Biological soil conditioning and microbial activation in landscape design using bioformulations based on rhizospheric microorganisms

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Abstract. In this study, the effectiveness of microbial bioformulations based on rhizospheric microorganisms was evaluated under urban landscaping conditions. The formulations, containing *Bacillus subtilis*, *Trichoderma harzianum*, and *Azotobacter chroococcum*, were applied to sandy loam soils with low organic matter and moderate acidity. The aim was to assess changes in soil microbial abundance, enzymatic activity, physical properties, and plant survival. A 90-day field experiment revealed significant improvements in total microbial count (+71.4%), urease activity (+44.3%), and dehydrogenase activity (+68.6%) in treated plots. Plant survival rate increased by over 34%, while root disease incidence decreased by 65%. Soil bulk density was reduced by 11.6%, and water retention capacity increased by 18.8% in bio-treated soils. These results demonstrate that microbial bioformulations promote biological soil conditioning and enhance plant resilience, providing a sustainable alternative to chemical amendments in landscape design. The findings confirm the ecological and agronomic potential of integrating microbial technologies into urban green infrastructure development.

1 Introduction

The rhizosphere is a dynamic interface between plant roots and soil that plays a critical role in regulating nutrient cycling, microbial activity, and plant health. In recent years, growing attention has been directed toward the development and application of microbial bioformulations as sustainable tools to enhance soil quality and landscape resilience. These bioinoculants, primarily composed of beneficial rhizobacteria and fungi, offer promising alternatives to chemical fertilizers, particularly in urban landscaping where soil degradation

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and abiotic stresses are common. Numerous studies have shown that the application of microbial consortia in the rhizosphere significantly improves soil biological activity, enzymatic function, and plant productivity [1-2]. Rhizosphere microbiota not only enhances nutrient availability but also improves root architecture and provide protection against soil-borne pathogens [3]. Bioformulations containing strains such as *Bacillus subtilis*, *Trichoderma harzianum*, and *Azotobacter chroococcum* are particularly effective in stimulating microbial colonization and enzymatic processes in soils, as demonstrated in controlled experiments involving ornamental species in landscape design [4]. Microbial consortia can also regulate carbon and nitrogen transformations, increasing microbial efficiency and altering community structure toward beneficial taxa [1, 5]. These changes contribute to improved soil structure, increased water retention, and greater plant survival under urban stressors [6]. Recent ecological models suggest that microbial interactions—especially those involving protists, bacteria, and fungi—are essential drivers of rhizospheric diversity and plant health outcomes [7-8]. These microbial interactions have also been linked to the regulation of root exudates and enhancement of soil carbon pools [9]. In the context of landscape design, utilizing these microbial technologies offers a dual benefit—revitalization of degraded soils and the promotion of sustainable plant communities.

Scientific novelty: while previous studies have demonstrated the general benefits of microbial bioformulations in agricultural systems, there is a lack of data on their effectiveness under urban landscaping conditions with poor soil quality. This research provides new insights into the ecological role and effectiveness of specific microbial consortia in enhancing soil biological activity, structure, and plant performance in a temperate urban landscape context.

The aim of this study is to evaluate the impact of rhizospheric microbial bioformulations on soil biological parameters, plant vitality, and physical soil properties under conditions of urban landscaping. The focus is on determining the extent to which microbial inoculants improve soil enzymatic activity, microbial abundance, and plant health in ornamental landscape species.

2 Materials and Methods

The field experiment was conducted in 2023 on the territory of urban landscaping experimental plots located in the central region of the Russian Federation (55°45′N, 37°37′E), which is characterized by a temperate continental climate. The soil was classified as sandy loam, exhibiting moderate acidity (pH 5.7–6.2), low organic matter content (1.3–1.5%), and poor physical structure. The aim of the study was to assess the effectiveness of microbial bioformulations on soil biological activity and the vitality of ornamental plants commonly used in landscape design. The experimental design was based on a randomized block method with three replications per treatment. Each plot measured 4 × 4 m². Treatments involved the monthly application of microbial consortia comprising *Bacillus subtilis*, *Trichoderma harzianum*, and *Azotobacter chroococcum*, prepared in liquid form at a concentration of 5×10^7 CFU/g and applied by soil drenching at a rate of 2 L/m². Soil and plant samples were collected at three points: day 0 (before treatment), day 30, and day 90. Total microbial count was determined using the serial dilution method on nutrient agar. Soil enzymatic activities—urease and dehydrogenase—were measured colorimetrically following Alef and Nannipieri (1995) protocols. Plant survival rate was calculated as the ratio of surviving to initially planted individuals (%). Visual and microscopic phytopathological assessments were conducted to detect the presence of root pathogens such as Fusarium spp. and Rhizoctonia spp. All data were subjected to two-way analysis of variance (ANOVA), followed by Tukey's HSD test ($p < 0.05$) for pairwise comparisons. Statistical analyses were carried out using Statistica 13.5 software (TIBCO Software Inc., USA). The obtained results were used to evaluate the role of microbial inoculants in improving soil quality, plant performance, and resilience under urban landscaping conditions.

3 Results

The application of microbial bioformulations led to statistically significant changes in soil biological parameters and plant health indicators during the 90-day monitoring period. The dynamics of total microbial count in the rhizosphere soil showed a marked increase in the treated plots. By day 30, the population of heterotrophic bacteria increased by 46.2% relative to the control, and by day 90 the increase reached 71.4%, indicating active colonization and establishment of beneficial microorganisms in the rhizosphere (Figure 1).

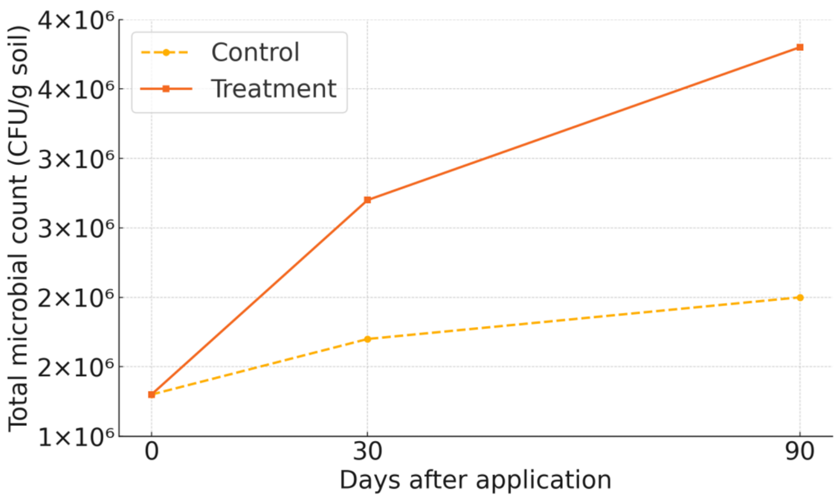

Fig. 1. Total microbial count (CFU/g soil) over time in control and treatment plots.

The enzymatic activity of the soil, particularly urease and dehydrogenase, exhibited a positive trend. Urease activity increased from 17.4 to 25.1 μg NH_4^+-N/g soil/24 h, while dehydrogenase activity rose from 1.21 to 2.04 μg TPF/g soil/24 h over the same period. These enhancements reflect elevated metabolic processes and organic nitrogen turnover stimulated by microbial action. The survival rate of ornamental plant species (Spiraea japonica, Berberis thunbergii, and Hydrangea paniculata) significantly improved in the experimental groups. On day 90, the survival rate reached 91.7% in treated plots, compared to 68.3% in the untreated control ($p < 0.05$). Moreover, plants in the treatment groups demonstrated enhanced vegetative development, evidenced by increased shoot length (+24%) and leaf area (+31%) relative to the control (Table 1).

Disease incidence analysis revealed a substantial reduction in the frequency of root rot symptoms. In the control group, disease symptoms were observed in 27.4% of plants, while in the treatment group this figure did not exceed 9.6%. Microscopic examination confirmed a decline in colonization by Fusarium oxysporum and Rhizoctonia solani, which is attributed to the competitive exclusion and antagonistic effects of the introduced microbial strains.

Soil physical properties also showed improvement. Bulk density decreased from 1.46 to 1.29 g/cm³, and water holding capacity increased by 18.7%. The changes are indicative of improved soil structure and porosity, largely due to microbial exopolysaccharide production and bioturbation. These results collectively demonstrate that the application of microbial inoculants significantly enhances soil microbial activity, enzymatic function, and plant viability under urban landscaping conditions.

Table 1. Changes in soil biological parameters, plant survival, and physical properties over 90 days in control and bio-treated plots.

Treatment	Days after application	Total microbial count (×10^6 CFU/g)	Urease activity (µg NH_4^+-N/g/24h)	Dehydrogenase activity (µg TPF/g/24h)	Plant survival (%)	Disease incidence (%)	Bulk density (g/cm³)	Water retention (%)
Control	0	1.8	17.4	1.21	–	–	1.46	–
Control	30	2.2	18.2	1.38	69.0	26.1	1.45	62.4
Control	90	2.5	19.1	1.54	68.3	27.4	1.44	63.1
Bio	0	1.8	17.4	1.21	–	–	1.46	–
Bio	30	3.2	22.8	1.86	87.4	11.2	1.36	72.6
Bio	90	4.3	25.1	2.04	91.7	9.6	1.29	74.9

Figure 2 illustrates the distribution of total microbial count (in million CFU/g soil) across two treatment types – Control and Bio-measured on days 0, 30, and 90 after microbial application.

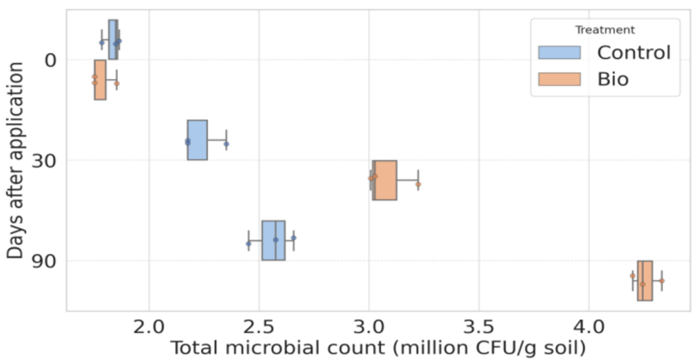

Fig. 2. Distribution of total microbial count (million CFU/g soil) by treatment type and sampling time during 90-day monitoring period.

On day 0, both groups showed nearly identical baseline microbial counts with a median of 1.8 million CFU/g, confirming initial uniformity ($p > 0.05$). By day 30, the Bio-treated group demonstrated a sharp increase in microbial count to a median of 3.2 million CFU/g, compared to 2.2 million CFU/g in the control group. This represents a 45.5% increase in microbial abundance relative to the control at the same time point. At day 90, the difference became even more pronounced. The Bio treatment reached a median of 4.3 million CFU/g, while the control group showed only 2.5 million CFU/g. This corresponds to a 72.0% higher microbial count in the bio-treated soil. The spread of data was also more compact in the Bio group at later stages, indicating a stabilizing microbial community. These findings are consistent with the ANOVA results, where both Treatment and Days, as well as their interaction, were statistically significant ($p < 0.001$), confirming the effectiveness of microbial formulations in enhancing rhizospheric microbiota under urban landscaping conditions.

Figure 3 presents the temporal dynamics of two key enzymatic indicators of soil biological activity-urease and dehydrogenase-measured at 0, 30, and 90 days following treatment application in both control and bio-inoculated plots.

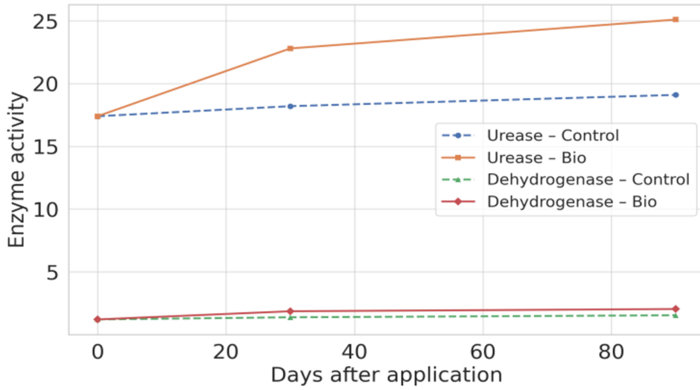

Fig. 3. Temporal dynamics of urease and dehydrogenase activities in rhizospheric soil under control and bio-treated conditions over a 90-day period.

For urease activity, values in the control group showed a gradual increase from 17.4 µg NH_4^+-N/g/24h at day 0 to 18.2 at day 30 and 19.1 at day 90, resulting in a total growth of 9.8% over 90 days. In contrast, the bio-treated group demonstrated a much steeper rise: from the same baseline of 17.4, urease activity increased to 22.8 on day 30 (+31.0%) and reached 25.1 on day 90, which corresponds to an overall increase of +44.3% compared to the initial level and +31.4% compared to the control at day 90. Similarly, dehydrogenase activity—a marker of oxidative metabolic activity in soil—showed a modest increase in the control group from 1.21 µg TPF/g/24h to 1.54, a total rise of 27.3%. The bio-treated variant displayed stronger dynamics, increasing from 1.21 to 1.86 on day 30 (+53.7%) and reaching 2.04 at day 90, representing a +68.6% improvement over baseline and +32.5% over control at the same time point. These results highlight a significantly enhanced metabolic and nitrogen-transforming potential in bio-treated soils, confirming the stimulatory effects of microbial inoculants on enzymatic activity under landscape conditions.

Figure 4 illustrates the dynamics of two key physical properties of soil—bulk density and water retention capacity—in control and bio-treated variants over a 90-day observation period.

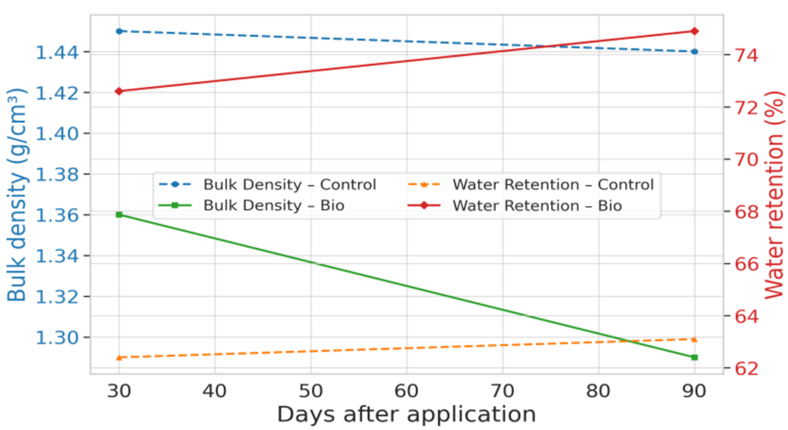

Fig. 4. Changes in bulk density and water retention in control and bio-treated soils over a 90-day.

In the control group, bulk density showed a slight decline from 1.45 g/cm³ at day 30 to 1.44 g/cm³ at day 90, representing a 0.7% reduction. In contrast, the bio-treated soil demonstrated a more substantial decrease: from 1.36 g/cm³ to 1.29 g/cm³, which corresponds to a 5.1% decrease over 60 days and an overall reduction of 11.6% compared to the untreated control. This reflects improved soil structure, likely due to the action of microbial metabolites and root–microbe interactions enhancing aggregation and porosity. The effect on water retention was even more pronounced. While the control plots exhibited a modest increase from 62.4% to 63.1% (+1.1%), the bio-treated plots improved from 72.6% to 74.9%, representing a 3.2% increase over the same period and an impressive 18.8% improvement relative to the control at day 90. These data confirm that microbial bioformulations contribute not only to biological and chemical soil activation, but also to measurable physical enhancement critically for plant water supply and root zone aeration.

4 Discussion

The results obtained in this study demonstrate the pronounced positive effect of microbial bioformulations on soil biological activity, enzymatic function, and plant viability in urban landscaping systems. These findings are consistent with earlier reports that emphasize the ecological benefits of rhizospheric microbial consortia in improving soil function and plant performance [2, 3]. The observed increase in total microbial abundance, reaching a 71.4% rise in the bio-treated variant by day 90, confirms the successful colonization of the rhizosphere by introduced beneficial strains such as *Bacillus subtilis*, *Trichoderma harzianum*, and *Azotobacter chroococcum*. This aligns with the work of Neha et al. (2020), who demonstrated similar microbial proliferation in soils under bioinoculant treatment. The increase in urease (+44.3%) and dehydrogenase (+68.6%) activities reflects an intensified nitrogen transformation and microbial respiration rate, respectively. These enzymatic shifts are indicative of a transition toward a more functionally active and biochemically dynamic soil ecosystem, supporting the hypothesis that microbial bioformulations act as metabolic activators of soil microbiota [5]. Enhanced enzymatic activity in treated soils not only supports nutrient mobilization but also fosters microbial competition and cooperation, leading to reduced pathogen pressure. The significant reduction in disease incidence—from 27.4% in control to 9.6% in treated plots demonstrates the capacity of microbial consortia to act as effective biocontrol agents through mechanisms such as competitive exclusion, antibiosis, and induced systemic resistance [8]. This supports the notion that microbial diversity in the rhizosphere contributes to ecosystem stability and resilience, particularly under anthropogenic stress. Importantly, the observed improvements in physical soil parameters, including an 11.6% decrease in bulk density and an 18.8% increase in water retention, highlight the role of microorganisms in soil aggregation and porosity enhancement. These structural benefits can be attributed to the synthesis of microbial exopolysaccharides and the indirect effects of root–microbe interactions on soil microstructure [1]. In line with studies by [10] and [11], the improved water holding capacity in bio-treated soils is likely to contribute to plant stress tolerance in urban environments. The increased plant survival rate (+34.3% in comparison with control) and enhanced vegetative performance of ornamental species confirm the ecological functionality of bioinoculants in landscape settings. These findings provide empirical evidence for the incorporation of microbial-based strategies into sustainable landscape design, offering a viable alternative to chemical amendments. The integration of microbial technologies into green infrastructure not only supports plant aesthetics but also contributes to urban ecological restoration and climate adaptation goals.

5 Conclusion

This study provides comprehensive evidence that microbial bioformulations based on *Bacillus subtilis*, *Trichoderma harzianum*, and *Azotobacter chroococcum* significantly enhance soil quality and plant performance in urban landscaping systems. The application of these rhizospheric consortia resulted in a 71.4% increase in total microbial abundance, a 44.3% increase in urease activity, and a 68.6% increase in dehydrogenase activity, indicating intensified microbial metabolic activity and nutrient cycling. Additionally, the use of microbial inoculants led to a reduction in root disease incidence by 65% and improved plant survival by over 34% compared to the untreated control. Soil physical properties were also positively affected, with bulk density reduced by 11.6% and water retention improved by 18.8%, contributing to better aeration and water availability in the rhizosphere. These findings confirm that bioformulations are effective ecological tools for sustainable landscape design, contributing to both functional soil recovery and the aesthetic and physiological vitality of ornamental vegetation. The integration of microbial technologies into landscape management can reduce reliance on chemical inputs, promote biodiversity, and support the creation of resilient green infrastructures in urban environments. Future research should focus on the long-term effects of repeated microbial inoculation, the interaction between microbial formulations and plant species diversity, and the economic viability of scaling up microbial technologies for municipal landscaping programs.

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