Differences in the Development of Lateral Roots among Several Cultivars in Japanese Radishes (*Raphanus sativus* L.) under Low Temperature Condition

Nobuyuki FUKUOKA

*Sand Dune Agricultural Experimental Station, Ishikawa Agricultural Research Center, Kaho, Ishikawa 929–1126, Japan*

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Seeds of cv. T-340 were sown in the field on 10 March and 9 April 1997. The lateral root (LR) production was progressively intensified in early sown plot (ESP) instead of late (LSP). Plants in the ESP produced roots with small mean weights, diameter and length as compared with those in the LSP. Early-sown seedlings were subjected to low soil temperature conditions, below approximately 12°C during the first growth stage. In a second experiment, seeds of cvs. T-340 and T-396 were sown in 2 experimental plots: a control plot and a high soil temperature plot (HTP). The minimum soil temperatures in the control were often remained below 10°C for about 25 days after sowing, but the temperatures in the HTP kept above 15°C during the growth period. When grown under the control, larger number of thickened LRs was observed in roots of cv. T-340, but roots of cv. T-396 did not exhibit this disorder at any soil temperature plot. In both cvs., hinder root elongation was observed in the control as compared with the HTP. In a third experiment, varietal difference in the occurrence of LR in roots of cvs. Oshin, T-340, T-396 and Tenpou was compared. Although no correlation was observed between the different cvs. and root growth, active meristematic activity was observed in the pericycle region outside the protoxylem in roots of susceptible cvs. Oshin and T-340. Conversely, this meristematic activity remained low in roots of resilient cvs. T-396 and Tenpou. These observations led to the hypothesis that the obstruction of root elongation caused by low soil temperatures (below 12°C) during the first growth period often results in activation of the basal region of the LR meristem in susceptible cvs. prompting LR development. However, in resilient cvs., the LR seldom occur, even when root elongation is inhibited by low soil temperatures that normally decrease the meristematic activity of this region.

Keywords: lateral root, meristematic activity, radish, soil temperature, varietal difference

INTRODUCTION

Japanese radish is an important vegetable crop in Japan that can be cultivated all year by correctly selecting the appropriate varieties. Generally, two differently timed crops, one sown from winter to spring and another from summer to autumn, are used for radish production. The properties related to the quality of radish grown in each of the two harvests are considerably different. Especially, when sown from midwinter to early in spring, thick and abundant lateral roots (LRs) appear to arise from the diarch longitudinal furrows and affect the value of marketable fresh roots.

Corresponding author: Nobuyuki Fukuoka, fax : +81-076-283-2204, e-mail : n-fuku@pref.ishikawa.jp
As for the occurrence of this physiological disorder, Oe et al. (1991) reported that the LR production was progressively intensified with advancing the sowing date in spring. Several investigators (Kawashiro and Takeda, 1986; Takeda et al., 1987) have reported that thermal conditions had a significant effect on the development of LRs, and that a larger number of thickened LRs tended to be produced when plants were exposed to low temperature conditions. Since low temperature conditions are known to affect optimal root growth adversely (Okayasu and Takahashi, 1987), the LRs production with growth retardant in roots due to low temperatures conditions may be largely responsible for this phenomenon. Conversely, it is well known that marked variation exists with respect to LR formation (Emura, 1996; Jinbo, 1992; Shiono et al., 1981). According to Okayasu and Takahashi (1987), root growth under low temperature conditions differs among varieties. This finding may indicate that the different responses by Raphanus varieties to different temperature conditions are largely due to variations in LR formation.

The present study sought to clarify the effect of thermal conditions on LR development, and to ascertain whether any correlations could be observed between varietal differences and root growth during the growth period.

MATERIALS AND METHODS

To examine the effect of culture conditions on the development of LRs, three experiments were conducted at the Experimental Farm of the Ishikawa Sand Dune Experimental Station.

Experiment 1. Effect of sowing date on the development of LRs
Raphanus sativus L. ‘T-340’ was sown in the field on 10 March and 9 April 1997 (designated as the ESP and LSP, respectively). Sixty g m\(^{-2}\) of dolomite, 25 g m\(^{-2}\) of N, P, O, and K, and 60 g m\(^{-2}\) of micro elemental manure containing 0.4% Mn, 0.3% B, 1.2% Fe and 0.03% Cu were applied approximately seven days before sowing. Each plot measured 1.5 m × 7 m and consisted of 3 rows per bed at a distance of 30 cm with hills spaced 30 cm apart, in an arrangement that resulted in a final density of approximately 7 plants m\(^{-2}\). Irrigation tubes were placed between the rows, and the beds were covered with white polyethylene mulch. On emergence, each hill containing three seeds was thinned to one. In the ESP treatment, the white polyethylene row cover (0.02 mm thick) applied at sowing time and suspended on arches that were 40 cm high 70 cm wide to form a tunnel above the rows for approximately 30 days after sowing (DAS). At 70 DAS, 15 plants were sampled randomly, and of those, 10 similar plants were selected. Leaf and root weight, root diameter and root length were measured. To examine the occurrence of LRs, LRs were classified into the following five groups by diameter (d): 0.5 mm ≤ d < 1 mm, 1 mm ≤ d < 1.5 mm, 1.5 mm ≤ d < 2 mm, 2 mm ≤ d < 2.5 mm, 2.5 mm ≤ d, respectively, and the number of roots within each category per plant was then counted. Daily minimum air temperature at a height of 30 cm and minimum soil temperatures at a depth of 10 cm were recorded throughout the growth period.

Experiment 2. Effect of high soil temperature treatment on the development of LRs
On 10 March, seeds of cvs. T-340 and T-396 were sown in 2 experimental plots in glass hot-houses: a control plot and a high temperature plot (HTP), each of which measured 1.5 m × 5 m. The application of fertilizers and culture conditions were the same as those used in Experiment 1. For the HTP, heating cables were laid at depths of 5 and 10 cm below the surface of radish-planted rows and a thermostat was set to keep the soil temperature above approximately 20°C from approximately 6 DAS to harvest. Control plants were grown under natural soil temperatures. Plant growth was measured at 20, 40, 61 and 73 DAS and LR formation was examined at 61 and 73 DAS. Daily minimum soil temperatures at a depth of 10 cm below the surface of the row were recorded.

Experiment 3. Varietal differences in the occurrence LRs
Seeds of cvs. Oshin, T-340, T-396 and Tenpou were sown in the field on 10 March 1997. The
amount of fertilizer and culture conditions were the same as those employed in Experiment 1. Plant growth was measured at 33 and 69 DAS and LR formation was examined at 69 DAS. For anatomical studies, roots of 33-day-old plants were sampled and the midregions of the roots were fixed, dehydrated, embedded in paraffin and cut longitudinally into 10 μm sections. The sections were stained with hematoxylin and eosin solution.

RESULTS

Experiment 1. Effect of sowing date on the development of LRs

The daily minimum soil temperatures in the ESP often remained below 10°C for approximately 22 DAS, and were 5 to 10°C lower than those in the LSP during the growth period (Fig. 1). Data describing the effect of sowing date on plant growth showed that the earlier sowing in spring prevented roots from growing well (Table 1). Plants in the ESP produced roots with small mean weights compared with those in the LSP. Likewise, the mean diameter and length of main root in the ESP were smaller than those in the LSP. Larger numbers of thickened LRs produced in the ESP compared to the LSP (Fig. 2). A significant increase in the number of LRs was observed in the following categories, 1 mm < diameter (d) < 1.5 mm, 2 mm < d < 2.5 mm, 2.5 mm < d, respectively.

Experiment 2. Effect of high soil temperature treatment on the development of LRs

The minimum soil temperatures in the HTP ranged from 14 to 25°C, with the lowest of 14.3 °C at 49 DAS (Fig. 3). Conversely, the temperatures in the control group were 5 to 15°C lower than those in the HTP and often remained below 10°C for about 25 DAS. The thermal conditions in soil largely affected root growth during the growth period (Table 2). In both varieties, except roots sampled from ‘T-396’ at 73 DAS, plants in the control group produced roots with low mean

![Fig. 1](image1.png) Changes in minimum air and soil temperatures during the growth period. Open and solid circles indicate temperatures when sowing occurred in March and April, respectively.

| Table 1 | The effect of sowing date on plant growth at 70 days after sowing. |
|----------------|----------------|----------------|----------------|
| Sowing date  | Leaf weight  (g) | Root weight (g) | Root diameter (cm) | Root length (cm) |
| March 10    | 307            | 733            | 6.2             | 35.1          |
| April 9     | 251            | 1377           | 7.6             | 45.5          |

Significance²

² NS and * indicate non-significance and significance at the P<0.05 level, respectively.
N. FUKUOKA

Fig. 2  The effect of sowing date on the occurrence of lateral roots at 70 days after sowing. Left and right bar graphs depict the results for plots sown March and April, respectively. Lateral roots were classified into the following five groups by diameter (d): 0.5 mm ≤ d < 1 mm (A), 1 mm ≤ d < 1.5 mm (B), 1.5 mm ≤ d < 2 mm (C), 2 mm ≤ d < 2.5 mm (D) and 2.5 mm ≤ d (E). NS and * indicate non-significance and significance at the P < 0.05 level, respectively.

Fig. 3  Changes in the minimum soil temperatures during the growth period. Open and solid circles indicate temperatures from the control and high soil temperature plots, respectively.

weights compared to those in the HTP. Likewise, the mean diameter and length in the control tended to be small as compared with those in the HTP. Plants of ‘T-340’ were characterized as having a large number of LRs which developed vigorously in plants exposed to low soil temperature conditions, while in ‘T-396’, there were fewer LRs and LR diameter rarely exceeds above 1.5 mm even under the low soil temperature conditions (Fig. 4).

Experiment 3. Varietal difference in the occurrence of LRs

Although ‘Oshin’ and ‘Tenpou’ sampled at 69 DAS appeared to have roots of greater width, statistical differences in root weight and/or root length were not evident among varieties (Table 3). Larger numbers of thickened LRs could generally be found in ‘Oshin’ and ‘T-340’ compared to ‘T-396’ and ‘Tenpou’ (Fig. 5). In the longitudinal sections of roots shown in Fig. 6, active proliferation of meristem tissue in the pericycle region outside the protoxylem point was observed in roots in ‘Oshin’ and ‘T-340’. Conversely, in ‘T-396’ and/or ‘Tenpou’, this meristematic activity was low even under low temperature conditions.
Table 2  The effect of soil temperature treatment on root growth during the growth period.

<table>
<thead>
<tr>
<th>Cultivar</th>
<th>Treatment</th>
<th>Root weight (g)</th>
<th>Root diameter (cm)</th>
<th>Root length (cm)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>20DAS</td>
<td>40DAS</td>
<td>61DAS</td>
</tr>
<tr>
<td>T-340</td>
<td>HTP</td>
<td>0.29a</td>
<td>14.2b</td>
<td>286a</td>
</tr>
<tr>
<td></td>
<td>CONT</td>
<td>0.11c</td>
<td>2.1c</td>
<td>185b</td>
</tr>
<tr>
<td>T-396</td>
<td>HTP</td>
<td>0.29a</td>
<td>21.7a</td>
<td>294a</td>
</tr>
<tr>
<td></td>
<td>CONT</td>
<td>0.16b</td>
<td>1.8c</td>
<td>176b</td>
</tr>
</tbody>
</table>

1 DAS: days after sowing. Means within a column with the same letter are not significantly different at the 5% level (Duncan’s multiple Range test).
Fig. 4 The effect of soil temperature treatment on the occurrence of lateral roots for the latter growth period. Solid and open columns indicate soil temperatures in the high temperature and control plots, respectively. Lateral roots were classified into the following six groups by diameter (d); 0.3 mm ≤ d < 0.5 mm (A), 0.5 mm ≤ d < 1 mm (B), 1 mm ≤ d < 1.5 mm (C), 1.5 mm ≤ d < 2 mm (D), 2 mm ≤ d < 2.5 mm (E) and 2.5 mm ≤ d (F). Means within a column with the same letter are not significantly different at the 5% level (Duncan’s Multiple Range test).

Table 3 Varietal difference of plant growth during the growth period.

| Cultivar | 33 DAS | | 69 DAS | |
|----------|--------|--------|--------|--------|--------|--------|--------|--------|
|          | LW (g) | RW (g) | RD (cm)| RL (cm)| LW (g) | RW (g) | RD (cm) | RL (cm) |
| Oshin    | 39a    | 7.0a   | 1.3a   | 18.8a  | 307a   | 733    | 6.2b    | 35      |
| T-340    | 36a    | 5.0bc  | 1.1c   | 14.6b  | 228c   | 730    | 5.7c    | 35      |
| T-396    | 29b    | 5.8ab  | 1.1c   | 18.9a  | 260b   | 733    | 5.8c    | 36      |
| Tenpou   | 22c    | 3.8c   | 0.9b   | 15.0b  | 268b   | 779    | 6.4a    | 36      |

*DAS: days after sowing. LW, RW, RD and RL represent leaf weight, root weight, root diameter and root length, respectively. Means within a column with the same letter are not significantly different at the 5% level (Duncan’s Multiple Range test).

Fig. 5 Varietal differences in the occurrence of lateral roots at the latter growth period. Lateral root was classified into the following five groups by diameter (d); 0.5 mm ≤ d < 1 mm (A), 1 mm ≤ d < 1.5 mm (B), 1.5 mm ≤ d < 2 mm (C), 2mm ≤ d < 2.5 mm (D) and 2.5 mm ≤ d (E). Means within a column with the same letter are not significantly different at the 5% level (Duncan’s Multiple Range test).
Fig. 6  Longitudinal sections of the mid. region of roots sampled from the cultivars of Oshin (A), T-340 (B), T-396 (C) and Tenpou (D), respectively. Each root was sampled on the 33rd day after sowing. Meristematic activity of cells at the basal portion of the lateral root differed among cultivars. LR and px indicate lateral root and protoxylem, respectively.

DISCUSSION

It is well known that the earlier seeds are sown in spring, the higher the occurrence of LRs will be in Japanese radish (Oe et al., 1991). Takeda et al. (1987) reported that plants subjected to a nighttime temperature regime of 3°C exhibited reduced LR production relative to plants with roots cultivated at −3°C. In this experiment, LR production tended to increase markedly and exhibit vigorous development when seeds of ‘T-340’ were sown early, as opposed to late, spring. In such instances, early-sown seedlings were subjected to a greater number of low soil temperature conditions, below approximately 12°C during the first growth stage than those of comparable ages in the LSP. Furthermore, in the early-sown crop, soil temperatures above 14°C appeared to have a lesser effect on LR production. These results strongly indicate that low soil temperatures act as a stimulus for LR production in ‘T-340’, and that cultivation at temperatures of approximately 12°C and below during the first growth stage often facilitates this phenomenon.

It is interesting to note that, in both cases, the low soil temperature condition inhibited optimal growth of the roots, especially with respect to root elongation. Takeda et al. (1987) demonstrated that the increase in the number of LRs was accompanied by a decrease in the length of the main root. In an experiment using daikon seeds that were sown from February to March, Moteki (1979) found that the elongation of roots sown later during this period occurred more rapidly than it did in the roots of those plants sown earlier. Furthermore, Kawashiro and Takeda (1986) illustrated that reducing root-zone temperature from 15°C to approximately 8°C resulted in decreased growth at the root. Since root elongation in daikon has been reported to be approximately 80% at 30 DAS
(Hayashi et al., 1958), it is likely that the suppression of root elongation during the first growth period in response to low soil temperatures is necessary for promoting LR production in the *Raphanus* root. In higher plants, if elongation of the main root is suppressed due to adverse conditions, a larger proportion of assimilative products tend to be transported in the direction of the lateral root branches instead of the root apex, resulting in active lateral development in this region (Russell, 1981).

Several investigators (Emura, 1996; Jinbo, 1992) have reported that the induction of LRs in *Raphanus* root varies among varieties. Similarly, the findings presented here revealed that varietal differences exist with respect to susceptibility of LR formation. In ‘T-396’, thermal conditions had no effect on LR production, which were rarely observed to develop even under low soil temperatures during the first growth period in spite of decreased root elongation. Furthermore, within the context of LR disorders, radishes sown in early spring can thus be classified as one of two types. One group develops LRs with high frequency and contains varieties such as Oshin and T-340, while the other is relatively resilient to LR production and is represented by varieties such as T-396 and Tenpou. However, there were no statistical differences in root growth between susceptible or resilient varieties. These findings show that different radish varieties exhibit markedly different responses to low soil temperature in so far as LR formation is concerned. In addition, the effect of low soil temperature on decreased root elongation due to increased LR development is relatively less apparent in the resilient cultivars.

In radish roots, LRs arise in the pericycle region outside the protoxylem, or in some cases slightly tangential to them, with the structure of LR resembling that of the non-fleshy region of the primary root (Hayward, 1951). Although root growth of Japanese radish is largely dependent on the active and continuous division and enlargement of parenchymatous cells (Watanabe, 1958), meristematic activity of these cells is known to differ among varieties (Kano and Fukuoka, 1994). In this experiment, active proliferation of parenchymatous cells was observed at the pericycle region outside the protoxylem in the susceptible varieties, while meristematic activity in the resilient varieties remained low in spite of earlier sowing. It therefore appears that the propensity of different varieties to produce LRs is closely correlated with the differences of meristematic activity at the basal regions of LRs.

In conclusion, the following assumption about the LR development can be made. If the susceptible varieties are subjected to low soil temperature (below 12°C) during the first growth period, fragilely root elongation due to low soil temperature results in active proliferation of meristematic tissue at the basal regions of the LRs. These enhanced meristematic activities often induce LR production. Conversely, in the resilient varieties, these cellular elements are not activated even when root elongation is suppressed by low soil temperatures and then active LR production seldom occur.

REFERENCES


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