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Herbivore Impacts on Two Morphologically Similar Bloom-Forming Ulva Species in a Eutrophic Bay

Herbivore impacts on two morphologically similar bloom-forming *Ulva* species in a eutrophic bay

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Abstract

Herbivore impacts on macrophyte growth vary with the identity of the herbivores and macrophytes, as well as under different abiotic conditions. This interaction is further complicated by anthropogenic alterations to the environment, such as eutrophication. In this study, we utilized *in situ* herbivore exclusion experiments and mesocosm feeding preference assays to examine the impacts of different herbivores on the growth of two morphologically similar, co-occurring macroalgal bloom *Ulva* species in a nutrient-rich environment. We found herbivory had a measurable impact on *Ulva* biomass, though the rate of consumption rarely surpassed growth for either *Ulva* species. We determined that the primary herbivores within the blooms were amphipods and mud crabs, and that their effects varied among study sites and months. Our results also confirmed that, even with a diverse suite of consumers, *Ulva* blooms are capable of escaping herbivore control, particularly early in the growing season when growth rates peak and herbivore activity is limited. Furthermore, our experiments revealed species-specific feeding preferences among herbivores, as well as differences in growth rates and chemistry between the two *Ulva* species, which likely influence bloom dynamics.

Keywords: Algal bloom, Eutrophication, Green tide, Herbivore, Top down control, *Ulva*

Introduction

The structure of macrophyte communities is affected by the availability of resources and the strength of herbivory; the relative contribution of these opposing forces can fluctuate considerably among different habitats (Shurin et al., 2002; Hillebrand et al., 2007; Gruner et al., 2008). Even among similar habitat types, the relationship between resource availability and herbivore pressure can vary depending upon the identity, diversity, and abundance of individual macrophyte and herbivore species present (e.g. Boyer et al., 2004; Burkepile & Hay, 2006; Sala et al., 2008; Vermeij et al., 2010; McLenaghan et al., 2011).

Anthropogenic impacts, including nutrient pollution, can significantly alter the relative influence of resource availability and herbivory on macroalgal communities. Within coastal ecosystems, eutrophication can promote the growth of fast growing ephemeral macroalgal species at the expense of perennial macroalgae and seagrasses (Valiela et al., 1997; Hauxwell et al., 2001; Worm & Lotze, 2006). In some instances, herbivory offsets this increase in biomass, preventing widespread changes to the ecosystem (Geertz-Hansen et al., 1993; Neckles et al., 1993; Williams & Ruckelshaus, 1993; Korpinen et al., 2007). However, in many cases nutrient enrichment enables macroalgal growth to surpass herbivore control (Horne et al., 1994; Hauxwell et al., 1998; Morgan et al., 2003; Worm & Lotze, 2006; Fox et al., 2012), resulting in macroalgal blooms. Moreover, persistent eutrophication can further lessen herbivore control by decreasing herbivore functional responses to the higher algal nutrient concentrations that occur with nutrient pulses (Russell & Connell, 2007) and increasing the occurrence of hypoxic events that result in herbivore mortality (Hauxwell et al., 1998; Berezina et al., 2007).

Beyond their potential to limit the occurrence or severity of a macroalgal bloom event, herbivores also have the potential to alter algal diversity or richness within a bloom by

58 preferentially consuming one or more algal species. This preference can be driven by algal
59 morphology, chemical defenses, or nutritional content (e.g. Van Alstyne et al., 2001; Van
60 Alstyne & Houser, 2003; Thornber et al., 2008), and results in an increase of less palatable
61 species. For example, Lotze & Worm (2000) observed that herbivores in the Baltic Sea preferred
62 to graze *Ulva* spp. (formerly *Enteromorpha*) over *Pilayella littoralis*, resulting in the dominance
63 of *P. littoralis* in Baltic blooms. However, mesocosm assays demonstrated that this preference
64 was only exhibited by certain herbivore species. Similarly, Nelson et al. (2008) found that the
65 distributional pattern of attached bloom-forming *Ulva* and *Ulvaria* in Washington, USA was due
66 to a combination of preferences by subtidal herbivores for *Ulva* and abiotic conditions in the
67 intertidal zone that restricted the range of chemically defended *Ulvaria*. Consequently, *Ulva* was
68 more abundant in the intertidal zone, while *Ulvaria* proliferated in the subtidal zone. These
69 studies highlight both the importance of herbivory in determining the macroalgal species present
70 within blooms, as well as how herbivore impacts can vary with herbivore identity and
71 environmental conditions.

72 Macroalgal blooms are an annual occurrence within many shallow, eutrophic areas of
73 Narragansett Bay, Rhode Island. In contrast to previously studied multi-species blooms, which
74 contained species from different genera that were distinctly different in morphology and/or
75 chemistry (eg. Lotze & Worm 2000; Nelson et al. 2008), these blooms are primarily composed
76 of the morphologically similar *Ulva compressa* Linnaeus and *U. rigida* C. Agardh. Which (if
77 either) of these *Ulva* species is dominant within these blooms varies spatially and temporally
78 (Guidone & Thornber, 2013), and it is currently unknown how abiotic and biotic factors
79 influence interactions between these species. Surveys of bloom sites within Narragansett Bay
80 indicated differences in the invertebrate community present within the blooms (Guidone and

Thornber, unpubl. data), offering a potential explanation for site-to-site differences in bloom species composition. Therefore, to explore herbivore impacts within these blooms, we conducted *in situ* herbivore exclusion experiments as well as a series of mesocosm feeding assays to determine: 1) whether herbivores that co-occur with *Ulva* blooms in the field have a measurable impact on *Ulva* biomass, 2) how the impacts of invertebrate grazing vary among species, sites of varying bloom severity, and throughout the growing season, 3) the consumption rates and feeding preferences of several abundant herbivore species on *U. compressa* and *U. rigida*, and 4) any physical or chemical differences between the two *Ulva* species that might explain herbivore preferences. We discuss our results within the context of the role of invertebrates in impacting algal bloom severity and species composition.

Materials and Methods

Study locale and species

Narragansett Bay, Rhode Island, is a well-studied estuary; the northern portions of the bay, as well as several of the bay's subestuaries, are heavily impacted by anthropogenic nutrient and chemical pollution (e.g. Granger et al., 2000; Calabretta & Oviatt, 2008; Deacutis, 2008; Oczkowski et al., 2008). Our field sites were located within Greenwich Bay, Rhode Island (Fig. 1), a subestuary of Narragansett Bay that experiences annual blooms dominated by *U. compressa* and *U. rigida* (Guidone & Thornber, 2013). Both *U. compressa* and *U. rigida* are distromatic blades lacking any distinguishing macroscopic features. Therefore, prior to the start of each experiment species identity of all blades was determined in the laboratory using cellular features based on previous molecular studies (Hofmann et al., 2010; Guidone et al., 2013).

All invertebrates used in our mesocosm feeding assays were collected from Oakland Beach Cove, Warwick, Rhode Island (Fig. 1). The species used in the mesocosm experiments were selected based upon their presence during our pilot *in situ* herbivore exclusion cage experiments (see below). Although abundant at all of our sites, mud snails (*Ilyanassa obsoleta*) were not included in these assays as our data indicate that, in this system, they rarely consume distromatic *Ulva* spp. (Guidone et al., 2010, 2012).

In situ herbivore exclusions

We conducted *in situ* herbivore exclusion experiments monthly from May-August 2009 and June-July 2010 at three field sites: Chepiwanoxet (CH), Warwick City Park (WCP), and Oakland Beach Cove (OBC; Fig. 1). These sites were selected to represent a range of distromatic *Ulva* bloom severity (low at CH, medium at WCP, and high at OBC) based on patterns of *Ulva* wrack accumulation (Guidone and Thornber, personal observation). WCP and OBC were located nearer to one another than either was to CH, with WCP and OBC approximately 250 m apart on opposite sides of a heavily impacted cove and CH in a separate portion of Greenwich Bay approximately 3,000 m away. Cages at CH and WCP were located at mean depths of 40-60 cm at low tide and were placed adjacent to areas where *Ulva* wrack accumulates on shore. Cages at OBC were at a mean depth of 20-30 cm at low tide; OBC cages were located in an area frequently inundated by drift *Ulva*. Herbivore exclusion cages were placed 30 m offshore of the mean low tide line, ensuring continual submersion throughout each experiment. Starting with the June 2009 experiment, water temperature was measured every half hour throughout each experiment at each site using temperature data loggers (Tidbit v2, Onset, Massachusetts, USA). Mean daily temperatures during 2009 and 2010 were analyzed for differences among sites and

126 months using two-way ANOVAs (JMP version 8, SAS Institute Inc., North Carolina, USA). To
127 estimate differences in dissolved inorganic nitrogen (DIN) availability among the sites, we
128 collected a single water sample from each site at the conclusion of each *in situ* experiment. DIN
129 levels were determined with a segmented flow autoanalyzer (model 303A, Astoria Pacific
130 International, Oregon, USA) by the University of Rhode Island's Watershed Watch.
131 In 2009, we examined herbivore impacts on *Ulva compressa.* For these experiments, we
132 placed a single pre-weighed blade of *U. compressa* within a mesh cage (12.7 cm x 8.3 cm x 6.4
133 cm). Prior to weighing, all blades were spun to a consistent dryness in a salad spinner
134 (approximately 20 rotations). Blade wet masses ranged from 0.7-1.0 g. Cages were constructed
135 from a 1 cm plastic frame covered with one of three mesh sizes (n = 5 for each mesh size). Pilot
136 studies conducted during 2008 indicated that these mesh sizes differentially excluded
137 invertebrates, creating three different communities among the cages. The smallest mesh (1 mm^2
138 pore opening) excluded all invertebrates except amphipods, isopods, polychaetes, and juvenile
139 (<3 mm carapace width) mud crabs. The medium mesh (16 mm^2 pore size) excluded large mud
140 crabs, hermit crabs, and large mud snails, allowing in shrimp, small mud crabs (<14 mm
141 carapace width), and small mud snails. The largest mesh (100 mm^2 pore size) excluded only
142 large predatory crabs and fish. Cages were secured by cable tying them to a PVC stake that was
143 driven securely into the substrate.
144 Cages remained in the field for nine to ten days, after which they were retrieved with all
145 of their contents; previous studies have used similar time frames to measure herbivore impacts
146 (eg. Lotze & Worm 2000; Nelson et al. 2008). To retain all mobile invertebrates, we placed each
147 cage within a plastic gallon zipper bag while the cage was still submerged. *U. compressa* final
148 wet mass was measured and all invertebrates found within the cages were counted and identified

149 to the family or species (when possible) level. In addition, to determine if potential herbivores
150 avoided entering the cages, we sampled co-occurring invertebrate densities at each field site on
151 the final day of each experiment. Invertebrate density was sampled with a 40 cm diameter mesh
152 net (1 mm pore diameter) at 3 m intervals along a 30 m subtidal transect positioned
153 perpendicular to the shoreline. Additionally, we recorded the water depth of each sample.
154 During pilot studies, we determined that mesh sizes smaller than 1 mm^2 restricted water
155 flow and light penetration, resulting in limited *Ulva* growth. Consequently, we were unable to
156 include a non-herbivore growth control treatment at our field sites. Therefore, to ensure the
157 different cage mesh sizes did not differentially impact *Ulva* growth, we monitored growth within
158 five cages of each mesh type held in seawater tables at the University of Rhode Island's Bay
159 Campus (described below). These concurrent cage mesh controls were run during each
160 experiment in 2009 and 2010.
161 In 2010, we examined herbivore impacts on both *U. compressa* and *U. rigida*. Within
162 each herbivore exclusion cage, we placed a known wet mass of both species. To accommodate
163 the additional *Ulva* biomass, we constructed cages with slightly larger dimensions (10 x 10 x 10
164 cm). All other methods were the same as described above.
165 We analyzed the change in *U. compressa* biomass during the 2009 experiments using a
166 three-way ANOVA for differences among months, sites, and cage types. For our 2010
167 experiments, the change in *U. compressa* and *U. rigida* biomass was analyzed for differences
168 among species, months, sites, and cage types using a fully factorial nested ANOVA, with species
169 nested within cage type to account for variation among cages. All data were tested for normality
170 and homogeneity of variance and transformed to meet these assumptions as needed. In addition,
171 we assessed the differences in invertebrate assemblages between our exclusion cages and net

samples using a one-way analysis of similarity (ANOSIM) with subsequent similarity of percentages (SIMPER) analysis. Invertebrate composition within the cages during each month of the experiment was further analyzed for differences among the study sites and cage types using a two-way crossed ANOSIM and SIMPER analysis. Prior to ANOSIM and SIMPER analyses, data were fourth-root transformed to increase the importance of rare species and Bray-Curtis similarities were calculated. All parametric statistics were conducted using JMP, while ANOSIM and SIMPER analyses were conducted with PRIMER-E (version 6, Primer-E Ltd., Plymouth UK).

Feeding preference assays

To determine herbivore feeding rates and assess whether *U. compressa* and *U. rigida* differed in their palatability, we conducted a series of paired-choice feeding experiments using each of six herbivores found at our field sites: juvenile (length <1 cm) Nereidae polychaetes (6 replicates), the amphipod *Gammarus mucronatus* (6 replicates), the hermit crab *Pagurus longicarpus* (7 replicates), the grass shrimp *Palaemonetes pugio* (8 replicates) and *P. vulgaris* (10 replicates), and the mud crab *Panopeus herbstii* (8 replicates; carapace width 9-18 mm). For each experiment, a single herbivore species was placed in a mesocosm with a piece of *U. compressa* and a piece of *U. rigida* of known wet mass (2-5 mg each). Due to the size disparity of our herbivores, we employed two mesocosm designs. For larger species (shrimp, mud crabs, and hermit crabs), we used 2.5 L plastic containers with mesh-covered holes in the sides for seawater flow; these were held in outdoor, flow-through seawater tables at the University of Rhode Island's Bay Campus (Narragansett, RI, USA). The seawater tables were supplied with filtered, ambient temperature seawater from Narragansett Bay. Smaller invertebrates (amphipods

and polychaetes) were placed within 250 ml shallow glass bowls with filtered, room temperature (approximately 21°C) seawater and an air stone; seawater was changed daily for the duration of these experiments. In addition, experiments utilizing the smaller herbivores, as well as the *P. pugio* experiment, included two individuals per replicate mesocosm, while mesocosms with larger species contained a single individual.

Each paired-choice experiment ran for 3 days, except for the *P. pugio* trial, which ran for 7 days to ensure no feeding occurred (see 'Results'). To account for autogenic changes in *Ulva* blade wet mass, an equal number of non-herbivore controls were run concurrently with each experiment in identical containers. In experiments where control *Ulva* blades grew significantly, we adjusted the final wet mass of all thalli by the amount of growth observed in the controls. Results of each paired-choice assay were analyzed using matched pairs t-tests.

Characteristics of Ulva compressa *and* Ulva rigida

To determine if algal chemical or physical properties might be responsible for feeding preferences, we assessed several characteristics that may influence *Ulva* palatability. For each herbivore exclusion experiment, we measured *Ulva* organic content as the percent ash-free mass of each thallus by combusting dried tissue samples in a muffle furnace at 500°C for two hours (Thornber et al., 2008). Organic content for 2009 was analyzed for differences among months, cage types, and sites using a three-way repeated measures ANOVA that compared organic content values from the start and end of each experiment. Data from 2010 were analyzed for differences among species, months, cage types, and sites with a fully-factorial repeated measures ANOVA, with species nested within cage type.

In addition, we determined the dimethylsulfoniopropionate (DMSP; a known chemical precursor to herbivore deterrents) levels (as % of dry weight) of *U. compressa* and *U. rigida* blades collected in June (n = 10) and October 2010 (n = 8). To measure DMSP, we first dried the algae at 60ºC for seven days and then shipped them to the Shannon Point Marine Center (Anacortes, WA). Approximately 0.1 g of each piece was weighed and sealed in a gas-tight vial with 4 ml of 4N sodium hydroxide. The vials were stored in the dark overnight in order to hydrolyze the DMSP, which resulted in the cleavage of DMSP and the production of the volatile compound dimethylsulfide (DMS). DMS concentrations were determined in the headspace of the vials by injecting 10 μL headspace samples into an SRI GC equipped with a Chromasil 330 column in a 90ºC oven and a flame photometric detector (125ºC). Commercially obtained DMSP (Center for Analysis, Spectroscopy and Synthesis, University of Groningen) was used as a standard. DMSP results were analyzed via two-way ANOVA for differences among species and collection date.

Lastly, we determined *Ulva* blade tissue toughness for ten blades of each species collected from WCP in September 2009. Tissue toughness was determined using a tissue penetrometer to measure ten randomly selected locations on each blade (Duffy & Hay 1991); results for each blade were averaged prior to analysis by t-test.

Results

Herbivore exclusion experiments - 2009

In our 2009 herbivore exclusion experiments, *Ulva compressa* growth differed significantly among months, sites, and cage types (Fig. 2, Table 1a). On average, *U. compressa* grew 2.5 to 3 times faster at CH (mean 121.59 mg d^{-1}) and OBC (mean 110.37 mg d^{-1}) than at

240 WCP (mean 40.03 mg d^{-1}), with a maximum mean growth rate in May of 407.52 ± 31.65 mg d^{-1}
241 at OBC. Among the cage types, *U. compressa* grew an average of 35-40% more in the large and
242 medium mesh cages than in the small mesh cages, though this was largely driven by the
243 significant growth differences among the cage types in May. Our generalized linear model for
244 growth rate of cage mesh controls yielded a nonsignificant whole model response ($F_{11, 48}$ = 1.79,
245 P = 0.081; mean growth = 99.73 ± 5.97 mg d^{-1}), demonstrating a lack of difference in algal
246 growth rate due to abiotic variation, such as light levels and water flow, caused by the cage mesh
247 types. This indicates that *in situ* differences among cage types within a site were due to the
248 differential herbivory of the invertebrate communities the cages created.
249 Invertebrate assemblages within the cages at WCP and OCB were exceedingly similar to
250 each other and significantly different from CH during all months in 2009 (see Online Resource
251 Table S1a). Between site dissimilarity was mostly accounted for by the varying abundance of
252 amphipod taxa, with polychaetes and panopeid mud crabs accounting for a majority of the
253 remaining dissimilarity (Fig. 3, Online Resource Table S2a). Additionally, assemblages in the
254 small mesh cages were significantly different from those in the medium and large mesh cages in
255 every instance except July, where invertebrates within the small and medium mesh cages were
256 similar (Online Resource Table S1b). Significant differences between the invertebrates within
257 the large and medium mesh cages were only observed during June and August 2009 (Online
258 Resource Table S1b). Although found across all cage types, amphipod taxa were most abundant
259 within the small mesh cages, while panopeid mud crabs and *Palaemonetes* spp. shrimp were
260 most abundant in the large and medium mesh cages. Nereid polychaetes showed no clear
261 preference for any cage type (Fig. 3, Online Resource Table S2b).
262

Herbivore exclusion experiments - 2010

In 2010, *Ulva* growth again differed significantly among sites and cage types, as well as species (Fig. 4, Table 1b). In addition, when averaged across all sites and both months, *U. compressa* grew approximately 2.5 times more than *U. rigida*. As in 2009, our generalized linear model for growth rate of cage mesh controls yielded a nonsignificant whole model response ($F_{17, 58} = 0.94$, $P = 0.54$), although mean *U. compressa* growth rates (June: 57.67 mg d^{-1} and July: 75.21 mg d^{-1}) in these cage mesh controls were approximately 60% higher than *U. rigida* (June: 36.23 mg d^{-1} and July: 45.07 mg d^{-1}; $F_{1, 17} = 8.07$, $P = 0.006$).

As in 2009, mean *Ulva* growth for all months was highest at CH (65.04 ± 19.64 mg d^{-1}), although growth was lowest at OBC (mean 7.44 ± 18.49 mg d^{-1}) rather than WCP (18.87 ± 16.24 mg d^{-1}; Fig. 4). Invertebrate assemblages among the months and cage types showed similar patterns to those found in 2009 (Online Resource Table S1c, d); however, the abundance of amphipods in the families Aoridae and Gammaridae were notably higher at WCP and OBC, while amphipods in the family Corophiidae were notably higher at CH in July 2010 (Fig. 3, Online Resource Table S2a).

Herbivore exclusion experiments - General patterns

Mean water temperatures in 2009 were significantly different among all months ($F_{2, 8} = 106.31$, $P < 0.0001$) and significantly higher at OBC than at CH ($F_{2, 8} = 6.24$, $p = 0.003$; Online Resource Table S3). However, the difference in mean daily temperature among the sites within any individual month was less than 1.5°C. Mean water temperatures in 2010 were significantly higher in July than in June ($F_{1, 5} = 20.44$, $p < 0.0001$) but did not differ among sites ($F_{2, 5} = 2.18$, $p = 0.12$; Online Resource Table S3).

Mean water nitrate levels in 2009 were markedly higher at WCP (118.25 ± 43.97 μg L^{-1}) and OBC (151.5 ± 53.78 μg L^{-1}) than CH (25.00 ± 6.24 μg L^{-1}). Mean water nitrate levels were lower in 2010, with less variation among sites (CH 11.00 ± 4.00; WCP 12.00 ± 1.00; OBC 25.00 ± 14.57 μg L^{-1}).

In both 2009 and 2010, the invertebrate community found within the exclusion cages differed significantly from those found via net sampling (2009 global R = 0.77, P = 0.001; 2010 global R = 0.719, P = 0.001; Table S4). This difference was mostly due to the greater abundance of *Ilyanassa obsoleta* within net samples and a greater abundance of amphipods, mud crabs, and polychaetes within the cages (Online Resource Table S4). A greater abundance of shrimp and hermit crabs within the net samples also contributed to the dissimilarity (Online Resource Table S4).

Additionally, in 2009 and 2010 we found no relationship between the growth rate of either *Ulva* species and the total number of invertebrates found within the exclusion cages (Online Resource Fig. S5). *Ulva* growth rates were also not related to invertebrate species richness or diversity (Shannon H') within the cages (data not shown). When we examined the relationship between *Ulva* growth and individual taxa by month, significant negative relationships (r^2 = 0.13 to 0.39) were found between *U. compressa* growth and the abundance of amphipods in the families Gammaridae and Melitidae in July 2009, and growth rates for both *Ulva* species with mud crab abundance in July 2010 (Online Resource Fig. S6).

Feeding preference assays

Herbivore consumption of *Ulva* thalli was readily apparent for all herbivores except *Palaemonetes pugio* (Fig. 5). Thallus consumption was evident due to grazing marks on the thalli

and/or the continual presence of green hued feces within the mesocosms. Of the five herbivores that consumed *Ulva*, both *Gammarus mucronatus* and *Panopeus herbstii* consumed significantly more *U. compressa* than *U. rigida* (*G. mucronatus* $t_5 = 6.106$, $P = 0.0017$; *P. herbstii* $t_7 = 3.22$, $P = 0.015$; Fig. 5). *P. herbstii* also had the highest consumption rate, consuming an average of 5.88 mg d^{-1} of *U. compressa* and 1.25 mg d^{-1} of *U. rigida.* In contrast to the other assays, both *Ulva* species grew in the *P. pugio* assay, with *U. rigida* growing approximately 6 mg d^{-1} more in the presence of *P. pugio* than in the control treatment ($t_7 = -2.71$, $P = 0.03$).

Characteristics of Ulva compressa *and* Ulva rigida

In 2009, the organic content of *U. compressa* tissue was lowest during May (67.59 ± 0.64%) and highest in August (77.34 ± 0.80%; $F_{3, 129} = 56.37$, $P < 0.0001$; Table 2). Organic content also differed among sites ($F_{2, 129} = 6.06$, $P = 0.0031$). Thalli at CH (mean 72.40 ± 0.68%) had the lowest organic content for all months except July. In 2010, organic content again differed among sites ($F_{2, 100} = 5.07$, $P = 0.008$), however CH had the highest values (mean 69.48 ± 0.89%; Table 2). Additionally, *U. compressa* organic content (mean 68.85 ± 0.76%) was 2-6% higher than *U. rigida* (mean 65.10 ± 1.19%), though this difference was not significant ($F_{3, 100} = 1.72$, $P = 0.17$). For both years, organic content did not change throughout the course of the experiments (2009: $F_{1, 129} = 0.0011$, $P = 0.97$; 2010: $F_{1, 100} = 3.57$, $P = 0.062$).

The percent content of DMSP differed significantly between the *Ulva* species ($F_{1, 53} = 23.69$, $P < 0.0001$), but the direction of this difference depended on the month in which the blades were sampled (Month*Species $F_{1, 53} = 44.25$, $P < 0.0001$). *U. compressa* thalli collected in June 2010 had DMSP levels that were only 14% higher (mean 1.74 ± 0.06%) than those of *U.*

rigida (mean 1.53 ± 0.06%). In contrast, DMSP levels in *U. rigida* collected during October 2010 (mean 2.39 ± 0.09%) were 130% higher than those of *U. compressa* (mean 1.04 ± 0.12%).

Tissue toughness did not differ between *Ulva compressa* and *U. rigida* ($t_{21} = 0.68$, $P = 0.51$).

Discussion

Our results demonstrate that a wide variety of invertebrates found at bloom-impacted sites consume *Ulva* and measurably reduce its biomass. *In situ,* this was evidenced by significant growth differences among the cage types. Unfortunately, we were unable to include an *in situ* herbivore exclusion cage control to assess total *Ulva* growth rates, since mesh sizes small enough to exclude amphipods severely limited *Ulva* growth. However, controls run at our mesocosm facility demonstrated *Ulva* growth rates were unaffected by the abiotic environments created by the mesh sizes used in this experiment. Therefore, while the lack of a non-herbivore control prevented us from calculating total herbivory, we can confidently attribute significant differences in *Ulva* growth among cage types to the differences in the herbivore communities the cages created.

Based on the growth differences among the cage types, the herbivores that had the largest impact on *Ulva* growth varied monthly in both study years. Amphipods exerted the greatest herbivore pressure early in the bloom season, while mud crabs, and possibly shrimp, exerted equal or greater pressure later in the summer. In 2009, the largest discernable herbivore impact was observed in May within the small mesh cages, which were mainly occupied by amphipods and polychaetes. Throughout the remaining months of 2009, *U. compressa* growth was similar in all cage types. While this could indicate that amphipod consumption decreased after May, it

more likely represents an increase in consumption by larger herbivores found in the medium and large mesh cages as 1) *U. compressa* in our cages grew less in these months than in May, despite continued bloom proliferation at these sites (Guidone and Thornber, 2013) and 2) mud crabs were more abundant in these later months.

We observed a similar pattern in 2010. During June 2010, *Ulva* in the small and medium mesh cages experienced the greatest herbivore consumption, indicating amphipods were the dominant herbivores. In contrast, in July 2010 greater herbivory was observed in the medium and large mesh cages, which had a greater abundance of panopeid mud crabs and the shrimp *Palaemonetes vulgaris*.

Of the taxa identified within the small mesh cages, amphipods in the Gammaridae family had the largest detectable impact on *Ulva* biomass. They were the most abundant herbivore in the OBC small mesh cages during May 2009 when 282.2 mg d^{-1} more biomass was consumed in the small mesh than the large mesh cages. Their abundance was also negatively correlated to *U. compressa* growth during July 2009. Our mesocosm feeding assays and previous study (Horne et al., 1994) have confirmed *Gammarus mucronatus* as an *Ulva* consumer, and indeed, this was the dominant gammarid species in our samples. *Melita nitida*, the predominant melitid amphipod in our samples, represents another potential *Ulva* consumer as melitid abundance was negatively correlated with *Ulva* growth during July 2009.

Within the medium and large mesh cages, the most influential herbivore taxon was panopeid mud crabs. We observed a negative correlation between their abundance and *Ulva* growth in July 2010. The mud crab *Panopeus herbstii* also had the highest *Ulva* consumption rate in our mesocosm experiments, consuming an average of 7.13 mg d^{-1} of *Ulva* tissue. Although this was the greatest per capita impact we observed in our feeding assays, it is within

377 the range reported for other *Ulva* consuming mesoherbivores (e.g. *Idotea baltica*, 2.9-7.3 mg d^{-1};
378 Hauxwell et al., 1998; Nicotri, 1980). Based on our SIMPER analyses and previous study (Fox et
379 al., 2012), *P. vulgaris* is also likely to be grazing *Ulva* in these cages. However, we did not
380 observe an *in situ* relationship between *P. vulgaris* abundance and *Ulva* growth during any study
381 month. Low consumption rates during mesocosm assays further indicate that *P. vulgaris* may
382 only play a small role in regulating *Ulva* bloom biomass in this system.
383 While herbivory had a negative impact on *Ulva* biomass during both years, herbivore
384 consumption rarely exceeded the rate of *Ulva* growth, corroborating previous studies that found
385 that herbivory could not control bloom proliferation in high nutrient areas (Horne et al., 1994;
386 Hauxwell et al., 1998; Morgan et al., 2003; Worm & Lotze, 2006; Fox et al., 2012). Indeed, if *U.*
387 *compressa* were growing at the maximum rate observed during our study (503.38 mg d^{-1} at OBC
388 during May 2009), it would take a minimum of 71 *P. herbstii* mud crabs or 149 *G. mucronatus*
389 amphipods to completely consume the daily biomass produced by a *single blade* of *U.*
390 *compressa*. Even within our cages where these species congregated, we never observed densities
391 this high. As a square meter of a bloom mat can be composed of several large blades to over 200
392 individual smaller blades (Guidone and Thornber, unpubl. data), it is easy to see how blooming
393 *Ulva* readily escapes the influence of herbivory. Moreover, the observed temporal mismatch
394 between the start of rapid *Ulva* growth in late spring and the onset of mud crab and shrimp
395 herbivory in June to July may enhance *Ulva* bloom proliferation in this system (Svensson et al.,
396 2012).
397 Given that many herbivores appeared concentrated within our cages in comparison to our
398 net samples, it is possible that our field data overestimated the potential for herbivore control in
399 this system. However, it should be noted that our net samples rarely coincided with macrophytes

400 or drifting *Ulva*, which serve as habitat for these types of herbivores (e.g. Norkko et al., 2000).
401 Therefore, it is also possible that our net samples represent an underestimate of the herbivore
402 abundance that would be found within an *Ulva* mat. If this is the case, our cages may represent a
403 better estimate of herbivore abundance within these blooms than our net samples.
404 Contrary to our expectations, *Ulva* growth rates were often greatest at CH, the site that
405 we identified as least bloom impacted. Based on our limited point sampling of water DIN,
406 nutrient levels cannot explain the variation in *Ulva* growth among sites or months; CH had lower
407 DIN levels than WCP and OBC in 2009 and similar DIN levels to the other sites in 2010.
408 Differences in temperature among the sites also fail to explain differences in growth rates; mean
409 daily temperature never differed more than 1.5°C among the three sites. In contrast, invertebrate
410 community composition did differ significantly between CH and the other two sites. In
411 particular, CH had fewer mud crabs during most months, and a complete absence of melitid
412 amphipods except during August 2009. While growth differences among the sites may have been
413 influenced by unmeasured abiotic variables (e.g. water flow, salinity, light levels), our results
414 suggest that minor differences in the herbivore community may have a measurable impact on
415 *Ulva* growth among eutrophic sites.
416 Although we observed strong impacts of individual taxa on *Ulva* growth within particular
417 months, we did not find an overall negative correlation between *Ulva* growth and total
418 invertebrate abundance, nor were negative correlations between particular taxa and *Ulva* growth
419 consistent across all study months. This may be partially due to the study methods. Although our
420 *in situ* experiments provided for an estimate of relative herbivore pressure among the different
421 herbivore communities created by our cages, lack of an herbivore exclusion treatment limited our
422 ability to detect patterns in the larger data set. However, the high degree of variation found

423 within our study among sites and months is consistent with previous work (Morgan et al., 2003)
424 and could be due to a number of additional factors.
425 First, it is possible that our assessment of the herbivore communities within the cages was
426 biased towards slower moving organisms. Fast moving animals, such as shrimp or juvenile fish,
427 may have darted from the large and medium mesh cages while they were being placed into bags
428 for transport to the laboratory. The ability of juvenile fish to quickly escape is supported by our
429 observation that *Fundulus heteroclitus* (a known *Ulva* consumer, Sly, 2013) is common at all
430 three field sites (Guidone and Thornber, unpubl. data), yet they were never present within any of
431 the cage or net samples.
432 Second, it is unlikely that all invertebrates within our cages were consuming *Ulva* tissue,
433 therefore a correlation between the total number of invertebrates and *Ulva* growth should not
434 necessarily be expected. Among the non-*Ulva* consumers, some may have facilitated *Ulva*
435 growth by consuming fouling organisms or contributing nutrients through their nitrogenous
436 wastes, further obscuring our detection of herbivory patterns. The positive influences of
437 removing fouling organisms have been demonstrated for amphipods (Kamermans et al., 2002),
438 snails (Jormalainen et al., 2003; Råberg & Kautsky, 2008; Guidone et al., 2010, 2012;), chitons
439 (Littler et al., 1995), and aquatic insect larvae (Dudley, 1992). Mussels (Aquilino et al., 2009)
440 and snails (Guidone et al., 2012) can also facilitate macroalgal growth via nitrogenous wastes,
441 though this mechanism is unlikely to have an impact at nutrient enriched sites or sites with short
442 water residence times (Yarrington et al., 2013). Furthermore, omnivorous individuals may have
443 only occasionally consumed *Ulva* tissue. Even among the suspected herbivores, some may
444 choose to not consume *Ulva*. For examples, we observed *Palaemonetes pugio* would not

consume any *Ulva* tissue during mesocosm assays, even after one week with no other food source provided.

Last, variation within our data may have resulted from herbivore preferences that fluctuate temporally or spatially with *Ulva* tissue quality or defensive chemistry. While we found no difference in tissue toughness and only small differences in organic content, DMSP values differed significantly between the two *Ulva* species and the direction of this difference differed over time. Intraspecific DMSP values of *Ulva* in the Pacific Northwest have also been shown to vary widely (Van Alstyne et al., 2007), and levels of DMSP in *Ulva* species can vary with light (Karsten et al., 1991), salinity (Karsten et al., 1992), and temperature (Lyons et al., 2010). As DMSP is a precursor to the herbivore deterrents acrylic acid and DMS (Van Alstyne et al., 2001; Van Alstyne and Houser, 2003), if DMSP levels of *U. compressa* and *U. rigida* vary over the course of the summer or among sites, then herbivore preferences may fluctuate accordingly. However, it is also worth noting that a clear pattern between herbivore consumption and *Ulva* DMSP levels could be obscured if not all herbivores in a given habitat are deterred by acrylic acid or DMS (Erickson et al., 2006; Van Alstyne et al., 2009). Therefore, a detailed examination of temporal and spatial patterns of *Ulva* DMSP levels, in conjunction with herbivore feeding assays, is needed to clarify the role of DMSP in herbivore *Ulva* preferences and overall bloom species composition in this system.

One consistent relationship that emerged from our 2010 *in situ* experiments is that *Ulva rigida* grew significantly less than *U. compressa*, even showing net biomass losses at WCP and OCB during July 2010. However, two lines of evidence suggest that this is not because *U. rigida* was consumed more than *U. compressa*. First, our cage controls held at our mesocosm facility demonstrated that *U. compressa* growth rates were greater than those of *U. rigida* in the absence

of herbivory. Had we been able to incorporate an *in situ* non-herbivore control, adjustments for growth rates would likely indicate *U. compressa* was consumed as much, or more than, *U. rigida*. Additionally, both herbivores that demonstrated a feeding preference in our mesocosm paired-choice feeding assays preferred *U. compressa* to *U. rigida*.

Our findings highlight the complex nature of invertebrate herbivore impacts on macroalgal growth when examined across months, sites, and bloom-forming species within a eutrophic system. Previous studies have demonstrated that both herbivore and macrophyte species identity are important factors to consider when determining herbivore impacts on macrophytes (e.g. Duffy et al., 2001; Duffy & Harvilicz, 2001). Indeed, within our mesocosm experiments, we observed that two *Palaemonetes* species had markedly different impacts on *Ulva* growth, with *P. vulgaris* consuming both *Ulva* species and *P. pugio* facilitating *U. rigida* growth. Additionally, our results point to growth rate and palatability differences between the morphologically similar *U. compressa* and *U. rigida* that likely influence species composition within these *Ulva* blooms, similar to previous studies of multi-species blooms with morphologically and/or chemically distinct species (eg. Lotze & Worm 2000; Nelson et al. 2008). While closely related species may form functional groups that can have similar ecosystem impacts (e.g. Steneck & Dethier, 1994), our results support the view that the complexity within a functional group may be just as relevant to ecosystem structure as differences across functional groups (e.g. Nelson et al., 2008; Thornber et al., 2008; Burkepile & Hay, 2010).

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502

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662

663 **Figures**

664

665 **Fig. 1** Map of Narragansett Bay showing the location of our herbivore exclusion field sites:
666 Chepiwanoxet (CH), Warwick City Park (WCP), and Oakland Beach Cove (OBC). Photos: C.
667 Deacutis

668 **Fig. 2** Mean daily change in *Ulva compressa* wet mass (± 1 SE) in small (S), medium (M), and
669 large (L) mesh cages at Chepiwanoxet (CH), Warwick City Park (WCP), and Oakland Beach
670 Cove (OBC) during 2009 herbivore exclusion experiments: a) May, b) June, c) July, and d)
671 August.

672 **Fig. 3** Mean abundance of the eight herbivores contributing most to the dissimilarity between
673 cage types and exclusion sites Chepiwanoxet (CH), Warwick City Park (WCP), and Oakland
674 Beach Cove (OBC) in May, June, July, and August 2009 and June and July 2010: a)
675 Corophiidae, b) Gammaridae, c) Aoridae, d) Unidentified gammarid amphipods, e) Panopeidae,
676 f) Melitidae, g) Nereidae, and h) *Palaemonetes vulgaris*

677 **Fig. 4** Mean daily change in *Ulva compressa* and *U. rigida* wet mass (± 1 SE) in small (S),
678 medium (M), and large (L) mesh cages during 2010 herbivore exclusion experiments: a) June
679 and b) July

680 **Fig. 5** Mean daily change in *Ulva compressa* and *U. rigida* wet mass (per individual herbivore, ±
681 1 SE) in paired-choice feeding assays. * indicates a significant ($p < 0.05$) difference

Figure
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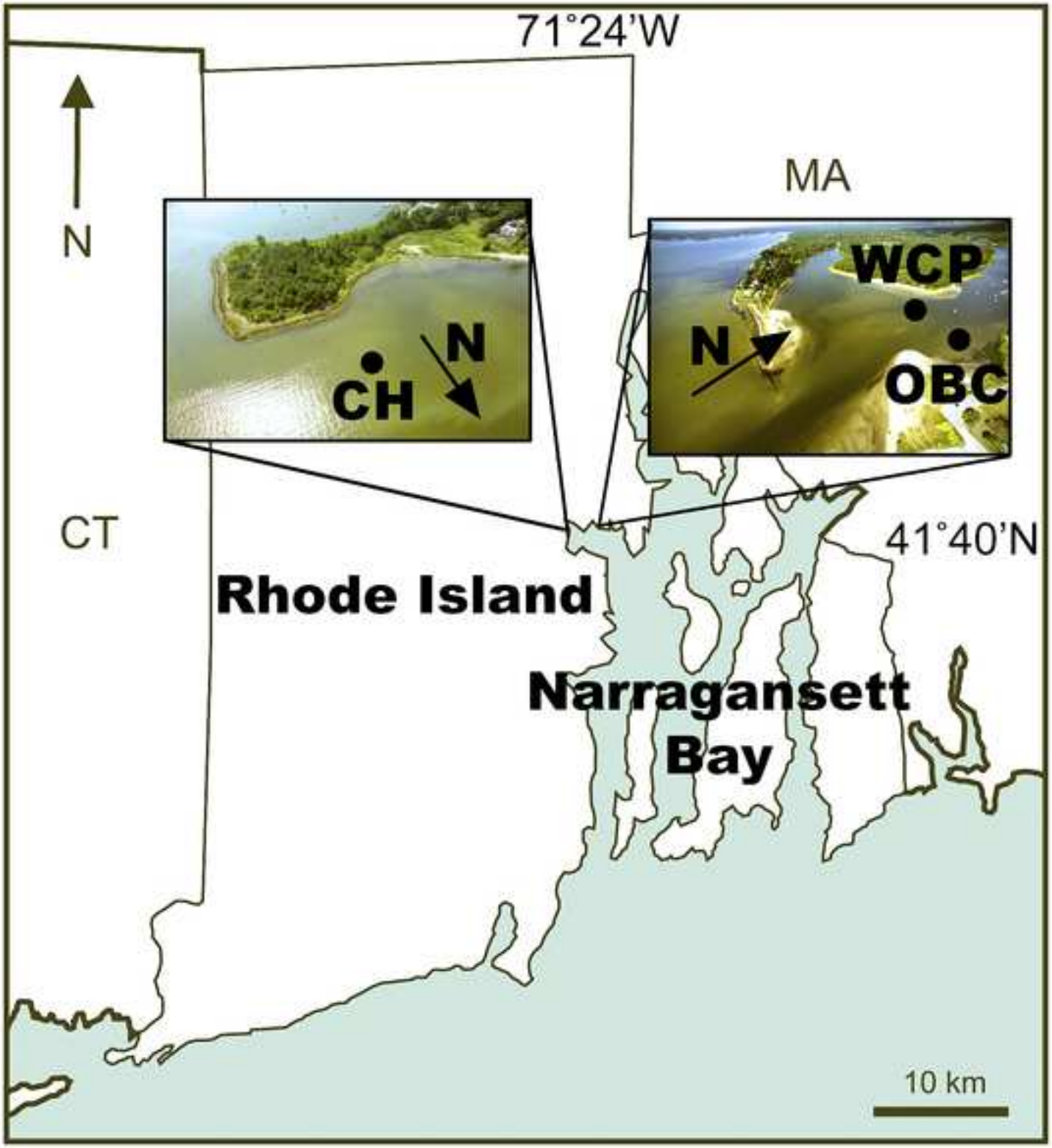

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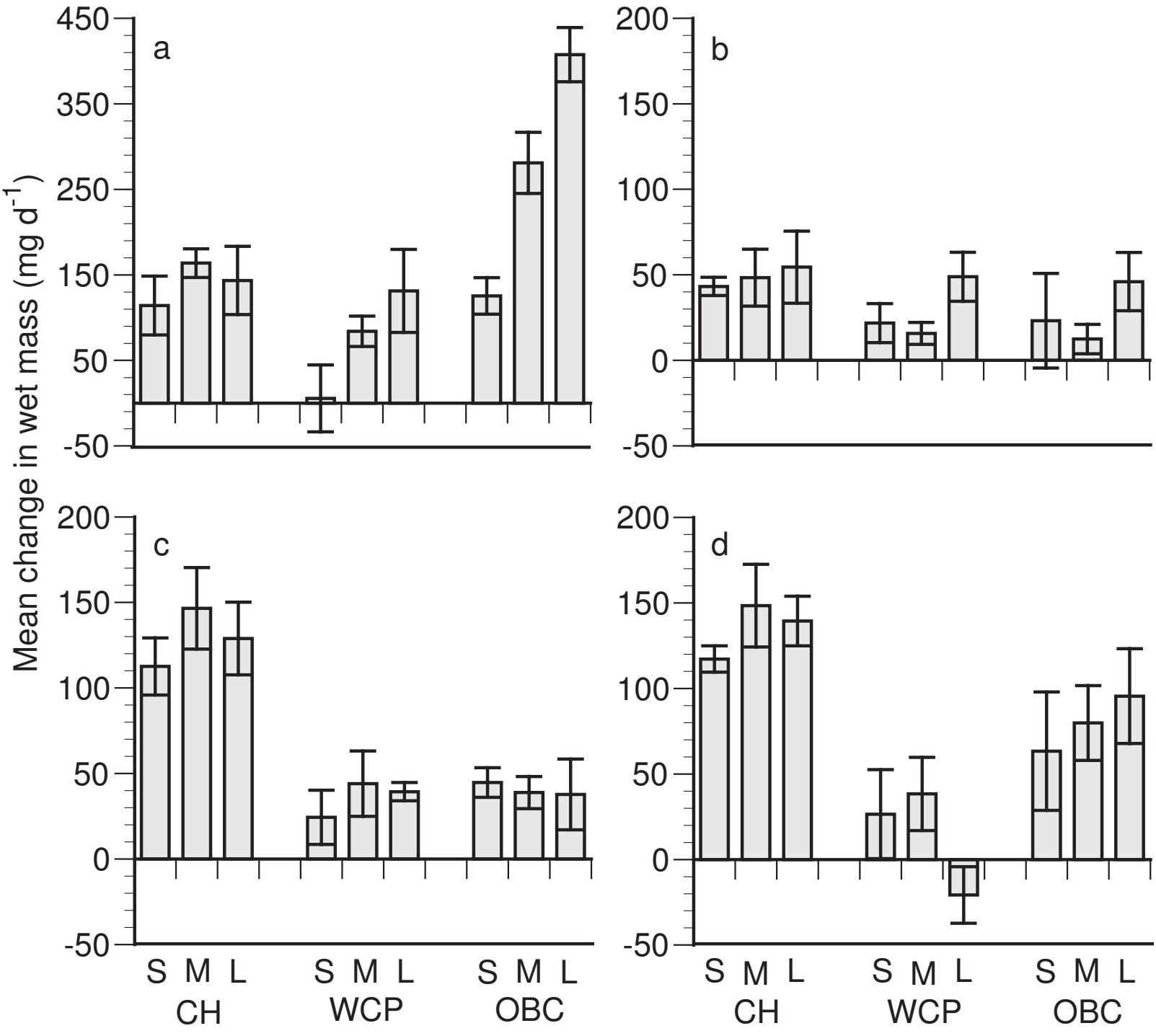

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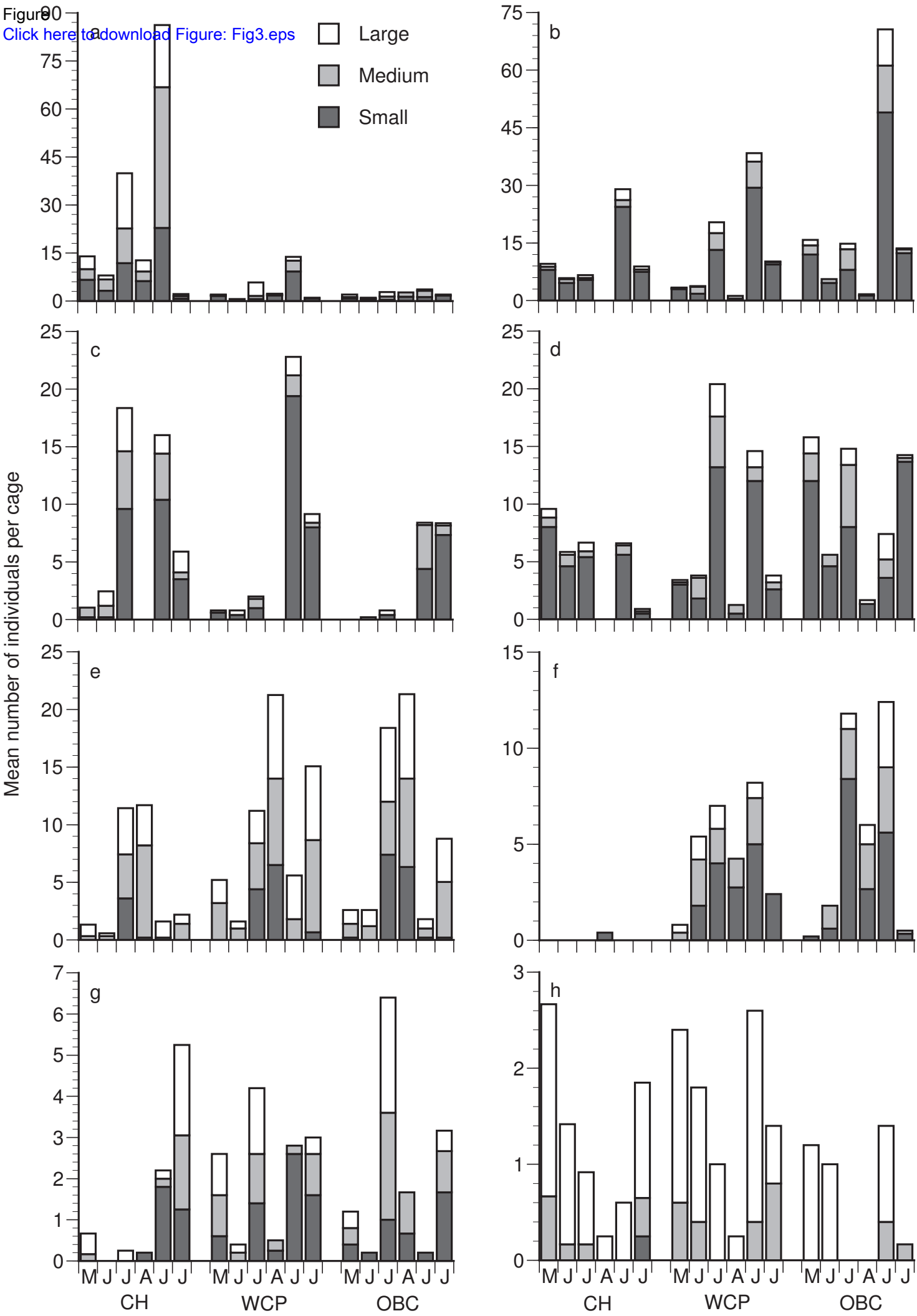

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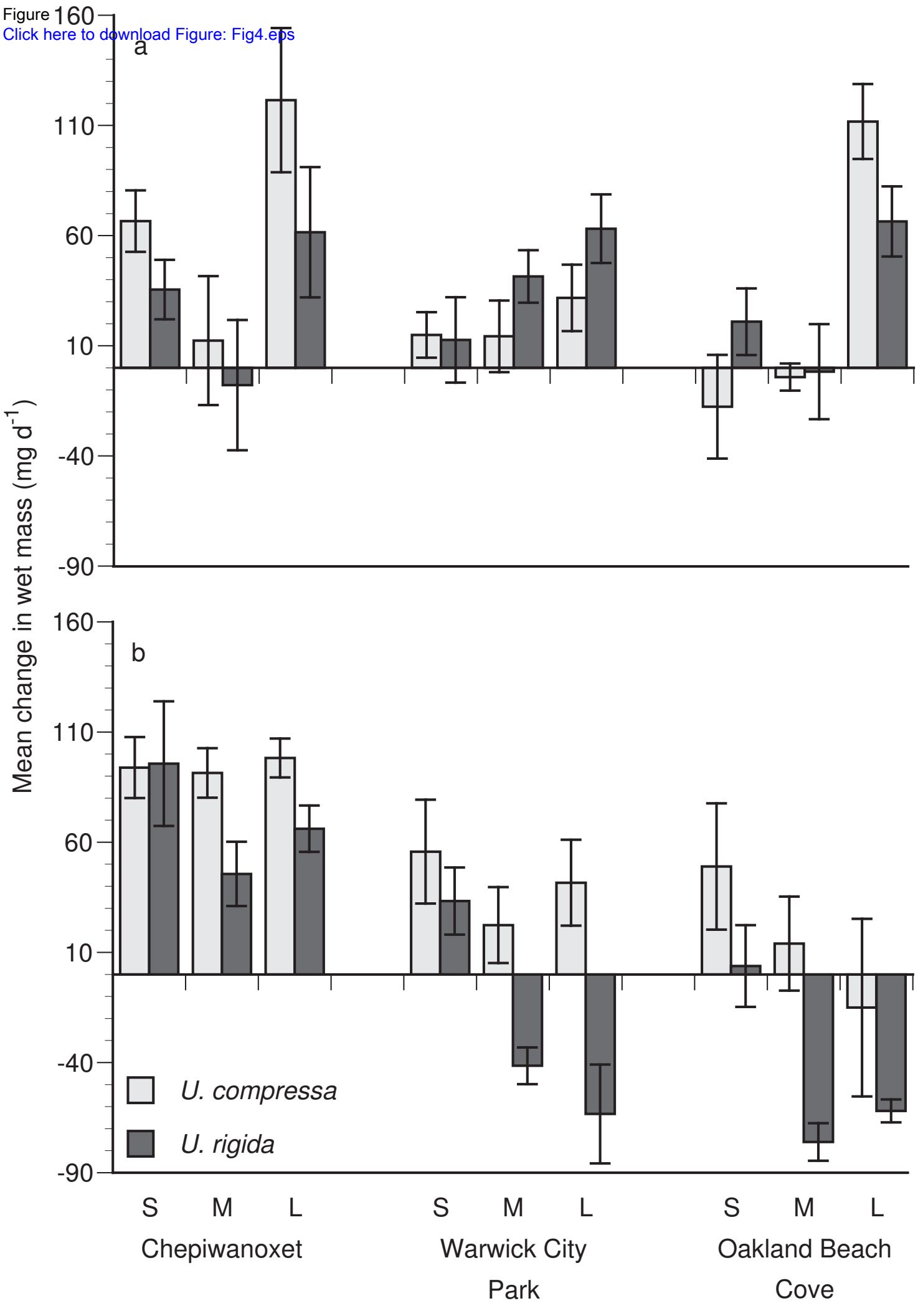

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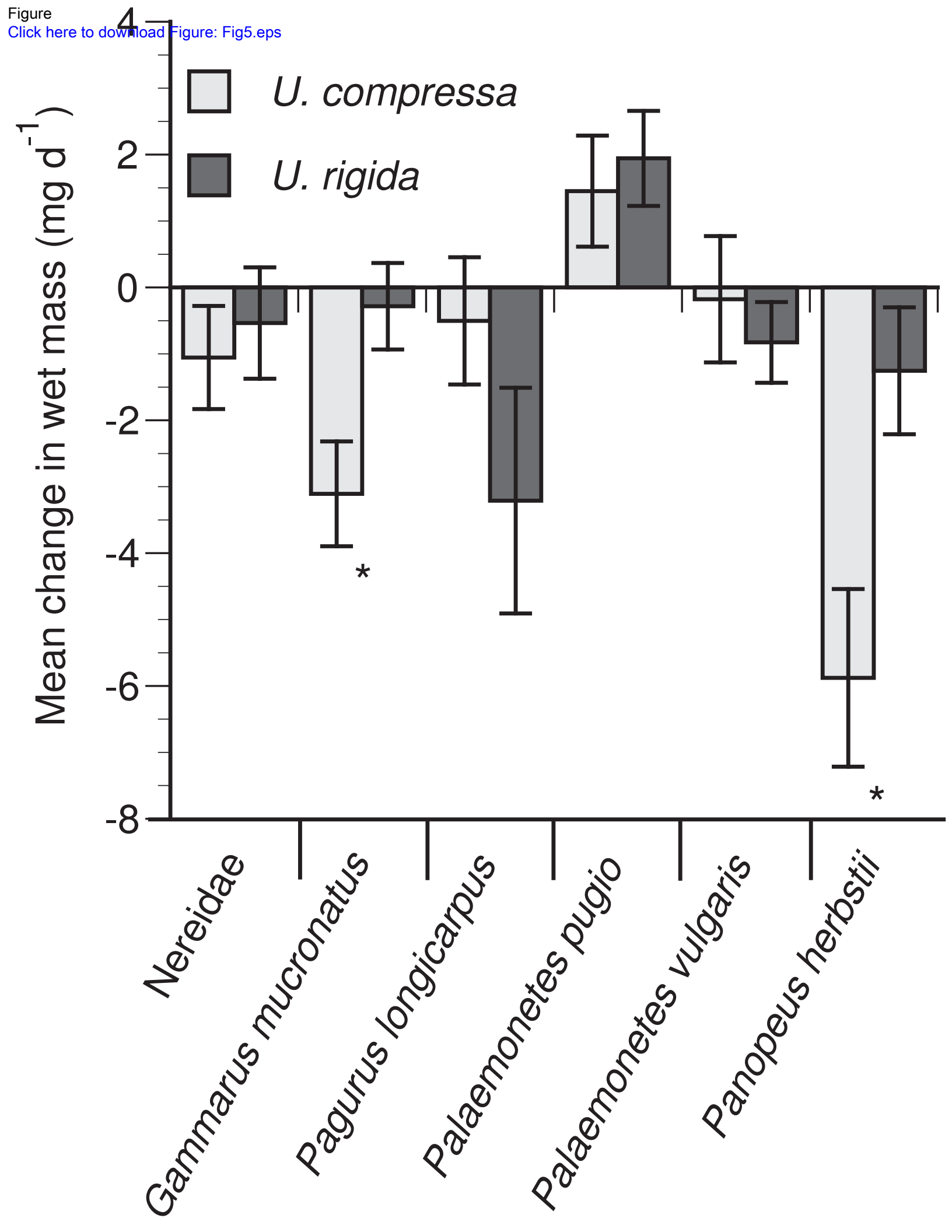

Table
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Table 1. Results of a) a three-way ANOVA on *Ulva compressa* growth during our 2009 herbivore exclusion experiments and b) a nested ANOVA on *U. compressa* and *U. rigida* growth during our 2010 herbivore exclusion experiments

a)

Source	df	SS	*F*	*P*
Month	3	386,266.43	51.06	< 0.0001
Site	2	187,807.01	37.21	< 0.0001
Cage type	2	56,062.20	11.11	< 0.0001
Month*Site	6	290,652.97	19.19	< 0.0001
Month*Cage type	6	105,767.61	6.98	< 0.0001
Site*Cage type	4	22,564.29	2.24	0.069
Month*Site*Cage type	12	66,465.06	2.19	0.015
Error	134	338,192.90		

b)

Source	df	SS	*F*	*P*
Month	1	4,818.37	2.59	0.11
Site	2	107,000.88	28.79	< 0.0001
Cage type	2	41,594.60	7.70	< 0.0001
Species [Cage type]	3	42,905.20	11.19	< 0.0001
Month*Site	2	45,162.94	12.15	< 0.0001
Month*Cage type	2	68,701.45	18.48	< 0.0001
Month*Species [Cage type]	3	25,855.26	4.64	0.004
Site*Cage type	4	10,006.56	1.35	0.26
Site*Species [Cage type]	6	2,267.73	0.20	0.98
Month*Site*Cage type	4	20,507.03	2.76	0.030
Month*Site*Species [Cage type]	6	29,567.29	2.65	0.018
Error	140	260,182.24		

Table
Click here to download Table: Table2.docx

Table 2. *Ulva compressa* and *U. rigida* tissue organic content

Species	Year	Month	Site	% Organic (± 1 SE)
U. compressa	2009	May	CH	66.07 ± 1.06
			WCP	68.53 ± 0.86
			OCB	68.25 ± 1.34
		June	CH	71.60 ± 1.04
			WCP	74.37 ± 0.71
			OCB	74.11 ± 1.22
		July	CH	76.42 ± 1.12
			WCP	76.91 ± 1.04
			OCB	75.66 ± 1.15
		August	CH	75.47 ± 1.24
			WCP	79.94 ± 1.80
			OCB	76.87 ± 0.71
U. compressa	2010	June	CH	70.68 ± 1.03
			WCP	69.68 ± 1.35
			OCB	69.68 ± 2.22
		July	CH	69.70 ± 1.11
			WCP	65.76 ± 1.02
			OCB	60.30 ± 4.34
U. rigida	2010	June	CH	69.59 ± 2.03
			WCP	66.10 ± 1.79
			OCB	67.08 ± 2.49
		July	CH	68.12 ± 2.35
			WCP	59.86 ± 3.51
			OCB	67.22 ± 3.01

Electronic Supplementary Material:

Guidone M*, Thornber CS, Van Alstyne KL. Herbivore impacts on two morphologically similar bloom-forming *Ulva* species in a eutrophic bay. *Hydrobiologia.*

*For correspondence: michele.guidone@armstrong.edu; affiliation: Armstrong State University

Table S1. Results from two-way crossed ANOSIMs for differences among sites and cage types for each month in our 2009 (a, b) and 2010 (c, d) herbivore exclusion experiments. R-values close to 1.00 indicate complete separation between groups while R-values close to 0 indicate little separation between groups. * indicates a significant pairwise tests ($p < 0.05$). (a, c) Tests for differences between site groups across all cage type groups. (b, d) Tests for differences between cage type groups across all site groups

(a)

	Chepiwanoxet	Warwick City Park	Oakland Beach
May: Global R = 0.677*			
Chepiwanoxet			
Warwick City Park	0.352*		
Oakland Beach	0.367*	0.033	
June: Global R = 0.287*			
Chepiwanoxet			
Warwick City Park	0.372*		
Oakland Beach	0.416*	0.089	
July: Global R = 0.505*			
Chepiwanoxet			
Warwick City Park	0.844*		
Oakland Beach	0.587*	0.047	
August: Global R = 0.189*			
Chepiwanoxet			
Warwick City Park	0.287*		
Oakland Beach	0.281*	-0.007	

(b)

Global R: P = 0.002	Small Mesh	Medium Mesh	Large Mesh
May: Global R = 0.233*			
Small Mesh			
Medium Mesh	0.248*		
Large Mesh	0.414*	0.031	
June: Global R = 0.328*			
Small Mesh			
Medium Mesh	0.234*		
Large Mesh	0.641*	0.123*	
July: Global R = 0.119*			
Small Mesh			
Medium Mesh	0.066		
Large Mesh	0.266*	0.047	
August: Global R = 0.133*			
Small Mesh			
Medium Mesh	0.111*		
Large Mesh	0.255*	0.042*	

(c)

	Chepiwanoxet	Warwick City Park	Oakland Beach
June: Global R = 0.424*			
Chepiwanoxet			
Warwick City Park	0.430*		
Oakland Beach	0.738*	0.093	
July: Global R = 0.193*			
Chepiwanoxet			
Warwick City Park	0.356*		
Oakland Beach	0.240*	-0.016	

(d)

	Small Mesh	Medium Mesh	Large Mesh
June: Global R = 0.299*			
Small Mesh			
Medium Mesh	0.330*		
Large Mesh	0.503*	0.054	
July: Global R = 0.216*			
Small Mesh			
Medium Mesh	0.365*		
Large Mesh	0.351*	-0.028	

Table S2. Results from a two-way crossed SIMPER analysis for average differences in herbivore assemblages between a) sites and b) cage types during our 2009 and 2010 herbivore exclusion experiments. Only the five taxa contributing the largest percentage of dissimilarity to a pair-wise comparison are shown

a)

Between site dissimilarity	Percentage contribution to average dissimilarity between sites		
Taxon	CH-WCP	CH-OBC	WCP-OBC
May 2009			
Corophiidae	16.29	14.23	8.92
Gammaridae	10.43	10.64	15.23
Lacuna vincta	8.13	9.76	—
Nereidae	6.56	6.48	11.17
Panopeidae	8.21	6.59	6.46
Phyllodocidae	7.68	8.55	10.87
Unidentified gammarid	7.59	7.76	10.21
Average between site dissimilarity	66.27	64.54	65.01
June 2009			
Aoridae	7.38	8.23	5.56
Corophiidae	13.72	15.09	5.45
Gammaridae	8.30	6.90	7.85
Melitidae	11.60	7.10	9.96
Panopeidae	5.09	7.79	8.25
Phyllodocidae	13.20	7.84	12.17
Unidentified gammarid	9.57	11.73	15.06
Unidentified polychaete	7.69	9.26	7.15
Average between site dissimilarity	64.78	67.73	57.41
July 2009			
Aoridae	14.59	15.97	10.03
Corophiidae	16.65	15.74	11.09
Gammaridae	9.17	8.10	7.59
Melitidae	14.14	15.28	10.41
Nereidae	8.87	11.58	10.85
Unidentified gammarid	6.98	8.18	9.12
Average between site dissimilarity	45.36	49.17	34.72
August 2009			
Corophiidae	15.19	14.83	8.49
Gammaridae	9.46	10.09	11.22

Melitidae	8.79	7.79	9.53
Panopeidae	10.11	9.91	10.31
Unidentified gammarid	10.20	10.17	11.05
Average between site dissimilarity	66.45	66.15	60.09
June 2010			
Aoridae	5.75	6.72	11.87
Corophiidae	17.77	19.06	11.87
Gammaridae	7.16	8.01	11.47
Ilyanassa obsoleta	7.50	5.69	4.41
Melitidae	12.64	15.59	6.26
Panopeidae	7.71	5.21	7.16
Phyllodocidae	6.75	6.70	7.48
Streblospio benedicti	6.71	8.43	6.18
Unidentified gammarid	8.20	9.13	12.90
Average between site dissimilarity	52.29	52.82	39.94
July 2010			
Aoridae	10.23	11.44	9.49
Gammaridae	9.74	11.78	11.09
Ilyanassa obsoleta	17.19	18.04	7.82
Nereidae	8.37	9.72	11.48
Panopeidae	10.79	9.81	14.09
Unidentified gammarid	7.38	7.92	10.85
Average between site dissimilarity	64.40	70.50	60.26

b)

Between cage type dissimilarity	Percentage contribution to average dissimilarity between cage types		
Taxon	Small-Medium	Small-Large	Medium-Large
May 2009			
Corophiidae	16.29	14.23	8.92
Gammaridae	10.43	10.64	15.23
Lacuna vincta	8.13	9.76	—
Nereidae	6.56	6.48	11.17
Panopeidae	8.21	6.59	6.46
Phyllodocidae	7.68	8.55	10.87
Unidentified gammarid	7.59	7.76	10.21
Average between cage type dissimilarity	65.94	71.19	60.65
June 2009			
Corophiidae	9.03	7.87	4.95
Gammaridae	16.38	15.43	8.70

Palaemonetes vulgaris	3.28	9.96	11.12
Panopeidae	10.06	7.01	9.22
Phyllodocidae	7.49	8.71	7.43
Unidentified gammarid	11.58	10.76	14.01
Unidentified polychaete	9.60	5.53	9.59
Average between cage type dissimilarity	61.89	76.68	62.12
July 2009			
Aoridae	10.43	7.54	9.33
Corophiidae	9.75	9.97	11.36
Gammaridae	14.35	12.10	10.38
Melitidae	7.63	11.72	8.23
Nereidae	9.30	10.31	8.44
Unidentified gammarid	10.04	9.87	11.86
Average between cage type dissimilarity	31.92	38.07	33.24
August 2009			
Corophiidae	8.22	9.19	9.08
Gammaridae	14.56	13.07	9.35
Melitidae	7.31	7.10	6.62
Palaemonetes vulgaris	2.79	7.03	8.10
Panopeidae	14.43	12.95	10.39
Unidentified gammarid	10.83	10.27	11.07
Average between cage type dissimilarity	60.05	67.91	60.41
June 2010			
Aoridae	9.04	12.60	11.75
Corophiidae	12.03	8.75	10.24
Gammaridae	13.56	15.15	10.26
Ilyanassa obsoleta	7.13	3.54	8.16
Palaemonetes vulgaris	3.44	9.47	10.42
Panopeidae	8.83	10.62	7.71
Unidentified gammarid	10.83	9.84	8.18
Average between cage type dissimilarity	42.33	51.72	42.16
July 2010			
Aoridae	11.01	11.59	8.66
Gammaridae	11.73	14.23	8.17
Ilyanassa obsoleta	10.53	9.93	12.21
Nereidae	7.30	10.78	12.19
Panopeidae	15.38	14.95	13.37
Unidentified gammarid	9.48	12.02	8.21
Average between cage type dissimilarity	69.67	75.49	56.41

Table S3. Average experimental temperatures (°C ± 1SE) at each field site.

	Chepiwanoxet	Warwick City Park	Oakland Beach
June 2009	19.58 ± 0.044	19.25 ± 0.057	19.41 ± 0.059
July 2009	23.58 ± 0.040	24.01 ± 0.061	24.16 ± 0.081
August 2009	26.12 ± 0.056	26.78 ± 0.065	27.05 ± 0.084
June 2010	22.90 ± 0.099	23.38 ± 0.087	23.96 ± 0.099
July 2010	25.75 ± 0.042	26.41 ± 0.058	26.59 ± 0.065

Table S4. Results from one-way SIMPER analyses for average similarity and differences between cage and net invertebrate assemblages during 2009 and 2010

	Percent contribution	Cumulative percent
Within sample similarity – 2009		
Cage – Average similarity: 35.31		
Mud crab	27.04	27.04
Corophiidae	19.97	47.02
Unidentifiable gammarid	17.38	64.40
Gammaridae	15.50	79.90
Melitidae	5.16	85.06
Phyllodocidae	3.57	88.64
Nereidae	3.03	91.67
Net – Average similarity: 45.70		
Ilyanassa obsoleta	69.45	69.45
Pagurus spp.	13.11	82.56
Crangon septemspinosa	5.16	87.72
Palaemonetes pugio	3.87	91.59
Within sample similarity – 2010		
Cage – Average similarity: 37.28		
Gammaridae	24.24	24.24
Aoridae	17.69	41.93
Mud crabs	17.02	58.95
Unidentified gammarids	9.48	68.42
Corophiidae	8.53	76.95
Nereidae	7.47	84.43
Melitidae	4.88	89.30
Ilyanassa obsoleta	3.90	93.20
Net – Average similarity: 47.34		
Ilyanassa obsoleta	89.73	89.73
Gammaridae	2.00	91.74

Between sample dissimilarity	Percentage contribution to average dissimilarity between cage and net invertebrate samples	
Taxon	2009	2010
Aoridae	3.87	11.00
Corophiidae	11.70	8.45
Gammaridae	10.16	14.55
Crangon septemspinosa	2.24	—
Ilyanassa obsoleta	12.08	14.27
Melitidae	5.42	5.65
Mud crabs	13.62	12.52

Nereidae	4.22	7.09
Pagurus spp.	4.26	—
Palaemonetes pugio	3.03	—
Palaemonetes vulgaris	4.19	4.75
Phyllodocidae	4.43	3.98
Unidentifiable gammarid	10.07	8.18
Unidentifiable polychaete	2.90	—
Total contribution of taxa	92.19	90.46
Average between sample type dissimilarity	91.95	90.73

Fig. S5. Correlations between the change in *Ulva* wet mass (mg d^{-1}) and total invertebrate abundance (per cage) in our herbivore exclusion experiments. A) 2009, B) 2010 *U. compressa*, C) 2010 *U. rigida*

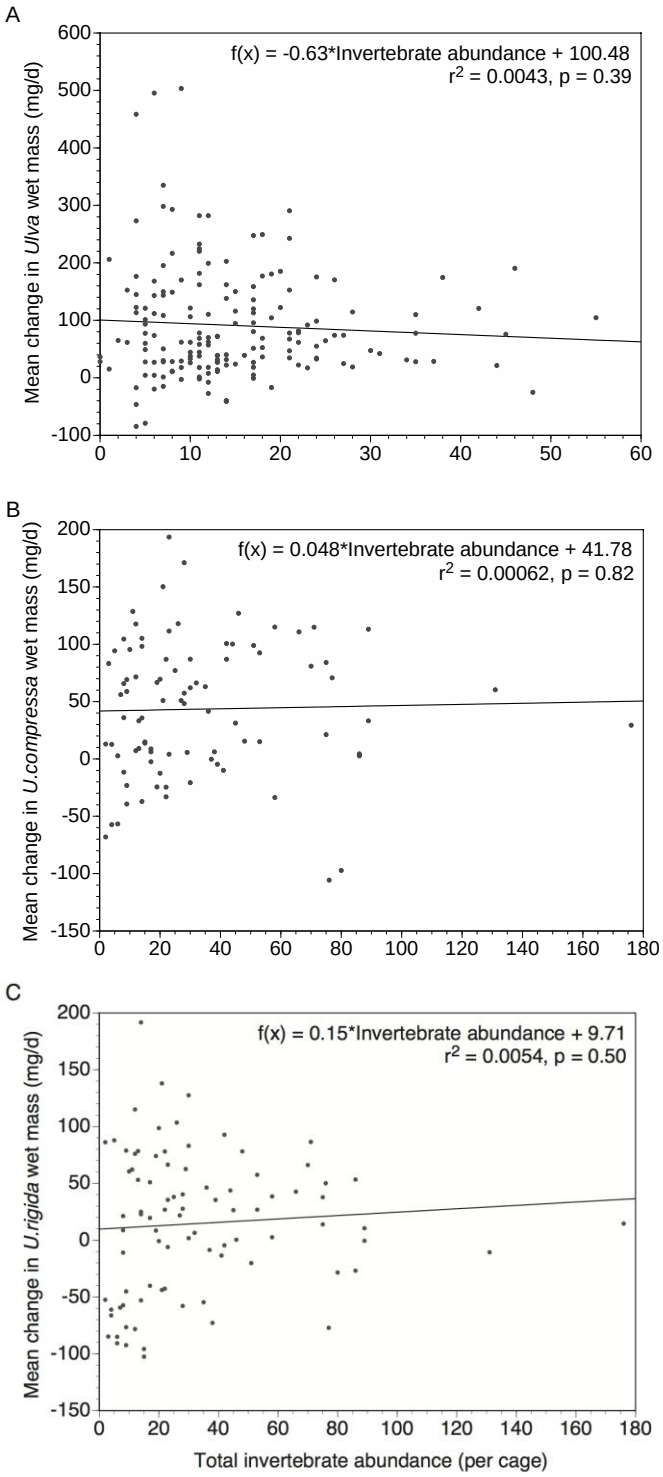

Fig. S6. Correlations between the change in *Ulva compressa* growth (mg d^{-1}) and A) Gammaridae abundance in July 2009, B) Melitidae abundance in July 2009, and C) Panopeidae mud crab abundance in July 2010. D) Correlation between *U. rigida* growth (mg d^{-1}) and Panopeidae mud crab abundance in July 2010