abcam

ab104836

Anti-Hexokinase II antibody [3D3]

Key facts

Isotype	IgG1
Host species	Mouse
Storage buffer	Preservative: 0.03% Sodium azide Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	Recombinant Fragment Protein within Human HK2. The exact immunogen used to generate this antibody is proprietary information. Database link P52789
Clone number	3D3
Purification technique	Affinity purification Protein G
Concentration	1 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →
Purification notes	Purified from tissue culture supernatant.

Reactivity data

Flow Cyt

Tested

Species	Human
Dilution info	-
Notes	ab170190 - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.

WB

Tested

Species	Human
Dilution info	1/500 - 1/2000
Notes	-

IHC-P

Tested

Species	Human
Dilution info	1/200 - 1/1000
Notes	-

Target data

See full target information HK2

Function	Catalyzes the phosphorylation of hexose, such as D-glucose and D-fructose, to hexose 6-phosphate (D-glucose 6-phosphate and D-fructose 6-phosphate, respectively) (PubMed:23185017, PubMed:26985301, PubMed:29298880). Mediates the initial step of glycolysis by catalyzing phosphorylation of D-glucose to D-glucose 6-phosphate (PubMed:29298880). Plays a key role in maintaining the integrity of the outer mitochondrial membrane by preventing the release of apoptogenic molecules from the intermembrane space and subsequent apoptosis (PubMed:18350175).

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

We do not recommend this combination. It is not covered by our product promise.

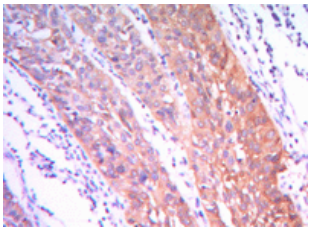
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

---

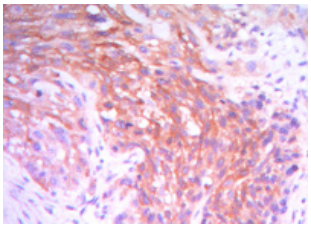
9 product images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Hexokinase II antibody [3D3] (ab104836)

Hexokinase II Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using mouse Anti-Hexokinase II antibody

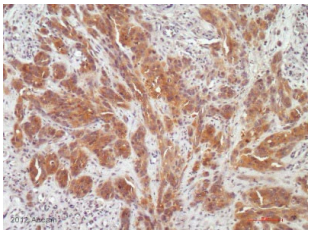
ab104836 at 1/200 dilution staining Hexokinase II in human esophagus cancer by Immunohistochemistry, Paraffin-embedded tissue with DAB staining.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Hexokinase II antibody [3D3] (ab104836)

Hexokinase II Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using mouse Anti-Hexokinase II antibody

ab104836 at 1/200 dilution staining Hexokinase II in human lung cancer by Immunohistochemistry, Paraffin-embedded tissue with DAB staining.



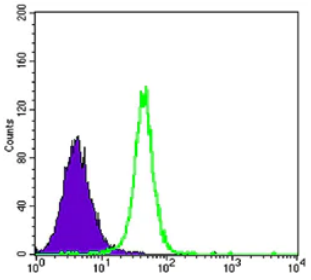
This image is courtesy of a customer review submitted by Ms Hadas Pahima

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Hexokinase II antibody [3D3] (ab104836)

Hexokinase II Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using mouse Anti-Hexokinase II antibody

Immunohistochemical analysis of human skin tissue, staining Hexokinase II with ab104836.

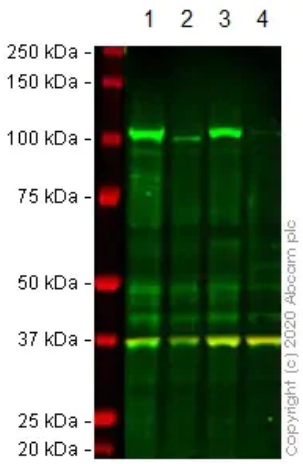
Tissue was fixed with paraformaldehyde and blocked with 10% BSA for 2 hours at room temperature; antigen retrieval was by heat mediation in citrate buffer. Samples were incubated with primary antibody (1/250 in 5% NGS + 10% BSA in PBS) for 16 hours at 4°C. An HRP-conjugated goat anti-mouse polyclonal IgG (1/250) was used as the secondary antibody.



Flow Cytometry - Anti-Hexokinase II antibody [3D3] (ab104836)

Hexokinase II Flow Cytometry staining using mouse Anti-Hexokinase II antibody

ab104836 staining Hexokinase II in K562 cells by Immunofluorescence (green) and negative control (purple).



Western blot - Anti-Hexokinase II antibody [3D3] (ab104836)

Hexokinase II Western blot staining using mouse Anti-Hexokinase II antibody

Lanes 1 - 4: Merged signal (red and green). Green - ab104836 observed at 102 kDa. Red - loading control ab181602 (Rabbit Anti-GAPDH antibody [EPR16891]) observed at 37kDa.
ab104836 was shown to react with Hexokinase II in wild-type HEK-293T cells in western blot. The band observed in HK2 CRISPR/Cas9 edited sample lane below 102 kDa may represent truncated forms and cleaved fragments. This has not been investigated further. Wild-type and HK2 CRISPR/Cas9 edited HEK-293T cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with ab104836 and ab181602 (Rabbit Anti-GAPDH antibody [EPR16891]) overnight at 4°C at a 1 in 500 Dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed (ab216772) and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed (ab216777) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-Hexokinase II antibody [3D3] (ab104836) at 1/500 dilution

Lane 1:
Wild-type HEK-293T cell lysate at 20 µg

Lane 2:
HK2 CRISPR/Cas9 edited HEK-293T cell lysate at 20 µg

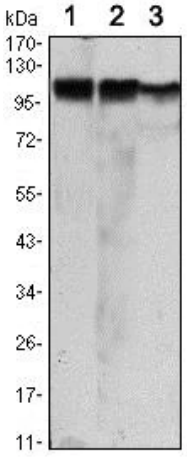
Lane 3:
HeLa cell lysate at 20 µg

Lane 4:
A549 cell lysate at 20 µg

Performed under reducing conditions.

Predicted band size: 102 kDa

Observed band size: 102 kDa



Western blot - Anti-Hexokinase II antibody [3D3] (ab104836)

All lanes:
Western blot - Anti-Hexokinase II antibody [3D3] (ab104836) at 1/500 dilution

Lane 1:
Jurkat cell lysate

Lane 2:
HeLa cell lysate

Lane 3:
HEK293 cell lysate

Predicted band size: 102 kDa

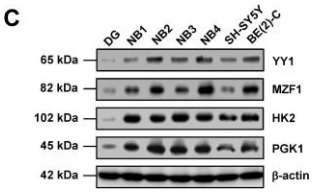


Image collected and cropped by CiteAb under a CC-BY license from the publication

Western blot - Anti-Hexokinase II antibody [3D3] (ab104836)

Hexokinase II Western blot staining using mouse Anti-Hexokinase II antibody

Western Blotting using Anti-Hexokinase II antibody [3D3], ab104836. Publication image from Fang, E. et al., 2020, Theranostics, 32042322. Legend direct from paper.

MZF1-uPEP/YY1/MZF1 expression is associated with NB outcome. (A) Representative images of immunohistochemical staining showing the expression patterns of MZF1-uPEP, YY1, and MZF1 in tumor cells of NB specimens (arrowheads, brown). Scale bars: 50 μm. (B) Kaplan-Meier curves indicating overall survival of 42 NB patients with high or low MZF1-uPEP immunostaining in 88 (GSE16476) NB cases with low or high expression levels of YY1 (cutoff value=1003.1). (C and D) Western blot (C) and real-time qRT-PCR (D, normalized to β-actin) assays showing the expression of YY1, MZF1, and target genes in normal dorsal root ganglia (DG), NB tissues (n=42), and NB cell lines. (E) Mining of a public microarray dataset (GSE16476) revealing the levels of YY1 in NB tissues with different status of age, death, or INSS stages. (F) The positive expression correlation of YY1 with MZF1, HK2, or PGK1 in 88 NB cases (GSE16476). (G) The mechanisms underlying MZF1-uPEP-suppressed tumor progression: as an uORF-encoded small peptide, MZF1-uPEP directly binds to YY1 to repress its transactivation, resulting in decreased transcription of MZF1 and downstream target glycolytic genes, and reduced aerobic glycolysis and tumor progression. Log-rank test for survival comparison in B. Student's t test compared the difference in D and E. Pearson's correlation coefficient analysis for gene expression in F. * P<0.05 vs. DG. Data are shown as mean ± s.e.m. (error bars) and representative of three independent experiments in C and D. Bars are means and whiskers (min to max) in E.

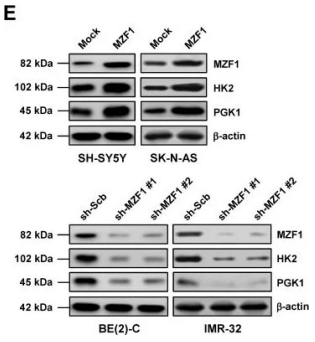


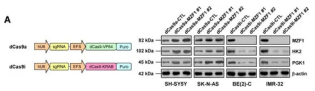
Image collected and cropped by CiteAb under a CC-BY license from the publication

Western blot - Anti-Hexokinase II antibody [3D3] (ab104836)

Hexokinase II Western blot staining using mouse Anti-Hexokinase II antibody

Western Blotting using Anti-Hexokinase II antibody [3D3], ab104836. Publication image from Fang, E. et al., 2020, Theranostics, 32042322. Legend direct from paper.

MZF1 facilitates the transcription of glycolytic genes in NB. (A) Venn diagram indicating the identification of glycolytic genes (left panel) and transcription factors (right panel) differentially expressed in 88 NB cases (GSE16476) with various status of age, death, and INSS stages, and the over-lapping analysis with potential transcription factors regulating glycolytic genes revealed by Genomatix program. The middle panel showing the potential transcription factors regulating expression of glycolytic genes. (B) Mining of a public microarray dataset (GSE16476) revealing the MZF1 levels in NB tissues with different status of age, death, or INSS stages. (C) Kaplan-Meier curve showing overall survival of 88 NB patients (GSE16476) with high or low MZF1 expression (cutoff value=226.6). (D and E) Real-time qRT-PCR (D, normalized to β-actin, n=4) and western blot (E) assays indicating the transcript and protein levels of MZF1, ALDOC, ENO1, GPI, HK2, LDHA, or PGK1 in SH-SY5Y and BE(2)-C cells stably transfected with empty vector (mock), MZF1, scramble shRNA (sh-Scb), or sh-MZF1. (F) ChIP and qPCR assays showing the binding of MZF1 to promoters of HK2 and PGK1 in SH-SY5Y, SK-N-AS, BE(2)-C, and IMR-32 cells stably transfected with mock, MZF1, sh-Scb, or sh-MZF1 (n=4). (G) Dual-luciferase assay indicating the promoter activity of HK2 and PGK1 with wild-type (WT) or mutant (Mut) MZF1 binding site in SH-SY5Y and BE(2)-C cells stably transfected with mock, MZF1, sh-Scb, or sh-MZF1 (n=6). Fisher's exact test for over-lapping analysis in A. Student's t test compared the difference in B. Log-rank test for survival comparison in C. Student's t test and ANOVA compared the difference in D, F and G. * P<0.05, ** P<0.01 vs. mock or sh-Scb. δP<0.05 vs. WT. Bars are means and whiskers (min to max) in B. Data are shown as mean ± s.e.m. (error bars) and representative of three independent experiments in D-G.



Western blot - Anti-Hexokinase II antibody [3D3] (ab104836)

Image collected and cropped by CiteAb under a CC-BY license from the publication

Hexokinase II Western blot staining using mouse Anti-Hexokinase II antibody

Western Blotting using Anti-Hexokinase II antibody [3D3], ab104836. Publication image from Fang, E. et al., 2020, Theranostics, 32042322. Legend direct from paper.

MZF1 promotes the aerobic glycolysis of NB cells. (A) Western blot assay indicating the expression of MZF1, HK2, and PGK1 in SH-SY5Y, SK-N-AS, BE(2)-C, and IMR-32 cells stably transfected with dCas9a control (dCas9a-CTL), dCas9a-MZF1, dCas9i control (dCas9i-CTL), or dCas9i-MZF1. (B) Seahorse tracing curves (left panel) and ECAR bars (right panel) of SH-SY5Y and BE(2)-C cells stably transfected with empty vector (mock), MZF1, scramble shRNA (sh-Scb), sh-MZF1, dCas9a-CTL, dCas9a-MZF1, dCas9i-CTL, or dCas9i-MZF1, and those treated with glucose (10 mmol・L-1), oligomycin (2 μmol・L-1), or 2-deoxyglucose (2-DG, 100 mmol・L-1) at indicated (4 replicates for each point). (C and D) Glucose uptake, lactate production, and ATP levels in SH-SY5Y (C) and BE(2)-C (D) cells stably transfected with mock, MZF1, sh-Scb, sh-MZF1, dCas9a-CTL, dCas9a-MZF1, dCas9i-CTL, or dCas9i-MZF1 (n=4). Student's t test and ANOVA compared the difference in B-D. ** $P<0.01$ vs. mock, sh-Scb, dCas9a-CTL, or dCas9i-CTL. Data are shown as mean ± s.e.m. (error bars) and representative of three independent experiments in A-D.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.