Hyper-active CRISPR-Cas9 variants for sensitive chemical detection through molecular recording
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Technology Summary

CRISPR-Cas (Clustered regularly interspaced short palindromic repeats- and their associated proteins) is a prokaryotic adaptive immune system that has generated intense interest as a gene-editing tool in synthetic biology. In the CRISPR immune response, Cas9 can associate with part of the integrase complex to record an invader’s (usually viral) foreign DNA, known as a spacer sequence, in between the repeats of the bacterial host’s CRISPR genomic locus. The spacer sequences are then processed into small RNAs that guide Cas nucleases to the invading DNA, thereby cutting and destroying the invader’s genome.

Our scientists have identified Cas9 point mutations that significantly promote the rate of spacer acquisition, or invader/viral DNA, into the bacterial host’s CRISPR locus. These highly ‘evolved’ Cas9 variants increase the frequency of foreign DNA integration into the host genome, making the integrated invader DNA easier to detect. When applied to synthetic biology technologies as molecular recording devices, they can potentially boost the detection sensitivity of low frequency events.

Application
Suitable as a molecular recording device in synthetic biology technologies for detecting low frequency events. Examples include:
- Trace (bio)chemical detection for industrial manufacturing processes in the food and beverage industry.
- Trace detection for environmentally-related molecules
- Diagnostic medical devices

Advantages
Enhances foreign DNA(spacer) acquisition rates by three fold

Stage of Development
A proof-of-concept demonstrates Staphylococcus aureus transformed with these Cas9 variants have 100-fold enhanced CRISPR adaptive immunity.

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Patent Information
Patent pending

Reference
- Heler et al. 2016. Mutations in Cas9 Enhance the Rate of Acquisition of Viral Spacer Sequences during the CRISPR-Cas Immune Response Molecular Cell 65:168-75
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