Product Datasheet

PKN3 Antibody - BSA Free
NBP1-30102

Unit Size: 100 ul

Store at 4C. Do not freeze.

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Reviews: 2 Publications: 6

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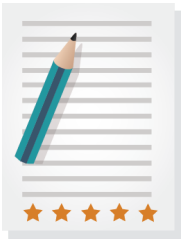
Updated 2/21/2025 v.20.1



NBP1-30102

PKN3 Antibody - BSA Free

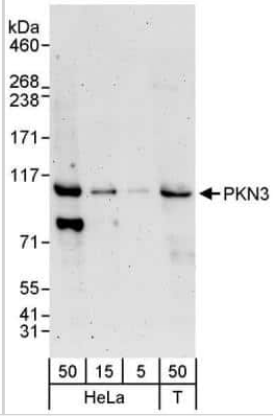
Product Information	
Unit Size	100 ul
Concentration	1.0 mg/ml
Storage	Store at 4C. Do not freeze.
Clonality	Polyclonal
Preservative	0.09% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	Tris-Citrate/Phosphate (pH 7.0 - 8.0)
Target Molecular Weight	99 kDa

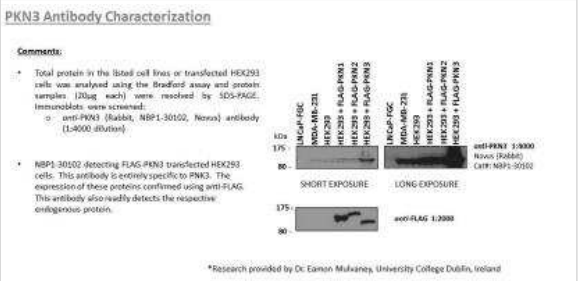
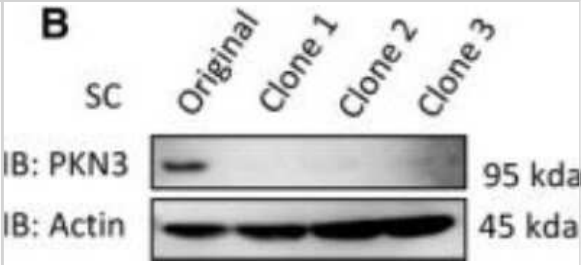
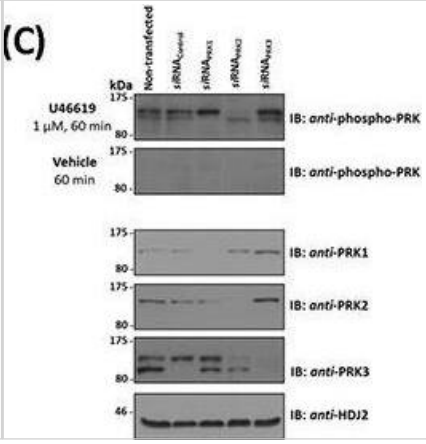
Product Description	
Host	Rabbit
Gene ID	29941
Gene Symbol	PKN3
Species	Human, Mouse
Reactivity Notes	Mouse reactivity reported in scientific literature (PMID: 30422386).
Immunogen	maps to a region between residue 225 and 275 of human protein kinase N3 using the numbering given in entry NP_037487.2

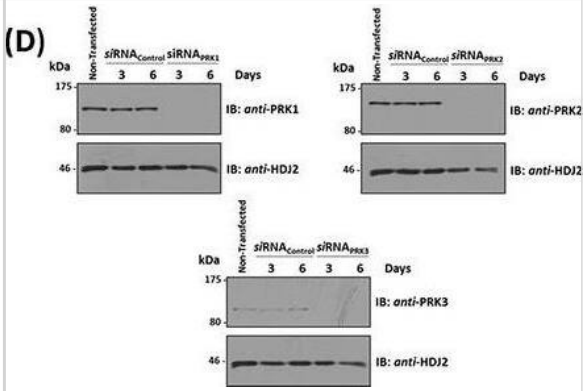
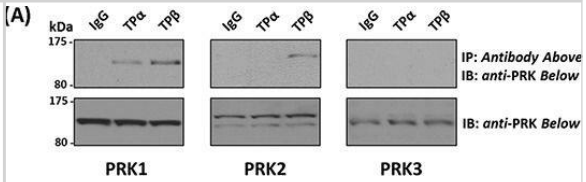
Product Application Details	
Applications	Western Blot, Immunoprecipitation (Negative), Knockout Validated
Recommended Dilutions	Western Blot 1:2000-1:10000, Immunoprecipitation (Negative), Knockout Validated Reactivity validation reported in (PMID: 30518210)

Images

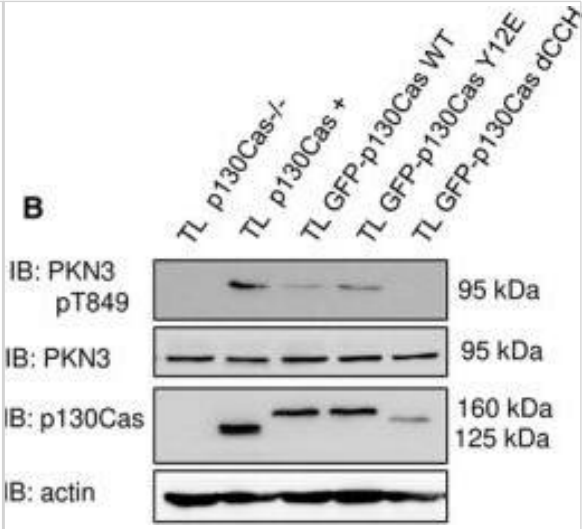
Western Blot: PKN3 Antibody [NBP1-30102] - Whole cell lysate from HeLa (5, 15 and 50 ug) and 293T (T; 50 ug) cells. Antibody used at 0.1 ug/ml.



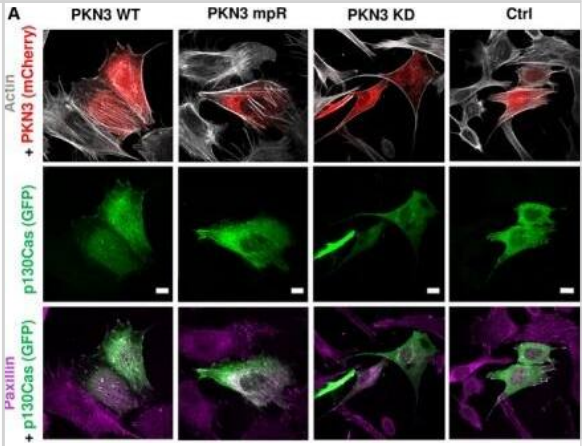
Western Blot: PKN3 Antibody [NBP1-30102] - Image submitted by a verified customer review.	
Western Blot: PKN3 Antibody [NBP1-30102] - PKN3 regulates growth in 2D and in 3D environment of Src-transformed MEFs through interaction with p130CaS. Immunoblot of SC cells and SC cells with PKN3 gene inactivated using CRISPR/CAS9 (SCpkn3-/-). Inactivation of PKN3 expression is visualized by antibody anti-PKN3. Antibody anti-actin was used as loading control. Image collected and cropped by CiteAb from the following publication (https://onlinelibrary.wiley.com/doi/abs/10.1002/1878-0261.12401), licensed under a CC-BY license.	
Western Blot: PKN3 Antibody [NBP1-30102] - Influence of TP Agonist on the Activation of PRK1, PRK2 & PRK3. Panel C. In order to identify the species of PRK subject to U46619-induced T-loop phosphorylation, PC-3 cells initially transfected for 72 hr with 30 nM siRNAPRK1, siRNAPRK2, siRNAPRK3 or, as controls, with a scrambled siRNAControl. Thereafter, cells serum starved & stimulated with U46619 for 60 min or with vehicle & then immunoblotted (20 µg/lane) with anti-phospho-PRK1Thr774/PRK2Thr816/PRK3Thr718 & with anti-PRK1, anti-PRK2, anti-PRK3 or, as loading controls, HDJ2 antisera, as indicated. Image collected & cropped by CiteAb from the following publication (https://www.oncotarget.com/lookup/doi/10.18632/oncotarget.4664), licensed under a CC-BY license. Not internally tested by Novus Biologicals.	

Western Blot: PKN3 Antibody [NBP1-30102] - Effect of siRNA-mediated disruption of PRK1, PRK2 & PRK3 expression on TP agonist-induced proliferation & migration of PC-3 cellsPanels A-D. PC-3 cells were transfected for 48 hr with 30 nM siRNAPRK1, siRNAPRK2, siRNAPRK3 or, as controls, with a scrambled siRNA (siRNAControl) prior to stimulation with U46619 (10 nM) or vehicle (0.0001% EtOH) for the indicated time specific to the assay, where non-transfected cells served as a reference. Panel A: For analysis of proliferation, 72 hr post-siRNA transfection PC-3 cells were stimulated for 48 hr with U46619 (10 nM) or vehicle (0.0001% EtOH). Panel B: For analysis of colony formation, 48 hr post-siRNA transfection PC-3 cells were stimulated with U46619 (10 nM) or vehicle (0.0001% EtOH) in soft agar & assessed 10 day post-treatment for colony formation. Panel C: For analysis of migration, 72 hr post-siRNA transfection PC-3 cells were stimulated for 8 hr with U46619 (10 nM) or vehicle (0.0001% EtOH). In Panels A-C, the bar charts show mean relative levels of PC-3 cell proliferation, colony formation & migration (± SEM, n ≥ 3), where levels in the vehicle-treated cells are assigned a value of 100%. The asterisks indicate where U46619-stimulation resulted in significant increases in proliferation, colony formation or migration by PC-3 cells compared to vehicle-treated cells, where ** & *** indicates p < 0.01 & p < 0.001, respectively. Panel D: Validation of the specificity & sustained siRNA-mediated disruption of PRK1, PRK2 & PRK3 expression was confirmed by immunoblot analysis of whole cell lysates (20 µg/lane) with the respective anti-PRK1/2/3 or, to validate protein loading, with anti-HDJ2 antisera where analysis was carried out 3 & 6 day post-transfection. Data n ≥ 3. Image collected & cropped by CiteAb from the following publication (https://www.oncotarget.com/lookup/doi/10.18632/oncotarget.4664), licensed under a CC-BY license. Not internally tested by Novus Biologicals.	
Western Blot: PKN3 Antibody [NBP1-30102] - Association of PRK1 & PRK2 with TPα & TPβ in prostate PC-3 cellsPanel A. PC-3 cells were immunoprecipitated with anti-TPα, anti-TPβ or, as controls, with the pre-immune (IgG) sera. Thereafter, immunoprecipitates (upper panels) or equivalent aliquots of whole cell lysates (20 µg/lane, lower panels) were immunoblotted (IB) with anti-PRK1, anti-PRK2 or anti-PRK3 antisera. The relative positions of the molecular size markers (kDa) are indicated to the left of the panels. Data shown are representative of at least three independent experiments (n ≥ 3). Panel B. PC-3 cells were incubated with U46619 (1 µM; 0–60 min) prior to immunoprecipitation with anti-TPα, anti-TPβ or, as controls, with the pre-immune (IgG) sera. Thereafter, immunoprecipitates (upper panels) or equivalent aliquots of whole cell lysates (20 µg/lane, lower panels) were IB with anti-PRK1, anti-PRK2 or anti-PRK3 antisera. Data n ≥ 3. Panel C. Bar charts show the mean relative levels of PRK1 or PRK2 associated with the anti-TPα or anti-TPβ immunoprecipitates, as determined by quantitative densitometry (± SEM), where levels associated with the respective immunoprecipitates in the absence of agonist are expressed as 1. The asterisks indicate where U46619 stimulation resulted in significant changes in complex-associated PRK1 or PRK2, where * & ** indicate p < 0.05 & p < 0.01, respectively. Image collected & cropped by CiteAb from the following publication (https://www.oncotarget.com/lookup/doi/10.18632/oncotarget.4664), licensed under a CC-BY license. Not internally tested by Novus Biologicals.	

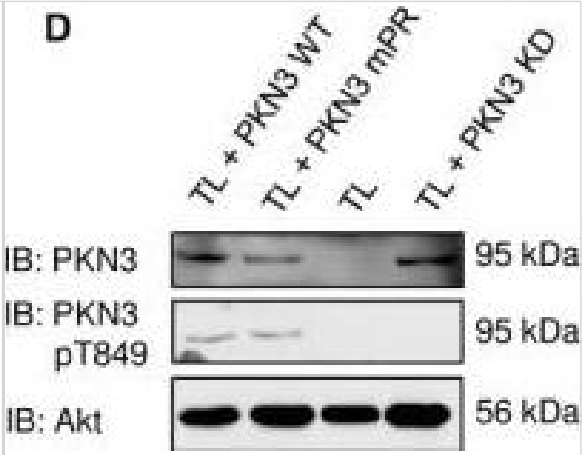
Western Blot: PKN3 Antibody [NBP1-30102] - PKN3 activity is important for stress fibers formation & is stimulated by the expression of p130Cas. (A) p130Cas−/−MEFs growing on FN□coated cover slips were co□transfected by GFP□p130Cas & mCherry□PKN3 fusion variant (WT, mPR, KD) or mCherry. After 48 h, cells were fixed & imaged by Leica TCS SP2 microscope (63×/1.45 oil objective). Stress fibers were visualized by Phalloidin (405) & focal adhesions by anti□Paxillin staining (2nd 633). Representative images are shown. Scale bars represent 20 µm. (B) p130Cas−/−MEFs or p130Cas−/−MEFs re□expressing p130Cas or transfected by GFP□fused p130Cas variants (WT, YE, dCCH) were lysed in RIPA buffer, blotted to nitrocellulose membrane, & analyzed for endogenous PKN3 activity by antibody anti□phosphoThr849 of PKN3 (pT849 PKN3). Expression of p130Cas mutants was verified by anti□p130Cas antibody & loading by anti□PKN3 & anti□actin antibody. (C) Densitometric quantification of PKN3 activity (pT849 PKN3 phosphorylation). The effect of p130Cas re□expression on PKN3 T849 phosphorylation was analyzed separately from the effect of transfected p130Cas mutants (indicated by a dotted line). Error bars indicate means ± SD from three independent experiments (four experiments for the left part). Statistical significance was evaluated by one□way repeated ANOVA followed by Turkey's post hoc test (*P < 0.05; **P < 0.01). (D) Lysates or (E) immunoprecipitates (by Flag sepharose) from p130Cas−/−MEFs re□expressing p130Cas & overexpressing PKN3 variants (WT, mPR, KD) were immunoblotted by anti□PKN3, anti□pT849 PKN3, & anti□Akt antibodies (loading control). Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/30422386), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



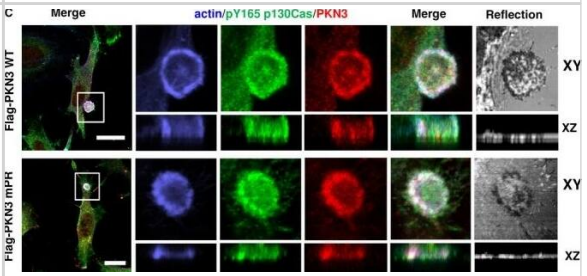
Immunocytochemistry/ Immunofluorescence: PKN3 Antibody [NBP1-30102] - PKN3 activity is important for stress fibers formation & is stimulated by the expression of p130Cas. (A) p130Cas−/−MEFs growing on FN□coated cover slips were co□transfected by GFP□p130Cas & mCherry□PKN3 fusion variant (WT, mPR, KD) or mCherry. After 48 h, cells were fixed & imaged by Leica TCS SP2 microscope (63×/1.45 oil objective). Stress fibers were visualized by Phalloidin (405) & focal adhesions by anti□Paxillin staining (2nd 633). Representative images are shown. Scale bars represent 20 µm. (B) p130Cas−/−MEFs or p130Cas−/−MEFs re□expressing p130Cas or transfected by GFP□fused p130Cas variants (WT, YE, dCCH) were lysed in RIPA buffer, blotted to nitrocellulose membrane, & analyzed for endogenous PKN3 activity by antibody anti□phosphoThr849 of PKN3 (pT849 PKN3). Expression of p130Cas mutants was verified by anti□p130Cas antibody & loading by anti□PKN3 & anti□actin antibody. (C) Densitometric quantification of PKN3 activity (pT849 PKN3 phosphorylation). The effect of p130Cas re□expression on PKN3 T849 phosphorylation was analyzed separately from the effect of transfected p130Cas mutants (indicated by a dotted line). Error bars indicate means ± SD from three independent experiments (four experiments for the left part). Statistical significance was evaluated by one□way repeated ANOVA followed by Turkey's post hoc test (*P < 0.05; **P < 0.01). (D) Lysates or (E) immunoprecipitates (by Flag sepharose) from p130Cas−/−MEFs re□expressing p130Cas & overexpressing PKN3 variants (WT, mPR, KD) were immunoblotted by anti□PKN3, anti□pT849 PKN3, & anti□Akt antibodies (loading control). Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/30422386), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



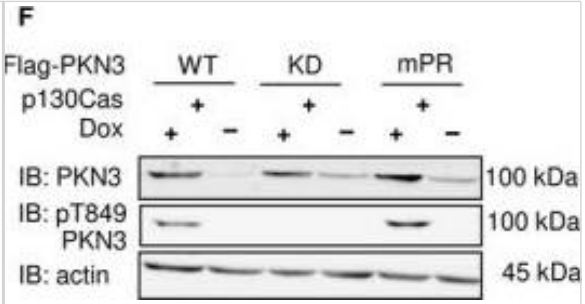
Western Blot: PKN3 Antibody [NBP1-30102] - PKN3 activity is important for stress fibers formation & is stimulated by the expression of p130Cas. (A) p130Cas−/−MEFs growing on FN□coated cover slips were co□transfected by GFP□p130Cas & mCherry□PKN3 fusion variant (WT, mPR, KD) or mCherry. After 48 h, cells were fixed & imaged by Leica TCS SP2 microscope (63×/1.45 oil objective). Stress fibers were visualized by Phalloidin (405) & focal adhesions by anti□Paxillin staining (2nd 633). Representative images are shown. Scale bars represent 20 µm. (B) p130Cas−/−MEFs or p130Cas−/−MEFs re□expressing p130Cas or transfected by GFP□fused p130Cas variants (WT, YE, dCCH) were lysed in RIPA buffer, blotted to nitrocellulose membrane, & analyzed for endogenous PKN3 activity by antibody anti□phosphoThr849 of PKN3 (pT849 PKN3). Expression of p130Cas mutants was verified by anti□p130Cas antibody & loading by anti□PKN3 & anti□actin antibody. (C) Densitometric quantification of PKN3 activity (pT849 PKN3 phosphorylation). The effect of p130Cas re□expression on PKN3 T849 phosphorylation was analyzed separately from the effect of transfected p130Cas mutants (indicated by a dotted line). Error bars indicate means ± SD from three independent experiments (four experiments for the left part). Statistical significance was evaluated by one□way repeated ANOVA followed by Turkey's post hoc test (*P < 0.05; **P < 0.01). (D) Lysates or (E) immunoprecipitates (by Flag sepharose) from p130Cas−/−MEFs re□expressing p130Cas & overexpressing PKN3 variants (WT, mPR, KD) were immunoblotted by anti□PKN3, anti□pT849 PKN3, & anti□Akt antibodies (loading control). Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/30422386), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Immunocytochemistry/ Immunofluorescence: PKN3 Antibody [NBP1-30102] - PKN3 colocalizes with p130Cas in lamellipodia & podosome rosettes. Representative images are shown. (A) p130Cas−/−MEFs plated on fibronectin (FN) were transfected by GFP□p130Cas, CFP□LifeAct, & mCherry□PKN3WT or mCherry□PKN3 mPR & imaged live 24 h after transfection. White arrow indicates lamellipodia. Histogram of dotted straight line is shown. (B) Quantification of mCherry□PKN3 WT, mCherry□PKN3 mPR, & mCherry localization to lamellipodia (LifeAct as marker) was calculated as described in methods (values are mean ± SD from three independent experiments, n > 50 measurements – 3 per cell; ***P < 0.001, one□way ANOVA on ranks followed by Dunn's post hoc test). (C) Src□transformed p130Cas−/−MEFs co□expressing p130Cas (SC) & mouse Flag tagged PKN3 WT or Flag□PKN3 mPR are shown. Cells were grown on FN□coated coverslips for 48 h, fixed, & stained for p130Cas by anti□pTyr165 p130Cas antibody (pY165 p130Cas; 2nd 405), for actin by Phalloidin 488 & for Flag□PKN3 by anti□Flag antibody (2nd 633). Reflection (670 nm) indicates fibronectin degradation. All scale bars represent 20 µm. Cell were imaged by Leica TCS SP8 microscope system equipped with Leica 63×/1.45 oil objective. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/30422386), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Western Blot: PKN3 Antibody [NBP1-30102] - PKN3 overexpression regulates growth of MEFs, & this effect requires PKN3–p130Cas interaction. (A) Immunoblotted lysates from MEFs p130Cas−/− or MEFs p130Cas−/− re□expressing p130Cas (p130Cas+) treated by Doxycycline (Dox) to induce expression of mCherry□PKN3 or mCherry alone. p130Cas presence detected by ani□p130Cas antibody & mCherry epitope by anti□mCherry antibody. (B) Dynamics of mCherry□PKN3 expression after supplementation w/ Dox shown by immunoblot w/ anti□mCherry antibody. (C–E) Effect of induced mCherry□PKN3 expression on cell growth. Representative graphs showing growth of MEFs p130Cas−/− re□expressing p130Cas (p130Cas+) (C) or MEFs p130Cas−/− (D) measured in real□time using xCELLigence RTCA (real□time cell analysis) system instrument. (E) Quantification of cell growth change induced by mCherry□PKN3 expression ('−' indicates inducible mCherry expression used as negative control). Slope ratios reflecting cell growth calculated from log growth phase of cell growth (indicated by dotted lines; see C & D). (F) Immunoblotted lysates from MEFs p130Cas−/− re□expressing p130Cas (p130Cas+) treated or not treated by Dox which induced expression of Flag□fused PKN3 variants (WT, mPR, KD, empty vector). Stimulated overexpression of PKN3 detected by anti□PKN3 antibody & its activity by antibody anti□pT849 PKN3. (G) Quantification of cell growth change stimulated by Dox□inducible expression of Flag□fused PKN3 variants (WT, mPR, KD) in MEFs p130Cas−/− re□expressing p130Cas (p130Cas +). All error bars indicate means ± SD calculated from 3 to 5 independent experiments (each in triplicates). Statistical significance always calculated between induced & noninduced cells & evaluated by one□way repeated ANOVA followed by Turkey post hoc test (***P < 0.001). Image collected & cropped by CiteAb from following publication (https://pubmed.ncbi.nlm.nih.gov/30422386), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Publications

Aine G O'Sullivan, Eamon P Mulvaney, B Therese Kinsella Regulation of protein kinase C-related kinase (PRK) signalling by the TPα and TPβ isoforms of the human thromboxane A 2 receptor: Implications for thromboxane- and androgen- dependent neoplastic and epigenetic responses in prostate cancer. Biochimica et biophysica acta. Molecular basis of disease 2018-11-19 [PMID: 28108419]

Arang N, Lubrano S, Ceribelli M et al. High-throughput chemogenetic drug screening reveals PKC-RhoA/PKN as a targetable signaling vulnerability in GNAQ-driven uveal melanoma Cell reports. Medicine 2023-10-13 [PMID: 37858338] (WB, Human)

Browne CM, Jiang B, Ficarro SB et al. A Chemoproteomic Strategy for Direct and Proteome-Wide Covalent Inhibitor Target-Site Identification J. Am. Chem. Soc. 2019-01-09 [PMID: 30518210] (KO, WB, Human)

Gemperle J, Dibus M, Koudelkova L et al. The interaction of p130Cas with PKN3 promotes malignant growth Mol Oncol. [PMID: 30422386] (WB, Mouse)

O'Sullivan AG, Mulvaney EP, Hyland PB, Kinsella BT. Protein kinase C-related kinase 1 and 2 play an essential role in thromboxane-mediated neoplastic responses in prostate cancer. Oncotarget 2015-09-22 [PMID: 26296974]

Collazos A, Michael N, Whelan RD et al. Site recognition and substrate screens for PKN family proteins. Biochem J;438(3):535-43. 2011-09-15 [PMID: 21749319]

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NBP2-33444PEP	PKN3 Recombinant Protein Antigen

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