Alleviating Peanut Allergy Using the CRISPR/Cas System

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Peanut allergy is the most common cause of severe or fatal food-associated anaphalaxis and results in approximately 200 deaths per year in the US alone. Although much research has been to develop treatments such as vaccines, a cure has yet to be created. To address this issue, we would like to genetically modify immunodominant allergen sequences found in peanut using the CRISPR (clustered regularly interspaced short palindromic repeats) /Cas9 system. In 2012 the CRISPR/Cas9 system was reported to be a powerful genome-editing tool. The precise targeting of the microbial system can be utilized to reveal the function of genes that influence phenotypes often seen in diseases and illnesses such as food allergies. This revolutionary RNA-guided gene-editing tool involves the introduction of double-strand breaks (DSB) at a specified location in target DNA. The formation of DSBs induces either the DNA repair mechanisms known as non-homologous end joining (NHEJ) or homologous recombination (HR) and consequently will lead to mutations in the target genome through an insertion/deletion of nucleotides. Depending on the repair mechanism, these insertions/deletions can be random or very specific. Herein we describe the use of CRISPR/Cas 9 to generate targeted disruption of a major allergen gene found in the peanut genome. Target site selection, as well as the design, construction, verification and use of guide RNAs (gRNAs) for sequence-specific CRISPR/Cas-mediated mutagenesis in *Arachis hypogaea* will be shown.