Effects of Soil Moisture Depletion for One Month before Flowering on Dry Matter Production and Ecophysiological Characteristics of Wheat Plants in Wet Soil during Grain Filling

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Abstract: The period from mid-March to April in the wheat-growing season in Japan corresponds to a wet period known as “Natanezuyu”. After this wet period, the weather remains rather dry until June. Fluctuations in soil moisture conditions during the growing season might be expected to affect the growth of wheat. Therefore, we compared the grain yield, dry matter production and ecophysiological characteristics of wheat grown with adequate moisture during the ripening stage after it had been grown under adequate (W-plot) or deficient (D-plot) soil moisture conditions for about one month before heading. The grain yield in the D-plot was higher by about 15 to 40% than that in the W-plot, with greater dry matter production. The larger dry weight in the D-plot resulted from a higher rate of crop growth before and after heading, which was due to a larger leaf area and higher net assimilation rate. During the ripening stage, leaf senescence of plants was delayed and the rate of photosynthesis fell more slowly in the D-plot than in the W-plot. Root systems developed better, resistance to water transport from root to leaves was lower, the exudation rate of roots was higher, and the cytokinin activity in xylem exudates from roots was higher in the D-plot. These characteristics of roots might have caused the significant difference in the growth and physiology of the aboveground parts of the plants. Our results indicate that encouragement of the development of the root systems, for example, by drainage during the wet period might be important for improving the grain yield of wheat in Japan.

Key words: Cytokinin, Dry matter production, Photosynthesis, Resistance to water transport, Root system development, Senescence, Soil moisture, Wheat.

The mean annual precipitation in Japan ranges from about 1,700 to 1,800 mm and is far greater than the annual mean evaporation. Therefore, crop plants in Japan seldom suffer from the severe water stress that is usually experienced by plants in and around semi-arid regions. However, the growth of crop plants is certainly influenced by fluctuations in soil moisture conditions in Japan. Soybean plants, an example of a summer crop, grow with vigorous expansion of leaves and shallow roots at the vegetative phase during the rainy period, known as “Baiu”, that precedes the hot, dry summer. Since transpiration of these plants exceeds water absorption as a result of strong solar radiation and the large atmospheric vapor pressure deficit in the summer, the plants can easily suffer from a mild water deficit, even when soil moisture is adequate and they are susceptible to early senescence (Hirasawa et al., 1994, 1998).

Wheat plants were originally adapted to cool and rather dry conditions, and many strains are cultivated in regions where the annual precipitation is less than 1,000 mm (Briggle and Curtis, 1987). Thus, much research into their growth, yield, and ecophysiological characteristics has been performed under conditions of severe water deficit (e.g., Gusta and Chen, 1987; Loss and Siddique, 1994; Ludlow and Muchow, 1990). However, there has been little detailed research into the effects of fluctuations in moisture conditions during the growing season on the ecophysiology of wheat plants, although moisture damage caused by excessive rainfall and poor drainage was examined in early studies in Japan (Noda and Ibaraki, 1961; Ohtani, 1949; Tokimasa, 1951; Yamazaki, 1952).

Wheat plants are grown from fall to early the next summer in Japan, except in some parts of Hokkaido. The internodes of plants start to elongate and to grow rapidly in mid-March and ears emerge in mid-April. The period from mid-March to April falls in the wet period known as “Natanezuyu”, when precipitation is high and the atmospheric vapor pressure deficit is small (Fig. 1). Wheat plants grow rapidly for one month
Fig. 1. Daily precipitation (■) and daily maximum atmospheric vapor pressure deficit (VPD; ●) from January to June in Tokyo (data from Tokyo District Meteorological Observatory). Average values for 30 years from 1970 to 1999. VPD was estimated from the daily maximum temperature and daily minimum vapor pressure.

Fig. 2. Amount of precipitation (white bars) and irrigated water (black bars) for five-day periods from January through June, 1995. A, B, and C correspond to the C-, W-, and D-plots, respectively. Total precipitation from March 13 to June 22 was 550.6 mm. Total irrigation for W- and D-plots during the indicated period was 288 mm and 268 mm, respectively. Arrows indicate the date of heading.

Materials and Methods

1. Plant materials

Wheat plants (Triticum aestivum L. cv. Bandowase) were grown at the University paddy farm in 1994 and 1995. The soil in the field was classified as loam to clay loam from 0 to 34 cm below the surface, light clay from 34 to 44 cm, sandy loam from 44 to 71 cm and light clay from 71 to 120 cm. The field was well-drained and the level of the underground water was always far lower than 1.5 m below the soil surface.

For the experiment in 1995, seeds were sown at the rate of 80 kg ha⁻¹, with a distance between rows of 32.5 cm, on November 16, 1994. As basal dressing, manure was applied at the rate of about 10 t ha⁻¹ and chemical fertilizer (14–14–14) was applied at the rate of 100, 100 and 100 kg ha⁻¹ for N, P₂O₅, and K₂O, respectively. Ammonium sulfate was applied as topdressing at the rate of 50 kg ha⁻¹ on February 22, 1995. Plants were grown under natural rainfall conditions until March 13. Some plants were grown under conditions of deficient soil moisture by withholding irrigation water from March 13 to April 17 (D-plot; Fig. 2). Other plants were grown with adequate soil moisture by irrigation with 10 mm of water per three days in March and 12 mm per three days in April during the same period (W-plot; Fig. 2), to reflect the average recorded precipitation. The plants in the D-plot were irrigated with 70 mm of water on April 17 and 27, respectively, such that the soil moisture in the D-plot increased almost to that in the W-plot. Plants in both plots were grown with adequate soil moisture by irrigation with 10 mm of water per three days in May 1. Irrigation was suspended from June 5, during the maturation period. The experimental field, 114 m² in area, was divided into four parts, and plastic board that extended 1 m below the soil were installed in order to isolate each part hydraulically. Moisture treat-

before heading under the wet conditions. After the wet period, even though precipitation does not decrease significantly, it is rather dry with high air temperatures and a large vapor pressure deficit, until early June (Fig. 1). Such moisture conditions increase evapotranspiration and might cause mild water stress and senescence in plants and, therefore, they might affect the growth and dry matter production after heading in wheat plants of a winter crop as in the case of the above-mentioned effects of the rainy season on soybean plants. They might, moreover, be one of the factors that suppress the grain yield of wheat in Japan.

In the present study, to investigate how the precipitation from mid-March to mid-April affects the growth and ecophysiological characteristics of wheat, we examined the dry matter production, grain yield and related characteristics of wheat plants that were growing with adequate soil moisture during the ripening stage after they had been grown with adequate or limited soil moisture for about one month before heading. Experiments were carried out in 1994 and 1995.
ments for the W- and D-plots were arranged randomly. The field was isolated from rain water, by covering it in whole with a movable roof (about 40% shading of solar radiation), whenever it rained after March 13. Heading occurred around April 22 and harvest was June 22.

For the experiment in 1994, irrigation was withheld from March 12 to April 16 and the treatment of soil in both plots was similar to that in 1995, with the exception that the plants in the D-plot were irrigated with about 100 mm of water every two weeks in May. Heading occurred around April 18. Plants were harvested on June 12. The W- and D-plots had three replicates, with about 22 m² for each replicate.

For comparison, wheat plants were also grown under natural rainfall conditions during the entire growth period in an adjacent field with the same soil conditions as those in the moisture-controlled field (G-plot; Fig. 2).

2. Measurements of leaf area, dry weight of aboveground parts and grain yield

Leaf area and dry weight of aboveground parts were determined for the plants in an area of 0.25 m² for each replicate that had an average number of stems. Sampled plants were dried at 90°C in a ventilated oven after they had been divided into leaf blade, leaf sheath plus stem, and panicle. The leaf blade area of some plants from each replicate was measured with an area meter (AAM -8; Hayashi Denko Inc., Tokyo, Japan). Specific leaf area was calculated as the leaf blade area divided by the dry weight of the leaf blade. The entire leaf area for each replicate was calculated by multiplying the dry weight of all the leaf blades by the specific leaf area. Plants in an area of 2.4 m² for each replicate were harvested for determination of unit area yield. Yield components for each replicate were determined for the plants in an area of 0.3 m² with average number of panicles.

3. Soil moisture and leaf xylem water potential

Soil moisture was measured with a tensiometer. The leaf xylem water potential was measured with a pressure chamber (model 3005; Soil Moisture Equipment Inc., Santa Barbara, CA, USA) as described by Hirasawa and Ishihara (1991)

4. Root length

Root length was measured with a minirhizotron root-observation system (Hirasawa et al., 1995). Transparent acrylic resin tubes of 60 mm in outer diameter and 54 mm in inner diameter for observation of roots had been placed in the soil at an installation angle of 60° from the soil surface a few years before the present experiments. Roots were observed with a fiber-optic system (model 13005; Soil Moisture Equipment Inc.) and recorded with a video camera (Victor Inc., Tokyo, Japan) and videotape recorder (Victor Inc.). Pictures were reviewed on a monitor and lengths of roots were measured by the line intersect method (Tennant, 1975).

5. Chlorophyll content and rate of photosynthesis

The chlorophyll content of a leaf was estimated for the central portion of a leaf blade on the main stem with a chlorophyll meter (CT-101; SME Inc., Kawasaki, Japan).

The rate of photosynthesis was measured with an acrylic assimilation chamber (30 mm × 40 mm × 4 mm) that was clamped over the central portion of an attached leaf. Then air with a controlled CO₂ concentration and controlled dew point was pumped into the chamber at a rate of 33.3 cm³ s⁻¹, via a mass flow controller (model SEC-521; STEC Inc., Tokyo, Japan). The concentration of CO₂ in the air pumped into the chamber during measurements was 346 ± 0.1 μL L⁻¹ and the dew point of the air pumped into the chamber was 9 ± 0.1°C, 10 ± 0.1°C and 10 ± 0.1°C in March, April and May, respectively. The difference in CO₂ concentration between the air pumped into and out of the chamber was measured with an infrared CO₂ analyzer (model ZAP; Fuji Electric Inc., Tokyo, Japan). The dew point of the air was measured with a dew point meter (Model 660; EG&G Inc., Walthan, MA, USA). Leaf temperature was measured with a copper-constantan thermocouple of 0.1 mm in diameter, which was attached to the lower surface of the leaf. Measurements were taken under artificial light from a halogen lamp (model LA-150; Hayashi-Tokei Inc., Tokyo, Japan) at 1,600 μmol m⁻² s⁻¹.

6. Resistance to water transport

The resistance to water transport from root to leaf (R) was measured under conditions of adequate soil moisture from the following equation, as detailed Hirasawa and Ishihara (1991):

\[ R = -\frac{\Psi_s}{T} \]

where \( \Psi_s \) is the leaf xylem water potential and \( T \) is the transpiration rate. The transpiration rate of an entire attached leaf was measured with an assimilation chamber in the daytime on a clear day. Air, with the dew point controlled to about 10°C, was pumped into the chamber at a rate of 66.7 cm³ s⁻¹. The difference in dew point between the air going into and out of the chamber was measured with a dew point meter. When the transpiration rate reached a constant value, the leaf xylem water potential was measured with a pressure chamber (model 13005; Soil Moisture Equipment Inc.).

7. Exudation rate and cytokinin activity in xylem exudates

Plants were cut at an internode about 3 cm below the third node from the flag leaf node on the main stem. The xylem exudate from the cut end was collected for about 15 h, from 1800 to 0900 of the following day, with cotton that was covered with a polyethylene bag during the collection period to prevent evaporation. The exudation rate was estimated from the increase in the weight of...
Xylem sap for the quantification of cytokinins was collected from 40 stems, excised at a height of 20 cm above the soil surface, with cotton for about 15 h during night, as described above. Xylem sap was weighed immediately after collection, supplemented with 99% ethanol, as a preservative, to a concentration of 80% ethanol, and stored at -20°C until analysis. Cytokinin activity was determined by the Amaranthus betacyanin bioassay (Biddington and Thomas, 1973) using Amaranthus tricolor. The xylem sap in 80% ethanol was evaporated on a rotary evaporator at 40°C to an aqueous residue. The residue was evaporated at 40°C, after purification with a Sep-pak C18 cartridge (Waters, Milford, MA, USA). The sample was then dissolved in 1.6 ml of 1/60 M phosphate buffer (pH 6.3) and used for the bioassay.

Results

1. Changes in soil and leaf xylem water potentials

The soil matric potential at a depth of 30 cm ranged from around -3.3 kPa to -8.1 kPa from March to the heading stage in the W-plot (Fig. 3). In the D-plot, it decreased gradually after irrigation had been suspended and fell to -51.3 kPa on April 17. The soil water potential in the W- and D-plots was similar and ranged from -7.4 kPa to -17.4 kPa at the ripening (grain filling) stage from April 27. In the C-plot, the soil matric potential changed similarly to that in the W-plot before the heading stage and was also very similar to those in the W- and D-plots after heading, except at the end of April and the end of May when the soil matric potential fell below -30 kPa because of limited rainfall.

The leaf xylem water potential in the D-plot changed similarly to that in the W-plot for about ten days after irrigation had been suspended (Fig. 4). It fell below that in the W-plot subsequently and was far lower on April 10 and 17, in particular. The leaf xylem water potential in the D- and W-plots decreased gradually as the grains of wheat ripened, but there was scarcely difference between these plots. The leaf xylem water potential in the C-plot resembled that in the W-plot before heading except on April 17, and tended to be lower than that in the W- and the D-plots after heading (data not shown).

The differences in soil water potential among plots in 1994 were similar to those in 1995 with the exception that the water potential in the D-plot decreased significantly at the end of May and the water potential in the C-plot was always the same as that in the W-plot.

2. Dry matter production, grain yield and yield components

In both 1994 and 1995, the dry weight of aboveground part at harvest in the D-plot was 17 to 32% higher than those in the W- and C-plots (Table 1). The dry weights from the W- and C-plots were almost equal. The grain yield in the D-plot was also 13 to 40% higher than those in the W- and C-plots. There was no significant difference in yield between the W- and C-plots. The harvest
Table 2. Yield components.

| Year | Plot | No. of panicles (m⁻²) | No. of kernels per panicle | No. of kernels (×10³ m⁻²) | 1000 kernel weight (g) |
|------|------|------------------------|---------------------------|---------------------------|------------------------|
| 1994 | W    | 714.7 a                | 32.5 a                    | 23.2 a                    | 27.2 a                 |
|      | D    | 757.2 a                | 32.4 a                    | 25.9 a                    | 30.2 a                 |
|      | C    | 692.5 a                | 31.7 a                    | 24.5 a                    | 29.6 a                 |
| 1995 | W    | 736.4 a                | 28.4 a                    | 20.8 a                    | 25.3 a                 |
|      | D    | 798.7 a                | 29.6 a                    | 23.6 a                    | 31.1 a                 |
|      | C    | 625.7 a                | 28.8 a                    | 18.1 a                    | 33.1 a                 |

Grain weight is given for moisture content of 12.5%. The number of panicles and kernels in 1994 were estimated from the grain yield, 1,000 kernel weight and the number of kernels per panicle. Values with different letters are significantly different at the 5% level for the respective years.

Fig. 5. Changes in leaf area index (LAI; A) and dry weight of aboveground parts (B) in plants in the W-plot (○) and the D-plot (●) in 1995.

Vertical bars represent standard deviations (n=2). An asterisk indicates significant difference at the 5% level between the W- and D-plots.

Fig. 6. Crop growth rate (CGR), net assimilation rate (NAR) and mean leaf area index (mean LAI) in plants of the W-plot (○) and the D-plot (●) in 1995.

Vertical bars represent standard deviations (n=2). An asterisk indicates significant difference at the 5% level between the plots.

3. Leaf area index, dry weight of aboveground parts, and growth analysis

Here, we describe only the results of experiments in 1995 because we obtained similar results in 1994. The leaf area index (LAI; Fig. 5A) of wheat plants was about 5 in both the W- and D-plots on March 13 prior to irrigation treatment. The LAI in the D-plot tended to be higher than that in the W-plot on April 27, at the heading stage. On June 1, at the ripening stage, the LAI in the D-plot tended to be still higher than that in the W-plot. The dry weight of aboveground parts was significantly heavier in the D-plot than that in the W-plot at the heading stage (Fig. 5B). The difference in dry weight between plots was even larger at the late ripening and harvest stages (Fig. 5B, Table 1).

The crop growth rate (CGR) of plants in the D-plot was significantly higher than that in the W-plot during the period from March 13 to April 27 and tended to be higher during the period from April 27 to June 1 (Fig. 6). The higher CGR in the D-plot resulted from a higher net assimilation rate (NAR) and a slightly higher
mean leaf area index (mean LAI) than those in the W-plot from March 13 to April 27. From April 27 to June 1, the higher CGR in the D-plot resulted from the higher NAR and also the higher mean LAI.

We investigated the morphological and physiological characteristics of plants in the W- and D-plots in order to identify factors responsible for the difference in NAR and LAI between the plants at the ripening stage. Even though a number of repeated measurements were taken, replicates were not taken into consideration in these measurements because the differences between replicates, in terms of growth and development, were small.

4. Root length

Differences in root length among soil layers in our experiments were smaller than those described in other reports (e.g., Box et al., 1989; Box and Rameur, 1993; Proffitt et al., 1985). However, the lengths of roots in the shallow soil layers were large and then they decreased until a depth of 60 cm, and increased again in the deeper soil layers. Roots in both W- and D-plots developed to a depth of 120 cm on March 20~22, about one week after the start of irrigation treatment (Fig. 7). The root length in the D-plot tended to be greater than that in the W-plot in many soil layers, and especially in soil layers from 40 cm to 80 cm and from 100 cm to 120 cm. The root length below a depth of 40 cm on April 20 and 21, just after irrigation treatment was completed, was greater in both plots. A difference in root length between the plots was observed at a depth of 40 cm to 120 cm, and especially in the layers from 40 cm to 80 cm and from 100 cm to 120 cm. The root length increased further in all soil layers in early June, at the ripening stage, in both plots. No further clear differences in root length were found except in the soil layer from 60 cm to 80 cm.

5. Chlorophyll content and photosynthetic rates of leaves

The estimated chlorophyll content of lower leaves differed between the plants in the W- and D-plots
before heading, during the irrigation treatment (Fig. 8).
The estimated content remained higher in the D-plot for the 5th and 4th leaves on March 27 and for the 7th leaves on April 8 and 9. The difference between the plants became marked during the ripening stage. The chlorophyll content tended to remain higher in the D-plot for the 7th, 8th and 9th leaves on May 3 and 4, and for all leaves on May 31.

The rate of photosynthesis was measured in the morning for eliminating the afternoon depression, if any (Fig. 9). The rate in lower leaves tended to decrease rapidly in plants in the W-plot during senescence, after heading, as compared with those in the D-plot. However, no differences in the rates of photosynthesis in lower leaves were observed between the plants before heading.

6. Resistance to water transport
We examined the resistance to water transport from roots to flag leaves of plants in the W- and D-plots during a period of intense transpiration during the daytime on a sunny day. The resistance tended to be lower in plants in the D-plot than that in the W-plot (Table 3). These results indicate that the capacity of plants for passive water absorption in the D-plot, with their better-developed root systems, was higher than that of plants in the W-plot (see also Hirasawa and Ishihara, 1991).

7. Exudation rate
The exudation rate was far higher in plants in the D-plot, with their better-developed root systems, than that of plants in the W-plot on May 4, at the middle-ripening stage (Table 4). These results indicate that the plants in the D-plot had roots with higher physiological activity than plants in the W-plot. The exudation rate from the plants in both groups decreased markedly on May 25, at the late-ripening stage.

8. Cytokinin activity in xylem exudate
The exudation rate tended to be higher and the mean concentration of cytokinin in terms of equivalence to BA was also tended to be higher in plants in the D-plot than in those in the W-plot (Table 5). As a result, cytokinin activity, calculated by multiplying the exudation rate by the concentration, was far higher in plants in the D-plot than in those in the W-plot.

Discussion
The growing season of grain crops usually extends for several months and sometimes for even longer. Growth conditions can have a considerable effect on dry matter production and yield, not only immediately but also in the long term, via the effects on both morphogenesis and physiology. We showed previously that summer field-crop plants develop vigorous shoots with poor root systems during vegetative growth under conditions of adequate soil moisture in the rainy season known as "Baiu", and that the poor root systems promote water stress and...
root length in the D-plot increased in deeper soil layers after the start of irrigation treatment, and this tendency leaves (data not shown). This difference resulted from the smaller reduction in than that in the W-plot at the heading stage (Fig. 5A). In addition, as compared with plants in the W-p10t, the reached and from the less extensive dying back of lower stem number after the maximum tillering had been potential in the daytime (Fig. 2) decreased to a greater extent in the D-plot. The factors responsible for the higher CGR in the D-plot were not clarified in this experiment which resulted from insertion of the minirhizotron tubes (Wiesler and Horst, 1994; Smit et al., 2000). The resistance to water transport in plants in the D-plot, with their better-developed root systems, was smaller than that in the W-plot (Table 3), indicating that plants in the D-plot had a greater capacity for water absorption (Hirasawa and Ishihara, 1991; Hirasawa et al., 1992a, b). Leaf senescence in the D-plot occurred later and the rate of leaf photosynthesis and the leaf area remained higher in the D-plot than in the W-plot (Figs. 5 and 9).

The CGR of plants in the D-plot was higher than that in the W-plot at the heading stage (Fig. 5A). This difference resulted from the smaller reduction in stem number after the maximum tillering had been reached and from the less extensive dying back of lower leaves (data not shown). It also resulted in a larger number of panicles on plants in the D-plot (Table 2). In addition, as compared with plants in the W-plot, the root length in the D-plot increased in deeper soil layers after the start of irrigation treatment, and this tendency became clearer at the heading stage. However, there were no distinct differences in root length between the plants at the late-ripening stage six weeks after the completion of the treatment. Nakagami et al. (1998) observed a significant difference in root system development between the wheat plants grown in the soil with moisture treatments similar to the present experiment. It was reported that, when soil moisture decreased, roots developed well especially in deeper soil, not only in the wheat (Box et al., 1989; Morita and Okuda, 1994; Proffitt et al., 1985) but also in soybean (Hida et al., 1995; Hirasawa et al., 1994, 1998), cowpea, rice (Angus et al., 1983), peanut, millet (Inanaga et al., 1996), and other plants. Wheat roots in the shallower soil in the present experiment were not so long, as in earlier studies by others (Box et al., 1989; Box and Ramseur, 1993; Proffitt et al., 1985), perhaps due to the dry and compacted surface of the soil, light leakage and temperature effects, which resulted from insertion of the minirhizotron tubes (Wiesler and Horst, 1994; Smit et al., 2000). The relation between water transport in plants in the D-plot, with their better-developed root systems, was smaller than that in the W-plot, with their better-developed root systems, was smaller than that in the W-plot, indicating that plants in the D-plot had a greater capacity for water absorption (Hirasawa and Ishihara, 1991; Hirasawa et al., 1992a, b). Leaf senescence in the D-plot occurred later and the rate of leaf photosynthesis and the leaf area remained higher in the D-plot than in the W-plot (Figs. 5 and 9).

The CGR of plants in the D-plot was higher than that in the W-plot for one month before heading, although the soil moisture (Fig. 1) and the leaf xylem water potential in the daytime (Fig. 2) decreased to a greater extent in the D-plot. One reason for the higher CGR in plants in the D-plot was their larger leaf area index, which resulted from the smaller reduction in stem number after the maximum tiller number stage and the less extensive dying back of lower leaves (Figs. 5, 6 and 8). Another reason was the higher NAR of plants in the D-plot (Fig. 6). The factors responsible for the higher NAR in the D-plot were not clarified in this experiment although it is likely that the delay in senescence of the lower leaves of the plants in the D-plot (Figs. 8 and 9) might have played a role. Even though wheat plants grew under low soil moisture conditions before heading, when their shoots developed rapidly, their leaf area tended to increase rather than decrease, and their NAR became higher. These features were clearly different.

Table 4. Xylem exudation rates (mg h-1) from the cut ends of main stems in 1995.

| Date       | W-plot | D-plot |
|------------|--------|--------|
| May 4 to 5 | 20.4 ± 8.3 | 31.4 ± 10.4 |
| May 25 to 26 | 1.3 ± 0.2 | 1.5 ± 0.2 |

Data represent means ± standard deviations (n = 10).

Xylem exudates were collected for about 15 h from the evening (1800) to the morning (0900) of the following day.

Table 5. Cytokinin activities of xylem exudates in 1995.

| Plot | Exudation rate (a) (pmol / 15 h) | Cytokinin concentration in exudate (b) (nM eq. BA) | Cytokinin activity in exudate (c) (pM eq. BA) |
|------|---------------------------------|---------------------------------|---------------------------------|
| W    | 8.0 ± 0.4                       | 4.9 ± 0.9                       | 39.2 ± 8.6                      |
| D    | 10.8 ± 1.8                      | 19.5 ± 19.6                     | 192.8 ± 174.0                   |

Xylem exudates were collected from forty stems, including tillers, for a single measurement from the evening of May 5 (1800) to the morning of May 6 (0900).

Data represent means ± standard deviations (n = 3).
from those of the growth of soybean plants (Hirasawa et al., 1994, 1998). Similar earlier experiments (Nakagami et al., 1998) indicated that a smaller reduction in photosynthesis with leaf aging, rather than a difference in canopy structure, contributes to the higher NAR in plants that are growing in the soil with decreased moisture. Our unexpected results might have resulted from a low transpiration rate of leaves and a high leaf xylem water potential due to a low atmospheric vapor pressure deficit in early spring, even in the soil with limited moisture (Figs. 1 and 4). Moreover, the photosynthetic rate of the wheat cultivar used for our experiments did not decrease until the leaf xylem water potential fell below -1.0 MPa (Hirasawa, 1999), which might also have contributed to the maintenance of a high NAR in the D-plot. Wheat plants are sometimes injured by heavy rains as a result of raised ground water levels and anaerobic conditions. When the ground water is raised to a level that is less than 30 cm below the surface of the soil, adverse effects can be observed (Noda and Ibaraki, 1961). The present experiments were conducted under conditions where the level of the ground water was more than 1.5 m below the soil surface and, also, the soil texture was not expected to cause serious drainage problems. We speculate that the increased dry matter production in the D-plot resulted from the enhancement of the growth of plants in the D-plot and not from the moisture damage to plants in the W-plot. However, the increase in soil moisture decreases the volume of air in the soil. This might induce some oxygen deficiency for root growth. Much irrigation also decreases available nutrients, such as nitrogen, due to leaching. This might induce the decrease in LAI and NAR. In our next report, we will discuss further details of the effects of a moderate reduction in soil moisture on dry matter production in wheat. In the present research, we can conclude at least that the conditions of precipitation in the wet period for about one month before heading could decrease dry matter production of wheat plants.

The most prominent factor responsible for the difference in CGR between plants in the W- and the D-plots after heading was the difference in senescence of plants. The LAI of plants in the D-plot remained high at the ripening stage (Fig. 5A). Moreover, the NAR in the D-plot at the ripening stage also remained high because the rate of photosynthesis remained high during senescence (Fig. 9). These results indicate that leaf senescence was suppressed in plants in the D-plot during ripening. A midday or afternoon reduction in the rate of photosynthesis on a clear day has also been observed in wheat plants after April (Koh and Kurumura, 1973), as in other crops (Hirasawa and Hsiao, 1999; Huck et al., 1983; Ishihara and Saito, 1987). The larger the resistance to water transport in plants, the larger the reduction in the rate of photosynthesis (Hirasawa et al., 1992a). The resistance to water transport in plants in the D-plot was smaller than that in the W-plot (Table 3). The reduction in the rate seems likely to have differed between the two plants in the late ripening stage when daytime leaf water potential decreased. But we did not take enough measurements on the reduction in the photosynthetic rate of plants.

The plants in the D-plot had more developed root systems than those in the W-plot (Fig. 7), and xylem exudation rate (Table 4), which reflects the physiological activity of roots, was higher in the D-plot than in the W-plot. Thus leaf senescence in the D-plot was delayed at the ripening stage. Plants with better-developed root systems are known to exhibit high physiological activity in their roots and delayed leaf senescence in rice (Jiang et al., 1988), soybean (Hirasawa et al., 1998), and maize (Kondo et al., 2000). Cytokinins inhibit leaf senescence (Jordi et al., 2000; Nooden et al., 1990; Van Staden et al., 1988) and larger amounts of cytokinins were transported from roots to shoots in cultivars of rice (Soejima et al., 1992, 1995) and sorghum (Ambler et al., 1992), when leaf senescence was delayed. In our present experiments, the cytokinin activities of xylem exudates from the roots of plants in the D-plot, with their better-developed root systems, were higher than those of plants in the W-plot (Table 5).

Our results suggest that a better-developed root system, high physiological activity of roots, and a large amount of cytokinins transported from roots to shoots might be related to the delayed leaf senescence and the maintenance of a large leaf area and high rate of photosynthesis in the plants in the D-plot. The capacity of roots for nitrogen absorption might also contribute to the maintenance of leaf area and a high rate of photosynthesis. Leaf senescence is delayed in cultivars of rice (Yamazaki et al., 1999) and maize (Kondo et al., 2000) that can absorb larger amounts of nitrogen than other cultivars. The actual status of nitrogen absorption and its effects on the maintenance of leaf area and photosynthesis during senescence remain to be examined, as well as the effects of cytokinins in the plants in the D-plot.

Our experiments showed that wheat plants in the D-plot, grown with low soil moisture for only one month before heading, when plants were growing rapidly, had better-developed root systems and their dry matter production after heading was greater than that of plants in the W-plot, grown with irrigation that reflected precipitation in an average year. We can expect that the moisture damage during this period promotes leaf senescence and decreases grain yield. Thus, in order to improve grain yield, it seems to be important to encourage the development of root systems, for example, by drainage during the rainy season. Furthermore, more attention should be paid to soil moisture conditions during ripening. The effects of soil moisture on plants after heading will be reported in a separate report.

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References

Ambler, J.R., Morgan, P.W. and Jordan, W.R. 1992. Amounts of zeatin and zeatin riboside in xylem sap of senescent and nonsenescent sorghum. Crop Sci. 32: 411-419.

Angus, J.F., Hasegawa, S., Hsiao, T.C., Liboon, S.P. and Zandza, H.G. 1983. The water balance of post-monsoonal dryland crops. J. Agric. Sci. 101: 699-710.

Biddington, N.L. and Thomas, T.H. 1973. A modified 

betacyanin bioassay for the rapid determination of cytokinins in plant extracts. Planta 111: 183-186.

Box, J.E. Jr., Smucker, A.J.M. and Ritchie, J.T. 1989. Minirhizotron installation techniques for investigating root responses to drought and oxygen stress. Soil Sci. Soc. Am. J. 53: 115-118.

Box, J.E. Jr. and Ramsay, E.L. 1993. Minirhizotron wheat root data: comparisons to soil core root data. Agron. J. 85: 1058-1060.

Briggle, L.W. and Curtis, B.C. 1987. Wheat worldwide. In E.G. Heyne ed., Wheat and Wheat Improvement. ASA, CSSA and SSSA, Madison. 1-32.

Gusta, L.V. and Chen, T.H.H. 1987. The physiology of water and temperature stress. In E.G. Heyne ed., Wheat and Wheat Improvement. ASA, CSSA and SSSA, Madison. 115-150.

Hida, Y., Hirasawa, T. and Ishihara, K. 1995. Differences in dry matter production and root system development between soybean cultivars under deficient soil moisture conditions. Jpn. J. Crop Sci. 64: 573-580.

Hirasawa, T. and Ishihara, K. 1991. On resistance to water transport to crop plants for estimating water uptake ability under intense transpiration. Jpn. J. Crop Sci. 60: 174-183.

Hirasawa, T., Tsuchida, M. and Ishihara, K. 1992a. Relationship between resistance to water transport and exudation rate and the effect of the resistance on the midday depression of stomatal aperture in rice plants. Jpn. J. Crop Sci. 61: 145-152.

Hirasawa, T., Gotou, T. and Ishihara, K. 1992b. On resistance to water transport from roots to the leaves at the different position on a stem in rice plants. Jpn. J. Crop Sci. 61: 153-158.

Hirasawa, T., Tanaka, K., Miyamoto, D., Takei, M. and Ishihara, K. 1994. Effects of pre-flowering soil moisture deficits on dry matter production and ecophysiological characteristics in soybean plants under drought conditions during grain filling. Jpn. J. Crop Sci. 63: 721-730.

Hirasawa, T., Takei, M. and Ishihara, K. 1995. A minirhizotron method for measuring root system of soybean plants growing in the field. Jpn. J. Crop Sci. 64: 78-85.

Hirasawa, T., Nakahara, M., Izumi, T., Iwamoto Y. and Ishihara, K. 1998. Effects of pre-flowering soil moisture deficits on dry matter production and ecophysiological characteristics in soybean plants under well irrigated conditions during grain filling. Plant Prod. Sci. 1: 8-17.

Hirasawa, T. and Hsiao, T.C. 1999. Some characteristics of reduced leaf photosynthesis at midday in maize growing in the field. Field Crops Res. 62: 53-62.

Hirasawa, T. 1999. Physiological characterization of the rice plant for tolerance of water deficit. In O.Boo, J.O'Toole and B.Hardy eds., Genetic Improvement of Rice of Water-Limited Environments. IRRI, Los Banos. 89-98.

Huck, M.G., Ishihara, K., Peterson, C.M. and Ushijima, T. 1983. Soybean adaptation to water stress at selected stages of growth. Plant Physiol. 73: 422-427.

Inanaga, S., Kitamura, H., Matsuzuka, A., Hirasawa, T. and Sugimoto, Y. 1996. Interspecific differences of drought tolerance among soybean, peanut and common millet during vegetative growth in Tottori Sand Dune. Sand Dune Res. 43: 29-35.

Ishihara, K. and Saito, K. 1987. Diurnal courses of photosynthesis, transpiration, and diffusive conductance in the single leaf of the rice plants grown in the paddy field under submerged condition. Jpn. J. Crop Sci. 56: 8-17.

Jiang, C.-Z., Hirasawa, T. and Ishihara, K. 1988. Physiological and ecological characteristics of high yielding varieties in rice plants. II. Leaf photosynthesis rates. Jpn. J. Crop Sci. 57: 139-145.

Jordi, W., Schapendonk, A., Davelaar, E., Stoopen, G.M., Pot, C.S., Van Rhiijn, J.A., Gan, S. and Amasino, R.M. 2000. Increased cytokinin levels in transgenic 

Pisum sativum-ipt 

tobacco plants have large direct and indirect effects on leaf senescence, photosynthesis and N partitioning. Plant Cell Environ. 23: 279-280.

Koh, S. and Kumura, A. 1973. Studies on matter production in wheat. I. Diurnal changes in carbon dioxide exchange of wheat plant under field conditions. Proc. Crop Sci. Soc. Jpn. 42: 227-235.

Kondo, A., Oosaka, T., Shigemori, I., Miki, K. and Hirasawa, T. 2000. Photosynthetic rate and nitrogen content of leaves and nitrogen content and cytokinin activity of xylem exudates in maize lines which differ in leaf color degradations after flowering. Jpn. J. Crop Sci. 69 (Extra 2): 112-113**.

Loss, S.P. and Siddique, K.H.M. 1994. Morphological and physiological traits associated with wheat yield increases in Mediterranean environments. Adv. Agron. 43: 107-153.

Ludlow, M.M. and Muchow, R.C. 1990. A critical evaluation of traits for improving crop yields in water-limited environments. Adv. Agron. 43: 107-153.

Morita, S. and Okuda, H. 1994. Effects of soil water condition on root development of wheat seedling with special reference to branching of primary seminal root. Jpn. J. Crop Sci. 63: 418-422.

Nakagami, K., Oosaka, T. and Hirasawa, T. 1998. Effects of soil moisture conditions from about one month before heading through ripening on dry matter production of wheat plants. Jpn. J. Crop Sci. (Extra 2): 112-113**.

Noda, K. and Ibaraki, K. 1961. Studies on ripening of wheat and barley in the warmer districts in Japan. 3. Relationship between ripening of wheat and barley and soil moisture content. Proc. Crop Sci. Soc. Jpn. 29: 251-252**.

Nooden, L.D., Singh, S. and Letham, S. 1990. Correlation of xylem sap cytokinin levels with monocarpic senescence in soybean. Plant Physiol. 93: 33-39.

Ohtani, Y. 1949. Studies on the growth hindrance in wheat and barley, caused by excessive soil moisture contents. (I) Effects of uprising ground water on the root development and the yield of wheat. Proc. Sci. Soc. Jpn. 18: 10**.

Proflitt, A.P.B., Berliner, P.B. and Oosterhuis, D.M. 1985. A comparative study of root distribution and water extraction efficiency by wheat grown under high- and low- frequency irrigation. Agron. J. 77: 655-662.

Smit, A.L., George E. and Groenwold, J. 2000. Root observations and measurements at (transparent) interfaces with soil. In A.L. Smit ed., Root Methods: A Handbook. Springer, Berlin. 236-
Soejima, H., Sugiyama, T. and Ishihara, K. 1992. Changes in cytokinin activities and mass spectrometric analysis of cytokinins in root exudates of rice plant (Oryza sativa L.). Plant Physiol. 100: 1724-1729.

Soejima, H., Sugiyama, T. and Ishihara, K. 1995. Changes in chlorophyll contents of leaves and in levels of cytokinins in root exudates during ripening of rice cultivars Nipponbare and Akenohoshi. Plant Cell Physiol. 36: 1105-1114.

Tennant, D. 1975. A test of a modified line intersect method of estimating root length. J. Ecol. 63: 993-1001.

Tokimasa, F. 1951. Research on the harm of the excessive moisture in the soil to barley and wheat. I. Harm of moisture in the various stage of wheat growth. Proc. Crop Sci. Soc. Jpn. 20: 171-173**.

Van Staden, J., Cook, E.L. and Nooden, L.D. 1988. Cytokinin and senescence. In L.D. Nooden and A.G. Leopold eds., Senescence and Aging in Plants. Academic Press, San Diego. 281-328.

Wiesler, F. and Horst W.J. 1994. Root growth of maize cultivars under field conditions as studied by the core and minirhizotron method and relationships to shoot growth. Z. Pflanzenernahr. Bodenk. 157: 351-358.

Yamazaki, T. 1952. Studies on the "excess-moisture injury" of upland crops in overmoist soil from view point of soil chemistry and plant physiology. Bull. Natl. Inst. Agri. Sci. B1: 1-98**.

Yamazaki, T., Ookawa, T. and Hirasawa, T. 1999. Casual factors of the difference in leaf nitrogen content at the ripening stage between rice cultivars, Akenohoshi and Nipponbare. Jpn. J. Crop Sci. 68 (Extra 2): 78-79***.

*In Japanese with English abstract.
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***In Japanese.