SANTA CRUZ BIOTECHNOLOGY, INC.

Cre Vector: sc-418923

BACKGROUND

The Cre Vector expresses Cre recombinase, a bacteriophage p1 enzyme that catalyzes site-specific DNA recombination between two LoxP sites, and can be used in conjunction with the CRISPR/Cas9 Knockout (KO) and Homology-Directed Repair (HDR) plasmids (1,2,3). CRISPR/Cas9 KO Plasmids enable the identification and cleavage of a specific gene encoding a protein of interest by disrupting the gene in the genomic DNA (5,8). HDR Plasmids provide a specific DNA repair template for the double-strand break created by the CRISPR/Cas9 KO Plasmids (4). When the CRISPR/Cas9 KO Plasmid is co-transfected with the HDR Plasmid, the HDR Plasmid incorporates a selection marker where Cas9-induced DNA cleavage has occurred. Cells containing the edited DNA can be isolated using the selection marker inserted during homology-directed repair. Following selection, cells can be transfected with the Cre Vector to excise the selection marker, such as a puromycin resistance gene. Cre recombinase binds and catalyzes the two LoxP sites that flank the DNA sequence (6,8). The DNA fragment in between the two LoxP sites is excised as circular DNA and the genomic DNA is rejoined by DNA ligase, leaving a single LoxP site in the genomic DNA (7).

REFERENCES

1. Sauer, B and Henderson, N. 1988. Site-specific DNA recombination in mammalian cells by the Cre recombinase of bacteriophage P1. Proc. Natl. Acad. Sci. USA 85: 5166-5170.
2. Nagy, A. 2000. Cre recombinase: the universal reagent for genome tailoring. Genesis 26: 99-109.
3. Long, D.P., et al. 2012. Progress in Cre/Lox site-specific recombination systems in higher eukaryotes. Yi Chuan 34: 177-189.
4. Mali, P., et al. 2013. RNA-guided human genome engineering via Cas9. Science 339: 823-826.
5. Hsu, P., et al. 2014. Development and applications of CRISPR-Cas9 for genome editing. Cell 157: 1262-1278.
6. Ma, Y. 2014. Generation of eGFP and Cre knockin rats by CRISPR/Cas9. FEBS J. 281: 3779-3790.
7. Ma, Y., et al. 2014. Generating rats with conditional alleles using CRISPR/Cas9. Cell Res. 24: 122-125.
8. Xue, W. 2014. CRISPR-mediated direct mutation of cancer genes in the mouse liver. Nature. E-published.

RESEARCH USE

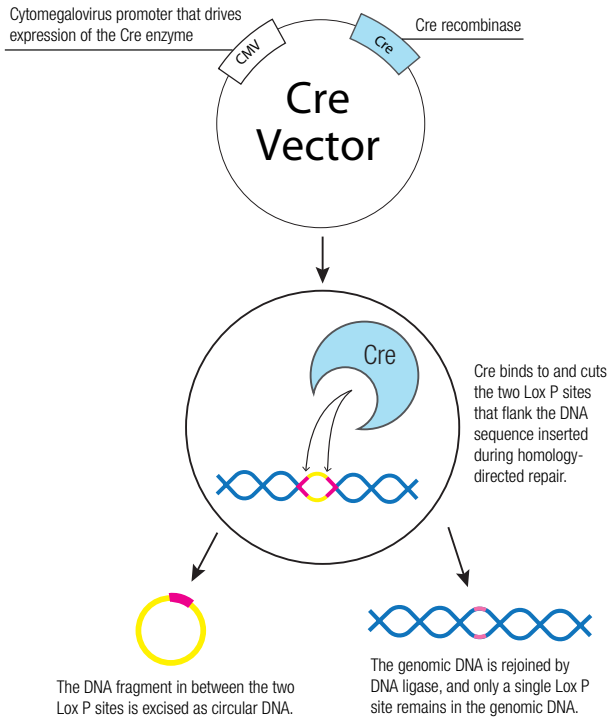
The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

PRODUCT

The Cre Vector is designed to specifically target the LoxP sites in DNA edited by the CRISPR/Cas9 KO Plasmid and the HDR Plasmid. Each vial contains 20 µg of lyophilized DNA plasmid. Suitable for up to 20 transfections.

APPLICATIONS

Cre Vector is recommended for the removal of genetic material flanked by LoxP sites.



SUPPORT REAGENTS

For optimal Cre Vector transfection efficiency, Santa Cruz Biotechnology's UltraCruz™ Transfection Reagent: sc-395739 (0.2 ml) and Plasmid Transfection Medium: sc-108062 (20 ml) are recommended.

STORAGE AND RESUSPENSION

Store lyophilized Cre Vector at 4° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C for long-term storage. Avoid repeated freeze thaw cycles.

Resuspend lyophilized Cre Vector in 200 µl of the provided ultrapure, sterile, DNase-free water. Resuspension of the plasmid DNA makes a 0.1 µg/µl solution in a 10 mM TRIS EDTA, 1 mM EDTA buffered solution.