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COMMENTS

Base editing technology

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The largest class of known human pathogenic mutations is point mutation, also known as single nucleotide polymorphism (SNP)[1]. Precise and efficient editing of these SNPs has been of great interest for the treatment of genetic disorders. Base editing is a genome editing method which can precisely induce point mutations on DNA or RNA at the target loci without generating double strand breaks[2–5]. DNA base editors are comprised of a catalytically impaired CRISPR-associated (Cas) nuclease fused with a base modification enzyme[2,4]. These modification enzymes are normally deaminases that operate only on single-stranded DNA (ssDNA) not double-stranded DNA. On pairing of single-guide RNA (sgRNA) and the target DNA sequence, a ssDNA bubble is generated and the DNA bases are modified by the deaminase enzyme[6]. There are two classes of DNA base editors. Cytosine base editors (CBEs) convert C·G base pairs to A·G base pairs[2] and adenine base editors (ABEs) conversely convert A·G base pairs to G·C base pairs[4]. RNA base editors have also been developed using Cas13 sgRNA-targeting methods[7].

Although base editors can generate point mutations at target loci, unanticipated edits are observed[8]. By fusing a second uracil DNA glycosylase inhibitor (UGI) to the C terminus of BE3, Komor et al.[9] found the product purity of BE3 could be greatly improved and indel frequency reduced. In addition to undesired edits at target sites, both CBEs and ABEs have the potential to induce off-target edits at the genome-wide level. Although early evidence suggested that off-target effects of base editing are rare[2,4,10], recent studies have revealed that substantial off-target edits were induced by CBEs[11,12]. These off-target edits were sgRNA-independent and probably induced by the deaminase[11,12]. As early studies only focus on a subset of loci that may experience off-target editing from Cas9 nuclease[10], which might explain why they did not find any off-target edits induced by CBEs. In addition, others have reported large amounts of RNA off-target effects induced by both CBEs and ABEs[13,14]. Researchers have also attempted to reduce off-target effects by engineering deaminases to provide improved base editors with high fidelity[13,14].

Genome editing tools with the capacity to efficiently and precisely correct pathological mutations in a genome are a robust technology that will undoubtedly impact all life sciences. Continued development of improved or newly generated base editing tools with higher efficiency and fidelity is needed to enhance the impact of this technology in the field.

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