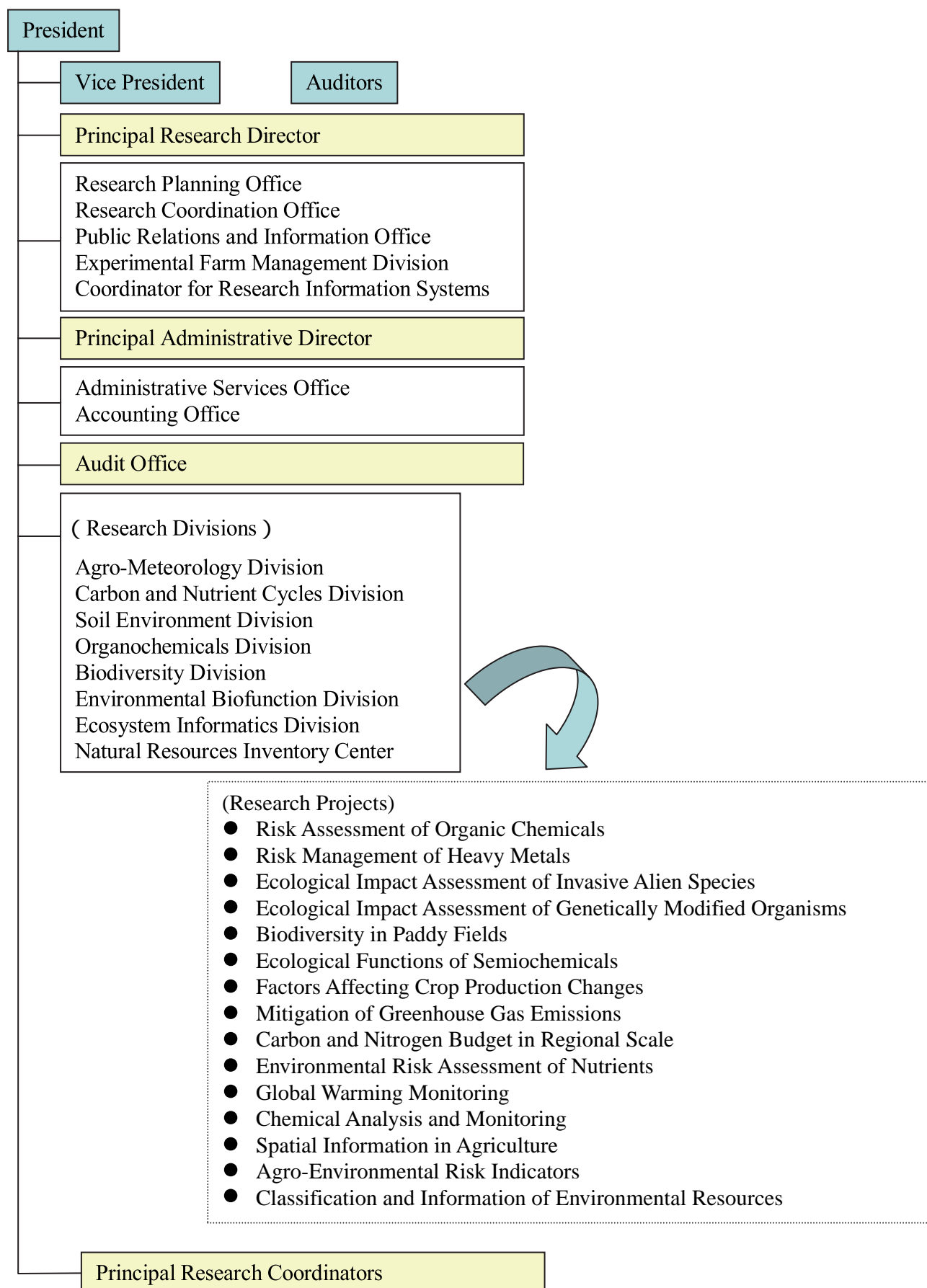


# ***Research Overview in 2008***



## Research Organization



## Summary of NIAES Research Projects

The National Institute for Agro-Environmental Sciences is emphasizing the following research themes in three research divisions during the medium-term research period (2006–2010) while continuing to prioritize and specialize in our work toward basic studies and research aimed at assuring the safety of agricultural production environments.

- A Assessing risks to agro-environments, and developing technologies to manage them**
- B Determining the structures and functions of agro-ecosystems and developing management technologies to elicit the functions of natural cycles**
- C Basic research to help elucidate the functions of agro-ecosystems**

Below we present the medium-term plan of the five-year medium-term research period and an overview of the research conducted in 2008.

- A. Assessing risks to agro-environments, and developing technologies to manage them**
  - 1) Development of risk management technologies for hazardous chemicals in agro-ecosystems**
    - (1) Development of a method to assess the risks of hazardous chemicals in agro-environments, and risk management technologies**

### Medium-term plan:

To reduce the contamination risk of cadmium, arsenic, radioactive substances, persistent organic pollutants including drin-family pesticides, and other chemical substances in agro-environments, we will

determine their dynamics and develop technologies to reduce risk. For organic chemicals such as pesticides, we will develop a model to predict their behavior in the environment, and also develop a way to assess their environmental risks by, for example, performing exposure tests using aquatic arthropods and other organisms. Additionally, we will develop technologies to restore soil contaminated with hazardous chemical substances using methods such as chemical washing or bioremediation, and develop techniques such as using cultivars with low absorption of these substances.

### Research Overview:

- a. Development of a method to assess the environmental risks of pesticides and other substances, and development of risk abatement technologies
  - (i) Development of a method to assess the risks of organic chemicals

We improved the spatial resolution (from 15° latitude × 30° longitude per grid cell to 5° latitude × 15° longitude per grid cell) of the multimedia model (NIAES-MMM), developed in 2007, which can assess the global dispersion of contamination by organic chemicals, and we made the model more sophisticated.

By measuring the change over time in the concentrations of various rice paddy herbicides and their metabolites in rivers we found that for a few herbicides, the concentrations of metabolites were higher than those of their parent compounds, which suggests the need to also take metabolites into consideration when assessing ecosystem impacts. For that purpose we improved the model for predicting pesticide concentrations in river

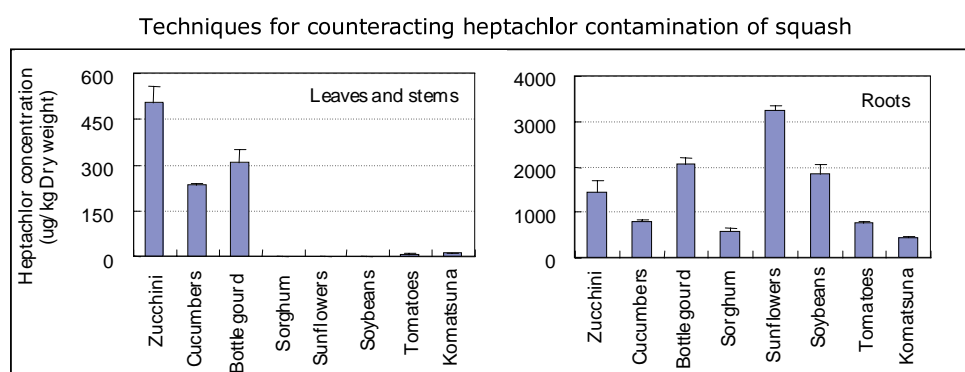


Fig. 1 Heptachlor concentrations in leaves and stems and roots of various young crop plants raised in soil with residual pesticides.

I: Standard error ( $n = 3$ )

In plants of the Cucurbitaceae family, heptachlors were detected prominently in the leaves and stems, but they were detected in very low concentrations in plants of other families. This phenomenon, the same as is seen with dieldrin absorption by Cucurbitaceae plants, revealed the specificity of Cucurbitaceae for the absorption of heptachlors.

water (PADDY-Large), thereby enabling it to also predict the behaviors of metabolites, and we confirmed the model's soundness by comparing its predictions with monitoring data from the midstream part of the watershed of Sakura River in Ibaraki.

(ii) Development of technologies to reduce the risks of organic chemicals

(Achievements ready for deployment: soil extraction method that can predict the concentration of residual dieldrin in cucumbers, and a technology to counteract heptachlor contamination of squash)

In recent years dieldrin has been detected in cucumbers in concentrations exceeding the residue standard, which has obliged producers to respond by introducing voluntary production curbs and other such measures. For this reason we examined ways to predict

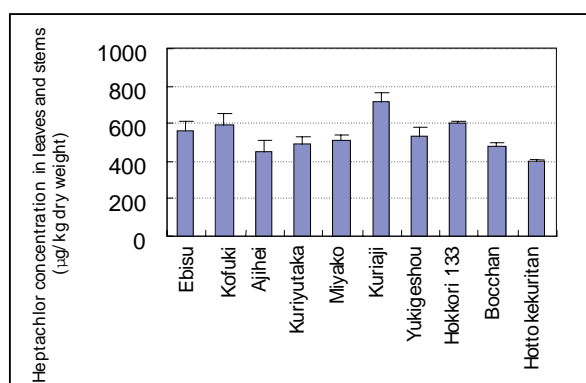


Fig. 2 Differences between squash cultivars in the concentrations of heptachlors in the leaves and stems of young plants raised in soil with residual pesticides.

I: Standard error ( $n = 3$ )

Differences between squash cultivars in their absorption of heptachlors raise hopes that using low-absorption cultivars can to an extent reduce contamination.

the residual dieldrin concentration in cucumbers before growing them. We found that the concentration of dieldrin in soil obtained by extraction with 50% methanol/water (v/v) indicated the dieldrin concentration in the body of cucumber plants regardless of the soil type. There are expectations that this research can be used as a method for judging the appropriateness of cucumber cultivation, and currently NIAES is conducting verification testing jointly with regional agricultural research institutes.

Additionally, in recent years heptachlors have been detected in squash in excess of the residue standard. Heptachlors disappear slowly in soil, and even now, more than 30 years since the banning of heptachlor as an insecticide (in 1975), there are residues in farmland. We therefore examined the applicability of several ways to counteract heptachlor in squash that were found to be effective for cucumbers: switching to low-absorption crops, selecting low-absorption cultivars, and reducing absorption of crops by using absorbent materials. We found that these techniques can all be used.

b. Development of a method to assess the risk of heavy metal contamination and a technology to remediate contaminated soil

(i) Development of a method to assess the risk of heavy metal contamination

We developed a method that can estimate the total soluble cadmium in soil from the cadmium isotope ratio of soil to which the stable cadmium isotope  $^{111}\text{Cd}$  has been added. This method can be used to assess the risk of soil cadmium uptake by crops.

(ii) Development of a technology to reduce heavy metal risk

To verify the effectiveness of a cadmium chemical washing method, which has been proven for rice paddy

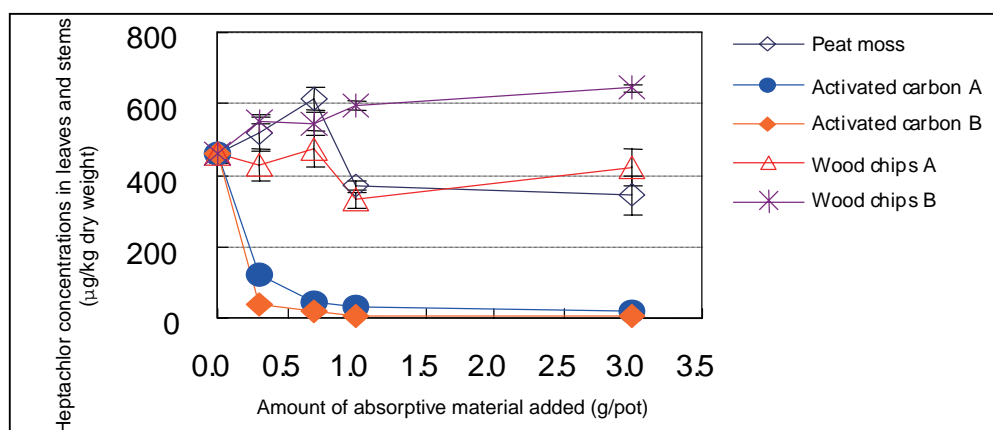


Fig. 3 Effectiveness of mixing adsorption material into soil in reducing absorption of heptachlors by squash.

I: Standard error ( $n = 3$ )

Just as with dieldrin in cucumbers, activated carbon material effectively reduced absorption of heptachlors by squash.

soil, as a measure to reduce the cadmium contamination of wheat, vegetables, and other crops grown in upland fields converted from rice paddies, we conducted a wheat cultivation experiment in a converted rice paddy which had been chemically washed in 2006, and also performed a pot cultivation test (with okra) using soil from the same chemically washed field. Results confirmed that in the chemically washed field the effectiveness of soil Cd removal was almost fully maintained: the Cd content in the unmilled grain from the washed field was reduced to about 50% of that from an unwashed plot; the reduction in the okra pot test was about 54%. These values were near the soil Cd removal rate, demonstrating the effectiveness of reducing Cd absorption risk by means of chemical washing.

To develop a phytoremediation technology for upland field soil contaminated with Cd, we cultivated rice plants (cultivars: IR8 and Chokoukoku) and sorghum (cultivars: G Sorgo and H Sorgo) under upland field conditions, and compared them for inter-cultivar differences in cadmium absorption in the aboveground parts. Chokoukoku, the upland-grown rice cultivar with the highest aboveground cadmium absorption, appeared to be a promising cultivar as a remediation crop for upland fields.

For the development of technologies using cultivars with low heavy metal absorption, the National Agriculture and Food Research Organization and the National Agricultural Research Center for Tohoku Region worked together in selecting five Ukei rice lines whose brown rice Cd concentration is low and whose cultivation characteristics are improved. The brown rice Cd concentrations of these five lines are about half those of generally used cultivars, raising expectations for their use as cultivars with low Cd absorption.

## **2) Development of technologies for managing the risks of exotic organisms and genetically modified organisms in agro-ecosystems**

### **(1) Development of technologies to assess ecosystem impacts and manage the risks of exotic organisms and genetically modified organisms**

#### **Medium-term plan:**

In order to prevent disturbance and damage to agro-ecosystems by exotic organisms (which invade or are introduced), we will determine the growth and propagation characteristics, allelopathy, and other attributes of exotic organisms, determine the extent of damage by exotic organisms, and predict their ecesis, spread, and damage. Also, we will identify the places where exotic organisms originate, and estimate the probability of invasion. Further, we will analyze the impacts of exotic organisms such as exotic natural enemy insects on closely related native species from aspects

such as competition and hybridization, and assess the risks that exotic organisms present to agro-ecosystems. Additionally, we will develop techniques using molecular markers and other means to quickly detect and monitor exotic organisms whose species are hard to identify. In order to appropriately assess the impacts of genetically modified organisms (GMOs) on ecosystems, we will develop a technology that uses DNA markers and other means to detect hybridization of genetically modified crops and their closely related species, such as soybeans and wild soybeans *Glycine max* subsp. *soja*, and determine the ecosystem impacts due to hybridization. To prepare for the coexistence of crops that are genetically modified and those that are not, we will develop a hybridization rate prediction model and cultivation techniques, such as maintaining isolation distance, to reduce hybridization.

#### **Research Overview:**

a. Assessing the ecosystem impacts of exotic organisms, and developing risk management techniques

(i) State of ecesis by specified exotic organisms

Field studies of rice paddies in the area of Lake Inba, Chiba Prefecture, found that the specified exotic organism alligator weed *Alternanthera philoxeroides* is damaging to agriculture because it infests dikes, paddies, and water channels, and that it is also distributed in narrow valley paddies, which are separate from these water channel systems. It was conjectured that the agricultural methods employed in these fields has much to do with the expanding distribution of this plant. China has serious infestations of alligator weed, and caution is needed.

We determined the distribution of the golden mussel *fortunei*, a specified exotic organism, from the mouth of the Tone River to its confluence with the Karasu River in Gumna Prefecture. In the Tone River we found the mussel from the river mouth to the 116-km point. The mussel was found not only in the Tone River's main channel, but also in nearby rivers, lakes, water channels, and water withdrawal facilities, and it was confirmed that the operation of water utilization facilities in Ibaraki, Chiba, and Gunma prefectures is adversely affected.

(ii) Relationship between exotic plant distribution and soil chemical characteristics

We conducted a survey of herbaceous plant communities around farmland in the northern Kanto region, and found that the invasion of exotic plants is frequent in places where surface soil pH is high and in soil with much available phosphorus.

(iii) A method of assessing exotic plant risks

We determined that the Australian weed risk assessment system, which has an established international reputation as a pre-entry plant risk

assessment system (a system which distinguishes between plants that would and would not become weeds in Japan, before the plants are introduced into Japan), can be used in Japan. We also proposed a method for deciding weed determination criteria which, in addition to a method that decides based on the past weed differentiation rate, also considers external factors such as economic benefit, and the accuracy of the assessment method. We hope these research accomplishments will be used by the Ministry of Environment and other administrative agencies that are responsible for laws related to exotic organisms.

b. Assessing ecological impacts of genetically modified organisms, and developing a risk management technique

(i) Research on assessing biodiversity impacts (research on escape and hybridization)

Because of reports of genetically modified oilseed rape that has sprung from spilled imported rape seeds at Kagoshima Port and several other Japanese ports, we carried out a test of competition with other weeds in a field by varying the planting time and harvest conditions of rape for the purpose of seeing whether this exotic cultivar will drive out other species and widen its distribution. Results showed that planting time and whether or not the plants were harvested considerably affected the growth of this cultivar; however, in every test plot the dense growth of other weeds suppressed the growth of the rape, with just a few individuals appearing and then dying in each case. This suggested only a small possibility that this rape will drive out other weed species.

It has been pointed out that when genetically modified soybeans are cultivated, they might hybridize with wild soybeans, a closely related species that grows wild in Japan. In an NIAES field we found that even when the two species are cultivated so that their flowering periods overlap to the maximum possible extent, and close enough for their branches to intertwine, the hybridization rate was extremely low. Because the frequency of hybridization is affected by the extent to which the flowering periods of both species overlap, we examined the geographical variation in wild soybean flowering characteristics for the purpose of creating a model to predict the possibility of hybridization based on overlapping flowering period. We found that of five wild soybean strains from Hokkaido, Akita, Ibaraki, Saga, and Miyazaki, in all except for the Miyazaki strain, the start of the flowering period was determined more by effective cumulative temperature than by day length, and that the farther south a strain's origin, the higher effective cumulative temperature it required. We also found that for the day of maximum flowering and the end of

flowering, the farther south a strain's origin, the shorter the day length it responds to, showing that the flowering maximum and end are governed by day length.

(ii) Coexistence research (research on suppressing hybridization)

To find the extent of long-range scattering of rice pollen and the extent of hybridization in large fields, we used a hybridization prediction model to prepare a hybridization rate prediction map for the seed parent field, efficiently collected a large number of samples, and found the relationship between the distance from the pollen parent and the hybridization rate.

To develop a technique to control the scattering of pollen from corn and other anemophilous crops to suppress hybridization, we tested the efficacy of suppressing hybridization with windbreak vegetation and by sprinkling water. By comparing the hybridization rates over three years we found that when separation from the pollen parent is 10 m or more, the hybridization suppression effect almost disappears, showing that the hybridization suppression effect is limited to that of a windbreak net.

## **B Determining the structures and functions of agro-ecosystems and developing management technologies to elicit the functions of natural cycles**

### **1) Determining and assessing the structure and functions of agro-ecosystems**

#### **(1) Illuminating the dynamics and biodiversity of the communities that make up agro-ecosystems**

##### ***Medium-term plan:***

To conserve agriculture-fostered biota and their biodiversity, we will investigate the plants, birds, insects, nematodes, microorganisms, and other organisms inhabiting farmland and surrounding areas. We will ascertain how their species composition and diversity is affected by the tilling of farmland and use of chemicals, crop changes and fallowing, changes in how surrounding vegetation and irrigation ponds are managed, and changes in the landscape structure of rice paddies and their surrounding areas. Additionally, we will use the results to find population stabilization factors by building a model to predict the dynamics of populations of indicator insects and other organisms in conjunction with changes in land use and other agricultural activities.

##### ***Research Overview:***

(i) Development of an information system, and finding the relationship between landscape structure and biodiversity

We created a system using Web-GIS to improve the agro-ecosystem portion of the Rural Landscape Information System (RuLIS) and to make the data of the agro-ecosystem portion (classified into 60 classes

nationwide) available to the public. Additionally, we used the Digital National Land Information gridded land use data with 1/10 cell subdivisions (about 100 m on a side), which are provided for multiple years and based on a national standard, to calculate the extent of land use mixing within a three-dimensional grid (about 1 km on a side), and showed that this can be used as an indicator of valuable landscape structures such as boundary areas between rice paddies and forest.

(ii) Ascertaining the effects of irrigation ponds, rice paddies, and their peripheral environments on the habitats of indicator insects

We assessed the importance of the disposition of irrigation ponds to dragonflies in comparison with the influences of a pond's internal environment and the land use in its surrounding area. We found that for dragonfly species with limited flying ability, the disposition of an irrigation pond is most important, and we found that for all species, pond disposition has about as important as the effects of internal pond environment and surrounding area land use. Dragonflies are one kind of indicator insect (insects which are environmental indicators) to watch amid changes in irrigation facilities and cultivation techniques and our results are important for effectively pursuing initiatives aimed at conserving the diverse dragonfly species that inhabit rural areas.

(iii) Finding the impacts of changes in agricultural methods, such as the use of chemicals, on aquatic plant communities near rice paddies

To find what effects agricultural methods such as herbicide use have on the aquatic plant communities near rice paddies, we performed simultaneous monitoring of aquatic plant communities and herbicide concentrations in irrigation channels near rice paddies. We found that

there was a pronounced impact on submerged plants (such as curly pondweed *Potamogeton crispus* L) when herbicides are applied the most (May), whereas the emergent plants cattails *Typha latifolia* and reeds *Phragmites australis* were hardly affected. Additionally, our research suggested the possibility that curly pondweed populations in irrigation channels kept repeating a cycle of disappearance and regeneration.

### (2) Unlocking the secrets of semiochemicals involved in expressing agro-ecosystem functions

#### Medium-term plan:

To maintain and improve agro-ecosystem functions, we plan to find and determine (1) the functions of the bioactive substances produced by plants such as those of the Rosaceae family, (2) the substances involved in the interactions among organisms, such as the semiochemicals involved in the propagation of the Pyraustinae and other insects, and (3) the substances that control the expression of genes involved in the decomposition of compounds such as persistent chlorinated aromatic compounds in bacteria groups such as those of the genus *Burkholderia*.

#### Research Overview:

(i) Determining the functions of the bioactive substances produced by plants such as those of the Rosaceae family

The activity of the cis-cinnamoyl glucosides in Thunberg's meadowsweet *Spiraea thunbergii* to inhibit plant growth hardly varied in alluvial soils. In volcanic ash soils and calcareous soils the inhibitory activity declined to between 1/2 and 1/10, but even then it was clear that the level of inhibitory activity was high enough to manifest its function in ecosystems. We will continue research to gain new knowledge that will lead to

Attractant using sex pheromones for predicting outbreaks of pear fruit moth.



Photo 1 Sex pheromone trap.

Rubber impregnated with sex pheromone constituents is put inside this trap as the attractant. Attracted insects are captured by the adhesive plate on the trap floor.



Photo 2 Male pear fruit moths captured in the sex pheromone trap.

The number of insects caught in the trap tells us whether adults have appeared, and the time period of their appearance. Such information enables farmers to efficiently take preventive measures such as insecticide applications.

developing new weed suppression technologies and to bettering our understanding of ecosystem biodiversity.

- (ii) Determining the functions of the semiochemicals involved in the propagation of insects such as those of the Pyraustinae  
(Achievements ready for deployment)

We extracted sex pheromones from female "tsuwabukinomeiga" moths, investigated the reactions of males to candidates for the active pheromone constituent, and identified three constituents that are necessary to attract males. Additionally, we extracted the sex pheromone from adult female pear fruit moths *Ectomyelois pyrivorella* and identified the attraction constituent; conducted a field study to examine the active constituents and their ratios, support amount and effective time span as attractant; and developed an attractant for predicting outbreaks.

- (iii) Understanding the functions of microorganisms that decompose biodegradable plastic  
(Achievements ready for deployment)

From among the more than 1,500 strains of fungi found living on the leaf surfaces of Gramineae crops, we selected a mold (strain 47-9) which belongs to the imperfect fungi and which has a particularly high activity for decomposing biodegradable plastic, and we found the conditions under which the enzyme activity is the greatest. When cultured in a liquid medium this mold has the excellent characteristic of producing only an enzyme of high purity which breaks down biodegradable plastic. When the enzyme solution from this fungus was applied to biodegradable plastic film (polybutylene succinate adipate, or PBSA) that had been placed over commercially sold potting soil, 91.2% (by weight) of the plastic was decomposed in 6 days.

This is a valuable finding that raises hopes for the development of a new technology that facilitates the decomposition of used biodegradable plastic products.

## **2) Understanding the mechanisms of change in agro-ecosystems, and developing technologies to counter them**

### **(1) Predicting impacts of changes in the global environment on agro-ecosystems, and assessing the risks to production**

#### **Medium-term plan:**

In order to assess on a field scale the changes in rice yield due to global warming and extreme weather events, we will develop an integrated rice paddy ecosystem response model that includes water, soil, rice plant cultivars, and cultivation conditions. Additionally, we will develop a regional-scale simple yield model, predict changes in mid-century rice yields mainly in Japan and other Asian countries on the basis of yields and water

resources, and develop a method for wide-area assessment on a regional scale of the risk of declining rice yields. Also, based on these results, we will construct scenarios to predict the impacts of climate change on food production.

#### **Research Overview:**

- (i) Field-scale impact assessment (development of a rice paddy ecosystem response model)

Free atmospheric CO<sub>2</sub> enrichment (FACE) experiments and global warming experiments over 2 years yielded quite consistent results for the effects of increased CO<sub>2</sub> (+200 ppm) and higher water temperature (+2°C). Specifically, subjecting plants to higher temperatures and CO<sub>2</sub> concentrations increased dry weight, but the extent of increase declined with each growth stage. Also, we found that the decreases in response of photosynthesis and the maximum carboxylation rate, which are related to the above, occur mainly via the decrease in photosynthesis-related proteins. This provided verification data which showed, under open-system conditions, the validity of our crop model's basic structure.

In an environmentally controlled chamber (a climatron) we studied the effects of nighttime temperature during the flowering period on the fertilization process in rice. It is known from past chamber experiments that sterility results when temperature at flowering time exceeds 35°C; however, this study found that even when daytime temperature is about 32°C if nighttime temperature is high, anthers burst with difficulty, which results in much sterility. The threshold nighttime temperature for this to happen is 28–30°C; and there are marked differences among cultivars in the sterility rate under high nighttime temperature conditions.

- (ii) Regional-scale impact assessments

To assess nationwide impacts in Japan, we analyzed the results of long-term field cultivation studies on rice cultivars at eight locations to gauge the impacts of climate change trends in recent years on rice growth throughout Japan. We found that the number of days from transplanting to heading has markedly shortened almost nationwide (on the average, about 7 days over 25 years). On the other hand, we discerned no significant tendency toward yield decline. Additionally, because the field-scale model previously developed soundly reproduced the above long-term change, it is conceivably valid for reanalysis of the situation and for predictions.

To assess impacts in East and Southeast Asia, we applied the regional-level yield model developed in 2007 to a wide area of East Asia (provincial yields in China). We found that the model can be used to reproduce and predict the spatial and temporal change in regional yields

based on climate, nitrogen application, and cultivar differences. A model for the rainfed rice district in northeastern Thailand found that values for planted area estimated on the basis of rainfall amount do a good job of qualitatively explaining the changes in planted area according to agricultural statistics. Combining the planted area prediction model with a growth/yield prediction submodel made it possible to calculate changes in planted area, yield, and production by grid cell based on changes in water resources.

## **(2) Determining the impacts of agricultural activities on material cycles**

### ***Medium-term plan:***

To help solve regional and global environmental problems related to greenhouse gases, nitrogen, and other substances arising from agricultural activities, we will determine the impacts of agricultural activities on material cycles and develop measures to mitigate the burden. For greenhouse gases, we will propose a technology system for efficient burden mitigation by quantitatively assessing the efficacy of reducing greenhouse gas emissions by means of cultivation and soil management technologies. At the same time, we will use soil-related databases to verify and improve models that describe soil carbon dynamics, and predict changes in the amount of carbon accumulation in the soil of Japan's farmland that occur in conjunction with changes in climate and human management. Additionally, by using an acidifying substance dynamics model, statistical data, and other means we will estimate and predict nitrogen flows and stocks as a function of food production, imports, and exports, and we will ascertain and predict the wide-area nitrogen cycle and environmental burden on the watershed scale and national scale in East Asia. On the watershed level, we will determine the dynamics of flows of nitrates, phosphates, and other nutrients in the pedosphere including in shallow groundwater, and develop a method to assess vulnerability to water pollution.

### ***Research Overview:***

a. Quantitative assessment of technologies to limit the generation of greenhouse gases from farmland

(i) Quantitative assessment of technologies to limit the generation of greenhouse gases  
(Achievements ready for deployment)

At nine sites throughout Japan we conducted verification tests on techniques to limit CH<sub>4</sub> generation through rice paddy water management, such as by starting the midsummer paddy drainage period earlier or by extending it. At many sites, improved water management, such as extending the midsummer drainage period, was able to effectively reduce the amount of CH<sub>4</sub> generated; however, CH<sub>4</sub> was not reduced where there

was much rainfall during the drainage period (Aichi Prefecture) or where CH<sub>4</sub> emissions are concentrated in the late part of the cultivation period (Tokushima and Kagoshima prefectures). We also found that there was little N<sub>2</sub>O increase due to water management, and we gathered data about the impacts on rice yield, quality, and other relevant items.

We developed a portable automatic sampler for greenhouse gases emitted from farmland. It is possible with this sampler to make very frequent measurements of greenhouse gases over the whole year throughout Japan, and to more accurately estimate the flux of greenhouse gas from farmland.

(ii) Creating an overall greenhouse gas balance database, and developing a wide-area prediction model

We completed a database showing nitrogen flow by prefecture in conjunction with agricultural production every five years from 1985 to 2005, and determined the amount of N<sub>2</sub>O emitted in accordance with the Tier 2 method of the IPCC guidelines. Comparing 1990 and 2005 revealed that N<sub>2</sub>O emissions decreased in some areas and increased in others, but we found that in many cases, N<sub>2</sub>O emissions increased in a certain area if there was an increase in nitrogen from livestock waste even if there was less nitrogen being applied to farmland. On the whole, Japan's livestock sector emits more N<sub>2</sub>O than the crop farming sector, suggesting that the N<sub>2</sub>O emissions from managing livestock waste hold more sway over an area's emissions.

For the wide-area estimation of CH<sub>4</sub> emissions using the DNDC-Rice model, we calculated the breakdown of drainage (speed of surface water elimination and groundwater level) by soil group based on the GIS data of the Basic Survey of Land Use and Infrastructure Development (FY1993), and created a database. We also modified the DNDC-Rice model so that it calculates soil water flux from soil type and groundwater level, and we confirmed that changes in CH<sub>4</sub> emissions can be estimated from groundwater level.

(iii) Influence of high CO<sub>2</sub> concentration and global warming on CH<sub>4</sub> emissions from rice paddies

To determine the influence of high CO<sub>2</sub> concentration and global warming on CH<sub>4</sub> emissions from rice paddies, we continued work of the previous fiscal year with experiments in Shizukaishi Town, Iwate Prefecture using FACE (outside air + 200 ppm CO<sub>2</sub>) and elevated water temperature (+2°C). In both years the effects on CH<sub>4</sub> flux were the same. We observed a large effect from higher temperatures (+29% to +59%), and, although not significant, a FACE effect (+6% to +39%) about the same as those of past experiments.

b. Development of a wide-area assessment method for the balances of carbon and nitrogen associated with

agricultural activities

- (i) Wide-area verification with a soil organic matter dynamics model, and nationwide estimation of changes in soil carbon accumulation

To perform a wide-area assessment of the reliability of a soil organic matter dynamics model (improved RothC) and to develop a method to improve model parameters, we used the data for 2,843 upland field locations (non-andosols, 1,251 locations; andosols, 1,592 locations) and 5,285 rice paddy locations from the Basic Soil Environment Survey (a fixed-point survey), and we compared the estimated values for soil carbon accumulation amount generated by the model with measured values. Model estimates were generally on the mark.

We applied the soil organic matter dynamics model (improved RothC) to farmland all over Japan and estimated the effectiveness of adding compost and green manure in terms of soil carbon accumulation. Using the carbon accumulation amount in 1990 as the initial value, we calculated the change in carbon accumulation over a 25-year period when (1) annually adding compost equivalent to 1.0 t-C/ha to all rice paddies and 1.5 t-C/ha to farmland other than paddies (compost scenario), (2) planting barley as an off-season crop on all rice paddies and adding the barley crop residue equivalent to 0.7 t-C/ha annually as green manure (rice paddy double-cropping scenario), and (3) using a combination of both. We assessed the effectiveness of soil carbon accumulation by the difference between these techniques and when nothing was added (no added organic matter scenario). Compared with the no added organic matter scenario, the compost scenario had an extra 32 million t-C, and the double-cropping scenario had an extra 11 million t-C.

- (ii) Determining the wide-area nitrogen cycle and environmental burden

We estimated the wide-area nitrogen balance and the impacts on the aquatic environment due to crop production, not only for food but also for biofuels, for five Southeast Asian countries (Indonesia, Myanmar, the Philippines, Thailand, and Vietnam) which have prospects for spare production capacity by increasing grain yield per unit area. To do this, we calculated surplus farmland under scenarios for grain yield change per unit area and food demand, and we estimated the amounts of biofuels that could be produced and the nitrogen burden when cultivating three crops (sugar cane, cassava, and oil palms). Our study estimated that in 2030 it would be possible to produce 30–40 million kL of biofuels on about 20% of the area currently planted with grain, but it found that the nitrogen burden would increase to about 8 million t-N. The 2005 nitrogen

burden was 3.4 million t-N, which in 2030 would rise to an estimated 6.1 million t-N if only food were produced. The study predicted that the nitrogen burden would increase by a factor of 1.3–1.5 in order to increase grain yield per unit area to free up surplus farmland and to produce energy crops.

- c. Development of a method to assess watershed water pollution risk based on an understanding of the dynamics of nutrient outflows from the pedosphere to bodies of water

- (i) Assessing the amount of nitrate nitrogen removed through denitrification in the shallow groundwater flow process

Utilizing an analysis method devised in 2007 that uses the stable isotope ratio of nitrogen in nitrate ions ( $\text{NO}_3^-$ ) and oxygen, we estimated the  $\text{NO}_3^-$  removal rate by denitrification at various locations in a topographically linked system (the watershed of Ishioka City, Ibaraki Prefecture) that comprises upland fields, orchards, and lowland rice paddies. The initial (pre-denitrification)  $\text{NO}_3^-$  concentration was determined from the removal rate and the shallow groundwater  $\text{NO}_3^-$  concentration, and the watershed's annual denitrification amount was estimated. We estimated that almost all of the  $\text{NO}_3^-$  in the watershed is formed by the nitrification of  $\text{NH}_4^+$  from chemical fertilizers. We also estimated that the  $\text{NO}_3^-$  removal rate is in the range of 10–70%, and that the influx concentration of  $\text{NO}_3^-$  into groundwater is 0.6–95 mg-N  $\text{L}^{-1}$ . Based on these results, it was estimated that in the areas of the watershed studied (5.0 ha with study wells) the  $\text{NO}_3^-$  groundwater load is 85.1 kg-N $\cdot$ ha $^{-1}\cdot$ y $^{-1}$ , of which 22.8 kg-N $\cdot$ ha $^{-1}\cdot$ y $^{-1}$  is denitrified in groundwater and removed.

- (ii) Estimating watershed phosphorus outflows due to surface runoff and flows through subsoil

We used a slope lysimeter to measure the phosphorus concentration in surface runoff water and the differences in runoff amount depending on soil. The runoff rate (=runoff amount/rainfall amount) from andosols was small at an average 0.3%, and both the runoff phosphorus concentration and phosphorus load were less than 1/10 those of yellow soil. Although the addition of compost to yellow soil reduced runoff amount, the concentration of runoff phosphorus tended to increase.

A rainfall infiltration experiment using a column found that the concentration of dissolved phosphorus in water discharged from the column was at most 0.01 mg-P  $\text{L}^{-1}$ , and that at least 90% of the phosphorus in water discharged from the lower part of the column is particulate phosphorus. Adding compost did not necessarily increase phosphorus surface runoff. due to improvement of soil's water permeability and reduction of dispersion

- (iii) Assessing the risk of groundwater and surface water pollution by nutrients on the watershed level

We analyzed the relationship between the estimated watershed nitrogen load based on cropping record and the groundwater nitrate nitrogen concentration. Analysis results for Kumamoto and Tochigi prefectures did not find good correspondence between the nitrogen potential concentration (NPC) calculated with 1- or 5-km grid cells and observed groundwater nitrate nitrogen concentration in the studied area. Groundwater nitrate nitrogen concentration had a log-normal distribution, and as NPC increased, there was an increasing probability that it would exceed the voluntary standard ( $10 \text{ mg L}^{-1}$ ). The relationship between NPC and the probability of groundwater nitrate nitrogen concentration exceeding  $10 \text{ mg L}^{-1}$  was different in the two prefectures, which suggested the involvement of regional factors. We examined soil groups as a factor influencing the decline in concentration of nitrate nitrogen in shallow groundwater in a riparian zone, and attempted an area estimate of nitrate nitrogen concentration in shallow groundwater in a watershed (Sakura River upstream area) including the riparian zone.

After further improvements to these methods, we intend to create maps for assessing the environmental vulnerability to water pollution by nitrate nitrogen and maps for assessing the risk of water pollution.

## **C Basic research to help elucidate the functions of agro-ecosystems**

### **1) Agriculture-related long-term environmental monitoring**

#### **(1) Long-term monitoring of agro-environments and the development of simple and highly accurate measurement methods**

##### **Medium-term plan:**

For the early detection of changes in agro-environment resources, we will perform long-term monitoring of the physical environment, which is the baseline of agro-ecosystems, and the fluxes of greenhouse gases including carbon dioxide and methane, as well as  $^{137}\text{Cs}$ ,  $^{210}\text{Pb}$ , and other isotopes in crops and soil. Also, we will develop simple and highly accurate measurement methods for the analysis and monitoring of organic arsenic compounds and other trace substances in the environment, including in crops.

##### **Research Overview:**

- a. Enhancing the technologies to detect and monitor changes in the physical environment and gas fluxes in relation to global warming

##### **(i) Greenhouse gas flux monitoring**

We continued gas flux observations at rice paddy sites in four locations with different meteorological

conditions and planting systems (single cropping rice in Mase, Tsukuba City; double cropping of rice and barley in Okayama; double cropping of rice and wheat in Jiangdu, Jiangsu Province, China; and double cropping of rice in Mymensingh, Bangladesh). From data gathered so far, we found that with single-cropped rice (Mase),  $\text{CO}_2$  corresponding to more than half of the net ecosystem production (NEP) during the cultivation period is emitted as ecosystem respiration (RE) during the non-cultivation period (the non-growth period is equivalent to soil respiration), which accounts for two-thirds of the year. In the paddies double cropped with rice and barley or wheat (Okayama and Jiangdu), we found that  $\text{CO}_2$  is absorbed nearly all year because the non-cropping period is short. In this way, the characteristics of each site (planting system) were determined. A comparison of the NEPs of various world ecosystems with the NEPs at the study sites found that while Mase ranks at about the middle of various ecosystem types, the other three rice paddy sites were distributed within the upper 10%. In other words, the ecosystems of rice paddies double cropped with rice and wheat/barley and paddies double cropped with rice have large NEPs, even in a worldwide comparison.

In four pasture sites in Japan (Nakashibetsu, Shizunai, Nasu-Shiobara, and Kobayashi) we gathered observation data on greenhouse gas flux in the fourth year of the monitoring. For the years 2005 to 2007, although the NEPs of all sites in the sections where compost had not been added varied greatly depending on the year ( $150\text{--}360 \text{ g-C}\cdot\text{m}^{-2}\cdot\text{year}^{-1}$ ), the 3-year average came between the values of the rice single-cropped paddies and the rice and wheat/barley double-cropped paddies. In general, there was a tendency for NEP to increase along with the length of the pasture growth period.

##### **(ii) Physical environment monitoring**

At the alpine vegetation line on the Tibetan Plateau, for which there are very few observation data, we gathered detailed meteorological data and analyzed precipitation data. We found that precipitation is concentrated in the monsoon season (June through September), and that the highest amounts fall in the mid-slope region around 5,100 m. It is known that the maximum rainfall occurs in mid-slope areas because water drops form due to adiabatic expansion of air currents along the slopes during the day, and we observed the same phenomenon on the Tibetan Plateau at elevations over 4,000 m.

- b. Long-term monitoring of radioactive substances and other substances in crops and soil, and development of simple and highly accurate methods to measure trace chemicals

- (i) Long-term monitoring of radioactive substances in agro-environments

We analyzed the radioactive strontium ( $^{90}\text{Sr}$ ) and radioactive cesium ( $^{137}\text{Cs}$ ) in the soils and in the rice and wheat produced in 2007 on observation fields throughout Japan. The analysis found that the levels were the same as those for grains produced in 2006, and discerned no marked changes in concentrations. We also analyzed the radioactive substances in spinach, garlic chives, and tea from our institute's environmental radiation study fields. The analysis found for uranium series nuclides that  $^{214}\text{Bi}$  was below  $0.1 \text{ Bq}\cdot\text{kg}^{-1}$ , and that  $^{210}\text{Pb}$  was about  $3 \text{ Bq}\cdot\text{kg}^{-1}$  in leaves of tea, and below  $1 \text{ Bq}\cdot\text{kg}^{-1}$  in the other two crops.  $^{131}\text{I}$ ,  $^{134}\text{Cs}$ , and  $^{137}\text{Cs}$  were not detected. Spinach, which has been continually measured, had the same levels as in 2006, and no marked changes in concentrations were observed.

To prepare for rebuilding the system for analyzing radioactivity in livestock products, we began cross-checking analyses of  $^{90}\text{Sr}$  in milk samples with the National Institute of Livestock and Grassland Science.

- (ii) Development of simple and highly accurate analysis methods for trace chemicals in agro-environments

Organic arsenic compounds are found in agro-environments in various forms, and because each has different toxicity and characteristics, we made enhancements to form-specific analysis methods. We studied an analysis method using liquid chromatography as pre-processing for ICP-MS, and identified the unknown organic arsenic compounds in reduced soil as dimethylphenylarsine sulfide and methylphenylarsine sulfide.

As part of an effort to develop a multi-component analysis method for persistent organic pollutants (POPs), a manual for a method to analyze POPs in river systems was prepared under joint international research (Japan, South Korea, Germany, and the United States). This analysis method, whose development was conducted in line with the standard operations procedural manual, has specific ways of analyzing POPs with multiple components (22 kinds).

## **2) Gathering, preserving, computerizing, and using environmental resources**

### **(1) Constructing an agro-environmental resource inventory and developing a way to use it**

#### ***Medium-term plan:***

To perform an overall assessment of the agro-environment, we will develop technologies to analyze data from microwave measurements, from the high temporal resolution satellite sensor MODIS, and from other remote sensing sources and we will use a geographical information system (GIS) and other tools to

develop new methods to ascertain the state of agricultural land use and develop indicators for the habitats of organisms. Additionally, we will develop a method of linking individual databases by using GIS as a common platform and a system to register and gather new information, and in that way contribute to the formulation of agro-environmental indicators. Also, along with expanding and enhancing individual databases of environmental resources, we will publicly release a proposed soil classification scheme that includes functional assessment of deep soils, and build an integrated soil database of cultivated and non-cultivated land. Further, for the efficient use of inventory data and other data, we will develop a basic statistical method and a method of visualizing that statistical method's results. We will also lend assistance as a sub-bank to the gene bank project operated by the National Institute of Agrobiological Sciences.

#### ***Research Overview:***

- a. Computerization and use of agro-environmental resources using remote sensing and a geographical information system

- (i) Development of a method to ascertain the wide-area state of agricultural land use by means of remote sensing analysis technologies

Using data from the high temporal resolution satellite sensor MODIS, we prepared general basic data sets for the Indochinese Peninsula region, the Heilongjiang Province region in China, and the Japan/Korean Peninsula region (2004–2007; 500 m and 250 m resolution), and we built a time series data set (6-day intervals) of various indicators for assessing the state of the Earth's surface. For the Indochinese Peninsula, we used a classification system that employs the pattern of changes over time of each pixel to create an agricultural land use classification map that focuses on rice paddy areas. We confirmed the map's soundness by comparing it with land use classification maps resulting from field surveys, and with detailed classification results of the Mekong Delta. These are useful for the two-dimensional comprehension of wide-area information on land use and cropping, and the data sets are already being put to uses such as Japan–Korea international joint research, and wide-area productivity assessments by the National Agriculture and Food Research Organization.

Analyses of the latest satellite images were used to determine the most recent state of land use change and ecosystem carbon stocks in the swidden agriculture zone of the Lao PDR. This found that swiddening land area has still not decreased. Through a detailed comparison of land use and cropping scenarios, we found a land use and cropping system that is promising in view of ecosystem carbon stock and profitability. There is great interest in

these research achievements among international development and environmental agencies, such as Germany's Agency for Technical Cooperation (GTZ).

- (ii) Developing systems to ascertain the state of agricultural land use using GIS analysis techniques (Achievement ready for deployment: Development of the Historical Agro-Environmental Browsing System (HABS))

We had been working on converting maps produced in the early Meiji period (the Rapid Survey Maps) to GIS data for the purpose of analyzing past land use. We have completed this conversion and have published the results on the internet. It is being used by universities, research institutes, and the like as base maps for biological surveys in research on rice paddy biodiversity and for other such applications. There are also expectations that it can be used, for example, as a basic source material for activities to preserve community-based forests.

Based on analyses using the Ministry of Environment's Existing Vegetation Maps and agricultural statistical data, we found that small rice paddies (less than about 100 m wide) found in narrow valleys are, although small in area, widely distributed throughout the Kanto region. They tend to run long distances along the contour and abut wooded areas, and as such, they play a vital role in maintaining the narrow valley rice paddy landscapes. Using the area of "abandoned farmland (paddies)" and the area of small rice paddies as indicators, we created a distribution map showing places where there is a danger that the valley rice paddy landscapes will disappear from villages in the Kanto region.

- b. Constructing an integrated inventory and developing a method to use it

- (i) Expansion and enhancement of individual inventories, and building an integrated agro-environmental inventory

In our work on an agro-environmental inventory system that displays the contents of various individual agro-environmental resource databases, such as those of soil and insects, over a map in a unified manner with the GIS as a common platform, we improved operability and the screen display, and gave access to everyone in NIAES. Security was improved, a manual was written on how to run and administer the system, and we are working on opening it to the general public. We also made other expansions and enhancements, such as adding new data to the inventory related to soil, microorganisms, and insects.

- (ii) Formulating a proposed comprehensive soil classification scheme

To formulate a proposed comprehensive soil

classification scheme, we performed surveys at six locations in Japan to create representative soil profiles, wrote descriptions of the profile forms, performed analyses of their general physical and chemical natures, and analyzed clay minerals. We also performed a soil survey that included forest soils for 1:50,000 soil sheet maps, compared the third revised edition of the farmland soil classification scheme (third draft) with the second proposal for a unified Japanese soil classification system by the Japanese Society of Pedology to determine their correspondence and differences, and then created soil maps that include forest land. To serve as shared data for creating the proposed classification scheme, we made national farmland soil maps for 1992, prepared standard data for creating upland field soil series and standard data for creating rice paddy soil series, and partially released them.

- (iii) Developing indicators for soil erosion, pesticides, and other agro-environment risks

We compared potential soil erosion amounts and data from nine locations where measurements had been made in the past, and found a significant correlation between the two ( $r^2 = 0.63$ ,  $P < 0.05$ ). On the other hand, we did not find a clear relationship between the average potential soil erosion amounts compiled by watershed and the 5-year average suspended solid concentrations at water quality observation points in each watershed.

We identified factors (exposure and toxicity) thought to be important as environmental risk indicators for pesticides used in places other than rice paddies (such as upland fields and orchards), and we established an assessment environment (a model watershed of 100 km<sup>2</sup> with 750 ha of upland fields and rivers) for calculating concentrations in river water.

- (iv) Developing basic statistical methods

We made improvements to the software we had been developing through the previous year for calculating phylogenetic diversity so that it can calculate not only the existing phylogenetic measure but also the new measure, the "Gromov metric." We used this software to actually calculate the phylogenetic diversity of a certain monophyletic group for DNA base sequence data.

- (v) Gene bank project

In accordance with the FY 2008 project plan, we registered 15 new MAFF microorganism strains including tomato black leaf mold *Pseudocercospora fuligena* and the Japanese stewartia *Stewartia pseudo camellia* mold *Colletotrichum acutatum*, and we performed studies of the microorganisms' characteristics covering 217 items. With regard to insects, in addition to what is in the project plan we introduced one new line (not damaging to brown rice planthopper *Nilaparvata*

## *Special Research Projects*

### **1. Selection of Functional Biodiversity Indicators and Development of Assessment Methods**

To promote sustainable agriculture in which agricultural productivity is compatible with conservation of biodiversity, environment-friendly (environment-preserving) farming systems have been developed and propagated in Japan. Little is known, however, about the effect of these farming systems on biodiversity in agro-ecosystems. Over the period from FY 2008–2012, NIAES has been conducting a research project on “Selection of functional biodiversity indicators and development of assessment methods” with the support of a Grant-in-Aid from the Ministry of Agriculture, Forestry and Fisheries of Japan. The objectives of the project are to develop indicators that can help evaluate the effects of environment-friendly farming on the conservation and enhancement of agrobiodiversity. The project focuses on the investigation of indicator organisms beneficial to agriculture, such as predators and parasitoids of agricultural pests. Essentially, the research compares the species and abundance of organisms in fields where environment-friendly farming and conventional farming are practiced; organisms abundant in the environment-friendly farming systems are selected as candidate indicator organisms.

The project includes the following two research subjects:

#### **(1) Selection of organisms as indicators of functional agrobiodiversity**

We expect that some indicator organisms will be vulnerable to differences in management (e.g., pesticide application) in each cropping field; thus their abundance will differ among plots. Other indicator organisms, however, may not be vulnerable to changes in management, but their populations may fluctuate at the landscape level because they move over wide areas or use different sites in a landscape as habitats. Hence, agrobiodiversity is to be analyzed at a crop field level and at a landscape level.

#### **(2) Development of simple methods of evaluating agrobiodiversity by using indicator organisms**

The evaluation methods that are developed need to be simple so that they can be applied at the farm level. The research will therefore establish simple methods of identifying indicator organisms and will develop efficient ways of monitoring these organisms. In addition, we will develop a system for applying the results of research subject (1) to the entire agro-ecosystem in order to predict changes in agrobiodiversity in Japan.

### **2. Research project for developing risk mitigation technologies for POPs in vegetables**

The concept of risk analysis has only recently been introduced to domestic food safety administration in Japan. It is therefore essential that we build a scientific and technical basis for risk mitigation of farm/marine products and livestock.

Twelve chemicals were selected as persistent organic pollutants (POPs) by the Stockholm Convention in 2001, and nine chemicals have been newly assigned in 2009. Fourteen of these POPs have been used as pesticides. They include dieldrin and heptachlor, which were recently detected in fruits of cucumber and pumpkin in amounts exceeding Japanese maximum residue levels (MRLs), even though their use was banned in the early 1970s.

We have studied these chemicals in past research projects, but we still have limited practical information on the risk analysis of POP residues in agricultural products.

This project has the following three study components, targeting mainly vegetables: (1) development of risk assessment technologies for POP exposure in the food chain; (2) development of risk management technologies for POP contamination; and (3) development of basal technologies for risk assessment and management of POPs.

The goal of the project is to develop and validate preventive (minimization of absorption by crops) and curative (remediation of soils) technologies for those POPs that were used domestically as pesticides in the past. We aim to keep POPs residues in crops at levels lower than the MRLs. By using these newly developed methods, together with good field practices, we hope to achieve risk mitigation of POPs. This project is divided into three large issues, composed by 21 action plans as a whole, and they are conducted by about 22 researchers from 13 organizations.

This project is one in a sequence of seven known collectively as the “Research project for ensuring food safety from farm to table”. The objective of the sequential project is to clarify the behavior of significant hazards, such as arsenic and cadmium (as described below), POPs, mycotoxins (nivalenol, deoxynivalenol), and pathogenic microbes, in order to develop simple and quick detection methods and methods of mitigating exposure risk in the journey of food products from arable lands and fishing waters to the table. The project began in FY 2008 and is scheduled to end in FY 2012. It is supported by the Ministry of Agriculture, Forestry and

Fisheries of Japan. The participants are not only NIAES, but also other non-designated independent administrative institutions, universities, prefectural agricultural experimental stations, and the private sector.

### 3. Development of risk mitigation technologies for arsenic and cadmium in crops

Arsenic is considered one of the most important toxic elements found in the environment because of its potential risk to human health. Food is a potentially important source of dietary arsenic intake. Rice (*Oryza sativa*) accumulates the highest amount of arsenic of all grain crops, largely because of the high plant availability of arsenic under reduced soil conditions. Rice is one of the world's major staple food crops, with daily intakes of up to 0.5 kg per head in Asian countries and 0.17 kg in Japan in 2002. Consequently, rice is a potentially major source of dietary arsenic for much of the world's population. The Ministry of Agriculture, Forestry and Fisheries of Japan (MAFF) has analyzed arsenic contents of staple crops in Japan. The arsenic concentration in brown rice ranged from 0.04 to 0.33 mg kg<sup>-1</sup> (average 0.16 mg kg<sup>-1</sup>;  $n = 199$ ) in 2003. The averages in other crops were wheat, 0.008, soybean, 0.005, and spinach 0.010 mg kg<sup>-1</sup>. Chemical speciation of arsenic influences its toxicity to humans. Inorganic arsenic is generally considered to be more toxic than methylated arsenic compounds

Recently, the CODEX Alimentarius Commission of the FAO and the WHO proposed a new international standard for Cd concentrations in a variety of staple foodstuffs; for fruiting vegetables this level is 0.05 mg Cd kg<sup>-1</sup> and for root vegetables it is 0.10 mg Cd kg<sup>-1</sup>. In a field and market basket survey conducted by the MAFF in 1998–2001 in Japan, about 7% of 381 samples of eggplant (*Solanum melongena*), 22% of 165 samples of okra (*Abelmoschus esculentus*), and 10% of 302 samples of taro (*Colocasia esculenta*) contained cadmium at concentrations above these limits. We therefore urgently need to develop technologies to suppress cadmium absorption by crops.

This project has the following two study components: (1) development of risk mitigation technologies for arsenic in paddy rice; and (2) development of risk mitigation technologies for cadmium in upland crops. The project began in FY 2008 and is scheduled to end in FY 2012. This research project is supported by a grant from the MAFF and conducted by 27 organizations carrying 54 themes (Research project for ensuring food safety from farm to table).

### 4. Assessment and extension of technologies for mitigating greenhouse gas emissions from agricultural soil

Greenhouse gas (GHG) emissions from Japan's agricultural sector are estimated to be 27 million tons-CO<sub>2</sub>. CH<sub>4</sub> emissions from rice paddies account for 21% of total agricultural sector emissions, and N<sub>2</sub>O emissions associated with N-fertilizer use account for 9.5%. Japan did not select "cropland management" in the first commitment period of the Kyoto Protocol, but the Japanese government will choose "management to reduce GHG emissions from agricultural soil" in the second commitment period of the Kyoto Protocol. Therefore, the government is expected to promote the introduction of farming practices that reduce GHG emissions by the agricultural sector.

A research project, entitled "Assessment and extension of technologies for mitigating greenhouse gas emissions from agricultural soil" focuses on evaluation of the effects of reducing CH<sub>4</sub> emissions by implementing new water management practices (longer periods of mid-season drainage) in paddy fields and by carbon sequestration through intensive soil management based on organic matter application. The project is sponsored by the Agricultural Production Bureau of MAFF, and NIAES is supervising the project activities of the two national institutes, 47 prefectural institutes, and one incorporated foundation. The research project runs from FY 2008–2012.

In this project, we will estimate the CH<sub>4</sub>-emission-reduction effects of water management at nine experimental paddy field sites in Japan. We will also confirm soil carbon sequestration factors for every region, soil type, and type of organic matter (e.g., plant residues, manure composts, and charcoals) at 68 sites in paddy fields, upland fields, greenhouses, and orchards. Moreover, we will evaluate soil carbon content to 30 cm depth at 3200 sites in farmers' fields by soil survey and will interview farmers about their soil management.

### 5. Assessment of Risks to Aquatic Ecosystems by Zinc and Other Heavy Metals Originating in Farming Areas

In recent years, to conserve aquatic organisms, an environmental quality standard for water and an effluent standard have been set for zinc. In rural districts there are concerns that emissions of zinc and other heavy metals from not only graywater but also buildings housing pigs and other livestock are sources of ecosystem pollution. The reason is that, because metals such as zinc and copper are essential elements for mammals, considerable

amounts are used daily by humans and the livestock industry. In the livestock industry especially, these elements are added in large amounts to pig and chicken feed to promote growth, and they are present in high concentrations in livestock waste. However, nearly all research so far deals with efforts related to human health risks, such as accumulation of heavy metals in farmland soil, and absorption and transport into agricultural crops. One sees no research at all from the perspective of ecological toxicity or ecosystem conservation, and hardly anything is known about the burden on river systems by zinc and other heavy metals, or their dynamics.

Accordingly, through this research we will determine the flow in rural watersheds of zinc and other heavy metals whose ecological toxicity is perceived as a problem, and create a dynamics model. Additionally, we will develop a technology for treatment of the zinc and other heavy metals in effluent from livestock buildings, and throw light on the water remediation function of wetlands. Then we will integrate these and assess the risk to aquatic ecosystems by zinc and other heavy metals arising in farming areas. Specifically, we will investigate the following items and provide basic knowledge on aquatic ecosystems in rural areas.

1) Determining the state and intensity of emissions of zinc and other heavy metals from livestock farms

Mainly with regard to pig farming, in which high amounts of zinc and copper are added to feed, we will investigate the concentrations of zinc and other metals in effluent, find the amount of runoff through interviews and other means, calculate the runoff loads of zinc and other heavy metals in river systems, and find the load intensity with reasonable accuracy.

2) Determining the state of emissions and runoff dynamics of zinc and other heavy metals in agricultural watersheds

In agricultural watersheds where many livestock farms are found, we will determine the load of zinc and other heavy metals from non point sources such as fields on which composted livestock waste has been applied. Additionally, we will determine the process by which the load of zinc and other heavy metals emitted from non point sources and livestock operations arrive in public waters from emission channels via streams. Further, we will carry out a study of aquatic biota in the rivers of rural watersheds, and will analyze changes in biota and its relationship to the loading from livestock farming and river water quality.

3) Developing a technology to lower the concentration of zinc in livestock industry wastewater

We will investigate the wastewater treatment processes operating at pig farms and other facilities, and will verify the effectiveness with which zinc

concentration in wastewater is lowered by reducing suspended matter. Additionally, with regard to those treatment processes for which reduction of zinc concentration is still insufficient, we will develop a method of lowering wastewater zinc concentration that can be used in livestock operations in order to stably keep the zinc concentration below the effluent standard.

4) Determining the function of wetlands in reducing loads of zinc and other heavy metals

We will quantify the effectiveness of wetlands and vegetation zones in reducing zinc, throw light on remediation processes such as soil adsorption and plant absorption, and explore ways of enhancing load reduction by artificial wetlands.

5) Assessing risks to aquatic ecosystems

For agricultural watersheds, we will create a GIS (Geographical Information System) model that expresses the emissions and dynamics of zinc and other heavy metals, and then we will assess the risk to public waters and ecosystems.

This project will be carried out in cooperation with National Institute for Agro-Environmental Sciences, National Agriculture and Food Research Organization/National Institute of Livestock and Grassland Science, Aichi Agricultural Research Center, and Nagoya Women's University; it is supported by the Ministry of the Environment from FY2008 to 2011.

## **6. Predicting acidification and nitrogen leaching in East Asian ecosystems with a catchment-scale model**

Emission rates of acidic substances in East Asia are likely to increase over the next several decades owing to the expected increases in energy and food demand. Natural ecosystems in the area are thus likely to be chronically exposed to atmospheric acidic deposition. The effects of chronic input of acidic substances on tropical ecosystems are not well known, because investigations of the effects in these ecosystems are quite limited compared with those in temperate and boreal areas. The objectives of our 3-year research project in collaboration with The University of Tokyo and the Acid Deposition and Oxidant Research Center are therefore to 1) clarify the changes in soil and stream water chemistry, such as acidification and nitrogen leaching, caused by atmospheric deposition in tropical ecosystems; and 2) predict the temporal trends in these changes under future emission scenarios.

Small catchment study sites have been established in a tropical rain forest in the Danum Valley area of Malaysia and in a tropical dry evergreen forest at Sakaerat Silvicultural Research Station, Thailand, and

catchment-scale monitoring of atmospheric depositions and soil and stream-water chemistry has begun in order to evaluate material cycles. Numerical models of soil chemistry changes and the nitrogen cycle are being developed on the basis of these data. The results of the first year of research indicate that there are prominent seasonal changes in soil and stream-water chemistry in response to the periodic dry and rainy seasons in Sakaerat. The model also shows similar changes in soil pH and in the concentrations of some soil elements. Various GIS data and statistical data on, for example, agriculture and households in the divisions or provinces containing the study sites are being collected, and nitrogen budgets are being evaluated to estimate the contribution of agriculture to acid deposition and the future trend in this contribution until 2030.

The research project has been conducted during FY2008-2010 period supported by the Global Environment Research Fund (C-082) of Japan's Ministry of the Environment.

### **7. Study of Changes in Chemical Form and Plant Uptake of Aromatic Arsenicals in Agricultural Soils**

In 2002, the inhabitants of the Kizaki area of the town of Kamisu in Ibaraki Prefecture exhibited uncommon clinical symptoms of the central nervous system. In 2003, diphenylarsinic acid (DPAA), and phenylarsonic acid (PAA) were detected in groundwater drunk by the inhabitants. In 2004, DPAA was detected in groundwater used for irrigation in Kamisu. DPAA and methylphenylarsinic acid (MPAA) were also detected in harvested paddy rice. There is therefore a need to measure the extent of pollution of the soil, water, and farm products, but there has been little research on plant uptake of aromatic arsenicals from soil.

We investigated methods of quantifying aromatic arsenicals in soil and in rice, which is widely cultivated

in the Kizaki area. DPAA, PAA, MPAA, dimethylphenylarsine oxide (DMPAO), and methyldiphenylarsine oxide (MDPAO) in soil and rice were extracted, separated by reverse-phase chromatography, and quantified by ICP-MS. For extraction of arsenicals from rice grain and straw, hot trifluoroacetic acid at 2.0 mol/L or hot 68%  $\text{HNO}_3$  gave better extraction efficiency than 50% MeOH. For extraction from soil, hot 68%  $\text{HNO}_3$  gave a better result than 1.0 mol/L  $\text{H}_3\text{PO}_4$  or NaOH. We also investigated the unknown arsenic species formed in the course of incubation of DPAA-amended soil under flooded conditions. The results of HPLC-TOF/MS analysis strongly suggested that some of the unknown species were dimethylphenylarsine sulfide and methyldiphenylarsine sulfide.

In an incubation study, we investigated the biogeochemical changes in DPAA in two types of agricultural soil, Kamisu soil and Tsukuba soil, under flooded or upland conditions. In flooded soils, DPAA can be converted to MDPAO by methylation; some of the DPAA can be converted to PAA, which is subsequently converted through MPAA to DMPAO. The concentration of DPAA in the Kamisu soil clearly decreased after 24 weeks' incubation. In sterilized soils of both types, the DPAA content was close to stable during the 24-week incubation, although small amounts of PAA were produced.

We investigated the uptake by rice of aromatic arsenicals from contaminated soil and from unpolluted soil amended with DPAA, PAA, MPAA, DMPAO, or MDPAO. In the contaminated soil, PAA and MPAA concentrations decreased and the DMPAO concentration increased under flooded conditions; however, their concentrations remained unchanged under upland conditions. DMPAO and MDPAO absorbed by the shoots were retained; MPAA absorbed by the shoots was translocated to the grain more easily than DMPAO and MDPAO.