Developmental aspects of barley drought tolerance

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POLAPGEN-BD: Biotechnological tools for breeding cereals with increased resistance to drought

Task 2. Genetic maps and the location of QTLs associated with barley resistance to the water deficiency

Project carried out under Innovative Economy Programme 2007-2013, Action 1.3, Subaction 1.3.1. within the subject „Biological progress in agriculture and environment protection“
THE AIM OF THE PROJECT

1. The general aim of the project was to enhance competitiveness of breeding companies producing cereal varieties and development of biotechnological services in Poland. This is the action that will lead to a biological progress.

2. Environmental protection.

3. Creating prototype biotechnological services, which could be provided to breeders and agricultural producers.
A system approach is achieved by adopting a model of tolerance to drought stress containing ecophysiological, morphological, anatomical, metabolic, proteomic, and molecular levels considered in the context of genetics.
THE AIM OF THE STUDY

Mapping of quantitative trait loci (QTL) affecting yield and its components in European/Syrian spring barley (*Hordeum vulgare* L.) cross combinations under optimal and water shortage conditions.
PLANT MATERIAL

Three populations consisted of 100 RILs ($F_8$) each, generated via SSD technique in POLAPGEN project:

Lubuski x Cam/B1/Cl, Maresi x Cam/B1/Cl, Georgie x Harmal.

Maresi (German, semidwarf malting variety)
Fig. Steps in the procedure of obtaining RIL combined with *in vitro* culture: a) spikes and immature kernels from which the embryos were dissected; b) embryos placed on B5 medium; c) embryos developed after 4 days of culture; d) plantlets after 8 days of *in vitro* culture; e) plants transplanted in pots; f) spike plants after flowering stage.
GREENHOUSE EXPERIMENT

1. The soil was taken from the Grabów Experimental Station (E 21º 39’, N 51º 21’);
2. The soil texture and properties were characterized by the Institute of Agrophysics, Lublin;
3. Fertilization according to the Institute of Soil Science and Plant Cultivation, Puławy;
4. Water retention curve pF;

The pF value is defined as a logarithm of the pressure p (expressed in cm of water head) necessary for water removal from soil capillaries.

- Levels of soil moisture:
  1. pF 2.2-3.0 – optimal conditions
  2. pF 3.2-3.6 – water deficit
Water shortage was imposed at the three leaf stage (phase 13 in the BBCH scale) and maintained for 10 days.
Water shortage was imposed at the flag leaf stage (phase 37 in the BBCH scale) and maintained for 14 days.
MEASUREMENTS OF SOIL MOISTURE

<table>
<thead>
<tr>
<th>pF</th>
<th>3.2</th>
<th>3.4</th>
<th>3.6</th>
<th>3.8</th>
<th>4.2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Humidity %w/w</td>
<td>8.0</td>
<td>6.75</td>
<td>5.7</td>
<td>4.75</td>
<td>3.6</td>
</tr>
</tbody>
</table>
PHENOTYPING

Traits

1. Grain weight per main spike (g)
2. Grain weight per lateral spike (g)
3. Grain weight per plant (g)
4. 1000-grain weight (g)
5. Length of main spike (cm)
6. Length of lateral spike (cm)
7. Number of grains per main spike
8. Number of grains per lateral spike
9. Number of spikelets per main spike
10. Number of spikelets per lateral spike
11. Fertility of main spike
12. Fertility of lateral spike
13. Length of main stem (cm)
14. Number of productive tillers per plant
15. Number of tillers per plant
16. Productivity of tillers
17. Straw weight per plant (g)
18. Heading stage (days)

Plants secured for biometric observations

Average of 10 plants
A consensus linkage map of barley constructed using three RIL mapping populations and used for QTL analysis.

Mikołajczak K. et al. Quantitative trait loci for yield and yield-related traits in spring barley populations derived from crosses between European and Syrian cultivars. under review
EXPERIMENTS CARRIED OUT IN THE GREENHOUSE

Variation of the spike morphology
EXPERIMENTS CARRIED OUT IN THE FIELD
Transnational Access Application Form

1. Details
Name(s) of submitting scientist(s): Anetta Kuczyńska, Paweł Krajewski
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2. Requested access to:
[Please provide the short name of the installation(s) to which you request access]
IPK APPP

3. Title of the project
[max. 200 characters]
Developmental aspects of barley drought tolerance

4. Project acronym
[max. 20 characters]
POLAPGEN-BIP
Setup experiment 1436PK
Phenotyping of 95 RIL barley lines plus 4 control varieties
8x replicates (2xcarrier per line; 4-pot config per carrier)
2x treatment conditions (control normal watering, stress conditions no watering)

Pre-cultivation phase: 4th of August – 3rd of September 2014
04.08.2014 Sowing of seeds in 96-well tray, soil mixture (Substrate 2 + Sand, 4:1)
24x seeds of each genotype distributed over 24x trays (1xreplicate/tray)
Greenhouse climate conditions:
16°C day/14°C night temperature
>65% relative humidity
Long day conditions (6.00-22.00) additional light
Continuous watering of trays as required (manually done by gardeners)

Transplanting of the plants in individual pots
20.08.2014 Transplanting of single plants in 3.5L pots for LemnaTec system
Soil mixture (Substrate 2 + Sand, 4:1)
Greenhouse climate conditions:
16°C day/14°C night temperature
>65% relative humidity
Long day conditions (6.00-22.00) additional light
Continuous watering of pots as required (manually done by gardeners)
Phenotyping: 3rd of September – 8th October 2014

01.09.2014  Transfer pots from greenhouse to LemnaTec phenotyping facilities
28das (days after sowing)

Greenhouse climate conditions:
20°C day/16°C night temperature
>65% relative humidity
Long day conditions (6.00-22.00) additional light
Automatically watering based on watering regime (target weight)

Start watering 03.09.2014
Start imaging 04.09.2014

08.09.2014  Start stress (35das)
No/low watering to 20% field capacity
Normal watering 70% FC (control)

22.09.2014  Start recovery phase
Adding 300ml absolute watering to each pot

23.09.2014  additional target watering to reach 90% FC

25.09.2014  Fertilization, Combo Hakaphos blau (200ml absolute volume per plant)

01.10.2014  Fertilization, Combo Hakaphos blau (200ml absolute volume per plant)

Rotation of the carriers: each day for one lane, every 3rd day in between the lane for 11 positions
All lines (RILs, varieties) (left) and varieties only (right)
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All lines (RILs, varieties) (left) and varieties only (right)
Post-cultivation: from 7th of October 2014
07.10.2015 Finish phenotyping, transfer of a selection of plants (around 800 plants) for post-cultivation, harvest

Fig. Biometric analysis of spikes:
A – main spikes collected from a pot;
B – measurement of spikelets number per spike;
C – measurement of spike length
Principal component biplot for traits observed after harvest; green – lines non-stressed, red – lines stressed
Thank you for your attention