Nitrogen Contents in Soil, Grains, and Straw of Hybrid Rice Differ When Applied with Different Organic Nitrogen Sources

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Abstract: In the rice–wheat (R–W) system, inorganic nitrogen (N) fertilizer (urea, etc.) is the largest component of the N cycle, because the supply of N from organic fertilizers is insufficient. But the 4% Initiative aims to improve organic matter and stimulate carbon sequestration in soils using best agronomic practices (sustainable management practices) which are economically, environmentally, and socially friendly. This research project was, therefore, designed to assess the impact of various organic sources (OS, animal manure versus plant residues), inorganic N (urea), and their different combinations on the N concentrations in soils and plants (i.e., grains and straw) of hybrid rice plants. The experiments were conducted on farmers' fields in Batkhela (Malakand), northwestern Pakistan, over 2 years (2011–2012 (Y1) and 2012–2013 (Y2)). The results revealed that N concentrations in soil as well as in rice plants ranked first when applied with urea-N, followed by the application of N in mixture (urea + OS), while the control plots (no N applied) ranked at the bottom. Among the six OS (three animal manures: poultry, sheep, and cattle; and three crop residues: onion, berseem, and wheat), application of N in the form of poultry manure was superior in terms of higher N concentrations in both soil and plants. Applying the required total N (120 kg N ha^{-1}) in the form of 75% N from urea +25% N from OS resulted in higher N concentrations in soil and plants in Y1. The required total N (120 kg N ha^{-1}) application in the form of 50% N from urea +50% N from OS produced higher N concentrations in soil and plants in Y2. It was concluded from the results, that combined application of N sources in the form of urea + OS can produce good performances in terms of higher N concentrations in soil as well as in rice plants under the R–W system. Integrated use of urea (N-fertilizer) with organic carbon sources (animal manures and crop residue) could sustain rice-based (exhaustive) cropping system.

Keywords: rice; organic source; urea; nitrogen; ratio; N concentration in soil; N concentration in rice plants

1. Introduction

Rice (*Oryza sativa* L.) and wheat (*Triticum aestivum* L.) are the two most commercially important crops, accounting for more than 40% of world food production. Farmers in Asia have been using the R–W system for more than 1000 years. Continuous cultivation of R–W system has resulted in an increase in nutrient mining and a decrease in yields of both crops [1]. The R–W crop-system is extremely nutrient-exhaustive and, thus, its continuous use has decreased soil fertility, leading to

nutrient deficiency [1–3]. Rice is very important food human and contribute about 35–60 of the dietary calories for almost 3 billion people [4]. The irrigated lowland rice system accounts for approximately 55% of the worlds harvested rice area and contributes approximately 76% of global rice production [4]. In contrast to wheat, 95% of the world's rice is grown in less developed nations, mainly in Asia, Africa, and Latin America. China and India are the world's largest developing and consuming rice production countries. By 2025, it is projected that about 60% more rice will have to be produced to meet the food needs of a growing world population. Rice is Asia's principal staple meal. By 2030, the world's demand for milled rice is expected to be around 533 million tons. The rice–wheat (R–W) rotation is the most important crop rotation system in Southern and Eastern Asia, with an estimated area of 21.9 million hectares in seven countries: Bangladesh, Bhutan, China, India, Myanmar, Nepal, and Pakistan [5]. Reference [6] indicates that rice and wheat yields have reached a plateau in the Indo-Gangetic plains region of four South Asian countries, namely, India, Pakistan, Nepal, and Bangladesh due to the decline in soil fertility [7–9] under a continuous R–W cropping system [2,8,10]. It is important to analyze the effects of R–W cropping systems on long-term sustainability and to compare the conventional practice of chemical fertilizers with the integrated use of organic manures [1,2,8]. Long-term studies are required to understand various plant, soil, climatic, and agronomic interactions and their effects on crop productivity [11–13]. The R–W system is considered the backbone of food self-sufficiency; unfortunately, it is faced with the problem of sole chemical fertilizer use without organic manure application [7]. The increased productivity factor and depletion of organic matter (soil organic carbon) and nutrients from soil are the consequences of modern R–W production [3,14,15].

Nitrogen is the most limiting nutrient for crop production [5,16]; therefore, efficient use of N is important for the economic sustainability under the R–W system [17,18]. The use of adequate N rates and sources is important for improvement in N use efficiency [19], reducing production costs and environmental pollution [19,20]. The sole use of chemical N-fertilizers without organic sources leads to a gradual decline in soil fertility and lower crop productivity under the R–W system [2,12]. Due to the global energy crisis and the high cost of N chemical fertilizers (urea, ammonium sulphate, etc.), there is a renewed interest in low-cost alternatives such as the use of organic manures [1,12,17]. Complementary use of organic fertilizers is advantageous, and a substantial amount of inorganic N-fertilizer may be saved under the R–W system [7,10,21–25]. For example, References [5,7,25] reported that application of N from organic manures increased total soil N and fertility.

Organic carbon sources include essential plant nutrients (macro and micro) which improve soil properties, increase soil fertility, microbial activities, and crop productivity [1,10,11]. Organic sources are considered to be promising renewable, nutrient-rich sources and can be used as a substitute for reducing the cost of chemical fertilizers [22,26–31]. Incorporation of crops residues (CRs) and animal manures (AMs) in soils contribute to increased N immobilization in crop systems by increasing the amount of C relative to N [1,10,22,32–34]. Organic practices make the R–W cropping system more sustainable without adverse effects on natural resources and the environment [10,16,18,22,35]. The chemical N-fertilizers on the other hand are very costly and cause environmental issues. Therefore, the use of organic manures (AM versus CR) in field crop production is becoming an ever more important feature of environmentally friendly and sustainable practice [10,22,28,36]. Therefore, the use of organic materials (AM and CR) needs to be promoted under the R–W system [1,10,22]. The objective of this research project was to investigate the impact of organic carbon sources (animal manures (i.e., cattle, poultry, and sheep) and crop residues (onion, wheat, and berseem)) applied alone or in various combinations with chemical N-source (urea) on rice productivity [7,10], N concentrations in soil and hybrid rice plants in the current study, and its residual effect on the subsequent wheat crop [8] under the R–W cropping system.

2. Materials and Methods

Field experiments were conducted to investigate the impacts of organic N sources (animal manures (AM) and crop residues (CR) and inorganic N source (urea) management on the nitrogen contents

in soil, grains and straw of hybrid rice (Pukhraj) and their residual effects on the subsequent wheat (cv. Siren-2010) crop under the R–W system. Six different sources of organic sources (OS) were used: three animal manures—poultry manure (PM), sheep manure (SM) and cattle manure (CM); and three crop residues—onion residues (ORs), berseem residues (BRs), and wheat straw (WS). The C:N ratio of urea was much lower (0.4:1) as compared with CM (19:1), PM (15:1), SM (16:1), OR (15:1), WS (125:1), and BR (20:1). The experiments were carried out on farmers' field in Batkhela, Malakand Division (northwestern Pakistan) during 2011–2012 (Y1) and 2012–2013 (Y2). Batkhela is situated at 71.6167 E and 34.6167 N (in decimal degrees) 71°58'17″ E and or 34°37' N (in degrees–minutes–seconds). The soil is moderately calcareous in nature ($CaCO_3$ = 7.18%), slightly alkaline (pH = 7.3), clay loam by texture, non-saline (ECe = 1.02 dS/m), low in soil fertility containing less organic matter (0.71%), total N (0.51%), extractable P (5.24 mg kg^{-1}), exchangeable potassium (AB-DTPA) (71 mg kg^{-1}), and Zn (0.93 mg kg^{-1}).The OSs were applied 30 days before transplanting, while the urea (chemical N) was applied in two equal splits: 50% at transplanting and 50% at 30 days after transplanting. In Y1, rice nursery was grown on 4 June 2011, and then the 25 day old seedlings were transplanted on 29 June 2011. In Y2, the rice nursery was grown on 11 June 2011, and then the 25 day old seedlings were transplanted on 5 July 2011. A uniform basal dose of 60 kg P ha^{-1} (as triple super phosphate) and 60 kg K ha^{-1} (as sulphate of potash) were applied uniformly to all plots at the time of transplanting. Nitrogen sources were not applied to the control plots for both years. After the harvest of the rice crop in October, the wheat variety "Siren-2010" was grown in November in both years as a subsequent crop under the R–W system.

The experiments were carried out in a randomized complete block design with four replications. The plot size was 12 m^2 (3 m × 4 m) with 300 single plants per plot, and the plant-to-plant distance was 20 cm. All plots were separated by 30 cm ridges to stop the movement of water/nutrients among different plots (treatments), and water was applied to each plot separately from a water channel. After maturity, 2 m^2 of rice within each treatment was harvested, dried, and weighed to calculate biomass [31]. These materials were then threshed, and rice grains were separated and weighed to calculate grain yield [10].

2.1. Determination of N Content in Soil, Straw, and Grains after Harvesting of Rice Crop

Soil total N (%) was determined calorimetrically. In brief, 0.2 g of soil/straw/grains was digested in 3 mL of concentrated H_2SO_4 in the presence of 1.1 g digestion mixture (K_2SO_4, CuSO4, and Se) on block digest for 4–5 h. The temperature was gradually increased up to 3500 C and maintained for 1 hr to appear a light greenish color. After cooling, the digest was transferred to volumetric flask and the volume made up to 100 mL with distilled water. Then, 20 mL of the digest was distilled with 5 mL both of 40% NaOH solution and boric acid mixed indicator and titrated against standard 0.005 M HCl. Nitrogen was calculated as 1 mL of 0.005 M HCl is equal to 70 µg. A blank reading was also taken at the same time:

$$\text{Total N }(\%) = \frac{(\text{sample} - \text{blank}) \times 0.005 \times 0.014 \times 100 \times 100}{\text{Weight of sample} \times 20}$$

2.2. Statistical Analysis

Data on nitrogen contents in soil, grains, and straw of hybrid rice (Pukhraj) were subjected to analysis of variance (ANOVA) according to the methods described for simple randomized complete block design combined over the years [37], and the means in different treatments were compared using the least significant difference (LSD) test ($p \leq 0.05$). The Statistics v.8.1 (Analytical Software, Tallahassee, FL, USA) was used for the statistical analysis [37].

3. Results

3.1. Total Soil Nitrogen after Rice Harvesting

Organic sources had a significant effect on the total soil nitrogen (TSN) in Y1, Y2, and when combined over the two years' mean data (Table 1). The years' mean data showed PM application produced the highest TSN (776 mg kg^{-1}), followed by CM (775 mg kg^{-1}), while the minimum TSN (562 mg kg^{-1}) was obtained with use of WS. While, TSN exhibited a non-significant response to Y × OS, yet it was higher in Y2 than Y1. In Y1, the TSN ranged between 545 (WS) and 748 mg kg^{-1} (CM); while in Y2, it ranged between 580 (WS) and 821 mg kg^{-1} (PM).

Table 1. Analysis of variance for soil nitrogen concentration (mg kg^{-1}) after harvesting of rice hybrid "Pukhraj" as affected by organic and inorganic fertilizer.

Sources of Variance	Degree of Freedom	Sum of Squares	Mean Squares	*F*-Values	Significance
Years (Y)	1	239,463.00	239,463.00	23.89	**
Blocks (Years)	6	60,119.96	10,019.99	1.60	-
Treatments	25	1,808,013.00	72,320.51	11.60	***
Control vs. Rest	(1)	395,841.00	395,841.00	63.52	***
Urea versus Pure OS (Organic Sources)	(1)	42,508.59	42,508.59	6.82	**
Among all OS (Sole + Mixtures)	(23)	1,369,663.00	59,550.57	9.55	***
Pure OS vs. Mixtures	<1>	173,510.40	173,510.40	27.84	***
Pure OS	<5>	345,009.40	69,001.87	11.07	***
Animal Manures (AM) versus Crop Residues (CR)	{1}	314,604.08	314,604.10	50.49	***
Mixtures	<17>	851,143.30	50,067.25	8.03	***
Ratios	{2}	125,548.20	62,774.10	10.07	***
Organic Sources in Mixtures	{5}	700,659.20	140,131.80	22.48	***
Ratios × Organic Sources	{10}	24,935.90	2493.59	0.40	ns
Y × Treatments	25	144,057.00	5762.28	0.92	ns
Y × Control vs. rest	(1)	33,435.13	33,435.13	5.36	*
Y × Urea vs. Pure OS	(1)	136.92	136.92	0.02	ns
Y × Among all OS	(23)	110,485.00	4803.69	0.77	ns
Y × Pure OS versus Mixtures	<1>	1172.77	1172.77	0.18	ns
Y × Pure OS	<5>	13,045.67	2609.13	0.41	ns
Y × AM vs. CR	{1}	7676.02	7676.02	1.23	ns
Y × Mixtures	<17>	96,266.52	5662.73	0.90	ns
Y × Ratios	{2}	5873.43	2936.71	0.47	ns
Y × Organic Sources in Mixtures	{5}	22,614.72	4522.94	0.72	ns
Y × Ratios × Organic Sources	{10}	67,778.36	6777.83	1.08	ns
Error	150	934,639.10	6230.92		
Total	207	3,186,292			

*, **, *** indicate that data are significant at the 5%, 1% and 0.1% level of probability, respectively. ns: non-significant data at the 5% level of probability; () splits of 25; <> splits of 23; {} splits of 17 and 5 degrees of freedom (DF). CV = 10.98.

The ratios in Y1, Y2, and the two years' mean had a substantial effect, while the years' × ratios (Y × R) interaction had a non-significant effect on the TSN of rice (Table 2). The mean data of the two years revealed that N application in the ratio of 25U:75OS (25% N from urea +75% N from organic sources) produced a significantly higher TSN (776 mg kg^{-1}), while the lower TSN was observed with 75U:25OS (75% N from urea +25% N from organic sources). The Y × R indicated that TSN was significantly higher in both years while using 25U:75OS.

Table 2. Soil nitrogen concentration (mg kg^{-1}) after harvesting of rice hybrid "Pukhraj" as affected by organic and inorganic N-fertilizer.

N Source	2011	2012	Mean
Cattle Manure	748	803	775
Poultry Manure	731	821	776
Sheep Manure	664	789	726
Onion leaves	586	618	602
Wheat Straw	545	580	562
Berseem Straw	603	653	628
Level of Significance	***	***	***
75U:25OS	678	737	707
50U:50OS	720	800	760
25U:75OS	731	821	776
Level of Significance	*	**	***
Urea + Organic sources			
Urea + Cattle Manure	748	851	800
Urea + Poultry Manure	751	863	807
Urea + Sheep Manure	783	835	809
Urea + Onion Leaves	705	777	741
Urea + Wheat Straw	577	649	613
Urea + Berseem Straw	695	739	717
Level of Significance	***	***	***
Planned Mean Comparison			
Control	530	471	501 b
Rest	684	756	720 a
Urea	624	688	656 b
Mixture	710	786	748 a
Pure OS	646	711	678 b
Mixture	710	786	748 a
Urea	624	688	656 a
Pure OS	646	711	678 a
Animal Manure	749	839	794 a
Crop Residues	639	695	667 b
Urea	624	688	656 b
Pure OS + Mix	694	767	730 a
Interactions	Significance	Interactions	Significance
Y × OS	ns	Y × U vs. Mix	ns
Y × ratios	ns	Y × OS versus Mix	ns
Y × mixtures	ns	Y × AM versus CR	ns
Y × control versus rest	*	Y × U versus OS + Mix	ns
Y × urea versus OS	ns		

*, **, *** indicate that data are significant at the 5%, 1% and 0.1% level of probability, respectively. ns: non-significant data at the 5% level of probability. Means followed by different letters in the same category are significantly different at the 5% level of probability.

The mixtures had a significant effect on TSN in Y1, Y2, and the two years' mean, while their interaction (Y × M) had a non-significant effect on TSN (Table 2). The TSN ranged from 577 (U + WS) to 783 mg kg^{-1} (U + SM) in Y1, and 649 mg kg^{-1} (U + WS) to 863 mg kg^{-1} (U + PM) in Y2. The years' mean data indicated that application of N in the form of U + SM resulted in significantly higher TSN (809 mg kg^{-1}), followed by U + PM (807 mg kg^{-1}), while U + WS had the lowest TSN content (613 mg kg^{-1}).

The planned mean comparison indicated that the rest plots (average of all N treated plots) had the higher TSN than control (N not applied). The TSN increased by 9.5% in the rest plots, decreased by 12.5% in the control plots in the two years lead to a significant year x control versus rest interaction. Application of sole urea (U) reduced the TSN as compared U + OS mixtures. The TSN increased by 9.4% in the sole urea plots in Y2 over Y1, but in contrast, the TSN increased by 9.5% in Y2 over Y1 in the mix plots that resulted in non-significant year × U versus mixture interaction. Application of mixtures (U + OS) produced higher TSN than the use of pure OS. The increase in TSN was 9.1% in Y2 over Y1 while using pure OS, but application of mixture increased the TSN by 9.7% in Y2 over Y1 that resulted in non-significant Y × OS versus mixture interactions. On the other hand, TSN was significantly higher with application of pure OS than sole urea. The TSN increased by 9.4% in Y2 than Y1, when sole urea was used, while in the case of pure OS, TSN increased by 9.1% in Y2 over Y1. Interestingly, TSN was significantly higher with application of AMs as compared with CRs. The TSN increased by 10.7% in Y2 over Y1 when AM was used, while in the case of CR, TSN increased by 8.2% in Y2 over Y1. The TSN increased with the application of pure OS + mixtures than sole urea. The TSN increased by 9.4% in Y2 over Y1 with sole urea, while in the case of pure OS + mix, TSN increased by 9.6% in Y2 over Y1 (Table 2).

3.2. Total Nitrogen Concentration in Rice Grains

Organic sources had a non-significant effect on total N in rice grains (TNG), in Y1, Y2, when combined over the two years, and their interaction (Table 3). The mean years' data showed that PM produced the highest TNG (1.54%), while the lowest TNG (1.33%) was produced with WS. Although TNG showed a non-significant response to Y × OS, it was relatively higher in Y2 than Y1. In Y1, the TNG ranged between 1.30 (WS) and 1.52% (PM), but the differences were not significant. In Y2, it was statistically higher while using PM (1.55%) and lower with WS (1.36%).

Table 3. Analysis of variance for nitrogen concentration in grains (%) of rice hybrid "Pukhraj" as affected by organic and inorganic fertilizer.

Sources of Variance	Degree of Freedom	Sum of Squares	Mean Squares	*F*-Values	Significance
Years (Y)	1	0.46	0.46	16.07	**
Blocks (Years)	6	0.17	0.02	1.08	-
Treatments	25	8.16	0.32	12.39	***
Control versus Rest	(1)	6.72	6.72	255.03	***
Urea versus Pure OS (Organic Sources)	(1)	0.14	0.14	5.48	*
Among all OS (Sole + Mixtures)	(23)	1.29	0.05	2.14	**
Pure OS versus Mixtures	<1>	0.34	0.34	13.17	***
Pure OS	<5>	0.24	0.04	1.86	ns
Animal Manures (AM) versus Crop Residues (CR)	{1}	0.18	0.18	6.85	**
Mixtures	<17>	0.70	0.04	1.57	ns
Ratios	{2}	0.00	0.00	0.08	ns
Organic Sources in Mixtures	{5}	0.68	0.13	5.18	***
Ratios × Organic Sources	{10}	0.01	0.00	0.06	ns
Y × Treatments	25	0.37	0.01	0.56	ns
Y × Control versus rest	(1)	0.10	0.10	4.03	*
Y × Urea versus Pure OS	(1)	0.00	0.00	0.23	ns
Y × Among all OS	(23)	0.26	0.01	0.43	ns
Y × Pure OS versus Mixtures	<1>	0.05	0.05	2.15	ns
Y × Pure OS	<5>	0.00	0.00	0.01	ns
Y × AM versus CR	{1}	5.84	5.84	2.21	ns
Y × Mixtures	<17>	0.20	0.01	0.45	ns
Y × Ratios	{2}	0.17	0.08	3.22	*
Y × Organic Sources in Mixtures	{5}	0.02	0.00	0.16	ns
Y × Ratios × Organic Sources	{10}	0.01	0.00	0.04	ns
Error	150	3.95	0.02		
Total	207	13.13			

*, **, *** indicate that data are significant at the 5%, 1% and 0.1% level of probability, respectively. ns: non-significant data at the 5% level of probability; () splits of 25; <> splits of 23; {} splits of 17 and 5 degrees of freedom (DF). CV = 11.09.

The interaction of years × ratios (Y × R) had a significant effect on TNG, while the ratios in Y1, Y2, and the average of the two years had a non-significant effect on the TNG of rice (Table 4). The years' mean data indicated that application of N in the ratios of both 50U:50OS and 25U:75OS produced higher TNG (1.52%) than 75U:25OS. The Y × R indicated that TNG (1.50%) was higher with 75U:25OS

in Y1, while the TNG was higher while using 25U:75OS in Y2. The TNG increased 12.19%, 9.11%, and 2.16% in Y2 over Y1 while using 25U:75OS, 50U:50OS, and 75U:25OS, respectively.

The mixtures in Y1 and Y2 had a significant effect on TNG of rice (Table 4), while the average of two years and interaction of years × mixtures (Y × M) had a non-significant effect. The TNG varied significantly between 1.33% (U + WS) to 1.54% (U + PM) in Y1, and between 1.49% (U + WS) to 1.68% (U + PM) in Y2. The average of the two years' data indicated that application of N in the form of U + PM produced relatively higher TNG (1.61%), while U + WS produced the lowest TNG (1.41%). The Y x Mix indicated that TNG increased with U + AM than U + CR in both years (Table 4). The percent increase in TNG in Y2 over Y1 ranged between 6.18–9ß.20% while using U + AM, but the increase in TNG in Y2 over Y1 was 6.42–10.47%, while using U + CR. The highest increase in TNG (10.47%) in Y2 over Y1 was obtained with U + WS as compared with the lowest increase (6.18%) with U + SM.

The planned mean comparison indicated that the rest plots produced significantly higher TNG than the control plots. The TNG increased by 6.23% in the rest plots in Y2 than Y1. In contrast, in the control plots the TNG decreased by 26.44% in Y2 than Y1. Application of sole urea produced higher TNG than mixtures (U + OS). The TNG increased by 2.93% in the sole urea plots in Y2 over Y1. Application of mixtures produced higher TNG than pure OS. The TNG increased with the application of both pure OS and mixtures in Y2 than Y1. However, the increase in TNG was 3.18% in Y2 over Y1 while using pure OS, but application of mixture increased the TNG by 7.93% in Y2 over Y1. The TNG was higher with sole urea than pure OS. The TNG increased by 3.18% in Y2 than Y1 while using pure OS, while in the case of sole urea, the TNG increased by 2.93% in Y2 over Y1. The TNG was higher with AM than CR. The TNG increased by 6.77% in Y2 over Y1 while using AM, while in the case of CR, the TNG increased by 6.87% in year over Y1. The TNG increased with the application of sole urea than pure OS + mixtures. The TNG increased by 6.82% in Y2 over Y1 with pure OS + mix, while in the case of sole urea, the TNG increased by 2.93% in Y2 over year (Table 4).

Table 4. Nitrogen concentration in grains (%) of rice hybrid "Pukhraj" as affected by organic and inorganic N-fertilizer.

N Source	2011	2012	Mean
Cattle Manure	1.45	1.51	1.48
Poultry Manure	1.52	1.55	1.54
Sheep Manure	1.41	1.45	1.43
Onion leaves	1.37	1.41	1.39
Wheat Straw	1.30	1.36	1.33
Berseem Straw	1.34	1.38	1.36
Level of Significance	ns	ns	ns
75U:25OS	1.50	1.53	1.51
50U: 50OS	1.45	1.59	1.52
25U:75OS	1.43	1.62	1.52
Level of Significance	ns	ns	ns
Urea + Organic sources			
Urea + Cattle Manure	1.51	1.66	1.58
Urea + Poultry Manure	1.54	1.68	1.61
Urea + Sheep Manure	1.50	1.59	1.54
Urea + Onion Leaves	1.45	1.56	1.50
Urea + Wheat Straw	1.33	1.48	1.41
Urea + Berseem Straw	1.42	1.52	1.47

Table 4. *Cont.*

Level of Significance	*	*	ns
	Planned Mean Comparison		
Control	0.63	0.51	0.56 b
Rest	1.47	1.56	1.51 a
Urea	1.61	1.66	1.63 a
Mixture	1.46	1.58	1.52 b
Pure OS	1.40	1.44	1.42 b
Mixture	1.46	1.58	1.52 a
Urea	1.61	1.66	1.63 a
Pure OS	1.40	1.44	1.42 a
Animal Manure	1.50	1.61	1.55 a
Crop Residues	1.38	1.49	1.43 b
Urea	1.61	1.66	1.63 a
Pure OS + Mix	1.44	1.55	1.49 a

Interactions	Significance	Interactions	Significance
Y × OS	ns	Y x U versus Mix	ns
Y × ratios	*	Y x OS versus Mix	ns
Y × mixtures	ns	Y x AM versus CR	ns
Y × control versus rest	***	Y x U versus OS + Mix	ns
Y × urea versus OS	ns		

*, *** indicate that data are significant at 5% and 0.1% level of probability, respectively. ns: non-significant data at the 5% level of probability. Means followed by different letters in the same category are significantly different at the 5% level of probability.

3.3. Total Nitrogen Concentration in Rice Straw

Organic sources had a non-significant effect on total nitrogen in straw (TNS) in Y1, Y2, when combined over the two years, and the interaction (Y × OS) on rice (Table 5). The two years' mean data revealed that use of PM produced higher TNS (0.55%), while the lower TNS (0.48%) was obtained with use of WS, while Y × OS had no substantial effect on TNS, yet it was relatively higher in Y2 than Y1. In Y1, the TNS ranged between 0.47 (WS) and 0.52% (CM and PM), but the differences were non-significant; while in Y2, it was statistically higher with SM (0.59%) and lower with WS (0.49%).

The ratios in Y1, Y2, and their interactive effect (Y × R) had a substantial effect on TNS, while the mean of the two years had no substantial effect on the TNS of rice (Table 6). The two years' mean data showed that N application in both ratios of 75U:25OS and 50U: 50OS produced the higher TNS (0.56%) than 25U:75OS. The Y × R showed that in Y1, application of 75U:25OS ratio, produced higher TNS, but in Y2, the higher TNS was produced by the ratio of 25U:75OS. The NG increased by 21.16%, 11.29%, and 1.48% in Y2 over Y1 while using 25U:75OS, 50U:50OS, and 75U:25OS, respectively.

Table 5. Analysis of variance for nitrogen concentration in straw (%) of rice hybrid "Pukhraj" as affected by organic and inorganic fertilizer.

Sources of Variance	Degree of Freedom	Sum of Squares	Mean Squares	*F*-Values	Significance
Years (Y)	1	0.16	0.16	14.06	**
Blocks (Years)	6	0.07	0.012	1.71	-
Treatments	25	0.64	0.02	3.71	***
Control versus Rest	(1)	0.29	0.29	42.43	***
Urea versus Pure OS (Organic Sources)	(1)	0.02	0.02	3.27	ns
Among all OS (Sole + Mixtures)	(23)	0.32	0.01	2.04	**
Pure OS versus Mixtures	<1>	0.05	0.05	8.24	**
Pure OS	<5>	0.02	0.00	0.74	ns
Animal Manures (AM) versus Crop Residues (CR)	{1}	0.02	0.01	2.63	ns
Mixtures	<17>	0.24	0.01	2.06	*
Ratios	{2}	0.00	0.00	0.14	ns
Organic Sources in Mixtures	{5}	0.18	0.03	5.34	***
Ratios × Organic Sources	{10}	0.05	0.00	0.81	ns
Y × Treatments	25	0.28	0.01	1.62	*
Y × Control versus rest	(1)	0.03	0.03	4.99	*
Y × Urea versus Pure OS	(1)	0.00	0.00	0.53	ns
Y × Among all OS	(23)	0.24	0.01	1.52	ns
Y × Pure OS versus Mixtures	<1>	0.00	0.00	0.75	ns
Y × Pure OS	<5>	0.00	0.00	0.17	ns
Y × AM versus CR	{1}	0.00	0.00	0.52	ns
Y × Mixtures	<17>	0.23	0.01	1.96	*
Y × Ratios	{2}	0.09	0.04	6.80	**
Y × Organic Sources in Mixtures	{5}	0.00	0.00	0.16	ns
Y × Ratios × Organic Sources	{10}	0.13	0.01	1.90	*
Error	150	1.04	0.00		
Total	207	2.22			

*, **, *** indicate that data are significant at the 5%, 1% and 0.1% level of probability, respectively. ns: non-significant data at the 5% level of probability; () splits of 25; <> splits of 23; {} splits of 17 and 5 degrees of freedom (DF). CV = 15.26.

The mixtures in Y1 had a significant effect, while in Y2, the two years' mean and interaction of years x mixtures (Y × M) had no substantial effect on TNS of rice (Table 6). The TNS varied between 0.49% (U + WS and U + OL) and 0.59% (U + PM) in Y1 and between 0.54% (U + WS) and 0.67% (U + PM) in Y2. The two years' average data showed that the use of U + PM as source of N produced relatively higher TNS (0.63%), while U + WS produced the lowest TNS (0.51%). The Y × mixtures indicated that TNS increased with U + AM than U + CR in both years (Table 6). The percent increase in TNS in Y2 over Y1 ranged between 8.72% and 11.61% while using U + AM, but the increase in TNS in Y2 over Y1 was 10.87–14.55% while using U + CR. The highest increase in TNS (14.55%) in Y2 over Y1 was obtained with U + BS as compared with lowest increase (8.72%) with U + WS.

Rest produced higher TNS than the control as indicated by the planned mean comparison. The TNS increased by 9.78% in the rest plots in Y2 over Y1, in difference in the control plots the TNS decreased by 22.48% in Y2 than Y1. Application of sole urea was higher in TNS than mixtures. The TNS increased by 3.24% in the sole urea plots in Y2 over Y1, but in contrast, the TNS increased by 11.70% in Y2 over Y1 in the mix plots that lead to a significant interaction of year × U versus Mix. Application of mixtures had higher TNS than pure OS. The TNS increased with application of both pure OS and mixtures in Y2 than Y1. However, the increase in TNS was 8.40% in Y2 over Y1 while using pure OS, but application of the mixture increased the TNS by 11.70% in Y2 over Y1 that resulted in non-significant Y × OS versus mixture interactions. The TNS was higher with sole urea than pure OS. The TNS increased by 8.40% in Y2 than Y1 while using pure OS, while in the case of sole urea, the TNS increased by 3.24% in Y2 over Y1 resulting in significant Y × U versus OS interaction. The TNS was higher with AM than CR. The TNS increased by 10.46% in Y2 over Y1 while using AM, while in the case of CR, the TNS increased by 11.46% in Y2 over Y1.

Table 6. Nitrogen concentration in straw (%) of rice hybrid "Pukhraj" as affected by organic and inorganic N-fertilizer.

N Source	2011	2012	Mean
75U:25OS	0.55	0.56	0.56
50U:50OS	0.53	0.60	0.56
25U:75OS	0.50	0.63	0.57
Level of Significance	*	*	ns
"Cattle Manure	0.52	0.57	0.54
Poultry Manure	0.52	0.57	0.55
Sheep Manure	0.50	0.59	0.54
Onion leaves	0.51	0.54	0.52
Wheat Straw	0.47	0.49	0.48
Berseem Straw	0.49	0.53	0.51
Level of Significance	ns	ns	ns
Urea + Organic sources			
Urea + Cattle Manure	0.54	0.59	0.57
Urea + Poultry Manure	0.59	0.67	0.63
Urea + Sheep Manure	0.55	0.61	0.58
Urea + Onion Leaves	0.49	0.58	0.54
Urea + Wheat Straw	0.49	0.54	0.51
Urea + Berseem Straw"	0.51	0.60	0.55
Level of Significance	**	ns	*
Planned Mean Comparison			
Control	0.40	0.32	0.36 b
Rest	0.53	0.59	0.56 a
Urea	0.60	0.62	0.61 a
Mixture	0.53	0.60	0.56b
Pure OS	0.50	0.55	0.52 b
Mixture	0.53	0.60	0.56 a
Urea	0.60	0.62	0.61 a
Pure OS	0.50	0.55	0.52 b
Animal Manure	0.55	0.61	0.58 a
Crop Residues	0.49	0.56	0.53 a
Urea	0.60	0.62	0.61 a
Pure OS + Mix	0.52	0.58	0.55 b

Interactions	Significance	Interactions	Significance
Y × OS	ns	Y × U versus Mix	*
Y × ratios	**	Y × OS versus Mix	ns
Y × mixtures	ns	Y × AM versus CR	ns
Y × control versus rest	*	Y × U versus OS + Mix	ns
Y × urea versus OS	*		

*, ** indicate that data are significant at 5% and 1% level of probability, respectively. ns: non-significant data at the 5% level of probability. Means followed by different letters in the same category are significantly different at the 5% level of probability.

4. Discussion

4.1. Nitrogen Concentration in Soil after Rice Harvest

The residual effect of organic and inorganic N sources on available N status revealed that a negative N content in soil was observed where N was applied in the form of mineral fertilizer (urea) or in control plot (N not applied). In the case of organic sources, higher N was noted in AM treated plots, while minimum N concentration was recorded in CR treated plots. Lower N concentration was found

with a ratio of 75U:25OS, while a maximum N concentration was obtained with 25U:75OS. Application of U + AM gave more soil N than U + CR. The soil N status was greater where N was supplied through organic and inorganic sources of fertilizers [21]. According to References [21–23], application of farm yard manure (FYM) increased soil organic carbon, available nitrogen, phosphorus and potassium (NPK) when compared with control treatment. Reference [38] suggested that the combined application of organic manures and NPK increases maize yield and soil N. References [5,25] reported that application of N from fertilizer, FYM, and poultry litter increased soil total N, and References [23,28] reported that total N increased in treatment where poultry manure was applied to the previous wheat crop. Similarly, References [8,28] reported that over a period of 20 years with FYM treatment, soil organic matter increased by 80% compared to only 10% with NPK. However, from an environmental and agronomic point of view, manure application was not a better strategy compared to NPK fertilization. Reference [8] also came to the conclusion that the residual effect of organic fertilizers on soil fertility. Wheat yield is in the following order: bird droppings + FYM followed by PM, FYM, and no organic sources. Rice yields and nitrogen availability in low-fertility soils have increased over three years of continuous organic fertilization [28]. However, this may depend on the type and amount of organic matter used. The residual effect of organic and inorganic materials on available N status revealed that a negative N content in soil was observed where N was applied in the form of mineral or in control plot. In the case of organic sources maximum N was noted in AM treated plots having low CN ratio (15:1 to 19:1), while minimum N concentration was recorded in CR treated plots having higher CN ratio (15:1 to 125:1). A lower N concentration was found with a ratio 75U:25OS, while a maximum N concentration was obtained from 25U:75OS in both years. Application of U + AM gave more soil N than U + CR. The soil N status was greater where N was supplied through organic and inorganic sources of fertilizers [21]. The study suggested the complementary use of organic and inorganic fertilizer is advantageous, and that a substantial amount of inorganic fertilizer may be saved for rice crop by the use of organic manures. Application of FYM increased soil organic carbon and available NPK when compared with control treatment [21,23]. Soil fertility-building in organic systems improves soil biological properties which subsequently influences N availability to crops [7,16,39,40]. High N-uptake under treatment where mineral N and organic N ratios were 75U:25OS and 50U:50OS N, respectively [24]. Maximum total N after each crop harvest was found in the treatment where organic sources were applied with low mineral N. This may be due to the greater quantity of organic sources of N applied. Reference [38] suggested that the combined application of organic manures and NPK increases maize yield and soil N.

4.2. Nitrogen Concentration in Rice Grains and Straw after Rice Harvest

Understanding crop nutrient removal can provide important information for soil fertility management by comparing total crop uptake with inputs from all sources [28]. The present results of our study demonstrated that N concentrations in rice grains and straw was high when mineral N was applied in the form of urea or in combination with animal manure (U + PM), while lower N concentration was noted where N was not applied or where N was applied in the form of organic N. Mineral fertilizers provided better plant growth and nutrient intake when grown in one season compared to organic fertilizers [28]. The residual effect of organic and inorganic materials on available N status revealed that a negative N content in soil was observed where N was applied in the form of mineral or in control plot. In the case of organic sources, maximum N was noted in AM treated plots, while minimum N concentration was recorded in CR treated plots. The residual effect of FYM with inorganic fertilizers played an important role in the grain yield of the next wheat crop by improving the absorption of NPK [41]. Improving soil fertility in organic systems improves the biological properties of the soil which subsequently affects the availability of nitrogen for crops [7,42]. Reference [24] reported high N-uptake with mineral N and organic N ratios than the control. Previous research has shown that compost and manure increase nutrient efficiency by slowly releasing nutrients and reducing nutrient losses [15,43]. The addition of organic fertilizers increases nutrient mobilization and soil microbial activity; improve nutrient mobilization and soil microbial activity; it can also help improve

nutrition as well as the root system of crops. The present results are consistent with the assumptions in References [44,45] which suggested that increasing soil nitrogen levels may have a positive effect on nitrogen uptake in maize. The effect of organic and inorganic N status revealed that a negative N content in soil was observed where N was applied in the form of mineral N. with organic sources maximum N with AM treated plots, while minimum N concentration was recorded in CR treated plots. Reference [21] reported that lower N concentration was found with a ratio of 75U:25OS, while maximum N concentration was obtained with 25U:75OS. Application of U + AM resulted in significantly higher soil N than U + CR. The soil N status was greater where N was supplied through organic and inorganic sources of fertilizers. The study suggested the complementary use of organic and inorganic fertilizer is advantageous and substantial amount of inorganic fertilizer may be saved for rice crop by the use of organic manures. According to References [21,23], FYM application increased the available NPK soil organic carbon content compared to control treatments. Improving soil fertility in organic systems improves the biological properties of the soil, which subsequently affects the availability of nitrogen for crops [7,40]. Likewise, Reference [24] reported high N-uptake under treatment where the mineral N and organic N ratio was higher 75U:25OS as compared with 50U:50OS N. The maximum total N after each crop harvest was found in the treatment where OA were applied with low mineral N. This may be due to the greater quantity of organic sources of N applied. According to Reference [38], combined application of organic manures and NPK increases maize yield and soil N. Likewise, References [5,25] reported that application of N from fertilizer, FYM, and poultry litter increased soil total N.

5. Conclusions

The N concentration in rice grains and straw increased with the application of mineral N or with combination of mineral N + AMs. The N concentration in grains and straw was reduced significantly when N was not applied (N control plots). The N concentration in grains and straw increased significantly with the application of 120 kg N ha^{-1} in the form of sole urea, and urea + PM. Plants grown in Y2 had more N concentration in their grains and straw than in Y1. The soil N content after rice harvest was higher in plots where N was applied in the form of sole (pure) organic sources (pure OS > mixtures > pure urea > control).

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