# Promotion of Ripening of Rice Plants by 3-Hydroxy-5-Methyl-Isoxazole under Low Temperature Conditions

### By YASUO OTA

Department of Physiology and Genetics, National Institute of Agricultural Sciences (Yatabe, Ibaraki, 305 Japan)

### Introduction

In the Tohoku and other important rice growing districts in northern Japan the rice crop suffers cool-weather damage about every three years. In these cool-weather districts, therefore, the stability of rice crop depends on how to combat with cool-weather damage.

3-Hydroxy-5-methyl-isoxazole (on the market with the trade name of Tachigaren) is already known to promote growth of rice plant roots, enhance their physiological activity and facilitate healthy growth of seedlings. Recently attention has been focussed on its action of promoting rice ripening at low temperatures.<sup>1,2,3)</sup> This report reviews results of a large number of experiments conducted by the author and many others to confirm the function of Tachigaren to promote ripening of rice plants.

# Discovery and confirmation of the function

In 1971 the cool-weather damage of growth delay type occurred. The heading of rice plants was delayed about 10 days. The weather was cool during the ripening period. In that year the cool-weather attacked rice plants not only in the northeastern parts of Japan but also in the mountaneous areas at the center of Japan. Unripened rice plants remained green and unharvested till late.

It happened in that year that, in an attempt to make sure if Tachigaren which promotes growth of rice seedlings would prevent the outbreak of sheath blight, a farmer applied Tachigaren to rice plants at the heading stage which had been delayed one week as compared with a normal year. Rice plants in the neighboring fields were unripe, while he found out that the ears of his rice plants treated by Tachigaren were yellow ripe. He harvested his treated crop and found that the yield was more than 20% higher than that of the untreated crop.

To confirm the reproducibility of this result tests were performed by the author in a green house for three years from 1977 through 1979. A rice variety "Nihonbare" was grown in pots, and at the heading stage a 1:500 dilution of Tachigaren liquid was applied to the rice plants. The pots were moved into a temperature-controlled green house kept at 20°C during the day time and 13°C during the night to simulate the weather condition which causes cool-weather damage to rice plants. Under this condition the effect of Tachigaren on rice ripening was studied in detail.

The observations obtained in three years are summarized in Table 1. Grain fertility of rice grown in the control plot was reduced considerably due to the low temperature. In the treated plot with Tachigaren, on the other hand, the fertility was 6 to 11% higher than that of the control plot.

#### 1) Position of rice grains on the panicle and grain ripening

The time of blooming of glumous flowers is

| Year | Plot    | Fertile grains/<br>panicle | Sterile grains/<br>panicle | Total grains/<br>panicle | Fertility<br>rate (%) |
|------|---------|----------------------------|----------------------------|--------------------------|-----------------------|
| 1077 | Control | 51                         | 12                         | 64                       | 84                    |
| 1911 | Treated | 53                         | 8                          | 62                       | 87                    |
| 1079 | Control | 13                         | 59                         | 72                       | 17                    |
| 1970 | Treated | 23                         | 58                         | 81                       | 28                    |
| 1979 | Control | 33                         | 25                         | 58                       | 57                    |
|      | Treated | 40                         | 22                         | 61                       | 65                    |

 Table 1. Effect of Tachigaren treatment on rice fertility under low temperature conditions

Note: 1977 Three hills, each composed of two seedlings/pot (1/2000 a). Three pots were assigned to each plot.

1978 A hill composed of two seedlings/pot (1/5000 a).

Five pots were assigned to each plot.

1979 Twenty seeds were sown roundly in a 1/5000 a. pot, and four pots were assigned to each plot.

A 1:500 dilution of Tachigaren liquid was sprayed over rice plants at the heading stage. Then all test plants were grown in a green house ( $20^{\circ}$ C during the day time and  $13^{\circ}$ C during the night).

related to the position of glumous flowers on a panicle. There is a difference of about one week in blooming time between the glumous flower which blooms first and that which blooms last. Ripening of rice grains is associated with the order of blooming.

Fig. 1 indicates the relationship between weight of each grain and the position of each grain on a panicle, determined with rice plants grown under both natural (normal) condition and cool weather condition in a temperaturecontrolled green house kept at 20°C during the day time and at 13°C during the night.

It is apparently shown in Fig. 1 that it is not all grains but only inferior grains that suffer physiological damage due to cool weather. The inferior grains are known to be located at definite positions of a panicle, and susceptible to adverse conditions. They usually bloom late. Under the cool weather, growth of inferior grains is remarkably delayed so that they do not reach full-ripe even at the end of the growing season. Accordingly, in studying the effect of the chemical, examination of inferior grains can reveal more accurate information than the observation of the whole ears. An example is given in Fig. 2, which demonstrates a remarkable effect of Tachigaren treatment on inferior

grains. This result was obtained from the experiment undertaken in the paddy field of the National Institute of Agricultural Sciences in 1980. As is clear from Fig. 2, the 2nd and 3rd grains (from the top) on the secondary rachis branch showed a remarkable effect of Tachigaren on grain weight. This fact indicates that Tachigaren is effective to inferior grains that are particularly vulnerable to cool weather damage.

#### 2) Effect of Tachigaren on carbohydrate translocation to panicles

To determine the effect of Tachigaren on carbohydrate translocation in rice plants,  $^{14}CO_2$  tracer studies were conducted with potted plants of var. Nihonbare. Tachigaren was applied at the heading stage. Immediately after the Tachigaren treatment, both treated and control plants were fed with  $^{14}CO_2$  to make them carry out  $^{14}CO_2$  assimilation. On the next day, the plants were moved into a green house kept at 20°C during the day time and 13°C during the night. The plants were sampled at 1 and 10 days after the application of Tachigaren and the distribution of  $^{14}C$  in ears was determined by autoradiography (Plate 1).

Effect of Tachigaren on translocation of



- Fig. 1. Relationship between weight of each grain and position of each grain on a panicle
  - Note: Grain weight was measured 4 weeks after heading. Each figure is the average weight in mg of grains on the same position of 20 panicles. Upper figures indicate grain weight in the low temperature plot and lower figures that in the optimum temperature plot. Solid grains indicate that their ripening was retarded by low temperature.

<sup>14</sup>C into ears was not detected 1 day after the treatment but after 10 days a clearly higher rate of <sup>14</sup>C translocation into grains on the lower halves of ears than in the control plot was recognized. This observation suggests that Tachigaren promotes the translocation of carbohydrate to inferior grains in particular.



- Fig. 2. Effect of Tachigaren on weight of different grains
  - Note: A 1:500 dilution of Tachigaren liquid was sprayed over rice plant (var. Nihonbare) at the heading stage. Grains on the 4th primary rachis branches (from the top) of panicles emerged later in each hill were weighed. Upper figures indicate grain weight in the treated plot and lower figures grain weight in the control plot. Figures in parentheses indicate ratios of the former to the latter.

# Field tests in areas attacked by cool weather damage

In 1980, rice plants in the northeastern part of Japan were severely damaged by cool weather. The Toyama Prefectural Agricultural Experiment Station located in this part undertook a field experiment with Tachigaren. Young seedlings of var. Fujiminori were transplanted in a field at an altitude of about 640 m on May 18, and a 1:500 dilution of Tachigaren was applied at the panicle-formation stage (July 22) and at the full heading time (August 19) at a rate of 150 l per

| Experiment Station (1980)                                   |                        |                        |  |  |  |
|---|------------------------|------------------------|--|--|--|
| Plot  | Straw weight<br>(kg/a) | Grain weight<br>(kg/a) |  |  |  |
| Control   | 75.8                   | 41.0(100)              |  |  |  |
| Sprayed at full head-<br>ing stage                          | 74.1                   | 50.4(123)              |  |  |  |
| Sprayed at panicle-forma<br>stage and full heading<br>stage | ution<br>g 77.9        | 50.5(124)              |  |  |  |

Table 2. Field test conducted by the To-

yama Prefectural Agricultural

10 a. The mean daily temperature during the period from heading to harvest was  $17.1^{\circ}$ C, which was rather unfavorable for rice ripening.

The result of the field test is summarized in Table 2. Grain yield in the treated plot sprayed with Tachigaren was higher by about 20% than in the control plot. Although the yield itself did not differ appreciably between a single application of Tachigaren at the full heading stage and two applications at the panicle-formation stage and full heading stage,



Fig. 3. Distribution of thickness of brown rice (Toyama Prefectural Agricultural Experiment Station, 1980)



1 day after <sup>14</sup>CO<sub>2</sub> assimilation

10 days after <sup>14</sup>CO<sub>2</sub> assimilation

- Plate 1. Autoradiographs showing translocation of <sup>14</sup>C-assimilates to the panicle
  - Note: Rice plants (var. Nihonbare) were treated with Tachigaren at the heading stage (Aug. 26). On August 27, following the assimilation of <sup>14</sup>CO<sub>2</sub> the plants were transferred into a temperature-controlled green house (20°C during the daytime and 13°C during the night).



Control

Sprayed at panicle-formation stage and heading stage

Plate 2. Effect of Tachigaren on ripening of rice grown in a cold-waterirrigated field (Tohoku National Agricultural Experiment Station, 1980)

an advantage of the application at the panicleformation stage was reflected in the higher winnowed paddy weight in the plot with two applications of Tachigaren.

In this field test a discovery of particular interest was made on thickness of brown rice. A larger portion of brown rice in the treated plot than rice in the control plot was found to belong to thick grain (2.1 to 2.2 mm thick) (Fig. 3). Furthermore, in a field test carried out by the National Tohoku Agricultural Experiment Station, the favorable effect of Tachigaren on rice grain development was clearly observed. As shown in Plate 2, its effect was apparently known even by gross inspection.

## A country-wide field test conducted from 1979 to 1981

A large number of field tests were carried out by national and prefectural agricultural experiment stations for 3 years from 1979 through 1981 as a trust project of the Japan Association for Advancement of Phytoregulators to determine the effect of Tachigaren



- Note : o: Treated at panicle-formation-meiosis stage •: Treated at booting-heading stage
- Fig. 4. Relationship between rate of increase (%) of ripening percentage in the Tachigaren-treated plot and ripening percentage in the control plot (data compiled from the country-wide field experiments conducted for 3 years)

under diversified conditions. The results obtained in the field tests undertaken all over the country were compiled and analyzed in order to make a more accurate evaluation of the effect of this chemical on rice ripening.

Fig. 4 shows the rate of increase in ripening percentage caused by Tachigaren, plotted against the ripening percentage of the control. As is evident from this figure, the lower the ripening percentage in the control plot, the higher was the rate of promotion of ripening in the treated plot, sprayed with Tachigaren. When the ripening percentage in the control plot was higher than 80%, Tachigaren gave only a little effect. In other words, Tachigaren was not effective under favorable conditions which allowed 80% or higher rates of ripening, but it induced a substantial increase in ripening percentage under unfavorable conditions which caused 70% or lower rates of ripening in the control plot.



Note: o: Treated at panicle-formation~meiosis stage •: Treated at booting~heading stage

Fig. 5. Relationship between the sum of daily temperatures for 40 days after heading and the rate of increase (%) of ripening percentage induced by Tachigaren treatment (data compiled from the country-wide field experiments conducted for 3 years) Fig. 5 shows the relationship between temperatures during the ripening period and the effect of Tachigaren to promote ripening. The effect of Tachigaren is not recognized when the temperature is high during the ripening period, but it is remarkable when the sum of average daily temperature for 40 days after heading is  $800^{\circ}$ C (average daily temp.  $20^{\circ}$ C) or less. In short, it can be said that Tachigaren exerts a marked effect on ripening at low temperatures causing retardation of ripening.



Note: o: Treated at panicle-formation~meiosis stage •: Treated at booting~heading stage

Fig. 6. Relationship between the proportion of thick grains to the total amount of grains produced in the Tachigaren-treated plot and ripening percentage in the control plot (data compilea from the country-wide field experiments conducted for 3 years)

As shown in Fig. 6, Tachigaren application increased the proportion (%) of thick grains to the total amount of grains produced. This effect was manifested irrespective of the ripening percentage in the control plot but the degree of effectiveness appears to be high under the conditions that favor the ripening of rice.

#### Summary

On the basis of a large number of experiments conducted at many places in the coun-

JARQ Vol. 17, No. 3, 1983

try, it was clarified that when the temperature is low during the ripening period, Tachigaren application is very effective in increasing rate of ripening and promoting grain development of rice. It is, therefore, desirable to apply Tachigaren at the full heading stage when an undesirably low temperature during the ripening period is expected, or when the heading is delayed for one week or more due to a low temperature before heading. This chemical may also be appreciated in the regions where unripe or waste rice grain problems occur chronically.

#### References

- Ota, Y.: The effect of Tachigaren on the promotion of ripening in rice plant under low temperature condition. Agr. and Hort., 56, 657-660 (1981) [In Japanese].
- Ota, Y.: The effect of 3-hydroxy-5-methylisoxazole on the promotion of ripening in rice plant under low temperature condition. *Chem. Regulation of plants*, 17, 145-149 (1982) [In Japanese].
- Ota, Y. & Yamamura, S.: The effect of growth regulators on the ripening in rice plant. J. Crop. Sci. Soc. Jpn., 49, sp. ed. 1, 161-162 (1980) [In Japanese].

(Received for publication, April 7, 1983)