Prime editing technology

Leaf rust is just the beginning

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IN THE RACE to develop disease resistant cultivars, new biotech tools can offer an inside track.

A research team at Agriculture and Agri-Food Canada (AAFC) Morden is exploring the use of prime editing, which is an emerging CRISPR technology. The project, *Genome Editing to Accelerate Pre-breeding Delivery of Improved Genetics,* is funded through the Canadian National Wheat Cluster*.

Building on research by Dr. Sylvia Cloutier (AAFC-Ottawa) on wheat's wild relatives with resistance to leaf rust, Dr. Andriy Bilichak and his research team have been working to introduce leaf rust resistance from goatgrass *(Aegilops tauschii)* into elite wheat cultivars.

Prime editing allows researchers to begin the breeding process in the lab, relying on tissue culture to test potential cultivars.

"This is a new technology with limited applications in crops. We are at proof-of-concept stage, with lots of unknowns. The technology works best when we know which DNA modifications need to be introduced to convert susceptible genes to the resistant ones," said Dr. Bilichak.

In the case of leaf rust, those genes are known. Working with one broad-spectrum gene (Lr34) and a narrow-spectrum gene (Lr21), they are testing the efficiency of various prime editing constructs to introduce precise edits in the complex wheat genome.

"In plant pathology, having a broad-spectrum gene is like the Holy Grail because it gives resistance to a number of pathogens. It recognizes all the leaf rust isolates but doesn't offer very strong resistance. Lr21 offers strong resistance to fewer isolates. A combination of different Lr genes can offer durable resistance to the leaf rust pathogen," said Bilichak.

The team is working with Fielder, a cultivar that is amenable to tissue culture, and which is susceptible to leaf rust.

Dr. Bilichak said, "With prime editing, if a cultivar is lacking a particular trait, you can precisely edit the DNA. Editing directly in an elite cultivar that is already otherwise well-adapted can cut the time needed to develop new cultivars in half."

Additionally, the team is leveraging a cis-genic approach, using native wheat genes and their natural regulatory elements, to enhance resistance to wheat leaf rust, under the leadership of Dr. John Laurie at AAFC Lethbridge.

The project also includes a strong discovery component led by Drs. Sylvie Cloutier, Frank M. You and Brent D. McCallum. By integrating wheat wild relatives with a series of genomics and bioinformatics tools, the team aims to identify and characterize novel genes conferring resistance to wheat leaf rust.

"If we can go back to the germplasm in gene banks to screen for resistance to various pathogens in crops' wild relatives, we'll be able to use this technology to introduce those genes in elite wheat lines," said Bilichak.

The research will lead to a better understanding of the potential for prime editing in crop breeding for wheat, as well as other grains.

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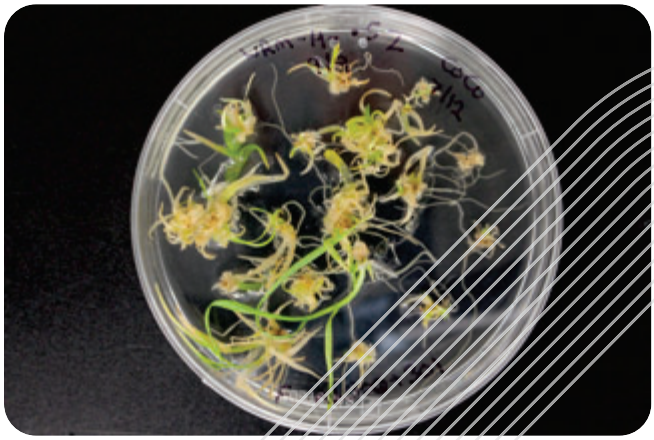

LEFT AND RIGHT: Wheat samples including genes from wild relatives growing in various conditions to determine resistance to leaf rust. | PHOTO CREDIT: DR. ANDRIY BILICHAK