

Our Latest Research Results - March 2011

In the Eye of the Beholder: The Effect of Rater Variability and Different Rating Scales on QTL Mapping

Authors: J.A. Poland, R. Nelson

Submitted to: Phytopathology

Breeding for disease resistance has been a long-standing and important objective for plant breeders. Reflecting this, many studies have been conducted to find molecular markers that are linked to genes conferring disease resistance in order to assist breeding efforts through marker assisted selection. These previous studies have been almost exclusively conducted using visual assessment of disease severity on the population of interest. Visual assessment of disease severity raises questions over the variability of ratings among different raters and how this affects the results and interpretations of these studies. In the present study, 22 individuals rated the same maize population for resistance to northern leaf blight, an important disease throughout the world. We then examined the impact of rater variability and different rating scales on the results of genetically mapping disease resistance genes. We found that despite considerable variability among raters, the genetic location of identified disease resistance genes was largely consistent. However, the estimated strength of that resistance was variable among raters. We concluded that differences between raters are real and may affect results in some cases.

Contact Jesse Poland, telephone 785-532-2709, email jesse.poland@ars.usda.gov

Genome-Wide Nested Association Mapping of Quantitative Resistance to Northern Leaf Blight in Maize

Authors: J.A. Poland, P. Bradbury, E.S. Buckler IV, R. Nelson

Submitted to: Proceedings of the National Academy of Sciences

There are two generally recognized forms of disease resistance in plants; quantitative and qualitative. Qualitative resistance is generally conditioned by a single gene of large effect. This type of resistance is very effective and useful in breeding resistant cultivars, however, cultivars developed with a single resistance gene often become susceptible due to mutations in the pathogen which render the resistance gene ineffective. On the other hand, quantitative resistance is generally

conditioned by multiple genes of small effect that can be combined to make resistant cultivars that are more robust to pathogen evolution. The mechanisms of quantitative resistance, however, are not well understood. In an effort to better understand quantitative disease resistance, we evaluated and analyzed a large maize population consisting of 5,000 inbred lines for resistance to northern leaf blight (NLB), an economically important disease of maize throughout the world. We found 30 genomic locations that were associated with resistance to NLB and explained most of the differences in resistance. We then used a technique termed nested-association mapping to identify genes associated with resistance and found multiple candidate genes for NLB resistance. These candidate genes were consistent with hypothesized classes of genes for quantitative resistance. Taken together, these results greatly expand the current understanding of quantitative disease resistance in plants.

Contact Jesse Poland, telephone 785-532-2709, email jesse.poland@ars.usda.gov

A Robust, Simple Genotyping-by-Sequencing (GBS) Approach for High Diversity Species

Authors: R.J. Elshire, J.C. Glaubitz, Q. Sun, J.A. Poland, K. Kawamoto, E.S. Buckler IV, S.E. Mitchell

Submitted to: PLoS One

Rapid developments in DNA sequencing technology are quickly reducing the cost of sequencing in many species. To utilize this new technology in diverse crop species with large genomes, we developed a simple procedure for making multiplex libraries for DNA sequencing. By multiplexing many samples the sequencing cost can be reduced to the point where many samples can be evaluated for genetics and breeding. We used a DNA restriction enzyme to target a limited portion of the genome in maize and barley for sequencing. Using DNA barcoded adapters, unique to each sample we were able to sequence 48 samples in single run. From this data, we identified 200,000 molecular markers in maize and 25,000 molecular markers in barley at a relatively low cost per sample. Using a simple protocol as described here, this genotyping-by-sequencing method can rapidly produce hundreds of thousands of molecular markers in many diverse species, enabling new genetics studies in under-researched crops, novel germplasm or thousands of samples in a plant breeding program.

Contact Jesse Poland, telephone 785-532-2709, email jesse.poland@ars.usda.gov

Resistance to Tan Spot and Insensitivity to Ptr ToxA in Wheat

Authors: A.J. Noriel, X. Sun, W. Bockus, G. Bai

Submitted to: Crop Science

Tan spot is a wheat leaf disease caused by the fungus *Pyrenophora tritici-repentis*. Tan spot is an important disease that can significantly reduce wheat yields worldwide. Host-selective toxins produced by the fungus are responsible for leaf damage. Growing resistant cultivars is an effective approach to reduce the losses caused by the disease. To identify resistance genes in common wheat, 380 wheat accessions from different geographical origins were evaluated for resistance to *P. tritici-repentis* race 1, the predominant race in the Great Plains of U.S. and western Canada, and for insensitivity to Ptr ToxA, a toxin produced by race 1. About 60% of accessions tested were resistant and only 24% were as susceptible as the susceptible check. A total of 230 accessions showed insensitivity to Ptr ToxA, but only 158 of them also showed resistance to race 1. The results suggest that insensitivity to Ptr ToxA is not the only factor for resistance to race 1, other factors such as Ptr ToxC might also contribute to tan spot damage in some accessions. The tan spot resistant accessions identified in this study should be useful sources for developing new tan spot resistant cultivars.

Contact Guihua Bai, telephone 785-532-1124, email guihua.bai@ars.usda.gov

Cysteine Digestive Peptidases Function as Post-Glutamine Cleaving Enzymes in Tenebrionid Stored Product Pests

Authors: I.A. Goptar, T.A. Semashko, S. A. Danilenko, E.N. Lysogorskaya, E.S. Oksenoit, D.P. Zhuzhikov, M.A. Belozersky, Y.E. Dunaevsky, B.S. Oppert, I.Y. Filippova, E.N. Elpidina

Submitted to: Biochimie

To be successful, stored product pests need to have enzymes capable of efficiently digesting their main dietary proteins, which have many glutamine and proline amino acids. We describe for the first time enzymes, glutamine-specific peptidases, in the gut of yellow mealworm larvae that are capable of digesting proteins at glutamine residues. Our characterization of these peptidases indicates that they are previously studied enzymes belonging to the class of cysteine peptidases, hypothesized to have evolved in storage pests to protect the insect from serine protease inhibitors found in cereals. We now propose that storage pests also have retained cysteine peptidases to efficiently digest cereal proteins. These results may be exploited to develop new control products for storage pests.

Contact Brenda Oppert, telephone 785-776-2780, email brenda.oppert@ars.usda.gov

Study of Hard Waxy Wheat Flours Differing in Pasting Properties

Authors: P.S. Garimella, Y.-C. Shi, L. Guan, J.D.

Wilson, R.A. Graybosch

Submitted to: Journal of Cereal Science

Starch in endosperm of normal wheat consists of ~ 25% amylose (mostly linear) and ~ 75% amylopectin (highly branched), whereas starch in waxy wheat, a valuable specialty wheat, endosperm is comprised of essentially all amylopectin. This waxy trait in wheat flour is responsible for differences in how starch melts, or pastes in excess water. This study was undertaken to determine the factors governing the pasting properties of waxy wheat flours and compare their pasting properties to those of normal and partial waxy wheat flours. Starches isolated from six hard waxy wheat flours had similar pasting properties, yet their corresponding normal and partial waxy flours had very different pasting properties. The differences in pasting properties were narrowed once endogenous α -amylase (a starch degrading enzyme) activity in the waxy wheat flours was inhibited by silver nitrate. Upon the action of protease, the extent of protein digestibility determined the viscosity profile in waxy wheat flours. Waxy wheat starch granules swelled more extensively and were more prone to α -amylase degradation than normal wheat starch. The protein matrix of the flour also affected the swelling of the starch granules which in turn also caused variations in pasting properties of waxy wheat flours.

Contact Jeff Wilson, telephone 785-776-2763, email jeff.d.wilson@ars.usda.gov

USDA-ARS Center for Grain and Animal Health Research

1515 College Avenue
Manhattan, KS 66502

800-627-0388
ars.usda.gov/npa/cgahr

