

# Newsletter

FOR INTERNATIONAL COLLABORATION

2

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Selling fish in Peru (Photo by T. Ishitani)

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# JIRCAS

JAPAN INTERNATIONAL RESEARCH CENTER FOR AGRICULTURAL SCIENCES



# Establishment of JIRCAS Fisheries Division

*Dr. Kunihiko Fukusho*

The Fisheries Division started its activities together with the reorganization of the former Tropical Agriculture Research Center (TARC) into JIRCAS on October 1, 1993. All the researchers were appointed on April 1, 1994 in the following fields:

Dr. Shigeo Hayase (squid ecology), Dr. Koji Nakamura (marine biochemistry), Dr. Motoyuki Hara (fish genetics), Mr. Katsuhisa Tanaka (marine chemistry), Mr. Satoshi Suyama (marine fish ecology), and Mr. Masashi Sekino (freshwater fish culture).

The Fisheries Division is in charge of research on marine and freshwater fisheries resource management, aquaculture, fisheries product processing, which are major themes for international collaboration.

In the field of fisheries sciences, international collaboration and technical transfer had mainly covered aspects relating to fishing technology, development of fishing gear, freshwater fish culture, and fish processing at first. However, mariculture technology and industry have experienced a substantial development in Japan for the past twenty years. Then, the transfer of these techniques was requested by the developing countries. To answer these needs, JICA and other organizations involved in

international collaboration have implemented various research projects on intensive mariculture and obtained fruitful results. Many developing countries which have introduced intensive culture technology from developed countries like Japan have succeeded in promoting mariculture of finfish and prawns for the supply of animal protein, and the development of export industries.

However, due to the insufficient knowledge about the aquatic ecosystems and environmental protection, and to the rapid development of the mariculture industry centered on economic aspects only, a large number of problems have arisen. Therefore, the JIRCAS Fisheries Division plans to implement collaborative research projects for studying the aquatic ecosystems in the developing countries in order to develop techniques for sustainable production of fish, methods of fish culture compatible with the preservation of the environment, and methods for efficient utilization of fisheries products.

Although the Division has a small number of researchers, it is being supported by nearly 430 scientists affiliated to the nine fisheries institutes of the Fisheries Agency, and will also implement joint research programs with other Divisions of JIRCAS.



*Director, Fisheries Division  
Aquaculture Biologist for Marine Finfish  
Born in Pusan, Korea in 1942. Graduated from  
Tokyo University of Fisheries in 1965.  
Completed master course of the TUF in 1967.  
Visiting researcher at Department of Fisheries,  
Faculty of Agriculture, Kyoto University (1967-  
1969). Since then, he has been engaged in studies  
on fry production of marine finfish carried out at  
the Mariculture Laboratory, Nagasaki  
Prefectural Institute of Fisheries (1969-1979).  
Ph. D., Faculty of Agriculture, Kyoto  
University, 1979. He moved to the National  
Research Institute of Aquaculture and carried out  
research on marine finfish breeding as Chief of  
the Genetic Breeding Section (1979-1989).  
During this period, he was dispatched as short-  
term expert to Indonesia, Singapore, Thailand  
and China for participating in international  
collaboration projects on marine finfish culture  
sponsored by JICA, FAO, and IDRC. He was  
appointed Chief of the Research Planning Section  
of the NRA (1989-1990). Thereafter, he was  
appointed Director of the Nikko Laboratory,  
NRIA. He joined JIRCAS in October, 1993, to  
assume the position of the Fisheries Division.*



*Photo: Aquaculture area in Java, Indonesia.*

## **JIRCAS International Symposium 1994 Satellite Symposium of the XXIVth International Horticultural Congress**

Fruit Production in the Tropics and Sub-Tropics, Kyoto, Japan, August 22-23, 1994

### **Keynote Address**

1. Role of Fruit Production in the Tropics and Sub-Tropics

*H. Kitagawa, Kagawa Univ., Japan*

### **Technical Reports**

#### **Session 1. Collection and Utilization of Tropical and Sub-Tropical Fruit Tree Genetic Resources**

1. Role of IPGRI in the Collection, Conservation and Utilization of Tropical and Sub-Tropical Fruit Tree Genetic Resources

*R.K. Arora and K.A. Okada, IPGRI*

2. Collection and Utilization of Tropical and

Sub-Tropical Fruit Tree Genetic Resources for Breeding in India

*P.L. Agarwal, ICAR, India*

3. Collection and Evaluation of Unused Tropical and Sub-Tropical Fruit Tree Genetic Resources in Malaysia

*W.K. Choo, Univ. Pertanian Malaysia*

4. Utilization of Genetic Resources of Tropical and Sub-Tropical Fruit Trees in Japan

*H. Daito, JIRCAS, Japan*

#### **Session 2. Pest Control of Tropical and Sub-Tropical Fruits**

1. Virus or Virus-Like Diseases of Citrus in Tropical and Sub-Tropical Zones

*M. Koizumi, FTRS, Japan*

2. Prevention and Control of Harmful Insects on Tropical and Sub-Tropical Fruits

*N.N. Kiem, VEGETEXCO, Vietnam*

#### **Session 3. Storage and Postharvest Physiology of Tropical Fruits**

1. Storage of Tropical Fruits in Thailand

*S. Ketsa, Kasetsart Univ.*

2. Postharvest Physiology of Tropical Fruits in the Philippines

*Ma. C. C. Lizada, Univ. the Philippines*

3. Postharvest Physiology of Avocado

*H. Inoue, Nihon Univ., Japan*

#### **Session 4. Cultivation of Temperate Zone Fruits in the Tropics and Sub-Tropics**

1. Cultivation of Grape Vine in Brazil

*M.M. Terra, Inst. Agro. Secao de Viticultura*

2. Cultivation of Grape Vine in Indonesia

*M. Winarno, Solok Res. Institute*

3. Breeding of Temperate Zone Fruits Required Low Chilling in Australia

*S.R. Sykes, CSIRO Div. Hort., Australia*



# JIRCAS Annual Research Promotion Meeting 1993

Norio Nakaya

Director, Planning and Coordination Division

With the purpose of reviewing various activities relating to the Tropical Agriculture Research Center (TARC) and the Japan International Research Center for Agricultural Sciences (JIRCAS) conducted in 1993 (fiscal year), the Annual Research Promotion Meeting was held at the JIRCAS Headquarters in Tsukuba on 8 February 1994. There were 51 participants including representatives from the Agriculture, Forestry and Fisheries Research Council Secretariat, the Fisheries Agency, the Forestry Agency, National Agriculture Research Center and other research institutes under the Ministry of Agriculture, Forestry and Fisheries. TARC was reorganized into JIRCAS last October for promoting research activities pertaining to wider research fields including fisheries science. The geographical zone covered by the Center was also extended to the tropical, sub-tropical, temperate and cool zone. The Director General of JIRCAS, Dr. Keiji Kainuma, briefly introduced the role and functions of the Center at the opening of the meeting, and asked all the participants

to extend their cooperation to the new Center.

During the meeting, TARC/JIRCAS activities were reviewed in detail through the exchange of information and the projects and proposals were evaluated. Among the research achievements, 11 subjects deemed particularly important are listed in the Table. Apart from the research findings, the outcome of discussions held during various seminars organized at JIRCAS was outlined. These seminars covered topics relating to fisheries science, eco-systems in brackish water and insects in Africa. The results of the 27th International Symposium, "Plant Genetic Resource Management in the Tropics", were also reported. The 28th Symposium on "Fruit Production in the Tropics and Sub-Tropics" will be held in Kyoto during the period of August 23-24, 1994. Recent progress in the joint projects between IRRI and ICRISAT/Government of Japan was outlined.

Special emphasis of the discussions was placed on studies for the development of

postharvest technology in the developing countries. These studies which had not been carried out as a major theme by TARC, will be promoted more intensively by JIRCAS in collaboration with the counterpart institutes.

The successful operation of the JIRCAS Visiting Research Fellowship Program was conveyed by the Director of the Okinawa Subtropical Station. Ten scientists from 9 countries (Bangladesh, Brazil, India, Indonesia, Malaysia, Nigeria, Philippines, Thailand, Vietnam) are jointly conducting studies involving 4 themes such as environmental control, heat-tolerance, salt-tolerance and genetic resources in collaboration with JIRCAS researchers.

The initiation of 2 new projects under the special budget was approved, i.e. 1) Agro-technical Studies for Rural Development in the Mekong Delta Area (Vietnam, 1994-98) and 2) Development of Technology for Aquaculture using Fertilizer in South-East Asia, (Thailand, 1994-98).

## Food Problem and the Role of Agricultural Technology Development in LDCs:

### Seminar organized within the framework of the 1994 Research Promotion Conference of JIRCAS

Keiji Ohga

JIRCAS has added socio-economic studies in connection with research on technology development as a new research field when TARC was reorganized into JIRCAS on Oct. 1st, 1993. In order to define the priorities for such studies, JIRCAS organized a seminar under the theme of "Food Problem and the Role of Agricultural Technology Development in LDCs" as a part of the 1994 Research Promotion Conference of JIRCAS.

Three agricultural economists and a former director general of TARC made presentations on related topics. The first report entitled: "Long Term World Food Projection" was presented by Mr. Keiji Ohga from JIRCAS, the second, "The Implication of Global Environmental Problems on World Food Situation" by Dr. Mitsuhiro Nakagawa from the National Research Institute of Agricultural Economics (NRIAE) and the third, "Food Policy and Needs for Agricultural Development in Asian Developing Countries" by Mr. Kunio Tsubota from NRIAE. The fourth report entitled "Appropriate Technology in Agricultural Development" was presented by Dr. Shiro Okabe currently working at the Research Center for Food and Agricultural Policy.

The chairman, Dr. Yoshikazu Ohno

from JIRCAS, summarized the discussions in the light of the new agenda for JIRCAS as follows.

The first task is to build a new "World Food and Agriculture Model" incorporating the relation between global environmental problems and food production in order to analyse the effects of environmental changes such as global warming on crop growth and alternative food policy choices. The second task is to study the food and agricultural situation in developing countries and characterize the countries in the light of their resource endowment and development stages. The third task is to study the development of rural zones in marginal areas such as mountainous areas and areas undergoing desertification. The fourth task is to work out Japan's international strategy for research cooperation and development assistance in the field of agriculture, forestry and fisheries, based on Japan's experiences in agricultural development.

**Table 1. Major Research Achievements in 1993**

No.	Subject	(Place of study)
1.	User's Manual for Tropical Agriculture Research Information Database (TROPIS) Systems	Japan
2.	Plant Response to Drought Stress and Gene Expression	Japan
3.	Detection of Mycoplasma-like Organisms in Southeast Asia using DNA Probes	Thailand
4.	New Rice Varieties "Dian Jing 34, 35, 36, 37, Registered in Yunnan Province	China
5.	Three New Cucumber Varieties of High Quality with Heat Tolerance and Disease Resistance for Sub-tropical Areas	China
6.	Genetic Diversification of <i>Bradyrhizobium</i> Rhizobia in Soil	Thailand
7.	Soil Properties in the Desert Region	China
8.	Incidence and Agrotechnical Control of Virus Diseases on Vegetables	Thailand
9.	Isolation and Characterization of Cellulolytic Bacteria from the Rumen of Tropical Ruminants	Malaysia
10.	Development of Breeding Colony of Lesser Malayan Mouse Deer	Malaysia
11.	Wheat Genetic Resources Lacking in Wx Protein	Japan

### Announcement JIRCAS E-MAIL ID

JIRCAS has been connected to the computer network "Internet" through MAFFIN-TISN. Anyone or any organization can send electronic mail (e-mail) to JIRCAS through Internet. The e-mail ID is "info@jircas.affrc.go.jp"



# Development of Direct Seeding Culture for Rice Double Cropping in the Tropical Countries

*Koji Kawashima*

JIRCAS (formerly TARC) and MADA (Muda Agricultural Development Authority) have carried out a collaborative research project on rice direct seeding culture since 1988 for five years, entitled: "Establishment of integrated crop production technology for rice cropping system in the Muda area, Malaysia".

During the conference, the research members who had participated in the above collaborative project outlined their activities and summarized the results. Several invited investigators from various national agricultural institutes who are interested in rice direct seeding culture made comments on the lectures. Furthermore, a Thai agronomist who had long been involved in this practice joined the conference and gave a report on direct seeding culture in Thailand; history, methods, technical problems, future trends and economic evaluation.

Many problems must be addressed in the implementation of rice direct seeding culture:

- 1) Weed problem. Besides *Echinochloa* species, the severe occurrence of other weed species and weedy rice that are resistant to herbicides was reported.
- 2) Due to water shortage, dry seeding culture plays a major role. However the instability of rice production associated with dry seeding culture remains to be solved.

3) For direct seeding, paddy varieties adapted to submerged fields, for example, must be selected.

4) Incidence of insect pests after the fallow period was reported to be a serious problem, depending on the insect species.

After the completion of the rice direct

seeding culture project, JIRCAS and MADA have initiated a new collaborative project since 1993 for five years entitled "Integrated research on sustainability of rice production on a long term basis with emphasis placed on biotic agents". Two JIRCAS researchers are being engaged in the project in the Muda area, Malaysia.



*Photo: Due to the labour shortage in the Southeast Asian countries, transplanting is being gradually replaced by direct seeding (Malaysia and Thailand).*

## Research Strategy for Postharvest Technology at JIRCAS

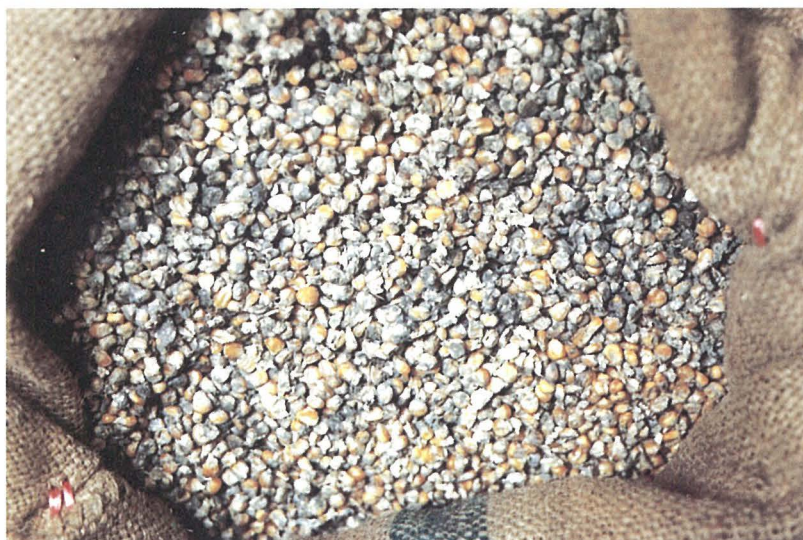
*Koji Kawashima*

At JIRCAS, the research activities related to postharvest technology will be conducted in collaboration with four research divisions as follows: Crop Production and Postharvest Technology Division, Animal Production and Grassland Division, Forestry Division and Fisheries Division.

Up to now, most of the collaborative studies had involved preharvest research areas such as irrigation, breeding, agronomy and crop protection. However, a large number of developing countries are requesting that postharvest problems be addressed by JIRCAS.

During the conference, the following subjects were taken up in relation to postharvest technology research : 1) Preservation of foods through the prevention of biological damage during postharvest storage. 2) Shelf life extension and quality improvement of products, including safety problems. 3) Food distribution technologies that will be compatible with the preservation of the natural environment. 4) Enhancement and stabilization of commodity values through the conversion of products into processed foods. 5) Production of processed foods with high quality. 6) Utilization of undeveloped new food resources.

The research divisions outlined specific research subjects currently implemented. Review of the research activities in the field of postharvest technology carried out at TARC and JIRCAS during the past 20 years revealed that only eleven researchers had been sent overseas mainly to India and Thailand. A questionnaire concerning the overseas collaborative postharvest research problems was distributed to the various national institutes under the Ministry of Agriculture, Forestry and Fisheries. The results of the questionnaire were discussed during the conference.



*Photo: Safety is essential for any food. Due to incomplete drying, agricultural products are often contaminated with mycotoxins (Thailand).*



# JIRCAS RESEARCH HIGHLIGHTS

## Plant response to drought stress and gene expression

Kazuko Yamaguchi-Shinozaki

It has become important recently to breed plants resistant to various environmental stresses to cope with the food crisis and environmental pollution. Recent progress in molecular biology and biotechnology enables to develop transgenic plants that are tolerant to various environmental stresses. For this purpose, it is most important to analyse the molecular process of plant response to environmental stresses. Plants respond to conditions of severe environmental changes or stresses, such as drought, low temperature, or high-salt by undergoing various physiological and developmental changes. Abscisic acid (ABA) production appears to play an important role in the ability of plants to tolerate these stresses. Drought or high-salt conditions induce dehydration of plant cells, which may trigger physiological and biochemical responses against such stresses. *Arabidopsis thaliana* has become a model plant for the analysis of molecular processes of plant growth and development. In order to analyse plant responses to dehydration stress at the molecular level, we cloned genes that are induced by dehydration with *Arabidopsis*. We isolated nine independent cDNAs for genes that are induced by dehydration by applying the technique of differential hybridization and named them RD (Responsive to Dehydration). The nucleotide sequences of the inserts in all nine clones have been determined and homologies to these nine sequences were examined in a protein sequence data base. Proteins encoded by RD19 and RD21 exhibit a significant homology to a number of thiolproteases which may degrade the denatured protein by dehydration. RD22 exhibits a local homology to an unidentified seed protein (USP) of *Vicia faba*. RD28 appears to encode a water channel membrane protein which functions in the transport of water molecules to control osmotic changes during water-stress. RD17 exhibits sequence homology to RAB proteins that protect mature seeds from dehydration. RD29 is identical with low temperature-inducible *cor78* or *lti78* gene, which encodes a hydrophilic protein. No sequence homologies were found for RD20, RD26 or RD2. These nine genes probably are involved in drought-tolerance and can be used to engineer drought-tolerant transgenic crops.

In order to learn more about the expression of drought-inducible genes, we analysed the induction of expression of RD genes by dehydration using Northern hybridization. All the RD genes were induced by water stress within 10 hours, whereas the timing of mRNA induction varied with the RD clones. The transcription of genes that hybridize to RD29 cDNA was induced very rapidly and at a high rate 20 min after the start of dehydration. RD29 is also induced by cold and high-salt stresses and ABA. The genomic clone corresponding to the RD29 cDNA (*rd29A*) was isolated and the expression of *rd29A* was examined in transgenic *Arabidopsis* and tobacco plants by fusion between the 5' flanking region of *rd29A* and the  $\beta$ -glucuronidase (GUS) reporter gene. This fusion gene was induced at significant levels by dehydration, cold, high-salt conditions and ABA in both transgenic *Arabidopsis* and tobacco plants. Histochemical analysis of GUS activity revealed that the *rd29A* promoter functions in almost all the organs and tissues of vegetative plants during water deficiency. We precisely analysed the *rd29A* promoter in both transgenic *Arabidopsis* and tobacco plants, and identified a novel cis-acting element containing 9 bp, TACCGA-CAT (DRE, Dehydration Responsive Element), that is involved in the rapid response of *rd29A* to conditions of dehydration or salinity. DRE is also involved in the induction by low temperature, but does not function in the ABA-responsive induction. Nuclear proteins that specifically bind to DRE were detected in *Arabidopsis* plants under either high-salt or normal conditions.

Different cis-acting elements seem to function in the induction of *rd29A* under conditions of dehydration, high salt, or low temperature. The *rd29A* promoter is useful for the expression of genes involved in environmental stress tolerance to develop transgenic crops that can grow under stress conditions, such as drought, high salt and low temperature.

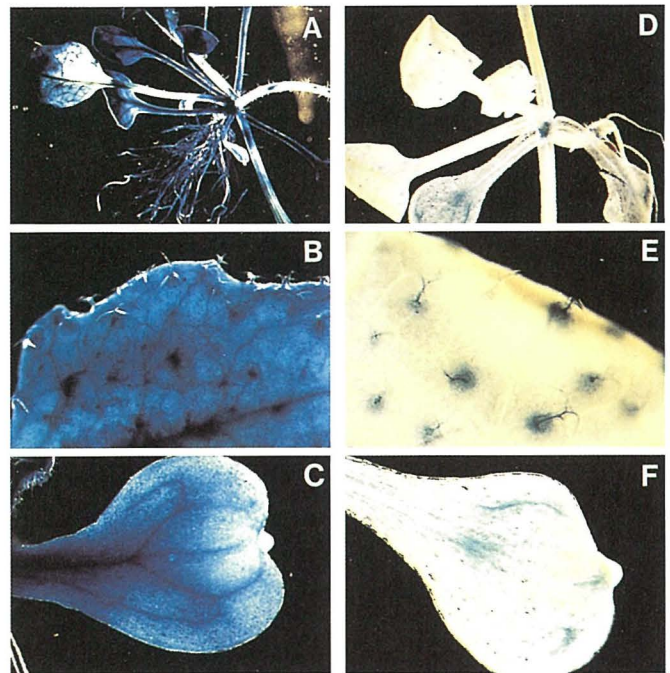


Photo: Histochemical localization of GUS activity in transgenic *Arabidopsis*. Transgenic *Arabidopsis* plants containing the *rd29A*/GUS fusion construct were either exposed to dehydration (A, B and C) or grown under normal conditions (D, E and F). GUS activity is shown in rosettes (A and D), leaves (B and E) and cotyledons (C and F).

## Incidence and control of major viruses of pepper and cucurbit in Thailand

Chiyoichi Noda

Six hundred and thirty-five virus-infected leaf samples of cucurbits were collected from 10 provinces in the central, northern, northeastern and southern regions of Thailand. The presence of 5 viruses was detected by indirect enzyme-linked immunosorbent assay (ELISA) and cucumber mosaic virus (CMV)



Photo: Symptoms caused by virus(es) on Thai melon in a field.



and papaya ringspot virus (PRSV) were found to be the major viruses in cucurbits. From pumpkin, loofah, wax gourd and bitter melon, PRSV was detected at the rates of 62, 40, 24 and 17%, respectively, which were the highest rates on each crop. Besides, both CMV and PRSV were detected from cucumber at high rates of 40 and 34%, respectively. Zucchini yellow mosaic virus (ZYMV) was also found to cause very severe symptoms on cucurbit crops though the incidence was not as high as that of CMV and PRSV. Since CMV, PRSV and ZYMV were found to be the most important viruses in cucurbit crops, the three viruses were isolated from naturally infected leaves to analyse their characteristics for identification based on the symptomatology, particle morphology, virus transmission, physical properties, host range and serology. The antisera of each virus were then produced. Ten pepper viruses were detected by ELISA in the samples collected from 13 provinces in Thailand. Results from the surveys indicated that chilli vein mottle virus (CVMV) and CMV were the most prevalent in all provinces. The two major viruses were isolated, identified and their antisera were produced.

Forty-seven cultivars of cucurbits from Thailand and Japan were tested for their reactions to CMV, PRSV and ZYMV in Kanchanaburi Province. The results indicated that three cucumber cultivars and lines, one calabash cultivar and two melon cultivars were immune (0% infection) or resistant (0.1-10% infection) to the three viruses. Out of 85 pepper cultivars and lines screened for CVMV and CMV resistance, four and three were immune and six and three were resistant to CVMV and CMV, respectively. VC 16a was found to be resistant under greenhouse conditions but tolerant under field conditions to both viruses.

Field tests using cultural and mechanical methods, and foliar sprays for the prevention of virus diseases of cucumber and pepper were conducted in Kanchanaburi Province. Cucumber and pepper were grown in plots with silver mulch spreading, silver tape hanging, oil spraying, skim milk spraying, blue net covering, white net covering, and excessive insecticide spraying together with spraying of insecticides at 2-week intervals. Yield of pepper from the plot sprayed with skim milk at 2-week intervals was 59.2 g per plant, while that from the plot with only insecticide spraying was 29.8 g. Plants from skim milk-sprayed plots grew better and the rates of insect infestation and virus infection were also lower than those from the control plot. As for cucumber, plants from the plot with a silver plastic film as mulch gave a higher yield of 89.7 kg, while the yield from the control plot was 60.0 kg. This cultural practice was also effective in reducing the insect and virus incidence. Conventional methods of excessive insecticide application were not effective in the control of virus diseases in both crops.

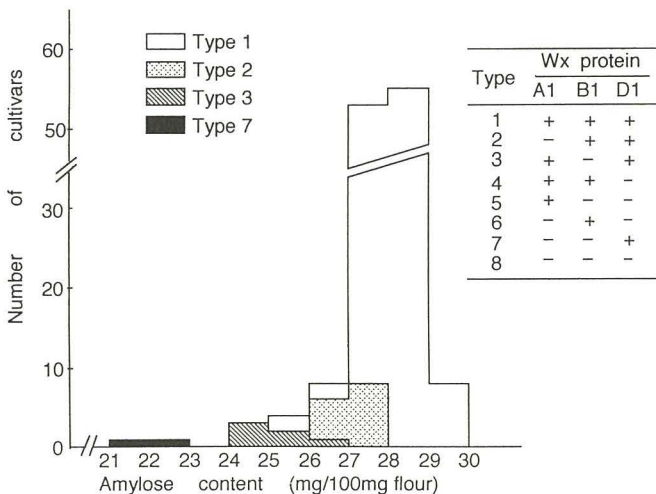


Fig. 1. Classification of apparent amylose contents of 133 Japanese cultivars on the basis of presence or absence of the Wx proteins. Amylose contents (mg per 100 mg flour) were cited from Kuroda *et al.* (1989). + and - in eight types indicate the presence and absence of each Wx protein, respectively.

## Wheat germplasm lacking waxy protein; a new mutant enables to produce waxy wheat

M. Yamamori, T. Nagamine and T. Nakamura<sup>1</sup>

Starch of wheat flour is composed of amylose (ca 25%) and amylopectin (ca 74%). The ratio of these components or amylose content affects the flour quality or palatability of Japanese noodles. Low amylose content tends to increase the glutinosity of noodles. The waxy (Wx) protein tightly bound to starch-granules is known to be involved in the synthesis of amylose. Since the Wx protein level in wheat is correlated to the amylose content, this protein is likely to play an important role in flour quality. Due to allohexaploidy, common wheat (*Triticum aestivum*) has three Wx proteins, Wx-A1, Wx-B1 and Wx-D1, and the genes coding for these proteins are located on chromosomes 7A, 4A and 7D, respectively.

In this study, we examined the deficiency of Wx protein in wheat germplasm derived from several countries and world regions (Table 1). Two electrophoretic techniques, SDS polyacrylamide gel (SDS-PAGE) and two-dimensional gel (2D-PAGE), allowed us to confirm the presence or absence of the three wheat Wx proteins.

Deficiency of the Wx-A1 and Wx-B1 proteins was observed in a few countries (Table 1). The frequency of wheat cultivars lacking the Wx-A1 protein was high in Turkey (51.9%), Japan (16.2%) and Korea (10.8%) in comparison with cultivars from the other countries (0.0 - 6.0%). Of the 159 cultivars deficient in the Wx-B1 protein, 51 originated from Australia and 25 from India. However, only one Chinese cultivar lacked the Wx-D1 protein. While nine Japanese cultivars were deficient in both the Wx-A1 and Wx-B1 proteins, no wheat cultivars lacked both Wx-A1 and Wx-D1 or Wx-B1 and Wx-D1, and all three Wx proteins.

Based on the deficiency of the Wx proteins, wheat was classified into eight types as shown in Fig. 1. Japanese cultivars in which apparent amylose contents had been determined by Kuroda *et al.* (1989) were classified on the basis of these criteria. The amylose contents seemed to decrease in the order of types 1, 2, 3 and 7.

From the cross between Japanese wheat belonging to type 7 and a Chinese wheat cultivar belonging to type 4, eight possible types were developed, which may enable to determine the amount of Wx protein and amylose produced in these wheat cultivars. In diploid cereals, e.g. corn and rice, mutants lacking Wx protein yield waxy endosperm which does not contain amylose. Furthermore, the existence of plants without Wx protein indicates that this protein is not essential for life. Therefore, the type 8 wheat obtained showed the waxy phenotype.

<sup>1</sup> Tohoku National Agricultural Experiment Station

Table 1. Distribution of wheat cultivars lacking the Wx-A1, Wx-B1 and Wx-D1 proteins revealed by the modified SDS-PAGE and 2D-PAGE

Origin	Number of cultivars examined*	Number of cultivars lacking the Wx protein		
		Wx-A1	Wx-B1	Wx-D1
Japan	462 (333)	75 (16.2%)	16	0
South & North Korea	93	10 (10.8)	1	0
China	303 (171)	3 ( 1.0 )	12	1
India	50	3 ( 6.0 )	25	0
Pakistan	85	0 ( 0.0 )	13	0
Afghanistan	59	0 ( 0.0 )	13	0
Turkey	156	81 (51.9)	0	0
Australia	127	1 ( 0.8 )	51	0
North America	315 (172)	3 ( 1.0 )	19	0
Western Europe	172	1 ( 0.6 )	4	0
Russia	133	0 ( 0.0 )	5	0
Total	1,960 (1,551)	177 ( 9.0%)	159	1

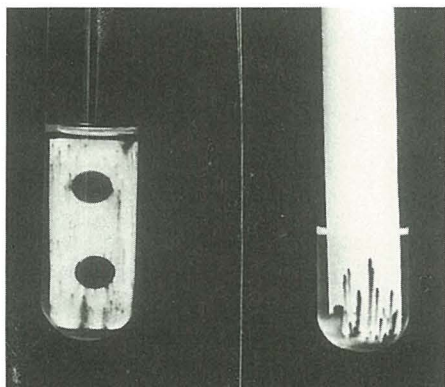
\* All cultivars (1,960) were subjected to the modified SDS-PAGE to determine the deficiency in the Wx-A1 protein. The 2D-PAGE was applied for 1,551 cultivars (333 Japanese, 171 Chinese, 172 North American and all the other cultivars) to determine the lack of the Wx-B1 and Wx-D1 proteins.



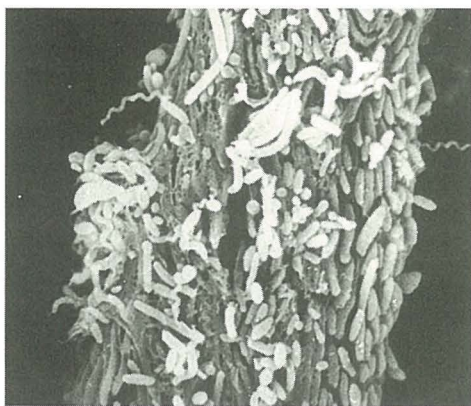
## Isolation and characterization of cellulolytic bacteria from the rumen of tropical ruminants

Kudo, H., Abdullah, N.\* , Ho, Y. W.\* and S. Jalaludin\*

Stomach of ruminants, unlike mono-gastric animals, is divided into three or four compartments, the first and largest of which is called rumen where consumed feed materials are stored and continuous anaerobic fermentation by a complex community of microorganisms (bacteria, protozoa, fungi and probably other unknown microorganisms) takes place. The special characteristic of ruminants is that they can utilize cellulose as energy source. Most ruminants in tropical countries receive cellulosic feed of low quality and its effective utilization is one of the characteristics of tropical ruminants. Although cellulose is one of the most important feed components for ruminants, this polymer is highly resistant to ruminal digestion and only a few genera among the rumen bacteria exhibit high levels of cellulase activity. Therefore, cellulose digesters are probably the most important microorganisms in the rumen of tropical ruminants. *Fibrobacter succinogenes* and *Ruminococcus flavefaciens* synthesize very active cellulases, which can hydrolyze crystalline cellulose. *R. albus* and some strains of *Butyrivibrio fibrisolvens* are also cellulolytic, but these organisms can only degrade the more amorphous non-crystalline types of cellulose. Since the isolation of cellulolytic bacteria has been difficult, especially for *F. succinogenes*, only a few strains of *F. succinogenes* have been isolated and used in the laboratory. Under the joint project with Universiti Pertanian Malaysia, a simple isolation method for cellulolytic bacteria was developed. Cellulolytic bacteria were isolated from the rumen of various ruminants with this new technique. Most of these cellulolytic bacteria were identified as *F. succinogenes*. *R. flavefaciens* was found less frequently, followed by *R. albus*. Many authors have reported that it is difficult to maintain cellulose-digesting bacteria in an active cellulolytic state during



Digestion of cellulose paper by pure culture of *Fibrobacter succinogenes* isolated from water buffalo.



Digestion by consortia of cellulolytic bacteria and non-cellulolytic bacteria (*Treponema*, *Spirochetes*). Cellulose paper was incubated in the rumen of Kedah Kelantan cattle for 2 hours.

repeated transfers. When we transferred both *F. succinogenes* and *R. flavefaciens* cultures every day in a cellulose broth medium lacking soluble carbohydrates, we were able to maintain a high level of cellulolytic activity until completion of the experiment after a 7-month period. Cultures transferred every 4 days maintained a cellulolytic activity throughout the same 7-month period, but this activity was not as strong as that in daily transferred cultures. Transfer at longer intervals (5 to 7 days) often led to the progressive loss of the ability to digest cellulose. The frequency of transfer of cultures of cellulolytic bacteria (*F. succinogenes*, *R. flavefaciens* and *R. albus*) affected the cellulolytic activity. When about top 30% active *F. succinogenes* and *R. flavefaciens* isolated from water buffalo and Kedah Kelantan (KK) cattle were compared, *F. succinogenes* and *R. flavefaciens* isolated from water buffalo exhibited a much stronger cellulase activity than the strains isolated from KK-cattle. *F. succinogenes* isolated from water buffalo showed a greater dependence on rumen fluid. *F. succinogenes* isolated from other ruminants showed a higher dry matter digestibility and gave higher fermentation products from cellulose paper when rumen fluid from water buffalo fed on cellulosic feed was used in the medium. Tests of the rumen fluid from water buffalo, KK-cattle, sheep and goat showed that the rumen fluid from water buffalo was most effective in cellulose digestibility, which may account for the fact that the water buffalo can utilize effectively cellulosic feed with a low quality. Although pure cultures of cellulolytic bacteria digest cellulose *in vitro*, the digestion does not proceed at a similar rate to that seen in the rumen unless consortia are formed with non-cellulolytic bacteria such as *Treponema* sp. *Butyrivibrio fibrisolvens*, indicating that cellulolytic bacteria interact with these non-cellulolytic bacteria to promote the digestion of cellulosic materials. We have shown that primary cellulolytic bacteria adhere to cellulose fibers by specific mechanisms, which can be reversed and enhanced. As we achieve a more complete understanding of the nature of cellulolytic microbial associations, we will explore ways to promote the formation of adherent multispecies populations in order to further accelerate the digestion of cellulose from feed in tropical countries.

(\* Universiti Pertanian Malaysia, Selangor, Malaysia)

## User's Manual for Tropical Agriculture Research Center Information Database Systems

Mitsuo Suzuki

Tropical Agriculture Research Center (presently JIRCAS) publishes six periodicals as follows;

1. Japan Agriculture Research Quarterly (JARQ)
2. Tropical Agriculture Research Series (TARS)
3. Technical Bulletin
4. Technical Document of TARC (in Japanese)
5. Tropical Agriculture Technical Series (in Japanese)
6. Research Reports on Tropical Agriculture (in Japanese)

The main subjects of these journals have been collected and compiled as a database for further use and 3,287 entries have been indexed until now.

This database is constructed by using a card type database software (Ninja4) for a stand alone personal computer and relational type database software (informix) for network use. Also, it can access telephone lines by using ISDN and a modem. Therefore anyone can access the database using adequate code from areas such as Okinawa Subtropical Station.

For retrieving, the database consists of 15 items including: journal name, volume/number, page number, year of publication, subject name, category keyword (main, sub-main, detail), plant/animal name, disease/pest name, chemical/fertilizer name, country/region, instruments, research No. Moreover, a user's manual has been published.

Anyone who wants to use this database must consult the JIRCAS database server and retrieval results are only in the printed form.



## Profile of JIRCAS Divisions (1)

### Research Information Division

#### — Collection and Analysis of Information and Research Coordination —

Research Information Division systematically collects and compiles information on agriculture, forestry and fisheries in developing countries. Research coordinators and other researchers in the division analyse the information to identify research priorities or to formulate JIRCAS research strategy. The results of the analysis are conveyed to the organizations concerned in Japan and overseas.

The effective implementation of international research programs requires accurate analysis of the characteristics of agriculture, forestry and of the agricultural technologies currently applied in various ecoregions and environments. The role played by these sectors in the life of the people that are closely related to the environmental conditions as well as the importance and trend for the national economy in general must be evaluated properly.

Research coordinators are responsible for collecting and compiling information in specific regions or relating to specific problem areas. The regions include Asia I (East and Southeast Asia), Asia II (South and West Asia and North Africa), sub-Saharan Africa, Latin America and developed countries (including International Organizations). Specific problem areas cover food demand and supply projection, environmental conditions and rural development. Research coordinators utilize the information collected to make proposals for new research projects and coordinate special research projects that involve several research divisions based on a multi-disciplinary approach.

The division is also in charge of the development of computer information processing technologies such as data base management, local networking application of multi-media computer for promoting dissemination and utilization of research information.

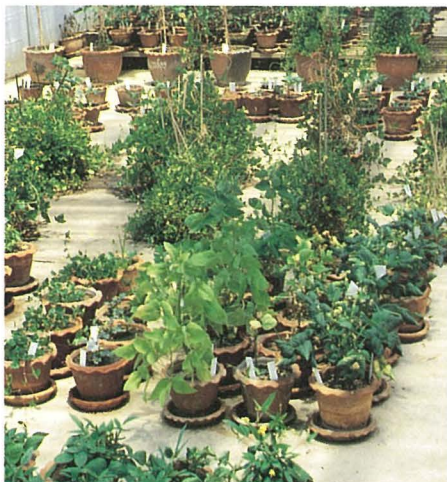


Photo: Multiplication of the seeds of wild *Vigna* germplasm at the Chainat Field Crops Research Center, Thailand for evaluation.

#### Mr. Keiji Ohga



*Director, Research Information Division  
Agricultural Economist. Born in Hokkaido in 1942. Graduated from Tokyo University. Joined Ministry of Agriculture, Forestry and Fisheries in 1967 and mainly worked as policy planning officer. During 1977-1980 worked for FAO in Rome as econometrician to develop World Food Model. During 1983-90 Senior Research Fellow of National Research Institute of Agricultural Economics and carried out research on food policy simulation model building. During 1990-1993 worked for IFPRI in Washington D.C. as Research Fellow and developed International Food Policy Simulation Model.*

#### Dr. Shoji Miyazaki



*Director, Biological Resources Division  
Born in Tokyo in 1943. Graduated from the Faculty of Agriculture, Tokyo University, in 1966. After working at the National Agriculture Experimental Station (1966-70), he carried out research on the introduction to Japan of tropical legumes at the Okinawa Branch of TARC (1970-75) and also on the phylogenetic relationships and classification of Asian *Vigna* at the National Institute of Agricultural Sciences (1976-81). He was appointed principal researcher for the soybean breeding program at Nagano Chushin Agriculture Experiment Station (1983-89) and then Chief of the Germplasm Storage Center, National Institute of Agrobiological Resources (1991-93), where he was in charge of the management of the genebank. In March 1993, he joined TARC as Director of the Eco-physiology Research Division and was appointed Director of the Biological Resources Division, in October 1993.*

## Biological Resources Division

The Biological Resources Division focuses on the conservation and utilization of biological resources in developing countries, by placing emphasis on the development and application of new biotechnological procedures to improve crops as well as on the diagnosis of pathogenic microbes, etc.

Currently, eight projects are being carried out, which can be classified into the following four categories.

1. Collection and evaluation of plant genetic resources: In collaboration with the Department of Agriculture, Thailand, wild species of Asian *Vigna* are being collected in Southeast Asia and evaluated for new gene sources in the mungbean breeding program at the Chai Nat Field Crops Research Center, Thailand. Recently, a collaborative research project on Andean root and tuber crops has been initiated in Ecuador between the International Potato Center (CIP) and JIRCAS.

2. Genetic diversity of Mycoplasma-like Organisms (MLOs): The occurrence of MLOs on sugarcane, sesame and rice plants in Thailand is being investigated in collaboration with Khon Kaen University, Thailand, utilizing newly developed DNA probes.

3. Stress tolerance of cowpea: To improve the stress tolerance of cowpea, an important crop in the sub-Saharan zone, eco-physiological studies are being carried out to evaluate germplasm and breeding lines of cowpea at an outreach of the International Institute of Tropical Agriculture (IITA) in Kano, Nigeria. Supporting studies are also being conducted in Tsukuba, using biotechnological procedures to identify gene(s) responsive to dehydration.

4. Improvement of rice, wheat and vegetables: In collaboration with the Yunnan Academy of Agricultural Sciences, China, a rice breeding project is being carried out in Yunnan Province, China, to develop rice varieties which are high-yielding with disease resistance and cold tolerance, utilizing rice genetic resources from China and Japan. Another collaborative rice breeding project is now being implemented in Malaysia. Also, a vegetable breeding project is being carried out between Shanghai Academy of Agricultural Sciences, China and JIRCAS, to develop new cucumber and green pepper varieties of high quality for subtropical summer cropping.

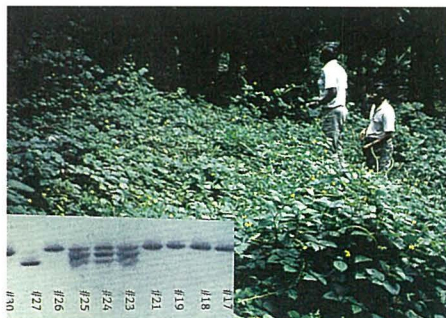


Photo: Collection of wild *Vigna* germplasm in Peninsular Malaysia.



## Environmental Resources Division

Environmental Resources Division started its research activities with fifteen staff members.

As a result of the imbalance in the supply of and demand for food, caused by the increase of the population, which exceeds the level of sustainable production from arable land in a given region, desertification and destruction of the ecosystems in agriculturally marginal lands are proceeding rapidly. To address these problems, research is carried out to identify and develop a technology to promote agricultural activities compatible with the preservation of the environment or ecosystems in lands that had not been easily utilized hitherto, such as arid lands, wetlands, sloped lands, etc., as well as to rehabilitate degraded agricultural lands. In addition, studies are carried out to develop a technology for sustainable agriculture by analysing the mechanisms of nutrient cycling within the ecosystems of arable land. Also to conserve the global environment, techniques are developed to reduce the generation of polluting substances.

Research subjects are as follows;

1. Development of technology for utilization and preservation of environmental resources in arid lands.

(1) Evaluation of large scale water evaporation in the marginal land areas in the

arid zone.

(2) Analysis of weathering process of rocks.

2. Development of technology for utilization and preservation of environmental resources in lowland swamps.

(1) Physiology and ecology of nitrogen-fixing microorganisms in marginal land areas – classification of rhizobia by gene engineering –

3. Development of technology for the re-

### Dr. Michio Araragi



*Director, Environmental Research Division Soil and environmental microbiologist. Born in Saga in 1936. Graduated from Kyushu University and Research Officer in Nat'l Inst. of Agr. Sciences, TARC and Kyushu Nat'l Agr. Exp. Station. Between 1971-74, as TARC staff, carried out research on Microflora of Tropical Paddy and Upland Farm Soils in Thailand.*

habilitation of degraded lands.

(1) Dynamic behavior of soil erosion and deterioration in tropical cultivated lands and development of technology for prevention of these processes.

4. Analysis of preservation function for global environment.

(1) Mechanism of methane formation and development of techniques for decreasing methane formation in humid tropical cultivated lands.

(2) Analysis of dynamics of land utilization.

(3) Development of methods of evaluation of environmental changes associated with the conversion of forest land into agricultural land in the tropics and strategy for the promotion of sustainable land use systems.

5. Analysis of nutrient cycle in arable ecosystems and development of technology for preservation of the environment.

(1) Genesis and characteristics of lowland soils in the tropics.

(2) Physiological and ecological studies of upland rice roots in savanna soils in South America.

(3) Analysis of the effect of pesticides on non-target biological organisms.

(4) Analysis of environmental factors controlling the disappearance of pesticides in soils.

The above studies are carried out abroad by researchers on long or short term assignments as well as at the Tsukuba headquarters.

## Crop Production and Postharvest Technology Division

— For the sustainable supply of high quality food —

### Dr. Koji Kawashima



*Director, Crop production and Postharvest Technology Division. Food Microbiologist: Born in Harbin, China in 1939. Graduated from Hokkaido University in 1962. Since then, he has been engaged in studies on Food Technology carried out mainly at the National Food Research Institute. He visited Malaysia as a member of an IAEA mission for the promotion of "Food Irradiation" program in 1980-81. He participated in a JICA project "Agricultural Products Processing Pilot Plant Project (AP4)" at Bogor Agricultural University, Indonesia, from 1982 to 1983. He joined TARC and was dispatched to Thailand from 1987 to 1990 where he carried out studies on the Quality Preservation of Maize. He returned to the National Food Research Institute, where he held the positions of Director of the Research Planning and Coordination Division and Director of Food Resources Division. In March 1992 he joined TARC again as Director of Research Division II.*

In the developing countries, it is essential to produce a stable and constant supply of food to meet the requirements of a growing population.

Self-reliance in food supply is the main priority for developing countries and many problems must be solved to achieve this objective.

The division will carry out studies covering the various steps of the sequence of the food chain from production to marketing of food products, including disciplines such as agronomy, plant protection (insect, disease and weed control), agricultural mechanization, irrigation, drainage, cropping systems, postharvest technology, food processing technology, farm management and agricultural economics.

Through the intensive utilization of limited arable land, sustainable production of many agricultural products could be achieved. Various technological problems must be addressed for the development of sustainable agriculture which should be compatible with the preservation of the natural environment.

In order to alleviate the water and labour shortage, new cultivation systems should be developed in the paddy production areas.

Research will be conducted for the identification of crop rotation systems for the promotion of sustainable agriculture

adapted to the local conditions.

The amount of chemical inputs used should be reduced and research on biological or cultural control will be promoted, including ecological studies of the natural enemies of insects and prediction of outbreaks of planthoppers that migrate over a long distance.

Postharvest studies of agricultural products will also be conducted. Quality improvement including safety and extension of shelf life of various products as well as insect control of grain storage facilities will be one of the main objectives in this division. Sustainable production of agricultural commodities will be supported by the development of markets for food products that are economically sound. Technology for the enhancement of the value of agricultural products will be developed with a view to contributing to the regional development of the respective countries. For integrated research projects, an economic evaluation will be carried out.





## JIRCAS International Symposium 1993 Plant Genetic Resource Management in the Tropics

The 27th International Symposium on Tropical Agriculture, sponsored by Japan International Research Center for Agricultural Sciences (JIRCAS) in cooperation with the National Institute of Agro-biological Resources (NIAR) was held in the auditorium of the Tsukuba Office of the Secretariat of the Agriculture, Forestry and Fisheries Research Council (SAFFRC), Tsukuba City, Ibaraki on August 24 and 25, 1993. The subject of the symposium covered the current situation and future prospects of plant genetic resource management in the tropics. It aroused much interest on the part of both the Japanese and foreign researchers, resulting in a large number of participants (more than 150), including those from the private sector or NGOs and many foreign researchers staying in Japan, in addition to the delegates from overseas.

The objective of the symposium was to discuss various aspects relating to the management of plant genetic resources, with emphasis placed on technical advances and international cooperation. The symposium consisted of a keynote address and four technical sessions as follows; 1) Exploration and Collection, 2) Evaluation and Utilization, 3) Conservation and 4) Data Management.

During the opening session, Dr. K. Kainuma, Director General of JIRCAS, gave the inaugural address, followed by the welcome addresses by Dr. E. Miwa, Deputy Director General of SAFFRC, and Dr. H. Fujimaki, Director General of NIAR. The symposium started with a keynote address on the Role of International Organizations in Global Plant Genetic Resources Management, delivered by Dr. M. Iwanaga, Deputy Director of IPGRI. He stated that international organizations can play an important role in supporting the development of the national capacity and developing a global system for plant genetic resource management.

The first session for the technical reports included 4 presentations on the exploration and collection of genetic resources, which dealt with the genetic erosion taking place in Asia and South Asia and emphasized the need for further collections and close collaboration with national programs as well as for raising funds for collection and conservation of genetic resources. The session also included 5 current topics on the country situation and the strategies for collection of plant genetic resources, reported by the delegates from Asian countries (Bangladesh, Sri Lanka, Vietnam, Japan and Thailand).

In the following session for the evaluation and utilization of genetic resources, there were 8 presentations, on the genetic variations of rice, sweet potato, potato and *Vigna* species, and on the successful breeding of rice, mungbean and cowpea. Highlights in this session were as follows: 1) it was reported that RFLP analysis had been

applied for the evaluation of genetic variation in rice and sweet potato, 2) promising varieties and clones resistant to the brown planthopper, 2 bruchid species and 2 parasitic weeds had been released using plant genetic resources including wild relatives. All the presentations provided important information on how to use genetic resources for crop breeding programs.

The third session included 4 presentations on international networks for conservation, cryopreservation, *in-situ* and *in-vitro* conservation. In the case of cryopreservation, recent significant achievements in desiccation, vitrification and conventional slow freezing methods were introduced. *In-situ* conservation which is a new concept was highlighted where evolutionary processes continue to operate in nature, thereby keeping the germplasm in tune with the changing environment. The problems relating to the conservation were thoroughly discussed from the viewpoint of the selection of area and species.

In the final session for the data management, the delegates from the NIAR described the sophisticated data base management system (DBMS) developed on the basis of a relational data model, and the new role of the DNA Information and Stock Center (DISC) which prepares mapping data bases in addition to distributing

DNA clones such as cDNA or RFLP markers. The DBMS of the NIAR gene bank can be used in other gene banks with some modifications.

At the end of the symposium, general conclusions were delivered by the chairmen, which can be summarized as follows; (1) The selection of the symposium theme was particularly timely and appropriate considering the need to conserve the vast pool of plant genetic resources for further promoting their utilization for human welfare.

(2) As for the conservation, it is essential that activities for the collection and conservation of plant genetic resources worldwide be undertaken by the international organizations in collaboration with the national programs.

(3) It is important that plant genetic resources are safely conserved, studied and utilized to sustain advances in crop productivity and to stabilize agricultural production.

(4) The need to carry out basic studies was also highlighted during the symposium. Japan with its strong research infrastructure and scientific capability should play a major role in the activities related to the management of plant genetic resources in the tropics.

The proceedings of the symposium will be published as International Agricultural Research Series (IARS) in the near future by JIRCAS. (Mitsunori Oka)

### First Seminar for the promotion of international collaborative studies on agricultural sciences

On January 13, 1994, the seminar entitled "Present Status and Prospects for International Collaborative Studies on Aquaculture" was held at JIRCAS. The seminar aimed at investigating the needs for international collaborative studies in the field of fisheries science, and also at introducing the activities of the JIRCAS Fisheries Division. The theme of the seminar was selected since aquaculture appears to be the most important and relevant field of fisheries science in developing countries.

Nearly 90 participants from national and prefectural institutes, universities, private sectors, and JIRCAS attended the seminar. Six speakers presented papers after the opening address given by Dr. Keiji Kainuma, Director General of JIRCAS.

After all the presentations were completed, the seminar was opened to discussions and six commentators and six advisors gave brief recommendations and suggestions, respectively.

Various kinds of information and suggestions, on how to promote international collaborative studies by the Fisheries Division of JIRCAS in the field of Aquaculture, were supplied throughout the seminar. Among them, it was strongly pointed out by several speakers that the exchange of information and good human relations between counterparts are the most important factors for the successful implementa-

tion of the projects. Program was as follows:

Opening address Dr. Keiji Kainuma  
(D.G. JIRCAS)  
Importance of international collaborative studies on aquaculture  
- past, present, and prospects -

Dr. Masaru Fujiya  
(Councillor, ICLARM)  
FAO international projects on aquaculture  
Dr. Toshihiko Matsusato  
(Senior Researcher, Nt. Inst. Fish. Sci.)  
JICA international projects on aquaculture  
Mr. Yasuo Tadokoro  
(Director, Fish. Tech. Coop. Div., JICA)  
Development of sustainable aquaculture technology with environmental preservation  
Dr. Motoyuki Hara  
(Senior Researcher, Fish. Div., JIRCAS)  
International collaborative studies conducted by universities belonging to the Ministry of Education

Dr. Koichi Ohwada  
(Prof. Ocean Res. Inst., Univ. of Tokyo)  
International technical cooperation on fisheries industry by the Fisheries Agency  
Mr. Ryoza Kaminokado  
(Director, Office of Overseas Fish. Coop.,  
Ocean. Fish. Dep., Fisheries Agency)

(Kunihiko Fukusho)



# New Research Projects 1994

## Evaluation and improvement of "Square" system in Mekong Delta

The cooperative research project on AFFCFS (agriculture, forestry and fisheries combined farming systems), "Square", will start from this year for a duration of 5 years. The detailed plan will be worked out within this year after discussions by both Vietnamese and Japanese authorities. The plan was proposed after the organization of a visit to Vietnam in 1990 and discussions

held with Dr. Nguyen Van Luat, Director of Cuu Long Delta Rice Research Institute (CLDRRI). The proposal was accepted since it was considered to be suitable for the multi-disciplinary approach adopted by the new Center, JIRCAS, which will extend its activities to fisheries.

Vietnam has exported rice since 1989 after the success recorded by the new Doi

Moi policy aiming at the promotion of rice production by giving farmers the right to use land for 15 years on contract with the government. It is reported that out of 2 million hectares of rice fields in the Mekong Delta, 147 thousand hectares are used for floating rice cultivation (water depth more than 1 m) and 320 thousand for deep water rice cultivation (0.5 - 1 m). Although problem soils such as acid sulphate, saline and peat soils are present in the Delta, soils are generally fertile and heat and rainfall are abundant. Various forms of rice-based farming systems are being implemented. The "Square" system consists of paddy fields (5 ha square-shaped plot), water ditches where shrimps and various fishes are raised and rows of trees which are sold on the market. The system seems to be advantageous both by enabling to increase the income through the production of diversified commodities and to avoid over-simplification of the ecosystems.

CLDRRI has studied the system for several years. Japanese researchers may be able to evaluate and improve it.

At present, the following areas require research: **Agronomy:** Weeding practices to avoid adverse effects on fish. **Fisheries:** Introduction of profitable fish species. **Forestry:** Evaluation and diversification of tree species. **Livestock:** Introduction of small-sized animals. **Farm management:** Improvement of profitability. **Ecology:** Significance of diversified ecosystems in relation to sustainability of agriculture.

The institutes with which collaborative studies may be undertaken include CLDRRI, Faculty of Agriculture of Cantho University and Agricultural Science Institute in South Vietnam.

(Kunio Hamamura)



Photo: Square farming area in Mekong Delta, Vietnam.

## Development of Technology for Sustainable Aquaculture in Southeast Asia

The first collaborative research project on aquaculture initiated by JIRCAS is scheduled to be carried out with the National Institute of Coastal Aquaculture (NICA), Department of Fisheries, Ministry of Agriculture and Cooperatives, in Songkhla, southern Thailand. A senior researcher, Dr. Motoyuki Hara, who is specialized in fish genetics will be dispatched to Songkhla to carry out the project, at first. Thereafter, researchers from the Fisheries Division of JIRCAS and National Fisheries Institutes, Fisheries Agency will join the project.

The project aims at developing aquaculture technology in Southeast Asia in taking account of the need to preserve the environmental conditions since the introduction of technology for intensive aquaculture of prawn and fish into these countries in recent years, has tended to destroy the environment of coastal areas, estuaries, and mangrove forests since the industry developed too rapidly and sought only economic benefits. As these phenomena have given rise to serious social problems, the collaborative studies aim at the de-

velopment of a hybrid form of aquaculture technology combining traditional methods and intensive ones.

This hybrid technology which may be referred to as semi-intensive aquaculture, involves the limited use of feeds or fertilization of brackish fish ponds with organic and inorganic materials. Although the productivity is likely to be lower than that of

intensive culture, sustainable production of animal protein which does not generate pollution and enables the preservation of the environment is essential. The project will involve various disciplines, including fish genetics, nutrition, diseases, physiology, and ecology and may require the use of biotechnological procedures.

(Kunihiko Fukusho)



Photo: Aquaculture of prawn in Bali, Indonesia.



# Oil Palm Frond As Ruminant Feed

*Motohiko Ishida<sup>1</sup>, Abu Hassan Osman<sup>2</sup>, Tadashi Nakui<sup>3</sup> and Fuminori Terada<sup>4</sup>*

Self-sufficiency in the products from the ruminant sector in Malaysia has been very low due to slow growth and liberal import policy. Research should be carried out to increase feed supply for ruminants as well as ruminant numbers. Meanwhile, Malaysia leads the world in palm oil production and 17 million tons of oil palm fronds (OPF) are produced on a dry matter basis annually as a by-product. Currently, OPF is left in the field as shown in Photo 1 and often becomes a breeding ground for pests and snakes. However, OPF could be exploited as a good feed source because it is available throughout the year within or in the vicinity of the livestock farms. In taking account of the problem of feed deficit in all the systems of production and the potential amount of fibrous residues from OPF, various activities or options should be considered to narrow down the gap in nutritional needs of ruminants in Malaysia.

## Preservation of OPF as silage

Dry matter content of OPF was 31.1%. The percentage of the dry matter of some components was as follows; crude protein : 4.2, organic cell contents : 25.7, nitrogen cell wall free extract (which represents available carbohydrates) : 22.3, neutral detergent fiber : 69.5. *In vitro* dry matter digestibility of OPF was 35.6%. These results suggested that OPF could be used as a feed.

The quantity of OPF produced daily in the oil palm estates ranges from 50 to 100 kg per hectare on a dry matter basis, depending on the pruning cycle or estate management. It may be possible to use OPF as feed on a large scale if it could be preserved with appropriate and practical methods. In our collaborative study, silage-making was selected and examined. First, water, molasses and urea were examined for use as additives to improve OPF silage quality by packing chopped OPF into 200 liter metal silo with or without the additives. The control silage showed a pH-value of 4.02 and contained 1.9% lactic acid on a dry matter basis. These results indicated that good quality silage could be obtained without any additives, if OPF was ensiled under anaerobic conditions (Photo 2). However, mold was found on top of the silo in the control, water and molasses treatment due to air leaking in the silo during the six-month period of storage. Meanwhile, the silage subjected to urea treatment had no mold. It was suggested that urea should be added at ensiling to prevent mold from growing under aerobic conditions. Forage can be preserved as silage, as long as it is kept under anaerobic conditions. However, once air penetrates into the silo after opening of the silo, the quality deteriorates due to aerobic secondary fermentation. As the humid conditions in the tropics promote aerobic deterioration, the use of silage is difficult in the tropics. Therefore, the effect of urea addition at ensiling on aerobic deterioration was examined in our study. OPF was packed in concrete bunker silos so that the urea concentration in silage was either 0, 1, 2 or 3% on a dry matter basis. One month after ensiling, each sample was exposed to the air and the temperature of the silage was continuously recorded for 144 hr. In the process of aerobic deterioration, heat is produced in silage. In this experiment, in silage without urea heat production started 8 hr after exposure to the air, while only 1% urea addition delayed the initiation of heat production by 28 hr. Besides, maximum temperature decreased by the addition of urea at ensiling. It was found that urea addition could alleviate aerobic deterioration in OPF silage after silos are opened in the tropics.

## Nutritive value of OPF silage

The voluntary intake of OPF silage without urea addition, with 3% urea addition and 6% urea addition was 39.9, 32.1 and 24.0 g dry matter per metabolic body size ( $W^{0.75}$  kg) per day, respectively when the bulls were fed each silage alone. Total digestible nutrients (TDN) of OPF silage, 3% and 6% urea OPF

were 46.1, 49.3 and 43.5, respectively based on digestion trials using Kedah-Kelantan bulls. It was found that the nutritive value of OPF silage was equivalent to that of rice straw and that the addition of less than 3% of urea at ensiling had no adverse effect on the nutritive value of OPF.

## Feeding OPF to beef cattle

Feeding trials were conducted, using 24 Australian Commercial Cross (ACC) bulls to examine the effect of the level of urea-treated OPF silage in the diet and urea treatment on the per-



Photo 1. Oil palm trees and oil palm fronds (OPF) piled up under the trees.



Photo 2. Preparation of oil palm frond silage using a metal drum silo.



formance of beef cattle. The urea concentration in the urea-treated silage was 3% at ensiling. The bulls (6 head per treatment) were fed either diet containing 10 (T1 treatment), 30 (T2 treatment), 50% (T3 treatment) urea-treated OPF silage or 50% OPF silage without urea addition (T4 treatment). The silage was fed to bulls with palm kernel cake (PKC)-based concentrate. After growth trials, the bulls were slaughtered for carcass evaluation. The results are shown in Table 1.

Table 1 shows that concentrate feed required for bulls to gain 1 kg live body weight and carcass fat decreased by feeding OPF silage and that lean meat production could not be significantly reduced by the incorporation of OPF silage in the diet up to 30% on a dry matter basis. These observations clearly showed that OPF silage could be utilized as a feed source for raising beef cattle and that the inclusion of OPF in the diet could reduce the feeding cost.

It was also confirmed in a feeding trial using Sahiwal/Friesian cows that OPF silage could be fed to lactating dairy cows as a roughage without adverse effects on the animal condition and milk flavor. It was suggested that the suitable level of OPF silage incorporation into the diet should reach 30%.

The press conference held at MARDI for announcing the results of the collaborative study has evoked an interest among feed technology experts in Malaysia, indicating that oil palm frond could be a promising feed resource. Many farmers are currently starting to feed oil palm frond to ruminants, using attachments fitted to tractors for the collecting and chopping in the estates.

<sup>1</sup>(National Grassland Research Institute)

<sup>2</sup>(Malaysian Agricultural Research and Development Institute, MARDI)

<sup>3</sup>(Hokkaido National Agricultural Experiment Station)

<sup>4</sup>(Kyusyu National Agricultural Experiment Station)

Table 1. Effect of oil palm frond level in the diet and urea addition at ensiling on performance of Australian Commercial Cross bulls

Items	Diet treatment			
	T1	T2	T3	T4
Live body weight, kg				
Initial weight	229.1	226.5	232.9	229.4
Final weight	396.3 <sup>a</sup>	366.4 <sup>ab</sup>	333.8 <sup>b</sup>	357.2 <sup>ab</sup>
Daily gain, kg/day	0.75 <sup>a</sup>	0.62 <sup>ab</sup>	0.45 <sup>c</sup>	0.57 <sup>bc</sup>
Feed intake				
OPF, kg DM/day	0.70 <sup>c</sup>	1.83 <sup>b</sup>	2.74 <sup>a</sup>	2.79 <sup>a</sup>
Concentrates, kg DM/day	6.32 <sup>a</sup>	4.26 <sup>b</sup>	2.74 <sup>c</sup>	2.79 <sup>c</sup>
Total diet, kg DM/day	7.02 <sup>a</sup>	6.10 <sup>ab</sup>	5.48 <sup>b</sup>	5.58 <sup>b</sup>
Feed: gain ratio				
OPF	0.96 <sup>d</sup>	3.03 <sup>c</sup>	6.21 <sup>a</sup>	4.96 <sup>b</sup>
Concentrates	8.69 <sup>a</sup>	7.06 <sup>b</sup>	6.21 <sup>bc</sup>	4.96 <sup>c</sup>
Total diet	9.65 <sup>b</sup>	10.09 <sup>b</sup>	12.39 <sup>a</sup>	9.92 <sup>b</sup>
Carcass weight, kg	237.2 <sup>a</sup>	210.2 <sup>ab</sup>	189.0 <sup>b</sup>	195.2 <sup>b</sup>
Dressing percentage	60.6 <sup>a</sup>	58.2 <sup>ab</sup>	57.6 <sup>ab</sup>	55.4 <sup>b</sup>
In carcass, kg				
Meat	127.8	121.5	107.0	116.7
Fat	76.4 <sup>a</sup>	58.1 <sup>ab</sup>	45.8 <sup>b</sup>	46.0 <sup>b</sup>
Bone	37.6	33.4	33.2	36.1
Percentage in carcass				
Meat	53.6	58.2	57.2	59.2
Fat	31.6 <sup>a</sup>	27.6 <sup>ab</sup>	24.2 <sup>b</sup>	23.7 <sup>b</sup>
Bone	16.0	16.1	17.7	18.4

T1, T2, T3 and T4: See the text.

<sup>a,b,c</sup> Means with different superscripts differ ( $p < 0.05$ ).

## Letters to the Editor from JIRCAS Visiting Scientists



Photo: Visiting research fellows in 1994; from left to right, Saturnina D. Halos, Bui Ba Bong, Nilse K. S. Yokomizo, Gbade O. Oyediran, Nurliani Bermawie, Sayed M. Hasan, Panie Temiesak, Ancha Srinivasan, M. Mofazzal Hossain and Prasop Virakornphanich.

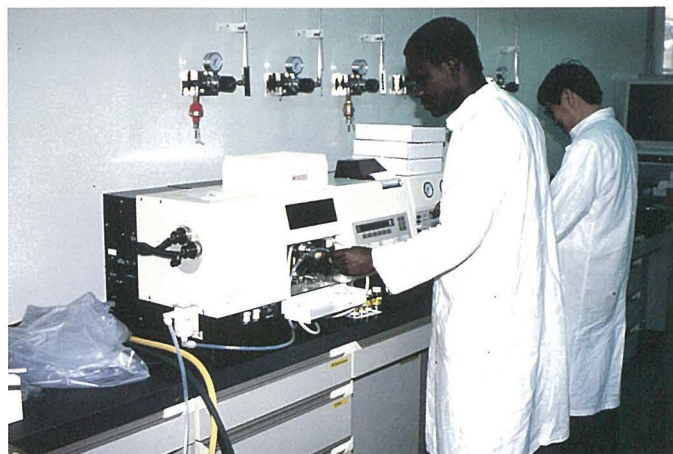


Photo: Research on PCR of sweet potato.



Photo: Methane emission study using gas chromatography.



### **JIRCAS VRF Program — A Timely Initiative for International Collaboration in Sustainable Agriculture**

Sustainability of agro-ecosystems has become an issue of global concern in the 1990s, in particular in the tropics, where the rising populations are putting a heavy demand on resource base, and the technologies adopted so far have not always been suitable for the already fragile environment.

Legumes are essential components of sustainable cropping systems. High temperature stress, especially during flowering, is a major constraint limiting their adaptation in the tropics. The predictions of global climate change also suggest that the frequency of hot weather will increase in the future. Improvement of the heat tolerance of legumes is, therefore, of immediate and increasing concern. To achieve this objective, I am examining genotypic variations and evaluating various selection techniques for heat tolerance in chickpea, pigeonpea, groundnut and soybean.

Dr. Ancha SRINIVASAN  
Postdoctoral Fellow (formerly),  
ICRISAT, Patancheru INDIA

### **A New Heat-Tolerant Leafy Vegetable for the Tropics**

Cabbage is an important and popular vegetable in several tropical countries. However, it grows during the winter season only.

Furthermore, seed production in the tropics is difficult due to the lack of low temperature for flower induction after completion of the juvenile phase. In addition, there is a large difference between winter and summer vegetable production. In Bangladesh, of the 2 million tons of vegetables (excluding tubers) produced annually, about 20 % is produced in summer, hence the vegetable shortage and malnutrition. The development of heat-tolerant leafy vegetables is, therefore, of considerable importance. We were successful in developing several heat-tolerant hybrids using interspecific crosses and protoplast fusion between cabbage and Chinese cabbage. In addition to the tolerance of high temperatures, these materials can produce seeds under tropical conditions.

I am currently, investigating various physiological and genetic characteristics of these hybrids under high temperature conditions. This information should enable Bangladesh as well as other tropical countries to increase vegetable production.

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Institute of Postgraduate Studies  
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### **International Collaboration for Rice Research in Vietnam**

Although rice production in Vietnam is currently increasing, the average rice yield is still low (3.1 t/ha). Nearly half of the 6.3 million ha planted to rice is subject to abiotic stresses (acid sulphate soils, salinity, deepwater and upland conditions). Productivity in these ecosystems is low and unstable. Meanwhile, productivity from high-yielding varieties has reached a plateau in large areas of irrigated ecosystems.

As a researcher from the Cuu Long Delta Rice Research Institute of Vietnam, I appreciate having the opportunity to carry out research under the JIRCAS Visiting Research Program 1993 on the improvement of salt tolerance in rice by employing somaclonal variation and somatic hybridization.

Dr. Bui Ba BONG  
Head, Division of Genetics and  
Plant Breeding  
Cuu Long Delta Rice Research  
Institute  
Omon, Cantho, VIETNAM

### **Wetland Development for Sustainable Rice Production in West Africa**

West Africa consists of about 30 million hectares of wetlands in the form of small inland valleys, river floodplains, inland basins and coastal wetlands, out of which inland valleys account for about one-third of the total area. West Africa is also faced with various environmental problems, such as forest destruction, savannization, desertification, and soil deterioration, which hamper agricultural development. As upland productivity rapidly decreases due to population pressure, lowland agriculture seems to be a valid alternative for food self-reliance.

Of the lowland ecosystems, inland valleys show the highest potential for sustainable development of rice farming. They account for about 20% of the rice area harvested and produce about 22% of the total rice in the region. The inland valley ecosystems have appropriate environments, especially topography and hydrolo-

gy, and are less subject to erosion than uplands, particularly when water control schemes are developed. Under proper management, sustainable continuous double or even triple rice cropping is possible without any fallow period. In addition, the lowlands are useful for the cultivation of other crops such as cowpea, groundnut and vegetables during the dry season.

The opportunity of taking part in the JIRCAS Visiting Research Fellowship Program under the theme "Development of Techniques for Environmental Control Using Plants and Microorganisms Specific to the Tropics and Subtropics", will strengthen our experience and interest in conducting further research on lowland utilization with emphasis placed on Low-Input Sustainable Agriculture (LISA).

Dr. Gbade OYEDIRAN  
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Ladoke Akintola University of  
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Ogbomoso, NIGERIA

### **An Approach to Alleviate the Deforestation Trend in Brazil**

Brazil is undergoing a serious process of deforestation. A survey of the natural cover of Sao Paulo State, the most developed region of Brazil, shows small patches of native vegetation which account for 6.0% of the whole surface, suggesting that in the next century only 3.0% of the primitive vegetation will remain.

This situation can be attributed to the clearing of forest land for the cultivation of coffee, sugar cane, soybeans, orange and other crops and the reckless extraction of minerals and valuable timber. To alleviate this situation, the Brazilian government has implemented a policy to promote sustainable use of these natural forests and concomitantly to stimulate the increase of the rate of reforestation which is estimated at around 2.8 million ha. Presently the demand for timber is about 35 million cubic meters, mostly from the fast-growing tree species *Eucalyptus* and *Pinus*.

The research concerned with the restoration of degraded areas that has been conducted at the ICRS-JIRCAS, using mycorrhizal fungi associated with forest



Photo: Research building for visiting scientists.



trees is highly relevant for this purpose. The use of appropriate mycorrhizal fungi will contribute to the reforestation program, by increasing timber productivity and reducing the need for cutting natural forest.

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YOKOMIZO  
Forestry Institute  
Environment Secretariat of  
Sao Paulo State  
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#### A Note on my Research Activities at ICRS

Salinity is a major constraint for rice production in many countries, especially in South and Southeast Asia. Plants respond to salinity in several ways ranging from readjustment of transport and metabolic processes to inhibition of growth. Many breeding attempts have been made to overcome this problem. In order to formulate an effective breeding strategy, however, a better understanding of the genetic and biochemical basis of salt tolerance is essential.

One approach is to identify salt-induced changes in protein synthesis. In many crops which have been studied, the level of salt-induced protein synthesis appears to be genetically related. To determine whether there are differences in plant response after salt treatment in terms of polypeptide changes, two rice varieties differing in salt tolerance, Pokkali (highly tolerant) and IR28 (sensitive) were used in the studies I am currently carrying out. Presently one dimension PAGE is used for the analysis and later, more detailed investigations will be conducted using two-dimensional PAGE. It is hoped that these studies will contribute to a better understanding of the mechanism underlying salt tolerance in rice.

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#### RAPD for the Detection of Genetic Variation in Sweet Potato and Yam

Restriction Fragment Length Polymorphism (RFLP) has been used for detecting genetic variation in several crops. New methods, independent of restriction sites and DNA probes, are being developed for the detection of polymorphism, using polymerase chain reaction (PCR) with a single short oligonucleotide arbitrary primer involving a set of random amplified polymorphic DNA (RAPD). RAPD requires only a nanogram amount of genomic DNA and thus enables to perform the analysis using a single seedling or even a small leaf from tissue culture. Random DNA segment amplification using single primers appears to be an effective and highly sensitive method for the detection of genetic variation and taxonomic classification in breeding programs and

germplasm preservation. I have developed suitable techniques for DNA preparation, determined the optimal chemical concentration in RAPD reaction and optimal temperature conditions for using the PCR machine for denaturation, annealing and extension of target DNA. I have also developed a protocol for RAPD reaction in sweet potato and yam using single 10-base oligonucleotide OPERON primers and DNA polymerase. I plan to use these techniques to examine the genetic variation both in field-grown and in tissue culture-derived plants.

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#### Crop Fit for Farmers

*"Phosphorus Absorption under Water-Stress"*

The economy of Thailand is based on agriculture. Crop yields and food production have increased during the past two decades, along with the concomitant decrease of the area under natural forest at an alarming rate by the application of new and conventional technology. In fact, when forests are cleared and when continuous farming is adopted, the soil physical, chemical, biological and nutritional properties deteriorate rapidly, and there are adverse effects on the macro and micro-climates. An example of the climatic change is the prolongation of the dry season and the deterioration of the soil water balance resulting in the increase of water runoff and soil erosion. To address these problems, the Thai Government has been promoting water-saving measures since the beginning of the year after the reduction of irrigation for paddy fields from the beginning of last year.

The research theme on the effect of endomycorrhiza on growth and phosphorus content of cowpea under water-stress is particularly timely and appropriate. Since Thai soils contain mainly kaolinitic clay, they are characterized by low available water-holding and low cation-exchange capacities, by acidity and a low level of available plant nutrients, particularly nitrogen and phosphorus.

The methods developed through research should be suited to the local conditions to bridge the gap between what farmers routinely produce and the record yields that are obtained in the experimental stations.

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#### In Vitro Germplasm Conservation for Sweet Potato Breeding

New trends in research and utilization, and traditional farming are useful for sweet potato breeding. Sweet potato is now processed as a snack food and food source. Breeders are proceeding to the selection for other properties that would make sweet potato more suitable for new food processing technologies. Pigments such as anthocyanins and beta-carotenes are also being studied as food-coloring products. In traditional horticulture in developing countries this crop compensates for rice crop failure. Breeding efforts are thus focused on enabling sweet potato to grow in soils with low fertility.

Such differences in plant breeding objectives require the availability of a wide variety of germplasm accessions. However, because sweet potato is asexually propagated, germplasm conservation is costly and difficult. Germplasm is maintained mainly in field plots. Research on *in vitro* conservation of sweet potato germplasm may enable to conserve and obtain a large collection of germplasm.

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THE PHILIPPINES

#### In Vitro Preservation of Yam (*Dioscorea* spp.) Germplasm

*Dioscorea* spp. (yam) is one of the vegetatively propagated crops in the tropical regions. This crop is sexually sterile which precludes the possibility of seed storage. The storage organs, i.e. tubers, are short-lived and susceptible to disease and pest damage. Storage of tissue cultures is a suitable method for the conservation of this crop.

I have been able to multiply plantlets and I have treated nodal cuttings of several accessions of *D. alata*, *D. bulbifera*, *D. esculenta*, *D. opposita* and *D. rotundata* under various *in vitro* conditions to reduce the growth rate for the short-term preservation test. Studies on the protein banding pattern through polyacrylamide gel electrophoresis are also being carried out in order to detect the effect of prolonged *in-vitro* storage on the genetic stability of the strains.

The interruption of the metabolic processes at ultra-low temperatures can prevent the genetic instability of the storage tissues. Alginate-coated beads of nodal segments were successfully prepared for the cryogenic storage. The objective is to obtain a sufficient level of dehydration of the encapsulated-nodal segments, by osmotic dehydration and air-drying methods, before the materials are immersed into liquid nitrogen for long-term storage.

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## ILCA and Collaborative Research program

*Kenji Togashi*

The International Livestock Centre for Africa (ILCA) was established in 1974. Its headquarters are in Ethiopia, located on a 36-hectare site on the eastern edge of the capital city of Addis Ababa. ILCA has research sites in Ethiopia (highland and semi-arid zones), Kenya (subhumid zone), Nigeria (humid and subhumid zones), Mali (subhumid and semi-arid zones) and Niger (semi-arid zone).

Over the past 20 years Africa's human population grew by about 3 % a year, while the total agricultural output has been growing at less than 1.5% a year. The number of Africa's people at risk of hunger is anticipated to reach 185 million in 2000. Most of the people are tied to agriculture for their well-being. Livestock in Africa provide meat, milk, eggs, blood, hides and skins for consumption at home or for sales in the market. In African farming systems, they also provide draught power, transport, fuel and manure which is only available for farmers and transfers nutrients from renewable sources, such as rangelands, to crop lands. In addition, animals have been a source of identity and prestige for the family and have been the basis for building and maintaining social relationships. Moreover, in the drier part of the region, livestock make it possible for people to live in areas which could not support crop farming.

Opportunities for increasing livestock production can be divided into three categories: expansion of production into new areas, increased productivity in areas where livestock are already kept and improvements in marketing.

### **Expanding production:**

Control or eradication of trypanosomiasis would open up to 10 million km<sup>2</sup> of land to livestock in the higher rainfall areas. This potentially productive forage zone could then carry as many as 120 million head of cattle.

### **Increasing productivity:**

Despite the large livestock population, the level of productivity is extremely low. Although one-seventh of the world's cattle are raised in Africa, they produce only one-fourteenth of the world's meat. In other words, 50 head of African cattle are required to produce 1 ton of meat, whereas semi-intensive grassland productive system in Australia produced 1 ton of meat from only 14 head.

### **Improvement of marketing facilities:**

Marketing facilities are barely adequate. Markets are often far from production areas, and prices offered to producers are low. Where roads and railways exist, freight charges are often prohibitive, and when animals are trekked over a long distance to market, substantial weight loss and death occur. As a result, relatively few potential consumers can afford to purchase livestock products regularly.

Despite the above opportunities, increased production will not come easily. The most common constraints are the low

genetic potential of Africa's livestock, inadequate nutrition and animal diseases. Most of the livestock in Africa subsist on range-land grasses. Very little formal supplementary feeding is practiced. Poorly fed animals suffer from higher mortality, especially among the young, even up to 50%. Technical solution, in some instances, to diseases has been available but only at a high cost.

The objective of collaborative programs lies in the solution of the constraints of low genetic potential and in the identification and/or production of cattle which show resistance or tolerance to environmental stress, including diseases. African producers have tended to select animals according to the ability to survive under adverse conditions such as diseases, fluctuating feed and water supply, poor feed and high temperature rather than high levels of production. In lower-potential and more highly fluctuating environments, the ability to

survive is the dominant selection criterion. However, selection for higher production is essential to raise livestock productivity and reduce the number of people in abject poverty. Characterization of indigenous and exotic cattle with emphasis on both the ability for survival represented by resistance or tolerance to diseases and the ability for higher production has not been practiced. Therefore, firstly, the collaborative research program aims at characterizing indigenous and exotic cattle which have been already introduced to Africa. However, analytical methods for simultaneous evaluation of animal survival and higher production are not available. The first objective of the ILCA/JIRCAS (TARC) collaboration is to establish a scientific analytical method, which will then be used to characterize indigenous and exotic livestock populations. In conclusion, animals in Africa have been traditionally used for human subsistence rather than for the generation of income.

However, there is an urgent need for African cattle improvement to include not only hardiness, but also higher production.



### **Dr. Y. Ohno Appointed to NARC**

Dr. Yoshikazu Ohno, Director of Research Information Division of JIRCAS, was appointed Research Coordinator General of NARC (National Agriculture Research Center) on 1 March 1994. He joined TARC in 1970 as a founding member and carried out research on plant-soil relations both at IRRI and in Brazil. After he was appointed Director of the Research Information Division at TARC in 1989, he devoted his time and energy to expanding the scope of English publications by increasing their number and upgrading their quality. He played a major role in the publication of the Newsletter. In addition he contributed significantly to the construction of database systems in the field of tropical agriculture.

### **Notice**

Due to the reorganization of the Tropical Agriculture Research Center (TARC) into the Japan International Research Center for Agricultural Sciences (JIRCAS), the TARC quarterly Newsletter is published as JIRCAS Newsletter twice a year.

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