Science Societies

Reaching for the ‘Top Shelf’

Breeding Barleys for Distilling That Don’t Contain Carcinogen Precursor

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To turn cereal grains into the whiskey in your old fashioned, a delicate dance of art and science is performed. The first step is the creation of barley malt, which can provide the base ingredient or the enzymatic power needed for distillation.

But some barley (*Hordeum vulgare* L.) used for malting produces glycosidic nitrile, a compound that can be a precursor to a carcinogen called ethyl carbamate. Luckily, glycosidic nitrile production can be bred out—like in the new glycosidic nitrile-null (GNO) barley variety 'Top Shelf,' recently released by Oregon State University and registered in the *Journal of Plant Registrations* (https://doi.org/10.1002/plr2.20366).

"Glycosidic nitrile itself is not necessarily the problem," says Campbell Morrissy, first author on the *Journal of Plant Registrations* registration who now works in the brewing industry. "It's just the fact that during the unique steps of distillation to make barley-based whiskey you … end up with ethyl carbamate."

As the malting and distilling industries become more aware of glycosidic nitrile, barley-breeding programs like Oregon State's are hard at work producing high-yielding,

climate-resilient GNO varieties. “How can we use barley breeding or plant breeding in general for a better future? Well, this is a great application,” says Ashley McFarland, the vice president and technical director of the American Malting Barley Association (AMBA), a trade organization representing the interests of barley malt producers and users. “If there's concern here from a human health perspective, why not solve some of those problems through genetic improvement?”

From Malt to Bottle

Glycosidic nitriles (GNs) are naturally occurring metabolites that act as defense mechanisms in a plant. They’re activated through enzymes released by insects chewing on a plant’s leaves, explains Harmonie Bettenhausen, a lecturer at the James B. Beam Institute for Kentucky Spirits at the University of Kentucky and the director of the Harwick College Center for Craft Food & Beverage. “It creates a little cyanide bomb and repels the pest,” Bettenhausen says.

Let’s drill down into that aforementioned whiskey-making dance. First comes malting, when grains are steeped in water and then exposed to air. Here, you want plump and uniform grain kernels, McFarland says. “The more plump and uniform they are, the better they’ll malt and the better extract potential there is,” she says, referring to the amount of sugar the malt will release with later mashing.

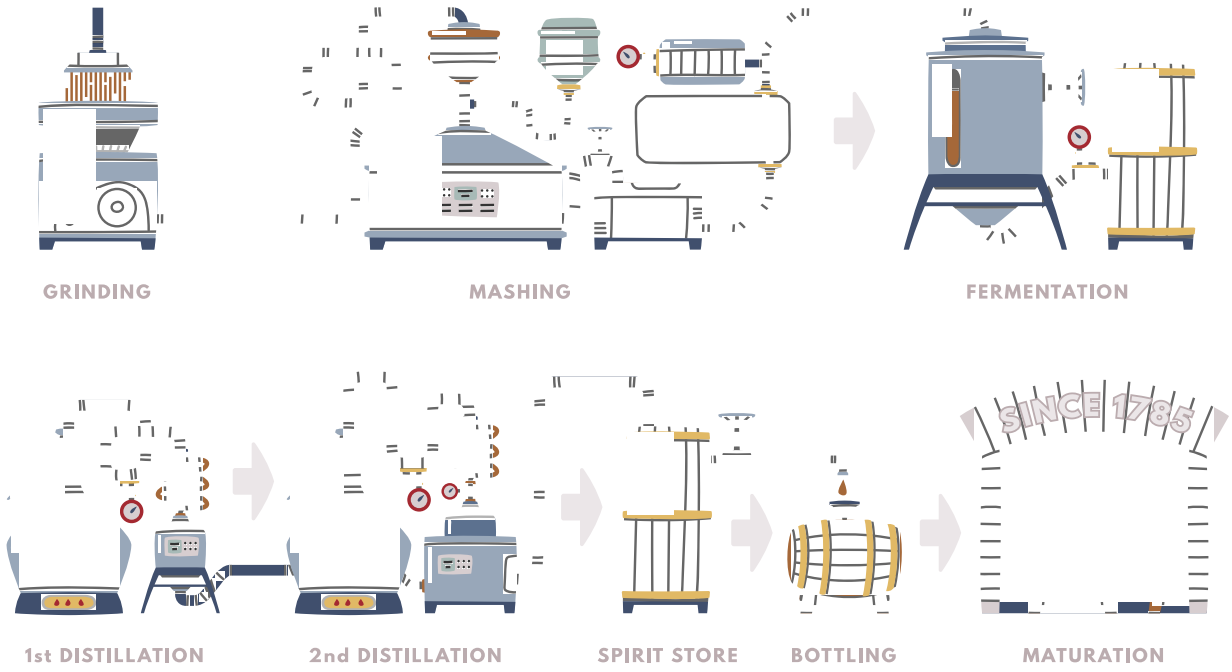


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After steeping, the kernels are germinated: roots and shoots emerge and the starches within break down via changes in temperature, moisture, and rotation delicately controlled by the maltster. Finally, the malt is “kilned” to stop germination and dry it out. Throughout malting, different changes and choices in the process can create different color and flavor outcomes. “It’s really recipe dependent,” McFarland says.

When malt whiskey is the end product, the dried malt is then ground into grist and mixed with hot water to produce a liquid called wort. The next step is fermentation: yeast is added after the wort is cooled and feeds on the sugars within to produce alcohol. Then comes distillation: the liquid goes into pot stills, which are typically copper but can vary in shape depending on the desired final taste of the spirit. When

heated, the alcohol within vaporizes into the stills' necks and re-liquifies when cooled. The process can be repeated, and the liquid is then housed in casks that are tucked away in warehouses for years while it matures into the liquor that is bottled, sold, and drank.

From Glycosidic Nitriles to Ethyl Carbamate



Photo courtesy of Adobe Stock/Kunz Husum.

During the fermentation process, GNs in the original malting barley are converted through several steps into hydrogen cyanide, which is then catalyzed into ethyl carbamate (EC) by ethanol and copper present in the distilling still (more EC can continue to be produced in the barrel as the whiskey is aged). Levels of GN in barley malt destined for distilling can differ depending on different moisture conditions during steeping and/or time and temperature changes during germination. There are a few ways distillers can mitigate EC via contact with the copper, but "the whole thing is just a little bit wonky," Bettenhausen says.

Ethyl carbamate can also be found at low levels in fermented foods like bread and in other alcoholic beverages like beer and wine. But it shows up at high levels in spirits because of this ethanol, copper, and hydrogen cyanide reaction. "When you make malt for brewing, there's no copper catalyst there, there's no distillation, so you don't

actually end up with ethyl carbamate regardless of GN in your barley," explains Morrissy of this beer/spirits difference.

At high doses, EC has been shown to cause cancer in lab mice and has been classified as "probably carcinogenic" by the International Agency for Research on Cancer and "reasonably anticipated to be a human carcinogen" by the U.S. National Toxicology Program. Ethyl carbamate was first identified in whiskey by the Liquor Control Board of Ontario in 1985, and researchers at the United Distillers International Research Center in Scotland continued investigating the issue into the 1990s. "Time went on and they discovered it was not only malted grains, but how we malted the grains," Bettenhausen says.

Today, EC is strictly regulated in distilled spirits at 150 parts per billion by the United Kingdom, the European Union, and Canada. In the United States, however, regulation is voluntary at 125 parts per billion. While breeding GN out of barley has been a main focus in the U.K., it hasn't been on the radar in the U.S. "There are regulatory pressures in Europe that are mandating a different approach," McFarland explains.



Clay Kaasa, Director of U.S. Grain Procurement at Great Western Malting Company, stands in a field of Top Shelf near Burley, ID. Photo courtesy of Pat Hayes.

The stricter regulations are tied in part to the role of Scotch whiskey as an important U.K. product and export. “That’s a large portion of their barley usage and is a very dominant industry in the alcohol beverage space,” Morrissy says. “Much of the Scotch whisky industry is malt whiskey, so 100% malted barley, unlike the United States, which is mostly a bourbon-based industry or rye-based industry and which is using a very small portion of malted barley.” Those processes use majority corn, rye, or wheat and just a small portion of barley malt. “But now we see high levels [of GN] even with small inclusions in the bourbon,” Bettenhausen adds.

Breeding for Null

In barley, GN production is controlled by one inherited cluster of genes that, via certain combinations of expression, lead to high, low, or no GN. As such, making GN0 barley is, at least genetically, a cinch. “This trait is single-gene-controlled, effectively, so you can remove a transcription factor and breed it out, and there doesn’t seem to be any sort of linkage with other issues or other genes or traits of interest,” Morrissy explains.

After EC and GN were identified as conc



Fusarium head blight on barley. Photo by Javier Segura/CIMMYT.

erns for malting, barley breeders in the U.K. began developing GN0 varieties in earnest. Initially, they may have selected for the trait accidentally when breeding for resistance to a pathogen called powdery mildew (*Blumeria graminis* f. sp. *hordei*), which uses a recognition factor connected to GN expression. "It's a fairly straightforward proposition—you've got barleys that do not produce the compound—there's apparently no negative effect on the agronomic or quality performance of those varieties," says Patrick Hayes, Professor Emeritus of Barley Breeding and Genetics at Oregon State. Now, around 90% of barley acreage planted in Scotland is GN0. But the U.S. lagged behind for a while. "There just wasn't a whole lot of interest in the GN0s in the U.S.," adds Hayes, an ASA and CSSA member.

Then about a decade ago, Hayes' program at Oregon State released the GN0 variety 'Full Pint' (https://bit.ly/3Y5RpPi). The appearance of the GN0 allele in the Oregon State breeding program and Full Pint's subsequent GN0 status was also a happy accident, Hayes says. "It's a testimony to the power of free exchange of germplasm between breeding programs because it just came kind of hitchhiking along with crosses that we had made with material from the ICARDA/CIMMYT program in Mexico"

"We had no idea that GN0 was in the program. The breeder there probably had no idea that GN0 was in the program," he continues. They didn't even know Full Pint was GN0 until they decided to screen a sample of their varieties for the trait. Lo and behold, it was—and the fact that the Oregon State researchers had the allele in their germplasm meant they could swiftly get to work once their colleagues in the malting and distilling industries started asking about varieties that didn't produce GN.

Other breeding efforts also began developing GN0 barleys. However, there is only one GN0 variety currently on AMBA's list of recommended malting barley cultivars, LCS

Odyssey. “Odyssey production is limited to a certain area, so we need other varieties to be able to be grown in other areas, and we need people to pick them up,” Bettenhausen says.

Odyssey, Full Pint, and most of the GNO varieties grown in the U.K. are spring varieties, meaning they are planted in the spring and harvested in the late summer. By contrast, winter-habit barleys planted in the fall or facultative ones that can be planted in either fall or spring can address some of the challenges of climate change, Hayes says. “Many of our spring barley production areas are increasingly characterized by super high temperatures and stresses,” he explains. These include water limitations in the West and hot, humid conditions in the upper Midwest that favor the development of diseases such as fusarium head blight. “What you’re trying to do is avail yourself of the opportunity to move into production areas where barley simply wasn’t grown because it couldn’t survive the winter,” Hayes says.

Creating Top Shelf



A seed increase of Top Shelf at the OSU Hyslop Farm near Corvallis, OR. Photo courtesy of Pat Hayes.

Oregon State's breeding program is focused on developing facultative varieties. "It gives farmers greater flexibility, but it can achieve the same level of low-temperature tolerance as the winter type," Hayes says. But they still develop and release winters along the way. "If you have a great winter variety, you're going to pursue it."

The program was "grinding along," in Hayes' words, producing double haploids and putting them out into field tests with the goal of developing a variety that had malting quality attributes for beer making. But after the GN issue came on their radar through conversations with colleagues including Bettenhausen, they decided to take the GN0 allele for a spin in what they called the "Distiller's Delight" trial, testing how they could modulate GN0 lines through nitrogen fertilization for different distilling outcomes.

The variety that became Top Shelf was added to the trial somewhat late in the game, Morrissy recalls. Its initial cross had been made in 2015, and it had gone through the typical pipeline of seed increases and yield trials. As with most field trials, "we had kind of the routine seven plagues of Egypt, if you will, that are always afflicting our crop," Hayes says. But Top Shelf consistently showed good agronomic potential and high yield. Often a selection that breeders are very keen on falls short along the pipeline, Hayes reflects. "Those are the heartbreak moments of plant breeding." But both Top Shelf and a sister variety calledGN0 Vivar didn't have any major flaws, he says.

Also, importantly, "it behaves itself in the malthouse," Hayes adds. "That's the great grief of malting barley breeding. We'll have things that are resistant to diseases and that have a high yield and so forth," and then the malting industry tests it and gives it the thumbs down, he says. Over several years, the researchers put Top Shelf through small-scale malting trials with onsite and partners' malting machines, a multi-day

process of steeping the barley and stimulating the biochemical process to break down the starch and protein packets within. When Top Shelf takes up moisture, it shows an appropriate amount of enzymatic activity, and the enzymes reach a target threshold that is modulated and balanced. “It meets a lot of profiles that are really desirable for both brewing and distilling,” Morrissy says.

And on that note, while Top Shelf’s GNO status makes it obviously attractive and applicable for distilling, its malt can be used for brewing beer too without issue. “That just gives farmers tons more options,” Morrissy says. “I think it's a great barley variety, period, GNO or not.”

What’s Next for GNO?



Scott Fisk and Margaret Krause, OSU, Clay Kaasa, Great Western Malting Company, and Scott Adams, Golden Valley Warehouse in a field of Top Shelf near Burley, ID. Photo courtesy of Pat Hayes.

Of course, farmers need plentiful seed to be able to plant Top Shelf in their fields. The program is in the process of doing seed increases, but that pipeline is a “tightrope act,” Hayes says, and one that takes time. Top Shelf was released under a nonexclusive

license, meaning anyone interested in selling the seed can contact Oregon State's Office of Commercialization and Corporate Development to obtain a license. Some of the program's industry partners are testing commercial-scale malting of the variety and working on producing larger amounts of the seed. "Right now, Top Shelf has the enviable status of there being more demand than there is supply," Hayes says.

Volume is a problem across the board for GN0 barley. If every distiller in the country wanted GN0 barley malt, there simply would need to be "more barley farmers [and] more people picking up GN0 varieties," Bettenhausen says. Barley farmers aren't always set up for success, she says, and novel varieties can present challenges on the ground. "It's tough when you don't know how something's going to grow and you don't know what to expect from it," she says. "Barley's very sensitive. Each variety's got a different personality, and it's tough every year. With things changing with the climate, we've got an even tougher situation."

Getting the word out about GN and EC in the first place is also a priority for the researchers. Over the last few years, Bettenhausen and colleagues have worked hard to communicate this information to the large distilling companies, connecting them with breeders like the team at Oregon State.

When McFarland first came on board at AMBA in 2021, she says the GN issue wasn't really part of the conversation. At the James B. Beam Institute Industry Conference at the University of Kentucky two years ago, she recalls chatting with Bettenhausen about how important the issue was—but it wasn't being discussed widely at the meeting. "Fast forward a year, I'm at the same conference, and now everyone's talking about it," McFarland says. "I definitely think it's on folks' radar now, but it's still a kind of new emerging issue in the industry that maybe not everyone understands fully yet."

The American Malting Barley Association has altered its breeding guidelines to state that they want barley breeders to be targeting GNO varieties from here on out as that's what barley end users will be increasingly looking for, McFarland says. A main driver of industry interest is getting out ahead of future regulations, she explains, and AMBA and their partners are just beginning to have those early conversations about how guidelines will evolve. While the EC limit in U.S.-made spirits is voluntary now, that may change in the future, especially in light of the growing popularity of American single malt whiskey. The single malt category is more prevalent in Europe but is just starting to catch on in the U.S., McFarland says. "People see a lot of opportunity for marketability with a single malt product."

It's going to be a matter of years before anyone can make an old fashioned with whiskey made from Top Shelf barley, single malt or not. But Hayes and colleagues did get to sample some "new make" made from the variety. Also evocatively called "white dog," new make is the stuff that comes immediately off the still—unaged whiskey that when sold illegally historically was known as moonshine. "We have done a tasting, and with the James Beam Institute, we did an experimental distillation," Hayes says. And that moniker? "We really enjoy naming our varieties," he says. Hayes credits his wife for engaging in the long car ride discussions that led to this one's label. "Top Shelf came up because it rolls off the tongue very easily, and it just conjures up visions of something that is the best."

Dig Deeper

Read about Top Shelf in the *Journal of Plant Registrations* here:

Morrissy, C. P., Filichkin, T., Fisk, S. P., Helgerson, L., & Hayes, P. M. (2024). Registration of ‘Top Shelf’ barley: The first glycosidic nitrile-null, winter malting cultivar to be released in North America. Journal of Plant Registrations, 18, 241–249. https://doi.org/10.1002/plr2.20366

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