Soil Microbiome Engineering through Synthetic Microbial Consortia for Sustainable Wheat Production under Climate Change

Dr. Viraj Thornwell

Pro. Department of Soil Fertility and Nutrient Management, Agro Frontier University, India

*Corresponding author: **Dr. Viraj Thornwell**

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ABSTRACT

The availability of wheat is severely affected by climate change, among other issues, and synthetic microbial consortia may help alleviate this problem. Purpose: This research focused on the effectiveness of designed soil microbiomes in terms of the wheat yield. Methods: A field study was conducted for two growing seasons, applying different climatic conditions and microbial consortium types in the study. Findings: The use of the designed microbial consortium was found to have increased the wheat yield by 28-35% and by 32-38%, respectively, under drought and heat stress. The microbial biomass carbon has risen by 46%, alongside a measurable increase in the microbial biodiversity indices. Conclusions: Engineered soil microbiomes can be used for developing climate-resilient wheat production systems.

Keywords: Soil microbiome engineering, Synthetic microbial consortia, Plant–microbe interactions, Climate change adaptation, Wheat production, Sustainable agriculture, Rhizosphere ecology, Nutrient cycling

1. Introduction

Wheat (*Triticum aestivum* L.) is one of the major crops worldwide, making up around 20% of the overall calorie needs of people across the world (Food and Agriculture Organization of the United Nations, 2023) [1]. However, climatic impacts, like extended drought and increased temperatures, greatly change the functioning of soil ecosystems, which are vital for obtaining nutrients and resisting stress (Asseng *et al*., 2015) [2] (Jansson and Hofmockel, 2020) [3]. Traditional agronomic methods of soil usage experienced the diminishing effects and led to ruin of environment instead (Lal *et al*., 2011) [4]. The microbiome of soil is essential in the functioning of ecosystems, recycling of nutrients, and health of plants (Trivedi *et al*., 2020) [5]. The region of the soil around plant roots called rhizosphere is the center of microbial activity and interactions between plants and microorganisms (Reinhardt *et al*., 2022) [6]. Throughout climate change, the functioning of microbes in this region is being disturbed by changes in moisture and temperature regimes, resulting in decreased productivity osf crops (Sharma and Singhvi, 2017) [7]. One of the promising ways of improving soil health and enabling crops to sustain climatic strains is through soil microbiome engineering realized through synthetic microbial consortia (Delgado-Baquerizo *et al*., 2023) [8] (Zhou *et al*., 2021) [9]. Unlike simple inoculants, synthetic consortia are designed to include functionally complementary microorganisms performing linked ecological functions (Gopal and Saha, 2022) [10]. The contents of synthesizing consortia include nitrogen-fixing bacteria, phosphate-dissolving bacteria, potassium-transmitting microorganisms, arbuscular mycorrhizal fungi, and beneficial bacteria (Kumar and Verma, 2018) [11]. Plant growth-promoting microorganisms (PGPM Despite resistance to climate challenges, the use of plant growth-promoting microorganisms leads to the accumulation of osmolytes, sodium ion, and effective functioning of antioxidant enzymes (Glick, 2012) [12]. Field experiments on the interactions between multi-component microorganisms and different climatic situations are still insufficient (Bhattacharyya and Jha, 2012) [13]. In this report, we tackle the gap in knowledge of designing a synthetic microbial community that is efficient in wheat cultivation and ensure the operation of the community as well as rhizosphere colonization, analyze the process of diversity and nutrient cycling in the soil, as well as the implications for wheat growth and productivity under climate conditions.

2. Materials and Methods

2.1. Study Site and Experimental Design

The field experiment was conducted at the Research Farm of the Indian Institute of Agricultural Research, New Delhi, India (28.64°N, 77.16°E, elevation 216 m) during two consecutive wheat-growing seasons (October 2022–May 2024). The site experiences a semi-arid climate with mean annual precipitation of 714 mm and mean growing-season temperatures of 22–32°C. The experimental soil was a sandy loam with initial soil organic carbon of 4.2 g kg^{-1}, pH 7.3, available phosphorus 12 mg kg^{-1}, and available potassium 156 mg kg^{-1}. The wheat cultivar 'DBW-187' was selected for its commercial importance and moderate susceptibility to drought and heat stress (Carvalhais *et al*., 2019) [15].

2.2. Synthetic Microbial Consortium

The engineered consortium comprised seven microbial taxa selected based on complementary functional traits and demonstrated plant growth-promoting capacity: (1) nitrogen-fixing bacteria: *Azospirillum brasilense* strain Sp245 and *Bacillus subtilis* strain GB03; (2) phosphate-solubilizing bacteria: *Bacillus megaterium* strain B14 and *Pseudomonas fluorescens* strain PfM1; (3)

potassium-mobilizing bacterium: *Bacillus mucilaginosus* strain KSM-K9; (4) arbuscular mycorrhizal fungi: *Rhizophagus irregularis* isolate INVAM-IL-169; and (5) beneficial antagonistic bacterium: *Bacillus subtilis* GB03. Microbial cells were cultured to mid-exponential phase and combined at a ratio of 10^9 CFU g^{-1} dry carrier material. Prior to field application, consortium members were validated for functional traits and tested for absence of phytotoxic effects (Peiffer *et al*., 2013) [16].

2.3. Experimental Treatments

The experiment employed a split-plot design with climate conditions as main plots (normal rainfall, drought simulating RCP 4.5, and combined heat and drought simulating RCP 8.5) and microbial treatments as subplots (T1 = control without consortium, T2–T4 = consortium at 10^8, 10^9, 10^{10} CFU g^{-1} soil respectively, T5 = consortium at 10^9 CFU g^{-1} plus biochar at 5 t ha^{-1}). Each treatment combination was replicated four times in randomized blocks, yielding 60 total plots.

2.4. Soil Microbiome and Health Assessment

Soil microbial community composition was determined through 16S rRNA gene amplicon sequencing of DNA extracted from rhizosphere soil samples collected at multiple growth stages. Metagenomic analysis identified functional genes associated with nitrogen cycling, phosphorus metabolism, and organic matter decomposition (Zhai *et al*., 2018) [21]. Soil microbial biomass carbon was quantified via the chloroform fumigation-extraction method. Enzyme activities (acid phosphatase, alkaline phosphatase, dehydrogenase, cellulase) were assayed using standard colorimetric procedures. Soil aggregate stability and water-holding capacity were measured according to established protocols. Rhizosphere colonization of consortium members was determined through quantitative real-time PCR targeting strain-specific genetic markers (Drogue *et al*., 2012) [17].

2.5. Plant Growth and Stress Assessments

Plant growth parameters (height, tiller number, leaf area index, dry biomass) were recorded at multiple Zadoks developmental stages. Root architecture parameters were quantified through core sampling and image analysis. Yield components (spike number, grain number per spike, thousand-grain weight) and grain yield were recorded at physiological maturity. Water Use Efficiency and Nutrient Use Efficiency were calculated as grain yield divided by seasonal water use and plant nutrient content, respectively (Philippot *et al*., 2012) [18]. Physiological responses to drought and heat stress were quantified through analysis of osmolytes and antioxidant enzyme activities in leaf tissues (Dennis *et al*., 2010) [19].

2.6. Statistical Analysis

All data were subjected to analysis of variance (ANOVA) using the general linear model procedure, with climate and microbial treatments as fixed factors. Treatment means were compared using the least significant difference (LSD) test at $P < 0.05$. Principal component analysis (PCA) was performed on microbial community composition data. Pearson correlation coefficients assessed relationships between microbial diversity and agronomic performance. All analyses were performed using R statistical software version 4.2.3 (Zhao and Wang, 2021) [20].

3. Results

Table 1: Experimental Site Characteristics, Soil Physicochemical Properties, and Wheat Cultivar Information

Parameter	Value/Description
Latitude/Longitude/Elevation	28.64°N, 77.16°E, 216 m a.s.l.
Climate	Semi-arid; annual precipitation 714 mm
Growing season temperature	Maximum 22–32°C, Minimum 8–18°C
Soil texture	Sandy loam (65% sand, 18% silt, 17% clay)
Soil organic carbon (initial)	4.2 g kg^{-1}
Soil pH (1:2.5 soil:water)	7.3
Available N/P/K (initial)	0.12%, 12 mg kg^{-1}, 156 mg kg^{-1}
Wheat cultivar	DBW-187
Experimental duration	October 2022–May 2024 (two seasons)
Plot size and replication	5 m × 4 m; 4 replicates per treatment

Table 2: Experimental Design: Climate and Microbial Consortium Treatments

Treatment	Climate	Consortium	CFU g^{-1}	Replications	Measurements
T1	Normal/Drought/Heat	Control	0	4 reps	Baseline measurements
T2	Normal/Drought/Heat	Consortium	10^8	4 reps	Growth, yield, microbiome
T3	Normal/Drought/Heat	Consortium	10^9	4 reps	Growth, yield, microbiome
T4	Normal/Drought/Heat	Consortium	10^{10}	4 reps	Growth, yield, microbiome
T5	Normal/Drought/Heat	Consortium + Biochar	10^9 + 5 t/ha	4 reps	Growth, yield, microbiome

Table 3: Composition of Synthetic Microbial Consortium: Taxa, Functional Groups, and Plant Growth-Promoting Traits

Microbial Species/Strain	Functional Group	Key Traits	Ecological Role
Azospirillum brasilense Sp245	N-fixing	Nitrogen fixation, IAA, ACC deaminase	Nitrogen provision, stress mitigation
Bacillus subtilis GB03	N-fixing/antagonistic	Nitrogen fixation, antibiotics, biofilm	Nitrogen provision, disease suppression
Bacillus megaterium B14	P-solubilizing	Phosphate solubilization, acid production	Phosphorus mobilization
Pseudomonas fluorescens PfM1	P-solubilizing	Phosphate solubilization, siderophores	Phosphorus and Fe mobilization, ISR
Bacillus mucilaginosus KSM-K9	K-mobilizing	Mineral weathering, organic acids	Potassium mobilization
Rhizophagus irregularis INVAM-IL-169	Arbuscular mycorrhizal	Root colonization, nutrient acquisition	Root extension, nutrient transfer, WUE
Bacillus subtilis GB03	Beneficial	Volatile organic compounds	Rhizosphere functions, stress tolerance

3.1. Soil Physicochemical Properties

Following consortium application over two growing seasons, significant improvements in soil physicochemical properties were observed (Table 4). Soil organic carbon increased from 4.2 g kg^{-1} (initial) to 6.8 g kg^{-1} in T4 treatments at the end of season two, representing a 61.9% increase (Zhai *et al*., 2018) [21]. Available nitrogen increased by 69.0%, available phosphorus by 133.3%, and available potassium by 57.1% (Zhai *et al*., 2018) [21]. Soil aggregate stability (mean weight diameter) improved from 1.94 mm to 2.67 mm, indicating enhanced soil structure and water-holding capacity (Zhai *et al*., 2018) [21]. Cation exchange capacity increased from 14.2 to 18.9 cmol kg^{-1} in T4 treatments (Zhai *et al*., 2018) [21].

Table 4: Changes in Soil Physicochemical Properties Following Microbial Consortium Application (End of Season 2)

Property	Control	T2 (10^8)	T3 (10^9)	T4 (10^{10})	T5 (+BC)
Soil organic carbon (g kg^{-1})	4.8	5.6	6.4	6.8	6.5
pH (1:2.5)	7.2	7.2	7.1	7.1	7.0
EC (dS m^{-1})	0.48	0.51	0.54	0.56	0.58
Available N (mg kg^{-1})	178	218	256	284	278
Available P (mg kg^{-1})	18	22	26	28	29
Available K (mg kg^{-1})	168	198	224	245	251
Cation exchange capacity (cmol kg^{-1})	14.2	15.8	17.4	18.9	19.6
Field moisture capacity (%)	22.4	24.1	26.8	28.6	29.2
Aggregate stability (mm)	1.94	2.16	2.42	2.67	2.71

3.2. Soil Biological Indicators and Microbial Dynamics

Soil microbial biomass carbon increased from 187 µg g^{-1} in controls to 273 µg g^{-1} in T4 treatments (45.9% increase). Shannon diversity indices increased from 6.84 to 7.62, indicating enhanced microbial functional diversity. Consortium members established stable populations in the rhizosphere, with Azospirillum and Bacillus taxa comprising 12–18% of total bacterial sequences versus <1% in controls (Chen *et al*., 2023) [22]. Enzyme activities showed dramatic responses to consortium application (Table 5), with acid phosphatase increasing from 42 to 78 µmol g^{-1} h^{-1} and dehydrogenase increasing by 62% (Chen *et al*., 2023) [22].

Table 5: Soil Biological Indicators: Microbial Biomass, Diversity, Enzyme Activities, and Rhizosphere Colonization

Indicator	Control	T2 (10^8)	T3 (10^9)	T4 (10^{10})	T5 (+BC)
Microbial biomass C (µg g^{-1})	187	224	248	273	267
Bacterial 16S rRNA ($\times 10^8$ g^{-1})	4.2	6.8	8.4	11.2	10.8
Fungal ITS ($\times 10^6$ g^{-1})	3.1	4.2	5.6	7.4	7.1
Shannon diversity index	6.84	7.12	7.38	7.62	7.56
Simpson diversity index	0.92	0.94	0.95	0.96	0.96
Acid phosphatase (µmol g^{-1} h^{-1})	42	58	70	78	76
Dehydrogenase (µmol g^{-1} $24h^{-1}$)	48	66	78	78	74
Cellulase (mg g^{-1} h^{-1})	2.4	3.2	4.1	4.8	4.6
Rhizosphere colonization (%)	8	38	62	82	79
AMF root colonization (%)	35	42	58	72	76

3.3. Plant Growth and Yield Performance Under Different Climate Scenarios

Plant height, dry biomass, and yield were significantly enhanced by consortium application (Table 6). Under normal conditions, grain yield increased from 6.14 t ha^{-1} (control) to 6.82 t ha^{-1} in T4 treatments (11.1% increase). Under drought stress, yield increased from 4.28 to 5.78 t ha^{-1} (35.0% increase). Under combined heat and drought stress, yields increased from 3.62 to 5.28 t ha^{-1} (45.9% increase), demonstrating remarkable stress mitigation (Panda *et al*., 2022) [23]. Water Use Efficiency improved from 1.84 kg m^{-3} (control) to 2.36 kg m^{-3} (T4) under drought (28.3% increase). Nutrient Use Efficiency for nitrogen improved by 23.7% (Panda *et al*., 2022) [23].

Table 6: Plant Growth, Physiological, and Yield-Related Characteristics Under Different Climate Treatments.

Parameter	Control	T2 (10^8)	T3 (10^9)	T4 (10^{10})	T5 (+BC)
Plant height normal (cm)	92.4	98.2	102.6	103.8	102.4
Plant height drought (cm)	78.6	88.4	94.2	97.2	95.8
Plant height heat+drought (cm)	68.4	81.2	89.6	87.8	86.4
Dry biomass normal (g m^{-2})	582	648	714	782	756
Dry biomass drought (g m^{-2})	412	502	584	625	612
Dry biomass heat+drought (g m^{-2})	298	382	456	562	548
Grain yield normal (t ha^{-1})	6.14	6.48	6.72	6.82	6.78
Grain yield drought (t ha^{-1})	4.28	4.98	5.42	5.78	5.64
Grain yield heat+drought (t ha^{-1})	3.62	4.28	4.84	5.28	5.14
Harvest index	0.42	0.44	0.45	0.46	0.45
WUE drought (kg m^{-3})	1.84	2.12	2.28	2.36	2.32
NUE normal (kg grain kg^{-1} N)	52.4	56.8	61.2	64.8	63.4

3.4. Stress Physiology and Tolerance Mechanisms

Consortium-treated plants accumulated significantly higher osmolytes under stress conditions. Leaf proline content increased from 2.1 µmol g^{-1} fresh weight (control, drought) to 4.8 µmol g^{-1} in T4 treatments (128.6% increase). Antioxidant enzyme activities were substantially elevated: superoxide

dismutase increased by 52–68% under stress, catalase by 38–54%, and ascorbate peroxidase by 61–78%. Leaf relative water content remained higher in consortium-treated plants (74–78%) versus controls (62–68%) under drought stress (Johnson *et al*., 2024) [24]. Membrane stability indices were significantly better in consortium treatments (63–71%) versus controls (41–52%) under heat stress, indicating superior membrane protection (Johnson *et al*., 2024) [24].

Table 7: Functional Analysis of Microbial-Mediated Nutrient Cycling and Stress Mitigation Mechanisms

Function/Mechanism	Key Enzyme/Gene	Product/Outcome	Field Impact
N-cycling (Azospirillum, Bacillus)	Nitrogenase (nifH)	Bioavailable NH_4^+, NO_3^-	69% increase in available N
P-cycling (B. megaterium, P. fluorescens)	Phosphatase enzymes	PO_4^{3-} from mineral reserves	133% increase in available P
K-cycling (B. mucilaginosus)	Mineral weathering enzymes	K^+ from feldspars, micas	57% increase in available K
Mycorrhizal symbiosis (R. irregularis)	Nutrient transporters	Extended root surface area	Root colonization 72% of cortex
Stress tolerance (all members)	ACC deaminase, IAA, VOCs	Osmolytes, antioxidants	35–46% yield increase under stress
Organic matter decomposition	Cellulase, protease, lipase	Available C, N, P sources	62% increase in dehydrogenase activity

Table 8: Correlation Analysis: Relationships Between Microbial Diversity, Soil Health Indicators, and Wheat Productivity

Variables	r	P-value	Strength	Interpretation
Microbial biomass vs. Grain yield	0.84	<0.001	Strong positive	Active microbiota drives yield
Shannon diversity vs. Grain yield	0.78	<0.001	Strong positive	Functional diversity supports productivity
Phosphatase activity vs. Available P	0.92	<0.001	Very strong	Microbial P-solubilization critical
Rhizosphere colonization vs. NUE	0.86	<0.001	Strong positive	Root colonization enhances uptake
Enzyme activity vs. Soil C	0.81	<0.001	Strong positive	Active microbes enhance C-cycling
Water-holding capacity vs. WUE	0.74	<0.001	Strong positive	Soil structure improves water retention
Osmolyte content vs. Stress tolerance	0.79	<0.001	Strong positive	Osmolytes indicate stress response
AMF colonization vs. Root length	0.88	<0.001	Very strong	Mycorrhizal symbiosis extends roots

Table 9: Comparative Summary of International Studies on Soil Microbiome Engineering and Synthetic Microbial Consortia in Cereal Crops

Reference	Crop	Consortium	Stress	Yield Increase	Key Finding
Chen *et al*. 2023	Wheat	3-member (N, P, AMF)	Drought	18–24	Single-stress simpler approach
Panda & Kumar 2022	Maize	5-member consortium	Heat stress	22–28	Multi-member advantage noted
Johnson *et al*. 2024	Wheat	7-member consortium	Drought+Heat	35–46	Complex stress enhanced effect
Sharma *et al*. 2023	Rice	4-member consortium	Flooding stress	16–21	Crop-specific response patterns
Present study	Wheat	7-member consortium	Drought+Heat	28–46	Optimized consortium effectiveness

4. Discussion

The present study demonstrates that engineered synthetic microbial consortia substantially enhance wheat productivity, soil health, and climate resilience through multiple integrated mechanisms. Our findings align with and extend recent literature on microbiome engineering (Berendsen *et al*., 2012) [25] (Kharabian Masouleh *et al*., 2020) [26]. The establishment and persistence of consortium members in the rhizosphere (72–85% relative abundance) exceeded expectations and demonstrated functional compatibility. The preferential survival of nitrogen-fixing bacteria and mycorrhizal fungi under water-limited conditions suggests that consortium design inadvertently selected for drought-tolerant species (Lupatini *et al*., 2017) [27]. This has important implications for developing region-specific consortia adapted to projected future climates.

Soil enzyme activities increased dramatically following consortium application, reflecting both increased biomass of hydrolytic microorganisms and enhanced metabolic activity. The 62% increase in dehydrogenase activity strongly indicates enhanced overall soil respiration and organic matter decomposition, consistent with literature demonstrating that PGPM enhance carbon cycling (Zhou *et al*., 2021) [9]. The substantial increases in phosphatase and other nutrient-cycling enzymes directly mediated the observed improvements in available phosphorus and potassium.

The extraordinary yield increases under stress (35–46%) merit careful interpretation. These gains represent genuine rescue of stress-induced yield depression rather than yield maximization above optimal conditions. Under combined heat and drought stress, control yields declined to 3.62 t ha^{-1} (42% below normal conditions), whereas consortium-treated yields declined only to 5.28 t ha^{-1} (23% below normal), demonstrating substantial stress mitigation (Johnson *et al*., 2024) [24].

The mechanisms underlying stress tolerance enhancement include enhanced osmolyte accumulation—particularly proline doubling under drought—likely facilitated through consortium-mediated enhancement of abscisic acid signaling and provision of amino acid precursors (Kumar and Verma, 2018) [11]. The sustained antioxidant enzyme activity under stress probably reflects improved electron transport efficiency and provision of essential micronutrient cofactors.

Superior maintenance of leaf relative water content in consortium-treated plants under drought (74–78% versus 62–68%) likely reflects improvements in both root system architecture and soil water-holding capacity. Enhanced root length density (32–48% increase) provides greater surface area for water acquisition, while consortium-driven improvements in soil structure increase available water capacity in upper soil profiles (Reinhardt *et al*., 2022) [6].

Future research should include multi-location trials across diverse soils and climates, longer-term studies (5–10 years) tracking microbiome stability and yield sustainability, investigation of disease suppression effects, mechanistic studies using controlled environments, and economic viability assessments. These directions will strengthen evidence for

practical adoption and regional adaptation of microbiome-based agricultural interventions.

5. Conclusion

Soil microbiome engineering with the use of synthetic microbial consortia is a viable option for improving wheat production outputs and climate change resilience. The seven-member engineered consortium, established in the rhizosphere, improved microbial processes, as well as soils' physical properties, which led to a considerable increase in grain productivity both under normal and stressing conditions, as confirmed by increases of 35–46% of the yield in stress conditions and 22–28% and 18–24% increases in two efficiency parameters: water and nutrients. The mechanisms of operation can be explained by improvements in nutrient cycling, root system development, and antioxidant defense and osmotic regulation. For researchers, this means that they should invest more in microbiome research and development of local consortia. endorses initiatives for promoting sustainable agriculture adaptation in climate-vulnerable area. Top recommendations include: (1) forming local consortia for different wheat-growing regions and soil varieties; (2) incorporating microbiome management with climate-smart agricultural practices; (3) making investments in the infrastructure for regional inoculum production; (4) creating farmer training system; and (5) supporting policies like certification systems and subsidies for early adopters. Microbiome-based methods are an important part of strategies for implementing sustainable, resilient and efficient agricultural systems capable for meeting demands for food security in the future.

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