



Minerva Access is the Institutional Repository of The University of Melbourne

Author/s:

Roussel, S;Poitevin, P;Day, R;Le Grand, F;Stiger-Pouvreau, V;Leblanc, C;Huchette, S

Title:

Haliotis tuberculata, a generalist marine herbivore that prefers a mixed diet, but with consistent individual foraging activity

Date:

2020-07-01

Citation:

Roussel, S., Poitevin, P., Day, R., Le Grand, F., Stiger-Pouvreau, V., Leblanc, C. & Huchette, S. (2020). *Haliotis tuberculata*, a generalist marine herbivore that prefers a mixed diet, but with consistent individual foraging activity. *Ethology*, 126 (7), pp.716-726. <https://doi.org/10.1111/eth.13020>.

Persistent Link:

<https://hdl.handle.net/11343/275517>

Author Manuscript

This is the author manuscript accepted for publication and has undergone full peer review but has not been through the copyediting, typesetting, pagination and proofreading process, which may lead to differences between this version and the [Version of Record](#). Please cite this article as [doi: 10.1111/eth.13020](https://doi.org/10.1111/eth.13020)

This article is protected by copyright. All rights reserved

1
2 DR SABINE ROUSSEL (Orcid ID : 0000-0001-9275-0818)

3
4
5 Article type : Research Paper

6
7
8 *Haliotis tuberculata*, a generalist marine herbivore that prefers a mixed diet, but with consistent
9 individual foraging activity

10
11 Running title: consistency of behaviour in a marine herbivore

12
13 Word count of the text (excluding abstract and references, title and authors) : 5183 words

14
15 Sabine ROUSSEL^{a,*}, Pierre POITEVIN^{a,b}, Robert DAY^{a,c}, Fabienne LE GRAND^a, Valérie
16 STIGER-POUVREAU^a, Catherine LEBLANC^{d,e}, Sylvain HUCHETTE^b

17
18
19 ^a Univ Brest, CNRS, IRD, Ifremer, LEMAR, F-29280 Plouzane, France

20 ^b France Haliotis, Kerazan Lilia, 29 880 Plouguerneau, France

21 ^c School of Biological Sciences, University of Melbourne, Parkville, Victoria, Australia 3010

22 ^d Sorbonne Universités, UPMC Univ Paris 06, UMR 8227, Integrative Biology of Marine
23 Models, Station Biologique de Roscoff, CS 90074, F-29688 Roscoff CEDEX, France

24 ^e CNRS, UMR 8227, Integrative Biology of Marine Models, Station Biologique de Roscoff, CS
25 90074, F-29688 Roscoff CEDEX, France

26
27 ^{*}Corresponding author: Sabine Roussel,

28 Univ Brest, CNRS, IRD, Ifremer, LEMAR, Rue Dumont d'Urville, F-29280 Plouzane, France

29 Tel.: +33 (0) 2 98 01 70 43

30 E-mail address: sabine.roussel@univ-brest.fr

31

32 CONFLICT OF INTEREST: no conflict of interest.

33

34 ACKNOWLEDGEMENTS

35 This work was supported by the IDEALG program, a long term investment grant from the

36 Agence Nationale de la Recherche (ANR) (grant number: ANR-10-BTBR-04).

37

38 ABSTRACT

39 While population foraging behaviour of herbivores has been extensively studied, individual

40 choice is still poorly understood. Very few studies have focused on the individual consistency of

41 foraging behaviour in marine herbivores. Because marine ectotherms are strongly influenced by

42 their environment and because a mixed diet is appropriate for herbivores, we hypothesized that

43 *Haliotis tuberculata*, a large marine gastropod, would not exhibit significant individual

44 consistency in foraging activity and would display generalist food choices. To test these

45 hypotheses, the behaviour of 120 abalone was studied using a choice test of eight macroalgal

46 species over 3-weeks, with video recording 24 hours a day. In addition, primary components,

47 secondary metabolites and toughness of the eight algae were measured. At the population level,

48 food choice was mainly related to the protein composition and the toughness of the macroalgae.

49 We found that *H. tuberculata* is a generalist species feeding on a variety of algae ($IS = 0.64$), even

50 if 21% of the individuals can be considered to be specialists. However, in contrast to our

51 hypothesis, highly consistent between-individual variation was observed in foraging activity (ICC

52 = 0.81 for time spent feeding and ICC = 0.74 for number of feeding visits per day). The high
53 individual consistency of foraging activity has some ecological and evolutionary implications
54 currently not understood for this marine herbivore.

55 Keywords: intraclass correlation, proportion similarity index, food choice, marine herbivore,
56 foraging activity, mollusc, individual consistency

57

Author Manuscript

58 INTRODUCTION

59

60 Herbivores have to feed on items that are highly variable and often nutritionally much poorer
61 than their own body (Sternler & Hessen, 1994). For herbivores with nutritionally poor prey items,
62 foraging choices are complex, and a mixed diet is a good food choice strategy in terms of nutrition
63 (Day & Cook, 1995). It has been suggested that food choice strategies of herbivores depend on
64 four major components in plants: protein content, chemical defences (distasteful or toxic
65 substances), chemical feeding stimulants (perhaps related to essential nutrients), and physical
66 defences such as the toughness of plant tissue (Scheidel & Bruelheide, 1999). As plants contain
67 much less protein than animals, protein content plays an important role in determining the
68 preferences of herbivores (Mattson, 1980). Protein composition is highly variable between algal
69 species (Jung, Lim, Kim, & Park, 2013; Mai, Mercer, & Donlon, 1994; Mai, Mercer, & Donlon,
70 1995) and between seasons and harvest sites (Renaud & Luong-Van, 2006; Schmid, Guiheneuf, &
71 Stengel, 2014; Villares, Fernandez-Lema, & Lopez-Mosquera, 2013). Another important
72 component determining food choices in abalone is the chemical defences of macroalgae. In marine
73 gastropods, post-ingestive consequences of secondary metabolites have been evaluated in only a
74 few cases, showing that growth is reduced by secondary metabolites in sea hares (Pennings &
75 Carefoot, 1995) and abalone (Fleming, 1995b; Winter & Estes, 1992). The effect of a third
76 potential determinant of feeding behaviour on marine herbivores, algae toughness, has been quite
77 controversial. Some concluded this is of only minor importance (Shepherd & Steinberg, 1992)
78 while others have observed important effects of such structural defences (Winter & Estes, 1992).
79 However, neither the effectiveness of any single factor nor the interactions between them is yet
80 fully understood. In addition, even if some food preferences can be determined at a population
81 level, individual foraging strategies can nuance these preferences.

82

83 Consistency of foraging behaviour can be studied with different approaches, historically separate
84 but conceptually similar. One approach focuses on food resource use to determine if the
85 population and individuals can be defined as generalist or specialist (Mather, Leite, & Batista,
86 2012). The determination of individual specialisation is important because apparently generalized
87 species might be composed of individual specialists using only a small part of the population's
88 food niche (Bolnick et al., 2003). This variation within the population can be considered as
89 adaptive in itself (Valen, 1965) and can persist over time and across contexts (Toscano, Gownaris,
90 Heerhartz, & Monaco, 2016). Some tools have been developed which allow quantitative
91 assessment of within- and between-individual variation in resource use (Bolnick, Yang, Fordyce,
92 Davis, & Svanback, 2002). Because generalist behaviour might result in ingesting a more optimal
93 set of dietary components for an herbivore with low locomotion abilities, we hypothesized that
94 *Haliotis tuberculata* would display a generalist algal choice strategy at the population level with
95 non-specialized individual food choices.

96 A second approach to study foraging behaviour consists in studying individual behavioural
97 consistency and personality. Consistency of individual behavioural patterns across time and
98 between different contexts has been reported in taxa ranging from fish to mammals, with most of
99 the studies done on insects, and focusing on mate preference and courtship (Bell, Hankison, &
100 Laskowski, 2009). Foraging types can be an important factor in determining community dynamics,
101 with a spatial structuration of behavioural type (Griffen, Toscano, & Gatto, 2012). Individual
102 consistency can also be used as a rough estimate of the heritability of a trait (Dohm, 2002;
103 Falconer & Mackay, 1996). As suggested by the lower heritability of morphological traits for
104 ectotherms in comparison to endotherms (Mousseau & Roff, 1987), we expected the heritability
105 and thus indirectly the consistency of foraging activity would be low in abalone. To our

106 knowledge, only a few experiments have been performed on the consistency of foraging behaviour
107 in marine ectotherms, with most of them done on predators (Koteja, Carter, Swallow, & Garland,
108 2003; McHuron, Hazen, & Costa, 2018; Missoweit, Engels, & Sauer, 2007; Morgan, Hassall,
109 Redfern, Bevan, & Hamer, 2019; Patrick et al., 2014; Toscano & Griffen, 2014), but none of them
110 performed on marine herbivores.

111
112 To test these hypotheses, the foraging behaviour of *Haliotis tuberculata*, a large marine herbivore
113 common in European sublittoral environments, was studied with three objectives: 1) to understand
114 the food choices at a population level in relation to important components of potential prey algae
115 (primary, secondary metabolites and toughness) and diurnal rhythm 2) to evaluate the prevalence
116 of individual specialisation in the population and 3) to evaluate the consistency of foraging activity
117 at the individual level.

118

119 MATERIAL AND METHODS

120

121 Animal origin and maintenance

122

123 Behavioural experiments were carried out at the France *Haliotis* organic abalone farm facilities
124 (48°36'46N; 4°33'30W) in Plouguerneau, France, between December 2011 and March 2012.
125 Chemical analyses of macroalgae were performed in the Marine Environmental Sciences
126 Laboratory (LEMAR, Brest, France) for phenolic compounds, proteins, lipids and carbohydrates;
127 in the University of Western Brittany (UBO, Brest, France) for nuclear Magnetic Resonance
128 HRMAS analyses (to detect phenolic compounds).

129

130 Adult *Haliotis tuberculata* (65-70 mm total shell length, 4 years old) were reared from systematic
131 mating between wild and farmed broodstocks, mainly to avoid inbreeding. From the age of 1 year
132 until the start of the experiment, they were reared in a cage-structure placed in the subtidal zone.
133 The abalone were randomly harvested from the structure without removing them from the black
134 plastic oyster seed collectors used to provide crevices for them (diameter: 140 mm). They were
135 transported to the land-based laboratory in a 50 L seawater tank. Once in the laboratory, eight
136 abalone were randomly assigned to each experimental tank. A reflective tag was gently glued to
137 each individual with cyanoacrylate glue, without removing the animal from the collectors, to
138 minimize any stress. The tags were designed to allow us to recognize each individual abalone in a
139 tank. Abalone were allowed to acclimate to the experimental tanks for 2 weeks before the start of
140 the experiment.

141

142 Materials

143

144 The six experimental tanks were grey, flat, square tanks with rounded corners, made of
145 epoxy painted fibreglass (1.10 x 1.10 x 0.20 m, water volume = 100 L), with rough plastic strips
146 on the inside edges to prevent abalone escaping. Each tank held 75 L.h⁻¹ of 3 µm mechanically
147 filtered and temperature regulated seawater (12.0 ± 0.5 °C), and was provided with an aeration
148 system. Eight pairs of plastic oyster seed collectors were uniformly distributed along the edges of
149 each tank to be used as hiding places. Eight plastic feeders (7 x 7 x 3 cm) were fixed onto the floor
150 at the center, at equal distances from the eight hiding places, to ensure equal access to each algal
151 species (Figure 1). Five of the tanks contained abalone. The last tank was used as a control to
152 follow natural degradation of the algae. The light:dark regime was 10:14 h (Light: 0830 to 1830
153 hours; Dark: 1830 to 0830 hours). To avoid stressful conditions during light changes, a transition

154 of light level was programmed over 30 minutes during dawn (0800 to 0830 hours) and dusk (1830
155 to 1900 hours) using a dimmer (Gold Star, Besser Elektronik, Italia). Tanks were cleaned twice a
156 week using a hose and water filters were changed every day. All tanks were continuously
157 videotaped by five digital cameras (TS-WD6001HPSC, Sygonix GmbH, Germany) linked to a 24h
158 recording device (TVVR 40021, Abus, Germany).

159

160

161 Experimental procedure

162

163 Food choices of abalone were studied using 8 macroalgal species: *Asparagopsis armata*,
164 *Palmaria palmata* (Rodophyta), *Enteromorpha intestinalis*, *Ulva lactuca* (Chlorophyta),
165 *Saccharina latissima*, *Saccorhiza polyschides*, *Laminaria digitata* and stipes of *Laminaria*
166 *hyperborea* (Phaeophyta). Fresh algae were collected at Pors Grac'h beach (48°37'58 N; 4°32'57
167 W), Plouguerneau, France. The macroalga provided during the acclimation period was *Palmaria*
168 *palmata*. From one to four years of age during commercial growth conditions, the eight algae had
169 been provided to these abalone on a regular basis by France Haliotis, with *Palmaria palmata* and
170 *Laminaria digitata* being the most important in terms of quantity. After the acclimation period, *P.*
171 *palmata* was removed 3 days prior to the start of the experiment.

172 To determine wet biomass intakes of abalone, the eight macroalgal species were weighed after
173 drying them gently with paper towels and each alga was randomly allocated to one feeder position
174 in each tank. The quantities of algae provided were chosen to provide ad libitum access (at least
175 half of the initial quantity remaining at the end of the period). After 4 days, the remaining algae
176 were removed from each feeder, dried in the same way and wet weighed. Abalone were then
177 starved for 3 days after this food choice trial and the same procedure was repeated during two

178 more weeks to study the food choices and foraging activity over a 3-week period. However, in
179 each tank the algal species were allocated to the same feeder positions during the two subsequent
180 trials. The natural change in weight of algae was measured in the control tank. The quantity of
181 algae ingested by all the abalone in a tank was calculated by removing the change of weight in the
182 control tank from the reduction in weight of the algae in the tanks containing abalone. The 3-week
183 experiment was replicated three times (in total, $n = 15$ tanks with abalone, $n = 3$ tanks with only
184 algae).

185

186 Food choices and foraging activity analysis

187

188 Experimental tanks were filmed 4 days per week from 1830 hours on Monday to 0900 hours on
189 Friday during a 3-week period. The videos were analysed at 16x speed, slowed to 4x at the
190 beginning and the end of each food intake period. The researchers were blind to the alga used in
191 the feeders during video analysis.

192

193 Mean feeding bout duration was calculated by subtracting the start feeding time from the stop
194 feeding time for each visit to the feeder, and averaged per day. Total time spent feeding was the
195 sum of all feeding durations per day. The number of feedings was the number of feeder visits done
196 per day. Individual foraging activity was followed using individual reflective tags. These
197 behavioural variables were used for the ICC analysis (see Statistical Analysis for details). For algal
198 preference, variables were calculated per alga for each tank, using the three weeks of individual
199 foraging behaviour. The number of times the abalone chose each of the eight algae during the 3-
200 weeks was recorded, to calculate the proportional similarity index as well as the prevalence of
201 individual specialisation in the population (see Statistical Analysis for explanations).

202

203 Algal chemical analysis

204

205 Sample collection

206 Samples from all algal species were collected on the 6th of February and the 5th of March. These
207 samples consisted of the same parts of the algal thalli as were available to the abalone during the
208 food choice experiment. In total, six samples (three per collection day) of 200 g, representative of
209 the algae distributed, were vacuum packed, immediately frozen and stored in a freezer at -20°C
210 before lyophilisation at -55°C for 96h. Dried algae were then crushed, to be used in all the
211 following tests except fatty acid analysis. For fatty acid analysis, samples of 2 g were frozen with
212 liquid nitrogen and stored in a freezer at -80°C before crushing. Dry matter content of each species
213 was determined by weighing the samples before and after freeze-drying.

214

215 Algal toughness

216 Algae were ranked from 1 to 8 according to their toughness (Steneck & Watling, 1982) and
217 visual evaluation of their thickness : 1 being the thinnest and softest alga, and 8 the thickest and
218 toughest alga (1 : *Asparagopsis armata* and *Enteromorpha intestinalis*, 3 : *Ulva lactuca*, 4 :
219 *Palmaria palmata*, 5 : *Saccharina latissima*, 6 : *Laminaria digitata*, 7 : *Saccorhyza polyschides*, 8
220 : stipes of *Laminaria hyperborea*).

221

222 Total protein content

223 Total protein was calculated from total nitrogen content of the samples by Kjeldahl's method, as
224 modified by Connan (2004). The homogenized samples (150 mg) were digested by boiling for 40
225 min in concentrated sulfuric acid (H₂SO₄), with a catalyst (K₂SO₄+CuSO₄+Se). The end product is

226 an ammonium solution. Excess base (NaOH) was added to the product to convert NH_4^+ to NH_3 .
227 After distillation in a Büchi (Büchi 323, Büchi Labortechnik, Switzerland), dissolved NH_3 was
228 recovered in 25 ml boric acid (H_3BO_3) solution containing Tashiro's indicator. Titration was
229 performed with sulfuric acid at 0.01M. Analysis of a blank was run in parallel to the analysis of
230 samples. The protein content (%) of the samples was calculated according to the following
231 formula: $((V_s - V_b) * 28 * F) / W$

232 Where:

233 V_b = mL titrant for the blank

234 V_s = mL titrant for the sample

235 W = Weight of sample in milligrams

236 F = Factor used to convert nitrogen percentage in a sample to percent protein, its value was 6.25

237

238

239 Total carbohydrate content

240 Total carbohydrate content (%) was measured by the spectrophotometric orcinol-sulfuric method,
241 using glucose as a standard. Algal powder (5 mg) was diluted in distilled water (5 mL) and
242 vortexed before sampling. Samples (200 μL , 3 replicates) were introduced into glass test tubes,
243 then 400 μL of orcinol-sulfuric reagent (1.5%) and 3 mL H_2SO_4 (60%) were added. The test tubes
244 were placed in a shaking water bath for 20 min at 80°C. The reaction was stopped by placing the
245 test tubes in an ice bath for 2 min and storing them in a dark room for 45 min. The absorbance of
246 all samples was measured at 510 nm using a spectrophotometer (Perkin Elmer).

247

248 Total lipid content and fatty acid analysis

249 Lipid extraction was conducted on 150 to 200 mg of algal powder. Aliquots were transferred in

250 glass test tubes previously heated for 6 h at 450 °C and containing 6 mL of a chloroform/methanol
251 mixture (2/1, v/v), in accordance with the Folch et al. (1957) method. The method is described in
252 detail in Roussel et al. (2019). Quantitative fatty acid spectra obtained by gas chromatography
253 were used to calculate the molar content of each fatty acid in the samples. Depending on the
254 number of double bonds they display, fatty acids (FA) were classified into three groups: saturated
255 FA (SFA, no double bond), monounsaturated (MUFA, only one double bond) and polyunsaturated
256 (PUFA, two or more double bonds). FA could also be differentiated by the position of the first
257 double bond from the terminal carbon: n-3 (omega 3) or n-6 (omega 6). The results used in the
258 PCA were: total lipid content (μg FA / mg dry matter), n-3/n-6 ratio, MUFA content (% of total
259 FA), PUFA content (% of total FA) and SFA content (% of total FA).

260

261 Phenolic compounds analysis

262 NMR HRMAS analysis was performed first to detect the presence of phenolic compounds.
263 Phenolic compound analysis was conducted only on three algal species where phenolic
264 compounds were detected: *Asparagopsis armata* (Rhodophyta), *Enteromorpha intestinalis*
265 (Chlorophyta) and *Saccharina latissima* (Phaeophyta). Extraction of phenolic compounds was
266 conducted in triplicate for each algal species by putting algal powder (400 mg) in an ethanol/water
267 solution (40 mL) for 3 h at 40°C in a shaking water bath. The amount of total phenolics in the 3
268 species extracts was determined using the Folin-Ciocalteu reagent following the method of
269 Slinkard and Singleton (1977) using gallic acid as a standard for *Asparagopsis armata* and
270 *Enteromorpha intestinalis* and phloroglucinol as a standard for *Saccharina latissima*. Samples (20
271 μL , 3 replicates for each extract) were introduced into microtiter plate wells, and then 10 μL of
272 Folin-Ciocalteu reagent, 40 μL NaCO_3 (7.5%), and 130 μL H_2O were added. The absorbance of all
273 samples was measured at 765 nm using a spectrophotometer (Labsystems multiskan MS) after

274 incubating at 70°C for 10 min. Results were expressed as milligrams gallic acid equivalent (GAE)
275 / g fresh algae sample for *Enteromorpha intestinalis* and *Asparagopsis armata* or milligrams
276 phloroglucinol equivalent (PE) / g fresh algae sample for *Saccharina latissima* per gram of fresh
277 weight.

278

279 Statistical Analysis

280

281 To study the general foraging activity of the population, the total number of feeder visits was
282 calculated over all the abalone in each tank during the day and night periods over the 3 weeks (N =
283 15 tanks in total). To compare the number of day versus night feeding visits, a Wilcoxon signed
284 rank test for paired data was used because the conditions to use a parametric test were not fulfilled.
285 In addition, the total number of feeding bouts initiated in each 2-hour period of the 24-hour cycle
286 was calculated per tank.

287

288 To study the population food choices, a Friedman two-way analysis of variance by ranks was
289 used. Behavioural variables were calculated per alga for each tank over the three weeks. This non-
290 parametric analysis was proposed by Lockwood (1998) and Siegel and Castellan (1988) for
291 multiple-choice feeding preference experiments. A rank of 1 corresponded to the algae eaten in
292 largest quantity while a rank of 8 corresponded to the algae eaten in the smallest quantity (Table
293 1). Post-hoc comparisons were performed with the method proposed by Siegel and Castellan
294 (1988).

295

296 Spearman correlations with adjusted p-value (Holm's correction) were performed to study the
297 relationships between the foraging behaviour per algae for each tank over the three weeks and the

298 chemical variables.

299

300 For each individual, the consistencies of their mean feeding bout duration, their total time spent
301 feeding per day and their number of feeding bouts per day, were studied over the three weeks. The
302 four daily values ($k = 4$ days per week) were averaged per week and per individual. Prior to
303 calculating repeatability statistics, the data were log-transformed when non-normality of residuals
304 was detected (Shapiro's test) or when variances were found to be significantly different (Levene's
305 test) to satisfy the assumption that there is a common population (residual) variance (Biro &
306 Stamps, 2015). Thereafter, ICC estimates for the transformed foraging variables and their 95%
307 confidence intervals were calculated using the psych package in R (Revelle, 2018), based on a
308 mean-rating ($k = 4$), consistency-agreement, 2-way mixed-effects model (weeks are considered
309 fixed effects but individuals are treated as random effects) with 120 individuals.

310

311 A proportional similarity index (PS) was calculated for each individual as well as the prevalence
312 of individual specialisation in the population (IS) using the RInSp R package (Zaccarelli,
313 Mancinelli, & Bolnick, 2013) to study whether abalone consistently act as specialists or generalists
314 with respect to algal choice. Abalone which did not feed at least four times during the 3-week
315 experiment (18 abalone out of 120) were not used for this individual specialisation analysis.

316

317 Ethical Note

318

319 Manipulation of individuals involved food limitation and diet modification. At the end of the
320 experiment, the abalone were removed from the tanks and placed in another farm husbandry
321 structure.

322

323 RESULTS

324

325 Nocturnal foraging activity

326 The abalone foraging behaviour varied between the day and night: the total number of feeder
327 visits was much higher during the night than during the day periods (night vs day period: 91 ± 9.4
328 vs 13 ± 2.4 visits per tank over the 3-week period, Wilcoxon signed rank test for paired data, $N =$
329 15, $P < 0.001$) (Figure 2).

330

331 Food choices of the population

332 The total quantity ingested varied between algal species (Freidman two-way analysis of variance
333 by ranks, $Q = 48.4$, $N = 15$, $P < 0.001$) (Figure 3): *Enteromorpha intestinalis* was the most
334 ingested alga, followed by *Asparagopsis armata*. On the other hand, *Laminaria digitata* and stipes
335 of *Laminaria hyperborea* were the algae ingested in the smallest quantities. *Ulva lactuca*,
336 *Saccharina latissima*, *Saccorhiza polyschides* and *Palmaria palmata* were ingested in intermediate
337 quantities, not significantly different from the most ingested alga or the least ingested alga.

338

339 The Freidman two-way analysis of variance by ranks showed that there were significant
340 differences between algal species in the number of feeding visits per day ($Q = 45.7$, $N = 15$, $P <$
341 0.001), the total time spent feeding each day ($Q = 42.4$, $N = 15$, $P < 0.001$) and the mean feeding
342 bout duration ($Q = 36.7$, $N = 15$, $P < 0.001$) (Figure 3). However, the ranks for the algae were
343 different depending on the variable (Figure 3): for example, *Enteromorpha intestinalis* was the
344 alga ranked first in terms of quantity ingested and for the number of visits, but was ranked fifth for
345 the mean feeding duration; while *Saccharina latissima* was ranked respectively fourth and fifth for

346 the quantity ingested and number of feeding visits, but second for the mean feeding bout duration.

347

348 Food choice in relation to algal characteristics

349 The quantity of algae ingested per day as well as the number of feeding visits per day were
350 significantly correlated to the protein content of the algae (respectively, $r_s = 0.40$ and $r_s = 0.54$) and
351 negatively correlated to algal toughness (respectively, $r_s = -0.50$ and $r_s = -0.51$) (Table 1). In
352 addition, the number of visits per day was positively correlated to total lipid content ($r_s = 0.35$), n-
353 3/n-6 ratio ($r_s = 0.35$) and negatively correlated to MUFA content ($r_s = -0.34$). No correlation was
354 observed between the mean feeding bout duration and any of the algal chemical components. The
355 total time spent feeding was correlated to the total protein content ($r_s = 0.39$) and to the total lipid
356 content ($r_s = 0.33$).

357

358 Individual and population specialisation

359 The *H. tuberculata* population consumed resources in a similar way, with a specialisation index
360 IS of 0.64 ($p < 0.001$). However, 21% of the individuals had a PS < 0.50 (Figure 4)

361

362 Individual foraging activity consistency

363 Very high consistency of individual foraging activity was observed for the total time spent
364 feeding per day (ICC = 0.81, $F_{119,238} = 5.2$, $p < 0.001$, 95% confidence interval = 0.74 – 0.86), as
365 well as for the number of feeder visits per day (ICC = 0.74, $F_{119,238} = 3.8$, $p < 0.001$, 95%
366 confidence interval = 0.64 – 0.81, log transformed data) and for the mean feeding bout duration
367 (ICC = 0.74, $F_{119,238} = 3.8$, $p < 0.001$, 95% confidence interval = 0.65 – 0.81, log
368 transformed data).

369

370

371 DISCUSSION

372 The aim of the experiment was to understand the foraging strategy of *H. tuberculata*, a marine
373 herbivore at both a population and an individual level. The food choices of *H. tuberculata* are
374 mainly correlated to the protein composition and the toughness of the macroalgae at the population
375 level. The population was composed mostly of generalist individuals with a few specialist
376 individuals. Surprisingly, foraging activity of individuals is highly consistent.

377

378 Nocturnal foraging activity

379

380 Foraging activity was mainly observed at night. Foraging activity was intense soon after sunset
381 and progressively increased until 22:00 to reach a stable level between 22:00 and 02:00. Nocturnal
382 feeding and movement has also been reported for other abalone species in laboratory studies
383 (Momma & Sato, 1970; Tahil & Juinio-Menez, 1999) and field conditions (Day & Branch, 2000;
384 Shepherd, 1973; Wood & Buxton, 1996). Even if light intensity is probably the main cue to
385 coordinate abalone circadian behaviour (Morikawa & Norman, 2003), a circatidal clock due to
386 tides cannot be excluded (Wilcockson & Zhang, 2008). There was no obvious sign of tidal
387 variation however, during this laboratory experiment.

388

389 Food choices of the population

390 Although local algal abundances may be the overriding factor determining the diet of abalone in
391 a particular area in the wild (Tutschulte & Connell, 1988; Wood & Buxton, 1996; Zeeman,
392 Branch, Peschak, & Pillay, 2012), as in many laboratory studies, the abalone we studied preferred
393 some algae over others. *Enteromorpha intestinalis*, with *Ulva lactuca* and *Asparagopsis armata*

394 were the preferred algae in term of quantity of algae ingested and numbers of feeding visits.
395 *Laminaria hyperborea* stipes were the least attractive. However, the total time spent feeding and
396 the mean feeding duration give additional information on abalone feeding behaviour. For example,
397 *Asparagopsis armata*, the second most ingested algae in terms of quantity, is at the sixth position
398 for the total time spent feeding and at the seventh position for the mean feeding duration. It is
399 clearly easily consumed.

400

401 Food choices based on primary compounds and toughness of algae

402

403 Optimal foraging theory implies that organisms should consume foods that maximize fitness by
404 selecting the most energy-rich and nutritional balanced items (Pyke, Pulliam, & Charnov, 1977).
405 The three most ingested algae, *Enteromorpha intestinalis*, *Ulva lactuca* and *Asparagopsis armata*
406 had the highest protein contents, while the lowest protein value was associated with the least
407 ingested alga, stipes of *Laminaria hyperborea*. This experiment shows that the total quantity of an
408 alga ingested and total number of feeding visits were correlated with total protein content but also
409 other parameters such as algal toughness. This is consistent with an emphasis on protein content in
410 abalone food choice and the lower importance of total carbohydrate (Fleming, 1995a). Energy in
411 the form of available carbon compounds is generally thought to be in ample supply in plants and
412 seaweeds because of photosynthesis, but this is not the case for nitrogen compounds, considered to
413 be a more limiting factor. Abalone species have a low lipid requirement (< 5% algal dry weight),
414 typical of herbivorous molluscs and fish (Viera et al., 2005). Total lipid content was only weakly
415 related to food choices. No correlation was found between the level of phenolic compounds and
416 *Haliotis tuberculata* food choices in our experiment, in contrast to the expected avoidance of algae
417 with phenolics (Shepherd and Steinberg 1992). This might be related to the very low phenolic

418 contents observed during this winter period, in the algal species analysed. Effects on feeding
419 behaviour might have been more important during spring or summer, when algae phenolic
420 contents are higher and probably would serve as feeding deterrents or inhibitors for abalone
421 (Fleming, 1995b; Stepto & Cook, 1996).

422 Algae toughness has been identified as a factor limiting seaweed consumption (Toth & Pavia,
423 2007). Some experiments have avoided this factor by mixing powdered algae with agar or
424 carrageenan, to remove its effect (Angell, Pirozzi, de Nys, & Paul, 2012; McShane, Gorfine, &
425 Knuckey, 1994). Abalone feed using a rasp-like radula, and the effectiveness of the radula teeth is
426 strongly influenced by the form and toughness of the food items. In our experiment, it seems that
427 the morphological structure of the *Haliotis tuberculata* radula allows them to graze more
428 effectively on filamentous and thinner algae such as *Asparagopsis armata* and *Enteromorpha*
429 *intestinalis* than on the less favoured thick and tough *Laminaria* leaves and stipes.

430

431 Consistent foraging activity

432 Highly significant foraging activity repeatability was observed during this experiment. Obtaining
433 accurate, reliable and repeatable phenotypic information is critical for better understanding of how
434 the personalities of individuals can be maintained under environments with different selective
435 pressures (Sih, Bell, & Johnson, 2004). At a population level, in contrast to our hypothesis,
436 foraging activity was very repeatable. In a meta-analysis performed on a wide range of species, the
437 average repeatability of behaviour was reported to be 0.37 (Bell et al., 2009). Foraging behaviour
438 repeatability was reported to be higher (Bell et al., 2009; McHuron et al., 2018) but nevertheless
439 ranged between 0.2 and 0.5 most of the time (Koteja et al., 2003; McHuron et al., 2018; Missoweit
440 et al., 2007; Morgan et al., 2019). In our experiment, the high ICC value and narrow 95%
441 confidence interval for abalone were probably obtained because the mean values for four days per

442 week of a large number of individuals were analysed, sufficient to study repeatability with
443 precision (Wolak, Fairbairn, & Paulsen, 2012). This result highlights that high consistency of
444 individual foraging activity can be observed for abalone, indicating that this herbivore has a rigid
445 foraging activity, at least in winter. The number of feeding visits per day might be a foraging
446 variable related to intrinsic factors (such as active or passive “type” personality). The total time
447 spent feeding per day and mean feeding bout duration have a lower ICC and might be more
448 influenced by extrinsic factors, such as the toughness and other properties of the algal species
449 eaten.

450

451 Individual and population specialisation

452 *H. tuberculata* is a generalist that feeds on a variety of algae (IS = 0.64). Most individuals were
453 opportunistic, feeding on a large range of algae. But 21% of the individuals can be considered as
454 specialists. Based on our observation of the data, some specialist individuals might choose the
455 same algae because they have a greater appetite for some physical attribute (e.g. the most tender
456 algae such as *A. armata* and *E. intestinalis*) or chemical attribute (e.g. the richest in protein such as
457 *P. palmata*). However for others, no trend can be easily observed. Because some abalone have
458 homing behaviour, their choices might be related to the algae that are closest to their hiding place.
459 Each abalone presumably evaluated the food differently and made decisions to select a suitable
460 diet. Individual differences can result from intrinsic factors (metabolism or individual experience)
461 or extrinsic factors (intraspecific competition or predation). Because these herbivores were reared
462 in the same sea-cage for three years, their previous individual experiences in relation to their
463 rearing environment are probably similar. Metabolism might be an important factor influencing the
464 differences in feeding preference (Hawlana, Hughes, & Schmitz, 2011; Holtmann, Lagisz, &
465 Nakagawa, 2017). Components of the algae previously consumed are also likely to influence

466 future preferences, to gain nutrients lacking in the past diet (Day & Fleming, 1992). In addition, it
467 should be noted that this experiment was performed in a laboratory context with a simplified
468 environment. In the wild, individuals have far more complex decisions to take, depending of the
469 external situation such as the presence of predators or hiding places, algal composition and
470 quantity, etc...

471 Choosing a mixed diet is probably the best compromise for large marine herbivores, consistent
472 with nutritional advantages and conferring significantly higher fitness than the average of single-
473 species diets, but not for the best single prey species (review by Lefcheck, Whalen, Davenport,
474 Stone, & Duffy, 2013). In a one-year experiment, we have confirmed the advantage of a mixed
475 diet foraging strategy on *Haliotis tuberculata*, showing that the mixed diet was the second best
476 strategy in terms of growth and reproductive development after a monospecific diet of the best
477 single alga, *P. palmata* (Roussel et al., 2019). In the wild, *P. palmata* is often likely to be
478 unavailable. By mixing different sources of algae, a multi-algal diet would allow the acquisition of
479 vitamins, essential fatty acids and amino acids present in different proportions in various algal
480 species. This result fits well the optimal diet theory in the sense that the mixed diet will produce
481 the highest fitness, even if the algae chosen do not have the highest energy value individually (Sih
482 & Christensen, 2001).

483

484 CONCLUSION

485

486 *H. tuberculata* is a generalist species in term of algal choice, with a small proportion of the
487 individuals being specialists and the majority being generalists. In addition, individual foraging
488 activity pattern was highly consistent over time, with some abalone feeding every night while
489 others fed only rarely. This study provides a new perspective on the foraging strategy of an

490 abalone species – a very widespread group of large herbivorous marine molluscs. Algal
491 preferences probably result from a compromise between the most useful algae from a nutritional
492 point of view, the physical ability of abalone to consume the food item and some individual
493 consistency. In a further study, it would be interesting to test the interaction of abalone personality
494 traits such as activity and exploration in a novel environment in addition to abalone food resource
495 use, to understand how variations among individuals contribute to patterns at the population level
496 (Toscano et al., 2016).

497

498 REFERENCES

499

500 Angell, A. R., Pirozzi, I., de Nys, R., & Paul, N. A. (2012). Feeding preferences and the
501 nutritional value of tropical algae for the abalone *Haliotis asinina*. *Plos One*, 7(6),
502 e38857. doi:10.1371/journal.pone.0038857

503 Bell, A., Hankison, S., & Laskowski, K. L. (2009). The repeatability of behaviour: a meta-
504 analysis. *Animal Behaviour*, 77(4), 771-783. doi:10.1016/j.anbehav.2008.12.022

505 Biro, P. A., & Stamps, J. A. (2015). Using repeatability to study physiological and behavioural
506 traits: ignore time-related change at your peril. *Animal Behaviour*, 105, 223-230.
507 doi:10.1016/j.anbehav.2015.04.008

508 Bolnick, D., Svanbäck, R., Fordyce, J., Yang, L., Davis, J., Hulsey, C. D., & Forister, M. (2003).
509 The ecology of individuals: incidence and implications of individual specialization. *The*
510 *American Naturalist*, 161(1), 1-28. doi:10.1086/343878

511 Bolnick, D. I., Yang, L. H., Fordyce, J. A., Davis, J. M., & Svanback, R. (2002). Measuring
512 individual-level resource specialization. *Ecology*, 83(10), 2936-2941.
513 doi:10.2307/3072028

- 514 Connan, S. (2004). Etude de la biodiversité des macroalgues des côtes de Bretagne et potentiel de
515 valorisation des substances UV-protectrices. PhD Thesis. (PhD), University of Western
516 Brittany, Brest.
- 517 Day, E., & Branch, G. M. (2000). Relationships between recruits of abalone *Haliotis midae*,
518 encrusting corallines and the sea urchin *Parechinus angulosus*. *South African Journal of*
519 *Marine Science*, 22, 137-144.
- 520 Day, R. W., & Cook, P. (1995). Bias towards brown algae in determining diet and food
521 preferences: The South African abalone *Haliotis midae*. *Marine and Freshwater*
522 *Research*, 46(3), 623-627. doi:10.1071/MF9950623
- 523 Day, R. W., & Fleming, A. E. (1992). The determinants and measurement of abalone growth. In
524 S. A. Shepherd, M. J. Tegner, & S. A. Guzman Del Proo (Eds.), *Abalone of the world:*
525 *biology, fisheries and culture* (pp. 141-168). Oxford: Fishing News Books.
- 526 Dohm, M. R. (2002). Repeatability estimates do not always set an upper limit to heritability.
527 *Functional Ecology*, 16(2), 273-280. doi:10.1046/j.1365-2435.2002.00621.x
- 528 Falconer, D. S., & Mackay, T. F. C. (1996). *Introduction to quantitative genetics*. 4th edition.
529 London, UK: Longman.
- 530 Fleming, A. E. (1995a). Digestive efficiency of the Australian abalone *Haliotis rubra* in relation
531 to growth and feed preference. *Aquaculture*, 134, 3-4. doi:10.1016/0044-8486(95)00055-7
- 532 Fleming, A. E. (1995b). Growth, intake, feed conversion efficiency and chemosensory preference
533 of the Australian abalone, *Haliotis rubra*. *Aquaculture*, 132, 3-4. doi:10.1016/0044-
534 8486(94)00347-Q
- 535 Folch, J., Lees, M., & Stanley, G. H. S. (1957). A simple method for the isolation and purification
536 of total lipides from animal tissues. *Journal of Biological Chemistry*, 226(1), 497-509.
- 537 Griffen, B. D., Toscano, B. J., & Gatto, J. (2012). The role of individual behavior type in

538 mediating indirect interactions. *Ecology*, 93(8), 1935-1943. doi:10.1890/11-2153.1

539 Hawlena, D., Hughes, K. M., & Schmitz, O. J. (2011). Trophic trait plasticity in response to
540 changes in resource availability and predation risk. *Functional Ecology*, 25(6), 1223-
541 1231. doi:10.1111/j.1365-2435.2011.01891.x

542 Holtmann, B., Lagisz, M., & Nakagawa, S. (2017). Metabolic rates, and not hormone levels, are a
543 likely mediator of between-individual differences in behaviour: a meta-analysis.
544 *Functional Ecology*, 31, 685–696. doi:10.1111/1365-2435.12779

545 Jung, K. A., Lim, S. R., Kim, Y., & Park, J. M. (2013). Potentials of macroalgae as feedstocks for
546 biorefinery. *Bioresource Technology*, 135, 182-190. doi:10.1016/j.biortech.2012.10.025

547 Koteja, P., Carter, P., Swallow, J., & Garland, T. (2003). Food wasting by house mice: variation
548 among individuals, families, and genetic lines. *Physiology & Behavior*, 80(2-3), 375-383.
549 doi:10.1016/j.physbeh.2003.09.001

550 Lefcheck, J. S., Whalen, M. A., Davenport, T. M., Stone, J. P., & Duffy, J. E. (2013).
551 Physiological effects of diet mixing on consumer fitness: a meta-analysis. *Ecology*, 94(3),
552 565-572. doi:10.2307/23436259

553 Lockwood, J. R. (1998). On the statistical analysis of multiple-choice feeding preference
554 experiments. *Oecologia*, 116(4), 475-481. doi:10.1007/s004420050612

555 Mai, K., Mercer, J. P., & Donlon, J. (1994). Comparative studies on the nutrition of two species
556 of abalone, *Haliotis tuberculata* L. and *Haliotis discus hannai* Ino: 2. Amino acid
557 composition of abalone and six species of macroalgae with an assessment of their
558 nutritional value. *Aquaculture*, 128(1–2), 115-130. doi:10.1016/0044-8486(94)90107-4

559 Mai, K., Mercer, J. P., & Donlon, J. (1995). Comparative studies on the nutrition of two species
560 of abalone, *Haliotis tuberculata* L. and *Haliotis discus hannai* Ino. 3. Response of abalone
561 to various levels of dietary lipid. *Aquaculture*, 134, 65-80. doi:10.1016/0044-

562 8486(95)00043-2

563 Mather, J. A., Leite, T. S., & Batista, A. T. (2012). Individual prey choices of octopuses: Are they
564 generalist or specialist? *Current Zoology*, 58(4), 597-603. doi:10.1093/czoolo/58.4.597

565 Mattson, W. J. (1980). Herbivory in relation to plant nitrogen-content. *Annual Review of Ecology*
566 *and Systematics*, 11, 119-161. doi:10.1146/annurev.es.11.110180.001003

567 McHuron, E. A., Hazen, E., & Costa, D. P. (2018). Constrained by consistency? Repeatability of
568 foraging behavior at multiple timescales for a generalist marine predator. *Marine Biology*,
569 165(8), 122. doi:10.1007/s00227-018-3382-3

570 McShane, P. E., Gorfine, H. K., & Knuckey, I. A. (1994). Factors influencing food selection in
571 the abalone *Haliotis rubra* (Mollusca: Gastropoda). *Journal of Experimental Marine*
572 *Biology and Ecology*, 176(1), 27-37. doi:10.1016/0022-0981(94)90195-3

573 Missoweit, M., Engels, S., & Sauer, K. P. (2007). Foraging ability in the scorpionfly *Panorpa*
574 *vulgaris*: individual differences and heritability. *Behavioral Ecology and Sociobiology*,
575 61(3), 487-492. doi:10.1007/s00265-006-0277-y

576 Momma, H., & Sato, R. (1970). The locomotion behavior of the disc abalone, *Haliotis discus*
577 *hannai* Ino, in a tank. *Tohoku Journal of Agricultural Research*, 21(1), 20-25.

578 Morgan, E. A., Hassall, C., Redfern, C. P. F., Bevan, R. M., & Hamer, K. C. (2019). Individuality
579 of foraging behaviour in a short-ranging benthic marine predator: incidence and
580 implications. *Marine Ecology Progress Series*, 609, 209-219. doi:10.3354/meps12819

581 Morikawa, Y., & Norman, C. P. (2003). Perception of light intensity by *Haliotis discus discus*
582 based on locomotor activity patterns. *Fisheries Science*, 69(3), 478-486.
583 doi:10.1046/j.1444-2906.2003.00648.x

584 Mousseau, T., & Roff, D. A. (1987). Natural selection and the heritability of fitness components.
585 *Heredity*, 59(2), 181-197.

586 Patrick, S. C., Bearhop, S., Grémillet, D., Lescroël, A., Grecian, W. J., Bodey, T. W., . . . Votier, S.
587 C. (2014). Individual differences in searching behaviour and spatial foraging consistency
588 in a central place marine predator. *Oikos*, 123(1), 33-40. doi:10.1111/j.1600-
589 0706.2013.00406.x

590 Pennings, S. C., & Carefoot, T. H. (1995). Postingestive consequences of consuming secondary
591 metabolites in sea hares (Gastropoda: Opisthobranchia). *Comparative Biochemistry and*
592 *Physiology C-Pharmacology Toxicology & Endocrinology*, 111(2), 249-256.
593 doi:10.1016/0742-8413(95)00044-o

594 Pyke, G. H., Pulliam, H. R., & Charnov, E. L. (1977). Optimal foraging - selective review of
595 theory and tests. *Quarterly Review of Biology*, 52(2), 137-154. doi:10.1086/409852

596 Renaud, S. M., & Luong-Van, J. T. (2006). Seasonal variation in the chemical composition of
597 tropical Australian marine macroalgae. *Journal of Applied Phycology*, 18(3-5), 381-387.
598 doi:10.1007/s10811-006-9034-x

599 Revelle, W. (2018). Package 'psych', version 1.8.12, [https://cran.r-](https://cran.r-project.org/web/packages/psych/psych.pdf)
600 [project.org/web/packages/psych/psych.pdf](https://cran.r-project.org/web/packages/psych/psych.pdf).

601 Roussel, S., Caralp, C., Leblanc, C., Le Grand, F., Stiger-Pouvreau, V., Coulombet, C., . . .
602 Huchette, S. (2019). Impact of nine macroalgal diets on growth and initial reproductive
603 investment in juvenile abalone *Haliotis tuberculata*. *Aquaculture*, 734385.
604 doi:10.1016/j.aquaculture.2019.734385

605 Scheidel, U., & Bruelheide, H. (1999). Selective slug grazing on montane meadow plants.
606 *Journal of Ecology*, 87(5), 828-838. doi:10.1046/j.1365-2745.1999.00402.x

607 Schmid, M., Guiheneuf, F., & Stengel, D. B. (2014). Fatty acid contents and profiles of 16
608 macroalgae collected from the Irish coast at two seasons. *Journal of Applied Phycology*,
609 26(1), 451-463. doi:10.1007/s10811-013-0132-2

- 610 Shepherd, S. A. (1973). Studies on southern Australian abalone (genus *Haliotis*). I. Ecology of
611 five sympatric species. *Australian Journal of Marine and Freshwater Research*, 24, 217-
612 257.
- 613 Shepherd, S. A., & Steinberg, P. D. (1992). Food preferences of three Australian abalone species
614 with a review of the algal food of abalone. In S. A. Shepherd, M. J. Tegner, & S. A.
615 Guzmán del Prío (Eds.), *Abalone of the world: biology, fisheries and culture* (pp. 169-
616 181). Oxford: Blackwell Scientific Publications.
- 617 Siegel, S., & Castellan, N. J. (1988). *Nonparametric statistics for the behavioural sciences*. New
618 York: McGraw-Hill International Editions.
- 619 Sih, A., Bell, A., & Johnson, J. C. (2004). Behavioral syndromes: an ecological and evolutionary
620 overview. *Trends in Ecology & Evolution*, 19(7), 372-378. doi:10.1016/j.tree.2004.04.009
- 621 Sih, A., & Christensen, B. (2001). Optimal diet theory: when does it work, and when and why
622 does it fail? *Animal Behaviour*, 61, 379-390. doi:10.1006/anbe.2000.1592
- 623 Slinkard, K., & Singleton, V. L. (1977). Total phenol analysis - automation and comparison with
624 manual methods. *American Journal of Enology and Viticulture*, 28(1), 49-55.
- 625 Steneck, R. S., & Watling, L. (1982). Feeding capabilities and limitation of herbivorous mollusks
626 - a functional group approach. *Marine Biology*, 68(3), 299-319. doi:10.1007/bf00409596
- 627 Stepto, N. K., & Cook, P. A. (1996). Feeding preferences of the juvenile South African abalone
628 *Haliotis midae* (Linnaeus, 1758). *Journal of Shellfish Research*, 15(3), 653-657.
- 629 Sterner, R. W., & Hessen, D. O. (1994). Algal nutrient limitation and the nutrition of aquatic
630 herbivores. *Annual Review of Ecology and Systematics*, 25, 1-29.
631 doi:10.1146/annurev.es.25.110194.000245
- 632 Tahil, A. S., & Juinio-Menez, M. A. (1999). Natural diet, feeding periodicity and functional
633 response to food density of the abalone, *Haliotis asinina* L., (Gastropoda). *Aquaculture*

- 634 Research, 30(2), 95-107. doi:10.1046/j.1365-2109.1999.00294.x
- 635 Toscano, B. J., Gownaris, N. J., Heerhartz, S. M., & Monaco, C. J. (2016). Personality, foraging
636 behavior and specialization: integrating behavioral and food web ecology at the individual
637 level. *Oecologia*, 182(1), 55-69. doi:10.1007/s00442-016-3648-8
- 638 Toscano, B. J., & Griffen, B. D. (2014). Trait-mediated functional responses: predator
639 behavioural type mediates prey consumption. *Journal Of Animal Ecology*, 83(6), 1469-
640 1477. doi:10.1111/1365-2656.12236
- 641 Toth, G. B., & Pavia, H. (2007). Induced herbivore resistance in seaweeds: a meta-analysis.
642 *Journal of Ecology*, 95(3), 425-434. doi:10.1111/j.1365-2745.2007.01224.x
- 643 Tutschulte, T. C., & Connell, J. H. (1988). Feeding-behavior and algal food of three species of
644 abalones (*Haliotis*) in southern-California. *Marine Ecology Progress Series*, 49(1-2), 57-
645 64. doi:10.3354/meps049057
- 646 Valen, L. V. (1965). Morphological variation and width of ecological niche. *The American*
647 *Naturalist*, 99(908), 377-390. doi:10.1086/282379
- 648 Viera, M. P., Pinchetti, J. L. G., de Vicose, G. C., Bilbao, A., Suarez, S., Haroun, R. J., &
649 Izquierdo, M. S. (2005). Suitability of three red macroalgae as a feed for the abalone
650 *Haliotis tuberculata coccinea* Reeve. *Aquaculture*, 248(1-4), 75-82.
651 doi:10.1016/j.aquaculture.2005.03.002
- 652 Villares, R., Fernandez-Lema, E., & Lopez-Mosquera, E. (2013). Seasonal variations in
653 concentrations of macro- and micronutrients in three species of brown seaweed. *Botanica*
654 *Marina*, 56(1), 49-61. doi:10.1515/bot-2012-0114
- 655 Wilcockson, D., & Zhang, L. (2008). Circatidal clocks. *Current biology*, 18(17), R753-R755.
656 doi:<https://doi.org/10.1016/j.cub.2008.06.041>
- 657 Winter, F. C., & Estes, J. A. (1992). Experimental evidence for the effects of polyphenolic

658 compounds from *Dictyoneurum californicum* Ruprecht (Phaeophyta: Laminariales) on
659 feeding rate and growth in the red abalone *Haliotis rufescens* Swainson. *Journal of*
660 *Experimental Marine Biology and Ecology*, 155(2), 263-277. doi:10.1016/0022-
661 0981(92)90067-K

662 Wolak, M. E., Fairbairn, D. J., & Paulsen, Y. R. (2012). Guidelines for estimating repeatability.
663 *Methods in Ecology and Evolution*, 3(1), 129-137. doi:10.1111/j.2041-210X.2011.00125.x

664 Wood, A. D., & Buxton, C. D. (1996). Aspects of the biology of the abalone *Haliotis midae*
665 (Linne, 1758) on the east coast of South Africa. 1. Feeding biology. *South African Journal*
666 *of Marine Science*, 17, 61-68. doi:10.2989/025776196784158590

667 Zaccarelli, N., Mancinelli, G., & Bolnick, D. I. (2013). RInSp: an R package for the analysis of
668 individual specialisation in resource use. *Methods in Ecology and Evolution*, 4(11), 1018-
669 1023. doi:10.1111/2041-210X.12079

670 Zeeman, Z., Branch, G. M., Peschak, T. P., & Pillay, D. (2012). Assessing the ecosystem effects
671 of the abalone *Haliotis midae* from its diet and foraging behaviour. *African Journal of*
672 *Marine Science*, 34(2), 205-214. doi:10.2989/1814232x.2012.675119

673

674

675

676 Table 1: Correlation between the primary components, secondary metabolites and toughness of 8
 677 algal and the foraging behaviour of 15 groups of 8 abalone. The correlation was performed on the
 678 average foraging value obtained per algae for each group of abalone four days per week during a
 679 3-week period. Significant correlations are highlighted in bold

680

	Quantity of algae ingested per day (g / day)	Time spent		
		feeding per day (min / day)	Number of feeding visits per day	Mean feeding bout duration
Dry matter content %	-0.34 (p = 0.01)	-0.04	-0.15	0.05
Algal toughness (rank)	-0.50 (p < 0.001)	-0.28	-0.51 (p < 0.001)	-0.00
Total protein content (%)	0.40 (p < 0.001)	0.39 (p < 0.001)	0.54 (p < 0.001)	0.12
Total carbohydrate content (%)	-0.14	0.01	0.12	-0.10
Total lipid content (µg FA / mg dry matter)	0.10	0.33 (p = 0.01)	0.35 (p < 0.01)	0.18
n-3/n-6 ratio	0.13	0.24	0.35 (p < 0.01)	0.06
MUFA content (% of total FA)	-0.21	-0.12	-0.34 (p = 0.01)	0.10
PUFA content (% of total FA)	0.01	0.21	0.17	0.14
SFA content (% of total FA)	-0.05	-0.20	-0.09	-0.19
Total phenolics (mg GAE or PE / g fresh algae sample)	0.41 (p < 0.001)	0.14	0.26	0.02

681

682

683

684 Figure 1: Design of the experimental tank

685

686 Figure 2. Number of feeder visits initiated each 2-h period of the 24-hour cycle of *Haliotis*
687 *tuberculata*. Eight abalone per tank were observed for 4 days per week during a 3-week period in a
688 total of 15 tanks. Mean \pm s.e.

689

690 Figure 3. Algal preferences of abalone. Foraging behaviour was followed four days per week
691 during a 3-week period in 15 tanks containing 8 abalone

692

693 Figure 4. Proportional similarity index (Psi) frequency in a population of 102 abalone studied
694 during 3 weeks. Abalone had the possibility to choose between eight different algae in a tank.

695