Root Growth of Two Soybean [\textit{Glycine max} (L.) Merr.] Cultivars Grown under Different Groundwater Level Conditions

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Abstract: Soybean yield is low in the fields with a low groundwater level during summer due to drought stress. By raising the groundwater level using Farm-Oriented Enhancing Aquatic System (FOEAS) the yield of soybean cultivar Sachiyutaka can be increased, but not that of Fukuyutaka. Here, we examined the effect of the groundwater level on root growth and its dynamics in these two cultivars. Three of the four experiments demonstrated that root elongation ceased just below groundwater level in both cultivars. However, when the groundwater level was kept at 35 cm or deeper, the root growth at an early growth stage was more vigorous at a deeper layer in Fukuyutaka than in Sachiyutaka, but at the mid-growth stage root growth in Sachiyutaka became similar to or exceeded that of Fukuyutaka. These results indicated that the optimum control technique for the groundwater level differed with the cultivar. The groundwater level for Sachiyutaka should be kept relatively high at an early growth stage. Further studies will be needed to clarify the optimum control technique for maximizing the yield of Fukuyutaka that have a fast root growth at an early growth stage.

Key words: Farm-Oriented Enhancing Aquatic System, Groundwater level, Minirhizotron, Root length density, Root weight density, Soybean.

In Japan, more than 80\% of soybean [\textit{Glycine max} (L.) Merr.] is cultivated in fields converted from paddy fields with hardpans that exist about 20 cm below the soil surface. The drainage speed of paddy fields is relatively slow because the paddy fields are constructed to keep the water level above the soil surface. In southwestern Japan, the optimum sowing time for soybean is during the rainy season. This wet weather lowers the germination and seedling rates and inhibits root growth into deep layers due to flooding. After the rainy season, the amount of rainfall in this area is generally small, causing drought stress to the soybean plants. The Farm-Oriented Enhancing Aquatic System (FOEAS) was developed in 2005 to solve the problems of soil water fluctuations (Wakasugi and Fujimori, 2009). This system has both subirrigation and underdrain systems and can thus keep the groundwater level constant at various heights (down to 35 cm to up to 10 cm from the soil surface), even in the rainy season and during drought.

The effects of the groundwater level on root growth and the water absorption patterns of root systems of soybean have been reported. Shimada et al. (1995) kept the groundwater level at 20, 40, and 70 cm using a lysimeter and revealed that the root systems of soybean elongated down to the depth of the groundwater level in all treatments. They also demonstrated that when the groundwater level was kept at 20 and 40 cm depths, the root length density was higher at the soil surface layer, but when it was kept at a 70 cm depth, root distribution peaked at the soil surface layer and just above the groundwater level. The latter finding was also reported by Reicosky et al. (1972) and Tanaka et al. (1994), who conducted deep (80 – 100 cm) groundwater level treatments. Reicosky et al. (1972) estimated that water absorption was dominant below 50 cm from the soil surface when the groundwater level was at 100 cm depth. In addition to the effects of deep groundwater, Nathanson et al. (1984), demonstrated that groundwater level treatments of 3 and 15 cm increased the root dry matter weight of soybean (cv. CPI26671) up to 2.6 and 1.4 times, respectively, at the mid-pod filling stage as compared with the control without controlling the groundwater level.

Previously, we examined the effect of FOEAS in southwestern Japan, in the fields under drought stress conditions with a low groundwater level during the...
summer season (Matsuo et al., 2013). The groundwater treatment by FOEAS increased the yield of Sachiyutaka, but did not have a clear effect on the yield of Fukuyutaka. Furthermore, the benefit of FOEAS on shoot growth in Sachiyutaka was observed at an early growth stage, but not in Fukuyutaka. Therefore, we hypothesized that these differences might be due to a difference in the root growth of the cultivar: Fukuyutaka had faster root growth, deeper root system or higher root length density than Sachiyutaka. To test this hypothesis, here we examined the root growth of the two cultivars in response to groundwater level by field, lysimeter and pot experiments. We discuss the optimum technique for controlling the groundwater level of each cultivar.

Materials and Methods

1. Plant materials

The soybean cultivars Fukuyutaka and Sachiyutaka were used in all experiments. The former was released in 1980 and has been cultivated widely in southwestern Japan. The latter was released in 2001, and is being used increasingly by farmers in this region.

2. Experiment 1: Field experiment 1

A field experiment was conducted in 2008 at the Kyushu Okinawa Agricultural Research Center (KARC), Chikugo, Fukuoka, Japan (33°12′N, 130°30′E, 10 m elevation) to measure the tap root depth of two cultivars grown without controlling the groundwater level. Although the experimental field was equipped with FOEAS, the groundwater level was not controlled; i.e. only drainage. The cultivation method was the same as used in our previous study (late sowing and without groundwater control in 2008; Matsuo et al., 2013). Briefly, the soil was lowland paddy soil (Typic Endoaquolls), and the experimental design was a randomized complete block design with three replications. Three seeds each of the two cultivars were sown at a spacing of 38 cm × 25 cm on 30 July 2008. The total area and each plot area was 94.0 and 15.7 m², respectively. The seedlings were thinned to two plants per hill after establishment. The fields received 5 g m⁻² P₂O₅ and 5 g m⁻² K₂O in the form of chemical fertilizer and 100 g m⁻² magnesium lime one day before sowing. Chemical fertilizer was applied and incorporated into the soil to a depth of 15 cm by rotary tillers and no additional fertilizer was applied. After sowing, three groundwater level treatments were conducted: the groundwater level was kept at 20 cm below the soil surface (20 cm), at 35 cm below the soil surface (35 cm), and not adjusted (Control). Intertillage and ridging (called “baido”), a normal practice in Japan to control lodging and weeds (Wada et al., 2006), was carried out at 25 DAS. Therefore, the soil surface after this practice was elevated to the level between the cotyledon and primary leaf (about 10 cm). Application of insecticides and herbicides, and manual weeding control were done as necessary.

Data for weather, groundwater level and soil water potential at 10 and 30 cm depths were described previously (Matsuo et al., 2013). Root systems at two positions per replicate down to a 30 cm depth from the soil surface were sampled with a soil sampler (length; 30 cm, inner diameter; 5 cm, HS-30S, Fujiwara Scientific, Co., Ltd., Tokyo, Japan) at 21 DAS, at the R1 stage (beginning to bloom; Fehr et al., 1971, 43 DAS for Fukuyutaka and 38 DAS for Sachiyutaka), at the R4 stage (pod setting stage, 64 DAS for Fukuyutaka and 59 DAS for Sachiyutaka), and at the R6 stage (full size seed in top four nodes, 89 DAS for Fukuyutaka and 81 DAS for Sachiyutaka) to determine the root length density (cm cm⁻²; RLD). Soil samples that contained hypocotyls were further divided into sections of 0 – 15 cm and 15 – 30 cm and stored in a refrigerator at 4°C before analysis. The soil was carefully removed from the root samples with tap water and the nodules were completely removed. The RLD was analyzed with a WinRHIZO (Regent Instruments, Montreal, QC, Canada). Root dry weight was determined after oven-drying at 80°C for at least 3 days and the root weight density (g cm⁻³; RWD) was then calculated.

3. Experiment 2: Field experiment 2

A field experiment was conducted in 2010 at the KARC to evaluate root growth in response to groundwater level controlled by FOEAS in the same fields as in Experiment 1. The cultivation method was the same as reported previously (Matsuo et al., 2013). Briefly, the soil was lowland paddy soil (Typic Endoaquolls). The experimental design was a split plot design with three replications. The sowing date was 16 July 2010 and planting density was 75 cm × 25 cm. Total area and that of each plot was 617.6 m² and at least 17.5 m², respectively. The fields received 5 g m⁻² P₂O₅, and 5 g m⁻² K₂O in the form of chemical fertilizer and 100 g m⁻² magnesium lime one day before sowing. Chemical fertilizer was applied and incorporated into the soil to a depth of 15 cm by rotary tillers and no additional fertilizer was applied. Just after sowing, three groundwater level treatments were conducted: the groundwater level was kept at 20 cm below the soil surface (20 cm), at 35 cm below the soil surface (35 cm), and not adjusted (Control). Intertillage and ridging (called “baido”), a normal practice in Japan to control lodging and weeds (Wada et al., 2006), was carried out at 25 DAS. Therefore, the soil surface after this practice was elevated to the level between the cotyledon and primary leaf (about 10 cm). Application of insecticides and herbicides, and manual weeding control were done as necessary.

To test this hypothesis, here we examined the root growth of the two cultivars in response to groundwater level by field, lysimeter and pot experiments. We discuss the optimum technique for controlling the groundwater level of each cultivar.

4. Experiment 3: Lysimeter experiment

A lysimeter experiment was conducted in 2010 at the KARC to nondestructively analyze the root growth dynamics. The lysimeters were 5 m in length × 2 m in width × 2 m in depth. Because of the limited number, only two groundwater level treatments (i.e., 20 and 35 cm) were
conducted. The groundwater level was maintained by adding tap water all the time and draining redundant water through rubber hoses. For each treatment, two lysimeters were used. One lysimeter consisted of seven rows. About two months before the experiment, three cellulose acetate butyrate minirhizotron observation tubes (180 cm long × 5 cm in diameter; Bartz Technol., Co., Santa Barbara, CA, USA) per lysimeter were installed at an angle of 45° from the vertical down to 50 cm. The section of the tube above the soil surface was wrapped with aluminum foil and capped with a rubber stopper. In each treatment, one lysimeter had three or four successive rows per cultivar and the other lysimeter was opposite from that of another lysimeter (i.e., if three rows were used in one lysimeter, four rows were used in the other lysimeter for each cultivar). One observation tube was set at the middle position of three rows and two tubes at the middle position of four rows. Three observation tubes were used for each treatment and cultivar. Three seeds each of two cultivars were sown at a spacing of 70 cm × 15 cm on 19 July 2010. The seedlings were thinned to one plant per hill after establishment. The amount of fertilizer and application method used was the same as in Experiment 1. A BTC 100X Minirhizotron Video Microscope (Bartz Technol., Co.) was used to collect the photographs (13.5 mm long × 18 mm wide) of the root systems at 20, 40 and 61 DAS. Photographs were taken at three positions (left, upper and right sides) per tube. Scanned images were analyzed by the line intersection method (Tennant, 1975) and the RLD calculated by this minirhizotron was designated RLDM (cm^3 cm^-2). One observation tube was regarded as one replication and average values of three positions were used for representative values of each replication. The results are shown at every 5 cm interval down to a 40 cm depth.

During the experiment, soil water potential at a depth of 15 cm only for Fukuyutaka was monitored with tensiometers (HD-001, Sensez Co., Ltd. Tokyo, Japan) throughout the experiment and the average value in the 20 cm, 35 cm and Control treatment during growth was −4.0 ± 0.2 kPa, −5.9 ± 0.3 kPa and −7.9 ± 1.3 kPa (mean ± standard error; n = 3), respectively. This result indicated that severe water stress was not imposed on the plants throughout the experiment. Three medium-sized plants from five pots were selected and their above-ground parts were removed at 21, 42 and 56 DAS. The pots were then set in a low-temperature chamber at 4°C before analysis. Soil samples were separated into 0 – 10, 10 – 20, 20 – 30, 30 – 40 and 40 – 50 cm sections. The soil was then carefully removed from the root systems with tap water and the nodules were completely removed. The RLD was analyzed with a WinRHIZO, which could also estimate root-diameter distribution. Roots were grouped by diameter, using the classification of Böhm (1979): very fine (< 0.5 mm), fine (0.5 – 2 mm), small (2 – 5 mm), medium (5 – 10 mm), large (10 – 20 mm) and very large (> 20 mm). We designated very fine and fine roots as thin roots and the others as thick roots. Root dry weight was determined after oven-drying at 80°C for at least 3 d and then the RWD was calculated.

5. Experiment 4: Pot experiment

The pot experiment was conducted in a climate-controlled chamber (14 hr light/10 hr dark cycle, 25°C, 70% humidity and a photon flux density of ~1000 μmol s^-1 m^-2). The pots (12.5 cm diameter and 55 cm long) were filled with 6.2 kg of air-dried lowland paddy soil collected near the KARC, and had four drainage holes (ϕ = 10 mm) perforated 3 cm from the bottom. The pots were filled with soil, but a 5 cm space was left at the top (0.99 g cm^-3 of bulk density). Before sowing, the pots were immersed in tap water and then excess soil water was drained. This process minimized the fluctuation of soil moisture condition at sowing. Four seeds were sown and the seedlings were thinned to one plant per pot after establishment. Three water treatments (20 cm, 35 cm and Control) were conducted just after sowing. Five pots were used for each treatment and cultivar. The pots were placed in water baths of 400 L (102 cm length; 86 cm width; 46 cm height) for the 20 and 35 cm treatments and in plastic trays (5 cm height) for the Control. The water baths used for the 20 and 35 cm treatment had drainage holes at the position of 20 and 35 cm, respectively. Float-type magnet valves were used to keep the water level stable. When the water level decreased below a certain threshold (for example, a threshold of 21 cm in the 20 cm treatment), the valve opened and tap water was added to the water bath. The water table was monitored with water level loggers (U20-001-04, Onset Computer Corp., Bourne, MA, USA) and the fluctuation of the water level was within 1 cm. Redundant water was drained through the drainage hole of the water bath. Soil water potential at a depth of 15 cm only for Fukuyutaka was monitored with tensiometers (HD-001, Sensez Co., Ltd. Tokyo, Japan) throughout the experiment and the average value in the 20 cm, 35 cm and Control treatment during growth was −4.0 ± 0.2 kPa, −5.9 ± 0.3 kPa and −7.9 ± 1.3 kPa (mean ± standard error; n = 3), respectively. This result indicated that severe water stress was not imposed on the plants throughout the experiment. Three medium-sized plants from five pots were selected and their above-ground parts were removed at 21, 42 and 56 DAS. The pots were then set in a low-temperature chamber at 4°C before analysis. Soil samples were separated into 0 – 10, 10 – 20, 20 – 30, 30 – 40 and 40 – 50 cm sections. The soil was then carefully removed from the root systems with tap water and the nodules were completely removed. The RLD was analyzed with a WinRHIZO, which could also estimate root-diameter distribution. Roots were grouped by diameter, using the classification of Böhm (1979): very fine (< 0.5 mm), fine (0.5 – 2 mm), small (2 – 5 mm), medium (5 – 10 mm), large (10 – 20 mm) and very large (> 20 mm). We designated very fine and fine roots as thin roots and the others as thick roots. Root dry weight was determined after oven-drying at 80°C for at least 3 d and then the RWD was calculated.

6. Statistical analysis

Statistical analysis was conducted by using SPSS v. 19.0 (SPSS 19.0, SPSS Inc., Chicago, IL). In Experiment 1, one-way analysis of variance (ANOVA) was conducted to test the effect of cultivar on rooting depth. Significant (P < 0.05) cultivar effects were explored by Fisher’s protected LSD. In experiments 2, 3 and 4, two-way ANOVA was conducted to test the effects of cultivar, groundwater level treatment and their interaction on each parameter. When the F test of ANOVA exceeded the 0.05 probability level, significance (P < 0.05) was explored by Fisher’s
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Results and Discussion

The rooting depth of Fukuyutaka and Sachiyutaka grown without groundwater is shown in Fig. 1. Fukuyutaka had a significantly \((P < 0.05)\) deeper tap root system than Sachiyutaka throughout the measurement period. The rooting depth of Fukuyutaka at 35 DAS was 56 cm, while that of Sachiyutaka was 43 cm. Manavalan et al. (2010) analyzed the tap root length of 8 cultivars by seedling assays which they developed, and demonstrated that the tap root length was 20 – 50 cm at 21 DAS and about 50 – 70 cm at 35 DAS. The values of tap root depth in Experiment 1 fell within the ranges reported by Manavalan et al. (2010). The greater potential of tap root elongation of Fukuyutaka may be more beneficial to acquire more water and nutrition at an early growth stage in field conditions, where the groundwater level is low.

However, no significant cultivar differences in RLD and RWD were observed in the field conditions (Experiment 2), except for RLD and RWD at 0 – 15 cm depth at the R4 stage (Table 1), and no significant effect of groundwater level on these traits was detected. There was no cultivar

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Table 1. Effects of cultivar and groundwater level treatment on root length density (RLD) and root weight density (RWD) at depths of 0 – 15 and 15 – 30 cm at 21 DAS, and at R1, R4 and R6 stages in Experiment 2.

| Stage | Cultivar | Groundwater | Soil depth (cm) | RLD (cm cm\(^{-3}\)) | RWD (g cm\(^{-3}\)) |
|-------|----------|-------------|----------------|---------------------|---------------------|
|       |          |             | 0 – 15         | 15 – 30             | 0 – 15              | 15 – 30              |
| 21 DAS\(^a\) | Fukuyutaka | 1.93        | 0.86           | 1.46                | 0.05                |
|        | Sachiyutaka | 1.74        | 0.97           | 1.42                | 0.05                |
|        | 20 cm     | 1.70        | 0.79           | 1.49                | 0.05                |
|        | 35 cm     | 1.82        | 1.12           | 1.56                | 0.06                |
|        | NT\(^e\)  | 1.98        | 0.83           | 1.27                | 0.06                |
| R1\(^b\) | Fukuyutaka | 1.14        | 0.44           | 7.12                | 0.27                |
|        | Sachiyutaka | 1.46        | 0.74           | 9.42                | 0.44                |
|        | 20 cm     | 1.50        | 0.64           | 7.96                | 0.19                |
|        | 35 cm     | 1.07        | 0.55           | 10.09               | 0.37                |
|        | NT        | 1.33        | 0.58           | 6.74                | 0.50                |
| R4\(^c\) | Fukuyutaka | 1.32 \(^a\) | 0.38           | 10.54b               | 0.16                |
|        | Sachiyutaka | 1.04 \(^a\) | 0.69           | 14.94a               | 1.22                |
|        | 20 cm     | 0.98        | 0.59           | 12.89               | 1.09                |
|        | 35 cm     | 1.21        | 0.60           | 13.26               | 0.41                |
|        | NT        | 1.35        | 0.41           | 12.08               | 0.57                |
| R6\(^d\) | Fukuyutaka | 1.42        | 0.90           | 9.51                | 0.27                |
|        | Sachiyutaka | 1.00        | 0.78           | 10.09               | 0.26                |
|        | 20 cm     | 1.38        | 1.06           | 9.68                | 0.08                |
|        | 35 cm     | 1.16        | 0.67           | 9.83                | 0.40                |
|        | NT        | 1.09        | 0.79           | 9.91                | 0.30                |

\(^a\) Days after sowing.
\(^b\) 43 and 38 DAS for Fukuyutaka and Sachiyutaka, respectively.
\(^c\) 64 and 59 DAS for Fukuyutaka and Sachiyutaka, respectively.
\(^d\) 89 and 81 DAS for Fukuyutaka and Sachiyutaka, respectively.
\(^e\) Non-treated control.
\(^i\) Means in columns followed by the different letters are significantly \((P < 0.05)\) different.
Table 2. Effects of cultivar and groundwater level treatment on root length density (RLD) measured with minirhizotron (RLDM) at 20, 40 and 61 DAS in Experiment 3 (lysimeter experiment).

| DAS* | Cultivar | Groundwater | Soil depth (cm) |
|------|----------|-------------|----------------|
|      |          |             | 0 - 5          | 5 - 10         | 10 - 15        | 15 - 20        | 20 - 25        | 25 - 30        | 30 - 35        | 35 - 40        |
| 20   | Fukuyutaka | 0.34 | 0.34 | 0.21 | 0.12 | 0.12 | 0.09 a | 0.06 | 0.02 a |
|      | Sachiyutaka | 0.35 | 0.30 | 0.14 | 0.11 | 0.12 | 0.01 b | 0.02 | 0.00 b |
|      | 20 cm     | 0.29 | 0.31 | 0.22 | 0.11 | 0.02 b | 0.02 | 0.00 b | 0.00 b |
|      | 35 cm     | 0.38 | 0.33 | 0.18 | 0.09 | 0.13 a | 0.08 | 0.06 a | 0.02 a |
| 20   | Fukuyutaka | 0.25 | 0.31 | 0.25 | 0.13 | 0.01 | 0.03 | 0.00 b | 0.00 b |
|      | Sachiyutaka | 0.43 | 0.37 | 0.17 | 0.10 | 0.23 | 0.15 | 0.11 aA | 0.03 aA |
| 20 cm| 20 cm     | 0.33 | 0.30 | 0.19 | 0.08 | 0.02 | 0.01 | 0.00 | 0.00 |
|      | 35 cm     | 0.37 | 0.30 | 0.09 | 0.15 | 0.22 | 0.00 | 0.03 B | 0.01 B |
| 40   | Fukuyutaka | 0.31 | 0.28 | 0.16 | 0.13 | 0.09 | 0.08 | 0.07 | 0.01 |
|      | Sachiyutaka | 0.49 | 0.28 | 0.23 | 0.25 | 0.17 | 0.11 | 0.04 | 0.00 |
|      | 20 cm     | 0.31 | 0.31 | 0.26 | 0.22 | 0.00 b | 0.01 b | 0.00 b | 0.00 b |
|      | 35 cm     | 0.45 | 0.37 | 0.26 | 0.20 | 0.09 a | 0.09 a | 0.07 a | 0.01 a |
| 61   | Fukuyutaka | 0.27 | 0.23 | 0.18 | 0.16 | 0.11 | 0.13 | 0.08 | 0.01 |
|      | Sachiyutaka | 0.38 | 0.21 | 0.19 | 0.25 | 0.19 | 0.19 | 0.14 | 0.01 |
|      | 20 cm     | 0.21 b | 0.24 | 0.20 a | 0.18 | 0.05 b | 0.02 b | 0.00 | 0.00 |
|      | 35 cm     | 0.31 a | 0.28 | 0.19 b | 0.18 | 0.12 a | 0.13 a | 0.08 | 0.01 |

* Days after sowing.

b Main effects are shown, but, means in columns followed by the different letters are significantly (P < 0.05) different.

c Simple main effects are shown, if significant cultivar × groundwater level treatment interactions on the RLDM are detected by two-way ANOVA. Means in columns followed by the different small or capital letters are significantly (P < 0.05) different within cultivar or groundwater level treatment, respectively.

× groundwater level interaction on them in Experiment 2 (data not shown). Shimada et al. (2012) conducted the field experiment with FOEAS in the Kanto region, in which groundwater levels were set at 20 and 32 cm depth and measured RLD after the R6 stage. They reported that RLD was concentrated in the top (0–15 cm) soil layer, regardless of groundwater levels, while RLD at depths of 15–30 cm tended to be higher in 32 cm treatment than in the other treatments. Interestingly, seed yield in 32 cm treatment was also higher than that in the control (without controlling groundwater table). Their results might at least partly explain why root growth at deeper layer contributes seed production. However, there was no consistent trend in the effect of groundwater level or cultivar on the root growth in Experiment 2, although we measured the root growth at various growth stages. A possible explanation was that intertillage and ridging were performed to control lodging and weeds, and thus the height of the ground surface was raised to about 10 cm. Therefore, the actual sampling depth might be about 20 cm, although we attempted to sample down to 30 cm. A modified sampling method should be used to detect the effect of groundwater level on the root traits in the field conditions; top soil layer (about 10 cm) had better be removed before root sampling, if intertillage and ridging were performed. At R4 stage, RLD in Fukuyutaka at a depth of 0–15 cm was significantly higher than that of Sachiyutaka, while the RWD in Sachiyutaka at this depth was significantly higher than that of Fukuyutaka. These results indicated that Fukuyutaka might possess more fine roots than Sachiyutaka and that Sachiyutaka had a thicker root system than Fukuyutaka at R4 in this study.

To further clarify the effects of cultivar and groundwater level on root traits, we conducted Experiments 3 and 4. In Experiment 3, the root growth dynamics was nondestructively analyzed with a minirhizotron at 20, 40 and 61 DAS (Table 2). At 20 DAS, a significant effect of cultivar on the RLDM was observed at depths of 25–30 cm and 35–40 cm. A significant effect of groundwater level was observed at depths of 20–40 cm, though not at 25–30 cm. A cultivar × groundwater level interaction on the RLDM was detected at 30 cm and deeper depths. In Fukuyutaka, RLDM in the 35 cm treatment was significantly (P < 0.05) higher than that in the 20 cm treatment while there was no significant difference in the RLDM among groundwater level treatments in Sachiyutaka. In the 35 cm treatment, RLDM in Fukuyutaka was significantly (P < 0.05) higher than that in Sachiyutaka. This result indicates that Fukuyutaka had a greater ability to expand its root systems at an early growth stage than Sachiyutaka, when the groundwater level was at a depth of 35 cm, while no significant cultivar difference of root growth was observed when the groundwater level was...
at a depth of 20 cm. No cultivar effect and cultivar × groundwater level interaction on RLDM were observed at 40 and 61 DAS, but RLDM in Sachiyutaka was higher than that in Fukuyutaka in both the 20 and 35 cm treatments at 40 and 61 DAS. The effect of groundwater level on RLDM was detected at 40 and 61 DAS. Because the root system hardly existed below a depth of 20 cm in the 20 cm treatment, RLDM below a 20 cm depth in the 35 cm treatment was higher than that in the 20 cm treatment.

The results obtained in Experiment 3 indicated that root growth of soybean ceased near the groundwater level and that Fukuyutaka had a greater root growth rate than Sachiyutaka below a 25 cm depth at an early growth stage, when the groundwater level existed at a 35 cm depth. This difference in root growth with the cultivar at an early growth stage partly coincided with the results in Experiment 1. Shimada et al. (1995) also reported that in the lysimeter experiment in which groundwater levels were

| DAS | Cultivar | Groundwater | RLD (cm cm⁻³) | 0–10 | 10–20 | 20–30 | 30–40 | 40–50 |
|-----|----------|-------------|---------------|------|------|------|------|------|
| 21  | Fukuyutaka | 20 cm       | 2.0           | 2.8  | 1.8  | 0.7  | 0.3 a |
|     | Sachiyutaka | NT         | 2.4           | 2.6  | 1.7  | 0.6  | 0.1 b |
| 21  | Fukuyutaka | 35 cm       | 2.6 a c       | 3.6 a | 0.9 b | 0.1 c | 0.0 c |
|     | Sachiyutaka | NT         | 2.1 b         | 2.4 b | 2.4 a | 0.6 b | 0.2 b |
| 21  | Fukuyutaka | 20 cm       | 1.9 c         | 2.2 c | 1.9 a | 1.2 a | 0.4 a |
|     | Sachiyutaka | NT         | 1.8           | 1.9  | 1.8  | 1.2  | 0.2  |
| 42  | Fukuyutaka | 20 cm       | 9.8           | 12.4  | 1.9  | 0.1  | 0.0  |
|     | Sachiyutaka | NT         | 4.6           | 6.5  | 10.0 | 3.4  | 0.2  |
| 42  | Fukuyutaka | 35 cm       | 2.8 b         | 3.9 b | 5.2 b | 5.9 a | 3.5 a |
|     | Sachiyutaka | NT         | 2.8           | 3.7  | 4.0  | 5.5  | 1.9  |
| 56  | Fukuyutaka | 20 cm       | 9.9 a         | 14.7  | 9.6  | 5.0 b | 2.3  |
|     | Sachiyutaka | NT         | 6.4 b         | 10.9  | 11.8 | 10.9 a | 3.4 |
| 56  | Fukuyutaka | 35 cm       | 15.4 a        | 21.6 a | 2.2 b | 0.0 c | 0.0 b |
|     | Sachiyutaka | NT         | 5.9 b         | 11.1 b | 17.7 a | 5.9 b | 0.4 b |
| 56  | Fukuyutaka | 35 cm       | 3.2 b         | 5.8 b | 12.3 a | 17.9 a | 8.1 a |
|     | Sachiyutaka | NT         | 21.4 a A a    | 27.7  | 2.4  | 0.9 b | 0.0 b |
| 56  | Fukuyutaka | 35 cm       | 6.2 b         | 13.0  | 20.3 | 5.9 ab | 0.6 b |
|     | Sachiyutaka | NT         | 2.2 b         | 3.5  | 6.2  | 9.2 ab | 6.3 ab |
| 56  | Fukuyutaka | 35 cm       | 9.3 ab        | 15.5  | 2.1  | 0.0 b | 0.0 b |
|     | Sachiyutaka | NT         | 5.6 ab        | 9.2  | 15.0 | 5.9 ab | 0.2 b |
| 56  | Fukuyutaka | 35 cm       | 4.3 b         | 8.1  | 18.4 | 26.6 aA | 9.9 aA |

\[\text{Table 3. Effects of cultivar and groundwater level treatment on root length density (RLD) at 21, 42 and 56 DAS in Experiment 4 (pot experiment).}\]

a Days after sowing.
b no treated control.
c For main effect, means in columns followed by the different letters are significantly \(P < 0.05\) different.
d Simple main effects are shown, if significant cultivar × groundwater level treatment interactions on the RLD are detected by two-way ANOVA. Means in columns followed by the different small or capital letters are significantly \(P < 0.05\) different within cultivar or groundwater level treatment, respectively.
kept at 20, 40 and 70 cm in depth the elongation of root systems of soybean (cv. Tamahomare) plants ceased just below groundwater level. Nagae (1972) also demonstrated that root systems of soybean (cv. Hachigatsu, Tamanishiki and Akiyoshi) did not elongate below the groundwater level. Although the number of cultivars whose root growth response to groundwater level was studied was only six, the results indicate that it may be common that root elongation of soybean ceases just below groundwater level, regardless of cultivar. This information will be useful for optimum control of groundwater level by the FOEAS.

In Experiment 4, Fukuyutaka and Sachiyutaka had similar RLD and RWD values at 21 DAS at 20 - 40 cm depths, regardless of groundwater level treatments (Tables 3 and 4). Although there were no significant differences between the two cultivars in RLD and RWD, Fukuyutaka had slightly larger values of RLD and RWD than Sachiyutaka at 40 - 50 cm. At 42 DAS, no significant groundwater level × cultivar interaction on RLD and RWD were found at any soil depths. However, Fukuyutaka

| DAS | Cultivar | Groundwater | RWD (g cm⁻³) | Soil depth (cm) |
|-----|---------|-------------|-------------|----------------|
| 21  | Fukuyutaka | 20 cm | 0.27 a | 0.09 | 0.05 | 0.02 | 0.01 a |
|     | Sachiyutaka | 20 cm | 0.23 b | 0.08 | 0.05 | 0.02 | 0.00 b |
|     |          | NT     | 0.24    | 0.07 | 0.06 a | 0.03 a | 0.01 a |
|     | Fukuyutaka | 20 cm | 0.25 | 0.12 | 0.03 | 0.00 | 0.00 |
|     |           | NT     | 0.28 | 0.07 | 0.06 | 0.03 | 0.02 |
|     | Sachiyutaka | 20 cm | 0.25 | 0.11 | 0.02 | 0.00 | 0.00 |
|     |           | NT     | 0.21 | 0.06 | 0.05 | 0.03 | 0.00 |
| 20 cm | 0.27 | 0.09 | 0.07 | 0.02 | 0.01 |
| 35 cm | 0.26 | 0.08 | 0.07 a | 0.02 b | 0.01 ab |
| NT     | 0.24 | 0.07 | 0.06 a | 0.03 a | 0.01 a |
| 20 cm | 0.27 | 0.09 | 0.07 | 0.02 | 0.01 |
| 35 cm | 0.26 | 0.08 | 0.07 a | 0.02 b | 0.01 ab |
| NT     | 0.24 | 0.07 | 0.06 a | 0.03 a | 0.01 a |
| 20 cm | 0.25 | 0.12 | 0.03 | 0.00 | 0.00 |
| 35 cm | 0.27 | 0.09 | 0.07 | 0.02 | 0.01 |
| NT     | 0.28 | 0.07 | 0.06 | 0.03 | 0.02 |
| 20 cm | 0.25 | 0.11 | 0.02 | 0.00 | 0.00 |
| 35 cm | 0.24 | 0.07 | 0.07 | 0.02 | 0.00 |
| NT     | 0.21 | 0.06 | 0.05 | 0.03 | 0.00 |

Table 4. Effects of cultivar and groundwater level treatment on root weight density (RWD) at 21, 42 and 56 DAS in Experiment 4.

*a Days after sowing.
*b no treated control.
*c For main effect, means in columns followed by the different letters are significantly (P < 0.05) different.
*d Simple main effects are shown, if significant cultivar × groundwater level treatment interactions on the RWD are detected by two-way ANOVA. Means in columns followed by the different small or capital letters are significantly (P < 0.05) different within cultivar or groundwater level treatment, respectively.
tended to have higher RLD and RWD than Sachiyutaka at depths of 30 – 50 cm in 35 cm and Control. These results partly supported the results obtained in Experiments 1 and 3, in which root growth of Fukuyutaka was greater than that of Sachiyutaka at an early growth stage. On the other hand, RLD in Sachiyutaka at depths of 20 – 50 cm, especially at 30 – 40 cm, was higher than those in Fukuyutaka at 56 DAS. A significant \((P < 0.05)\) difference in RLD among cultivars at depths of 30 – 50 cm was found only in the Control. Although groundwater level treatment was different in Experiments 3 and 4 (i.e., 35 cm in Experiment 3 and the Control in Experiment 4), root growth of Sachiyutaka might overtake that of Fukuyutaka at the middle growth stage when the groundwater level was below a depth of 35 cm. In addition, the effect of groundwater level on the RLD was clear. The depth of the peak RLD lowered as the groundwater level lowered, regardless of cultivar and growth stage (Table 3). However,
the effect of the groundwater level on the RLD did not always coincide with that on RWD. This was due to the fact that at least 62% of thick (i.e., heavy) roots were concentrated at the depths of 0–20 cm (Table 5), consequently RWD was larger at depths of 0–20 cm than at a deeper layer, in all cultivars and growth stages (Table 5). The distribution patterns of thin roots varied with the groundwater level treatment; the zone with the highest thin root rate shifted deeper as groundwater level lowered and this result was consistent with that of RLD. These results suggest that RWD does not reflect actual root distribution patterns in soybean.

The soil water potential at a depth of 15 cm in the 20 cm treatment was relatively high. This may cause anoxia stress for soybean plants. Nathanson et al. (1984) studied the effect of a groundwater level of 3 and 15 cm from the surface on physiology, growth, and yield of two soybean cultivars (cv. Fiskebry V and CPI26671) under glasshouse conditions. They reported that groundwater level treatments increased seed yield of two cultivars, especially in CPI26671, compared with that in the Control in which groundwater level was not controlled and frequent irrigations were applied to maintain field water capacity. The seed yield of CPI26671 in 3 and 15 cm treatments were 71 and 65% higher, respectively, than that in the Control. Their results indicated that soybean could tolerate to nearly saturated soil conditions. Shimada et al. (2012) reported recently that groundwater level kept at 20 cm with FOEAS increased soybean yield, as compared with the Control without controlling the groundwater level. Although a relatively high groundwater level is thought to be harmful to soybean due to anoxia, the reports by Nathanson et al. (1984) and Shimada et al. (2012) indicate that serious stresses which reduce growth and yield of soybean are not imposed on soybean even under the conditions of relatively high groundwater level.

In conclusion, the present experiments (except Experiment 2) clearly demonstrated that the root growth rate of Fukuyutaka was higher than that of Sachiyutaka at an early growth stage. This difference in root growth between the two cultivars might partly explain our previous finding that the groundwater level control was effective for Sachiyutaka, while its effect was unclear for Fukuyutaka. That is, the groundwater level control may be effective for Sachiyutaka whose root growth rate is low at an early growth stage, while its effect may be unclear in Fukuyutaka whose root systems are deep and large at an early stage. Ayars et al. (2006) suggested that the ideal groundwater level would be close to the bottom of the crop root zone early in the season and would recede as the root zone develops. Because the groundwater level can be raised at any time by FOEAS, it will be possible to control the groundwater level in accordance with root growth. Thus, root growth characteristics should be considered to achieve an optimum technique for controlling the groundwater level for each cultivar. This will lead to stable and high soybean production using FOEAS.

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