

Relationship between Deep Root Distribution and Root Penetration Capacity Estimated by Pot Experiments with a Paraffin and Vaseline Layer for Landraces and Recent Cultivars of Wheat

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Abstract : Root growth into deep soil is an important factor for stable production in wheat under drought conditions. Root penetrating capacity (RP) shown by pot experiments with a paraffin-Vaseline layer (PV layer) may be a useful indicator estimating deep rooting ability of wheat genotypes. Previously, we identified genotypes of durum wheat (*Triticum turgidum* L. var. *durum*) and bread wheat (*T. aestivum* L.) with diverse RP by the pot experiments. In this study, we investigated the root distribution of three Ethiopian landraces of durum wheat with high RP, three recent cultivars of durum wheat with low RP and one Japanese cultivar of bread wheat 'Haruyutaka' with low RP using: (1) pots with a PV layer, (2) root boxes, (3) artificial field and (4) a normal field to analyze the relationship between RP estimated by pot experiment and root development in the field. In the pot experiments, RP was evaluated by the number of roots penetrating through the PV layer (NRP). In the root-box and field experiments, the root distribution was evaluated by the number of roots on the vertical surface of soil as the root frequency (RF: root number cm⁻¹ soil surface). Ethiopian landraces had a significantly larger NRP than recent cultivars in the pot experiment. The root box and field experiments showed that Ethiopian landraces tended to have a higher RF than recent cultivars in deep soil layer. We concluded that RP estimated by pot experiments with a PV layer is a useful indicator of deep rooting ability under field conditions.

Key words : Compacted soil, Deep rooting, Genotypic difference, Hard soil, Root penetration, Soil compaction, *Triticum aestivum*, *Triticum turgidum*.

Deep rooting is a beneficial trait because deep roots collect water from deep soil under drought conditions (Hurd, 1974; Zegada-Lizarazu et al., 2006, 2007). Reduced root-length density in deeper soil layers has been reported to affect the grain yields of bread wheat (Oussible et al., 1992). A simple screening method to evaluate deep rooting ability of wheat genotypes is required to increase drought resistance in wheat.

An experimental method to estimate the penetrating capacity of roots (RP) using pots having a layer made from a mixture of paraffin and Vaseline (PV layer) has been developed for rice (Yu et al., 1995), cotton (Klueva et al., 2000) and wheat (Botwright-Acuna and Wade, 2005; Kubo et al., 2004, 2005, 2006), and the genotypes of these crops with high RP have been identified. The success of this experimental method is depends on identifying genotypes with a higher ability of deep rooting in the fields (Clark et al., 2002), but such information is lacking in wheat. In this study, we investigated whether the genotypic difference in RP shown by pot experiments would correspond to the ability of deep rooting in the field in wheat. The number of roots penetrating through a PV layer for

each plant (NRP) evaluated by the pot experiment was compared with the number of roots on the vertical surface (root frequency (RF): root number cm⁻² soil surface) in a root box and fields using three Ethiopian landraces with high RP and three recent cultivars bred by International Maize and Wheat Improvement Center (CIMMYT) of spring durum wheat (*Triticum turgidum* L. var. *durum*), and one Japanese cultivar of spring bread wheat (*T. aestivum* L.), evaluated in previous studies by Kubo et al. (2004, 2006).

Materials and Methods

1. Materials

Three Ethiopian landraces and three recent cultivars bred by CIMMYT of spring durum wheat, and one Japanese cultivar of spring bread wheat 'Haruyutaka' were used for the pot experiment. Ethiopian landraces 'Et-23 A-1-97 (Et-23)', 'Et-31 A-1-113 (Et-31)' and 'Et-47 B-3-115 (Et-47)', had a high RP and CIMMYT cultivars 'Plover'S' (Plover)', 'Snipa'S' (Snipa)' and 'Bittern'S' (Bittern)' had low RP, respectively, in the pot experiments (Kubo et al., 2004, 2006). 'Haruyutaka' is a major cultivar in Hokkaido, Japan, bred at the

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This study was supported in part by a grant-in-aid (No.13760010 and 15380010) from the Japan Society for the Promotion of Science.

Abbreviations : NRP, number of roots penetrating through the PV layer; NRR, total number of seminal and crown roots reaching PV layer; PV, mixture of paraffin and Vaseline; RF, root frequency (root number cm⁻¹ soil surface); RP, root penetration capacity; RPI, root penetration index (NRP/NRR); TRN, total number of seminal and crown roots.

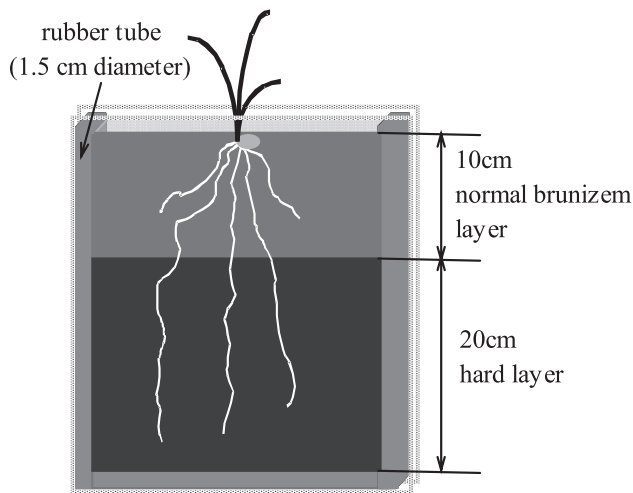


Fig. 1. Diagram of root box.

Hard soil treatment had a normal brunizem layer (0–10 cm depth) and hard soil layer (10–30 cm depth) while the control had only a normal brunizem layer.

Hokkaido Prefectural Agricultural Experiment Station in Kitami in 1985 and has a low RP (Kubo et al., 2006). Five or six genotypes of these seven genotypes were used in root box and field experiments.

2. Experiment 1 (Evaluation by pot experiments with a PV layer)

The experiment was done from May 2001 in a greenhouse at the Field Science Center for Northern Biosphere, Hokkaido University (Sapporo, Japan, 43°N, 141°E). The structure of pot, growth conditions of seedlings and experimental design were the same as those used by Kubo et al. (2006). Briefly, the pot consisted of a 10-cm tall cylinder placed on a 5-cm tall cylinder. The PV layer (3 mm thickness, 6 cm diameter) made from a mixture of 40% (in weight) paraffin and 60% Vaseline was placed between the two cylinders, and was fixed between the tubes. The cylinders were filled with non-compacted vermiculite. One seedling was grown in each pot. At six weeks after transplanting (approximately the heading stage of the seedling), number of roots penetrating through the PV layer by each plant (NRP) and total number of seminal and crown roots reaching PV layer by each plant (NRR) were counted. The root penetration index (RPI) was calculated as the proportion of NRP to NRR (Yu et al., 1995).

3. Experiment 2 (Evaluation in root box)

The experiment was done from September 2001, in the same greenhouse as in Exp. 1. The root box consisted of two transparent acrylic boards (35 cm width, 35 cm length, 3 mm thickness) and a rubber tube (15 mm diameter) sandwiched between the boards and fastened with clips at two points on each side (Fig. 1). Hard soil (compacted soil) and control

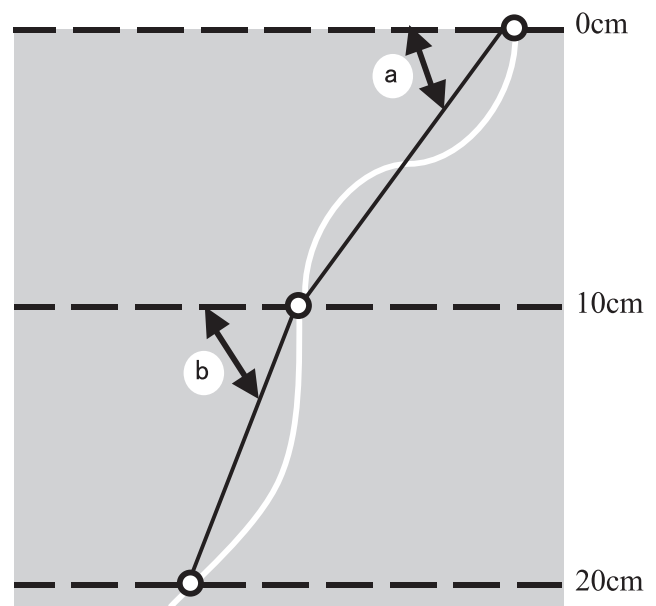


Fig. 2. Measurement of the growth angle of seminal roots.

The white curved line and bold dotted lines show the seminal root and each 10 cm depth, respectively. \circ is the intersection of the seminal root with each 10 cm line. The growth angle is defined as the angle between the horizontal line of each 10 cm depth and the line connecting each intersection of horizontal line and seminal root. a and b is the growth angle at depths 0–10 and 10–20 cm, respectively.

soil were used. For hard soil, the soil with a high clay content made from a mixture of 500 mL brunizem, 500 mL Bentonite (Calcium type, Kunimine Industries Co., Ltd., Tokyo, Japan) and 150 mL water was compacted at a depth of 10–30 cm and non-compacted normal brunizem layer was placed at a depth of 0–10 cm. As the control soil, only a normal brunizem layer was placed at a depth of 0–30 cm. In both hard and control soils, the soil bulk density was set at 1.40 g cm^{-3} . At the start of the experiment, 5.4, 9.0, 4.5 and 2.7 g m^{-2} of N, P_2O_5 , K_2O and MgO were applied. Three seeds of each genotype were sown at a depth of 1.0 cm, and the first seedling was allowed to continue growing. The soil was kept saturated with water by irrigation. The box was covered with aluminum foil to avoid increase in soil temperature by direct sunlight. Boxes were inclined at a 40° angle throughout the experiment so that roots elongated along the undersurface of the acrylic board. The experimental design was a completely randomized design with three replications.

At 23 days after sowing (DAS), root distribution, elongation rate of seminal roots, growth angle of seminal roots and the density of branching roots of seminal roots were measured. Root distribution was evaluated by a method similar to the profile wall method of Bohm (1976). Briefly, after dividing the acrylic boards into $1.0 \times 1.0 \text{ cm}$ grids, the number of visible roots in each grid was counted, and the root frequency (RF: root number cm^{-2} soil surface) was

Table 1. NRP, NRR and RPI surveyed by the pot experiment with a PV layer (Exp. 1).

	NRP ¹⁾	NRR ²⁾	RPI ³⁾
Et-23	19.0 ± 3.2 ^{4)ab5)}	18.7 ± 1.3 ^{ab}	1.00 ± 0.11 ^a
Et-31	15.7 ± 3.2 ^{abc}	21.3 ± 0.3 ^a	0.73 ± 0.14 ^{ab}
Et-47	22.7 ± 1.9 ^a	22.7 ± 1.3 ^a	1.00 ± 0.03 ^a
Plover	10.7 ± 1.5 ^{bc}	16.0 ± 1.0 ^b	0.68 ± 0.12 ^{ab}
Snipa	8.7 ± 2.0 ^{bc}	16.3 ± 0.9 ^b	0.54 ± 0.14 ^{ab}
Bittern	7.7 ± 1.9 ^c	18.7 ± 0.3 ^{ab}	0.41 ± 0.10 ^b
Haruyutaka	5.3 ± 1.2 ^c	16.0 ± 0.0 ^b	0.33 ± 0.08 ^b
P ⁶⁾	0.002	0.001	0.006
Ethiopian landraces ⁷⁾	19.1 ± 2.0	20.9 ± 2.0	0.91 ± 0.09
Recent cultivars ⁸⁾	9.0 ± 0.9	17.0 ± 1.5	0.54 ± 0.08
P ⁹⁾	0.010	0.055	0.037

¹⁾ Number of roots penetrating through the PV layer per plant.

²⁾ Total number of seminal and crown roots reaching PV layer per plant.

³⁾ Root penetration index = NRP / NRR.

⁴⁾ Mean ± Standard error (n = 3).

⁵⁾ Means followed by the common letters of each trait were not significantly different by the multiple test of Ryan-Einot-Gabriel-Welsch (P < 0.05).

⁶⁾ Probability of the difference among cultivars by ANOVA.

⁷⁾ Mean value in Ethiopian landraces (n = 3).

⁸⁾ Mean value in recent cultivars (n = 3).

⁹⁾ Probability of the significance of the difference between Ethiopian landraces and recent cultivars by the t-test.

calculated. The measuring area of RF was 5 cm on each side and 30 cm from the plant base on the lower side (10 cm width, 30 cm depth). The elongation rate of seminal roots (cm d⁻¹) was calculated by measuring the position of the root tips in every 24 hrs. The growth angle of seminal roots was defined as the angle between the horizontal line at every 10 cm depth and the line connecting each intersection of horizontal line and seminal root (Fig. 2). The branching root density of seminal roots (cm⁻¹) was calculated by counting the number of branching roots of more than 1 mm length. The soil hardness was measured for 500 mL clods of hard and control soil mixtures in the 500 mL glass beakers using Yamanaka's hardness tester (Fujiwara Scientific Co., Ltd., Tokyo, Japan) (Tada, 1987). Bulk density and water content of the soil clods at the beginning of experiment were the same as the soil mixture in the root box.

4. Experiment 3 (Evaluation in field with artificial soil layer)

The experiment was done from June 2001, in a greenhouse at the Field Science Center for Northern Biosphere, Hokkaido University on brown lowland soil (typic Udifluent) that was bare fallow in the previous year. First, we made an artificial soil tank. After removing the soil down to a depth of 30 cm, non-woven fabric was spread on the bottom and the walls. For the hard soil, soil with high clay content was made

by mixing the removed soil with 6% (in volume) of Bentonite and adequate water. This soil was placed in the tank at a depth of 10–30 cm, and pressure was applied by stamping over the soil. Then, the removed soil was dried and placed on the top of the hard soil at a depth of 0–10 cm. The control plots were filled with removed soil without stamping. Seedlings of six genotypes with a 5-cm long leaf sheath, which were germinated in cell-pots (each cell 3 × 3 × 4 cm), were transplanted to the artificial field in a split plot design (main plot - soil treatment; sub plot - genotype) with three replications. The planting distance was 20 cm between rows and 20 cm between plants. Each plot size was 0.6 m² (1 row × 15 plants). The number of plots was 36 (two treatments, six genotypes and three replications). Fertilizer (5.4, 9.0, 4.5 and 2.7 kg 10a⁻¹ of N, P₂O₅, K₂O and MgO, respectively) was applied just before transplanting. Pesticide and fungicide were applied to control aphid and ear scab. Weeds were removed frequently by hand to avoid disturbance of measurement of wheat root. Drip irrigation was applied daily at 1.25 L m⁻² to the rows of the plots. When all genotypes measured (93 days after transplanting), each replication was trenched down to a depth of 40 cm to measure the root distribution and soil hardness on the vertical surface. Soil hardness was measured at a depth of 10 cm in hard and control soils using Yamanaka's hardness tester. To evaluate the root distribution in the soil profile, vinyl sheets (10

Table 2. Root frequency (RF, cm⁻²)¹⁾ evaluated by the root box experiment (Exp. 2).

Depth (cm)	Genotype	Control	Hard	Hard/Control ²⁾
0–10	Et-23	1.30 ± 0.27 ³⁾ a ⁴⁾	1.48 ± 0.23 ^a	1.14
	Et-31	1.25 ± 0.34 ^a	1.63 ± 0.41 ^a	1.30
	Et-47	0.83 ± 0.10 ^a	0.99 ± 0.77 ^a	1.19
	Plover	0.85 ± 0.19 ^a	0.85 ± 0.29 ^a	1.00
	Snipa	1.07 ± 0.18 ^a	1.26 ± 0.30 ^a	1.18
	Bittern	1.01 ± 0.47 ^a	1.07 ± 0.21 ^a	1.06
	P ⁵⁾	0.545	0.445	
	Ethiopian landraces ⁶⁾	1.13 ± 0.10	1.37 ± 0.19	1.21 ± 0.05
	Recent cultivars ⁷⁾	0.98 ± 0.07	1.06 ± 0.12	1.08 ± 0.05
	P ⁸⁾	0.497	0.277	0.002
10–20	Et-23	1.56 ± 0.34 ^a	2.10 ± 0.18 ^a	1.35
	Et-31	1.48 ± 0.25 ^a	2.23 ± 0.27 ^a	1.51
	Et-47	1.41 ± 0.11 ^a	1.61 ± 0.11 ^{ab}	1.14
	Plover	0.67 ± 0.14 ^a	1.11 ± 0.14 ^{bc}	1.66
	Snipa	0.97 ± 0.09 ^a	0.61 ± 0.24 ^c	0.63
	Bittern	0.92 ± 0.21 ^a	1.11 ± 0.19 ^{bc}	1.21
	P	0.066	0.002	
	Ethiopian landraces	1.48 ± 0.04	1.98 ± 0.19	1.33 ± 0.11
	Recent cultivars	0.85 ± 0.09	0.94 ± 0.17	1.16 ± 0.30
	P	0.040	0.085	0.691
20–30	Et-23	2.39 ± 0.61 ^a	0.85 ± 0.52 ^a	0.36
	Et-31	1.08 ± 0.29 ^{ab}	0.67 ± 0.11 ^a	0.62
	Et-47	1.34 ± 0.52 ^{ab}	0.32 ± 0.13 ^a	0.24
	Plover	0.44 ± 0.11 ^b	0.52 ± 0.13 ^a	1.18
	Snipa	0.71 ± 0.02 ^{ab}	0.22 ± 0.09 ^a	0.31
	Bittern	0.59 ± 0.06 ^b	0.20 ± 0.08 ^a	0.34
	P	0.035	0.299	
	Ethiopian landraces	1.60 ± 0.40	0.61 ± 0.16	0.40 ± 0.11
	Recent cultivars	0.58 ± 0.08	0.31 ± 0.10	0.61 ± 0.29
	P	0.165	0.090	0.601

¹⁾ Number of visible roots on the soil surface.

²⁾ Relative value of the RF in the hard soil to that in the control.

³⁾ Mean ± standard error (n=3).

⁴⁾ Means followed by the common letters for each trait were not significantly different by the multiple test of Ryan-Einot-Gabriel-Welsch (P<0.05).

⁵⁾ Probability of the significance of the difference among genotypes by ANOVA.

⁶⁾ Mean value in Ethiopian landraces (n=3).

⁷⁾ Mean value in recent cultivars (n=3).

⁸⁾ Probability of the significance of the difference between Ethiopian landraces and recent cultivars by the t-test.

cm width, 30 cm depth for each plant) were placed to cover the vertical surfaces of the trench by fitting the hill and the upper center of the sheet, and visible roots of each plant were traced on the sheet by felt-tip pen. Then the sheet was divided into 1.0 × 1.0 cm grids, and the RF was calculated as in Exp. 2.

5. Experiment 4 (Evaluation in natural field)

The experiment was done from April 2002 in a field of the Field Science Center for Northern Biosphere, Hokkaido University. The preceding crop in the field was wheat. After plowing to about 20 cm depth, seeds of five genotypes were sown by hand in a randomized complete block design with three replications. The

planting distance was 25 cm between rows and 10 cm between plants. Each plot size was 3.0 m² (4 rows × 30 plants). Fertilization and control of insects, diseases and weeds were the same as in Exp. 3. At the panicle initiation (PI) stage (44–45 DAS) and maturing (MT) stage (108–109 DAS) of all genotypes, trenches were made vertically to the rows by using a power shovel down to depth 110 cm to measure the soil hardness and root distribution of each genotype, because roots of wheat reach down to depth 100 cm (Iwama and Yamaguchi, 1996). The trenches at PI stage and MT stage were made 2 m apart in each plot. Soil hardness was measured at every 2.5 cm in depth from the soil surface to 40 cm depth, and at every 5.0 cm depth from 40 to 100 cm depth in each replication by using Yamanaka's hardness tester as in Exps. 2 and 3. Visible roots of the central two plants of the four plants for each genotype were traced on vinyl sheets (25 cm width, 100 cm depth for each plot) to evaluate the RF of the vertical soil surface, as in Exps. 2 and 3. Grids of 2.5 × 2.5 cm were used to count the number of roots. The total number of seminal and crown roots (TRN) of each genotype was counted at the PI stage. Root length densities at depths of 7.5, 22.5, 37.5 and 60 cm were measured for each genotype of all replications by using the core-sampling method (Bohm, 1976) and the modified line intersect method (Tennant, 1975). The volume of core used in this study was 100 cm³.

6. Statistical analyses

All analyses of variances and correlation analyses of the measured traits were done by using software SPSS (Ver.7.5.1J, SPSS Japan, Tokyo, Japan). Genotypic difference of each characteristic was considered to be significant at 10% provability level.

Results

1. Experiment 1

The temperature of PV layer was from 15 to 25°C during the experiment, and its hardness was from 0.38 to 0.57 MPa, 0.45 MPa on average. NRP, NRR and RPI differed significantly among the genotypes (Table 1). The NRP in Ethiopian landraces was about twice as large as that in recent cultivars and 'Haruyutaka'. The NRR and RPI were also larger in Ethiopian landraces than in recent cultivars.

2. Experiment 2

Soil hardness measured by using clods was 0.05 ± 0.01 and 1.10 ± 0.03 MPa (mean ± standard error, n=3) in control and hard soils, respectively. The vertical soil surface was divided into three layers, 0–10, 10–20 and 20–30 cm in depth to compare the RF in each layer among genotypes. Genotypic difference of RF was significant at depths 10–20 cm in both treatments and 20–30 cm in the control (Table 2). The RF in Ethiopian landraces was higher than in recent cultivars

at the depth of 10–20 cm in both hard and control soils and at the depth of 20–30 cm in the hard soil ($P < 0.10$). The relative value of the RF calculated as the proportion of RF in the hard soil to RF in the control was higher in Ethiopian landraces than in recent cultivars at the depth of 0–10 cm. At the depths of 10–20 and 20–30 cm, there was little difference between Ethiopian landraces and recent cultivars. The growth angle of seminal root at the depth of 0–10 cm in hard soil treatment was larger in Ethiopian landraces than in recent cultivars (Table 3). The branching root density of seminal roots at the depth of 0–10 cm in the hard soil was significantly higher in recent cultivars than in Ethiopian landraces ($P < 0.10$). Other characteristics showed little difference between Ethiopian landraces and recent cultivars. There were no significant differences in the relative value (Hard/Control) of each characteristic between Ethiopian landraces and recent cultivars.

3. Experiment 3

Soil hardness at the depth of 10 cm was 0.20 ± 0.02 and 4.14 ± 0.57 MPa (mean ± standard error, n=3) for control and hard soils, respectively. Differences in RF among genotypes were significant at the depths of 10–20 and 20–30 cm in the hard soil ($P < 0.10$) (Table 4). In the comparison between Ethiopian landraces and recent cultivars, Ethiopian landraces had significantly higher RF than recent cultivars at the depth of 10–20 cm in hard soil. There was no significant difference in the relative values of RF in the hard soil to RF of the control between Ethiopian landraces and recent cultivars at any soil depth.

4. Experiment 4

The relationship between root length density, assessed by using the core-sampling method and then by the line-intersection method shown in Fig. 3 confirmed the effectiveness of RF for estimating the root distribution. The root length density and RF showed a significant positive correlation.

Fig. 4 shows the soil hardness at the different depths from the soil surface at the PI stage. Soil hardness increased with increasing depth, and was the highest value at 1.84 MPa at depth 30 cm. Below a depth of 30 cm, the hardness values decreased with increasing depth. We defined four soil layers depending on the degree of hardness: shallow layer (S layer; 0–22.5 cm depth) with low hardness; hard soil layer (H layer; 22.5–32.5 cm depth) with increasing hardness; below hard soil layer (BH layer; 32.5–42.5 cm depth) with decreasing hardness; deep layer (D layer; 42.5–100 cm depth) with low hardness.

In the comparison of root length density, there was no significant genotypic difference at any soil depth (Table 5). The genotypic difference in RF was significant at the BH layer in PI stage (Table 6). 'ET-

Table 3. Characteristics of seminal roots evaluated by a root box experiment (Exp. 2).

Group	Control	Hard	Hard/Control ¹⁾
<i>Seminal root growth angle (°: 0–10 cm depth)</i>			
Ethiopian landraces ²⁾	78.5 ± 0.6 ³⁾	80.9 ± 1.3	1.03
Recent cultivars ⁴⁾	77.5 ± 1.5	71.9 ± 1.7	0.93
P ⁵⁾	0.559	0.014	
<i>Seminal root growth angle (°: 10–20 cm depth)</i>			
Ethiopian landraces	79.8 ± 1.7	83.5 ± 0.2	1.05
Recent cultivars	74.7 ± 6.0	81.5 ± 1.5	1.09
P	0.461	0.253	
<i>Seminal root growth rate (mm d⁻¹)</i>			
Ethiopian landraces	17.23 ± 0.96	11.40 ± 0.30	0.66
Recent cultivars	16.80 ± 1.35	12.50 ± 0.58	0.74
P	0.807	0.176	
<i>Branching root density on seminal roots (cm⁻¹: 0–10 cm depth)</i>			
Ethiopian landraces	1.85 ± 0.10	2.38 ± 0.11	1.29
Recent cultivars	2.16 ± 0.44	2.81 ± 0.14	1.30
P	0.531	0.074	
<i>Branching root density on seminal roots (cm⁻¹: 10–20 cm depth)</i>			
Ethiopian landraces	3.99 ± 0.29	2.91 ± 0.03	0.73
Recent cultivars	3.69 ± 0.26	2.61 ± 0.38	0.71
P	0.501	0.474	
<i>Branching root density on seminal roots (cm⁻¹: 20–30 cm depth)</i>			
Ethiopian landraces	2.53 ± 0.28	1.06 ± 0.34	0.42
Recent cultivars	2.31 ± 0.43	0.95 ± 0.43	0.41
P	0.702	0.854	

¹⁾ Relative value of the RF in the hard soil to that in the control.

²⁾ Mean value in Ethiopian landraces (n=3).

³⁾ Mean ± Standard error (n=3).

⁴⁾ Mean value in recent cultivars (n=3).

⁵⁾ Probability of the significance of the difference between Ethiopian landraces and recent cultivars by the t-test.

47' was largest, and then 'ET-31', 'Bittern', 'Plover' and 'Haruyutaka'. In MT stage, RF significantly differed among genotypes in the H layer (P<0.10). Comparison between Ethiopian landraces and recent cultivars showed that Ethiopian landraces had significantly higher RF than recent cultivars in the BH (P<0.10) and D layers in MT stage. TRN at the PI stage of Ethiopian landraces was about twice as large as recent cultivars and 'Haruyutaka'.

5. Comparison of pot and field experiments

Simple correlation analyses were done between NRP in the pot experiment and RF in Exps. 3 and 4 (Fig. 5). NRP evaluated by pot experiment showed significant positive correlations with RF at depths 10–20 cm and

20–30 cm in the hard soil treatment in Exp. 3 and with RF in the BH layer at the PI stage in Exp. 4.

Discussion

The average hardness of the PV layer during the experiment was 0.45 MPa, which was similar to that of the PV layer used by Kubo et al. (2004, 2005, 2006) at 0.44–0.50 MPa to assess the RP in durum and bread wheat genotypes. The maximum RP of bread wheat, measured with a pot containing artificially compacted hard soil layer, is 0.30–0.40 MPa (Tanakamaru et al., 1998). The hardness of the PV layer in this study was suitable for surveying the RP of durum and bread wheat genotypes. Ethiopian landraces had larger NRP than recent cultivars. This result is the same as that in

Table 4. Root frequency (RF, cm²)¹⁾ evaluated in an artificial field (Exp. 3).

Depth (cm)	Genotype	Control	Hard	Hard/Control ²⁾
0–10	Et-23	2.34 ± 0.25 ³⁾ a ⁴⁾	1.92 ± 0.18 ^a	0.82
	Et-31	2.16 ± 0.63 ^a	1.60 ± 0.30 ^a	0.74
	Et-47	2.44 ± 0.36 ^a	2.00 ± 0.22 ^a	0.82
	Plover	2.01 ± 0.50 ^a	1.72 ± 0.15 ^a	0.86
	Snipa	2.74 ± 0.44 ^a	1.92 ± 0.41 ^a	0.70
	Bittern	2.34 ± 0.39 ^a	1.77 ± 0.17 ^a	0.76
	P ⁵⁾	0.729	0.866	
	Ethiopian landraces ⁶⁾	2.31 ± 0.08	1.84 ± 0.12	0.79 ± 0.03
	Recent cultivars ⁷⁾	2.36 ± 0.21	1.80 ± 0.06	0.77 ± 0.05
	P ⁸⁾	0.836	0.801	0.729
10–20	Et-23	1.30 ± 0.11 ^a	0.77 ± 0.09 ^a	0.59
	Et-31	1.42 ± 0.42 ^a	0.75 ± 0.18 ^a	0.53
	Et-47	1.60 ± 0.32 ^a	0.96 ± 0.07 ^a	0.60
	Plover	1.09 ± 0.04 ^a	0.57 ± 0.17 ^a	0.52
	Snipa	1.06 ± 0.25 ^a	0.64 ± 0.08 ^a	0.60
	Bittern	1.16 ± 0.17 ^a	0.59 ± 0.28 ^a	0.51
	P	0.216	0.075	
	Ethiopian landraces	1.44 ± 0.09	0.83 ± 0.07	0.57 ± 0.02
	Recent cultivars	1.10 ± 0.03	0.60 ± 0.02	0.55 ± 0.03
	P	0.022	0.032	0.450
20–30	Et-23	0.55 ± 0.32 ^a	0.36 ± 0.00 ^a	0.65
	Et-31	0.47 ± 0.20 ^a	0.15 ± 0.04 ^a	0.32
	Et-47	0.68 ± 0.39 ^a	0.56 ± 0.20 ^a	0.82
	Plover	0.45 ± 0.16 ^a	0.15 ± 0.05 ^a	0.33
	Snipa	0.69 ± 0.34 ^a	0.17 ± 0.05 ^a	0.25
	Bittern	0.53 ± 0.25 ^a	0.14 ± 0.07 ^a	0.26
	P	0.524	0.076	
	Ethiopian landraces	0.57 ± 0.06	0.36 ± 0.12	0.60 ± 0.15
	Recent cultivars	0.56 ± 0.07	0.15 ± 0.01	0.28 ± 0.03
	P	0.920	0.162	0.101

¹⁾ Number of visible roots on the soil surface.

²⁾ Relative value of the RF in the hard soil to that in the control.

³⁾ Mean ± standard error (n=3).

⁴⁾ Means followed by the common letters of each trait were not significantly different by the multiple test of Ryan-Einot-Gabriel-Welsch (P<0.05).

⁵⁾ Probability of the significance of the difference among genotypes by ANOVA.

⁶⁾ Mean value in Ethiopian landraces (n=3).

⁷⁾ Mean value in recent cultivars (n=3).

⁸⁾ Probability of the significance of the difference between Ethiopian landraces and recent cultivars by the t-test.

previous studies (Kubo et al., 2004, 2006) indicating that Ethiopian landraces had excellent RP compared with recent cultivars.

RF evaluated by counting the visible roots on the vertical soil surface in Exp. 4 and root length density measured by using core-sampling and then by using the line intersection method was positively correlated.

This suggested that RF could be an alternative method to evaluate the number of roots in field conditions in this study, as reported by Bohm (1976).

NRP was larger in Ethiopian landraces than in recent cultivars in the pot experiment (Exp. 1). The RF at the depth 10–20 cm in the control and hard soils evaluated in an artificial field (Exp. 3) was significantly

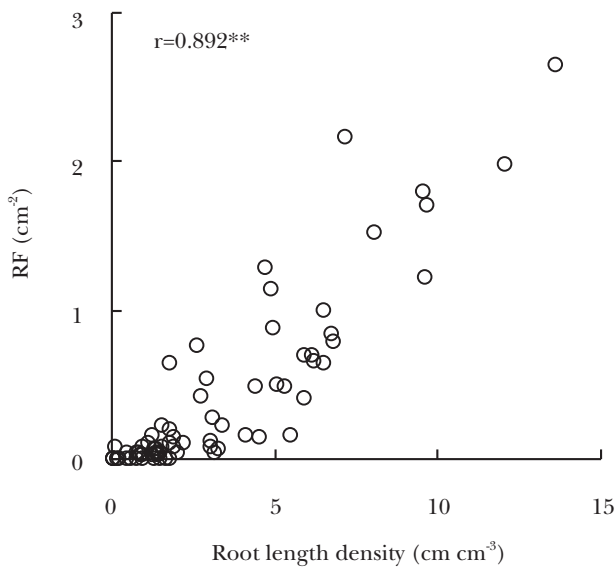


Fig. 3. Relationship between root length density evaluated by using the core-sampling method and RF evaluated by using the tracing method in a natural field (Exp. 4). RF indicates the number of visible roots on the soil surface. ** shows significant at $P < 0.01$.

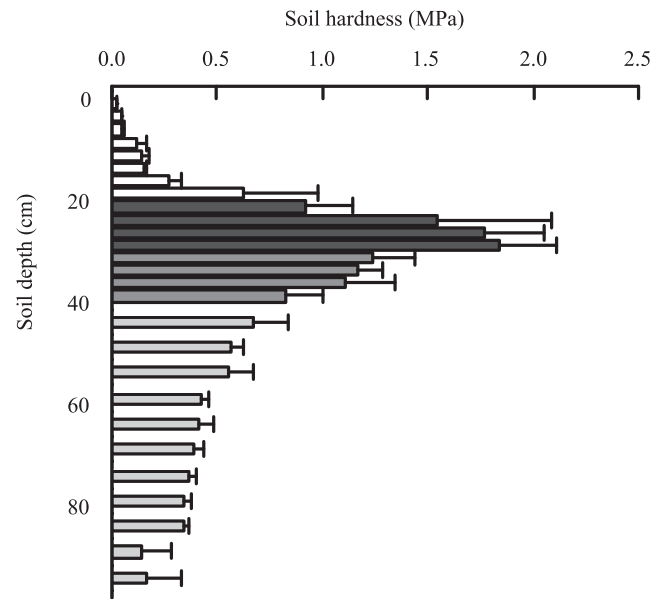


Fig. 4. Soil hardness at the PI stage in a natural field (Exp. 4). □ : Shallow layer (S layer; 0–22.5 cm depth), ■ : Hard soil layer (H layer; 22.5–32.5 cm depth), ▨ : Below the hard soil layer (BH layer; 32.5–42.5 cm depth), ▩ : Deep layer (D layer; 42.5–100 cm depth).

Table 5. Root length density measured by core-sampling method and the modified line intersect method at PI stage in a natural field (Exp. 4).

	Depth			
	7.5 cm	22.5 cm	37.5 cm	60.0 cm
Et-31	8.22 ± 2.71 ^{1) a2)}	3.21 ± 1.36 ^a	2.43 ± 0.78 ^a	1.69 ± 0.81 ^a
Et-47	7.20 ± 0.43 ^a	2.26 ± 0.58 ^a	1.48 ± 0.41 ^a	0.97 ± 0.40 ^a
Plover	6.65 ± 1.53 ^a	4.10 ± 1.53 ^a	1.08 ± 0.18 ^a	0.54 ± 0.27 ^a
Bittern	7.90 ± 2.14 ^a	3.82 ± 0.83 ^a	1.63 ± 0.13 ^a	0.62 ± 0.26 ^a
Haruyutaka	7.23 ± 1.34 ^a	2.97 ± 0.68 ^a	1.46 ± 0.16 ^a	0.49 ± 0.41 ^a
P ³⁾	0.800	0.454	0.139	0.349
Ethiopian landraces ⁴⁾	7.71 ± 0.51	2.74 ± 0.48	1.96 ± 0.48	1.33 ± 0.36
Recent cultivars ⁵⁾	7.28 ± 0.63	3.96 ± 0.14	1.34 ± 0.28	0.58 ± 0.04
P ⁶⁾	0.644	0.132	0.388	0.174

¹⁾ Mean \pm Standard error (n=3).

²⁾ Means followed by the common letters of each trait are not significantly different by the multiple test of Ryan-Einot-Gabriel-Welsch ($P < 0.05$).

³⁾ Probability of the significance of the difference among cultivars by ANOVA.

⁴⁾ Mean value in Ethiopian landraces (n=2).

⁵⁾ Mean value in recent cultivars (n=2).

⁶⁾ Probability of the significance of the difference between Ethiopian landraces and recent cultivars by the t-test.

higher in Ethiopian landraces than in recent cultivars. The RF at the depth of 10–20 and 20–30 cm of the hard soil in the artificial field (Exp. 3) showed significant correlations with NRP evaluated by the pot experiment (Exp. 1). The RF in the BH layer of the PI stage (Exp. 4) also was significantly correlated with NRP. These results suggest that a large NRP is positively correlated with deep rooting in the fields. The

NRP evaluated by the pot experiments with a PV layer can be a helpful indicator of the deep root distribution in the wheat genotypes. The large NRR and TRN in Ethiopian landraces in Exps. 1 and 4, respectively, may also contribute to deep root distribution, increasing the opportunity to grow through the cracks and biopores in the soil. In addition, the higher RPI in Ethiopian landraces than in recent cultivars in Exp. 1

Table 6. Root frequency (RF, cm⁻³) and TRN evaluated in a natural field (Exp. 4).

Genotype	RF ¹⁾				TRN ⁶⁾ (Plant ⁻¹)
	S layer ²⁾ (0–22.5 cm)	H layer ³⁾ (22.5–32.5 cm)	BH layer ⁴⁾ (32.5–42.5 cm)	D layer ⁵⁾ (42.5–100 cm)	
<i>PI stage</i> ⁷⁾					
Et-31	1.08 ± 0.31 ⁸⁾ a ⁹⁾	0.11 ± 0.02 ^a	0.06 ± 0.01 ^{ab}	0.01 ± 0.01 ^a	103.7 ± 5.3 ^a
Et-47	1.01 ± 0.15 ^a	0.25 ± 0.04 ^a	0.11 ± 0.04 ^a	0.02 ± 0.01 ^a	91.8 ± 5.5 ^a
Plover	1.10 ± 0.05 ^a	0.12 ± 0.02 ^a	0.03 ± 0.00 ^b	0.01 ± 0.01 ^a	58.0 ± 3.0 ^b
Bittern	0.90 ± 0.05 ^a	0.14 ± 0.05 ^a	0.06 ± 0.00 ^{ab}	0.01 ± 0.01 ^a	57.8 ± 7.4 ^b
Haruyutaka	1.68 ± 0.51 ^a	0.13 ± 0.05 ^a	0.02 ± 0.01 ^b	0.01 ± 0.01 ^a	55.5 ± 8.4 ^b
P ¹⁰⁾	0.375	0.271	0.045	0.596	0.001
Ethiopian landraces ¹¹⁾	1.05 ± 0.04	0.18 ± 0.07	0.09 ± 0.03	0.02 ± 0.00	97.8 ± 6.0
Recent cultivars ¹²⁾	1.00 ± 0.10	0.13 ± 0.01	0.04 ± 0.02	0.01 ± 0.00	57.9 ± 0.1
P ¹³⁾	0.712	0.571	0.280	0.845	0.022
<i>MT stage</i> ¹⁴⁾					
Et-31	0.79 ± 0.28 ^a	0.14 ± 0.06 ^a	0.09 ± 0.03 ^a	0.02 ± 0.00 ^a	
Et-47	0.79 ± 0.09 ^a	0.14 ± 0.03 ^a	0.07 ± 0.02 ^a	0.03 ± 0.02 ^a	
Plover	0.63 ± 0.19 ^a	0.16 ± 0.08 ^a	0.06 ± 0.03 ^a	0.01 ± 0.00 ^a	
Bittern	0.77 ± 0.24 ^a	0.14 ± 0.03 ^a	0.05 ± 0.01 ^a	0.01 ± 0.00 ^a	
Haruyutaka	0.76 ± 0.13 ^a	0.18 ± 0.08 ^a	0.06 ± 0.02 ^a	0.00 ± 0.00 ^a	
P	0.879	0.079	0.835	0.377	
Ethiopian landraces	0.79 ± 0.00	0.14 ± 0.00	0.08 ± 0.01	0.02 ± 0.00	
Recent cultivars	0.70 ± 0.07	0.15 ± 0.01	0.06 ± 0.00	0.01 ± 0.00	
P	0.343	0.586	0.055	0.034	

¹⁾ Number of visible roots on the soil surface.

²⁾ Shallow layer.

³⁾ Hard soil layer.

⁴⁾ Below the hard soil layer.

⁵⁾ Deep layer.

⁶⁾ Total number of seminal and crown roots by each plant.

⁷⁾ Panicle initiation stage.

⁸⁾ Mean ± Standard error (n=3).

⁹⁾ Means followed by the common letters of each trait are not significantly different by the multiple test of Ryan-Einot-Gabriel-Welsch (P<0.05).

¹⁰⁾ Probability of the significance of the difference among cultivars by ANOVA.

¹¹⁾ Mean value in Ethiopian landraces (n=2).

¹²⁾ Mean value in recent cultivars (n=2).

¹³⁾ Probability of the significance of the difference between Ethiopian landraces and recent cultivars by the t-test.

¹⁴⁾ Maturing stage.

may relate to the elongating ability of individual root. The little differences in the relative value (Hard/Control) of the RF between Ethiopian landraces and recent cultivars may suggest that the interaction between root distribution and soil condition is not significant among the genotypes used in this study.

Differences in RF in the root box (Exp. 2) and field experiments (Exps. 3 and 4) were relatively smaller than the difference in NRP evaluated by pot experiment (Exp. 1). In addition, every RF value in the field experiments had larger standard errors than that in the root box experiment (Exp. 2). This may be due to characteristics of the roots and soil. The structure

of the root system in field conditions is determined by the growth angle, length and branching pattern of roots (Fitter, 1987). Genotypic differences in the growth angle (Oyanagi et al., 1991), length (Sharma and Lafever, 1992) and branching pattern (Blaha and Janacek, 1997) of wheat roots were reported. The root box experiment in this study showed that recent cultivars had a higher density of branching roots on seminal roots than Ethiopian landraces. The relatively small difference in RF among genotypes in the root box and field experiments may have been caused by the genotypic differences of characteristics independent of RP. Cracks and biopores can be an

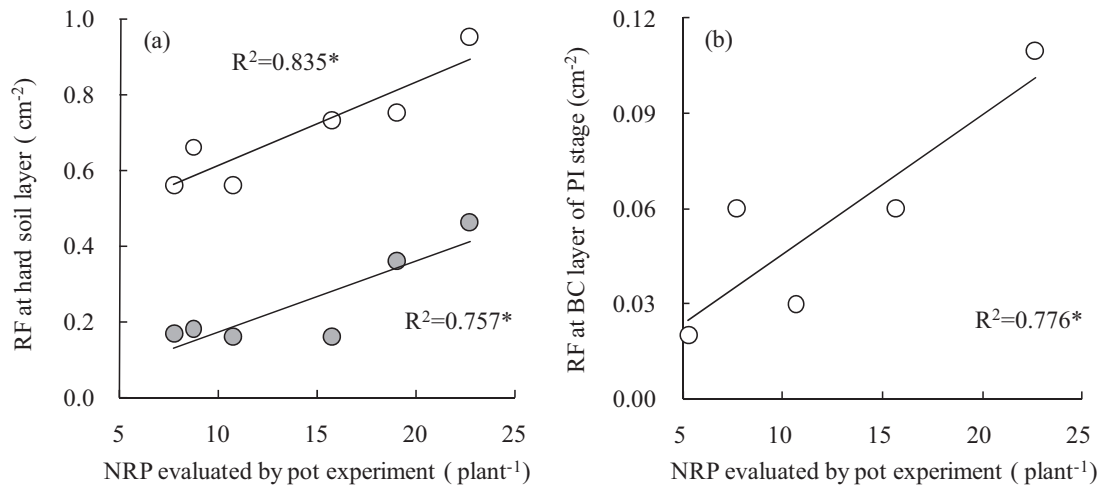


Fig. 5. Relationship of NRP surveyed in the pot experiment (Exp. 1) with RF in the compacted soil layer in an artificial field (Exp. 3) (a) and below the hard soil layer in a natural field (Exp. 4) (b).

NRP indicates the number of roots penetrating through the PV layer by each plant. RF indicates the number of visible roots on the soil surface. In (a), ○ : Soil layer at 10–20 cm depth; ● : Soil layer at 20–30 cm depth. * shows significant at $0.01 \leq P < 0.05$.

alternative route of plant roots to form a deep root system, because roots can elongate in the cracks and biopores to avoid the mechanical impedance (Ehlers et al., 1983; Wang et al., 1986; Nakamoto, 2000). The relatively small difference in RF among genotypes in the root box and field experiments may have been caused by such gaps in the soil or between soil and acrylic board.

The aboveground growth and yield of these wheat genotypes had little relationship with root distribution in Exp. 4 (data not shown). This may be because the deep root distribution related to RP was not a major contributor to aboveground growth in the tested conditions (e.g. sufficient precipitation, about 300 mm in the growth period). In west Asia and northern Africa, wheat is sown at the end of the wet season, so that the wheat grows in the dry season. Deep rooting ability of Ethiopian landraces may be a critical factor for aboveground growth in environments where soil water stored in deep soil is a major water source for plants, such as west Asia and northern Africa.

In conclusion, RP evaluated by the pot experiment may be a helpful indicator of the deep root distribution in the wheat genotypes. The landraces used in this study may be useful genetic resources to improve root traits of modern cultivars of durum wheat. Quantitative trait loci that control RP in wheat have been detected (Kubo et al., 2007). Improvement of deep root penetration in wheat can be accelerated by the combination of field evaluation, pot screening and marker-assisted selection.

Acknowledgements

Special thanks are due to I. Furumichi of the Research Faculty of Agriculture, Hokkaido University

for her excellent technical assistance in data collection, and A. Yanagisawa of the Hokkaido Prefectural Kitami Agricultural Experiment Station and T. Tominaga of the Graduate School of Agriculture, Kyoto University for technical advices. Experimental management was conducted by N. Moki and S. Ichikawa of the Field Science Center for Northern Biosphere, Hokkaido University.

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