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12 **Avoiding the dry season: dispersal time and syndrome mediate seed dormancy in**

13 **grasses in Neotropical savanna and wet grasslands**

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27   **Abstract**

28   **Question:** In seasonal rainfall systems, seed dormancy is a strategy to avoid

29   germination and seedling emergence in the dry season. Grass species in Brazilian

30   savannas (*Cerrado*) show variation in seed dispersal timing and mechanisms, and occur

31   in different habitat types (distinguished by soil moisture) within a seasonal rainfall

32   environment. However, it is unknown whether dormancy has evolved in these systems

33   as a dominant way in which germination is deferred, or how it correlates with other key

34   traits such as dispersal, where known trade-offs exist for avoiding competition. We

35   asked whether seed germination and dormancy vary with dispersal and abiotic factors in

36   savanna systems. Specifically, we assessed dormancy by comparing seeds: (1) from

37   species living in habitats with contrasting soil moisture during the dry season (open

38   savannas *versus* wet grasslands); (2) dispersed at different times (early in the wet

39   season, late in the wet season and in the dry season) and (3) showing alternate dispersal

40   syndromes (barochoric *versus* anemochoric).

41   **Location:** Open savannas and wet grasslands in Central Brazil.

42   **Methods:** We collected seeds of 29 grass species and tested viability and dormancy

43   using germination trials of fresh seeds, which was then repeated after dry storage of 3,

44   6, 9 and 12 months. Generalized Linear Mixed Models were used to test whether the

45   degree of dormancy was dependent on habitat type, seed dispersal time and seed-

46   dispersal syndrome.

47   **Results:** Seeds from wet grasslands lived longer and had consistently higher

48   germination rates than seeds from open savannas. Additionally, fresh seeds dispersed

49   late in the wet season had higher levels of seed dormancy compared to seeds dispersed

50   early in the wet season. Finally, we found that anemochoric seeds had lower levels of

51   dormancy than barochoric seeds.

52   **Conclusions:** Seed dormancy among Neotropical grasses was higher for seeds of

53   species from dry habitats, dispersed late in the wet season, and with short-distance

54 dispersal (barochory). These results suggest that seed dormancy is a key mechanism by

55 which seedlings avoid seedling emergence in the dry season, an effect offset by habitat

56 specific soil moisture availability. The trade-off between dormancy and seed dispersal

57 suggests that both strategies are costly and had non-additive benefits.

58 **Keywords:** *Cerrado*; Dry storage; Germination; Open savanna; Poaceae; Wet

59 grassland.

60 **Introduction**

61   Savannas are biomes composed of a continuous grass layer and scattered trees,

62 under a seasonal climate consisting of an annual cycle of wet and dry seasons (Scholes

63 & Archer 1997). The *Cerrado* is a Neotropical biome in Central Brazil dominated by

64 savannas, but also containing grasslands and forests. Soil moisture is one of the main

65 determinants of vegetation physiognomies; for example, rainforests are associated with

66 shaded valleys and riparian areas while wet grasslands occur in more open habitats

67 where the water table reaches close to the surface (Cianciaruso & Batalha 2008). In both

68 of these examples, soils are moist throughout the year. However, in savanna

69 physiognomies the water table is further below the soil surface (Rossatto et al. 2012),

70 leading to soils and the herbaceous layer drying out during the annual dry season. Grass

71 species within the *Cerrado* biome are mostly restricted to these open physiognomies of

72 savannas and grasslands due to their shade intolerance.

73   In savannas, rainfall seasonality is among the major limiting factors determining

74 seed germination and seedling establishment, due to its direct effects on water

75 availability in the soil (Jurado & Flores 2005; Baskin & Baskin 2014). Rainfall

76 seasonality is therefore likely to be a strong selective pressure acting on seed dormancy.

77 Indeed, seed dormancy has been found in a higher number of species in many seasonal

78 rainfall environments in comparison to less seasonal environments (Jurado & Flores

79 2005; Baskin & Baskin 2014). Grasses are one of the most highly represented families

80 (Poaceae) in the herbaceous layer of the *Cerrado* (around 600 species, Filgueiras et al.

81 2014), with species from wet grasslands and open savannas occurring under the same

82 seasonal macro-climate. However, in wet grasslands the water table is situated near the

83 surface throughout the year (Cianciaruso & Batalha 2008). Hence, the impact of rainfall

84 seasonality in the soil is buffered by the water table and the soil surface stays moist for

85 longer during the dry season (Cianciaruso & Batalha 2008). Subsequently, if seed

86 dormancy is a strategy of species from seasonal environments to deal with water

87 shortage, this selective pressure would be weaker in wet grasslands. Thus, we would

88 expect a greater seed dormancy among seeds of species occurring in open savannas in

89 comparison to seeds of species occurring in wet grasslands.

90 　　　Grass flowering and fruiting follow a seasonal pattern restricted to the wet

91 season in open savannas and wet grasslands (Munhoz & Felfili 2007; Ramos et al.

92 2014), presumably due to resource availability (i.e. water) to allow reproduction.

93 However, there is considerable variation in the timing of seed dispersal among grass

94 species (Munhoz & Felfili 2007; Ramos et al. 2014), with some species dispersing early

95 in the wet season, others dispersing later, and some species dispersing seeds even during

96 the dry season. Seeds dispersed early in the wet season would have the entire growing

97 season to germinate and recruit, whereas seeds dispersed late in the wet season, or

98 during the dry season (when low amounts of rain usually occurs in Brazilian savannas),

99 would very probably face extreme water shortages and low relative air humidity, which

100 potentially could be a risky strategy for seedling survival. Avoiding germination prior to

101 and during the dry season would be a key strategy for reducing the probability of

102 seedling mortality during the dry season, and seed dormancy would therefore play an

103 important role in delaying seed germination to the next wet season. Consequently, we

104 would expect greater seed dormancy among seeds dispersed late in the wet season, and

105 during the dry season, in comparison to seeds dispersed early in the wet season.

106 Moreover, to synchronize germination in the next wet season, we would expect dormant

107 seeds to maintain viability and overcome dormancy through dry-storage.

108 　　　While the season of seed release may be an important driver for variation in

109 dormancy, dispersal also plays a critical role determining the level of competition faced

110 by emerging seedlings. In broad terms, seed dormancy can reduce extinction risk by

111 spreading germination over time (Bulmer 1984), while seed dispersal can reduce risk by

112 spreading seeds and, consequently, germination over space (Venable & Brown 1988).

113 Parent-offspring (Ellner 1986) and sibling competition can significantly reduce plant

114 fitness (Satterthwaite 2010; Baskin & Baskin 2014; Saatkamp et al. 2014). Both seed

115 dormancy and dispersal are costly strategies. Thus, a trade-off between seed dormancy

116 and dispersal is often reported between these two bet-hedging traits (Venable and

117 Brown 1988). Theoretical studies in particular have reported that mean dispersal

118 distance decreases with increasing dormancy (e.g. Venable & Lawlor 1980; Cohen &

119 Levin 1991), although this relationship is not always supported where positive temporal

120 correlations in environment exist (Snyder 2006). Nevertheless, few empirical

121 assessments of the trade-off between dormancy and dispersal have been made (Rees

122 1993).

123   The diaspore morphology of savanna grass species is highly variable. Among

124 several dispersal syndromes, seeds can be characterized by structures which facilitate

125 wind dispersal (anemochory), such as winged bracts, hairy bracts and hairy rachis.

126 These structures can be completely absent and dispersal is therefore likely to occur

127 through gravity (barochory) (Ernst et al. 1992). Barochoric seeds fall near the mother-

128 plant, while anemochoric seeds have the opportunity to disperse far from the mother-

129 plant. Anemochory could therefore be favoured to deal with spatial unpredictability and

130 competition. Hence, we would expect to find a greater seed dormancy among

131 barochoric seeds in comparison to anemochoric seeds if there was evidence of a

132 dispersal-dormancy trade-off.

133   Seeds of grass species can be non-dormant or physiologically dormant (Baskin

134 & Baskin 1998), but the occurrence and mechanisms of seed dormancy in grass species

135 from Brazilian ecosystems are poorly understood. Furthermore, investigating the effects

136 of selective pressures resulting from abiotic factors, or how dormancy is related to

137 dispersal, is important for understanding the ecology and evolution of plant traits. The

138 seasonal macro-climate of the Brazilian savanna, the micro-climatic differences in open

139 savannas and wet grasslands habitats as a consequence of variations of the water table,

140 and the high variability in seed dormancy and dispersal syndromes of savanna grasses

141 make the *Cerrado* an ideal ecological system to investigate germination strategies.

142 Thus, our objectives were to investigate the germination, viability (*i.e.* longevity) and

143 dormancy levels of both freshly collected and dry-stored seeds: (1) from species living

144 in habitats with contrasting soil moisture during the dry season (open savannas *versus*

145 wet grasslands); (2) dispersed at different times (early in the wet season, late in the wet

146 season and in the dry season) and (3) displaying alternate dispersal syndromes

147 (barochoric *versus* anemochoric).

148 **Methods**

149 Study area

150   The *Cerrado* is the largest Neotropical savanna and covers around 2 million

151 square kilometers in Central Brazil. The climate of the region is seasonal with two well

152 defined seasons: a wet season from October to March (mean annual precipitation from

153 800 to 2000 mm; Oliveira-Filho & Ratter 2002) and a dry season from April to

154 September (Fig. 1). The study was conducted in two protected areas of Brasília:

155 Fazenda Água Limpa at the University of Brasília (FAL - 15º58'43.06"S and

156 47º56'21.41"W, 1.197 m above sea level) and the National Park of Brasília (PNB -

157 15º38'46.22"S and 48º00'19.75"W, 1.178 m above sea level). FAL and PNB are

158 composed of patches of savanna, forest and grassland. This study was conducted in

159 open savannas and wet grasslands. We selected two wet grassland sites in each area,

160 four open savanna sites in PNB and two open savanna sites in FAL. Vegetation in open

161 savannas and wet grasslands are composed mainly of grasses and forbs, with a few

162 scattered trees occasionally visible in open savannas. Soils in open savannas are well

163 drained, poor in nutrients and rich in aluminum. On the other hand, soils in wet

164 grasslands are rich in organic matter and the water table is near the surface (about 20–80

165 cm) (Cianciaruso & Batalha 2008), making the soil moist for most of the year

166 (Cianciaruso & Batalha 2008; Fidelis et al. 2013).

167 To characterize the soil moisture of the studied areas (FAL and PNB) we

168 measured the water potential (MPa/Ψ) of six open savanna sites and four wet grassland

169 sites every 30 days during the dry season, from June to August in 2015. Soil samples

170 were collected during the dry season to verify whether soils of wet grasslands stayed

171 wet longer than the soils of open savannas after the end of the wet season. We collected

172 soil samples in three plots previously set in each area equidistant (50 m) from each other

173 in a triangle arrangement, totaling 18 plots in open savannas and 12 plots in wet

174 grasslands. The plots were divided into three subsamples, one for each month of

175 collection. In each subsample soil samples were collected at two depths: between 2 to 3

176 centimeters and between 10 to 11 centimeters. These depths were selected because 1)

177 most seeds in the soil seed banks in Brazilian savannas were shown to occur in the first

178 few centimeters below the surface (Andrade & Miranda 2014), and 2) root biomass of

179 grasses was shown to occur mostly between 0 to 20 cm depth in Brazilian grasslands

180 (Fidelis et al. 2013), so a depth between 2 to 11 cm is well within the *perfil-position* that

181 roots of grass seedlings would grow during the initial development stages of

182 germination. The soil samples were placed in hermetically sealed sample cups and

183 stored in a coolbox. The water potential was measured in the laboratory with a WP4C

184 water potential meter (Decagon Devices, Pullman-USA, 2015), within two days of

185 sampling.

186   Seed collection

187   The dispersal units in grasses are generally complex structures composed of a

188 caryopsis (fruit), a caryopsis with bracts (lemma and palea) or occasionally a caryopsis

189 with inflorescence structures attached (hereafter called seeds). Seeds of 28 native

190 perennial grass species and one annual *Digitaria lehmanniana* (Table 1) from open

191 savannas and wet grasslands habitats were collected in 2012 and 2013, in FAL and

192 PNB. Plants were monitored regularly to ensure that seeds were mature at dispersal and

193 then collected by hand. After collection the seeds were stored in paper bags at room

194 temperature (27 ºC -maxima and 17 ºC -minima), measured throughout dry storage with

195 a thermometer. The average relative air humidity in the region (Distrito Federal) was

196 43-80% during the period of study (data from BDMEP/INMET). For accurate

197 identification, we collected vouchers of the studied grass species, which were deposited

198 in the Embrapa Genetic Resources and Biotechnology (CEN) Herbarium, headquartered

199 in Brasilia.

200   The monitoring of the grass populations selected for this study allowed us to

201 estimate the period of seed dispersal for each species. Thus, we classified the grass

202 species into three groups according to their dispersal times: a) species dispersing early

203 in the wet season (October to January); b) species dispersing late in the wet season

204 (February to April) and c) species dispersing in the dry season (May to September).

205 Based on their external morphology, we categorized the seeds into one of two dispersal

206 syndromes: anemochoric or barochoric (van der Pijl 1982). We considered those seeds

207 with structures that facilitate dispersal by wind, such as winged bracts and/or presenting

208 hairs in the bracts as anemochoric. Seeds without winged bracts or hairs were

209 considered barochoric (Table 1).

210   Germination, viability and dormancy levels of freshly harvested and dry-stored seeds

211   To determine the level of dormancy and to test the effect of dry storage on the

212 level of dormancy, viability and germination of seeds for each species, germination

213 trials were conducted with both freshly collected seeds as well as with seeds dry-stored

214 for three, six, nine and twelve months. The germination experiments were conducted in

215 germination chambers regulated set at an alternating temperature cycle of 28/18°C

216 day/night, under a photoperiod of 12h of white light. These temperatures were set

217 according to the average minimum and maximum temperatures recorded during the wet

218 season (Fig. 1), which represents the growing season for most savanna species in the

219   *Cerrado*, including grasses. Seeds were placed in petri dishes lined with two sheets of

220   filter paper and moistened with distilled water. The germination was recorded daily for

221   up to 30 days, using radicle emergence as a criterion for seed germination. Five

222   replicates of 20 seeds each were used for each species for each treatment, except for

223   *Paspalum maculosum, Agenium goyazense*, *Homolepis longispicula* and *Schizachyrium*

224   *sanguineum*, where we used five replicates of 10 seeds each per treatment due to the

225   limited quantity of seeds. After each germination trial, the viability of the ungerminated

226   seeds was tested using 1% tetrazolium chloride solution. The ungerminated seeds were

227   placed in contact with tetrazolium solution for 24 hours in the dark at 30 ºC in a

228   germination chamber. We considered the seeds whose embryos were stained dark pink

229   or red as viable. The total viability of each seed sample was set as the number of

230   germinated seeds during the experiments plus the positive results from the tetrazolium

231   test. The results for the viability were used to measure seed longevity across dry-storage

232   times.

233   The level of dormancy was interpreted as the difference between the estimated

234   viability and the number of germinated seeds for each seed sample (see statistical

235   analysis). As low germination can be the result of low seed viability of the seed sample

236   and not dormancy *per se*, we verified whether seed viability varied between treatments

237   (*i.e.* dispersal syndromes, habitat of seed collection and dry storage times; see Results

238   and Tables S1 to S5).

239   Statistical analysis

240   All analyses were done using the R 3.1.2 statistical platform (R Core Team

241   2014). To analyse the differences in soil water potential of wet grasslands and open

242   savannas during the dry season months we used Generalized Linear Mixed Models

243   (GLMM, normal distribution, see Zuur et al. 2009). As we intended to test for these

244   differences at each month (June, July and August), we made a model for each month

245   separately. We used the water potential as response variable, and included a two-way

246   interaction with habitat (wet grassland or open savanna) and depth (2-3 or 10-11 cm) as

247   fixed independent variables. We included the studied areas (FAL or PNB) and plots (3

248   *per* site) nested as random factors in the models. As the water potential did not follow a

249   normal distribution and ranged from negative to zero values, we log transformed (+1)

250   data. As it is not possible to obtain the log of negative values, we multiplied the data by

251   -1 to change it to a positive value before performing the log transformation. We made

252   *post hoc* pairwise comparisons between levels of the significant fixed factors: habitat

253   and depth. We used the single step method for P-values adjustment and the *glht*

254   command of multcomp package for the pairwise comparisons (Hothorn et al. 2008).

255   We used Generalized Linear Mixed Models (GLMM, binomial distribution, see

256   Zuur et al. 2009) (R package: lme4; see Bates et al. 2014) to test whether the effect of

257   dry storage (zero, three, six, nine and 12 months) on seed viability and germination is

258   dependent on seed dispersal time (early in the wet season, late in the wet season and in

259   the dry season), seed-dispersal syndrome (anemochoric or barochoric) and habitat of

260   seed collection (open savannas or wet grasslands). We analysed seed viability and

261   germination (presence/absence) as the response variables, separately. In this analysis,

262   we used each seed as an experimental unit. We included a two-way interaction term

263   between habitat, seed dispersal time and seed-dispersal syndrome and dry storage

264   treatment (zero, three, six, nine and 12 months), as fixed independent variables. We

265   included genus, species and replication as nested random factors in all models. As the

266   seeds placed inside the petri dishes are under the same environment, which can result in

267   autocorrelation of errors (Sileshi 2012), we included the number of replicates (5 *per*

268   species) at the random component of the model in order to control for autocorrelation of

269   errors. As all interactions were significant, it was not necessary to perform a model

270   selection, so we made only a single step with the full model and we used likelihood ratio

271   tests (LRT) to test the significance of the fixed independent variables. We made *post*

272   *hoc* pairwise comparisons between levels of the significant fixed factors: dry storage

273   and habitat, dry storage and seed dispersal time and dry storage and seed-dispersal

274   syndrome. We used the single step method to P-values adjustment and the *glht*

275   command of multcomp package for the pairwise comparisons (Hothorn et al. 2008).

276   **Results**

277   Habitat seasonality and germination

278   The soil water potential at each depth differed between habitat types during the

279   dry season months of June, July and August, with the exception of 10 – 11 cm in June

280   and July (Fig. 2; Table 2). In wet grasslands, the soil water potential was higher than in

281   open savannas for all dry season months (Fig. 2). The mean soil water potential at

282   shallower depths (2 – 3 cm) was higher in wet grasslands than in open savannas,

283 ranging from -0.5 MPa (June) to -1.2 MPa (August) and from -3.2 MPa (June) to -5

284 MPa (August), respectively (Fig. 2; Table 2).

285   The germination of grass seeds during dry storage was influenced by habitat of

286 seed collection (Likelihood-Ratio Test, LRT_4 = 27.583, $P<0.001$; Fig. 3). The

287 germination among species from open savannas increased after six months of dry

288 storage relative to freshly collected seeds and stayed constant until twelve months of dry

289 storage (Table 3; Fig. 3). On the other hand, the germination increased at a greater rate

290 for species from wet grasslands after three months of dry storage relative to freshly

291 collected seeds, and also stayed constant until twelve months of dry storage (Table 3;

292 Fig. 3). Similar to seed germination, seed viability response over storage time was also

293 influenced by species habitat (LRT_4 = 10.323, $P<0.05$). In open savannas the seed

294 viability was lower after nine (β = -0.57, $P<0.05$) and twelve (β = -0.75, $P<0.05$)

295 months of dry storage in comparison to freshly collected seeds, while in wet grasslands

296 the seed viability did not differ during storage time (Table 3; Table S1 - supplementary

297 data).

298 Dormancy level comparison against dispersal time and dispersal syndrome

299   The germination of grass seeds during dry storage was influenced by the seed

300 dispersal time (LRT_8 = 110.738, $P<0.001$; Fig. 4). Freshly collected seeds dispersed

301 early in the wet season and in the dry season had a higher probability of germination

302 than seeds dispersed late in the wet season (Table 3; Fig. 4). After six months of dry

303 storage the probability of germination was no longer influenced by the dispersal time of

304 the seeds (Fig. 4). After six months of dry storage, early-dispersed seeds had lower

305 viability than late dispersed seeds in the wet season (β = -0.9635, $P<0.05$; Table S4 -

306 supplementary data). After twelve months of dry storage the viability of early-dispersed

307 seeds was lower than late-dispersed seeds (β = -0.9258, $P<0.05$; Table S4 -

308 supplementary data) and of seeds dispersed in the dry season (β = -2.2161, $P<0.001$;

309 Table S4 - supplementary data).

310   We found a significant effect of seed-dispersal syndrome (LRT_4 = 28.785,

311 $P<0.001$; Fig. 5) on germination probability during dry storage. The germination in

312 barochoric seeds was lower than in anemochoric ones for freshly collected seeds and

313 also after three months of dry storage (Table 3; Fig. 5). After six months of dry-storage

314 the probability of germination was no longer influenced by the seed-dispersal syndrome

315 (Fig. 5). The viability of barochoric and anemochoric seeds did not differ (Table S2 -

316 supplementary data), except at six months of dry-storage, when barochoric seeds

317 showed a lower viability than anemochoric seeds (Table S2 - supplementary data).

Discussion

319   There were contrasting effects of dry-storage on the germination and viability of

320 species from open savannas and wet grasslands. In open savannas the germination of

321 fresh seeds was initially low, increasing after six months of storage, but then was

322 followed by seed mortality after nine months of storage. These results indicate that

323 germination in seeds of species from open savannas may be delayed due to dormancy,

324 at most, until the onset of the next wet season. On the other hand, seeds of species from

325 wet grasslands were longer-lived than seeds from open savannas, remaining viable after

326 one year of dry storage. Additionally, germination levels increased and stayed

327 consistently high after three months of dry storage.

328   In wet grasslands, the soil remained wet during the dry season in comparison to

329 open savannas, presenting values around -1 MPa at 2 to 3 cm depth (Fig. 2). Water

330 potential of around -1 MPa is not limiting for the germination of grass species (Qi &

331 Redmann 1993), thus suggesting that the water potential measured in the wet grasslands

332 studied here would not limit the germination of seeds present in the soil. Moreover, the

333 soil moisture and the high percentages of germination of grass species from wet

334 grasslands suggest that germination might occur even during the dry season.

335   Although rainfall seasonality does not seem to be a strong selective pressure on

336 seed germination of species from wet grasslands, due to the high soil moisture even

337 during the dry season, other factors such as the pressure of stablished vegetation, have

338 been demonstrated to negatively influence seedling establishment in savannas

339 (Zimmermann et al. 2008). Wet grasslands can be strongly competitive environments

340 for seedlings, as they have dense and closed aboveground plant biomass of around 765

341 g m^{-2} (Fidelis et al. 2013). Fire frequently occurs in wet grasslands and can reduce

342 competition by removing the established vegetation (Zimmermann et al. 2008). The

343 high longevity of seeds, combined with high germination rates, can result in an

344 opportunistic strategy for taking advantage of gaps by grass species from wet

345 grasslands, for example after a fire event, to allow growth and establishment in a less

346 competitive environment.

347 *Seed dormancy at the end of the wet season may avoid risky germination*

348　　　　We show that seed dormancy is related to the dispersal time of Neotropical grass
349 species (Fig. 4). Seeds dispersed late in the wet season showed lower probability of
350 germination than seeds dispersed either early in the wet season, or dispersed during the
351 dry season. The high levels of dormancy among late dispersed seeds may represent a
352 drought-avoidance syndrome, a strategy already observed among grass species of
353 savanna environments (Mott 1978; Veenendaal et al. 1996; McIvor & Howden 2000;
354 Scott et al. 2010; Salazar et al. 2011), and Melastomataceae species from high
355 grasslands in other parts of Brazil (Silveira et al. 2012). Seeds dispersed in the
356 beginning of the wet season are expected to have about seven months of relatively
357 stable water availability to enable germination and establishment. On the other hand,
358 seed germination at the end of the wet season would be very risky, since seedlings
359 would not have time enough to grow and acquire a minimal size that enable them to
360 tolerate the harsh conditions expected during the dry season. Thus, the presence of seed
361 dormancy here might prevent germination during periods when there is a low
362 probability of recruitment, and hold back emergence until the onset of the next wet
363 season.

364　　　　Contrary to our expectations, the germination of seeds dispersed in the dry
365 season was not different from seeds dispersed early in the wet season. Seeds dispersed
366 during the dry season showed low levels of seed dormancy. Since sporadic rains are
367 expected to occur in the dry season (Fig. 1), we did not expected to find high
368 germination in seeds dispersed during this time. However, the amount of rain is
369 extremely low (Fig. 1) and may potentially not be sufficient to induce germination.
370 Additionally, the following wet season occurs relatively soon after dispersal and, by not
371 having seed dormancy, these dry season dispersed seeds may rapidly germinate and take
372 advantage of the entire growing season to establish.

373 *Trade-off between seed dispersal and level of dormancy*

374　　　　We found a negative relationship between seed dispersal and seed dormancy.
375 Anemochoric seeds germinated to a significantly greater degree and consequently had
376 lower levels of dormancy than barochoric seeds. As far as we know, this is the first
377 study to demonstrate a trade-off between seed dispersal and dormancy across species
378 from the same family (Poaceae). Moreover, most of both the theoretical and empirical
379 studies investigating the evolution of seed dispersal and seed dormancy have been made

380 for plants from unpredictable environments, mostly in deserts (Ellner & Shmida 1981;

381 Volis & Bohrer 2013). Our between-species comparison in a seasonal rainfall ecological

382 system provides empirical results showing that barochoric grass seeds are significantly

383 more dormant than anemochoric ones.

384   Seed dispersal and seed dormancy may be viewed as bet-hedging strategies to

385 enable species to cope with environmental patch heterogeneity and climatic

386 unpredictability (Bulmer 1984; Venable & Brown 1988), the former by spreading risk

387 over space (Bulmer 1984), and the latter by spreading risk over time (Venable & Brown

388 1988). However, even in the absence of patch heterogeneity, seed dispersal can be

389 favored by kin selection (Venable & Brown 1988). Additionally, in environments

390 without inter-annual variation in precipitation, seed dormancy can be advantageous

391 where sibling competition is high (Volis & Bohrer 2013). Although water availability

392 affects seedling establishment in savannas (Medina & Silva 1990; Higgins et al. 2000)

393 and might explain seed dormancy, it does not explain our results that anemochoric seeds

394 had lower seed dormancy than barochoric seeds. In open savannas and wet grasslands of

395 the *Cerrado* there is no clear patch heterogeneity, as exists in deserts, so it is less likely

396 that this has driven a seed dispersal-dormancy trade-off in our studied species.

397 Alternatively, we suggest that competition between siblings, conspecifics or

398 heterospecifics, might drive the dispersal-dormancy trade-off found in our study

399 species.

400   Conspecific competitor densities during seedling growth negatively influences

401 plant fitness (Orrock & Christopher 2010). By dispersing far and not being dormant,

402 grass seeds with an anemochoric dispersal syndrome might enhance the chances of a

403 seed landing in a more open patch, enabling it to take advantage of sites with lower

404 competition for resources (Ellner 1988; Venable & Brown 1988; Cohen & Levin 1991).

405 Moreover, fire is a frequent disturbance in Brazilian open savannas and wet grasslands,

406 and fire occurrence can open up gaps in the herbaceous vegetation. Thus, these

407 anemochoric species may be favored by germinating earlier and growing in gaps opened

408 by fire events. In contrast, the outcome for barochoric seeds is that they fall near to the

409 mother plant and their own siblings. Because they disperse throughout the wet season,

410 there is sufficient moisture to enable seed germination, and seedlings could grow in

411 clumps if no other mechanism were available. Thus, seed dormancy can be a way to

412 reduce immediate germination and distribute seedling emergence over time.

413 Alternatively, greater dormancy among barochoric seeds could give seeds more time to

414 enable a secondary dispersal event. Indeed, some of the barochoric grass species studied

415 have structures recognized to favor secondary dispersal, including *Echinolaena inflexa*

416 and *Icnanthus camporum*, which both have elaiosomes that are known to attract ants

417 (Giladi 2006). Furthermore, *Aristida* species have awns, which can attach to rodents

418 providing potential longer-distance dispersal.

419 In conclusion, our results show that seed dormancy is a strategy to synchronize

420 the germination of grasses at the beginning of the wet season in seasonal environments.

421 The coevolution of both strategies – seed dormancy and the timing of seed dispersal –

422 contribute to enabling seeds to avoid germination during periods with low chances of

423 successful recruitment, such as at the end of the wet season. We show, using a large

424 representative number of grass species from Neotropical savannas, that seed dormancy

425 is a critical life-history trait that allows persistence in seasonal environments with dry

426 periods. Moreover, our results also provide empirical evidence for a trade-off between

427 seed dispersal and seed dormancy. These results indicate that rainfall seasonality is not

428 the only selective pressure driving the evolution of seed dormancy in grass species.

429 Furthermore, we suggest that competition can have an important influence selecting for

430 the timing of germination of seeds from seasonal and predictable environments.

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554 **Supplementary material**

555 Table S1. *Post hoc* comparisons of probability of seed viability between dry storage

556 months according to habitat types.

557 Table S2. *Post hoc* comparisons of probability of seed viability between seeds with

558 contrasting dispersal syndromes across dry storage months.

559 Table S3. *Post hoc* comparisons of probability of seed viability between dry storage

560 months according to the timing of seed dispersal.

561 Table S4. *Post hoc* comparisons of probability of seed viability between seeds dispersed

562 in different periods across dry storage months.

563 Table S5. Percentage dormancy (D), seed germination (G) and viability (V) of grass

564 species from *Cerrado* according to dispersal time, seed-dispersal syndrome and habitat

565 of seed collection along twelve months of dry storage.

Table 1. Time of seed dispersal, seed-dispersal syndrome and habitat of occurrence of grass species from open savannas and wet grasslands of Central Brazil.

Species	Seed-dispersal syndrome	Seed dispersal (Season)	Habitat	Site
Agenium goyazense (Hack.) Clayton	Barochory	Dry	Open savanna	PNB
Andropogon leucostachyus Kunth	Anemochory	Early	Wet grassland	FAL
Anthaenantia lanata (Kunth) Benth.	Anemochory	Early	Open savanna	PNB
Aristida gibbosa (Nees) Kunth	Barochory	Dry	Open savanna	FAL
Aristida recurvata Kunth	Barochory	Dry	Open savanna	PNB
Aristida riparia Trin.	Barochory	Dry	Open savanna	FAL
Aristida setifolia Kunth	Barochory	Late	Open savanna	FAL
Arthropogon villosus Nees	Barochory	Early	Wet grassland	PNB
Axonopus siccus var. siccus (Nees) Kuhlm.	Barochory	Late	Open savanna	FAL
Ctenium cirrhosum (Nees) Kunth	Anemochory	Late	Open savanna	PNB
Digitaria lehmanniana Henrard	Barochory	Late	Wet grassland	PNB
Echinolaena inflexa (Poir.) Chase	Barochory	Late	Open savanna	FAL
Elionurus muticus (Spreng.) Kuntze	Anemochory	Early	Open savanna	PNB
Eragrostis polytricha Nees	Barochory	Early	Wet grassland	PNB
Homolepis longispicula (Döll) Chase	Anemochory	Early	Wet grassland	PNB
Ichnanthus camporum Swallen	Barochory	Late	Open savanna	FAL
Mesosetum ferrugineum (Trin.) Chase	Anemochory	Early	Wet grassland	PNB
Panicum olyroides Kunth	Barochory	Early	Open savanna	PNB
Paspalum carinatum Humb. & Bonpl. ex Flüggé	Anemochory	Late	Open savanna	FAL

Paspalum erianthum Nees ex. Trin.	Anemochory	Early	Open savanna	PNB
Paspalum glaucescens Hack.	Barochory	Late	Open savanna	PNB
Paspalum guttatum Trin.	Anemochory	Early	Open savanna	PNB
Paspalum maculosum Trin.	Barochory	Late	Wet grassland	PNB
Paspalum pectinatum Nees ex Trin.	Anemochory	Early	Open savanna	PNB
Paspalum polyphyllum Nees	Anemochory	Dry	Wet grassland	FAL
Saccharum villosum Steud.	Anemochory	Early	Wet grassland	PNB
Sacciolepis myuros (Lam.) Chase	Barochory	Dry	Wet grassland	FAL
Schizachyrium sanguineum (Retz.) Alston	Barochory	Dry	Open savanna	PNB
Setaria parviflora (Poir.) Kerguélen	Barochory	Late	Open savanna	PNB

Table 3. Mean of percentage of level of dormancy (D), seed germination (G) and viability (V) of grass species from *Cerrado* according to dispersal time, seed-dispersal syndrome and habitat of

Time period of seed dispersal: Early (October - January), late in the wet season (February - April) and during the dry season (May - September). Site of occurrence: PNB (National Park of Brasília) and FAL (Água Limpa farm).

Table 2. Multiple comparisons of differences in soil water potential between open savannas and wet grasslands from Central Brazil during the dry months.

Month	LRT_1	P	Habitat	Depth (cm)	$\beta \pm SE$	P
June	17.562	P<0.001	OS – WG = 0	2-3	0.6346 ± 0.2192	P<0.05
			OS – WG = 0	10-11	0.4418 ± 0.2189	P=0.05
July	21.405	P<0.001	OS – WG = 0	2-3	0.6161 ± 0.2334	P<0.05
			OS – WG = 0	10-11	0.4293 ± 0.2328	P=0.08
August	39.049	P<0.001	OS – WG = 0	2-3	0.9121 ± 0.2199	P<0.05
			OS – WG = 0	10-11	0.6127 ± 0.2194	P<0.05

OS = Open Savannas, WG = Wet Grasslands. $\beta \pm$ Standard Error.

seed collection along twelve months of dry storage.

| | Dry storage (Months) | | | | | | | | | | | | | | |
	0			3			6			9			12		
	D	G	V	D	G	V	D	G	V	D	G	V	D	G	V
Dispersal time															
Early in the wet season	17	60	74	11	58	70	10	60	65	9	68	76	27	44	50
Late in the wet season	90	8	70	71	18	69	71	24	64	52	29	58	53	26	52
Dry season	55	34	73	32	44	81	49	54	77	36	38	67	24	59	81
Habitat															
Open savanna	58	30	72	39	40	70	43	39	63	34	40	63	48	27	47
Wet grassland	39	45	72	40	48	78	23	61	76	23	62	76	19	55	64
Seed dispersal															
Anemochoric seeds	15	60	71	5	63	67	11	61	68	4	66	67	22	44	44
Barochoric seeds	74	21	73	64	29	77	52	37	68	47	36	68	47	34	61

Level of dormancy (D) = number of ungerminated viable seeds over total viable seeds.

566
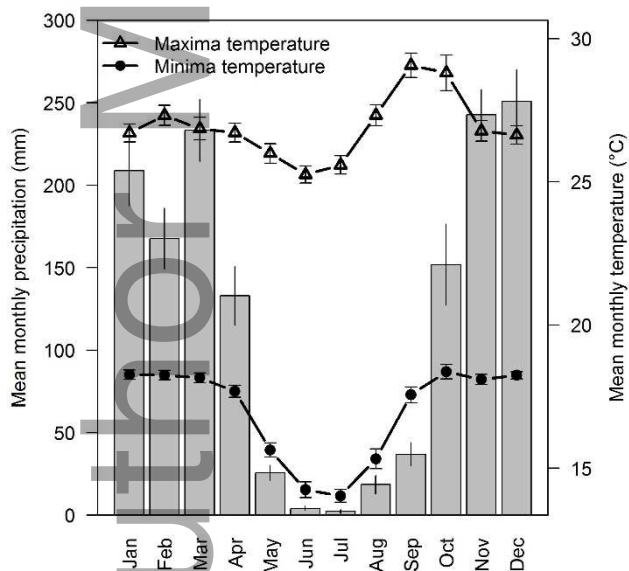
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568 Figure 1. Mean monthly minimum and maximum temperatures and precipitation in
569 Brasília-DF from 1994 to 2014. Data from BDMEP/INMET.
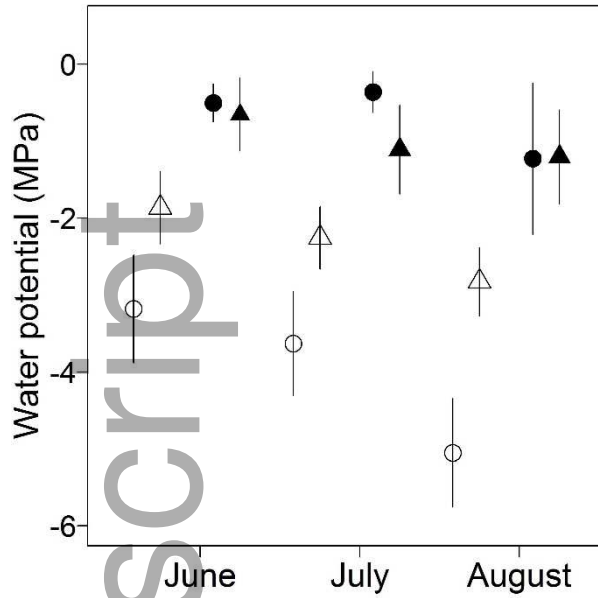
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2 Figure 2. Soil water potential (MPa) of wet grasslands and open savannas during the dry

3 season. Open symbols = open savannas; Closed symbols = wet grasslands; Circles = 2 -

4 3 cm depth; Triangles = 10 - 11 cm depth. Mean + 95% Confidence Interval.

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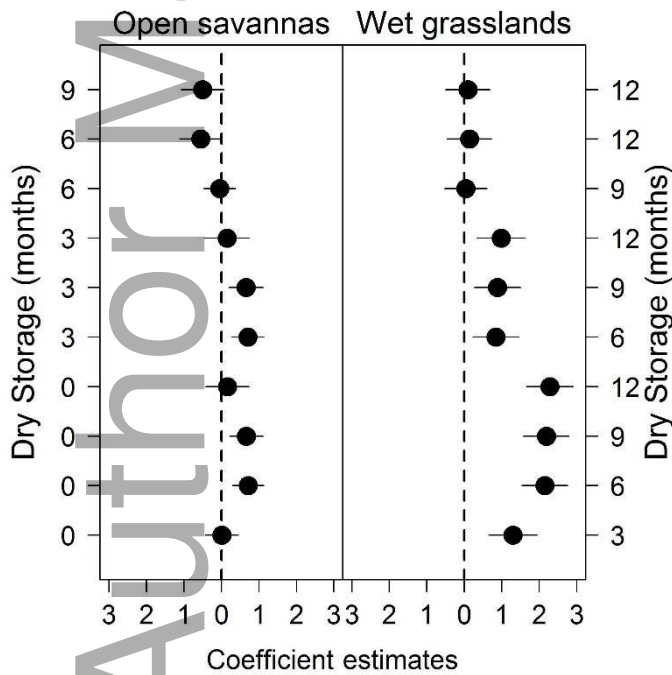
6

7 Figure 3. Coefficient estimates from pairwise multiple comparisons of germination

8 probability between months of dry storage in grass species from open savannas and wet

9 grasslands. The closed circles denote the mean values, the error bars denote 95% of

1   lower and upper confidence intervals. The central dotted line (zero value) means no

2   statistical difference.
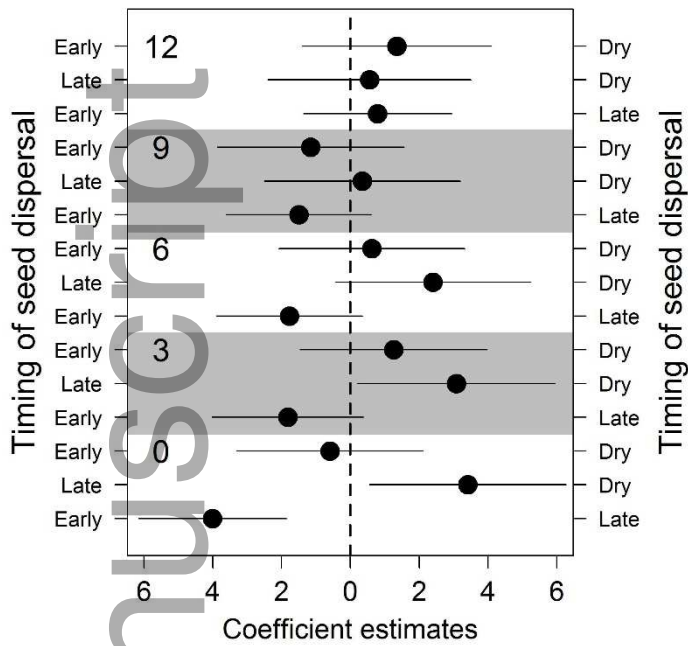
3

4   Figure 4. Coefficient estimates from pairwise multiple comparisons of germination

5   probability between grass seeds dispersed early, late in the wet season and in the dry

6   season along dry storage (0, 3, 6, 9 and 12 months). The closed circles denote the mean

7   values, the error bars denote 95% of lower and upper confidence intervals. The central

8   dotted line (zero value) means no statistical difference.
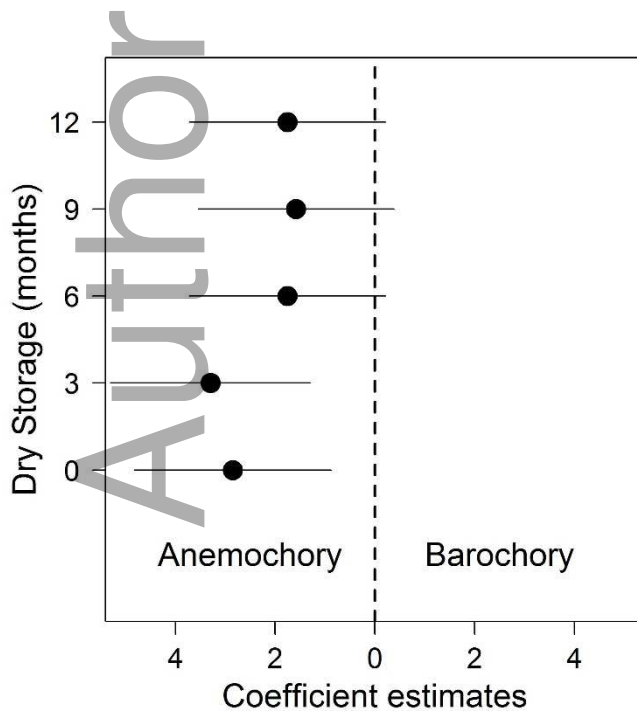
9

1 Figure 5. Coefficient estimates from pairwise multiple comparisons of germination

2 probability between grass species with anemochory and barochory dispersal syndromes

3 along dry storage (0, 3, 6, 9 and 12 months). The closed circles denote the mean values,

4 the error bars denote 95% of lower and upper confidence intervals. The central dotted

5 line (zero value) means no statistical difference.