Docket #: S21-085

Using high-throughput single-strand DNA profiling for profiling CRISPR targeting of DNA sequences (CasKAS)

Stanford inventors have developed the CasKAS method for profiling CRISPR off-targets using single-stranded DNA (ssDNA) mapping. Binding of CRISPR protein to DNA generates ssDNA structures, which can be a sensitive biochemical signal of CRISPR occupancy. CasKAS uses recently developed KAS-seq assay for mapping ssDNA structures to identify DNA sequences that are associated with CRISPR proteins.

CRISPR technology has great potential in biomedical research and medical practices, yet the off-target effects of CRISPR can bring risks to patients' health in clinical applications. There are numerous approaches to experimentally map off-target effects. However, some of these methods involve combination of complex molecular biology protocols that prevent them being widely adopted, while other approaches suffer from background and specificity issues.

Unlike current methods, this new CasKAS method is fast and cheap. It only uses very simple molecular biology procedures, and it is highly accessible to labs with no high-level technological expertise. Moreover, CasKAS is applicable to all different types of DNA-targeting CRISPR proteins and it can be applied to primary non-dividing cells to profile both CRISPR occupancy and CRISPR cleavage.

Stage of development
Proof of concept

Applications

- CRISPR genome and epigenome editing
- Personalized therapy

Advantages

- Quick and cheap
- Simple procedures and more accessible
- Applicable to all types of DNA-targeting CRISPR proteins
- Applicable to primary non-dividing cells

Publications

- Marinov, G. K. et al. Direct profiling of genome-wide dCas9 and Cas9 specificity using ssDNA mapping (CasKAS). *Cold Spring Harbor Laboratory* (2021).

Patents

- Published Application: WO2022216555

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