2 in Lackey et al. 2019), and tested for sex-specific effects.

1.3. Methods

1.3.1. Fairy Shrimp Data Collection

1.3.1.1. Pond Sampling

At MCNP, fairy shrimp typically occur only in ponds with nonpermanent hydroperiods (dry at least once every few years) due to high rates of predation by paedomorphs in permanent ponds (Bohonak and Whiteman 1999). Based on historical occurrences (Wissinger et al. 1999) and occurrences during pilot sampling for this study, we chose nine nonpermanent ponds in which to sample fairy shrimp. In 2021 and 2022, we sampled each pond once per week between mid-June and early August ($N = 8$ weeks), which represents the typical window during which metamorphs are present in ponds (Whiteman et al. 1994).

Fairy shrimp are often patchily distributed throughout a pond (S. Wissinger, unpublished data), and we therefore established several non-overlapping transects that ran along invisible lines between shoreline points and the pond center. The number of transects per pond (range: 3 – 8) and the length of each (range: 1 – 8 m) varied by pond size and shape, and together represented adequate spatial coverage for mean count and biomass estimates. For transects that were > 3 m and occurred in water depths > 25 cm, we threw and retrieved a 1.2-mm mesh

zooplankton net (0.058 m² opening) attached to a rope that was knotted at each meter to enable replicated throws. Retrievals occurred in the middle of the water column. Along shorter and shallower transects, we conducted D-net (1.2-mm mesh; 0.065 cm² opening) sweeps and recorded the water depth when the net was not entirely submerged. We took a top-down photo (iPhone SE, 2nd generation) of each sample in a plastic tray and returned fairy shrimp to the appropriate section of the pond.

1.3.1.2. Length-Weight Regression

Throughout the 2021 season, we collected fairy shrimp vouchers (N = 133 individuals) across the observed size range and stored them in 90% ethanol. Each fairy shrimp was photographed indoors on a whiteboard with a field ruler and placed in tinfoil weigh boats with known weights. All samples were dried for 48 hours at 50-60°C and weighed to the nearest 0.001 mg on a Sartorius MC5 microbalance. We measured fairy shrimp length—defined as the top of the head to the base of the thorax (see Eriksen & Belk 1999 Figure 1.2)—with program Fiji (Schindelin et al. 2012). In program R version 4.2.0 (R Core Team, 2022), we constructed a log-log length-weight linear regression (Figure A.1), described by the formula

$$\log(\text{weight}) = 2.22706 * \log(\text{length}) - 3.69561, \quad (\text{Eq. 1.1})$$

that resulted in high explanatory power among our two morphological variables ($P < 0.001$; $R^2 = 0.91$).

1.3.1.3. Biomass Density Estimates

We calculated fairy shrimp biomass density (mg m⁻³) estimates using the data photos from each transect. First, we measured the length of each fairy shrimp in a given photo in Fiji,

converted each length to a biomass estimate using the length-weight regression formula (Eq. 1.1), and summed all biomasses in a given transect. Second, we converted our transect biomass estimates into volumetric density measurements by dividing each value by the total volume of water sampled during the transect. Sample volumes equaled the surface area of the net opening that was fully submerged multiplied by the length of the transect. Third, we calculated the average density of fairy shrimp biomass across all transects for each date-pond combination.

1.3.2. Metamorph Data Collection

1.3.2.1. Diet Composition

Salamanders are routinely captured in MCNP ponds as part of a long-term capture-mark-recapture study that has occurred annually since 1988. On two occasions each year, we assessed diet for metamorphs captured in nonpermanent ponds using gastric lavage (Joly 1987). Annual diet sampling dates aligned with peak (mid-July) and declining (late July) fairy shrimp densities in many ponds (Figure 1.1). Routine demographic data (e.g., weight, snout-vent length [SVL], breeding condition) were collected for each individual prior to gastric lavage, and we calculated body condition by dividing weight by SVL (Whiteman et al. 2012). There are positive relationships between body condition and fitness metrics for both male (male-male competition and female choice; Whiteman 1997, Whiteman et al. 1999) and female salamanders (egg production; Lackey et al. 2019) in this population. Stomach contents were stored in separate vials in 90% ethanol. All salamander handling was approved by both the Murray State University and Rocky Mountain Biological Laboratory Institutional Animal Care and Use Committee (IACUC).

For each stomach sample, we separated diet taxa into twelve orders (Anomopoda [water fleas], Acari [mites], Anostraca [fairy shrimp], Araneae [spiders], Calanoida [copepods], Coleoptera [beetles], Diptera [true flies], Hemiptera [true bugs], Hymenoptera [ants/wasps],

Lepidoptera [moths], Odonata [dragonflies], Trichoptera [caddisflies]) under a stereoscopic microscope. Diet items were counted by heads (eyes and/or mouthparts) and, for each sample, diet categories were placed in separate vials. We treated Anomopoda and Calanoida as one zooplankton prey group to expedite counting. Ingested non-food items such as conifer needles and woody debris were not included in analyses. We dried all samples (48 hours at 50-60°C) and weighed them with a Sartorius MC5 microbalance to obtain biomass estimates for each prey category per stomach sample. Since some diet items were partially digested—whether they were counted (head present) or not—we recognize that our biomass estimates will be biased lower than the true values available for energy assimilation by metamorphs. Potential biases are also introduced by differential digestibility across diet taxa, however these are likely to be relatively small compared to the variation in their total biomasses, particularly for a primary prey taxon such as fairy shrimp.

1.3.2.2. Prey Selection

We tested metamorph prey selection across a gradient of fairy shrimp density using a microcosm experiment with a four-member prey assemblage common in nonpermanent ponds in this preserve. Twenty-five plastic boxes were filled with approximately 53 x 37 x 8 cm of filtered pond water (500-µm mesh). Each of our five treatments—with five replicates each—included the same relative abundances of fifth instar caddisfly larvae (*Asynarchus nigriculus*, N = 10), mosquito larvae (*Aedes* spp., N = 10), and adult copepods (Calanoida spp., N = 10). We then varied fairy shrimp density across our treatments as follows: no fairy shrimp, low fairy shrimp density (N = 10), medium fairy shrimp density (N = 25), high fairy shrimp density (N = 50), and a control fairy shrimp density (N = 25) without salamanders. The control treatment was used to

assess prey mortality due to factors other than metamorph predation, such as abiotic conditions and predation by other prey taxa. After collecting demographic data and emptying stomach contents using gastric lavage, twenty metamorphs from the same nonpermanent pond were haphazardly assigned to treatment replicates. While each treatment contained a mixture of metamorph sexes and ages, group sample sizes were small within each treatment, and we did not consider variation in prey selection across these two demographic variables. We placed lids with several air holes on boxes to prevent metamorphs from escaping. The experiment began at 18:00 and we returned to count the number of remaining prey in each category at 11:00 the following morning, 17 hours after the experiment start. At the conclusion of the selection experiment, we quantified the rate of fairy shrimp consumption across metamorph treatments by putting 10 fairy shrimp in each box—including the no fairy shrimp treatment—and recording with a stopwatch the time of each successful fairy shrimp capture. We continuously observed boxes for the first 10 minutes and then performed a final count of remaining fairy shrimp after 20 minutes.

To assess metamorph prey selection in the experiment, we used the Manly-Chesson selection index (Manly 1974, Chesson 1978, 1983),

$$\alpha_i = \frac{r_i / e_i}{\sum_{i=1}^m r_i / e_i}, \quad (\text{Eq. 1.2})$$

where α_i is a relative measure of selection, r_i is the proportion of prey item i in the diet (i.e., dietary frequency), e_i is the proportion of prey item i in the environment (i.e., environmental frequency), and m is the number of prey types in the environment. For each prey type, the index quantifies proportional use relative to proportional availability. The index ranges from 0 (complete avoidance) to 1 (strongest possible selection), and $\alpha_i > 1/m$ indicates prey selection while $\alpha_i < 1/m$ indicates prey avoidance. Given that we did not include any vegetation or detritus in boxes and each box was relatively small compared to the size of metamorphs, our

experiment only quantified prey selection and did not capture the potentially different costs of searching for each prey taxa in natural environments.

1.3.3. Statistical Analysis

All statistical analyses were performed in program R. To test for spatiotemporal trends in fairy shrimp biomass density in ponds, we used a two-part approach to analyze semi-continuous zero-inflated data. First, we used generalized linear mixed models (GLMMs) with a binomial error distribution in the glmmTMB package (Brooks et al. 2017) to model fairy shrimp presence-absence. Models included two discrete (year and pond) and one continuous (week) fixed effects and a random intercept of transect to account for non-independence of repeated sampling. Second, we used linear mixed models (LMMs) in the lme4 package (Bates et al. 2015) to model positive density values only. Each model included two discrete (year and pond) and one continuous (week) fixed effects, one interaction (pond $\times$ week), and a random intercept of transect. For both model types, we included a quadratic term of week to account for the non-linear trend of fairy shrimp biomass. For the LMMs, we log transformed the response variable to meet model assumptions of residual normality. Main and interactive effects were examined with an analysis of variance (ANOVA) using Type II Wald χ^2 tests. When categorical model terms were statistically significant ($P \leq 0.05$) we constructed post-hoc contrasts to assess pairwise comparisons using the emmeans package (Lenth et al. 2020). We report error terms as standard errors unless otherwise noted.

We used non-parametric multivariate techniques to evaluate differences in the composition of prey taxa across stomach samples. Samples were alternately grouped into temporal (year and sampling period), spatial (historical pond hydroperiod and bench elevation),

and demographic (metamorph cohort class and sex) categories, and differences between groups were tested with analysis of similarity (ANOSIM) based on a Bray-Curtis ranked dissimilarity matrix of absolute biomass values (vegan package; Oksanen et al. 2017). Cohorts were grouped into four classes—oldest (first captured in 1996–2003, $N = 18$), middle (2004–2011, $N = 18$), youngest (2012–2020, $N = 17$), and unknown ($N = 42$)—to better incorporate a continuous variable with many unknown values into analyses. ANOSIM tests compare the mean of pairwise ranked dissimilarities among between- versus within-group sample pairings (Clarke 1993). Test statistic (R) values near 1 suggest dissimilarity between groups, values near 0 suggest an even distribution of high and low ranks with and between groups, and negative values suggest that dissimilarities are greater within than between groups. For statistically significant group differences, we performed post-hoc indicator species analyses using the indicpecies package (Cáceres and Legendre 2009). This approach uses an association index that is based on Pearson correlation coefficients between a binary variable indicating whether the stomach sample belongs to the groups under consideration (e.g., male and female) and a quantitative variable containing the biomass of the prey taxa (Cáceres et al. 2010). A permutation test is performed (iterations = 10000) that compares the observed association index with a distribution obtained by randomly reordering the data. The P -value for positive and negative diet taxa-group relationships is the proportion of the permutations that yield a higher and lower association value than that of the observed data, respectively. The prey composition of stomach samples were visualized with non-metric multidimensional scaling (NMDS), which is an ordination technique based on a rank dissimilarity matrix that aligns with the approach of an ANOSIM statistical test. Ellipses drawn around each group represent ± 1.96 standard deviations from the mean diet compositions.

We accounted for potential biases in our diet analyses in two ways. First, we evaluated the independence of diet compositions for metamorphs that were sampled more than once (20 out of 73 unique individuals) by calculating Pearson's r and testing if NMDS x - and y -values were highly correlated between the first and second capture. The correlation coefficient for both sets of values were below $|0.3|$ (Figure A.2). Second, since metamorphs are able to move between ponds, gut content provenance may not be exclusively from the pond of capture. However, we believe that this is unlikely among our samples based on the individuals' capture histories. Out of 96 stomach samples, 78 of the metamorphs were recorded by capture-mark-recapture sampling in the same pond during the previous capture that year. Over ninety percent of metamorphs that return to the pond system in a given year visit only one pond (H. Whiteman, unpublished data). Thirteen metamorphs were not captured in any pond that year prior to the date of diet sampling and are assumed to have immigrated into the pond directly from the terrestrial environment. Three metamorphs had their prior capture in a permanent pond, but their diets were predominantly fairy shrimp (mean $\pm$ SD: $97.4 \pm 2.8\%$), which suggests that each had been in the pond of capture for at least several days. Finally, two metamorphs were last captured in a different nonpermanent pond one month and two weeks prior to the date of gastric lavage. However, no rainy nights—which are when most inter-pond movements in July occur (Whiteman et al. 1994)—occurred in the days preceding gastric lavage in both instances (USDA Natural Resources Conservation Service 2022).

To evaluate metamorph prey selection, we constructed LMMs to test for differences among mean Manly-Chesson selection index values (α). Models contained fixed effects of treatment and diet taxon, their interaction, and a random intercept among replicates to account for non-independence among diet taxa that were exposed to the same salamander. Certain

comparisons (e.g., selection for fairy shrimp across treatments) required excluding the no fairy shrimp treatment from the dataset used in the model. To evaluate the number of fairy shrimp consumed in a 20-minute period after the selection experiment, we constructed a linear model with a fixed effect of treatment. For both analyses, we investigated statistically significant terms in the ANOVA with post-hoc contrasts that were adjusted for multiple comparisons with Tukey *P*-value corrections.

Lastly, we examined the relationship between the contribution of fairy shrimp to metamorph diets and the prey taxon's density in ponds in two ways. First, we calculated the mean a) total fairy shrimp biomass and b) percent contribution of fairy shrimp in metamorph diets for each date-pond combination. Each of these diet metrics was plotted against the density of fairy shrimp biomass in ponds, and we fit four-parameter logistic regressions to the relationships using the *drc* package (Ritz et al. 2015). We defined the lower and upper asymptotes with minimum and maximum observed values, respectively, allowed the model to estimate the inflection point, and manually adjusted the slope parameter until the curve best approximated the relationship. We removed two points from these plots for which fairy shrimp density in the pond was zero, but metamorphs had recently consumed the prey taxon. This was likely the result of a rapid decline in fairy shrimp abundance (pond 13, Figure 1.1) due to pond drying and water temperatures exceeding the critical thermal maximum of fairy shrimp. Second, we evaluated the relationship between metamorph body condition and the biomass of fairy shrimp in stomach contents. We constructed a LMM with the natural exponential function (e^x), a fixed effect of sex, their interaction, and a random intercept among ponds. We modeled this relationship only for metamorphs with fairy shrimp present in their stomachs to explicitly test how variation in fairy shrimp consumption influences metamorph body condition. We did not

measure other environmental variables that contributed to variation in body condition for metamorphs that did not consume fairy shrimp.

1.4. Results

1.4.1. Spatiotemporal Patterns in Fairy Shrimp Density

The presence of fairy shrimp among all nonpermanent ponds in both years followed a concave-down quadratic relationship across eight weeks that illustrated the hatch-grow-die phenology of fairy shrimp in these ponds ($\chi^2 = 45.51$, $df = 1$, $P < 0.001$). Presence also varied inter-annually with fairy shrimp twice as likely to occur in ponds in 2021 (0.82 ± 0.03) than in 2022 (0.40 ± 0.04 ; $\chi^2 = 73.93$, $df = 1$, $P < 0.001$). Spatially, occurrences of fairy shrimp were distributed heterogeneously among ponds, with the lowest and highest probabilities of presence in ponds six (0.16 ± 0.05) and fifteen (0.97 ± 0.02), respectively ($\chi^2 = 115.37$, $df = 8$, $P < 0.001$). Fairy shrimp biomass density followed similar spatiotemporal patterns as fairy shrimp presence-absence. Overall densities (conditioned on presence) among ponds and years exhibited a concave-down quadratic relationship across eight weeks ($\chi^2 = 113.46$, $df = 1$, $P < 0.001$), densities were higher in 2021 ($142.0 \pm 16.2 \text{ mg m}^{-3}$) than in 2022 ($27.6 \pm 3.9 \text{ mg m}^{-3}$; $\chi^2 = 138.0$, $df = 1$, $P < 0.001$), and densities ranged from lowest to highest in ponds six ($2.0 \pm 1.4 \text{ mg m}^{-3}$) and fifteen ($1322.4 \pm 309.8 \text{ mg m}^{-3}$), respectively ($\chi^2 = 631.91$, $df = 8$, $P < 0.001$; Figure 1.1). In addition, the quadratic trend of weekly fairy shrimp densities differed across ponds ($\chi^2 = 19.35$, $df = 8$, $P = 0.01$).

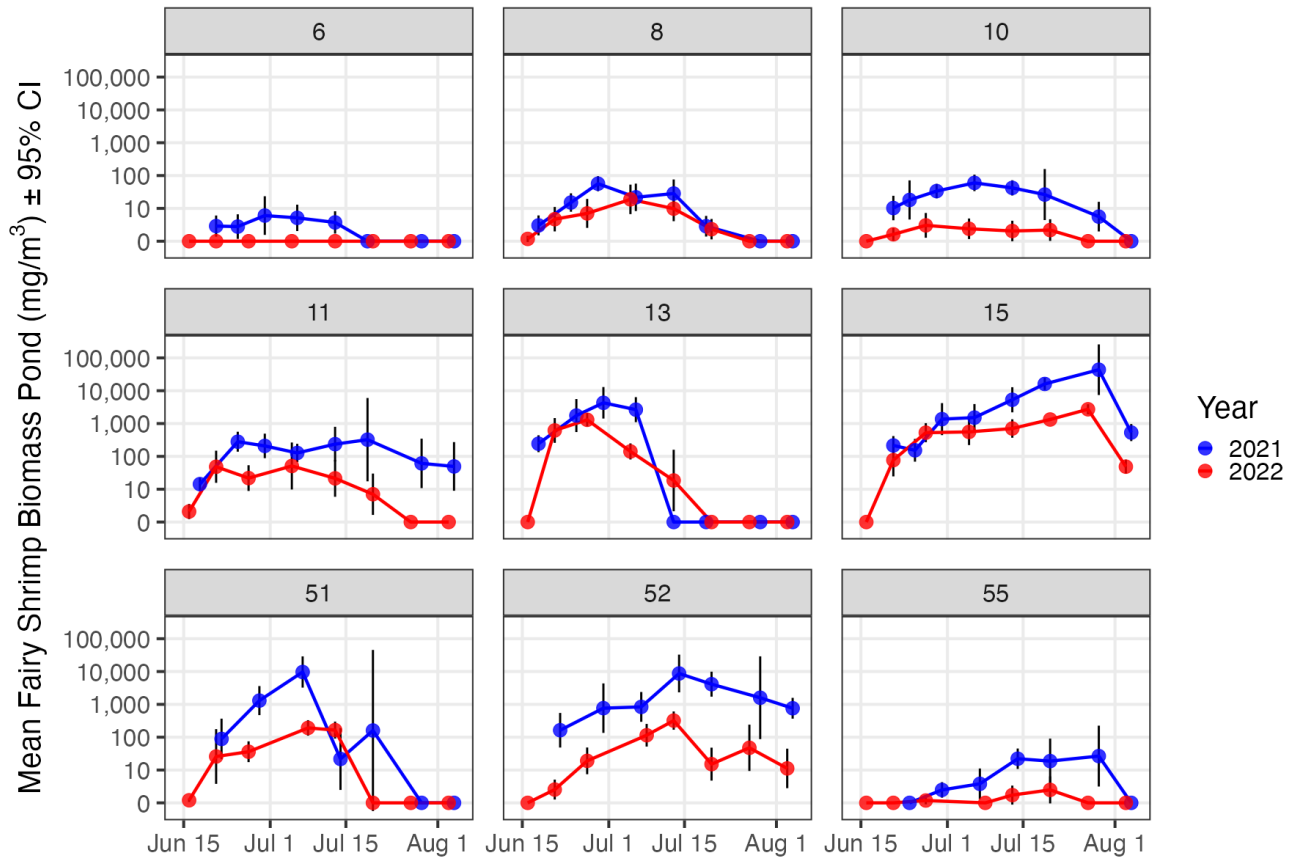


Figure 1.1. Mean density of fairy shrimp biomass in nine nonpermanent ponds at Mexican Cut Nature Preserve across eight weeks in 2021 and 2022. Pond numbers are displayed above each plot.

1.4.2. Metamorph Diet Composition

We collected stomach samples from 35, 21, 23, and 17 metamorphs on July 12, 2021; July 27, 2021; July 11, 2022; and July 25, 2022, respectively. Samples had an average of 81.8 ± 107.4 mg of total prey (range: 0.006 – 533.2 mg), and a mean observed richness of 2.3 ± 0.9 diet taxa (range: 1 – 5 taxa; Figure 1.2). The composition of diet taxa differed intra-annually (sampling period: $R = 0.21$, $P < 0.001$), but not inter-annually (year: $R = -0.007$, $P = 0.56$, Figure 1.3). Zooplankton were an indicator of mid-July metamorph diets (correlation statistic = 0.27, $P = 0.008$), while moths—a terrestrial subsidy—were indicative of late July diets (correlation

statistic = 0.32, $P < 0.001$; Figure 1.3). Metamorphs did not exhibit statistically significant differences in diet across either of our spatial groups, elevation ($R = -0.04$, $P = 0.72$) and hydroperiod ($R = -0.16$, $P = 0.99$, Figure 1.3). In contrast, diet composition varied across both metamorph demographic groups—sex ($R = 0.06$, $P = 0.02$) and cohort class ($R = 0.08$, $P = 0.03$, Figure 1.3)—with similar magnitudes of dissimilarity within each group. Fairy shrimp (correlation statistic = 0.27, $P = 0.007$) and ants (correlation statistic = 0.20, $P = 0.03$) were indicator taxa for female and male diets, respectively, while zooplankton (correlation statistic = 0.36, $P = 0.005$), beetles (correlation statistic = 0.31, $P = 0.02$), and caddisfly larvae (correlation statistic = 0.34, $P = 0.009$) were indicative of the youngest, middle aged, and oldest metamorphs, respectively.

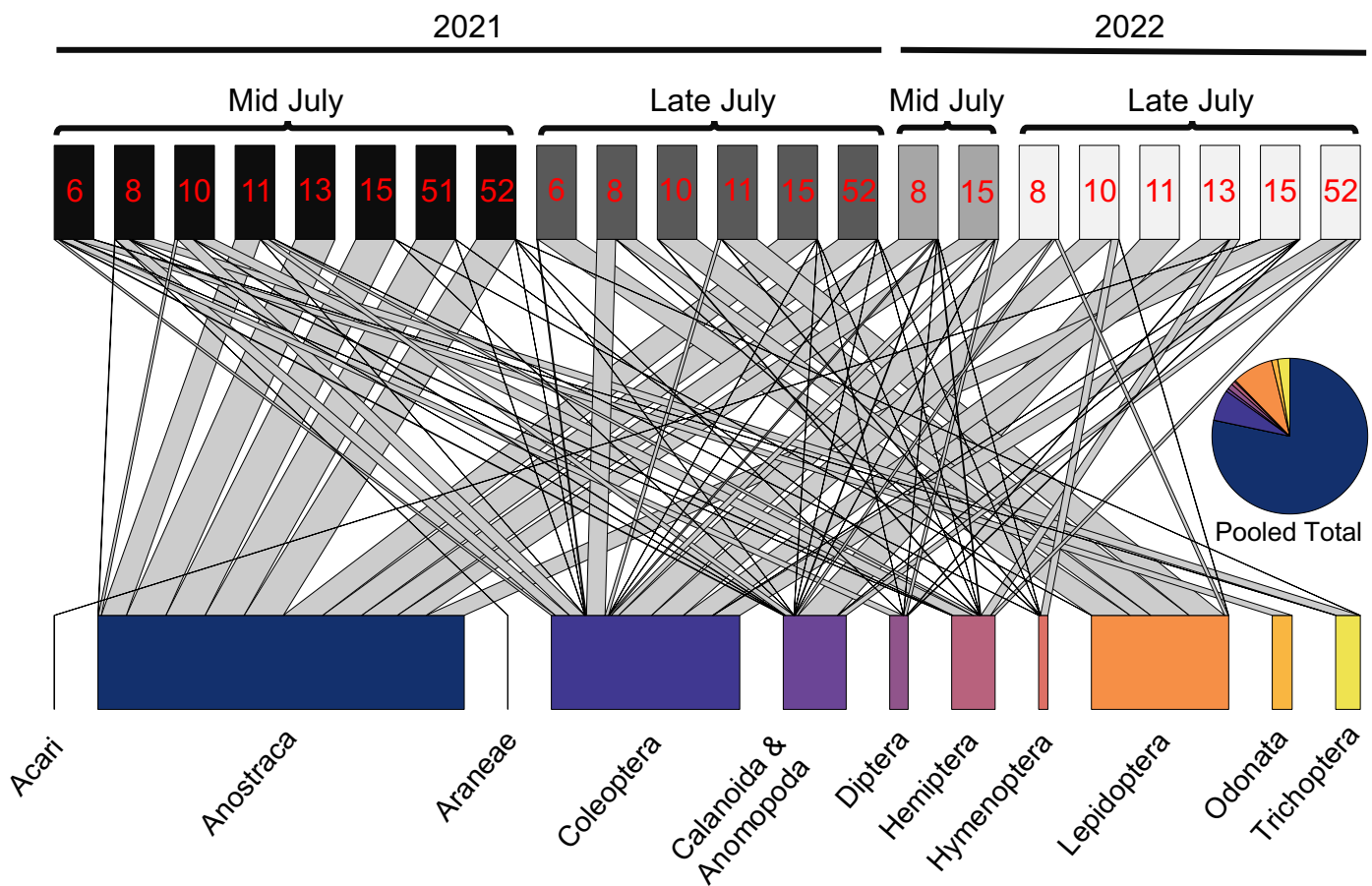


Figure 1.2. Taxonomic composition of metamorph diets evaluated through morphological examinations of stomach contents (N = 96). The thickness of each line represents the percent biomass of a diet category from pooled metamorph stomach samples for each date-pond combination. The lines for each date-pond combination sum to 100%. The width of each prey taxa block is the sum of the widths (percentage values) of the lines feeding into it. The pie chart illustrates the composition of prey taxa biomass pooled across all 96 stomach samples. We did not collect stomach samples from metamorphs in pond 55 and therefore the pond is not represented here. Pond numbers are illustrated in red text. See methods section for common names of prey orders.

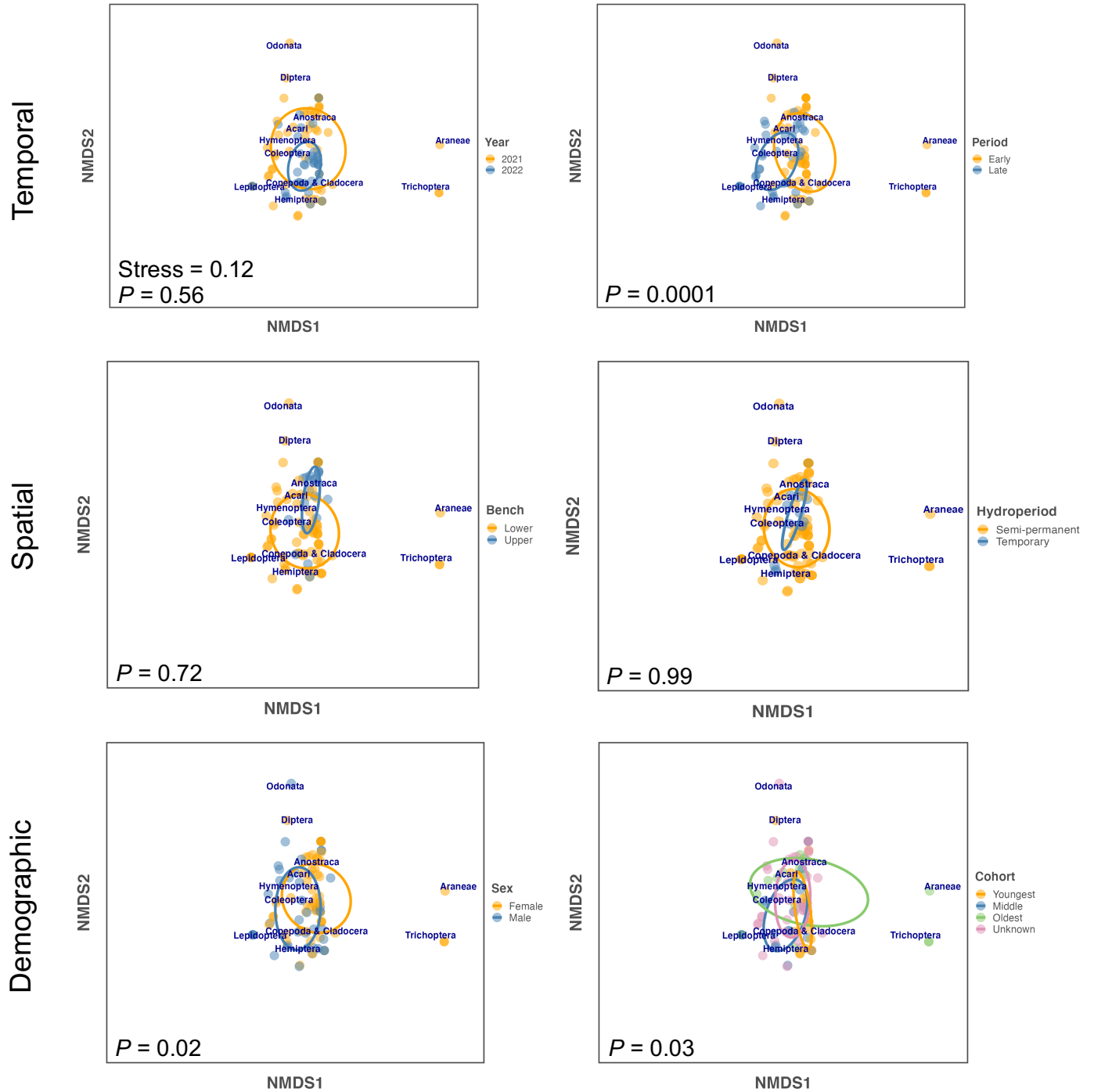


Figure 1.3. Non-metric multidimensional scaling (NMDS) ordinations displaying the similarity of metamorph diets. Each point represents an individual stomach sample, and points are alternately color-coded based on temporal, spatial, and demographic groupings. Points closer together are more similar in diet composition. Ellipses drawn around each group represent ± 1.96 standard deviations from the respective mean diet composition. P -values are from analysis of similarity (ANOSIM) tests based on a Bray-Curtis dissimilarity matrix of absolute biomass values. See methods section for common names of prey orders.

1.4.3. Metamorph Prey Selection

The three fairy shrimp densities (low, medium, high) did not affect the prey selection values in our microcosm experiment (treatment $\times$ diet taxon: $\chi^2 = 4.37$, $df = 6$, $P = 0.63$; Figure 1.4). In each of these three treatments, metamorphs selected most strongly for fairy shrimp followed by mosquito larvae, while they selected most strongly against *Asynarchus* caddisfly larvae followed by copepods. All pairwise differences between prey selection means were statistically significant ($P < 0.05$) and 17 out of 20 metamorphs displayed the experiment-wide order of preference. In the treatment without fairy shrimp, metamorph selection against caddisfly larvae was similar to those in the three treatments with fairy shrimp (no vs. low, $P > 0.99$; no vs. medium, $P > 0.99$; no vs. high, $P = 0.97$). However, selection values were greater for both copepods (no vs. low, $P = 0.03$; no vs. medium, $P = 0.002$; no vs. high, $P = 0.02$) and mosquito larvae (no vs. low, $P < 0.001$; no vs. medium, $P < 0.001$; no vs. high, $P < 0.001$) in the absence of fairy shrimp (Figure 1.4). In the control treatment, there were no declines in *Asynarchus* larvae (mean count at end of experiment: 10 ± 0), negligible declines in mosquito larvae (9.8 ± 0.2) and copepods (9.4 ± 0.4) possibly due to observer error, and small declines in fairy shrimp (22.6 ± 1.0) due to predation by *Asynarchus*. Lastly, the mean number of fairy shrimp consumed over the 20-minute foraging trial did not differ across treatments, with many individuals consuming the majority of fairy shrimp ($F_{3,16} = 0.59$, $P = 0.63$, Figure 1.4). However, the fewest fairy shrimp consumed in a replicate within each treatment monotonically declined from seven to zero from the no fairy shrimp to the high fairy shrimp treatment.

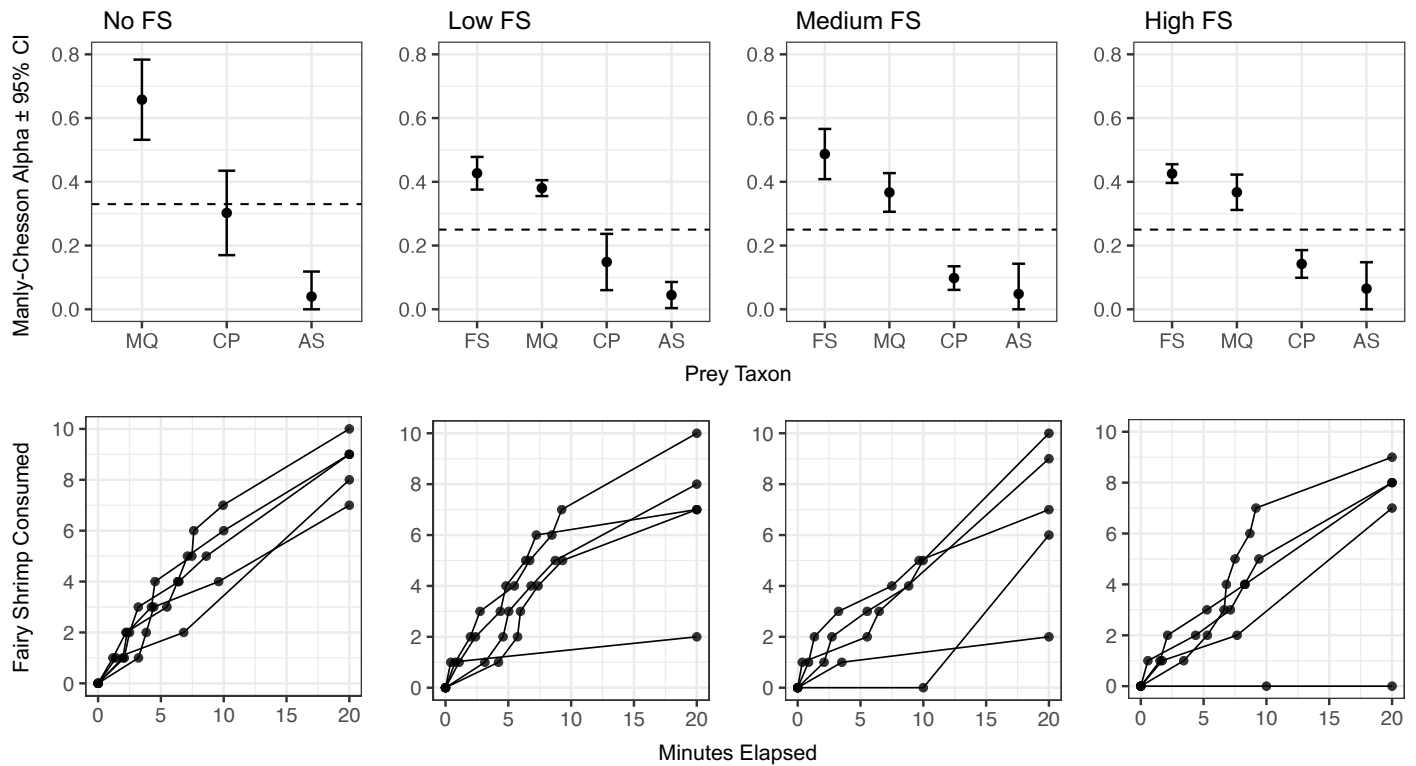


Figure 1.4. Results from the metamorph prey selection experiment across a gradient of fairy shrimp density. The top row displays the Manly-Chesson selection index (α) for metamorph foraging among four prey taxa: fairy shrimp (FS), mosquito larvae (MQ), copepods (CP), and *Asynarchus nigriculus* caddisfly larvae (AS). Points represent mean α values across five replicates. The dashed horizontal line represents no selection ($1/\text{number of prey categories}$), while points above and below the line indicate prey selection and avoidance, respectively, by metamorphs. The prey only control treatment is not shown here (see results section). The bottom row displays the rate of fairy shrimp consumption by metamorphs at the end of the selection experiment. Each line represents one replicate. We continuously monitored consumption for the first ten minutes and then performed a final count after 20 minutes.

1.4.4. Fairy Shrimp in Metamorph Diets

Fairy shrimp dominated the prey biomass in metamorphs' diets, with the prey taxon accounting for 78% of total prey biomass across both years (Figure 1.2). Many metamorphs had diets that were either predominantly fairy shrimp (43% with >90% fairy shrimp by biomass) or completely lacked the prey taxon (41% with no fairy shrimp). Among sexes, both male and female consumption of fairy shrimp biomass exhibited a type III predator functional response

with similar maximum rates of increase (Figure 1.5A, B). However, females exhibited a strong bimodal distribution in the mean percent contribution of fairy shrimp to their diet that was not observed in males. In other words, females either did not consume this prey resource entirely or they predominantly consumed it ($> 80\%$ of diet biomass). This resulted in a type III functional response for females with a much greater maximum rate of increase than in males (Figure 1.5C, D). Fairy shrimp absence in female diet occurred exclusively in ponds with lower fairy shrimp densities ($< 100 \text{ mg m}^{-3}$), while diets mostly comprised of the prey taxon occurred across the full range of fairy shrimp densities observed in ponds. Diets with intermediate fairy shrimp consumption was more common in males, with several mean values ranging between 5% and 63% of diet biomass. Similar to females, male diets mostly consisted of fairy shrimp ($> 90\%$) when individuals were in ponds with the highest fairy shrimp densities.

For metamorphs that consumed fairy shrimp ($56/96 = 58\%$), body condition exhibited a positive exponential relationship with respect to the biomass of fairy shrimp in their stomachs ($P < 0.001$, $R^2 = 0.53$, Figure 1.6). Across a range of biomass values from ~ 0.1 to 100 mg , the effect of fairy shrimp consumption on body condition was slightly positive. For metamorphs that consumed the most fairy shrimp biomass ($100 - 533 \text{ mg}$), the positive effect of consuming this prey taxon on body condition increased dramatically. These individuals with the highest fairy shrimp consumption achieved body condition values above the maximum value observed among metamorphs that did not consume fairy shrimp (0.39 g cm^{-1}). This exponential relationship did not differ by sex ($\chi^2 = 0.66$, $df = 1$, $P = 0.42$), and there were no overall differences in body condition among sexes ($\chi^2 = 1.68$, $df = 1$, $P = 0.20$). However, among metamorphs at the top of the exponential curve with body condition values $> 0.39 \text{ g cm}^{-1}$, 10 out of 12 (83%) were female.

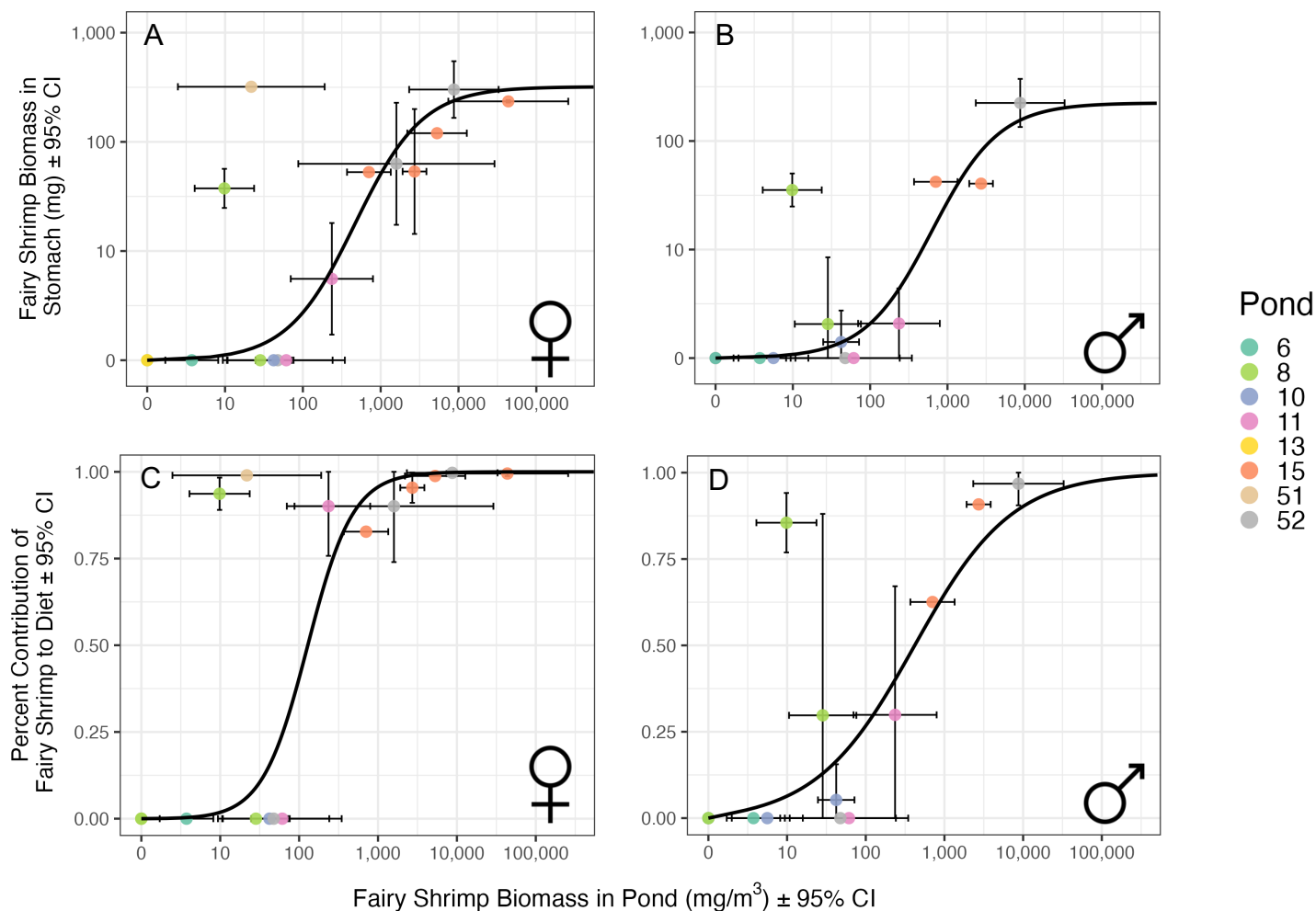


Figure 1.5. Relationships between fairy shrimp density in ponds and sex-specific metamorph consumption of fairy shrimp. Points represent mean values for date-pond combinations. Points with positive y-values that do not have vertical error bars indicate that only one metamorph was sampled for that date-pond combination. Four-parameter logistic regression curves were fit to the mean values.

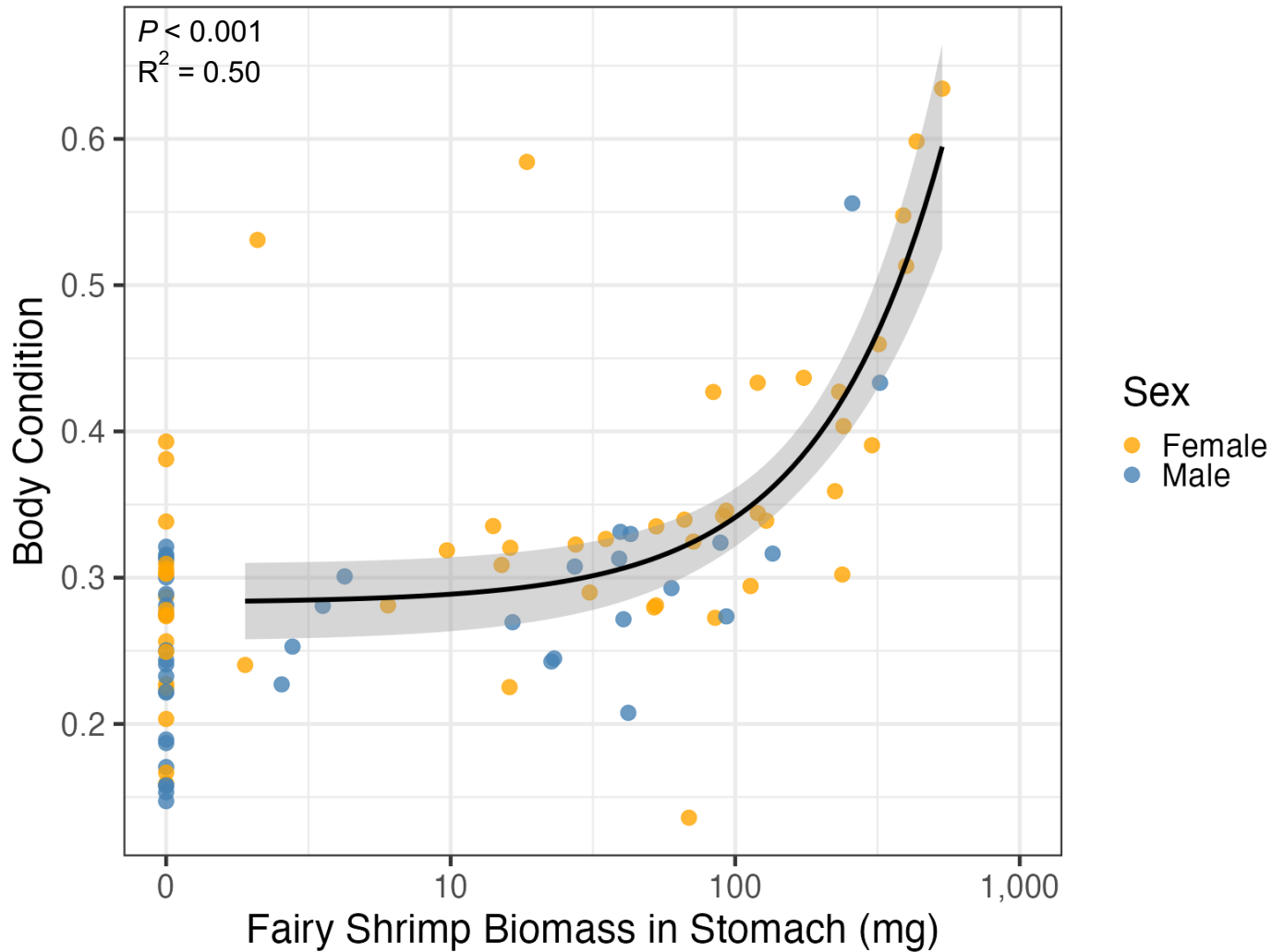


Figure 1.6. Relationship between the biomass of fairy shrimp in metamorph stomachs and metamorph body condition (g cm^{-1}). Each point represents an individual metamorph. The regression line in the plot was fit with the natural exponential function (e^x) only, while the statistical model (P and R^2 values in upper left) included an interaction with sex and a random intercept among ponds. Metamorphs with no fairy shrimp in their stomach are plotted here, but are not included in the regression line or statistical model. The shaded gray area around the regression line represents the 95% confidence interval.

1.5. Discussion

In a polyphenic population of Arizona tiger salamanders, dispersal-capable metamorphs move between ponds and may exploit prey resources in ponds with nonpermanent hydroperiods. In this study, we found that the overall metamorph diet composition in these ponds was diverse and changed across the summer, but not across years, nonpermanent hydroperiod types, or elevation. In particular, our results support prior research and show that fairy shrimp are a primary prey taxon for metamorphs in nonpermanent ponds (Whiteman et al. 1994, 1996; Bohonak and Whiteman 1999, Wissinger et al. 1999, Denoël et al. 2007). Fairy shrimp represent a boom-and-bust resource over the course of eight weeks from mid-June to early August and dominate the total prey biomass of metamorphs on several occasions during this period. Results from our prey selection experiment further confirmed that metamorphs select for fairy shrimp over other prey taxa common in nonpermanent ponds in this ecosystem. Lastly, we found evidence in support of our hypothesis that consumption of fairy shrimp contributes to fitness gains in metamorphs that balances a life-history trade-off with paedomorphs. In particular, diets of female metamorphs either consisted predominantly of fairy shrimp or lacked the prey taxon completely, while intermediate consumption was more common in males. The body condition of females that consumed high fairy shrimp biomass were among the highest across all 96 metamorphs for which we evaluated diet. For a population in which body condition is positively correlated with fecundity and metamorphs live longer on average than paedomorphs (Lackey et al. 2019), metamorphic females with high fairy shrimp consumption may play an important role in recouping initial fitness deficits incurred by the morph's later average age at first reproduction relative to paedomorphs, including the production of energetically expensive eggs. More broadly,

our findings highlight how prey resources can shape landscape heterogeneity, and how in turn this heterogeneity can maintain phenotypic trade-offs within populations.

The consumption of a diverse prey assemblage (twelve orders) by metamorphs in nonpermanent ponds has been well documented in this wetland complex (Whiteman et al. 1994, Wissinger et al. 1999, Denoël et al. 2007). Besides salamander larvae that persist between pond drying events in some ponds, metamorphic adults are the only aquatic vertebrates in nonpermanent ponds and act as keystone predators that influence prey distributions and abundances (Dodson 1970, Sprules 1972). We found differences in metamorph diet composition across sampling periods (mid- vs. late July), but not years, which highlights the stronger role of seasonal prey phenologies than inter-annual variation in determining prey availability for metamorphs. In a subalpine ecosystem where many invertebrates complete their full life cycle in a summer, prey availability can change dramatically over a few weeks. For instance, moths were mostly absent in metamorph diets in mid-July, but became a common prey item in late July when recently metamorphosed adult moths were trapped on pond surfaces. If we had evaluated metamorph diets across a longer time interval, we would expect more indicator taxa contributing to temporal differences in diet. Metamorph diets varied significantly less among our spatial groups, although our inferences were limited by largely unequal sample sizes in both sets of groups. Even so, we expect the larger source of spatial variation in metamorph diets to occur between nonpermanent and permanent ponds due to strong differences in conspecific density (i.e., paedomorph presence) and drying regime not captured in our two spatial groupings (Whiteman et al. 1994).

In addition to spatiotemporal drivers of diet composition, predator diets can vary based on demographic factors, such as age and sex (Breed et al. 2009, Wollrab et al. 2013, De Pascalis

et al. 2020). The indicator taxa associated with diet differences among metamorph cohorts—with the youngest, middle, and oldest associated with zooplankton, beetles, and caddisflies, respectively—are sequentially larger and provide evidence for an ontogenetic dietary niche shift based on gape widths in this population (Wissinger et al. 1999). Metamorph diet also varied across sexes, with ants and fairy shrimp most indicative of male and female diets, respectively. Ant consumption in our study was low overall (present in six individual diets), but their association with male diets highlights a sex-specific interaction with a terrestrial subsidy. This pattern did not extend to other terrestrial subsidies though, with non-significant differences in moth consumption across sexes ($P = 0.83$). Fairy shrimp were a much more common diet item than ants in our study, and as a result, the positive association between the prey taxon and females could have larger consequences for sex-specific differences in body condition than the male metamorph-ant association. Metamorphs in this wetland complex are known to have a strong affinity for fairy shrimp (Whiteman et al. 1994, 1996; Wissinger et al. 1999, Denoël et al. 2007), but our findings are the first to uncover sex-specific differences in fairy shrimp consumption by metamorphs.

The life-history trade-off (age at first reproduction vs. longevity) between morphs in this population is, in part, maintained by sex-specific reproductive advantages in each morph (Lackey et al. 2019). Specifically, male and female body condition contributes towards fitness gains in paedomorphs and metamorphs, respectively. Male paedomorphs have greater average lifetime reproductive opportunities than male metamorphs when accounting for the potentially higher mating success of males with higher body condition (i.e., male-male competition and female choice; Whiteman 1997, Whiteman et al. 1999). In contrast, female metamorphs have greater average lifetime egg production than female paedomorphs due to higher average body condition.

If metamorph fitness is largely tied to female body condition, we would expect there to be a selective pressure to consume a high-quality diet for this sex. While multivariate analyses of diet composition revealed a positive association between female metamorphs and fairy shrimp, this alone does not explain how the diet taxon potentially balances a life-history trade-off between metamorphs and paedomorphs.

Evaluating the relationship between the biomass of fairy shrimp in metamorph stomachs and their body condition underscores not only the contribution of the prey taxon to higher body condition, but also the requirement of high fairy shrimp consumption for metamorphs to achieve the highest body condition values. Among the 96 metamorphs for which we evaluated diet, the maximum body condition value was 0.39 g cm^{-1} (mean $\pm$ SD: $0.25 \pm 0.06 \text{ g cm}^{-1}$) for the 40 metamorphs (42%) that had not recently consumed fairy shrimp. Of the remaining metamorphs with fairy shrimp in their stomachs, body condition values of $> 0.39 \text{ g cm}^{-1}$ were predominantly observed in female metamorphs that had consumed the highest biomasses of fairy shrimp. There were two high-condition females that had consumed relatively low biomasses of fairy shrimp (Figure 1.6), perhaps suggesting that there are alternative dietary pathways to achieving high body condition. However, these individuals had few prey in their stomachs, and during their captures two weeks earlier, they recorded the two highest fairy shrimp biomass and body condition values we observed. First, this finding indicates that indeed high fairy shrimp consumption appears to be the only pathway for metamorphs—and females in particular—to achieve the highest body condition values. Second, high body condition values are not driven entirely by the biomass of prey in metamorph stomachs. One of these two individuals had 433.04 mg of prey (99.5% fairy shrimp) in her stomach during an initial capture on July 12, 2021 and 17.54 mg (96.6% fairy shrimp) two weeks later, yet her body condition value only dropped from

0.60 to 0.58 g cm⁻¹. This indicates that the high body condition conferred by fairy shrimp consumption is quickly assimilated as energy stores that can be utilized for egg production.

The importance of fairy shrimp to metamorphic female fecundity in this population has resulted in sex-specific functional responses that could have wide-ranging food-web effects. In optimally foraging predators, prey items are ranked by their ratio of caloric value to handling time (i.e., profitability), and prey items are sequentially added to diets from most to least profitable as the more profitable items become increasingly scarce (Pyke et al. 1977). This ‘top-down’ selection of prey items results in the encounter rate with the most profitable prey item determining the predator’s diet composition (Goss-Custard 1977, Krebs et al. 1977). Both male and female metamorphs exhibit similar type III functional responses as they encounter variable fairy shrimp densities. Above a density threshold, higher fairy shrimp densities resulted in higher fairy shrimp consumption by metamorphs (Figure 1.5A, B). However, the role of fairy shrimp densities in controlling metamorph diet composition is starkly different between the sexes. Males display a gradual increase in fairy shrimp selectivity (i.e., percent composition in diet) as densities of the prey taxon increase (Figure 1.5D). In contrast, females exhibit a sharp bifurcation in which they switch from consuming no fairy shrimp to predominantly fairy shrimp at intermediate prey densities (Figure 1.5C). The sex-specific shapes of these two functional response curves could lead to differential top-down structuring of nonpermanent pond food webs based on variation in the adult sex ratio (ASR) of metamorphs. For instance, if the ASR in a pond with moderate fairy shrimp density (~100 – 1000 mg m⁻³) is skewed toward females, metamorph predation will be skewed toward individuals that consume predominantly fairy shrimp. As the ASR in this pond shifts towards males, there will be more individuals that consume a mixed diet. Within a range of intermediate densities, apparent competition exists between fairy shrimp and

other members of the nonpermanent-pond prey assemblage that is mediated by the metamorph ASR. In contrast, patterns of fairy shrimp consumption are similar between metamorph sexes at the lowest (do not consume) and highest (predominantly consume) densities. If high fairy shrimp densities decline rapidly in a pond (e.g., ponds 13, 51; Figure 1.1) and metamorphs remain, there will be strong apparent competition between fairy shrimp and other prey, but it will operate independently of metamorph sex.

The long-term effects of metamorphs' top-down predation pressure in these ponds is further mediated by the different life-history strategies among the prey assemblage. Fairy shrimp need a dispersal agent (e.g., metamorphs; Bohonak & Whiteman 1999) to travel between ponds, and if metamorph predation is high enough in ponds with low fairy shrimp densities, the prey taxon could become absent until recolonization (e.g., pond 6; Figure 1.1). Fairy shrimp at MNCP likely represent a metapopulation with reliably occupied ponds (e.g., ponds 15, 52; Figure 1.1; S. Wissinger unpublished data) and others that 'blink on and off' based on factors that include metamorph predation (Levins 1969). In contrast, many of the other prey taxa that metamorphs consume (e.g., beetle and fly larvae) have life histories that involve aquatic larval and terrestrial adult stages. Even when apparent competition results in strong annual top-down pressure on insect larvae in a nonpermanent pond, these emergent taxa can continually recolonize ponds through oviposition by winged adults from other ponds (i.e., source-sink metapopulation dynamics; Pulliam 1988). As a result, the long-term effects of metamorph predation in these nonpermanent ponds—which are themselves the result of a dispersal life-history trade-off of a top predator—are in part mediated by the differences in dispersal ability among the prey assemblage.

Despite the strong patterns in fairy shrimp consumption observed in our study, the validity of prey taxon absence can be difficult to confirm with gut content analyses, particularly for soft-bodied prey that are readily digested (e.g., fairy shrimp; Hoenig et al. 2022). Future studies that incorporate stable isotopes or fecal DNA would allow for heightened sensitivity that could indicate whether metamorphs consume a few fairy shrimp at low densities or, as our results suggest, avoid the prey taxon altogether. However, these approaches are limited in their ability to evaluate prey life stages and total prey biomass, and they should therefore be incorporated in conjunction with morphological examinations of diet (e.g., Matley et al. 2018, Horswill et al. 2018, Tverin et al. 2019, Bonin et al. 2020).

Our findings provide an example of the role of a primary prey taxon in balancing a life-history trade-off in a polymorphic population. Specifically, consumption of the prey taxon allows one of the morphs to better occupy the end of the life-history trade-off that is predicated on high female body condition. More broadly, our study highlights the importance of preserving landscape heterogeneity to maintain intrapopulation phenotypic variation (McLeod et al. 1981, Lind and Johansson 2007, Sargeant et al. 2007, Darimont et al. 2009). As certain land-use (Gossner et al. 2016, Liang et al. 2019) and climatic (Magurran et al. 2015, Hidasi-Neto et al. 2019) changes homogenize landscapes, natural selection may eventually select against variation in certain traits. For instance, investment in dispersal capability at the expense of early-age reproduction may result in lower fitness than the opposing life-history strategy if there is little spatiotemporal variation in resource quality. Over the past several decades in our study system, mean annual air temperatures have increased, and maximum and minimum snow water equivalent values have decreased (USDA Natural Resources Conservation Service 2022). Populations of organisms that are nonpermanent hydroperiod specialists—such as fairy shrimp—

may become less viable as climatic baselines continue to shift. These potential losses of resources may ultimately decrease the phenotypic diversity of populations among traits that evolved in response to landscape heterogeneity.

Chapter 2

Integrating Initial Molecular Diet Data Into a Declining Species' Known Diet History: A Case Study in the North Atlantic Rocky Intertidal

2.1. Chapter Abstract

At temperate latitudes, the overwintering period for birds is often longer than subsequent periods, such as migration and breeding. Lower daily foraging success during this period may produce population-level impacts that are similar to or larger than those reported during shorter periods of the annual cycle. Purple Sandpipers (*Calidris maritima*) overwinter at high northern latitudes along North Atlantic rocky coastlines. A population in the Gulf of Maine experienced steep declines over a recent ten year period (2004–2014), yet it is unclear when in the annual cycle the strongest limitation occurred. Warming of sea surface temperatures in the Gulf of Maine accelerated recently and are correlated with declines of marine invertebrates that are common in Purple Sandpiper diet. In this study, we collected 246 Purple Sandpiper fecal samples over two years in Maine (USA) and used DNA metabarcoding to describe winter diet. We identified 24 taxonomic groups, which included many expected prey taxa (e.g., marine snails) and several less expected taxa (e.g., terrestrial insects). Surprisingly, barnacles dominated Purple Sandpiper diet, while mussels—a common diet item in previous studies—were absent. Our study is the first to employ molecular techniques to describe Purple Sandpiper diet, and as a result, we may have uncovered an unknown diet composition that was difficult to detect with morphological techniques. Alternatively, our results may be at least in part due to changes in biological processes, such as a recent dietary shift in this population during a period of increased warming in the Gulf of Maine. The relationship between a putative dietary shift in our study and the recent declines in overwintering Purple Sandpipers in Maine is intriguing, but unclear and requires further tests of winter food limitation. Regardless, utilizing a novel technique to

understand Purple Sandpiper diet has shed light on food resources during the long overwintering period.

2.2. Introduction

Animal diet during the least productive annual periods can reveal the impact of seasonally specific food limitation on longer term population dynamics (Newton 1998). At temperate latitudes, the overwintering period can be a time of relative food scarcity compared to more productive breeding months, during which failure to secure adequate food can result in mortality through starvation or hypothermia (Suter and van Eerden 1992, Gardner et al. 2017). Overwintering condition can affect fitness for bird species in other periods of the annual cycle as well, with carry-over effects influencing survival and fecundity during the subsequent migratory and breeding periods, respectively (Jansson et al. 1981, Reudink et al. 2009). For bird populations that remain at high northern latitudes during the overwintering period, short diurnal foraging periods and high thermoregulatory costs can make fulfilling daily energy requirements a challenge.

Many shorebird (Charadriiformes, suborders Charadrii and Scolopaci) populations are experiencing steep declines (Rosenberg et al. 2019). Drivers of these declines are likely numerous—climate change is altering the phenology of many species' high latitude breeding regions (Kwon et al. 2019), coastal zones that are utilized during the non-breeding period continue to be altered by humans (Small and Nicholls 2003), and long-distance migratory flights cross political boundaries with different management approaches (Watts and Turrin 2016). Diet has also been suggested as a contributing factor to declines in diverse avian taxa from seabirds to aerial insectivores (Kitaysky et al. 2006, Spiller and Dettmers 2019). For shorebirds specifically,

horseshoe crab harvesting has been linked to Red Knot (*Calidris canutus*) declines in Delaware Bay (Niles et al. 2009), and coastal development along the Yellow Sea has been tied to population declines of many species that migrate along the East Asian-Australasian flyway (Chan et al. 2019). While these two examples highlight the importance of adequate foraging opportunities during migration, the quality of overwintering sites can also influence shorebird performance and survival throughout the annual cycle (Swift et al. 2020). The overwintering period is relatively long compared to breeding and migratory periods for many avian species, and thus smaller magnitudes of daily individual foraging success may produce population-level impacts over the full overwintering period that are similar to or larger than those reported during shorter periods of the annual cycle. Additionally, some physiological processes that covary with winter diet quality—thermoregulation and pre-migratory fattening—play an outsized role in determining survival during this period or those immediately following (Prop and Deerenberg 1991, Ruthrauff et al. 2015).

Foraging ecology has also been implicated in determining the poleward-limit of an overwintering range—the distributional equivalent of population dynamics (Holt and Keitt 2000, Pulliam 2000). For instance, many shorebirds that breed at high latitudes migrate to regions that maintain above-freezing temperatures during the boreal winter, in part due to the inactivity of their sand- and mud-dwelling primary prey in the coldest regions (Evans and Smith 1975, Pienkowski 1983). Purple Sandpipers (*Calidris maritima*) are a notable exception, with populations overwintering as far north as 71 °N along North Atlantic coastlines (Strann and Summers 1990, Summers et al. 1990b, 1998). Their ability to inhabit northern locations with below-freezing winter temperatures is thought to be largely related to the availability of primary

prey in rocky intertidal habitats that typically remain ice-free (Dierschke 1994, Burton and Evans 1997, Summers et al. 1998).

Purple Sandpiper winter abundance in Maine declined sharply over a recent ten-year period, with a 49% decrease between 2004 and 2014 (Maine Department of Inland Fisheries and Wildlife 2015). It is unclear when in the annual cycle this population experiences the strongest limitation, but changes to the prey community in overwintering regions could be playing a role. Between 2004 and 2013, the Gulf of Maine was one of the fastest warming marine bodies of water in the world (Pershing et al. 2015). Increases in sea surface temperatures in this region have been attributed to declines of some invertebrates that have historically been common in Purple Sandpiper winter diet, such as blue mussel (*Mytilus edulis*) (Feare 1966, Mallory et al. 2016, Lesser 2016, Matoo et al. 2021). Additionally, the invasion of the European green crab (*Carcinus maenas*) along western North Atlantic shorelines has led to declines in shellfish stocks in numerous northeastern U.S. states (Glude 1955, Welch 1968, Ens et al. 2022). Taken together, these landscape-scale changes may be adversely impacting the quality of overwintering habitats, the effects of which could be realized as reduced overwinter survival, migratory performance, or breeding productivity.

In this study, we use DNA metabarcoding of feces to describe Purple Sandpiper winter diet in the western North Atlantic. In recent years, this technique has become increasingly common in the study of avian diets (Liu et al. 2018, McClenaghan et al. 2019, Bourbour et al. 2021). Advantages of this approach include lower risk to sampled birds, larger sample sizes, and higher-resolution descriptions of diet over more traditional visual inspections of gut contents or feces (Hoenig et al. 2022). By using DNA sequences to identify diet items, metabarcoding lessens biases toward prey items with identifiable, indigestible fragments, but the technique is

not without its own biases. For instance, metabarcoding cannot quantify absolute prey biomasses—which are valuable for understanding predator energetics—nor can it resolve differences in life stage or body size of a prey taxon (Gerwing et al. 2016, Alberdi et al. 2019). This study is one of only a few to describe the winter diet of Purple Sandpiper populations in the western North Atlantic (Smith and Bleakney 1969, Mallory et al. 2016), and it is the first to use DNA metabarcoding with this species globally. Our primary objective is to describe winter diet with an eye toward future tests of winter food limitation. We contextualize our results with previous studies on Purple Sandpiper winter diet and long-term trends in prey abundances in the Gulf of Maine. Our second objective is to understand how Purple Sandpiper diet diversity changes across tidal heights. Individuals tend to forage near the tide line throughout the tidal cycle, crossing intertidal microhabitats that exhibit macroalgal and invertebrate zonation across a vertical gradient (Stephenson and Stephenson 1949, Southward 1958, Payne and Pierce 2020). If Purple Sandpipers are generalist foragers, we hypothesize that feces collected in lower intertidal zones will have greater dietary diversity due to correlations between tidal height and intertidal invertebrate community composition (Menge and Sutherland 1987, Scrosati and Heaven 2007).

2.3. Methods

2.3.1. Fecal Sample Collection

We collected Purple Sandpiper fecal samples on five occasions, with two sampling dates (February 23, April 23) during 2018 and three sampling dates (February 3, March 1, April 12) during 2019. Individuals begin arriving on their wintering grounds in Maine (USA) from late October to early November, and typically depart for their breeding grounds in April or May (Payne and Pierce 2020). Purple Sandpipers can be found along most stretches of rocky intertidal

coastline in Maine, but they occur in highest densities on exposed offshore islands and ledges in mid-coast Maine (Mittelhauser et al. 2013). Due to the relative inaccessibility of the highest-density sites, sampling dates were wholly a function of weather conditions rather than study species or prey phenology. Winter marine conditions in the Gulf of Maine are characterized by turbulent seas that frequently make small craft passage unsafe. Our five sampling occasions represent some of the few winter days when seas were forecasted to remain calm throughout the day.

We surveyed ten sites in total within two regions of the Maine coast, Sorrento (Frenchman Bay; three sites) and South Addison (Pleasant, Wohoa, and Western Bays; seven sites; Figure 2.1). These regions have been extensively surveyed by study authors, allowing for direct knowledge of local conditions and previous locations of Purple Sandpipers (Mittelhauser et al. 2013). During the overwintering period when Purple Sandpipers are found in rocky intertidal habitats, individuals typically roost at high tide and feed throughout the remainder of the tidal cycle (Feare 1966). To align fecal collection with periods of foraging, field days typically began at a high falling tide and concluded before a full tidal cycle or nightfall. As we passed islands and ledges by boat, we scanned shorelines with 10x binoculars. We prioritized locations that had flocks of >25 individuals in order to maximize the number of fecal samples collected and avoid landings at locations with few birds. We observed flocks for at least 15 minutes after initial sighting to allow further defecation, but most were likely foraging for a longer duration prior to our arrival. We did not observe any individuals arriving during our observation period.

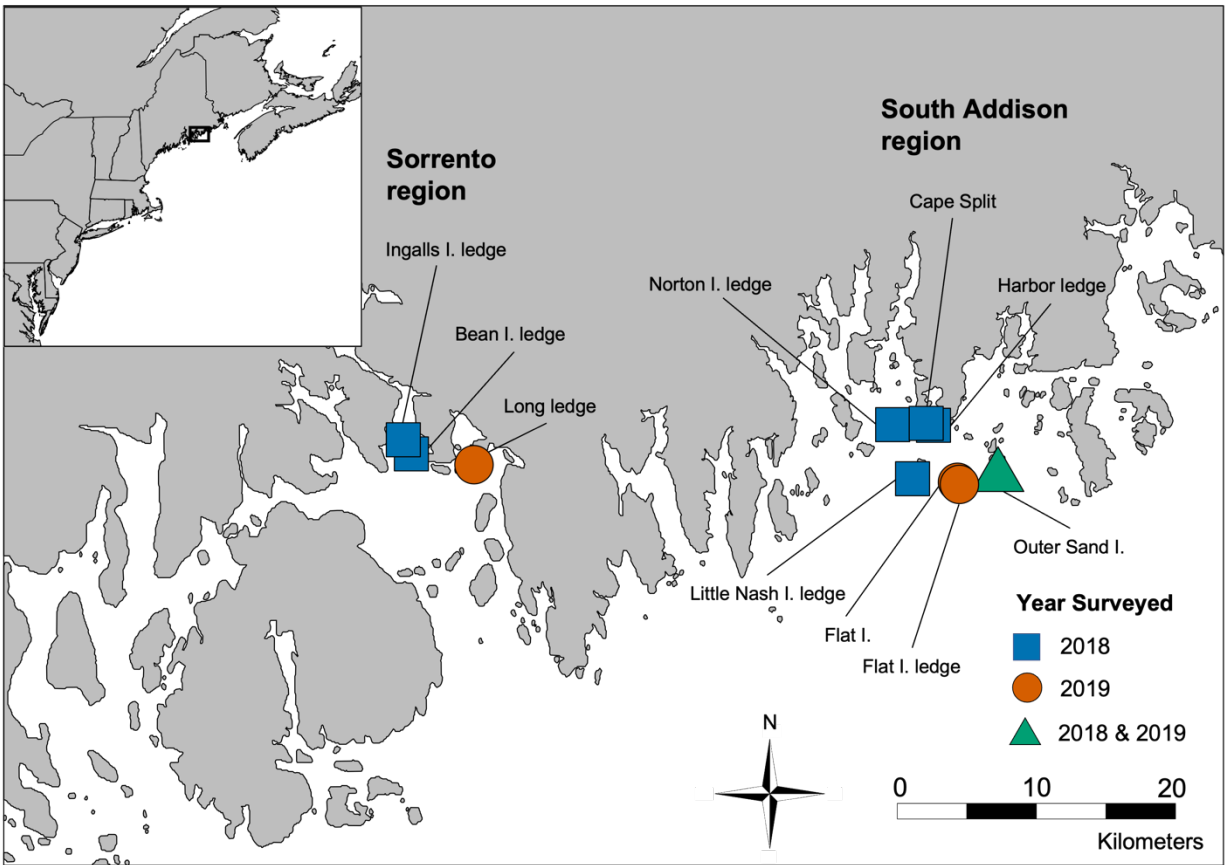


Figure 2.1. Purple Sandpiper fecal collection sites in Maine. During two consecutive winters (February – April 2018 and 2019) we visited locations in the Sorrento and South Addison regions. We collected a total of 246 fecal samples over five sampling occasions. See Table 2.1 for sample sizes collected at each site.

Fecal samples were collected directly off of the substrate (e.g., rock or seaweed) with a plastic fork or spoon. Each plastic utensil was used only once to ensure that there was no cross-contamination between fecal samples. Each sample was wrapped in a unique tinfoil sheet and placed in a small plastic bag. We collected fecal samples at a site until at least one of the following conditions (assessed sequentially) was true: a) marine conditions became less favorable, b) we ran out of unused utensils, c) we collected all fecal samples, or d) we collected a

majority samples and we opted to save unused utensils for other sites. There was a high likelihood that all fecal samples collected were those of the target species for two reasons. First, Purple Sandpipers are the only regularly wintering shorebird in this region. Second, bird richness is low in these habitats and we predominantly collected fecal samples on recently exposed substrate that was fully inundated at higher tides, and where we observed only Purple Sandpipers foraging.

Given the high sensitivity of molecular techniques and the risk of environmental contamination, we collected negative and positive field controls. Negative controls consisted of rock scrape ($N = 1$) and seawater ($N = 2$) samples to account for the presence of non-fecal DNA that originated from the utensil scraping the substrate or seawater splashing feces, respectively. Positive controls required eliminating all potential sources of environmental DNA contamination in the field. To achieve this, we built a fecal sampling box (Figure B.2.) that allowed for the collection of feces directly from individuals ($N = 3$). The box was divided into 15 cells that had a hardware cloth floor, and below each cell was a plastic container lined with tinfoil. Individuals were placed in the sampling box for a maximum of 30 minutes, and after birds were released each fecal sample was wrapped in tinfoil and placed in a small plastic bag. We used a handheld rocket net (Super Talon Net Gun; www.net-gun.com) fired from the bow of the boat to capture birds. We targeted small flocks (<10 individuals) given that we only used this approach for collecting control samples and not as a primary method of collection. Purple Sandpipers allowed close approach, ensuring that all individuals were captured over exposed intertidal. We did not collect control samples at all sites, and as a result we treated these samples as study-wide controls rather than site-specific controls. Bird handling was conducted under the University of Maine Institutional Animal Care and Use Committee permit #A2018-02-01 and United States

Geological Survey federal bird banding permit #06400. Fecal samples were put in an icebox at the conclusion of the field day, and all samples were stored in a buffer solution (RNAlater) in individual 10 mL vials within 24 hours of collection.

2.3.2. Fecal Sample DNA Metabarcoding

2.3.2.1. DNA Extraction, Amplification, and Sequencing

DNA was extracted from samples using a DNeasy PowerSoil HTP kit (Qiagen LLC, Germantown, MD) following manufacturer's directions with the modification that 20 μ L of proteinase K (20 mg mL⁻¹) was added to each sample and incubated for ~30 min at 65 °C prior to the sample homogenization step.

Samples libraries were prepared for sequencing using a two-step PCR method (Maslo et al. 2022). The first PCR was performed to amplify a 180 base pair (bp) region of the COI gene using primer pair LCO11490 (5'-GGTCAACAAATCATAAAGATATTGG-3') and CO1-CFMRa (5'-GGWACTAATCAATTTCCAAATCC-3') (Jusino et al. 2019). Universal tails were added to these primers to allow for the use of universal barcodes in a second PCR reaction to dual-index each sample. Each 15 μ L reaction contained 1X Platinum II Hot-Start Green PCR Master Mix (Invitrogen), 0.16 μ g μ L⁻¹ Bovine Serum Albumin (Thermo Fisher) and 0.2 μ M primers. The PCR conditions were 5 min at 94 °C, followed by 35 cycles of 94 °C for 60 s, 60 °C for 90 s, and 72 °C for 60 s, with a final elongation of 10 min at 72 °C.

A second PCR was performed using amplicons from the initial PCR to add unique 8 bp indexes as well as the Illumina P5/P7 sequencing adapters to each sample. The 25 μ L reactions contained 1X Platinum II Hot-Start Green PCR Master Mix and 0.4 μ M of each primer. PCR conditions were 2 min at 98 °C, followed by 10 cycles of 30 s at 98 °C, 20 s at 60 °C, and 30 s at 72 °C, with a final elongation of 5 min at 72 °C. Products were visualized on a 1% agarose gel,

purified with AMPure XP beads (Beckman Coulter), and sample concentrations were normalized using the SequalPrep normalization plate kit (Invitrogen). Normalized samples were pooled at equal volumes and sequenced on an Illumina MiSeq instrument using a v2 2 x 150 cycle reagent kit.

2.3.2.2. Reference Library and Bioinformatics

Bioinformatics were performed using the QIIME2 platform (Bolyen et al. 2019). Demultiplexed samples were imported into QIIME2 and sequences were trimmed to 110 bp using the cutadapt plugin (Martin 2011). Sequences were denoised using the DADA2 plugin (Callahan et al. 2016) and representative sequences were taxonomically assigned using a curated reference database from the Barcode of Life project library (BOLD; www.boldsystems.org). Briefly, we downloaded all animal sequences from BOLD (Jan 2019), filtered out sequences that did not have a “COI-5P” marker code, removed gap characters and sequences <100 bp, and dereplicated sequences using a least common ancestor consensus taxonomy method.

We merged amplicon sequence variants (ASVs; i.e. sequences) that shared taxonomic assignments into taxonomic groups. The lowest taxonomic rank we assigned to ASVs was genus in an effort to increase the certainty of taxonomic assignments over species-level data. Furthermore, we did not expect our study’s conclusions to meaningfully change with species-level diet data given that many invertebrate genera have small morphological differences between species that do not contribute to a level of variation relevant to our study. We next performed several filtering steps to ensure that we reported high-quality diet data. First, we rarefied samples to a sequencing depth of 10,013 reads, which represented a balance between minimum sample quality (i.e., total read count) and the retention of the most number of samples.

Second, we removed any taxonomic groups that were not identified to at least the level of phylum ($N = 1$). Third, we removed taxonomic groups that were extremely unlikely diet taxa given their known distributions ($N = 2$). Lastly, we removed taxonomic groups from samples that represented $< 1\%$ of the total number of reads in a sample in an effort to remove any possible sample contamination or misidentification due to sequence degradation. While this approach may remove some rare diet taxa that were ingested at low frequencies, the primary objective of this study is to accurately identify the major components of Purple Sandpiper winter diet rather than describe total diet breadth.

2.3.3. Statistical Analysis

To compare diet across fecal samples, we used two metrics—relative read abundance (RRA) and percent of occurrence (POO)—that each highlight different aspects of diet composition. RRA is defined as a taxonomic group's read count relative to the total number of reads in a sample. This approach allows for estimates of the relative abundances of diet taxa biomass in fecal samples, although relative abundances can be skewed by DNA extraction or PCR amplification biases and must be interpreted with caution (Krehenwinkel et al. 2017). POO converts read counts into presence-absence data and gives equal weight to each taxonomic group in a sample (e.g., if there are six taxonomic groups present in a sample, each would have a POO value of $1/6$). This metric reduces potential amplification biases and highlights taxonomic group richness in samples, but it can be a conservative view of diet composition as rare taxa will be overrepresented relative to abundant taxa.

We used two different diversity indices to compare Purple Sandpiper diet diversity across intertidal zones: Shannon diversity index and Faith's phylogenetic diversity. Calculating several

diversity estimates allowed us to assess the robustness of diet diversity patterns across indices that each emphasize different aspects of the diet community. For each diversity index, residuals were non-normally distributed, and as a result we used Kruskal-Wallis non-parametric tests to compare whether there were differences between intertidal zone groups (high, middle, low). Pairwise differences between groups were calculated using Wilcoxon rank sum tests, and significance values were adjusted for multiple comparisons using Benjamini-Hochberg corrections. Given that sampling was opportunistic and subject to time and safety constraints, we were not able to achieve a fully crossed study design in which fecal samples were collected in all intertidal zones during each sampling occasion. To compare diet diversity across the two winters, we used the non-parametric Mann–Whitney U test.

For all diversity analyses, we accounted for differences in the tidal height between the time of diet taxa ingestion and defecation by correcting for estimated gut passage duration. Purple Sandpipers have an estimated gut passage duration of 40 minutes based on field observations (Dierschke 1994), which is similar to estimates from captive experiments with another *Calidris* species (Sánchez et al. 2012). We subtracted 1 hour—allowing 20 minutes for boat landing and sampling preparation—from the time of initiating fecal collection to estimate the time of prey ingestion. Using tidal charts, we then estimated the tide height at the time of ingestion. Given that Purple Sandpipers tend to forage and defecate near the tide line throughout the tidal cycle, it is reasonable to assume that the estimated location of ingestion corresponded roughly with the tide line. We assumed that the error generated with this approach is minimal relative to the 3 broad intertidal zone positions (high, middle, low) to which we assigned bird foraging locations. We defined “high” as < 2 hours from (either before or after) the closest high tide, “middle” as 2–4 hours away from high tide, and “low” as > 4 hours away from high tide.

High, middle, and low intertidal zones were defined as the portion of the intertidal that is newly exposed (falling tide) or newly submerged (rising tide) during our defined tidal windows. Unless otherwise noted, errors are reported in standard deviations.

2.4. Results

We collected 246 Purple Sandpiper fecal samples from ten sites (mean: 22.4 ± 20.3 samples; range: 4–62 samples) in the Sorrento and South Addison regions during 2018 and 2019. In addition, we collected three positive field controls (fecal samples from the bird box) and three negative field controls (one rock scrape and two seawater samples) across four sites. We sequenced and amplified COI gene fragments in each sample, and after rarefying samples' library sizes to 10,013 reads, we retained 192 fecal samples and 4 control samples (57 fecal samples and 2 seawater samples were removed). After the various sequence filtering steps, we retained sequences making up 24 taxonomic groups across 196 samples, with 62.5% of the groups identified to genus, 20.8% to order, 12.5% to class, and 4.2% to phylum (Table 2.1, Figure 2.2).

Out of the 24 taxonomic groups identified, there were only 4 “core” taxonomic groups present in $\geq 10\%$ of all fecal samples: genus *Semibalanus* (barnacles; present in 88.5% of samples), phylum Arthropoda (arthropods; 26.0%), class Insecta (insects; 21.4%), and genus *Apohyale* (amphipods; 20.3%). The remaining 20 taxonomic groups had a mean occurrence across all fecal samples of 2.9% ($\pm 2.9\%$; range: 0.5–9.9%), which included many expected taxa such as genus *Littorina* (marine snails), genus *Gammarellus* (amphipods), and class Polychaeta (marine worms), and several less expected taxa such as terrestrial orders Diptera (true flies) and Lepidoptera (moths and butterflies). Across all fecal samples, the mean richness was 2.2

taxonomic groups (± 1.5 ; range: 1–8). *Semibalanus* dominated Purple Sandpiper diet across years, ordinal dates, and sites, accounting for 71.4% (1,372,997 out of 1,922,496) of all dietary sequence reads across all fecal samples. Forty-six percent of fecal samples contained only *Semibalanus* reads, and only 11% of samples contained no *Semibalanus* reads (Table 2.1, Figure 2.2). Even when barnacles were present with other taxonomic groups in a fecal sample, the taxon often dominated RRAs. Out of 82 fecal samples that had *Semibalanus* reads and at least one other taxonomic group present, 60 (73%) had *Semibalanus* RRAs greater than 0.50 (Figure 2.2).

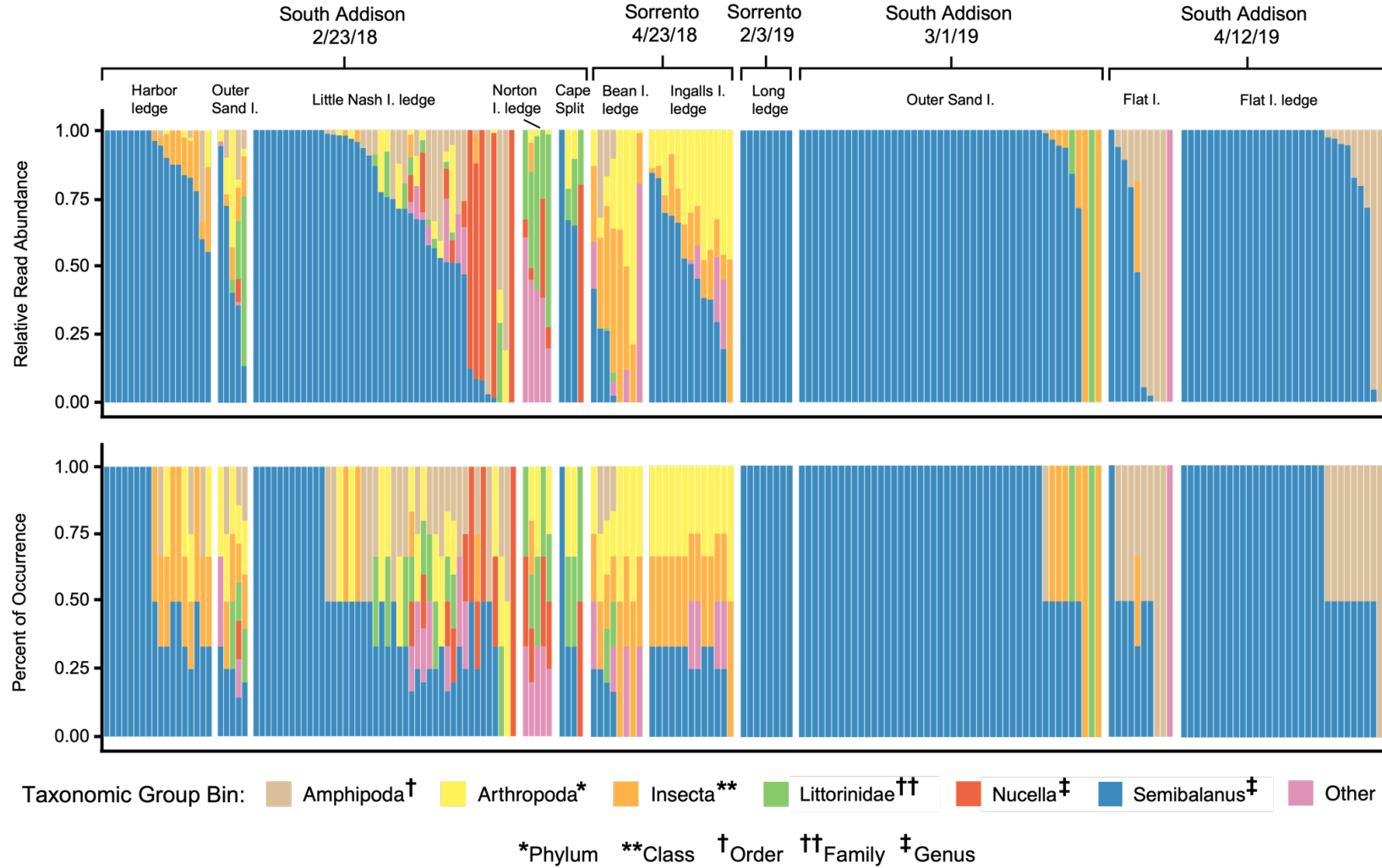


Figure 2.2. Purple Sandpiper diet composition at ten sites over a two-year sampling period. Each column represents an individual fecal sample. We identified a total of 24 taxonomic groups, but we merged groups into seven bins in an effort to aid visual clarity. See Table 2.1 for a full list of taxonomic groups. The top panel visualizes the sequencing data as relative read abundances (RRA), which are proportional summaries of read counts. The bottom panel shows percent of occurrence (POO), which displays equally-weighted presence-absence of taxonomic group bins.

Table 2.1. Site-level percent of occurrence (POO) for the 24 taxonomic groups identified in Purple Sandpiper diet. POO values represent the percent of fecal samples with a taxonomic group present. The total column represents the POO across all fecal samples. Taxonomic groups are color-coded into seven bins that match the visualization of the sequencing data in Figure 2.2.

Taxonomic Group						2018						2019					
Phylum	Class	Order	Family	Genus	Common Name	Harbor ledge	Outer Sand I.	Little Nash I. ledge	Norton I. ledge	Cape Split	Bean I. ledge	Ingalls I. ledge	Long ledge	Outer Sand I.	Flat I.	Flat I. ledge	Total
Annelida	Clitellata	Enchytraeida			Segmented worms	0	20	2.3	0	0	0	0	0	0	0	0	1
Annelida	Polychaeta				Marine worms	0	20	0	0	0	0	0	0	0	0	0	0.5
Annelida	Polychaeta	Phyllodocida			Marine worms	0	0	0	40	0	0	0	0	0	0	0	1
Annelida	Polychaeta	Phyllodocida	Polynoidae	Lepidonotus	Marine worms	0	0	9.1	100	0	0	0	0	0	0	0	4.7
Arthropoda					Arthropods	22.2	100	34.1	60	50	100	100	0	0	0	0	26
Arthropoda	Hexanauplia	Harpacticoida	Ectinosomatidae	Microsetella	Copepods	0	0	0	0	0	37.5	15.4	0	0	0	0	2.6
Arthropoda	Hexanauplia	Monstrilloida	Monstrillidae	Monstrilla	Copepods	0	0	2.3	0	0	25	15.4	0	0	0	0	2.6
Arthropoda	Hexanauplia	Sessilia	Archaeobalanidae	Semibalanus	Barnacles	100	100	90.9	0	75	50	92.3	100	93.5	70	96.8	88.5
Arthropoda	Insecta				Insects	55.6	80	6.8	20	0	100	92.3	0	6.5	0	0	21.4
Arthropoda	Insecta	Coleoptera			Beetles	0	0	0	0	0	0	0	0	0	10	0	0.5
Arthropoda	Insecta	Diptera			True flies	5.6	0	2.3	0	0	0	0	0	0	0	0	1
Arthropoda	Insecta	Diptera	Chironomidae	Halocladius	True flies	0	0	0	0	0	0	23.1	0	0	0	0	1.6
Arthropoda	Insecta	Diptera	Limoniidae	Dicranomyia	True flies	0	0	0	0	0	0	0	0	4.3	0	0	1
Arthropoda	Insecta	Diptera	Limoniidae	Erioptera	True flies	5.6	0	2.3	0	0	0	0	0	0	0	0	1
Arthropoda	Insecta	Lepidoptera			Moths	27.8	20	2.3	0	0	62.5	46.2	0	2.2	0	0	9.9
Arthropoda	Malacostraca	Amphipoda	Ampithoidae	Ampithoe	Amphipods	0	0	15.9	0	0	0	0	0	0	0	0	3.6
Arthropoda	Malacostraca	Amphipoda	Gammarellidae	Gammarellus	Amphipods	0	0	20.5	0	0	0	0	0	0	0	0	4.7
Arthropoda	Malacostraca	Amphipoda	Hyalidae	Apothyale	Amphipods	16.7	60	27.3	0	0	37.5	0	0	2.2	80	29	20.3
Cnidaria	Hydrozoa	Leptothecata	Campanulariidae	Obelia	Hydrozoans	0	0	2.3	0	0	0	0	0	0	0	0	0.5
Mollusca	Gastropoda	Littorinimorpha	Littorinidae	Lacuna	Periwinkles	0	0	9.1	100	25	0	0	0	0	0	0	5.2
Mollusca	Gastropoda	Littorinimorpha	Littorinidae	Littorina	Periwinkles	0	60	13.6	0	50	25	0	0	4.3	0	0	7.8
Mollusca	Gastropoda	Neogastropoda	Muricidae	Nucella	Dog whelks	0	20	22.7	80	25	0	0	0	0	0	0	8.3
Rotifera	Bdelloidea				Rotifers	0	0	0	0	0	0	0	0	0	10	0	0.5
Rotifera	Bdelloidea	Adinetida	Adinetidae	Adineta	Rotifers	0	0	0	0	0	0	0	0	0	10	0	0.5
Number fecal samples						18	5	44	5	4	8	13	8	46	10	31	192

Taxonomic Group Bin: Amphipoda Arthropoda Insecta Littorinidae Nucella Semibalanus Other

Given the ubiquity of barnacles in rocky intertidal zones, the contact with the intertidal substrate during fecal collection, and the sensitivity of molecular analyses, it was critical that we demonstrated that the high abundance of *Semibalanus* was not a result of environmental contamination. Two out of 3 positive field controls contained *Semibalanus* reads, with one sample containing exclusively this diet taxon (Figure B.2). Furthermore, the rock scrape negative field control did not contain any *Semibalanus* reads, and both seawater negative field controls did not contain total read counts above the rarefaction level. Notably, we did not record any occurrences of mussels (*Mytilus* spp.)—a common winter diet taxon in other studies—across the 196 fecal samples.

Purple Sandpiper diet richness was significantly higher (Mann–Whitney $U = 7714$, $P < 0.001$) in the first winter of fecal collection (mean: 3.1 ± 1.6 taxonomic groups, range: 1–8 taxonomic groups) than in the second winter (mean: 1.3 ± 0.5 taxonomic groups, range: 1–3 taxonomic groups). The occurrences of nine out of the eleven total taxonomic groups present in the second winter were restricted to ten fecal samples, with the remaining 85 fecal samples containing only *Semibalanus* and *Apohyale* reads. This further highlights the fact that diet richness was low in the large majority of fecal samples during the second winter.

There were significant differences in diet diversity across intertidal zones for Shannon diversity index [$H(2) = 74.364$, $P < 0.001$] and Faith’s phylogenetic diversity [$H(2) = 87.185$, $P < 0.001$]. For each index, all pairwise differences were statistically significant ($P < 0.001$), with diversity of diet taxa increasing monotonically from high to low intertidal zones (Figure 2.3). Lastly, the prevalence of barnacles in Purple Sandpiper diet was negatively related to tidal height for both RRA ($\beta = -0.052$, $P = 0.001$, $R^2 = 0.055$) and POO ($\beta = -0.076$, $P < 0.001$, $R^2 = 0.108$; Figure 2.4). In other words, fecal samples were less likely to be predominantly barnacle reads at

lower intertidal zones. However, these two trends explained relatively little variance, and barnacles reads were still commonly recorded in low intertidal samples at some sites.

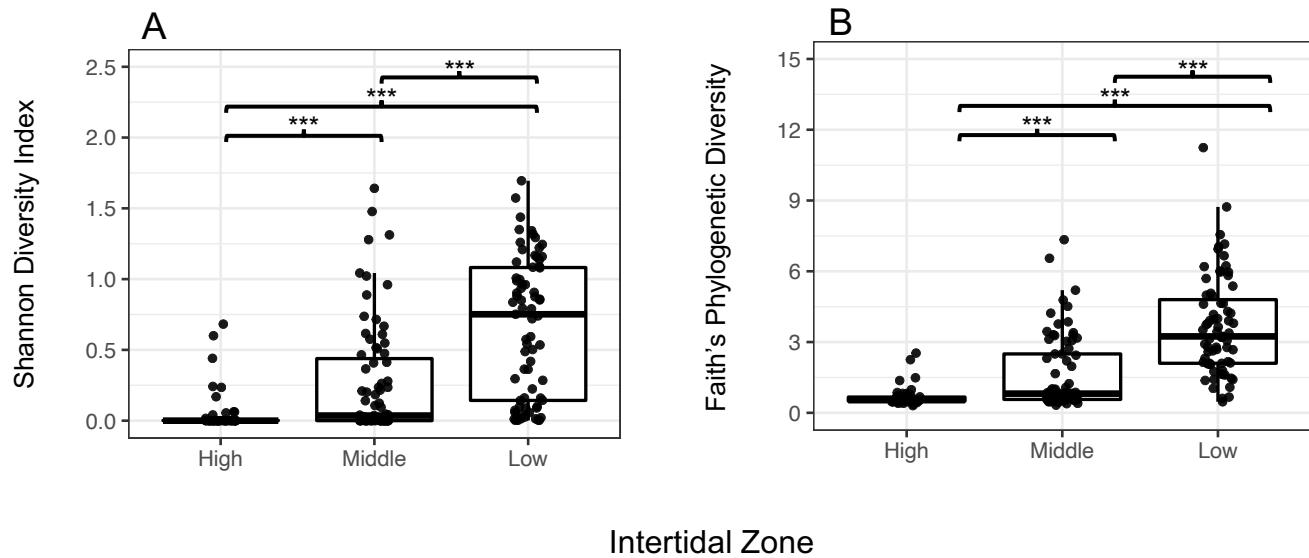


Figure 2.3. Purple Sandpiper diet diversity across three intertidal zones as measured by (A) Shannon Diversity Index and (B) Faith's Phylogenetic Diversity. Each point represents a fecal sample. In each plot, all pairwise differences are significant (***) denotes $P < 0.001$).

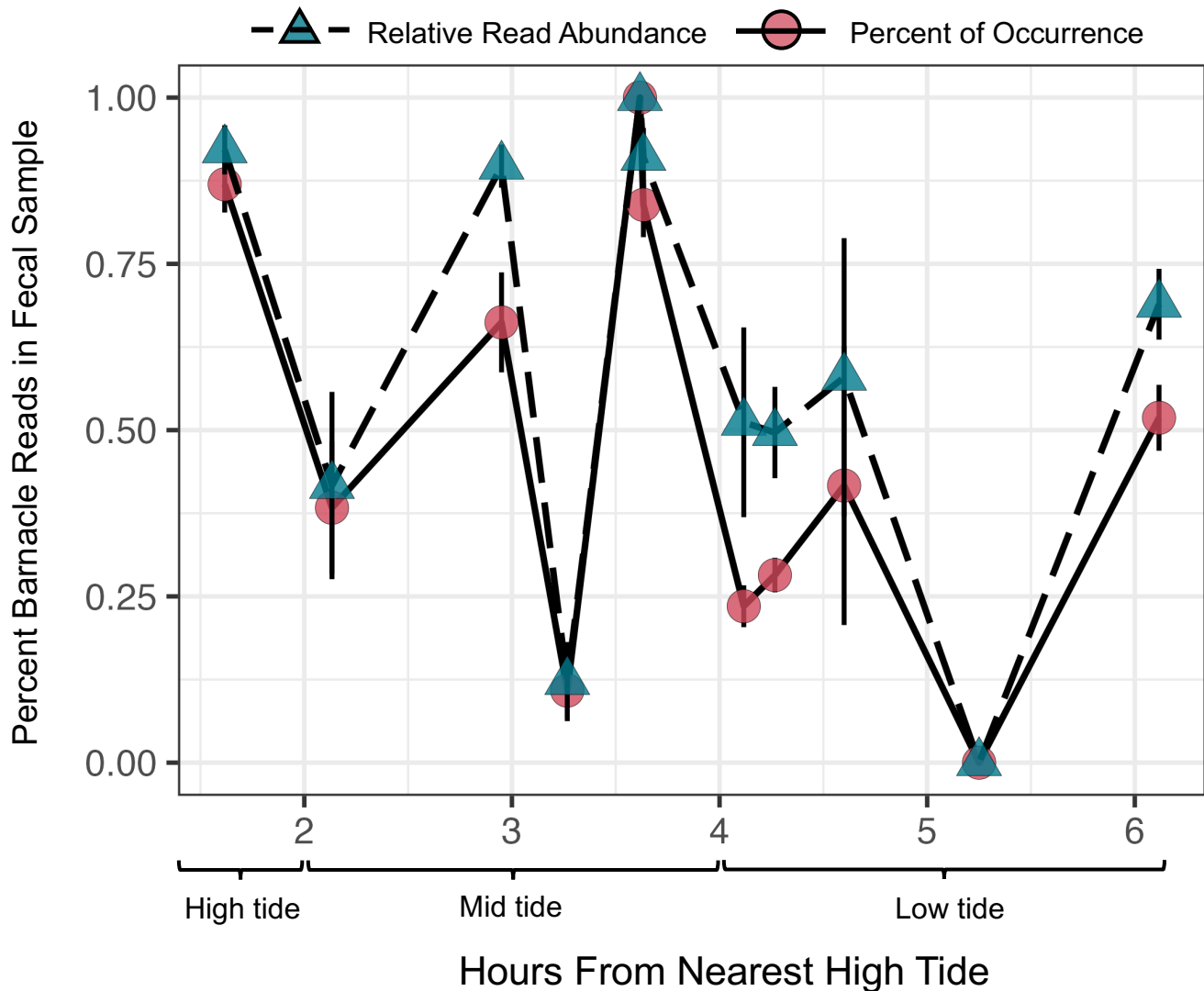


Figure 2.4. The mean ($\pm$ SE) percentage of barnacle reads in Purple Sandpiper fecal samples across the tidal cycle. Each x-axis value with points represents a sampling occasion ($N = 11$). The mean value is an average of all fecal samples collected during a sampling occasion. For each sampling occasion, means are displayed for relative read abundance (RRA) and percent of occurrence (POO) data. RRAs are proportional summaries of read counts, and POOs are equally-weighted presence-absence data.

2.5. Discussion

Declines in migratory bird populations are often driven by a multitude of factors, making the diagnosis of declines and the prescription of management solutions difficult. In considering

limiting factors during a population's full annual cycle, the overwintering period is a relatively long-duration stage during which low quality resources can reduce a population's overwinter survival rate or subsequent migratory performance and breeding productivity (Jansson et al. 1981, Suter and van Eerden 1992, Reudink et al. 2009, Gardner et al. 2017). The quality of certain resources, such as food, may be paramount for species that overwinter at higher latitudes and rely in part on their diet for optimal thermoregulatory performance in cold, daylight-limited regions.

After recent declines in overwintering Purple Sandpipers in Maine, we collected 246 fecal samples over two consecutive winters and used a metabarcoding approach to describe their winter diet in Maine. We found very high occurrences and relative abundances of barnacles, which while present in other studies, has never been described as a primary winter diet taxon in this bird species. Combined with the absence of mussels—a common winter diet taxon in previous studies—in all fecal samples, our results suggest a significant dietary shift could have occurred in this population's winter diet. Our findings also support our hypothesis that Purple Sandpiper diet diversity increases from high to low intertidal zones, mirroring rocky intertidal zonation patterns of benthic invertebrate diversity along northwest Atlantic coastlines (Scrosati and Heaven 2007).

Two notable spatiotemporal diversity patterns emerged from our diet data. First, our results confirmed our hypothesis that Purple Sandpiper diet diversity increases monotonically from high to low intertidal zones (Figure 2.3). The congruence between observed diet patterns and regional rocky intertidal zonation patterns of invertebrate diversity suggests that Purple Sandpipers are generalist foragers, but we did not quantify prey availability at our sites and therefore this finding remains speculative. Second, diet diversity was considerably higher in the

first winter than in the second winter despite collecting a similar number of fecal samples in each season ($N = 97$ and 95 , respectively) at comparable ordinal dates. The distribution of tide heights during fecal collection, however, was partially confounded by year. We collected the most fecal samples in the second winter (2019) at Outer Sand Island (46 out of 95 samples), which is the only sampling occasion in our study when we collected fecal samples at high tide. Conversely, all five sampling occasions at low tide (Little Nash Island ledge, Norton Island ledge, Cape Split, Ingalls Island ledge, Outer Sand Island) occurred in 2018. Benthic invertebrate diversity in western North Atlantic rocky intertidal ecosystems increases monotonically moving from high to low intertidal zones, and as a result, a skewed distribution of tidal heights during collection would likely bias the comparison of diversity patterns between years (Scrosati and Heaven 2007). Tide height alone, however, does not explain the inter-annual differences in diet diversity, as sampling occasions at middle tide heights had higher mean richness in the first winter (mean $\pm$ SD: 3.0 ± 1.8 , $N = 26$) than the second winter (mean $\pm$ SD: 1.3 ± 0.6 , $N = 49$). Other possible explanatory factors include spatial variation in the prey community across sites and inter-annual variation that could impact either the macroinvertebrate community or bird foraging behavior (e.g., weather).

Fecal metabarcoding analyses of diet often reveal wider dietary niches than studies using morphological examination of stomach contents or fecal samples (Gerwing et al. 2016, Ingala et al. 2021). Among previous studies on Purple Sandpiper winter diet in North America, however, our individual dietary niche estimates were narrower. Mallory et al. (2016) collected gut contents from 25 Purple Sandpipers (five individuals each from five locations in Nova Scotia) and identified a total of nine dietary items (congeners grouped into one diet item). Among the ‘core’ dietary items (present in $\geq 10\%$ of all samples) in their study, five were present in our study

(*Littorina*, *Lacuna*, and *Nucella* snails; Chironomidae insects; and Gammarid amphipods) and three were absent (*Mytilus* mussels, *Corallina* algae, and Brachyura crabs). The absence of algae and crabs is not surprising given that the two taxa were not common (each present in 4 out of 25 birds = 16%) in Mallory et al. (2016), and our primers were not designed to amplify autotroph DNA. The mean diet richness of fecal samples in our study (mean: 2.2 ± 1.5 taxonomic groups; range: 1–8 taxonomic groups) was lower than that of gut content samples in Mallory et al. (2016) (mean: 4.8 ± 2.0 diet items; range: 2–9 diet items). A comparison of dietary breadth with Smith and Bleakney (1969) is difficult given that the authors examined the gut contents of only five individuals from two locations in Nova Scotia. Gut contents contained primarily *Littorina* snails, and the only other diet item recorded was small fragments of *Balanus balanoides* (synonymous with *Semibalanus balanoides*).

The total diet breadth in our study (N = 24 taxonomic groups; Table 2.1) was larger than in the two previous studies of Purple Sandpiper winter diet in North America. However, many of the novel diet items we described are small, soft-bodied organisms that occurred in low frequencies, and would likely be missed in gut content analyses. Moreover, our relatively large sample size likely contributed to increased diet breadth. The infrequent occurrence of seven insect taxonomic groups from three orders (Coleoptera, Diptera, Lepidoptera) suggests that Purple Sandpipers opportunistically forage in supratidal freshwater and terrestrial ecosystems at higher tides or that organisms originating from these environments are consumed in the rocky intertidal. Metabarcoding of a congener's (Semipalmated Sandpiper; *Calidris pusilla*) fecal samples from the Bay of Fundy also revealed several unrecorded insect diet taxa that indicates the use of diverse ecosystems (Gerwing et al. 2016). The winter diet of a European population of Purple Sandpipers (N = 37 individuals from eight sites) consisted of > 30 diet items (congeners

grouped) from gut contents, including Diptera associated with supratidal seaweed wrack (Summers et al. 1990a). So while our study may have identified novel diet breadth regionally, other Purple Sandpiper populations are known to have broad winter diets that include low frequencies of taxa found outside the intertidal.

Uncovering a population's total diet breadth can be worthwhile for wildlife managers if novel, low-frequency diet items indicate use of additional habitat types in which survival is different than in core habitats. The occasional use of supratidal habitats that our diet data suggest is unlikely to represent such a scenario. Rather, the most striking aspects of our winter diet study from a management perspective are the dominance of barnacles and the absence of mussels. Barnacles have been recorded in Purple Sandpiper winter diet in both North America and Europe, but they have been reported with relatively low frequency (Smith and Bleakney 1969, McKee 1982, Summers et al. 1990a, Dierschke 1994) or absent altogether (Feare 1966, Mallory et al. 2016). Along rocky intertidal shorelines, barnacles are often common in the high intertidal zone (Benedetti-Cecchi 2001). In some instances, barnacles have been present in Purple Sandpiper diet at higher tidal heights, but absent at lower tidal heights where benthic invertebrate diversity is considerably higher (Dierschke 1994). However, in our study barnacles were a common diet item throughout the tidal cycle (Figure 2.4). In contrast, mussels were not detected in any fecal sample, which is surprising given that the bivalve occurred frequently in many descriptions of winter diet (Feare 1966, Strann and Summers 1990, Summers et al. 1990a, Dierschke 1994, Mallory et al. 2016). It is unclear whether the absence of mussels in diets was the result of non-selection of the prey by Purple Sandpipers, site absence due to normal distributional patterns, or site absence resulting from regional population declines. Other populations of overwintering shorebirds for which *Mytilus* mussels are a primary prey have faced

steep declines of the bivalve as well (Morten et al. 2022). Future research should monitor whether there are parallels in population-level responses to declines in mussels across shorebird populations and species.

The initial use of molecular techniques to describe a species' diet often reveals previously unknown species interactions (e.g., [Riemann et al. 2010](#), [Leray et al. 2015](#), [Gerwing et al. 2016](#), [Granquist et al. 2018](#)). While it is perhaps unsurprising that our study produced a novel result by uncovering a high prevalence of barnacle consumption, it is unclear whether this result is due in larger part to the sensitivity of our methodology or recent shifts in this population's winter diet. Other shorebird species that occur in rocky intertidal habitats—such as Ruddy Turnstones (*Arenaria interpres*) and Black Oystercatchers (*Haematopus bachmani*)—commonly eat barnacles, and individuals have been observed consuming only the soft-bodied parts of barnacles while leaving the outer plates attached to the substrate (Marsh 1983, Robinson et al. 2018). While barnacle plate fragments have been observed in Purple Sandpiper stomachs and feces (Smith and Bleakney 1969, Summers et al. 1990a, Dierschke 1994), and individuals have been observed chipping barnacles from rocks (McKee 1982), barnacles could be underrepresented in morphological analyses of diet if the majority of barnacle predation targets the soft-bodied flesh. Ingestion of plate fragments could be incidental or from an infrequent attack behavior targeting the entire barnacle. Shorebirds must crush hard-shelled prey in their gizzard, and the act of crushing and processing shell waste can be a digestive bottleneck that limits intake rates (Wanink and Zwarts 1985, Van Gils et al. 2005, Ruthrauff et al. 2015).

Alternatively, Purple Sandpipers may consume primarily soft-bodied recruits that settle on rocky substrates in the winter and spring, providing high densities of poorly defended prey (Minchinton and Scheibling 1991, Pineda et al. 2006). During limited foraging observations in

our study, individuals frenetically pecked at the substrate while moving, which is consistent with this hypothesis. Determining the nature of this predator-prey relationship through diet analyses is difficult due to methodological limitations—morphological examinations of gut contents and feces will likely miss small cyprid and juvenile barnacles, and metabarcoding does not provide information on the life stages of identified taxa (Hoenig et al. 2022). Detailed foraging observations with high frame rate video would be especially insightful in addressing this question.

A second interpretation of our results is that there has been a shift in the winter diet of the population that overwinters in the Gulf of Maine. While there is uncertainty as to whether barnacles have been underrepresented in previous diet studies, the conspicuous absence of mussels in our study may be reflective of changes in the prey community in this region. When contextualizing a taxon's absence from a metabarcoding study, it is important to consider whether the study's primers were likely to amplify that taxon's DNA from samples. We believe that it is unlikely that our primers were able to amplify common mollusks such as *Lacuna*, *Littorina*, and *Nucella* snails, but not *Mytilus*. Moreover, our reference library contained many *Mytilus* spp. sequences.

While we lack a replicated study from mid-coast Maine to directly compare our results to, a recent study in Nova Scotia (nearest site to our study locations = 191 km) found mussels in Purple Sandpiper stomachs at all five sampling locations and in 72% of birds (N = 25; Mallory et al. 2016). The taxon has been a common winter diet item in Europe as well (Feare 1966, Strann and Summers 1990, Summers et al. 1990, Dierschke 1994). Changing environmental conditions in the Gulf of Maine—such as warming sea surface temperatures, declining pH, and species invasions—have been linked to declines of intertidal benthic invertebrates over the last two

decades (Petratis and Dudgeon 2020, Ens et al. 2022). In particular, blue mussels (*Mytilus edulis*) have experienced steep declines, with recruitment declining 15.7% per year from 1997–2018 (Petratis and Dudgeon 2020). Other mollusk taxa in this study have also experienced declines in the Gulf of Maine, but to a much lesser degree (*Semibalanus balanoides* recruitment = –5.0% per year, *Nucella lapillus* abundance = –3.8% per year, *Littorina littorea* abundance = –3.2% per year). Faced with widespread reductions in a common winter diet item (mussels), Purple Sandpipers may have increased consumption rates of other abundant diet items (barnacles) to meet their daily energy budgets.

The effects of a shift in winter diet largely depend on the interaction between prey items' caloric value and climatic demands on thermoregulation. The caloric content of dry *Semibalanus balanoides* flesh (N = 3; mean: 4554 cal/g $\pm$ 21.59) is 13% greater than that of dry *Mytilus* spp. flesh (N = 5; mean: 4028 cal/g $\pm$ 27.46) in the Gulf of Maine (Sungail et al. 2013), indicating that barnacles could be adequate for meeting daily energy budgets. However, it is unclear if the energy necessary for processing barnacles is similar to mussels, and if the youngest barnacles life stages on the substrate (e.g., cyprid larvae prior to metamorphosis) contain comparable caloric content to the adult values reported above.

Even if the mean daily caloric intake is lower in the contemporary diet of this Purple Sandpiper population, potential reduction in energy consumed may be offset by lowered metabolic demands due to warming average and minimum winter air temperatures along the Maine coast (Maine Climate Office 2022). This hardy shorebird—which has heavy plumage and carries relatively little fat during the winter compared to other shorebirds—appears to have little difficulty fulfilling daily energy requirements during typical winter conditions (Summers et al. 1992, Dierschke 1994). If Purple Sandpipers often meet their daily energy requirements prior to

high tide, individuals could compensate for small decreases in diet quality by increasing total foraging time.

Our study highlights potential challenges in comparing the first metabarcoding diet study for a species with previous morphological diet studies (e.g., [Soininen et al. 2013](#), [Cuff et al. 2022](#)). In particular, if molecular results differ from those obtained with traditional techniques and the consumer population is experiencing declines, resource managers should consider the relative contribution of methodological and biological processes in producing the novel diet results. The dominance of barnacles in the winter diet of Purple Sandpipers is a novel finding, but given the taxon's occurrence in other studies, we cannot distinguish between changes in methodological bias or a diet shift in a warming region. The absence of mussels—a common diet item in many studies of Purple Sandpiper winter diet that has experienced steep declines in our study area—lends support to the idea that our results are driven, at least partially, by biological rather than methodological processes. Optimizing specific primer sets for identifying marine taxa in shorebird diets is an important next step, as is being done in numerous eDNA studies in different ecosystems ([Forsman et al. 2022](#)). Our study demonstrates that the primers we used can amplify a broad range of marine taxa. However, we also do not necessarily know what taxa are missing due to poor amplification. Primers that better characterize marine invertebrates likely exist and should be evaluated in concert with diet items that have been morphologically identified as well as having been tested with mock communities.

The relationship between a putative dietary shift suggested by our study and the recent declines in overwintering Purple Sandpipers in Maine is intriguing, but unclear. Changes to a population's prey base during the relatively long overwintering period could negatively impact within-season survival or subsequent migratory performance or breeding productivity ([Jansson et](#)

al. 1981, Reudink et al. 2009). Alternatively, as the western North Atlantic Ocean has rapidly warmed in recent decades, there may be a northward shift in this population's winter distribution, which has been documented for numerous shorebird species in Europe (Rehfisch et al. 2004, Austin and Rehfisch 2005). In such cases, local declines may not be reflective of population- and flyway-level trends. Future research on Purple Sandpiper diet in the western North Atlantic should monitor barnacle consumption across years and locations, better understand their net caloric value, and test for relationships between variation in diet and physiological performance of individual sandpipers. Incorporating experimental or observational approaches (e.g., Brun et al. 2022) would also allow for greater understanding of the relationship between barnacle life stages and Purple Sandpiper predation.

Chapter 3

Bed-scale impact and recovery of a commercially important intertidal seaweed*

*Johnston, E. M., H. N. Mittelstaedt, L. A. Braun, J. F. Muhlin, B. J. Olsen, H. M. Webber, A. J. Klemmer (2023). Bed-scale impact and recovery of a commercially important intertidal seaweed. *Journal of Experimental Marine Biology and Ecology* 561:151869.

3.1. Chapter Abstract

As the value of ecosystem-based management (EBM) approaches is increasingly recognized in marine ecosystems, it is critical that the impacts of resource harvest are assessed at various spatial scales. This is particularly true for habitat-forming resources, such as wild seaweeds, that act as foundation species by physically structuring ecosystems. The impacts of spatially heterogeneous harvest may change with scale, and have different management implications based on the ecosystem process or organism under consideration. *Ascophyllum nodosum* (hereafter rockweed) is a canopy-forming fucoid seaweed endemic to rocky coastlines in the North Atlantic Ocean that has been harvested for centuries. We conducted a Before-After Control-Impact study of commercial rockweed harvest at 38 sites across the coast of Maine (USA) from 2018 to 2020 in an effort to understand impact and one-year recovery of two rockweed bed structural characteristics, height and biomass, at a scale similar to a single harvest event. Our results indicate that rockweed harvest is spatially heterogeneous at the scale of the rockweed bed, and as a result, the effect sizes of harvest at this scale are smaller than those reported in previous studies that assessed smaller spatial scales. Mean rockweed biomass recovered to pre-harvest values after one year of recovery, but mean rockweed height remained lower at impacted sites. While post-harvest recovery was generally high in our study, sites that experienced higher intensities of harvest were less likely to fully recover height or biomass one

year post-harvest. Our findings provide resource managers with a bed-scale perspective that can inform EBM approaches, particularly for population-level management of harvested resources and impacts of harvest on highly mobile organisms—such as birds and fish—that interact with these ecosystems at larger spatial scales.

3.2. Introduction

The harvest and recovery of seaweeds (macroalgae) has been studied widely across littoral zones and latitudes (Keser et al., 1981; Mafra Jr. and Cunha, 2006; Ulaski et al., 2020; Westermeier et al., 2019). There is high interest in seaweeds' response to harvest given their ecological role as foundation species (Dudgeon and Petraitis 2005, Schmidt et al. 2011) and economic value for coastal communities (Rebours et al. 2014). Many seaweed species exhibit relatively fast growth rates (Mafra Jr. and Cunha 2006, Reed et al. 2008), and the ability of some to recover biomass a few years after harvest is often highlighted in examples of sustainable fisheries (e.g., Marquez et al., 2014; Vea and Ask, 2011). However, there has been a paradigm shift towards ecosystem-based management (EBM) in recent decades—particularly in marine ecosystems—as resource managers recognize the importance of maintaining not only the population sizes of targeted resources, but also trophic interactions throughout associated food webs (Pikitch et al. 2004, Arkema et al. 2006).

Assessing the food-web impacts of commercial seaweed harvest requires that the scale of inquiry overlaps with the scales of both human harvest and the food web itself. The proportion of harvested individuals can change across spatial scales and result in variable impacts to habitat quality depending on the home range size and movement type of the organism under consideration (Grindal and Brigham 1999, Leonard et al. 2008). Along many North Atlantic

rocky intertidal zones, *Ascophyllum nodosum* (hereafter rockweed)—a canopy-forming fucoid seaweed—dominates the intertidal seaweed assemblage (Vadas et al. 2004, Guiry and Morrison 2013). Rockweed has experienced a rise in global harvest over the past several decades and, as a result, there have been concerns about the ecological sustainability of harvesting practices (Seeley and Schlesinger 2012, Lotze et al. 2019). Some jurisdictions report rockweed harvest by total landed weight or percent biomass removed across a landscape-scale area on the order of tens of square kilometers (i.e., sector; Ugarte and Sharp, 2012), yet these metrics lack context at the site-level scale over which a single harvest event occurs and some highly mobile organisms—such as birds and fish—utilize the ecosystem. Furthermore, impacts associated with small, intensely harvested patches (e.g., several m²; Walder, 2015) may be different than those associated with rockweed beds that experience spatially heterogeneous harvest typical of commercial practices (DFO, 1999). Quantifying harvest impacts at the harvest-site scale (i.e., hundreds to thousands of square meters) will allow EBM approaches to simultaneously assess population-level management of the harvested resource and the impacts of commercial harvest on mobile organisms that interact with these ecosystems at relatively large spatial scales.

In addition to assessing harvest impacts at different spatial scales, resource managers must consider the representativeness of research protocols to commercial harvest in their jurisdiction. For instance, some studies use harvest intensities that cut rockweed fronds below the legal harvest height in the area of study (Keser et al., 1981; Black and Miller, 1991; Fegley, 2001). Studies may also create uniformly harvested treatment plots with hand shears that are not typical of patchy commercial harvest (Keser et al., 1981; Fegley, 2001; Walder, 2015). Recovery thresholds may be crossed at these high harvest intensities, but species may show greater resilience at lower harvest intensities (Keser et al. 1981). As a result, harvest regulations

informed by research that lacks representativeness to commercial practices can lead to uncertainty in the outcomes of management decisions.

Our objective was to design an experiment to assess the effects of commercial rockweed harvest by measuring impact and recovery at the same spatial scale as the harvest. We conducted a Before-After Control-Impact (BACI) experiment using commercial rockweed harvest practices at 38 sites across the coast of Maine (USA; Figure 3.1). To assess impact and recovery, we measured mean bed height and biomass during three time periods: pre-harvest (2018), harvested (2019), and one year post-harvest (2020). Given the large site sizes in our study and the limitations of sampling their full spatial extent, we use a two-part approach in our analysis. First, we tested whether our sampling methodology detected harvest at impact sites relative to control sites. Second—contingent on detecting harvest—we quantified the impact and recovery of rockweed height and biomass at the rockweed bed scale. We hypothesized that harvest impacts to both rockweed characteristics will be spatially heterogeneous at a site, resulting in harvest impacts that are detectable at the site scale, but of smaller effect size than those reported in previous studies with smaller treatment plot sizes. Additionally, we hypothesized that sites with greater harvest intensity (i.e., biomass loss) will show less recovery one year later. We define recovery as a return to pre-harvest baseline values in height and biomass. While this approach defines impact and recovery strictly by these two morphological characteristics of the target resource, and does not consider any direct or indirect impacts on other species in the ecosystem, our study provides a harvest-scale perspective that can inform EBM recommendations in rockweed ecosystems.

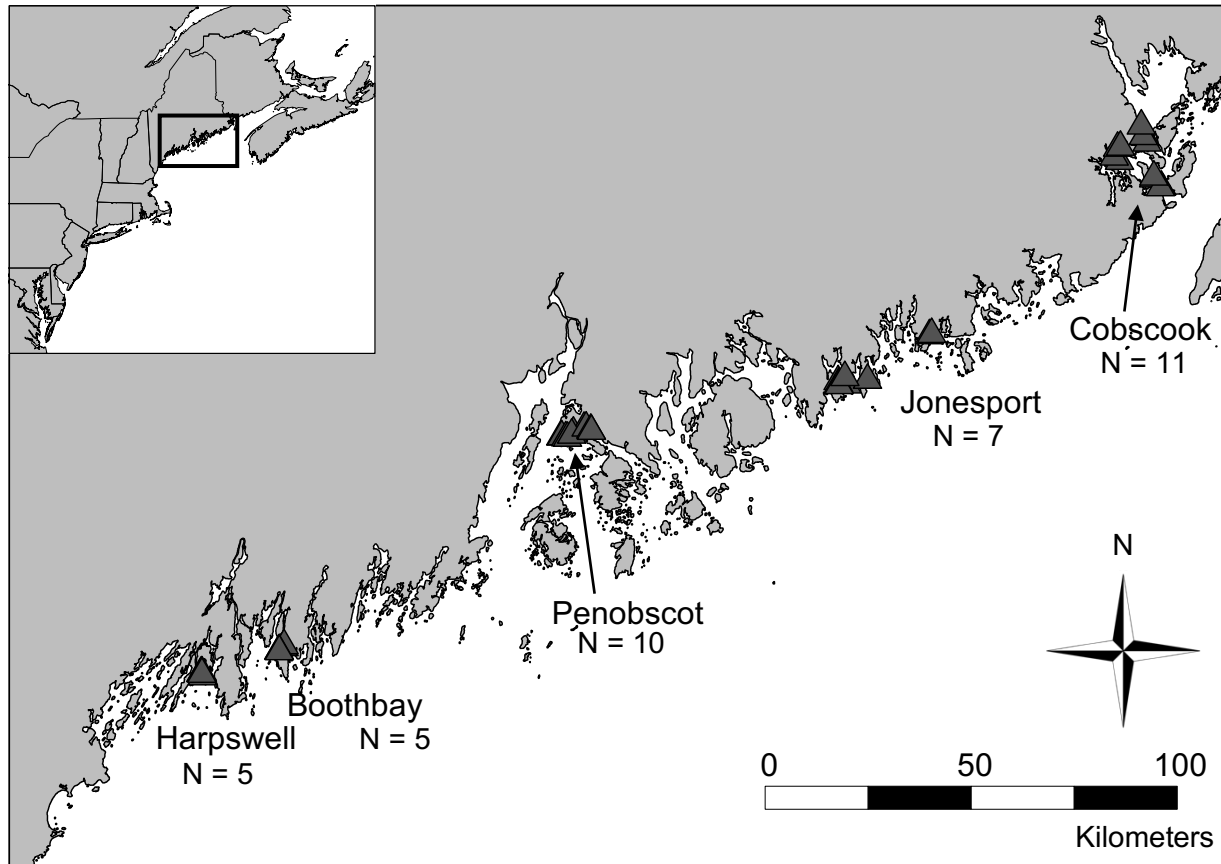


Figure 3.1. The Before-After Control-Impact (BACI) experiment conducted at 38 sites across five regions. Regional treatment totals were as follows: Harpswell 2 control, 3 impact; Boothbay 2 control, 3 impact; Penobscot 4 control, 6 impact; Jonesport 4 control, 3 impact; Cobscook 7 control, 4 impact. Treatments are not visually differentiated due to high symbol overlap at the current scale. Harpswell and Boothbay were combined into a single “Midcoast” region during analyses to better balance sample sizes across regions.

3.3. Methods

3.3.1. Study Design

The BACI study design (Stewart-Oaten et al. 1986) is commonly used to assess the impacts of disturbance, and has been frequently used to assess the impacts of rockweed harvest (e.g., Kay, 2015; Kelly et al., 2001; Trott and Larsen, 2012; Ugarte et al., 2006; Walder, 2015). BACI experiments allow for strong causal inference—spatial replication (control vs. impact)

separates the effects of natural environmental variation and experimental treatment, while temporal replication (before vs. after) assesses whether post-disturbance differences between control and impact groups were present pre-disturbance. We used a repeated-measures BACI design (Green 1993) with rockweed surveys at each site before harvest (August–October 2018), immediately after harvest (August–December 2019), and one year after harvest (August–November 2020). Our annual survey window was informed by typical harvest schedules and avoided biomass assessment during the late spring reproduction of rockweed when reproductive receptacles can increase seaweed weight considerably (Vadas et al. 2004).

3.3.2. Site Selection and Rockweed Harvest

One of our primary objectives was to maximize the representativeness of rockweed harvest to commercial practices in our region (Maine). Therefore, we worked with several rockweed harvest companies—Acadian Seaplants (N = 28 sites), North American Kelp (N = 5 sites), and Source Incorporated (N = 5 sites)—and private landowners to establish 38 sites along the Maine coast. Based on their own criteria, each company defined their geographic boundaries of harvestable shoreline within which research sites could be established, resulting in five study regions—Harpwell, Boothbay, Penobscot, Jonesport, and Cobscook (Figure 3.1). We initially began the study with another company using knife-harvesting in the mid-coast region, but after initial efforts resulted in only two viable sites (one treatment, one control) we did not have enough replication to control for either regional or harvest method differences and these two sites were removed from the study. Within each company boundary, we used Google Earth satellite imagery to identify potential sites that a) were non-cliff rocky intertidal habitat and b) had extensive rockweed beds suitable for commercial-scale harvest. Additionally, we worked with

harvesters to select candidate sites that were not harvested within at least the past three years. Sites were located either on the mainland or near-shore islands to ensure harvest and survey accessibility. Typical substrata included bedrock and boulders.

From the pool of candidate sites, we first selected sites for which we received landowner permission to access the intertidal. We next selected sites until we reached the maximum number of sites a company could harvest given their anticipated availability of harvesters and equipment, or until our pool of candidate sites was exhausted. While harvest companies defined the candidate patch boundaries—providing direct application to industrial practices—we assigned treatment type (control or impact) to each site. Landowner permissions to harvest rockweed prevented a fully spatially random treatment assignment, but we ensured that there was a spatial mixture of control ($N = 19$) and impact ($N = 19$) sites within each of the five predefined regions. In addition, we attempted to ensure that there was no geomorphological bias between treatment type (e.g., a mixture of coastline features such as points, coves, etc.). Sites were vertically bounded by the high and low tide lines, and measured 100 m horizontally (parallel to the shoreline), a size representative of the scale at which commercial harvesters operate (A. Feibel, personal communication). We define a rockweed bed at the scale of the harvest event in this study (i.e., a study site), but we recognize that these beds are often part of larger ecologically contiguous units.

Control sites were unharvested throughout our study, and all impact sites were harvested once between June and November 2019 by commercial harvesters under current Maine regulations (40.6 cm minimum cutting height). We provided companies with Global Positioning System (GPS) coordinates of site boundaries, and instructed companies to harvest as they would during commercial operations. Rockweed harvest occurred by two methods: mechanical

harvester vessel (N = 13) and hand rake (N = 6; see Rockweed Plan Development Team et al. 2014 for details on each method). We personally observed multiple site harvests with each company (N = 7) to confirm that their practices aligned with typical harvest practices. In addition, harvest companies reported total seaweed harvest landings from our study sites when possible.

3.3.3. Rockweed Height and Biomass Assessment

Rockweed height and biomass assessment allowed us to measure variation in both harvest intensity (at our impact sites) and in natural sources of variation (at all sites), such as rockweed growth and frond breakage due to storms. We conducted rockweed assessments once per site during each time period—pre-harvest (2018), harvested (2019), and one year post-harvest (2020). Our methodology was adapted from the Maine Department of Marine Resources' Fishery Management Plan for Rockweed (Rockweed Plan Development Team et al., 2014). During low tide, we haphazardly placed two 10-meter transects in separate areas of the rockweed bed (> 20 m apart), parallel to the shoreline. Transects were placed left- and right-of-center at a middle tidal height, which is the most productive zone for rockweed and where most harvest takes place (Stengel and Dring, 1997; A. Feibel, personal communication). We marked transect locations with Garmin GPS handheld units and placed each transect in the same location during each survey year (GPS manufacturer reported error of ≤ 3 m).

Along each 10-m transect, five 0.5 x 0.5 m quadrats (0.25 m²) were sampled every two meters, placed along alternating sides of the transect. At each quadrat, we used a three-step process to assess rockweed structural characteristics. First, we counted the number of holdfasts (the part of the rockweed individual that attaches to the substrate). Holdfasts, and therefore

rockweed individuals, were considered separate if they were > 0.5 cm apart (Kay et al., 2016). Our surveys included holdfast count to allow us to assess variation in rockweed density between time periods.

Second, we haphazardly selected three rockweed individuals and measured their length from the holdfast to the tip of the longest frond. If the longest frond significantly overestimated canopy height (e.g., a single outlier frond), we measured the length of the tallest frond in the group that represented the top of the canopy. Our haphazard selection of rockweed individuals could have been biased toward longer algae that were more conspicuous, which would result in mean height values more representative of canopy structure than all individuals within a site. However, we would not expect this potential bias to change across treatments or time periods (i.e., affect inferences in our study).

Third, we placed all rockweed from holdfasts within the quadrat into a mesh bag and weighed it with a spring scale. We left several inches between the holdfast and the bottom of the cinched mesh bag to allow slack for weighing and to prevent damage to the rockweed individuals. Small individuals that had holdfasts < 2 cm in diameter with fronds that did not exceed 30 cm were excluded from rockweed surveys. All sampling was non-destructive in an effort to isolate changes in rockweed characteristics to either natural variation or harvest, and not from sampling methods.

Commercial rockweed harvest is not homogeneous across the bed and leads to variation in local biomass removal (Rangeley and Davies 2000, Ugarte et al. 2006). The primary objective of our sampling methodology was to estimate height and biomass at the site scale. We did not ensure that there was perfect spatial overlap between harvest and either our quadrats or our transects, and harvesters were unaware of the location of sampling within each site. We expected

that conventional commercial practices would result in a spatially heterogeneous harvest that would only partially overlap with our precise sample locations, and thus our measurements would provide us with a representative assessment of harvest impact at the site, but not the holdfast, scale. Importantly, our design was not intended to precisely estimate biomass removed from the site, but to capture typical within-site variability. This approach allowed us to estimate average changes in height and biomass across a given site, and to provide an informative, unbiased average of harvest impact across our 19 impact sites, incorporating both among- and within-site variation.

3.3.4. Statistical Analysis

All statistical analyses were performed in program R version 4.1.2 (R Core Team, 2022). Regional differences in pre-harvest rockweed height and biomass were analyzed using linear mixed models (LMMs) in the lme4 package (Bates et al. 2015). We included region as a fixed effect and site, transect, and quadrat as hierarchical sets of nested random intercepts to account for spatial non-independence.

Tests of the impacts of rockweed harvest on height and biomass were divided into three separate analyses. Transects and sites were the experimental units in these analyses and we therefore calculated mean height and biomass values over five and ten quadrats, respectively, at each site. First, we assessed whether our sampling methodology detected rockweed harvest at impact sites relative to control sites. We performed contingency χ^2 analyses to examine the relationship between treatment type and the number of transects at a site that declined in mean rockweed height and biomass between the pre-harvested and harvested time periods. Second, we tested the impact of rockweed harvest across all study sites with LMMs that had two main effects

and their interaction: treatment (control or impact) and time period (pre-harvest, harvested, one year post-harvest). To quantify the impact of harvest, we calculated standardized and unstandardized effect sizes, respectively,

$$\ln\left(\frac{\bar{X}_{\text{Harv,I}}}{\bar{X}_{\text{Harv,C}}}\right) - \ln\left(\frac{\bar{X}_{\text{Pre,I}}}{\bar{X}_{\text{Pre,C}}}\right) \quad (\text{Eq. 3.1})$$

$$| \bar{X}_{\text{Harv, I}} - \bar{X}_{\text{Pre, I}} | + | \bar{X}_{\text{Harv, C}} - \bar{X}_{\text{Pre, C}} | \quad (\text{Eq. 3.2})$$

where $\bar{X}_{ij}$ equals the estimated marginal mean $\bar{X}$ during time period i (Pre = pre-harvest, Harv = harvested) at sites with treatment type j (I = impact, C = control; Rosenberg et al., 2013). Reporting both metrics allowed us to make comparisons with other studies (standardized) while also providing context within the current study (unstandardized). For each effect size, we calculated reduced bias-corrected bootstrap 95% confidence intervals (Tibbe and Montoya 2022). Third, we tested whether the impact of rockweed harvest varied by study region with LMMs that included a three-way interaction between treatment, time period, and region.

In the LMMs for all three analyses, we included a random intercept of site nested within region in order to account for repeated sampling at sites grouped by region. Models were assessed for homoscedasticity using plots of model residuals versus fitted values and for normality using Q-Q plots. Kenward-Roger approximations for degrees of freedom were used to calculate P values using the lmerTest package (Kuznetsova et al. 2017). One degree of freedom planned contrasts were used to investigate specific hypotheses using the emmeans package (Lenth et al., 2020). Throughout the analyses we combined Harpswell (N = 5 sites) and Boothbay (N = 5 sites)—which are geographically proximate—into a single “Midcoast” region in order to better balance sample sizes across regions.

We also examined the relationship between harvest intensity—measured as the change in mean rockweed biomass at a site over the first time interval (pre-harvest to harvested)—and the a) amount of post-harvest growth and b) recovery of each rockweed characteristic. Post-harvest growth was defined as the absolute amount of change in either height or biomass during the second time interval (harvested to one year post-harvest), while recovery was defined as the percent of the baseline (pre-harvest) value observed at the end of our study (one year post-harvest). We used linear regressions with a single explanatory variable (harvest intensity) to assess these relationships.

During preliminary analyses, we removed four statistical outliers (i.e., ≥ 3 SD from the mean) from final analyses (Figure C.1). First, we removed the biomass value from one quadrat and the height value from one alga that were 6.73 kg 0.25m⁻² heavier and 68 cm longer than the next highest values, respectively. Second, we removed one Jonesport impact site with a mean biomass value 4.1 SD above the mean in the pre-harvest time period. The site with the next most extreme biomass value in the first time period was 1.6 SD away from the mean. We also removed one Cobscook impact site where we recorded 373% more holdfasts between the pre-harvest and harvested time periods, which corresponded to a 166% increase in biomass. This change in holdfasts was 6.1 SD above the mean, with the next most extreme value 2.1 SD away from the mean. Each of these decisions was motivated by the fact that our transects were unlikely to measure the exact same rockweed individuals during each time period, and, therefore, it was important that we identified extreme sampling outliers that resulted from our sampling design.

3.4. Results

Mean holdfast count increased slightly at each consecutive time period, but the treatment $\times$ time period interaction was not statistically significant ($F_{2,72} = 0.37$, $P = 0.69$), indicating that changes in rockweed densities would not impact the inferences in our study. In addition, there was no statistically significant effect of harvest observation on the change in rockweed biomass between the pre-harvest and harvested time periods ($F_{1,17} = 1.78$, $P = 0.20$).

3.4.1. Pre-harvest: Regional Comparison

Mean rockweed height and biomass were relatively homogeneous across the coast of Maine. Thirty-eight out of 114 rockweed assessments occurred in 2018 before harvest, providing a coastwide comparison of rockweed characteristics exclusive of the short-term impacts of rockweed harvest. Given that commercial-scale rockweed harvest has occurred in Maine since the 1970s, inherent in our regional ‘unharvested’ comparisons are variable harvest histories across the coast. Mean rockweed height did not differ significantly between regions, with means ($\pm$ SE) ranging from 89.6 ± 8.0 cm (Harpwell) to 113.5 ± 8.0 cm (Boothbay) ($F_{4,32.89} = 1.45$, $P = 0.24$; Figure 3.2A). Similarly, mean rockweed biomass was not significantly different between regions, with means ($\pm$ SE) ranging from 8.3 ± 1.4 kg m⁻² (Harpwell) to 12.7 ± 1.1 kg m⁻² (Jonesport) ($F_{4,33} = 1.26$, $P = 0.31$; Figure 3.2B). The variances of nested random intercepts indicate that within-group variation in rockweed height increases monotonically from the quadrat to site spatial scale, while for biomass the smallest variation occurs at the site scale (Table C.1).

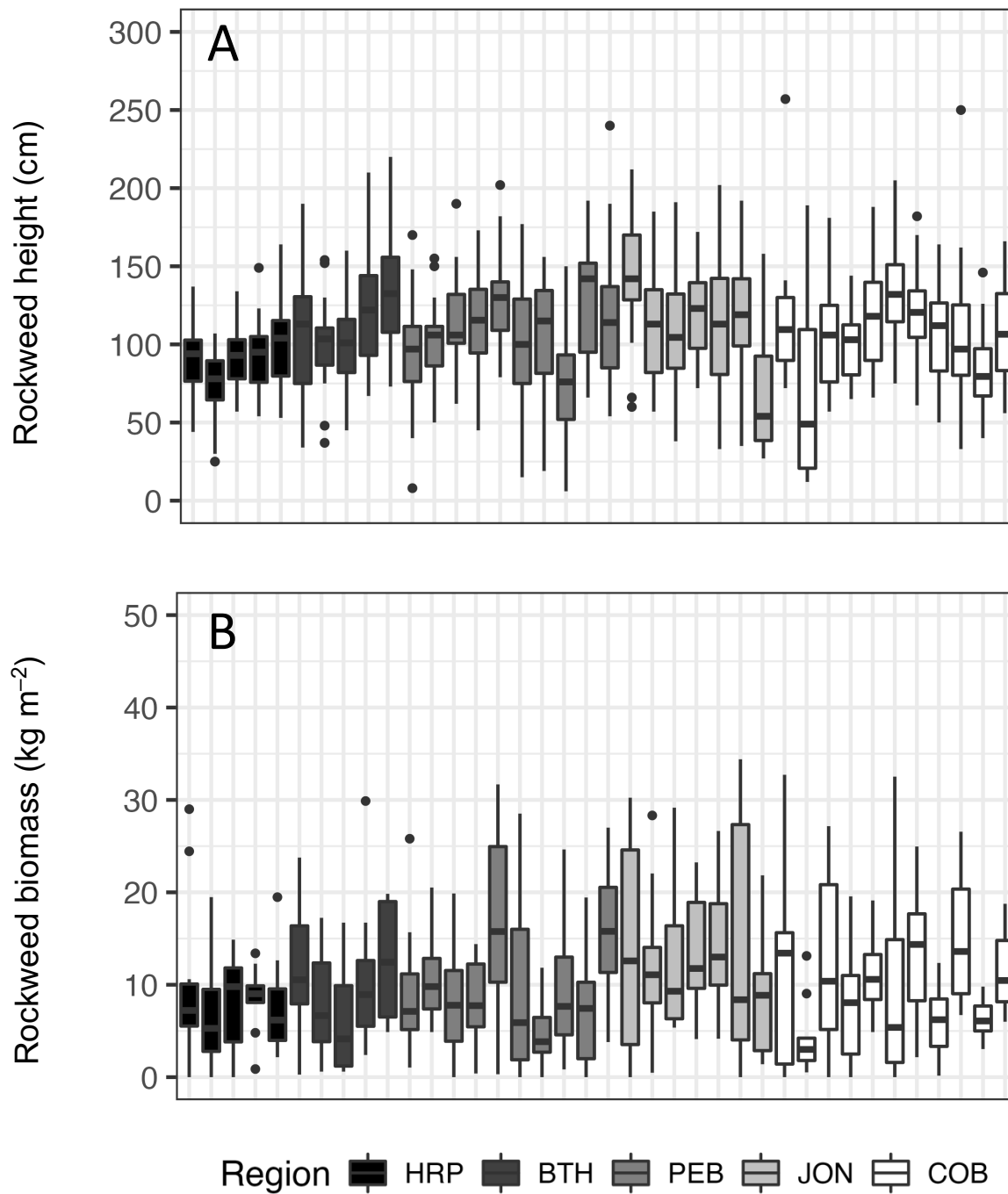


Figure 3.2. Regional comparisons of (A) mean rockweed height and (B) biomass at 38 sites across five regions. For each site, boxplots are comprised of 30 values for height and 10 values for biomass. All differences between region group means are non-significant ($\alpha = 0.05$) for both rockweed characteristics. Only surveys from the pre-harvest time period at control and impact sites are included. Regional abbreviations are as follows: HRP = Harpswell, BTH = Boothbay, PEB = Penobscot, JON = Jonesport, and COB = Cobscook.

3.4.2. Pre-harvest to Harvested: Impact of Harvest

Across several analyses assessing the impacts of harvest—described below—there was marginal statistical support for an effect of treatment ($0.10 > P > 0.01$). The effect of treatment on the number of transects that declined in mean rockweed height or biomass between the pre-harvest and harvested time periods was near the alpha level (0.05) in our study for both height ($\chi^2 = 5.42, P = 0.07$) and biomass ($\chi^2 = 7.25, P = 0.03$). For each bed characteristic, both transects declined more often at impact sites (height: $N = 7$ impact and 4 control; biomass: $N = 8$ impact and 1 control), while neither transect declined more often at control sites (height: $N = 4$ impact and 11 control; biomass: $N = 5$ impact and 7 control). The spatially heterogeneous patterns of commercial harvest and natural disturbance were apparent (Figure C.3), with only one transect declining in mean height and biomass at several control and impact sites (Table 3.1).

The treatment $\times$ time period interaction was close to the alpha level in our study for both mean height ($F_{2, 72} = 2.63, P = 0.08$) and biomass ($F_{2, 72} = 3.25, P = 0.04$; Table 3.2). Planned contrasts indicated marginal support for greater declines in mean height and biomass at impact sites than control sites (height, $t_{1, 72} = -1.91, P = 0.06$; biomass, $t_{1, 72} = -2.21, P = 0.03$; Table 3.2, Figure 3.3). The standardized effect size of harvest on mean biomass (-0.28, 95% CI = -0.58 to 0.003) was more than double the effect size on mean height (-0.11, 95% CI = -0.25 to 0.04; Table 3.3). These magnitudes of impact equated to unstandardized effect sizes of -11.9 cm (95% CI = -27.3 to 3.8 cm) for height and -2.97 kg/m² (95% CI = -6.03 to 0.08 kg/m²) for biomass. For both rockweed characteristics, the mean increase at control sites (height = 8.5 cm, biomass = 2.08 kg/m²) contributed more than the mean decrease at impact sites (height = -3.4 cm, biomass = -0.89 kg/m²) toward the effect sizes. Lastly, the impact of harvest varied by region for both

rockweed characteristics (Figure 3.4). Midcoast was the only region to experience statistically significant declines in rockweed height or biomass following harvest (height, $t_{1,60} = -2.26$, $P = 0.03$; biomass, $t_{1,60} = -3.03$, $P = 0.004$; Table C.2, Figure C.2).

3.4.3. Harvested to One Year Post-harvest: Regrowth After Harvest

Mean rockweed height increased slightly during the post-harvest interval at both control and impact sites (3.2 and 2.2 cm, respectively; $t_{1,72} = -0.15$, $P = 0.88$; Figure 3.3C). In contrast to height, the post-harvest increase in mean rockweed biomass was significantly greater at impact sites than at control sites (3.28 and 0.31 kg/m², respectively; $t_{1,72} = 2.21$, $P = 0.03$; Figure 3.3F). As a result, the standardized effect size of harvest on regrowth was higher for biomass (0.27, 95% CI = -0.003 to 0.55) than for height (-0.007, 95% CI = -0.15 to 0.13).

Across regions, post-harvest changes in mean height were small and there were no statistically significant effects of treatment. Increases in biomass were greater at impact sites than at control sites in all four regions, but none of these treatment differences were statistically significant (Midcoast: $t_{1,60} = 1.12$, $P = 0.27$, Penobscot: $t_{1,60} = 1.17$, $P = 0.25$, Jonesport: $t_{1,60} = 0.47$, $P = 0.64$, Cobscook: $t_{1,60} = 1.50$, $P = 0.14$; Figure 3.4).

3.4.4. Pre-harvest to One Year Post-harvest: Recovery from Harvest

At the end of the study, mean height at impact sites (107.7 ± 4.3 cm) remained below mean height at control sites (115.4 ± 4.3 cm), indicating a lack of recovery to pre-harvest values ($t_{1,72} = -2.05$, $P = 0.04$; Figure 3.3C). Less than half of the impact sites (9/19 = 47%) showed complete recovery in rockweed height (range: 67.1 – 177.9%; mean $\pm$ SD: $102.5 \pm 27.8\%$), but most of the sites that exhibited partial recovery (7/10 = 70%) were within 80% of original height

values. In contrast to height, biomass exhibited a complete recovery on average across all sites ($t_{1, 72} = -0.003$, $P = 1.00$; Figure 3.3F). Furthermore, the majority of impact sites (14/19 = 74%) showed complete recovery in rockweed biomass and many sites exceeded pre-harvest values (range: 51.6 – 201.1%; mean $\pm$ SD: $130.5 \pm 38.8\%$).

Recovery varied across regions for both rockweed characteristics. In Midcoast, mean height at impact sites remained lower than at control sites after one year of recovery ($t_{1, 60} = -3.20$, $P = 0.002$; Figure 3.4A). Mean biomass also remained lower at impact sites in this region, but there was less statistical support for a difference between treatments ($t_{1, 60} = -1.92$, $P = 0.06$; Figure 3.4B). In the other three regions (Penobscot, Jonesport, and Cobscook), there were no statistically significant effects of treatment on recovery for either rockweed characteristic (Table C.2).

3.4.5. Harvest Intensity Gradient

There was no effect of harvest intensity—defined as the change in mean rockweed biomass between pre-harvest and harvested time periods—on the post-harvest growth of rockweed height ($P = 0.74$, $R^2 = 0.007$; Figure 3.5A). In contrast, there was a positive effect of biomass change on height recovery ($P = 0.006$, $R^2 = 0.36$; Figure 3.5B). In other words, sites that experienced greater harvest intensities were less likely to return to pre-harvest values of rockweed height by the end of the study. Unlike height, post-harvest growth of biomass tended to be higher at sites that experienced higher harvest intensities ($P = 0.01$, $R^2 = 0.33$; Figure 3.5C). However, there was also a positive effect of biomass change on biomass recovery ($P = 0.006$, $R^2 = 0.37$; Figure 3.5D), indicating that sites that experienced greater harvest intensities were less likely to return to pre-harvest values of rockweed biomass after one year of recovery.

Table 3.1. Contingency tables assessing the relationship between treatment and changes in rockweed height and biomass between the pre-harvest and harvested time periods.

Rockweed Characteristic			
Treatment	Number of Transects With Mean Decline in Rockweed Characteristic (Pre-harvest to Harvested)		
Height			
	Zero	One	Two
Control	11	4	4
Impact	4	8	7
		χ^2	5.42
		df	2
		P	0.07
Biomass			
	Zero	One	Two
Control	7	11	1
Impact	5	6	8
		χ^2	7.25
		df	2
		P	0.03

Table 3.2. Results from the linear mixed-effects models testing the impact of rockweed harvest on rockweed height and biomass at the scale of the rockweed bed. The significance levels ($P \leq 0.05$ in bold) are calculated from F -statistics of type III hypothesis tests, based on Kenward–Roger approximations for degrees of freedom. Marginal R^2 considers only the variance of the fixed effects, while conditional R^2 considers the variance of both fixed and random effects. The planned contrasts represent the effect of treatment on (top to bottom) harvest impacts, post-harvest growth, and one-year recovery, respectively. Abbreviations are as follows: Pre = Pre-harvest, Harv = Harvested, Yr Post = One Year Post-harvest, I = Impact, and C = Control.

Rockweed characteristic	Source of variation					
Height	<i>Fixed effect</i>	Variance	SD	<i>df</i>	<i>F</i>	<i>P</i>
	Treatment			1, 35.04	0.44	0.51
	Time Period			2, 72	1.39	0.25
	Treatment x Time Period			2, 72	2.63	0.08
	<i>Random effect</i>					
	Region	6.53	2.56			
	Region(Site)	125.61	11.21			
	Residual	185.80	13.63			
	<i>Planned contrast</i>	Estimate	SE	<i>df</i>	<i>t</i>	<i>P</i>
	$(\bar{X}_{\text{Harv, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Harv, C}} - \bar{X}_{\text{Pre, C}})$	-11.93	6.25	1, 72	-1.91	0.06
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Harv, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Harv, C}})$	-0.91	6.25	1, 72	-0.15	0.88
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Pre, C}})$	-12.84	6.25	1, 72	-2.05	0.04
Marginal R^2 : 0.05, Conditional R^2 : 0.44						
Biomass	<i>Fixed effect</i>	Variance	SD	<i>df</i>	<i>F</i>	<i>P</i>
	Treatment			1, 34.26	0.22	0.64
	Time Period			2, 72	6.82	0.002
	Treatment x Time Period			2, 72	3.25	0.04
	<i>Random effect</i>					
	Region	0.81	0.90			
	Region(Site)	3.12	1.77			
	Residual	8.62	2.94			
	<i>Planned contrast</i>	Estimate	SE	<i>df</i>	<i>t</i>	<i>P</i>
	$(\bar{X}_{\text{Harv, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Harv, C}} - \bar{X}_{\text{Pre, C}})$	-2.98	1.35	1, 72	-2.21	0.03

Table 3.2 Continued

$(\bar{X}_{Yr Post, I} - \bar{X}_{Harv, I}) \neq (\bar{X}_{Yr Post, C} - \bar{X}_{Harv, C})$	2.97	1.35	1, 72	2.21	0.03
$(\bar{X}_{Yr Post, I} - \bar{X}_{Pre, I}) \neq (\bar{X}_{Yr Post, C} - \bar{X}_{Pre, C})$	-0.004	1.35	1, 72	-0.003	1.00

Marginal R^2 : 0.11, Conditional R^2 : 0.39

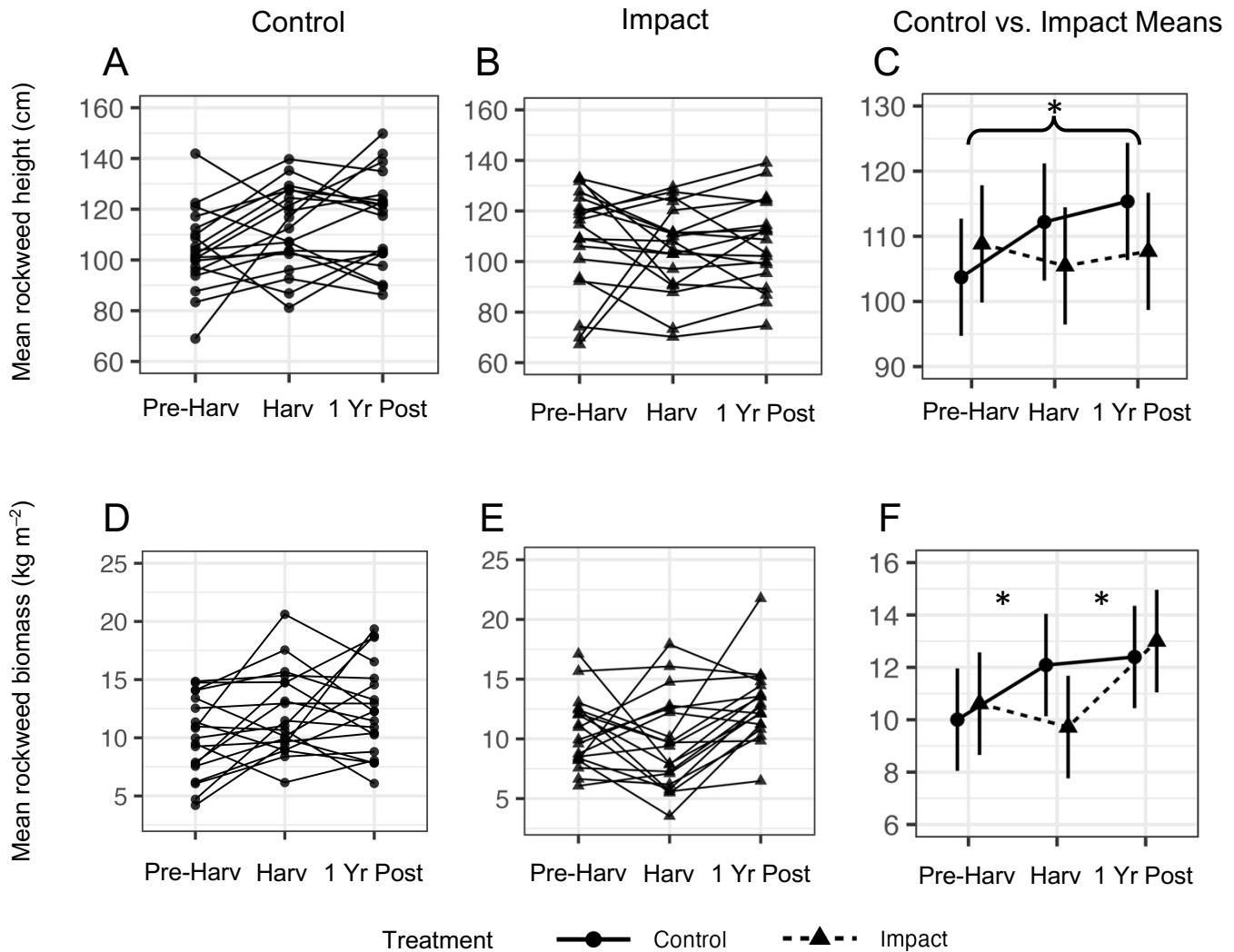


Figure 3.3. Change in mean rockweed height and biomass across three time periods. Each point in panels A, B, D, and E represents a mean site value, and a line connects a given site through time. Error bars for each point are not shown in these plots to aid visual clarity. In panels C and F, points represent treatment means $\pm$ 95% confidence intervals. Asterisks indicate statistically significant differences ($\alpha = 0.05$) in the change in height or biomass between treatments for a given time interval. Pre-Harv = Pre-harvest, Harv = Harvested, and 1 Yr Post = One Year Post-Harvest.

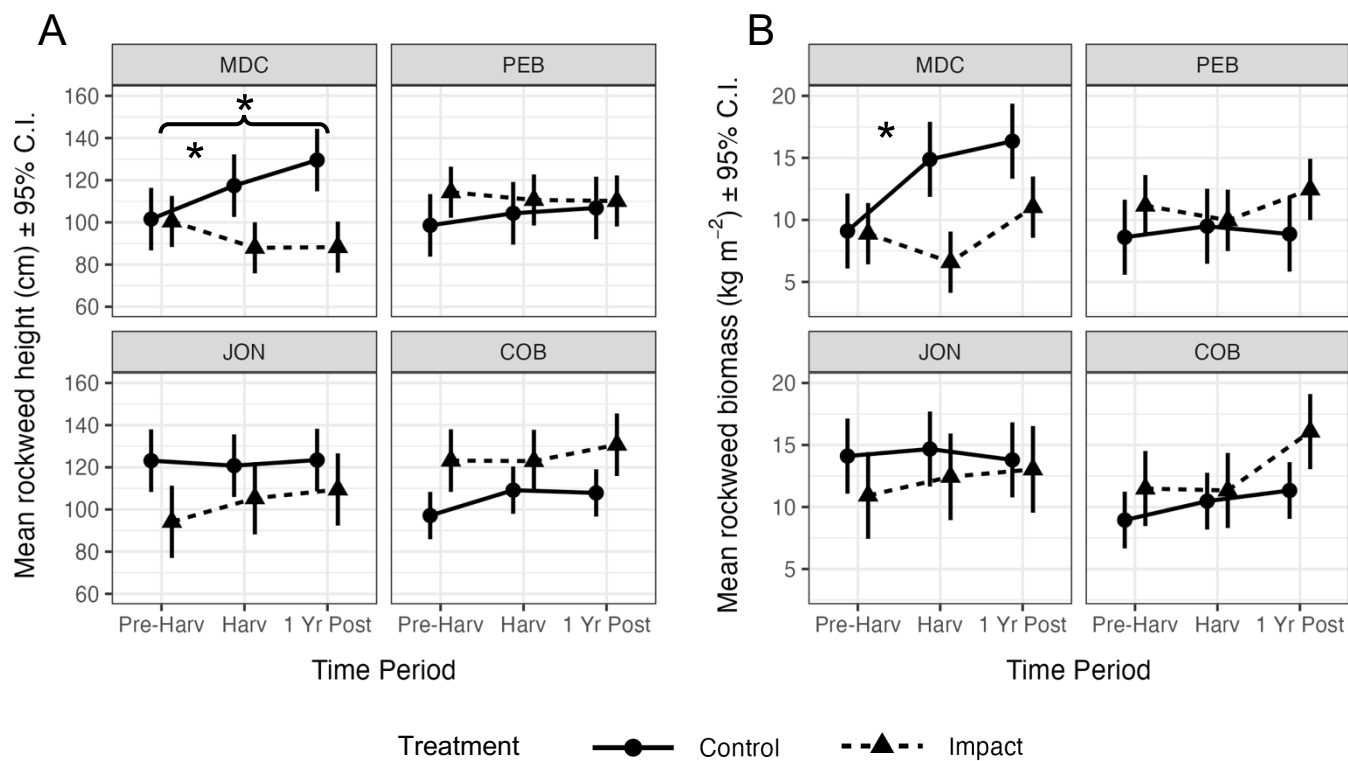


Figure 3.4. Change in mean (A) rockweed height and (B) biomass across three time periods in each study region. Asterisks indicate statistically significant differences ($\alpha = 0.05$) in the change in height or biomass between treatments for a given time interval. Abbreviations are as follows: Pre-Harv = Pre-harvest, Harv = Harvested, 1 Yr Post = One Year Post-harvest, MDC = Midcoast, PEB = Penobscot, JON = Jonesport, and COB = Cobscook.

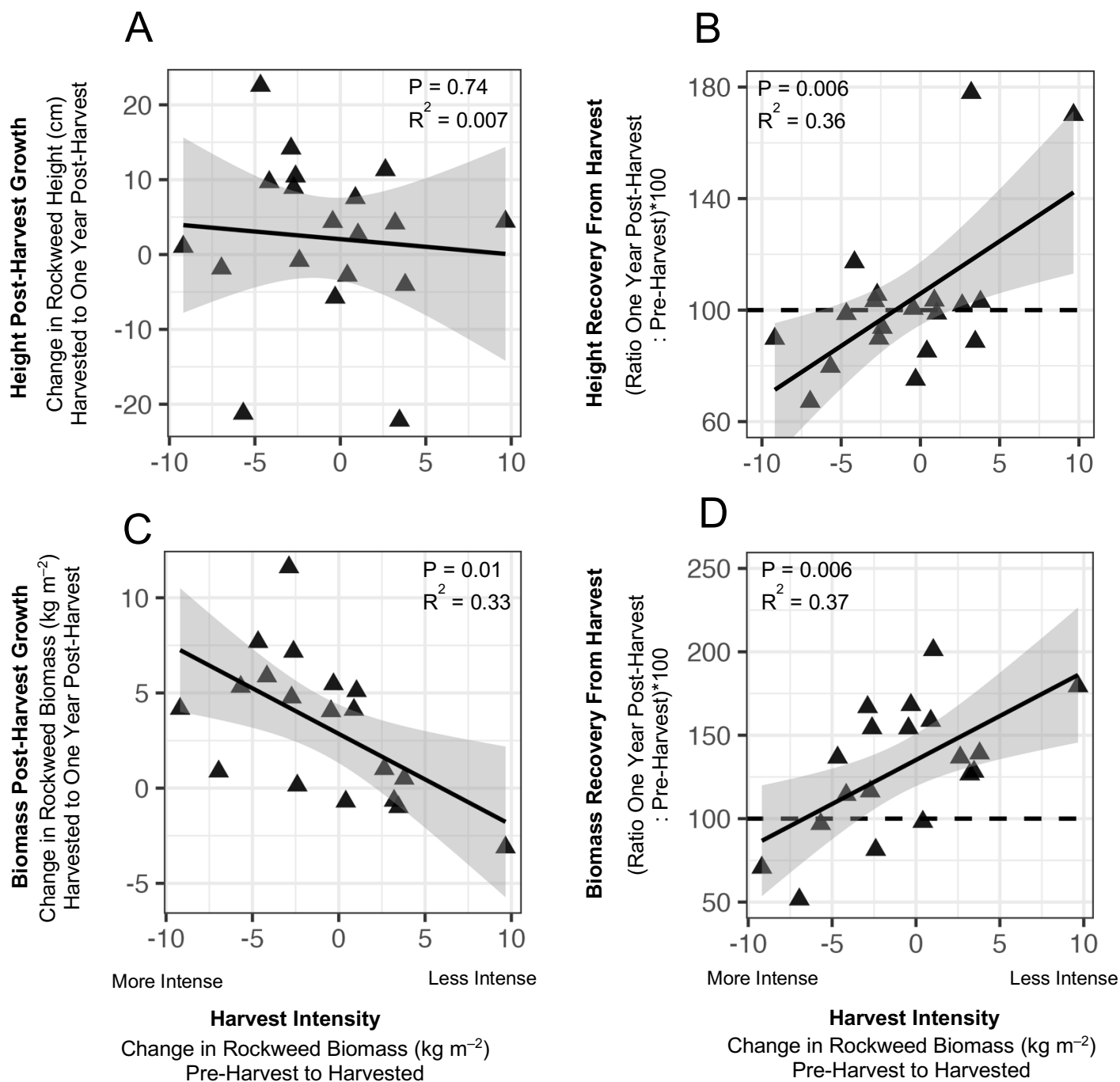


Figure 3.5. Post-harvest growth rate and one-year recovery of rockweed (A, B) height and (C, D) biomass across a gradient of harvest intensity. Only impact sites are displayed. Dashed horizontal lines in panels B and D indicate 100% recovery to pre-harvest values. The gray shading around the regression lines represents the 95% confidence interval.

Table 3.3. Review of studies assessing the impact of rockweed harvest on rockweed height and biomass. Standardized and unstandardized effect sizes are calculated when possible (formulas provided in table note), and main results are provided otherwise. Some effect sizes were calculated from study figures with ImageJ. Studies with complete rockweed removal as the main treatment were not considered. In the Harv. Type column, C = commercial harvest and R = researcher harvest. Sites are defined as separate rockweed beds, while plots are defined as the spatial extent at which each treatment was applied. In cases when a full rockweed bed represents a treatment (e.g., this study), sites and plots are equal. Regional regulations (minimum cutting height) are provided for studies for which we were able to calculate an effect size.

Study	Region	Harv. Type	Sites	Treatments	Total Trtmnt. Plots Per Site	Plot Size	Rockweed Characteristic	Standardized BACI Effect Size (95% CI)	Unstandardized BACI Effect Size (95 % CI)	Length of Recovery Data
Johnston et al. 2023 (this study)	Maine (USA)	C	38	Unharvested, commercial harvest (40.6 cm min. cut)	1	Mean $\pm$ SD = 3866 $\pm$ 3003 m ²	Height	-0.11 (-0.25, 0.04)	-11.9 cm (-27.3, 3.8)	1 year
							Biomass	-0.28 (-0.58, 0.003)	-2.97 kg/m ² (-6.03, 0.08)	
Kay 2015	Nova Scotia (Canada)	C	1	Unharvested, commercial harvest (12.7 cm min. cut)	2	3750 m ²	Height	-0.20	-22 cm	4 weeks
							Biomass	-0.29	-3.3 kg/m ²	
Walder 2015	New Brunswick (Canada), Maine	R	3	Unharvested, 40.6 cm	18, 30, 40	4 m ²	Height	-1.04	-89.7 cm	3 Months
							Biomass	--	--	
Ugarte et al. 2006	New Brunswick	C	1	Unharvested, commercial harvest (12.7 cm min. cut)	5	64 m ²	Height	-0.21	-13.8 cm	2 years
							Biomass	-0.52	-5.36 kg/m ²	
Kelly et al. 2001	Ireland	C	2	Unharvested, mechanical harvest, hand harvest (no min. cutting height)	3	50 m x intertidal width	Height	Hand: -1.42 Mechanical: -0.58	Hand: -63.7 cm Mechanical: -40.8 cm	11-17 months
							Biomass	--	--	
Main Results If Unable to Calculate Effect Sizes										
Lauzon-Guay et al. 2021	New Brunswick, Nova Scotia	C	NB = 21 sectors, NS = 39 sectors.	Unharvested (4 NB sectors sampled at end of study only), commercial harvest	1	Rockweed bed (sizes unknown)	No significant decrease in rockweed biomass in NB or NS harvest sectors over 25 years, but biomass in unharvested NB sectors was 7% higher than in harvested sectors at end of study. Average rockweed height decreased by 7.8 cm in NS and increased by 13.8 cm in NB. Rockweed height in unharvested NB sectors was similar to values in harvested sectors.			Repeated harvesting across 25 years. Harvest intervals unknown.
Gendron et al. 2018	Quebec (Canada)	R	1	15 cm, 30 cm	10	25 m ²	Three-year recovery period maximizes harvested biomass. 15-cm cutting height often provided higher biomass yields than the 30-cm cutting height, especially at longer recovery periods.			1,2,3,4, or 5 year recovery intervals studied over 28 years of repeated harvesting

Table 3.3 Continued

Phillippi et al. 2014	Maine	R, C	2	Unharvested, 20.3 cm (hand shears), 40.6 cm (hand shears); commercial harvest	54 (hand shears); 2 (comm. harvest)	4 m ² (hand shears); 60 m x intertidal width	No data provided on rockweed harvest. Results focus on macroinvertebrate responses to harvest.	None
Trott and Larsen 2012	Maine	C	1	Unharvested, commercial harvest	2	~683 m ²	No difference in average biomass between control and impact plots two months post-harvest.	Two months
Blinn et al. 2008	New Brunswick	C	12 (year 1), 8 (year 2)	Unharvested, commercial harvest	1	Rockweed bed (sizes unknown)	No pre-harvest data. No significant difference in rockweed density or height between unharvested and harvested sites.	None
Sutherland 2005	New Brunswick	C	43 across six sectors	Commercial harvest	1	Rockweed bed (sizes unknown)	Assessed rockweed beds in six harvest sectors. Compared to an earlier study of the same beds (Smith 2000), average rockweed bed height and biomass were lower in half of the sectors.	None
Hamilton and Nudds 2003	New Brunswick	R	1	Unharvested, 50% biomass removal	12	24.5 m ²	No data provided on impact of rockweed harvest. Rockweed volume recovered one year post-harvest, while height did not.	1 year
Fegley 2001	Maine	R	4	Unharvested, 18 cm, 36 cm	9	25 m ²	88% biomass recovery two years post-harvest for 36 cm treatment. 12.3 cm average growth in first year post-harvest for 36 cm treatment vs. 11.9 cm average growth for control.	2 years
Lazo & Chapman 1996	Nova Scotia	R	1	Unharvested, low intensity (15 min.), medium intensity (30 min.), high intensity (45 min.)	21	336 m ²	18% (SD = 5), 60% (SD= 19) and 70% (SD = 18) of the stand biomass was removed in the low-, medium-, and high-intensity treatments. Post-harvest growth was higher in harvested than unharvested plots.	2 years
Ang et al. 1993	Nova Scotia	C	1	Unharvested, commercial harvest	6	400 m ²	Localized biomass loss of 80% after harvest. Results focus on the effect of harvest on rockweed size structure and mortality.	None
Keser et al. 1981	Maine	R	10	Complete removal, 15 cm, 25 cm	3	5 m x intertidal width	Weighed harvested biomass rather than biomass of remaining algae. 15 cm and 25 cm yields were highest on the first harvest and declined with successive harvests.	3 years
Boaden & Dring 1980	Northern Ireland	C	1	Unharvested, 10-15cm	2	~1,000 m ²	Rockweed internodal length and lateral branching increased after harvest, but provided 20% less shore cover than in the uncut area.	3 years
Baardseth 1970	Norway	C	1	Commercial harvest	1	809 m ²	Variable recovery (55% to >100%) of beds, measured as harvest yield, three years after harvest	3 years

Note: Standardized effect sizes = $\ln(\frac{\bar{X}_{Harv,I}}{\bar{X}_{Harv,C}}) - \ln(\frac{\bar{X}_{Pre,I}}{\bar{X}_{Pre,C}})$. Unstandardized effect sizes = $(\bar{X}_{Harv,I} - \bar{X}_{Pre,I}) - (\bar{X}_{Harv,C} - \bar{X}_{Pre,C})$. $\bar{X}_{i,j}$ represents the mean height or biomass value during the i th time period (Pre = Pre-harvest, Harv = Harvested) at sites with j treatment type (I = impact, C = control). We were not able to calculate effect size 95% confidence intervals for studies other than our own due to data limitations.

3.5. Discussion

We conducted a Before-After Control-Impact study with commercially harvested rockweed beds at 38 sites across five regions in Maine to quantify the bed-scale impact and short-term (i.e., one year) recovery from rockweed harvest. Our findings support our prediction that harvest impacts are spatially heterogeneous at the scale of the rockweed bed, resulting in declines in mean rockweed height and biomass that were relatively small in their effect sizes relative to the effects on the most impacted rockweed individuals. These trends varied by region, with impacts most pronounced in Midcoast. Both rockweed bed characteristics increased during the year following harvest; however, there was no effect of treatment on the post-harvest growth in height, while there was a greater increase in post-harvest biomass at impact sites than control sites. Sites that experienced greater harvest intensities had lower recovery of height and biomass after one year, but overall, recovery was greater than we expected, with many impact sites showing >80% recovery in height and biomass. These findings highlight the ability of rockweed beds to initiate recovery soon after harvest. Lastly, the statistical support for an effect of treatment in several of our analyses was marginal ($0.10 > P > 0.01$) due to high between-site variation among treatments in our study.

3.5.1. Pre-harvest: Regional Comparison

Across a large portion of the Maine coast, mean rockweed height and biomass were relatively homogeneous. While there was high inter-site variation within regions, our results indicate that any regional differences in rockweed harvest histories or environmental conditions have not led to significant landscape-scale differences in rockweed bed height or biomass among our study sites that span the coast of Maine. Furthermore, the range of mean site biomass values

in our study (3.53 – 21.76 kg/m²) did not differ markedly from previous assessments of rockweed biomass in Maine (Rockweed Plan Development Team et al. 2014; see their Table 2).

Variance in rockweed height and biomass increased monotonically across hierarchical spatial scales, but in opposite directions for the two characteristics. For biomass, variance was larger within transects than within sites, but this phenomenon may have been a methodological outcome rather than a biological one. While transects were typically in areas where rockweed fully covered the substrate, we only weighed algae with holdfasts inside the quadrat. Quadrats that contained no holdfasts had biomass values of zero, which contributed to high variance in some transects. In contrast, measuring height was predicated on rockweed presence and mean values were not expressed in areal units. Variance in height within quadrats was less than half of the variance within transects and sites. This may be due in part to patterns of disturbance—natural or anthropogenic—that create variation in height at similar spatial scales to some of our sampling scales (e.g., transect, site), but not others (quadrat).

3.5.2. Pre-harvest to Harvested: Impact of Harvest

The bed-scale impact of rockweed harvest on biomass was roughly twofold greater than on height. These findings are similar to other studies of commercial rockweed harvest in the western North Atlantic (Kay, 2015; Ugarte et al., 2006; Table 3.3). Taller rockweed individuals tend to have a greater proportion of their biomass in the upper canopy compared to smaller individuals (Ugarte et al., 2006), and as a result, the proportion of a mature individual's biomass and height lost during harvest may not be equal. The effects of differential harvest impacts on height and biomass for other species within the associated food web are likely taxa-specific. For instance, canopy height may be a more important bed characteristic for the youngest class of

Common Eider (*Somateria mollissima*) ducklings that feed in rockweed habitats by dabbling in floating canopies at higher tides (Hamilton 2001). Conversely, some macroinvertebrates such as isopods forage on rockweed epiphytes (Pavia et al. 1999) and may be most impacted by reductions in the surface area (i.e., biomass) of rockweed beds. Height and biomass may also provide different roles in modifying abiotic conditions in rockweed habitats. For instance, average air temperatures along the Maine coast are increasing (Maine Climate Office, 2022) and rockweed's ability to provide thermal refugia during low tide may be differentially impacted by reductions in height versus biomass.

The effect sizes of harvest on both height and biomass were lower in our study than in other studies for which we could calculate effect sizes (Table 3.3). Small, researcher-harvested plots (Walder, 2015) and locations with no minimum cutting heights (Kelly et al. 2001) unsurprisingly exhibited the largest effect sizes (only height assessed in both studies). Furthermore, minimum cutting height in Atlantic Canada (12.7 cm) is considerably shorter than in Maine (40.7 cm) and may explain some of the larger effect sizes in other studies (Kay, 2015; Ugarte et al., 2006). However, the effect size of harvest on biomass in our study (-0.28) was very close to the value (-0.29) in the only other study to conduct commercial harvest at the scale of the rockweed bed for which we could calculate effect sizes (Kay, 2015). Even though these studies occurred in locations with different minimum cutting heights, harvesters cannot precisely control the cutting height of algae at higher tides (Ugarte et al., 2006), and the exploitation rates may have been similar among comparable harvest efforts. Future rockweed harvest studies should provide data that allow for the calculation of effect sizes, which will enable managers to assess whether -0.3 is a typical effect size of harvest on biomass across regions and harvest methods.

The impact of rockweed harvest on both bed characteristics was variable across our four study regions. The southernmost region (Midcoast) was the only region to exhibit statistically significant declines in height or biomass among impact sites. Standardized effect sizes were roughly threefold stronger for both characteristics in this region (height: -0.28 [95% CI = -0.54 to -0.02]; biomass: -0.78 [95% CI = -1.16 to -0.41]) than for our study-wide estimates (Table 3.3). This region contained only mechanical harvest—while the other regions contained a mixture of mechanical and hand harvest—but it also was the only region harvested by two of the harvesting companies. These possible explanatory factors were not fully crossed in our study design and we therefore are unable to speculate as to whether the observed regional differences in harvest impacts were due in larger part to harvest method or company. The main objective of our study was to assess the average impact of harvest protocols presently operating in Maine. However, this result highlights the importance of monitoring a harvested resource across the full spatial scale of exploitation.

Lastly, we acknowledge the limitations of our sampling design given the fact that there were several impact sites that increased in mean height ($N = 5$) and biomass ($N = 8$) between the pre-harvest and harvested time periods. This is likely due to a lack of spatial overlap between transects and harvest at these sites. Despite this limitation, our study had high replication among each treatment (19 impact and 19 control sites), and we believe that treatment means (Figures 3.3C, 3.3F) are informative of an average impact of harvest at the scale of the rockweed bed. It would be valuable to sample a small number of commercially harvested sites extensively (e.g., nine 10-m transects equally stratified by tidal height) and compare the estimates of impact (i.e., effect sizes) and associated confidence intervals to those obtained using our sampling

methodology at the same sites. These data can test the ability of our methods to serve as a valid index of the average bed-scale impacts of rockweed harvest across a large number of sites.

3.5.3. Harvested to One Year Post-harvest: Regrowth After Harvest

One year after harvest, there was an effect of treatment on the post-harvest growth of rockweed biomass, but not height, indicating that the two rockweed characteristics responded differently immediately after harvest. These findings are generally congruent with results from other harvest studies that have evaluated rockweed height and biomass growth over comparable post-harvest intervals (Fegley 2001, Ugarte et al. 2006). Mature, unharvested rockweed individuals tend to have a bimodal distribution of frond lengths, with a few large fronds that form the canopy and high densities of small, suppressed fronds that grow slowly due to low sub-canopy irradiance levels (Cousens 1985, Ang et al. 1996). Rockweed canopies float on the surface of the water during higher tides and are the portion of the algae typically harvested (Ugarte et al. 2006). A common hypothesis is that rockweed harvest results in reduced canopies that allow growth of suppressed shoots due to increased light exposure (Baardseth, 1970; Cousens, 1985; Ugarte et al., 2006; Vadas and Wright, 1986). Furthermore, harvest can lead to increased lateral branching during regrowth that increases total biomass relative to canopy height (Boaden and Dring 1980). While our study lacks data on the mechanistic underpinnings of these processes, our results support the idea that a significant proportion of post-harvest growth occurs below frond apices in the canopy. This likely explains why there was greater growth of biomass (weighing whole alga) than height (a maximal value) after harvest at impact sites. Our results might suggest that individual rockweed morphology becomes more dense during short-term recovery (Fegley 2006), but we cannot definitively conclude this given that we compared site-

level averages in height and biomass rather than explicitly measuring the morphological change of marked individuals.

Regional variation in post-harvest biomass growth was largely related to the magnitude of initial declines after harvest. This is apparent in the V-shaped trends at impact sites in the Midcoast and Penobscot regions (Figure 3.4B). However, biomass increased considerably in Cobscook during the recovery interval, despite weak initial declines from harvest. Site-level data reveal that declines and gains in biomass between the pre-harvest and harvested time periods resulted in little average change in the region, but post-harvest gains at the two sites that lost biomass were particularly high (Figure C.2).

3.5.4. Pre-harvest to One Year Post-harvest: Recovery from Harvest

One year post-harvest, mean rockweed biomass fully recovered at impact sites relative to control sites, while mean height remained lower at impact sites. The ability of rockweed to recover a large proportion (> 80%) of its biomass soon after harvest, and even exceed initial values in some instances, has been observed widely across regions and harvest protocols (Baardseth 1970, Fegley 2001, Hamilton and Nudds 2003, Ugarte et al. 2006, Trott and Larsen 2012, Gendron et al. 2018). Despite greater biomass growth at sites with higher harvest intensities, it is important to note that the likelihood of complete height and biomass recovery after one year was lower at these more disturbed sites. As a result, if the goal is to maintain tall algal canopies, resource managers may need to allow longer recovery intervals at sites with higher harvest intensities, even in the presence of higher rates of biomass growth.

While rockweed height may not recover at the same rate as biomass, it is important to consider the ecological implications of a) the difference in height observed at the end of the

study (control: 115.4 ± 4.3 cm, impact: $107.7 \text{ cm} \pm 4.3$) and b) potential changes in the height-biomass relationship in harvested rockweed beds. Ultimately, resource managers must consider thresholds of ‘recovery’ that vary by resource stakeholder. For instance, rockweed harvesters or blue carbon initiatives may be most interested in the total biomass of a bed, while wildlife managers may focus on retaining bed characteristics (e.g., clump density) that benefit another taxon of interest.

Another critical factor to consider in rockweed management is the recovery interval between repeated harvests. Our study was extensive in spatial scale, but it was limited in temporal scale. It is unclear how observed trends would change in years two and three of recovery, and whether biomass at impact sites would continue to increase at a faster rate than height. We recognize that resource managers must think about harvest intervals and recovery of the resource at multi-year and decadal scales (e.g., Gendron et al., 2018; Lauzon-Guay et al., 2021). While we cannot say with certainty that the magnitude of rockweed harvest observed in this study is sustainable across repeated harvests and shifting environmental conditions, our data suggest that recovery, especially in biomass, can occur within a single year at the bed-scale using current commercial methods.

3.5.5. Conclusions

As ecosystem-based management (EBM) approaches become more widely adopted in marine ecosystems, it is important to consider management at several spatial scales. For instance, commercial rockweed harvesting in regions of Maine and Atlantic Canada is regulated at the scale of the individual alga (40.6 cm and 12.7 cm minimum cutting heights, respectively) and at the scale of the sector (17% annual biomass removal). Examining rockweed harvest impacts and

growth rates in the patches of highest harvest intensity is critical for understanding how the target resource responds to disturbance. However, when harvest is spatially heterogeneous at the bed scale—as is the case with commercial rockweed harvest—quantifying harvest impacts at larger spatial scales can benefit resource management in EBM frameworks.

Our findings indicate that the impact of commercial rockweed harvest on two common bed characteristics (height and biomass) is smaller at the scale of the rockweed bed than at smaller scales that do not capture the spatial heterogeneity of harvest impacts. This study represents an initial step toward making EBM recommendations in these ecosystems, and the applicability of this perspective depends on the spatial scale at which resource managers target their actions. Highly mobile organisms such as birds and fish may utilize rockweed habitats at the bed scale or larger and respond to bed averages more closely than smaller scale impact estimates. Mobile macroinvertebrates, on the other hand, may relocate within beds to areas of either the highest or lowest harvest impacts, with potentially little changes in their abundance at the bed scale within some ranges of spatial heterogeneity. Further research is needed to understand the implications of our findings for other members of rockweed ecosystems, but our findings underscore the importance of taking bed-scale impacts into consideration. Future harvest impact studies should include recovery data of sufficient duration (e.g., 1–3 years) and prior harvest history whenever possible to understand how the various components of these ecosystems respond after repeated commercial harvest.

Chapter 4

Lack of Strong Bottom-up Forcing of a Rocky Intertidal Resource Harvest on a High Trophic-level Consumer Group

4.1 Chapter Abstract

The development of ecosystem-based management (EBM) frameworks for resource harvest has increased in importance over the past several decades as ecosystems face numerous anthropogenic stressors. In these frameworks, resource managers must consider the suite of interactions that comprise food webs and how resource harvest drives responses in non-target organisms. In rocky intertidal zones along North Atlantic coastlines, rockweed (*Ascophyllum nodosum*)—a canopy-forming brown seaweed—has been commercially harvested for centuries, yet the vast majority of research on the effects of harvest have focused on the responses of the target resource and the macroinvertebrate assemblage. In this study, we used a Before-After Control-Impact experiment to assess the bottom-up effects of commercial rockweed harvest on a high trophic-level consumer group (birds) in Maine (USA). Among the full intertidal bird assemblage, we found no treatment effect immediately following harvest, while there was a small (1.2 birds) positive effect size of harvest during the one-year post-harvest recovery interval. Several of the trends observed for the full assemblage appear to be driven more strongly by the low tide than the high tide assemblage. Independent of treatment, patch visitation by birds was low in our study (60% of surveys recorded no birds) and highlights questions about the use of rocky intertidal habitats relative to other habitat types in their home ranges. Overall, there was no evidence for strong bottom-up forcing of rockweed harvest on birds' patch visitation, but if similar intensities of harvest occurred in areas with higher bird densities there may be larger changes in patch visitation. To our knowledge, there are no comparable assemblage-level bird

studies in this system that can provide context to our results. Further research of rockweed-bird interactions is needed to more confidently incorporate this high trophic-level consumer group into an EBM framework for a rocky intertidal resource harvest.

4.2 Introduction

The management of many marine fisheries has historically employed a single-species approach that focused on the regeneration of the target organism's biomass (Hall and Mainprize 2004). As the number and magnitude of stressors to marine ecosystems has grown, the importance of management approaches that consider broader ecosystem processes—such as maintaining food web connectivity—has grown concomitantly (e.g., ecosystem-based management (EBM) paradigm; Pikitch et al. 2004). The resiliency of marine ecosystems to current and future global change will be buoyed by fisheries that view harvested organisms as members of food-web networks that interact with other organisms (Holsman et al. 2020).

In an EBM framework, managers must consider the impacts of resource harvest on non-target organisms. Exploitation of one food-web member can initiate top-down or bottom-up forces that result in multi-species responses (Reisewitz et al. 2006, Lorentsen et al. 2010). In particular, the harvest of habitat-forming autotrophs is well suited to EBM approaches given their potential to influence higher trophic levels. For example, harvest impacts on autotrophs can limit flows of energy to consumers, and the modification of autotroph (i.e., habitat) structural characteristics can disrupt food web connections if some members emigrate to other habitats (Dugan et al. 2003, Orr 2013, Leaver et al. 2021).

Many commercially harvested wild seaweeds (macroalgae) form structurally complex canopies that increase the abundance and diversity of associated macrofauna (Wikström and

Kautsky 2007, Schmidt et al. 2011). Despite this, resource managers frequently focus management objectives on the regeneration of the target organism (Lotze et al. 2019). This is likely due, in part, to a paucity of research on the effects of seaweed harvest on high-level consumers, such as birds (but see Hamilton & Nudds 2003, Lorentsen et al. 2010). Primary consumers can be negatively impacted by seaweed harvest (Fegley 2001, Walder 2015), but the magnitude of a basal-level perturbation often attenuates across trophic levels (Borer et al. 2006). As a result, it is unclear empirically and theoretically whether top consumers are likely to be impacted by seaweed harvest.

A canopy-forming fucoid seaweed—rockweed (*Ascophyllum nodosum*)—has been commercially harvested in North Atlantic rocky intertidal zones for centuries (Pereira et al. 2020). Rockweed is considered a foundation species (Dudgeon and Petraitis 2005) that contributes structural complexity to habitats (Schmidt et al. 2011), food resources (i.e., epiphytes) for primary consumers (Pavia et al. 1999), and foraging habitat for higher-level consumers (Rangeley and Kramer 1998). Due to its commercial and ecological value, the effects of rockweed harvest have been studied extensively, but much of the research has focused on the responses of the harvested organism (i.e., rockweed; Keser et al. 1981, Ugarte et al. 2006, Johnston et al. 2023) and primary consumers (e.g., macroinvertebrates; Trott & Larsen 2012, Phillippi et al. 2014, Walder 2015) to harvest.

In this study, we quantify the patch-level response of an intertidal bird assemblage to commercial rockweed harvest to understand how basal-level perturbations affect top trophic levels in an intertidal food web (Figure 4.1). Birds are a suitable high-level consumer group with which to test the impacts of rockweed harvest for several reasons. First, there are positive correlations between characteristics of rockweed habitats and the presence and behavior of some

bird groups, but it is unclear whether rockweed harvest changes these relationships (Hamilton 2001, Johnston et al. 2021). Second, unlike fish, birds can be monitored from upland observation points that minimize changes in patch visitation during surveys. Finally, birds garner high public interest and are an effective animal group through which to communicate research findings to lay audiences (Ainsworth and Burns 2020).

To test the effect of commercial rockweed harvest on the intertidal bird community, we conducted a repeated-measures Before-After Control-Impact (BACI) experiment (Green 1993) at 40 sites across the Maine (USA) coast (Figure 4.2). We defined each study site as a patch and assessed if rockweed harvest affected birds' patch visitation (i.e., count) relative to unharvested control patches. Measuring patch visitation in a BACI framework allowed us to assess whether resource harvest alters the use of habitat patches for birds (e.g., Heyman 2010). We quantified patch visitation by performing bird point-count surveys across three time periods (pre-harvest, recently harvested, and one year post-harvest).

We hypothesized that a negative effect of rockweed harvest on patch visitation would be relatively small for two reasons. First, there is little evidence that commercial rockweed harvest decreases macroinvertebrate abundances or diversity in our region (Maine; Trott & Larsen 2012, Phillippi et al. 2014). If birds perceive and use prey abundance to inform patch use in rockweed habitats, we would not expect large changes in patch visitation when prey abundances have not significantly declined. Second, if rockweed harvest negatively impacts prey resources, birds may not perceive this difference until they have entered a patch (i.e., their perceptual range; Lima & Zollner 1996). We would still expect to observe a negative effect of rockweed harvest if birds spend less time in patches due to the decreased likelihood that our surveys overlapped with birds' patch visitation at impacted sites. However, for this to occur, prey densities would need to fall

below birds' threshold for decreased time spent in a patch. Rockweed harvest could affect patch visitation for reasons other than prey densities as well. Birds may be attracted to recently harvest sites that represent novel heterogeneity along rockweed-dominated coastlines (Robertson et al. 2013). Lastly, we hypothesized that any impacts of rockweed harvest on bird count will vary across bird groups that use the patches at different tidal stages (i.e., high and low tide assemblages) due to differences in bird foraging mode.

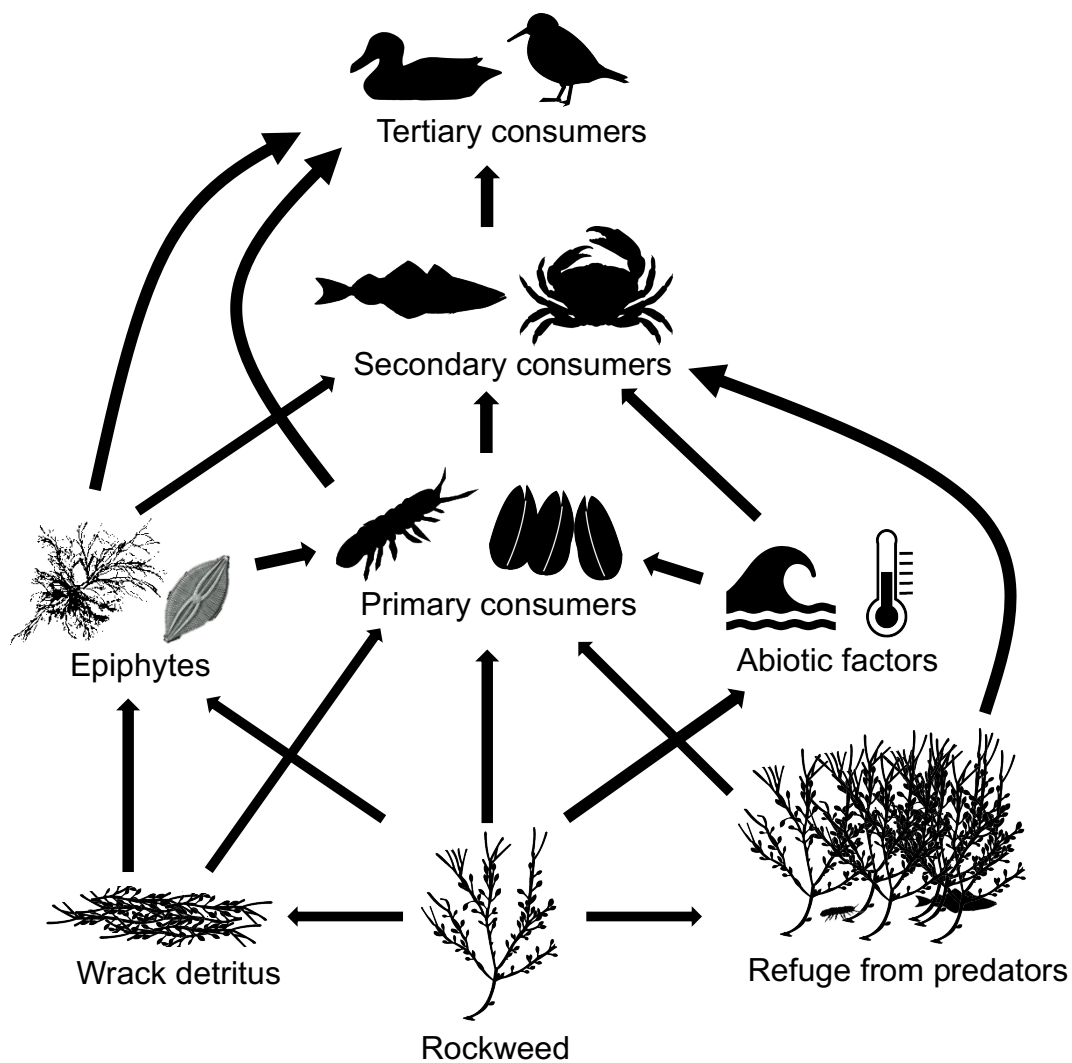


Figure 4.1. Interaction web displaying common trophic and non-trophic interactions in rockweed-dominated rocky intertidal zones in Maine. Arrows only represent linkages and variation in size does not indicate interaction strengths.

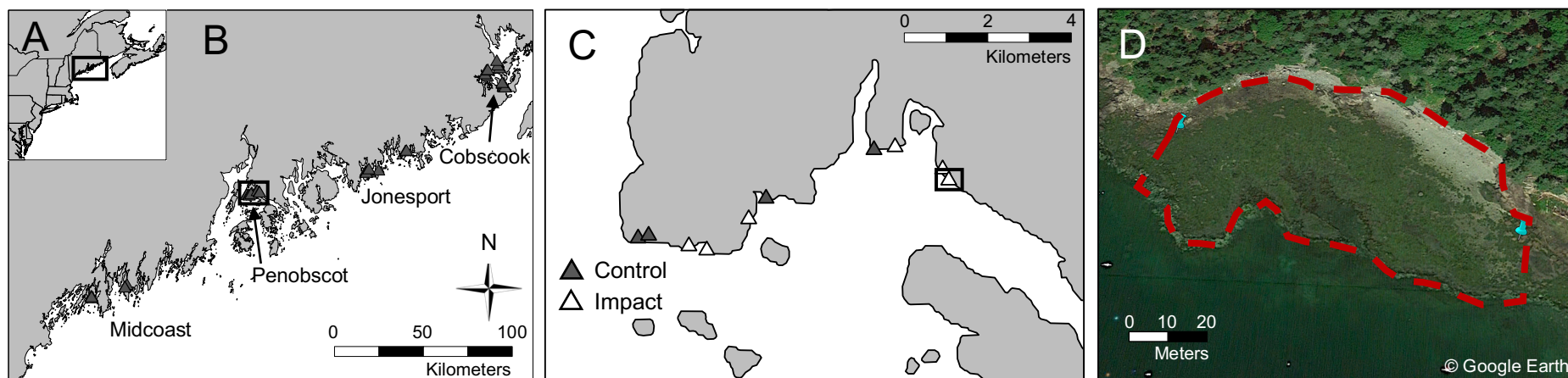


Figure 4.2. Hierarchical spatial scales displaying study sites. Our research occurred (A) along the Maine coastline in the northwestern Atlantic Ocean where we established (B) 40 sites in four regions. Within each region, (C) site placement was determined by the location of harvest-quality rockweed beds and landowner access permissions. Each site (D) represented a rockweed habitat patch that was 100 meters parallel to the shoreline and vertically bounded by the high and low tide lines. Regional treatment totals were as follows: Midcoast 4 control, 6 impact; Penobscot 4 control, 6 impact; Jonesport 4 control, 4 impact; Cobscook 7 control, 5 impact.

4.3. Methods

4.3.1. Study Design

We designed our study to measure an intertidal bird assemblage's use of rockweed-habitat patches within constituent home ranges (i.e., third order habitat selection; Johnson 1980) and to test whether commercial rockweed harvest affected patch use. We used a repeated-measures BACI framework that consisted of bird and rockweed surveys across three time periods—pre-harvest (August–October 2018), recently harvested (August–December 2019), and one year post-harvest (August–November 2020)—to quantify changes in birds' patch use immediately following rockweed harvest and after one year of recovery. One year of regrowth can result in considerable recovery of rockweed biomass in this system (Hamilton and Nudds 2003, Ugarte et al. 2006, Johnston et al. 2023).

Certain aspects of our study design were either informed or constrained by several considerations: maximizing the representativeness of harvest to commercial practices in our region, capturing bird use of rockweed habitats, and survey feasibility. First, we worked with several rockweed harvesting companies and private landowners to establish 40 study sites ($N = 19$ control and 21 impact) across five regions in Maine (Fig 3.1B). These conversations ensured that all of our sites were of harvestable size and quality, as determined by those in the industry who make these decisions. After site selection, we assigned treatment level (impact or control) based on landowner permissions and without input from harvest companies. A detailed methodology of the site selection process and rockweed harvest protocols can be found in Johnston et al. (2023). All impact sites were harvested once between June and November 2019 by commercial harvesters using their regular procedures under current Maine regulations (40.6

cm minimum cutting height), while control sites were unharvested throughout our study, although some sites likely have been harvested historically.

Second, the scale of our habitat patches were set to align with the scales of both industrial harvest and bird use. We defined a habitat patch (i.e., study site) as an area 100 m in length parallel to the shoreline and vertically bounded by the high and low tide lines (mean area $\pm$ SD = $3866 \pm 3003 \text{ m}^2$; Figure 4.2D). This patch size is a) similar to the scale over which a single commercial harvest event occurs (A. Feibel, personal communication) and b) large enough to encompass foraging bouts for our target bird species while keeping bird detection high, particularly for surveys during lower tides at sites with high rugosity.

We did not include the shallow subtidal in our patches given that rockweed is an intertidal species and the presence of predominantly subtidal birds is not determined by intertidal macroalgae (Johnston et al. 2021). During higher tides, it was sometimes difficult to know whether a given bird was inside (intertidal) or outside (subtidal) of the patch. We marked uncertain observations on the datasheet and recreated birds' positions over low tide imagery in Google Earth using the coordinates of the observer, the compass bearings to the birds, and the distances to the birds. Out of 14 birds that we assessed with this approach, we removed eight that were outside patch boundaries.

Sites were clustered within each region (nearest neighbor distance: 102 – 5041 m, mean $\pm$ SD: $664 \pm 885 \text{ m}$; Figure 4.2C) to allow multiple harvests and bird surveys within a single day. Given that the home range sizes of coastal birds are typically larger than our inter-site distances (Herring and Collazo 2005, Anderson et al. 2019), it is possible that birds were counted in more than one patch during a single survey day. As such, it is important to interpret our counts as a metric of bird patch use and not one of bird population size. Our study was designed to capture

the total exposure of patches to the intertidal bird assemblage, and does not rely on assumptions of site independence that are common in studies of population biology (MacKenzie et al. 2002). Lastly, we used an annual survey window from August to December for all research activities in order to ensure that a) rockweed harvest occurred outside the window of rockweed reproduction and b) bird surveys overlapped with the autumn migratory period for shorebird species (Hicklin 1987, Vadas et al. 2004), which use the rocky intertidal during autumn migration (Johnston et al. 2021).

4.3.2. Bird and Rockweed Surveys

We conducted bird point-count surveys at each site once before harvest (2018), twice immediately after harvest (2019), and twice one year after harvest (2020). This unbalanced survey design was a result of increased sampling in the second and third time periods due to low bird counts during initial surveys. Treatment assignments and survey timing within each of the three time steps were conducted such that the distribution of patch size and the tidal height and ordinal date of surveys were similar across all levels of treatment $\times$ time period (Figure D.1).

Following the rocky intertidal bird survey methodology in Johnston et al. (2021), we conducted 20-minute point counts from a fixed upland location during which we counted individuals of all species present within the site boundary and recorded the distance (using a laser range finder) and compass bearing between the observer and each bird or flock of birds. Ten-power magnification binoculars were used at most sites and a forty-power magnification spotting scope was used at the largest sites. Observers were obscured behind upland vegetation or at a distance that would not disturb birds in the study plot. If birds flushed before the survey began, we counted species to the best of our ability and estimated distances and compass bearings from the point of observation. Birds that did not interact with intertidal habitat (i.e.,

flyovers) were not counted. We did not limit our surveys to particular diurnal hours given that time of day is not correlated with bird use of the rocky intertidal zone in our region (Herring & Collazo 2005, Anderson et al. 2019). In addition, we avoided conducting bird surveys within 24 hours after harvest to decrease the impact of harvester presence on bird site use.

Rockweed bed height and biomass were measured once per time period at each site. We calculated mean site values for each rockweed characteristic by averaging values across two 10-m transects that each contained five 0.5×0.5 m quadrats. A detailed description of our rockweed assessment methodology can be found in Johnston et al. (2023).

4.3.3. Statistical Analysis

We tested the impact of commercial rockweed harvest on three groups: all birds, low tide assemblage, and high tide assemblage. Bird species ($N = 25$) were grouped into tidal height assemblages (Figure D.2) based on the substrate (exposed benthos or water column) that is most representative of their predominant foraging modes. For the species that commonly forage in both substrates (*Larus* spp.; $N = 3$), we summed counts of each within two-hour windows centered on high and low tides, and we placed each species in the tidal height group with a greater total count. We did not place counts of these three species in both assemblages based on tidal stage due to a lack of information about the substrate use associated with each observation.

All statistical analyses were performed in program R version 4.2.0 (R Core Team, 2022). To test the effect of commercial rockweed harvest on patch visitation, we used zero-inflated generalized linear mixed models (ZI GLMMs) in the glmmTMB package (Brooks et al. 2017) for each of our three bird assemblages (all birds, low tide assemblage, high tide assemblage). All three models included two fixed effects (treatment and time period), their interaction, and a

random intercept of site nested within region. ZI GLMMs allowed us to account for non-normality distributed count data and a high incidence of zero counts (60% across all surveys). This approach allowed us to evaluate the effect of rockweed harvest on both bird presence (binomial) and count if present (conditional poisson). The binomial model estimates the probability of an extra zero such that a positive variable coefficient indicates a higher probability of absence, while a positive variable coefficient in the conditional model indicates a positive effect on abundance (Brooks et al. 2017). All model diagnostics were examined with the DHARMA package (Hartig 2017) to ensure adequate model fit before interpreting parameters. Main and interactive effects for each model component (binomial and conditional) were examined with an analysis of variance (ANOVA) using Type II Wald χ^2 tests. When interactive effects were statistically significant ($P \leq 0.05$) we constructed post-hoc contrasts to assess pairwise comparisons using the emmeans package (Lenth et al. 2020). To quantify the magnitude of harvest impact and post-harvest recovery, we calculated unstandardized effect sizes,

$$(X_{i+1, I} - X_{i, I}) - (X_{i+1, C} - X_{i, C}), \quad (\text{Eq. 4.1})$$

where $X_{i,j}$ equals point estimate X during time period i at sites with treatment type j (I = impact, C = control). Point estimates for each treatment-time period combination were derived with the ‘predict’ function based on fully specified (i.e., binomial and conditional poisson) models. For each effect size, we computed reduced bias-corrected bootstrap 95% confidence intervals (iterations = 1000; Tibbe & Montoya 2022). We were unable to compute effect size confidence intervals for the low tide assemblage due to repeated failures of model convergence that resulted from excess zero counts after random sampling with replacement. However, the effect size point estimates for this assemblage were all < 1 bird, which is below biological relevance for our system even if the estimates had little error.

In addition to testing for effects of rockweed harvest, we directly evaluated the relationship between two rockweed bed characteristics—mean biomass and height—and bird presence and conditional count for all three bird groups. This allowed us to incorporate environmental variation not captured by our treatment assignments, such as different harvest intensities at impacted sites (Johnston et al. 2023) and natural sources of disturbance (e.g., ice scour, extreme wave action) at any site. We modeled these relationships using ZI GLMMs with a fixed effect of the focal rockweed characteristic and a random intercept of site nested within region. For significant relationships, we generated predicted probabilities or counts for only the binomial or conditional poisson model, respectively.

For all analyses, we removed four large bird flocks ($N = 54, 22, 22$, and 20 birds) to avoid a) issues of non-independence among birds in flocks (Sundar 2006) and b) outlier counts in analyses that had high leverage. However, these flocks all occurred at post-harvest impact sites and we assess potential implications of removing these observations on our findings in the discussion section.

4.4. Results

Patch visitation was low across 200 total surveys at 40 patches, with no birds recorded during 119 surveys (60%) and 2.8 ± 5.3 (mean $\pm$ SD) birds present when counts were greater than zero (Figure 4.3A). Some bird surveys occurred at considerably later ordinal dates during the immediately post-harvest time period (August 6 – December 6) than during the pre-harvest time period (August 13 – October 26) due to constraints imposed by rockweed harvest schedules. Six out of 21 impact sites were harvested after October 30. While this precluded observations of some migratory species during the latest surveys (i.e., November and December), most of the

species recorded in our study were year-round residents along the Maine coast. We observed 25 bird species throughout the study, but many of these species were recorded infrequently and four year-round residents—Common Eider (*Somateria mollissima*), Double-crested Cormorant (*Phalacrocorax auratus*), American Crow (*Corvus brachyrhynchos*), and Herring Gull (*Larus argentatus*)—accounted for 60% of all observations. Thirteen species were associated with the high tide assemblage and twelve with the low tide assemblage (Figure D.2). Counts were significantly lower in the Penobscot region (mean $\pm$ SD = 0.4 ± 1.1 birds, N = 50 surveys) than in the other three regions ($F_{3, 196} = 5.33$, $P = 0.002$; Figure 4.3B).

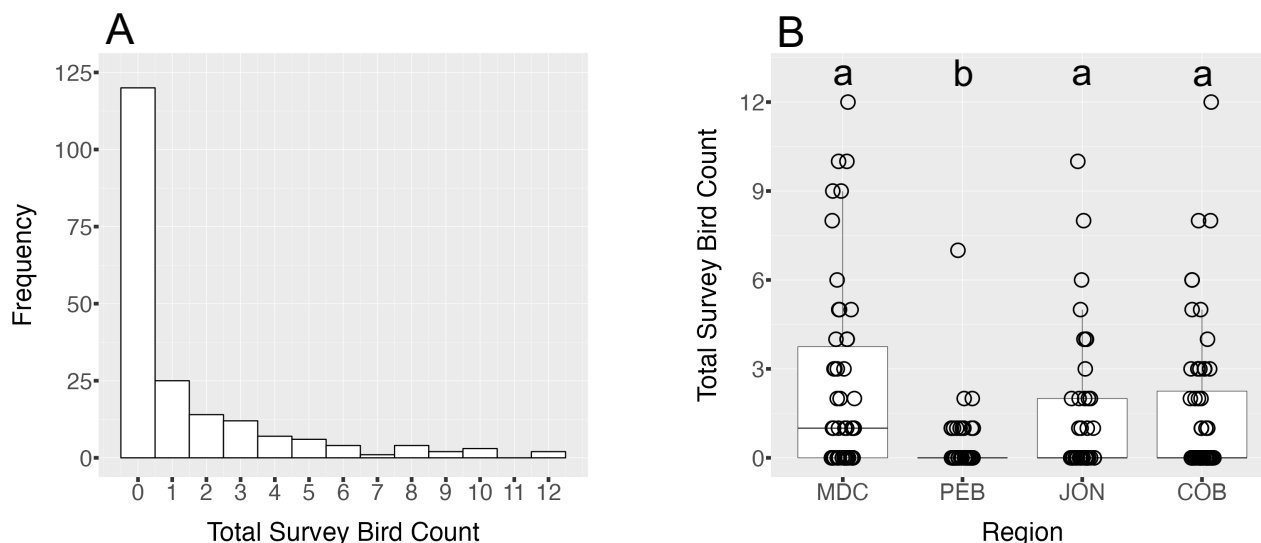


Figure 4.3. (A) Histogram of overall bird counts (N = 200 surveys), and (B) bird counts across four regions in Maine. Regions that do not share a letter had statistically significant ($\alpha = 0.05$) differences in bird counts. Abbreviations are as follows: MDC = Midcoast, PEB = Penobscot, JON = Jonesport, and COB = Cobscook.

Among all birds, the treatment $\times$ time period interaction was statistically significant for both the binomial ($\chi^2 = 7.96$, $df = 2$, $P = 0.02$) and conditional ($\chi^2 = 6.85$, $df = 2$, $P = 0.03$) model components (Table 4.1). Immediately following harvest, there was no treatment effect on

bird presence ($t_{1,184} = -0.003$, $P > 0.99$), but there was marginal statistical support for a negative effect of harvest on conditional counts ($t_{1,184} = -1.93$, $P = 0.055$). During the post-harvest recovery period, there was a positive effect of harvest on bird presence ($t_{1,184} = -2.821$, $P = 0.005$; Fig. 4A), but there was no treatment effect on conditional counts ($t_{1,184} = -0.30$, $P = 0.76$).

The overall effect sizes—which incorporate both binomial and conditional poisson model components—for the two time period intervals either had confidence intervals that crossed zero or were quite small. The initial effect size of harvest (pre-harvest to recently harvested) was -0.66 birds, but the 95% CI ($-2.59 - 0.96$) indicated that this effect may not differ from zero. In contrast, the effect size of post-harvest recovery (recently harvest to one-year post-harvest) was 1.2 birds (95% CI = $0.14 - 2.37$), which indicates a small, positive residual effect of harvest during the first year of recovery (Figure 4.4A).

Independent of site treatment, there was no relationship between the presence or conditional count of birds and average rockweed biomass ($\chi^2 = 2.04$, $df = 1$, $P = 0.15$; $\chi^2 = 1.16$, $df = 1$, $P = 0.28$; respectively). Average rockweed height also had no effect on conditional bird counts ($\chi^2 = 1.44$, $df = 1$, $P = 0.23$). There was marginal support for a negative effect of height on bird presence in a patch ($\chi^2 = 3.84$, $df = 1$, $P = 0.0501$), with the probability of presence decreasing from 76% (95% CI: 48–92%) to 32% (95% CI: 16–54%) across the full range of mean rockweed bed heights observed in our study (67–150 cm; Fig. 3.7A).

For the low tide bird assemblage, birds visited study patches during 54 out of 200 (27%) surveys, and in total we recorded 147 birds. The treatment $\times$ time period interaction was statistically significant for the conditional component ($\chi^2 = 6.82$, $df = 2$, $P = 0.03$) and marginally non-significant for the binomial component ($\chi^2 = 5.89$, $df = 2$, $P = 0.053$; Table 4.1). Immediately following harvest, there was no effect of harvest on the presence of this bird group

($t_{1, 184} = -0.25$, $P = 0.80$), while there was a negative effect of harvest on conditional counts ($t_{1, 184} = -2.34$, $P = 0.02$). During the post-harvest recovery period, there was a positive effect of harvest on bird presence ($t_{1, 184} = -2.29$, $P = 0.02$), but not conditional counts ($t_{1, 184} = -0.02$, $P = 0.99$).

The overall effect sizes of harvest were < 1 bird for the low tide assemblage. Immediately following harvest the effect size was -0.86 birds (95% CI not calculated, see methods), while it was 0.66 birds during the recovery period (Figure 4.4B). There was no statistically significant relationship between patch visitation and mean rockweed bed height for the low tide assemblage (binomial: $\chi^2 = 0.08$, $df = 1$, $P = 0.78$; conditional: $\chi^2 = 0.58$, $df = 1$, $P = 0.45$). In contrast, the conditional count of this group exhibited a positive, accelerating relationship with mean rockweed biomass ($\chi^2 = 12.19$, $df = 1$, $P < 0.001$), while presence did not ($\chi^2 = 2.06$, $df = 1$, $P = 0.15$). Predicted conditional counts increased from 0.6 to 4.5 birds across the full range of mean biomass values ($4\text{--}20 \text{ kg m}^{-2}$) at sites for which we observed this assemblage (Figure 4.5B).

Among the high tide bird assemblage, birds visited study patches during 45 out of 200 (23%) surveys, and in total we recorded 135 birds. The treatment $\times$ time period interaction was not statistically significant for either the binomial ($\chi^2 = 2.45$, $df = 2$, $P = 0.29$) or conditional ($\chi^2 = 4.66$, $df = 2$, $P = 0.10$) model components, indicating that rockweed harvest did not effect this group's patch visitation in our study (Table 4.1, Figure 4.4C). Furthermore, there was no statistically significant relationship between visitation and mean rockweed biomass (binomial: $\chi^2 = 2.09$, $df = 1$, $P = 0.15$; conditional: $\chi^2 = 0.32$, $df = 1$, $P = 0.57$) or height (binomial: $\chi^2 = 0.68$, $df = 1$, $P = 0.41$; conditional: $\chi^2 = 0.58$, $df = 1$, $P = 0.45$).

Table 4.1. Results of analysis of variance Type II Wald χ^2 tests for main and interactive effects for zero-inflated generalized linear mixed models. Each model is composed of a binomial component that models the process of zero versus positive counts and a conditional poisson component that separately models the process of counts among non-zero counts only. Statistically significant results ($\alpha = 0.05$) are in bold.

Bird group	Source of variation	Binomial			Conditional		
		<i>df</i>	Wald χ^2	<i>P</i>	<i>df</i>	Wald χ^2	<i>P</i>
All birds	Treatment	1	1.50	0.009	1	2.10	0.15
	Time Period	2	1.93	0.02	2	0.91	0.63
	Treatment $\times$ Time Period	2	7.96	0.02	2	6.85	0.03
Low tide assemblage	Treatment	1	0.82	0.36	1	0.23	0.63
	Time Period	2	2.52	0.28	2	0.58	0.75
	Treatment $\times$ Time Period	2	5.89	0.053	2	6.82	0.03
High tide assemblage	Treatment	1	0.37	0.55	1	0.06	0.81
	Time Period	2	1.37	0.50	2	4.14	0.13
	Treatment $\times$ Time Period	2	2.45	0.29	2	4.66	0.10

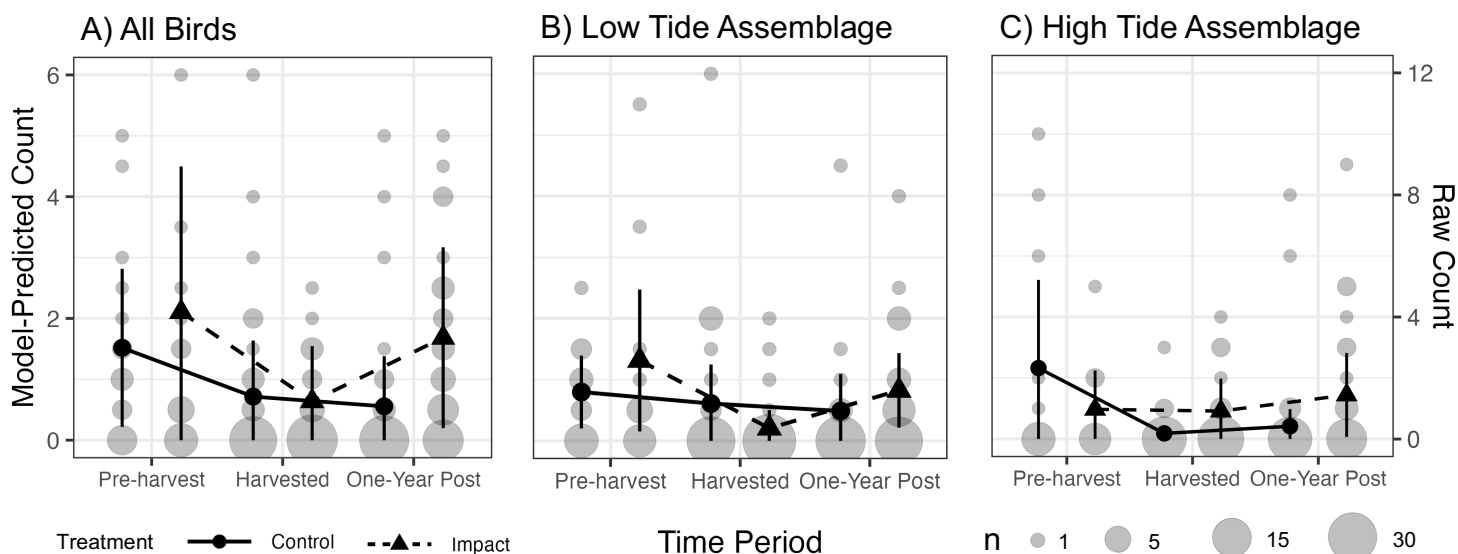


Figure 4.4. Responses of intertidal bird assemblages to commercial rockweed harvest. Black points are model-predicted estimates based on zero-inflated generalized linear mixed models. Predictions incorporate both binomial and conditional poisson model components. Error bars represent 95% confidence intervals. Gray points are raw counts that are scaled based on the number of observations for a given discrete value.

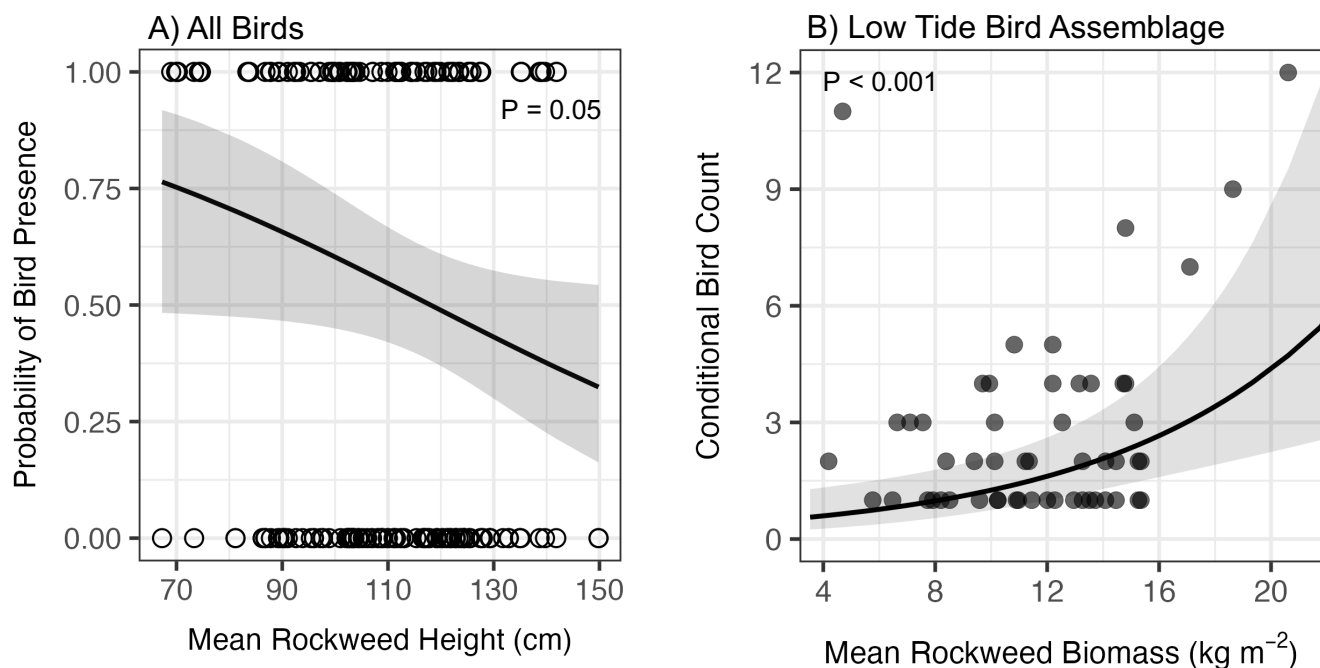


Figure 4.5. Relationships between mean rockweed bed characteristics and birds' patch visitation. Lines were derived from predictions based on statistically significant model components and the gray envelopes represent 95% confidence intervals. The points in each plot are the raw data.

4.5. Discussion

We examined the effect of commercial seaweed harvest on patch visitation by birds at 40 sites along a North Atlantic intertidal zone. Overall, we observed low patch visitation across treatments and tidal heights in our study. Sixty percent of surveys recorded no birds, and when birds visited patches, they were often in small groups (mean individuals per species per site visit $\pm$ SD = 2.1 ± 1.9 individuals). As a result, the effect sizes of harvest were small in comparison to the relative commonness of frequently observed species (e.g., Common Eider, American Crow, Herring Gull) and some confidence intervals overlapped zero. The only supported effect size of harvest was an increase of 1.2 birds (95% CI = 0.14 – 2.37) at impacted sites during the post-harvest recovery interval. Independent of our treatment assignments, bird presence in a patch was less likely at sites with longer mean rockweed height. When we grouped birds into tidal height assemblages, the effects of harvest on the overall bird assemblage appeared to be driven more strongly by the response of the low tide assemblage. However, this group was also the only to exhibit a positive relationship between abundance at visited patches and rockweed biomass. The high tide bird assemblage displayed no detectable responses to rockweed harvest or either rockweed bed characteristic, which suggests that the range of environmental variation captured by our variables does not affect patch visitation. Our findings support our hypotheses that the effects of commercial rockweed harvest on birds are relatively weak and assemblage-specific, and they also highlight questions about use of rockweed habitats relative to other habitat types within coastal birds' home ranges.

The lack of a treatment effect at recently harvested sites indicates that the disturbance associated with a single commercial harvest event does not immediately effect patch visitation. This suggests that birds were equally likely to arrive at recently harvested patches as control

patches, and that they did not spend less time there relative to control patches on average. However, assessing whether there were behavioral changes due to harvest during equal-length stays in patches would require further data on bird behavior and prey densities. The small positive effect of harvest on birds' patch visitation during the post-harvest recovery interval could be a response to increases in the abundances of macroinvertebrate prey during this period (Braun 2022). In other systems, disturbances such as fire (Evans 1984, Panzer 2002, Joern 2005), flooding (Swanson et al. 1998, Gallardo et al. 2009), and resource harvest (Wilzbach et al. 2005, Wootton 2012) can result in positive biotic responses in lower trophic level taxa during post-disturbance recovery. This could result in positive bottom-up forces that diminish across successively higher trophic levels (Borer et al. 2006) and result in relatively weak positive effects on top-level consumers, which would align with our findings. Overall, the effects of commercial rockweed harvest on intertidal birds were small, but if similar intensities of harvest occurred in areas with higher bird densities there may be larger changes in patch visitation.

The two supported relationships between rockweed bed characteristics and birds' patch visitation indicate that these top consumers are responsive to certain aspects of habitat structure in this system. While our BACI experiment tested the effects of commercial rockweed harvest in Maine, the discrete grouping of control and impact treatments did not capture the full variation in rockweed height and biomass across sites. For instance, we observed decreases and increases in mean values of both rockweed characteristics at some control and impact sites, respectively, likely due to factors such as natural disturbance and high spatial heterogeneity of harvest (Johnston et al. 2023). The negative relationship between the probability of bird presence in a patch and mean rockweed height could represent either an indirect relationship in which rockweed height influences prey distributions or abundances, or a direct relationship whereby

foraging efficiency is moderated in part by the mean height of the bed. Among sites with more diverse intertidal seaweed assemblages, Common Eider presence at sites was positively associated with shorter-canopied seaweeds (Johnston et al. 2021). For benthivores in both tidal height assemblages, seaweed habitats with shorter canopies may improve their ability to locate and capture prey. It is important to note that this relationship had marginal statistical support ($P = 0.0501$) and that the 95% confidence interval included equal probabilities of site presence and absence across the entire range of mean rockweed heights (Figure 4.5A). Future investigations of this relationship would be strengthened by measures of foraging activity and larger sample sizes that allow for species-level analyses.

We also observed a positive relationship between conditional counts of the low tide assemblage and mean rockweed biomass in patches. This may be in response to positive correlations between rockweed biomass and the densities of some macroinvertebrate prey taxa (Johnson & Scheibling 1987, H. Mittelstaedt unpublished data) that result from increases in rockweed surface area (i.e., epifaunal species) or suitable refuge from predators. Notably, our observed relationship provides evidence for an upper limit on conditional counts of the low tide assemblage based on mean rockweed biomass in a patch (Figure 4.5B). While we removed three large flocks of this assemblage from analyses, two of these were migratory shorebirds. Migrants may be more likely to visit patches independent of patch quality than resident species due to limited knowledge of local resource, particularly outside of core stopover sites with higher interannual site fidelity (Petit 2000, Németh and Moore 2007).

Upper limits on species abundances set by environmental gradients are common in foraging ecology and result from constraints imposed by limiting environmental factors (Lancaster and Belyea 2006). The most frequently observed species in the low tide assemblage—

American Crow and Herring Gull—are habitat and dietary generalists for which variation in presence and sub-maximal abundance in rockweed patches is determined by environmental variables not measured in this study. However, the maximum abundance of this assemblage in rockweed beds may be determined by the maximum density of prey. This relationship is indicative of positive bottom-up forces in rockweed habitats that highlights the role of a foundation species in indirectly influencing high-level consumers. Our findings that lower mean rockweed height increases the probability of bird presence while greater mean biomass increases conditional counts for the low tide assemblage may be antagonistic from a management perspective, particularly without further behavioral data. Future research that considers additional structural characteristics of rockweed beds (e.g., mean circumference, density, degree of branching) will be beneficial in disentangling these effects.

When we consider patch visitation across all of our sites, perhaps the most notable finding was the high incidence of zero counts. In part, this could be a methodological outcome. When the size of home ranges are large relative to patch sizes and the duration of surveys relatively short, animal locations are unlikely to overlap with a large number of patches (Thomas and Taylor 2006). On the other hand, common zero counts could be biologically meaningful and point to the fact that all bird species recorded in our study occupy much wider ecological niches than rockweed habitats, and rocky intertidal zones in general. There are obligate habitat specialists in other intertidal ecosystems in Maine, such as Piping Plovers (*Charadrius melodus*) along sandy beaches and Saltmarsh Sparrows (*Ammospiza caudacuta*) in tidal saltmarshes, but there are no rocky intertidal specialists in our region outside of Purple Sandpipers (*Calidris maritima*), which are only present in the winter outside of our survey window. Birds in the high tide assemblage were frequently observed in subtidal habitats even during periods of intertidal

inundation (e.g., Common Eider, Double-crested Cormorant), and common low tide species (e.g., American Crow, Herring Gull) are habitat and dietary generalists that are commonly found in supratidal or upland habitats during all tidal stages. A small relative contribution of rockweed habitats toward the total resource needs of Maine coastal birds could lead to low bird densities in these habitats, but this needs to be more rigorously tested. The low bird counts in the Penobscot region (Fig. 3B) suggest that spatial distributions of coastal birds in Maine are unlikely to be uniform, and patch visitation in rockweed habitats is likely partially a function of regional bird densities or distances to roost sites outside of the intertidal.

Despite partially overlapping with the southward migratory period of shorebirds, our surveys did not record enough patch visits in this group (see Figure D.2 for a species list) to robustly assess their response to rockweed harvest. Migratory shorebirds are highly spatiotemporally variable and may require a different survey methodology than ours to best capture their use of rockweed habitats. In addition to better aligning the full survey window with their migratory period, modifications could include longer observation periods due to the use of multiple habitat types across the full tidal cycle, larger patch sizes that encompass more contiguous habitat, and greater proximity to high-use stopover and roost sites (Alerstam et al. 1992, Burger et al. 1997). Despite these challenges, understanding the effects of rockweed harvest on shorebirds should remain a high priority. Migratory shorebirds lack the local knowledge that resident species have about patch quality and are under greater energetic constraints, which could both make changes in patch quality more impactful to them relative to resident species. Furthermore, shorebird presence in seaweed-dominated intertidal zones has been positively correlated with the depth of seaweed beds and the percent cover of rockweed in the seaweed assemblage (Johnston et al. 2021). Obtaining high resolution habitat use data across

longer intervals with satellite-tracked individuals may clarify the role of rocky intertidal habitats in the daily, seasonal, and annual cycles of coastal birds (Schwemmer et al. 2016, Stantial and Cohen 2020).

Most bird populations are subject to a multitude of limiting factors of varying strengths (Newton 1998), and whether commercial rockweed harvest represents a biologically meaningful limiting factor relative to other such factors is beyond the scope of our study. However, among the habitat use indices considered in this study, the small effect sizes of harvest on the full intertidal assemblage either had confidence intervals that crossed zero or were positive. The relationship between the disturbances commonly caused by commercial rockweed harvest in Maine and intertidal bird visitation appears unlikely to be of sufficient magnitude to cause population-level impacts in the region's resident bird species. Whether this changes over repeated harvesting of a patch or expansion of harvest activities is presently unclear. A few previous studies have found seaweed harvest to negatively affect waterbird (Double-crested Cormorant and Common Eider) foraging efficiency, but the applicability of their protocols to commercial rockweed harvest in our region is low. Seaweeds were either completely removed by scraping the substrate (kelp harvest; Lorentsen et al. 2010) or cut uniformly with hand shears at harvest intensities much higher than those typical of commercial harvest in our region (rockweed; Hamilton & Nudds 2003).

Further research is needed to more confidently incorporate a high trophic-level consumer group (birds) into an EBM framework of this rocky intertidal resource harvest. There is a robust literature on the impacts of rockweed harvest on the target resource (Baardseth 1970, Keser et al. 1981, Lazo and Chapman 1996, Kelly et al. 2001, Ugarte et al. 2006, Gendron et al. 2018, Lauzon-Guay et al. 2021) and macroinvertebrate assemblage (Fegley 2001, Trott & Larsen 2012,

Phillippi et al. 2014, Walder 2015, Braun 2022), but, to our knowledge, there are no comparable assemblage-level bird studies in this system that can provide context to our results. While the theory of EBM in marine ecosystems is well developed, gaps remain in evaluating biotic interactions and responses to anthropogenic disturbances (Garcia and Cochrane 2005, O'Boyle et al. 2005, Lester et al. 2010, Carey et al. 2014, Dannheim et al. 2020). Our study prioritized the replication of sites across a large geographic area and selected sites based on their suitability to rockweed harvest, but future studies of rockweed harvest-bird interactions may modify elements of our survey design (i.e., behavioral protocol, longer observation periods) and conduct more surveys at fewer sites that are selected in part based on their potential for bird use.

REFERENCES

- Ainsworth, G. B., and G. L. Burns (2020). ‘Although I use science, it’s an emotional thing’: conservation practitioners’ use of positive affect to frame messages about threatened birds. *Australasian Journal of Environmental Management* 27:351–377.
- Alberdi, A., O. Aizpurua, K. Bohmann, S. Gopalakrishnan, C. Lynggaard, M. Nielsen, and M. T. P. Gilbert (2019). Promises and pitfalls of using high-throughput sequencing for diet analysis. *Molecular Ecology Resources* 19:327–348.
- Alerstam, T., G. A. Gudmundsson, and K. Johannesson (1992). Resources for Long Distance Migration: Intertidal Exploitation of *Littorina* and *Mytilus* by Knots *Calidris Canutus* in Iceland. *Oikos* 65:179–189.
- Anderson, C. M., H. G. Gilchrist, R. A. Ronconi, K. R. Shlepr, D. E. Clark, D. V. C. Weseloh, G. J. Robertson, and M. L. Mallory (2019). Winter home range and habitat selection differs among breeding populations of herring gulls in eastern North America. *Movement Ecology* 7:1–11.
- Ang, P. O., G. J. Sharp, and R.E. Semple (1993). Changes in the population structure of *Ascophyllum nodosum* (L.) Le Jolis due to mechanical harvesting. *Hydrobiologia* 260/261:321–326.
- Ang, P. O., G. J. Sharp, and R. E. Semple (1996). Comparison of the structure of populations of *Ascophyllum nodosum* (Fucales, Phaeophyta) at sites with different harvesting histories. *Hydrobiologia* 326–327:179–184.
- Arkema, K. K., S. C. Abramson, and B. M. Dewsbury (2006). Marine ecosystem-based management: from characterization to implementation. *Frontiers in Ecology and the Environment* 4:525–532.
- Austin, G. E., and M. M. Rehfish (2005). Shifting nonbreeding distributions of migratory fauna in relation to climatic change. *Global Change Biology* 11:31–38.
- Baardseth, E. (1970). Synopsis of biological data on knobbed wrack: *Ascophyllum nodosum* (Linnaeus) Le Jolis. Food and Agriculture Organization of the United Nations.
- Bates, D., M. Mächler, B. Bolker, and S. Walker (2015). Fitting Linear Mixed-Effects Models Using lme4. *Journal of Statistical Software* 67:1–48.
- Benedetti-Cecchi, L. (2001). Variability in abundance of algae and invertebrates at different spatial scales on rocky sea shores. *Marine Ecology Progress Series* 215:79–92.
- Black, R., and R. J. Miller (1991). Use of the intertidal zone by fish in Nova Scotia. *Environmental Biology of Fishes* 31:109–121.

- Blinn, B. M., A.W. Diamond, and D. J. Hamilton (2008). Factor Affecting Selection of Brood-rearing Habitat by Common Eider (*Somateria mollissima*) in the Bay of Fundy, New Brunswick, Canada. *Waterbirds* 31:520-529.
- Boaden, P. J. S., and M. T. Dring (1980). A quantitative evaluation of the effects of *Ascophyllum* harvesting on the littoral ecosystem. *Helgoländer Meeresuntersuchungen* 33:700–710.
- Bohonak, A. J., and H. H. Whiteman (1999). Dispersal of the fairy shrimp *Branchinecta coloradensis* (Anostraca): Effects of hydroperiod and salamanders. *Limnology and Oceanography* 44:487–493.
- Bolyen, E., J. R. Rideout, M. R. Dillon, N. A. Bokulich, C. C. Abnet, G. A. Al-Ghalith, H. Alexander, E. J. Alm, M. Arumugam, F. Asnicar, Y. Bai, et al. (2019). Reproducible, interactive, scalable and extensible microbiome data science using QIIME 2. *Nature Biotechnology* 37:852–857.
- Bonin, M., C. Dussault, J. Taillon, N. Lecomte, and S. D. Côté (2020). Combining stable isotopes, morphological, and molecular analyses to reconstruct the diet of free-ranging consumers. *Ecology and Evolution* 10:6664–6676.
- Borer, E. T., B. S. Halpern, and E. W. Seabloom (2006). Asymmetry in community regulation: effects of predators and productivity. *Ecology* 87:2813–2820.
- Bourbour, R. P., C. M. Aylward, C. W. Tyson, B. L. Martinico, A. M. Goodbla, T. E. Ely, A. M. Fish, A. C. Hull, and J. M. Hull (2021). Falcon fuel: metabarcoding reveals songbird prey species in the diet of juvenile Merlins (*Falco columbarius*) migrating along the Pacific Coast of western North America. *Ibis* 163:1282–1293.
- Braun, L. (2022) Understanding the Impacts of Anthropogenic Effects and Habitat Variability Interactions on Maine's Rocky Intertidal Ecosystem. Master of Science Thesis. University of Maine, Maine, USA.
- Breed, G. A., I. D. Jonsen, R. A. Myers, W. D. Bowen, and M. L. Leonard (2009). Sex-specific, seasonal foraging tactics of adult grey seals (*Halichoerus grypus*) revealed by state–space analysis. *Ecology* 90:3209–3221.
- Brooks, M., E., K. Kristensen, K. Benthem J. ,van, A. Magnusson, C. Berg W., A. Nielsen, H. Skaug J., M. Mächler, and B. Bolker M. (2017). glmmTMB Balances Speed and Flexibility Among Packages for Zero-inflated Generalized Linear Mixed Modeling. *The R Journal* 9:378–400.
- Brun, L., J. Schneider, E. M. Carrió, P. Dongre, P. Taberlet, V. D. Waal, and L. Fumagalli (2022). Focal vs. fecal: Seasonal variation in the diet of wild vervet monkeys from observational and DNA metabarcoding data. *Ecology and Evolution* 12:e9358.
- Burger, J., L. Niles, and K. E. Clark (1997). Importance of beach, mudflat and marsh habitats to migrant shorebirds on Delaware Bay. *Biological Conservation* 79:283–292.

- Burton, N. H. K., and P. R. Evans (1997). Survival and winter site-fidelity of Turnstones *Arenaria interpres* and Purple Sandpipers *Calidris maritima* in northeast England. *Bird Study* 44:35–44.
- Cáceres, M. D., and P. Legendre (2009). Associations between species and groups of sites: indices and statistical inference. *Ecology* 90:3566–3574.
- Cáceres, M. D., P. Legendre, and M. Moretti (2010). Improving indicator species analysis by combining groups of sites. *Oikos* 119:1674–1684.
- Callahan, B. J., P. J. McMurdie, M. J. Rosen, A. W. Han, A. J. A. Johnson, and S. P. Holmes (2016). DADA2: High-resolution sample inference from Illumina amplicon data. *Nature Methods* 13:581–583.
- Carey, M. P., P. S. Levin, H. Townsend, T. J. Minello, G. R. Sutton, T. B. Francis, C. J. Harvey, J. E. Toft, K. K. Arkema, J. L. Burke, C.-K. Kim, et al. (2014). Characterizing coastal foodwebs with qualitative links to bridge the gap between the theory and the practice of ecosystem-based management. *ICES Journal of Marine Science* 71:713–724.
- Chan, Y.-C., H.-B. Peng, Y.-X. Han, S. S.-W. Chung, J. Li, L. Zhang, and T. Piersma (2019). Conserving unprotected important coastal habitats in the Yellow Sea: Shorebird occurrence, distribution and food resources at Lianyungang. *Global Ecology and Conservation* 20:e00724.
- Chesson, J. (1978). Measuring Preference in Selective Predation. *Ecology* 59:211–215.
- Chesson, J. (1983). The Estimation and Analysis of Preference and Its Relationship to Foraging Models. *Ecology* 64:1297–1304.
- Clarke, K. (1993) Non-parametric multivariate analyses of changes in community structure. *Australian Journal of Ecology* 18:117–43.
- Cook, A. G., and R. F. Denno (1994). Planthopper plant interactions: feeding behavior, plant nutrition, plant defense and host plant specialization. In *Planthoppers: Their Ecology and Management* (R. F. Denno and T. Perfect, Editors). Chapman & Hall, New York, pp. 114–39.
- Cousens, R. (1985). Frond size distributions and the effects of the algal canopy on the behaviour of *Ascophyllum nodosum* (L.) Le Jolis. *Journal of Experimental Marine Biology and Ecology* 92:231–249.
- Cuff, J. P., F. M. Windsor, M. P. T. G. Tercel, J. J. N. Kitson, and D. M. Evans (2022). Overcoming the pitfalls of merging dietary metabarcoding into ecological networks. *Methods in Ecology and Evolution* 13:545–559.
- Dannheim, J., L. Bergström, S. N. R. Birchenough, R. Brzana, A. R. Boon, J. W. P. Coolen, J.-C. Dauvin, I. De Mesel, J. Derweduwen, A. B. Gill, Z. L. Hutchison, et al. (2020). Benthic

- effects of offshore renewables: identification of knowledge gaps and urgently needed research. *ICES Journal of Marine Science* 77:1092–1108.
- Darimont, C. T., P. C. Paquet, and T. E. Reimchen (2009). Landscape heterogeneity and marine subsidy generate extensive intrapopulation niche diversity in a large terrestrial vertebrate. *Journal of Animal Ecology* 78:126–133.
- De Pascalis, F., S. Imperio, A. Benvenuti, C. Catoni, D. Rubolini, and J. G. Cecere (2020). Sex-specific foraging behaviour is affected by wind conditions in a sexually size dimorphic seabird. *Animal Behaviour* 166:207–218.
- Denno, R. F., L. W. Douglass, and D. Jacobs (1986). Effects of Crowding and Host Plant Nutrition on a Wing-Dimorphic Planthopper. *Ecology* 67:116–123.
- Denno, R. F., M. J. Raupp, D. W. Tallamy, and C. F. Reichelderfer (1980). Migration in Heterogeneous Environments: Differences in Habitat Selection Between the Wing Forms of the Dimorphic Planthopper, *Prokelisia Marginata* (Homoptera: Delphacidae). *Ecology* 61:859–867.
- Denoël, M., H. H. Whiteman, and S. A. Wissinger (2007). Foraging tactics in alternative heterochronic salamander morphs: trophic quality of ponds matters more than water permanency. *Freshwater Biology* 52:1667–1676.
- DFO (1999). The impact of rockweed harvest on the habitat of southwest New Brunswick. DFO Maritimes Regional Habitat Status Report 99/2E.
- Dierschke, V. (1994). Food and feeding ecology of purple sandpipers *Calidris maritima* on rocky intertidal habitats (Helgoland, German Bight). *Netherlands Journal of Sea Research* 31:309–317.
- Dodson, S. I. (1970). Complementary Feeding Niches Sustained by Size-Selective Predation1. *Limnology and Oceanography* 15:131–137.
- Dudgeon, S., and P. S. Petraitis (2005). First year demography of the foundation species, *Ascophyllum nodosum*, and its community implications. *Oikos* 109:405–415.
- Dugan, J. E., D. M. Hubbard, M. D. McCrary, and M. O. Pierson (2003). The response of macrofauna communities and shorebirds to macrophyte wrack subsidies on exposed sandy beaches of southern California. *Estuarine, Coastal and Shelf Science* 58S:25–40.
- Ens, N. J., B. Harvey, M. M. Davies, H. M. Thomson, K. J. Meyers, J. Yakimishyn, L. C. Lee, M. E. McCord, and T. G. Gerwing (2022). The Green Wave: reviewing the environmental impacts of the invasive European green crab (*Carcinus maenas*) and potential management approaches. *Environmental Reviews* 30:306–322.
- Eriksen, C. H., and D. Belk (1999). Fairy shrimps of California's puddles, pools, and playas. Mad River Press, Eureka, California.

- Evans, E. W. (1984). Fire as a Natural Disturbance to Grasshopper Assemblages of Tallgrass Prairie. *Oikos* 43:9.
- Evans, P. R., and P. C. Smith (1975). Studies of shorebirds at Lindisfarne, Northumberland. 2. Fat and pectoral muscle as indicators of body condition in the Bar-tailed Godwit. *Wildfowl* 26:64–76.
- Feare, C. J. (1966). The winter feeding of the Purple Sandpiper. *British Birds* 59:165–179.
- Fegley, J. C. (2001). Ecological implications of rockweed, *Ascophyllum nodosum* (L.) Le Jolis, harvesting. Ph.D. Thesis. University of Maine, Maine, USA.
- Fegley, J. C. (2006). Morphological, Population and Biomass Studies of Rockweed (*Ascophyllum nodosum*) in Quahog Bay and Taunton Bay. Coastal Environmental Consulting and Planning.
- Forsman, A. M., B. D. Hoenig, S. A. Gaspar, J. D. Fischer, J. Siegrist, and K. Fraser (2022). Evaluating the impacts of metabarcoding primer selection on DNA characterization of diet in an aerial insectivore, the Purple Martin. *Ornithology* 139:1–17.
- Frank, S. A., and M. Slatkin (1990). Evolution in a Variable Environment. *The American Naturalist* 136:244–260.
- Gallardo, B., S. Gascón, M. García, and F. A. Comín (2009). Testing the response of macroinvertebrate functional structure and biodiversity to flooding and confinement. *Journal of Limnology* 68:315–326.
- Garcia, S. M., and K. L. Cochrane (2005). Ecosystem approach to fisheries: a review of implementation guidelines. *ICES Journal of Marine Science* 62:311–318.
- Gardner, J. L., E. Rowley, P. de Rebeira, A. de Rebeira, and L. Brouwer (2017). Effects of extreme weather on two sympatric Australian passerine bird species. *Philosophical Transactions of the Royal Society B: Biological Sciences* 372:20160148.
- Gendron, L., A. Merzouk, P. Bergeron, and L. E. Johnson (2018). Managing disturbance: the response of a dominant intertidal seaweed *Ascophyllum nodosum* (L.) Le Jolis to different frequencies and intensities of harvesting. *Journal of Applied Phycology* 30:1877–1892.
- Gerwing, T. G., J.-H. Kim, D. J. Hamilton, M. A. Barbeau, and J. A. Addison (2016). Diet reconstruction using next-generation sequencing increases the known ecosystem usage by a shorebird. *The Auk: Ornithological Advances* 133:168–177.
- Glude, J. B. (1955). The Effects of Temperature and Predators on the Abundance of the Soft-Shell Clam, *Mya Arenaria*, in New England. *Transactions of the American Fisheries Society* 84:13–26.
- Goss-Custard, J. D. (1977). Optimal foraging and the size selection of worms by redshank, *Tringa totanus*, in the field. *Animal Behaviour* 25:10–29.

- Gossner, M. M., T. M. Lewinsohn, T. Kahl, F. Grassein, S. Boch, D. Prati, K. Birkhofer, S. C. Renner, J. Sikorski, T. Wubet, H. Arndt, et al. (2016). Land-use intensification causes multitrophic homogenization of grassland communities. *Nature* 540:266–269.
- Granquist, S. M., R. Esparza-Salas, E. Hauksson, O. Karlsson, and A. Angerbjörn (2018). Fish consumption of harbour seals (*Phoca vitulina*) in north western Iceland assessed by DNA metabarcoding and morphological analysis. *Polar Biology* 41:2199–2210.
- Green, R. H. (1993). Application of repeated measures designs in environmental impact and monitoring studies. *Australian Journal of Ecology* 18:81–98.
- Grindal, S. D., and R. M. Brigham (1999). Impacts of forest harvesting on habitat use by foraging insectivorous bats at different spatial scales. *Écoscience* 6:25–34.
- Guiry, M. D., and L. Morrison (2013). The sustainable harvesting of *Ascophyllum nodosum* (Fucaceae, Phaeophyceae) in Ireland, with notes on the collection and use of some other brown algae. *Journal of Applied Phycology* 25:1823–1830.
- Hall, S. J., and B. Mainprize (2004). Towards ecosystem-based fisheries management. *Fish and Fisheries* 5:1–20.
- Hartig, F., L. Lohse (2022) DHARMa: Residual Diagnostics for Hierarchical (Multi-Level/Mixed) Regression Models. R package version 0.4.6. Available from <https://CRAN.R-project.org/package=DHARMa>.
- Hamilton, D. J. (2001). Feeding Behavior of Common Eider Ducklings in Relation to Availability of Rockweed Habitat and Duckling Age. *Waterbirds* 24:233–241.
- Hamilton, D., and T. Nudds (2003). Effects of predation by common eiders (*Somateria mollissima*) in an intertidal rockweed bed relative to an adjacent mussel bed. *Marine Biology* 142:1–12.
- Hedrick, P. W. (1986). Genetic Polymorphism in Heterogeneous Environments: A Decade Later. *Annual Review of Ecology and Systematics* 17:535–566.
- Herring, G., and J. A. Collazo (2005). Habitat Use, Movements and Home Range of Wintering Lesser Scaup in Florida. *Waterbirds* 28:71–78.
- Heyman, E. (2010). Clearance of understory in urban woodlands: Assessing impact on bird abundance and diversity. *Forest Ecology and Management* 260:125–131.
- Hicklin, P. W. (1987). The migration of shorebirds in the Bay of Fundy. *The Wilson Bulletin* 99:540–570.
- Hidasi-Neto, J., D. C. Joner, F. Resende, L. de M. Monteiro, F. V. Faleiro, R. D. Loyola, and M. V. Cianciaruso (2019). Climate change will drive mammal species loss and biotic homogenization in the Cerrado Biodiversity Hotspot. *Perspectives in Ecology and Conservation* 17:57–63.

- Hoenig, B. D., A. M. Snider, A. M. Forsman, K. A. Hobson, S. C. Latta, E. T. Miller, M. J. Polito, L. L. Powell, S. L. Rogers, T. W. Sherry, D. P. L. Toews, et al. (2022). Current methods and future directions in avian diet analysis. *Ornithology* 139:1–28.
- Holsman, K. K., A. C. Haynie, A. B. Hollowed, J. C. P. Reum, K. Aydin, A. J. Hermann, W. Cheng, A. Faig, J. N. Ianelli, K. A. Kearney, and A. E. Punt (2020). Ecosystem-based fisheries management forestalls climate-driven collapse. *Nature Communications* 11:4579.
- Holt, R. D., and T. H. Keitt (2000). Alternative causes for range limits: a metapopulation perspective. *Ecology Letters* 3:41–47.
- Horswill, C., J. A. Jackson, R. Medeiros, R. W. Nowell, P. N. Trathan, and T. C. O’Connell (2018). Minimising the limitations of using dietary analysis to assess foodweb changes by combining multiple techniques. *Ecological Indicators* 94:218–225.
- Ingala, M. R., N. B. Simmons, C. Wultsch, K. Krampis, K. L. Provost, and S. L. Perkins (2021). Molecular diet analysis of neotropical bats based on fecal DNA metabarcoding. *Ecology and Evolution* 11:7474–7491.
- Jansson, C., J. Ekman, and A. von Brömssen (1981). Winter Mortality and Food Supply in Tits *Parus* spp. *Oikos* 37:313–322.
- Joern, A. (2005). Disturbance By Fire Frequency and Bison Grazing Modulate Grasshopper Assemblages in Tallgrass Prairie. *Ecology* 86:861–873.
- Johnson, D. H. (1980). The Comparison of Usage and Availability Measurements for Evaluating Resource Preference. *Ecology* 61:65–71.
- Johnson, S. C., and R. E. Scheibling (1987). Structure and dynamics of epifaunal assemblages on intertidal macroalgae *Ascophyllum nodosum* and *Fucus vesiculosus* in Nova Scotia, Canada. *Marine Ecology Progress Series* 37:209–227.
- Johnston, E., A. Klemmer, E. Blomberg, A. Baron, V. Watson, L. Tudor, L. Welch, and B. Olsen (2021). Macroalgae composition alters occupancy of multiple bird guilds in rocky intertidal communities. *Marine Ecology Progress Series* 659:29–47.
- Johnston, E. M., H. N. Mittelstaedt, L. A. Braun, J. F. Muhlin, B. J. Olsen, H. M. Webber, and A. J. Klemmer (2023). Bed-scale impact and recovery of a commercially important intertidal seaweed. *Journal of Experimental Marine Biology and Ecology* 561:151869.
- Joly, P. (1987) Le régime alimentaire des amphibiens: méthodes d’étude. *Alytes* 6:11–17.
- Jusino, M. A., M. T. Banik, J. M. Palmer, A. K. Wray, L. Xiao, E. Pelton, J. R. Barber, A. Y. Kawahara, C. Gratton, M. Z. Peery, and D. L. Lindner (2019). An improved method for utilizing high-throughput amplicon sequencing to determine the diets of insectivorous animals. *Molecular Ecology Resources* 19:176–190.

- Kay, L. (2015). Canopy and Community Structure of Rockweed Beds in Nova Scotia and New Brunswick: Regional Variation and Effects of Commercial Harvest and Proximity to Aquaculture.
- Kay, L. M., T. D. Eddy, A. L. Schmidt, and H. K. Lotze (2016). Regional differences and linkage between canopy structure and community composition of rockweed habitats in Atlantic Canada. *Marine Biology* 163:251.
- Kelly, L., L. Collier, M. J. Costello, M. Diver, S. McGarvey, S. Kraan, J. Morrissey, and M. D. Guiry (2001). Impact Assessment of Hand and Mechanical Harvesting of *Ascophyllum nodosum* on Regeneration and Biodiversity. Marine Institute Marine Resource Series No. 19.
- Keser, M., R. L. Vadas, and B. R. Larson (1981). Regrowth of *Ascophyllum nodosum* and *Fucus vesiculosus* under various harvesting regimes in Maine, USA. *Botanica Marina* 24:29–38.
- Kitaysky, A. S., E. V. Kitaikaia, J. F. Piatt, and J. C. Wingfield (2006). A mechanistic link between chick diet and decline in seabirds? *Proceedings of the Royal Society B: Biological Sciences* 273:445–450.
- Krebs, J. R., J. T. Erichsen, M. I. Webber, and E. L. Charnov (1977). Optimal prey selection in the great tit (*Parus major*). *Animal Behaviour* 25:30–38.
- Krehenwinkel, H., M. Wolf, J. Y. Lim, A. J. Rominger, W. B. Simison, and R. G. Gillespie (2017). Estimating and mitigating amplification bias in qualitative and quantitative arthropod metabarcoding. *Scientific Reports* 7:17668.
- Kuznetsova, A., P. B. Brockhoff, and R. H. B. Christensen (2017). lmerTest Package: Tests in Linear Mixed Effects Models. *Journal of Statistical Software* 82.
- Kwon, E., E. L. Weiser, R. B. Lanctot, S. C. Brown, H. R. Gates, G. Gilchrist, S. J. Kendall, D. B. Lank, J. R. Liebezeit, L. McKinnon, E. Nol, et al. (2019). Geographic variation in the intensity of warming and phenological mismatch between Arctic shorebirds and invertebrates. *Ecological Monographs* 89:1–20.
- Lackey, A. C. R., M. P. Moore, J. Doyle, N. Gerlanc, A. Hagan, M. Geile, C. Eden, and H. H. Whiteman (2019). Lifetime Fitness, Sex-Specific Life History, and the Maintenance of a Polyphenism. *The American Naturalist* 194:230–245.
- Lancaster, J., and L. R. Belyea (2006). Defining the limits to local density: alternative views of abundance-environment relationships. *Freshwater Biology* 51:783–796.
- Lauzon-Guay, J.-S., R. A. Ugarte, B. L. Morse, and C. A. Robertson (2021). Biomass and height of *Ascophyllum nodosum* after two decades of continuous commercial harvesting in eastern Canada. *Journal of Applied Phycology*. <https://doi.org/10.1007/s10811-021-02427-x>

- Lazo, L., and A. R. O. Chapman (1996). Effects of harvesting on *Ascophyllum nodosum* (L.) Le Jol. (Fucales, Phaeophyta): a demographic approach. *Journal of Applied Phycology* 8:87–103.
- Leaver, J., J. C. Carstens, and M. I. Cherry (2021). Impact of informal timber harvesting on habitat structure and bird assemblages in a coastal forest of the Eastern Cape, South Africa. *Bird Conservation International* 31:219–238.
- Lenth, R., P. Buerkner, M. Herve, J. Love, H. Reibl, and H. Singmann (2020) Estimated marginal means. R package version 1.5.2-1. Available from <https://CRAN.R-project.org/package=emmeans>.
- Leonard, T. D., P. D. Taylor, and I. G. Warkentin (2008). Landscape Structure and Spatial Scale Affect Space Use by Songbirds in Naturally Patchy and Harvested Boreal Forests. *The Condor* 110:467–481.
- Leray, M., C. P. Meyer, and S. C. Mills (2015). Metabarcoding dietary analysis of coral dwelling predatory fish demonstrates the minor contribution of coral mutualists to their highly partitioned, generalist diet. *PeerJ* 3:e1047.
- Lesser, M. P. (2016). Climate change stressors cause metabolic depression in the blue mussel, *Mytilus edulis*, from the Gulf of Maine. *Limnology and Oceanography* 61:1705–1717.
- Lester, S. E., K. L. McLeod, H. Tallis, M. Ruckelshaus, B. S. Halpern, P. S. Levin, F. P. Chavez, C. Pomeroy, B. J. McCay, C. Costello, S. D. Gaines, et al. (2010). Science in support of ecosystem-based management for the US West Coast and beyond. *Biological Conservation* 143:576–587.
- Levins, R. (1969). Some Demographic and Genetic Consequences of Environmental Heterogeneity for Biological Control. *Bulletin of the Entomological Society of America* 15:237–240.
- Liang, C., G. Yang, N. Wang, G. Feng, F. Yang, J.-C. Svenning, and J. Yang (2019). Taxonomic, phylogenetic and functional homogenization of bird communities due to land use change. *Biological Conservation* 236:37–43.
- Lima, S. L., and P. A. Zollner (1996). Towards a behavioral ecology of ecological landscapes. *Trends in Ecology & Evolution* 11:131–135.
- Lind, M. I., and F. Johansson (2007). The degree of adaptive phenotypic plasticity is correlated with the spatial environmental heterogeneity experienced by island populations of *Rana temporaria*. *Journal of Evolutionary Biology* 20:1288–1297.
- Liu, G., A. B. A. Shafer, X. Hu, L. Li, Y. Ning, M. Gong, L. Cui, H. Li, D. Hu, L. Qi, H. Tian, and B. Wang (2018). Meta-barcoding insights into the spatial and temporal dietary patterns of the threatened Asian Great Bustard (*Otis tarda dybowskii*) with potential implications for diverging migratory strategies. *Ecology and Evolution* 8:1736–1745.

- Lorentsen, S.-H., K. Sjøtun, and D. Grémillet (2010). Multi-trophic consequences of kelp harvest. *Biological Conservation* 143:2054–2062.
- Losey, J. E., J. Harmon, F. Ballantyne, and C. Brown (1997). A polymorphism maintained by opposite patterns of parasitism and predation. *Nature* 388:269–272.
- Lotze, H. K., I. Milewski, J. Fast, L. Kay, and B. Worm (2019). Ecosystem-based management of seaweed harvesting. *Botanica Marina* 62:395–409.
- MacKenzie, D. I., J. D. Nichols, G. B. Lachman, S. Droege, J. A. Royle, and C. A. Langtimm (2002). Estimating Site Occupancy Rates When Detection Probabilities Are Less Than One. *Ecology* 83:2248–2255.
- Mafrá Jr., L. L., and S. R. Cunha (2006). *Sargassum Cymosum* (Phaeophyceae) in Southern Brazil: Seasonality of Biomass, Recovery After Harvest and Alginate Yield. *Journal of Coastal Research*:1847–1852.
- Magurran, A. E., M. Dornelas, F. Moyes, N. J. Gotelli, and B. McGill (2015). Rapid biotic homogenization of marine fish assemblages. *Nature Communications* 6:8405.
- Maine Climate Office (2022). Statewide Monthly/Seasonal Temperature and Precipitation. Climate Change Institute, University of Maine, Orono, ME, USA. Available from https://mco.umaine.edu/data_monthly/?cdiv_id=me&var_id=t2anom&mon_id=ann
- Maine Department of Inland Fisheries and Wildlife (2015). Maine’s Wildlife Action Plan. Maine Department of Inland Fisheries and Wildlife, Augusta, ME, USA.
- Mallory, M. L., M. F. Elderkin, N. C. Spencer, T. A. Betsch, A. C. Russell, and P. L. Mills (2016). Diet of *Calidris maritima* (Purple Sandpiper) during the Winter in Nova Scotia, Canada. *Northeastern Naturalist* 23:205–210.
- Manly, B. F. J. (1974). A Model for Certain Types of Selection Experiments. *Biometrics* 30:281–294.
- Marquez, G. P. B., W. J. E. Santiañez, G. C. Trono, M. N. E. Montaña, H. Araki, H. Takeuchi, and T. Hasegawa (2014). Seaweed biomass of the Philippines: Sustainable feedstock for biogas production. *Renewable and Sustainable Energy Reviews* 38:1056–1068.
- Marsh, C. P. (1983). The role of avian predators in an Oregon rocky intertidal community.
- Martin, M. (2011). Cutadapt removes adapter sequences from high-throughput sequencing reads. *EMBnet.journal* 17:10–12.
- Maslo, B., R. L. Mau, K. Kerwin, R. McDonough, E. McHale, and J. T. Foster (2022). Bats provide a critical ecosystem service by consuming a large diversity of agricultural pest insects. *Agriculture, Ecosystems and Environment* 324:107722.

- Matley, J. K., G. E. Maes, F. Devloo-Delva, R. Huerlimann, G. Chua, A. J. Tobin, A. T. Fisk, C. A. Simpfendorfer, and M. R. Heupel (2018). Integrating complementary methods to improve diet analysis in fishery-targeted species. *Ecology and Evolution* 8:9503–9515.
- Matoo, O. B., G. Lannig, C. Bock, and I. M. Sokolova (2021). Temperature but not ocean acidification affects energy metabolism and enzyme activities in the blue mussel, *Mytilus edulis*. *Ecology and Evolution* 11:3366–3379.
- Matsumura, M. (1996). Genetic analysis of a threshold trait: density-dependent wing dimorphism in *Sogatella furcifera* (Horváth) (Hemiptera: Delphacidae), the whitebacked planthopper. *Heredity* 76:229–237.
- McClenaghan, B., E. Nol, and K. C. R. Kerr (2019). DNA metabarcoding reveals the broad and flexible diet of a declining aerial insectivore. *The Auk: Ornithological Advances* 136:1–11.
- McKee, J. (1982). The winter feeding of Turnstones and Purple Sandpipers in Strathclyde. *Bird Study* 29:213–216.
- McLeod, M. J., D. J. Hornbach, S. I. Guttman, C. M. Way, and A. J. Burky (1981). Environmental Heterogeneity, Genetic Polymorphism, and Reproductive Strategies. *The American Naturalist* 118:129–134.
- Menge, B. A., and J. P. Sutherland (1987). Community Regulation: Variation in Disturbance, Competition, and Predation in Relation to Environmental Stress and Recruitment. *The American Naturalist* 130:730–757.
- Minchinton, T. E., and R. E. Scheibling (1991). The Influence of Larval Supply and Settlement on the Population Structure of Barnacles. *Ecology* 72:1867–1879.
- Mittelhauser, G. H., L. Tudor, and B. Connery (2013). Abundance and Distribution of Purple Sandpipers (*Calidris maritima*) Wintering in Maine. *Northeastern Naturalist* 20:219–228.
- Morten, J. M., R. A. Burrell, T. D. Frayling, A. N. Hoodless, W. Thurston, and L. A. Hawkes (2022). Variety in responses of wintering oystercatchers *Haematopus ostralegus* to near-collapse of their prey in the Exe Estuary, UK. *Ecology and Evolution* 12:e9526.
- Németh, Z., and F. R. Moore (2007). Unfamiliar stopover sites and the value of social information during migration. *Journal of Ornithology* 148:369–376.
- Newton, I. (1998). *Population Limitation in Birds*. Academic Press, San Diego, CA, USA.
- Nijhout, H. F. (2003). Development and evolution of adaptive polyphenisms. *Evolution & Development* 5:9–18.
- Niles, L. J., J. Bart, H. P. Sitters, A. D. Dey, K. E. Clark, P. W. Atkinson, A. J. Baker, K. A. Bennett, K. S. Kalasz, N. A. Clark, J. Clark, et al. (2009). Effects of Horseshoe Crab

Harvest in Delaware Bay on Red Knots: Are Harvest Restrictions Working? *BioScience* 59:153–164.

- O’Boyle, R., M. Sinclair, P. Keizer, K. Lee, D. Ricard, and P. Yeats (2005). Indicators for ecosystem-based management on the Scotian Shelf: bridging the gap between theory and practice. *ICES Journal of Marine Science* 62:598–605.
- Oksanen, J., G. Simpson, F. Blanchet, R. Kindt, P. Legendre, P. Minchin, R. O’Hara, P. Solymos, M. Stevens, E. Szoecs, H. Wagner, M. Barbour, M. Bedward, B. Bolker, D. Borcard, G. Carvalho, M. Chirico, M. De Caceres, S. Durand, H. Evangelista, R. FitzJohn, M. Friendly, B. Furneaux, G. Hannigan, M. Hill, L. Lahti, D. McGlinn, M. Ouellette, E. Ribeiro Cunha, T. Smith, A. Stier, C. Ter Braak, and J. Weedon (2017). *vegan*: Community Ecology Package. R package version 2.6-4. Available from <https://CRAN.R-project.org/package=vegan>.
- Orr, K. (2013). Predicting the ecosystem effects of harvesting beach-cast kelp for biofuel.
- Panzer, R. (2002). Compatibility of Prescribed Burning with the Conservation of Insects in Small, Isolated Prairie Reserves. *Conservation Biology* 16:1296–1307.
- Pavia, H., H. Carr, and P. Åberg (1999). Habitat and feeding preferences of crustacean mesoherbivores inhabiting the brown seaweed *Ascophyllum nodosum* (L.) Le Jol. and its epiphytic macroalgae. *Journal of Experimental Marine Biology and Ecology* 236:15–32.
- Payne, L. X., and E. P. Pierce (2020). Purple Sandpiper (*Calidris maritima*), version 1.0. In *Birds of the World* (S.M. Billerman, Editor). Cornell Lab of Ornithology, Ithaca, NY, USA.
- Pereira, L., L. Morrison, P. S. Shukla, and A. T. Critchley (2020). A concise review of the brown macroalga *Ascophyllum nodosum* (Linnaeus) Le Jolis. *Journal of Applied Phycology*. <https://doi.org/10.1007/s10811-020-02246-6>
- Petit, D. (2000). Habitat use by landbirds along Nearctic–Neotropical migration routes: implications for conservation of stopover habitats. *Studies in Avian Biology* 20:15–33.
- Petratis, P. S., and S. R. Dudgeon (2020). Declines over the last two decades of five intertidal invertebrate species in the western North Atlantic. *Communications Biology* 3:591.
- Phillippi, A., K. Tran, and A. Perna (2014). Does intertidal canopy removal of *Ascophyllum nodosum* alter the community structure beneath? *Journal of Experimental Marine Biology and Ecology* 461:53–60.
- Pienkowski, M. (1983). Surface activity of some intertidal invertebrates in relation to temperature and the foraging behaviour of their shorebird predators. *Marine Ecology Progress Series* 11:141–150.
- Pikitch, E. K., C. Santora, E. A. Babcock, A. Bakun, R. Bonfil, D. O. Conover, P. Dayton, P. Doukakis, D. Fluharty, B. Heneman, E. D. Houde, et al. (2004). Ecosystem-Based Fishery Management. *Science* 305:346–347.

- Pineda, J., V. Starczak, and T. Stueckle (2006). Timing of successful settlement: demonstration of a recruitment window in the barnacle *Semibalanus balanoides*. *Marine Ecology Progress Series* 320:233–237.
- Prop, J., and C. Deerenberg (1991). Spring staging in brent geese *Branta bernicla*: feeding constraints and the impact of diet on the accumulation of body reserves. *Oecologia* 87:19–28.
- Pulliam, H. R. (1988). Sources, Sinks, and Population Regulation. *The American Naturalist* 132:652–661.
- Pulliam, H. R. (2000). On the relationship between niche and distribution. *Ecology Letters* 3:349–361.
- Pyke, G. H., H. R. Pulliam, and E. L. Charnov (1977). Optimal Foraging: A Selective Review of Theory and Tests. *The Quarterly Review of Biology* 52:137–154.
- R Core Team (2022) R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna. www.r-project.org.
- Rangeley, R. W., and J. Davies (Editors) (2000). Gulf Of Maine Rockweed: Management in the face of scientific uncertainty. Proceeding of the Global Programme of Action Coalition for the Gulf of Maine (GPAC) workshop. Huntsman Marine Science Centre.
- Rangeley, R. W., and D. L. Kramer (1998). Density-Dependent Antipredator Tactics and Habitat Selection in Juvenile Pollock. *Ecology* 79:943–952.
- Rebours, C., E. Marinho-Soriano, J. A. Zertuche-González, L. Hayashi, J. A. Vásquez, P. Kradolfer, G. Soriano, R. Ugarte, M. H. Abreu, I. Bay-Larsen, G. Hovelsrud, et al. (2014). Seaweeds: an opportunity for wealth and sustainable livelihood for coastal communities. *Journal of Applied Phycology* 26:1939–1951.
- Reed, D. C., A. Rassweiler, and K. K. Arkema (2008). Biomass Rather than Growth Rate Determines Variation in Net Primary Production by Giant Kelp. *Ecology* 89:2493–2505.
- Reed, T. E., R. S. Waples, D. E. Schindler, J. J. Hard, and M. T. Kinnison (2010). Phenotypic plasticity and population viability: the importance of environmental predictability. *Proceedings of the Royal Society B: Biological Sciences* 277:3391–3400.
- Rehfishch, M. M., G. E. Austin, S. N. Freeman, M. J. S. Armitage, and N. H. K. Burton (2004). The possible impact of climate change on the future distributions and numbers of waders on Britain's non-estuarine coast. *Ibis* 146:70–81.
- Reisewitz, S. E., J. A. Estes, and C. A. Simenstad (2006). Indirect food web interactions: sea otters and kelp forest fishes in the Aleutian archipelago. *Oecologia* 146:623–631.

- Reudink, M. W., P. P. Marra, T. K. Kyser, P. T. Boag, K. M. Langin, and L. M. Ratcliffe (2009). Non-breeding season events influence sexual selection in a long-distance migratory bird. *Proceedings of the Royal Society B: Biological Sciences* 276:1619–1626.
- Riemann, L., H. Alfredsson, M. M. Hansen, T. D. Als, T. G. Nielsen, P. Munk, K. Aarestrup, G. E. Maes, H. Sparholt, M. I. Petersen, M. Bachler, and M. Castonguay (2010). Qualitative assessment of the diet of European eel larvae in the Sargasso Sea resolved by DNA barcoding. *Biology Letters* 6:819–822.
- Ritz, C., F. Baty, J. C. Streibig, and D. Gerhard (2015) Dose-Response Analysis Using R. *PLoS One* 10:e014602.
- Robertson, B. A., J. S. Rehage, and A. Sih (2013). Ecological novelty and the emergence of evolutionary traps. *Trends in Ecology & Evolution* 28:552–560.
- Robinson, B. H., H. A. Colett, L. M. Phillips, and A. N. Powell (2018). Are prey remains accurate indicators of chick diet? A comparison of diet quantification techniques for Black Oystercatchers. *Wader Study* 125:20–32.
- Rockweed Plan Development Team, C. Bartlett, S. Redmond, J. Arbuckle, B. Beal, S. Brawley, S. Domizi, L. Mercer, D. Preston, G. Seaver, N. Sferra, P. Thayer, and R. Ugarte (2014). Fishery Management Plan for Rockweed (*Ascophyllum nodosum*). Maine Department of Marine Resources, Augusta, Maine, USA.
- Roff, D. A. (1986). The Evolution of Wing Dimorphism in Insects. *Evolution* 40:1009–1020.
- Rosenberg, C., H. Rothstein, and J. Gurevitch (2013). Effect Sizes: Conventional Choices and Calculations. In: *Handbook of Meta-Analysis in Ecology and Evolution* (eds Koricheva, J., Gurevitch, J., Mengersen, K.). Princeton University Press, Princeton, NJ, pp. 61–71
- Rosenberg, K. V., A. M. Dokter, P. J. Blancher, J. R. Sauer, A. C. Smith, P. A. Smith, J. C. Stanton, A. Panjabi, L. Helft, M. Parr, and P. P. Marra (2019). Decline of the North American avifauna. *Science* 366:120–124.
- Roulin, A. (2004). The evolution, maintenance and adaptive function of genetic colour polymorphism in birds. *Biological Reviews* 79:815–848.
- Ruthrauff, D. R., A. Dekinga, R. E. Gill, J. A. van Gils, and T. Piersma (2015). Ways to be different: foraging adaptations that facilitate higher intake rates in a northerly-wintering shorebird compared to a low-latitude conspecific. *Journal of Experimental Biology* 218:1188–1197.
- Sánchez, M. I., F. Hortas, J. Figuerola, and A. J. Green (2012). Comparing the potential for dispersal via waterbirds of a native and an invasive brine shrimp. *Freshwater Biology* 57:1896–1903.

- Sargeant, B. L., A. J. Wirsing, M. R. Heithaus, and J. Mann (2007). Can environmental heterogeneity explain individual foraging variation in wild bottlenose dolphins (*Tursiops* sp.)? *Behavioral Ecology and Sociobiology* 61:679–688.
- Schindelin, J., I. Arganda-Carreras, E. Frise, V. Kaynig, M. Longair, T. Pietzsch, S. Preibisch, C. Rueden, S. Saalfeld, B. Schmid, J.-Y. Tinevez, et al. (2012). Fiji: an open-source platform for biological-image analysis. *Nature Methods* 9:676–682.
- Schmidt, A., M. Coll, T. Romanuk, and H. Lotze (2011). Ecosystem structure and services in eelgrass *Zostera marina* and rockweed *Ascophyllum nodosum* habitats. *Marine Ecology Progress Series* 437:51–68.
- Schwemmer, P., F. Güpner, S. Adler, K. Klingbeil, and S. Garthe (2016). Modelling small-scale foraging habitat use in breeding Eurasian oystercatchers (*Haematopus ostralegus*) in relation to prey distribution and environmental predictors. *Ecological Modelling* 320:322–333.
- Scrosati, R., and C. Heaven (2007). Spatial trends in community richness, diversity, and evenness across rocky intertidal environmental stress gradients in eastern Canada. *Marine Ecology Progress Series* 342:1–14.
- Seeley, R. H., and W. H. Schlesinger (2012). Sustainable seaweed cutting? The rockweed (*Ascophyllum nodosum*) industry of Maine and the Maritime Provinces: Sustainable seaweed cutting? *Annals of the New York Academy of Sciences* 1249:84–103.
- Skúlason, S., and T. B. Smith (1995). Resource polymorphisms in vertebrates. *Trends in Ecology & Evolution* 10:366–370.
- Small, C., and R. J. Nicholls (2003). A Global Analysis of Human Settlement in Coastal Zones. *Journal of Coastal Research* 19:584–599.
- Smith, P. C., and J. S. Bleakney (1969). Observations on oil pollution and wintering Purple Sandpipers, *Erolia Maritima* (Brunnich), in Nova Scotia. *Canadian Field Naturalist* 83:19–22.
- Soininen, E. M., L. Zinger, L. Gielly, E. Bellemain, K. A. Bråthen, C. Brochmann, L. S. Epp, G. Gussarova, K. Hassel, J.-A. Henden, S. T. Killengreen, et al. (2013). Shedding new light on the diet of Norwegian lemmings: DNA metabarcoding of stomach content. *Polar Biology* 36:1069–1076.
- Solbreck, C. (1978). Migration, diapause, and direct development as alternative life histories in a seed bug, *Neacoryphus bicruc*. In *The evolution of insect migration and diapause* (H. Dingle, Editor). Springer, New York, pp. 195–217.
- Southward, A. J. (1958). The Zonation of Plants and Animals on Rocky Sea Shores. *Biological Reviews* 33:137–177.

- Spiller, K. J., and R. Dettmers (2019). Evidence for multiple drivers of aerial insectivore declines in North America. *The Condor: Ornithological Applications* 121:1–13.
- Sprules, W. G. (1972). Effects of Size-Selective Predation and Food Competition on High Altitude Zooplankton Communities. *Ecology* 53:375–386.
- Stantial, M. L., and J. B. Cohen (2020). Miniature GPS Tags Provide High Resolution Location Data for a Small-Bodied Threatened Shorebird. *Waterbirds* 43.
- Stengel, D., and M. Dring (1997). Morphology and in situ growth rates of plants of *Ascophyllum nodosum* (Phaeophyta) from different shore levels and responses of plants to vertical transplantation. *European Journal of Phycology* 32:193–202.
- Stephenson, T. A., and A. Stephenson (1949). The Universal Features of Zonation Between Tide-Marks on Rocky Coasts. *The Journal of Ecology* 37:289–305.
- Stewart-Oaten, A., W. W. Murdoch, and K. R. Parker (1986). Environmental Impact Assessment: “Pseudoreplication” in Time? *Ecology* 67:929–940.
- Strann, K.-B., and R. W. Summers (1990). Diet and diurnal activity of Purple Sandpipers *Calidris maritima* wintering in northern Norway. *Fauna Norvegica Serie C, Cinclus* 13:75–78.
- Subramaniam, B., and M. D. Rausher (2000). Balancing Selection on a Floral Polymorphism. *Evolution* 54:691–695.
- Summers, R. W., T. Piersma, K.-B. Strann, and P. Wiersma (1998). How do Purple Sandpipers *Calidris maritima* survive the winter north of the Arctic circle? *Ardea* 86:51–58.
- Summers, R. W., S. Smith, M. Nicoll, and N. K. Atkinson (1990a). Tidal and sexual differences in the diet of Purple Sandpipers *Calidris maritima* in Scotland. *Bird Study* 37:187–194.
- Summers, R. W., K.-B. Strann, R. Rae, and J. Heggas (1990b). Wintering Purple Sandpipers *Calidris maritima* in Troms County, Northern Norway. *Ornis Scandinavica* 21:248–254.
- Summers, R. W., L. G. Underhill, M. Nicoll, R. Rae, and T. Piersma (1992). Seasonal, size- and age-related patterns in body-mass and composition of Purple Sandpipers *Calidris maritima* in Britain. *Ibis* 134:346–354.
- Sundar, K. S. G. (2006). Flock Size, Density and Habitat Selection of Four Large Waterbirds Species in an Agricultural Landscape in Uttar Pradesh, India: Implications for Management. *Waterbirds* 29:365–374.
- Sungail, J., A. C. Brown, K. Alpert, and J. Maurukas (2013). Prey selection by Gulf of Maine green crabs (*Carcinus maenas*), rock crabs (*Cancer irroratus*) and American lobsters (*Homarus americanus*): A laboratory study. *Journal of Experimental Marine Biology and Ecology* 449:294–303.

- Suter, W., and M. R. van Eerden (1992). Simultaneous Mass Starvation of Wintering Diving Ducks in Switzerland and the Netherlands: a Wrong Decision in the Right Strategy? *Ardea* 80:229–242.
- Sutherland, B. (2005). An Independent Study and Review of the New Brunswick Rockweed Harvest – Phase 2. Eastern Charlotte Waterways Inc., Black Harbour, New Brunswick.
- Swanson, F. J., S. L. Johnson, S. V. Gregory, and S. A. Acker (1998). Flood Disturbance in a Forested Mountain Landscape. *BioScience* 48:681–689.
- Swift, R. J., A. D. Rodewald, J. A. Johnson, B. A. Andres, and N. R. Senner (2020). Seasonal survival and reversible state effects in a long-distance migratory shorebird. *Journal of Animal Ecology* 89:2043–2055.
- Thomas, D. L., and E. J. Taylor (2006). Study Designs and Tests for Comparing Resource Use and Availability II. *Journal of Wildlife Management* 70:324–336.
- Tibbe, T. D., and A. K. Montoya (2022). Correcting the Bias Correction for the Bootstrap Confidence Interval in Mediation Analysis. *Frontiers in Psychology* 13:810258.
- Trott, T. J., and P. F. Larsen (2012). Evaluation of short-term changes in rockweed (*Ascophyllum nodosum*) and associated epifaunal communities following cutter rake harvesting in Maine. Maine Department of Marine Resources.
- Tuttle, E. M., A. O. Bergland, M. L. Korody, M. S. Brewer, D. J. Newhouse, P. Minx, M. Stager, A. Betuel, Z. A. Cheviron, W. C. Warren, R. A. Gonser, and C. N. Balakrishnan (2016). Divergence and Functional Degradation of a Sex Chromosome-like Supergene. *Current Biology* 26:344–350.
- Tverin, M., R. Esparza-Salas, A. Strömberg, P. Tang, I. Kokkonen, A. Herrero, K. Kauhala, O. Karlsson, R. Tiilikainen, M. Vetemaa, T. Sinisalo, et al. (2019). Complementary methods assessing short and long-term prey of a marine top predator – Application to the grey seal-fishery conflict in the Baltic Sea. *PLOS ONE* 14:e0208694.
- Ugarte, R. A., G. Sharp, and B. Moore (2006). Changes in the Brown Seaweed *Ascophyllum nodosum* (L.) Le Jol. Plant Morphology and Biomass Produced by Cutter Rake Harvests in Southern New Brunswick, Canada. *Journal of Applied Phycology* 18:351–359.
- Ugarte, R., and G. Sharp (2012). Management and production of the brown algae *Ascophyllum nodosum* in the Canadian maritimes. *Journal of Applied Phycology* 24:409–416.
- USDA Natural Resources Conservation Service (2022). SNOwpack TELEmetry Network (SNOTEL). NRCS. <https://data.nal.usda.gov/dataset/snowpack-telemetry-network-snotel>. Accessed 2023-03-16.
- Vadas, R. L., and W. A. Wright (1986). Recruitment, growth and management of *Ascophyllum nodosum*. *Actas II Congreso Algas Marinas Chilenas*: 101-113.

- Vadas, R. L., W. A. Wright, and B. F. Beal (2004). Biomass and Productivity of Intertidal Rockweeds (*Ascophyllum nodosum* LeJolis) in Cobscook Bay. *Northeastern Naturalist* 11:123–142.
- Van Gils, J. A., S. R. D. Rooij, J. Van Belle, J. Van Der Meer, A. Dekinga, T. Piersma, and R. Drent (2005). Digestive bottleneck affects foraging decisions in red knots *Calidris canutus*. I. Prey choice: *Prey choice under a digestive constraint*. *Journal of Animal Ecology* 74:105–119.
- Vea, J., and E. Ask (2011). Creating a sustainable commercial harvest of *Laminaria hyperborea*, in Norway. *Journal of Applied Phycology* 23:489–494.
- Walder, C. E. (2015). Making the Cut: Benthic Community Responses to Rockweed (*Ascophyllum nodosum*) Harvesting.
- Wanink, J., and L. Zwarts (1985). Does an optimally foraging oystercatcher obey the functional response? *Oecologia* 67:98–106.
- Watts, B. D., and C. Turrin (2016). Assessing hunting policies for migratory shorebirds throughout the Western Hemisphere. *Wader Study* 123:6–15.
- Welch, W. R. (1968). Changes in abundance of the green crab, *Carcinus maenas* (L.), in relation to recent temperature changes. *Fishery Bulletin* 67:337–345.
- West-Eberhard, M. J. (1989). Phenotypic Plasticity and the Origins of Diversity. *Annual Reviews* 20:249–278.
- Whiteman, H. H. (1997). Maintenance of Polymorphism Promoted by Sex-Specific Fitness Payoffs. *Evolution* 51:2039–2044.
- Whiteman, H. H., J. J. Gutrich, and R. S. Moorman (1999). Courtship Behavior in a Polymorphic Population of the Tiger Salamander, *Ambystoma tigrinum nebulosum*. *Journal of Herpetology* 33:348–351.
- Whiteman, H. H., S. A. Wissinger, and A. J. Bohonak (1994). Seasonal movement patterns in a subalpine population of the tiger salamander, *Ambystoma tigrinum nebulosum*. *Canadian Journal of Zoology* 72:1780–1787.
- Whiteman, H. H., S. A. Wissinger, and W. S. Brown (1996). Growth and foraging consequences of facultative paedomorphosis in the tiger salamander, *Ambystoma tigrinum nebulosum*. *Evolutionary Ecology* 10:433–446.
- Whiteman, H. H., S. A. Wissinger, M. Denoël, C. J. Mecklin, N. M. Gerlanc, and J. J. Gutrich (2012). Larval growth in polyphenic salamanders: making the best of a bad lot. *Oecologia* 168:109–118.

- Wikström, S. A., and L. Kautsky (2007). Structure and diversity of invertebrate communities in the presence and absence of canopy-forming *Fucus vesiculosus* in the Baltic Sea. *Estuarine, Coastal and Shelf Science* 72:168–176.
- Wilzbach, M. A., B. C. Harvey, J. L. White, and R. J. Nakamoto (2005). Effects of riparian canopy opening and salmon carcass addition on the abundance and growth of resident salmonids. *Canadian Journal of Fisheries and Aquatic Sciences* 62:58–67.
- Wissinger, S. A., A. J. Bohonak, H. H. Whiteman, and W. S. Brown (1999). Habitat permanence, salamander predation, and invertebrate communities. In *Invertebrates in Freshwater Wetlands of North America: Ecology and Management* (D. P. Batzer, R. B. Rader and S. A. Wissinger, Editors). Wiley, New York, pp. 757–789.
- Wissinger, S. A., W. S. Brown, and J. E. Jannot (2003). Caddisfly life histories along permanence gradients in high-altitude wetlands in Colorado (U.S.A.): Caddisfly life histories in wetlands. *Freshwater Biology* 48:255–270.
- Wollrab, S., A. M. de Roos, and S. Diehl (2013). Ontogenetic diet shifts promote predator-mediated coexistence. *Ecology* 94:2886–2897.
- Wootton, J. T. (2012). River Food Web Response to Large-Scale Riparian Zone Manipulations. *PLoS ONE* 7:e51839.
- Zera, A. J. (1984). Differences in Survivorship, Development Rate and Fertility Between the Longwinged and Wingless Morphs of the Waterstrider, *Limnopus canaliculatus*. *Evolution* 38:1023–1032.
- Zera, A. J., and R. F. Denno (1997). Physiology and Ecology of Dispersal Polymorphism in Insects. *Annual Review of Entomology* 42:207–230.
- Zera, A. J., and S. Mole (1994). The physiological costs of flight capability in wing-dimorphic crickets. *Population Ecology* 36:151–156.

APPENDIX A: Chapter 1 Supplemental Material

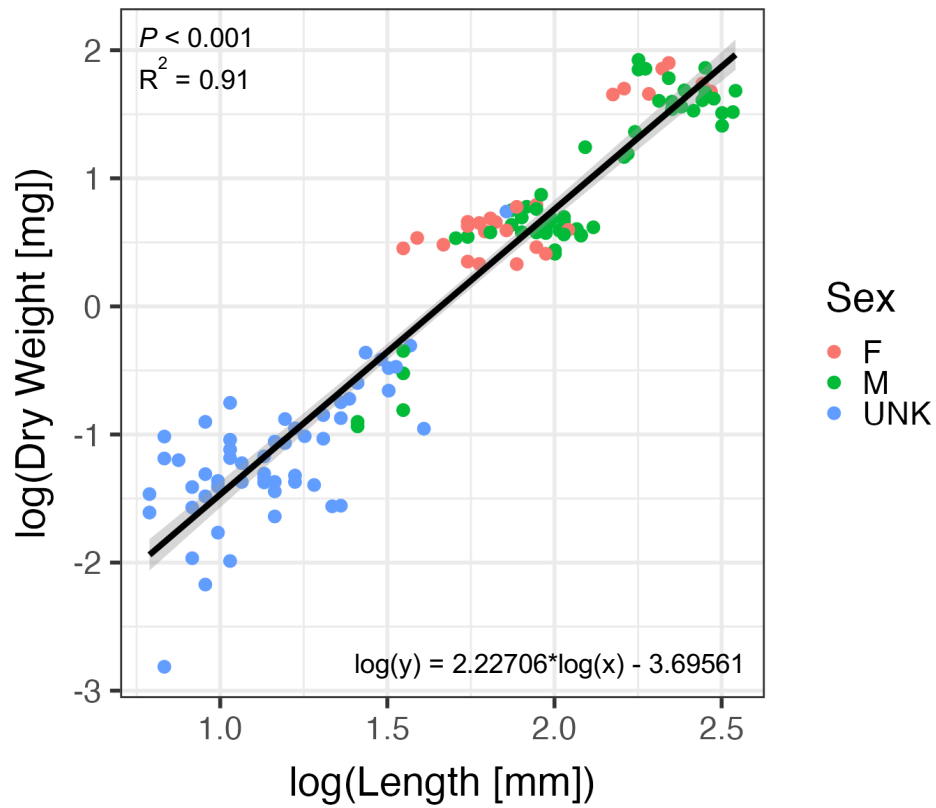


Figure A.1. Length-weight regression for 133 Colorado fairy shrimp (*Branchinecta coloradensis*) from nonpermanent ponds at Mexican Cut Nature Preserve. The shaded gray area around the regression line represents the 95% confidence interval.

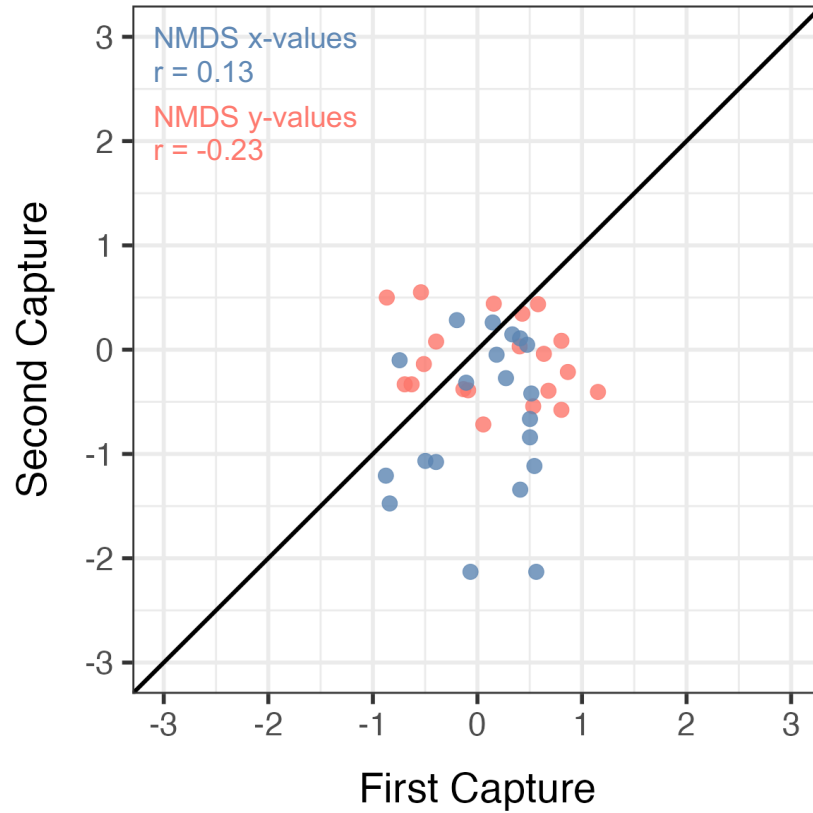


Figure A.2. Correlation among metamorphs' diet composition between the first and second captures. We collected multiple gut content samples for 20 out of 73 metamorphs. NMDS coordinates represent the prey compositions of stomach samples in two-dimensional space. The diagonal black line represents $y = x$ (i.e., correlation coefficient = 1).

APPENDIX B: Chapter 2 Supplemental Material

A



B

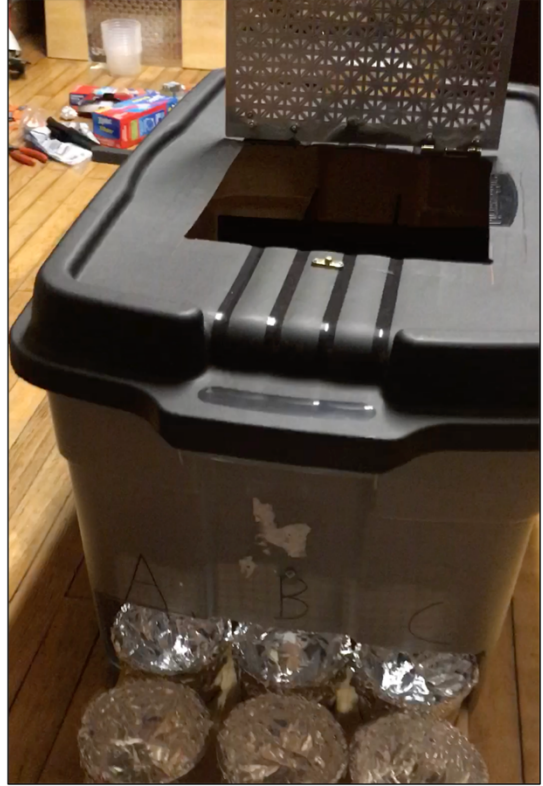


Figure B.1. Fecal sampling box for positive field controls ($N = 3$). After capture, Purple Sandpipers were placed in cells (A) for a maximum of 30 minutes. Individual birds sat on the hardware cloth floor and defecated into the tinfoil-lined containers below. The top covers of cells are not pictured. Samples were removed by sliding the bottom panel through the front opening (B).

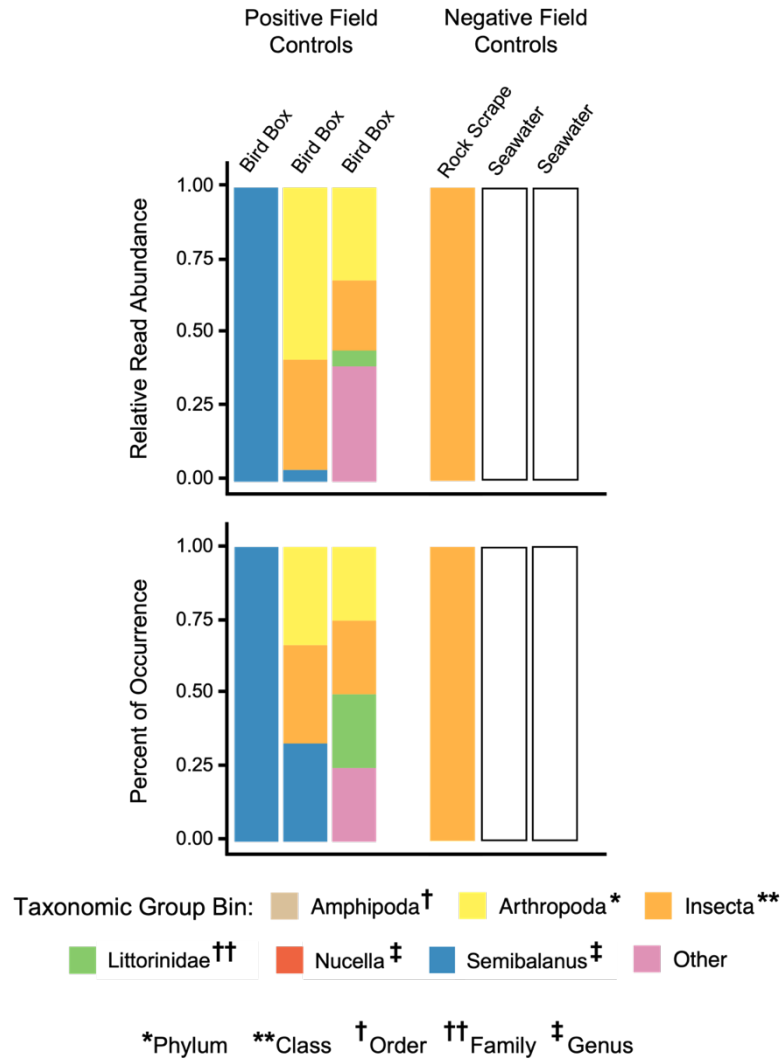


Figure B.2. Taxonomic group reads from fecal metabarcoding sequencing for positive and negative field controls. Positive field controls are Purple Sandpiper fecal samples collected in the fecal sampling box (Figure 2.1). Negative field controls are rock scrape and seawater samples. Each column represents an individual fecal sample. Taxonomic groups are grouped into seven bins in an effort to aid visual clarity. The top panel visualizes the sequencing data as relative read abundances (RRA), which are proportional summaries of read counts. The bottom panel shows percent of occurrence (POO), which displays equally-weighted presence-absence of taxonomic group bins. Both seawater samples did not have total read counts above 10,013 reads, and therefore they were removed from the analysis after rarefying samples.

APPENDIX C: Chapter 3 Supplemental Material

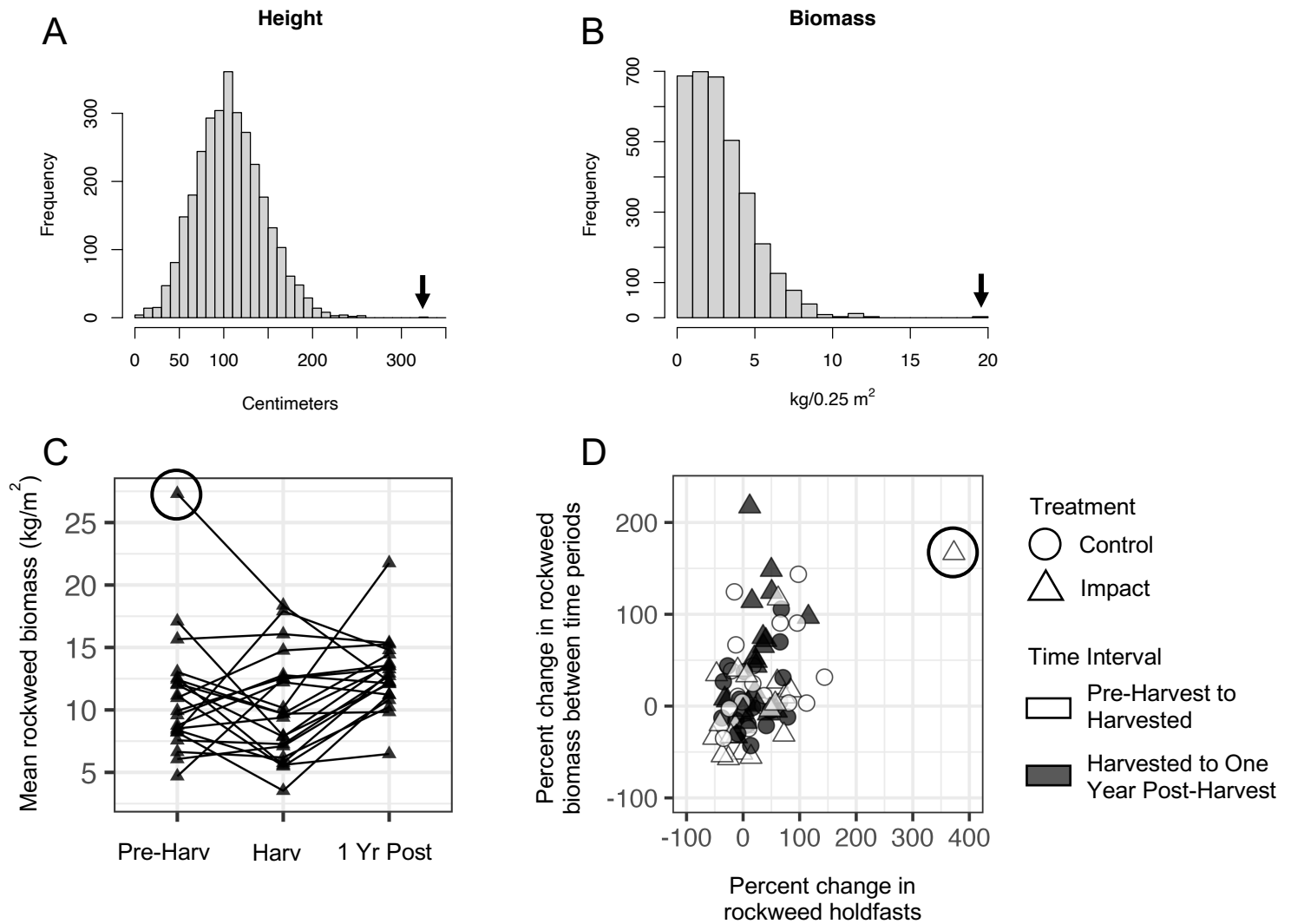


Figure C.1 Outliers removed from final analyses. Outliers in panels A and B are single height (alga) and biomass (quadrat) values, respectively, while outliers in panels C and D represent one impact site each. The legend applies to panel D only.

Table C.1. Results of linear mixed models comparing pre-harvest rockweed height and biomass across our study regions. The significance levels ($P \leq 0.05$ in bold) are calculated from F -statistics of type III hypothesis tests, based on the Kenward–Roger approximations for degrees of freedom. Marginal R^2 considers only the variance of the fixed effects, while conditional R^2 considers the variance of both fixed and random effects. Quadrat is included as a source of variation for only the height model given that we measured three rockweed individuals in each quadrat but only recorded one biomass value per quadrat.

Rockweed characteristic	Source of variation	Variance	SD	df	F	P
Height	<i>Fixed effect</i>					
	Region			4, 32.89	1.45	0.24
	<i>Random effect</i>					
	Region(Site)	195.21	13.97			
	Region(Site(Transect))	158.67	12.60			
	Region(Site(Transect(Quadrat)))	64.78	8.05			
	Residual	1013.55	31.84			
Marginal R^2 : 0.03; Conditional R^2 : 0.32						
Biomass	<i>Fixed effect</i>					
	Region			4, 33	1.26	0.31
	<i>Random effect</i>					
	Region(Site)	0.008	0.09			
	Region(Site(Transect))	0.07	0.26			
	Residual	0.24	0.49			
Marginal R^2 : 0.02; Conditional R^2 : 0.25						

Table C.2. Planned one degree of freedom contrasts assessing the effect of treatment on (top to bottom) harvest impacts, post-harvest growth, and one-year recovery, respectively, for rockweed height and biomass in each study region. Contrasts were derived from linear mixed models with a three-way interaction between treatment, time period, and region as fixed effects and site as a random effect. Statistically significant results ($\alpha = 0.05$) are in bold. $\bar{X}_{i,j}$ represents the mean height or biomass value during the i th time period (Pre = Pre-harvest, Harv = Harvested, Yr Post = One Year Post-harvest) at sites with j treatment type (I = Impact, C = Control).

Region	Planned contrast	Estimate	SE	df	t	P
Height						
Midcoast	$(\bar{X}_{\text{Harv, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Harv, C}} - \bar{X}_{\text{Pre, C}})$	-28.33	12.60	1, 60	-2.26	0.03
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Harv, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Harv, C}})$	-11.80	12.60	1, 60	-0.94	0.35
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Pre, C}})$	-40.14	12.60	1, 60	-3.20	0.002
Penobscot Bay	$(\bar{X}_{\text{Harv, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Harv, C}} - \bar{X}_{\text{Pre, C}})$	-9.39	12.60	1, 60	-0.75	0.46
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Harv, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Harv, C}})$	-3.01	12.60	1, 60	-0.24	0.81
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Pre, C}})$	-12.40	12.60	1, 60	-0.99	0.33
Jonesport	$(\bar{X}_{\text{Harv, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Harv, C}} - \bar{X}_{\text{Pre, C}})$	13.10	14.90	1, 60	0.88	0.38
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Harv, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Harv, C}})$	1.50	14.90	1, 60	0.10	0.92
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Pre, C}})$	14.60	14.90	1, 60	0.98	0.33
Cobscook Bay	$(\bar{X}_{\text{Harv, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Harv, C}} - \bar{X}_{\text{Pre, C}})$	-12.25	12.20	1, 60	-1.01	0.32
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Harv, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Harv, C}})$	9.13	12.20	1, 60	0.75	0.46
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Pre, C}})$	-3.12	12.20	1, 60	-0.26	0.80
Biomass						
Midcoast	$(\bar{X}_{\text{Harv, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Harv, C}} - \bar{X}_{\text{Pre, C}})$	-8.07	2.66	1, 60	-3.03	0.004
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Harv, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Harv, C}})$	2.97	2.66	1, 60	1.12	0.27
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Pre, C}})$	-5.10	2.66	1, 60	-1.92	0.06
Penobscot Bay	$(\bar{X}_{\text{Harv, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Harv, C}} - \bar{X}_{\text{Pre, C}})$	-2.07	2.66	1, 60	-0.78	0.44
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Harv, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Harv, C}})$	3.12	2.66	1, 60	1.17	0.25
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Pre, C}})$	1.05	2.66	1, 60	0.40	0.69

Jonesport	$(\bar{X}_{\text{Harv, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Harv, C}} - \bar{X}_{\text{Pre, C}})$	0.94	3.15	1, 60	0.30	0.77
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Harv, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Harv, C}})$	1.47	3.15	1, 60	0.47	0.64
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Pre, C}})$	2.42	3.15	1, 60	0.77	0.45
Cobscook Bay	$(\bar{X}_{\text{Harv, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Harv, C}} - \bar{X}_{\text{Pre, C}})$	-1.68	2.59	1, 60	-0.65	0.52
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Harv, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Harv, C}})$	3.89	2.59	1, 60	1.50	0.14
	$(\bar{X}_{\text{Yr Post, I}} - \bar{X}_{\text{Pre, I}}) \neq (\bar{X}_{\text{Yr Post, C}} - \bar{X}_{\text{Pre, C}})$	2.21	2.59	1, 60	0.85	0.40

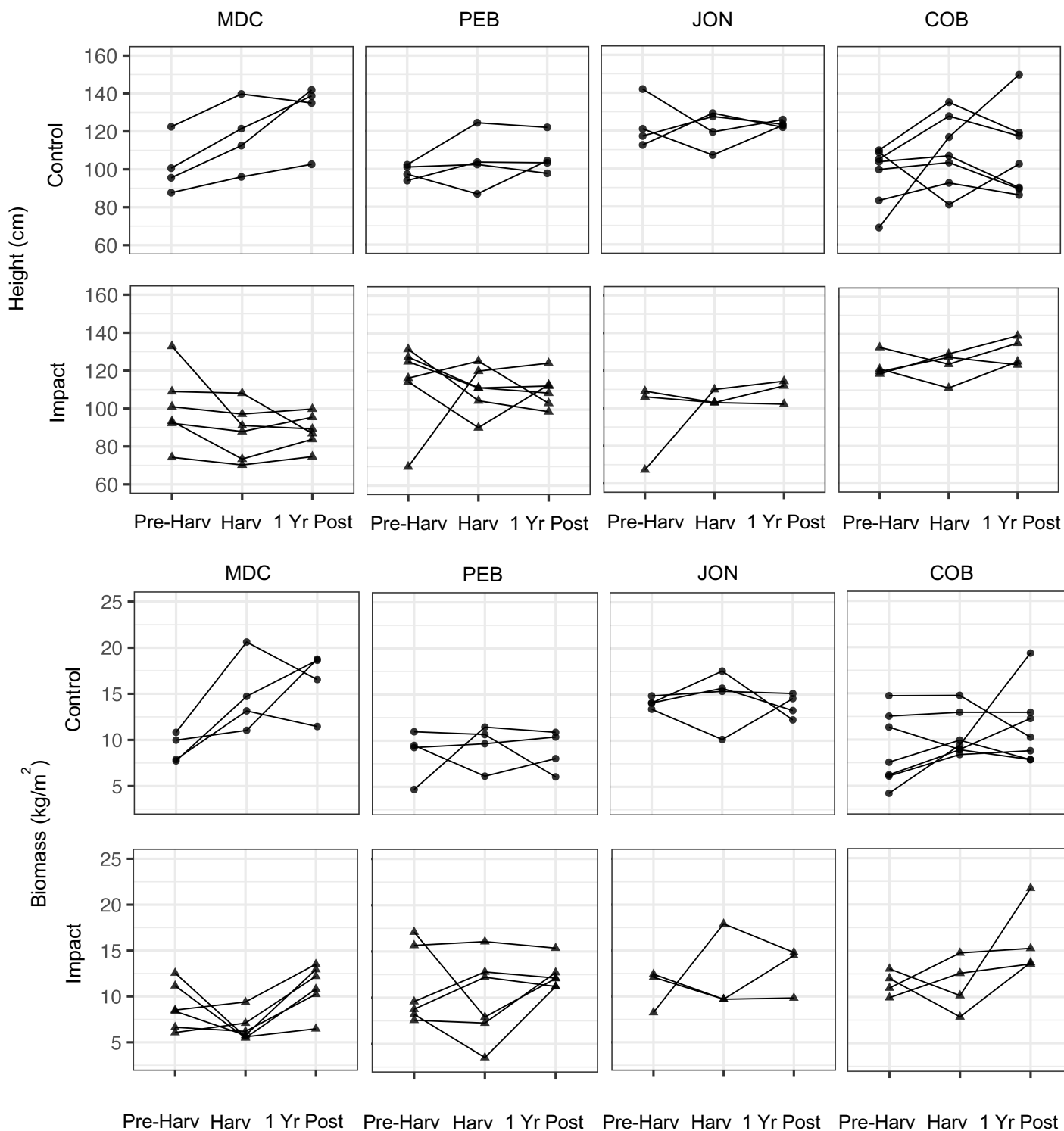


Figure C.2. Change in mean rockweed height and biomass across three time periods in each study region. Each point represents a mean site value, and a line connects a given site through time. Error bars for each point are not shown to aid visual clarity. Abbreviations are as follows: Pre-Harv = Pre-harvest, Harv = Harvested, 1 Yr Post = One Year Post-harvest, MDC = Midcoast, PEB = Penobscot, JON = Jonesport, and COB = Cobscook

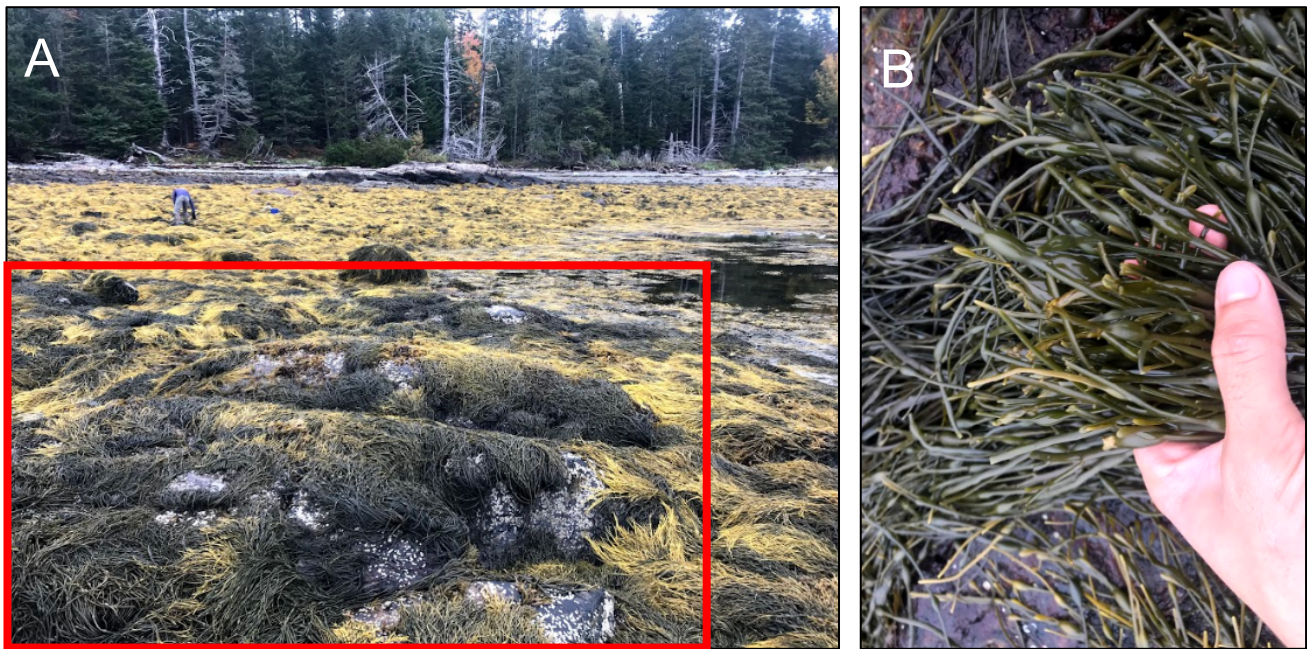


Figure C.3. Photographs of an impact site immediately following harvest. The red box in panel A indicates where the majority of harvest occurred at this site. Panel B provides a close-up view of a harvested rockweed individual.

APPENDIX D: Chapter 4 Supplemental Material

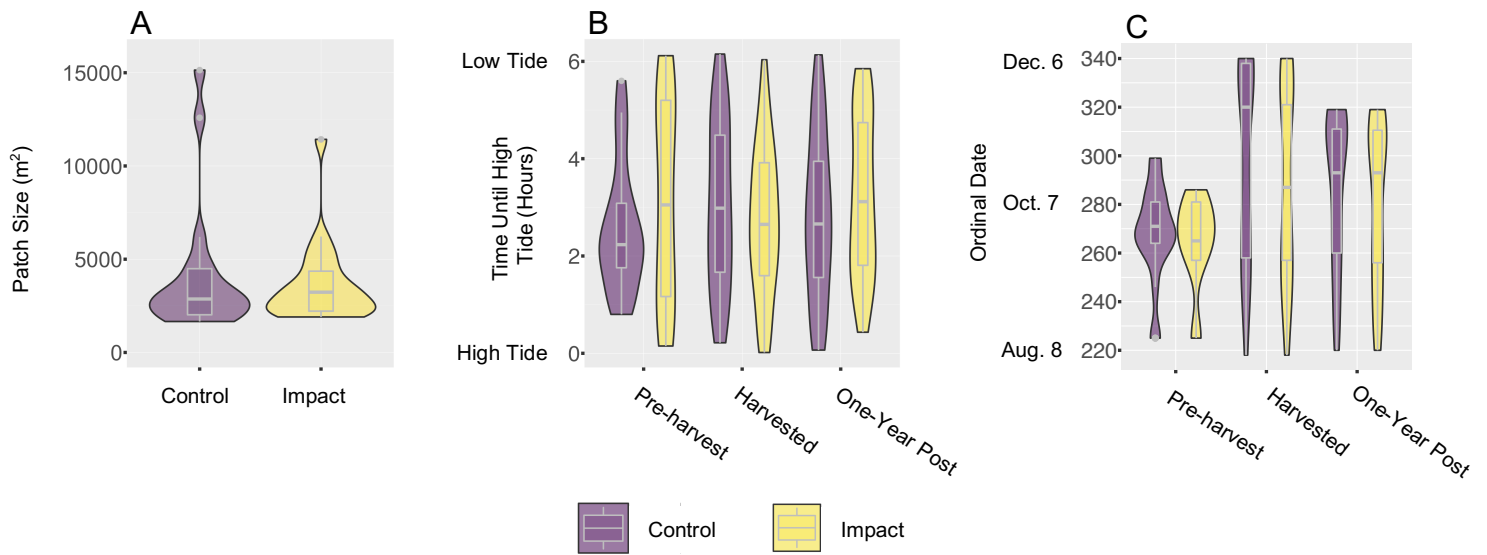
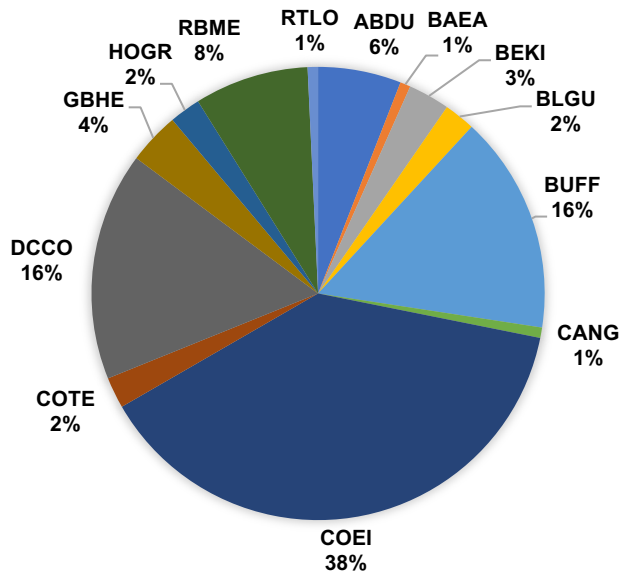
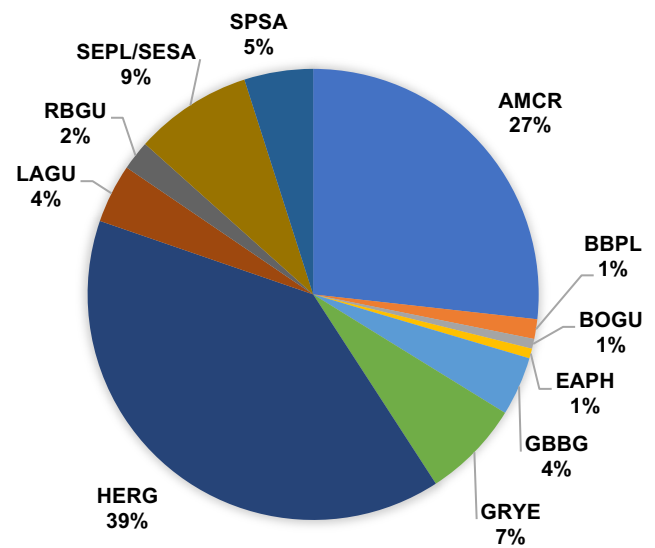


Figure D.1. (A) Site- and (B, C) survey-level covariates. For patch size, $P = 0.21$ for an effect of treatment; for survey tidal height, $P = 0.24$ for an effect of treatment $\times$ time period; and for ordinal date $P = 0.95$ for an effect of treatment $\times$ time period.



High Tide Assemblage



Low Tide Assemblage

Figure D.2. Species compositions of high- (N = 135 birds) and low-tide (N = 147 birds) intertidal bird assemblages. Outlier flocks that were removed from analyses are not included here. Counts of SEPL and SESA were combined due to several counts of mixed-species flocks. Abbreviations are as follows: ABDU = American Black Duck, AMCR = American Crow, BAEA = Bald Eagle, BBPL = Black-bellied Plover, BEKI = Belted Kingfisher, BOGU = Bonaparte's Gull, BLGU = Black Guillemot, BUFF = Bufflehead, CANG = Canada Goose, COEI = Common Eider, COTE = Common Tern, DCCO = Double-crested Cormorant, EAPH = Eastern Phoebe, GBBG = Great Black-backed Gull, GBHE = Great Blue Heron, GRYE = Greater Yellowlegs, HERG = Herring Gull, HOCR = Horned Grebe, LAGU = Laughing Gull, RBGU = Ring-billed Gull, RBME = Red-breasted Merganser, RTLO = Red-throated Loon, SEPL = Semipalmated Plover, SESA = Semipalmated Sandpiper, SPSA = Spotted Sandpiper.

BIOGRAPHY OF THE AUTHOR

Elliot Johnston was born on July 9, 1991 and raised in Buffalo, New York. He graduated from Nichols School in 2010 and earned a Bachelor of Arts in International Relations from the University of Pennsylvania in 2014. He began a Master's degree in Ecology and Environmental Science at the University of Maine in 2018 and switched his degree track to a Ph.D. in 2020. During his graduate studies, Elliot was honored to receive the Chase Distinguished Research Assistantship during the 2022-23 academic year as well as the 2022-23 College of Natural Sciences, Forestry, and Agriculture's George F. Dow Award. Elliot is a candidate for the Doctor of Philosophy degree in Ecology and Environmental Sciences from the University of Maine in May 2023.