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# Diurnal Changes of α-Amylase Activity and Starch Content of Rice (*Oryza sativa* L.) Seedlings

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**Abstract** : Diurnal changes of α-amylase activity and starch content were examined in the seedlings of three types of rice (*Oryza sativa* L.), i.e., Indica type (cv. IR24), Japonica x Indica type (cv. Suweon 258) and Japonica type (cv. Nipponbare). The results obtained are summarized as follows :

1. In all varieties the α-amylase activity was lower during the daytime (6 a.m. to 6 p.m.) than durng the nighttime (6 p.m. to 6 a.m.). The activity was highest between 4 a.m. and 6 a.m., and rapidly decreased between 7 a.m. and 9 a.m..
2. The average activity of α-amylase in a day was highest in IR24, a little lower in Suweon258, and lowest in Nipponbare, which was about 30% of that in IR24.
3. The diurnal changes of starch content showed a tendency converse to those of α-amylase activity : viz, starch content was lower during the nighttime than during the daytime in all varieties.
4. The average of starch content in a day was highest in Suweon258, a little lower in IR24 and lowest in Nipponbare.

**Key words** : α-Amylase, Diurnal changes, Rice seedlings, Starch content.

**水稲幼植物におけるα-アミラーゼ活性と澱粉含量の日変化**：田中尚道・*西川欣一・*秋田謙司（神戸大学大学院自然科学研究科・*神戸大学農学部）

**要　旨**：水稲品種 IR 24 号（インド型），水原 258 号（日印交雑種）及び日本晴（日本型）を用いて，播種後 20 日目（葉齢約 4.0）の葉鞘におけるα-アミラーゼ活性と澱粉含量の日変化について調査し，以下のような結果を得た．

1．α-アミラーゼ活性の日変化はいずれの品種も午前 6 時から午後 6 時（昼間）に比べて午後 6 時から午前 6 時（夜間）の方が高かった．特にα-アミラーゼ活性の高い時刻は 3 品種とも，午前 4 時から午前 6 時の間で，その後いずれの品種も午前 7 時から午前 9 時にかけて著しく低下する傾向がみられた．

2．品種間でみると，α-アミラーゼ活性は IR 24 号が最も高く推移し，水原 258 号でもほぼ同様の活性を示したが，日本晴は IR 24 号の約 30% の低い活性で推移した．

3．澱粉含量の日変化は，いずれの品種もα-アミラーゼ活性とは逆に午前 6 時から午後 6 時（昼間）の方が午後 6 時から午前 6 時（夜間）より含量は高かった．

4．品種間でみると，澱粉含量の最も多かったのは水原 258 号で，IR 24 号もほぼ同様の値で推移した．一方，日本晴は両品種に比べて澱粉含量は低く推移した．これは，品種特性（耐肥性）の違いによるものではないかと思われた．

**キーワード**：α-アミラーゼ活性，水稲幼植物，澱粉含量，日変化．

Amylase activity of rice plants has been studied[7,12,15,17,18] mainly in types, forms and distribution of grains[17], and it is well known that rice amylase is composed of multiple forms[18].

Rice varieties of Japonica type are generally assumed to contain amylase not only in the grains but also in the leaves and stems. Since rice plants accumulate starch in the leaf-sheath at the seedling stage[1-4,13,22], amylase activity may have a connection with the distribution of starch in the leaf-sheath[14]. Murakami[4] reported the relationship between sampling time in a day and sugar and starch contents, and the clarified that the starch content was the most variable in a day.

As previously reported[10], we clarified the diurnal changes of β-amylase activity and its isozyme pattern in alfalfa seedlings. We then intended to find the cross relationship between varietal characteristics and diurnal changes in amylase activity in rice seedlings.

The present paper deals with the investigation of the distribution of amylase, starch and their diurnal changes in the leaf-sheath of rice seedlings, using 3 varieties of different type.

## Materials and Methods

In September 1987, rice plants were grown in a greenhouse, the air temperature of which was controlled at 25/20°C (day/night) between 6 a.m. and 6 p.m. under natural daylight.

Rice varieties IR24, Suweon258 and Nipponbare grown in seedling cases were used as materials. The 40 seeds were sown (1cm × 1cm) in a plastic seedling case. Each seedling case (5cm × 15cm × 10cm) contained 0.8kg of soil and 0.5g each N, $P_2O_5$ and $K_2O$.

On a very fine day, twenty days after seeding (plant age 4.0), starch content and amylase activities were measured in 20 seedlings sheath sampled at every 2 hours during the day.

For starch analyses the dry samples were ground using a vibration mill into less than 100 mesh. The starch content were analyzed without 80% hot ethanol extraxt, after the residues were hydrolyzed by 0.7N sulfuric acid at 80°C for 1hr and analyzed by the Somogi-Nelson method.

Amylase activity was measured by the method described previously[10]. In the measurement of $\alpha$-amylase activity corrections were made for the difference in co-existant $\beta$-amylase in the enzyme preparations. Amylase acuivity is expressed in terms of mg of maltose per ml of each extraction solution.

## Results

The diurnal changes of amylase activity in leaf-sheath is shown in Fig.1. In all varieties the $\alpha$-amylase activity exhibited a decrease to increase during the daytime to nighttime. Suweon258 and IR24 showed a higher amylase activity than that of Nipponbare.

The diurnal changes of starch content of leaf-sheath is shown in Fig.2. The starch content of leaf-sheath was highest in Suweon258, and next in IR24; Nipponbare showed the lowest starch content. The changes of starch content in a day showed the same tendency, in all varieties, starch content was higher during the daytime than during the nighttime.

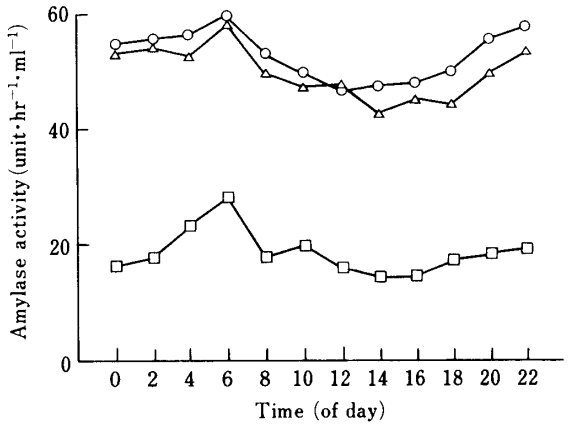


Fig. 1. Changes in amylase activity in the leaf-sheath of rice seedlings during a day.
○ : IR24, △ : Suweon258, □ : Nipponbare.

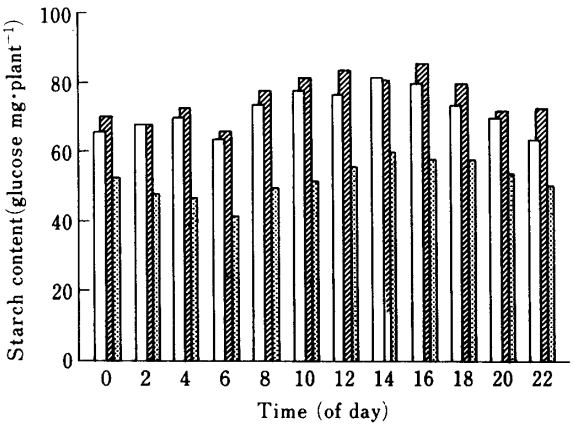


Fig. 2. Changes in starch content in the leaf-sheath of rice seedlings during a day.
□: IR24, ▨ : Suweon258, ▩ : Nipponbare.

Table 1. Average of amylase activity and starch content in leaf-sheath in a day

| Variety | 6 a.m. to 6 p.m. amylase (unit·hr$^{-1}$·m$l^{-1}$) | 6 a.m. to 6 p.m. starch (mg) | 6 p.m. to 6 a.m. amylase (unit·hr$^{-1}$·m$l^{-1}$) | 6 p.m. to 6 a.m. starch (mg) |
|---|---|---|---|---|
| IR 24 | 59.57 | 88.17 | 65.45 | 79.33 |
| Suweon 258 | 56.38 | 92.83 | 61.37 | 83.67 |
| Nipponbare | 21.60 | 62.67 | 23.70 | 58.83 |

Table 1. shows the average amylase activity and starch content in a day (during the daytime and during the nighttime). In all cultivars the average $\alpha$-amylase activity was higher during the nighttime than during the daytime. However, in all varieties the average starch content was higher during the daytime than during the nighttime.

## Discussion

The diurnal changes in the amylase activity showed the same tendency in all cultivars ; viz, higher during the nighttime than during the daytime. Indica and Japonica×Indica type showed a higher amylase activity than Japonica type in a day. This may also be related to characteristics of rice plants, especially to difference of adaptability for heavy manuring[8,9,20]. Shinke et al[18]. reported that floating rice was found to increase during the daytime and decrease during the nighttime[13,14]. This may also be related to the balance of the rate of photosynthesis and respiration rate in a day. Therefore, diurnal changes of starch content may also be correspond to the rise and decline of total sugar content. Murakami[4,5] reported the relationship between sampling time and sugar and starch content in $C_3$ and CAM plants. Her report suggested that the sugar content increased in the nighttime, but starch content decreaesd during nighttime, and the difference in each organ, especially starch content was greater in the daytime. In the present study only leaf-sheath was measured, but we obtained almost the same results as in her report. There results may also suggest that the varietal differences of amylase activity and starch content might reflect characteristics of rice varieties, especially to difference of adaptability for heavy manuring, because higher starch accumulation was higher amylase activity. Therefore, in the future, it may be necessary to clarify the relationship between amylase activity, starch content and physiological characteristics of rice varieties.

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** In Japanese. The title has been tentatively translated by the present authors.