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| Abstract             | <p>Sexes of dioecious species may have dimorphic responses to environmental variation due to differences in resource requirements and reproductive costs. We analyzed the effect of aridity/relative shrub cover, and vicinity to shrub patches on morpho-chemical traits of sexes of the dioecious perennial grass <i>Poa ligularis</i> in patchy arid ecosystems in northern-central Patagonia. We hypothesized that sexes of <i>P. ligularis</i> have dimorphic responses in morpho-chemical traits in relation to the environmental variation induced by aridity/relative shrub cover and vicinity to shrub patches. We selected seven sites across a gradient of increasing aridity and relative shrub cover. We randomly collected 5–10 <i>P. ligularis</i> plants per site registering the sex (female or male) and location with respect to shrub patches (shrubs patch or inter-patch). For each plant, we assessed morpho-chemical traits (height of the vegetative tillers, length/dry weight/area of blades, specific blade area, nitrogen and soluble phenol concentration in blades). Sexes showed dimorphic responses in height of vegetative tillers, blade length, and blade area with respect to vicinity to shrub patches; and in variation of soluble phenolics in blades in relation to aridity/relative shrub cover. Responses in both sexes were opposite to those expected by aridity, highlighting the role of favorable environments induced by shrub canopies on dimorphic responses of sexes of <i>P. ligularis</i>. Resource-rich microsites associated with shrub canopies promoted increased plant performance of females with high</p> |  |

reproductive costs while resource-poor open areas, favorable for pollen dispersal, induced improved chemical defenses of males. These results are consistent with the resource availability hypothesis.

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Keywords (separated by '-') Arid Patagonia - Inter-patch areas - Plant defenses - Sexual dimorphism - Shrub patches - Vegetative growth

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Footnote Information

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2 **Variation of morphological and chemical traits in sexes**  
3 **of the dioecious perennial grass *Poa ligularis* in relation to shrub**  
4 **cover and aridity in Patagonian ecosystems**

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8 **Abstract** Sexes of dioecious species may have dimorphic  
9 responses to environmental variation due to differences in  
10 resource requirements and reproductive costs. We analyzed  
11 the effect of aridity/relative shrub cover, and vicinity to  
12 shrub patches on morpho-chemical traits of sexes of the  
13 dioecious perennial grass *Poa ligularis* in patchy arid  
14 ecosystems in northern-central Patagonia. We hypothe-  
15 sized that sexes of *P. ligularis* have dimorphic responses in  
16 morpho-chemical traits in relation to the environmental  
17 variation induced by aridity/relative shrub cover and  
18 vicinity to shrub patches. We selected seven sites across a  
19 gradient of increasing aridity and relative shrub cover. We  
20 randomly collected 5–10 *P. ligularis* plants per site regis-  
21 tering the sex (female or male) and location with respect to  
22 shrub patches (shrub patch or inter-patch). For each plant,  
23 we assessed morpho-chemical traits (height of the vegeta-  
24 tive tillers, length/dry weight/area of blades, specific blade  
25 area, nitrogen and soluble phenol concentration in blades).  
26 Sexes showed dimorphic responses in height of vegetative  
27 tillers, blade length, and blade area with respect to vicinity  
28 to shrub patches; and in variation of soluble phenolics in  
29 blades in relation to aridity/relative shrub cover. Responses  
30 in both sexes were opposite to those expected by aridity,  
31 highlighting the role of favorable environments induced by  
32 shrub canopies on dimorphic responses of sexes of *P.*  
33 *ligularis*. Resource-rich microsites associated with shrub

canopies promoted increased plant performance of females 34  
with high reproductive costs while resource-poor open 35  
areas, favorable for pollen dispersal, induced improved 36  
chemical defenses of males. These results are consistent 37  
with the resource availability hypothesis. 38

**Keywords** Arid Patagonia · Inter-patch areas · Plant 40  
defenses · Sexual dimorphism · Shrub patches · Vegetative 41  
growth 42

**Introduction** 43

Vegetation in arid ecosystems consists of a two phase 44  
mosaic formed by shrubby plant patches with high cover 45  
alternating with areas of sparse vegetation or bare soil 46  
(Aguiar and Sala 1999; Tongway and Ludwig 2005). Plant 47  
patches may improve the local microenvironment by cre- 48  
ating sheltered areas with reduced temperature and evap- 49  
orative demand, high concentration of organic matter, 50  
nutrients and seeds in soils, thus favoring plant establish- 51  
ment and growth in their vicinity (Burke et al. 1999; 52  
Guterman 2000; Erickson et al. 2005; Busso et al. 2012). 53  
The influence of plant patches on the microenvironment 54  
decreases from the center of the patch canopy to the 55  
neighboring bare soil areas (Burke et al. 1999; Bertiller 56  
et al. 2002; Tongway and Ludwig 2005). Areas of bare soil 57  
have not only lower soil organic matter and nutrients but 58  
also are more exposed to wind and water erosion than the 59  
soil beneath plant patches (Zaady et al. 1996; Tongway and 60  
Ludwig 2005). Moreover, shrubs dominating plant patches 61  
usually have chemical and morphological anti-herbivore 62  
defenses providing biotic refuges against herbivores for 63  
neighboring herbaceous plants with high growth rates and 64  
low chemical and morphological defenses (Milchunas and 65

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66 Noy-Meir 2002; Rebollo et al. 2002; Oosterheld and  
67 Oyarzábal 2004).

68 Dioecious species with sexual dimorphism may opti-  
69 mize the use of resources in space and time across gradi-  
70 ents of micro-environmental conditions induced by plant  
71 patches (Freeman et al. 1980; Bierzychudek and Eckhart  
72 1988; Dawson and Geber 1999). Some studies reported  
73 spatial segregation of sexes with females occupying  
74 microsities with high resource availability and males dom-  
75 inating in open microsities with low resources but favorable  
76 for pollen dispersal (Dawson and Ehleringer 1993; Eppley  
77 et al. 1998; Delph 1999; Obeso 2002; Eppley 2005). This  
78 pattern of spatial segregation could be related to differen-  
79 tial traits between sexes such as growth rates (Shea et al.  
80 1993; Graff et al. 2013); photosynthesis, transpiration, and  
81 water uptake rates (Dawson and Geber 1999; Retuerto et al.  
82 2000); and reproductive patterns (Cipollini and Whigham  
83 1994; Delph 1999). Moreover, sexes may differ in  
84 responses to herbivory, parasitism, and competitive ability  
85 affecting not only their spatial organization but also sex  
86 ratios (Agren et al. 1999; Graff et al. 2013). Optimization  
87 of resource use in space and time might be particularly  
88 important in resource-limiting environments where sexual  
89 dimorphism and sex spatial segregation would be expected  
90 to be greater (Gehring and Linhart 1993; Dawson and  
91 Geber 1999).

92 The dioecious perennial grass *Poa ligularis* Nees ap.  
93 Steudel, is widespread in across the aridity gradient of  
94 Patagonia (Argentina) and is one of the most preferred  
95 species for herbivores (Correa 1978; Ares et al. 1990). The  
96 aridity gradient in Patagonia is positively related to the  
97 absolute and relative shrub cover (Moreno et al. 2010).  
98 Previous studies in Patagonia reported differences between  
99 the spatial organization of males and females of this spe-  
100 cies which could be attributed to sexual dimorphism  
101 (Bertiller et al. 2002; Pazos et al. 2007; Graff et al. 2013).  
102 Moreover, other studies reported increased expression of  
103 mesophytic traits (i.e., N concentration in leaves, specific  
104 leaf area) and lowered chemical defenses in *P. ligularis*  
105 plants with increasing aridity/relative shrub cover across  
106 regional gradients, highlighting that these trends were  
107 opposite to those expected by aridity and consistent with  
108 enhanced microenvironments induced by shrub canopies  
109 (Moreno and Bertiller 2012). However, this study did not  
110 address whether sexes were dimorphic in these responses.  
111 Our objective was to analyze the effect of aridity/relative  
112 shrub cover, and vicinity to shrub patches on morpholog-  
113 ical and chemical traits of sexes of the dioecious perennial  
114 grass *P. ligularis* in patchy arid ecosystems in northern-  
115 central Patagonia. We hypothesized that sexes of *P. ligu-*  
116 *laris* have dimorphic responses in morpho-chemical traits

in relation to the environmental variation induced by 117  
aridity/relative shrub cover and vicinity to shrub patches. 118

## Materials and methods 119

### Species and study area 120

We selected the dioecious species *P. ligularis* which is a 121  
perennial grass species with a wide geographical distribu- 122  
tion in northern-central Patagonia, Argentina (Correa 123  
1978). We identified seven sites across a gradient of aridity 124  
(aridity index from 3.7 to 7.3) encompassing the distribu- 125  
tion area of *P. ligularis* (Bertiller et al. 2006; Moreno and 126  
Bertiller 2012). The aridity index (AI) of each site was 127  
taken from Moreno and Bertiller 2012 and was calculated 128  
as: 129

$$AI = PET/MAP,$$

where PET (mean annual potential evapotranspiration) 31  
=  $69.4 \times MAT$  (mean annual temperature) (UNESCO 132  
1979; Le Houérou 1990), and MAP is the mean annual pre- 133  
cipitation of each site. High AI values indicate high aridity. 134  
This aridity gradient was positively associated with a gradient 135  
of temperature, shrub cover (23–92 % of relative shrub cover 136  
estimated as the percentage of absolute shrub cover relative 137  
to the total plant cover), and shrub species richness (Moreno 138  
et al. 2010). The data of absolute and relative shrub cover were 139  
taken from Moreno and Bertiller (2012). The total and shrub 140  
species cover at each site were estimated using cover cate- 141  
gories with increments of 1 % within a representative vege- 142  
tation stand of about 1 ha (minimal area sensu Mueller- 143  
Dombois and Ellenberg 1974). This sampling followed the 144  
protocols used in previous patchy vegetation studies in arid 145  
Patagonia (Bertiller and Ares 2008; Moreno et al. 2010) 146  
characterized by a two-phase mosaic formed by shrubby plant 147  
patches (mean diameters ranging from 50 to more than 148  
100 cm) with high cover alternating with areas of sparse 149  
vegetation or bare soil (Aguar and Sala 1999; Soriano et al. 150  
1994; Bertiller et al. 2002). The percentages of species cover 151  
were estimated by at least two observed, in plots of ca. 152  
 $10 \times 10$  m within the sampling area (1 ha). The relative 153  
shrub cover (RSC) was calculated as the percentage of shrub 154  
cover relative to the total plant cover. After that, the sites were 155  
assigned to one of the following RSC groups: low 23–46 %; 156  
mid 47–69 %; high 70–92 %, which were also associated with 157  
three AI levels (low, less than 4.2; mid, 4.3–6.3; high, more 158  
than 6.4, respectively) (Table 1). 159

160 Sampling sites represent dominant vegetation units in  
161 the southern Monte Phytogeographical Province and in the  
162 Occidental, Central, and San Jorge Gulf Districts of the

**Table 1** Study site identification and Phytogeographical Province (*P* Patagonian, *M* Monte)-Phytogeographical District (*C* Central, *O* Occidental, *SJG* San Jorge Gulf) in parentheses, location in latitudes and altitudes, mean annual precipitation (MAP), mean

annual temperature (MAT), aridity index (AI), percentage of relative shrub cover (RSC) of each study site, and AI/RSC group (low, less than 4.2/23–46 %; mid, 4.3–6.3/47–69 %; high, more than 6.4/70–92 %), as in Fig. 1

| Site                       | Location                   | MAP   | MAT   | AI   | RSC % | AI/RSC group |
|----------------------------|----------------------------|-------|-------|------|-------|--------------|
| Gastre (P-O)               | 42°23'31"S, 68°56'41"W     | 150.0 | 7.95  | 3.68 | 23    | Low          |
| Pampa del Castillo (P-SJG) | 45°41'11.6"S, 67°53'26.6"W | 150.0 | 8.58  | 3.97 | 31    | Low          |
| Rio Mayo (P-O)             | 45°35'19.5"S, 70°20'14.8"W | 150.0 | 9.29  | 4.30 | 57    | Mid          |
| Rawson (P-C)               | 43°13'1.6"S, 65°0.1'29.1"W | 183.3 | 13.30 | 5.04 | 62    | Mid          |
| Telsen (M)                 | 42°33'33.5"S, 66°33'50.4"W | 137.5 | 12.76 | 6.44 | 84    | High         |
| Dique F. Ameguino (P-C)    | 43°44'40.4"S, 66°20'21.9"W | 130.0 | 12.38 | 6.61 | 89    | High         |
| Sierra Chata (M)           | 42°45'42.5"S, 66°0.1'2.5"W | 125.0 | 13.16 | 7.31 | 92    | High         |

165 Patagonian Phytogeographic Province. The vegetation in  
 166 the sites of the southern Monte Phytogeographical Province  
 167 had a random patchy structure (1.5–2.0 m tall), character-  
 168 ized by the shrubland of *Larrea divaricata* Cav. with  
 169 perennial grass species *Nassella tenuis* (Phil.) Barkworth,  
 170 *P. ligularis* and *Pappostipa speciosa* (Trin. & Rupr.  
 171 Romasch) (Soriano 1950; Ares et al. 1990; Bisigato and  
 172 Bertiller 1997; León et al. 1998; Mazzarino et al. 1998).  
 173 The sites of the Occidental District were characterized by a  
 174 patchy grass-shrub steppe (0.6–1.8 m tall) dominated by *P.*  
 175 *ligularis*, *Jarava* spp., *Senecio filaginoides* De Candolle,  
 176 *Adesmia volkmannii* Phil., and *Mulinum spinosum* (Cav.)  
 177 Pers. (Ares et al. 1990; León et al. 1998). The sites of the  
 178 Central District were characterized by a patchy shrubby  
 179 steppe dominated by *Chquiraga avellanadae* Lorentz  
 180 (50–80 cm tall) with perennial grass species (*Pappostipa*  
 181 spp., *P. ligularis*, *Poa dusenii* Hackel in Dusén, *Hordeum*  
 182 *comosum* Prel, and *Bromus pictus* Hooker) (León et al.  
 183 1998). The vegetation of the site in the San Jorge Gulf  
 184 District (30–50 cm) was characterized by a grass-shrub  
 185 steppe dominated by *Festuca pallescens* (St-Yves) Parodi  
 186 and *P. ligularis* with scattered tall and medium shrubs (*A.*  
 187 *volckmannii*, *M. spinosum*, *Nardophyllum obtusifolium*  
 188 Hooker et Arnott, and *S. filaginoides*) (León et al. 1998).  
 189 All dominant shrubs have strong morphological and/or  
 190 chemical defenses (Moreno et al. 2010). All the study sites  
 191 have been grazed by low densities of wild ungulates and by  
 192 sheep since early 1900 (Ares et al. 1990; Baldi et al. 2004).

### 193 Sampling

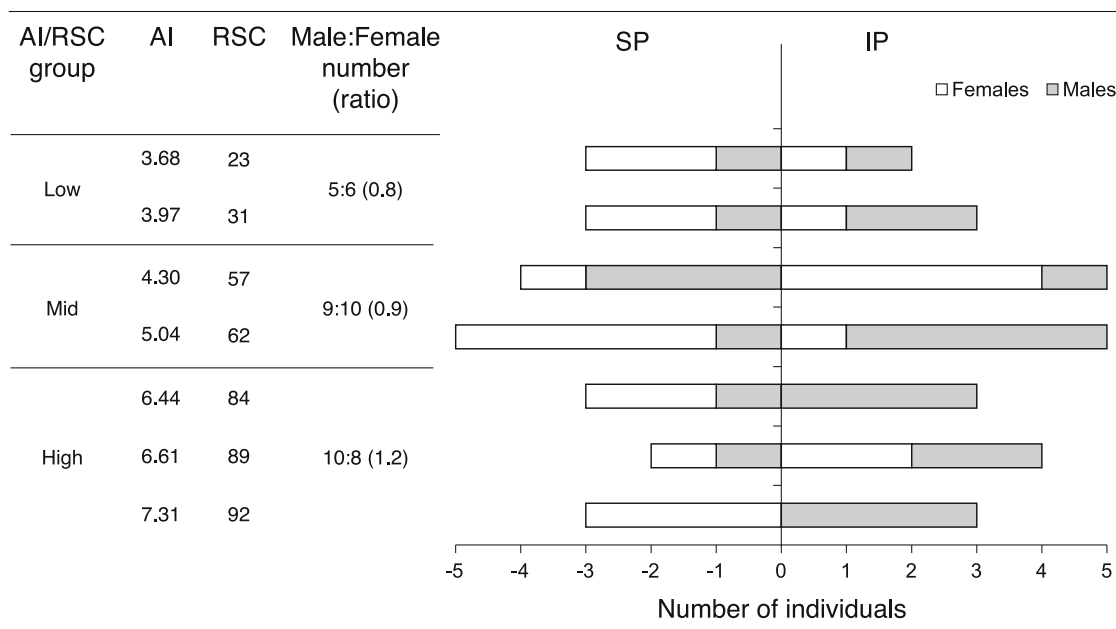
194 In this study, we reanalyzed data on attributes of *P. ligu-*  
 195 *laris* plants reported for the 7 sampling sites by Moreno  
 196 and Bertiller (2012) adding non published data on sex and  
 197 location with respect to shrub patches of each plant.  
 198 Sampling was carried out in November–December 2007,  
 199 during the transition from vegetative to early reproductive  
 200 phenological stages. Plants of *P. ligularis* were randomly  
 201 collected at each site within a representative vegetation

stand of about 1 ha (minimal area sensu Mueller-Dombois 202  
 and Ellenberg 1974) at each site (Moreno et al. 2010). We 203  
 registered the sex (male, female) of each randomly selected 204  
 plant and its location with respect to shrub patches (SP: *P.* 205  
*ligularis* plants located at the edge or underneath shrub 206  
 patches, and IP: *P. ligularis* plants located at inter-patch 207  
 bare areas). The number of collected plants varied between 208  
 sites (from 5 to 10) depending on plant size. This allowed 209  
 us to ensure a minimum number of plants of each sex 210  
 ( $n = 5$ ) with enough green leaf biomass to perform each 211  
 chemical analysis and a balanced number of males and 212  
 females per AI/RSC group (Fig. 1). We define a ‘shrub 213  
 patch’ as a group of perennial plant species, including at 214  
 least one shrub life form, such that the projection of its 215  
 canopy over the soil was separated from neighbor patches 216  
 by at least 15 cm of bare ground. In those plants collected 217  
 at vegetative stage, some tillers were separated, trans- 218  
 planted, and followed until flowering. This procedure 219  
 allowed us to assess the sex in most vegetative plants. It 220  
 should be noted that this species does not have labile sex 221  
 expression (Bertiller et al. 2000, 2002; Graff et al. 2013). 222  
 This sampling effort resulted in different number of 223  
 females and males per site although the total number of 224  
 plants of per sex was the same (24) across sites (Fig. 1). 225

### Morphological and chemical traits 226

The collected plants were brought to the laboratory, we 227  
 measured morphological traits in all collected plants (24 228  
 plants of each sex), and then we took samples for chemical 229  
 analyses (chemical traits). Sample collection for the mea- 230  
 surement of morphological and chemical traits was per- 231  
 formed under protocols established in previous studies 232  
 (Cornelissen et al. 2003; Bertiller et al. 2006). Among 233  
 morphological traits, we assessed: (a) the height of vege- 234  
 tative tillers from the base up to the top of the longest leaf 235  
 in 3 tillers per plant, (b) the length, area and dry mass of the 236  
 blade of the youngest full expanded green leaf selected of 5 237  
 different tillers per plant, and (c) the specific blade area 238





**Fig. 1** Aridity-related shrub cover (AI/RSC) groups: low, less than 4.2/23–46 %; mid, 4.3–6.3/47–69 %; high, more than 6.4/70–92 %, aridity index (AI), relative shrub cover (RSC), male:female of *P.*

*ligularis* number (ratio) per AI/RSC group, and number of females (white bars) and males (black bars) per location (SP shrub patch, IP inter-patch) at each study site across the AI gradient

239 (SBA) of these blades as blade area per blade dry mass.  
 240 The length and area of leaf blades were measured on digi-  
 241 talized images. After that, blades were dried at 60 °C for  
 242 48 h and weighed to estimate the dry mass. Chemical  
 243 analyses were assessed in remainder full expanded green  
 244 blades without signs of deterioration (uniformity in the  
 245 coloration and absence of damage by herbivore or patho-  
 246 gens) of each plant dried at 60 °C for 48 h. We firstly  
 247 assessed N concentration by semi-micro Kjeldahl in indi-  
 248 vidual plants with sufficient green leaf biomass for this  
 249 analysis (24 males and 23 females). After that, we assessed  
 250 total soluble phenolics by the Folin-Ciocalteu method  
 251 using 50 % methanol as extract solution and tannic acid as  
 252 standard (Waterman and Mole 1994) in those plants with  
 253 enough remnant green leaf biomass for this analysis (19  
 254 males and 19 females). In all cases the minimum number of  
 255 plants per sex and AI/RSC group was 5.

## 256 Statistical analyses

257 The effects of the fixed factors location (SP, shrub patch;  
 258 IP, inter-patch), sex (male, female), and aridity (AI)-rela-  
 259 tive shrub cover (RSC) and their interactions on plant  
 260 morphological and chemical traits were evaluated by three  
 261 way ANOVA. We used means per plant for statistical  
 262 analyses. The variables blade area and specific blade area  
 263 were logarithmic (ln) transformed, to meet ANOVA  
 264 assumptions. The significance level was set at  $P \leq 0.05$   
 265 throughout the study. We used the statistical package SPSS

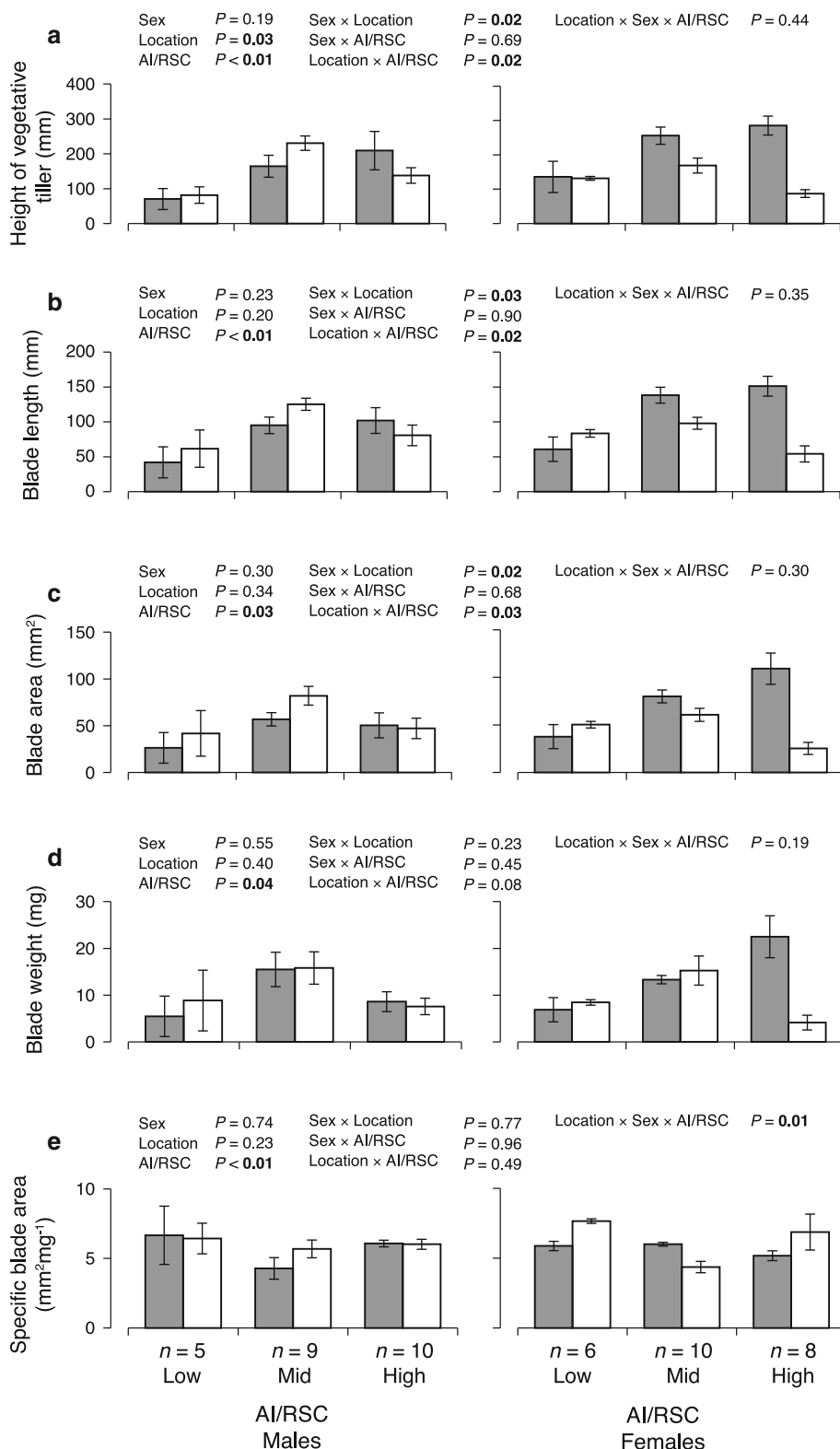
7.5 for Windows (Norusis 1997) to perform statistical 266  
 analyses. 267

## Results 268

### Variation of morphological traits of sexes with AI/RSC and location with respect to shrub patches 269 270

271 We found significant interaction effects for sex and loca-  
 272 tion, and location and AI/RSC on the height of vegetative  
 273 tillers ( $F_{1,36} = 5.22$ ;  $F_{2,36} = 4.37$ ), the blade length  
 274 ( $F_{1,36} = 4.66$ ;  $F_{2,36} = 4.29$ ), and the blade area  
 275 ( $F_{1,36} = 5.23$ ;  $F_{2,36} = 3.60$ ) of *P. ligularis* (Fig. 2a–c,  
 276 respectively). The values of these traits were higher in  
 277 females associated with shrub patches (SP) than in those at  
 278 inter-patch areas (IP), while height of vegetative tiller,  
 279 blade length, and blade area did not vary between SP and  
 280 IP locations in males. In both sexes, height of vegetative  
 281 tiller and blade length increased with increasing aridity in  
 282 SP, while in IP the highest values of both traits occurred at  
 283 mid values of AI/RSC. We find a significant effect of AI/  
 284 RSC on blade weight ( $F_{1,36} = 3.46$ ) being the largest  
 285 values of this trait at mid AI/RSC (Fig. 2d). The specific  
 286 blade area (SBA) varied in relation to the interaction of the  
 287 three factors ( $F_{2,36} = 5.20$ ) (Fig. 2e). SBA of females was  
 288 higher at SP than at IP at mid values of AI/RSC. Both AI/  
 289 RSC and location with respect to shrub patches, did not  
 290 affect SBA in males.

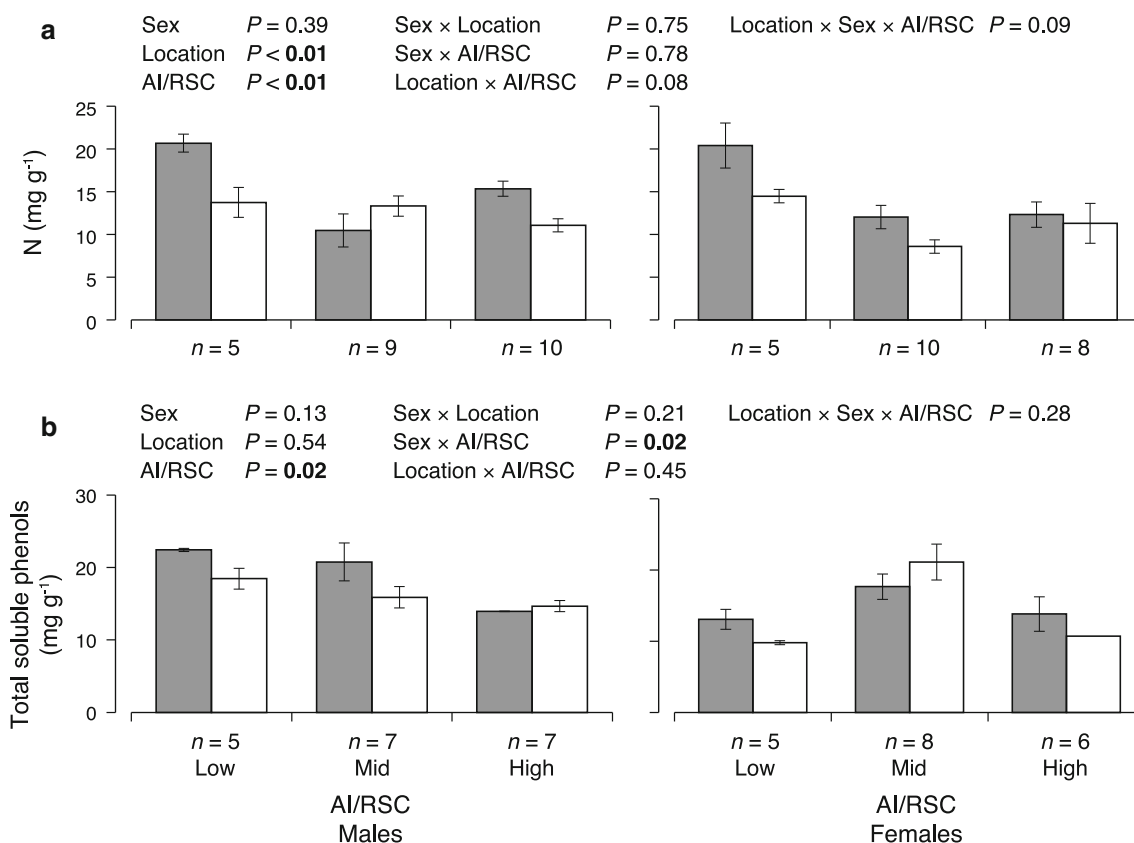
**Fig. 2** Mean values  $\pm$  SE of morphological traits (**a** height of vegetative tillers, **b** blade length, **c** blade area, **d** blade weight, **e** specific blade area) in males and females of *P. ligularis* at each location [*SP* shrub patch (*black bars*), *IP* inter-patch (*white bars*)], at each group of aridity index/relative shrub cover (AI/RSC): low, less than 4.2/23–46 %; mid, 4.3–6.3/47–69 %; high, more than 6.4/70–92 %. Results of ANOVA at the *top* of each panel and the total number of plants by AI/RSC group at the *bottom* of the figure



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**Fig. 3** Mean values  $\pm$  SE of chemical traits (**a** nitrogen, **b** total soluble phenols) in males and females of *P. ligularis* at each location [SP shrub patch (black bar), IP interpatch (white bar)], at each group of aridity index/relative shrub cover (AI/RSC): low, less than 4.2/23–46 %; mid, 4.3–6.3/47–69 %; high, more than 6.4/70–92 %.

Results of ANOVA at the top of each panel and the total number of plants by AI/RSC group at the bottom of the figure. The number of samples analyzed was 24 males and 23 females for N concentration and 19 males and 19 females for the total soluble phenolic analysis. Bars without error bars represent values of only one plant

## 291 Variation of chemical traits of sexes with AI/RSC 292 and location with respect to shrub patches

293 N concentration in green leaves of plants of *P. ligularis*  
294 decreased significantly with increasing AI/RSC  
295 ( $F_{2,35} = 11.04$ ) and this trait was higher in SP than in IP  
296 ( $F_{1,35} = 8.63$ ) (Fig. 3a). We found a significant combined  
297 effect of sex and AI/RSC on the total soluble phenolic  
298 concentration in green leaves ( $F_{2,27} = 4.29$ ). Phenolic  
299 concentration in males decreased with increasing AI/RSC,  
300 while in females this trait did not vary significantly in  
301 relation to location and AI/RSC (Fig. 3b).

## 302 Discussion

303 In Patagonia, as in other arid ecosystems, increasing aridity  
304 is associated with increasing shrub cover (Schlesinger et al.  
305 1990; Moreno et al. 2010). Fertile and sheltered microsites  
306 nearby well-defended shrub patches constitute refuges  
307 against water shortage and herbivores for herbaceous plants

with high growth rates and low structural and chemical 308  
defenses (Milchunas and Noy-Meir 2002; Adler et al. 309  
2004; Bertiller et al. 2002; Busso et al. 2012). Sexes of *P.* 310  
*ligularis* grew either associated with shrub patches or at the 311  
inter-patch areas across the aridity/relative shrub cover 312  
gradient and had dimorphic responses only in some mor- 313  
phological and chemical traits, thus partially supporting our 314  
hypothesis. Height of vegetative tillers, blade length, and 315  
blade area varied between sexes depending on the location 316  
with respect to shrub patches, while dimorphic variation of 317  
soluble phenolics in green leaves depended on the aridity/ 318  
relative shrub cover gradient. On the other hand, the 319  
specific blade area was the only trait influenced by the three 320  
factors (sex, aridity/relative shrub cover and location with 321  
respect to shrub patches). Dimorphic responses were 322  
opposite to those expected by aridity since in all cases trait 323  
mesophytism was the highest in areas with high aridity and 324  
relative shrub cover. These results are consistent with 325  
augmented offer of favorable microenvironments for plant 326  
growth created by shrub canopies (Adler et al. 2004; Ber- 327  
tiller et al. 2002). Shrubs patches in arid ecosystems induce 328

329 the formation of fertility islands, and provide sheltered  
 330 microsities and refuges against water shortage and herbi-  
 331 vores thus promoting the establishment and persistence of  
 332 herbaceous plants with high growth rates and low structural  
 333 and chemical defenses in their vicinity (Milchunas and  
 334 Noy-Meir 2002; Adler et al. 2004; Bertiller et al. 2002;  
 335 Rotundo et al. 2006; Golodets et al. 2009; Busso et al.  
 336 2012). Accordingly, the functional dimorphism of sexes  
 337 with respect to the shrubby matrix may allow them to  
 338 optimize the use of limiting resources in space and time  
 339 (Dawson and Geber 1999). In this sense, our results at the  
 340 patch scale also provided evidence of the positive effect of  
 341 shrub canopies on females which were more responsive  
 342 than males to variations in the microenvironment induced  
 343 for shrub patches. Females associated with shrub patches  
 344 showed more mesophytic traits (i.e., were taller and had  
 345 increased blade length and area) than those in inter-patch  
 346 areas at sites with mid and high values of aridity index and  
 347 relative shrub cover. This would indicate that females are  
 348 able to use microsities with abundant resources and refuges  
 349 to increase vegetative growth thus coping with their high  
 350 energetic costs of reproduction. In contrast, males with  
 351 lower reproductive costs than females were less responsive  
 352 to resource availability at the shrub patch scale as reported  
 353 elsewhere (Dawson and Ehleringer 1993; Eppley et al.  
 354 1998; Delph 1999; Bertiller et al. 2000; Obeso 2002;  
 355 Eppley 2005). Females showed also to be more responsive  
 356 in the specific blade area than males, to environmental  
 357 changes induced by shrub canopies, but in this case these  
 358 differences were only observed at mid values of aridity/  
 359 relative shrub cover. This could be associated with low  
 360 phenotypic plasticity of females at more predictable and  
 361 stable environments at the wettest and the driest sites (due  
 362 high precipitation and high offer of protected microsities,  
 363 respectively) and with increased phenotypic plasticity  
 364 under more unpredictable and unstable environmental  
 365 conditions (Valladares et al. 2006; Moreno and Bertiller  
 366 2015). These results on larger responsiveness of vegetative  
 367 traits in females than in males may be an evidence of the  
 368 functional dimorphism of *P. ligularis* sexes in relation to  
 369 resource and habitat use in patchy environments with a  
 370 wide variation in the resource spectrum (Gehring and  
 371 Linhart 1993; Dawson and Geber 1999) and are consistent  
 372 with higher competitive ability in females than in males of  
 373 *P. ligularis* (Bertiller et al. 2002).

374 On the other hand, our results showed that sexes were  
 375 dimorphic in relation to chemical defenses but, in this case,  
 376 males were more responsive than females. The concen-  
 377 tration of soluble phenolics in green leaves of males sig-  
 378 nificantly increased with decreasing aridity/relative shrub  
 379 cover while females maintained similar values of phenolic  
 380 concentration in blades across the aridity/relative shrub

cover gradient. Comparatively, the levels of soluble phe- 381  
 nolics in blades of females were lower than those of males 382  
 at low aridity/relative shrub cover and similar or slightly 383  
 higher than those in males with mid and high aridity/rela- 384  
 tive shrub cover values in accordance with those reported 385  
 by Graff et al. (2013) for similar environments. This con- 386  
 trast could be associated with differential requirements of 387  
 males and females. Females with higher reproductive costs 388  
 than males probably allocate more nutrients and carbohy- 389  
 drates to reproductive function than the latter (Agren et al. 390  
 1999) thus maintaining lower fixed levels of chemical 391  
 defenses across the aridity/relative shrub cover gradient. 392  
 This is consistent with the resource availability hypothesis 393  
 (Coley et al. 1985) since females usually occupy more 394  
 resource-rich microenvironments than males (Bertiller 395  
 et al. 2002). On the other hand, chemical defenses in males 396  
 with low reproductive costs were variable and decreased 397  
 with increasing levels of anti-herbivore defenses provided 398  
 the shrub matrix (Moreno et al. 2010) but probably also 399  
 with more favorable microenvironments provided by shrub 400  
 canopies as predicted by the resource availability hypoth- 401  
 esis. Thus, males were more protected by defensive com- 402  
 pounds in open environments with low shrub cover but 403  
 more favorable for pollen dispersal by wind (Obeso et al. 404  
 1998). 405

406 Furthermore, sexes did not show dimorphic responses in  
 407 N concentration in blades in relation to aridity/relative  
 408 shrub cover, or location with respect to shrub patches. Both  
 409 sexes of *P. ligularis* showed decreasing N concentration in  
 410 green leaves with increased aridity/shrub relative cover,  
 411 without differences between locations. This variation was  
 412 that expected by aridity and could be attributable to  
 413 decreasing N concentration in soil with increasing aridity  
 414 due to lower litter input, higher litter recalcitrance, and  
 415 lower soil microbial activity at the driest than at the wettest  
 416 sites (Zaady et al. 1996; Carrera and Bertiller 2010;  
 417 Tongway and Ludwig 2005; Moreno and Bertiller 2012).

418 In conclusion, our results highlighted dimorphic  
 419 responses to environmental variation between females and  
 420 males of *P. ligularis*. Morphological traits of females were  
 421 more responsive than males to environmental differences  
 422 induced by shrub patches. In this sense, the vicinity of  
 423 shrubs enhanced mesophytism of morphological traits of *P.*  
 424 *ligularis* females in accordance with increased plant per-  
 425 formance in favorable environments probably to cope with  
 426 high costs of reproduction. In contrast, males of *P. ligularis*  
 427 were more responsive than females in chemical traits  
 428 related to anti-herbivore defenses. This fact is consistent  
 429 with the requirement of anti-herbivore protection in open  
 430 areas favorable for pollen dispersal. Both female and male  
 431 responses were consistent with the resource availability  
 432 hypothesis.

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