Japan ISSN 0917-2378

Research Highlights '93

September 1993

TARC

TROPICAL AGRICULTURE RESEARCH CENTER

Research Period: 1990–91
Research Site: Yunnan Academy of Agricultural Sciences, the People's Republic of China

Procedure for the Evaluation of Field Resistance and Identification of Genotypes for True Resistance of Rice Varieties to Blast Disease in Yunnan Province, China

Yoshikatsu FUJITA[1], Jiarui LI[2] and Chengyun LI[2]
1) Hokuriku National Agricultural Experiment Station, Japan, 2) Yunnan Academy of Agricultural Sciences, the People's Republic of China

In Yunnan Province, japonica rice is cultivated in the paddy fields located at an elevation of less than 1,500 m above the sea level. Rice blast disease severely affects japonica rice in Yunnan province, China. Basic and reliable measures to control this disease consist of the development and use of resistant varieties. Varietal resistance to blast disease is divided into true resistance and field resistance. The former is determined by the all-or-none disease response of a certain variety to a certain race of the fungus, and the latter is determined by the severity of the disease on a certain variety caused by the compatible race of the fungus. Use and development of resistant varieties with true resistance genes and a high level of field resistance are the most effective measures to control blast disease. Field resistance of rice varieties harbouring true resistance genes can not be assessed in test fields in the absence of compatible races. Seedlings of the japonica rice varieties were inoculated at the fourth leaf stage by spraying with a spore suspension of compatible races. Inoculated seedlings were heated with an electric heater at night in the greenhouse. These inoculation and heating procedures have resulted in such a successful infection that it has become possible to determine accurately the relative resistance of the japonica rice varieties to blast disease.

Out to eighty-one indica rice varieties tested, nineteen varieties were resistant to all the isolates collected in the japonica rice-growing area in Yunnan Province. These indica varieties were divided into nine groups, on the basis of the reaction patterns to the Japanese isolates or the Yunnan isolates collected in the indica rice-growing area in Yunnan Province. It is assumed that these indica varieties harbour at least five true resistance genes to be identified yet.

Table1. Classification of the indica rice varieties based on reaction patterns to nine isolates of blast fungus

Group	Genotype*	Isolates (race) Y90-13 (037)	Y90-9 (007)	Y90-71 (102)	Y90-48 (001)	Y90-73 (114t)	TH74-9 (177)	TH81-02-3 (137b)	88A (433)	TH81-04 (437)	Varieties
I		S	S	–	–	–	–	–	–	–	Xihonggu
II	A	–	S	–	–	–	–	–	–	–	Shanyanggu, Luozagu, Jiuyuehuangpigu, Jiuyuegu (1)
III	A	–	S	–	–	S	–	–	–	–	Shanhuagu
IV	B · C	S	–	–	–	–	–	–	–	–	Jinchigu, Lazigu (2)
V	B	S	–	–	–	–	–	–	–	S	Haodonglang
VI	C	S	–	–	–	–	S	–	–	–	Xiajiubaigu
VII	E	S	S	–	–	–	–	–	–	–	Yuanjianbaigu, Jianchigu, Luopinger, Matigu, Landigu, Xiaomangzhong
VIII	D	S	–	S	–	–	–	–	–	–	Haomenglai, Taixianyihao
IX	E	S	S	S	–	–	–	–	–	–	Honzaogu

S: Susceptible reaction, –: Resistant reaction, *: Unknown genotype

Research Period: 1990–92
Research Site: Okinawa Branch, Tropical Agriculture Research Center

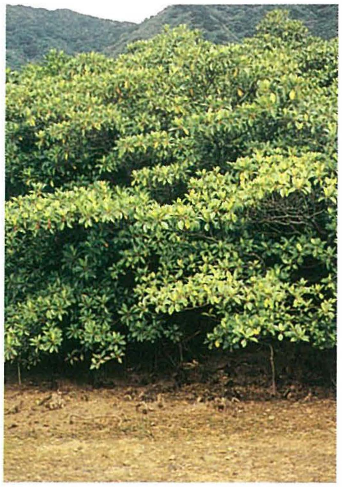


Fig. 1.
Bruguiera gymnorrhiza

Fig. 2.
Natural *Rhizophora stylosa* and planted *R. stylosa* on Ishigaki Island

Optimum Salinity Level in Relation to Photosynthetic Rate in Mangroves

Shigeki FURUYA
Okinawa Branch, Tropical Agriculture Research Center

Mangrove swamp forests are distributed in river mouths and bays in the subtropical zone of Japan. *Bruguiera gymnorrhiza* (Fig.1) and *Rhizophora stylosa* (Fig.2) are the dominant species in Yaeyama islands, while *Kandelia candel* is dominant in Okinawa Islands, Okinawa Prefecture. The other species of mangrove in Japan are *Avicennia marina*, *Sonneratia alba*, *Lumnitzera racemosa*.

Recently, mangroves have been planted on the beach. However, the percentage of establishment of planted species is often low. Therefore, the effect of environmental conditions on the growth of mangroves needs to be investigated to increase the percentage of establishment of planted mangroves. Factors such as wave hight, soil pH, salinity level, temperature may be related to the growth of mangroves. In this study, the effects of salinity level and temperature on the growth and photosynthetic activity of mangroves were studied.

Key words: mangrove, salinity, temperature, photosynthesis

Materials and Methods

Seedlings of *B. gymnorrhiza* and *R. stylosa* were planted in 1/5000a wagner pots filled with sand. The seedlings were cultivated under flooded conditions using fresh water for irrigation. When the plants reached a height of 40–50cm, salinity treatments consisting of control (fresh water), seawater (salinity level: 1), seawater with increased salinity level (salinity level: 1.5) and the following dilutions of seawater, 1/8, 1/4, 1/2 salinities, were initiated. Two weeks after the treatment, the photosynthetic rate, transpiration rate and stomatal conductance of leaves, were measured with a portable infra-red gas analyzer in a controlled glasshouse. Air temperature in the glasshouse was 25°C. Light intensity at the leaf surface was 450 μmol/m^2/sec. Effect of the temperature on the photosynthetic rate and stomatal diffusive conductance was analyzed with an infra-red carbonic acid gas analyzer and hygrometer in the laboratory. Light intensity at the time of the measurement was 1066 μmol/m^2/sec. Leaf length measurements were taken 19 days after salinity treatments.

Results and Discussion

Leaf length in *B. gymnorrhiza* was maximum at a 1/8 salinity level of seawater, and that of *R. stylosa* was maximum at the salinity level of fresh water (Fig. 3). Leaf length of both *B. gymnorrhiza* and *R. stylosa* decreased with the increase in the salinity level. Photosynthetic rate was maximum at a 1/8 salinity level of seawater in *B. gymnorrhiza* and that of *R. stylosa* was maximum at the salinity level of fresh water (Fig. 4). Photosynthetic rate decreased with the increase in the salinity level.

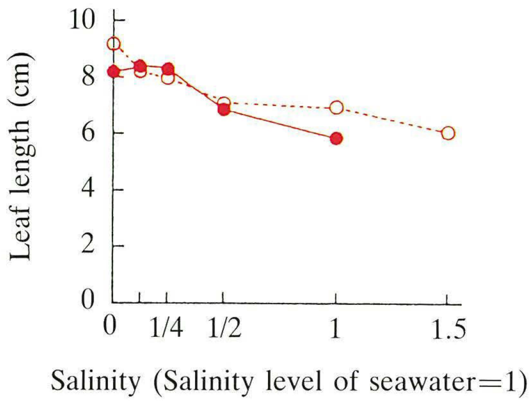


Fig. 3.
Effect of salinity level of culture solution on growth of leaves
● *Bruguiera gymnorrhiza*
○ *Rhizophora stylosa*

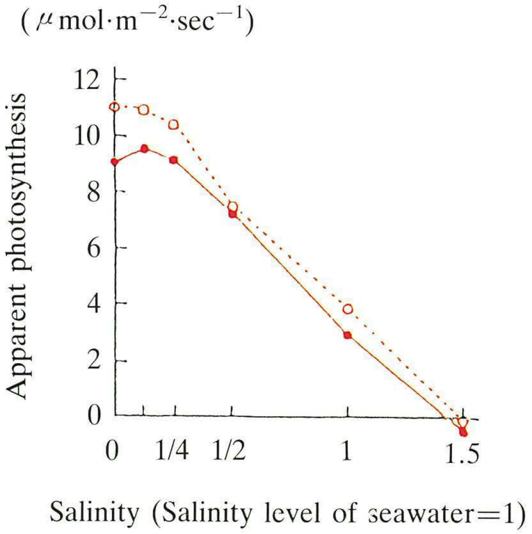


Fig. 4.
Effect of salinity level of culture solution on apparent photosynthesis
● *Bruguiera gymnorrhiza*
○ *Rhizophora stylosa*

Optimum temperature in relation to the photosynthetic rate in *B. gymnorrhiza* and *R. stylosa* was 25°C (Fig. 5). The decrease of the photosynthetic rate at a low temperature was less pronounced in *R. stylosa* compared with *B. gymnorrhiza*. Stomatal conductance started to decrease at a salinity level of more than 1/4 of seawater (Fig. 6), a low temperature below 20°C and a high temperature over 30°C (Fig.7). Closure of stomata is related to the decrease of the photosynthetic rate under high salinity, low temperature and high temperature conditions.

Although it is generally considered that strong winds and high waves control the growth of mangroves, it was found that a high salinity level also affects the growth of mangroves. Therefore, river mouths and coastal areas where subterranean water flows out, may be suitable for the planting of mangroves.

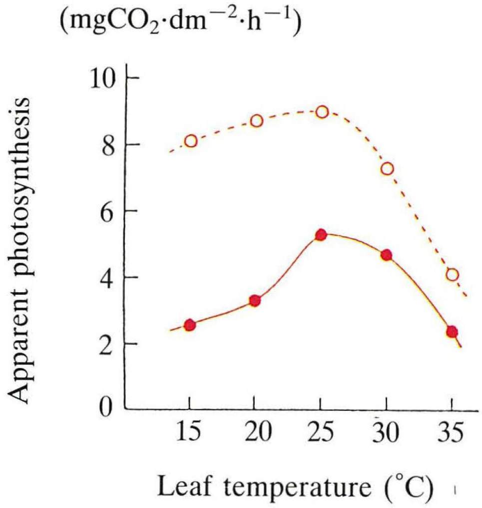


Fig. 5.
Effect of leaf temperature on apparent phtosynthesis
● *Bruguiera gymnorrhiza*
○ *Rhizophora stylosa*

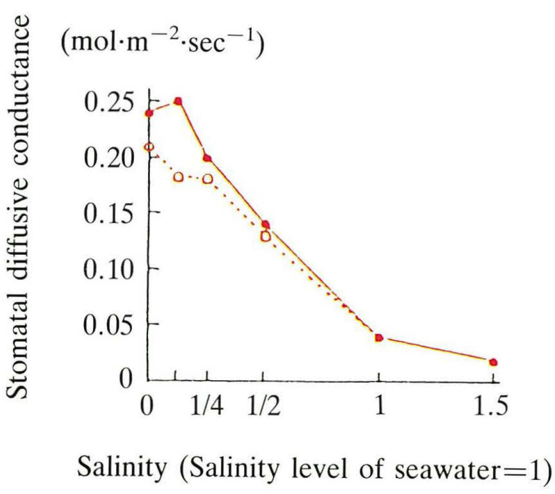


Fig. 6.
Effect of salinity level of culture solution on stomatal diffusive conductance
● *Bruguiera gymnorrhiza*
○ *Rhizophora stylosa*

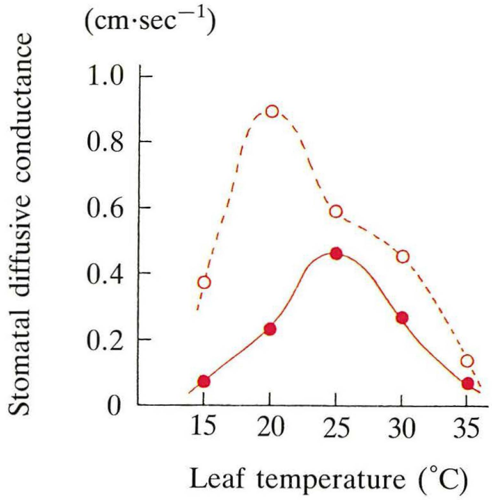


Fig. 7.
Effect of temperature on stomatal diffusive conductance

● *Bruguiera gymnorrhiza*
○ *Rhizophora stylosa*

References

1) Furuya, S. (1992): Effects of salinity on growth, photosynthesis, stomatal diffusive conductance in mangrove. Japan J. Trop. Agr. 36 Extra issue 2, 12–13.

Nitrogen Fixation of Mungbean (*Vigna radiata*) in Thailand

Research Period: 1988–91
Research Site: Department of Agriculture, Thailand

Toshifumi MURAKAMI[1)], Settha SIRIPIN[2)], Precha WADISIRISUK[1)], Nantakorn BOONKERD[1)], Tadakatsu YONEYAMA[3)], Tadashi YOKOYAMA[4)] and Hideo IMAI[4)]
1) Department of Agriculture, Thailand, 2) Kasetsart University, Thailand, 3) National Agriculture Research Center, 4) Tropical Agriculture Research Center

Fig. 1.
Mungbean grown in Tak fa area in central Thailand (pod-maturing stage)

Soils in the tropics are, in general, extremely infertile and crop productivity is very low. Improvement of productivity with the use of chemical fertilizers (especially nitrogen) is, however, very difficult due to economic factors in these areas. The effective use of the symbiosis between legume crops and *Rhizobium* bacteria may enable to achieve a high yield and decrease the cost of production under such conditions.

Mungbean which is a pulse crop characterized by an early maturity (60 days) is widely cultivated in tropical Asia due to the supply of easily digestible protein source. In Thailand, it is one of the most important crops for export. The current research project was initiated to maximize mungbean yield in Thailand through the utilization of the symbiosis. However, information about nitrogen fixation in mungbean is limited. As a first step of study, the changes in the nitrogen-fixing ability of mungbean with growth and other related parameters were analyzed.

Key words: Mungbean, Nitrogen fixation, ^{15}N natural abundance method, %Ndfa, ARA

Materials and Methods

Several varieties (lines) of mungbean (KPS 1, UT 1, VC 2768A etc.) were grown in concrete pots filled with Gray lowland soil or in an experimental field. The plants were periodically sampled to analyze the parameters related to nitrogen fixation. Mungbean plants in farmers' fields in the central region of Thailand were also collected.

The nitrogen-fixing activity was determined by the acetylene reduction assay. The cut roots bearing nodules were incubated in 10% acetylene gas in a 250ml volume flask and the amount of evolved ethylene gas was determined using a gas chromatograph equipped with a flame ionization detector. The contribution of nitrogen fixation was determined by the ^{15}N natural abundance method. The shoot/pod samples were pulverized in a mill, digested in heated concentrated sulfuric acid and steam-distilled. The evolved ammonia was collected in 1N-sulfuric acid, converted to nitrogen gas and the ^{15}N natural abundance was determined by mass spectrometry (Yoneyama, 1987). The contribution was expressed as %Ndfa (% nitrogen derived from air), which was calculated from the differences between the $\delta^{15}N$ value of N-fixing and neighboring non N-fixing plants as follows:

$$\%Ndfa = \frac{\delta^{15}N_{\text{non-fixing plant}} - \delta^{15}N_{\text{fixing plant}}}{\delta^{15}N_{\text{non-fixing plant}} - \delta^{15}N_{\text{fixing plant grown in air N}}} \times 100$$

where $\delta^{15}N$ is the ^{15}N natural abundance expressed as the ‰ ^{15}N excess to the atmospheric N_2.

Results and Discussion

The root nodules were formed at 6 to 7 days after sowing (DAS) and the number increased three fold (10 to 14, 30 to 40 and 55 to 60 DAS) during the growth of the plant regardless of nitrogen application (Fig. 2). In the absence of nitrogen treatment (N0), the nitrogen-fixing activity (ARA) appeared at 12 DAS, increased rapidly after flowering (34 DAS), reached a peak at 40 DAS and increased again at about 60 DAS (Fig. 4). The resumption of the increase agreed well with the increase of the nodule number. Similar phenomena had been reported in faba bean (Mukhtar et al., 1988) and lupin (Witty et al., 1988), although the physiological mechanism remained unknown. In the nitrogen application treatment (75 kgN/ha, N75), the activity appeared at 14 DAS and was maintained at a lower level (Fig. 5). The application of a large amount of nitrogen resulted in a decrease of the nodule number to 54%, nodule weight to 26%, nodule diameter to 72% and ARA to 19% compared with the absence of treatment.

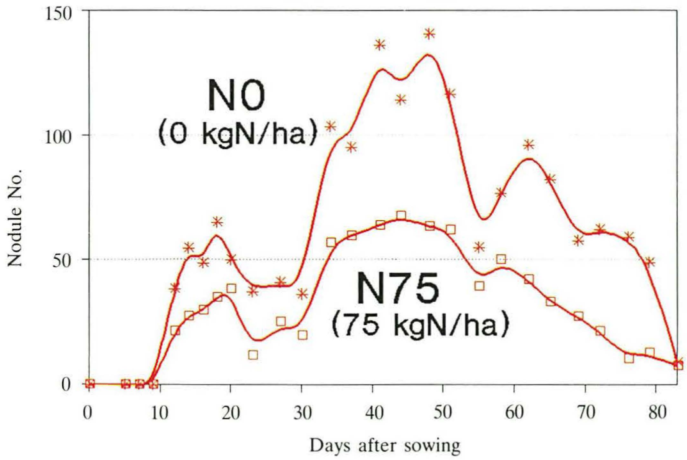


Fig. 2.
Changes in nodule number with plant growth in the pot experiment
Data were expressed as the average values of 3 varieties (lines); VC 2768A, VC 4000-7, VC 2768A/VC 1560D line 2.

Fig. 3.
Changes in %Ndfa with plant growth in the pot experiment
Data were expressed as average values of the 3 varieties (lines).

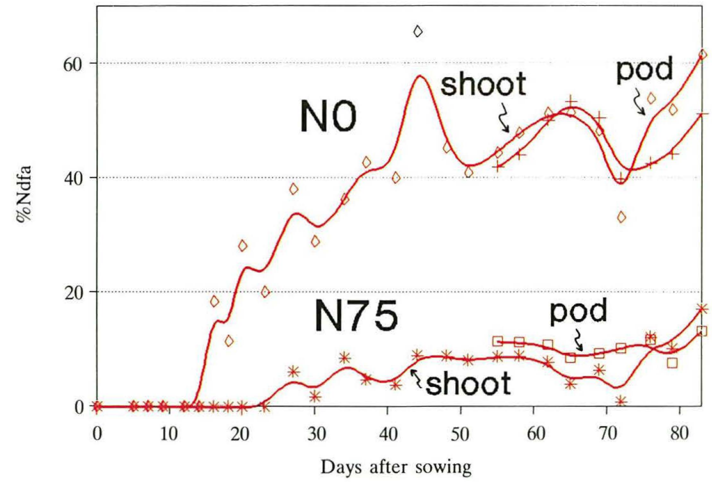


Fig. 4.
Nitrogen accumulation by mungbean in the absence of treatment
ARA; acetylene reduction activity, nfa; nitrogen derived from air, nfs; nitrogen derived from soil. Data were expressed as average values of the 3 varieties (lines).

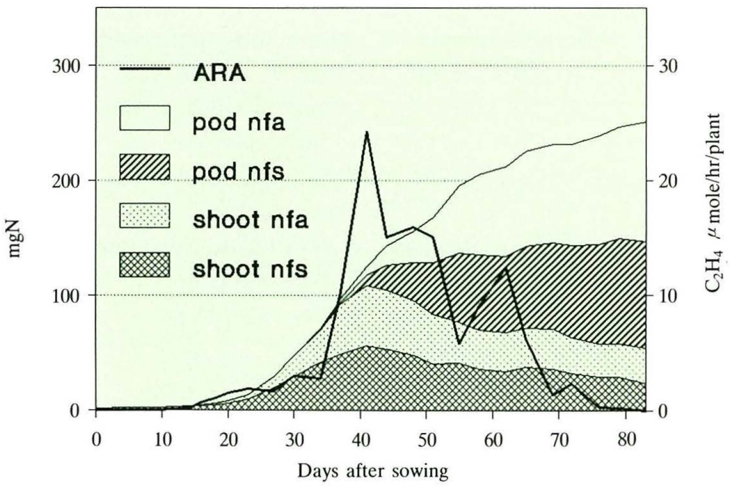
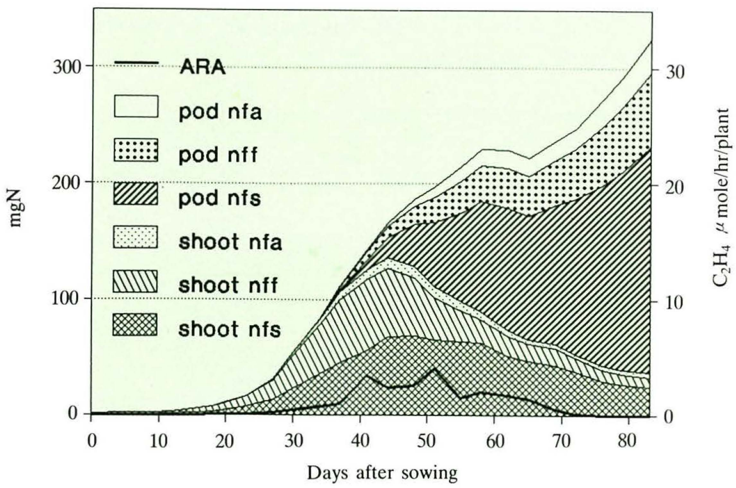


Fig. 5.
Nitrogen accumulation by mungbean in N75 treatment
ARA; acetylene reduction activity, nfa; nitrogen derived from air, nff; nitrogen derived from fertilizer, nfs; nitrogen derived from soil. Data were expressed as average values of the 3 varieties (lines).



The contribution of nitrogen fixation in mungbean (%Ndfa) increased rapidly after 16 DAS, reached a value of 40% at the flowering stage and ranged from 40 to 60% in the absence of treatment (Fig. 3). The major nitrogen source of the plant was soil (seed) nitrogen from germination to the flowering stage and thereafter both soil and air nitrogen (Fig. 4). In the N75 treatment, the contribution was less than 10% during plant growth (Fig. 3). The major nitrogen sources were fertilizer and soil nitrogen until flowering and thereafter soil nitrogen (Fig. 5). The part of stored nitrogen in the shoot was translocated to pods in both nitrogen treatments. The application of a large amount of nitrogen suppressed the absorption of air nitrogen, while fertilizer nitrogen was absorbed. However, the application of a small amount of nitrogen may enhance seedling growth, when nitrogen fixation has not yet been initiated. The application of "starter nitrogen" (19 kgN/ha, recommended amount in Thailand) was observed in the field. Although nitrogen fixation was not suppressed, shoot weight and grain yield did not increase in the 13 varieties examined. The %Ndfa of the 13 mungbean varieties at the pod-maturing stage (around 60 DAS) ranged from 40 to 60% and small differences were observed among some varieties at 5% significance level.

The mature mungbean samples (45-60 DAS) were collected from 36 farmers' fields. They bore very few nodules and the %Ndfa value was 23.2±18.1% on an average, i.e. 1/3 to 1/2 of the values recorded in the pot or field experiments described above. The indigenous *Rhizobium* bacteria in the farmers' fields may not be comparable to the cultivated varieties. The selection of *Rhizobium* bacteria suitable for the host plant is important for enhancing nitrogen fixation. Basic research involving the classification of the bacteria, especially in the tropics should be carried out for effective selection.

References

1) Murakami, T. et al. (1991): The nitrogen fixing ability of mungbean (*Vigna radiata*). In Proceedings of the Mungbean Meeting 90, p. 187–198, Bangkok.
2) Mukhtar, N.O. and Abu Naib, A. Sayda (1988): Biological nitrogen fixation by faba bean, lentil, and chickpea in the traditional versus the new areas of the Sudan. In Nitrogen fixation by legumes in Mediterranean agriculture. (Eds.) D.P. Beck and L.A. Materon. p. 331–344.
3) Witty, J.E. and Minchin, F.R. (1988): Measurement of nitrogen fixation by the acetylene reduction assay; Myths and Mysteries. ibid. p. 331–344.
4) Yoneyama, T. (1987): N_2 fixation and natural ^{15}N abundance of leguminous plants and *Azolla*. Bull. Natl. Inst. Agrobiol. Resour. 3, 59–87.

Research Period: 1990–92
Research Site: Tropical Agriculture Research Center

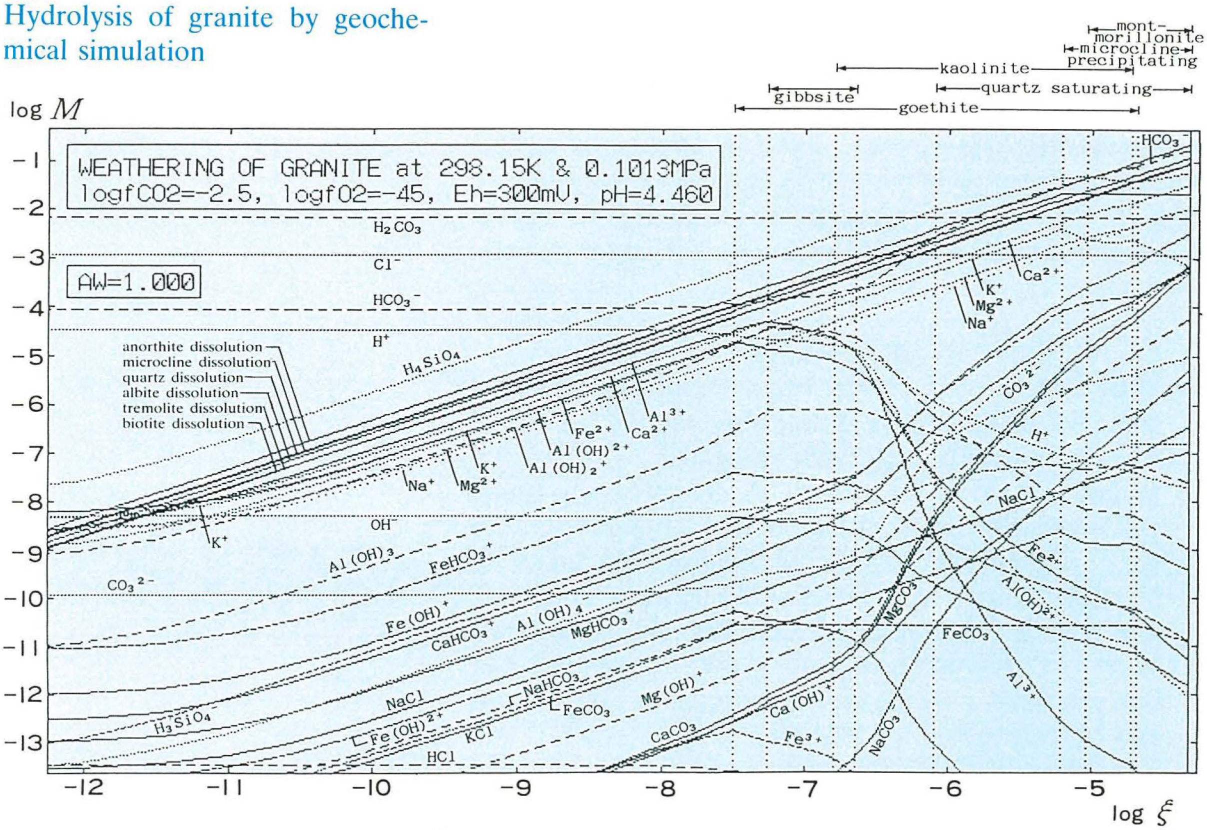
Mechanism of Rock-Weathering in Tropical Dry Areas

Tamao HATTA
Tropical Agriculture Research Center

The mechanism of rock-weathering in tropical dry areas is important in studies on desertification. The present studies dealt with the changes in the chemical and physical properties of standard rocks during experimental weathering. The formation of secondary minerals and the behavior of chemical species in aqueous solution were studied by geochemical simulation which is an analytical method of rock decomposition. The physical weatherability of rocks as a cause of rock disintegration can be determined by the relation between the TMA (Thermo-Mechanical Analysis) -values and pore ratio.

Key words: weathering, dry area, geochemical simulation, thermo-mechanical analysis

Fig. 1.
Hydrolysis of granite by geochemical simulation



Chemical Weathering

The process of chemical weathering can be simulated as a reaction occurring between rock-forming minerals and an aqueous solution[1]. The changes in the amount (grams) of chemical species/1000g of solution (M) and the degree of saturation of secondary minerals, shown in Fig. 1, were calculated as increments of the progression of the reaction (ξ) in the hydrolysis of standard granite as an example. The changes in the values of the chemical parameters were complex. The formation of saturated mineral zoning by weathering is represented by the reaction area ($10cm^2$) in the profile (Fig. 2). Satisfactory results were obtained in the alteration to laterite.

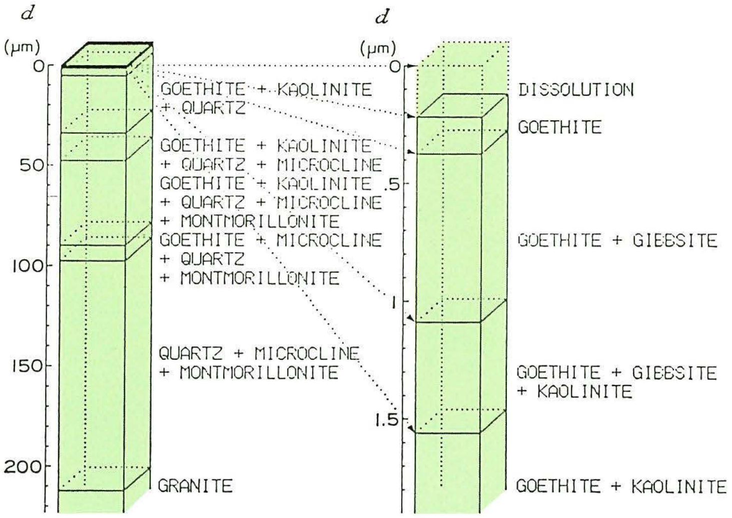


Fig. 2.
Mineral zoning in simulated weathering profile
d: depth from reaction surface

Physical Weathering

Physical weathering takes place when rocks on the earth surface are broken into fragments or grains. Weathering by thermal expansion only occurs under very hot and very dry climatic conditions. The behavior of thermal expansion and shrinkage of 14 standard rocks* in the temperature ranges of 0 to +100°C and −125 to +550°C was examined by TMA**[2]. The rocks characterized by a large thermal expansion contain a large amount of quartz. The samples of granite were fractured during the experiment (Fig. 3). A new stability series of physical rock-weathering can be determined based on the thermal expansion-shrinkage coefficient and the pore ratio*** in each rock type (Fig. 4). This order reflects the geomorphological characteristics in a desert area.

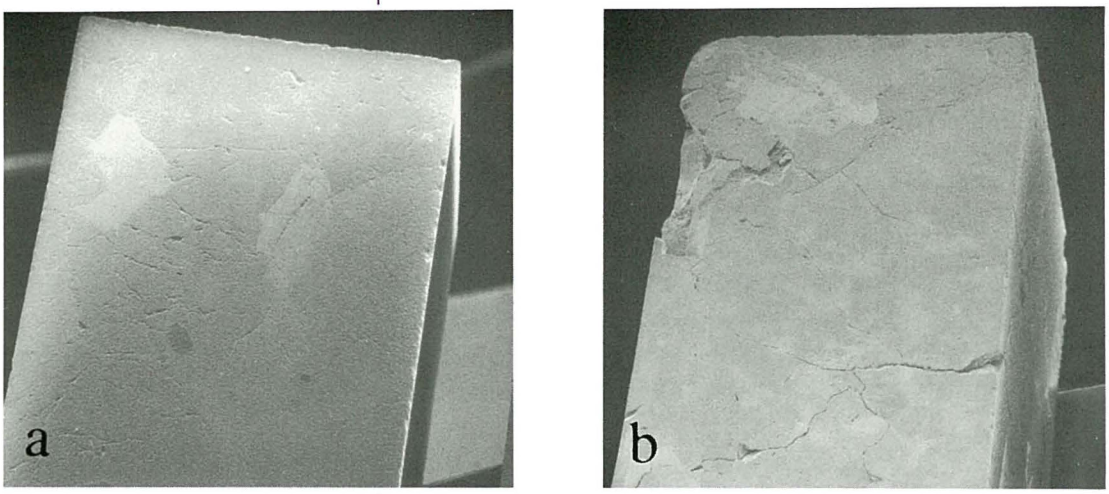


Fig. 3.
SEM photographs showing the onset of granite(a) and disintegration of granite by TMA (b)

Weathering Process in Tropical Dry Areas

Although there is no natural water at present, many hydrous minerals (e.g. goethite, gibbsite, kaolinite etc.) occur on the earth surface in a desert area. The formation of these secondary minerals can be explained by geochemical simulation that describes the water-rock interaction. As

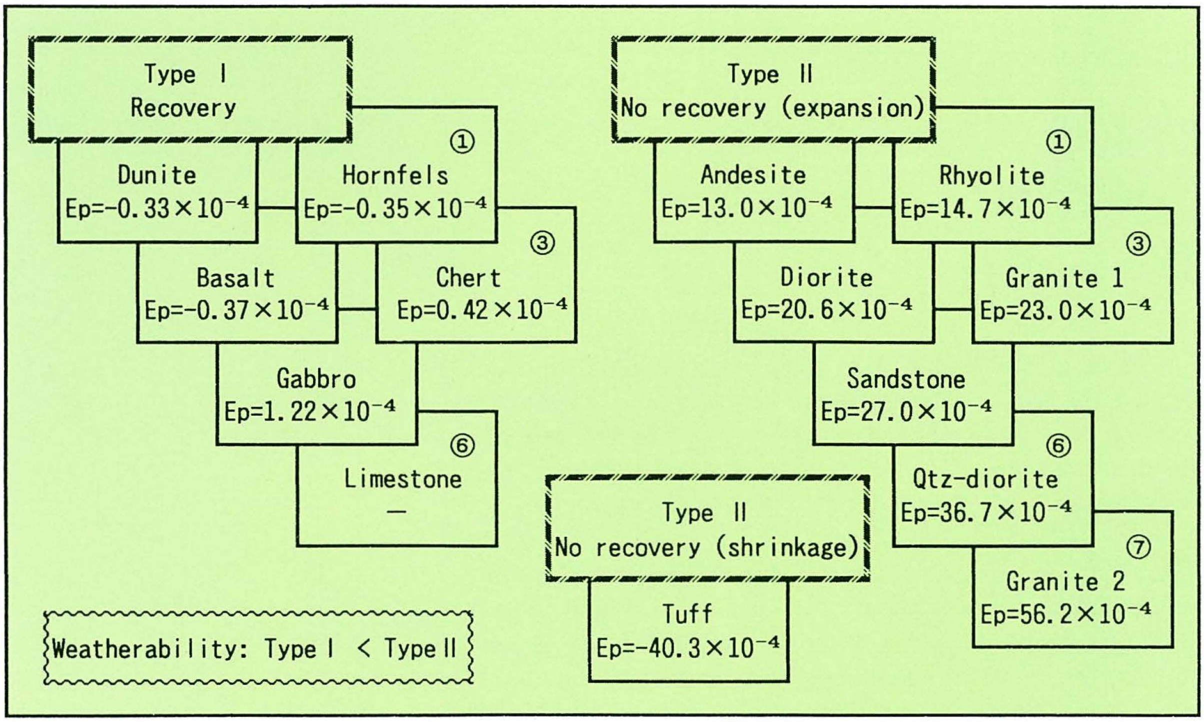


Fig. 4.
Rock stability series in physical weathering

natural water is essentially involved in soil genesis, it is considered that the secondary minerals forming soils in a tropical dry area have not been formed under the present climatic conditions.

The rock stability series in physical weathering indicates that the rocks characterized by a large thermal expansion are not prone to weathering, because the pores in rocks play an important role in the alleviation of various environmental stresses. The importance of the thermal expansion coefficient of rocks on the earth surface in tropical dry areas has been well documented. The physical weatherability of rocks can be studied by the determination of thermal coefficients and the physical properties of each rock type.

References

1) Hatta, T. (1991): Evaluation of physical properties of weathering products by geochemical simulation. J. Chem. Soc. Jpn. 1991, 509–513.
2) Hatta, T. et al. (1992): The expansion-shrinkage behavior of rocks by TMA over the range of 0 — +100°C and −125 — +550°C. Abst. 29th Inter. Geol. Cong. 3, 712.

* Samples of standard rocks (3.54mm × 3.54mm × 20.00mm) were cut from large blocks, and then the surfaces were polished by abrasives (#100).

** The TMA-values were obtained at a heating rate of $5^{\circ}C min^{-1}$ and holding time of 20 min at maximum temperature with 5 cycles.

*** $Ep = \alpha (1 + e)^{-1}$, Ep: index of physical weathering, α: thermal expansion-shrinkage coefficient, and e: pore ratio.

Research Period: 1990–92
Research Site: Okinawa Branch, Tropical Agriculture Research Center


Photo 1.
Ringspot on papaya fruit caused by PRSV-P

Detection of Two Plant Viruses of Papaya by ELISA

Tetsuo MAOKA and Tomio USUGI
Okinawa Branch, Tropical Agriculture Research Center

Papaya (*Carica papaya* L.) is one of the most widely grown and economically valuable fruits in the tropics and subtropics. This large herbaceous, dicotyledonous plant with a single stem and a crown of large, palmate and lobed leaves grows fast and produces fruits 8–10 months after being transplanted in the field. The extensive adaptation of the plant and the wide acceptance of its fruits offer a considerable potential for local and export markets. Like banana, pineapple and mango, papaya is one of the important cash crops in the tropics and subtropics.

The presence of a destructive disease caused by papaya ringspot virus (PRSV-P) is the main obstacle to wide-scale planting of this fruit tree. PRSV-P is as a major limiting factor for growing papaya in Hawaii, Florida, the Caribbean countries, South America, Africa, Australia, Thailand, Malaysia and Taiwan. In papaya, the virus causes mosaic and distortion of leaves and ringspots on fruit. PRSV-P belongs to the potyvirus group, with flexuous, filamentous particles about 780 nm in size. It is transmitted mechanically and by many species of aphids in a non-persistent manner. PRSV-P has a narrow host range that includes species of three dicotyledonous families: Caricaceae, Chenopodiaceae and Cucurbitaceae.

In Japan, a virus-like disease was first observed in the northern part of Okinawa Mainland in 1954 and wihtin 6 years, the virus had destroyed most of the papaya trees on Okinawa Mainland, Miyako Island and Ishigaki Island. Initially, the virus was reported to be PRSV-P, since it caused mosaic with or without distortion or yellowing of leaves and ringspots on fruits. The virus belongs to the potyvirus group, with flexuous, filamentous particles about 800 nm in size. It was transmitted mechanically and by some species of aphids in a non-persistent manner and had a narrow host range that included species of Caricaceae and Cucurbitaceae. However, in 1987, since it was found that the virus had no serological relationship with PRSV-P, the virus was considered to be a new one and was designated as papaya leaf distortion mosaic virus (PLDMV). Thereafter it appeared that all the viral diseases on papaya were caused by PLDMV and that PRSV-P did not occur in Japan. However, the first Japanese isolate of PRSV-P was detected from papaya on Miyako Island in 1991 (Photo 1).

As PRSV-P and PLDMV belong to the potyvirus group and develop similar symptoms on papaya, it is very difficult to distinguish them. In this study, we attempted to develop a field survey system using ELISA. We prepared two antisera to PRSV-P and PLDMV and were able to distinguish both viruses using ELISA.

The studies were implemented under the Tropical Agriculture Research Program of TARC.

Key words: papaya, papaya ringspot virus (PRSV-P), papaya leaf distortion mosaic virus (PLDMV), ELISA

Materials and Methods

PRSV-P was obtained from a distorted papaya leaf with mosaic and ringspot symptoms on Miyako Island in June, 1991. PLDMV was obtained from a distorted leaf of papaya with mosaic symptoms on Ishigaki Island in April, 1990. These isolates were inoculated to either *Cucumis metuliferus* or papaya. In the case of PRSV-P, the virus propagated after three cycles of local lesion isolation using *Chenopodium quinoa*. Antisera were produced by rabbits through injections with purified viruses. Double-antibody-sandwich enzyme-linked immunosorbent assay (DAS-ELISA) was performed according to the method of Clark and Adams (1984). In the field investigations, each sample was homogenized with 20 fold (v/w) PBS-T. The ELISA absorbance values at A^{405} were measured with a Microplate Reader Model 450 (BIO RAD).

Results and Discussion

The usefulness of the two kinds of antisera was evaluated with DAS-ELISA. Fig. 1 shows that, the assays were very sensitive and enabled to detect PRSV-P and PLDMV in crude extracts from infected papaya plants at dilutions of 10^3. The ELISA values of healthy papaya leaves were very low.

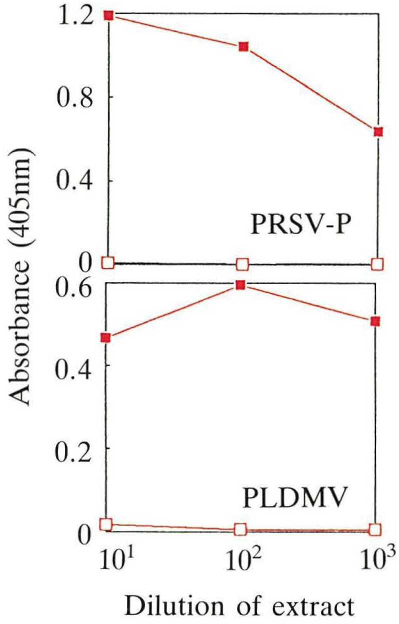


Fig. 1.
Detection of PRSV-P and PLDMV in crude extracts of papaya plants by ELISA

■; infected, □; healthy.

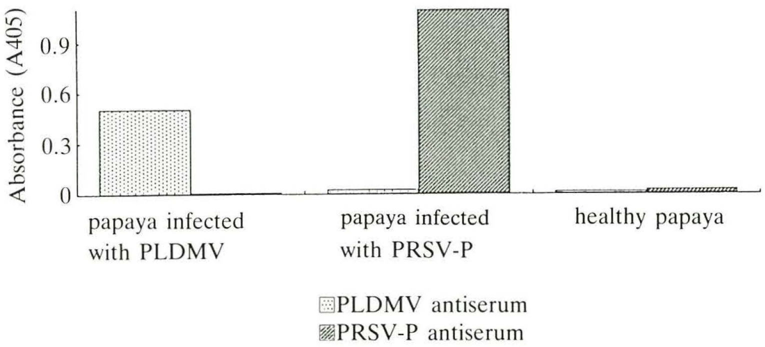


Fig. 2.
Detection of virus antigens in crude extracts of papaya plants by ELISA using PRSV-P antiserum and PLDMV antiserum

Table 1.
Distribution of PLDMV and PRSV-P on Ishigaki Island

PLDMV	PRSV-P	Double infection	Other virus*	Negative	Total
88	7	1	3	1	100

* Negative reaction by ELISA, but virus particles were observed by electron microscopy.

Specific responses of each antiserum were determined by comparison of homologous and heterologous reactions. As shown in Fig. 2, the PRSV-P antiserum reacted with PRSV-P and did not react with PLDMV by ELISA. PLDMV antiserum recognized only the homologous virus. There were no heterologous reactions between the two viruses. These results indicate that the ELISA system enables to detect the viruses in the field.

In small scale examinations by crude sap inoculation to *C. amaranticolor*, PRSV-P was not detected on Ishigaki Island. We, then, collected 100 papaya samples in fields on Ishigaki Island and subjected them to ELISA. Table 1 shows that PLDMV was detected from 88 plants, PRSV-P from 7 plants and both viruses from one plant. Although most of the viruses on papaya on Ishigaki Island consisted of PLDMV, PRSV-P could also be detected even in a low population.

Further studies should be carried out to analyze the epidemiology of the two viruses. Although we could very easily detect and distinguish the two viruses in field-grown papaya by ELISA, we were not able to differentiate the viruses by merely observing the symptoms induced on papaya.

References

1) Maoka, T. and Usugi, T. (1992): Comparisons of symptoms on some papaya cultivars inoculated with papaya virus isolates from Ryukyu Islands and Amami-Oshima, and studies on the serological diagnosis. Ann. Phytopathol. Soc. Japan 58, 623 (in Japanese).
2) Maoka, T. and Usugi, T. (1993): Type P isolate of papaya ringspot virus (PRSV-P) and papaya leaf distortion mosaic virus (PLDMV) isolated from Ryukyu Island. Ann. Phytopathol. Soc. Japan 59, 335 (in Japanese).

Comparison of Biotypes of Brown Planthopper Populations Collected in the Indochina Peninsula and Japan.

Takashi WADA[1)] and Kiyomitsu ITO[2)]
1) Kyushu National Agricultural Experiment Station, Japan,
2) Tropical Agriculture Research Center

Research Period: 1992
Research Site: Tropical Agriculture Research Center and Kyushu National Agricultural Experiment Station, Japan

Rice planthoppers, *Nilaparvata lugens* (BPH) and *Sogatella furcifera*, which became a threat to rice production in Japan and the Asian countries, do not hibernate in Japan. They invade Japan in June and July, every year from South China due to the monsoon. Chinese entomologists also showed that there is a large number of immigrants annually in April and May in South China from countries further south. The local planthopper population, thereafter, in South China originates from these immigrants. Thus, the origin of the Japanese planthoppers may be traced to some areas in the Indochina Peninsula. In this paper we compared the biotypes (virulence of BPH populations on resistant rice varieties) of the BPH populations including those from Indochina. The objective of this study is to determine the origin of Japanese BPH.

Key words: *Nilaparvata lugens*, biotype, migration, resistant rice variety, Indochina, Vietnam

Fig. 1.
Brown planthopper, *Nilaparvata lugens*

Materials and Methods

Eight BPH populations were collected in 1992 in the rice fields of the Indochina Peninsula and in Japan: four populations in the fields of the Muda area, Malaysia, Central Plain, Thailand, Mekong Delta, Vietnam (two populations) in the tropics, two populations in the fields of the Red River Delta, Vietnam in the subtropics, and two populations in the fields of Kyushu in the temperate zone of Japan. The insects used for the experiment were basically newly-emerged female macropters from the progenies reared for a few to several generations. Half of the planthoppers used were brachypters for the Malaysian population. The standard rice varieties used were TN1 (susceptible check), Mudgo (resistance gene: *Bph1*), IR26 (*Bph1*), ASD7 (*bph2*), Rathu Heenati (*Bph3*) and Babawee (*bph4*). We measured the amount of honeydew excreted on rice by an insect during 48 h using the parafilm sachet method. Since the resistance of these varieties is caused by the inhibition of planthopper feeding, the quantity of honeydew which reflects the feeding activity can be used as an index of the virulence of the insect on rice.

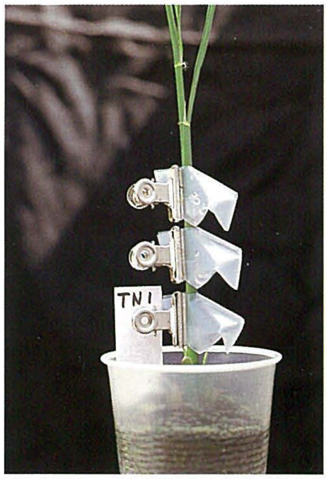


Fig. 2.
Parafilm sachets used to collect the honeydew excreted by a planthopper

Results and Discussion

The mean amount of honeydew excreted for 48 h by a female adult on TN1 ranged from 23.6 to 61.8 mg, mainly depending on the wing forms and rice stages in the experiment (Table 1). Therefore, we calculated the relative excretion index (REI) which indicates the mean amount of the excretion on a resistant variety relative to the mean of the excretion on TN1 (=100) to compare all the populations on a same scale.

Table 1. Relative excretion index (REI)[1)] of honeydew excreted by a female macropter on five standard rice varieties

Population	Insects used	Mean[2)] on TN1 (mg)	REI on resistant rice varieties (resistance genes)					
			TN1 (none)	Mudgo (Bph1)	IR26 (Bph1)	ASD7 (bph2)	Rathu heenati (Bph3)	Babawee (bph4)
Kyushu A	48	23.6	100	3.2 a	32.8 ab	10.6 a	4.5 ab	6.9 ab
Kyushu B	54	30.7	100	13.9 ab	34.8 ab	15.6 a	3.5 b	4.6 ab
Red River Delta A	58	24.5	100	21.0 b	51.6 abc	26.8 a	2.8 ab	4.0 a
Red River Delta B	53	27.7	100	4.7 a	41.8 ab	13.6 a	0.5 a	2.8 a
Mekong Delta A	52	32.5	100	21.2 ab	59.7 bc	72.9 b	8.5 b	20.1 b
Mekong Delta B	54	32.3	100	19.1 ab	78.7 c	97.1 b	6.1 ab	9.3 ab
Thailand	57	26.5	100	11.1 a	58.1 bc	75.0 b	2.7 ab	3.4 ab
Malaysia[3)]	56	61.8	100	4.3 a	20.1 a	75.3 b	0.5 a	6.4 ab

1) Mean of the amount of honeydew excreted on a resistant rice variety relative to the mean of the honeydew amount excreted on TN1 (=100).
2) Mean amount of honeydew on TN1.
3) Approximately half of the insects used were brachypters in the population.

Eight BPH populations were classified into two groups according to the virulence on ASD7. It was moderately resistant to the Japanese and Red River Delta populations. The REIs ranged from 10.6 to 26.8 for the populations collected in temperate and subtropical fields. On the other hand, the REIs on ASD7 ranged from 72.9 to 97.1 for the Malaysian, Thai and Mekong Delta populations indicating that the resistance of ASD7 entirely broke down in the tropical fields of the Indochina Peninsula.

IR26 was moderately susceptible to all the populations except for the Malaysian one. The REI values were slightly higher for the Thai and Mekong Delta populations than for the Japanese and Red River Delta populations. The rate of the planthoppers which excreted more than 10 mg (another index reflecting the biotype properties of the BPH population) was significantly higher for the Thai and Mekong Delta populations, too. Therefore, the virulence of these populations on IR26 appears to be slightly higher than that of the Japanese and Red River Delta populations. Although the Malaysian population is a tropical population, it was less capable of feeding on IR26 and Mudgo which have the *Bph1* resistance gene.

Rathu Heenati and Babawee still remained highly resistant to all the populations except for the Mekong Delta populations. These populations contained a few percentages of planthoppers capable of attacking either variety.

As a result, the biotype of the Japanese populations bore a close resemblance to that of the Red River Delta populations. In the Red River Delta, the planthopper density increases every year from late April along with flowering of the spring crop (the 1st crop of the year) and the southwest monsoon becomes strong from May onwards. Therefore, the ecological and meteorological conditions are conducive to a mass exodus of the planthoppers from the Red River Delta in spring. Although further studies should be carried out, it is, however, likely that the planthoppers in the subtropics of the Indochina Peninsula are involved in the large migration system of planthoppers which extends to the temperate zone of East Asia.

Research Period: 1989–91
Research Site: Faculty of Forestry, Kasetsart University, Thailand

Analysis of Reflectance and Transmittance of Leaf and Diffused Light Conditions under Canopies of Fast-growing Trees in the Tropics

Kiyoshi NAKASHIMA[1)], B. THAIUTSA[2)], S. KAITPRANAET[2)] and M. JAMROENPRUKSA[2)]
1) Forestry and Forest Products Research Institute, Japan,
2) Faculty of Forestry, Kasetsart University, Thailand

In the tropics, forest deterioration has made rapid progress in the past decades due to excessive logging, shifting cultivation or forest clearing for agricultural production. Preservation of forest resources and reforestation of degraded forest lands are an important task in the tropical countries. Production of wood and food is, however, essential in those countries. These conflicting requirements, namely the need to preserve forests and increase food production, have to be met at the same time. Although agroforestry is considered to be only one of the techniques that could contribute to solving these problems in the tropics, ecological and physiological studies on the properties of trees and crops in agroforestry systems are limited. The quality and quantity of light energy affect the production of agricultural crops in agroforestry systems. Therefore it is very important to control the light conditions between trees or under the canopies of tree stands for the cultivation of crops in agroforestry systems. To analyze and estimate the actual lights conditions under fast-growing tree stands in the tropics, spectral reflectance and transmittance of light energy of leaves and light conditions between trees and under canopies of those species were studied during the collaborative research project entitled "Rehabilitation of degraded forest lands and agroforestry in the tropics" with Kasetsart University, Thailand.

Key words: light condition, fast-growing trees, tropical forest, agroforestry

Materials and Methods

Reflectance and transmittance of leaf were measured in the range from 300 to 1100 nm with a portable spectroradiometer (LI-1800, LI-COR). Photosynthetic photon flux density (PPFD, μmol/s/m^2), illuminance (IL, lux) and integrated light energy (IT, W/m^2) were calculated from these data. Leaves of fast-growing trees were collected at Ratchaburi Forest Experiment Station, Royal Forest Department (RFD) in Ratchaburi. Diffused light conditions under the canopy of several tree species were determined with the spectroradiometer (LI-1800) and photometers (Minolta T-1) in the Northeastern Reforestation Center in Sie Sa Ket, Sakaerat Re-afforestation Project, Sakaerat, Doi Angkhan Royal Project in Chiangmai and Ratchaburi Forest Experiment Station. The relative light intensity (RLI) was calculated from these data.

Results and Discussion

1) Characteristics of reflectance and transmittance of light energy of fast-growing tree leaves

Part of the solar radiation energy is reflected on the surface of a leaf and the rest is absorbed by the mesophyll cells of leaf (Fig. 1). The spectrum of transmitted light of a leaf is, therefore, very different from that of solar radiation. In fast-growing tree species, the absorbance of light energy of a leaf was very high for blue, green and red light, ranging from 400 to 700 nm wavelength which is called "photosynthetically active radiation (PAR)", but low for the far-red light in the range 700 to 1100 nm.

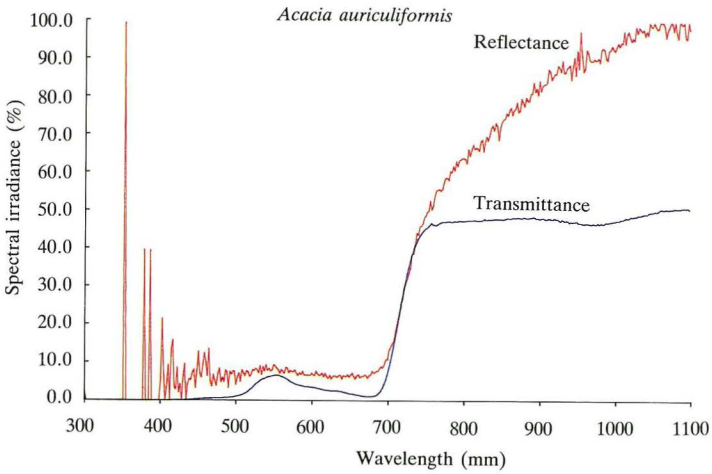


Fig. 1.
Spectral reflectance and transmittance of a fast-growing tree leaf

Transmittance of light energy of a leaf varies with its age, too. Transmittance was higher in a young leaf than in a mature or an old one for green and red light in the range from 500 to 700 nm but it was lower in an old leaf than in a young or a mature one for far-red light in the range from 700 to 1100 nm. These results were similar to those reported in other plant species.

Reflectance of light energy on the surface of a leaf was considerably high for far-red light but was very low in the range of PAR. However, the transmittance of a leaf was very low in the range of PAR and high for far-red light (Fig. 1).

As reflectance and transmittance on the surface of a leaf were significantly different among each tree species shown in Table 1, these leaf traits could become useful for differentiating various species.

Table 1.
Relative light intensity (RLI) under canopies, transmittance and reflectance of leaves, and opening of canopies in several tree stands

Species	RLI (%)	PPFD[1] Transmittance (%)	PPFD[1] Reflectance (%)	Opening of canopy[2] (%)
Melaleuca argentea	83.28	2.32	1.71	73.8
Eucalyptus exserta	76.44	3.41	4.01	36.5
Melaleuca symphyocarpa	75.34	3.54	6.54	63.0
Eucalyptus brassiana	71.07	6.30	2.03	52.5
Acacia leptocarpa	63.66	2.48	6.50	64.7
Eucalyptus bigalerita	57.07	4.73	2.18	50.2
Acacia auriculiformis	56.67	2.53	5.88	49.5
Acacia julifera	53.26	2.68	1.66	64.1
Acacia torulosa	52.56	2.30	2.01	54.6
Grevillea parallela	49.85	2.32	2.22	46.9
Acacia aulacocarpa	48.89	3.46	6.62	35.0
Grevillea pinnatifida	41.27	2.91	4.07	28.3
Eucalyptus tereticornis	39.00	8.48	2.06	27.4
Eucalyptus torelliana	33.20	6.44	1.56	28.7
Acacia polystachya	27.52	3.02	6.73	37.7
Petalostigma pubescens	25.79	3.01	1.92	29.4
Melia azedarach	21.06	4.12	6.46	25.4
Azadirachta indica	15.84	2.75	2.42	20.2
Peltophorum dasyrachis	8.00	1.89	0.85	15.2

1) Photosynthetic Photon Flux Density for PAR(400–700nm), figures show relative PPFD for control.
2) Relative area of an opening in a canopy.

2) Characteristics of light conditions under canopies of fast-growing trees

Under the closed canopy of fast-growing trees, most of the natural radiation (direct solar radiation and sky radiation) energy was reflected or absorbed by leaves of the canopy and only part of the radiation reached the ground. The diffused light under the canopy was very different from that of natural radiation and the spectrum exhibited approximately a constant energy at all wavelengths, whereas it varied remarkably at each wavelength for natural radiation (Fig. 2). The diffused light energy was extremely low for both the range of PAR and far-red light and was different in each tree species.

Light conditions under a canopy also varied with the tree age and the spacing of tree stands. In *Acacia auriculiformis* and *A. mangium* stands at 2 × 2m and 2 × 4m spacings, the relative light intensity (RLI) decreased rapidly to less than 20 % in three or four years after the trees were planted and the decrease of RLI continued gradually with tree growth subsequently. In the *A. leptocarpa* stands planted at 2 × 2m, 2 × 4m and 2 × 6m spacings, a large amount of energy of diffused light was observed at all wavelenghts in the stands with a wider spacing (Fig. 3).

Relative light intensity under the canopy was closely

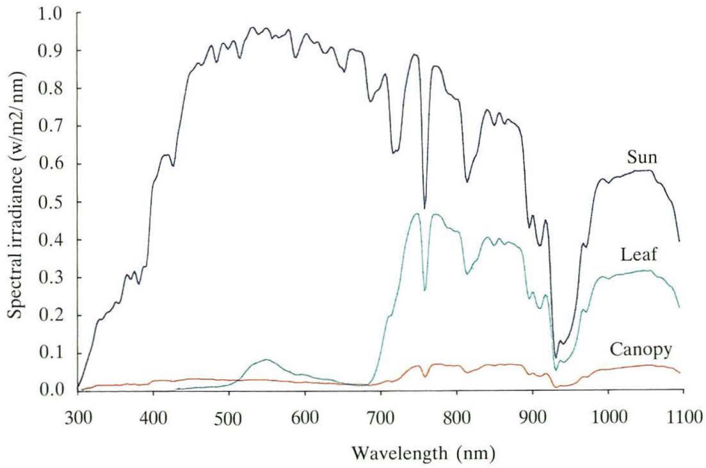


Fig. 2.
Spectral energy of direct sunlight, transmitted light of a leaf and diffused light under the canopy of *Acacia auriculiformis* stand
Sun: Direct sunlight, Leaf:Transmitted light of a leaf, Canopy: Diffused light under a canopy

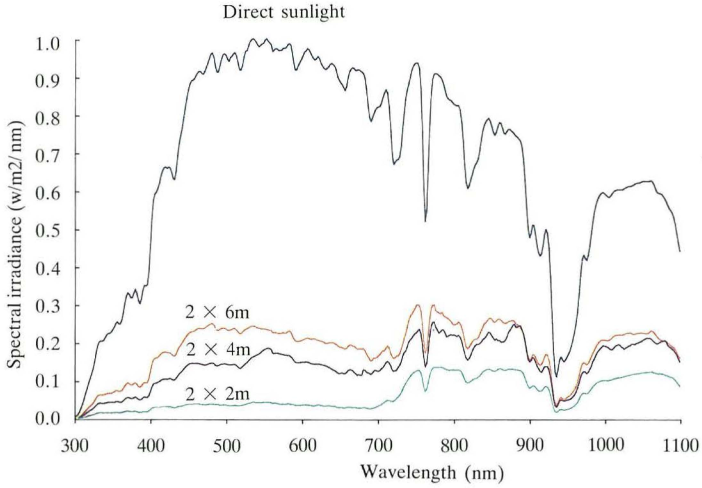


Fig. 3.
Difference in diffused light energy under canopies of *Acacia leptocarpa* stands at 2 × 2m, 2 × 4m and 2 × 6m spacings

related to the area of an opening in the canopy but not with the reflectance or transmittance in a leaf (Table 1). This fact suggests that the light conditions under a canopy mainly depend on the structure of the stand's canopy.

To estimate the light conditions between trees or under a canopy and to control them, further experimental studies on the crown structure in each tree species should be carried out.

References

1) Morikawa, Y., S. Sasaki and S. Asakawa (1976): Growth of pine and birch seedlings under light with different spectral compositions and intensities. J. Jpn. For. Soc. 58, 174–178.
2) Morikawa, Y., S. Sasaki and S. Asakawa (1980): Spectral composition of light on the forest floor and the growth of tree seedlings. Jpn. J. For. Environ. 25 (1), 1–7
3) Sato A. and R.V. Dalmacio (1991): Maize production under an intercropping system with fast-growing tree species: A case in the Philippines. JARQ 24, 319–356.

Fig. 4.
Hemispherical photograph of the canopy in a fast-growing tree stand.

List of Technical Bulletins of the Tropical Agriculture Research Center

No. 1. Plant-water relationship of indica rice in Malaysia (1971) by Katsuo SUGIMOTO

2. Maize in India - a review of literature (1971) by Haruo MIKOSHIBA

3. Nitrogen application technology for tropical rice as determined by field experiments using ^{15}N tracer technique (1973) by Takeo KOYAMA, Chittana CHAMMEK and Natee NIAMSRICHAND

4. Varietal difference of Thai rice in the resistance to phosphorus deficiency (1973) by Takeo KOYAMA, Chittana CHAMMEK and Patoom SNITWONGSE

5. Effect of transplanting time on rice plant growth and grain yield in Thailand (1973) by Takeo KOYAMA and Chittana CHAMMEK

6. Studies on ecology and control of the rice gall midge in Thailand (1974) by Terunobu HIDAKA, Precha VUNGSILABUTR and Sawang KADKAO

7. Studies on rice yellow orange leaf virus disease in Thailand (1975) by Toshihiko HINO, Luasark WATHANAKUL, Nopporn NABHHEERONG, Preecha SURIN, Ubol CHAIMONGKOL, Somkid DISTHTAPORN, Methie PUTTA, Dara KERDCHOKCHAI and Arunee SURIN

8. Chilli virus diseases in Sri Lanka (1975) by Miyoji SUGIURA, C.H.HEMACHANDRA and G.H.HEMACHANDRA

9. Varietal differences of photosynthetic efficiency and dry matter production in Indica rice (1976) by Yoshikazu OHNO

10. Forest and forest products pest problems in the Philippines (1977) by Katsura MORIMOTO and Romeo S.RAROS

11. Some pathogens of Cercosporiosis collected in Brazil (1978) by Toshihiko HINO and Hasime TOKESHI

12. (1) The prevalence of maize and soybean diseases in Thailand (1979) by Takehito SUZUI and Thanawatt KAMPHANGRIDTHRONG
(2) *Phytophthora* spp. isolated from some economic plants in Thailand (1979) by Takehito SUZUI, Ubol KUEPRAKONE and Thanawatt KAMPHANGRIDTHRONG

13. Studies on microflora in tropical paddy and upland farm solis (1979) by Michio ARAGAKI, Banharn TANGCHAM, Wisit CHOLITKUL and Samnao PHETCHAWEE

14. Farm management studies (1981) by Masanobu YAMASHITA, WONG Hin Soom and S.JEGATHEEAN

15. The brown planthopper, *Nilaparvata lugens* (Stal) (Homoptera, Delphacidae) at the IRRI, the Philippines (1982) by Kazushige SOGAWA

16. Studies on the control of downy mildew disease of maize in tropical countries of Asia (1983) by Haruo MIKOSHIBA

17. Phosphate response of rice in Indonesian paddy fields (1984) by Masanori MIYAKE, M. ISMUNADJI, Iskandar ZULKARNAINI and Sismiyati ROECHAN

18. Improvement of weak foundation of paddy fields by subsurface, drainage system field studies in the Muda irrigation project area (1984) by Tsuneo YAMASHITA

19. Study on soil productivity of paddy fields in Thailand (1984) by Satoru MOTOMURA, Ashara SEIRAYOSAKOL and Wisit CHOLITKUL

20. Multiple cropping on paddy field in Thailand (1986) by Sadao HATTA

21. Virus diseases of rice and legumes in the tropics (1986) by Toshihiro KAJIWARA and Shoshin KONNO

22. Salt tolerance of *Atriplex nummularia* (1987) by Yasutaka UCHIYAMA

23. Screening and genetic analysis of wide compatibility in F_1 hybrids of distant crosses in rice, *Oryza sativa* L. (1987) by Hiroshi IKEHASHI and Hitoshi ARAKI

24. Characteristics and genesis of volcanic ash soils in the Philippines (1988) by Hiroo OTSUKA, Aurello A. BRIONES, Nonilona P. DAQUIADO and Fernando A. EVANGELIO

25. Soils of northeast plateau, Thailand (1989) by Masanori MITSUCHI, Pichai WICHAIDIT and Saeree JEUNGNIJNIRUND

26. Analysis of light climate under the canopy of a man-made forest in the tropics (1989)by Yoichi KANAZAWA, Shozo NAKAMURA and Roberto V. DALMACIO

27. Management of irrigation systems for rice double cropping culture in the tropical monsoon area (1990) by Yoshinobu KITAMURA

28. Genetic diversity and landrace differentiation of mungbean, *Vigna radiata* (L.) Wilczek, and evaluation of its wild relatives (the subgenus *Ceratotropis*) as breeding materials (1991) by Norihiko TOMOOKA

Research Highlights '92

September 1992

Tropical Agriculture Research Center
Ministry of Agriculture, Forestry and Fisheries
1-2, Ohwashi, Tsukuba, Ibaraki, 305 Japan