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Influence of Inorganic Fertilizers with Nano Urea on Soil Health Parameters and Quality Parameters under different planting geometry of Rice Crop (*Oryza sativa* L.)

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Abstract

An experiment was conducted at the Agronomy Research Farm of Acharya Narendra Deva University of Agriculture and Technology in Kumarganj, Ayodhya, Uttar Pradesh, India, during the Kharif seasons of 2022 and 2023. The study aimed to evaluate the effects of planting geometry and inorganic fertilizers, combined with nano urea, on soil health parameters and quality of rice crop. The experiment utilized a split plot design with three replications, incorporating three planting geometries—C_1 (20 cm × 10 cm), C_2 (20 cm × 15 cm), and C_3 (20 cm × 20 cm) in the main plot. The subplot treatments included five fertilizer levels: F_1 (100% RDF (150:60:40)), F_2 (100% RDF + two foliar sprays of nano urea at 3000 ml ha^{-1} during the Tillering and Panicle Initiation stages), F_3 (75% RDF + two foliar sprays of nano urea), F_4 (50% RDF + two foliar sprays of nano urea), and F_5 (Control with no fertilizers). Results showed that the combination of treatment C_1 (20 cm × 10 cm) with F_2 (100% RDF + two foliar sprays of nano urea at 3000 ml ha^{-1} during the Tillering and Panicle Initiation stages) was optimal for enhancing the soil health and quality of rice grain, resulting in maximum yield attributes and yield (q ha^{-1}) of rice across both years of the study.

Keywords.
PI, Nano urea.

1. Introduction

Rice (Oryza sativa L.) is a vital staple food originating from Southeast Asia, accounting for 20% of global calorie consumption. As the second-largest producer of rice, India cultivates it on 450.57 lakh hectares, yielding 122.27 million tons annually. However, productivity lags behind that of many other countries, highlighting the potential for improvement. Transplanting seedlings at a closer spacing of 20 cm × 10 cm resulted in significantly higher grain (63 q ha^{-1}) and straw (162 q ha^{-1}) yields compared to wider spacings of 20 cm × 20 cm and 20 cm × 15 cm, although it was comparable to the 15 cm × 15 cm planting geometry **(Kawat *et al.,* 2002)**. The soil nitrogen availability is often insufficient **(Vigneau *et al.,* 2011),** leading farmers to apply more nitrogen fertilizers to achieve higher crop yields. This can result in nitrogen over-fertilization, which ultimately hinders optimal plant productivity **(Rubio-Covarrubias *et al.,* 2009)**. Foliar applications can improve nutrient absorption and minimize soil pollution. Nano-fertilizers, especially the innovative nano-urea developed by IFFCO, offer a cost-effective and controlled nutrient supply. Over 1 mm of urea prill contains 55,000 nitrogen particles **(Prem Baboo, 2021).** As a more cost-effective option than conventional urea, nano urea is currently considered the superior alternative to traditional urea fertilizers.

2. MATERIAL AND METHODS

The experiment was carried out at the Agronomy Research Farm of Acharya Narendra Deva University of Agriculture and Technology in Kumarganj, Ayodhya, Uttar Pradesh, during the Kharif seasons of 2022 and 2023. The site has a sub-tropical climate and features alluvial calcareous soil, categorized as silty loam, which is low in organic carbon and nitrogen but moderate in phosphorus and high in potassium. Utilizing a Split Plot Design with three replications, the study focused on three planting geometries: C_1 (20 cm × 10 cm), C_2 (20 cm × 15 cm), and C_3 (20 cm × 20 cm). The subplot treatments consisted of five fertilizer levels: F_1 [100% RDF (150:60:40)], F_2 [100% RDF plus foliar spray of nano urea at 3000 ml ha^{-1} during the Tillering and Panicle Initiation stages], F_3 [75% RDF plus nano urea], F_4 [50% RDF plus nano urea], and F_5 [Control with no fertilizers]. The rice variety Sarju-52 was manually transplanted on July 2, 2022, and July 13, 2023.

Nano urea, obtained from the Indian Farmers Fertilizer Cooperative Limited (IFFCO), was applied at a rate of 3000 ml ha^{-1} (4 ml per liter) in two doses during the Tillering and Panicle Initiation stages.

2.1 Soil health parameters:

Soil samples were collected from a surface depth of 0-15 cm near the plant root (rhizosphere) and were taken in small polythene bags from each plot according to treatments. The soil samples were thoroughly air-dried, ground, and passed through a 2 mm mesh sieve and analysed for microbial Enzymatic parameters, *i.e.,* urease activity, dehydrogenase activity and alkaline phosphatase activity.

2.1.1 urease activity

Urease activity in soil was assayed by quantifying the ratio of release of NH4 + from the hydrolysis of urea (Tabatabai and Bremner, 1972). 5g of soil was taken in a 50 ml volumetric flask, after adding 0.2 ml of toluene and 9 ml THAM buffer, the flask was swirled for a few seconds to mix the contents and 1ml of 0.2M urea solution was added and swirled the flask again for a few seconds. Then the flask was stoppered and placed in an incubator at 37°C. After 2 hours, the stopper was removed, and approximately 35 ml of KCl-Ag2SO4 solution was added, swirled the flask for a few seconds, and allowed the flask to stand until the contents have cooled to room temperature (about 5 min). The contents were made to 50 ml by addition of KCl-Ag2SO4 solution, the flask was stoppered and inverted several times to mix the contents. NH+4- N was determined in the resulting soil suspension, by pipetting out 20 ml aliquot of the suspension distilling with 0.2 g of MgO for 4 min. The ammonium boric acid complex was titrated against standard 0.05 N H_2SO_4.Controls were performed by following the procedure described for assay of urease activity, but for the addition of 1ml of 0.2M urea solution after the addition of KCl-Ag_2SO_4 solution. The further urease activity was calculated and expressed in (µg NH+4 -N released g^{-1} $2h^{-1}$).

2.1.2 Alkaline Phosphatase activity

Alkaline phosphatase activity was estimated as described by Tabatabai and Bremner (1969). For its determination, modified universal buffer (MUB; pH 11), Toluene p-nitrophenyl phosphate (PNP), calcium chloride, and sodium hydroxide reagents were used. For estimation of alkaline phosphatase activity, 2.0 g of air-dried soil samples were taken and moistened with 2.0 ml of distilled water followed by incubation for 24 hours. Following incubation, 0.2 ml of toluene, 4.0 ml of MUB, and 1.0 ml of freshly prepared PNP were added and thoroughly mixed before incubating at 37 ^{0}C for 1.0 hours. After incubation, 1.0 ml of 0.5 M $CaCl_2$, and 4.0 ml of 0.5 M NaOH were added and mixed thoroughly. The contents were filtered and centrifuged for 10 minutes at 10,000 rpm. The supernatant solution was collected and used for recording the absorbance of a spectrophotometer at 400 nm. A standard curve was prepared to calculate alkaline phosphatase activity and the values were expressed in terms of µg g^{-1} of soil day^{-1}.

2.1.3 Dehydrogenase activity

Dehydrogenase enzyme activity was estimated as per the method described by Casida *et al.* (1964) using reagents like triphenyl tetrazolium chloride (TTC) and methanol (AR grade). For estimation of dehydrogenase activity, 5 g of soil was taken in a screw-capped test tube and moistened with 1.0 ml of distilled water followed by incubation for 24 hours at 30 ^{0}C. After incubation, samples were added to 1.0 ml of freshly prepared 3% TTC solution. These test tubes were incubated in the dark for 24 hours at 30 °C. After incubation, 10 ml of methanol was added to the test tubes, as an extracting agent, and the contents were well mixed by rotation. The contents were filtered and centrifuged for 10 minutes at 10,000 rpm. A supernatant solution [Tri phenyl formazan (TPF) – pink colour] was collected and used for recording the absorbance of a spectrophotometer at 485 nm. The concentration of TTF in the soil was determined by the standard curve prepared. Further dehydrogenase activity was calculated and expressed in µg TPF g^{-1} soil, 24 h^{-1}.

2.2 Protein content in grain (%):

Protein content in rice grains was estimated separately by multiplying the nitrogen content of grain '6.25' as determined by modified Nessler's reagent method. The nitrogen content was multiplied by a factor 6.25 (AOAC, 1970).

Protein content (%) = Nitrogen content (%) × 6.25

3. RESULTS AND DISCUSSION

3.1 Soil Health Parameters

3.1.1 urease activity (µg NH+4 -N released g^{-1} $2h^{-1}$)

The data pertaining to urease activity in soil after harvest of rice crop as influenced by treatment are presented in Table:1 clearly indicates that planting geometry and inorganic fertilizers with nano urea have significant effect on urease activity during both the years of experiment.

Data further revealed that maximum urease activity 226.27 and 241.48 (µg NH+4 -N released g^{-1} $2h^{-1}$) during the year 2022 and 2023 respectively was recorded under treatment C_1 (20 cm × 10 cm), which was at par with C_2 (20 cm × 15 cm) and was significantly higher than rest of the treatments.

Among fertilizer levels, application of F_2 [100% RDF + Two foliar spray of nano urea @ 3000 ml ha^{-1} (Tillering and PI stage)] was recorded significantly maximum urease activity *i.e.,* 227.32 and 238.25 (µg NH+4 -N released g^{-1} $2h^{-1}$) during the year 2022 and 2023 respectively, which was at par with application of F_3 [75% RDF + Two foliar spray of nano urea @ 3000 ml ha^{-1} (Tillering and PI stage)] and F_1 (100% RDF; 150:60:40). While significantly higher than the rest of the fertilizer levels during both years. It might be due to soil urease which plays a major role in catalysis of the hydrolysis of urea to ammoniacal form, which will be subsequently oxidized by nitrifiers to nitrate form, which increases the utilization rate of nitrogen fertilizer. Similar results were also reported by Senthil kumar *et al.* (2000) and Ramlakshmi (2011).

3.1.2 Alkaline Phosphatase activity (µg *P-Nitrophenol* g^{-1} of soil day^{-1})

The data pertaining to Alkaline Phosphatase activity in soil after harvest of rice crop as influenced by treatment are presented in Table:2 clearly indicates that planting geometry and inorganic fertilizers with nano urea have significant effect on dehydrogenase activity during both the years of experiment.

Data further revealed that maximum Alkaline Phosphatase activity 181.43 and 196.09 (µg *P-Nitrophenol* g^{-1} of soil day^{-1}) during the year 2022 and 2023 respectively was recorded under treatment C_1 (20 cm × 10 cm), which was at par with C_2 (20 cm × 15 cm) and was significantly higher than rest of the treatments.

Among fertilizer levels, application of F_2 [100% RDF + Two foliar spray of nano urea @ 3000 ml ha^{-1} (Tillering and PI stage)] was recorded significantly maximum alkaline phosphatase activity *i.e.,* 182.02 and 198.01(µg *P-Nitrophenol* g^{-1} of soil day^{-1}) during the year 2022 and 2023 respectively, which was at par with application of F_3 [75% RDF + Two foliar spray of nano urea @ 3000 ml ha^{-1} (Tillering and PI stage)]. While significantly higher than the rest of the fertilizer levels during both years. Similar results were also reported by Dotaniya *et al.* (2014).

3.1.3 Dehydrogenase activity (µg TPF g^{-1} soil, 24 h^{-1})

The data pertaining to dehydrogenase activity in soil at after harvest of rice crop as influenced by treatment are presented in Table:2 clearly indicates that planting geometry and inorganic fertilizers with nano urea have significant effect on dehydrogenase activity during both the years of experiment.

Data further revealed that maximum dehydrogenase activity 153.75 and 166.87 (μg TPF g^{-1} soil, 24 h^{-1}) during the year 2022 and 2023 respectively was recorded under treatment C_1 (20 cm × 10 cm) being at par with C_2 (20 cm × 15 cm), which was significantly higher than rest of the treatments.

Among fertilizer levels, application of F_2 [100% RDF + Two foliar spray of nano urea @ 3000 ml ha^{-1} (Tillering and PI stage)] was recorded significantly maximum dehydrogenase activity *i.e.,* 155.35 and 163.64 (μg TPF g^{-1} soil, 24 h^{-1}) during the year 2022 and 2023 respectively, which was at par with application of F_3 [75% RDF + Two foliar spray of nano urea @ 3000 ml ha^{-1} (Tillering and PI stage)]. While significantly higher than the rest of the fertilizer levels during both years. The dehydrogenase activity is commonly used as an indicator of biological activity in soils which are essential for maintaining soil fertility. Similar results were also reported by Burns, (1978) and Joachim *et al.,* (2008).

3.2 Quality Parameters

3.2.1 Protein content in grain (%)

The data on protein content in grain of rice as influenced by that planting geometry and inorganic fertilizers with nano urea was recorded for both the cropping seasons have been presented in Table:2

Data pertaining to protein content (%) of grain in rice clearly indicates that planting geometry has non-significant effect on protein content (%) in grain during both the years of experiment. However, maximum protein content 8.44 and 8.75 % in grain was recorded under treatment C_1 (20 cm × 10 cm) during year 2022 and 2023. The minimum protein content 7.50 and 7.69 % in grain was recorded under treatment C_3 (20 cm × 20 cm) during year 2022 and 2023. Similar findings were also found by Singh and Verma (2006), Dewedi *et al.,*(2006).

Among fertilizer levels, application of F_2 [100% RDF + Two foliar spray of nano urea @ 3000 ml ha^{-1} (Tillering and PI stage)] was recorded significantly maximum protein content in grain *i.e.,* 8.50 and 8.81 % in both the year 2022 and 2023 respectively, which was at par with application of F_3 [75% RDF + Two foliar spray of nano urea @ 3000 ml ha^{-1} (Tillering and PI stage)] and F_1 (100% RDF; 150:60:40). While significantly higher than the rest of the fertilizer levels during both years. This increment in protein content might be due to increment in nitrogen content in grain and straw, as nitrogen (N) in vegetative organs (tillers and leaves). Similar findings were also found by Havlin *et al.* (2014), and Khanday *et al.* (2017).

3.2.2 Protein yield (Kg ha^{-1})

The data pertaining to protein yield of rice as influenced by that planting geometry and inorganic fertilizers with nano urea recorded for both the cropping seasons have been presented in Table:2.

Data pertaining to protein yield of rice clearly indicates that planting geometry have significant effect on protein yield of rice during both the years of experiment. The maximum protein yield 407.65 and 441.00 kg ha^{-1} was recorded under treatment C_1 (20 cm × 10 cm), which was at par with C_2 (20 cm × 15 cm), during year 2022 and 2023 and was significantly higher than rest of the treatments. Similar findings were also found by Singh and Verma (2006).

Among fertilizer levels, application of F_2 [100% RDF + Two foliar spray of nano urea @ 3000 ml ha^{-1} (Tillering and PI stage)] was recorded significantly maximum protein yield *i.e.,* 455.60 and 488.07 kg ha^{-1} during the year 2022 and 2023 respectively, which was at par with application F_3 [75% RDF + Two foliar spray of nano urea @ 3000 ml ha^{-1} (Tillering and PI stage)] and F_1 (100% RDF; 150:60:40). While significantly higher than the rest of the fertilizer levels during both years. This increment in protein content (%) might be due to increment in nitrogen content in grain and straw, as nitrogen (N) in vegetative organs (tillers and leaves) is gradually transferred to developing grain and participates in protein synthesis after plants enter reproductive growth. Similar findings were also found by Havlin *et al.* (2014), and Khanday *et al.* (2017).

4. CONCLUSION

Based on the above findings, it is evident that treatment C_1 (20 cm × 10 cm) coupled with the application of F_2 [100% RDF + foliar spray of nano urea @ 3000 ml ha^{-1} (during Tillering and PI stages)] exhibited good soil health parameters; urease activity (µg NH+4 -N released g^{-1} 2h^{-1}), Alkaline Phosphatase activity (µg *P-Nitrophenol* g^{-1} of soil day^{-1}) and Dehydrogenase activity (µg TPF g^{-1} soil, 24 h^{-1}) as well as quality parameters viz; protein content in grain (%) and protein yield (Kg ha^{-1}) of rice across both 2022 and 2023.

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Table: 1 Effect of different planting geometry and inorganic fertilizers with nano urea on soil urease activity (µg NH+4 -N released g^{-1} $2h^{-1}$), Alkaline phosphatase activity (µg *P-Nitrophenol* g^{-1} of soil day^{-1}.) and dehydrogenase activity in soil (µg TPF g^{-1} soil, 24 h^{-1}) at harvest of rice crop.

Treatments	Urease		Alkaline phosphatase		Dehydrogenase	
	2022	2023	2022	2023	2022	2023
(A) Planting geometry						
C_1 - 20 cm×10 cm	226.27	241.48	181.43	196.09	153.75	166.87
C_2 - 20 cm×15 cm	214.55	225.90	169.24	185.24	143.36	155.29
C_3 - 20 cm×20 cm	205.67	212.63	160.36	175.62	133.15	138.02
SEm±	4.14	4.28	3.51	3.67	2.80	3.01
C.D. (P=0.05)	16.67	17.26	14.14	14.81	11.27	12.03
(B) Fertilizers levels						
F_1 - 100% RDF (150:60:40)	216.62	227.55	171.09	187.31	144.10	152.94
F_2 -100% RDF + Two foliar spray of nano urea @ 3000 ml ha^{-1} (Tillering and PI stage)	227.32	238.25	182.01	198.01	155.35	163.64
F_3 - 75% RDF + Two foliar spray of nano urea @ 3000 ml ha^{-1} (Tillering and PI stage)	220.41	231.34	175.10	188.43	147.89	156.73
F_4- 50% RDF + Two foliar spray of nano urea @ 3000 ml ha^{-1} (Tillering and PI stage)	210.74	216.67	165.43	181.43	138.22	147.06
F_5 - Control (no fertilizers)	202.39	214.54	157.08	173.08	129.87	139.93
SEm±	4.47	4.60	3.53	3.61	2.96	3.10
C.D. (P=0.05)	13.05	13.44	10.29	10.55	8.63	9.04

Table: 2 Effect of different planting geometry and inorganic fertilizers with nano urea on Protein content in grain (%) and Protein yield (Kg ha^{-1}) of rice crop.

Treatments	Protein content in grain (%)		Protein yield (Kg/ha)	
	2022	2023	2022	2023
(A) Planting geometry				
C_1 - 20 cm×10 cm	8.44	8.75	407.65	441.00
C_2 - 20 cm×15 cm	7.75	8.06	358.05	383.66
C_3 - 20 cm×20 cm	7.50	7.69	318.00	342.97

SEm±	0.11	0.15	13.70	15.01
C.D. (P=0.05)	**NS**	**NS**	55.23	60.53
(B) Fertilizers levels				
F_1 - 100% RDF (150:60:40)	8.00	8.31	395.20	427.97
F_2 -100% RDF + Two foliar spray of nano urea @ 3000 ml ha^{-1} (Tillering and PI stage)	8.50	8.81	455.60	488.07
F_3 - 75% RDF + Two foliar spray of nano urea @ 3000 ml ha^{-1} (Tillering and PI stage)	8.31	8.63	433.78	469.47
F_4- 50% RDF + Two foliar spray of nano urea @ 3000 ml ha^{-1} (Tillering and PI stage)	7.88	8.13	372.72	400.00
F_5 - Control (no fertilizers)	7.56	7.63	254.02	267.81
SEm±	0.17	0.21	20.90	22.33
C.D. (P=0.05)	**0.51**	**0.62**	61.26	65.21