SANTA CRUZ BIOTECHNOLOGY, INC.

p-PKR (Thr 446): sc-16565

SANTA CRUZ BIOTECHNOLOGY

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. 1989. Protein phosphorylation controls translation rates. J. Biol. Chem. 264: 20823-20826.
2. Samuel, C.E. 1993. The eIF2α protein kinases, regulators of translation in eukaryotes from yeasts to humans. J. Biol. Chem. 268: 7603-7606.

CHROMOSOMAL LOCATION

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

SOURCE

p-PKR (Thr 446) is available as either goat (sc-16565) or rabbit (sc-16565-R) polyclonal affinity purified antibody raised against a short amino acid sequence containing Thr 446 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-16565 P, (100 µg peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% BSA).

APPLICATIONS

p-PKR (Thr 446) is recommended for detection of Thr 446 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), immunohistochemistry (including paraffin-embedded sections) (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.

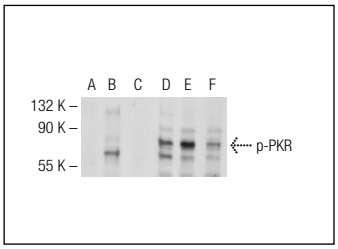
Molecular Weight of p-PKR: 68 kDa.

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

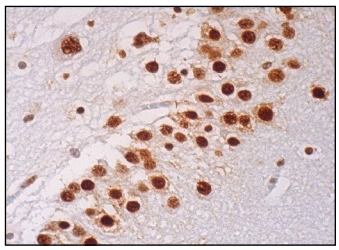
STORAGE

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

DATA



Western blot analysis of PKR phosphorylation in untreated (**A**,**D**), calyculin treated (**B**,**E**) and calyculin and lambda protein phosphatase (sc-200312A) treated (**C**,**F**) HeLa whole cell lysates. Antibodies tested include p-PKR (Thr 446)-R: sc-16565-R (**A**,**B**,**C**) and PKR (M-515): sc-1702 (**D**,**E**,**F**).



p-PKR (Thr 446): sc-16565. Immunoperoxidase staining of formalin fixed, paraffin-embedded human hippocampus tissue showing nuclear staining of neuronal cells and glial cells.

SELECT PRODUCT CITATIONS

1. Dreikhausen, U., et al. 2005. NFκB-repressing factor inhibits elongation of human immunodeficiency virus type 1 transcription by DRB sensitivity-inducing factor. Mol. Cell. Biol. 25: 7473-7483.
2. Zhang, P. and Samuel, C.E. 2008. Induction of protein kinase PKR-dependent activation of interferon regulatory factor 3 by vaccinia virus occurs through adapter IPS-1 signaling. J. Biol. Chem. 283: 34580-34587.
3. Guan, Z., et al. 2010. Interaction of Hsp40 with influenza virus M2 protein: implications for PKR signaling pathway. Protein Cell 1: 944-955.
4. Paquet, C., et al. 2012. The PKR activator PACT is induced by Aβ: involvement in Alzheimer's disease. Brain Pathol. 22: 219-229.
5. Liu, L., et al. 2012. Influenza A virus induces interleukin-27 through cyclooxygenase-2 and protein kinase A signaling. J. Biol. Chem. 287: 11899-11910.
6. Weissbach, R. and Scadden, A.D. 2012. Tudor-SN and ADAR1 are components of cytoplasmic stress granules. RNA 18: 462-471.

RESEARCH USE

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

PROTOCOLS

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