



Use of seed balls as a technique to improve seedling emergence and establishment in an area degraded by the disposal of urban solid waste

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Abstract

Direct seeding is used for the restoration of extensive areas, but seed germination, seedling stabilization, and survival depend on the development of adequate techniques. This study evaluated plant emergence, survival, establishment, and growth using seed balls with different substrates for the restoration of a decommissioned landfill area in the municipality of Diamantina, Minas Gerais, Brazil. Seed balls made with manure and 45%, 60%, and 75% clay, containing seeds of four tree species and two herbaceous species, composed the treatments with low (T1), medium (T2), and high (T3) clay content. Seedling emergence was assessed after 60 days, and survival, establishment, and growth (final height–initial height) were evaluated after 12 months. Seed balls containing 60% clay (T2) resulted in the highest seedling emergence and height growth, while survival and establishment were generally greater in the treatments with medium and high clay contents than in the low-clay treatment. Overall, the medium-clay formulation provided the most favorable balance among the evaluated recruitment indicators. Under the environmental conditions of this study, seed balls containing 60% clay improved early plant recruitment and establishment, indicating their potential use in the initial stages of ecological restoration of degraded areas.

Keywords Abandoned areas · Seed balls · Ecological restoration · Direct seeding · Soils

Introduction

The reduction of environmental quality is one of the main problems associated with deficient solid waste management in urban areas. The disposal of such waste in open

dumps increases the concentration of nutrients and chemical substances, reducing oxygen in the soil and water and increasing pollution by slow-decomposing materials (Brand and Spencer 2023). Reduced soil infiltration after the deactivation of these sites may increase the occurrence of floods and erosive processes (Brand and Spencer 2020), in addition to facilitating invasion by exotic plants and reducing the quality of the natural soil structure and function and natural regeneration (Assis et al. 2020), making the application of environmental restoration techniques in these areas necessary.

Direct seeding is a large-scale viable revegetation technique in locations invaded by plants (Atondo-Bueno et al. 2018), but low survival rates reduce its efficiency (Pérez et al. 2019). Therefore, technologies aimed at improving seed viability and plant establishment in the field have been enhanced (Taylor et al. 2020). Because early recruitment is often a major bottleneck in restoration programs, understanding factors that improve seedling emergence and

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persistence during the first year is essential, although these responses do not necessarily predict long-term ecosystem recovery.

The germination and emergence of *Acacia acuminata*, *Calothamnus sanguineus*, *Hakea laurina*, and *Melaleuca seriata* were higher when their seeds were coated with different substrates made from soil (Harshani et al. 2024). The dispersal of seeds of *Lindernia procumbens*, a threatened species in Europe, improved when coated with a natural and synthetic polymer and adhered to rice seeds (Corli et al. 2024). Additionally, pellets containing activated carbon protected grass seeds from herbicide effects (Brown et al. 2019).

Seed germination, establishment, and protection against predation and deterioration—especially by insects such as ants, as well as small mammals and birds—are improved when seeds are coated with substrates composed of water, clay, and soil, as well as fertilizers (Gornish et al. 2019; Andrés et al. 2024). Climate change and severe droughts increase the need to create microhabitats favorable to plants with improved water retention (Mirsaifi et al. 2024). Seed coating, particularly with multiple species combined and aggregated, increases the size, weight, and surface texture of the pellet, improving distribution at the restoration site and reducing seed dispersal by wind or rain (Hoose et al. 2019; Harshani et al. 2024). This technique, still little known in the restoration of degraded areas (Overdyck et al. 2013; Jones et al. 2014), facilitates the distribution of seeds with different ecological functions within a single nucleus, simulating natural conditions (Jones et al. 2014; Gornish et al. 2019; Holbert et al. 2019).

In addition to tree species, fast-establishing herbaceous species were included in the seed balls as functional nurse components to improve early restoration performance. Herbaceous plants were not included as final successional species, but as temporary facilitator species. These species may rapidly provide soil cover, reduce erosion, buffer soil temperature, enhance moisture retention, add organic residues, and help suppress invasive grasses during the initial phase of ecosystem recovery. Their inclusion aimed to complement woody species establishment under the stressful conditions commonly found in degraded landfill areas.

The objective of this study was to evaluate the emergence, survival, establishment, and growth of plants using seed balls with different clay contents in the restoration of a decommissioned urban solid waste disposal site.

Materials and methods

The experiment was established in an area of the former waste disposal site (open dump) of the city of Diamantina, Minas Gerais, Brazil, located on the JK campus of the Federal University of the Jequitinhonha and Mucuri Valleys (UFVJM) (18° 12' 17" and 43° 34' 08", 1390 m altitude), with a humid temperate climate characterized by dry winters and rainy summers, according to the Köppen classification. The mean annual precipitation and temperature were 1,468 mm and 18 °C, respectively. The predominant vegetation in the study area is *cerrado campestre* (Ribeiro and Walter 2008). The soil is classified as dystrophic Quartzipsamment (*Neossolo Quartzarênico*) with a high sand content and low organic matter, indicating reduced nutrient availability (Freitas et al. 2012).

The dump site, decommissioned in 2003, was covered with approximately 50 cm of soil and fenced for revegetation using non-native tree species and sowing of exotic grasses (*Melinis minutiflora* and *Urochloa* spp.) to accelerate ground cover and reduce erosion. Fourteen years after revegetation, the grasses, with high dominance and biomass production, covered the soil. In March 2017, chemical control of the dominant exotic grass (*Melinis minutiflora*) was carried out using the herbicide glyphosate. The herbicide glyphosate was chosen due to its low residual activity; it is practically biologically inactive due to its strong binding to soil components (Duke 2020). Therefore, it does not interfere with the establishment of subsequently introduced plant species. The chemical weeding consisted of the application of glyphosate herbicide (1440 g ha⁻¹) using a CO₂ pressurized costal sprayer (Guarany, Backpack Fire Pump—S4 20 l, Brazil, Itu—SP). After the grass was controlled, the area was prepared (Fig. 1A), and, 20 days later, the seed balls were scattered around the site. Controlling the grass was necessary to favor the development of native species, since *M. minutiflora* is a highly competitive species and could interfere with their growth.

Seeds of tree and herbaceous species commonly used in restoration projects or recognized for rapid establishment and soil cover functions were selected (Table 1). In addition to tree species, fast-establishing herbaceous species were included in the seed balls as functional nurse components to improve early restoration performance. Herbaceous plants were not included as final successional species, but as temporary facilitator species. These species may rapidly provide soil cover, reduce erosion, buffer soil temperature, enhance moisture retention, add organic residues, and help suppress invasive grasses during the initial phase of ecosystem recovery. Their inclusion aimed to complement woody species establishment under the stressful conditions commonly found in degraded landfill areas. Tree species were

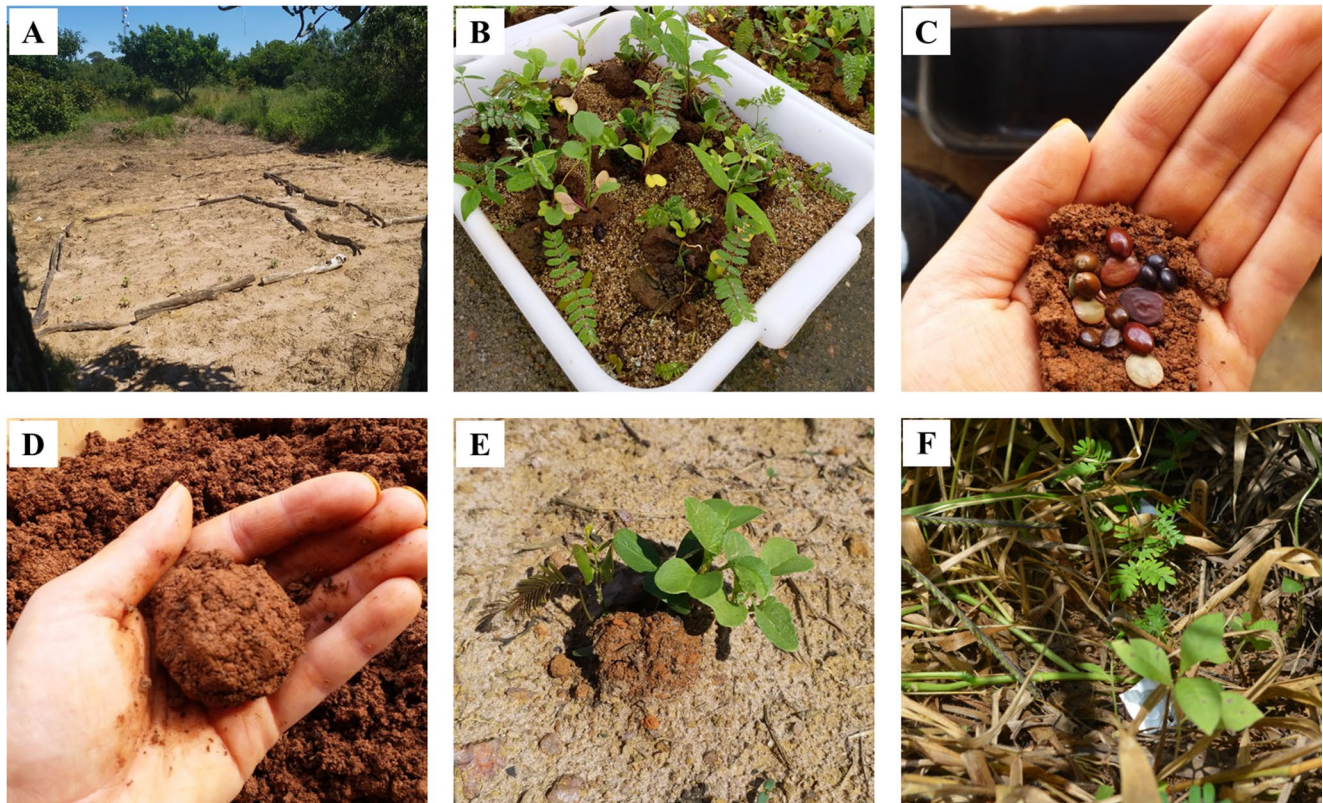


Fig. 1 **A** Image of the area prepared after controlling *Melinis minutiflora* with glyphosate. **B** Germination test of the seeds used. **C** and **D** Illustration of the preparation of seed balls. **E** and **F** Development of the seed balls in the field

Table 1 Species, families, number of seeds per seed pod (NS), Lifestyle (trees or herbaceous plants), Functional Group (pioneer, early secondary, and temporary facilitator), life cycle (perennial or annual), origin (native or exotic), and germination (%) of the species used in the seed balls

Species	Families	NS	Lifestyle	Functional group	Life cycle	Origin	Germination (%)
<i>Senegalia polyphylla</i>	Fabaceae	4	Tree	Early secondary	Perennial	Native	86
<i>Piptadenia gonoacantha</i>	Fabaceae	4	Tree	Pioneer	Perennial	Native	56
<i>Senna macranthera</i>	Fabaceae	4	Tree	Pioneer	Perennial	Native	82
<i>Crotalaria ochroleuca</i>	Fabaceae	2	Herbaceous	Temporary facilitator	Annual	Exotic	74
<i>Raphanus sativus</i>	Brassicaceae	2	Herbaceous	Temporary facilitator	Annual	Exotic	78

chosen for medium-term vegetation structuring, whereas herbaceous species were included to promote early ground cover and improve microsite conditions for seedling establishment. The seeds were stored in a cold chamber at 6 °C and 40% relative humidity and used after germination tests (Fig. 1B). Seeds of *Senna macranthera* were subjected to dormancy breaking (scarification with sandpaper), and all seeds used, including those of this species, were placed in a BOD-type (Biochemical Oxygen Demand) (ADAMO, Model 72, Brazil – São Paulo—SP) germination chamber with a 12 h photoperiod and temperature of 30 °C for 30 days, after which their viability was assessed.

The experimental design adopted was a randomized complete block design with four replicates. Each block consisted of three experimental plots (one per treatment), totaling 12 plots. Each plot had an area of 10 m² (4 × 2.5 m) and

was considered the experimental unit for all statistical analyses. Within each plot, 15 seed balls were distributed, totaling 180 seed balls across the entire experimental area. Each seed ball contained 16 seeds of the plant species described in Table 1. Seed balls were treated as sampling subunits and were used solely to calculate plot-level means, not as independent replicates. The treatments consisted of different clay proportions in the seed ball matrix. Thus, the experimental factor was the clay content of the formulation: T1 – low (45%), T2 – medium (60%), and T3 – high clay content (75%). For the preparation of the seed balls, the seeds were randomly distributed in a substrate composed of soil and manure. The proportion of clay was adjusted according to the treatments (45%, 60%, and 75% of the original soil used to make the seed balls). Therefore, the proportion of manure was 55%, 40%, and 25%, respectively. The manure

Table 2 Soil chemical and texture characteristics of soil used in the preparation of seed balls

Variables	Values
<i>Chemical properties of soil</i>	
pH in H ₂ O (mg dm ⁻³)	4.84
P (mg dm ⁻³)	0.14
K ⁺ (mg dm ⁻³)	14.18
Ca ²⁺ (cmolc dm ⁻¹)	0.03
Mg ²⁺ (cmolc dm ⁻¹)	0.01
Al ³⁺ (cmolc dm ⁻¹)	0.76
H+Al (cmolc dm ⁻¹)	2.73
SB (cmolc dm ⁻¹)	0.08
t (cmolc dm ⁻¹)	0.83
T (cmolc dm ⁻¹)	2.80
m (%)	90.80
V (%)	2.72
Organic matter (dag kg ⁻¹)	2.90
<i>Physical attributes of the soil</i>	
Total sand (%)	26.50
Clay (%)	60.00
Silt (%)	13.50

used was from dairy cattle fed corn silage. The manure was acquired from a farm in the Diamantina region, Minas Gerais, Brazil. Both the soil and the manure were dried in the shade in a well-ventilated area and then sieved through a 2 mm sieve. The balls were made with an average diameter of 5 cm and an average weight of 70 g. The characterization of the soil used to prepare the seed balls is shown in Table 2. The mixture was moistened, homogenized, and then manually compacted to form the seed balls, which were subsequently dried in the shade for 72 h (Fig. 1C and D). The experimental design is illustrated in Fig. 2. For statistical

analyses, data obtained from the 15 seed balls within each plot were aggregated by calculating means, with the plot considered the independent experimental unit.

Seedling development was monitored in the field (Fig. 1E and F). The percentage of emergence was evaluated at 60 days. Survival (%) (proportion of emerged seedlings that remained alive 12 months after sowing), establishment (plants m⁻²)=density of live individuals per unit area 12 months after sowing, representing the net result of recruitment after emergence and mortality processes), and growth (final height at 12 months minus initial height – evaluated 180 days after seed ball dispersal) were also evaluated. Because seed balls progressively disintegrated after field deployment, treatment effects observed at 12 months were interpreted as carry-over consequences of early recruitment conditions rather than persistent direct structural effects of intact seed balls.

The data were subjected to analysis of variance (ANOVA) and, when significant ($\alpha < 0.05$), means were compared using Tukey's test ($\alpha < 0.05$) in R software, version 3.5.0 (2018).

Results

A total of 1,177 seedlings (40.86%) of tree and herbaceous species emerged from the 2,880 seeds broadcast in the seed balls. The treatment with medium clay content (T2) resulted in the highest emergence percentage (Fig. 3).

Seedling emergence of the tree species *S. polyphylla* was higher in T1 than in T3; for *P. gonoacantha*, T2 was higher

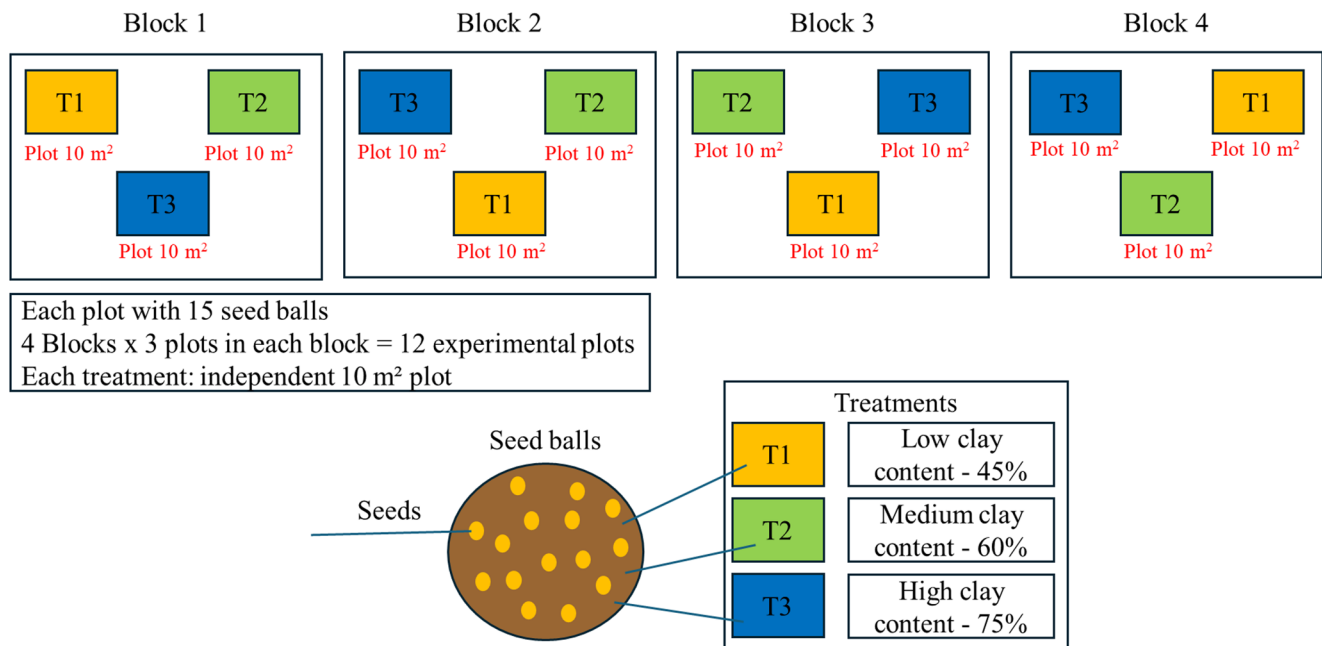
**Fig. 2** Experimental design

Fig. 3 Seedling emergence (%) in the field sixty days after the implementation of the experiment with seed balls (treatments with low (T1), medium (T2) and high (T3) clay content). Columns with mean and bars of standard deviation. Columns with the same letter do not differ by Tukey's test at 5% significance

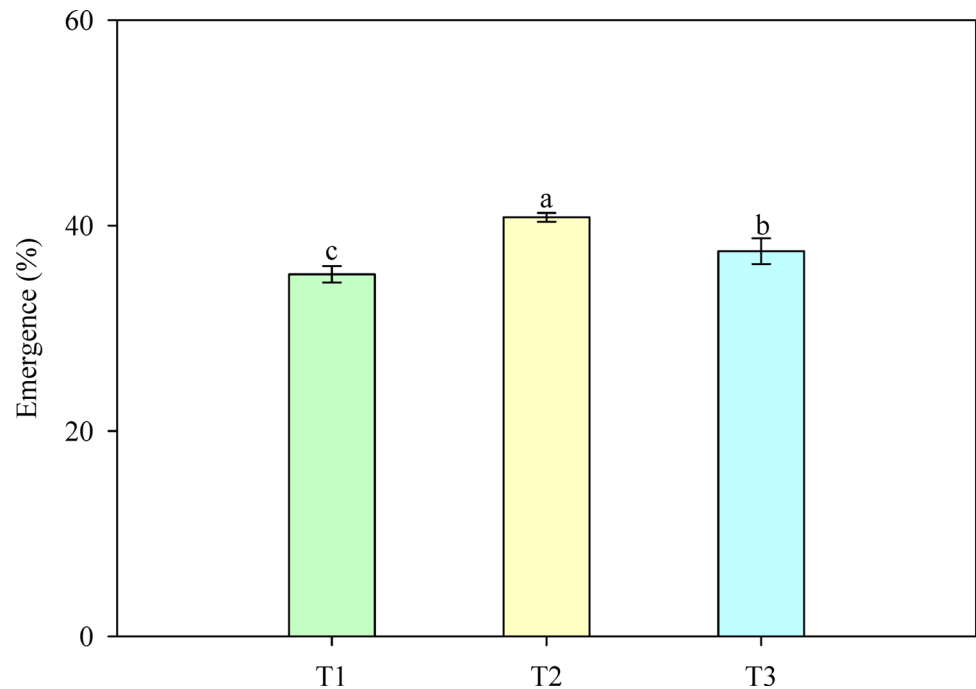


Table 3 Emergence (mean±standard deviation) (%) by plant species in the field sixty days after the release of seed balls with low clay content (T1), medium clay content (T2) and high clay content (T3)

Habit	Species	Treatments		
		T1	T2	T3
Trees	<i>Senegalia polyphylla</i>	36.0±1.8 a	33.0±2.6 ab	31.0±1.8 b
	<i>Piptadenia gonoacantha</i>	17.0±1.8 b	24.0±2.4 a	19.0±2.6 ab
	<i>Senna macranthera</i>	23.0±1.4 b	33.0±1.4 a	30.0±2.6 a
Herbaceous	<i>Crotalaria ochroleuca</i>	50.8±1.7 b	59.5±2.6 a	55.0±2.2 ab
	<i>Raphanus sativus</i>	49.5±1.7 b	54.5±1.3 a	52.5±1.7 ab

than T1; and for *S. macranthera*, T2 and T3 were higher than T1 (Table 3). Seedling emergence of the herbaceous species *C. ochroleuca* and *R. sativus* was higher in T2 compared to T1 (Table 3).

The survival percentage of tree seedlings was similar between treatments T2 and T3 and higher than in treatment T1 (Fig. 4A). At the species level, *S. polyphylla* showed higher survival in T3, *P. gonoacantha* had higher survival in T2 than in T1, and *S. macranthera* exhibited a higher value in T3 compared to T1 (Fig. 4B).

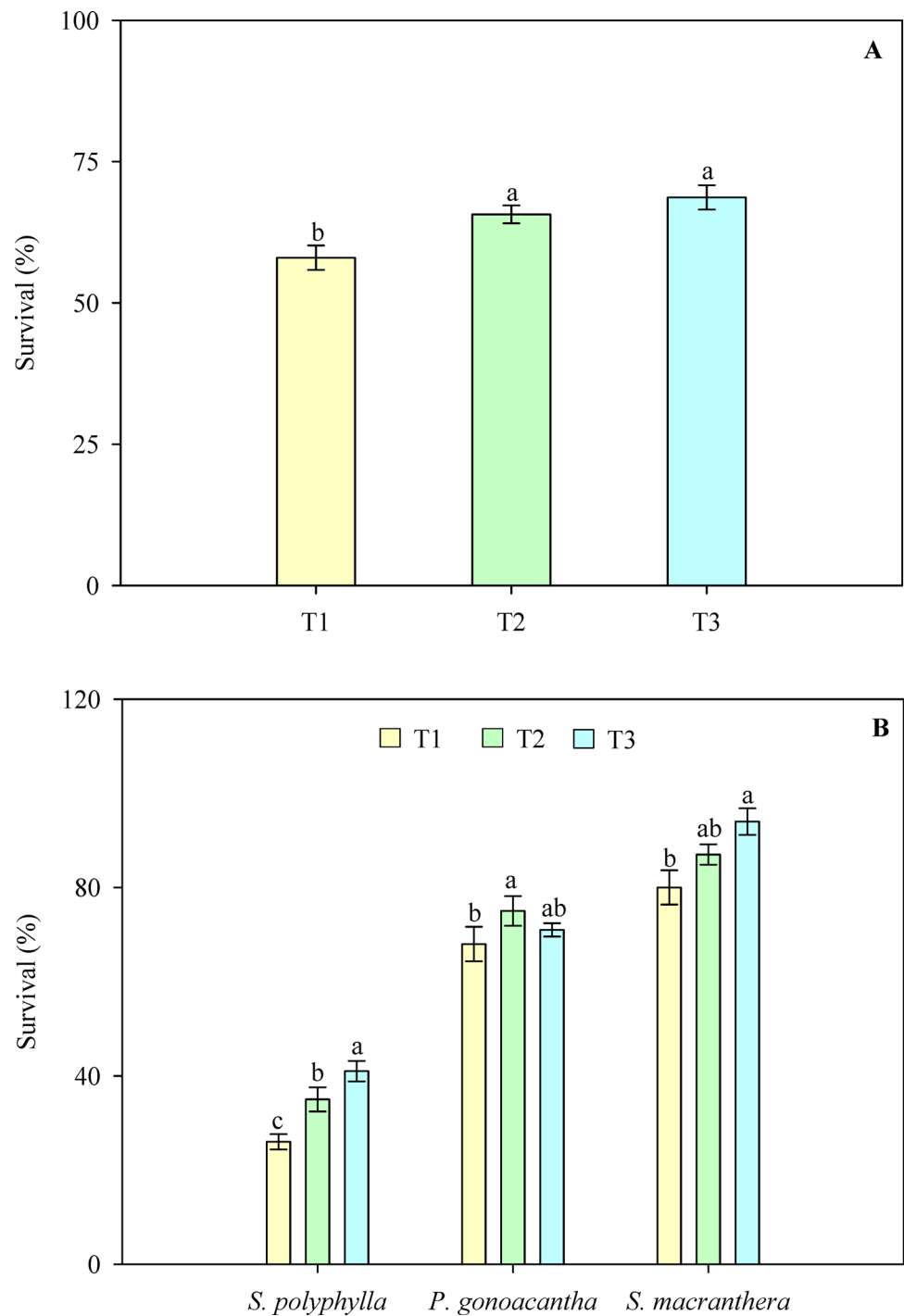
Seedling establishment was higher in treatments T2 and T3 compared to T1 (Fig. 5A). At the species level, *S. polyphylla* showed greater establishment in T2 and the lowest in T1; *P. gonoacantha* presented higher values in T2 compared to the other treatments; and *S. macranthera* exhibited similar values across all treatments (Fig. 5B).

The height increment of tree species after 12 months was greater in treatment T2 than in T1 and T3 (Table 4).

Discussion

The seedling emergence rate (40.86%) was high compared with the general average of 10% reported in other direct seeding studies (Oliveira et al. 2019). However, germination varies according to the position of the seed within the sphere (surface or center), moisture, and compaction of the material composing the seed balls (Lieurance et al. 2024), as well as temperature. Additionally, some species may exhibit delayed emergence (Harshani et al. 2024), but these individuals may be important for maintaining plant diversity (Robinson et al. 2024). The higher emergence percentage of tree and herbaceous species in the treatment with 60% clay (medium – T2) indicates an improvement to the microsite resulting from the combined effects of clay content and manure, including increased water retention (Scalon et al. 2003). Differences between T2 and T3 may be associated with variations in infiltration and gas exchange (Madsen et al. 2016; Benvenuti 2023), which are lower in T3 due to its high clay content (75%), reducing germination and, consequently, seedling emergence. In soils with low clay content, larger particles form larger pores (macropores) filled with air, whereas clay-rich soils contain smaller particles forming smaller pores (micropores) that retain water (Fermino 2002), thereby reducing germination in clayey substrates (Madsen et al. 2016). Conversely, the lower seedling emergence in T1 can be explained by the soil/substrate structure

Fig. 4 Plant survival by treatment (A) and species (B) in the field 12 months after the implementation of the experiment with seed balls (treatments with low (T1), medium (T2) and high (T3) clay content). Columns with the same letter, by plant species, do not differ by Tukey's test at 5% significance



(Hillel 1982) with 45% clay, whose less clayey composition accelerates disintegration, exposing the seeds to herbivory, desiccation, and wind. Because clay proportion increased at the expense of manure content, treatment effects should be interpreted as the result of interacting physical (clay-mediated) and organic (manure-mediated) influences on micro-site conditions.

Although not statistically compared, herbaceous species exhibited numerically higher germination values than tree

species. The higher germination percentage of herbaceous species confirms previous findings for degraded, high-light environments (Souza et al. 2021), yet with lower values than those reported in similar studies, which ranged from 65.3 to 85.6% (Paz et al. 2016; Souza et al. 2021). Moreover, seed germination and seedling development depend on other factors such as seed size (Caldas et al. 2024), with smaller seeds, such as *Crotalaria ochroleuca* and *Raphanus sativus*, germinating more easily in environments with high

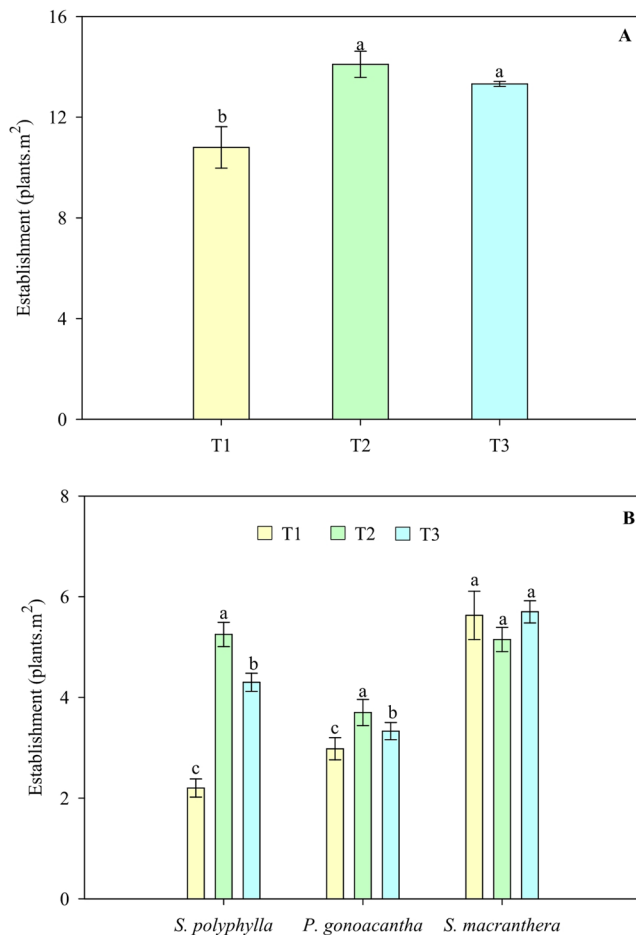


Fig. 5 Total number of plants (A) and by species (B) established per square meter in the field 12 months after sowing with seed balls with low (T1), medium (T2) and high (T3) clay content (mean $\pm$ standard deviation). Bars with the same letter, by species, do not differ by Tukey's test at 5% significance. Means followed by the same letter in each column do not differ by Tukey's test at a 5% significance level

Table 4 Increase in plant height sixty days after direct sowing with seed balls in treatments with low clay content (T1), medium clay content (T2) and high clay content (T3)

Height increase	
Treatments	Height (cm)
Low clay content (T1)	6.4 $\pm$ 1.5 b
Medium clay content (T2)	7.7 $\pm$ 1.2 a
High clay content (T3)	6.5 $\pm$ 1.9 b

solar radiation (Lai et al. 2016). Beyond emergence performance, herbaceous species may play an important facilitative role during the first stages of restoration. Rapid canopy formation near the soil surface can reduce raindrop impact, decrease evaporation, increase litter inputs, and create more favorable microsites for woody seedling recruitment. Thus, their inclusion in seed balls should be interpreted not as a final successional target, but as a temporary functional strategy to accelerate ecosystem recovery. Finally, the medium

clay content treatment enhanced germination of herbaceous species compared with the low clay treatment, indicating an improvement in the microsite due to greater water retention and adequate gas exchange. For tree species, a similar pattern was observed, except for *S. polyphylla*, which exhibited higher germination in the low clay treatment compared with the high clay treatment, possibly due to a greater requirement for gas exchange during germination relative to the other species.

The higher survival of tree seedlings in T2 and T3 confirms that seed balls with greater clay content and lower manure proportion are more suitable for seedling development and maturation, and consequently, survival, due to the increased retention and availability of water and nutrients in clayey substrates (Maluf et al. 2015). Because manure content decreased with increasing clay proportion, differences in survival may also reflect variation in localized nutrient availability in addition to changes in moisture retention. The lower survival in T1 is attributed to its sandier texture, and reduced water retention, which impair plant performance, reinforcing the importance of soil moisture for plant survival (Ritchie et al. 2020). Numerically, *P. gonoacantha* and *S. macranthera* presented higher survival compared with *S. polyphylla*, which may be related to seed size and ecological group. Plants with smaller seeds, having thinner seed coats, tend to show greater survival (Ritchie et al. 2020) due to higher germination percentages (Guzmán et al. 2023). *S. polyphylla* seeds are the largest among the tested species, increasing exposure as the seed balls disintegrated, a factor that may have reduced survival through predation in T1 and T2 (Overdyck et al. 2013). Furthermore, this species is a secondary species with lower performance under full sunlight (Alves et al. 2021). *S. macranthera* and *P. gonoacantha* are pioneer species, characterized by rapid growth and good performance under full sunlight, improving conditions for non-pioneer species (Silva et al. 2015).

The higher number of established plants in the treatments with medium (T2) and high (T3) clay content was expected, since increased water and nutrient retention in clay soils benefits plants (Pradeep and Bell 2025; Leewis et al. 2022; Miao and Wang 2022). This result is better interpreted as an indirect consequence of improved initial recruitment conditions than as a direct, persistent effect of the seed balls after 12 months. During the initial post-sowing period, the clay and manure matrix likely increased local water retention, reduced the risk of desiccation, and temporarily provided nutrients, thus favoring root and shoot development. Seedlings that achieved greater initial vigor may have had a greater capacity to survive and compete after the disintegration of the seed balls, resulting in a higher final establishment density. In this context, the relatively higher manure content in T2 compared with T3 may have contributed to

improved early nutrient availability, which is particularly relevant in the nutrient-poor soil of the study area. However, the greater establishment of *P. gonoacantha* and *S. polyphylla* in T2 compared with T3 may indicate that substrates with very high clay content are more compact, reducing root growth and seedling establishment (Ribeiro et al. 2010; Silva et al. 2022). This contrasts with *S. macranthera*, which showed similar establishment across treatments probably due to its high plasticity in soils of varying textures, making it suitable for ecological restoration of degraded areas (Silva et al. 2015).

The greater height increase observed in T2 after 12 months is more plausibly interpreted as an indirect carry-over effect of more favorable early recruitment conditions than as a persistent direct effect of the seed ball structure. A moderate clay content may have provided a better balance between moisture retention and aeration during early seedling development, while T3 may have been too compact and T1 may have had less favorable moisture conditions. Since manure content varied inversely with the clay ratio, physical and nutritional factors may have contributed to the greater cumulative growth observed in T2. Adequate movement of water and nutrients to the plant favors stomatal conductance and growth (Koehler et al. 2024). On the other hand, the high clay content in T3 may increase water and nutrient retention (Nicoloso et al. 2008), but its high microporosity tends to reduce aeration and increase penetration resistance, limiting root elongation and nutrient absorption (Stefanoski et al. 2013). The lower height growth in T1 may be due to the smaller root-soil contact surface in sandy soils (Rosolem et al. 1999) and the low soil water potential, reducing plant transpiration through stomatal closure, limiting CO₂ absorption and photosynthesis (Demicheli et al. 2023) and, consequently, reducing plant growth (Chauhan et al. 2023). Once the seed balls disintegrated, seedlings that had achieved an initial size or biomass advantage were likely better able to acquire light, water, and nutrients, tolerate environmental stress, and compete with neighboring vegetation. Since the manure content varies inversely with the proportion of clay, there are both physical (related to clay) and nutritional (related to manure) contributions to seedling growth. This developmental head-start may explain the greater cumulative height growth recorded in T2.

Seed balls containing 60% clay provided the most favorable balance between clay and manure proportions, enhancing early recruitment processes (emergence and initial growth), generating carry-over benefits that resulted in greater plant density after 12 months. These results suggest that both substrate physical properties and organic inputs acted together to determine seedling performance. In addition, combining woody and herbaceous species in seed balls represents a promising functional strategy, as herbaceous

plants can rapidly occupy the site and improve conditions for subsequent tree establishment. The practical implications of this study highlight the potential of seed balls as an accessible, low-cost, and viable technique for ecological restoration, particularly in degraded areas where soil infertility, erosion, and seed predation limit plant establishment. The superior performance of seed balls with 60% clay demonstrates that simple modifications to substrate composition enhance and benefit germination, survival, and initial growth, improving restoration success. This technique can be integrated into restoration programs, environmental compensation projects, and community initiatives, facilitating the reintroduction of native species and accelerating fauna recovery in abandoned, disturbed, or biologically invasive areas. By reducing seed loss, improving microenvironmental conditions, and enabling the sowing of multiple species, seed balls represent a promising strategy for restoring ecosystem structure and function across a wide range of degraded landscapes.

Although emergence, survival, establishment, and early growth are important indicators of restoration progress, these short-term responses should not be interpreted as definitive evidence of long-term restoration success. Vegetation trajectories in degraded environments are strongly influenced by subsequent competition, climatic variability, species interactions, and reinvasion processes. Continued monitoring will be necessary to determine whether the early advantages observed in T2 persist over time and translate into greater canopy cover, higher native species persistence, improved soil quality, and more stable plant community development.

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Declarations

Conflict of interest The authors declare that they have no conflict of interest.

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