SANTA CRUZ BIOTECHNOLOGY, INC.

p-PKR (Thr 451): sc-16815

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The Power to Question

BACKGROUND

An interferon-inducible, RNA-dependent protein serine/threonine kinase, PKR has various designations. Mouse PKR is known as DAI, dsJ, PI kinase, p65, p67 or TIK, whereas human PKR is known as p68 or p69. PKR phosphorylates its substrate, a subunit of protein synthesis initiation factor eIF-2, on Ser 51 to inhibit translation. PKR contains two dsRNA binding motifs required for its activation by dsRNA. Three kinds of regulation of PKR enzymatic activity occur, and these include transcriptional regulation in response to interferon, an autoregulatory mechanism controlling PKR expression at the level of translation, and post-translational regulation by RNA mediated autophosphorylation. Human PKR contains at least 15 autophosphorylation sites, but only Thr 446 and Thr 451 in the activation loop are critical for its kinase activity. Thr 446 is the *in vivo* autophosphorylation site of PKR. Mutation of threonine to alanine at position 446 substantially reduces PKR function, and mutant kinase containing Ala 451 is completely inactive.

REFERENCES

1. Hershey, J.W.B. 1989. Protein phosphorylation controls translation rates. J. Biol. Chem. 264: 20823-20826.
2. Samuel, C.E. 1993. The eIF-2α protein kinases, regulators of translation in eukaryotes from yeasts to humans. J. Biol. Chem. 268: 7603-7606.
3. Tanaka, H. and Samuel, C.E. 1994. Mechanism of interferon action: structure of the mouse PKR gene encoding the interferon-inducible RNA-dependent protein kinase. Proc. Natl. Acad. Sci. USA 91: 7995-7999.
4. Romano, P.R., Garcia-Barrio, M.T., Zhang, X., Wang, Q., Taylor, D.R., Zhang, F., Herring, C., Mathews, M.B., Qin, J. and Hinnebusch A.G. 1998. Autophosphorylation in the activation loop is required for full kinase activity *in vivo* of human and yeast eIF-2α kinases PKR and GCN2. Mol. Cell. Biol. 18: 2282-2297.
5. Zhang, F., Romano, P.R., Nagamura-Inoue, T., Tian, B., Dever, T.E., Mathews, M.B., Ozato, K. and Hinnebusch, A.G. 2001. Binding of double-stranded RNA to protein kinase PKR is required for dimerization and promotes critical autophosphorylation events in the activation loop. J. Biol. Chem. 276: 24946-24958.

CHROMOSOMAL LOCATION

Genetic locus: EIF2AK2 (human) mapping to 2p22.2.

SOURCE

p-PKR (Thr 451) is available as either goat (sc-16815) or rabbit (sc-16815-R) polyclonal affinity purified antibody raised against a short amino acid sequence containing Thr 451 phosphorylated PKR of human origin.

PRODUCT

Each vial contains 200 µg IgG in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

Blocking peptide available for competition studies, sc-16815 P, (100 µg peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% BSA).

APPLICATIONS

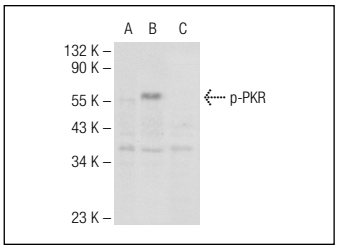
p-PKR (Thr 451) is recommended for detection of Thr 451 phosphorylated PKR of human origin by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000), immunoprecipitation [1-2 µg per 100-500 µg of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

Suitable for use as control antibody for PKR siRNA (h): sc-36263, PKR shRNA Plasmid (h): sc-36263-SH and PKR shRNA (h) Lentiviral Particles: sc-36263-V.

Molecular Weight of p-PKR: 68 kDa.

Positive Controls: HeLa + Calyculin A cell lysate: sc-2271 or HeLa + IFN-α + Calyculin A cell lysate: sc-24684.

DATA



p-PKR (Thr 451)-R: sc-16815-R. Western blot analysis of PKR phosphorylation in untreated (**A**), calyculin A treated (**B**) and calyclulin A and lambda protein phosphatase (sc-200312A) treated (**C**) HeLa whole cell lysates.

SELECT PRODUCT CITATIONS

1. Steinart, S., Kroll, T.C., Taubert, I., Pusch, L., Hortschansky, P., Höffken, K., Wölfl, S. and Clement, J.H. 2008. Differential expression of cancer-related genes by single and permanent exposure to bone morphogenetic protein 2. J. Cancer Res. Clin. Oncol. 134: 1237-1245.
2. Pataer, A., Swisher, S.G., Roth, J.A., Logothetis, C.J. and Corn, P.G. 2009. Inhibition of RNA-dependent protein kinase (PKR) leads to cancer cell death and increases chemosensitivity. Cancer Biol. Ther. 8: 245-252.
3. Pataer, A., Raso, M.G., Correa, A.M., Behrens, C., Tsuta, K., Solis, L., Fang, B., Roth, J.A., Wistuba, I.I. and Swisher, S.G. 2010. Prognostic significance of RNA-dependent protein kinase on non-small cell lung cancer patients. Clin. Cancer Res. 16: 5522-5528.

STORAGE

Store at 4° C, **DO NOT FREEZE**. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

RESEARCH USE

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