

PABP siRNA (h2): sc-108012

BACKGROUND

PABP, for Poly(A)-binding protein, is an essential, well-conserved, multifunctional protein involved in translational initiation, mRNA biogenesis and degradation. PABP is required for the shortening of the 3' poly(A) tail of eukaryotic mRNA and translation initiation. The interaction between PABP and eukaryotic translation initiation factor 4G (eIF4G) facilitates translational initiation of polyadenylated mRNAs. This interaction is mediated, at least in part, by eIF4G, which bridges the mRNA termini by simultaneously binding PABP and the cap-binding protein, eIF4E. With lower affinities, PABP can also associate with non-poly(A) sequences. The physiological consequences of these PABP/RNA interactions are far from clear but may include functions such as translational silencing. PABP is a modular protein, with four N-terminal RNA-binding domains and an extensive C-terminus. During poliovirus infection, cleavage of eIF4GII and PABP have been proposed to contribute to complete host translation shutoff. The human PABP gene maps to chromosome 8q22.3 and encodes a 633 amino acid protein.

REFERENCES

1. Online Mendelian Inheritance in Man, OMIM™. 2000. Johns Hopkins University, Baltimore, MD. MIM Number: 604679. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
2. Chekanova, J.A., et al. 2001. Analysis of an essential requirement for the poly(A)-binding protein function using cross-species complementation. *Curr. Biol.* 11: 1207-1214.
3. Svitkin, Y.V., et al. 2001. Poly(A)-binding protein interaction with eIF4G stimulates picornavirus IRES-dependent translation. *RNA* 7: 1743-1752.
4. Deo, R.C., et al. 2001. X-ray structure of the human hyperplastic discs protein: an ortholog of the C-terminal domain of poly(A)-binding protein. *Proc. Natl. Acad. Sci. USA* 98: 4414-4419.
5. Mohr, E., et al. 2001. Vasopressin mRNA localization in nerve cells: characterization of *cis*-acting elements and *trans*-acting factors. *Proc. Natl. Acad. Sci. USA* 98: 7072-7079.
6. Wakiyama, M. and Miura, K. 2002. Inhibition of translation and progesterone-induced maturation of *Xenopus* oocytes by expressing the amino-terminal portion of the eukaryotic translation initiation factor 4G. *Biosci. Biotechnol. Biochem.* 66: 185-187.

CHROMOSOMAL LOCATION

Genetic locus: PABPC1 (human) mapping to 8q22.3.

PRODUCT

PABP siRNA (h2) 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PABP shRNA Plasmid (h2): sc-108012-SH and PABP shRNA (h2) Lentiviral Particles: sc-108012-V as alternate gene silencing products.

For independent verification of PABP (h2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-108012A, sc-108012B and sc-108012C.

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 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

PABP siRNA (h2) is recommended for the inhibition of PABP expression in human 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 μ M in 66 μ l. 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.

GENE EXPRESSION MONITORING

PABP (10E10): sc-32318 is recommended as a control antibody for monitoring of PABP gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor PABP gene expression knockdown using RT-PCR Primer: PABP (h2)-PR: sc-108012-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Eisermann, K., et al. 2015. Poly(A) binding protein cytoplasmic 1 is a novel co-regulator of the androgen receptor. *PLoS ONE* 10: e0128495.

RESEARCH USE

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