

**EFFECT OF TEMPERATURE, REDOX AND DISSOLVED OXYGEN  
ON METHANE FLUX OF RICE SOIL ECOSYSTEM AND DESIGNING  
MITIGATION STRATEGIES TO MINIMIZE GLOBAL WARMING  
POTENTIAL**

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**2011**

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*Thesis submitted in part fulfillment of the requirement for the Degree of  
MASTER OF SCIENCE IN AGRICULTURAL METEOROLOGY to the  
Tamil Nadu Agricultural University, Coimbatore*

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## **CERTIFICATE**

This is to certify that the thesis entitled "**EFFECT OF TEMPERATURE, REDOX AND DISSOLVED OXYGEN ON METHANE FLUX OF RICE SOIL ECOSYSTEM AND DESIGNING MITIGATION STRATEGIES TO MINIMIZE GLOBAL WARMING POTENTIAL**" submitted in part fulfillment of the requirement for the award of the degree of **MASTER OF SCIENCE IN AGRICULTURAL METEOROLOGY** to the Tamil Nadu Agricultural University, Coimbatore is a record of bonafide research carried out by **Mr. A. SANKAR** under my supervision and guidance and that no part of this thesis has been submitted for the award of other degree, diploma, fellowship or other similar titles or prizes and that the work has not been published in part or full in any scientific or popular journal or magazine.

Place: Coimbatore  
Date : 28.06.2011

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## ABSTRACT

### EFFECT OF TEMPERATURE, REDOX AND DISSOLVED OXYGEN ON METHANE FLUX OF RICE SOIL ECOSYSTEM AND DESIGNING MITIGATION STRATEGIES TO MINIMIZE GLOBAL WARMING POTENTIAL

By  
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Rice fields have to be considered as a significant source of greenhouse gases and rice field eco systems account for about 60 Tg methane per year or about 12 per cent of the global annual methane emission (IPCC, 1996). The emission of green house gases from rice ecosystem is likely to increase in the days to come due to intensification of rice cultivation. Temperature, redox potential, dissolved oxygen, organics and seasonal variations are among the factors that influence production of methane in soil. Recently, it has been reported that growing Cyanobacteria and *Azolla* in rice fields could reduce methane flux. Keeping in view the above facts, the present investigation was carried out to assess the methane emission from the rice cultivation under different organic manure and exploring the correlation between methane flux and air/soil temperature. As climate change mitigation strategy, the potential of Blue green algae and *Azolla* in minimizing methane flux at source in paddy field ecosystem has also been investigated.

The present investigation was conducted at Anbil Dharmalingam Agricultural College and Research Institute, Trichy, during the *Rabi* seasons of 2010-11. It is well known that temperature strongly affects methanogenesis and increasing temperature leads to higher methane emission. In the present investigation, the marginal reduction in soil and water temperature in BGA and *Azolla* applied plots as a result of higher oxygen diffusion would be one among the factors that contributed to low methane flux. The air

temperature surrounding the top of the rice plant had relatively small effect on conductance of methane in to atmosphere by rice plants. Interestingly the higher methane flux in all treatments was recorded during 51 and 52 standard weeks (60-75 DAT), that also registered high mean maximum and minimum air temperature.

Redox status of soil and dissolved oxygen content in the standing water are indirect indicators of methane flux pattern from rice ecosystem. The combined application of BGA and *Azolla* recorded higher redox potential followed by the application of BGA and *Azolla* individually in all growth stages of rice in the experimental plots. Similarly the BGA and *Azolla* application individually and in combination enhanced the dissolved oxygen concentration in the standing water in all growth stages while the dissolved oxygen concentration was minimum in farm yard manure and green leaf manure applied plots. BGA and *Azolla* application in rice cultivation as biofertilizers minimize methane flux by enhancing the soil redox and dissolved oxygen that are unfavorable to methane generating methanogens. The methane fluxes were maximum during flowering stage in all treatments and showed decreasing trend towards maturity. The higher rates of CH<sub>4</sub> production during the flowering stage were due to the degradation of the available organic carbon in the form of root exudates. The methane emission was found to be maximum in farmyard manure and green leaf manure applied plots in all growth stages. Blue green algal and *Azolla* application recorded the lowest methane flux in all growth stages. The higher dissolved oxygen and redox values recorded in algal and *Azolla* applied plots resulted in low methane emission values.

In the present study, combined application of organics and blue green algae not only recorded higher yield, but found to emit less methane in paddy cultivation than the application of organics alone. The mean methane flux in farm yard manure and green leaf manure applied plot was 58.54 mg m<sup>2</sup> day<sup>-1</sup>, while the flux was reduced to 20 per cent due to BGA and *Azolla* application (46.37 mg m<sup>2</sup> day<sup>-1</sup>). The present field study reiterates that biofertilization of paddy fields with blue green algae and *Azolla* is a potential climate change mitigation strategy due to their effect in minimizing methane emission, besides yield enhancement by nitrogen fixation.

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## LIST OF ABBREVIATIONS USED

%	Per cent
@	at the rate of
°C	Degree Celsius
BGA	Blue green algae
CD	Critical Difference
CDZ	Cauvery delta zone
CH <sub>4</sub>	Methane
cm	Centimeter
CO <sub>2</sub>	Carbon dioxide
CO <sub>2</sub> e	Carbon dioxide equivalent
DAT	Day after transplanting
dS m <sup>-1</sup>	Deci Siemens per metre
E longitude	East longitude
EC	Electrical conductivity
EF	Emission factor
Eh	Redox potential
FAO	Food and Agriculture Organization of the United Nations
Fig.	Figure
FYM	Farm yard manure
g kg <sup>-1</sup>	Gram per kilogram
g	Gram
GHG	Greenhouse gas
GLM	Green leaf manure
GWP	Global Warming Potential
ha	Hectare

ha <sup>-1</sup>	Per hectare
hrs	Hours
IPCC	Intergovernmental Panel on Climate Change
IRRI	International Rice Research Institute
K	Potassium
kg ha <sup>-1</sup>	Kilogram per hectare
kg	Kilogram
km <sup>-2</sup> yr <sup>-1</sup>	kilometer square per year
m	Meter
m <sup>-2</sup>	Per meter square
mg l <sup>-1</sup>	Milligram per liter
mg m <sup>-2</sup> day <sup>-1</sup>	Million gram per meter square per day
mg	Milli gram
min	Minutes
mm	Millimeter
MMTCO <sub>2</sub> E	Million metric tonnes of CO <sub>2</sub> equivalent
MOB	Microbial oxidizing bacteria
mt/yr	Million tonnes per year
mV	Millivolt
N latitude	North latitude
N	Nitrogen
N <sub>2</sub> O	Nitrous Oxide
NS	Non significant
OC	Organic Carbon
P	Phosphorus
panicle <sup>-1</sup>	per panicle

pH	Hydrogen ion concentration
ppb	parts per billion
ppm	parts per million
SEd	Standard Deviation
t	Tonnes
Tg	Teragram
US-EPA	Unite States Environmental Protection Agency
Yr <sup>-1</sup>	Per year

## CHAPTER 1

### INTRODUCTION

Climate change has many facets, which include changes in long-term trends in temperature and rainfall regimes with increasing year-to-year variability and a greater prevalence of extreme events. Global warming induced by increasing concentration of greenhouse gases (GHGs) in the atmosphere is a matter of great environmental concern. Methane (CH<sub>4</sub>), carbon dioxide (CO<sub>2</sub>), nitrous oxide (N<sub>2</sub>O) and chlorofluorocarbon are the GHGs which have strong infrared absorption bands and trap part of the thermal radiation from the earth surface (Rasmussen and Khalil, 1986; Battle *et al.*, 1996). In past years, concerns have been expressed all over the world over rising levels of CH<sub>4</sub> @ 0.7 to 1.1 per cent year<sup>-1</sup> because of its role in global climate change (Rasmussen and Khalil, 1981; Scheel *et al.*, 1989).

According to the Agriculture chapter of the IPCC Fourth Assessment Report (Smith *et al.*, 2007), approximately 40-50 per cent of the Earth's surface is managed for agricultural purposes and contributes 10-12 per cent of global GHG emissions, which was around 5.1-6.1 Pg CO<sub>2</sub>-eq yr<sup>-1</sup> in 2005. This is made up of 3.3 Pg CO<sub>2</sub>-eq yr<sup>-1</sup> from CH<sub>4</sub> and 2.8 Pg CO<sub>2</sub>-eq yr<sup>-1</sup> from N<sub>2</sub>O emissions. If indirect emissions from agrochemical production, distribution and on-farm operations are also included, an extra 0.4-1.6 Pg CO<sub>2</sub>-eq yr<sup>-1</sup> (0.8-3.2 %) can be attributed to agriculture, meaning that in total direct and indirect emissions from agricultural activity and land use change to agricultural use could contribute as much as 32.2 per cent of all GHG emissions (Bellarby *et al.*, 2008).

Methane gas is the most abundant organic species of the atmosphere, primarily a biogenic gas produced in anoxic sediments by the methanogenic bacteria, as the terminal product of organic carbon mineralization (Zehnder, 1978). Methane is the most important component of the Global Warming Potential (GWP) of rice production due to the interactive nature of carbon and nitrogen cycles in rice fields. Once methane is generated in the submerged soils of rice-fields, it escapes to the troposphere through ebullition, molecular diffusion and plant mediated transport. About 90 per cent of CH<sub>4</sub> produced in the sediment is transported through rice plants, as they have well developed aerenchyma to serve as conduits for supply of atmospheric O<sub>2</sub> to roots for respiration and release of CH<sub>4</sub> to the troposphere (Neue and Sass, 1994).

Methane production and consumption in soil are the biological-mediated processes and therefore influenced by the prevalent weather condition, water regime, soil properties and various cultural practices like irrigation and drainage, organic amendments, fertilization and rice cultivars. Temperature, irrigation, redox potential, fertilization, available carbon and seasonal variations are among the factors that influence production of methane in soil (Allen *et al.*, 2003; Lindau *et al.*, 1990; Yagi and Minami, 1990). While the organic carbon from the rice soil and organic amendments serve as sources for CH<sub>4</sub> production, several other factors limit methanogenesis in various ways in wetland rice-fields. Methane production is negatively correlated with soil redox potential and positively correlated with soil temperature, soil carbon content and rice growth. Temperature plays an important role in the activity of soil microorganisms, including those involved in methane emission. The correlation between the diurnal variations in the CH<sub>4</sub> emission rate and air or soil temperature has been reported by many researchers (Seiler *et al.*, 1984; Schutz *et al.*, 1989a). Temperature is a major factor causing the seasonal variation in the CH<sub>4</sub> emission rate during continuous flooding, although the development of reducing conditions and rice growth also affect seasonal variation (Khalil *et al.*, 1998; Watanabe *et al.*, 2001).

Minimizing the methane emission from rice cultivation is considered as an important climate change mitigation strategy. In the global CH<sub>4</sub> cycle, biological processes consume substantial amount of CH<sub>4</sub>. The only known biological sink for atmospheric CH<sub>4</sub> is its oxidation in aerobic soils by methanotrophs or Methane-Oxidizing Bacteria (MOB), which can contribute up to 15 per cent to the total global CH<sub>4</sub> destruction. The photosynthetic systems such as Blue Green Algae (BGA) and Azolla are also known to minimize the global warming potential in flooded rice by enhancing the dissolved oxygen content in the soil water interface that ultimately suppresses the activity of methanogens. These nitrogen fixing biological systems can be used to reduce methane flux from flooded rice ecosystem besides their ability in supplementing nitrogen to the rice crop (Lakshmanan *et al.*, 2010).

Keeping in view the above facts, the present investigation entitled **“Effect of temperature, redox and dissolved oxygen on methane flux of rice soil ecosystem and**

**designing mitigation strategies to minimize global warming potential**” was carried out with the following objectives.

- Assessing the methane emission from the rice cultivation in sodic soil of Cauvery delta zone (CDZ) under different organic manure and exploring the correlation between methane flux and air/soil temperature.
- Assessing the role of photosynthetic microbial systems such as Blue green algae (BGA) and *Azolla* on rice soil redox and dissolved oxygen contents.
- Assessing the potentials of Blue green algae and *Azolla* in minimizing methane flux at source in paddy field ecosystem as climate change mitigation strategy.

## CHAPTER II

### REVIEW OF LITERATURE

Globally rice production is estimated to have contributed 44 per cent of agricultural methane (CH<sub>4</sub>) emissions in 2000 and 16 per cent of total non-CO<sub>2</sub> agricultural emissions on a CO<sub>2</sub> equivalent basis (Verge *et al.*, 2009). In 2005, 97 per cent of emissions from rice cultivation were from developing countries and South and East Asia were responsible for 82 per cent of this, as it is a dominant food source in this region (US-EPA, 2006). They also estimated that Asia as a whole contributed 82 per cent of CH<sub>4</sub> emissions from rice in 2000, using a global EF of  $2.77 \times 10^{-5}$  Tg CH<sub>4</sub> km<sup>-2</sup> yr<sup>-1</sup> (calculated according to Mosier *et al.*, 1998) to calculate total emissions from the sector from the total area under rice cultivation from the FAO (2004) database.

Methane (CH<sub>4</sub>) is one of the important greenhouse gases that absorb infrared radiation and increases global mean surface temperature. Methane is present at about 1774±1.8 ppb in the atmosphere (IPCC, 2007). It is primarily a biogenic gas produced in anoxic sediments by the methanogenic bacteria, as the terminal product of organic carbon mineralization (Zehnder, 1978). Over 50 per cent of the global annual methane emission is of anthropogenic origin and the cultivation of irrigated rice accounts for up to 12 per cent of this efflux (IPCC, 2007). The submerged conditions in rice paddies and the rice plants are actively involved in methane production and transport. The submerged rice fields, characterized by oxygen depletion due to high moisture and relatively high organic substrate levels offer an ideal environment for the activity of methanogenic bacteria.

The increasing concentrations of trace gases such as carbon dioxide, methane and nitrous oxide in the earth's atmosphere are of global concern because of their potential influence on atmospheric chemistry and climate pattern (Houghton *et al.*, 1995). Studies conducted between 1978 and 1988 indicate that atmospheric CH<sub>4</sub> concentration is increasing at about 1.0 per cent yr<sup>-1</sup> (Crutzen, 1991). However, the recent trend in atmospheric CH<sub>4</sub> concentration shows that the rate of increase has slowed down, with an annual increase of 0.7 per cent (Steele *et al.*, 1992; Khalil and Rasmussen, 1993). The anthropogenic sources of CH<sub>4</sub> include rice fields, domestic ruminants, biomass burning, landfills, coal mining, oil and natural gas flaring, animal wastes and domestic sewage

(Crutzen, 1991; Khalil and Rasmussen, 1981). Rice fields alone may account for about 15–20 per cent of global atmospheric CH<sub>4</sub> budget (US-EPA, 1990; Minami and Neue, 1994). Currently, global research is intended at estimating the source strength of rice fields and identifying as well as developing mitigation technologies for CH<sub>4</sub> emission from flooded rice fields.

Methane emissions from rice fields are determined mainly by water regime and organic inputs but they are also influenced by soil type, weather, tillage management, chemical and biofertilizers that are reviewed briefly in this chapter.

## **2.1. GLOBAL RICE PRODUCTION**

Rice is the most economically important food crop in many developing countries providing two thirds of the calorie intake of more than 3 billion people in Asia and one third of the calorie intake of nearly 1.5 billion people in Africa and Latin America (FAO, 1995). Recently, in several developed countries such as North America and European Union, rice consumption has increased due to food diversification and immigration.

Rice is grown on more than 140 million hectares worldwide and is the most heavily consumed staple food on earth. Ninety per cent of the world's rice is produced and consumed in Asia and 90 per cent of rice land is at least temporarily flooded. The unique semi-aquatic nature of the rice plant allows it to grow productively in places no other crop could exist, but it is also the reason for its emissions of the major greenhouse gas, methane. Rice is one of the most important cereal crops supplying 20 per cent of the total food calories in the world (IRRI, 2005). Generally, rice cultivation is sustainably carried out in flooded paddy fields because the flooded water washes out substances harmful to rice growth, supplies micronutrients and weakens the activities of pathogenic bacteria and fungi with the reductive conditions of the soil. However, the strict reductive conditions cause CH<sub>4</sub> production by methanogens. Because of the demand for food production to feed the increasing human population, paddy fields will continue to be one of the most important sources of global CH<sub>4</sub> emission.

According to the Food and Agriculture Organization (FAO, 2010) of the United Nations, 80 per cent of the world rice production comes from 7 countries. India is the second major rice producing (26%) country in the world after China (32.7%).

## 2.2. GLOBAL WARMING POTENTIAL OF GREEN HOUSE GASES

The Global Warming Potential (GWP) of a greenhouse gas is the ratio of global warming, or radiative forcing both direct and indirect from one unit mass of a greenhouse gas to that of one unit mass of carbon dioxide over a period of time. Hence this is a measure of the potential for global warming per unit mass relative to carbon dioxide.

### Global Warming Potential (GWP) of major greenhouse gases.

Gas	Lifetime (years)	Global Warming Potential (Time Horizon in Years)		
		20 Yrs	100 Yrs	200 Yrs
Carbon Dioxide CO <sub>2</sub>		1	1	1
Methane CH <sub>4</sub>	12.0	62	25	7
Nitrous Oxide N <sub>2</sub> O	114	275	298	156

Different gases have different Global Warming Potential (GWP):

The potency of a greenhouse gas is referred to as its global warming potential.

The common unit is referred to as a carbon dioxide equivalent or CO<sub>2</sub>e.

Carbon dioxide (CO<sub>2</sub>) = 1 CO<sub>2</sub>e

Methane (CH<sub>4</sub>) = 25 CO<sub>2</sub>e

Nitrous oxide (N<sub>2</sub>O) = 298 CO<sub>2</sub>e

To convert tons of methane to CO<sub>2</sub>e, simply multiply by 25. (Forster *et al.*, 2007)

Most of the global emissions come from the combustion of fossil fuels releasing greenhouse gas to the atmosphere. Agricultural emissions come from other greenhouse gases, namely methane (CH<sub>4</sub>) and nitrous oxide (N<sub>2</sub>O) in addition to CO<sub>2</sub>. While CH<sub>4</sub> and N<sub>2</sub>O emissions are far less in quantity in the atmosphere; they have a much more potent impact on the climate. The characteristics of heat retention by CH<sub>4</sub> in the atmosphere is higher than that of carbonic gas (CO<sub>2</sub>) and thus lead the Kyoto Protocol to establish an equivalence between them by a factor of 21 in mass (1 ton of CH<sub>4</sub> is equivalent to 21 tonnes of CO<sub>2</sub>) as regards greenhouse effect generation.

Methane in the earth's atmosphere is an important greenhouse gas with a global warming potential of 25 times when compared to CO<sub>2</sub> over a 100 year period although accepted figures probably represents an underestimate (Shindell *et al.*, 2009). Methane emission from the rice reached 6.15 Tg yr<sup>-1</sup>, accounting for 18 per cent of the national CH<sub>4</sub> emission in 1994. Compared to CO<sub>2</sub>, methane is relatively short-lived and its atmospheric perturbation lifetime is 12 years (IPCC 2007).

Major anthropogenic methane sources in 2005 included enteric fermentation of ruminants (30%) of anthropogenic CH<sub>4</sub> emissions, natural gas and oil systems (18%), landfills (12%), wetland rice cultivation (10%), wastewater (9%), coal mining (6%) and livestock manure (4%) (US-EPA, 2006). The agriculture production (ruminant livestock, manures and rice grown under flooded conditions) currently accounts for about half of global anthropogenic methane emissions.

Methane is generated when organic matter decays in anaerobic conditions. Natural methane sources include wetlands, termites, oceans and gas hydrates (Milich, 1999). Recently, Keppler *et al.* (2006) have suggested large-scale methanogenesis by plants in aerobic conditions. It has been calculated that plants could account for up to 45 per cent of global methane emissions.

Looking at emission trends, CH<sub>4</sub> emissions and atmospheric concentrations have increased markedly since pre-industrial times. Atmospheric concentrations of CH<sub>4</sub> increased from pre-industrial values of about 715 ppb to about 1774 ppb in 2005 and exceed by far the natural range over the last 6,50,000 years (IPCC, 2007). Bousquet *et al.* (2006) reported rising of anthropogenic methane emissions since 1999 while these emissions were decreased in the 1990s. The latest increased in anthropogenic emissions has been masked by a coincident decrease in natural CH<sub>4</sub> emissions, mostly from wetlands and also reported that very large fluctuations in the growth rate of atmospheric methane concentrations from one year to the next. The inter-annual variability seems to be dominated by wetland methane emission over periods. US-EPA (2006) projected an increasing global anthropogenic emission trend until 2020.

Annual CH<sub>4</sub> emission, estimated from the analysis of air trapped in polar ice were 180 Tg year<sup>-1</sup> during the 15th century (1 Tg = 10<sup>12</sup> g) and 200 Tg year<sup>-1</sup> at the beginning

of the 18th century. The recent estimates of the International Panel for Climate Changes (IPCC) are around 300 Tg in 2000 and between 400 and 600 Tg in 2010.

IPCC (2001) reported that the earth's atmospheric methane concentration has increased by about 150 per cent since 1750 and it accounts for 20 per cent of the total radiative forcing from all of the long-lived and globally mixed greenhouse gases.

### **2.3. METHANE AS GREENHOUSE GAS (GHG)**

Global anthropogenic methane emissions are projected to increase by 15 per cent to 7,904 million metric tonnes carbon dioxide equivalent (MMTCO<sub>2</sub>E) by 2020. From 2010 to 2020, the relative contributions of the agriculture, coal mining and landfill sectors are projected to remain relatively constant, changing by less than 1 per cent of global anthropogenic methane.

In terms of total amount of production worldwide, the scientist's first guesses were between 60 and 240 million tonnes of methane per year. About 10 to 30 per cent of present annual methane production comes from plants. The evidence of direct methane emissions from plants also explains the unexpectedly high methane concentrations over tropical forests, measured only recently via satellite by a research group from the University of Heidelberg (<http://news.mongabay.com/2006/0111-mpg.html>).

The increase of methane concentration has been identified as the second largest contributor to global warming. United States Environmental Protection Agency (US-EPA) estimated that CH<sub>4</sub> would be responsible for 22.9 per cent of the observed effect of the total concentration accumulated since the pre-industrial era until today.

Methane is a hydrocarbon and the primary component of natural gas. Methane is also a potent and abundant greenhouse gas, which makes it a significant contributor to climate change, especially in the near term (10-15 years). Methane is emitted during the production and transport of coal, natural gas and oil. Emissions also result from livestock and other agricultural practices and from the decay of organic waste in municipal solid waste landfills and certain wastewater treatment systems. Global anthropogenic methane emissions for 2010 were estimated at 6,875 million metric tonnes of CO<sub>2</sub> equivalent (MMTCO<sub>2</sub>E). Approximately 50 per cent of these emissions come from the five sources

targeted by the Global Methane Initiative (GMI): agriculture, coal mines, landfills, oil and natural gas systems and wastewater.

### **2.3.1. Sources of methane**

Agricultural sources alone produce 3,290 million metric tons of carbon dioxide (CO<sub>2</sub>) equivalents or 51 per cent of worldwide anthropogenic methane emissions (US-EPA, 2006). The Intergovernmental Panel on Climate Change (IPCC) included some of agricultural categories in greenhouse gas inventories like enteric fermentation, manure management, rice cultivation, agricultural soils, prescribed burning of savannas and field burning of agricultural residues. The majority (59 %) of methane emissions from agriculture are generated from enteric fermentation from live stock and farm animals. Rice cultivation is the second largest source of agricultural methane, producing 20 per cent of agricultural methane emissions. Manure management produces 7 per cent of worldwide agricultural methane emissions and other agricultural sources produce 14 per cent.

Methane emissions from domestic ruminants are estimated to be about 80 Mt/yr, with a range of 65–100 Mt/yr (IPCC, 1992; Hogan, 1993). He also reported that Cattle and buffalo account for about 80 per cent of the global annual CH<sub>4</sub> emissions from domestic livestock. Non-ruminant livestock make a relatively small contribution (Crutzen *et al.*, 1986; Gibbs and Leng, 1993; Johnson *et al.*, 1993).

#### ***2.3.1.1. Methane emissions from Indian paddy fields***

Rice cultivation is the second largest contributor of global agricultural methane after enteric fermentation. Global methane emissions from rice cultivation are expected to rise by 22 per cent by 2020 from 2000 levels, according to the US-EPA (US-EPA, 2006). According to the International Rice Research Institute (IRRI), the world will need nearly 700 million tons of rice annually (almost 10 per cent above current production) to meet expected consumption rates in 2015. Rice cultivation generally occurs in flooded paddy fields. The flooding of fields leads to anaerobic conditions where degradation of organic matter by methanogenic bacteria produces methane. It is released from submerged soils to the atmosphere by diffusion and ebullition and through the roots and stems of rice plants (Neue, 1993).

India has 42.3m ha (28.55 per cent of the global paddy area) under paddy cultivation which is the largest area at global level. Estimation of methane emission from Indian paddy fields, therefore acquires a special significance.

### ***2.3.1.2. Methane emissions from flooded soils***

Methane is produced as the terminal step of the anaerobic breakdown of organic matter in wetland rice soils. Methane is exclusively produced by methanogenic bacteria that can metabolize only in the strict absence of free oxygen and at redox potentials of less than  $-150$  mV (Wang, 1986). Most methanogens are neutrophilic, with an optimal pH of 6-8. Methanogens rely on a plethora of other microorganisms to provide them with the few substrates they can catabolize hydrogen, carbon dioxide, formate, acetate, methanol, methylamines and methy sulfides (Conrad, 1989 and Garcia, 1990). In wetland rice soils, methane is largely produced by transmethylation of acetic acid and to some extent by the reduction of carbon dioxide (Takai, 1970).

The formation of methane is preceded by the production of volatile acids. On flooding, short-term evolution of hydrogen immediately follows the disappearance of oxygen, carbon dioxide increases and with decreasing carbon dioxide, methane formation increases (Neue and Scharpenseel, 1984; Takai *et al.*, 1956). The delay of methane production depends on the pattern of soil reduction, pH, substrate availability and temperature. In tropical flooded rice soils, where soil temperatures are 25-30°C, methane production in alkaline and calcareous soils may start hours after flooding, in neutral soils it is delayed two to three weeks and in acid soils methane may only be formed five or more weeks after flooding. Methane production is negatively correlated with soil-redox potential and positively correlated with soil temperature, soil carbon content and rice growth (Neue and Roger, 1994).

The rate and pattern of organic matter addition and decomposition determine the rate and pattern of methane formation. Methane production generally increases during the cropping season although the population density of methanogens remains fairly stable (Schutz *et al.*, 1989). Easily degradable crop residues, fallow weeds and soil organic matter are the major source for initial methane production. At later growth stages of rice, root exudates, decaying roots and aquatic biomass seem to be more important. Methane production is enhanced in the rooted soil zones (Sass *et al.*, 1991).

### **2.3.2. Factors regulating methane emission**

Methane production and consumption in soil are the biological-mediated processes and therefore influenced by the prevalent weather condition, water regime, soil properties and various cultural practices like irrigation and drainage, organic amendments, fertilization and rice cultivars. Temperature, irrigation, redox potential, fertilization, available carbon and seasonal variations are among the factors that influence production of methane in soil (Allen *et al.*, 2003; Lindau *et al.*, 1990; Yagi and Minami, 1990). The factors are discussed below individually.

#### **2.3.2.1. Temperature**

Temperature plays an important role in the activity of soil microorganisms, including those involved in methane emission. Watanabe *et al.* (2005) reported that temperature is a major factor causing seasonal variation in CH<sub>4</sub> emission rates during continuous flooding and that higher cumulative temperature leads to higher total CH<sub>4</sub> emissions.

The temperature influences methane production by regulating (i) anaerobic carbon mineralization and availability of alternative electron acceptors and (ii) methanogenic activity. At a higher temperature mineralization increases and more carbon substrate becomes available, resulting in a faster depletion of the alternative electron acceptor pool. However, the influence of temperature on methane production is mainly through its effect on methanogenic activity. Most of the methanogenic bacteria display optimum rates of methane production at around 30 - 35°C with very little methanogenesis between 5 and 15°C (Yagi *et al.*, 1997). The methane emission rates increase sharply when the soil temperature rises from 20 to 25°C, corresponding with a temperature coefficient (Q<sub>10</sub>) of about 4 (Holzapfel-Pschorn and Seiler, 1986).

Allen *et al.* (2003) suggested that elevated CO<sub>2</sub> and higher temperatures increased CH<sub>4</sub> emissions in flooded rice soils due to greater root exudation or root sloughing mediated by increased seasonal total photosynthetic CO<sub>2</sub> uptake. However, no quantitative relationship between seasonal CH<sub>4</sub> emission and temperature was found in field experiments that monitored CH<sub>4</sub> emission for more than three years from the same plots (Schutz *et al.*, 1989; Wassmann *et al.*, 1993; Nugroho *et al.*, 1996).

The diurnal variation in the CH<sub>4</sub> emission rate is mainly controlled by temperature, as other factors significantly affecting CH<sub>4</sub> emission do not change markedly in such a short period. The correlation between the diurnal variations in the CH<sub>4</sub> emission rate and air or soil temperature has been reported by many researchers (Seiler *et al.*, 1984; Schutz *et al.*, 1989a). Temperature is also a major factor causing the seasonal variation in the CH<sub>4</sub> emission rate during continuous flooding, although the development of reducing conditions and rice growth also affect seasonal variation (Khalil *et al.*, 1998; Watanabe *et al.*, 2001).

### **2.3.2.2. Water regime**

The water regime of soil is an important factor for the gas exchange between soil and atmosphere and has a direct impact on the processes involved in methane emission. For methanogenesis to take place, it is of primary importance that the soils should have enough moisture to create an anoxic condition. Drainage is a major modifier of seasonal methane emission pattern. A single mid-season drainage may reduce seasonal methane emission. This emission could be reduced further by intermittent irrigation yielding a 30 per cent reduction as compared to mid-season drainage (Lu *et al.*, 2000).

Water regime has been recognized as one of the most important practices that affect CH<sub>4</sub> emission in rice production. Much research has focused on CH<sub>4</sub> and N<sub>2</sub>O emissions in rice paddies under various agricultural managements, including water regime, synthetic and organic fertilizer application etc. (Cai *et al.*, 1997; Khalil *et al.*, 1998; Zheng *et al.*, 1999).

An episode of midseason drainage for 7-10 days rather than continuous flooding is commonly adopted in China to inhibit ineffective tillers, remove toxic substances and improve roots activities (Gao *et al.*, 1992). Thus, an intermittent flooding practice is very efficient in reducing methane emission without a significant effect on grain yield. The percolating water also transports organic solutes and dissolved gases into the subsoil or groundwater where leached methane may be oxidized or released to the atmosphere. In a four year study in northern India, it has been observed that low emissions are indirectly caused by high percolation rates of the soil and the frequent water replenishments cause constant inflow of oxygen in the soil (Jain *et al.*, 2000).

### **2.3.2.3. Redox potential (Eh)**

Methanogenesis occurs only under anaerobic conditions. A sufficiently low value (-150 mV) of redox potentials (Eh) is required for methane production and it is negatively related to methane emission. In soils with high contents of Fe and organic matter, the Eh value falls to -50 mV and may slowly decline further to -200 mV over a period of one month. Soils low in active Fe with high organic matter attain lower Eh values much faster, may be within week after submergence. Flooded rice soils may have Eh values as low as -250 to -300 mV, while Eh of -150 to -190 mV is needed for methane formation (Wang *et al.*, 1993).

Methane was emitted during intermittent irrigation because the soil Eh at a depth of 5 cm remained lower than -200 mV, which stimulates CH<sub>4</sub> emission. Generally, CH<sub>4</sub> emission occurs at lower than -150 mV and increases with a decrease in soil Eh (Masscheleyn *et al.*, 1993; Wang *et al.*, 1993). On the other hand, O<sub>2</sub> consumption is the first process of the soil reduction after flooding that occurs at higher than 300 mV. Therefore, water management based on a specific time span is not necessarily sufficient to decrease CH<sub>4</sub> emission.

Hou *et al.* (2000) indicated that an intermediate range of soil Eh between approximately -100 and 200 mV may be sufficient to minimize the emissions of CH<sub>4</sub> and N<sub>2</sub>O. Although controlled drainage is necessary for sound rice growth and is effective in decreasing CH<sub>4</sub> emission, it is necessary to reconsider the span and frequency of drainage for minimizing the total CO<sub>2</sub>-equivalent emission.

### **2.3.2.4. Mineral fertilizers**

Methane fluxes are strongly influenced by the type, method and the rate of fertilizer application. Sulphate-containing fertilizers reduced methane emission. Aerts and Ludwig, 1997; Aerts and Toet, 1997; Lindau *et al.*, 1991, reported that methane emissions would increase with fertilizer applications. In contrast, Cai *et al.*, 1997; Wang *et al.*, 1992, reported that CH<sub>4</sub> emissions decreased with fertilizer application. Plant nutrients (N, P, S etc.) either naturally occurring or added as fertilizer to paddy soil may affect the emission of CH<sub>4</sub> by either influencing the growth of the rice plant or methanogenic microbial communities (Lindau *et al.*, 1991; Wang *et al.*, 1992; Kimura *et al.*, 1992; Delwiche and

Cicerone, 1993). Sass *et al.*, 1990 has been observed the correlation of CH<sub>4</sub> emission with plant biomass (both above and underground).

Phosphogypsum, a sulphate-containing by-product of industrial production of phosphoric acid, reduced methane emission by 56 - 73 per cent when applied in combination with urea. Adhya *et al.* (1994) reported inhibition of methane emission from flooded rice field with application of P fertilizer as single super phosphate. Applying phosphogypsum (calcium sulfate dihydride) in combination with urea has been determined to reduce methane emissions by more than 70 per cent (Metra-Corton *et al.*, 2000).

There are some reports pointed that microbial methanogenesis process in anoxic soil is stimulated by organic amendments (Yagi and Minami, 1990; Lauren and Duxbury, 1993; Denier van der Gon and Neue, 1995) and inhibited by nitrate (Bollag and Czlonkowski, 1973; Kitada *et al.*, 1993) and sulphate (Cappenberg, 1975; Westermann and Ahring, 1987; Achtnich *et al.*, 1995).

#### **2.3.2.5. Soil pH**

Most of the methanogens are neutrophilic hence methane production is most efficient in the pH range 6.4 - 7.8. Methanogenesis is a highly pH sensitive process and a small change in the pH value sharply lowers the methane production. Below pH 5.8 and above 8.8, methane production in the soil suspension is almost completely inhibited.

#### **2.3.2.6. Organic amendments**

Methane emissions are generally enhanced by organic inputs into the soil such as straw or manure amendment. The increment in CH<sub>4</sub> emissions following organic inputs depends on quantity, quality and timing of the application (Yagi and Minami, 1990; Sass *et al.*, 1991; Denier van de Gon., 2000). Rice straw and manure are typically applied before transplanting resulting in an emission peak during the first half of the growing season. High temperatures in the weeks following the incorporation of these materials result in a pronounced emission peak whereas low temperatures during this period diminish this peak (Wassmann *et al.*, 2000).

Organic residue amendments have been ordinarily accepted to improve soil fertility for rice production in China. Incorporation of crop residues provides a source of

readily available C and N, which is believed to induce a higher CH<sub>4</sub> release from rice fields (Denier van der Gon and Neue, 1995; Wang, 1999).

Decomposition of incorporated organic material is the predominant source of methanogenic substrates in early stage of rice growing (Watanabe and Roger, 1985) and CH<sub>4</sub> emission via ebullition during this period contributes significantly to the total emissions of this gas (Shangguan and Wang, 1993). In general, the highest CH<sub>4</sub> emissions are observed in fields receiving organic amendments. Decomposition of organic materials offers the predominant source of methanogenic substrates, particularly in the early stage of rice development.

Application of organic manure and crop residues enhances the methanogenesis process. The amount of methane formed in paddy soils is positively correlated with soil organic carbon and water soluble organic carbon and other factors, such as bacterial population and oxidizing capacity of the soil are not limiting. Incorporation of rice straw in the soil increases methane production by 120 - 800 per cent over that of unamended soil. Methane emission is highest with the incorporation of straw followed by straw compost, zero tillage with straw mulching and least with straw ash application (Setyanto *et al.*, 2000).

Application of biogas spent slurry as a manure results in lowering methane emission compared to application of farm yard manure (FYM) from rice fields (Dabnath *et al.*, 1996). Use of biogas slurry therefore could be a mitigation option for minimizing methane flux from flooded rice fields.

It has been reported that addition of fresh organic sources to the rice soil increases the availability of methanogenic substrates and thereby enhances CH<sub>4</sub> production and emission (Neue, 1993). Application of organic sources such as rice straw, *Azolla*, blue-green algae (BGA), green manure (leguminous and nonleguminous) and animal excreta to rice soils is one of the common cultural practices. The organic sources used were rice straw, cellulose, *Azolla* (a water fern harboring a nitrogen-fixing blue-green alga, *Anabaena*) compost, blue-green algae compost, farmyard manure (FYM) and green manure (GM). Composted organic sources are known to support low production of CH<sub>4</sub> when compared with the fresh organic sources (Debnath *et al.*, 1996).

### **2.3.2.7. Cultivars and crop growth stages**

The effects of cultivar on methane emission rates have been reported by a few researchers (Parashar *et al.*, 1990; Lindau *et al.*, 1995; Watanable *et al.*, 1995 and Wang *et al.*, 1997). Neue and Roger (1993) reported large differences in root oxidizing power among rice cultivars. Because of the cultivar controlled differences in methane source strength, in methane transport capacity and in root oxidation power, opportunities exist for screening and breeding rice cultivars that give a low methane emission rate.

Cultivar influences methane source strength by providing the soil with root exudates and leaf littering, which are positively correlated to aboveground dry matter and root weight. Differences in methane emission rates were not proportional to differences in methane source indicating difference in transport capacity among rice cultivars (Wang *et al.*, 1997). He also showed a difference in methane emission rates among rice cultivars, attributed mainly to cultivar-controlled differences in methane source strengths. He also reported that the ideal rice cultivars for reducing methane emission should have high harvest index, less ineffective tillers and high root oxidizing power

There are some significant variations in the quantities of methane emitted from soils growing different cultivars. The high yielding varieties like IR-64 show moderately high emission (Setyanto *et al.*, 2000). Such differences in CH<sub>4</sub> emission from the rice cultivars could be due to the differences in amounts of root exudates produced per plant, the CH<sub>4</sub> oxidizing capacity of the roots and the population level of methanogenic bacteria in roots.

The highest CH<sub>4</sub> peaks were observed at flowering and heading stages, which could be related to the development of intense reducing conditions in the rice rhizosphere (Neue and Roger, 1993; Adhya *et al.*, 1994; Chidthaisong *et al.*, 1999), increased availability of labile organic matter and root exudates (Schutz *et al.*, 1989; Bouwman, 1991; Wassmann *et al.*, 1993; Denier van Der Gon and Neue, 1995; Inubushi *et al.*, 2003) and enhanced conductivity of CH<sub>4</sub> via rice plant as supported by Mariko *et al.*(1991). Ali *et al.* (2008, 2009) reported that methane emission rate was comparably lower at the initial rice growing stage, which increased significantly with plant growth and with the development of soil reductive condition during rice growing seasons and also reported that as plant reached maturity CH<sub>4</sub> emission rates gradually decreased and finally

dropped to minimum levels, even though that the field was still flooded and could still possibly produce CH<sub>4</sub>. This indicates that rice plant growth and physiological parameters play vital roles in controlling CH<sub>4</sub> emission in the flooded paddy soil.

#### **2.3.2.8. Inhibitors**

Sahrawat, K. L. (1989) reported that nitrification is generally used to mean biological oxidation of ammonium to nitrate via nitrite effected respectively by *Nitrosomonas* and *Nitrobacter* species of nitrifying bacteria, although nitrification inhibitors are defined as compounds or materials that specifically inhibit or retard the oxidation of ammonium to nitrite without affecting the subsequent oxidation of nitrite to nitrate.

Banerjee and Mosier (1989) demonstrated that encapsulated calcium carbide is a slow release source of acetylene that inhibits nitrification and reduces nitrous oxide fluxes in flooded soils. Acetylene also has been reported to inhibit methane production. Keerthisinghe *et al.* (1993) found that nitrapyrin and acetylene nitrification inhibitors significantly reduced methane emission in flooded rice.

#### **2.3.3. Oxidation of methane in flooded soils**

Methane oxidizing bacteria (methanotrophs) are abundant in the oxidized flooded water soil interface and in the rice rhizosphere. They sequentially oxidize methane to carbon dioxide via methanol, formaldehyde and formate. Oxygen is essential for the growth of methanotrophs, but the required partial pressure may be low (Cicerone and Oremland, 1988). Methane oxidation greatly limits diffusion of methane to the atmosphere. Ammonium ion inhibited methane oxidation in studies with pure cultures of methanotrophs (Hyman and Wood 1983; Whittenbury *et al.* 1970). Up to 60 per cent of the methane produced during a rice growing season may be oxidized before it reaches the atmosphere (Holzapfel-Pschorn *et al.* 1986; Sass *et al.* 1991).

Rice plants supply atmospheric oxygen to the roots for respiration via a special vascular system, the aerenchyma. The aerenchyma has its own openings at the leaf sheath (Nouchi *et al.*, 1991) and the gas supply to the roots is independent of transpiration and stomatal gas exchange. Oxygen diffusion from rice roots constitutes an important part of the roots oxidizing power, aside from enzymatic hydrogen peroxide production. Because

of the abundance of methane-oxidizing bacteria present in the rhizosphere, the rhizosphere's potential for methane oxidation is high.

De Bont *et al.* (1978) counted ten times more methane oxidizing bacteria in the rhizosphere than in the bulk anaerobic soil and one third more than in the oxidized soil water interface. They found significant increases in methane emission by the rice cultivar IR36 when methane oxidation was suppressed with acetylene at the soil water interface. However, acetylene had only a small effect on emission rates when applied to the rhizosphere. They concluded that the use of oxygen by reduced substances and microbes other than methanotrophs at the region of the root soil interface exceeds the supply of oxygen by the root.

#### **2.3.4. Temporal and spatial variation in methane emission**

The methane emission generally shows strong diurnal and seasonal variation. The seasonal variation in methane emission depend on factors like growing stage of rice, temperature, day-length, solar radiation, humidity, water regimes, fertilization and weeding etc. During the early vegetative period, maximum methane emission occurs at both noon and night while during the late vegetative period, it occurs only at night. It is due to transportation of less oxygen to the root system of rice plants at night resulting in less methane oxidation and higher emission. The temporal and spatial variations of methane production are related to rice root biomass, which might depend on cultivars and soil. The first maximum may occur within four weeks after flooding and is governed by CH<sub>4</sub> production from soil organic matter and organic amendments (Neue *et al.*, 1994).

#### **2.3.5. Pathway of methane emission from rice fields**

Submerged paddy soils without rice plants released methane to the atmosphere through diffusion of dissolved methane, ebullition of its gas bubbles. Ebullition of methane gas generally occurs when the partial pressure of entrapped methane within the soil exceeds the hydrostatic pressure (Mattson and Likens, 1990), while the diffusion of gases in water is approximately 10,000 times slower than in air (Armstrong, 1979; Wang *et al.*, 1995). Therefore, large portions of methane may remain trapped in the flooded soil and the diffusive exchange of gases nearly ceases. However, some methane emission through diffusion process continues through the soil-water and air-water interface.

### **2.3.5.1. Ebullition**

Ebullition is the dominant transport pathway after the initial flooding, during land preparation and when rice plants are still small (Wassmann *et al.*, 1996). Neue *et al.*, 1994 reported that the ebullition is a major methane release pathway during land preparation and early rice growth stage.

### **2.3.5.2. Diffusion across water surface**

Diffusion of gases in water is  $10^4$  times slower than in air and therefore, exchange of gases almost stops when soils are waterlogged. The actual diffusion of methane from rice fields is a function of its supply to the floodwater, its concentration in the floodwater and prevailing wind speed. Diffusion through the floodwater is usually less than 1 per cent of the total flux. It is suggested that the rate-limiting step in plant-mediated  $\text{CH}_4$  transport is its diffusion across the root/shoot junction (Armstrong, 1979; Wang *et al.*, 1995).

### **2.3.5.3. Transport through rice plant**

Methane produced in sediment flows into the roots, to the shoots and out of the leaves of aquatic plants (Dacey and Klug, 1979). Nouchi *et al.* (1990) reported that methane was mostly released from the culms (aggregation of leaf sheaths) of rice plants but not from the leaf blades. They also found that micro pores of the aerenchyma, different from stomata, at the abaxial epidermis of the leaf sheaths as gas exchange sites.

Lee *et al.* (1981); Nouchi *et al.* (1990) and Seiler *et al.* (1984) reported that stomatal closing during darkness did not significantly affect methane emission rates from rice plants and dismissed stomata as release sites of methane. Nouchi *et al.* (1990) found methane emissions only up to 13 cm from the base of culm. Methane diffused through rice plants according to the concentration gradient between the soil and the atmosphere (Denier van der Gon and van Breemen, 1993).

The  $\text{CH}_4$  formed in flooded soils and sediments can migrate to the surface and be emitted into the atmosphere by one of three different pathways. Kruger *et al.* (2001) reported that first diffusion can take place in solution towards the surface, in the course of which a substantial proportion of the  $\text{CH}_4$  oxidation (10-40 per cent) in rice fields happens. Second, sufficient gas may be produced for bubbles to form in the water layer and force their way to the surface by the process of ebullition. The speed of this process

prevents any significant oxidation (Conrad, 1989). The third route is via the continuous air spaces aerenchyma of vascular rice plants which are adapted to life in flooded environments (Thomas *et al.*, 1996 and Lloyd *et al.*, 1998). These structures have evolved to transport oxygen needed for root respiration and cell division, but serve equally well as channels for the transport of methane from the root environment to the atmosphere.

## **2.4. FEASIBLE METHANE MITIGATION OPTIONS FOR RICE CULTIVATION**

Mitigation options must achieve both reduced methane emission and increased sustainable production of rice and some of the possible mitigation strategies to minimize the global warming potential of rice fields are discussed.

- Water management.
- Direct seeding.
- Use of chemical fertilizers.
- Use of different rice cultivars.
- Improved tillage and crop residue management practices.
- Use of Phytosynthetic Blue green algae in rice cultivation.
- Dual cropping of *Azolla* in rice.

### **2.4.1. Water management**

Water management practices (intermittent drainage) aimed at reducing methane emissions are feasible. Increasing water percolation would add oxygen-rich water to the reduced soil layer and decrease methane production. In drought-prone rain-fed areas, water is too valuable to be drained. Higher water percolation requires more water and may cause detrimental leaching of nutrients. Significant quantities of methane may also be leached and subsequently released to the atmosphere elsewhere (Kimura, 1992). Temporarily aerating the soil by stopping irrigation enhances methane oxidation and decreases methane formation as well as total emission. But intermittent aeration may increase gaseous losses of nitrogen as nitrous oxide (nitrification-dinitrification). Drying of rice terraces at hillsides can cause severe cracking and collapse of the terrace construction.

#### ***2.4.1.1. Temporary drainage of rice fields***

Draining stimulates root development and accelerates decomposition of organic materials in the soil, resulting in more mineralized nitrogen available for plant uptake. Midseason drainage aerates the soil, interfering with the anaerobic conditions and thereby interrupting methane production (NASA, 2002). In the Beijing region of China, the local practice of drying field's midseason has been found to reduce methane emission rates by 23 per cent, as compared with continuous flooding (Wang *et al.*, 2000).

Research conducted by Lu *et al.* (2000) reported that the methane emission reduction associated with midseason drainage could be as high as 44 per cent. Field experiments conducted in the Philippines results showed that the field drying at the mid tillering stage reduced methane emissions by 15 to 80 per cent, as compared with continuous flooding, without having a significant effect on rice yield (Wassmann *et al.*, 2000). Studies conducted by Parashar *et al.* (2002) in India reported that controlled irrigation can effectively reduce methane without affecting yield.

#### **2.4.2. Direct seeding (Versus Transplanting)**

Direct seeding of pre-germinated rice instead of transplanting rice seedlings has been shown to reduce methane emissions due to shorter flooding periods and decreased soil disturbances. The research conducted by IWMI, 2007 revealed that methane emission reductions were an unintended benefit of direct seeding. Ko and Kang, (2000) reported that transplanting at 30 days old seedlings, direct seeding on wet soil and direct seeding on dry soil reduced methane emissions by 5, 13 and 37 per cent respectively, when compared with transplanting eight days old seedlings. The results of Metra-Corton *et al.* (2000) showed 16 to 54 per cent reduction in methane emissions in direct seeding (versus transplanting).

Despite the emission reductions and labor savings associated with direct seeding, transplanting rice seedlings has a number of benefits that are important to consider for the farmer. These include higher yields and an assurance that transplanting is done at the optimal time for the plants (Bouman, 1991).

#### **2.4.3. Use of chemical fertilizers**

The direct impact of chemical fertilizer applications on methane emission is not clear. Because most methane is emitted through the rice plant, improved rice growth

(more tiller and roots) in response to fertilizer application increases emission. But source and mode of application may also have direct effects (Schutz *et al.* 1989). Sulfate-containing fertilizer reduces methane emission. Sulfate-reducing bacteria compete with methanogens for the limited hydrogen, but the amount of sulfate normally added as fertilizer seems to be insufficient to have significant effects.

Most of the increases in Asian rice production resulting from the “Green Revolution” have been attributed to increased nitrogen use. Nitrogen use may also have the benefit of resulting in lower methane emissions. Incorporating urea into the soil has been shown to reduce methane emissions; however, surface-applied urea resulted in nearly 20 per cent greater emissions compared to unfertilized fields (US-EPA, 1991).

Nitrogen fertilizers are commonly used in rice production to increase grain yields. Urea and ammonium sulfate account for 80–90 per cent of the total nitrogen fertilizer demanded in rice cultivation (FAO, 2010). Lindau *et al.* (1991) found that the methane emission increased with urea application. In contrast, Wang *et al.* (1993) reported, no change in emission with urea application and a decrease in methane production with ammonium nitrate application as a nitrogen source. Cai *et al.* (1997) has been reported that methane emission on an average decreased by 42 and 60 per cent in the ammonium sulphate treatments and 7 and 14 per cent in the urea treatments at rates of 100 and 300 kg N ha<sup>-1</sup> respectively compared to control.

Urea accounts for approximated 80 per cent of the nitrogen applied to rice in Asia and ammonium sulfate accounts for approximately 6 per cent (De Datta, 1981). Most farmers apply nitrogen fertilizer in two or three splits. The first split is applied during final land preparation or shortly after planting and the remainder is broadcast at later growth stages, especially at panicle initiation. To minimize volatilization losses of nitrogen fertilizer, it is recommended to place the fertilizer granules 10 cm deep in the soil during the final land preparation. In general, potassium and phosphorus fertilizer are basal-applied during final land preparation. Potassium chloride is the principal fertilizer source of potassium and superphosphate is the primary source of phosphorus fertilizer. On acid rice soils, phosphate rock may be applied.

Impacts of various other culture practices (land preparation, seeding and transplanting, pest control and harvest) on methane emission have not yet been studied in

detail. A few observations at the International Rice Research Institute (IRRI) reveal that soil disturbances caused by current culture practices release large amounts of soil-entrapped methane. The increased adoption of direct seeding (wet and dry seeding) instead of transplanting is likely to reduce methane emission. In direct-seeded rice, flooding periods are shorter and cultural disturbance of reduced soils is minimized.

Several million hectares of the Indo Gangetic plains are alkaline soils (Abrol *et al.*, 1985). Gypsum ( $\text{CaSO}_4$ ) inputs into the soil reduced  $\text{CH}_4$  emissions by 29 to 46 per cent in field study in Louisiana (Lindau *et al.*, 1993) and by 55 to 70 per cent in the Philippines (Denier van der Gon and Neue, 1994). Gypsum application can be beneficial to neutralize the pH of alkaline soils. In the rice-wheat belt, gypsum application may offer some potential to combine the purposes of improving soil fertility and mitigating emissions.

#### **2.4.4. Use of different rice cultivars**

The variety (cultivar) of rice has been shown to have an effect on methane emissions. Wang *et al.* (2000) reported that some varieties transmit more methane than others. They also reported that the use of cultivar Zhongzhou (modern japonica) reduced methane emissions by approximately 50 percent when compared with Jingyou (japonica hybrid) and Zhonghua (tall japonica) in the Beijing region of China. Wang and Adachi 2000 revealed that that rice cultivars with small root systems, high root oxidative activity, high harvest indices (the ratio of harvested yield to total yield) and productive tillers (shoots) are likely to produce less methane than other cultivars in China.

Studies conducted by Shin and Yun, (2000) reported some rice cultivars can emit more than twice as much as others in Korea. The Annada rice variety (commonly used in Andhra Pradesh, a major rice growing region in India) has high yield and comparatively low methane emissions, which might mitigate emissions from paddy fields in that region (Parashar *et al.*, 2002).

Additional research is needed to identify important plant features, screening the existing rice varieties and develop new varieties that meet all the requirements for pest resistance and high productivity as well as methane reduction (US-EPA, 1991). Although low methane emitting rice cultivars have been identified, methane emission reductions

due to cultivar selection have been shown to be less significant than those identified due to modifying water management regimes or adding organic amendments (Lu *et al.*, 2000). In addition, the rice yield of the low methane emitting cultivars would need to be evaluated; if the low CH<sub>4</sub> emitting rice cultivar produced less rice, then more rice would need to be cultivated to meet the demand for rice and therefore overall methane emissions may increase.

The wide variation of these traits and related emission rates among cultivars opens the possibility for breeding rice cultivars with low methane emission potential. The inheritance of underlying traits and relationships to yield potential has yet to be elucidated.

#### **2.4.5. Improved tillage and crop residue management practices**

Methane emissions are very intense during the tilling stage of rice field preparation, which can account for more than 80 per cent of total annual emissions. Wetland tillage was compared with dry land or zero tillage, results in an earlier onset of methanogenesis and therefore it contributes to greater methane production during the growing season (US-EPA, 1991). Zero tillage results in the lowest methane emissions and is a practice that involves leaving crop residue in place to compost and mulch the soil. Increased land and water productivity, as well as improved net income, has attracted some farmers to adopt zero tillage techniques when preparing rice fields for wheat planting. Lack of familiarity with the zero tillage technique is a major constraint for small farmers to adopt new practices (IWMI, 2007). There is also potential for certain harmful pests such as the stem borer to survive on the unincorporated residue or stubble. Some crop residue management techniques can be implemented in the near term; however, deploying new machinery and training is a longer term endeavor.

#### **2.4.6. Role of Blue Green Algae in minimizing methane emission from rice fields**

Blue green algae (Cyanobacteria / BGA) which colonize in the rice fields have been reported to be responsible for the long term fertility of paddy fields. On an average BGA contributes 25-30 kg nitrogen per hectare per season and leads to 10-15 per cent increase in rice productivity. BGA helps in improving soil health, increasing soil water holding capacity and availability of several nutrients. The growth and proliferation of

BGA also helps in reducing methane emission, buffering the pH of soil and water besides preventing weed cover (Rao and Burns, 1991).

A yield improvement of rice of between 5 per cent and 25 per cent was found when fields were inoculated with BGA even in the presence of 100-150 kg N ha<sup>-1</sup> as fertilizer (Sprent and Sprent, 1990; Yanni, 1992).

Cyanobacteria are well adapted to a wide range of environmental conditions and have been widely employed as inoculants for enhancing soil fertility and improving soil structure, besides enhancing crop yields especially in rice (Venkataraman, 1972; Kaushik, 2004; Nayak *et al.*, 2004; Dhar *et al.*, 2007). The favorable conditions provided by the rice fields for nitrogen fixation by these organisms leads to enhanced plant-available N in soil and yield improvement of rice (Roger *et al.*, 1993; Mandal *et al.*, 1998). Cyanobacteria also add organic matter, synthesize and liberate amino acids, vitamins and auxins, reduce oxidizable matter content of the soil, provide oxygen to the submerged rhizosphere, ameliorate salinity, buffer the pH, solubilize phosphates and increase the efficiency of fertilizer use in crop plants (Mandal *et al.*, 1998; Kaushik, 2004). Enhancement of rice seed germination, root and shoot growth, weight of rice grains and their protein content and the fertilizing action of N<sub>2</sub>-fixing cyanobacteria have been generally attributed to the release of synthesized nitrogenous compounds either by decomposition of the cells or excretion (Venkataraman and Neelakantan, 1967; Nayak *et al.*, 2004). Singh, (1950) and Oikarinen, (1996) reported their influence on the physical, chemical and biological properties of the soil and soil water interface excluding supplying nitrogen in rice fields.

Prasanna *et al.*, (2002) reported that the photo synthetically generated O<sub>2</sub> is of crucial importance in the maintenance of redox potential and oxygenic conditions of soil despite frequent flooding. Laboratory simulation experiments were undertaken using soil cores from rice fields inoculated with cyanobacterial/*Azolla* biofertilizers. The results revealed a rapid decrease in headspace concentrations of methane in the soil cores from these treatments as compared to the treatments involving the use of chemical fertilizers. This can be attributed to the significant role of oxygen evolved by cyanobacteria during photosynthesis as oxygen donors to methylotrophs or direct enhancement of methane oxidation.

The less investigated beneficial effects of cyanobacteria include curbing of ammonia volatilization, suppressing weeds, reducing methane emission, transformation of P, Fe, Mn, Zn Cu, pesticide degradation and reclamation of wastelands/degraded soil (Mandal *et al.*, 1993; Prasanna *et al.*, 2002).

#### **2.4.7. Dual cropping of *Azolla* in minimizing methane emission from flooded rice fields**

*Azolla* is a genus of aquatic ferns found floating in swamps, ditches, lakes and rivers. Due to its aquatic nature, rapid growth, ability to fix 30 - 60 kg N ha<sup>-1</sup> in 30 days (due to symbiosis with *Anabaena* and blue-green algae) and high N content *Azolla* has been used as a green manure or a dual crop in rice cultivation for many years (Wagner, 1997). Application of organic substrates, including green manure, often increased the CH<sub>4</sub> flux from flooded rice paddy (Yagi and Minami, 1990; Denier van der Gon and Neue, 1995; Wassmann *et al.*, 1996).

Watanabe *et al.* (1989) reported that a dual crop of *Azolla* resulted in lowering of CH<sub>4</sub> efflux while increasing the rice yield. Bharati and Mohanty *et al.* (2000) indicated that dual cropping of *Azolla* reduced methane flux and yet increased grain yield similar to that of urea application. The decrease in methane efflux in plots with dual crop of *Azolla* could be related to the release of oxygen in the standing water by the growing *Azolla* leading to less reduced conditions in the soil (Bharati and Mohanty *et al.*, 2000). The methanotrophs oxidize methane to carbon dioxide deriving energy from this reaction. *Azolla* is an interesting option for harvesting greenhouse gases such as CO<sub>2</sub> (Prasanna and Kumar, 2002).

Wang *et al.* (1993) reported that treatment of soil with dual crop of *Azolla* registered a higher redox potential leading to low CH<sub>4</sub> flux. High Dissolved Oxygen (DO) in the floodwater might retard CH<sub>4</sub> emission from rice field by promoting CH<sub>4</sub> oxidation at the soil water interface (Hanson and Hanson, 1996). Leaching of oxygenated water by percolation in flooded rice fields would inhibit methanogenesis by keeping the soil in a more oxidized state as well as stimulate CH<sub>4</sub> oxidation in the reduced soil layer (Kimura *et al.*, 1992). The mean DO<sub>2</sub> concentration was higher in field plots with a dual crop of *Azolla* indicating the role of *Azolla* in enriching the standing water with oxygen. In dual cropping of *Azolla*, the growing fern forms a mat above the standing water, but unlike many other aquatic plants is not particularly known to release oxygen through its floating roots (Ashton and Walmsley, 1976). CH<sub>4</sub> emission from a flooded field was low

during the active growth of algae and this was attributed to the release of oxygen from the algal mat during photosynthesis (Wang *et al.*, 1995). *Azolla* can promote aerobic transformations such as methane oxidation through enhanced aeration of the flood water in rice fields (Prasanna and Kumar, 2002).

## CHAPTER III

### MATERIALS AND METHODS

A field experiment was conducted at Anbil Dharmalingam Agricultural College and Research Institute, Trichy during *Rabi* (2010 - 2011) season to study the methane emission rates from rice field under different organic amendments, besides understanding the correlation between methane flux and air/ water temperature. The trial was also aimed to assess the potentials of the photosynthetic diazotrophs such as Blue green algae and *Azolla* in minimizing methane flux of rice ecosystem as a possible climate change mitigation strategy. The details of the materials used and methods adopted in the field experiment are furnished in this chapter.

#### 3.1. Field experiment

##### 3.1.1. Location

The field experiment was carried out in the 'A1C' block of farm of Anbil Dharmalingam Agricultural College and Research Institute, Trichy. The farm is situated at 10° 45'N latitude, 78° 36'E longitude and at an altitude of 85 m above mean sea level.

##### 3.1.2. Climate and weather

The experiment location having the climate of Semi-Arid Tropics experiences a mean annual rainfall of 843 mm distributed over 48 rainy days. The mean maximum temperature and minimum temperature are 34.8°C and 24.7°C respectively. The relative humidity ranged from 87 to 96 per cent in the forenoon and 66 to 87 per cent in the afternoon.

The experimental field falls under the tropical climate and the meteorological parameters such as temperature, relative humidity, rainfall, evaporation, sunshine hours, wind velocity and solar radiation recorded during the period of experimentation are furnished in Table.5 and Fig.2.

##### 3.1.3. Season

The field experiment was conducted during *Rabi*, 2010-2011.

##### 3.1.4. Soil characteristics

The soil of the experimental field was sandy clay loam, taxonomically classified as isohyperthermic Vertic Ustropet, having 191 kg ha<sup>-1</sup> of available nitrogen, 27.5 kg ha<sup>-1</sup> of available phosphorus and 240 kg ha<sup>-1</sup> of available potassium. The detailed physico-chemical properties of the soil of experimental field are presented in Table 4. The methods used for analysis of soil properties are presented in Table 1.

**Table 1. Methods employed for soil analysis**

<i>Particulars</i>	<b>Method</b>	<b>Reference</b>
<b>I. Soil analysis</b>		
A. Mechanical analysis	Robinson's International pipette method	Piper (1966)
B. Chemical analysis		
1. Available nitrogen	Alkaline permanganate method	Subbiah and Asija (1956)
2. Available phosphorus	Using 0.5 M NaHCO <sub>3</sub> of pH 8.5 using colorimeter	Olsen <i>et al.</i> (1954)
3. Available potassium	Flame photometric method using neutral normal ammonium acetate extract	Stanford and English (1949)
4. Organic carbon	Chromic acid wet digestion method	Walkley and Black (1934)
5. pH (Soil: water = 1: 2)	Using glass electrode in the ELICO pH meter	Jackson (1973)
6. Electrical conductivity (Soil: water = 1: 2)	Using ELICO conductivity bridge	Jackson (1973)

### **3.1.5. Crop and variety**

The rice variety TNAU (R) TRY1 was chosen for the study and it has duration of 135 days. The Green manure *Sesbania aculeata* was raised in a separate field and incorporated in the field as green leaf manure before planting as per the treatment. The characteristics features of TNAU (R) TRY1 used in the study are given in Table 2.

**Table.2. Characteristics of TNAU (R) TRY 1 rice variety**

<b>Characters</b>	<b>Details</b>
Parentage	IR 578-172-2-2/BR 1-2B-1-19
Duration	135-140 days
Average yield	4011 kg ha <sup>-1</sup>
1000 grain weight	24.0 g
Grain type	Medium
Habit	Erect
Leaf sheath	Green
Panicle	Long, moderately compact
Grain type	Medium
Grain length	6.2 mm
Breadth (at middle)	2.4 mm
Thickness	1.8 mm

### **3.2. Methods**

#### **3.2.1. Design and layout**

The experiment was laid out in Randomized Block Design (RBD) with three replications. The layout of the experiment is presented in Fig.1.

#### **3.2.2. Treatments**

The details of the treatments imposed in the experiment are given below

##### **Treatment details**

- T<sub>1</sub> - Control
- T<sub>2</sub> - Blue Green Algae (BGA)
- T<sub>3</sub> - *Azolla*
- T<sub>4</sub> - Farm Yard Manure (FYM)

- T<sub>5</sub> - Green Leaf Manure (GLM)
- T<sub>6</sub> - BGA + *Azolla*
- T<sub>7</sub> - FYM + GLM
- T<sub>8</sub> - BGA + *Azolla* + FYM + GLM

### 3.2.3. Plot size

- Gross plot - 5 x 4 m
- Net plot - 4.6 x 3.6 m

**Table 3. Experimental details**

<b>Particulars</b>	<b>Crop and other details</b>
Season	Late Samba (Rabi)
Period	September, 2010–February, 2011
Spacing	20×10 cm (50 hills m <sup>-2</sup> )
Design	Randomized Block Design
Treatments	8
Replications	3
Date of sowing	15.09.2010
Nursery duration	19 days
Date of transplanting	02.10.2010
Main field duration	129 days
Date of Harvesting	07-02-2011

### **3.3. Cultivation details**

#### **3.3.1. Field preparation**

The experimental field was thoroughly puddled with tractor mounted cage wheel and soft permeable layer of soil was achieved. Then the field was levelled properly with wooden level board. The plots were laid out according to the experimental layout with irrigation cum drainage channels all around the field.

#### **3.3.2. Seeds and sowing**

A seed rate of 40 kg ha<sup>-1</sup> of rice variety (TRY1) was used for the experiment. The seeds were treated with Carbendazim @ 2 g kg<sup>-1</sup> of seeds for protection against seed borne diseases. After 24 hours of fungicidal treatment, the seeds were treated with *Azospirillum* @ 600 g ha<sup>-1</sup> of seeds. The treated seeds were soaked in water for 24 hours to induce sprouting. The sprouted seeds were sown uniformly in the well prepared nursery maintaining thin film of water. The sowing was done on 15.09.2010.

#### **3.3.3. Transplanting**

Seedlings of 19 days old were pulled out from the nursery and transplanted on the same day with a spacing of 20 x 10 cm (50 hills m<sup>-2</sup>) on 02.10.2010.

#### **3.3.4. Gap filling**

The plots were sufficiently irrigated before thinning and gap filling. Gap filling was done at 10 days after transplanting (DAT) to maintain uniform population of 50 hills m<sup>-2</sup>.

#### **3.3.5. Irrigation**

Irrigation was given by maintaining 2 cm of water at transplanting. As the seedlings growth progressed, the water level was slowly increased up to 5 cm level. The irrigation was withheld 10 days prior to harvest.

#### **3.3.6. Fertilizer application**

A recommended dose of 187.5:50:50 kg NPK ha<sup>-1</sup> was applied to the crop uniformly to all the treatments. The entire dose of phosphorous as single super phosphate (16 % P<sub>2</sub>O<sub>5</sub>) was applied as basal. Potassium in the form of muriate of potash (60 % K<sub>2</sub>O) was applied in two equal splits, one at basal and other at panicle initiation stage. Nitrogen

in the form of urea (46 % N) was applied in four equal splits at basal, active tillering, panicle initiation and flowering stages.

### **3.3.7. Farm Yard Manure application**

Well decomposed FYM was applied @ 12.5 tonnes ha<sup>-1</sup> to the respective treatments.

### **3.3.8. Green leaf manure application**

Green leaf manure (*Azadiracta indica*, *Pongamia glabra* and *Calotropis gigantea*) was collected from the nearby fields and incorporated @ 6.25 tonnes ha<sup>-1</sup> in the specific treatmental plots.

### **3.3.9. BGA flakes application**

Composite BGA inoculum, collected from the mother inoculum field at wetlands of Tamil Nadu Agricultural University, Coimbatore containing *Anabaena sp*, *Nostoc sp* and *Westiellopsis sp* was applied in the form of flakes. The BGA was applied @ 10 kg ha<sup>-1</sup> during ten days after transplanting.

### **3.3.10. Azolla application as dual crop**

*Azolla microphylla* inoculum @ 1 tonnes per hectare was applied 10 days after transplanting as per treatment. Water level was maintained at 5 cm throughout the growth and development of the fern till 10 days before harvesting.

### **3.3.11. Weed management**

Pre-emergence application of Butachlor @ 1.25 kg a.i. ha<sup>-1</sup> was applied on three DAT followed by two hand weeding at 30 and 60 DAT to ensure weed free condition.

### **3.3.12. Plant protection**

Leaf folder attack was noticed at tillering and panicle initiation stages and timely plant protection measure was taken up with Monocrotophos 36 WSC @ 1 liter ha<sup>-1</sup>.

### **3.3.13. Harvest**

The crop was harvested on 07.02.2011, upon physiological maturity. The border rows on either side were removed from the field. The net plot was then harvested plot wise, hand thrashed, sun dried and weighed at 14 per cent moisture level. The grain yield

was converted to express in  $\text{kg ha}^{-1}$ . The straw was also collected plot wise sun dried, weighed and reported as  $\text{kg ha}^{-1}$ .

### **3.4. Yield attributes**

#### **3.4.1. Productive tillers**

Ear bearing tillers alone were counted in tagged plants and expressed as productive tillers  $\text{m}^{-2}$ .

#### **3.4.2. Number of filled and ill filled grains**

Ten panicles were collected at random and threshed individually and the number of filled and ill filled grains were counted and recorded as filled and ill filled grains and expressed in numbers panicle<sup>-1</sup>.

#### **3.4.3. Test weight**

Thousand filled grains were sampled from the produce in each plot and weighed at 14 per cent moisture content and expressed in grams (g).

#### **3.4.4. Panicle wt**

The weight of the randomly selected 10 panicles was recorded individually and the mean value was expressed in grams (g).

### **3.5. Yield**

#### **3.5.1. Grain yield**

The harvested plants from the net plot area were threshed manually and each plot grain was thoroughly sun dried, cleaned and weighed. Grain yield was corrected to 14 per cent moisture content as suggested by Yoshida and Paraoq, (1976) and expressed in  $\text{kg ha}^{-1}$ .

#### **3.5.2. Straw yield**

The yield of straw from each net plot area was weighed after sun drying and expressed in  $\text{kg ha}^{-1}$ .

### 3.5.3. Harvest index (HI)

HI was worked out by using the formula

$$\text{HI} = \frac{\text{Economic yield}}{\text{Biological yield}} \times 100$$

### 3.6. CH<sub>4</sub> flux measurements

Plant-mediated CH<sub>4</sub> emission flux from the experimental plots was measured by closed chamber method of Adhya *et al.*, (1994) at regular intervals from transplanting to harvest. Samplings for CH<sub>4</sub> flux measurements were made at 09:00-10:00 hours and 15:00-16:00 hours and the average of morning and evening fluxes were used as the flux value for the day. For measuring CH<sub>4</sub> emission, eighteen rice hills were covered with a locally-fabricated transparent acrylic sheet chamber (59.3 cm length, 59.3 cm width and 87.8 cm height). A battery-operated fan was fixed for air circulation (avoid plant suffocation) to mix the air inside the chamber and draw the air samples into air-sampling bags (Tedlar®). The air samples from the sampling bags were analyzed for CH<sub>4</sub>. Each chamber was placed on the soil surface with 4-5 cm inserted into the soil, 10 minutes prior to each sampling for equilibration to reduce the disturbance to the sampling site.

#### 3.6.1. CH<sub>4</sub> estimation

The CH<sub>4</sub> was estimated in a Shimadzu GC-2014 gas chromatograph equipped with FID. The gas samples were introduced into the analyzer by filling the fixed loop (1.0 mL) on the sampling valve. Samples were injected into the column system by starting the analyzer which was automatically activates the valve and back flush the samples according to the time programmed. The retention time of CH<sub>4</sub> was between 4 to 4.17 min. The GC was calibrated before and after each set of measurements using 1 ppm, 2.3 ppm and 5 ppm of CH<sub>4</sub> (Chemtron® science laboratories Pvt. Ltd., Mumbai) as primary standard curve linear over the concentration ranges used. The minimum detectable limit for CH<sub>4</sub> was 1 ppm. CH<sub>4</sub> flux was determined by peak area and CH<sub>4</sub> flux was expressed as mg m<sup>-2</sup> day<sup>-1</sup> using the equation given by Lantin *et al.* (1995). The registered methane flux values are furnished in **Table 10**.

### 3.7. Soil analyses

Measurements for redox potential and dissolved oxygen concentration were done with each set of CH<sub>4</sub> flux measurement. The redox potential (Eh) of the field soil was measured by inserting a combined water proof ORP/ redox meter (Eutech Instruments, USA) to the root region and measuring the potential difference in mV (Satpathy, 1997). The Eh of soil was measured (rhizosphere to bulk soil interface) in the morning and afternoon at different points near the flux measurement setup and averaged for the day and furnished in **Table 8**.

Dissolved oxygen concentration at the soil–floodwater was measured using an Azide modification iodimetric method and expressed as mg l<sup>-1</sup>. Soil chemical components were analyzed from field soils sampled by inserting auger (2 cm diameter) to a depth of 5-7 cm in between two rice hills. The analyzed values were presented in **Table 9**.

### **3.7.1. Soil temperature and water temperature**

Soil temperature and water temperature were measured in each treatment during the entire crop period. Soil temperature reading was taken with mercury in glass thermometers (15 cm depth) which were placed in each treatment and water temperature was measured with ordinary thermometer. Soil and water temperatures were recorded at 10.00 and 15.00 hrs and averaged for the day. The mean data are presented in **Table 6** and **Table 7** respectively.

### **3.8. Statistical analysis**

The data collected were subjected to statistical analysis in Factorial Randomized Block Design following the method of Gomez and Gomez (1984). Whenever the treatment difference were found significant (F test) critical difference were worked out at five per cent probability level and the values were furnished. If there are no significant difference between treatments, it was denoted by the symbol NS.

## CHAPTER IV

### EXPERIMENTAL RESULTS

Field experiment was conducted at Anbil Dharmalingam Agricultural College and Research Institute, Trichirapalli during *Rabi* (2010 - 2011) to study the effect of temperature, redox and dissolved oxygen on methane flux of rice soil ecosystem and designing mitigation strategies to minimize global warming potential. *Azolla* (as dual crop), Blue Green Algae (as biofertilizers), Green Leaf Manure and Farm Yard manure were used in the experimental field to estimate their influence on methane flux, redox potential and dissolved oxygen concentration from the flooded rice field. As temperature influences methane emissions, air, soil and water temperatures were recorded from the field experiment at periodical intervals. The results obtained from this experiment are presented in this chapter. As minimizing methane emission from rice eco system is considered to be the prime climate change mitigation strategy in agriculture, the importance of photosynthetic Blue green algae and *Azolla* in methane emission reduction at source was studied and the findings are also detailed in this chapter.

#### **4.1. Initial Physiochemical properties of experimental soil**

The data on initial soil characteristics of the experimental site are presented in Table 4. The experimental soil was sandy clay loam in texture. The soil pH was 9.1 and non - saline (EC of 0.35 dS m<sup>-1</sup>). The organic carbon content was medium (0.49 %). With regard to nutrient status, the soil was low in N (191 kg ha<sup>-1</sup>) and P (27.5 kg ha<sup>-1</sup>) and medium in K (240 kg ha<sup>-1</sup>) levels. The CEC of the soil was 3.46 cmol (p<sup>+</sup>) kg<sup>-1</sup> with an ESP of 24.86. Considerable population of microorganisms (bacteria, fungi and actinomycetes) and enzyme (dehydrogenase, phosphatase and urease) activities were also observed in the experimental field soil.

#### **4.2. Weather prevailed during crop growth period**

The data on weather condition prevailed during crop growth period was recorded and furnished in **Table 5**.

During the crop growth period the lowest maximum temperature of 29.6 °C was noticed at 52<sup>th</sup> standard week and the lowest mean minimum temperature (19.4 °C) was registered at 4<sup>th</sup> standard week. The highest morning mean relative humidity (95.4 %) was recorded at 4<sup>th</sup> standard week.

was recorded at 4<sup>th</sup> week. The lowest morning mean relative humidity (87.7 %) was registered at 38<sup>th</sup> standard week. The lowest evening mean relative humidity (66.2 %) was observed at 6<sup>th</sup> standard week and the highest evening mean relative humidity (87.4 %) was recorded at 48<sup>th</sup> standard week. The highest cumulative sun shine hour (56.18) was recorded at 42<sup>nd</sup> standard week and the lowest sun shine hours (7.5 hrs) was noticed at 6<sup>th</sup> standard week. Higher value of mean evaporation 8.3 mm was recorded at 42<sup>nd</sup> standard week. Then the lowest mean evaporation (1.5 mm) was recorded at 5<sup>th</sup> standard week. The higher value of rainfall (202.6 mm) was observed at 48<sup>th</sup> standard week.

At the time of planting (40<sup>th</sup> standard week), the mean relative humidity of 90 per cent was recorded at 07.22 hrs and the mean relative humidity of 76 per cent was noticed at 14.22 hrs with a total rainfall of 7 mm. The mean maximum and minimum temperature were 33.5<sup>0</sup>C and 26.4 <sup>0</sup>C respectively. The mean evaporation rate was 6.3 mm and the cumulative sunshine hours were 41.2 hrs.

During 51<sup>st</sup> standard week (flowering), the mean maximum and minimum temperature were 31.1 <sup>0</sup>C and 23.6 <sup>0</sup>C respectively. The total rainfall was 50 mm with cumulative sunshine hours of 24.12 hrs.

During heading stage (2<sup>nd</sup> standard week), the mean evaporation rate was 1.7 mm and the cumulative sunshine hours was 30.5 mm. The mean maximum and minimum temperature were 31.0 <sup>0</sup>C and 22.8 <sup>0</sup>C respectively.

### **4.3. Soil temperature (°C)**

The mean data on soil temperature (°C) of flooded sodic soil in different treatments during crop growth period was recorded and presented in **Table 6**.

The mean soil temperature in different treatments was high (28.7 °C) at 75 DAT, while it was low during 60 DAT.

During early stage, the treatment T<sub>1</sub>, T<sub>7</sub> (FYM + GLM) and T<sub>5</sub> (GLM) registered significantly higher soil temperature of 28.8 °C followed by the treatments T<sub>4</sub> (28.7), T<sub>8</sub> (28.6) and T<sub>6</sub> (28.5) respectively. The treatments T<sub>1</sub>, T<sub>5</sub> and T<sub>7</sub> were on par with one another. The plots treated with BGA (T<sub>2</sub>) and *Azolla* (T<sub>3</sub>) recorded significantly lower value of soil temperature (28.4 °C) and other treatments were on par with each other.

At 15 days after transplanting (15 DAT), the plots treated with FYM + GLM (T<sub>7</sub>) recorded high soil temperature (28.5 °C) followed by T<sub>1</sub>, T<sub>4</sub> and T<sub>8</sub> which were on par. The plots treated with *Azolla* (T<sub>2</sub>) had shown significantly lower value of soil temperature (27.9 °C) than all treatments followed by T<sub>2</sub> and T<sub>6</sub>.

During 30 days after transplanting (30 DAT), the plots treated with FYM (T<sub>4</sub>), GLM (T<sub>5</sub>) and the plots treated with FYM + GLM (T<sub>7</sub>) recorded soil temperatures of 28.3 °C and were on par. The plots treated with *Azolla* (T<sub>3</sub>) registered the lower value of soil temperatures (27.8 °C) followed by BGA (T<sub>2</sub>) and BGA + *Azolla* (T<sub>6</sub>).

At maximum tillering stage (45 DAT), the control plots (T<sub>1</sub>) and the plots applied with FYM + GLM (T<sub>7</sub>) recorded higher soil temperature value (28.6 °C). The plots treated with BGA (T<sub>2</sub>), *Azolla* (T<sub>3</sub>) and BGA + *Azolla* (T<sub>7</sub>) registered the lowest water temperature of 28.2 °C. During flowering stage (75 DAT), the plots treated with *Azolla* (T<sub>3</sub>), BGA + *Azolla* (T<sub>7</sub>) recorded the lowest soil temperature (28.4 °C).

During the late growth stage (120DAT), the plots treated with GLM + FYM (T<sub>7</sub>) registered the highest soil temperature (28.7 °C). The plots treated with *Azolla* (T<sub>3</sub>) had shown the lowest soil temperature (28.2 °C).

#### **4.4. Water temperatures (°C)**

The mean data on water temperatures (°C) of flooded sodic soil in different treatments during crop growth period were recorded and presented in **Table 7**.

During early stages (15 DAT), the control plots (T<sub>1</sub>) had shown highest value of water temperature (31°C) followed by T<sub>5</sub>, T<sub>7</sub>, T<sub>2</sub>, T<sub>6</sub> and T<sub>3</sub>. The treatments T<sub>5</sub> and T<sub>7</sub> and T<sub>2</sub> and T<sub>6</sub> were at par. The plots treated with *Azolla* had shown significantly lower value of water temperature (30.3 °C) than all treatments.

At maximum tillering stage (45 DAT), the plots applied with FYM + GLM (T<sub>7</sub>) recorded higher water temperature value (30.7 °C) while *Azolla* applied plots (T<sub>3</sub>) recorded the lowest water temperature (30 °C).

During flowering stage (75 DAT), the plots treated with FYM + GLM (T<sub>7</sub>) recorded significantly higher water temperature (30.7 °C) and the same trend was also noticed during

120 days after transplanting, with GLM + FYM (T<sub>7</sub>) registered the highest water temperature (31.4 °C). The plots treated with *Azolla* (T<sub>3</sub>) had shown the lowest water temperature (30.2 °C).

#### 4.5. Redox potential (mV)

The redox potential (mV) values measured in different treatments during crop stages in the root region of rice plants are presented in the **Table 8**.

Redox potential values in the root region of rice had shown significant variation between treatments and the time of sampling. During planting, the average redox potential value of treated plots was -33 mV which showed decreasing trend during growth stages of rice in all treatments. The lowest redox potential value (-130 mV) was recorded at 60 DAT.

Among treatments, combined application of BGA and *Azolla* (T<sub>6</sub>) registered a redox potential value of -43 mV, followed by the plots treated with BGA (-53 mV) and *Azolla* (-60 mV) individually. The plots treated with FYM + GLM (T<sub>7</sub>) registered the lowest redox potential (-107 mV).

During maximum tillering stage (45 DAT), the plots treated with BGA + *Azolla* (T<sub>6</sub>) had shown a redox potential of 173 mV while it was -161mV in plots applied with FYM + GLM (T<sub>7</sub>). At flowering stage (75 DAT), the same trend was noticed.

At 120 DAT, the plots treated with BGA + *Azolla* (T<sub>6</sub>) recorded the highest redox potential of -29 mV. FYM + GLM (T<sub>7</sub>) treated plots recorded the lowest redox potential of -59mV.

#### 4.6. Dissolved oxygen concentration in the soil-floodwater interface

The dissolved oxygen concentrations (mg l<sup>-1</sup>) in different treatments of flooded sodic soil are given in **Table 9**. Significant variation in dissolved oxygen concentration in rice field was noticed between treatments and growth stages.

The average dissolved oxygen concentration of treated plots showed decreasing trend during growth stages in all the treatments. The lowest dissolved oxygen concentration of 0.92 mg l<sup>-1</sup> was noticed during 75 DAT.

Among the treatments T<sub>2</sub> (BGA) and T<sub>3</sub> (*Azolla*) registered mean maximum dissolved oxygen concentration of 2.05 mg l<sup>-1</sup> and 2.03 mg l<sup>-1</sup>, respectively. The plots applied with FYM + GLM (T<sub>7</sub>) recorded the lowest dissolved oxygen concentration of 1.31 mg l<sup>-1</sup>.

At 45 DAT (maximum tillering stage) and 75 DAT (flowering stage), the plots with BGA + *Azolla* (T<sub>6</sub>) recorded the highest dissolved oxygen concentration of 1.95 and 1.27 mg l<sup>-1</sup> respectively while the plots treated with FYM + GLM (T<sub>7</sub>) recorded the lowest dissolved oxygen concentration (1.03 mg l<sup>-1</sup>).

Also during maturity stage (120 DAT), BGA + *Azolla* (T<sub>6</sub>) application enhanced the dissolved oxygen content which was 2.06 mg l<sup>-1</sup> while green leaf manure application (T<sub>5</sub>) resulted in a dissolved oxygen concentration of 1.04 mg l<sup>-1</sup>.

#### 4.7. Methane flux

The data on methane emissions from different treatments in flooded sodic soil are presented in the **Table 10**. Methane flux pattern varied significantly both in treatments and time of sampling.

The mean methane emission was lowest at the time of planting (8.25 mg m<sup>-2</sup> day<sup>-1</sup>) and progressed steadily with the time period. The emission was highest on 75 DAT (90.64 mg m<sup>-2</sup> day<sup>-1</sup>) which was 47.03 mg m<sup>-2</sup> day<sup>-1</sup> during 60<sup>th</sup> DAT. A decreasing trend in methane emission was noticed towards harvest.

Among the treatments, plots which received FYM + GLM (T<sub>7</sub>) recorded the highest methane emissions of 58.54 mg m<sup>-2</sup> day<sup>-1</sup>, followed by T<sub>5</sub> and T<sub>4</sub>. (53.49 and 50.08 mg m<sup>-2</sup> day<sup>-1</sup> respectively). The control plot with out organic manure recorded mean methane emission of 48.81 mg m<sup>-2</sup> day<sup>-1</sup>, while Blue green algal and *Azolla* applied plots registered the lowest emission in all growth stages with a mean emission of 30.30 mg m<sup>-2</sup> day<sup>-1</sup>.

During maximum tillering stage (45 DAT), the mean methane emission was 35.16 mg m<sup>-2</sup> day<sup>-1</sup>. The plots treated with FYM + GLM recorded the highest value of methane flux (49.03 mg m<sup>-2</sup> day<sup>-1</sup>) while BGA + *Azolla* treatment registered the lowest methane flux of 22.56 mg m<sup>-2</sup> day<sup>-1</sup> during maximum tillering stage. The same trend was also noticed during flowering stage (75 DAT) with highest methane emissions of 122.69 mg m<sup>-2</sup> day<sup>-1</sup> recorded in the plots treated with FYM + GLM (T<sub>7</sub>).

Prior to harvest, during 120 DAT, the plots treated with FYM + GLM (T<sub>7</sub>) recorded the highest methane flux of 55.36 mg m<sup>-2</sup> day<sup>-1</sup>. The plots applied with BGA + *Azolla* registered the lowest methane flux of 30.66 mg m<sup>-2</sup> day<sup>-1</sup>.

## **4.8. Yield attributes**

### **4.8.1. Productive tillers m<sup>-2</sup>**

The mean data on Productive tillers m<sup>-2</sup> in rice under different treatments were recorded during physiological maturity and are presented in **Table 11**.

The number of productive tillers m<sup>-2</sup> differed significantly between treatments. The plots treated with BGA + *Azolla* + FYM + GLM (T<sub>8</sub>) produced significantly higher number of productive tillers (413) followed by T<sub>6</sub> (395), T<sub>2</sub> (378), T<sub>7</sub> (368), T<sub>3</sub> (357), T<sub>4</sub> (349), T<sub>5</sub> (341) and T<sub>1</sub> (273). The control plots recorded the lowest number of productive tillers.

### **4.8.2. Panicle length**

The mean data on length of panicle (cm) in different treatments in rice field were recorded and presented in **Table 11**.

In general the plots treated with BGA + *Azolla* + FYM + GLM (T<sub>8</sub>) registered more panicle length (24.49 cm) followed by the treatments T<sub>6</sub> (24.01 cm) and T<sub>2</sub> (23.80 cm). The plots treated with BGA (T<sub>2</sub>) and BGA + *Azolla* (T<sub>6</sub>) recorded 23.8cm and 24.01cm of panicle length and they were at par. The treatments T<sub>2</sub> (23.80 cm) and T<sub>7</sub> (23.66 cm) were at par. The treatments T<sub>3</sub> (23.44 cm), T<sub>4</sub> (23.39 cm) and T<sub>7</sub> (23.66 cm) were at par. Similarly the treatments T<sub>3</sub> (23.44 cm), T<sub>4</sub> (23.39 cm) and T<sub>5</sub> (23.30 cm) were at par. The control plot (T<sub>1</sub>) recorded significantly lower of panicle length (22.23 cm).

### **4.8.3. Panicle weight (g)**

The mean data on Panicle weight (g) are presented in **Table 11**.

The plot treated with BGA + *Azolla* + FYM + GLM (T<sub>8</sub>) recorded significantly more panicle weight (5.72g) followed by T<sub>6</sub>, T<sub>7</sub>, T<sub>2</sub>, T<sub>3</sub>, T<sub>4</sub>, T<sub>5</sub> and T<sub>1</sub>. The plots applied with BGA (T<sub>2</sub>) and the plots treated with *Azolla* recorded 5.15g and 5.10g respectively and were at par. The control plots (T<sub>1</sub>) had shown significantly the lowest panicle weight (4.71g) than other treatments.

### **4.8.4. Filled grains hill<sup>-1</sup>**

The mean data on Filled grains hill<sup>-1</sup> in different treatments are documented in **Table 12** and maximum number of filled grains hill<sup>-1</sup> (183) was observed in plots treated with BGA + Azolla + FYM + GLM (T<sub>8</sub>). The plots applied with BGA + Azolla (T<sub>6</sub>) and the plots applied with BGA (T<sub>2</sub>) recorded 174 and 164 hill<sup>-1</sup> respectively. The treatments T<sub>5</sub>, T<sub>3</sub> and T<sub>4</sub> registered 158, 156 and 154 hill<sup>-1</sup> respectively and they were at par. The control plots (T<sub>1</sub>) had shown significantly the lowest number of filled grains hill<sup>-1</sup> (146).

#### **4.8.5. Ill-filled grains hill<sup>-1</sup>.**

The mean data on ill-filled grains hill<sup>-1</sup> are presented in **Table 12**.

The control plots (T<sub>1</sub>) recorded significantly more number of ill-filled grains hill<sup>-1</sup> (17) followed by the plots treated with GLM (T<sub>5</sub>) (16) and FYM (T<sub>4</sub>) (15). The treatments T<sub>3</sub> and T<sub>4</sub> produced 14 ill-filled grains hill<sup>-1</sup> and they were at par. The plots applied with BGA + Azolla + FYM + GLM (T<sub>8</sub>) recorded significantly lower number of ill filled grain hill<sup>-1</sup> (13).

#### **4.8.6. Test weight (g)**

The mean data on test weight (g) of the grain for different treatments are presented in **Table 12**. The highest (23.61g) and lowest (23.56g) 1000 grain weight was observed in the plots treated with BGA + Azolla + FYM + GLM (T<sub>8</sub>) and control (T<sub>1</sub>) respectively. The plots T<sub>3</sub> and T<sub>7</sub> recorded 23.6 g and there was no significant influence between treatments.

#### **4.8.7. Grain yield**

The mean data on grain yield kg ha<sup>-1</sup> in different treatments in sodic soils of flooded rice field are presented in **Table 13**.

The plots treated with BGA + Azolla + FYM + GLM (T<sub>8</sub>) produced significantly higher grains yield (3847 kg ha<sup>-1</sup>). The treatments T<sub>2</sub> (3646 kg ha<sup>-1</sup>) and T<sub>6</sub> (3581 kg ha<sup>-1</sup>) were at par. The treatments T<sub>3</sub> (3287 kg ha<sup>-1</sup>), T<sub>4</sub> (3255 kg ha<sup>-1</sup>) and T<sub>5</sub> (3188 kg ha<sup>-1</sup>) were also at par. The control plots (T<sub>1</sub>) registered the lowest grain yield (3040 kg ha<sup>-1</sup>).

#### **4.8.8. Straw yield (Kg ha<sup>-1</sup>)**

The mean data on straw yield are presented in **Table 13**.

The plots applied with BGA + *Azolla* + FYM + GLM (T<sub>8</sub>) produced significantly high straw yield (5778 Kg ha<sup>-1</sup>) when compared to other treatments and the control plots (T<sub>1</sub>) produced the lowest straw yield (4668 Kg ha<sup>-1</sup>).

#### **4.8.9. Harvest Index**

The mean data on Harvest Index in different treatments are presented in **Table 13**.

The plots treated with BGA recorded significantly the highest value of harvest index (40.7). The treatments T<sub>2</sub>, T<sub>6</sub>, T<sub>7</sub> and T<sub>8</sub> recorded significantly higher value of harvest index that were 40.7, 40.5, 40, and 39.4 respectively and were at par. The treatments T<sub>1</sub>, T<sub>6</sub> and T<sub>8</sub> were also at par. The treatments T<sub>1</sub>, T<sub>3</sub>, T<sub>4</sub> and T<sub>5</sub> recorded the lowest value of harvest index when compared with other treatments.

#### **4.9. Influence of temperature, redox potential and dissolved oxygen on methane flux**

The influence of soil temperature, redox potential and dissolved oxygen concentration on mean methane flux in different treatments is summarized in **Table 14**.

The mean soil and water temperature were low in blue green algal and *Azolla* applied plots (28.2 and 28<sup>0</sup>C respectively) and higher in farm yard manure and green leaf manure applied plots. The mean dissolved oxygen concentration and redox potential values were high in plots applied with blue green alga and *Azolla* (2.20 mg l<sup>-1</sup> and -43 mV respectively) while green leaf and farm yard manure applied plots recorded low dissolved oxygen and redox values ( 1.31 mg l<sup>-1</sup> and -107 mV respectively).

The higher dissolved oxygen and redox values in BGA and *Azolla* applied plots recorded lowest methane flux value of 30.30 mg m<sup>-2</sup> day<sup>-1</sup> while green leaf and farm yard manure applied plots recorded higher methane flux ( 58.54 mg m<sup>-2</sup> day<sup>-1</sup> ) due to lower dissolved oxygen and redox status.

## CHAPTER V

### DISCUSSION

The present investigation entitled “Effect of temperature, redox and dissolved oxygen on methane flux of rice soil ecosystem and designing mitigation strategies to minimize global warming potential” was conducted at Anbil Dharmalingam Agricultural College and Research Institute, Trichy, during the *Rabi* seasons of 2010-11.

The results of the experiment presented in the preceding chapter have been discussed and elucidated in this chapter with the help of suitable reasons and evidences available on the topic of investigation. The significant results, where ever found to be necessary, have also been illustrated through bar and line diagrams in order to predict the effect of experimental factors. In order to make the things more illustrative, the factors and possible reasons of variation obtained due to treatment differences have been discussed in this chapter according to the objectives of the present investigation.

The weather parameters prevailed during crop growing period are presented in **Fig. 2**.

#### **5.1. Influence of organic manure and photosynthetic systems on soil /water temperature and methane flux.**

The field experiment was conducted to study the influence of temperature (air, soil and water) under different organic amendments on methane flux in rice cultivation. As redox potential, soil temperature and dissolved oxygen in the flooded rice soil are major factors influencing the methane flux; their concentrations were monitored in all treatments throughout the growth stages to derive the correlation between temperature/ redox/ dissolved oxygen and methane flux.

Minimizing CH<sub>4</sub> flux in rice cultivation is an important climate change mitigation strategy and hence the influence of photosynthetic systems such as blue green algae (BGA) and *Azolla* on soil redox, dissolved oxygen and CH<sub>4</sub> emission was studied. As BGA and *Azolla* supply nitrogen and other growth regulators to the rice crop besides CH<sub>4</sub> emission reduction, their role in enhancing the yield in rice cultivation was also quantified in the field experiment.

The experimental soil was sandy clay loam in texture with the pH of 9.1. The organic carbon content was medium with low N and medium K levels. The plots applied with farm yard manure and green leaf manure separately (T<sub>4</sub> and T<sub>5</sub>) and also in combination (T<sub>7</sub>) recorded higher soil and water temperature (**Fig.3 & 4**). The decomposition of organics and mineralization processes would have enhanced the soil temperature in these plots. The plots treated with BGA and *Azolla* registered lower soil and water temperature and the same trend was also noticed during all growth stages such as maximum tillering, flowering and maturity stages. The BGA and *Azolla* form a mat over the water surface and minimize the penetration of solar radiation. More over BGA and *Azolla* being photosynthetic systems releases oxygen into soil water interface that ultimately minimizes the water and soil temperature in the experimental plots. Prasanna *et al.* (2002) reported that cyanobacteria releases oxygen during photosynthesis into the standing water that can diffuse into soil.

Nouchi *et al.* (1994) attempted to model the methane flux from rice paddies using a diffusion model. They suggested that the conductance of rice plants for methane transport is very sensitive to temperature. It is well known that temperature strongly affects methanogenesis (Lindau *et al.*, 1993) and increasing temperature leads to higher methane emission. Methane flux was reached maximum at higher temperature in water logged soils. The rate of methane formation was very small below 20 °C, because the conversion rate of substrate to CH<sub>4</sub> depends on the maximum and minimum temperature. (Schutz *et al.*, 1989). In the present investigation, the marginal reduction in soil and water temperature in BGA and *Azolla* applied plots as a result of higher oxygen diffusion would be one among the factors that contributed to low methane flux. In general, the simple diffusion coefficient of dissolved gases in solution increases with increasing temperature. Hosono and Nouchi (1997) reported that the process by which methane enters in to rice root from the root surface in contact with water is affected by the temperature which is confirmed by the results of the present investigation.

In the present study the air temperature surrounding the top of the rice plant had relatively small effect on conductance of methane in to atmosphere by rice plants. Interestingly the higher methane flux in all treatments was recorded during 51 and 52 standard weeks (60-75 DAT), that also registered high mean maximum and minimum air

temperature. Hosono and Nouchi (1997) reported that increase in the air temperature slightly influenced the methane conductance.

### **5.3. Influence of organic manure and photosynthetic systems on soil redox potential (Eh) of experimental field.**

Redox status of soil is an indirect indicator of methane flux pattern from rice ecosystem (Wang *et al.*, 1993) and soils with lower redox potential are usually associated with high methane flux. Hence the redox potential was measured in all the treatments during different crop growth stages (**Fig.5**). The combined application of BGA and *Azolla* recorded higher redox potential followed by the application of BGA and *Azolla* individually in all growth stages of rice in the experimental plots. The redox potential value was the lowest in treatments applied with farm yard manure and green leaf manure (T<sub>7</sub>) which are evident by the low mean redox potential of -107mV in T<sub>7</sub>. Methane production mostly occurs in the soil microenvironment where the redox status is expected to be lower (Neue, 1993). Bharati and Mohanty (2000) found that *Azolla* dual cropping in rice registered a higher redox potential leading to low methane flux under Blue green algal application in rice fields. The results of the present investigation are in line with the earlier findings that BGA and *Azolla* application in rice cultivation as biofertilizers minimize methane flux by enhancing the soil redox that is unfavorable to methane generating methanogens.

### **5.4. Influence of organic manure and photosynthetic systems on dissolved oxygen and methane flux**

Dissolved oxygen is important parameter that plays a major role in methane emission from rice field. The lower level of flooded water dissolved oxygen is associated with higher methane emission. In the present investigation significant variation in dissolved oxygen concentration was observed between treatments. The dissolved oxygen concentration showed a decreasing trend in all treatments during growth stages and this might be due to the enhanced microbial activity in rice soil rhizosphere and this is in line with the earlier results of Sethunathan *et al.* (2000).

The BGA and *Azolla* application individually and in combination enhanced the dissolved oxygen concentration in the standing water in all growth stages while the

dissolved oxygen concentration was minimum in farm yard manure and green leaf manure applied plots. BGA and *Azolla* are aerobic photosynthetic organisms and in the medium of their growth, they release a lot of oxygen during photosynthesis. As a result when they grow in rice fields they make the standing water highly oxygenated. When there is profuse growth of BGA and *Azolla*, the surface layer of the soil absorbs enough oxygen through diffusion to become aerobic in nature and prevents the development of highly reduced conditions underneath it. Mandal *et al.* (1998) and Lakshmanan *et al.* (1994) reported similar findings that BGA application increased the dissolved oxygen content in the standing water of rice field. Prasanna *et al.* (2002) also reported the beneficial effect of cyanobacteria in decreasing the headspace concentration of methane due to higher dissolved oxygen concentration that enhanced the methane oxidation at source.

## **5. 6. Influence of organic manure and photosynthetic systems on methane flux**

The mean methane emission rate exhibited variation between treatments and growth stages. The methane fluxes were maximum during flowering stage in all treatments and showed decreasing trend towards maturity. Purkait *et al.* (2005) reported that methane flux varies with the age of the rice crop. In case of Rabi crop, the emission rate starts increasing a few days after transplanting and becomes maximum in 60 days and then decreases. In the present study in all treatments the peak methane flux was observed at flowering stage of the crop. Methane flux was low in all plots during the first week after transplantation (**Fig.6**). The higher rates of CH<sub>4</sub> production during the flowering stage was due to the degradation of the available organic carbon in the form of root exudates. Low CH<sub>4</sub> flux during the early growth stage of the rice plant was due to low levels of methanogenesis and poor conduction of CH<sub>4</sub> from the soil to the atmosphere. (Nouchi, 1994).

Rice plants make at least two main contributions to the process of methane emission. First, rice plants release organic substances into the rhizosphere by biomass litter and root exudation. Second, rice plants provide channels for gas transport. Like other vascular plants rooted in anoxic sediments, rice plants are thought to release oxygen into the rhizosphere (De Bont *et al.*, 1978), supporting methane oxidation. Meanwhile methane produced in the sediments diffuses into the cell-wall water of the root cells, gasifies into root cortex and then is mostly related through the microspores in the leaf

sheaths into atmosphere. The CH<sub>4</sub> flux pattern during rice growth stages recorded in this study is found to agree with the earlier reports of Nouchi (1994); Sass and Fisher (1996).

The methane emission was found to be maximum in farm yard manure and green leaf manure applied plots in all growth stages. Easily degradable organic substrates are the main sources of methane generation in wet land rice soils and hence application of farm yard manure and green leaf manure triggered methane emission in the present investigation. Neue and Roger (1993) obtained similar results with various organic manures in their field study. Watanabe *et al.*, (1989) also reported that incorporation of organic manure resulted in higher methane flux.

Blue green algal and *Azolla* application recorded the lowest methane flux in all growth stages. The higher dissolved oxygen and redox values recorded in algal and *Azolla* applied plots resulted in low methane emission values. Moreover BGA can promote aerobic transformation such as methane oxidation through enhanced aeration of the flood water in rice fields as reported by Prasanna *et al.* (2002). Oxygen released during photosynthesis by cyanobacteria (BGA) in to the standing water can diffuse in to the soil and provide aerobic condition, not congenial for methanogenesis. Mandal *et al.* (1993) also documented the beneficial effects of BGA and *Azolla* in minimizing methane flux in wetland rice fields through enhancing the dissolved oxygen content in the standing water. Cyanobacteria are important components of wetland rice ecosystem and their role as biofertilizers may be more significant if they also contribute as oxygen donors for methanotrophs (Bodelier *et al.*, 2000).

### **5.7. Effect of organic manure and photosynthetic systems on rice yield.**

Rice yield was significantly higher in the plots applied with organic manure (FYM and GLM) and biofertilizers (BGA and *Azolla*). Even though the methane flux is found to be high due to organics, application of organic manure is encouraged in rice cultivation due to higher yield and soil health. In the present study, combined application of organics and blue green algae not only recorded higher yield, but found to emit less methane in paddy cultivation than the application of organics alone (**Fig. 7 & 8**). The mean methane flux in farm yard manure and green leaf manure applied plot was 58.54 mg m<sup>2</sup> day<sup>-1</sup>, while the flux was reduced to 20 per cent due to BGA and *Azolla* application (46.37 mg m<sup>2</sup> day<sup>-1</sup>). Bharati and Mohanty (2000) emphasized that

application of BGA and *Azolla* reduced methane flux without reducing rice yields and can be used as a practical mitigation option for minimizing the global warming potential of rice ecosystem. The present field study reiterates that biofertilization of paddy fields with blue green algae and azolla is a potential climate change mitigation strategy due to their effect in minimizing methane emission, besides yield enhancement by nitrogen fixation.

## CHAPTER VI

### SUMMARY AND CONCLUSION

Agriculture accounts for approximately one-fifth of the annual increase in anthropogenic greenhouse gases and rice cultivation has been accredited as one of the most important sources of anthropogenic methane. The present investigation was carried out to study the effect of temperature, redox and dissolved oxygen on methane flux of rice soil ecosystem and designing mitigation strategies to minimize global warming potential, during *Rabi* season, 2010-11 at Anbil Dharmalingam Agricultural College and Research Institute, Trichy. Methane efflux observation was tested in three replicated randomized block design. All other agronomic practices were adapted as per normal recommendations.

The observations on methane flux, yield attributes and yield characters were recorded and the data collected so far, averaged and tabulated for statistical analysis. Further, homogeneity of the data was tested and if the data was found homogenous the pooling was done. The results based on pooled analysis of homogenous data have been described and discussed in previous two chapters. The salient results of present investigation on variation in soil redox, dissolved oxygen, methane flux, yield attributes and yield are summarized here under:

The experimental soil was sandy clay loam in texture with the pH of 9.1. The organic carbon content was medium with low N and medium K levels.

The plots applied with farm yard manure and green leaf manure separately (T<sub>4</sub> and T<sub>5</sub>) and also in combination (T<sub>7</sub>) recorded higher soil and water temperature. Marginal reduction in soil and water temperature in BGA and *Azolla* applied plots as a result of higher oxygen diffusion would be one among the factors that contributed to low methane flux.

The air temperature surrounding the top of the rice plant had relatively small effect on conductance of methane in to atmosphere by rice plants. The higher methane flux in all treatments was recorded between 60 and 75 days after transplanting, which also registered high mean maximum and minimum air temperature.

Redox status of soil and dissolved oxygen content in the standing water are indirect indicators of methane flux pattern from rice ecosystem. The combined application of BGA and *Azolla* recorded higher redox potential followed by the application of BGA and *Azolla* individually in all growth stages of rice in the experimental plots. The redox potential value was the lowest in treatments applied with farm yard manure and green leaf manure (T<sub>7</sub>) which are evident by the low mean redox potential of -107mV in T<sub>7</sub>.

BGA and *Azolla* application individually and in combination enhanced the dissolved oxygen concentration in the standing water in all growth stages while the dissolved oxygen concentration was minimum in farm yard manure and green leaf manure applied plots. When there is profuse growth of BGA and *Azolla*, the surface layer of the soil absorbs enough oxygen through diffusion to become aerobic in nature and prevents the development of highly reduced conditions underneath it.

The mean methane emission rate exhibited variation between treatments and growth stages. The methane fluxes were maximum during flowering stage in all treatments and showed decreasing trend towards maturity. Low CH<sub>4</sub> flux during the early growth stage of the rice plant was due to low levels of methanogenesis and poor conduction of CH<sub>4</sub> from the soil to the atmosphere. The higher rates of CH<sub>4</sub> production during the flowering stage was due to the degradation of the available organic carbon in the form of root exudates.

The methane emission was found to be maximum in farm yard manure and green leaf manure applied plots in all growth stages. Blue green algal and *Azolla* application recorded the lowest methane flux in all growth stages. The higher dissolved oxygen and redox values recorded in algal and *Azolla* applied plots resulted in low methane emission values.

Combined application of organics and blue green algae not only recorded higher yield, but found to emit less methane in paddy cultivation than the application of organics alone.

The mean methane flux in farm yard manure and green leaf manure applied plot was  $58.54 \text{ mg m}^{-2} \text{ day}^{-1}$ , while the flux was reduced to 20 per cent due to BGA and *Azolla* application ( $46.37 \text{ mg m}^{-2} \text{ day}^{-1}$ ).

The present investigation gives evidence that Blue green algae and *Azolla* could reduce global warming potential from flooded rice soils at the levels of green house gas production, transport and oxidation besides enhancing the yield.

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# **Effect of temperature on methane flux of rice soil ecosystem and designing mitigation strategies to minimize global warming potential**

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## **Research Findings**

The present investigation was conducted at Anbil Dharmalingam Agricultural College and Research Institute, Trichy, during the *Rabi* seasons of 2010-11. It is well known that temperature strongly affects methanogenesis and increasing temperature leads to higher methane emission. In the present investigation, the marginal reduction in soil and water temperature in BGA and *Azolla* applied plots as a result of higher oxygen diffusion would be one among the factors that contributed to low methane flux. The air temperature surrounding the top of the rice plant had relatively small effect on conductance of methane in to atmosphere by rice plants. Interestingly the higher methane flux in all treatments was recorded during 51 and 52 standard weeks (60-75 DAT), that also registered high mean maximum and minimum air temperature.

Redox status of soil and dissolved oxygen content in the standing water are indirect indicators of methane flux pattern from rice ecosystem. The combined application of BGA and *Azolla* recorded higher redox potential followed by the application of BGA and *Azolla* individually in all growth stages of rice in the experimental plots. Similarly the BGA and *Azolla* application individually and in combination enhanced the dissolved oxygen concentration in the standing water in all growth stages while the dissolved oxygen concentration was minimum in farm yard manure and green leaf manure applied plots. BGA and *Azolla* application in rice cultivation as biofertilizers minimize methane flux by enhancing the soil redox and dissolved oxygen that are unfavorable to methane generating methanogens. The methane fluxes were maximum during flowering stage in all treatments and showed decreasing trend towards maturity. The higher rates of CH<sub>4</sub> production during the flowering stage were due to the degradation of the available organic carbon in the form of root exudates. The methane emission was found to be maximum in farmyard manure and green leaf manure applied plots in all growth stages. Blue green algal and *Azolla* application recorded the lowest methane flux in all growth stages. The higher dissolved oxygen and redox values recorded in algal and *Azolla* applied plots resulted in low methane emission values. In the present study, combined application of organics and blue green algae not only recorded higher yield, but found to emit less methane in paddy cultivation than the application of organics alone. The mean methane flux in farm yard manure and green leaf manure applied plot was 58.54 mg m<sup>2</sup> day<sup>-1</sup>, while the flux was reduced to 20% due to BGA and *Azolla* application (46.37 mg m<sup>2</sup> day<sup>-1</sup>). The present field study reiterates that biofertilization of paddy fields with blue green algae and *Azolla* is a potential climate change mitigation strategy due to their effect in minimizing methane emission, besides yield enhancement by nitrogen fixation.

**Table.4. Physiochemical properties of the experimental field**

<b>Parameters</b>	<b>Value</b>
<b>A. Mechanical analysis</b>	
Clay (%)	29.1
Silt (%)	13.1
Sand (%)	57.0
Textural class	Sandy clay loam
<b>B. Physical analysis</b>	
Field capacity (%)	43.25
Permanent wilting point (%)	32.51
Available soil moisture (%)	10.74
Bulk density ( $\text{Mg m}^{-3}$ )	1.39
<b>C. Chemical analysis</b>	
pH (1: 2 of soil : water)	9.1
Electrical conductivity ( $\text{dS m}^{-1}$ )	0.35
Organic carbon (%)	0.49
Available nitrogen ( $\text{kg ha}^{-1}$ )	191.0
Available phosphorus ( $\text{kg ha}^{-1}$ )	27.5
Available potassium ( $\text{kg ha}^{-1}$ )	240.0
<b>D. Biological properties</b>	
Bacteria ( $10^8$ CFU $\text{g}^{-1}$ of soil)	16
Fungi ( $10^4$ CFU $\text{g}^{-1}$ of soil)	6
Actinomycetes ( $10^3$ CFU $\text{g}^{-1}$ of soil)	3
Dehydrogenase ( $\mu\text{g}$ of TPF $\text{g}^{-1}$ of soil)	5.8
Phosphatase ( $\mu\text{g}$ of PNPP $\text{g}^{-1}$ of soil)	11.2
Urease ( $\mu\text{g NH}_4\text{-N g}^{-1}$ of soil $\text{h}^{-1}$ )	8.8

**Table.5. weather prevailed during crop growing period**

Std. week	Date	Mean Temp (°C)		Mean RH (%)		R.F (mm)	Evaporation (mm)	Sun shine (hr)
		Max.	Min.	7.22 hrs	14.22 hrs			
37	Sep. 09-15	35.5	23.4	92	81	50	3.0	24.12
38	Sep. 16-22	35.6	23.4	87.7	77	27.6	4.1	32.2
39	Sep. 23-29	34	23.1	91.2	80.7	36.6	4.0	29.5
40	Sep. 30-Oct.06	33.5	26.4	90	75.7	7	6.3	41.2
41	Oct.07-13	35	27.2	89.2	73.2	22	7.7	49.1
42	Oct.14-20	36.1	26.4	87.8	73.7	6	8.3	56.1
43	Oct. 21-27	35.4	25.8	90.1	70.4	3	6.2	38.4
44	Oct.28- Nov.03	35.8	24.8	92.8	81.4	70.6	5.9	37.7
45	Nov. 04-10	35	23.8	91.2	81.2	10.5	5.8	35.3
46	Nov. 11-17	34.3	23.6	91.2	85.8	41.5	3.9	31.3
47	Nov. 18-24	32	20.9	93	85.5	51.3	4.5	30.7
48	Nov. 25-Dec.01	30.5	22.4	93.7	87.4	202.6	2.7	20.3
49	Dec. 02-08	31.1	24.3	90.2	84.4	46.6	3.0	23.2
50	Dec. 09-15	36.2	27.2	93.8	83.2	17.3	3.8	51.8
51	Dec. 16-22	36.4	27.5	93.5	82.8	31.2	2.1	23.5
52	Dec. 23-29	29.5	24.1	93.8	78.7	2.2	3.2	48.7
1	Dec.30-Jan.05	30.5	24.2	93.2	84.7	10	1.8	26.4
2	Jan. 06-12	31	22.8	94.7	86.4	0	1.7	30.5
3	Jan. 13-19	30.8	21.7	89.2	80.5	0	2.4	27.2
4	Jan. 20-26	30.5	19.4	95.4	84.7	0	2.2	68.7
5	Jan.27- Feb.02	30.6	22.2	94.1	66.5	0	1.5	52.6
6	Feb. 03-09	31.8	20.3	94.7	66.2	0	1.9	39.8

**Table.10. Methane flux ( $\text{mg m}^{-2} \text{ day}^{-1}$ ) of experimental plots planted to rice.**

Treatment	Days After Transplanting									
	0	15	30	45	60	75	90	105	120	Mean
T <sub>1</sub>	7.82	11.05	16.25	39.21	51.52	97.45	87.12	83.79	45.12	<b>48.81</b>
T <sub>2</sub>	7.81	8.64	14.28	25.27	37.71	71.98	61.98	58.65	36.98	<b>35.92</b>
T <sub>3</sub>	7.82	8.28	13.63	24.61	35.42	69.77	59.43	56.10	34.10	<b>34.35</b>
T <sub>4</sub>	8.47	11.10	16.65	40.28	52.20	99.00	89.00	85.66	48.33	<b>50.08</b>
T <sub>5</sub>	8.75	11.81	17.21	43.15	56.45	107.50	97.50	88.17	50.83	<b>53.49</b>
T <sub>6</sub>	7.81	7.82	12.75	22.56	30.80	61.33	51.00	48.00	30.66	<b>30.30</b>
T <sub>7</sub>	9.56	12.68	18.25	49.03	64.89	122.69	102.69	91.69	55.36	<b>58.54</b>
T <sub>8</sub>	7.97	10.84	15.56	37.18	47.26	95.39	83.72	79.72	39.72	<b>46.37</b>
<b>Mean</b>	<b>8.25</b>	<b>10.28</b>	<b>15.57</b>	<b>35.16</b>	<b>47.03</b>	<b>90.64</b>	<b>79.06</b>	<b>73.97</b>	<b>42.64</b>	
<b>S.Ed.</b>	<b>0.18</b>	<b>0.05</b>	<b>0.06</b>	<b>0.69</b>	<b>0.74</b>	<b>0.85</b>	<b>0.81</b>	<b>0.70</b>	<b>0.92</b>	
<b>CD (P=0.05)</b>	<b>0.38</b>	<b>0.11</b>	<b>0.13</b>	<b>1.48</b>	<b>1.58</b>	<b>1.81</b>	<b>1.75</b>	<b>1.50</b>	<b>1.97</b>	

T<sub>1</sub> - Control  
T<sub>2</sub> - BGA  
T<sub>3</sub> - Azolla  
T<sub>4</sub> - FYM

T<sub>5</sub> - GLM  
T<sub>6</sub> - BGA + Azolla  
T<sub>7</sub> - FYM + GLM  
T<sub>8</sub> - BGA + Azolla + FYM + GLM

**Table 8. Variation in redox potential (mV) in the root region of rice plants.**

Treatment	Days After Transplanting									
	0	15	30	45	60	75	90	105	120	Mean
<b>T<sub>1</sub></b>	-28	-56	-121	-138	-145	-96	-92	-55	-40	<b>-86</b>
<b>T<sub>2</sub></b>	-25	-37	-60	-86	-93	-65	-53	-31	-26	<b>-53</b>
<b>T<sub>3</sub></b>	-27	-41	-61	-90	-110	-76	-65	-38	-30	<b>-60</b>
<b>T<sub>4</sub></b>	-39	-58	-124	-137	-151	-102	-97	-56	-44	<b>-90</b>
<b>T<sub>5</sub></b>	-39	-63	-137	-149	-157	-116	-108	-64	-51	<b>-98</b>
<b>T<sub>6</sub></b>	-27	-31	-47	-70	-80	-55	-40	-20	-19	<b>-43</b>
<b>T<sub>7</sub></b>	-39	-70	-147	-161	-173	-128	-118	-70	-59	<b>-107</b>
<b>T<sub>8</sub></b>	-38	-46	-96	-119	-131	-89	-76	-45	-37	<b>-75</b>
<b>Mean</b>	<b>-32.75</b>	<b>-50.25</b>	<b>-99.12</b>	<b>-118.75</b>	<b>-130</b>	<b>-90.85</b>	<b>81.25</b>	<b>47.37</b>	<b>-38.25</b>	
<b>S.Ed.</b>	<b>0.48</b>	<b>0.39</b>	<b>1.54</b>	<b>6.62</b>	<b>0.93</b>	<b>1.17</b>	<b>0.50</b>	<b>2.33</b>	<b>0.42</b>	
<b>CD (P=0.05)</b>	<b>1.04</b>	<b>0.84</b>	<b>3.31</b>	<b>14.20</b>	<b>2.00</b>	<b>2.50</b>	<b>1.07</b>	<b>5.01</b>	<b>0.91</b>	

T<sub>1</sub> - Control  
T<sub>2</sub> - BGA  
T<sub>3</sub> - Azolla  
T<sub>4</sub> - FYM

T<sub>5</sub> - GLM  
T<sub>6</sub> - BGA + Azolla  
T<sub>7</sub> - FYM + GLM  
T<sub>8</sub> - BGA + Azolla + FYM + GLM

**Table 9. Dissolved oxygen concentration (mg l<sup>-1</sup>) in the soil-floodwater interface in flooded sodic soil planted to rice**

Treatment	Days After Transplanting									
	0	15	30	45	60	75	90	105	120	Mean
<b>T<sub>1</sub></b>	3.07	2.14	1.78	1.28	0.96	0.81	1.02	1.53	1.31	<b>1.54</b>
<b>T<sub>2</sub></b>	3.56	3.00	2.29	1.75	1.34	1.19	1.34	2.07	1.92	<b>2.05</b>
<b>T<sub>3</sub></b>	3.38	2.91	2.26	1.83	1.33	1.17	1.32	2.14	1.96	<b>2.03</b>
<b>T<sub>4</sub></b>	2.95	2.09	1.55	1.06	0.83	0.72	0.78	1.35	1.22	<b>1.39</b>
<b>T<sub>5</sub></b>	2.78	1.94	1.42	1.19	0.75	0.62	0.83	1.21	1.04	<b>1.31</b>
<b>T<sub>6</sub></b>	3.79	3.16	2.48	1.95	1.44	1.27	1.48	2.19	2.06	<b>2.20</b>
<b>T<sub>7</sub></b>	2.76	2.02	1.48	1.03	0.70	0.66	0.72	1.28	1.14	<b>1.31</b>
<b>T<sub>8</sub></b>	3.23	2.36	2.04	1.41	1.02	0.90	1.08	1.91	1.77	<b>1.75</b>
<b>Mean</b>	<b>3.19</b>	<b>2.45</b>	<b>1.91</b>	<b>1.44</b>	<b>1.05</b>	<b>0.92</b>	<b>1.07</b>	<b>1.71</b>	<b>1.55</b>	
<b>S.Ed.</b>	<b>0.02</b>	<b>0.01</b>	<b>0.10</b>	<b>0.02</b>	<b>0.03</b>	<b>0.01</b>	<b>0.01</b>	<b>0.02</b>	<b>0.02</b>	
<b>CD (P=0.05)</b>	<b>0.04</b>	<b>0.04</b>	<b>0.20</b>	<b>0.05</b>	<b>0.06</b>	<b>0.02</b>	<b>0.03</b>	<b>0.04</b>	<b>0.04</b>	

T<sub>1</sub> - Control  
T<sub>2</sub> - BGA  
T<sub>3</sub> - Azolla  
T<sub>4</sub> - FYM

T<sub>5</sub> - GLM  
T<sub>6</sub> - BGA + Azolla  
T<sub>7</sub> - FYM + GLM  
T<sub>8</sub> - BGA + Azolla + FYM + GLM

**Table 6. Soil Temperature (<sup>0</sup>C) prevailed during crop growing period of rice.**

	<b>T<sub>1</sub></b>	<b>T<sub>2</sub></b>	<b>T<sub>3</sub></b>	<b>T<sub>4</sub></b>	<b>T<sub>5</sub></b>	<b>T<sub>6</sub></b>	<b>T<sub>7</sub></b>	<b>T<sub>8</sub></b>	<b>Mean</b>
<b>0 DAT</b>	28.8	28.4	28.4	28.7	28.8	28.5	28.8	28.6	<b>28.6</b>
<b>15 DAT</b>	28.4	28.0	27.9	28.4	28.3	28.1	28.5	28.4	<b>28.3</b>
<b>30 DAT</b>	28.4	28.0	27.8	28.3	28.3	28.1	28.3	28.2	<b>28.2</b>
<b>45 DAT</b>	28.6	28.2	28.2	28.4	28.4	28.2	28.6	28.5	<b>28.4</b>
<b>60 DAT</b>	27.9	27.4	27.3	27.6	27.7	27.5	27.9	27.6	<b>27.6</b>
<b>75 DAT</b>	28.9	28.5	28.4	28.7	28.8	28.4	28.6	28.6	<b>28.6</b>
<b>90 DAT</b>	28.6	28.4	28.3	28.6	28.5	28.3	28.5	28.5	<b>28.5</b>
<b>105 DAT</b>	28.8	28.5	28.4	28.8	28.8	28.5	28.6	28.6	<b>28.6</b>
<b>120 DAT</b>	28.7	28.3	28.2	28.6	28.7	28.4	28.6	28.5	<b>28.5</b>
<b>Mean</b>	<b>28.5</b>	<b>28.2</b>	<b>28.0</b>	<b>28.4</b>	<b>28.4</b>	<b>28.1</b>	<b>28.5</b>	<b>28.3</b>	

T<sub>1</sub> - Control  
T<sub>2</sub> - Blue Green Algae (BGA)  
T<sub>3</sub> - Azolla  
T<sub>4</sub> - Farm Yard Manure (FYM)

T<sub>5</sub> - Green Leaf Manure (GLM)  
T<sub>6</sub> - BGA + Azolla  
T<sub>7</sub> - FYM + GLM  
T<sub>8</sub> - BGA + Azolla + FYM + GLM

\*DAT: Days after transplantation

**Table 7. Water temperature (<sup>0</sup>C) prevailed during crop growing period of rice.**

	<b>T<sub>1</sub></b>	<b>T<sub>2</sub></b>	<b>T<sub>3</sub></b>	<b>T<sub>4</sub></b>	<b>T<sub>5</sub></b>	<b>T<sub>6</sub></b>	<b>T<sub>7</sub></b>	<b>T<sub>8</sub></b>	<b>Mean</b>
<b>0 DAT</b>	30.9	30.3	30.3	30.8	30.8	30.5	30.9	30.6	<b>30.6</b>
<b>15 DAT</b>	31.0	30.5	30.3	30.8	30.9	30.5	30.9	30.7	<b>30.7</b>
<b>30 DAT</b>	30.4	29.7	29.6	30.1	30.3	29.8	30.3	30	<b>30.0</b>
<b>45 DAT</b>	30.6	30.2	30	30.5	30.6	30.2	30.7	30.5	<b>30.4</b>
<b>60 DAT</b>	30.6	30.0	29.9	30.5	30.6	30.1	30.6	30.3	<b>30.3</b>
<b>75 DAT</b>	30.6	30.0	29.9	30.5	30.6	30.1	30.7	30.3	<b>30.3</b>
<b>90 DAT</b>	30.9	30.3	30.1	30.9	31.0	30.4	30.9	30.7	<b>30.7</b>
<b>105 DAT</b>	31.1	30.4	30.2	31.1	31.1	30.6	31.2	30.8	<b>30.8</b>
<b>120 DAT</b>	31.1	30.4	30.2	31.2	31.2	30.6	31.4	30.7	<b>30.9</b>
<b>Mean</b>	<b>30.8</b>	<b>30.2</b>	<b>30.1</b>	<b>30.7</b>	<b>30.8</b>	<b>30.3</b>	<b>30.8</b>	<b>30.5</b>	

T <sub>1</sub>	-	Control	T <sub>5</sub>	-	Green Leaf Manure (GLM)
T <sub>2</sub>	-	Blue Green Algae (BGA)	T <sub>6</sub>	-	BGA + Azolla
T <sub>3</sub>	-	Azolla	T <sub>7</sub>	-	FYM + GLM
T <sub>4</sub>	-	Farm Yard Manure (FYM)	T <sub>8</sub>	-	BGA + Azolla + FYM + GLM

\*DAT: Days after transplantation

**Table 11. Number of productive tillers, length of panicle and panicle weight of above ground biomass of experimental field.**

<b>Treatments</b>	<b>Productive tillers/m<sup>2</sup></b>	<b>Length of panicle (cm)</b>	<b>Panicle weight (g)</b>
<b>T<sub>1</sub>- Control</b>	273	22.2	4.71
<b>T<sub>2</sub> - BGA</b>	378	23.8	5.15
<b>T<sub>3</sub> - Azolla</b>	357	23.4	5.10
<b>T<sub>4</sub> - FYM</b>	349	23.4	5.00
<b>T<sub>5</sub> - GLM</b>	341	23.3	4.84
<b>T<sub>6</sub> - BGA + Azolla</b>	395	24.0	5.41
<b>T<sub>7</sub> - FYM + GLM</b>	368	23.7	5.23
<b>T<sub>8</sub> - BGA + Azolla + FYM + GLM</b>	413	24.5	5.72
<b>SEd</b>	<b>3.6</b>	<b>0.13</b>	<b>0.04</b>
<b>CD (P=0.05)</b>	<b>7.71</b>	<b>0.29</b>	<b>0.08</b>

**Table 12. Treatments effects on filled grains, ill-filled grains and test weight of experimental plots planted to rice**

<b>Treatments</b>	<b>Filled grains/hill</b>	<b>Ill-filled grains/hill</b>	<b>Test weight (g)</b>
<b>T<sub>1</sub>- Control</b>	146	17	23.56
<b>T<sub>2</sub> - BGA</b>	165	14	23.60
<b>T<sub>3</sub> - <i>Azolla</i></b>	156	14	23.59
<b>T<sub>4</sub>- FYM</b>	154	15	23.59
<b>T<sub>5</sub>- GLM</b>	158	16	23.59
<b>T<sub>6</sub>- BGA + <i>Azolla</i></b>	175	13	23.6
<b>T<sub>7</sub>- FYM + GLM</b>	160	14	23.59
<b>T<sub>8</sub>- BGA + <i>Azolla</i> + FYM + GLM</b>	183	13	23.61
<b>SEd</b>	<b>1.48</b>	<b>0.24</b>	<b>0.02</b>
<b>CD (P=0.05)</b>	<b>3.17</b>	<b>0.51</b>	<b>NS</b>

**Table 13. Mean CH<sub>4</sub> flux, grain yield (at 14% moisture content), straw yield and harvest index of experimental field.**

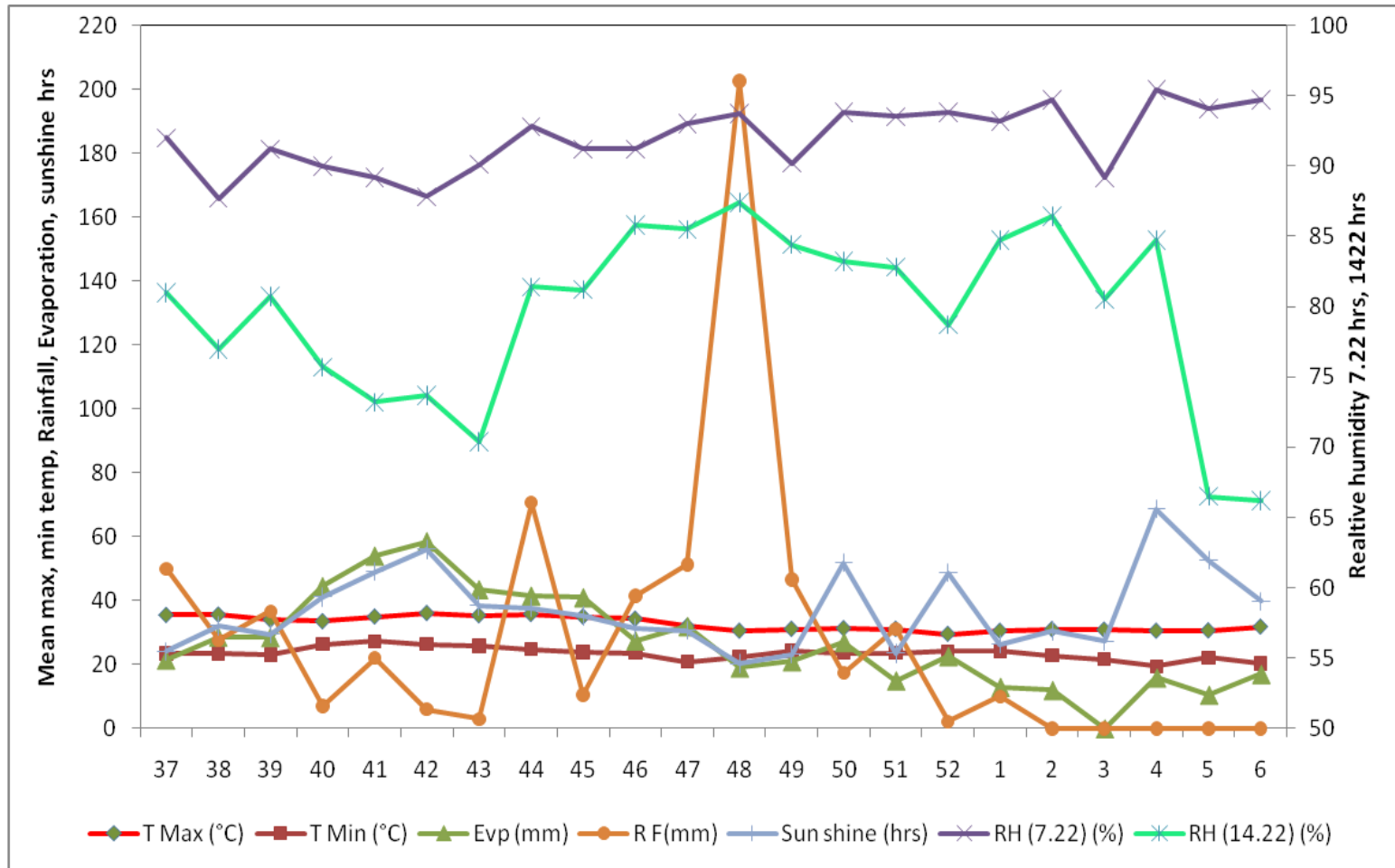
<b>Treatments</b>	<b>Mean CH<sub>4</sub> flux (mg m<sup>-2</sup> day<sup>-1</sup>)</b>	<b>Grain yield (kg ha<sup>-1</sup>)</b>	<b>Straw yield (kg ha<sup>-1</sup>)</b>	<b>Harvest Index</b>
<b>T<sub>1</sub>- Control</b>	48.81	3040	4668	39.4
<b>T<sub>2</sub> - BGA</b>	35.92	3646	5307	40.7
<b>T<sub>3</sub> - Azolla</b>	34.35	3287	5172	38.9
<b>T<sub>4</sub>- FYM</b>	50.08	3255	5099	39.0
<b>T<sub>5</sub> - GLM</b>	53.49	3188	5013	38.9
<b>T<sub>6</sub> - BGA + Azolla</b>	30.03	3685	5551	39.9
<b>T<sub>7</sub> - FYM + GLM</b>	58.54	3581	5250	40.5
<b>T<sub>8</sub> - BGA + Azolla + FYM + GLM</b>	46.37	3847	5778	40
<b>SEd</b>		<b>40.3</b>	<b>42.05</b>	<b>0.38</b>
<b>CD (P=0.05)</b>		<b>86.45</b>	<b>90.2</b>	<b>0.81</b>

**Table 14. Influence of temperature, redox potential and dissolved oxygen on methane flux.**

<b>Treatments</b>	<b>Mean soil temperature (°C)</b>	<b>Mean water temperature (°C)</b>	<b>Mean Dissolved Oxygen mg l<sup>-1</sup></b>	<b>Mean redox potential (mV)</b>	<b>Mean methane flux (mg m<sup>-2</sup> day<sup>-1</sup>)</b>
<b>T1</b>	28.5	30.8	1.54	-86	48.81
<b>T2</b>	28.2	30.2	2.05	-53	35.92
<b>T3</b>	28.0	30.1	2.03	-60	34.35
<b>T4</b>	28.4	30.7	1.39	-90	50.08
<b>T5</b>	28.4	30.8	1.31	-98	53.49
<b>T6</b>	28.1	30.3	2.20	-43	30.30
<b>T7</b>	28.5	30.8	1.31	-107	58.54
<b>T8</b>	28.3	30.5	1.75	-75	46.37

- |                |   |                        |                |   |                          |
|----------------|---|------------------------|----------------|---|--------------------------|
| T <sub>1</sub> | - | Control                | T <sub>5</sub> | - | Green Leaf Manure (GLM)  |
| T <sub>2</sub> | - | Blue Green Algae (BGA) | T <sub>6</sub> | - | BGA + Azolla             |
| T <sub>3</sub> | - | Azolla                 | T <sub>7</sub> | - | FYM + GLM                |
| T <sub>4</sub> | - | Farm Yard Manure (FYM) | T <sub>8</sub> | - | BGA + Azolla + FYM + GLM |

**Fig.2. Weather prevailed during crop growing period**



**Fig.8. Influence of temperature, redox potential and dissolved oxygen on methane flux.**

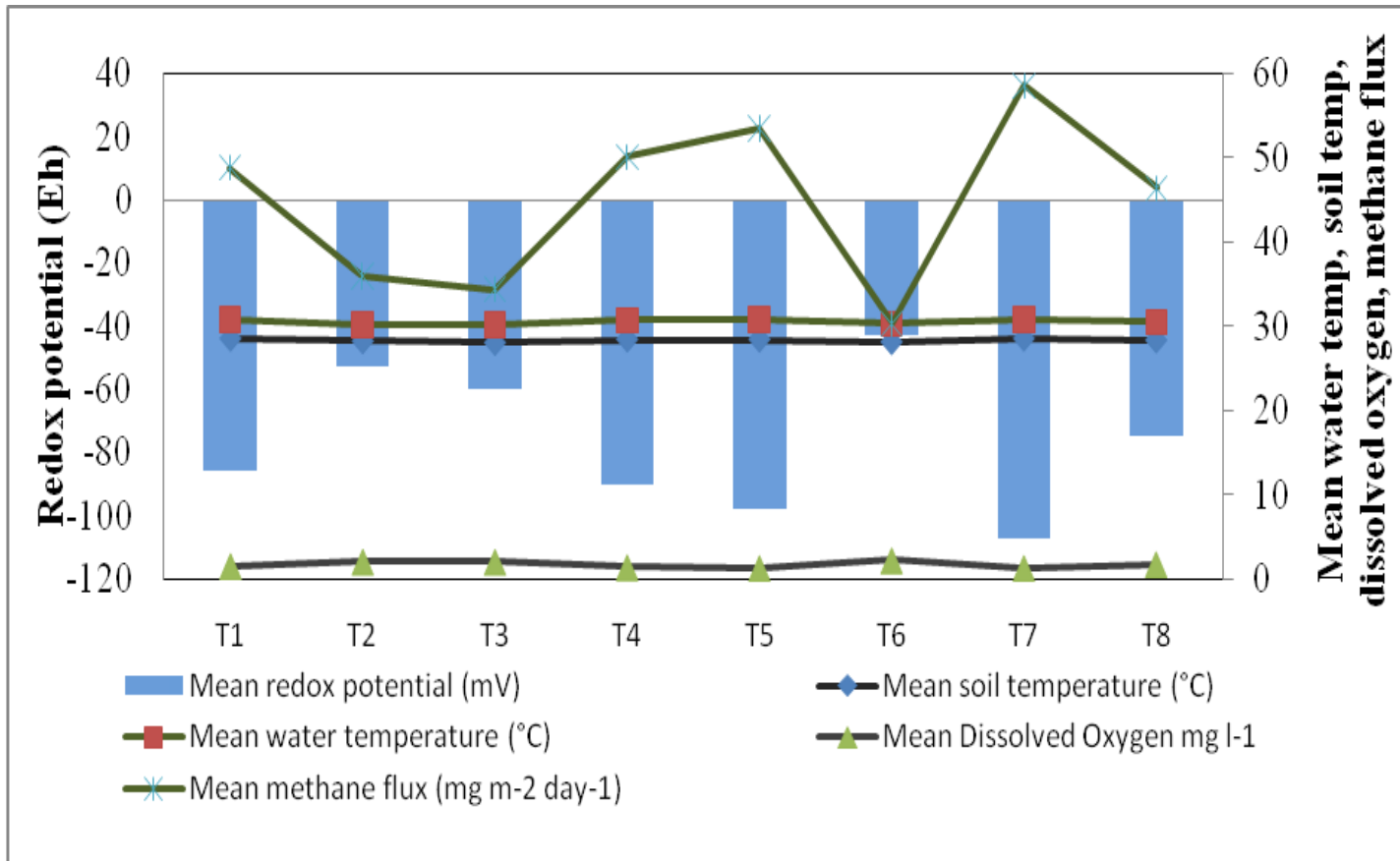
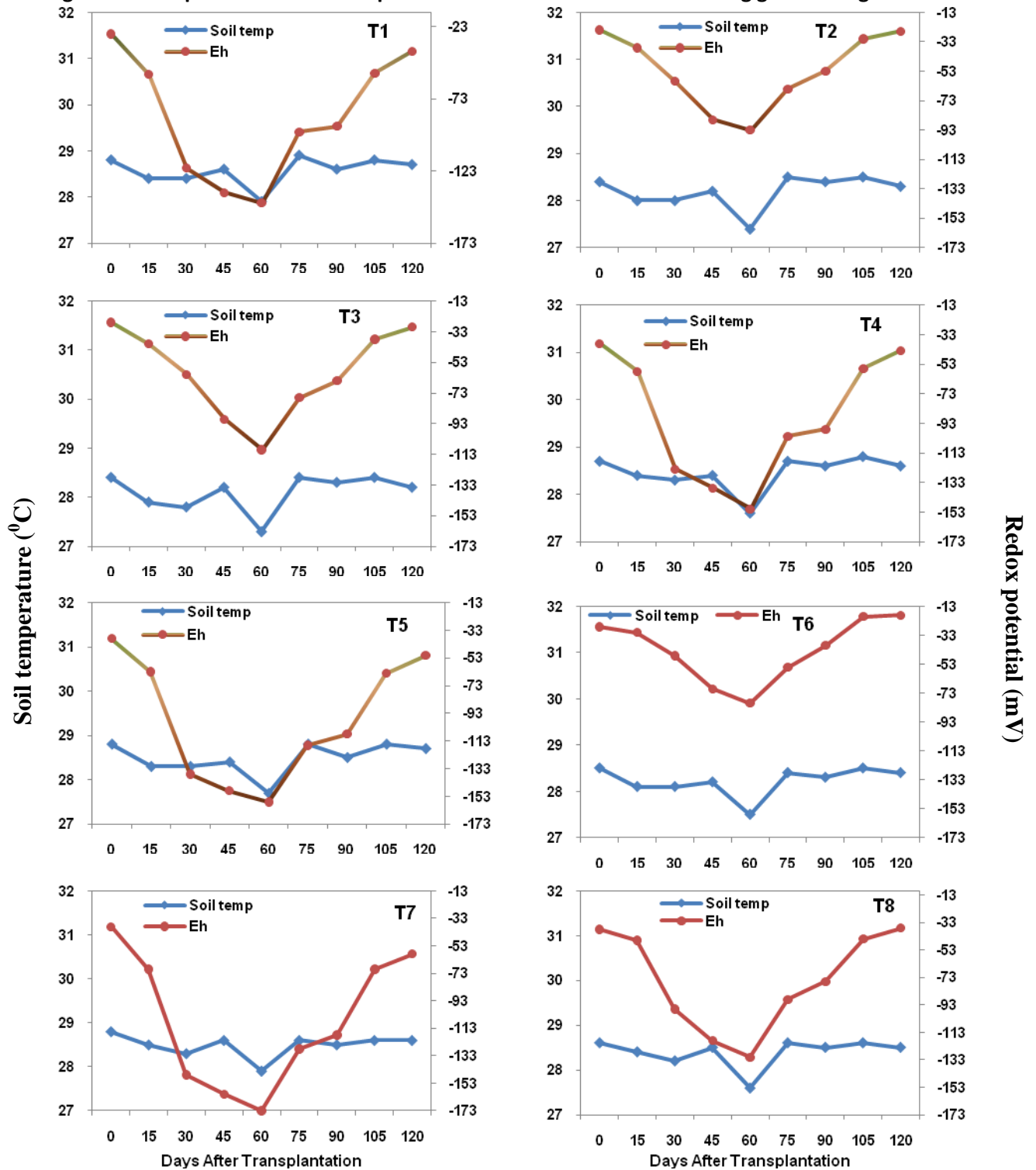


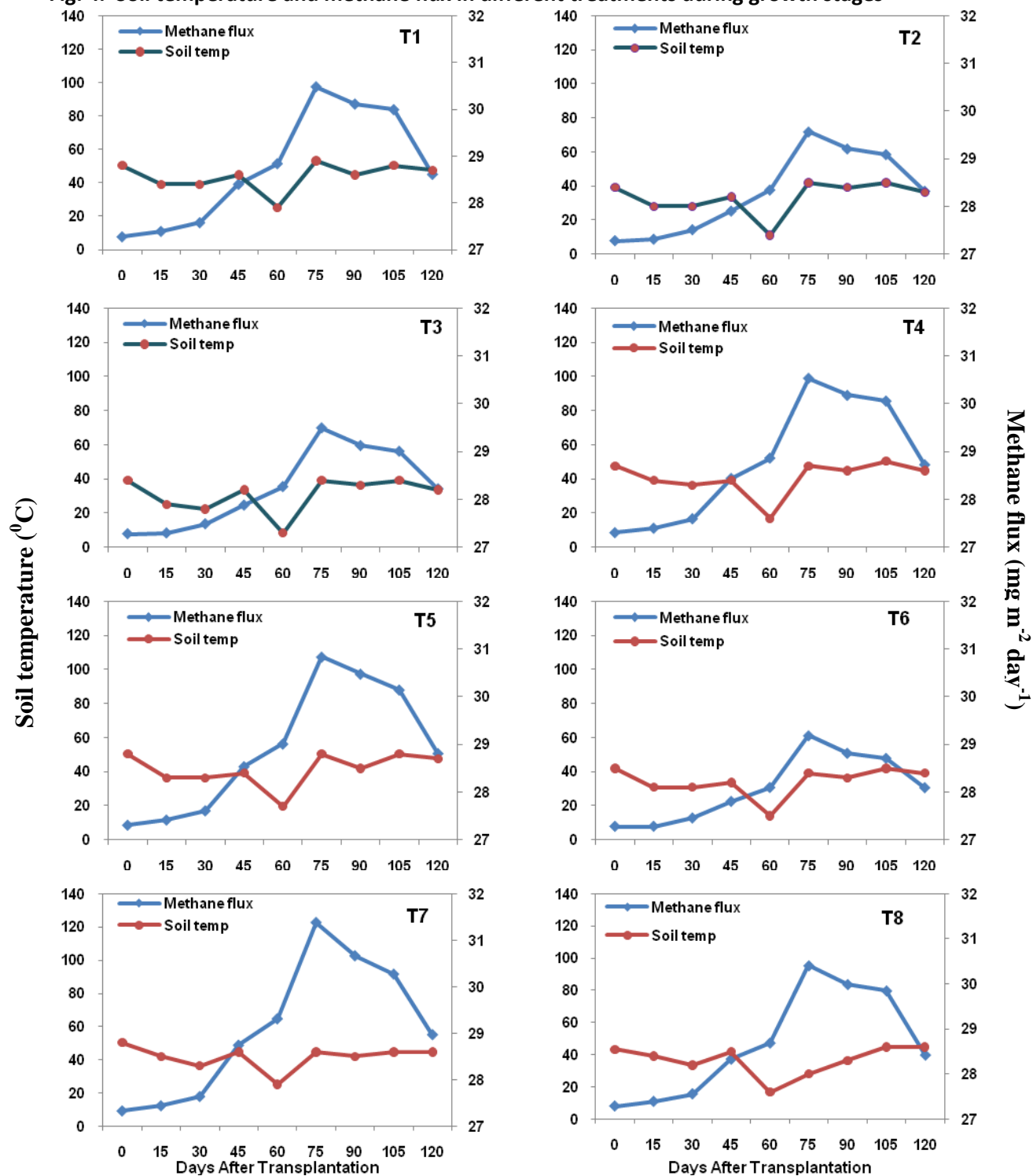
Fig. 3. Soil temperature and Redox potential in different treatments during growth stages



T<sub>1</sub> - Control  
 T<sub>3</sub> - Azolla  
 T<sub>5</sub> - Green Leaf Manure (GLM)  
 T<sub>7</sub> - FYM + GLM

T<sub>2</sub> - Blue Green Algae (BGA)  
 T<sub>4</sub> - Farm Yard Manure (FYM)  
 T<sub>6</sub> - BGA + Azolla  
 T<sub>8</sub> - BGA + Azolla + FYM + GLM

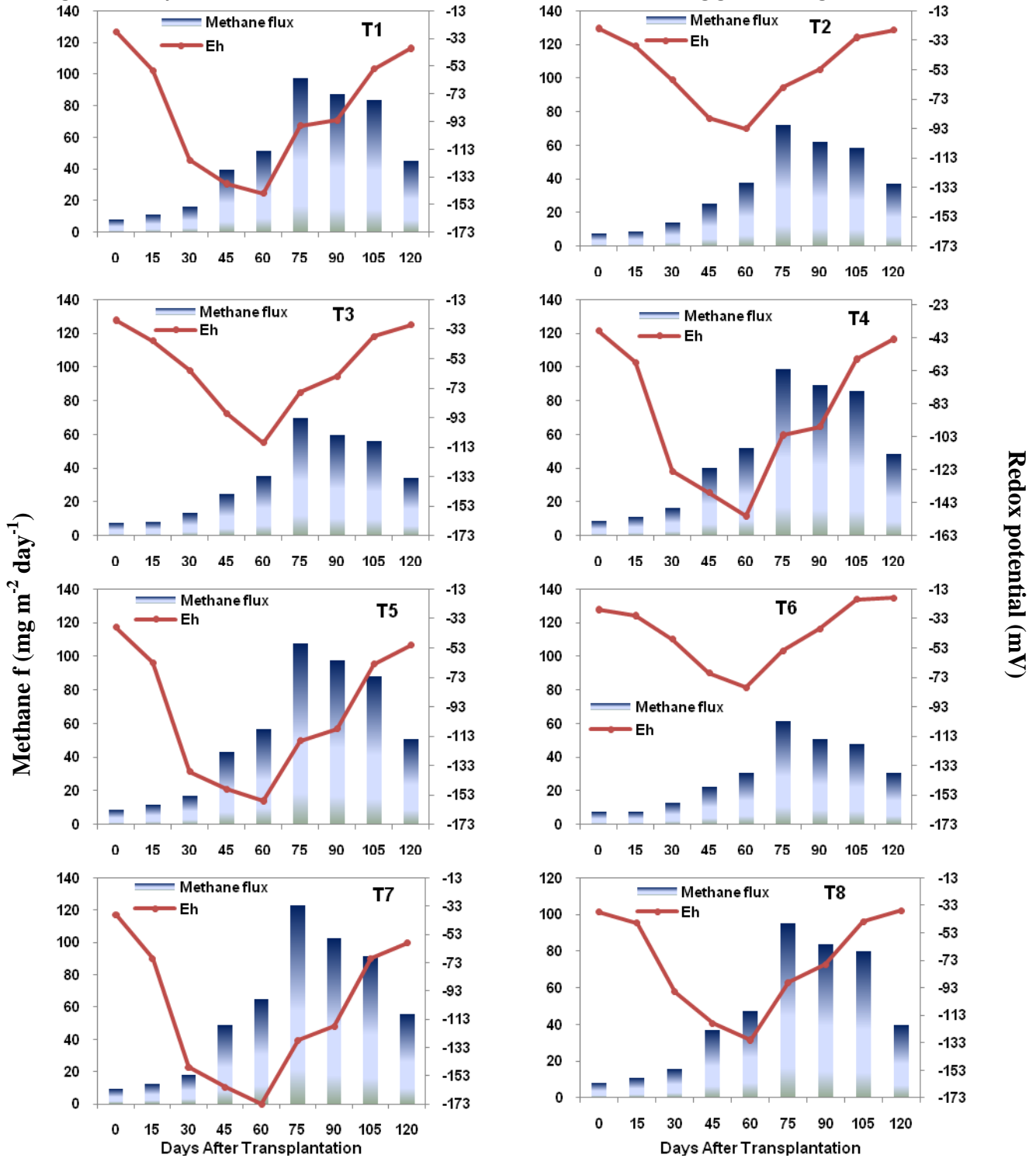
Fig. 4. Soil temperature and methane flux in different treatments during growth stages



T<sub>1</sub> - Control  
 T<sub>3</sub> - Azolla  
 T<sub>5</sub> - Green Leaf Manure (GLM)  
 T<sub>7</sub> - FYM + GLM

T<sub>2</sub> - Blue Green Algae (BGA)  
 T<sub>4</sub> - Farm Yard Manure (FYM)  
 T<sub>6</sub> - BGA + Azolla  
 T<sub>8</sub> - BGA + Azolla + FYM + GLM

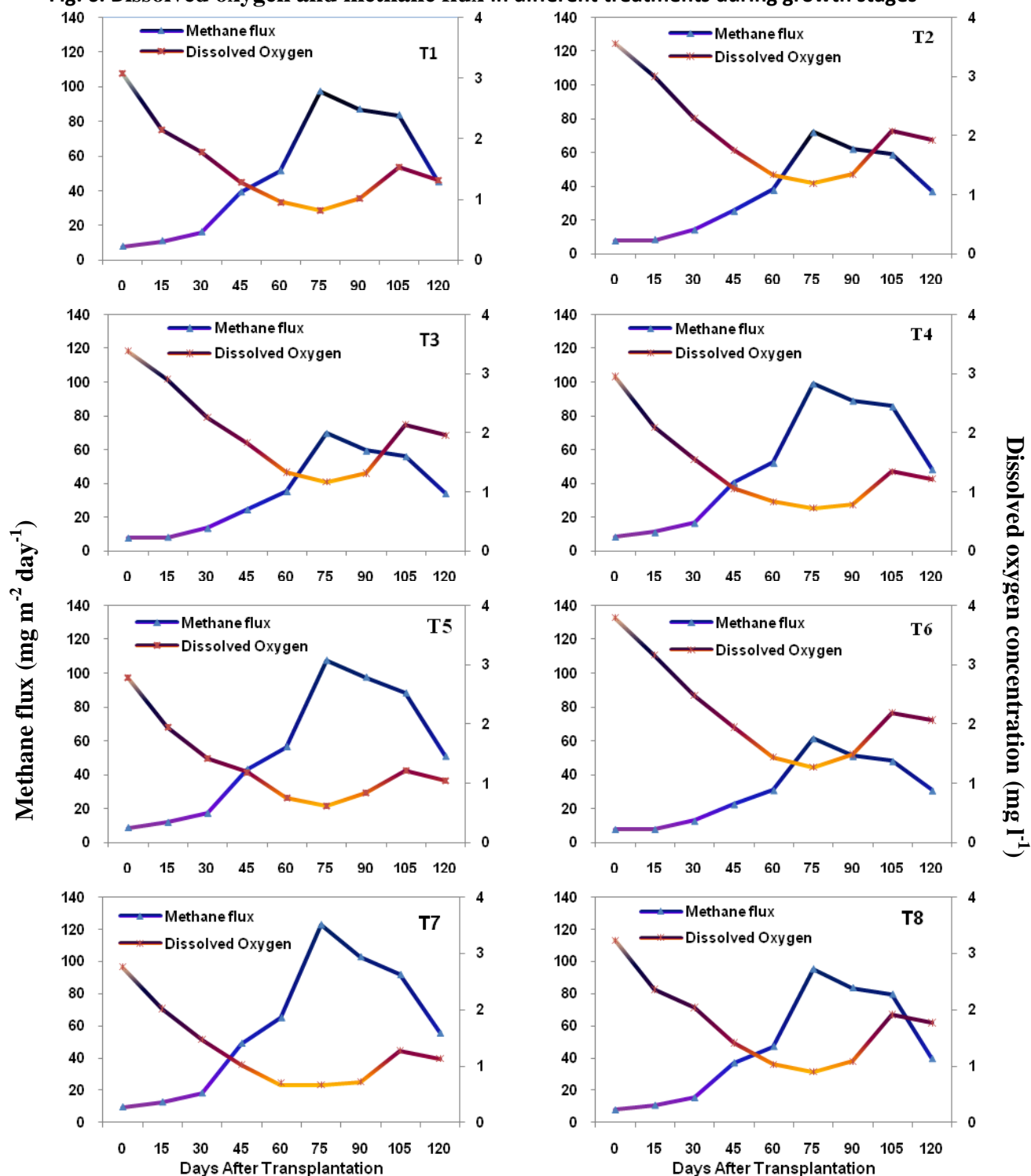
Fig.5. Redox potential and methane flux in different treatments during growth stages



T<sub>1</sub> - Control  
 T<sub>3</sub> - Azolla  
 T<sub>5</sub> - Green Leaf Manure (GLM)  
 T<sub>7</sub> - FYM + GLM

T<sub>2</sub> - Blue Green Algae (BGA)  
 T<sub>4</sub> - Farm Yard Manure (FYM)  
 T<sub>6</sub> - BGA + Azolla  
 T<sub>8</sub> - BGA + Azolla + FYM + GLM

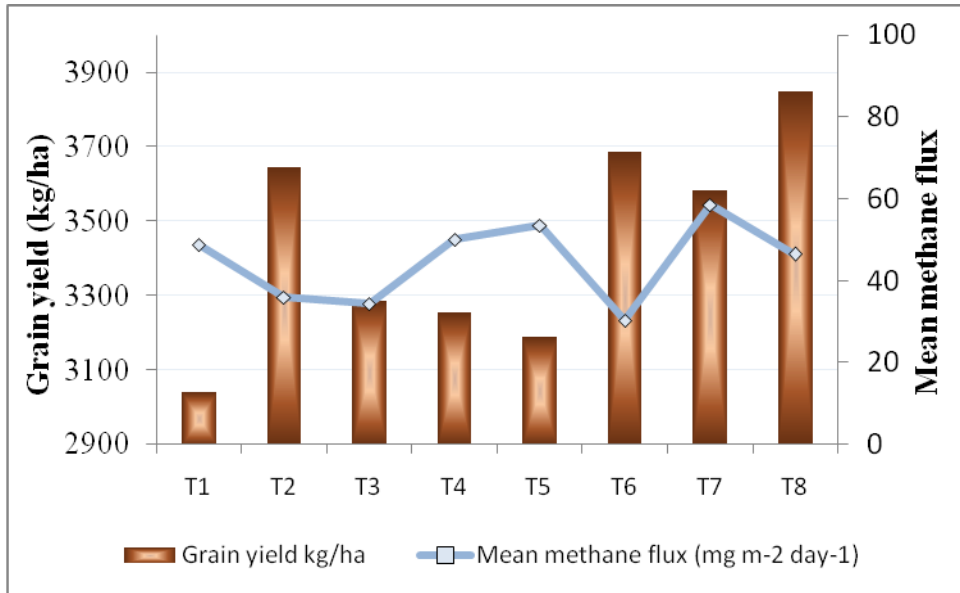
Fig. 6. Dissolved oxygen and methane flux in different treatments during growth stages



T<sub>1</sub> - Control  
 T<sub>3</sub> - Azolla  
 T<sub>5</sub> - Green Leaf Manure (GLM)  
 T<sub>7</sub> - FYM + GLM

T<sub>2</sub> - Blue Green Algae (BGA)  
 T<sub>4</sub> - Farm Yard Manure (FYM)  
 T<sub>6</sub> - BGA + Azolla  
 T<sub>8</sub> - BGA + Azolla + FYM + GLM

**Fig.7. Mean methane flux and crop yield**



- |                |   |                         |                |   |                          |
|----------------|---|-------------------------|----------------|---|--------------------------|
| T <sub>1</sub> | - | Control                 | T <sub>2</sub> | - | Blue Green Algae (BGA)   |
| T <sub>3</sub> | - | Azolla                  | T <sub>4</sub> | - | Farm Yard Manure (FYM)   |
| T <sub>5</sub> | - | Green Leaf Manure (GLM) | T <sub>6</sub> | - | BGA + Azolla             |
| T <sub>7</sub> | - | FYM + GLM               | T <sub>8</sub> | - | BGA + Azolla + FYM + GLM |

**Plate 1. Experimental plots applied with Green Leaf Manure (T<sub>5</sub>) and Farm Yard Manure (T<sub>4</sub>)**



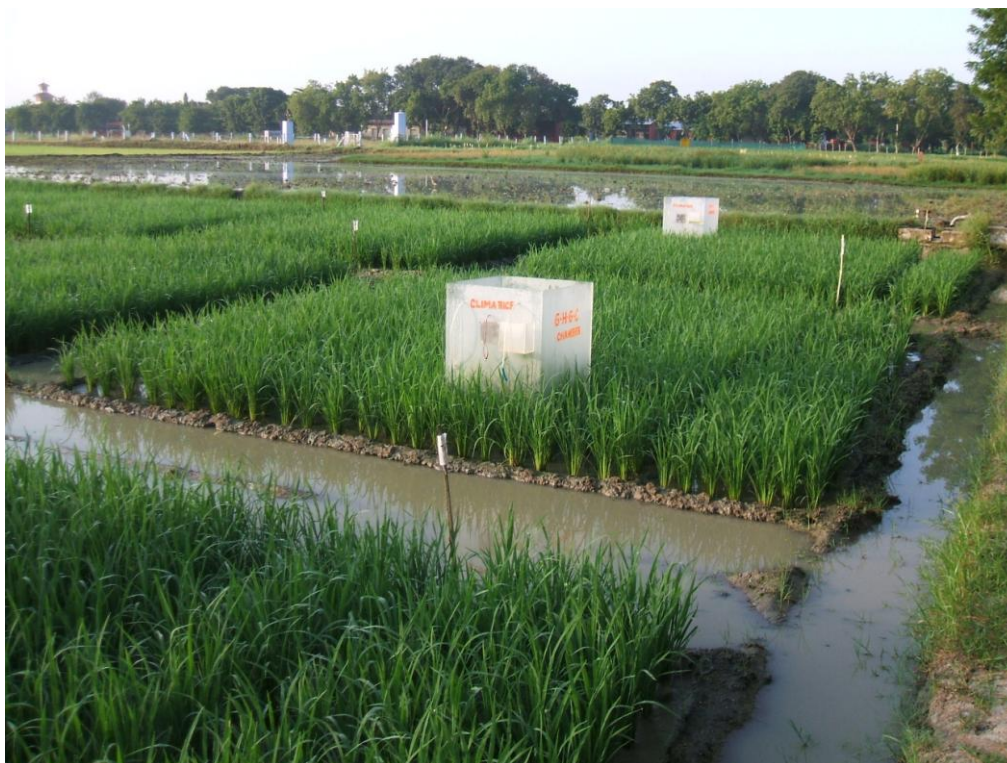
**Plate 2. The experimental field while transplanting**



**Plate 3. Over view of experimental field**



**Plate 4. Experimental field with methane collection chamber**



**Plate 5. Colonization of Blue Green Algae in experimental plot (T<sub>2</sub>)**



**Plate 6. Azolla as dual crop in the experiment plot (T<sub>3</sub>)**



**Plate 7. Wire netting to retain Azolla in the plot**



**Plate 8. Collection of methane using methane collection chamber in the field**



**Plate 9. Measurement of soil and water temperature from the experimental plots**



**Plate 10. Automatic weather station for recording weather parameters**



**Plate 11. Field board with experimental details**



**Plate 12. Methane analysis using gas chromatography**

