

UNIVERSIDADE FEDERAL DO RIO GRANDE DO SUL  
INSTITUTO DE BIOCIÊNCIAS  
PROGRAMA DE PÓS-GRADUAÇÃO EM ECOLOGIA

TESE DE DOUTORADO

PROCESSOS INICIAIS DE RESTAURAÇÃO ECOLÓGICA EM ÁREAS  
DEGRADADAS POR MINERAÇÃO DE CARVÃO

EDILANE ROCHA-NICOLEITE

PORTO ALEGRE, SETEMBRO DE 2015

# **PROCESSOS INICIAIS DE RESTAURAÇÃO ECOLÓGICA EM ÁREAS DEGRADADAS POR MINERAÇÃO DE CARVÃO**

**EDILANE ROCHA-NICOLEITE**

**TESE DE DOUTORADO APRESENTADA AO  
PROGRAMA DE PÓS-GRADUAÇÃO EM  
ECOLOGIA, DO INSTITUTO DE  
BIOCIÊNCIAS DA UNIVERSIDADE  
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PARTE DOS REQUISITOS PARA OBTENÇÃO  
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**ORIENTADOR: PROF<sup>a</sup>. DR<sup>a</sup>. SANDRA  
CRISTINA MÜLLER  
COORIENTADOR: PROF. DR. GERHARD  
ERNST OVERBECK**

**COMISSÃO EXAMINADORA**

**PROF<sup>a</sup>. DR<sup>a</sup>. VANILDE CITADINI-ZANETTE  
PROF. DR. PAULO LUIZ DE OLIVEIRA  
PROF. DR. DEMETRIO LUIS GUADAGNIN**

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Tudo o que um sonho precisa para ser  
realizado é alguém que acredite que  
ele possa ser realizado.

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que com amor, prazer e coragem estamos  
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## **RESUMO**

Tendências atuais em restauração evidenciam a importância de propiciar o retorno das funções ecossistêmicas e ecológicas. Porém muitas áreas possuem baixo potencial para reestabelecimento de processos ecológicos devido à perda da resiliência, o que faz da restauração ecológica um processo desafiador. Para o sucesso das atividades propostas, é essencial que no decorrer do projeto os sistemas sejam capazes de retomar sua trajetória sucessional e dinâmica temporal, sem necessidade de futuras intervenções humanas. O objetivo desta tese foi avaliar os processos iniciais de restauração ecológica em áreas degradadas por mineração de carvão, através da análise de trajetórias sucessionais envolvendo: (1) taxas de crescimento e mortalidade de mudas nativas introduzidas e sua relação com atributos funcionais de plantas; (2) chuva de sementes associada, ou não, ao uso de poleiros artificiais e sua relação com regenerantes de espécies lenhosas; e (3) regeneração natural e sua relação com fatores abióticos e bióticos. Mudas de espécies nativas introduzidas sob condições limitantes (devido ao alto nível de impacto) apresentaram diferentes performances e relações com atributos, por vezes distintas do esperado para espécies sob condições naturais. A limitação de sementes nas áreas em restauração pode ser, potencialmente, reduzida pelo uso de poleiros artificiais, aumentando especialmente sementes de espécies não-pioneiras e zoocóricas. No entanto, não foi verificada relação entre a chuva de sementes e a taxa de recrutamento. Fatores abióticos, relacionados à composição química do solo, e fatores bióticos, como a presença de gramíneas exóticas, foram as variáveis com maior influência negativa sobre a regeneração natural. Este estudo evidencia a possibilidade do retornar do processo de sucessão natural em áreas de mata ciliar que foram profundamente alteradas por mineração de carvão, em um tempo relativamente curto. Entretanto, recomendamos o uso de objetivos realísticos e intermediários, bem como monitoramentos detalhados nas fases iniciais.

**Palavras-chave:** taxas de crescimento e mortalidade; chuva de sementes; poleiros artificiais; regeneração natural.

## **ABSTRACT**

In current projects of ecological restoration, the return of ecosystem functions and natural processes is an important aim. However, many areas have lost their resilience due the high damage they have been submitted to, and this makes the ecological restoration a challenging process. For the success of the proposed activities, it is essential that during the project, the ecosystem can return to its successional trajectory without necessity of strong human intervention in the future. The goal of this thesis was to evaluate the initial processes of ecological restoration in areas severally degraded by coal mining, through analyses of successional trajectories regarding: (1) growth and mortality rates of planted samplings and their relationship to functional traits; (2) seed rain, associated or not, to perches and its relationship to natural regeneration of woody species; and (3) natural regeneration of woody species and its relationship to abiotic and biotic variables. Planted saplings under limited conditions (due the high impact of mining) showed to have distinct performance and trait relationships, somewhat different to the expected for species under natural conditions. The study indicates that the limitation of seeds in areas under ecological restoration can be reduced by the use of perches, increasing especially the number of seeds of non-pioneer and zoocoric species. Nevertheless, we did not found a relation between seed rain and recruitment rate. Soil chemistry (abiotic variable) and the cover of exotic grasses (biotic variable) were the variables with the strongest negative impact on natural regeneration. We conclude that it is possible to return to successional processes in areas of riparian forest severely damaged by coal mining activities in a relatively short time, but recommend the use of realistic, intermediate restoration goals and detailed monitoring in early restoration phases.

**Key words:** growth and mortality rates; seed rain; perches; natural regeneration.

## SUMÁRIO

<b>INTRODUÇÃO GERAL.....</b>	<b>9</b>
Organização da tese.....	13
Mineração de Carvão em Santa Catarina: impactos e recuperação ambiental.....	17
Áreas de estudo.....	20
<b>CAPÍTULO 1: Can plant functional traits predict demographic rates of planted tree species in ecological restoration?.....</b>	<b>23</b>
<b>CAPÍTULO 2: Seed rain and plant establishment patterns in forest restoration on coal mining degraded areas.....</b>	<b>46</b>
<b>CAPÍTULO 3: Forest restoration after severe degradation by coal mining: lessons from the first years of monitoring.....</b>	<b>72</b>
<b>CONSIDERAÇÕES FINAIS.....</b>	<b>109</b>
<b>REFERÊNCIAS BIBLIOGRÁFICAS.....</b>	<b>112</b>

## LISTA DE FIGURAS

### INTRODUÇÃO GERAL

**Figura 1.** Aspecto geral da área A, localizada no município de Lauro Müller, SC , no primeiro (A) e no quarto ano de restauração.....21

**Figura 2.** Aspecto geral da área B, localizada no município de Lauro Müller, SC , no primeiro (A) e no quarto ano de restauração.....21

**Figura 3.** Aspecto geral da área C, localizada no município de Lauro Müller, SC , no primeiro (A) e no quarto ano de restauração.....22

**Figura 4.** Aspecto geral (A) e detalhe do interior (B) da área D, localizada no município de Lauro Müller, SC , 10 anos após o inicio do processo de restauração.....22

### CAPÍTULO 1

**Figure 1.** Significant linear models between (a) the crown growth rate and leaf nitrogen content (LNC, log transformed;  $R^2=0.19$ ;  $p=0.03$ ;  $F=5.20$ ;  $DF=17$ ) and between (b) the crown area ( $m^2$ , log transformed) and leaf phosphorus content (LPC, log transformed;  $R^2=0.27$ ;  $p=0.01$ ;  $F=7.79$ ;  $DF=17$ ). Species labels can be seen in Table 1.....33

**Figure 2.** Ordination diagram (PCA) of 19 species described by their functional traits (A) and the significant linear model (B) between the crown growth rate and the first ordination axis of the PCA ( $R^2=0.20$ ;  $p=0.03$ ;  $F=5.59$ ;  $DF=17$ ). Species labels can be seen in Table 1.....34

**Figure 3.** Linear regression between the variation of the height growth rate (HGR) and the leaf area (LA - log transformed;  $R^2=0.22$ ;  $p=0.02$ ;  $F=6.00$ ;  $DF=17$ ). Species labels can be seen in Table 1.....34

**Figure 4.** Linear regression between the relative mortality rate and the leaf dry-matter content (LDMC; log transformed;  $R^2=0.18$ ;  $p=0.01$ ;  $F=6.42$ ;  $DF=23$ ). Species labels can be seen in Table 1.....35

**Figure 5.** CCA ordination (canonical correspondence analysis) results ordered in multivariate space along the first two canonical axes ( $p=0.05$ ). The sample units are the tree species coded as showed in table 1. On the red color follows the response variables (relative mortality rate = MR; relative growth rates: HGR = height growth rate; HGRsd = height growth rate standard deviation; DGR = diameter growth rate; DGRsd= diameter growth rate standard deviation; CGR = crown growth rate; CGRsd = crown growth rate standard deviation; Crown = crown area). The blue color correspond to the functional traits (mH = Maximum adult height; LA = Leaf area; LDMC = Leaf dry matter content; LNC = Leaf nitrogen content; LPC = Leaf phosphorus content; SLA = Specific leaf area; WD = woody density).....36

## CAPÍTULO 2

**Figure 1.** Number and species richness of seeds (log-transformed) collected in seed traps with and without perches, considering all seed species (A and B) and only non-grass species (C and D). The box indicating the variability inside the lower and the upper quartiles and the vertical line (whiskers) indicate variability outside the upper and lower quartiles; the individual points indicate the outliers. Mean values of groups differed significantly ( $p<0.005$ ) for all four variables.....56

**Figure 2.** Value of community weighted mean of traits (CWM) of sampled seeds, comparing seed traps with and without perches (% of zoocoric species, A), successional group (% of pioneer species, B), and seed color (% of black seeds, C; % of white seeds, D). The box indicating the variability inside the lower and the upper quartiles and the vertical line (whiskers) indicate variability outside the upper and lower quartiles; the individual points indicate the outliers. Mean values of groups differed significantly ( $p<0.05$ ) for all variables.....57

**Figure 3.** Value of community weighted mean traits (CWM) of sampled seeds, comparing seed traps with and without perches A; seed length (mm), B: seed diameter (mm) and C: seed form (relation between length and diameter). The box indicating the variability inside the lower and the upper quartiles and the vertical line (whiskers) indicate variability outside the upper and lower quartiles; the individual points indicate the outliers. Mean values of plots A and B presented a weak relation ( $p<0.06$ ) and the mean values of plot C differed significantly ( $p<0.001$ ).....58

**Figure 4.** Relationship between patterns of seed rain and plant recruitment at the restoration sites for seed traps with perches. The co-inertia analysis shows the ordination of plots (distinguished by plot number) described by seed rain and woody plant recruitment based on abundances. Shorter arrows indicate better adjustment of patterns in both sets of data (seed rain species and recruits of woody species). The PCAs of both data sets are shown separately at the bottom (recruitment patterns at the left; seed rain patterns at the right; the species codes are in the species list: Appendix 1 for seed rain, Appendix 3 for recruits).....59

**Figure 5.** Linear regressions of woody species recruitment (number and height) and the patterns of woody species seed rain (A), exotic grasses cover (B), and forbs cover (C and D) in the plots.....60

### CAPÍTULO 3

**Figure 1.** Ordination diagram of the areas under ecological restoration (A, C and D) based on woody species of the regenerating stratum and on the proportion of herb categories (exotic grasses (*exgr*), native grasses (*nagr*) and forbs), measured through three evaluations (A, B and C areas: 10, 22 and 34 months after tree planting; D area: 6, 7 and 8 years after tree planting). The numbers identify the plots of each area at time. Most closely correlated species are shown in the respective diagram (Altr – *A. triplinervia*; Auin – *A. inulifolium*; Bagl – *B. glaziovii*; Base – *B. semiserrata*; Batr – *B. triplinervium*; Cacaj – *C. cajan*; Cegl - *C. glaziovii*; Citrus - *Citrus* sp.; Crce – *C. celtidifolius*; Eued – *E. edulis*; Eupi – *E. picturatum*; Euru – *E. rufescens*; Leau – *L. australis*; Magu – *M. guianensis*; Mica – *M. cabucu*; Myco – *M. coriacea*; Ocpu – *O. puberula*; Piad – *P. aduncum*; Pito – *P. tomentosa*; Piza – *P. zapallo*; Rico – *R. communis*; Scte – *S. terebinthifolia*; Sebr – *S. brasiliensis*; Soma – *S. mauritianum*; Sops – *S. pseudocapsicum*; Sovi – *S. viarum*; Vettw – *V. tweedieana*; Wain – *W. indica*).....83

**Figure 2.** Venn diagram with the adjusted R<sup>2</sup> results of the variance partition (RDAp) according to the subsets of variables significantly related to the response matrices (see Table 3), (Y<sub>R</sub>) considering only the composition of woody species regenerating and (Y<sub>T</sub>) considering all plots evaluated, even those without regenerating woody species, but with the addition of herbaceous categories cover as response variables.....84

**Figure 3.** Principal coordinates ordination diagram of all plots described by the abundance of woody regenerating species plus the cover of herbs categories (matrix  $Y_T$ ), according to the four studied sites. Most correlated species and herbs categories to both axes are plotted in the diagram (italic font: Anru – *A. rugulosa*; Auin – *A. inulifolium*; Base – *B. semiserrata*; Boma – *B. macrophylla*; Casi – *C. silvestris*; Cegl – *Cecropia glaziovii*; Karu – *Kaunia rufescens*; Magu – *Matayba guianensis*; Mica – *M cabucu*; Mica – *Miconia cabucu*; Mise – *bnnn*; Myco – *Myrsine coriacea*; Piad – *Piper aduncum*; Scte – *Schinus terebinthifolia*; Trmi – *T. micrantha*; Vesp – *Vernonanthura* sp.; bold font: Exgr – exotic grasses; Nagr – native grasses; Forb - forbs). Furthermore, abiotic (soil chemistry: Ca – calcium; H+Al – potential acidity; P – phosphorus; (Ca+Mg)/K – relation between that cations) and biotic variables (herbaceous species: Ursu – *U. subquadripala*; Urbi – *U. brizantha*; Urmu – *U. mutica*; Papu – *P. pumilum*; structural variables of upper stratum: Ba – basal area; Dens – density) were placed according to its axes correlation value.....85

## LISTA DE TABELAS

### CAPÍTULO 1

**Table 1.** List of species considered for the study with the number of measured individuals in the last year (N in 3<sup>rd</sup> year), their mortality rate (MR per year), and their relative growth rate considering height (RGR-H), basal diameter (RGR-D), and crown area (RGR-C), followed by their standard deviation (sd). Species without values for growth rates were not included in the analysis due to the low number of individuals (see methods).....32

**Table 2.** Spearman correlation coefficients between functional traits and relative growth rate or its standard deviations (sd), considering plant height (RGR-H), basal diameter (RGR-D) and crown area (RGR-C). Additionally, we considered crown area ( $m^2$ ) in the last evaluation. Significant values are in bold ( $p<0.01=**$ ,  $p<0.05=*$ , and  $p<0.10$ ).....33

### CAPÍTULO 2

**Table 1.** Overview of the seed rain, in seed traps with and without perches, considering percentage of grasses and non-grasses in terms of seedling number, number of seeds per  $m^2 \cdot year^{-1}$  and species richness of the seed rain.....55

## **LISTA DE APÊNDICES**

### **CAPÍTULO 1**

**Appendix 1.** List of species considered for the study with their species code and the number of measured individuals to access the functional traits (N): leaf area (LA); specific leaf area (SLA); leaf dry-matter content (LDMC); leaf nitrogen concentration (LNC); leaf phosphorous concentration (LPC); maximum height (mH); wood density (WD).....44

**Appendix 2.** Ordination diagram (PCA) of 25 species described by their functional traits (A) and the significant linear model (B) between the relative mortality rate and the first ordination axis of the PCA ( $r^2=0.13$ ;  $p=0.03$ ;  $F=4.89$ ;  $DF=23$ ).....45

### **CAPÍTULO 2**

**Appendix 1.** Species collected on the seed rain sampling recorded in areas process of ecological restoration.....68

**Appendix 2.** Abundance of seed rain of exotic, native and doubtful species and the richness of seeds.....70

**Appendix 3.** Species collected on the wood natural regeneration (seedling recruits) in areas process of ecological restoration.....70

**Appendix 4.** Relationship between patterns of seed rain and plant recruitment at the restoration sites for seed traps without perches ( $RV=0.46$ ;  $p=0.16$ ). The co-inertia analysis shows the ordination of plots (distinguished by plot number) described by seed rain and woody plant recruitment based on abundances. Shorter arrows indicate better adjustment of patterns in both sets of data (seed rain species and recruits of woody species). The PCAs of both data sets are shown separately at the bottom (recruitment patterns at the left; seed rain patterns at the right; the species codes are in the species list: Appendix 1 for seed rain, Appendix 3 for recruits).....71

## CAPÍTULO 3

<b>Appendix 1.</b> Species of the regenerating woody species recorded in areas in process of ecological restoration, considering the last evaluation (areas A, B and C: 34 months after initial intervention with tree planting; areas D: 8 years after initial intervention with tree sowing). Given is abundance per area (standardized to individuals per 100 m <sup>2</sup> ) at each study area.....	94
<b>Appendix 2.</b> Species sampled in the woody regenerating stratum at the reference areas R2 and R1 given is abundance per area (standardized per 100 m <sup>2</sup> ).....	97
<b>Appendix 3.</b> Ecological traits of woody species sampled in the regeneration stratum at the restoration sites (A, B, C and D), considering the last evaluation (areas A, B and C: 34 months after initial intervention with tree planting; area D: 8 years after initial intervention with tree sowing) and at remnant forests (average of values recorded at the two reference areas).....	100
<b>Appendix 4.</b> Species sampled in the upper stratum (dhb>5 cm) of area D, given is abundance standardized per 100 m <sup>2</sup> .....	101
<b>Appendix 5.</b> Species sampled on the upper stratum of the reference areas R2 and R1, given is abundance per area (standardized to individuals per 100 m <sup>2</sup> ).....	102
<b>Appendix 6.</b> Species sampled in the herbaceous stratum at the areas under ecological restoration, given is mean cover per area.....	105
<b>Appendix 7.</b> Cover of herb categories, given are mean cover and standard deviation (SD) per area under ecological restoration.....	107
<b>Appendix 8.</b> Species sampled in the herbaceous stratum at the reference areas, given is mean cover.....	107
<b>Appendix 9.</b> Subset of variables selected from each set of predictor variables (based on adjusted R <sup>2</sup> value) to explain Y <sub>R</sub> (only plots with presence of regenerating woody species) or Y <sub>T</sub> (all plots described by regenerating woody species and the cover of herbs categories). All variables were significant at a level of p<0.005.....	108

## **INTRODUÇÃO GERAL**

A Sociedade Internacional de Restauração ecológica (SER 2004) define o termo restauração como o processo que auxilia a recuperação de um ambiente alterado, nos mais diferentes níveis, a fim de recriar um ecossistema sustentável e resiliente, muito semelhante ao natural. Este conceito, foi adotado para o desenvolvimento deste estudo. Tendências atuais em restauração evidenciam a importância de propiciar o retorno das funções ecossistêmicas e ecológicas (Miller & Hobbs 2007; Schrama *et al.* 2012), o que pode ser realizado de duas formas principais, restauração ativa e passiva, sendo que a escolha entre uma ou outra é baseada principalmente no grau de impacto ambiental que a área está exposta.

Algumas áreas, mesmo após diversos impactos ambientais, possuem potencial para reestabelecimento da vegetação natural sem a necessidade de intervenções diretas, processo conhecido como restauração passiva (Holl & Aide 2011). Uma das vantagens da restauração passiva é o reduzido custo, relacionado apenas as atividades de manutenção e manejo, quando necessárias. No entanto, a decisão entre qual estratégia deve ser adotada (se ativa ou passiva) depende dos objetivos estabelecidos e da resiliência natural da área, embora as limitações orçamentárias também influenciem na escolha (Bradley *et al.* 2010; Holl & Aide 2011).

Muitas áreas possuem baixo potencial para reestabelecimento de processos ecológicos devido à perda da resiliência (Kanowski & Catterall 2003; Chazdon 2008), tornando a restauração ativa necessária. As intervenções incluem desde construção topográfica ou de canais de rios até a reintrodução de plantas e propágulos ou manipulação ativa de regimes de distúrbios, como fogo por exemplo (Holl & Aide 2011). O plantio de espécies nativas é o método comumente empregado para sistemas florestais (Omeja *et al.* 2011; Peña-Domene *et al.* 2013), embora, além de oneroso, traga consigo inúmeros desafios, relacionados ao

desenvolvimento de espécies nativas que possuem condições específicas de desenvolvimento, muitas das quais ainda desconhecidas e com inúmeras interações ecológicas.

Para o sucesso da restauração, é essencial que os sistemas sejam capazes de retomar sua trajetória sucessional e dinâmica temporal, sem necessidade de futuras intervenções humanas, momento em que o sucesso do projeto é atingido (Block *et al.* 2001). Para isso tem sido priorizado o aumento da riqueza de espécies introduzidas, a escolha de espécies facilitadoras, a condução da regeneração natural, entre outros métodos que visam o retorno das funções naturais (Martínez-Garza & Howe 2003; Chazdon *et al.* 2007; Viers *et al.* 2012). Estes conceitos têm substituído modelos e métodos determinísticos baseados, apenas, em práticas silviculturais, com reduzido número de espécies (Martínez-Garza & Howe 2003; Lima *et al.* 2009; Rodrigues *et al.* 2011).

Atualmente, plantios com alta diversidade de espécies nativas têm sido recomendados, por acreditar-se que promovem mudanças das condições microclimáticas, aumento da complexidade estrutural da vegetação e o desenvolvimento das camadas de serapilheira (Vieira & Gandolfi 2006). As plantas introduzidas ou em regeneração, podem conduzir o processo de restauração e sua velocidade, seja através de interações ecológicas ou das taxas de crescimento (Temperton & Zirr 2004; Cheung *et al.* 2009; Muñiz-Castro *et al.* 2012).

Um dos benefícios associados ao plantio de espécies nativas é a capacidade de acelerar a cobertura de copas no ambiente (maior sombreamento), criando microambientes florestais que contribuem para o crescimento e sobrevivência de plântulas, com a diminuição da competição com espécies herbáceas. Além disso, a cobertura de copas no sistema auxilia os processos de decomposição no solo e de oxidação da matéria orgânica, controlando processos erosivos e outras funções ecossistêmicas (Kanowski *et al.* 2003; Melo *et al.* 2007; Muñiz-Castro *et al.* 2012).

No entanto, estudos sobre a estrutura fisionômica e funcional de florestas restauradas com plantio de espécies nativas ainda são escassos, constituindo uma grande lacuna de conhecimento em relação à avaliação dos processos associados à restauração, tendo em vista a complexidade das questões envolvidas (Lima *et al.* 2009). Também cabe destacar que a heterogeneidade dos ambientes em restauração dificulta comparações, pois cada área tem suas peculiaridades e está exposta a processos dinâmicos naturais e antrópicos, resultando no recrutamento de diferentes espécies de plantas com cenários de regeneração distintos (Klein *et al.* 2009).

Além do simples plantio de mudas de espécies nativas, outros métodos associados a melhoria microclimática devem ser priorizados, visando a revitalização de processos criadores de condições específicas, para que a dinâmica natural de espécies nativas seja reestabelecida no ambiente perturbado. Neste sentido, a criação de núcleos de regeneração através de técnicas que estimulem a resiliência do ambiente, podem ser fundamentais, tal como, o uso de poleiros artificiais para atração e aumento da chuva de sementes (Reis *et al.* 2010).

Estes métodos de atração de dispersores e chuva de sementes são essenciais para a restauração de sistemas florestais tropicais, onde a principal forma de regeneração natural é por meio da chuva de sementes, a qual depende da distância de áreas fonte de propágulos e dos bancos de sementes do solo e de plântulas, que por ventura permaneceram nos sítios degradados (Cheung *et al.* 2009). As diferentes maneiras como os diásporos são dispersos e a frequência com que atingem ambientes favoráveis para o estabelecimento da plântula é que determinam a riqueza e a distribuição espacial das populações de plantas (van der Pijl 1972). Ao mesmo tempo, a comunidade vegetal em estabelecimento (espécies em regeneração espontânea e introduzidas) é altamente dependente das condições físicas e químicas do solo que, se não forem adequadas, limitam seu desenvolvimento (Lima *et al.* 2012), tornando a restauração uma atividade extremamente complexa. Neste sentido, fatores abióticos associados à regeneração

natural, principalmente aqueles ligados às condições físicas e químicas do solo, também são determinantes para o estabelecimento e desenvolvimento de plântulas e, consequentemente, dos padrões de recrutamento de espécies (Marimon *et al.* 2010; Catterall *et al.* 2012).

Ferreira *et al.* (2010) afirmam que mesmo em áreas onde o solo sofreu intensas alterações físicas, a regeneração natural pode contribuir com mais de 50% da diversidade de espécies após seis anos de plantio, desde que inicialmente sejam introduzidas espécies arbóreas com potencial de facilitação. A facilitação promovida por espécies arbóreas está relacionada, principalmente, à cobertura das copas, à atratividade de dispersores e à melhoria das condições abióticas na escala local (Remor 2004).

Outro desafio importante da ecologia de restauração reside em como avaliar o sucesso da restauração ecológica e transformar os resultados obtidos em subsídios para futuros projetos, potencializando acertos que venham propiciar sustentabilidade temporal aos sistemas restaurados, de modo similar aos naturais (Block *et al.* 2001; Viers *et al.* 2012). A heterogeneidade dos ambientes em restauração dificulta comparações, pois cada ambiente está atrelado a diferentes históricos de degradação, diferentes contextos de paisagem, diferentes técnicas utilizadas, entre outros (Holl & Aide 2011), o que resulta em particularidades nos processos naturais e antrópicos que conduzem a diferentes cenários de regeneração (Hobbs *et al.* 2011).

Uma forma para avançar na restauração é conhecer os pontos críticos e fatores chaves das metodologias aplicadas, possibilitando o desenvolvimento de uma base de dados que permita avaliar e refinar as estratégias prescritas para a restauração de áreas degradadas e a correção de eventuais problemas, bem como o estabelecimento de parâmetros robustos para avaliação do sucesso da restauração (Brancalion *et al.* 2009). Ou seja, o monitoramento e ações de *feedback* devem fazer parte de qualquer ação de restauração.

## *Organização da Tese*

O conhecimento de processos que atuam nas fases iniciais da restauração pode fornecer resultados que subsidiem o manejo adaptativo contribuindo para o sucesso do projeto, e ao mesmo tempo avaliando a sua trajetória de acordo com as teorias ecológicas aplicadas, gerando também um aprofundamento teórico. Com uma abordagem integradora, associada a padrões de trajetórias sucessionais, esta tese de doutorado está estruturada em 3 capítulos que relacionam aspectos aplicados à restauração ecológica de matas ciliares em áreas severamente impactadas pela mineração de carvão, no sul do Brasil.

Estes impactos causados pela mineração de carvão são de abrangência regional e estão associados a áreas submetidas a intervenções ativas de restauração com reduzida resiliência. Independente do cenário, objetivou-se construir um desenho que possibilitasse a avaliação de processos ecológicos associados à restauração florestal como um todo, que em geral passa pelo plantio de espécies arbóreas nativas, visando a melhoria das condições microclimáticas locais e o favorecimento dos demais processos sucessionais necessários para as fases iniciais da restauração.

No capítulo 1 é apresentado o manuscrito “**Can plant functional traits predict demographic rates of planted tree species in ecological restoration?**”, que traz uma abordagem de taxas vitais (crescimento e mortalidade) das espécies introduzidas como mudas, e sua relação com atributos funcionais das plantas. O plantio de espécies nativas para restauração ecológica não é realizado apenas com o objetivo de reestruturar o ambiente florestal, como acreditava-se no passado onde eram preconizados modelos determinísticos (Rodrigues *et al.* 2011). As espécies introduzidas possuem importante papel na melhoria das condições locais, como microclima, propriedades do solo, e fornecimento de abrigo e recursos alimentares para dispersores (Parrotta *et al.* 1997; Holl *et al.* 2000; Martínez Ramos & Orth

2007). Assim, o sucesso no estabelecimento das espécies florestais introduzidas é fundamental para ocorrência de outros processos ecológicos, como aumento da chuva de sementes, da regeneração natural e da recolonização da fauna local, os quais garantirão a sustentabilidade do projeto em longo prazo (Ribeiro *et al.* 2009).

Diversos estudos indicam uma relação entre as taxas de crescimento e mortalidade de plantas e seus atributos funcionais e estes vêm sendo amplamente estudados por potencializarem uma resposta global para a performance individual das espécies, associadas às comunidades vegetais (Poorter *et al.* 2008; Wright *et al.* 2010). No entanto, poucos estudos avaliaram a relação entre atributos funcionais de plantas e o desenvolvimento das espécies introduzidas para restauração ecológica (Poorter *et al.* 2008; Martínez-Garza *et al.* 2013; Andrade *et al.* 2014). Considerando a variação dos atributos funcionais de plantas e sua resposta nas taxas de crescimento e sobrevivência das espécies introduzidas (Violle *et al.* 2007), esta relação pode ser uma importante ferramenta para prever trajetórias de comunidades em restauração ecológica contribuindo também para o manejo e monitoramento do projeto.

Assim, o objetivo deste capítulo foi avaliar as taxas de crescimento das plantas nativas introduzidas através dos parâmetros de altura, diâmetro basal e área da copa, bem como, sua relação com atributos funcionais das plantas, de áreas em processo de restauração ecológica de áreas severamente impactadas por mineração de carvão. Este capítulo teve como questão central: Atributos funcionais de plantas predizem taxas de crescimento e de mortalidade de plantas introduzidas para restauração ecológica de áreas intensamente degradadas?

A chegada e o estabelecimento de espécies nativas é o processo que potencializa a auto-mantenção de áreas em restauração (Chazdon 2008). Neste sentido estruturamos o capítulo 2 “**Seed rain and plant establishment patterns in forest restoration on coal mining degraded areas**”, onde foi avaliada a contribuição de poleiros artificiais como técnica adicional ao aumento de propágulos na área. Além da avaliação da chuva de sementes através de coletores

com e sem poleiros artificiais, também foi avaliada a regeneração natural de espécies lenhosas (arbustos e árvores) e a cobertura de plantas herbáceas, bem como as relações entre estas variáveis e os padrões da chuva de sementes.

Na restauração de ecossistemas florestais tropicais, o principal filtro que limita a regeneração natural é a baixa disponibilidade de sementes, frequentemente associada à falta de dispersores e de banco de sementes no solo (Holl 1999; Chazdon 2003; Florentine & Westbrooke 2004; Baur 2014). O uso de poleiros artificiais tem sido relatado como uma ferramenta adicional para atrair dispersores (Graham & Page 2012; Heeemann *et al.* 2012; Cavallero *et al.* 2013), formando núcleos onde a vegetação poderia se estabelecer mais rapidamente e assim diminuindo as consequências do filtro associado à baixa disponibilidade de propágulos.

É fundamental que a chegada de sementes promovida pelo uso de poleiros, porém, também esteja associada ao aumento nas taxas de recrutamento de espécies lenhosas, para que esta estratégia seja de fato favorável (Graham & Page 2012). Assim, o objetivo deste capítulo foi avaliar a riqueza, a composição e a densidade da chuva de sementes, em termos de espécies e grupos funcionais, e suas relações com o recrutamento de espécies lenhosas. Considerando que a presença de gramíneas exóticas invasoras é considerada um outro fator limitante importante ao processo de restauração, por impedir ou retardar o recrutamento de espécies nativas (Martinez-Ramos & Soto-Castro 1993; Ortega-Piecka *et al.* 2011), também se avaliou a relação entre o recrutamento e a cobertura de plantas herbáceas nas categorias de gramíneas nativas e exóticas, bem como outras herbáceas (*forbs*).

Uma das metas recorrentes de projetos de restauração ecológica que utilizam técnicas ativas de intervenção é obter a auto-manutenção do sistema e seus processos sem o contínuo auxílio humano, o que assegura o sucesso da restauração ecológica no tempo. Para tanto, um dos processos fundamentais para esta trajetória é a regeneração natural, que ocorre

de forma passiva. Assim, foi estruturado um terceiro capítulo “**Forest restoration after severe degradation by coal mining: lessons from the first years of monitoring**”, onde são explorados os padrões de regeneração natural e suas relações com fatores bióticos e abióticos. O estabelecimento de uma comunidade de plantas que se assemelhe à natural consiste em um desafio devido a diversas limitações relacionadas ao estabelecimento de espécies nativas, especialmente em áreas altamente impactadas (Fields-Johnson *et al.* 2012; Zhenqi *et al.* 2012; Bauman *et al.* 2013). O objetivo deste capítulo foi avaliar a regeneração natural de arbustos e árvores em áreas em processo de restauração ecológica após severo impacto ambiental associado à mineração de carvão. Paralelamente foram analisados parâmetros abióticos (química e estrutura do solo) e bióticos associados a regeneração de espécies lenhosas.

Em situações onde houve mudança completa das condições naturais, como em áreas de mineração, por exemplo, o substrato não apresenta suas características originais de estrutura física e química e, frequentemente, encontra-se altamente impactado (Kämpf *et al.* 1997; Bauman *et al.* 2013). Ainda, a presença de espécies exóticas invasoras pode constituir uma barreira adicional ao estabelecimento de uma nova comunidade (Nyamai *et al.* 2011; Bennett *et al.* 2012). Assim, a trajetória sucesional estabelecida no início das atividades de restauração, descrita por padrões de regeneração natural, pode variar amplamente de acordo com as características locais relacionadas tanto a fatores abióticos quanto bióticos (Block *et al.* 2001).

Com o desenvolvimento destes capítulos, buscou-se explorar diferentes aspectos da restauração ecológica com base em projetos que estão sendo aplicados para restauração de áreas de mata ciliar em uma região intensamente degradada pela mineração de carvão, vinculando assim os processos ecológicos à prática que tem sido aplicada. Neste sentido, a coleta de dados de campo foi realizada em áreas em processo de restauração ecológica, porém sem instrumentação e/ou controle das técnicas que foram empregadas. A construção desta tese foi, então, com base na avaliação de diversos parâmetros e no monitoramento de projetos

implementados por empresas legalmente responsáveis pela restauração de algumas áreas da região.

### ***Mineração de Carvão em Santa Catarina: impactos e recuperação ambiental***

No Brasil, as principais jazidas de carvão mineral localizam-se na Região Sul, nos estados de Paraná, Santa Catarina e Rio Grande do Sul. A Bacia Carbonífera de Santa Catarina, ocupa, aproximadamente, 100 km de comprimento e 20 km de largura, entre a Serra Geral a Oeste e o maciço granítico da Serra do Mar a Leste, seguindo a orientação Norte-Sul (Krebs 2005). Com a forte demanda por carvão mineral, a partir de 1970, a exploração passou a ser realizada em grandes minas, ocasionando maior contaminação do meio ambiente (Gomes *et al.* 2008).

Para compreensão do impacto originado por esta atividade, cabe esclarecer os métodos empregados à mineração de carvão, que são a lavra subterrânea e a céu aberto. A lavra subterrânea, é utilizada quando a jazida de carvão se encontra em camadas mais profundas (superior a 30 m de profundidade), e os principais impactos ao ecossistema são relacionados à deposição de rejeitos da mineração na superfície, após o beneficiamento do minério (Koppe & Costa 2008).

Já a lavra a céu aberto, realizada quando a jazida de carvão se encontra próxima à superfície do solo (até 30 m de profundidade), envolve a remoção de grandes volumes de rochas removidas para acesso às camadas de carvão (denominadas estéril da mineração) para cada tonelada de carvão produzida. Este método causa a formação de cavas para acesso à camada de carvão e pilhas de material resultante da escavação (Koppe & Costa 2008), geralmente cônicas com até 20 m de altura, caracterizando a inversão dos horizontes do solo e das rochas, em

relação às suas posições estratigráficas originais, com vegetação e solo depositados na base e estéreis da mineração nas camadas superiores (Citadini-Zanette 1999; Lopes *et al.* 2009).

Com a disposição sem controle dos estéreis e dos rejeitos, o solo não foi preservado e muitas áreas foram abandonadas após a mineração, ocasionando diversos problemas ambientais, como a geração de drenagem ácida de mina (DAM), erosão, emissão de gases para atmosfera e impacto visual (Koppe & Costa 2008). Entre estes problemas, o mais sério é a DAM, resultante da oxidação da pirita e à presença de outros sulfetos presentes no carvão e também encontrados nos rejeitos do beneficiamento. Como consequência desse processo ocorre a formação de água extremamente ácida (pH inferior a 2,0) e enriquecida com ferro, alumínio, sulfato e metais pesados, tais como chumbo, manganês e cádmio (Silva *et al.* 2011).

Esta intensa contaminação originou impactos ambientais em três bacias hidrográficas (bacia do Rio Araranguá, do Rio Tubarão e do Rio Urussanga), com aproximadamente 6.500 ha em superfície (Brasil 2012b), causando também sérios problemas de saúde pública associada à população que vive no entorno destas áreas.

Visando a enfrentar o passivo ambiental decorrente da mineração de carvão, o Ministério Público Federal propôs uma ação civil pública (processo 93.8000533-4) em 1993, perante a Justiça Federal de Santa Catarina. Mais tarde, em 2000, foi proferida a sentença, na qual condenou os réus (empresas carboníferas e a União), solidariamente, a apresentarem projetos de recuperação ambiental para a região que compõe a bacia carbonífera catarinense (JFSC 2015).

Para otimizar a execução da sentença, em 2006 criou-se o Grupo Técnico de Assessoramento ao Juízo (GTA), que se constitui um inovador instrumento de autogestão, com representantes técnicos de todas as partes e também com a presença de pessoas externas ao processo e relevantes à questão ambiental (JFSC 2015). Este grupo foi responsável pela elaboração de um documento que especifica como devem ser realizadas as atividades de

recuperação ambiental na região chamado “Critérios técnicos para a recuperação e para a reabilitação das áreas degradadas” (Brasil 2013).

Este documento traz importantes definições, dentre as quais se destaca a distinção entre Áreas de Preservação Permanente (APP) e demais áreas (Não APP), com diferentes recomendações, considerando a importância ambiental de cada local. Considerando as definições já recomendadas por lei (Brasil 2012a), bem como a maior sensibilidade ambiental e sua importância ecossistêmica, definiu-se que todas as áreas de APP devem ser recuperadas visando a formação de floresta característica do bioma Mata Atlântica (no âmbito deste estudo, a definição considerada restauração).

No cenário de degradação ambiental ao qual a região está exposta, muitas áreas de APP, especialmente matas ciliares, estão completamente desprovidas de suas características originais, com deposição de material contaminante nas margens dos rios e, por vezes no próprio leito (Rocha-Nicoleite *et al.* 2013). Assim, para a restauração destas áreas, recomenda-se que todo o material contaminante seja removido, possibilitando a reconstrução do solo em um nível que permita a implantação de espécies arbóreas nativas e o restabelecimento de floresta típica do Bioma Mata Atlântica (Brasil 2013).

Visando atender a sentença e as recomendações do Ministério Público Federal e da Justiça Federal, os réus responsáveis pela recuperação das áreas têm estabelecido e executado projetos de recuperação e restauração ecológica, embora ainda carentes de técnicas, e especialmente de avaliações e monitoramento relacionando prática e teoria.

Portanto, a região de mineração de carvão do estado de Santa Catarina, foco desta tese, apresenta sérios problemas ambientais, com áreas desprovidas de vegetação florestal, e com solos e recursos hídricos altamente impactados. Problemas estes que são globais, uma vez que as atividades de mineração são extremamente impactantes e distribuem-se ao redor do mundo, com complexidade semelhante à encontrada no sul de Santa Catarina (Campos *et al.*

2003; Zipper *et al.* 2011; Zhenqi *et al.* 2012). Frente a esta problemática, e considerando a necessidade de estudos que relacionem prática e teoria no campo da ecologia da restauração, estabeleceu-se este cenário como foco de estudo desta tese.

### **Áreas de estudo**

Para este estudo foram selecionadas quatro áreas de mata ciliar que estão em processo de restauração ecológica, com histórico de uso e intervenções semelhantes, aqui denominadas A, B, C e D, e dois remanescentes florestais, denominados LM e SD, como áreas de referência. As áreas A, B, C e LM localizam-se no município de Lauro Müller, na bacia hidrográfica do rio Tubarão, com altitude de aproximadamente 220 m acima do nível do mar. As áreas D e SD localizam-se em Siderópolis, na Bacia Hidrográfica do rio Araranguá, com altitude de aproximadamente 120 m acima do nível do mar. A distância entre os dois locais é de aproximadamente 20 km. A região de estudo está inserida no bioma Mata Atlântica (Ribeiro *et al.* 2009), possui clima Subtropical Úmido com Verão Quente (Cfa) segundo a classificação de Köppen (1948), com temperatura média anual de 19°C e a precipitação média anual de 1.600 mm.

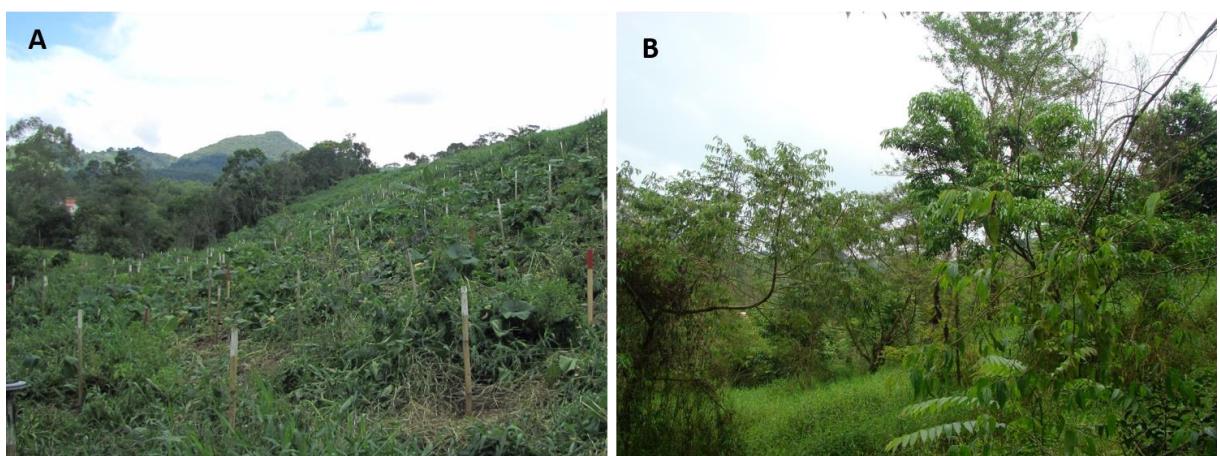
Em todas as áreas em restauração houve mineração de carvão à céu aberto e deposição de materiais contaminantes, sendo que para restauração, houve remoção destes materiais, construção do solo e revegetação. Para a revegetação, foram introduzidas gramíneas exóticas anuais (áreas B e C) e perenes (áreas A e D), seguida da introdução de espécies arbóreas nativas, através de semeadura direta de *Mimosa scabrella* Benth. e introdução de 20 núcleos com seis espécies arbóreas nativas em cada núcleo (área D) ou através do plantio de mudas de espécies arbóreas nativas com densidade de 2.500 mudas/ha (áreas A, B. e C). A riqueza de espécies arbóreas introduzidas foi de 30 espécies nas áreas A, B e C e de 16 espécies

na área D. Estas intervenções foram realizadas entre 2009 e 2010 nas áreas A, B e C (período da exploração de carvão: 1997), e entre 2002 e 2005 no caso da área D (exploração de carvão entre 1950 e 1989). As áreas foram avaliadas por um período de quatro anos, e foram amostradas conforme as metodologias descritas em cada capítulo. Um aspecto geral das áreas estudadas nesta tese pode ser verificado nas figuras 1, 2, 3 e 4.

As áreas utilizadas como referência, são áreas sem histórico conhecido de utilização e, portanto, em estágio avançado de regeneração natural, embora situadas em um contexto de fragmentação da paisagem.



**Figura 1.** Aspecto geral da área A, localizada no município de Lauro Müller, SC , no primeiro (A) e no quarto ano de restauração.



**Figura 2.** Aspecto geral da área B, localizada no município de Lauro Müller, SC , no primeiro (A) e no quarto ano de restauração.



**Figura 3.** Aspecto geral da área C, localizada no município de Lauro Müller, SC , no primeiro (A) e no quarto ano de restauração.



**Figura 4.** Aspecto geral (A) e detalhe do interior (B) da área D, localizada no município de Lauro Müller, SC , 10 anos após o inicio do processo de restauração.

# CAPÍTULO 1

**CAN PLANT FUNCTIONAL TRAITS PREDICT DEMOGRAPHIC  
RATES OF PLANTED TREE SPECIES IN ECOLOGICAL  
RESTORATION?**



# **Can plant functional traits predict demographic rates of planted tree species in ecological restoration?**

Edilane Rocha-Nicoleite<sup>1,2</sup>; Gerhard Ernst Overbeck<sup>1,3</sup>; Sandra Cristina Müller<sup>1,4</sup>

1 – Graduate Program in Ecology, Universidade Federal do Rio Grande do Sul, Av. Bento Gonçalves 9500, 91501970, Porto Alegre, RS, Brazil.

2 – Clean Coal Research Centre, Associação Beneficente da Indústria Carbonífera de Santa Catarina, Rua Pascoal Meller 73, 88805-380, Criciúma, SC, Brazil, (e-mail: edilane.rocha@satc.edu.br).

3 – Department of Botany, Universidade Federal do Rio Grande do Sul, Av. Bento Gonçalves, 9500, 91540-000 Porto Alegre, Brazil.

4 – Department of Ecology, Universidade Federal do Rio Grande do Sul, Av. Bento Gonçalves, 9500, 91540-000 Porto Alegre, Brazil.

## **Abstract**

In ecological restoration, native trees are planted to accelerate vegetation recovery, to improve site conditions and to facilitate natural regeneration for long-term restauration. In the context of a restoration project of a site degraded by coal mining in southern Brazil, we aimed to evaluate the relative growth rates (based on height, crown and stem base diameter) and mortality rate for each specie, and the relationship of these features to principal plant functional traits. Once a year, during three years, we evaluate 685 individuals belonging to 25 species and 15 botanical families. Relations between relative growth rates, mortality rate and functional traits were investigated by help of Spearman's rank correlation and by multiple linear regression models. Principal canonical analysis and Canonical correspondence analysis were applied to show multi-trait patterns. Regression models of RGR of crown area and crown size were significant when using the traits leaf nitrogen (LNC) and leaf phosphorus content (LPC), respectively. Relative growth rates related to stem basal diameter and height did not presented significant relation to any functional trait. The mortality rate responded significantly when

using leaf dry-matter content as predictor variable, with a positive relation. Our study showed that some traits could be related to growth and mortality rates of young trees, but the way what such relations occurred were still unclear and contrary to what is expected based on the literature. Facing this, we believe that more and broader studies encompassing vital rates and functional traits of species are needed in order to make use of the results of theoretical ecology to practical ecological restoration.

**Key-words:** relative growth rate; mortality rate; native tree plantation; coal mining restoration.

## **Introduction**

Areas affected by severe degradation, such as by coal mining, need to be actively restored due to the very low potential to return to previous conditions without any assistance (Bradley *et al.* 2010; Holl & Aide 2011). In forest regions, the most common method for ecological restoration is planting of native trees (Omeja *et al.* 2011; Peña-Domene *et al.* 2013). The objectives of tree plantings go well beyond the vegetation component itself. Introduced trees play an important role for the improvement of site conditions (microclimate, soil properties) and can provide habitat and resources for fruit-eating animals, which may contribute to seed dispersal (Parrotta *et al.* 1997; Holl *et al.* 2000; Martínez Ramos & Orth 2007). These processes, however, depend on the successful establishment and canopy development of trees by selection of trees with rapid growth thus can be important for restoration projects (Melo *et al.* 2007; Ribeiro *et al.* 2009; Omeja *et al.* 2011).

Plant functional traits have been employed worldwide as useful parameters to predict plant fitness and plant growth abilities (Poorter *et al.* 2008; Wright *et al.* 2010). However, only few studies evaluate the relation between plant traits and plant development in ecological restoration (Poorter *et al.* 2008; Martínez-Garza *et al.* 2013; Andrade *et al.* 2014). Functional traits are any morphological, physiological or phenological features that can be measured at the individual level, and they impact plant fitness via their effects on growth, reproduction and survival (Violle *et al.* 2007). As functional response traits vary among species and may influence ecosystem functions (Díaz *et al.* 2004), working with traits in ecological restoration can be useful to forecast trajectories of plant communities and to improve project management and monitoring (Asanok *et al.* 2013; Martínez-Garza *et al.* 2013). This also is true for seeded or planted species, due to their importance for shaping ecological processes at restoration sites (Laughlin 2014a). Quantifying the relation of functional traits with growth and

survival of plants under different restoration conditions may be useful for the selection of species with better performance in the field.

Several key traits have been proposed to be universally important for plant performance and to represent relatively independent aspects of plant ecological strategies (Poorter *et al.* 2008). Among these, the following seven functional traits related to resource acquisition and allocation patterns can be highlighted: leaf surface area (LA), specific leaf area (SLA), leaf nitrogen (LNC) and phosphorus (LPC) content, leaf dry-matter content (LDMC), wood density (WD), and potential plant height ( $H_{max}$ ). These traits represent whole-plant traits, leaf traits and stem traits (Pérez-Harguindeguy *et al.* 2013) and seem to express three different dimensions in terms of plant form and performance (Laughlin 2014b). They have been directly related to environmental conditions, especially considering light, soil and water features of the sites, and the ability of plants in establishment and persistence (Westoby & Wright 2006). Leaf trait states can compose a ‘leaf economics spectrum’ (Wright *et al.* 2004) that varies from species with fast growth (high SLA, LNC, LPC), i.e. with the potential for quick returns of investments of nutrients and dry mass in leaves to slow growing species with long leaf lifespan (low SLA, LNC, LPC, high LDMC). This turnover is associated to photosynthetic rates and herbivore resistance (Westoby & Wright 2006; Laughlin 2014a). Leaf area, besides its influence on light interception, is further related to the maintenance of favorable leaf temperatures and to water-use efficiency in conditions of high solar radiation and low water availability (Ackerly *et al.* 2002), which are common limitations in early phases of ecological restoration. Wood density represents a trade-off between growth rate and survival and also can be interpreted in terms of resistance to decay, storage capacity and mechanical strength (Wright *et al.* 2010).

In this study, we aimed to evaluate the relative growth rates (RGR) and the mortality rate (MR) of planted tree species used in active restoration of areas severely degraded

by coal mining and their relationships to plant functional traits. Our central question was if plant functional traits were good predictors of RGR and MR of planted tree species. We predicted that (1) species with higher LA, SLA, LNC and LPC should present higher demographic rates due to their potential of rapid resource acquisition (fast-growth species), while (2) species with higher WD and Hmax should present a higher mortality rate as they are often associated to shade tolerant species (late successional) and should not perform well the under environment with high light availability at our restoration site. Trees planted in restoration areas often are under very specific, often limiting, soil conditions or may suffer severely from competition with grasses, which may also affect growth rates (van Breugel *et al.* 2011; Campoe *et al.* 2014). Thus, we were further interested to understand which trait values characterize species with higher variability in their demographic rates, which may be indicative of greater tolerance of high variability of abiotic conditions in restoration sites.

## Methods

### *Study site*

This study was conducted in the southern part of the Atlantic Rainforest (Ribeiro *et al.* 2009), in southern Santa Catarina state, Brazil. Climate in the region is humid subtropical, with annual temperature of 19 °C and mean precipitation of 1600 mm (Alvares *et al.* 2014).

We conducted this research at three sites ( $28^{\circ}26'S$ ,  $49^{\circ}23'W$  –  $28^{\circ}25'S$ ,  $49^{\circ}25'W$ ) currently under ecological restoration. All sites were originally covered by riparian forest and had been degraded by surface coal mining with deposition of spoils between 1975 and 1997. After the end of mining activities, all sites were abandoned until 2009 and 2010, when the restoration procedures were initiated. The principal restoration goal was the reestablishment of the pre-mining riparian forest. Restoration of these sites followed the general procedures

applied in the region (Rocha-Nicoleite *et al.* 2013): after removal of spoils to stop ongoing contamination, the topography was restructured by a mixture of clay or sand of about 2 m in depth. A thin layer of organic matter (ca. 2 cm), in this case peat or broiler litter, was then distributed over that substrate to facilitate vegetation development. After that, seeds of exotic grasses (*Urochloa brizantha* (Hochst. ex A. Rich.) R.D. Webster, *Avena sativa* L. and *Lolium multiflorum* Lam.) were sown in order to rapidly establish vegetation cover (required by law), followed by planting of native trees. A total of 25 species were used in varying quantities, with a planting distance of 2 x 2 m between trees. Monitoring of tree development (height, crown and stem sizes – see below) occurred in permanent research plots maintained without any silvicultural practices. The young trees (planted seedlings) never received any nutritional support.

#### *Data collection and analysis*

We monitored species with at least 10 individuals at the first evaluation, which was about one year after the tree planting. Thus, only seedlings that survived this first year were considered. Altogether, we evaluated 685 individuals belonging to 25 species and 15 botanical families along three consecutive years with one measurement per year.

Relative growth rates were calculated for: height (from the base of the stem to the end of the crown), crown projection area (based on two measurements of the crown diameter), stem base diameter (measured at the base of the stem). For the calculation of growth rates, we considered only those 19 species that had 10 or more living individuals at the last sampling date. Relative growth rates (RGR) were calculated as  $(\log[X_t] - \log[X_0])/\text{time}$ , where  $X_0$  and  $X_t$  correspond to the parameter under evaluation (here: height, area of crown, or base diameter) at the first and last measurement, respectively (Poorter *et al.* 2008). Additionally, we also calculated the variation of the growth rate (standard deviation) among individuals for each species. Annual mortality rates (MR) were calculated as  $(\log[N_0] - \log[N_t])/\text{time}$ , where  $N_0$  and

$N_t$  are the number of individuals of each species at the first and last measurement, respectively (Poorter *et al.* 2008).

We evaluated seven functional traits: leaf area (LA,  $\text{cm}^2$ ); specific leaf area (SLA,  $\text{mm}^2 \cdot \text{mg}^{-1}$ ); leaf dry-matter content (LDMC,  $\text{mg} \cdot \text{g}^{-1}$ ); leaf nitrogen concentration (LNC, % per mass); leaf phosphorus concentration (LPC, % per mass); maximum adult height ( $H_{\max}$ , m); and wood density (WD,  $\text{g} \cdot \text{cm}^{-3}$ ). The plant functional traits were obtained from our own database of plants functional traits, which contains trait data of tree species in southern Brazil. Leaf traits measurements followed the protocol of Pérez-Harguindeguy *et al.* (2013). For each leaf trait, at least five individuals per species were sampled, within their natural distribution range. Maximum adult height and wood density data came from published studies or existing databases (Lorenzi 2002; Carvalho 2003, 2006; Chave *et al.* 2006; Carvalho 2008).

Relations between the parameters RGR, MR, and functional traits were first investigated through Spearman's rank correlation. To evaluate if functional traits predicted survival and growth rates, we used multiple linear regression models with a step-wise ascending procedure. In the multiple regression models, the final combination of traits for each growth rate (RGR of height, crown area, stem diameter) and mortality (MR) was retained according to the F-value criteria (Crawley 2014). When necessary, data were log-transformed to satisfy tests of normality. The average of the crown area in the last year was also considered in the analyses as a response variable in order to visualize this important structural pattern of the species (the potential to quickly form a canopy) and its relationships with the other parameters. In addition to consideration of each functional trait separately, we also used the full set of traits in the models of RGR and the MR, using a multivariate approach. For that, we first applied a principal component analyses (PCA) for the species described by the traits and considered the first three axes as potential descriptors in the models.

Finally, to explore the multivariate patterns of functional traits and the growth rates of the species, we applied a canonical correspondence analysis (CCA), performed on two matrices: the species described by their growth rates and the species described by their functional traits as the independent (explanatory) variable.

All analyses were performed using the package *vegan* (Vegan: Community ecology package) on the R platform (R Development Core Team, <http://www.Rproject.org>).

## Results

Relative growth rates (RGR) considering plant height, basal diameter or crown area were measured for 19 species, while the mortality rate (MR) was estimated for 25 species (Table 1). *Eugenia brasiliensis* and *Cupanea vernalis* were the species that presented the higher mortality rate. Considering the relative growth rate the species that highlight were *Schinus terebinthifolius*, *Handroanthus chrysotrichus*, *Alchornea triplinervia*, *Psidium cattleyanum* and *Citharexylum myrianthum*. Mean values of functional traits for each species evaluated are showed in Appendix 1. Some significantly correlations between the parameters of species growth and the functional traits could be shown (Table 2).

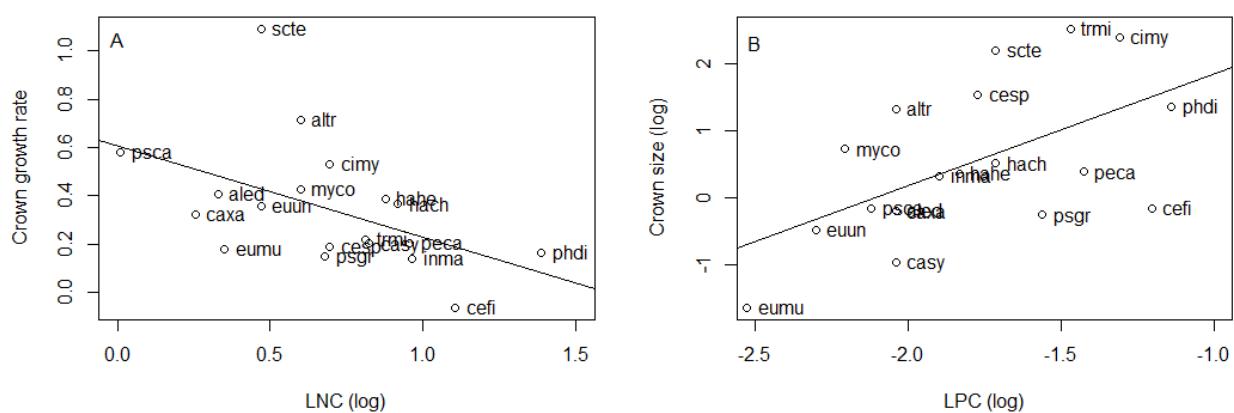
**Table 1.** List of species considered for the study with the number of measured individuals in the last year (N in 3<sup>rd</sup> year), their mortality rate (MR per year), and their relative growth rate considering height (RGR-H), basal diameter (RGR-D), and crown area (RGR-C), followed by their standard deviation (sd). Species without values for growth rates were not included in the analysis due to the low number of individuals (see methods).

Family	Species	Species Code	N	MR	RGR-H (sd)	RGR-H (sd)	RGR-D (sd)	RGR-C (sd)	RGR-C (sd)	
Anacardiaceae	<i>Schinus terebinthifolius</i> Raddi	scte	112	0.00	0.35	0.17	0.51	0.18	1.09	0.43
Apocynaceae	<i>Peschiera catharinensis</i> (A. DC.) Miers	taca	26	0.00	0.15	0.13	0.13	0.19	0.20	0.36
Bignoniaceae	<i>Handroanthus chrysotrichus</i> (Mart. ex A. DC.) Mattos	hach	10	0.03	0.30	0.14	0.30	0.14	0.37	0.42
	<i>Handroanthus heptaphyllus</i> (Vell.) Mattos	hahe	28	0.10	0.24	0.12	0.26	0.13	0.39	0.32
	<i>Handroanthus umbellatus</i> (Sond.) Mattos	haum	6	0.18	-	-	-	-	-	-
Cannabaceae	<i>Trema micrantha</i> (L.) Blume	temi	10	0.08	0.12	0.17	0.22	0.17	0.22	0.35
Euphorbiaceae	<i>Alchornea triplinervia</i> (Spreng.) Müll. Arg.	altr	10	0.06	0.31	0.15	0.40	0.13	0.72	0.30
Fabaceae	<i>Inga marginata</i> Willd.	inma	14	0.08	0.13	0.27	0.27	0.18	0.14	0.53
	<i>Mimosa scabrella</i> Benth.	misc	8	0.04	-	-	-	-	-	-
Malvaceae	<i>Ceiba speciosa</i> (A. St.-Hil.) Ravenna	cesp	23	0.00	0.15	0.18	0.22	0.15	0.19	0.27
	<i>Pseudobombax grandiflorum</i> (Cav.) A. Robyns	psgr	18	0.00	0.22	0.15	0.24	0.17	0.15	0.31
Meliaceae	<i>Cedrela fissilis</i> Vell.	cefi	56	0.03	0.16	0.15	0.19	0.11	-0.07	0.43
Myrtaceae	<i>Campomanesia xanthocarpa</i> Mart. ex O. Berg	caxa	28	0.11	0.23	0.17	0.22	0.18	0.32	0.40
	<i>Eugenia brasiliensis</i> Lam.	eubr	6	0.31	-	-	-	-	-	-
	<i>Eugenia involucrata</i> DC.	euin	9	0.18	-	-	-	-	-	-
	<i>Eugenia multicostata</i> D. Legrand	eumu	34	0.05	0.06	0.16	0.12	0.12	0.18	0.31
	<i>Eugenia uniflora</i> L.	euun	45	0.03	0.16	0.19	0.19	0.10	0.36	0.34
	<i>Psidium cattleyanum</i> Sabine	psca	17	0.05	0.31	0.14	0.40	0.15	0.58	0.33
Phytolaccaceae	<i>Phytolacca dioica</i> L.	phdi	12	0.00	0.37	0.09	0.22	0.15	0.16	0.43
Primulaceae	<i>Myrsine coriacea</i> (Sw.) R. Br. ex Roem. & Schult.	myco	15	0.08	0.30	0.20	0.26	0.21	0.43	0.35
Rubiaceae	<i>Posoqueria latifolia</i> (Rudge) Schult.	pola	8	0.06	-	-	-	-	-	-
Salicaceae	<i>Casearia sylvestris</i> Sw.	easy	13	0.16	0.10	0.28	0.26	0.16	0.20	0.57
	<i>Allophylus edulis</i> (A. St.-Hil., A. Juss. & Cambess.) Hieron. ex Niederl.	aled	18	0.12	0.18	0.22	0.24	0.15	0.41	0.36
	<i>Cupania vernalis</i> Cambess.	cuve	4	0.26	-	-	-	-	-	-
Verbenaceae	<i>Citharexylum myrianthum</i> Cham.	cimy	20	0.00	0.38	0.11	0.35	0.11	0.53	0.28

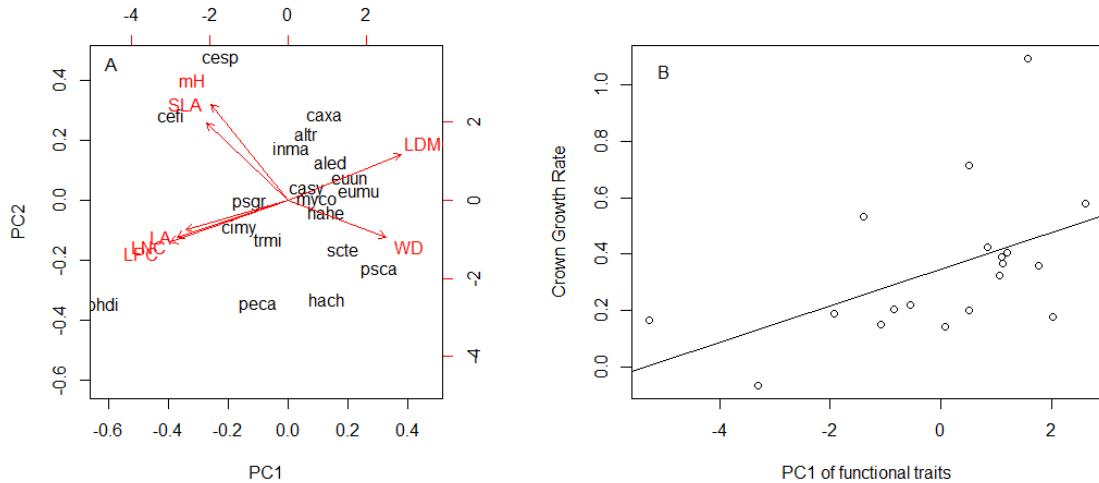
**Table 2.** Spearman correlation coefficients between functional traits and relative growth rate or its standard deviations (sd), considering plant height (HGR), basal diameter (DGR) and crown area (CGR). Additionally, we considered crown area ( $m^2$ ) in the last evaluation. Significant values are in bold ( $p < 0.01 = **$ ,  $p < 0.05 = *$ , and  $p < 0.10$ ).

Functional traits	HGR	HGRsd	DGR	DGRsd	CGR	CGRsd	Crown Area
Leaf area	0.16	<b>-0.53*</b>	-0.13	-0.34	<b>-0.44*</b>	-0.30	-0.02
Specific leaf area	-0.07	0.14	<b>-0.41*</b>	-0.21	-0.35	0.22	0.02
Leaf dry-matter content	-0.10	0.32	0.26	-0.18	0.29	-0.08	<b>-0.42*</b>
Leaf nitrogen concentration	-0.11	-0.29	-0.10	-0.06	<b>-0.55**</b>	<b>0.41*</b>	0.28
Leaf phosphorus concentration	0.24	<b>-0.55**</b>	-0.01	0.01	-0.30	0.18	<b>0.54*</b>
Maximum adult height	-0.03	0.05	-0.16	-0.17	<b>-0.48*</b>	-0.11	-0.07
Wood density	0.13	-0.03	0.21	-0.22	<b>0.42*</b>	0.10	<b>-0.41*</b>

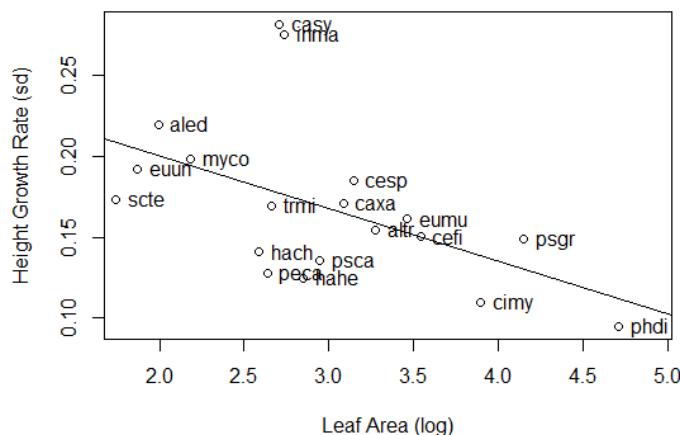
Regression models of RGR of crown area and of last crown size were significant when using the traits leaf nitrogen (LNC) and leaf phosphorus content (LPC), respectively, as predictor variables (Fig. 1). RGR of stem basal diameter and of height did not present any significant regressions to any trait. Considering the first axis of the multivariate space trait (PCA, Fig. 2a), we also could see a significant positive relation with crown growth rate (Fig. 2b). Considering the intraspecific variation in terms of growth rates, only the variability observed for height growth rate (HGRsd) showed a significant association to the species traits, in this case a negative relation to the leaf area (LA) (Fig. 3).



**Figure 1.** Significant linear models between (a) the crown growth rate and leaf nitrogen content (LNC, log transformed;  $R^2=0.19$ ;  $p=0.03$ ;  $F=5.20$ ;  $DF=17$ ) and between (b) the crown area ( $m^2$ , log transformed) and leaf phosphorus content (LPC, log transformed;  $R^2=0.27$ ;  $p=0.01$ ;  $F=7.79$ ;  $DF=17$ ). Species labels can be seen in Table 1.

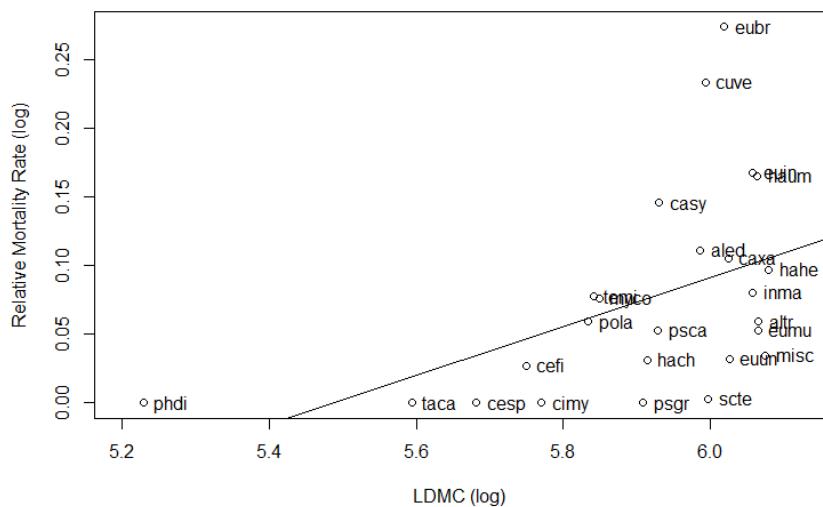


**Figure 2.** Ordination diagram (PCA) of 19 species described by their functional traits (A) and the significant linear model (B) between the crown growth rate and the first ordination axis of the PCA ( $R^2=0.20$ ;  $p=0.03$ ;  $F=5.59$ ;  $DF=17$ ). Species labels can be seen in Table 1.



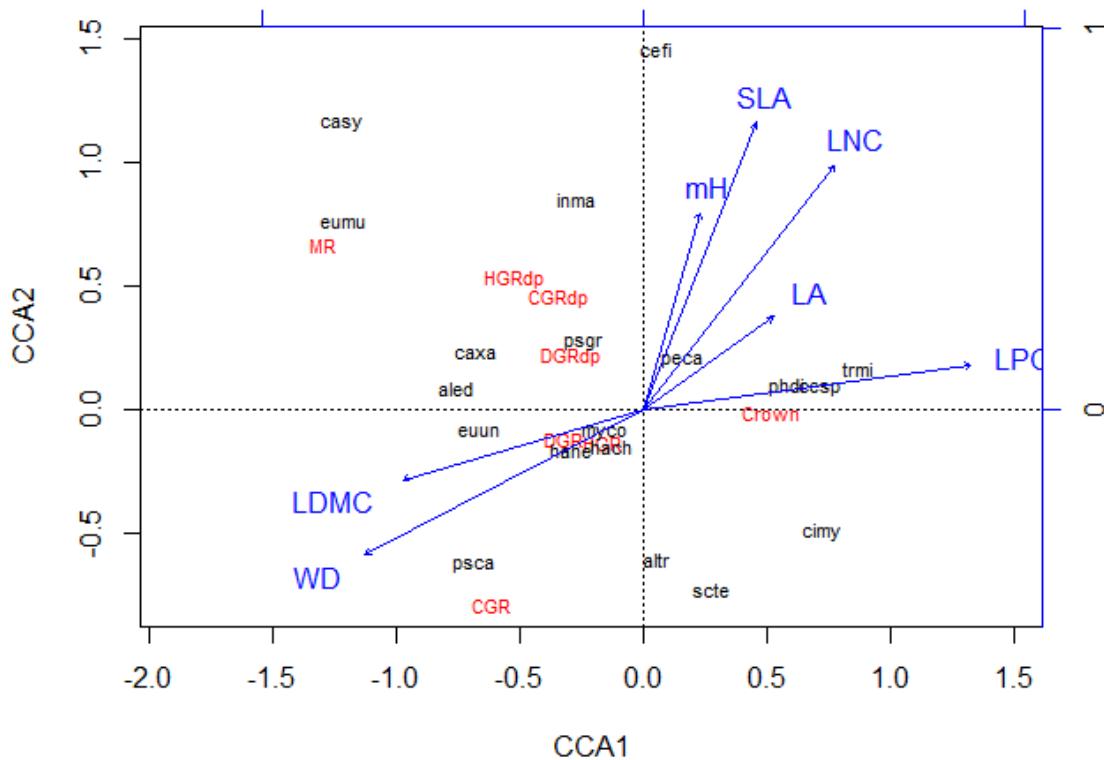
**Figure 3.** Linear regression between the variation of the height growth rate (HGR) and the leaf area (LA - log transformed;  $R^2=0.22$ ;  $p=0.02$ ;  $F=6.00$ ;  $DF=17$ ). Species labels can be seen in Table 1.

For the mortality rate, we only found a significant model when using leaf dry-matter content (LDMC) as predictor variable, with a positive relation (Fig. 4). When considering the first ordination axis of the PCA done for the 25 species used for estimation of MR (Appendix 2A), a very similar regression pattern associated to the LDMC was found (Appendix 2B).



**Figure 4.** Linear regression between the relative mortality rate and the leaf dry-matter content (LDMC; log transformed;  $R^2=0.18$ ;  $p=0.01$ ;  $F=6.42$ ; DF=23). Species labels can be seen in Table 1.

In the CCA, the set of functional traits explained 56% (axis 1= 40%; axis 2=11%) of the total growth rates variation of the species set (19 species). The first CCA axis was strongly and positively correlated with LDMC and WD and negatively correlated with LPC, LNC and SLA (Fig. 5). The species *Psidium cattleyanum*, *Eugenia uniflora* and *Allophylus edulis* showed high correlations to the first axis, and so did crown growth rate. Species in the left portion of the diagram had a lower growth rate, but higher mean values of crown area, being further associated with LPC. Species with higher variation in their growth rates were positively correlated to the axis 2, and associated with higher values of LNC and SLA.



**Figure 5.** CCA ordination (canonical correspondence analysis) results ordered in multivariate space along the first two canonical axes ( $p=0.05$ ). The sample units are the tree species coded as showed in table 1. On the red color follows the response variables (relative mortality rate = MR; relative growth rates: HGR = height growth rate; HGRsd = height growth rate standard deviation; DGR = diameter growth rate; DGRsd= diameter growth rate standard deviation; CGR = crown growth rate; CGRsd = crown growth rate standard deviation; Crown = crown area). The blue color correspond to the functional traits (mH = Maximum adult height; LA = Leaf area; LDMC = Leaf dry matter content; LNC = Leaf nitrogen content; LPC = Leaf phosphorus content; SLA = Specific leaf area; WD = woody density).

## Discussion

Based on their plant traits, the species planted in the restoration project could be placed along a gradient from rapid resource acquisition (high SLA, LA, LNC, LPC) to conservation of resources (high LDMC and WD) (Díaz *et al.* 2004; Wright *et al.* 2007). These differences, however, were only weakly related with differences in the vital rates of species,

and some of them were even contradictory to our expectations. In general, only the relative growth rate (RGR) of the crown area parameter responded to variation in functional traits. Lower values of LA, LNC, and LPC and higher values of WD and LDMC characterized species with higher RGR regarding crown area. Under the conditions of our study areas, individuals of species with more conservative strategies of resource used had higher relative growth than those classified as fast-growth species according to the ‘leaf economics spectrum’ approach (Wright et al. 2004). On the other hand, the mortality rate was higher for species with higher WD and LDMC, as we had expected. Altogether, these results, indicate that conservative species have a smaller crown size and grow relatively more over a year, even with a higher probability of dying than the species with strategies of fast resource acquisition.

Contrarily to our expectation, species with higher LNC – an indicative of species with higher photosynthetic capacity – tended to have a higher crown growth rate during the studied period. Nevertheless, the average size of the crown area was positively related to LPC, which indicates that species with higher LPC tend to have large crowns. Despite the size of their crowns, these species did not grow faster under the conditions of the studied restoration sites. High SLA, LNC and LPC are commonly associated with high photosynthetic capacity and faster turnover of plant leaves (low leaf-life span), which in turn should permit flexible response to spatial patchiness of light and soil resources, giving short-term advantages (Westoby et al. 2002). However, as observed in this study, opposite species strategies seem to have an overall advantage for planted seedlings, maintaining a relative crown growth rate under the harsh conditions of the restoration sites (e.g. high grass competition, light incidence, and temperature fluctuation).

Using the traits together in a multivariate approach, similar results as for single traits emerged (Fig. 2 and Fig. 5). Species with higher crown growth rate (e.g. *A. edulis*, *Eugenia multicostata*, *E. involucrata*, *P. cattleyanum*) were further associated to higher LDMC

and WD (first axis of the PCA, Fig 2a), contrasting to species with higher SLA, LPC, and LNC (e.g. *Phytolacca dioica*, *Citharexylum myrianthum*, *Cedrela fissilis*). These results emphasize that species that invest more to maintain the size of the crown do not grow continuously, leading to a lower increase rate along the first years after planting. In forest restoration, canopy closure is a key process, because it may change the microclimate by adding shade, which should decrease the cover of competing grasses and improve the natural regeneration (Martínez-Garza *et al.* 2013). Thus, both species with high crown growth rate and species with large crowns are essential to improve this process, but there does not seem to be a positive relation between these two parameters among the young individuals of the studied tree species.

The SLA is one of the traits that are commonly used to predict the performance of plants (Poorter *et al.* 2008; Asanok *et al.* 2013), but in our study it did not present any strong significant correlation with the growth rate parameters. Some weak relationships could be seen in the overall correlation matrix and under the multivariate approach (Fig. 5), where species with higher SLA and LNC were somewhat related to a higher intraspecific variation in the growth rates. This may be attributed to local variability in terms of soil nutrients and neighbor conditions (low differences in terms of light, humidity and competition), thus restricting the overall performance of fast-growth plants that usually demand high concentrations of resources. On the other hand, more conservative species (with high LDMC and WD) generally require less nutrients and can survive in such environment conditions. We additionally saw that the species standard deviation in height growth rate (i.e. young trees of one species are growing more heterogeneous in terms of height) was negatively related to LA, which may indicate that species with low LA are more sensitive to the surrounding conditions. As noticed by Campoe *et al.* (2014b), local conditions may have a significant role on plant growth performances, so that we did not find clear patterns of relative growth only considering the functional traits.

It has been shown that species with high LDMC are more likely to survive and grow well under harsh conditions (Martínez-Garza *et al.* 2013). In our study, species with high LDMC also grew more in crown area, however their mortality rate was also high, at least when considering those young trees of species that already had survived the first 12 months after planting. High LDMC can be related to non-pioneer species, since they tend to have longer leaf lifespan, lower SLA, and the nutrient return is slower (Martinéz-Garza *et al.* 2013). In our case, we suppose that young trees of rainforest trees characterized with high LDMC content were more susceptible to dying due to their inability to overcome the grass ground layer, present in most of studied restoration sites. Some species with these features, high LDMC and high mortality rate (*e.g. Eugenia brasiliensis*, *E. involucrata*, *Cupania vernalis*), were not considered for the growth rates analysis, since they had too few individuals in the last survey. On the other hand, plants with thicker leaves, which also express high dry-matter content, can also be related to nutrient-poor soils and high light conditions (Westoby *et al.* 2002). So, in this study, surviving individuals of species with high LDMC were able to grow more, at least in terms of their crown.

We still know very little about functional traits and vital rates (growth and mortality) of tree species on ecological restoration conditions, where they often are under very specific environmental and resource conditions, and even on natural forest conditions to better conclude about the potential of predictability of traits on plant growth rates (Paine *et al.* 2015). On the other hand, this knowledge can bring important insights for restoration ecology theory as well as for ecological restoration practices, improving the success of the projects (Laughlin 2014a). Our study showed that some traits could be related to the growth and mortality rates of planted seedlings, but the way what such relations occurred were still unclear and contrary to some finds of the literature. For the species we studied, no clear relation between plant traits and growth and mortality rates existed. Facing this, we believe that more and broader studies

encompassing growth rates and functional traits of species are needed in order to apply the results of theoretical ecology in practical ecological restoration.

## Acknowledgments

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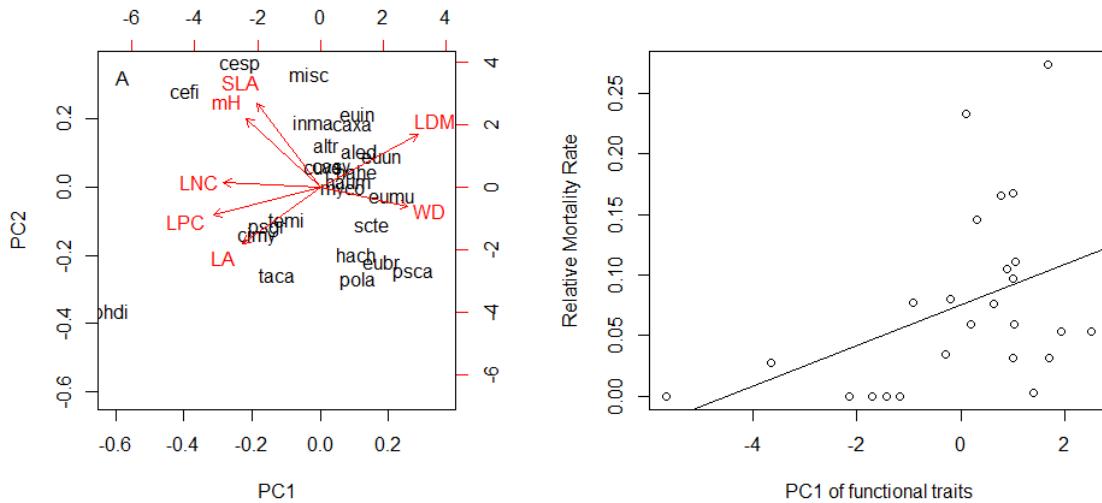
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**Appendix 1.** List of species considered for the study with their species code and the number of measured individuals to access the functional traits (N): leaf area (LA); specific leaf area (SLA); leaf dry-matter content (LDMC); leaf nitrogen concentration (LNC); leaf phosphorous concentration (LPC); maximum height (mH); wood density (WD).

Family	Species	Species Code	N	LA	SLA	LDMC	LNC	LPC	mH	WD
Euphorbiaceae	<i>Alchornea triplinervia</i> (Spreng.) Müll. Arg.	altr	7	26.50	8.04	430.56	1.82	0.13	28.00	0.47
Sapindaceae	<i>Allophylus edulis</i> (A. St.-Hil., A. Juss. & Cambess.) Hieron. ex Niederl.	aled	5	7.35	14.79	397.67	1.39	0.13	19.00	0.65
Myrtaceae	<i>Campomanesia xanthocarpa</i> Mart. ex O. Berg	caxa	9	21.87	12.82	413.25	1.29	0.13	28.00	0.86
Salicaceae	<i>Casearia sylvestris</i> Sw.	casy	8	14.97	12.07	376.05	2.28	0.13	22.00	0.71
Meliaceae	<i>Cedrela fissilis</i> Vell.	cefi	7	34.51	23.58	313.92	3.02	0.30	30.00	0.49
Malvaceae	<i>Ceiba speciosa</i> (A. St.-Hil.) Ravenna	cesp	1	23.18	22.77	293.37	2.00	0.16	30.00	0.39
Verbenaceae	<i>Citharexylum myrianthum</i> Cham.	cimy	8	49.14	12.25	320.48	2.00	0.27	24.00	0.60
Sapindaceae	<i>Cupania vernalis</i> Cambess.	cuve	8	21.14	10.79	400.74	1.72	0.19	25.00	0.66
Myrtaceae	<i>Eugenia brasiliensis</i> Lam.	eubr	5	59.07	8.71	411.05	1.10	0.07	16.00	0.71
Myrtaceae	<i>Eugenia involucrata</i> DC.	euin	3	19.60	21.06	427.09	1.80	0.10	17.00	0.76
Myrtaceae	<i>Eugenia multicostata</i> D. Legrand	eumu	4	31.90	9.33	430.23	1.42	0.08	21.00	0.92
Myrtaceae	<i>Eugenia uniflora</i> L.	euun	8	6.50	14.04	413.78	1.60	0.10	18.00	0.83
Bignoniaceae	<i>Handroanthus chrysotrichus</i> (Mart. ex A. DC.) Mattos	hach	6	13.33	9.38	370.24	2.50	0.18	16.00	1.05
Bignoniaceae	<i>Handroanthus heptaphyllus</i> (Vell.) Mattos	hahe	6	17.25	12.83	436.77	2.40	0.16	20.00	0.98
Bignoniaceae	<i>Handroanthus umbellatus</i> (Sond.) Mattos	haum	2	7.29	8.80	430.00	2.50	0.15	20.00	0.67
Fabaceae	<i>Inga marginata</i> Willd.	inma	7	15.43	11.93	427.48	2.62	0.15	26.00	0.58
Fabaceae	<i>Mimosa scabrella</i> Benth.	misc	3	0.38	15.92	434.49	3.00	0.14	25.00	0.56
Primulaceae	<i>Myrsine coriacea</i> (Sw.) R. Br. ex Roem. & Schult.	myco	6	8.90	11.60	346.85	1.82	0.11	19.00	0.59
Apocynaceae	<i>Peschiera catharinensis</i> (A. DC.) Miers	taca	7	13.94	10.61	268.82	2.60	0.24	16.00	0.55
Phytolaccaceae	<i>Phytolacca dioica</i> L.	phdi	5	111.04	15.28	186.68	4.00	0.32	26.00	0.44
Rubiaceae	<i>Posoqueria latifolia</i> (Rudge) Schult.	pola	7	24.53	4.64	341.63	1.48	0.12	17.00	0.57
Malvaceae	<i>Pseudobombax grandiflorum</i> (Cav.) A. Robyns	psgr	5	63.49	9.34	368.23	1.97	0.21	25.00	0.39
Myrtaceae	<i>Psidium cattleyanum</i> Sabine	psca	8	19.07	5.21	375.73	1.01	0.12	18.00	1.12
Anacardiaceae	<i>Schinus terebinthifolius</i> Raddi	scte	5	5.71	9.54	402.11	1.60	0.18	16.00	0.80
Cannabaceae	<i>Trema micrantha</i> (L.) Blume	temi	9	14.35	10.70	344.17	2.25	0.23	18.50	0.35

**Appendix 2.** Ordination diagram (PCA) of 25 species described by their functional traits (A) and the significant linear model (B) between the relative mortality rate and the first ordination axis of the PCA ( $r^2=0.13$ ;  $p=0.03$ ;  $F=4.89$ ; DF=23).



## CAPÍTULO 2

### SEED RAIN AND PLANT ESTABLISHMENT PATTERNS IN FOREST RESTORATION ON COAL MINING DEGRADED AREAS



# **Seed rain and plant establishment patterns in forest restoration on coal mining degraded areas**

Edilane Rocha-Nicoleite<sup>1,2</sup>; Gerhard Ernst Overbeck<sup>3</sup>; Sandra Cristina Müller<sup>1</sup>

1 – Graduate Program in Ecology, Department of Ecology, Universidade Federal do Rio Grande do Sul, Av. Bento Gonçalves 9500, 91540-000, Porto Alegre, RS, Brazil;

2 – Clean Coal Research Centre, Associação Beneficente da Indústria Carbonífera de Santa Catarina, Rua Pascoal Meller 73, 88805-380, Criciúma, SC, Brazil (e-mail: edilane.rocha@satc.edu.br);

3 – Department of Botany, Universidade Federal do Rio Grande do Sul, Av. Bento Gonçalves, 9500, 91540-000, Porto Alegre, Brazil.

## **Abstract**

Seed arrival and plant establishment are two fundamental processes for the ongoing forest restoration of degraded areas. The use of perches to attract seed dispersers has been used to improve seed rain, although there is no consensus about the recruitment effectiveness near the perches. In a high-degraded region by coal mining, in southern Brazil, we aimed at evaluating species richness and abundance of the seed rain and seedling recruitment through of sampling with or without perches, considering also seed traits, co-variation in composition patterns and relations with the herbaceous layer. Seed rain was evaluated every two weeks for one year and we sampled the recruitment of woody species, and the percentage cover of the herbaceous layer (native grasses, exotic grasses and forbs) in 26 plots. We compared the differences in seed rain traits between seed traps with and without perches by paired t-test, and the relation between seed rain and seedling recruitment by co-inertia analysis. We also performed linear regressions

between the proportion of plant recruitment and the abundance of seed rain of grasses and non-grass species, and between the percentage of cover of grasses and forbs. The presence of perches enhanced the seed rain, especially of non-grass species. The proportion of seeds of zoothorax and non-pioneer species was higher as well, which highlight the importance of perches to overcome the barrier of dispersal. However, the effectiveness of recruitment of woody species was not directly related to the seed rain. This recruitment was positively related to the cover of forbs. So, even increasing the abundance of desired seed, perches alone did not enhance the establishment of woody plants, which in turn seems to be related to local ecological conditions, as the proportional cover of forbs and exotic grasses.

**Key-words:** seed traps; recruitment; woody species; ecological restoration.

## **Introduction**

Arrival and establishment of native species are important processes in the recovery of degraded plant communities and ensure the self-sustainability of restored areas (Chazdon 2008). Multiple biotic and abiotic filters influence the specific composition of the community that emerges from restoration, in terms of species and functional traits (Zhan-Huan *et al.* 2013; Barnes & Chapman 2014; von Gillhaussen *et al.* 2014). Areas under restoration might follow different trajectories of succession and even achieve different stable states according to local environmental conditions or priority effects (von Gillhaussen *et al.* 2014). For tropical forest restoration, barriers that limit secondary succession in degraded sites (Holl 1999; Florentine & Westbrooke 2004) are mostly related to the low availability of seeds, usually in consequence of the lack of seed bank and of dispersers (Chazdon 2003; Baur 2014) and to the distance to propagule sources (Pereira *et al.* 2013).

The high abundance of exotic species in degraded areas, especially of invasive grasses that compete with native species, can constitute a particularly strong barrier to the establishment of native species (Martinez-Ramos & Soto-Castro 1993; Ortega-Piecka *et al.* 2011), sometimes exacerbated by the presence of allelopathic substances (Zimmerman *et al.* 2000). Timing of seedling emergence may also play a critical role for the development of the community under restoration. Often, areas under restoration suffer high propagule pressure from exotic species that dominate in the extant communities (Wainwright *et al.* 2012). Thus, knowledge about the processes underlying the arrival and establishment of new species, whether desired (native species) or not (invasive species), is essential to predict and manage changes in the restoration process and to define realistic restoration goals (D'Antonio 1993; Wainwright *et al.* 2012; Byun *et al.* 2013).

Perches have been widely used to attract seed dispersers (Heelemann et al., 2012; Cavallero et al 2013; Graham & Page 2012) and to aid in the establishment of nuclei where the development of the community occurs more rapidly (Yarranton & Morrison 1974). The use of perches works well to attract dispersers in fragmented landscapes, as it provide a place to frugivorous birds rest on and defecate bringing propagules to into deforested zones (Graham & Page 2012). Nonetheless, no consensus about the effectiveness regarding number of recruits below or around the perches exists (Graham & Page 2012; Heelemann *et al.* 2012; Cavallero *et al.* 2013), making further studies necessary in order to establish general rules.

In this study, we evaluate species composition, richness and density of the seed rain and of seedling recruitment in areas degraded by coal mining in the first phase of a restoration process, which has the overall goal of restoring a similar original riparian forest. We evaluated the recruitment patterns and the seeds of traps associated or not to perches and tested the following hypothesis: (1) Seed traps below perches should present higher richness and abundance of seeds, especially seeds of non-grass and zoolochoric species with great seed sizes. (2) There should be a tight relationship between seed rain and natural seedling recruitment, concerning species composition, abundance and traits of seeds sampled in traps. (3) Higher seed rain of woody species should be related to higher recruitment of woody species, but (4) only in areas where the cover of exotic grasses does not delay or impede seedling recruitment.

## Materials and methods

### *Study sites*

Research was conducted at three sites (from lat 28°S 26'28'', long 49°W 23'42'' to lat 28°S 25'51'', long 49°W 25'27'') in southern Santa Catarina state that are currently under

ecological restoration. Originally, the region was completely covered by Atlantic rain forest *sensu stricto* (Ribeiro *et al.* 2009), a global biodiversity hotspot (Myers *et al.* 2000). All sites were coal mined (surface mining with deposition of spoils over the areas) between 1950 and 1989, and restoration interventions were carried out between 2009 and 2010. The restoration project was established in riparian zones and the principal goal was the reestablishment of a forest structure similar to the pre-mining riparian forest communities.

Restoration techniques followed the procedures commonly applied in the region: the first step to restore these areas was the removal of spoils to stop ongoing contamination. After this, a new substrate was formed, composed of clay or sand (depth ca. 2 m), in order to provide support for vegetation and to construct the topography. On the top of this new soil, a thin layer of organic matter (usually a layer of approximately 2 cm of peat and broiler litter) was added. Then, in order to rapidly establish a vegetation cover on the soil (which is required by law), all sites were sown with exotic grasses, followed by planting or seeding of native trees. In the studied sites the following exotic grasses were introduced: *Urochloa brizantha* (Hochst. ex A. Rich.) R.D. Webster (18 kg/ha) in one of the areas, *Avena sativa* L. and *Lolium multiflorum* Lam. (30 kg/ha) in the other two. Following this, 30 native tree species were planted with a density of 2500 individuals/ha.

Few natural forest remnants exist in the vicinity of the restoration site, i.e. few sources of propagules for the sites under restoration exist. Proportion of forest inside a buffer zone of 500 m varied from 6 to 30% among the three sites, and inside the buffer of 1000 m it varied from 11 to 45%. These distances did not have apparent influences on the seed rain patterns.

### *Sampling design and data collection*

To assess the recruitment pattern of woody species, we established 26 plots of 10 × 30 m (six in one area and 10 in the other two). Within each of these plots, we established three subplots of 5 × 5 m to sample the woody regeneration component and to estimate the percentage cover of the herbaceous layer (native grasses, exotic grasses and forbs). Close to each plot (ca. 6 m) we placed a pair of seed traps, one with and another without perch (height: 8 m). The seed traps consisted of a wooden frame of 1 m<sup>2</sup> with a cloth of mesh-size 1 mm<sup>2</sup>, fixed at 1 m above the ground.

Seed trap content was collected every two weeks for one year (May/2011 to April/2012) and samples were dried and sorted out into morpho-species counted and identified. Seeds were identified to the species level when possible by comparing them to seeds collected from individuals that were fructifying during the experiment, or by consulting local botanists and specimens deposited at the Laboratory of Plant-Animal Interaction at the Universidade do Extremo Sul Catarinense (UNESC).

Natural regeneration of woody species, defined as all individuals with height ≥ 50 cm and dbh ≤ 5 cm, was evaluated in January 2014 in the subplots. All plants were identified to the species level in the field or by help of the literature/or and consultation to herbaria and specialists. Classification into families follows APG III (2009) and the species nomenclature follows MOBOT (2010).

Additionally, we collected traits information for the species present in the seed rain and in the regeneration layer. For all fully identified species (or genus with consistent trait stages) we considered the following traits: successional category (pioneer or non-pioneer), dispersal mode (zoocoric, anemocoric and authocoric) and original distribution range (native or exotic). For seeds sampled in the traps, including identified species and morpho-species, we measured seed size (width and length) and noted the colour (black, white and brown).

### *Data analysis*

We aimed at (1) testing the effects of the perches in the seed rain concerning differences according to the traits of the seeds, (2) evaluating linear relationship between the abundance of seed rain per category of seeds (grass or woody species) and the proportion of woody plants recruitment, and (3) analyzing the relationships between the composition of seed rain and of plant recruitment throughout all sampling areas, i.e. at the community level.

The seed rain data was organized in two matrices. Matrix **B<sub>S</sub>** contained the seed rain species described by traits, and matrix **W<sub>S</sub>** the plots (with or without perches, 26 plots per group) described by the abundance of seed rain species. The seedling recruitment data was also organized in a similar way: **B<sub>R</sub>** was the matrix of recruiting species by traits and **W<sub>R</sub>** a matrix of plots described by the abundance of recruiting species. For this species set, data from subplots were pooled to the plot level. We also calculated community-weighted mean of traits (CWM) for the seed rain data, through matrix multiplication ( $CWM = W_S B_S'$ ) (Garnier et al., 2004).

Total abundance and richness of seeds in seed traps were compared between sampling units with and without perches by paired t-test based on log-transformed data. This analysis was also done considering only non-grass species. To analyze the effect of perches on expression of functional traits, we further performed a paired t-test comparing sampling units with or without perches concerning the CWM of traits per unit.

To evaluate the relation between the patterns of sampling units described by either the seed rain or seedling recruitment, we conducted a co-inertia analysis using the log-transformed matrices **W<sub>S</sub>** and **W<sub>R</sub>**. For this, the sampling units of **W<sub>S</sub>** were separated according to the information of units with or without perches and two analyses were run. The co-inertia axes were calculated maximizing the covariance of the factorial scores generated in the separate ordinations of the two input tables, carrying out at PCA for each matrix (Dray et al 2003). The

co-inertia method illustrates de co-structure between both matrices for every sample unit showing pairs that belonging to the same locality that are linked by an arrow. A Monte-Carlo permutation test was used to check the significance of the correlation between the two data sets. The correlation between the two matrices is given by RV-coefficient, which varies between 0 and 1: the closer the coefficient to 1, the stronger the correlation between the tables (Thioulouse *et al.* 1997).

Finally, we performed linear regressions to see the relations between the proportion of plant recruitment and the abundance of seed rain of grasses or woody species, and also considering the percentage of cover of grasses and forbs.

All statistical analyses were carried out on the R platform (R Development Core Team 2009), using package FD (Laliberté & Legendre 2010) for calculation of the CWM matrix and package ADE4 (Thioulouse *et al.* 1997) for CoIA analysis.

## Results

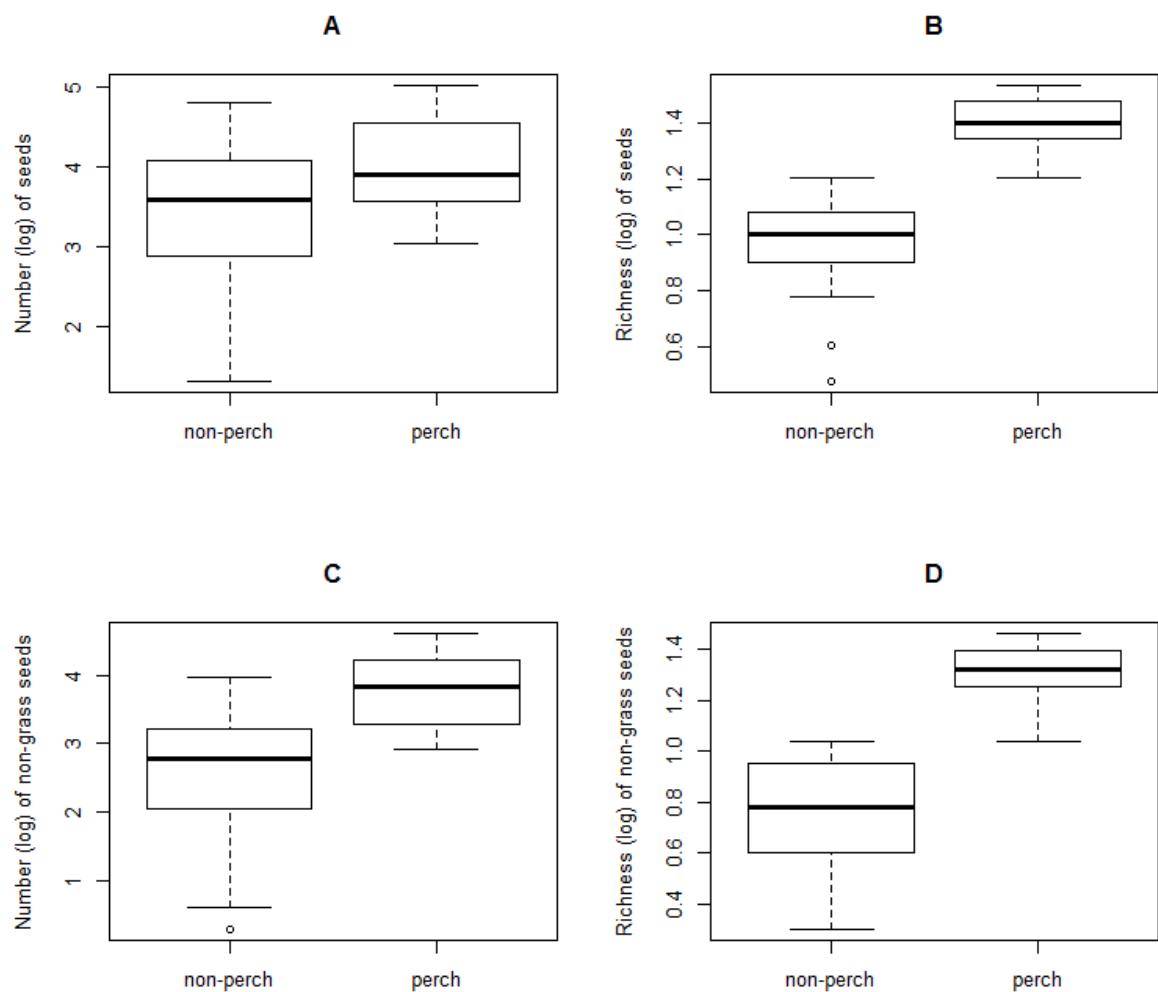
### *Seed rain patterns in traps with and without perches*

We registered 32.828 seed m<sup>2</sup> year<sup>-1</sup>. Of these, 61.9% were from grasses and 38.1% from other families (non-grass), including forbs, shrubs and tree species (Table 1). Fifty-one morpho-species were sampled, of which 10 species could not be identified at all (the other 41 species were identified at least until family level, see appendix 1). Annual distribution of the seeds abundance and richness did not present much variation, yet we could see a decrease between August and October and a concentration of arriving seeds during the summer (Appendix 2).

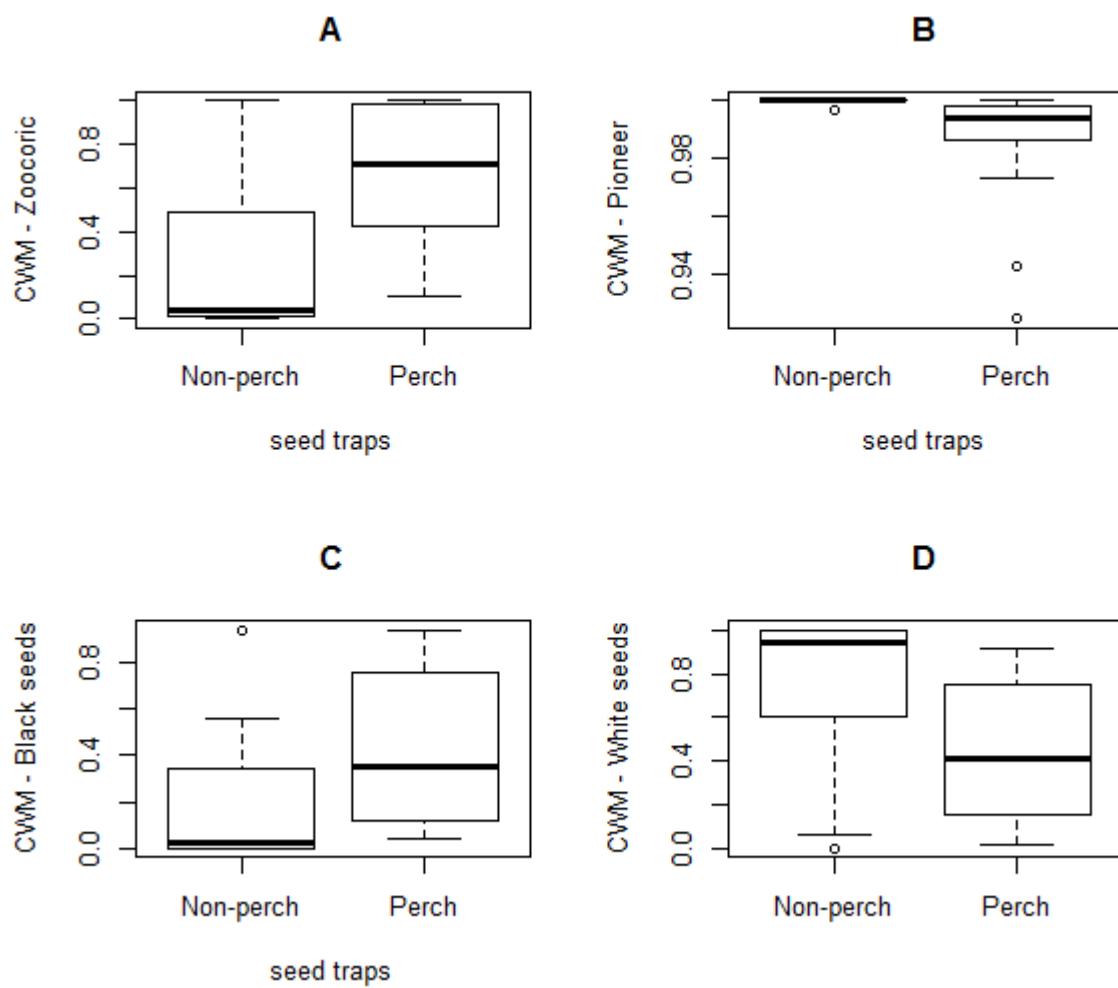
**Table 1.** Overview of the seed rain, in seed traps with and without perches, considering percentage of grasses and non-grasses in terms of seedling number, number of seeds per m<sup>2</sup>.year<sup>-1</sup> and species richness of the seed rain.

Seed traps	Seed abundance (m <sup>2</sup> .year <sup>-1</sup> )		Richness of seed rain	
	Grass	Non-grass	Grass	Non-grass
All	20312 (61.9%)	12526 (38.1%)	12	39
With perches	8845 (44.6%)	10980 (55.4%)	11	38
Without perches	11467 (88.1%)	1546 (11.9%)	12	20

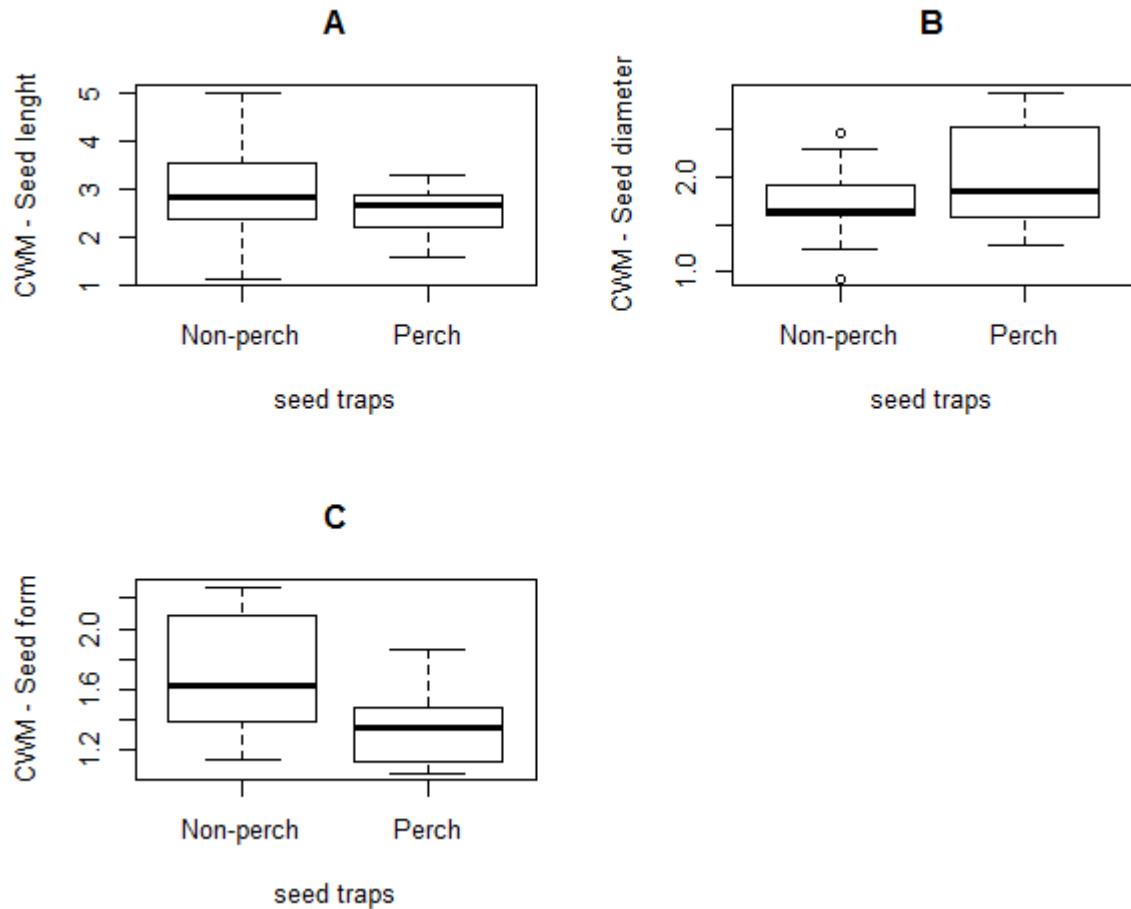
The presence of perches increased significantly both the number of species and the total abundance of seeds, either considering all seeds or only non-grass seeds (Fig. 1). Seed traps with and without perches also differed regarding CWM traits of seeds (Fig. 2). Under perches, most of the seeds collected were dispersed by animals (zoochoric) and the predominant color of the seeds was black, while in seed traps without perches, the proportion of seeds from pioneer species and with white color were higher (Fig. 2). The size of seeds also presented differences, although the difference for length was not significant ( $p=0.06$ ). However the relation between the length and diameter (length:diameter), which is an indicative of seed form (values around 1 indicate round seeds), was significantly higher in seed traps without perches (Fig. 3).



**Figure 1.** Number and species richness of seeds (log-transformed) collected in seed traps with and without perches, considering all seed species (A and B) and only non-grass species (C and D). The box indicating the variability inside the lower and the upper quartiles and the vertical line (whiskers) indicate variability outside the upper and lower quartiles; the individual points indicate the outliers. Mean values of groups differed significantly ( $p<0.005$ ) for all four variables.



**Figure 2.** Value of community weighted mean of traits (CWM) of sampled seeds, comparing seed traps with and without perches (% of zoocoric species, A), successional group (% of pioneer species, B), and seed color (% of black seeds, C; % of white seeds, D). The box indicating the variability inside the lower and the upper quartiles and the vertical line (whiskers) indicate variability outside the upper and lower quartiles; the individual points indicate the outliers. Mean values of groups differed significantly ( $p < 0.05$ ) for all variables.

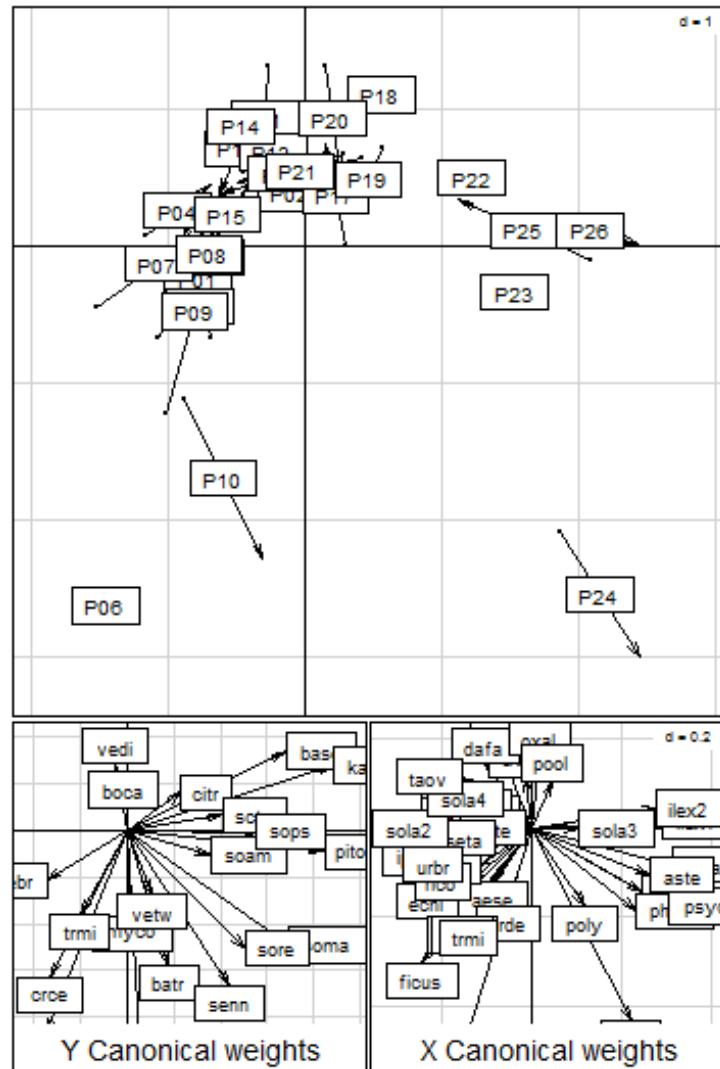


**Figure 3.** Value of community weighted mean traits (CWM) of sampled seeds, comparing seed traps with and without perches A; seed length (mm), B: seed diameter (mm) and C: seed form (relation between length and diameter). The box indicating the variability inside the lower and the upper quartiles and the vertical line (whiskers) indicate variability outside the upper and lower quartiles; the individual points indicate the outliers. Mean values of plots A and B presented a weak relation ( $p<0.06$ ) and the mean values of plot C differed significantly ( $p<0.001$ ).

#### Relationships between seed rain and plant recruitment

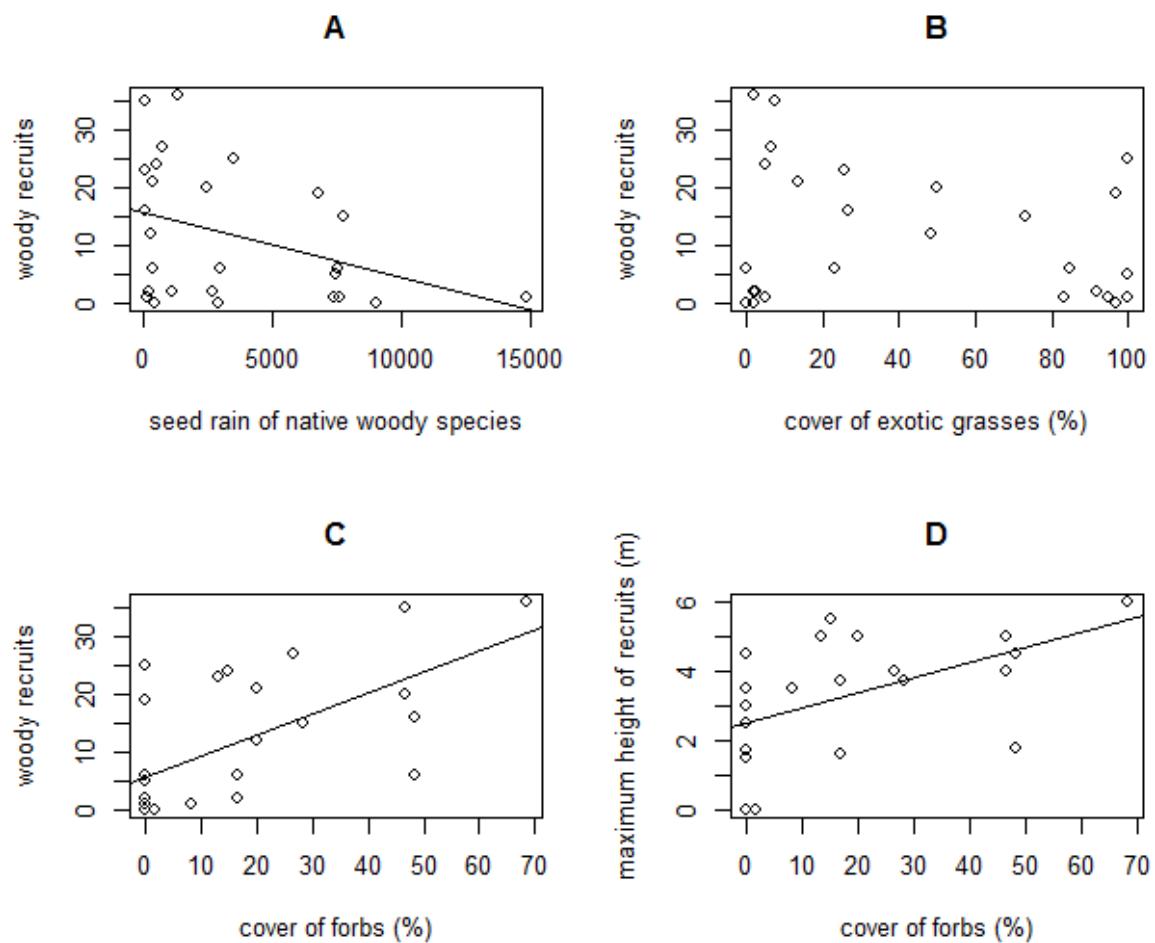
Co-inertia analyses of the seed rain and recruit species showed significant co-variation between both datasets as when considerate only seed traps with perches ( $RV= 0.45$ ;  $p= 0.02$ ). The first axis explained 18.9% and the second axis 5.1% of the co-variation (Fig. 4). The first axis of the co-inertia represents a gradient from plots with higher establishment of woody species (e.g. *Kaunia rufescens*, *Baccharis semiserrata*, *Piptocarpha tomentosa* and *Solanum pseudoquina*), at the right, to plots where density of wood species was markedly

lower, at the left. The pattern of seed rain followed that of established woody plants, that is, on the right side, species that were absent in the plots of the left side could be found. The seed traps without perches, did not presented significative co-variation (Appendix 4).



**Figure 4.** Relationship between patterns of seed rain and plant recruitment at the restoration sites for seed traps with perches. The co-inertia analysis shows the ordination of plots (distinguished by plot number) described by seed rain and woody plant recruitment based on abundances. Shorter arrows indicate better adjustment of patterns in both sets of data (seed rain species and recruits of woody species). The PCAs of both data sets are shown separately at the bottom (recruitment patterns at the left; seed rain patterns at the right; the species codes are in the species list: Appendix 1 for seed rain, Appendix 3 for recruits).

The seed rain of native woody species did not improve the abundance in recruitment of plants. In contrast to our expectations, a weak negative linear relation was found (Fig. 5a,  $R^2=0.10$ ;  $F=3.95$ ;  $p=0.05$ ). No relationship existed between recruitment of woody species with the cover of exotic grasses in the plots (Fig. 5b). On the other hand, we found that the cover of forbs linearly increased the number of native woody plants recruitment and these recruits tended to be taller (Fig. 5c and 5d,  $R^2=0.37$ ;  $F=16.31$ ;  $p<0.001$  and  $R^2=0.24$ ;  $F=8.98$ ;  $p=0.006$ , respectively).



**Figure 5.** Linear regressions of woody species recruitment (number and height) and the patterns of woody species seed rain (A), exotic grasses cover (B), and forbs cover (C and D) in the plots.

## **Discussion**

Both dispersal and recruitment of seeds are fundamental for plant community establishment on areas under ecological restoration (Howe *et al.* 2010). In our study, the presence of perches resulted in higher abundance and richness of seeds of zoothchorous and non-pioneer species compared to the control seed traps without perches, where the presence of non-grasses was lower. However, places with higher seed rain of non-grass species were not characterized by higher recruitment of woody species. We had expected a negative influence of high cover by exotic grasses on woody plant recruitment (Batten *et al.* 2005b; von Gillhaussen *et al.* 2014). However, contrarily to our expectation, the recruitment of woody species was independent on the cover of exotic grasses. On the other hand, recruitment had a slight increase in plots with high cover of forbs, which were established naturally through propagules present in the thin layer of organic matter placed over the new soil and/or from the seed rain. In the following, we discuss potential mechanisms underlying the patterns of initial restoration processes on high-degraded areas of tropical forest.

The increase in seed rain due to the presence of perches has been shown in many studies (Graham & Page 2012; Heelemann *et al.* 2012; Cavallero *et al.* 2013), and this is true especially for seeds dispersed by animals and non-pioneer species, as seen in our study. This indicates that some seeds were brought from surrounding forest areas by birds attracted by the perches (Cole *et al.* 2010; Cavallero *et al.* 2013). Dispersal limitation is predominant in degraded and fragmented landscapes, in consequence of the scarcity of animals, since tropical forest trees are mainly animal-dispersed and degraded places do not present much refuge and food for animals (Martinez-Ramos & Soto-Castro 1993; Hubbell *et al.* 1999). Our results indicate that seeds arrive, but still in rather small numbers. If we compare the proportion of non-grass species with grasses (38% vs. 62%) and consider that within these non-grasses only

a small portion are of zoothorax non-pioneer species, we might conclude that many of desired species are limited by dispersal and the majority of seeds sampled apparently were dispersed from within the degraded zones (high number of grasses and potential weed species). Even that, the presence of perches greatly enhanced the arriving of non-grass species (with perches 55% vs. 12% without) and within this group a higher proportion of seeds could be identified as zoothorax non-pioneer species (Fig. 2), which highlight the importance to overcome the barrier of dispersal.

Seed traits differed between seed traps with and without perches, which that the presence of perches, besides increase the number and richness of seeds, also helped to attract a set of seeds with distinct traits, benefiting biodiversity and meeting the goals of ecological restoration (Holl 1999; Halme *et al.* 2013). The use of perches often increases the number of zoothoric and non-pioneer species (Cole *et al.* 2010; Cavallero *et al.* 2013), although the relation with other seed traits and the use of perches are not clear yet, such as with seed color, as found by us (Barnes & Chapman 2014). Even though the length and diameter of seeds did not present great differences, the seed form (relation between length:diameter) did. Seeds of rounded form were more abundant in traps with perches; while more elongated seeds were in traps without. Grass and Asteraceae species commonly present elongated seeds and, especially for Asteraceae, they are known as typically anemocoric species (van der Pijl 1972). On the other hand, rounded seeds are frequently associated to fleshy fruits dispersed by animals (Howe & Westley 1997), which might justify its higher proportion in traps under perches.

Seedling recruitment and seed rain of traps under perches were related concerning patterns of species composition distribution and abundance (Fig. 3), which does not necessarily mean that the same species have been found in the two sets of data (regeneration and seed rain). This is a convergent pattern, expressed through co-inertia analysis ( $RV = 0.45$ ), showing that plots with distinct composition of seed rain also are distinct regarding composition of recruits.

The ordination diagram is showing that many plots with a large variety of seed rain composition have almost any regenerating species (top left plots), while some of the two other set of plots (top right and bottom left) have already some woody species in the regeneration strata (e.g. *Senecio brasiliensis*, *Trema micrantha*, *Myrsine coriacea*, *Piptocharpa tomentosa*, *Solanum pseudoquina*) and they were also distinguished by their seed rain composition (e.g. *Ilex* sp., *Psychotria* sp., *Solanum* sp., *Ficus* sp., *Smilax* sp., *Paspalum* sp., *Trema micrantha*, *Urochloa brizantha*). Interestingly was that for seed rain without perches, co-inertia analysis was not significant. The correspondence between the seeds arriving under the perches (probably through birds) and the regeneration patterns of woody species, clearly show the presence of dispersal limitation in natural regenerating, even considering that the effective number of recruits was small.

The convergent pattern observed in the co-inertia analysis led us to expect that plots with a higher proportion of woody species in the seed rain would also present a higher proportion of recruitments, but this was not true. Abundance of seed rain and recruitment of woody species were even negatively related to each other (Fig. 4a). We interpret this as an indicative that local establishment filters are stronger than the dispersal limitation filter. High dispersion in the data could be seen, potentially related to local environmental conditions, such as temperature, soil chemistry, humidity, and the competition pressure by exotic grasses, which limit even species planted as seedlings (Peña-Domene *et al.* 2013).

The presence of different filters acting first on seed arrival probability and then on the establishment of plants in our total set of study plots, related both to landscape and site features, is certainly limiting the development of the plant community and preventing restoration success (D'Antonio 1993; Batten *et al.* 2005; Avio *et al.* 2013). The recruitment of woody species is often negatively influenced by cover of exotic grasses (Batten *et al.* 2005; Ortega-Piecka *et al.* 2011; von Gillhaussen *et al.* 2014), but this was not significant in our

study. Nevertheless, more woody plants established in places with higher coverage of forbs. As the cover of forbs and grasses was indirectly related in the study areas (Pearson correlation= -0.26), exotic grasses might still limit the recruitment of woody species, even though enhancement of establishment success at sites with more forbs was clearer, which may be related to different site conditions. It has been shown that forbs are less competitive to woody recruits and that, at the same time, they reduce microclimatic extremes (Holl 1999). Our results indicate the importance of priority effects for restoration success (von Gillhaussen *et al.* 2014), which can here be related to the time that different groups of species arrived the sites. If forbs covered the restoration areas, the ongoing restoration trajectory might be very different than if exotic grasses achieved high cover and outcompeted other plant life forms.

In this study we emphasize the importance of two processes, seed rain and seedling recruitment, and their relationships for the ongoing ecological restoration of high-degraded sites. The use of perches effectively enhances the seed rain of desired woody species, and let to establishment of functionally different species. The use of perches thus also increases functional diversity in restoration. Furthermore, the natural establishment of woody species was improved by the cover of forbs. It thus may be beneficial for restoration success to seed forbs instead of grasses (and more so, exotic invasive grasses, as had been used in our case) to establish a first vegetation cover and retain soil erosion, or to prevent the establishment of exotic invasive grasses from surrounding areas.

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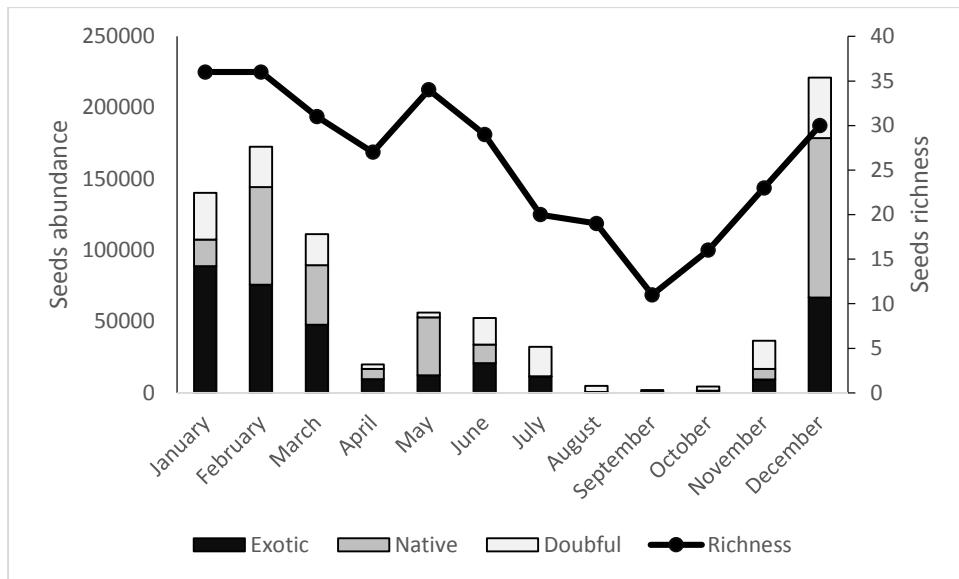
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**Appendix 1.** Species collected on the seed rain sampling recorded in areas process of ecological restoration.

Family	Species	Species code	Abundance do seeds (m <sup>2</sup> /year)
Anacardiaceae	<i>Schinus terebinthifolius</i> Raddi	scte	4.37
Aquifoliaceae	<i>Ilex</i> sp. 1	ile1	1.83
Aquifoliaceae	<i>Ilex</i> sp. 2	ile2	0.52
Asteraceae	Asteraceae 1	Ast1	344.08
Cannabaceae	<i>Trema micrantha</i> (L.) Blume	trmi	25.54
Clethraceae	<i>Clethra scabra</i> Loisel.	clsc	341.08
Convolvulaceae	<i>Ipomea</i> sp.	ipom	7.50
Erythroxylaceae	<i>Erythroxylum deciduum</i> A. St.-Hil.	erde	0.63
Euphorbiaceae	<i>Alchornea</i> sp.	alch	2.08
Euphorbiaceae	<i>Ricinus communis</i> L.	rico	23.75
Lamiaceae	<i>Aegiphila sellowiana</i> Cham.	aese	0.90
Magnaliaceae	<i>Talauma ovata</i> A. St.-Hil.	taov	0.38
Melastomataceae	<i>Miconia cabucu</i> Hoehne	mica	1.48
Melastomataceae	<i>Miconia</i> sp.	mico	1252.67
Moraceae	<i>Ficus</i> sp.	ficu	35.54
Oxalidaceae	<i>Oxalis</i> sp.	oxal	41.04
Phyllantaceae	<i>Hyeronima alchorneoides</i> Allemão	hyal	1.19
Phytolaccaceae	<i>Phytolacca americana</i> L.	pham	1529.29
Poaceae	<i>Avena sativa</i> L.	avsa	0.46
Poaceae	<i>Echinochloa</i> sp.	echi	4920.13
Poaceae	<i>Paspalum</i> sp. 1	pas1	11.08
Poaceae	<i>Paspalum</i> sp. 2	pas2	78.27
Poaceae	<i>Paspalum urvillei</i> Steud.	paur	4213.25
Poaceae	<i>Setaria</i> sp.	seta	9.19
Poaceae	Poaceae 1	poa1	21.48
Poaceae	Poaceae 2	poa2	32.38
Poaceae	Poaceae 3	poa3	619.04
Poaceae	Poaceae 4	poa4	110.21
Poaceae	Poaceae 5	poa5	11.58
Poaceae	<i>Urochloa brizantha</i> (Hochst. ex A. Rich.)		
Poaceae	R.D. Webster	urbr	128.88
Polygonaceae	<i>Polygonum</i> sp.	poly	259.27
Portulacaceae	<i>Portulaca cf. oleracea</i> L.	pool	8.17
Primulaceae	<i>Myrsine cf. umbellata</i> Mart.	myum	65.60
Rosaceae	<i>Rubus rosaefolius</i> Focke	ruro	14.73
Rubiaceae	<i>Psychotria</i> sp. 1	psy1	4.98
Rubiaceae	<i>Psychotria</i> sp. 2	psy2	0.96
Solanaceae	<i>Solanum</i> sp. 1	sol1	1.33
Solanaceae	<i>Solanum</i> sp. 2	sol2	1986.08
Solanaceae	<i>Solanum</i> sp. 3	sol3	7.63
Solanaceae	<i>Solanum</i> sp. 4	sol4	1.25
Solanaceae	Solanaceae 1	sola	27.63
Unidentified	Unidentified 1	uni1	213.35
Unidentified	Unidentified 2	uni2	5.29
Unidentified	Unidentified 3	uni3	0.38
Unidentified	Unidentified 4	uni4	0.54

Unidentified	Unidentified 5	uni5	16.87
Unidentified	Unidentified 6	uni6	16.96
Unidentified	Unidentified 7	uni7	13.19
Unidentified	Unidentified 8	uni8	1.12
Unidentified	Unidentified 9	uni9	2.67
Unidentified	Unidentified 10	uni10	1.06
<hr/>			
Total			16418.88

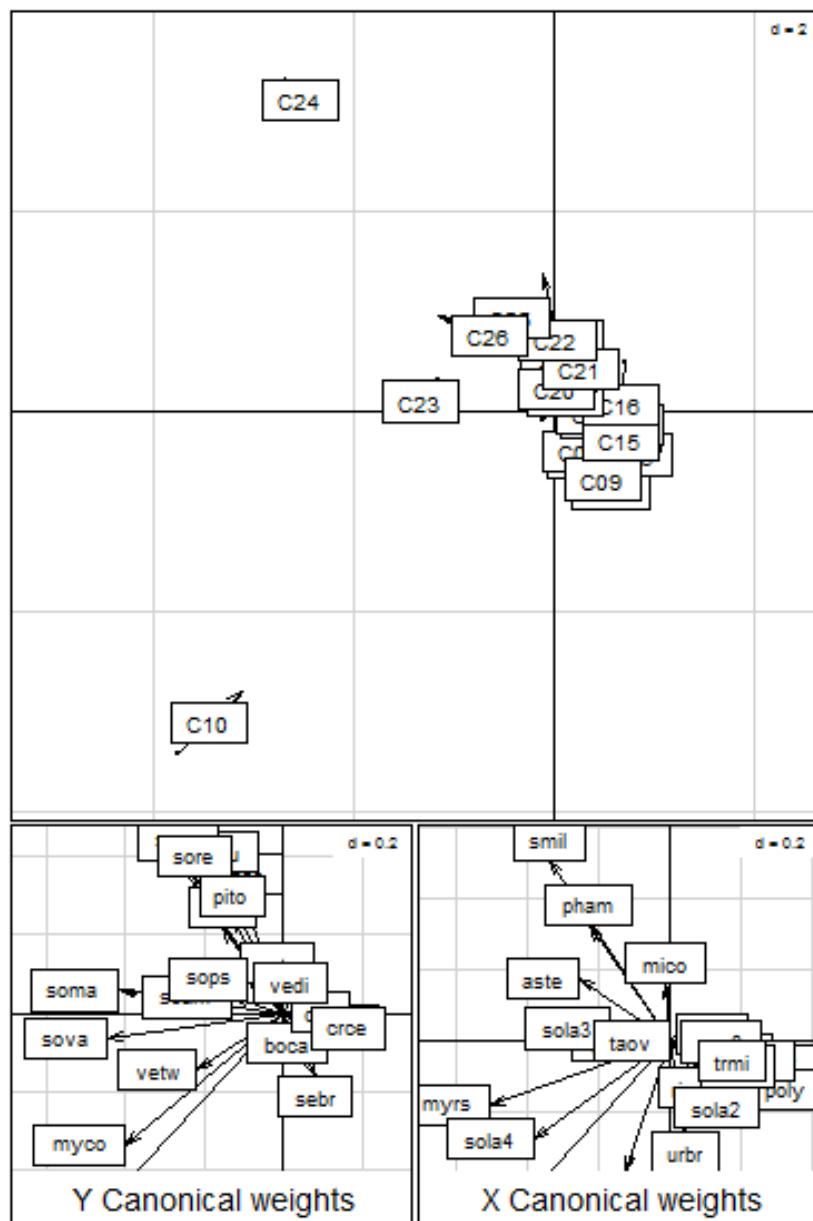
**Appendix 2.** Abundance of seed rain of exotic, native and doubtful species and the richness of seeds.



**Appendix 3.** Species collected on the wood natural regeneration (seedling recruits) in areas process of ecological restoration.

Family	Species	Specie code	Abundance
Anacardiaceae	<i>Schinus terebinthifolius</i> Raddi	scte	3
Asteraceae	<i>Baccharidastrum triplinervium</i> (Less.) Cabrera	batr	2
Asteraceae	<i>Baccharis semiserrata</i> DC.	base	86
Asteraceae	<i>Kaunia rufescens</i> (P.W. Lund ex DC.) R.M. King & H. Rob.	karu	62
Asteraceae	<i>Piptocarpha tomentosa</i> Baker	pito	22
Asteraceae	<i>Senecio brasiliensis</i> (Spreng.) Less.	sebr	22
Asteraceae	<i>Vernonanthura discolor</i> (Spreng.) H. Rob.	vedi	1
Asteraceae	<i>Vernonanthura tweedieana</i> (Baker) H. Rob.	vetw	17
Cannabaceae	<i>Trema micrantha</i> (L.) Blume	trmi	1
Euphorbiaceae	<i>Croton celtidifolius</i> Baill.	crce	20
Fabaceae	<i>Senna</i> sp.	senn	1
Lamiaceae	<i>Aegiphila sellowiana</i> Cham.	aese	1
Primulaceae	<i>Myrsine coriacea</i> (Sw.) R.Br. ex Roem. & Schult.	myco	10
Rutaceae	<i>Citrus</i> sp.	citr	1
Solanaceae	<i>Solanum americanum</i> Mill.	soam	1
Solanaceae	<i>Solanum mauritianum</i> Scop.	soma	18
Solanaceae	<i>Solanum pseudocapsicum</i> L.	sops	3
Solanaceae	<i>Solanum reflexum</i> Dunal	sore	4
Solanaceae	<i>Solanum variabile</i> Mart.	sova	27
Urticaceae	<i>Boehmeria caudata</i> Sw.	boca	4

**Appendix 4.** Relationship between patterns of seed rain and plant recruitment at the restoration sites for seed traps without perches ( $RV=0.46$ ;  $p=0.16$ ). The co-inertia analysis shows the ordination of plots (distinguished by plot number) described by seed rain and woody plant recruitment based on abundances. Shorter arrows indicate better adjustment of patterns in both sets of data (seed rain species and recruits of woody species). The PCAs of both data sets are shown separately at the bottom (recruitment patterns at the left; seed rain patterns at the right; the species codes are in the species list: Appendix 1 for seed rain, Appendix 3 for recruits).



## CAPÍTULO 3

### FOREST RESTORATION AFTER SEVERE DEGRADATION BY COAL MINING: LESSONS FROM THE FIRST YEARS OF MONITORING



# **Forest restoration after severe degradation by coal mining: lessons from the first years of monitoring**

Edilane Rocha-Nicoleite<sup>1,2</sup>, Mari Lucia Campos<sup>3</sup>, Guthieri Teixeira Colombo<sup>2</sup>, Gerhard Ernst Overbeck<sup>1,4</sup> & Sandra Cristina Müller<sup>1,5</sup>

<sup>1</sup>Universidade Federal do Rio Grande do Sul, Graduate Program in Ecology, Av. Bento Gonçalves 9500, 91540-000, Porto Alegre, RS, Brasil

<sup>2</sup>Associação Beneficente da Indústria Carbonífera de Santa Catarina, Clean Coal Research Centre, Rua Pascoal Meller 73, 88805-380, Criciúma, SC, Brasil;

<sup>3</sup>Universidade do Estado de Santa Catarina, Department of Soil Science, Av. Luiz de Camões 2090, 88520-000, Lages, SC, Brasil;

<sup>4</sup>Universidade Federal do Rio Grande do Sul, Department of Botany, Av. Bento Gonçalves, 9500, 91540-000, Porto Alegre, RS, Brasil.

<sup>5</sup>Universidade Federal do Rio Grande do Sul, Department of Ecology, Av. Bento Gonçalves, 9500, 91540-000 Porto Alegre, RS, Brasil.

## **Abstract**

We analysed woody plant species regeneration in the first years of restoration in areas severely degraded by coal mining in the South Brazilian Atlantic Rainforest. In the four riparian forest under restoration and in two forest remnants, we collected data on composition and cover of the herbaceous layer (including invasive grasses), abundance and richness of introduced trees and spontaneous woody regeneration, traits woody species, and soil chemistry. We compared characteristics of the woody regeneration with the forest remnants as target communities and evaluated the dynamics of regeneration over three years by principal coordinates analyses

(PCoA). The influence of biotic and abiotic variables for woody species regeneration was analysed by partial redundancy analysis (RDAP). We found great variation in relation to community composition and structure between restored sites. Most of this variation was explained by variables related to soil chemistry, introduced trees and cover of exotic grasses. Fertilization of soil, as commonly recommended, seems to increase cover of exotic grasses, which clearly impede the development of planted trees and woody regeneration. Over time, an increase in woody species establishment could be observed. Although the restoration of abandoned mining areas is a challenge for restoration ecology, our results, despite the short period of observation and high mortality of planted trees, do allow for the conclusion that the restoration of successional trajectories after severe degradation is possible. We highlight the importance to monitor the initial process of restoration and of (re)defining intermediate goal and project targets, following an adaptive management approach.

**Keywords** – Adaptive management; Invasive grasses; Herbaceous cover; Soil fertilization; Atlantic Rainforest; Woody regeneration; Tree planting; Restoration Ecology.

## **Introduction**

The ecological restoration of areas affected by mining activities can be very difficult and often requires different engineering approaches and multidisciplinary knowledge (Zipper *et al.* 2011; Adibee *et al.* 2013). Reduction and containment of pollution and repairing past environmental damage often are primary goals to establish vegetation close to natural condition (ecological restoration) have been conducted worldwide, but severe limitations related to native species establishment and initiation of natural succession processes exist (Fields-Johnson *et al.* 2012; Zhenqi *et al.* 2012; Bauman *et al.* 2013). Often, these are consequence of strong interventions required to change the abiotic and biotic characteristics of the degraded systems.

Plantings of species is considered an important strategy to overcome recruitment limitations (Omeja *et al.* 2011), also enhancing, in the long run, succession through the attraction of seed dispersing animals and the creation of favourable microclimatic conditions. However, after surface-mining activities, the soil often needs to be completely reconstructed. The resulting substrate presents no natural conditions regarding physical structure and chemical composition, and generally is strongly compacted (Kämpf *et al.* 1997; Bassett *et al.* 2005), reducing survival of plants. The presence of invasive species, mostly exotic grasses which often are planted, can constitute an additional major difficulty in post-mining restoration projects (Nyamai *et al.* 2011; Bennett *et al.* 2012). Grasses limit the establishment and diversity of native plant species because they are strong competitors. Additionally, they often increase the incidence of fire (Veldman *et al.* 2009) which further impedes the colonization by native woody species (Martínez-Garza *et al.* 2005; McGlone *et al.* 2012).

The trajectories of plant communities in restoration projects can vary widely (Chazdon *et al.* 2007). Understanding the role of the principal biotic and abiotic factors during

all restoration phases is very important to improve ecological theory and practice (Block *et al.* 2001; Miller & Hobbs 2007). Differences to the planned or desired trajectories may become obvious early in restoration. In highly impacted lands, monitoring of restoration success in the first years is especially important, as successional pathways are initiated and may then drive regeneration processes and vegetation dynamics for long periods (Zipper *et al.* 2011; Fields-Johnson *et al.* 2012; Zhenqi *et al.* 2012; Bauman *et al.* 2013). Early monitoring is crucial for the implementation of adaptive management that responds to the development of the restoration site (Lindenmayer & Likens 2009).

Here, we present results from a study situated in a region severely affected by coal mining in the southern part of the Brazilian Atlantic Rainforest. For many decades, before rise of environmental consciousness, no attempts to reduce pollution or restoration had been undertaken at all in these areas. Waste material from coal mines was deposited without any concern. Runoff water from polluted areas and from mines has caused severe contamination of watersheds in the past 130 years when the coal mining began in this region (Silva *et al.* 2013). At present, in rivers pH values may reach 3.0 and environmental conditions present severe health risk for human population in the region (Santos *et al.* 2008; Silva *et al.* 2011; Silva *et al.* 2013). In 2000, restoration of all riparian zones in the region became mandatory (Brasil 2013), and remnants were defined as reference sites. Since this time, restoration projects have been planned and implemented, but restoration success has not been assessed thoroughly in these areas characterized by high environmental damage.

We studied the regeneration of shrubs and trees in four areas undergoing ecological restoration and looked specifically at the first years of these projects. Our goals were: (1) to evaluate the composition, abundance and ecological features of woody species that are regenerating at the restoration sites, (2) to assess changes in species composition during three years of monitoring, (3) to analyse the influence of abiotic and biotic variables over patterns of

natural regeneration and (4) to develop realistic restoration aims for the initial phase of restoration of highly degraded sites. We expected responses of species composition to key abiotic and biotic variables, such as soil nutrients and abundance of invasive grasses and planted trees. We also expected different patterns and trajectories in relation to the age of the restoration areas, with the oldest area (six years) being already more similar to the reference forests.

## Methods

### *Study region*

This study was conducted in southern Santa Catarina state, Brazil. Study areas are under humid subtropical climate conditions, with annual temperature of 19 °C and rainfall average of 1600 mm (Alvares *et al.* 2014). Originally, this region was completely covered by Atlantic Rainforest (Ribeiro *et al.* 2009; Teixeira *et al.* 2009), part of a global biodiversity hotspot (Myers *et al.* 2000).

We conducted this research at four sites (28°34' S, 49°25' W - 28°25' S, 49°25' W) currently under ecological restoration. All sites had been degraded by surface coal mining with deposition of spoils in riparian zones. Original vegetation was riparian forest. The principal restoration goal is the reestablishment of the pre-mining forest communities. As reference areas, we studied two riparian forest remnants without any history of mining (areas R1 and R2) in the same region (R1: 28°34' S - 49°24' W; R2: 28°26' S - 49°25' W).

Restoration of four study sites followed the general procedures applied in the region: the first step to restore these areas was removal of spoils to stop ongoing contamination. After this, a new substrate was created, composed of clay or sand (depth ca. 2 m), in order to provide support for vegetation and to construct the topography. Soil construction was finished

with addition of a thin layer organic matter (approximately 2 cm), e.g. peat and broiler litter. Then a vegetation cover was established by sowing of exotic grasses (in order to rapidly cover the soil, which is recommended by law), followed by planting or seeding of native trees.

In the studied sites the following exotic grasses were introduced: in area A *Urochloa decumbens* (Stapf) R.D. Webster (Poaceae; 18 kg.ha<sup>-1</sup>), in areas B e C *Avena sativa* L. (Poaceae) and *Lolium multiflorum* Lam. (Poaceae; 30 kg.ha<sup>-1</sup>) and in area D *Melinis minutiflora* P. Beauv. (Poaceae; 15 kg.ha<sup>-1</sup>). Following this, 30 native tree species were planted with a density of 2500 individuals per hectare in areas A, B and C. In area D, one native tree, *Mimosa scabrella* Benth. (Fabaceae), was sowed (3 kg.ha<sup>-1</sup> with latter cutting of some individuals to reach a density of around 5000 individuals per hectare) and seedlings of 16 species were planted at 20 nucleus. These interventions were carried out between 2009 and 2010 in areas A, B and C (coal mined between 1975 and 1997) and between 2002 and 2005 in area D (coal mined between 1950 and 1989).

### *Vegetation surveys*

Per area, six (areas B and D) or 10 (areas A and C) plots of 10 × 30 m were established for sampling (a) survived planted trees and (b) woody plants in the upper stratum (diameter at breast height (dbh) ≥ 5 cm). In subplots of 1 × 1 m (three per plot), we recorded the composition of the herbaceous component, by visual estimation of cover of all species. In subplots of 5 × 5 m (three per plot), the woody regeneration stratum, composed of individuals with height ≥ 50 cm and ≤ 5 cm dbh, was sampled. Additionally, we estimated the cover of three categories of herbs: exotic grasses, native grasses, and forbs. The regeneration stratum and the categories of herbs were evaluated three times (A, B and C areas: 10, 22 and 34 months after tree planting; D area: 6, 7 and 8 years after tree planting). The reference areas were evaluated based on 30 plots with 10 × 10 m in each remnant forest (R1 and R2) to sample the upper

stratum, with one subplot of  $5 \times 5$  m to regenerating stratum and four  $1 \times 1$  m subplots to the herbaceous component within each plot. Sampling procedures were the same as at the restoration sites.

All plant species were identified in the field or a sample was collected for later identification by help of bibliography and/or consultation of herbaria and specialists. Classification into families follows APG III (2009). For all regenerating woody species, some ecological traits considered important for the restoration process were compiled from the literature.

#### *Analysis of soil properties*

Samples of the surface soil layer (0–20 cm) were collected in each subplot of  $5 \times 5$  m at the restored and remnant sites for evaluation of soil chemistry composition. Laboratory methods followed Tedesco *et al.* (1995). The samples were dried at a temperature of 60 °C for a 24 hours. Soil *pH* was determined in water solution by a potentiometer using soil/solution ratio 1:1; soil organic matter content (*OM*) by Walkley Black method,  $H+Al$  and  $Al^{3+}$  were determined by NaOH titulometry. *P*, *K* and *Na* elements were extracted using a Mehlich<sup>-1</sup> extractor and determined by flame photometry (*K*, *Na*) and colorimetric method (*P*). Calcium ( $Ca^{+2}$ ) and Magnesium ( $Mg^{+2}$ ) were extracted using KCl<sup>1</sup> mol L<sup>-1</sup> and measured using atomic absorption spectrophotometry.

#### *Data analysis*

As plots of  $10 \times 30$ –m were considered as sampling units, density data obtained in subplots were pooled together and extrapolated to 100 m<sup>2</sup> to standardize the analyses.

The temporal dynamics of natural regeneration were analysed by Principal Coordinate Analyses (PCoA), using abundance data of the woody regeneration stratum and

cover of herb categories recorded at the three sampling dates. Data were previously standardized and centralized within variables. Chord distance between sampling units was used as similarity measure. This analysis was done independently for each study site to preserve the particularity of each area, avoiding statistical comparisons between ongoing restoration areas and aiming at identifying successional trajectories.

We evaluated the proportion of potential biotic and abiotic influences on natural regeneration patterns considering different multiple data sets in partial Redundancy Analyses (RDAp) (Borcard *et al.* 2011). Response matrices were species abundances in the woody regeneration stratum ( $Y_R$ , i.e. sampling units without any woody individuals were not considered) and species abundances together with cover of herb categories ( $Y_T$ , i.e. including sampling units without any woody individuals). Explanatory matrices were soil chemistry, structural variables of the upper stratum (upper stratum for area D and planted trees for areas A, B and C, considered species abundances, species richness, and basal area), and abundances of the herbaceous component. Response matrices were based on data of the third evaluation. Both  $Y_R$  and  $Y_T$  were Hellinger transformed (Legendre & Gallagher 2001) and then submitted to RDAp, considering the values of adjusted  $R^2$  (Peres-Neto *et al.* 2006).

Before RDAp procedures, each original explanatory matrix (soil chemistry, structural variables of superior stratum, and species composition of herbaceous component) was submitted to a forward selection analysis to identify the most important subset of variables explaining variance of the response matrix. This was done with the BIOENV function, considering  $p<0.01$  after 999 randomizations (Blanchet *et al.* 2008). This and the RDAp were done by using the package *vegan* (Vegan: Community ecology package) on the R platform (R Development Core Team, <http://www.Rproject.org>)

Dispersion patterns of natural regeneration according to studied sites were shown by ordination diagrams of PCoA based on matrix  $Y_T$  and plotting the biotic and/or abiotic

variables that were significant in explaining natural regeneration in the areas (evidenced by RDAP results). Ordination analyses were carried out by MULTIV software (available at <http://ecoqua.ecologia.ufrgs.br/ecoqua/software.html>).

## Results

### *Species richness and ecological traits of regenerating woody species*

Altogether, 47 woody species were detected in the regenerating woody stratum at the third evaluation (Appendix 1). Considerable differences concerning richness and abundance of this stratum existed among areas. In area B, only five species (abundance: 2.44 ind.100m<sup>-2</sup>) were sampled, while in area D, 37 species were found (abundance: 71.11 ind.100m<sup>-2</sup>). Even if we only consider the areas with the same age of restoration (A, B, C), strong differences in composition and abundance were obvious.

At the two remnant sites, 106 species were found (abundance 22 ind.100m<sup>-2</sup>) in the woody regenerating stratum, with few compositional differences between each other (Appendix 2). Only nine species were recorded at sites under restoration and reference sites (*Boehmeria caudata*, *Casearia silvestris*, *Euterpe edulis*, *Inga marginata*, *Matayba guianensis*, *Miconia cabucu*, *Miconia sellowiana*, *Piper aduncum* and *Zanthoxylum rhoifolium*).

The ecological traits (Appendix 3) of the woody species found in the regeneration stratum at the restoration sites were indicative of a very early succession stage of woody vegetation, as shown by the high percentage of anemochorous species and of pioneer species. Area D, the area with longer time of restoration, was somewhat more similar to reference sites, despite clear differences in species richness and composition.

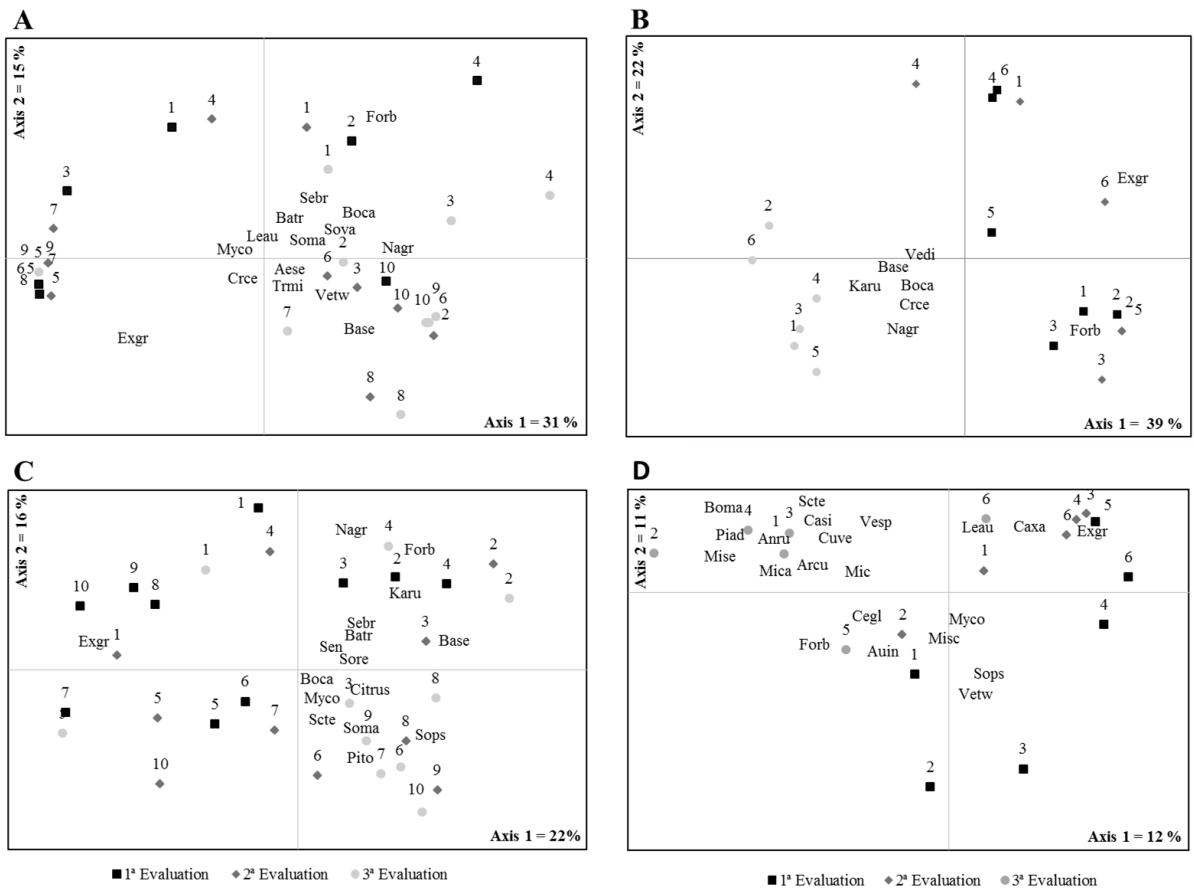
Only D area presented an upper stratum (dbh≥5 cm), with 14 species (abundance 11 ind.100m<sup>-2</sup>) (Appendix 4). In contrast, the reference areas presented 123 species (abundance

20 ind.100m<sup>-2</sup>) in this stratum (Appendix 5) and only three species were shared with area D (*Alchornea triplinervia*, *Annona rugulosa* and *Cecropia glaziovii*).

In the herbaceous layer of the restored areas, we sampled 9.3 species.m<sup>-2</sup>, and 86 species of 34 families (Appendix 6), with area C presenting highest richness. All areas presented high cover of exotic grasses, especially areas B and A with 83% and 74% of mean cover, respectively (Appendix 7). In the reference areas, we sampled 0.1 species.m<sup>-2</sup>, and 27 species of 17 botanic families (Appendix 8).

#### *Regenerating dynamics of woody species*

Dynamics in the regenerating stratum along the time of evaluations were high for all areas, although the patterns varied considerably among sites (Fig. 1). For all areas, changes were more evident when comparing the first and third evaluation, where most part of the plots shifts completely the side of the diagrams. Exotic grasses, especially in areas A, B and C, markedly dominated plots in the first evaluation. In the third evaluation, the regeneration of woody species became stronger in all areas, indicating that a large number of species arrived and/or increased in abundance.

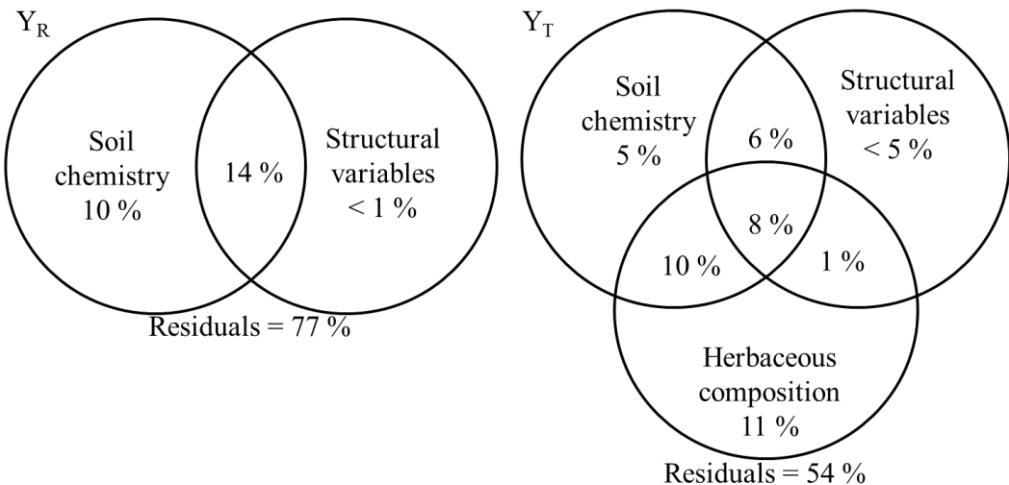


**Figure 1.** Ordination diagram of the areas under ecological restoration (A, C and D) based on woody species of the regenerating stratum and on the proportion of herb categories (exotic grasses (*exgr*), native grasses (*nagr*) and forbs), measured through three evaluations (A, B and C areas: 10, 22 and 34 months after tree planting; D area: 6, 7 and 8 years after tree planting). The numbers identify the plots of each area at time. Most closely correlated species are shown in the respective diagram (Altr – *A. triplinervia*; Auin – *A. inulifolium*; Bagl – *B. glaziovii*; Base – *B. semiserrata*; Batr – *B. triplinervium*; Cacaj – *C. cajan*; Cegl - *C. glaziovii*; Citrus - *Citrus sp.*; Crce – *C. celtidifolius*; Eued – *E. edulis*; Eupi – *E. picturatum*; Eur – *E. rufescens*; Leau – *L. australis*; Magu – *M. guianensis*; Mica – *M. cabucu*; Myco – *M. coriacea*; Ocpu – *O. puberula*; Piad – *P. aduncum*; Pito – *P. tomentosa*; Piza – *P. zapallo*; Rico – *R. communis*; Scte – *S. terebinthifolia*; Sebr – *S. brasiliensis*; Soma – *S. mauritianum*; Sops – *S. pseudocapsicum*; Sovi – *S. viarum*; Vetw – *V. tweedieana*; Wain – *W. indica*).

#### Influence of abiotic and biotic variables on natural regeneration

In pRDA, different subsets of explanatory variables were chosen for each of the two response matrices ( $Y_R$  and  $Y_T$ ) (Appendix 9), and 24% and 46% of the variation of the matrices  $Y_R$  and  $Y_T$ , respectively, were explained by the abiotic and biotic variables analysed (Fig. 2). For matrix  $Y_R$ , the major part of explanation was achieved with the

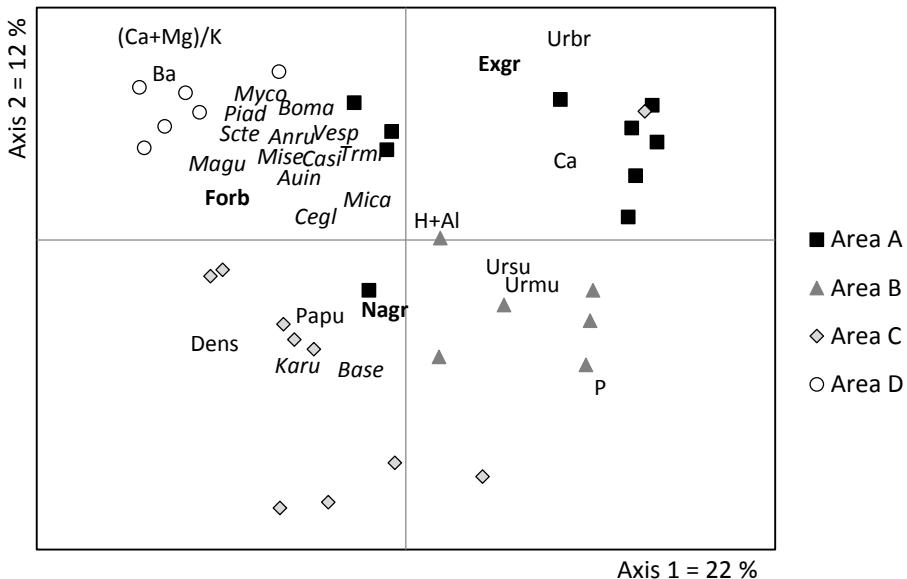
sharing between both sets of explanatory variables (soil chemistry and vegetation structure; Fig. 2  $Y_R$ ). For matrix  $Y_T$ , species composition of the herbaceous component presented the highest explanation, followed by the shared portion between herbaceous composition and soil chemistry, and between all subsets analysed (Fig. 2  $Y_T$ ).



**Figure 2.** Venn diagram with the adjusted  $R^2$  results of the variance partition (RDAp) according to the subsets of variables significantly related to the response matrices (see Table 3), ( $Y_R$ ) considering only the composition of woody species regenerating and ( $Y_T$ ) considering all plots evaluated, even those without regenerating woody species, but with the addition of herbaceous categories cover as response variables.

Ordination of the full dataset (using data from last evaluation) showed clear overall differences among the four areas (Fig. 3). Plots of areas A and B were placed on the same side of the diagram, and were associated to high cover of exotic grasses, especially *Urochloa subquadripila*, *U. brizantha* and *U. mutica*. These areas were also characterized by high values of  $P$ ,  $Ca^{+2}$  and  $H+Al$  in the constructed soil. The proportion of woody species regenerating was clearly low for areas A and B, contrasting with plots on the left side of the diagram, where areas C and D were situated, although clearly separated along the second axis. Plots of area D were positively related to density and richness of planted trees and upper stratum and to the basal areas of the upper stratum.

Plots of areas C were highly associated with the two pioneer shrubs *Baccharis semiserrata* and *Kaunia rufescens*.



**Figure 3.** Principal coordinates ordination diagram of all plots described by the abundance of woody regenerating species plus the cover of herbs categories (matrix  $Y_T$ ), according to the four studied sites. Most correlated species and herbs categories to both axes are plotted in the diagram (italic font: *Anru* – *A. rugulosa*; *Auin* – *A. inulifolium*; *Base* – *B. semiserrata*; *Boma* – *B. macrophylla*; *Casi* – *C. silvestris*; *Cegl* – *Cecropia glaziovii*; *Karu* – *Kaunia rufescens*; *Magu* – *Matayba guianensis*; *Mica* – *M. cabucu*; *Mica* – *Miconia cabucu*; *Mise* – *bnnn*; *Myco* – *Myrsine coriacea*; *Piad* – *Piper aduncum*; *Scte* – *Schinus terebinthifolia*; *Trmi* – *T. micrantha*; *Vesp* – *Vernonanthura* sp.; bold font: *Exgr* – exotic grasses; *Nagr* – native grasses; *Forb* - forbs). Furthermore, abiotic (soil chemistry: *Ca* – calcium; *H+Al* – potential acidity; *P* – phosphorus;  $(Ca+Mg)/K$  – relation between that cations) and biotic variables (herbaceous species: *Ursu* – *U. subquadripila*; *Urbi* – *U. brizantha*; *Urmu* – *U. mutica*; *Papu* – *P. pumilum*; structural variables of upper stratum: *Ba* – basal area; *Dens* – density) were placed according to its axes correlation value.

## Discussion

In ecological restoration, reference areas are commonly used to evaluate the development of areas under restoration, based on parameters such as abundances of species and proportion of species with ecological characteristics considered typical for the vegetation type in question, or indicative or desirable ecological process. However, in

highly degraded systems that demand strong intervention, such as the post-mining areas studied by us, direct comparison should be done with caution. Abundance of species has been pointed out not to be a clear indicator before the successional phase of the area under restoration does not match that of the reference community (Martinez-Ramos & Soto-Castro 1993; Chazdon *et al.* 2007).

Highly impacted lands need a considerable time to allow the establishment of natural regeneration processes (Liu *et al.* 2011; Zipper *et al.* 2011; Zhenqi *et al.* 2012), and that areas like those studied by us still differ considerably from the reference areas thus is not surprising. However, even after a relatively short time (*e.g.* in area D, eight years after beginning of restoration) we could see some similarities with the references sites, especially in the proportion of zoolochoric and non-pioneer species. Although still a long way from the reference communities, and at distinct successional phases, the difference of this site to the other three sites suggests that, despite the strong degradation and necessity for severe intervention such as soil construction, more time should allow for convergence of the vegetation to that from reference areas, contradicting those that criticize this kind of attempt to be totally infeasible for areas with this or similar kind of degradation (Tozer *et al.* 2011).

The high variability in species composition and abundance among sites seems to be determined both by the soil chemical composition (abiotic component) and by the development of upper stratum (biotic component). The values found for these sets of variables showed some clear relations among these factors, as well as with the herbaceous component, here dominated by exotic grasses, that may exert strong competition for the establishment of woody species individuals (Batten *et al.* 2005a; Avio *et al.* 2013b; Hui-na *et al.* 2014) and also increase the risk of fires (Veldman *et al.* 2009). Plots with high content of phosphorus (*P*) were related to high abundance of grasses and to lower

development of the planted trees, which resulted in low natural regeneration of woody species. This is probably a consequence of the way soil fertilization is commonly conducted in restoration projects together with the recommendation of seeding of grasses to rapidly cover the soil: nutrient values follow guidelines for agricultural systems, which favours the development of invasive grasses (Högberg 2007; Jung & Lal 2011; Calonego & Rosolem 2013; Fonte *et al.* 2014). Knowledge on nutritional requirements of native species is mostly missing; one study on native species from our region has shown that *P* uptake presented significant higher growth just for few pioneer species (Pasqualini *et al.* 2007). Interactions between soil variables seem also to occur. High *P* content, for instance, infers directly on absorption of other cations, as the micronutrient zinc (*Zn*), reducing its availability (Bingham 1963). Similarly the cations *Ca*, *Mg* and *K* compete and influence the absorption of each other, thus the equilibria of levels of these cations shows directly in the nutrition of the plants. In our study, the relation *Ca+Mg/K* was associated with plots better characterized by natural regeneration of native woody plants. While currently the nutritional demand of native plants are little known, a better knowledge of these relations can be an important guidance for the way the soil in restoration areas is prepared before introduction of native species, and more specific studies are necessary to improve restoration projects.

It is well known that the development of introduced trees strongly influences the progress of natural regeneration (Parrotta *et al.* 1997b; Omeja *et al.* 2011a), as seen in area D under the canopy of *Mimosa scabrella*, a fast growth pioneer. The canopy closure changes local conditions (e.g. microclimate), increases the potential for arrival of seeds (perches) and facilitates the establishment of native trees through the exclusion of invasive grasses by shading (Chazdon 2003; Lindenmayer *et al.* 2010; Nyamai *et al.* 2011; Omeja *et al.* 2011; Rodrigues *et al.* 2011). Besides, *M. scabrella* is a legume species

capable to fix nitrogen, which is considered of high relevance to improve degraded soil conditions (Lammel *et al.* 2013). The establishment and growth of first introduced species might then crucially contribute to restoration success due facilitation for other species and also trigger the further community development. These results and the importance of priority effects on community assembly processes (von Gillhaussen *et al.* 2014), make us think about which species should be selected for planting in restoration of highly degraded lands. While on the one hand the restoration aim may be the high diversity of species (Rodrigues *et al.* 2011), the first restoration steps should aim to re-establish the vegetation structure and conditions (Parrotta *et al.* 1997; Omeja *et al.* 2011) that will enable ecosystem and community assembly processes without further human interventions. At the same time, it should be carefully analysed which plant traits are able to survive under the rather harsh environmental conditions, e.g. on completely reconstructed soils.

Altogether, our study shows the necessity to establish realistic goals for each step or phase of the project, and the first goal should be related to canopy closure (Parrotta *et al.* 1997; Omeja *et al.* 2011) and soil improvement. Under strongly constraining abiotic and biotic conditions, far away from the reference state, the establishment of high species diversity is no such realistic goal, even though this might be the final goal (Failing *et al.* 2013). If we set initial and intermediate goals, the whole restoration process becomes more realistic. The perspective of restoration as a process implies that evaluation of restoration success considers initial, intermediate and final goals. Especially the analysis of trajectories over time seems important. Once trajectories stabilized and show a clear direction (which was not the case in the first three years of study for our young sites), it can be evaluated whether or not they will, in the future, converge with the reference community. To determine intermediate goals for projects of restoration looks like an

important step to evaluate trajectories and make decisions, such as that related with adaptive management.

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**Appendix 1.** Species of the regenerating woody species recorded in areas in process of ecological restoration, considering the last evaluation (areas A, B and C: 34 months after initial intervention with tree planting; areas D: 8 years after initial intervention with tree sowing). Given is abundance per area (standardized to individuals per 100 m<sup>2</sup>) at each study area.

Family / Species	Abundance (100 m <sup>2</sup> )			
	A	B	C	D
<b>Anacardiaceae</b>				
<i>Schinus terebinthifolius</i> Raddi	0.00	0.00	0.40	5.11
<b>Annonaceae</b>				
<i>Annona rugulosa</i> (Schltdl.) H.Rainer	0.00	0.00	0.00	1.78
<b>Arecaceae</b>				
<i>Archontophoenix cunninghamiana</i> H. Wendl. & Drude	0.00	0.00	0.00	0.44
<i>Euterpe edulis</i> Mart.	0.00	0.00	0.00	0.22
<b>Asteraceae</b>				
<i>Austroeupatorium inulifolium</i> (Baker) H. Rob.	0.00	0.00	0.00	1.33
<i>Baccharidastrum triplinervium</i> (Kunth) R.M. King & H. Rob.	0.13	0.00	0.13	0.00
<i>Baccharis semiserrata</i> (Less.) Cabrera	1.07	0.22	10.27	0.00
<i>Kaunia rufescens</i> (P.W. Lund ex DC.) R.M. King & H. Rob.	0.00	0.44	8.00	3.33
<i>Piptocarpha tomentosa</i> (Spreng.) Less.	0.13	0.00	2.80	0.00
<i>Senecio brasiliensis</i> Baker	2.93	0.00	0.00	0.00
<i>Vernonanthura discolor</i> DC.	0.00	0.22	0.00	0.22
<i>Vernonanthura</i> sp.	0.00	0.00	0.00	5.56
<i>Vernonanthura tweedieana</i> (Spreng.) H. Rob.	0.53	0.00	1.73	0.22
<b>Cannabaceae</b>				
<i>Trema micrantha</i> (L.) Blume	0.13	0.00	0.00	0.44
<b>Euphorbiaceae</b>				
<i>Alchornea glandulosa</i> (Spreng.) Müll. Arg.	0.00	0.00	0.00	0.44
<i>Alchornea triplinervia</i> Baill.	0.00	0.00	0.00	3.78
<i>Croton celtidifolius</i> Poepp.	1.87	1.33	0.00	2.67
<b>Fabaceae</b>				
<i>Inga marginata</i> (DC.) Kuntze	0.00	0.00	0.00	0.22

<b>Family / Species</b>	<b>Abundance (100 m<sup>2</sup>)</b>			
	<b>A</b>	<b>B</b>	<b>C</b>	<b>D</b>
<i>Mimosa bimucronata</i> (Vogel) H.S. Irwin & Barneby	0.00	0.00	0.00	1.11
<i>Mimosa scabrella</i> Benth.	0.00	0.00	0.00	0.89
<i>Senna</i> sp.	0.00	0.00	0.13	0.00
<b>Lamiaceae</b>				
<i>Aegiphila sellowiana</i> Cham.	0.13	0.00	0.00	0.00
<b>Lauraceae</b>				
<i>Ocotea puberula</i> (Rich.) Nees	0.00	0.00	0.00	0.22
<b>Melastomataceae</b>				
<i>Leandra australis</i> (Cham.) Cogn.	0.00	0.00	0.00	1.56
<i>Miconia cabucu</i> (DC.) Naudin	0.00	0.00	0.00	1.33
<i>Miconia ligustroides</i> Hoehne	0.00	0.00	0.00	0.22
<i>Miconia sellowiana</i> Naudin	0.00	0.00	0.00	1.56
<b>Myrtaceae</b>				
<i>Psidium guajava</i> L.	0.00	0.00	0.00	0.44
<b>Nyctaginaceae</b>				
<i>Pisonia zapallo</i> Griseb.	0.00	0.00	0.00	0.22
<b>Oleaceae</b>				
<i>Ligustrum lucidum</i> W.T. Aiton	0.00	0.00	0.00	0.22
<b>Piperaceae</b>				
<i>Piper aduncum</i> Aubl.	0.00	0.00	0.00	6.67
<i>Piper arboreum</i> L.	0.00	0.00	0.00	0.22
<b>Primulaceae</b>				
<i>Myrsine coriacea</i> (Sw.) R.Br. ex Roem. & Schult.	0.93	0.00	0.40	21.56
<b>Rosaceae</b>				
<i>Rubus rosifolius</i> Sm.	0.00	0.00	0.00	1.11
<b>Rutaceae</b>				
<i>Citrus</i> sp.	0.00	0.00	0.13	0.00
<i>Zanthoxylum rhoifolium</i> Lam.	0.00	0.00	0.00	0.22

Family / Species	Abundance (100 m <sup>2</sup> )			
	A	B	C	D
<b>Salicaceae</b>				
<i>Casearia silvestris</i> Sw.	0.00	0.00	0.00	0.44
<b>Sapindaceae</b>				
<i>Cupania vernalis</i> Aubl.	0.00	0.00	0.00	0.22
<i>Matayba guianensis</i> Cambess	0.00	0.00	0.00	0.44
<b>Solanaceae</b>				
<i>Solanum americanum</i> Dunal	0.00	0.00	0.13	1.11
<i>Solanum mauritianum</i> L.	0.53	0.00	1.87	0.00
<i>Solanum pseudocapsicum</i> Mart.	0.00	0.00	0.40	0.44
<i>Solanum reflexum</i> Mill.	0.00	0.00	0.53	0.00
<i>Solanum variabile</i> Scop.	2.00	0.00	1.60	1.78
<b>Urticaceae</b>				
<i>Boehmeria caudata</i> Hornem	0.27	0.22	0.13	0.00
<i>Boehmeria macrophylla</i> Snelth.	0.00	0.00	0.00	2.89
<i>Cecropia glaziovii</i> Sw.	0.00	0.00	0.00	0.44
<b>Total – individuals.100 m<sup>2</sup> (species richness)</b>	<b>10.67 (12)</b>	<b>2.44 (5)</b>	<b>28.67 (15)</b>	<b>71.11 (37)</b>

**Appendix 2.** Species sampled in the woody regenerating stratum at the reference areas R2 and R1 given is abundance per area (standardized per 100 m<sup>2</sup>).

Family	Species	Abundance (100m <sup>2</sup> )	
		R2	R1
Annonaceae	<i>Duguetia lanceolata</i> A. St.-Hil.	0.00	0.07
	<i>Guatteria australis</i> A. St.-Hil.	0.23	0.03
	<i>Annona neosericea</i> H. Rainer	0.03	0.00
	<i>Xylopia brasiliensis</i> Spreng.	0.10	0.00
Apocynaceae	<i>Aspidosperma parvifolium</i> A. DC.	0.03	0.07
Aracaceae	<i>Bactris setosa</i> Mart.	0.00	0.17
	<i>Euterpe edulis</i> Mart.	4.20	5.67
	<i>Geonoma gamiova</i> Barb. Rodr.	0.07	0.57
Bignoniaceae	<i>Jacaranda puberula</i> Cham.	0.00	0.03
Boraginaceae	<i>Cordia silvestris</i> Fresen.	0.03	0.00
Burseraceae	<i>Protium kleinii</i> Cuatrec.	0.10	0.10
Cardiopteridaceae	<i>Citronella paniculata</i> (Mart.) R.A. Howard	0.10	0.00
Chrysobalanaceae	<i>Hirtella hebeclada</i> Moric. ex DC.	0.00	0.07
Clethraceae	<i>Clethra scabra</i> Pers.	0.03	0.00
Clusiaceae	<i>Garcinia gardneriana</i> (Planch. & Triana) Zappi	0.40	1.40
Combretaceae	<i>Buchenavia kleinii</i> Exell	0.03	0.00
Cyatheaceae	<i>Alsophila setosa</i> Kaulf.	0.30	0.00
	<i>Cyathea corcovadensis</i> (Raddi) Domin	0.23	0.00
Elaeocarpaceae	<i>Sloanea guianensis</i> (Aubl.) Benth.	0.00	0.13
Euphorbiaceae	<i>Actinostemon concolor</i> (Spreng.) Müll. Arg.	0.33	1.63
	<i>Pachystroma longifolium</i> (Nees) I.M. Johnst.	0.03	0.00
Peraceae	<i>Pera glabrata</i> (Schott) Poepp. ex Baill.	0.10	0.00
Fabaceae	<i>Dahlstedtia pentaphylla</i> (Taub.) Burkart	0.00	0.03
	<i>Inga marginata</i> Kunth	0.13	0.13
	<i>Inga sessilis</i> (Vell.) Mart.	0.00	0.10
	<i>Senna multijuga</i> (Rich.) H.S. Irwin & Barneby	0.07	0.00
	<i>Zollernia ilicifolia</i> (Brongn.) Tul.	0.03	0.07
Lauraceae	<i>Aiouea saligna</i> Meisn.	0.10	0.00
	<i>Cinnamomum glaziovii</i> (Mez) Kosterm.	0.00	0.03
	<i>Endlicheria paniculata</i> (Spreng.) J.F. Macbr.	0.20	0.33
	<i>Nectandra megapotamica</i> (Spreng.) Mez	0.20	0.00
	<i>Nectandra membranacea</i> (Sw.) Griseb.	0.10	0.00
	<i>Nectandra oppositifolia</i> Nees & Mart.	0.13	0.03
	<i>Ocotea catharinensis</i> Mez	0.03	0.07
	<i>Ocotea indecora</i> Schott ex Meissner	0.03	0.07
	<i>Ocotea laxa</i> (Nees) Mez	0.07	0.10
	<i>Ocotea mandiocana</i> A. Quinet	0.07	0.03
Magnoliaceae	<i>Magnolia ovata</i> (A. St.-Hil.) Spreng.	0.20	0.00
Malpighiaceae	<i>Byrsinima niedenzuiana</i> Skottsb.	0.03	0.03
Melastomataceae	Not identified	0.00	0.03
	<i>Leandra dasytricha</i> (A. Gray) Cogn.	0.27	0.10
	<i>Miconia cabucu</i> Hoehne	0.20	0.03
	<i>Miconia cubatanensis</i> Hoehne	0.03	0.00

Family	Species	Abundance (100m <sup>2</sup> )	
		R2	R1
Meliaceae	<i>Miconia latecrenata</i> Triana	0.07	0.00
	<i>Miconia sellowiana</i> Naudin	0.03	0.00
	<i>Ossaea amygdaloïdes</i> Triana	0.00	0.03
	<i>Cabralea canjerana</i> (Vell.) Mart.	0.50	0.17
	<i>Guarea macrophylla</i> Vahl	0.50	0.47
	<i>Trichilia lepidota</i> Mart.	0.00	0.20
Monimiaceae	<i>Trichilia pallens</i> C. DC.	0.10	0.33
	<i>Mollinedia calodonta</i> Perkins	0.07	0.00
	<i>Mollinedia schottiana</i> (Spreng.) Perkins	0.77	1.30
Moraceae	<i>Brosimum glaziovii</i> Taub.	0.00	0.03
	<i>Sorocea bonplandii</i> (Baill.) W.C. Burger, Lanj. & Wess.		
	Boer	1.20	1.63
Myristicaceae	<i>Virola bicuhyba</i> (Schott ex Spreng.) Warb.	0.07	0.17
Myrtaceae	<i>Calyptranthes grandifolia</i> O. Berg	0.03	0.00
	<i>Calyptranthes lucida</i> Mart. ex DC.	0.10	0.10
	<i>Calyptranthes</i> sp.	0.00	0.03
	<i>Campomanesia guaviroba</i> (DC.) Kiaersk.	0.03	0.00
	<i>Eugenia bacopari</i> D. Legrand	0.00	0.23
	<i>Eugenia beaurepairiana</i> (Kiaersk.) D. Legrand	0.00	0.03
	<i>Eugenia handroana</i> D. Legrand	0.07	0.00
	<i>Eugenia stigmatosa</i> DC.	0.17	0.07
	<i>Eugenia verticillata</i> (Velloso) Angely	0.03	0.07
	Not identified	0.00	0.07
Nyctaginaceae	<i>Marlierea eugenoides</i> (Cambess.) D. Legrand	0.27	0.00
	<i>Marlierea excoriata</i> Mart.	0.20	0.23
	<i>Myrceugenia myrcioides</i> (Cambess.) O. Berg	0.03	0.10
	<i>Myrceugenia</i> sp.	0.03	0.00
	<i>Myrcia aethusa</i> (O. Berg) N. Silveira	0.00	0.10
	<i>Myrcia floribunda</i> Miq.	0.00	0.13
	<i>Myrcia glabra</i> (O. Berg) D. Legrand	0.00	0.07
	<i>Myrcia multiflora</i> (Lam.) DC.	0.00	0.03
	<i>Myrcia pubipetala</i> Miq.	0.37	0.27
	<i>Myrcia spectabilis</i> DC.	0.00	0.13
Ochnaceae	<i>Myrcia splendens</i> (Sw.) DC.	0.03	0.00
	<i>Myrcia tijucensis</i> Kiaersk.	0.13	0.17
	<i>Myrciaria floribunda</i> (H. West ex Willd.) O. Berg	0.10	0.07
	<i>Guapira opposita</i> (Vell.) Reitz	0.73	1.30
	<i>Pisonia ambigua</i> Heimerl	0.03	0.00
	<i>Ouratea parviflora</i> Engl.	0.57	0.67
	<i>Heisteria silvianii</i> Schwacke	0.07	0.00
	<i>Hieronyma alchorneoides</i> Allemão	0.03	0.03
	<i>Piper aduncum</i> L.	0.30	0.17
	<i>Ardisia guianensis</i> (Aubl.) Mez	0.00	0.17
Primulaceae	<i>Myrsine hermogenesii</i> (Jung-Mend. & Bernacci) M.F.Freitas & Kin.-Gouv.	0.03	0.07
	<i>Quiina glaziovii</i> Engl.	0.03	0.27

Family	Species	Abundance (100m <sup>2</sup> )	
		R2	R1
Rosaceae	<i>Prunus myrtifolia</i> (L.) Urb.	0.03	0.00
Rubiaceae	<i>Bathysa australis</i> (A. St.-Hil.) Benth. & Hook. f.	0.23	0.03
	<i>Coussarea contracta</i> (Walp.) Müll. Arg.	0.13	0.00
	<i>Faramea montevidensis</i> (Cham. & Schltl.) DC.	0.50	0.50
	Not identified	0.03	0.00
	<i>Posoqueria latifolia</i> (Rudge) Schult.	0.20	0.17
	<i>Psychotria brachypoda</i> Müll. Arg.	0.00	0.20
	<i>Psychotria leiocarpa</i> Cham. & Schltl.	0.33	0.33
	<i>Psychotria suterella</i> Müll. Arg.	1.17	1.13
	<i>Psychotria vellosiana</i> Benth.	0.37	0.00
Rutaceae	<i>Rudgea jasminoides</i> (Cham.) Müll. Arg.	0.83	2.40
	<i>Esenbeckia grandiflora</i> Mart.	0.93	0.47
	<i>Zanthoxylum rhoifolium</i> Lam.	0.00	0.03
Sabiaceae	<i>Meliosma sellowii</i> Urb.	0.03	0.03
Salicaceae	<i>Casearia silvestris</i> Sw.	0.07	0.00
Sapindaceae	<i>Matayba guianensis</i> Aubl.	0.10	0.10
Sapotaceae	<i>Chrysophyllum inornatum</i> Mart.	0.07	0.03
	<i>Chrysophyllum viride</i> Mart. & Eichler	0.07	0.00
Urticaceae	<i>Boehmeria caudata</i> (Poir.) Bonpl.	0.03	0.00

**Appendix 3.** Ecological traits of woody species sampled in the regeneration stratum at the restoration sites (A, B, C and D), considering the last evaluation (areas A, B and C: 34 months after initial intervention with tree planting; area D: 8 years after initial intervention with tree sowing) and at remnant forests (average of values recorded at the two reference areas).

Ecological traits	area A		area B		area C		area D		Remnant forests	
	Sp.	Ind.	Sp.	Ind.	Sp.	Ind.	Sp.	Ind.	Sp.	Ind.
Density/100m <sup>2</sup> (Absolute)	1.6 (12)	10.6 (80)	0.6 (5)	1.5 (11)	2 (15)	28.6 (215)	4.9 (37)	42.6 (320)	10.3 (77)	22.8 (684)
Anemochory (%)	41.7	46.3	80.0	45.5	33.3	80.0	10.8	13.1	3.7	1.7
Autochory (%)	0.0	0.0	0.0	0.0	6.7	0.5	8.1	3.1	8.6	8.8
Zoochory (%)	41.7	33.8	20.0	54.5	46.7	13.5	70.3	75.0	85.6	89.2
Non-Pioneer species (%)	0.0	0.0	0.0	0.0	0.0	0.0	18.9	6.3	83.9	92.3
Pioneer species (%)	83.3	80.0	100	100	80.0	93.5	67.6	87.8	14.1	7.4
Exotic (%)	0.0	0.0	0.0	0.0	0.0	0.0	8.1	2.8	0	0

**Appendix 4.** Species sampled in the upper stratum (dhb>5 cm) of area D, given is abundance standardized per 100 m<sup>2</sup>.

Family	Species	Abundance
Anacardiaceae	<i>Schinus terebinthifolius</i> Raddi	0.67
Annonaceae	<i>Annona neosalicifolia</i> H. Rainer	0.44
	<i>Annona rugulosa</i> (Schltdl.) H. Rainer	0.44
Cannabaceae	<i>Trema micrantha</i> (L.) Blume	3.56
Euphorbiaceae	<i>Alchornea triplinervia</i> (Spreng.) Müll. Arg.	1.33
Fabaceae	<i>Inga marginata</i> Kunth	1.11
	<i>Mimosa bimucronata</i> (DC.) Kuntze	0.44
	<i>Mimosa scabrella</i> Benth.	30.67
	<i>Schizolobium parahyba</i> (Vell.) S.F. Blake	1.11
Primulaceae	<i>Myrsine coriacea</i> (Sw.) R. Br. ex Roem. & Schult.	0.89
Rutaceae	<i>Citrus</i> sp.	0.22
Solanaceae	<i>Solanum mauritianum</i> Scop.	0.22
Urticaceae	<i>Boehmeria caudata</i> Sw.	0.44
	<i>Cecropia glaziovii</i> Snethl.	0.67
Dead	-	8.67

**Appendix 5.** Species sampled on the upper stratum of the reference areas R2 and R1, given is abundance per area (standardized to individuals per 100 m<sup>2</sup>).

Family	Species	Abundance/100m <sup>2</sup>	
		R2	R1
Annonaceae	<i>Annona neosericea</i> H. Rainer	0.20	0.30
	<i>Annona rugulosa</i> (Schltdl.) H. Rainer	0.10	0.00
	<i>Duguetia lanceolata</i> A. St.-Hil.	0.00	0.07
	<i>Guatteria australis</i> A. St.-Hil.	0.33	0.00
	<i>Xylopia brasiliensis</i> Spreng.	0.03	0.03
Apocynaceae	<i>Aspidosperma parvifolium</i> A. DC.	0.20	0.20
	<i>Aspidosperma tomentosum</i> Mart.	0.00	0.10
Aquifoliaceae	<i>Ilex paraguariensis</i> A. St.-Hil.	0.03	0.03
Araliaceae	<i>Didymopanax morototoni</i> (Aubl.) Decne. & Planch.	0.07	0.33
Arecaceae	<i>Euterpe edulis</i> Mart.	4.13	2.83
	<i>Syagrus romanzoffiana</i> (Cham.) Glassman	0.20	0.00
	<i>Piptocarpha tomentosa</i> Baker	0.00	0.03
Asteraceae	<i>Vernonanthura discolor</i> (Spreng.) H. Rob.	0.07	0.10
	<i>Jacaranda puberula</i> Cham.	0.03	0.10
Bignoniaceae	<i>Cordia silvestris</i> Fresen.	0.10	0.00
Burseraceae	<i>Protium kleinii</i> Cuatrec.	0.33	0.37
Cardiopteridaceae	<i>Citronella paniculata</i> (Mart.) R.A. Howard	0.03	0.03
Celastraceae	<i>Maytenus floribunda</i> Reissek	0.00	0.20
	<i>Maytenus glaucescens</i> Reissek	0.03	0.00
Chrysobalanaceae	<i>Hirtella hebeclada</i> Moric. ex DC.	0.07	0.03
Clethraceae	<i>Clethra scabra</i> Pers.	0.27	0.00
Clusiaceae	<i>Garcinia gardneriana</i> (Planch. & Triana) Zappi	0.13	0.23
Cyatheaceae	<i>Alsophila setosa</i> Kaulf.	4.67	0.60
	<i>Cyathea corcovadensis</i> (Raddi) Domin	0.23	0.00
	<i>Cyathea delgadii</i> Sternb.	0.27	0.00
	<i>Cyathea phalerata</i> Mart.	0.03	0.10
Elaeocarpaceae	<i>Sloanea guianensis</i> (Aubl.) Benth.	0.00	0.47
Euphorbiaceae	<i>Actinostemon concolor</i> (Spreng.) Müll. Arg.	0.40	0.30
	<i>Alchornea glandulosa</i> Poepp.	0.03	0.00
	<i>Alchornea triplinervia</i> (Spreng.) Müll. Arg.	0.03	0.17
	<i>Pachystroma longifolium</i> (Nees) I.M. Johnst.	0.03	0.00
	<i>Sapium glandulatum</i> (Vell.) Pax	0.07	0.03
	<i>Tetrorchidium rubrivenium</i> Poepp.	0.10	0.50
Fabaceae	<i>Inga sessilis</i> (Vell.) Mart.	0.07	0.17
	<i>Lonchocarpus nitidus</i> (Vogel) Benth.	0.03	0.00
	<i>Machaerium stipitatum</i> (DC.) Vogel	0.03	0.00
	<i>Piptadenia gonoacantha</i> (Mart.) J.F. Macbr.	0.03	0.00
	<i>Senna multijuga</i> (Rich.) H.S. Irwin & Barneby	0.07	0.00
Lauraceae	<i>Zollernia ilicifolia</i> (Brongn.) Vogel	0.07	0.00
	<i>Aiouea saligna</i> Meisn.	0.03	0.00
	<i>Cinnamomum glaziovii</i> (Mez) Kosterm.	0.03	0.07
	<i>Endlicheria paniculata</i> (Spreng.) J.F. Macbr.	0.03	0.17
	Not identified	0.03	0.00
	<i>Nectandra megapotamica</i> (Spreng.) Mez	0.17	0.07

Family	Species	Abundance/100m <sup>2</sup>	
		R2	R1
	<i>Nectandra membranacea</i> (Sw.) Griseb.	0.03	0.00
	<i>Nectandra oppositifolia</i> Nees & Mart.	0.30	0.50
	<i>Ocotea catharinensis</i> Mez	0.00	0.10
	<i>Ocotea elegans</i> Mez	0.03	0.00
	<i>Ocotea indecora</i> (Schott) Mez	0.03	0.00
	<i>Ocotea laxa</i> (Nees) Mez	0.03	0.03
	<i>Ocotea mandiocana</i>	0.00	0.10
	<i>Ocotea puberula</i> (Rich.) Nees	0.00	0.03
	<i>Ocotea silvestris</i> Vattimo-Gil	0.07	0.03
	<i>Ocotea urbaniana</i> Mez	0.03	0.00
	<i>Persea willdenovii</i> Kosterm.	0.10	0.00
Magnoliaceae	<i>Magnolia ovata</i> (A. St.-Hil.) Spreng.	0.03	0.07
Malpighiaceae	<i>Byrsonima ligustrifolia</i> A. Juss.	0.00	0.07
	<i>Byrsonima niedenzuiana</i> Skottsb.	0.03	0.03
Malvaceae	<i>Luehea divaricata</i> Mart.	0.03	0.00
	<i>Pseudobombax grandiflorum</i> (Cav.) A. Robyns	0.03	0.10
Melastomataceae	<i>Miconia cabucu</i> Hoehne	0.60	0.03
	<i>Miconia ligustroides</i> (DC.) Naudin	0.10	0.00
Meliaceae	<i>Cabralea canjerana</i> (Vell.) Mart.	0.27	1.07
	<i>Cedrela fissilis</i> Vell.	0.00	0.13
	<i>Guarea macrophylla</i> Vahl	0.40	0.17
	<i>Trichilia lepidota</i> Mart.	0.00	0.23
	<i>Trichilia pallens</i> C. DC.	0.07	0.03
Monimiaceae	<i>Mollinedia schottiana</i> (Spreng.) Perkins	0.10	0.13
	<i>Mollinedia triflora</i> (Spreng.) Tul.	0.17	0.00
Moraceae	<i>Brosimum glaziovii</i> Taub.	0.10	0.17
	<i>Ficus adhatodifolia</i> Schott ex Spreng.	0.00	0.07
	<i>Ficus luschnathiana</i> (Miq.) Miq.	0.07	0.00
	<i>Sorocea bonplandii</i> (Baill.) W.C. Burger, Lanj. & Wess. Boer	0.23	0.43
Myristicaceae	<i>Virola bicuhyba</i> (Schott ex Spreng.) Warb.	0.30	0.13
Myrtaceae	<i>Calyptranthes grandifolia</i> O. Berg	0.07	0.07
	<i>Calyptranthes lucida</i> Mart. ex DC.	0.00	0.13
	<i>Calyptranthes</i> sp.	0.00	0.03
	<i>Eugenia beaurepairiana</i> (Kiaersk.) D. Legrand	0.00	0.13
	<i>Eugenia handroana</i> D. Legrand	0.03	0.03
	<i>Eugenia</i> sp.	0.00	0.03
	<i>Eugenia stigmatosa</i> DC.	0.07	0.00
	<i>Eugenia verticillata</i> (Velloso) Angely	0.03	0.00
	Not identified	0.00	0.20
	<i>Marlierea eugenoides</i> (Cambess.) D. Legrand	0.10	0.00
	<i>Marlierea excoriata</i> Mart.	0.23	0.10
	<i>Myrceugenia myrcioides</i> (Cambess.) O. Berg	0.00	0.03
	<i>Myrcia aethusa</i> (O. Berg) N. Silveira	0.03	0.10
	<i>Myrcia pubipetala</i> Miq.	0.07	0.20
	<i>Myrcia spectabilis</i> DC.	0.00	0.17
	<i>Myrcia splendens</i> (Sw.) DC.	0.30	0.00

Family	Species	Abundance/100m <sup>2</sup>	
		R2	R1
	<i>Myrcia tijucensis</i> Kiaersk.	0.17	0.17
	<i>Myrciaria floribunda</i> (H. West ex Willd.) O. Berg	0.03	0.00
	<i>Myrciaria pliniooides</i> D. Legrand	0.00	0.07
Nyctaginaceae	<i>Guapira opposita</i> (Vell.) Reitz	0.47	0.90
Ochnaceae	<i>Quiina glaziovii</i> Engl.	0.07	0.17
Oleaceae	<i>Chionanthus filiformis</i> (Vell.) P.S. Green	0.03	0.00
	<i>Heisteria silvianii</i> Schwacke	0.07	0.27
Peraceae	<i>Pera glabrata</i> (Schott) Poepp. ex Baill.	0.17	0.10
Phyllanthaceae	<i>Hieronyma alchorneoides</i> Allemão	1.00	0.73
	<i>Margaritaria nobilis</i> L. f.	0.00	0.13
Primulaceae	<i>Myrsine parvula</i> (Mez) Otegui	0.17	0.00
	<i>Myrsine umbellata</i> Mart.	0.03	0.00
Rubiaceae	<i>Bathysa australis</i> (A. St.-Hil.) Hook. f. ex K. Schum.	1.77	0.63
	<i>Cordiera concolor</i> (Cham.) Kuntze	0.00	0.03
	<i>Coussarea contracta</i> (Walp.) Müll. Arg.	0.00	0.03
	<i>Faramea montevidensis</i> (Cham. & Schltl.) DC.	0.27	0.30
	<i>Posoqueria latifolia</i> (Rudge) Schult.	0.30	0.00
	<i>Psychotria suterella</i> Müll. Arg.	0.27	0.03
	<i>Psychotria vellosiana</i> Benth.	0.23	0.00
	<i>Rudgea jasminoides</i> (Cham.) Müll. Arg.	0.03	0.20
Rutaceae	<i>Esenbeckia grandiflora</i> Mart.	0.10	0.03
	<i>Zanthoxylum rhoifolium</i> Lam.	0.03	0.00
Sabiaceae	<i>Meliosma sellowii</i> Urb.	0.03	0.23
Salicaceae	<i>Casearia silvestris</i> Sw.	0.40	0.27
	<i>Xylosma pseudosalzmanii</i> Sleumer	0.03	0.00
Sapindaceae	<i>Matayba guianensis</i> Aubl.	0.43	0.27
Sapotaceae	<i>Chrysophyllum inornatum</i> Mart.	0.00	0.13
	<i>Chrysophyllum viride</i> Mart. & Eichler	0.00	0.03
Theaceae	<i>Laplacea angustifolia</i> O.C. Schmidt	0.03	0.00
Urticaceae	<i>Cecropia glaziovii</i> Snethl.	0.07	0.00
	<i>Coussapoa microcarpa</i> (Schott) Rizzini	0.00	0.03
Verbenaceae	<i>Citharexylum myrianthum</i> Cham.	0.13	0.00
Not identified	Not identified	0.03	0.00

**Appendix 6.** Species sampled in the herbaceous stratum at the areas under ecological restoration, given is mean cover per area.

Family	Species	Mean cover (%)			
		A	B	C	D
Bignoniaceae	<i>Amphilophium crucigerum</i> (L.) L.G. Lohmann	0.0	0.0	0.0	3.0
Amaranthaceae	<i>Alternanthera tenella</i> Colla	29.1	0.0	10.0	20.0
	<i>Amaranthus lividus</i> Roxb.	20.3	13.1	20.1	0.0
Apiaceae	<i>Centella asiatica</i> (L.) Urb.	10.0	1.0	1.0	45.0
Apocynaceae	Not identified	0.0	0.0	0.0	1.0
Araliaceae	<i>Hydrocotyle bonariensis</i> Lam.	0.0	5.0	0.0	0.0
	<i>Hydrocotyle ranunculoides</i> L. f.	0.0	0.0	55.3	0.0
Asteraceae	<i>Ageratum conyzoides</i> L.	0.0	3.0	20.5	3.0
	<i>Baccharis glaziovii</i> Baker	0.0	0.0	13.3	0.0
	<i>Baccharis trimera</i> (Less.) DC.	25.0	0.0	0.0	0.0
	<i>Eclipta prostrata</i> (L.) L.	2.0	0.0	0.0	0.0
	<i>Emilia coccinea</i> (Sims) G. Don	0.0	0.0	1.0	0.0
	<i>Erechtites hieraciifolius</i> (L.) Raf. ex DC.	2.0	3.0	0.0	0.0
	<i>Erechtites valerianifolius</i> (Link ex Spreng.) DC.	19.2	4.5	12.5	15.0
	<i>Gamochaeta coarctata</i> (Willd.) Kerguélen	2.0	0.0	0.0	0.0
	Not identified	0.0	0.0	11.5	0.0
	<i>Mikania campanulata</i> Gardner	0.0	3.0	30.1	0.0
	<i>Mikania micrantha</i> Kunth	0.0	0.0	18.2	9.8
	<i>Podocoma notobellidiastrum</i> (Griseb.) G.L. Nesom	0.0	0.0	1.0	1.8
Begoniaceae	<i>Senecio brasiliensis</i> (Spreng.) Less.	0.0	0.0	3.3	8.0
	<i>Solidago chilensis</i> Meyen	10.7	0.0	8.6	9.7
	<i>Sonchus oleraceus</i> L.	1.0	0.0	0.0	0.0
	<i>Vernonanthura tweedieana</i> (Baker) H. Rob.	0.0	0.0	5.0	2.5
	<i>Begonia cucullata</i> Willd.	0.0	0.0	2.5	0.0
	<i>Drymaria cordata</i> (L.) Willd. ex Schult.	0.0	3.3	22.0	36.2
	<i>Commelinaceae</i>				
	<i>Commelina diffusa</i> Burm. f.	7.8	2.0	1.0	25.2
	<i>Commelina erecta</i> L.	10.0	10.0	0.0	41.0
	<i>Tradescantia fluminensis</i> Vell.	5.0	0.0	0.0	31.7
Convolvulaceae	<i>Ipomea</i> sp.	20.0	15.0	23.5	0.0
	<i>Cucurbitaceae</i>				
	<i>Cayaponia</i> sp.	0.0	0.0	0.0	12.0
	<i>Cucurbita moschata</i> Duch.	0.0	33.2	0.0	0.0
Cyperaceae	<i>Cyperus hermaphroditus</i> (Jacq.) Standl.	0.0	3.0	3.0	1.0
	<i>Cyperus</i> sp.	6.4	1.0	9.0	5.0
	<i>Rhynchospora gigantea</i> Link	0.0	0.0	13.2	0.0
	<i>Rhynchospora holoschoenoides</i> (Rich.) Herter	1.0	4.0	2.0	0.0
	<i>Dryopteridaceae</i>				
	<i>Rumohra adiantiformis</i> (G. Forst.) Ching	0.0	0.0	1.0	0.0
Fabaceae	<i>Desmodium adscendens</i> (Sw.) DC.	0.0	0.0	0.0	4.3
	<i>Linderniaceae</i>				
	<i>Lindernia dubia</i> (L.) Pennell	0.0	0.0	1.0	0.0
Lythraceae	<i>Cuphea carthagenensis</i> (Jacq.) J.F. Macbr.	1.0	8.0	25.2	0.0
	<i>Malvaceae</i>				
	<i>Sida rhombifolia</i> L.	2.0	0.0	8.6	15.0
Melastomataceae	<i>Tibouchina clinopodifolia</i> Cogn.	0.0	0.0	1.0	0.0
	<i>Onagraceae</i>				
	<i>Ludwigia leptocarpa</i> (Nutt.) H. Hara	13.0	12.6	33.9	15.7
Oxalidaceae	<i>Oxalis conorrhiza</i> Jacq.	2.0	4.7	4.5	1.0
	<i>Phyllanthaceae</i>				
	<i>Phyllanthus corcovadensis</i> Müll. Arg.	0.0	8.0	2.0	0.0

Family	Species	Mean cover (%)			
		A	B	C	D
Phytolaccaceae	<i>Phytolacca americana</i> L.	28.3	5.7	14.4	2.0
Poaceae	<i>Axonopus</i> sp.	15.0	10.0	18.0	11.3
	<i>Bromus brachyanthera</i> Döll	0.0	15.0	0.0	0.0
	<i>Cynodon dactylon</i> (L.) Pers.	20.0	0.0	16.5	0.0
	<i>Digitaria</i> sp.	29.0	19.0	22.5	3.0
	<i>Echinochloa crus-galli</i> (L.) P. Beauv.	30.7	0.0	0.0	0.0
	<i>Echinochloa polystachya</i> (Kunth) Hitchc.	38.0	23.3	17.5	0.0
	<i>Eleusine indica</i> (L.) Gaertn.	2.0	0.0	20.0	0.0
	<i>Hymenachne donacifolia</i> (Raddi) Chase	0.0	0.0	2.0	0.0
	<i>Ichnanthus pallens</i> (Sw.) Munro ex Benth.	0.0	0.0	0.0	12.8
	<i>Lolium multiflorum</i> Lam.	0.0	2.5	1.0	0.0
	<i>Melinis minutiflora</i> P. Beauv.	0.0	0.0	0.0	46.7
	<i>Olyra humilis</i> Nees	0.0	0.0	0.0	7.6
	<i>Panicum maximum</i> Jacq.	52.0	0.0	25.0	0.0
	<i>Parodiophylochloa ovulifera</i> (Trin.) Zuloaga & Morrone	0.0	0.0	0.0	5.0
	<i>Paspalum conjugatum</i> P.J. Bergius	0.0	0.0	54.5	0.0
	<i>Paspalum dilatatum</i> Poir.	0.0	0.0	5.0	0.0
	<i>Paspalum inaequivalve</i> Raddi	0.0	6.0	0.0	3.0
	<i>Paspalum notatum</i> Alain ex Flüggé	0.0	0.0	52.1	0.0
	<i>Paspalum pumilum</i> Nees	10.0	0.0	38.3	0.0
	<i>Paspalum urvillei</i> Steud.	66.0	15.0	31.3	0.0
	<i>Paspalum vaginatum</i> Sw.	5.0	0.0	50.0	0.0
	<i>Urochloa brizantha</i> (Hochst. ex A. Rich.) R.D. Webster	56.9	0.0	5.0	0.0
	<i>Urochloa subquadripala</i> (Trin.) R.D. Webster	43.9	83.4	63.0	58.6
	<i>Uruchloa mutica</i> (Forssk.) T.Q. Nguyen	0.0	76.1	2.0	0.0
Polygonaceae	<i>Polygonum acuminatum</i> Kunth	0.0	0.0	0.0	5.0
	<i>Polygonum punctatum</i> Elliott	32.0	20.0	12.7	0.0
Portulacaceae	<i>Portulaca oleracea</i> L.	2.0	0.0	0.0	0.0
Pteridaceae	<i>Doryopteris pedata</i> (L.) Féé	0.0	0.0	2.0	1.0
	<i>Pityrogramma calomelanos</i> (L.) Link	0.0	0.0	1.5	0.0
Rubiaceae	<i>Richardia brasiliensis</i> Gomes	0.0	4.7	37.3	0.0
	<i>Spermacoce dasycephala</i> (Cham. & Schltld.) Delprete	0.0	2.0	100.0	1.0
Sapindaceae	<i>Serjania</i> sp.	0.0	0.0	0.0	15.0
Scrophulariaceae	<i>Buddleja stachyoides</i> (Benth.) E.M. Norman	0.0	0.0	17.7	11.5
Solanaceae	<i>Solanum americanum</i> Mill.	19.3	2.0	22.8	3.0
Thelypteridaceae	<i>Thelypteris dentata</i> (Forssk.) E.P. St. John	0.0	0.0	21.0	6.8
Vitaceae	<i>Cissus verticillata</i> (L.) Nicolson & C.E. Jarvis	0.0	0.0	0.0	5.0
Zingiberaceae	<i>Hedychium coronarium</i> J. Koenig	0.0	20.0	0.0	0.0
Not identified	Not identified	0.0	0.0	3.5	15.0
	Not identified	0.0	0.0	36.0	0.0
	Not identified	0.0	0.0	10.0	0.0
	Not identified	0.0	1.0	4.0	5.3

**Appendix 7.** Cover of herb categories, given are mean cover and standard deviation (SD) per area under ecological restoration.

Categories of herbs	Average percentage of cover ± SD			
	A	B	C	D
Exotic grasses	4.57±14.29	3.61±7.33	13.85±20.76	4.33±11.19
Native grasses	73.92±33.19	83.18±27.26	48.05±36.76	51.00±39.79
Forbs	26.57±32.55	11.83±19.92	46.69±32.75	44.80±35.57

**Appendix 8.** Species sampled in the herbaceous stratum at the reference areas, given is mean cover.

Family	Species	Mean of cover (%)	
		R2	R1
Anemiaceae	<i>Anemia phyllitidis</i> (L.) Sw.	3.75	0.00
Asteraceae	<i>Mikania scandens</i> Willd.	3.00	3.00
Athyriaceae	<i>Diplazium cristatum</i> (Desr.) Alston	15.25	0.00
	<i>Diplazium plantaginifolium</i> (L.) Urb.	9.00	3.00
Blechnaceae	<i>Blechnum brasiliense</i> Desv.	11.63	11.40
	<i>Salpichlaena volubilis</i> (Kaulf.) J. Sm.	37.00	0.00
Cyperaceae	<i>Pleurostachys gaudichaudii</i> Brongn.	0.00	9.00
	<i>Pleurostachys urvillei</i> Brongn.	10.91	0.00
Dennstaedtiaceae	<i>Dennstaedtia</i> sp.	9.00	0.00
Dryopteridaceae	<i>Ctenitis anniesii</i> (Rosenst.) Copel.	0.00	37.00
	<i>Didymochlaena truncatula</i> (Sw.) J. Sm.	42.00	8.40
Lomariopsidaceae	<i>Lastreopsis amplissima</i> (C. Presl) Tindale	0.00	9.00
	<i>Polybotrya osmundacea</i> Humb. & Bonpl. ex Willd.	13.33	8.47
Marantaceae	<i>Lomariopsis marginata</i> (Schrad.) Kuhn	0.00	3.00
Marattiaceae	<i>Calathea monophylla</i> Koern.	12.40	27.81
Musaceae	<i>Danaea elliptica</i> Sm.	0.00	9.00
	<i>Marattia raddiana</i> Schott	18.00	0.00
Orchidaceae	<i>Heliconia farinosa</i> Raddi	22.53	21.83
	<i>Cyclopogon polyaden</i> (Vell.) F.S.Rocha & Waechter	0.00	3.00
Poaceae	<i>Prescottia stachyodes</i> Lindl.	3.00	3.00
	<i>Ichnanthus pallens</i> Döll	3.00	0.00
Polypodiaceae	<i>Olyra glaberrima</i> Raddi	0.00	9.88
	<i>Doryopteris multipartita</i> (Fée) Sehnem	0.00	3.00
Rubiaceae	<i>Lindsaea</i> sp.	3.00	0.00
	<i>Pteris denticulata</i> Sw.	0.00	18.00
Thelypteridaceae	<i>Coccocypselum geophiloides</i> Wawra	0.00	3.00
	<i>Thelypteris dentata</i> (Forssk.) E.P. St. John	3.00	12.19

**Appendix 9.** Subset of variables selected from each set of predictor variables (based on adjusted R<sup>2</sup> value) to explain Y<sub>R</sub> (only plots with presence of regenerating woody species) or Y<sub>T</sub> (all plots described by regenerating woody species and the cover of herbs categories). All variables were significant at a level of p<0.005.

Response matrices		
Explanatory matrices	Matrix Y <sub>R</sub>	Matrix Y <sub>T</sub>
	Organic matter (OM)	Calcium (Ca)
	Calcium (Ca)	H+Al
Soil chemistry	H+Al	Phosphorus (P)
	Phosphorus (P)	Relation Ca+Mg/K
	Relation Ca+Mg/K	
	Species richness	Individuals density
Structural variables (upper stratum or planted trees)	Individuals density	Steam basal area
	Stem basal area	
	(not significant)	<i>Urochloa subquadripala</i>
Herbaceous composition		<i>Urochloa brizantha</i>
		<i>Urochloa mutica</i>
		<i>Paspalum pumilum</i>

## **CONSIDERAÇÕES FINAIS**

Neste estudo foram avaliados processos e técnicas associadas a restauração ativa de áreas severamente impactadas pela mineração de carvão, em processo inicial de restauração ecológica, implementadas pelas empresas responsáveis pelo passivo ambiental.

Evidenciou-se intensa variabilidade de dados associada às avaliações realizadas, embora processos ecológicos fundamentais ao sucesso do projeto tenham sido registrados. No capítulo 1, verificou-se que os atributos foliares, amplamente relacionados à performance de certos grupos de plantas, apresentaram poucas relações significativas. Uma das possibilidades para as baixas relações encontradas neste estudo é que os atributos se referem a valores médios de espécie em condições naturais e indivíduos, sob condições de estresse, podem apresentar ampla variação em suas respostas de crescimento. Assim, acreditamos que as mudas introduzidas estejam apresentando diferentes performances quando comparadas a indivíduos sob condições naturais.

Os resultados indicaram que as espécies com estratégias mais conservadoras de recursos apresentaram maior crescimento em copa, embora também tenham tido maior probabilidade de morrer que espécies com estratégia de crescimento rápido. Consideramos que avaliações entre atributos funcionais de plantas e taxas de crescimento e mortalidade ainda são pouco frequentes para áreas em processo de restauração e precisam ser melhor avaliadas. Com base neste estudo, apenas podemos indicar que atributos funcionais de plantas tiveram uma baixa predictabilidade em relação ao desempenho de crescimento e mortalidade das mudas nas áreas estudadas.

A presença de poleiros aumentou consideravelmente a chuva de sementes nas áreas avaliadas, especialmente de espécies não-gramíneas, assim como a proporção de espécies zoocóricas e não-pioneiras. Assim o uso de poleiros artificiais parece um método adicional importante para áreas em processo de restauração, contribuindo para quebra de barreiras associadas ao processo de dispersão.

Porém, o aumento nas taxas de recrutamento de espécies lenhosas não foi diretamente associado à presença de poleiros artificiais, mas sim à cobertura de plantas herbáceas não-gramíneas (*forbs*). Considerando ainda que houve uma relação significativa entre os padrões de distribuição das espécies da chuva de sementes sob poleiros e da regeneração natural, é provável que fatores locais estejam determinando as diferenças nas trajetórias sucessionais destas áreas. Além da potencial facilitação da cobertura de herbáceas não-gramíneas no recrutamento de lenhosas, o efeito da excessiva cobertura por gramíneas exóticas em limitar o recrutamento de espécies lenhosas foi também evidenciado no capítulo 3.

Claramente, os fatores abióticos relacionados às características do solo têm influência sobre os processos ecológicos relacionados à regeneração natural. O prejuízo à regeneração natural parece ser potencializado pela combinação das variáveis de composição química do solo e a presença de gramíneas exóticas com alta cobertura.

No entanto, este estudo também evidencia a possibilidade de retorno ao processo de sucessão natural em áreas de mata ciliar que foram profundamente alteradas por mineração de carvão, em um tempo relativamente curto. Vários resultados aqui apresentados são importantes para o desenvolvimento teórico da ecologia da restauração e para a prática da implementação das técnicas a campo. Por exemplo, as diferenças encontradas entre as áreas alertam para a necessidade de conhecer o inicio das trajetórias sucessionais, obtidas pelo monitoramento das áreas, que pode contribuir efetivamente

para projetos de restauração através da melhoria das práticas utilizadas (monitoramento adaptativo). Faz-se necessário que as técnicas de manejo e intervenção sejam avaliadas durante os primeiros anos de monitoramento e adaptadas, quando necessário, ao desenvolvimento das comunidades durante todas as fases de restauração, até que o sistema possa ser considerado auto-sustentável e resiliente.

Este estudo também enfatiza a necessidade de evitar ou controlar a invasão por espécies vegetais exóticas competitivas (como gramíneas perenes), sendo altamente recomendada a não utilização de tais espécies no processo de cobertura vegetal inicial, por comprometer a trajetória sucessional.

A variabilidade observada nos padrões estruturais e na dinâmica temporal das comunidades de plantas regenerantes demonstram as distintas possibilidades de trajetórias nos primeiros anos da restauração, mesmo em áreas cuja intervenção foi similar. Embora os padrões de trajetórias não sejam indicativos precisos de sucesso, as avaliações nas fases iniciais mostram-se fundamentais para tomadas de decisões relacionadas ao manejo no início da restauração, com importância fundamental para a continuidade dos processos sucessionais a longo prazo.

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