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Microbial Biomass Carbon, Activity of Soil Enzymes, Nutrient Availability, Root Growth, and Total Biomass Production in Wheat Cultivars under Variable Irrigation and Nutrient Management

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Abstract: Intensive mono-cropping without a balanced supply of nutrients and declining water resources are degrading soil health, and as a consequence, agriculture production is becoming unsustainable and causing environmental degradation. The field experiment was conducted during *Rabi* season to assess the effect of an irrigation schedule, nutrient management, and wheat (*Triticum aestivum* L.) varieties on soil microbial biomass carbon (SMBC) and soil enzymes activities. Two nutrient levels, recommended rate of chemical fertilizer (RDF) and 50% RDF + 50% recommended dose of nitrogen (RDN) through farmyard manure (FYM) designated as Integrated Nutrient Sources (INS), and three irrigations levels, one irrigation at crown root initiation (CRI), two irrigations at CRI and flowering stages, and five irrigations at all main stages of the crop (CRI, tillering, jointing, flowering, and grain filling) were allocated to main-plots while four varieties of wheat, HD 2967, WR 544, HD 2987, and HD 2932, were allocated to sub-plots. The results revealed that SMBC and activities of dehydrogenase, alkaline phosphatase enzymes, and acid phosphatase were higher under restricted irrigation (irrigation at CRI stage) than other irrigation schedules. SMBC, dehydrogenase, acid phosphatase, and alkaline phosphatase activities were 73.0 µg g soil^{-1}, 86.0 µg TPF g soil^{-1}d^{-1}, 39.6 µg PNP g soil^{-1} h^{1}, and 81.8 µg PNP g^{-1} soil h^{-1}, respectively, with the use of INS that was higher than RDF. Root weight and root volume followed a similar pattern. Applying single irrigation at CRI left behind the maximum available nitrogen (166.4 kg ha^{-1}) in soil compared to other irrigation schedules and it was highest (149.31 kg ha^{-1}) with the use of INS. Moreover, total organic carbon (TOC) was 0.44 and 0.43% higher with irrigation at CRI stages and the use of INS, respectively. The INS with single irrigation at the CRI stage is important to improve the root growth, SMBC dehydrogenase, alkaline phosphatase, and acid phosphatase enzyme activity in the wheat production system.

Keywords: microbial biomass carbon; irrigation schedules; nutrient management; organic carbon; soil enzymes; biomaterials

1. Introduction

Wheat constitutes the major staple food crop for people around the world. India is a privileged country in wheat production and is considered as the second-largest producer of wheat in the globe, registering a historic wheat production of 97.44 million tons (Mt) during 2016–2017 with an average productivity of 3.17 t ha^{-1}. The rice-wheat is a major growing system in the countries of South-Asia, which grows about 13.5 million hectares (Mha), more than three-quarters of which are in India. The adoption of high yielding and the use of fertilizer-responsive varieties led to the intensive application of fertilizers. The continuation of the rice-wheat growing system on the same land for several decades has resulted in a noticeable reduction of soil organic matter (SOM), soil fertility, and eventually reduced soil productivity. Cereal-based cropping systems are very exhaustive, especially concerning nutrients [1], and thus, cause mining of soil nutrients.

Nutrient imbalance in rice-wheat cropping system (RWCS) is escalating especially for major nutrients—nitrogen (N), phosphorous (P), potassium (K), and sulfur (S) in the soil and thus, deterioration in soil quality [2,3]. The SOM is considered as the source of energy for diverse soil micro-organisms. Low organic matter availability under RWCS limits microbial activity that in turn affects nutrient availability and crop performance. The assessment of soil enzyme activity may provide valuable information on the main reactions that contribute in slowing decomposition of SOM and nutrients transformation in the soil [4]. Soil quality is strongly associated with soil enzymes because of their relationship to soil biology and ease of measurement [5]. The assessment of the activity of soil enzymes is very much necessary to know the soil microbial activity concerning the cropping system, moisture, and nutrient levels [6]. The root:shoot (R/S) ratio has an important effect on the ecological succession because species follow different strategies for the vegetative growth for light or root growth to reach sufficient water and nutrients. It is critical to understand the whole plant complexity on the levels of roots and shoot. This comprehensive approach may assist in the accurate interpretations of experimental findings.

Agricultural practices that enhance soil properties have been receiving more attention from researchers across the globe. Any change in the practices of soil management as well as land utilization may alter the activities of soil enzymes [7]. Root exudates' qualitative and quantitative composition is determined by plant species, cultivar, growth stage, and different environmental factors, such as temperature, pH, soil type, and availability of micro-organisms [8,9]. In wheat, deficit irrigation (three irrigations) enhanced soil respiration, soil dehydrogenase activity, and SMBC by 79, 8.18, and 27.17%, respectively, over the recommended five irrigations [10]; however, such effects are location specific. Water regime is a critical factor in production, and irrigation will also enhance the production in the dry season or under water-stressed conditions as found in a previous investigation on wheat associating between irrigation and productivity [11]. Declining soil fertility is a big apprehension for agriculture sustainability, and most of the time the researchers' focus is/was on the effect of nutrient and irrigation on crop yield, while fewer studies were done to assess its effect on soil health. We hypothesized that irrigation schedule and nutrient management will have a beneficial effect on improving soil microbial activity, nutrient availability, and root characteristics of wheat crop. The study offers a unique way to assess soil biological behavior under varying irrigation regimes and will provide good information on the interaction between soil biology and irrigation management. This information can then be used for soil health management particularly under irrigation water-stress conditions. Therefore, in the current study, an attempt was made to explore the impact of nutrient management, irrigation scheduling, and wheat varieties on the activity of different enzymes in the soil as well as soil health parameters.

2. Materials and Method

2.1. Study Area

Field experiments were conducted during the winter seasons of 2011–2012 and 2012–2013 at the Indian Agricultural Research Institute, New Delhi, (28.4° N, 77.1° E, and 228.6 m above mean sea level). The field soil was sandy clay loam, neutral in reaction (pH 7.9), low in available N (168.3 kg ha^{-1}) and organic carbon (0.38%), medium in available P (11.9 kg ha^{-1}), available K (241.5 kg ha^{-1}), and DTPA-extractable available Zn (0.68 mg kg $soil^{-1}$).

2.2. Treatments and Crop Management

The experimental design was a split-split plot design replicated thrice (Figure S1). Main-plot treatments consisted of 2 nutrient levels, *viz.* 120:60:40 N, P_2O_5, and K_2O kg ha^{-1}—a recommended dose of chemical fertilizer (RDF) and 50% RDN + 60 kg N (50% RDN) through FYM-integrated nutrient sources (INS), and 3 levels of irrigation, *viz.* 1 irrigation at crown root initiation (CRI) stage, 2 irrigations at CRI and flowering stages, and 5 irrigations at all critical stages of the crop (CRI, tillering, jointing, flowering, and grain filling stage). Sub-plot treatments consisted of 4 wheat varieties, *viz.* HD 2967, WR 544, HD 2987, and HD 2932. The calendar of operation is mentioned in Table S1. The half dose of N through DAP and urea and the full dose of P_2O_5 and K_2O through DAP and MOP were uniformly incorporated in the soil at the sowing. The second half of N was applied in two equal doses (at CRI and the boot-leaf stage). The FYM was applied a week before sowing as per treatment. The FYM contained 0.5% N, 0.2% available P_2O_5, and 0.5% of available K_2O. The NPK doses were adjusted by subtracting the amounts of nutrients coming from FYM, and the remaining doses were supplied as urea, di-ammonium phosphate, and muriate of potash, respectively.

2.3. Wheat Varieties

HD 2967 (Pusa Sindhu Ganga): The plant height ranges from 82 to 108 cm with a mean height of 96 cm. This variety has a semi-spreading growth habit with profuse tillering. The leaves are green, waxy, semi-erect, tapering, dull-white colored, non-pubescent glumes, and dense ear. The grains are ovate shape, amber-colored, hard, lustrous, and have an approximately 1000-grain weight of 40 g. This variety matures in 129–143 days and produces 5.04 t ha^{-1} grain.

HD 2932: This variety is recommended for cultivation in different states of India. The plant height of this variety is 84 cm. The test weight is 43 g. It takes 109 days to mature. The minimum yield of the variety is 4.17 t ha^{-1}.

HD 2987 (Pusa Bahar): It is also known as Pusa Bahar and suitable for timely sown rainfed conditions. Under 2–3 irrigations, it gives 4.0–4.5 t ha^{-1} within 115 days.

WR 544 (Pusa Gold): It is an early maturing (126–134 days) variety with a plant height of 80–90 cm. The yield potential of the variety is about 3.1–3.5 t ha^{-1}.

2.4. Data Collection and Lab Analysis

At the flowering stage, the dry root weight (DRW) and root volume (RV) were determined in two representative hills from the sampling rows by separate dugout from a depth of 20 cm (including soil mass from root zone). The roots and the attached soil mass were placed in a plastic bag with fine mesh and subjected to running water. The clean roots were cut-off from the attached shoots, then each plant root was placed in a graduated measuring cylinder filled with distilled water. For analyzing N status in soil, soil sampling was done after crop harvest. The increase in water level in the cylinder was recorded as root volume [12–14]. The roots and shoots were dried in an oven at 70 ± 2 °C until reaching a constant recorded weight. The root:shoot ratio (RSR) was determined by dividing DRW by shoot dry weight [12]. The dehydrogenase enzyme was estimated following Klein et al. [15]. Alkaline phosphatase and acid phosphatase activities were measured colorimetrically by spectrophotometer [16]. The SMBC was determined by a chloroform fumigation–extraction

method [17]. The soil samples were collected at a depth of 0–15 cm before starting the experiment and at the end of the experiment. The organic carbon was determined by wet digestion [18]. The available nitrogen was determined by alkaline permanganate (KMnO4) procedure [19]. The irrigation water quantity was calculated using volumetric flow value of Parshall flumes and multiplied by the time required to irrigate a plot.

2.5. Statistical Analysis

The data of the different parameters were subjected to the analysis of variance (ANOVA) for the split-split plot design with the help of MSTAT-C software in collaboration with King Saud and Princess Nourah bint Abdulrahman Universities. The results are expressed at a 5% level of significance (p = 0.05). The principal component analysis (PCA) was also performed to group nine soil quality indicators and root characteristics into uncorrelated statistical factors based on their correlation structure. Further discriminant analysis was used to identify the discriminating statistical factor(s) that appeared most differentiating among the different factors (nutrient, irrigation, and varieties).

3. Results

3.1. Root Parameters and Total Biological Yield

The effect of irrigation schedules, nutrient management, and varieties on root length, root volume, root dry weight, and total biological yield (TBY) of wheat was found significant. A significantly higher root volume was observed when wheat was irrigated only once at the CRI stage compared to irrigations given at all critical stages during both seasons (Table 1). The use of INS resulted in significantly higher root volume than RDF. The minimum root volume was observed in HD 2932. Root dry weight was significantly higher with irrigation applied at the CRI stage than other irrigation schedules (Table 2). A significantly higher root dry weight was recorded from INS compared to RDF. Among different varieties, the root dry weight of HD 2987 was higher than other varieties and the minimum root dry weight was observed in HD 2932. Root length was significantly higher with irrigation at the CRI stage over other irrigation schedules. Root length was minimum when irrigations were applied at all critical stages (Table 3). Nutrient levels could not influence root length during both years. However, a significantly higher root length was recorded in HD 2987 than other varieties and the lower root length was observed in HD 2932.

The TBY was significantly higher in the plots irrigated at all critical growth stages than other plots. Among the varieties, HD 2987 gave significantly higher TBY than WR 544 and HD 2932 (Table 4). Among different interactions, HD 2967 produced the highest TBY with 100% RDF under assured irrigation followed by the same variety provided with INS and assured irrigation; both were significantly superior to other combinations. Under limited irrigated conditions, HD 2987 produced significantly higher TBY than other varieties applied with INS and irrigated twice at CRI and flowering stages.

Table 1. Interaction effect of irrigations schedules, nutrient levels, and varieties on root volume (cm^{-3}) at 50% flowering stage.

Varieties	IR1		IR2		IR5		Mean
	RDF	INS	RDF	INS	RDF	INS	
HD 2967	5.6	5.7	4.3	4.6	3.9	4.4	4.7
WR 544	4.8	5.4	4.2	4.5	3.8	4.0	4.5
HD 2987	4.7	5.4	4.7	5.0	4.1	4.4	4.7
HD 2932	4.7	4.9	4.0	4.5	4.1	4.2	4.4
Mean	4.9	5.4	4.3	4.7	4.0	4.2	
Mean Irrigation	5.2		4.5		4.1		

Table 1. *Cont.*

Varieties	IR1		IR2		IR5		Mean
	RDF	INS	RDF	INS	RDF	INS	
Mean Nutrient Levels		RDF		INS			
		4.4		4.8			
		S.Em. ±		CD (p = 0.05)			
IR × NL		0.06		0.20			
IR × V		0.06		0.16			
NL× V		0.05		0.13			
IR × NL× V		0.08		0.22			

Note: IR1—crown root initiation stage (CRI), IR2—CRI and flowering stages, IR3—CRI, tillering, jointing, flowering, and grain filling stage, RDF—recommended dose of fertilizer (RDF), INS—50% RDN + 60 kg N (50% RDN) through FYM-integrated nutrient sources, IR—Irrigation schedules, NL—Nutrient levels, and V—Varieties.

Table 2. Interaction effect of irrigation schedules and nutrient management levels and varieties on root length 50% flowering stage (cm).

Varieties	IR1		IR2		IR5		Mean
	RDF	INS	RDF	INS	RDF	INS	
HD 2967	250.5	254.0	216.3	209.6	203.8	204.9	223.2
WR 544	226.2	247.5	201.0	211.2	185.7	189.0	210.1
HD 2987	242.0	227.0	232.3	221.0	206.1	200.0	221.4
HD 2932	230.2	218.0	206.1	203.9	191.7	206.0	209.3
Mean	237.2	236.6	213.9	211.4	196.8	200.0	
Mean Irrigation	236.9		212.7		198.4		
Mean Nutrient Levels		RDF		INS			
		216.0		216.0			
		S.Em. ±		CD (p = 0.05)			
IR × NL		2.29		7.21			
IR × V		1.6		4.7			
NL× V		1.3		3.8			
IR × NL× V		2.3		6.6			

Note: IR1—crown root initiation stage (CRI), IR2—CRI and flowering stages, IR3—CRI, tillering, jointing, flowering, and grain filling stage, RDF—recommended dose of fertilizer, INS—50% RDN + 60 kg N (50% RDN) through FYM—integrated nutrient sources, IR—Irrigation schedules, NL—Nutrient levels, and V—Varieties.

Table 3. Interaction effect of irrigation schedules and nutrient management levels and varieties on root weight (mg) 50% flowering stage.

Varieties	IR1		IR2		IR5		Mean
	RDF	INS	RDF	INS	RDF	INS	
HD 2967	323.0	376.9	268.2	307.5	231.1	294.1	300.1
WR 544	300.6	355.5	261.8	303.3	237.7	267.6	287.7
HD 2987	293.9	370.0	295.3	346.8	257.0	303.0	311.0
HD 2932	293.0	327.6	241.4	310.5	245.7	291.7	285.0
Mean	302.6	357.5	266.7	317.0	242.9	289.1	
Mean Irrigation	330.1		291.8		266.0		
Mean Nutrient Levels		RDF		INS			
		270.7		321.2			
		S.Em. ±		CD (p = 0.05)			

Table 3. *Cont.*

Varieties	IR1		IR2		IR5		Mean
	RDF	INS	RDF	INS	RDF	INS	
IR × NL		3.9		12.3			
IR × V		2.7		7.7			
NL× V		2.2		6.3			
IR × NL× V		3.8		10.9			

Note: IR1—crown root initiation stage (CRI), IR2—CRI and flowering stages, IR3—CRI, tillering, jointing, flowering, and grain filling stage, RDF—recommended dose of fertilizer, INS—50% RDN + 60 kg N (50% RDN) through FYM—integrated nutrient sources, IR—Irrigation schedules, NL—Nutrient levels, and V—Varieties.

Table 4. Interaction effect of irrigation schedules, nutrient management levels, and varieties on total biomass yield (tons ha^{-1}).

Varieties	IR1		IR2		IR5		Mean
	RDF	INS	RDF	INS	RDF	INS	
HD 2967	7.9	9.0	9.2	9.1	14.6	14.0	10.6
WR 544	8.4	8.5	8.9	9.1	11.0	10.8	9.4
HD 2987	9.7	10.2	10.8	11.1	12.4	12.1	11.0
HD 2932	8.3	8.6	8.8	9.9	10.9	10.4	9.5
Mean	8.6	9.1	9.4	9.8	12.2	11.8	
Mean Irrigation	8.8		9.6		12.0		
Mean Nutrient Levels	RDF		INS				
	10.1		10.2				
	S.Em. ±		CD (p = 0.05)				
IR × NL	0.3		1.1				
IR × V	0.2		0.6				
NL× V	0.2		0.5				
IR × NL× V	0.3		0.9				

Note: IR1—crown root initiation stage (CRI), IR2—CRI and flowering stages, IR3—CRI, tillering, jointing, flowering, and grain filling stage, RDF—recommended dose of fertilizer, INS—50% RDN + 60 kg N (50% RDN) through FYM—integrated nutrient sources, IR—Irrigation schedules, NL—Nutrient levels, and V—Varieties.

3.2. Soil Enzymes

Significantly higher dehydrogenase activity was recorded in plots irrigated once at the CRI stage than other irrigation schedules (Table 5). Among nutrient levels, INS produced higher dehydrogenase activity than 100% RDF. A significantly higher dehydrogenase was recorded from HD 2987 than other varieties. Among different combinations of irrigation schedules and varieties, a significantly higher dehydrogenase activity was recorded in HD 2967 when irrigated at the CRI stage only compared to other combinations. However, when two irrigations were given first at the CRI stage and second at the flowering stage, the variety HD 2987 recorded a significantly higher dehydrogenase activity than other varieties.

Higher alkaline phosphatase and acid phosphatase activities were found under one irrigation only applied at the CRI stage (IR_1) compared to two other irrigation schedules (Tables 6 and 7). The use of INS led to significantly higher alkaline phosphatase and acid phosphatase activities in the soil over 100% RDF. Among different varieties, the HD 2987 exhibited significantly higher activity of acid and alkaline phosphatase activity. Among different combinations of irrigation schedules, varieties, and nutrient management, significantly higher acid phosphatase and alkaline phosphatase activity were recorded in HD 2967 irrigated only once at the CRI stage and fertilized with INS, compared to any other variety × IR × NL combinations.

Table 5. Interaction effect of nutrient management levels, irrigation schedules, and varieties on dehydrogenase (µg TPF/g soil/d).

Varieties	IR1		IR2		IR5		Mean
	RDF	INS	RDF	INS	RDF	INS	
HD 2967	104.1	104.4	78.3	78.5	76.4	76.7	86.4
WR 544	92.2	92.5	79.6	79.8	73.1	73.4	81.8
HD 2987	97.0	97.4	99.4	99.7	87.7	88.0	94.8
HD 2932	89.0	89.2	81.3	81.7	72.2	71.3	80.8
Mean	95.6	95.9	84.7	84.9	77.4	77.3	
Mean Irrigation	95.7		84.8		77.3		
Mean Nutrient Levels	RDF		INS				
	85.9		86.0				
	S.Em. ±		CD (p = 0.05)				
IR × NL	1.12		3.52				
IR × V	1.1		3.3				
NL× V	0.9		2.7				
IR × NL× V	1.6		4.6				

Note: IR1—crown root initiation stage (CRI), IR2—CRI and flowering stages, IR3—CRI, tillering, jointing, flowering, and grain filling stage, RDF—recommended dose of fertilizer, INS—50% RDN + 60 kg N (50% RDN) through FYM—integrated nutrient sources, IR—Irrigation schedules, NL—Nutrient levels, and V—Varieties.

Table 6. Interaction effect of nutrient management levels, irrigation schedules, and varieties on acid phosphates (µg PNP/g soil/h).

Varieties	IR1		IR2		IR5		Mean
	RDF	INS	RDF	INS	RDF	INS	
HD 2967	38.1	48.2	28.7	36.1	28.2	35.2	35.8
WR 544	34.6	41.8	30.0	36.0	26.8	33.8	33.8
HD 2987	34.9	45.6	37.5	44.9	32.5	40.2	39.2
HD 2932	33.7	40.0	28.4	39.1	25.1	34.8	33.5
Mean	35.3	43.9	31.1	39.0	28.1	36.0	
Mean Irrigation	39.6		35.1		32.1		
Mean Nutrient Levels	RDF		INS				
	31.5		39.6				
	S.Em. ±		CD (p = 0.05)				
IR × NL	0.6		1.9				
IR × V	0.7		1.9				
NL× V	0.5		1.5				
IR × NL× V	0.9		2.6				

Note: IR1—crown root initiation stage (CRI), IR2—CRI and flowering stages, IR3—CRI, tillering, jointing, flowering, and grain filling stage, RDF—recommended dose of fertilizer, INS—50% RDN + 60 kg N (50% RDN) through FYM—integrated nutrient sources, IR—Irrigation schedules, NL—Nutrient levels and V—Varieties.

Table 7. Interaction effect of nutrient management levels, irrigation schedules, and varieties on alkaline phosphates (µg PNP/g soil/h).

Varieties	IR1		IR2		IR5		Mean
	RDF	INS	RDF	INS	RDF	INS	
HD 2967	76.4	100.8	56.7	75.2	54.8	70.6	72.4
WR 544	69.4	87.5	59.2	74.9	52.2	67.8	68.5
HD 2987	69.9	95.3	74.1	93.4	63.1	80.6	79.4
HD 2932	67.6	83.8	56.0	81.3	48.7	69.9	67.9
Mean	70.8	91.8	61.5	81.2	54.7	72.2	
Mean Irrigation	81.3		71.4		63.4		
Mean Nutrient Levels	RDF		INS				
	62.3		81.8				
	S.Em. ±		CD (p = 0.05)				
IR × NL	1.2		3.7				
IR × V	1.3		3.8				
NL× V	1.1		3.1				
IR × NL× V	1.9		5.4				

Note: IR1—crown root initiation stage (CRI), IR2—CRI and flowering stages, IR3—CRI, tillering, jointing, flowering, and grain filling stage, RDF—recommended dose of fertilizer, INS—50% RDN + 60 kg N (50% RDN) through FYM—integrated nutrient sources, IR—Irrigation schedules, NL—Nutrient levels and V—Varieties.

3.3. Total Organic Carbon and Microbial Biomass Carbon

Significantly higher organic carbon (OC) (0.44%) was recorded when the crop was irrigated once at the CRI stage (Table 8). A slightly higher OC was recorded for plots treated with INS than RDF. Among varieties, the HD 2987 showed significantly higher OC (0.46%) than other varieties. Furthermore, variety HD 2987 irrigated twice at CRI and flowering stages and nutrients supplied from INS gave significantly higher OC values than the rest of the variety × IR × NL combinations. SMBC was significantly higher in INS than the 100% RDF applied plot. Irrigation at CRI resulted in significantly higher SMBC than other irrigation schedules (Table 9). Cultivation of HD 2987 variety led to higher SMBC than other varieties. Among different combinations of irrigation schedules and varieties, a significantly higher SMBC was recorded from HD 2967 irrigated once at the CRI stage than other combinations.

Table 8. Interaction effect of nutrient management levels, irrigation schedules, and varieties on soil organic carbon (%).

Varieties	IR1		IR2		IR5		Mean
	RDF	INS	RDF	INS	RDF	INS	
HD 2967	0.39	0.49	0.46	0.33	0.38	0.33	0.40
WR 544	0.41	0.43	0.33	0.40	0.37	0.49	0.41
HD 2987	0.53	0.39	0.49	0.55	0.33	0.45	0.46
HD 2932	0.36	0.51	0.31	0.47	0.42	0.42	0.41
Mean	0.42	0.45	0.40	0.43	0.38	0.42	
Mean Irrigation	0.44		0.42		0.40		
Mean Nutrient Levels	RDF		INS				
	0.40		0.43				

Table 8. *Cont.*

Varieties	IR1		IR2		IR5		Mean
	RDF	INS	RDF	INS	RDF	INS	
		S.Em. ±	CD (p = 0.05)				
IR × NL		0.017	0.05				
IR × V		0.02	0.05				
NL× V		0.014	0.04				
IR × NL× V		0.024	0.07				

Note: IR1—crown root initiation stage (CRI), IR2—CRI and flowering stages, IR3—CRI, tillering, jointing, flowering, and grain filling stage, RDF—recommended dose of fertilizer, INS—50% RDN + 60 kg N (50% RDN) through FYM—integrated nutrient sources, IR—Irrigation schedules, NL—Nutrient levels, and V—Varieties.

Table 9. Interaction effect of nutrient management levels, irrigation schedules, and varieties on microbial biomass carbon (µg/g soil).

Varieties	IR1		IR2		IR5		Mean
	RDF	INS	RDF	INS	RDF	INS	
HD 2967	67.6	89.2	50.2	66.6	48.4	62.4	64.1
WR 544	61.4	77.4	52.4	66.3	46.1	60.0	60.6
HD 2987	61.9	84.3	65.5	82.6	55.9	71.3	70.3
HD 2932	59.8	74.1	49.6	72.0	43.1	69.9	61.4
Mean	62.7	81.2	54.4	71.9	48.4	65.9	
Mean Irrigation	72.0		63.1		57.1		
Mean Nutrient Levels		RDF		INS			
		55.1		73.0			
		S.Em. ±	CD (p = 0.05)				
IR × NL		1.1	3.3				
IR × V		1.2	3.4				
NL× V		1.0	2.8				
IR × NL× V		1.7	4.8				

Note: IR1—crown root initiation stage (CRI), IR2—CRI and flowering stages, IR3—CRI, tillering, jointing, flowering, and grain filling stage, RDF—recommended dose of fertilizer, INS—50% RDN + 60 kg N (50% RDN) through FYM—integrated nutrient sources, IR—Irrigation schedules, NL—Nutrient levels, and V—Varieties.

3.4. Soil Available Nitrogen

Soil available N was significantly higher when the wheat crops were irrigated once at the CRI stage than other irrigation schedules and minimum N availability was observed for plots that were irrigated five times at all critical stages (Table 10). The plots treated with INS showed 28.2% higher available N in soil compared to RDF.

Table 10. Interaction effect of nutrient management levels, irrigation schedules, and varieties on available nitrogen (kg/ha).

Varieties	IR1		IR2		IR5		Mean
	RDF	INS	RDF	INS	RDF	INS	
HD 2967	181.46	210.11	116.81	149.28	110.24	152.65	153.42
WR 544	127.69	189.83	104.70	123.30	92.01	98.67	122.70
HD 2987	127.12	212.21	129.20	129.27	107.65	135.50	140.16
HD 2932	121.30	161.46	118.77	137.54	60.89	91.93	115.31
Mean	139.39	193.40	117.37	134.85	92.70	119.69	
Mean Irrigation	166.40		126.11		106.19		

Table 10. *Cont.*

Varieties	IR1		IR2		IR5		Mean
	RDF	INS	RDF	INS	RDF	INS	
Mean Nutrient Levels		RDF		INS			
		116.49		149.31			
		S.Em. ±		CD (p = 0.05)			
IR × NL		2.58		8.12			
IR × V		2.29		6.57			
NL× V		1.87		5.37			
IR × NL× V		3.24		9.30			

Note: IR1—crown root initiation stage (CRI), IR2—CRI and flowering stages, IR3—CRI, tillering, jointing, flowering, and grain filling stage, RDF—recommended dose of fertilizer, INS—50% RDN + 60 kg N (50% RDN) through FYM—integrated nutrient sources, IR—Irrigation schedules, NL—Nutrient levels, and V—Varieties.

3.5. Discriminant Analysis

PCA was performed to describe the overall pattern of sensitivity of all measured soil parameters and to extract the most variable parameter(s) out of it based on factor loadings from each principal component (PC). In this case, three PCs were retained for interpretation having eigenvalues greater than 1.0 accounted for 100% variation (Table 11). PC1, which contributed 71.3% variation, screened soil DHA because of the highest loading (0.93). Similarly, in PC2 (16.8% variation) only root length was retained after excluding other indicators. PC3 (11.7% variation) screened SOC as the most sensitive parameters among all showing maximum factor loadings.

Table 11. Principal component and loading related to soil quality indicators and root characteristics.

Indicators	Principal Components		
	PC1	**PC2**	**PC3**
Eigen value	6.424	1.518	1.057
Variability %	71.380	16.870	11.750
RV	0.388	**0.891**	0.238
RL	0.182	0.824	0.537
RDW	0.633	0.773	−0.042
DHA	**0.930**	0.234	0.284
Ac.P	0.926	0.266	0.266
Al.P	0.928	0.265	0.263
SOC	−0.068	−0.368	**−0.928**
SMBC	0.829	0.559	0.003
N	0.415	0.002	0.910

Values representing the highest component loading are depicted with bold font in each principal component. RV—root volume, RL—root length, RDW—root dry weight, DHA—dehydrogenase, Ac.P—acid phosphatase, Al.P—alkaline phosphatase, SOC—soil organic carbon, SMBC—soil microbial biomass carbon, N—available N.

Discriminant analysis with different soil quality indicators and root characteristics had generated two significant canonical discriminant functions (function 1 and 2) contributing 92.6 and 6.9% of the total variation, respectively (Figure 1. The results revealed that nutrient management practice (INS), varieties (WR 544 and HD 2987), and irrigation practices (IR1 and IR2) have shown a positive effect on soil quality indicators and root characteristics.

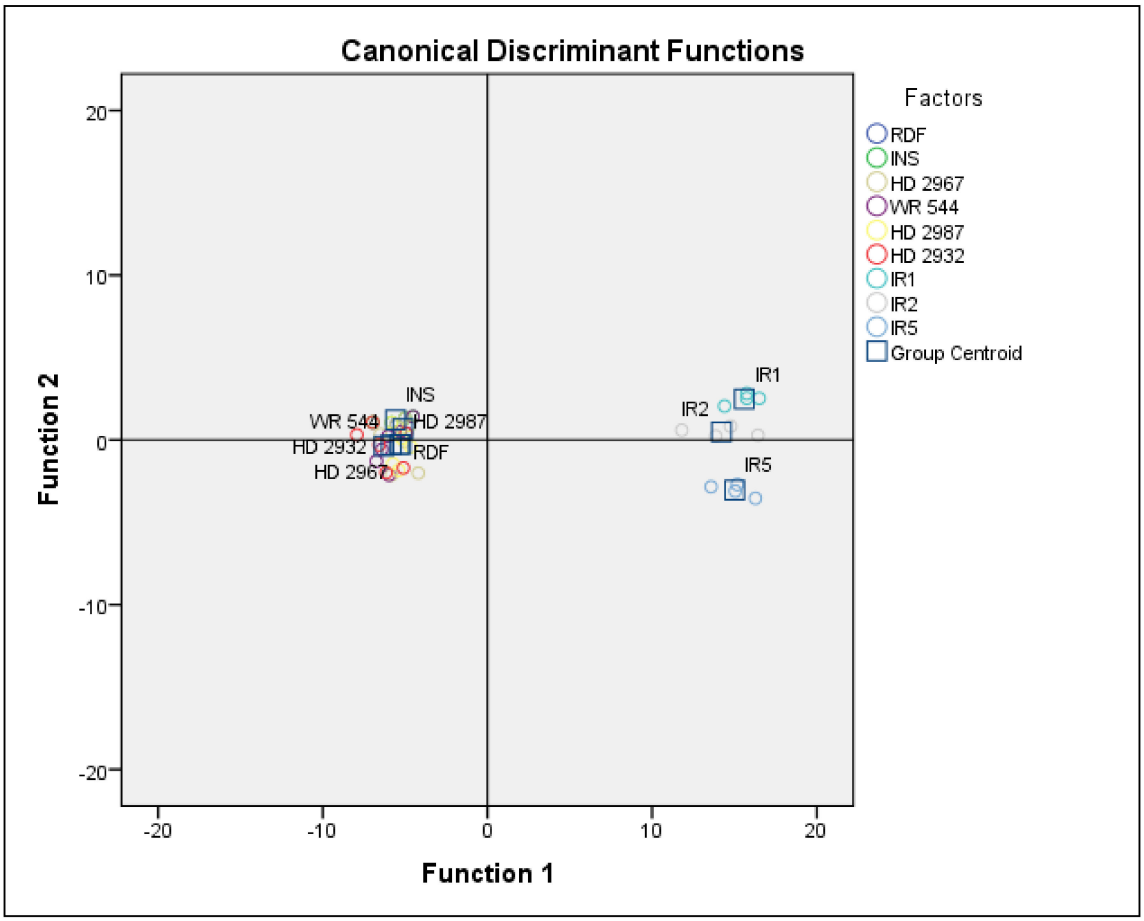


Figure 1. Bi-plot of canonical discriminant functions for separation of different factor effects.

4. Discussion

4.1. Soil Microbial Activity

All the soil enzymes, SMBC, and OC increased with the increased root dry weight of all wheat varieties. Irrespective of varieties, significantly higher acid phosphatase, dehydrogenase, alkaline phosphatase activities, and SMBC were recorded when the wheat crop was irrigated once at the CRI stage compared to other irrigation schedules during both seasons (Tables 4–6 and 8). This might be due to a higher rhizo-deposition under deficit-irrigation applied plots. Rhizo-deposition forms an important source of C and nutrients for soil microbes. Higher rhizo-deposition by the plants in response to drought is primarily because the up-regulation of this operation may offset the main negative impacts on plants. Rhizo-deposition is higher under dry conditions because under such (drought) conditions, plants may need lubrication to assist the roots in moving through the dry soil and to sustain healthy root-soil association [20–22]. Rhizo-deposits are mainly composed of mucilage that has an essential effect on lubrication. Rhizo-deposits might include different substances such as ions (e.g., H^+, OH^-, HCO_3^-), mucilage, sugars, organic acids, amino acids, and enzymes [23]. These rhizo-deposits might be produced in an active or passive manner [24] and may include substances released from dead root tissues [25]. Under drought conditions, there is a tendency for increased rhizo-deposition and increasing root mortality, which also controls cell membrane integrity that causes loss of solutes, which are a source of C and hardly distinguished from the rhizo-deposition increase of carbon [20]. When the root got damaged, less re-absorption of rhizo-deposits led to a further increase in the amount of carbon (C) that is measured in the rhizosphere [20]. Recent studies indicate a large amount of C (2 and 11%) is fixed during photosynthesis that is lost by rhizo-deposition [26].

Between two nutrient levels, INS exhibited significantly higher dehydrogenase, acid phosphatase, alkaline phosphatase, and SMBC over 100% RDF in both seasons, which might be attributed to the effect of FYM that increase the OC and stimulates soil microbial activity; OC acts as a source of energy for the bacterial growth in organically treated plots. It was higher due to the increased microbial biomass [27]. Wheat variety HD 2987 exhibited significantly higher dehydrogenase, acid phosphatase, alkaline phosphatase activities, and SMBC than other varieties. This variety possessing the ability to resist adverse conditions by altering the root:shoot ratio might be a reason for providing higher root surface area for the growth and development of microorganisms. Higher soil enzyme activity may be due to the release of volatile compounds and enzymes by microbial activities under adverse conditions [9]. The interaction effect of irrigation schedules and varieties revealed that significantly higher dehydrogenase, alkaline phosphatase, acid phosphatase activities, and SMBC were measured for HD 2967 with irrigation at CRI stages and in INS than other combinations. This might be due to higher enzyme activity, root length, volume, and dry weight in HD 2967, which got amplified under INS.

Moreover, the genetic characteristics of HD 2987 enabled this variety to perform well under limited irrigation. The higher root parameters like volume, length, and dry matter of this variety might be the reason for higher biomass yield under limited irrigation. Higher root density in the upper soil depth (0–15 cm) resulted from a higher moisture regime due to irrigation at IW:CPE{(irrigation water (IW) and cumulative pan evaporation (CPE)} ratio of 0.90 that eliminated water-stress, thus enabling better root growth into deeper layers in wheat [10]. The enzymatic activity and SMBC were also found higher in its rhizosphere. Higher microbial activities in the rhizosphere facilitate the mineralization of less available soil and applied nutrients and make them available to plant under limited moisture conditions.

4.2. Available Nutrient Status in the Soil at the Harvest

Available N status was higher under deficit-irrigated plots, such as irrigation at the CRI stage compared to the other irrigation timings. The plots irrigated once at the CRI stage produced lower biological yield than the plot irrigated at all critical growth stages, which could be the reason for higher available N in the less irrigated plot because of reduced nutrient uptake [28]. The nutrient transportation from the roots to the shoot is limited due to decreased imbalance in active transport, transpiration rate, and possible membrane permeability, which may result in lower uptake of nutrients by the roots [29–31]. Therefore, drought causes reductions in the availability of different nutrients and reduced nutrient transport in plants in general [30]. Drought minimizes nitrate reductase activity, which affects negatively the N uptake in many species, including lettuce [32], maize [33], and cowpea [34]. Under a water deficit, N deficiency occurs [35], which reduces plant growth and produces chlorosis [36]. A healthy plant has more uptake of nutrients compared to a crop grown under water-stressed conditions. Wheat crops grown under well-irrigated conditions absorbed a greater amount of nutrients and reduced nutrient levels in the soil. Ladha et al. [37] and Tiwari [38] reported a reduction in soil organic matter as well as an increase in major nutrients (N, P, K, and S) and micronutrients (Zn, Fe, and Mn) deficiencies attributed to over mining by cereals.

Nutrient uptake under drought stress could be further enhanced by increasing the root: shoot ratio in drought-stressed environments [39]. The application of 50% RDF + 50% RDN through FYM increased the amount of available N to 100% RDF during both years. This could be explained by the residual effect of decomposed organic manure within the soil following crop harvest [40]. Su et al. [41], Jiang et al. [42], and Dass et al. [43] have also confirmed that FYM application separately or combined with some inorganic fertilizer enhanced the nutrient composition of the soil. Among different varieties, significantly higher available N was recorded after harvest of HD 2967 than other varieties. This might be due to lower nutrients uptakes of this variety.

Under limited irrigation, the HD 2967 had higher root parameters while decreasing water limitation increased the root parameters of HD 2987 and this tendency is true for soil enzyme activities, too. Using mean values of these varieties masks the different plant reactions for drought stress. Several researchers found a positive correlation between organic matter (OM) content and dehydrogenase [44–47]. As the root biomass production enhanced the activity of soil enzymes and also improved SMBC and OC in this study, root biomass production should be given priority because it helps to maintain soil fertility. The N availability showed a positive correlation with the activity of soil enzymes. The high activity of soil enzymes indicates the soil has a high microbial population and it helped the mineralization of organic matter. Similar results were also obtained by several researchers and they found that the activity of dehydrogenase is very important and might be used as an indicator of the microbial activity of soils [48–50] because they are available in all microbes [45,47]. Dehydrogenases are associated with microbial biomass (MB), which may affect the decomposition of organic matter [51]. Under well-irrigated conditions, nutrient availability was exhausted by adequate soil moisture, but under deficit irrigation, microbes played a greater role in making soil nutrients available to plant in fewer quantities.

The discriminant analysis revealed that DHA and SOC are the most distinguishing soil indicators among all other soil attributes (Figure 1) extracted from three principal components. Significantly higher content of SOC and DHA discriminated well both nutrient (INS) and irrigation factors (IR1) systems from the rest of the treatments. INS, IR1, and variety (HD 2987) were further separated from each other due to significant difference in root length. HD 2987 has been found to be associated with the highest DHA, SOC, and root length, which contributed to separate the system from a cluster of other combinations, which were found to be grouped together. The soil quality indicators (DHA, SOC) and root length were found greatly influenced due to irrigation practices, and as the soil water content increases, the soil microbial activity improves. Soil ecology deals with the interaction between inhabitant soil microbes and their surroundings influencing various soil attributes and processes (soil water availability, nutrient cycling, and biodiversity interactions) finally forming the basis for delivering ecosystem services. The results reveal that irrespective of nutrient management and varieties, the irrigation (soil water availability) was very important to improve the soil quality indicators.

5. Conclusions

This study, which evaluated the effect of an irrigation schedule, nutrient management, and cultivar, provides first-time insights on the interaction effects of irrigation and nutrient management on the root characteristics and soil microbial activity. The results revealed that irrigating crop at the CRI stage is very much essential to improve crop root length (236.9 cm), root volume (5.2 cm^3), and root dry weight (330.1 mg) compared to irrigations at all critical stages during both seasons. Furthermore, the higher organic carbon (0.44%), soil available N (166.40 kg N/ha), soil microbial biomass carbon (72.0 μg kg^{-1}), dehydrogenase activity (95.7 μg TPF/g soil/d), and acid phosphatase activities (39.6 μg PNP/g soil/h) were observed under irrigating crop at CRI stage. However, a significantly higher total biomass was noticed with irrigating wheat crop at all critical stages. Likewise, integrated nutrient supply has recorded higher root parameters and soil microbial activity over the recommended dose of fertilizers. Growing variety HD 2987 was found beneficial in improving total biomass yield and root growth, and its performance under limited irrigation conditions was also found satisfactory. The current study confirms that the irrigations at all critical stages might result in a higher yield but lead to nutrient mining. Continuity of such a scenario may lead to a lower residual nutrient in the soil and an overall decline in soil health in the long-run. The reverse trend was found with the reduced irrigation frequency, and even though this practice produced a lower yield, it reduced the mining of nutrients and retained a higher amount of nutrients in the soil. Among nutrient management, integrated nutrient supply (50% RDF + 50% RDN through FYM) was found to be more productive and maintained good soil health and fertility compared to 100%

RDF. Thus, the future focus should not be on exploiting yield potential through the use of 100% chemical fertilizer and advanced agronomic management like assured irrigation, but rather on promoting other practices that enhance and promote rhizosphere function and the biogeochemical cycle for healthy soil.

Supplementary Materials: The following are available online at https://www.mdpi.com/article/10.3390/agronomy11040669/s1, Figure S1: The layout of the field experiment, Table S1: Detail of the field operation.

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