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1 **Reproductive phenology of two co-occurring Neotropical mountain**
2 **grasslands**

3
4 Soizig Le Stradic^{1,2,3}, Elise Buisson¹, G. Wilson Fernandes² & Leonor Patrícia Cerdeira
5 Morellato³

6 ¹ UMR CNRS/IRD 7263/237 IMBE - Institut Méditerranéen de Biodiversité et d'Ecologie –
7 Université d'Avignon et des Pays de Vaucluse, IUT, Agroparc, BP 61207, 84 911 Avignon
8 cedex 9, France.

9 ² Ecologia Evolutiva & Biodiversidade / Instituto de Ciências Biológicas, Universidade Federal
10 de Minas Gerais, 30161-970 Belo Horizonte MG, CP 486, Brazil.

11 ³ UNESP Universidade Estadual Paulista, Instituto de Biociências, Departamento de Botânica,
12 Laboratorio de Fenologia, Rio Claro, São Paulo, Brazil

13 Corresponding author: Soizig Le Stradic email: Soizig.lestradic@gmail.com

14 **Abstract:**

15 Aim: Climate tends to explain phenological variations in tropical ecosystems. However, water
16 availability and nutrient content in soil strongly affect plant communities, especially those on
17 old, climatically-buffered, infertile landscapes (OCBILs), and may impact these ecosystems'
18 plant reproductive phenology over time. Here, we compare the reproductive phenology of
19 sandy and stony tropical mountain grasslands, two co-occurring herbaceous communities of the
20 *campo rupestre* OCBILs. We asked whether flowering, fruiting and dispersion are seasonal in
21 both grasslands, and whether these phenophases differ due to variation in soil properties. We
22 also asked whether the phenological strategies and the numbers of flowers and fruits differ
23 between these two grasslands as soil conditions vary.

24 Location: Serra do Cipó, Minas Gerais, Brazil.

25 Methods: The phenology of herbaceous species of sandy and stony grasslands was monitored
26 monthly over two consecutive years.

27 Results: Sandy and stony grasslands flowered and fruited all year long. We did not find a
28 distinctive seasonal pattern at the community level of either studied grassland. However,
29 flowering, fruiting and dissemination occurred in stony grasslands mainly during the rainy
30 season, while sandy grassland species flowered in both seasons and fruited and disseminated
31 mainly during the dry season, as observed in other Cerrado savanna vegetations. Flower and
32 fruit production was higher in sandy grasslands than in stony grasslands, which may be linked
33 to higher water retention in sandy grassland soils. In both communities, species of Cyperaceae,
34 Eriocaulaceae and Xyridaceae contributed the most to overall production, whereas Poaceae and
35 Velloziaceae, two important families in *campo rupestre*, barely participated in the
36 reproductive phenology during our two-year survey.

37 Conclusions: Despite a strong seasonal climate, there was no reproductive seasonal pattern at
38 the community level in *campo rupestre*. This first investigation of Neotropical grassland
39 phenology indicates that the differential soil content may constrain the grasslands' reproductive
40 phenology and restrict reproduction of stony grassland species to the most favorable season.
41 Further studies of grassland phenology are necessary to disentangle the relative importance of
42 soil, climate and other triggers, especially fire.

43

44 **Keywords:** Circular statistics, dissemination, flowering, fruiting, phenological strategy,
45 rupestrian grassland, seasonality, savanna.

46 **Nomenclature:** Brazilian Flora 2020 in construction. Rio de Janeiro Botanical Garden.
47 Available at: < <http://floradobrasil.jbrj.gov.br/> >. Accessed on: 04 Sep. 2017

48

49 **Running head:** Reproductive phenology of neotropical grasslands

50

51 **Introduction**

52 Plant phenology studies recurrent reproductive events such as flower budding,
53 flowering, fruiting, and seed germination, as well as vegetative events like leaf flushing and
54 shedding. These events link different groups within a community, such as decomposers,
55 herbivores, pollinators, or seed dispersers (Morellato et al. 2016). Climate is the main factor
56 controlling the timing of phenological events in plants, especially in seasonal environments
57 (Rathcke & Lacey 1985; Fenner 1998; Ramirez 2002; Morellato et al. 2013), where the dry
58 season limits the time available for growth, seed maturation and germination, and may decrease
59 the diversity of phenological strategies in plant communities (Sarmiento & Monasterio 1983;
60 Seghieri et al. 1995; Ramirez 2002; Batalha & Martins 2004).

61 Few studies have addressed the reproductive phenology of Neotropical grasslands, and
62 the effect of climate seasonality on savanna and grassland phenology is yet to be demonstrated
63 (Morellato et al. 2013; but see Rocha et al. 2016). In the Brazilian Cerrado savanna, most
64 herbaceous species flower late in the wet season; conversely, in Brazilian and Venezuelan
65 woody savannas, flowering occurs during the dry season (Monasterio & Sarmiento 1976;
66 Ramirez 2002; Batalha & Martins 2004; Tannus et al. 2006). Besides the presence of a dry
67 season, soil characteristics, such as water retention capacity or soil texture, are important
68 determinants of the vegetation structure of old growth tropical grasslands (Veldman et al. 2015;
69 Silveira et al. 2016). However, the relevance of soil as an environmental factor affecting plant
70 species phenology is still poorly understood (but see Borchert 1994; Cardoso et al. 2012).
71 Moreover, to our knowledge, no study has yet assessed whether soil type can affect plant
72 phenology in tropical grasslands. Phenology defines the temporal structure of a community,
73 including plant life cycles and interactions between plants and animals. Therefore,
74 understanding how the environment (i.e. climate and soil conditions) affects and/or modifies
75 phenology is fundamental, with applications that can improve management actions and guide
76 conservation practices and restoration strategies (Andersen et al. 2005; Morellato et al. 2016;
77 Buisson et al. 2017).

78 Old-growth grasslands are species-rich ecosystems that developed over many centuries.
79 Among these old-growth grasslands are the Brazilian “*campo rupestre*” (or rupestrian
80 grasslands), which are fire-prone mountain grasslands within the Cerrado domain (Silva &
81 Bates 2002) and are regarded as OCBILs (old, climatically-buffered, infertile landscapes)
82 (Silveira et al. 2016). The *campo rupestre* sustains a highly diverse vegetation composed of
83 numerous endemic species (Giulietti et al. 1997; Fernandes 2016; Silveira et al. 2016). We
84 focused our study on the mosaic of co-occurring stony and sandy grasslands (Le Stradic et al.
85 2015, see also Appendix S1) and the dominant herbaceous plant communities in *campo rupestre*

86 (Le Stradic et al. 2015). Under the same climate, soil condition and water availability are
87 thought to play a major role as environmental filters that favor stress-tolerant species in both of
88 these grasslands (Negreiros et al. 2014). Stony grasslands are often located on slopes and have
89 coarser soil, lower water retention and higher mineral nutrient concentration. Sandy grasslands
90 are located in flatter areas, and soils present higher water retention and lower mineral nutrient
91 concentration (Le Stradic et al. 2015). Each grassland soil's distinct properties result in the
92 filtering of distinct functional trait values (Negreiros et al. 2014). In contrast to sandy
93 grasslands, the plants in stony grasslands present narrower leaves, higher stress tolerance, lower
94 competitiveness and higher sclerophylly, which are all traits associated with drier soil
95 conditions related to lower water retention. Therefore, we hypothesize that the phenological
96 strategies and flower and fruit production should also differ between the two grasslands.

97 Here, we examine co-occurring herbaceous communities of stony and sandy grasslands
98 under the same climate to determine whether their reproductive phenology varies due to
99 different soil structure and composition. We address the following questions: (i) Do the plant
100 communities of these two mountain grasslands present seasonal reproductive (flower, fruit,
101 dispersion) phenological patterns? Do the phenological patterns differ between grassland types?
102 To assess the seasonality of the two grasslands, we applied circular analysis as proposed by
103 Morellato et al. (2010). We expected reproductive patterns to be seasonal, with a flower peak
104 during the rainy season and a fruiting peak towards the end of the rainy season. This strategy
105 has been observed in other grasslands and savannas under tropical seasonal climate. (ii) Do the
106 phenological strategies differ between stony and sandy grasslands? To define the phenological
107 strategies of each plant species, we considered frequency, duration, timing and amplitude
108 (Newstrom et al. 1994) of three phenophases: flowering, fruiting and dissemination. We
109 expected stony grassland communities to present short to medium seasonal flowering and
110 fruiting strategies restricted to the rainy season due to lower water retention in soils, while sandy
111 grassland would show a larger suite of reproductive strategies, as its soil presents higher water
112 retention throughout the year. Finally, to precisely evaluate the differences in phenological
113 response between these two grasslands, we assessed not only the phenological strategies but
114 also the flower and fruit production, addressing the question: (iii) Do the numbers of flowers
115 and fruits produced differ between the two grassland types? The harsher soil conditions should
116 limit flower and fruit production in stony grasslands, so we expected flower and fruit production
117 to be higher in sandy grasslands.

118 **Material & Methods**

119 *Study area*

120 Our study area is located in southeastern Brazil, in the southern portion of the Espinhaço
121 Mountain Range, within the Morro da Pedreira Environmental Protection Area, a buffer zone
122 of the Serra do Cipó National Park (state of Minas Gerais). *Campo rupestre* is the main
123 vegetation formation in the Espinhaço Mountains (Silveira et al. 2016). The climate is seasonal,
124 with a cold dry season from April to September and a warm rainy season from October to March
125 (Appendix S1). The mean annual precipitation is 1,622 mm, and the mean annual temperature
126 is 21.2 °C (Madeira & Fernandes 1999). We observed transitional seasons between the two
127 main seasons, from dry to rainy and from rainy to dry, defined as September-October and
128 March-April, respectively (Appendix S1).

129 Sandy and stony grasslands of *campo rupestre* are species-rich vegetations dominated
130 by Poaceae, Cyperaceae, Xyridaceae, Eriocaulaceae and some Velloziaceae, as well as forb and
131 sub-shrub species belonging to Asteraceae or Melastomataceae. Stony grasslands are
132 characterized by the presence of *Vellozia* spp., which are strongly associated with dry

133 environments (Porembski & Barthlott 2000). In sandy grasslands, Poaceae and Cyperaceae are
134 predominant (Appendix S2, Le Stradic et al. 2015). The soil in stony grasslands is coarser, with
135 a high density of quartzic stones, and retains less water than sandy grassland soil (Le Stradic et
136 al. 2015). The total N, P, K, Ca²⁺, Mg²⁺ concentrations and C_{org} content are significantly higher,
137 and the soil more acidic, in stony grasslands than in sandy grasslands. In both grasslands, P and
138 C_{org} content and pH vary seasonally: during the dry season, P concentrations are significantly
139 higher, while C_{org} contents and pH are significantly lower (Le Stradic et al. 2015). Fire is a
140 driver of vegetation dynamics in *campo rupestre*. In the study area, fires usually occur during
141 the transition from dry to rainy season (September-October), with low to moderate fire
142 frequency (Figueira et al. 2016, Alvarado et al. 2017).

143 *Plant survey*

144 To study the phenological patterns of the two main types of *campo rupestre* grassland,
145 we selected five locations containing each of the two types, totaling 10 grassland sites: 5 sandy
146 and 5 stony. Each pair of grasslands were less than 100 m apart. Each location covered between
147 0.23 ha to 2.36 ha; lay between 1,091 m and 1,310 m altitude; presented slopes between 4% to
148 17%; had a different fire history; and was not grazed (Appendix S3). Together, the 10 sites
149 spread over an area covering around 560 ha of *campo rupestre*. At each of these ten sites, we
150 randomly set up 10 permanent quadrats of 1 m² each, totaling 100 permanent quadrats (50 on
151 sandy grasslands and 50 on stony grasslands). A detailed floristic survey was carried out in
152 these 10 sites in 2009 (Le Stradic et al. 2015), and the phenological survey was performed
153 monthly in the same quadrats, during two consecutive years, from November 2009 to October
154 2011. For each quadrat, we recorded all species, and for each species we counted (1) the total
155 number of individuals or clumps (i.e. cluster of the same species, probably clones) and (2) the
156 number of all reproductive stems in each phenophase (i.e. observable stage or phase in the
157 annual life cycle of a plant): (i) flower (including flower buds and open flowers); (ii) fruit
158 (including unripe and ripe fruits); and (iii) dissemination (open fruits with seed dispersal, or
159 empty fruits following a month with fruit production). Thereafter, the number of reproductive
160 stems presenting flowers, fruits or dissemination signs were designated by number of flowers,
161 number of fruits and number of dissemination signs, respectively.

162 *Phenological strategy analysis*

163 We adapted the classification of Newstrom et al. (1994)—which considers frequency,
164 timing, amplitude (numbers of flowers, fruits, and dissemination signs), and the duration of
165 each phenophase—to describe and classify the phenological strategies of each species occurring
166 on sandy and stony grasslands. We classified each species' phenology according to five
167 phenological strategies (adapted from Newstrom et al. 1994): Continuous (C), phenological
168 events present continuously over the year; Irregular/episodic (I), irregular phenological events
169 over the year; Short (SS), phenophase lasting up to two months; Medium (SM), phenophase
170 lasting from two to six months; Extended (SE), phenophase lasting more than six months but
171 less than one year (Appendix S4). SS, SM and SE are seasonal strategies. For each species, we
172 defined three periods of occurrence for each phenological strategy: (R) or rainy season (from
173 October to March), (D) or dry season (from April to September), and (T) or the dry-to-rainy
174 and rainy-to-dry transitional seasons, as defined above.

175 *Flower and fruit production*

176 For each grassland type, all reproductive stems with flower or fruit in each quadrat,
177 hereafter referred to as flower and fruit production, were counted and further analyzed for the
178 seven most important families in our grasslands, according to the survey performed by Le
179 Stradic (2015): Asteraceae, Cyperaceae, Eriocaulaceae, Melastomataceae, Poaceae,

180 Velloziaceae and Xyridaceae. The remaining species were classified as other forbs or other sub-
181 shrubs, because the families were represented by few species.

182 *Statistical analyses*

183 To characterize the seasonality of flowering, fruiting and dissemination in stony and
184 sandy grasslands, we applied circular statistical analyses as described by Morellato et al. (2010),
185 using the total number of species at the peak of flowering, fruiting and dissemination per month.
186 For each phenophase, the number of species exhibiting a given phenological event on the peak
187 date (i.e. the date when the highest number of flowers, fruits or dissemination signs was
188 observed) of each month was treated as a circular frequency distribution, with data grouped at
189 30° intervals (30° = interval between two months) and January as the starting point (15°). The
190 mean angle μ represents the mean date of the flowering, fruiting or dissemination period, and r
191 is the measure of the concentration of the circular distribution of frequencies around the mean
192 angle. Values of r range from 0 (when data are completely dispersed in all angles) to 1 (when
193 all the data are concentrated around one angle or date) (Zar 1996). The Rayleigh test (i.e. test
194 of non-uniformity) assesses the null hypothesis, i.e. there is no sample mean direction of a set
195 of points on a circle. A significant Rayleigh test indicates a significant mean angle or mean
196 date. When species are concentrated around the mean angle or date, the phenological pattern is
197 seasonal (Morellato et al. 2010). We could not compare significant mean angles or seasonal
198 phenological patterns between sandy and stony grasslands, because a significant mean angle
199 was never present for both grasslands for the same phenological event. Circular analyses were
200 performed using the software Oriana 3.0 (Kovach Computing Services 2012).

201 To describe the different phenological strategies encountered in *campo rupestre*, the numbers
202 of species associated with each strategy (i.e. Continuous, Irregular, Seasonal Short, Seasonal
203 Medium and Seasonal Extended) were analyzed using permutation tests performed in R
204 package «coin» (Zeileis et al. 2008). Permutation tests are a type of statistical significance test
205 that obtains the population distribution by calculating the sample statistics under every possible
206 permutation of the observed data points. The calculated statistic of the permutation tests is z
207 when one explanatory variable is tested, or maxT when two or three explanatory variables are
208 tested.

209 To analyze flower and fruit production at each site, according to the grassland type and plant
210 families, GLM procedures were performed assuming a Poisson distribution and a logarithmic
211 link function. The numbers of flowers and fruits were the response variables, while grassland
212 types and families were the categorical variables (Crawley 2007).

213 These analyses were performed with R version 3.2 (R Core Team. 2015. R: A language and
214 environment for statistical computing. Vienna, Austria).

215 **Results**

216 We recorded 146 species in sandy grasslands and 155 species in stony grasslands,
217 mainly perennial species (138 species or 94.5% in the stony grasslands, and 151 species or
218 97.4% in the sandy grasslands, Table 1, Appendix S5). In stony and sandy grasslands, 26.7%
219 and 34.1% of species, respectively, did not reproduce during the observation period (Table 1,
220 Appendix S6).

221 *Seasonality of flowering, fruiting and dissemination on stony and sandy grasslands.*

222 Stony and sandy grasslands presented species flowering, fruiting and disseminating all year
223 long, with few variations between seasons (Fig. 1, Appendix S1). Sandy grasslands presented

224 a significant seasonal mean flowering peak date in March ($Z=5.53$, $p<0.01$) and a seasonal
225 mean fruiting peak date in May ($Z=4.71$, $p<0.01$), but both r scores were low (0.26 and 0.24,
226 respectively, Fig. 1 a and c; Appendix S7), indicating a very low seasonality. Dissemination
227 time did not show a significant mean peak date (μ) or seasonality (Fig. 1 e, Appendix S7). Stony
228 grasslands did not show a significant seasonality for flowering or fruiting (Fig. 1 b, d, Appendix
229 S7) and showed only marginally significant mean dissemination peak dates in December
230 ($Z=3.26$, $p<0.05$, with $r=0.19$, Fig. 1 f, Appendix S7), also due to a barely unimodal distribution,
231 though peak activity intensified during the wet season for the three phenophases (Fig. 1 b, d, f).

232 *Phenological strategies in stony and sandy grasslands*

233 Sandy and stony grasslands showed no significant differences regarding the proportion of
234 phenological strategies for either flowering, fruiting or dissemination (Table 2). For all
235 phenophases, in both grasslands, less than 20% of species presented continuous or irregular
236 strategies (Table 2, Appendix S4 a, b), and over 80% of species showed seasonal strategies. In
237 both grasslands, about 80% of species showed seasonal short (SS, Appendix S4 c, d) to seasonal
238 medium (SM, Appendix S4 e, f) flowering strategies, while around 60% of species presented
239 seasonal medium (SM) to seasonal extended (SE, Appendix S4 g, h) fruiting strategies (Table
240 2). There was no significant difference among the three seasonal fruiting strategies in stony
241 grasslands (Table 2). For dissemination, between 82% and 79% of species had an SS or SM
242 strategy in both grasslands (Table 2).

243 SS species in both grasslands flowered and fruited mainly during the rainy season. SM species
244 in sandy grasslands flowered primarily during the dry season, but those in stony grasslands
245 flowered during both rainy and dry seasons (Fig. 2, Appendix S8). A large share of species
246 presented an SM fruiting strategy, occurring mostly during the dry season in sandy grasslands,
247 but in stony grasslands during the rainy season (Fig. 2, Appendix S8). Seasonal extended (SE)
248 species of both grasslands fruited during the dry season (Fig 2, Appendix S8). SS species in
249 sandy grasslands disseminated during both rainy and dry seasons, while SS species in stony
250 grasslands disseminated mainly in the rainy season (Fig. 2, Appendix S8). Conversely, SM
251 species in sandy grassland disseminated mostly in the dry season, while stony grasslands
252 species disseminated in both seasons (Fig. 2, Appendix S8); some SM species disseminated
253 also during the transitional seasons (Fig. 2, Appendix S8). SE species in both sandy and stony
254 grasslands disseminated in the dry season (Fig. 2, Appendix S8).

255 *Flower and fruit production*

256 The mean numbers of flowers and fruits produced per site were significantly higher in sandy
257 than in stony grasslands (227.7 ± 26.6 flowers/10 m² and 211.6 ± 12.48 flowers/10 m²
258 respectively, $z=-2.42$ $p=0.01$; 532.6 ± 45.9 fruits/10 m² and 464.8 ± 47.5 fruits/10 m²,
259 respectively, $z=-6.78$ $p<0.001$). Cyperaceae, Xyridaceae and Eriocaulaceae produced the
260 largest numbers of flowers and fruits in both grasslands (Table 3). Poaceae presented the highest
261 number of species in both grasslands but produced few seeds (Table 3). Asteraceae,
262 Velloziaceae, Xyridaceae and other sub-shrubs had higher flower and fruit production in stony
263 grasslands, while Cyperaceae, Poaceae, and other forbs had higher flower and fruit production
264 in sandy grasslands (Table 3).

265 **Discussion:**

266 **Although** *campo rupestre* is characterized by a strong seasonal climate, our results did
267 not show the expected distinct reproductive seasonal patterns at the community level. Both
268 grasslands produced flowers and fruits all year long (see also Rocha et al. 2016). We observed
269 only very low seasonality, and then only for the sandy grasslands. Most studies show that

270 climate seasonality influences tropical grassland phenology, triggering the flowering of
271 herbaceous species during mid to late rainy season (Batalha & Martins 2004; Tannus et al.
272 2006). We did not find the expected differences in the proportions of phenological strategies
273 between stony and sandy grasslands. Despite the lack of seasonality at the community level,
274 most species in both grassland types present seasonal strategies, whereas only small portions
275 present continuous and irregular phenological strategies.

276 Most species in both grasslands presented a short flowering peak during the rainy
277 season, as found in other seasonal grassland ecosystems in Venezuela and Cameroon
278 (Monasterio & Sarmiento 1976; Seghieri et al. 1995; Ramirez 2002) and in the Brazilian
279 Cerrado (Batalha & Martins 2004; Tannus et al. 2006; Munhoz & Felfili 2007). This
280 phenological pattern is often related to the rainy season's higher water availability and the
281 following dry season's acute water shortage, which are characteristic of a seasonal system.
282 Available water is crucial for herbs to complete their vegetative growth and accumulate the
283 carbohydrates that enable them to flower (Sarmiento & Monasterio 1983; Ramirez 2002;
284 Batalha & Martins 2004). In the studied sandy grasslands, flowering also occurred during the
285 dry season, probably because sandy grassland soils retain more water than the coarser stony
286 grassland soils. Biotic factors may also play an influential role in the reproductive phenology
287 of the studied grasslands. Most species had short or medium seasonal flowering strategies,
288 indicating that the specific flowering period coincides with not only the best environmental
289 conditions, but also the availability of biotic resources such as pollinators (Guerra et al. 2016;
290 Brito et al. 2017). The need for specific pollinators such as large bees, wasps and/or dipterans
291 (most active during the dry season) may favor a dry seasonal medium strategy (Freitas &
292 Sazima 2006, Carstensem et al, 2014, Appendix S2).

293 Phenological fruiting strategies differed between the two grasslands. In sandy
294 grasslands, fruiting occurred mainly during the dry season, with a higher number of species
295 disseminating during the dry and transitional seasons. Most species that fruited during the dry
296 season adopted an SM or SE fruiting strategy, depending on the time required for fruit
297 maturation and seed dispersal. Long fruiting seasons are already described for Cerrado
298 herbaceous and sub-shrub species: anemochoric and autochoric species produce and
299 disseminate fruits during the dry season, whereas zoochorous species do so most during the
300 rainy season (Batalha & Martins 2004; Munhoz & Felfili 2007; Camargo et al. 2013). In *campo*
301 *rupestre*, anemochory and autochory are the two main seed dispersal syndromes (Conceição et
302 al. 2007; Dutra et al. 2009). The end of the dry season is marked by stronger and more frequent
303 winds (Fernandes 2016) and is therefore the optimal period for fruit and seed dissemination. In
304 stony grasslands, more species produced fruits and disseminated during the rainy season than
305 the dry season, possibly related to seed dispersal by water and/or more favorable conditions for
306 germination (Silveira et al. 2012). No study has reported hydrochory (but see Trovó & Stützel
307 2011; Goldenberg et al. 2015), but the importance of water as a dispersal mechanism in *campo*
308 *rupestre* should not be underestimated: sandy grasslands are regularly flooded during the rainy
309 season, and sedge seeds are known to be buoyant (Leck & Schütz 2005).

310 Our results highlighted significant differences in flower and fruit production between
311 sandy and stony grasslands, with the expected higher flower and fruit production in sandy
312 grasslands. These differences may be explained by low water availability in stony compared to
313 sandy grasslands, but this hypothesis needs to be experimentally tested. In both grasslands, all
314 flowers and fruits were produced by around 10% of individuals from a limited number of
315 species. Cyperaceae, Eriocaulaceae and Xyridaceae species produced most of the flowers and
316 fruits in both grasslands, whereas important and species-rich families of *campo rupestre*, such
317 as Poaceae and Velloziaceaea (Le Stradic et al. 2015), barely reproduced during our two-year

318 phenological survey (see also Neves & Damasceno-Junior 2011). While this can be explained
319 as an irregular reproductive event (i.e. does not occur every year), we suggest that, as the *campo*
320 *rupestre* is a fire-prone environment, numerous species are resprouters (Le Stradic et al. 2015;
321 Le Stradic et al. 2017) and need fire as a stimulus to induce flowering and fruiting (Munhoz &
322 Felfili 2007; Conceição et al. 2013). Such a strategy would allow them to avoid years of sub-
323 optimal climate and expend more resources in optimal years (Venable 2007). Some studies
324 have pointed out that recurrent burnings are not favorable for sexual reproduction because it
325 reduces seed supply, even for species occurring in tropical fire-prone environments;
326 consequently, vegetative reproduction may increase under frequent fire regimes (Hoffmann
327 1998; Setterfield 2002), such as in *campo rupestre* (see Furley & Ratter 1988).

328 This is the first quantitative study examining the phenology of *campo rupestre*
329 herbaceous flora. Understanding variations in reproductive strategies requires further studies of
330 grassland phenology, and long-term phenology monitoring would better assess climate
331 influences on tropical mountain grasslands. To better comprehend the dynamics of fire-prone
332 tropical grasslands, future phenology studies should focus on patterns of flower and fruit
333 production driven by fire regimes. The small proportion of individuals participating in the
334 phenology of both grasslands implies a slow succession process, a potential limitation on the
335 natural regeneration of *campo rupestre* after strong disturbances, and a low capacity for
336 colonization of degraded areas by *campo rupestre* species. Hence, we must learn more about
337 limited seed source and the lack of clear phenology and reproductive strategies for producing
338 quality seeds, a strong constraint on the conservation and ecological restoration of *campo*
339 *rupestre* (Le Stradic et al. 2014; Buisson et al. 2017).

340

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Table 1. Total numbers and percentages (%) of species surveyed in the sandy and stony grasslands at Serra do Cipó (Minas Gerais, Brazil), classified as perennial and annual species and as presenting at least one or no reproductive events during the study (flower, fruit and/or dissemination).

	Sandy grasslands		Stony grasslands	
	Number of species	Number of individuals	Number of species	Number of individuals
Perennial	138 (94.5%)	29 100	151 (97.4%)	31 327
Annual	8 (5.5%)	127	4 (2.6%)	27
with reproductive event	107 (73.3%)	2,691 (9.2%)	89 (65.9%)	2,226 (7.1%)
without reproductive event	39 (26.7%)	26,409 (90.8%)	46 (34.1%)	29,101 (92.9%)
Total	146	29 227	155	31 354

Table 2. Distribution of species of sandy and stony grasslands, at Serra do Cipó (Minas Gerais, Brazil), according to the phenological strategies for phenological event and grassland type. Mean number (\pm standard error) of species and percentage of species (indicated in brackets) for each strategy, according to the total number of species for each grassland type. Phenological strategies: Continuous (C), Irregular (I), Seasonal Short (SS), Seasonal Medium (SM), Seasonal Extended (SE). For each phenological event, permutation tests were performed within each grassland type to compare occurrence of phenological strategies therein (maxT test). Letters indicate significant differences between strategies within each grassland type. Permutation tests were performed between grassland type (Z tests); * means p-value $P < 0.05$, otherwise tests were not significant.

	Strategies	Sandy grasslands	Stony grasslands	
Flowering	C	2.0 ± 0.4^a (4 %)	3.0 ± 0.3^a (7 %)	
	I	3.8 ± 0.7^a (8 %)	3.4 ± 0.7^a (8 %)	
	SS	20.0 ± 0.9^b (40 %)	19.6 ± 1.4^b (43 %)	
	SM	20.2 ± 1.6^b (40 %)	17.0 ± 2.3^b (37 %)	
	SE	4.0 ± 0.3^c (8 %)	2.2 ± 0.4^a (5 %)	*
	Permutation tests - differences between strategies		maxT = 2.96, p = 0.016	maxT = 3.23, p = 0.006
Fruiting	C	3.0 ± 0.4^a (7 %)	3.8 ± 0.4^a (9 %)	
	I	2.4 ± 0.5^a (5 %)	3.4 ± 0.7^a (9 %)	
	SS	9.6 ± 0.5^b (21 %)	9.4 ± 1.3^b (22 %)	
	SM	16.8 ± 0.7^c (36 %)	14.2 ± 1.8^b (33 %)	
	SE	14.2 ± 1.4^c (31 %)	11.2 ± 1.2^b (27 %)	
	Permutation tests - differences between strategies		maxT = 3.10, p = 0.002	maxT = 2.95, p = 0.01
Dissemination	C	-	-	
	I	3.2 ± 0.6^a (7 %)	2.6 ± 0.5^a (6 %)	
	SS	20.6 ± 0.9^b (42 %)	12.4 ± 1.5^b (28 %)	*
	SM	19.8 ± 1.4^b (40 %)	22.8 ± 1.8^c (51 %)	
	SE	5.6 ± 0.4^c (11 %)	6.4 ± 0.6^d (15 %)	
	Permutation tests - differences between strategies		maxT = 2.81, p = 0.007	maxT = 3.68, p = 0.001

Table 3. Flower and fruit production per site (i.e. average number of reproductive stems with flower or fruit per site and standard error), for the main families based on peak production, in sandy (Sa) and stony (St) grasslands at Serra do Cipó (Minas Gerais, Brazil). Results of the GLM procedures (family: Poisson, link: log) are indicated. Letters indicate significant differences between families among grassland types.

	Mean number of reproducing species / site		Flower production			Fruit production						
	Sa	St	Sa	St	GLM procedures	Sa	St	GLM procedures				
Asteraceae	3.4 ± 1.2	3.2 ± 0.4	7.6 ± 1.9	a	23.2 ± 8.4	a	8.44 ***	5.4 ± 1.5	a	8.7 ± 2.4	a	2.75 **
Cyperaceae	11.0 ± 0.4	10.6 ± 1.6	251.2 ± 25.6	b	158.1 ± 21.6	b	-14.42 ***	290.6 ± 29.6	b	170.8 ± 27.0	b	-17.43 ***
Eriocaulaceae	4.8 ± 0.5	5.6 ± 1.3	70.5 ± 14.8	c	68.9 ± 15.6	c	-0.42 ^{NS}	51.6 ± 12.5	c	49.1 ± 11.2	c	-0.78 ^{NS}
Other Forbs	9.4 ± 1.2	5.2 ± 1.4	22.5 ± 3.4	d	14 ± 3.2	d	-4.40 ***	8.5 ± 2.7	d	1.9 ± 0.5	d	-5.90 ***
Melastomataceae	2.4 ± 0.6	2.2 ± 0.5	20.2 ± 6.9	d	14 ± 5.9	d	-3.33 ***	33 ± 14.0	e	41.7 ± 15.4	e	3.17 ***
Poaceae	4.0 ± 0.6	4.4 ± 0.8	29.7 ± 6.7	e	14.8 ± 4.9	d	-6.92 ***	29.7 ± 8.8	e	9.1 ± 4.6	a	-9.87 ***
Other Sub-shrubs	4.0 ± 1.3	6.0 ± 0.5	13.1 ± 4.4	f	38.2 ± 14.8	e	10.57 ***	7.8 ± 2.1	d	37.9 ± 15.0	e	12.71 ***
Velloziaceae	0.8 ± 0.2	1.2 ± 0.4	0.5 ± 0.3	g	4.7 ± 3.7	f	4.76 ***	1.3 ± 0.6	f	11.9 ± 5.4	f	7.58 ***
Xyridaceae	11.0 ± 1.4	7.4 ± 1.8	157.2 ± 36.0	h	227.6 ± 28.1	g	11.28 ***	138.2 ± 31.3	g	177.6 ± 23.5	b	6.99 ***
GLM procedures			<i>p</i> >0.001		<i>p</i> >0.001			<i>p</i> >0.001		<i>p</i> >0.001		

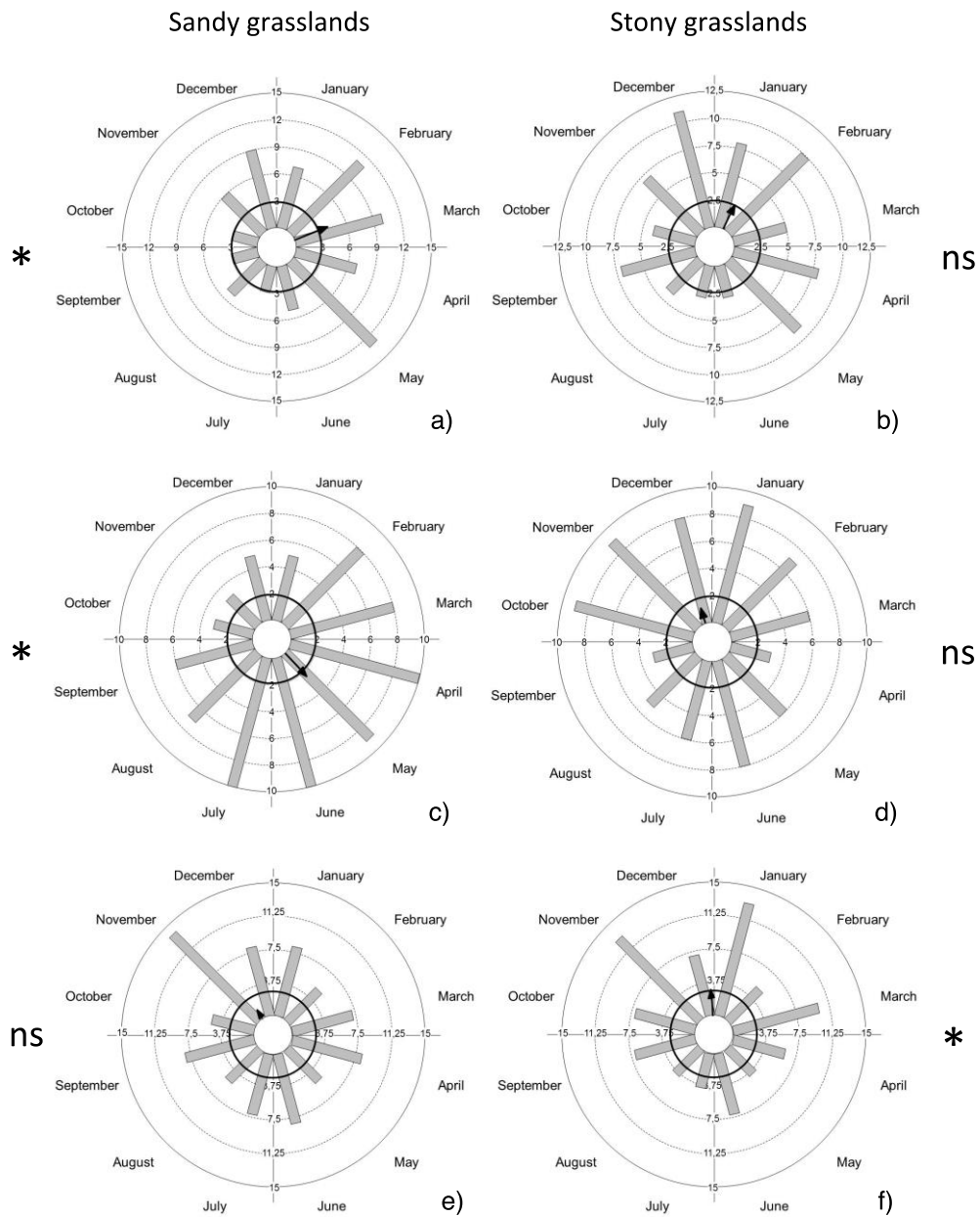


Fig. 1. Reproductive phenology of sandy (left) and stony (right) grasslands at Serra do Cipó (Minas Gerais, Brazil). Total monthly number of species at peak of flowering (a) (b), fruiting (c) (d), and dissemination (e) and (f) are presented. Arrows point to the mean date or mean angle (μ); the length of the arrow indicates the concentration around the mean, i.e. the degree of seasonality; and the black circle is the significant threshold. Results of Rayleigh test are indicated for each graphic: ns = non-significant and * = p value < 0.05. All results of statistical tests are available in Appendix S7

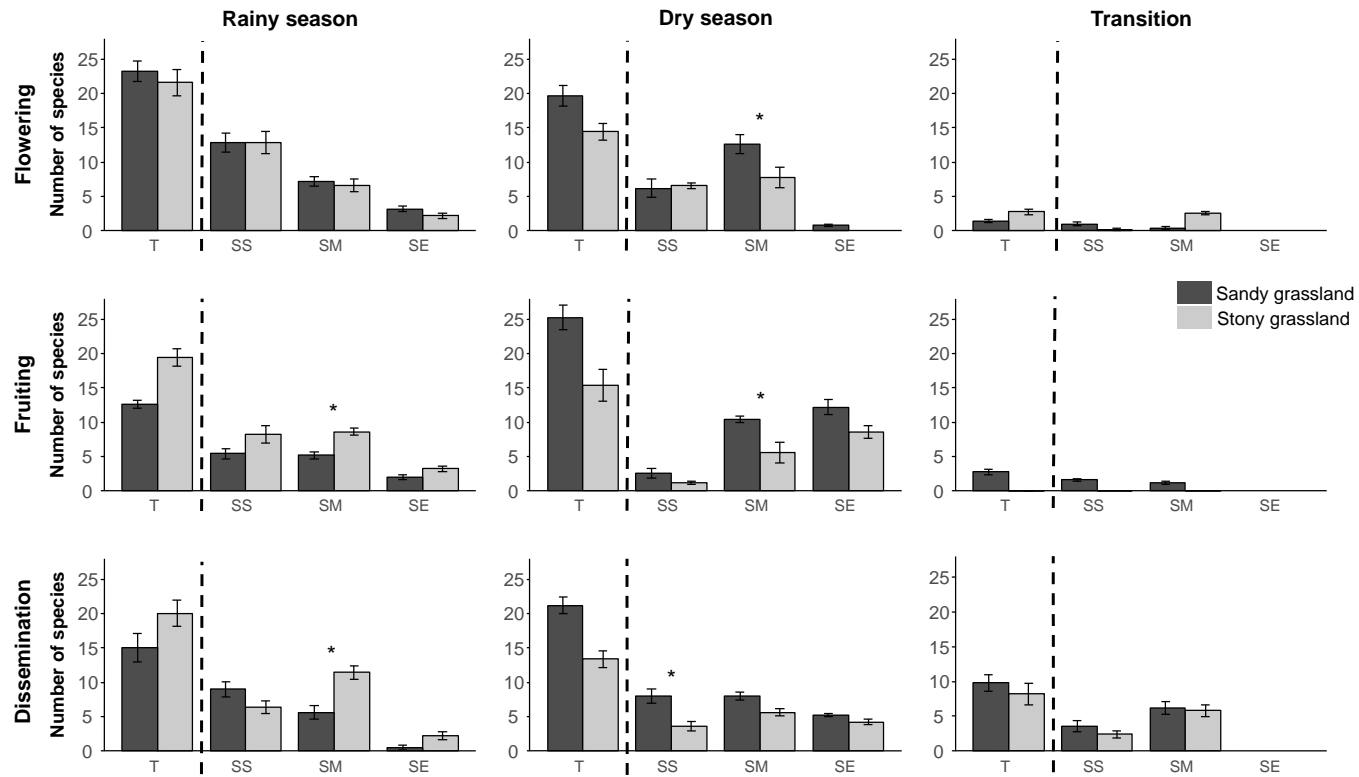


Fig. 2: Mean number of species per phenological event (flowering, fruiting, dissemination) according to the seasons—rainy season, dry season and transition—for the two grassland types: sandy grassland and stony grassland, at Serra do Cipó (Minas Gerais, Brazil). The total mean number of species (T) per phenological event is represented first and is then decomposed according to the three seasonal phenological strategies: Short (SS), Medium (SM), Extended (SE) seasonal strategy. * indicates significant difference with $p < 0.05$ between sandy and stony grasslands. A detailed table of all statistical test results is available in Appendix S8.

Supporting Information to the paper Le Stradic S. et al. Reproductive phenology of two co-occurring Neotropical mountain grasslands. *Journal of Vegetation Science*. Appendix S1. Distribution of the mean monthly temperatures (°C) at 6:00am and 1:00pm, and cumulative rainfall (mm). Mean percentage of species with flowers, fruits and dissemination, per site in sandy grasslands and stony grasslands, from November 2009 to October 2011 at Serra do Cipó, MG.

Supporting Information to the paper Le Stradic S. et al. Reproductive phenology of two co-occurring Neotropical mountain grasslands. *Journal of Vegetation Science*. Appendix S2. General view of *campo rupestre* at Serra do Cipó (Minas Gerais, Brazil), detailed pictures of sandy grasslands and stony grasslands, and pictures of some *campo rupestre* species from the most common families.

Supporting Information to the paper Le Stradic S. et al. Reproductive phenology of two co-occurring Neotropical mountain grasslands. *Journal of Vegetation Science*. Appendix S3. Main characteristics of each study site.

Supporting Information to the paper Le Stradic S. et al. Reproductive phenology of two co-occurring Neotropical mountain grasslands. *Journal of Vegetation Science*. Appendix S4. Examples illustrating the phenological strategies used in this study for some selected species occurring in sandy grasslands and stony grasslands at Serra do Cipó (Minas Gerais, Brazil).

Supporting Information to the paper Le Stradic S. et al. Reproductive phenology of two co-occurring Neotropical mountain grasslands. *Journal of Vegetation Science*. Appendix S5. List of species surveyed in the sandy and stony grasslands at Serra do Cipó (Minas Gerais, Brazil), showing reproductive activity in the period of study, along with their habitat of occurrence, the phenological strategy and the reproductive timing.

Supporting Information to the paper Le Stradic S. et al. Reproductive phenology of two co-occurring Neotropical mountain grasslands. *Journal of Vegetation Science*. Appendix S6. List of species surveyed in the sandy and stony grasslands at Serra do Cipó Minas Gerais, Brazil, that did not show reproductive activity in the period of study, along with their habitat of occurrence.

Supporting Information to the paper Le Stradic S. et al. Reproductive phenology of two co-occurring Neotropical mountain grasslands. *Journal of Vegetation Science*. Appendix S7. Results of circular statistical analyses on the flowering, fruiting and dissemination phenology of sandy and stony plant communities at Serra do Cipó (Minas Gerais, Brazil).

Supporting Information to the paper Le Stradic S. et al. Reproductive phenology of two co-occurring Neotropical mountain grasslands. *Journal of Vegetation Science*. Appendix S8. Mean number of species per phenological event (flowering, fruiting, dissemination) and seasonal phenological strategy, according the grassland types and timing at Serra do Cipó (Minas Gerais, Brazil).