
POPULAR ARTICLE

Key enzymes involved in CRISPR systems and their roles

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Manuscript received: December 5,2025; Decision on manuscript, December20,2024; Manuscript accepted: January12, 2025

The primary enzyme in CRISPR gene editing is Cas9, acting as "molecular scissors" guided by RNA to cut specific DNA sequences, but other Cas (CRISPR-associated) proteins like Cas12a (Cpf1) and Cas13 are also used, with Cas13 targeting RNA instead of DNA, offering versatility for different editing or detection tasks.

Key enzymes and their roles

Cas9: The most famous enzyme, it creates double-strand breaks in DNA at targeted locations, allowing scientists to insert, delete, or modify genes.

Cas12 (e.g., Cas12a): Another DNA-targeting enzyme that processes its own guide RNA and cuts double-stranded DNA, often used in diagnostics and offering different cutting patterns than Cas9.

Cas13: An RNA-targeting enzyme that cleaves RNA, useful for techniques like RNA knockdown and detection, not DNA editing.

Base Editors (e.g., ABE, CBE): Modified Cas9 enzymes (often nickases, which cut only one strand) fused with other enzymes like deaminases to change single DNA bases without creating double-strand breaks, as noted in this National Institutes of Health (NIH) article.

Prime editors (PE): A sophisticated system combining a Cas9 nickase with a reverse transcriptase and a special guide RNA (pegRNA) to directly write new genetic information, allowing for insertions, deletions, and all base changes.

How they work together

These enzymes work with specially designed guide RNAs (gRNAs) that tell the Cas enzyme where to go in the genome, essentially directing the "scissors" to the precise DNA or RNA sequence to be edited or detected, as explained on MedlinePlus and Your Genome.

The CRISPR system relies on several key enzymes (Cas proteins) to function as a form of adaptive immunity in bacteria and archaea, and as a powerful genome editing tool in molecular biology. The primary role of these Cas enzymes is to recognize and cleave target nucleic acids (DNA or RNA) as guided by a small RNA molecule.

In the table note that engineered variants, such as nickase Cas9 (nCas9) (which only cuts one DNA strand) and dead Cas9 (dCas9) (which binds but does not cut DNA), are used in applications like base editing and epigenome editing when fused with other enzymes like deaminases or reverse transcriptase.

Below are the key enzymes involved in CRISPR systems and their roles in tabular form

Enzyme Name	Role in the CRISPR System
Cas9	The most widely used endonuclease (a type of nuclease) that uses a single guide RNA (sgRNA) to locate and induce a double-strand break in target double-stranded DNA (dsDNA). It possesses RuvC and HNH nuclease domains, each cleaving one DNA strand.
Cas12a (Cpf1)	Another type V single-protein endonuclease that targets and cleaves dsDNA, often resulting in a staggered (sticky) cut rather than the blunt end produced by Cas9. It has a single RuvC nuclease domain.
Cas13	A type VI single-protein ribonuclease (RNase) that is guided by a crRNA to target and degrade specific single-stranded RNA (ssRNA) sequences. It also has a non-specific "collateral" RNase activity upon target binding.
Cas1 and Cas2	A complex of proteins essential for the adaptation stage (spacer acquisition), which involves the processing and integration of fragments of foreign DNA into the host's CRISPR array as new "spacer" segments.
Cas3	The signature protein for Type I systems, which has both helicase (unwinds DNA duplexes) and endonuclease activity to degrade targeted DNA.
Cas10	The signature protein for Type III systems, which functions as a multi-domain nuclease targeting both DNA and RNA, and also has cyclic oligoadenylate (cOA) synthesis activity for signal amplification.
RNase III	A host cell ribonuclease that works with Cas9 in the Type II system to process the precursor crRNA (pre-crRNA) into mature crRNAs for use in targeting.