When the microclimate does matter: differences in the demographic, morphometric and reproductive variables of *Fucus guiryi* from two nearby populations

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Introduction

Marine forests and canopy-forming macroalgae provide essential ecosystemic services from cold to warm latitudes, whose positive effects cascade through their communities and sustain whole ecosystems. However, global change is leading to the retreat and extinction of many populations over the past decade (Riera et al., 2015). This process is particularly enhanced at southern-range and peripheral populations, which are more vulnerable to climatic stress due to their exposure to higher absolute temperatures, despite local conditions can ameliorate microclimatic conditions, thus providing climatic refugia (Lourenço et al., 2016; Melero-Jiménez et al., 2017).

This study aims at comparing the demographic, morphometric and reproductive-ecological variables of two close populations of *Fucus guiryi* [Zardi, Nicastro, E.S.Serrão & G.A.Pearson] at the Strait of Gibraltar (Tarifa-TRF, Guadalmesí-GDM, ca. 8 km away).

Material and methods

Seasonal sampling surveys took place between spring 2018 and spring 2019, at diurnal low spring tides. All the variables were studied in spring and fall, while in summer and winter only individual length and reproductive status were recorded. Microclimate of each sampling site was characterized from direct observations, deployment of *in situ* temperature loggers during summer 2018, and retrieval of meteorological and oceanographic variables from public databases. For the demographic variables, non-destructive quadrats of 0.16 m^2 (n=4) were sampled randomly within the intermediate intertidal zone. Length and reproductive status (vegetative, incipient, mature apices) were determined for each individual. Cover was calculated from 5 equidistant transects perpendicular to the highest water mark, by recording the exact intervals where *F. guiryi* was present. At GDM, cover was estimated from pictures due to the low extent and cover of the population. Morphometric variables were measured at each site and sampling from 10 individuals (individual bushiness - IB, length of the frond between the holdfast and the first dichotomy - LF, frond width-WF and receptacle size (height-RH, width-RW, length-RL). Two-way factorial ANOVA was applied to test the main and interactive effects among sites and season, except when data lacked and one-way ANOVA or t-tests were performed. Reproductive status was analyzed by means of PERMANOVA.

Results and discussion

Microclimatic conditions differed greatly due to local characteristics of each sampling site. TRF presented a higher proportion of bare rock covered by algae, more abrupt rock surface with abundant tidal pools of 10-100 cm depth. This ambient prevents water loss and ameliorates abiotic stress during emersion. In contrast, *F. guiryi* at GDM is disposed as patches on flat or slightly angled surfaces of the flysch platforms, and outside their canopies, there is mainly acorn-barnacle-covered-bare rock.

Maximum air temperature registered did not exceed 30°C, while at the upper intertidal distributional limits temperature raised up to 41.5±2.8°C at TRF and 44.3±1.0°C, at GDM. Nevertheless, no significant differences in mean temperatures (21-24.6°C) or mean values of maximum (32-27°C) and minimum temperature (17-18.3°C) were obtained. Canopy reduced temperature by 5-7°C and attenuated irradiance by 10-16%, corresponding to irradiances below 100 μmol m^{-2} s^{-1}, values highly consistent with those reported for several populations of *F. guiryi* from Portugal (Monteiro et al., 2019).

On the other hand, TRF was exposed to higher waves than at GDM, due to the shore position in relation to the predominant wave direction (W). In turn, individuals of *F. guiryi* are exposed to higher environmental stress at GDM, due to the higher maximum temperature, less frequent watering, patchier distribution, and more direct wave impact that can lead to plant detachment.

Regarding demographic variables, sampling site and season had a significant effect on the thalli length of *F. guiryi*, with no significant interaction among factors. *Fucus guiryi* from both sites experienced parallel seasonal dynamics in individual length, showing the expected growing season from spring to summer observed in many temperate macroalgae, when maximum size is reached (Figure 1). However, individuals from TRF were 1.4-1.8 times longer than at GDM (Figure 1). Both populations differed in their minimum (TRF 1.4±0.6 cm; GDM 0.8±0.2 cm), mean (TRF 5.8±1.0 cm; GDM 3.5±0.9 cm) and maximum (TRF 11±2 cm; GDM 7±2 cm) thalli length across seasons. As a result, both had different population structure and heterogeneity. TRF had a significantly higher percentage of individuals of size classes 6-12 cm than GDM, whereas at the latter 84% of individuals were below 4 cm ($p<0.01$). In addition, the percentage of new recruits (0-2 cm) at GDM has significantly increased during the study, probably due to the frond breakage by wave storms after summer 2018.

Fucus guiryi from TRF had twice the population density and cover than at GDM (average density 55±26 *vs.* 26±15 individuals m^{-2}; average cover 14±7% *vs.* 7±3%). Population density has declined by 50% for both populations along the study, with no significant interactions among factors, whereas algal cover remained unaltered across seasons.

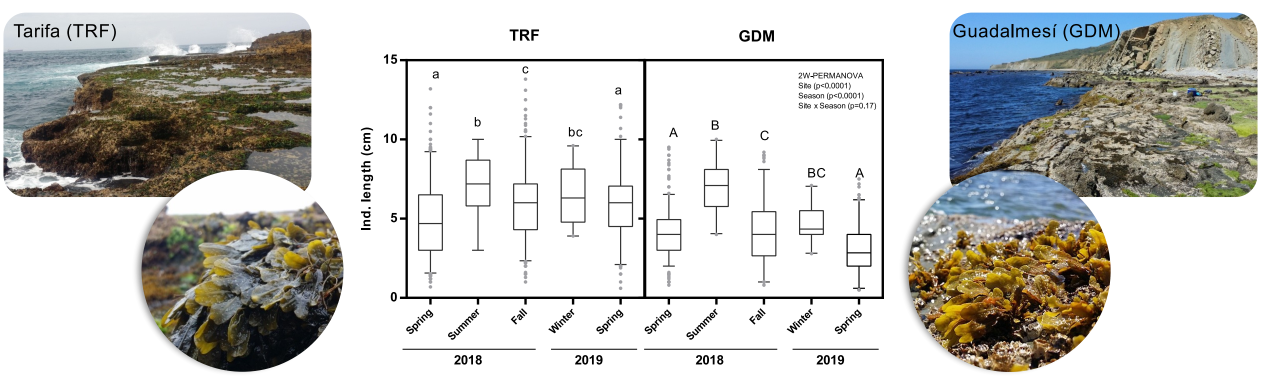

Figure 1: Seasonal changes in individual length of *F. guiryi* from two populations at the Strait of Gibraltar (TRF-Tarifa, GDM- Guadalmesí).

In spring 2018, *F. guiryi* from GDM presented lower bushiness than in TRF (GDM, 0.8±0.2). In fall, 25-52% of individuals from GDM experienced severe breakage of apical fronds, after a period of extreme air summer temperatures (up to 45.6°C in August) and an intense wave action in early autumn, which constrained the measure of all the morphological variables. *Fucus guiryi* from both populations presented similar LF and WF across the study (averaged values, LF: 1.4±0.5 cm; WF: 1.1±0.2 cm). LF was 3 times lower than those reported for Portuguese populations, while WF was more similar to *F. guiryi* from the Canary Islands (Riera et al., 2015) or *F. spiralis* from Portuguese populations (Zardi et al., 2015). Receptacle size varied among sites and samplings with interactive effects. Receptacles became thicker from spring to fall for both populations, being 2-times thicker for GDM specimens (0.4±0.1 *vs.* 0.8±0.2 cm). RL and RW remained invariable within each site, and only in fall 2018, receptacles were longer and wider for TRF specimens. Also, RW from specimens of GDM declined by 50% from spring to fall, probably derived from the disruptive stress experienced by this population after summer 2018.

Reproductive population increased throughout the study period, from 16-36% to 45-60%. Despite seasonality did not affect the percentage of reproductive individuals within each population, in spring 2019 TRF had a greater density of reproductive individuals than GDM (21±3% *vs.* 7±5, Figure 2). Minimum length of reproductive individuals neither experienced seasonal changes but differed greatly between sites, with 50% shorter individuals at GDM (2.2±0.8 cm) than in TRF (4.6±1.2 cm).

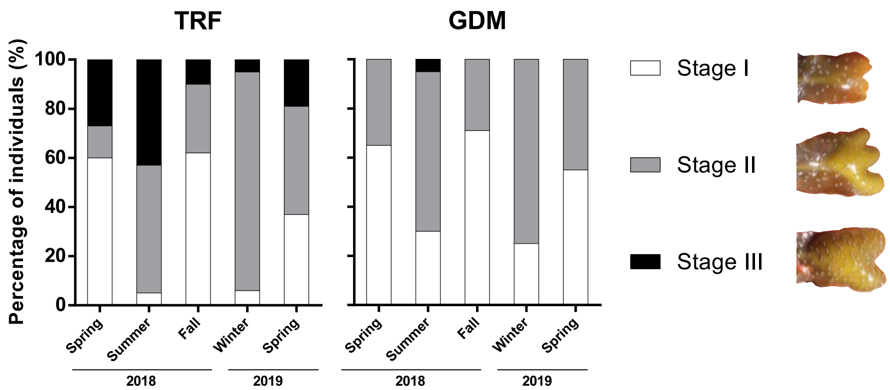

Figure 2: Seasonal changes in the percentage of individuals of *F. guiryi* at each reproductive stage (ST I- Vegetative; ST II- developing receptacles; ST III- Mature receptacles) from the two studied populations (TRF-Tarifa; GDM-Guadalmesí). Images of each stage are illustrated.

Table 1: Percentage of apical tips of *Fucus guiryi* from TRF at each reproductive stage (ST I: Vegetative; ST II: Developing receptacles; ST III: Mature receptacles) during the study period. Different letters indicate significant differences among seasons within each stage (p <0.05).

	Spring	Summer	Fall	Winter	Spring'19
ST I	97.91±7.25^{a}	50.95±29.98^{b}	95.76±10.13^{a}	69.5±20.06^{c}	86.83±19.92^{bc}
ST II	1.68±6.71^{a}	38.53±32.05^{b}	4.24±10.13^{c}	30.25±20.42^{b}	12.43±19.69^{d}
ST III	1.03±5.69^{a}	10.53±19.29^{abc}	1.58±7.72^{b}	0.25±1.12^{c}	2.37±7.05^{ab}

Site had a clear influence on the percentage of individuals reaching mature receptacles (p<0.001), with a significantly higher proportion of individuals at stage III at TRF (0-41%), for all seasons (Figure 2). *Fucus guiryi* at TRF exhibited a higher proportion of mature plants (27±19%), whereas at GDM it was only reported in summer'18 with less than 5% of the individuals (Figure 2), for which most of the individuals had developing receptacles (stage II, 35±11%). At TRF, proportion of individuals with mature receptacles increased by 15% from spring to summer 2018, when 41% of reproductive individuals reached stage III, with also a significant decrease in the % of vegetative apices (Table 1).

In conclusion, demographic, morphological and reproduction-related variables for the populations of *F. guiryi* from the Strait of Gibraltar were in good agreement with those from rear-edge populations from southern Portugal (eg. individual length, bushiness) (Zardi et al., 2015). Interestingly, specimens from TRF deviated from the correlation more than those from GDM (eg. higher bushiness, individual length and fertility at the growing season, and higher population extent, cover and density), suggesting that Tarifa acts as a true climate refugia for *F. guiryi* (Lourenço et al., 2016).

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