# Gene Snipper™ Cas9 Nickase (H840A)

**CATALOG#:**

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|---|---|
| M1098-50 | 50 pmol (50 µl) |
| M1098-250 | 250 pmol (25 µl) |

**SOURCE:** Recombinant *Streptococcus pyogenes* Cas9 (CRISPR associated protein 9) Nickase (H840A) protein purified from *E. coli*

**MOLECULAR WEIGHT:** ~160 kDa

**PURITY:** The protein is at least 95% pure by SDS PAGE

**CONCENTRATION:**

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|---|---|
| M1098-50 | 1000 nM |
| M1098-250 | 10 µM |

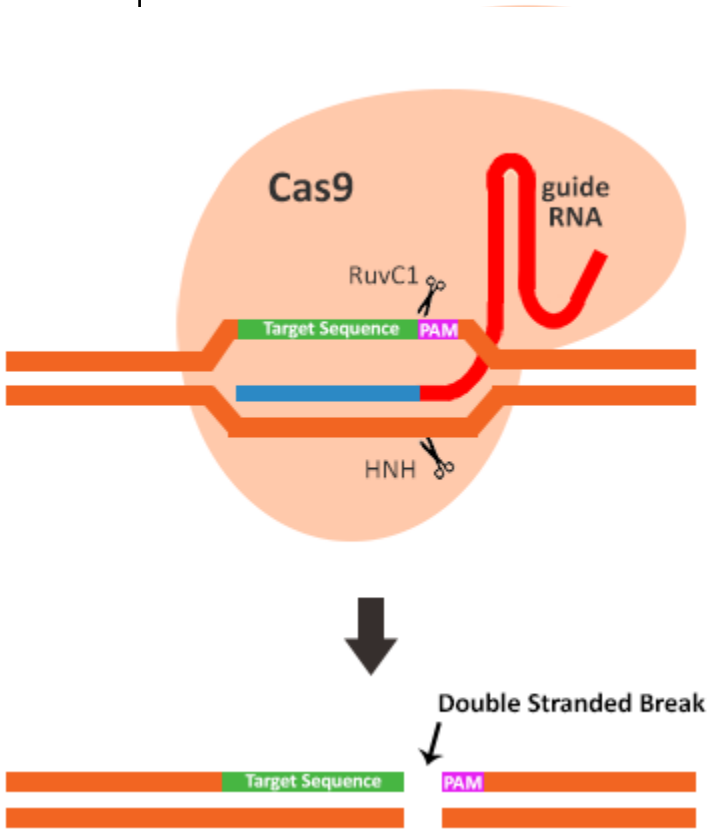
**FORM:** Colorless liquid. Enzyme supplied with 10X Reaction Buffer (200 mM HEPES, 50 mM $MgCl_2$, 1 M NaCl,1 mM EDTA, pH 6.5)

**RECONSTITUTION:** 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM DTT, 300 mM NaCl, and 50% (v/v) Glycerol

**STORAGE CONDITIONS:** Store all components at -20°C. Avoid repeated freeze/thaw cycles. All components are stable for 1 year from the date of shipping when stored and handled properly.

**DESCRIPTION:** Cas9 Nuclease Nickase H840A, *S. pyogenes*, is an RNA-guided endonuclease that catalyzes site specific nicking of a single strand of double stranded DNA. The Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR)/Cas9 system is the latest RNA-guided, endonuclease tool in genome editing which allows for very specific genomic disruption and replacement. One concern with the current CRISPR Cas9 technology is the potential off-target effects of the Cas9 nuclease activity. To improve the off-target mutagenic effects of this system, the Cas9 Nickase H840A Protein was developed with a H840A mutation in its HNH-like nuclease domain. This mutant form results in the generation of single strand nick instead of a double stranded break (DSB), like that generated by the Cas9 Nuclease, at the target site. Since a single strand break, or nick, is normally quickly repaired through the homology directed repair pathway using the intact complementary DNA strand as the repair template, off-target effects of the Cas9 Nickase is minimized. To utilize Cas9 Nickase for genome editing, two gRNAs, instead of one is required. The two gRNAs will be designed on opposite DNA strands but with close proximity to ensure that a DSB is induced once the two strands are nicked by the Cas9 Nickase. This paired Cas9 Nickase modification reduces off target effects because the two gRNAs need to work together to produce a DSB. Once the DSB is created, either the NHEJ or HDR pathway will be activated to complete the genome editing process. The Cas9 Nickase can also be used to create nucleotide modifications by homologous recombination if a repair template DNA containing the desired modification is introduced along with the gRNA and Cas9 Nickase.

**BIOLOGICAL ACTIVITY:** The activity of the protein in in vivo is confirmed by CRISPR Genome Cleavage Detection Kit.




***FOR RESEARCH USE ONLY! Not to be used on humans.***

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