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Targeted introduction of heritable point mutations into the plant mitochondrial genome

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摘要	<p>The development of technologies for the genetic manipulation of mitochondrial genomes remains a major challenge. Here we report a method for the targeted introduction of mutations into plant mitochondrial DNA (mtDNA) that we refer to as transcription activator-like effector nuclease (TALEN) gene-drive mutagenesis (GDM), or TALEN-GDM. The method combines TALEN-induced site-specific cleavage of the mtDNA with selection for mutations that confer resistance to the TALEN cut. Applying TALEN-GDM to the tobacco mitochondrial nad9 gene, we isolated a large set of mutants carrying single amino acid substitutions in the Nad9 protein. The mutants could be purified to homochondriomy and stably inherited their edited mtDNA in the expected maternal fashion. TALEN-GDM induces both transitions and transversions, and can access most nucleotide positions within the TALEN binding site. Our work provides an efficient method for targeted mitochondrial genome editing that produces genetically stable, homochondriomic and fertile plants with specific point mutations in their mtDNA.</p>
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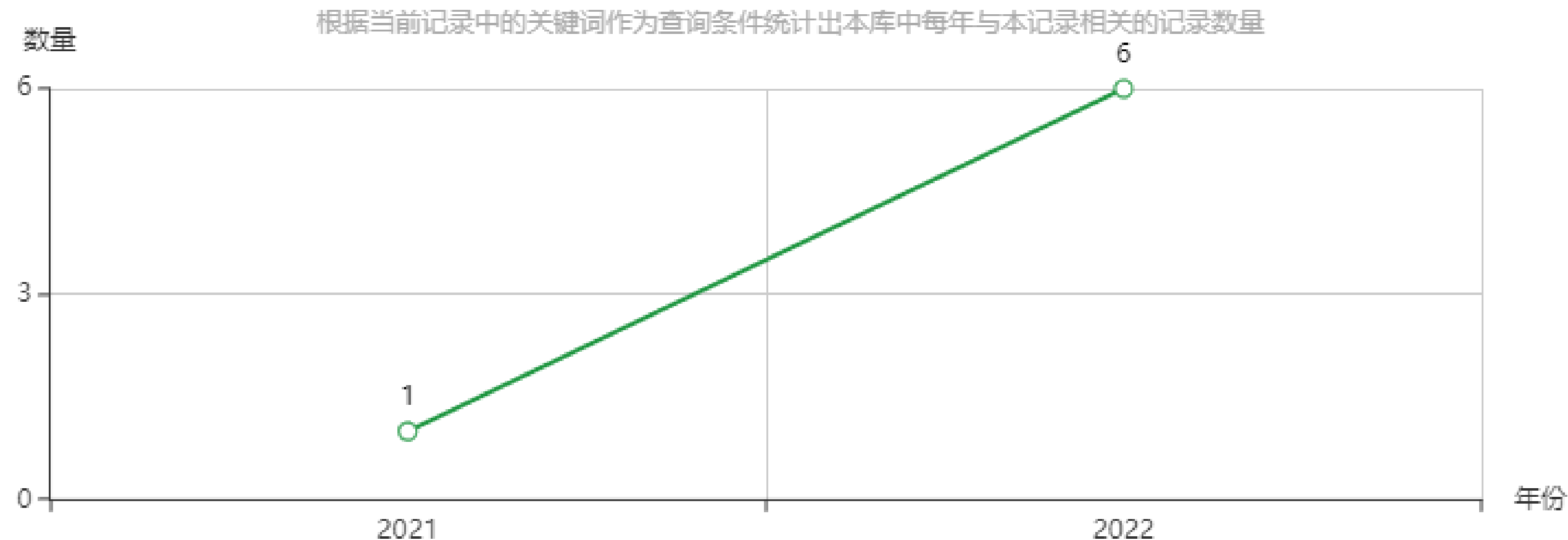
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