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Review

Strategies to lower greenhouse gas level by rice agriculture

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The Earth's average temperature has risen to about 1°F in the past 100 years and is projected to rise another 3 to 10°F in the next 100 years. Human activity in last few decades has increased the concentration of various greenhouse gases, leading to increased CO_2, methane, tropospheric ozone, CFCs and nitrous oxide. To normalize the nature health many researchers around globe devote their life to searching a good way to reduce greenhouse gases, therefore global warming has been taken a full flag attention worldwide. In this minireview we introduced different agriculture strategies used so far to reduce greenhouse gases. The concept principally focuses on transgenic plants and integrated management system. The transgenic rice (basically a C3 plant) harboring C4 photosynthetic genes phosphoenolpyruvate carboxylase (PEP) carboxylase (PEPC) and pyruvate orthophosphate dikinase (PPDK) has been showed to increased photosynthetic capacity and efficiency of carbon dioxide assimilation. However, many drawbacks that is, field test, stability of the transgenic lines are unavoidable. Taiwan farmers utilized commercial hormone and fertilizer combination to rice growing. Very recently the integrated management system is set up based on plant physiological needs. It did increase the rice yield per growing unit area and grain bearing rate. The fixation of CO_2 to carbon was therefore expected to be greater, which indirectly reduced the amount of CO_2 in the field. On the other hand, the emission of methane may also be reduced with better CO_2 assimilation and the less carbon delivered to the ground. This strategy can both increase the rice yield and have benefits on global warming mitigation. In short-term, the integrated management system, an eco-farming approach, would be a better solution than transgenic plants.

Key words: Greenhouse gas, rice, C4 plant, PEP carboxylase, pyruvate orthophosphate dikinase.

INTRODUCTION

Global warming is defined as the increase in the average measured temperature of the Earth's near-surface air and oceans, and its projected continuation. The rising of Earth's temperature is govern by exotic components like water vapor, carbon dioxide (CO_2), ozone, methane (CH_4), nitrous oxide (N_2O), and chlorofluorocarbons (CFCs) that absorb heat and thus increase atmospheric temperatures, and results in disasters like hurricanes, droughts and floods which are becoming more frequent. The unexpected temperature rising is mainly because of the increase in atmospheric green-house gases due to anthropogenic activities: man cuts down the forest for timber or clear land for agricultural farming, industries or increasing human habitations. This extensive deforestation reduces the number of trees available to absorb carbon dioxide. The large scale me-thane gas produced in the atmosphere is from natural wetlands, rice paddies and livestock. Biomass burning, natural gas production, termite landfills and coal mining also release methane. Because of overexploitation of fossil fuel and other natural resources, it has become difficult to maintain the global energy balances between the atmosphere and the earth surface, and thus earth temperature is rising. Agriculture practices such as rice based cropping system is considered as the major greenhouse gas emitter (Neue and Minami, 1994; Banker et al., 1995; Wassmann and Aulakh, 2000), which contributes a major portion of all

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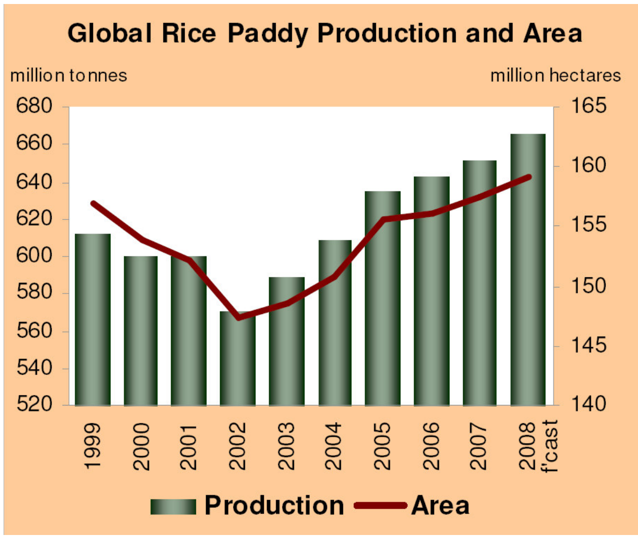

Figure 1. Global rice paddy production and area. Adapted from FAO Rice Market Monitor (2008).

global emissions. Rice absorbs sorbs carbon from the atmosphere, but if the plant cannot utilize it efficiently, the carbon is dispersed into the soil, where it converts to methane. Under these situations, global warming nowadays has become an important issue.

Rice is an important economic crop in the world. It serves as a main food source for more than 3 billion people around the world in 2004, especially in Asia. More than 90% of the world's harvested rice area is in Asia. According to the estimates from the latest Food and Agriculture Organization (FAO) of the United Nations, paddy production increased by 1.4% in 2007 to 650 million tones. The expansion of production would be on account of a 1.0 percent increase in the area harvested to 157.5 million hectares, and of a modest gain in average yields from 4.12 to 4.14 tones per hectare (Figure 1).

With rapid population growth, most Asian countries encounter domestic supply constraints. They have to import rice even to supplement their own rice production. The problems raised from the need of rice production have become an internationally hot issue. Uncertainties become even greater as rice cultivation itself has a significant effect on global warming through the emission of greenhouse gases, carbon dioxide and methane.

GREENHOUSE GASES

Carbon dioxide, methane, nitrous oxide, chloro-fluorocarbons comprise the major greenhouse gases (Figure 2). Among these greenhouse gases, both carbon dioxide and methane play major roles in greenhouse effect. These gases in combination with water vapour make a blanket like layer around the earth and prevent heat from escaping to keep the global average temperature within biologically active range. This effect is called greenhouse effect and the gases which can absorb the heat radiation from the earth are therefore called greenhouse gases. Greenhouse gases are increasing rapidly as a result of anthropogenic activities, particularly continued deforestation and the burning of fossil fuels to provide living space and energy for an expanding population.

CARBON DIOXIDE

Carbon dioxide is the most important anthropogenic greenhouse gas which leads to an increase in atmospheric temperature and continues to heat for decades to centuries. It represents 77% of total anthropogenic greenhouse gas emissions in 2004 (Figure 2). Although carbon dioxide is not a powerful greenhouse gas but it plays major role in greenhouse effect. Carbon dioxide is the sole source of carbon used by all plants during photosynthesis. Photosynthesis is a process by which plants use sunlight, water, and carbon dioxide to produce carbohydrates and other biological compounds, which reduces the amount of carbon dioxide in the air. This, in turn, helps reduce global warming. Thus plants behave as the "lungs of the Earth". The efficiency of carbon dioxide assimilation in rice, a C3 plant, would be diminished by photorespiration. If the efficiency of carbon dioxide assimilation is increased, the amount of fixed carbons in rice increases and hence lowers the concentration of carbon dioxide in the atmosphere.

METHANE

Methane in the Earth's atmosphere is an important greenhouse gas with an extreme global warming potential. Methane emission will have 25 times the impact on temperature of carbon dioxide emission of the same mass in the next 100 years (Climate Change, 2001). Methane is a radioatively active trace gas with approximate 21 times more infrared absorbing capacity per molecule than carbon dioxide (Prather et al., 1996). Methane plays a very important role as a potent greenhouse gas for three reasons: 1. It has large trend of increasing concentrations, 14% of total anthropogenic greenhouse gas emissions in 2004 (Figure 2); 2. It has strong absorbance and emission of infrared radiation; 3. The location of its absorption and emission bands are at wavelengths where carbon dioxide and water vapor are not absorbed (Dickinson and Cicerone, 1986).

The elevated atmospheric CO_2 concentrations in flooded soil leads to anaerobic decomposition (methanogenic activity) of organic matter that leads to the formation and emission of various trace gases which are generally not found in well-aerated upland soil (Butterbach-Bahl, 1992). Among these anaerobically produced trace gases, methane is very common (Neue and Minami, 1994). Flooded rice fields are a significant source

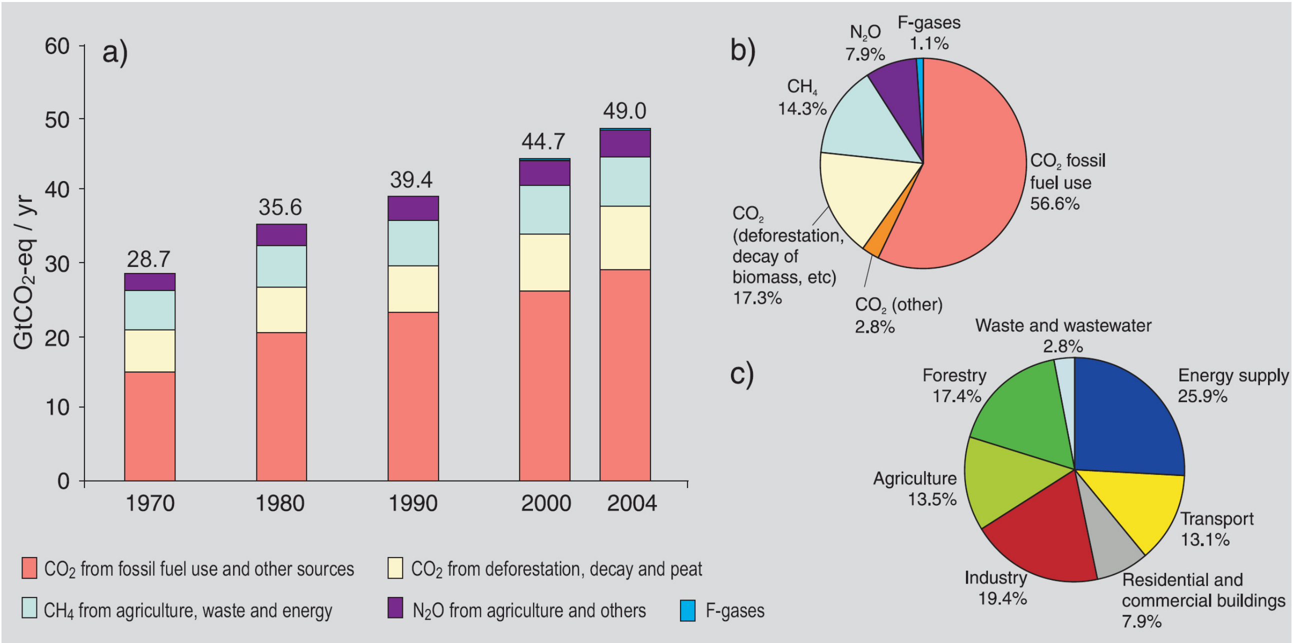

Figure 2. Global anthropogenic greenhouse gases emissions. (a) Global annual emissions of anthropogenic greenhouse gases from 1970 to 2004. (b) Share of different anthropogenic greenhouse gases in total emissions in 2004 in terms of CO_2. (c) Share of different sectors in total anthropogenic greenhouse gases emissions in 2004 in terms of CO_2. Adapted from Climate Change (2007).

of atmospheric methane as a result of a complex array of soil processes involving plant microbe interactions in the anaerobic microenvironments. Methanogenic bacteria in soil utilises the carbon sources supplied by the rice plants and other incorporated organic substrate to produce a massive amount of methane.The correlation between methane emission and grain yield was demonstrated by Denier van der Gon et al. (2002). The emission of methane was supervised at different degree of spikelet removal. As more spikelet was removed, the rice fields showed high methane emission and reduced grain filling. Low rice yield leads to photosynthetic production of excess carbon that is partially transferred below ground, where it can act as a carbon source for methanogensis (Minoda and Kimura, 1994; Minoda et al., 1996; Cheng et al., 2007). Their results strongly suggest that methane emission from rice agriculture can be curtailed by optimizing rice production to maximum rice yield. According to this outcome, it shows that methane emission decreases when grain yield increases.

NITROGEN GASES FROM RICE FIELD

Among the greenhouse gases, nitrous oxide (N_2O) is about 310 times more capable to trap heat than carbon dioxide on a molecular basis. Due to heavy use of nitrogenous fertilizers in rice field, this gas is produced but not in significant amount to make effect on earth surface (De Datta and Buresh, 1989). Both carbon dioxide and methane are important greenhouse gases involved in rice cultivation. In practical, many people try to reduce greenhouse gases by avoiding flooding in rice field and increasing carbon assimilation in crops.

DELIVERING C4 TRAITS TO C3 PLANTS TO INCREASE THE FIXATION OF CARBON DIOXIDE

Carbon fixation is a process found in autotrophs, usually driven by photosynthesis, whereby carbon dioxide is converted into organic compounds. In plants, there are three types of carbon fixation during photosynthesis: C3, C4 and crassulacean acid metabolism (CAM). C3 plants such as rice use an enzyme called ribulose bisphosphate (RuBP) carboxylase (RuBisCO) which fixes atmospheric carbon dioxide into a stable 3-carbon compound called 3-phosphoglycerate (3-PGA). This reaction occurs in C3 plants as the first step of the Calvin cycle in the chloroplast. Although RuBisCO favors carbon dioxide to oxygen (approximately 3 carboxylations per oxygenation), but at high oxygen concentration RuBisCO often incorporates an oxygen molecule into the RuBP instead of a carbon dioxide molecule. This breaks the RuBP into a three-carbon sugar that can remain in the Calvin cycle, and two molecules of glycolate which is oxidized into carbon dioxide, which waste the cell's energy (ATP is not produced). However, at high oxygen concentration the competition of oxygen with carbon dioxide molecules at the active site of Rubisco results in a loss of some fixed carbon molecule in form of carbon dioxide in a process known as photorespiration (Ogren, 1984). Oxidation of ribulose-1, 5-bisphosphate (RuBP) severely diminishes the efficiency of carbon dioxide assimilation in C3 plants.

In C4 plants, carbon dioxide is drawn out of malate and into four-carbon compound malate and into Calvin cycle rather than directly from the air. This additional step elevates the concentration of carbon dioxide around RubisCO, and the high CO_2 concentration block the photorespiration and hence enhances the efficiency of carbon dioxide assimilation. Under optimal growth condition, C4 plants are more productive than C3 plants (Brown, 1999). C4 plants exhibit higher water and nitrogen use efficiencies and results in an increased dry matter production.

Researchers all over the world attempt to improve the carbon dioxide assimilation in C3 plants based on C4 plants. A consortium of agricultural scientists is setting out to re-engineer photosynthesis in rice in hope of boosting yields by 50%. They outlined a number of ways rice could be turned to a C4 plant such as through conventional hybridization and transgenic plants (Normile, 2006). Conventional hybridization between C3 and C4 plants may take a lot of time and is available only in few plant genera. Even more, most C3-C4 hybrids showed infertility due to abnormal chromosome pairing (Brown and Bouton, 1993). With the development of biotechnology, the most direct way to transfer C4 traits to C3 plants is by plant genetic engineering. A successful accomplishment is the simultaneously overexpession of phosphoenolpyruvate (PEP) carboxylase (PEPC) and pyruvate orthophosphate dikinase (PPDK) in rice. PEPC is an enzyme that catalyzes the first step of C4 cycle to fix carbon dioxide in mesophyll cells of leaf and are absent in C3 plants. In C4 plants, 4-carbon com-pounds are synthesized by PEP carboxylase using PEP and carbon dioxide. On the other side, PPDK catalyzes the conversion of pyruvate to PEP which is the substrate of PEP carboxylase. Combined overexpression of PEP carboxylase and PPDK in rice plants enhances its photosynthetic capacity linked to an increased provision of PEP for the Shikimate pathway. Ku et al. (2001) have reported that the transgenic plants exhibit a 35% increase in the photosynthetic capacity and also grain yield increased by 22%. Furthermore, the transgenic rice plants tend to have a high photosynthetic rate due to a high level of stomata conductance and intercellular carbon dioxide.

This increased photosynthetic capacity of transgenic rice leads to increase efficiency of carbon dioxide assimilation, which greatly reduces carbon dioxide in atmosphere. On the other hand, the methane emission in rice field reduced with elevated grain yield. Transgenic plants with combined overexpression PEPC and PPDK in rice successfully reach the goal to reduce the emission of greenhouse gases although the mechanism is still unclear and waiting for detailed analysis. Several C4-cycle enzymes were introduced to C3 plants to improve the efficiency of photosynthesis (Hausker et al., 2002). With no expectation, the genes engineered into rice and other C3 plants are enzymes and metabolite transporters in photosynthesis. All these enzymes and transporters involved in the C4 pathway also occur in C3 plants, although with lower activities and different tissue specificities. These enzymes may have distinct function in C3 plants (Table 1).

Genetic engineering of C4 traits to C3 plants all elevate the enzyme activity several fold in rice, but may not always work to advance photosynthesis in all C3 plants. After overespression and increasing the activities of these genes in rice, it is likely to perturb metabolism or trigger compensation changes in metabolic fluxes. These transgenic plants sometimes show different functions from previous expectation and may be an effective tool to figure out the crosstalk between different metabolic pathways. However, they have no benefit on reducing greenhouse gases.

There has been significant progress in the overexpression of the key enzymes of C4 type biochemistry in transgenic C3 plants. Nevertheless, it is still uncertain whether this approach will be sufficient to suppress photorespiration. Apart from this misgiving, there are still lots of problems encountered. As with traditional hybridization, transgenic plants with combined overexpression of PEPC and PPDK in rice also have a lower fertility. It is a tremendous barrier when shifting to practical application. In addition, whether C4 traits can be stably inherited in the following generation needs to be evaluated. Furthermore, several-years field tests must be executed in transgenic plants before marketing. This is not only for human safety but also for environmental protection.

REDUCTION OF GREENHOUSE GASES BY IMPROVED MANAGEMENT SYSTEM

One of the most prominent greenhouse gases mitigation options in agriculture is to improve cropland management including agronomy, nutrient management, tillage/residue and water management (including irrigation and drainage) and set-aside/agroforestry (Smith et al., 2008). The researchers from AgNeeds, an agriculture company in Taiwan, set up a management system based on the physiological needs at different growth stages of plants to improve conventional fertilizer management system to achieve the optimal efficiency of land use and fertilizer utilizetion. This management system increases the rice yield by three products: by adding Pollen Extract, Rootking and Actzyme Plus at different growth stages. Pollen Extract is a special plant extract able to activate cells to make complete internal organelles for energetic biochemical reactions resulting in plant overall development. Rootking, contains organic acids, vitamin B groups and special ingredients, is a unique formulation designed to increase root hair density to greatly enhance absorption surface of nutrients for sufficient fertilizer utilization. Actzyme Plus facilitates degradation of organic matters in

Table 1. Some proposed functions of 'C4-cycle enzymes' and transporters in C3 plants.

C4-cycle enzymes and transporters	Location	Proposed metallic function
Enzymes		
PEPC	Non-green tissues	Recapture of CO_2 respired
	Leaves	Anaplerotic supply of carbon skeleton for amino acid biosynthesis
		Buffering cytosolic OH^- formation during nitrate reduction by malic acid formation.
	Stomatal guard cells	Formation of malic acid during stomatal opening
NADP-ME	Fruits	De-acidification of vacuoles. Provision of reducing equivalents and carbon skeletons for gluconeogenesis.
		In combination with PEPC involved in PH-stat. In combination with NAD-MDH involved in NADH/NADPH conversion
	Seeds	Provision of reducing equivalents and carbon skeletons for fatty acid biosynthesis.
	Leaves	In vascular bundles, provision of reducing equivalents for lignin biosynthesis, Stress responses.
NAD-ME	Leaves	Together with PEPC involved in anaplerotic provision of carbon skeletons for amino acid biosynthesis.
NADP-MDH	Leaves	Reduction of OAA in the chloroplast. Shuttling excessive redox equivalent (malate valve) into the cytosol (mitochondria)
PEPCK	Non green tissues	Gluconeogenetic PEP production from OAA
	Trichomes	Involvement in secondary metabolism.
		Largely unresolved. (PEP production For the shikimate pathway?)
	Stomatal guard cells	Gluconeogenetic PEP regeneration n from pyruvate stomatal closure
Transporters		
Pyruvate		
Malate OAA	Leaves	Malate valve, see NADP-MDH
PPT		Privision of PEP for the shikimate pathway inside the chloroplast (also fatty acid biosynthsis.

Adapted from Häusler et al. (2002).

the soil. Biological enzymes within this reagent enhance plant-friendly microbes and suppress soil pathogens. Foliar spray of Actzyme Plus enhances photosynthetic products accumulation for rice grain growth. Application of Actzyme optimize carbon/nitrogen ratio resulting in high quantity and high quality rice grains. It is different from transgenic plants or hybridization; they offer extra nutrients to rice based on the plant physiological needs. In seedling stage, they bring basic nutrients, nitrogen, phosphate, and potassium and micro-elements and rare micro-elements to activate metabolism reaction of rice. Until vegetative growth, nitrogen uptake is enlarged to promote leaf elongation and expansion to enhance photosynthesis. Proper biochemical metabolism facilitates carbon conversion into carbohydrate contents of rice grains when flower and grain development (reproductive growth) occurs. More carbon is converted into nutrients; less carbon being released into soil, meanwhile the methane metabolized by anaerobic organism was reduced. Healthy root system was promoted by addition of friendly micro-flora environment in soil which facilitates fertilizer absorption. While crop develops at the optimal physiological status, applied fertilizers will be converted efficiently into tissues and grains as food. It has been scientifically shown rice yield increase can capture the atmospheric carbon dioxide into biomass; additionally better yield performance can reduce methane emission from the land. This is a practical case which elevate rice yield and reduce greenhouse gases. Under the same cultivation condition farmers in Pingtung County, Taiwan did not change their major fertilizers and cultivation machines. After using the management system from AgNeeds, the productions of rice paddy in the first crop season are list in Table 2. The rice variety is Japonica Tainan 11. According to the information from the National Plant Genetic Resource Center of Taiwan Agriculture Research Institute, the average grain yield of Tainan 11 is 7,497 kg/ha.

The proportion of carbon element in carbon dioxide is 12/44 (3/11). When plants absorbed 110 g carbon dioxide, 30 g carbon in the carbohydrates and cellulose by photosynthesis are acquired. There are about 30 g carbon in

Table 2. Area and rice yield of four different farmers using AgNeeds products.

Name	Avg. area of farms (ha)	Wet weight (kg)	Wet weight (kg/ha)	Dry weight (kg/ha)
Mr. A	0.775946	9600	12372	9898
Mr. B	0.290980	3780	12991	10393
Mrs. C	0.290980	4260	14640	11712
Mr. D	0.969932	11820	12186	9749

Adapted from http://tw.myblog.yahoo.com/agneeds-tw.
Dry weight is equal to wet weight multiplied by 0.8.

80 g carbohydrates. According to the food composition table, 100 g rice contains 81.0 to 82.0 g carbohydrates and cellulose excluding fatty acids, protein and other nutrient containing carbon (Table 3). As a result, we estimate that with increase of 100 g rice, 110 g carbon dioxide in the atmosphere would be reduced. With one extra kilogram rice production, 1100 g carbon dioxide in the atmosphere would be absorbed. The cultivation areas of the farmers in Pingtung County cooperated with AgNeeds are 2.23 hectares in total. Compare to the average grain yield, the cultivation management made the yield increase of 6555 kg. Their work reduce 6555 X (1.1/1) = 7.2 tonnes of carbon dioxide in the world.

Applying management system of AgNeeds, rice yield per unit farming area can increase up to 30%. The majority of rice biomass comes directly from the atmospheric carbon dioxide which is fixed by photosynthesis and transformed biologically into valuable food, rice grains. The more biomass obtained, the more carbon dioxide is transformed to rice grains and the less carbon delivered to soil. This kind of eco-farming system from AgNeeds not only increases the rice yield but also mitigate greenhouse gas emission.

Table 3. Nutrients factors about rice.

Nutrition facts serving: 100 g	White rice	Jasmine
Calories (kcal)	361	355
Moisture (water) (g)	10.2	11.9
Total Fat (g)	0.8	0.7
Dietary Fiber (g)	0.6	0.8
Calcium (mg)	8	5
Phosphorus (mg)	87	65
Potassium (mg)	111	113
Sodium (mg)	31	34
Vitamin B1 (mg)	0.07	0.12
Vitamin B2 (mg)	0.02	0.02
Niacin (g)	1.8	1.5
Protein (g)	6	6.1
Carbohydrates (g)	82.0	81.1

Adapted from Thai Food Composition Table by Institute of Nutrition, Mahidol University (Puwastein et al., 1999).

CONCLUSION

United Nations and other sources indicate that world population could grow upto about 8.5 billion by 2025 (Keyfitz, 1989) and to 11 billion by the end of the coming century (UNFPA, 1990). Global agricultural production of rice will need to increase several times from present levels to meet the needs of food. However, agriculture accounts for 14 and 52% of global anthropogenic carbon doxide and methane emissions, respectively. The emission of greenhouse gases in rice paddy could be mitigated accompanied with increased irrigation, fertilizer use, and developing new high-yield rice varieties. However, the cost is too high to farmers and they are reluctant to drain the field or apply measure by employing more labors. For this reason, it is urgent that we develop other strategies to lower global warming, by biotechnology to manipulate genes or by integrated management system. Genetic engineering is an effective strategy to control greenhouse gases from rice, but it needs long-term field test and may have a potential side effect on human safety and our environment. Integrated management system is also an effective and even better strategy. The integrated management system can be applied to rice field immediately to relieve the global warming right away. In this case, integrated management system seems a more powerful way to reduce greenhouse gases in short-term. There will be a great potential to stabilize or even reduce greenhouse gases from rice fields while increasing rice production without dramatically changing cultural practices if we can combine both technologies in the future.

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