ArciTect™ Cas9 Nickase

Modified Cas9 nuclease for the generation of single-strand breaks in CRISPR-Cas9 genome editing

Catalog #	76007	10 µg	1 µg/µL
	76008	50 µg	1 µg/µL
	76009	100 µg	4 µg/µL

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Product Description

ArciTect™ Cas9 Nickase is a modified version of the Cas9 recombinant protein from Streptococcus pyogenes. This version of Cas9 contains a deactivating D10A mutation, leaving only one active nuclease domain that introduces a single-strand break on the DNA strand complementary to the guide RNA (gRNA). ArciTect™ Cas9 Nickase requires association with a gRNA, composed of ArciTect™ tracrRNA (Catalog #76016) and ArciTect™ crRNA (Catalog #76010), to form a ribonucleoprotein (RNP) complex. Typically, two gRNAs are targeted to opposite DNA strands in the same region, and the two single-strand breaks create an overhanging double-strand break of defined length. This method enables the creation of small deletions for knockout models and further reduces off-target cleavage events for gene repair or insertions via homology-directed repair. ArciTect™ Cas9 Nickase also contains a nuclear localization signal at the N-terminus, ensuring that the RNP complex translocates to the nucleus, thereby increasing the efficiency of genome editing. As the RNP complex is fully functional upon transfection, it allows for immediate activity following translocation to the nucleus. The RNP complex is degraded over 48 hours, allowing sufficient time for genome editing to occur while reducing off-target effects that can be caused by the continuous presence of the RNP complex. Using the RNP system also circumvents the laborious process of generating stable Cas9-expressing cell lines, saving time and reducing the risk of off-target effects due to leaky inducible expression systems. The S. pyogenes Cas9 uses the protospacer adjacent motif (PAM) sequence NGG (where N can be any nucleotide). The enzyme will not cleave without a genomic PAM site downstream of the target sequence.

Properties

Storage: Store at -20°C.
Shelf Life: Stable for 30 months from date of manufacture (MFG) on label.
Formulation: 10 mM Tris buffer, 300 mM NaCl, 0.1 mM EDTA, 1 mM DTT, 50% glycerol, pH 7.4.
Molecular Weight: 160 kDa

Directions for Use

For complete instructions on CRISPR-Cas9 genome editing, refer to the Technical Bulletin: Human Pluripotent Stem Cell Genome Editing Using the ArciTect™ CRISPR-Cas9 System (Document #27084), available at www.stemcell.com or contact us to request a copy.

Related Products

For related products, including other genome editing tools, specialized cell culture and storage media, supplements, antibodies, cytokines, and small molecules, visit www.stemcell.com or contact us at techsupport@stemcell.com.

References

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Hultquist JF et al. (2016) A Cas9 ribonucleoprotein platform for functional genetic studies of HIV-host interactions in primary human T cells. Cell Rep 17(5): 1438–52.

Kim S et al. (2014) Highly efficient RNA-guided genome editing in human cells via delivery of purified Cas9 ribonucleoproteins. Genome Res 24(6): 1012–9.

Liang X et al. (2015) Rapid and highly efficient mammalian cell engineering via Cas9 protein transfection. J Biotechnol 208: 44–53.

Ran FA et al. (2013) Double nicking by RNA-guided CRISPR Cas9 for enhanced genome editing specificity. Cell 154(6): 1380–9.

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