

Effects of pasture-burning management on anuran communities in subtropical Brazilian grasslands

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Abstract

Anthropogenic fire is a worldwide event that affects many ecosystems and organisms. In Southern Brazil, grassland management with fire has been highly employed since the mid-18th century. Although the practice is regulated by federal law (prescribed fire), there is no detailed information about the impacts of this practice on the small, non-volant fauna. We evaluated the effect of fire management on anuran species richness and community composition in Brazilian grassland areas that have adopted this practice for more than 15 years. Our results show that burning practices lead to a reduction of anuran richness. About 37% of the species occur exclusively in sites free of fire. Sites with fire management have low densities of taller grass and shrubs, which could reduce habitat availability for some anuran species. Nestedness and turnover components of beta diversity did not differ within and between treatments, but there was a tendency for a nestedness organization of the community in burned sites, suggesting that sites with fire management are a subsample of sites where fire is absent. Our results pointed out that prescribed fire practices have potentially negative effects on the anuran diversity. These results suggest that the changes in vegetation, in particular percentages of shrub cover, affect habitat suitability for some species. Therefore, anuran communities tend to become less diverse and lack arboreal species where fire occurs.

Abstract in Portuguese is available with online material.

KEYWORDS

amphibians, Beta diversity, Brazil, fire, grasslands, pasture

1 | INTRODUCTION

Grasslands play an important role in agriculture and animal production and at the same time serve as habitat for several species. For many centuries, grasslands have been managed for animal production and pasture, resulting in a negative impact on several species (Metzger et al., 2010). Vegetation burning, for example, is a worldwide management practice that has been applied to facilitate cattle production in grasslands and savanna-like habitats (Boldrini, 2009; Fidelis

& Pivello, 2011; Pillar & Lange, 2015; Ramos-Neto & Pivello, 2000). Fire has a great power in shaping the ecosystem structure (Bond et al., 2005; Bond & Keeley, 2005), and the effects of natural and anthropogenic fires on native fauna have been extensively studied in the last decades (Masterson et al., 2008). Considering this, habitat management practices, which include vegetation burning, are a powerful trigger to ecosystem modification (Hobbs & Huenneke, 1992).

Fire can harm animals directly by increasing mortality rates through burns, as well as through asphyxiation or intoxication

caused by smoke inhalation. Additionally, fire promotes changes in vegetation structure, creating harsher environmental conditions (e.g., dryer microclimate) and loss of microhabitats for shelter and foraging (Pilliod et al., 2003). This negative effect is more dramatic for less mobile organisms, particularly anurans, as many species have permeable skin and high association with moist habitats (Duellman & Trueb, 1994). However, some studies indicate that, in some cases, burning processes can increase species richness (Klaus & Noss, 2016; Mester et al., 2015; Schurbon & Fauth, 2003) and abundance for some species (Brown et al., 2011; Hossack et al., 2013; Kirkland Jr et al., 1996; Perry et al., 2012). The positive effects of fire on anuran fauna are related to changes in the vegetation structure, which leads to the creation of new microhabitats (Bixby et al., 2015). Consequently, we could argue that, although the fires affect the anurans negatively (Allingham & Harvey, 2013; Cano & Leynaud 2010; Pilliod et al., 2003), they could favor some species (Klaus & Noss, 2016; Mester et al., 2015; Schurbon & Fauth, 2003). However, it is important to consider the species' adaptation to environments that have historically evolved with the periodic action of burn (Brooks et al., 2004).

The native vegetation cover on the highlands of Southern Brazil has been managed with fire for the last 7400 years (Behling et al. 2004), suggesting anthropogenic fires since the arrival of the first humans, but with greater intensity since the mid-18th century (Boldrini, 2009; Fidelis et al., 2010). At present days, these grasslands suffer from prescribed burning for pasture management, which is allowed by Brazilian laws (Pillar & Lange, 2015). Natural fire is part of this ecosystem dynamics, with some plant species, for example, adapted to burning events (Overbeck et al., 2018). However, anthropogenic fire, used with the intent of preparing the land for cattle ranching (fire management), has caused fire events to occur with a frequency and intensity far greater than those expected from natural causes (Ferrando et al. 2016) or by human up to mid-18th century (Boldrini, 2009; Fidelis et al., 2010). Thus, despite being an ecosystem integrated and shaped by fire, it is expected that fire caused by human activity from the last century brings negative impacts on biodiversity. However, these impacts are poorly understood or, for some taxa, completely unknown (Ferrando et al. 2016). Although negative effects of fire on anurans were reported in studies from Neotropical habitats (Cano & Leynaud 2010; Papp & Papp 2000; Rocha et al. 2008), research studies disagree on the positive and negative effects of fire on the whole system (Carlucci et al., 2016; Luza et al., 2014; Overbeck et al., 2016, 2018; Pillar et al., 2009; Pillar & Vélez, 2010). However, most studies evaluating the effects of fire on frogs were carried out in temperate and tropical forests, with few evaluating savanna environments and even fewer evaluating grasslands (dos Anjos et al., 2021).

Results of manipulative grassland-burning experiments showed that fire induces taxonomic and/or functional changes in assemblages of spiders, edaphic detritivorous invertebrates, grasshoppers, and thrips, but also evidenced strong community resilience due to the rapid (ca. 6–12 months) return to unburned-like conditions (Podgaiski et al., 2013, 2014, 2018). Natural experiments, comparing

managed and unmanaged (not management by fire) grasslands, evidenced similar richness but distinct taxonomic compositions for ground-dwelling ants (Albuquerque et al., 2017), while small non-flying mammals showed simplified species assemblages under live-stock and burning management (Luza et al., 2016; Pedó et al., 2010). However, the effects of fire on amphibians in this region are still uncertain.

As presented above, there is some disagreement on the negative effects of fire on Brazilian grasslands. Regarding anurans, habitat loss generated by the expansion of agriculture and animal production areas is considered a main threat to their conservation in Brazil. The southern Brazilian grasslands encompass about 84 amphibian species, of which 14% are endemic (Santos et al., 2014). Beyond the threats imposed by habitat loss and fragmentation (Garcia & Vinciprova, 2003), fire could represent a significant and poorly evaluated impact on amphibian communities in this region. In this study, we performed field samplings in grasslands of Southern Brazilian highlands to evaluate the effects of grassland burning management on anurans assemblages. We hypothesize that (1) species richness will be higher where fire management is absent, (2) the abundance of individuals will be higher in sites without fire management, and (3) sites without fire management will present a larger number of exclusive species.

2 | METHODS

2.1 | Study site

Sampling was performed in the highland grasslands of Southern Brazil. The region is locally called “Campos de Cima da Serra”, which is part of the Atlantic Forest domain. The sample units were distributed in the municipalities of Camará do Sul and São Francisco de Paula (29°10'35.81" S and 50°10'06.93"O) (Figure 1). The habitat is formed by a wide area of grasslands interspersed by a mosaic of forest (*Araucaria* forest) patches (Pillar & Quadros, 1997). Following Köppen, the local climate is subtropical with an average air temperature of 18.5°C. The rainfall is well-distributed through the seasons, with an average annual precipitation of 2252 mm.

2.2 | Sampling design

We selected four sampling sites in areas with regular fire management and four in areas without fire management (totalling eight sampling sites with two treatments: with and without fire practices). We were unable to apply a random distribution of sampling sites due to the low availability of sites without fire practices. Thus, we adopted a preferential sampling design based on a map of fire spots obtained from time-series satellite images from the last 15 years provided by the Brazilian Space Agency (Instituto Nacional de Pesquisas Espaciais). Afterwards, eligible sites for each treatment were checked personally to achieve detailed information about the use of fire by

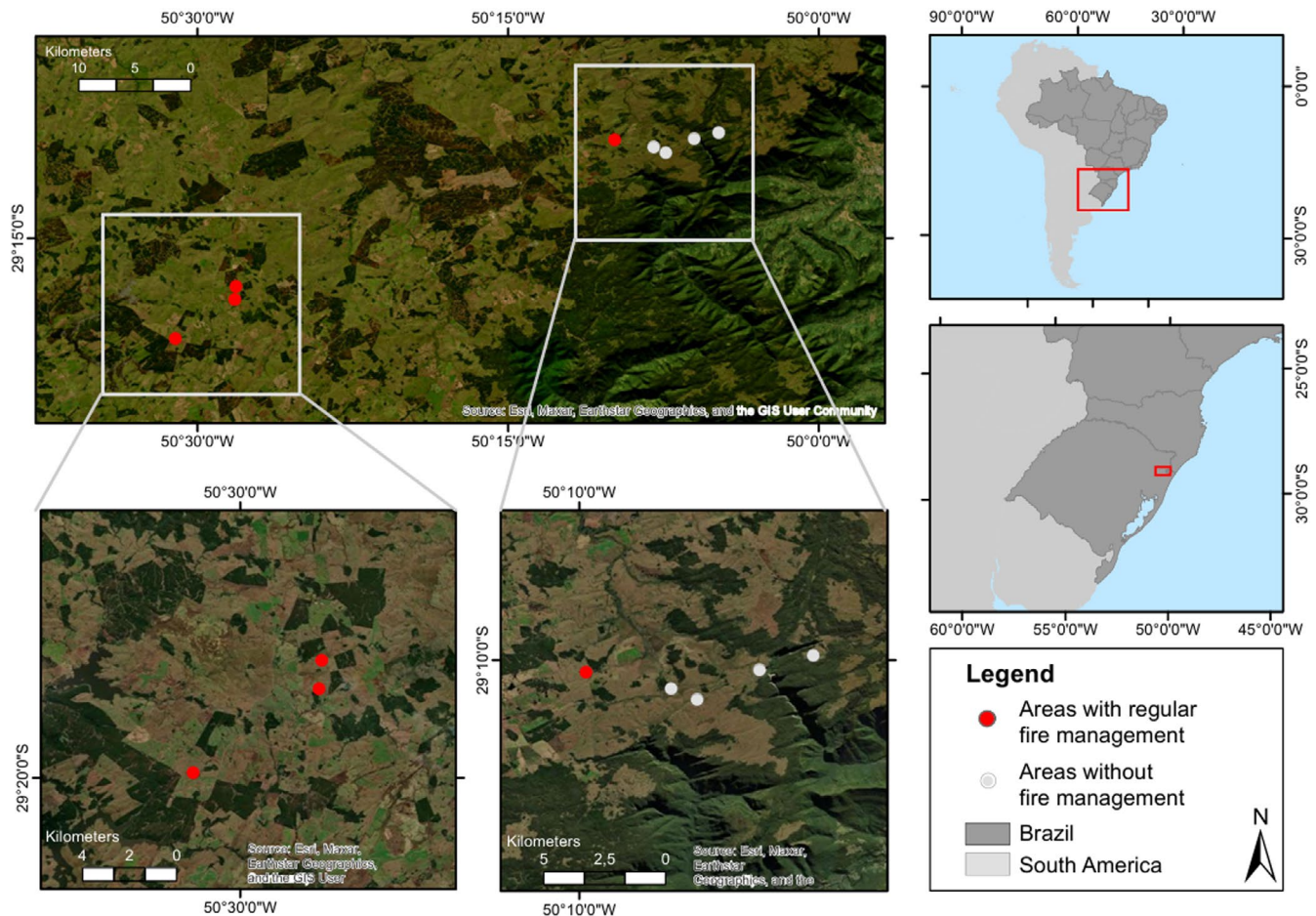


FIGURE 1 Map of sampling locations. Red dots indicate ponds in areas with regular fire management and gray dots indicate ponds in areas without fire management. Map prepared by Gabriela Morais Olmedo.

landowners (cattle ranchers): frequency of burning and extension and limits of the burning area. Distances between sampling sites were at least 5 km between treatments and at least 2 km within the same treatment (Figure 1). We performed a Mantel test to evaluate the spatial independence of the sampling sites, which revealed them to be spatially independent ($p > .05$).

All sampling sites with fire management shared the following characteristics: presence of similar densities of cattle, fire applied seasonally from July to September, annual burning in the last 15 years, and burning practices based on a traditional method for cattle management (see supplementary material—Data S1). We confirmed the existence and frequency of burning practices by interviewing landowners and cross-referencing this information with satellite images from both the current year and the past 15 years. The burning area was at least three times larger than the sampling site and encompassed the whole sampling site plus the surrounding area. All sampling sites without fire management were located within an area of 13,141.05 hectares, which, according to official records, has been excluded from fire for at least 15 years. The sampling sites within this area are situated in protected areas or conservation units, such as Parque Nacional Aparados da Serra, where the use of fire for cattle management was discontinued at least 15 years ago.

2.3 | Assessing anuran diversity

We recorded anuran species in each sampling site through surveys in natural ponds used as breeding sites. Surveys based on breeding sites are a frequent procedure in studies of amphibians and allow a fast and reliable assessment of local diversity (Heyer et al. 1994, Madalozzo et al., 2017). Selected breeding sites are in areas of the field that retain rainwater and form marshy zones. They resemble temporary ponds with shallow water, averaging 20 cm, up to 1 m in the deepest part (Figure S1). Due to the flat landscape, the ponds lack steep banks, allowing the water to blend into the surrounding habitat. Thus, fire is gradually halted as it reaches the moist soil before reaching the pond basin. We monitored one pond in each sampling site, which was selected in a preferential manner based on the presence of a large number of species and the criteria of homogeneity of their configuration as follows: all ponds have a circular design, are less than 1 m deep, and have total area varying from 500 m² to 580 m². It is important to highlight that additional ponds per site were monitored during the initial phase of the sampling. However, only those with the previously described characteristics were used as breeding sites by anurans during the sampling period. Selected ponds were monitored monthly from September to October 2016

TABLE 1 Coordinates of sampling site and microhabitat variables surveyed in the eight sampling units in the subtropical fields of southern Brazil.

| Sampling sites | Latitude | Longitude | AD (cm) | FV (%) | CSV (%) | AHG (cm) | pH | ECW (ms/cm) | BOD (mg/L) |
|----------------|------------|------------|---------|--------|---------|----------|------|-------------|------------|
| PF1 | -29.299113 | -50.470093 | 19.4 | 1 | 1 | 17.8 | 7.37 | 27.5 | 17.8 |
| PF2 | -29.288531 | -50.469056 | 27.9 | 1 | 1 | 16.0 | 8.24 | 22.0 | 22.1 |
| PF3 | -29.331199 | -50.518006 | 13.8 | 2 | 1 | 24.7 | 7.80 | 24.8 | 19.9 |
| PF4 | -29.171044 | -50.164293 | 31.3 | 2 | 1 | 11.9 | 7.80 | 24.8 | 19.9 |
| PWF1 | -29.177064 | -50.133022 | 36.0 | 3 | 3 | 39.2 | 7.95 | 23.9 | 20.6 |
| PWF2 | -29.181207 | -50.123423 | 35.1 | 1 | 2 | 52.5 | 7.85 | 24.5 | 20.2 |
| PWF3 | -29.170270 | -50.100430 | 23.1 | 4 | 3 | 28.7 | 7.87 | 24.4 | 20.2 |
| PWF4 | -29.164955 | -50.080631 | 24.8 | 1 | 2 | 15.7 | 7.89 | 24.2 | 20.3 |

Note: BOD, Biological oxygen demand; ECW, Electrical conductivity of water; CSV, Categories of percentage of shrub vegetation; FV, Categories of percentage of floating vegetation; PF, Sampling sites with fire; PWF, Sampling sites without fire.

Abbreviations: %, Values in percentage; AD, Average depth; AHG, Average height of grasses.

(three samplings per pond), which corresponds to the peak of the breeding season of anurans in southern Brazil (Both et al., 2008; Santos et al., 2008). We used two methods to detect anuran species: automated recording systems (Bridges & Dorcas, 2000; Hutto & Stutzman, 2009) and tadpole sampling with a dip net (Heyer et al., 1994; Vasconcelos & Rossa-Feres, 2005; Both et al., 2011). The combination of these techniques is considered one of the most effective methods in amphibian inventories (Hsu et al., 2005; Madalozzo et al., 2017; Silva, 2010).

2.4 | Calling surveys (automated recording systems)

From September to October 2016, we performed three calling surveys per pond. All ponds were sampled simultaneously. To perform calling surveys, we installed one digital audio recorder (Sony ICD-PX312/PX312F) at the margin of each pond, placed 1.5 m above the ground (Figueira et al., 2015). Recording periods went from 6 p.m. to 6 a.m. (12 h of full recording), generating a total of 288 recorded hours (3 surveys × 8 ponds × 12 recorded hours per sampling). Each 12-h audio record (one night) in a sample unit was considered a sample. We inspected the entire recorded file by extracting 5-min-long subsamples from each recorded hour. Species were identified based on audio reference guides for calling. Estimations of the abundance of calling males were made using abundance classes (adapted from Bertoluci & Rodrigues, 2002): (a) 1–4 individuals in calling activity; (b) 5–9 individuals; (c) 10–20 individuals; or (d) >20 individuals. For statistical analysis, we used the highest level recorded during one night of sampling for each sampling site.

2.5 | Tadpole sampling

We performed dip net sampling during the daytime to capture tadpoles. The dip net was moved from the border to the centre of the

pond to cover the whole area of the pond (Vasconcelos & Rossa-Feres, 2005). Captured tadpoles were immediately euthanized using a benzocaine solution and transferred to the laboratory for identification under a stereomicroscope. Identification was done with the aid of identification keys. Animal capture and handling were authorized by the federal regulatory agency and the committee of ethics in animal use and experimentation (permits #55308–1).

2.6 | Microhabitat evaluation

We performed microhabitat measurements using four quadrats of 5 × 5 m in an area surrounding the breeding site (pond). Each quadrat was placed between 30 and 40 m from the margin of the pond in the four cardinal directions. For each quadrat, we performed visual estimations of: (a) % of shrub vegetation (SV): 1 = none; 2 = 1%–25%; 3 = 26%–50%; 4 = 51%–75%; and 5 = 76%–100%; and (b) average grass cover (Table 1). We also measured the percentage of the pond area occupied by emergent vegetation according to the following classes: 1 = none; 2 = 1%–25%; 3 = 26%–50%; 4 = 51%–75%; and 5 = 76%–100% (Table 1). We also measured the mean depth (cm) and the following water chemical–physical parameters: pH, dissolved oxygen (mg/mL), and electric conductivity (ms/cm) (Table 1). Measurements were conducted using a multi-parameter water quality meter (HORIBA) and collected on each sampling day.

2.7 | Data analysis

All analyses were performed using R (R Development Core Team, 2017). The normal distribution of records and homogeneity of their variances were evaluated using the Shapiro–Wilk test and the Levene test, respectively. We used a Student *t*-test (Callegari-Jacques, 2003) to compare species richness between sample units with and without fire management. We used a non-parametric multidimensional scaling (NMDS) to evaluate the variation of anuran

community composition between treatments based on the Jaccard similarity index. This procedure creates a stress value from the original distance matrix and calculated distances. Stress values close to zero represent a better fit between the original and calculated distance matrices. This is a method that produces object ordinations from any distance matrix (Legendre & Legendre, 2012). Afterwards, variables obtained from each sample unit were added to the ordination by the envifit function. We used a Similarity Analysis (ANOSIM) (Clarke, 1993) estimated from the Jaccard similarity to evaluate differences in species composition between treatments (with and without fire). The analysis was performed using the “vegan package” (Oksanen et al., 2017). We were unable to apply a completely random sampling site distribution due to the limitation on permits to access non-public lands. Additionally, the only available areas free of fire are limited to a single locality.

We conducted a species indicator analysis in areas with and without fire management following Dufrêne and Legendre (1997). The indicative values (IndVal) generated by this analysis are based on species abundance (specificity) and frequency (fidelity). To evaluate the specificity, we used a matrix of species abundance in each sample unit based exclusively on calling records. We adapted this strategy to avoid bias in the sampling effort of species abundance from different field methods (calling estimation \times tadpole capturing). We used minimal values of individuals from each abundance category to be more conservative in abundance estimations. This analysis was performed using the Indval function of the labdsv package (Roberts, 2016). Beta diversity (β sor) and its partitioning between compounds' turnover (β sim) and nestedness (β nes) were obtained following Baselga (2010). The turnover component explains the changes in community composition by substitution between pairs of species. Nestedness indicates the loss of species between compared pairs (Baselga 2010). This analysis was performed with R software, using the function “beta.pair” from package “betapart” (Baselga 2010). Components values of turnover and nestedness from the eight sampling sites were pulled into three groups: pairs with fire management ($N=6$), pairs without fire management ($N=6$), and pairs with fire management–without fire management ($N=16$). We adopted this comparison to verify which component has a greater influence on beta diversity.

3 | RESULTS

We recorded 16 anuran species belonging to three families: Bufonidae (1 species), Hylidae (9), and Leptodactylidae (6) (Table 2). Species richness was higher in sites without fire management ($N=15$) than in those with fire management ($N=10$) ($t=-3.549$, $p=.001$) (Figure 2). Six species (*Boana faber*, *B. leptolineata*, *Dendropsophus microps*, *D. sanborni*, *Physalaemus nanus*, and *Rhinella icterica*) were recorded exclusively in sites without fire. Only *Leptodactylus gracilis* was recorded exclusively in sites with fire management. The remaining species ($N=9$) were recorded in both treatments.

The NMDS analysis (Figure 3) showed differences in species composition between sites with and without fire management (stress=0.02), which was reinforced by the analysis of similarity (ANOSIM; $R^2=.59$, $p=.03$). The envifit function also showed that the variation in species composition was related to the fire ($R^2=.59$, $p=.026$). In addition, ANOSIM showed an influence of SV on the anuran species composition ($R^2=.68$, $p=.038$). Sites without fire showed higher percentages of SV, and this was associated with the presence of *Physalaemus nanus* and *Dendropsophus microps*. This positive relationship between the absence of fire and anurans was greater in sites with higher shrub density (sample units W1 and W3). This result indicates that sites with fire present changes in both the percentage of SV and anuran community composition. The remaining variables of microhabitat did not show significant variation between treatments (average depth: $R^2=.58$; $p=.111$; flooding vegetation categories: $R^2=.15$, $p=.67$; average grass height: $R^2=.18$, $p=.591$; pH: $R^2=.21$, $p=.544$; electric conductivity: $R^2=.20$, $p=.553$; biological oxygen demand: $R^2=.20$, $p=.547$).

Pseudis cardosoi, *Scinax squalirostris*, *Leptodactylus plaumanni*, and *Dendropsophus minutus* reached the highest abundance category (>20 individuals) in at least one sample unit of each treatment. For these species, fire management does not seem to affect their abundance (Table 3).

The IndVal analysis indicated *Boana leptolineata* as an indicator species of sites without fire management (IndVal=1, $p=.028$). This species was recorded exclusively in sites without burning activities. In addition, *B. leptolineata* was recorded in all four sample units (high fidelity level), being one of the most abundant species in these sites.

Differences in beta diversity (β sor) components, turnover (β sim) and nestedness (β nes), were not statistically significant between sampling sites. There was no difference between the role of nestedness or turnover as drivers to species change either between sites within the same treatment (with fire: $t=-.4707$, $p=.328$; without fire: $t=-.9354$, $p=.196$) or between treatments (with \times without fire: $w=97$, $p=.133$). However, beta diversity values suggest different patterns in results from comparison within and between treatments. Mean turnover values within treatments were higher (β sim=.126%–56.92%) than those of nestedness (β nes=.103%–63.39%) (Figure 4). On the other hand, nestedness between treatments showed higher mean values (β nes=.202–63.09%) than turnover (β sim=.118%–36.9%), suggesting that anuran communities in sites with fire management are mostly subsets of the communities found in sites without fire.

4 | DISCUSSION

Our data indicate that fire management favors the reduction of species diversity in grassland habitats of southern Brazil. A similar scenario was also observed in the Argentinean Chaco (Cano & Leynaud 2010). Changes in species composition generated by anthropogenic fire were also recorded in North America (Wilgers & Horne, 2006) and South Africa (Masterson et al., 2008). In Brazil,

TABLE 2 ANURAN species recorded in sites with and without fire management in subtropical grasslands of southern Brazil.

| Family/Species | PF1 | PF2 | PF3 | PF4 | PWF1 | PWF2 | PWF3 | PWF4 |
|--------------------------------|-----|-----|-----|-----|------|------|------|------|
| Bufonidae | | | | | | | | |
| <i>Rhinella icterica</i> | | | | | A | | | A/T |
| Hylidae | | | | | | | | |
| <i>Dendropsophus microps</i> | | | | | A | | A | |
| <i>Dendropsophus minutus</i> | T | A/T | | A/T | A/T | A/T | A/T | A/T |
| <i>Dendropsophus sanborni</i> | | | | | A | A | A | A |
| <i>Boana faber</i> | | | | | T | | | T |
| <i>Boana leptolineata</i> | | | | | A/T | A | A/T | A |
| <i>Boana pulchella</i> | A/T | A/T | A | A/T | A | T | A | |
| <i>Pseudis cardosoi</i> | A/T | A/T | A/T | A/T | A/T | A/T | A/T | A/T |
| <i>Scinax granulatus</i> | | A | | A | A | A | A | A/T |
| <i>Scinax squalirostris</i> | A | A | A | A/T | A | A | A/T | |
| Leptodactylidae | | | | | | | | |
| <i>Leptodactylus gracilis</i> | | | A | | | | | |
| <i>Leptodactylus luctator</i> | | A | A/T | A | A | A | A/T | |
| <i>Leptodactylus plaumanni</i> | A | A | A | A | A | A | A | A |
| <i>Physalaemus carrizorum</i> | T | | T | A/T | A | A/T | A | A |
| <i>Physalaemus nanus</i> | | | | | A | | | |
| <i>Physalaemus cuvieri</i> | | | | A | | A | | A |
| Species richness (sample unit) | 5 | 7 | 6 | 9 | 14 | 11 | 11 | 10 |
| Total number of species | 10 | | | 15 | | | | |

Note: A, record of adults; PF, with fire management sample unit; T, record of tadpoles; PFW, without fire management sample unit.

Rocha et al. (2008) observed that frogs from burned sites had fewer prey items per stomach than those from unburned sites, which suggests that fire has negatively affected the species studied. In contrast, De Oliveira Drummond et al. (2018) observed an increase in anuran species richness after a fire event, which demonstrates the need for more studies in this line of research to better understand the effects of fire on Neotropical anurans. However, it is worth highlighting that the study of De Oliveira Drummond et al. (2018) was carried out in a transition zone between the Atlantic Forest and the Cerrado, a region that is affected by natural fires. Therefore, frogs from this region may be more adapted to fire than frogs from southern Brazil.

The negative effects of fire on amphibian diversity have been reported in many ecosystems (Cano & Leynaud 2010; Papp & Papp 2000; Rocha et al. 2008). Such effects include death by overheating and intoxications due to fire-emanating gases (Frizzo et al., 2011; Pilliod et al., 2003; Smith et al., 2012), as well as changes in habitat configuration and a decrease in prey availability (Allingham & Harvey, 2013; Frizzo et al., 2011; Pilliod et al., 2003; Rocha

et al., 2008). At the same time, negative impacts vary according to the ecological traits of species, driving differences in their responses to fire (Morais et al., 2011). Some species occurred only in sites without fire, such as *Rhinella icterica*, *Dendropsophus microps*, *D. sanborni*, *Boana faber*, *B. leptolineata*, and *Physalaemus nanus*. Evidence about the ability of fire to change species compositions is shared by other studies (Matthews et al., 2010; Pilliod et al., 2003; Rochester et al., 2010).

At the same time, *Pseudis cardosoi*, *Scinax squalirostris*, *Leptodactylus plaumanni*, and *Dendropsophus minutus* reached the highest abundance category (4=>20 individuals) in at least one sample unit of each treatment. Some studies found that low-intensity fire does not cause a great impact on anuran assemblages (Lemckert et al., 2004; Morais et al., 2011). Some of them argue that some species are even favored by fire regimes (Kirkland Jr et al., 1996; Klaus & Noss, 2016; Mester et al., 2015; Schurbon & Fauth, 2003). Many studies highlighted the effects of fire on the vegetation cover (Allingham & Harvey, 2013; Cano & Leynaud, 2010; Schurbon & Fauth, 2003). Apparently, arboreal

anurans are more affected by these modifications in vegetation (Friend, 1993; Papp & Papp, 2000). Our results reinforce this hypothesis since most arboreal species occurred exclusively in sites without fire (*Dendropsophus microps*, *D. sanborni*, *Boana faber*, *B. leptolineata*). Although not all species exclusive to these sites were arboreal (e.g., *Rhinella icterica*), we believe that fire generates a loss of microhabitats, including shelter from predators and extreme climatic conditions. Another non-arboreal species exclusive to non-burned sites is *Physalaemus nanus*, a terrestrial and forest-associated species (Kwet et al., 2010). We believe that its presence on sites without fire is related to the existence of a denser vegetation cover. The vegetation structure is one of the most

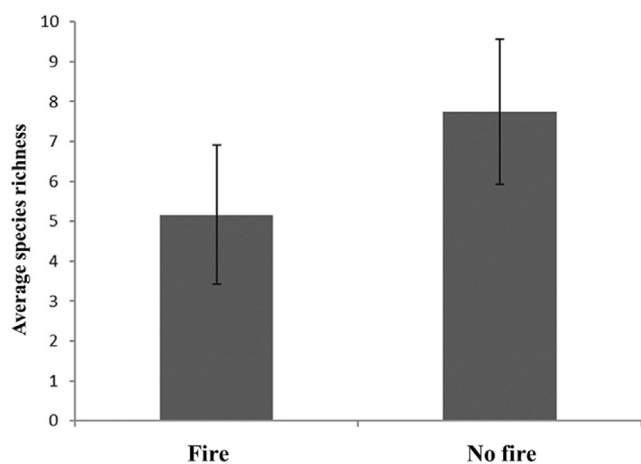


FIGURE 2 Differences in species richness (average and standard deviation) between areas with and without fire management.

representative components of the habitat related to the organization of amphibian communities (Allingham & Harvey, 2013). The presence of *Dendropsophus microps*, *D. sanborni*, and *Physalaemus nanus*, which were positively affected by the availability of shrubs, suggests that fire has an indirect effect on anurans by modifying the vegetation structure. We observed that SV and tall grass are present only in sites without fire. Many studies reported a direct relationship between vegetation height and species richness in anuran assemblages (Allingham & Harvey, 2013; Silva et al., 2012). Taller vegetation would offer more opportunities for protection and breeding activities (e.g., calling sites, oviposition sites) (Hazell et al. 2001; Bertoluci & Rodrigues, 2002; Hazell et al., 2001). Especially in grassland habitats, vegetation height is a crucial mechanism in determining microclimatic conditions. The reduction of vegetative biomass leads to rapid changes in daytime temperature, humidity, and insolation levels, characterizing a harsh habitat (Pilliod et al., 2003).

At the same time, the small leptodactylid *Leptodactylus gracilis* was exclusive to sites with fire. This species uses natural burrows or ground depressions for shelter and calling sites (Kwet et al., 2010; Maneyro et al., 2017). This could guarantee a higher probability of escaping from fire (see Raison et al., 1986). The species indicator analysis (IndVal) indicated *Boana leptolineata* as indicative of sites without fire. This species has affinities with floating vegetation and marginal vegetation of temporary and permanent ponds, which are used as calling sites (Maneyro et al., 2017; Reinke & Deiques, 2010). The low availability of this kind of microhabitat could be a limiting factor to the persistence of this species in frequently burned sites.

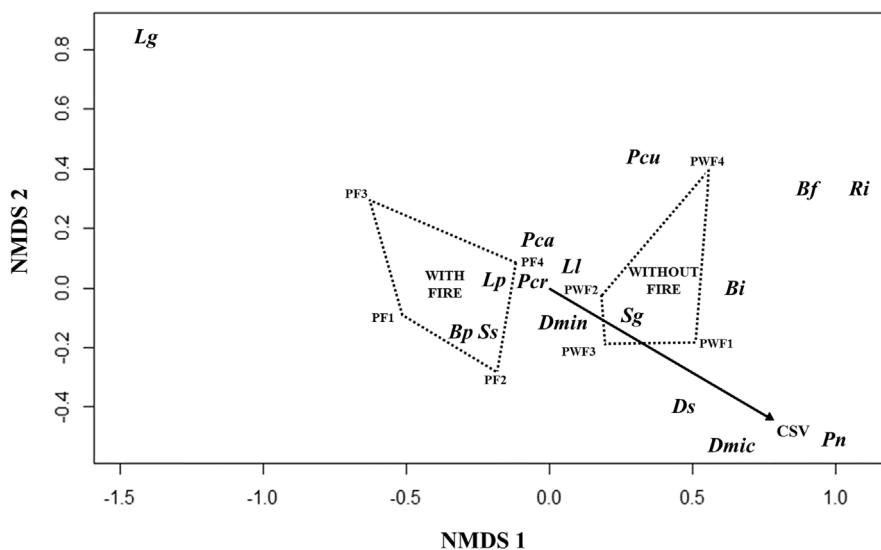


FIGURE 3 Non-metric multidimensional scaling (NMDS) considering the similarity in species composition between sites with and without fire management (Jaccard similarity). The ordination represents the two types of management (with and without fire) and the categories of the percentage of shrub vegetation (CSV) that significantly influenced the composition of species ($p < 0.05$), where: P Bf, *Boana faber*; Bl, *Boana leptolineata*; Bp, *Boana pulchella*; Dmic, *Dendropsophus microps*; Dmin, *Dendropsophus minutus*; Ds, *Dendropsophus sanborni*; Lg, *Leptodactylus gracilis*; LI, *Leptodactylus luctator*; Lp, *Leptodactylus plaumanni*; Pcr, *Pseudis cardosoi*; Pcu, *Physalaemus cuvieri*; Pca, *Physalaemus carrizorum*; Pn, *Physalaemus nanus*; Ri, *Rhinella icterica*; PF, sample unit with fire; Sg, *Scinax granulatus*; Ss, *Scinax squalirostris*; PWF, sample unit without fire.

TABLE 3 Abundance categories of species calling in sample units located in areas with and without fire management in subtropical grasslands in southern Brazil.

| Species | PF1 | PF2 | PF3 | PF4 | PFW1 | PFW2 | PFW3 | PFW4 |
|--------------------------------|-----|-----|-----|-----|------|------|------|------|
| <i>Dendropsophus microps</i> | | | | | | | | |
| <i>Dendropsophus minutus</i> | | | | | | | | |
| <i>Dendropsophus sanborni</i> | | | | | | | | |
| <i>Boana leptolineata</i> | | | | | | | | |
| <i>Boana pulchella</i> | | | | | | | | |
| <i>Leptodactylus gracilis</i> | | | | | | | | |
| <i>Leptodactylus luctator</i> | | | | | | | | |
| <i>Leptodactylus plaumanni</i> | | | | | | | | |
| <i>Physalaemus carrizorum</i> | | | | | | | | |
| <i>Physalaemus nanus</i> | | | | | | | | |
| <i>Physalaemus cuvieri</i> | | | | | | | | |
| <i>Pseudis cardosoi</i> | | | | | | | | |
| <i>Rhinella icterica</i> | | | | | | | | |
| <i>Scinax granulatus</i> | | | | | | | | |
| <i>Scinax squalirostris</i> | | | | | | | | |
| Number of calling species | 4 | 7 | 5 | 9 | 13 | 10 | 11 | 7 |

Note: Mean number of individuals calling:

| | | | |
|-----|-----|-------|-----|
| 1–4 | 5–9 | 10–20 | >20 |
|-----|-----|-------|-----|

PF, with fire management sample unit; PFW, without fire management sample unit.

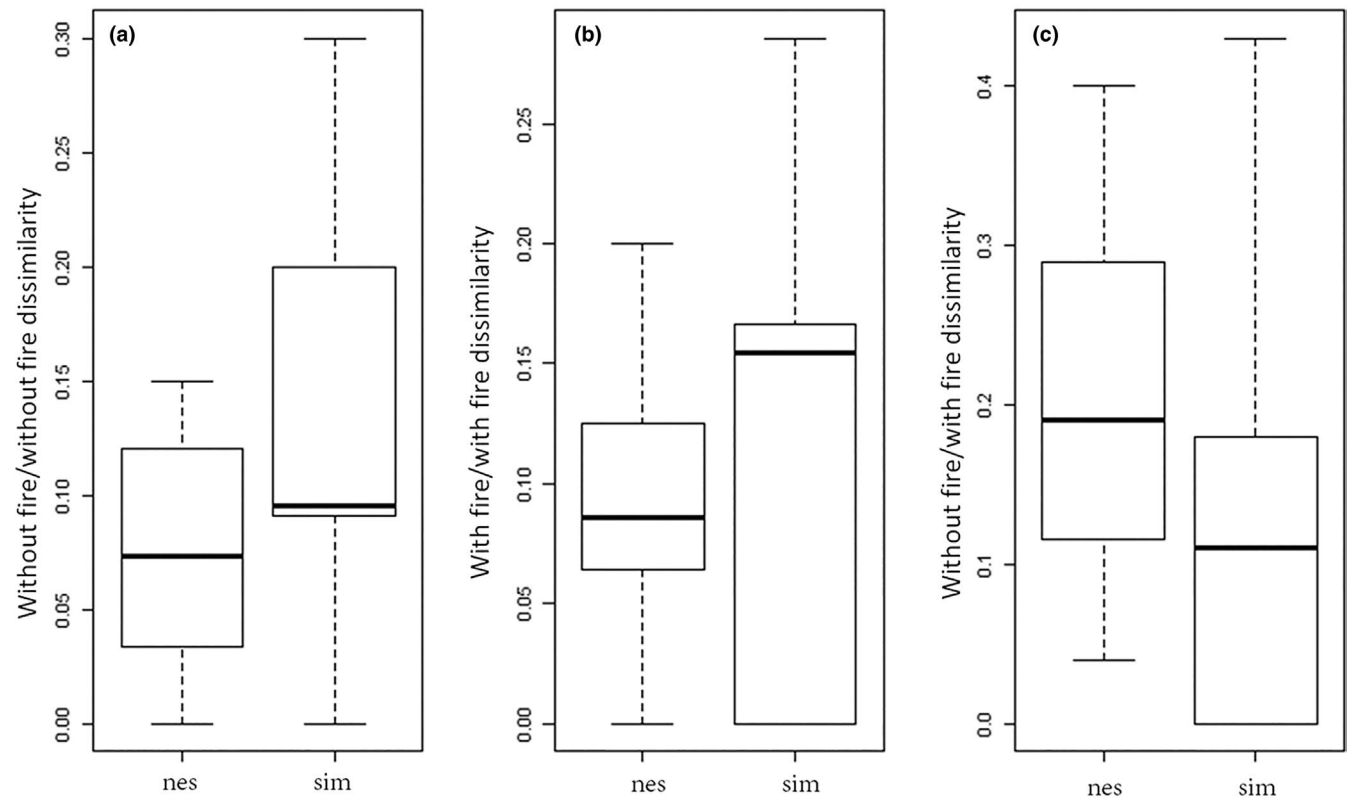


FIGURE 4 Relative contribution of species turnover (β -Simpson) and nestedness (β -nestedness) for total beta-diversity in the three groups of pairwise comparisons: (a) without fire management–without fire management (6 pairs), (b) with fire management–with fire management (6 pairs), (c) without fire management–with fire management (16 pairs) in subtropical grasslands of southern Brazil, where nes, Nestedness and sim, Turnover.

The beta diversity components revealed no difference between treatments. However, it was possible to observe a trend for a nested organization of the community in burned sites, suggesting that sites with fire are a subsample of sites where fire is absent. In a study carried out in the same region, but in forest fragments within a grassland matrix, a pattern of species turnover (i.e., exchange) was observed (de Oliveira Drummond et al., 2018). These results may be related to the origin of the studied environment, which is characterized by forest expansion over the grassland matrix (Behling et al. 2004). We must reinforce that the negative effects of fire on anurans could be a complex issue. More than death by burning, fire causes habitat changes that can harm anurans. For example, in the same area as our study, Schuck et al. (2024) observed that fire was associated with changes in the bacteria that inhabit the skin of frogs, in addition to an increase in pathogen loads.

4.1 | Conclusions

This study sheds some light on the impacts of fires on such sensitive animals. Our results are worrying since the effects of fire on anurans are poorly studied in comparison with other vertebrates (Pastro et al., 2014) and need to be investigated in many more species and habitats. Sites without the presence of fire developed taller vegetation and a higher density of shrubs, which could be viewed as an improvement in habitat complexity. Therefore, we hypothesize that this increase in complexity generates an increase in microhabitat opportunities, favoring a higher species richness in comparison with constantly burned sites. This is the first study evaluating the effects of fire management on anurans in Brazilian subtropical grasslands. Thus, we suggest that fire management should not only consider the impacts on the fauna but also allow the maintenance of tall grass and shrubs around ponds.

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DATA AVAILABILITY STATEMENT

The data that support the findings of this study are openly available in the Dryad Digital Repository: <https://doi.org/10.5061/dryad.jwstajqkw>.

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SUPPORTING INFORMATION

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