



## RYBP siRNA (m): sc-77379

### BACKGROUND

The products of the Polycomb group (PcG) of genes are necessary for the maintenance of transcriptional repression of a number of important developmental genes, including the homeotic genes. RYBP is a member of the mammalian polycomb complex. RYBP (RING1- and YY1-binding protein) interacts specifically with the E2F2 and E2F3 family members, dependent on the marked box domain in these proteins. YY1 and RYBP, in combination with either E2F2 or E2F3, can stimulate Cdc6 promoter activity synergistically, at G<sub>1</sub>/S of the cell cycle. RYBP also complexes with both RING1 proteins (RING1 and RING1B) and with M33, two mutually interacting sets of proteins of the mammalian Polycomb complex. RING1 binds RYBP and M33 through the same C-terminal domain, whereas the RYBP-M33 interaction takes place through an M33 domain not involved in binding. RYBP is widely expressed with highest levels in lymphoid tissues and placenta.

### REFERENCES

1. García, E., et al. 1999. RYBP, a new repressor protein that interacts with components of the mammalian Polycomb complex, and with the transcription factor YY1. *EMBO J.* 18: 3404-3418.
2. Zheng, L., et al. 2001. The death effector domain-associated factor plays distinct regulatory roles in the nucleus and cytoplasm. *J. Biol. Chem.* 276: 31945-31952.
3. Sawa, C., et al. 2002. YEA1/RYPB and YAF2 are functionally distinct members of a cofactor family for the YY1 and E4TF1/hGABP transcription factors. *J. Biol. Chem.* 277: 22484-22490.
4. Schlisio, S., et al. 2002. Interaction of YY1 with E2Fs, mediated by RYBP, provides a mechanism for specificity of E2F function. *EMBO J.* 21: 5775-5786.

### CHROMOSOMAL LOCATION

Genetic locus: Rybp (mouse) mapping to 6 D3.

### PRODUCT

RYBP siRNA (m) is a pool of 2 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see RYBP shRNA Plasmid (m): sc-77379-SH and RYBP shRNA (m) Lentiviral Particles: sc-77379-V as alternate gene silencing products.

For independent verification of RYBP (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-77379A and sc-77379B.

### RESEARCH USE

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

### PROTOCOLS

See our web site at [www.scbt.com](http://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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

### APPLICATIONS

RYBP siRNA (m) is recommended for the inhibition of RYBP 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  $\mu$ M in 66  $\mu$ 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

RYBP (A-1): sc-374235 is recommended as a control antibody for monitoring of RYBP 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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  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 $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  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 RYBP gene expression knockdown using RT-PCR Primer: RYBP (m)-PR: sc-77379-PR (20  $\mu$ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.