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Transmission of on-target and off-target mutations in a series of CRISPR/Cas9-induced soybean plants

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There is considerable interest in editing DNA sequences for crop improvement using CRISPR/Cas technologies. However, the genetic changes that can result from these technologies, including the relative frequency of sequence alterations that occur at the intended locus (on-target) compared to other locations in the genome (off-target) have not been well characterized. To address this question, we performed genome-wide resequencing of ten soybean T0 plants that were transformed with a range of different CRISPR/Cas9 transgenes, looking to identify novel SNPs, small insertion-deletions, and larger structural variants. To gauge the transmission rate of on-target and off-target mutations, we also sequenced eight T1 or T2 progeny from selected individuals, including individuals where the CRISPR/Cas9 transgene was still present and others where the transgene had segregated away. This talk will highlight recent findings from this work, including the presence of mutations in the CRISPR/Cas9 plants compared to control plants, and the relationship between transgene transmission and the inheritance of on-target and off-target mutations. Lastly, we will also discuss the possible factors that could contribute to the appearance of off-target mutations in these lines, including natural de novo mutation processes, tissue culture, transformation, and the activity of the CRISPR/Cas9 reagents, per se.