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Growth Dynamics and Yield of Zero-Till Maize as Influenced by Microbial Consortia Treated Paddy Residue Incorporation

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Authors' contributions

This work was carried out in collaboration among all authors. 'All authors read and approved the final manuscript.

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ABSTRACT

A field experiment was conducted at College farm, Professor Jayashankar Telangana State Agricultural University, Rajendranagar, Hyderabad during 2020-21 and 2021-22 to assess the influence of paddy residue management practices at different fertility levels on certain growth parameters and the yield of *rabi* maize succeeding *kharif* rice in a strip plot design. The growth analysis indicated that the leaf area index, crop growth rate, net assimilation rate and leaf area duration remained the same with residue management practices at 0-30 DAS and 30-60 DAS but between 60 DAS-harvest, maize leaf area index (LAI), crop growth rate (CGR), net assimilation rate (NAR) and leaf area duration (LAD) was significantly increased by incorporation of crop residues treated with microbial consortia and SSP. Different fertility levels affected leaf area index, crop growth rate and leaf area duration at all the growth intervals whereas net assimilation rate remained unaffected. Interaction effect of residue management practices and fertility levels on leaf area index, crop growth rate, net assimilation rate and leaf area duration was found non significant at 0-30 DAS

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and 30-60 DAS, while at 60 DAS-harvest integration of residues and microbial consortia along with chemical fertilizers resulted in a significant increase of LAI, CGR and LAD with no significant improvement in NAR. In 2020-21 and 2021-22, the grain yield and straw yield were higher in the incorporation of crop residues treated with microbial consortia and single super phosphate (SSP) in combination with 125 % recommended dose of fertilizers (RDF) which was on par with 100% RDF and 75 % RDF and was significantly superior over other treatments.

Keywords: Growth parameters; fertility levels; microbial consortia; rabi maize; residue management; yield.

1. INTRODUCTION

A large amount of rice residue is annually produced in the rice growing countries. A significant amount of rice straw has also been left in the field after the grain has been harvested as a result of the use of mechanical farming. India produces roughly 500 Mt of agricultural residues annually, according to the National Policy for Management of Crop Residues (NPCMR). Burning crop remnants causes a significant loss of plant nutrients, especially organic carbon. It was discovered that the entire amount of organic carbon, as well as almost 80-90 percent of nitrogen, 25 percent phosphorus, 20 percent potassium, 50 percent sulphur, and 25 percent of phosphorus were lost as gaseous and particulate debris [1]. The crop residues can be used in a efficient and utilizable manner other than burning. Adoption of appropriate tillage practices by retention of residue helps to reduce physical disturbance of the soil and leads to the accumulation of more soil organic matter by enhancing microbial population [2].

In the green revolution era, intensive agriculture with the use of high analysis inorganic fertilizers alone beyond the threshold requirement of crop and soil resulted in diminishing soil fertility and posed a stern hazard to sustainable crop production. Availability of fertilizers at subsidized rates and faster output in terms of yield increment encouraged the farmers towards imbalanced use of fertilizers. This not only led to a downward spiraling of natural resources but also made us to depend on non-renewable resources both of which posed a hazard to sustainable agriculture [3]. Thus, there is an immediate necessity for an alternate sustainable nutrient management strategy to overcome this dawning challenge [2].

Microbial consortia are a type of biofertilizers that involves symbiotic interaction of two or more microbial groups for enhancing the efficiency. They intensify the turnover of soil organic matter

and mobilize various nutrients for plant use and also aid in metabolic actions like fixation of nitrogen from atmosphere, solubilization of soil P, release of fixed K and Zn, thereby increase their availability to crop [4] in a sustainable way apart from improving soil health [5]. Incorporating a microbial consortium made up of *Azotobacter, Trichoderma harzianum* and *Pseudomonas fluorescens* into the supply of nutrients can both decrease the need for chemical fertilisers while also improving their performance [6]. Thus the present experiment was planned with the objective to determine the effect of integrated application of residues treated with microbial consortium along with NPK fertilizers on crop growth dynamics and yield of maize *(Zea mays)* as it is the third most important cereal after rice and wheat in the world.

2. MATERIALS AND METHODS

2.1 Study Region

A two year study (2020–2022) on the rice-maize system was conducted at the Agricultural College Farm of Professor Jayashankar Telangana State Agricultural University with 17°32'22"N latitude and 78°41'11"E longitudeand an altitude of 550 m above the mean sea level. Before the initiation of the actual experiment, rice crop was grown uniformly with traditional farmers practice for homogenization of soil fertility and collection of residues for the experiment. The soil of the experimental field was sandy clay loam, deep and free from gravels. Base line soil samples from 0 to 15 cm depth were collected and analyzed before the initiation of the experiment. The soil had 0.18 % of soil organic carbon as determined by the wet digestion method [7], 145 kg ha⁻¹ of available soil nitrogen examined by the alkaline permanganate method [8], 38 kg ha $^{-1}$ of available P_2O_5 analyzed by spectrophotometer using Olsens method and 277 kg ha⁻¹ of available potassium (K) determined by neutral normal ammonium acetate method and analyzed by a flame photometer [9]. The soil pH was 7.84 (1:2.5, soil and water ratio) and a bulk density of 1.42 g cm−3 was estimated by the core sampler method [10]. The NPK content in maize plant was 2.3, 0.6 and 1.9, respectively. Rice was grown during the rainy season and harvested in the winter season during both years of study. Maize was sown under zero tillage after the harvest of rice. The amount of rainfall received and temperature variation from sowing to harvest were measured for the entire growing season.

2.2 Methodology

The experiment was laid out in a strip plot design with three replications. The experiment consisted of combinations of residue management practices and fertility levels as follows:

M₁: Burning residue before sowing, M₂: Surface retention of residues, M3: Removal of residues before sowing, M_4 : Incorporation at 15 DAS, M_5 : Incorporation + SSP at equivalent to 'P' dose at 15 DAS, M_6 : Spraying consortia of decomposers @ 10% of residue weight + surface retention, M_7 : Spraying consortia of decomposers @ 10% of residue weight + incorporation at 15 DAS, $M₈$: Spraying consortia of decomposers @ 10% of residue weight + incorporation at 15 DAS + SSP at equivalent to 'P' dose and three fertility levels $(S_1: 75 \%$ RDF, $S_2: 100\%$ RDF and $S_3: 125 \%$ RDF).

Leaf area index, crop growth rate, net assimilation rate and leaf area duration at intervals for the entire growing season were computed.

2.3 Leaf Area Index (LAI)

The leaf area of five contiguous plants from the border rows leaving the extreme row was measured at 30, 60 DAS and at harvest in maize by using the leaf area meter image analysis system. After computing the leaf area as explained above, the leaf area index was calculated by using the following formula as suggested by Watson [11].

LAI= Leaf area (cm²) / Ground area (cm²)

2.4 Crop Growth Rate (CGR)

Crop growth rate can be defined as the rate of dry matter production per unit ground area per unit time. It was calculated during 0– 30 DAS, 30– 60 DAS and 60 DAS – harvest in maize as per the formula given by Watson, 1956.

Crop growth rate =
$$
\frac{W_2 - W_2}{t_2 - t_1} \times \frac{1}{P}
$$
 (g m – 2 day – 1)

where,

 W_1 and W_2 are the total dry weight (g) at time t_1 and t_2 respectively and

P is the ground area occupied by the plants (m^2) .

2.5 Net Assimilation Rate (NAR)

The net assimilation rate indirectly indicates the rate of net photosynthesis. It is expressed as dry matter produced (g) per unit of leaf area (m^2) per unit time (day). It was calculated by the formula outlined by Gregory, [12]. NAR was calculated at 0 – 30 DAS, 30– 60 DAS and 60 DAS – harvest in maize.

Net assimilation rate = $(\mathsf{W}_2 - \mathsf{W}_1) \times (\mathsf{Log}_e \mathsf{A}_2 - \mathsf{Log}_e \mathsf{A}_1)$ (g m⁻² day⁻¹) $(t_2 - t_1)$ (A₂ – A₁)

where,

 W_1 and W_2 are the total dry weight (g) at time t_1 and $t₂$ respectively.

 A_1 and A_2 are leaf area (m²) at time t_1 and t_2 respectively.

2.6 Leaf Area Duration (LAD)

Leaf area duration is the integration part of leaf area over a while [11]. It is a measure of its ability to produce leaf area on unit area of land throughout its life. LAD was calculated 0– 30 DAS, 30– 60 DAS and 60 DAS – harvest in maize.

Leaf area duration (days) = A_1 + A_2 x (t₂ – t₁) 2

where,

 A_1 and A_2 are leaf area index at time t_1 and t_2 respectively

For the determination of grain yield, the kernels from the air-dried cobs from each net plot were separated, dried and threshed after proper cleaning to obtain 14 per cent moisture. The weight of grains of each plot was recorded separately and expressed as grain yield in kg ha⁻¹. Stover from the net plot area was weighed after thorough drying under the sun and expressed as stover yield in kg ha⁻¹.

3. RESULTS AND DISCUSSION

3.1 Leaf Area Index

The yield is determined by the extent of assimilating synthesis, and leaf area is a fundamental physiological parameter that controls this process. The canopy design, which in turn is determined by agronomic techniques like plant variety, plant density, crop and fertilizer management, etc., determines optimal leaf area. Up to 60 DAS, all treatments generally improved leaf area; after that point, a downward tendency toward maturity due to leaf senescence was seen. An overview in Table 1 showed that throughout both of the experimentation years, residue management and fertility level treatments did not significantly affect leaf area at 30 and 60 DAS. On the other hand, during both trial years, there were notable changes in leaf area between treatments at harvest. The interaction between residue management and fertility levels, remained significant over both years at the harvest stage only.

During 2020-21, leaf area index was higher with the incorporation of residues treated with microbial consortia and SSP followed by incorporation treated with consortia and incorporation treated with SSP. The boost in leaf area index was due to a significant increase in leaf primordial, a high rate of cell division and cell expansion [13]. On the other hand, significantly lower LAI was noticed with the removal of residues which was in equivalence with incorporation, retention + consortia, surface retention and *in-situ* burning. During 2021-22, similar trend was observed with incorporation +consortia +SSP, incorporation+consortia and incorporation+SSP while bare incorporation was found superior over retention + consortia, surface retention, *in-situ* burning and removal.

A significantly higher leaf area index was registered with fertility level of 125 % RDF over 100 % RDF and 75 % RDF. The greater amounts of cellular protoplasm as well as increased amounts of proteins all of which contributed to increase in leaf area with higher fertilizer levels.

The results find support from the findings of Amanullah et al. [14] who reported increase in leaf area index with the increase in nitrogen rate.

No interaction effect was found at 30 and 60 DAS. At harvest, perusal of the data reveled that the incorporation of residues treated with

consortia and SSP in combination with 75 % RDF resulted in higher leaf area index which was similar to 100 % RDF and 75 % RDF. A similar trend was observed with incorporation of residues treated with consortia alone. Lower leaf area index was observed under removal with 75 % RDF, *in-situ* burning with 75 % RDF, retention with 75 % RDF, retention + consortia with 75 % RDF and incorporation with 75 % RDF. Similar findings of increased leaf area in the chickpea crop with combined application of chemical fertilizers and microbial inoculants (Rhizobium + PSB + PGPR) were also reported by Rani et al. [15].

3.2 Crop Growth Rate

Crop growth rate increased from sowing to 60 DAS thereafter showed a declining trend which might be due to assimilation translocation to the seed, the senescence and leaf fall at later stage. Residue management practices had a significant impact on crop growth rate at 60 DAS – harvest only (Table 1 & 2) during 2020-21 and 2021-22. The crop growth rate was higher under incorporation $+$ microbial consortia $+$ SSP, while it was lower in removal, *in-situ* burning of residues, incorporation and residue retention under zero tillage which were on par with each other during 2020-21. During 2021-22, similar results were attained with incorporation + microbial consortia + SSP but mere incorporation was found effective over removal, *in-situ* burning, retention and retention + microbial consortia indicating incorporation effect dominating over the burning, removal and retention of residues. This is similar to results from Meena et al. [16] where residue addition resulted in higher crop growth rate under zero- till conditions.

Fertility levels also influenced the crop growth rate at all the intervals of observation which was enhanced significantly by 125 % RDF over 100 % RDF and 75 % RDF. A decrease in the crop growth rate of maize when supplied with 75% RDF might be due to a decrease in leaf area index at low rate of fertilizer application. These results corroborate those obtained by Pandey et al. [17], who reported a significant increase in CGR and NAR in maize hybrids grown under sufficient phosphorous levels.

The crop growth rate was not influenced by the interaction of both residue management practices and fertility levels at 0-30 DAS and 30- 60 DAS but at 60 DAS-harvest, incorporation + microbial consortia + SSP in combination with 125 % RDF recorded higher crop growth rate which was on par with incorporation $+$ microbial consortia+ SSP with 100 % RDF and

incorporation + microbial consortia + SSP with 75 % RDF. The similar trend was observed with combination of incorporation + microbial

Table 1. Leaf area index of maize as influenced by residue management options and fertilizer levels during 2020 and 2021

Table 2. Interaction effect of residue management and fertilizer levels on leaf area index of maize at harvest during 2020-21

Treatment	Leaf area index (harvest)						
	Fertilizer levels						
Residue management	75 % RDF (S_1)	100 % RDF (S ₂)	125 % RDF (S_3)	Mean			
Burning (M_1)	1.58	2.05	2.68	2.10			
Retention (M_2)	1.58	2.19	2.62	2.13			
Removal (M_3)	1.51	2.01	2.54	2.02			
Incorporation (M_4)	1.70	2.42	3.15	2.42			
Incorporation + SSP (M_5)	2.33	2.81	2.87	2.67			
Retention + consortium (M_6)	1.61	2.19	2.72	2.26			
Incorporation + consortia	2.84	2.91	3.00	2.91			
(M ₇)							
Incorporation $+$ consortium $+$	3.08	3.18	3.19	3.15			
$SSP(M_8)$							
Mean	2.02	2.47	2.84				
For comparison the mean of	SEM+		CD.				
Residue management	0.06		0.20				
Fertilizer levels	0.06		0.27				
Main plot at same level of	0.08		0.26				
sub plot							
Sub plot at same level of	0.26		0.78				
main plot							

Table 3. Interaction effect of residue management and fertilizer levels on leaf area index of maize at harvest during 2021-22

Fig. 1. Crop growth rate as influenced by residue management and fertility levels during 2020-21

consortia with 125 % RDF and incorporation + SSP with 125 % RDF. A significantly lower crop growth rate was obtained with the treatment combination of removal of residues with 75 % RDF which was at par with *in-situ* burning of residues with 75 % RDF, surface retention with 75 % RDF and retention + microbial consortia with 75 % RDF.

3.3 Net Assimilation Rate (g m-2 day-1)

The residue management effect was significant at 60 DAS - harvest only, with highest increase in net assimilation rate under incorporation + microbial consortia + SSP which was at par with incorporation + microbial consortia, incorporation + SSP and incorporation (Table 3). During the first and second year of study, lower values of net assimilation rate were observed under surface retention which was equivalent to *in-situ* burning, removal and retention + microbial consortia.

Fertility levels resulted in a non significant increase in net assimilation at 0 – 30 DAS, 30 – 60 DAS and 60 DAS – harvest intervals during both the years of experimentation. Similarly no interaction effect was found between residue the *rabi* seasons of 2020-21 and 2021-22.

management practices and fertility levels during

Table 4. Interaction effect of residue management and fertilizer levels on crop growth rate (g m-2 day-1) of maize at 60 DAS-harvest during 2020-21

Table 5. Interaction effect of residue management and fertilizer levels on crop growth rate (g m-2 day-1) of maize at 60 DAS-harvest during 2021-22

Fig. 2. Crop growth rate as influenced by residue management and fertility levels during 2021-22

Fig. 3. Net assimilation rate (g m-2 day-1) of maize as influenced by residue management and fertilizer levels during 2020-21

3.4 Leaf Area Duration (Days)

Leaf area duration (LAD) indicates duration for which a certain canopy size is maintained in the field. It is a combined measurement between leaf area index and time. From Tables 4 & 5, it can be inferred that fertility levels exerted a significant influence on LAD at all the intervals while, residue management practices exerted a significant influence at 60 DAS –harvest only.

Among the residue management treatments, during 2021-22 and 2021-22, at 60 DAS– harvest, incorporation + microbial consortia + SSP resulted in significantly higher leaf area duration over other treatments *viz.,* incorporation + consortia, incorporation + SSP, incorporation, retention + consortia, retention, *in-situ* burning and removal.

Leaf area duration was higher with 125 % RDF over 100 % RDF and 75 % RDF. Colomb *et al.* [18] reported a 13.5 % reduction in leaf area duration in field-grown maize under phosphorous deficiency. Higher leaf area duration at higher fertility levels might be due to efficient interception and utilization of solar radiation per unit area.

The interaction effect on leaf area duration was significantly influenced by both residue management practices and fertility levels during 2021-22. At 60 DAS – harvest, significantly higher leaf area duration was recorded with the combination of incorporation + microbial consortia + SSP with 125 % RDF which was on par with incorporation + microbial consortia + SSP with 100 % RDF and incorporation + microbial consortia+ SSP with 75 % RDF. Significantly lower leaf area duration was recorded with the treatment combination of removal of residues combined with 75 % RDF which was at par with *in-situ* burning of residues with 75 % RDF, surface retention with 75 % RDF and retention + microbial consortia with 75 % RDF.

Fig. 4. Net assimilation rate (g m-2 day-1) of maize as influenced by residue management and fertilizer levels during 2021-22

Fig. 5. Leaf area duration (days) of maize as influenced by residue management and fertilizer levels during 2020-21

Fig. 6. Leaf area duration (days) of maize as influenced by residue management and fertilizer levels during 2021-22

3.5 Grain Yield (kg ha-1)

Data furnished in Tables 6 &7 indicated that grain yield was significantly influenced by residue management, fertility levels and their interaction.

Among all the residue management practices tested, incorporation + microbial consortia + SSP recorded significantly higher grain yield in comparison to the rest of the practices. During 2020-21 and 2021-22, on average, there was an increase of 23 and 26 %, respectively in incorporation + consortia + SSP treatment over *in-situ* burning treatment. In this study, it is observed that surface retention, removal, *in-situ* burning and retention + consortia recorded lower maize yield in comparison to incorporation. This was associated with an increase in root biomass carbon in residue incorporated plots, which significantly improved the yield of crops [19]. The effect of incorporation on enhancing grain yields was reported by several researchers [20-22].

Concerning different fertility levels tested, higher maize grain yield was obtained with 125 % RDF followed by 100 % RDF and 75 % RDF. The results are supported by the findings of Aulakh, [23] that higher fertility results in higher yields.

The interaction effect when tested with residue management practices and fertility levels was found significant with the treatment combinations tested. Higher grain yield was evident from incorporation + microbial consortia + SSP with 125 % RDF but it was found equivalent with that of incorporation + microbial consortia + SSP with 100 % RDF and incorporation + consortia + SSP with 75 % RDF treatment combinations. Significantly lower grain yield was recorded with the treatment combination of removal with 75 % RDF which was on par with that of *in-situ* burning with 75 % RDF, surface retention with 75 % RDF and retention + consortia with 75 % RDF combinations. Similar results were obtained by

Singh *et al.* (2014) that application of 75 % RDF + *Rhizobium* inoculation increased the root volume through better root development, nodulation, more nutrient availability resulting in vigorous plant growth and dry matter production which in turn resulted in better flowering, pod formation and ultimately seed yield.

3.6 Straw Yield (kg ha-1)

The data on maize straw yield (kg ha $^{-1}$) as influenced by residue management and fertility levels is furnished in Tables 6 &7.

Significantly higher maize straw yield (kg ha $^{-1}$) after harvest was attained by the adoption of different residue management methods. Incorporation + microbial consortia+ SSP registered significantly higher straw yield followed by incorporation + microbial consortia, incorporation + SSP and incorporation. On the other hand, significantly lower straw yield was observed under removal which was on par with *in-situ* burning, surface retention, retention + microbial consortia among the different residue management methods.

Table 7. Yield (Kg ha-1) of maize as influenced by residue management and fertility levels during 2020-21

Treatment	Grain yield (Kg ha ⁻¹)			Straw yield (Kg ha ⁻¹)				
	Fertilizer levels							
Residue management	75 % RDF (S_1)	100 % RDF (S_2)	125 % RDF (S_3)	Mean	75 % RDF (S_1)	100 % RDF (S_2)	125 % RDF (S_3)	Mean
Burning	4412	5161	5991	5188	5030	6232	7052	6105
(M_1)								
Retention (M_2)	4488	5438	5869	5265	4295	6245	7941	6160
Removal	4039	5255	6220	5171	5284	6180	6666	6043
(M_3)								
Incorporation (M_4)	4564	5418	6239	5407	4649	6553	8061	6421
Incorporation + SSP (M_5)	5381	5923	6088	5797	5611	7492	7956	7020
Retention + consortium (M_6)	4724	5355	5941	5340	5537	6254	7268	6353
Incorporation + consortia (M ₇)	6132	6211	6273	6205	7508	7581	7702	7597
Incorporation + consortium + SSP (M_8)	6487	6529	6787	6601	7932	8164	8357	8151
Mean	5028	5661	6176		5731	6838	7625	
For comparison the mean of	SEM+		CD		SEM+		CD	
Residue management	125.2		379.8		159.1		482.7	
Fertilizer levels	127.7		501.6		184.0		722.5	
Main plot at same level of sub plot	139.9		405.3		217.9		631.2	
Sub plot at same level of main plot	413.9		1199.1		666.9		1932.1	

Treatment	Grain yield (Kg ha ⁻¹)				Straw yield (Kg ha ⁻¹)			
	Fertilizer levels							
Residue	75 %	$\%$ 100	125 $\%$	Mean	75 $\%$	100 %	125 %	Mean
management	RDF	RDF	$RDF(S_3)$		RDF	RDF	RDF	
	(S_1)	(S_2)			(S_1)	(S_2)	(S_3)	
Burning	4828	5210	5807	5282	5170	6396	7219	6262
(M_1)								
Retention (M_2)	4909	5359	5723	5330	4816	6417	7797	6343
Removal	4520	5403	5801	5241	4352	6221	8041	6205
(M_3)								
Incorporation (M_4)	5383	5771	6292	5815	6206	6873	7887	6989
Incorporation +	5867	6291	6489	6216	6970	7750	7814	7511
SSP (M_5)								
Retention +	4976	5377	5854	5402	5060	6490	7839	6463
consortium (M_6)								
Incorporation +	6518	6631	6693	6614	7915	8055	8143	8038
consortia $(M7)$								
Incorporation +	6929	6971	7096	6999	8269	8568	8828	8555
consortium + SSP								
(M_8)								
Mean	5491	5877	6219		6095	7096	7946	
For comparison	SEM+		CD		SEM+		CD	
the mean of								
Residue	125.1		379.7		169.2		513.3	
management								
Fertilizer levels	82.8		325.2		167.4		657.3	
Main plot at same	100.1		290.0		218.2		632.3	
level of sub plot								
Sub plot at same	238.7		691.7		647.4		1875.4	
level of main plot								

Table 8. Yield (Kg ha-1) of maize as influenced by residue management and fertilizer levels during 2021-22

The performance of maize was better under residue addition due to the improvement of soil fertility by N mineralization from the residues resulting in higher grain and stover yields. Also, the benefits of zero tillage are accrued only when residues are retained than burnt or removed from the soil. Better crop growth and yield under zero tillage with residue addition as compared to conventional tillage with residue removal have been reported earlier by many researchers [24,25,16].

Irrespective of residue management practices,the higher straw yield was obtained with 125 % RDF as compared to 100 % RDF. The significantly lower straw yield was reported with 75 % RDF among fertility levels. This corroborates with earlier reports by Patel et al. [26], who reported that higher straw yield was obtained with 100 % RDF compared to 50 % RDF and 75 % RDF due to optimum supply of nitrogen and phosphorous which plays a crucial role in physiological processes in a plant resulting in increased growth and yield.

The significant interaction effect was noticed with residue management practices and fertility levels. Among all the tested combinations, incorporation $+$ microbial consortia $+$ SSP with 125 % RDF recorded significantly higher straw yield but it was statistically at par with that of incorporation + microbial consortia + SSP with 100% RDF and incorporation + microbial consortia + SSP with 75 % RDF combinations. On the other hand, the significantly lower straw yield was recorded with the treatment combinations of removal with 75 % RDF which was statistically comparable with that of *in-situ* burning with 75 % RDF, retention + microbial consortia with 75 % RDF and surface retention with 75 % RDF during *rabi*, 2020-21 and 2021- 22.

4. CONCLUSION

From the present study, it can be concluded that the incorporation of residues treated with microbial consortia and SSP with 75 % RDF led to enhanced growth parameters and improved yield over *in-situ* burning and removal with 125 % RDF.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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