Anti-VRK1 (human) antibody, mouse monoclonal (5D1)

71-600 100 μg

Function; The VRK1 gene encodes serine/threonine kinase VRK1 (Vaccinia-Related Kinase 1; 396 aa, 45.5 kDa) which is involved in Golgi disassembly during the cell cycle following phosphorylation by PLK3 during mitosis, and required to induce Golgi fragmentation. It acts by mediating phosphorylation of a downstream target protein 'Thr-18' of p53/TP53 and may thereby prevent the interaction between p53/TP53 and MDM2. It also phosphorylates casein and histone H3. Phosphorylation of the BANF1 gene product disrupts its ability to bind DNA, reduces its binding to LEM domain-containing proteins and causes its relocalization from the nucleus to the cytoplasm.

Involvement in disease Defects in VRK1 are the cause of pontocerebellar hypoplasia type 1A (PCH1A); also called pontocerebellar hypoplasia with infantile spinal muscular atrophy or pontocerebellar hypoplasia with anterior horn cell disease. PCH1A is characterized by an abnormally small cerebellum and brainstem, central and peripheral motor dysfunction from birth, gliosis and anterior horn cell degeneration resembling infantile spinal muscular atrophy

Applications

1. Western blotting (1/200~1/1,000 dilution). Use of highly sensitive chemiluminescence reagents such as Lumi-Light Plus (Roche) or ImmunoStar®LD (Wako, Tokyo) are recommended.
2. Immunoprecipitation (assay dependent)
3. Immunofluorescence staining (1/100 dilution)
4. Immunohistochemistry (assay dependent)
5. ELISA (assay dependent)

Reactivity; Human VRK1 protein. Not tested with other species.

Immunogen ; Synthetic peptide corresponding to N-terminus of human VRK1, MPRVKAAQAGRQSSAKRHL-C

Product; Mouse monoclonal antibody (5D1) produced in serum-free medium and purified by propriety chromatography under mild conditions (90~98% pure).

Isotype; IgG1 kappa

Form; 1 mg/ml in PBS with 50% glycerol, filter-sterilized.

Database links; SwissProt: Q99986 Human ,
Entrez Gene: 7443 Human

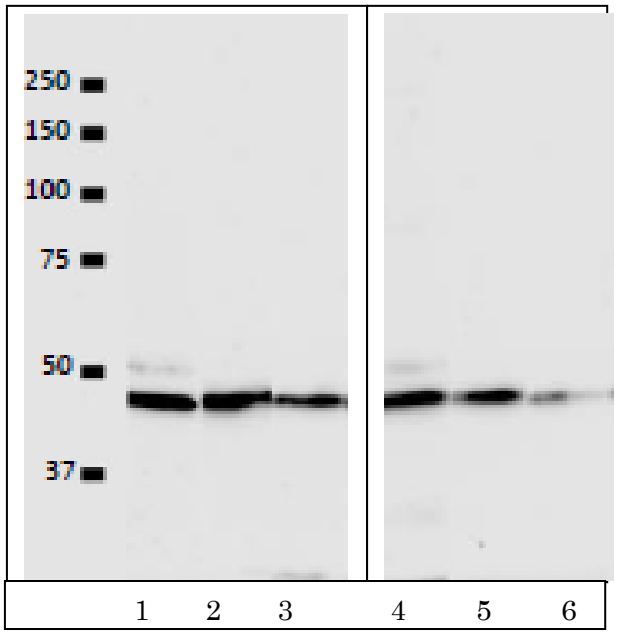

Fig.1. Western blot detection of VRK1 in the crude extracts of human cells. Lanes 1, 2, 3; HeLa cell extract (5x10^4 cells) with antibody dilutions at 1/100, 1/500, 1/1000. Lanes 4, 5, 6; U2OS cell extract (5x10^4 cells) with the antibody dilutions at 1/100, 1/500, 1/1,000. As secondary antibody, Alexa488 goat anti-mouse IgG was used. ImmunoStar ® LD (Wako, Tokyo) was used as chemiluminescence reagent and images were taken with BIO-RAD ChemiDocXRS.

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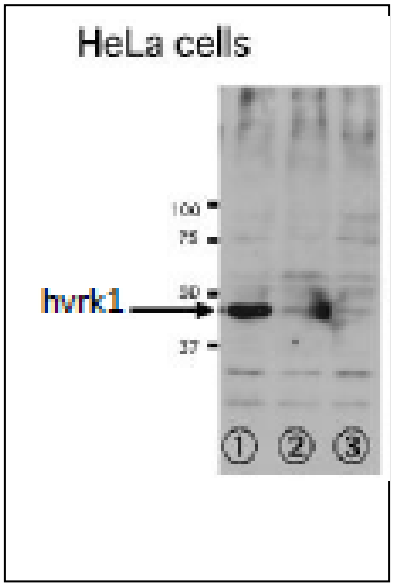

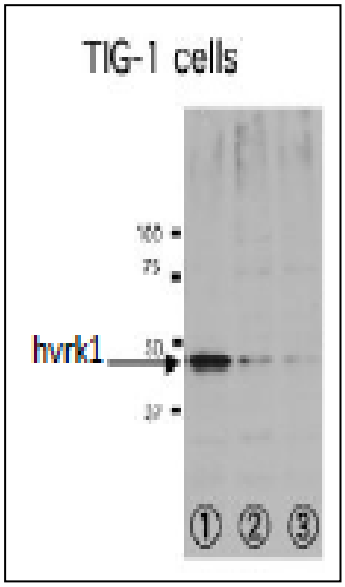

Fig.2. Inhibition of VRK1 expression in human cells treated by RNAi. specific to VRK1.

Lane 1; Luciferase RNAi (control). Lane2; VRK1-1 RNAi. Lane 3; VRK1-2 RNAi.. Antibody at 1/500 dilution. Lumi-Light Plus (Roche) was used as chemiluminescence reagent. Extracts from 5x10^4 cells.

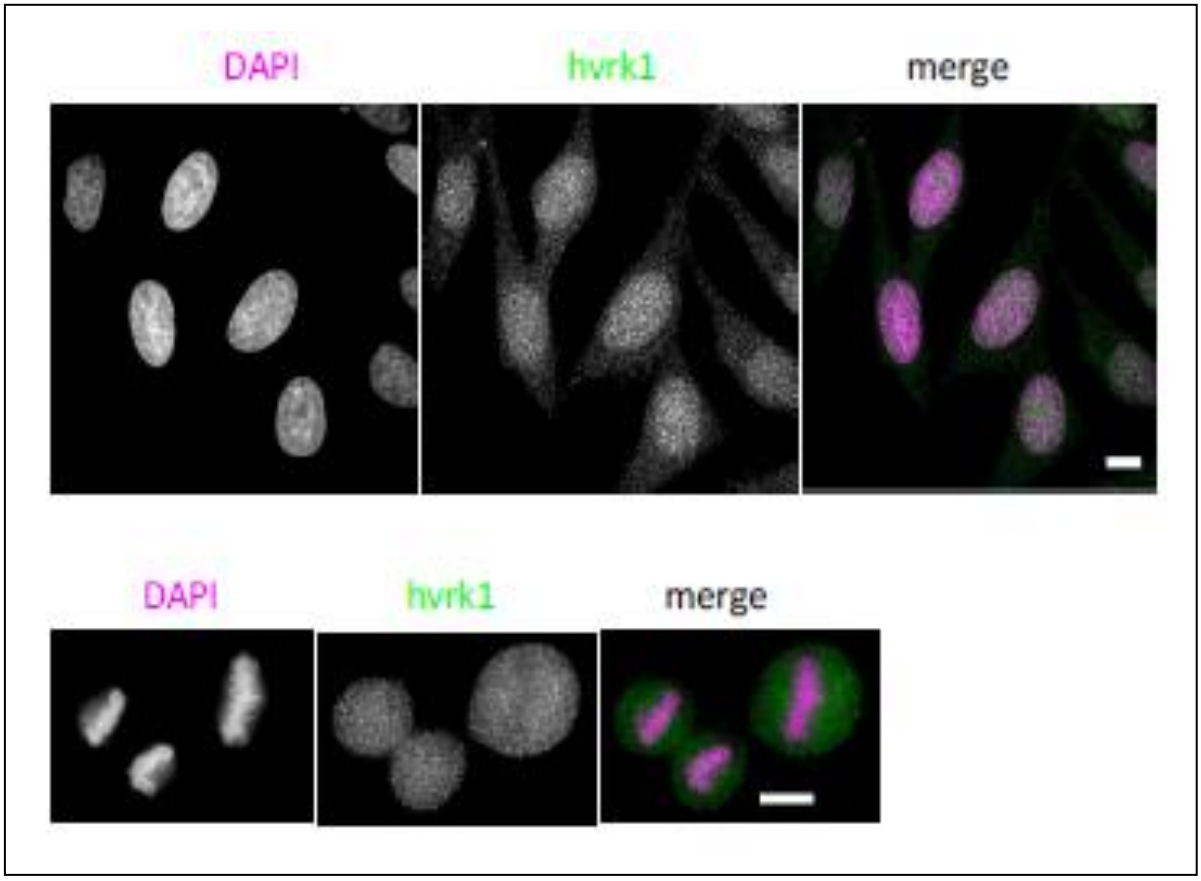

Fig.3. Immunoflorescence staining of VRK1 in HeLa cells, PA fixed.

Top; Interphase cells were fixed with paraformaldehyde and stained with the anti-human VRK1 antibody (hvrk1) at 1/100 dilution (center), DNA was stained with DAPI (left) and two images were merged (right; merge).

Bottom; Metaphase cells. At metaphase, VRK1 dots were solely detected in nuclei.

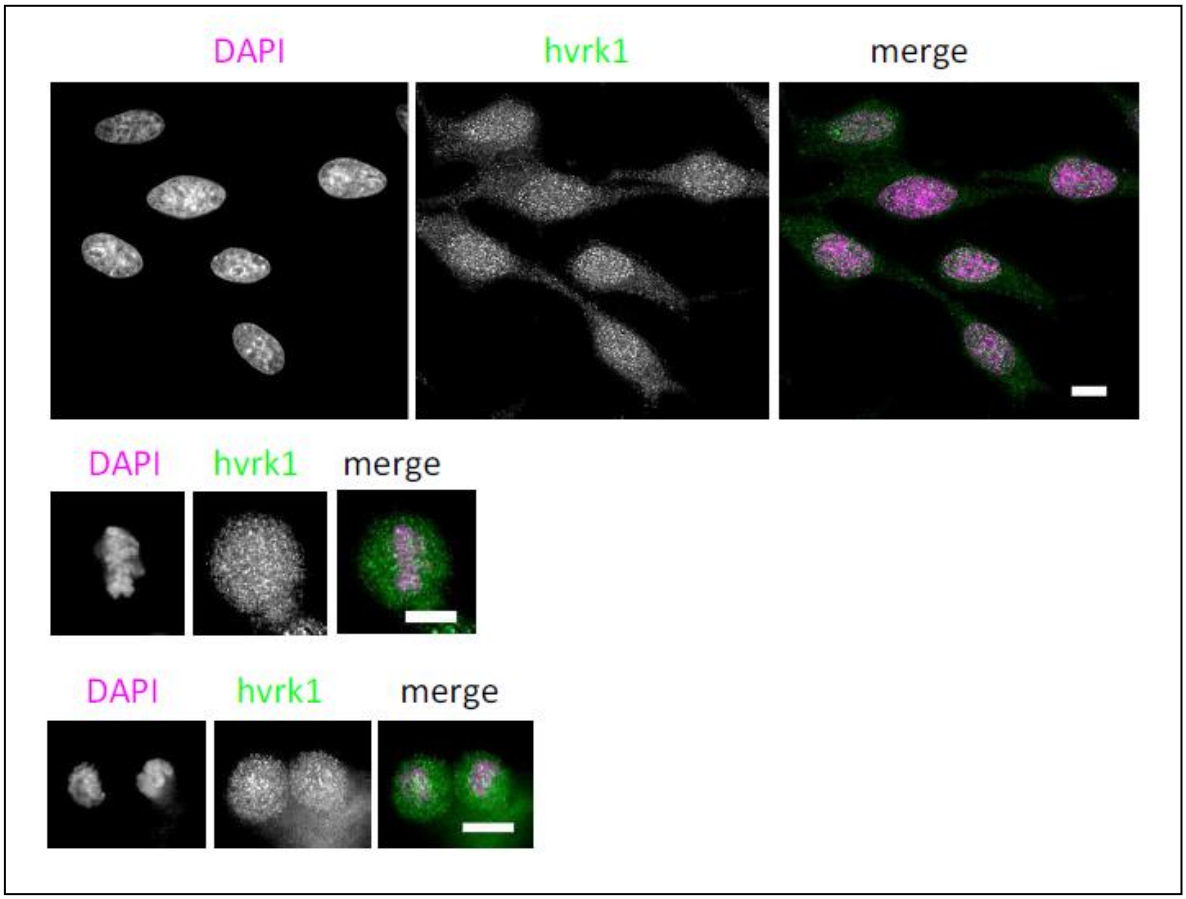

Fig.4. Immunoflorescence staining of VRK1 in HeLa cells, methanol fixed.

Top; Interphase cells were fixed with methanol and stained with the anti-human VRK1 antibody (hvrk1) at 1/100 dilution (center), DNA was stained with DAPI (left) and two images were merged (right; merge).

Center and bottom; Metaphase cells. At metaphase, VRK1 dots were solely detected in nuclei.

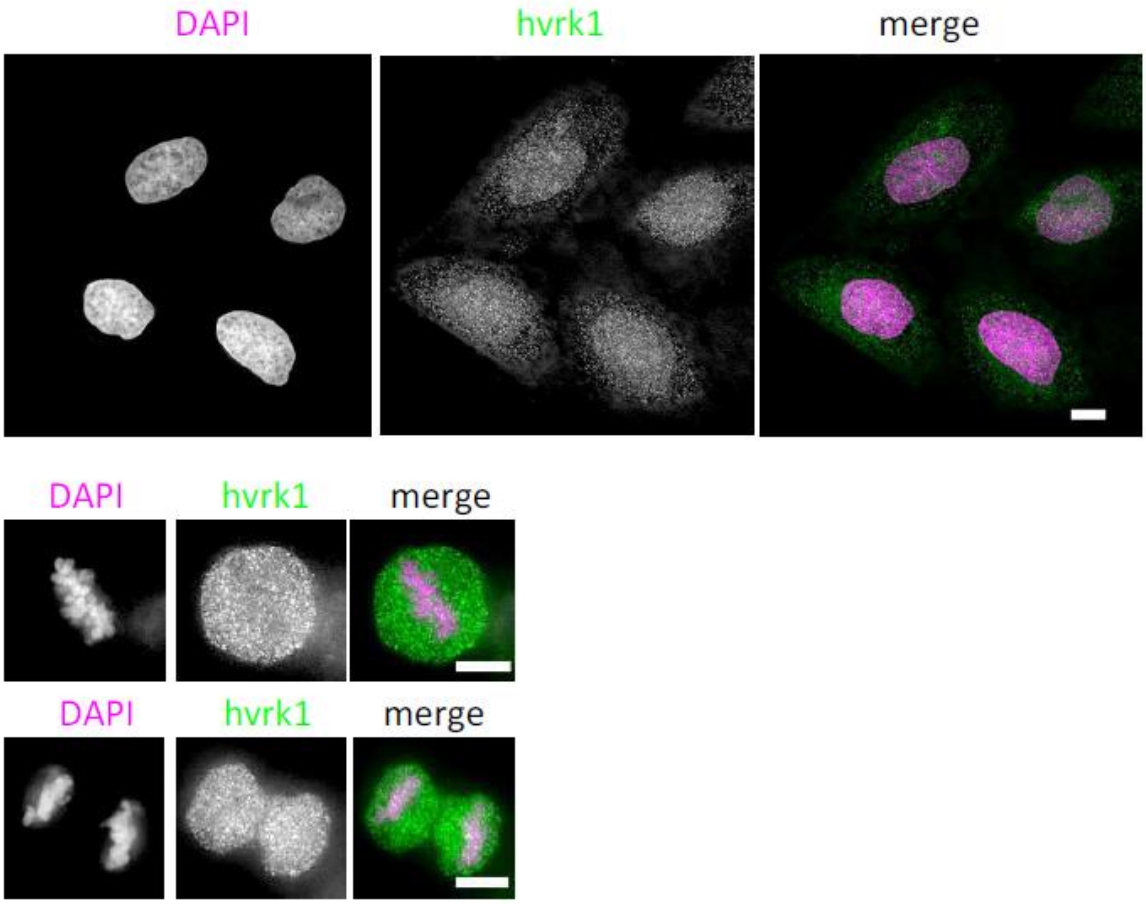

Fig. 5. Immunoflorescence staining of VRK1 in U-2 OS cells, formaldehyde fixed.

Top; Interphase cells were fixed with formaldehyde and stained with the anti-human VRK1 antibody (hvrk1) at 1/100 dilution (center), DNA was stained with DAPI (left) and two images were merged (right; merge).

Center and bottom; Metaphase cells. At metaphase, VRK1 dots were solely detected in nuclei.

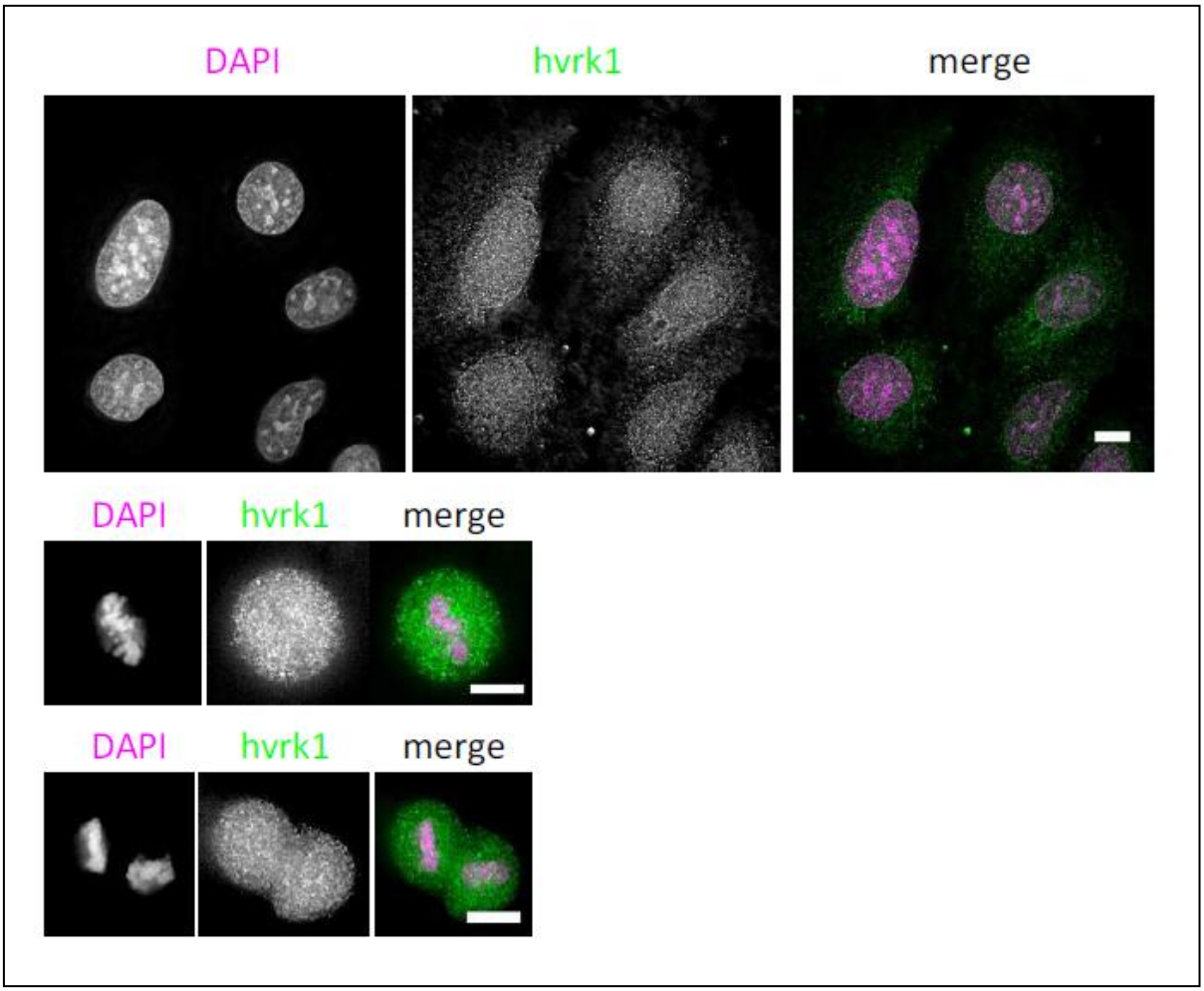

Fig. 5. Immunoflorescence staining of VRK1 in U-2 OS cells, methanol fixed.

Top; Interphase cells were fixed with methanol and stained with the anti-human VRK1 antibody (hvrk1) at 1/100 dilution (center), DNA was stained with DAPI (left) and two images were merged (right; merge).

Center and bottom; Metaphase cells. At metaphase, VRK1 dots were solely detected in nuclei.