SANTA CRUZ BIOTECHNOLOGY, INC.

Separase siRNA (m): sc-72041

BACKGROUND

Separase is a cysteine protease that triggers anaphase in all eukaryotes by participating in separation of sister chromatids during mitosis. Once activated, Separase hydrolyzes the SCC1 subunit of cohesin, the chromosomal protein complex responsible for sister chromatid cohesion. Separase and cohesin are highly conserved from yeasts to humans. When the cell is not dividing, Separase is prevented from cleaving cohesin through its association with another protein, securin. When anaphase is signaled, the securin is ubiquitinated and hydrolyzed by APC/cyclosome, releasing the active separase. Separase is transiently activated between the two meioses and may also be involved in homolog separation.

REFERENCES

1. Agarwal, R., et al. 2002. Mitotic regulation: the fine tuning of Separase activity. Cell Cycle 1: 255-257.
2. Zou, H., et al. 2002. Anaphase specific auto-cleavage of Separase. FEBS Lett. 528: 246-250.
3. Waizenegger, I., et al. 2002. Regulation of human Separase by securin binding and autocleavage. Curr. Biol. 12: 1368-1378.
4. Sullivan, M., et al. 2003. A non-proteolytic function of Separase links the onset of anaphase to mitotic exit. Nat. Cell Biol. 5: 249-254.
5. Chestukhin, A., et al. 2003. Processing, localization, and requirement of human Separase for normal anaphase progression. Proc. Natl. Acad. Sci. USA 100: 4574-4579.
6. Chestukhin, A., et al. 2003. Western blot screening for monoclonal antibodies against human Separase. J. Immunol. Methods 274: 105-113.

CHROMOSOMAL LOCATION

Genetic locus: Espl1 (mouse) mapping to 15 F3.

PRODUCT

Separase siRNA (m) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Separase shRNA Plasmid (m): sc-72041-SH and Separase shRNA (m) Lentiviral Particles: sc-72041-V as alternate gene silencing products.

For independent verification of Separase (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-72041A, sc-72041B and sc-72041C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNAses and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Separase siRNA (m) is recommended for the inhibition of Separase expression in mouse cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 µM in 66 µl. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Separase gene expression knockdown using RT-PCR Primer: Separase (m)-PR: sc-72041-PR (20 µl, 508 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.