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Article

Lime Application Reduces Methane Emissions from a Double-Cropped Rice Field under the Substitution of Chemical Fertilizers by Pig Manure

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Abstract: *Background and Aims* The substitution of chemical fertilizers by organic manure plays a critical role in sustainable crop production. Nevertheless, organic amendments promote the global warming potential (GWP) in rice paddies due to increased methane (CH₄) emissions. Increasing evidence shows that lime application can reduce CH₄ emissions from acidic paddy soils, while it is still not clear whether liming can reduce the GWP in rice fields under organic manure substitution. *Methods* A two-year field experiment was conducted to investigate the impacts of pig manure substitution and lime application on grain yield, CH₄ and nitrous oxide (N₂O) emissions in a subtropical double-cropped rice field in China. The experiment consisted of three treatments: chemical fertilization (CF, 100% N from urea), 50% of chemical N substituted by pig manure (1/2N + M), and the 1/2N + M treatment plus lime amendment (1/2N + M + L). *Results* On average, 1/2N + M reduced rice yield by 5.65% compared to CF, while lime application had no effect on rice yield. Mean cumulative CH₄ emissions were 218.8% higher in 1/2N + M than in CF, whereas 1/2N + M + L reduced CH₄ emissions by 36.6% compared to 1/2N + M. Neither pig manure substitution nor lime application affected N₂O emissions. Consequently, 1/2N + M increased the GWP and greenhouse gas intensity (GHGI) by 214.6% and 228.3%, respectively, compared to CF. In contrast, 1/2N + M + L reduced the GWP and GHGI by 36.4% and 36.5% compared to 1/2N + M. *Conclusion* Lime application can mitigate CH₄ emissions and GWP induced by pig manure amendment in double-cropped rice fields.

Keywords: soil acidification; organic amendment; methane; nitrous oxide; rice yield

1. Introduction

Chemical fertilizer application is the most effective practice to enhance crop yield. Nevertheless, long-term chemical fertilizer application alone, particularly chemical nitrogen (N), may lead to soil acidification and the decline of soil fertility [1,2]. Combined application of chemical fertilizers and organic manure can not only reduce the rate of chemical fertilizers but also recycle organic wastes [3,4]. Furthermore, many studies suggest that an appropriate substitution of chemical fertilizers by organic manure can effectively enhance soil fertility without compromising crop yield [5–7]. However, organic amendment has been shown to promote methane (CH₄) emissions from rice paddies [8]. CH₄ is the second most important greenhouse gas after carbon dioxide, while rice paddies are a significant source of atmospheric CH₄ leading to global warming [9,10]. Thus, it is urgent to mitigate CH₄ emissions from paddy fields under organic manure amendment to improve the sustainability of rice production.

Limes are usually applied to alleviate soil acidification and increase rice yield in acidic paddy fields. Increasing evidence shows that lime application can reduce CH₄ emissions from acidic paddy soils [11,12]. Lime application raises soil pH and enhances microbial activity, thus promoting the mineralization of organic matter in cropland soil and finally reducing carbon (C) substrates for CH₄ production [13,14]. Meanwhile, lime application may promote root growth and improve soil structure, thus enhance oxygen availability in rice rhizosphere and stimulate CH₄ oxidation [15–17]. However, to the best of our knowledge, it is not clear yet whether liming can reduce CH₄ emissions from rice fields under organic manure amendment.

China is the largest rice producer in the world, with the double rice cropping system accounting for approximately 33% of the total rice cultivation area [18]. However, those double-cropped rice fields are primarily located in subtropical regions in China. The inherent low soil pH and long-term chemical N application has led to soil acidification and yield loss in double-cropped rice fields [19,20]. Concurrently, China is the largest pork producer in the world [21]. The subtropical region is also one of the major pig farming areas in China [18]. The substitution of chemical fertilizers by pig manure can not only reduce the rate of chemical fertilizers but also facilitate the recycle of organic wastes from pig farms, thereby playing a critical role in sustainable agroecosystems in subtropical China [22]. Previous studies have shown that liming can reduce CH₄ emissions from rice fields, and have suggested that the combined application of chemical fertilizers and organic manure can enhance soil fertility [6,23–25]. Nevertheless, it is unknown whether lime application could mitigate CH₄ emissions in double-cropped rice fields under pig manure amendment. Therefore, in the present study, we conducted a two-year field experiment to investigate the impacts of lime application on rice yield, CH₄ and N₂O emissions under the substitution of chemical fertilizers by pig manure in a double rice cropping system. We hypothesized that i) pig manure substitution could maintain rice yield while boosting CH₄ emissions; and ii) lime application could reduce CH₄ emissions induced by pig manure amendment in the double-cropped rice paddies.

2. Materials and Methods

2.1. Experimental Site

The field experiment was conducted at the Jiangxi Institute of Red Soil (28°15'30"E, 116°20'24"N), Jinxian Country, Jiangxi Province from 2019 to 2020. This site has a subtropical monsoon climate with a mean annual precipitation of 1684 mm and a mean annual temperature of 17.7 °C. The dynamics of the average air temperature and precipitation during 2019 and 2020 are presented in Figures S1 and S2, respectively. The paddy soil is classified as Typic Stagnic Anthrosol. The main soil properties (0–15cm) before the experiment are as follows: pH (5:1, H₂O: soil, *w/w*) 5.2, organic matter 20.3 g kg⁻¹, total N 1.2 g kg⁻¹, total phosphorus (P) 0.5 g kg⁻¹, total potassium (K) 9.2 g kg⁻¹, alkaline hydrolyzable-N 146.3 mg kg⁻¹, available P 26.3 mg kg⁻¹, and available K 82.1 mg kg⁻¹.

2.2. Experiment Design

The experiment was arranged in a completely randomized block design, with three treatments: CF (chemical fertilizers), 1/2N + M (50% of chemical N substituted by pig manure), 1/2N + M + L (50% of chemical N substituted by pig manure plus lime application). The experiment consisted of three blocks, each containing three plots. Each plot was 6-m-long and 5-m-wide. In the CF plots, N, P, and K fertilizers were applied at rates of 150, 75, and 75 kg ha⁻¹ per rice season, respectively. All the P and K fertilizers were applied as basal fertilizers in the form of calcium magnesium phosphate and potassium chloride, respectively. The urea was split and applied twice: 50% as basal fertilizer and 50% at tillering stage. For the manure-added plots, pig manure was applied as basal fertilizer to substitute all the basal chemical N (i.e., 50% of the total N rate) in each rice season, and the remaining 50% of N was applied by urea at the tillering stage. The rate of pig manure was calculated according to its moisture and N content. The fresh pig manure was collected from a nearby pig farm with the moisture content of 71.3%, total N 26.3 g kg⁻¹, total P 34.2 g kg⁻¹, and total K 13.9 g kg⁻¹. Since the manure application rate was based on its N content, the rate of P was higher in the manure-added

plots compared to the CF treatment, while the deficient K was replenished with potassium chloride in the manure treatments. In the lime-added plots, slaked lime ($\text{Ca}(\text{OH})_2$) was applied only once at a rate of 2.45 t ha^{-1} . Both pig manure and limes were evenly spread across the plots before tillage. The rice cultivars 'Qiliangyou 2012' and 'Taiyou 871' were selected for the early and late rice seasons, respectively. The early rice seeding was transplanted at a hill spacing of $13 \text{ cm} \times 25 \text{ cm}$ with four seedlings per hill, while the late rice seeding was transplanted at a hill spacing of $13 \text{ cm} \times 16 \text{ cm}$ with two seedlings per hill. All the rice straw was returned to the respective plots in each treatment. Water management in each plot was as follows: flooding in the early rice growth stage followed by a mid-season drainage and then intermittent irrigation after reflooding. Chemicals were applied to intensively control weeds and pests.

2.3. Sampling and Measurement

The CH_4 and N_2O emissions were measured at one-week intervals during the rice growth season with the static chamber-gas chromatography technique. After transplanting, a polyvinyl chloride frame (inner size $50 \times 50 \times 15 \text{ cm}$) was inserted into the soil in each plot. Each frame included six rice plants. While sampling, an opaque steel chamber ($50 \times 50 \times 50 \text{ cm}$ or $50 \times 50 \times 100 \text{ cm}$ according to the height of rice plants) wrapped with a heat-insulating layer and equipped with a fan inside the top to blend gases was installed on the frame. The gas samples were collected at each sampling event between 9:00–11:00 am at 10-minute intervals for a total period of 30 min. The CH_4 and N_2O concentrations of the samples were measured by the Agilent 7890A gas chromatograph (Agilent Technologies, Santa Clara, CA, USA). Cumulative CH_4 or N_2O emissions were calculated for each rice growth season as the accumulation at two adjacent intervals of the measurement. Global warming potential (GWP) on a 100-year time scale was calculated according to the Equation (1), and greenhouse gas intensity (GHGI) (i.e., yield-scaled GWP) was calculated according to the Equation (2) [26]. As the experiment only lasted for 2 years, soil C sequestration was not included in the GWP to avoid the noise of short-term fluctuations [27].

$$\text{GWP (kg CO}_2 \text{ eq ha}^{-1}\text{)} = 27 \times \text{CH}_4 \text{ (kg ha}^{-1}\text{)} + 273 \times \text{N}_2\text{O (kg ha}^{-1}\text{)} \quad (1)$$

$$\text{GHGI (kg CO}_2 \text{ eq kg}^{-1}\text{)} = \text{GWP/grain yield (kg ha}^{-1}\text{)} \quad (2)$$

For the grain yield measurement, 200-hill rice plants were harvested from each plot at maturity and then adjusted to a moisture of 14%. In addition, five soil cores at 0–15 cm soil depth were randomly collected in each plot after late rice harvest in 2019 and 2020. Soil pH was measured by the pH meter using air-dried and 2-mm sieved soil at a distilled-water to soil ratio of 5:1 (*w/w*). The soil organic matter (SOM) was determined using wet digestion with $\text{H}_2\text{SO}_4\text{-K}_2\text{CrO}_7$ [28].

2.4. Statistical Analyses

Three-way analyses of variance (ANOVAs) were conducted to examine the effects of treatment (CF, $1/2\text{N} + \text{M}$, and $1/2\text{N} + \text{M} + \text{L}$), crop season (early and late rice), study year, and their interactions on rice yield, CH_4 and N_2O emissions, seasonal GWP and seasonal GHGI. All statistical analyses were conducted by JMP Pro 13 (SAS Institute Inc. Cary, NC, USA). Two-way ANOVAs were carried out to examine the effects of treatment (CF, $1/2\text{N} + \text{M}$ and $1/2\text{N} + \text{M} + \text{L}$), study year, and their interactions on soil pH and SOM.

3. Results

3.1. Grain Yield

Averaged across years and crop seasons, the $1/2\text{N} + \text{M}$ treatment significantly reduced rice yield by 5.7% compared to CF, primarily due to the decrease in the number of panicles (Tables 1 and S1). Relative to the CF treatment, $1/2\text{N} + \text{M}$ significantly increased grain filling by 5.2%, but decreased panicle number by 7.9%. There was no significant difference in the spikelet number per panicle and

1000-grain weight between CF and 1/2N + M treatments (Table S1). In addition, no significant difference in grain yield was observed between 1/2N + M and 1/2N + M + L (Table 1).

Table 1. Cumulative CH₄ and N₂O emissions, area-scaled global warming potential (GWP), yield, and greenhouse gas intensity (GHGI) during the crop growing season as affected by treatment, crop (early and late rice), and study year. *F*-values are provided for interactions.

	CH ₄ (kg ha ⁻¹)	N ₂ O (g ha ⁻¹)	GWP (kg CO ₂ eq ha ⁻¹)	Yield (kg ha ⁻¹)	GHGI (kg CO ₂ eq kg ⁻¹)
Treatment (T) ^a					
CF	149c	413a	4123c	7146a	0.60c
1/2N + M	475a	534a	12973a	6742b	1.97a
1/2N + M + L	301b	444a	8257b	6686b	1.25b
Crop (C) ^b					
Early rice	283b	330b	7740b	6753b	1.16b
Late rice	333a	598a	9162a	6963a	1.38a
Year (Y) ^c					
2019	331a	405b	9043a	7375a	1.26a
2020	286b	522a	7860b	6342b	1.28a
<i>F</i> -values					
T*C	11.2***	NS	11.2***	199***	9.6***
T*Y	13.8***	NS	13.8***	NS	9.9***
C*Y	60.4***	NS	60.3***	NS	246***
T*C*Y	21.2***	NS	21.2***	NS	49.6***

CF, 1/2N + M and 1/2N + M + L represent chemical fertilization, 50% of chemical N substituted by pig manure and 50% of chemical N substituted by pig manure combined with lime amendment, respectively. Significant interactive effects are indicated by * (0.01 < *P* ≤ 0.05), ** (0.001 < *P* ≤ 0.01), or *** (*P* ≤ 0.001). Values followed by different lowercase letters are significantly different among treatments, crops or years. ^a Values were averaged across crops and years. ^b Values were averaged across treatments and years. ^c Values were averaged across treatments and crops.

3.2. CH₄ Emissions

The dynamics of CH₄ fluxes exhibited similarity across rice seasons and treatments. Main CH₄ emissions were detected before midseason drainage with the peaks of CH₄ fluxes occurring at the early tillering stage except during the early rice season in 2020 when no apparent peak was present.

Averaged across crops and study years, 1/2N + M increased cumulative CH₄ emissions by 219.8% compared to CF. Relative to the 1/2N + M treatment, 1/2N + M + L reduced CH₄ emissions by 36.6% (Table 1). There was a significant interactive effect on cumulative CH₄ emissions among treatments, crops, and study years (Table 1). Compared to the CF treatment, 1/2N + M increased CH₄ emissions in the early rice growth season by 466.2% in 2019 and by 173.6% in 2020. Relative to the 1/2N + M treatment, 1/2N + M + L reduced CH₄ emissions in the early rice growth season by 53.6% in 2019 and 41.8% in 2020. Similarly in the late rice growth season, 1/2N + M increased CH₄ emissions by 155.6% in 2019 and 156.4% in 2020, compared to CF. Relative to the 1/2N + M treatment, 1/2N + M + L reduced CH₄ emissions in the late rice growth season by 20.0% in 2019 and 25.6% in 2020 (Figure 3). In addition, compared to the 1/2N + M treatment, the lime-induced reduction in annual CH₄ emissions in the 1/2N + M + L treatment were greater in 2019 (-40.1%) than in 2020 (-32.0%).

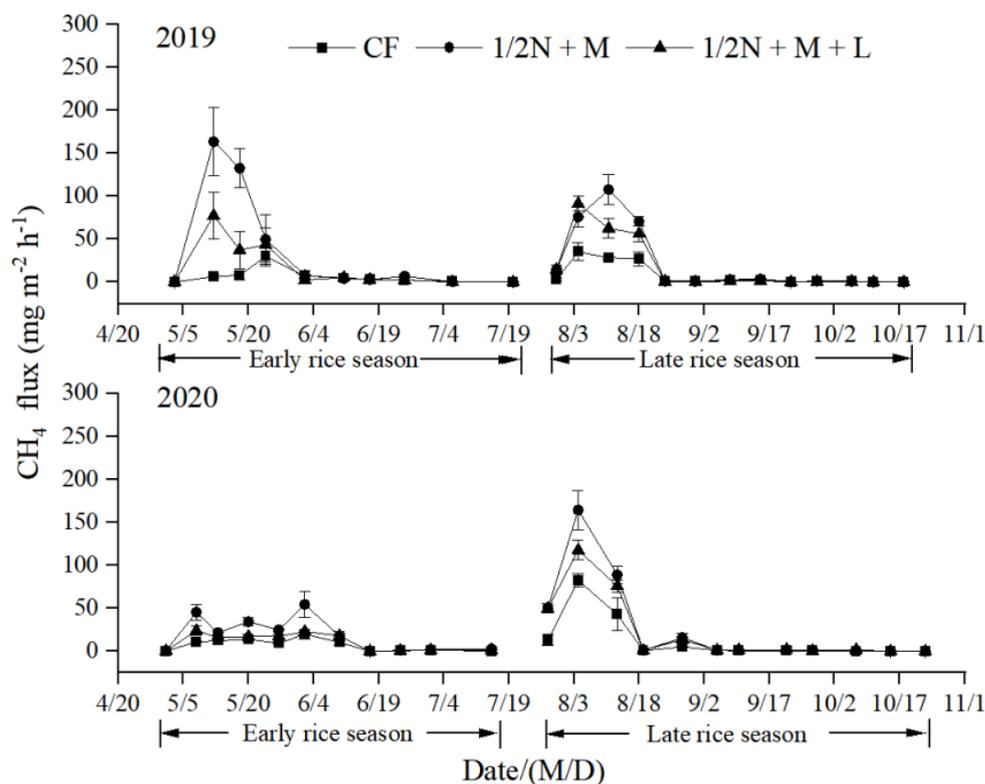


Figure 1. Effects of pig manure and lime application on CH_4 fluxes over two years. CF, 1/2N + M, and 1/2N + M + L represent chemical fertilization, 50% of chemical N substituted by pig manure, and 50% of chemical N substituted by pig manure combined with lime amendment, respectively. Error bars represent the standard deviation of mean ($n=3$).

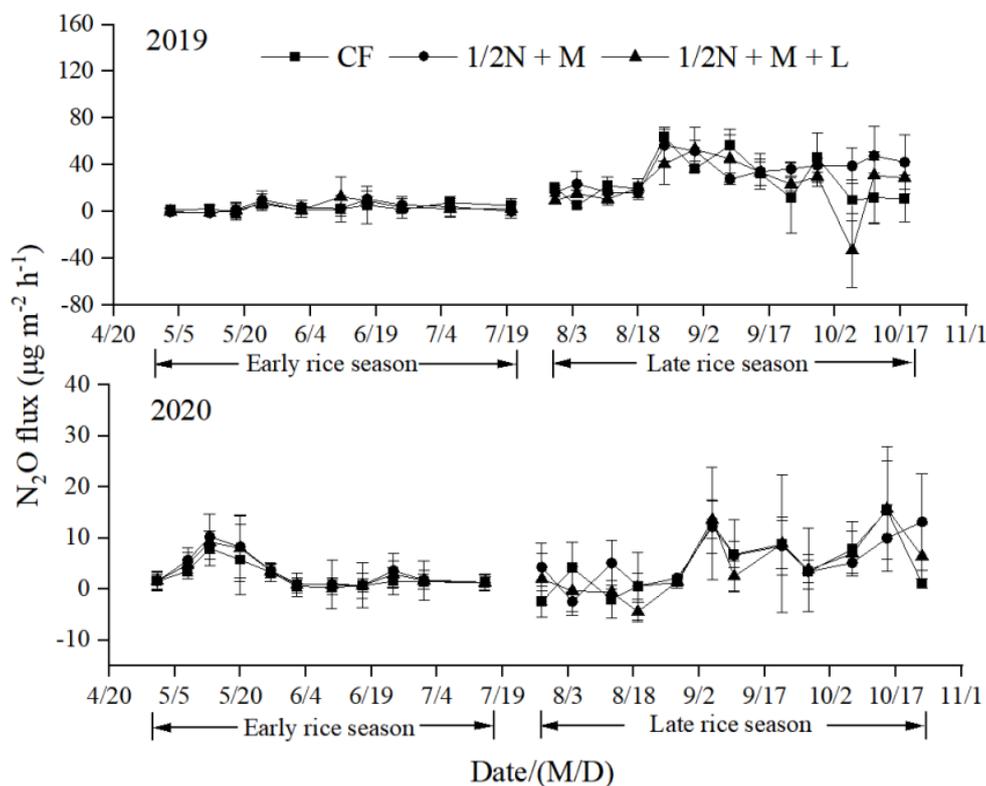


Figure 2. Effects of pig manure and lime application on N_2O fluxes over two years. CF, 1/2N + M, and 1/2N + M + L represent chemical fertilization, 50% of chemical N substituted by pig manure, and 50% of chemical N substituted by pig manure combined with lime amendment, respectively. Error bars represent the standard deviation of mean ($n=3$).

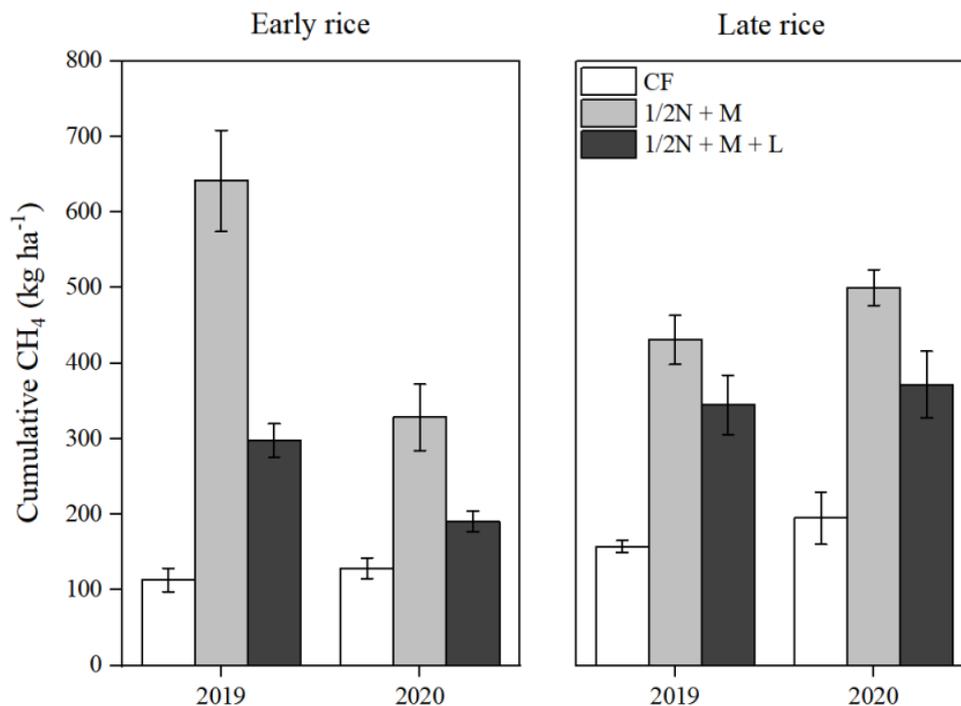


Figure 3. Effects of pig manure substitution and lime application on cumulative CH_4 emissions over the two years, as there was significant treatment \times crop \times year interaction. CF, 1/2N + M, and 1/2N + M + L represent chemical fertilization, 50% of chemical N substituted by pig manure, and 50% of chemical N substituted by pig manure combined with lime amendment, respectively. Error bars represent the standard deviation of the mean ($n=3$).

3.3. N_2O Emissions

Over the two-year experimental period, significant variations in N_2O fluxes were observed in the rice paddy, particularly during the late rice seasons. In the early rice seasons, the N_2O fluxes seemed to be weak sources or sinks of atmospheric N_2O in all treatments due to the abundant rainfall. Conversely, higher N_2O fluxes and greater variation were observed in the late rice season, particularly following mid-season drainage. Overall, no significant differences in cumulative N_2O emissions were detected among the three treatments. However, the cumulative N_2O emissions were higher in the late rice season compared to the early rice (Table 1).

3.4. GWP and GHGI

Since CH_4 emissions accounted for most of the GWP (i.e., by an average of 98.5%) in the present experiment, the GWP exhibited similar trends with CH_4 emissions (Table 1, Figures 3 and 4). Pig manure application significantly increased the GWP in all crop seasons, while the addition of lime markedly mitigated GWP. Averaged across years and crop seasons, 1/2N + M increased the GWP by 214.9% compared to CF. Meanwhile, 1/2N + M + L decreased the GWP by 36.4% compared to the 1/2N + M treatment (Table 1). Relative to the CF treatment, 1/2N + M increased GWP in the early rice growth season by 457% in 2019 and by 153.4% in 2020. Compared to the 1/2N + M treatment, 1/2N + M + L decreased GWP in the early rice growth season by 53.4% in 2019 and by 41.4% in 2020. Relative to the CF treatment, 1/2N + M increased GWP in the late rice growth season by 168.5% in 2019 and by 152.7% in 2020. Compared to 1/2N + M, 1/2N + M + L decreased GWP in the late rice growth season

by 20.1% in 2019 and by 25.4% in 2020 (Figure 4). The lime-induced reduction in the annual GWP in the 1/2N + M + L treatment was 39.9% and 31.8% in 2019 and 2020 compared to 1/2N + M, respectively. Similar to cumulative CH₄ emissions, the mitigation effect of lime application on the GWP was stronger in 2019 than in 2020.

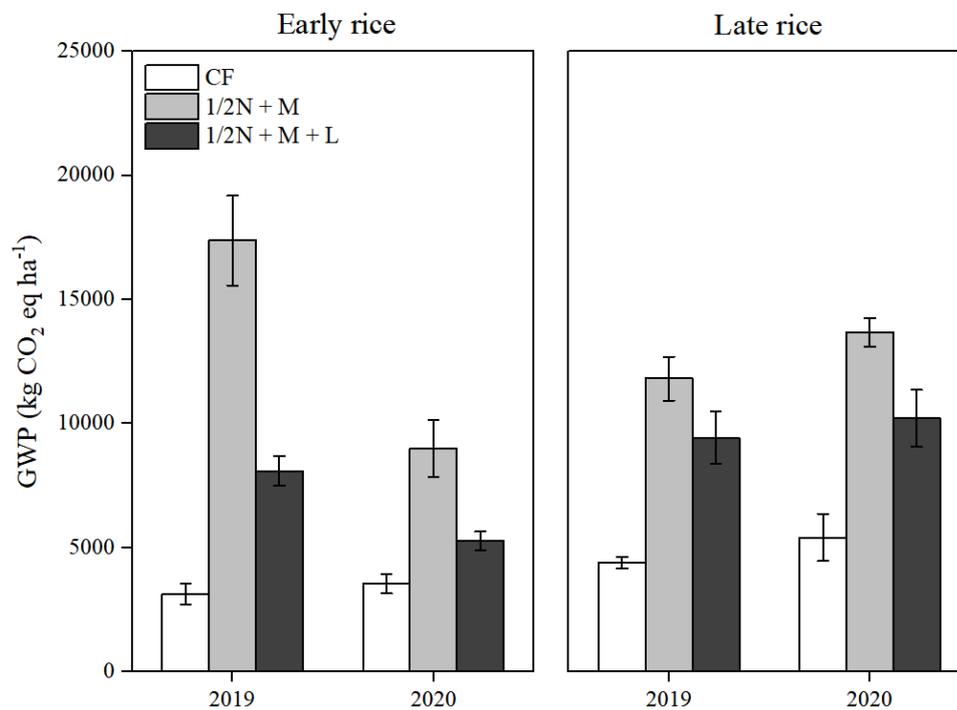


Figure 4. Effects of pig manure substitution and lime application on GWP over the two years, as there was significant treatment × crop × year interaction. CF, 1/2N + M, and 1/2N + M + L represent chemical fertilization, 50% of chemical N substituted by pig manure, and 50% of chemical N substituted by pig manure combined with lime amendment, respectively. Error bars represent the standard deviation of the mean ($n=3$).

Nearly identical to the GWP, relative to CF, 1/2N + M increased the GHGI by 229% averaged across years and crop seasons, while 1/2N + M + L reduced the GHGI by 36.3% compared to 1/2N + M (Table 1). Compared to the CF treatment, 1/2N + M increased GHGI in the early rice growth season by 481.1% in 2019 and by 164.2% in 2020. Relative to the 1/2N + M treatment, 1/2N + M + L reduced GHGI in the early rice growth season by 50.7% in 2019 and by 39.1% in 2020. Similar patterns were observed in the late rice growth season. Compared to the CF treatment, 1/2N + M increased GHGI in the late rice growth season by 194% in 2019 and by 164% in 2020. Relative to the 1/2N + M treatment, 1/2N + M + L reduced GHGI in the early rice growth season by 21.7% in 2019 and by 28.4% in 2020 (Figure 5). Compared to 1/2N + M, the lime-induced reduction in the annual GHGI in 1/2N + M + L was 39.1% and 31.6% in 2019 and 2020, respectively.

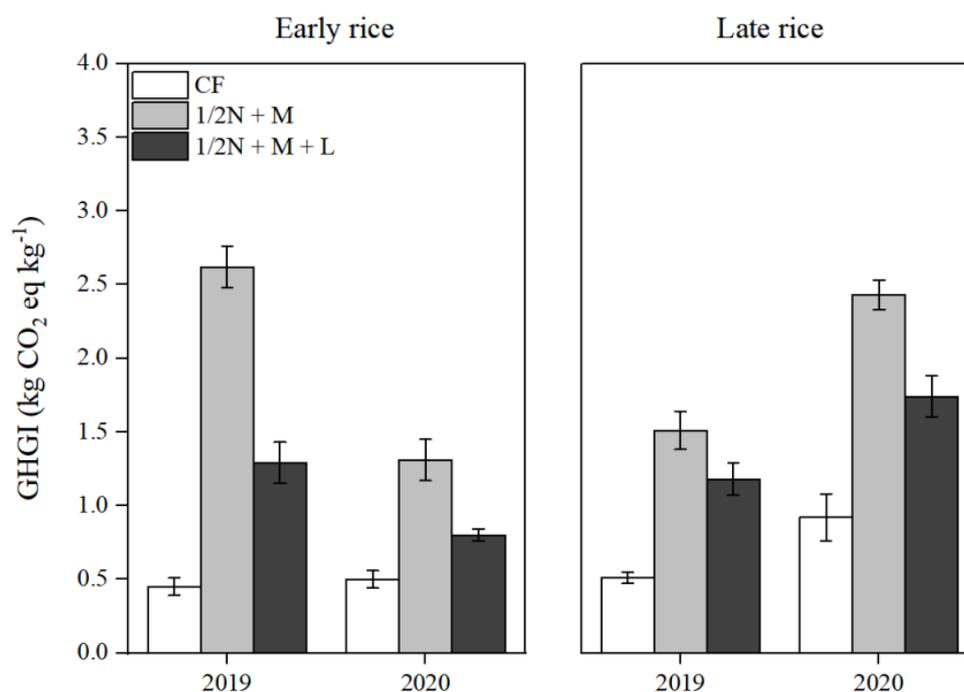


Figure 5. Effects of pig manure substitution and lime application on GHGI over the two years, as there was significant treatment \times crop \times year interaction. CF, 1/2N + M, and 1/2N + M + L represent chemical fertilization, 50% of chemical N substituted by pig manure, and 50% of chemical N substituted by pig manure combined with lime amendment, respectively. Error bars represent the standard deviation of the mean ($n=3$).

3.5. Soil Properties

Averaged across years, 1/2N + M increased the concentration of SOM (+8.9%) compared to the CF treatment, while there was no significance on SOM between 1/2N + M and 1/2N + M + L (Table 2). Relative to the 1/2N + M, 1/2N + M + L increased soil pH (+0.4 unit), whereas there was no significance in soil pH between CF and 1/2N + M (Table 2).

Table 2. Soil pH and soil organic matter (SOM) as affected by treatment and study year at maturity of late rice.

	pH	SOM (g kg ⁻¹)
Treatment (T) ^a		
CF	5.4b	19.0b
1/2N + M	5.5b	20.7a
1/2N + M + L	5.9a	20.4a
Year (Y) ^b		
2019	5.7a	20.1a
2020	5.6a	20.0a
F-values		
T	22.3***	11.7***
Y	NS	NS

CF, 1/2N + M and 1/2N + M + L represent chemical fertilization, 50% of chemical N substituted by pig manure and 50% of chemical N substituted by pig manure combined with lime amendment, respectively. There were no significant interactions between treatment and year (T \times Y) for soil properties. Significant effects are indicated by * (0.01 < P \le 0.05), ** (0.001 < P \le 0.01), or *** (P \le 0.001). Values followed by different lowercase letters are significantly

different among treatments or years. ^a Values were averaged across years. ^b Values were averaged across treatments. ^c Values were averaged across treatments and crops.

4. Discussion

4.1. Effect of Pig Manure Substitution on Rice Yield, CH₄ and N₂O Emissions

Many results indicate that partial substitution of chemical fertilizers by manure can increase the content and availability of soil nutrients, promote soil structure and microbial activity, and subsequently boost rice yield [29–31]. However, the effect on rice yield may depend on the substitution ratio (SR) of manure. Compared to chemical fertilizers, high SR of manure may fail to enhance and even decrease rice yield due to the insufficient N supply during the early crop growth stage [6,32,33]. Contrary to our initial hypothesis, our results showed that the substitution of 50% chemical N by pig manure significantly reduced rice yield compared to full chemical fertilization (Table 1). In the present study, pig manure substituted 50% of the total N fertilizer and was entirely applied as basal fertilizer, and thus no chemical N was applied in the manure-added treatments before rice transplanting. The release of N from pig manure is considerably slower than that from chemical fertilizers [34]. Additionally, the application of organic manure to paddy soils may promote microbial N immobilization [35]. Furthermore, the shorter growth duration of double-cropped rice compared to single rice further restricts N uptake from pig manure [36]. Thus, the high SR of pig manure may lead to insufficient N supply in the early stages of rice growth, thereby inhibiting rice tillering and reduced the number of panicles at maturity (Table S1). In contrast to long-term experiments where manure amendment promotes the build-up of soil nutrients and improves soil fertility [30,37], the present short application duration (only two years) may limit the beneficial effects of pig manure on rice growth [38,39]. Indeed, Zhang et al. proved that the response of crop yield to manure substitution was positively related to the experiment duration and suggested that the high SR of pig manure may contribute to increased crop yield only after long-term application [40]. Therefore, we suggest that the substitution of 50% chemical N by pig manure may be too high to maintain grain yield in the short term within this double rice cropping system.

It is generally acknowledged that organic C input increases CH₄ emissions from paddy fields [8,27,35]. Consistent with previous studies, our results showed that CH₄ emissions were largely increased under the substitution of chemical fertilizers by pig manure (Figure 3). The increase in CH₄ emissions under pig manure amendment was primarily due to the increase in the abundance of methanogens, as organic C addition provides abundant substrates for their activity [41]. Furthermore, the decomposition of pig manure consumes a large amount of oxygen and promotes the decrease of soil oxidation-reduction potential, thereby providing a suitable environment for microbial methanogenesis [42,43].

Previous studies have showed that the combined application of organic manure and reduced chemical fertilization rates can effectively reduce N₂O emissions from upland soils [44,45]. Coupled C and N cycling and soil C accumulation under combined organic and chemical fertilization may contribute to the microbial retention of active N, enhance the abundance of denitrifying bacteria, and promote the conversion of N₂O to N₂, thus reducing the N₂O emissions [46]. Nevertheless, our study indicated that the substitution of chemical fertilizers by pig manure had no significant effect on N₂O emissions from the paddy field compared to the chemical fertilization. Under flooding conditions in paddy fields, N₂O produced by denitrification is primarily reduced to N₂, resulting in lower N₂O emission factors compared to upland soils [47]. We speculated that the negligible N₂O emissions and the large variation of N₂O fluxes in flooded rice-cropping seasons may reduce the ability to detect the treatment effects [48].

4.2. Effect of Liming on Rice Yield, CH₄ and N₂O Emissions under Pig Manure Substitution

Lime application can effectively increase soil pH in paddy fields and alleviate the adverse effect of soil acidification on rice growth [49,50]. Previous studies have proved that liming can not only improve the growth of rice roots but also promote the mineralization of organic matter, thus

increasing nutrient uptake and grain yield [51–53]. In contrast, our results showed that liming did not increase rice yield under pig manure substitution. Firstly, the abundant organic anions in pig manure may react with limes and thus weaken the positive effect of lime application on rice growth [54]. Secondly, as fresh pig manure contains a large deal of NH_4^+ , lime application may promote ammonia volatilization in rice fields [55], thereby leading to an insufficient supply of available N during the critical early growth stages. Indeed, our previous results showed that liming can promote the number of panicles due to the increase in rice N uptake [51], whereas no significant effects were found under pig manure amendment in the present study.

Consistent with previous studies [6,11,12], our results suggested that lime application significantly reduced CH_4 emissions from rice paddies with pig manure amendment. Firstly, lime application can enhance the activity of soil enzymes related to C and N mineralization, promote the decomposition of organic materials, thus reducing the availability of organic C substrates for methane production [23,56]. Secondly, the rise in soil pH decreases the concentration of soil NH_4^+ due to promoted ammonia volatilization in rice fields [55]. The decrease in soil NH_4^+ may instead favor methane oxidation, as there is substrate competition between soil NH_4^+ and CH_4 for methane mono-oxygenase when methanotrophs possess sufficient N sources of their own [41,57]. In addition, the mitigation of CH_4 emissions due to lime application was stronger in the first study year than in the second year in the present study (Figure 3). This can be attributed to the depletion of OH^- in limes with the extension of application duration, resulting in a reduced alkaline effect [50]. Correspondingly, our results also showed that the increase in soil pH under lime application was lower in the second study year than in the first year (Table 2). Consequently, the promotion effect of lime on the mineralization of organic matter in pig manure may also be weakened [58]. Meanwhile, our results showed that the reduction in seasonal CH_4 emissions due to lime application was greater in the early rice season than in the late rice season, particularly in the first year (Figure 3). As limes were only applied once before the transplanting of early rice, the alkaline effect of limes was greatest in the early season of the first year and then gradually weakened as discussed above.

Previous studies have reported varying effects of liming on N_2O emissions from acidic soils, ranging from positive to negative, and even neutral outcomes [23,59,60]. On the one hand, liming in acidic soils may favor ammonia oxidizing bacteria (AOB) over ammonia oxidizing archaea (AOA), ultimately increasing N_2O emission due to the higher potential for N_2O per mole NH_3 oxidized of AOB [17,61,62]. On the other hand, lime application could decrease N_2O emissions by increasing the abundance of genes encoding nitrite reductase (*nirK*) and nitrous oxide reductase (*nosZ*), thereby limiting N_2O emissions [60,63,64]. Many studies have proved a negative correlation between soil pH and the ratio of denitrification products ($\text{N}_2\text{O}/(\text{N}_2+\text{N}_2\text{O})$) within the pH range of 5 to 8 [65–68]. Thus, the rise in soil pH due to lime application is supposed to reduce N_2O emissions. However, several studies also reported that lime application did not significantly affect N_2O emissions, possibly due to the conflicting effect of liming on N_2O production and consumption [23,66,67]. Our results also showed that liming had no significant effect on N_2O emissions from the paddy field, but the underlying mechanisms remain unclear.

Consistent with previous results, the present study showed that CH_4 emissions contributed to most of the GWP in the double rice cropping system [6,69]. As lime application reduced CH_4 emissions without significant effect on N_2O emission or grain yield, liming reduced the GWP and GHGI induced by the substitution of chemical N by pig manure. Furthermore, we expect that long-term addition of organic manure under lime application may further decrease GWP and GHGI through soil C sequestration [27]. However, appropriate substitution ratios of chemical fertilizers by pig manure should be determined to maintain rice yield without compromising food security [32]. Meanwhile, as the liming effect on raising soil pH weakens gradually, its persistent effect on CH_4 emission mitigation needs further examination in the future [50].

5. Conclusions

Compared to chemical fertilization, the substitution of pig manure with 50% chemical N reduced rice yield, while significantly promoted CH_4 emissions. Neither pig manure substitution nor liming

significantly affected N₂O emissions from the paddy field. Lime application had no significant effect on rice yield but significantly mitigated CH₄ emissions induced by pig manure addition, and thus reduced the GWP and GHGI. Consequently, combining pig manure substitution with lime application is effective to not only alleviate soil acidification and improve soil fertility in the long term, but also mitigate the greenhouse effect induced by manure addition in rice paddies with acidic soils.

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