Opinion

The promise of gene-editing in plant agriculture

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From the genomes of microbes have risen two novel gene-editing technologies that hold great promises in plant agriculture. Although gene-editing techniques such as the uses of zinc-finger nuclease (the "ZFN" method) and of transcription activator-like effector nuclease (the "TALEN" system) have been around for some time,[1] they have not find traction in plant agriculture applications because they are expensive as well as lack programming efficiency to have potential for broad applications with ease.

The novel CRISPR-Cas9[2–5] and NgAgo-gDNA[6] gene-editing technologies have now been shown to be more easily programmable and may be broadly adaptable to have great potential in plant agriculture, although both have limitations. For example, the CRISPR-Cas9 system relies on the presence and proximity of protospacer adjacent motifs ("PAM")[7] while the NgAgo-gDNA appears to rely on a high salinity[8,9] alkaline[9] or high temperature[10] environment for the unwinding of the double-strand DNA helix of the gene targets for their "guide" nucleotides (crRNA for CRISPR-Cas9 and ss-gDNA for NgAgo-gDNA, respectively) to target their nuclease activities. The former's reliance on PAM may limit the range and selection of gene targets that may be edited. The latter's apparent reliance on a high salinity, alkaline, or temperature environment, although a challenge, may be less of a constrain because plant cells have in general reasonably high tolerance to such environmental stress, at least for a short period of time likely to be sufficient for the "editing" of the gene to complete. This challenge shall await further experimentations by plant researchers who can manipulate their experimental conditions to overcome. Together with plant tissue culture techniques, these two new gene-editing technologies may found great applications of significance in plant agriculture especially with their "multiplexing" capabilities since polyploidy is an important characteristic of most modern crop plants.[11]

Recent advances in the use of molecular biological techniques in the studies of cell and developmental biology in plants have illuminated a long list of potential target genes, the inactivation or suppression of which by gene-editing technologies, may bring forward highly desirable agricultural traits.

For example, the methods of specifically applying CRISPR-Cas9 to genetically modify plants and fungi is the subject of a patent application by The Penn State Research Foundation[12] and further described in a recent report of the successful use of CRISPR-Cas9 by Yang at Pennsylvania State University to edit one of a family of six genes encoding polyphenol oxidase that reduced the enzyme's activity by 30% and hence slowed the "browning" of the common white button mushroom (*Agaricus bisporus*).[13] This new trait will prolong the shelf life of the product and enhance its value and attractiveness to consumers.

Other genes that have been identified in the past to be responsible for fruit ripening may similarly be suppressed or inactivated entirely to control fruit ripening for improved storage, transport, and shelf characteristics. For example, ethylene is a hydrocarbon gas widely recognized to be the fruit-ripening hormone. Two key enzymes, the ACC synthase and the ACC oxidase, are important in the biosynthesis of ethylene.[14] They may be suppressed or inactivated using the new gene-editing technologies to slow or totally prevent fruit-ripening. The gene-editing technologies may be preferable to and more effective than the older antisense-RNA technology because the latter is known to be "leaky" in its gene-suppression (not total or permanent inactivation).[13] By "editing" the genes directly, the end result may more likely be an improved suppression of the known genes. Then at the appropriate time, ethylene or its active analogue, propylene, may be introduced to facilitate fruit-ripening when it is desirable. Fruits that are sensitive to undesirable over-ripening causing great value loss include apples, apricots, banana, guava, Kiwifruit, mango, papaya, passion fruit, peach, pear, persimmon, plum, sapodilla, and tomato.[15] Most of these are high cash-value crops.

Other potential target plant genes of agronomic value to suppress or inactivate have been reported by Yanofsky et al.,[16] at the University of California San Diego. The flowering of plants may be suppressed to allow energy be more channeled to support enhanced vegetable growth since flowering consumes 25 to 35% of the energy of a typical plant.[17] The lignin biosynthesis of plants may also be enhanced to reduce lodging and loss.[18] Harvest loss due to lodging has been documented to be significant in crops like rice[19] and corn.[20] Pod-shattering or pre-mature seed-pod breakage is also a significant cause of harvest loss in pod-bearing crops such as oil seeds (rape or Canola) and soybean.[21] Seed pod-shattering can cause up to 50% seed/bean loss and represents a risk of significant value loss to farmers. Certain genes may be suppressed or inactivated to prevent pod-shattering.[22]

Although gene suppression or inactivation by gene-editing is the most straight forward approach to acquire more desirable plant traits, the gene-editing technologies can be applied to "seek-and-replace" specific features of any DNA and hold further promises. National and multinational agricultural biotech giants and seed companies are taking notice and have begun using and investing heavily in the use of these technologies. Other desirable traits and characteristics that may be achieved include drought-resistance, ablation of self-pollination to enable hybrid seed production, elimination of allergenic proteins, oil composition manipulation, lessening susceptibility to diseases etc. Alliances between DuPont and Caribou Biosciences; and between Dow et al.,[23] are signs of great things to come and hold promise to benefit the global food supply.

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Alan Paau
Advance Immune Medical Systems Inc, USA

Correspondence: Alan Paau, Advance Immune Medical Systems Inc., USA, Tel 1-607-229-9810, Email apaau@hotmail.com

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Conflict of interest

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