

TGF-β Fibrosis Pathway Antibody Kit

HAK21004



Contains Product	Specification	Applications	Species reactivity	MW(kDa)
alpha smooth muscle Actin[ET1607-53]	20µl	WB,IF,CellIF-Tissue,IHC-P,FC,mIHC	H,M,R	42 kDa
COL1A1[ET1609-68]	20µl	WB,IHC-P	H	139 kDa
Smad2.3[RT1566]	20µl	WB,IF,IHC-P	H,M,R	55-60kDa
Smad2[ET1604-22]	20µl	WB,IF,CellIHC-P,IF,FC	H,M,R	52 kDa
Phospho-Smad3(S423,S425)[ET1609-41]	20µl	WB,IF,CellIF-Tissue,IHC-P	H,M	48 kDa
YKL-40 / CHI3L1[EM1902-14]	20µl	WB,IHC-P,IF-Cell	H,M	43 kDa
TGF beta Receptor II[ER1917-66]	20µl	WB,ELISA,IHC-P,IF	H,M,R,C,Pg,C,Rab.	62 kDa
TGF beta 1[HA721143]	20µl	WB,IHC-P,IF-Tissue	H,M,R	44 kDa
HRP-Alpha anti-Rabbit IgG Fc, Recombinant VHH[HA1031]	100µl	IP,ELISA,IHC-P,WB	Rab	
HRP-Goat anti-Mouse IgG[HA1006]	100µl	WB,ELISA,IHC-P	M	

Description:

Transforming growth factor-β The fibrosis pathway Antibody Sampler Kit provides an economical method to study the activation of transforming growth factor-β/ Smad2 / 3 signaling pathway leads to the expression of Pro fibrogenic genes in cells or tissues, including alpha smooth muscle Actin, COL1A1/Collagen I and YKL-40 / CHI3L1 were up-regulated in activated fibroblasts. The kit contains enough antibodies to perform at least two Western blotting tests on each primary antibody. And also includes secondary reagent for detection of these antibodies.

Storage Buffer:

1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction:

Store at +4°C after thawing. Aliquot store at -20°C. Avoid repeated freeze / thaw cycles.

Background

Transforming growth factor-β (TGF-β) superfamily members are critical regulators of cell proliferation and differentiation, developmental patterning and morphogenesis, and disease pathogenesis.

In the context of fibrosis, TGF-β signaling to SMAD2/3 is one of the biggest drivers of the profibrotic program. TGF-β elicits signaling through three cell surface receptors: type I (RI), type II (RII), and type III (RIII). Activated type I receptors associate with SMAD2/3 and phosphorylate them on a conserved carboxy terminal SXS motif. In the context of fibrosis, SMAD2/3 activation upregulates expression of profibrotic genes such as COL1A1 and other ECM modulators that modify the extracellular matrix of the tissue. TGF-β/ SMAD2/3 signaling also induces expression of α-Smooth Muscle Actin in fibroblasts, causing transformation of these cells to myofibroblasts. Injury to the tissue attracts macrophages and other immune cells and the fibrotic tissue soon becomes a site of inflammation. In this pro-fibrotic, pro-inflammatory environment, YKL-40, also known as Chitinase-3-like protein 1 (CHI3L1), is secreted. YKL-40 is a pro-inflammatory glycoprotein that also contributes to the progression of fibrosis.

Database links:

UniProt ID: P62736, P62737, P62738, P02452, 282187Cow, Q15796, Q15796, Q62432, O70436, P84022, Q8BUN5, P36222, Q61362, P37173, P01137, P04202, P17246

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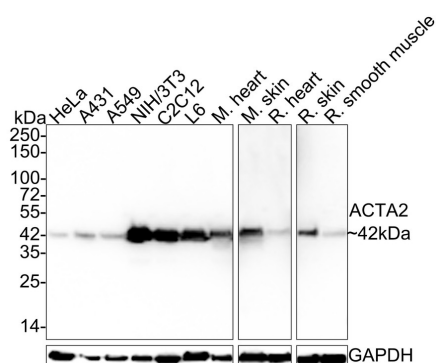


Fig1: Western blot analysis of alpha smooth muscle Actin on different lysates with Rabbit anti-alpha smooth muscle Actin antibody (ET1607-53) at 1/5,000 dilution.

Lane 1: HeLa cell lysate

Lane 2: A431 cell lysate

Lane 3: A549 cell lysate

Lane 4: NIH/3T3 cell lysate

Lane 5: C2C12 cell lysate

Lane 6: L6 cell lysate

Lane 7: Mouse heart tissue lysate

Lane 8: Mouse skin tissue lysate

Lane 9: Rat heart tissue lysate

Lane 10: Rat skin tissue lysate

Lane 11: Rat smooth muscle tissue lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 42 kDa

Observed band size: 42 kDa

Exposure time: 5 seconds;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDm/TBST for 1 hour at room temperature. The primary antibody (ET1607-53) at 1/5,000 dilution was used in 5% NFDm/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:100,000 dilution was used for 1 hour at room temperature.

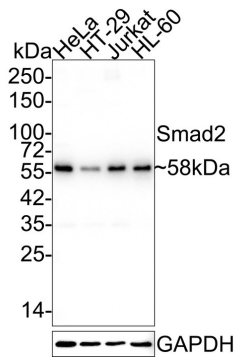


Fig2: Western blot analysis of Smad2 on different lysates with Rabbit anti-Smad2 antibody (ET1604-22) at 1/5,000 dilution.

Lane 1: HeLa cell lysate

Lane 2: HT-29 cell lysate

Lane 3: Jurkat cell lysate

Lane 4: HL-60 cell lysate

Lysates/proteins at 15 μ g/Lane.

Predicted band size: 52 kDa

Observed band size: 58 kDa

Exposure time: 1 minute 20 seconds;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDm/TBST for 1 hour at room temperature. The primary antibody (ET1604-22) at 1/5,000 dilution was used in 5% NFDm/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:50,000 dilution was used for 1 hour at room temperature.

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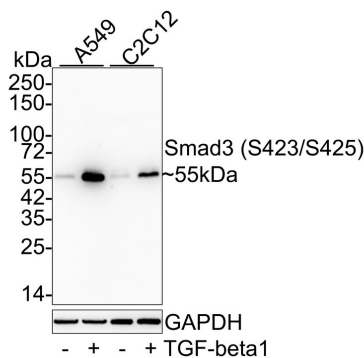
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Fig3: Western blot analysis of Phospho-Smad3(S423/S425) on different lysates with Rabbit anti-Phospho-Smad3(S423/S425) antibody (ET1609-41) at 1/2,000 dilution.



Lane 1: A549 whole cell lysate

Lane 2: A549 treated with 5ng/mL TGF-beta1 for 24 hours whole cell lysate

Lane 3: C2C12 whole cell lysate

Lane 4: C2C12 treated with 5ng/mL TGF-beta1 for 24 hours whole cell lysate

Lysates/proteins at 15 µg/Lane.

Predicted band size: 48 kDa

Observed band size: 55 kDa

Exposure time: 3 minutes 30 seconds;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1609-41) at 1/2,000 dilution was used in 5% NFDM/TBST at 4°C overnight. Goat Anti-Rabbit IgG- HRP Secondary Antibody (HA1001) at 1:50,000 dilution was used for 1 hour at room temperature.

Note: All products are “FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE”.

Background References

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