Applying CRISPR beyond Arabidopsis thaliana

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Arabidopsis thaliana. Credit: Wikipedia.

Few technologies have made as big a splash in recent years as CRISPR/Cas9, and rightfully so. CRISPR/Cas9, or clustered regularly interspaced palindromic repeats (CRISPR) and associated genes, is a bacterial gene editing toolbox that allows researchers to edit genomic sequences much more precisely and efficiently than previously possible, opening up doors to new ways of doing research. As with many new biotechnologies, the application of CRISPR in biology began with genetic model organisms such as *Arabidopsis thaliana*. In research presented in a recent issue of *Applications in Plant Sciences*, Shengchen Shan and colleagues review the prospects for expanding the use of CRISPR for research beyond genetic model plant species.

"Developing CRISPR/Cas9 in nongenetic models holds enormous potential in plant biology," says Shan, a Ph.D. candidate at the University of Florida, Gainesville. "Almost all CRISPR applications have focused on genetic models, like *Arabidopsis* and major crops. However, many nongenetic models are economically important and/or biologically significant."

Indeed, while our in-depth knowledge of the biology of genetic model species makes them enormously valuable for research, these species represent only a tiny sample of plant diversity. Much could be gained from expanding genetic studies of non-model species, including through the use of new tools like CRISPR.

Polyploidy, or whole genome duplication, is one example Shan and colleagues highlight of a research area that CRISPR could revolutionize. "We are very excited about future CRISPR/Cas9 applications in studies of evolutionary biology, especially the role of polyploidy in plant evolution. Polyploidy is a major evolutionary force in land plants, but the genotype-phenotype relationships following genome doubling are largely unknown," explains Shan. "CRISPR/Cas9 could be used to disrupt the observed genetic patterns in polyploids, and therefore, by comparing phenotypes of the mutant and wild-type plants, researchers will be able to reveal the biological underpinnings of many intriguing genetic consequences, such as nonrandom gene losses following polyploidy."

Researchers looking to study non-genetic model species using CRISPR will face some technical obstacles, including difficulties in genetic transformation, the process of introducing and expressing foreign genes in a plant species. However, looking to transformation systems established in closely related species can help. According to Shan, "The biggest barrier is developing the transformation system. Although we have summarized a few commonly used transformation strategies, plant transformation is a very species-specific process." He goes on to note, "As we reviewed, both transformation and CRISPR systems from a phylogenetically closely related genetic model species can be adapted for an initial attempt of establishing CRISPR in a newly studied plant species." In this way, the development of transformation and CRISPR systems in genetic models has helped pave the way for expanding CRISPR beyond these species.

The research done using CRISPR in genetic model species thus far has lowered the bar for entry in other ways too, as researchers have optimized techniques, hammered out details, and built up expertise in the use of this exciting new technology. "For people studying ecology or evolutionary biology, using a state-of-the-art technique in molecular biology research, like CRISPR/Cas9, might seem intimidating," says Shan. "However, based on our own experience, with the help from collaborators from a molecular biology lab, it is not that hard!"

More information: Shengchen Shan et al, Considerations in adapting CRISPR/Cas9 in nongenetic model plant systems, *Applications in Plant Sciences* (2020). DOI: 10.1002/aps3.11314

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