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Estratégias de persistência e regeneração em campo sujo de Cerrado após o fogo

Talita Marques Zupo

Tese apresentada ao Instituto de Biociências do Câmpus de Rio Claro, Universidade Estadual Paulista, como parte dos requisitos para obtenção do título de Doutor em Ciências Biológicas - Biologia Vegetal.

Maio - 2017

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À pequena vida que nascerá, com a certeza de que
as crianças renovam as esperanças...

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essa conquista com um sorriso maroto
debaixo do bigode!

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“Não precisamos de mais tempo,
precisamos de um tempo que seja nosso.”

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Índice

Resumo

Em ambientes onde ocorre o fogo, as espécies de plantas podem regenerar via rebrote, semente ou através de ambas as estratégias. Os mecanismos de regeneração são favorecidos ou limitados tanto pelas condições ambientais (produtividade e disponibilidade de recursos) quanto pelo regime de queima, incluindo sua frequência e época. Assim, o objetivo principal desta tese foi investigar a regeneração pós-fogo de espécies de uma comunidade de campo sujo de Cerrado localizada na Reserva Natural Serra do Tomabdor (Goiás). Avaliamos o papel da regeneração via rebrote e via semente e procuramos avaliar os fatores que estão envolvidos no rebrote das plantas após serem submetidas a diferentes tipos de distúrbio, e distúrbios que ocorreram em diferentes frequências e épocas do ano, visto que tanto o tipo do distúrbio quanto a época e a frequência em que ele ocorre podem afetar a regeneração das plantas. Verificamos que o rebrotamento é a principal estratégia de regeneração pós-fogo, ocorrendo principalmente a partir de gemas localizadas em estruturas subterrâneas que estão altamente protegidas pelo solo, e que o recrutamento via semente tem um papel pequeno na regeneração. No entanto, a tolerância das sementes a altas temperaturas é uma característica importante que permite o recrutamento quando as condições ambientais forem favoráveis. Também verificamos que as respostas pós-fogo, relacionadas tanto com o rebrote como com a floração, estão relacionadas com a disponibilidade de recursos no ambiente pós-fogo. A regeneração mais devagar das plantas queimadas no inicio da estação seca está provavelmente relacionada com o maior número de meses que estas plantas foram sujeitas a déficit hídrico, sendo que, após a estação chuvosa, as plantas queimadas nas diferentes épocas do ano apresentaram tamanhos similares. A regeneração após três queimas consecutivas não foi diferente da regeneração após queimas com intervalos maiores, sugerindo que a quantidade de reservas armazenadas não foi um fator limitante. Além disso, o fogo induziu a floração das espécies em relação a plantas não queimadas, sugerindo que o fogo é um fator importante na reprodução das espécies.

Abstract

In fire-prone ecosystems, plant species can persist by resprouting, recruiting from seeds, or use both strategies. Such regeneration mechanisms can be favored or limited by both environmental conditions (site productivity and resource availability) and fire regimes, such as fire frequency and season. The aim of this thesis was investigate the post-fire regeneration mechanism of Cerrado plant species from an open savanna community located at the Reserva Natural Serra do Tombador (Goiás, Central Brazil). Thus, we evaluated the role of post-fire resprouting and post-fire seeding as regeneration mechanisms in Cerrado open savannas. Moreover, we evaluated the factors driving resprouting responses after plants are subjected to different disturbance types and to disturbances in different seasons and frequencies. We verified that resprouting is the main post-fire regeneration mechanism in Cerrado, originating mainly from buds located in underground structures, and that post-fire seeding plays a minor role in the post-fire regeneration of this community. However, seed tolerance is an important seed trait as it allows for seed germination and recruitment when favorable environmental conditions arise. Our results suggest that post-fire resprouting is driven mainly by availability of resources present in the post-fire environment. The slower recovery of plants burned during early-dry season fires is most likely related to the prolonged exposure to water deficits as plants endured all five months of the dry season. However, after the rainy season, plants burned in the different fire seasons had similar sizes. We found no difference in resprouting responses between annually and biennially burned plants, and plants resprouted vigorously after three consecutive burns, suggesting that amount of stored reserves was not a limiting factor. Moreover, fire induced flowering in most species in relation to unburnt plants, suggesting fire is an important factor for the reproduction of these species.

Introdução Geral

O fogo é um importante fator ecológico em vários ecossistemas do mundo como, por exemplo, nas savanas africanas (Trollope 1982), australianas (Morgan 1999) e neotropicais (Coutinho 1990; Sarmiento 1993; Simon et al. 2009), e nos ecossistemas mediterrâneos (Keeley et al. 2012). A passagem do fogo leva a um aumento na incidência de luz e na disponibilidade de nutrientes (Kauffmann et al. 1994; Whelan 1995; Neary et al. 1999), criando condições favoráveis para a regeneração da vegetação. Em ambientes onde ocorre o fogo, as espécies de plantas são classificadas de acordo com suas estratégias de regeneração em: espécies germinadoras (*seeders*), que morrem com a passagem do fogo e dependem da germinação de suas sementes e posterior estabelecimento de suas plântulas para manutenção das populações; espécies rebrotadoras (*resprouters*), que rebrotam após a passagem do fogo, rapidamente reocupando os espaços; e, espécies rebrotadoras facultativas (*facultative resprouters*), que podem regenerar tanto via rebrote como pelo recrutamento de sementes (Bond & Midgley 2001,2003; Pausas et al. 2004; Pausas & Keeley 2014).

O mecanismo de regeneração pós-fogo, via rebrote ou via semente, é favorecido ou limitado tanto pelas condições ambientais (produtividade e disponibilidade de recursos) quanto pelo regime de queima. Este é caracterizado por sua frequência (intervalo de retorno do fogo), intensidade (calor que é liberado), época (período em que ocorre a queima) e tipo de material combustível que é consumido (e.g. fogo de copa ou fogo de superfície; Bond & Van Wilgen 1996; Bond & Keeley 2005). Por exemplo, ambientes sujeitos a frequentes fogos de superfície de baixa intensidade que consomem rapidamente o estrato herbáceo, como as savanas (Miranda et al. 2002; Bond & Keeley 2005; Pausas & Ribeiro 2013), favorecem as espécies rebrotadoras (Bellingham & Sparrow 2000; Bond & Midgley 2001, 2003; Pausas & Keeley 2014). Já as espécies germinadoras são favorecidas nos ambientes sujeitos a fogos de alta intensidade que consomem a copa de espécies arbóreas e arbustivas, ocorrendo em intervalos longos, uma vez em várias décadas, como os que ocorrem nos ambientes de vegetação mediterrânea (Keeley et al. 2011; Keeley et al. 2012; Pausas & Keeley 2014).

A germinação pós-fogo está normalmente associada com a presença de um banco de sementes resistente ao fogo e com o atraso da germinação até o período pós-fogo (Pausas et al. 2004; Pausas & Keeley 2014). Assim, a germinação é frequentemente desencadeada por diferentes mecanismos relacionados ao fogo como, por exemplo, choques de temperatura (Moreira et al. 2010; Moreira and Pausas 2012) e fumaça (Keeley and Pausas 2016). Estudos realizados com espécies de vários ecossistemas suscetíveis ao fogo observaram que choques de temperatura (simulando a passagem do fogo) podem aumentar a germinação, principalmente em espécies com sementes impermeáveis, já que as altas temperaturas quebram a dormência física das sementes, possibilitando então a germinação (Williams et al. 2003; Reyes and Trabaud 2009; Moreira et al. 2010; Moreira and Pausas 2012). Por outro lado, diversos estudos mostraram que, para muitas espécies, a germinação não é afetada pelos choques de temperatura, mas suas sementes sobrevivem às altas temperaturas indicando tolerância a estas (Gashaw and Michelsen 2002; Clarke and French 2005; Jaureguiberry and Diaz 2015; Le Stradic et al. 2015)

Já a capacidade de rebrotar está associada com a sobrevivência das gemas e com a quantidade de reservas armazenadas para sustentar o rebrote (Bond & Midgley 2001; Clarke et al. 2013). A sobrevivência das gemas depende, dentre outros fatores, da proteção e localização das gemas (Clarke et al. 2013). Assim, as espécies que rebrotam a partir de suas partes aéreas (e.g. gemas epicórmicas) possuem as gemas protegidas por uma cortiça grossa ou localizadas em camadas mais profundas do tronco, como ocorre com muitas espécies arbóreas nas savanas africanas (Charles-Dominique et al. 2015), australianas (Burrows et al. 2010) e neotropicais (Dantas and Pausas 2013). Outras espécies possuem gemas basais ou subterrâneas localizadas em estruturas especializadas, como o lignotuber e xilopódio, protegidas pelo solo (Noble 2001; Simon et al. 2009; Simon and Pennington 2012; Clarke et al. 2013; Fidelis et al. 2014).

A capacidade de rebrotar é considerada uma característica ancestral entre as angiospermas. No entanto, ela não é só geneticamente determinada, sendo influenciada tanto pelas condições ambientais (produtividade e disponibilidade de recursos), como pelo regime do fogo (Bond & Midgley 2003; Bond & Keeley 2005; Pausas & Ribeiro 2013). Dentre os fatores que caracterizam o regime do fogo, a frequência e a época em que ocorre a queima podem afetar o rebrotamento das plantas de diversas maneiras. Intervalos pequenos entre queimas podem causar um esgotamento na quantidade de reservas armazenadas (Miyanishi and Kellman 1986; Canadell & López-Soria 1998; Enright et al. 2011), de modo que pode haver uma diminuição no número de ramos rebrotando (Medeiros & Miranda 2008) ou no vigor do rebrote (biomassa rebrotada, Paula & Ojeda 2006). Por outro lado, intervalos longos entre queimas tendem a resultar em fogos de maior severidade e intensidade, já que há maior quantidade de biomassa acumulada, o que também pode afetar a resposta da vegetação (Whelan 1995).

A resposta pós-fogo das plantas também pode ser afetada pela época em que ocorre a queima (Cespedes et al. 2014; Hmielowski et al. 2014). A umidade do solo e da vegetação varia ao longo do ano, afetando alguns parâmetros do fogo, como a taxa de propagação e sua intensidade e severidade (Govender et al. 2006; Pivello et al. 2010). O material combustível tende a ser mais inflamável no final da estação seca, já que há maior quantidade de biomassa morta, em relação ao material combustível no início da estação seca. Assim, queimas no início da estação seca costumam ser menos intensas que queimas no meio ou no final da estação seca (Williams et al. 1999; Govender et al. 2006; Knapp et al. 2009). Além disso, a sazonalidade é um fator importante na dinâmica da vegetação em muitos ecossistemas, onde as reservas de amido podem ser utilizadas durantes os períodos de crescimento da planta que ocorrem durante a estação chuvosa (Filgueiras 2002), diminuindo, assim, a quantidade armazenada para sustentar o rebrote, caso o fogo ocorra logo após tais períodos.

No Cerrado, o fogo está presente há milhões de anos (Simon et al. 2009), e o regime de fogo é caracterizado por frequentes fogos de superfície de baixa intensidade (Miranda et al. 2002). Nas fisionomias campestres e savânicas, as gramíneas perenes constituem o grupo de espécies mais frequente e as principais contribuintes para a biomassa total (Silva & Ataroff 1985; Almeida 1995). A biomassa aérea das gramíneas é acumulada rapidamente durante a estação chuvosa, e se torna inflamável durante o período seco, levando ao fogo (Bond & Keeley 2005; Pivello et al. 2010). No Cerrado, o fogo natural causado por raios é comum na estação chuvosa, mas também nos meses de transição, principalmente na transição da estação seca para a chuvosa (Setembro e Outubro; Ramos-Neto & Pivello 2000; Medeiros & Fiedler 2004; Alvarado et al. 2017). Assim como em outras savanas mésicas, a frequência de fogo no Cerrado é alta, com um intervalo de retorno de fogo de 3-4 anos (Daldegan et al. 2014; Pereira-Junior et al. 2014), não sendo limitada pela quantidade de material combustível (Alvarado et al. 2017). Deste modo, a recorrência do fogo é determinante na manutenção de fisionomias abertas de Cerrado, uma vez que a exclusão do fogo pode favorecer o estabelecimento de espécies lenhosas (Hoffmann et al. 2012; Durigan & Ratter 2016).

Deste modo, o objetivo principal desta tese foi investigar a regeneração pós-fogo de espécies de campo sujo de Cerrado presentes na Reserva Natural da Serra do Tombador, Cavalcante, Goiás (Fig. 1). Para tal, esta tese foi dividida em três capítulos, sendo que no primeiro **"Post-fire regneration strategies in a Cerrado open savana"**, avaliamos as diferentes estratégias de regeneração pós-fogo, classificando as espécies de campo sujo de acordo com tais estratégias (via rebrote e/ou via semente). Verificamos a capacidade de rebrote de espécies após episódios de fogo e também a tolerância e o estímulo de propágulos, por meio de experimentos de germinação e viabilidade, após serem submetidos a choques térmicos no laboratório. No segundo capítulo, **"Do different disturbance types affect resprouting and flowering responses of Cerrado species?"**, avaliamos os mecanismos da regeneração via rebrote de nove espécies de diferentes grupos funcionais, utilizando o corte e a queima como tratamentos experimentais. O terceiro capítulo, intitulado **"Effects of fire regimes on resprouting and flowering of Cerrado species",** buscamos avaliar se diferentes regimes de fogo resultam em diferenças na organização da comunidade vegetal por meio de queimadas experimentais realizadas em diferentes épocas (precoce, modal e tardia) e em diferentes frequências (bienal, anual e sem queima).

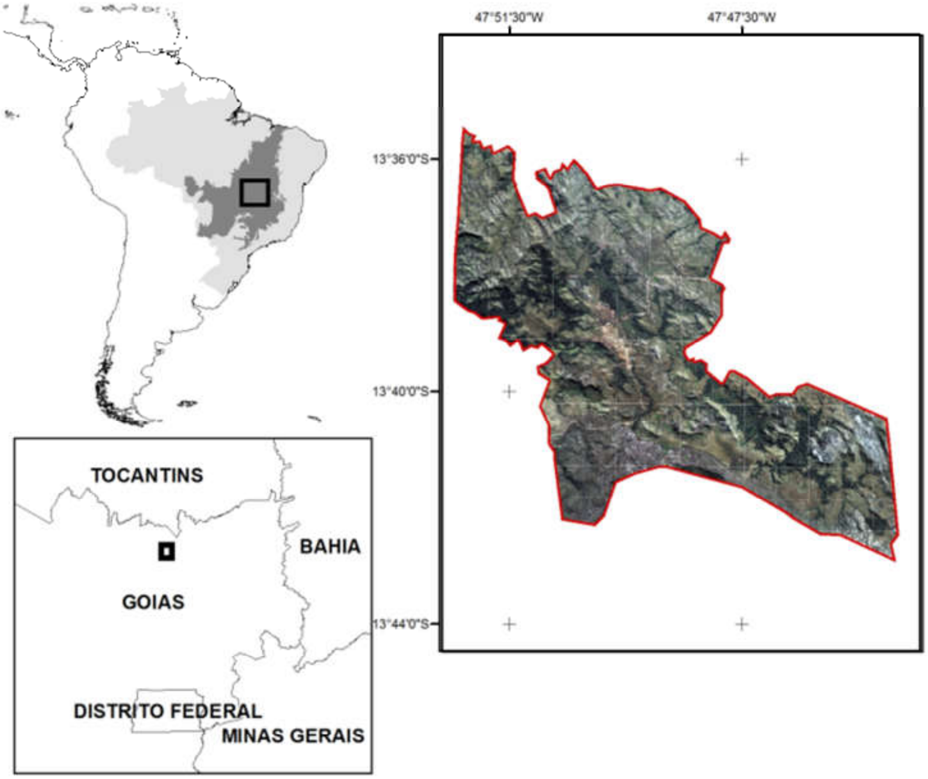

Figura 1. Localização da Reserva Natural Serra do Tombador, no Estado de Goiás, Centro-Oeste do Brasil.

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Capítulo 1

Post-fire regeneration strategies in a Cerrado open savanna

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Abstract

Fire is a natural disturbance present in many ecosystems throughout the world where plant species can persist by resprouting, recruiting from seeds or both. We evaluated the post-fire regeneration mechanisms for 41 Cerrado species, including grasses, forbs and shrubs, from an open savanna community in Central Brazil. We examined both resprouting ability after experimental fires, accouting for bud location, and seed response to heat shocks caused by fire, where seeds were subjected to heat shocks of 100^{o}C for one and three minutes, 200^{o}C and a control (untreated seeds). All species were able to resprout (R+) after fire, mainly from buds located in underground structures, but also from aerial and basal buds. Seeds of most species resisted heat shocks of 100^{o}C for one and three minutes, but heat treatments of 200^{o}C decreased seed viability of nearly 40% of species, and only seven species showed heat-stimulated germination. Thus, 81% of the community was classified as R+PT (resprouters with heat-tolerant propagules) and 17% was R+PS (resprouters with heat-stimulated propagules). Resprouting is the main post-fire regeneration strategy in Cerrado open savannas, and although post-fire seeding plays a minor role in Cerrado, we found heat-stimulated germination in 17% (seven species) of our community, demonstrating that such strategy may be coupled with resprouting as a post-fire regeneration strategy for some species. Additionally, we argue that fire-tolerance could be considered as an important seed trait that enables seed germination when favorable conditions arise.

Keywords: resprouting, seeding, germination, Neotropical savanna, fire ecology

Introduction

Fire is a main disturbance in many ecosystems worldwide (Bond and van Wilgen 1996; Bond and Keeley 2005) and both resprouting, defined as the production of new shoots after aboveground biomass removal, and seed recruitment have been recognized as key regeneration traits, where species may have one trait, both or none (Bond and Midgley 2001, 2003; Vesk et al. 2004; Pausas et al. 2004; Paula and Keeley 2014). Thus, classification for post-fire plant responses are usually based on whether individuals are killed or survive a fire episode by resprouting, and whether a plant species forms a fire-resistant seed bank with post-fire germination and recruitment (Pausas et al. 2004, Clarke et al. 2015).

Such mechanisms of post-fire regeneration are favored or limited by both the environmental conditions (site productivity and resource availability) and the fire regime at a given site, which include fire frequency, intensity, season and patterns of fuel consumption (Bond and Midgley 2003; Bond and Keeley 2005; Clarke et al. 2005; Pausas and Ribeiro 2013; Keeley et al. 2016). Different fire regimes select different functional traits, thus plant species seem to be adapted to a specific fire regime rather than to fire itself (Pausas and Keeley 2009; Keeley et al. 2011).

For instance, crown fires, such as those occurring in Mediterranean-like vegetation, consume canopy biomass of woody species and are considered high intensity fires that occur once in several decades (Keeley et al. 2011; Keeley et al. 2012; Pausas and Keeley 2014). In such systems, high temperatures and prolonged soil heating could affect resprouting responses by damaging buds or bud-forming tissues (Vesk et al. 2004; Wright and Clarke 2007), and recruitment from seeds would be conspicuous (Bond and Midgley 2003; Pausas and Keeley 2014; Clarke et al. 2015). In Australian crown-fire systems, around 40% of species are facultative species, displaying both post-fire resprouting and seeding, and up to 57% of species are obligate seeders, regenerating exclusively from seeds after fires (Clarke et al. 2015).

On the other hand, open vegetation, such as savannas and grasslands, undergo surface fires, which have relatively low intensity, fast rates of spread (rapidly consuming the grassy layer), and can reoccur within a few years interval (Miranda et al. 2002; Bond and Keeley 2005; Pausas and Ribeiro 2013), most likely favoring resprouting species (Bellingham and Sparrow 2000; Bond and Midgley 2001, 2003; Pausas and Keeley 2014). Location, protection and number of buds are main drivers of post-fire resprouting and adult individuals can resprout from aerial, basal or belowground buds, given that the meristematic tissues survive the high temperatures caused by fire (Clarke et al. 2013; Clarke et al. 2015; Charles-Dominique et al. 2015).

Post-fire seeding is often coupled with delaying germination to a single point in time, where germination may be triggered by different fire-related cues, such as heat shocks (Auld and Denham 2006; Reyes and Trabaud 2009; Moreira et al. 2010; Moreira and Pausas 2012), temperature fluctuations (Santana et al. 2010, 2013; Musso et al. 2015) and smoke (Keeley and Fotheringham 2000; Keeley and Pausas 2016). Heat shocks caused by fire have been reported to stimulate germination in several species of fire-prone ecosystems, mainly in hard-seeded species where high temperatures may break physical dormancy enabling germination (Auld and O'Connell 1991; Williams et al. 2003; Reyes and Trabaud 2009; Moreira et al. 2010; Moreira and Pausas 2012). Nonetheless, some species have been shown to be unaffected by heat shocks, although their seeds survived the exposure to high temperatures, which could be considered as a tolerance trait in face of fires (Jaureguiberry and Diaz 2015; Le Stradic et al. 2015; Ribeiro et al. 2015; Fichino et al. 2016; Fidelis et al. 2016; Zupo et al. 2016).

Fire has been present in Cerrado for at least four million years (Simon et al. 2009), and fire regimes are characterized by frequent surface fires of relatively low intensity fueled by grasses (Coutinho 1982; Kauffman et al. 1994; Miranda et al. 2002, 2009). After fires, many woody and herbaceous species resprout from dormant buds (Coutinho 1990; Medeiros and Miranda 2008), but the role of post-fire seeding as a regeneration mechanism in Cerrado remains unclear, even though such strategy has been observed in grasslands and savannas that undergo surface fires (Overbeck &

Pfadenhauer 2007; Clarke et al. 2015). Studies evaluating seed responses after heat-shocks in Cerrado have done so with few species and not on a community level approach (Ribeiro et al. 2013; Le Stradic et al. 2015; Fichino et al. 2016; Ramos et al. 2016). Moreover, such studies have not taken into account plant resprouting ability. In this context, we aimed to investigate the role of both post-fire resprouting and post-fire seeding as regeneration strategies for 41 species, including shrubs, forbs and grasses, from a Cerrado open savanna community. We assessed: (1) resprouting ability after experimental fires, accounting for bud location and (2) propagule stimulation and/or resistance, through assessment of seed germination and viability after heat shocks applied in the laboratory.

Material and Methods

Study area and seed collection

The study was conducted at the Serra do Tombador Nature Reserve (RNST), located in Central Brazil (47°45'; 47°51' W and 13°35'; 13°38' S, 8900 ha, 560-1118 m a.s.l). The climate has a marked dry season from May to September (total precipitation during this period is <60 mm) and a wet season from October to April, when most of the annual precipitation is concentrated (1300-1500 mm/year). Minimum temperatures may be <15°C and maximum can reach up to 36°C (Fundação Grupo Boticário 2011). The vegetation of the study area is characterized as a *campo sujo*, an open savanna vegetation with a continuous herbaceous layer dominated by grasses and scattered shrubs (Coutinho 1978). The RNST has been subjected to natural and anthropogenic fires, especially in grassland and savanna sites, with a mean fire return interval of 3 years (Daldegan et al. 2014).

Seeds from 41 different species from *campo sujo*, including different growth forms (shrubs, forbs and graminoids), were used in this study (Table 1). Seeds were collected throughout different seasons of the year between 2014 and 2016 from the most representative families in the area, especially Fabaceae (15 species), Asteraceae (4 species), Lamiaceae (4 species) and Poaceae (4 species). Seeds from all species were collected from several individuals (n >10), except for *Dyckia* *brasiliana* where seeds were collected from 3 individuals. Seeds were kept stored at low temperatures (<20°C), usually for less than three months, prior to the experiments.

Resprouting ability

We addressed data of resprouting ability for the same study species from which we collected seeds. To do so, we observed individuals (usually > 5 individuals per species) in the field after fires. From 2012 to 2016, two fire experiments have been set up in the RNST (see Gorgone-Barbosa et al. 2015; Rissi et al. 2017) where several plots were burned throughout the years. During this period, the area has been monitored every three months, where we observed conspicuous post-fire regeneration by resprouting of the species used in this study, and, whenever possible, determined the location the resprouting shoots were originating from: underground, aerial or basal buds.

Propagule germination and survival

Germination experiments were carried out to evaluate seed response to heat shocks simulating fire passage. Germination data for twenty-five of the 41 species used in this study are found in Table 2. For the remaining species, we compiled data from other studies, which used the same treatments as described below, from species collected in the same area (Novaes 2015; Nunes 2016; Zirondi 2016; Zirondi et al. unpubl. data; see Table 1). Seeds of all species were subjected to four different treatments prior to germination tests: (1) heat treatment of 100°C for 1 minute; (2) heat treatment of 100°C for 3 minutes; (3) heat treatment of 200°C for 1 minute; (4) control (untreated seeds). This combination of temperature and time was determined according to previous studies that observed heat pulses for Cerrado open savannas (Miranda et al. 1993; Daibes et al. unpubl. data). For each species, we used five replicates containing 20 seeds in each treatment (n = 100 seeds/treatment). Treatments were applied separately for each replicate in an electric oven to ensure independence of data (Morrison and Morris 2000). Seeds of *Vellozia* spp., *Xyris* sp., *Dyckia brasiliana*, *Mimosa pteridifolia* and *Senna corifolia* were not subjected to the 100°C - 3 minute treatment due to the limited number of seeds available (Novaes 2015; Nunes 2016; Zirondi 2015; Zirondi et al. unpubl. data).

After treatments, seeds were placed in Petri dishes with two layers of filter paper saturated with distilled water, and put in germination chambers at 27ºC (12 h of light). Seed germination was recorded three times a week during 30 days and germination was determined by radicle emergence (Bewley et al. 2013). Physiological dormancy (PD) was assigned for species which had less than 30% of germination in relation to the number of viable seeds in the control treatment, during the 30 days of the tests (Baskin & Bakin 2014; Dayrell et al. 2017). Species that had germination percentage between 30 and 70% in relation to its number of viable seeds were assigned to have 'intermediate' dormancy, while those with >70% of germination were considered non-dormant (see also Dayrell et al. 2017). Hard-coated seeds with no visible signs of imbibition by the end of the tests were assigned as physically dormant (PY). In such cases, seeds were then manually scarified with a sandpaper and allowed to imbibe and germinate for one week. Within this period, the germinated seeds were counted as viable, and thus we also assured they had no combined physiological dormancy.

Viability of ungerminated seeds of the visibly imbibed soft-coated seeds was analyzed at the end of each experiment by means of the Tetrazolium test at 1% solution. Those stained red were considered viable, those not stained or showing flaccid tissues were considered dead (Lakon 1949). The average of viable seeds (germinated + stained) from control replicates was used as the initial viability.

Classification of strategies and data analysis

Regarding resprouting ability, the species used in the study were classified as R+, species which presented post-fire resprouting, and R-, species that are not able to resprout and were killed by fires (Pausas et al. 2004); although such pattern was not observed among the study species (see Results). Location of resprouting shoots after fire were defined as originating from underground, basal or aerial buds (Clarke et al. 2013).

We defined three types of germination response regarding heat treatments based on Paula and Pausas (2008) and Jaureguiberry and Díaz (2015): (a) heat-tolerant propagules (PT), germination or viability of at least one of the heat treatments equal to the control treatment; (b) heat-stimulated propagules (PS), germination of at least one of the heat treatments higher than the control treatment and (c) heat-sensitive propagules (P-), germination or viability of all heat treatments significantly lower than the control treatment.

One-way analysis of variance applied to randomization tests (Euclidean distance between sampling units, 10000 iterations) were carried out to evaluate differences in germination and viability percentage between the different treatments (fixed factor) for each species. All statistical analyses were performed using MULTIV (Pillar 2008).

Results

All species were able to resprout (R+) after fire, mainly from buds located in underground structures (25 species, 66%), but also from aerial (9 species, 24%) buds, such as *Mimosa kalunga*, *Vellozia glochidea*, *V. squamata*, and *V. tubiflora*, and basal (4 species, 10%) buds, such as *Aristida riparia* and *Xyris* sp. (Table 1, Fig. 1). We did not observe non-resprouting (R-) species (Table 1).

Most species (81%) had propagules which resisted at least one of the heat treatments, and were thus classified as heat-tolerant (PT). Seven species (17%) had enhanced germination after at least one of the heat shock treatments and were classified as heat-stimulated (PS) and only one species (*Mimosa pteridifolia*) showed heat-sensitive propagules (P-, Table 1), with decreased germination and viability percentage in all heat treatments in relation to its control. In summary, 81% of the community was R+PT (resprouters with heat-tolerant propagules), 17% was R+PS (resprouters with heat-stimulated propagules) and 2% was R+P- (resprouter with heat-sensitive propagules) regarding regeneration strategies in face of fires (Table 1, Fig. 2). Moreover, species with heat-stimulated propagules were evenly distributed among underground, basal and aerial resprouters (Fig. 3).

Table 1. Family, species, growth form, initial viability, dormancy class or nondormancy (PY= physical dormancy, PD = physiological dormancy, MPD = morphophysiological dormancy, ND = nondormant), bud location (aerial, basal, underground), and classification of regeneration strategy according to resprouting and seed germination responses to heat: R+ PT = *resprouter and heat-tolerant propagules;* R+PS = *resprouter and heat-stimulated propagules;* R+P- = *resprouter and heat-sensitive propagules.* *type of dormancy is inferred from information on germination and on characteristic of seeds in that family.

Family	Species	Growth form	Initial viability (%)	Dormancy	Bud location	Regeneration strategy
Asteraceae	*Aldama grandiflora* (Gardner) E.E. Schill. & Panero	Forb	56	ND	underground	R+PT
	Lessingianthus buddleiifolius (Mart. ex DC.) H. Rob.	Shrub	30	ND	underground	R+PT
	Lessingianthus floccosus (Gardner) H. Rob.	Shrub	26	ND	underground	R+PT
	Lessingianthus hoveaefolius (Gardner) H. Rob.	Shrub	13	ND	underground	R+PT
Bromeliaceae	*Dyckia brasiliana*[1] L.B. Sm.	Forb	68	ND	undetermined	R+PT
Calophyllaceae	*Kielmeyera rubriflora* Cambess.	Shrub	29	ND	underground	R+PT
Cyperaceae	*Bulbostylis paradoxa* (Spreng.) Lindm.	Graminoid	84	ND	aerial	R+PT
Dilleniaceae	*Davilla elliptica* A. St.-Hi.	Shrub	85	MPD*	aerial	R+PT
Fabaceae	*Bauhinia dumosa*[4] Benth.	Shrub	98	ND	underground	R+PT
	Chamaecrista claussenii (Benth.) H.S. Irwin & Barneby	Shrub	66	PY	underground	R+PT
	Chamaecrista ochrosperma[2] (H.S. Irwin & Barneby) H.S. Irwin & Barneby	Shrub	84	PY	underground	R+PT
	Harpalyce brasiliana Benth.	Shrub	51	PY	underground	R+PT
	Mimosa claussenii[2] Benth.	Shrub	80	PY	aerial	R+PT
	Mimosa kalunga[2] Marc. F. Simon & C.E. Hughes	Shrub	48	PY	aerial	R+PS
	Mimosa leiocephala[4] Benth.	Shrub	74	PY	underground	R+PS
	Mimosa pteridifolia[3] Benth.	Shrub	60	ND	underground	R+P-
	Mimosa gracilis Benth.	Forb	47	ND	underground	R+PT
	Mimosa[2] sp1	Shrub	91	PY	aerial	R+PT
	Mimosa[2] sp2	Shrub/Tree	100	ND	aerial	R+PT
	Senna cana[2] (Nees & Mart.) H.S. Irwin & Barneby	Shrub/Tree	90	PY	undetermined	R+PS

	Senna corifolia[2] (Benth.) H.S. Irwin & Barneby	Shrub	81	PY	underground	R+PT
	Senna silvestris[2] (Vell.) H.S. Irwin & Barneby	Shrub/Tree	84	PY (intermediate)	undetermined	R+PT
	Stylosanthes sp.	Shrub	100	PY (intermediate)	underground	R+PS
Gentianaceae	*Deianira pallescens* Cham. & Schltdl.	Forb	81	ND	underground	R+PT
Iridaceae	*Trimezia juncifolia* (Klatt) Benth. & Hook. f.	Forb	73	ND	underground	R+PT
Lamiaceae	*Cyanocephalus coriaceus* (Benth.) Harley & J.F.B. Pastore	Sub-shrub	82	ND	underground	R+PT
	Hypenia brachystachys (Pohl ex Benth.) Harley	Sub-shrub	22	ND	underground	R+PT
	Hypenia sp.	Forb	71	ND	underground	R+PT
	Medusantha mollissima (Benth.) Harley & J.F.B. Pastore ex Benth.	Sub-shrub	66	ND	underground	R+PT
Lythraceae	*Diplusodon punctatus* Pohl	Shrub	53	PD	underground	R+PS
Malvaceae	*Melochia* sp.	Shrub	86	PY	underground	R+PS
Melastomataceae	*Tibouchina melastomoides*[3] (Naudin) Cogn.	Forb	44	ND	underground	R+PT
Poaceae	*Elionurus* sp.	Graminoid	88	ND	basal	R+PT
	Aristida riparia Trin.	Graminoid	35	PD	basal	R+PT
	Aristida sp.	Graminoid	42	PD	basal	R+PS
	Axonopus aureus	Graminoid	72	ND	underground	R+PT
Velloziaceae	*Vellozia glochidea*[4] Pohl	Subshrub	99	ND	aerial	R+PT
	Vellozia squamata[4] Pohl	Shrub	57	ND	aerial	R+PT
	Vellozia tubiflora[4] (A.Rich.) Kunth	Shrub	48	ND	aerial	R+PT
Verbenaceae	*Lippia lupulina* Cham.	Sub-shrub	25	ND	underground	R+PT
Xyridaceae	*Xyris* sp.	Graminoid	81	ND	basal	R+PT

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[1] Experimental data from R.B. Novaes (2015)

[2] Experimental data from J.S. Nunes (2016)

[2] Experimental data from H.L. Zirondi (2015)

[3] Experimental data from H.L. Zirondi et al. (unpubl. data)

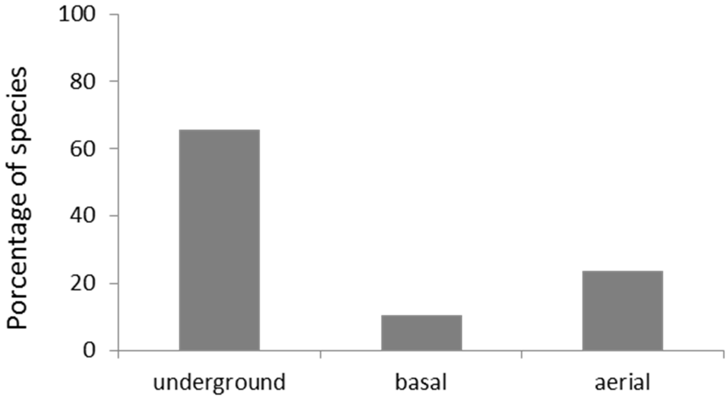

Figure 1. Porcentage of species with resprouts originating from underground, basal and aerial buds in the community (n=38)

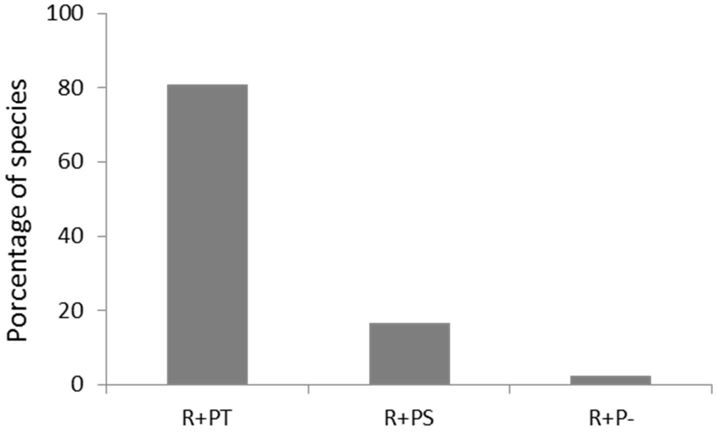

Figure 2. Porcentage of species that were classified as R+PT (resprouter and heat-tolerant propagules), R+PS (resprouter and heat-stimulated propagules) and R+P- (resprouter and heat-sensitive propagules) in the community (n=41)

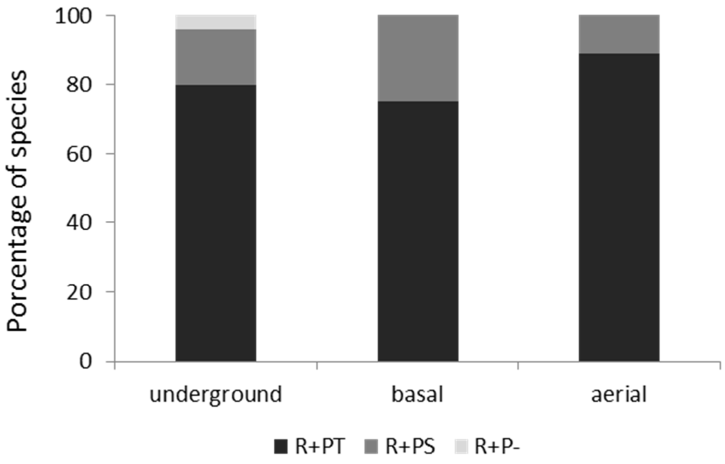

Figure 3. Porcentage of species that were classified as R+PT (resprouter and heat-tolerant propagules), R+PS (resprouter and heat-stimulated propagules) and R+P- (resprouter and heat-sensitive propagules) separately for each resprouting strategy according to bud location (underground, basal or aerial)

Twenty-five species (61%) were non-dormant and sixteen species (39%) were dormant, of which two species were classified with intermediate dormancy (Table 1, Fig. 4a). Among dormant species, twelve species (75%) had physical dormancy (PY), three species (19%) had physiological dormancy (PD) and only one had morpho-physiological dormancy (MPD, Fig. 4b). Most dormant species (59%) were classified as heat-tolerant (PT) and 41% were heat-stimulated (PS); all species that had heat-stimultaed propagules had dormant seeds (Fig. 5).

Most species survived exposure of 100°C for one and three minutes, but nearly 50% of species decreased their germination and viability in the 200°C treatment (Fig. 6). Overall, non-dormant species had germination percentages over 50% in the control and heat treatments of 100°C for one and three minutes, but six species had germination percentages below 30% in all treatments (*Hypenia brachstachys, Kielmeyera rubriflora, Lessingianthus buddleifolius, Lessingianthus floccosus, Lessingianthus hovaeafolius* and *Lippia lupulina,* Table 2). For 56% of non-dormant species, the germination and viability percentage was not affected by any of the heat treatments, but for 44% of species, heat shocks of 200°C decreased both germination and viability percentages. For dormant species, the germination percentages in the control were below 20% for all species, except *Senna silvestris* and *Stylosanthes* sp. (over 40%). Additionally, heat treatments of 200^{o}C decreased seed viability of five (nearly 30%) species (*Harpalyce brasiliana, Mimosa claussenii*, *Mimosa kalunga*, *Melochia* sp. and *Senna silvestris*) in relation to their control.

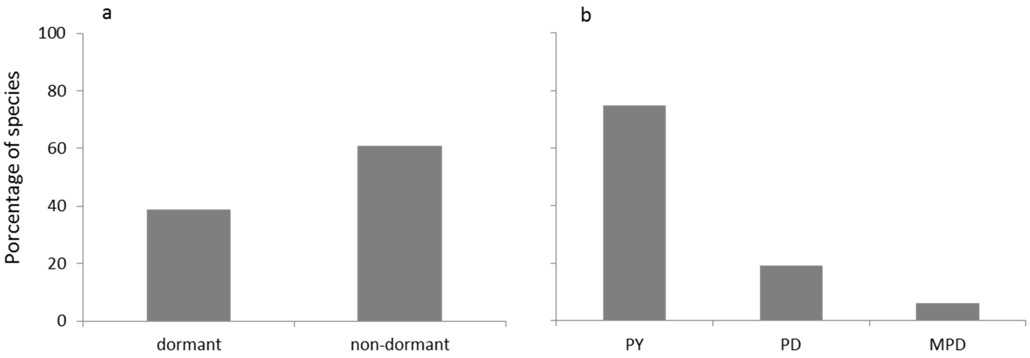

Figure 4. a) Porcentage of species that were classified as dormant and non-dormant in the community (n=41) and b) percentage of species in each of the following dormancy classes: physical dormancy (PY), physiological dormancy (PD) and morpho-physiological dormancy (MPD)

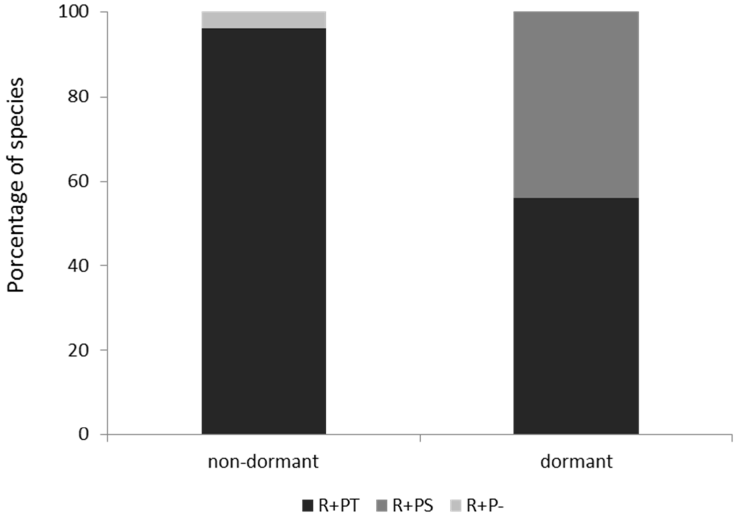

Figure 5. Porcentage of species that were classified as R+PT (resprouter and heat-tolerant propagules), R+PS (resprouter and heat-stimulated propagules) and R+P- (resprouter and heat-sensitive propagules) separately for dormant and non-dormant species

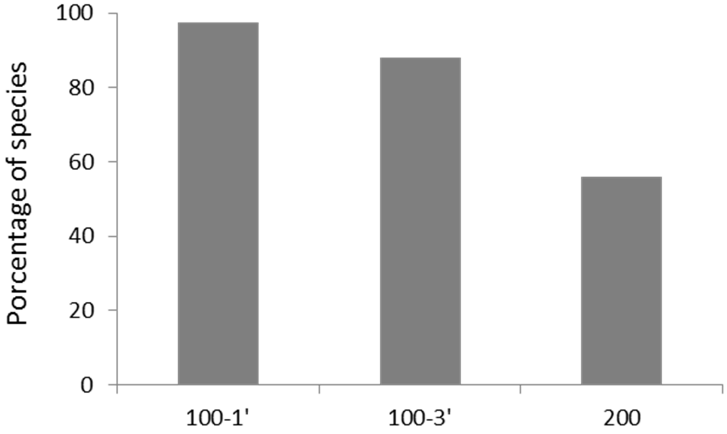

Figure 6. Porcentage of species that survived heat-shocks of 100^{o}C - 1’, 100^{o}C - 3’ and 200^{o}C in the community (n=41)

Table 2. Seed germination and seed viability (%, mean ± SE) of 25 species after being exposed to heat shocks of 100°C – 1’, 100°C - 3’, 200°C - 1’. Control refers to untreated seeds. Different letters mean significant differences (p≤0.05) among treatments; statistical comparisons were performed separately for each species.

Species	Germination				Viability			
	control	100°C - 1’	100°C - 3’	200°C - 1’	control	100°C - 1’	100°C - 3’	200°C - 1’
Aldama grandiflora	56 ± 6 a	62 ± 12 a	31 ± 18 ab	11 ± 11 b	56 ± 6 a	62 ± 12 a	31 ± 18 ab	11 ± 11 b
Lessingianthus buddleiifolius	30 ± 2 a	35 ± 3.3 a	25 ± 7 ab	11 ± 6 b	30 ± 2 a	35 ± 3 a	25 ± 7 ab	11 ± 6 b
Lessingianthus floccosus	24 ± 3 a	24 ± 4 a	7 ± 4 b	0 ± 0 b	26 ± 3 a	24 ± 4 a	8 ± 4 b	0 ± 0 b
Lessingianthus hoveaefolius	12 ± 8	10 ± 6	2 ± 2	0 ± 0	13 ± 8	10 ± 6	2 ± 2	0 ± 0
Kielmeyera rubriflora	29 ± 8	22 ± 4	26 ± 8	12 ± 6	29 ± 8	22 ± 4	26 ± 8	12 ± 6
Bulbostylis paradoxa	81 ± 6	74 ± 5	75 ± 12	67 ± 17	84 ± 4	75 ± 5	85 ± 6	80 ± 10
Davilla elliptica	0	0	0	0	85 ± 1 a	89 ± 3 a	84 ± 4 a	3 ± 2 b
Chamaecrista claussenii	18 ± 3	11 ± 4	5 ± 4	11 ± 6	66 ± 5 a	54 ± 5 a	18 ± 6 ab	51 ± 8 b
Harpalyce brasiliana	10 ± 4	20 ± 13	25 ± 6	18 ± 9	51 ± 6 a	23 ± 12 ab	27 ± 6 b	18 ± 9 b
Mimosa gracilis	44 ± 9 a	42 ± 8 a	41 ± 7 a	5 ± 5 b	49 ± 10 a	44 ± 9 a	45 ± 8 a	10 ± 9 b
Stylosanthes sp.	45 ± 10 a	46 ± 9 a	77 ± 7 b	94 ± 2 b	100 ± 0	98 ± 1	98 ± 1	94 ± 2
Deianira pallescens	64 ± 11	72 ± 4	58 ± 7	51 ± 8	81 ± 5	83 ± 5	82 ± 3	62 ± 6
Trimezia juncifolia	69 ± 9 a	76 ± 6 a	71 ± 11 a	7 ± 5 b	73 ± 10 a	81 ± 6 a	85 ± 4 a	12 ± 4 b
Cyanocephalus coriaceus	82 ± 4 a	66 ± 6 a	71 ± 10 a	49 ± 13 b	82 ± 4 a	66 ± 6 a	73 ± 9 a	51 ± 11 b
Hypenia brachystachys	22 ± 7	21 ± 6	21 ± 2	14 ± 5	22 ± 7	21 ± 6	21 ± 2	14 ± 5
Hypenia sp.	71 ± 10	74 ± 3	81 ± 6	54 ± 9	71 ± 10	75 ± 3	82 ± 5	56 ± 9
Medusantha mollissima	66 ± 9	66 ± 11	58 ± 4	39 ± 16	66 ± 9	66 ± 11	58 ± 4	39 ± 16
Diplusodon punctatus	9 ± 2 a	24 ± 5 b	15 ± 4 ab	26 ± 6 b	52 ± 6	58 ± 9	51 ± 10	53 ± 4
Melochia sp.	12 ± 3 a	10 ± 3 a	46 ± 16 ab	43 ± 8 b	86 ± 2 a	92 ± 3 a	75 ± 8 ab	57 ± 11 b
Elionurus sp.	81 ± 3 a	82 ± 2 a	88 ± 1 a	52 ± 10 b	85 ± 4 a	82 ± 2 a	88 ± 1 a	52 ± 10 b
Aristida riparia	4 ± 3	0 ± 0	10 ± 6	12 ± 6	35 ± 10	42 ± 9	34 ± 10	48 ± 7
Aristida sp.	4 ± 1 a	8 ± 5 a	22 ± 12 a	27 ± 7 b	41 ± 9	45 ± 6	28 ± 11	40 ± 8
Axonopus aureus	66 ± 6	80 ± 5	72 ± 7	76 ± 11	72 ± 6	80 ± 5	75 ± 5	80 ± 9
Lippia lupulina	21 ± 8 ab	28 ± 4 a	11 ± 6 ab	7 ± 4 b	25 ± 9	31 ± 4	17 ± 7	11 ± 6
Xyris sp.	79 ± 7 a	80 ± 11 ab	-	48 ± 39 b	81 ± 2 a	81 ± 4 ab	-	62 ± 15 b

Discussion

All species were able to resprout after fires (R+) and non-resprouting obligate seeding species (R-) were not observed, indicating that resprouting is the main post-fire regeneration strategy in the Cerrado open savanna. Post-fire resprouting and post-fire seeding are considered independent traits, in which species can present one trait, both or none (Pausas et al. 2004; Pausas and Keeley 2014). Although post-fire seeding is more common in ecosystems that undergo high intensity crown fires (Pausas and Keeley 2014; Clarke et al. 2015), such strategy has been observed in fire-prone ecosystems that are subjected to surface fires: around 7% of species were considered obligate seeders in subtropical grasslands of Southern Brazil (Overbeck and Pfadenhauer 2007), while in the Australian savannas, nearly 50% of species are facultative resprouters, displaying both post-fire resprouting and seeding, and 7% are obligate seeders (Clarke et al. 2015). However, only 17% of our community (7 species) had heat-stimulated propagules (R+PS), while the vast majority of species had heat-tolerant propagules, but were not stimulated (R+PT; 33 species), suggesting that post-fire seeding plays a minor role in post-fire regeneration in the Cerrado open savannas. Nonetheless, germination triggered by fire-related cues had not been previously observed among Cerrado species (Le Stradic et al. 2015; Fichino et al. 2016; Ramos et al. 2016), and our results suggest that such strategy may be coupled with resprouting as a post-fire regeneration strategy for some species.

The selective factors driving the evolution of resprouting and seeding strategies in fire-prone environments have been subject to much debate (Bond and Midgley 2003; Keeley et al. 2011; Pausas and Keeley 2014). It has been suggested that sites with high resource availability and productivity favor resprouting species because post-fire regeneration is very rapid and plants quickly reoccupy the space (Clarke et al. 2005, 2013; Pausas and Keeley 2014). Thus, under such conditions resprouters would have a competitive advantage since growth responses are almost immediate (Lamont et al. 2011). Additionally, climate and soil productivity interact through different mechanisms in affecting fire regimes, especially fire frequencies (Pausas and Bradstock

2007; Pausas and Ribeiro 2013). Although the Cerrado system has been recognized to have nutrient poor soils (Ratter et al. 1997), its climate is typical of the moister savanna regions of the world, with an average annual precipitation of 800-2000 mm (Ratter et al. 1997) and a productivity of around 900 g.m^{-2} two years after fire (Rissi 2016). In such nutrient poor soils, increasing moisture leads to higher fire frequencies as time to reach critical fuel levels is reduced (Pausas and Ribeiro 2013; Wuest et al. 2016). Environments with high disturbance frequencies, such as the Cerrado (Pereira Júnior et al. 2014; Alvarado et al. 2017), have also been suggested to favor resprouting species, mainly because frequent disturbances inhibit seedling recruitment to maturity (Bellingham and Sparrow 2000; Bond & Midgley 2003).

Alternatively, sites with slow post-fire regeneration and large and long lasting post-fire gaps, where competition for resources is reduced, will select for seedling recruitment over resprouting (Keeley et al. 2016). This could explain the lack of obligate seeding species and the low number of species that could potentially regenerate after fires by both strategies (facultative resprouters) in our community. The vigorous resprouts, especially of the abundant grasses that dominate the Cerrado open savannas, quickly close post-fire gaps, and one year after fires bare soil gaps are rare and usually small (personal observation). However, bare soil gaps have been shown to influence seed germination due to enhanced daily temperature fluctuations in Cerrado open savannas (Daibes et al. 2017).

Furthermore, post-fire seeding is coupled with the acquisition of a fire resistant seed bank and delaying germination to a single point in time (i.e. after fires) when more resources are available (Pausas and Keeley 2014). Seed dormancy, especially physical dormancy, can thus be considered a key trait in fire-prone ecosystems as it can be related to post-fire recruitment (Keeley et al. 2011; Ooi et al. 2014). Although all species that had heat-stimulated propagules in our community had dormant seeds, most of which were physically dormant, the percentage of dormant species (around 40%) is relatively low compared to the expected percentage for savanna communities (c.a. 70%, Baskin and Baskin 2014), but similar to what has been found for Cerrado *campo rupestre* (Dayrell et al. 2017). Moreover, less than half of the dormant species showed heat-stimulated propagules, which might indicate that the presence of seed dormancy in this system is most likely not linked to fire recruitment, as has been suggested for the Mediterranean fire-prone ecosystems (Moreira and Pausas 2012; Ooi et al. 2014; Pausas and Keeley 2014), but rather be driven by phylogeny (Dayrell et al. 2017).

Additionally, few Cerrado species form persistent seed banks (Velten and Garcia 2007; Salazar et al. 2011) and recruitment from the seed bank does not seem to be linked to fire, but rather to the onset of the rainy season (Andrade and Miranda 2014; Escobar & Cardoso 2015). Nonetheless, all species (except for *M. pteridifolia*) were resistant to one-minute heat shocks of 100^{o}C and most could resist three-minute heat shocks of 100^{o}C, suggesting seed tolerance to these temperatures (R+PT), as has been observed for species of other fire-prone grassy ecosystems (Gashaw and Michelsen 2002; Clarke and French 2005; Dayamba et al. 2008), including the Cerrado (Le Stradic et al. 2015; Fichino et al. 2016; Ramos et al. 2016). During Cerrado fires, temperatures 1cm belowground are negligible, rarely increasing over 80^{o}C (Zupo et al. unpubl. data), thus, fire-tolerance could be an important seed trait as seeds that are occasionally stored in the soil seed bank are able to survive a fire episode and recruit in the following rainy season.

On the other hand, seeds of nearly 50% of species showed lower germination and viability percentages when exposed to 200^{o}C for one minute. Thus, recently dispersed seeds that have not been incorporated to the soil seed bank at the time of fire are likely to be killed (Daibes et al. 2017) as temperatures on the soil surface frequently exceed 200^{o}C (Pivello et al. 2010; Gorgone-Barbosa et al. 2015). In fact, it is more likely that seedling establishment of resprouting species occur in the intervals between fires (Pausas and Keeley 2014), thus, the ability of juveniles to resprout in such frequent fire regimes is essential for successful establishment. Most species resprouted from underground bud banks, which are highly protected since they are insulated by soil (Choczynska and Johnson 2009; Clarke et al. 2013). These buds are usually found on specialized structures, such as xylopodia and underground stems, constituting the geoxyle growth form, or underground trees, present in both the Cerrado (Simon et al. 2009; Simon and Pennington 2012) and in African savannas (Maurin et al. 2014), but lacking among taxa from the Australian savannas (Clarke et al. 2015). Maurin et al. (2014) suggest that the geoxylic growth form is advantageous in areas of high precipitation and frequent fires, as the moist savannas of Africa and Brazil.

Furthermore, around 25% of non-dormant species had low germination and viability percentages under control conditions, mainly due to a great amount of embryo-less seeds (data not shown), as has been observed for other Cerrado communities (Le Stradic et al. 2015; Dayrell et al. 2017). Lamont et al. (2011) argue that low fecundity among resprouting species may be due to accumulation of deleterious somatic mutations, given that resprouters live through many fire cycles and accumulation of mutant alleles and chromosomal aberration is a time-dependent process. Thus, low seed sets may be an inevitable consequence of the longevity of resprouters. However, resprouting can enhance fitness where seed production and seedling recruitment are limited, given that resprouters regenerate rapidly and produce seeds more quickly (Lamont et al. 2011, Keeley et al. 2011, Pausas & Keeley 2014). Nonetheless, resprouting can have major impacts on plant populations as population turnover is reduced and dependence on seeds for maintaining populations seems to be of minor importance (Bond and Midgley 2001), suggesting that perhaps the ultimate role of seeds, given that resprouting is the main regeneration mechanism, would be dispersal rather than recruitment.

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Capítulo 2

Do different disturbance types affect resprouting and flowering responses of Cerrado species?

Talita Zupo & Alessandra Fidelis

Abstract

Resprouting is a key regenerative trait that allows species to persist in face of many different disturbances, including fire. However, unlike other disturbances, the high temperatures associated with fire can be lethal to meristematic tissue, potentially affecting resprouting responses. We evaluated how different disturbance types affected the resprouting responses and flowering of nine plant species from a Cerrado open savanna in Central Brazil. Individuals of four shrubs, two forbs and three grass species were permanently tagged and subjected to clipping at base or fire. We measured number of shoots and height (shrubs and forbs) and diameter and height (grasses) prior to disturbance and three, six and twelve months after treatments were applied. Presence or absence of reproductive shoots or tillers was also accounted for. Individuals of all species were able to resprout after both disturbance types, mostly from buds located belowground, thus protected by soil. Additionally, fire stimulated production of new shoots for most shrub species and probability of flowering in two shrub and one forb species. Our results help explain the mechanisms of fire effects in Cerrado, suggesting that such fires cause no damage to the belowground bud bank and that for some species, fire, and not only aboveground biomass removal, determines resprouting success, which may be due to the increased resource availability and reduced competition of the post-fire environment.

Keywords: Clipping, fire, flowering, Neotropical savanna, regeneration, resprouting

Introduction

Resprouting is defined as the ability of a plant species to form new shoots after the loss of part or most of its aboveground biomass (Cornelissen et al. 2003), thus being a key regenerative trait that allows species to persist after a wide range of disturbances (Bond and Midgley 2001, 2003; Vesk and Westoby 2004; Clarke et al. 2013; Pausas et al. 2016). Resprouting increases fitness, especially in ecosystems where disturbance frequencies are high and recruitment is low, as resprouters regenerate rapidly and produce seeds more quickly (Bellingham and Sparrow 2000; Lamont et al. 2011; Keeley et al. 2011). It is a common trait in many plants throughout the world and considered an ancient trait among angiosperms (Bond and Midgley 2003; Keeley et al. 2012).

For a plant to be able to resprout after a disturbance, it needs both surviving meristems (dormant buds or bud forming tissues) and stored reserves that support regrowth (Bond and Midgley 2001, 2003; Lamont et al. 2011; Clarke et al. 2013). Bud location and protection, as well as bud number and amount of resources available to fund regrowth are important drivers of resprouting ability, however, it is a complex trait associated with several mechanisms under different disturbance regimes (Clarke et al. 2013).

Although resprouting has been studied as a response to different disturbance types, such as wind, drought, herbivory and, especially, fire (Bond and Midgley 2001, 2003; Vesk and Westoby 2004; Clarke et al. 2013; Pausas and Keeley 2014; Zeppel et al. 2015), there are major differences among these disturbances which would limit generalizations (Pausas et al. 2016). The high temperatures associated with fire can be lethal to meristematic tissue (buds and cambium), and plants that are able to resprout after other disturbances may not be able to resprout after fire (Vesk and Westoby 2004; Vesk et al. 2004). Thus, in fire-prone ecosystems, plants resprouting from aboveground parts (epicormic buds) must protect their buds with bark or have their buds located deep in the wood, as do many woody species present in the Australian, African and Neotropical savannas (Burrows et al. 2010; Dantas and Pausas 2013; Charles-Dominique et al. 2015), while other species have belowground bud banks located in specialized structures (e.g. xylopodia and lignotubers) that are highly protected as they are insulated by soil (Simon et al. 2009; Simon and Pennington 2012; Clarke et al. 2013; Fidelis et al. 2014). Furthermore, fire, unlike other disturbances, reduces competition and enhances resource (water, nutrient, light and space) availability (Whelan 1995; Neary et al. 1999), creating a favorable environment for vegetative recovery.

Resprouting is also strongly influenced by the disturbance regime (Whelan 1995; Bond and Midgley 2001; Vesk and Westoby 2004; Clarke et al. 2013), where frequency, intensity, and severity can affect resprouting responses. Different fire regimes can affect resprouting by altering production of new shoots, where increasing fire frequency, intensity and severity would decrease resprouting success, either by permanently damaging dormant buds or by depleting resources stored for regrowth (Canadell and Lopez-Soria 1998; Vesk and Westoby 2004; Wright and Clarke 2007; Enright et al. 2011). On the other hand, clipping, which has been used as a fire surrogate without the lethal effects of fire (high temperatures), would only damage aboveground biomass, leaving the bud bank found in underground organs intact and thus be characterized as less intense/severe disturbance (Cruz et al. 2003; Vesk and Westoby 2004; Calvo et al. 2005). Thus, in many fire-prone ecosystems subjected to intense fire regimes, clipped plants usually show a higher resprouting response in relation to burned plants (Vesk and Westoby 2004, Vesk et al. 2004). However, factors present in the post-fire environment, such as decreased competition and enhanced resource availability (Whelan 1995; Neary et al. 1999), or even species-specific traits (as for example, the presence of belowground bud banks, Clarke et al. 2013) might be related to the success in plant regeneration after fire.

Fire has been present in Cerrado for millions of years (Simon et al. 2009; Simon and Pennington 2012) and fire regimes, as in other savanna ecosystems, are characterized by frequent fires of relatively low intensity fueled by grasses (surface fires, Miranda et al. 2002). However, these fires can be considered severe since they consume most of the aboveground biomass (Miranda et al. 2009; Rissi 2016). For many species in Cerrado, new shoots can be produced from dormant buds located in well-developed underground structures (Appezzato-da-Glória et al. 2008; Simon et al. 2009; Simon and Pennington 2012) and aboveground biomass is quickly recovered (1-2 years, Rissi 2016), enabling frequent fires. The aim of this study was to test how different disturbance types affected the resprouting and flowering responses of dominant Cerrado species belonging to different functional groups, using clipping and burning as experimental treatments. Since Cerrado fires are frequent and of low intensity, resprouting responses between burned and clipped plants should be similar (Vesk and Westoby 2004). Thus, comparing plant regeneration of clipped and burned plants would help understand plant responses to fire and gain insight into what drives resprouting responses in these ecosystems.

Material and methods

Study Area

The study was conducted at the Serra do Tombador Nature Reserve (RNST), located in Central Brazil (47°45'; 47°51' W and 13°35'; 13°38' S, 8900 ha, 560-1118 m a.s.l). The regional climate is tropical with wet summers and dry winters with an average annual rainfall between 1300 and 1500 mm (Fundação Grupo Boticário 2011). The vegetation of the study area is characterized as a *campo sujo,* an open savanna with a continuous herbaceous layer dominated by grasses and scattered shrubs (Coutinho 1978), which produces fine fuel loads that are able to burn frequently. The RNST faces natural and anthropogenic fires and data for 2001-2010 indicated that fires were most frequent in grassland and savanna sites and typically occurred in a 3-yr interval (Daldegan et al. 2014).

Study species and treatments

Nine dominant species belonging to different functional groups were selected for this study: *Chamaecrista claussenii* (Benth) H.S. Irwin & Barneby, *Harpalyce brasiliana* Benth., *Mimosa leiocephala* Benth. and *Bauhinia dumosa* Benth. (all Fabaceae shrub species); *Mimosa gracilis* Benth. (Fabaceae) and *Ipomoea fiebrigii* Hassl. ex O'Donell (Convolvulaceae, forb species); and the three grass species were *Mesosetum ferrugineum* (Trin.) Chase, *Mesosetum loliiforme* (Hochst. ex Steud.) Chase and *Oncorachis ramosa* (Zuloaga & Soderstr.) Morrone & Zuloaga (Poaceae). All shrub and forb species have buds on underground structures insulated by soil, two grass species (*M. lolliforme* and *O. ramosa*) are rhizomatous with belowground buds and one (*M. ferrugineum*) is caespitose with buds on the soil surface protected by leaf bases.

Treatments consisted of clipping at base or subjecting to prescribed burnings during experimental fires conducted in the area. Both treatments were applied in July (2013), mid-dry season, in a *campo sujo* area that had last been subjected to fire two years earlier (2011). We clipped plants at the base, since the study species resprout from basal (*M. ferrugineum*) or belowground bud banks (all other species), and did not remove the surrounding vegetation around clipped individuals. For each treatment (burned and clipped), ten individuals of each species were randomly selected, permanently tagged and measured prior to disturbance, and three, six and twelve months after the disturbance. In both treatments, marked individuals of the same species were at least 1m apart from each other. In all surveys we measured maximum height and total number of shoots for shrubs and forbs, and height and diameter of tussocks for grass species. For all species, presence or absence of reproductive shoots or tillers was accounted for in every survey carried out.

Experimental fires were head fires and conducted with the assistance of the reserve's fire brigade being carried out at the beginning of the day for safety reasons. Mean fire intensity was 2707±1899 kW.m^{-2} and mean rate of spread was 0.23±0.1 m.s^{-1} (Rissi et al. 2017). After fires, all aboveground biomass was dead, either by being totally consumed (grasses and forbs) or by having tissue death from high temperatures (T. Zupo pers. obs).

Statistical analyses

We used mixed models to analyze post-disturbance resprouting response. To estimate differences in total number of shoots we used Generalized Linear Mixed Models (GLMMs, Zuur et al. 2009) with a Poisson distribution. To estimate differences in diameter and height, we used linear mixed models (LMMs, Zuur et al. 2009) with a Gaussian distribution after log transformation to fit model assumptions. We fitted GLMMs or LMMs for each species separately with treatment and time as fixed effects, and pre-disturbance size (total number of shoots – or diameter - and height prior to disturbance) as a covariate. We fitted a random intercept model with individuals as random factors. The fire treatment was taken as the baseline for the intercept in the models. GLMMs were fitted by maximum likelihood assuming a Laplace approximation to the likelihood function and LMMs were fitted by maximum likelihood. All the analyses were carried out with the ''lme4'' package (Bates et al. 2015) in R software (R Core Development Team 2016).

To evaluate how the treatments affected flowering, we used GLMs with a binomial distribution, given that presence of flowering was a binary data. We fitted the GLMs for each species separately using the subsequent flowering period for each species, except for *M. lolliforme* since we did not observe reproductive tillers in any of the census carried out. The subsequent flowering period for *M. ferrugineum* and *O. ramosa* was in October 2013, for *H. brasiliana* and *M. gracilis* in February 2014, for *M. leiocephala* in May 2014 and for *B. dumosa*, *C. clausenii*, *and I. fiebrigii* in July 2014. The logits for the intercept (baseline) refer to the fire treatment.

Results

Individuals of all species were able to resprout after both clipping and burning. In general, treatment had a strong effect on total number of shoots and height for shrub species, where burned individuals resprouted with a greater number of shoots and height ($p \leq 0.05$, Table 1). Three months after treatments were applied, all shrub species (except for *Mimosa leiocephala*, Table 1, Fig. 1a), showed at least 2-fold more shoots after fires in relation to being clipped (*B. dumosa:* 9.5±1.3 and 3.3±0.9 shoots; *H. brasiliana*: 8.4±2.1 and 2.3±0.4; *C. claussenii*: 3±0.4, and 1.5±0.5, for burned and clipped individuals, respectively, Fig. 1b-d). The number of resprouting shoots decreased significantly over time only for *H. brasiliana*, from 8.1±2.1 shoots three months after fire to 4.1±0.6 shoots 12 months after fire ($p<0.0001$, Fig. 1c). The number of shoots of both forb species, *M.*

gracilis and *I. fiebrigii*, did not differ significantly between treatments or over time (Table 1, Fig. 2 a-b).

As expected, height of all shrub and forb species was strongly affected by time, with maximum height increasing significantly over time (Table 1, Fig. 1 and 2). Among shrub species, maximum height differed between burned and clipped individuals only for two species, with taller resprouting shoots in burned individuals (three months after fire - *H. brasiliana:* 45.9±4.2 cm and 18.2±3.2 cm; and *C. claussenii:* 36±3.6 cm and 21.3±5 cm, burned and clipped individuals, respectively, p=0.01 and p=0.004; Fig. 1c and d). Additionally, maximum height of both forb species was not affected by treatment: 12 months after treatments were applied, *I. fiebrigii* showed 27±2 cm and 32.9±2 cm in burned and clipped individuals, respectively, and *M. gracilis* showed 60.4±5.1 cm and 51.4±7.4 cm in burned and clipped individuals, respectively (Fig. 2).

In general, grass species showed no significant differences between treatments for diameter of tussocks, where clipped individuals regenerated just as burned ones (Table 2, Fig. 3). Moreover, diameter of both *O. ramosa* and *M. lolliforme* increased over time, in both burned and clipped individuals (Fig. 3a-b). Height of all grass species also increased over time, but for *O. ramosa* and *M. ferrugineum*, burned individuals were taller than clipped ones (Table 2, Fig. 3a and c).

For three species, clipping treatments had a negative effect on flowering in relation to fire (Table 3). Burned individuals of the shrub species *H. brasiliana* and *C. claussenii* had significantly greater chances of flowering (70% for both) in relation to clipped individuals (10 and 20%, respectively). The probability of flowering for the forb species *M. gracilis* was 70% after fire and only 20% after clipping (Table 3). No significant differences in flowering were found for grass species between burned and clipped plants (Table 3).

Table 1 Parameters estimated by the mixed models for total number of shoots (GLMMs) and height (LMMs) of shrub and forb species in relation to treatments (burned and clipped), time (3, 6 and 12 months after disturbance) and pre-disturbance size (total number of shoots prior to disturbance). Significant coefficients (p≤0.05) are given in bold font.

		Number of shoots			**Height**		
Species	Effects	Coefficients	Standard error	*P* values	Coefficients	Standard error	*P* values
Mimosa leiocephala	intercept	1.79	0.26	<0.0001	1.82	1.00	0.02
	treatment clipped	-0.22	0.18	0.2	-0.11	0.08	0.16
	time	-0.02	0.02	0.14	**0.08**	0.01	0.001
	pre-disturbance size	0.01	0.05	0.9	0.4	0.21	0.06
Bauhinia dumosa	intercept	1.91	0.22	<0.0001	-0.46	1.79	0.79
	treatment clipped	**-0.76**	0.14	<0.0001	-0.36	0.21	0.1
	time	-0.03	0.01	0.06	**0.11**	0.03	0.0003
	pre-disturbance size	**0.09**	0.04	0.03	**0.79**	0.37	0.04
Harpalyce brasiliana	intercept	1.59	0.22	<0.0001	1.46	1.42	0.31
	treatment clipped	**-0.85**	0.19	<0.0001	**-0.37**	0.14	0.01
	time	**-0.06**	0.02	<0.0001	**0.08**	0.02	<0.0001
	pre-disturbance size	**0.2**	0.05	<0.0001	0.48	0.33	0.15
Chamaecrista claussenii	intercept	0.66	0.33	0.05	3.62	1.47	0.02
	treatment clipped	**-0.69**	0.18	0.0001	**-0.56**	0.17	0.004
	time	0.02	0.02	0.49	**0.06**	0.02	0.01
	pre-disturbance size	0.16	0.11	0.17	-0.09	0.39	0.81
Mimosa gracilis	intercept	-0.27	0.3	0.373	-5.05	2.58	0.06
	treatment clipped	-0.47	0.27	0.083	-0.57	0.27	0.051
	time	0.04	0.03	0.162	**0.18**	0.03	<0.0001
	pre-disturbance size	**0.21**	0.06	0.0002	**1.83**	0.65	0.012
Ipomoea fiebrigii	intercept	1.46	0.30	<0.0001	1.85	1.25	0.147
	treatment clipped	-0.34	0.20	0.093	-0.10	0.15	0.491
	time	-0.02	0.01	0.071	**0.04**	0.01	0.014
	pre-disturbance size	**0.12**	0.03	0.0001	0.34	0.335	0.351

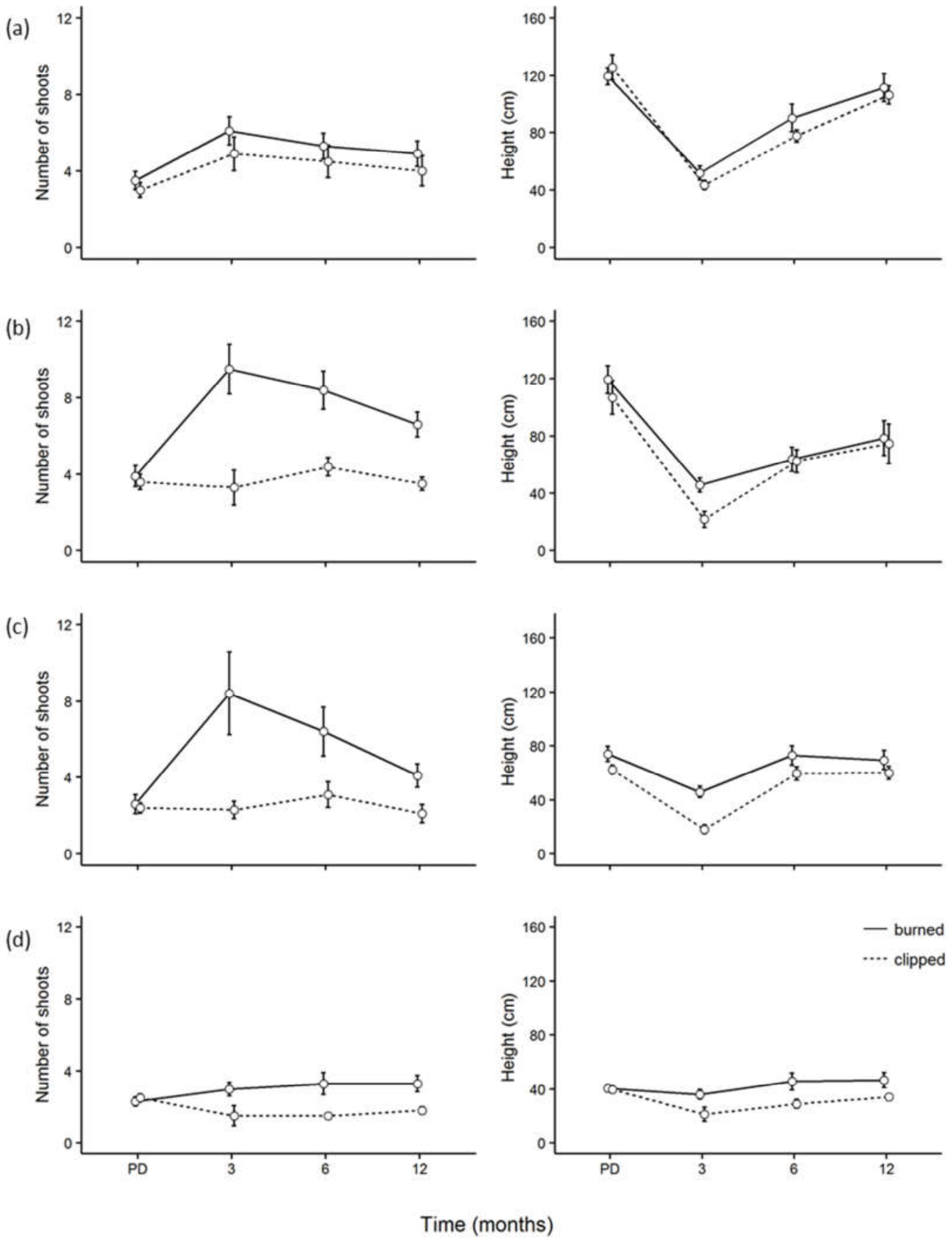

Figure 1. Number of shoots and height (mean ± SE) for a) *Mimosa leiocephala*, b) *Bauhinia dumosa*, c) *Harpalyce brasiliana* and d) *Chamaecrista claussenii* in burned (solid line) and clipped (dashed line) individuals prior to disturbance (PD) and three, six and twelve months after treatments were applied in a campo sujo area of the Serra do Tombador Nature Reserve

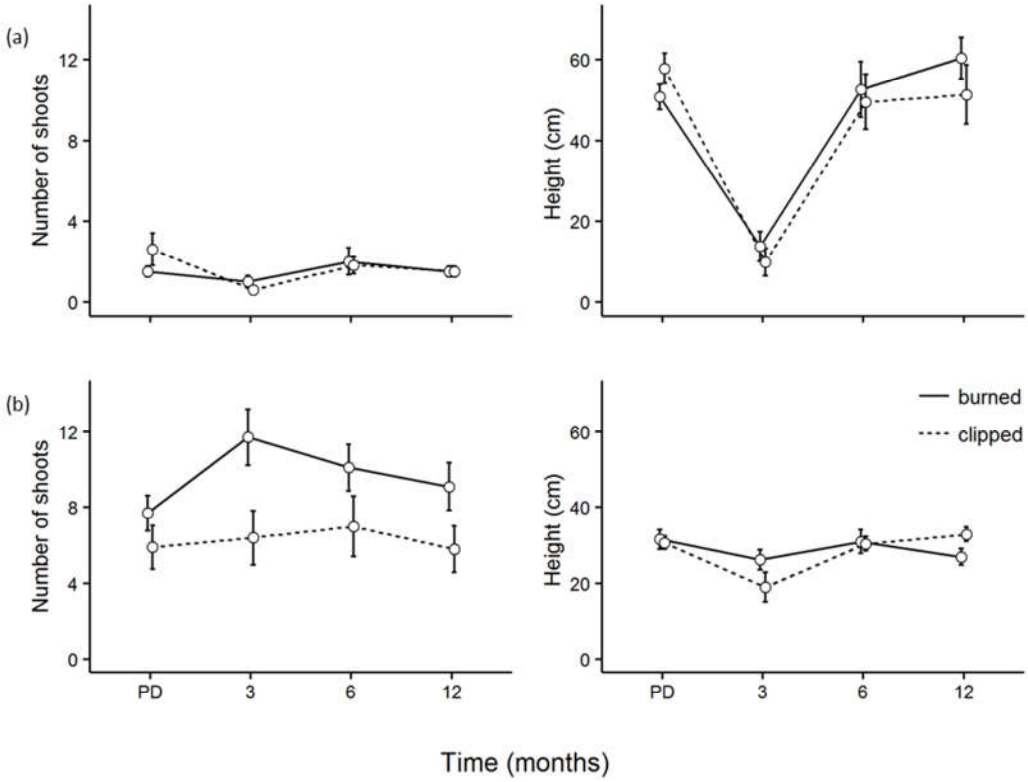

Figure 2. Number of shoots and height (mean ± SE) for a) *Mimosa gracilis and* b) *Ipomoea fiebrigii*, in burned (solid line) and clipped (dashed line) individuals prior to disturbance (PD) and three, six and twelve months after treatments were applied in a campo sujo area of the Serra do Tombador Nature Reserve

Table 2 Parameters estimated by the mixed models for diameter and height (LMMs) of grass species in relation to treatments (burned and clipped), time (3, 6 and 12 months after disturbance) and pre-disturbance size (total number of shoots prior to disturbance). Significant coefficients (p≤0.05) are given in bold font.

Species	Effects	**Diameter**			**Height**		
		Coefficients	Standard error	*P* values	Coefficients	Standard error	*P* values
Oncorachis ramosa	intercept	0.75	0.24	0.004	1.77	0.47	<0.0001
	treatment clipped	-0.01	0.08	0.98	**-0.35**	0.11	0.008
	time	**0.05**	0.01	<0.0001	**0.06**	0.01	<0.0001
	pre-disturbance size	**0.59**	0.09	<0.0001	**0.43**	0.14	0.008
Mesosetum loliiforme	intercept	1.99	0.31	<0.0001	-1.13	1.01	0.263
	treatment clipped	-0.12	0.07	0.138	-0.03	0.11	0.773
	time	**0.02**	0.00	0.008	**0.14**	0.01	<0.0001
	pre-disturbance size	0.12	0.14	0.381	**0.90**	0.30	0.008
Mesosetum ferrugineum	intercept	2.16	0.51	<0.0001	1.41	0.77	<0.0001
	treatment clipped	-0.16	0.09	0.091	-0.08	0.08	0.316
	time	0.00	0.00	0.454	**0.05**	0.01	<0.0001
	pre-disturbance size	0.12	0.19	0.533	0.44	0.22	0.06

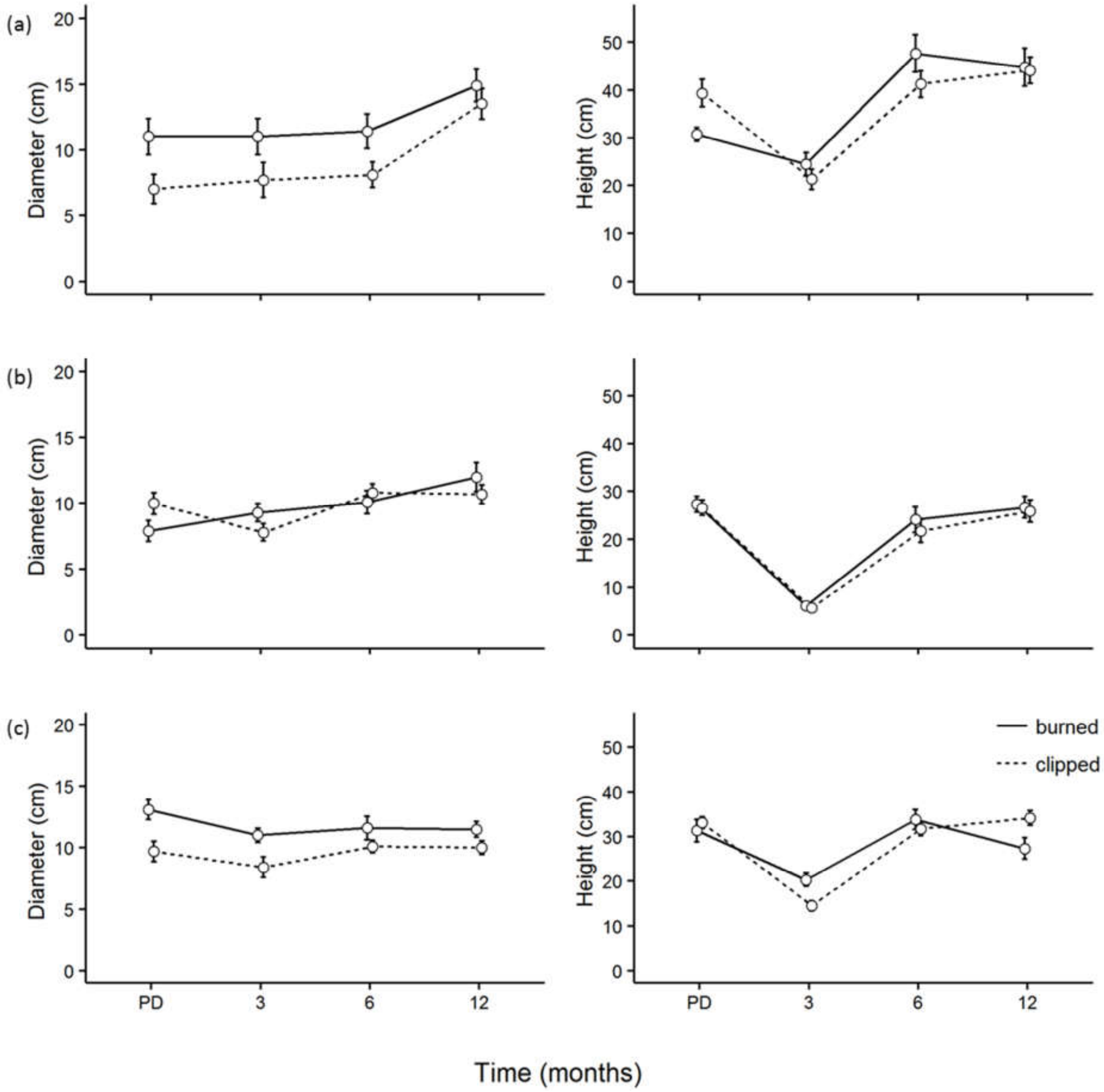

Figure 3. Diameter and height (mean ± SE) for a) *Oncorachis ramosa*, b) *Mesosetum lolliiforme* and c) *Mesosetum ferrugineum* in burned (solid line) and clipped (dashed line) individuals prior to disturbance (PD) and three, six and twelve months after treatments were applied in a *campo sujo* area of the Serra do Tombador Nature Reserve

Table 3 Parameters estimated by the GLMs for flowering responses of all species to burning and clipping treatments. Coefficients correspond to the logodds of flowering and its corresponding probability is also shown. Significant coefficients (p≤0.05) are given in bold font.

Species	Effects	Coefficients	Standard error	Probability	*P* values
Mimosa leiocephala	intercept	2.19	1.05	0.9	0.037
	treatment clipped	-1.32	1.49	0.7	1.0
Bauhinia dumosa	intercept	0.405	0.64	0.6	0.53
	treatment clipped	-1.253	0.94	0.3	0.185
Harpalyce brasiliana	intercept	0.847	0.69	0.7	0.21
	treatment clipped	**-3.04**	1.25	0.1	0.015
Chamaecrista claussenii	intercept	0.847	0.69	0.7	0.21
	treatment clipped	**-2.23**	1.04	0.2	0.03
Mimosa gracilis	intercept	0.847	0.69	0.7	0.21
	treatment clipped	**-2.23**	1.04	0.2	0.03
Ipomoea fiebrigii	intercept	1.386	0.79	0.8	0.07
	treatment clipped	-1.791	1.02	0.4	0.07
Oncorachis ramosa	intercept	0.405	0.64	0.6	0.53
	treatment clipped	-1.25	0.94	0.3	0.185
Mesosetum ferrugineum	intercept	1.25	0.80	0.77	0.11
	treatment clipped	-1.25	1.02	0.5	0.22

Discussion

Overall, burned shrubs had a greater resprouting response than clipped ones, while forb and grass species resprouted similarly after the different treatments, which might indicate that aboveground biomass removal was the main driver of resprouting responses in these species. Although both disturbance types lead to loss of aboveground biomass, fire may cause additional impacts because of the high temperatures reached during a fire episode, which can be lethal to meristematic tissue, such as buds and cambium (Clarke et al. 2013; Michaletz et al. 2012). In our study, individuals of all species could resprout after both fire and clipping, indicating that these fires did not damage buds or bud-forming tissues.

Resprouting ability is determined by the maintenance of a viable bud bank influenced by bud location and protection (Clarke et al. 2013). All species (except *M. ferrugineum*) have buds located belowground that are usually well protected by soil, insulating the heat generated from fire (Choczynska and Johnson 2009; Gagnon et al. 2010, 2015; Clarke et al. 2013). Soil heating is mainly determined by duration of combustion, or how long fuels burn, where faster fires tend to burn with shorter residence times (Neary et al. 1999; Choczynska and Johnson 2009; Gagnon et al. 2010, 2015). Cerrado surface fires have fast rates of spread and low residence times (Miranda et al. 2009; Pivello et al. 2010), and although temperatures at soil surface can reach up to 700ºC in Cerrado open savannas (Miranda et al. 1993; Pivello et al. 2010), temperatures 1cm belowground usually increase up to around 50ºC (Miranda et al. 1993) and only occasionally reach over 80ºC (Zupo et al. unpubl. data). Thus, the shorter fire residence time in grassy systems may explain the high resilience of species to such fire regimes (Scott et al. 2010). However, in more sclerophyllous systems, higher temperatures and prolonged soil heating has been measured (Vesk et al. 2004; Wright and Clarke 2007; Santana et al. 2013), which could potentially damage buds and bud-forming tissues and affect resprouting responses.

Nonetheless, previous studies comparing clipping and burning have shown positive and negative effects on resprouting. In other surface fire systems, Hmieloski et al. (2014) reported a smaller number of shoots after fire in relation to clipping for *Quercus nigra* in pine-grassland ecosystems, but Peterson and Drewa (2014) observed that fire and clipping showed similar effects on plant responses in oak barrens, savanna-type vegetation dominated by oaks, of Central North America. Likewise, Fidelis et al. (2012) also found similar recovery responses of shrub species between burned and mowed plots in subtropical grasslands. In Mediterranean-type vegetation that undergo crown fires, which are typically hotter and more intense, Calvo et al. (2005) and Fernandez et al. (2013, 2015) observed no differences in shrub recovery after fire and clipping, however, shrub recovery was greater after fire than after mastication in a Californian chaparral (Potts et al. 2010).

Another major difference between fire and other disturbances is that fire also depletes the litter layer and changes nutrient dynamics: there is a flush of new resources after fire that results from biomass combustion and ash deposition (Kauffman et al. 1994; Whelan 1995; Neary et al. 1999). The early resprouting response of the Mediterranean-type shrub *Erica australis* was highly variable among sites and largely driven by water and nutrient availability, both from storage and from the soil (Cruz et al. 2002, 2003). Thus, the post-fire environmental conditions, with increased resource availability, may explain both the greater number of shoots in shrub species (except for *M. leiocephala*) and the higher probability of flowering in two shrub and one forb species after fire in relation to clipping. Additionally, fire creates gaps where competition for resources are reduced (Whelan 1995), which could also explain the greater number of shoots found for three shrub species, since we did not remove the vegetation around clipped plants. Moreover, the distribution of nutrients in a post-fire environment may be influenced by the vegetation structure on a fine-scale, with a higher accumulation of ash and organic matter around large plants (Rodríguez et al. 2009), which might account for why only shrub species showed higher resprouting responses after fire.

The removal of aboveground biomass and depletion of the litter layer caused by fire also increases light availability at the soil surface and would be yet another resource-related change experienced by the burned individuals, since post-fire environments have a higher solar radiation than unburned ones (Fidelis and Blanco 2014), and could stimulate both flowering and recovery responses in plants, especially grasses (Brewer et al. 2009; Fidelis et al. 2012; Araújo et al. 2013). Due to the increase in light and nutrient availability, grasses, for example, may use the available solar radiation more efficiently, exhibiting increased net photosynthesis, leaf production and N uptake in post-fire environments (Knapp 1985), and are thus considered the most resilient group in grasslands and savannas (Bond 2004; Scott et al. 2010). However, we have not found a higher resprouting response among burned plants of grass species, reinforcing that the removal of aboveground biomass was sufficient to stimulate resprouting and that fire heating did not damage the buds. The same result was found for both forb species, although fire stimulated more individuals of *Mimosa gracilis* to flower. Other than resource related factors, direct effect of heat and smoke have also been reported as fire-related flowering cues (Light et al. 2007; Lamont and Downes

2011). In subtropical grasslands, burned plots showed a higher number of grass species flowering than mowed ones, indicating that fire might trigger flowering in this group (Fidelis and Blanco 2014). Flowering response of grass species usually occur within one month after fire (A. Fidelis et al. unpubl. data) and thus, we might not have been able to properly account for flowering among grass species in our experiment (observations every three months).

Our results can help elucidate the mechanisms of fire effects in Cerrado open savannas and suggest that these fires cause no damage both to buds located in the belowground bud bank and to buds on the soil surface protected by leaf bases. Additionally, the post-fire environment, associated with reduced competition and enhanced resource availability, seems to be an important factor affecting both resprouting and flowering responses. Thus, for some species, fire, and not only aboveground biomass removal, determines resprouting success. Moreover, this study contributes to the long-needed knowledge on resprouting behavior outside the Mediterranean ecosystem (Pausas et al. 2016), bringing valuable results for the Neotropical savannas.

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Capítulo 3

Effects of fire regime on resprouting and flowering of Cerrado species

Talita Zupo & Alessandra Fidelis

Abstract

Resprouting responses of species can be strongly affected by fire regimes, such as fire season and frequency. We conducted experimental fires to test the effects of fire season and frequency on resprouting and flowering of nine Cerrado open savanna species in Central Brazil. We predicted that late-dry season fires and annual fires would result in greater negative impacts on resprouting and flowering responses. We conducted experimental fires in three different seasons (early, mid and late-dry season; burned in 2013) and frequencies (biennial, annual and unburned; burned from 2013-2015). Individuals of four shrubs, two forbs and three grass species were permanently tagged and measured before and after fires. For shrubs and forbs we measured number of shoots and height and for grasses we measured diameter and height. Early-dry season fires had a negative impact on resprouting height in relation to mid and late-dry season fires, most likely limited by water availability as plants resprouted during the dry season. Resprouting responses were not significantly different between annually and biennially burned plants and plants resprouted vigorously after three consecutive burns. Moreover, fire, and not a specific fire season, seems to be more important for reproduction as flowering was not affected by fire season for most species and fire induced flowering compared to unburned plants. These results suggest that these plants display a rather conservative strategy in terms of utilizing stored reserves, both by rapidly recovering their photosynthetic tissue after disturbance and by suppressing flowering during periods without fires.

Keywords: fire season, fire frequency, post-fire regeneration, resprouting vigour, Neotropical savanna

Introduction

Many plant species resprout after disturbances that kill their aboveground tissues, thus resprouting can be considered a key regenerative trait that allows species to persist in fire-prone ecosystems (Bond and Midgley 2001, 2003; Vesk and Westoby 2004; Clarke et al. 2013). Resprouting is an ancient trait among angiosperms (Bond and Midgley 2003; Keeley et al. 2012), however, resprouting responses are not only genetically determined, but strongly affected by both the environmental conditions (site productivity and resource availability) and the fire regime at a given site, such as fire frequency, intensity, and season (Bond and Midgley 2003; Bond and Keeley 2005; Clarke et al. 2005; Pausas and Ribeiro 2013).

The ability of plants to resprout after fires can be limited by both the availability of surviving meristems (bud or bud-forming tissues) and amount of stored reserves that support regrowth (Clarke et al. 2013). Thus, location and protection of buds are key to successful resprouting in fire-prone ecosystems, where species often protect their buds with thick corky barks (Dantas and Pausas 2013) or leaf sheaths (Lamont et al. 2004), have buds buried deep in the wood (Burrows et al. 2010; Charles-Dominique et al. 2015) or have belowground bud banks located in specialized structures (e.g. xylopodia and lignotubers) insulated by soil (Simon et al. 2009; Simon and Pennington 2012; Clarke et al. 2013; Fidelis et al. 2014). In grasslands and savannas, which are subjected to surface fires, fire frequency may affect bud survival as fuel accumulation from longer fire intervals may lead to more intense and hotter fires (Govender et al. 2006; Pivello et al. 2010; Fidelis et al. 2010; Zupo et al. unpubl. data). However, if disturbance frequency is too high, resprouting responses might be reduced, as storage reserves may not have been completely replenished (Miyanishi and Kellman 1986; Enright et al. 2011; Paula and Ojeda 2006, 2009). Repeated disturbances can also affect reproductive output of species as stored resources that would be mobilized for reproduction would otherwise be allocated to sustain regrowth of vegetative structures (Knox and Morrison 2005, Brewer et al. 2009).

Timing of fire may affect vegetation recovery through several mechanisms, especially in ecosystems with seasonal climate (Knapp et al. 2009; Hmielowski et al. 2014). One model that may explain the effects of fire season on plant responses is related to differences in fire parameters across seasons (Williams et al. 2004; Wright and Clarke 2007). Plant and soil moisture content vary throughout the year, which in turn will affect plant flammability, rates of fire spread, fire intensity and severity (Govender et al. 2006; Simpson et al. 2016, Rissi et al. 2017). It is generally believed that fires burning early in the dry season tend to be less severe as fuel moisture contents are still higher than in mid and late-dry season fires, period in which there is a greater amount of dry biomass (Whelan 1995; Govender et al. 2006; Knapp et al. 2009; Rissi et al. 2017).

However, if fire parameters between seasons are similar, then differences related to plant physiology and phenology are more likely to be noticed (Knapp et al. 2009), as carbohydrate reserves available for plant growth may vary according to season (Wurth et al. 2005; Schutz et al. 2009; Cruz and Moreno 2001). Storage reserves are relatively lower during the growing season (wet season in tropical climates), increasing to higher amounts during the dry season due to the variation in allocation between growth and storage (Wurth et al. 2005; Schutz et al. 2009). Differences in resprouting responses have often been interpreted as caused by differences in stored carbohydrate reserves at the time of the disturbance (Bowen and Pate 1993; Hmielowski et al. 2014), but higher water and nutrient availability in the soil have also been proposed to account for such variation (Wildy and Pate 2002, Cruz et al. 2002, 2003a).

Fire has been present in Cerrado for millions of years (Simon et al. 2009) and fire regimes are characterized by frequent surface fires of relatively low intensity fueled by grasses (Miranda et al. 2002). The grassy fuel loads become dry and highly flammable during the dry season and natural fires caused by lighting are common, mainly in the wet season but also during the early (May) and, especially, late-dry seasons (September and October; Ramos-Neto and Pivello 2000; Medeiros and Fiedler 2004; Alvarado et al. 2017). Currently, however, a great number of wildfires in Cerrado are caused by human ignition, mostly during the mid-dry season (July and August, Ramos-Neto and Pivello 2000; Medeiros and Fiedler 2004; Pivello 2011). After fires, most Cerrado species resprout (Zupo et al. unpubl. data) and aboveground biomass is quickly recovered (1-2 years, Cianciaruso et al. 2010; Rissi 2016), enabling frequent fires. Thus, as in other mesic savannas, fire frequency in Cerrado is high, where the typical fire return interval is between 3-4 years, but some areas, especially open savannas and grasslands, are subjected to higher frequencies (intervals of 1 or 2 years, Daldegan et al. 2014; Pereira-Júnior et al. 2014).

In this context, we investigated the effects of different fire regimes on regeneration of species from a Cerrado open savanna. More specifically we asked how fire season (early, mid and late-dry season fires) and frequency (biennially, annually and unburned) would affect resprouting and flowering of Cerrado species. We predict that if late-dry season fires are more severe, then early-dry season fires are expected to have a higher resprouting and flowering response. However, if fire parameters among seasons are similar, then differences in resprouting responses are expected to be limited by resource availability, both from stored reserves and from the soil. We also predict that if resprouting responses are limited by time allowed for carbohydrate storage to be replenished, then annual burns will have reduced resprouting and flowering responses in relation to biennial burns.

Material and methods

Study Area and study species

The study was conducted at the Serra do Tombador Nature Reserve (RNST), located in Central Brazil (47°45'; 47°51' W and 13°35'; 13°38' S). The regional climate is tropical with wet summers and dry winters with an average annual rainfall between 1300 and 1500 mm, most of which falls during the wet season (Fundação Grupo Boticário 2011). The RNST has an area of approximately 8900 ha, where different Cerrado physiognomies are present, but the *campo sujo*, an open savanna type with a continuous herbaceous layer and scattered shrubs prevails. Data for 2001-

2010 indicated that fires in the RNST were most frequent in grassland and savanna sites and typically occurred in a 3-yr interval. During that period, 69% of the RNST had suffered at least one fire episode, 33% was burned at least twice and around 10% was burned four times (Daldegan et al. 2014).

Nine dominant species belonging to different functional groups were selected for this study: four shrub species, *Chamaecrista claussenii* (Benth) H.S. Irwin & Barneby, *Harpalyce brasiliana* Benth., *Mimosa leiocephala* Benth. and *Bauhinia dumosa* Benth. (all Fabaceae); two forb species, *Mimosa gracilis* Benth. (Fabaceae) and *Ipomoea fiebrigii* Hassl. ex O'Donell (Convolvulaceae); and three grass species (Poaceae), *Mesosetum ferrugineum* (Trin.) Chase, *Mesosetum loliiforme* (Hochst. ex Steud.) Chase and *Oncorachis ramosa* (Zuloaga & Soderstr.) Morrone & Zuloaga. All shrub and forb species have buds on underground structures insulated by soil, two grass species (*M. loliiforme* and *O. ramosa*) are rhizomatous with belowground buds and one (*M. ferrugineum*) is caespitose with buds on the soil surface protected by leaf sheaths.

Experimental design

In 2013, we established 30 x 30m plots in an open savanna area that had last been burned in August 2011. Plots were established with a distance of at least 4 m from each other, in a randomized block design, and treatments within each block were randomly assigned to each plot (4 plots/treatment). To prevent fire from spreading, 3 m wide firebreaks were established around each plot. In every experimental fire carried out, head fires were conducted separately in each plot early in the morning or late in the afternoon with the assistance of the reserve's fire brigade. After fires, all aboveground biomass was dead, either by being totally consumed (grasses and forbs) or by having tissue death due to high temperatures (shrubs, topkill, T. Zupo pers. obs).

Fire season effects on resprouting and flowering

In 2013, four 30 x 30 m plots were assigned to each of the following treatments: early-dry (burned at the beginning of the dry season in May 2013, EF), mid-dry (burned during mid-dry season in July 2013, MF) and late-dry (burned at the end of the dry season in October 2013, LF; Fig. 1). Experimental fires in all plots were conducted as described above. Mean fire intensity ranged from 503±119 $kW.m^{-1}$ in early-dry (EF) to 3009±1408 $kW.m^{-1}$ in late-dry season fires (LF); and rate of fire spread from 0.05±0.01 $m.s^{-1}$ in early-dry (EF) to 0.35±0.15 $m.s^{-1}$ in late-dry season fires (Rissi et al. 2017, Table 1).

In each plot five individuals of each species were randomly selected, permanently tagged and measured prior to fires (May, July and October 2013, respectively) and three, six and twelve months after fires. A total of 20 individuals of each species were tagged in each treatment (EF, MF, LF). Marked individuals of the same species were at least 1 m apart from each other. In all surveys we measured maximum height, total number of shoots and number of reproductive shoots for shrubs and forbs, and height and diameter of tussocks for grass species, as well as number of reproductive tillers.

Fire frequency effects on resprouting and flowering

Also in July 2013, four 30 x 30 m plots were assigned to each of the following treatments: biennial (burned during mid-dry season in 2013 and 2015, B), annual (burned during mid-dry season in 2013, 2014 and 2015, A) and unburned (kept protected from fire from 2011-2016, UB; Fig. 1). Plots belonging to the MF treatment described above were later burned in July 2015, becoming the biennial treatment (B). Additionally, in May 2014, an accidental fire burned one of the unburned (UB) plots, henceforth having only three replicates. Experimental fires in all plots were conducted as described above. Fire intensity in biennial plots ranged from 2707±1899 in 2013 to 2782±1357 $kW.m^{-1}$ in 2015, while in annual plots fire intensity ranged from 808±112 in 2013 to 1091±205 $kW.m^{-1}$ in 2015 (Table 1).

In each treatment (A, B, UB), 20 individuals of each species were randomly selected, permanently tagged and measured as described above (5 individuals/plot). Measurements were performed prior to fires (July 2013, dry season), and every six months after the fires until July 2016.

Thus we carried out surveys in the wet season of 2014, 2015 and 2016 and in the dry season of 2014, 2015 and 2016. We were unable to properly survey the unburned plots (UB) in the wet season of 2016.

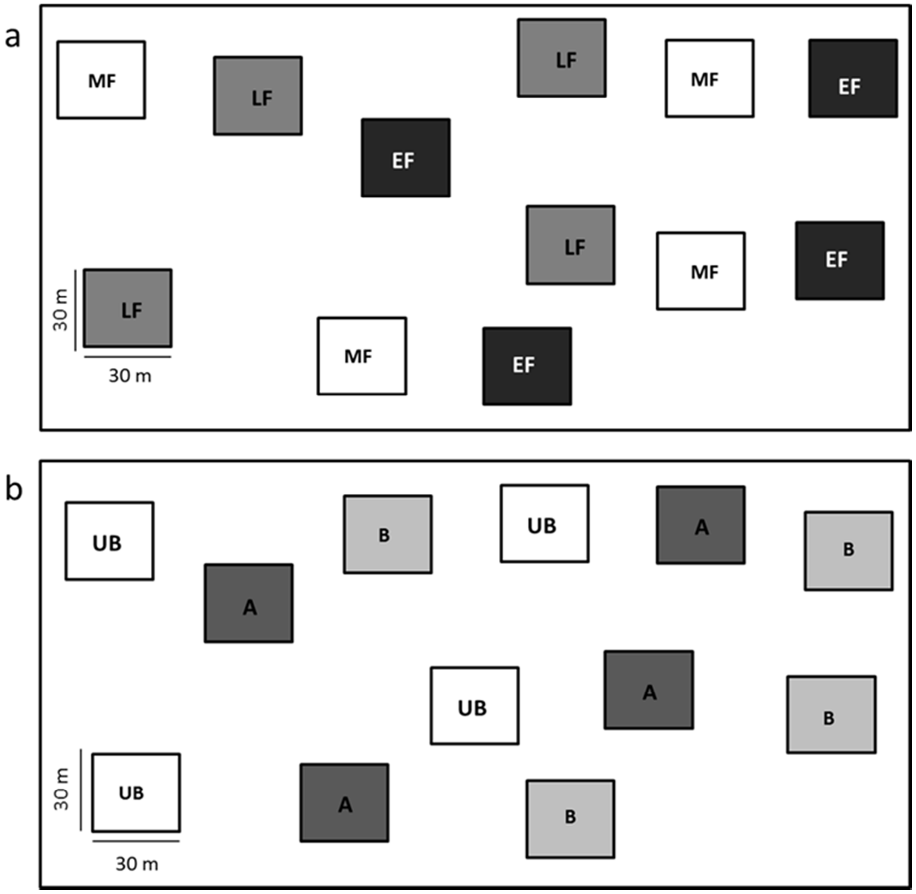

Figure 1. Experimental design of the prescribed burning experiments carried out in an open savanna in Central Brazil: a) effects of fire season, with plots that were burned in early (EF), mid (MF) and late-dry season (LF) in 2013 and b) effects of fire frequency, with plots burned biennially (B, 2013 and 2015) and annually (A, 2013, 2014 and 2015) during mid-dry season (July), and unburned plots (UB, kept protected from fire from 2013 - 2016).

Table 1. Fire intensity and rate of spread (mean ± SD) for a) fire season: early, mid and late-dry season fires and b) fire frequency: biennial and annual fires burned in 2013, 2014 and 2015.

		Fire intensity (kW.m^{-1})	Rate of spread (m.s^{-1})
a) Fire season[a]	Early	503 ± 119	0.05 ± 0.01
	Mid	2707 ± 1899	0.23 ± 0.13
	Late	3009 ± 1408	0.35 ± 0.15
b) Fire frequency[b]			
2013	Biennial	2707 ± 1899	0.23 ± 0.13
	Annual	808 ± 112	0.18 ± 0.02
2014	Biennial	-	-
	Annual	941 ± 548	0.15 ± 0.05
2015	Biennial	2782 ± 1357	0.2 ± 0.06
	Annual	1091 ± 205	0.16 ± 0.04

[a] Data from Rissi et al. (2017)

[b] Data from Zupo et al. (unpubl. data)

Statistical analyses

We used mixed models to analyze post-fire resprouting response. To estimate differences in total number of shoots we used Generalized Linear Mixed Models (GLMMs, Zuur et al. 2009) with a Poisson distribution. To estimate differences in diameter and height, we used linear mixed models (LMMs, Zuur et al. 2009) with a Gaussian distribution after log transformation to fit model assumptions. We fitted GLMMs or LMMs for each species separately with treatment and time as fixed effects, and pre-disturbance size (total number of shoots – or diameter - and height prior to fires) as a covariate. Thus, for fire season we used early (EF), mid (MF) and late-dry season fires (LF) as treatments, where EF was taken as the baseline for the intercept in the models, and time (3, 6 and 12 months after fires) as a continuous variable. For fire frequency we used unburned (UB), Biennial (B) and Annual (A) fires as treatments, where UB was taken as the baseline for the intercept in the models, and time (6, 12, 18, 24, 30 and 36 months after fires) as a continuous variable. In both cases, we fitted a random intercept model with individuals and plots as random factors, since the same individual was measured repeatedly over time and individuals were nested within plots. *Post-hoc* Tukey's tests were performed to evaluate multiple comparisons among treatments ($\alpha = 0.05$).

We used GLMMs with a binomial distribution to estimate probability of flowering under each treatment, given that presence of flowering was a binary data. We fitted a random intercept model with treatment as fixed factors and plots as random factors, since individuals were nested within plots. Models were fitted for each species separately using data from the flowering period of the subsequent year for each species. Unburnt plants were used to determine the flowering period for each species, except for grass species since we did not observe reproductive tillers in unburnt plants in any of the census carried out.

Hence, to evaluate how fire season affected probability of flowering of *Harpalyce brasiliana* and *Mimosa gracilis* we used data from January/February 2014 for individuals of each treatment (early, mid and late); for *Mimosa leiocephala* we used data from May 2014; and for *Bauhinia dumosa*, *Chamaecrista claussenii* and *Ipomoea fiebrigii* we used the data from July 2014. Although we were able to account for reproductive tillers after early and mid-dry season fires for two grass species (*Mesosetum ferrugineum and Oncorachis ramosa*), we were unable to do so after late-dry season fires since they had already dispersed their seeds in the subsequent survey carried out. Moreover, we did not observe reproductive tillers in *Mesosetum loliiforme* in any of the surveys carried out. Thus, we were unable to estimate probability of flowering between fire seasons for any of the grass species.

To evaluate how fire frequency affected probability of flowering, we performed GLMMs for each year separately: for the grass species, *M. ferrugineum* and *O. ramosa*, we used data from October 2013, 2014 and 2015; for *H. brasiliana* and *M. gracilis* we used data from February 2014 and 2015; for *M. leiocephala* we used data from May 2014, 2015 and 2016; and for *B. dumosa*, *C. claussenii*, and *I. fiebrigii* we used the data from July 2014-2016. We were unable to estimate probability of flowering for *H. brasiliana* and *M. gracilis* for the year of 2016 as we were unable to properly survey the unburned (UB) plots in February 2016 as described above. We were also unable to estimate probability of flowering for the grass species *M. lolliforme* for the reasons described above.

GLMMs were fitted by maximum likelihood assuming a Laplace approximation to the likelihood function and LMMs were fitted by maximum likelihood. All the analyses were carried out with the ''lme4'' (Bates et al. 2015) and multcomp (Hothorn et al. 2008) package in R software (R Core Development Team 2016).

Results

Fire season

All species could resprout after fires, in all seasons. Fire season did not affect the number of shoots for two shrubs species, *B. dumosa* and *H. brasiliana* (Table 2, Fig. 2a-b). However, for *M. leiocephala*, total number of shoots regenerating after EF (e.g. 10.8±1.8 shoots three months after early fires) was greater in relation to both MF (6.3±0.7 shoots, $p<0.0001$) and LF (5.9±0.8 shoots, $p=0.01$; Fig. 2c). The opposite pattern was found for *C. claussenii*, where the total number of shoots after EF (1.3±0.2 shoots three months after early fires) was smaller in relation to both MF (3.2±0.4 shoots, $p=0.001$) and LF (3.9±0.4 shoots, $p<0.0001$; Fig. 2d). Additionally, season of burn did not affect total number of shoots for both forb species, *M. gracilis* and *I. fiebrigii* (Table 2, Fig. 3a-b). Over the course of one year after fires, total number of shoots decreased through time for all shrub species, except for *C. claussenii* (Table 2, Fig. 2d) and for one forb species*, I. fiebrigii* (Table 2, Fig. 3b), in which time did not affect the total number of shoots.

Maximum height of all shrub and forb species was affected by time, with an increase in height throughout the first twelve months after fires (Table 2, Fig. 2 and 3). Maximum height of shrub species was smaller after EF (15.6±2.1, 16.2±2.1 and 17.2±2.6 cm three months after fire for *H. brasiliana, M. leiocephala* and *C. claussenii*, respectively) in relation to both MF (44.2±2.9, 50.7±4.1, 35.8±2.3) and LF (54.9±2.7, 67.8±3.8, 37.6±1.8 cm; Table 2, Fig. 2 b-d), except for *B.*

dumosa, where maximum height was not affected by fire season (Table 2, Fig. 2a). However, twelve months after fires, individuals of all shrub species had similar maximum heights in all treatments (Fig. 2). Height of the forb species *M. gracilis* was greater after late-dry season fires in relation to both early and mid-dry season fires (Table 2, Fig. 3a), while *I. fiebrigii* showed the same pattern as the shrub species, with smaller height after early fires in relation to both mid and late-dry season fires (Table 2, Fig. 3b).

Fire season only affected diameter of *M. ferrugineum*, which showed a greater diameter after early season fires in relation to both mid and late-dry season fires (Table 3, Fig. 4a). Diameter of *O. ramosa* and *M. loliiforme* increased over time in all treatments (Table 3, Fig. 4b-c). In general, height of all grass species was greater after late-dry season fires in relation to both early and mid-dry season fires (Table 3, Fig. 4). As expected, height increased over time for all grass species.

For most species, fire season had no effect on flowering (Table 4, Fig. 5). However, *H. brasiliana* had significantly greater chances of flowering after both mid and late-dry season fires, (72 and 95%, respectively) in relation to EF (20%, p=0.02 and p<0.001, respectively; Table 3, Fig. 5). *C. claussenii* had significantly greater chances of flowering after MF (79%) in relation to both early (45%, p=0.02) and LF (37%, p=0.02; Table 3, Fig. 5). Proportion of flowering individuals was >80% for *M. leiocephala* and around 45% for *B. dumosa*, irrespective of burning season (Fig. 5). Proportion of individuals of forb species flowering was also unaffected by fire season (Fig. 5).

Table 2. Parameters estimated by the mixed models for total number of shoots (GLMMs) and height (LMMs) of shrub and forb species in relation to treatments (early, mid and late dry season burns) and time (3, 6 and 12 months after disturbance). Significant coefficients ($p \leq 0.05$) are given in bold.

		Number of shoots			**Height**		
Species	Effects	Coefficients	Standard error	*P* values	Coefficients	Standard error	*P* values
Bauhinia dumosa	intercept	1.36	0.19	<0.0001	2.32	0.55	<0.0001
	Mid-dry season	0.36	0.23	0.11	0.18	0.11	0.12
	Late-dry season	0.16	0.23	0.48	0.09	0.11	0.387
	time	**-0.04**	0.01	<0.0001	**0.06**	0.01	<0.0001
Harpalyce brasiliana	intercept	1.29	0.18	<0.0001	0.14	0.8	0.85
	Mid-dry season	0.24	0.18	0.19	**0.75**	0.09	<0.0001
	Late-dry season	0.06	0.18	0.74	**0.77**	0.09	<0.0001
	time	**-0.06**	0.01	<0.0001	**0.06**	0.01	<0.0001
Mimosa leiocephala	intercept	1.66	0.12	<0.0001	-0.06	0.52	0.91
	Mid-dry season	**-0.49**	0.11	<0.0001	**0.73**	0.09	<0.0001
	Late-dry season	**-0.27**	0.11	0.01	**0.71**	0.08	<0.0001
	time	**-0.03**	0.01	<0.0001	**0.10**	0.01	<0.0001
Chamaecrista claussenii	intercept	0.37	0.16	0.02	1.36	0.9	0.13
	Mid-dry season	**0.45**	0.14	0.001	**0.46**	0.11	0.002
	Late-dry season	**0.70**	0.14	<0.0001	**0.36**	0.12	0.003
	time	0.01	0.01	0.4	**0.06**	0.01	<0.0001
Mimosa gracilis	intercept	0.17	0.19	0.393	0.58	1.19	0.62
	Mid-dry season	-0.25	0.14	0.076	0.10	0.17	0.54
	Late-dry season	0.16	0.15	0.284	**0.71**	0.16	<0.0001
	time	0.01	0.01	0.576	**0.09**	0.02	<0.0001
Ipomoea fiebrigii	intercept	1.77	0.19	<0.0001	1.37	0.52	0.01
	Mid-dry season	-0.01	0.20	0.98	**0.35**	0.10	0.001
	Late-dry season	0.4	0.21	0.05	**0.51**	0.10	<0.0001
	time	**-0.02**	0.01	<0.0001	**-0.02**	0.01	0.003

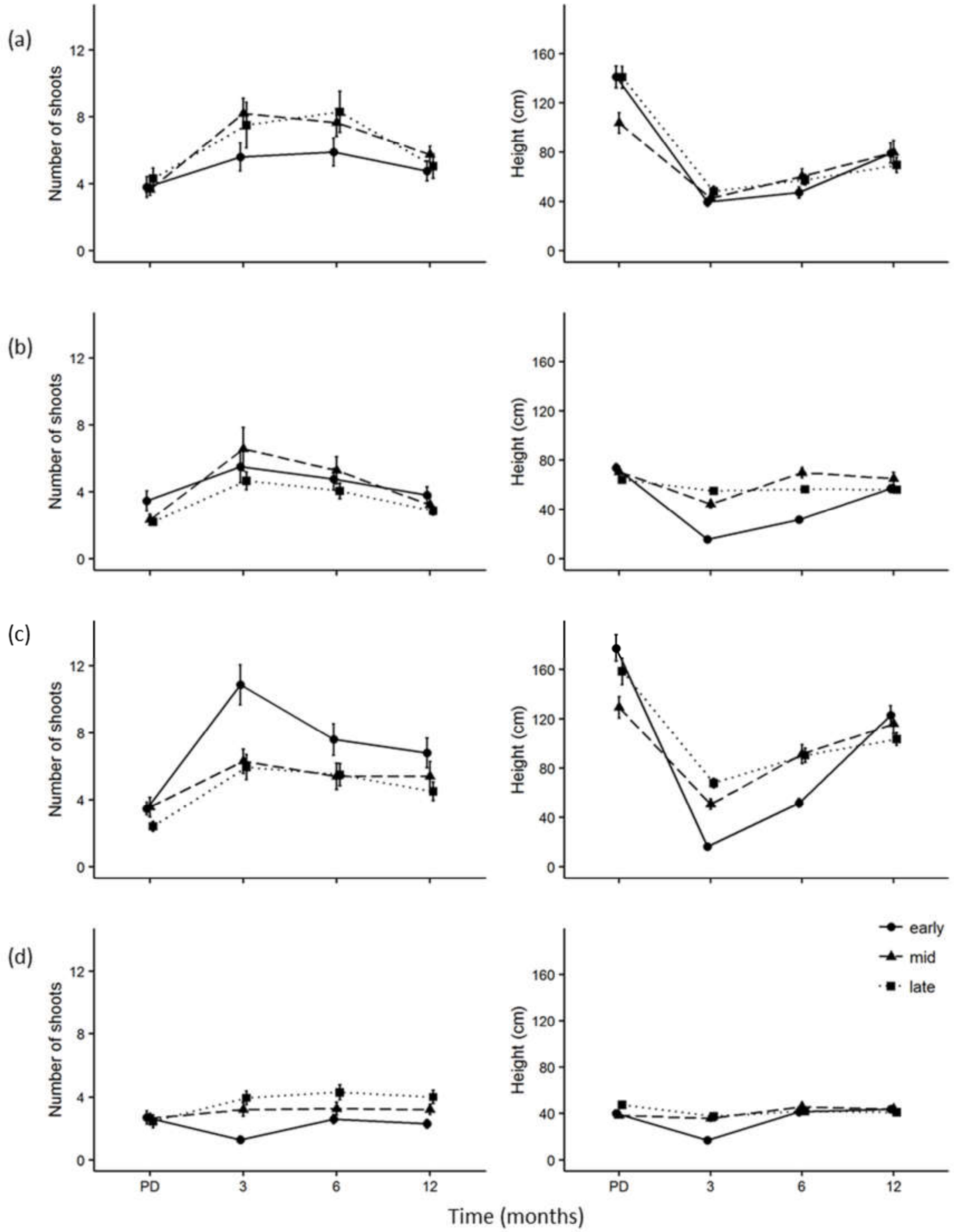

Figure 2. Number of shoots and height (mean±SE) for the shrub species a) *Bauhinia dumosa*, b) H*arpalyce brasiliana*, c) *Mimosa leiocephala* and d) *Chamaecrista claussenii* in early (solid line), mid (dashed line) and late-dry season burns (dotted line) prior to disturbance (PD) and three, six and twelve months after treatments were applied in an open savanna area of the Serra do Tombador Nature Reserve

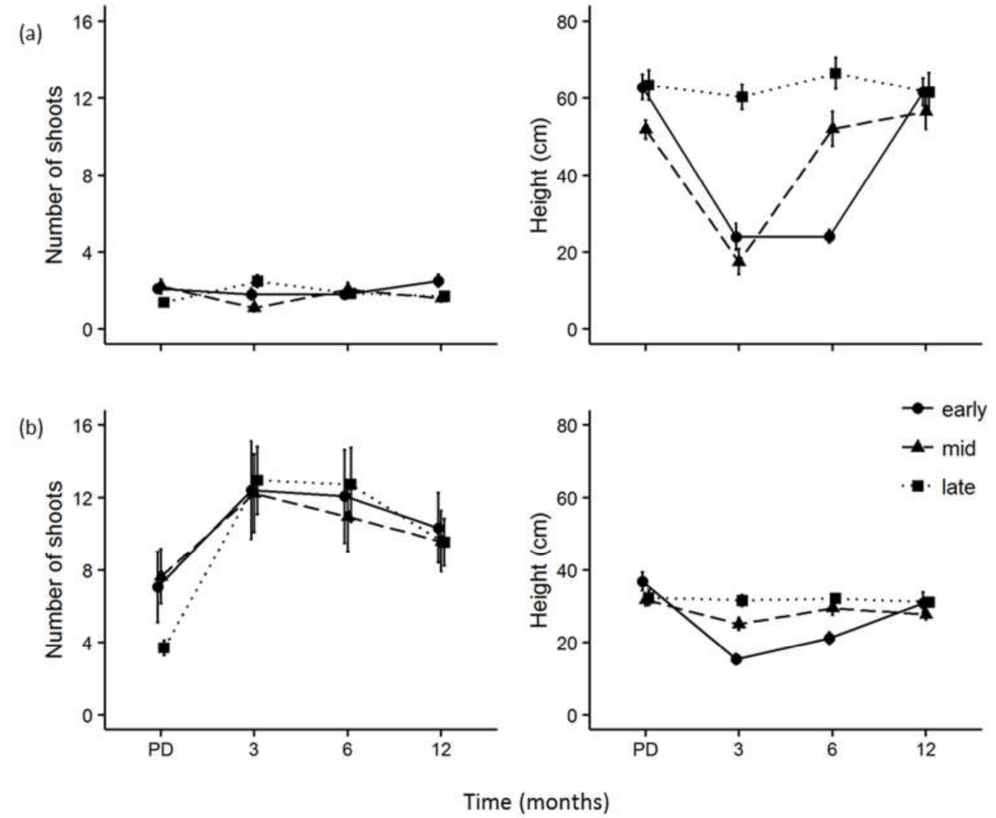

Figure 3. Number of shoots and height (mean ± SE) for forbs species a) *Mimosa gracilis* and b) *Ipomoea fiebrigii* in early (solid line), mid (dashed line) and late dry season burns (dotted line) prior to disturbance (PD) and three, six and twelve months after treatments were applied in an open savanna area of the Serra do Tombador Nature Reserve

Table 3. Parameters estimated by the mixed models for diameter and height (LMMs) of grass species in relation to treatments (early, mid and late dry season burns) and time (3, 6 and 12 months after disturbance. Significant coefficients (p≤0.05) are given in bold.

		Diameter			**Height**		
Species	Effects	Coefficients	Standard error	*P* values	Coefficients	Standard error	*P* values
Mesosetum ferrugineum	intercept	0.87	0.22	0.0002	2.54	0.42	<0.0001
	Mid-dry season	**-0.17**	0.05	0.0003	0.01	0.07	0.81
	Late-dry season	**-0.27**	0.05	<0.0001	**0.16**	0.07	0.03
	time	0.00	0.00	0.89	**0.01**	0.01	0.007
Mesosetum loliiforme	intercept	1.28	0.15	<0.0001	0.03	0.55	0.95
	Mid-dry season	-0.07	0.06	0.25	**0.17**	0.08	0.03
	Late-dry season	-0.08	0.06	0.19	**0.62**	0.08	<0.0001
	time	**0.01**	0.00	0.005	**0.09**	0.01	<0.0001
Oncorachis ramosa	intercept	1.69	0.18	<0.0001	1.61	0.45	0.006
	Mid-dry season	0.03	0.09	0.71	**0.12**	0.05	0.017
	Late-dry season	-0.10	0.09	0.28	**0.31**	0.05	<0.0001
	time	**0.06**	0.01	<0.0001	**0.04**	0.01	<0.0001

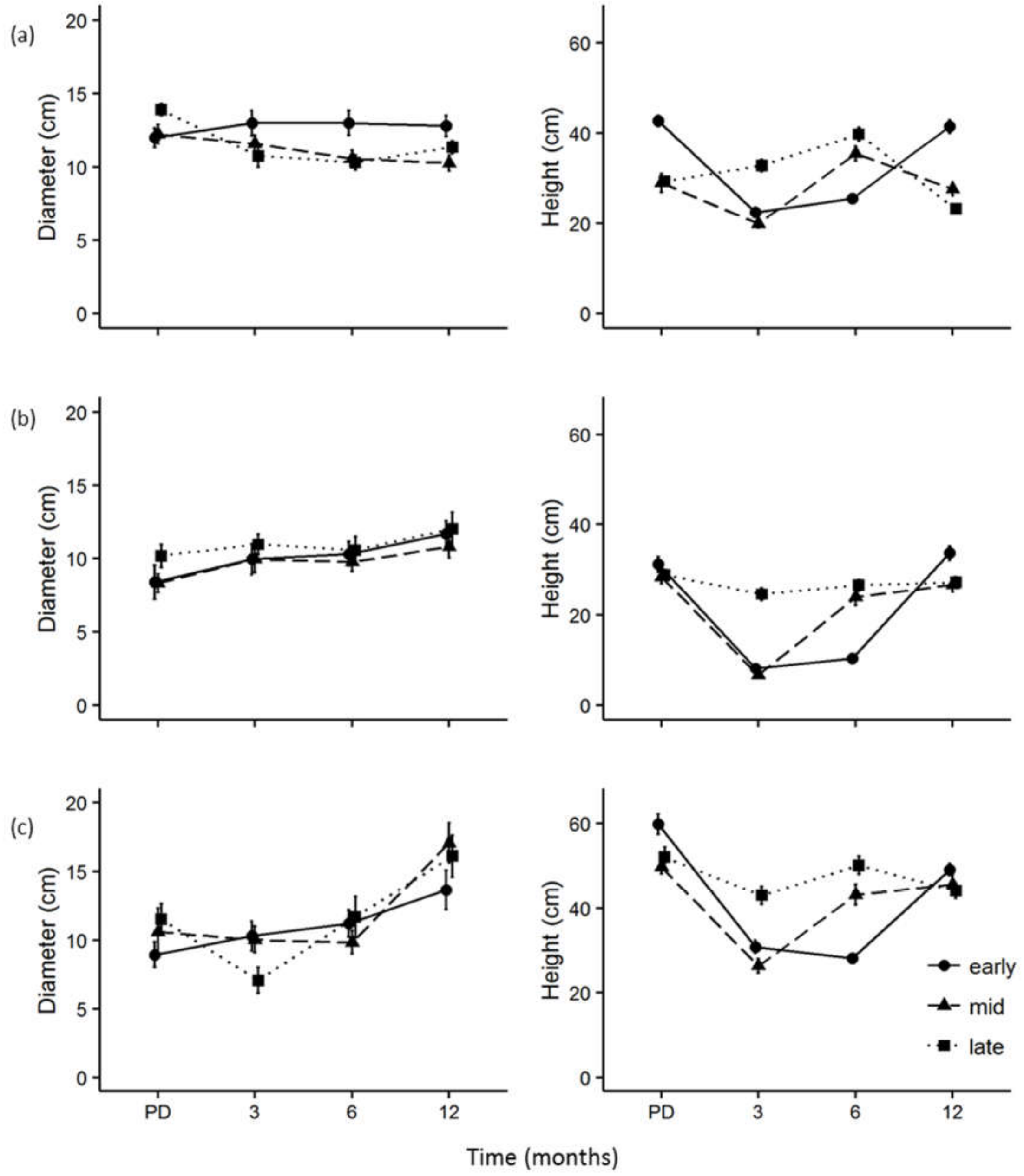

Figure 4. Diameter and height (mean ± SE) for the grass species a) *Mesosetum ferrugineum*, b) *Mesosetum loliiforme* and c) *Oncorachis ramosa* individuals in early (solid line), mid (dashed line) and late dry season fires (dotted line) prior to disturbance (PD) and three, six and twelve months after treatments were applied in an open savanna area of the Serra do Tombador Nature Reserve

Table 4. Parameters estimated by the GLMMs for flowering responses of shrub and forb species to different fire seasons. Coefficients correspond to the logodds of flowering; significant coefficients (p≤0.05) are given in bold.

Species	Effects	Coefficients	Standard error	*P* values
Bauhinia dumosa	intercept	0.0	0.4	1
	Mid-dry season	-0.02	0.6	0.75
	Late-dry season	0.0	0.6	1
Harpalyce brasiliana	intercept	-1.36	0.56	0.01
	Mid-dry season	**2.34**	0.77	0.002
	Late-dry season	**4.33**	1.17	<0.001
Mimosa leiocephala	intercept	2.34	1.38	0.09
	Mid-dry season	0.39	1.61	0.8
	Late-dry season	0.58	1.63	0.7
Chamaecrista claussenii	intercept	-0.2	0.44	0.6
	Mid-dry season	**1.58**	0.71	0.02
	Late-dry season	-0.33	0.65	0.6
Mimosa gracilis	intercept	0.0	0.4	1
	Mid-dry season	0.6	0.6	0.3
	Late-dry season	0.3	0.6	0.6
Ipomoea fiebrigii	intercept	1.55	0.82	0.06
	Mid-dry season	0.16	1.15	0.88
	Late-dry season	-0.26	1.08	0.81

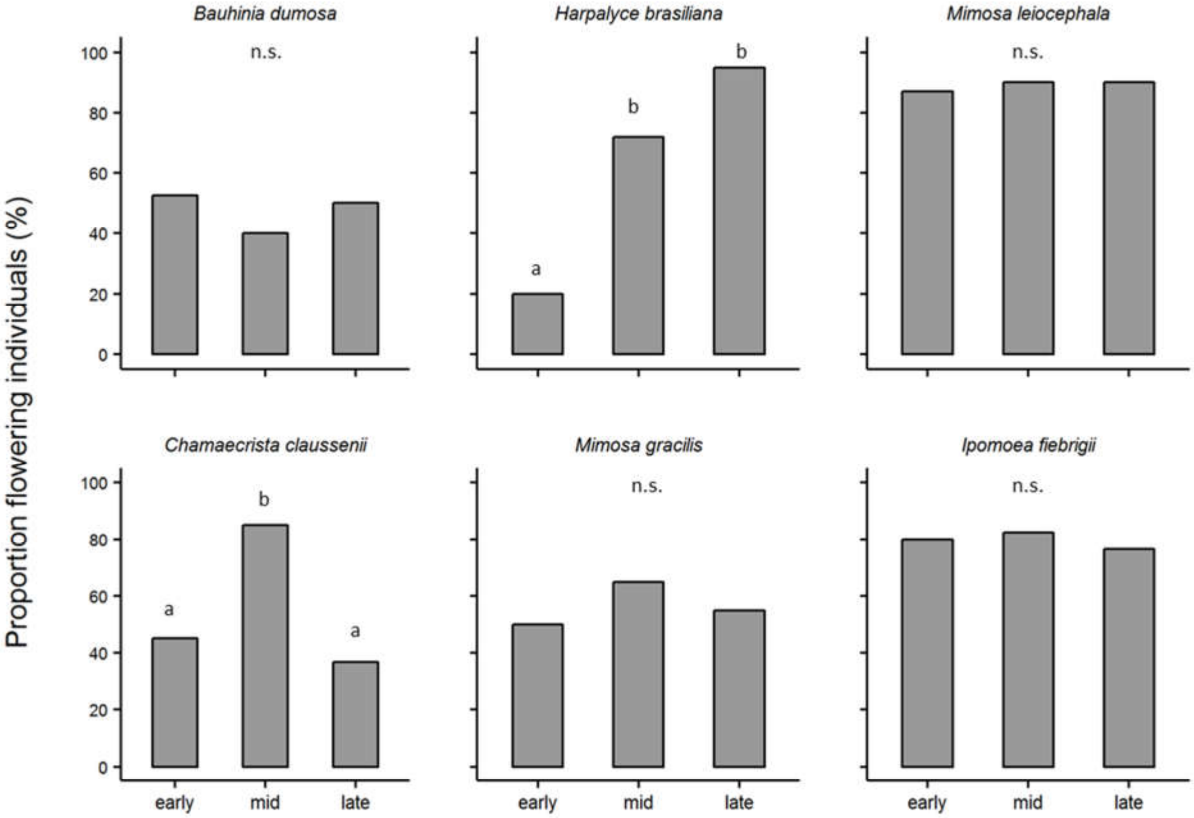

Figure 5. Proportion of flowering individuals for shrub and forb species in early, mid and late-dry season fires in the subsequent flowering period for each species of an open savanna area of the Serra do Tombador Nature Reserve. Different smallcase letters indicate significant differences ($p \leq 0.05$) among treatments

Fire frequency

All individuals of all species were able to resprout after both biennial and annual fires. Total number of shoots of shrub and forb species was greater after both biennial and annual fires in relation to unburned plants (Table 5, Fig. 6 and 7), but no differences were found between individuals that were burned biennially and those that were burned annually (in July 2016, *B. dumosa*: 5.1±0.6 and 6±0.9 shoots; *H. brasiliana*: 3.8±0.6 and 5.1±0.9 shoots; *M. leiocephala*: 5.6±0.8 and 5±0.9 shoots; *C. claussenii*: 3.4±0.3 and 3±0.3 shoots, after biennial and annual fires, respectively, all $p > 0.05$, Fig. 6 a-d). Among shrub species, time only affected total number of shoots of *B. dumosa,* which decreased over time (Table 5, Fig. 6a). Total number of shoots of the forb species *I. fiebrigii* was, in general, greater in individuals that were annually burned in relation to those that were biennially burned (p=0.02), since total number of shoots decrease considerably over time (Table 5, Fig. 7a). However, by July 2016 burned individuals showed similar number of shoots in both treatments: 9.1±1.4 and 8.7±0.9 shoots in biennial and annual treatments, respectively, while unburned individuals had 2.1±0.4 shoots (Table 5, Fig. 7a).

Height of *B. dumosa* was greater in unburned individuals in relation to both biennially and annually burned plants (Table 5, Fig. 6a), while for *C. claussenii* height in burned individuals (B and A), was greater in relation to unburned plants (Table 5, Fig 6d). Unburned individuals of *M. leiocephala* were only greater than annually burned ones, while no differences in height were found for *H. brasiliana* (Table 5, Fig. 6b-c). However, by July 2016 all shrub species showed similar heights in all treatments (Fig. 6). For the forb species, no differences in height were found for *I. fiebrigii* while annually burned individuals of *M. gracilis* were significantly greater than both unburned and biennially burned ones (Table 5, Fig. 7b).

Fire frequency had no effect in both diameter and height of the grass species *M. ferrugineum* (Table 6, Fig. 8a). *M. loliiforme* had significantly greater diameter in individuals that were both biennially and annually burned in relation to unburned ones, while height was only greater in annually burned individuals in relation to unburned ones (Table 6, Fig. 8b). Diameter of *O. ramosa* in annually burned individuals was significantly smaller than in both biennially burned and unburned plants (Table 6, Fig. 8c). Additionally, *O. ramosa* showed a significant increase in diameter over time (Table 6, Fig. 8c).

Flowering for *B. dumosa* and *H. brasiliana* was only greater after fires in relation to unburned individuals in 2014 (Table 7, Fig. 9). Flowering of *M. leiocephala* was similar in all treatments (biennial, annual and unburned), every year (2014, 2015 and 2016; Fig. 9). The shrub species *C. claussenii* and the forb species *I. fiebrigii* had similar flowering responses, with a higher number of flowering individuals following fires in all years; additionally, flowering occurred every year following fire in annually burned plants (Table 7, Fig. 9). Flowering of the grass species *M.*

ferrugineum and *O. ramosa* was only observed after fires, in both treatments (A and B), with no flowering observed in unburned plants (Table 7, Fig. 10).

Table 5. Parameters estimated by the mixed models for total number of shoots (GLMMs) and height (LMMs) of shrub and forb species in relation to treatments (unburned, biennial, annual fires) and time. Significant coefficients (p≤0.05) are given in bold.

		Number of shoots			**Height**		
Species	Effects	Coefficients	Standard error	*P* values	Coefficients	Standard error	*P* values
Bauhinia dumosa	intercept	0.67	0.12	<0.0001	2.95	0.59	0.00
	Biennial	**0.89**	0.12	<0.0001	**-0.28**	0.11	0.01
	Annual	**0.87**	0.13	<0.0001	**-0.38**	0.11	<0.001
	time	**-0.01**	0.00	<0.0001	0.002	0.01	0.76
Harpalyce brasiliana	intercept	-0.05	0.15	0.75	0.51	0.72	0.48
	Biennial	**0.71**	0.16	<0.0001	-0.003	0.09	0.97
	Annual	**0.79**	0.15	<0.0001	-0.15	0.09	0.13
	time	-0.002	0.002	0.31	-0.02	0.01	0.09
Mimosa leiocephala	intercept	0.42	0.17	0.02	1.93	0.78	0.01
	Biennial	**0.59**	0.21	0.005	-0.03	0.1	0.78
	Annual	**0.75**	0.21	<0.001	**-0.24**	0.1	0.03
	time	0.002	0.02	0.36	0.03	0.02	0.13
Chamaecrista claussenii	intercept	-0.12	0.16	0.43	1.45	0.32	0.00
	Biennial	**0.55**	0.12	<0.0001	**0.22**	0.06	<0.001
	Annual	**0.61**	0.12	<0.0001	**0.13**	0.06	0.02
	time	-0.001	0.003	0.56	**0.02**	0.01	<0.001
Mimosa gracilis	intercept	0.09	0.15	0.55	-0.42	1.42	0.76
	Biennial	0.16	0.17	0.34	0.42	0.21	0.05
	Annual	**0.41**	0.16	0.01	**0.63**	0.21	0.003
	time	**-0.01**	0.004	0.008	**-0.08**	0.03	0.006
Ipomoea fiebrigii	intercept	0.99	0.11	<0.0001	2.6	0.89	0.003
	Biennial	**0.68**	0.12	<0.0001	-0.22	0.14	0.11
	Annual	**0.99**	0.12	<0.0001	-0.07	0.13	0.61
	time	**-0.01**	0.002	<0.0001	-0.02	0.02	0.25

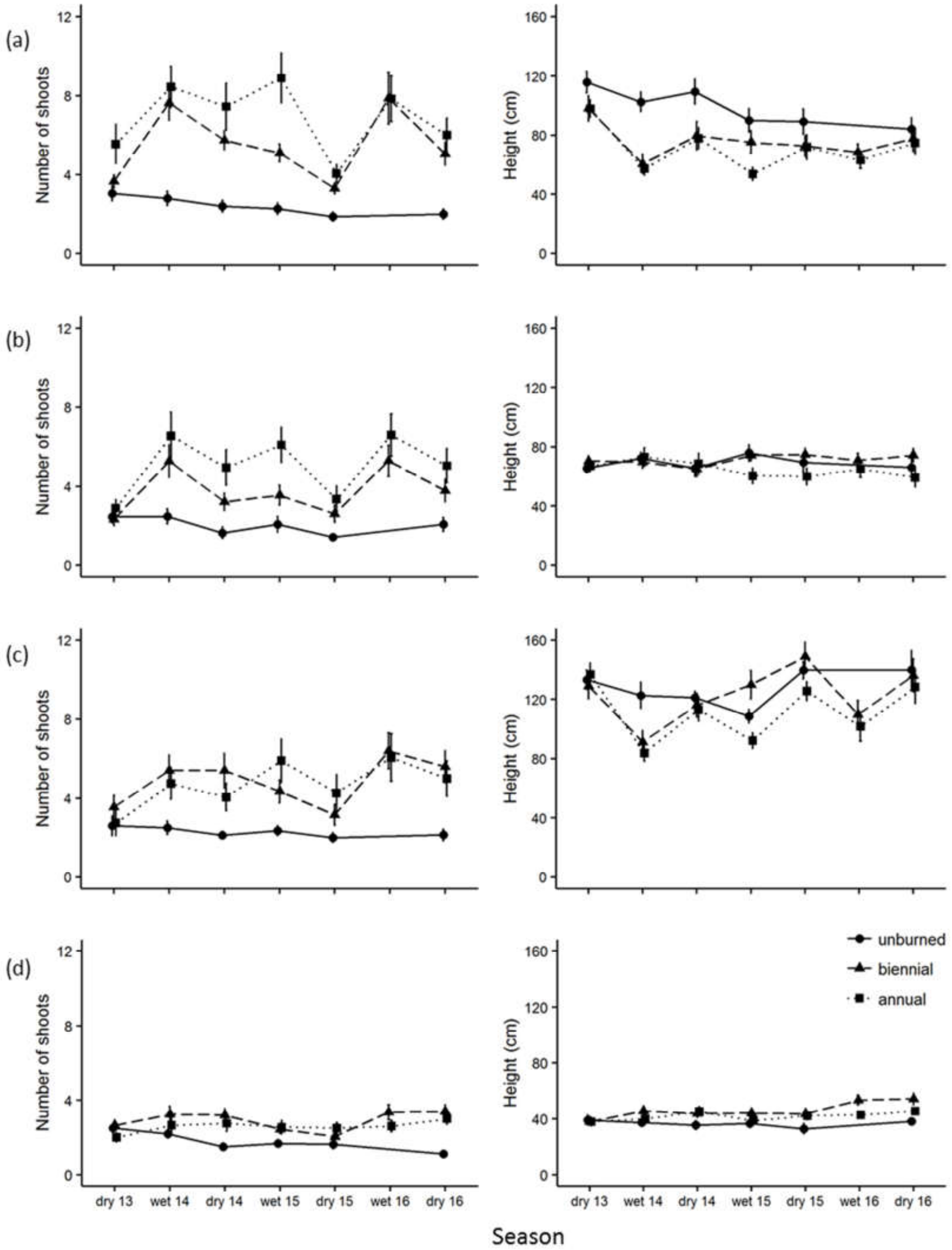

Figure 6. Number of shoots and height (mean ± SE) for a) *Bauhinia dumosa*, b) H*arpalyce brasiliana*, c) *Mimosa leiocephala* and d) *Chamaecrista claussenii* in unburned (solid line), biennial (dashed line) and annual (dotted line) fires prior to fires (dry 2013) and every six months after fires until 2016 in an open savanna area of the Serra do Tombador Nature Reserve. Biennial fires were set in the dry season of 2013 and 2015; annual fires in the dry season of 2013, 2014 and 2015.

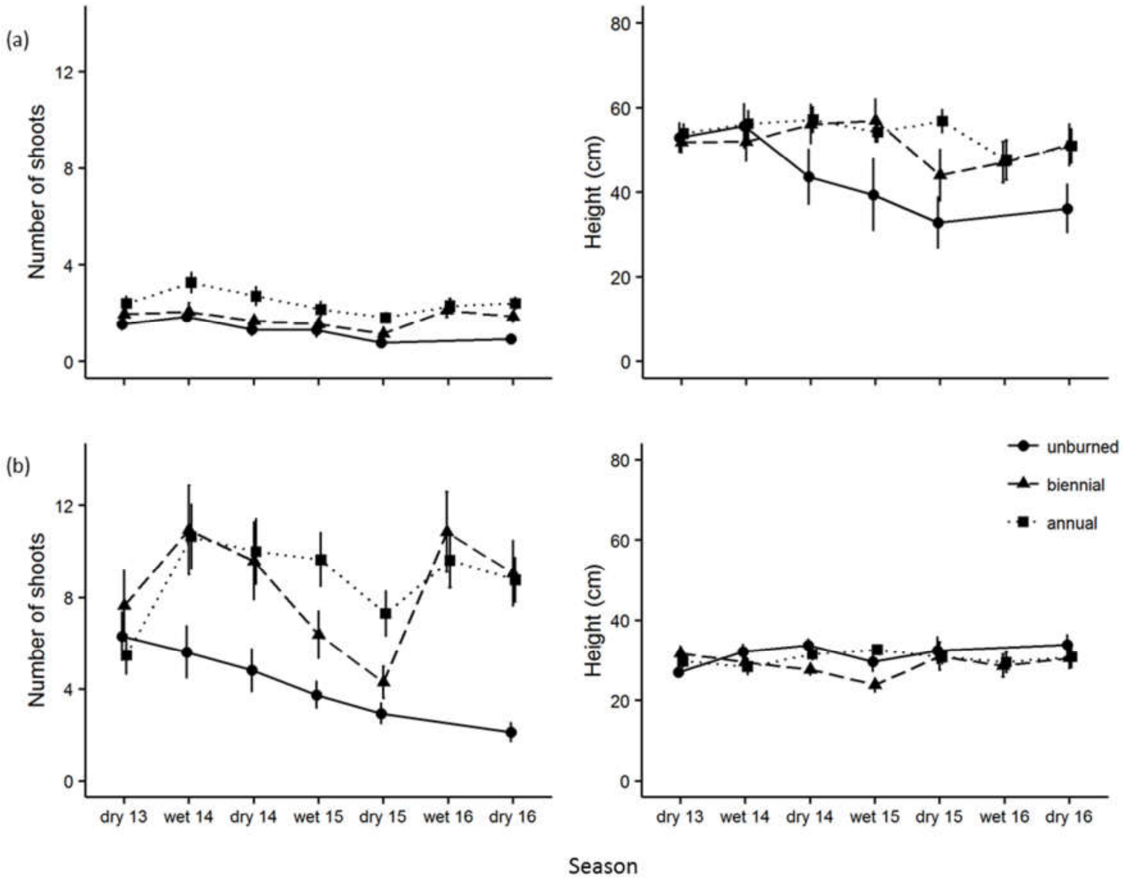

Figure 7. Number of shoots and height (mean ± SE) for a) *Mimosa gracilis* and b) *Ipomoea fiebrigii* in unburned (solid line), biennial (dashed line) and annual (dotted line) fires prior to fires (dry 2013) and every six months after fires until 2016 in an open savanna area of the Serra do Tombador Nature Reserve. Biennial fires were set in the dry season of 2013 and 2015; annual fires in the dry season of 2013, 2014 and 2015.

Table 6. Parameters estimated by the mixed models for diameter and height (LMMs) of grass species in relation to treatments (unburned, biennial and annual fires) and time. Significant coefficients (p≤0.05) are given in bold.

		Diameter			**Height**		
Species	Effects	Coefficients	Standard error	*P* values	Coefficients	Standard error	*P* values
Mesosetum ferrugineum	intercept	1.73	0.18	0.0	2.74	0.21	0.0
	Biennial	-0.06	0.04	0.08	0.02	0.03	0.58
	Annual	-0.04	0.04	0.22	-0.005	0.04	0.89
	time	-0.002	0.006	0.97	-0.005	0.006	0.42
Mesosetum loliiforme	intercept	1.55	0.16	0.0	1.54	0.38	<0.001
	Biennial	**0.26**	0.06	<0.001	0.1	0.06	0.08
	Annual	**0.28**	0.06	<0.001	**0.13**	0.06	0.03
	time	**0.04**	0.01	<0.001	0.01	0.01	0.06
Oncorachis ramosa	intercept	2.24	0.15	0.0	2.36	0.46	0.0
	Biennial	-0.14	0.07	0.05	-0.11	0.05	0.05
	Annual	**-0.31**	0.07	<0.001	**-0.15**	0.05	0.005
	time	**0.04**	0.01	0.003	0.007	0.01	0.4

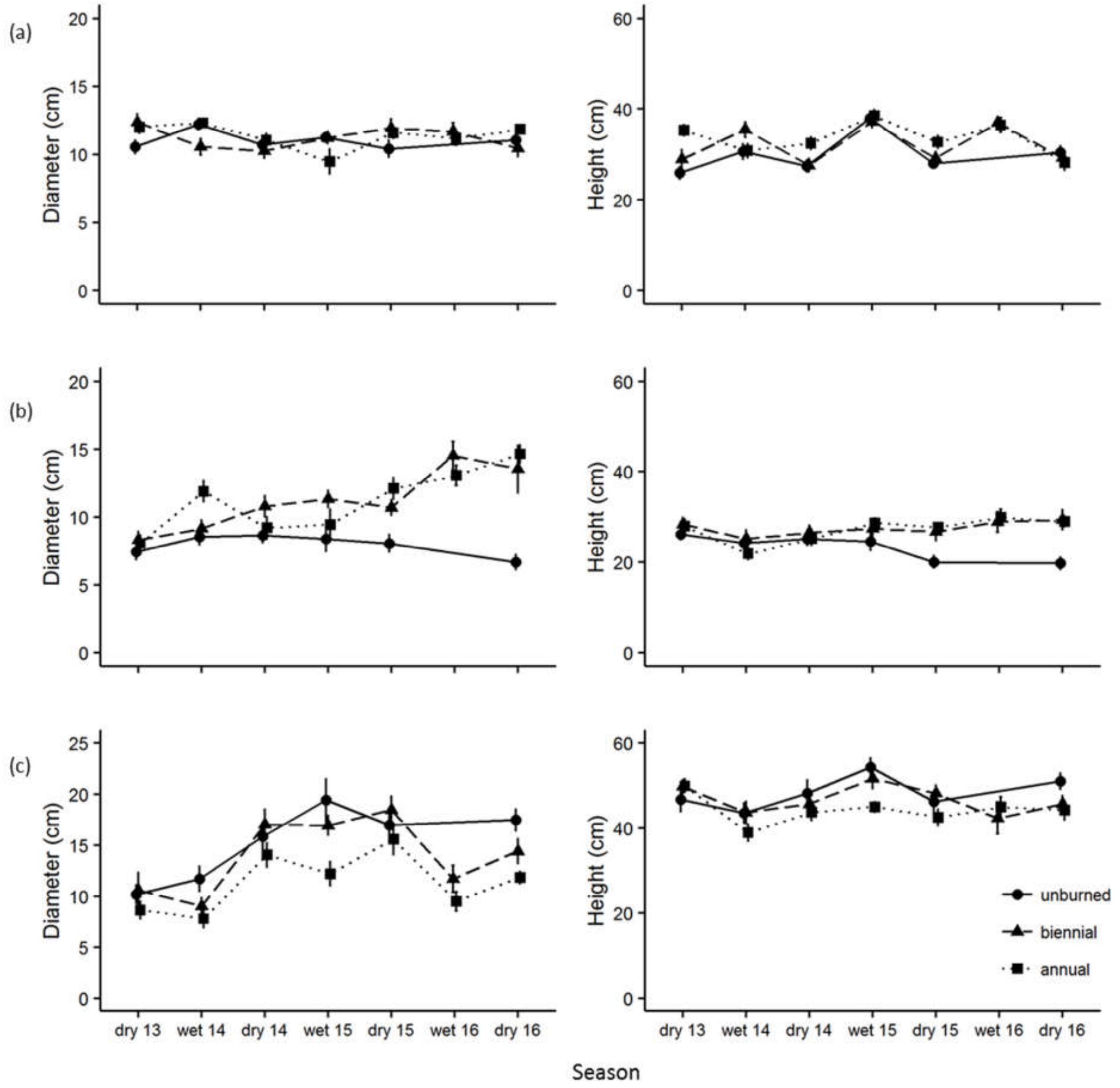

Figure 8. Number of shoots and height (mean ± SE) for a) *Mesosetum ferrugineum*, b) *Mesosetum loliiforme* and c) *Oncorachis ramosa* in unburned (solid line), biennial (dashed line) and annual (dotted line) fires, prior to fires (dry 2013) and every six months after fires until 2016 in an open savanna area of the Serra do Tombador Nature Reserve. Biennial fires were set in the dry season of 2013 and 2015; annual fires in the dry season of 2013, 2014 and 2015.

Table 7. Parameters estimated by the GLMMs for flowering responses of species to the treatments (unburned, biennial and annual fires) in each year. Coefficients correspond to the logodds of flowering; significant coefficients (p≤0.05) are given in bold.

		Year 2013/2014			**Year 2014/2015**			**Year 201/2016**		
Species	Effects	Coefficients	Standard error	*P* values	Coefficients	Standard error	*P* values	Coefficients	Standard error	*P* values
Bauhinia dumosa	intercept	-1.75	0.01	<0.001	-2.32	1.19	0.05	-1.12	0.76	0.13
	Biennial	**1.3**	0.59	0.03	-1.12	1.64	0.49	0.42	0.97	0.66
	Annual	**2.21**	0.01	<0.0001	1.53	1.38	0.26	0.89	0.96	0.35
Harpalyce brasiliana	intercept	-1.45	0.56	0.009	1.23	0.73	0.08	-	-	-
	Biennial	**2.4**	0.76	0.001	0.41	0.95	0.67	-	-	-
	Annual	0.99	0.76	0.17	0.18	0.89	0.84	-	-	-
Mimosa leiocephala	intercept	2.77	1.03	0.007	1.88	0.76	0.013	1.39	0.68	0.04
	Biennial	-1.03	1.21	0.38	-0.13	0.98	0.89	0.34	0.91	0.7
	Annual	-1.45	1.17	0.22	-0.19	0.99	0.84	-0.36	0.85	0.67
Chamaecrista claussenii	intercept	-2.81	1.18	0.017	-3.09	1.47	0.03	-2.81	1.18	0.017
	Biennial	**4.7**	1.57	0.002	-0.41	1.83	0.8	**4.7**	1.57	0.002
	Annual	**3.04**	1.32	0.02	**4.25**	1.87	0.03	**3.04**	1.32	0.02
Mimosa gracilis	intercept	-2.97	1.05	0.004	-2.64	1.03	0.01	-	-	-
	Biennial	**3.6**	1.18	0.002	-0.31	1.46	0.83	-	-	-
	Annual	**3.3**	1.17	0.004	1.79	1.44	0.12	-	-	-
Ipomoea fiebrigii	intercept	-0.15	0.63	0.8	-0.81	0.57	0.16	-2.09	1.46	0.15
	Biennial	1.82	1.07	0.09	0.68	0.37	0.37	4.43	2.49	0.07
	Annual	**3.16**	1.39	0.02	**2.85**	0.99	0.004	**5.12**	2.49	0.04
Mesosetum ferrugineum	intercept	-2.94	1.02	0.004	-3.24	1.46	0.02	-2.69	1.03	0.01
	Biennial	**5.89**	1.45	<0.0001	-0.1	1.68	0.9	**5.58**	1.45	<0.0001
	Annual	**4.68**	1.2	<0.0001	**5.85**	2.29	0.01	**5.53**	1.45	<0.0001
Oncorachis ramosa	intercept	-2.94	1.02	0.04	-2.98	1.09	0.006	-2.94	1.02	0.004
	Biennial	**3.15**	1.12	<0.0001	0.0	1.47	1	**3.72**	1.14	0.001
	Annual	**3.35**	1.12	<0.0001	**3.85**	1.26	0.002	**2.94**	1.12	0.008

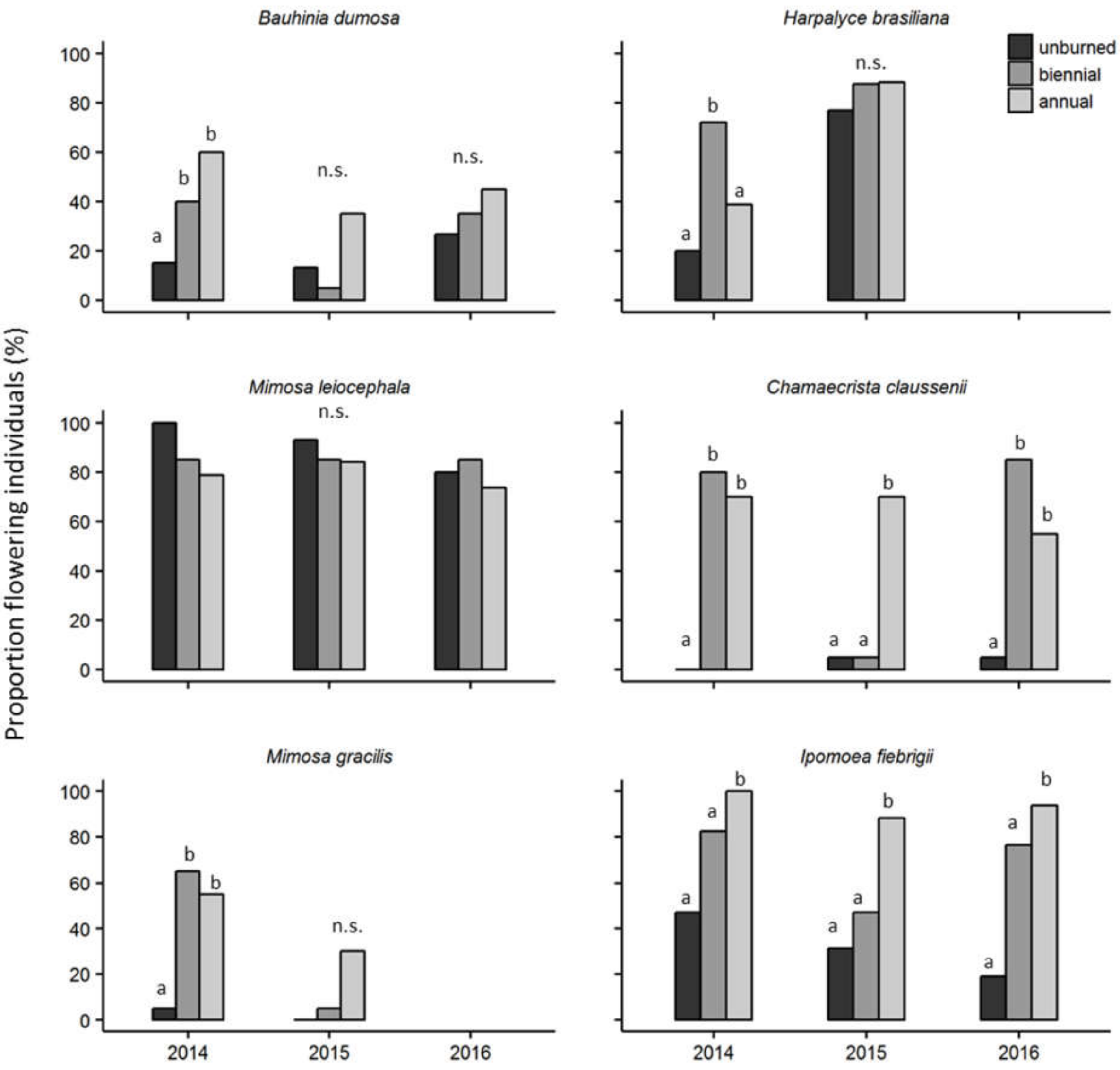

Figure 9. Proportion of flowering individuals for shrub and forb species in unburned, biennially and annually burned plants for each year following fires (2014, 2015 and 2016) in an open savanna area of the Serra do Tombador Nature Reserve. Different smallcase letters indicate significant differences (p≤0.05) among treatments

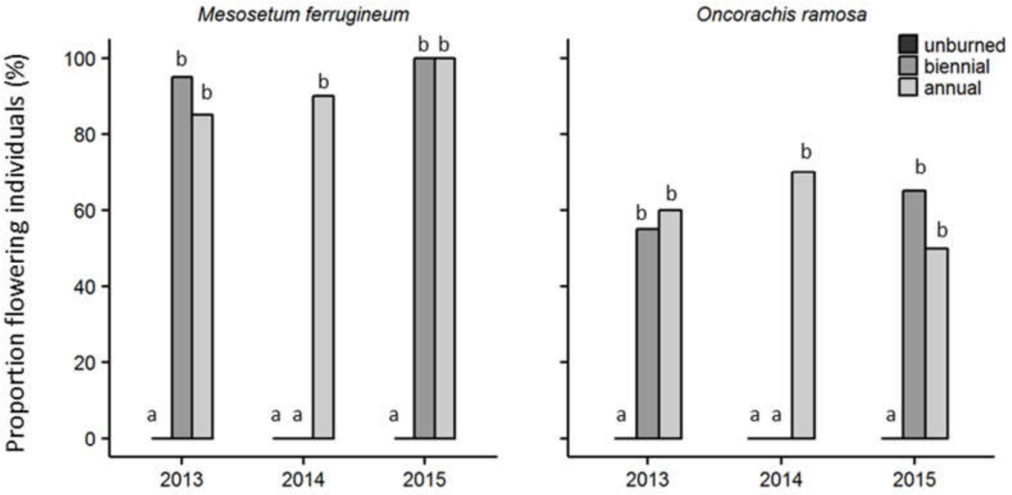

Figure 10. Proportion of flowering individuals for two grass species in unburned, biennially and annually burned plants for each year following fires (2013, 2014 and 2015) in an open savanna area of the Serra do Tombador Nature Reserve. Different smallcase letters indicate significant differences ($p \leq 0.05$) among treatments

Discussion

All individuals of all species were able to resprout after fires in all seasons, suggesting that the belowground temperatures reached in the different fire seasons did not damage buds. For most species, no differences in number of shoots or diameter were found between treatments, except for *C. claussenii* (shrub), which had a higher number of shoots after mid and late-dry season fires, and *M. leiocephala* (shrub) and *M. ferrugineum* (grass), which had higher number of shoots and diameter after early-dry season fires. However, differences in height were found for most species, where height after mid and late-dry season fires was greater than after early-dry season fires. Considering that both number of shoots and height can be considered as estimates for resprouting vigour (Enright et al. 2011; Fernandez et al. 2013), the overall effects of fire season observed in this study point to a negative effect of early-dry season fires on resprouting vigour, contrary to our initial prediction that late-dry season fires would be more detrimental due to greater amount of dead fuel loads and higher flammability, and consequently greater fire intensity and severity.

Although plant moisture contents, fire intensity and rates of fire spread varied greatly across fires seasons, there were no significant differences in these parameters among seasons (Rissi et al. in press). Thus, the differences in resprouting responses observed were likely not due to differences in fire parameters (intensity, severity and rates of fire spread) across seasons. Additionally, fire intensity and severity have recently been shown to be poor predictors of post-fire vegetation responses in surface fire systems as they do not account for belowground soil heating, hence being poor indicators of damage to bud-bearing belowground organs and seeds (Gagnon et al. 2010, 2015). Although we did not measure belowground temperatures, temperatures 1cm belowground usually increase up to around 50ºC (Miranda et al. 1993), ensuring survival of belowground buds (Choczynska and Johnson 2009; Gagnon et al. 2010).

After fires, resprouting plants rely on carbohydrate reserves to sustain regrowth and respiration demands until the plant has recovered enough photosynthetic tissue to support these costs (Chapin et al. 1990; Bowen and Pate 1993; Schutz et al. 2009). Therefore, resprouting responses could be limited by amount of stored reserves, and variation of resprouting responses across seasons could be attributed to differences in amount of carbohydrates available at the time of disturbance, where plants would regenerate slower and with less vigorous resprouts if topkilled when their reserves are low (Bowen and Pate 1993; Landhausser and Liefer 2002; Drewa et al. 2006; Hmielwoski et al. 2014). In tropical climates, storage reserves are relatively lower during the wet (growing) season, increasing to higher amounts during the dry season (Wurth et al. 2005; Schutz et al. 2009). Thus, amounts of stored reserves should be lower during the early-dry season in relation to the amounts in mid and late-dry season and could explain the less vigorous resprouting responses found after early-dry season fires as growth was relatively slower in these plants.

However, several studies have found no correlation between amount of stored reserves and resprouting responses (Hoffmann et al. 2000; Wildy and Pate 2002; Cruz et al. 2003a), and resprouting plants of different fire-prone ecosystems seem to have more than enough stored carbohydrates to support regrowth after a single disturbance event (Canadell and Lopez-Soria 1998;

Hoffmann et al. 2000; Cruz and Moreno 2001; Wildy and Pate 2002; Cruz et al. 2003b), hence other factors may be limiting. For instance, Cruz et al. (2002, 2003a) found that variation in resprouting responses of a Mediterranean-type shrub was largely driven by site water and nutrient availability. Wildy and Pate (2002) reported that variation in shoot growth of resprouting *Eucalyptus kochii*, a Western Australian shrub, was more related to climatic conditions, where less than 10% of root reserves were used to support regrowth. Hoffmann et al. (2000) reported that regrowth of the resprouting Cerrado tree *Kielmeyera coriacea* was not limited by amount of nonstructural carbohydrates, but might have been limited by nutrient availability. We argue that the post-fire environment, mainly rainfall patterns after fire (Giljohann et al. 2017), may explain the differences in resprouting responses observed in this study. Recovery after early-dry season fires was slower as the resprouting plants endured all five months of the dry season, however, after the wet season, plants burned in different seasons had similar sizes, both in number of shoots and in height. Community regeneration showed the same patterns, where regeneration of aboveground biomass of open savanna was slower after early-dry season fires in relation to mid and late-dry season fires, but after the rainy season, aboveground biomass was similar among treatments (Rissi 2016).

Resprouting responses after three consecutive annual burns were not different than after biennial burns. These results suggest that, contrary to our prediction, resprouting responses of these plants were not limited by fire frequency. Repeated disturbances have been shown to reduce carbohydrate reserves, thus affecting resprouting responses (Canadell and Lopez-Soria 1998; Paula and Ojeda 2009). However, the interval needed for restoring carbohydrate reserves to pre-burn levels can be highly variable among species. Schutz et al. (2009) reported that root reserves of a South African savanna tree were replenished within one year after fires. Miyanishi and Kellman (1986) reported that the starch concentrations of two Neotropical savanna shrubs were restored to pre-burn levels in a period between one to two years after fires, suggesting that shorter fire intervals could result in cumulative depletions of root reserves. Nonetheless, after five consecutive annual burns, root starch concentrations of both shrub species were depleted by 50% compared to unburnt plants, indicating these species would be able to resprout after many consecutive burns before depleting all stored reserves. However, Hoffmann et al. (2000) suggested that plants in frequently burnt systems might display a rather conservative strategy, using small amounts of stored reserves to support regrowth, quickly reverting to photosynthesis to actively support the costs of respiration and growth. Our results show that, overall, species had a higher number of shoots after fire, compared to unburnt plants, with rapid recovery of photosynthetic tissue, which could indicate such strategy and explain the high resilience of these species to frequent fires.

Additionally, fire, rather than a specific burning season, seems to be more important for reproduction: fire induced flowering of all species, except *M. leiocephala,* which flowered prolifically irrespective of fire season and frequency. Thus, fire can be considered a critical factor for the reproduction of these species, especially grasses, as has been reported for other savanna and grassland systems (Hiers et al. 2000; Brewer et al. 2009; Fidelis and Blanco 2014), including the Cerrado (Araújo et al. 2013; Rissi 2016). Post-fire flowering could also be seen as a conservative strategy of plants in frequently disturbed environments: by suppressing flowering during periods without fire, plants would be storing resources that could be used for growth, including clonal growth (Brewer et al. 2009). Moreover, it would also allow plants to avoid unfavorable conditions, ensuring flowering, and consequently seed dispersal and recruitment, would occur in more favorable conditions as the post-fire environment is associated with reduced competition and enhanced resource availability (Kauffman et al. 1994; Whelan 1995; Neary et al. 1999; Fidelis and Blanco 2014). Brewer et al. (2009) suggest that frequent fires could increase fitness of species by increasing the opportunities for reproduction.

Our results have shown that, contrary to previous belief, cool fires at the beginning of the dry season have negative effects on resprouting responses, at least during the early resprouting stages. Surprisingly, three consecutive burns did not affect resprouting and flowering responses of the species, indicating that these species are highly resilient to frequent fire regimes. However, further studies should be carried out with annual burns for a longer period to evaluate if resprouting responses would be affected. We also argue that quantifying amounts of stored reserves at the time of burning and after fires are necessary to fully understand the mechanisms of such high resilience in frequently burned ecosystems.

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Considerações Finais

O principal objetivo desta tese era investigar a regeneração pós-fogo de espécies de Cerrado de campo sujo. Assim, avaliamos o papel da regeneração via rebrote e via semente. Além disso, procuramos avaliar os fatores que estão envolvidos no rebrote das plantas após serem submetidos a diferentes tipos de distúrbio, e distúrbios que ocorreram em diferentes frequências e épocas do ano, visto que tanto o tipo do distúrbio quanto a época e a frequência em que ele ocorre podem afetar a regeneração das plantas.

Deste modo, no primeiro capítulo, verificamos que o rebrotamento é a principal estratégia de regeneração pós-fogo e ocorre principalmente a partir de gemas localizadas em estruturas subterrâneas que estão altamente protegidas pelo solo, que é um excelente isolante térmico (Choczynska & Johnson 2009). Além disso, verificamos que o recrutamento via semente tem um papel pequeno na regeneração pós-fogo do Cerrado, tendo em vista que poucas espécies apresentaram aumento na germinação após serem expostas às altas temperaturas. No entanto, as sementes de 96% das espécies foram consideradas tolerantes, sobrevivendo após sendo expostas a 100^{o}C. Durante o fogo, as temperaturas abaixo do solo raramente atingem acima dos 100^{o}C (Zupo et al. dados não publicados), portanto, a tolerância das sementes a esta temperatura sugere que sementes que estiverem enterradas no banco de sementes do solo podem sobreviver ao fogo, e o recrutamento pode ocorrer quando as condições ambientais forem favoráveis. Porém, a sobrevivência das sementes diminui com temperaturas mais altas, como aquelas encontradas na superfície do solo durante as queimas (Daibes et al. 2017), diminuindo a probabilidade de sementes recém-dispersas de sobreviver ao fogo. Deste modo, a regeneração via rebrote é essencial para a manutenção das populações em ambientes que, como o Cerrado, sofrem distúrbios frequentes (Coutinho 1990; Pivello 2011; Durigan & Ratter 2016), apresentam alta produtividade (Kauffman et al. 1994) e possuem poucas espécies que formam bancos de semente persistentes (Salazar et al. 2011; Andrade & Miranda 2014).

Tendo em vista que o rebrotamento é o principal mecanismo de persistência das populações em tais ambientes, no capítulo 2, avaliamos se a reposta das plantas a diferentes tipos de distúrbio ocasionariam em respostas semelhantes em relação ao rebrotamento e à floração. Verificamos que o fogo não foi um distúrbio mais severo que o corte, uma vez que todos os indivíduos de todas as espécies rebrotaram tanto após serem cortados como após serem queimados. Isto sugere que as temperaturas causadas pelo fogo não danificaram as gemas. Além disso, verificamos que, tanto para as espécies herbáceas quanto para as gramíneas, não houve diferenças no rebrotamento entre plantas cortadas e queimadas, sugerindo que a remoção da biomassa aérea foi o fator determinante. No entanto, para três espécies arbustivas, observamos um maior número de ramos em plantas queimadas em relação às cortadas, sugerindo que outros fatores, além da remoção da biomassa, estariam atuando. Também verificamos um aumento na floração para uma espécie arbustiva e uma herbácea em plantas queimadas. Concluímos que o aumento na disponibilidade de recursos após o fogo, resultante tanto da combustão da biomassa e posterior deposição das cinzas, quanto da redução da competição, é um fator importante e que pode influenciar as respostas relacionadas ao rebrote e floração das espécies.

No terceiro capítulo, verificamos os padrões da regeneração pós-fogo em diferentes regimes de queima, por meio de queimadas experimentais em diferentes épocas do ano (precoce – início da estação seca, modal – meio da estação seca e tardia – final da estação seca) e em diferentes frequências (bienal, anual e sem queima). Observamos que as queimas no início da estação seca (precoce) afetaram negativamente o rebrotamento para a maioria das espécies estudadas em relação a queimas modais e, principalmente, queimas tardias. No entanto, as diferenças foram significativas para altura e não para número de ramos, indicando que estas não devem ter sido relacionadas a diferenças nos parâmetros do fogo (por exemplo, intensidade, taxa de propagação e temperatura) entre as diferentes épocas. As diferenças encontradas se devem a diferenças na disponibilidade de recursos, principalmente do ambiente, já que as plantas queimadas no início da estação seca estão sujeitas a um número maior de meses com déficit hídrico. Além disso, depois da estação chuvosa, os indivíduos apresentaram tamanhos similares (em número de ramos e altura) em todos os tratamentos, indicando que a disponibilidade de água no ambiente é um fator determinante na regeneração pós-fogo no Cerrado. Assim, queimas no início da seca resultaram em rebrotes menos vigorosos.

Também observamos que queimas anuais não afetaram a regeneração das plantas em relação a queimas bienais, sugerindo que o rebrotamento não foi limitado pela quantidade de reservas armazenadas, ao menos não com três queimas consecutivas. Os indivíduos queimados, tanto anualmente quanto bienalmente, apresentaram um número maior de ramos em relação a indivíduos não queimados, recuperando rapidamente sua biomassa aérea e, portanto, o tecido fotossinteticamente ativo, sugerindo que estas plantas optem por uma estratégia mais conservadora em relação ao uso das reservas armazenadas, voltando rapidamente a utilizar os produtos da fotossíntese para sustentar o crescimento e manutenção. No entanto, estudos que quantifiquem a quantidade de reservas armazenadas antes e depois da queima são necessários para um melhor entendimento dos mecanismos que permitem a alta resiliência das espécies de Cerrado a regimes de fogo tão frequentes. Com isso conseguiremos compreender não só quanto tempo leva para restaurar o estoque de reservas para os níveis antes da queima, mas também a quantidade de reservas que é mobilizada após tais distúrbios. Além disso, verificamos que o fogo, e não uma época de queima específica, parece ser o estímulo mais importante na reprodução das espécies destes ambientes. O fogo induziu a floração na maioria das espécies, em relação a indivíduos não queimados, de modo que a ocorrência do fogo pode aumentar o fitness das espécies, já que aumenta as chances de muitas plantas reproduzirem.

Assim, as espécies de Cerrado campo sujo podem ser consideradas excelentes rebortadoras, com gemas altamente protegidas localizadas em estruturas subterrâneas, e que rebrotam vigorosamente caso a disponibilidade de recursos do ambiente pós-fogo não seja limitante. Deste modo, a capacidade de rebrotar possibilita a persistência das populações nestes ambientes e o papel das sementes para a regeneração é secundário, estando mais relacionado com a dispersão já que o fogo induz a floração das espécies.

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