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Chapter 3- Genetics and Physiology of Crop Water Use

Prem S. Paul
University of Nebraska at Lincoln, ppaul2@unl.edu

Monica Norby
University of Nebraska-Lincoln, mnorby1@unl.edu

Gillian Klucas
University of Nebraska-Lincoln

Ashley Washburn
University of Nebraska-Lincoln, awashburn2@unl.edu

Elizabeth Banset
University of Nebraska-Lincoln, ebanset1@unl.edu

See next page for additional authors

Authors
Prem S. Paul, Monica Norby, Gillian Klucas, Ashley Washburn, Elizabeth Banset, and Vicki Miller

3

GENETICS AND PHYSIOLOGY OF CROP WATER USE

Genetics and Physiology of Crop Water Use

Panel

Marty Matlock
Professor of Ecological Engineering, University of Arkansas

Sally Mackenzie
Ralph and Alice Raikes Chair, Plant Sciences, and
Director, Center for Plant Science Innovation, University of Nebraska–Lincoln

Roberto Tuberosa
Professor of Biotechnology Applied to Plant Breeding, University of Bologna, Italy

Richard Richards
Chief Research Scientist, Commonwealth Scientific and
Industrial Research Organisation (CSIRO), Australia

James Specht, Moderator
Haskins Professor in Plant Genetics, University of Nebraska–Lincoln

The session explored key issues and challenges in developing crops that can produce more yield with less water, including plant breeding techniques, corn water use modeling and transitioning plant innovations from the laboratory to the field. The panelists brought many years of experience and perspectives from different areas of expertise. Each panelist gave an overview of his or her subject area and the panel then responded to audience questions.

From left: Sally Mackenzie, Roberto Tuberosa, Richard Richards and Marty Matlock

A Global Assessment of Corn Water Use As Affected by Climate, Genetics and Scarcity

Marty Matlock, University of Arkansas

Marty Matlock described a high-resolution water assessment model he and colleagues are developing to determine how much water corn uses globally and to evaluate the balance between rainwater stored as soil moisture (green water) and water from surface water or groundwater sources (blue water). With a framework for assessing these characteristics, the model can analyze various scenarios, such as climate change and water demand by region.

Marty Matlock

"Our quest is to develop a modeling framework that has utility for decision-makers," Matlock said.

To achieve high resolution, Matlock and his colleagues divided the globe into geospatial resolution cells of 5 minutes by 5 minutes, or about 10 kilometers by 10 kilometers. After inputting data for each cell, the researchers ran the model to determine yield. Comparing the results between the model's predicted yield and observed data, the model was calibrated using high-resolution input and yield data available for the U.S. heartland (Corn Belt). From potential yield data, researchers can determine water demand.

Matlock and his colleagues chose the CERES-Maize simulation model embedded in the Decision Support System for Agrotechnology Transfer (DSSAT) because it uses daily rainfall inputs. It is therefore sensitive to critical threshold water scarcity, a more important element for kernel development than annual rainfall. Using the CERES model required collecting and entering daily data sources into each cell for each characteristic. Temperature and radiation data were acquired from the Climate Research Unit; precipitation data were acquired first from the Tropical Rainfall Measuring Mission and later from the National Climatic Data Center; and soil characteristics came from the ISRIC-World Inventory of Soil Emission Potentials soil dataset.

After running the model, Matlock's team assessed its predicted values against global crop yield data obtained from Foley et al. published in *Science* magazine in 2005. The model did well in dryland regions, but predictions did not match observed yields in wetter regions. To calibrate the model using the highest geospatial resolution yield data, they focused on the U.S. heartland region, inputting high-resolution soil, temperature and rainfall data.

"We're modeling one stalk of corn and extrapolating that to the world," Matlock said. "If I really wanted this model to be right, I'd quit right now. All models are wrong; some models are useful. The question is, is there utility with this

Genetics and Physiology of Crop Water Use

model? And I would argue that, yes, there's strong utility because of its process-based development."

To establish the model's parameters, Matlock and his colleagues developed a set of parameters based on what other researchers use to model at the field or plot level. They first performed calibration runs on a 40-county region, then on a larger region spanning several hundred counties. For single cultivars, the model is sensitive to the four parameters that define the way a single corn stalk responds to precipitation and temperature. In the case of a single cultivar, the predicted versus observed graphs were not effective. However, modeling using nine cultivars and selecting the cultivar that best fit yield resulted in good calibration between predicted and observed yield. Mapping the results showed these four variables are associated with other important variables as well.

The next step will be evaluating the model's ability to adequately predict water use. The model then can be used to analyze land use impacts on blue water resources; to determine a stress-related water footprint using regional stress factors; and to develop a series of water stress indices, including the impact on base flow under various scenarios, such as climate change, population change and industrial demand.

A lack of regional high-spatial and high-temporal data remains a problem, Matlock said. In addition, he continued, "We lack integrated models for the outcomes of concern: the 'so what?' part. We have to build that from scratch because life cycle assessment, risk-based models just don't cut it for these sorts of social and economic impacts."

Plant Research Innovations in the University: When Will They Apply to the Real World?

Sally Mackenzie, University of Nebraska–Lincoln

Despite tremendous innovations in plant research today, the challenges of integrating research into the real world leave many of those innovations stuck in the laboratory, Sally Mackenzie said. She described the approach taken by the Center for Plant Science Innovation at the University of Nebraska–Lincoln (UNL) to move research to the field.

Sally Mackenzie

Some of the research occurring in universities includes innovations to improve seed nutrient content, modify plant architecture for water use efficiency and alter properties to enhance shelf life.

Many innovations stem from the ability to sequence the genomes of major crop species, which is helping researchers understand the genes and mechanisms that one day may improve plant tolerance to drought and other beneficial characteristics.

These innovations and capabilities are already happening in the laboratory, Mackenzie said, adding that "the innovation is not what limits our ability to actually come up with some interesting solutions."

If universities have laid the technological groundwork, why are they not real players in the dialogue? she asked. On most campuses, the link between the lab and true agriculture biotechnology is nonexistent. In contrast, the Center for Plant Science Innovation is building its approach around taking research to the field. Such an approach requires four things:

- Broadening in-house capabilities in crop transformation. On most campuses, transformations stay in model species in the lab.
- Building capacity for large-scale, APHIS-compliant transgene field testing to see whether transformations will come to fruition.
- Designing a critical mass of researchers to facilitate interactions, a "center" concept that often fails because the departmental nature of university settings hinders the free flow of ideas.
- Providing cross-departmental and cross-disciplinary accessibility, a huge challenge facing university researchers, particularly in interacting with government researchers.

Although UNL's center has met each of these requirements, moving to commercialization remains challenging. Much of the problem lies with the U.S. regulatory system, Mackenzie said, which often requires case-by-case review by at least two regulatory agencies before innovations can be grown large scale. The process creates paperwork and enormous expense that can preclude public sector participation.

"The innovations sit on the shelf, and this is going to be a huge challenge, I predict, for the Bill & Melinda Gates Foundation and for anyone else who really wants to integrate these kinds of technologies," Mackenzie said. "There are really two conduits to allowing anything to come out for public use, and that's right now, as far as we're concerned, DuPont and Monsanto."

These companies have the ability to manage the regulatory process, but their involvement is limited compared to the available innovations. The average American consumer is unwilling to pay more for many valuable products that, for example, enhance growth capability in response to abiotic stress, Mackenzie said. This situation creates a logjam for biotechnology opportunities.

To help UNL participate in moving innovations to the field, the university is establishing Nebraska Innovation Campus, a collaboration of academia, industry and government. Other universities are establishing similar opportunities to let researchers participate more meaningfully in moving needed innovations from their laboratories to real world applications.

Genetics and Physiology of Crop Water Use

Mapping and Cloning QTLs for Drought Tolerance in Durum Wheat and Maize

Roberto Tuberosa, University of Bologna, Italy

Roberto Tuberosa presented data on research projects for mapping and cloning quantitative trait loci (QTLs) to increase yield in durum wheat and maize. QTLs are stretches of DNA closely linked to two or more genes that underlie a phenotypic characteristic. Both projects used forward genetics, in which biparental crosses identify the genes and QTLs underlying the crop's adaptive response to drought. The genes' DNA can then be sequenced and annotated. "The reason I like the QTL approach is that we ask the plant, 'What is important?'" Tuberosa said. "We do not go in with a preconceived hypothesis. I personally think it's a little bit dangerous, particularly when we deal with complex traits such as drought tolerance, to go in with the candidate gene approach."

Roberto Tuberosa

Wheat QTL Mapping

Tuberosa described finding a QTL important for drought resistance in durum wheat. Drought tolerance is important because sensitivity to drought not only decreases yield but also impairs the flower's quality and the quality of the final product. He defined drought tolerance as not merely surviving drought, but also producing high yields under a wide range of water availability. Tuberosa collaborates with the seed company Produttori Sementi Bologna.

The project's objective, which Tuberosa coordinated among 10 partners throughout the Mediterranean Basin, is to identify QTLs for yield water use efficiency in related strains, grown across environments with a broad range of water availability. Researchers used biparental, or linkage, mapping with Svevo and Kofa, a durum desert wheat grown in Arizona. In rainfed and irrigated field trials, the researchers obtained a tenfold range in yield, demonstrating the negative effect of drought and allowing them to test the mapping population across a broad range of yield potential. Interestingly, the morphophysiological trait that best correlated yield with the environment and genetics was peduncle length.

Tuberosa and his colleagues identified two major QTLs on two different chromosomes that account for a large portion of phenotypic variation among the genotypes tested. A significant portion of the phenotypic variability stems from the QTL interactions. Grain weight had the largest effect on yield. Now, the researchers are fine-mapping and cloning the QTLs and both chromosomes in conjunction with the TriticeaeGenome project.

Maize QTL Mapping

Tuberosa's QTL mapping and cloning research in maize focused on two traits important for drought tolerance: root architecture, because of its importance in avoiding the negative effects of drought, and flowering time, because phenology is the most important trait enabling plants to complete a life cycle before drought stress damages yield.

A wind storm had caused stalk lodging in an isogenic line with high root abscisic acid (ABA), but not in the line with low ABA. The team discovered that high-ABA plants had a higher root clump. Test crosses in both well watered and water-stressed conditions performed by collaborator Yu-Li in China demonstrated a significant effect on grain yield. Notably, the most productive genotype had a lower root mass.

They discovered the opposite effect in another recently isogenized QTL for root architecture, underscoring Tuberosa's point that "different QTLs, but also the same QTL, can have different effects according to the genetic background."

Tuberosa described a process to shortcut moving from QTL course-mapping to fine-mapping using introgression line libraries instead of isogenization. To date, he and his colleagues have identified three major QTLs and are fine-mapping one.

Genomics provides many opportunities to identify candidate genes for yield and traits important for drought resistance, Tuberosa concluded, but phenotyping remains the major constraint.

Breeding for Water Productivity in Temperate Cereals

Richard Richards, CSIRO, Australia

Richard Richards is optimistic that water productivity can be doubled in many environments but believes advances will be gradual. He described his successes using a trait-based approach to developing improved varieties of wheat and other temperate cereals.

Richards emphasized the need to develop a benchmark for water use efficiency by eliminating concepts such as drought tolerance and drought resistance that are not easily measurable and may be unrelated to productivity.

Richard Richards

Genetics and Physiology of Crop Water Use

Benchmarking would allow scientists to measure improved genotypes and farmers to compare successes from year to year or from changes in practices. "I really want to get rid of this idea of drought tolerance and drought resistance," Richards said.

Conventional breeding has been remarkably successful and will continue as the cornerstone of improvements and the benchmark for future gains. Yet, despite tremendous yield gains from new varieties, these successes are not enough. Greater yield improvements are needed.

Richards described the logical relationship between water use and yields. Soil evaporation sets a lower limit of water required to obtain any yield, but as available water increases, yield does, too. That relationship's slope forms a boundary limiting potential yield based on available water. Yet, most farms' yields fall well below the current potential. Filling the gap between actual and potential yields would double production, Richards said.

Improving management systems, such as stubble retention and earlier sowing, would have the biggest impact on filling that gap. "They're going to have a much more immediate impact; they're going to be adopted more widely; and that's where the biggest gains are going to be made, in many cases," he said.

But better genetics is also important, and surprisingly, the most important genes for drought resistance are those that promote a healthy root system. Water productivity in dry environments requires a healthy root system to use the water and nutrients effectively. Therefore, genes that resist rootworm and cereal cyst nematodes and tolerate acidic soils have provided major breakthroughs in drought resistance.

Harvesting wheat from experimental plots

By improving the factors limiting water productivity, it is also possible to raise the maximum potential yield beyond current levels. Because heritability for yield is low in highly variable rainfed systems (a high-yield variety one year may produce a low yield the next due to different conditions), Richards and his colleagues use a trait-based approach to find new water-productive varieties. Other advantages to a trait-based approach include the ability to focus on genetic variability for the most important trait; faster genetic gains because the heritability of the trait may be higher than yield; more cost-effective evaluations other than yield; ability to conduct work out of season; greater amenability to marker-assisted selection; and the potential to pyramid multiple yield-enhancing traits.

To increase yields in a water-limited environment, a selected trait must increase water use, water use efficiency or harvest index. One example of a successful variety developed using this approach involved a trait that selects for 13C molecules, which offers a 10 to 12 percent yield advantage in very dry environments. A variety packaged with this and other beneficial traits, such as disease resistance, was released.

Richards and his colleagues are working on other traits believed to be extremely important in dry environments, including seedling establishment, shoot and root vigor, and transpiration efficiency. Surprisingly, they found it most effective to select for each trait under favorable conditions rather than unfavorable or drought conditions. Although they have identified quantitative trait loci (QTLs), which are stretches of DNA closely linked to two or more genes for a trait, they have found selection for phenotype, not markers or QTLs, to be the most efficient selection method.

"The phenotype is the limiting factor in every single case," Richards concluded. "Important traits for crop improvement are complex, and in many cases molecular markers, or QTLs, may not be very effective. [Phenotypic information] is the massive challenge … and we've just started to realize this."

Questions and Answers

Moderator James Specht: ***What are the biggest possibilities for accelerating yield productivity rate of gain?***

Richard Richards sees enormous opportunities in improving the overall health of root systems. "I think it's an area of absolutely essential investment in the future," he said. "I think we really have to understand what's going on below ground to make further gains."

Roberto Tuberosa considers genomics an important tool to increase genetic variability and the heritability of beneficial traits. Knowing the exact gene or quantitative trait locus limiting a useful trait provides opportunities for manipulating that trait. Close collaboration with breeders who understand the limiting factors in the field is necessary.

Sally Mackenzie agreed that an integrated approach is necessary, particularly for understanding phenotypes. Scientists have examined phenotypes from a genetic perspective without understanding the metabolic phenomena underlying the processes. A greater understanding of metabolic biochemistry is needed, and that will take a systems approach.

Audience question: ***How would you recommend distributing limited resources to improve water use efficiency?***

Mackenzie would put more funding into broad, interdisciplinary training in the plant breeding community, particularly as technologies integrate.

Richards agreed, adding that integration must include farmers who, with their intimate knowledge of growing crops, will make important observations and should be encouraged to work with academia. He also emphasized the need for bold initiatives, such as the International Rice Research Institute's C4 Rice Project.

Genetics and Physiology of Crop Water Use

Marty Matlock believes information technology is an important area for funding, noting the highest levels of technology for earth observation available to U.S. farmers come from the French and Indian governments.

Tuberosa suggested rethinking what plant breeding means and training plant breeders to coordinate teams of different specialists and to recognize the value of each piece. "The entire interdisciplinary effort has a bigger value than the value that each single person brings into this relay."

Audience question: ***What advice would you give young scientists starting in the field?***

Mackenzie described UNL's educational approach, which emphasizes training students to integrate and manage new biotechnologies and comply with federal guidelines; capitalize on bioinformatics; communicate effectively; and understand and influence policy.

Tuberosa believes private-public partnerships are valuable avenues for training graduate students to work in teams and produce research more easily translatable to seed industry and farmers' needs.

Richards urged students to find their passion and to seek collaborators. "The more you can collaborate and the more you can discuss, the more value you're going to have, the more success you're going to have."

Audience question: ***What are the key improvements necessary for modeling water use efficiency, such as data availability and focusing on potential rather than actual yield?***

Data availability is the biggest limitation, Matlock answered. It is impossible to calibrate or validate a model without knowing "what was." Potential yield also is a critical variable. However, it is impossible to evaluate the impact of changing environmental conditions, climate or resource availability on actual yield using potential yield. Both are necessary.

Irrigated fields under conservation agriculture in Mexico

Audience question: ***What is Richards's experience with canopy air temperature as a wheat-screening tool?***

It's an exciting area of research, Richards said. The coolness of the canopies has been an important selection tool but greater understanding is needed to fully harness it.

Audience question: ***Why does Tuberosa urge caution regarding the candidate gene strategy?***

The more complex the trait, the less heritable it is, Tuberosa responded. The relationship between a specific gene and a phenotype becomes blurred. In those situations, the candidate gene approach becomes less effective, or at least riskier.

Audience question: *To what extent do epigenetic factors contribute to breeding programs?*

Epigenetics plays a huge role in traits, Mackenzie answered. For example, perturbing the mitochondria can reprogram the plant to grow fundamentally differently in response. However, understanding how epigenetics will influence breeding strategies is just beginning.

Audience question: *The highest-yielding cultivars in irrigated conditions often produce the highest yields under stress conditions, as well. Is it symmetrical, or is it a process of selecting genotype by environment?*

Richards said he emphasizes selecting under favorable environments because it maximizes genetic variants; therefore, heritability is higher, and greater genetic gains are made. In a bad year, those yields may be the lowest, but farmers are unlikely to do well regardless.

Audience question: *Maize and rice are sensitive to drought stress at flowering. Does Richards believe that selection for tolerance to stress at flowering without directly selecting under stress is possible?*

Richards described the mechanism that causes sterility under stress conditions. If researchers can keep some of the genes that switch off, causing sterility, to remain on, fertility of the pollen grains will increase. "It's an exciting area of research, and I think it's going to have massive consequences in so many of our crops," he said.

Moderator Specht: *Concluding remarks?*

"What is good in nature for survival, I don't think is good for crops," Tuberosa said. Sometimes researchers must "fool" the crops into not reacting strongly to environmental cues.

Understanding how Mother Nature has learned to survive difficult environments provides important clues for crops, Mackenzie said. "But I would tend to agree these really drought-hardy materials that we've been able to come up with would probably not be of much agronomic interest."

Matlock suggested moving beyond thinking of information as privileged toward a corporate reporting framework that encourages information to move from the laboratory to the field and higher "so that we can actually inform our future rather than react."

Richards appealed to the youngest audience members to be excited by the challenges and to integrate and discuss ideas as much as possible. "I want to emphasize the value of our young people here and the potential impact that they can have in some very, very exciting areas of science in a very, very uncertain world."