Using Genomics to Improve Drought Tolerance in Corn Hybrids

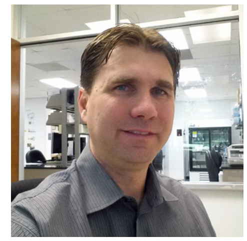
Syngenta uses the HiSeq® 2500 System to understand the mode of action behind their new Artesian hybrid corn trait.

Introduction

Seed corn companies have used traditional plant breeding for decades to select for drought tolerance in corn. Marker-assisted selection has been one of the genetic tools they've employed, enabling the identification of genes in corn that appear to perform better under water stress. It's an indirect process where a trait of interest is selected, not based on the trait itself, but on a marker linked to it[1]. This approach can produce a reasonably good product, but leaves scientists with no information about why it performs better than a traditional hybrid.

Water stress in corn is a complex process, affecting different mechanisms at different sites within the plant. Not surprisingly, several genes confer drought tolerance in corn. Syngenta is using the Illumina HiSeq 2500 System to understand the biology behind these drought-tolerant traits. In the process, they're establishing a more informed breeding strategy for their new "Agrisure Artesian" corn hybrid. "Our goal in developing Artesian was to increase yield of corn grown in the presence of drought stress with no change in yield in the absence of stress," said Duane A. Martin, Ph.D., Product Lead in Commercial. "We were able to, through a unique scientific process, carefully select genes that provide multiple means for the plant to use water more efficiently. Artesian corn is protected from drought all season long. That's what makes it different from anything else in the industry."

iCommunity spoke to Dr. Martin and Joseph Clarke, Ph.D., Principal Scientist - Genomics to Breeding, to learn more about how Syngenta used a genomics approach to develop Artesian hybrid corn.



Duane A. Martin, Ph.D., is Product Lead in Commercial and Joseph Clarke, Ph.D., is a Principal Scientist - Genomics to Breeding at Syngenta

Q: What are some of the challenges you face in the corn industry?

Duane Martin (DM): The corn industry and the crop protection industry have been successful in controlling plant diseases, weeds, and insects. What remains a challenge industry wide, is ameliorating abiotic stress, ie crop stress due to non-living factors such as temperature and rain. With the need to increase top-line crop production and efficiency, optimizing water use and drought tolerance is an area of interest to the industry.

Q: What is a corn hybrid?

DM: A corn hybrid is the result of a single cross between two inbred lines, the progeny of which is then planted and harvested. Prior to hybrid corn development in the 1930s, corn globally was a self-pollinated crop with yields less than half of what we have today. Every seed corn company has what they call a library of genetics—literally thousands of inbred corn plants. The combinations of those inbred corn plants produce the hybrid corn plant that is sold to growers. The grower plants it one year, harvests it, sells it, and goes back to the seed company to buy new seed for the next year. You can't save hybrid corn seed and plant it the next year because the genes segregate differently in progeny. The next generation will consist of a bunch of odd looking corn plants.

Q: What approach did Syngenta use in developing Artesian corn hybrids?

DM: Rather than study how corn hybrids perform in drought conditions, we took a more holistic approach. Our premise was that the hybrids should make the most of available water. A grower may use limited or supplemental irrigation, they may be a dry land corn grower in an area that is nearly always under water stress, or they may be a highly productive grower in the central or eastern Corn Belt. Those considerations formed our approach to water use in corn, which was different than our competitors, who generally focused on a more traditional, drought tolerance approach in low-yield environments.

Joe Clarke (JC): Evidence suggested there are multiple protections against drought that operate together so we started with a hypothesis-driven candidate gene approach. Early in the process we looked through our genetic library and initially nominated several candidate genes that we believed would affect different plant structures or processes positively to allow the plant to respond to water stress better. From the larger pool, we were able to focus on a set of core genes that met our criteria. Our process also enabled us to identify specifically the right version (allele) of these genes and where on the genome they resided. We then used highly managed field trials to measure and validate the allelic effects.

Q: How did you select the candidate genes?

JC: The candidate genes were selected through a hypothesis-driven approach represented by the integration of microarray-based expression profiling across several crop and model species, relevant genetic information, public literature, and functional annotations. Comparative genomics and extensive sequencing was used to define the "target" candidate gene in corn. The target corn sequence was then converted into a series of genotypic assays intended to distinguish the allele/haplotype for each candidate gene across a diverse population. The genotypic information was used to assess the effect of each allele towards phenotypic diversity with regard to performance under drought. Our success was well above the 5–10% success rate often attributed to the non-hypothesis driven approaches of the time.

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Q: How did you validate the performance of the candidate genes in the field?

DM: We used managed stress environments where we essentially take corn with these genetic backgrounds and grow them in areas that receive no rainfall during growing season. The only water the plant receives is through a sophisticated irrigation system where we closely control and track the water supplied through moisture sensors. We take these plants with the genes in question to the edge of death due to lack of water. Then we recharge the soil profile, allow the plant to recover, and measure yield and final effect on the plants.

Q: What were the results?

DM: We were able to validate performance in the field and determine roughly what each gene candidate contributed to the water-stress response. We were able to move a variable number of these genes into the right genetic backgrounds and produce the Artesian corn hybrids that we ultimately sell to growers. With the ability to identify multiple genes that make up the Artesian trait, we don't have just one or two good ways to protect the corn plant against drought, we have multiple modes of action against drought that help to protect the plant all season long. The performance of Artesian is roughly 2× what we see from other technology in the field, because we've identified functions, processes, and structures that affect the plant's ability to deal with water stress. Under water stress, the plants yield up to about 15% more than a corn hybrid without the Artesian trait. Under extreme water stress, such as a prolonged, significant drought, we've increased corn yield as much as 40% with the Artesian trait versus a hybrid without the trait.

Q: How do you connect the trait to the mode of action?

JC: One of the distinguishing features in our development of Artesian is that it was based on a hypothesis-driven approach. Understanding the modes of action helps understand what is changing inside the plant at the genomic level to make it drought tolerant.

DM: A key loss in corn due to drought is what's called a barren ear, an ear of corn that sets few to no kernels. One of the modes of action that we've identified is a gene that appears to make kernel sets much more consistent in these Artisan hybrids, even under water stress, than in other hybrids. It's that specificity that allowed us to define the genes we're after, describe in general what we think they do, and move them into a testing system. We concluded that under extreme drought conditions this gene performed the way we thought it would. It became one of several core genes that we would locate in our breeding program and use for Artisan hybrids.

JC: Illumina is enabling us to build on the Artesian success by contributing towards the next generation of Artesian alleles at higher resolutions of trait impact and potential modes of action. We are also using Illumina platforms in the expression profile space to help better understand and leverage the modes of action assigned to the current Artesian product release. Our newer Illumina-based expression data sets on the HiSeq System offer several advantages over the existing hybridization-based expression data with regard to resolving potential modes of action.

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Q: What do your customers think of the Artesian hybrids?
DM: Artesian is a fun project to work on because it's a very visual technology. When growers plant these hybrids and see them under water stress, next to another corn hybrid, they see a dramatic difference between the corn plants. Under very hot drought stress conditions, an Artesian hybrid will generally be tall and dark green with its leaves fully unfolded. It looks like it's growing more comfortably than other hybrids, which are likely to be shorter, have poor color, and have the leaves rolled up to conserve water. Growers loathe seeing that response in their fields.

In 2012, I talked to a grower in Colorado, which is a very dry area to grow corn. It was so dry that he lost his entire crop, even his Artesian hybrids. But he called us to say that his Artesian hybrids lasted weeks longer than any other hybrid that he had. He was so enthused that he could go out there and see this visual difference in his field. He said he would likely not see that level of drought again for 20 years, but it proved to him that Artesian corn hybrids were different than the ones he was growing previously.

Q: What impact has genomic technology had on the project?
JC: Artesian was launched in 2012, but the project started in the early 2000s. The plant genomic world changed radically around 2007, facilitated by Illumina. We migrated to Illumina GoldenGate® arrays when they became available to help with the marker introgression aspects after the Artesian alleles were defined. We used GoldenGate arrays to help with the genotyping, characterize the populations, and to help with the molecular breeding and stacking of the alleles.

In 2008, we dove into large-scale Illumina-based sequencing projects in corn to increase our marker platform by an order of magnitude or two. The information was so much better. It was a clear decision to move from the candidate gene/Sanger sequencing approaches that defined the original Artesian to Illumina-based genome-wide association studies (GWAS) to define the next product releases under the Artesian brand. At the time, the cost of analyzing data on a gene-by-gene basis almost equaled the cost of taking an Illumina-based approach over the whole genome.

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Q: How has the success of the Artesian project impacted your approach to hybrid development?
DM: We've seen such success with our project in corn that we're anxious to try a similar approach in other crops. Water is such a limiting resource in so many areas and so many crop production systems. We could use a similar approach, in wheat, which is grown predominantly in the western states under extremely variable precipitation and in many cases under very dry conditions. If we could introduce wheat varieties that might yield 15% better under water stress, it would be a significant addition to agriculture as well as to our business.

JC: Because the hypothesis-driven, knowledge-driven approach worked it means we can continue taking time to understanding the biology behind what the GWAS results are telling us. In addition to using Illumina technology to discover SNP markers that identify GWAS alleles for us to breed with, we can use it to understand the biology behind why these alleles are doing what they're doing. There's some biology driving the decision making and the HiSeq 2500 System is helping us resolve that biology.

Illumina • 1.800.809.4566 toll-free (U.S.) • +1.858.202.4566 tel • techsupport@illumina.com • www.illumina.com

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