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A Rhizobacterial Consortium Improves Crop Productivity, Stress Tolerance, and Soil Health

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Abstract
Plant growth-promoting rhizobacteria (PGPR) offer an environmentally benign route to sustaining crop productivity while curtailing dependence on synthetic agrochemicals. This study isolated, characterised and functionally screened rhizobacteria recovered from four contrasting agricultural soils, and evaluated elite strains and a defined consortium across a graded series of controlled, stress, biocontrol and field experiments conducted over two cropping seasons. Forty-eight morphologically distinct isolates were obtained, of which twelve were shortlisted and assigned to six genera by 16S rRNA gene sequencing, with Bacillus and Pseudomonas predominating. In vitro screening identified three elite strains — Bacillus velezensis PGPR-4 and PGPR-8 and Pseudomonas putida PGPR-9 — that combined strong phosphate solubilisation, indole-3-acetic acid (IAA) production, siderophore synthesis and broad-spectrum antifungal activity; a fourth strain, Azospirillum brasilense PGPR-3, contributed vigorous nitrogen fixation. A compatible consortium of the four strains consistently outperformed the individual strains, raising germination, root and shoot length, biomass, chlorophyll content and nutrient uptake under controlled conditions, and mitigating drought and salinity stress through improved water relations, ionic balance and antioxidant defence. The consortium suppressed three soil-borne fungal pathogens with a biocontrol efficacy of approximately 65 %. Under field conditions the consortium alone approached the yield of the full recommended fertiliser dose, while the consortium combined with 75 % of the recommended dose exceeded it (5.0 t ha^{-1}), demonstrating a substantial fertiliser-substitution effect. Inoculation also enhanced soil microbial biomass, enzyme activities and nutrient status while preserving microbial diversity.

Keywords: plant growth-promoting rhizobacteria; microbial consortium; biofertiliser; abiotic-stress tolerance; biocontrol; soil health; integrated nutrient management

1. Introduction

The intensification of agriculture over recent decades has been sustained largely through the heavy application of synthetic nitrogen and phosphatic fertilisers. While these inputs have delivered substantial gains in productivity, their continued and often indiscriminate use has been accompanied by declining nutrient-use efficiency, soil acidification and salinisation, the loss of soil organic carbon and microbial diversity, and the eutrophication of water bodies and emission of greenhouse gases associated with nutrient losses. In many intensively cropped soils the nitrogen-use efficiency of applied fertiliser has fallen to only around a third of that applied, so that the greater part of the nutrient supplied is lost to the environment rather than converted

into yield. There is therefore a pressing need for technologies that can maintain or improve productivity while reducing reliance on synthetic inputs and safeguarding the biological health of the soil.

Plant growth-promoting rhizobacteria (PGPR) — beneficial bacteria that colonise the rhizosphere and root surface — have emerged as a leading candidate for this purpose. PGPR promote plant growth through a combination of direct and indirect mechanisms, including biological nitrogen fixation, the solubilisation of insoluble soil phosphorus, the production of phytohormones such as indole-3-acetic acid (IAA), the synthesis of iron-chelating siderophores, and the suppression of phytopathogens through the production of antibiotics, lipopeptides, volatile compounds and cell-wall-degrading enzymes. By enhancing nutrient acquisition and modulating root architecture, and by conferring tolerance to abiotic and biotic stresses, PGPR can improve both the yield and the nutritional quality of crops while permitting a reduction in the application of synthetic fertilisers and pesticides.

A recurrent obstacle to the wider adoption of microbial inoculants has been the inconsistency of their performance under field conditions, where introduced strains must establish themselves within a competitive resident community and contend with variable environmental and edaphic conditions. Increasing evidence indicates that multi-strain consortia, which combine functionally complementary strains, deliver broader and more stable benefits than single strains, because functional redundancy buffers the inoculant against the failure of any one function under unfavourable conditions. The present study was designed to test the hypothesis that indigenous rhizobacterial isolates possessing multiple plant growth-promoting attributes, deployed individually and as a defined consortium, could enhance crop productivity and soil quality while permitting a reduction in the quantity of chemical fertiliser applied. To this end a structured screening cascade was employed, progressively narrowing a large isolate collection to a small number of elite strains, which were then evaluated across a continuum of controlled, stress, biocontrol and field experiments.

2. Materials and Methods

2.1 Study sites and rhizosphere sampling

Rhizospheric soil and root samples were collected from agricultural fields under cereal and legume cultivation at four geographically distinct sites (Site A–D), selected to span a gradient of soil texture, reaction, electrical conductivity, organic carbon and management history. Sampling was carried out during the active vegetative growth stage, when rhizosphere microbial activity and root exudation are greatest. Healthy plants were excavated together with their adhering soil; the loosely attached bulk soil was removed, and the soil firmly adhering to the root surface was collected aseptically into sterile containers. Three composite samples were prepared per site, transported cool, and processed within twenty-four hours. Soil physicochemical properties (texture, pH, electrical conductivity, organic carbon, available N, P and K, and microbial biomass carbon) were determined by standard soil-testing protocols prior to microbiological processing.

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2.2 Isolation, purification and characterisation

Bacteria were isolated by serial dilution and spread-plating on nutrient agar and selective media, including nitrogen-free media for the enrichment of free-living diazotrophs. Morphologically distinct colonies were purified by repeated streaking and preserved both as agar slants and as glycerol stocks under deep-freeze conditions. Isolates were characterised morphologically (colony form, pigmentation, Gram reaction, cell shape, motility and endospore formation) and biochemically (catalase, oxidase, citrate, indole, methyl-red, Voges–Proskauer and urease tests, carbohydrate fermentation, ammonia production and starch and gelatin hydrolysis). Definitive identification of the shortlisted isolates was achieved by amplification and sequencing of the 16S rRNA gene using universal bacterial primers, comparison of sequences against reference databases, and construction of a bootstrap-supported phylogenetic tree.

2.3 Screening for plant growth-promoting traits

All isolates were screened in vitro for nitrogen-fixation potential (growth on nitrogen-free medium and nitrogenase activity), phosphate solubilisation (halo formation on Pikovskaya's agar, the phosphate-solubilisation index, and soluble phosphorus released into liquid culture), IAA production (colorimetric assay in tryptophan-supplemented medium), siderophore production (chrome azurol S agar, expressed as percentage siderophore units), and antagonism against the soil-borne fungal pathogens

Fusarium oxysporum, Rhizoctonia solani and Macrophomina phaseolina by the dual-culture technique. Each assay was performed in triplicate. On the basis of the comparative trait assessment, three elite strains were selected for detailed evaluation and combined with the strongest diazotroph to form a defined consortium, the compatibility of whose members was confirmed by cross-streak assays.

2.4 In planta, stress, biocontrol and field evaluation

Elite strains and the consortium were grown to a standardised cell density; seeds were inoculated by immersion in the bacterial suspension with a carrier and adhesive, and uninoculated controls received sterile broth. Plant growth was evaluated first in sterile growth pouches and then in pot culture (completely randomised design, four replicates), measuring germination, seedling vigour, root and shoot length, dry biomass, chlorophyll content, the maximum quantum efficiency of photosystem II (Fv/Fm), and tissue N, P and K. Abiotic-stress tolerance was assessed under controlled drought (soil maintained at a defined fraction of field capacity) and salinity (irrigation to a defined electrical conductivity) in a factorial design, measuring relative water content, proline, chlorophyll, the shoot Na^+/K^+ ratio and superoxide dismutase (SOD) activity. Biocontrol potential was assessed by in vitro dual-culture antagonism and by in planta disease incidence and severity in Fusarium-challenged plants, with biocontrol efficacy computed as the proportional reduction in disease incidence relative to the challenged control. Field experiments were laid out in a randomised complete block design (four replicates) over two cropping seasons, comparing an uninoculated control (T1), the recommended dose of fertiliser (T2, 100 % RDF), individual elite-strain inoculation (T3), consortium inoculation (T4), and the consortium combined with 75 % RDF (T5). Data were

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analysed by analysis of variance with mean separation by least significant difference, together with correlation, regression and principal component analysis.

3. Results and Discussion

3.1 Soil characteristics and isolate recovery

The four sampling soils differed markedly in their physicochemical properties (Table 1), ranging from slightly acidic sandy loams (Sites A and D) to a near-neutral loam (Site B) and a moderately alkaline clay loam (Site C). The loam and clay loam soils of Sites B and C, which exhibited the highest organic carbon, available nutrients and microbial biomass carbon, yielded the greatest number of isolates and the broadest generic diversity, consistent with the established principle that soil organic matter and nutrient status are primary determinants of microbial abundance and diversity. A total of forty-eight morphologically distinct colonies were recovered across the four sites and preserved for characterisation.

Soil property	Site A	Site B	Site C	Site D
Texture	Sandy loam	Loam	Clay loam	Sandy loam
pH (1:2.5 H_2O)	6.8	7.4	8.1	6.5
Electrical conductivity (dS m^{-1})	0.42	0.58	0.91	0.39
Organic carbon (%)	0.54	0.71	0.66	0.48
Available N (kg ha^{-1})	218	264	241	202
Available P (kg ha^{-1})	14.2	19.6	16.8	12.1
Available K (kg ha^{-1})	178	226	254	165
Microbial biomass C (mg kg^{-1})	182	241	209	171

Table 1. Baseline physicochemical properties of rhizospheric soils at the four sampling sites (mean of three composite samples per site).

3.2 Morphological, biochemical and molecular identification

The isolates displayed considerable morphological diversity, with a mixture of Gram-positive and Gram-negative, predominantly rod-shaped cells, and both endospore-forming and non-spore-forming types. Catalase activity was detected in all twelve shortlisted isolates, an attribute that enhances survival under the oxidative conditions of the rhizosphere, while ammonia production and the hydrolysis of starch and gelatin — traits associated with nutrient mobilisation and biocontrol — were prominent among the Gram-positive, spore-forming isolates. Sequencing of the 16S rRNA gene assigned the twelve shortlisted isolates to six genera (Table 2), with the closest type-strain similarities ranging from 98.6 % to 99.7 %. The genera Bacillus and Pseudomonas predominated, in agreement with the morphological and biochemical profiles and with the general predominance of these robust, readily culturable genera reported for rhizosphere communities. A phylogenetic tree confirmed the clustering of isolates into well-supported clades corresponding to their assigned genera.

Isolate	Closest species	Genus	Similarity (%)
PGPR-1	Pseudomonas fluorescens	Pseudomonas	99.2

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PGPR-2	Bacillus subtilis	Bacillus	99.5
PGPR-3	Azospirillum brasilense	Azospirillum	98.7
PGPR-4	Bacillus velezensis	Bacillus	99.6
PGPR-5	Azotobacter chroococcum	Azotobacter	98.9
PGPR-6	Bacillus amyloliquefaciens	Bacillus	99.4
PGPR-7	Enterobacter cloacae	Enterobacter	98.8
PGPR-8	Bacillus velezensis	Bacillus	99.7
PGPR-9	Pseudomonas putida	Pseudomonas	99.3
PGPR-10	Azotobacter vinelandii	Azotobacter	98.6
PGPR-11	Bacillus subtilis	Bacillus	99.5
PGPR-12	Serratia marcescens	Serratia	98.9

Table 2. Molecular identification of the twelve shortlisted PGPR isolates based on 16S rRNA gene sequencing. Species names are italicised in the text; similarity is to the nearest type strain.

3.3 Plant growth-promoting traits

Functional screening revealed that plant growth-promoting capacity was unevenly distributed, with a minority of strains combining strong performance across several traits and the remainder excelling in only one or two (Table 3). The diazotrophic genera Azospirillum and Azotobacter (PGPR-3, PGPR-5, PGPR-10) showed the most vigorous growth on nitrogen-free medium and the highest nitrogenase activity, whereas the Bacillus velezensis isolate PGPR-8 emerged as the outstanding multifunctional strain, ranking first or near first for the phosphate-solubilisation index (4.2), IAA production (72 µg mL^{-1}), siderophore production (52 % SU) and antifungal activity (71 % inhibition). The Pseudomonas putida isolate PGPR-9 and the Bacillus velezensis isolate PGPR-4 also combined several strong traits. The prominence of fluorescent pseudomonads among the siderophore producers is consistent with their well-known capacity to synthesise high-affinity siderophores such as pyoverdine, while the strong antifungal activity of the Bacillus isolates reflects the rich repertoire of cyclic lipopeptides — surfactin, iturin and fengycin — that this genus produces to disrupt fungal membranes and inhibit spore germination. This uneven distribution of traits, in which no single isolate excelled in every function, provided the principal justification for the assembly of a multi-strain consortium.

Isolate	N-fixation	PSI	IAA (µg mL^{-1})	Siderophore (% SU)	Antifungal (% inhib.)	Multi-trait score
PGPR-1	+	2.1	18	21	41	Moderate
PGPR-2	+	3.4	42	35	48	High
PGPR-3	+++	2.8	31	28	32	Moderate
PGPR-4	+	3.9	58	44	58	High
PGPR-5	+++	2.3	22	19	26	Moderate
PGPR-6	+	3.1	49	38	61	High
PGPR-7	+	2.6	27	24	37	Moderate
PGPR-8	++	4.2	72	52	71	Very high

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PGPR-9	++	3.7	61	46	66	Very high
PGPR-10	+++	2.0	15	16	24	Moderate
PGPR-11	+	3.3	45	33	49	High
PGPR-12	+	2.9	38	29	44	Moderate

Table 3. Comparative summary of plant growth-promoting traits of the twelve shortlisted PGPR isolates. N-fixation: + weak, ++ moderate, +++ strong; PSI = phosphate-solubilisation index; % SU = percentage siderophore units.

On this basis, three elite isolates (PGPR-4, PGPR-8 and PGPR-9) were selected for detailed evaluation and combined with the strongest diazotroph, PGPR-3 (Azospirillum brasilense), to constitute a defined consortium integrating vigorous nitrogen fixation with phosphate solubilisation, phytohormone production and biocontrol. Cross-streak assays confirmed that the constituent strains were mutually compatible — an essential precaution, since strains effective in isolation may inhibit one another in combination.

3.4 Plant growth under controlled conditions

Inoculation significantly enhanced every measured growth parameter, with the consortium consistently outperforming the individual strains. Germination increased from approximately 72 % in the control to 86–91 % for individual strains and about 95 % for the consortium, attributable to phytohormone-stimulated mobilisation of seed reserves and the suppression of seed- and soil-borne pathogens. Root length increased from 9.8 cm in the control to 17.8 cm under the consortium (about 82 %), shoot length from 21.4 to 34.7 cm (about 62 %), and dry biomass from 0.92 to 1.73 g plant^{-1} (about 88 %). Among individual strains, PGPR-8 produced the greatest enhancement, consistent with its high IAA-producing capacity. Inoculation also raised leaf chlorophyll content and Fv/Fm, and improved tissue N, P and K (Table 4), providing a direct mechanistic link between the in vitro traits of the strains and the in planta growth response. The magnitude of the root and shoot responses is comparable to that reported for effective growth-promoting rhizobacteria in the recent literature. The consistent superiority of the consortium indicates a synergistic effect arising from functional complementarity, as the simultaneous improvement of nitrogen and phosphorus availability coupled with hormonally driven root expansion relieves multiple growth constraints concurrently.

Treatment	**Tissue N (%)**	**Tissue P (%)**	**Tissue K (%)**	**Chlorophyll (mg g^{-1})**	**Fv/Fm**
Control	1.62	0.21	1.84	3.1	0.74
PGPR-4	2.04	0.29	2.11	3.6	0.78
PGPR-8	2.21	0.33	2.24	3.9	0.80
PGPR-9	2.12	0.30	2.18	3.7	0.79
Consortium	2.38	0.37	2.41	4.2	0.82

Table 4. Effect of PGPR inoculation on tissue nutrient content, chlorophyll and photosystem-II efficiency (Fv/Fm) under controlled conditions (mean of four replicates).

3.5 Abiotic-stress tolerance

Both drought and salinity markedly reduced growth in uninoculated plants, but inoculation substantially mitigated these effects (Table 5). Under drought, leaf relative water content

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declined from 88 % (well-watered) to 61 % in uninoculated stressed plants but was maintained at 82 % under the consortium, while proline accumulation — a marker of osmotic adjustment — rose further under inoculation, from 38 to 61 µmol g^{-1}. These responses reflect the promotion of deeper root systems, exopolysaccharide-mediated improvement of soil water retention, and modulation of stomatal behaviour. Under salinity, the shoot Na^+/K^+ ratio rose from 0.42 (non-saline) to 1.31 in uninoculated stressed plants but was held to 0.71 under the consortium, while SOD activity increased from 29 to 47 U mg^{-1}, indicating a strengthened antioxidant defence against reactive oxygen species. The consortium consistently provided the greatest amelioration, but among individual strains PGPR-8 was most effective under drought and PGPR-9 under salinity — a divergence consistent with the desiccation tolerance and exopolysaccharide production of Bacillus and the siderophore and antioxidant-inducing capacity of fluorescent pseudomonads. The complementary stress specialisations of the strains provide a further rationale for the consortium, whose functional redundancy conferred robust protection across both stress types. Notably, the relative advantage of inoculation was greater under stress than under non-stressed conditions, indicating that PGPR benefits become most pronounced precisely when plants are most challenged.

Parameter	Stress (no PGPR)	Stress + individual strain	Stress + consortium	Improvement (%)
Root length (cm) — drought	7.6	11.8	13.4	76
Shoot length (cm) — drought	16.2	23.1	25.9	60
Relative water content (%) — drought	61	76	82	34
Proline (µmol g^{-1}) — drought	38	52	61	61
Root length (cm) — salinity	8.1	12.0	13.1	62
Na^+/K^+ ratio — salinity	1.31	0.86	0.71	−46
SOD activity (U mg^{-1}) — salinity	29	41	47	62

Table 5. Effect of PGPR inoculation on growth and physiological parameters under drought and salinity stress. Improvement is for the consortium relative to the uninoculated stressed control; a negative Na^+/K^+ value denotes a desirable reduction.

3.6 Biocontrol potential

The consortium exhibited the strongest in vitro antagonism, inhibiting mycelial growth of Fusarium oxysporum, Rhizoctonia solani and Macrophomina phaseolina by 72–78 %, while the Bacillus isolate PGPR-8 achieved 64–71 %. The strong, broad-spectrum antagonism of the Bacillus isolates and the consortium is attributable to the production of cyclic lipopeptides such as iturin and fengycin, which disrupt fungal membranes, together with cell-wall-degrading enzymes (chitinases and glucanases) and volatile antifungal compounds confirmed in sealed-

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plate assays. The siderophore-producing Pseudomonas isolate PGPR-9 further contributed through competition for iron. In planta, inoculation reduced Fusarium wilt incidence from approximately 68 % in the challenged control to 24 % under the consortium, corresponding to a biocontrol efficacy of about 65 % — a value comparable to that reported for effective Bacillus-based biocontrol agents. The operation of multiple, complementary suppression mechanisms accounts for the robustness of the activity and reduces the likelihood of pathogens rapidly evolving resistance, as they might against a single mode of action. The marked inhibition of Macrophomina phaseolina, a pathogen of particular concern in drought-prone systems, is noteworthy, as the coincidence of biocontrol activity with drought-tolerance promotion within the same strains offers an integrated route to managing compound stresses.

3.7 Field performance and fertiliser substitution

Under field conditions, sustained across two cropping seasons, inoculation significantly enhanced plant height, tiller number, leaf area and yield attributes, although the magnitude of the effects was more modest than in pot experiments — a well-documented attenuation reflecting competition from the indigenous microbial community. The uninoculated control yielded 3.2–3.4 t ha^{-1} and the full recommended fertiliser dose 4.6–4.8 t ha^{-1}. Critically, the consortium applied without supplementary fertiliser achieved 4.4–4.6 t ha^{-1}, approaching the full-dose yield, while the integrated treatment combining the consortium with 75 % RDF produced the highest yield of all, 4.9–5.1 t ha^{-1}, exceeding the full fertiliser dose (Table 6). The yield gains arose from improvements in multiple yield components — productive tillers, grains per panicle and thousand-grain weight — rather than any single attribute. This fertiliser-substitution effect, permitting a 25 % reduction in chemical fertiliser without yield penalty, is consistent with recent field studies and carries substantial economic and environmental benefits: lower production costs for resource-limited farmers, and reduced nutrient losses to water and atmosphere. The superiority of the integrated treatment reflects the complementary roles of biological and chemical inputs — chemical fertiliser meeting immediate nutrient demand while PGPR enhance the efficiency with which applied and indigenous nutrients are acquired, fix additional nitrogen, mobilise fixed phosphorus, and improve root development and stress resilience. The proportional benefit of inoculation was stable between seasons, an encouraging indication of consistent performance under realistic conditions.

Treatment	Productive tillers (m^{-2})	Grains panicle^{-1}	1000-grain wt (g)	Grain yield (t ha^{-1})
Control	248	96	21.4	3.3
RDF (100 %)	312	124	23.8	4.7
PGPR-8	286	112	22.9	4.2
Consortium	301	119	23.4	4.5
Consortium + 75 % RDF	326	131	24.6	5.0

Table 6. Effect of field treatments on yield attributes and grain yield (mean of two cropping seasons, four replicates per treatment). RDF = recommended dose of fertiliser.

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3.8 Soil health and statistical synthesis

Inoculation exerted a broadly positive and ecologically benign influence on the soil ecosystem. Soil microbial biomass carbon and the activities of dehydrogenase, urease and alkaline phosphatase were all significantly increased, with the consortium producing the greatest enhancements; dehydrogenase activity, for example, rose from 42 to 98 µg triphenyl-formazan g^{-1} day^{-1}. These enzymes catalyse the conversion of organic nutrient forms into plant-available inorganic forms, providing a mechanistic link between the soil-biological and plant-level effects of inoculation. Available soil N, P and K at harvest were higher in inoculated plots, indicating a residual-fertility benefit extending beyond the standing crop, while high-resolution community analysis showed a moderate enrichment of beneficial taxa without loss of overall microbial diversity — a desirable property indicating that the introduced strains acted in concert with, rather than against, the indigenous community. Analysis of variance confirmed that the treatment effects on growth, yield, physiological and soil parameters were highly significant ($p < 0.01$) with low coefficients of variation, and mean separation confirmed that the consortium and integrated treatments differed significantly from the control. Correlation analysis revealed strong positive associations between IAA production and root length and between phosphate solubilisation and both biomass and yield, while regression analysis showed that root length was predictable from IAA production and grain yield from plant biomass. Principal component analysis separated the treatments along a primary axis of productivity and soil biological activity, with the consortium and integrated treatments at the high-productivity end and the control at the opposite extreme, confirming the tight coupling between agronomic productivity and soil biological health.

4. Conclusion

This study demonstrates that indigenous rhizobacterial strains possessing multiple plant growth-promoting attributes can substantially enhance crop productivity, confer tolerance to drought and salinity, suppress soil-borne fungal pathogens, improve nutrient-use efficiency and enhance soil health, and that these benefits are greatest when functionally complementary strains are combined within a consortium and integrated with judicious chemical-fertiliser management. The central and most consistent finding is the superiority of the multifunctional consortium over individual strains across virtually every parameter and condition examined, arising from the integration of nitrogen fixation, phosphate solubilisation, phytohormone production and biocontrol within a single inoculant. Of particular significance for sustainability is the demonstration that the consortium could partially substitute for chemical fertiliser, with the integrated treatment matching or exceeding full-dose yields while permitting a 25 % reduction in fertiliser, and that these agronomic benefits were achieved in conjunction with, rather than at the expense of, soil health. The principal limitations — the single-location nature of the field trials, albeit across two seasons, and the reliance on culture-dependent characterisation — indicate the need for multi-location validation and culture-independent community analysis. Notwithstanding these limitations, the consistency, coherence and statistical robustness of the findings provide a strong evidence base for the deployment of

multifunctional PGPR consortia within integrated nutrient-management strategies for climate-resilient, environmentally responsible crop production.

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