Phenotypic Variations of Seedling Root Architectural Traits of Wheat Species under Different Growth Media

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Abstract

Understanding of the behaviour and the function of root system architecture (RSA; the spatial configuration of a root system) is critically important to improve water and nutrient uptake of the plant. However, root traits are highly dependent upon the soil type and soil moisture content. Therefore, the present study, consisting of two experiments, was carried out under controlled environment conditions to identify phenotypic variations of early seedling root architectural traits of three ancient wheat species together with modern bread wheat. In the first experiment, two week old vertically grown root systems on germination paper, soaked in nutrient media, were imaged and images were analysed using root phenotyping software *RootNav*. Seedling root architectural traits of two week old seedlings grown in compost and sand were compared in the second experiment where root samples were scanned and analysed using *WinRHIZO* software. Both experiments were set up according to completely randomised design with 20 replicates. Number of seminal roots, total root length, maximum width, maximum depth, convex hull, tip angle of the seminal roots, emergence angle of the seminal roots were significantly different between genotypes in the first experiment where the greatest number of seminal roots and the largest tip angle leading to the widest root systems was observed in emmer 2. Spelt and bread wheat genotypes produced moderate numbers of seminal roots and deep-narrow root systems. There was a significant interaction between genotype and media for number of seminal roots, total root length, root volume, root diameter, root biomass, root to shoot ratio and specific root length in the second experiment.

Keywords: Ancient wheat, Growth medium, Image analysis, Modern bread wheat, System architecture
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Introduction

Understanding of the behaviour and the function of root system architecture (RSA; the spatial configuration of a root system) is critically important to improve water and nutrient uptake of the plant. The root system plays a major role in determining soil exploration, nutrient acquisition and providing anchorage to the plant while maintaining plant-soil microbe interactions (Hirel *et al.*, 2007). Plants have an ability to adjust their morphology, physiology, growth and development with the influence of the growth conditions around them. Modification of RSA is one of the adaptations to survive under resource limited conditions. The growth and development of root architecture of the plant species modify RSA enabling them to uptake nutrients by proliferating lateral roots hence increase foraging capacity within nutrient rich soil layers (Walch-Liu *et al.*, 2006). Consideration of plant productivity and RSA under different environmental conditions is therefore important (Wu *et al.*, 2005). The present study was carried out to identify phenotypic variations of early seedling root architectural traits of ancient wheat species together with modern bread wheat under different growth conditions.

Materials and Methods

Two controlled environment experiments were set up to study early seedling root architecture of three hulled wheat species of cultivated einkorn (*Triticummonococcum* L), cultivated emmer (*T. dicoccum*) and spelt (*T. spelta* L), together with modern bread wheat (*T. aestivum*) in different growth media. The experiments were conducted at Sutton Bonington Campus, University of Nottingham, United Kingdom. Ten genotypes were used in the first experiment; three einkorn (1, 2 and 3), two emmer (1 and 2), three cultivars of spelt (SB, Oberkulmer and Tauro) and two cultivars of modern bread wheat (Xi 19 and JB Diego) where they grew vertically on germination paper (GP), soaked in liquid nutrient mediain a controlled environment room. Two week old seedlings were imaged using a Nikon D5100 DSLR camera and images were analysed using root phenotyping software *RootNav* (Pound *et al.*, 2013). In the second experiment, root system architecture of fifteen genotypes, including ten genotypes used in the previous experiment and five more bread wheat

cultivars (Hobbit, Glasgow, Gallant, Forno and Beaver) were compared in sand and compost medium. The experiment was conducted in the glasshouse under natural day-light. Root samples of two week old seedlings were digitized at 400 dpi resolution and 256 grey contrasts (Tiff format) with a scanner with transparency adapter (WinRHIZO STD 1600+, Regent Instruments Inc., Quebec; Canada). The scanned images of the root systems were analysed using WinRHIZO regular V.2002c software (WinRHIZO STD 1600 +, Regent Instruments Inc., Quebec; Canada). The experiments were set up according to completely randomised design with 20 replicates.

The software used in experiment one produced the readings of number of seminal roots, average length of seminal roots, total root length, tip angle and emergence angle of seminal root, maximum width, maximum depth, width to depth ratio and convex hull. Total root length, root volume, average root diameter, root to shoot ratio (R:S) and root length in different diameter classes (less than 0. 5 m, 0.5 to 1 mm, 1 to 1.5 mm, 1.5 to 2 mm, 2 to 2.5 mm, 2.5 to 3mm, 3 to 3.5 mm, 3.5 to 4 mm, 4 to 4.5 mm and more than 4.5 mm) were recorded in the second experiment. However, due to complexity, only root length at less than 0.5 mm diameter (very fine roots), 0.5 to 1 mm (fine roots) and sum of the values of all other classes as more than 1 mm (thick roots) were considered. Specific root length (SRL) was calculated based on original data of the root scanning results.

Results and Discussion

The greatest number of seminal roots (5.27, 7.30, 7.40 in germination paper [GP], compost and sand, respectively at $P < 0.001$) was recorded in emmer 2, while the lowest number of seminal roots on GP (3.75) and sand (4.60) was observed in bread wheat cv. JB Diego. Einkorn 1 produced least number of seminal roots (4.90) in compost. Further, all of the ancient wheat genotypes produced more seminal roots in sand than the other medium (Table 1). Total root length was higher in emmer 1 (63.92 cm, $P < 0.001$) but not significantly different from emmer 2 and spelt Oberkulmeron GP where the shortest total root length was shown in einkorn 1 which was 46% shorter than emmer 1. Emmer 2 recorded the longest root length both in sand and compost medium. All genotypes, except bread wheat cv. JB Diego, recorded longer roots in sand than in compost. Averaged across species, total root length washigher in emmer followed by spelt then bread wheat and einkorn in both media. Emmer, spelt, bread wheat and einkorn produced 18.8, 20.7, 14.5 and 5.7% longer roots in sand than compost, respectively (Table 1).

Average convex hull of bread wheat and emmer was not significantly different but spelt and einkorn species were significantly lower than both bread wheat and emmer on GP. Average tip angle of seminal roots was significantly different between genotypes in the range of 16.7° to 35.8° ($P < 0.001$) Emmer 2 recorded largest tip angle of seminal roots while narrow tip angles of seminal roots were observed in spelt SB. On average, the largest tip angle was recorded in emmer (31.9°) followed by bread wheat (27.6°) then einkorn (23.2°) and spelt (18.7°). Emmer had 42, 27 and 14% wider tip angle than spelt, einkorn and bread wheat, respectively. Average emergence angle of seminal root was 21.6° to 35.1° between genotypes ($P < 0.01$) where einkorn 1 showed widest emergence angles while spelt Oberkulmer had the narrowest emergence angle which was 61% narrower than einkorn 1 (Fernando *et al.*, 2014).

The effect of genotype, medium and the interaction were highly significant ($P < 0.001$) for root volume, root diameter, root biomass, root to shoot ratio and SRL in the second experiment. The average root diameter of the genotypes was between 0.22 to 0.32 mm in compost and 0.27 to 0.33 mm in sand where Spelt Tauro recorded the highest average root diameter (0.32, $P < 0.001$) in compost and bread wheat cv. JB Diego showed the highest value in sand (0.33, $P < 0.001$). The thickest roots were observed in sand medium. Genotypes grown in sand produced the highest root volume. The highest value was recorded in emmer 2 for both sand and compost medium. Lowest root volume was observed in einkorn 3 for compost and einkorn 1 for sand. Overall, root volume of two week old seedlings was higher in emmer and spelt than bread wheat and einkorn in both growing media. R: S was significantly different between genotypes, the lowest R: S value was observed in einkorn 1 and 3 in compost and all einkorn cvs in sand. R: S was higher in sand than in compost. SRL was significantly different between genotypes and the highest value was recorded in einkorn species which had the thinnest roots, in both compost and sand.

Table 1: Number of seminal roots and total root length of the genotypes used in experiment 1 and 2

Genotype	Number of seminal roots			Total root length (cm)		
	Experiment 1	Experiment 2		Experiment 1	Experiment 2	
		Sand	Compost		Sand	Compost
JB Diego	3.75	4.60	5.50	49.03	161	180
Xi 19	4.19	5.50	5.70	49.93	244	214
Beaver	-	5.70	6.00	-	237	168
Forno	-	5.60	5.90	-	199	176
Gallant	-	6.20	5.70	-	257	188
Glasgow	-	5.40	5.70	-	223	196
Hobbit	-	5.60	5.40	-	205	182
Spelt Tauro	4.03	6.20	5.30	52.36	230	195
Spelt SB	3.95	5.80	5.30	53.04	277	213
Spelt Ober	4.36	6.10	5.30	60.69	272	210
Emmer 1	4.96	6.60	5.20	63.92	225	212
Emmer 2	5.27	7.40	7.30	60.20	321	231
Einkorn 1	3.76	5.50	4.90	34.46	229	180
Einkorn 2	4.40	6.30	5.40	42.07	222	158
Einkorn 3	4.53	5.60	5.20	44.73	216	136
SED						
GT (*df*)	0.23 (*151*)***	0.15 (*545*)***		3.05 (*151*)***	11.17 (*545*)***	
M (*df*)		0.06 (*545*)***			4.08 (*545*)***	
GT x M (*df*)		0.21 (*545*)***			15.79 (*545*)***	

In this study, large phenotypic variation of roots in response to various growth conditions were found among ancient wheat species together with bread wheat. Root systems in the first experiment, where roots grow vertically on the germination paper in nutrient media, was much smaller than those grown in sand and compost at two weeks. Root growth of the seedlings in sand was very strong for all species, suggesting that root growth was encouraged by limited supply of nutrient. On the other hand, foraging capacity of the roots is very high under resource limited conditions. Root length in < 0.5 mm diameter class was greater in sand suggesting that, all genotypes produced thinner roots, therefore increased surface area. Species variation for number of seminal roots, total root length, root volume, root biomass production and average diameter of the roots were very high among ancient wheat species. Emmer 2 had highest number of seminal roots, total root length and root volume, under any growth conditions at early seedling stage.

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