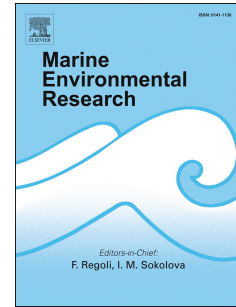


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**Enhanced nutrient loading and herbivory do not depress the resilience of subtidal canopy
forests in Mediterranean oligotrophic waters**

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24 Abstract

25 The interaction between top-down and bottom-up forces determines the recovery
26 trajectory of macroalgal forests exposed to multiple stressors. In an oligotrophic
27 system, we experimentally investigated how nutrient inputs affected the recovery of
28 *Cystoseira brachycarpa* following physical disturbance of varying intensities, both inside
29 forested areas and at the boundary with sea urchin barrens. Unexpectedly, *Cystoseira*
30 forests were highly resilient to disturbance, as they were able to recover from any
31 partial damage. In general, the addition of nutrients sped up the recovery of *Cystoseira*.
32 Thus, only the total canopy removal, in combination with either low nutrient availability
33 or intense grazing pressure, promoted the expansion of mat-forming algae or urchin
34 barrens, respectively. Our study suggests that the effects of enhanced nutrient levels
35 may vary according to the trophic characteristics of the waterbody, and hence, are likely
36 to vary among regions of the Mediterranean basin.

39 Keywords

40 Subtidal rocky reefs, Macroalgal forests, Sea urchin barrens, Mat-forming macroalgae,
41 Bottom-up versus top-down regulation, Disturbance, Multiple stressors, Resilience

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49 **1. Introduction**

50 Resilience is a key property of ecosystems, defining their capacity "to absorb
51 disturbance and reorganize while undergoing changes so as to retain the identity,
52 structure and functions of the original system, as opposed to switching to alternative
53 systems" (Holling, 1973). Experimental tests of resilience generally involve applying
54 pulse disturbances (i.e. acute events) of increasing intensity, in order to identify
55 conditions that could be tolerated and absorbed by the system without crossing a
56 critical threshold of change (Standish et al., 2014). On appropriate timescales, most
57 terrestrial and aquatic systems can fully recover in the absence of chronic,
58 anthropogenic stressors (Jones and Schmits, 2009). On the other hand, co-occurring
59 stressors often generates unpredictable interactive effects (Crain et al., 2008; Darling
60 and Cote, 2008). Trajectories of recovery from a pulse disturbance may divert towards
61 persistent alternative assemblages depending on the outcomes of the interaction
62 between top-down (predation or herbivory) and bottom-up forces (nutrient inputs),
63 (Scheffer and Carpenter, 2003; Scheffer et al., 2008; Connell et al., 2011; Reed et al.,
64 2011; Ortiz et al., 2018). Assessing the effects of these forces on ecological systems is a
65 crucial area of research, as they can decrease resilience and reduce the intensity of
66 disturbance (i.e. the threshold) required to switch an ecosystem to an alternative,
67 generally less complex and productive system.

68 Although macroalgal forests have demonstrated a high degree of resilience to
69 stressors compared to other habitats (e.g., corals or mangroves, Krumhansl et al., 2016
70 and references therein), their replacement by less diverse and productive assemblages
71 has been described worldwide (Benedetti-Cecchi et al., 2001; Gorgula and Connel, 2004;
72 Airoidi and Beck, 2007; Ling et al., 2015). Macroalgal forests are often extirpated by sea
73 urchin overgrazing and replaced by encrusting coralline-dominated assemblages (i.e.

74 urchin barrens), generally as a consequence of human over-exploitation of top
75 predators (Filbee-Dexter and Scheibling, 2014). When sea urchin density increases
76 above a threshold level, destructive grazing can trigger the shift to barrens, even in the
77 absence of other sources of disturbance (Filbee-Dexter and Scheibling, 2014). However,
78 barrens can also develop at a relatively low density of sea urchins following disturbance
79 events that reduce canopy biomass (Ebeling et al., 1985; Carnell and Keough, 2016).

80 In addition to grazing, reduced water quality and excessive nutrient loads cause a
81 diffuse decline in macroalgal forests and the proliferation of opportunistic, mat-forming
82 algae (Kennelly, 1987; Gorgula and Connell, 2004; Strain et al., 2014). In particular,
83 opportunistic filamentous species are able to rapidly colonise and retain space at the
84 expense of canopy-forming algae and, once established, can inhibit canopy recovery
85 through sediment trapping (Gorman and Connell, 2009; Piazzini and Ceccherelli, 2017a).
86 Yet, nutrient enrichment seems not to directly trigger the shift to filamentous algal
87 mats, as intact canopy stands have proved to be resistant to nutrient enrichment
88 (Falkenberg et al., 2012). In addition, the effects of enhanced nutrient loading on
89 canopy-forming macroalgae are likely dependent upon background nutrient levels,
90 since, in oligotrophic systems, moderate levels of nutrient enrichment can increase the
91 growth and hasten the recovery of canopy-forming species (Agatsuma et al., 2014).
92 Some studies have experimentally investigated the effects of disturbance on macroalgal
93 forests in combination with nutrient enrichment (Carnell and Keough, 2014; Piazzini and
94 Ceccherelli, 2017a), but the effects of physical disturbance have generally been explored
95 by completely removing canopy-forming macroalgae (Kraufvelin, 2007; Carnell and
96 Keough, 2014). Thinning macroalgae to attain different density levels in factorial
97 combination with nutrient enhancement allows identifying the intensity of damage that
98 could be absorbed by the system before switching to the alternative assemblage.

99 In addition, despite compelling evidence that species interactions differ at
100 boundaries between habitats (Fagan et al., 1999; Konar and Estes, 2003; Ries et al.,
101 2004; Bulleri and Benedetti-Cecchi, 2006), no study has investigated how variations in
102 the intensity of disturbance and nutrient loading influence canopy recovery inside
103 forests versus at the boundary with urchin barrens. Canopy recovery can be expected to
104 differ at the boundaries as a consequence of the fact that these boundaries are
105 characterized by different biotic (e.g. grazing pressure, spores and zygote supply) and
106 abiotic (e.g. hydrodynamic forces, sediment deposition and light) conditions compared
107 with the interior of forests. For example, the removal or thinning of canopy-forming
108 algae along the boundary of forested patches can facilitate the entrance of sea urchins
109 from barrens and stimulate an active grazing behaviour (Konar and Estes, 2003).

110 Using oligotrophic Mediterranean rocky reefs dominated by the canopy-forming
111 brown alga *Cystoseira brachycarpa* J. Agardh as a model system, we experimentally
112 assessed the importance of nutrient inputs in regulating macroalgal recovery from
113 physical disturbances of varying intensity (intact canopy stands, canopy rarefied to 70%
114 or 30% of cover, total canopy removal) inside forested areas and at the boundary with
115 barren areas.

116 We predicted that canopy recovery after 2 years from the start of the experiment would
117 be lower at the boundary with barrens than inside forests. More specifically, we
118 expected that thinning canopies below a given threshold at boundaries would facilitate
119 sea urchin grazing and, hence, result in the expansion of barrens. Enhanced nutrient
120 levels were expected to reduce canopy recovery by either promoting the development
121 of mat-forming algae or that of barrens by increasing grazing pressure as a consequence
122 of the higher nutritional value of algal tissues (lower C/N ratio; Balata et al., 2010;

123 Bakker and Nolet, 2014). Alternatively, enhancing nutrient levels may accelerate canopy
124 recovery by releasing macroalgae from N and P limitation.

125

126 **2. Materials and Methods**

127 **2.1 Study site and experimental design**

128 The study was done at Capraia Island in the Tuscany Archipelago (NW
129 Mediterranean, 43.05° N, 9.85° E) between June 2014 and July 2016. This volcanic
130 island is located 27.8 km from the mainland and is included in an MPA (Tuscany
131 Archipelago Natural Park). The study site, located on the south-eastern coast of the
132 island, is exposed to intense hydrodynamism (Bulleri et al. 2018). Shallow rocky reefs
133 between 2 and 8 m are dominated by macroalgal forests formed by the furoid canopy-
134 former *Cystoseira brachycarpa*. Forests are fragmented by patches dominated by mat-
135 forming algae and/or sea urchin barrens. Stands of *C. brachycarpa* host a rich
136 assemblage of benthic invertebrates including sponges, hydroids, anemones, spirorbids,
137 gastropods, bryozoans and ascidians, and a high diversity of seaweeds, either epiphytes
138 on its fronds or inhabiting the understory (Sales et al., 2012). Subtidal *Cystoseira* forests
139 have high ecological value, as they represent nursery areas for littoral fishes, and
140 provide food and shelter for many organisms (Cheminée et al., 2013). Gaps of canopy
141 removal or thinning are usually generated by intense storms (Navarro et al., 2011) or
142 by the grazing activity of the fish *Sarpa salpa* (Vergés et al., 2009) and of sea urchins
143 (Hereu 2006). Two species of sea urchins, namely *Paracentrotus lividus* and *Arbacia*
144 *lixula*, coexist in Mediterranean rocky reefs, where they create and maintain barren
145 grounds consisting of bare rock, and encrusting coralline and non-calcified prostrate
146 algae, and a low abundance of invertebrates (e.g. *Balanophyllia europaea*, *Vermetus* sp.,
147 encrusting bryozoans). Mat-forming algae, generally scarce beneath the canopy, include

148 foliose (e.g. *Dictyota* spp., *Padina pavonica*), filamentous (e.g. *Sphacelariales*), siphonous
149 algae (e.g. *Acetabularia acetabulum*, *Caulerpa cylindracea*) and corticated rhodophytae
150 (e.g. *Laurencia obtusa*, *Gastroclonium* sp.), (Bulleri et al., 2017).

151 At the beginning of the experiment, we identified 32 round boulders about 15 m²
152 in surface area, along a 600 m stretch of coast dominated by a subtidal *C. brachycarpa*
153 forest: 16 boulders were entirely covered by *C. brachycarpa* (hereafter, canopy beds),
154 whilst 16 hosted both canopy stands and barrens of variable width (3-12 m²), actively
155 maintained by sea urchins (Fig. 1). On each boulder, the density of sea urchins, *A. lixula*
156 and *P. lividus*, was quantified in 12 randomly placed 50x50 cm at 9 different times
157 throughout the duration of the experiment. On boulders with both macroalgal forests
158 and barrens, *A. lixula* was the most common sea urchin (mean density \pm SE = 3.344 \pm
159 0.168 \cdot m⁻²), while *P. lividus* was present only occasionally (mean density \pm SE = 0.136 \pm
160 0.024 \cdot m⁻²) (Fig. A.1 in Appendix A). Sea urchins were not found on boulders totally
161 covered by *C. brachycarpa*.

162 On each boulder, an area of 1.5 by 0.5 m was permanently marked in the middle
163 of canopy beds (> 1 m from the edge) or at the boundary with barren grounds, using
164 epoxy putty (Fig. 1). Experimental manipulation of physical disturbance and nutrient
165 loading were applied at the scale of the entire area. In each area, we permanently
166 marked 3 plots of 20x20 cm at their corners with epoxy putty (Fig. 1).

167 Boulders characterized by different habitats (i.e. full canopy cover versus mixed
168 canopy-barren habitat) were randomly assigned to different levels of disturbance
169 intensity (Fig. 1). Within the marked area, *C. brachycarpa* was either left untouched or
170 its cover was reduced by 30%, 70% or 100%. *C. brachycarpa* was removed with
171 hammer and chisel, paying attention not to damage understory organisms around the
172 holdfasts, to avoid artefact effects. Reducing the canopy cover by 30% and 70%,

173 respectively, simulated moderate and intense disturbances. Canopy cover reduction
174 was carried out by removing randomly identified individuals, in order to obtain a
175 homogeneous thinning of the canopy.

176 Two boulders for each combination of habitat and disturbance intensity were
177 randomly allocated to nutrient enrichment (Fig. 1). Nutrient levels were elevated using
178 slow release fertilizer pellets (Osmocote 6 months, 17:11:10 N:P:K) contained in plastic
179 net bags (1-mm mesh size). Nutrient enrichment was designed to simulate conditions
180 comparable to those recorded in urban areas of the same basin (Balata et al. 2010).
181 Eight mesh bags containing 100 g of fertilizer each were fixed in every area, by means of
182 steel hooks permanently fixed to the substratum. Nutrient bags were spaced out
183 between plots within an area, and their spatial distribution guaranteed the enrichment
184 of the whole area, while it did not influence adjacent boulders (Bulleri et al. 2012).
185 Nutrient bags were replaced every three months, in order to ensure the maintenance of
186 experimental conditions. Dissolution rates of fertilizer pellets vary through time
187 according to temperature and hydrodynamic regimes (Worm et al., 2000), yet this
188 approach provides a gradual release and realistically tracks natural fluctuations of
189 nutrient concentration in the water (Balata et al., 2010). The weight of fertilizer in each
190 nutrient bag was measured before deployment. Upon retrieval, nutrient bags were
191 dried in a muffle for 28 hours at 60 °C and the amount of fertilizer that had not
192 dissolved was re-weighted, in order to estimate the average nutrient release rate per
193 day over the duration of the experiment (Carnell and Keough, 2014). The amount of
194 fertilizer released did not vary among areas characterized by different habitats or
195 disturbance intensities (Table B.1, Fig. B.1 in Appendix B), and nutrients dissolved at an
196 average rate of 4,9 g per area (0.5 x 1.5 m) per day. In addition, at two random dates
197 during the experiment, water samples were collected from 30 enriched plots and 16

198 plots kept at ambient nutrient concentration. Two samples were taken approximately 3
199 cm above each plot, using a 60 ml syringe. Samples were immediately filtered (0.45 μm)
200 and frozen prior to transport to the laboratory for analysis. Nutrient concentrations in
201 seawater were determined using a continuous-flow AA3 Auto-Analyzer (Bran-Luebbe),
202 following standard methods (Grasshoff et al., 1999). Samples were analysed in triplicate
203 to obtain an average value for each sample. The study site is naturally oligotrophic, with
204 ambient nutrient concentrations much lower than those reported in coastal areas of the
205 same region (Balata et al., 2010; Gennaro and Piazzzi 2011). Although absolute
206 concentrations of nutrients in enriched plots were moderate ($\text{NO}_3 = 0.319 \pm 0.024$
207 $\mu\text{mol/l}$, $\text{NO}_2 = 0.141 \pm 0.022$ $\mu\text{mol/l}$, $\text{PO}_4 = 0.137 \pm 0.021$ $\mu\text{mol/l}$), higher concentrations of
208 nitrate, nitrite and phosphate were achieved in enriched plots compared to natural
209 environments (Fig. B.2 in Appendix B). However, while phosphate enhancement
210 attained levels comparable to those observed in urbanized coastal areas (Balata et al.,
211 2010), nitrate and nitrite concentrations were significantly lower than those reported in
212 other studies (Balata et al., 2010; Gennaro and Piazzzi 2011).

213 After two years since the beginning of the experiment, we visually sampled
214 experimental units, estimating percentage cover of algae and invertebrates by means of
215 a 20x20 cm plastic frame subdivided in 25 subquadrats. For each of the subquadrats, a
216 score from 0 to 4% was given to each taxon according to their relative cover (0= absent;
217 1 = occupation of 1/4 of the space; 2 = occupation of 2/4 of the space; 3 = occupation of
218 3/4 of the space; 4 = total occupation of the space) and the total percentage cover of
219 each taxon was calculated by summing the values across the subquadrats.

220

221 **2.2 Statistical analyses**

222 Effects of habitat, disturbance intensity and nutrient enrichment on the recovery of
223 benthic assemblages were tested by means of a permutational multivariate analysis of
224 variance (PERMANOVA, Anderson, 2001) performed on a Bray-Curtis dissimilarity
225 matrix of untransformed data, including the cover of *C. brachycarpa*. The model included
226 four factors: Habitat (fixed, with two levels: inside macroalgal forests versus boundary
227 between macroalgal forests and barrens), Disturbance Intensity (fixed, with four levels:
228 0%, 30%, 70%, 100% canopy removal, orthogonal to Habitat and Nutrient), Nutrient
229 (fixed, with two levels: ambient concentration, enhanced nutrients, orthogonal to
230 Habitat and Disturbance Intensity), and Boulder (random, nested within Habitat x
231 Disturbance Intensity x Nutrient, with two levels). Pairwise *a-posteriori* comparisons
232 were performed to assess differences among factor levels. Multivariate patterns were
233 visualized using nonmetric multidimensional scaling (nMDS) on the basis of a Bray-
234 Curtis dissimilarity matrix.

235 Benthic species of algae and invertebrates were divided into three broad
236 morphological groups, i.e. canopy, barren and mat-forming algae. The percentage cover
237 of each assemblage group was analysed with an ANOVA with the same design described
238 for PERMANOVA. Cochran's C-test was used to check for homogeneity of variances and
239 Student-Newman-Keuls (SNK) tests were used for the ranking of the means.

240

241 **3. Results**

242 Effects of canopy disturbance on the structure of benthic assemblages varied
243 among habitats (Table 1, Fig. 2). After 2 years, assemblages in plots exposed to the
244 initial removal of 70% or 100% of the canopy in both habitats were still different from
245 those in non-disturbed areas (0% removal). Following the total removal of the canopy,
246 the structure of the macroalgal assemblage inside the forest significantly differed from

247 that at the boundary with sea urchin barrens (Table 1, Fig. 2). Nutrient enrichment
248 modified the structure of assemblages and its effects were independent of the habitat or
249 disturbance intensity (Table 1). Enhanced nutrient loads favoured the recovery of *C.*
250 *brachycarpa*, while reducing the proliferation of mat-forming algae (Table 2, Fig. 3-4).

251 Inside forests, *C. brachycarpa* had fully recovered in areas where it had been
252 disturbed by removing up to 30% or 70% after 25 months (Table 2, Fig. 3). Canopy
253 recovery in totally cleared areas occurred only at enhanced nutrient concentrations
254 (Table 2, Fig. 3). Mat-forming algae became dominant after total canopy removal, with
255 cover values exceeding 70% at ambient nutrient concentration (Table 2, Fig. 4, Fig. A.2
256 in Appendix A). Nutrient enrichment had, in fact, a negative effect on mat-forming algae
257 in areas of total removal, and a similar negative tendency was observed in 70% canopy
258 removal areas (Table 2, Fig. 4, Fig. A.2 in Appendix A).

259 At the margins with barrens, nutrient addition had no influence on canopy
260 recovery in areas that were exposed to 30% clearance. The recovery of *C. brachycarpa*
261 in 70 % canopy removal areas was partial at ambient nutrient levels, while it was
262 complete at enhanced nutrient levels (Table 2, Fig. 3). In contrast, there was no canopy
263 recovery in total removal areas, regardless of nutrient levels (Table 2, Fig. 5). In fact,
264 these areas were dominated by species, such as encrusting corallines or bryozoans, that
265 characterize urchin barrens. Barrens increased only at the margins of areas where the
266 canopy had been totally removed, while no significant increment was observed in 30%
267 or 70% canopy removal areas or in undisturbed canopy boundaries (Table 2, Fig. 5, Fig.
268 A.3 in Appendix A).

269

270 **4. Discussion**

271 Inside canopy forests, in the absence of herbivores, *C. brachycarpa* was highly
272 resilient, as it totally recovered after removal of 30% or 70% of the canopy. Nutrient
273 enhancement promoted the complete recovery of *C. brachycarpa* canopy after total
274 removal and, contrary to our expectation, it did not stimulate the proliferation of mat-
275 forming algae. The positive effect of nutrient enrichment on growth and fertility of
276 Laminariales and Fucales has been observed in several systems (Hernández-Carmona et
277 al., 2001; Agatsuma et al., 2014; Tiffany et al., 2016). Our results suggest that, in
278 oligotrophic systems, a moderate nutrient enrichment can enhance the resilience of
279 canopy stands by increasing their competitive ability with turf-forming species.
280 Falkenberg et al. (2012) have shown that moderate nutrient enrichment strengthens
281 the ability of adult kelp individuals to maintain competitive dominance over mat-
282 forming algae and, by shading the substratum, inhibit their proliferation. While the
283 experiment by Falkenberg et al. (2012) tested kelp dominance of adult individuals (i.e.
284 resistance capability), the rapid recovery of *C. brachycarpa* in areas that were totally
285 cleared inside forests suggests that also its establishment can benefit from greater
286 nutrient availability.

287 The positive effect of moderate nutrient enrichment on the resilience of *C.*
288 *brachycarpa* is only apparently in contrast with the negative effect of eutrophication,
289 which is correlated with the historical decline of canopy stands across the
290 Mediterranean (Thibaut et al., 2005). For example, in the Adriatic Sea, eutrophic
291 conditions caused a drastic regression of *Cystoseira* populations, some of which
292 recovered when nutrient levels were reduced (Iveša et al., 2016). At smaller spatial
293 scales, detrimental effects of high nutrient loads on *Cystoseira* canopies have been
294 described in urbanized areas (Mangialajo et al., 2008; Piazzini and Ceccherelli, 2017a). In
295 habitats where macroalgal assemblages are dominated by opportunistic species,

296 ephemeral algae can rapidly propagate vegetatively, monopolize gaps opened by
297 disturbance events and competitively exclude canopy recruits (Kennelly 1987,
298 Korpinen and Jormalainen, 2008; Gorman and Connell, 2009). Even in the absence of
299 disturbance events, high nutrient loads can cause the decline of intact *Cystoseira* forests
300 (Piazzi and Ceccherelli, 2017a; Piazzi and Ceccherelli, 2017b). Yet, it is still debated
301 whether the regression of forests is due to the direct, negative effect of high nutrient
302 concentrations on adult *Cystoseira* or if it is mediated by the increase of opportunistic
303 algal species, represented by erect algae with fast growth and nutrient uptake rates
304 higher than *Cystoseira* species in eutrophic conditions. However, low or moderate
305 nutrient concentrations like those obtained in our experiment, have been reported to
306 advantage complex macroalgae, such as Fucales, over ephemeral species, as, in addition
307 to efficient external uptake, they can also use internal N stores (Foldanger Pedersen and
308 Borum, 1997). Regardless of the underpinning mechanism, our results suggest that in
309 oligotrophic systems as our, moderate nutrient enhancement is indeed able to foster the
310 persistence of canopies by sustaining its resilience to disturbance.

311 In the absence of herbivores, a combination of bottom-up effects and intensity of
312 physical disturbance is expected to control the persistence of canopy beds over long
313 time scales (Dayton et al., 1992; 1999). In some systems, such as Californian kelp
314 forests, the effects of disturbance overwhelm bottom-up processes and drive the
315 changes in the structure of macroalgal assemblage (Reed et al., 2011). In contrast, in our
316 system, even intense disturbance events that totally removed the canopy were not
317 sufficient by themselves to facilitate turfing algae dominance inside continuous canopy
318 beds when nutrients levels were moderately increased. Settlement and growth of
319 embryos released from adult plants at the margins of cleared areas, likely contributed
320 to *C. brachycarpa* recovery. Since the dispersal of *Cystoseira* embryos is very limited in

321 space (Mangialajo et al., 2012), it is possible that its recovery would decrease with
322 increasing extent of disturbed areas. After 25 months, a partial recovery of *Cystoseira*
323 canopy inside continuous canopy beds (18.7% of cover, Fig. 3) was observed also at
324 ambient nutrient levels. As reported for other systems, it is likely that nutrient
325 depletion might only delay canopy recovery to longer time scales (Dayton et al., 1992).

326 *Cystoseira brachycarpa* was more vulnerable to disturbance at the margin with
327 barren patches. While a total recovery of the canopy was observed for moderate
328 intensity of disturbance (removal of 30% of the canopy), severe perturbations caused a
329 substantial replacement of the canopy, which was substituted either by mat-forming
330 algae or by urchin barrens, respectively where 70% or 100% of the canopy was
331 removed. The vulnerability of the canopy at the edge of barrens is in contrast with the
332 findings by Konar and Estes (2003) and Bulleri and Benedetti-Cecchi (2006), who
333 detected a greater resilience and stability through time in the identity of habitats at
334 boundaries compared to clearings embedded within homogeneous habitat types. Worth
335 noting is that, in the latter study, algal assemblages were dominated by turf-forming
336 species, most of which are able to acquire space also through vegetative propagation.
337 Contrarily from those studies, intense disturbance (removal of 70% of the canopy) did
338 not cause the expansion of one of the two adjacent habitat types, but rather the
339 proliferation of a diverse algal assemblage made of ephemeral and opportunistic
340 species, which is otherwise suppressed by the presence of the competitive dominant *C.*
341 *brachycarpa*. At the edge of the forest, space occupation by mat-forming macroalgae in
342 70% canopy removal areas could be the result of either lower *C. brachycarpa* zygote
343 supply and/or increased growth rates under higher light levels in comparison with
344 areas inside the forest.

345 Yet, at enhanced nutrient concentrations, *C. brachycarpa* fully recovered and
346 prevented the colonization by these algal mats. The high resilience of *C. brachycarpa* to
347 severe disturbance (removal of 70% of the canopy) can be a result of its relatively fast
348 vegetative growth in all seasons of the year, differently from other *Cystoseira* species
349 that grow fronds only during spring. The species is characterized by a caespitose thallus
350 (i.e. growing in clusters), with blended stolon axes growing prostrate and generating
351 new root disks wherever they get in contact with the substratum, thus originating
352 numerous new cauloids and fronds (Sales and Ballesteros, 2012). At enhanced nutrient
353 concentrations, *C. brachycarpa* might have rapidly proliferated and expanded after
354 disturbance, thus building a barrier against the penetration of sea urchins in the midst
355 of canopy thalli. The sea urchin *Arbacia lixula*, which is the most abundant species at our
356 study site, is known not to be able to flex down *Cystoseira* fronds to the substratum and
357 feed on its fronds, differently from *Paracentrotus lividus* (Agnetta et al., 2015). The rapid
358 thickening of the canopy might have prevented *C. brachycarpa* germlings from grazing
359 during the first reproductive season after disturbance (i.e. January - February), thus
360 favoring the regrowth and rapid proliferation of the canopy within the first year after
361 disturbance. This is supported also by the high cover of *C. brachycarpa* observed after
362 only one year in nutrient enriched areas compared to natural environments (mean
363 cover in nutrient enriched areas = $71.3\% \pm 10.4$, at ambient nutrient concentrations =
364 $40.3\% \pm 4.5$, $n = 6$).

365 The efficacy of dense canopies in creating a mechanical barrier against the
366 penetration of sea urchins has been experimentally demonstrated by Konar and Estes
367 (2003), who showed how the sweeping motion of kelps or of their mimics might inhibit
368 urchins from crossing the boundary between barrens and canopy beds. If that was the
369 case also in our study, as suggested by direct observations of sea urchins only at the

370 edge or outside the canopy during the experiment (authors personal observation), we
371 may expect that, similarly to what observed within canopy beds, the complete recovery
372 of the canopy could have only been delayed to longer time scales by the grazing action
373 of sea urchins during the first few months after disturbance.

374 The expansion of barrens observed at the edge with forests where the canopy had
375 been totally removed, independently from the addition of nutrients, suggests that, in the
376 absence of mechanical barriers, the grazing activity of sea urchins prevented any
377 recovery of *C. brachycarpa*, either due to encroachment from lateral areas or to the
378 development of newly settled recruits. This is in accordance with the findings of Piazzini
379 and Ceccherelli (2017a), and confirms the relevant role of *Arbacia lixula* in maintaining
380 barrens, even at reduced densities (Bonaviri et al., 2011; Bulleri, 2013). At sea urchin
381 densities such as those found at our study site, intense disturbance appears as an
382 essential condition for the enlargement of barren areas. This result agrees with the
383 findings of Bulleri et al. (2018), who found a positive effect of wave-exposure on the
384 abundance of sea urchins and the extension of barren patches in the Tuscan
385 Archipelago. Several examples in the literature describe the formation of urchin barrens
386 following the removal of kelp forests due to intense storms (Dayton, 1985; Ebeling et al.,
387 1985).

388 In conclusion, our study brings novel evidence of unexpected resilience of
389 *Cystoseira* forests exposed to single and combined pressures, including mechanical
390 disturbance, moderate nutrient addition and low herbivore grazing pressure. At the
391 same study site, reducing the cover of intertidal *Cystoseira* canopies below 25% has
392 been shown to cause algal turf dominance, even in the absence of other stressors (Rindi
393 et al., 2018). In the subtidal, however, *C. brachycarpa* appears to recover, at least
394 partially, to any partial removal. Only the total removal of the canopy, in combination

395 with either low nutrient availability (this study), or intense grazing pressure (e.g. Piazzini
396 and Ceccherelli, 2017a; Ling et al., 2015), can limit its recovery.

397 Unexpectedly, addition of nutrients facilitated *Cystoseira* recovery rather than
398 representing a further stressor. Thus, our study suggests that the effects of enhanced
399 nutrient levels on the resilience of canopy forests vary according to the trophic
400 characteristics of the waterbody, and, hence, are likely to vary among regions of the
401 Mediterranean basin. Our results change the current view of the effects of nutrient
402 inputs on canopy-forming macroalgae being exclusively negative (Piazzini and
403 Ceccherelli, 2017a; Gorgula and Connell, 2009; Korpinen and Jormalainen, 2008).
404 Indeed, moderate enhancement of nutrient levels at oligotrophic sites appears to
405 sustain subtidal forests resilience. As such, strategies for the management of canopy-
406 forming macroalgae should be tailored to the biotic and abiotic characteristics of
407 regional rocky reefs.

408

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414

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418 Capraia Island for their assistance during fieldwork.

419

420 **Literature cited**

- 421 • Agatsuma, Y., Endo, H., Yoshida, S., Ikemori, C., Takeuchi, Y., Fujishima, H., Nakajima,
422 K., Sano, M., Kanezaki, N., Imai, H., Yamamoto, N., Kanahama, H., Matsubara, T.,
423 Takahashi, S., Isogai, T., Taniguchi, K., 2014. Enhancement of *Saccharina* kelp
424 production by nutrient supply in the Sea of Japan off southwestern Hokkaido, Japan.
425 J. Phycol. 26, 1845-1852.
- 426 • Agnetta, D., Badalamenti, F., Ceccherelli, G., Di Trapani, F., Bonaviri, C., Gianguzza, P.,
427 2015. Role of two co-occurring Mediterranean sea urchins in the formation of
428 barren from *Cystoseira* canopy. Estuar. Coast. Shelf Sci. 152, 73-77.
- 429 • Airoidi, L., Beck, M.W., 2007. Loss, status and trends for coastal marine habitats of
430 Europe. Oceanogr. Mar. Biol. Annu. Rev. 45, 345-405.
- 431 • Anderson, M.J., 2001. A new method for non-parametric multivariate analysis of
432 variance. Austral Ecol. 26, 32-46.
- 433 • Bakker, E.S., Nolet, B.A., 2014. Experimental evidence for enhanced top-down
434 control of freshwater macrophytes with nutrient enrichment. Oecologia 176, 825-
435 836.
- 436 • Balata, D., Piazzzi, L., Nesti, U., Bulleri, F., Bertocci, I., 2010. Effects of enhanced loads
437 of nutrients on epiphytes on leaves and rhizomes of *Posidonia oceanica*. J. Sea Res.
438 63, 173-179.
- 439 • Benedetti-Cecchi, L., Pannacciulli, F., Bulleri, F., Moschella, P.S., Airoidi, L., Relini, G.,
440 Cinelli, F., 2001. Predicting the consequences of anthropogenic disturbance: large-
441 scale effects of loss of canopy algae on rocky shores. Mar. Ecol. Prog. Ser. 214, 137-
442 150.

- 443 • Bonaviri, C., Fernandez, T.V., Fanelli, G., Badalamenti, F., Gianguzza, P., 2011. Leading
444 role of the sea urchin *Arbacia lixula* in maintaining the barren state in southwestern
445 Mediterranean. *Mar. Biol.* 158, 2505-2513.
- 446 • Bulleri, F., Cucco, A., Dal Bello, M., Maggi, E., Ravaglioli, C., Benedetti-Cecchi, L., 2018.
447 The role of wave-exposure and human impacts in regulating the distribution of
448 alternative habitats on NW Mediterranean rocky reefs. *Est. Coast. Shelf Sci.* 201, 114-
449 122.
- 450 • Bulleri, F., Benedetti-Cecchi, L., Ceccherelli, G., Tamburello, L., 2017. A few is enough:
451 a low cover of a non-native seaweed reduces the resilience of Mediterranean
452 macroalgal stands to disturbances of varying extent. *Biol. Invasions* 19, 2291-2305.
- 453 • Bulleri, F., 2013. Grazing by sea urchins at the margins of barren patches on
454 Mediterranean rocky reefs. *Mar. Biol.* 160, 2493-2501.
- 455 • Bulleri, F., Russell, B.D., Connell, S.D., 2012. Context-dependency in the effects of
456 nutrient loading and consumers on the availability of space in marine rocky
457 environments. *PLOS ONE* 7(3), e33825.
- 458 • Bulleri, F., Benedetti-Cecchi, L., 2006. Mechanisms of recovery and resilience of
459 different components of mosaics of habitats on shallow rocky reefs. *Oecologia* 149,
460 482-492.
- 461 • Carnell, P.E., Keough, M.J., 2014. Spatially variable synergistic effects of disturbance
462 and additional nutrients on kelp recruitment and recovery. *Oecologia* 175, 409-416.
- 463 • Carnell, P.E., Keough, M.J., 2016. The influence of herbivory on primary producers
464 can vary spatially and interact with disturbance. *Oikos* 125, 1273-1283.
- 465 • Cheminée, A., Sala, E., Pastor, J., Bodilis, P., Thiriet, P., Mangialajo, L., Cottalorda, J.,
466 Francour, P., 2013. Nursery value of *Cystoseira* forest for Mediterranean rocky reef
467 fishes. *J. Exp. Mar. Biol. Ecol.* 442, 70-79.

- 468 • Connell, S.D., Russell, B.D., Irving, A.D., 2011. Can strong consumer and producer
469 effects be reconciled to better forecast "catastrophic" phase-shift in marine
470 ecosystems? *J. Exp. Mar. Biol. Ecol.* 400, 296-301.
- 471 • Crain, C.M., Kroeker, K., Halpern, B.S., 2008. Interactive and cumulative effects of
472 multiple human stressors in marine systems. *Ecol. Lett.* 11, 1304-1315.
- 473 • Darling, E.S., Côté, I.M., 2008. Quantifying the evidence for ecological synergies. *Ecol.*
474 *Lett.* 11, 1278-1286.
- 475 • Dayton, P.K., Tegner, M.J., Edwards, P.B., Riser, K.L., 1999. Temporal and spatial
476 scales of kelp demography: the role of oceanographic climate. *Ecol. Monogr.* 69, 219-
477 250.
- 478 • Dayton, P.K., Tegner, M.J., Parnell, P.E., Edwards, P.B., 1992. Temporal and spatial
479 patterns of disturbance and recovery in a kelp forest community. *Ecol. Monogr.* 62,
480 421-445.
- 481 • Dayton, P.K., 1985. The structure and regulation of some South American kelp
482 communities. *Ecol. Monogr.* 55, 447-468.
- 483 • Ebeling, A.W., Laur, D.R., Rowley, R.J., 1985. Severe storm disturbances and reversal
484 of community structure in a southern California kelp forest. *Mar. Biol.* 84, 287-294.
- 485 • Fagan, W.F., Cantrell, R.S., Cosner, C., 1999. How habitat edges change species
486 interactions. *Am. Nat.* 153, 165-182.
- 487 • Falkenberg, L.J., Russell, B.D., Connell, S.D., 2012. Stability of strong species
488 interactions resist the synergistic effects of local and global pollution in kelp forests.
489 *PLOS ONE*, 7:e33841.
- 490 • Filbee-Dexter, K., Scheibling, R.E., 2014. Sea urchins barrens as an alternative stable
491 states of collapsed kelp ecosystems. *Mar. Ecol. Prog. Ser.* 495, 1-25.
- 492 • Foldanger Pedersen, M., Borum, J., 1997. Nutrient control of estuarine macroalgae:

- 493 growth strategy and the balance between nitrogen requirements and uptake. Mar.
494 Ecol. Prog. Ser. 161, 155-163.
- 495 • Gennaro, P., Piazzzi, L., 2011. Synergism between two anthropic impacts: *Caulerpa*
496 *racemosa* var. *cylindracea* invasion and seawater nutrient enrichment. Mar. Ecol.
497 Prog. Ser. 427, 59-70.
- 498 • Gorgula, S.K., Connell, S.D., 2004. Expansive covers of turf-forming algae on human-
499 dominated coast: the relative effects of increasing nutrient and sediment loads. Mar.
500 Biol. 145, 613-619.
- 501 • Gorman, D., Connell, S.D., 2009. Recovering subtidal forests in human-dominated
502 landscapes. J. Appl. Ecol. 46, 1258-1265.
- 503 • Grasshoff, K., Ehrhardt, M., Kremling, K., Anderson, L., 1999. Methods of seawater
504 analysis. Wiley-Vch Verlag, Weinheim 600 p.
- 505 • Hereu, B., 2006. Depletion of palatable algae by sea urchins and fishes in a
506 Mediterranean subtidal community. Mar. Ecol. Prog. Ser. 313, 95-103.
- 507 • Hernández-Carmona, G., Robledo, D., Serviere-Zaragoza, E., 2001. Effect of nutrient
508 availability on *Macrocystis pyrifera* recruitment and survival near its southern limit
509 off Baja California. Bot. Mar. 44, 221-229.
- 510 • Holling, C.S., 1973. Resilience and stability of ecological systems. Annu. Rev. Ecol.
511 Syst. 4, 1-23.
- 512 • Iveša, L., Djakovac, T., Devescovi, M., 2016. Long-term fluctuations in *Cystoseira*
513 populations along the west Istrian Coast (Croatia) related to eutrophication patterns
514 in the northern Adriatic Sea. Mar. Poll. Bull. 106, 162-173.
- 515 • Jones, H.P., Schmitz, O.J., 2009. Rapid recovery of damaged ecosystems. PLOS ONE,
516 4(5):e5653.
- 517 • Kennelly, S.J., 1987. Inhibition of kelp recruitment by turfing algae and consequences

- 518 for an Australian kelp community. *J. Exp. Mar. Biol. Ecol.* 112, 49-60.
- 519 • Konar, B., Estes, J.A., 2003. The stability of boundary regions between kelp beds and
520 deforested areas. *Ecology* 84, 174-185.
- 521 • Korpinen, S., Jormalainen, V., 2008. Grazing and nutrients reduce recruitment
522 success of *Fucus vesiculosus* L. (Fucales: Phaeophyceae). *Est. Coast. Shelf Sci.* 78, 437-
523 444.
- 524 • Kraufvelin, P., 2007. Responses to nutrient enrichment, wave action and disturbance
525 in rocky shore communities. *Aquat. Bot.* 87, 262-274.
- 526 • Krumhansl, K.A., Okamoto, D.K., Rassweiler, A., Novak, M., Bolton, J.J., Cavanaugh,
527 K.C., Connell, S.D., Johnson, C.R., Konar, B., Ling, S.D., Micheli, F., Norderhaug, K.M.,
528 Pérez-Matus, A., Sousa-Pinto, I., Reed, D.C., Salomon, A.K., Shears, N.T., Wernberg, T.,
529 Anderson, R.J., Barrett, N.S., Buschmann, A.H., Carr, M.H., Caselle, J.E., Derrien-
530 Courtel, S., Edgar, G.J., Edwards, M., Estes, J.A., Goodwin, C., Kenner, M.C., Kushner,
531 D.J., Moy, F.E., Nunn, J., Steneck, R.S., Vázquez, J., Watson, J., Witman, J.D., Byrnes,
532 J.E.K., 2016. Global patterns of kelp forest change over the past half-century. *Proc.*
533 *Nat. Acad. Sci., USA* 113, 13785-13790.
- 534 • Ling, S.D., Scheibling, R.E., Rassweiler, A., Johnson, C.R., Shears, N., Connell, S.D.,
535 Salomon, A.K., Norderhaug, K.M., Pérez-Matus, A., Hernández, J.C., Clemente, S.,
536 Blamey, L.K., Hereu, B., Ballesteros, E., Sala, E., Garrabou, J., Cebrian, E., Zabala, M.,
537 Fujita, D., Johnson, L.E., 2015. Global regime shift dynamics of catastrophic sea
538 urchin overgrazing. *Philos. Trans. R. Soc. Lond., B* 370, 1-10.
- 539 • Mangialajo, L., Chiantore, M., Cattaneo-Vietti, R., 2008. Loss of furoid algae along a
540 gradient of urbanization, and structure of benthic assemblages. *Mar. Ecol. Prog. Ser.*
541 358, 63-74.

- 542 • Mangialajo, L., Chiantore, M., Susini, M.L., Meinesz, A., Cattaneo-Vietti, R., Thibaut, T.,
543 2012. Zonation patterns and interspecific relationships of fucoids in microtidal
544 environments. *J. Exp. Mar. Biol. Ecol.* 412, 72-80.
- 545 • Navarro, L., Ballesteros, E., Linares, C., Hereu, B., 2011. Spatial and temporal
546 variability of deep-water algal assemblages in the Northwestern Mediterranean: the
547 effects of an exceptional storm. *Est. Coast. Shelf Sci.* 95, 52-58.
- 548 • Ortiz, J.C., Wolff, N.H., Anthony, K.R.N., Devlin, M., Lewis, S., Mumby, P.J., 2018.
549 Impaired recovery of the Great Barrier Reef under cumulative stress. *Science*
550 *Advances* 4, eaar6127.
- 551 • Piazzi, L., Ceccherelli, G., 2017a. Concomitance of oligotrophy and low grazing
552 pressure is essential for the resilience of Mediterranean subtidal forests. *Mar. Poll.*
553 *Bull.* 123, 197-204.
- 554 • Piazzi, L., Ceccherelli, G., 2017b. Eutrophication affects the resistance of fucoids to an
555 introduced alga spread. *Mar. Environ. Res.* 129, 189-194.
- 556 • Reed, D.C., Rassweiler, A., Carr, M.H., Cavanaugh, K.C., Malone, D.P., Siegel, D.A., 2011.
557 Wave disturbance overwhelms top-down and bottom-up control of primary
558 production in California kelp forests. *Ecology* 92, 2108-2116.
- 559 • Ries, L., Fletcher, R.J., Battin, J., Sisk, T.D., 2004. Ecological responses to habitat
560 edges: mechanisms, models, and variability explained. *Annu. Rev. Ecol. Evol. Syst.* 35,
561 491-522.
- 562 • Standish, R.J., Hobbs, R.J., Mayfield, M.M., Bestelmeyer, B.T., Suding, K.N., Battaglia,
563 L.L., Eviner, V., Hawkes, C.V., Temperton, V.M., Cramer, V.A., Harris, J.A., Funk, J.L.,
564 Thomas, P.A., 2014. Resilience in ecology: abstraction, distraction, or where the
565 action is? *Biol. Conserv.* 177, 43-51.

- 566 • Sales, M., Ballesteros, E., 2012. Seasonal dynamics and annual production of
567 *Cystoseira crinita* (Fucales: Ochrophyta)-dominated assemblages from the
568 northwestern Mediterranean. *Sci. Mar.* 76, 391-401.
- 569 • Sales, M., Ballesteros, E., Anderson, M.J., Ivesa, L., Cardona, E., 2012. Biogeographical
570 patterns of algal communities in the Mediterranean Sea: *Cystoseira crinita*-
571 dominated assemblages as a case study. *J. Biogeogr.* 39, 140-152.
- 572 • Scheffer, M., Carpenter, S.R., 2003. Catastrophic regime shifts in ecosystems: linking
573 theory to observation. *Trends Ecol. Evol.* 18, 648-656.
- 574 • Scheffer, M., van Nes, E.H., Holmgren, M., Hughes, T., 2008. Pulse-driven loss of top-
575 down control: the critical-rate hypothesis. *Ecosystems* 11, 226-237.
- 576 • Strain, E.M.A., Thomson, R.J., Micheli, F., Mancuso, F.P., Airolidi, L., 2014. Identifying
577 the interacting roles of stressors in driving the global loss of canopy-forming to mat-
578 forming algae in marine ecosystems. *Glob. Change Biol.* 20, 3300-3312.
- 579 • Thibaut, T., Pinedo, S., Torras, X., Ballesteros, E., 2005. Long-term decline of the
580 populations of Fucales (*Cystoseira* spp. and *Sargassum* spp.) in the Albères coast
581 (France, North-western Mediterranean). *Mar. Poll. Bull.* 50, 1472-1489.
- 582 • Tiffany, A., Stephens, C., Hepburn, D., 2016. A kelp with integrity: *Macrocystis*
583 *pyrifera* prioritises tissue maintenance in response to nitrogen fertilisation.
584 *Oecologia* 182, 71-84.
- 585 • Vergés, A., Alcoverro, T., Ballesteros, E., 2009. Role of fish herbivory in structuring
586 the vertical distribution of canopy algae *Cystoseira* spp. in the Mediterranean Sea.
587 *Mar. Ecol. Prog. Ser.* 375, 1-11.
- 588 • Worm, B., Reusch, T.B.H., Lotze, H.K., 2000. In situ nutrient enrichment: methods for
589 marine benthic ecology. *Int. Rev. Hydrobiol.* 85, 359-375.

591 **Table 1.** Permutational multivariate analysis of variance (PERMANOVA) on the effect of
 592 Habitat (H, inside the macroalgal forest, at the boundary between forest and barren),
 593 Disturbance Intensity (D, 0%, 30%, 70%, 100% canopy removal), and Nutrient (N,
 594 ambient concentration, enhanced nutrients) on the benthic assemblage among
 595 experimental Boulders (B). Pairwise tests for pairs of levels are reported for the
 596 interaction between Habitat and Disturbance intensity. * $P < 0.05$, ** $P < 0.01$, *** $P <$
 597 0.001; CR = canopy removal.
 598

Source of variation	df	MS	<i>Pseudo-F</i>
Habitat (H)	1	10967	8.594***
Disturbance intensity (D)	3	25785	20.206***
Nutrient (N)	1	4101.1	3.214*
H x D	3	7463.9	5.849***
H x N	1	1122.9	0.880
D x N	3	1966.2	1.541
H x D x N	3	1102.2	0.864
Boulder (H x D x N)	16	1276.1	3.323***
Residual	64	384.05	

<i>Pair-wise test for pairs of levels of Habitat</i>	
0% CR	Inside forest = Boundary
30% CR	Inside forest = Boundary
70% CR	Inside forest = Boundary
100% CR	Inside forest \neq Boundary

<i>Pair-wise test for pairs of levels of Disturbance intensity</i>	
--	--

Inside forest	0% CR = 30% CR	0% CR ≠ 70% CR
	0% CR ≠ 100% CR	30% CR = 70% CR
	30% CR ≠ 100% CR	70% CR ≠ 100% CR
Boundary	0% CR = 30% CR	0% CR ≠ 70% CR
	0% CR ≠ 100% CR	30% CR ≠ 70% CR
	30% CR ≠ 100% CR	70% CR ≠ 100% CR

599

600 **Table 2.** Analyses of variance (ANOVA) on the effect of Habitat (inside the macroalgal
601 forest, at the boundary between forest and barren), Disturbance Intensity (D, 0%, 30%,
602 70%, 100% canopy removal), and Nutrient (N, ambient concentration, enhanced
603 nutrients) on the cover of canopy algae, barren, and mat-forming algae among
604 experimental Boulders (B). * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$

605

Source of variation	Canopy algae			Barren		Mat-forming algae	
	df	MS	<i>F</i>	MS	<i>F</i>	MS	<i>F</i>
Habitat (H)	1	1939.5	3.17	6927.5	<0.01**	1536.0	2.07
Disturbance (D)	3	17622.6	28.80***	2195.8	0.03*	8089.8	10.91***
Nutrient (N)	1	2375.1	3.88	239.1	0.53	4121.3	5.56*
H x D	3	2284.5	3.73*	1976.6	0.04*	742.8	1.00
H x N	1	11.7	0.02	214.5	0.55	326.3	0.44
D x N	3	2630.7	4.30*	630.1	0.39	3385.6	4.57*
H x D x N	3	2582.3	4.22*	637.4	0.38	764.0	1.03
Boulder (H x D x N)	16	611.9	0.10	590.0	<0.01***	741.4	2.42**
Residual	64	386.6		169.6		305.7	

606

607 **Legend to Figures:**

608 **Figure 1.** Schematic description of the experimental design. At the beginning of the
609 study, 16 boulders totally covered by *Cystoseira* and 16 boulders partially covered by
610 *Cystoseira* were selected at the study site. One 1.5 x 0.5 m area was selected inside the
611 macroalgal forest (level 1 of the factor Habitat) on each of the boulders totally covered
612 by *Cystoseira* or at the boundary between the macroalgal forest and the barren (level 2
613 of the factor Habitat) on each of the boulders only partially covered by macroalgal
614 forests. Two boulders of each type of Habitat were then assigned to each combination of
615 Disturbance intensity (Control = macroalgal canopy left untouched; 30% CR = 30%
616 canopy removal; 70% CR = 70% canopy removal; 100% CR = 100% canopy removal)
617 and Nutrients (Ambient versus Enhanced). Three replicate 20x20 cm plots were
618 marked in each with epoxy putty. Eight nutrient releasing bags were fixed among plots
619 in each area.

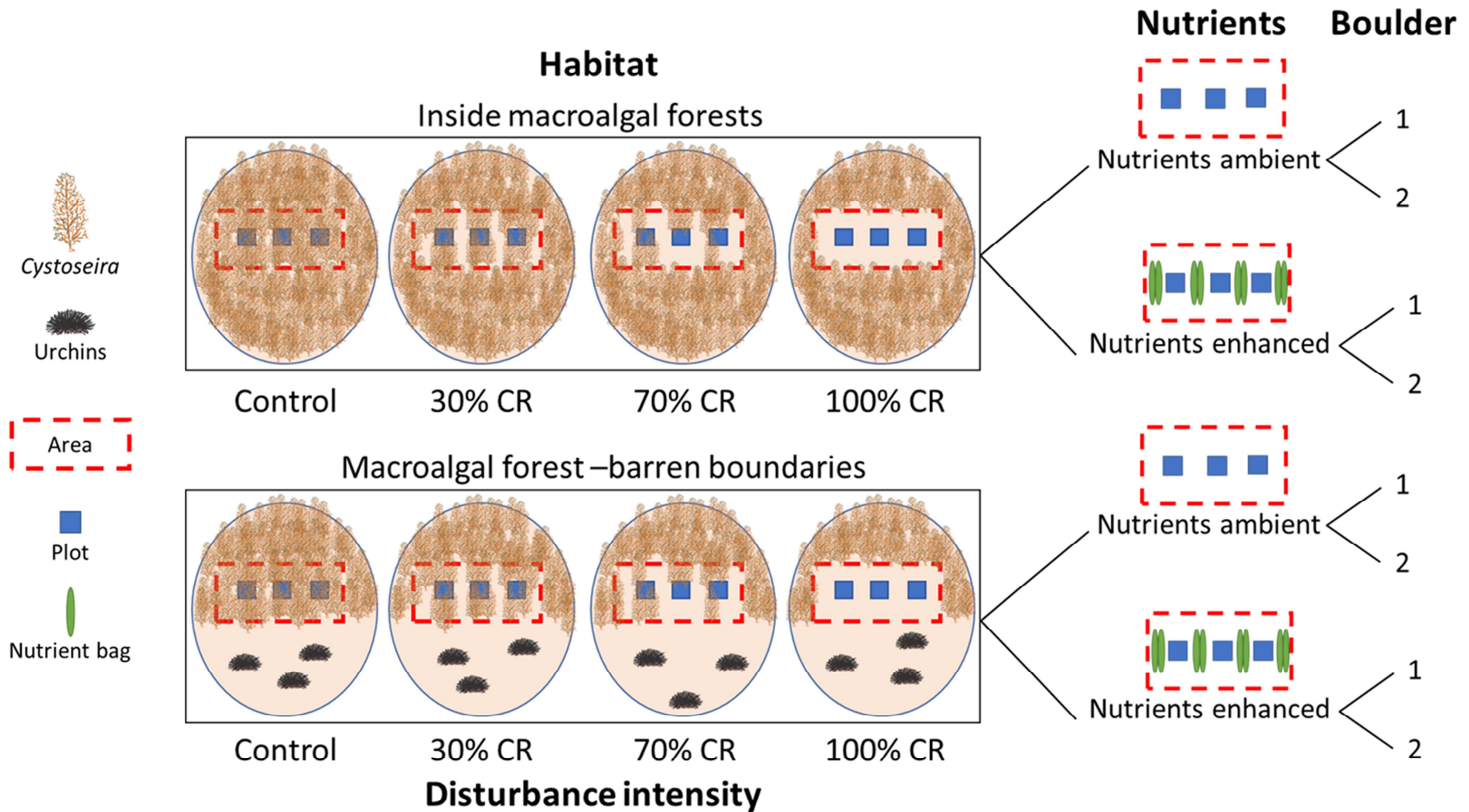
620 **Figure 2.** nMDS ordination plots comparing assemblages exposed to different
621 disturbance intensities (red = 0% canopy removal, yellow = 30% canopy removal, green
622 = 70% canopy removal, blue = 100% canopy removal) in plots embedded within
623 different habitat. Different symbols characterize habitats (square = inside the
624 macroalgal forest, circle = boundary between the macroalgal forest and the barren).
625 Stress = 0.12.

626 **Figure 3.** Percentage cover (mean + SE, $n = 12$) of canopy of *Cystoseira brachycarpa* in
627 plots characterized by different habitat (left = inside the macroalgal forest, right =
628 boundary between the macroalgal forest and the barren), disturbance intensity (0%,
629 30%, 70%, 100% canopy removal) and nutrient loads (black bars = ambient
630 concentration, grey bars = enhanced nutrients). Letters above the columns indicate the

631 outcome of SNK (student-Newman-Keuls) tests, with significant differences at $P < 0.05$.

632 **Figure 4.** Percentage cover (mean + SE, $n = 12$) of mat-forming algae in plots
633 characterized by different disturbance intensity (0%, 30%, 70%, 100% canopy
634 removal) and nutrient loads (black bars = ambient concentration, grey bars = enhanced
635 nutrients). Letters above the columns indicate the outcome of SNK (student-Newman-
636 Keuls) tests, with significant differences at $P < 0.05$.

637 **Figure 5.** Percentage cover (mean + SE, $n = 12$) of barren in plots characterized by
638 different habitat (white bars = inside the macroalgal forest, grey bars = boundary
639 between the macroalgal forest and the barren) and disturbance intensity (0%, 30%,
640 70%, 100% canopy removal). Letters above the columns indicate the outcome of SNK
641 (student-Newman-Keuls) tests, with significant differences at $P < 0.05$

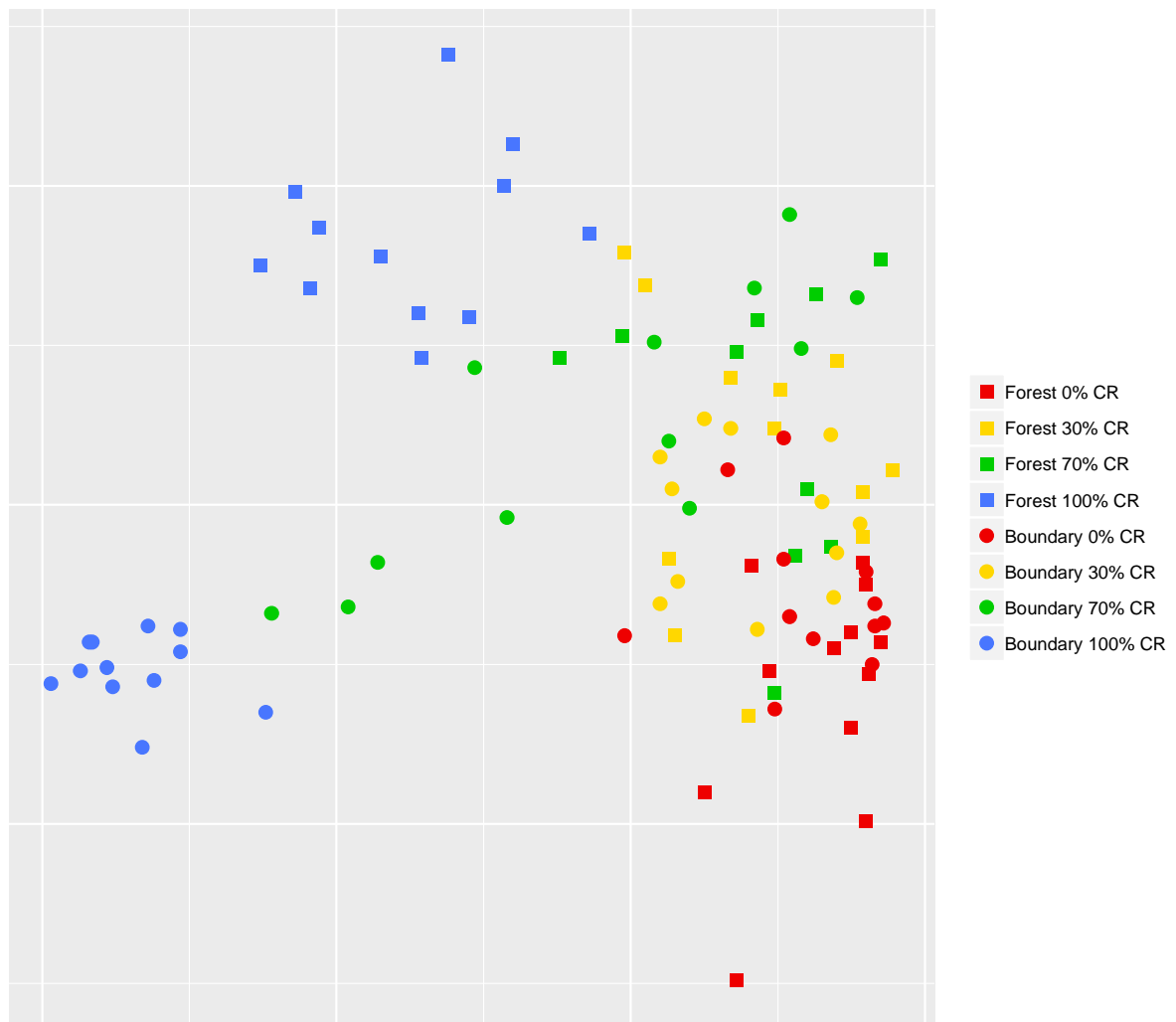


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643

Figure 1.

644

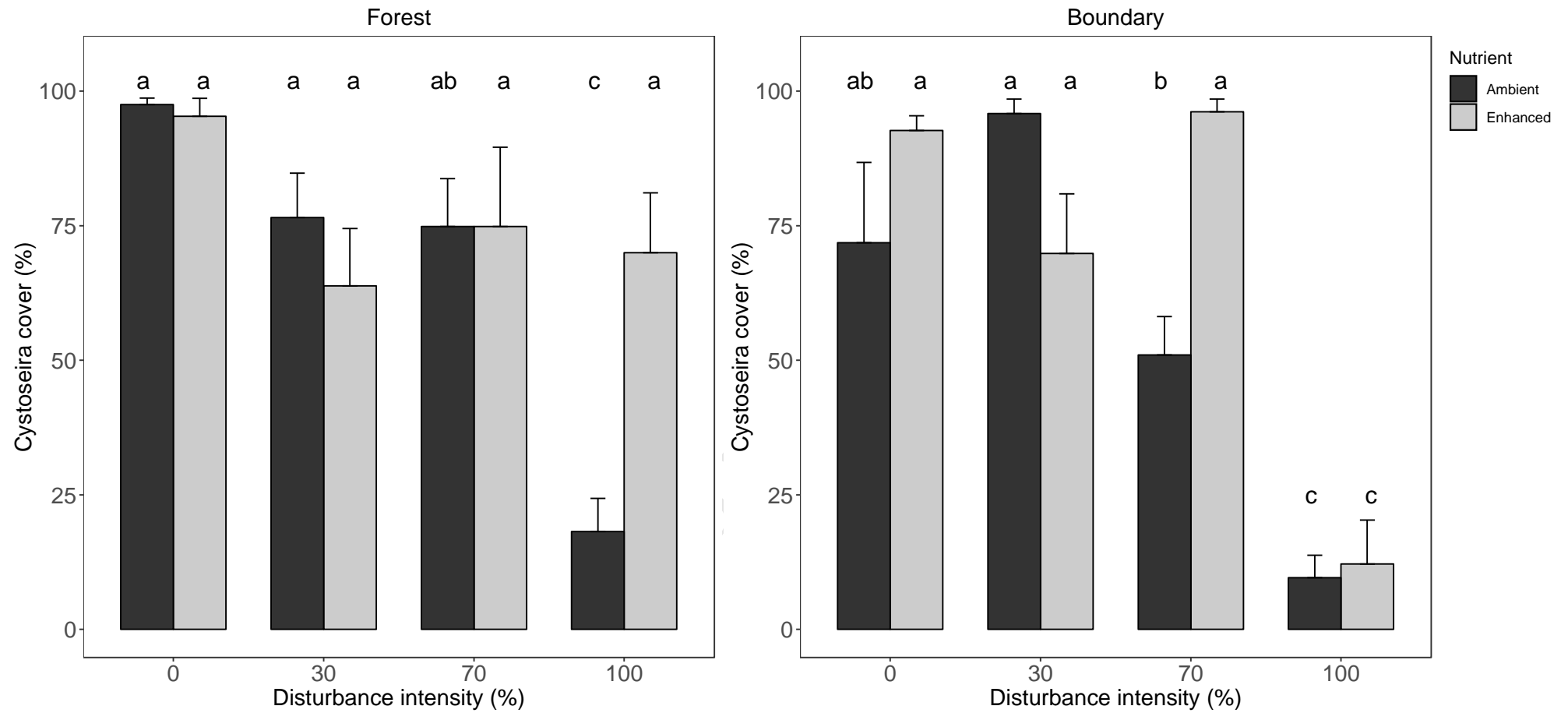


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Figure 2.



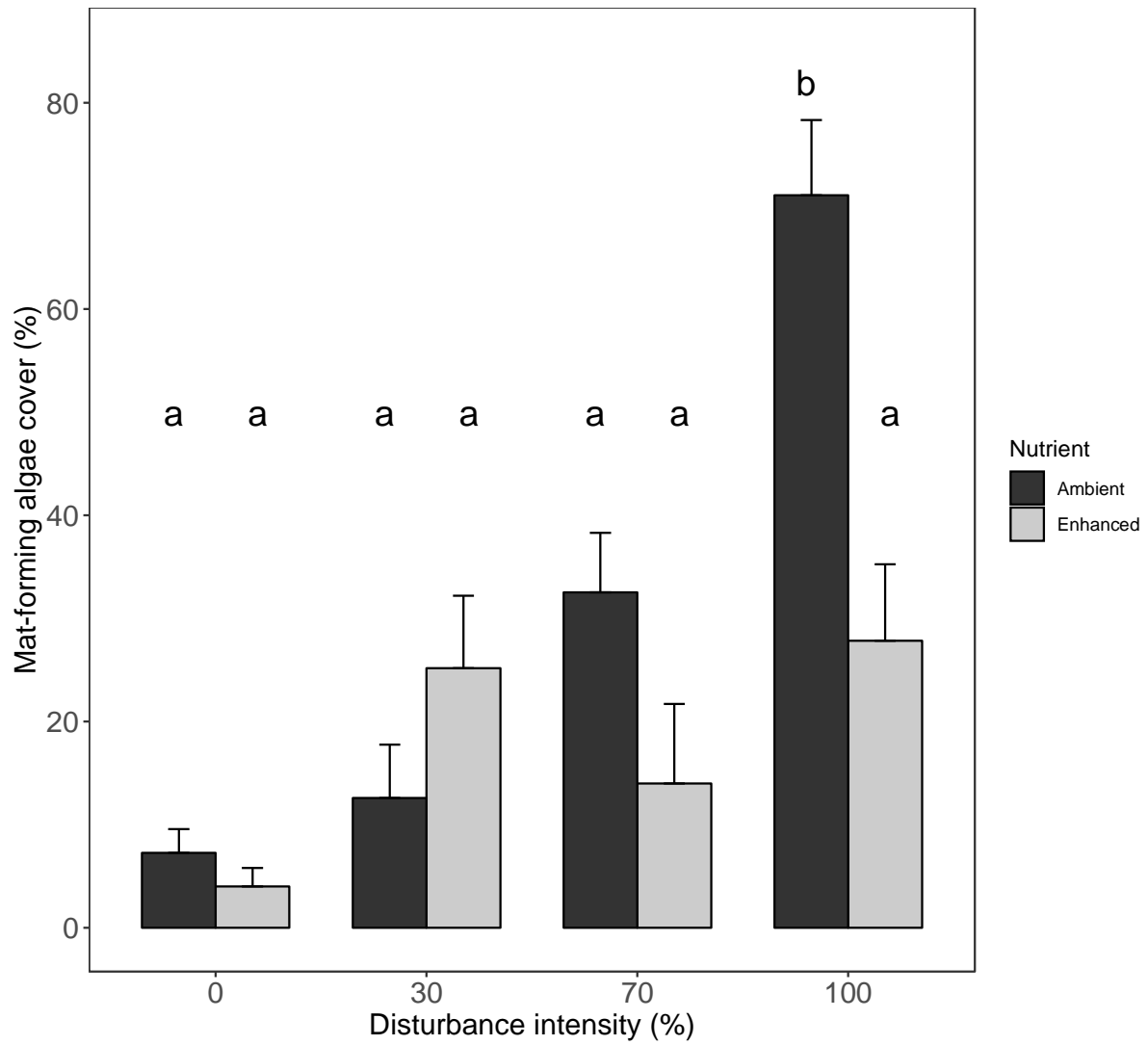
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Figure 3.

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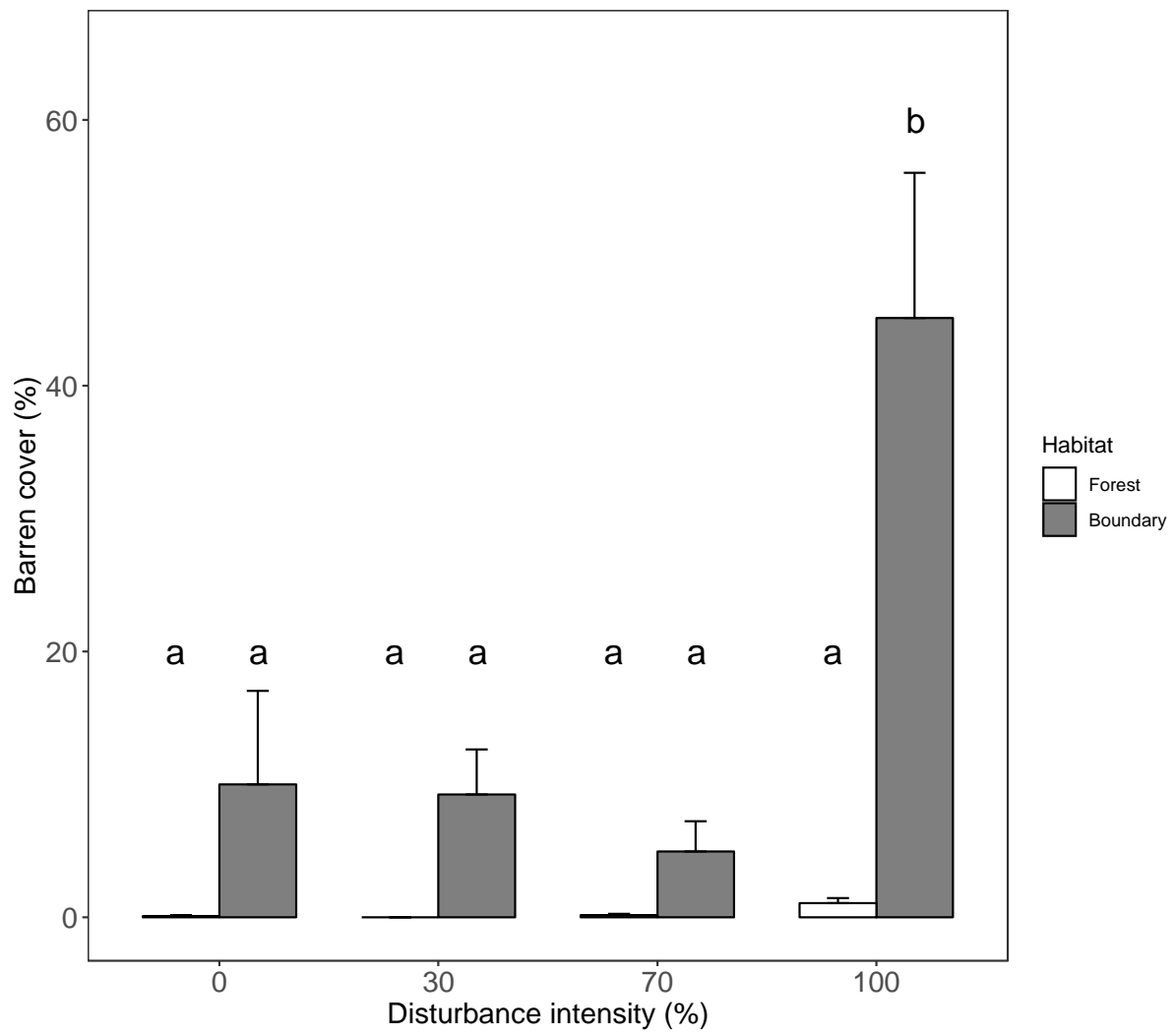
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Figure 4.

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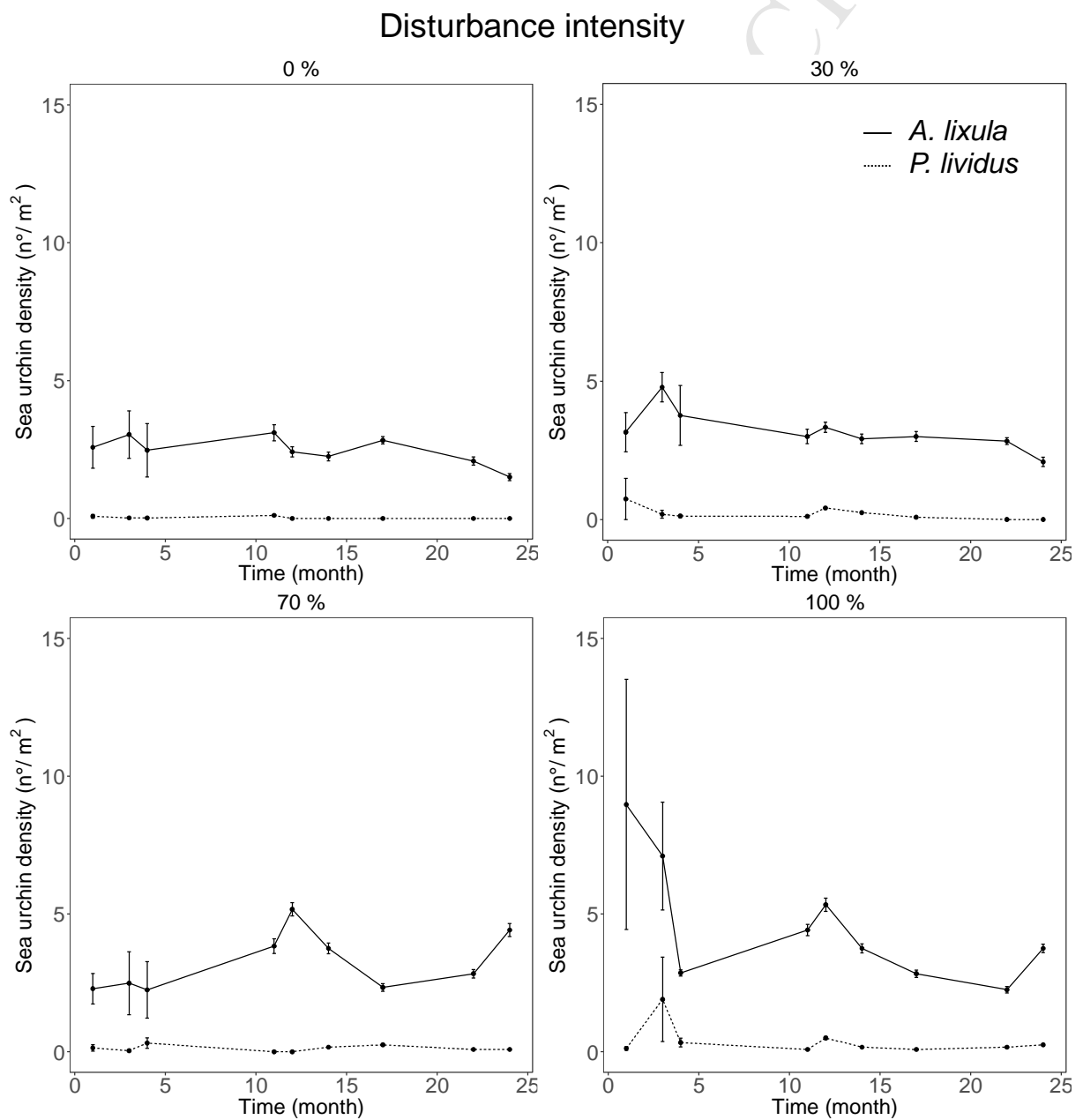
658

Figure 5.

659 **Appendix A.**

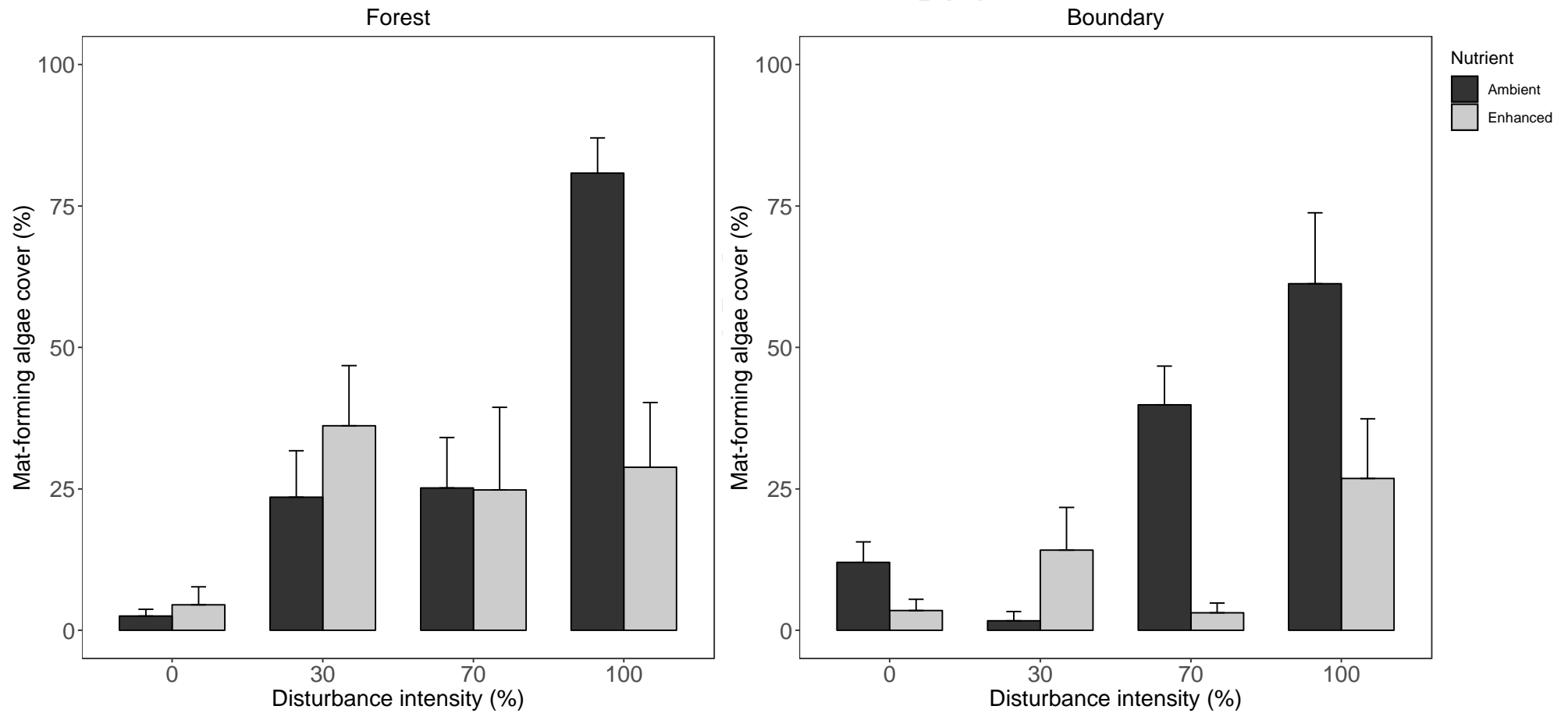
660 **Figure A.1** Density of the two species of sea urchins (*Arbacia lixula* and *Paracentrotus*
 661 *lividus*) estimated at different sampling times in 12 50x50 m plots, haphazardly placed
 662 in barren grounds at the boundary with *Cystoseira* forests on boulders characterized by
 663 the presence of both habitats. No sea urchins were found on boulders totally covered by
 664 *Cystoseira* forests.

665



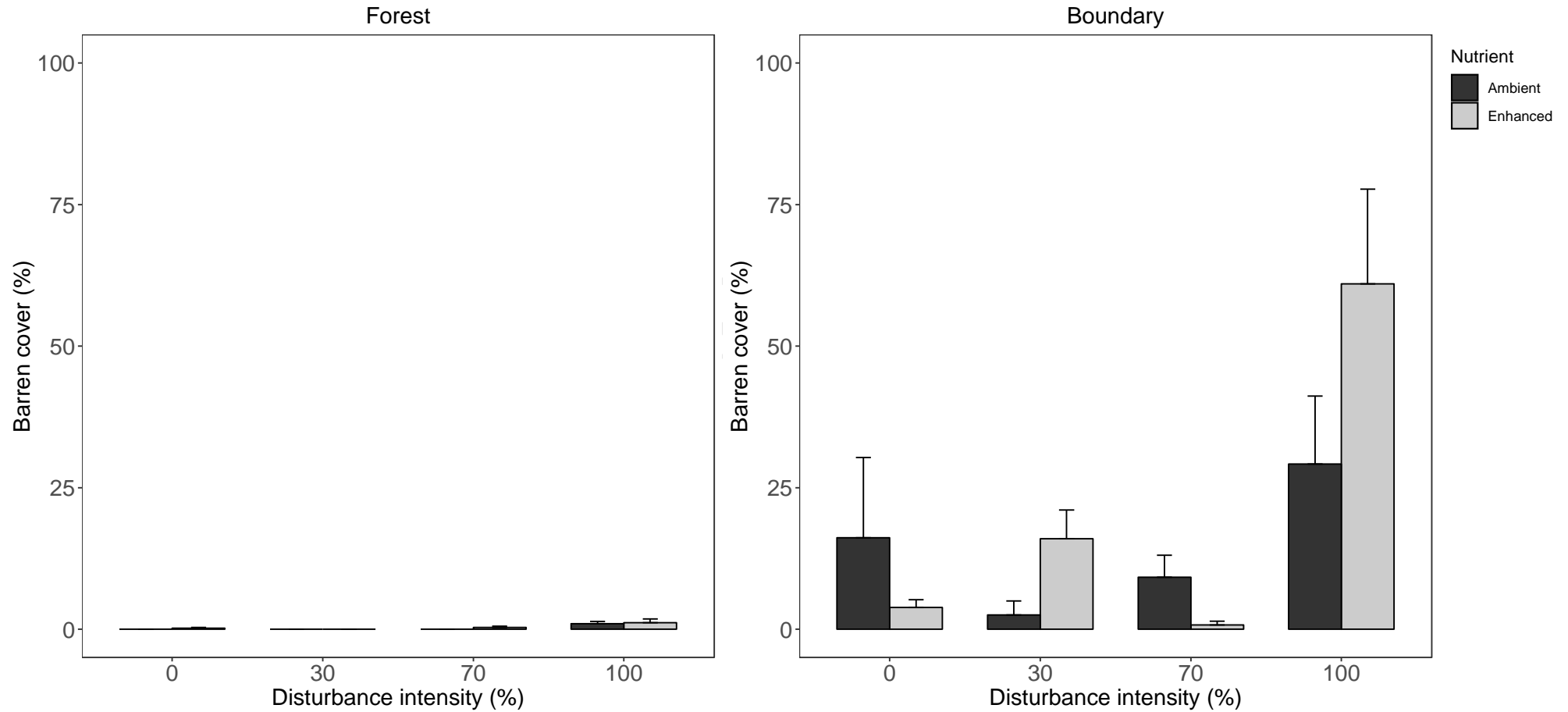
666

667 **Figure A.2** Percentage cover (mean + SE, $n = 12$) of mat-forming algae in plots characterized by different habitat (left = inside the
668 macroalgal forest, right = boundary between forest and barren), disturbance intensity (0%, 30%, 70%, 100% canopy removal) and
669 nutrient loads (black bars = ambient concentration, grey bars = enhanced nutrients).



670

671 **Figure A.3** Percentage cover (mean + SE, $n = 12$) of barren in plots characterized by different habitat (left = inside the macroalgal forest,
672 right = boundary between forest and barren), disturbance intensity (0%, 30%, 70%, 100% canopy removal) and nutrient loads (black
673 bars = ambient concentration, grey bars = enhanced nutrients).



674
675

676 **Appendix B.**

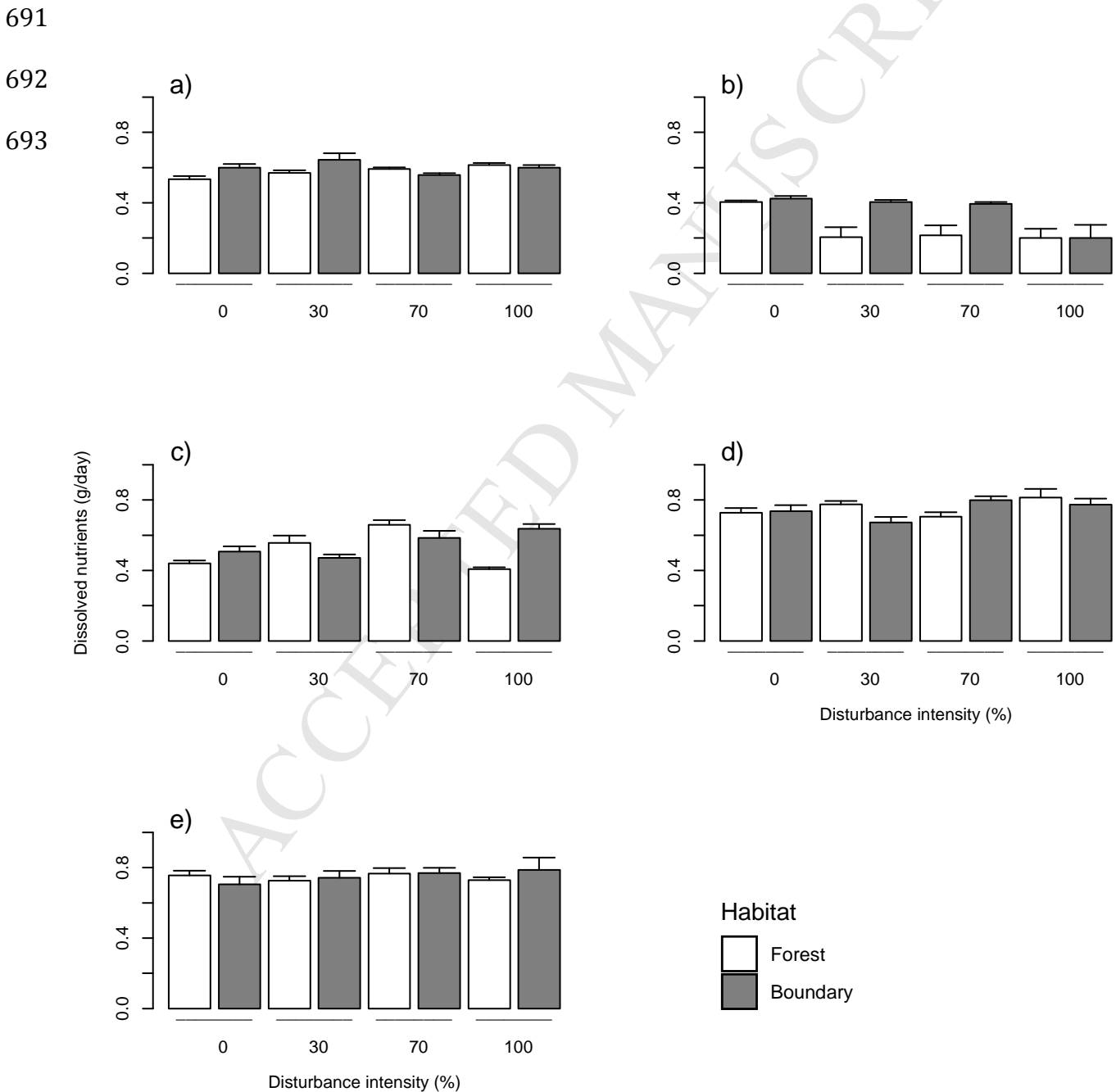
677

678 **Table B.1** Analyses of variance (ANOVA) comparing the dry weight of dissolved
 679 fertilizer pellets after retrieval of bags from areas characterized by different Habitat (H,
 680 inside the macroalgal forest, at the boundary between the macroalgal forest and the
 681 barren) and Disturbance Intensity (D, 0%, 30%, 70%, 100% canopy removal) at
 682 different times since the beginning of the experiment. * $P < 0.05$, ** $P < 0.01$, *** $P <$
 683 0.001

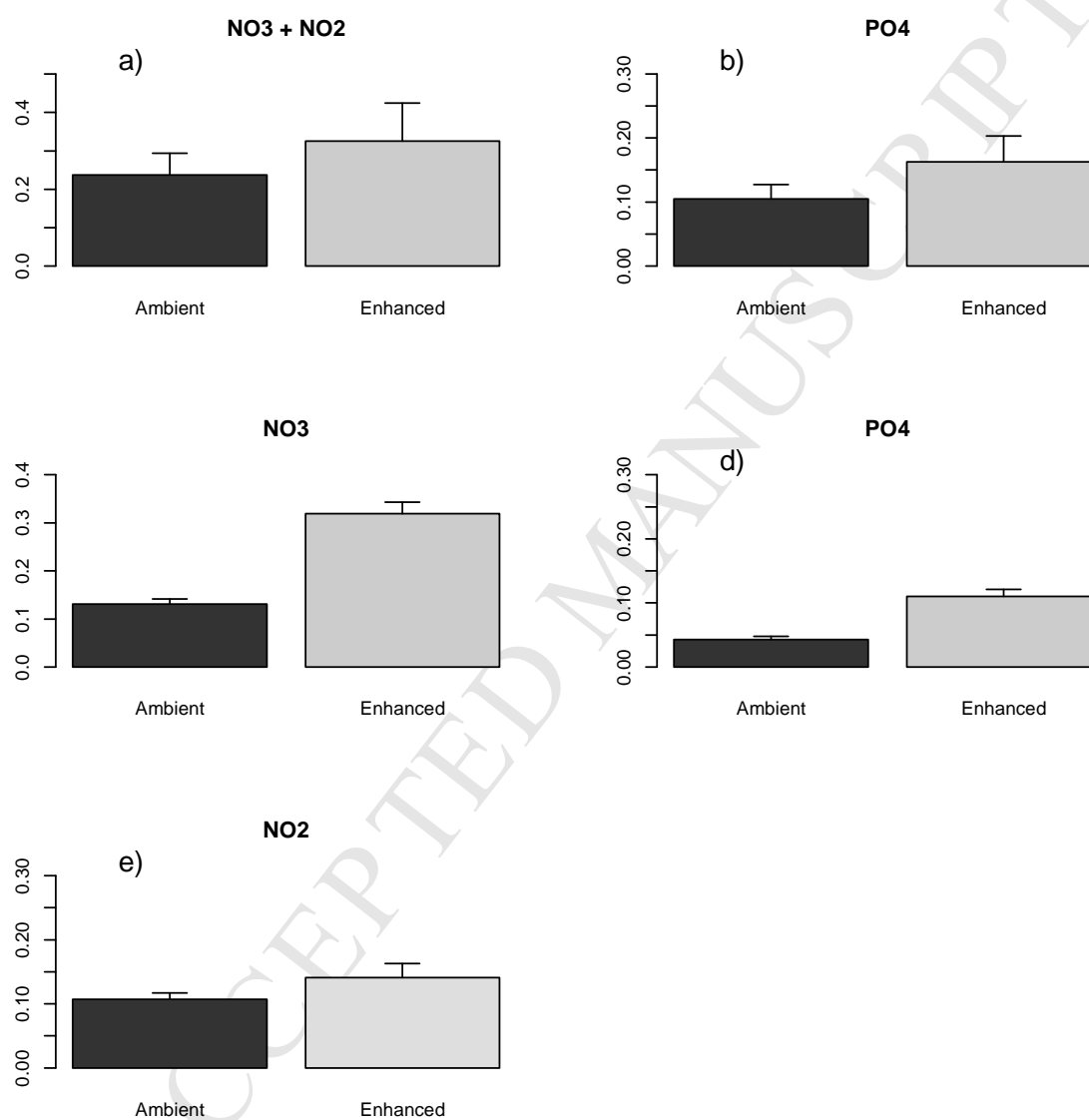
		4 months		9 months		12 months	
Source of variation	df	MS	<i>F</i>	MS	<i>F</i>	MS	<i>F</i>
Habitat (H)	1	0.0022	1.17	0.0364	0.85	0.0068	0.64
Disturbance (D)	3	0.0019	0.99	0.0319	0.75	0.0157	1.47
H x D	3	0.0029	1.57	0.0117	0.27	0.0209	1.97
Residual	8	0.0019		0.0428		0.0107	
Cochran's test		ns		ns		ns	
		14 months		17 months			
Source of variation	df	MS	<i>F</i>	MS	<i>F</i>		
Habitat (H)	1	0.0001	0.00	0.0004	0.07		
Disturbance (D)	3	0.0025	0.59	0.0023	0.41		
H x D	3	0.0062	1.47	0.0043	0.77		
Residual	8	0.0042		0.0056			
Cochran's test		ns		ns			

684

685 **Figure B.1** Histograms showing the dry weight of dissolved fertilizer pellets after
 686 retrieval of bags (grams/day) from areas characterized by different Habitat (white bars
 687 = inside the macroalgal forest grey bars = boundary between forest and barren) and
 688 Disturbance Intensity (0%, 30%, 70%, 100% canopy removal). Dissolved fertilizer
 689 pellets were evaluated after a) 4 months, b) 9 months, c) 12 months, d) 14 months and
 690 e) 17 months since the beginning of the experiment.



694 **Figure B.2** Histograms showing the concentration of nutrients in seawater ($\mu\text{mol/l}$), at
695 two random times during the experiment. Time 1: a (nitrate and nitrite), b (phosphate);
696 Time 2: c (nitrate), d (phosphate), e (nitrite). For each sampling time, at ambient
697 concentration: $n = 16$ (black bars); in nutrient enriched boulders: $n = 30$ (grey bars).



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Highlights

- We tested the effects of nutrient enhancement and herbivory on canopy recovery
- *C. brachycarpa* was highly resilient and recovered to any partial mechanical damage
- Nutrient addition speeded up canopy recovery rather than favoring mat-forming algae
- At the forest margin, total canopy removal promoted the expansion of urchin barrens