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Feature Article Offshore Environmental Research

The argument for protecting the environment is based on the fear that the biotopes will be unable to withstand the disturbances caused by human activities, along with a deep concern for the future generations, in particular the under-privileged groups in the society.

Despite the development and use of modern measurements and technologies, far too little is actually being done for the promotion of sustainable development. There is not even a clear agreement as to what sustainable development is. Under these conditions, to enable the development of common standards for sustainable production in the field of agriculture, forestry and fisheries, scientific research will play a crucial role.

Coastal zones sharply illustrate the problems and policy challenges faced locally as well as on a global scale. These zones are under increasing environmental pressure and are showing an unacceptable level of environmental degradation as a result of population growth, urbanization, reckless adoption of aquaculture and other multiple and often conflicting changes in resource utilization. The alleviation of these problems and the adoption of practical and sustainable economic development policy objectives will require innovative policy responses. Integrated coastal zone management (ICZM) is one of such comprehensive policy/management options. ICZM requires the formulation of a wide range of general principles, as well as the combination of economically efficient and sustainable resource utilization with prevention strategies.

In many coastal areas, maintenance or expansion of the regional economy is a major, often the primary, objective. However, the benefits associated with the development in coastal

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economies, such as tax revenues, tourist expenditures, and employment may be reduced as a result of the deterioration of water quality.

In the context of regional and area economic development, the objectives of ICZM can be expressed in two forms: the negative and positive aspects. On the negative side, people



will have to bear the costs of normal production, net coastal management, discharge reduction, mangrove or beach restoration, coastal protection, cost for repair of existing damage and the administrative cost of management. The positive factors include improved income from increased gross regional product combined with benefits from improved environmental quality processes which are an international focus of interest. Fo example, by using natural microbial foods and taking advantage of the capacity of highly productive mangrove ecosystems to absorb effluents, aquaculture can become an eco-friendly process (Fig. 1), that will provide the fishermen with a higher income in the long term.

In addition, we should remember the words of the famous novelist Ryotaro Shiba: "People of the late 20th century, having gained the knowledge that they are merely one part of Nature, have again begun to fear Nature in the way people feared the gods in ancient times. The period in which man regards himself as master of Nature will come to an end as we draw increasingly close to the 21st century. Human beings do not live on their own, but are kept alive by a great presence."



Fig. 1. Use of natural food chain and coastal purification procedures for aquaculture.

Collaborative Research Project

Collaborative Research Projects between ICIPE and JIRCAS

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The International Centre of Insect Physiology and Ecology (ICIPE), based in Nairobi, Kenya, was established in 1970 as an advanced research institute amidst growing global concern about the misuse and overuse of synthetic pesticides. ICIPE was registered as an intergovernmental organization in 1986, with governance through a 16-member international Governing Council. Due to its broader mandate encompassing health and environment as well as agriculture, ICIPE has remained outside the CGIAR system, whose work is primarily commodity-oriented. Interested donors judiciously established a Sponsoring Group (SGI) in 1980 to help guarantee scientific excellence and funding.

Today, ICIPE continues to follow its original mandate of developing technologies to alleviate world poverty and to ensure food security and good health for the peoples of the ropics through management and control of both harmful and useful arthropods. The Centre's current objectives aim at improving and promoting the activities of the "4H's" (human, animal, plant and environmental health) interdisciplinary teams of scientists engaged in research related to ecosystems science, behavioral biology and chemical ecology, molecular







Sign at the entrance of ICIPE



Nymphs of *Schistocerca gregaria* (Photo by S. Tanaka, Natl. Inst. Sericult. Entomol. Sci.)

biology and biotechnology and social sciences. Research support is provided by biomathematics, animal breeding and quarantine and biosystematics units, and computer and information services. The 360-strong staff members originate from 19 countries, primarily in developing regions. Programs are mainly focused on plant pest management, disease vector management, insect biodiversity conservation and utilization, and capacity building from post-doctoral to farmer level.

Collaboration is a by-word at ICIPE, with partners from 45 universities throughout the world, including 30 African universities in ARPPIS (The African Regional Postgraduate Programme in Insect Science), other IARCs and CGIAR centres, more than 20 advanced research institutions, NARS in Africa and beyond, and NGOs.

JIRCAS is one of these partners and JIRCAS-ICIPE collaborative research started from 1994 and then the project: "Biorational Approaches to Long-term and Sustainable Management of Desert Locusts in East and North Africa" was conducted during the period 1995-1999, which produced fruitful results on the mechanism of change from the "solitary" to harmful "gregarious" phase of desert locusts, based mainly on physiological and endocrinological studies. Presently these studies are being taken over to the new five-year project: "Physiological and Ecological Studies for the Development of IPM for Economically Important Pests in Africa" funded by the Ministry of Agriculture, Forestry and Fisheries of Japan.

Besides locusts, another objective of the new project is to focus on the biological control of stemborers of staple cereals in East Africa. The spotted stemborer, *Chilo partellus*, an exotic cereal pest that was accidentally introduced into Africa from Asia in the 1930s, has been responsible for crop losses of up to 80%. A natural enemy of the stemborer, a parasitic wasp *Cotesia flavipes*, was imported by ICIPE from Pakistan in 1991 and has been found to be one of the most effective biological control agents for this destructive pest. However, for further suppression of stemborer densities, other promising natural enemies will be studied and introduced in the near future.

JIRCAS Research Highlight Utilization of Oil Palm Residues as a Raw Material for Pulp and Paper

African oil palm, *Elaeis guineensis*, is one of the most important plants in Malaysia. It produces palm oil and palm kernel oil, which are widely used in food and other industries such as detergents and cosmetics. Malaysia is the world's largest producer and exporter of the oil, accounting for approximately 10% of the world's oil and fat production (1998). The total area of oil palm plantations is close to 3.2 million hectares, which account for almost 50% of the land under cultivation in Malaysia.

Although oil from the palm tree is an excellent product for the country, residues from oil palm have not been used sufficiently. One of the abundant lignocellulosic residues consists of empty fruit bunches (EFB), which are left behind after removal of oil palm fruits for the oil refining process at palm oil refineries. Tons of EFB are discharged from each refinery regularly, amounting to 16 million tons per annum in the year 2000. Some of them are used as fuel at the factories, but most of them are left unused.

Converting this lignocellulosic waste to paper-making pulp is a promising way of utilization, as the consumption of paper is increasing dramatically in the Southeast Asian countries including Malaysia. Several investigations have been carried out on EFB pulping, whereas studies on bleaching of EFB pulp have been very limited so far. Collaboration work between JIRCAS and Universiti Sains Malaysia (USM) has, therefore, focused on the bleaching of chemical pulp from EFB, especially using total chlorine-free (TCF) methods.

TCF bleaching was examined for EFB chemical pulps such as kraft, soda and soda-anthraquinone pulps. TCF processes include oxygen (O_2) delignification, ozone (O_3) bleaching,

acid treatment, hydrogen peroxide (H₂O₂) bleaching and alkali extraction. It was found that a pulp brightness of 75% can be obtained for EFB kraft pulp by a bleaching sequence of O_2 - acid - O_3 - H_2O_2 . The brightness is required to be above 80% for commercial bleached kraft pulp based on JIS Standard. Mechanical properties such as tensile and tear strength of handsheets produced from this pulp were found to be comparable to those of hardwood pulps. Furthermore, since the TCF bleaching method removes most of the coloring compounds, that is lignin, in EFB, the bleaching ability is assumed to be similar to that of bleaching methods using chlorine. Soda pulp and soda-anthraquinone pulp of EFB, which contain less lignin than EFB kraft pulp, have also reached a brightness level of 70~75% with a single bleaching stage using O_2 or O_3 . This also indicates that the coloring materials in EFB can be easily removed without using any chlorine com pounds.

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Based on this study, we observed that the total chlorinefree bleaching method can be applied to EFB chemical pulps and that EFB is suitable as a raw material of chemical pulp. Although the bleaching and even pulping methods for producing EFB pulp for paper-making have not been optimized yet, there is a great potential for the utilization of this oil palm lignocellulosic residue in pulp and paper industries. Currently, the world trend in pulp bleaching is toward the application of non-chlorine processes because of environmental concern. Once the oil palm by-product becomes a raw material for pulp production, it will be essential to develop TCF bleaching processes in future.



Oil palm tree



Empty fruit bunch (left), fibrous strands of EFB (right top) and paper sheets made of TCF-bleached EFB pulp (right bottom).

Simple and Rapid Technique for the Identification of an Aromatic Thai Rice Variety (Khao Dawk Mali 105)

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In order to meet consumers' demand for food, a large quantity of rice (*Oryza sativa* L.) is produced in many parts of the world. Aromatic rice types, i.e. rice varieties having a stronger aroma than ordinary rice, have become increasingly popular in Southeast Asian countries, Europe and the U.S.A.

An aromatic variety, Khao Dawk Mali 105 (KDML105), is mainly produced in Northeast Thailand. Being recognized for its better quality, market demand for this variety has increased both in Thailand and overseas. Although increase of production is urgently needed, the cultivation is limited due to infertile and drought-striken sandy soils. Moreover, erratic rainfall at the beginning of the rainy season and labor shortage for transplanting are other constraints on the production of KDML 105. As a matter of fact, no objective method for the identification of such an aromatic variety has been developed. As a result, varieties different from KDML 105 are often marketed under the name of KDML 105.

In Japan, since rice is distributed as brown rice which can germinate, information about varieties can be obtained. In contrast, in Southeast Asia, rice is mainly marketed as milled rice unable to germinate. To identify varieties of milled rice in a mixture with varieties of similar appearance, conventional methods such as determination of the length-width ratio are not suitable.

Molecular marker techniques based on the polymerase chain reaction (PCR), have provided more objective methods for the identification of grain varieties. These techniques (i.e., microsatellite analysis, randomly amplified polymorphic DNA (RAPD) and amplified fragment length polymorphism (AFLP)) could be very useful for plant-based identification; microsatellite analysis is particularly useful for simple and rapid analysis. To apply this technique to milled rice, however, high quality DNA must be extracted from a single grain of milled rice. It is difficult to extract DNA, because the main components (starch and proteins) of milled rice show the same solubility as that of DNA. It should be emphasized that enzyme-inhibiting polysaccharides often contaminate the "purified" DNA.



from rice market samples by PCR using primer set RM17. Arrowheads indicate the position of the amplified fragments of authentic KDML105. Extracting procedures for simple and rapid identification of grain varieties require breakage of the cell wall, disruption of the cell membrane, protection of DNA from endogenous nucleases and finally removal of polysaccharides.

In our laboratory, we developed a DNA extraction method based on a freeze-and-thaw cycle for extracting purified, high molecular weight DNA from grains. The developed technique does not require expensive equipment and materials, and is not a time-consuming procedure.

Microsatellite analytical method for cultivar identification was used for extracting DNA. We analyzed rice samples obtained from Bangkok, Suwannaphum (Northeast Thailand) and Kuala Lumpur markets as well as selected varieties different from KDML 105 (Bangkok and Kuala Lumpur market rice samples). Fig. 1 shows the results of microsatellite analysis of market samples on agarose gel. Some of the rice samples originating from Kuala Lumpur and Bangkok markets did not match the KDML 105 variety.

A simple and rapid method for cultivar identification of milled rice is illustrated in Fig. 2. By applying the improved method, we were able to obtain DNA for microsatellite analysis within 2 h and the whole process of identification required 6 h. Time saving and the possibility to avoid the use of expensive instruments or toxic chemicals in the present method are an attractive alternative to existing methods of identification. The results obtained showed that DNA extracted from tested samples was of high quality and hence could be used in PCR-based techniques for cultivar identification even in laboratories with a moderate level of technology. Another feature of this developed technique was the saving in time and cost as well as the safety of the procedure.



Genetic Variation in Amylose Content and Lipoxygenase-3 Activity of Seeds from Rice Genetic Resources in Yunnan, China

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Yunnan province in China is a center for genetic diversity in Asian cultivated rice (*Oryza sativa* L.). We studied the biochemical properties of rice grains using a wide variety of genetic resources from Yunnan and Japan.

The amylose content in endosperm starch is a major determinant of eating, cooking, and processing of rice grains. Starch in nonglutinous endosperm contains both amylose and amylopectin, whereas starch in glutinous endosperm contains only amylopectin and lacks amylose, or contains a small amount of amylose. The eating quality of cooked rice is negatively correlated with the amylose content in nonglutinous rice. Cooler temperature during grain filling considerably affects the increase of the amylose content of rice plants cultivated in paddy fields at an altitude of 1500 - 2100 m above sea level in Yunnan province. To improve the eating quality, YAAS breeders are implementing a breeding program to develop new rice varieties with a low content of amylose.

We screened 581 varieties from Yunnan (n=376) and Japan (n=205) for the endosperm amylose content (Fig. 1). Yunnan rice showed a 1.7 times larger variation than Japanese rice based on the Shannon-Wiener index (H'). Upland rice exhibited the largest variation in the amylose content



Fig. 1. Frequency distribution of varieties differing in the endosperm amylose content in Japanese rice and three groups of Yunnan indigenous rice. Varieties with less than 3% belong to glutinous rice.

among the varietal groups from Yunnan: Indica paddy rice (H'=0.734), Japonica paddy rice (H'=0.844), and upland rice (H'=0.909). Upland rice varieties with a low amylose content (5-11%) are expected to be useful for the breeding of new varieties with improved eating quality in highland areas.

Rice grain deterioration and the development of staleness during storage are serious problems that reduce quality. It is difficult for developing countries to construct temperaturecontrolled warehouses that would help ensure proper treatment of harvested rice. Lipid degradation is considered to be responsible for the deterioration during rice storage. Lipoxygenase enzyme catalyzes the oxidation of polyunsaturated fatty acids containing a 1,4-pentadiene moiety into conjugated hydroperoxy fatty acids. The lack of lipoxygenase in rice grains may contribute to the reduction of oxidative deterioration during storage.

We screened the germplasm collection from Yunnan for (the lipoxygenase-3 (LOX-3) content in embryos, by using monoclonal antibodies against LOX-3, in collaboration with Dr. Y. Suzuki, National Agriculture Research Center, Japan. We detected 22 varieties that lacked LOX-3, the major component among isozymes in rice seeds, after screening 108 varieties from Yunnan rice genetic resources. Most of the varieties that lacked LOX-3 belonged to upland rice, and to Groups 4 and 6 based on the esterase isozyme genotype of *Est1*, *Est2*, and *Est3* (Fig. 2).

Major agronomic features of the varieties lacking LOX-3 include early heading, red and round grains, long culms, and glabrous leaves and hulls. These varieties showed a poor agronomic performance in the paddy fields because of severe lodging before harvest. The lack of LOX-3 was inherited as



Fig. 2. Relationship between lipoxygenase-3 (LOX-3) activity in seeds and rice varietal groups based on genotypes of esterase isozymes (*Est1*, *Est2*, and *Est3*) in leaves. a single recessive trait in a cross between CI-115 lacking LOX-3 and Norin 20 having LOX-3. CI-115 is a pure line selected from the indigenous upland variety Chongtui. No tight linkage was observed between the lack of LOX-3 and the red pericarp derived from CI-115.

Varieties lacking LOX-3 could help reduce off-flavor formation in rice stored in warehouses that are not temperaturecontrolled. We are currently developing breeding lines lacking LOX-3 by crossing CI-115 with improved varieties that are agronomically superior.

Workshop

JIRCAS International Workshop:

"Brackish Water Mangrove Ecosystems – Productivity and Sustainable Utilization" February 29 – March 1, 2000, Tsukuba, Japan

Twenty-one technical papers and 3 panel papers were presented at this workshop, which was attended by 114 participants. The workshop focused on the socio-ecological conditions in the Matang, Merbok and Lumut mangrove ecosystems in Malaysia, the main sites of the collaborative study entitled "Productivity and Sustainable Utilization of Brackish Water Mangrove Ecosystems," which was carried out in Malaysia from April 1995 to March 2000 by JIRCAS, Fisheries and Forestry Agencies in Japan, and the Fisheries Research Institute (FRI), the University of Malaya (UM) and the Forest Research Institute (FRIM) in Malaysia. The workshop consisted of 5 sessions: (1) Structure, management and reforestation of mangrove areas; (2) Nutrients, plankton and fish species in brackish water mangrove areas; (3) The ecological significance of benthic animals in the mangrove forest biotope; (4) Aquaculture in mangrove estuary areas; and (5) General Discussion on collaborative work on ecosystem research in brackish water mangrove areas.

This international workshop was held in the final year of the JIRCAS comprehensive project mentioned above, for which two rivers in Matang (the Selinsing and Sangga), one river each in Merbok (the Merbok) and in Lumut (the Dinding) were selected as the research sites in Malaysia. The characteristic differences between the three mangrove areas are that the ratios of mangrove forest area to river area are 4.7 in Matang, 2.1 in Merbok and 1.1 in Lumut. During this workshop, a wide range of results were reported, including the following: (1) the brackish water area of the River Selinsing in Matang was found to be important as a breeding ground for penaeid shrimp, (2) the freshwater areas of the River Sangga in Matang act as a feeding site for *Arridae* spp., (3) areas of the River Merbok are feeding sites for *Scianidae* spp. and (4) the freshwater areas of the River Dinding in Lumut act as a migration site for marine fish species, including *Leiognathidae* spp. The range of species and the ecological features of prawns and benthic animals were also elucidated in the three mangrove areas. Concerning the diversity of fish, in the Matang mangrove estuary area, the volume of fish catches was greater than that in the Merbok and Lumut ones. The diversity index of fish, however, showed a higher value in the Merbok than in the other two mangrove areas. In the Lumut area, the volume of fish catches and diversity index were the lowest of the three areas.

Socio-economic studies in these mangrove areas have additionally revealed that in well-managed areas, such as the Matang Mangrove Forest Reserve, mangrove forests generate significant social and economic advantages. On the other hand, inappropriate management of mangrove areas causes socio-economic losses due to the exhaustion of available resources.

JIRCAS would like to thank various people, the chairmen, speakers and delegates, for their excellent presentations and for the stimulating discussions during the workshop.

(Masachika Maeda)

JIRCAS International Workshop Brackish Water Mangrove Ecosystems -Productivity and Sustainable Utilization-

Feb. 29 ~ Mar. 1, 2000 International Congress Center "Epochal Tsukuba"

Organized by Japan International Research Center for Agricultural Sciences

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Symposium

JIRCAS International Symposium

The 7th JIRCAS International Symposium on "Agricultural Technology Research for Sustainable Development in Developing Regions" will be held on November 1-2 at Epochal-Tsukuba in conjunction with the 30th anniversary of TARC-JIRCAS. This is an opportune time to re-assess the progress of agricultural development in developing countries and to re-examine the roles and contributions of the different actors involved in development, from farmers to national research and extension organizations, non-governmental organizations, the private sector, as well as government and international research institutes including JIRCAS.

For further information, please contact: Dr. M. Yajima Secretariat of the Organizing Committee for the 7th JIRCAS Symposium: Fax: +81-298-38-6342 E-mail: sympo7@jircas.affrc.go.jp

PEOPLE



Dr. Masa IWANAGA, a plant geneticist, became Director of JIRCAS's Biological Resources Division on April 1, succeeding Dr. Tsuguhiro Hoshino who was appointed Director of the Department of Crop Physiology and Quality, National Agriculture Research Center. Dr. Iwanaga worked as Deputy Director General (Programmes) at the International Plant Genetic Resources Institute (IPGRI), Rome, Italy, for more than seven years. Before IPGRI, he worked for CIAT and CIP for three and ten years, respectively. Dr. Iwanaga holds a MS degree from Kyoto University and Ph. D. from the University of Wisconsin-Madison.



Dr. Osamu ITO, a plant physiologist, became Director of JIRCAS's Environmental Resources Division on April 1, succeeding Dr. Tadao Hamazaki who became Director of the Department of Natural Resources, National Institute of Agro-Environmental Sciences. Dr. Ito worked as International Research Coordinator for Thailand at JIRCAS's Research Information Division for six months after spending three years at the International Rice Research Institute as Director of the Agronomy, Plant Physiology and Agro-Ecology Division.

Dr. Makie KOKUBUN left JIRCAS on April 1, to take up his appointment as Professor of the Graduate School of Agriculture, Tohoku University. Dr. Kokubun served as International Research Coordinator for South America at JIRCAS's Research Information Division for four years.

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