Hindawi
Applied and Environmental Soil Science
Volume 2019, Article ID 3075153, 8 pages
https://doi.org/10.1155/2019/3075153

Research Article

Lysinibacillus sphaericus as a Nutrient Enhancer during Fire-Impacted Soil Replantation

A. M. Aguirre-Monroy, J. C. Santana-Martínez, and J. Dussán

Centro de Investigaciones Microbiológicas (CIMIC), Departamento de Ciencias Biológicas, Universidad de Los Andes, Carrera 1 No. 18A-10 J-206, Bogotá, Colombia

Correspondence should be addressed to J. Dussán; jdussan@uniandes.edu.co

Received 19 October 2018; Revised 5 February 2019; Accepted 14 February 2019; Published 12 March 2019

Academic Editor: Claudio Cocozza

Over the past ten years, more than twenty fires have affected the El Noviciado estate located in Cerro Majuy, Colombia, leading to a loss of soil nutrients and infertility. *Lysinibacillus sphaericus*, a Gram-positive, mesophilic, and spore-forming bacterium, can be used in soil amendment in the replantation processes, given its ability to fix nitrogen, and nitrify, and solubilize phosphorus, increasing soil nutrients used for plant growth. In this study, we evaluated the soil-amendment potential of *L. sphaericus* by monitoring the nutrient content of a selected fragment of soil in the El Noviciado estate. For this purpose, we added a mixture of *L. sphaericus* OT4b.31, OT4b.49, CBAM5, III(3)7, and 2362 strains and determined the ammonium, nitrites, nitrates, phosphorus, and indole acetic acid concentrations in soil. *Alnus acuminata* sbsp. *acuminata*, a native model plant known for its restoration effect, was used for replantation. Results indicated that soils with added *L. sphaericus* presented significant differences in ammonium, nitrites, nitrates, phosphorus, and indole acetic acid concentrations when compared to control soils. Further, results showed no significant differences between soil that had been pre-inoculated in greenhouse and soil directly inoculated in field. We propose that *L. sphaericus* could be a good nutrient enhancer and plant growth promoter that can be used for the amendment of fire-impacted soils and replantation treatments.

1. Introduction

Wildfires significantly change soil properties and composition with physical effects such as altered pore size, water repellency, runoff response, and aggregates stability [1], and biological effects consisting of changes ranging from vegetation lost or removal to changes in biomass production and the composition of microbial communities [1, 2]. High temperatures reached during a long fire can decrease microbial biomass and, in extreme cases, lead to soil sterilization. Fires also alter microbial communities, stimulating the proliferation of microorganisms with resistant structures or those capable of growing in polluted environments containing combustion products, but inhibiting the communities without these traits [2, 3]. Chemical effects include changes in the availability of nutrients. Organic matter combustion leads to an increase in C, while some other nutrients such as N and P are volatilized in short-term due to high temperatures [3, 4]. However, processes of volatilization occur partially to the soil N and P [2]. Inorganic forms of these nutrients can also be generated during fires, resulting in a greater number of available compounds for plants, but it can be lost without plant uptake as a result of vegetation removal. Concerning N from organic matter, fires transform it into ammonium and nitrates, which can be lost by denitrification and leaching. During fires, orthophosphates, the only available P form for plants uptake, are primarily released from organic P pool in litter [5]. Nevertheless, high-intensity fires remove the litter by combustion, depleting the organic P source [6]. Additionally, orthophosphates can precipitate to slightly available mineral forms [2].

The effects of fire on the physical, chemical, and biological properties of soil are related to each other. For instance, the reduction of soil microbial biomass is closely related to the mid- and long-term decrease of nutrients given that soil fungi and bacteria are essential in N and P cycling [7]. Water repellency and runoff result in moisture decrease, leading to stressful conditions for bacteria and fungi on topsoil, which, in turn, cause a decrease in the metabolic rate of these organisms and consequently a loss of organic matter [8, 9]. Changes in soil pH lead to changes in soil microbial communities and their activity, for example, influencing fungal/bacteria ratio [10].

Bacteria whose activity affects soil chemical properties can be plant-growth promoting bacteria (PGPB). Part of the bacteria classified in this group are reported to play an important role in nitrogen and phosphorus cycling, since they have the ability to fix nitrogen, and nitrify, and solubilize phosphorus, increasing soil nutrients that plants use for their growth [11]. Studies reported the use of PGPB in soil restoration, showing an enhancement of soil nutrients when adding an inoculum of PGPB consortium in degraded soils during replantation with native trees [12] and the direct effect of these bacteria inoculum on plant growth [13].

Lysinibacillus sphaericus is a spore-forming, aerobic, and mesophilic Gram-positive bacterium. Previous studies reported the isolation of this bacterium from soil and water and its activity as a PGPB [11]. There are also reports of *L. sphaericus* being used to promote the production of phytohormones, such as indole acetic acid (IAA) [11], involving in the radicular plant growth and plant nutrient uptake [14]. Furthermore, the genomes of *L. sphaericus* strains OT4b.31, CBAM5, and III(3)7 contain annotated genes that are involved in nitrogen cycling [11, 15–17].

Along with the soil nutrient amendment with PGPB, impacted forests and soils are also reclaimed with replantation methods [12, 18]. An evaluation of tree species in a replantation process found establishment and utility to be the most determinant tree characteristics for successful reforestation [19]. Accordingly, *Alnus acuminata* sbsp. *acuminata* is a native tree distributed in the Americas, from Mexico to northern Argentina, which gives rise to the cloud forests present in the Andes given its ability to establish in dry, wet, and very wet areas. The importance of this tree is based on its restoring effect when reforesting, which consists of improved soil fertility, the formation of litter cover that increases the organic matter in soil, soil conservation, slopes stabilization, and nitrogen fixing capacity through the formation of symbiotic nodules. In addition, *A. acuminata* sbsp. *acuminata* exhibits fast growth and good adaptability to nutrient-poor soils [20].

In Colombia, the El Noviciado estate, located in the Majuy hills, is a natural reserve, preserving primary and secondary forests, which allow soil protection [21]. Temperatures in the region range from 5 to 14°C. A section of the estate is designated to native forest self-regeneration and replantation [21], given that it has been affected by over twenty fires over the last 10 years, with the most destructive in 2013. This wildfire caused the complete loss of 65 ha of forests and severe damage in terms of soil nutrients content and fertility [22]. Despite the fact that some PGPBs are reported as good enhancers of soil nutrients for replantation of damaged soils [7], there are no reports for the action of *L. sphaericus*, its effects on fire-impacted soil nutrients, and its role in replantation in growth temperatures lower than the optimum range reported (14–37°C) [15]. For these reasons, the aim of this study was to determine the effect of *L. sphaericus* on fire-impacted soil nutrients while replanting with *A. acuminata* sbsp. *acuminata* seedlings. For this purpose, a fragment of fire-impacted soil in the El Noviciado estate was selected for field trials, where the mean temperature is 13.5°C, ranging from 5 to 14°C [22]. Further, the nitrogen (ammonium, nitrates, and nitrites), phosphorus, and IAA contents were monitored after a mixture of *L. sphaericus* OT4b.31, 4OT4b.49, CBAM5, III(3)7, and 2362 strains were added.

2. Materials and Methods

2.1. Site Description. El Noviciado estate (4°50′19.7″N, 74°05′31.9″W) is located in the Cota municipality, Cundinamarca, Colombia. It is classified as cool savanna and is located at latitude 2550–2975 m.a.s.l. The mean precipitation for the area is of 800 mm^3 a year, and it has a mean temperature of 13.5°C, ranging from 5 to 14°C. The region has two rainy periods a year—one from April to May and the other from September to December—and two dry periods a year—one from January to February and the other from July to August. The estate covers 331 ha, and it is situated in an Andean bioclimatic floor, consisting of a characteristic hilly landscape and Andean forest vegetation. Soil in the estate is classified as half-dry, which consists of 57% clay and sandstone. It also has a high content of cinder and organic matter, but its fertility is very low [22]. The soil is slightly acidic with a moderate to high Al saturation, a moderate and low organic C content, and a low N, P, K, and Ca content. Soil exposes an average drainage with pronounced slopes and high susceptibility to erosion [22].

2.2. Field Seedlings Treatments. Four treatments were established for field trials (Table 1), each consisted of 15 four-month-old *Alnus acuminata* sbsp. *acuminata* seedlings. All individuals in the treatments were randomly distributed and transplanted in a fragment of 40 m × 25 m (length × width) of fire-impacted soil at the El Noviciado estate. Each seedling was watered biweekly in field with 400 mL. Thirty of the total seedlings were previously maintained in greenhouse conditions and watered in two-day intervals. In greenhouse, two preliminary treatments were established: 15 seedlings were watered with a mixture of five *L. sphaericus* strains (explained below) every six weeks during the four months of growth (GB) and 15 were maintained with no bacterial addition (G) as a negative control. Before transplantation, the other 30 seedlings, which were not submitted to greenhouse conditions, were assigned to two different treatments (B) and its respective negative control (C). Treatments GB and B were watered with a mixture of five *L. sphaericus* strains at week zero and week six. Week zero corresponded to the transplanting date.

2.3. L. sphaericus Strains Inoculum. Five *L. sphaericus* strains previously reported as PGPB [11] were used in this study:

Table 1: Field trial treatments with their respective conditions. Greenhouse (G) indicates seedlings were maintained previously in green house conditions. Bacterial inoculum (B) indicates the mixture of the 5 *L. sphaericus* strains was added to seedlings.

Treatments	Greenhouse (G)	Bacterial inoculum (B)
GB	✓	✓
G	✓	X
B	X	✓
C	X	X

OT4b.31, OT4b.49, III(3)7, CBAM5, and 2362. Strains OT4b.31 and OT4b.49 were originally isolated in Colombia from coleopteran larvae. Strain III(3)7 was originally isolated in Colombia from oak forest soil. Strain CBAM5 was originally isolated from subsurface soil of oil well explorations in Colombia's Eastern Plains. All four strains OT4b.31, OT4b.49, III(3)7, and CBAM were isolated by CIMIC and belong to its culture collection. Finally, the WHO reference strain 2362 originally isolated from adult *Simulium damnosum* (Diptera: Simuliidae) in Nigeria was acquired from the Pasteur Institute.

For the initial activation, each of the five strains was grown in nutrient broth (Oxoid) at 30°C for 16 h in order to obtain an overnight (ON) liquid culture. Following this, the overnight cultures were transferred into 6 autoclaved trays containing 500 mL of SPC agar and were incubated for 24 h at 30°C. The bacterial inoculum of each strain was collected from the trays, resuspended on a 0.85% saline solution, and stored at 4°C, until they were added to the field trials. In the estate, the five strains inocula were combined and diluted in a 16 L water pail, and then 400 mL were used to water each of the 30 individuals of the GB and B treatments. The inoculum was added twice during the entire study: once at week zero right after sample collection, and once at week six. Final concentrations of the strains OT4b.31, OT4b.49, III(3)7, CBAM5, and 2362 in water were $\sim10^7$, $\sim10^8$, $\sim10^9$, $\sim10^8$, and $\sim10^8$, respectively.

2.4. Nutrients and Bacterial Titer Determination. Nine soil samples were collected per treatment every third week, beginning in week zero to week 12. Each sample consisted of ~40 g of soil collected at 15 cm depth from the bottom of seedlings. Further, soil samples in the treatments were randomly grouped into three and combined to simplify the measurements.

The concentrations of ammonium, nitrites, nitrates, and phosphorus in the three combined soil samples per treatment were determined using commercial Merck Test kits refs. 114752 (0.010–3.00 mg/L NH_4), 1.14776.0002 (0.07–3.28 mg/L NO_2), 109713 (0.4–110.7 mg/L NO_3), and 114848 (0.0057–11.46 mg/L PO_4), respectively.

Indole acetic acid concentrations in the combined soil samples were measured following the protocol suggested by Sarwar et al. for the colorimetric determination of auxins [14], but tryptophan addition was skipped, since *L. sphaericus* presents a tryptophan operon and the five strains used were reported to produce IAA *in vitro,* without the addition of tryptophan to the medium [23]. Finally, cultivable bacterial titer was also determined for each combined sample using the plating method on SPC agar [24].

2.5. Statistical Analyses. R v3.1.1 [25] and Rstudio v 1.1.383 [26] software were used for statistical analyses. The Shapiro–Wilcoxon test [27] was used to evaluate the data for normality. Ammonium, nitrites, nitrates, phosphorus, and IAA concentrations were analyzed using an analysis of variance (ANOVA) followed by a Tukey–Kramer test to determine significant differences between averages between the treatments. Cases where data were not normally distributed, a Kruskal–Wallis test followed by a Nemenyi test was used to determine significant differences between averages between treatments. A principal components analysis (PCA) was used as a multivariate analysis to elucidate the relationships between nutrient concentrations, time, indole acetic acid production, and cultivable bacterial titers.

3. Results and Discussion

3.1. Free Phosphorus Concentration and L. sphaericus Growth. Comparisons between free phosphorus concentrations in all four treatments showed significant differences at weeks three, six, nine, and 12 (Figure 1; ANOVAs: $F_{W3} = 30.88$, $P_{W3} < 0.0001$, $F_{W6} = 10.71$, $P_{W6} = 0.0036$, $F_{W9} = 49.68$, $P_{W9} < 0.0001$, and $F_{W12} = 4.906$, $P_{W12} = 0.0321$, respectively). Regarding GB, free phosphorus concentrations did not show significant differences through week zero to week 12 (Figure 1; *t*-test: $P = 0.981$) or when comparing them to control G at the end of the study (week 12) (Figure 1). However, at week nine, a significant difference was found between GB and G treatments, and a decreasing tendency was observed in the G treatment (Figure 1). Considering B, free phosphorus concentrations showed a rising tendency contrary to control C, which decreased through time (Figure 1), and a significant difference was found between both treatments at weeks three, six, nine, and 12. Also, significant differences were found between week zero and week 12 in the B treatment (Figure 1; *t*-test: $P = 0.02917$). Even though treatments GB and B both had PGPB added, only the soil without a previous treatment showed significant differences in free phosphorus concentrations from week zero to week 12. This was probably due to the GB treatment already having slightly high concentrations of the nutrient by week zero as a result of *L. sphaericus* activity on soil during the four months in greenhouse, as it was inoculated before the transplanting date.

As bacterial count in treatments GB and B increased throughout the whole study, from week zero to week 12 (Figure 2), we suggest that there might be a relationship between the action of the bacteria and the free phosphorus concentrations in these treatments, probably due to the role of *L. sphaericus* in solubilizing this nutrient [28, 29]. Considering that phosphorus is an essential macronutrient that determines plant growth [30], the role of microorganisms in this nutrient cycling is crucial, and it can be fundamental in a replantation process in fire-impacted soil. Phosphorus in soil is mostly found in an insoluble form or associated with Fe,

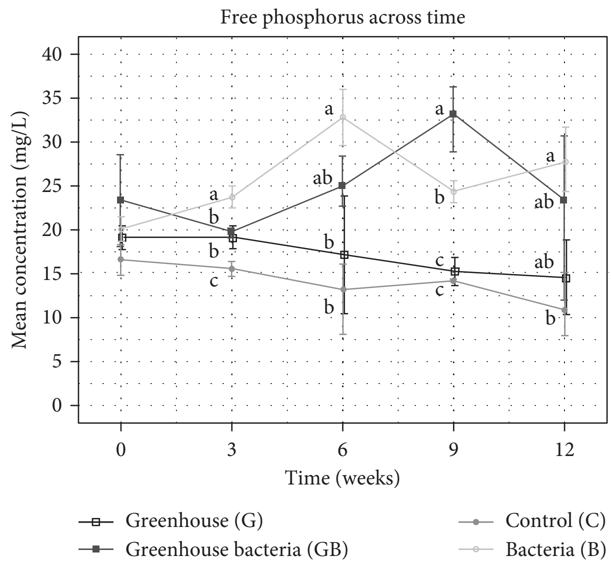

Figure 1: Free phosphorus concentrations per treatment across time. Means within a time measured followed by the same letter are not significantly different according to the Tukey–Kramer test or Nemenyi test in case of no normality.

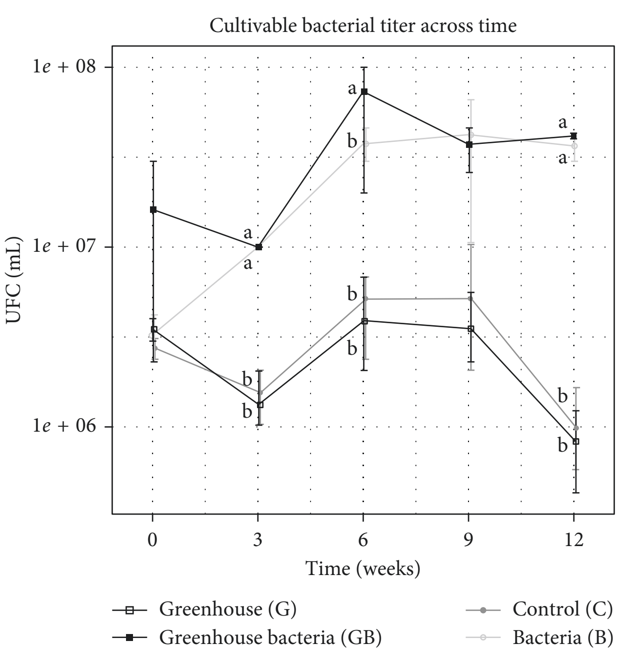

Figure 2: Cultivable bacterial titers per treatment across time. Means within a time measured followed by the same letter are not significantly different according to the Tukey–Kramer test or Nemenyi test in case of no normality.

Al, and Mn oxides, which are difficult to assimilate by plants [30]. The role of PGPB in this nutrient cycling consists of their capacity to synthesize organic acids, so that the consequent soil acidification can lead to phosphorus release from mineral phosphate, turning it into soluble and available forms for plants uptake [30, 31]. Studies reported *L. sphaericus* solubilization activity in silicate medium agar and biochar, supporting our results [28, 32]. Presumably, *L. sphaericus* c-binding capacity followed by the secretion of organic acids and phosphatases is the mode of action that allows the solubilization of phosphorus even when undergoing pH changes [28, 32]. Our results, together with these reports, indicate the potential for the use of this microorganism in fire-impacted soil amendment in replantation processes, due to its capacity to release phosphorus from biochar.

Additionally, in both treatments, B and GB, an oscillating pattern was observed for free phosphorus concentrations, similar to the one described by Rodríguez and Fraga [30], where phosphorus levels rise and fall. They propose an explanation that after an increase of the nutrient, the consequent decrease in phosphorus concentrations is the result of uptake of the nutrient as a source by plants and soil microorganisms [30, 33].

Cultivable bacterial titer across time showed significant differences between treatments in weeks three, six, and 12 (Figure 2; ANOVA: $F_{W3} = 505.3$, $P_{W3} < 0.0001$, $F_{W6} = 5.874$, $P_{W6} < 0.0202$, and $F_{W12} = 130.3$, $P_{W12} < 0.0001$). As expected, treatments with *L. sphaericus* presented higher bacterial titers than treatments with no bacteria added, which presumably had less microorganisms once environment was affected by wildfires. Significant differences were also found for treatments GB and B between week zero and week 12 (Figure 2; *t*-test: $P = 0.0009$ and $P = 0.0344$). These results together indicate that *L. sphaericus* might have a great potential to establish in soils impacted by wildfires, even in lower growth temperatures than the optimum range reported [15], such as the one previously mentioned for the El Noviciado estate (ranging from 5 to 14°C) [22]. As a result, the increased bacterial titer can indicate a more active bacterial metabolism and a consequent increase in soil nutrients.

3.2. Ammonium, Nitrite, and Nitrate Concentration and L. Sphaericus Growth. Results of soil ammonium concentrations showed significant differences in weeks three, nine, and 12 (Figure 3; ANOVA: $F_{W3} = 10.44$, $P_{W3} = 0.0039$, $F_{W9} = 20.68$, $P_{W9} = 0.0004$, and $F_{W12} = 43.46$, $P_{W12} < 0.0001$). Despite the fact that ammonium concentrations tend to increase in GB and B treatments, no significant differences were found between week zero and week 12 (Figure 3; *t*-test: $P = 0.2146$ and $P = 0.0892$, respectively). However, in contrast to these treatments, C and G controls showed a decreasing tendency. Nitrification or denitrification processes might influence diminishing soil ammonium concentration [34], yet results exhibited that PGPB can help stabilize soil ammonium levels or even increase them when compared to soil with no bacteria, as shown in our results and in a previous study [11].

With regard to nitrite concentrations, significant differences were observed in weeks three, nine, and 12 (Figure 4; ANOVA: $F_{W3} = 53.49$, $P_{W3} \leq 0.0001$, $F_{W9} = 13.70$, $P_{W9} = 0.0016$, and $F_{W12} = 10.59$, $P_{W12} < 0.0037$). Similarly to ammonium, nitrite concentrations in the GB and B treatments tended to increase across time; however, results between week zero and week 12 did not show significant differences (Figure 4; *t*-test: $P = 0.2616$ and $P = 0.2588$,

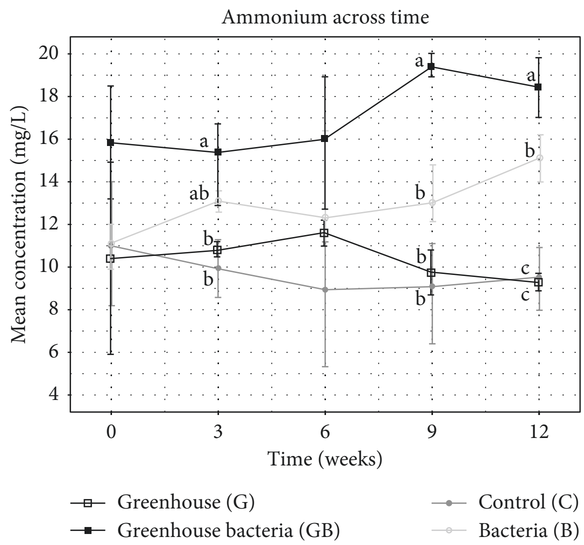

Figure 3: Ammonium concentrations per treatment across time. Means within a time measured followed by the same letter are not significantly different according to the Tukey–Kramer test or Nemenyi test in case of no normality.

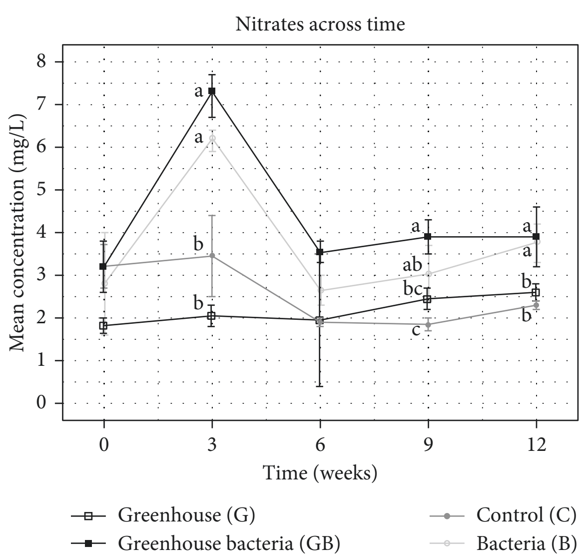

Figure 4: Nitrite concentrations per treatment across time. Means within a time measured followed by the same letter are not significantly different according to the Tukey–Kramer test or Nemenyi test in case of no normality.

respectively). Nitrites are the most unstable nitrogen form, and once they are present in soil, they are rapidly transformed into other more stable compounds [35].

Nitrate concentrations exhibited significant differences in weeks nine and 12 (Figure 5; ANOVA: $F_{W9} = 16.21$, $P_{W9} = 0.0009$, and $F_{W12} = 14.37$, $P_{W12} < 0.0013$), suggesting the nitrifying activity of the mixture of the 5 *L. sphaericus* strains used. In this case, nitrate concentrations presented in treatment B were significantly different between weeks zero and 12 (Figure 5; *t*-test: $P = 0.0411$), but, once again, in treatment GB they were not (Figure 5; *t*-test: $P = 0.1817$).

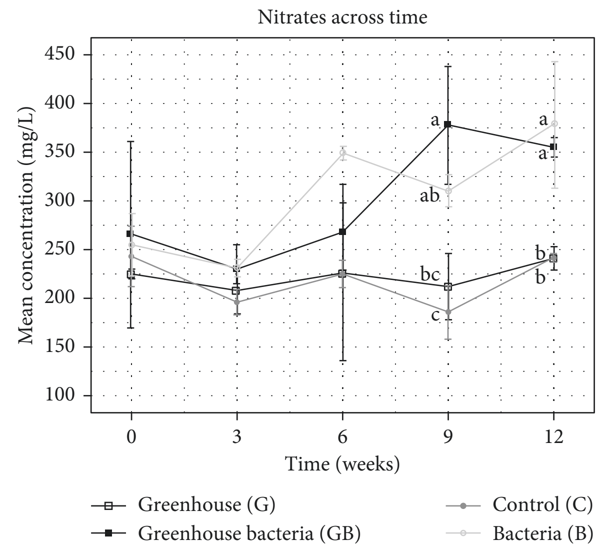

Figure 5: Nitrate concentrations per treatment across time. Means within a time measured followed by the same letter are not significantly different according to the Tukey–Kramer test or Nemenyi test in case of no normality.

The GB treatment was previously inoculated, and bacterial activity was probably already evident by week zero.

A relationship between the ammonium, nitrite, and nitrate concentrations was found in cases where *L. sphaericus* was added. By week three nitrite concentrations in GB and B treatments were higher than ammonium and nitrate concentrations, respectively, to their corresponding controls (Figures 3–5). In this sense, when nitrogen is fixed in the form of ammonium, the nitrifying bacteria in soil turn ammonium into nitrites, but they are rapidly converted into nitrates that can then be taken up by the seedlings [11]. Oscillations in all nitrogen-form concentrations (ammonium, nitrite, and nitrate) in treatments GB and B reflect the bacterial activity and their role in nitrogen cycling [36, 37], which is consistent with the bacterial titers found across time (Figure 2). Moreover, these results are supported by a previous study, where it was shown that while nitrogen is fixed as ammonium in the soil, nitrates are available for plant uptake ultimately, thanks to bacterial metabolism [11]. We propose that PGPB are essential in replantation programs because they provide the only means by which to transform N sources into available forms for plant uptake.

3.3. Indole Acetic Acid Production. Given that *L. sphaericus* presents a tryptophan operon, this bacterium has the ability to synthesize tryptophan amino acid [11, 23, 38], which is one of the precursors to indole acetic acid [39, 40]. In this sense, results showed that indole acetic acid could be determined directly from soil samples without the addition of tryptophan. In our study, indole acetic acid showed significant differences by weeks nine and 12 in the treatments where the mixture of the 5 *L. sphaericus* strains was added (Figure 6; Kruskal–Wallis: $H_{W9} = 10.38$, $P_{W9} = 0.0156$; ANOVA: $F_{W9} = 8.876$, $P_{W9} = 0.0063$), in contrast to control soils, suggesting a relationship with the presence of the bacteria. This result

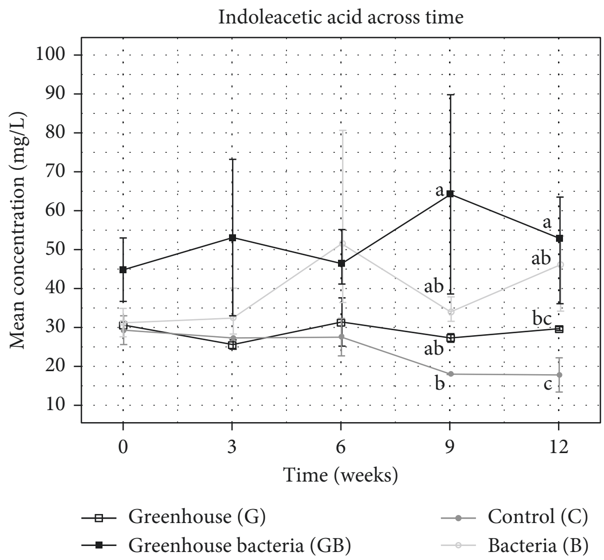

Figure 6: Indole acetic acid per treatment across time. Means within a time measured followed by the same letter are not significantly different according to the Tukey–Kramer test or Nemenyi test in case of no normality.

indicated the bacterial ability to promote the plants' auxins production and its own capacity of producing IAA [11, 14]. There is also an oscillatory pattern in the auxin concentrations observed over time, suggesting that while IAA is produced by the plant and the bacteria, it is also used for root growth, allowing the roots of the seedlings to expand and take nutrients from the soil more efficiently [41, 42]. Thus, *L. sphaericus* plays a crucial role in fire-impacted soil replantation processes, as it can enhance nutrients in soil and improve seedlings' nutrient uptake capacity through auxin production and its IAA production-promoting activity.

3.4. Principal Components Analysis. The analysis showed two noticeable clusters of dots, differentiated by treatments with and without the addition of *L. sphaericus* addition, separated horizontally from left to right (Figure 7). Cultivable bacterial titers; time; ammonium, nitrite, nitrate, and phosphorus concentrations; and indole acetic acid production all have a significant effect on the clustering distribution, with less influence from time and nitrites (Figure 7). We suggest that the distribution shown indicates the influence of bacteria in soil amendment regarding all variables. Thus, the addition of *L. sphaericus* to GB and B treatments enhanced the availability of nutrients and the production of auxins. As such, we consider *L. sphaericus* to be a good candidate for use in reforestation or replantation processes.

4. Conclusions

The importance of this study is related to the need to preserve the natural reserve in the El Noviciado estate, due to the role it plays in the reduction of factors that cause the degradation of biodiversity and ecosystem characteristics. We consider decisive the contribution of native vegetation replantation processes to revert the destructive processes

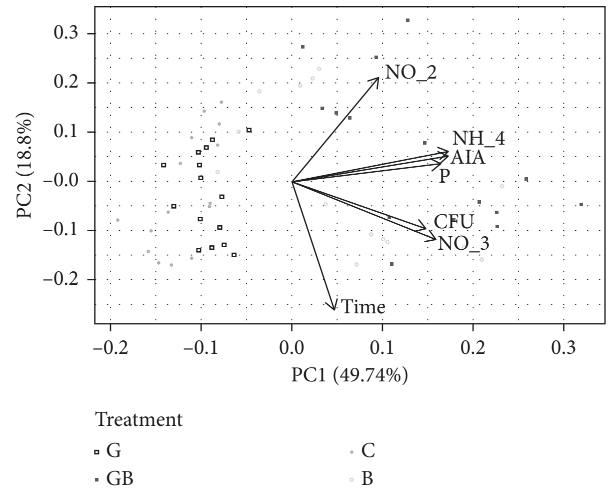

Figure 7: Autoplot showing the results of the principal components analysis (PCA) to illustrate the relationships between treatments and soil determinations. The length of the vectors indicates the influence of the parameter in the ordination of the data.

and improve environmental conditions for preventing forest fires. This study is the first report of *L. sphaericus* used in fire-impacted soils in lower than previously reported growth temperatures. We propose that *L. sphaericus* can grow and have an active metabolism at a temperature range of 5 to 14°C, reported as the temperature range for the study site [22]. Given the fact that our study showed a significant difference in the free phosphorus, nitrogen, and indole acetic acid contents in soil when the bacterium was added, we suggest that *L. sphaericus* could be a good nutrient enhancer and plant growth promoter that can be used for fire-impacted soil amendment and replantation purposes. Finally, we successfully donated and transplant 60 *Alnus acuminata* sbsp. *acuminata* seedlings for the reforestation of a fire-impacted fragment of soil in the El Noviciado estate.

Data Availability

The obtained data used to support the findings of this study are included within the supplementary information file.

Conflicts of Interest

The authors declare no conflicts of interest.

Acknowledgments

The authors are grateful to the School of Sciences at Universidad de los Andes, Colombia, for funding this study. Special thanks go to the staff at El Noviciado for their support in the fieldwork and to A. Delecluse from the Pasteur Institute for kindly donating the *L. sphaericus* WHO reference strain. The authors also thank Tiziana Laudato, native speaker and professional in scientific writing, for the English grammar corrections provided. This project was funded by the School of Sciences at Universidad de los Andes, Colombia.

Supplementary Materials

Supplementary Table 1 contains the collected raw data used to support the analysis of this study. (*Supplementary Materials*)

References

[1] S. H. Doerr and A. Cerdà, "Fire effects on soil system functioning: new insights and future challenges," *International Journal of Wildland Fire*, vol. 14, no. 4, pp. 339–342, 2005.

[2] G. Certini, "Effects of fire on properties of forest soils: a review," *Oecologia*, vol. 143, no. 1, pp. 1–10, 2005.

[3] L. F. DeBano, "The effect of fire on soil properties: proceedings—management and productivity of Western Montane forest soils," General Technical Report INT-GTR-280, US Department of Agriculture, Forest Service, Intermountain Research Station, Ogden, UT, USA, 1991.

[4] M. Francos, X. Úbeda, P. Pereira, and M. Alcañiz, "Long-term impact of wildfire on soils exposed to different fire severities: a case study in Cadiretes massif (NE Iberian Peninsula)," *Science of the Total Environment*, vol. 615, pp. 664–671, 2018.

[5] B. J. Cade-Menun, S. M. Berch, C. M. Preston, and L. M. Lavkulich, "Phosphorus forms and related soil chemistry of podzolic soils on Northern Vancouver island. II: the effects of clear-cutting and burning," *Canadian Journal of Forest Research*, vol. 30, no. 11, pp. 1726–1741, 2000.

[6] H. Knicker, "How does fire affect the nature and stability of soil organic nitrogen and carbon? A review," *Biogeochemistry*, vol. 85, no. 1, pp. 91–118, 2007.

[7] P. Radhapriya, A. Ramachandran, R. Anandham, and S. Mahalingam, "*Pseudomonas aeruginosa* RRALC3 enhances the biomass, nutrient and carbon contents of *Pongamia pinnata* seedlings in degraded forest soil," *PLoS One*, vol. 10, no. 10, Article ID e0139881, 2015.

[8] L. F. DeBano, S. M. Savage, and D. A. Hamilton, "The transfer of heat and hydrophobic substances during burning," *Soil Science Society of America Journal*, vol. 40, pp. 779–782, 1976.

[9] S. Manzoni, J. P. Schimel, and A. Porporato, "Responses of soil microbial communities to water stress: results from a meta-analysis," *Ecology*, vol. 93, no. 4, pp. 930–938, 2012.

[10] E. V. Blagodatskaya and T. H. Anderson, "Interactive effects of pH and substrate quality on the fungal-to-bacterial ratio and qCO_2 of microbial communities in forest soils," *Soil Biology and Biochemistry*, vol. 30, no. 10-11, pp. 1269–1274, 1998.

[11] S. A. Martínez and J. Dussán, "*Lysinibacillus sphaericus* plant growth promoter bacteria and lead phytoremediation enhancer with *Canavalia ensiformis*," *Environmental Progress & Sustainable Energy*, vol. 37, no. 1, pp. 276–282, 2017.

[12] A. Ramachandran and P. Radhapriya, "Restoration of degraded soil in the Nanmangalam reserve forest with native tree species: effect of indigenous plant growth-promoting bacteria," *Scientific World Journal*, vol. 2016, Article ID 5465841, 9 pages, 2016.

[13] Y. Bashan, B. G. Salazar, M. Moreno, B. R. Lopez, and R. G. Linderman, "Restoration of eroded soil in the Sonoran desert with native leguminous trees using plant growth-promoting microorganisms and limited amounts of compost and water," *Journal of Environmental Management*, vol. 102, pp. 26–36, 2012.

[14] M. Sarwar, M. Arshad, D. A. Martens, and W. T. Frankenberger, "Tryptophan-dependent biosynthesis of auxins in soil," *Plant and Soil*, vol. 147, no. 2, pp. 207–215, 1992.

[15] T. D. Peña-Montenegro and J. Dussán, "Genome sequence and description of the heavy metal tolerant bacterium *Lysinibacillus sphaericus* strain OT4b. 31," *Standards in Genomic Sciences*, vol. 9, no. 1, p. 42, 2013.

[16] T. D. Peña-Montenegro, L. Lozano, and J. Dussán, "Genome sequence and description of the mosquitocidal and heavy metal tolerant strain *Lysinibacillus sphaericus* CBAM5," *Standards in Genomic Sciences*, vol. 10, no. 1, p. 2, 2015.

[17] A. Rey, L. Silva-Quintero, and J. Dussán, "Complete genome sequencing and comparative genomic analysis of functionally diverse *Lysinibacillus sphaericus* III(3)7," *Genomics Data*, vol. 9, pp. 78–86, 2016.

[18] N. R. Sousa, A. R. Franco, M. A. Ramos, R. S. Oliveira, and P. M. Castro, "Reforestation of burned stands: the effect of ectomycorrhizal fungi on *Pinus pinaster* establishment," *Soil Biology and Biochemistry*, vol. 43, no. 10, pp. 2115–2120, 2011.

[19] M. A. McDonald, A. Hofny-Collins, J. R. Healey, and T. C. R. Goodland, "Evaluation of trees indigenous to the Montane forest of the blue mountains, Jamaica for reforestation and agroforestry," *Forest Ecology and Management*, vol. 175, no. 1–3, pp. 379–401, 2003.

[20] C. Ospina, R. Hernández, D. Gómez et al., *Guías Silviculturales Para El Manejo De Especies Forestales Con Miras A La Producción De Madera En La Zona Andina Colombiana: El Aliso O Cerez*, Vol. 37, CENICAFE-Federación Nacional de Cafeteros. Manizales, Chinchinám, Caldas, Colombia, 2005.

[21] Cideter SAS, Revisión general PBOT Cota, documento de diagnóstico, Municipio de Cota, 2014, http://www.cota-cundinamarca.gov.co/Ciudadanos/Notificaciones_Terceros/Documento%20Diagn%C3%B3stico.pdf.

[22] F. Econat and C. de Bogotá, *Plan de Manejo Ambiental (PMA)*, Universidad de Los Andes, Bogota, Colombia, 2013.

[23] K. Srisucharitpanit, M. Yao, B. Promdonkoy, S. Chimnaronk, I. Tanaka, and P. Boonserm, "Crystal structure of BinB: a receptor binding component of the binary toxin from *Lysinibacillus sphaericus*," *Proteins: Structure, Function, and Bioinformatics*, vol. 82, no. 10, pp. 2703–2712, 2014.

[24] J. Lederberg and E. M. Lederberg, "Replica plating and indirect selection of bacterial mutants," *Journal of Bacteriology*, vol. 63, no. 3, pp. 399–406, 1952.

[25] R Core Team, "R: a language and environment for statistical computing," February 2017, http://www.Rproject.org.

[26] RStudio Team, "RStudio: integrated development for R," RStudio, February 2017, http://www.rstudio.com/.

[27] J. H. Zar, *Biostatistical Analysis*, Prentice-Hall, Upper Saddle River, NJ, USA, 4th edition, 1999.

[28] H. He, T. T. Qian, W. J. Liu, H. Jiang, and H. Q. Yu, "Biological and chemical phosphorus solubilization from pyrolytical biochar in aqueous solution," *Chemosphere*, vol. 113, pp. 175–181, 2014.

[29] C. Gómez-Garzón, A. Hernández-Santana, and J. Dussán, "Comparative genomics reveals *Lysinibacillus sphaericus* group comprises a novel species," *BMC Genomics*, vol. 17, no. 1, p. 709, 2016.

[30] H. Rodríguez and R. Fraga, "Phosphate solubilizing bacteria and their role in plant growth promotion," *Biotechnology Advances*, vol. 17, no. 4-5, pp. 319–339, 1999.

[31] A. K. Halder, A. K. Mishra, P. Bhattacharyya, and P. K. Chakrabartty, "Solubilization of rock phosphate by *Rhizobium* and *Bradyrhizobium*," *Journal of General and Applied Microbiology*, vol. 36, no. 2, pp. 81–92, 1990.

[32] Z. Naureen, N. U. Rehman, H. Hussain et al., “Exploring the potentials of *Lysinibacillus sphaericus* ZA9 for plant growth promotion and biocontrol activities against phytopathogenic fungi,” *Frontiers in Microbiology*, vol. 8, p. 1477, 2017.

[33] P. Illmer and F. Schinner, “Solubilization of inorganic phosphates by microorganisms isolated from forest soils,” *Soil Biology and Biochemistry*, vol. 24, no. 4, pp. 389–395, 1992.

[34] T. H. DeLuca, M. D. MacKenzie, M. J. Gundale, and W. E. Holben, “Wildfire-produced charcoal directly influences nitrogen cycling in ponderosa pine forests,” *Soil Science Society of America Journal*, vol. 70, no. 2, pp. 448–453, 2006.

[35] K. H. Stern, “High temperature properties and decomposition of inorganic salts part 3, nitrates and nitrites,” *Journal of Physical and Chemical Reference Data*, vol. 1, no. 3, pp. 747–772, 1972.

[36] E. M. Souza, L. S. Chubatsu, L. F. Huergo et al., “Use of nitrogen-fixing bacteria to improve agricultural productivity,” *BMC Proceedings*, vol. 8, no. 4, p. O23, 2014.

[37] L. Spolaor, L. S. A. Gonçalves, O. J. A. P. D. Santos et al., “Plant growth-promoting bacteria associated with nitrogen fertilization at topdressing in popcorn agronomic performance,” *Bragantia*, vol. 75, no. 1, pp. 33–40, 2016.

[38] C. Gómez-Garzón, A. Hernández-Santana, and J. Dussán, “A genome-scale metabolic reconstruction of *Lysinibacillus sphaericus* unveils unexploited biotechnological potentials,” *PLoS One*, vol. 12, no. 6, Article ID e0179666, 2017.

[39] R. T. Vendan, Y. J. Yu, S. H. Lee, and Y. H. Rhee, “Diversity of endophytic bacteria in ginseng and their potential for plant growth promotion,” *Journal of Microbiology*, vol. 48, no. 5, pp. 559–565, 2010.

[40] T. Kunthic, B. Promdonkoy, T. Srikhirin, and P. Boonserm, “Essential role of tryptophan residues in toxicity of binary toxin from *Bacillus sphaericus*,” *BMB Reports*, vol. 44, no. 10, pp. 674–679, 2011.

[41] T. M. Tien, M. H. Gaskins, and D. H. Hubbell, “Plant growth substances produced by *Azospirillum brasilense* and their effect on the growth of pearl millet (*Pennisetum americanum* L.),” *Applied and Environmental Microbiology*, vol. 37, no. 5, pp. 1016–1024, 1979.

[42] C. L. Patten and B. R. Glick, “Role of *Pseudomonas putida* indole acetic acid in development of the host plant root system,” *Applied and Environmental Microbiology*, vol. 68, no. 8, pp. 3795–3801, 2002.