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Original Research

Effect on Efficient Utilization and Availability of Nitrogen in Paddy Field under Rural Domestic Reclaimed Water Irrigation

Menghua Xiao[1*], Shizong Zheng[1], Yuanyuan Li[1, 2]**

[1]Zhejiang Institute of Hydraulics and Estuary (Zhejiang Institute of Marine Planning and Design), Hangzhou, 310020, P.R. China
[2]North China University of Water Resource and Electric Power, Zhengzhou, 450045, P.R. China

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Abstract

Rural domestic reclaimed water (RDRW) brings in a large amount of nitrogen that could affect the nitrogen supply capacity of soil and the absorption and utilization of crops. Four kinds of irrigation water sources (primary and secondary treated water R1 and R2, purified water R3, and river water CK) and three kinds of fertilization gradients (10%, 30%, and 100% conventional nitrogen fertilizer reduction of N1, N2, and N0) were set up to study the effects on the efficient utilization and availability of nitrogen in paddy rice. ^{15}N tracer technology combined with fertilizer equivalent methods was used. The results showed that the nitrogen absorbed and utilized for soil and crop systems mainly came from fertilizer nitrogen (NF), soil nitrogen (NS), and reclaimed water nitrogen (NRW). NS was the main source of nitrogen uptake by plants. NRW use efficiency (RWNUE) was not directly proportional to the nitrogen concentration in RDRW, while NRW residue rate (RWNRE) was inversely proportional to it. Compared with CK, the absorption and utilization of nitrogen were inhibited, and the contribution rates of NF and NRW were both decreased under RDRW irrigation. Under N1 and N2, the NF relative substitution equivalent (RFE) of R1, R2, and R3 was 28.1% and 56.3%, 13.6% and 46.6%, 1.3% and 5.4%, respectively. Since reducing the fertilization gradient can effectively improve NRW availability, 30% and 10% nitrogen reduction fertilization were recommended for R1 and R2 irrigation, which can fully utilize the effectiveness of nitrogen in reclaimed water in paddy fields.

Keywords: rural domestic sewage, fertilization gradient, efficiency and availability, 15N tracer technology, fertilizer equivalent method

*e-mail: liya66720@126.com;
**e-mail: menghuaxiao@aliyun.com
Tel.: +86 15903662916

Introduction

Rice is one of the most important food crops in the world, however, it has a strong risk of non-point source pollution emissions with high water and nitrogen consumption during the growth period [1]. The contradiction of water shortages in China is becoming increasingly prominent. Even in southern China, water shortages caused by worse water quality are still prominent [2]. With the large water consumption of industry and urban areas, the gap in agricultural irrigation water reaches 60 billion m^3 every year, which is still increasing year by year. With the acceleration of new rural construction, the discharge of rural domestic sewage is increasing. Compared with urban sewage, rural domestic sewage can be used as fertilizer resources with the characteristics of a small water quality difference, scattered water volume, high content of N and P, good biodegradability, and generally without toxic substances such as heavy metals [3, 4]. However, it is discharged in an extensive manner, with scattered distribution and wide coverage [5]. Compared with conventional water sources, reasonable irrigation of rural sewage can realize low-cost treatment of sewage, reduce the amount of fresh water and the input of fertilizer, and improve soil fertility [6]. Therefore, the safe treatment of rural domestic sewage for agricultural irrigation is an effective way to solve the dilemma of the water ecological environment and alleviate the contradiction between supply and demand of water resources [7].

Rural domestic sewage discharge standards in China carry out the first-class B standard of the discharge standard of pollutants for municipal sewage treatment plants [8]. An annual output of domestic sewage is as high as 8.395 billion tons, with about 2.7 million natural villages in China, which will bring in about 16800 tons of nitrogen to rivers, lakes, and reservoirs. Irrigation and reuse of RDRW could realize the utilization of sewage resources; meanwhile, it would lead to a large amount of nitrogen surplus in the soil environment, which would then affect the supply capacity of soil nitrogen and finally affect nitrogen absorption and utilization in the soil and crop systems. It was found that it can save 75% of the clean water amount with reclaimed water irrigation, and the substitution efficiency of nitrogen fertilizer was about 35.8% [9].

Scholars at home and abroad found that excessive fertilization in paddy fields increased the risk of nitrogen leaching, resulting in nitrogen use efficiency decreasing and endangering the ecological environment [10-12]. However, the absorption and utilization mechanisms of nitrogen, respectively, from RDRW, fertilizer, and soil by crops are very different due to the complex components of RDRW. In recent years, scholars at home and abroad have used ^{15}N tracer technology to quantify nitrogen use efficiency [13, 14], which can distinguish the nitrogen sources of fertilizer or soil [15, 16] and compare the effectiveness of different nitrogen application rates and nitrogen sources. Moreover, the fertilizer equivalent method is usually used to study the effectiveness of organic fertilizer nitrogen on crop growth. In order to compare the chemical fertilizer substitution equivalent of organic nitrogen under different organic fertilizers and different application rates, it is necessary to calculate the relative substitution equivalent of organic fertilizer, which is the chemical fertilizer substitution equivalent of organic fertilizer divided by the amount of organic fertilizer nitrogen applied [17].

Although the recycling of RDRW can replace a certain amount of fertilizer, there is still a lack of research on the distribution of nitrogen in soil and crop systems, the efficient utilization of nitrogen, and its availability for crop growth. The traditional difference method for calculating nitrogen use efficiency and its availability cannot avoid the impact of soil nitrogen under RDRW irrigation. Therefore, in this study, different from existing research, ^{15}N isotope tracing technology combining with fertilizer equivalent methods was used to put forward the objectives as follows; (1) to study the migration and distribution characteristics of nitrogen from different sources in soil and crop system, (2) to obtain the dynamics of crop absorption, soil residue, and loss of nitrogen, respectively, coming from soil, fertilizer, and RDRW, and (3) to explore the mechanism of nitrogen efficient utilization under RDRW irrigation. The innovation of this study is to study the coupling utilization characteristics of crops between NF and NRW and explore the efficient nitrogen utilization mechanism under RDRW irrigation. The results will provide a theoretical reference for exploring how to achieve a high yield of rice, high nitrogen utilization, and a reduction in nitrogen fertilizer application under RDRW irrigation.

Materials and Methods

Experimental Site

This study was carried out in Jinhua rural domestic reclaimed water test base (120°10′E, 28°48′N) of Zhejiang Province from June to October from 2020 to 2022. The annual average rainfall is 1787 mm, the annual average evaporation is 930.2 mm, the annual average temperature is 17.5º, and the frost free period is 245 days. A domestic sewage disposal with a design scale of 400 m^3/d has been built, which was studied as the source of rural domestic reclaimed water (RDRW) in this study. The treatment process adopted the secondary biological treatment process (primary treatment was a conventional process, and secondary treatment adopted the A^2O process improved), and the effluent quality met the first-class B standard of the discharge standard of pollutants for the municipal sewage treatment plant. An ecological pond with a sewage storage capacity of 3000 m^3 was built to store and purify the secondary treated water of RDRW.

There were 36 cylindrical measuring barrels set in the greenhouse with a diameter of 400 mm and a depth of 1000 mm. The soil in the measuring barrels retains the original soil structure, and the physical and chemical properties of different soil layers are shown in Table 1.

Experimental Design

The experimental research was carried out on RDRW irrigation of paddy rice. The rice variety was Jiayou Zhongke 13-1, which was planted by sowing, and 10 plants were reserved in each measuring barrel after emergence. Four kinds of irrigation water sources were used: namely, primary treated water of RDRW (R1), secondary treated water of RDRW (R2), ecological pond water (R3), and river water (CK). Each irrigation water source was connected to the measuring barrel through the field irrigation pipe network. The water quality indexes of different water sources are shown in Table 2. Controlled irrigation and drainage modes were adopted, which was different from the previous water-saving irrigation. The core of irrigation and drainage regulation was to increase the consumption of RDRW and save access to fresh water. The water level regulation of controlled irrigation and drainage in paddy fields is shown in Table 3. Three nitrogen fertilizer gradients were set, namely 10% nitrogen fertilizer reduction N1 (90% conventional fertilization), 30% nitrogen fertilizer reduction N2 (70% conventional fertilization), and no nitrogen application N0. The conventional nitrogen application rate was 225 kg/ha, according to local custom. By calculation, the three nitrogen application

Table 1. PH, electrical conductivity (EC), soluble salt, total nitrogen (TN), total phosphorus (TP), Organic matter, ammonium-nitrogen (NH^{4+}-N) and nitrate-nitrogen (NO_3^--N) in measuring barrels.

Soil depth	pH	EC (mS/m)	Soluble salt (g/kg)	TN (%)	TP (%)	Organic matter (g/kg)	NH_4^+-N (mg/kg)	NO_3^--N (mg/kg)
0-20 cm	5.56	2.6	0.44	0.12	0.069	17.7	8.24	2.84
20-40 cm	5.88	2.9	0.27	0.09	0.032	14.8	5.75	2.69
40-60 cm	6.15	2.8	0.26	0.07	0.027	12.9	4.71	2.5

Table 2. Description and statistics of chemical oxygen demand (COD), linear alkylbenzene sulfonates (LAS), ammonium-nitrogen (NH_4^+-N) and nitrate-nitrogen (NO_3^--N) in different irrigation water sources (mg/L).

Water sources	Indicator	Maximum value	Minimum value	Standard deviation	Mean value	Kurtosis	Skewness
R1	COD	84	15	26.794	29.5	5.855	2.410
	LAS	0.88	0.06	0.315	0.25	5.199	2.247
	NH_4^+-N	11.9	8.25	1.645	9.647	-1.782	0.916
	NO_3^+-N	0.061	0.016	0.019	0.034	-1.452	0.642
R2	COD	59	10	16.783	24.1	0.719	1.291
	LAS	0.16	0	0.058	0.048	-0.425	0.827
	NH_4^+-N	11.9	3.52	2.837	7.712	-0.946	-0.174
	NO_3^+-N	6.25	0.01	2.455	1.364	1.238	1.687
R3	COD	62	11	12.735	24.15	0.710	1.553
	LAS	0.32	0	0.132	0.042	-1.215	0.826
	NH_4^+-N	5.45	2.34	0.634	4.415	0.478	0.473
	NO_3^+-N	3.16	0.345	0.928	0.823	1.382	1.275
CK	COD	56	7	15.712	23.45	0.710	1.251
	LAS	0.1	0	0.041	0.035	-1.875	0.418
	NH_4^+-N	1.49	0.116	0.394	0.711	0.143	0.393
	NO_3^+-N	2.56	0.624	0.578	1.048	4.680	2.078

Note: Standard deviation is the arithmetic square root of variance, reflecting the dispersion of a data set, mean value is the average value of a data set, kurtosis is a statistic that describes the gradient of all value distribution patterns, and skewness is a statistic that describes the distribution form of data, reflecting the symmetry of a data set.

Table 3. Standard of water level regulation of irrigation and drainage in paddy field (mm).

Upper and lower limit	Turning green stage	Early tillering stage	Later tillering stage	Jointing-booting stage	Heading-flowering stage	Milky stage
Upper limit of sewage	0	10	10	10	10	10
Lower limit of sewage	30	50	Exposing field	50	50	50
Upper limit of storage	50	70		100	100	100

Note: The values of the upper and lower limit were water depth maintained by farmland

gradients were 5.5 g/barrel, 4.28 g/barrel, and 0 g/barrel, respectively. Fertilizer was applied three times, namely base fertilizer, tillering fertilizer, and panicle fertilizer, accounting for 50%, 30%, and 20%, respectively. The nitrogen fertilizer was urea marked by ^{15}N (the nitrogen mass fraction was 46%, and the abundance was 10%). Each irrigation water source and nitrogen application gradient were designed for 3 repetitions; therefore, 36 test treatments were set in total. The layout of the barrel test is shown in Fig. 1.

Indicators and Methods

The soil and plants were sampled at the main growth stage of rice. The soil samples of 0-20 cm, 20-40 cm, and 40-60 cm soil layers were taken from each treatment by the 5-point method. At the same time, typical plant samples were selected. The samples were brought back to the laboratory for air drying, put into the drying oven (85 °C), dried to a constant weight, and then crushed by a pulverizer. The plant and soil samples were sieved at 60 mesh and 120 mesh, respectively, to determine the TN content and ^{15}N abundance. TN content was determined by the semi micro Kjeldahl method, and ^{15}N abundance was determined by nitrogen isotope mass spectrometry.

Irrigation water consumption was measured throughout the whole growth period. When there was a water layer in the field, the depth of the water layer before and after irrigation was recorded by a measuring needle, and the difference between the two was the irrigation amount. When there was no water layer in the field, the irrigation amount was directly recorded by the water meter. TN concentration in irrigation water was measured by alkaline potassium persiflage digestion-ultraviolet spectrophotometry.

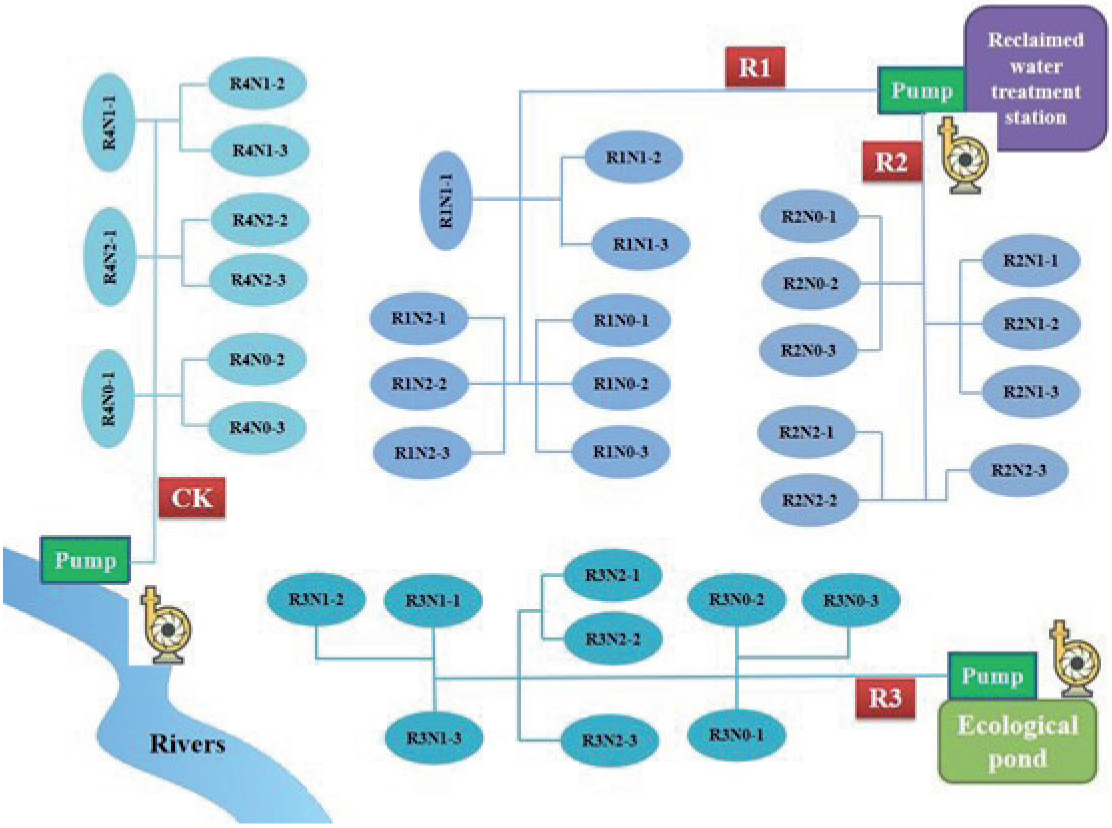

Fig. 1. Layout of barrel test.

Data Analysis

For RDRW irrigation, the nitrogen absorbed and utilized for soil and crop growth mainly comes from three parts, namely fertilizer nitrogen (NF), soil nitrogen (NS), and nitrogen brought in by RDRW (NRW). The nitrogen calculation for each part was as follows:

The proportion of nitrogen absorbed by crops from ^{15}N labeled urea (%PNF, %) or the proportion of soil nitrogen from ^{15}N labeled urea (%SNF, %) was calculated as follows:

$$\%PNF(\%SNF) = \frac{atom\%15N_{assay} - atom\%15N_{CK}}{atom\%15N_{fertilizer} - atom\%15N_{CK}} \times 100$$

Where $atom\%15N_{assy}$ refers to the ^{15}N abundance of plant or soil samples in the treatment with fertilization (%), $atom\%15N_{CK}$ refers to ^{15}N abundance of plant or soil samples in the treatment without fertilization (%), and $atom\%15N_{fertolizer}$ refers to ^{15}N abundance of labeled urea for the test (%).

The amount of nitrogen absorbed by each part of the crop comes from NF (PNF, g/plant) or the proportion of nitrogen in soil comes from NF (SNF, g/barrel) was calculated as follows:

$$PNF(SNF) = \frac{N_{assay} \times \%PNF(\%SNF)}{100}$$

Where N_{assay} refers to crop nitrogen uptake (g/plant) or soil total nitrogen (g/barrel). The nitrogen uptake of crops is the biomass multiplied by the nitrogen content of crops measured in the experiment, and the TN in soil is the soil quality multiplied by the nitrogen content of soil measured in the experiment.

When using clear water for irrigation, the nitrogen absorbed by crops (PNF and PNS) mainly comes from NF and NS, so PNS can be considered equal to the amount of crop nitrogen absorption minus PNF. The initial soil mineral nitrogen content of the barrel test is very low, and short-term RDRW irrigation will not have a significant impact on soil nitrogen mineralization. Therefore, it can be considered that PNS contents after irrigation with different water qualities are the same. Based on the above, for RDRW irrigation, the absorption and utilization of NRW by crops (PNRW) can be considered to be equal to the amount of nitrogen absorbed by crops minus PNF and PNS.

NF use efficiency (FNUE, %) represents the proportion of PNF in NF application, and NF residue rate in soil (FNRE, %) represents the proportion of SNF in NF application. Therefore, the NF loss rate (FNLE, %), is 100 minus FNUE and FNRE. The NRW use efficiency (RWNUE, %) is the ratio of the PNRW content to the nitrogen content brought in RDRW for irrigation. The calculation method of NRW residue rate (RWNRE, %) and loss rate (RWNLE, %) is consistent with that of NF.

The fertilizer equivalent method was used to study NRW availability on rice growth. Firstly, the regression relationship between the nitrogen application amount and the PNF was established, and then PNRW was substituted into the above regression equation to obtain the fertilizer nitrogen substitution equivalent (FE) of NRW. Divide the FE of NRW to obtain the fertilizer nitrogen relative substitution equivalent (RFE) of NRW to illustrate NRW availability.

Additionally, the data calculation and diagramming were completed by origin. ANOVA analysis was carried out by SPSS Statistics 19.

Results

SNF Distribution

SNF distributions under different irrigation water sources and fertilization gradients in rice fields are shown in Fig. 2. Under the same water source irrigation for N1 and N2 gradients, the SNF content of the 0-20 cm soil layer was higher than that of the 40-60 cm soil layer at the end of the growth period. Under R1 irrigation, for the N1 gradient, the SNF contents of the 0-20 cm and 20-40 cm soil layers were lower than those for N2, with a decrease of 46.6% and 33.3%, respectively, and the SNF content of the 40-60 cm soil layer was higher than those for N2, with an increase of 9.4%. Under R2 irrigation, the SNF content of each soil layer decreased with an increase in soil depth and gradually increased with the advance of the growth period. For the N1 gradient, compared with the N2 gradient, the SNF content of the 0-20 cm soil layer was increased by 3.7%, and the SNF contents of the 20-40 cm and 40-60 cm soil layers were decreased by 6.1% and 24.1%, respectively. It indicated that high fertilization had an obvious effect on the accumulation of SNF in the surface soil (0-20 cm) under R2 irrigation. Under R3 irrigation, for the N1 gradient, the SNF contents of 0-20 cm and 40-60 cm soil layers at the end of the growth period were higher than that at the beginning of the growth period, but the SNF content of 20-40 cm soil layer showed the opposite trend. For the N2 gradient, the change in SNF content was opposite to that of N1. Under CK irrigation, the SNF content of each soil layer was basically higher at the beginning than those at the end of the growth period, with an increase of 58.7%, 8.5%, and 64.8% at the N1 gradient, respectively, in 0-20 cm, 20-40 cm, and 40-60 cm soil layers, compared with that at the N2 gradient.

Under the same fertilization gradient, the accumulation of SNF content in soil at the end of the growth period was shown that, for the N1 gradient, the SNF contents in 0-20 cm and 20-40 cm soil layers were the largest under R2 irrigation, which were 68.3%, 55.4%, 81.2%, and 26.1%, 50%, and 47.8% lower than those of R1, R3, and CK, respectively, and the SNF content in the 40-60 cm soil layer was larger under R1 and R3 irrigation (R1≈R3), with a decrease of 37.1%

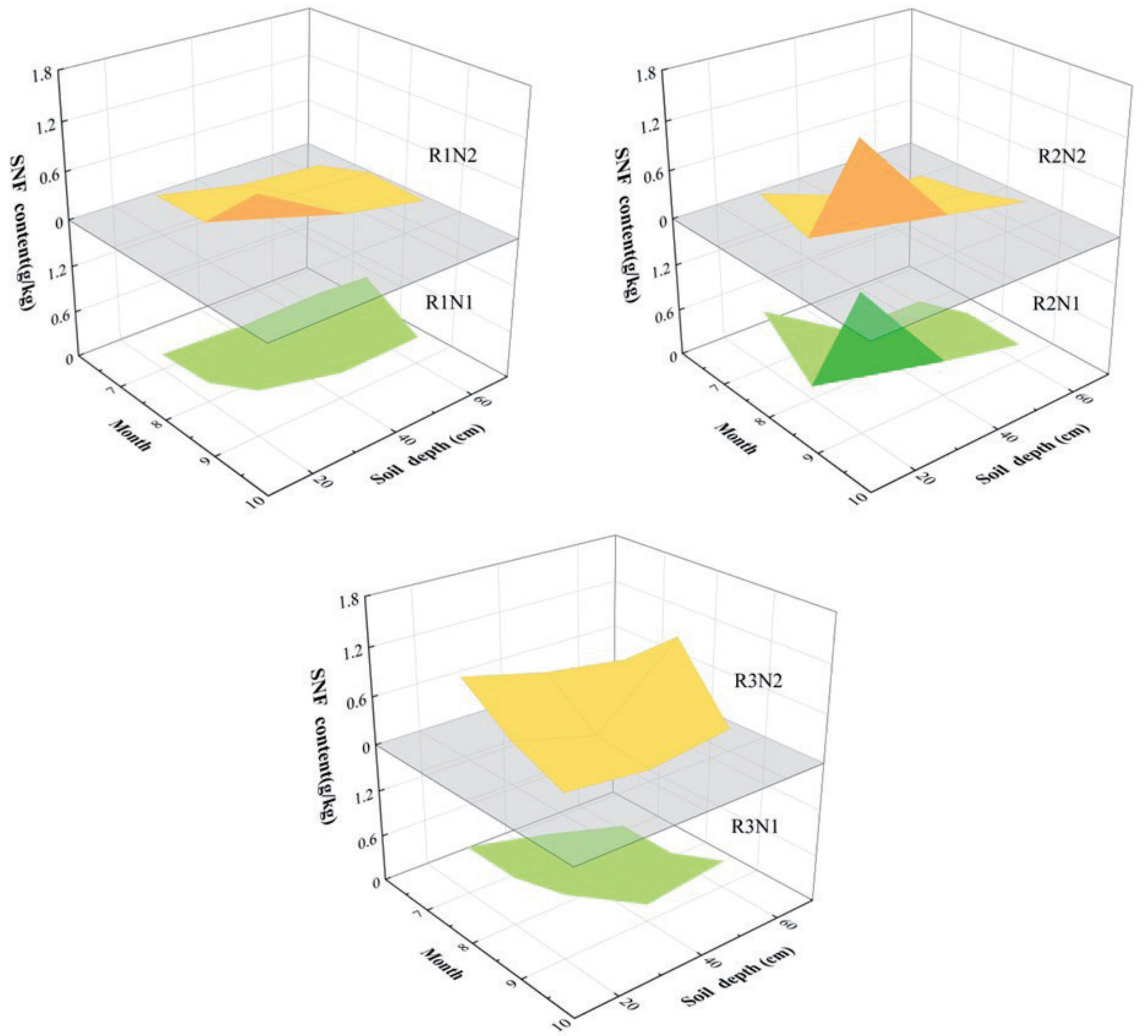

Fig. 2. Nitrogen distributions in soil from fertilizer nitrogen (SNF) under different irrigation water sources and fertilization gradients.

and 28.6% under R2 and CK, respectively. Under the N2 gradient, for 0-20 cm and 20-40 cm soil layers, the SNF contents under R1 and R2 irrigation were significantly higher than those of R3 and CK, and they were the largest in the 0-20 cm soil layer under R2 irrigation, with those of R1, R3, and CK lower by 38.9%, 85.6%, and 87.8%, respectively. For the 20-40 cm soil layer, the SNF content was the largest under R1 irrigation, with those of R2, R3, and CK lower by 3.9%, 70.6%, and 56.9%, respectively. For the 40-60 cm soil layer, the SNF content was the lowest under CK irrigation, and those of R1, R2, and R3 were basically the same.

SNRW Distribution

SNRW distributions under different irrigation water sources and fertilization gradients in rice fields are shown in Fig. 3. Under R1 irrigation, for a 0-20 cm soil layer, the SNRW content was gradually increased with an increase in fertilization amount. The SNRW content at the N1 gradient was 1.8 times and 38.1 times that of N2 and N0, respectively. For the 20-40 cm soil layer, the accumulation of SNRW content was the largest at the N1 gradient, with a decrease of 82.2% and 77.2%, respectively, at the gradients of N2 and N0. For the 40-60 cm soil layer, the change in SNRW content was opposite to that of the 0-20 cm soil layer, which may be due to the acceleration of rice plant absorption, utilization, and transformation of NRW under high fertilization conditions. For the N1 gradient, the SNRW contents of 0-20 cm and 20-40 cm soil layers gradually increased with the advance of the growth period, while they showed the opposite trend at the N2 and N0 gradients. The SNRW content of the 40-60 cm soil layer under each fertilization gradient gradually decreased with the advance of the growth period, indicating that nitrogen was surplus and accumulated in the soil at the N1 gradient under R1 irrigation.

Under R2 irrigation, the SNRW content of the 0-20 cm soil layer at the N1 and N2 gradients increased significantly with the advance of the rice growth period, but decreased slightly at the N0 gradient. At the end of the growth period, the SNRW content of the 40-60 cm soil layer decreased by 57.4% at the N1 gradient and increased by 47.4% and 53.7% at the N2 and N0 gradients, respectively. The SNRW content of each soil layer at the N1 gradient decreased with an increase in soil depth, while it showed the opposite trend at the N2 and N0 gradients. The SNRW content of the 0-20 cm soil layer was the largest at the N1 gradient, which was 1.4 times and 5 times that at N2 and N0, respectively, and it was the smallest of the 40-60 cm soil layer at the N1 gradient, which was 33.6% and 85% of that at N2 and N0, respectively.

Under R3 irrigation, for a 0-20 cm soil layer, the SNRW content increased with an increase in fertilization, and it was 67.6% and 77.9% lower at the N2 and N0 gradients, respectively, than that at the N1 gradient. For the 40-60 cm soil layer, the SNRW content was the smallest at the N1 gradient, which was 76.9% and 80.4% of that at N2 and N0, respectively. The SNRW content of the 0-20cm soil layer at the N1 and N0 gradients accumulated significantly with the advance of the growth period, which was 7.4 times and 2.1 times, respectively, but decreased by 30.7% at the N2 gradient compared with that at the beginning of the growth period, while the opposite trend was shown in the 40-60 cm soil layer. Therefore, an appropriate reduction in nitrogen fertilizer application was conducive to promoting the utilization of NRW in the 0-20 cm soil layer, while excessive fertilization could enrich the SNRW content and hinder the absorption and utilization of NRW.

NF Distribution in Soil and Crop Systems

The NF distributions in soil and crop systems under different irrigation water sources and fertilization gradients are shown in Table 4. Under CK irrigation, PNF and FNUE were the largest, at 0.7653 g/plant and 62.7%, respectively. Compared with CK, PNF, and FNUE under R1, R2, and R3, they were reduced by 10.7%, 29.2%, 8.6%, and 10.6%, 29.6%, and 8.6%, respectively. For the N1 gradient, compared with N2, SNF was increased by 28.8%, but FNUE was decreased by 9.5%, indicating that high fertilization may exceed the nitrogen demand of crops. Under R2 irrigation, SNF and FNRE were the largest, which were 0.8371g/barrel and 17.4%, respectively, with a decrease of 37.5%, 59.3%, 72.4%, and 35.9%, 60.9%, and 73.3% under R1, R3, and CK, respectively. For the N1 gradient, compared with N2, SNF was increased by 5.4%, but FNRE was decreased by 17.8%. LNF and FNLE were 1.8865 g/barrel and 38.5%, respectively, with a decrease of 13.5%, 7.2%, 15.3%, and 14.7%, 6.6%, and 9.2% under R1, R3, and CK, respectively. For the N1 gradient, compared with N2, LNF and FNLE were increased by 35.4% and 5.4%, respectively. It indicated that higher fertilization can increase PNF and SNF contents, but reduce FNUE and FNRE, so it can significantly improve

Table 4. Distributions of fertilizer nitrogen (NF) in soil and crop system under different irrigation water sources and fertilization gradients.

Treat-ment	NF (g/barrel)	PNF (g/ plant)	FNUE (%)	SNF (g/barrel)	FNRE (%)	LNF (g/barrel)	FNLE (%)
R1N1	5.5	0.7609b	55.3b	0.4162d	7.6e	2.0402a	37.1b
R1N2	4.28	0.6065d	56.7b	0.6309b	14.7c	1.2231e	28.6e
R2N1	5.5	0.6296d	45.8c	0.8311a	15.1b	2.1505a	39.1a
R2N2	4.28	0.4536e	42.4d	0.8431a	19.7a	1.6225c	37.9ab
R3N1	5.5	0.7843b	57.0b	0.4593c	8.4d	1.9035b	34.6c
R3N2	4.28	0.6149d	57.5b	0.2222f	5.2f	1.5982c	37.3ab
CKN1	5.5	0.8544a	62.1a	0.2766e	5.0f	1.8058b	32.8d
CKN2	4.28	0.6761c	63.2a	0.1854g	4.3g	1.3902d	32.5d
Variance analysis							
R	-	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)
N	-	** (P = 0.00)	NS (P = 0.54)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)
RN	-	NS(P = 0.94)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)

Notes:PNF, SNF, and LNF represented nitrogen fertilizer (NF) absorption by crops, residue in soil, and losses.
FNUE, FNRE, and FNLE represented NF use efficiency, residue rate in soil and loss rate.
R and N represented irrigation water resource and nitrogen fertilization gradient, respectively.
NS = not significant at the 0.05 level.
* = significant at the 0.05 level.
** = significant at the 0.01 level.

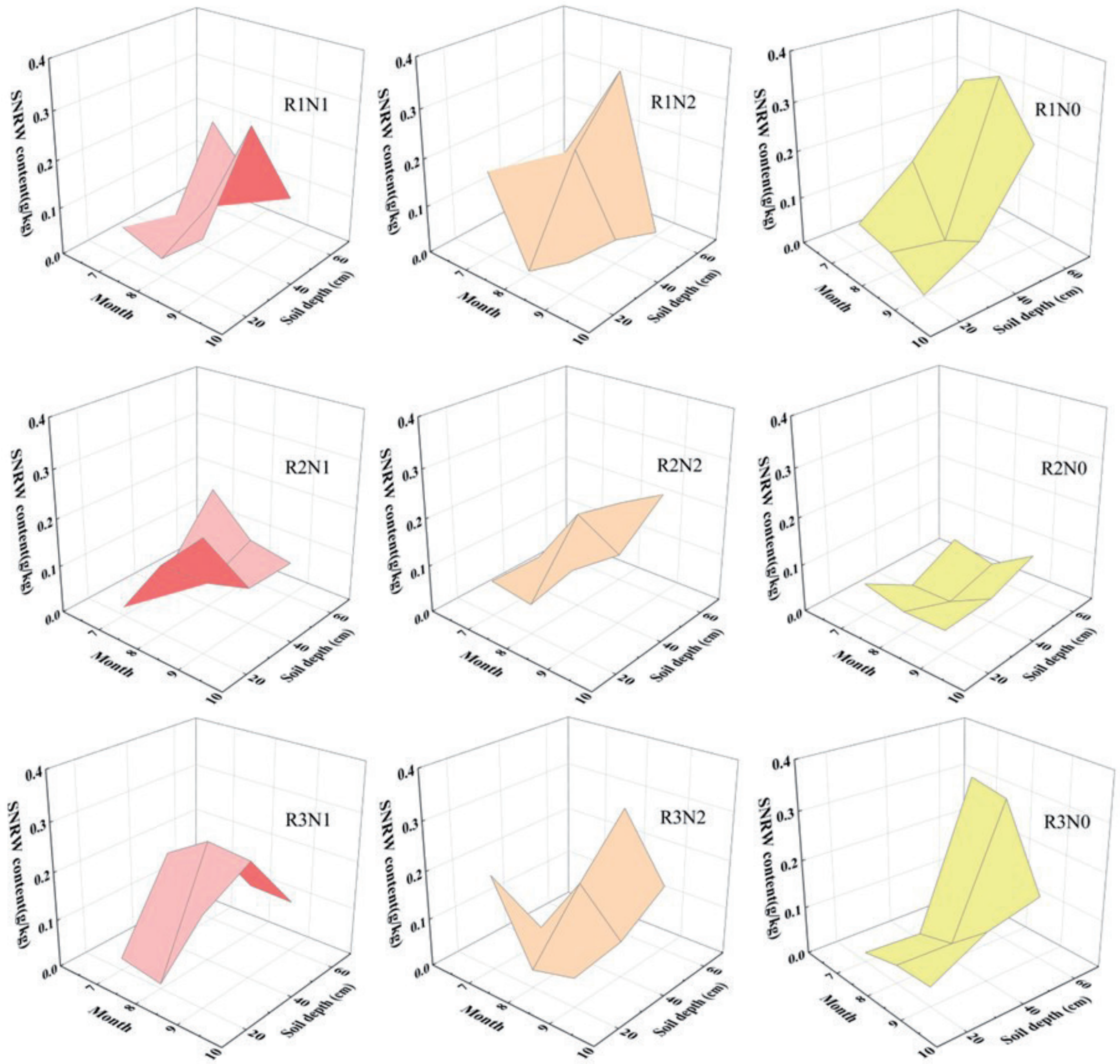

Fig. 3. Nitrogen distributions in soil from rural domestic reclaimed water (SNRW) under different irrigation water sources and fertilization gradients.

FNLE. In addition, the results of variance analysis showed that, except FNUE did not respond positively to the nitrogen fertilization gradient, and PNF did not respond positively to the interaction of irrigation water source and nitrogen fertilization gradient, the responses of other index values to irrigation water source, nitrogen fertilization gradient, and the interaction of two factors showed extremely significant differences (P<0.01).

NRW Distribution in Soil and Crop Systems

NRW distributions in soil and crop systems under different irrigation water sources and fertilization gradients are shown in Table 5. The nitrogen content brought in by irrigation water sources increased with the increase in nitrogen concentration in RDRW, and there were significant differences among water sources. Under different irrigation water sources, PNRW content was directly proportional to the change in nitrogen content in RDRW. Compared with R1, the PNRW content was reduced by 66.0% and 90.8% under R2 and R3, respectively. RWNUE was the largest under R1, and there was little difference between R2 and R3. Both SNRW and RWNLE were the largest under R2 irrigation and the smallest under R1 irrigation. RWNRE decreased with the increase in nitrogen concentration in RDRW, which was both twice that under R2 and R3 irrigation. LNRW was the smallest under R3 irrigation, which was 4.9 times and 10.8 times that under R1 and R2 irrigation, respectively. It indicates that the higher the nitrogen concentration in RDRW, the greater the PNRW content, however, RWNUE was not directly proportional to the nitrogen concentration, and RWNRE was inversely proportional to the nitrogen concentration.

Under different fertilization gradients, PNRW and RWNUE were the largest at the N2 gradient and the smallest at the N1 gradient. Compared with N1, PNRW was increased by 88.0% and 56.6% at N2 and N0, respectively. Therefore, appropriate fertilizer application

Table 5. Distributions of nitrogen brought in by reclaimed water (NRW) in soil crop system under irrigation water sources and fertilization gradients.

Treat-ment	NRW (g/barrel)	PNRW (g/ plant)	RWNUE (%)	SNRW (g/barrel)	RWNRE (%)	LNRW (g/ barrel)	RWNLE (%)
R1N1	1.65a	0.2306b	55.9c	0.1626b	9.9e	0.565b	34.2b
R1N2	1.68a	0.3534a	84.1a	0.0418f	2.5g	0.2246d	13.4e
R1N0	1.64a	0.3530a	86.1a	0.095d	5.8f	0.133e	8.1f
R2N1	1.21b	0.0607e	20.1f	0.1264c	10.4e	0.8408a	69.5a
R2N2	1.22b	0.1798c	59.0b	0.1765a	14.5d	0.3243c	26.6c
R2N0	1.24b	0.0779d	25.1e	0.0609e	4.9f	0.8675a	70.0a
R3N1	0.27d	0.0105g	15.6g	0.1761a	65.2a	0.0519f	19.2d
R3N2	0.34c	0.0341f	40.1d	0.0902d	26.5c	0.1134e	33.4b
R3N0	0.28d	0.0416f	59.4b	0.0914d	32.6b	0.0222g	7.9f
Variance analysis							
R	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)
N	NS (P = 0.09)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)
RN	NS (P = 0.30)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)

Notes: PNRW, SNRW, and LNRW represented nitrogen brought in by RDRW (NRW) absorption by crops, residue in soil, and losses.
RWNUE, RWNRE, and RWNLE represented NRW use efficiency, residue rate in soil and loss rate.
R and N represented irrigation water resource and nitrogen fertilization gradient, respectively.
NS = not significant at the 0.05 level.
* = significant at the 0.05 level.
** = significant at the 0.01 level.

was conducive to improving the plant's absorption and utilization of NRW, while excessive fertilizer application would play an inhibitory role. SNRW and RWNRE increased with the increase in fertilization. For the N1 gradient, compared with N2 and N0, SNRW and RWNRE were increased by 50.8%, 88.1%, and 45.3%, 49.4%, respectively. LNRW and RWNLE were the largest at the N1 gradient and the smallest at the N2 gradient, and RWNLE at the N1 gradient was 67.4% and 42.9% higher than that at N2 and N0, respectively. Therefore, SNRW and LNRW were the largest and PNRW was the least at the N1 gradient, while PNRW was the largest and LNRW was the least at the N2 gradient. In addition, the results of variance analysis showed that the fertilization gradient and the interaction of the irrigation water source and fertilization gradient had no significant effect on the NRW ($P>0.05$), while the irrigation water source, fertilization gradient, and their interaction had an extremely significant effect on the other indexes ($P<0.01$).

Nitrogen Uptake and Utilization by Rice Plants

The uptake amount and utilization rate of NF by each part of the plants under different irrigation water sources and fertilization gradients are shown in Fig. 4. It showed that uptake and utilization of NF by rice grain were less than those of rice leaf and stem, which was mainly due to the late formation of rice grain, and nitrogen absorption mainly occurs in the middle and late stages of rice growth. Under the same irrigation water source, at N1 and N2 gradients, the average NF uptake for rice grain was 0.2521 g/plant and 0.1955 g/plant, respectively, with little difference between utilization rates, which were 18.33% and 18.25%, respectively. The average NF uptake for rice leaf and stem was 0.5052 g/plant and 0.3923 g/plant, respectively, and the utilization rates were 36.73% and 36.68%, respectively. Under the same fertilization gradient, for rice grain, the NF uptake decreased with the increase in nitrogen concentration of RDRW. Compared with CK, the NF uptake under R1, R2, and R3 irrigation decreased by 15.0%, 27.4%, and 6.9%, respectively. As for rice leaf and stem, the average NF uptake was consistent with that of rice grain. Compared with CK, the NF uptake under R1, R2, and R3 irrigation decreased by 8.5%, 30.1%, and 9.4%, respectively. It indicated the absorption of NF by rice grain and rice leaf and stem was hindered under RDRW irrigation. The variance analysis is shown in Table 6. It showed that the irrigation water quality had a very significant difference on the NF uptake and utilization of each part of the plant ($P<0.01$), and the fertilization gradient had a very significant difference on the NF uptake ($P<0.01$), but had no significant effect on the utilization rate of each part of the plant ($P>0.05$).

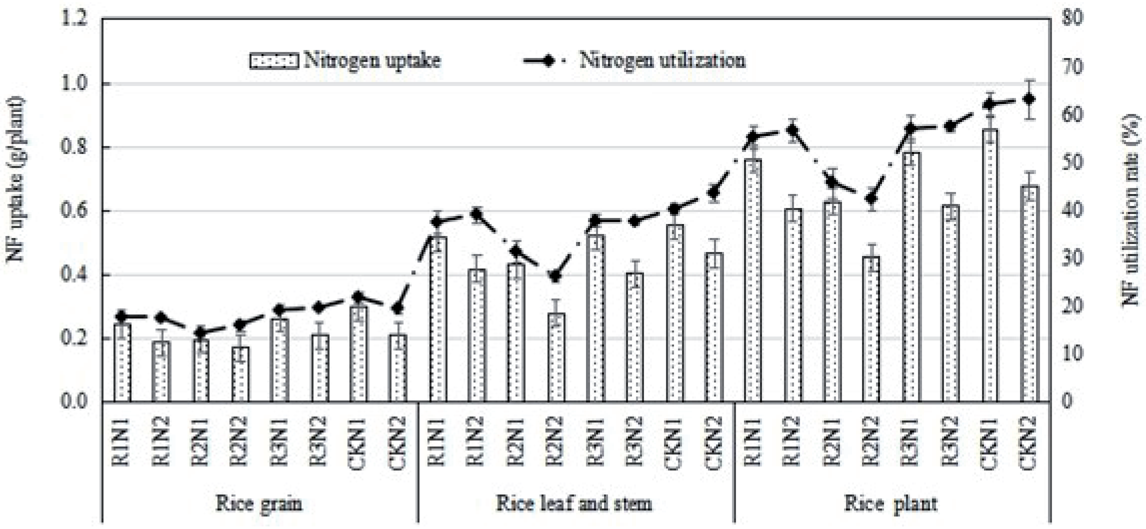

Fig. 4. Uptake amount and utilization rate of fertilizer nitrogen (NF) by each part of rice plant.

Table 6. Variance analysis of uptake amount and utilization rate of fertilizer nitrogen (NF) by each part of rice plant.

Treatment	Grain		Leaf and stem		Plant	
	Uptake-N (g/ plant)	Utilization rate (%)	Uptake-N (g/ plant)	Utilization rate (%)	Uptake-N (g/ plant)	Utilization rate(%)
R	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)	** (P = 0.00)
N	**(P = 0.00)	NS (P = 0.26)	** (P = 0.00)	NS (P = 0.88)	** (P = 0.00)	NS (P = 0.54)
RN	** (P = 0.00)	**(P = 0.00)	* (P = 0.04)	** (P = 0.00)	NS (P = 0.94)	** (P = 0.00)

Notes: R and N represented irrigation water resource and nitrogen fertilization gradient, respectively.
NS = not significant at the 0.05 level.
* = significant at the 0.05 level.
** = significant at the 0.01 level.

Nitrogen uptake by rice plants from fertilizer, RDRW, and soil is shown in Fig. 5. It showed that NS was an important source of nitrogen absorption by rice plants, regardless of CK or RDRW irrigation. According to sections 3.3 and 3.4, the amount of fertilization increased from 4.28 g/barrel to 5.5 g/barrel, and PNF under CK irrigation was increased from 0.68 g/plant to 0.85 g/plant, while from 0.61~0.63 g/plant to 0.45~0.78 g/plant under RDRW irrigation, PNRW was decreased from 0.19 g/plant to 0.10 g/plant under RDRW irrigation, which indicated that with an increase of nitrogen fertilizer application, the contribution rate of NF was increased, but compared with CK irrigation, the contribution rate of NF was reduced under RDRW irrigation, the nitrogen absorption and utilization of RDRW was inhibited, and the contribution rate of NRW was decreased. With the increase in nitrogen concentration in RDRW and the decrease in fertilization gradient, PNRW and PNS increased, but PNF decreased.

NRW Availability in Rice Plants

The responses of PNF and FE to nitrogen application (x) under different irrigation water sources are shown in Fig. 6. The regression equations between nitrogen application rate and PNF under R1, R2, and R3 irrigation were $PNF = -0.0028x^2 + 0.1535x + 5E-15$, $PNF = 0.007x2 + 0.0762x + 4E-15$, $PNF = -0.0009x2 + 0.1474x + 5E-15$, respectively, with the FE of PNRW of 1.5453~3.1233 g/barrel, 0.7468~1.9926 g/barrel, and 0.0712~0.2820 g/barrel, respectively. It showed that the FE of PNRW increased with an increase in the nitrogen concentration of RDRW. In order to quantify the availability of NRW on rice plant growth, the regression equation between FE and nitrogen application of NRW was established, which were $FE = -0.0981x2 + 0.2526x + 3.1233$, $FE = -0.2304x2 + 1.2317x + 0.9407$, and $FE = -0.0217x2 + 0.0809x + 0.282$ under R1, R2, and R3 irrigation, respectively. RFE of R1N1, R2N1, R3N1, R1N2, R2N2, and R3N2 was calculated as 28.1%, 13.6%, 1.3%, 56.3%, 46.6%, and 5.4%, respectively. It indicated that the PNRW significantly improved with an increase in the nitrogen concentration of RDRW, while the RFE decreased with an increase in the nitrogen fertilization gradient. Therefore, a lower fertilization gradient can replace more NF under RDRW irrigation.

Discussion

The efficient utilization of nitrogen in paddy fields is the key to achieving a high yield of rice, reducing nitrogen fertilizer input, and establishing environment-friendly agriculture. In the soil and crop systems, the amount of nitrogen fertilizer applied significantly changed the distribution and transformation of nitrogen in the soil and crop systems. This study showed that the PNF increased with an increase in nitrogen fertilizer

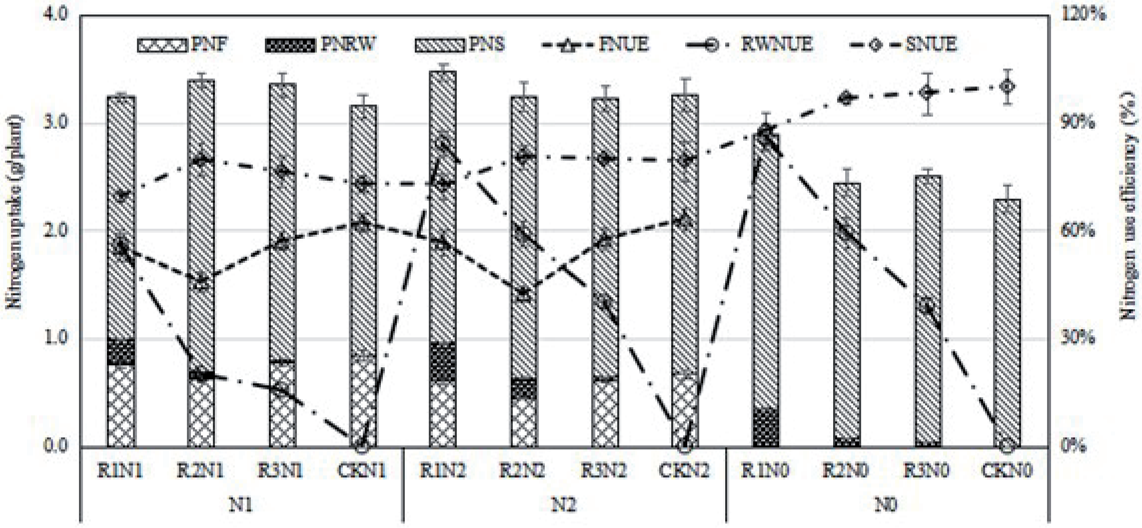

Fig. 5. Nitrogen uptake by rice plant from fertilizer, rural domestic reclaimed water (RDRW) and soil.
Notes: PNF, PNRW, and PNS represented nitrogen uptake by rice plants form fertilizer, RDRW, and soil. FNUE, RWNUE, and SNUE represented use efficiency of NF, NRW, and NS.

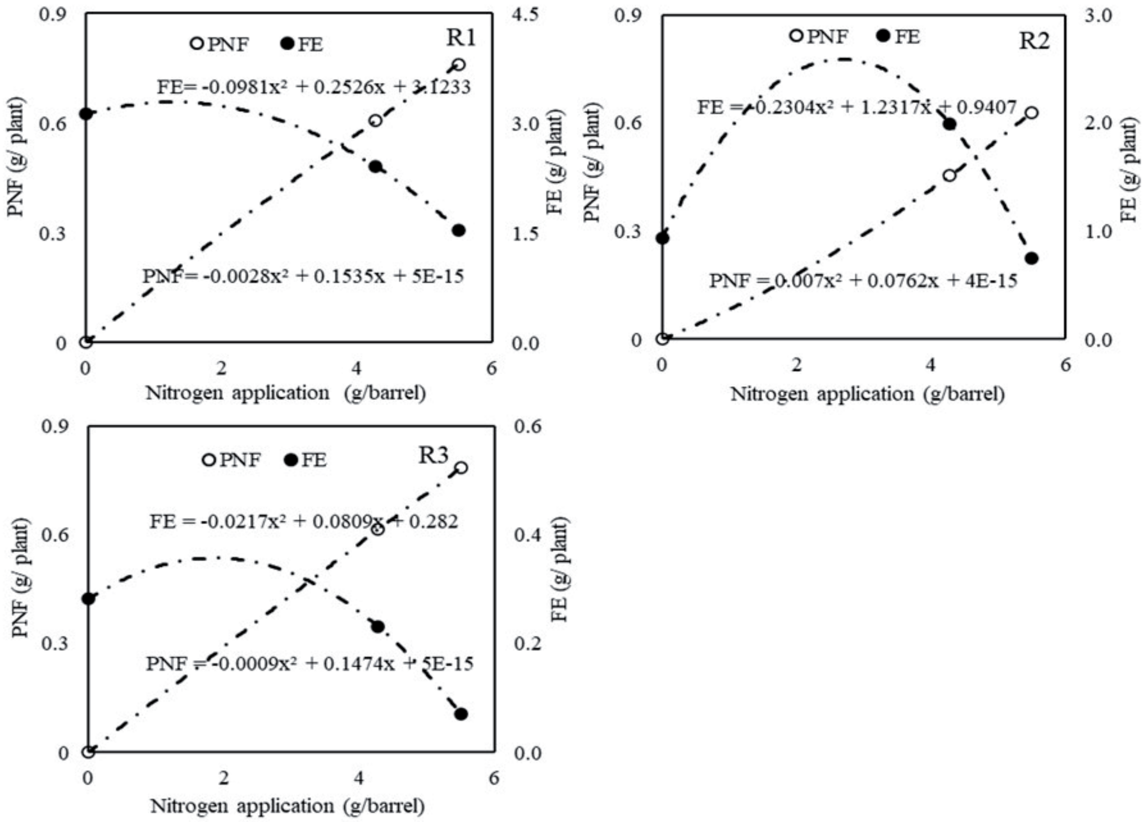

Fig. 6. The response of fertilizer nitrogen absorption by rice plants (PNF) and fertilizer nitrogen substitution equivalent (FE) to nitrogen application under different irrigation water sources.

application, but the FNUE decreased. This was similar to the results of Zhang [18], who found that high nitrogen fertilization increased the absorption of NF but reduced the absorption of NS by rice plants, resulting in an increase in nitrogen content in paddy soil and a decrease in nitrogen use efficiency. In addition, nitrogen content in paddy soil will increase with an increase in nitrogen content in reclaimed water and irrigation water [19]. This study found that there was a surplus of nitrogen in paddy soil, and NF absorption by rice plants was hindered under RDRW irrigation with high nitrogen concentration (R1) and high fertilization gradient (N1). Therefore, in order to ensure a higher FNUE, the joint action of R1 water resource irrigation and N1 fertilization gradient should be avoided. Guo studied the effects of water quality and nitrogen application rate on soil nitrogen balance and nitrate leaching loss through field experiments [20]. It was concluded that the combination of sewage irrigation and a high nitrogen application rate expanded the apparent nitrogen loss and nitrate leaching loss, which was basically consistent with the results of this paper.

Due to RDRW's rich nitrogen content, reasonable irrigation could promote crop growth and increase crop yield [21]. Rice plants irrigated with RDRW could absorb nitrogen not only from the soil and fertilizer, but also from RDRW. In this study, the ^{15}N tracer combined with the fertilizer equivalent method was used to quantitatively evaluate the NRW availability for rice growth. It showed that PNRW was significantly improved with an increased nitrogen concentration in RDRW. There was a quadratic curve relationship between NRW availability and the amount of nitrogen fertilizer applied. Under a high fertilization gradient (N1), SNRW and LNRW increased, while PNRW and RWNUE decreased, and the RFE for NRW decreased, which showed that the RFE was the highest under R1N2 treatment. Therefore, reducing fertilizer application was conducive to improving the efficient utilization and availability of NRW on rice plants. Li [22] and Zaragoza [23] found that reclaimed water irrigation can promote nitrogen absorption, but increasing nitrogen application reduced the NRW availability for corp growth, which was basically consistent with the results of this study. However, the above results had a short test cycle and lacked long-term experimental verification. Whether long-term RDRW irrigation would cause a large amount of nitrogen loss and the impact mechanism on the nitrogen utilization of soil and crop systems still needs to be further studied.

Conclusions

The effects of different irrigation water sources and different fertilization gradients on the efficient utilization and availability of nitrogen in paddy rice were studied using ^{15}N tracer technology combined with the fertilizer equivalent method. The main conclusions were as follows:

(1) SNF was accumulated significantly in the 0-20 cm soil layer under RDRW irrigation at a lower fertilization gradient (N2), which was significantly higher for R2 irrigation than that for R1 irrigation at a higher fertilization gradient (N1). The SNRW content with fertilization was significantly higher than that without fertilization in the 0-20 cm soil layer, which decreased gradually with an increase in fertilization in the 40-60 cm soil layer.

(2) PNF and SNF content increased, but FNUE and FNRE decreased, resulting in FNLE significantly increasing at the N1 gradient. The nitrogen content in irrigation water increased with an increase in nitrogen concentration in RDRW, but RWNUE was not directly proportional to the nitrogen concentration in RDRW. SNRW and LNRW were the highest, but RWNUE was the lowest under the N1 gradient, while RWNUE was the highest and LNRW was the lowest under the N2 gradient.

(3) NS was the main source of nitrogen uptake by rice. With the increase in nitrogen fertilizer application, plant uptake and the contribution rate of NF increased. Compared to the N2 gradient, the PNF content at the N1 gradient increased by 28.8%. Compared with CK irrigation, the absorption of NF by plants was hindered, the contribution rate of NF decreased, the absorption and utilization of nitrogen were inhibited, and the contribution rate of NRW was reduced under RDRW irrigation.

(4) Reducing the fertilization gradient can effectively improve NRW availability. Under N1 and N2 fertilization gradients, the RFE of R1, R2, and R3 was 28.1% and 56.3%, 13.6% and 46.6%, 1.3% and 5.4%, respectively. It is suggested that 30% and 10% reductions of nitrogen fertilization can be used for R1 and R2 irrigation, which can not only reduce fertilizer application, but also fully utilize the effectiveness of nitrogen in reclaimed water and reduce nitrogen loss in paddy fields.

Abbreviations

RDRW, rural domestic reclaimed water; NF, fertilizer nitrogen; NS, soil nitrogen; NRW, reclaimed water nitrogen; SNF, nitrogen in soil from NF; SNRW, nitrogen in soil from NRW; PNF, nitrogen absorbed by crops from NF; PNS, nitrogen absorbed by crops from NS; PNRW, nitrogen absorbed by crops from NRW; LNF, NF losses; LNRW, NRW losses; FNUE, NF use efficiency; FNRE, NF residue rate in soil; FNLE, NF loss rate; RWNUE, NRW use efficiency; RWNRE, NRW residue rate in soil; RWNLE, NRW loss rate; SNUE, NS use efficiency; FE, NF substitution equivalent; RFE, NF relative substitution equivalent.

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Conflicts of Interest

The authors declare that they have no conflicts of interest to report regarding the present study.

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