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Reintroduction of vascular epiphytes in forest restoration plantations

Frederico Domene

Thesis presented to obtain the degree of Doctor in Science. Program: Forest Resources. Option in: Conservation of Forest Ecosystems

Piracicaba
2018

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versão revisada de acordo com a resolução CoPGr 6018 de 2011

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RESUMO

Reintrodução de epífitas vasculares em plantios de restauração florestal

Espécies epífitas são um componente fundamental da estrutura e dinâmica das florestas tropicais. Apesar de ser frequentemente encontrada em florestas maduras, essas espécies possuem dificuldade para se restabelecer em florestas jovens (plantios de restauração floresal e florestas secundárias). Avaliações recentes de florestas jovens demonstram que houve sucesso na recuperação de carbono, principalmente pelo sucesso na sobrevivência e desenvolvimento das espécies arbóreas, que produzem uma estrutura florestal semelhante à encontrada nas florestas maduras. Entretanto a riqueza de espécies é muito inferior a original, principalmente quando comparamos as epécies epífitas. Os objetivos desta tese foram responder questões essenciais ao enriquecimento de florestas jovens, através do transplante artificial de espécies epífitas, principalmente o efeito da idade do plantio, o local de transplante, o tamnho das mudas e da espécie hospedeira utilizada na sobrevivência e desenvolvimento das espécies epífitas transplantdas. Oito espécies epífitas (Araceae, Bromélias, Cactus e Orquídeas) foram selecionadas. Nós estudamos o armazenamento de sementes, a germinação de sementes, os fatores abióticos (Água, Luz e Nutrientes) que afetam a produção de mudas e os métodos de transplante para florestas jovens, utilizando mudas e semeadura direta. Todas as epécies são ortodoxas mantendo pontencial de germinação superior a 80% após 16 meses de armazenameto. Duas espécies de cactus são fotobásticas positivas, germinando somente com presença de luz. O crescimento das mudas em casa de vegetação é fortemente influênciado pela oferta de nutrientes. Tanto o local de fixação quanto o tamanho das plantas utilizadas não interfere na sobrevivência e desenvolvimento das espécies epífitas transplantadas. A idade da floresta também não é um fator importante no sucesso das epífitas transplantadas, sendo possível utilizar florestas jovens (12-15 anos). A semeadura direta pode ser um método promissor pra enriquecer plantios jovens, porém ajustes precisam ser realizados para proporcionar maiores índices de sobrevivência das plantuas. O transplante de espécies epífitas para florestas jovens é uma estratégia viável para superar as limitações naturais de recolonização que essas florestas apresentam. Pesquisas futuras e monitoramento podem comprovar o sucesso no estabelecimento de espécies epífitas transplantadas para determinar a generalidade de nossas observações.

Palavras-chave: Restauração florestal; Enriquecimento artificial; Epifítas; Orquídea; Bromélia; Cactus; Mata Atlântica

ABSTRACT

Reintroducion of vascular epiphytes in forest restorarion plantations

Epiphytic species are a key component of the structure and dynamics of tropical forests. Although frequently found in mature forests, these species have difficulty reestablishing in young forests (forest restoration plantings and secondary forests). Recent assessments of young forests shows that there has been success in carbon recovery, mainly because of the success in survival and development of tree species, which produce a forest structure similar to that found in mature forests. However, the species richness is much lower than the original one, especially when comparing the epiphyte species. The objective of this thesis was to answer questions essential to the enrichment of young forests, through the artificial transplantation of epiphytic species, mainly the effect of planting age, the place of transplant, the size of the seedlings and the host species used in the survival and development of the species transplant epiphytes. Eight epiphytic species (Araceae, Bromeliads, Cacti and Orchids) were studied. We evaluated potendial seed storage, seed germination, abiotic factors (Water, Light and Nutrients) that affect seedling production and transplant methods for young forests using seedlings and direct seeding. All epices are orthodox maintaining germination potential higher than 80% after 16 months of storage. Two species of cactus are positive photobatic, germinating only in the presence of light. The growth of the seedlings in greenhouse is strongly influenced by the nutrient supply. Both the fixation site and the size of seedlings do not interfere in the survival and development of the transplanted epiphyte species. The age of the forest is also not an important factor in the success of the transplanted epiphytes, being possible to use young forests (12-15 years). Direct sowing may be a promising method to enrich young plantations, but adjustments need to be made to provide higher survival rates for the seedlings. The transplantation of epiphytic species into young forests is a viable strategy to overcome the natural limitations of recolonization that these forests present. Future research and monitoring of transplanted epiphytes may determine the generality of our observations.

Keywords: Forest restoration; Artificial enrichment, Epiphytes; Orchids; Bromeliad; Cactus, Atlantic Forest

1. INTRODUCTION

Epiphytes are plants that grow directly on the trunk, branches, or leaves of other plants without the emission of haustoria structures (specialized extensions for uptaking sap from the host plant). The plants that provide support are called forophytes or hosts (Zotz, 2016). These organisms can be classified according to the type of relationship they establish with the phorophyte: i) accidental epiphytes - frequently terrestrial plants that do not present specialized modifications to the environment of the canopy, being rarely observed as epiphytes; ii) facultative epiphytes - plants occupying both terrestrial and canopy environments; iii) hemiepiphytes - plants that maintain a communication, functional or not, with the soil; and iv) holoepiphytes - plants that survive without any contact with the soil of the forest or with the vascular system of the phorophyte (Zotz, 2016). There are approximately 29,000 epiphytic species, including all three types described above (except accidental epiphytes), belonging to 876 genera and 84 families. In some tropical montane forests, the representativeness of epiphytes can reach 50% of the local vascular flora, indicating the importance of this life form to the composition and biomass of these forest types (Gentry & Dodson, 1987). In Brazil, the Atlantic Forest harbours approximately 1,927 species, 201 genera, and 29 families of vascular epiphytes (Flora do Brasil, 2020).

Epiphytic species increase the structural complexity of forests by creating additional microhabitats and by adding a large biomass and surface area over tree trunks and branches, and in the forest canopy (Gentry & Dodson, 1987; Nadkarni et al., 2001), which serves as a habitat for a myriad of invertebrate species, amphibians, and other taxonomic groups that integrate the still little known canopy communities. Epiphytes contribute, therefore, to important ecological processes in tropical forests, as well as to the maintenance of a relevant part of their biodiversity. The pattern of water and nutrient cycling in forest ecosystems is influenced by the epiphytic community, either by the interception of the nutrients loaded by rainwater (Jordan et al., 1980) and their release to other plants (Lowman & Rinker, 2004); in many cases the litter produced by these organisms exceeds the concentration of nutrients than that produced by arboreal individuals (Nadkarni & Matelson, 1991). Many species of fauna present in tropical ecosystems use resources offered by epiphytic species, including fruit, floral nectar, water, small invertebrates living in epiphytes (Ellwood & Foster, 2004), and nesting.

As consequence of the aforementioned values for biodiversity conservation and ecosystem functioning, the recolonization regenerating forests or restoration plantations by epiphytes is critical for restoration success. Forest restoration projects, which have been mostly focused on the reintroduction of shrub and tree species, have not either contributed to the active reintroduction of epiphytes. Practitioners frequently expect that once the forest phisiognomy is reestablished, the other plant life forms typical of tropical forest ecosystems (e.g. epiphytes and lianas) will naturally colonize these new ecosystems by dispersing seeds. Contrary to these expectations, monitoring of second-growth forests has shown failed epiphyte recolonization, even after 100 years of regeneration (Martin et al., 2014). Unsuccessful regeneration of epiphytes could be explained by a myriad of factors associated to dispersal limitation (e.g., lack of seed sources, landscape isolation, lack of seed dispersers) and establishment limitation (e.g., lack of appropriate hosts, microclimate conditions, and herbivory). The interactions between hosts and epiphytes are not either well understood. Some researchers speculate that host characteristics, such as the texture and pH of the bark and chemical exudates released to the trunk and branches may facilitate or inhibit the colonization and development of epiphyte species. These studies have mainly developed in temperate and subtropical regions (e.g. Frei & Dodson, 1972, Benzing, 1978, Schlesinger & Marks, 1977).

Little is known about the relative importance of dispersal and establishment limitation in the regeneration dynamics of epiphytes, which prevent the use of ecological knowledge to guide the active reintroduction of epiphytes in forests undergoing restoration. The reintroduction of epiphyte species in restored forests is a new field in restoration. Some experiments involving the transplanting of epiphytes into forests have already been carried out and have found promising results (Pett-Ridge & Silver, 2002, Schneffknecht et al., 2012, Fernandez et al., 2016 Duarte & Gandolfi, 2017). However, these experimental reintroductions did not last for more than a year, so the long-term sustainability of reintroduced epiphyte populations remain unclear. Kowledge gaps include the definition of the epiphytes species with the best performance, best transplanting time, the characteristics of the hosts that influence the establishment and development of epiphyte species, and survival rates seedlings of different sizes.

Given the emerging need to reintroduce epiphytes in restoration projects and the relevant knowedge gaps associated to this activity, we aimed with this thesis to elucidate the most basic ecological and practical aspects of epiphyte reintroduction in tropical forest restoration. The thesis was organized in three core chapters, in which different technical and scientific aspects of epiphytes reintroduction were explored. In the first chapter, entitled "Methods for epiphytes species reintroduction in restored forests", we studied seed germination and storage, abiotic resources controlling seedling production, the effect of transplanting microsites and seedling size in establishment performance, and tested an innovative method for direct seeding of epiphytes. The second chapter, entitled "Successful reintroduction of orchids and bromeliads in a young restored forest", describes the reintroduction performance of four epiphytic species to a young forest restoration plantation. We evaluated how the identity and physical characteristics of host trees, especially the roughness of the bark, affects the survival and development of the transplanted epiphyte species. Finally, the third chapter, entitled "Reintroduction of vascular epiphytes in tropical forest restoration: Effects of forest age and phorophyte identity", presents the results of the reintroduction of eight epiphytic species transplanted to five restoration forests of a wide range of ages. Finally, we presented a chapter with final consideration about the major findings of the thesis and the recommendation for future works.

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2. METHODS FOR EPIPHYTES SPECIES REINTRODUCTION IN RESTORED FORESTS

Abstract

Tropical forest restoration has been essentially performed through natural regeneration and plantations of nursery-grown tree seedlings. Epiphyte species have failed, however, to recolonized both restoration plantations and second-growth forests, thus justifying the development of methods for their effective reintroduction in restored forests. Here, we developed and tested different methods for reintroducing vascular epiphytes in tropical forest restoration. We based our studies in one Araceae, two bromeliads, and three cacti epiphitic species and a 25 years old restoration plantation in theAtlantic Forest. We first investigated the requirements for seed storage and germination, and found that two cacti species had photoblastic seeds and all species had orthodox seeds with >80% germination after 16 months of storage. Secondly, we investigated the effects of the availability of water (using substrate water holding capacity as a proxy), light, and nutrient on seedling growth in nursery conditions, and found that nutrient supply was the most limiting factor. We then tested a novel approach for direct seeding of epiphytes, based on the production of an artisanal paper sheets, with seeds mixed in the pulp, that were stapled on tree trunks. Seedling emergence was high, but most seedlings were cut by herbivores. Finally, we tested the combined effect of seedling size (two classes) and transplanting site (trunk and forks) on establishment performance, but these factors did not impacted seedling survival and growth fro two years. Producing epiphyte seeds and seedlings and transplanting them to restoration proved to be viable enough to justify further investments in this activity.

Keywords: Atlantic Forest; Canopy ecology; Enrichment planting; Epiphytism; Seedling production; Seed germination; Tropical forest restoration

2.1. INTRODUCION

Forest restoration has advanced remarkably over the past decades, counting today with a large menu of methodological options to produce planting stocks and reintroduce native species on degraded lands (Rodrigues et al., 2009). The vast majority of these methods were developed for, and are applied to trees, which represent only one of the many plant life forms that make up forests, especially in the wet tropics. Trees are the dominant life form, but lianas, herbs, epiphytes and other life forms are essential elements of the diversity, structure, and functioning of forests, and thus should also be considered when assessing restoration success. The premise that these other life forms would spontaneously recolonize regenerating forests or restoration plantations has not been corroborated (Martin et al. 2014; Garcia et al., 2016; Mansourian et al., 2017), thus highlighting the need to develop specific methods for reintroducing other life forms

(Duarte & Gandolfi 2017). The recolonization of epiphytes has been especially problematic (Lisboa et al., 1991, Kanowski et al., 2003, Woods et al., 2013; Martin et al. 2014).

Epiphytes are plants that grow directly on the trunk, branches, or leaves of other plants, called as forophytes or hosts, without the emission of haustoria structures (specialized extensions for uptaking sap from the host plant; Zotz, 2016). They are essential component of tropical forests, accounting for up to a third of their plant diversity (citação) and play important contributions to nutrient cycling (Lowman & Rinker 2004), water accumulation, habitat provision, and ecological interactions (Nadkarni & Matelson 1989, Scheffers et al., 2013). The poor epiphyte recolonization of tropical restored forests may then lead to incomplete recovery of their diversity, structure, and functioning. Restoration ecology and ecological restoration have then clearly to advance in the front of other plant life forms, with special consideration for epiphytes in tropical forests.

The difficulty of epiphytes to recolonize forests can be associated to a myriad of factors linked to dispersal limitation (see Cascante-Marín et al., 2008) and establishment limitation (e.g., homogeneity of host trees, lack of microhabitats and the little microclimatic differentiation; Barthlott et al., 2001; Woods et al., 2015). Some of these constrains could be artificially overcome by reintroducing epiphytes through direct seeding or seedling transplant in forests undergoing restoration. However, little is known about the production of epiphyte seeds and seedlings, there is no method available for the direct seeding of epiphytes – which could reduce transplanting costs but is especially challenging due to the use of trunks as substrate –, nor specific technical guidelines for transplanting nursery-grown seedlings to trees.

Here, we developed and tested different methods for reintroducing vascular epiphytes in tropical forest restoration. We based our studies in one Araceae, two bromeliads, and three cacti epiphitic species and a 25 years old restoration plantation in the Atlantic Forest of Brazil. We first investigated the requirements for seed storage and germination, and the effects of the availability of water, light, and nutrient on seedling growth in nursery conditions. Once seed and seedling production was explored, we moved to transplanting methods, and tested a novel approach based on the production of an artisanal paper sheets, with seeds mixed in the pulp, that were stapled on tree trunks. Finally, we tested the combined effect of seedling size (two classes) and transplanting site (trunk and forks) on seedling establishment performance.

2.2. METHODS

2.2.1. STUDY SPECIES

We worked with two epiphytic bromeliads [*Aechmea bromeliifolia* (Rudge) Baker; *Billbergia zebrina* (Herb.) Lindl.]; three epiphytic cacti [*Epiphyllum phyllanthus* (L.) Haw.; *Lepismium cruciforme* (Vell.) Miq.; and *Rhipsalis cereuscula* Haw.], and one hemi-epiphytic Araceae species (*Philodendron bipinnatifidum* Schott ex Endl). We selected these species because i) their importance for vertebrate communities (the bromeliads are pollinated by hummingbirds and the other species by insects; all species produce fleshy fruits dispersed by birds, except the Araceae species, which is dispersed by monkeys − Rowley, 1980; Gottsberger & Jr, 1984; Scrok & Varassin, 2011; Guaraldo et al. 2013; Pansarin et al., 2016); ii) the widespread distribution of these species throughout most Brazilian states (Flora do Brasil 2020), which makes them good model species for other ecological contexts; iii) the occurrence of these native species in isolated trees and old-growth forests – but not in restoration plantations – of the study region; and iv) the availability of seeds at the early stages of the research. We collected ripo fruits from at least 10 individuals per species, in forest fragments in the region of Piracicaba-SP, southeastern Brazil. Due to limitation of seeds, some species were not used in all the experiments further described (Table 2.1).

Table 2.1. Species studied and experiments developed

	Experiments			
Species	Seed production	Seedling production	Direct seeding	Seedling transplanting
A. bromeliifolia	X	X	X	X
B. zebrina	X	X	X	X
E. phyllanthus	X	X	X	X
L. cruciforme	X		X	
P. bipinnatifidum	X	X	X	X
R. cereuscula	X		X	

2.2.2. STUDY SITE

This study was carried out in a restored forest that was planted between the years of 1988 e 1990. The planting was carried out with high diversity of native species (>120 espécies).

The restoration area has 21 hectares (ha) and is located around the public reservoir for supply of the municipality of Iracemápolis (22°34′51″ S, 47°31′08″ W), Sao Paulo, Braszil. The restored forest is surrounded by sugar cane plantation. Seasonal Semideciduous Atlantic Forest is the main vegetation present in the study area and is part of the “Interior” biogeographical zone, the second most threatened zone in the biome with only 7% forest cover remaining (Ribeiro et al. 2009). The climate is classified as Cwa (humid subtropical) with a dry winter and hot summer, according to Koppen calssification (Alvares et al., 2013). After approximately 25 years of restoration, it is still rare to find any epiphytic species naturally recolonizing trunks and branches of the trees; only a few individuals have colonized the site, mostly the bromeliad Tillandsia recurvata (L.) L. and the fern Pleopeltis sp. (nomenclature follows Reflora, 2016), which are found growing everywhere, including electrical wires and roofs.

2.2.3. Experiments

A) Seed production: Aiming at understanding the basic seed ecological behavior of epiphyte species to allow the application of the information gathered in direct seeding and seedling production in nurseries, we studied the requirements of epiphyte species for seed germination and storage. We extracted seeds by by placing them on a wire-mesh screen and rinsing abundantly in running water. Seeds were dried on shade conditions (20% seeds average water content) and then used for seed storage and germination experiments. Germination tests were performed in plastic boxes kept inside germination chambers, at constant temperature of 25°C, and exposed (light) or not to light (dark). We used four replicates of 25 seeds per species and treatment (light and dark conditions). Seed germination was evaluated every three days, based on radicle emission, until no further germination was observed. We compared treatments through a t-test. For assessing seed storage potential, we established a factorial experiment with two factors (temperature and container permeability) and two levels each (4°C and 25°C; pearmeable and impermeable container), with four replicates. Every month, and for a period of, at least 16 months, we took a sample of 20 seeds per replicate and installed germination tests.

B) Seedling production: Aiming at understanding the resources demands for producing epiphyte seedlings in nursery conditions and estimate which resources could limit the performance of reintroduced epiphyte seedlings, we established an experiment to assess the effects of water, light, and nutrient availability on seedling production. We first transplanted the seedlings obtained in the seed storage experiment to seed trays filled with an organic, forestry substrate, and were grown in there until they had a well developed root system. We then used the seedlings to establish a factorial experiment with three factors (water, light, and nutrient availability) and two levels each, with five replicates. The two light availability levels − high and low (≅ 50% and 15% of direct sunlight, respectivelly) − were obtained by covering the plots submitted to low light availability with a 50% plastic shade cloth. The two water availability levels were created by transplanting the seedlings to plastic pots of 0.415 liters filled with substrates with contrasting water holding capacity: gravel (low water retention) and organic, nutrient free substrate (high water retention). The two nutrient availability levels were created by the spraying a solution of 10 ml l^{-1} of NPK fertilizer (Polifertil® 8-8-8 + micronutrients) every 15 days in plants receiving the high nutrient availability treatment, while the low availability treatment did not receive any application and relied solely on the scarce nutrients of the substrate and irrigation water. All seedlings were grown inside a greenhouse covered with plastic, direct illumination outside the greenhouse was approximately 2000 $\boldsymbol{\mu mol\, m^{-2}\, s^{-1}}$ (n = 5 days in February 2014), and seedlings were irrigated every day with 13 liters (Irrigation three periods a day; 10 minutes per period; sprinkler flow rate: 26 liters/hour). The experiment was monitored every three months, for a period of 12 months (January - December 2014), by measuring the height, number of leaves, and length of the largest leaf of each seedling; one individual per block was randomly picked at the end of the experiment and over-dried for biomass estimation. The experiment was analyzed through an analysis of variance (ANOVA) followed by a Tukey HSD test for significant results.

C) Direct seeding: We first produced an artisanal paper mixing epiphyte seeds in the pulp, which was used for producing circular paper sheets (16 cm in diameter) containing 25 seeds of one single species (Figure S1a). We stapled four paper sheets per host tree (one sheet per species), for a total of six trees of *Myroxylon peruiferum* L.f. (Fabaceae), at 1.5 meters from the ground level and facing North. Seedling emergence (Figure S1b) and mortality were monitored for seven weeks.

C) Seedling transplanting: We tested the combined effect of seedling size (two classes) and transplanting site (trunk and forks) on seedling establishment performance, in a factorial design with 5 replicates, using various host tree species. Two seedling size classes were determined, based on the average fresh weight of seedlings of each species: *A. bromeliifolia* (big: 18.6 ± 3,8g; small: 6.1 ± 1.6g), *B. zebrina* (big: 14.8 ± 7.3g; small: 7.0 ± 3.7g), *E. phyllanthus* (big: 19.0 ± 9.7g; small: 2.6 ± 1.4g), *P. bipinnatifidum* (big: 18.8 ± 4.5g; small: 3.1 ± 1.3g). Transplanted seedlings were momitored according to length: Aracea – main stem, from base to leaf top; bromeliads – main stem, from base to leaf top; cactus – main filocladium, from base to leaf top, for a total of two years. The experiment was evaluated through a Chi-square test.

2.3. Results

The seeds of *L. cruciforme* and *R. cereuscula* required light for germination (positive photoblast), while those of *A. bromeliifolia* and *E. phyllanthus* were slight benefited by light exposition (Figure 2.1). All species had orthodox seeds with >80% germination after 16 months of storage.

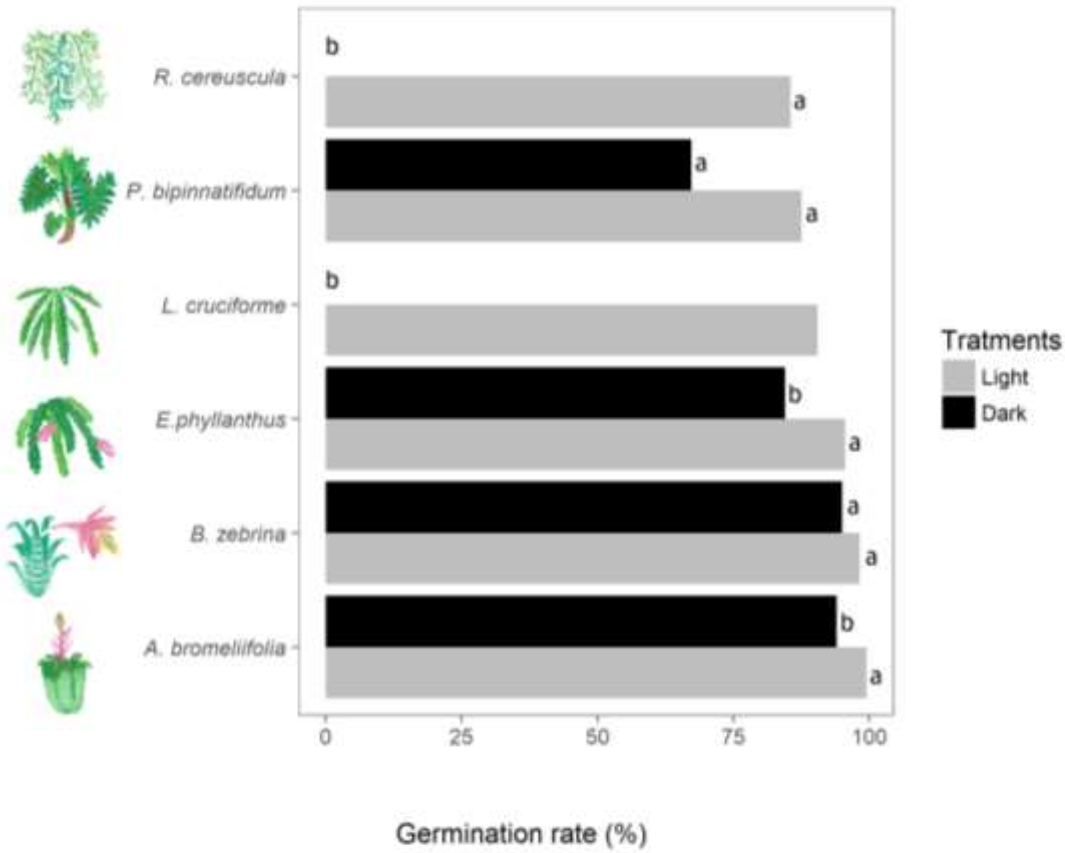

Figure 2.1: Effect of presence (Light) and absence of light (Dark) on the germination rate (%) of six epiphytic species: *Aechmea bromeliifolia* (Rudge) Baker; *Billbergia zebrina* (Herb.) Lindl.; *Epiphyllum phyllanthus* (L.) Haw.; *Lepismium cruciforme* (Vell.) Miq.; *Philodendron bipinnatifidum* Schott ex Endl e *Rhipsalis cereuscula* Haw. Different letters on the bars indicate a significant difference between the means by the *t*-test at the 5% level.

The germination rate of the seeds is significantly affected according to the methods used for storage, with storage temperature being the main factor that provokes the maintenance of the germination rate over time (Figure 2.2). Seed viability was better preserved at low storage temperature, whereas container permeability only affected the viability of *B. zebrina* and *P. bipinnatifidum* seeds, which were benefited by an impermeable container (Figure 2.2).

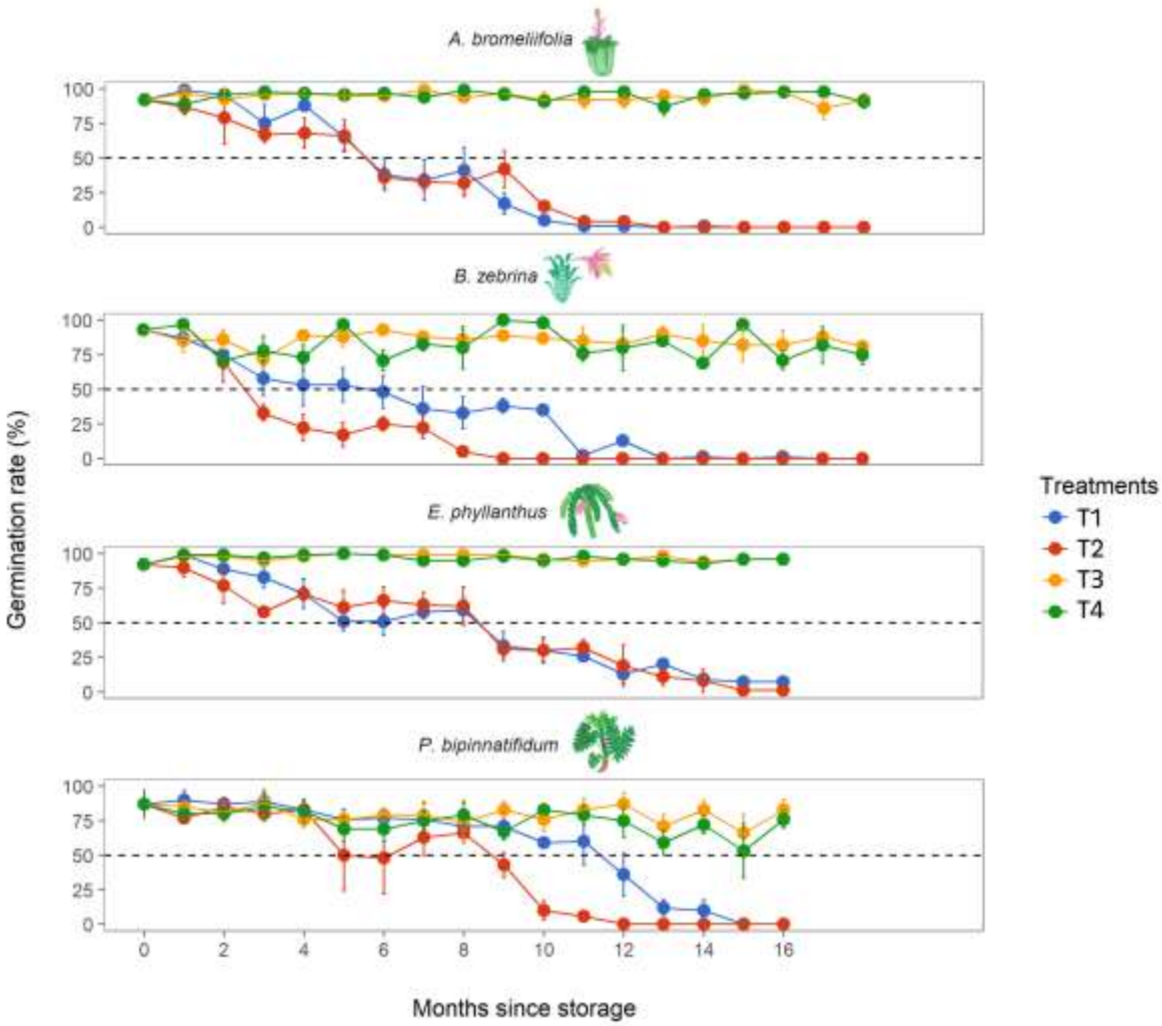

Figure 2.2: Germination rate (%) of four species: *Aechmea bromeliifolia* (Rudge) Baker; *Billbergia zebrina* (Herb.) Lindl.; *Epiphyllum phyllanthus* (L.) Haw.; *Philodendron bipinnatifidum* Schott ex Endl, after storage in two types of container and in two temperature conditions: T1 - Waterproof container /25°C; T2 - Pervious container /25°C ; T3 – Waterproof container /4°C; T4 - Pervious container/4°C.

The availability of all resources, but especially nutrients and water, affected seedling growth in nursery conditions (Figure 2.3; Figure S2-5). As expected, the maximum growth was obtained when all resources were fully available, whereas the lowest growth was obtained for full light exposition and reduced nutrient and water availability; light + water benefited most species, while *A. bromeliifolia* was especially benefited by higher light availability (Figure 2.3).

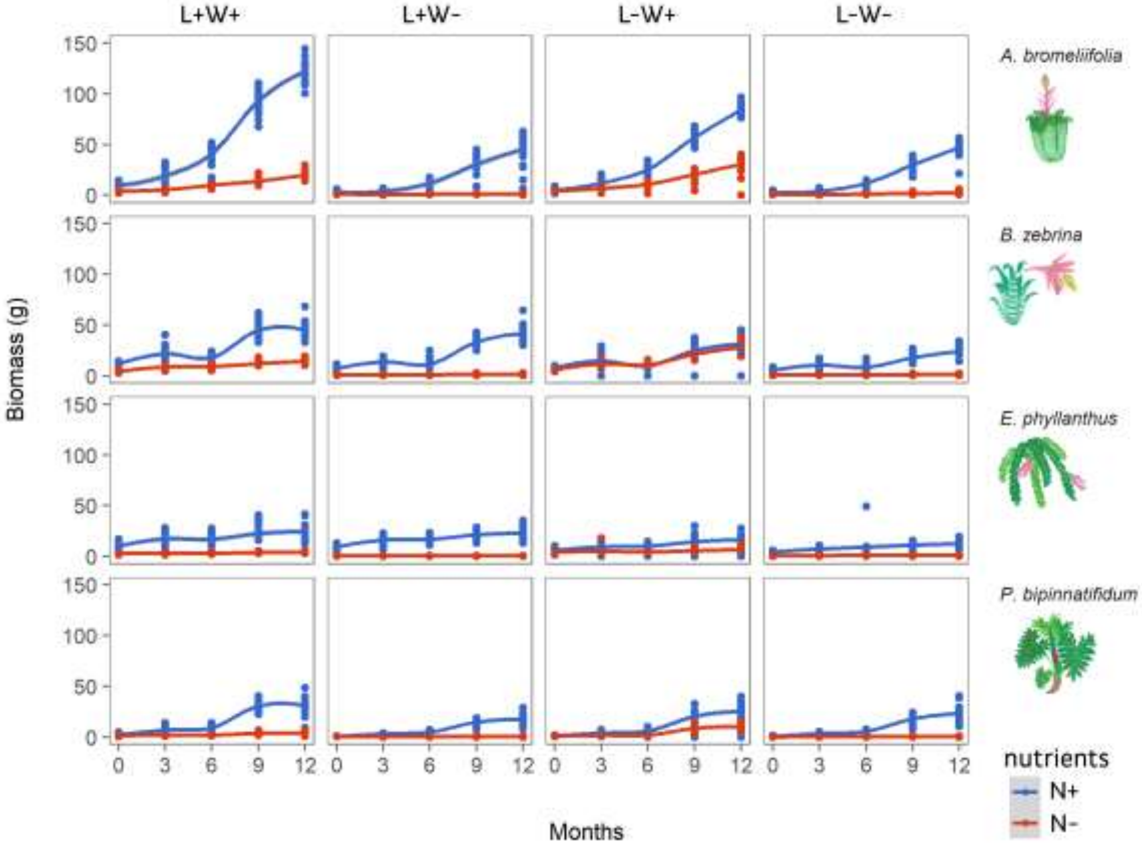

Figure 2.3: Biomass accumulation (g) for four epiphytic species: *Aechmea bromeliifolia* (Rudge) Baker; *Billbergia zebrina* (Herb.) Lindl.; *Epiphyllum phyllanthus* (L.) Haw. and *Philodendron bipinnatifidum* Schott ex Endl, cultivated in greenhouse with different light conditions (L+ ≅ 50% of direct sunlight; L- ≅ 15% of direct sunlight), water availability (W+ Substrate with water retention; W- Substrate without water retention) and nutrient supply (N+ Nutrient supply; N- Nutrient restriction). All four species were cultivated for 12 months.

Seedling emergence was fast and high, especially for *A. bromeliifolia*; *B. zebrina*; *E. phyllanthus* (Fig. 2.4), in the direct seeding experiment, but all seedlings were further consumed by leaf cutter ants. Neither the size of seedlings not the site of transplant (trunk or fork) affected seedling survival and growth (Table S2.1).

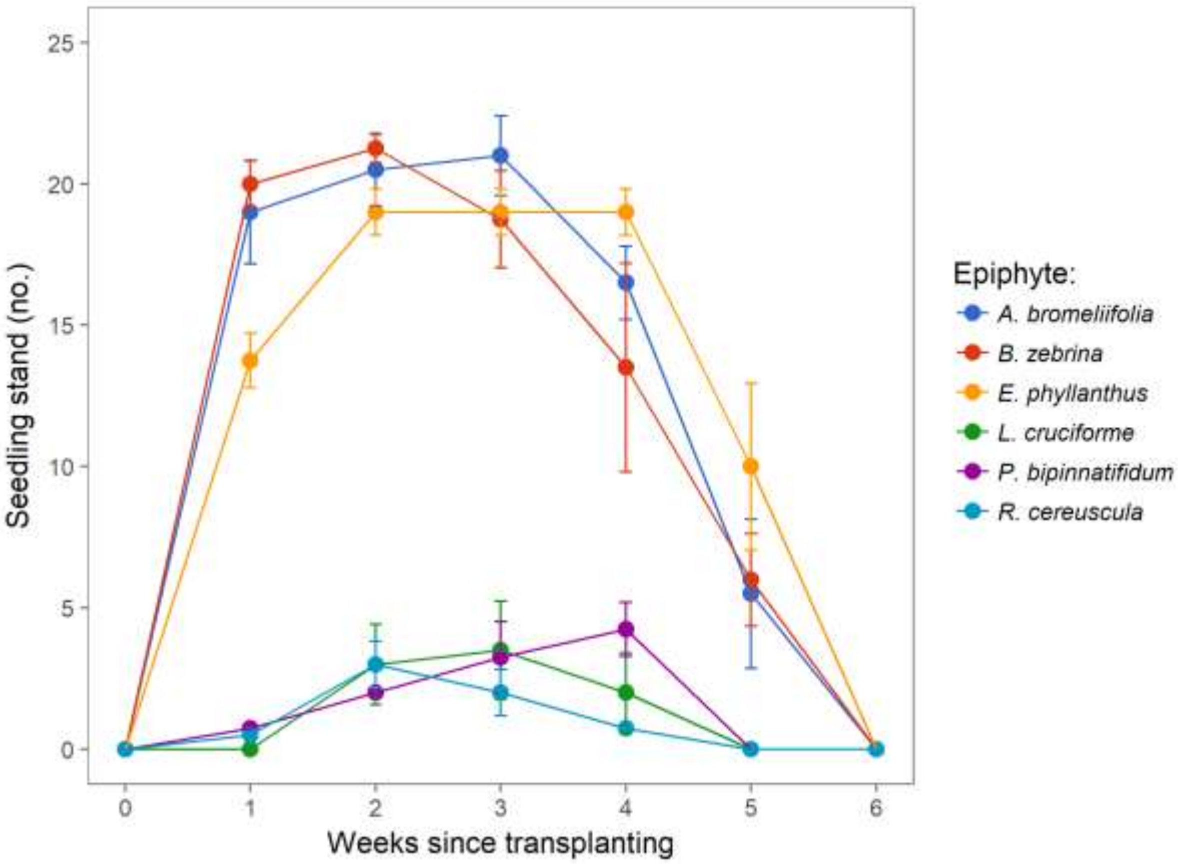

Figure 2.4: Seedling stand (no.) for six epiphytic species: *Aechmea bromeliifolia* (Rudge) Baker; *Billbergia zebrina* (Herb.) Lindl.; *Epiphyllum phyllanthus* (L.) Haw.; *Lepismium cruciforme* (Vell.) Miq.; *Philodendron bipinnatifidum* Schott ex Endl e *Rhipsalis cereuscula* Haw. transplanted to a restoration forest located in the city of Iracemápolis - SP. Transplantation was performed through a circular paper containing 25 seeds. Each paper contained seeds of only one species epiphytes.

2.4. Discussion

Our results demonstrate that the epiphytic species studied have high germination potential, they have orthodox seeds, maintaining germination potential even after long periods of storage. These characteristics allow the development of techniques for the enrichment of forest restoration plantations through direct seeding, a cheaper and faster method compared to transplanting using seedlings. The production of seedlings in a greenhouse is a simple procedure. The supply of nutrients accelerates the vegetative growth of the seedlings and reduces the time necessary for the species to enter the reproductive phase. Therefore, it is possible to obtain seedlings of epiphytic species in a shorter time, reducing production costs. In addition, because seedlings are already transplanted in an adult phase, they will more readily play the expected ecological roles, especially the production of flowers and fruits, favoring seed dispersal. The transplant site in the host tree and the size of the seedlings does not seem to be an important factor in the survival and development of the species, so it is not necessary to search for specific sites during the transplant, which facilitates the field activities.

The germination pattern of the epiphytic species reflects adaptations for canopy regeneration, since this environment is very dynamic, always undergoing changes in the light regime (e.g. deciduous, falling branches). Negative photoblastic species have a larger regeneration niche and can germinate in the innermost areas of the crown, where relative humidity and seed predation are lower (Benıítez-Rodrıíguez et al., 2004). In contrast, two species of cacti that have positive photoblastism probably germinate in the brighter parts of the crown. The positive photoblastism of the two species of cacti (*L. cruciformis* and *R. cereuscula*) may be related to the small size of the seeds (Flores et at., 2006). The longevity of epiphytic species is maintained when they are stored at low temperatures, evidencing that storage temperature is the main factor for maintaining the germination rate of the species (Dickie et al., 1990). In case that is not possible to storage seeds in low temperature, the planning of seedling production or enrichment through direct seeding is an important factor because even at environment temperature, germination after 6 months is approximately 50%

The slow growth of the epiphyte species is related to the environmental conditions in which these species develop, mainly the irregular availability of water and nutrients (Zotz 2016). Under natural conditions, nutrient uptake by epiphytes is associated with water uptake, as nutrients arrive in pulses when rainwater washes the leaves and trunks of trees. The supply of water and nutrients promoted maximum growth for the four species, when they are cultivated in greenhouses, which is consistent with other experiments involving the growth of epiphyte species (Hietz et al. 2002; Schmidt & Zotz 2002, Laube & Zotz 2003). Therefore, when producing epiphytic species with the objective of enriching restoration plantation, is important to offer nutrients, mainly to reduce the time for seedlings production and for the transplanted species are already in their adult phase.

The two species of bromeliads transplanted to two sites (Trunk and Fork) presented low survival rates due to the high rate of herbivory suffered, demonstrating that there are some environmental filters in the restoration plantations that affect the survival of these species. For the other two species, *E. phyllanthus* and *P. bipinnatifidum*, the survivorship rate is comparable to other projects that studied epiphyte transplantation (Pett-Ridge & Silver 2002, Schneffknecht et al., 2012 and Fernandez et al., 2016). Transplanting through direct seeding may be an alternative to transplanting seedlings. The germination rate is higher than other experiments that performed the same type of experiment (Winkler et al., 2005; Mondragon & Calvo-Irabien, 2006), probably due to the higher humidity provided by the seed paper method. Despite the high germination potential, the seedlings were unable to establish, mainly due to herbivory. This phenomenon may have occurred due to the implantation method used: fixation of all papers of the same specie in a single host. Thus, when finding a paper, the vector responsible for herbivory, found the entire population. To avoid herbivory and to succeed with direct seeding some possibilities must be studied, among them, the preparation of papers with seeds of several epiphytic species, to diversify the host trees that will receive the seeds seeds or even the use of some substance that repilates herbivores. The first results showed that direct seeding is an alternative to enrich forest restoration plantations, with only a few adjustments needed to make this methodology a reality

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APPENDECES

Figure S1 – Paper sheet with epiphyte specie (*E. phyllanthus*) transplanted to a host tree specie (*M. peruiferum*).

Table S2.1. Survival and growth of four epiphytic species: *Aechmea bromeliifolia* (Rudge) Baker; *Billbergia zebrina* (Herb.) Lindl.; *Epiphyllum phyllanthus* (L.) Haw.; *Philodendron bipinnatifidum* Schott ex Endl, transplanted to a forest in restoration plantation (25 years). Seedlings of two categories were used (big and small) and two attachment sites in the host tree (fork and trunk).

Epiphytes	**Seedling**	**Site**	**Survival**	**Growth**
A. bromeliifolia	Big	Fork	0.4	14.10
A. bromeliifolia	Big	Trunk	0.6	11.73
A. bromeliifolia	Small	Fork	0	-
A. bromeliifolia	Small	Trunk	0.2	-1.00
B. zebrina	Big	Fork	0.2	-19.20
B. zebrina	Big	Trunk	0.2	12.40
B. zebrina	Small	Fork	0.2	4.40
B. zebrina	Small	Trunk	0.4	-6.30
E. phyllanthus	Big	Fork	1	11.40
E. phyllanthus	Big	Trunk	1	-8.80
E. phyllanthus	Small	Fork	0.8	25.70
E. phyllanthus	Small	Trunk	1	29.00
P. bipinnatifidum	Big	Fork	0.6	32.47
P. bipinnatifidum	Big	Trunk	0.6	-0.67
P. bipinnatifidum	Small	Fork	0.6	-4.47
P. bipinnatifidum	Small	Trunk	1	12.60

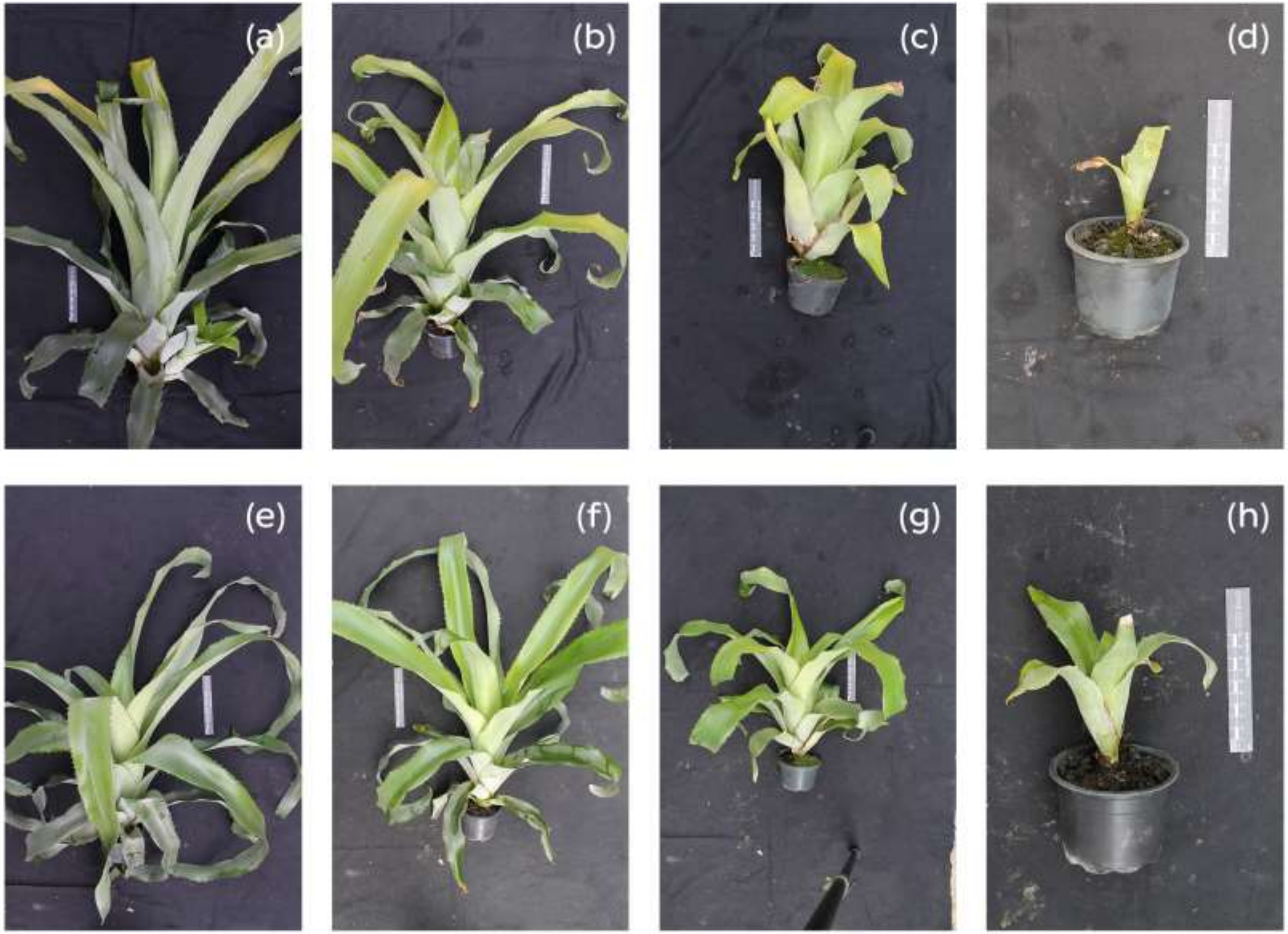

Figure S2. *Aechmea bromeliifolia* (Rudge) Baker cultivated in a greenhouse for 12 months on different lighting conditions, water availability and nutrient supply (a) L+W+N+; (b) L+W-N+; (c) L+W+N-; (d) L+W-N-; (e) L-W+N+; (f) L-W-N+; (g) L-W+N-; (h) L-W-N-

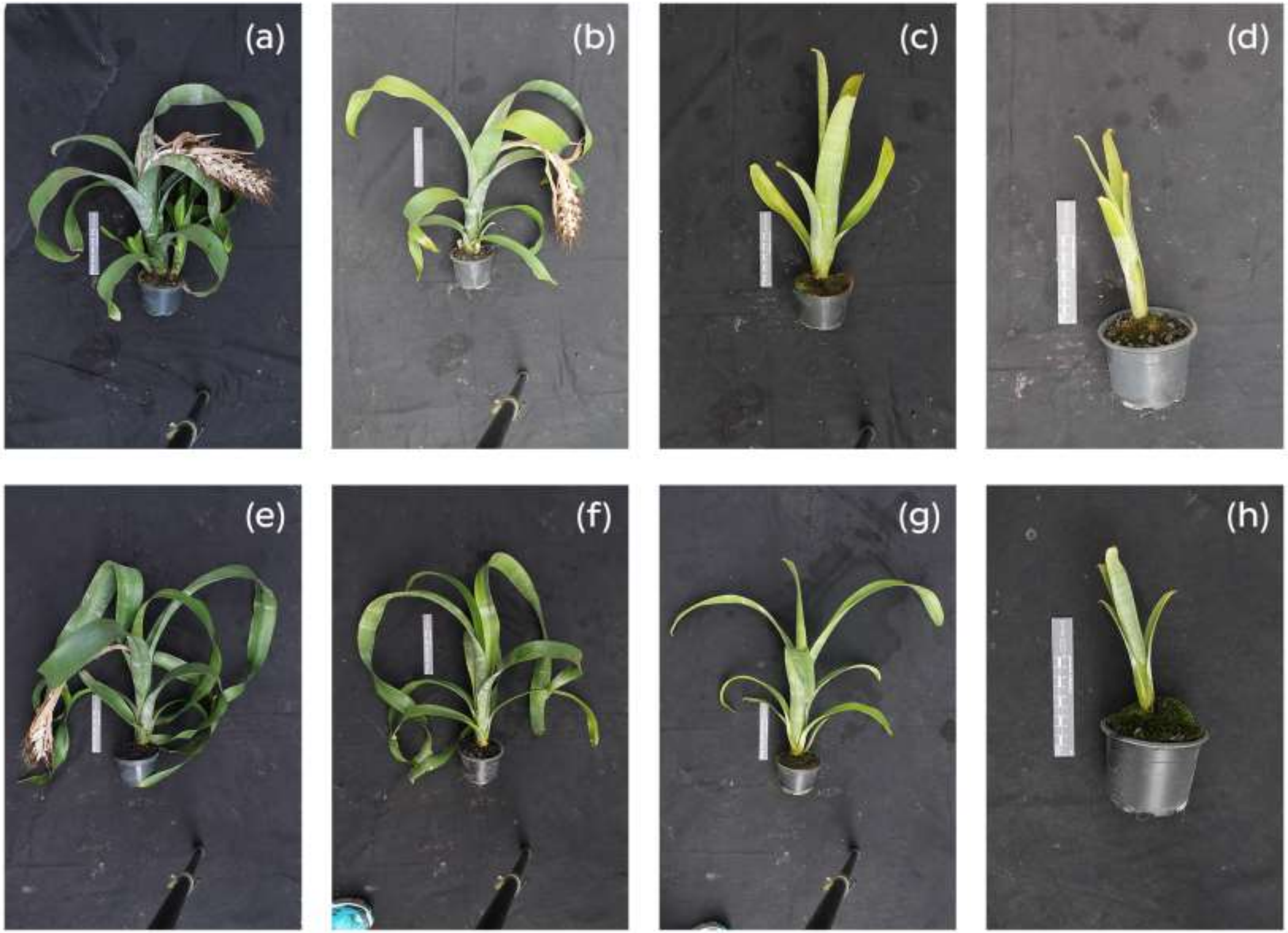

Figure S2. *Billbergia zebrina* (Herb.) Lindl. cultivated in a greenhouse for 12 months on different lighting conditions, water availability and nutrient supply: (a) L+W+N+; (b) L+W-N+; (c) L+W+N-; (d) L+W-N-; (e) L-W+N+; (f) L-W-N+; (g) L-W+N-; (h) L-W-N-

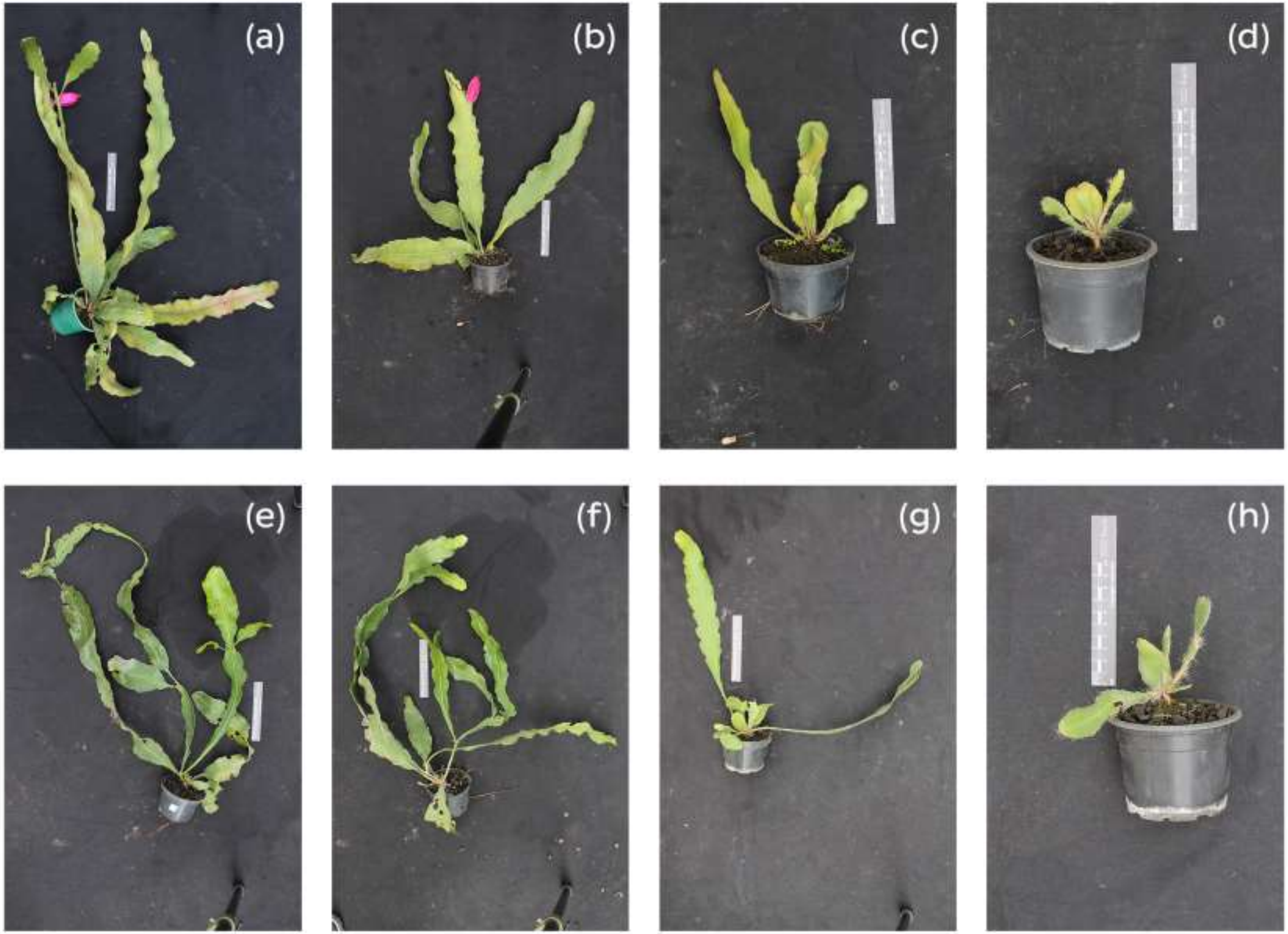

Figure S2. *Epiphyllum phyllanthus* (L.) Haw. cultivated in a greenhouse for 12 months on different lighting conditions, water availability and nutrient supply: (a) L+W+N+; (b) L+W-N+; (c) L+W+N-; (d) L+W-N-; (e) L-W+N+; (f) L-W-N+; (g) L-W+N-; (h) L-W-N-

Figure S2. *Philodendron bipinnatifidum* Schott ex Endl cultivated in a greenhouse for 12 months on different lighting conditions, water availability and nutrient supply: (a) L+W+N+; (b) L+W-N+; (c) L+W+N-; (d) L+W-N-; (e) L-W+N+; (f) L-W-N+; (g) L-W+N-; (h) L-W-N-

3. SUCCESSFUL REINTRODUCTION OF ORCHIDS AND BROMELIADS IN A YOUNG RESTORED TROPICAL FOREST

Abstract

Epiphytic plants are fundamental to the structure and dynamics of tropical forests; however, they have difficulty colonizing secondary forests and forest restoration plantations due to limitations on dispersal, germination, and establishment, thus limiting the conservation value of restored forests. Artificial reintroduction of epiphyte seedlings produced in greenhouses may be an alternative to overcome the barriers encountered in natural colonization. In this research, we monitored the survival and development of four epiphytic species transplanted into a young, Seasonal Semideciduous Atlantic Forest restoration plantation in southeastern Brazil. We transplanted two orchids (*Cattleya forbesii* Lindl. and *Oncidium flexuosum* Sims) and two bromeliads (*Aechmea bromeliifolia* (Rudge) Baker and *Billbergia distachya* (Vell.) Mez) on 90 host trees in a six year old restoration plantation. We selected six host tree species, which varied in bark roughness. Survival, growth and production of flowers and fruits were monitored for approximately six years. Survival varied among transplanted epiphyte species: orchids (*C. forbesii*: 63.3%; and *O. flexuosum*: 10.0%); bromeliads (*A. bromeliifolia*: 4.4%; *B. distachya*: 43.3%). Host tree identity strongly influenced survival for all species, as well growth, flowering, and fruiting for *B. distachya* and *C. forbesii*. Survival, flowering and fruiting of *B. distachya* and *C. forbesii*, and also growth (length) for *C. forbesii*, were favored by bark roughness. Three out of four epiphyte species produced flowers and two produced fruits, suggesting that even young forests provide suitable habitat for growth and reproduction, though not necessarily for seed germination and establishment of young seedlings. Future research and monitoring will evaluate epiphyte germination in this and additional sites to determine the generality of our observations.

Keywords: Epiphytes; Artificial reintroduction; Enrichment planting; Dispersal limitation; Forest restoration; Atlantic forest

3.1. Introduction

Epiphytic species account for more than half of plant diversity in some tropical ecosystems (Gentry & Dodson, 1987). They increase the structural complexity of forests adding biomass and surface area to forest canopies (Gentry & Dodson, 1987 and Nadkarni et al., 2001), and by creating unique microhabitats (Scheffknecht et al., 2012) which serves as a habitat for insect populations (Fernandes et al., 2016, Ellwood & Foster, 2004) and other taxonomic groups. Epiphytes affect water and nutrient cycling in forest ecosystems (Gotsch et al., 2016, Lowman & Rinker 2004), provide food for wildlife (Nadkarni & Matelson 1989 and Cruz-Angón & Greenberg 2005), and are important for biological interactions, preventing co-exinctions (Guaraldo et. al., 2013).

Forest ecosystem degradation drastically compromises the epiphytic community. Even regenerating forests more than 100 years old do not always recover the reference-level epiphyte biodiversity (Martin et al., 2014), possibly due to the limitations of dispersion (Mondragon & Calvo-Irabien, 2006), specific conditions for germination (Goode & Allen, 2009), and microclimatic conditions (Toledo-Aceves & Wolf, 2008). Furthermore, recolonization by epiphyte species is limited both by the absence of propagules in secondary forests and by the lack of surrounding forest cover (Vellend, 2003 and Reid et al., 2016). These facts underscore the importance of helping the recolonization of forest ecosystems by epiphyte species.

Host specificity is an important parameter for the ecology of epiphyte species. Structural host specificity measures the performance (e.g. occupancy, abundance and fitness parameters) of any epiphyte species on a given host tree species relative to other host tree species (Wagner et al. 2015). Determining the degree of host specificity is important in a conservation context because specialist species are generally more vulnerable to habitat alterations and climate change (Clavel et al., 2011) and threatened by coextinction with their hosts (Tremblay et al., 1998; Colwell et al., 2012). Studies that investigated the relationship between epiphytes and hosts were carried out in natural forests, in which several uncontrolled variation factors (e.g. preferential dispersal) can influence the results and mask the effects of host specificity (Wagner et al., 2015). Therefore, restoration forests offer an opportunity to study the interactions between epiphytic and host species, since abiotic factors can be controlled.

Restoration has already been considered as the "acid test of ecology," since its success checks our understanding of ecosystems (Bradshaw 1987) and may be a way to more closely explore how epiphytes can be restored in these new ecosystems. Since it is difficult to reestablish epiphyte biodiversity levels in restored forests and given the ecological importance of these species to the functioning and structure of these ecosystems, it is necessary to understand how to overcome the natural limitations that prevent the reestablishment of epiphyte species, promoting the conservation of these species in the restored forests. Three main questions guided the study: Is variance in the survival, growth and reproduction of transplanted epiphytes explained by host tree species? Do host tree preferences vary depending on epiphyte species identity? Do young restoration forests provide appropriate conditions for the establishment and development of epiphyte species?

3.2. Methods

3.2.1. Study Area and Epiphyte Species

This study was carried out in a restored forest that was planted in March 2004, using 20 different regional Brazilian native tree species (Campoe et al., 2010). The restoration area has 4.5 hectares (ha) and is located at the Anhembi Forest Research Station (AFRS), owned by the University of São Paulo (22°58′04″ S, 48°43′40″ W), situated at an elevation of 460 m. The restored forest is surrounded by eucalyptus plantation and by water reservoir. The AFRS has an area of 663.49 ha and land use is predominantly occupied with forest plantations of eucalyptus and pine trees, for research purposes. The land use in the landscape in which the AFRS is inserted is characterized by degraded pastures with small fragments of secondary forest. Seasonal Semideciduous Atlantic Forest is the main vegetation present in the study area and is part of the "Interior" biogeographical zone, the second most threatened zone in the biome with only 7% forest cover remaining (Ribeiro et al. 2009).

The climate is classified as Cwa (humid subtropical) with a dry winter and hot summer, according to Koppen calssification (Alvares et al., 2013). Over the 69 months of the study, mean, maximum, and minimum monthly temperature averaged 22.7°C, 33.5°C, 11.9°C and rainfall averaged 1248 mm year-1, with 68% falling between October and March. There was a strong drought in 2014 – the most severe over the last 52 years – in which precipitation declined 40% compared to historical records (CEPAGRI, 2017, Marengo et al. 2015). After 12 years of restoration, it is still rare to find any epiphytic species naturally recolonizing trunks and branches of the trees; only a few individuals have colonized the site, mostly the bromeliad *Tillandsia* recurvata (L.) L. and the fern *Pleopeltis* sp. (nomenclature follows Reflora, 2016), which are found growing everywhere, including electrical wires and roofs. There are more than 350 species of vascular epiphytes in regional, old-growth forest (Reflora, 2016). Epiphyte species constitute 13% of plant diversity in Semideciduous Seasonal Atlantic forest, and this rate may increase with the description of new species (Reflora, 2016).

We worked with four epiphyte species: *Aechmea bromeliifolia* (Rudge) Baker (Bromeliaceae), *Billbergia distachya* (Vell.) Mez (Bromeliaceae), *Cattleya forbesii* Lindl. (Orchidaceae) and *Oncidium flexuosum* Sims (Orchidaceae). The species selection was based on two criteria: Frist, the availability for seed collection (Bromeliads) and for seedlings acquisition in commercial orchids, as the propagation of this species is a sensitive and time-consuming process. Second, the natural occurrence of the four species in forest fragments of the study region. Orchids are critical partners in some coevolved mutualisms (Nilsson, 1992 and Ramírez, 2009). They are also the most diverse epiphyte family (Benzing, 1990), and many are threatened with extinction (Leão et al., 2014). Bromeliads are keystone species that provide microrefugia and specialized habitats, mainly in a seasonal forest (Brandt et al., 2016). These bromeliads also produce fleshy fruits during the dry season when trees are not fruting (pers. oberverv.)

3.2.2. Nursery

Orchids were purchased from a commercial laboratory (Biorchids Comercial de Plantas Ltda; Várzea Grande, São Paulo, Brasil.) as seedlings, and bromeliad seedlings were produced through seeds collected from at least 10 wild individuals per species in Seasonal Semideciduous Atlantic Forest fragments near the city of Piracicaba (São Paulo state). Bromeliad seeds were removed from fruits and germinated in germination chambers with constant photoperiod (12 hours of light and 12 hours of darkness). Orchid and bromeliad seedlings were transplanted into a sphagnum substrate and cultivated for six months in a greenhouse. Neither orchids nor bromeliads were fertilized. Mean length (cm) at transplantation to the restored forest was 4.8 (*A. bromellifolia*), 9.0 (*B. distachya*), 4.3 (*C. forbesii*), and 3.7 (*O. flexuosum*) (Figure 3.1).

Figure 3.1: Comparison of the development of four epiphytic species: *Cattleya forbesii* Lindl. *Oncidium flexuosum* Sims.; *Aechmea bromeliifolia* (Rudge) Baker and *Billbergia distachya* (Vell.) Mez, transplanted to a young restored tropical forest. In the upper line the photos are referring to the transplant period. The bottom line shows the species in the last evaluation. Orchids were monitored for 2089 days, bromeliads for 1717 days. The length and shoot values are mean ± standard deviation of the difference between the individual size at beginning of the experiment and the size at the last evaluation.

3.2.3. Transplantation Methods

We transplanted epiphytes onto 90 host trees in three blocks of young, restored forest. In each block, we selected six host tree species (five individuals per species per block), which varied in bark roughness (Lorenzi, 2000; table S3.2). Planted trees were arranged in the same configuration in each experimental block. Experimental blocks were separated by <60 meters and were relatively homogeneous. The host tree species were chosen because they are frequent in secondary forests and in forest restoration plantations of the Atlantic Forest. The host individuals were chosen randomly, inside the restored forest. Trees <6 m from the edge of the experimental blocks were excluded from the study.

Host tree species included: *Acacia polyphylla* (DC.), *Erythrina mulungu* (Mart. ex Benth.), *Hymenaea courbaril* (Hayne ex Lee et Lang.), *Jacaranda cuspidifolia* (Marth.), *Tabebuia impertiginosa* (Marth ex Standl.) and *Peltophorum dubim* (Spreng. ex Taub.). On each host tree, we transplanted four individual epiphytes, one of each species. Epiphytes were affixed at two meters height, facing north, using palm fibers and natural fiber rope (Sisal). Orchids were transplanted in December 2010; bromeliads were transplanted in December 2011 (Figure 3.1).

3.2.4. Monitoring

Epiphyte survival and growth were monitored each three months for the first year, each six months for the second year, and annually for subsequent years through September 2016. Growth and reproductive parameters measured included length (cm), number of stems, flowering and fruiting. For orchids, growth was assessed by measuring the length of the largest stem and sum of the number of stems (Zotz, 1995). The two species grow sympodially, usually developing one stem per year, which are sourced from a main rhizome. However, in some individuals the branching of the rhizome occurs, giving rise to two or more growth points. For bromeliads, the grow was assessed by measuring the stem from the base of the root to the tip of the longest leaf. When the individuals of both species reproduced by the emission of a ner lateral stem, this growth was accounted for by the sum of the stems.

Flowring and fruting was measured by the simple presence of flower and fruits, not taking into account the quantity of flowers or fruits by individuals. The results were expressed as percentage of plants that had flowers or fruits.

3.2.5. Data Analysis

We performed survival analysis to assess differences in time until an event (death) occurred in each treatment per species. We used a Kaplan–Meier estimator of survival probability (Therneau, 2012), that is defined as the probability of surviving in a given length of time while considering time in many small intervals. This method calculates the probability of recall of an event (death) in a specific period of time (interval between evaluations). Successive probabilities of survival are multiplied to reach the final estimate (Altman, 1992). The log-rank test was used to test whether the difference between survival times between the treatments. The comparison is performed by testing the null hypothesis (no difference in survival between treatments).

Two-way analyses of variance (ANOVA) was used to compare independent effects and interactions between block, host tree effect, and bark roughness on growth of the four epiphyte species studied. Statistical analysis were performed with R version 3.2.5 (R Core Team, 2014), using the “OIsurv”, “lme4” and “lmerTest” packages.

3.3. Results

After 69 months epiphyte survival varied strongly by species: *A. bromeliifolia* (4.4%); *B. dystachya* (43.3%); *C. forbesii* (63.3%) and *O. flexuosum* (10.0%) (Figure 3.2). Among surviving individuals, mean length growth by species ranged from 3.8-14.8 cm and stem count ranged from 1-9 (Table S3.1). Three species (*B. distachya*, *C. forbesii and O. flexuosum*) produced flowers, and two of these (*B. distachya*, *C. forbesii*) produced fruits.

The identity of host tree species significantly influenced survival of the four epiphytic species ($P \leq 0.03$), and host preferences were strongly species-specific for *B. distachya* and *C. forbesii* with greatest survivorship (Figure 3.3). Host tree identity also influenced growth, flowering, and fruiting for B. distachya and C. forbesii but not for *A. bromeliifolia* or *O. flexuosum*. Survival, flowering and fruiting of *B. distachya* and *C. forbesii*, and also growth (length) for *C. forbesii*, were favored by bark roughness ($P \leq 0.05$).

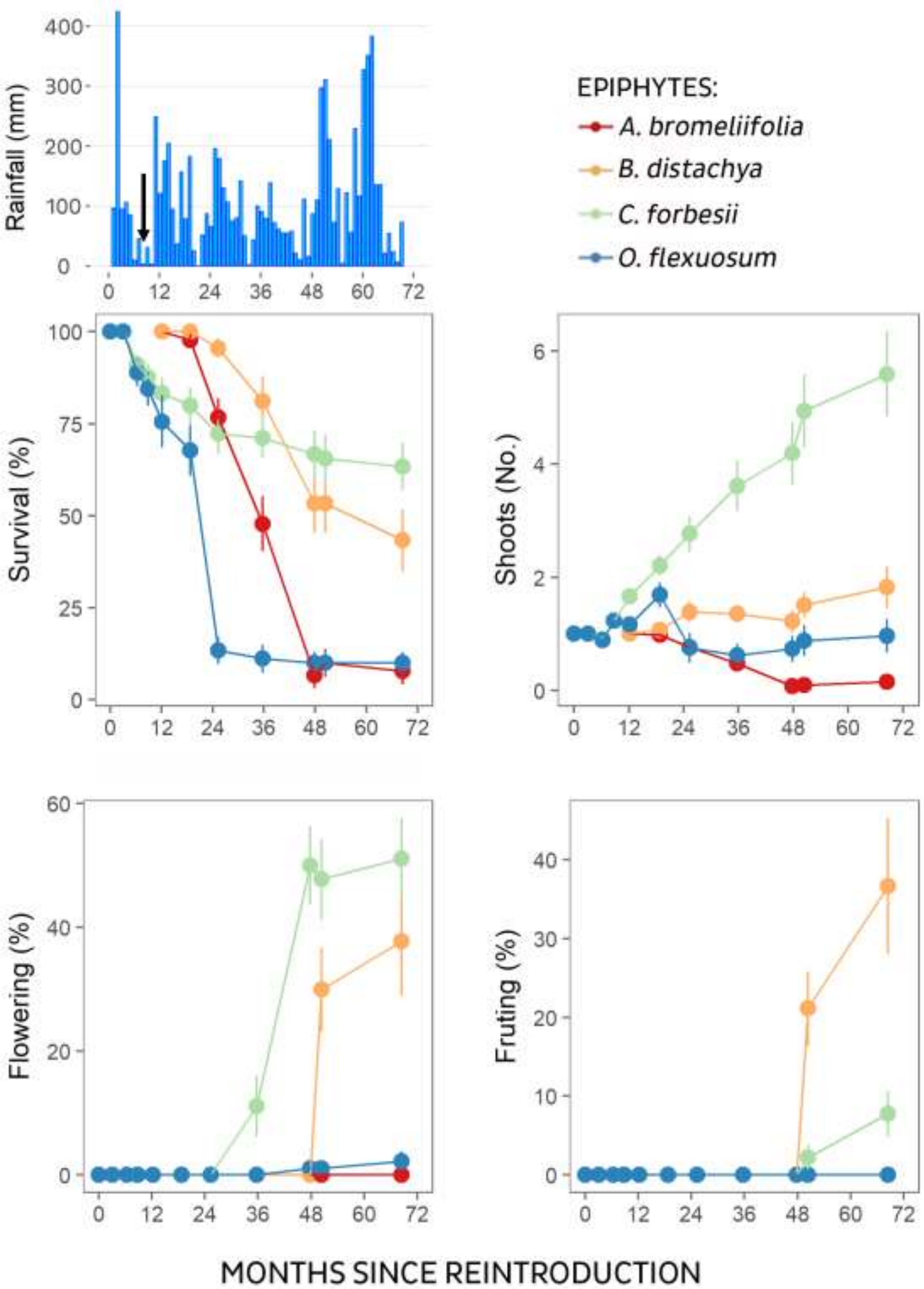

Figure 3.2 – Survival, growth, flowering and fruiting of four epiphytic species transplanted into a young Seasonal Semideciduous Atlantic Forest restoration plantation, in relation to rainfall (top row), 2010 – 2016. ACH - *Aechmea bromeliifolia* (Rudge) Baker (Bromeliaceae); BIL – *Billbergia distachya* (Vell.) Mez (Bromeliaceae); CAT – *Cattleya forbesii* Lindl. (Orchidaceae); ONC – *Oncidium flexuosum* Sims (Orchidaceae). Bars represent the standard error.

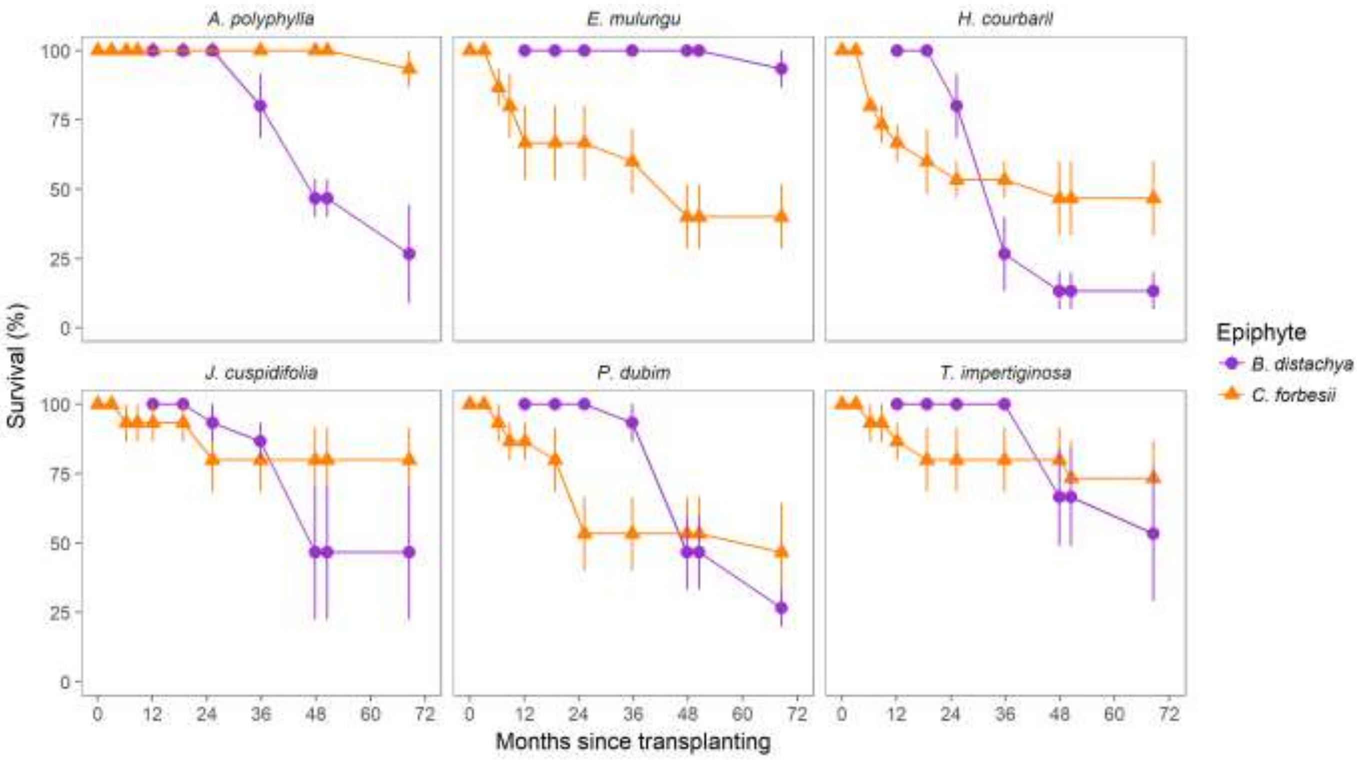

Figure 3.3: Differential epiphyte survival among host trees when transplanted to a young Seasonal Semideciduous Atlantic Forest restoration plantation, 2010-2016.

3.4. Discussion

Our long-term results demonstrate that it is possible for epiphytic species transplanted into a young, Seasonal Semideciduous Atlantic Forest to survive, flower, and fruit. Two species (an orchid and a bromeliad) were severely affected by the dry season in the first years after transplantation, showing that climatic conditions in the first years after transplantation are an important ecological filter for the survival of epiphytic species in this ecosystem (Fig. 3.2). Nevertheless, transplantation assisted the other species to overcome dispersal and early establishment limitations. Few previous studies have evaluated epiphyte reintroduction success, and even fewer have monitored transplanted individuals for five years (Wendelberger & Maschinski, 2016), though at least one study suggests that it may take even longer to evaluate the success of these reintroductions (Duquesnel et al., 2017). Bromeliad survival after one year in our study (*A. bromeliifolia* – 76.7%; *B. distachya* – 95.6%) was comparable to early survivorship in Puerto Rico (Pett-Ridge & Silver 2002; 97% survival after 6 months), Mexico (Schneffknecht et al., 2012; 70% survival after 7 months), and Costa Rica (Fernandez et al., 2016; 65-95% survival after 9 months, Duarte 2017).

We also identified clear host preferences (Figure. 3.3), which differed between an orchid and a bromeliad. This finding supports the use of a high diversity of native tree species in restoration plantations to provide enough habitat variability for the further recolonization of a relevant share of the regional epiphyte species pool. Existing literature finds mixed support for host preferences among vascular epiphytes (Calloway et al., 2002). Bark roughness could help explain the differential preferences of C. forbesii, which uses velamen to collect water and nutrients directly from the atmosphere (Benzing, 1990). However, more research is needed on epiphyte host preferences to improve the choice of the best host trees for new epiphytic transplants.

Biology of the species may explain the survival difference between the epiphyte species. Amog the orchids, *C. forbesii* has thicker roots and leaves compared to *O. flexuosum*, which may reflect water retention and the ability to absorb water. For bromeliads, *B. distachya* individuals showed red leaves (Figure 3.1), a reaction to environments with high exposure to sunlight (Pittendrigh 1948, Barthlott 2001). This phenomenon was not observed for *A. bromeliifolia*. The ability to adapt to environments with high luminosity and the thickness of leaves and roots may explain the survival difference between the epiphyte species. There are no records in the literature on the behavior of the species studied in natural forests, therefore, we recommend that further research be done with these four species to better understand what factors affect the survival and development of these species.

Although some orchid and bromeliad individuals started producing flowers and fruits, it is not possible to say that the transplanted epiphytes will become self-sustaining over time, giving rise to new individuals, as there may be strong limitations to germination and establishment in young tropical forests (e.g. water availability, light quality, presence of symbiotic fungi) (Castro-hernández et al. 1999, Mondragon & Calvo-Irabien, 2006). Therefore, future monitoring will be required to describe the dynamics of the population increase in this forest (Duquesnel et al., 2017). However, the transplanted epiphytes could contribute to the population at the landscape level, because the seeds produced by mature individuals could colonize older forests and trees outside the forest undergoing restoration.

The reintroduction of epiphytic species into tropical forests is a challenging new field of ecological restoration and can represent a valuable opportunity for controlled ecological studies to investigate the interaction of epiphytes and host trees. Our results demonstrate that it is possible to be successful with this activity, but more research still needs to be done so that this action offers concrete results. The next experiments should evaluate direct seeding to evaluating germination and early establishment limitations; the epiphytes reintroduction seedlings with different sizes, identifying the best size to perform the transplant and whether introducing epiphyte to forests of different ages makes a difference.

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APPENDECES

Table S3.1. Epiphyte survival, growth, flowering, and fruiting. All individuals used for analysis of survival, flowering, and fruiting. Only surviving individuals were used to calculate growth. Orchids were monitored for 2089 days, bromeliads for 1717 days.

Epiphyte species	**Survival (%)**	**Growth (no. shoots)**	**Growth (length cm ± sd)**	**Flowering (%)**	**Fruiting (%)**
Cattleya forbesii	63.3	7.8 ± 3.3	11.6 ± 6.1	55.6	8.9
Oncidium flexuosum	10.0	8.7 ± 4.9	14.8 ± 11.8	2.2	0.0
Aechmea bromeliifolia	4.4	1.0 ± 0.0	3.8 ± 9.9	0.0	0.0
Billbergia distachya	43.3	3.2 ± 1.7	14.8 ± 6.7	42.2	38.9

T**able S3.2.** Bark texture e DBH (cm) of host tree species. The bark texture classification was obtained through literature review

Family	**Host species**	**Bark texture[1]**	**DBH (cm)**
Fabaceae	*Acacia polyphylla* (DC.) 1	Harsh	13.25 ±5.14
Fabaceae	*Erythrina mulungu* (Mart. ex Benth.)1	Rough	19.24±8.63
Fabaceae	*Hymenaea courbaril* (Hayne ex Lee et Lang.)1	Smooth	10.89±3.55
Bignoniaceae	*Jacaranda cuspidifolia* (Marth.)1	Rough	9.21±4.59
Fabaceae	*Peltophorum dubim* (Spreng. ex Taub.)1	Smooth	9.77±4.60
Bignoniaceae	*Tabebuia impertiginosa (Marth ex Standl.).*	Rough	9.64±4.59

[1] Lorenzi, H. 2002. Árvores brasileiras. Manual de identificação e cultivo de plantas arbóreas nativas do brasil. Instituto Plantarum, Nova Odessa, BR

4. RESTORING VASCULAR EPIPHYTES COMMUNITIES IN TROPICAL REFORESTATION PLANTATIONS: EFFECTS OF FOREST AGE AND PHOROPHYTE IDENTITY

Abstract

Epiphyte species have important structural and ecological functions in forest ecosystems, although they are neglected in forest restoration projects, due to the prioritization of shrub and tree species planting. Even after several years of planting, it is rare to observe epiphyte species colonizing restored forests. With this project we try to answer key questions for the reintroduction of epiphytic species in forest restoration plantations, mainly: (i) Does the age of forest restoration planting affect the survival and development of transplanted epiphyte species? (ii) Is the survival and development of the transplanted epiphyte species influenced by the roughness of the bark of host trees? We transplanted 780 seedlings of eight epiphytic species – one Araceae; two bromeliads; three cacti and two orchids – for five forest restoration plantations with ages varying from six to 57 years. For each forest restoration plantation, we transplanted the eight epiphytic species to 20 host trees (five individuals per host tree species). We monitored the survival (%) and growth for 47 months. Survival ranged from (0 to 80%) while the length (cm) of the species ranged from (13.4 - 59.3). Both the age of forest restoration planting and the roughness of the host tree bark did not affect the survival and development of the eight transplanted epiphyte species. The enrichment of restoration plantations through the artificial transplantation of epiphytic species is a viable strategy to overcome the natural limitations of recolonization suffered by epiphyte species. Monitoring of established epiphytic communities will be necessary to assess whether, in addition to surviving, transplanted species will be able to perform the expected ecological functions.

Keywords: Artificial reintroduction; Adaptative management, Restoration projetcts; Restored Forests; Epiphytes

4.1. INTRODUCION

Tropical forests are ecosystems of great importance for the conservation of biodiversity (Gardner et al. 2009), standing out for the generation of multiple ecosystem services that guarantee the survival of millions of people worldwide (Molnar et al. 2004), and the supply of large quantities of timber and non-timber forest products (Simões & Lino, 2003). The Atlantic Forest is an example of this type of forest. In addition to providing ecosystem services that support more than 100 million people, it has a high biodiversity level, compared to other world ecosystems, with approximately 20,000 species of plants. making it a priority ecosystem for the conservation of global biodiversity (Myers et al., 2000, Laurence, 2009). However, its ecological and social importance was not enough to spare this ecosystem of degradation. The Atlantic Forest has been destroyed for several centuries due to real estate speculation and the various cycles and economic pulses that existed in its domain, resulting in intense habitat loss (Dean, 1996).

Today, only 12% of the vegetation present in this biome remains, which are arranged in highly fragmented landscapes (Ribeiro et al., 2009). This situation further weakens the Atlantic Forest, since fragmentation has consequences on the connectivity and mechanisms responsible for the maintenance of biological diversity over time (Collinge, 1996; Serrano et al., 2002). In this context, ecological restoration is one of the most promising tools to reestablish minimum levels of landscape connectivity to deal with species extinction rates. In areas with low resilience, where the potential for local self-recovery is not sufficient to restore a succession trajectory sufficient to lead to the restoration of the ecosystem, planting tree seedlings has been used as the main method of restoration (Rodrigues et al. 2009). To favor the ecosystem functioning and to recover part of the lost biological diversity with the restoration, the use of a high diversity of arboreal species in the plantations has been prioritized (Ribeiro et al., 2009), whereas it is expected that other forms of plants (epiphytes, lianas, herbaceous and shrubs) colonize these areas over time, without the need for direct reintroduction (Kageyama & Gandara, 2000; Rodrigues et al., 2009).

However, recent evaluations of forest restoration plantations in the Atlantic Forest show that epiphytes have presented very low recolonization levels, even in older plantations. (Damasceno, 2005, Garcia, 2016). Hypotheses such as absence of well-preserved areas that serve as a source of propagules, or, in the case of the existence of forests near the restored areas, it can be expected that the high exigency of this group to colonize such areas is making this process difficult. In a recent review on the restoration of biomass and biodiversity throughout secondary succession in tropical forests worldwide, Martin et. al. (2014) observed that tree richness recovers much faster than that of epiphytes, which did not reach the values observed in the reference ecosystem even after 100 years of succession. This highlights that ecological restoration faces a great challenge, which is the development of techniques for the enrichment of restoration plantations with epiphytes, since these species will certainly have greater restrictions to restore enough levels of biodiversity, especially when inserted in very fragmented landscapes.

Epiphytes are plants that settle directly on the trunk and branches of other trees without the emission of haustoria structures (extensions that suck sap from the host plant). The plants that provide support are called forophytes or hosts (Zots, 2016). These species play an importante role in forest ecosystems: provide wildlife for invertebrates (Ellwood & Foster 2004), they are importance in the interception of water and nutrients cycling (Jordan et al., 1980). Many species of fauna present in tropical ecosystems use resources offered by epiphytic species, including fruit, floral nectar, water, small invertebrates living in these organisms and material for nesting. This work had as objective to answer two fundamental questions for the transplanting of epiphytic species restoration plantations: (i) Does the age of forest restoration planting affect the survival and development of transplanted epiphyte species? (ii) Is the survival and development of the transplanted epiphyte species influenced by the roughness of the bark of host trees?

4.2. METHODS

4.2.1. Study Epiphyte Species

We work with eight epiphyte species native of the Atlantic Forest. An Araceae: *Philodendron bipinnatifidum* Schott ex Endl; two bromeliads: *Aechmea bromeliifolia* (Rudge) Baker; *Billbergia zebrina* (Herb.) Lindl.; three cacti: *Epiphyllum phyllanthus* (L.) Haw.; *Lepismium cruciforme* (Vell.); *Rhipsalis cereuscula* Haw. and two orchids: *Catasetum fimbriatum* (C.Moren) Lindl and *Oncidium flexuosum* (Kunth) Lindl. With exception of the orchids, which were acquired by seedlings in commercial orchid (Bioriquids®), all others species were harvested from at least 10 individuals, located in forest fragments in the region of Piracicaba. The fruits were processed, and seeds germinated in laboratory. Orchid seedlings and seedlings from the other tree families were transplanted into plastic trays with 98 cells, used *sphagnum* as substrate. They were cultivated in greenhouse for six months.

The availability of seeds and seedlings, the geographic distribution (Flora of Brazil 2020) and the interaction with the fauna, through the pollination of the fruits and dispersion of the seeds were the criteria used to choose the epiphytic species. Except for orchids in which seeds are dispersed by the wind, the other six species exhibit zoocoric dispersion (Rowley, 1980, Gottsberger & Jr 1984, Scrok & Varassin 2011, Guaraldo et al 2013, Pansarin et al., 2016).

4.2.2. Study Sites

Five forest restoration plantations, located in the state of São Paulo, in the municipalities of Anhembi, Araras, Iracemápolis, Cosmópolis and Mogi-Guaçu, were selected for this experiment. In the year in which the epiphyte species were transplanted (December 2012), forest restoration plantations ranged in age from six to 57 years. All forest restoration plantations were carried out in river banks or water reservoirs. For 12 months, we evaluated the micromlimate (temperature and relative humidity) inside the five restoration sites with data loggers (Hobo Pro V2). We also mesure the opening of the canopy through a concave spherical densitometer (Table S4.1).

In all forest restoration plantations, we sampled the diversity and abundance of epiphytic species. Plots of 900 m² (20 x 45 m) were randomly installed within each forest, always respecting a distance of at least 30 meters from the edges. All the tree individuals present within the plot were sampled. In each tree we measured the diameter at 1.30 meters of the soil and looked for epiphytic species. When necessary, we used binoculars to assist in the search and identification of species. When the trees were hosts for epiphytic species, we identified the epiphyte species in the field and counted the number of individuals

4.2.3. Epiphyte reintroduction

We transplanted epiphytes to 100 host trees. In each restoration plantation, we chose four host tree species. For each host tree specie, we selected 5 individuals, totalizing 20 host trees in each restoration plantation. The host tree individuals were chosen randomly, within each restored forest plantation. Trees that were <10 meters from the edge were excluded from the study. Host tree species were selected based on the roughness of the bark and its frequency in secondary forests. Whenever possible, the same host tree species were replicated in the five experimental areas. In the case where restoration plantation did not have the host species used, we chose other host species that have similar bark roughness (Table 4.1.). The host species were: *Cariniana legalis* (Mart.) Kuntze; *Cedrela fissilis* Vell.; *Centrolobium tomentosum* Benth.; *Hymenaea courbaril* L. and *Myroxylon peruiferum* L.f.

In each host tree, we transplanted eight individuals of epiphytes, one of each epihyte species studied. The epiphytes were always fixed facing the north, at a minimum height of 8 meters from the ground level. We used palm fibers to stabilize the individuals in the trunk and the lashing was done with natural fibers (Sisal). The order of the species to be tied was chosen at random. Transplants were performed in January 2013.

Table 4.1. Host trees species used in each plantation of forest restoration. The value in parenthesis equals the age of planting at the time of transplanting of the epiphyte species.

Host tree species	Forest Restoration Plantation				
	Anhembi (6)	Araras (10)	Cosmópolis (57)	Iracemápolis (24)	Mogi-Guaçu (13)
C. legalis	X	X	X	X	X
C. fissilis		X		X	X
C. tomentosum	X		X	X	
H. courbaril	X	X	X		X
M. peruiferum	X	X	X	X	X

4.2.4. Performace of reintroduced epiphytes

Epiphyte survival and growth were monitored each six months for the first and second year, and annually for subsequent years through Feburary 2017. Growth parameters measured included length (cm), number of leaves and number of stems. The number of leaves and the number of stems were evaluated in the same way for all eight species (counting). The evaluation of the length varied according to the evaluated species, adapting the predictive variable according to the morphology of the evaluated species. For, *A. bromeliifolia*; *B. zebrina* e *P. bipinnatifidum* the length was evaluated through the same protocol, measuring the size between the base of the individual to the end of the leaf. For *C. fimbriatum* and O. flexuosum the growth was evaluated through the largest stem (Zotz, 1995). The two species grow sympodially, usually developing one stem per year, which are sourced from a main rhizome. However, in some individuals the branching of the rhizome occurs, giving rise to two or more growth points. For *E. phyllanthus*; *L. cruciformis* e *R. cereuscula* the growth was evaluated through the largest branch (junction of the phyloclados). The growth results for each species were analyzed according to the best predictor of growth. We also mesure reproductive parameters (flowering) based on the presence or absence of this event in each species.

4.2.5. Data analysis

We performed survival analysis to assess differences in time until an event (death) occurred in each treatment per species. We used a Kaplan–Meier estimator of survival probability (Therneau, 2012) and log-rank tests to evaluate differences among treatments in host effect, and bark roughness. Two-way analyses of variance (ANOVA) was used to compare independent effects and interactions between, host tree effect, and bark roughness on growth of the eight epiphyte species studied. Statistical analysis were performed with R version 3.2.5 (R Core Team, 2014), using the “OIsurv”, “lme4” and “lmerTest” packages.

4.3. RESULTS

All five forest restoration plantations present low diversity and abundance of epiphyte species. Only the plantations located in the municipalities of Araras, Iracemápolis, and Cosmopolis had, respectively, 0.06; 0.05; 0.12% of sampled host trees occupied by epiphyte species. All epiphyte species sampled were of the *Tillandsia* sp. and were represented by only 1 individual in each host tree (Table S4.2)

After 47 months, survival varied greatly among species when they were transplanted to restoration plantations with different ages (Figure 4.1): *A. bromeliifolia* (0.0-0.50); *B. zebrina* (0.0 - 0.85); *C. fimbriatum* (0.0 - 0.40); *E. phyllanthus* (0.10 - 0.60); *L. Cruciformis* (0.25 - 0.70); *O. flexuosum* (0.20 - 0.80); *R. cereuscula* (0.15 - 0.65). The species *P. bipinnatifidum* died in all areas. In general, the species obtained better survival results in the younger areas. The species *O. flexuosum* produced flowers, but no individual produced fruits. The length (cm) of the species varied from (13.4 - 59.3) while the count in the number of stems ranged from 1-8. We identify the preference of the epiphytes by specific hosts (Table 4.2). The survival, growth and production of flowers were influenced by the characteristics of the host species ($p > 0.05$).

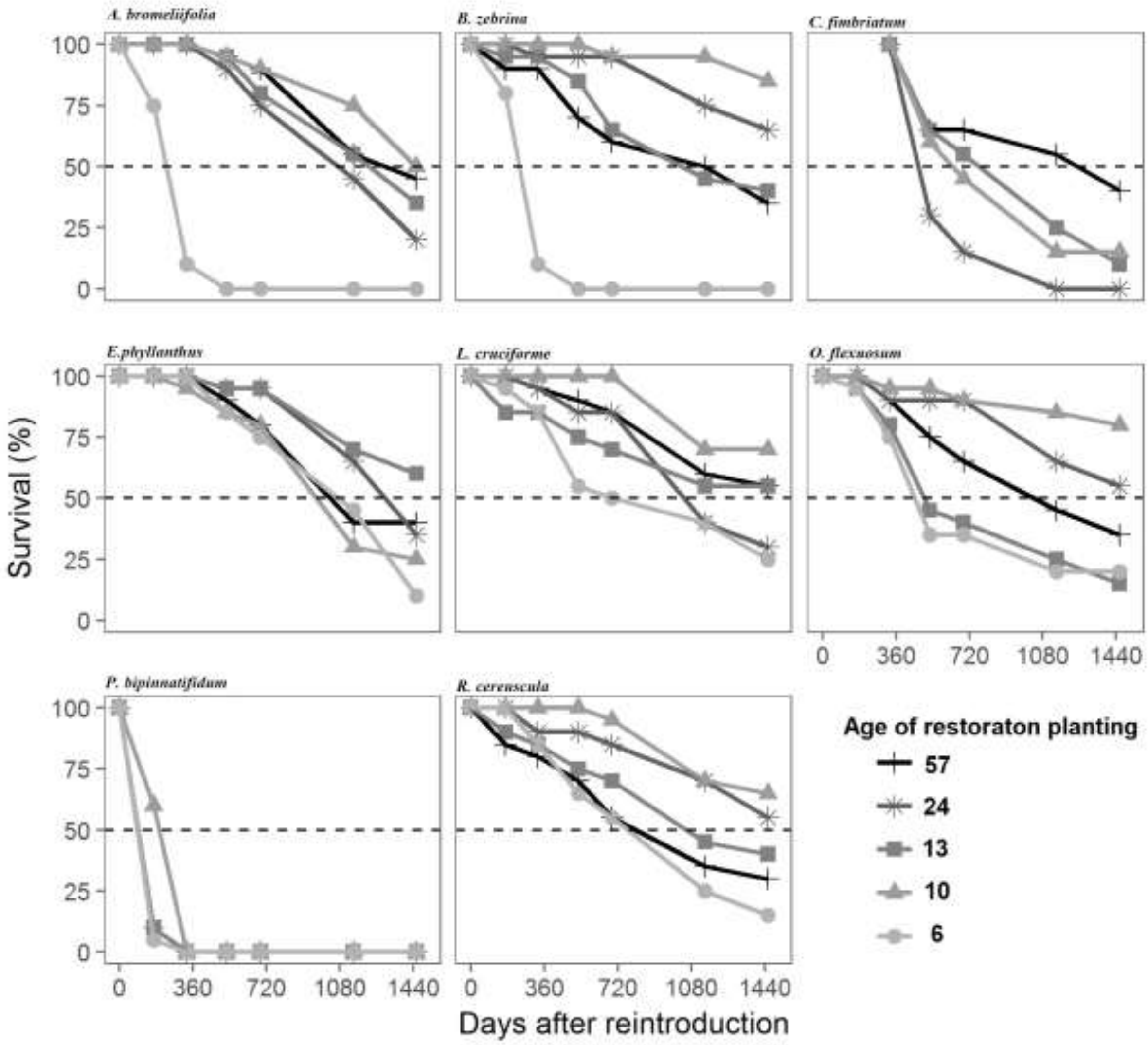

Figure 4.1: Survival (%) of eight epiphytic species: *A. bromeliifolia*; *B. zebrina*; *C. fimbriatum; E. phyllanthus*; *L. cruciform*; *O. flexuosum*; *P. bipinnatifidum* and *R. cereuscula*, transplanted to 20 host trees located in five forest restoration plantations with ages varying from six to 57 years.

Table 4.2 – Survival of epiphytes species in relation to a host species used

Epiphyte species	Host Tree Species					
	C. tomentosum	*M. peruiferum*	*C. fissilis*	*H. courbaril*	*C. legalis*	P value
A. *bromeliifolia*	30.0	25.0	86.7	26.7	25.0	0.005
B. zebrina	50.0	60.0	80.0	40.0	50.0	0.211
C. fimbriatum	20.0	20.0	13.3	13.3	15.0	0.944
E. phyllanthus	20.0	28.0	80.0	35.0	20.0	0.012
L. cruciformis	26.7	52.0	66.7	60.0	32.0	0.244
O. flexuosum	26.7	48.0	66.7	20.0	44.0	0.097
R. cereuscula	20.0	44.0	73.3	60.0	16.0	<0.001
P. bipinnatifidum	0.0	0.0	0.0	0.0	0.0	-

4.4. DISCUSSION

Our results show that forest restoration plantations do not recover naturally the diversity and abundance of epiphyte species. This limitation can be overcome through artificial reintroduction. The transplanted epiphyte species survive and grow and, in some cases, flourish, evidencing that over the years, the expected ecological processes will occur. The age of planting of forest restoration does not seem to be an important factor, and it is possible to reintroduce epiphytic species in young forests. Contrary to our expectations, host tree species do not seem to interfere in species survival and growth, however, new research should be carried out to clarify the species-host relationship.

The specie *P. bipinnatifidum* does not seem to adapt to the method used for transplantation, since all individuals died in the first year of the study (Figure 4.1). Probably the high mortality rate is related to the fact that this species is hemi-epiphyte, therefore other strategies should be developed to reintroduce this specie. The other sevem species obtained acceptable rates of survival and development, demonstrating that the transplantation of seedlings overcomes the natural limitations (dispersion and establishment) imposed on the colonization of host trees by epiphytic species. During the first year, the survival of the eight transplanted species was like other studies (Pett-Ridge & Silver 2002; Schneffknecht et al., 2012; Fernandez et al., 2016; Duarte & Gandolfi, 2017). As the development of epiphytic species varies over the years (Zotz et al., 2005), future studies evaluating transplantation of epiphyte species should monitor the experiments over the years to verify the success of these reintroductions (Duquesnel et al., 2017).

Transplanting seedlings of epiphytes for forest restoration plantations is a promising technique to enrich these new ecosystems, because the natural limitations of seed dispersal and establishment of seedlings are overcome. An alternative to the use of seedlings would be the use of direct seeding, however, low seed germination rates (Winkler et al., 2005; Mondragon and Calvo-Irabien, 2006) and the uncertainty that the transplanted seeds will be able to overcome the environmental filters, favors the use of seedlings. Furthermore, the transplanting of seedlings allows the use of adult plants, which are already in their reproductive stage, consequently the ecological processes will happen more quickly, and transplanted epiphytes can be seed sources, assisting the colonization of new areas.

Contrary to our expectations, the older forest restoration plantation with did not provide the best survival rates for transplanted epiphytes. For some species, the best survival rates occurred in young forest restoration plantation. When transplanting epiphytes to two young forest restoration plantation, Duarte & Gandolfi, 2017, obtained high survival rates in the first year, demonstrating that factors other than age should influence the survival of epiphyte species. Therefore, future projects involving the enrichment forest restoration plantation through the transplant of epiphytic species can occur in young forests. Transplantation of epiphytic species into young forests can be a strategy to accelerate the forest restoration, since several epiphytic species are attractive to fauna, favoring the entry of new species into these new ecosystems.

The difference in the bark roughness of host trees did not provide differences in the survival of the epiphyte species. The effect of bark roughness on the epiphyte species is more expressive in the process of germination and establishment of the seedlings (Callaway et al., 2002). As in our experiment the transplantation occurred through seedlings, probably the filters that act on the seeds were overcome, not being possible to observe interaction between epiphyte and host. In addition, because restoration plantations are young areas, perhaps planted trees have not yet expressed the bark characteristics that are expected when the same species become adults (Flores-Palacios and Garcia-Franco, 2006; Wagner et al., 2015).

Forest restoration plantaton provide an excellent opportunity to test new ideas that will aid in the evolution of restoration ecology. Studying the transplantation of epiphyte species and their interactions with these newly restored ecosystems offers an opportunity to overcome the limitations that epiphytic species face to naturally recolonize forest restoration plantations. New enrichment methods should be tested, thus increasing the survival rate of the transplanted epiphyte species.

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APPENDECES

Table 4.1 – Characteristics of the forest restoration plantations used for the transplantation of eight epiphytic species.

County	Year of planting [1]	Age at the time of transplantation of epiphytic species [1]	Area (ha)[1]	Coordinates [1]	Tmax (ºC)	Tmin (ºC)	Tmed (ºC)	UR max (%)	UR min (%)	UR med (%)	Canopy cover (%)	Characteristics of restoration plantation [1]
Anhembi	2006	6	3.9	22º42’20” S 48º10’01” O	37.2	8.34	21.7	100	14.2	72.5	86.6	Forest restoration plantation of 150 x 300 m, located on the banks of the Tietê River, at the Experimental Station of Forestry Sciences of Anhembi (ESALQ / USP). A total of 80 regional native species were planted, spaced 3 x 2 m, with random distribution of planting groups. At the time of epiphytic transplanting, the canopy of the planting had approximately 8 m, with absence of regenerating species and presence of invading grasses in some stretches of the planting.
Mogi-Guaçu	2002	10	11.5	22º22’23” S 46º58’3” O	38.3	6.4	21.5	100	16.7	69.2	86.7	Forest restoration plantation with 150 meters wide, around a reservoir of water, inside the São Marcelo Park, owned by Internation Paper. A total of 101 regional native species were planted, equally distributed among pioneers and non-pioneers, spaced 3 x 2.5 m. At the time of epiphyte transplantation, the canopy was approximately 15 m high, with the presence of regenerating species, mainly pioneer species.
Araras	1999	13	8.5	22º26’26” S 47º21’41” O	36.0	9.2	22.1	100	16.9	71.1	89.2	Forest restoration plantation with approximately 50 meters around one of the dams of Usina São João, where a native and exotic regional planting was carried out, spaced 3 x 2 m, with random distribution of the planting groups, being unknown the number of species planted

County	Year of planting [1]	Age at the time of transplantation of epiphytic species [1]	Area (ha)[1]	Coordinates [1]	Tmax (ºC)	Tmin (ºC)	Tmed (ºC)	UR max (%)	UR min (%)	UR med (%)	Canopy cover (%)	Characteristics of restoration plantation [1]
												At the time of epiphytic transplantation, the canopy was approximately 18 m high, with a low index of regenerants and presence of invasive species.
Iracemápolis	1988-1990	24	21	22º34’38” S 47º30’32” O	36.9	6.63	22.2	100	17.0	70.7	90.7	Forest restoration plantation of approximately 60 m in the surroundings of the water reservoir for the public supply of Iracemápolis. A high diversity plantation was planted, with approximately 140 native and exotic species, spaced 3 x 3 m. In this plantation was adopted the system of modules containing 9 individuals from the four successional classes (initial, secondary, late secondary and climax). At the time of transplanting the epiphytes, the canopy was approximately 25 m. In the sub-forest regeneration, invasive species such as *Clausena excavata*, *Triplaris americana* and *Leucaena leucocephala*.
Cosmópolis	1955-1969	57	15.2	22º40’19” S 47º12’16” O	38	10.5	24.1	100	17.14	72.8	85.8	Planting of forest restoration of variable width (approximately 200m) on the banks of the Jaguari River. A total of 71 shrub and tree species were planted, with regional natives being preferred, with no spacing or defined alignment, with approximately one tree per 10 m². At the time of transplanting the epiphytes, the canopy was approximately 40 meters high, with large individuals, and very developed understory, being possible to find regenerants of several of shrubs and trees.

[1]Adaptado de Silva, C. C. 2013. Potencial de espécies nativas para a produção de madeira serrada em plantios de restauração florestal. Dissertaçao de Mestrado. 99p.

Table S4.2. Characterization of epiphyte species in five forest restoration plantations with ages varying from six to 57 years. Plot size: 900 m²

Forest restoration plantation	**Age**	**Number of trees**	**Basal area (m²)**	**No. epiphytes species**
Anhembi	**6**	102	1.64	0
Mogi-Guaçu	**10**	72	1.74	0
Araras	**13**	50	3.52	5
Iracemápolis	**24**	49	2.38	3
Cosmópolis	**57**	72	1.74	9

5. FINAL CONSIDERATIONS

In this work we verified that the epiphyte species have natural limitations to colonize forest restoration plantations. The assisted reintroduction, using transplant of seedlings or direct seeding, are promising methodologies that can reverse this situation, favoring the establishment of epiphytic species in the forest restoration plantation. When using seedlings, it is possible to obtain high survival rates. For some species, it is also possible to obtain fruits and flowers, thus reestablishing the ecological functions performed by epiphytic species in forest ecosystems. The transplanting of epiphytes through direct seeding is an alternative to the transplanting of seedlings, but adjustments in this method are still necessary, because although they germinate easily, the seedlings face difficulties in the firing, mainly due to the high hebivory rates.

We study the reintroduction of epiphytic species in the context of forest restoration, but this theme should also be extended to secondary forests, since these ecosystems provide similar challenges and difficulties to restored forests. Few epiphytic species can colonize secondary forests, and often, epiphyte species found in secondary forests have low conservation value, as it is possible to find these species in highly degraded areas and even in urban areas (e.g. *Tillandsia sp*.). Therefore, secondary forests are not favoring the conservation of epiphyte species. This phenomenon also reflects the possibility of epiphytic species colonizing new areas, since if they were present in the secondary forests, their propagules and seeds could be more easily dispersed to new areas.

Another factor that contributes negatively to the conservation of epiphyte species is the negligence of the public authorities. Often, when an enterprise or public utility needs to degrade or even deforest an area with native vegetation, one of the counterparts required by the public authorities may be compensation for the deforested area through the planting of new areas. However, in this new planting, only tree and shrub species are planted, that is, this mechanism allows the exchange of a forest in which different life forms coexist (trees, shrubs, herbs, palms, lianas, epiphytes) by a planting that does not respect, and that it will be difficult to recover all the biodiversity of the deforested forest. Therefore, favoring epiphytes and other forms of life is an essential mechanism for environmental compensation projects to play the role of biodiversity conservation that is expected of them.

Forest restoration projects also have the same bias as environmental compensation projects. Particularly in the state of São Paulo, where this research was carried out, the criteria adopted to monitor the success of restoration plantations are based on tree and shrub species (native vegetation cover (%); ha), number of native regenerating species), that is, epiphytic species and other forms of life are not monitored and are not accounted for to determine the success of restoration projects. It is evident that the public agencies are based on the premise that the forest structure will allow the arrival of new forms of life. However, as we demonstrate in this work, this phenomenon does not occur naturally, so artificial reintroduction is necessary. In order for new forests from restoration projects to effectively fulfill their expected role in biodiversity conservation, more attention should be paid to epiphytes and other forms of life.

In addition to the reduction in populations of epiphytes due to the degradation of natural forests, predatory exploitation is also a prominent factor for epiphytic species, especially orchids, due to their high commercial value. Many epiphytic species are removed from the naturian forests, increasing the risk of local extinction, affecting the conservation of these species and the communities in which these species were suppressed. Forest restoration plantations, as well as secondary forests, provide an opportunity to reverse this situation, since epiphyte species can be reintroduced into these ecosystems with two objectives: local conservation and income generation. For this to become a viable alternative both ecologically and economically, further research will be needed to understand the biology and ecology of epiphyte species. Many species have as reproductive strategy the production of lateral buds, which are transformed into shoots, being a clone of the mother plant. These shoots can be managed to give rise to a new individual, which can be used to enrich new forests in restoration and secondary forests, as well as be commercialized, diversifying the income generation of rural owners.

The reintroduction of epiphyte species is a universe within the restoration ecology. Through this research we have demonstrated that it is possible to succeed with the transplanting of seedlings and epiphyte species, which allows restored forests and forest restoration plantations to become ecosystems with a greater degree of complexity, favoring new networks of interactions between species. It is necessary to continue testing new methods of reintroduction, mainly to find out which methods are more efficient according to the transplanted epiphyte species.