AGRICULTURE

Acclimation of methane emissions from rice paddy fields to straw addition

Yu Jiang[1,2], Haoyu Qian[1], Shan Huang[3], Xingyue Zhang[4], Ling Wang[5], Li Zhang[6], Mingxing Shen[7], Xiaoping Xiao[8], Fu Chen[9], Hailin Zhang[9], Changying Lu[7], Chao Li[8], Jun Zhang[1], Aixing Deng[1], Kees Jan van Groenigen[2]*, Weijian Zhang[1]*

Straw incorporation is a common long-term practice to improve soil fertility in croplands worldwide. However, straw amendments often increase methane (CH_4) emissions from rice paddies, one of the main sources of anthropogenic CH_4. Intergovernmental Panel on Climate Change (IPCC) methodologies to estimate CH_4 emissions from rice agriculture assume that the effect of straw addition remains constant over time. Here, we show through a series of experiments and meta-analysis that these CH_4 emissions acclimate. Effects of long-term (>5 years) straw application on CH_4 emissions were, on average, 48% lower than IPCC estimates. Long-term straw incorporation increased soil methanotrophic abundance and rice root size, suggesting an increase in CH_4 oxidation rates through improved O_2 transport into the rhizosphere. Our results suggest that recent model projections may have overestimated CH_4 emissions from rice agriculture and that CH_4 emission estimates can be improved by considering the duration of straw incorporation and other management practices.

INTRODUCTION

Rice is a staple food for almost half of all people, and global rice demand is projected to increase by 28% in 2050 (*1*, *2*). Yet, rice yields have stagnated in 35% of all rice-growing regions (*3*). Rice yield improvement has been limited largely by the depletion of soil fertility due to prolonged agricultural activity (*3*, *4*). Straw incorporation is a common management practice to improve soil fertility and boost crop yields (*5*, *6*). However, straw additions to rice paddies increase emissions of the potent greenhouse gas methane (CH_4) (*5*, *7*). This is important because rice paddies are a main source of CH_4; rice agriculture is responsible for approximately 11% of global anthropogenic CH_4 emissions (*8*), and rice has the highest greenhouse gas intensity among the main food crops (*9*, *10*).

Methane emissions from rice paddies are determined by the balance between CH_4 production and oxidation (*7*). Methane is produced by methanogens under anaerobic conditions, and its production is affected by C availability (*7*). In flooded rice paddies, straw incorporation usually stimulates CH_4 production (*11*, *12*). This increase in CH_4 production may, in turn, stimulate methanotrophic growth and CH_4 oxidation (*13*, *14*), but methanotrophic growth can be limited by low O_2 concentrations (*7*, *15*). Because larger rice plants stimulate O_2 transport into the rhizosphere, management practices can affect CH_4 oxidation rates through their effect on plant growth (*16*, *17*). Thus, the net effect of straw incorporation on CH_4 emissions likely depends on several mechanisms that may operate at different time scales, making long-term predictions of CH_4 emissions challenging.

Intergovernmental Panel on Climate Change (IPCC) methodologies to estimate the effect of straw amendments on regional and global CH_4 emissions use straw application rate as the main predictor (*18*, *19*). Similarly, the CH4MOD model (*20*, *21*), a widely used model to predict CH_4 emissions from rice agriculture (*19*, *22*), estimates effects of straw incorporation on CH_4 emissions from straw application rate and straw type (Supplementary Text). Both methodologies assume that the duration of straw incorporation does not affect CH_4 emissions, but this assumption has never been thoroughly tested.

Here, we conducted three complementary experiments to determine the effects of straw additions on CH_4 emissions. First, in two long-term field experiments, we measured the effect of straw incorporation on CH_4 emissions over time in two of China's most common rice cropping systems, that is, double rice and middle rice (see Materials and Methods). Second, using soils from the same field experiments, we grew rice in pots and measured the effect of straw incorporation on rice biomass and the abundance of methanogenic and methanotrophic microbes. Third, to test for effects of straw incorporation that are not mediated through its effect on rice growth, we conducted an incubation experiment without rice plants, again using soils of the same field experiments. Last, to test the generality of our findings, we conducted a meta-analysis to quantify the effect of straw incorporation on CH_4 emissions.

RESULTS

Straw incorporation increased CH_4 emissions in both field experiments (Fig. 1), confirming numerous previous studies (*5*, *7*, *11*, *12*). However, we also found a straw × year interaction, whereby straw application increased CH_4 emissions less strongly in the later years of the experiments. In the double rice system, straw incorporation increased CH_4 emissions in year 2 of the experiment by 266%, but only by 87% in year 13 (Fig. 1A and fig. S1A). In the middle rice system, methane emissions in year 13 were higher than in year 8, but only in plots without straw application (Fig. 1B and fig. S1B). The average daily temperature was higher in year 13 than in year 8 in the middle rice system (fig. S2),

---

[1]Institute of Crop Sciences, Chinese Academy of Agricultural Sciences, Ministry of Agriculture, Beijing 100081, China. [2]Department of Geography, College of Life and Environmental Sciences, University of Exeter, Exeter EX4 4 RJ, UK. [3]Ministry of Education and Jiangxi Key Laboratory of Crop Physiology, Ecology and Genetic Breeding, Jiangxi Agricultural University, Nanchang 330045, China. [4]Department of Land, Air and Water Resources, University of California, Davis, Davis, CA 95616, USA. [5]School of Biotechnology, Jiangsu University of Science and Technology, Zhenjiang 212018, China. [6]Institute of Applied Ecology, Nanjing Agricultural University, Nanjing 210095, China. [7]Institute of Agricultural Sciences in Taihu Lake District, Jiangsu Academy of Agricultural Sciences, Suzhou 215100, China. [8]Institute of Soil and Fertilizer, Hunan Academy of Agricultural Sciences, Changsha 410125, China. [9]College of Agronomy and Biotechnology, Key Laboratory of Farming System, Ministry of Agriculture of China, China Agricultural University, Beijing 100193, China.
*Corresponding author. Email: zhangweijian@caas.cn (W.Z.); kj.vangroenigen@exeter.ac.uk (K.J.V.G.)

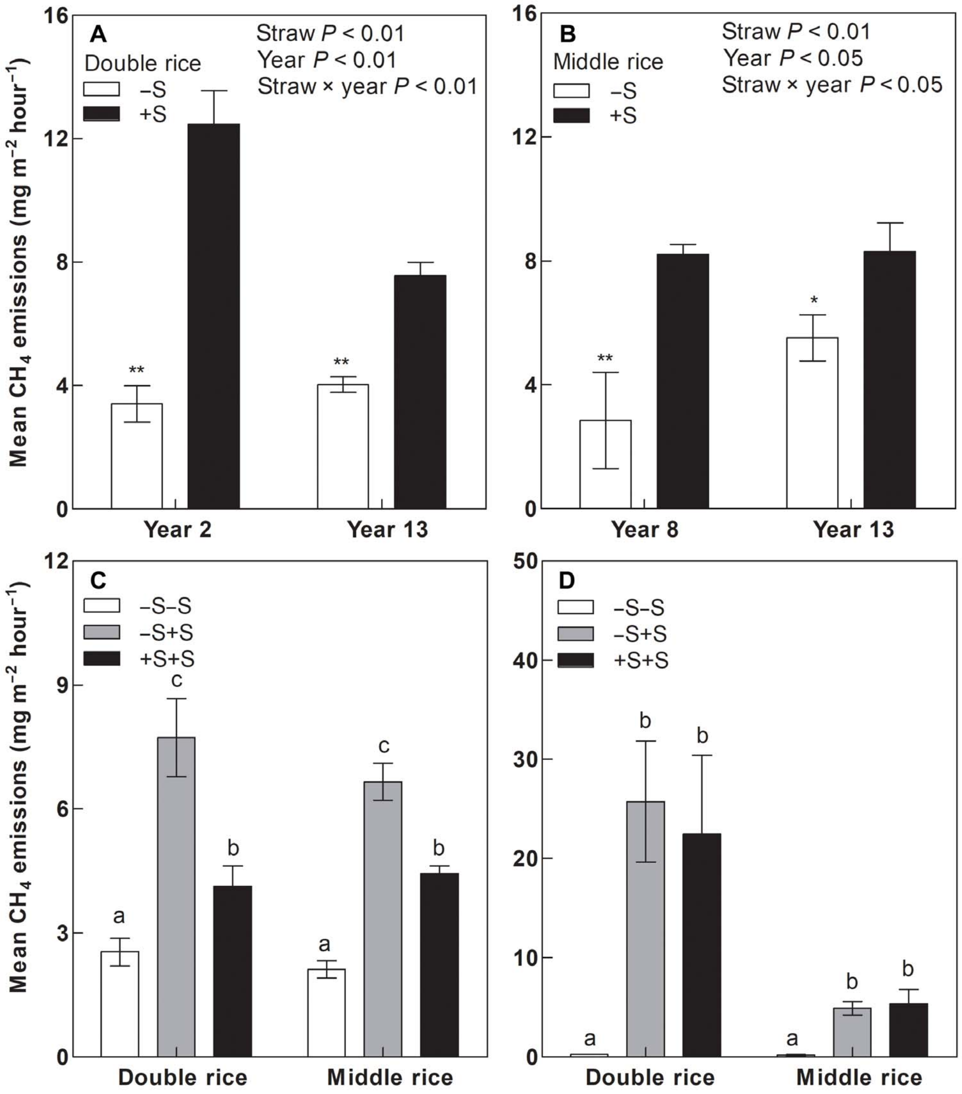

Fig. 1. CH_4 emissions from two rice cropping systems, as affected by straw incorporation and experimental duration. Results are shown for field experiments in a double rice system (**A**) and a middle rice system (**B**), and for a pot experiment (**C**) and an incubation experiment (**D**) using soils from both systems. −S, no straw incorporation in the field; +S, straw incorporation in the field; −S−S, no straw addition to −S soils; −S+S, straw addition to −S soils; +S+S, straw addition to +S soils. "*" and "**" denote significant effects of straw addition within the year at $P < 0.05$ and $P < 0.01$, respectively. Different characters represent significant differences ($P < 0.05$). Error bars represent SD ($n = 3$).

possibly explaining the higher CH_4 emissions in plots without straw application in year 13. Straw incorporation increased CH_4 emissions by 186% in year 8 of the experiment, but only by 50% in year 13 (Fig. 1B and fig. S1B). Similarly, straw incorporation in the pot experiment stimulated CH_4 emissions from both systems, but treatment effects were two to three times smaller for soils that had previously received straw in the field (Fig. 1C and fig. S3). In the incubation experiment, straw addition also increased CH_4 emissions in both systems, but the effect was similar for soils with and without a history of straw incorporation (Fig. 1D and fig. S4).

In our meta-analysis, straw incorporation increased CH_4 emission more strongly in short-term experiments (≤5 years) than in long-term experiments (>5 years; Fig. 2A), although the average straw application rate in short-term experiments [4.6 metric tons (MT) ha^{-1}] was lower than in long-term experiments (5.2 MT ha^{-1}; dataset S1). Experimental duration accounted for more of the variation in responses of CH_4 emission to straw addition than a wide range of other environmental and experimental factors (fig. S5A).

Long-term straw addition in the field increased the abundance of both methanogens and methanotrophs (table S1). However, in the pot experiment, straw addition removed the initial differences in the abundance of methanogens between soils with and without a history of straw incorporation at 4 and 9 weeks after rice transplanting (Fig. 3, A and B). In contrast, straw incorporation increased methanotrophs more strongly in soils with a history of straw incorporation than in soils without such a history (Fig. 3, C and D).

The effects of straw incorporation on rice growth increased with experimental duration. In the double rice system, straw incorporation

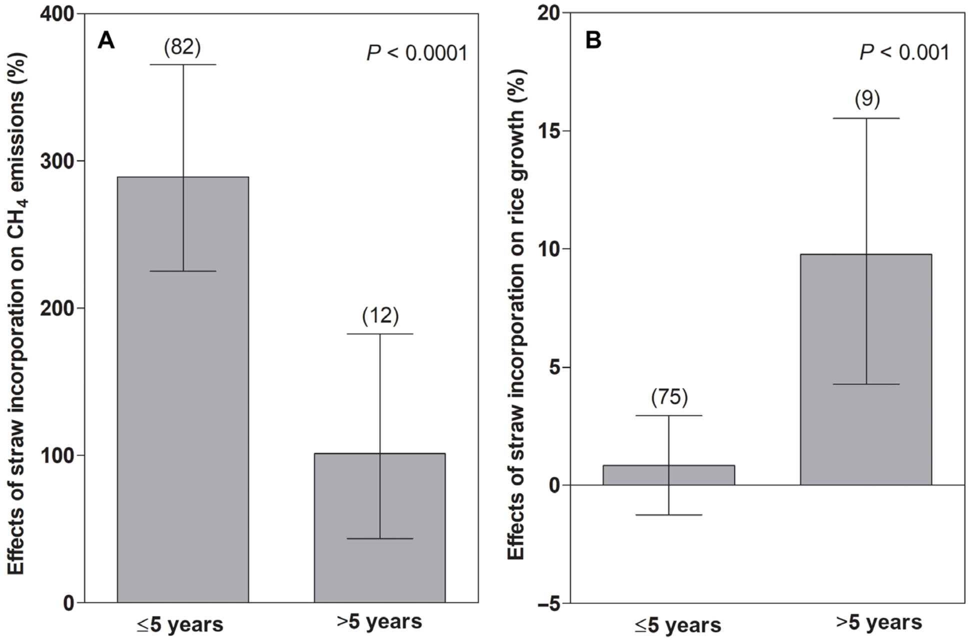

Fig. 2. Results of a meta-analysis on the effect of straw incorporation on CH_4 emissions and rice growth in short-term (≤5 years) and long-term (>5 years) experiments. Results for CH_4 emissions (**A**); results for rice growth (**B**). The total number of observations included in each category is displayed in parentheses. Error bars indicate 95% confidence intervals.

did not affect rice biomass in year 2, but it significantly increased rice biomass in year 13 (Fig. 4A). Similarly, in the middle rice system, straw application did not affect rice biomass in year 8, but it significantly increased rice biomass in year 13 (Fig. 4B). In the pot experiment, straw addition to −S soils tended to reduce the root biomass and aboveground biomass, whereas straw addition to +S soils increased root biomass and aboveground biomass (Fig. 4, C and D). In our meta-analysis, whereas straw incorporation did not affect rice growth in short-term experiments, it increased rice growth in long-term experiments (Fig. 2B). As with CH_4 emissions, experimental duration explained more of the variation in responses of plant growth to straw addition than a wide range of other environmental and experimental factors (fig. S5B).

DISCUSSION

Our field experiments, our pot experiment and our meta-analysis consistently indicate that the effect of straw incorporation on CH_4 emissions decreases over time. These results suggest that short-term studies overestimate the effects of straw incorporation of seasonal CH_4 emissions in real-world rice cropping systems, where straw addition is typically added for many years (*5*, *6*).

Our results can be explained by a difference in the temporal response of methanogens and methanotrophs to straw addition. Because methanogens are typically limited by C availability (*7*, *11*), methanogenic growth and CH_4 production increase quickly after the implementation of straw incorporation practices (*11*, *12*). This quick response was confirmed by our pot experiment, where straw addition increased methanogens to the same extent in soils with or without a history of straw incorporation. In contrast, straw addition stimulated methanotrophic growth more strongly in soils that previously received straw, indicating an increase in the response of CH_4 oxidation rates over time. We propose three different explanations for our findings. First, the effect of straw incorporation on rice plant growth increases over time in our meta-analysis, our field experiments, and our pot experiment. Because larger rice plants stimulate O_2 transport into the rhizosphere, they can stimulate methanotrophic growth (*16*, *17*). Straw incorporation likely stimulated plant growth by increasing soil N and P availability, as indicated by higher total N and available P concentrations in both field experiments (table S1). Our finding of increased plant growth corroborates a recent comprehensive meta-analysis on straw-induced changes in rice plant growth (*6*). Second, improved soil fertility with long-term straw incorporation can stimulate algal growth, which, in turn, increases dissolved O_2 concentrations of floodwater (*23*). Third, high soil N availability may directly stimulate methanotrophic growth under high CH_4 concentrations (*24*). Yet, our incubation experiment indicates that the time-dependent effect of straw incorporation does not occur in the absence of rice plants, suggesting that long-term straw incorporation facilitated methanotrophic growth mainly by stimulating plant growth and O_2 transport.

To compare our results directly to IPCC predictions, we plotted both observed and predicted effects of straw incorporation on CH_4 emissions against experimental duration (See Materials and Methods). Because straw application rates increased with experimental duration (fig. S6), IPCC predictions of straw incorporation effects on CH_4 emissions increased as well. However, observed effects of straw incorporation decreased with experimental duration (Fig. 5). On average, the IPCC Tier 1 methodology estimated a 193% increase in CH_4 emissions due to long-term straw incorporation for the studies in our dataset. Yet, long-term straw incorporation stimulated the CH_4 emissions by only 101% (Fig. 2A), i.e., a positive effect that is almost twice as small as the IPCC estimates. Thus, our results suggest that the IPCC methodologies overestimate long-term CH_4 emissions from rice paddies with straw addition. These overestimates likely reflect that the IPCC

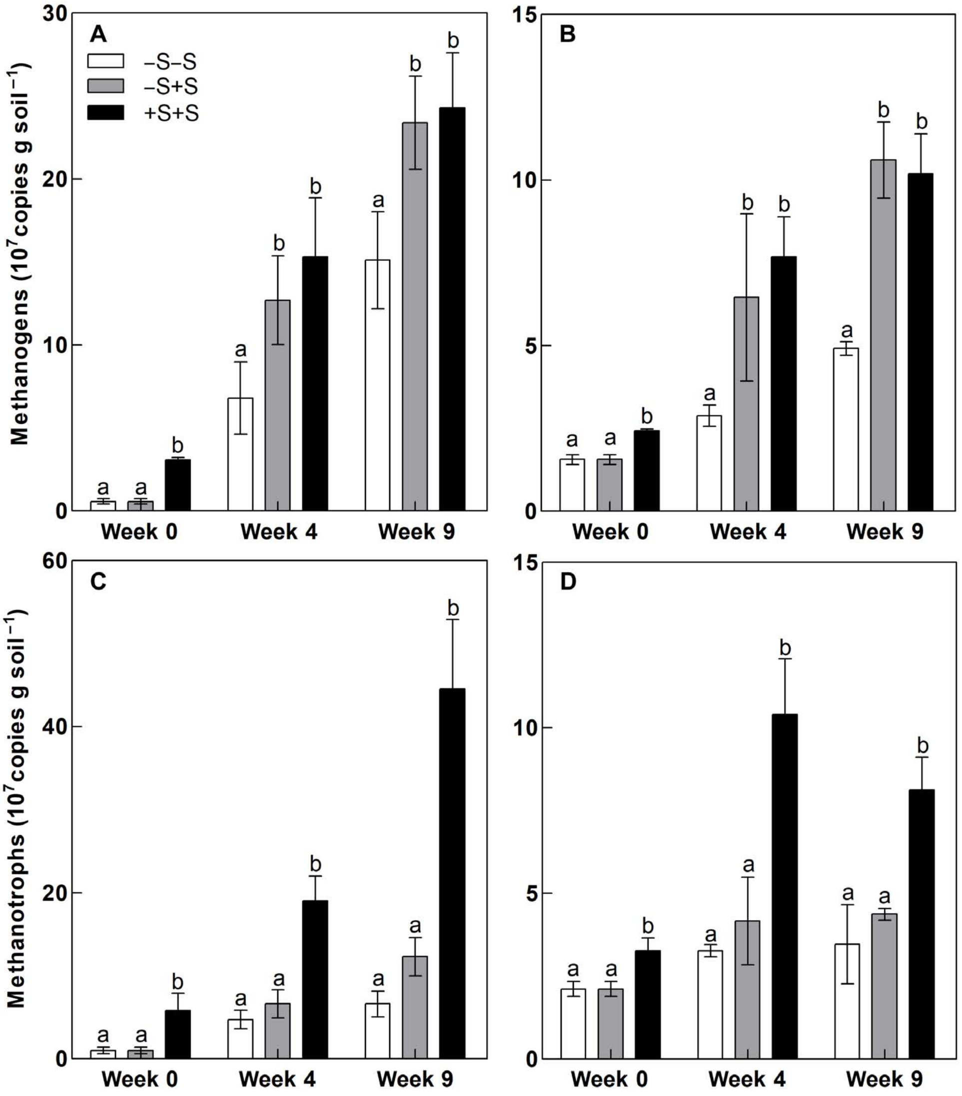

Fig. 3. The abundance of methanogens and methanotrophs as affected by straw incorporation. Results are from a pot experiment on soils of a double rice (**A** and **C**) and a middle rice system (**B** and **D**). −S−S, no straw addition to −S soils; −S+S, straw addition to −S soils; +S+S, straw addition to +S soils. Different characters represent significant differences ($P < 0.05$). Error bars represent SD ($n = 3$).

methodology is informed by short-term experiments (*18*), which typically show stronger treatment effects (Figs. 1, 2, and 5). To improve the accuracy of CH_4 emission estimates, we suggest that the IPCC methodologies add duration into the conversion factor for straw incorporation.

Similarly, the CH4MOD model suggests that because of high crop straw retention, CH_4 emissions from China's rice paddies gradually increased by 65.8% from the 1970s to the 2000s (*22*). In China, straw is incorporated in about 40% of cropland, and this number is widely expected to increase (*25*), suggesting further increases in CH_4 emissions from China's rice paddies (*22*). The CH4MOD model, which was also calibrated using a short-term experiment (*26*), assumes that CH_4 oxidation rates throughout the growing season correlates with relative biomass (that is, the ratio of rice aboveground biomass to maximum aboveground biomass of growing season; Supplementary Text) (*20*, *21*). Yet, our results and recent studies show that absolute biomass and CH_4 concentrations significantly affected the abundance of methanotrophs and CH_4 oxidation rate (*14*, *16*, *17*). Because long-term straw incorporation stimulates absolute plant biomass, the CH4MOD model likely underestimates CH_4 oxidation rates and overestimates CH_4 emission from rice paddies with straw incorporation. The CH4MOD model could possibly be improved by considering the factors affecting the CH_4 oxidation rates, such as CH_4 production and absolute biomass.

Our study focuses solely on the effects of straw incorporation. However, other types of organic matter amendment such as manure, cover crop, and compost also gradually increase rice plant growth and soil fertility (*27*), suggesting that positive effects of these amendments on CH_4 emissions (*7*, *27*) may decrease over time as well. Temporal variation in crop growth responses to management practices should be considered when estimating CH_4 emissions from rice agriculture.

Our experiments and meta-analysis, the IPCC methodologies, and the CH4MOD model all focus on CH_4 emissions during the growing

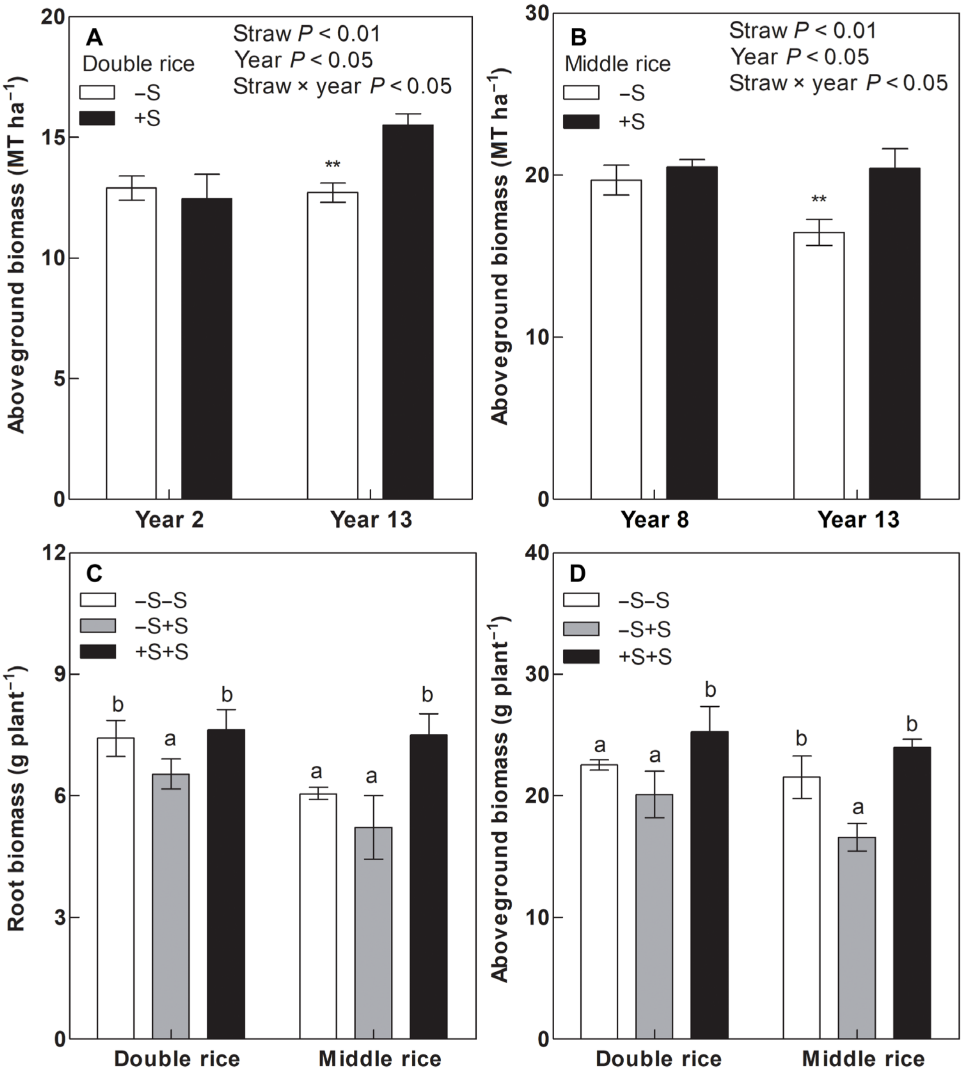

Fig. 4. Aboveground biomass and root biomass as affected by straw incorporation. Aboveground biomass in a double rice system (**A**) and a middle rice system (**B**). −S, no straw incorporation; +S, straw incorporation. "**" denotes significant effects of straw addition within the year at $P < 0.01$. Root biomass (**C**) and aboveground biomass (**D**) in a pot experiment. −S−S, no straw addition to −S soils in pots; −S+S, straw addition to −S soils in pots; +S+S, straw addition to +S soils in pots. Different characters represent significant differences within rice system ($P < 0.05$). Error bars in all panels represent SD ($n = 3$).

season. However, CH_4 emission during the fallow season can be substantial in rice systems where paddies remain flooded year-round (*28*). Under these conditions, annual CH_4 emission with straw addition may acclimate to a smaller extent because the mechanism responsible for acclimation is contingent on the presence of rice plants. Moreover, because long-term straw addition stimulates rice growth and straw production, it may even increase off-season CH_4 emissions in these systems. However, approximately 90% of paddies with long fallow periods are drained during fallow season (*29*), minimizing CH_4 emissions during this period. Thus, our results that CH_4 emissions with straw addition acclimate over time are likely representative for the vast majority of rice systems.

Besides its direct effect on CH_4 emissions, straw incorporation affects numerous other aspects of rice paddies, many of which affect the overall greenhouse gas budget of rice cropping systems. Long-term straw incorporation enhances soil C stocks, which may further offset its positive effects on CH_4 emissions (*5*). For instance, straw additions increased the soil organic carbon (SOC) content by 20 to 30% in our field experiments (table S1). Because straw incorporation tends to increase soil fertility (*5*, *6*), it may reduce the need for fertilizer input to sustain rice productivity, thereby reducing indirect C emissions from fertilizer production and transport (*30*). Other aspects of rice agriculture that are not related to greenhouse gas emissions should not be ignored either. For instance, compared with open-field burning, straw incorporation leads to favorable impacts on human health and air quality (*31*). Straw incorporation can benefit soil biota growth, which will enhance soil biodiversity and health (*32*). Last, long-term straw incorporation usually increases rice yields (*5*, *6*), indicating that straw amendments may contribute to improved food security. Agricultural policy decisions involving straw management need to consider numerous trade-offs between positive and negative impacts of straw incorporation on rice production, greenhouse gas emissions, and the

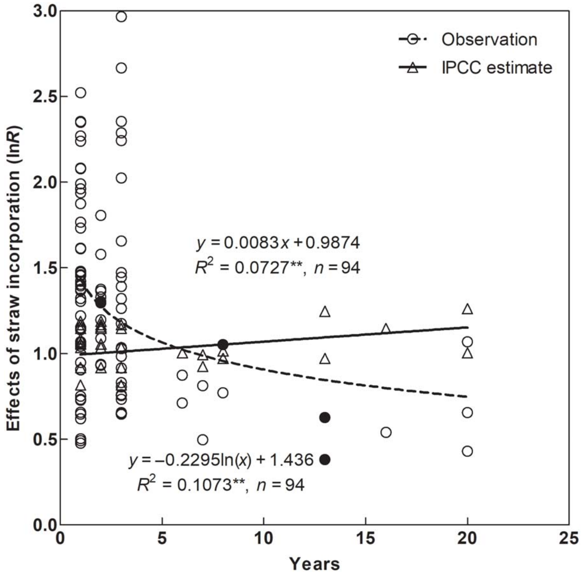

Fig. 5. Comparison between observed effects of straw incorporation on CH_4 emissions from rice paddies and IPCC estimates. Both the observed effects and IPCC estimates are plotted against experimental duration. Results are based on 94 observations; observations from our field experiments are indicated by black circles. IPCC estimates are based on 94 matching observations of straw incorporation rates. "**" denotes significance at $P < 0.01$.

environment. Our findings suggest that, within this context, the positive effects on CH_4 emissions are smaller than previously assumed.

MATERIALS AND METHODS

Field experiments

In 2005, we established two long-term experiments to study the effects of straw addition on crop growth and CH_4 emissions from rice paddies. One experiment was established in a double rice cropping system at Changsha (28.1°N, 112.3°E) and the other experiment in a middle rice system (rice-wheat rotation) system at Suzhou (31.9°N, 120.6°E). Together, these experiments represent two major Chinese rice production areas and cropping systems. The Changsha site is characterized by a subtropical monsoonal humid climate, with a mean annual temperature of 16.8°C and a mean annual precipitation of 1358 mm. The Suzhou site has a northern subtropical monsoon climate with a mean annual temperature of 15.7°C and a mean annual precipitation of 1094 mm. The initial soil properties in the double rice system (0 to 10 cm) and middle rice system (0 to 20 cm) are reported in table S1.

Both field experiments included two treatments: straw incorporation (+S) and no straw incorporation (−S). Each experiment had a completely randomized design with three replicates for each treatment. Plots in the double rice system were 66.7 m^2 (8.34 m by 8.0 m), and plots in the middle rice system were 26 m^2 (4.0 m by 6.5 m). In the +S treatments at both sites, all straw was chopped into about 10-cm-long pieces. In the double rice system, after early rice harvest, all rice straw was incorporated into the soil (0- to 15-cm depth) by plowing. After the late rice harvest, all straw was left on the soil surface during the fallow season and was incorporated into the soil by plowing before transplanting the early rice seedlings. In the middle rice system, after rice and wheat harvest, all rice or wheat straw was incorporated into the soil (0- to 15-cm depth) by plowing. In the −S treatments, all aboveground rice straw was removed in the double rice system; in the middle rice system, residual stubbles (20 cm) were left in all plots.

In the double rice system, early rice and late rice were transplanted in middle April and early July and were harvested in early July and middle October, respectively. Fertilizer was applied at 150 kg N ha^{-1} as urea, 45 kg P_2O_5 ha^{-1}, and 52 kg K_2O ha^{-1} for both the early and late growing seasons. In the middle rice system, rice seedlings were transplanted in middle June and harvested in late October. Wheat was seeded in early November and harvested in late May. All plots received 225 kg N ha^{-1} as urea and 150 kg K_2O ha^{-1} during the rice season and 225 kg N ha^{-1} and 90 kg K_2O ha^{-1} during the wheat season. At both sites, water regimes consisted of early flooding, followed by midseason drainage, followed by intermittent irrigation in all plots. At the double rice site, we grew an Indica rice cultivar (*Oryza sativa L. cv. Yizao 9*) in the early rice season and an Indica rice cultivar (*Oryza sativa L. cv. Xiangwanxian 13*) in the late season in all years. At the middle rice site, we grew a Japonica rice cultivar (*Oryza sativa L. cv. Suxiangjing 100*). All other management practices followed local recommendations.

We measured CH_4 emissions from rice paddies during the late rice season of 2006 and 2017 in the double rice system (i.e., 2 and 13 years of straw incorporation) and in 2012 and 2017 in the middle rice system (i.e., 8 and 13 years of straw incorporation). Because we focused on the effects of straw incorporation within the same rice season on CH_4 emissions, we only measured the CH_4 emissions during the late rice season in the double rice system. In the double rice system, straw application rate in year 13 (7.3 MT ha^{-1}) was higher than in year 2 (6.1 MT ha^{-1}). In the middle rice system, the application rate of straw was about 4.2 MT ha^{-1} in years 8 and 13. In the double rice system, mean air temperatures during the growth season of 2006 (26.9°C) and 2017 (26.8°C) were similar (fig. S2A). In the middle rice system, the average temperature in 2017 (26.7°C) was higher than in 2012 (25.8°C), especially early in the growing season when CH_4 emissions were high (fig. S2B).

Pot experiment

The pot experiment with three replicates was conducted under open-field conditions at the Chinese Academy of Agricultural Science, Beijing (40.0°N, 116.3°E), China. In early June 2017, before straw was incorporated, soils were collected from both field sites described above. At both sites, we collected four soil samples (0 to 20 cm) from each of the six plots by shovel. Soil properties are shown in table S2. Samples were combined per plot, air-dried, and sieved (6-mm mesh size) to remove stones.

Plastic pots (height, 20 cm; diameter, 20 cm) were filled with 5.0 kg of dry soil. A nylon mesh bag (diameter, 8 cm; height, 10 cm; mesh size, 37 μm) was placed in the center of each pot to create two soil compartments: the central rooted compartment and the outside nonrooted compartment. Fresh wheat straw (2 to 5 mm) was incorporated into the soil at a rate equivalent to 6 MT ha^{-1}. Thus, straw incorporation in pots with −S soils (−S+S) mimicked the 1st year of straw incorporation, whereas straw incorporation in the pots with +S soils (+S+S) mimicked the 13th year of straw incorporation. We included pots with −S soil without straw incorporation (−S−S) as the control treatment. Two healthy rice seedlings were transplanted into the root bag in early July. Fertilizers N, P, and K were applied at 225 kg N ha^{-1}, 150 kg P_2O_5 ha^{-1}, and 150 kg K_2O ha^{-1}, respectively, in each pot. A water layer of 2 to 3 cm was kept during the rice growth period. We finished the experiment 9 weeks after rice transplanting; approximately 90% of total seasonal CH_4 emissions was produced during this period (fig. S1).

Incubation experiment

We conducted an incubation experiment with three replicates using the same soils as in the field and pot experiments. As with the pot experiment, the incubation experiment consisted of three treatments: −S soils without straw, −S soils with straw, and +S soils with straw. In the straw addition treatments, fresh wheat straw (2 to 5 mm) was mixed with soils at a rate equivalent to 6 MT ha^{-1}. We added 15 g of soil with or without straw to 40-ml beakers and kept a water layer of 2 to 3 cm. Soils were incubated at 30°C in the dark for 4 weeks.

Sampling and measurement methods

In the field and pot experiments, we measured CH_4 emissions at 7-day intervals by the static closed-chamber method (*27*). We measured CH_4 emissions from 1 or 2 weeks after rice transplanting to harvest. On each sampling day, we collected four gas samples at 5- or 10-min intervals for each plot or pot. Gas samples were taken between 9:00 and 11:00 a.m. The CH_4 concentrations were determined by a gas chromatograph (Agilent 7890A, USA). Emission rates of CH_4 (F) were calculated as follows

$$F = \Delta C/\Delta T \times V/A$$

where $\Delta C/\Delta T$ is the change in CH_4 concentration (mg liter^{-1} hour^{-1}) in the chamber determined by linear regression, V is the volume of the chamber (liter), and A is the enclosed surface area (m^2). For our flux rate estimates, we only accepted measurements for which $r^2 > 0.90$; less than 5% of the measurements were discarded.

In the incubation experiment, we measured CH_4 emissions at 3-day intervals. We put the beakers into 500-ml jars and sealed the jars. We collected 10 ml of gas from the jar at 0 and 24 hours after sealing the jar. Emission rates of CH_4 (F) were calculated as follows

$$F = \Delta C/\Delta T \times V/S$$

where $\Delta C/\Delta T$ is the change in CH_4 concentration (μg liter^{-1} hour^{-1}) in the jar, V is the volume of the jar (liter), and S is the soil weight (g).

In the field experiment, we measured the abundances of methanogens and methanotrophs of air-dried soils after 12 years of straw incorporation. In the pot experiment, we collected fresh soil samples from the root bags 4 weeks after rice transplanting, when CH_4 emissions were relatively high and significantly different among treatments, and 9 weeks after rice transplanting at the end of the experiment. We used the Power Soil DNA Isolation kit (MoBio, USA) to extract soil DNA from 0.25 g of soil. The copy numbers of *mcrA* and *pmoA* genes represent the abundances of methanogens and methanotrophs, respectively, and were quantified using quantitative real-time polymerase chain reaction (PCR). We used the primer pairs mcrAf/mcrAr and A189f/A682r to quantify the *mcrA* and *pmoA* genes, respectively (*33*, *34*). We performed the quantitative real-time PCR in CFX96 (Bio-Rad, USA).

In the field experiment, rice yield was measured at maturity stage, and aboveground biomass was calculated by rice yield and harvest index. In the pot experiment, we harvested plants and measured aboveground biomass and root biomass 9 weeks after rice transplanting. Rice plants were oven-dried at 70°C to achieve a constant weight.

Statistical analysis

We analyzed the data on CH_4 emissions, biomass, and soil properties from the field experiment by two-way analysis of variance (ANOVA) (that is, straw and year) and/or independent sample t test. A one-way ANOVA was used to analyze the data from the pot and incubation experiments. Differences between treatments were analyzed by using the least significant difference test. All analyses were performed with the statistical package SPSS 18.0. Differences between treatments were considered significant at $P < 0.05$.

Meta-analysis

We collected peer-reviewed papers on straw incorporation and CH_4 emission from the China National Knowledge Infrastructure and Web of Science. These peer-reviewed papers were published in Chinese or English before October 2017. The studies had to meet the following criteria to be included in our dataset: (i) The experiment was conducted under field conditions with replicates; (ii) experimental duration was clearly stated; (iii) all management practices besides straw addition (e.g., N fertilizer rate and water management) needed to be the same between the treatment and control; (iv) crop straw was incorporated into the soils within the same rice season (i.e., less than 30 days before rice transplanting); (v) the application rate of straw was between 3 and 7.5 MT ha^{-1}; and (vi) the rice paddies were under flooded conditions before the jointing stage. Criteria 4 to 6 were applied to ensure that all experiments in our dataset were representative of real-world conditions (*18*, *21*, *22*) and that results could be compared to IPCC estimates.

In total, 24 published papers including 94 observations met our criteria (table S2 and dataset S1). For each study, we tabulated rice growth data (that is, aboveground biomass or rice yield) if these were available. If a paper reported both data of aboveground biomass and rice yield, then we used the data of aboveground biomass. For each experiment in dataset S1, we quantified the effects of straw incorporation on CH_4 emissions through calculating the natural logarithm of the response ratio (R)

$$\ln R = \ln(xs/xc)$$

where xs and xc are the values of the variables (CH_4 emissions and rice growth) for the treatment with and without straw incorporation, respectively (*35*). We weighted ln$R$ by the inverse of its variance and estimated missing variances using the average coefficient of variation across our dataset (*36*).

Several factors are known to affect CH_4 emissions from rice paddies (*7*). To test whether these factors affected straw addition responses, we extracted the following information for each study in our dataset: SOC (gram per kilogram), mean annual temperature (degree Celsius), water management (continuous flooding, midseason drainage, or intermittent irrigation), rice cultivar (Japonica or Indica), inorganic N application rate (kilogram per hectare), cropping system (single rice, rice wheat, or double rice), straw application rate (metric ton per hectare), straw type (rice or wheat), and experimental duration (≤5 or >5 years). We used the "glmulti" package in R to determine the relative importance of the experimental factors in affecting treatment effects, analyzing our data with all possible models that could be constructed using combinations of the experimental factors (*36*–*38*). The relative importance of the experimental factors was calculated as the sum of Akaike weights derived for all the models in which the factor occurred, where the Akaike weights represent the relative likelihood of a model. On the basis of the outcome of the model selection procedure, we used a Wald test to determine whether treatment effects were statistically different between experimental classes.

We used the rma.mv function in the "metafor" package (*39*) to perform a mixed-effects meta-analysis in R, including "paper" as a random effect because several papers contributed more than one effect size. To ease interpretation, the results of ln$R$ were back-transformed and reported as the percentage change [$(R - 1) \times 100$]. We used the logarithmic function of the statistical package SPSS 18.0 to describe correlation between treatment effects and durations in our dataset.

IPCC methodology

According to the IPCC Tier 1 methodology, the effect of straw incorporation on CH_4 emissions (S) is estimated as follows

$$S = (1 + \mathrm{ROA} \times \mathrm{CFOA})^{0.59}$$

where S is the scaling factor (i.e., emissions in straw amended plots divided by emissions in nonamended plots), ROA is the application rate of straw, and CFOA is a conversion factor for straw incorporation (*19*). Because straw was incorporated into soils within the same rice season in our dataset, the value of CFOA is 1. Note that the scaling factor S is directly comparable to the response ratio R in the meta-analysis.

SUPPLEMENTARY MATERIALS

Supplementary material for this article is available at http://advances.sciencemag.org/cgi/content/full/5/1/eaau9038/DC1

Supplementary Text

Fig. S1. CH_4 emissions from two rice cropping systems, as affected by straw incorporation and experimental duration.

Fig. S2. Daily average air temperature during the rice growing season.

Fig. S3. CH_4 emissions from two rice cropping systems, as affected by straw incorporation.

Fig. S4. CH_4 emissions from two rice cropping systems, as affected by straw incorporation.

Fig. S5. Model-averaged importance of the predictors of the straw addition effect on CH_4 emissions and rice growth.

Fig. S6. The correlation between straw incorporation rate and experimental duration for studies included in our meta-analysis.

Table S1. Soil properties in two rice cropping systems, as affected by 12 years of straw incorporation.

Table S2. Overview of the straw incorporation studies included in our meta-analysis.

Dataset S1. Overview of all experimental observations from straw incorporation experiments that were used for the meta-analysis on CH_4 emissions and rice plant growth.

References (*40*–*63*)

REFERENCES AND NOTES

1. C. Zhu, K. Kobayashi, I. Loladze, J. Zhu, Q. Jiang, X. Xu, G. Liu, S. Seneweera, K. L. Ebi, A. Drewnowski, N. K. Fukagawa, L. H. Ziska, Carbon dioxide (CO_2) levels this century will alter the protein, micronutrients, and vitamin content of rice grains with potential health consequences for the poorest rice-dependent countries. *Sci. Adv.* **4**, eaaq1012 (2018).
2. N. Alexandratos, J.Bruinsma, *World Agriculture Towards 2030/2050: The 2012 Revision* (Food and Agriculture Organization of the United Nations, 2012).
3. D. K. Ray, N. Ramankutty, N. D. Mueller, P. C. West, J. A. Foley, Recent patterns of crop yield growth and stagnation. *Nat. Commun.* **3**, 1293 (2012).
4. N. D. Mueller, J. S. Gerber, M. Johnston, D. K. Ray, N. Ramankutty, J. A. Foley, Closing yield gaps through nutrient and water management. *Nature* **490**, 254–257 (2012).
5. C. Liu, M. Lu, J. Cui, B. Li, C. Fang, Effects of straw carbon input on carbon dynamics in agricultural soils: A meta-analysis. *Glob. Chang. Biol.* **20**, 1366–1381 (2014).
6. S. Huang, Y. Zeng, J. Wu, Q. Shi, X. Pan, Effect of crop residue retention on rice yield in China: A meta-analysis. *Field Crop Res* **154**, 188–194 (2013).
7. R. Conrad, Microbial ecology of methanogens and methanotrophs. *Adv. Agron.* **96**, 1–63 (2007).
8. IPCC, *Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change* (Cambridge Univ. Press, 2013).
9. K. M. Carlson, J. S. Gerber, N. D. Mueller, M. Herrero, G. K. MacDonald, K. A. Brauman, P. Havlik, C. S. O'Connell, J. A. Johnson, S. Saatchi, P. C. West, Greenhouse gas emissions intensity of global croplands. *Nat. Clim. Chang.* **7**, 63–68 (2017).
10. B. Linquist, K. J. van Groenigen, M. A. Adviento Borbe, C. Pittelkow, C. van Kessel, An agronomic assessment of greenhouse gas emissions from major cereal crops. *Glob. Chang.Biol.* **18**, 194–209 (2012).
11. R. Conrad, M. Klose, Y. Lu, A. Chidthaisong, Methanogenic pathway and archaeal communities in three different anoxic soils amended with rice straw and maize straw. *Front. Microbiol.* **3**, 4 (2012).
12. Y. Lu, R. Wassmann, H. U. Neue, C. Huang, C. S. Bueno, Methanogenic responses to exogenous substrates in anaerobic rice soils. *Soil Biol. Biochem.* **32**, 1683–1690 (2000).
13. Y. Zheng, L.-M. Zhang, Y.-M. Zheng, H. Di, J.-Z. He, Abundance and community composition of methanotrophs in a Chinese paddy soil under long-term fertilization practices. *J. Soil. Sediment.* **8**, 406–414 (2008).
14. Y. Cai, Y. Zheng, P. L. E. Bodelier, R. Conrad, Z. Jia, Conventional methanotrophs are responsible for atmospheric methane oxidation in paddy soils. *Nat. Commun.* **7**, 11728 (2016).
15. R. S. Hanson, T. E. Hanson, Methanotrophic bacteria. *Microbiol. Rev.* **60**, 439–471 (1996).
16. Y. Jiang, K. J. van Groenigen, S. Huang, B. A. Hungate, C. van Kessel, S. Hu, J. Zhang, L. Wu, X. Yan, L. Wang, J. Chen, X. Hang, Y. Zhang, W. R. Horwath, R. Ye, B. A. Linquist, Z. Song, C. Zheng, A. Deng, W. Zhang, Higher yields and lower methane emissions with new rice cultivars. *Glob. Chang. Biol.* **23**, 4728–4738 (2017).
17. K. Ma, Q. Qiu, Y. Lu, Microbial mechanism for rice variety control on methane emission from rice field soil. *Glob. Chang. Biol.* **16**, 3085–3095 (2010).
18. X. Yan, K. Yagi, H. Akiyama, H. Akimoto, Statistical analysis of the major variables controlling methane emission from rice fields. *Glob. Chang. Biol.* **11**, 1131–1141 (2005).
19. IPCC, *2006 International Panel for Climate Change Guidelines for National Greenhouse Gas Inventories*; www.ipcc-nggip.iges.or.jp/public/2006gl/
20. Y. Huang, R. L. Sass, F. M. Fisher Jr., A semi-empirical model of methane emission from flooded rice paddy soils. *Glob. Chang. Biol.* **4**, 247–268 (1998).
21. Y. Huang, W. Zhang, X. Zheng, J. Li, Y. Yu, Modeling methane emission from rice paddies with various agricultural practices. *J. Geophys. Res.* **109**, D08113 (2004).
22. W. Zhang, Y. Yu, Y. Huang, T. Li, P. Wang, Modeling methane emissions from irrigated rice cultivation in China from 1960 to 2050. *Glob. Chang. Biol.* **17**, 3511–3523 (2011).
23. G. R. Krishnayya, T. L. Setter, R. K. Sarkar, P. Krishnan, I. Ravi, Influence of phosphorus application to floodwater on oxygen concentrations and survival of rice during complete submergence. *Exp. Agric.* **35**, 167–180 (1999).
24. J. Schimel, Rice, microbes and methane. *Nature* **403**, 375–377 (2000).
25. Y. Zhao, M. Wang, S. Hu, X. Zhang, Z. Ouyang, G. Zhang, B. Huang, S. Zhao, J. Wu, D. Xie, B. Zhu, D. Yu, X. Pan, S. Xu, X. Shi, Economics- and policy-driven organic carbon input enhancement dominates soil organic carbon accumulation in Chinese croplands. *Proc. Natl. Acad. Sci. U.S.A.* **115**, 4045–4050 (2018).
26. H. Schütz, W. Seiler, R. Conrad, Processes involved in formation and emission of methane in rice paddies. *Biogeochem.* **7**, 33–53 (1989).
27. Q. Shang, X. Yang, C. Gao, P. Wu, J. Liu, Y. Xu, Q. Shen, J. Zou, S. Guo, Net annual global warming potential and greenhouse gas intensity in Chinese double rice-cropping systems: A 3-year field measurement in long-term fertilizer experiments. *Glob. Chang. Biol.* **17**, 2196–2210 (2011).
28. M. Martínez-Eixarch, C. Alcaraz, M. Viñas, J. Noguerol, X. Aranda, F. X. Prenafeta-Boldú, J. A. Saldaña-De la Vega, M. del Mar Català, C. Ibáñez, Neglecting the fallow season can significantly underestimate annual methane emissions in mediterranean rice fields. *PLOS ONE* **13**, e0198081 (2018).
29. X. Yan, H. Akiyama, K. Yagi, H. Akimoto, Global estimations of the inventory and mitigation potential of methane emissions from rice cultivation conducted using the 2006 Intergovernmental Panel on Climate Change guidelines. *Global Biogeochem. Cycles* **23**, GB2002 (2009).
30. X. Huang, C. Chen, H. Qian, M. Chen, A. Deng, J. Zhang, W. Zhang, Quantification for carbon footprint of agricultural inputs of grains cultivation in China since 1978. *J. Clean. Prod.* **142**, 1629–1637 (2017).
31. X. Huang, M. Li, J. Li, Y. Song, A high-resolution emission inventory of crop burning in fields in China based on MODIS thermal anomalies/fire products. *Atmos. Environ.* **50**, 9–15 (2012).
32. Z. Zhang, X. Zhang, M. Xu, S. Zhang, S. Huang, W. Liang, Responses of soil micro-food web to long-term fertilization in a wheat–maize rotation system. *Appl. Soil Ecol.* **98**, 56–64 (2016).
33. A. J. Holmes, A. Costello, M. E. Lidstrom, J. C. Murrell, Evidence that participate methane monooxygenase and ammonia monooxygenase may be evolutionarily related. *FEMS Microbiol. Lett.* **132**, 203–208 (1995).
34. P. E. Luton, J. M. Wayne, R. J. Sharp, P. W. Riley, The *mcrA* gene as an alternative to 16S rRNA in the phylogenetic analysis of methanogen populations in landfill. *Microbiol.* **148**, 3521–3530 (2002).
35. L. V. Hedges, J. Gurevitch, P. S. Curtis, The meta-analysis of response ratios in experimental ecology. *Ecology*, **80**, 1150–1156 (1999).
36. K. J. van Groenigen, C. W. Osenberg, C. Terrer, Y. Carrillo, F. A. Dijkstra, J. Heath, M. Nie, E. Pendall, R. P. Phillips, B. A. Hungate, Faster turnover of new soil carbon inputs under increased atmospheric CO_2. *Glob. Chang. Biol.* **23**, 4420–4429 (2017).
37. C. Terrer, S. Vicca, B. A. Hungate, R. P. Phillips, I. C. Prentice, Mycorrhizal association as a primary control of the CO_2 fertilization effect. *Science* **353**, 72–74 (2016).
38. J. Chen, Y. Luo, K. J. van Groenigen, B. A. Hungate, J. Cao, X. Zhou, R.-w. Wang, A keystone microbial enzyme for nitrogen control of soil carbon storage. *Sci. Adv.* **4**, eaaq1689 (2018).

39. W. Viechtbauer, Conducting meta-analyses in R with the metafor package. *J. Stat. Softw.* **36**, 1–48 (2010).
40. J. Shen, H. Tang, J. Liu, C. Wang, Y. Li, T. Ge, D. L. Jones, J. Wu, Contrasting effects of straw and straw-derived biochar amendments on greenhouse gas emissions within double rice cropping systems. *Agric. Ecosyst. Environ.* **188**, 264–274 (2014).
41. L. Xia, S. Wang, X. Yan, Effects of long-term straw incorporation on the net global warming potential and the net economic benefit in a rice–wheat cropping system in China. *Agric. Ecosyst. Environ.* **197**, 118–127 (2014).
42. J. Xiong, D.-Q. Zhang, X.-j. Shi, J.-P. Ni, D.-T. Xie, Z.-j. Mu, Effects of different long-term fertilization and crop residue management on methane emissions from paddy fields with purple soil. *J. Southwest China Normal Univ.* **38**, 98–102 (2013).
43. P. Hou, G. Li, S. Wang, X. Jin, Y. Yang, X. Chen, C. Ding, Z. Liu, Y. Ding, Methane emissions from rice fields under continuous straw return in the middle-lower reaches of the Yangtze River. *J. Environ. Sci.* **25**, 1874–1881 (2013).
44. Z. Gong, S. Yan, C. Yan, J. Wang, H. Zhang, Effect of rice straw retention and temperature on methane emission in rice field in cold region. *J. Northeast Agr. Univ.* **46**, 8–15 (2015).
45. B. Xie, Z. Zhou, B. Mei, X. Zheng, H. Dong, R. Wang, S. Han, F. Cui, Y. Wang, J. Zhu, Influences of free-air CO_2 enrichment (FACE), nitrogen fertilizer and crop residue incorporation on CH_4 emissions from irrigated rice fields. *Nutr. Cycl. Agroecosyst.* **93**, 373–385 (2012).
46. X. Zheng, Z. Zhou, Y. Wang, J. Zhu, Y. Wang, J. Yue, Y. Shi, K. Kobayashi, K. Inubushi, Y. Huang, S. Han, Z. Xu, B. Xie, K. Butterbach-Bahl, L. Yang, Nitrogen-regulated effects of free-air CO_2 enrichment on methane emissions from paddy rice fields. *Glob. Chang. Biol.* **12**, 1717–1732 (2006).
47. X. Li, J. Ma, Y. Yao, S. Liang, G. Zhang, H. Xu, K. Yagi, Methane and nitrous oxide emissions from irrigated lowland rice paddies after wheat straw application and midseason aeration. *Nutr. Cycl. Agroecosyst.* **100**, 65–76 (2014).
48. G. Liu, H. Yu, J. Ma, H. Xu, Q. Wu, J. Yang, Y. Zhuang, Effects of straw incorporation along with microbial inoculant on methane and nitrous oxide emissions from rice fields. *Sci. Total Environ.* **518–519**, 209–216 (2015).
49. N. Hu, B. Wang, Z. Gu, B. Tao, Z. Zhang, S. Hu, L. Zhu, Y. Meng, Effects of different straw returning modes on greenhouse gas emissions and crop yields in a rice–wheat rotation system. *Agric. Ecosyst. Environ.* **223**, 115–122 (2016).
50. J. Wang, X. Zhang, Z. Xiong, M. A. K. Khalil, X. Zhao, Y. Xie, G. Xing, Methane emissions from a rice agroecosystem in South China: Effects of water regime, straw incorporation and nitrogen fertilizer. *Nutr. Cycl. Agroecosyst.* **93**, 103–112 (2012).
51. Z. Xiong, Y. Liu, Z. Wu, X. Zhang, P. Liu, T. Huang, Differences in net global warming potential and greenhouse gas intensity between major rice-based cropping systems in china. *Sci. Rep.* **5**, 17774 (2015).
52. Y. Sui, J. Gao, C. Liu, W. Zhang, Y. Lan, S. Li, J. Meng, Z. Xu, L. Tang, Interactive effects of straw-derived biochar and N fertilization on soil C storage and rice productivity in rice paddies of Northeast China. *Sci. Total Environ.* **544**, 203–210 (2016).
53. G.-Y. Kim, J. Gutierrez, H.-C. Jeong, J.-S. Lee, M. D. M. Haque, P. J. Kim, Effect of intermittent drainage on methane and nitrous oxide emissions under different fertilization in a temperate paddy soil during rice cultivation. *J. Korean Soc. Appl. Biol. Chem.* **57**, 229–236 (2014).
54. Y.-f. Zhang, L.-g. Chen, P.-p. Zhu, C.-s. Zhang, J. Sheng, Z.-c. Wang, J.-c. Zheng, Preliminary study on effect of straw incorporation on net global warming potential in high production rice-wheat double cropping systems. *J. Agro-Environ. Sci.* **31**, 1647–1653 (2012).
55. H.-h. Wang, M.-x. Shen, C.-y. Lu, Y-c. Zhang, T.-d. Wu, L.-l. Shi, X.-w. Zhou, Effect of patterns of straw returning to field on methane and nitrous oxide emissions during rice-growing season in a rice-wheat double cropping system. *Jiangsu J. Agric. Sci.* **30**, 758–763 (2014).
56. L. Zhang, J. Zheng, L. Chen, M. Shen, X. Zhang, M. Zhang, X. Bian, J. Zhang, W. Zhang, Integrative effects of soil tillage and straw management on crop yields and greenhouse gas emissions in a rice–wheat cropping system. *Eur. J. Agron.* **63**, 47–54 (2015).
57. W. Wang, X. Wu, A. Chen, X. Xie, Y. Wang, C. Yin, Mitigating effects of ex situ application of rice straw on CH_4 and N_2O emissions from paddy-upland coexisting system. *Sci. Rep.* **6**, 37402 (2016).
58. J. Liu, P. Wu, X. Xie, X. Fu, Q. Shen, S. Guo, Methane emission from late rice fields in Hunan red soil under different long-term fertilizing systems. *Acta Ecol. Sin.* **28**, 2878–2886 (2008).
59. G. Chu, Z. Wang, H. Zhang, L. Liu, J. Yang, J. Zhang, Alternate wetting and moderate drying increases rice yield and reduces methane emission in paddy field with wheat straw residue incorporation. *Food Energy Secur.* **4**, 238–254 (2015).
60. Y. Wang, C. Hu, B. Zhu, H. Xiang, X. He, Effects of wheat straw application on methane and nitrous oxide emissions from purplish paddy fields. *Plant Soil Environ.* **56**, 16–22 (2010).
61. J. Ma, X. Li, H. Xu, Z. Cai, K. Yagi, Effects of nitrogen fertiliser and wheat straw application on CH_4 and N_2O emissions from a paddy rice field. *Soil Res.* **45**, 359–367 (2007).
62. J. Ma, E. Ma, H. Xu, K. Yagi, Z. Cai, Wheat straw management affects CH_4 and N_2O emissions from rice fields. *Soil Biol. Biochem.* **41**, 1022–1028 (2009).
63. J. Ma, H. Xu, Y. Han, Z. Cai, K. Yagi, Short-term effects of wheat straw incorporation into paddy field as affected by rice transplanting time. *Soil Res.* **46**, 281–287 (2008).

Acknowledgments

Funding: This work was supported by the National Key Research and Development Program of China (2016YFD0300903, 2017YFD0300104, 2016YFD0300501, and 2015BAC02B02), the Special Fund for Agro-scientific Research in the Public Interest (201503118 and 201503122), the China Agriculture Research System–Green Manure (CARS-22-G-16), the Central Public-interest Scientific Institution Basal Research Fund of Institute of Crop Science, the Innovation Program of CAAS (Y2016PT12 and Y2016XT01), and the GEF Project of Climate Smart Staple Crop Production in China (P144531). **Author contributions:** Y.J. and W.Z. designed the research. Y.J., H.Q., S.H., X.Z., L.W., L.Z., M.S., X.X., H.Z., F.C., C. Li, C. Lu, J.Z., and A.D. conducted the experiments and data collection. Y.J., H.Q., S.H., K.J.v.G., and W.Z. contributed to data analysis and interpretation and wrote the manuscript. All authors commented on and approved the final manuscript. **Competing interests:** The authors declare that they have no competing interests. **Data and materials availability:** All data needed to evaluate the conclusions in the paper are present in the paper and/or the Supplementary Materials. Additional data related to this paper may be requested from the authors.

Submitted 29 July 2018
Accepted 6 December 2018
Published 16 January 2019
10.1126/sciadv.aau9038

Citation: Y. Jiang, H. Qian, S. Huang, X. Zhang, L. Wang, L. Zhang, M. Shen, X. Xiao, F. Chen, H. Zhang, C. Lu, C. Li, J. Zhang, A. Deng, K. J. van Groenigen, W. Zhang, Acclimation of methane emissions from rice paddy fields to straw addition. *Sci. Adv.* **5**, eaau9038 (2019).

Science Advances

Acclimation of methane emissions from rice paddy fields to straw addition

Yu Jiang, Haoyu Qian, Shan Huang, Xingyue Zhang, Ling Wang, Li Zhang, Mingxing Shen, Xiaoping Xiao, Fu Chen, Hailin Zhang, Changying Lu, Chao Li, Jun Zhang, Aixing Deng, Kees Jan van Groenigen and Weijian Zhang

Sci Adv **5** (1), eaau9038.
DOI: 10.1126/sciadv.aau9038