

KLF4 Polyclonal Antibody

Catalog NumberPA1-095

Product data sheet

Details		Species Reactivity	
Size	200 µL	Species reactivity	Human, Mouse
Host/Isotope	Rabbit / IgG	Tested Applications	Dilution *
Class	Polyclonal		
Type	Antibody	Western Blot (WB)	1:500-1:2,000
Immunogen	Recombinant N-terminal fragment (amino acids 1-118) of human KLF4	Immunocytochemistry (ICC/IF)	1-2 µg/mL
		* Suggested working dilutions are given as a guide only. It is recommended that the user titrate the product for use in their own experiment using appropriate negative and positive controls.	
Conjugate	Unconjugated		
Form	Liquid		
Concentration	0.5 mg/mL		
Purification	Protein A		
Storage buffer	PBS with 1mg/mL BSA		
Contains	0.05% sodium azide		
Storage Conditions	-20°C		

Product specific information

The predicted molecular weight of KLF4 is ~55kD. The migration of this protein by SDS-PAGE was observed slightly higher (~60kD) with both PA1-095 and a benchmark antibody. PA1-095 detects a predominant band at ~60kD, and a nonspecific band of unknown origin at ~80kD in both whole cell lysates and nuclear extracts. For best results, StartingBlock T20 (TBS) Blocking Buffer (Product # 37543) is recommended.

Background/Target Information

Kruppel like factor 4 (KLF4) protein is encoded by the KLF4 gene located on chromosome 9 in humans. It is also known as gut-enriched Kruppel-like factor (GKLF) or epithelial zinc-finger protein (EZf) and belongs to a family of conserved Kruppel-like factor family of DNA binding zinc finger transcription factors. KLF4 exerts its functional relevance across varied cellular pathways. As one of the four Yamanaka factors, it plays a key role in reprogramming differentiated cells into pluripotent stem cells. Most studies have focused on its specific role in endothelial cell signaling, adipogenesis, differentiation, embryogenesis and cell cycle control. Localization plays an important role in the biological significance of this protein wherein nuclear localization is associated with its function as an oncogene and cytosolic localization with tumor suppression specific effect.

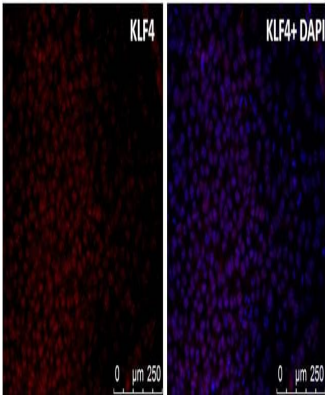
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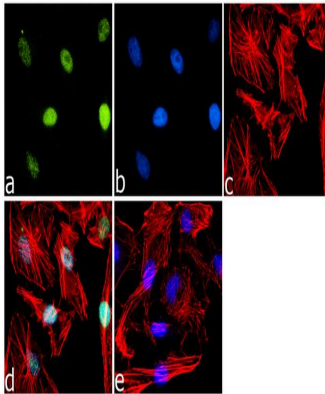
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Product Images For KLF4 Polyclonal Antibody



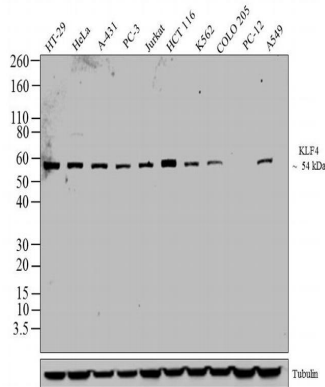
KLF4 Antibody (PA1-095) in ICC/IF

Immunofluorescent analysis of KLF4 (red) in human embryonic stem cell H9 line grown on irradiated MEF-feeder layer. The cells were fixed with 4% paraformaldehyde at room temperature for 10 min and permeabilized with 0.25% Triton-X 100 for 5 min and blocked with the 10% BSA in PBS for 30 min at 37°C. Cells were stained with a KLF4 polyclonal antibody (Product # PA1-095) at a dilution of 1:200 in 3% BSA/PBS blocking buffer overnight at 4°C, and then incubated with a RRX-conjugated donkey anti-rabbit IgG secondary antibody at a dilution of 1:500 for 1 hour at room temperature. Nucleus DNA (blue) was stained with DAPI (Product # D1306).



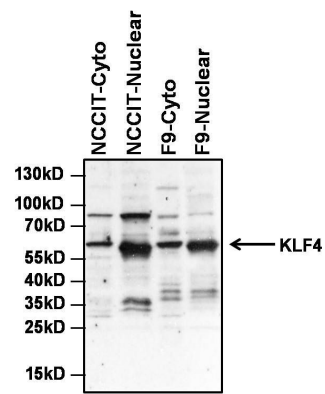
KLF4 Antibody (PA1-095) in ICC/IF

Immunofluorescence analysis of KLF4 was done on 70% confluent log phase HeLa cells. The cells were fixed with 4% paraformaldehyde for 15 minutes, permeabilized with 0.25% Triton™ X-100 for 10 minutes, and blocked with 5% BSA for 1 hour at room temperature. The cells were labeled with KLF4 Rabbit Polyclonal Antibody (Product # PA1-095) at 1 µg/mL in 1% BSA and incubated for 3 hours at room temperature and then labeled with Goat anti-Rabbit IgG (Heavy Chain) Superclonal™ Secondary Antibody, Alexa Fluor® 488 conjugate (Product # A27034) at a dilution of 1:2000 for 45 minutes at room temperature (Panel a: green). Nuclei (Panel b: blue) were stained with SlowFade® Gold Antifade Mountant with DAPI (Product # S36938). F-actin (Panel c: red) was stained with Alexa Fluor® 555 Rhodamine Phalloidin (Product # R415, 1:300). Panel d is a merged image showing nuclear localization. Panel e is a no primary antibody control. The images were captured at 60X magnification.



KLF4 Antibody (PA1-095) in WB

Western blot analysis was performed on whole cell extracts (20 µg lysate) of HT-29 (Lane 1), HeLa (Lane 2), A-431 (Lane 3), PC-3 (lane 4), Jurkat (lane 5), HCT 116 (lane 6), K562 (lane 7), COLO 205 (lane 8), PC-12 (lane 9) and A549 (lane 10). The blots were probed with Anti-KLF4 Rabbit Polyclonal Antibody (Product # PA1-095, 1:500-1:1500 dilution) and detected by chemiluminescence using Goat anti-Rabbit IgG (Heavy Chain) Superclonal™ Secondary Antibody, HRP conjugate (Product # A27036, 0.4 µg/mL, 1:2500 dilution). A 54 kDa band corresponding to KLF4 was observed across cell lines tested expect for PC-12. Known quantity of protein samples were electrophoresed using Novex® NuPAGE® 4-12 % Bis-Tris gel (Product # NP0322BOX), XCell SureLock™ Electrophoresis System (Product # EI0002) and Novex® Sharp Pre-Stained Protein Standard (Product # LC5800). Resolved proteins were then transferred onto a nitrocellulose membrane with iBlot® 2 Dry Blotting System (Product # IB21001). The membrane was probed with the relevant primary and secondary Antibody following blocking with 5 % skimmed milk. Chemiluminescent detection was performed using Pierce™ ECL Western Blotting Substrate (Product # 32106).



KLF4 Antibody (PA1-095) in WB

Western blot analysis of KLF4 was performed by loading 25 µg of NCCIT and F9 cytoplasmic and nuclear extracts, and 10 µL of PageRuler Prestained Protein Ladder (Product # 26616) per well onto a 4-20% Tris-HCl polyacrylamide gel. Proteins were transferred to a PVDF membrane and blocked with StartingBlock T20 (TBS) Blocking Buffer (Product # 37543) for at least 1 hour at room temperature. The membrane was probed with a KLF4 polyclonal antibody (Product # PA1-095) at a dilution of 1:1000 overnight at 4°C on a rocking platform, washed in TBS-0.1% Tween-20, and probed with an HRP-conjugated goat anti-rabbit IgG secondary antibody (Product # 31460) at a dilution of 1:40,000 for at least 30 minutes. Chemiluminescent detection was performed using SuperSignal West Dura (Product # 34075). NOTE: Cytoplasmic and nuclear extracts were obtained using NE-PER Nuclear Protein Extraction Kit (Product # 78833).

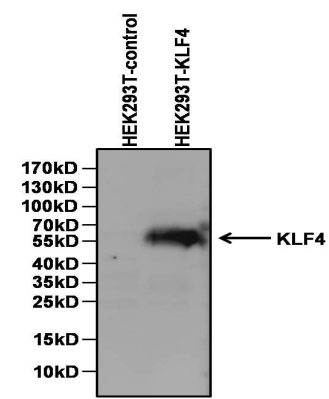
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**KLF4 Antibody (PA1-095) in WB**

Western blot analysis of KLF4 was performed by loading 20 µg of HEK293T lysates from cells transfected with a control vector (left lane) or a KLF4 overexpression plasmid (right panel) and 10 µL of PageRuler Prestained Protein Ladder (Product # 26616) per well onto a 4-20% Tris-HCl polyacrylamide gel. Proteins were transferred to a PVDF membrane and blocked with 5% BSA in TBST for at least 1 hour. The membrane was probed with a KLF4 polyclonal antibody (Product # PA1-095) at a dilution of 1:1000 overnight at 4°C on a rocking platform, washed in TBS-0.1% Tween-20, and probed with an HRP-conjugated goat anti-rabbit IgG secondary antibody (Product # 31460) at a dilution of 1:40,000 for at least 30 minutes. Chemiluminescent detection was performed using SuperSignal West Pico (Product # 34080).



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