International Journal of Current Advanced Research
ISSN: O: 2319-6475, ISSN: P: 2319-6505, Impact Factor: SJIF: 5.995
Available Online at www.journalijcar.org
Volume 6; Issue 11; November 2017; Page No. 7740-7743
DOI: http://dx.doi.org/10.24327/ijcar.2017.7743.1216

Research Article

METHANE EMISSION FROM PADDY FIELDS INFLUENCED BY FERTILIZATION AND DUAL CROPPING WITH *AZOLLA*

Yoganathan .K* and Ganesh .P

Department of Microbiology, Annamalai University, Annamalainagar, Tamilnadu, India

A R T I C L E I N F O

Article History:
Received 16th August, 2017
Received in revised form 25th September, 2017
Accepted 3rd October, 2017
Published online 28th November, 2017

Key words:
Methane emission, *Azolla*, dual cropping, Soil redox potential and fertilization

A B S T R A C T

Paddy fields are thought to be major greenhouse gases emitters. Several studies have investigated the reduction of greenhouse gases emission from paddy fields by applying AWD irrigation (alternative wetting and drying), a used method of water management practice and dual cropping method during rice growing seasons. This study was carried out to investigate the influence of fertilization and dual cropping of *Azolla* with rice cultivation on methane emissions and environmental variables in flooded paddy fields. The paddy fields are located in confluence point ofVellar and Manimuktha river basin in Peruvarappur village, Tamilnadu, India. The results showed that the organic fertilization increased the methane emission, increase in Total carbon (TC) level and decrease in pH of bulk soil of OFPF treatment. The dual cropping of *Azolla* with rice significantly degreased the methane emission, likely decreasing redox potential (Eh) in root region of rice in CFPF+*Azolla* treatment.

INTRODUCTION

Rice, one of the most major cereals, has a very long history in Asian continent especially in Indian sub-continent. The demand for rice is rising year by year with the increasing population. The more activities take place in agriculture, the more they can directly affect the environment and indirectly add to the environmental load. A paddy field is a flooded parcel of arable land used for growing rice and other semiaquatic crops thought to be a major emitter of greenhouse gases such as methane, nitrous oxide, and carbon dioxide [1]. Rice fields are generally considered as an important source of atmospheric methane (CH_4) [2]. The global CH_4 emission rate from rice fields were estimated to be 20-40 Tg per year [3,4] which accounts for approximately 11% of the total methane emissions [5]. The global warming potential (GWP) of CH_4 is 25 times greater than that of CO_2 on a mass basis and 100year time horizon [6]. Therefore, management approaches to reduce the CH_4 emissions from paddy fields have attracted intensive studies. Many techniques involving the control of resources like water and fertilizers are known as ways to increase rice production. Few studies have conducted to study the methane emission with water management. For example, Yagi *et al*., [1] showed that percolation of soil organic matters and nutrients in rice field soils through water drainage decreases methane gas emission.Continuous flooding of paddy fields is known to release a substantial amount of greenhouse gas.

Corresponding author:* **Yoganathan .K
Department of Microbiology, Annamalai University, Annamalainagar, Tamilnadu, India

The controlling of water, and in particular midseason drainage, is an effective way for reducing the methane flux from rice fields [7-9].

Azolla is a heterosporouspteridophyte with an extensive distribution in tropical aquatic ecosystems, such as ditches, swamps and lakes [10,11]. Additionally, *Azolla* can retard NH_3 volatilization by lowering the flooded water pH [when applied urea and serves as a temporary nitrogen resource [12]. Methane production, transport and oxidation in paddy soils are strongly influenced by environmental variables, including temperature, soil redox potential (Eh), rice variety, pH, fertilizer type and other factors [13-16].This present study employed to instigate the influence of fertilization on dual cropping of paddy with *Azolla* on methane emission.

MATERIALS AND METHODS

Study area

A field experiment was conducted in paddy fields in Peruvarappur village at Cuddalore district, Tamilnadu, India, in 2017. It has the average annual rainfall of 1273 mm during the last 25 years and 80% of area in this village is covered by river basin that was naturally formed by the confluence (latitude 11°41’85’’ N, longitude 79°46’70’’ E) of the rivers of Vellar and Manimuktha river, that is located at about 1 km away from Peruvarappur village and covering about 131 ha of cultivable field. The soil type of this village is classified as rich and fertile grimy clay that is appropriate for abundance rice production in confluence basin areas.

Experimental designs

In order to study the effect of fertilization and dual cropping with *Azolla* as a co-culture on methane emission, three treatments were established: 1) paddy field supplemented with organic composites (26.4% organic C, 2.5% total N, 1.6% P_2O_5 and 1.3% K_2O, made of composted farmyard manure, rice straw and green manures), 2) paddy field was supplemented with Conventional (Chemical) fertilizers (NPK inorganic fertilizer, in which 391 kg ha^{-1} urea [46% N], 750 kg ha^{-1} superphosphate and 183 kg ha^{-1} potassium chloride [mixed and pilled]) and chemical pesticides and insecticides [17] and 3) rice cropping with dual cropping of *Azolla* under recommended fertilization (NPK+*Azolla*). The treatments 1, 2 and 3 were respectively named as OFPF, and CFPF and CFPR+*Azolla.*

The *Azollacaroliniana* Willd. was allowed to grow after being inoculated into NPK+*Azolla* treatment (third treatment) at 1 Mg ha^{-1} 7 d after rice transplantation (the coverage density of *Azolla* was 0.76 kg m^{-2} when it reached full coverage above the flooded water (12 d after inoculation). The other treatments (1 and 2) were supplemented with their respective fertilizer one before paddy transplantation. On 8th- June- 2017, the fields were transplanted with 30 days old (*Oryzasativa* L. var.) nursery seedlings and harvested on 21st- August 2017. During the paddy cropping period, proper irrigation was maintained and the methane emission was measured at 10 days interval.

Measurements of CH_4 flux rates

CH_4 flux rate from three treatments were measured during the 10 days of interval up to 80th day. The Closed chamber method was used to measure the CH_4 emission as described previously [18,19]. Briefly, air gas samples were collected from square-shaped glass chambers (45 X 45 X 100 cm^3) covering five rice hills using 50-mL gas-tight syringes at 30 min after closing the top of the chamber. Gas samplings were carried out three times (8am, 12pm, 4pm) per days to obtain average CH_4 emissions. CH_4 concentrations were measured by a gas chromatograph (GC-2010, Shimadzu, Tokyo, Japan) equipped with a Porapak NQ column (Q 80–100 mesh) and a flame ionization detector. The temperatures of column, injector and detector were 100, 200 and 200 °C, respectively. Helium and hydrogen were used as carrier and burning gases, respectively. CH_4 emission rates from the rice paddies were calculated according to the following equation:

$$F = \frac{dc}{dt} \times h \times \rho \frac{273}{(273 + T)}$$

here, F is the CH_4 flux (mg m^{-2} h^{-1}), dc/dt is the slope of the curve of the gas concentration versus time, h is the headspace height (m) of the chamber, ρ is the gas density (kg m^{-3}) at standard state, and T is the air temperature (°C) inside the chamber.

During the experimental period, Total carbon (TC) concentration in soil and flooded water was determined colorimetrically by the method recommended by Nelson and Sommers, [20]. The pH of soil and flooded waterwas measured using the water sediment samples collected in triplicate at 10 day interval [21]. The soil Redox potential (Eh) of the root region (approximately 10 cm depth) was directly measured *in situ* (from six randomly picked points near rice plants for each treatment) using the depolarization method with an automatic Oxidation Reduction Potential (ORP) tester (CN61M/FJA-6, China).

Statistical analysis

Statistical analyses of the methane emission rate and abundance data were conducted using the LSD (least significant difference) test at the 0.05 probability level. All statistical analyses were performed using SPSS 17, MS-Excel.

RESULTS

Methane emission

Figure-1 showed the variations in methane emission in paddy fields. The amounts of methane emission were significantly higher in the treatment OFPF than other treatments. CFPF and CFPF+ *Azolla* treatments showed significantly similar trends in CH_4 flux. Throughout the experiment, the maximum CH_4 flux rate was observed at 40thday (26.71±0.30 mg m^{-2} h^{-1}) in OFPF treatment moreover, it was dropped up to 16.56±0.50 mg m^{-2} h^{-1} at 50th day of operation. In CFPF and CFPF+ *Azolla* treatments, the maximum CH_4 flux rate of 11.09±0.03 mg m^{-2} h^{-1} and 8.08±0.02 mg m^{-2} h^{-1} was observed respectively at 20th day of operation. From all treatments, the CH_4 flux rate was increased in vegetative and reproductive stages, and then it was decreased in maturation stage. Throughout the experiment, the maximum seasonal average of CH_4 flux 18.84±1.0 mg m^{-2} h^{-1} was found in OFPF treatment, it was 73.99% and 84.97% higher than CFPF and CFPF+ *Azolla*treatments, respectively. The results indicate that the application of *Azolla*in paddy field significantly reduced the CH_4 flux rate up to 42% than CFPF treatment.

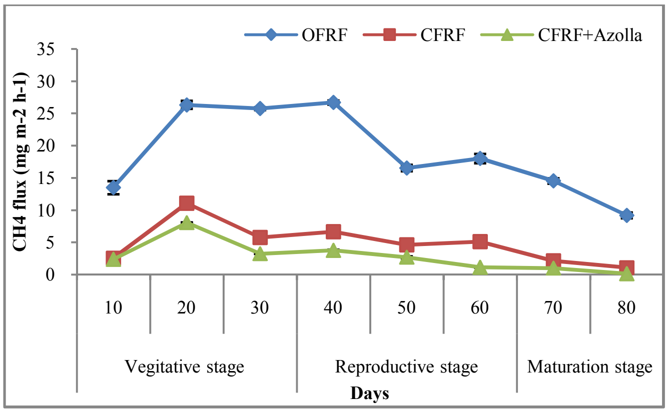

Figure 1 Methane emission rate from paddy fields

The pH of bulk soil of treatments were measured, that results showed, the pH in OFPF treatment (ranged 6.80-6.90, average: 6.88) was slightly acidic when compared to other treatments of CFPF (ranged 6.80-7.33, average: 7.07) and CFPF+*Azolla* (ranged 6.76-7.20, average: 7.01), when compared with flooded water, that did not noticed any variation in the average (6.86) in OFPF treatment. In CFPF+*Azolla* treatment, *Azolla*growth on surface of the flooded water significantly reduced the pH level.

The maximum TC concentration in soil of 29.34±0.80 g kg^{-1} was observed in OFPF treatment then it was decreased upto10.43±0.10 g kg^{-1}. The decrease in TC was directly reflected in drop of CH_4 flux rate. It indicates, the CH_4 emission positively correlated with TC concentration in bulk soil of the fields. The TC level in soil of CFPF and CFPF+*Azolla* treatments (Average, 14.82 and 14.35 g kg^{-1})

were significantly lower than the OFPF treatment. It was indicating, the soil TC level was influenced by fertilization however the *Azolla* as a co-crop did not giveany impact on TC level in soil.

in the water [26-28], which subsequently lowers the consumption of CO_2 in the flooded water and thus the pH. The low Eh in flooded soil is vital to the normal functioning of methanogens due to their anaerobic characteristics.

Table 1 Variations in Physico-chemical factors of paddy field under the different fertilization and influence of *Azolla* from day 10 to day 80th day after rice transplantation.

Treatments	Days after transplantation								
	10	20	30	40	50	60	70	80	Mean
Bulk soil pH									
OFPF	6.8±0.01	6.8±0.05	6.8±0.11	6.8±0.20	6.8±0.07	6.8±0.19	6.9±0.09	6.8±0.04	6.81
CFPF	6.8±0.03	7.1±0.56 [a]	7.1±0.08 [a]	6.9±0.08 [a]	7.1±0.05 [a]	6.9±0.02	7.3±0.06	7.1±0.05 [a]	7.07
CFPF+*Azolla*	6.7±0.02	7.0±0.07	6.8±0.08 [a]	6.8±0.12	7.2±0.10	6.9±0.12	7.2±0.09 [a]	7.1±0.06	6.96
Flooded water pH									
OFPF	6.8±0.02 [a]	6.8±0.07	6.9±0.12	6.8±0.08	6.8±0.08	7.0±0.12	6.9±0.03	6.8±0.07	6.85
CFPF	6.9±0.06	7.1±0.07	7.1±0.08 [a]	6.9±0.03 [a]	7.1±0.07	7.1±0.03 [a]	7.3±0.07	7.1±0.03 [a]	7.07
CFPF+*Azolla*	6.7±0.04	6.8±0.14	6.8±0.07 [a]	6.7±0.08	6.8±0.12	6.9±0.08 [a]	7.2±0.07 [a]	7.1±0.12	6.87
Bulk soil Total carbon (TC, g kg-[1])									
OFPF	21.7±0.13	24.4±0.37	25.4±0.65 [a]	29.3±0.96	21.7±0.34	17.2±0.45 [a]	11.2±0.52	10.4±0.09	20.1
CFPF	17.3±0.52 [a]	19.2±0.65 [a]	20.2±0.19	21.3±0.45	12.4±0.52	11.9±0.65	9.5±0.13 [a]	6.4±0.45	14.7
CFPF+*Azolla*	16.7±0.65	18.3±0.52 [a]	20.3±0.45	19.3±0.65	11.7±0.13	12.5±0.46	10.2±0.52	5.4±0.65	14.3
Flooded water Total carbon (TC, g kg-[1])									
OFPF	13.7±0.11	14.4±0.14	15.6±0.45	19.3±0.3 [a]	11.7±0.06 [a]	17.2±0.13	10.2±0.48 [a]	6.4±0.44	13.5
CFPF	4.3±0.45 [a]	5.2±0.44	5.2±0.45	4.3±0.19	3.4±0.11 [a]	4.9±0.44	2.5±0.11	1.4±0.45 [a]	3.9
CFPF+*Azolla*	11.7±0.19	12.3±0.09	10.4±0.11	11.3±.44	10.7±0.45	9.5±0.11 [a]	8.2±0.45 [a]	8.4±0.11	10.3
Bulk soil redox potential (Eh)									
OFPF	-49.9±1.4	-76.5±5.7 [a]	-87.3±1.4	-91.5±5.7 [a]	-119.4±1.4	-157.6±8.3 [a]	-142.8±5.7	-109.4±5.7	-104.3
CFPF	-56.1±1.5	-78.4±1.4	-103.8±4.5	-105.6±1.1	-134.4±5.7	-164.5±6.7 [a]	-154.8±3.8	-113.5±7.9	-113.9
CFPF+*Azolla*	-45.3±5.7 [a]	-65.6±3.7 [a]	-98.8±5.7 [a]	-99.6±1.4	-123.4±4.6	-156.6±8.6	-157.9±5.7 [a]	-111.4±5.8	-107.3

Values are expressed in the mean of 3 replications. [a] Values are presented as r with P values in the parentheses, significant differences ($P < 0.05$).

The flooded water TC level (Average 13.57 g kg-[1]) in OFPF treatment significantly lowered than the soil TC (Average 20.19 g kg-[1]). In other hand, *Azolla* as a co-crop significantly increased the flooded water TC in CFPF+*Azolla* treatment than compared to CFPF treatment. The Organic fertilization significantly increased the TC level in soil in OFPF treatment; it resulted to increase the CH_4 flux rate. The maximum average of soil Eh of -113.9 was observed in CFPF followed by CFPF+*Azolla* (-107.3) and OFPF (-104.3). The results showed the soil redox potential positively correlated with methane emission in OFPF (Table-1).

DISCUSSION

The influence of fertilization and *Azolla* as a dual crop with paddy on CH_4 flux rate was examined in this study. In order to examine the supporting factors for the CH_4 flux, the soil pH and TC were estimated throughout this experiment. The results were indicating, the organic fertilization on paddy fields (OFPF) notably increased the CH_4 flux then compared to other treatments. For this, the TC in the OFPF bulk soil acts as a supporting factor. The pH value is an environmental parameter that is correlated with CH_4 production in paddy fields and also acts as one of the key determinants [22,23]. In general, the methanogenic activity requires the optimal pH range of 6.8-7.0, and lower or higher pH could resulted with the low methanogenic activity and reduced the methane emission [24,25].

In the present study, the significant decrease in pH in bulk soil of CFPF+*Azolla* treatment than CFPF due to the culturing of *Azolla*, resulted reduction of methane emission due to the Chemical of fertilization. The pH decrease may be related to the fact that the floating *Azolla* absorbs most available solar radiation and consequently limits the photosynthesis of algae

Soils with low Eh rates are usually results with higher methane emissions [29] In addition, the oxygen is crucial to methanotrophic bacteria as an activator for methane oxidation but also very significant as an electron acceptor [30]. Compared with NPK, the decrease in the methane emission rate and cumulative emissions of NPK+*Azolla* after paddy transplantation is likely to be attributable to the depression of flooded water and bulk soil pH and the promotion root region Eh.

CONCLUSION

In the present study, we observed that, the organic fertilization in paddy field notably increased the CH_4 flux rates when compared to the other treatments. *Azolla* as a co-crop with the paddy significantly reduced the CH_4 flux rates when compared to the CFPF treatment. For minimizing the methane emission in paddy field during the cropping period of the rice, Conventional (chemical) fertilization along with *Azolla* as co-crop by dual cropping method gives significant reduction in methane emission.

Acknowledgements

The authors thank Thamizhazhagan and Balakumarfor theirvaluable support during the manuscript preparation, Devadoss for maintenance of selected agricultural fields. The authors would like to thank the analytical laboratory, Center for Advanced Study in Marine Biology, Parangipet for analytical supports.

References

1. Yagi, K., Minami, K., Ogawa, Y. Effect of water percolation on methane emission from rice paddies: a lysimeter experiment. *Plant Soil* 198: 193-200, (1998)
2. Lee, C.H.,*et al*. Effect of Chinese milk vetch (Astragalussinicus L.) as a green manure on rice productivity and methane emission in paddy soil. Agric. Ecosyst. *Environ*. 138, 343-347, (2010).

3. Sass, R.L., Fisher, F. M., Ding, A. & Huang, Y. Exchange of methane from rice fields: National, regional, and global budgets. *J. Geophys. Res*. 104, 26943 (1999).
4. Yan, X., Akiyama, H., Yagi, K., Akimoto, H. 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 (2009).
5. Montzka, S.A., Dlugokencky, E.J., Butler, J.H. Non-CO2 greenhouse gases and climate change. Nature 476, 43-50 (2011).
6. IPCC. Climate Change 2007 - The Physical Science Basis: Working Group I Contribution to the Fourth Assessment Report of the IPCC at < https://books.google.com/books?hl= zh-CN&lr= &id= 8-m8nXB8GB4C&pgis= 1> (Cambridge University Press, 2007).
7. Berger, S., Jana, I., Seo, J., Kang, H., Gebauer, G. A record of N2O and CH4 emissions and underlying soil processes of Korean rice paddies asaffected by different water management practices. *Biochemistry* 115: 317-332, (2013).
8. Hadi, A., Inubushi, K., Yagi, K. Effect of water management on greenhouse gas emissions and microbial properties of paddy soils in Japan and Indonesia. *Paddy Water Environ* 8: 319-324, (2010).
9. Nishimura, S., Sawamoto, T., Akiyama, H., Sudo., S, Yagi, K. Methane and nitrous oxide emissions from a paddy field with Japanese conventional water management and fertilizer application. *Global Biogeochem Cycles* 18, (2004).
10. Ying, Z., Boeckx, P., Chen, G. X. &Cleemput, O. Van. Influence of *Azolla* on CH 4 emission from rice fields. Nutr. Cycl. *Agroecosystems* 58, 321-326 (2000).
11. Bocchi, S., Malgioglio, A. Azolla-Anabaena as a Biofertilizer for Rice Paddy Fields in the Po Valley, a Temperate Rice Area in Northern Italy. *Int. J. Agron*. 2010, 1-5 (2010).
12. Cissé, M., Vlek, P.L.G. Influence of urea on biological N2 fixation and N transfer from Azolla intercropped with rice. *Plant Soil* 250, 105-112 (2003).
13. Kimura, M., Murase, J., Lu, Y. Carbon cycling in rice field ecosystems in the context of input, decomposition and translocation of organic materials and the fates of their end products (CO2 and CH4). *Soil Biol. Biochem*. 36, 1399-1416 (2004).
14. Liu, Y. *et al*. Evaluating oxidation-reduction properties of dissolved organic matter from Chinese milk vetch (Astragalussinicus L.): a comprehensive multi-parametric study. *Environ. Technol*. 35, 1916-27 (2014).
15. Aulakh, M.S., Wassmann, R., Bueno, C.,Rennenberg, H. Impact of root exudates of different cultivars and plant development stages of rice (Oryza sativa L.) on methane production in a paddy soil. *Plant Soil* 230, 77-86 (2001).
16. Yan, X., Yagi, K., Akiyama, H., Akimoto, H. Statistical analysis of the major variables controlling methane emission from rice fields. Glob. *Chang. Biol*. 11, 1131-1141 (2005).
17. Wang, J., Song, Y., Ma, T., Raza, W., Li, J., Howland, J.G., Huang, Q., Shen, Q. Impacts of inorganic and organic fertilization treatments on bacterial and fungal communities in a paddy soil. *Applied Soil Ecology*. 112 -42-50 (2017).
18. Ali, M.A., Lee, C.H., Kim, S.Y., Kim, P.J. Effect of industrial by-products containing electron acceptors on mitigating methane emission during rice cultivation. *Waste Manag* 29: 2759-2764(2009).
19. Lee, C.H., Park, K.D., Jung, K.Y., Ali, M.A., Lee, D., Gutierrez, J., Kim, P.J. Effect of Chinese milk vetch (Astragalussinicus L.) as a green manure on rice productivity and methane emission in paddy soil. *AgricEcosyst Environ* 138: 343-347(2010).
20. Nelson, D.W., Sommers, L.E. Total carbon, organic carbon, and organic matter. p. 961-1010. In: Black, C.A., ed. Methods of soil analysis. Part 3. Chemical methods. Soil Science of America and American Society of Agronomy, Madison, WI, USA (1996).
21. APHA, Standard methods for the examination of water and wastewater, 21st edn. American Public Health Association, Washington (2005).
22. Lauber, C.L., Hamady, M., Knight, R. Fierer, N. Pyrosequencing-Based Assessment of Soil pH as a Predictor of Soil Bacterial Community Structure at the Continental Scale. *Appl. Environ. Microbiol*. 75, 5111-5120 (2009).
23. Tripathi, B.M. *et al*. Soil pH and biome are both key determinants of soil archaeal community structure. *Soil Biol. Biochem*. 88, 1-8 (2015).
24. Ye, R. *et al*. pH controls over anaerobic carbon mineralization, the efficiency of methane production, and methanogenic pathways in peatlands across an ombrotrophic–minerotrophic gradient. *Soil Biol. Biochem*. 54, 36-47 (2012).
25. Svensson, B.H., Rosswall, T. In situ Methane Production from Acid Peat in Plant Communities with Different Moisture Regimes in a Subarctic Mire. Oikos 43, 341-350 (1984).
26. De Macale, M.A.R., Vlek, P.L.G. The role of *Azolla* cover in improving the nitrogen use efficiency of lowland rice. *Plant Soil* 263, 311-321 (2004).
27. Vlek, P.L.G., Diakite, M.Y., Mueller. H. The role of Azolla in curbing ammonia volatilization from flooded rice systems. *Fertil. Res*. 42, 165-174 (1995).
28. Tamizhazhagan and Pugazhendy. Physico - chemical parameters from the Manappadaiyur and swamimalai fresh water Ponds*, Indo Am. J. Pharm. Sci, 3(5)* 444-449(2016).
29. Bharati, K., Mohanty, S.R., Singh, D.P., Rao, V.R., Adhya, T.K. Influence of incorporation or dual cropping of Azolla on methane emission from a flooded alluvial soil planted to rice in eastern India. *Agric. Ecosyst. Environ.* 79, 73-83 (2000).
30. Conrad, R. Microbial Ecology of Methanogens and Methanotrophs. 96, 1-63 (2007).

****