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Mesocotyl Elongation in Rice Seedlings Grown from Immature Grains

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In three cultivars of **japonica** paddy rice, mesocotyl length of seedlings grown in the dark from grains obtained 10 and 20 days after **anthesis** was greater than 10 mm, while mesocotyl length decreased with the maturity of the grain and it was less than 10 mm in grain obtained 40 days after anthesis. Abscisic acid (ABA) stimulated the elongation of mesocotyl to more than 10 mm in a cultivar of **japonica** paddy rice in the dark, though it inhibited the elongation of coleoptile. It seems that morphological characters of these seedlings were similar to those of the seedlings grown from immature grains. From the above results, it is suggested that ABA-like substances were contained in the mesocotyl of the paddy rice seedlings grown from immature grains.

INTRODUCTION

The sativa rice varieties of the world were firstly grouped by Kato and coworkers (1928, 1930) into *japonica* and *indica* sub-species on the basis of morphological differences, serological reactions and sexual affinities.

According to Hamada (1937), the mesocotyl of the *japonica* type of rice usually did not elongate to more than **10** mm even in total darkness, although that of the *indica* type would elongate to more than 10 mm.

Recently, in a rice cultivar of the *japonica* type it was found that the mesocotyl lengths of seedlings grown from imperfectly ripened grain were about 3 times longer than those grown from fully ripened grain (Inouye and Hibi, 1972). This result suggested that if immature rice grains of the *japonica* type are incubated, the mesocotyl may elongate as long as that of the *indica* type.

The mesocotyl lengths of seedlings grown in the dark in aseptic conditions were examined. The seedlings were grown from grains obtained at different stages of maturity after anthesis.

MATERIALS AND METHODS

The materials used were from four cultivars of paddy rice plants of the *japonica* type, Nihonbare, Norin-22, Saikai-112 and Reiho. The first and the second

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of these are early maturing varieties, and the third and the fourth are late maturing ones grown in the southern part of Japan. Plants were grown in pots (a/ 5000) under field conditions. All except the glumous flowers which were fertilized from the 3rd to the 5th day after the beginning of heading were cut off. About 30 grains were set on each panicle. Plants of a late maturing variety, Reiho, were then grown under controlled temperatures of 20, 25 or 30 °C in a phytotron, and under field conditions, respectively. In the other three varieties, the plants were grown in a controlled temperature of 20 °C and under field conditions. After 10, 20, 30, 40 or 50 days of these treatments, about 30 grains were obtained from each of the varieties and stored in an air conditioned room at 20 °C for further 50 days.

In order to examine the effect of ABA on the elongation of mesocotyl and coleoptile, Reiho was used as a material. In this experiment, seeds were obtained 50 days after anthesis under field conditions and stored in an air conditioned room at 20 $^\circ C$ for about three months.

After removing the husk, the grain was disinfected in 75 % alcohol for 30 sec, then in 3 % hydrogen peroxide for 3 min and finally washed one or two times with sterilized water. Unless otherwise mentioned, two grains were sown in a glass tube (18×250 mm) containing about 6 ml of 0.6 % plain agar medium. The tubes were placed in the dark at a constant temperature of 30 °C for about 12 days. Each experimental treatment consisted of about 8 tubes.

RESULTS AND DISCUSSION

Effect of temperature during the development of the grain after anthesis on the mesocotyl and coleoptile elongations of the seedlings

Results obtained are given *in* Table 1. The average mesocotyl length of seedlings grown from grains obtained at each temperature and 10-20 days after anthesis was greater than 10 mm with an exception for the grain obtained at 25 °C and 20 days after anthesis. Mesocotyl length decreased with maturity of the grain. On the other hand, the average coleoptile length increased with the maturity of the grain. Therefore, on an average, the total length of the seedling (length of the mesocotyl plus coleoptile) was similar for the grain harvested at different stages of maturity and at different temperatures.

Varietal differences in mesocotyl and coleoptile elongations of seedlings grown from grains at different stages of maturity

The results obtained at 20 $^{\circ}$ C and under field conditions were almost the same in each variety. Therefore, varietal differences at 20 $^{\circ}$ C are shown in Table 2.

In an early and in two late maturing varieties, the average mesocotyl length of seedlings grown from grains obtained 10 and 20 days after anthesis was more than 10 mm. In an early maturing variety, Nihonbare, the mesocotyl length was 16 mm even in the seedlings grown from grains obtained 30 days after anthesis. In the other early maturing variety, however, it was shorter than 10 mm in the seedlings grown from grains obtained 20 days after anthesis. In all the varieties

Table 1. Effect of temperature during the development of the grains after anthesis on the mesocotyl and coleoptile elongations of the seedlings grown from this grain. Mesocotyl (mm)

Days after	Temperature (°C)*			
anthesis	20	25	30	Field
20 30 40	$\begin{array}{c}(2)\ 65\pm 39.7\\(10)\ 25\pm 13.7\\(11)\ 5\pm\ 1.4\\(15)\ 3\pm\ 1.0\end{array}$	(13) 22 ± 16.4 (10) 8 ± 2.3 (5) 2 ± 0.9 (6) 4 ± 2.4	$ \begin{pmatrix} 9 \\ 5 \end{pmatrix} 16 \pm 12.2 (5) 14 \pm 1.7 (4) 5 \pm 2.7 (10) 7 \pm 3.1 $	$ \begin{array}{c} (0) \\ (3) \\ (12) \\ (12) \\ 6\pm 2.6 \\ (15) \\ 6\pm 2.1 \end{array} $

Coleoptile (mm)

Days after

Temperature (°C)*

5		1	, <i>,</i>	
anthesis	20	25	30	Field
10 20 30 40	$\begin{pmatrix} 2 \\ 10 \\ 18f9.5 \\ (11) \\ 24\pm 6.3 \\ (15) \\ 34\pm 7.3 \end{pmatrix}$	$\begin{array}{c} (13) \ 21 \pm 9.8 \\ (10) \ 30 \pm 9.9 \\ (5) \ 38 \text{f} 1.7 \\ (6) \ 36 \pm 5.8 \end{array}$	(9) 22 ± 13.7 (5) 29 ± 14.3 (4) 33 ± 3.6 (10) 37 ± 3.3	

In each lot, fifteen seeds were sown and incubated at 30° C for 12 days in the dark. The number of seedlings that grow is enclosed in parentheses.

*Temperatures during the development of the grains after anthesis.

Table 2. Varietal differences in mesocotyl and coleoptile elongations of seedlings grown from grains at different stages of maturity at 20°C. Mesocotyl (mm)

Days after	Varieties			
anthesis	Nihonbare	Norin-22	Saikai-112	Reiho
$10 \\ 20 \\ 30 \\ 40 \\ 50$		$\begin{array}{c} (1) \ 30 \\ (14) \ 8 \pm 3.3 \\ (15) \ 6 \pm 3.0 \\ (13) \ 4 \pm 1.3 \\ (15) \ 3 \pm 1.8 \end{array}$	$\begin{array}{c} (1) & 30 \\ (15) & 33 \pm 21.5 \\ (4) & 4 \pm & 1.6 \\ (15) & 5 \pm & 3.6 \\ (15) & 3 \pm & 1.8 \end{array}$	$\begin{array}{cccccccccccccccccccccccccccccccccccc$

Coleoptile (mm)

Days after	Varieties				
anthesis	Nihonbare	Norin-22		Saikai-112	Reiho
10 20 30 40 50		$ \begin{array}{c} (1) & 3 \\ (14) & 32 \pm 9.1 \\ (15) & 44 \pm 10.1 \\ (13) & 42 \pm 5.6 \\ (15) & 47 \pm 10.4 \end{array} $		$\begin{array}{ccccc} (1) & 4 \\ (15) & 10 \pm & 4.8 \\ (& 4j & 39 \pm & 4.9 \\ (15) & 43 \pm & 13.3 \\ (15) & 43 \pm & 6.3 \end{array}$	(2) 4±1.4 (10) 18f9.5 (11) 24f6.3 (15) 34±7.3 (15) 39±7.4

In each lot, fifteen seeds were sown and incubated at 30° C for 12 days in the dark. The number of seedlings that grow is enclosed in parentheses.

used in this experiment, the average mesocotyl length decreased with the maturity of the grain. On the other hand, the average length of the coleoptile increased with the maturity of the grain. Therefore, the average seedling length in each variety was nearly the same for grain of all stages of maturity. Effect of ABA on the mesocotyl and coleoptile elongations of rice seedlings

Recently, Takahashi (1972, 1973) found that ABA stimulated the elongation of mesocotyl to more than **20** mm in a *japonica* type of paddy rice under darkness, though it inhibited the elongation of shoot above coleoptilar node. It seems that the morphological characters of these seedlings were similar to those of the seedlings grown from immature grains,

Two seeds were sown on medium contained of 0.6 % agar and various concentrations of ABA (Sigma). Results are given in Table 3.

Jeressi				
Concentrations	Number of	Length (mm)		
of ABA (ppm)	seedlings	Mesocotyl	Coleoptile	
Control 0.1 0.25	18 16 17 17	$\begin{array}{c} 1\pm \ 0.6 \\ 1\pm \ 0.4 \\ 3\pm \ 2.4 \\ 15\pm 10.9 \end{array}$	24 ± 3.5 22 ± 0.9 17 ± 4.3 12 ± 7.0	
0.5 1.0 2.5 5:0	16 17 17 17	25f12.9 17t14.4 0 0	$7 \pm 1.0 \\ 7 \pm 2.2 \\ 3 \pm 1.0 \\ 3 \pm 0.9$	

 Table 3. Effect of ABA on the mesocotyl and coleoptile elongations in a paddy rice cultivar Reiho, japonica type.

After sowing, they were incubated at $30^\circ\mathrm{C}$ for 14 days in the dark.

The average mesocotyl length of seedlings increased with concentration of ABA in the medium and it was longer than 10 mm in the medium which contained 0.25-1.0 ppm ABA. Against this, the average coleoptile length of seedlings decreased with the concentration of ABA. At the concentration of 0.5 ppm ABA, the average mesocotyl length was at its maximum, whereas the average coleoptile length of seedlings was about one third times shorter than that of control ones. It seems that the morphological characters of these seedlings were similar to those of the seedlings grown from immature grains.

According to Oritani and Yoshida (1971), activity levels of ABA-like substances in rice grains were much higher in immature grains than those of fully ripened ones.

From the above results, it is suggested that ABA-like substances may be contained as stimulating factors in the mesocotyl of seedlings grown from immature grains in paddy rices of *japonica* type.

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