

## Effects of Water Depth and Temperature on the Seedling Growth of Wild Rice, *Zizania palustris* L.

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**Abstract** : Under the circumstance of measures for reorganizing paddy field utilization in Japan, the aquatic habit and adaptability to cold climates of wild rice (*Z. palustris* L.) have stimulated interest in the possibility of commercial cultivation in the northern Japan. To investigate the effects of water depth and temperature on the growth at seedling and early vegetative stages, two experiments were carried out. The glasshouse experiment using Netum showed that seedling growth in terms of dry weight at 30 days after transplanting was accelerated by water depth of 2 cm. In the growth cabinet experiment using K2, increases in dry matter occurred in the range of 2 to 6 cm, being due to increased number and size of leaves and roots. Although plant height was not affected or only marginally affected by water depths, spindly growth with narrow and light color leaves appeared when plants were grown at the depths more than 8 cm. Under the controlled environments, seedling growth was much better in plants grown at 20°C than those at 12°C. There were no interactions between temperatures and water depths, suggesting that temperature is an independent factor of water depth.

**Key words** : Dry weight, Seedling establishment, Temperature, Water depth, *Zizania palustris*.

ワイルドライス幼植物体の生育に及ぼす水深と温度の影響 : 源馬琢磨・三浦秀穂・林 克昌 (帯広畜産大学)

**要 旨** : わが国の水田利用再編対策のもとで、ワイルドライス (アメリカマコモ) のもつ水生植物としての特性や寒冷地での適応性は、北日本での実用的な水田栽培の可能性に対する興味を刺激している。本研究では、幼植物体の生育に及ぼす水深と温度の影響を検討するため、二つの実験を行った。実験1は Netum を用いてガラス室で、実験2は K2 を用いて人工気象室で実施した。移植後30日目の乾物重でみた生育は、実験1では水深2 cm で、実験2では2~6 cm で促進された。これらには葉と根の数とサイズの増加が貢献していた。草丈は水深の違いによって大きく影響されないが、8 cm 以上の水深で栽培したとき徒長気味の生育を示し、幼植物体の乾物重は大きく低下した。実験2でみた温度の影響は強く表れ、12°C に比べ20°C での生育が優った。温度と水深の間には相互作用がなく、これら二つの要因は独立して幼植物体の生育に影響を及ぼすとみられた。

**キーワード** : 温度, 活着, 乾物重, 水深, ワイルドライス。

Wild rice, *Zizania palustris* L., is an annual grass grown in flooded conditions. It is the only cereal indigenous to many lakes and slow-moving streams in northern Minnesota, U.S.A. and southern Canada. The grains were traditionally harvested for a major food by the Ojibway and Sioux tribes of native Americans in areas along the Great Lakes. Today, wild rice is enjoyed by many as a nutritious cereal with a unique texture and nut-like flavor. Minnesota leads the world in wild rice production from natural stands and cultivated fields<sup>6)</sup>. Commercial paddy cultivation in this state is developed recently and has expanded. This is largely attributed to the discovery of genotypes with less shattering than those in natural stands<sup>1,3)</sup>. On the other hand, most commercial harvesting as the lake wild rice in

Canada is carried out from unmanaged natural stands in lakes and small streams.

Although wild rice production in Japan has not started yet, both the political measures for reorganizing paddy field utilization and the high price of seed due to the limited supply have stimulated interest in the possibility of cultivating wild rice. Furthermore, in terms of an excellent amino acid composition in grains<sup>5,7)</sup> and adaptability to cold climates, wild rice has fascinated us as a new genetic resource to improve rice (*Oryza sativa*) germplasm<sup>8,10)</sup> since the genus *Zizania* is a genera closely related to the genus *Oryza*.

Attempts to propagate this plant by transplanting seedlings into farmer's paddy field have initiated in our laboratory but not still been very successful. It has been noticed that problems which should be overcome are associated with poor establishment of transplant-

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ed seedlings, probably due to uneven size of seedlings and inappropriate water depth. When an intensive cultivation including nursery and transplanting of seedlings is aimed, two main problems must be solved. The first is about methods for preparing a sufficient number of germinated seeds or seedlings with uniform size and the second is in regard to water management for ensuring the subsequent growth.

For the first, we have reported that without loss of viability freshly harvested wild rice seeds enable to be maintained by dry storage for six months or more, and then germinate by soaking in cold water for more than 20 days<sup>2)</sup>. This has brought an easier and sufficient supply of seedlings for spring sowing experiments in our laboratory.

For water management, there are several reports which mentioned that the effect of water depth changed with the growth stages<sup>9)</sup>, while the growth was normal when a water depth was maintained below 10.4 cm throughout its life cycle<sup>11)</sup>. In Minnesota, to control weeds the critical period for maintaining at least 15 cm water depth is recommended during the first 8 to 10 weeks of the season<sup>6)</sup>. However, few informations are available about seedling growth after transplanting. In this paper we describe results of two experiments, in which seedlings are grown under different levels of water depth in a glasshouse (Experiment 1) and in controlled environment cabinets (Experiment 2), to investigate effects of water depth and temperature on the growth at early vegetative stages.

### Materials and Methods

Netum was used in Experiment 1 and K2 was used in Experiment 2. These cultivars have shatter resistance and are released for Minnesota wild rice growers<sup>3)</sup>. The seeds were harvested in paddy field at middle September, and stored under a dry condition at 5°C for seven to eight months. To overcome dormancy the seeds of both experiments were soaked in cold tap water at 4–6°C for about one month. Subsequently these seeds were germinated in tap water at room temperature. During the soaking and germination, water was changed everyday to prevent molding.

In Experiment 1, seedlings around 5 cm in height were transplanted into plastic pots

(20 × 17 cm ; diameter × depth) at the rate of eight plants per pot which filled with wet soil from paddy field. The planting density approximated 250 seedlings m<sup>-2</sup>. Three levels of water depth, 2cm, 5cm and 8 cm, were compared. On July 20, 1990, the pots were arranged with four replicates in a glasshouse. Water temperatures at different levels of water depth were recorded using a hand held data logger (Memory Sensor, MES-801 : Koito Industries, Ltd.).

In Experiment 2, seedlings with the first two or three leaves were normally submerged in 1/5000 a Wagner's pots containing wet soil from paddy field. Five seedlings per pot, corresponding to 50 seedlings m<sup>-2</sup>, were retained at the three levels of water depth, 2 cm, 6 cm and 10 cm. The experiment was initiated from June 4, 1991, and the seedlings were grown in two controlled environment cabinets with different mean air temperatures ; one cabinet was maintained at 20°C (at 22°C for day and 18°C for night) and the other at 12°C (at 14°C for day and 10°C for night). The photoperiod was provided by natural daylight in both of the cabinets. This experiment was replicated six times. To minimize positional effects pots were rerandomized weekly.

After 30 days all plants were sampled in both experiments, and plant height, the maximum root length and dry weight of whole plant including roots were measured on plant basis. In Experiment 2, number of roots, number of leaves and dry weights, which were separated into the shoots and roots, were also recorded. Dry weights were determined on material dried at 75°C for 72 hours. The data were subjected to analysis of variance.

### Results

#### 1. Experiment 1 ; Netum in glasshouse

During growing period, daily water temperature was fluctuated from 15 to 22°C. But differences in the temperature among the water depths were less than 1°C.

The effect of water depth on growth significantly appeared on plant dry weight. Table 1 shows that compared to plants grown at the depth of 2 cm, 18% and 27% decrease in dry matter occurred in plants at the depths of 5 cm and 8 cm, respectively. The hinder growth of both shoots and roots seemed to contribute to the dry matter reduction at the 5 cm and 8

cm water depths (data not shown). The effect on dry matter production at the 5 cm depth was similar to that at the 8 cm depth. Although plant height was not affected by the water depth, the morphological status was dependent on the water depth at which plants were grown. At the depth of 8 cm, most plants showed spindly growth with narrow and light green leaves.

## 2. Experiment 2 ; K2 in growth cabinets

The results of analysis of the variation are given in Table 2 and show that the differences between two temperature treatments, 20°C and 12°C, is a major source of the variation in all characters measured. The differential effects due to water depth were also found in five characters except plant height. The temperature × water depth interactions were not significant in any characters, suggesting that water depth and temperature independently affected the growth of wild rice seedlings.

Plants grown at 20°C were about 20 cm taller than those at 12°C. In the 12°C treatment, plant height was affected by the water depth where plants at the depth of 10 cm were 5 cm taller than those at the depth of 2 cm. The number of leaves and roots per plant, and the maximum root length increased as water depth decreased at both temperatures, while differences in the number of roots and leaves between the depths of 2 cm and 6 cm were not

significant. Plants grown in shallower water depths produced longer and more roots, resulting in higher production of dry matter at the depths of 2 cm and 6 cm than that at the depth of 10 cm. Leaf and stem growth was also affected by the water depths, in which an erect type phenotype was produced at the depths of 2 cm and 6 cm.

Table 1. The effect of water depth on three seedling characters of wild rice, *Netum*, grown in a glasshouse (Experiment 1)

| Water depth                       | Plant height (cm)  | Max. root length (cm) | Dry weight /plant (g) |
|-----------------------------------|--------------------|-----------------------|-----------------------|
| 2 cm                              | 23.8a <sup>#</sup> | 38.6a                 | 0.106a                |
| 5 cm                              | 23.8a              | 32.0b                 | 0.087b                |
| 8 cm                              | 24.8a              | 31.2b                 | 0.077b                |
| Significant effect of water depth | ns                 | ns                    | **                    |

<sup>#</sup>; The values within a column not followed by the same letter are significantly different at 5% level.

\*\* ; Significant at 1% level. ns ; Non-significant.

Table 2. The effects of temperature and water depth on six seedling characters of wild rice, K2, grown in controlled environment cabinets (Experiment 2)

| Temperature           | Water depth | Plant height (cm)  | No. of leaves /plant | No. of roots /plant | Max. root length (cm) | Dry weight/plant |          |           |
|-----------------------|-------------|--------------------|----------------------|---------------------|-----------------------|------------------|----------|-----------|
|                       |             |                    |                      |                     |                       | Shoot (g)        | Root (g) | Total (g) |
| 12°C                  | 2 cm        | 24.8a <sup>#</sup> | 7.00b                | 9.45b               | 29.0b                 | 0.096a           | 0.086b   | 0.183b    |
|                       | 6 cm        | 28.1ab             | 7.30b                | 9.88b               | 25.0b                 | 0.104a           | 0.064ab  | 0.169b    |
|                       | 10 cm       | 30.1b              | 6.39a                | 7.12a               | 21.5a                 | 0.047a           | 0.039a   | 0.087c    |
| 20°C                  | 2 cm        | 48.1c              | 9.10d                | 17.48d              | 38.4d                 | 0.472c           | 0.282d   | 0.756d    |
|                       | 6 cm        | 49.0c              | 9.95e                | 18.82d              | 40.3d                 | 0.533c           | 0.334d   | 0.867d    |
|                       | 10 cm       | 47.0c              | 8.45c                | 14.88c              | 33.5c                 | 0.330b           | 0.221c   | 0.550c    |
| Significant effect of |             |                    |                      |                     |                       |                  |          |           |
| Temperature           |             | ***                | ***                  | ***                 | ***                   | ***              | ***      | ***       |
| Water depth           |             | ns                 | **                   | *                   | **                    | *                | *        | *         |
| Interaction           |             | ns                 | ns                   | ns                  | ns                    | ns               | ns       | ns        |

<sup>#</sup>; The values within a column not followed by the same letter are significantly different at 5% level. \*, \*\*, \*\*\* ; Significant at 5%, 1% and 0.1% levels, respectively. ns ; Non-significant.

### Discussion

It was found in the present experiments that effects of water depth on wild rice seedlings and early vegetative growth clearly appeared on dry matter production of whole plant including roots. Under the uncontrolled temperature in Experiment 1, seedlings grown at the shallowest treatment of 2 cm water depth produced the highest dry matter. The reduction in dry matter at the 5 cm water depth seemed to be due to poor growth of some seedlings affected by the high planting density rather than water depth. This suggests that if the planting density is kept an appropriate level to ensure seedling establishment, water depths less than 5 cm is suitable during early vegetative growth stages. On the other hand, under the constant temperature in Experiment 2, seedling growth at the depth of 6 cm tended to be superior to that at 2 cm depth. This minor disagreement with Experiment 1 might be caused by differential response of cultivars to temperature, planting density and their combined effects. Furthermore, our preliminary experiment and other reports<sup>9)</sup> indicated that water depth at the ground level or 0 cm made seedlings wither. Thus water management less than 2 cm should be avoided in seedlings and subsequent vegetative stages.

Based on these informations, we tried transplanting experiments in grower's paddy field. Fig. 1 shows the growth of 20-day old seedlings of K2 after transplanting in which seedlings with the first two or three leaves were replanted at the middle of June. As evident from this, water depths less than 6 cm result in successful establishment and better subsequent growth of seedlings transplanted.

After a problem associated with establishment of seedlings comes to a satisfactory solution, the effect of water depth on subsequent growth and final grain yield should be assessed. Thomas and Stewart<sup>9)</sup> reported that yield in terms of dry weight of plant parts was decreased by water depths less than 8 cm under controlled environments. In the present experiments, although plant height was not affected or only marginally affected by different levels of water depth, morphological status changed with water depths. Plants grown at water depths of more than 8 cm showed an 'exhausted' phenotype with spindly type.

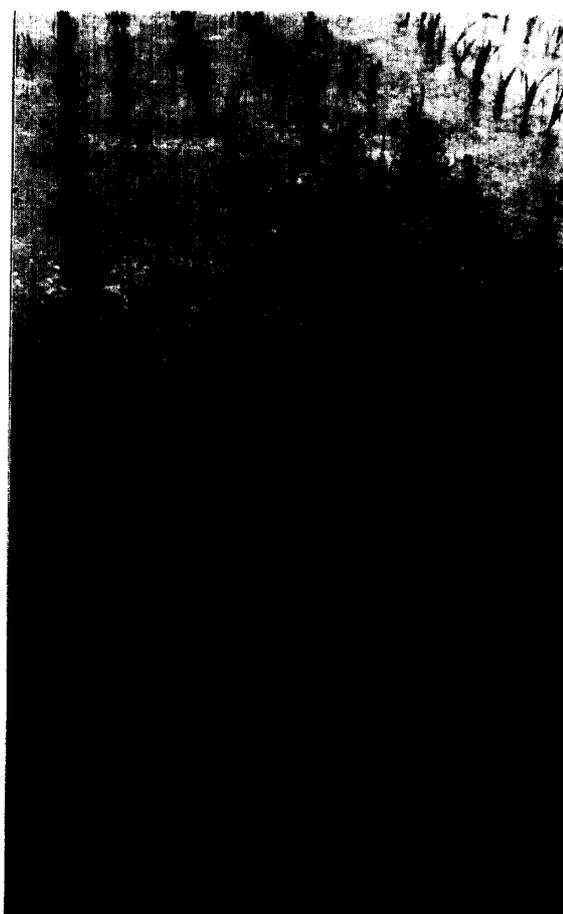


Fig. 1 The growth of 20-day old seedlings of K2 after transplanting in grower's paddy field. The seedlings with the first two or three leaves were replanted at the middle of June and the water depth was kept less than 6 cm.

Thus whether such 'exhausted' plants with less number of roots than plants grown at 2–6 cm depths produce less tillers and lodge earlier is of interest, and should be examined under paddy fields. It would be at least incorrect to assume that the unchanged water management at vegetative and reproductive phases would be profitable for grain yield production since the effect of water depth changes with the growth phase<sup>9)</sup>.

In addition to water depth, there are some factors affecting seeding success for commercial production of wild rice. As a biological factor, influence of transplanting injury on establishment and subsequent growth must be assessed because the number of ears per unit area can greatly affect the economical yield. Also chemical and physical factors of paddy fields used would be considered. Lee and

Stewart<sup>4)</sup> carried out sowing experiments at many sites with shallow water depth in north-western Ontario. The sites suitable for commercial production were characterized by an appropriate amount of sediment phosphorus and sediment soil types. As shown in Fig. 1, it is expected that soil fertility and sediment soil types in paddy fields distributed over the Tokachi district would not be limiting factors for establishment of seedlings. However, we have many uncultivated areas with shallow water depth in the northern Japan, in which possibility of commercial wild rice cultivation is uncertain.

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