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Effect of inoculum spraying on rice (*Oryza sativa*) residue decomposition kinetics

REKHA[1], LANDE SATISH DEVRAM[1]*, LIVLEEN SHUKLA[1], INDRA MANI[1], ROAF A PARRAY[1], SUKANTA DAS[2], KAPIL A CHOBHE[1], RAJEEV KUMAR[1] and TAPAN K KHURA[1]

ICAR-Indian Agricultural Research Institute, New Delhi 110 012, India

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ABSTRACT

Rice (*Oryza sativa* L.) residues on Indian farms have remained a major challenge due to its voluminous and unmanageable quantity and short window for sowing successive crops. A study was carried out at research farm of the ICAR-Indian Agricultural Research Institute, New Delhi during 2019–21, to develop a microbial inoculum spraying system for efficient decomposition of rice residues. The study involved the evaluation of flood nozzles at different operational parameters like operating pressure (1.5, 2.5 and 3.5 kg/cm^2), nozzle heights (50, 60 and 70 cm) and forward speeds (2.5 and 3 km/h). The inoculum spray was characterized in terms of Volume Median Diameter (VMD), Number Median Diameter (NMD) and Droplet Density (DD). The decomposition kinetics was studied in terms of total organic carbon, total nitrogen, phosphorus, potassium and N-acetyl glucosamine observed at an interval of 10 days and 20 days after inoculums application. VMD and NMD were found in the range of 347–243 µm and 77.67–87.8 µm respectively, whereas droplet density ranging from 252.5–403.9 droplets/cm^2 to spray microbial inoculum using flood nozzle. The C:N ratio of rice residue was reduced by 19.96% and 36.77% respectively after 10 and 20 days of rice residue decomposition compared to control. Carbon content reduced from 47.98–41.24% and total nitrogen content increased from 0.532–0.728 % after 20 days of decomposition. The N-acetyl glucosamine content of rice residue increased from 0.77–3.53 mg/g after 20 days indicating that target-oriented microbial inoculum spraying accelerates the decomposition kinetics of rice residues.

Keywords: Droplet density, Flood nozzle, Microbial inoculum, NMD, VMD

In India, crop residue burning has emerged as a challenge in the agricultural production system due to annual air pollution and declining soil health (Venkatramanan *et al*. 2021). The rice (*Oryza sativa* L.) crop alone contributes more than 50% of the crop residues as combine harvested rice field yield about 12.5 t/ha (Singh *et al*. 2011). The voluminous nature of rice residue and the short window of 15–20 days for the sowing of the next successive crops are major bottlenecks in management. Due to the slow degradation of rice residues within the field, sowing of the next crop instigate farmers for open field burning (Niveta Jain *et al*. 2021) and incurred nutrient loss and depletion of soil organic C. Incorporation of agricultural residues into the soil improves nutrients, soil biota, infiltration rate, water holding capacity, bulk density, cation exchange capacity and soil structure (Thanh *et al.* 2016, Lohan *et al*. 2018, Chivenge *et al*. 2019). The application of a consortium of microorganisms (fungi, bacteria, and actinomycetes) accelerates the *in-situ* incorporation of rice residue along with fungal microbial inoculum results the conversion of residues into compost and thereby it can enhance soil fertility and soil health which can lead to increased agricultural productivity, improved soil biodiversity, reduced ecological risks and a healthier environment (Singh *et al*. 2014, Zhang *et al*. 2017). The effectiveness of microbial inoculum to hasten *in-situ* decomposition can be improved through application rate and its uniformity over rice residue. The suspended fungal particles in the inoculum solution adversely affect the distribution of the microbial inoculum over the rice residue while spraying. The performance of spray nozzles to apply microbial inoculum can be improved through the assessment of spray characteristics such as droplet size, droplet density and drift. The understanding of the nexus between inoculum spray pattern and decomposition kinetics is of prime importance. This will be beneficial in enhancing the residue decomposition, thereby, improving the soil health and reducing the menace of rice residue burning.

[1]ICAR-Indian Agricultural Research Institute, New Delhi; [2]ICAR-Indian Agricultural Statistics Research Institute, New Delhi. *Corresponding author email: satishiari@gmail.com

MATERIALS AND METHODS

The present study was carried out at research farm of the ICAR-Indian Agricultural Research Institute, New Delhi during 2019-21 to investigate the rice residue decomposition kinetics due to variation in the sprayer design parameters. The understanding of rice residue decomposition pattern in a combine harvested field and sprayed with fungal microbial inocula (Pusa Decomposer, a consortium of seven fungi). Operating conditions were simulated in the laboratory similar to that of the combine harvested field condition to facilitate *in-situ* decomposition of rice residue as decomposition kinetics were greatly influenced due to rate of application and uniformity of microbial over the residues. To apply microbial inocula over the rice residues (loose straw and stubbles) after combine harvesting; an experimental set-up was designed and developed for laboratory simulation. It facilitates the spraying of microbial inocula over the rice residues as per the forward speeds of combine harvesting. To simulate crop parameters, several rice residue samples (var. Pusa Sugandh 5) were collected from experimental farms during combine harvesting by SMS-mounted combine harvester. As the application rate of microbial inoculum was influenced by nozzle operating pressure and forward speed of the combine harvester, similar spraying conditions were maintained for selected forward speeds, i.e. 2.5 and 3.5 km/h. The fungal microbial inoculum was diluted with water with a dilution ratio of 1:10 and sprayed over the rice residue with various combinations of pressure levels and forward speeds.

Microbial inoculum spraying system comprises flood type nozzle, stepper motor (Torque-10.0 kg-cm), motor driver, timing gear belt and pulley, diaphragm pump (Capacity: 4–6 l/min), pressure relief valve, storage tank and battery (12V, 7AH capacity). A stepper motor and timing gear belt and pulley were used to run the nozzle at different speeds. The flood nozzle was used to spray fungal microbial inoculum as the spray solution contains suspended fungi particles diluted with water. Also, the operating pressure of the flood nozzle ranges between 1.53–5.57 kg/cm^2 to deliver discharge of 3.35–5.12 l/min with a spray width of 2.5–3.4 m for wider coverage as compared to other spray nozzles.

The performance of the flood nozzle was evaluated for operating pressure (1.5, 2.5 and 3.5 kg/cm^2), the height of the nozzle (50, 60 and 70 cm) and forward speed (2.5 and 3.0 km/h). The effect of operational parameters on spray deposition in terms of droplet size (VMD and NMD) and droplet density (DD) for flood-type nozzles were studied by using water-sensitive papers. To study rice residue decomposition kinetics, the stubbles of size 10-30 cm and loose residue of size 5–20 cm were placed in plastic trays which were positioned exactly below the center of the nozzle assembly.

Water-sensitive papers were placed on rice stubbles to qualify the spray deposition of fungal microbial inoculum. Scanned water-sensitive papers (Image resolution: 600 dpi in .jpg format) were used to determination of the spray droplet size and droplet density by using ImageJ (Product version: 2006.02.01) software. These images were further processed with a threshold tool and USDA Automatic paper analysis tool to obtain data on Volume Median Diameter (VMD) and Number Median Diameter (NMD) and Droplet Density (DD) as these spray parameters influence microbial inoculum penetration into the residue, spray uniformity and percentage of spray coverage over the rice residues and ultimately accelerated decomposition of rice residues.

VMD is a droplet diameter where 50% of the total liquid volume is in a droplet of a smaller diameter; whereas NMD is defined as a diameter that divides the total number of droplets into two equal parts, i.e. half the total number of droplets is contained in smaller droplets and half in larger droplets (Lefebvre A H 1993).

Droplet density: The droplet density signifies the quality of the spray as it directly affects the volume of the spray. The ratio of the total count of droplets per unit area was used to compute droplet density.

The rice residue decomposition study was carried out for the straw samples the after being inoculated with fungal microbial inocula through a spraying experimental setup. The collected residue samples were used to quantify the decomposition pattern in terms of total organic carbon, total nitrogen, phosphorus, potassium and N-acetyl glucosamine in intervals of 10 days and continued till 20 days. The effect of spray operational parameters on rice residue decomposition was studied and analysed (Table 1).

Ash carbon of rice residue was measured by a muffle furnace. Residue samples of 5 g were taken and placed in a crucible specially designed to withstand the muffle furnace's high temperature (500°C). The weight of the samples was again taken after 24 h to determine carbon content (Anisuzzaman *et al*. 2015, Devi *et al*. 2017).

$$\text{Ash carbon (\%)} = \frac{\text{Weight of sample after process}}{\text{Weight of sample before process}} \times 100$$

Total nitrogen: Total nitrogen was measured by the Kjeldahl method which consists of three processes, i.e.

Table 1 Plan of an experiment for rice residue decomposition

Independent variables	Levels	Dependent variable
Pressure	3 (1.5, 2.5 and 3.5 kg/cm^2)	Carbon Total nitrogen
Speed	2 (2.5 and 3 km/h)	C:N ratio Phosphorous Potassium N-acetyl glucosamine

Treatment:

T_0, Rice residue without spray

T_1, Rice residue with spray at 2.5 km/h speed and pressure of 1.5 kg/cm^2

T_2, Rice residue with spray at 2.5 km/h speed and pressure of 2.5 kg/cm^2

T_3, Rice residue with spray at 2.5 km/h speed and pressure of 3.5 kg/cm^2

T_4, Rice residue with spray at 3 km/h speed and pressure of 1.5 kg/cm^2

T_5, Rice residue with spray at 3 km/h speed and pressure of 2.5 kg/cm^2

T_6, Rice residue with spray at 3 km/h speed and pressure of 3.5 kg/cm^2

digestion, distillation and titration (Jackson 1973, Bremner and Mulvaney 1982).

$$\text{Total nitrogen (\%)} = \frac{\{(\text{ml of } H_2SO_4 \text{ sample} - \text{ml of } H_2SO_4 \text{ blank}) \times N \times 4\}}{\text{Weight of the sample}} \times 100$$

where N, normality of H_2SO_4.

Phosphorus: The phosphorus content of the rice residue was measured using a spectrophotometer using 420 nm wavelengths. The phosphorus concentration based on absorbance versus concentration was determined using a standard curve.

Potassium: The potassium of rice residue was measured by using a flame photometer (Knudsen *et al.* 1982, Devi *et al.* 2017). The potassium concentration was calculated with standard reference and using the below given formula.

$$\text{Potassium (\%)} = \text{Concentration of K in ppm from graph} \times \frac{\text{Dillution factor} \times 100}{\text{Weight of sample (g)} \times 1000}$$

N-acetyl glucosamine: The N-acetyl glucosamine content was determined to represent variation in microbial fungus growth through biochemical changes in structural components (Kumar *et al.* 2008) of the samples of the residues based on absorbance using spectrophotometer at 530 nm wavelength.

The data obtained were statistically analyzed using SAS statistical software. Multivariate analysis was used to interpret treatment main effects, interaction effects, and significance at different treatment combinations.

RESULTS AND DISCUSSION

Microbial inoculum application to the rice residue alters composition of rice residue nutrients due to the action of microbes during decomposition. The variation in spray droplet size was determined at three different nozzles operating pressure, nozzle height and nozzle operating speed. A study was carried out to decompose rice residue within an available time window of 15–20 days before sowing of next successive *rabi* crops. *In-situ* rice residue decomposition can be accelerated through proper spraying system design parameters as it depends on the amount of microbial inoculum applied and its deposition pattern over the residues. The rate of decomposition of rice residue was quantified at different time intervals, i.e. before microbial inoculum application (0 days) and after microbial inoculum application (10 and 20 days).

Variation in droplet size of fungal microbial inocula: The droplet size of microbial inoculum in terms of VMD and NMD was determined and influenced by the spray operational parameters. VMD was found in the range of 347–243 µm. It decreased with an increase in pressure and decrease in speed and vice-versa. VMD decreased by 2.59%, 2.71% and 2.78%, respectively for operating pressures of 1.5 kg/cm^2, 2.5 kg/cm^2 and 3.5 kg/cm^2 during change in forward speed from 2.5 to 3 km/h at 50 cm height. Similar trends were obtained at 60 and 70 cm height of the nozzle (Fig 1). NMD was also found to decrease in the range of 87.08–77.67 µm and 86.16–75.41 µm 2.5 km/h and 3 km/h forward speed, respectively. It decreased with an increase in pressure and decrease in speed and vice-versa. With increase in forward speed from 2.5–3 km/h at 50 cm height and for operating pressure of 1.5 kg/cm^2, 2.5 kg/cm^2 and 3.5 kg/cm^2, NMD decreased by 1.06%, 2.63% and 2.9% respectively. Similar trends were obtained at 60 and 70 cm height of the nozzle (Fig 1).

Variation in droplet density of the fungal microbial inocula: The variation in microbial inoculum droplets applied over the rice residue influenced the decomposition. Droplet density was found in the range of 275–403.9 droplets/cm^2 and 252.5–395 droplets/cm^2 at 2.5 km/h and 3 km/h forward speeds, respectively. It increased with increase in pressure and decrease in forward speed and vice-versa. At 50 cm nozzle height, droplet density increased by 1.98%, 1.95% and 2.25% respectively for operating pressure of 1.5 kg/cm^2, 2.5 kg/cm^2 and 3.5 kg/cm^2 during increase in forward speed from 2.5 to 3 km/h. SA similar trend of decreasing was obtained at 60 and 70 cm height of nozzle (Fig 1). The effect of different pressures of 1.5, 2.5 and 3.5 kg/cm^2 and different speeds of 2.5 and 3 km/h on droplet density was found significant at 5% level of significance. However, the interaction effect of treatments was found insignificant. A

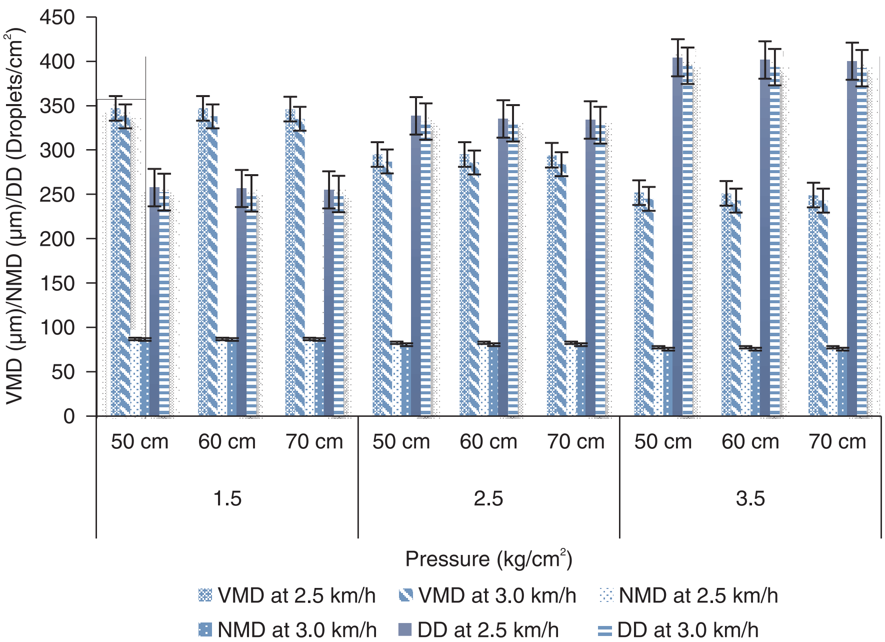

Fig 1 Effect of nozzle operating pressure and operating speed on droplet size (VMD and NMD) and Droplet Density (DD).

Table 2 ANOVA for the effect of operational parameters of flood type nozzle on droplet density

ANOVA Table - Dependent Variable: Droplet Density (DD)

Source	DF	Sum of squares	Mean square	F-ratio	P-value
Replication	2	462.9777	231.4889	3.9196	0.0294*
Pressure	2	186979.6327	93489.8164	1582.9891	<.0001*
Speed	1	545.2431	545.2431	9.2322	0.0045*
Pressure × Speed	2	23.7242	11.8621	0.2009	0.8190
Height	2	100.7200	50.3600	0.8527	0.4352
Pressure × Height	4	5.9460	1.4865	0.0252	0.9987
Speed × Height	2	1.3289	0.6645	0.0113	0.9888
Pressure × Speed × Height	4	2.5938	0.6484	0.0110	0.9997
Error	34	2008.0074	59.0590	.	.
Total	53	190130.1740	.	.	.

*Significance at 5% level.

high value of R^2 of >0.98 was observed which showed the strong relationship between droplet density with operating pressure and forward speed (Table 2).

Variation in carbon, total nitrogen and C: N of rice residue: During the decomposition of rice residue, the broken down complex residue molecules into simpler ones by the action of microbes resulted in a change of carbon content from 47.98–41.24% and total nitrogen content from 0.532–0.728% after 20 days interval of decomposition (Table 3). The treatment T_3 and T_6 showed a higher decrease in the carbon content of rice residue of 47.08–41.24% compared to the remaining T_1, T_2, T_4 and T_5 treatments. The carbon content of rice residue was reduced by 5.73% and 11.68% after 10 and 20 days of decomposition compared to untreated. The treatment T_3 and T_6 showed a higher increase in total nitrogen content of rice residue ranging from 0.532–0.728% compared to the remaining T_1, T_2, T_4 and T_5 treatments. Similarly, the total nitrogen content of rice residue showed 17.85% and 39.66% higher after 10 and 20 days of decomposition compared to untreated. The results of the study revealed that there was a change in carbon and total nitrogen content of rice residue with increase in a decomposition time interval.

Reduction in C:N ratio of rice residue is considered one of the important criteria in assessing its stability and maturity. Higher initial C: N ratio of the residues slow down the rate of decomposition, whereas a low initial C: N ratio causes strong NH_3 emission thereby increase in decomposition rate. The results of the study revealed that the reduction in C:N ratio ranged from 88.49:1 to 54.55:1 after 20 days of decomposition (Fig 2). The maximum decrease on C:N ratio was found with T_3 and T_6 treatment compared to remaining T_1, T_2, T_4 and T_5 treatments. The C:N ratio of rice residue was reduced to 19.96% and 36.77% after10 and 20 days of rice residue decomposition compared to control. It was decreased with an increase in time of decomposition. The spraying of microbial inoculum over the rice residue influenced the decomposition in terms of reduction in the C: N ratio. The change in carbon and total nitrogen content of rice residue was found significant for decomposition time at a 5% level of significance.

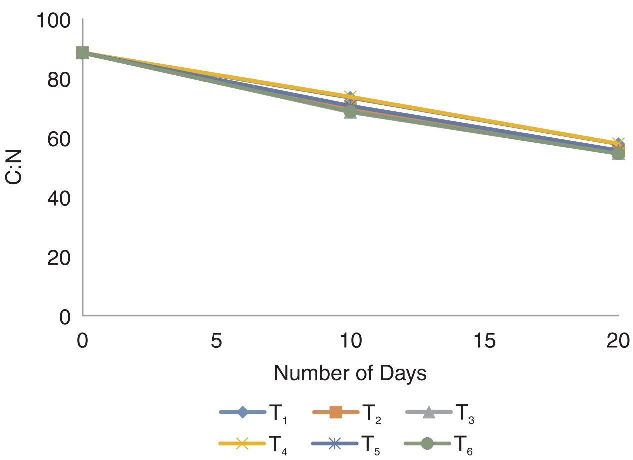

Fig 2 Variation in C : N ratio of rice residue under different treatment.

Table 3 ANOVA for the effect of decomposition time on carbon content, total nitrogen, potassium, phosphorus and N-acetyl glucosamine of rice residue under different treatments

Variable	DF	C		N		K		P		N-acetyl glucosamine	
		F value	P value	F value	P value	F value	P value	F value	P value	F value	P value
Treatment	5	1.955	0.158	1.440	0.279	10.147	0.0005*	0.103	0.989	2.721	0.072
Time	2	123.426	<.0001*	1319.938	<.0001*	558.872	<.0001*	12.137	0.0002*	6089.338	<0.0001*
Treatment × Time	10	0.307	0.972	0.9301	0.524	2.745	0.0207*	0.015	1.000	0.472	0.892

DF, Degree of Freedom; *Significance at 5% level.

Change in phosphorus and potassium content of rice residue under different treatments: The change in phosphorus and potassium content of residue during decomposition was evaluated. An increase in phosphorus content from 0.072–0.753 and potassium content from 0.116–0.157% after 20 days of rice residue decomposition was observed. The results of the study revealed that, there was increase in phosphorus and potassium and content with increase in time of decomposition. The treatment T_3 and T_6 showed higher increase in both phosphorus and potassium content compared to that in T_1, T_2, T_4 and T_5 treatments. The phosphorus content of rice residue showed 2.78% and 4.16% higher after 10 and 20 days of decomposition compared to before microbial inoculum application. Similarly, the potassium content of rice residue showed 15.5% and 30.17% higher after 10 and 20 days of decomposition compared to without microbial inoculum. The change in potassium and phosphorus content of rice residue was found significantly affected by varying decomposition times at 5% level of significance. However, the effect of treatment and interaction effect of treatment and time was found to be insignificant**.**

Change in fungal growth in terms of N-acetyl glucosamine of rice residue under different treatments: The change in microbial fungus growth influences the rice residue decomposition. The effect of spray parameters on change in fungus growth was studied in terms of change in N-acetyl glucosamine content of rice residue. The change in N-acetyl glucosamine content of rice residue increased from 0.77–3.53 mg/g of residue sample after 20 days of decomposition. The N-acetyl glucosamine content of rice residue showed 1.61 times and 3.51 times higher after 10 and 20 days of decomposition compared to untreated. The results of the study revealed an increase in fungus growth with increase in decomposition time of rice residue. From the ANOVA, the change in N-acetyl glucosamine of rice residue was found significantly affected by decomposition time at 5% level of significance.

Rice residue decomposition was influenced by VMD, NMD and droplet density of microbial inoculum spraying. The precision and uniformity in the application of microbial inoculum over the rice residue were affected by operational pressure and forward speed of operations and not dependent on nozzle operating height, whereas their interaction effects were found insignificant. The significant variation in C:N ratio of rice residue was found to decrease with an increase in decomposition time, i.e. after 20 days indicating that the target-oriented microbial inocula spraying system can hasten the rate of decomposition of rice residue for *in-situ* management.

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