MINUTES

Study Group Genetics & Breeding/IIRB, AG Beta Rüben/GPZ

21 September 2017, Institute of Sugar Beet Research, Göttingen (D)
22 September 2017, KWS SAAT SE, Einbeck (D)

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21st September 2017, IfZ

Bernward Märländer and Anne-Katrin Mahlein welcomed the group to Göttingen and presented the IfZ to the participants. Stephanie Kluth presented the IIRB.

Andreas Loock as chair of the IIRB Genetics and Breeding study group welcomed the members of the AG Beta Rüben of GPZ, the IIRB study group members as well as the external guests.

Bernd Weisshaar (Univ. Bielefeld): Beet genome sequence and steps to its improvement

From 2008 to 2011 the GABI BeetSeq project was set up with the aim of establishing a sugar beet reference genome sequence. It was based on the GABI Beet project (1999-2004) and the GABI Beet physical map (BPM) 2004-2007. In these projects the basics for further development of molecular markers and tools have been developed.

Gina Capistrano-Goßmann (CAU Kiel): Identification of Rz2 by direct mapping in a crop wild relative in situ population

Beet necrotic yellow vein virus (BNYVV) is transmitted by soilborne *Polymyxa betae* and causes yield reductions of up to 80% worldwide. More than 50% of the European production area is affected. Partial resistance is conferred by 'Holly' (*Rz1*) or WB 42 (*Rz2*) resistance types. *Rz1* resistance breakdown has been observed in the USA as well as in Europe. Climate warming might influence the increase of resistance-breaking strains.

Frequently used segregating populations in breeding are germplasm collections, elite breeding materials, and synthetic populations. This research referred to a segregating wild beet population in Kalundborg with permanent outcrossing, a low LD around the *Rz2* locus and high resolution mapping of *Rz2*. It can be regarded as a model for gene mapping in in situ populations.

Material was developed with 200 testcross families, the *Rz2* locus was mapped by sequencing and the resistance of the 200 testcross families was analysed. *Rz2* segregation was observed in the Kalundborg population which was not under Rhizomania selection pressure. In the process of mapping-by-sequencing, the genomic region containing *Rz2* was identified as well as 19 new markers associated to rhizomania resistance. MBS showed two pools of homozygous resistant and susceptible wild beet plants. In susceptible plants, a functional allele of *Rz2* was absent. The use of crop wild relative *in situ* populations can be regarded as a new strategy for gene identification.

Nadine Dally (CAU Kiel): Unraveling the genetic mechanisms of bolting time control in sugar beet

Cultivated beets are biennial crops, whereas annual, perennial and biennial plants are found in wild beets. Breeding winter beets is a challenge, controlled bolting would impact the vegetative growth for crop production and the reproductive growth for seed production and would require expertise about bolting and flowering time regulation. The pseudoresponse regulator BTC1 and the zinc finger transcription factor BvBBX19 were identified as two major bolting time regulators in beet by positional cloning approaches. Both genes act upstream of the flowering time genes BvFT1 and BvFT2. Mutations in either gene, BTC1 or BvBBX19, turn an annual into a biennial plant as they regulate the two FT homologs BvFT1 and BvFT2 to induce bolting, so mutant hybrids may bolt delayed or even fail to bolt after winter.

Max Teutsch (CAU Kiel): Hairy roots as an efficient test system for genome editing in sugar beet

CRISPR/CAS is functional in sugar beet hairy roots. Hairy roots provide a reliable, cost and time efficient system. PCR-restriction assays allow fast and reliable mutant detection. There is great potential for the development of HDR technology before *A. tumefaciens* transformations.

During coffee break, the group was shown IfZ facilities.

Shahid Siddique (Univ. Bonn): Plant basal resistance against parasitic nematodes

The sugar beet cyst nematode *Heterodera schachtii* is distributed worldwide in sugar-beet producing areas and can infect more than 200 species. Plant immune responses were analysed in *Arabidopsis* by checking the up/down regulation of immune genes and distinguish wound responses from nematode recognition. 'NemaWater' was produced by incubating J2 for 24 hours in water on a shaker, centrifuging and using the supernatant for analyses of secreted nematode compounds. NemaWater was found to activate ROS burst and inhibit growth in *Arabidopsis*, it elicited immune responses in host plants. After pre-treatment of plants with NemaWater to induce resistance to nematodes, the number of female nematodes in treated plants decreased strongly compared to controls. Results suggest that NemaWater contains potential elicitors recognized by a surface-localized receptor in plants leading to the activation of PTI-like responses. A knock out mutant for NILR1 was found to be more susceptible to nematodes and to be impaired in PTI responses.

J. Roeb (JKI, Braunschweig): Resistance and tolerance of sugar beet genotypes against the beet cyst nematode (Heterodera schachtii)

In field experiments on 3 resp. 4 locations in 2015 and 2016, 12 sugar beet genotypes (4 susceptible, 3 resistant, 5 tolerant) were tested for their resistance and tolerance towards *H. schachtii*. Nematodes were sampled at sowing (Pi) and harvest (Pf) in two soil depths. Cysts were extracted by the centrifugal flotation method, and cysts, eggs and juveniles were counted per 500 ml soil. Sugar beet yield and quality were determined. Variable degrees of partial resistance were observed, tolerant genotypes could be classified as partially resistant. The corrected sugar yield did not necessarily correlate with nematode tolerance. Field trials results were not necessarily reproducible in greenhouse experiments: Whereas the results of replication rates Pf/Pi in field trials and several greenhouse trials differed, the same order in the degree of tolerance could be observed in the number of eggs and juveniles/cyst.

M. Varrelmann (IfZ): Perspectives for the development of resistance biotests against different pathogens

Giving the example of rhizomania resistance biotests, biotests for the determination of storage rot susceptibility, and resistance biotests of sugar beet against *Helicobasidium purpureum*, the talk evaluated the perspectives for the development of resistance biotests. Molecular methods can be used in BNYVV resistance tests in the greenhouse. BNYVV fluorescence labeling allows evaluating the virus replication and movement by a UV lamp. Sugar beet genotypes differ in their level of susceptibility against storage rots. Biotest to determine storage rot susceptibility with whole field grown beets are time consuming and costly, but can be more efficient with greenhouse grown beets and an infection assay with *Fusarium graminearum*. Biotests could be further optimised by inoculating species mixtures, also on field grown beets. Analyses of transcriptomic response could identify possible resistance factors. The causes for violet root rot (*H. purpureum)* and its epidemiology as well as control measures are largely unknown. Resistance in sugar beet has not yet been shown; the development of resistance biotests to identify and evaluate factors of predisposition, susceptible hosts and their effect on spread and disease severity is required. Inoculation success in the field strongly depends on the year respectively environment, ranging from no infections to very strong infections which do not allow discrimination. Both greenhouse and field biotests have their advantages and limitations and remain challenging.

A. Wauters: Presence and identification of 'blue-grey' or 'silver' beet symptoms

New disease symptoms in sugar beet were observed since 2012 in Belgium (blue-grey matt color of leaves with a silvering of leaf surface in mature leaves and a cracking leaf tissue with increased visibility of leaf veins, browning of vascular bundle rings in the root tip). Symptoms were observed in 20 seed lots in 2016, and in 54 seed lots in 2017. Similar symptoms were reported from other European countries. Sugar beets from all breeding companies are concerned. *Curtobacterium flaccumfaciens* pv. *betae* (syn. *Corynebacterium betae*) was determined as the causal agent of 'silvering disease of beet'. An identification based on biomolecular techniques confirmed the presence of *C. flaccumfaciens pv. betae.* Further research is required to identify host range, pathogen survival, disease development etc.

C. Hoffmann (IfZ): Causes for genotypic variability in storage losses

Variety differences occur in the susceptibility of sugar beet to damage and root tip breakage. Less root tip breakage results in a lower infestation with moulds and rots during storage, a lower accumulation of invert sugar and lower sugar losses. Damage susceptibility is due to dry matter composition, affected by variety and environment. In most cases the damage susceptibility and thus marc content can explain storage losses, however, in some cases there seems to be an additional reason for losses.

A.-K. Mahlein (IfZ): Phenotyping the response of sugar beet to biotic and abiotic stress by optical sensors

Host-pathogen interactions differ in time and space, resulting in specific spectral signatures. Sensor technologies that measure these spectral signatures can therefore be used for a reproducible, objective and automatic detection of plant diseases, as well in precision agriculture where it can assist in detecting site-specific diseases as is plant phenotyping. Here genotypes can be evaluated in different environments to be able to select the relevant genotypes. Leaf structural changes that are induced by plant infections can be measured by sensor-based diagnosis methods.

22 September 2017, KWS SAAT SE

The meeting continued the next day at KWS SAAT SE.

KWS is among the 5 top seed companies worldwide. 17% of its net sales are spent on research and development. 60% of activities are focused on corn, 32% on sugar beet.

Maik Gertz: Development of CONVISO SMART sugar beets at KWS

Breeding for specific traits must focus on fast integration of the target traits. The development of ALS resistant varieties was presented. The trait “ALS resistance” was introduced in the germplasm by marker assisted backcrossing approaches. To speed up the breeding progress big populations of different backcross generations were screened foreground for ALS resistance and background for the elite line genome. By using this strategy the development of CONVISO varieties was sped up significantly.

Jens Lein: Molecular breeding for new varieties

All KWS sugar beet breeding programs are assisted by molecular markers. To establish efficient tools, adapted technologies have to be developed in collaboration within scientific networks as GABI. Each program needs specific support and a customized composition of marker applications. To use these molecular tools, molecular breeding needs a comprehensive understanding of the breeding material. Markers have been developed and are used for monogenic, oligo or polygenic inherited traits. The pro-active development of genomics resources allow a just-in-time marker development and transfer in breeding routine. All marker approaches as Marker Assisted Selection, QTL-based and Predictive Breeding significantly increase the gain in selection and will help to ensure the high competitiveness of the sugar beet crop.

Enrico B. Scheuermann: Georeferenced data in future breeding applications

Spatial applications become more and more important. A large amount of data needs to be made available in a fast way to all who require it in the breeding process. The spatial and temporal documentation is a challenge. After field measurements and the determination of the field boundaries, data can be imported and trials created with the help of GIS. Trial export is done from GIS to GNSS. The central platform is called 'Field explorer', its usability is important. The spatial layout of trials is interfaced with breeding data.

Field visit

During the field visit the group was demonstrated the functioning of the KWS quadrocopter and was shown ALS tolerant sugar beet demonstration plots.