Gene editing and the future of sunflower production

By Don Lilleboe

A discussion on gene editing – what it is and what it may mean for sunflower production – was presented at last year's National Sunflower Association's (NSA) Summer Seminar by Brent Hulke and Dylan Moxley.

Hulke is a sunflower research geneticist with the USDA Agricultural Research Service in Fargo, North Dakota. Moxley is a graduate student in the Rieseberg Lab, in the Department of Botany at the University of British Columbia.

Changing the DNA sequence

Gene editing has gathered much attention in recent years among scientists working in human and plant biotechnology research. Gene editing refers to the utilisation of biotech techniques to orchestrate changes to specific DNA sequences within the genomes of living organisms, so it is indeed a method of genome engineering.

However, unlike genetically modified organisms (GMOs), in which a gene is inserted into one species from a different species (transgenic), the gene editing concept in plants focuses on engineering the plant so that it expresses its own genes in a way that is different to the standard expression. In that sense, gene edited plants could offer a less controversial route to breeding progress than the GMO pathway. Both methods, though, should be considered a process towards a destination rather than the destination itself.

Developing technology

The Rieseberg Lab is well known and highly regarded for its work with high-throughput genomic methods, bioinformatics, ecological experiments and evolutionary theory, studying the origin and evolution of species, domesticated plants and weeds. The sunflower genus (*Helianthus*) is a primary focus at the lab.

Moxley explained to the NSA audience that 'genome engineering' in the broad sense is used to better understand the function of a specific gene, to introduce desirable traits and/or to remove or modify a trait. Its advantages include the ability to rapidly generate desired lines, saving time and resources.

Genome editing systems to date, Moxley noted, have included recombinant DNA technologies such as gene guns and agrobacterium transgenics. While these technologies have the benefit of being very versatile in terms of moving genes from one organism to another, they have been controversial among the general public.

The possibilities of CRISPR

Moxley observed that true gene editing refers to lesser-known systems (e.g. zinc finger and TALENs) and the now more recognisable CRISPR. CRISPRs are specialised stretches of RNA that guide the process to the target DNA sequence. The protein Cas9 (CRISPR-associated) is an enzyme that acts like a pair of molecular scissors, capable of cutting strands of DNA.

CRISPR technology has emanated from the natural defence mechanisms of bacteria (agrobacterium-mediated transformation). While it holds exciting possibilities for editing genomes of sunflowers and other plants, it comes with some big challenges. With sunflower specifically, the challenges include low transformation efficiencies (< 1%) and the lack of a stable transformation pipeline.

USDA's Hulke told Summer Seminar attendees that using pollen as a foreign DNA carrier currently offers the most promise when it comes to editing the sunflower genome via CRISPR technology. It's inexpensive and routine (once the method has been optimised), he noted.

Not surprisingly, though, there are significant research obstacles, too. The regulatory environment should be less of an obstacle than that of GMOs, Hulke added, since gene editing does not include the insertion of foreign DNA into the genome. However, bringing it to commercialisation is complicated by legal issues involving intellectual property rights.

If, and when, CRISPR technology becomes a reality in sunflower production, Hulke envisions several targets for application. *Sclerotinia* and *Phomopsis* resistance genes would certainly be on that list, as well as genes affecting seed size and oil composition. Other areas of research would be downy mildew, and rust and herbicide resistance.

For now, though, the utilisation of CRISPR technology in sunflower breeding "is still in the hypothetical phase," Hulke said. Biologists are in the process of mastering its fundamentals in 'amenable' species; and sunflower is not currently amenable due to transformation issues. Plus, there are regulatory issues (not so much in the US, but rather abroad*), as well as continuing legal debates regarding intellectual property rights.

So, while the potential for gene editing may be promising, making it a reality would come with a lot of, as yet, unresolved scientific and commercial complications.

*In July 2018, the Court of Justice of the European Union issued a judgement that organisms created through many newer genome editing techniques are to be regulated as genetically modified organisms (GMOs) in the EU. "This decision subjects such organisms, and food and feed products containing these organisms, to expensive and lengthy approval processes as well as traceability, labelling and monitoring obligations," noted a USDA Foreign Agricultural Service report on this development.

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