B-51. 2. 5 Studies on the Transportation of Methane and Nitrous Oxide through Rice Plants

Contact Person: Kazuyuki Yagi
Environmental Resources Division
Japan International Research Center for Agricultural Sciences
Ohwashi 1-2, Tsukuba, Ibaraki
305-8686 Japan
Tel,0298-38-6353, Fax,0298-38-6651
E-mail: kyagi@jircas.affrc.go.jp

Total Budget for FY 1998: 2,000,000 Yen

Abstract

Eleven varieties of rice plants were water-cultivated at greenhouse condition for testing the transportation potentials of methane and nitrous oxide. The growth of rice was followed during the whole vegetation season. The transportation rate was found to be controlled both by methane concentration in the solution and by plant conductance. Measurements on conductance of rice plants indicated that, for methane, the conductance ranged from 0.3±0.2 (LM) to 1.21±0.7 (YB) μmol/min/mM/plant at the tillering stage and varied between 0.6±0.3 (N36) and 2.5±0.7 (LM) μmol/min/mM/plant at the reproduction stage. For nitrous oxide, the conductance varied from 0.029 μmol/min/mM/plant (LM) to 0.079 μmol/min/mM/plant (N3).

Plant conductance was significantly correlated with the plant physical size. Multiple linear regression indicated that the conducting resistance of the plant mainly comes from the roots. In most case, the maturation status of the root greatly affects the transportation potentials of the plant. Results also showed the conductance of methane was, in general, higher than that of nitrous oxide.

Key Words: Conductance, Cultivar, Global warming, Greenhouse gas, Gas transport

Introduction

Rice paddy methane is one of the major sources which constructed atmospheric methane budget (IPCC, 1994). Methane produced in the rice paddy soils was transported into atmosphere by rice plants (Cicerone and Shetter, 1981). Plant mediated transport, molecular diffusion at the water-air interface, and gas bubble ebullition are the three major pathways for the transportation of the methane from paddy soils to the atmosphere (Neue et al., 1994). Among the three major conduits, the flow of methane through the plant aerenchyma seems to be the most important (Cicerone and Shetter, 1981; Holzapfel-Pschorn and Seiler,1986; Nouchi et al., 1990; Yagi et. al., 1996, Neue et. al., 1997), while under normal circumstances, the ebullition of high methane concentration bubbles and diffusion through water gas interface accounted about only 20% of total emitted methane during the entire rice cultivation period (Neue et. al.,1994). Some studies even claimed that methane emission from rice paddies into the atmosphere through rice plant may account as high as 90% of the total methane flux (Holzapfel-Pschorn and Seiler,1986; Butterbachbahl et. al., 1997).

Previous studies quantitatively described the methane transport from the rhizosphere to the atmosphere through rice plant by using some modified diffusion models (Nouchi et. al., 1994). However, these efforts were greatly hampered by the uncertainties concerning rice plant conductance. Especially there is lack of knowledge about how much the conductance would deviate from current level depending on the maturation status and variety difference (Watanabe et. al., 1995). Further more, the knowledge about the quantified influence on the conductance from the factors concerning the physical and physiological parameters of the plants are rare (Nouchi et. al., 1994).

The widely accepted theories on methane transportation through the rice plant claimed that the methane produced from anaerobic processes was first dissolved in the soil water. In some cases the gas bubbles containing methane and CO_2 may directly contact with some part of the root surface and even the stem base of the rice plants. Such a gas-root contact would make the methane completely gasified in the root cortex. Gasified methane was then diffused into the shoots by the plant aerenchyma and forming an upward concentration gradient. Methane is finally released through the both micropores located in leaf sheath and gaps of junction between nodal plate and the leaf sheath (Nouchi et. al., 1990). The driving force for methane transportation is the methane concentration difference between atmosphere and soil pore water, while influence of other factors like transpiration and plant physiological activities was temporarily excluded (Whiting and Chanton, 1996). Some environmental parameters like temperature of the root zone and pressure of the gas bubbles have already been tested by some specific studies (Hosono and Nouchi, 1997a; Hosono and Nouchi, 1997b).

If we take the conception that methane transportation is governed by molecular diffusion (Armstrong, 1990, Beckett et al, 1987, Deniear van der Gon and Breemen, 1993) and possibly the thermo-osmosis (Dacey, 1980, 1987, Schröder et al, 1986), efforts for knowing the conductance variation should also be focused on the influence of maturation status and rice variety. Because usually, these two factors on a great extend determined the plant physiological properties. Some studies have recently focused the relationship between methane emission rate and biomass of the rice plant and even rice variety (Watanabe et al., 1995; Huang et al., 1997; Sigren et al., 1997). To cope with the uncertainties remained, we therefore conducted this study to deal with the relationship between plant conductance potential and its physical and physiological properties.

Method and Measurement

1. Rice plant

Eleven varieties of rice cultivar were used for the experiment (Table 1). Nine varieties were obtained locally in Japan. One was from IRRI (International Rice Research Institute, Philippines), another one from the United States. Some varieties were cultivated in past, and some varieties are even currently used for agriculture.

Table 1. Rice seeds used for the experiment and conductance

No	Full name	Origin	Classified No.	Abv.	Conductance, CH_4, umol/min/mM/Plant		Conductance, N_2O umol/min/mM/Plant	
					Till.	Reprod.	Till.	Reprod.
1	ASAHI1	Japan	040412	A1	0.57±0.20	1.15±0.34		0.058
2	GINBOUZU	Japan	040671	GB	0.72±0.26	1.30±0.95	0.068±0.008	
3	IR36	IRRI	140095	IR36	0.30±0.09	1.65±0.19		
4	LEMONT	USA	00059772	LM	0.30±0.23	2.55±0.28	0.029±0.008	
5	NOURIN25	Japan	040640	N25	0.45±0.05	1.15±0.36		
6	NOURIN3	Japan	040638	N3	0.76±0.18	0.65±0.06	0.079	0.058
7	NOURIN36	Japan	040641	N36	0.74±0.33	0.62±0.40	0.065	
8	NOURIN37	Japan	920936	N37	0.74±0.48	0.66±0.03	0.060	
9	SHINRIKI	Japan	000118	SR	0.52±0.09	0.92±0.38	0.030±0.004	
10	TAMANISHIKI	Japan	040566	TN	0.31±0.16	1.88±0.92		
11	YUBAE	Japan	920989	YB	1.21±0.66	0.76±0.14		

Classified No. was the ID. number of Gene Bank, National Institute of Agro-Biological Resources, MAFF, Japan. Abbreviations were used for the discussion of this paper. Till. stands for tillering stage, Reprod. stands for reproduction stage.
Conductance showed is the average value ± SD from three independent measurements.
Data without SD come from single measurement.

Rice plants were water-cultured in a greenhouse chamber, assembled by Motoyama Ltd., Japan, at temperature of 30°C/20°C (day/night). Relative humidity in the chamber was around 60-75% by the free water vaporization from culture solution. Sunlight intensity is identical to the daily sunlight outside chamber of the season. Rice seeds were germinated on a Saran net floating in a plastic box which was filled with distilled water. At the fourth leaf emergence (25-30 days after sowing), seedlings were transplanted to container (28cm in wide, 35cm in length, 30cm in depth). Culture solution was composed according to Baba (1965) with a little modification based on the maturation status of the rice plants and the experience of previous studies (Nouchi et al, 1990). In order to make young seedling adept to the culture solution, during the first week, culture solution was ten times diluted. During the following vegetation period, the culture solution was replaced every week to maintain a stable chemical composition of the culture solution. For rice growing, the lower part of the stem was wrapped with a urethane sponge belt and inserted into the holes on Styrofoam plate. The plate was placed over a box (22L in size) filled with culture solution.

2. Measurement of the transportation

The system for measuring the methane and nitrous oxide emission rate from the top of the rice plant whose roots was soaked in a culture solution containing high concentrations of methane and nitrous oxide is shown in Fig. 1. The static chamber was a self-made 10.8 L PVC cylinder, which was 15 cm in diameter and 60 cm in height. A small outlet of 7 cm in diameter at bottom was used for introducing rice plant. The leaves and stems of the plant were closed by the PVC chamber and properly sealed from surrounding air by a rubber stopper and module clay. A small fan was mounted at the top of the chamber to make the air inside the chamber well mixed. The whole system was placed in an artificially, environmentally controlled growth chamber (Motoyama, Japan), which maintained an irradiation of 300 μmol/m^2/s (PPFD, photosynthetic Photon Flux Density), and a constant temperature of 25°C.

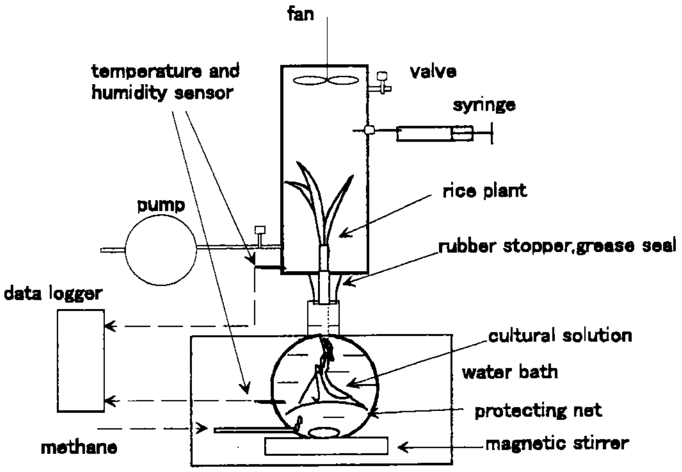

Figure 1. Device for testing methane transportation. The methane flux was measured in a closed chamber (PVC, 10.8L). The rice plant was held at lower part of the stem by a rubber stopper and sealed with model clay from surrounding atmosphere.

Before each set of measurement, purified air was drawn by an air pump (AP-115RN, Rei-Sea Co. Ltd) into the chamber to make the methane and CO_2 concentration inside the chamber return to normal level (for methane, less than 8ppm; for CO_2 less than 500ppm). The purified air was generated by a membrane filter and a serially connected cooling dryer (DH-105, Komatsu Electronics Inc.). Methane transportation rate was calculated by using the concentration increase inside the PVC chamber over time (Fig. 1). The temperature of culture solution flask was maintained at 25°C by a water bath (CTE42A, Yamato Komatsu). The culture solution was well mixed during the experiment by running a magnetic stirrer at the bottom of the flask. The methane concentration in the culture solution was previously found stable during 1-2 hour measurement (Hosono and Nouchi, 1997a).

3. Measurement of the physiological properties of rice plant.

The monitored physiological parameters of the rice plant include: height of the plant, maximum root length, numbers of node-leaf sheath junction (methane releasing site), numbers of stem, total root length, volume of roots, root pore space volume, stem volume, stem pore space volume, leaf area, stem fresh weight, leaf fresh weight, root fresh weight, stem dry weight, root dry weight, and leaf dry weight. The node-leaf sheath junction was believed to be the major sites for methane releasing at upper part of the rice plant (Nouchi et al., 1990).

4. Measurement of methane, nitrous oxide and carbon dioxide

Quantitatively, methane and CO_2 was determined by a gas chromatograph (GC-9AM, Shimadzu) installed with a Propack Q 80-100mesh packed column (1m, 0.5mm id.). A dehumidifier was used as a pre-column. Detectors were a TCD for testing CO_2 and a FID for testing methane connected serially. The amount of methane dissolved in the cultural solution was tested by direct injecting of 1 μL cultural solution into the GC. Nitrous oxide was tested by ECD using a Propack Q 80-100 mesh packed column for separation (GC-14B, Shimadzu). Aquatic nitrous oxide was measured by summating head space and solution nitrous oxide, which was kept in a 50ml glass vial.

Results and discussion

1. Effect of rice variety on the conductance

Measurements on the rice conductance were carried out twice. One is around the tillering stage (35-45 day-old); another is in the reproduction stage (70-80 day-old). For methane, the conductance of three rice plants was measured for each stage and every variety. The minimum conductance was found from the plants of LM at the tillering stage, 0.3±0.2 μmol/min/mM/plant; while highest conductance also associated with LM at reproduction stage, 2.5±0.7μmol/min/mM/plant. During the tillering stage, the conductance was relatively lower than that in the reproduction stage, ranging from 0.3±0.2 μmol/min/mM/plant (LM) to 1.2±0.7 μmol/min/mM/plant (YB). While during the reproduction stage, conductance ranged between 0.6±0.3 μmol/min/mM/plant (N36) and 2.5±0.7 μmol/min/mM/plant (LM). Four Japanese varieties, N25, N3, N36 and N37 showed similar conductance. Among these varieties, N25, N36 and N37 have genetic relationship (Hokuriku Natl. Argric. Exp. Stn., 1979). For nitrous oxide, the measurements, which were carried out both at the tillering stage and the reproduction stage, indicated that the conductance varied from 0.029 μmol/min/mM/plant (LM) to 0.079 μmol/min/mM/plant (N3).

2. Effect of maturation on the conductance

The first round of measurements was carried out when most of rice plants are still young. Due to the effect of variety, plants of some varieties like YB and N3 maturated a little faster than others. The maturation difference also existed during the second round of measurement. Some short vegetation varieties like, GB and A1 even produced a few young grains while most of other plants were still coining. Maturation status of the plants used for conductance measurement is shown in Fig. 2. Corresponding methane conductance of the plants from different varieties was listed in Table 1. For methane, in general trend, the conductance increased with maturation of the rice plant. Most of the tested varieties showed a significant difference in conductance between the tillering stage and the reproduction stage. From the tillering stage to reproduction stage, the plants of AI, GB, IR36, LM and TN, grew rapidly, while conductance also increased accordingly. Slow growing varieties, like N3, N36, N37 and SR, did not exhibit a significant increase in conductance. For nitrous oxide, such trend was not strong.

3. Influence of plant physical parameters on the conductance.

Compared with all the results obtained, the most significant feature which we found during the experiment, is that plant conductance methane and nitrous oxide, in a general trend, correlated with the size of the plant, that is, the bigger the size of the rice plants the larger the conductance. These results coincide with the results of some studies which related methane emission with variety and yields (Huang et al., 1997).

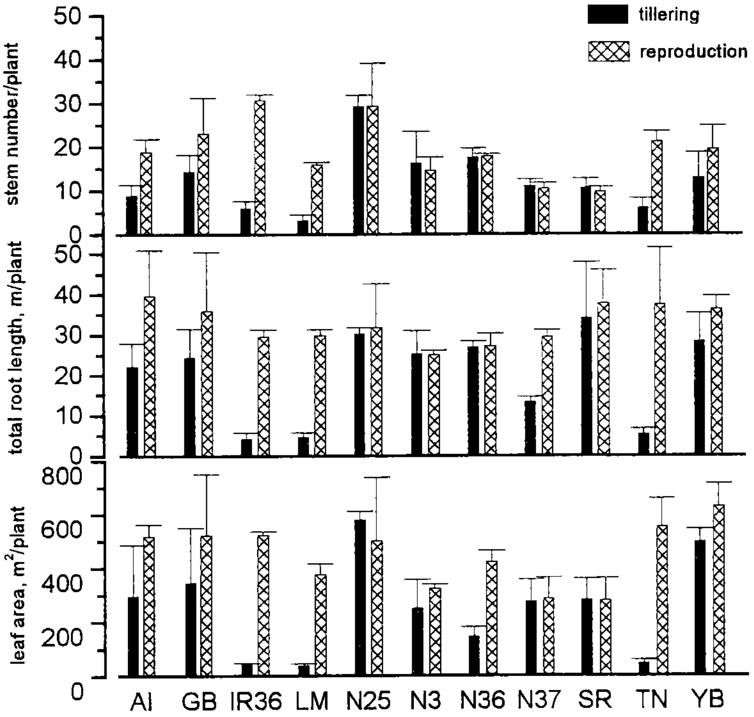

Figure 2. Maturation status at of the rice plant at tillering stage and reproduction stage. All average results come from three independent measurements. Error bar indicated the standard deviation.

Strong relationships between the conductance and most of the parameters concerning physical size were found by simple linear correlation computation. These parameters include: root volume, root fresh weight, root overall-perimeter, root maximum length, stem pore space, root dry weight, stem volume, leaf fresh weight, stem fresh weight, leaf dry weight, leaf area, above-biomass fresh weight, stem number, numbers of releasing site, above-biomass dry weight and stem dry weight (Fig. 3). However, plotting the conductance against all the measured parameters indicated that there is no such one, which can be much more prominent than rest of other factors which may determine the conductance under all circumstances. It is seemingly, from our observations, that the high conductance usually associated with the plants whose roots are white, burlier, and highly maturated. According to the data obtained both in the tillering stage and the reproduction stage, we found that the conductance is more likely to relate with root properties, that is, root total volume, root fresh weight, root overall perimeter, and root maximum length. Figure 3 indicated that the plots of conductance against some plant physiological parameter.

4. Correlation between methane and nitrous oxide conductance.

Cultural solution containing methane and nitrous oxide provided a contacting surface for the diffusion of the two gases. These two gases would first gasify on the root cortex. Gasified gases were then diffused into the shoots by the plant aerenchyma and simultaneously forming an upward concentration gradient. Such a concentration gradient would drive the methane or nitrous go through the micro-pores located on the leaf sheath and the gaps of junction between nodal plate and the leaf sheath (Nouchi et. al., 1990). Within this transportation scheme, if both methane and nitrous oxide follow the similar transportation mechanisms, the conductance should be to some extent correlated. Our observations confirmed these arguments. The methane conductance correlated with nitrous oxide conductance, which was tested simultaneously. The correlation coefficient reached 0.7336 (Fig. 4). Scattering of the data points suggested that mechanisms for methane and nitrous oxide may not completely the same.

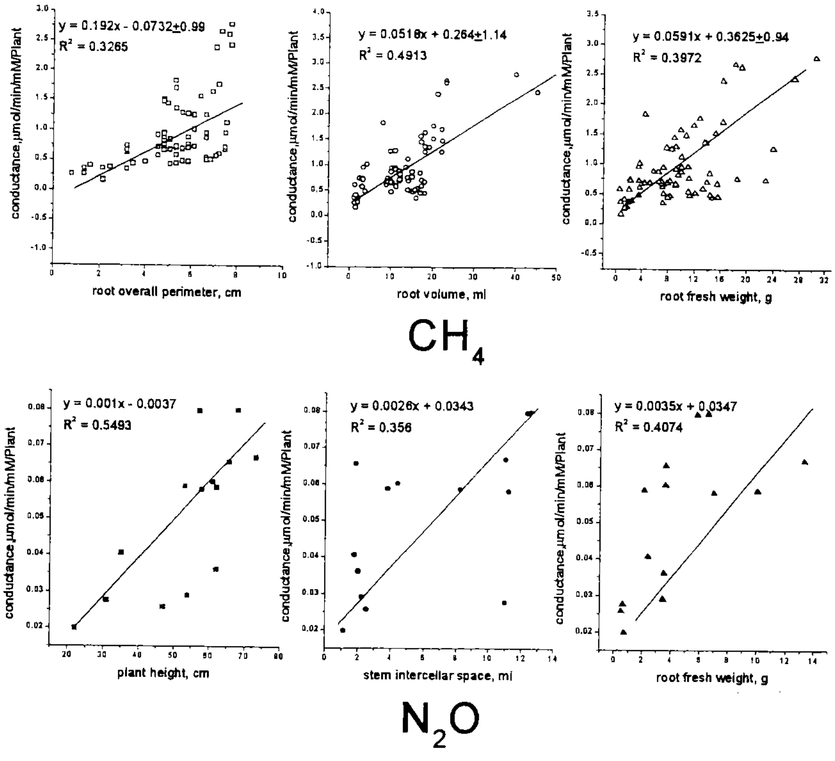

Figure 3. Correlation of methane and nitrous oxide conductance with some plant physical parameters.

Quantitatively, the conductance of methane and nitrous oxide were not in same magnitude. Methane conductance was, 9.3-19.1 (16.1 in average) times higher than that of nitrous oxide (Table 1, Fig. 4). The detailed reason for such discrepancy is difficult to be elucidated in this study. We speculated that following factors might contribute to the difference: 1) the molecular weight of nitrous oxide is 44, while that of methane is 16. At room temperature, such difference would make thermal motion of the nitrous oxide about 2 times less than that of methane and consequently cause a big difference in diffusion rate (Fu, 1979); 2) the molecular radius of nitrous oxide is 1.48Å, while that of methane is 1.09Å. Small molecular may easily go through the plant tissue and micro-pores located at the root surface. 3) The Henry constant for methane in the cultural solution is higher than that of nitrous oxide. According to our measurements, the Henry constant for methane is 1765 atm/M, while that for nitrous oxide is 12.6 atm/M. Larger Henry constant would cause a faster diffusion of dissolved gas. 4) Saturated solubility of nitrous oxide (15.6 mM, this study) is higher than that of methane (1.31 mM, this study). Higher solubility tends to trap more gas and therefore cause a slow transportation. Our results do not response the conclusions obtained by Yu et al (1997). In their study, they found that nitrous oxide was transport in a similar way as that of methane. It is very difficult to estimate how much the difference between the laboratory results and field experiment data. However, the conclusions from ours indicated that the plant conductance of nitrous oxide is much small than that of methane. In addition, since solubility of nitrous oxide is much higher than that of methane, we believe, in field condition, transportation of nitrous oxide from paddy soils to atmosphere is not mainly through rice plant. Diffusion of nitrous oxide from water surface and bubbling may take a large percent of the transportation.

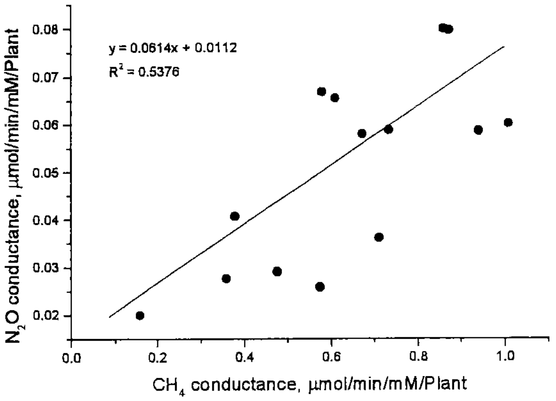

Figure 4. Correlation between methane and nitrous oxide conductance

Conclusion

Methane dissolved in the soil water of paddy fields is transported into the atmosphere through plant aerenchyma. The transportation rate was controlled both by methane concentration in the solution and by plant conductance. Measurements on conductance of rice plants from 11 varieties indicated that methane conductance ranged from 0.3±0.2 (LM) to 1.21±0.7 (YB) μmol/min/mM/plant at the tillering stage and varied between 0.6±0.3 (N36) and 2.5±0.7 (LM) μmol/min/mM/plant at reproduction stage. For nitrous oxide, the conductance varied from 0.029 μmol/min/mM/plant (LM) to 0.079 μmol/min/mM/plant (N3).

Plant conductance was significantly correlated with the plant physical size. This is because the aerenchyma system of a plant is roughly proportional to its physical size. For each specific species, since the gas transport from the culture solution to the atmosphere involved with several steps, there is not such a factor, which can act as the dominant role, which control the methane conductance of the plant. The conducting resistance of the plant mainly comes from the roots. In most case, the maturation status of the root greatly affects the transportation potentials of the plant. Due to the differences of physical and chemical properties, for rice plant, methane conductance is higher than nitrous oxide conductance.

Reference

Amstrong,W., 1971, Radial oxygen losses from intact rice toots as affected by distance from the apex respiration and waterlogging, Physiol.Plant, 25, 192-197.

Baba, I., Y.Takashsi, 1956, Water and sand culture methods, in Crop Science eds Togri Y., Associ. of Agri. Tech.Tokyo, 157.

Beckett, P.M., W.Armstrong W., S.H.F.W.Justin, J. Armstrong, 1988, On the relative importance of connective and diffusive gas flows in plant aeration, New Phytol., 110, 463-468.

Butterbachbal,K., H. Papen, H.Rennenberg, 1997, Impact of gas transport through rice cultivars on mehtane emission from rice paddy fields, Plant Cell & Environment, 20, 1175-1183.

Cicerone, R.J., Shetter, J.D., C.C.Delwiche, 1983, Seasonal variations of mehtane flux from a california rice paddy, J.Geophys. Res., 88,11022-11024.

Dacey,J.W.H., 1980, Internal winds in water lilies and adaptation for life in anaerobic sediments., Science, 210, 1017-1019.

Denier van der Gon, H.A.C. and N.Van Breemen, 1993, Diffusion-controlled transport of methane from soil to atmosphere as mediated by rice plants, Biogeochemistry, 21, 177-190.

Hokuriku Nati. Agri. Exp. Stn., 1979, List of breed rice varieties, systematic sources and variety name (Intelligent Document No. 25), Hokuriku Nati. Agri. Exp. Stn. ed. Agro-fishery Technology and Information Association Press, Japan.

Holzapfel-Pschorn, A, W.Seiler, 1986, Methane emission during a cultivation period from an Italian rice paddy, J.Geophys. Res., 91,11803-11814.

Hosono,T., I.Nouchi, 1997a, The dependence of methane transport in rice plants on the root zone temperature, Plant and Soil, 191,233-240.

Hosono T. and I. Nouchi, 1997b, Effect of gas pressure in the root and stem base on methane transport through rice bodies, Plant and soil, 195,65-73.

Huang,Y., R.L.Sass,F.M.Fisher, 1997, Methane emission from Texas rice paddy soils--quantitative multi-year dependence of CH4 emission on soil, cultivar and grain yield, Global Change Biology, 3, 479-489.

International Panel on Climatic Change, 1994, Radiative Forceing Rate of Cimate Change and an Estimation of the IPCC IS92 Emission Scenarios, in Climate Change, J.T.Houghton, L.G.Mera Filho, J.Bruce, H.Lee, B.A.Callander, E.F.Hailes, N.Marris and K. Maskell ed., Cambridge Univ Press, New York.

Liang, G.H., A.D.Dayton, C.C.Chu and A.J.Casady, 1975, Heritability of stmatal density and distribution on leaves of grain sorghum, Crop Science, 15, 567-570.

Neue,H.U., R.Wassmann, H.K.Kludze, W.Bujun, R.S.Lantin, 1997, Factors and processes controlling methane emissions from rice fields, Nutrient Cycling in Agroecosystems, 49, 111-117.

Neue,H.U., R.Wassmann, R.S.Lantin, M.C.A lberto, J.B.Aduna, 1994, Ebullition of methane, IRRI, 19-3,36.

Nouchi, I., S.Mariko, K.Aoki, 1990, Mechanism of methane transport from the rhizosphere to the atmosphere through rice plants, Plant Physiol., 94,59-66

Nouchi,I., T,Hosono, K.Aoki, K.Minami, 1994, Seasonal variation in methane flux from rice paddies associated with methane concentration in soil water, rice biomass and temperature, and its modeling, Plant and Soil, 161, 195-208.

Raskin I., H.Kende, 1983, How does deep water rice solve its aeration problem, Plant Physiol., 72, 447-454.

Raskin I., H.Kende, 1985, Mechanism of aeration in rice, Science, 228, 327-329.

Schröder,P., W.Grosse, D.Woermann, 1996, Localization of thermo-osmotically active partition in young leaves of Nuphar lutea, J. Exp. Bot, 37, 1450-1461.

Sigren LK, Byrd GT, Fisher FM & Sass RL, 1997, Comparison of soil acetate concentrations and methane production, transport, and emission in two rice cultivars. Global Biogeochem. Cycles, 11, 1-14.

Wang, Z.P., D.Zeng, W.H.Patrick, 1997, Characteristics of methane oxidation in a flooded rice soil profile, Nutrient Cycling in Agroecosystems, 49, 97-103.

Watanabe,A. M.Kaiwara, T.Tashiro, M.Kimura, 1995, Influence of rice cultivar on methane emission from paddy fields, Plant and Soil, 176,51-56.

Whiting,G.J., J.P.Chanton, 1996, Control of the diurnal pattern of methane emission from emergent aquatic macrophytes by gas transport mechanisms Aquatic Botany 52, 237-253.