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Evaluating the effect of bio-fertilizers in mitigating GHGs in puddled rice (*Oryza sativa*. L)

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Rice (Oryza sativa L.) is the important cereal food grain crop grown extensively in tropical and sub-tropical region of the world (Kumar et al., 2014). The staple food for more than 50% of the world's population is rice. Rice is raised over 114 countries and accounts for nearly 11% of the world's agricultural land. India ranks first in terms of area (45.1 M ha) and second in production (104.80 million tones) behind China. To meet the demand by 2050, India must produce roughly 140 million tonnes of rice (Statista, 2021). With 50 and 60 per cent of CH₄ and N₉O emissions coming from agriculture (Rivera JE and Chará, 2021), it is thought to be a significant source of GHGs. Methane (CH₄) and nitrous oxide are two of the main GHGs that are emitted when rice is conventionally transplanted $(N_{2}O)$. It has been determined that rice fields are a significant source of CH₄, accounting for 11% of CH₄ emissions worldwide (Smith et al., 2007). Furthermore, there are variances in the quantity of greenhouse gas emissions from various rice establishment techniques. Emission of GHGs from rice fields is very sensitive to rice management strategies. Bio-fertilizers proved to be a promising option for rice production, besides, have the advantages of lowering the methane emission in transplanted rice. Hence, a study has been conducted to quantify the GHG emission and

mitigation potential of BGA (Cyanobacteria) and Azolla in puddled rice.

The field experiment was conducted at Agricultural College & Research Institute, Madurai during samba, 2021 in C block and Field Number 47. The experimental soil was sandy clay loam in texture with alkaline pH (8.14) and EC (0.2 dSm⁻¹). Soil organic carbon was medium (0.5%) in status and soil available nitrogen was found to be low (206 Kg ha⁻¹), whereas, the available phosphorous (35.2 Kg ha⁻¹) and potassium (358 Kg ha⁻¹) were high in the experimental soil. In this study, the treatments viz, T_1 : SRI method of rice cultivation with organic farming standard of package, T2: SRI method of rice cultivation with inorganic farming standard of package SOP, T₃ : T₁+BGA application @ 10 Kg ha⁻¹, T₄ : T₂+ BGA application @ 10 Kgha⁻¹,T₅ -T₁+Azolla application @ 250 Kg ha⁻¹, $T_6 - T_2 + Azolla$ application @ 250 Kg. were arranged in Randomized Block Design with four replications using the variety, ADT 54 with a plot size of 5×4 m. Seeds were soaked with Bacillus subtilis @ 10 g, Azospirillum @ 30 g and Phosphobacteria @30 g per Kg of seeds. Well decomposed FYM @ 1.25 Kg, neem cake @ 50 g and gypsum @ 100 g per m² were applied as basal 10 days after sowing. Green manure crop (Sesbania aculeata) was raised before rice transplanting and incorporated in-situ at flowering stage.



It was followed by application of neem cake @ 250 Kg ha-1 and Gypsum @ 500 Kg ha-1 as basal. Seedlings were dipped with Azospirillum (1 Kg ha-1) + Phosphobacteria (1 Kg ha-1) in 40 liters of water for 15-30 minutes before transplanting. CH4 and N2O flux were determined using the IRGA Sensor closed-chamber technique. The closed chamber in this system contains a small infrared CO₀ gas analyzer. This system does not need air tubes or pumps for circulating air, so it is expected to offer the advantages of mobility and durability. This system was verified by a comparison with measurements made by using a closeddynamic-chamber (CDC) system. Gas sampling began at active tillering and flowering stages. The accumulated gases inside the chambers were collected using 100 mg plastic syringes after one hour of chamber closure and followed by infusion into an empty aluminum foil gas collecting bag. The sampling time was between 9:00 a.m. and 10:00 a.m. during each sampling day. The gas samples were transported to the laboratory for analysis by gas chromatography within a few hours. The concentrations of CH₄ and N₂O were analyzed with a gas chromatograph meter equipped with an electron capture detector for NoO analysis and a flame ionization detector for CH₄ analysis. The plant height was measured from the ground level to the tip of the top most fully opened leaf or flag leaf at active tillering and panicle initiation stages, while, at harvest stage, it was measured up to the tip of the panicle. The mean values were expressed in cm. Four 0.25 m⁻² quadrants were randomly placed in each net plot, and the total number of tillers was counted at tillering, panicle initiation, and harvest stages and expressed as No. m⁻². The LAI of rice was calculated at the tillering and panicle initiation stages using the formula.

Where,

- L = Maximum length of 3rd leaf blade from the top (cm)
- B = Maximum breadth of the same leaf (cm)
- K = Constant factor (0.75)
- N= Number of leaves per plant.

The number of panicle bearing tillers in each of the net plot (0.25 m⁻²) was counted at four random locations and expressed as No. m⁻². After threshing, cleaning, drying and winnowing, the grain yield from each net plot area was recorded. The final grain yield was calculated at 14 per cent moisture content and expressed in Kg ha⁻¹. The non-significant treatments were denoted by NS, and the significant treatments were calculated at 5 per cent probability level

The effect of treatments on the growth, yield parameters and yield of rice, ADT 54 was significant. Among the treatments, SRI method of rice cultivation with inorganic farming (T_{2}) has recorded significantly taller plants (133) cm), higher number of tillers plant¹ (19.8), maximum LAI (5.73), more productive tillers (328 m⁻²) and maximum grain yield (5285 Kg ha⁻¹) (Table 1). This was comparable with SRI with inorganic farming + BGA 10 Kg ha⁻¹ (T_{λ}), SRI with inorganic farming + Azolla 250 Kg ha⁻¹ and SRI with organic farming + Azolla 250 Kg ha-1. As regards SRI with organic farming practices, addition of bio-fertilizers, either Azolla or BGA had significant influence on the above said parameters. SRI with organic farming resulted in significantly lower yield (3350 Kg ha⁻¹) with less panicles m⁻² which was followed by SRI with organic farming + BGA 10 Kg ha⁻¹ and SRI with organic farming + Azolla 250 Kg ha⁻¹.

Treatment	Plant height (cm)	No.of tillers plant ⁻¹	LAI	Panicles m ⁻²	Grain yield (Kg/ha)
T ₁ - SRI with organic farming	121.3	12.8	4.80	196	3350
T_2 - SRI with inorganic farming	133.0	19.8	5.73	328	5285
$\rm T_3$ - SRI with organic farming + BGA 10 Kg ha $^{-1}$	119.8	14.5	5.10	240	3675
$\rm T_{4 -}SRI$ with inorganic farming + BGA 10 Kg $\rm ha^{\text{-}1}$	129.8	21.0	6.05	324	5387
$\rm T_5$ - SRI with organic farming + Azolla 250 Kg $\rm ha^{\cdot 1}$	124.8	16.8	4.93	268	3900
$\rm T_6^{-}$ SRI with inorganic farming + Azolla 250 Kg ha $^{\rm 1}$	125.7	17.0	6.13	348	5650
SEd	4.59	1.91	5.45	21	252
CD (<i>P=0.05</i>)	9.60	3.98	0.28	42	526

Table 1. Effect of treatments on growth, yield attributes and yield



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Methane emission was estimated during tillering and flowering stage of rice using IRGA sensor in closed chamber method. Emission of methane was less during flowering stage compared to the vegetative stage (Table 2; Fig.1). At active tillering stage, the values ranged from as low as 4.14 mg m⁻² hr⁻¹ in SRI with inorganic farming + *Azolla* 250 Kg ha⁻¹ to as high as 5.33 mg m⁻² hr⁻¹ in SRI with organic farming. This was due to liberation of photosynthetic oxygen in paddy water by *Azolla* and BGA (Malyan *et al.* 2016) which increased the dissolved oxygen concentration in flooded water, and eventually decreased the CH_4 emission from paddy soil by enhancing the CH_4 oxidation (Ali *et al.* 2015) and Malyan *et al.* (2019) observed that application of Azolla along with reduced dose of N fertilizer lowered the GHG intensity in rice by 16 to 19%.

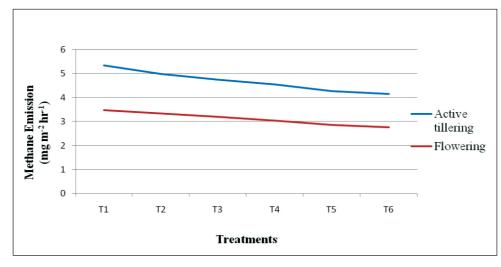


Fig 1. Effect of treatments on methane emission during active tillering and flowering stage

Treatment –	Methane Emission (mg m ⁻² hr ⁻¹)		Available Nutrients (Kg ha ⁻¹)			
	Active Tillering	Flowering	SOC (g kg ⁻¹)	N	Р	K
T_1 - SRI with organic farming	5.33	3.47	5.6	206	36.7	360
T ₂ -SRI with inorganic farming	4.97	3.34	5.4	204	34.4	362
T_{3} - SRI with organic farming + BGA 10 Kg ha ⁻¹	4.74	3.19	5.7	212	38.4	363
$T_{4-}SRI$ with inorganic farming + BGA 10 Kg ha ⁻¹	4.54	3.04	5.6	208	36.3	364
$\rm T_5$ - SRI with organic farming + Azolla 250 Kg ha $^{\rm 1}$	4.25	2.86	5.9	219	38.5	363
$\rm T_6$ – SRI with inorganic farming + Azolla 250 Kg $\rm ha^{\textsc{-1}}$	4.14	2.77	5.6	214	37.9	366
SEd	0.18	0.10	0.09	2.4	0.6	3.1
CD (P=0.05)	0.55	0.33	0.28	7.1	1.9	NS

Table 2. Effect of treatments on methane emission and soil nutrient status

Application of *Azolla* significantly reduced the methane emission irrespective of organic and inorganic nutrient management. As regards BGA, there was no significant influence on methane emission when it was added to organic or inorganic practices. The similar trend followed during the flowering stage also. Rose *et al.* (2014) reported that the bio-fertilizer containing plant growth promoting microorganisms could replace between 23 and 52 % of nitrogen (N) fertilizer without loss of yield. Ali *et al.* (2014) also reported that *Anabaena azollae* in combination with

urea and silicate fertilization decreased the total seasonal CH $_4$ flux by 12 % and increased rice grain yield by 10.6 %.

Adoption of organic nutrient management practices, *Azolla* @ 250 Kg ha⁻¹ recorded higher soil organic carbon of 5.9 g Kg⁻¹, which was statistically comparable with the same organic combination with BGA @ 10 Kg ha⁻¹. With regard to soil available nitrogen and phosphorous, the same combination registered higher values (219 & 38.5 Kg ha⁻¹) which was statistically comparable with other



organic nutrient management practices with bio-fertilizer combinations and inorganic nutrient management practice with *Azolla* 250 Kg ha⁻¹. The soil available potassium did not show any variation among the nutrient management practices.

The cost of cultivation was comparatively higher (Rs. 60000 ha⁻¹) under organic farming practices which may be due to the higher cost of organic inputs. The gross return was high (Rs. 118650 ha⁻¹) in SRI with inorganic farming + *Azolla* 250 Kg ha⁻¹ due to the higher grain yield which was reflected in the BCR too (Table 3).

Treatments	Cost of cultivation (Rs ha-1)	Gross Return (Rs ha-1)	B:C
T ₁ - SRI with organic farming	52000	70350	1.35
T ₂ -SRI with inorganic farming	46500	110985	2.39
$\rm T_3$ - SRI with organic farming + BGA 10 Kg $\rm ha^{-1}$	56000	77175	1.38
T_{4-} SRI with inorganic farming + BGA 10 Kg ha ⁻¹	48000	113140	2.36
$\rm T_5^{ -}$ SRI with organic farming + Azolla 250 Kg $\rm ha^{-1}$	60000	81900	1.37
$\rm T_6\mathchar`-SRI$ with inorganic farming + Azolla 250 Kg ha 1	50000	118650	2.37

Table 3. Effect of treatments on cost economics and BCR

Grain yield of ADT 54 was higher in SRI with inorganic farming + *Azolla* 250 Kg ha⁻¹ with very less methane emission during active tillering and flowering stages with higher post harvest soil fertility status. However, the B:C Ratio was higher at SRI with inorganic farming and it was closely followed by SRI with inorganic farming + Azolla 250 Kg ha⁻¹. Hence, SRI with inorganic farming + Azolla 250 Kg ha⁻¹ may be recommended for getting higher yield, reduced methane emission, higher post harvest soil fertility status and higher BCR for rice variety, ADT 54.

Author contributions

All the authors contributed to the article and approved the submitted version.

Compliance with ethical standards

Yes

Conflict of interests

No commercial or financial relationships that could be construed as a potential conflict of interest.

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