

paxillin siRNA (m): sc-36197

BACKGROUND

Paxillin is a focal adhesion phosphoprotein that is localized to the cytoskeleton. Phosphorylation of paxillin has been shown to occur in response to PDGF treatment, v-src transformation or cross-linking of integrins. FAK (focal adhesion kinase) and PYK2 have been shown to phosphorylate paxillin. FAK phosphorylates paxillin specifically on Tyr 118 *in vitro*. However, FAK phosphorylation does not seem to be required for the recruitment of paxillin to cell adhesion sites. Paxillin may play a role in signal transduction, regulation of cell morphology and the recruitment of structural and signaling molecules to focal adhesions. It has been shown that the amount of paxillin is reduced in mitotic cells by proteolytic downregulation and that paxillin is alternatively phosphorylated on serine rather than on tyrosine and serine during mitosis.

REFERENCES

1. Graham, I.L., et al. 1994. Complement receptor 3 (CR3, Mac-1, integrin $\alpha M\beta 2$, CD11b/CD18) is required for tyrosine phosphorylation of paxillin in adherent and nonadherent neutrophils. *J. Cell Biol.* 127: 1139-1147.
2. Salgia, R., et al. 1995. Molecular cloning of human paxillin, a focal adhesion protein phosphorylated by P210^{BCR/ABL}. *J. Biol. Chem.* 270: 5039-5047.
3. Bellis, S.L., et al. 1995. Characterization of tyrosine phosphorylation of paxillin *in vitro* by focal adhesion kinase. *J. Biol. Chem.* 270: 17437-17441.
4. Brown, M.C., et al. 1996. Identification of LIM3 as the principal determinant of paxillin focal adhesion localization and characterization of a novel motif on paxillin directing vinculin and focal adhesion kinase binding. *J. Cell Biol.* 135: 1109-1123.

CHROMOSOMAL LOCATION

Genetic locus: Pxn (mouse) mapping to 5 F.

PRODUCT

paxillin 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see paxillin shRNA Plasmid (m): sc-36197-SH and paxillin shRNA (m) Lentiviral Particles: sc-36197-V as alternate gene silencing products.

For independent verification of paxillin (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-36197A, sc-36197B and sc-36197C.

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

paxillin siRNA (m) is recommended for the inhibition of paxillin 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 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

paxillin (B-2): sc-365379 is recommended as a control antibody for monitoring of paxillin 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 paxillin gene expression knockdown using RT-PCR Primer: paxillin (m)-PR: sc-36197-PR (20 μ l, 443 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Velasco-Velázquez, M.A., et al. 2008. Reduced paxillin expression contributes to the antimetastatic effect of 4-hydroxycoumarin on B16-F10 melanoma cells. *Cancer Cell Int.* 8: 8.
2. Sharifi, M.N., et al. 2016. Autophagy promotes focal adhesion disassembly and cell motility of metastatic tumor cells through the direct interaction of paxillin with LC3. *Cell Rep.* 15: 1660-1672.
3. Murrey, M.W., et al. 2020. Adhesion, motility and matrix-degrading gene expression changes in CSF-1-induced mouse macrophage differentiation. *J. Cell Sci.* 133: jcs232405.

RESEARCH USE

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