PLANT SCIENCE TODAY
Vol 12(sp4): 01-11
https://doi.org/10.14719/pst.12322

eISSN 2348-1900

RESEARCH ARTICLE

Integrated use of sewage sludge and chemical fertilizers enhances growth, biochemical traits and stress tolerance in the rice-wheat cropping system

Pavan Singh[1], Arvind[2], Aakash[3], Kamlesh Meena[4], Nimit Kumar[5], Basant Kumar Dadrwal[6], Jyotsna Setty[7]*, Y V Singh[8], Hanuman Singh Jatav[9], Shyam Kishor Patel[1] & Amit Kumar[1]

[1]Faculty of Agricultural Sciences, GLA University, Mathura 281 406, Uttar Pradesh, India
[2]Department of Soil Science and Agricultural Chemistry, Narain College, Shikohabad 283 135, Uttar Pradesh, India
[3]Department of Agronomy, Ranjeet Singh Memorial Post Graduate College, Dhampur, Bijnor 246 761, Uttar Pradesh, India
[4]ICAR-Indian Institute of Vegetable Research, Krishi Vigyan Kendra, Malhana, Deoria 274 506, Uttar Pradesh, India
[5]Department of Agricultural Economics, College of Agriculture Sciences, Teerthanker Mahaveer University, Moradabad 244 001, Uttar Pradesh, India
[6]Sri Karan Narendra Agriculture University, Jobner, Jaipur 303 329, Rajasthan, India
[7]Department of Plant Physiology, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi 221 005, Uttar Pradesh, India
[8]Department of Soil Science and Agricultural Chemistry, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi 221 005, Uttar Pradesh, India
[9]Department of Soil Science and Agricultural Chemistry, Sri Karan Narendra Agriculture University, Jobner, Jaipur 303 329, Rajasthan, India

*Correspondence email - settyjyotsna@gmail.com

Received: 17 October 2025; Accepted: 16 December 2025; Available online: Version 1.0: 24 December 2025

Cite this article: Pavan S, Arvind, Aakash, Kamlesh M, Nimit K, Basant KD, Jyotsna S, Singh YV, Hanuman SJ, Shyam KP, Amit K. Integrated use of sewage sludge and chemical fertilizers enhances growth, biochemical traits and stress tolerance in the rice-wheat cropping system. Plant Science Today. 2025;12(sp4):01-11. https://doi.org/10.14719/pst.12322

Abstract

Sewage sludge (SSL), historically regarded as an environmental challenge, is increasingly recognized as a nutrient-rich organic amendment with considerable potential for sustainable agricultural use. This study evaluated the combined effects of SSL and chemical fertilizers (CF) on the growth, physiological performance and biochemical characteristics of rice (*Oryza sativa* L.) and wheat (*Triticum aestivum* L.) cultivated under a rice-wheat cropping system. The experiment, conducted over two consecutive cropping seasons, demonstrated that integrated nutrient management involving SSL and CF significantly improved plant height, tiller number, chlorophyll concentration, protein and starch contents, membrane stability index (MSI) and chlorophyll stability index (CSI). The most pronounced improvements were observed under the treatment comprising 30 t ha^{-1} SSL in combination with 100 % of the recommended dose of fertilizer (RDF). Furthermore, stress-associated metabolites such as proline, phenolics and malondialdehyde (MDA) were significantly lower under SSL-CF treatments, indicating reduced oxidative and abiotic stress and enhanced physiological resilience. These findings reveal that SSL not only supplies essential nutrients but also enhances physiological efficiency and stress tolerance in cereal crops. Nonetheless, the potential risks associated with heavy metal accumulation underscore the need for systemic monitoring and regulated application strategies. Collectively, the study provides robust evidence that the integration of SSL with CF improves soil fertility, maintains crop productivity and supports environmentally sound nutrient management for long-term agricultural sustainability in intensive cereal-based systems.

Keywords: chlorophyll stability index; membrane integrity; rice-wheat cropping system; sewage sludge; sustainable agriculture

Introduction

For centuries, farmers have relied on organic residues to replenish soil fertility and sustain crop productivity. With rapid urbanization and industrialization, however, the generation of municipal SSL, a nutrient-rich by-product of wastewater treatment which has substantially increased worldwide. Traditionally considered a waste material, SSL has gradually shifted into focus as a potential bioresource for sustainable agriculture due to its considerable content of organic matter (OM), macro and micronutrients (1-3). In many countries, applying SSL to croplands is seen as a dual strategy, reducing the burden of waste disposal while enhancing soil fertility and crop yields.

The conversion of wastewater into SSL encompasses a series of mechanical, biological and chemical processes, producing a semi-solid matrix enriched with essential nutrients such as nitrogen, phosphorus, potassium and micronutrients critical for plant development (4). Numerous studies have shown that amendments with sewage SSL enhance soil physical properties, including structure, porosity and water-holding capacity, as well as nutrient availability, thereby promoting improved growth and yield in crops such as wheat (*Triticum aestivum* L.), faba bean (*Vicia faba* L.), spinach (*Spinacia oleracea* L.) and cucumber (*Cucumis sativus* L.) (5-9).

In particular, the synergistic application of SSL and CF has been shown to optimize nutrient supply and promote sustainable productivity in intensive cropping systems, including the rice-wheat rotation prevalent in South Asia (10, 11). Beyond agronomic benefits, SSL use significantly alters plant biochemical responses. Carbohydrates, proteins, lipids, proline and phenolic compounds-key metabolites regulating growth and stress responses-are often influenced by SSL addition. For instance, carbohydrate accumulation, closely tied to nitrogen and phosphorus availability, has been widely observed in SSL-amended soils (12). Similarly, increased proline accumulation under environmental stress has been reported in crops grown with wastewater and sludge treatments, highlighting its role as a biochemical protectant (13).

Phenolic compounds, central to plant defence against abiotic and biotic stresses, also exhibit variability under SSL application (14). Thus, studying the biochemical responses of cereal and vegetable crops to SSL is critical, especially given their direct implications for human and animal consumption. However, the advantages of SSL application must be carefully weighed against environmental risks. Continuous use or excessive application rates may lead to soil acidification, salinity and the accumulation of potentially toxic heavy metals such as Pb, Cr and Hg, which adversely affect soil microbial communities and may bioaccumulate in edible plant parts (13). These risks necessitate the development of region-specific guidelines and monitoring frameworks that account for soil type, climate variability and crop sensitivity to ensure safe and sustainable SSL utilization (15).

Within this context, the rice-wheat cropping system provides a particularly relevant model for studying SSL integration, as it underpins food security for nearly half of the world's population. Intensive cultivation of rice and wheat has led to widespread nutrient depletion and soil degradation, necessitating sustainable soil fertility management strategies. While previous studies have explored the role of SSL in enhancing crop yields, limited research has comprehensively assessed its impact on both growth parameters and biochemical responses in the rice-wheat system.

Therefore, the present study was undertaken to investigate the combined effects of SSL and CF on growth attributes, physiological parameters and biochemical composition of rice and wheat. Specifically, aimed to evaluate plant height, tiller number, chlorophyll, protein, starch, proline, phenols and stress biomarkers such as MDA under integrated nutrient management. By linking agronomic performance with physiological and biochemical responses, this study provides novel insights into the safe and effective use of SSL in cereal-based cropping systems.

Materials and Methods

Experimental site

The experiment was performed at the Agricultural Research Farm of Banaras Hindu University, Varanasi, India (25°19′ N, 83°00′ E; 128.93 m asl), located in the Northern Gangetic Alluvial Plain (Inceptisol). The study comprised two consecutive rice-wheat cropping cycles during 2019-2020 (cycle I: rice and wheat) and 2020-2021 (cycle II: rice and wheat), following four earlier cycles (2015-2019) established under the same experimental layout (Table 1, Fig. 1). The rice cultivar *O. sativa* (hybrid 'Arize 6444') and wheat cultivar *T. aestivum* ('HD 2967') were used consistently throughout the experimental period.

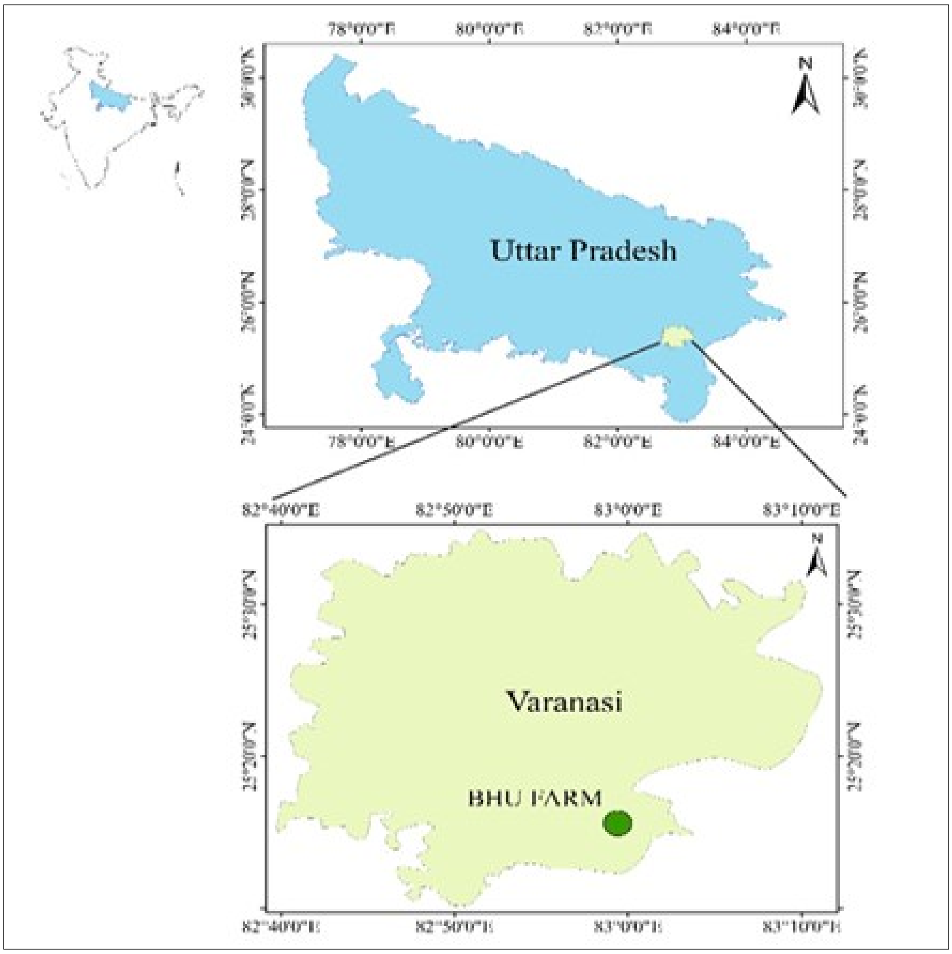

Fig. 1. Location of experimental trial.

Table 1. Cropping history of the experimental field

Year	*Kharif*	*Rabi*
2014-2015	Rice	Wheat
2016-2017	Rice	Wheat
2017-2018	Rice	Wheat
2018-2019	Rice	Wheat
2019-2020	Rice	Wheat
2020-2021	Rice	Wheat

Soil and sewage sludge characteristics

SSL was air-dried, thoroughly homogenized and applied to the experimental field before rice transplanting. Organic carbon was determined using the Walkley-Black wet oxidation method, while total nitrogen was estimated by the Kjeldahl method. The initial soil was alkaline, with a pH of 8.24, low electrical conductivity (0.15 dS m^{-1}) and low organic carbon content (4.60 g kg^{-1}) as well as limited available nitrogen (141.72 kg ha^{-1}). Available phosphorus (17.42 kg ha^{-1}), potassium (132.74 kg ha^{-1}) and sulphur (14.65 mg kg^{-1}) were within moderate levels. Micronutrient concentrations in the soil were 42.65 mg kg^{-1} Fe, 2.17 mg kg^{-1} Cu, 1.02 mg kg^{-1}Zn and 11.41 mg kg^{-1} Mn.

The SSL applied in this study had a slightly acidic pH of 6.6, electrical conductivity of 3.17 dS m^{-1}, organic carbon 8.67 %, total nitrogen 1.76 %, total phosphorus 1.29 % and total potassium 1.15 %. Total micronutrient and heavy metal contents (mg kg^{-1}) in the SSL were Fe 490.27, Cu 240.63, Zn 184.27, Mn 246.08, Cd 7.30, Cr 49.20, Ni 27.43 and Pb 39.53.

Experimental design and treatments

The present study was integrated into an ongoing long-term field experiment initiated during 2015-2016. Treatments were arranged in a randomized block design (RBD) with three replications. Ten treatment combinations comprising the RDF and graded levels of SSL were evaluated (Table 2).

The SSL was applied only during the first year at the time of rice establishment. Basal applications included half of the nitrogen along with the full recommended doses of phosphorus and potassium, while the remaining nitrogen was top-dressed in two equal splits at 30 and 60 days after transplanting or sowing (DAT/DAS).

Table 2. Treatment details of the experiment

Treatments number	Treatment description
T_0	Control
T_1	100 % RDF
T_2	100 % RDF + 20 t ha^{-1} SSL
T_3	100 % RDF + 30 t ha^{-1} SSL
T_4	50 % RDF + 20 t ha^{-1} SSL
T_5	60 % RDF + 20 t ha^{-1} SSL
T_6	70 % RDF + 20 t ha^{-1} SSL
T_7	50 % RDF + 30 t ha^{-1} SSL
T_8	60 % RDF + 30 t ha^{-1} SSL
T_9	70 % RDF + 30 t ha^{-1} SSL

Plant sampling

Fully expanded leaves were sampled from each plot at 30 DAT/ DAS. Leaves were washed thoroughly under tap water, portions were preserved at -20 °C for biochemical analyses (proline, phenols and MDA), while the remaining were oven-dried at 60 °C and ground (< 0.4 mm particle size) for carbohydrate analysis.

Physio-biochemical analyses

Chlorophyll, starch, total soluble protein content, MSI and CSI

Total chlorophyll content was quantified according to the protocol (16). Starch was estimated by anthrone method. Total soluble protein content (TSPC) was estimated using the Lowry method (17). MSI and CSI of leaves were determined (18, 19).

Stress markers (proline, phenolic and MDA)

Proline content was estimated and expressed as µg g^{-1} fresh weight (FW), while total phenols were quantified in shoots (20, 21). MDA content was measured using the thio-barbituric acid (TBA) reactive substance (TBARS), the degree of lipid peroxidation in the shoots was assessed and was determined by the TBA reaction method.

Principal component analysis (PCA), grid correlation matrix, heat map, scatter plot and dose response curve

PCA was used to examine relationships among all parameters in different seasons. Pearson's correlation analysis, performed using Origin Pro 2025b, evaluated correlations between all parameters, ranging from +1 to -1 and visualized with colour coding. Additionally, a heat map (olive green to yellow) was used to display the various growth attributes, physio-biochemical parameters and key stress indicators. Dose response curve and scatter plot with linear relationship between parameters were analysed using Origin Pro and R-studio.

Statistical analysis

Data were subjected to one-way analysis of variance (ANOVA) using OriginPro 2025b and differences among treatments were evaluated for significance using Tukey's test at the 5 % probability level. Heat maps were generated by normalizing the values and applying a colour scale, olive green (100 %, high), light green (50 %, medium) and yellow (1 %, low).

Results

Growth attributes

The integrated application of SSL and CF exerted a significant influence on plant height and tiller production in both rice and wheat across growing seasons (Table 3, Fig. 2, 3). Instead of reiterating individual numerical values, the results are presented in terms of treatment-wise trends. In both crops and seasons, treatments receiving full RDF in combination with SSL consistently outperformed reduced-RDF, indicating a synergistic interaction between organic and inorganic nutrient sources. The tallest plants were recorded under T_3 (100 % RDF + 30 t ha^{-1} SSL), followed closely by T_2 (100 % RDF + 20 t ha^{-1} SSL), while the control (T_0) showed the lowest growth. Where applicable, statistical significance was carefully interpreted in accordance with ANOVA results. In several cases, T_3 was statistically comparable to T_1 (100 % RDF) but remained numerically superior, suggesting enhanced nutrient availability rather than a simple substitution effect.

Tiller number exhibited a trend similar to that observed for plant height. Maximum tillering occurred under integrated SSL-RDF treatments, with T_3 producing the highest tiller density in both crops. Reduced RDF treatments (T_4-T_9) showed a decline in tiller production, indicating that SSL alone was insufficient to fully compensate for reduced mineral fertilizer inputs.

Table 3. Effect of conjoint application of sewage sludge and fertilizer on growth attributes of rice and wheat

Treatments	Plant height (cm)				Tillers per running (m)			
	2019 I-rice	2019-2020 I-wheat	2020 II-rice	2020-2021 II-wheat	2019 I-rice	2019-2020 I-wheat	2020 II-rice	2020-2021 II-wheat
T_0	44.77 ± 2.58d	17.77 ± 0.42e	43.61 ± 1.15e	17 ± 0.65e	20.89 ± 0.25e	32.98 ± 2.23e	20.53 ± 0.85c	30.36 ± 1.78e
T_1	73.13 ± 1.58ab	29.4 ± 2.05bcd	66.67 ± 1.11bc	29.93 ± 2.06bc	53.66 ± 4.02abc	65.77 ± 2.15ab	54.1 ± 2.54a	65.67 ± 2.37ab
T_2	73.33 ± 2.03ab	33.27 ± 2ab	72.31 ± 1.21a	31.87 ± 0.81ab	56.91 ± 0.98ab	67.28 ± 1.85ab	55.15 ± 0.68a	66.24 ± 0.92ab
T_3	78 ± 2.52a	37.27 ± 0.85a	70.16 ± 1.59ab	36.24 ± 1.74a	59.24 ± 1.94a	71.82 ± 1.23a	56.01 ± 0.45a	69.91 ± 1.53a
T_4	62.47 ± 1.13c	23.87 ± 1.01d	60.9 ± 1.18d	22.98 ± 1.24d	44.34 ± 1.22d	54.06 ± 1.2d	42.94 ± 0.79b	52.17 ± 1.3d
T_5	64.37 ± 3.15bc	25.93 ± 1.16d	62.85 ± 0.42cd	25.32 ± 0.19cd	46.42 ± 1.74cd	55.95 ± 1.51cd	44.47 ± 1.2b	54.36 ± 1.1cd
T_6	67 ± 0.46bc	27.3 ± 0.4cd	65.13 ± 1.29bcd	26.63 ± 0.47bcd	49.74 ± 0.5bcd	59.84 ± 1.22bcd	49.28 ± 2.52ab	57.76 ± 1.89cd
T_7	64.23 ± 1.08bc	26.04 ± 0.54cd	63.67 ± 0.5cd	25.54 ± 0.58cd	46.85 ± 0.68cd	59.12 ± 3.06bcd	44.69 ± 1.54b	57.43 ± 1.3cd
T_8	67.47 ± 1.54bc	27.83 ± 0.92bcd	66.23 ± 0.88bcd	27.43 ± 0.5bcd	54.2 ± 1.48abc	61.97 ± 1.15bcd	48.98 ± 2.75ab	60.19 ± 1.2bc
T_9	69.93 ± 0.92abc	31.83 ± 0.88abc	68.18 ± 0.89abc	29.9 ± 1.03bc	55.09 ± 1.52abc	64.42 ± 0.84abc	50.74 ± 0.31ab	61.86 ± 1.55bc

Mean values within the same column having alike alphabets differ non-significantly ($p \leq 0.05$), while different alphabets show a significant difference ($p \leq 0.05$). Mean (±SE) was taken from three replicates for each treatment.Treatments : T0 -without Fertilizer, T1 - 100 % RDF, T2 - 100 % RDF + 20 t ha-1 SSL, T3 - 100 % RDF + 30 t ha-1 SSL, T4-50 % RDF + 20 t ha-1 SSL, T5 -60 % RDF + 20 t ha-1 SSL, T6 - 70 % RDF + 20 t ha-1 SSL, T7 - 50 % RDF + 30 t ha-1 SSL, T8 - 60 % RDF + 30 t ha-1 SSL and T9 - 70 % RDF + 30 t ha-1 SSL.

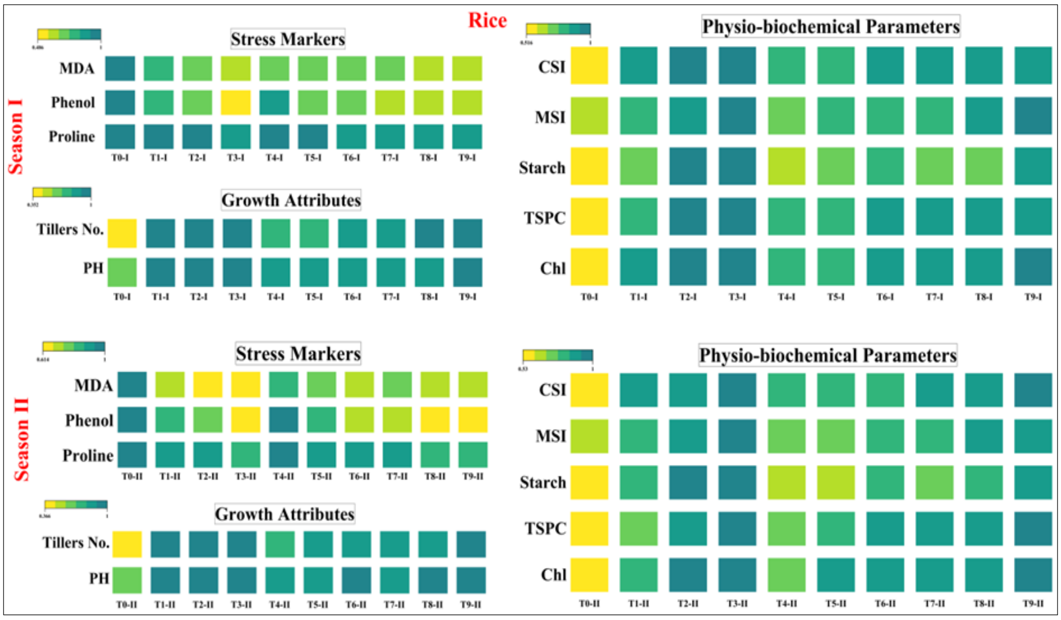

Fig. 2. Heat map of growth attributes, physio-biochemical parameters and stress indicators observed under different treatments in rice crop under two seasons. Olive green is for higher value (100 %), light green is for medium (50 %) and yellow is for the lowest values (1 %). All the data obtained were first normalized to bring the value of the parameters in the range of 0-1 to provide an unbiased colour code. (T_0-I to T_9-I denotes treatments of season I and T_0-II to T_9-II denotes treatments of season II).

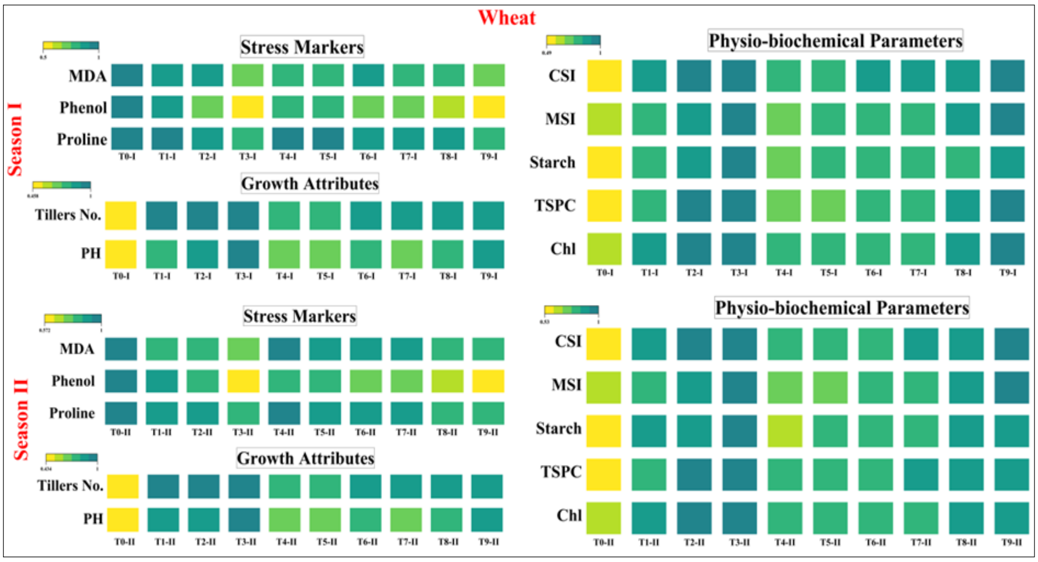

Fig. 3. Heat map of growth attributes, physio-biochemical parameters and stress indicators observed under different treatments in wheat crop under two seasons. Olive green is for higher value (100 %), light green is for medium (50 %) and yellow for the lowest values (1 %). All the data obtained were first normalized to bring the value of the parameters in the range of 0-1 to provide an unbiased colour code. (T_0-I to T_9-I denotes treatments of season I and T_0-II to T_9-II denotes treatments of season II).

Physio-biochemical parameters (Chlorophyll, TSPC, Starch, MSI and CSI)

Integrated nutrient management significantly enhanced chlorophyll concentration, TSPC, starch accumulation, MSI and CSI in both crops (Tables 4, 5, Fig. 2, 3). Chlorophyll content increased markedly under SSL-RDF combinations, with T_3 consistently exhibiting the highest values across seasons. This enhancement reflects improved nitrogen and magnesium availability, essential for chlorophyll biosynthesis, supported by the slow mineralization of organic nitrogen from SSL. Protein accumulation followed a similar trend. Higher TSPC under integrated treatments indicates improved nitrogen assimilation efficiency, resulting from the combined effect of readily available mineral nitrogen and sustained organic nitrogen release from SSL.

Starch content increased significantly under SSL-RDF treatments, particularly T_3 and T_2. The positive linear relationship between starch concentration and tiller number highlights carbohydrate availability as a key physiological driver of tiller

Table 4. Effect of conjoint application of sewage sludge and fertilizer on physio-biochemical parameters of rice and wheat

Treatment	Total chlorophyll content (mg g^{-1} FW)				TSPC (mg g^{-1} FW)			
	2019	2019-2020	2020	2020-2021	2019	2019-2020	2020	2020-2021
	I-rice	I-wheat	II-rice	II-wheat	I-rice	I-wheat	II-rice	II-wheat
T_0	1.02 ± 0.02F	1 ± 0.003f	1 ± 0.02g	0.99 ± 0.015g	7.69 ± 0.01e	8.58 ± 0.19f	7.1 ± 0.04c	8.15 ± 0.06e
T_1	1.58 ± 0.01cd	1.34 ± 0.025c	1.45 ± 0e	1.33 ± 0.024bcd	11.42 ± 0.36cd	12.31 ± 0.11cd	9.83 ± 0.09b	11.66 ± 0.24d
T_2	1.7 ± 0.03b	1.46 ± 0.021b	1.69 ± 0.01b	1.42 ± 0.018ab	13.8 ± 0.33ab	14.69 ± 0.3ab	12.14 ± 0.09ab	13.54 ± 0.04ab
T_3	1.82 ± 0.02a	1.56 ± 0.012ab	1.79 ± 0.01a	1.49 ± 0.022a	14.88 ± 0.26a	15.67 ± 0.1a	13.38 ± 0.1a	14.64 ± 0.32a
T_4	1.39 ± 0.02e	1.2 ± 0.019e	1.35 ± 0.01f	1.14 ± 0.021f	11.63 ± 0.32d	11.19 ± 0.03e	10.17 ± 0.07b	11.34 ± 0.55d
T_5	1.51 ± 0.02d	1.27 ± 0.019cde	1.52 ± 0de	1.2 ± 0.026ef	11.74 ± 0.22cd	11.63 ± 0.17de	11 ± 0.96ab	11.5 ± 0.31d
T_6	1.64 ± 0.01bc	1.27 ± 0.005de	1.6 ± 0.02bcd	1.24 ± 0.031def	12.66 ± 0.32bcd	12.55 ± 0.3cd	11.54 ± 0.61ab	12.04 ± 0.17cd
T_7	1.55 ± 0.01d	1.29 ± 0.008cd	1.53 ± 0.03cde	1.24 ± 0.021def	12.86 ± 0.32bcd	12.75 ± 0.36c	11.35 ± 0.72ab	12.52 ± 0.16bcd
T_8	1.66 ± 0.02bc	1.34 ± 0.017e	1.62 ± 0.01bc	1.29 ± 0.001cde	12.96 ± 0.27bcd	13.85 ± 0.22b	12.19 ± 0.45ab	13.02 ± 0.19bc
T_9	1.68 ± 0.02b	1.49 ± 0.011ab	1.66 ± 0.03b	1.37 ± 0.016bcd	13.26 ± 0.47bc	14.58 ± 0.19b	12.65 ± 0.56a	13.48 ± 0.06ab
Treatment	Starch content (mg g^{-1} FW)				CSI (%)			
	2019	2019-2020	2020	2020-2021	2019	2019-2020	2020	2020-2021
	I-rice	I-wheat	II-rice	II-wheat	I-rice	I-wheat	II-rice	II-wheat
T_0	17.49 ± 0.35f	14.5 ± 0.35f	18.1 ± 0.25g	14.6 ± 0.16g	49.5 ± 0.35f	46.35 ± 0.83f	48.5 ± 0.93f	47.4 ± 0.56f
T_1	24.13 ± 0.43cd	23.4 ± 0.43cd	25.13 ± 0.54d	24.5 ± 0.47bc	76.52 ± 0.52cd	73.37 ± 0.27cd	71.4 ± 0.6c	70.3 ± 1.61cd
T_2	31.77 ± 0.08a	26.4 ± 0.08a	29.99 ± 0.53b	25.1 ± 0.1b	86.65 ± 1.4a	83.5 ± 0.09a	79.4 ± 1.22ab	76.97 ± 0.74b
T_3	33.6 ± 0.12a	29.5 ± 0.12a	32.09 ± 0.52a	27.5 ± 0.7a	89.47 ± 0.65a	86.32 ± 0.9a	83.8 ± 0.04a	82.7 ± 0.56a
T_4	21.05 ± 0.14e	20.5 ± 0.14e	19.6 ± 0.46g	18.6 ± 0.11f	69.28 ± 1.01e	66.13 ± 1.58e	64.4 ± 1.34e	63.3 ± 1.38e
T_5	23.43 ± 0.44d	22 ± 0.44d	21.96 ± 0.3f	21.4 ± 0.03e	73.16 ± 0.53de	70.01 ± 0.07de	65.3 ± 0.68e	64.2 ± 0.53e
T_6	25.81 ± 0.34cd	23 ± 0.34c	25.52 ± 0.15d	21.8 ± 0.18de	78.2 ± 1.06bc	75.05 ± 1.56bc	70.5 ± 0.7d	69.4 ± 0.51d
T_7	23.88 ± 0.46d	22.5 ± 0.46d	22.94 ± 0.05ef	22 ± 0.03de	78.41 ± 0.65bc	75.26 ± 0.27bc	72.3 ± 1.05cd	71.2 ± 0.41cd
T_8	25.16 ± 0.21cd	23.7 ± 0.21cd	24.57 ± 0.59de	23.2 ± 0.37cd	80.77 ± 1.05bc	77.62 ± 0.65b	76 ± 1.11bc	74.9 ± 1.09bc
T_9	29.19 ± 0.68b	25.5 ± 0.68b	27.58 ± 0.34c	23.8 ± 0.09bc	82.03 ± 1.11b	78.88 ± 0.33b	78 ± 0.56b	76.57 ± 1.04b

Mean values within the same column having alike alphabets differ non-significantly ($p \leq 0.05$), while different alphabets show a significant difference ($p \leq 0.05$). Mean (±SE) was taken from three replicates for each treatment.Treatments : T0 -without Fertilizer, T1 - 100 % RDF, T2 - 100 % RDF + 20 t ha-1 SSL, T3 - 100 % RDF + 30 t ha-1 SSL, T4-50 % RDF + 20 t ha-1 SSL, T5 -60 % RDF + 20 t ha-1 SSL, T6 - 70 % RDF + 20 t ha-1 SSL, T7 - 50 % RDF + 30 t ha-1 SSL, T8 - 60 % RDF + 30 t ha-1 SSL and T9 - 70 % RDF + 30 t ha-1 SSL.

Table 5. Effect of conjoint application of sewage sludge and fertilizer on stress markers parameters of rice and wheat

Treatment	Proline content (µg mg^{-1} FW)				Phenol content (µg mg^{-1} FW)			
	2019	2019-2020	2020	2020-2021	2019	2019-2020	2020	2020-2021
	I-rice	I-wheat	II-rice	II-wheat	I-rice	I-wheat	II-rice	II-wheat
T_0	89.67 ± 0.93a	91.67 ± 0.6a	92.22 ± 1.3a	94 ± 1.2a	6.77 ± 0.1a	6.0 ± 0a	6.5 ± 0.1a	6.1 ± 0.08a
T_1	85.43 ± 1.35abc	86 ± 0.58abc	86.12 ± 0.7bc	86.36 ± 0.98bc	5.6 ± 0.11b	5.3 ± 0.12b	5.3 ± 0.1b	5.4 ± 0.13b
T_2	82.67 ± 2.35bcd	81.67 ± 3.26bcd	84 ± 2.08bc	83.33 ± 2.65bcd	4.5 ± 0.08de	4 ± 0.08d	4.9 ± 0.01c	4.8 ± 0.07cd
T_3	77.33 ± 0.72d	74.67 ± 30d	78.33 ± 0.65de	77.67 ± 0.79d	3.3 ± 0.04g	3.0 ± 0.04f	4 ± 0.04g	3.5 ± 0.03g
T_4	86.50 ± 0.2ab	87.17 ± 1.02ab	88.37 ± 0.24ab	88.17 ± 0.39ab	5.9 ± 0.09b	4.6 ± 0.09c	6.15 ± 0.05a	5.1 ± 0.03bc
T_5	83.17 ± 1.3abcd	84.17 ± 1.28abcd	84.34 ± 0.62bc	84.17 ± 0.72bcd	5 ± 0.11c	4.5 ± 0.01c	5.4 ± 0.04b	4.8 ± 0.11cd
T_6	81 ± 0.87bcd	80.6 ± 1.15bcd	83.57 ± 0.69bcd	83.6 ± 0.58bcd	4.6 ± 0.06cd	4.1 ± 0.04d	4.8 ± 0.11cd	4.5 ± 0.1de
T_7	80.30 ± 2.78bcd	78.63 ± 2.73bcd	81.35 ± 1.4cde	80.97 ± 1.5cd	4.3 ± 0.08def	4 ± 0.07d	4.5 ± 0.09de	4.4 ± 0.11de
T_8	79.07 ± 0.45cd	77.07 ± 1.61cd	78.54 ± 0.95de	79.4 ± 1.68d	4.1 ± 0.08ef	3.8 ± 0.03de	4.4 ± 0.09ef	4.2 ± 0.04ef
T_9	76.63 ± 0.37d	75.63 ± 1.46d	76.97 ± 1.02e	77.63 ± 1.19d	4 ± 0.09f	3.5 ± 0.06e	4.1 ± 0.03fg	3.9 ± 0.09fg
Treatment	MDA (mg g^{-1})				(MSI) (%)			
	2019	2019-2020	2020	2020-2021	2019	2019-2020	2020	2020-2021
	I-rice	I-wheat	II-rice	II-wheat	I-rice	I-wheat	II-rice	II-wheat
T_0	0.06 ± 0a	0.055 ± 0a	0.06 ± 0a	0.06 ± 0a	45.89 ± 0.84g	43±0.96h	45.89 ± 0.32f	40.5 ± 0.53h
T_1	0.045 ± 0b	0.048 ± 0b	0.043 ± 0cdef	0.051 ± 0cdef	57.66 ± 1.29ef	55.4±1.15de	59.4 ± 1.51cd	52.9 ± 0.69de
T_2	0.041 ± 0cde	0.046 ± 0bc	0.04 ± 0fg	0.048 ± 0fg	66.91 ± 0.03bc	61.51±1.25bc	63.4 ± 0.59bc	59.01 ± 1.44bc
T_3	0.038 ± 0e	0.04 ± 0d	0.038 ± 0g	0.046 ± 0g	73.5 ± 1.07a	68.1±1.03a	71.8 ± 1.61a	65.6±1.16a
T_4	0.044 ± 0bc	0.045 ± 0bc	0.049 ± 0b	0.057 ± 0ab	54.34 ± 0.85f	48.94±0.1g	54.34 ± 1.02de	46.44 ± 0.63g
T_5	0.044 ± 0bc	0.045 ±0bc	0.046 ± 0bc	0.054 ± 0bc	56.42 ± 0.91ef	51.02±0.48fg	51.3 ± 0.27e	48.52 ± 0.13fg
T_6	0.043 ± 0bc	0.048 ±0b	0.044 ± 0cde	0.052 ± 0cde	59.74 ± 1.15de	54.34±1.1def	56.5 ± 1.38de	51.84 ± 0.84ef
T_7	0.042 ± 0bcd	0.045 ± 0bc	0.045 ± 0cd	0.053 ± 0cd	59 ± 1.47def	53.6±0.78ef	58.3 ± 0.52cd	51.1 ± 0.37ef
T_8	0.039 ± 0de	0.043 ± 0cd	0.042 ± 0def	0.05 ± 0def	63.5 ± 0.03cd	58.1±0.33cd	62 ± 1.16bc	55.6 ± 0.09cd
T_9	0.038 ± 0e	0.041 ± 0d	0.041 ± 0efg	0.049 ± 0efg	68.7 ± 0.32b	63.3±0.26b	66 ± 0.96b	60.8 ± 0.13b

Mean values within the same column having alike alphabets differ non-significantly ($p \leq 0.05$), while different alphabets show a significant difference ($p \leq 0.05$). Mean (±SE) was taken from three replicates for each treatment.Treatments : T_0 -without Fertilizer, T_1 - 100 % RDF, T_2 - 100 % RDF + 20 t ha^{-1} SSL, T_3 - 100 % RDF + 30 t ha^{-1} SSL, T_4-50 % RDF + 20 t ha^{-1} SSL, T5 -60 % RDF + 20 t ha^{-1} SSL, T_6 - 70 % RDF + 20 t ha^{-1} SSL, T_7- 50 % RDF + 30 t ha^{-1} SSL, T8 - 60 % RDF + 30 t ha^{-1} SSL and T_9 - 70 % RDF + 30 t ha^{-1} SSL.

initiation and maintenance, especially pronounced in (Fig. 2,3). MSI and CSI were significantly improved under integrated treatments. Higher MSI and CSI values indicate reduced membrane injury and better preservation of photosynthetic pigments, reflecting lower oxidative damage and improved cellular integrity under balanced nutrient supply.

Stress markers (phenol, proline, MDA)

Stress-related metabolites responded inversely to growth and physiological parameters (Table 5, Fig. 2, 3). Proline and phenolic contents were highest in the control and declined significantly under SSL-RDF treatments, with T_3 consistently recording the lowest values. This reduction reflects decreased stress severity rather than suppression of metabolic activity, indicating improved nutrient and osmotic balance. MDA, a marker of lipid peroxidation, was significantly reduced under integrated treatments. Lower MDA content under SSL-RDF combinations indicates mitigation of oxidative stress and enhanced membrane protection, consistent with higher MSI values.

Multivariate and dose-response analyses

PCA explained > 94 % of the total variance in both crops (Fig. 4, 5). Quadrants dominated by growth and physiological traits represent nutrient sufficiency and low stress, whereas quadrants associated with proline, phenolics and MDA represent stress-dominated conditions. Treatments T_2, T_3 and T_9 clustered with positive growth and biochemical attributes, indicate superior nutrient management efficiency, while T_0 clustered with stress indicators, confirms nutrient deficiency-induced stress. Correlation matrix analysis (Fig. 6A, B) reveals a positive correlation between growth attributes and physio-biochemical parameters in both crops in every season. Further correlation matrix showed a negative correlation between stress indicators and other parameters in both crops. In addition, the scatter plot illustrates the relationship between starch concentration (mg g^{-1} FW) and tiller number per running meter (Fig. 7A – D). This emphasizes the pivotal role of starch in promoting tiller formation, suggesting that relatively small increments in starch concentration can lead to

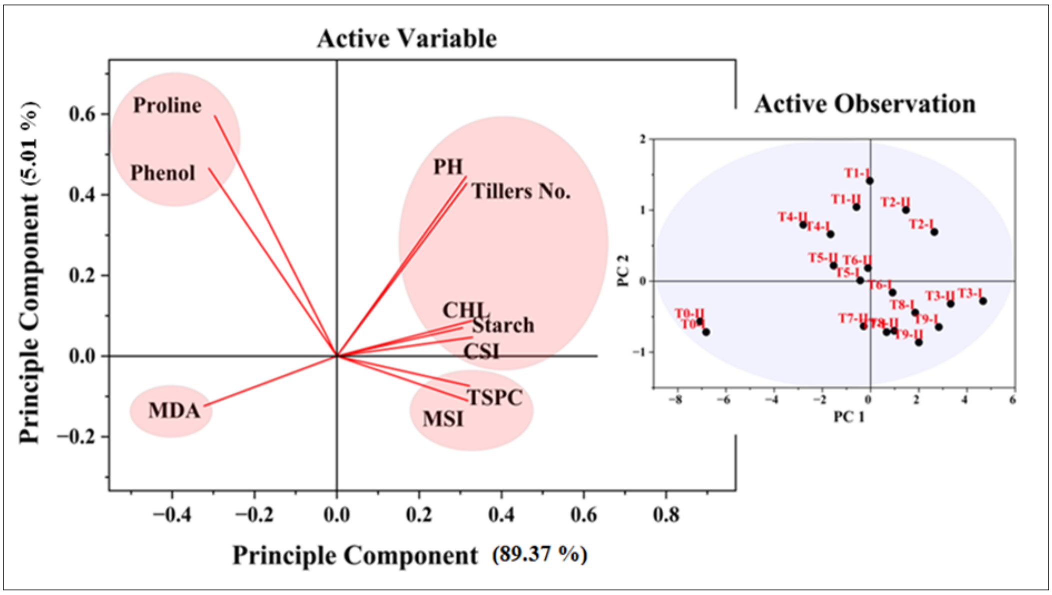

Fig. 4. PCA with various combinations of RDF and SSL treatment conditions in rice under two seasons. Arrows represent the parameters on the corresponding dimensions (PC1 and PC2), where PC2 expressed most of the variability in the data. (T_0-I to T_9-I denotes treatments of season I and T_0-II to T_9-II denotes treatments of season II).

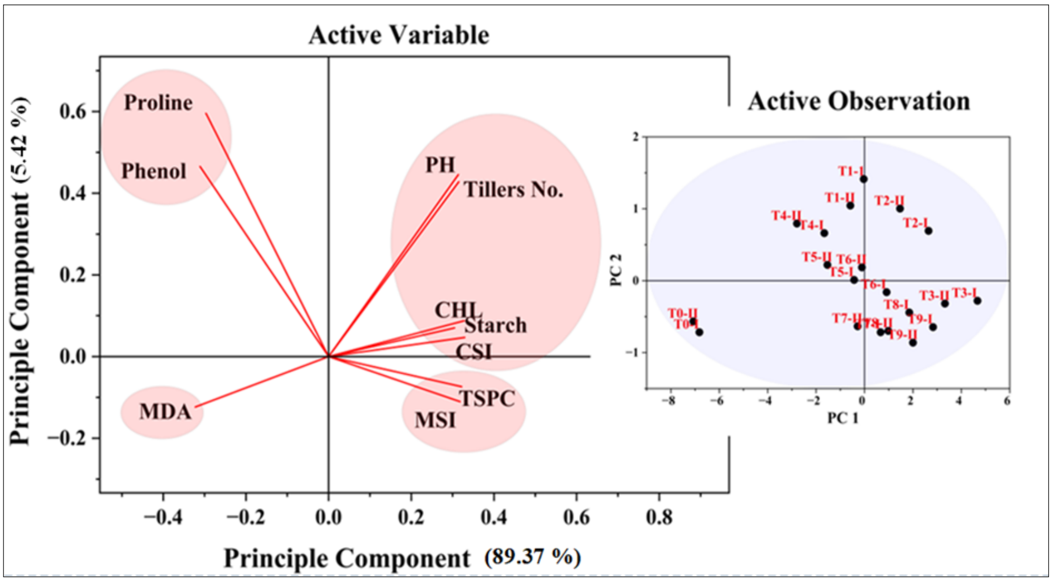

Fig. 5. The principal component analysis with various combinations of RDF and SSL treatment conditions in wheat under two seasons. Arrows represent the parameters on the corresponding dimensions (PC1 and PC2), where PC2 expressed most of the variability in the data. (T_0-I to T_9-I denotes treatments of season I and T_0-II to T_9-II denotes treatments of season II).

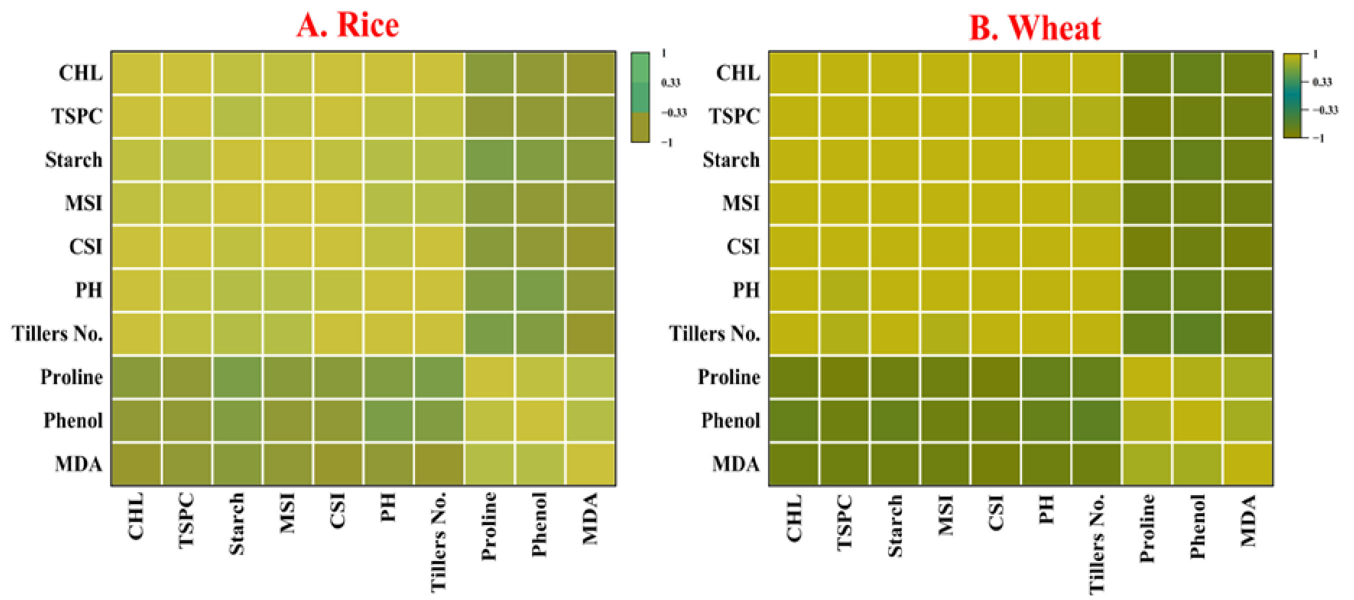

Fig. 6. Grid correlation matrix shows the correlation between all parameters with colour code (A) rice and (B) wheat.

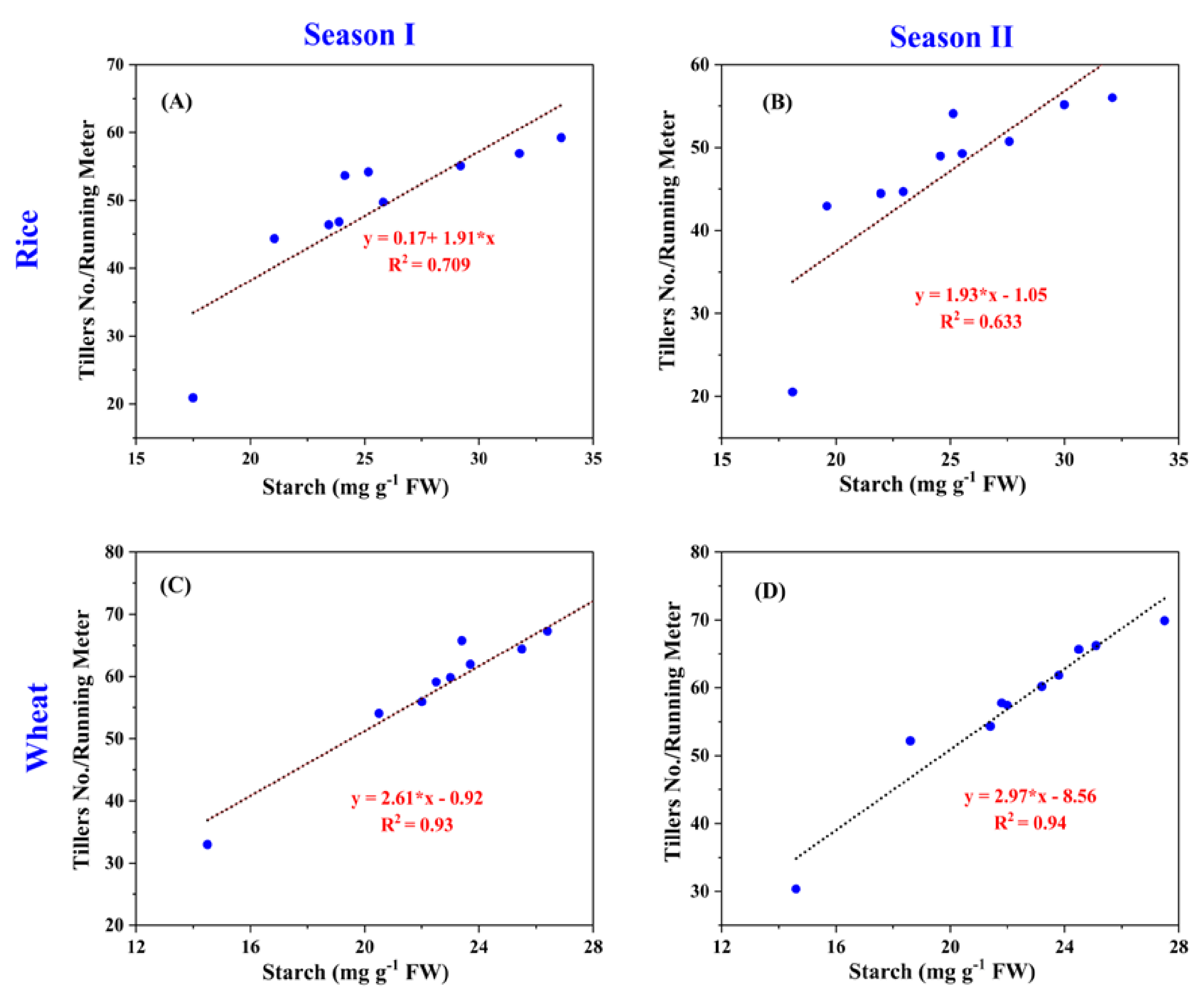

Fig. 7. The scatter plot indicating a linear relationship between the number of tillers per running meters and starch content (mg g^{-1} FW), during I season rice (A), wheat (C), II season rice (B) and wheat (D).

substantial increases in tiller number. Dose-response curves demonstrated a clear stimulatory response up to T_3, followed by a decline under reduced RDF treatments and partial recovery at higher SSL doses (Fig. 8). This confirms an optimal SSL-RDF interaction threshold rather than a linear response.

Discussion

Growth attributes

These findings demonstrate that wheat and rice exhibit differential responses to escalating SSL application rates. The significant improvement in plant height observed under SSL application can be attributed to its ability to enhance soil OM content, physical structure and the availability of essential nutrients, including nitrogen and phosphorus and micronutrients. These factors stimulate cell division and elongation, ultimately promoting vertical growth. Comparable results have been observed in cereals, where the application of SSL, alone or in combination with chemical fertilizers, enhanced vegetative growth by improving nutrient availability and soil health (7, 22).

Evidence from rice and wheat further supports that SSL-CF integration optimizes nutrient uptake, leading to significant improvements in plant height (11, 23). The increase in tiller number under SSL treatments is likely due to a combination of factors, including improved soil fertility, increased microbial activity and gradual nutrient release, which collectively enhance tiller initiation and survival. SSL also improves root proliferation, thereby facilitating efficient water and nutrient uptake that directly supports tiller production (24). These findings align with earlier reports in cereals, where SSL combined with CF resulted in higher vegetative branching and tillering capacity due to sustained nutrient availability (7, 11).

Physiological and biochemical responses

The physiochemical parameters (chlorophyll, starch, proteins, CSI and MSI) displayed great significant changes with treatment of SSL with CF. The addition of SSL generally enhances soil texture, increases OM content and improves total sugar and protein levels in plants (25). Similarly, it was observed that cucumber plants grown in SSL exhibited higher soluble protein content (26). The use of SSL in forest plantations has also been shown to promote tree growth, enhance wood production and improve several soil properties (27). SSL applications positively influenced the biochemical composition of maize plants, including sugars, proteins, free amino acids, proline and antioxidant enzyme activity (28). It was also found that increasing sludge levels led to enhanced sugar and protein content in *Brassica juncea* (29).

The increase in chlorophyll concentration under integrated nutrient management highlights the importance of balanced nitrogen and magnesium availability for chlorophyll biosynthesis. The slow-release nature of SSL nutrients, together with the immediate availability of CF, ensures a steady supply for chlorophyll formation. These results agree with earlier studies reporting synergistic effects of organic-inorganic nutrient sources on chlorophyll accumulation in cereals (30). The increased protein accumulation under combined SSL-CF treatments is likely attributable to enhanced nitrogen availability and improved nitrogen assimilation efficiency. The combination of quickly available nitrogen from CF and the slow mineralization of SSL-derived nitrogen sustains protein synthesis during active vegetative growth. Additionally, SSL improves microbial activity and enzymatic processes related to nitrogen metabolism, which further supports protein accumulation in plant tissues (31, 32).

The rise in starch levels suggests that integrated nutrient management enhances photosynthetic performance and assimilate partitioning. Nitrogen plays a crucial role in regulating starch biosynthesis via the activation of key enzymes, while SSL improves soil conditions and microbial processes that sustain carbohydrate metabolism (33, 34). This dual mechanism likely explains the higher starch accumulation in SSL-CF treatments compared to sole applications. The scatter plot demonstrated the linear relationship between starch concentration and tiller number per running meter in both crops. Together, these findings reinforce the notion that enhanced carbohydrate accumulation not only provides energy but also serves as a regulatory factor influencing the capacity of cereals to initiate and sustain productive tillers, highlighting starch metabolism as a key physiological determinant of crop productivity.

Higher MSI values under integrated nutrient management indicate greater membrane stability, reflecting improved antioxidant defence and osmotic adjustment. The combined nutrient availability from SSL and CF reduces oxidative stress, protecting membrane lipids and proteins from peroxidation. These findings are consistent with previous reports that organic amendments improve cell integrity by enhancing antioxidant

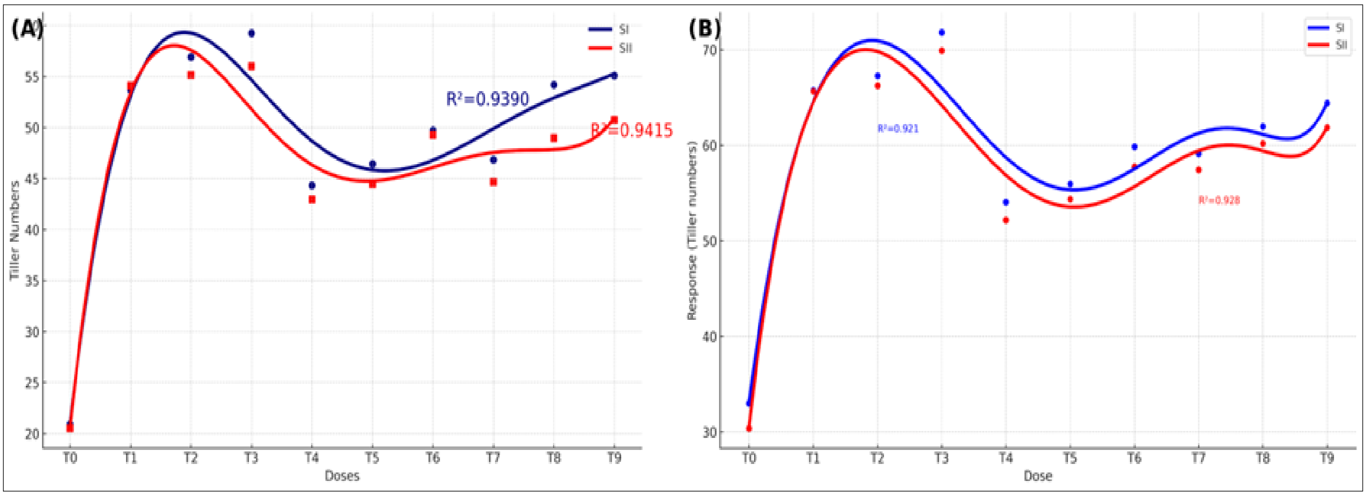

Fig. 8. The dose response curve of tiller numbers of (A) rice and (B) wheat cultivars at different combinations of CF and SSL (T_0-T_9) at two seasons (SI and SII).

activity (35, 36). CSI improvement under SSL-CF integration suggests better preservation of chlorophyll pigments and thylakoid membranes due to reduced oxidative degradation. Such stability under integrated nutrient regimes indicates that improved soil fertility and reduced stress enhance the persistence of photosynthetic machinery (37, 38).

Proline, known for its role as an osmoprotectant in plant metabolism, was found to decrease in treatments with SSL (39). This reduction suggests that plants experienced less stress under these conditions and thus, proline cannot be used as a stress marker in these cases. Conversely, proline levels were higher in control treatments, indicating its typical role as a stress indicator under nutrient-deficient conditions. Proline accumulation is a well-documented plant response to abiotic stress, functioning as an osmolyte, membrane stabilizer and enzyme protector (40). The observed decline in proline levels under SSL-CF treatments may reflect reduced stress severity due to better water and nutrient availability. Similar reductions have been reported in wheat, where adequate nutrient management suppressed stress-induced proline accumulation (41).

Phenolics are vital defence metabolites that provide resistance against pathogens and abiotic stress factors (1). The reduced phenol levels in SSL-amended plants may suggest lower stress intensity, but this could also compromise plant defence capacity against pathogens, necessitating careful management of SSL inputs to balance growth and defence trade-offs.

MDA is a biomarker of lipid peroxidation and oxidative stress (42). Lower MDA content in SSL-CF treatments indicates reduced oxidative damage, likely due to enhanced antioxidant capacity and better cellular stability (43, 44). Previous reports confirm that organic amendments mitigate ROS-mediated stress by strengthening antioxidant defences, which aligns with the present observations (45). However, cautioned that while SSL offers benefits as organic compost, it may also cause genetic damage in plants (46). There are potential health risks associated with consuming crops grown in sewage SSL-amended soils, as sludge may contain harmful substances like heavy metals, organic pollutants and pathogens (47). In the current study, treatments T_3 and T_9, which included RDF at 70 % and 100 % along with 30 % SSL, showed significant increases in chlorophyll, soluble sugar and protein contents, with T_3 displaying the most pronounced improvement. The C: N ratio of the SSL was not determined, which may influence decomposition dynamics and nutrient mineralization patterns.

Conclusion

This study demonstrates that integrated application of SSL with CF provides synergistic benefits to rice and wheat by enhancing growth, photosynthetic efficiency, carbohydrate accumulation and cellular stability while reducing oxidative stress. The combined use of 100 % RDF with 30 t ha^{-1} SSL (T_3) consistently produced the most favourable physiological and biochemical responses. Reductions in proline, phenolics and MDA under integrated treatments reflect reduced nutrient and osmotic-stress severity, rather than metabolic suppression, confirming the role of balanced nutrient availability in stress mitigation. From a practical perspective, SSL should be applied at moderated rates in combination with mineral fertilizers rather than as a sole nutrient source. While higher SSL rates improved plant performance, field-scale feasibility, transportation costs, regulatory thresholds and farmer adoption must also be considered. Although heavy-metal concentrations in soil and grains were not quantified in this study, their potential accumulation remains a critical concern. Regular monitoring of soil and crop metal content is therefore recommended when SSL is used repeatedly. Overall, integrating SSL with RDF represents a viable strategy for improving nutrient use efficiency, sustaining productivity and supporting circular bio-economy practices, if application rates are regulated and long-term soil health is monitored.

Acknowledgements

The authors express their sincere gratitude to the General Manager of the Ganga Pollution Control Unit, Uttar Pradesh Jal Nigam, Bhagwanpur Sewage Treatment Plant (STP), for providing sewage sludge for research purposes. The authors also acknowledge the Department of Soil Science and Agricultural Chemistry, Institute of Agricultural Sciences, Banaras Hindu University, for providing infrastructure and invaluable support.

Authors' contributions

PS carried out the experiments, investigations, methodology, critically reviewed the manuscript and wrote the original manuscript and edited the final draft. A^1, A^2, KM, NK and BKD assisted in writing and editing the final draft. JS participated in methodology and critical review, performed statistical analysis and edited the final draft. YVS did conceptualization, supervision, validation and provided resources. HSJ, SKP and AK edited the final draft. All authors read and approved the final manuscript. [A^1 - Arvind; A^2 - Akash]

Compliance with ethical standards

Conflict of interest: The authors do not have any conflicts of interest to declare.

Ethical issues: None

References

1. Singh A, Agrawal M. Biochemical responses of sesame to sewage sludge application. Ecotoxicol Environ Saf. 2010;73(8):1744-50.
2. Kumar V, Chopra AK. Agronomical performance of high-yielding cultivar of eggplant (*Solanum melongena* L.) grown in sewage sludge-amended soil. Res Agric. 2016;1(1):1-24. https://doi.org/10.1007/s40003-021-00546-y
3. Swain A, Singh SK, Mohapatra KK, Patra A. Sewage sludge amendment affects spinach yield, heavy metal bioaccumulation and soil pollution indexes. Arab J Geosci. 2021;14(8):1-18. https://doi.org/10.1007/s12517-021-07078-3
4. Demirbas A, Edris G, Alalayah WM. Sludge production from municipal wastewater treatment in sewage treatment plant. Energy Sources Part A. 2017;39(10):999-1006. https://doi.org/10.1080/15567036.2017.1283551
5. Eid EM, Alrumman SA, El-Bebany AF, Hesham AEL, Taher MA, Fawy KF. Effects of different sewage sludge amendment rates on heavy metal bioaccumulation, growth and biomass of cucumber (*Cucumis sativus* L.). Environ Sci Pollut Res. 2017;24(19):16371-82. https://doi.org/10.1007/s11356-017-9289-6

6. Eid EM, Alrumman SA, Farahat EA, El-Bebany AF. Prediction models for evaluating the uptake of heavy metals by cucumbers (*Cucumis sativus* L.) grown in agricultural soils amended with sewage sludge. Environ Monit Assess. 2018;190(9):501. https://doi.org/10.1007/s10661-018-6885-y

7. Latare AM, Kumar O, Singh SK, Gupta A. Direct and residual effect of sewage sludge on yield, heavy metals content and soil fertility under rice-wheat system. Ecol Eng. 2014;69:17-24. https://doi.org/10.1016/j.ecoleng.2014.03.066

8. Singh RP, Agrawal M. Effects of sewage sludge amendment on heavy metal accumulation and consequent responses of *Beta vulgaris* plants. Chemosphere. 2007;67(11):2229-40. https://doi.org/10.1016/j.chemosphere.2006.12.019

9. Sharma P, Jha AB, Dubey RS, Pessarakli M. Effect of sewage sludge on spinach productivity and soil health. Int J Recycl Org Waste Agric. 2019;8(1):51-60.

10. Jatav HS, Singh SK, Jatav SS, Rajput VD, Sushkova S. Feasibility of sewage sludge application in rice-wheat cropping system. Eurasian J Soil Sci. 2021;10(3):207-14. https://doi.org/10.18393/ejss.880677

11. Jatav SS, Singh SK, Parihar M, Alsuhaibani AM, Gaber A, Hossain A. Application of sewage sludge in a rice (*Oryza sativa* L.)-wheat (*Triticum aestivum* L.) system influences growth, yield, quality and heavy metals accumulation in the northern Gangetic alluvial plain. Life. 2022;12(4):484. https://doi.org/10.3390/life12040484

12. Hao J, Tan J, Zhang Y, Gu X, Zhu G, Wang S, et al. Sewage sludge-derived nutrients and biostimulants stimulate rice leaf photosynthesis and root metabolism to enhance carbohydrate, nitrogen and antioxidant accumulation. Chemosphere. 2024;352:141335. https://doi.org/10.1016/j.chemosphere.2024.141335

13. Singh A, Agrawal M. Potential benefits and risks of land application of sewage sludge. Waste Manag. 2008;28(2):347-58. https://doi.org/10.1016/j.wasman.2006.12.010

14. Pasqualone A, Delvecchio LN, Lacolla G, Piarulli L, Simeone R, Cucci G. Effect of composted sewage sludge on durum wheat: productivity, phenolic compounds, antioxidant activity and technological quality. J Food Agric Environ. 2014;12:276-80.

15. Duan B, Feng Q. Risk assessment and potential analysis of the agricultural use of sewage sludge in Central Shanxi Province. Int J Environ Res Public Health. 2022;19(7):4236. https://doi.org/10.3390/ijerph19074236

16. Hiscox JD, Israelstam GF. A method for the extraction of chlorophyll from leaf tissue without maceration. Can J Bot. 1979;57(12):1332-4. https://doi.org/10.1139/b79-163

17. Bradford MM. A rapid and sensitive method for the quantitation of microgram quantities of protein utilising the principle of protein-dye binding. Anal Biochem. 1976;72(1-2):248-54. https://doi.org/10.1006/abio.1976.9999

18. Aazami MA, Asghari-Aruq M, Hassanpouraghdam MB, Ercisli S, Baron M, Sochor J. Low temperature stress mediates the antioxidant pool and chlorophyll fluorescence in *Vitis vinifera* L. cultivars. Plants. 2021;10(9):1877. https://doi.org/10.3390/plants10091877

19. Kaloyereas SA. A new method of determining drought resistance. Plant Physiol. 1958;33(3):232. https://doi.org/10.1104/pp.33.3.232

20. Marín Velázquez JA, Andreu Puyal P, Carrasco Miral A, Arbeloa Matute A. Determination of proline concentration, an abiotic stress marker, in root exudates of excised root cultures of fruit tree rootstocks under salt stress. 2010.

21. Xu C, Zhang Y, Cao L, Lu J. Phenolic compounds and antioxidant properties of different grape cultivars grown in China. Food Chem. 2010;119(4):1557-65. https://doi.org/10.1016/j.foodchem.2009.09.042

22. Abbas T, Rizwan M, Ali S, Adrees M, Zia-ur-Rehman M, Qayyum MF, et al. Effect of biochar on alleviation of cadmium toxicity in wheat (*Triticum aestivum* L.) grown on Cd-contaminated saline soil. Environ Sci Pollut Res. 2018;25(26):25668-80. https://doi.org/10.1007/s11356-017-8987-4

23. Rehman RA, Rizwan M, Qayyum MF, Ali S, Zia-ur-Rehman M, Zafar-ul-Hye M, et al. Efficiency of various sewage sludges and their biochars in improving selected soil properties and growth of wheat (*Triticum aestivum*). J Environ Manag. 2018;223:607-13. https://doi.org/10.1016/j.jenvman.2018.06.081

24. Latare AM, Singh SK, Kumar O. Yield and profitability of rice-wheat sequence with conjunctive application of sewage sludge and chemical fertilisers. Indian J Fertilisers. 2017;13(7):50-61.

25. Mazen A, Faheed FA, Ahmed AF. Study of potential impacts of using sewage sludge in the amendment of desert reclaimed soil on wheat and jews mallow plants. Braz Arch Biol Technol. 2010;53:917-30. https://doi.org/10.1590/S1516-89132010000400022

26. Wyrwicka A, Urbaniak M, Przybylski M. The response of cucumber plants (*Cucumis sativus* L.) to the application of PCB-contaminated sewage sludge and urban sediment. Peer J. 2019;7:e6743. https://doi.org/10.7717/peerj.6743

27. Mosquera-Losada MR, López-Díaz L, Rigueiro-Rodríguez A. Sewage sludge fertilisation of a silvopastoral system with pines in north-western Spain. Agrofor Syst. 2001;53(1):1-10. https://doi.org/10.1023/A:1012239419829

28. Abdel Latef AA, Sallam MM. Changes in growth and some biochemical parameters of maize plants irrigated with sewage water. Austin J Plant Biol. 2015;1(1):1004.

29. Dar MI, Naikoo MI, Khan FA, Green ID. Assessing the feasibility of sewage sludge applications for the cultivation of *Brassica juncea* L.: metal accumulation, growth, biochemical and yield responses. Environ Sci Renew Res. 2018;1(1):1-10.

30. Ghouili E, Hidri Y, M'Hamed HC, Somenahally A, Xue Q, Znaïdi IEA, et al. Date palm waste compost promotes plant growth and nutrient transporter gene expression in barley (*Hordeum vulgare* L.). *S* Afr J Bot. 2022;149:247-57. https://doi.org/10.1016/j.sajb.2022.06.018

31. Sankati J, Mishra AK, Banerjee P, Pidikiti P. Improved nutrient management and fertilizer optimization in rice-based cropping systems. In: Key Drivers and Indicators of Soil Health Management: Transitioning from Conventional to Regenerative Agriculture. Singapore: Springer Nature Singapore; 2024. p. 115-36. https://doi.org/10.1007/978-981-97-7564-4_7

32. Liu Y, Lan X, Hou H, Ji J, Liu X, Lv Z. Multifaceted ability of organic fertilizers to improve crop productivity and abiotic stress tolerance: review and perspectives. Agronomy. 2024;14(6):1141. https://doi.org/10.3390/agronomy14061141

33. Zhen S, Deng X, Li M, Zhu D, Yan Y. 2D-DIGE comparative proteomic analysis of developing wheat grains under high-nitrogen fertilization revealed key differentially accumulated proteins that promote storage protein and starch biosynthesis. Anal Bioanal Chem. 2018;410(24):6219-35. https://doi.org/10.1007/s00216-018-1230-4

34. Lv X, Ding Y, Long M, Liang W, Gu X, Liu Y, et al. Effect of foliar application of various nitrogen forms on starch accumulation and grain filling of wheat (*Triticum aestivum* L.) under drought stress. Front Plant Sci. 2021;12:645379. https://doi.org/10.3389/fpls.2021.645379

35. Kumar S, Trivedi PK. Glutathione S-transferases: role in combating abiotic stresses including arsenic detoxification in plants. Front Plant Sci. 2018;9:751. https://doi.org/10.3389/fpls.2018.00751

36. Khan I, Mahmood S, Chattha MU, Chattha MB, Ahmad S, Awan MI, et al. Organic amendments improved the productivity and bio-fortification of fine rice by improving physiological responses and nutrient homeostasis under salinity stress. Plants. 2023;12(8):1644. https://doi.org/10.3390/plants12081644

37. Vishwakarma M, Kulhare PS, Tagore GS. Chlorophyll content in leaves of wheat as influenced by inorganic, organic and integrated sources of nutrient application. Int J Plant Soil Sci. 2021;33:31-45. https://doi.org/10.9734/ijpss/2021/v33i230412

38. Fathi A, Shiade SRG, Kianersi F, Altaf MA, Amiri E, Nabati E.

Photosynthesis in cereals under drought stress. 2024. https://doi.org/10.1201/b22922-38

39. Abd El-Samad HM, Abd Nabi SH, Abd El-Hakeem KN. Interactive effect of sewage sludge application with phytohormones IAA or SA on three broad bean cultivars. Am J Plant Sci. 2020;11(6):880-95. https://doi.org/10.4236/ajps.2020.116064
40. Hosseinifard M, Stefaniak S, Ghorbani Javid M, Soltani E, Wojtyla Ł, Garnczarska M. Contribution of exogenous proline to abiotic stress tolerance in plants: a review. Int J Mol Sci. 2022;23(9):5186. https://doi.org/10.3390/ijms23095186
41. El Moukhtari A, Cabassa-Hourton C, Farissi M, Savouré A. How does proline treatment promote salt stress tolerance during crop plant development? Front Plant Sci. 2020;11:1127. https://doi.org/10.3389/fpls.2020.01127
42. Del Río LA, Sandalio LM, Corpas FJ, Palma JM, Barroso JB. Reactive oxygen species and reactive nitrogen species in peroxisomes: production, scavenging and role in cell signalling. Plant Physiol. 2006;141(2):330-5. https://doi.org/10.1104/pp.106.078204
43. Gill SS, Tuteja N. Reactive oxygen species and antioxidant machinery in abiotic stress tolerance in crop plants. Plant Physiol Biochem. 2010;48(12):909-30. https://doi.org/10.1016/j.plaphy.2010.08.016
44. Sharma P, Jha AB, Dubey RS, Pessarakli M. Reactive oxygen species, oxidative damage and antioxidative defence mechanism in plants under stressful conditions. J Bot. 2012;2012:217037. https://doi.org/10.1155/2012/217037
45. Farooq M, Wahid A, Kobayashi N, Fujita D, Basra SMA. Plant drought stress: effects, mechanisms and management. In: Lichtfouse E, Navarrete M, Debaeke P, Véronique S, Alberola C, editors. Sustainable Agriculture. Dordrecht: Springer; 2009. p. 153-88. https://doi.org/10.1007/978-90-481-2666-8_12
46. Martins MNC, de Souza VV, da Silva Souza T. Genotoxic and mutagenic effects of sewage sludge on higher plants. Ecotoxicol Environ Saf. 2016;124:489-96. https://doi.org/10.1016/j.ecoenv.2015.11.031
47. Lamastra L, Suciu NA, Trevisan M. Sewage sludge for sustainable agriculture: contaminants' contents and potential use as fertilizer. Chem Biol Technol Agric. 2018;5(1):1-6. https://doi.org/10.1186/s40538-018-0122-3

Additional information

Peer review: Publisher thanks Sectional Editor and the other anonymous reviewers for their contribution to the peer review of this work.

Reprints & permissions information is available at https://horizonepublishing.com/journals/index.php/PST/open_access_policy

Indexing: Plant Science Today, published by Horizon e-Publishing Group, is covered by Scopus, Web of Science, BIOSIS Previews, Clarivate Analytics, NAAS, UGC Care, etc
See https://horizonepublishing.com/journals/index.php/PST/indexing_abstracting

Publisher information: Plant Science Today is published by HORIZON e-Publishing Group with support from Empirion Publishers Private Limited, Thiruvananthapuram, India.