

UNIVERSIDADE ESTADUAL PAULISTA "JÚLIO DE MESQUITA FILHO" INSTITUTO DE BIOCIÊNCIAS – RIO CLARO



# PROGRAMA DE PÓS-GRADUAÇÃO EM CIÊNCIAS BIOLÓGICAS (BIOLOGIA VEGETAL)

## Avaliação de diferentes técnicas de manejo para o controle de gramíneas invasoras em unidade de conservação de Cerrado

# **KAREN PATRÍCIA CASTILLIONI**

Dissertação apresentada ao Instituto de Biociências do Câmpus de Rio Claro, Universidade Estadual Paulista, como parte dos requisitos para obtenção do título de Mestre em Ciências Biológicas (Biologia Vegetal).

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#### CERTIFICADO DE APROVAÇÃO

TÍTULO: Avaliação de diferentes técnicas de manejo para o controle de gramineas invasoras em unidade de conservação de Cerrado

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"Mas na profissão, além de amar tem de saber. E o saber leva tempo pra crescer."

Rubem Alves

#### **RESUMO GERAL**

As invasões biológicas são consideradas uma das maiores ameaças à biodiversidade global. Agravado pelas atividades humanas, o problema das invasões por gramíneas africanas já atinge grande parte do Cerrado. A fim de minimizar os danos causados pela invasão, faz-se necessária a avaliação de técnicas de manejo em ecossistemas invadidos. Neste sentido, este estudo teve como objetivo principal avaliar a eficiência de diferentes técnicas de manejo para o controle de Melinis minutiflora e Urochloa decumbens em áreas de Cerrado em regeneração, na Estação Experimental de Itirapina-SP (EEI), assim como a resposta da vegetação nativa a esses tratamentos. Além disso, teve-se como objetivo identificar o manejo mais viável em termos de benefício/custo na área de estudo, ou seja, o tratamento capaz de promover a maior redução de biomassa total por unidade monetária. Os seguintes tratamentos foram testados na área de estudo: aplicação do herbicida glifosato (He), Corte (Ct), combinação do herbicida glifosato seguido de corte (He+Ct), e controle (Co). Os tratamentos foram aplicados em parcelas de 4x4m (6 réplicas/tratamento - total de 24 parcelas para Melinis minutiflora e 3 réplicas/tratamento – total de 12 parcelas para Urochloa decumbens). Em cada parcela, foi realizado o levantamento da cobertura vegetal (%) nas categorias: gramínea invasora viva (Melinis minutiflora e Urochloa decumbens), graminóide nativa (exceto Poaceae), herbácea nativa (exceto graminóides e Poaceae), arbusto nativo ( $\leq 1,5$  m), além da biomassa morta total (nativa e invasora) e solo nu em oito subparcelas de 1x1m. A biomassa aérea também foi coletada dentro de cada unidade amostral, utilizando-se uma subparcela de 0,5x0,5m. A biomassa foi então separada em laboratório nas seguintes categorias: gramínea invasora morta e viva (Melinis minutiflora e Urochloa decumbens), graminóide nativa (exceto Poaceae), herbácea nativa (exceto graminóides e Poaceae), arbusto nativo e biomassa nativa morta. Após a triagem, o material foi levado para secagem em estufa a 70°C durante 3 dias e depois pesado. Os levantamentos foram realizados antes da aplicação dos tratamentos e depois a cada quatro meses (total de 12 meses). Para se testar a diferença das variáveis avaliadas entre os tratamentos, foram realizadas análises de variância de um fator (tratamento) em cada tempo de observação para cada espécie separadamente. Em curto prazo, ambos os tratamentos de He e He + Ct foram os mais eficazes no controle das duas espécies invasoras. Entretanto, as herbáceas foram o grupo funcional nativo mais negativamente afetado pela aplicação do herbicida. Além disso, o acúmulo de biomassa morta também promovido por este tratamento foi capaz de afetar a estrutura e dinâmica da vegetação. Por outro lado, Ct estimulou a regeneração das espécies invasoras. Portanto, em curto prazo, para o controle de *Melinis minutiflora* e *Urochloa decumbens* sugere-se o uso de He + Ct. Porém é importante considerar que este tratamento também afetou a estrutura da vegetação, ao aumentar a cobertura de solo nu, o que pode facilitar a regeneração de espécies nativas e de exóticas. Além da avaliação das técnicas de manejo, a análise de seu benefício/custo mostrou que os tratamentos testados apresentam a mesma eficácia em pequena escala (1 m<sup>2</sup> e 16 m<sup>2</sup>), no entanto, em áreas maiores (5000 m<sup>2</sup> e 10000 m<sup>2</sup>), He + Ct para *Melinis minutiflora* e *Urochloa decumbens* foi o tratamento mais viável. Em curto prazo, os resultados reforçam a necessidade de uma avaliação de eficiência econômica ao planejar o controle de espécies invasoras para que os recursos para conservação sejam utilizados de forma mais eficaz.

Palavras-chave: Glifosato. Invasão biológica. Análise benefício/custo. *Melinis minutiflora*. *Urochloa decumbens*.

#### ABSTRACT

Biological invasion is considered a major threat to global biodiversity. Aggravated by human activities, the problem of invasions by African grasses has already reached most part of Cerrado. In order to minimize the damage caused by the invasion, it is essential that management techniques are assessed in invaded ecosystems. In this sense, this study aimed to evaluate the efficiency of different management techniques to the control of Melinis minutiflora and Urochloa decumbens in Cerrado areas under natural regeneration at the Experimental Station of Itirapina-SP (ESI), as well as the response of the native plant community to the treatments. In addition, it was analyzed the benefit/cost ratio of each technique in the study area, i.e., the treatment able to lead to the greatest reduction of the invasive species total biomass by monetary unit. The following treatments were tested: Cut (Ct), Herbicide application (He): glyphosate, combination of Herbicide (glyphosate) followed by Cut (He + Ct) and control (Co) (no intervention). The treatments were applied in 4x4m plots (6 replicates/treatment- total of 24 plots for Melinis minutiflora and 3 replicates/treatment- total of 12 plots for Urochloa decumbens). In each plot, we performed the survey of the vegetation cover (%) in the categories: live invasive grass (Melinis minutiflora and Urochloa decumbens), native graminoids, native forbs, native shrubs ( $\leq 1.5$ m), total dead biomass and bare soil in eight subplots of 1x1m. All aboveground biomass was also collected within each sample unit by using a subplot of 0.5x0.5m. Biomass was separated in the laboratory in the following categories: dead and live invasive grass (Melinis minutiflora and Urochloa decumbens), native graminoids, native forbs, native shrubs and dead biomass, then dried (at 70°C for 3 days) and weighed. The surveys were carried out before treatment application and then every four months (total 12 months). To test the difference of measured variables among treatments, analysis of variance of one factor (treatment) were performed at each observation period for each species separately. In the short term, He and He + Ct were the most effective treatments to control both invasive species. However, forbs were the native functional group most negatively affected by herbicide application. Moreover, dead biomass accumulation was also influenced by this treatment, which was able to affect the structure and dynamics of the vegetation. On the other hand, Ct enhanced invasive species regeneration. Thus, He + Ct would be the treatment suggested, considering it resulted in Melinis minutiflora and Urochloa decumbens control, but increased bare soil cover which may facilitate native and exotic species regeneration. In addition to the evaluation of the management techniques, the benefit/cost ratio showed that the treatments tested presented the same efficacy in small

scale (1  $\text{m}^2$  and 16  $\text{m}^2$ ), however, in large areas (5000  $\text{m}^2$  and 10000  $\text{m}^2$ ), He + Ct for *Melinis minutiflora* and *Urochloa decumbens* showed to be most viable treatment. In the short term, the results reinforce the need for an assessment of economic efficiency when planning the control of invasive species in order to use more effectively the conservation funds.

**Keywords:** Glyphosate. Biological invasion. Benefit/cost analysis. *Melinis minutiflora*. *Urochloa decumbens*.

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#### INTRODUÇÃO GERAL

É amplamente reconhecido que todas as partes do mundo têm sido afetadas pela introdução de espécies exóticas, desde o continente com mais alta biodiversidade até ilhas mais isoladas (CASSEY *et al.*, 2005). Como consequência, as invasões biológicas têm sido consideradas uma das principais ameaças à biodiversidade (DUKES; MOONEY, 1999), capazes de levar à extinção as espécies nativas (D'ANTONIO; VITOUSEK, 1992) e desafiar a conservação da biodiversidade e dos recursos naturais (SIMBERLOFF *et al.*, 2013). Por esta razão, a invasão biológica tem sido assunto de eminente importância e objeto de estudo de muitos autores (REJMÁNEK, 2000; RICHARDSON *et al.*, 2000a; PYŠEK *et al.*, 2004), sendo um consenso de que as invasões são indesejáveis para a manutenção da função de padrões e processos ecológicos e evolutivos (CASSEY *et al.*, 2005).

O processo da invasão biológica é o resultado da ampla dispersão e expansão de uma espécie exótica, introduzida de maneira acidental ou intencional por atividades humanas (PYŠEK, 1995; RICHARDSON *et al.*, 2000a). Estas espécies se dispersam do local de sua introdução, integram-se nas comunidades nativas, afetando sua dinâmica, e, em muitos casos, competindo e eliminando as espécies nativas (RICHARDSON *et al.*, 2000b). Como consequência, as espécies invasoras também modificam alguns aspectos da estrutura e funcionamento de ecossistemas invadidos (VILÀ *et al.*, 2011), alterando a composição e estrutura da comunidade invadida, assim como das propriedades do ecossistema, como ciclagem de nutrientes, produtividade ou frequência de distúrbios (VITOUSEK, 1990).

Gramíneas invasoras apresentam, no geral, um padrão de rápido crescimento e grande produção de biomassa, tornando-se potenciais competidoras com as espécies nativas, modificando severamente o ambiente invadido (D'ANTONIO; VITOUSEK, 1992). Sua presença pode levar a uma redução na absorção de luz pelas espécies nativas, limitando, principalmente, o estabelecimento de plântulas (D'ANTONIO; VITOUSEK, 1992; CABIN *et al.*, 2002). Além disso, o regime de fogo pode ser alterado, pois as gramíneas invasoras podem tornar o sistema mais inflamável (D'ANTONIO; VITOUSEK, 1992), como mostrado por Rossiter *et al.* (2013) para savanas australianas: a presença da gramínea africana *Andropogon gayanus* Kunth. aumentou a intensidade e frequência do fogo, causando a mortalidade de árvores e reduzindo seu recrutamento (ROSSITER *et al.*, 2003). No Cerrado, a altura das chamas e as temperaturas do fogo foram mais altas em áreas onde *Urochloa* 

*brizantha* (A. Rich.) R.D. Webster (gramínea africana invasora) estava presente, o que pode afetar a sobrevivência principalmente de arbóreas (Gorgone-Barbosa *et al.* 2015).

Nas Américas, *Melinis minutiflora* Beauv., *Hyparrhenia rufa* (Nees) Stapf., *Panicum maximum* Jacq., *Brachiaria mutica* (Forsk.) Stapf. e *Pennisetum clandestinum* Hochst. foram as primeiras espécies introduzidas (gramíneas africanas C<sub>4</sub>), enquanto que, mais recentemente, *Andropogon gayanus* Kunth, *Pennisetum purpureum* Schumach., *Chloris gayana* Kunth, *Digitaria decumbens* Stent, e muitas espécies de *Brachiaria (Urochloa) (U. decumbens, U. humidicola, U. brizantha e U. dictyoneura)* passaram a ser utilizadas como pastagem em áreas de savanas tropicais e regiões desflorestadas (WILLIAMS; BARUCH, 2000). No entanto, estas gramíneas se tornaram invasoras, promovendo uma drástica mudança na vegetação nativa (WILLIAMS; BARUCH, 2000). No Brasil, *Melinis minutiflora* Beauv. (Figura 1) teve seus primeiros registros descritos em 1812 (PARSONS, 1972), enquanto *Urochloa decumbens* Staf. (Figura 2), na década de 1950 (LORENZI, 2008) e são atualmente algumas das espécies que mais comumente invadem o Cerrado (PIVELLO; SHIDA; MEIRELLES, 1999b).

Figura 1. Invasão por *Melinis minutiflora* em uma área de Cerrado em regeneração natural na Estação Experimental de Itirapina-SP.



Figura 2. Invasão por *Urochloa decumbens* em uma área de Cerrado em regeneração natural na Estação Experimental de Itirapina-SP.



As espécies *Melinis minutiflora* ("capim-gordura") e *Urochloa decumbens* ("braquiária") passaram a afetar negativamente espécies de gramíneas nativas, exercendo uma forte pressão competitiva sobre elas (PIVELLO; SHIDA; MEIRELLES, 1999b). Além disso, as gramíneas africanas também são capazes de substituir algumas espécies herbáceas, influenciando assim toda a comunidade vegetal (PIVELLO *et al.*, 1999a). Atualmente, a presença destas espécies é considerada uma das maiores ameaças à conservação do Cerrado (PIVELLO; SHIDA; MEIRELLES, 1999b; DURIGAN *et al.*, 2007), pois podem produzir e acumular uma grande quantidade de biomassa (MARTINS; LEITE; HARIDASAN, 2004). Em áreas invadidas por *Melinis minutiflora*, há um aumento no sombreamento que afeta negativamente o estabelecimento e sobrevivência de espécies de árvores nativas (HOFFMANN; HARIDASAN, 2008).

Deste modo, devido aos impactos negativos que as espécies invasoras podem causar, elas são motivo de preocupação para a conservação da biodiversidade (VILÀ *et al.*, 2011). Gestores de áreas de conservação precisam estabelecer prioridades para que populações de espécies invasoras sejam reduzidas (USHER, 1988). Neste sentido, o uso de técnicas de manejo visa minimizar o processo de invasão em uma área ou até mesmo reduzir sua probabilidade de sucesso (DAVIS, 2009). Ações mecanizadas ou manuais (Figura 3) como o arranquio (GROVES, 1989), abafamento ou duplo corte raso (GORGONE-BARBOSA, 2009) e até mesmo o fogo podem ser usados como ferramentas para o controle de invasoras (CLOUT; WILLIAMS, 2009). Outra opção é o uso de controle químico (Figura 4), caracterizado pela ação sobre enzimas envolvidas na biossíntese de aminoácidos, na inibição da fotossíntese, na síntese de lipídios e na mitose (DUKE, 1990).

Figura 3. Manejo mecânico com roçada (corte raso) em (a) *Melinis minutiflora* e (b) *Urochloa decumbens*, em uma área de Cerrado em regeneração natural na Estação Experimental de Itirapina-SP.



Figura 4. Manejo químico (aplicação de herbicida) em (a) *Melinis minutiflora* e (b) *Urochloa decumbens*, em uma área de Cerrado em regeneração natural na Estação Experimental de Itirapina-SP.



Um dos principais herbicidas utilizados é o glifosato (N-(fosfonometil) glicina) (BAYLIS, 2000; KETTENRING; ADAMS, 2011). Ele é classificado como não seletivo, de ação sistêmica e de amplo espectro de ação (GALLI; MONTEZUMA, 2005). A utilização do glifosato provou ser eficiente no controle de espécies invasoras em áreas agriculturáveis (HARMONEY; STAHLMAN; HICKMAN, 2004; BOTTOMS *et al.*, 2011) e até mesmo em áreas naturais em processo de restauração (BELL; EASLEY; GOODMAN, 2008). Por outro lado, a sua utilização pode impactar negativamente a vegetação nativa (SULLIVAN; SULLIVAN, 2003). Por exemplo, uma única aplicação deste herbicida foi capaz de matar plântulas e indivíduos jovens de arbustos nativos (*Pimelea spicata* R. Br.) na Austrália

(MATARCZYK *et al.*, 2002) e também promoveu uma redução na riqueza de espécies do banco de sementes no Pampa (campos temperados) na Argentina (RODRIGUEZ; JACOBO, 2012).

No Brasil, a aplicação do herbicida é permitida em unidades de conservação que contemplem seu uso em seu plano de manejo (INSTRUÇÃO NORMATIVA IBAMA Nº7, de 02 de julho de 2012). No Parque Nacional de Brasília (DF), o manejo de *Melinis minutiflora* com uso de herbicida, arranquio e fogo reduziu a cobertura da invasora, favorecendo assim, a expansão da vegetação nativa (MARTINS *et al.*, 2011).

Deste modo, estudos que avaliem os efeitos do herbicida, não apenas no controle das espécies invasoras, como também na dinâmica e estrutura da vegetação nativa são de extrema importância, principalmente para os tomadores de decisão de unidades de conservação de Cerrado que convivem com este grande problema e necessitam de uma solução viável.

Outro motivo de preocupação na escolha da técnica de manejo mais eficiente em unidades de conservação é o seu custo (USHER, 1988). Bilhões de dólares já foram gastos com os impactos de plantas invasoras (PYŠEK; RICHARDSON, 2010) e gestores de unidades de conservação podem enfrentar dificuldades no controle destas espécies devido a limitações orçamentárias (ANDREU; VILÀ; HULME, 2009). Assim, decisões têm sido tomadas de forma arbitrária ou baseadas nos recursos disponíveis para o controle de invasoras (EPANCHIN-NIELL; HASTINGS, 2010).

Grande parte dos estudos sobre cálculo de custos para o manejo de gramíneas invasoras ainda é voltada para a agricultura, devido aos prejuízos econômicos que estas espécies podem ocasionar (SNIPES *et al.*, 1984; JUDICE *et al.*, 2006). Apesar das implicações orçamentárias potencialmente significativas da pesquisa, muitos estudos desconsideram os custos do controle de espécies invasoras (KETTENRING; ADAMS, 2011) e poucos têm medido a efetividade do custo de seu manejo em áreas naturais (por exemplo, DEHNEN-SCHMUTZ; PERRINGS; WILLIAMSON, 2004; GORGONE-BARBOSA, 2009; MCCONNACHIE *et al.*, 2012). Portanto, além da avaliação do efeito da técnica de manejo utilizada no controle das invasoras, estudos sobre o custo da aplicação de tal manejo são essenciais para subsidiar ações dos tomadores de decisão em áreas protegidas.

Dada a importância que informações sobre o manejo de espécies invasoras representam para gestores de áreas de conservação e tomadores de decisões, este trabalho tem como objetivo fornecer dados que subsidiem o uso de técnicas de manejo para o controle de plantas invasoras em áreas de conservação. Assim sendo, propõe:

- <u>Capítulo 1</u>: Verificar o efeito de diferentes técnicas de manejo para o controle de gramíneas invasoras em áreas de Cerrado e, ao mesmo tempo, verificar como tais técnicas afetam a vegetação nativa. Tem como objetivos específicos, em curto prazo (1 ano):
  - 1.1. Avaliar se os diferentes tratamentos (corte, aplicação de herbicida e, aplicação do herbicida seguido de corte) afetam a cobertura e biomassa das gramíneas invasoras, *Melinis minutiflora* e *Urochloa decumbens* em áreas de Cerrado em regeneração natural.
  - 1.2. Verificar como as técnicas acima citadas influenciam a biomassa e cobertura de diferentes grupos funcionais (graminóides, herbáceas e arbustos) da comunidade vegetal nativa.
  - 1.3. Examinar como as técnicas de manejo aplicadas afetam a estrutura da comunidade vegetal.
- <u>Capítulo 2</u>: Analisar a relação de benefício/custo dos diferentes tratamentos testados para o controle das gramíneas invasoras. A análise considerou o tempo de um ano de manejo em áreas de áreas de 1 m<sup>2</sup>, 16 m<sup>2</sup>, 5000 m<sup>2</sup> e 10000 m<sup>2</sup>. A relação de benefício/custo foi obtida através da quantidade de biomassa da invasora total viva e morta (em quilogramas) que é reduzida por unidade monetária (dólar).

O estudo foi realizado na Estação Experimental de Itirapina-SP (Figura 5), na Zona de Manejo Florestal, uma área de Cerrado em regeneração natural desde 1999, após a remoção de uma plantação de *Pinus oocarpa* (Figura 6). Atualmente, a área de estudo encontra-se altamente invadida pelas gramíneas *Melinis minutiflora* e *Urochloa decumbens*.

Figura 5. Mapa da localização de Itirapina-SP, com destaque para a Estação Experimental de Itirapina (EEI - no tracejado). (Adaptado do Plano de manejo integrado das unidades de Itirapina-SP, ZANCHETTA *et al.*, 2006).



Figura 6. Local de estudo (Zona de Manejo Florestal) em uma área de Cerrado em regeneração natural na Estação Experimental de Itirapina-SP.



Assim, a dissertação foi organizada em dois capítulos elaborados sob a forma de artigos científicos preparados para publicação. O primeiro capítulo será submetido para a revista *Forest Ecology and Management* e o segundo para *Natureza e Conservação – (Brazilian Journal of Nature Conservation)*.

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# CAPÍTULO I

# Evaluation of different management techniques to control African invasive grasses in Cerrado natural reserves

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#### Abstract

Biological invasion is a threat to global biodiversity. Intensified by human activities, African grasses have already reached most part of Cerrado. In order to minimize the damage caused by invasion, it is essential that management techniques be implemented in invaded ecosystems. In this study, we evaluated the efficiency of different management techniques to control Melinis minutiflora and Urochloa decumbens in a Cerrado area under regeneration. Moreover, we analyzed the effects of the treatments on the native vegetation. We tested the following treatments: cut, herbicide (glyphosate), herbicide (glyphosate) + cut, and control (no intervention). Plots (4x4m) were established in the area where patches of invasive grasses could be observed (6 replicates/treatment - total of 24 plots for Melinis minutiflora and 3 replicates/treatment - total of 12 plots for Urochloa decumbens). In each plot, vegetation cover (%) was estimated in the following categories: live invasive grass (Melinis minutiflora and Urochloa decumbens), native graminoid, native forb, native shrub ( $\leq 1.5$  m), total dead biomass (native + invasive) and bare soil (8 subplots of 1x1m). All aboveground biomass was sampled within each sample unit, using a subplot of 0.5x0.5m. After that, biomass was separated into: live and dead invasive grass (Melinis minutiflora and Urochloa decumbens), native graminoid, native forb, native shrub and dead native biomass. The biomass was ovendried (70 °C for 3 days) and weighted. Vegetation surveys were carried out before treatment application and then every four months (total of 12 months). To test the differences of the measured variables among treatments, one-way analyses of variance (factor: treatment) were performed at each time of observation and for each species. In the short-term, herbicide and herbicide + cut were the most effective treatments to control both invasive species. On the other hand, forb was the native functional group mostly negatively affected by herbicide application. Moreover, the accumulation of dead biomass in herbicide plots affected vegetation structure and dynamics in the area. Therefore, in the short-term, herbicide + cut would be the recommend technique, considering that it effectively controlled both invasive species and did not result in biomass accumulation, although it provided more bare soil that may facilitate not only native, but also invasive species regeneration.

Key-words: Glyphosate, biological invasion, *Melinis minutiflora*, *Urochloa decumbens*, tropical savanna.

#### 1. Introduction

Biological invasions are a threat to biodiversity (Dukes and Mooney, 1999), often causing the extinction of native species (D'Antonio and Vitousek, 1992). Plants introduced by humans can establish and become widely spread and invade disturbed, natural or seminatural communities (Pyšek, 1995; Richardson *et al.*, 2000). Grass invasions can negatively affect native plant community (D'Antonio and Vitousek, 1992). The presence of invasive grasses can alter the microclimate (D'Antonio and Vitousek, 1992). Light absorption by native woody species is reduced, affecting species growth and seedling recruitment (D'Antonio *et al.*, 1998; Cabin *et al.*, 2002, Hoffmann and Haridasan, 2008). In areas dominated by invasive grasses, native plant communities biomass and diversity are reduced (Flory and Clay, 2009). Moreover, invasive grasses can modify fire regimes by altering fuel load quantity, resulting thus in changes in fire behavior (D'Antonio and Vitousek, 1992, MacGranahan *et al.*, 2013, Gorgone-Barbosa *et al.*, 2015), which could lead to higher mortality of shrubs and trees and as a consequence, to drastic changes in plant community structure (Gorgone-Barbosa *et al.*, 2015).

*Melinis minutiflora* and *Urochloa decumbens* are some of the African grasses dominant in most parts of Cerrado (Pivello *et al.*, 1999a, 1999b). In Brazil, these species were introduced for cattle grazing (Lorenzi, 2008), considering that *M. minutiflora* was first documented in 1812 (Parsons, 1972), whilst *U. decumbens*, in the 1950's (Lorenzi, 2008). Both species are able to accumulate a large amount of aboveground biomass (Lannes *et al.*, 2012) and consequently, promote a strong competitive pressure on the native herbaceous community (Pivello *et al.*, 1999b). In some invaded areas of Cerrado, *M. minutiflora* represented 60% of the total biomass of the ground layer (Martins *et al.*, 2011). Where *M. minutiflora* is dominant, its biomass is negatively correlated with the survival of native seedlings due to competition for light (Hoffmann and Haridasan, 2008). Thus, in Cerrado, the presence of these invasive species is considered one of the major threats to its conservation (Durigan *et al.* 2007). Since Cerrado is considered one of the hotspots of biodiversity (Myers *et al.*, 2000), efforts for its conservation should be implemented and hence, the control of invasive grasses should be a priority.

The ecological impacts of invasive plants reinforce the need to control these species (D'Antonio *et al.*, 2004) in order to preserve natural systems and to reestablish biodiversity and native species function in the long-term (Hulme, 2006). In this sense, field experiments with the grass *Microstegium vimineum*, invasive in forests of the eastern United States, have shown positive results by the reduction of its biomass and regeneration of the native community cover by applying hand-weeding (Flory, 2010). Clipping was another control technique that reduced invasive grass cover by 90% and doubled the cover of native species (Wilson and Partel, 2003). Although the eradication of the perennial grass *Agropyron cristatum* in the prairie in Canada was not achieved, clipping allowed the coexistence of native species with the invasive grass (Wilson and Partel, 2003). Herbicide application is another option, which is encouraged by a wide range of products with fast visible results (Luken and Seastedt, 2004). Its efficacy has been demonstrated by the control of *Bothriochloa* and *Dichanthium* spp. in Texas coastal prairie (Ruffner and Barnes, 2010). Moreover, combining herbicide and mowing showed to effectively control *Bothriochloa ischaemum* in the prairie in South-Central USA (Robertson *et al.*, 2013).

Most studies focus on the application of different techniques to control invasive species, but native community responses to the treatments are usually overlooked ). Although studies have reported the role of herbicide as being an effective tool in controlling invasive plants for restoration (Bell *et al.*, 2008) or in natural grasslands (Ruffner and Barnes, 2010), its use remains questionable and should be evaluated in different systems. This is because the use of herbicide reduced both native forb density and cover in grasslands in the Great Plains
(Sheley and Denny, 2006). Moreover, it was reported that glyphosate reduced richness and diversity of native seed bank in semi-natural habitats in the Pampas grassland. As a consequence, the regeneration of native vegetation would mainly depend on the arrival of propagules from adjacent areas (Rodriguez and Jacobo, 2012).

Therefore, considering that management of invasive species must be incorporated in management plans of natural reserves, it is important to assess the effects and impacts of the treatments on both invasive species and native plant communities. Thus, we examined the effects of different management techniques (cut, herbicide, herbicide and cut, control) to control the invasive grasses *Melinis minutiflora* and *Urochloa decumbens* in areas of Cerrado in regeneration. In addition, we aimed to evaluate how these techniques affect the structure, cover and biomass of the native plant community. We hypothesized that glyphosate would be more effective in controlling invasive grasses, but it would affect negatively the native plant community, mostly graminoids, by reducing their biomass and cover compared to the other treatments.

#### 2. Material and methods

#### 2.1. Study area and species

The study was carried out in a Cerrado area under natural regeneration since 1999, after the removal of pine plantation (*Pinus oocorpa*). The study area is at the Experimental Station of Itirapina (ESI), Southeast Brazil (22° 15' a 22° 15' S, - 47° 45' e 47° 51' W, 710 - 830 m a.s.l.). The ESI is covered by *Pinus* spp. and *Eucalyptus* spp plantations, and fragments of native vegetation of cerrado and riparian forests (Zanchetta *et al.*, 2006). The climate is mesothermal, characterized by a rainy season from October to March, and a marked dry season from April to September. The average annual rainfall is 1459 mm and the average

annual temperature is 21.9 °C (Zanchetta *et al.*, 2006). The study area is highly invaded by two African grasses: *Urochloa decumbens* (Stapf) RD Webster and *Melinis minutiflora* Beauv. (Poaceae), both C<sub>4</sub> perennial grasses (Klink and Joly, 1989). *Urochloa decumbens* persists in low fertility soils, forming a large amount of biomass (Jakelaitis *et al.*, 2004). It propagates by seeds and rhizomes (Lorenzi, 2008). *Melinis minutiflora* is a forage grass of rapid growth (Lorenzi, 2008), introduced in Brazil for cattle grazing (Lorenzi, 2008). In Cerrado, these species found favorable climate and soil conditions, spreading and competing with the native vegetation, displacing species from the herbaceous layer (Pivello *et al.*, 1999a).

# 1.2. Experimental design

We established plots of 4x4 m (6 replicates/treatment=24 plots for *Melinis minutiflora* and 3 replicates/treatment=12 plots for *Urochloa decumbens*) in the study area, in patches where the invaded species could be found. Each plot had initially at least  $\geq$ 50% cover of each invasive grass (live and dead). The four treatments were applied in June/2013 for *Melinis minutiflora* and in October/2013 for *Urochloa decumbents*. The application date was performed during the flowering time of each species. Treatments were randomly established among the replicates, with the criteria that the plots with herbicide application were at least 2 m away from the control and cut plots in order to avoid any influence of the herbicide on the plants.

The treatments applied were: 1) Cut (Ct): all above ground biomass was cut by using a weed whacker and removed; 2) Herbicide (He): glyphosate application by spraying only the tussocks of the invasive grasses, and reapplication after six months; 3) Herbicide + Cut (He + Ct): application of glyphosate as described above and removal of the above ground

biomass seven days after herbicide application; and **4**) **Control (Co)**: no treatment application.

The herbicide used was Roundup<sup>®</sup> (480 g/l of Glyphosate Isopropylamine Salt and 360 g/l of the equivalent acid of N-(phosphonomethyl)glycine (Glyphosate)). A non-ionic adhesive spreader Silwet L-77 AG (Heptamethyltrisiloxane modified with methyl ether aliloxi polyethylene glycol- polyether copolymer and silicone -1.000 g/L) was added to assure complete and uniform deposition of the herbicide. Moreover, blue indicator dye was used to mark the tussocks that were spread with the herbicide. The amount of glyphosate and the above-mentioned products were applied as suggested by the label: 1.5% of glyphosate, 0.05% of the herbicide adjuvant and 0.8% of indicator dye.

# 1.3. Vegetation survey and biomass sampling

In each plot, a grid of 1x1m (total of 16 subplots) was established. Eight subplots were selected, and we estimated vegetation cover (%) according to the different groups: live invasive grass (*Melinis minutiflora* and *Urochloa decumbens*), native graminoid, native forb, native shrub ( $\leq 1.5$  m), total dead biomass (native and invasive) and bare soil. Additionally, in each subplot two vegetative heights of the plant were randomly recorded, giving the average heights of the two invasive species and of the native functional groups.

All the aboveground biomass was sampled within each plot in a subplot of 0.5x0.5m randomly selected in the grid. The biomass was stored in paper bags in the field. In the laboratory, it was separated into: dead and live invasive grass (*Melinis minutiflora* and *Urochloa decumbens*), native graminoid, native forb, native shrub and native dead biomass. Biomass was oven-dried (70 °C, 3 days) and then weighted.

Vegetation surveys were carried out before the application of treatments for each species and then, every four months for a year (T0 – before treatments, T1 – 4 months, T2 - 8 months, and T3 – 12 months after treatments application).

#### 1.4. Data analysis

Data analysis was performed separately for each invasive grass. To verify differences of the measured variables at each time of observation among the treatments, non-parametric one-way analysis of variance (factor: treatment) were conducted by using Kruskal-Wallis test. All analyses were performed with STATISTICA (StatSoft, Inc 2007).

#### 3. Results

# 3.1. Treatment effects on the invasive grasses

Before treatments application (T0), *Melinis minutiflora* height was the same among treatments (H = 1.84, P = 0.604, Table 1). Four months after treatment application (T1), He and He + Ct treatments had lower height than Co plots (He -  $1.4\pm9.4$  cm, H = 18.16, P < 0.001; He+Ct -  $5.2\pm5.7$  cm, H = 18.16, P = 0.022, Co -  $35\pm16.3$  cm). After the reapplication of herbicide (T2), He plots showed shorter tussocks than Co ( $29.9\pm25.6$  cm, H = 16.43, P = 0.05) and cut plots ( $43.2\pm17.8$  cm, H = 16.43, P = 0.003). One year after the application of treatments (T3), herbicide and herbicide+cut plots still had heights < 7 cm, whilst Ct had *M. minutiflora* tussocks 2-fold taller than Control (Co =  $35\pm36.5$ ; Ct =  $72.9\pm22.2$  cm, P > 0.05).

*Urochloa decumbens* tussocks were the same height before treatments application (H = 1.26; P = 0.74, Table 1). Herbicide significantly reduced the height of the invasive grass tussocks in comparison to control plots eight months after treatments application (T2; 2.0±3.6 cm, H = 9.25, P = 0.039). One year after treatment application, tussocks of *U. decumbens* in He were shorter than 4 cm, whilst control and cut plots had tussocks > 40 cm (Table 1)

*M. minutiflora* cover was the same among treatments at T0 (Figure 1a). At T1, herbicide significantly reduced *M. minutiflora* cover in 63% (from 63.33% at T0 to 0.33% at T1, H = 17.75, P < 0.001, Figure 1b). Eight months and one year after treatments application, the cover of *M. minutiflora* in He and He+Ct plots was lower than in Ct plots (Figure 1c and d, T2: He - H = 16.49, P = 0.002, He+Ct - P = 0.008; T3: He - H = 14.64, P = 0.009, He+Ct - P = 0.004). Cut tended to increase the invasive grass cover since T2, whilst in Co plots, an opposite trend was found: the cover of *Melinis minutiflora* changed from 62.7±17.2% at T0 to 18.0±20.8 % at T3.

Similar pattern was found for *U. decumbens* invaded plots: there was a significant decrease of its cover in 48% at T1 in He plots (from 51.88% at T0 to 3.75% at T1, H = 9.58, P = 0.013, Figure 1b). At T2, the cover of *U. decumbens* in He and He+Ct was lower than 15% (Figure 1c). One year after treatments application, herbicide plots had a lower cover of the invasive grass in comparison to control plots (Figure 1d, H = 9.46; P = 0.04). Contrary to the pattern found for *Melinis minutiflora*, in Co plots, the cover of *Urochloa decumbens* was nearly the same at T0 (56.3±13.1 %) and T3 (50.8±7.8 %).

Table 1. Invasive grass, graminoids, forbs and shrubs height (mean±SD, cm) correspondent to *Melinis minutiflora* and *Urochloa decumbens* plots: before (T0), four (T1), eight months (T2) and one year (T3) after treatments application: Control (Co), Cut (Ct), Herbicide (He) and Herbicide + Cut (He + Ct), in a Cerrado area under natural regeneration at the Experimental Station of Itirapina. Statistically significant *P*-values ( $P \le 0.05$ ) are in bold. Different letters indicate significant differences among treatments for each time of observation ( $P \le 0.05$ ). The absence of letters indicates no significant differences.

Groups	Treatment				
	Со	Ct	He	He + Ct	P-value
M. minutiflora plots					
M. minutiflora					
Т0	$100.3\pm17.3$	$104.4\pm33.7$	$100.6 \pm 19.1$	$97.2\pm21.1$	0.604
T1	$35.0 \pm \mathbf{16.3^a}$	$10.3\pm7.7^{\text{ab}}$	$1.4\pm9.4^{\text{b}}$	$5.2\pm5.7^{\text{b}}$	<0.001
T2	$29.9\pm25.6^{ac}$	$43.2\pm17.8^{a}$	$1.7\pm5.5^{\text{b}}$	$2.8\pm6.8^{\text{bc}}$	<0.001
Т3	$35.0\pm36.4^{ab}$	$72.9\pm22.2^{\mathbf{a}}$	$5.8 \pm 14.1^{\text{b}}$	$6.2\pm15.0^{\text{b}}$	0.004
Graminoids					
ТО	$4.8\pm21.0$	$1.4\pm9.7$	$3.6 \pm 17.3$	$3.9 \pm 17.4$	0.862
T1	$1.6\pm9.0$	0.0	0.0	0.0	0.099
T2	$7.1\pm26.3$	$0.2 \pm 1.4$	$0.3 \pm 1.1$	$0.7\pm3.6$	0.721
Т3	$4.3\pm17.0$	0.0	$1.1\pm5.0$	$0.3\pm1.3$	0.549
Forbs					
Т0	$23.7\pm27.8$	$33.6\pm33.0$	$22.5\pm24.2$	$20.4\pm32.7$	0.753
T1	$11.9\pm20.2$	$6.8 \pm 11.3$	$7.9\pm24.1$	$4.6\pm 6.0$	0.835
T2	$19.6\pm22.2^{a}$	$18.6\pm16.1^{a}$	$4.4\pm6.6^{\text{b}}$	$10.6 \pm 15.0^{\text{ab}}$	0.002
Т3	$11.5\pm17.2$	$9.1 \pm 14.0$	$4.7\pm13.9$	$7.3\pm13.1$	0.436
Shrubs					
ТО	$20.9\pm50.4$	$7.9\pm26.5$	$11.4\pm35.3$	$2.8 \pm 14.7$	0.439
T1	$8.9\pm24.8^{a}$	$3.8\pm10.6^{a}$	$2.8\pm14.1^{\mathbf{a}}$	$0.2\pm1.4^{\mathbf{a}}$	0.046
T2	$11.8\pm30.0$	$5.6\pm20.6$	$0.2 \pm 1.4$	$3.9 \pm 14.6$	0.397
Τ3	$18.3\pm40.1$	16.1 ±35.3	$1.9 \pm 11.6$	$5.4 \pm 15.5$	0.079

Table 1. (Continued).

Groups	Treatment				
	Со	Ct	He	He + Ct	<i>P</i> -value
U. decumbens plots					
U. decumbens					
T0	$54.4 \pm 12.6$	$55.5\pm21.3$	$51.6 \pm 14.3$	$59.8\pm8.2$	0.739
T1	$43.2\pm11.8$	$37.5 \pm 12.3$	$13.0\pm22.0$	$36.6 \pm 15.2$	0.147
T2	$57.6\pm7.4^{\mathbf{a}}$	$48.6 \pm 18.0^{\text{ab}}$	$2.0\pm3.6^{b}$	$10.3 \pm 17.4^{\text{ab}}$	0.026
Т3	$41.3\pm11.3^{\text{a}}$	$40.9 \pm 10.0^{a}$	$3.9\pm8.0^{a}$	$11.6 \pm 15.6^{a}$	0.030
Graminoids					
TO	$14.8\pm21.9$	$4.8 \pm 12.9$	$9.4 \pm 19.0$	$7.8\pm23.2$	0.505
T1	$2.8\pm9.5$	$9.0\pm20.0$	$5.7 \pm 11.0$	$1.2\pm5.9$	0.194
T2	$4.6\pm13.3$	$16.2\pm27.7$	$8.9 \pm 18.3$	$1.5\pm5.0$	0.298
Т3	$3.7\pm12.7$	$7.2\pm14.3$	$5.8 \pm 12.4$	0.0	0.307
Forbs					
T0	$14.3\pm15.0$	$14.8\pm23.2$	$12.8\pm15.2$	$10.2\pm15.1$	0.875
T1	$24.3 \pm 17.7$	$11.8 \pm 17.3$	$13.3\pm10.4$	$16.3\pm17.1$	0.432
T2	$12.4\pm19.3$	$10.7\pm15.6$	$6.4\pm11.5$	$18.6\pm28.7$	0.487
Т3	$15.3\pm15.5$	$4.3\pm9.2$	$7.0\pm10.4$	$10.7\pm17.9$	0.258
Shrubs					
T0	$12.6\pm29.0$	$8.5\pm24.6$	$4.8 \pm 14.0$	$13.3\pm33.4$	0.740
T1	$9.3\pm22.9$	$5.2 \pm 14.4$	$2.7\pm13.3$	$3.1 \pm 15.1$	0.426
T2	$2.5\pm12.5$	0.0	$8.4\pm22.8$	0.0	0.170
Т3	$8.8\pm27.0$	$15.5\pm28.5$	$9.6\pm21.9$	$12.5\pm29.0$	0.710



Figure 1. Live *Melinis minutiflora* and *Urochloa decumbens* cover (Mean; Box: Mean±SE; Whisker: Mean±SD, %): (a) before, (b) four, (c) eight months and (d) one year after treatments application: Control (Co), Cut (Ct), Herbicide (He) and Herbicide + Cut (He + Ct), in a Cerrado area under natural regeneration at the Experimental Station of Itirapina. Different letters indicate significant differences among treatments ( $P \le 0.05$ ).

Live biomass of *M. minutiflora* represented >17% of the total biomass in the plots before treatments were applied (Table 2). Four months after treatments application, no live biomass of the invasive grass could be found in He plots, whilst He + Ct showed less than 0.5 g.m<sup>-2</sup> (Table 2). The same pattern was found at T2, and one year after treatments application, cut plots tended to accumulate more biomass than control plots (H = 11.84, P = 0.48) and He + Ct had the lowest amount of live biomass of *M. minutiflora* (H = 11.84, P = 0.007, Table 2).

Dead *M. minutiflora* biomass represented >48% of the total biomass at T0 (Table 2), accumulating more than 860 g.m<sup>-2</sup>. In opposite to what was found for live biomass, herbicide plots tended to have the highest amount of dead biomass at T1 and T2, while cut plots had the lowest values (Table 2). At T3, control plots accumulated 968±438.4 g.m<sup>-2</sup>, and the lowest amount of biomass was observed in He+Ct plots (170.7±190 g.m<sup>-2</sup>, H = 14.05, P = 0.01, Table 2).

*U. decumbens* live biomass quantities were lower than *Melinis minutiflora* at T0, varying from 67.3 to 159.4 g.m<sup>-2</sup> (Table 2). Immediately after treatments application (T1), a significant reduction in biomass amount was found in He in comparison to Co plots (H = 8.61, P = 0.03). At T3, no biomass of *U. decumbens* was found in He+Ct plots, whilst Ct plots tended to show the highest amounts of accumulated biomass (H = 7.58, P = 0.55). Following the same pattern observed for *Melinis minutiflora* live biomass, throughout the year, a reduction from 159.4 to 58.5 g.m<sup>-2</sup> was observed in Co plots.

Dead *U. decumbens* biomass represented > 32% of the total biomass at T0. However, four months after treatments application, the same amount of this grass was accumulated in He and Co plots, whereas in Ct and He + Ct plots showed opposite trend, accumulating less biomass. At T2 and T3, the same tendency was observed, however, cut and herbicide + cut increasingly showed more biomass than the previous times study (Table 2).

Table 2. Live and dead *Melinis minutiflora* and *Urochloa decumbens* biomass (mean±SD, g.m<sup>-2</sup>): before (T0), four (T1), eight months (T2) and one year (T3) after treatments application: Control (Co), Cut (Ct), Herbicide (He) and Herbicide+Cut (He+Ct), in a Cerrado area under natural regeneration at the Experimental Station of Itirapina. Statistically significant *P*-values ( $P \le 0.05$ ) are in bold. Different letters indicate significant differences among treatments for each time of observation ( $P \le 0.05$ ). The absence of letters indicates no significant differences.

Invasive	Treatment				
species	Со	Ct	He	He + Ct	<i>P</i> -value
M. minutiflora					
Live					
T0	$482.5\pm260.1$	$326.1\pm287.1$	$264.1\pm257.4$	$270.3\pm165.1$	0.308
T1	$38.6 \pm \mathbf{43.8^a}$	$6.7\pm9.3^{ab}$	0 <b>b</b>	$0.3\pm0.8^{ab}$	0.006
T2	$51.8\pm84.2^{\mathbf{a}}$	$39.1\pm59.5^{a}$	$0^{\mathbf{a}}$	$0^{\mathbf{a}}$	0.038
Т3	$33.1\pm41^{ab}$	$316.1\pm216.2^{\mathbf{a}}$	$13.7\pm33.4^{\text{ab}}$	$O^{\mathbf{b}}$	0.007
Dead					
Т0	$913.5\pm562$	$856.9\pm309.9$	$968.7\pm260.7$	$948.8\pm543.1$	0.962
T1	$612.0\pm230$	$371.0\pm225.7$	$717.3\pm247.4$	$363.5\pm200.6$	0.051
T2	$957.2\pm426.7^{a}$	$201.9\pm76.8^{\text{b}}$	1168.7±269.3 <sup>a</sup>	$538.3\pm353.1^{ab}$	0.001
Т3	$968.0{\pm}438.4^{a}$	$324.5\pm247.3^{ab}$	$789.5\pm322.5^{a}$	$170.7 \pm 190^{\text{b}}$	0.002
U. decumbens					
Live					
Т0	$159.4\pm158.2$	$88.2\pm97.3$	$67.3 \pm 44.3$	$105.6\pm55.6$	0.715
T1	$254.7\pm72.5^a$	$51.4 \pm 49.3^{ab}$	$3.8\pm4.0^{\text{b}}$	$116.3\pm91.1^{\text{ab}}$	0.035
T2	$105.2\pm55.9^{\rm a}$	$88.3 \pm \mathbf{95.4^a}$	$0^{\mathbf{a}}$	$0^{\mathbf{a}}$	0.023
Т3	$58.5\pm49.6$	$128.3\pm32.7$	$34.9\pm60.4$	0	0.055
Dead					
Т0	$333.8\pm55.7$	$298.3\pm330.7$	$414.1\pm114.8$	$475.3\pm466.3$	0.813
T1	$305.7 \pm 152.7$	$7.2\pm4.9$	$342.5 \pm 324.9$	$11.5\pm7.6$	0.227
T2	$236.1\pm155.9$	$84.0\pm75.1$	$373.5\pm56.5$	$99.7\pm96.8$	0.084
Τ3	$348.3 \pm 183.8$	$268.7\pm94.3$	$322.6\pm253.2$	$211.4\pm89.5$	0.788

# 3.2. Treatment effects on vegetation structure and functional groups

The treatments also resulted in important changes in the vegetation structure, regarding bare soil (Figure 2) and total dead biomass cover (Figure 3). Before treatments application, less than 5% of bare soil was found in *M. minutiflora* plots (Figure 2a). However, four (Figure 2b) and eight months (Figure 2c) after the application of treatments, Ct

significantly increased bare soil cover to > 25%, opening more gaps in the vegetation than in control plots (T1: H = 16.99, P = 0.02; T2: H = 16.83, P = 0.02, Figure 2b and c). He + Ct also increased bare soil cover in plots invaded by *Melinis minutiflora* at T2 (H = 16.83, P = 0.03, Figure 2b) and T3 (H = 12.91, P = 0.03, Figure 2d) compared to Control plots. Plots invaded by *U. decumbens* showed the same trend found for *Melinis minutiflora*: more open spaces within the vegetation after the application of Ct and He + Ct (Figure 2b and c). However, one year after treatments application, He+Ct plots showed higher cover of bare soil only in relation to control plots (Figure 2d, H = 7.67, P = 0.04).

The opposite trend can be observed for dead biomass in *M. minutiflora* plots: more than 50% of total dead biomass was found in the experimental plots at T0 (Figure 3a). Right after the application of treatments (T1), He plots had more dead biomass cover than control (H = 12.73, P = 0.008). At T2, He plots showed the same amount of dead biomass cover than control plots (H = 17.32, P = 0.85, Figure 3b), but they accumulated more biomass than Ct (P < 0.0001) and He + Ct plots (P = 0.03). One year after the application of treatments, cut plots had lower cover of dead biomass in comparison to control plots (H = 17.82, P = 0.03, Figure 3d). In experimental plots invaded by *U. decumbens*, a different trend was found: immediately after the application of treatments (T1, Figure 3b), He plots showed a higher cover of dead biomass than He+Ct (H = 9.70, P = 0.027), although neither of them differed from control plots. At T2 (Figure 3c) and T3 (Figure 3d), the cover of dead biomass was the same for all treatments (T2: H = 5.76, P = 0.12; T3: H = 4.12, P = 0.24).



Figure 2. Bare soil cover (Mean; Box: Mean±SE; Whisker: Mean±SD, %) in *Melinis minutiflora* and *Urochloa decumbens* plots (a) before, (b) four, (c) eight months and (d) one year after treatments application: Control (Co), Cut (Ct), Herbicide (He) and Herbicide + Cut (He + Ct), in a Cerrado area under natural regeneration at the Experimental Station of Itirapina. Different letters indicate significant differences among treatments ( $P \le 0.05$ ).



Figure 3. Total dead biomass cover (Mean; Box: Mean±SE; Whisker: Mean±SD, %) in *Melinis minutiflora* and *Urochloa decumbens* plots (a) before, (b) four, (c) eight months and (d) one year after treatments application: Control (Co), Cut (Ct), Herbicide (He) and Herbicide + Cut (He + Ct), in a Cerrado area under natural regeneration at the Experimental Station of Itirapina. Different letters indicate significant differences among treatments ( $P \le 0.05$ ).

In *Melinis minutiflora* invaded plots, before treatments application, graminoids average height was  $3.4\pm16.8$  cm, forbs  $25.1\pm29.9$  cm and shrubs  $10.7\pm34.6$  cm (Table 1). Graminoids height was not influenced by treatments application (H = 6.26, P = 0.099), however, Ct tended to decrease graminoids height at T2 (H = 1.33, P = 0.721), as well He + Ct at T3 (H = 2.11, P = 0.549, Table 1). On the other hand, forbs had their height decreased at T2 in He plots in comparison to Co (H = 14.66, P = 0.006) and Ct plots (H = 14.66, P = 0.006, Table 1). Shrubs were lower in He + Ct plot immediately after treatments application (H = 7.98, P = 0.046, T1). However, at T2 and T3, no significant differences could be observed among treatments (P = 0.39 and P = 0.07 respectively, Table 1).

The height of the functional groups found in plots invaded by *Urochloa decumbens* were not affected by the treatments, at any time of observation (P > 0.05, Table 1). The height of graminoids tended to decrease throughout the year of the experiment: 14.8±21.9 at T0 to  $3.7\pm12.7$  at T3 in Co plots (Table 1). Shrubs showed the same pattern found for graminoids, whilst forbs height tended to be the same in control plots before and one year after treatments application (P = 0.258, Table 1).

Cover of graminoids in plots invaded by *Melinis minutiflora* was not affected by the treatments at any time of observation (Table 3, P > 0.05). Graminoids cover was lower than 3%. Control plots had 1±5.3% of graminoids cover at T0 and 2.9±12.2% at T3. One year after treatments application, cut plots had no graminoids (Table 3), and He and He+Ct plots had less than 1% of graminoids cover. Forbs were negatively affected by He at T2: forbs cover was lower than in control (H = 13.3, P = 0.04) and cut plots (P = 0.003). However, one year after treatments application, forbs cover was the same among treatments (H = 3.17, P = 0.37, Table 3). Shrubs cover was significant reduced immediately after treatments application (T1) in He and He + Ct (H = 8.77, P = 0.03). At T3, shrubs tended to have lower cover (He:  $0.5\pm2.6\%$ , He+Ct:  $3.5\pm9.2\%$ ) than in control plots ( $6.4\pm13.8\%$ , H = 5.6, P = 0.13, Table 3).

None of the functional groups cover was significantly affected by the treatments in plots invaded by *Urochloa decumbens*, at any time of observation (Table 3, P > 0.05). Graminoids cover in control plots at T0 was 7.7±12.9% and decreased to 1.5±5% at T3. The same pattern was found for all treatments, except for the cut plots (T0: 5±15%; T3: 6.5±12.2%, Table 3). No significant differences were found for forbs cover (P > 0.05), although Ct tended to reduce it subsequently after treatments application (Table 3). On the other hand, shrubs cover tended to increase in all treatments from T0 to T3 (Table 3).

Before treatments application, the total biomass was composed by <1% of graminoids and forbs, and <7% of shrubs biomass in *Melinis minutiflora* invaded plots (Table 4). Treatments did not affect any of the functional groups biomass, at any time of observation (P< 0.05). However, some trends could be observed. Graminoids biomass was not registered in any experimental plot after treatments application, except for Ct plots at T2 (Table 4). Forbs tended to be negatively affected by He and Ct, since the amount of forbs biomass decreased in these treatments, whilst in control plots, it increased nearly 40-fold from T0 to T3 (Table 4). After treatments application, shrubs biomass was not registered at T1 and T2 (except in Co plots, 5%). However, at T3 they were not found only in Ct plots (3.5%, Table 4).

In *Urochloa decumbens* plots at T0, graminoids biomass represented <1% of the total biomass, forbs < 14%, and shrubs < 13%. The same pattern was found for plot invaded by *U. decumbens:* treatments did not affect any of the functional groups biomass. However, a trend could be observed for forbs: biomass tended to decrease in all treatments from T0 to T3, being not registered in Ct and He+Ct plots. On the other hand, shrubs biomass increased from T0 to T3 in the same treatments (Ct, He+Ct, Table 4).

Table 3. Graminoids, forbs and shrubs cover (mean±SD, %) correspondent to *Melinis minutiflora* and *Urochloa decumbens* plots: before (T0), four (T1), eight months (T2) and one year (T3) after treatments application: Control (Co), Cut (Ct), Herbicide (He) and Herbicide + Cut (He + Ct), in a Cerrado area under natural regeneration at the Experimental Station of Itirapina. Statistically significant *P*-values ( $P \le 0.05$ ) are in bold. Different letters indicate significant differences among treatments for each time of observation ( $P \le 0.05$ ).

Functional groups	Treatment				
-	Со	Ct	He	He + Ct	<i>P</i> -value
M. minutiflora plots					
Graminoids					
T0	$1.0\pm5.3$	$0.4 \pm 2.9$	$2.3\pm10.8$	$1.7\pm9.1$	0.780
T1	$0.8\pm5.8$	0	0	0	0.391
T2	$2.8 \pm 11.1$	$0.2 \pm 1.4$	$0.3\pm1.2$	$0.2 \pm 1.4$	0.825
Т3	$2.9\pm12.2$	0.0	$0.9\pm3.4$	$0.3\pm1.6$	0.549
Forbs					
Т0	$9.9 \pm 12$	$15.6 \pm 14.8$	$6.0\pm8.8$	$11.5\pm13.9$	0.229
T1	$7.0 \pm 11.8$	$7.0\pm9.4$	$2.2\pm5.0$	$8.3 \pm 11.5$	0.093
T2	$14.4\pm16.3^{a}$	$20.4 \pm 17.0^{a}$	$4.0\pm7.8^{\textit{b}}$	$13.6\pm16.5^{ab}$	0.004
Т3	$9.0\pm13.4$	$6.5\pm8.6$	$3.1\pm 6.8$	$7.6 \pm 11.6$	0.365
Shrubs					
Т0	$4.4\pm11.4$	$2.9\pm8.8$	$1.1 \pm 4.5$	$2.5\pm8.1$	0.641
T1	$3.1\pm8.0^{a}$	$2.5\pm8.4^{a}$	$0.4\pm2.3^{\mathbf{a}}$	$0.1\pm0.7^{\mathbf{a}}$	0.032
T2	$5.0\pm13.3$	$2.0\pm7.4$	$0.1\pm0.7$	$2.1\pm7.7$	0.419
Т3	$6.4\pm13.8$	$6.1\pm13.1$	$0.5\pm2.6$	$3.5\pm9.2$	0.132
U. decumbens plots					
Graminoids					
Т0	$7.7 \pm 12.9$	$5.0 \pm 15.0$	$10.4\pm20.7$	$2.1\pm 6.1$	0.616
T1	$1.7\pm6.4$	$3.5\pm8.0$	$8.3 \pm 18.2$	$0.6\pm3.1$	0.521
T2	$2.5\pm7.2$	$9.8 \pm 19.3$	$5.8 \pm 12.9$	$0.6\pm2.2$	0.392
Т3	$1.5\pm5.0$	$6.5\pm12.2$	$5.0\pm11.0$	0	0.180
Forbs					
Т0	$8.2\pm11.2$	$14.0\pm26.0$	$6.3\pm9.1$	$6.5\pm11.8$	0.787
T1	$13.1\pm10.3$	$9.2 \pm 11.9$	$10.8 \pm 14.3$	$19.6\pm13.4$	0.433
T2	$10.0\pm15.3$	$8.5\pm13.2$	$7.0\pm12.4$	$9.8\pm8.9$	0.738
Т3	$12.3\pm13.1$	$2.9\pm5.3$	$9.0\pm13.0$	$11.3\pm8.8$	0.374
Shrubs					
Т0	$2.9\pm8.7$	$2.7\pm7.5$	$1.3 \pm 3.4$	$2.5\pm 6.9$	0.613
T1	$4.0\pm10.2$	$1.5\pm3.5$	$1.5 \pm 7.1$	$0.4\pm2.0$	0.223
T2	$1.7\pm8.2$	0	$4.8 \pm 11.5$	0	0.172
Т3	$3.3\pm9.2$	7.7 ± 12.7	8.5 ± 15.8	$5.4 \pm 11.6$	0.557

Table 4. Graminoids, forbs and shrubs biomass (mean±SD, g.m<sup>-2</sup>) correspondent to *Melinis minutiflora* and *Urochloa decumbens* plots before: (T0), four (T1), eight months (T2) and one year (T3) after treatments application: Control (Co), Cut (Ct), Herbicide (He) and Herbicide + Cut (He + Ct), in a Cerrado area under natural regeneration at the Experimental Station of Itirapina. Statistically significant *P*-values ( $P \le 0.05$ ) are in bold. Different letters indicate significant differences among treatments for each time of observation ( $P \le 0.05$ ). The absence of letters indicates no significant differences.

Functional groups	Treatment				
	Со	Ct	He	He + Ct	<i>P</i> -value
M. minutiflora plots					
Graminoids					
ТО	$11.3\pm27.8$	0.0	0.0	0.0	0.391
T1	0.0	0.0	0.0	0.0	1.000
T2	0.0	$0.007 \pm 0.016$	0.0	0.0	0.391
Т3	0.0	0.0	0.0	0.0	1.000
Forbs					
ТО	$0.8 \pm 1.3$	$7.9\pm19.3$	$3.7\pm7.9$	$6.2\pm9.3$	0.724
<b>T</b> 1	$0.2\pm0.4$	$2.0\pm4.5$	0.0	0.0	0.248
T2	$34.6\pm37.9$	$13.1\pm22.8$	0.0	$10.0 \pm 19.7$	0.206
Т3	$33.8\pm81.9$	$5.5\pm10.0$	$0.2\pm0.6$	$9.5\pm10.9$	0.557
Shrubs					
Τ0	$54.2 \pm 123.7$	$110.0\pm229.7$	0.0	0.0	0.206
T1	0.0	0.0	0.0	0.0	1.000
T2	$54.7 \pm 106.0$	0.0	0.0	0.0	0.099
T3	$3.0\pm7.2$	0.0	$2.3\pm5.5$	$20.1\pm35.0$	0.439
U. decumbens plots					
Graminoids					
Τ0	0.0	$7.3 \pm 12.6$	0.0	0.0	0.391
T1	$2.3\pm3.3$	0.0	0.0	0.0	0.087
T2	$3.5\pm6.1$	$3.3\pm5.7$	0.0	0.0	0.529
Т3	$66.8\pm31.3$	0.0	$114.3 \pm 198.0$	0.0	0.082
Forbs					
Τ0	$66.5\pm115.2$	$152.8\pm168.2$	$0.3\pm0.5$	$3.9\pm5.1$	0.473
<b>T</b> 1	$45.2\pm72.2$	$4.5\pm7.8$	$3.8\pm 6.6$	$29.2\pm39.7$	0.681
T2	$11.8 \pm 19.7$	$12.8\pm22.2$	$13.2\pm22.9$	$9.1\pm15.8$	0.937
Т3	$3.1 \pm 4.1$	0.0	$0.4\pm0.7$	0.0	0.059
Shrubs					
Τ0	$24.5\pm42.4$	0.0	$132.0\pm228.6$	0.0	0.529
T1	0.0	0.0	0.0	0.0	1.000
T2	0.0	0.0	0.0	0.0	1.000
T3	0.0	$51.8\pm89.7$	$5.0\pm8.6$	$3.2\pm5.5$	0.733

# 4. Discussion

# 4.1. Treatment effects on the invasive grasses

Studies in Cerrado have shown positive responses of glyphosate in controlling *Urochloa decumbens* (Durigan *et al.*, 1998) and *Melinis minutiflora* when combined to fire and hand removal (Martins *et al.*, 2011). In this study, as hypothesized, the use of glyphosate proved to be efficient in controlling both invasive species in a Cerrado area under regeneration. Herbicide reduced approximately 59% (from 63.33% at T0 to 4.71% at T3) *M. minutiflora* cover and in 95% of its live biomass (from 264.08 g.m<sup>-2</sup> at T0 to 13.72 g.m<sup>-2</sup> at T3), whilst *U. decumbens* cover was reduced 49% (from 51.88% at T0 to 2.88% at T3) and live biomass in 48% (from 67.28 g.m<sup>-2</sup> at T0 to 34.85 g.m<sup>-2</sup> at T3), one year after its application. Moreover, the combination of herbicide + cut was also an effective management technique, showing similar results to herbicide application: reduction in 58% (from 62.81% at T0 to 0 g.m<sup>-2</sup> at T3), while 53% (from 61.67% at T0 to 9.04 at T3) *U. decumbens* cover and in 100% live biomass (from 105.56 g.m<sup>-2</sup> at T0 to 0 g.m<sup>-2</sup> at T3).

Not only cover and biomass of the invasive grasses were reduced in He and He + Ct plots, but also tussock height. The reapplication of herbicide was also an important factor that contributed to control the invasive species, as observed at T2, when both species reached low values of cover and biomass.

On the other hand, the cut treatment showed to be ineffective to control the invasive grasses, since no significant differences were observed to the Co plots. After one year, cut plots had an increase in 9% (from 65.94% at T0 to 56.67% at T3) *M. minutiflora* cover and live biomass continued similar to before treatments application, decreasing only approximately 3% (from 326.08 g.m<sup>-2</sup> at T0 to 316.15 g.m<sup>-2</sup> at T3). For *U. decumbens*, there

was an increase in 45% of its live biomass (from 88.24 g.m<sup>-2</sup> at T0 to 128.31 g.m<sup>-2</sup> at T3), showing the high capacity of these two species to regenerate after being cut.

Studies that used similar techniques to control these exotic species showed that, in exception of clipping once, a double-clipping could be an effective method to reduce both invasive species biomass (Gorgone-Barbosa, 2009). The ineffectiveness of a single cut to control *Melinis minutiflora* was also demonstrated by Silva *et al.* (2013). However, other studies showed that annually cut was efficient to reduce *Melinis minutiflora* and increase native grass cover, although this treatment led to negative effects on native dicots cover in long term (Sato *et al.*, 2013).

Despite the positive effect of herbicide in controlling both invasive grasses, this treatment led to a high accumulation of dead biomass, which can have negative effects on native plant community. The dead biomass of these grasses are fuel material that can increase the risk of wildfires (D'Antonio and Vitousek, 1992). Gorgone-Barbosa *et al.* (2015) observed that the higher the amount of *Urochloa brizantha* dead biomass, the more intense are fires in Cerrado areas. Due to the higher accumulation of dead biomass at the end of the dry season, the risks of intense fires in this period also increase. Moreover, the presence of *U. brizantha* alters fire behavior, leading to fires of higher temperature and flames, which could influence vegetation structure by topkilling trees and shrubs (Gorgone-Barbosa *et al.*, 2015). *Melinis minutiflora* can also alter fire behaviour, since fire prediction models simulating fire behavior in Cerrado demonstrated that its presence resulted in rapidly fire spread, at high intensity and with high flames (Mistry and Berardi, 2005). Therefore, the high accumulation of dead biomass promoted by the use of herbicide could lead to more intense fires, with higher temperature and flames, which could negatively affect vegetation in invaded areas.

Additionally, the high cover and amount of live and dead biomass of invasive species could act as a mechanical barrier that can negatively influence the native plant community,

because they can shade seedlings and young individuals of other species, limiting their growth (D'Antonio *et al.*, 1998; Cabin *et al.*, 2002). Seedling establishment of woody species from Cerrado was negatively affected by *Melinis minutiflora* biomass, due to the reduction of light as a consequence of shading (Hoffmann and Haridasan, 2008). Therefore, herbicide+cut could be an alternative to the use of herbicide alone, due to the removal of dead biomass after herbicide application.

However, we observed that, after the removal of aboveground biomass in Ct and He + Ct plots, *Urochloa decumbens* occupied the spaces left by *Melinis minutiflora* removal (results not shown), demonstrating its high efficiency in occupying newly open spaces, replacing both native and other invasive species (in this case, *Melinis minutiflora*). These treatments probably resulted in an increase in the availability of resources or a decline in the uptake of liming resources by the removed species, so the site became more susceptible to invasion (Davis *et al.*, 2000). Thus, when more resources became available, species with efficient dispersal mechanisms, rapid growth and high productivity behaved opportunistically (Rejmánek *et al.*, 2013).

# 4.2. Treatment effects on the vegetation structure and functional groups

Although techniques to control invasive species are crucial to facilitate the restoration process of disturbed areas, most treatments must be evaluated concerning their effects on native plant communities (Cabin *et al.*, 2002). In this study, as hypothesized, forbs were negatively affected by herbicide, which reduced their height and cover compared to other treatments eight months after treatments application in plots invaded by *Melinis minutiflora*. Whilst herbicide + cut showed the trend to affect graminoids establishment. However it is important to notice that all functional groups had low representativeness in all treatments before their application, highlighting the negative effect of the invasive species on the native plant community, as already shown by Pivello *et al.* (1999b). Therefore, although most of the

treatments did not affect any of the functional groups, the initial condition of the plant community should be considered and thus, treatments did not enhance vegetation regeneration.

The inappropriate use of glyphosate, including its repeated application to previously weakened plants may eliminate plant individuals or result in a disadvantage on the their competitive ability (Matarczyk *et al.*, 2002). The effect of herbicide on the herbaceous layer is consistent to what found by Aigner and Woerly (2011), who tested the use of the herbicide and cut to control an annual invasive grass (*Aegilops triuncialis*) in open fields in California. The authors found that both treatments negatively affected forbs cover, whereas glyphosate eliminated a native grass and tended to eradicate another rare annual forb (*Navarretia jepsonii*). Other forb species (*Aster ericoides*) had its frequency reduced after the application of glyphosate in the prairie (Tunnell *et al.*, 2006). Therefore, the use of herbicide should be carefully analyzed, not only on its effectiveness in controlling invasive species, but also on its effect on native species. Moreover, its effects in the environment, such as on soil biota, edaphic components, groundwater and surface water.

The structure of the plant community was significantly modified by some treatments, since aboveground biomass removal in Ct and He + Ct plots led to opening of gaps, exposing more bare soil. As a result, there was an increase in light availability at ground level (D'Antonio *et al.*, 1998; MacDougall and Turkington, 2005) which can stimulate the regeneration of vegetation by providing a favorable microclimate for the establishment of new species (Cabin *et al.*, 2002), especially for shade-intolerant plant species (Iglay *et al.*, 2010). Also, the newly created gaps represent new opportunities for plant establishment, since invasive species occupied these spaces before, impeding the establishment of native species. However, as mentioned before, the new gaps are also new sites available for invasion. In this case, additional techniques could be implemented to improve native species establishment

over the invasive ones. Seeding or planting of native species is often a critical step after applying treatments to control invasive species in the prairie (Dennehy *et al.*, 2011). The removal of *Pennisetum setaceum* and consequent addition of native species proved to be an efficient technique to enhanced native species in dry forest in Hawaii (Cabin *et al.*, 2002). In this case, favorable microsites were created and the establishment of native species was facilitated. Moreover, species that are not able to resprout should be planted, as demonstrated for the seasonal deciduous forest in abandoned pastures in Brazil (Sampaio *et al.*, 2007). Hence, not only the application of techniques to control invasive species should be applied in some areas, but also restoration tools should be used in order to recolonize former invaded areas after their control.

#### **5.** Conclusions

In short-term, He and the combination of He + Ct were efficient to control *Melinis minutiflora* and *Urochloa decumbens*, since invasive grasses in these plots had their height, cover and live biomass reduced. Although the use of herbicide led to the decrease of invasive grasses, a high accumulation of dead biomass of these species could be observed in these plots, which could lead to negative effects on plant community. Therefore, the removal of the dead biomass should be performed after the application of herbicide (He + Ct). However, the use of cut alone is not recommended, since it enhanced invasive species

On the other hand, native forbs tended to decrease in plots where herbicide was applied and thus, it affected negatively forb community. Therefore, studies about techniques to control invasive species should consider their effects on native vegetation as well.

Hence, although both He and He + Ct were effective to control *Melinis minutiflora* and *Urochloa decumbens*, He + Ct would be the suggested treatment, since it did not result in biomass accumulation and provided more bare soil, which can make more light available for

species regeneration. However, more bare soil availability could also result in reinvasion, and thus techniques that promote the occupation of spaces by native species could also be used to facilitate their establishment in the area, such as seeding of natives after the control of the invasive species. Finally, it is essential to consider that reducing the cover and biomass of invasive after a year of management does not guarantee their long-term control. Monitoring the effectiveness of treatments should be continued, since seeds of invasive grasses can be viable in the seed bank and seedlings of these species can establish again.

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# CAPÍTULO II

Section: Essays & Perspectives

# Benefit/cost analysis of different management techniques to control invasive grasses in natural reserve of Cerrado

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## ABSTRACT

Biological invasions are one of the major causes of species extinction. In Brazil, African grasses have invaded Cerrado areas, particularly natural reserves. The benefit/cost rate of techniques to control invasive species provides information that can minimize the invasion process in protected areas. Thus, the present study aimed to analyze the benefit/cost ratio of cut, herbicide, herbicide+cut, and control (no intervention) to the control of Melinis minutiflora and Urochloa decumbens. Treatments were applied on 4x4m plots (6 replicates/treatment to Melinis minutiflora and 3 replicates/treatment to Urochloa decumbens). In 0.5x0.5m plots, all the aboveground biomass was sampled and separated into total invasive grass (live and dead) of *M. minutiflora* and *U. decumbens*, dried (70°C for 3 days) and weighed. The surveys were conducted before the treatments application and every four months (12 months total). The benefit/cost ratio was determined by the amount of biomass (kg) reduced by currency (U.S. Dollar) after 1 year. In small areas  $(1 \text{ m}^2 \text{ and } 16 \text{ m}^2)$ , all tested treatments have similar benefit/cost for both invasive species. While, herbicide+cut for Melinis minutiflora and Urochloa decumbens was the most efficient treatment in terms of benefit/cost ratio to control the species in larger areas (5000 m<sup>2</sup> and 10000 m<sup>2</sup>). Our study provides important information for management decisions of invasive grasses in Cerrado areas and reinforces the need for an assessment of economic efficiency when planning the control of invasive species in order to use the funds for conservation more effectively.

Keywords: protected areas, biological invasions, herbicide, *Melinis minutiflora*, *Urochloa decumbens*.

# INTRODUCTION

Biological invasions are among the main causes of species extinction (D'Antonio & Vitousek 1992) and it is estimated that billions of dollars have been spent with the impacts of invasive species (Pyšek and Richardson, 2010). Alien species with great ability to disperse to new habitats distant from the site of its introduction can start the invasion process (Pyšek, 1995; Richardson *et al.*, 2000) and its control can be very difficult and cost spending (Mack *et al.*, 2000). The negative impacts of invasive plants are the decrease of the biomass and diversity of native species (Flory and Clay, 2009), as well changes in fire regimes (D'Antonio and Vitousek 1992), which can be a great problem for managers in natural reserves.

The harmful consequences of the presence of these species are widely recognized in many parts of the world, as well as the need to reduce their current and future impact (Pyšek & Richardson 2010). African grasses, such as *Melinis minutiflora* P. Beauv. and *Urochloa decumbens* Stapf. are exotic species introduced in Brazil for animal feeding (Lorenzi 2008) that have widely spread, becoming invasive and competing with native herbaceous species (Pivello *et al.* 1999a). As a result, invasive grasses are one of the main threats to Cerrado nature reserves (Pivello *et al.* 1999a; Durigan *et al.* 2007). These African grasses are highly competitive because they promote a competitive pressure on the native species (Pivello *et al.*, 1999b) and accumulate a large amount of biomass (Hoffmann *et al.* 2004), increasing the risk of wildfires occurrence (D'Antonio and Vitousek, 1992).

In order to minimize the invasion process, different strategies can be used to control invasive species (Clout and Williams, 2009). Techniques that remove aboveground biomass, such as cutting or mowing are examples of the mechanical methods that can be used, although they can become expensive because of the great need of labor (Wittenberg and Cock, 2001). Herbicide is a chemical method (Galli and Montezuma, 2005), that has fast and evident

results on the control of invasive species (Luken and Seastedt, 2004). Herbicide (fluazifop-Pbuty) proved to be effective in reducing the invasive annual grass *Microstegium vimineum* (Trin.) A. Camus growth in deciduous forests in Indiana (USA) (Emery *et al.*, 2013) and the perennial grass *Schedonorus phoenix* (Scop.) Holub. in the grassland in Kentucky (USA) (Ruffner and Barnes, 2010). The herbicide spraying is encouraged by a wide variety of products that are efficient in controlling invasive species (Luken and Seastedt, 2004) with reduced labor efforts (Radosevich *et al.*, 2007). In addition, in Brazil herbicide spraying is allowed by law in protected areas that include its use in their management plan (Normative Ruling IBAMA N°7, of July 2<sup>nd</sup>, 2012).

The herbicide glyphosate is usually recommended because of its low toxicity and rapid degradation in soil (Sprankle *et al.*, 1975). However, some studies showed negative effects of the use of this herbicide on the native plant community, such as on the endangered Australian populations of *Pimelea spicata* R.Br. (Matarczyk *et al.*, 2002) and on reducing richness and diversity of native species on the seed bank in temperate grasslands (Rodriguez and Jacobo, 2012).

Additionally to the question of which management technique might be the most efficient, managers should be aware of the costs of controlling invasive species which can use a large part of protected area budget (Usher, 1988). The view that invasive species are dangerous, destructive and lead native plant community to extinction can bring the motivation that these species should be eradicated at all costs (Smith *et al.*, 2006). Moreover, given the budgetary constraints, managers may face difficulties when planning the management of these species (Andreu *et al.*, 2009), so that the management is recommended only when the benefits exceed the costs (Wiles 2004).
Few studies have measured the benefit/cost ratio to control invasive species (Dehnen-Schmutz *et al.*, 2004; Gorgone-Barbosa 2009; McConnachie *et al.*, 2012). Most studies are used for agriculture, due to the economic damage that invasive species can cause (Snipes *et al.* 1984; Judice *et al.* 2006). Economic analysis are essential to support management decisions and to define research priorities, considering the lack of information about the costs and benefits of invasive management (Wiles, 2004). Furthermore, these data would allow managers to make a decision as fast as possible in order to avoid further expansion of such species within protected areas.

In the United States (Pimentel *et al.*, 2005) and South Africa (McConnachie *et al.*, 2012), there is a great concern with the assessment of the economic damage caused by invasive species within protected areas. In Brazil, however, the calculation of management costs is not yet a widespread practice among policy-makers and managers of natural reserves. Therefore, this study aimed to evaluate the benefit/cost ratio in the short-term (1 year) of management techniques to control the total biomass (live and dead) produced by invasive grasses (*Melinis minutiflora* and *Urochloa decumbens*) in a Cerrado conservation unit. Thus, it is intended to provide information that supports management practices of invasive grasses, as well as conservation actions.

#### MATERIAL AND METHODS

#### Study site and study species

This study was carried out at the Florest Management Zone, at the Experimental Station of Itirapina (ESI, Southeastern Brazil, 22° 15' a 22° 15' S, - 47° 45' e 47° 51' W, 710 - 830 m a.s.l.). The Forest Management Zone consists of an area under natural regeneration after the removal of a *Pinus oocarpa* stand in 1999 (Zanchetta, personal communication). The Experimental Station of Itirapina (ESI) is characterized by having remnants of *cerrado sensu* 

*stricto*, *cerradão* and riparian forests. However, the area is mostly covered by *Pinus* spp. and *Eucalyptus* spp. plantations. In addition, some areas covered by *cerrado* were used as pasture and, therefore, the vegetation in these sites is strongly disturbed, although there can be seen species of native flora (Zanchetta *et al.* 2006).

The climate is mesothermal and seasonal with a very marked rainy (October to March) and dry season (April to September, Zanchetta *et al.* 2006). Annual average precipitation is 1459 mm and temperature  $21.9^{\circ}$ C (Zanchetta *et al.* 2006). The study area is highly invaded by *Melinis minutiflora* Beauv. and *Urochloa decumbens* (Stapf) RD Webster. Both are C<sub>4</sub> Poaceae grasses from South Africa (Klink & Joly 1989; Martins 2006). In Brazil, *M. minutiflora* was first described near Rio de Janeiro in 1812 (Parsons, 1972) and currently this species is already found in almost all regions of Brazil (Martins, 2006), invading mainly areas of *campo cerrado* and *campo sujo* in São Paulo state (Pivello *et al.* 1999a; 1999b). *U. decumbens* was introduced in Brazil in the 1950's (Lorenzi 2008) and it is observed in the states of São Paulo, Minas Gerais and Goiás in natural reserves of Cerrado (Lorenzi 2008).

## Experimental design

Plots of 4x4m (6 replicates/treatment= 24 plots for *Melinis minutiflora* and 3 replicates/treatment= 12 plots for *Urochloa decumbens*) were established in the study area in patches invaded by the African grasses. Each plot had at least  $\geq$  50% of invasive grasses cover. The treatments tested were: **Cut** (**Ct**): removal of all aboveground biomass; **Herbicide** (**H**): glyphosate at 1.5% application by spraying only on the tussocks of the study invasive grasses, and reapplication after six months; **Herbicide**+**Cut** (**He**+**Ct**): application of glyphosate at 1.5% on the tussocks of invasive grasses (reapplied after six months) and removal of all aboveground biomass after seven days, and **Control** (**Co**): no treatment. For biomass sampling, we sampled all the aboveground biomass of 0.5x0.5m subplots in each

sampling unit and stored in paper bags in the field. In the laboratory, the biomass was separated into: total invasive grass (live and dead) of *Melinis minutiflora* and *Urochloa decumbens*, dried (70°C, 3 days) and weighted.

#### Benefit/cost ratio of treatments

The cost of each treatment was calculated for one-year of management. Initially, costs were calculated for the management of areas of 1 m<sup>2</sup> and 16 m<sup>2</sup>, the later corresponding to treatments in the current experiment. Moreover, these costs were extrapolated to larger areas  $(5.000 \text{ m}^2 \text{ and } 10.000 \text{ m}^2)$ . In order to extrapolate data, all calculations were based in the time and costs to apply the treatments in a 1m<sup>2</sup> area (Table 3). Costs were based on prices for the year 2013, corresponding to the year of treatments application, so they may have had adjustments in subsequent years. Furthermore, it is important to consider that the calculations were performed only regarding the work of one man. In case of more people and in larger areas, costs would increase with tools and labor (men working), but it would demand less time. The classification of the categories of costs is described in Table S1. Moreover, in order to avoid the accumulation of biomass in the protected area, its final destination could be burned or used for other purposes. The estimated cost for discard was based on the amount of biomass removed from 1 m<sup>2</sup> in Ct and He+Ct plots at T0 (before the treatments application). This amount was extrapolated to larger areas of 16 m<sup>2</sup>, 5000 m<sup>2</sup> (0.5 ha) and 10000 m<sup>2</sup> (1 ha) (Table 1).

Table 1. Total biomass (mean, kg, dead and alive) of *Melinis minutiflora* and *Urochloa decumbens* removed from areas of 1 m<sup>2</sup> and extrapolated to larger areas of 16 m<sup>2</sup>, 5.000 m<sup>2</sup> and 10.000 m<sup>2</sup> before the application of the treatments: Cut (Ct), Herbicide (He) and Herbicide+Cut (He+Ct) in a Cerrado area under natural regeneration at the Experimental Station of Itirapina (Southeastern Brazil).

$(kg.m^{-2})$	(kg.16 m <sup>-2</sup> )	$(kg.5000 m^{-2})$	$(kg.10000 m^{-2})$
1.18	18.93	5914.85	11829.70
1.23	19.72	6163.98	12327.97
1.22	19.51	6095.45	12190.90
0.39	6.18	1932.73	3865.47
0.48	7.70	2407.00	4814.00
0.58	9.29	2904.47	5808.93
	( <b>kg.m</b> <sup>-2</sup> ) 1.18 1.23 1.22 0.39 0.48 0.58	(kg.m <sup>-2</sup> )(kg.16 m <sup>-2</sup> ) $1.18$ $18.93$ $1.23$ $19.72$ $1.22$ $19.51$ $0.39$ $6.18$ $0.48$ $7.70$ $0.58$ $9.29$	(kg.m <sup>-2</sup> )(kg.16 m <sup>-2</sup> )(kg.5000 m <sup>-2</sup> ) $1.18$ $18.93$ $5914.85$ $1.23$ $19.72$ $6163.98$ $1.22$ $19.51$ $6095.45$ $0.39$ $6.18$ $1932.73$ $0.48$ $7.70$ $2407.00$ $0.58$ $9.29$ $2904.47$

The total area managed per day (6 hours work) was based on the estimated time that a worker takes to manage (apply the treatments) an area of 16 m<sup>2</sup> (in each plot, 0.5 h in Ct plots and 0.3 h in He plots). Thus, in a day work in Ct plots, it was possible to manage 192 m<sup>2</sup>, whereas for herbicide application, a total of 291 m<sup>2</sup> would be managed. The combined treatment of herbicide+cut, was carried out using more than one day (one day for the herbicide application, another day for biomass removal). Moreover, another day should be added for herbicide reapplication (six months after the first application). Finally, calculation of the benefit/cost ratio for each treatment was performed in order to compare effectiveness of different treatments. Thus, the benefit was represented by the difference between the biomass sampled after one year (T3) in the control group and each treatment biomass for *Melinis minutiflora* and *Urochloa decumbens*, determining biomass reduction in efficiency (kg).

benefit/cost ratio, i.e., the amount of biomass (in kilograms) that is reduced by monetary unit (U.S. Dollar average for the year 2013, year of management).

## RESULTS

#### Benefit/cost ratio of the treatments

The benefit of each treatment (Table 2) showed that herbicide+cut was the treatment that mostly reduced the total invasive biomass (live and dead) of both African grasses in areas of 1 m<sup>2</sup>, 16 m<sup>2</sup>, 5000 m<sup>2</sup> and 10000 m<sup>2</sup>. Based on the amount of biomass found before treatments application (T0: Table 1) and one year after treatments application (T3: Table S2) for *M. minutiflora*, the total invasive biomass was reduced in 83% (from 1.22 kg.m<sup>-2</sup> at T0 to 0.17 kg.m<sup>-2</sup> at T3) by herbicide+cut treatment, whilst other treatments reduced less than 46% (Ct: from 1.18 kg.m<sup>-2</sup> at T0 to 0.64 kg.m<sup>-2</sup> at T3; He: from 1.23 kg.m<sup>-2</sup> at T0 to 0.80 kg.m<sup>-2</sup> at T3). In He plots, there was a great accumulation of *M. minutiflora* dead biomass (Table S2), which decreased the benefit of this treatment. A large amount of *Urochloa decumbens* live biomass in Ct plots (Table S2) affected its benefit, increasing in about 3% the invasive grass biomass (from 0.39 kg.m<sup>-2</sup> at T0 to 0.40 kg.m<sup>-2</sup> at T3). For this species, herbicide+cut reduced 64% (from 0.58 kg.m<sup>-2</sup> at T0 to 0.21 kg.m<sup>-2</sup> at T3).

Table 2. Benefit (mean, kg) one year after treatments application: Cut (Ct), Herbicide (He) and Herbicide+Cut (He+Ct), from areas of 1 m<sup>2</sup> and extrapolated to larger areas of 16 m<sup>2</sup>, 5000 m<sup>2</sup> and 10000 m<sup>2</sup> in a Cerrado area under natural regeneration at the Experimental Station of Itirapina (Southeastern Brazil).

Treatment	(kg.m <sup>-2</sup> )	$(kg.16 m^{-2})$	$(kg.5000 m^{-2})$	$(kg.10000 m^{-2})$
Melinis minutiflora				
Ct	0.36	5.77	1802.13	3604.27
Не	0.20	3.17	989.47	1978.93
He+Ct	0.83	13.29	4152.23	8304.47
Urochloa decumbens				
Ct	0.01	0.16	48.93	97.87
He	0.05	0.79	246.93	493.87
He+Ct	0.20	3.13	977.00	1954.00

Considering only the costs of each treatment for the management of a 1 m<sup>2</sup> area, the cheapest treatment was herbicide, whilst the most expensive was herbicide+cut (Table 3). The difference in the cost of management (cut and herbicide+cut) for each species is related to the costs for biomass discard, particularly for *M. minutiflora*, which accumulates a large amount of biomass (Table 1). When the costs were extrapolated to larger areas (> 16 m<sup>2</sup>), herbicide + cut was still the most expensive treatment (Table 4). The added costs of more trips to the site to application and reapplication of the herbicide, followed by cut increased the cost of labor and transportation, and thus, herbicide+cut was the less economic treatment. Furthermore, the need of more of materials used for the application of herbicide (herbicide, herbicide adjuvant and dye) contributed to the increased cost of such technique.

However, the benefit/cost ratio showed that for both invasive grasses, the treatments differed only in areas larger than 5000 m<sup>2</sup> (Figure 1), and were similar in small areas (1 m<sup>2</sup> and 16 m<sup>2</sup>). When increasing the managed area (> 5000 m<sup>2</sup>), there is an increase in materials,

days of work and spends with transport, which makes herbicide more expensive than cut (Table 4), mainly because of the need of reapplication and costs of its material. Thus, for *Melinis minutiflora* (Figure 1a), in areas of 5000 m<sup>2</sup> and 10000 m<sup>2</sup>, the treatments herbicide+cut and cut showed to be the most viable in terms benefit/cost ratio. While for *Urochloa decumbens* (Figure 1b), herbicide+cut and herbicide showed similar results, considering the benefit of cut was lower than for herbicide (Table 2), which made herbicide a treatment of more benefit/cost than cut in large areas.



Figure 1. Benefit/cost ratio (kg/US\$) of the treatments: Cut (Ct), Herbicide (He) e Herbicide+Cut (He+Ct) to control (a) *Melinis minutiflora* and (b) *Urochloa decumbens* in 1  $m^2$ , 16  $m^2$ , 5000  $m^2$  and 10000  $m^2$  areas in a Cerrado area under natural regeneration invaded by both species at the Experimental Station of Itirapina (Southeastern Brazil).

				Ct	Ĩ	Ie	He+C1	
	Costs category	Price (US\$)	Total	Quant.	Total	Quant.	Total (US\$)	Quant.
Labor	Payment for 6 hours work (US\$/day)	23.26	1	23.26	7	46.51	c.	69.77
	Fuel (US\$/L)	1.34	1	1.34			1	1.34
	Oil (US\$/unit)	1.40	1	1.40			1	1.40
Materials	Herbicide (US\$/unit)	27.44			1	27.44	1	27.44
	Herbicide adjuvant (US\$/unit)	10.23	•		1	10.23	1	10.23
	Indicator dye (US\$/unit)	33.95	•		1	33.95	1	33.95
	Weed whacker (US\$/unit)	403.97	1	403.97		ı	1	403.97
Tools	Weed whacker blade (US\$/unit)	18.26	1	18.26	•		1	18.26
	Backpack herbicide sprayer (US\$/unit)	64.11	•	•	1	64.12	1	64.12
	P.P.E. kit (US\$/unit)	54.14	•		1	54.15	1	54.15
	Gasoline $(10 \text{ km} = 1 \text{*US}\$2, 89)$	1.34	1	1.34	7	2.69	ω	4.03
Transport	Driver (US\$/day)	23.26	1	23.26	7	46.51	ς	69.76
ſ	Depreciation (US\$30,860.46/1825)	16.90	1	16.90	2	8.30	ŝ	12.45
	Maintenance (US\$/day)	4.15	1	4.15	0	33.82	ω	50.73
biomass discard	Plastic bags (US\$/100 unit)	32.37	1	32.37		·	1	32.37
Total per m <sup>2</sup>				526.26		327.72		853.98

Table 3. Management cost of a 1m<sup>2</sup> area to the treatments of: Cut (Ct), Herbicide (He) and Herbicide+Cut (He+Ct) in experimental plots invaded · t Itin otal Statio at the Ev. --- ite. - והיווות с ро č .; Jan. d Hrachlad tiflo ..... by Melinis (Sol

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Table 4. Management cost (US\$) extrapolated to 5000 m<sup>2</sup> and 10000 m<sup>2</sup> the treatments of: Control (Co), Cut (Ct), Herbicide (He) and Herbicide+Cut (He+Ct) in experimental plots invaded by *Melinis minutiflora* and *Urochloa decumbens*, in a Cerrado area under natural regeneration at the Experimental Station of Itirapina. (Southeastern Brazil).

		Treatment	
	Ct	He	He+Ct
Melinis minutiflora			
$5000 \text{ m}^2$	2 656.21	2 720.89	5 386.84
$10000 \text{ m}^2$	4 890.19	5 269.10	10 178.78
Urochloa decumbens			
$5000 \text{ m}^2$	2 441.39	2 720.89	5 214.70
$10000 \text{ m}^2$	4 460.55	5 269.10	9.834.50

### DISCUSSION

The benefit/cost analysis is critical because most scientific research on invasive species has as main goal the study of their ecology, rather than the analysis of cost-effective strategies to control these species (Andreu *et al.* 2012). However, management decisions can be driven by non-ecological factors because managers typically choose actions that are the most cost efficient (Maxwell *et al.*, 2015). Therefore, this fact reinforces the importance of the results of this study and the need of economic assessment of different management techniques associated to its effectiveness.

Analysis of benefits and costs provides information on the best strategy to be followed, mainly to help decision-makers and managers of natural reserves according to the available financial resources (Born *et al.*, 2005). Resources for dealing with invasive species are limited and the monitoring of the efficiency of management techniques allows practices to be adapted to these resources more effectively (McConnachie *et al.*, 2012). The management of smaller invaded areas can be cheaper than in larger areas because managers can selectively treat invaded spots that are more accessible and less costly to control (Clout and Williams, 2009). Thus, in the short-term, this study demonstrated that in small areas of 1 m<sup>2</sup> and 16 m<sup>2</sup>, the benefit/cost ratio of the treatments are similar and can be used to control the invasive species. In this case, herbicide which was the cheapest treatment, as found by Rodrigues *et al.*, 2009 and was also effective in controlling invasive grasses (Castillioni *et al.*, Chapter 1) would be in this study the technique recommended for small areas.

In larger areas of 5000 m<sup>2</sup> and 10000 m<sup>2</sup>, herbicide+cut and cut showed to have the best benefit/cost ratio for *Melinis minutiflora* and herbicide+cut and herbicide for *Urochloa decumbens*, that means that these treatments resulted in a great reduction in efficiency (kg) of the invasive grasses per monetary unit (US\$). Although cut (for *M. minutiflora*) or herbicide (*U. decumbens*) could also be used to control the invasive species in the mentioned scale, it is important to consider that the treatments have different ecological effects. The evaluation of the treatments showed that cut is likely to stimulate the regeneration of both African grasses, whereas herbicide leads to a large accumulation of dead biomass which may affect negatively the native plant community (Castillioni *et al*, Chapter 1). On the other hand, herbicide+cut proved to be efficient in controlling both invasive species and increasing gaps, exposing the soil (Castillioni *et al.*, 1998), which may stimulate the regeneration and establishment of native species (Cabin *et al.*, 2002).

The stimulation of *M. minutiflora* and *U. decumbens* regeneration in Ct plots may consist in a threat to native vegetation because these species can displace native herbaceous species in Cerrado (Pivello *et al.*, 1999b). On the other hand, the accumulation of invasive grass in He plots means fuel material that can increase fire risk in the area (D'Antonio and

Vitousek, 1992) and may affect negatively native species, limiting their establishment and growth (D'Antonio *et al.* 1998; Cabin *et al.* 2002).

Moreover, although, a single cut could not control the invasive grasses (Castillioni *et al.*, Chapter 1), studies that assessed cost effectiveness of similar management techniques found that for *U. decumbens*, double-clipping resulted in the best benefit/cost value in areas from 100 to 500 m<sup>2</sup>, while clipping and digging showed the best ratio for *M. minutiflora* (Gorgone-Barbosa 2009).

Additionally, although our study provides information for only one year of management, it is recommended that long-term actions on the impact of management should be considered and studied before its widespread practice (Smith *et al.*, 2006). Moreover, it is recognized that in this study was only performed a preliminary analysis of the benefit/cost ratio, so it is not ignored the fact that, when extrapolating the size of the managed area, some things can change. For example, in larger areas, other methods of applying the treatments may be used, such as mechanized or semi mechanized techniques, so costs with labor and days of work would decrease. Therefore, the limitations of this study are recognized as it considers only one man working with non-mechanized techniques.

In addition, the best controlling technique depends on the target invasive species and on the situation, considering control levels of invasive plants are variable depending on climatic conditions of each region, the soil type and the physiological characteristics of invasive plants (Lorenzi, 2014). Many invasion characteristics can influence the cost of longterm management, such as dispersion pattern of the species, area size and probability of reinvasion (Epanchin-Niell and Hastings, 2010). It is also important to assess the effect of management options on native vegetation, searching for the most benefic treatment in order to safeguard native species especially in large-scale managed areas (Hulme 2006; Flory & Clay 2009) and to support restoration strategies (Ansley and Castellano, 2006).

Therefore, our study provides important information for management decisions of invasive grasses in Cerrado areas. Additionally, it reinforces the need for economic efficiency evaluation in invasive control projects so management practices can be adjusted to the use of conservation funds more effectively.

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Table S1. Classification of costs of different categories to apply the treatments: cut, herbicide and herbicide+cut in a Cerrado area under natural regeneration at the Experimental Station of Itirapina (Southeastern Brazil).

Category	Description			
Labor	Payment for 6 hours work.			
	<u>Roundup<sup>®</sup> NA Glyphosate (1 L)</u> : used 0.46 mL for 1 $m^2$			
	<u>Herbicide adjuvant (250 mL)</u> : spreading agent; used 1 mL per 0.015 $m^2$			
	<u>Indicator dye (1 L)</u> : used 0.25 ml per 1 $m^2$			
Materials	Water: for herbicide dilution; 15ml of herbicide diluted in 1L of water			
	<u>Fuel</u> : for weed whacker operation; 1L of gas to mow $64 \text{ m}^2$			
	<u>Two-stroke engine oil (200 mL)</u> : for weed whacker better functioning; used in 384 $m^2$			
	<u>Plastic bags (200 L)</u> : for biomass discard. Capacity to support up to 6kg.			
	Weed whacker (protective glasses and ear plugs are included)			
Tools/ Personal	Weed whacker blade			
Protective Equipment (P.P.E.)	Backpack herbicide sprayer (20 L)			
	<u>P.P.E. kit</u> : long pants, long sleeve shirt, hat, face shield, goggles, respirator, chemical resistant gloves and boots.			
	These costs were based on a utilitarian vehicle type pick-up (brand: Chevrolet S10 Double Cabin LS 4x2 Flexpower, year: 2013).			
Transport	<u>Fuel</u> : calculated from the estimated mileage within the protected area to the area of study and return. <u>Driver</u> : payment for 1 day drive			
	<u>Vehicle depreciation</u> : based on the vehicle price, divided by 1825 days, which is equivalent to the time of five years of depreciation of the good (Receita Federal, 2014).			
	<u>Vehicle maintenance</u> : based on the average annual cost of mechanical maintenance (SINDIREPA 2014), added to the annual cost of vehicle document taxes (property taxes (IPVA), insurance for injuries caused by motor vehicles (DPVAT) and licensing). The annual maintenance costs were divided by 365 (equivalent to one year) and converted to a daily equivalent.			

Table S2. Live and dead biomass (kg.m<sup>-2</sup>) one year after treatments application: Control (Co), Cut (Ct), Herbicide (He) and Herbicide+cut (He+Ct), from 1 m<sup>2</sup> area in a Cerrado area under natural regeneration at the Experimental Station of Itirapina (Southeastern Brazil).

Treatment	Со	Ct	He	He+Ct
Melinis minutiflora				
Live	$0.03\pm0.04$	$0.32\pm0.22$	$0.01\pm0.03$	0
Dead	$0.97\pm0.44$	$0.32\pm0.25$	$0.79\pm0.32$	$0.17\pm0.19$
Total	$1.00\pm0.47$	$0.64\pm0.41$	$0.80\pm0.30$	$0.17\pm0.19$
Urochloa decumbens				
Live	$0.06\pm0.05$	$0.13\pm0.03$	$0.03\pm0.06$	0
Dead	$0.35\pm0.18$	$0.27\pm0.09$	$0.32\pm0.25$	$0.21\pm0.09$
Total	$0.41\pm0.23$	$0.40\pm0.11$	$0.36\pm0.21$	$0.21\pm0.09$

## **CONSIDERAÇÕES FINAIS**

No primeiro capítulo desta dissertação, verificamos que, em curto prazo, os tratamentos de herbicida e combinação de herbicida seguido de corte foram os mais eficazes para o controle das invasoras *Melinis minutiflora* e *Urochloa decumbens*. Nossos resultados corroboram com estudos que comprovaram a eficácia do glifosato, entretanto é importante que se considere como seu uso é capaz de alterar a estrutura da vegetação e afetar a comunidade vegetal nativa. Isso porque com a aplicação do herbicida, houve um grande acúmulo de biomassa morta da invasora no local, o que pode consistir em um risco de incêndio na área e limitar o estabelecimento e crescimento de plântulas nativas. Além disso, foi demonstrado que o corte promoveu a regeneração das gramíneas invasoras e não seria o tratamento indicado para o controle destas espécies. Portanto, recomenda-se o tratamento de herbicida + corte, capaz de promover o controle de *Melinis minutiflora* e *Urochloa decumbens* e ainda disponibilizar mais solo nu para que outras espécies nativas possam regenerar. No entanto, considerando que a maior disponibilidade de solo nu pode também promover a reinvasão na área, sugerimos, como complemento ao manejo, a implantação de técnicas de enriquecimento com espécies nativas que promovam sua ocupação nestes espaços.

Na segunda parte do trabalho, encontramos que na análise da relação benefício/custo, todos os tratamentos apresentam similares resultados em pequena escala (1 m<sup>2</sup> e 16 m<sup>2</sup>), no entanto, em áreas maiores (5000 m<sup>2</sup> e 10000 m<sup>2</sup>), o herbicida+corte para *Melinis minutiflora* e *Urochloa decumbens* promoveram grande redução de biomassa total por unidade monetária. Estes são resultados importantes, considerando que poucos estudos realizam essa análise da relação benefício/custo de tratamentos. Com isso, o monitoramento da eficiência das técnicas de manejo por meio dos dados apresentados permite que práticas sejam adaptadas a recursos financeiros de forma mais eficaz.

Com base nestes resultados, concluímos que o herbicida + corte pode ser utilizado como tratamento eficiente no controle das gramíneas invasoras, além disso, também é um dos tratamentos de melhor benefício/custo para ambas as espécies. No entanto, é fundamental que se considere que estes resultados são referentes a um estudo de curto prazo, com aplicação dos tratamentos e monitoramento de seus efeitos por apenas um ano. Isso porque, o controle destas espécies em um ano de manejo não garante seu controle em longo prazo. Deste modo, o contínuo monitoramento é necessário, pois as sementes das gramíneas invasoras podem estar viáveis no banco de sementes, ou estas espécies podem se estabelecer novamente por meio da pressão de propágulos.

# APÊNDICE

Ilustração em (a) dos quadros utilizados de 1x1m (externo) para levantamento da cobertura vegetal e de 0,5x0,5m (interno) para amostragem de biomassa aérea e, em (b) pesquisadora realizando a coleta de biomassa em parcela invadida por *Urochloa decumbens* na Estação Ecológica de Itirapina – SP.



Parcelas experimentais (a) Controle, (b) Corte, (c) Herbicida e (d) Herbicida + Corte antes (T0) da aplicação dos tratamentos em parcelas invadidas por *Melinis minutiflora*, em uma área de Cerrado em regeneração natural na Estação Experimental de Itirapina.



Parcelas experimentais (a) Controle, (b) Corte, (c) Herbicida e (d) Herbicida + Corte antes (T0) da aplicação dos tratamentos em parcelas invadidas por *Urochloa decumbens*, em uma área de Cerrado em regeneração natural na Estação Experimental de Itirapina.



Parcelas experimentais (a) Controle, (b) Corte, (c) Herbicida e (d) Herbicida + Corte um ano (T3) após a aplicação dos tratamentos em parcelas invadidas por *Melinis minutiflora*, em uma área de Cerrado em regeneração natural na Estação Experimental de Itirapina.



Parcelas experimentais (a) Controle, (b) Corte, (c) Herbicida e (d) Herbicida + Corte um ano (T3) após a aplicação dos tratamentos em parcelas invadidas por *Urochloa decumbens*, em uma área de Cerrado em regeneração natural na Estação Experimental de Itirapina.

