



The importance of seed mass for the tolerance to heat shocks of savanna and forest tree species

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Keywords

Climatic change; Heat shock; Neotropical savanna; Seed mass; Seed tolerance

Nomenclature

Lista de Espécies da Flora do Brasil
(<http://reflora.jbrj.gov.br/jabot/listaBrasil/ConsultaPublicaUC/ConsultaPublicaUC.do>)

Received 2 December 2014

Accepted 23 April 2015

Co-ordinating Editor: John Morgan

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Introduction

Tropical forests and savanna are important centres of biodiversity, reflecting complex evolutionary histories involving co-evolution and niche differentiation (Murphy & Bowman 2012). Environmental conditions, such as soil nutrient and water availability, are known to determine the distribution of these biomes (Ruggiero et al. 2002; Bond 2008; Hirota et al. 2011). Furthermore, fire is suggested to play a central role in the maintenance of savanna–forest boundaries (Hoffmann et al. 2009, 2011; Staver et al. 2011a). Fire frequency in savanna ecosystems is relatively high mainly due to the almost continuous occurrence of grasses that provides fuel for fire events, especially during

the dry season (Bond 2008). Conversely, fire incidence in forest ecosystems is low due to the existence of a dense canopy that tends to exclude grasses and maintain a lower biomass understorey (Hennenberg et al. 2006). Studies demonstrated that savanna tree species are more fire-tolerant than forest tree species leading to lower rates of topkill than forest trees (Hoffmann et al. 2009). This higher tolerance found in savanna tree species is clearly linked to adaptations to survival in fire-prone environments, such as increased bark thickness (Hoffmann et al. 2003).

High temperatures generated by a fire can also affect several aspects of seed biology, including seed viability and germination patterns (Keeley & Fotheringham 2000; Auld & Denhan 2006), affecting especially the soil seed bank

Abstract

Questions: Does seed mass influence the tolerance of seeds to the effects of heating in fires? Is the tolerance to heat shocks during fire events dependent mostly on seed mass itself or to other traits linked to the species ecological origin, e.g. non-fire-prone (forest) and fire-prone (savanna) environments?

Location: Savanna and forest ecosystems of the *Cerrado* biome, Brazil.

Methods: Heat shocks were applied to seeds of 17 selected common tree species in savanna (ten species) or forest ecosystems (seven species) of the *Cerrado* biome. After being submitted to a heat shock, seeds were allowed to germinate in chambers set at 30 °C. Germination percentage was calculated for each treatment and species. For comparison purposes, species were grouped into two functional groups (forest and savanna) and also into seed size classes (small, medium and large). We used generalized linear model to analyse heat shock effects on seed germination of savanna and forest tree species, and how seed size affected the germination response.

Results: Seed mass influenced seed tolerance to heat shock. Larger seeds had higher survival when subjected to heat shock, independent of species' ecological origin. In addition, seeds from species occurring in fire-prone environments (savanna) were more tolerant to high levels of heat shock than seeds from species occurring in non-fire-prone environments (forest).

Conclusion: In response to global climatic change, fire regimes in savannas of South America are expected to become more frequent and intense, even expanding into currently non-fire-prone environments. In this sense, our findings indicate that species from savannas (fire-prone environments) might have an adaptive advantage to these consequences of climatic changes, potentially leading to expansion of the savanna ecosystems.

(Stark et al. 2008; Keeley et al. 2011). Soil seed banks occurring in fire-prone environments, such as savannas, are shown to have good tolerance to the high temperatures (Williams et al. 2005; Scott et al. 2010). In contrast, fires might reduce soil seed banks density (Kennard et al. 2002; Martins & Engel 2007) and viability (Cochrane 2003) from species occurring in tropical forests, where the fire frequency is lowest compared to the frequencies found in savannas (Cochrane 2009). Thus, the low tolerance of seeds, seedlings and saplings of forest species to fire might strongly limit their encroachment into savanna ecosystems (Fensham et al. 2003; Bond et al. 2005; Hoffmann et al. 2009; Geiger et al. 2011; Scott et al. 2012).

Differences in functional traits might explain the different plant responses to changes in environmental conditions (Barbosa et al. 2014a,b). Seed traits, such as seed size (usually measured as mass), have long been regarded as important to determine the reproductive biology of plant species (Silvertown 1981; Leishman et al. 2000). For example, seed trait differences influence the probability of seedling establishment by affecting the distance that seeds disperse, the likelihood that they will reach suitable habitats for germination and the probability of survival (Westoby et al. 1992).

It is suggested that large-seeded species have an advantage in stress tolerance, including a higher tolerance to fire effects (Westoby et al. 2002; Moles et al. 2004; Lahoreau et al. 2006; Moles & Westoby 2006). However, small-seeded species are suggested to have advantage in seed production, fecundity and dispersion, due the high numbers of seeds they produce (Leishman et al. 2000; Henery & Westoby 2001; Muller-Landau et al. 2008; Muller-Landau 2010). Furthermore, the production of large or small seeds by the plant species is suggested to be associated to the bet-hedging responses to variations in environmental conditions (see Cohen 1966). Large seeds are linked to conservative bet-hedging response of plants inhabiting environments where reproductive success is highly variable and unpredictable (Venable 1989). Although plants with large seeds produce less offspring in copious years, they produce at least a few seeds in unfavourable years, when small-seeded species do not have sufficient resources to survive (Philippi & Seger 1989).

Although extensive research on how seed size variation affects plant performance has been carried out in forest and savanna species (Jurado & Westoby 1992; Pearson et al. 2002; Baraloto et al. 2005; Lahoreau et al. 2006), little is known about the effects of seed size on the germination response of species occurring in fire-prone and in non fire-prone environments (Hoffmann 2000; Gashaw & Michelsen 2002). Understanding the differences in forest and savanna species responses to fire and whether seed mass might influence it is crucial to a better understanding

of the role of fire on the dynamics of savanna–forest boundaries.

Due to the high frequency of fire (Hennenberg et al. 2006) and to the high tolerance to fire of soil seed banks in savanna ecosystems (Cochrane 2009; Scott et al. 2010), we would expect higher tolerance to heat shocks in seeds from savanna tree species compared with seeds from forest tree species, independent of the seed mass (Hypothesis 1). As in fire-prone environments large seeds showed higher tolerance to heat shocks than small ones (see Escudero et al. 2000; Gashaw & Michelsen 2002), we expect that among savanna tree species larger seeds are more tolerant to heat shocks than smaller ones (Hypothesis 2). Due to the low tolerance of forest tree species to fire (Hoffmann et al. 2009), we expect no strong differences in germination success between large and small seeds when subjected to heat shock (Hypothesis 3). We tested these hypotheses using tree species from the *Cerrado* biome, a Brazilian biome that contains both savanna and forest ecosystems.

Methods

The *Cerrado* biome covers about 2 million km², representing ca. 22% of the land surface of Brazil, extending from the southern borders of the Amazonian forest to outlying areas in the southern states of São Paulo and Paraná, its distribution being highly coincident with the plateaux of central Brazil (Oliveira-Filho & Ratter 2002). The typical vegetation landscape of the *Cerrado* biome consists of savanna of very variable structure, termed *cerrado sensu lato*, on the well-drained interfluves, with gallery forests or other moist vegetation following the watercourses (hereafter referred as ‘forest’ ecosystem; Oliveira-Filho & Ratter 2002). However, around 70% of the *Cerrado* biome is covered by a unique vegetation cover, the *cerrado sensu stricto* (hereafter referred as ‘savanna’ ecosystem), a typical savanna vegetation dominated by trees and shrubs often 3 to 8-m tall and giving more than 30% crown cover but with a fair amount of herbaceous vegetation, mainly grasses species between the woody vegetation (Oliveira-Filho & Ratter 2002).

Although at macro-climate scale forest and savanna ecosystems may share similar rainfall and temperature regimes (Furley 1999), at local scale there are clear differences; for example, savanna ecosystems are exposed to frequent fires while forest ecosystems are not (Hoffmann et al. 2009, 2012). Studies pointed to fire as a major driver of the vegetation dynamics in forest–savanna boundaries in the *Cerrado* biome (Geiger et al. 2011; Hoffmann et al. 2012). Therefore, this biome offers an excellent opportunity to conduct studies on the adaptations of forest and savanna species to fire.

Study sites

Seed samples were collected in three distinct sites: (1) at the Federal University of Roraima (UFRR; 02°38'N, 60°49'W), located 15 km north of Boa Vista, Roraima state, Brazil, at 77 m a.s.l. The climate of this area is classified as tropical with a dry winter (Aw) under the Köppen-Geiger classification (Peel et al. 2007), with around 1600 mm mean annual precipitation (MAP; Araújo et al. 2001). The driest months are between Dec and Mar (10% of annual precipitation), and in this period, the mean minimum and maximum temperatures are 23 and 33 °C, respectively. During the peak of the rainy season (May to Aug – 60% of annual precipitation), the mean maximum and minimum temperatures are 22 and 31 °C, respectively (Miranda & Absy 2000); (2) IBGE Ecological Reserve (RECOR; 15°55'S, 47°52'W) and (3) *Água Limpa* farm (FAL; 15°56'S, 47°55'W). These two adjacent sites (2 and 3) are located near Brasília, Federal District, Brazil, at an elevation of approximately 1100 m and ca. 1500 mm MAP (Geiger et al. 2011). In these sites, the climate is also classified as Aw in Köppen-Geiger (Peel et al. 2007), with a pronounced dry season from May to Sept, in which the mean minimum and maximum temperatures are 14 and 26 °C, respectively. During the rainy season, the main minimum and maximum temperatures are 18 and 30 °C, respectively (Lenza & Klink 2006).

Plant species and seed collection

This study tried to include a phylogenetically comparative context during selection of the species. However, species of the *Cerrado* biome have a low phylogenetic clustering among the plant families, due to the persistence of long-lived resprouting species from different plant lineages (Silva & Batalha 2010). Moreover, there is a loose relationship between families found in forest and savanna ecosystems (Oliveira-Filho & Ratter 1995). As this study focus on the effects of fire (e.g. heat shock) on seed viability and germination in a broad range of representative species of this biome, we selected 17 common tree species in accordance to their origin either in savanna (ten species) or forest ecosystems (seven species), their frequency of occurrence in the collection areas (Ratter et al. 2003) and the availability of individuals during the seed dispersal season (Table 1). Mature seeds were collected from at least ten individuals of each species, from different populations located in savanna (FAL and UFRR) and forest (RECOR and UFRR) areas. The savanna and forest ecosystems where the seeds were collected were at least 100 m apart. After collection, seeds were stored in paper bags under laboratory conditions (22–24 °C, 65–85% relative humidity) for a maximum of 1 month before the start of the germination experiments.

To estimate the average seed dry mass per species, 60 seeds per species were used. Each seed was weighed before and after drying in an oven (Nova Técnica NT 516, Piracicaba, SP, Brazil) at 105 °C for 24 h.

Heat shocks treatments

To test the effects of heat shocks on seed germination rates, the seeds were placed in a temperature-controlled oven with air circulation and renewal (Marconi MA035, Piracicaba, SP, Brazil), adjusted to provide one of the following temperatures: 80, 110, 140, 170 and 200 °C. These heat shocks were applied to the seeds for 2.5 or 5.0 min. These combinations of temperatures and times exposure were chosen based on field recordings taken in the soil of Brazilian savanna ecosystems (surface and/or shallow depths) during a prescribed fire (see Miranda et al. 1993). Control treatments consisted of seeds not submitted to heat shock.

General procedures

Four replicates of 15 seeds were placed into Petri dishes, a total of 60 individual seeds for each heat (ten levels) and species (17 levels). The replicates for each species were placed in oven separately. After being submitted to a heat shock, the seeds were allowed to germinate in Petri dishes (15-cm diameter) with two sheets of filter paper moistened with distilled water (15 ml each plate), which was replenished every 2 days. The Petri dishes were placed in germination chambers (Marconi MA402, Piracicaba, SP, Brazil) set at 30 °C, a temperature within the range considered optimal for the germination of *Cerrado* species (see Brancalion et al. 2010), with 12 h/12 h light/dark (white light, 30 $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$). Germination was scored daily by counting and removing the germinated seeds until no germinated seed was detected. The criterion used for germination was the emergence of the radicle, followed by its geotropic curvature to avoid false germination (for more detailed description see Labouriau 1983). A tetrazolium viability test (for methodological details see Moore 1973) was performed to check whether heat shocks killed non-germinated seeds. Similar procedure was performed on the non-germinated seeds of control treatments.

Data analyses

For comparison purposes, the species were grouped into two functional groups (forest and savanna) and also into seed size classes (Table 1). In most studies of seed size, classes (normally termed small, medium and large) rather than seed masses of individual seeds have been used (see Seiwa 2000; Leishman 2001). Thus, species were grouped into three seed size classes: small (six species), medium

Table 1. Dispersal period, dry seed mass (mean \pm SD), seed size class and seed dormancy of forest and savanna tree species studied.

Plant species	Family	Ecosystem/Collection area	Dispersal period	Dry seed mass (mg)	Seed size class	Seed dormancy
<i>Curatella americana</i>	Dilleniaceae	Savanna/UFRR	February to May	8.7 \pm 0.9	1	Physical
<i>Bowdichia virgilioides</i>	Fabaceae	Savanna/UFRR	February to May	15.8 \pm 1.1	1	No
<i>Roupala montana</i>	Proteaceae	Savanna/FAL	September to December	22.2 \pm 1.9	1	No
<i>Himatanthus articulatus</i>	Apocynaceae	Savanna/UFRR	February to April	22.3 \pm 2.2	1	No
<i>Jacaranda puberula</i>	Bignoniaceae	Forest/RECOR	July to September	26.2 \pm 3.7	1	No
<i>Handroanthus impetiginosus</i>	Bignoniaceae	Forest/RECOR	September to November	33.1 \pm 1.2	1	No
<i>Tachigali rubiginosa</i>	Fabaceae	Forest/RECOR	July to August	53.1 \pm 4.5	2	No
<i>Anadenanthera peregrina</i>	Fabaceae	Forest/UFRR	March to May	56.2 \pm 3.6	2	No
<i>Dalbergia densiflora</i>	Fabaceae	Forest/RECOR	July to October	58.3 \pm 2.2	2	No
<i>Tachigali subvelutina</i>	Fabaceae	Savanna/FAL	August to October	84.3 \pm 8.6	2	No
<i>Qualea grandiflora</i>	Vochysiaceae	Savanna/FAL	August to October	86.9 \pm 3.0	2	No
<i>Kielmeyera coriacea</i>	Clusiaceae	Savanna/FAL	September to October	123.5 \pm 2.5	3	No
<i>Tabebuia aurea</i>	Bignoniaceae	Savanna/FAL	September to October	135.3 \pm 8.6	3	No
<i>Anadenanthera falcata</i>	Fabaceae	Forest/RECOR	August to September	139.2 \pm 12.5	3	No
<i>Eriotheca pubescens</i>	Malvaceae	Savanna/FAL	August to November	175.8 \pm 10.0	3	No
<i>Dimorphandra mollis</i>	Fabaceae	Savanna/FAL	August to December	227.4 \pm 5.4	3	Physical
<i>Copaifera langsdorffii</i>	Fabaceae	Forest/RECOR	June to October	415.5 \pm 13.3	3	No

(five species) and large (six species), which differed by an order of magnitude. Seed mass ranged from 8.7 to 33.1 mg (mean 21.4 mg) in the small size class, 53.1 to 86.9 mg (mean 66.8 mg) in the medium size class, and 123.5 to 415.5 mg in the large size class (mean 202.7 mg).

The percentage viability and germination of the seeds before heat shock treatments were calculated for each size class, within each ecosystem (savanna and forest). Subsequently, one-way ANOVA was performed to compare these parameters according to seed size class.

To understand how heat shocks affect the seed germination of savanna and forest tree species, and how these responses were affected by seed size, we used the germination data (absence or presence) in a GLM. The ecosystem of occurrence of the species (forest vs savanna), the heat shock treatments (80, 110, 140, 170 and 200 °C), the exposure times (2.5 and 5.0 min) and seed size classes (small, medium and large) were included as fixed variables. The tree species were included in the model as random variables.

Generalized linear model analyses were used to analyse the data sets, using binomial error structure with logit-link function. The analysis were conducted in R (v 3.0.2; R Foundation for Statistical Computing, Vienna, AT) using the *lmer* function of the *package lme4* (<http://cran.r-project.org/web/packages/lme4/lme4.pdf>). The output from *lmer* includes SE estimates for the random effects and coefficients for the fixed effects (Pinheiro & Bates 2000;

Baayen et al. 2008; Zuur et al. 2009). Step-wise backward and forward analysis was used to select the best-fit model. To test the significance of the terms in the statistical model we ran Monte Carlo Markov Chain simulations (10 000 interactions), using R (<http://cran.r-project.org/doc/manuals/R-lang.pdf>).

Results

The best-fitting model took into account every variable studied (ecosystem, seed mass, temperature and time), as well as their interactions (Table 2). Seeds from forest and savanna species showed similar tolerance to a heat shock of 80 and 110 °C for 2.5 and 5.0 min, regardless of seed size class (Figs 1, 2). However, increases in the exposure temperature led to differences in the germination response, depending on the seed size class and functional group. Small seeds were more sensitive to the higher heat shocks than medium and large ones, for both forest and savanna species (Figs 1, 2). However, higher temperatures were more harmful to forest (Fig. 1) than to savanna (Fig. 2) seeds.

When the exposure time was increased from 2.5 to 5.0 min, small and medium seeds of forest species showed low or no tolerance to a heat shock of 110 °C and above, while large seeds showed tolerance up to 140 °C (Fig. 1b). For savanna species, medium and large seeds tolerated temperatures of 110 and 140 °C for up to 5.0 min, and

Table 2. Ecosystem, temperature, time and seed size effects on the germination response of seeds of savanna and forest tree species subjected to a heat shock.

Source of variation	df	F-ratio	P-value
Ecosystem	1	4.770	0.043
Seed size	2	10.850	0.001
Temperature	4	406.496	<0.00001
Time	1	203.789	<0.0001
Ecosystem × Seed size	2	0.339	0.717
Ecosystem × Temperature	4	16.715	<0.001
Ecosystem × Time	1	2.601	0.107
Seed size × Temperature	8	11.241	<0.001
Seed size × Time	2	5.471	0.004
Temperature × Time	4	17.448	<0.001
Ecosystem × Seed size × Temperature	8	4.912	<0.001
Ecosystem × Seed size × Time	2	4.321	0.014
Ecosystem × Temperature × Time	4	2.929	0.020
Seed size × Temperature × Time	8	10.628	<0.001
Ecosystem × Seed size × Temperature × Time	8	2.958	0.003

large seeds tolerated temperatures of 170 °C for 5.0 min (Fig. 2b). Heat shocks of 170 and 200 °C for 5.0 min were lethal for seeds of most seed classes, for both forest (Fig. 1b) and savanna (Fig. 2b) species. The increase in exposure time to a heat shock was more harmful to forest (Fig. 1) than savanna seeds (Fig. 2), especially for small and medium seeds.

The one-way ANOVA performed to compare seed viability and germination of the seeds before the heat shocks did not show differences among these parameters, within each ecosystem and according to seed size class (Table 3). Tetrazolium tests showed that the viability of the non-germinated seeds was low within the unheated seeds, and

zero among the heated seeds after the treatments. In this sense, it seems that the viability and germination percentages were substantially equivalent, regardless of the treatment applied, i.e. non-germinated seeds lost their viability as a result of the heat shocks.

Discussion

In this paper, we aimed to test the effects of heat shocks on germination success of tropical savanna and forest tree species. We compared seeds of different size classes exposed to different heat treatments for different time periods. We found that seeds from species occurring in fire-prone environments (savanna) were more tolerant to heat shocks than seeds from species occurring in non-fire-prone environments (forest). Moreover, seed mass also determined the tolerance to heat shocks. Larger seeds tolerate better the high temperatures applied, independent of species ecological origin.

Although several studies have evaluated fire effects on the savanna–forest dynamics (Staver et al. 2011b; Hoffmann et al. 2012; Scott et al. 2012), there are few comparative studies regarding how fire might modulate this balance through its effects on seed germination (Ribeiro & Borghetti 2014). Seeds from species occurring in fire-prone vegetation are expected to have a high tolerance to fire (Williams et al. 2005; Scott et al. 2010). Indeed, fire reduced viability of the soil seed banks in tropical forests (Cochrane 2003; Slik et al. 2010) and the results from this study are in agreement with this statement, thus confirming the negative effect of fire on forest seed banks.

The seed tolerance to heat shocks also depends on the fire temperature and duration (Keeley & Fotheringham 2000). In our study, there was no significant difference in

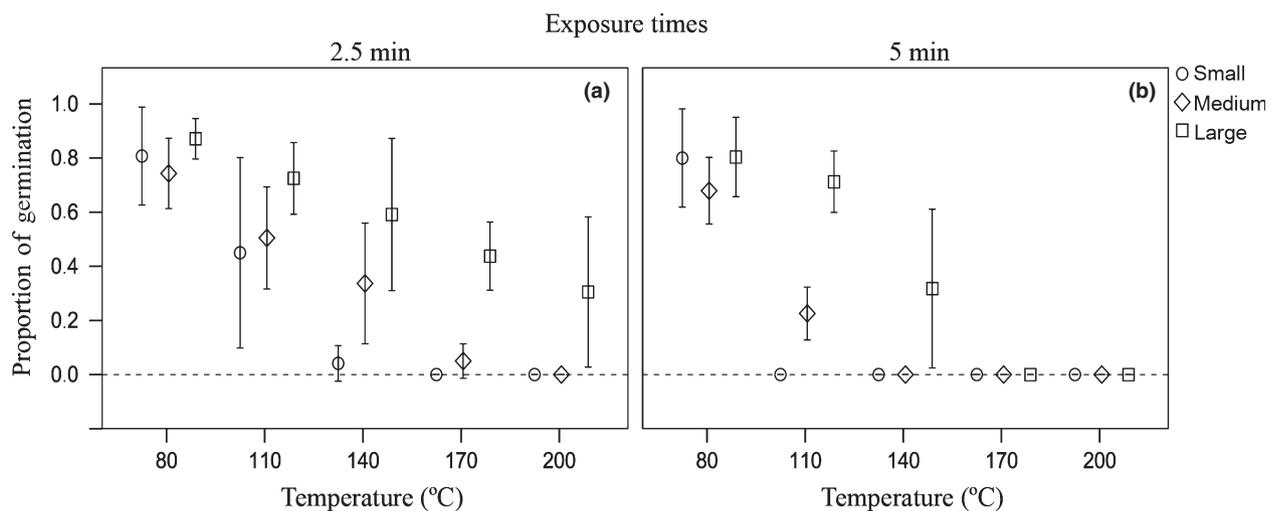


Fig. 1. Seed germination response of forest tree species subjected to heat shocks, according to their size class. (a) 2.5 min; (b) 5 min. Error bars: 95% CI.

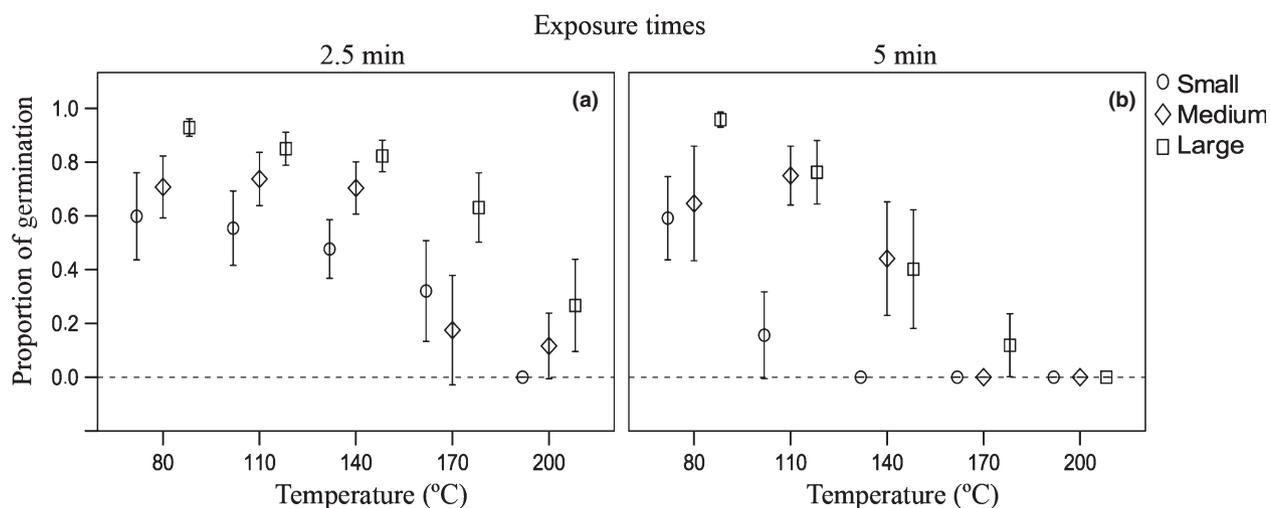


Fig. 2. Seed germination response of savanna tree species subjected to heat shocks, according to their size class. (a) 2.5 min; (b) 5 min. Error bars: 95% CI.

Table 3. One-way ANOVA performed to compare the percentages of seed viability and germination before heat shock treatments for each size class and within each ecosystem. Data expressed as mean \pm CI (95%).

Ecosystem	Seed size class	Viability (%)	Germination (%)	df	F-ratio	P-value
Forest	1	90.0 \pm 13.1	89.1 \pm 15.2	1	0.01	0.91
	2	78.4 \pm 12.0	75.1 \pm 15.0	1	0.36	0.56
	3	81.5 \pm 11.5	80.0 \pm 13.3	1	0.05	0.80
Savanna	1	71.4 \pm 18.3	66.8 \pm 22.2	1	0.40	0.54
	2	80.1 \pm 10.8	75.1 \pm 13.2	1	0.68	0.57
	3	95.7 \pm 4.3	94.9 \pm 5.1	1	0.20	0.66

germination rates between savanna and forest species when seeds were heated up to 80 °C. Indeed, different studies have shown that heat shocks up to 80 °C have minimal impact on seed viability, irrespective of the ecosystem in which the species occur (Escudero et al. 2000; Hanley & Lamont 2000; Thomas et al. 2007; Ribeiro et al. 2013; Ribeiro & Borghetti 2014). However, higher temperatures and/or longer times of exposure led to a significant decrease in seed viability (Gashaw et al. 2002; Reyes & Trabaud 2009) and/or germination success (Gashaw & Michelsen 2002; Thomas et al. 2007; Reyes & Trabaud 2009). In general (in agreement with Hypothesis 1), seeds of savanna species were more tolerant to heat shocks than seeds of forest species. In this study almost all savanna species tolerated heat shocks up to 140 °C for 5.0 min and up to 200 °C for 2.5 min (Figs 1, 2). These results confirm the high tolerance of savanna seeds and the low tolerance of forest seeds to heat shocks (Hoffmann 2000; Ribeiro & Borghetti 2014). Furthermore, our results also show that heat shock of 200 °C for 5.0 min was lethal to all seeds, both for forest and savanna species. Indeed, heat shock of 200 °C

for up to 5.0 min was previously reported to be lethal for seeds of both savanna and forest ecosystems (Schmidt et al. 2005; Ribeiro & Borghetti 2014). As temperatures of this magnitude were reported to occur close to the soil surface during a fire event in the Cerrado biome (Miranda et al. 1993; Neves & Miranda 1996), these results indicate that the survival of seeds present in the soil seed banks will depend largely on the position large and small seeds occupy with respect to the soil surface during a fire event.

In light of the evolutionary role of fire, little is known about which seed traits may best predict the germination response of forest and savanna species (Hoffmann 2000; Gashaw & Michelsen 2002) and whether these traits might be relevant to the ability of the seeds to tolerate the high temperatures generated during a fire event (Pausas et al. 2004; Bond & Keeley 2005). While few studies found that small seeds are more tolerant to heat shock than large seeds (Hanley et al. 2003), several others studies have shown an opposite relationship (Bradstock et al. 1994; Escudero et al. 2000; Gashaw & Michelsen 2002; Delgado et al. 2008). Our study demonstrated that seed size was an

important functional trait related to seed tolerance to high temperatures (Hypothesis 2). Larger seeds showed higher tolerance to heat shocks than smaller ones, irrespective of the heat treatment applied or the seed origin (savanna or forest species). These findings might help to explain why trees with larger seeds are more dominant in open savannas areas (Hoffmann 2000; Lahoreau et al. 2006) where fire intensity and temperatures are higher.

High temperatures usually have a positive effect on the germination of seeds that exhibit some degree of dormancy, frequently by removing coat-imposed dormancy (Herranz et al. 1998; Baker et al. 2005; Reyes & Trabaud 2009). In this study, this was the case for *Curatella americana* (Dilleniaceae) and *Dimorphandra mollis* (Fabaceae), whose physical dormancy was overcome by the heat treatments. Seeds of many Fabaceae species are characterized by the presence of physical dormancy (Baskin et al. 2000), which might be broken by a heat treatment. However, most Fabaceae species used in this study show no evidence of physical dormancy (see Table 1), since they imbibed without any scarification treatment. A comparison within this family reveals that species from fire-prone ecosystems are able to tolerate higher temperatures than species from non-fire-prone ecosystems (Gashaw & Michelsen 2002; Dayamba et al. 2008; Zuloaga-Aguilar et al. 2011; Ribeiro & Borghetti 2014), indeed, our results corroborate this pattern regardless of seed size and degree of dormancy presented by the Fabaceae seeds.

Variation in environmental conditions is suggested to be responsible for the large differences found in seed size among plant species (Leishman & Westoby 1994; Leishman et al. 2000; Baraloto et al. 2005; Moles et al. 2005; Rees & Venable 2007). Moreover, model outcomes propose that stress tolerance is a factor that favours the maintenance of large seeds in plant communities (Muller-Landau 2010). Our results provide general support for these models, as larger seeds were more tolerant to heat shocks (consequently to fire) than smaller ones, within and among functional groups. In contrast with our expectation (Hypothesis 3), we found a high tolerance to heat shock in larger seeds among forest species (*Anadenanthera falcata*, *A. peregrina* and *Copaifera langsdorffii*); these species were able to germinate after being subjected to 170 °C (Fig 2a). Besides their occurrence in forest ecosystems, *A. falcata*, *A. peregrina* and *C. langsdorffii* are also found in the savanna-forest boundary of the Brazilian Cerrado (Freitas & Oliveira 2002), might suggest that forest species that produce large (and heavy) seeds may be able to survive and recruit into savanna ecosystems. The production of large seeds is suggested as linked to conservative bet-hedging germination success when adult fitness is reduced (Venable 1989; Walck et al. 2011). Hence, species with large seeds

might have an advantage during years with unfavourable environmental conditions, producing at least a few seeds and viable offspring, when small-seeded species do not have sufficient resources to survive during these unfavourable environmental conditions. Consequently, our results suggest that forest species that produce small seeds would have their recruitment in savanna ecosystems largely limited by the high frequency of fires in these habitats.

Fire regimes in savannas of South America are expected to change over the next decades in response to global climatic change (Guariguata et al. 2008), becoming more frequent and intense (Siqueira & Peterson 2003) and even expanding into non fire-prone environments (Dale et al. 2001). Thus, more frequent and intense fires may act as a selective pressure to the advantage of species that already occur in fire-prone ecosystems, and limit the recruitment of less fire-tolerant species, usually forest species, in those ecosystems subjected to fire. Our results suggest that higher fire frequency and/or fire intensity due to climate change may select for those species that produce larger seeds, in both forest and savanna ecosystems, and limit the recruitment of species that produce small seeds, irrespective of whether they come from fire-prone or non fire-prone environments.

Acknowledgements

We acknowledge CAPES (CAPES/NUFFIC process 019/2010 and CAPES/PNADB process 451/2010), CNPq (process 476297/2004-4) and the University of Brasília, through their Deanship of Research and Post-Graduation, for financial support.

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