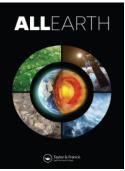


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To cite this article: Paul Abayomi Sobowale Soremi, Ngonidzashe Chirinda, Eduardo Graterol & Maria Fernanda Alvarez (2023) Potential of rice (Oryza sativa L.) cultivars to mitigate methane emissions from irrigated systems in Latin America and the Caribbean, All Earth, 35:1, 149-157, DOI: 10.1080/27669645.2023.2207941

To link to this article: https://doi.org/10.1080/27669645.2023.2207941

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Published online: 09 May 2023.

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# Potential of rice (*Oryza sativa* L.) cultivars to mitigate methane emissions from irrigated systems in Latin America and the Caribbean

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#### ABSTRACT

In irrigated rice fields, plant-mediated transfer of CH<sub>4</sub> from submerged soils to the atmosphere raise the possibility of genotypic differences in CH<sub>4</sub> emissions. Previous research has been contradictory, and varietal differences in rice CH<sub>4</sub> emissions in Latin America have not been examined. A field experiment in Colombia tested whether irrigated rice emissions might be reduced using a breeding line, an inbred variety, and two rice hybrids. Data was collected on CH<sub>4</sub> emissions, phenotypic, root, and grain yield parameters. Variations observed in CH<sub>4</sub> emissions, grain yield, root length, and root surface area were in the order Hybrid 2 > Hybrid 1 > breeding line > inbred variety. CH<sub>4</sub> emissions per unit area were between 29% and 62% greater for the hybrids than the inbred variety and breeding line, and CH<sub>4</sub> emissions per unit grain yield were comparable across genotypes. Our findings suggest that differences in root characteristics and aboveground biomass explain genetic influences on CH<sub>4</sub> emissions. The transition to low-emission rice production systems can be accelerated by using differences in productivity and root qualities among cultivars.

#### Key policy highlights

- CH<sub>4</sub> emission intensities were comparable among the breeding line, inbred variety and hybrids.
- Breeding for shorter roots, lower root volume, and surface area without compromising yields may be beneficial in reducing rice-based methane emissions.
- High-yielding, low-CH<sub>4</sub> emission rice should be classified as a clean development technology that qualifies for carbon credits.

#### 1. Introduction

Deficits in rice supply are common in Latin America and the Caribbean (LAC) region, where per capita rice consumption is approximately 30 kg per year. However, imports are used to make up the shortfall (Durand-Morat & Bairagi, 2021; Maclean et al., 2002). Irrigated rice, a known anthropogenic source of methane (CH<sub>4</sub>), currently accounts for over 70% of the LAC region's total rice-growing area (Espinosa, 2002). Expansion of rice cultivation areas through the utilisation of available land and water resources is possible in countries such as Brazil, Colombia, and Venezuela (EMSAL 2021).

Globally, rice production is responsible for an estimated 10–12% of total CH<sub>4</sub> emissions from anthropogenic sources (Ciais et al., 2013). Asian countries account for 90% of total global rice production, which is significantly higher than the estimated<5% of global rice production in the LAC region (EMSAL 2021). The disproportionately lower rice growing area in LAC implies a minor contribution of rice production systems to global methane emission compared to the Asian region (FAOSTAT, 2018). Nonetheless, rice production systems in the LAC region should aim to harness irrigation water resources to produce more rice without increasing total anthropogenic CH<sub>4</sub> emissions (S. F. Islam et al., 2018).

Methanogens that produce  $CH_4$  flourish under anaerobic conditions characteristic of flooded rice fields (Sass & FM, 1997). Molecular diffusion, ebullition, and plant-assisted transport are the mediums used to carry  $CH_4$  from the soil to the atmosphere (Khosa et al., 2010). Previous studies have reported that>90% of methane emitted from rice fields is lost through plantmediated transport (Butterbach-Bahl et al., 1997; Holzapfel-Pschorn et al., 1986; Wassmann & Aulakh, 2000). Yet, despite the importance of plants in

**ARTICLE HISTORY** 

Received 24 August 2022 Accepted 24 April 2023

#### **KEYWORDS**

Climate change mitigation; sustainable rice production; rice cultivars; methane emissions

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transmitting  $CH_4$  from the soil to the atmosphere, research on varietal differences in  $CH_4$  emissions is limited.

Climate change mitigation efforts in rice production systems have primarily focused on cultural practices, including fertiliser placement (Gaihre et al., 2015, 2018; S. M. M. Islam et al., 2018, 2022), agricultural residue management (Wassmann et al., 1993; Yagi et al., 1997), and irrigation management (Cheng et al., 2022; S. M. M. Islam et al., 2016, 2020; Wassmann et al., 2000). However, currently, it is very challenging for farmers to change their fertilisation practices, residue and irrigation management in the absence of supportive legislation and economic incentives (Chirinda et al., 2018). To overcome this challenge exploiting varietal differences in CH<sub>4</sub> emissions has been proposed as a more cost-effective technique for decreasing CH<sub>4</sub> emissions in LAC because the associated adjustment would not dramatically alter farming practices (Chirinda et al., 2018; Gogoi et al., 2008).

Previous research (Gogoi et al., 2008; Qin et al., 2015; Wang et al., 2017) has revealed that genotypic differences in CH<sub>4</sub> emissions exist, implying the possibility of selecting rice varieties with lower CH<sub>4</sub> emissions. Gogoi et al. (Gogoi et al., 2008) observed that in northeast India, high-yielding rice varieties such as IR-36 had lower CH<sub>4</sub> emissions compared to traditional varieties. In the Gogoi et al. (Gogoi et al., 2008) study, the differences in CH<sub>4</sub> emissions were attributed to differences in aboveground biomass accumulation. They conclude that cultivating high-yielding rice varieties and hybrids may be key for food security and climate change mitigation (Gogoi et al., 2008). In a separate study, Zhang et al. (Zhang et al., 2019) examined a rice hybrid and variety in China and found that root form and physiological parameters were adversely and significantly linked with total CH<sub>4</sub> emissions. The rice hybrid used in the study by Zhang et al. (Zhang et al., 2019) produced less CH<sub>4</sub> and had higher yields than those observed in the studied rice variety.

The concept of genotypes that emit less  $CH_4$  without compromising yield is a compelling solution for fighting climate change and achieving food security goals (Balakrishnan et al., 2018). Therefore, monitoring  $CH_4$  emissions from different varieties and hybrids is necessary to identify genotypes that reduce  $CH_4$  emissions without compromising grain yield. Furthermore, to guide future breeding efforts, it is necessary to understand the mechanisms that drive  $CH_4$  emission reductions. In the present study, We hypothesised that hybrids produce less  $CH_4$  per unit yield because they had higher harvest indexes than varieties.

### 2. Materials and methods

From June to October 2019, a designed experiment was conducted on a rice field at the International Center of Tropical Agriculture (CIAT) in Cali, Colombia. The field was laid out in a randomised complete block design with four treatments in four replicates. Four rice cultivars were used in the study: a Colombian inbred variety (135 days maturity), a CIAT experimental breeding line previously chosen for its increased root length (130 days maturity), and two hybrids [Hybrid 1 (126 days maturity) and Hybrid 2 (128 days maturity)] chosen for their higher yield performance in CIAT field trials compared to check-inbred-varieties. On the 17th of June 2019, twenty-one-day-old rice seedlings were transplanted into field plots. The inbred variety and breeding line had similar plant populations (250,000 plants per hectare) but higher than those used for the hybrids (111,111 plants per hectare). Table 1 shows the soil parameters in the experimental field. The neutral soil pH (7.4) and high available phosphorous (49 mg kg<sup>-1</sup>) levels indicate good soil conditions for rice production. All the experimental plots were under continuous flooding during the growing season, and water was constantly applied to maintain the flooded conditions in plots with all the rice genotypes. The experimental site was mechanically prepared, irrigated and puddled before seedling transplanting. The gross plot size was  $20 \text{ m}^2$ , while the harvested net plot was  $12 \text{ m}^2$ .

Soil property	Unit	Value
рН	-	7.42
Nitrogen	g kg <sup>-1</sup>	1.03
Available phosphorus	mg kg <sup>-1</sup>	49.39
Cation exchange capacity	cmol kg <sup>-1</sup>	37.1
Soil organic matter	g kg <sup>_1</sup>	42.6
Calcium	cmol kg <sup>-1</sup>	18.3
Magnesium	cmol kg <sup>-1</sup>	9.5
Potassium	cmol kg <sup>-1</sup>	0.7
Sodium	cmol kg <sup>-1</sup>	0.6
Iron	mg kg <sup>-1</sup>	14.0
Manganese	mg kg <sup>-1</sup>	105.7
Copper	mg kg <sup>-1</sup>	3.1
Zinc	mg kg <sup>-1</sup>	21.9
Boron	mg kg <sup>-1</sup>	0.5
Sulphur	mg kg <sup>-1</sup>	44.5
Oxidized carbon	g kg <sup>-1</sup>	18.75

Table 1. Soil properties at the study site.

Nitrogen fertiliser was applied at 180 kg N ha<sup>-1</sup> and 200 kg N ha<sup>-1</sup> for the varieties and hybrids, respectively, in three split applications of 10%, 70%, and 20% at 2, 10, and 55 days after transplanting, respectively. Similar rates of phosphorus (65 kg  $ha^{-1}$ ) and potassium (90 kg  $ha^{-1}$ ) were applied to all plots. Five aboveground plant samples were obtained from each treatment gross plot at four growth phases. The samples were dried at 60°C for 72 hours. The dry weight of plants from different net plots at harvest maturity was divided into leaves, stems, roots, and panicles. AgroTronix MT-PRO moisture metre was also used to determine the grain moisture content at harvest maturity. At maturity, root length, volume, and surface area were measured according to (Ogawa et al., 2014). Roots from five plants were collected with a core sampler, washed, and scanned with an EPSON scanner before being analysed with WinRhizo.

From July 10 to 25 September 2019, gas measurements were conducted on 16 separate days during the rice growing season. Around fertilisation, gas samples were collected more frequently. Specifically, four gas samples were collected from each treatment plot one day before and three days after fertilisation. For the rest of the rice growing season, gas measurements were conducted weekly until harvest. All samplings were in the morning (8:00 to 11:00 a.m.). The staticclosed chamber technique was utilised for gas sampling (31). Plastic buckets (114 L volume and 80 cm height) were used with custom-made chamber bases (40 cm height) that contained a canal, allowing for a water seal during chamber deployment. The static chambers had vents to facilitate pressure equilibration and a fan to mix the air (Chirinda et al., 2017). During each sampling campaign, at four 15-minute intervals (0, 15, 30, 45 mins), gas samples (20 mL) were obtained from the closed static chambers in each treatment plot using a syringe and injected into pre-evacuated vials before analysis. The methane concentration in the vials was determined using gas chromatography (Shimadzu GC-20C14) equipped with a Porapak K column and a flame ionisation detector (FID). The carrier gas was nitrogen (N<sub>2</sub>). Methane fluxes were calculated using the gas concentration rate, ideal gas equation and sampling time.

$$CH_{4flux} = \frac{\Delta C}{\Delta t} \times \frac{V}{A} \times p \times \frac{273}{273 + T}$$

 $\Delta C/\Delta t$  is the concentration change (ppm-CH<sub>4</sub> h<sup>-1</sup>); V is chamber volume (m<sup>3</sup>); A is chamber area (m<sup>2</sup>);  $\rho$  is gas density (0.717 kg m<sup>-3</sup> at 0°C); and T is the mean air temperature inside the chamber (°C). The cumulative fluxes for the monitoring period were calculated using linear interpolation and expressed as emissions per unit area (absolute emissions). CH<sub>4</sub> emission intensities were calculated using the ratio of cumulative emissions to grain yield, which was expressed in mg  $CH_4$  per kg grain dry matter yield.

#### 3. Statistical analyses

The normality of the data was checked. We used SAS's proc mixed method to evaluate the data on crop parameters because it was normally distributed. The models investigated the effects of genotype on various crop parameters. As random effects, the replicates were included. Pairwise comparisons were performed using Differences of Least Square Means in conjunction with these models. The methane data were not normally distributed, and none of the transformation procedures yielded variance homogeneity or normal distribution. To examine the paired difference in methane emissions between the genotypes, we utilised a non-parametric approach (the Wilcoxon Signed Rank test), which does not need the assumption of normality. While non-parametric methods may not provide the same breadth of statistical insights as parametric methods, they are helpful when flux data is non-normal (Corre et al., 1996).

## 4. Results

Aboveground biomass accumulation was comparable in all the genotypes during the vegetative growth stage (Figure 1). However, the hybrids had significantly higher aboveground biomass than the other genotypes at 50% flowering and physiological maturity. Hybrid 2 had a significantly longer root system than other genotypes at maturity (Table 2). Root volume and surface area followed the same trend as root length, but the breeding line's values were comparable to those in Hybrid 2 (which had the biggest root volume) and higher than those in Hybrid 1 and the inbred variety.

Tiller numbers at maximum tillering, flowering and physiological maturity were significantly higher for the inbred variety, which had a higher plant count (25 per  $m^2$ ) than the hybrids (11 per  $m^2$ ) (Table 3). However, despite the high plant count (25 per m<sup>2</sup>), the breeding line had similar tillers to the hybrids at maximum tillering and flowering. The hybrids produced similar grain yields, but the yield was higher than the other genotypes (Table 4). For Hybrid 1 and Hybrid 2, the relative increase in yield over the inbred variety was 26.4% and 26.7%, respectively. Hybrids 1 and 2, correspondingly, exhibited 46.0% and 46.3% higher grain yields than the breeding line. The different genotypes had similar daily methane fluxes (Figure 2). Cumulative methane emissions followed the order: Hybrid 2 > Hybrid 1 > breeding line > inbred variety (Table 4). Hybrid 2 emitted 18.5% more than Hybrid 1, which emitted 22.2% more

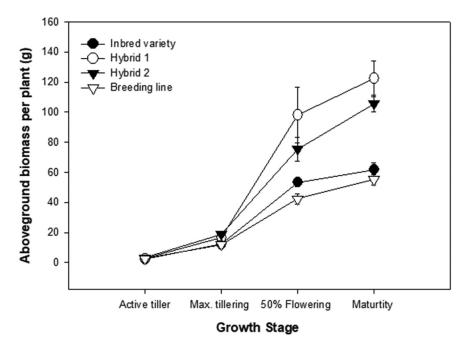


Figure 1. Aboveground biomass accumulation by the four rice cultivars at four growth stages (active tillering, maximum tillering, 50% flowering and maturity). The error bars represent the standard error of the means.

Table 2. Root parameters measured during the rice-grow
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Genotype	Root length at physiological maturity (cm)	Root volume at physiological maturity (cm <sup>3</sup> )	Root surface area at physiological maturity (cm <sup>2</sup> )
Inbred variety	2763 <sup>a</sup>	6.4 <sup>a</sup>	465ª
Breeding line	3567ª	8.4 <sup>ab</sup>	607 <sup>ab</sup>
Hybrid 1	2908ª	8.1ª	540 <sup>a</sup>
Hybrid 2	3826 <sup>b</sup>	11.0 <sup>b</sup>	725 <sup>b</sup>

**Table 3.** Tiller numbers (per m<sup>2</sup>) counted during three growth stages of the rice-growing seasons.

Genotype	Maximum tillering	Flowering	Physiological maturity
Inbred variety	348 <sup>a</sup>	345 <sup>a</sup>	378 <sup>a</sup>
Breeding line	236 <sup>b</sup>	213 <sup>b</sup>	217 <sup>b</sup>
Hybrid 1	273 <sup>bc</sup>	258 <sup>bc</sup>	267 <sup>c</sup>
Hybrid 2	283 <sup>bc</sup>	268 <sup>bc</sup>	291 <sup>c</sup>

than the breeding line, while the breeding line emitted 3.2% more  $CH_4$  compared to the inbred variety. Even though the hybrids had much higher absolute  $CH_4$  emissions (i.e. emissions per unit area) than the breeding line and inbred variety,  $CH_4$  emission intensities (i.e. emissions per unit yield) were comparable across genotypes.

During the maximum tillering period, the mean daily CH<sub>4</sub> fluxes were significantly higher in the hybrids than in the in-bred variety and breeding line (Table 5). However, the mean daily CH<sub>4</sub> fluxes were similar in the in-bred variety and the breeding line. On the other hand, mean daily CH<sub>4</sub> emissions were similar for the hybrids. Though not significantly different, similar patterns in mean daily CH<sub>4</sub> fluxes were observed during the flowering period. During the rice maturing period, the daily CH<sub>4</sub> fluxes were significantly higher in Hybrid 2 than in the breeding line (Table 4). When we excluded the breeding line and only focused on the commercial hybrids and in-bred variety, we observed linear relationships ( $r^2$ >0.98) between mean daily CH<sub>4</sub> emissions and selected plant parameters, including aboveground biomass at maximum tillering, root surface area, volume and length (Figure 3).

#### 5. Discussion

Based on the dominance of rice-mediated CH<sub>4</sub> transmission from the submerged soil to the atmosphere (Jia & Cai, 2003), previous studies have shown that genetic variations in rice influence CH<sub>4</sub> emissions from irrigated rice systems (Baruah et al., 2010; Jia & Cai, 2003; Qin et al., 2015). The current study rejected our hypothesis that the studied rice hybrids produce less CH<sub>4</sub> per unit yield. However, we observed differences in absolute CH4 emissions, with the inbred variety and breeding line resulting in significantly lower  $CH_4$  emissions than those obtained for the rice hybrids. Though we explored several plant parameters (supplementary material), focusing on the commercial inbred variety and the hybrids, we observed a strong relationship between mean daily CH<sub>4</sub> emissions to aboveground at maximum tillering and root parameters

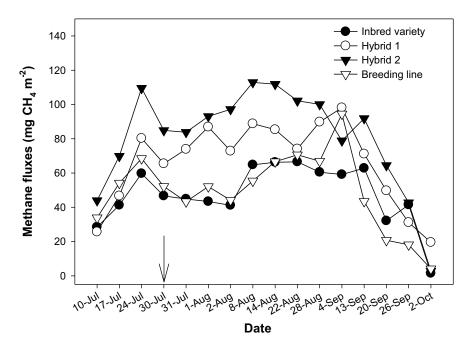


Figure 2. Methane emission dynamics of four rice cultivars observed during the monitoring period. Arrow indicates the date of the third N fertilisation.

Table 4. Cumulative CH<sub>4</sub> emissions, grain yield and grain quality parameters.

Genotype	Cumulative methane (mg CH <sub>4</sub> m <sup>-2</sup> )	Emission intensity (mg $CH_4$ kg <sup>-1</sup> dry matter)	Grain yield (kg ha <sup>-1</sup> )	1000 grain weight (g)	Harvest index (%)
Inbred variety	4338 <sup>a</sup>	0.73	6333ª	25.9ª	45.3
Breeding line	4482 <sup>a</sup>	0.75	5801ª	26.2 <sup>ab</sup>	48.3
Hybrid 1	5761 <sup>b</sup>	0.65	8607 <sup>b</sup>	24.2 <sup>b</sup>	43.0
Hybrid 2	7068 <sup>c</sup>	1.19	8647 <sup>b</sup>	23.9 <sup>b</sup>	52.0

**Table 5.** Mean daily methane fluxes (mg  $CH_4$  m<sup>-2</sup> d<sup>-1</sup>) during the maximum tillering, flowering and physiological maturity period.

Genotype	Maximum Tillering	Flowering	Maturity
Inbred variety	49.8 <sup>a</sup>	62.1ª	45.5 <sup>ab</sup>
Breeding line	50.5 <sup>a</sup>	77.3 <sup>a</sup>	27.5 <sup>ª</sup>
Hybrid 1	82.9 <sup>b</sup>	87.3 <sup>a</sup>	50.7 <sup>ab</sup>
Hybrid 2	101.1 <sup>b</sup>	93.7 <sup>a</sup>	66.4 <sup>b</sup>

**Note**: In each column, for each growth stage, values with the same letter are not significantly different.

(root length, volume and surface area) measured during the physiological maturity period. This finding is in corroboration with previous studies that observed differences in phenotypic parameters such as aboveground biomass and root characteristics are major drivers of differences in CH<sub>4</sub> emissions between rice genotypes (Baruah et al., 2010; Khosa et al., 2010; Su et al., 2015). The lower grain yields and CH<sub>4</sub> emissions observed in the inbred variety, despite its higher tiller numbers, suggest that several of these tillers were unproductive, and the high tiller numbers did not translate to high CH<sub>4</sub> emissions. The low CH<sub>4</sub> emissions may be because high tiller numbers in the inbred variety may have enhanced CH<sub>4</sub> oxidation due to a bigger volume of aerenchyma increasing oxygen movement from the atmosphere to the rhizosphere (Bhattacharyya et al., 2019).

The close alignment between CH<sub>4</sub> emissions and different root parameters is unsurprising as the roots of different genotypes have been reported to influence the soil methanotrophic community composition in various ways (Aulakh et al., 2000). Root influences on the methanotrophic community are probably driven by the fact that different rice cultivars appear to devote more photosynthetic products to root exudation than others (Win et al., 2012). The root exudates provide an organic substrate for microbial organisms that soil methanogens can use as C sources for CH<sub>4</sub> production (Setvanto et al., 2004). In addition, the release of root exudates, including simple sugars (electron donors), enhances soil anaerobiosity creating conditions conducive to CH<sub>4</sub> production by affecting the soil redox potential (Gutierrez et al., 2014; Wassmann & Aulakh, 2000). Wassmann and Aulakh (Wassmann & Aulakh, 2000) further highlight that variations in the quantity and quality of exudates may emanate from differences in rice genotypes. In this study, we did not monitor the content and composition of root exudates; however, we suspect that the root length, volume and surface area may have influenced the quantity of root exudates released into the rhizosphere (Williams et al., 2022). Hybrid 2, which had the largest root length, volume and surface area at maturity, consistently had the highest CH<sub>4</sub> emissions throughout the season,

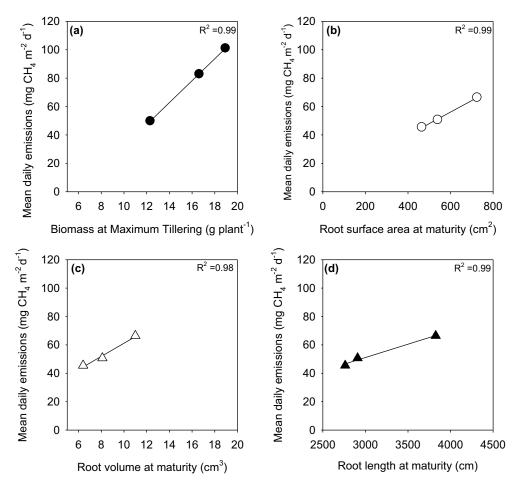


Figure 3. For the commercial genotypes only (excluding the breeding line), the linear relationships between mean daily methane emissions (measured during the respective periods) and (a) biomass at maximum tillering, (b) root surface area at maturity, (c) root volume at maturity, and (d) root length at maturity.

further suggesting the importance of root characteristics in driving  $CH_4$  production. On the other hand, the inbred variety with the least aboveground biomass at maximum tillering, 50% flowering and maturity, the shortest root length and the smallest root volume and surface area also had the lowest absolute  $CH_4$ emissions.

In corroboration with observations made by Minoda and Kimura (Minoda & Kimura, 1994), and Aulakh et al (Aulakh et al., 2001), the high aboveground biomass observed in the hybrids at 50% flowering and physiological maturity may have contributed to increased root exudates and, consequently, the high CH<sub>4</sub> emissions. On the other hand, the high aboveground biomass observed for the hybrids may have resulted in greater photosynthetic rates that translated to higher grain yields compared to those observed for the inbred variety and breeding line (Ogawa et al., 2014; Pittelkow et al., 2013; Zhou et al., 2017). According to our findings, the hybrids had high grain yields and CH<sub>4</sub> emissions, while the inbred variety and breeding line had low grain yields and low emissions resulting in similar  $CH_4$  emission intensities for all the studies genotypes.

Under continuously flooded rice production systems,  $CH_4$  oxidation in the anaerobic soil is expected

to be low. However, the rice aerenchyma system may also support the movement of atmospheric oxygen into the rhizosphere to support root respiration and contribute to  $CH_4$  oxidation (Gerard & Chanton, 1993). Previous studies have shown that about 90% of produced  $CH_4$  during rice maturation is oxidised (Sass et al., 1992; Schütz et al., 1989), which may explain the reduction in the emitted  $CH_4$  from 13 September 2019 (88 days after transplanting).

The main objective of rice farmers is to maximise yields. Our findings show that adopting rice hybrids is critical for achieving this goal. However, farmers have to incur upfront costs of seeds and additional inputs (i.e. fertilisers) on the assumption of recuperating these costs to form a higher-yielding crop. However, other factors, such as extreme weather events caused by climate change and variability and pest and disease attacks, may negatively impact the yield potential resulting in losses for farmers. Inbred varieties whose seeds can be reused without compromising yields and require lower fertiliser inputs may thus be attractive for resource-limited productivity farmers, but their is lower. Notwithstanding the importance of inbred varieties in sustainable agriculture, similarities of CH<sub>4</sub>

emission intensities among the genotypes suggest the possibility of simultaneously meeting food security and climate change mitigation goals through hybrids. Nonetheless, to further reduce  $CH_4$  emissions, it may be necessary to consider combining the adoption of rice hybrids with other proven practices such as Alternate Wetting and Drying (AWD) and residue management.

#### 6. Conclusion

Our findings suggest that breeding for shorter roots, smaller root volume, and surface area without sacrificing yields could help reduce rice-based CH<sub>4</sub> emissions. To further encourage the use of high-yielding, low-CH<sub>4</sub> emission rice cultivars, further research should be conducted to generate additional evidence that supports the classification of those genotypes as clean development technologies that qualify for carbon credits. While low absolute emissions could result from a low-yielding rice variety (per unit area), farmers will need more acreage to produce enough rice to alleviate the region's rice shortages. Promoting hybrids may be a more promising strategy for simultaneously achieving food security and emission reduction targets. However, hybrid seeds are more expensive than inbred varieties, which may make the latter to be a more attractive and sustainable option for resourceconstrained farmers. Therefore, to support sustainable rice production in the LAC region, improved management and technological options should be promoted considering the multiple socio-economic contexts.

#### **Acknowledgments**

This research was supported by the CGIAR Research Program on Climate Change, Agriculture and Food Security (CCAFS) and the Global Research Alliance on Agricultural Greenhouse Gases (GRA) through their CLIFF-GRADS program. CCAFS is carried out with support from CGIAR Trust Fund and through bilateral funding agreements. For details, please visit https://ccafs.cgiar.org/ donors. We are grateful to the International Center for Tropical Agriculture (CIAT), Palmira, Colombia, with the rice program the Physiology Group and Phenomics Platform for their support with different methodologies in physiology and root development and the Latin American Fund for Irrigated Rice (FLAR), Palmira, Colombia, for hosting the first author who was the recipient of the scholarship and to the Government of New Zealand for providing the financial support.

#### **Disclosure statement**

No potential conflict of interest was reported by the authors.

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#### Data availability statement

The data used to support the findings of this study are available from the corresponding author upon request.

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