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Cultivated sweet potatoes contain *Agrobacterium* inserted T-DNA that is absent in wild sweet potato relatives. A natural GMO?

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Genetic modification (GM) of food crops is a very hot topic in today's society. When researchers discovered that certain bacteria had the ability to transfer their own DNA into plant genomes, the applications seemed endless. By taking advantage of this transfer DNA (T-DNA) mechanism, scientists can selectively transfer specific genes into the target plant to provide agronomic or health benefits. For example, Bt corn contains bacterial genes that prevent significant yield loss due to insect damage, and golden rice contains genes from corn and bacteria that together generate vitamin A precursors not typically found in rice, providing a nutritional benefit in the staple food crop.

Two bacterial species within the genus *Agrobacterium* (*A. tumefaciens* and *A. rhizogenes*) have been studied in tremendous depth, and both possess this natural ability to insert T-DNA into the genome of over 92 different families of plants. Exhaustive studies of these organisms have allowed scientists to understand this mechanism extremely well (Pitzschke and Hirt 2010). Even though scientists are equipped with fundamental knowledge of the T-DNA insertion mechanism, a mistrust of genetic engineering has arisen in the general population over recent years. Opposition to GM crops can stem from fear of the unknown, a misunderstanding of the underlying science, feelings of GM being "unnatural", or views that the risks of GM food outweigh the rewards. Regardless of the source, this mistrust has significantly stalled scientific progress in the area of developing transgenic crops. Therefore, the recent finding from Kyndt et al. 2015 that *A. rhizogenes* T-DNA is present in all

commercial sweet potato cultivars, but absent in wild sweet potato relatives, leads to an interesting discussion point.

This study found two regions of the cultivated sweet potato (*Ipomoea batatas*) genome called *lbT-DNA1* and *lbT-DNA2*, that were homologous to known *A. rhizogenes* genes. The authors generated DNA probes specific to these T-DNA regions to prove these sequences were in the sweet potato genome using a Southern blot. In addition, they examined the *lbT-DNA1* region of a second sweet potato cultivar and found that it contained the *lbT-DNA1* sequences, but had evolved to have a duplication and inversion of these genes, plus a transposon insertion into the region. These differences between the two cultivars imply that the original *lbT-DNA1* insertion event took place in an ancient sweet potato ancestor, allowing the region sufficient time to differentiate after the original insertion event. A genome walking experiment revealed that *lbT-DNA1* is located in an intron of an actively transcribed sweet potato F-box gene, raising the question of whether or not these *lbT-DNA1* genes are also transcribed. Indeed, RT-qPCR results show that at least four of the *lbT-DNA1* genes and two of the *lbT-DNA2* genes are expressed in every tissue type tested (although their bar graph is lacking error bars!).

The real impact of this study comes from the large survey of cultivated and wild sweet potato lines for the presence or absence of *lbT-DNA1*. The survey showed all 291 cultivated lines contained *lbT-DNA1*, but none of the wild sweet potato relatives tested had the *lbT-DNA1* insertion. This supports their hypothesis of an ancient insertion event – perhaps prior to human domestication of sweet potato. The fact that *lbT-DNA1* is found in every cultivated line, but no wild lines, implies that this insertion may have had an agronomic benefit that was selected for by humans. Sweet potato cultivars have retained these genes, which adds to the argument that these genes could still be having an agronomic benefit. The authors also identified two different *lbT-DNA2* alleles, and used this information to generate a segregating population from parents with the two different *lbT-DNA2* alleles in order to identify a benefit to having one allele or the other. After examining 76 of the progeny lines, the authors concluded that no direct benefit could be distinguished between the alleles. Additional testing for potential agronomic benefits of retaining *lbT-DNA1* remains to be done. Nonetheless, a T-DNA insertion providing a beneficial trait (to humans) is precisely the goal of modern plant biotechnology, and it appears to have happened at least once without human intervention.

After reviewing this paper, our Genetics Journal Club discussed the possible implications of these findings. Where does the fear of GM technology stem from? Why is conventional breeding and chemical mutagenesis more accepted than GM biotechnology methods of creating genetic diversity? Could the findings in Kyndt et al. 2015 help alleviate the fear of *Agrobacterium* mediated GM methods in crops? But perhaps the most important question discussed was, what can we do as scientists to initiate a healthy, active conversation about the subject?

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