Engineering CRISPR/Cas9 to mitigate host contamination for 16S amplicon sequencing

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Video Byte

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Abstract

Microbes play essential roles in plant health for sustainable agriculture. A better understanding of plant microbiota could lead to strategies for regulating plant growth, enhancing stress tolerance, and reducing pathogenesis. High-throughput 16S rRNA gene sequencing is a useful method for studying bacterial community structures. Unfortunately, in plants, contamination of the 16S rRNA genes from the mitochondrion and plastid hinders sensitivity. Now, new research presents a novel method of reducing host contamination for plant microbiota profiling. The technique uses Cas9 and specific guide RNA to cut 16S rRNA targets, removing host contamination for 16S sequencing. In a feasibility test, Cas-16S-seq reduced the fraction of rice 16S rRNA gene sequences from 63.2% to 2.9% in roots and from 99.4% to 11.6% in leaf samples. Cas-16S-seq can reduce noise from host plants, increasing the yield of bacterial gene data without additional cost, and does not introduce a bias when profiling plant microbes, making it a useful new tool for plant microbiomics.