

## Rising optimism fuels falling numbers research

By Camille M. Steber and Kimberly Garland Campbell

Since 2011, Pacific Northwest growers have suffered major losses from discounts due to low falling numbers (FN). In 2016 alone, it's estimated that Washington farmers lost anywhere from \$30 million to \$200 million due to FN discounts.

The first Falling Numbers Summit was convened in February 2017 when researchers and wheat industry representatives identified two major needs: improved testing and breeding varieties with improved resistance to low FN.

The second Falling Number Summit was held in Portland in late January 2019. This article is intended as an update on solving the challenges of low FN. Go to [steberlab.org](http://steberlab.org) for more information.

Dr. Camille Steber with the Agricultural Research Service of the U.S. Department of Agriculture (USDA-ARS) opened the second summit by describing the FN test, its foibles and the causes of low FN. The Hagberg-Perten FN test measures starch degradation in wheat based on how many seconds it takes a stirrer to fall through a gravy made by boiling a water-whole grain meal mixture. Starch is digested by the enzyme alpha-amylase. More alpha-amylase causes more starch digestion, allowing the stirrer to fall faster through thinner gravy. Overseas customers require an FN of more than 300 seconds to avoid the risk of poor end-use quality due to elevated alpha-amylase.

FN varies with atmospheric pressure because the test uses boiling water, and the temperature at which water boils depends on pressure. Altitude and weather (high and low pressure systems) cause differences in atmospheric pressure. Previous altitude correction tables had large altitude increments and no adjustment below 2,000 feet, leading to over- or under-correction.

Dr. Stephen Delwiche (USDA-ARS) used a hyperbaric chamber to derive an equation for continuous correction of FN based on barometric pressure. This should reduce problems with grain shipments testing at below 300 seconds at their destination after testing at above 300 seconds upon departure.

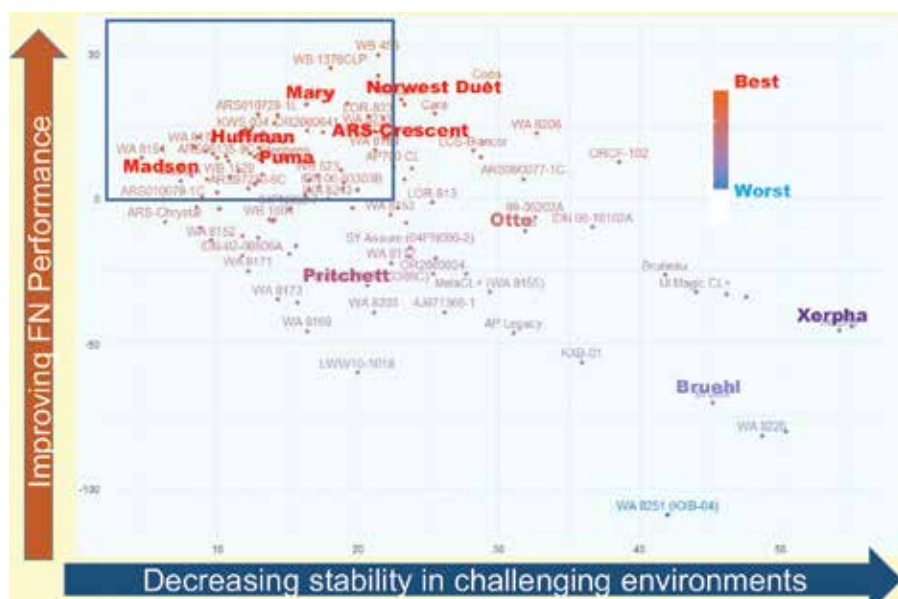


Camille Steber (left) and Kim Garland Campbell, both scientists with the Agricultural Research Service of the U.S. Department of Agriculture, address the second Falling Number Summit which took place in Portland Jan. 29-30 and featured a dozen scientists talking about the status of their research on both preharvest sprout and late maturity alpha-amylase.

Paul Katovich provided an elevator industry perspective on low FN. As general manager of HighLine Grain Growers Cooperative, Katovich is in the unenviable position between exporters who need grain with an FN of more than 300 seconds and farmers who are potentially docked if grain is under that threshold. Between 2002 and 2019, Katovich has experienced nine low FN events.

The elevator industry does "blend off" low FN grain, but it takes a great deal of sound grain to raise a low FN grain lot based on a liquefaction equation. While a sample measured at or above 270 seconds can be blended with sound grain to meet the 300 second threshold, an FN of 100 seconds is toxic; it cannot be blended off and can ruin a whole elevator of high FN grain, Katovich said.

HighLine Grain values its multigenerational relationship with farmers and wants to avoid degrading this



**Figure 1.** A factor analytic model was used to compare the tendency of cultivars to have low falling numbers (FN) between 2013 and 2016 based on FN testing of the Washington State University variety trials. The vertical axis shows increasingly good performance for FN while the horizontal axis shows an increasing tendency to respond poorly to challenging environments. Varieties in the boxed region have better resistance to low FN.

trust through harsh FN discounts. A typical discount schedule docks farmers 25 cents per bushel for every 25 seconds below 300; that means 50 cents for a sample below 275.

To avoid such harsh penalties, HighLine has gone to a continuous discount schedule of a half-penny per point (second) discount, but, as Katovich said, “It doesn’t take many mistakes to go broke. We are taking a calculated risk. We are trying to absorb as much of that risk for the farmer as possible.”

Elevated alpha-amylase in grain resulting in low FN has two causes: pre-harvest sprout (PHS) and late maturity alpha-amylase (LMA). PHS occurs when rain falls on mature grain before harvest with alpha-amylase providing fuel/sugar for germinating seedling growth, just as Mother Nature intended. In contrast, the purpose of alpha-amylase produced during LMA is unclear. LMA is triggered by a cold shock during the soft dough stage of grain filling.

Because PHS and LMA have different genetic and environmental triggers, wheat breeders must select separately for both. Washington State University (WSU) researcher Galina Mikhaylenko and Dr. Alecia Kiszonas (USDA-ARS) have shown that PHS appears to have a stronger impact on cake quality than LMA. Future research is intended to determine whether LMA-affected grain poses a lower risk to end-use quality.

Dr. Andrew Ross from Oregon State University (OSU) provided a historical perspective on falling numbers and improving the FN test. Ross was introduced to falling numbers in 1984 as a postdoctoral scientist in Australia. He says the main problem is using FN as a predictor of end-use quality instead of using a direct measure of quality (like baking a cake).

“The falling number test has its issues, but it is a valuable metric in the marketplace because the FN test can identify wheat with a risk of poor performance in end-use applications,” he said.

Ross is one of the inventors of the Rapid-Visco Analyzer (RVA), a test that is

more accurate than FN but only if the slower version (12 minutes) RVA test is run. Regardless of the testing method developed, there will always be a problem with “noise” at the 300 second threshold. That’s because alpha-amylase levels are the strongest determinant of FN below 300 seconds, but above 300 seconds, many other factors impact FN scores.

Two alternatives to the FN test were discussed during the second Falling Numbers Summit. Chun-Peng (James) Chen (Ph.D. student) and Dr. Zhiwu Zhang (WSU) have embarked on a mission to separate low and high FN grain using hyperspectral images of intact grain. This will be done using machine learning and the sort of mathematical modeling used for facial recognition. Since this is a new project, its accuracy is still unknown. It will be valuable, however, if it provides a cheap, fast, nondestructive method to prevent inadvertent mixing of high and low FN grain.

Another test method is being pursued at WSU by Drs. Amber Hauvermale, Andrew McCubbin and Mike Pumphrey. Hauvermale has produced monoclonal antibodies to the wheat alpha-amylase protein in order to create a highly sensitive ELISA “strip test.” While such assays use milled grain, they are accurate and are currently being used to detect GMO crops, pregnancy and disease. Researchers are investigating whether both methods can be used to differentiate between LMA and PHS, something the FN test cannot do.

The best way to reduce the risk of low FN is to select cultivars with higher resistance to low FN. WSU Ph.D. student Stephanie Sjoberg and Dr. Kimberly Garland Campbell (USDA-ARS) are developing statistical methods to quantify the genetic differences in varieties



It was standing room only at the Falling Number Summit where scientists, students, farmers and the elevator industry gathered to hear the latest on efforts to develop a better test to measure alpha amylase activity as well as work aimed at weeding out susceptible varieties.

and their responses to challenging environments.

A Factor Analytic Model allows researchers to separate genetic and environmental effects, looking both at a variety's overall FN performance (y-axis) and at the response to increasingly challenging environments (x-axis). See Figure 1. Looking at soft white winter varieties over three years showed that while newer releases like Otto and Pritchett have not yet reached an ideal level of resistance, they are superior to older varieties like Bruehl and Xerpha. Other varieties like UI-WSU Huffman, Mary, Trifecta, Puma, ARS-Crescent, ARS-Castella, LCS Hulk and Norwest Duet already show better resistance to low FN.

While PHS is an age-old problem, LMA was discovered mainly by Australian scientists in the 1980s. To select for LMA resistance, breeders need a reliable test that can be conducted in the greenhouse. WSU graduate student Chang (Chloe) Liu, Pumphrey and Steber found that LMA screening in the greenhouse is complicated. There is approximately a five-day window in grain development during which a cold temperature shock can induce LMA. The timing of this window varies between cultivars. Different plants, different tillers and different grains in a spike reach this stage at different times. The Northwest cultivars that Liu has examined don't induce LMA with a heat shock, but do with cold shock.

Given the labor-intensive nature of LMA and PHS screening, mapping resistance loci is a goal that's needed to speed up the breeding process. Dr. Jianli Chen from the University of Idaho (UI) has identified molecular markers linked to FN, visible sprouting and LMA in spring wheat. Since some of these loci fall on the same chromosomes as previously identified loci, future work will examine if any of these traits are controlled by similar genes.

Dr. Shantel Martinez from Cornell University mapped FN and PHS tolerance loci in soft white winter wheat while working at WSU with Dr. Arron Carter (WSU) and Steber. These markers will be used for genomic selection in WSU wheat breeding programs and others.

Efforts are also underway to use the power of gene editing to address the

FN problem without generating GMO wheat. Oregon State University wheat breeder, Dr. Robert Zemetra, is developing an efficient CRISPR/Cas9 system targeting a gene on chromosome 4A known to provide PHS tolerance in wheat and barley breeding programs on multiple continents.

Finally, Dr. Daolin Fu (UI) described using chemically induced mutations along with gene editing to create modified versions of the wheat alpha-amylase gene, TaAmy3. TaAmy3 is produced during seed development, not during LMA and sprouting. He will examine if mutations in TaAmy3 lead to higher FN.

Although much progress has been made since the first Falling Number Summit, challenges remain. A "fix" remains elusive, but research toward better management is already helping protect wheat export customers, elevators and farmers. ■

## More on FN

The Washington Grain Commission (WGC) podcast, *Wheat All About It!*, devotes three episodes to the Jan. 29-30 Falling Number Summit held in Portland. Listen to Paul Katovich of HighLine Grain explain the disconnect between the elevator industry and farmers in episode 113; to researchers explaining their work in episode 114; and to Oregon State University Professor Andrew Ross clarifying why a 60-year old test is still the best available, episode 115. Access the podcast on iTunes, Stitcher, Overcast, Pocket Cast, FM Player or by going to the WGC website at [wagains.org](http://wagains.org) and clicking on summaries. ■