

This is an Open Access document downloaded from ORCA, Cardiff University's institutional repository:<https://orca.cardiff.ac.uk/id/eprint/114028/>

This is the author's version of a work that was submitted to / accepted for publication.

Citation for final published version:

Retuerto, Rubén, Sanchez Vilas, Julia and Varga, Sandra 2018. Sexual dimorphism in response to stress. *Environmental and Experimental Botany* 146 , pp. 1-4.
10.1016/j.envexpbot.2017.12.006

Publishers page: <http://dx.doi.org/10.1016/j.envexpbot.2017.12.006>

Please note:

Changes made as a result of publishing processes such as copy-editing, formatting and page numbers may not be reflected in this version. For the definitive version of this publication, please refer to the published source. You are advised to consult the publisher's version if you wish to cite this paper.

This version is being made available in accordance with publisher policies. See <http://orca.cf.ac.uk/policies.html> for usage policies. Copyright and moral rights for publications made available in ORCA are retained by the copyright holders.



1 **EDITORIAL**

2 **Sexual dimorphism in response to stress**

3 Rubén Retuerto¹, Julia Sánchez Vilas², Sandra Varga³

4 ¹Universidade de Santiago de Compostela, Department of Functional Biology, Lope Gómez
5 de Marzoa s/n, 15782 Santiago de Compostela (Spain) ruben.reuerto@usc.es

6 ²Cardiff University, Organisms and Environment Division, Cardiff School of Biosciences,
7 The Sir Martin Evans Building, Museum Avenue, Cardiff, CF10 3AX (UK)
8 sanchezvilasj@cradiff.ac.uk

9 ³University of Lincoln, College of Science, School of Life Sciences, Joseph Banks
10 Laboratories, Green Lane, Lincoln, LN6 7TS (UK) svarga@lincoln.ac.uk

11

12 Sexual dimorphism in secondary traits (differences between the sexes in characteristics others
13 than the sexual organs) is widespread in plants (Geber and Dawson, 1999; Barrett and Hough
14 2013). Sex differences in ecological, morphological and physiological traits have been
15 commonly attributed to the different costs of reproduction associated with the male and the
16 female function (Reznick, 1985; Obeso, 2002) and are usually linked to trade-offs between
17 allocation to reproduction and to other plant functions (e.g., to growth and/or defence). Such
18 trade-offs are likely to be modified under more stressful conditions (Bazzaz and Grace,
19 1997). In fact, plants are continuously exposed to stressful biotic and abiotic environmental
20 factors during all their life cycle, which usually occur simultaneously (Suzuki et al., 2014).
21 Biotic factors include competitors, symbionts, parasites, pathogens, and herbivores. Abiotic
22 factors include extreme temperatures, water, light and nutrient availability, heavy metal, CO₂,
23 or UV radiation. As sessile organisms, plants have developed a wide range of responses to
24 cope with stress, ranging from escape to tolerance and avoidance (Lerner, 1999).

25 Understanding how environmental stress affects plant communities is especially
26 important in plants with sexually dimorphic breeding systems, as the sexes usually
27 experience the environmental pressures in a different way. Because the female function is
28 generally associated with a greater demand for resources, needed to produce both flowers and
29 seeds, female plants are usually assumed to perform worse than males under stress and male-
30 biased sex ratios are commonly found in more stressful environments (Barrett et al., 2010).
31 Importantly, highly skewed sex ratios as a consequence of sex specific responses could lead
32 to population declines and even extinction with potential effects on the structure and stability
33 of communities, which has been very rarely studied (Munné-Bosch, 2015; Hultine et al.,
34 2016; Petry et al., 2016). Likewise, sex-specific differences in stress tolerance and habitat-
35 related differences in performance put dimorphic species especially at risk due to the rapid
36 environmental changes underway. It is urgent to resolve uncertainties in order to increase our

37 ability to predict the extent to which the on-going environmental changes may impact
38 dimorphic species. The goal of this special issue was to expand our knowledge on sex-
39 specific responses to environmental stress in plants.

40 In the opening paper of this issue, Charlesworth (2017) highlights dioecious species
41 as interesting systems for studying the evolution of sexual dimorphism. Once separate sexes
42 evolve from hermaphroditism, adaptive divergence in traits of males and females can be
43 expected as the result of sex-specific selection pressures associated with their reproductive
44 function (Barrett and Hough, 2013). However, there are still many unresolved issues
45 concerning how and why trait differences between the sexes have evolved. Here,
46 Charlesworth (2017) discusses approaches for obtaining evidence for the occurrence of
47 adaptation after dioecy has evolved, and to what extent adaptive changes are sexually
48 antagonistic (having beneficial effects on one sex, but detrimental in the opposite sex).
49 Charlesworth (2017) suggests a number of interesting questions for future empirical work on
50 dioecious plants aimed at increasing our understanding of the genetics and evolution of
51 sexual dimorphism.

52 Combined with previous studies, evidence suggests that there is not yet a clear pattern
53 in the responses of the sexes to environmental stresses, and their differences seem to vary
54 depending on the species considered and the type of stress, demanding a greater range of
55 studies that allow generalisations (Juvany and Munné-Bosch, 2015). As emphasised by
56 Suzuki et al. (2014), recent studies have shown that the response of plants to multiple
57 simultaneous stresses cannot be extracted from the response to individual stress, highlighting
58 the need for more and larger studies encompassing multiple stress factors. In this Special
59 Issue, Strømme et al. (2017) and Canelles et al. (2017) consider this integrative approach and
60 incorporate climatic gradients into their studies. Global warming and associated climate
61 change are predicted to greatly influence the levels of Ultraviolet –B (UV-B) radiation at
62 Earth’s surface (Williamson et al., 2014). Strømme et al. (2017) consider the extent to which
63 the interaction between UV-B radiation and temperature, two factors rarely studied together,
64 can yield divergent responses between the sexes in the dioecious tree *Populus tremula*. They
65 study male and female plants grown along a natural temperature and UV-B gradient in
66 central Norway; reducing UV-B using specific screening filters and measuring effects on
67 growth, timing of terminal bud formation and bud break, carbon and nitrogen content and
68 concentrations of phenolic compounds. They found that elevation and UV-B radiation
69 affected concentrations of nitrogen and phenolics in stems and leaves. They also found that
70 increased elevation had a negative effect on plant growth and promoted autumnal bud
71 formation; but most importantly, the magnitude and direction of sexual differences depended
72 on elevation. Moreover, males were more sensitive to the effects of UV-B, with delayed bud
73 formation and enhanced growth at the highest elevation under the UV-B attenuation
74 treatment.

75 The relationship among reproduction patterns, floral dimorphism and environmental
76 stress is considered across a climatic gradient by Canelles et al. (2017). In their integrative
77 study on *Silene acaulis*, a widespread gynodioecious plant of boreo-alpine ecosystems, they
78 found that females showed larger ovary volumes and longer styles, but smaller corolla widths

79 and petal length than hermaphrodites. However, plant size and flower number per plant did
80 not differ significantly between sexes, although flower production was positively related to
81 plant size in both sexes and to fruit production in females, but not in hermaphrodites. The
82 study reveals a strategy in females to compensate for differences in reproductive investment,
83 and thus for the maintenance of gynodioecy, through higher fruiting production and other
84 components of fitness than hermaphrodites. Unexpectedly, environment severity was not
85 related to most of the biological traits studied, and only germination success was negatively
86 correlated with suitability index, regardless of sex. The authors suggest that the low overall
87 influence of climatic suitability in explaining differences in production may respond to
88 particularities of each population (e.g. interspecific competition, soil quality, pollinators,
89 etc.), and to the longevity of *S. acaulis*, which can attain well over 100 years. The hypothesis
90 that hermaphrodites could lose their female function in more severe climatic environments,
91 and therefore that gynodioecy may decline in favour of dioecy, was not clearly supported by
92 their results, although the authors claimed that more studies considering pollen quantity and
93 quality are needed to test this hypothesis.

94

95 The impact of climate change on performance of the sexes in the dioecious tree
96 *Populus tremula* is also the focus of the paper by Sobuj et al. (2017). They investigated the
97 single and combined effects of elevated temperature and CO₂ concentration on the growth
98 and carbon-based secondary metabolites synthesis in stem bark of male and female plantlets.
99 They observed that overall height growth in males was greater than in females. However,
100 under elevated temperature, height growth and diameter were considerably greater in females
101 than in males. From this result, the authors infer variation in optimum growth temperature
102 between females and males in *P. tremula*. Contrary to expectations that elevated CO₂ will
103 have no effect on growth, since the initial stimulation of CO₂ is not usually sustained through
104 time in broad-leaved trees (Vu et al., 2002), diameter incremented more in females than in
105 males in response to CO₂. However, the authors did not find effects of sex on biomass,
106 suggesting that the absence of a cost of reproduction in *P. tremula* seedlings might explain
107 the smaller intersexual differences in growth. Flavonoids, phenolic acids and salicylates are
108 important compounds in protecting plants against a wide range biotic and abiotic stress
109 (Lindroth and Clair, 2013; Julkunen-Tiitto et al., 2015). In line with this, Sobuj et al. (2017)
110 suggest that increased concentrations of these metabolites may provide a potential fitness
111 advantage in the face of environmental changes. In this respect, they found that females had a
112 higher total concentration of flavonoids in comparison with males. However, under elevated
113 CO₂, females reduced the synthesis whereas it was increased in males. Regarding phenolic
114 acids, the total concentration was greater in females than in males. In both sexes, the
115 synthesis of phenolic acids was reduced under elevated temperature, but it was increased
116 under elevated CO₂. The same trend was observed for salicylates, except that under elevated
117 CO₂ the excess synthesis only occurred in male plants. Overall, males tended to have higher
118 growth, while females tended to have greater concentrations of phenolics. This study
119 concludes that under scenarios with concomitant increases in CO₂ and temperature, the sexes
120 of *P. tremula* will probably grow more and accumulate lower levels of phenolics, although
121 the reported effects may be more pronounced in sexually mature plants. These three studies

122 highlight once more how the response of the sexes may be context-dependent and the need to
123 carry out further research including multiple interacting factors.

124 The profound effect that environmental stress can have on sex ratios in dioecious
125 plant populations is highlighted in the paper by Simancas et al. (2017). Whilst sex-lability is
126 common in plant species, the mechanisms underlying changes in sex expression are not well
127 understood. In their work, they found that when dioecious *Urtica dioica* plants were exposed
128 to phosphate starvation, males delayed their transition to flowering, and the sex ratio of the
129 experimental population shifted towards females. Seed production may be negatively affected
130 by this stress, and they hypothesised that the observed shift in sex ratio may serve as a
131 mechanism to compensate for seed production at the population level. In addition, their
132 experiment involved hormonal profiling that unravelled differences between the sexes in
133 response to phosphate starvation. In particular, they found that this stress increased the level
134 of compounds related to herbivory defence only in females. Stressful environmental
135 conditions may modify not only the number of flowers produced but also the sexual
136 expression of these flowers. In a contribution by Buide et al. (2017) demonstrated sexual
137 plasticity in gynodioecious-gynomonoecious *Silene littorea*, with a reduction in the total
138 amount of flowers produced in shaded plants but an increase in the proportion of female
139 flowers. Even though the physiological mechanisms were not investigated, plant hormones
140 together with resource availability are the probable causes.

141 Biased-sex ratios may impact, and be impacted, by interactions with other organisms
142 aboveground. Moritz et al. (2017) investigated the relatively well-established sex-biased
143 herbivory preference in dioecious willows using a large-scale approach. The authors created
144 experimental stands mimicking commercial plantation designs that contained either females,
145 males or even-sex mixes, and monitored herbivory damage by roe deer after one growth
146 season. Their results showed that mixed-sex plots were more preferred over mono-sex plots,
147 however they could not demonstrate an individual preference for one sex within mixed-sex
148 plots. The authors attributed these results to the differences in secondary metabolite profiles
149 between the sexes rather than the total content of secondary metabolites or nutrients. Studies
150 like this highlight the need for large-scale manipulative field experiments to evaluate
151 herbivory choice and to understand the mechanisms and consequences behind sex-biased
152 herbivory.

153 While the previous studies highlight the evolutionary forces driving sexual
154 dimorphism in response to stress, extra efforts should be directed towards elucidating the
155 molecular and physiological mechanisms underlying sex-specific responses. For example,
156 previous studies have reported that sexual dimorphic plants may have sex-specific
157 adaptations to heavy metal contamination, with most of these studies showing that males
158 exhibited a greater adaptive capacity than females when exposed to heavy metal stress (e.g.,
159 Chen et al., 2011; Han et al., 2013, but see Sanchez Vilas et al., 2016). In this Special Issue,
160 Qin et al. (2017), hypothesised that application of zinc, a metal that acts as a signal to
161 mediate the response to oxidative stress, could reduce lead-induced stress more in female
162 individuals than in males. To test this hypothesis, they examined sex differences in

163 morphological characteristics, biomass, gas exchange, cell ultrastructure and antioxidant
164 enzyme activity in saplings of a dioecious tree species widely planted as an important
165 economic species, the mulberry (*Morus alba*). Their study shows that sexes of mulberry
166 saplings differed in morphological traits, growth, gas exchange, enzyme activity, electrolyte
167 leakage and cellular ultrastructure in response to lead and zinc. Overall, lead stress negatively
168 affected females more than males, suggesting that males could have better detoxification
169 mechanisms and adaptation strategies in response to lead stress, and consequently, to be a
170 better candidate for phytoremediation. The application of zinc improves lead tolerance in
171 both sexes but, as hypothesised, the relieved was more pronounced in females than in males

172 We hope that the papers in this Special Issue motivate further research on the study of sexual
173 dimorphism in response to environmental stress. Given the increasing concerns over the
174 effects of global change on biological communities, there is a pressing need for further
175 research on the mechanisms underlying adaptive responses of plants to environmental
176 changes. Dioecious species are ideal comparative systems in which to investigate adaptation,
177 since they allow us to examine the functional significance of sexual dimorphism without the
178 confounding effects of differences in geography, history and ecology (Dawson and Bliss,
179 1989; Dawson and Geber, 1999). The rapid global changes underway will provide new
180 chances to study sex differences in adaptation. Changes in the climate will surely induce sex-
181 specific adjustments in the patterns of distribution of sexually dimorphic species, which will
182 move to new environments, disrupting their physiology, phenology and reproduction, as
183 testified by four of the papers published in this Special Issue.

184
185 An understanding of the basic mechanistic factors that underlay the responses of plants to
186 multiple stresses should benefit from integrative studies across multiple levels of biological
187 organisation, analysing transcriptional, metabolomic and physiological data. Plant biology
188 has become an increasingly data-rich field of research fuelled by the rapid advances in
189 analytical techniques. However, although the use of the “-omics” approaches (i.e.: genomic,
190 proteomic, metabolomic) could provide novel insights into the proximate mechanisms
191 responsible for sexual divergence in the responses to stress, they are still used insufficiently
192 in this field of research. Fortunately, five of the papers that appear in this Special Issue follow
193 this approach and use some of these promising tools for research on sex-related differences in
194 dioecious plants in response to stress.

195
196 The lack of general patterns emerging from the studies in this Special Issue are certainly
197 related to species-specific responses to stress and to the different nature (biotic and abiotic)
198 and intensities of the stresses studied, which could differentially modify the trade-offs
199 between allocation to reproduction and to other plant functions. Differences in the
200 reproductive status of the experimental plants (four studies in this Special Issue considered
201 pre-reproductive plants) could also explain the inconsistencies found, since intersexual
202 differences in growth, reproduction and physiology may be more pronounced after sexual
203 maturation (Dawson and Geber, 1999). Because sexes may differ in the timing of key
204 developmental stages (e.g., germination, flowering, senescence) (Lloyd and Webb, 1977) and
205 in the frequency and intensity of reproductive events, measurements at a single point in time

206 may not realistically reflect time-integrated patterns (Sanchez Vilas et al., 2012). Future
207 research would benefit from examining long-term differences in addition to short-term ones
208 for a thorough understanding of the quantitative relationships between male and female
209 reproductive efforts over the whole growing season, or even over the whole life-cycle of
210 perennial plants. Lastly, little is known on patterns of dimorphism in below-ground processes
211 and structures. Considering that sexual dimorphism would be the outcome of below versus
212 above processes (Varga et al., 2017), and allocation to above and below-ground sinks (Harris
213 and Pannell, 2008), studies that will focus on these aspects would be especially useful to
214 make a compelling story of sexual dimorphism in plants under stress.
215
216

217 ACKNOWLEDGEMENTS

218 We are grateful to Sergi Munné-Bosch for inviting us to guest edit this Special Issue and for
219 his help and support throughout. We are also indebted to the authors for their contributions
220 and to the many reviewers for their helpful constructive comments on the manuscripts.
221 Thanks also to John Pannell and Sergi Munné-Bosch for their comments on this manuscript.
222
223
224
225

226 References

- 227 Barrett, S. C. H., Hough, J., 2013. Sexual dimorphism in flowering plants. *J. Exp. Bot.* 64,
228 67–82.
- 229 Barrett, S.C.H., Yakimowski, S.B., Field, D.L., Pickup, M., 2010. Ecological genetics of sex
230 ratios in plant populations. *Phil. Trans. R. Soc. B.* 365, 2549e2557.
- 231 Bazzaz, F. A., Grace, J., 1997. *Plant Resource Allocation*. Academic Press.
- 232 Buide, M. L., del Valle, J. C., Castilla, A. R., Narbona, E., 2017. Sex expression variation in
233 response to shade in gynodioecious-gynomonoecious species: *Silene littorea*
234 decreases flower production and increases female flower proportion. *Env. Exp. Bot.*
235 XX, XX–XX.
- 236 Canelles, Q., Saura-Mas, S., Brotons, L., García, M. B., Lloret, F., Vilellas, J., Morris, W. F.,
237 2017. Environmental stress effects on reproduction and sexual dimorphism in the
238 gynodioecious species *Silene acaulis*. *Env. Exp. Bot.* XX, XX–XX.
- 239 Charlesworth, D., 2017. XXX. *Env. Exp. Bot.* XX, XX–XX.
- 240 Chen, L., Han, Y., Jiang, H., Korpelainen, H., Li, C., 2011. Nitrogen nutrient status induces
241 sexual differences in responses to cadmium in *Populus yunnanensis*. *J. Exp. Bot.* 62,
242 5037–5050.
- 243 Dawson, T.E., Bliss, L.C., 1989. Patterns of water use and the tissue water relations in the
244 dioecious shrub, *Salix arctica*: the physiological basis for habitat partitioning between
245 the sexes. *Oecologia* 79, 332-343.
- 246 Dawson, T.E., Geber, M.A., 1999. Sexual dimorphism in physiology and morphology. In:
247 Geber, M.A., Dawson, T.E., Delph, L.F. (Eds.), *Gender and Sexual Dimorphism in*
248 *Flowering Plants*. Springer, pp. 175-215.

- 249 Geber, M.A., Dawson, T E., Delph, L.F., 1999. Gender and Sexual Dimorphism in Flowering
250 Plants. Springer.
- 251 Han, Y., Wang, L., Zhang, X., Korpelainen, H., Li, C., 2013. Sexual differences in
252 photosynthetic activity, ultrastructure and phytoremediation potential of *Populus*
253 *cathayana* exposed to lead and drought. *Tree Physiol.* 33, 1043–1060.
- 254 Harris, M.S., Pannell, J.R., 2008. Roots, shoots and reproduction: sexual dimorphism in size
255 and costs of reproductive allocation in an annual herb. *Proc. R. Soc. B.* 275, 2595–
256 2602.
- 257 Hultine, K.R., Grady, K C., Wood, T.E., Shuster, S.M., Stella, J.C., 2016. Climate change
258 perils for dioecious plant species. *Nature Plants*, 16109.
- 259 Julkunen-Tiitto, R., Nenadis, N., Neugart, S., Robson, M., Agati, G., Vepsäläinen, J., Zipoli,
260 G., Nybakken, L., Winkler, B., Jansen, M.A.K., 2015. Assessing the response of plant
261 flavonoids to UV radiation: an overview of appropriate techniques. *Phytochem. Rev.*
262 14, 273–297.
- 263 Juvany, M., Munné-Bosch, S., 2015. Sex-related differences in stress tolerance in dioecious
264 plants: a critical appraisal in a physiological context. *J. Exp. Bot.* 66, 6083–6092.
- 265 Lerner, H.R., 1999. *Plant Responses to Environmental Stresses: from phytohormones to*
266 *genome reorganization.* CRC Press.
- 267 Lindroth, R.L., Clair, S.B.S., 2013. Adaptations of quaking aspen (*Populus tremuloides*
268 Michx.) for defense against herbivores. *For. Ecol. Manage.* 299, 14–21.
- 269 Lloyd, D.G., Webb, C.J., 1977. Secondary sex characters in plants. *Bot. Review* 43, 177–216.
- 270 Moritz, K. K., Parachnowitsch, A. L., Julkunen-Tiitto, R., Björkman, C., Ayres, M. P.,
271 Stenberg, J. A., 2017. Roe deer prefer mixed-sex willow stands over monosexual
272 stands but do not discriminate between male and female plants. *Env. Exp. Bot.* XX,
273 XX–XX.
- 274 Munné-Bosch, S., 2015. Sex ratios in dioecious plants in the framework of global change.
275 *Env. Exp. Bot.* 109, 99–102.
- 276 Obeso, J.-R., 2002. The costs of reproduction in plants. *New Phytol.* 155, 321–348.
- 277 Petry, W.K., Soule, J.D., Iler, A.M., Chicas-Mosier, A., Inouye, D.W., Miller, T.E.X.,
278 Mooney, K.A., 2016. Sex-specific responses to climate change in plants alter
279 population sex ratio and performance. *Science* 353, 69–71.
- 280 Qin, F., Liu, G., Huang, G., Dong, T., Liao, Y., Xu, X., 2017. Zinc application alleviates the
281 adverse effects of lead stress more in female *Morus alba* than in males. *Env. Exp.*
282 *Bot.* XX, XX–XX.
- 283 Reznick, D., 1985. Costs of reproduction: an evaluation of the empirical evidence. *Oikos* 44,
284 257–267.
- 285 Sánchez Vilas, J., Bermúdez, R., Retuerto, R., 2012. Soil water content and patterns of
286 allocation to below- and above-ground biomass in the sexes of the subdioecious plant
287 *Honckenya peploides*. *Ann. Bot.* 110, 839–848.
- 288 Sanchez Vilas, J., Campoy, J.G., Retuerto, R., 2016. Sex and heavy metals: Study of sexual
289 dimorphism in response to soil pollution. *Env. Exp Bot.* 126, 68–75.
- 290 Simancas B, Cotado A, Müller M, Munné-Bosch S., 2017. Phosphate starvation during the
291 transition phase increases the sex ratio and 12-*oxo*-phytodienoic acid contents in
292 females of *Urtica dioica*. *Env. Exp. Bot.* 145, 39–46.

293 Sobuj, N., Virjamo, V., Zhang, Y., Nybakken, L., Julkunen-Tiitto, R., 2017. Impacts of
294 elevated temperature and CO₂ concentration on growth and phenolics in the sexually
295 dimorphic *Populus tremula* (L.). *Env. Exp. Bot.* XX, XX–XX.
296 Strømme, C. B., Julkunen-Tiitto, R., Olsen, J. E., Nybakken, L. 2017. The dioecious *Populus*
297 *tremula* displays interactive effects of temperature and ultraviolet-B along a natural
298 gradient. *Env. Exp. Bot.* XX, XX–XX.
299 Suzuki, N., Rivero, R.M., Shulaev, V., Blumwald, R.M., 2014. Abiotic and biotic stress
300 combinations. *New Phytol.* 203, 32–43.
301 Varga, S., Vega-Frutis, R., Kytoviita, M.M., 2017. Competitive interactions are mediated in a
302 sex-specific manner by arbuscular mycorrhiza in *Antennaria dioica*. *Plant Biol.* 19,
303 217-226.
304 Vu, J.C.V., Newman, Y.C., Allen, L.H., Gallo-Meagher, M., Zhang, M.Q., 2002.
305 Photosynthetic acclimation of young sweet orange trees to elevated growth CO₂ and
306 temperature. *J. Plant Physiol.* 159, 147–157.
307 Williamson, C.E., Zepp, R.G., Lucas, R.M., Madronich, S., Austin, A.T., Ballaré, C.L.,
308 Norval, M., Sulzberger, B., Bais, A.F., McKenzie, R.L., Robinson, S.A., Häder, D.-P.,
309 Paul, N.D., Bornman, J.F., 2014. Solar ultraviolet radiation in a changing climate.
310 *Nat. Clim. Chang.* 4, 434–441.
311
312