

Effects of mild alternate wetting and drying irrigation and rice straw application on N₂O emissions in rice cultivation

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Abstract. The shortage of water resources and the decline in soil organic matter (SOM) are ~~critical important~~—limiting factors affecting the ~~improvement of~~improvement in rice productivity, while alternate wetting and drying (AWD) irrigation and recycling application of rice straw

~~return~~ are considered ~~favour~~favorable mitigation measures. However, ~~theirs~~ impact of such measures on rice yield and greenhouse gas (GHG) emissions, especially nitrous oxide (N₂O) emissions, needs to be further clarified; ~~to ensure that which is essential for the development of~~ agronomic ~~practises~~measures for save water ~~savings~~, conserve soil ~~fertilization~~, and reduce GHG ~~reduction~~. Therefore, we explored the effects of mild AWD irrigation combined with on-site rice straw ~~recycling~~return on N₂O emissions and rice yield through rice pot experiments. This experiment included two irrigation methods (continuous flooding (CF) irrigation and mild AWD irrigation)–, two nitrogen (N) application levels (0 and 225 kg N ha⁻¹) and two rice straw (S) return levels (0 and 9000 kg ha⁻¹), for a total of 10 treatments, and each treatment had three replicates. ¹⁵N-urea and ¹⁵N-S were added to the soil, ~~respectively~~. The results showed that N₂O emissions were ~~primarily~~mainly affected by urea application and irrigation methods, ~~withand~~ urea application being most important~~was the main reason~~. Compared with CF irrigation, mild AWD irrigation increased cumulative N₂O emissions, with an average increase of 28.8%. In addition, adding rice straw to mild AWD irrigation further stimulated N₂O emissions by 18.1%. Under the condition of urea application, compared with CF irrigation, mild AWD irrigation increased the yield-scaled N₂O emissions by 17.9%, and the addition of rice straw further promoted the yield-scaled

N₂O emissions under mild AWD irrigation by 17.4% but reduced the global warming potential (GWP) (methane (CH₄) + N₂O) by 62.9%. Under the condition of urea application, compared with CF irrigation, mild AWD irrigation reduced the uptake of soil-derived N and aboveground biomass of rice, but did not reduce rice yield. Therefore, mild AWD irrigation combined with rice straw ~~returning~~return may be a promising agronomic ~~method~~measure to ~~maintain~~ensure rice yield, reduce ~~the~~greenhouse ~~gases~~effect, and ~~protect~~ ~~maintain~~or improve soil fertility.—

1 Introduction

Rice is ~~at~~the staple food for more than half of the world's population, and ensuring rice production is crucial to food security (Tang and Cheng, 2018). More than 135 million hectares of rice are cultivated worldwide, and approximately 90% of paddy fields are submerged (Wang et al., 2017a). Feeding a growing population under water scarcity ~~is~~will be a major challenge to Asia's food security in the coming decades (Lampayan et al., 2015). In China, more than 60% of freshwater resources are consumed by rice cultivation every year, which ~~represents not only~~causes a great waste of freshwater ~~and~~but ~~also~~ causes many environmental problems, such as nonpoint source pollution, eutrophication ~~of water~~ ~~bodies~~, and GHG emissions (Liao et al., 2020). Therefore, it is urgent to

explore new methods for managing paddy field fertilization ~~management~~
~~measures~~ that can ensure high rice yield, and reduced water waste and
environmental pollution.

AWD irrigation is an effective water-~~conserv~~saving irrigation
method that can save approximately 23% of ~~fresh-water~~freshwater
resources compared with CF irrigation (Bouman and Tuong, 2001; Chu et
al., 2014). There are usually two ~~approaches~~types of to alternate wetting
and drying irrigation: severe AWD irrigation (soil water potential $\geq -30 \pm 5$
kPa) and mild AWD irrigation (soil water potential $\geq -15 \pm 5$ kPa) (Zhou et
al., 2017). Severe AWD irrigation could reduce rice yield by 22.6% due
to water stress, but under mild AWD irrigation, rice yield can be~~was~~
stable or slightly increased (Carrijo et al., 2017; Zhou et al., 2017).
Therefore, mild AWD irrigation may offer~~be~~ a more promising paddy
field management model. Previous studies have shown that AWD
irrigation can significantly reduce CH₄ emissions but considerably
significantly promote s N₂O emissions, while GWP mitigation is
dependent on the magnitude of the increase in the release of N₂O
emissions (Lagomarsino et al., 2016; Liang et al., 2017; Kritee et al.,
2018; Tan et al., 2018). Therefore, exploring the impact of mild AWD
irrigation on GHG emissions, especially N₂O-~~emissions~~, is conducive to
reducing paddy field emissions and maximizing agricultural and
environmental benefits.

Long-term cultivation without organic matter supplementation leads to serious degradation of cultivated land and ~~reduction of~~reduction in soil organic matter (SOM) content (Zhou et al., 2021), which is not conducive to rice production and sustainable agricultural development (Chen et al., 2016). Straw return to the field is ~~considered to be an~~ valuable important measure for improving SOM (Huang et al., 2021), and it is beneficial for reducing the environmental pollution caused by burning straw or discarding it randomly (Wang et al., 2018). In addition, rice straw return may ~~also~~ cause changes in paddy field GHG emissions (Naser et al., 2007; Ye et al., 2017; Sun et al., 2018; Yu et al., 2021), N use efficiency (Liu et al., 2021), and rice yield (Chen et al., 2016; Ku et al., 2019). Although there have been many studies on mild paddy AWD irrigation or rice straw return, few studies have focused on the effects of mild AWD irrigation combined with rice straw return on rice cultivation. Under the conventional fertilization mode, whether mild AWD irrigation combined with rice straw ~~returning~~return can achieve the optimal goals of water saving, yield increase, and ~~reduction of~~reduction in greenhouse gas emissions ~~has remained unclear~~needs to be further explored. Therefore, the purpose of this study was to investigate the effect of mild AWD irrigation combined with rice straw ~~returning~~return on N₂O emissions and rice yield in rice cultivation, and to explore the supply of N to rice growth ~~supply of~~ from the soil, urea, and rice straw ~~using to rice growth through~~

¹⁵N ~~labeling~~labelling technology. ~~Our initial~~ ~~We put forward the~~
~~following~~ hypotheses ~~were that~~: 1) Mild AWD irrigation would promote
N₂O ~~emission~~emissions in rice cultivation; ~~and~~ 2) Mild AWD irrigation
would maintain or promote rice yield.

2 Materials and methods

2.1 Experimental ~~set-up~~setup

~~A~~The pot experiment was conducted in an open greenhouse at the
Shenyang Experimental Station of the Institute of Applied Ecology,
Liaoning Province, China (43 °32'N, 123 °23'E) from June 17th to October
27th, 2020. The test soil was an Alfisol with a total C content of 16.01
g/kg and a total N content of 1.36 g/kg.

The pot experiment ~~used~~~~was designed with~~ a ~~completely~~ random
block design, including 30 pots (30 cm diameter × 20 cm height). This
experiment included two irrigation methods, two N application levels,
and two rice straw return levels, with three replicates of each combination,
for a total of 30 rice pots (urea and straw were ~~labelled~~labeled with ¹⁵N,
respectively). The two irrigation methods were CF irrigation and mild
AWD irrigation. CF irrigation maintained a water level depth of
approximately 3-5 cm throughout the rice-growing season. Mild AWD
irrigation water management in the first 7 days was consistent with CF
irrigation and ~~allowed to evaporate under monitoring; then it was~~
~~naturally pursued. W~~when the soil negative pressure gauge reached -15

kPa, it was sub-flooded to a depth of 3-5 cm again and then naturally allowed to ~~dryied up~~ again. This step was repeated until harvest. CF and mild AWD irrigation were halted 2 weeks before harvest. N was applied at 0 kg N ha⁻¹ (CK (control check) and ¹⁵S (rice straw)) and 225 kg N ha⁻¹ (106.13 mg kg⁻¹ dry soil) (¹⁵U (urea), U¹⁵S (urea + rice straw) and ¹⁵US (urea + rice straw)). The abundance of urea ¹⁵N was 10.20%. Urea was applied three times: base fertilizer 40% (June 17th), tiller topdressing 30% (August 4th) and heading topdressing 30% (August 25th). Rice straw return was applied at 0 kg ha⁻¹ (CK and ¹⁵U) and 9,000 kg ha⁻¹ (4.25 g kg⁻¹ dry soil) (¹⁵S, U¹⁵S and ¹⁵US). The total N content of un~~labelled~~~~labeled~~ rice straw was 0.72%, and the isotope abundance of ¹⁵N was 0.59%. The total N content of ~~labelled~~~~labeled~~ rice straw was 0.73%, and the ¹⁵N isotope abundance was 22.94%. The rice straw was ground and applied together with the base fertilizer. Phosphate fertilizer was superphosphate (150 kg P₂O₅ ha⁻¹), and potassium fertilizer was potassium chloride (185 kg K₂O ha⁻¹) as a one-time application of basic fertilizer. Every pot was filled with 10.51 kg (9 kg dry soil) of sieved (2 mm) fresh soil. Two hills of rice were planted in each pot. At maturity, the rice yield and aboveground biomass were recorded after being oven dried (105 °C for 0.5 h and 60 °C for 12 h).

2.2 Soil sample collection and analysis

At the regreening stage, tillering stage, jointing stage, booting stage,

filling stage, and maturity stage, five points were randomly selected from the 0-10 cm soil layer of each pot and mixed. The soil $\text{NH}_4^+\text{-N}$ and $\text{NO}_3^-\text{-N}$ were extracted with 2 mol L^{-1} KCl solution (Wu et al., 2019), filtered and ~~analysed~~analyzed with a continuous flow ~~analyser~~analyzer (AA3, Bran + Luebbe, Germany). The extraction of soil $^{15}\text{N-NH}_4^+\text{-N}$ ~~was follow by~~followed Yu et al. (2020). Soil microbial biomass N (MBN) was fumigated with chloroform, extracted with 0.5 mol L^{-1} K_2SO_4 (soil: solution = 5 g: 20 ml) (Joergensen et al., 1996), and determined by a TOC ~~analyser~~analyzer (Elementar vario TOC Analyzer, Germany). The soil $^{15}\text{N-NH}_4^+\text{-N}$ content, $^{15}\text{N-MBN}$ and ^{15}N of rice aboveground biomass were determined by a stable isotope ratio mass spectrometer (253 MAT, Thermo Finnigan, Germany).

2.3 Gas sampling and calculation

The static chamber method was used to determine the N_2O flux (Li et al., 2018a). The static chamber with a top seal made of transparent plexiglass consisted of two parts, namely, the base and the gas collecting chamber. The base had a diameter of 31 cm, a groove in the middle, and a height of 10 cm. The gas-collecting chamber had a diameter of 30 cm and a height of 70 cm. A small fan and a thermometer were installed in the gas-collecting chamber. N_2O was collected every two days in the first week after fertilization or irrigation and every seven days ~~during on the~~ other ~~periods~~days. N_2O was sampled at 8:00–11:00 a.m. each sampling

day. Every pot was sealed with water when N₂O was collected. Three gas samples were collected at 0, 30 and 60 min after the chamber was airtight, and N₂O was collected with a 50 mL injector and then injected into 200 mL gasbags.

The N₂O concentration was ~~analysed~~~~analyzed~~ ~~using~~by a gas chromatograph (Agilent 7890B, Gas Chromatograph, Delaware, USA).

The calculation of N₂O fluxes was as follows (Li et al., 2018a):

$$F = \rho \times h \times dc/dt \times 273/(273 + T)$$

where F is the N₂O flux ($\mu\text{g m}^{-2} \text{ h}^{-1}$); ρ is the N₂O standard-state density (1.964 kg m^{-3}); h is the chamber height above the soil (m); c is the N₂O concentration; dc/dt is the slope of the N₂O concentration curve, estimated using a linear regression model (Vitale et al., 2017); 273 is the gas constant; and T is the average air temperature inside the chamber during N₂O collection (°C).

Cumulative N₂O emissions (CE) were calculated using the following formula according to Wang et al. (2011):

$$CE (\text{kg N}_2\text{O ha}^{-1}) = \sum_{i=1}^n \left(\frac{F_i + F_{i+1}}{2} \right) (t_{i+1} - t_i) \times 24 \times 10^{-2}$$

where i is the various sampling times, t is the sampling date, n is the total measurement time and 10^{-2} is the conversion factor.

The contribution of ¹⁵N markers to NH₄⁺-N and MBN and the calculation of the N source of the aboveground biomass of rice was

~~follow by~~ followed Ma et al. (2015). Yield-scaled N₂O was calculated as the ratio between N₂O and rice yield (Li et al., 2018a).

2.4 Statistical analysis

All analyses were performed using SPSS Statistics 16.0 (SPSS, Inc., Chicago, USA). One-way ANOVA was conducted to test the treatment effects with Duncan's test. Significant differences were set at alpha = 0.05. Univariate analysis of variance was used to ~~analyse~~analyze the response of cumulative N₂O emissions to irrigation method, N level and rice straw application (Table 2). Tables and figures were prepared with Excel 2016 (Microsoft Corp., USA) and Origin 8 (Origin Lab Corp., USA), respectively. The data in the figures and tables are the average value ± standard error.

3 Results

3.1 N₂O flux

Three higher N₂O flux peaks appeared after basal fertilizer and two topdressing treatments (Figure 1), and the N₂O flux peak after basal fertilizer application was significantly larger than the last two peaks. After basal fertilizer was applied, the N₂O flux peaks of CF irrigation and mild AWD irrigation were similar. After the first topdressing, the N₂O flux peak of mild AWD irrigation was significantly greater than that of CF irrigation, ~~about~~approximately 1.4 and 9.1 times under ~~the~~ U and US treatments, respectively. In contrast, the N₂O flux peak of CF irrigation

after the second topdressing was 3.5 and 1.6 times higher than that of mild AWD irrigation under [the](#) U and US treatments, respectively. In addition to the above three peaks, the N₂O flux of CF irrigation was close to zero, and mild AWD irrigation had a lower flux peak with alternating wet and dry conditions. The flux of N₂O ranged from -103.93 $\mu\text{g m}^{-2} \text{h}^{-1}$ to 2,770.50 $\mu\text{g m}^{-2} \text{h}^{-1}$. The N₂O flux appeared negative in the late stage of rice growth. The N₂O fluxes of CF irrigation and mild AWD irrigation were similar in the later stage of rice growth, indicating that drainage had little effect on it. During the entire [rice](#) growth cycle ~~of rice~~, the N₂O flux of the CK and S treatments was low. There were significant differences in the peak N₂O fluxes between the different treatments. Compared with CK, the application of rice straw alone significantly promoted the N₂O flux on the first day after CF irrigation and mild AWD irrigation. Compared with U, the addition of rice straw in CF irrigation reduced the N₂O flux, while the addition of rice straw in mild AWD irrigation increased the N₂O flux.

3.2 Soil NH₄⁺-N, NO₃⁻-N and MBN concentrations

The soil NH₄⁺-N, NO₃⁻-N, and MBN concentrations varied with the growing stage of rice (Fig. 2A, 2C and Fig. 3A). The soil NH₄⁺-N concentration first increased and then decreased. The NH₄⁺-N concentration under CF irrigation and mild AWD irrigation was low in the late [rice](#) growth period ~~of rice~~ (Fig. 2A). The concentration of NO₃⁻-N in CF-irrigated soil showed a trend of first increasing and then decreasing

and was at a low level in the later growth period of rice, while the concentration of NO_3^- -N in mild AWD irrigation showed a trend of first decreasing and then increasing, ~~with repeated then-decreas~~esing and ~~then~~ increasesing (Fig. 2C). The concentration of MBN in CF-irrigated soil first decreased and then increased and then decreased to a lower level, while the concentration of MBN in mild AWD-irrigated soil decreased with the growth stage of rice (Fig. 3A).

There were significant differences in the NH_4^+ -N, NO_3^- -N, and MBN concentrations between the different treatments. In CF irrigation and mild AWD irrigation, the U treatment had a higher NH_4^+ -N concentration in the early stage of rice growth, while the other treatments had no significant difference, but as rice grew, the NH_4^+ -N concentration of the US treatment increased, which was significantly greater than that of the U, S and CK treatments (Fig. 2A). In CF irrigation, the NO_3^- -N concentration of the U treatment was slightly higher than that of the other treatments, and all treatments had little ~~variancedifference~~. In mild AWD irrigation, the NO_3^- -N concentration of the US treatment was significantly higher than that of the U, S and CK treatments. The U treatment had a higher NO_3^- -N concentration than the S and CK treatments in the later stage of rice growth (Fig. 2C). The US treatment in CF irrigation and mild AWD irrigation had the highest MBN concentration during the rice growth period (Fig. 3A).

Figure 2B shows that the $\text{NH}_4^+\text{-N}$ in CF irrigation and mild AWD irrigation mainly came from urea rather than rice straw, and the combined application of urea and rice straw further promoted the release of $\text{NH}_4^+\text{-N}$ from urea. Regardless of CF irrigation or mild AWD irrigation, rice straw N was difficult to utilize by microorganisms in the first year under single rice straw application, but rice straw and urea combined application significantly promoted the utilization of rice straw N by microorganisms. Urea combined with rice straw application may be more easily utilized by microorganisms than urea applied alone (Fig. 3B).

3.3 Sources of aboveground biomass N in rice

As shown in Fig. 4, under CF irrigation and mild AWD irrigation, compared with CK, a single application of rice straw did not increase the aboveground N absorption of rice, while the U and US treatments significantly promoted the aboveground N absorption of rice. Under mild AWD irrigation, the US treatment reduced N uptake in rice shoots compared with the U treatment. The U and US treatments under CF irrigation promoted the N uptake of the aboveground rice more than those under mild AWD irrigation.

With different irrigation methods, the effects of urea and rice straw addition on the N absorption of the aboveground rice were varieddifferent. Compared with the CK and S treatments, the U and US treatments under CF irrigation significantly promoted the absorption of soil N by rice,

while only [the](#) S and U treatments had significant differences under mild AWD irrigation. Compared with mild AWD irrigation, the U and US treatments under CF irrigation significantly promoted the absorption of soil N by rice. Regardless of the irrigation and fertilization method, the soil was the main source of N in the aboveground parts of rice, followed by urea and finally rice straw (Fig. 4).

3.4 Cumulative N₂O emissions, rice agronomic properties and yield-scaled N₂O emissions

In addition to the CK treatment, compared with CF irrigation, mild AWD irrigation significantly promoted the accumulation of N₂O during the rice growth period, with an average increase of 28.8% (Table 1). Under CF irrigation, there was no significant difference in the accumulation of N₂O between S and CK or between US and U. However, the addition of rice straw under mild AWD irrigation significantly increased the accumulation of N₂O by 18.1% (Table 1). Compared with the CK and S treatments, the U and US treatments significantly promoted cumulative N₂O emissions under the two irrigation modes. As shown in Table 2, irrigation methods, N application level, and rice straw return affected cumulative N₂O emissions, of which the N application level had the greatest impact. The interaction between irrigation level and N fertilizer or rice straw significantly affected cumulative N₂O emissions.

As shown in Table 1, compared with CF irrigation, mild AWD

irrigation significantly reduced rice aboveground biomass under US treatment but had no effect on other treatments. Regardless of whether CF irrigation or mild AWD irrigation was applied, there was no significant difference in the rice aboveground biomass between S and CK or between US and U. Compared with the CK and S treatments, the U and US treatments significantly promoted the rice aboveground biomass under the two irrigation modes. Irrigation level had no effect on rice yield under all treatments. Regardless of CF irrigation or mild AWD irrigation, rice yield under the U and US treatments was significantly higher than that under the CK and S treatments, but there was no difference between the former two and the latter two.

In addition to the CK treatment, compared with CF irrigation, mild AWD irrigation significantly promoted yield-scaled N₂O emissions during the rice growth period. Under urea application conditions, compared with CF irrigation, mild AWD irrigation increased yield-scale N₂O emissions by 17.9%, and the addition of rice straw further promoted yield-scale N₂O emissions by 17.4% under mild AWD irrigation conditions (Table 1). Regardless of CF irrigation or mild AWD irrigation, yield-scaled N₂O emissions under the U and US treatments were significantly higher than those under the CK and S treatments. Rice straw addition had no effect on the yield-scaled N₂O emissions under CF irrigation but significantly increased the yield-scaled N₂O emissions

under mild AWD irrigation.

4 Discussion

4.1 Effects of irrigation methods, N levels, and rice straw return on N₂O emissions

N₂O ~~emission is~~emissions are significantly affected by water-filled pores and mineral N (NH₄⁺-N and NO₃⁻-N) content (Allen et al., 2010). The N₂O emission peak in CF irrigation occurred only after N application, while mild AWD irrigation caused other N₂O emission peaks, which might have been caused by the change in soil moisture conditions by mild AWD irrigation (Zhou et al., 2020). The peak of N₂O after fertilization may be ~~due to the fact that~~ because a large amount of N application increases the soil inorganic N concentration (Fig. 2A and 2C), which in turn promotes the generation of N₂O, which ~~mainly comes~~comes from the denitrification process (Wang et al., 2017b; Yano et al., 2014). During the denitrification process, it is easier for microorganisms to use NO₃⁻-N as an electron acceptor (Fig. 2C), which affects the reduction process of N₂O, resulting in an increase in the ratio of N₂O/N₂ in the denitrification products (Pérez et al., 2000). Our results showed a negative N₂O emission flux at the later stage of rice growth, which may be due to the decrease ~~of~~in surface soil N₂O concentration due to the strengthening of the N₂O reduction process or the weakening of the N₂O diffusion process in the soil profile, which allowed atmospheric N₂O to diffuse back into the soil

(Chapuis-Lydie et al., 2007). Mild AWD irrigation promoted cumulative N₂O emissions by 28.8% on average, which was ~~as proposed in our~~ ~~the same as Hypothesis hypothesis~~ 1. Similar results were found in previous studies, which may be due to the increased N₂O produced by nitrification and denitrification due to water level alternation (Liang et al., 2017; Zhou et al., 2020) and temperature change (Wu et al., 2019) of mild AWD irrigation. ~~In order to~~ To reduce N₂O emissions from paddy fields, researchers generally regulate N₂O production by optimizing N management (Liang et al., 2017), applying inhibitors to control the N supply rate of N fertilizers (Wu et al., 2019). ~~, etc.~~ In both CF and mild AWD irrigation, there was no obvious N₂O emission peak at the later growth stage of rice, which may have been due to the decrease in soil inorganic N content (Fig. 2A and 2C) and microbial biomass (Fig. 3A).

Compared with CK and S, N fertilizer application (U and US) significantly increased N₂O cumulative emissions and was the most ~~notable important~~ of N₂O generation (Table 2), mainly because N fertilizer application provided sufficient substrates for soil nitrification and denitrification to generate N₂O (Fiedler et al., 2017; Wu et al., 2021). The peak of N₂O emissions after base fertilizer application was larger than that after two topdressing, which might have been due to a higher N application rate and simultaneous nitrification and denitrification in initial flooding (Mathieu et al., 2006; Wang et al., 2017b). The peak of N₂O

emissions after the two topdressing treatments with CF irrigation was similar, while the peak of N₂O emissions ~~after~~ the first topdressing treatment with mild AWD irrigation was significantly larger than that ~~following~~ the second topdressing treatment. This may be because the soil environment (temperature, moisture, etc.) changed little in long-term flooding under CF irrigation (Lagomarsino et al., 2016; Verhoeven et al., 2018; Congreves et al., 2019), while the ~~variations~~ difference in the soil environment of the two top dressings under mild AWD irrigation changed the utilization of fertilizer N by microorganisms (Fig. 3B). Therefore, reducing the amount of the first top dressing under mild AWD irrigation and maintaining flooding for approximately one week after fertilization may be ~~benefit~~ beneficial to N₂O reduction (Liao et al., 2020). The negative value of N₂O emissions appeared at the later stage of rice growth, indicating that the paddy field could also become a sink for N₂O (van Groenigen et al., 2015), which might be caused by the lack of N supply at the later stage. Increasing the second topdressing might be beneficial for alleviating N deficiency (Liao et al., 2020).

~~Unlike the~~ Different from CF irrigation, the addition of rice straw under mild AWD irrigation conditions promoted N₂O emissions (Table 1), probably because the alternation of wet and dry conditions promoted the decomposition of rice straw (Andren et al., 1993; Buchen et al., 2016; Chen et al., 2016), which was beneficial to the growth of microorganisms

(Fig. 3A), ~~and~~ thus promoting the production of N₂O (Said-Pullicino et al., 2014; Wang et al., 2018; Wu et al., 2021). In addition, compared with U and S, US promoted microbial absorption of urea and rice straw N (Fig. 3B), which also proved that US treatment was more conducive to the growth of microorganisms.

4.2 Effects of irrigation methods, N levels, and rice straw return on rice production and yield-scaled N₂O emissions

The change in irrigation method did not cause differences in rice yield (Table 1), but under the US treatment, mild AWD irrigation significantly reduced the aboveground biomass of rice and the uptake of soil N by rice (Table 1 and Fig. 4), which was consistent with our Hypothesis ~~hypothesis~~–2. Previous studies have also shown that mild AWD irrigation can stabilize or increase rice yield. This may be because mild AWD irrigation can promote the transport of nutrients from stems and leaves to grains during the reproductive growth stage of rice, while inhibiting ineffective tillering and increasing the number of effective panicles, thereby reducing ~~the~~ excessive vegetative growth of rice (Carrijo et al., 2017; Li et al., 2018b; Liao et al., 2020; Zhang et al., 2009). This may also be an important reason for the decrease in the uptake of soil N by rice under AWD irrigation. Urea application was a key factor in improving rice yield (Wang et al., 2017a) but also aggravated soil N uptake by rice, and soil was the largest source of N for rice in all

treatments (Fig. 4). Compared with U, under CF irrigation and mild AWD irrigation, US reduced the uptake of soil-derived N by rice, and the trend was more obvious under mild AWD irrigation, ~~although~~. Although the trend was not significant, rice straw return may be an effective way to maintain long-term soil fertility (Fig. 4).

In our study, mild AWD irrigation, urea application and rice straw return all increased yield-scaled N₂O emissions (Table 1), mainly due to improved soil aeration and increased inorganic N and rice straw decomposition, resulting in more N₂O production (Andren et al., 1993; Buchen et al., 2016; Chen et al., 2016; Lagomarsino et al., 2016; Fiedler et al., 2017; Verhoeven et al., 2018; Congreves et al., 2019; Wu et al., 2021). Although mild AWD irrigation had higher yield-scaled N₂O emissions than CF irrigation, the GWP (CH₄ + N₂O) under mild AWD irrigation was significantly lower than that under CF irrigation and decreased by 8.1%, 57.9%, 11.8% and 62.9% under CK, S, U and US, respectively (Table S1). Therefore, mild AWD irrigation combined with rice straw return may be a promising agronomic measure that ~~not only~~ maintains ~~ensures~~ rice yield, ~~and~~ slows the greenhouse effects (CO₂ emissions are not considered), ~~and~~ but also reduces soil fertility consumption.

5 Conclusions

The effects of irrigation methods, N levels and rice straw return on

N₂O emissions were explored through pot experiments ~~using~~with rice. We found that N₂O emissions were ~~mainly—affected~~affected by urea application and irrigation methods, ~~with~~and urea application being the most important~~was the main reason~~. Compared with CF irrigation, mild AWD irrigation increased cumulative N₂O emissions, with an average increase of 28.8%. In addition, adding rice straw to mild AWD irrigation further stimulated N₂O emissions by 18.1%. Under the condition of urea application, compared with CF irrigation, mild AWD irrigation increased the yield-scaled N₂O emissions by 17.9%, and the addition of rice straw further promoted the yield-scaled N₂O emissions under mild AWD irrigation by 17.4% but reduced the GWP (CH₄ + N₂O) by 62.9%. Under the condition of urea application, compared with CF irrigation, mild AWD irrigation reduced the ~~—~~uptake of soil-derived N and aboveground biomass of rice, but did not reduce rice yield. Therefore, mild AWD irrigation combined with rice straw ~~returning~~return may ~~offer~~be ~~—~~a promising agronomic measure to maintain high~~ensure~~ rice yield, reduce ~~the~~greenhouse effects, and maintain or improve soil fertility.

Data availability. Original data are available upon request. Material necessary for this study's findings is presented in the paper.

Author contributions. Kaikuo Wu, Wentao Li, Zhanbo Wei and Zhi Dong conceived and designed the experiments; Yue Meng and Na Lv

performed the experiments; Kaikuo Wu analyzed the data; Kaikuo Wu and Lili Zhang wrote the paper and all authors approved submission of the paper. All authors have read and agreed to the published version of the manuscript.

Competing interests. The authors declare that they have no conflict of interest.

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Review statement.

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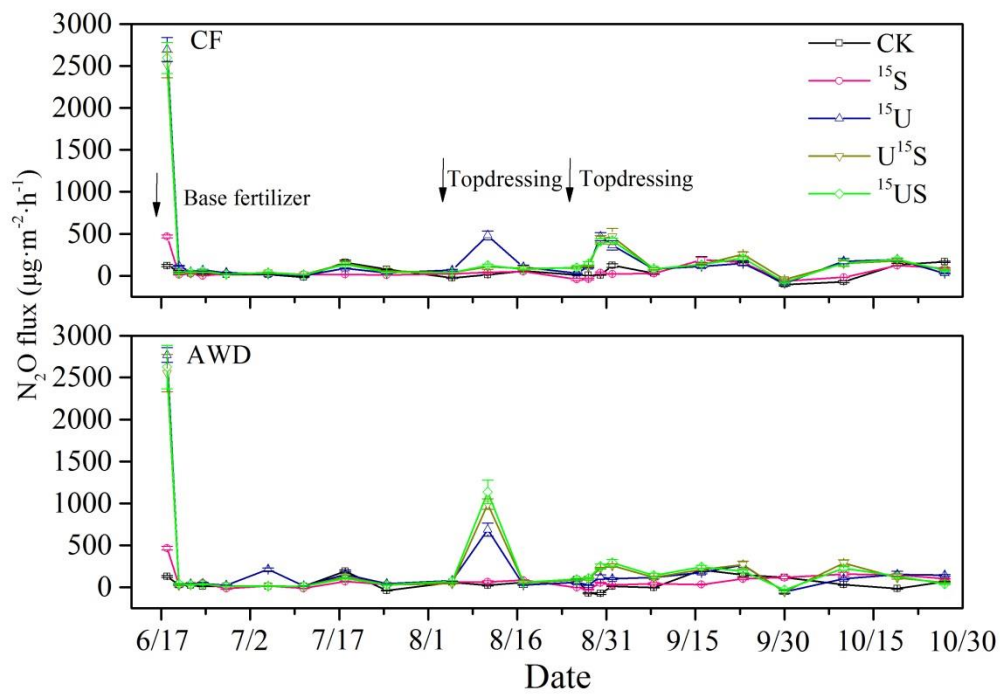
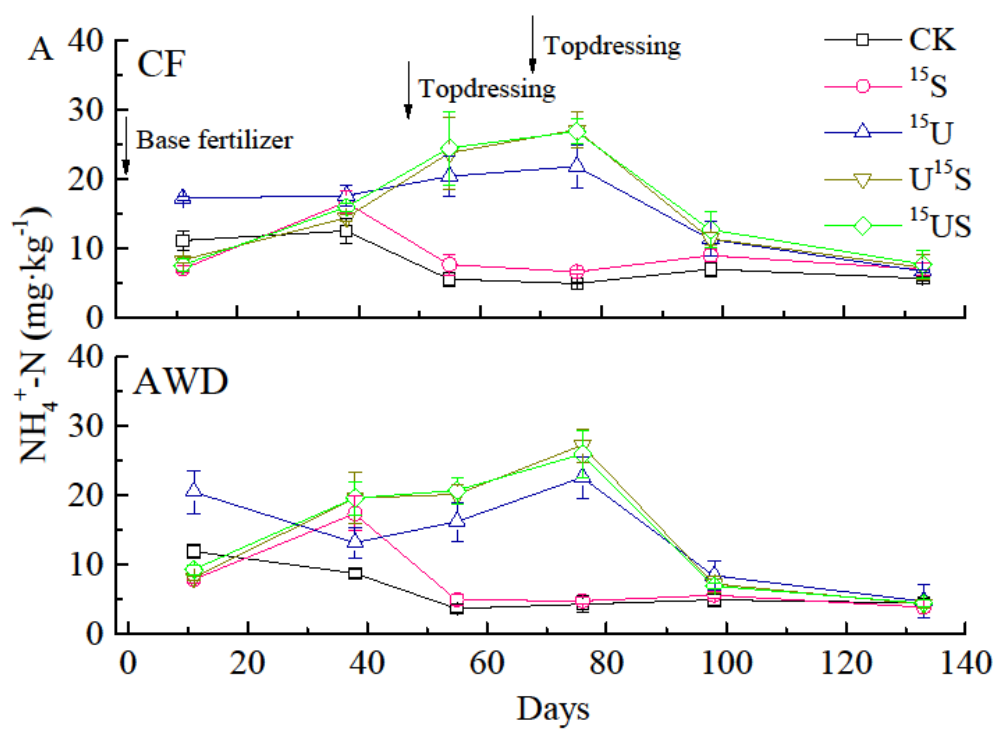
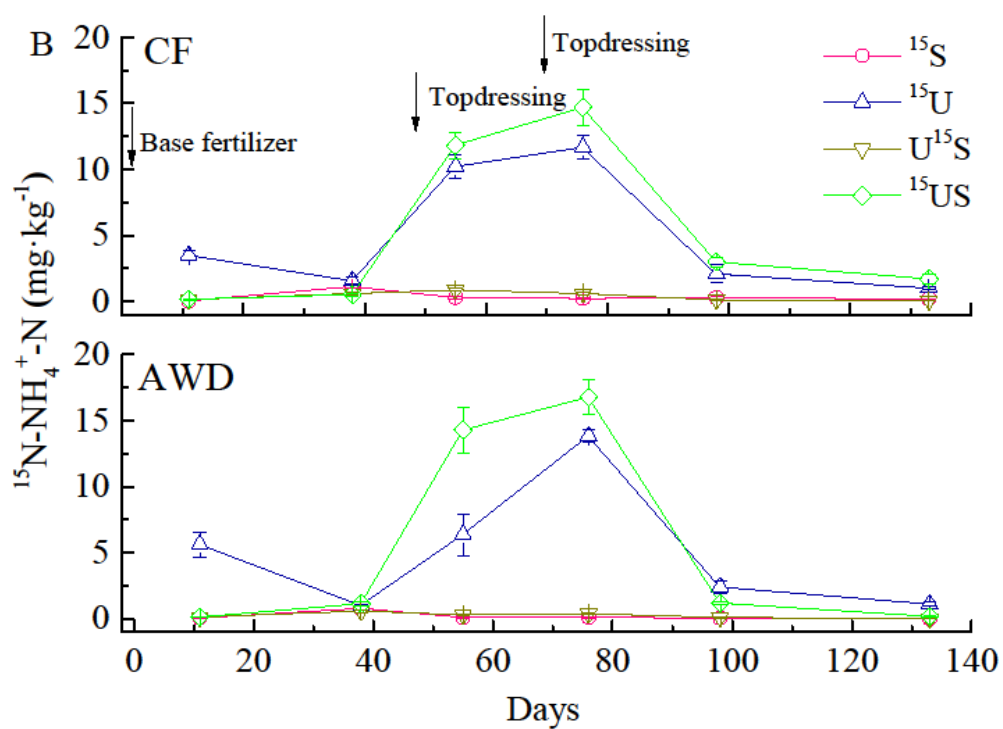


Fig. 1 Effects of different treatments on nitrous oxide (N_2O) flux. Bars represent standard errors (n=3), the same below.



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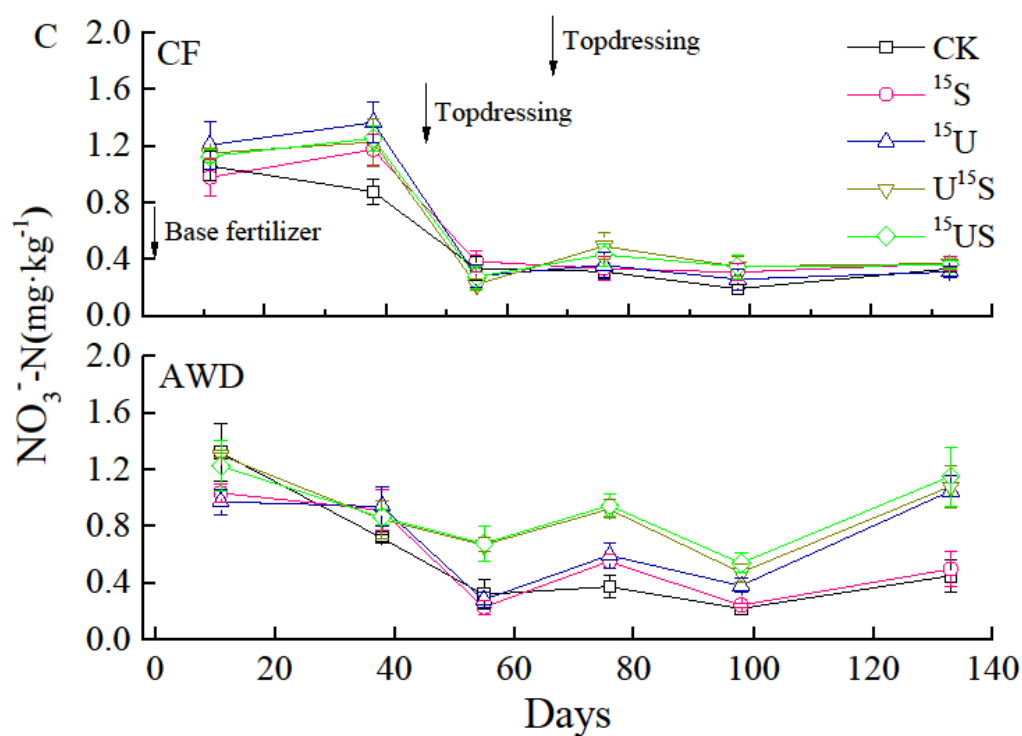


Fig. 2 Changes in soil ammonium nitrogen (NH_4^+ -N) concentration (A), the contribution of ^{15}N markers to NH_4^+ -N (B) and changes in soil nitrate nitrogen (NO_3^- -N) concentration (C) during the growth period of rice (n=3).

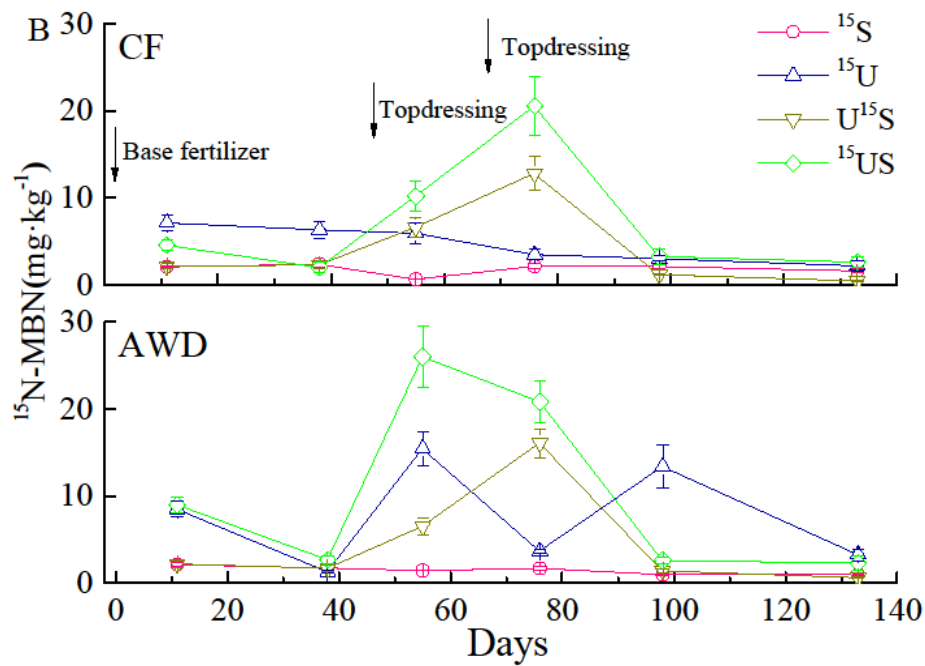
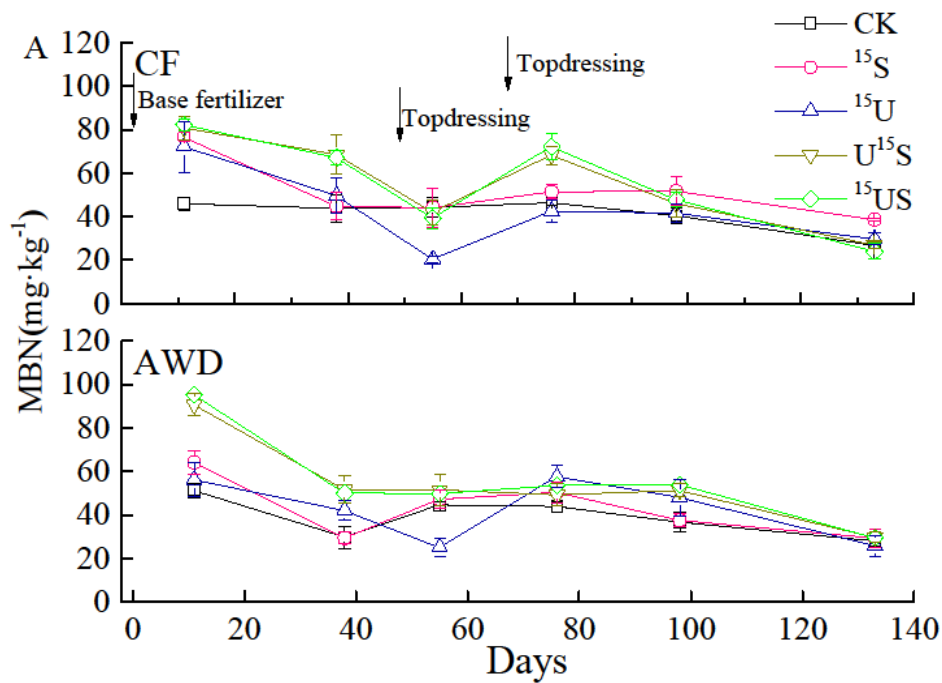


Fig. 3 Changes in the concentration of microbial biomass N (MBN) (A) in the soil during the rice growth period and the contribution of ^{15}N markers to MBN (B) (n=3).

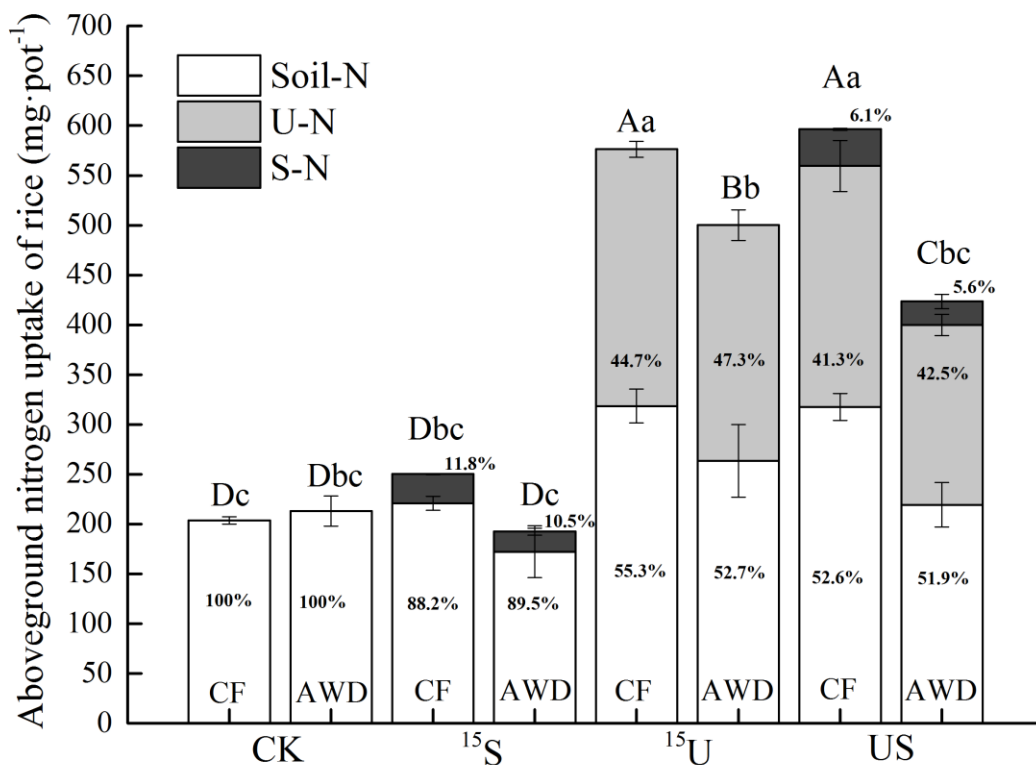


Fig. 4 The source of nitrogen in the aboveground biomass of rice at the maturity stage (n=3). CF: continuous flooding irrigation, AWD: mild alternate wetting and drying irrigation, Soil-N: Soil derived nitrogen, U-N: Urea derived nitrogen, S-N: Rice straw derived nitrogen. Different capital letters indicate significant differences in total nitrogen uptake of rice above ground ($P < 0.05$), and different lower case letters indicate significant differences in soil nitrogen supply ($P < 0.05$).

Table 1 Effects of different treatments on cumulative nitrous oxide (N₂O) emissions, rice aboveground biomass, rice yield and yield-scaled N₂O emission. The values denote means \pm standard errors (n=3). Different lowercase letters indicate significant differences ($P < 0.05$).

Treatments		Cumulative N ₂ O emissions kg ha ⁻¹	Rice aboveground biomass g pot ⁻¹	Rice yield g pot ⁻¹	Yield-scaled N ₂ O emission g kg ⁻¹
CF	CK	1.48 \pm 0.06 ef	37.11 \pm 2.53 f	21.63 \pm 1.81 b	0.24 \pm 0.01 d
	¹⁵ S	1.24 \pm 0.05 f	37.99 \pm 2.69 f	20.91 \pm 1.63 b	0.21 \pm 0.03 d
	¹⁵ U	4.02 \pm 0.30 c	82.54 \pm 10.39 ab	36.56 \pm 2.75 a	0.39 \pm 0.04 c
	U+ ¹⁵ S	3.89 \pm 0.09 c	87.58 \pm 7.70 a	35.98 \pm 1.72 a	0.38 \pm 0.02 c
	¹⁵ U+S	3.76 \pm 0.02 c	80.99 \pm 19.54 abc	35.85 \pm 2.50 a	0.37 \pm 0.03 c
AWD	CK	1.64 \pm 0.15 e	45.31 \pm 3.07 ef	22.92 \pm 1.07 b	0.25 \pm 0.02 d
	¹⁵ S	2.07 \pm 0.17 d	34.78 \pm 4.86 f	20.42 \pm 0.46 b	0.36 \pm 0.02 c
	¹⁵ U	4.48 \pm 0.12 b	64.44 \pm 7.33 bcd	34.81 \pm 1.64 a	0.46 \pm 0.03 b
	U+ ¹⁵ S	5.05 \pm 0.28 a	62.92 \pm 2.49 cde	34.50 \pm 1.76 a	0.53 \pm 0.01 a
	¹⁵ U+S	5.29 \pm 0.25 a	60.44 \pm 6.74 de	34.85 \pm 1.18 a	0.54 \pm 0.03 a

Table 2 Cumulative nitrous oxide (N₂O) emissions in response to irrigation method, nitrogen level, and straw returning. * indicated significant treatment effects within a main category ($P < 0.05$), *** indicated significant treatment effects within a main category ($P < 0.001$).

Factors	Cumulative N ₂ O emissions
Irrigation method (I)	88.576***
Nitrogen level (N)	1525***
Straw (S)	6.393*
I×N	6.275*
I×S	26.288***
N×S	1.426
I×N×S	0.178