Topsoil depth influences the recovery of rupestrian grasslands degraded by mining

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ABSTRACT: Close association of iron mining and ferruginous rupestrian grassland places this ecosystem in a special condition of vulnerability, with a large number of degraded areas requiring restoration. Seedling transplantation and topsoil translocation can be used to recover native vegetation in degraded areas. This study aimed to experimentally test the application of two different topsoil depths (0.20 and 0.40 m) in a degraded area. We assessed the vegetation’s natural recovery and the survival of transplanted native species from rescue operations in four 200 m² plots established in each topsoil depth. There was no influence of topsoil depth on the plant species survival, while the vegetation cover was greater on the thicker topsoil. However, exotic species with invasive potential contributed substantially to this vegetation cover, requiring management planning. Some planted native species stood out for their survival ability in the topsoil 49 months after planting. Application of 0.20 m topsoil layer showed to be able to provide native species’ natural recovery and survivorship.

Keywords: canga, campo rupestre, ecological restoration, differential survival, flora rescue.
INTRODUCTION

Ecological restoration aims to develop effective methods to restore the structural and floristic characteristics of an ecosystem (Gann and Lamb, 2006). This is particularly important in tropical non-forest ecosystems where deforestation has been expanding rapidly, producing strong effects on ecosystem functioning (Fernandes et al., 2016a). Moreover, the knowledge of restoration and resilience in these ecosystems is still limited (Buisson et al., 2018). Seedling transplantation, in situ germination of native seeds, and propagule germination from the topsoil are ways to re-establish indigenous vegetation (Skrindo and Pedersen, 2004; Fernandes et al., 2016b; Gomes et al., 2017).

Topsoil can be defined as the upper layer of soil, with marked presence of biological activity and organic matter. Revegetation from topsoil is based on germination from the propagule bank followed by the natural succession, defined as the nonseasonal, directional and continuous pattern of colonization and extinction by species populations on a site (Skrindo and Pedersen, 2004). Topsoil translocation has been applied experimentally in a wide range of ecosystems and shows potential as a method for restoring species diversity (Tozer et al., 2012; Merino-Martín et al., 2017). Studies of topsoil application to restore biodiverse tropical grasslands are important for effective restoration guidance (Pilon et al., 2018; Onésimo et al., 2021). The process exploits propagules present in the topsoil of a donor site to recreate a similar species assemblage at a recipient site (Tozer et al., 2012). When topsoil removal is necessary for mining, the soil may then be used as a substrate and propagules source for the reclamation or restoration of degraded areas (Holmes, 2001; Mendonça, 2013; Ferreira et al., 2015). This technique enables transposing some ecological filters of the natural regeneration process, such as seed availability, dispersion, and germination (Ferreira et al., 2015).

The Iron Quadrangle (QF) is a large ore reserve located in southeastern Brazil, with approximately 7000 km². The region harbors the headwaters of important Brazilian watersheds and a vegetation complex associated with ferruginous substrates (Jacobi et al., 2007; Ataíde et al., 2011; Fernandes, 2016a; Schaefer et al., 2016). The ferruginous rupestrian grassland, also known as canga, comprises a huge plant species diversity and high rates of endemism (Viana and Filgueiras, 2008; Fernandes, 2016b). Regional mining activity strongly impacts biodiversity and ecosystem services (e.g., recharge and water regulation, pollinators, natural pest controllers, etc.), requiring restoration actions in a large number of degraded areas (Fernandes and Ribeiro, 2017). Currently, 44.09 % of Minas Gerais State (258610.7 km²) is under concession to mining companies (Rezende, 2016; Pena et al., 2017). The soil structure of ferruginous rupestrian grasslands determines peculiar thermal and water behavior, which favors the establishment of highly specialized vegetation, extremely sensitive to the loss of soil cover (Jacobi et al., 2007; Fernandes, 2016a,b). The environmental uniqueness, high diversity, low resilience and accelerated degradation of this ecosystem have created a conflicting context and an immediate challenge for its conservation and restoration (Fernandes et al., 2016b).

In this study, we experimentally tested the application of two different topsoil depths (0.20 and 0.40 m) in a degraded area at the Iron Quadrangle. We assessed the natural vegetation recovery and the survival of transplanted seedlings of native species obtained from rescue operations in both topsoil depths.

MATERIALS AND METHODS

Study area

The experiment was settled up in January 2009 at the mine Capão Xavier (CPX) - Vale, located in Iron Quadrangle, south portion of Espinhaço Range, southeastern Brazil.
(20° 2’ 59” S, 43° 58’ 43” W). The climate is Cwa, according to the Köppen classification system, with rainy hot summer and dry winters. The warmer month exceeds 22 °C and the coldest month is below 18 °C in average temperature (Reboita, 2015).

**Topsoil**

The experimental area was capped with the topsoil from a donor site in the same mine. The topsoil was mainly composed of ferruginous concretions and the following granulometry: 44 % coarse sand, 28 % clay, 19 % silt and 9 % fine sand. The topsoil was removed from the excavation area by surface scraping (0.00-0.40 m) with a crawler tractor. After transporting to the experimental area, the material was scattered using loader and backhoe. The final thickness adjustment of the topsoil was performed manually. It was systematically established four 200 m² (20 × 10 m) plots with 0.20 m topsoil and four with 0.40 m topsoil. The plots were 2 m apart from each other. Each plot was subdivided into four subplots (5 × 10 m) to receive the native species planting, as specified in the following subsection.

Small soil fertilization was carried out aiming at the basic replacement of macronutrients. A mixture of the following nutrients was prepared and manually incorporated into the topsoil in all plots: magnesium thermophosphate (soluble P₂O₅, citric acid 2 %, Ca 20 %, Mg 7 %, B 0.10 %, Zn 0.55 %, Mn 0.12 %, Cu 0.05 %, and Mo 0.006 %), magnesium sulfate (Mg 9 % and S 12 %), and NPK 20-0-8.

**Species selection and planting**

The species *Arthrocereus glaziovii* (K.Schum.) N.P. Taylor & Zappi, *Sinningia rupicola* (Mart.) Wiehler, *Anthurium minarum* Sakur. & Mayo, *Hoffmannseggia crispat"a* (Thunb.) H.G.Jones, *Billbergia elegans* Mart. ex Schult. & Schult.f., *Epidendrum secundum* Jacq., *Pleroma heteromallum* (D. Don) D.Don, *Vellozia compacta* Mart. ex Schult. & Schult.f., *Vellozia caruncularis* Mart. ex Seub., *Vellozia graminea* Pohl, *Vriesea minarum* L.B.Sm., *Clusia arrudea* Planch. & Triana ex Engl., *Dyckia consimilis* Mez, *Paliavana sericiflora* Benth. and *Cupania sp.* L. were selected for the experimental planting, according to previous floristic survey in the surrounding area and to seedling availability in the mining company’s greenhouse (data not shown). These seedlings were obtained from rescue operations in the Capão Xavier mine area. On the rescue system adopted by the company, part of the rescued plants was planted in woody boxes (0.50 × 0.30 × 0.20 m dimension) and kept in the greenhouse for 6 to 12 months before planting. The boxes were taken to the experimental area and the seedlings were separated manually for planting in each subplot. Seedling size and age varied among species, once they were from rescue operations in the field. Nevertheless, the plants were visually selected to avoid plant size heterogeneity between experimental units when planting.

A total of 800 seedlings manually planted in shallow pits (0.10 × 0.10 × 0.10 m). Each subplot received 25 seedlings in the same spatial arrangement, totaling 100 seedling per plot, as following: *A. glaziovii* (N = 20), *S. rupicola* (N = 4), *A. minarum* (N = 8), *H. crispat"a* (N = 20), *B. elegans* (N = 4), *E. secundum* (N = 4), *P. heteromallum* (N = 4), *V. compacta* (N = 4), *V. caruncularis* (N = 4), *V. graminea* (N = 4), *V. minarum* (N = 8), *C. arrudea* (N = 4), *D. consimilis* (N = 4), *P. sericiflora* (N = 4), and *Cupania sp.* (N = 4). The planting was carried out in the rainy season, between January 20 and February 5 of 2009. However, due to low rainfall after the planting, irrigation by water truck was needed, according to the company availability and logistics.

**Vegetation assessment**

After 49 months, planted seedling survival, and vegetation composition and cover were assessed in each topsoil depth treatment. Survival was calculated for each species as
the percentage of living individuals in relation to the initial number of planted individuals, at plot level. The vegetation cover was measured by a visual qualitative-quantitative estimative of five randomly placed quadrats (1 × 1 m) in each subplot. Vegetation composition and cover were assessed by the Relevé Method (Braun-Blanquet, 1932). The sampling was taken by five quadrats (1 × 1 m) placed 2 m apart from each other along a 10 m transect, in each subplot. The relative cover of each species found was visually estimated in each quadrat. Scientific names of the plant species were verified at The International Plant Names Index (IPNI, 2020) and Flora do Brasil 2020 databases.

**Statistical analyses**

T-test was applied to evaluate total species survival and total plant cover in function of topsoil depth. The Mann-Whitney test was performed to evaluate the survival of each species in function of topsoil depth. All analysis were performed using the R 3.4.0 statistical platform (R Development Core Team, 2020).

**RESULTS**

**Planted seedling survival**

Plant survival was not influenced by topsoil depth, whether considering the total plants or each species. Nevertheless, seven species showed 50% survival or more (Cupania sp., V. caruncularis, A. minarum, C. arrudea, B. elegans, D. consimilis, and P. heteromallum) in both topsoil depth treatments, while V. graminea and S. warmingii were not found in the study area after the 49 months (Table 1). The species V. caruncularis showed 100% survival in the 0.20 m topsoil treatment.

**Vegetation cover and composition**

After 49 months, the 0.40 m topsoil plots showed greater vegetation cover than the 0.20 m topsoil plots (t(30) = -4.38; p = 0.00013) (Figure 1). The non-planted species (plants that emerged from the topsoil and/or colonizers arrived from seed rain or secondary dispersal) that most contributed to the vegetation cover in the 0.20 m topsoil treatment

### Table 1. Mean survival rates (%) of native species 49 months after planting in the topsoil experiment in the mine Capão Xavier – Vale. Plant species were from flora rescue operations

| Species                     | N per plot | 0.20 m topsoil | 0.40 m topsoil |
|-----------------------------|------------|----------------|----------------|
| Anthurium minarum           | 8          | 56.25          | 62.50          |
| Arthrocereus glaziovii      | 20         | 31.25          | 35.00          |
| Bilbergia elegans           | 4          | 56.25          | 81.25          |
| Hoffmannseggella crispata   | 20         | 47.50          | 52.50          |
| Clusia arrudea              | 4          | 56.25          | 50.00          |
| Cupania sp.                 | 4          | 81.25          | 87.50          |
| Dickyia consimilis          | 4          | 56.25          | 50.00          |
| Epidendrum secundum         | 4          | 18.75          | 31.25          |
| Paliavana sericiflora       | 4          | 25.00          | 6.25           |
| Sinningia warmingii         | 4          | 0.00           | 0.00           |
| Pleroma heteromallum        | 4          | 75.00          | 62.50          |
| Vellozia caruncularis       | 4          | 100.00         | 75.00          |
| Vellozia compacta           | 4          | 62.50          | 43.75          |
| Vellozia graminea           | 4          | 0.00           | 0.00           |
| Vrisea minarum              | 8          | 46.87          | 28.12          |

N: number of individuals planted in each plot.
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was *Achyrocline satureioides* (Lam.) DC. with about 20 % relative cover, followed by *Andropogon bicornis* L., *Axonopus siccus* Kuhlm., and *Microstachys corniculata* (Vahl) Griseb (Table 2). On the other hand, the non-planted species that most contributed to the vegetation cover in the 0.40 m topsoil was the exotic *Melinis minutiflora* P.Beauv. with about 15 % relative cover, followed by *M. corniculata*, *Triumfetta semitrioloba* Jacq., and *A. satureioides* (Table 2).

A total of seven non-native species were present in the 0.20 m topsoil (*Bidens Pilosa* L., *Melinis minutiflora* P.Beauv., *Melinis repens* (Willd.) Zizka, *Crocosmia crocosmiiflora* (Lemoine) N.E.Br., *Oxalis corniculate* L., *Thelypteris dentata* (Forssk.) E.P.St.John and *Pilea microphylla* (L.) Liebm.), and four in the 0.40 m topsoil depth (*B. pilosa*, *M. minutiflora*, *Plantago major* L. and *T. dentata*). All these species are exotic from Brazil, except *T. dentata*, that is native from Brazil but not from rupestrian grassland; it is a forest species. The non-native species contributed with 7 % to the 0.20 m topsoil cover and 19.42 % to the 0.40 m topsoil cover (Figure 2).

A total of 58 species (51 native) were recorded in the 0.20 m topsoil plots, while 45 species (41 native) were recorded in the 0.40 m topsoil plots. Typical species from ferruginous rupestrian grassland (*Mimosa calodendron* Mart. ex Benth., *Stachytarpheta glabra* Cham., *Vellozia albiflora* Pohl, *Cryptanthus schwackeanus* Mez, *Lychnophora pinaster* Mart., and *Portulaca hirsutissima* Cambess.) were recorded in the natural recovery at both topsoil depth.

Among the 15 planted species, 11 were found in the 0.20 m topsoil plots after 49 months (*A. glaziovii*, *A. minarum*, *H. crispata*, *B. elegans*, *P. heteromallum*, *V. compacta*, *V. caruncularis*, *V. graminea*, *V. minarum*, *D. consimilis* and *P. sericiflora*); while 10 species were found in the 0.40 m topsoil plots (*A. minarum*, *H. crispata*, *B. elegans*, *E. secundum*, *P. heteromallum*, *V. compacta*, *V. caruncularis*, *V. minarum*, *D. consimilis* and *Cupania sp.*).

**DISCUSSION**

Floristic and ecological studies on both flora rescue and topsoil use are important to increase the knowledge of restoration practices and vegetation establishment, especially in threatened ecosystems such as the ferruginous rupestrian grassland. Our study showed...
Table 2. Relative coverage (%) of plant species present in the experimental area in the mine Capão Xavier - Vale, comprising two treatments of different topsoil depth, 49 months after topsoil application

| Species                              | 0.20 m topsoil | 0.40 m topsoil |
|--------------------------------------|----------------|----------------|
| Achyrocline satureioides             | 20.72          | 11.31          |
| Ageratum conyzoides                  | 0.08           |                |
| Ageratum fastigiatum                 | 1.43           | 3.55           |
| Alternanthera brasiliana             | 0.48           | 0.32           |
| Andropogon bicornis                  | 11.11          | 1.34           |
| Andropogon ingratu                   | 0.95           |                |
| Anturium minarum                     | 0.48           | 0.24           |
| Arthrocereus glaziovii               | 0.16           |                |
| Axonopus siccus                      | 10.94          | 8.21           |
| Baccharis dracunculifolia            | 0.71           | 0.55           |
| Bilbergia elegans                    | 0.32           | 0.32           |
| Bidens pilosa                        | 0.56           | 0.55           |
| Bulbostylis fimbriata                | 2.38           | 1.26           |
| Cecropia pachystachya                | 0.79           | 0.24           |
| Chamaesyce hyssopifolia              | 0.08           |                |
| Chromolaena laevigata                | 0.08           |                |
| Clusia arrudea                       | 0.16           |                |
| Conyza canadensis                    | 0.87           | 0.63           |
| Commelina erecta                     | 0.48           | 6.85           |
| Crocosmia crocosmiiflora             | 0.40           |                |
| Cryptanthus schwakeanus              | 0.32           | 0.08           |
| Cupania sp.                          | 0.39           |                |
| Cyperus sp                          | 0.16           |                |
| Dyckia consimilis                    | 0.32           | 0.24           |
| Emilia fosbergii                     | 2.46           | 0.16           |
| Epidendrum secundum                  | 0.24           |                |
| Eragrostis articulata                | 0.24           |                |
| Eriope macrostachya                  | 0.20           | 0.39           |
| Eragrostisspp.                       | 0.08           |                |
| Gnaphalium sp.                       | 0.48           |                |
| Hoffmannseggella crispata            | 1.03           | 0.08           |
| Ipomoea ramosissima                  | 0.47           |                |
| Lippia gracilis                      | 1.19           | 1.34           |
| Lychnophora pinaster                 | 0.24           | 0.08           |
| Marsypianthes chamaedrys             | 0.32           |                |
| Melinis minutiflora                  | 3.57           | 15.16          |
| Melinis repens                       | 0.32           |                |
| Mimosa calodendron                   | 0.32           | 2.45           |
| Myrtaceae indeterminada              | 1.51           |                |
| Oxalis corniculata                   | 0.16           |                |
| Panicum sellowii                     | 0.16           |                |
| Phylanthus rosellus                  | 0.64           |                |
| Pilea microphylla                    | 0.24           |                |

Continue
that the topsoil technique could support the recovery of native species and provide persistence of transplanted native species. Our findings suggest that 0.20 m topsoil may be enough to play this role. Results pointed a potential of, after four years from topsoil application, 57 % vegetation cover using 0.20 m topsoil, with 93 % contribution of 51 different native species; while 79 % vegetation cover using 0.40 m topsoil, with 80.6 % contribution of 41 native species.

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**Table 1.** Coverage of native and non-native species in each topsoil depth after 49 months of topsoil application in an experiment in the mine Capão Xavier - Vale to restore ferruginous rupestrian grassland.

| Species                      | Cover (%) |
|------------------------------|-----------|
| Plantago major               | 0.08      |
| Portulaca hirsutissima       | 0.08      |
| Polygala paniculata          | 0.95      |
| Microstachys corniculata     | 10.48     |
| Senna obtusifolia            | 0.40      |
| Setaria parviflora           | 0.16      |
| Sida glaziovii               | 4.92      |
| Sporobolus sp.               | 2.06      |
| Spermacoce verticillata      | 0.71      |
| Stachytarpheta glabra        | 1.83      |
| Tagetes sp.                  | 1.98      |
| Thelypteris dentata          | 0.32      |
| Pleroma heteromalum          | 3.65      |
| Triumfetta semitrioloba      | 2.78      |
| Trachypogon spicatus         | 0.32      |
| Vellozia albiflora           | 1.03      |
| Verbena litoralis            | 0.32      |
| Vellozia caruncularis        | 0.16      |
| Vellozia compacta            | 0.79      |
| Vellozia graminea            | 0.08      |
| Vernonanthura phosphorica    | 0.37      |
| Vrisea minarum               | 0.24      |

**Figure 2.** Coverage of native and non-native species in each topsoil depth after 49 months of topsoil application in an experiment in the mine Capão Xavier - Vale to restore ferruginous rupestrian grassland.
Although total vegetation cover was greater in 0.40 m topsoil than in 0.20 m topsoil, it was under substantial contribution of non-native species (almost 20 % relative cover). Moreover, the highly invasive African grass *M. minutiflora* presented the higher relative cover in the 0.40 m topsoil treatment, more than four times if compared with its cover in the 0.20 m topsoil treatment. As invasive plants have fast growth and high competitive ability, they create adverse microclimatic conditions for the native species establishment, hindering their development (Blossey, 1999; Hilário et al., 2011). Therefore, attention to their control is needed in ecological restoration practices. The management of invasive species is fundamental for the success of the restoration process started by the topsoil use, also requiring long-term monitoring (Blossey, 1999; Ferreira et al., 2015; Gomes et al., 2017).

A greater species richness was found in the 0.20 m topsoil treatment, which can be due to the germination ability of rupestrian grassland native species. The germination conditions and the substrate characteristics in the restoring areas provide information about plant establishment (Vázquez-Yanes and Orozco-Segovia, 1993). Temperature and light are important factors that control germination and seed dormancy, being crucial for plant growth regulation and development (Baskin and Baskin, 1988). Many rupestrian grassland species have small seeds with photodormancy (Abreu and Garcia, 2005; Nunes et al., 2016); thus germination should be lower in deeper soils.

The survival performance of some native species can be highlighted, such as *V. caruncularis*, *P. heteromallum*, *Cupania sp.*, and *V. compacta* (only in 0.20 m topsoil) with above 60 % mean survival rates 49 months after planting. Additionally, many saplings of *P. heteromallum* were found in the area, indicating its sexual propagation.

The recording of typical species of ferruginous rupestrian grassland, that were not introduced by planting (*M. calodendron*, *S. glabra*, *V. albiflora*, *C. schwackeanus*, *L. pinaster*, and *P. hirsutissima*), suggests that these species can form seed banks and/or propagate from the surrounding environment. The presence of these species reinforces the potential of the topsoil use for restoration in this ecosystem. They were able to emerge from and/or colonize the topsoil at both tested depths.

Among the non-planted species, including the exotic ones, 30 were also found by Mendonça (2013) in an adjacent area, which are: *Ditassa linearis* Mart., *Oxypetalum appendiculatum* Mart., *Baccharis dracunculifolia* DC., *L. pinaster*, *Bulbostylis capillaris* (L.) C.B.Clarke, *Bulbostylis fimbrillata* (Nees) C.B.Clarke, *Alchornea triplinervia* (Spreng.) Müll. Arg., *M. corniculata*, *M. calodendron*, *Eriope macrostachya* Mart. ex Benth., *S. warmangii*, *Myrcia splendens* (Sw.) DC., *Peperomia decora* Dahlst., *Microtea paniculata* Moq., *Andropogon ingradius* Hack., *A. siccus*, *Eragrostis rufescens* Schrad. ex Schult., *Eragrostis polytricha* Nees, *Eragrostis articulata* (Schrank) Nees, *Panicum sellowii* Nees, *Sporobolus metallicolus* Longhi-Wagner & Boechat, *Andropogon bicornis* L., *Setaria parviflora* (Poir.) Kerguélen, *Trachypogon spicatus* (L.f.) Kuntze, *P. hirsutissima*, *Portulaca mucronate* Link, *Calibrachoa elegans* (Miers) Stehmann & Semir, *Lantana fucata* Lindl., *Lippia gracilis* Schauer, and *S. glabra*. These data indicate the potential role of the topsoil as a source of quality propagules and/or as functional receptor of surrounding propagules for the establishment of native species.

**CONCLUSIONS**

Use of topsoil for ecological restoration in ferruginous rupestrian grasslands has shown to be a potentially effective technique, but the management of invasive species is extremely necessary and essential to be considered in the restoration planning. The 0.20 m topsoil showed lower coverage by exotic species compared to the thicker topsoil.

The native species *Vellozia caruncularis*, *Cuspania sp.*, and *Pleroma heteromallum* stood out for their performance in this experiment, being considered good candidates for planting
with restoration purposes in ferruginous rupestrian grasslands. Studies that monitor species establishment and performance in the topsoil at a long-time scale are needed to improve the knowledge of the technique's effectiveness and identify complementary restoration practices.

Overall results suggest that a topsoil depth of 0.20 m allows the establishment of a variety of native species, both transplanted from rescue operations and colonizing species (from topsoil propagule bank or surrounding dispersal). Native species cover and richness were greater in 0.20 m topsoil compared to 0.40 m topsoil.

**AUTHOR CONTRIBUTIONS**

**Conceptualization:** Lina Andrade Lobo Rezende (equal) and Luiz Eduardo Dias (lead).

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**Writing – review & editing:** Geraldo Wilson Fernandes (equal), Luiz Eduardo Dias (supporting) and Vanessa Matos Gomes (lead).

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