

## Seedling morphology of different wheat genotypes at early stages under hydrocultural conditions

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### SUMMARY

*Consuming “sprouted seeds” is one of the most important factors of a healthy diet. An experiment was conducted in the University of Debrecen, Research Centre of Nyíregyháza (Hungary) in 2014 to analyse some morphological traits of four winter wheat genotypes (Triticum aestivum L.) and one spelt (T. spelta) variety. Our results showed that the spelt wheat variety “Franckenkorn” could maintain higher root length throughout the experimental period. On average, both “Perbetei” and “Franckenkorn” varieties could maintain higher root number compared to the other varieties. The extensive breeding line “1401 HK” had the highest shoot length throughout the whole experiment, being significantly higher than the landrace variety “Perbetei” and both of the varieties “KG Bendegúz” and “KG Kunhalom”. It could be concluded that “KG Bendegúz” cultivar and “Perbetei” landrace seem to be the most suitable for aquaculture techniques. In addition, “1401 HK” breeding line can be the most suitable for the production of juice since the minimal required shoot length (12 cm) was achieved within the shortest period of time after sowing (9 days). This breeding line and “Franckenkorn” can also be suitable for production of “wheatgrass”, because it is consumed without roots. Further research is needed to evaluate nutritional values of these genotypes.*

**Keywords:** aquaculture; shoot and root parameters; *Triticum aestivum*; *Triticum spelta*

### INTRODUCTION

The consumption of “sprouted seeds” as a food is becoming increasingly important in a healthy diet. In addition to fresh consumption (“ready-to-eat” sprouts), they are also present in the market as a processed product (dried or toasted). The final product for consumption may be different depending on the method of production and the plant parts to be consumed (with or without seeds, roots or young leaves/cotyledons) (Benincasa et al., 2019); for example, the “wheatgrass” means the shoots of the 6–10-day old wheat seedlings (Pant et al., 2013).

The nutritional value of wheatgrass is given by the easily accessible essential trace elements (Kulkarni et al., 2007) and high antioxidant content, which reaches its peak on about 7<sup>th</sup>–8<sup>th</sup> day of growing (Kulkarni et al., 2006). Peptides and amino acids also increase its nutritional value. The way of production is often aquaculture, which is also suitable for the production of fodder (Benincasa et al., 2019; Ndaru et al., 2020).

The hydroponic cultivation conditions are also suitable for performing various laboratory tests. For example, hydroculture experiments can be used for the evaluation of stress tolerance capacity (Ashraf and Ashraf, 2016; Rizwan et al., 2016), and the colonization capacity of roots with bacteria (Morales et al., 1996). Studying morphological characters such as root system (Ayalew et al., 2015; Kabir et al., 2015) or developmental traits of different genotypes, including early growth rate, can also be evaluated in this method, which can provide the knowledge that can be useful to both breeders and growers.

Seedling vigor is one of the most important prerequisites for establishing successful stand of crop plants (Baloch et al., 2012), it affects the final yield (Rauf et al., 2007). The roots, known as ‘the hidden half’, play a main role in plant growth, they absorb

water and nutrients from the soil, and can store them properly (Kutman et al., 2012; Smith and De Smet, 2012; Gaju et al., 2014), and they support the plants against lodging (Smith and De Smet, 2012). In wheat, there are two types of roots: the seminal roots and the nodal roots (Kirby, 2002). During germination, 3–6 seminal roots, in addition to the radicle, emerge. In the field, the seminal root system can reach more than 1 m deep and support the plant throughout its life cycle (Sylvester-Bradley et al., 2008). The nodal (crown or adventitious) roots, unlike the seminal roots, don’t grow directly from the seed, but arise from the basal nodes of the stem and begin to extend when tillering starts (Kirby, 2002).

The root system of wheat is relatively complex, and because of the obstacles of selection for traditional root traits, architectural root properties that are expressed at early stages of crop development and determine the growth and functioning of the mature root system later in the season may be more suitable as selection criteria in crop improvement programs (Manschadi et al., 2006). These quantitative traits, called the root system architecture (RSA), including root number and length, tip and emergence angles, rooting width and depth among other traits, have been used to describe the root spatial configuration in soil (Pound et al., 2013; Atkinson et al., 2015).

However, measuring root traits in the field is very difficult since extraction of all intact roots from the soil is very time-consuming, expensive and requires much labor. Alternatively, many indoor methods have been applied, like using hydroponic culture (Ayalew et al., 2015; Kabir et al., 2015), gelfilled chambers (Manschadi et al., 2008; Christopher et al., 2013), sand-filled pots (Waines and Ehdai, 2007; Hamada et al., 2012), soil-filled pots (Cao et al., 2014), clear pots (Richard et al., 2015), a germination paper-based ‘pouch and wick’ system (Atkinson et al., 2015), a



paper-based ‘cigar roll’ system (Li et al., 2011; Bai et al., 2013) and soil-filled columns scanned by X-rays (Gregory et al., 2009). There is widespread evidence for genotypic variations in the root characteristics of many crop species (Ludlow and Muchow, 1990). In wheat, such traits include the depth of rooting, root elongation rate, root distribution at depth, xylem vessel diameter, and root to shoot dry matter ratio (Hoad et al. 2001; Manske and Vlek 2002; Manschadi et al., 2006).

The aim of our experiment was to study some morphological traits, namely root length, root number and shoot length at the early growing stage (5–10 days after emergence) in different wheat genotypes in hydroculture conditions. In the experiment 2 wheat varieties, a breeding line, a landrace and a spelt wheat variety were included assuming their good nutritional

values (Abdel-Aal and Rabalski, 2008; Dotlačil et al., 2010).

## MATERIALS AND METHODS

The experiment was conducted in the University of Debrecen, Research Centre of Nyíregyháza (Hungary) during the year 2014.

### Plant material

Four winter wheat genotypes (*Triticum aestivum* L.), i.e. “Perbetei” (landrace), “1401 HK” (breeding line), “KG Bendegúz”, “KG Kunhalom” (varieties) and a spelt wheat variety (*Triticum spelta*) “Franckenkorn” were tested under hydroponic culture conditions. Table 1 lists the main characteristics of the materials used in the experiment.

Table 1. The main characteristics of selected wheat genotypes in the experiment

Variety	“Perbetei”	“1401 HK”	“KG Bendegúz”	“KG	“Franckenkorn”
Maturity	late	late	mid-early	mid-late	mid-late
1000-seed Weight (g)	41	38	47	42	80
Stem high (cm)	high (125–135)	high (105–120)	Short (90–100)	high (110–120)	145–165
Biomass weight	high	high	mid	mid	high
Yield t ha <sup>-1</sup>	3–4	3–6	7–9	7–9	2–5
Resistance to lodging	weak	weak	good	good	good

### Experimental setting conditions

The seeds were soaked in 10 Liters of aerated tap water for 24 h in darkness at 22±2 °C (Policarpo et al., 2007), using an air pump. 10-day-old sprouted winter wheat cultures were produced in high purity without any microbial infection using 8 g l<sup>-1</sup> zeolite (sodium and calcium aluminium silicate in crystalline form) in tap water instead of disinfectants (Wang et al., 2012). After the swelling, the surplus water was removed. The fully imbibed grains were placed in transparent polystyrene boxes (d=5.5 cm) (Figure 1), with different numbers of seeds depending on the thousand-seed-weight of genotypes („KG Bendegúz”: 105 pcs; „KG Kunhalom”: 130 pcs, „Franckenkorn”: 100 pcs, „1401 HK”: 130 pcs, „Perbetei”: 120 pcs) in order to create only one layer of seeds. After that, the seeds were kept in darkness at 22 °C for 48 h before they were cultured in plant growth chamber (Angelantoni CHL1500) under 12 h illumination, at 70% relative humidity and at 20 °C for 10 days, because 20 °C was found to be the best to prevent infections and to obtain shoots in high quality (Dziki et al., 2015). Water supply was made twice a day by spraying of distilled water to avoid excess water.

Figure 1. General view of wheat hydroculture experiment



### Measurements and statistical analyses

The experiment was initiated with 15 boxes per genotype, and three boxes were used for observation at four sampling times. Growth parameters (length of shoots, length and number of roots) were observed on 5<sup>th</sup>, 8<sup>th</sup>, 9<sup>th</sup> and 10<sup>th</sup> days: 10 seedlings per box were measured in three repetitions.

Data were analysed by ANOVA followed by Tukey-test ( $P \leq 0.05$ ) using SPSS 26.0 software.

## RESULTS AND DISCUSSION

### Root length

Excessive growth of root system in hydroponic wheat culture is undesirable because fibrous roots

require thorough processing before consumption. In addition, crowded rhizosphere creates circumstances which can enhance the growth of harmful microorganisms. Furthermore, the development of an extensive root system consumes a lot of energy and oxygen (Wetherell, 1988). Production of wheat seedlings for pressed juice also required shoot length up to 15 cm with a minimum of the root length (Koneva et al., 2018). Root system architecture of wheat shows large genetic variation among different ploidy levels, between domesticated and wild forms, and within each of the diploids, tetraploids and hexaploids (Manschadi et al., 2008; Ayalew et al., 2015). In our study we also observed significant differences in root development of wheat genotypes (Table 2).

Table 2. Root length (mm) of 10-day-old wheat seedlings

Day	“Perbetei”	“1401 HK”	“KG Bendegúz”	“KG Kunhalom”	“Franckenkorn”
5	28.1 <sup>c3</sup>	37.4 <sup>c2</sup>	29.4 <sup>c3</sup>	30.9 <sup>c3</sup>	43.4 <sup>d1</sup>
8	56.8 <sup>b3</sup>	69.2 <sup>b2</sup>	50.1 <sup>b3</sup>	84.3 <sup>b1</sup>	96.1 <sup>c1</sup>
9	72.9 <sup>a3</sup>	105.0 <sup>a12</sup>	68.4 <sup>a3</sup>	93.8 <sup>ab2</sup>	110.7 <sup>b1</sup>
10	60.9 <sup>ab3</sup>	94.5 <sup>a2</sup>	48.6 <sup>b3</sup>	101.6 <sup>a2</sup>	124.0 <sup>a1</sup>

- Same letters indicate no significant difference at .05 level among days within certain genotype.
- Same numbers indicate no significant difference at .05 level among genotypes within certain day.

The spelt wheat variety “Franckenkorn” could maintain higher root length throughout the experimental period. The extensive breeding line “1401 HK” had significantly higher root length than the landrace variety “Perbetei” during the whole experiment. Both varieties “KG Bendegúz” and “KG Kunhalom” had a very similar root length on day 5 compared to “Perbetei”, however, the roots of “KG Kunhalom” were significantly longer than both “KG Bendegúz” and “Perbetei” thereafter.

The root length of “Perbetei”, “1401 HK” and “KG Bendegúz” reached their maximum on day 9, whereas the rest showed the highest values of this trait at the last day of measurements (day 10). In their experiment, Wojciechowski et al. (2009) measured the root length of 7 different wheat Near-isogenic lines (NILs) (cv. Mercia) in gel chamber every second day until 12 days after germination. They reported that the different NILs had different root length through days, especially at day 10, where Rht-D1b line showed significantly less root length compared to Rht12 line. All studied lines showed continuously increasing root length over time

(Wojciechowski et al., 2009). Baloch et al. (2012) also reported that the seedlings of 16 different spring wheat genotypes had different root length after 10 days of applying distilled water, with the difference being significant for certain genotypes. Moreover, Wojciechowski et al. (2009) observed a relationship between root length and grain mass; i.e. longer roots developed from bigger seeds. In our experiment, correlation between root length and grain mass could not be detected (data not shown), probably because of the small differences in the 1000-seed-weight of the majority of genotypes (38–48 g). However, the longest roots developed from “Franckenkorn” seedlings, which have the highest grain mass (75–95 g).

### Root number

On average, both “Perbetei” and “Franckenkorn” varieties could maintain higher root number compared to the other varieties, followed by “1401 HK”, “KG Bendegúz” and “KG Kunhalom”, respectively (Table 3).

Table 3. Root number of 10-day-old wheat seedlings

Day	“Perbetei”	“1401 HK”	“KG Bendegúz”	“KG Kunhalom”	“Franckenkorn”
5	3.0 <sup>b2</sup>	3.0 <sup>b2</sup>	3.0 <sup>b2</sup>	3.2 <sup>c2</sup>	3.7 <sup>b1</sup>
8	4.7 <sup>a1</sup>	4.5 <sup>a12</sup>	4.4 <sup>a12</sup>	4.0 <sup>b2</sup>	4.4 <sup>a12</sup>
9	4.8 <sup>a1</sup>	4.5 <sup>a1</sup>	4.3 <sup>a12</sup>	3.6 <sup>bc2</sup>	4.8 <sup>a1</sup>
10	5.0 <sup>a1</sup>	4.8 <sup>a1</sup>	4.5 <sup>a1</sup>	4.6 <sup>a1</sup>	4.5 <sup>a1</sup>

- Same letters indicate no significant difference at .05 level among days within certain genotype.
- Same numbers indicate no significant difference at .05 level among genotypes within certain day.

From a daily point of view, all varieties, except for “Franckenkorn”, reached the highest root number by the end of the experiment and there were no significant differences in the root number between genotypes on 10<sup>th</sup> day. Previously, the number of seminal roots was measured for 26 wheat cultivars representing some of the most widely grown varieties in Australia and 3 CIMMYT genotypes (Manschadi et al., 2008). The authors reported that the number of seminal roots varied between wheat genotypes. While Yitpi genotype developed consistently five seminal root axes, genotypes Petrie, EGA Wedgetail, and Babax were the genotypes with the lowest number of seminal roots. Moreover, cluster analysis of the seminal root number indicated that the wheat genotypes formed 4 discrete groups. Groups 1 and 2 consisted of eight genotypes each, while Groups 3 and 4 included nine and four cultivars, respectively. Wheat cultivars in Groups 1 and 2 exhibited a low number of seminal roots, whereas cultivars in Groups 3 and 4 comprised genotypes expressing a high number of seminal roots. Xie et al. (2017) made a cross between a bread wheat cultivar ‘Forno’ and spelt landrace ‘Oberkulmer’, and the subsequent F5 recombinant inbred lines (RILs) mapping population consisting of 226 lines was used in

the study. The authors reported that there was a large variation in the g root system architecture (RSA), including root number, among the different RILs derived from Forno x Oberkulmer.

### Shoot length

The extensive variety “1401 HK” had the highest shoot length throughout the whole experiment, being significantly higher than the landrace variety “Perbetei” and both of the varieties “KG Bendegúz” and “KG Kunhalom”, with an average shoot length of 133.8 mm at the end of experiment. “KG Bendegúz” had the lowest shoot length with an average of 103.4 mm on 10<sup>th</sup> day (Table 4). In their experiment, Akbarimoghaddam et al. (2011) measured the shoot length of 6 wheat genotypes on 8<sup>th</sup> day after germination; they found out that the different genotypes showed different shoot length, ranging from 15.61 cm to 22.11 cm; however, these differences were insignificant (Akbarimoghaddam et al., 2011). The same conclusion was also reported (Baloch et al., 2012) with the differences being significant among the studied genotypes, and the shoot length of the 10-day-old seedlings ranging between 12.82 and 17.11 cm.

Table 4. Shoot length (mm) of 10-day-old wheat seedlings

Day	“Perbetei”	“1401 HK”	“KG Bendegúz”	“KG Kunhalom”	“Franckenkorn”
5	20.6 <sup>c2</sup>	29.9 <sup>c1</sup>	20.0 <sup>c2</sup>	18.4 <sup>d2</sup>	27.0 <sup>d1</sup>
8	80.3 <sup>b3</sup>	96.6 <sup>b1</sup>	79.4 <sup>b3</sup>	90.8 <sup>c12</sup>	87.9 <sup>c2</sup>
9	107.3 <sup>a2</sup>	129.6 <sup>a1</sup>	107.0 <sup>a2</sup>	104.1 <sup>b2</sup>	110.0 <sup>b2</sup>
10	108.7 <sup>a23</sup>	133.8 <sup>a1</sup>	103.4 <sup>a3</sup>	121.6 <sup>a12</sup>	128.0 <sup>a1</sup>

- Same letters indicate no significant difference at .05 level among days within certain genotype.
- Same numbers indicate no significant difference at .05 level among genotypes within certain day.

## CONCLUSIONS

It is very important to choose those winter wheat genotypes for hydroculture growing whose short roots or small root systems are genetically encoded, in addition to the development of an adequate shoot systems, since any treatment, which inhibited root growth, might reduce shoot growth (Wetherell, 1988). In this regard, “KG Bendegúz” cultivar and “Perbetei” landrace seemed to be the most suitable for aquaculture techniques. On the other hand, “1401 HK” breeding line can be the most suitable for the production of juice since the minimal required shoot length (12 cm) was achieved within the shortest

period of time after sowing (9 days). This breeding line and “Franckenkorn” can also be suitable for production of “wheatgrass”, because it is consumed without roots. Moreover, “Franckenkorn” showed high antioxidant capacity in Abdel-Aal and Rabalski studies (2008). Further research is needed to evaluate nutritional values of these genotypes.

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