

## Anti-Smad2/3 Rabbit mAb

Purified Recombinant Rabbit Monoclonal Antibody Catalog # R013762

## **Product Information**

Application ELISA, IF (Cell)/ICC, WB, IHC-P, IF (Tissue-P)

Reactivity Human, Mouse, Rat

**Dilution** WB 1:1,000~1:4,000; IHC-P 1:200; IF 1:100

Host Rabbit

Clone No. 14L86C22
Isotype IgG

Label Unconjugated

Immunogen Recombinant protein of human Smad2

Format Affinity purified monoclonal antibody supplied in PBS with 0.01% sodium azide and 50% glycerol, pH 7.3.

Storage Shipped on wet ice. Store at -20°C. Stable for 24 months from date of receipt. Aliquoting is unnecessary for -20°C storage.

Precautions Anti-Smad2/3 Rabbit mAb [14L86C22] is for research use only and not for use in diagnostic or therapeutic procedures.

## **Protein Information**

Synonyms SMAD3, MADH3, Mothers against decapentaplegic homolog 3, MAD homolog 3, Mad3, Mothers against DPP homolog 3,

hMAD-3, JV15-2, SMAD family member 3, SMAD 3, Smad3, hSMAD3, JV18, MADH2, MADR2, JV18-1, hMAD-2,

hSMAD2.

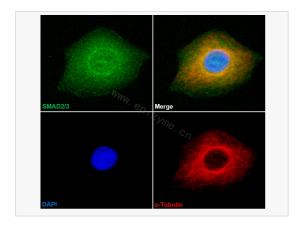
Calculated MW: 52 kDa; Observed MW: 58-62 kDa

Uniprot ID P84022, Q15796 Gene ID 4087/4088

Background Members of the Smad family of signal transduction molecules are components of a critical intracellular pathway that transmit

 $TGF-\beta$  signals from the cell surface into the nucleus. Three distinct classes of Smads have been defined: the receptor-regulated Smads (R-Smads), which include Smad1, 2, 3, 5, and 8; the common-mediator Smad (co-Smad), Smad4; and the antagonistic or inhibitory Smads (I-Smads), Smad6 and 7 . Activated type I receptors associate with specific R-Smads and phosphorylate them on a conserved carboxy terminal SSXS motif. The phosphorylated R-Smad dissociates from the receptor and forms a heteromeric complex with the co-Smad (Smad4), allowing translocation of the complex to the nucleus. Once in the nucleus,

 $Smads\ can\ target\ a\ variety\ of\ DNA\ binding\ proteins\ to\ regulate\ transcriptional\ responses\ .$ 



Immunofluorescence - Anti-Smad2/3 Rabbit mAb [14L86C22]

Sample: Hela cells

The cells were fixed with 4% paraformal dehyde (10 min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 5% BSA in 0.1% PBS-Tween for 0.5 .

hours.

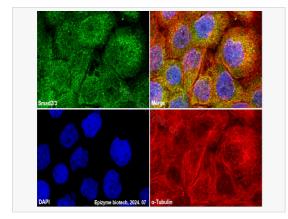
Primary antibodies: R013762 at 1:100 dilution and α-tubulin Mouse Monoclonal

Antibody (Cat. No. LF209) at 1:100 dilution

Secondary antibodies: Goat anti-Rabbit (488) at 1:1,000 dilution (shown in green) and

Goat anti-Mouse (555) at 1:1,000 dilution (shown in red)

Nuclei were stained with DAPI (shown in blue).



Immunofluorescence - Anti-Smad2/3 Rabbit mAb [14L86C22]

Sample: A431 cells

The cells were fixed with 4% paraformaldehyde (10 min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 5% BSA in 0.1% PBS-Tween for 0.5 hours

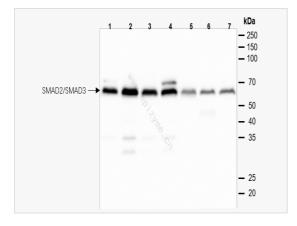
Primary antibodies: R013762 at 1:100 dilution and  $\alpha\text{-tubulin}$  Mouse Monoclonal

Antibody (Cat. No. LF209) at 1:100 dilution

Secondary antibodies: Goat anti-Rabbit (488) at 1:1,000 dilution (shown in green) and

Goat anti-Mouse (555) at 1:1,000 dilution (shown in red)

Nuclei were stained with DAPI (shown in blue).



Western Blot - Anti-Smad2/3 Rabbit mAb [14L86C22]

All lanes: R013762 at 1:4,000 dilution

Lane 1: C2C12 (mouse myoblasts epithelial cell) whole cell lysates

Lane 2: Jurkat (human T lymphocytic leukemia cell) whole cell lysates

Lane 3: HCT116 (human colorectal carcinoma epithelial cell) whole cell lysates

Lane 4: RAW264.7 (mouse mononuclear macrophage leukemia epithelial cell)

Lane 5: HepG2 (human hepatocellular carcinoma epithelial cell) whole cell lysates

Lane 6: Rat stomach whole tissue lysates

Lane 7: Balb/c mouse liver whole tissue lysates

Lysates/proteins at 10  $\mu g$  per lane.

 $Secondary\ antibody:\ Goat\ Anti-Rabbit\ IgG(H\!+\!L),\ HRP\ Conjugated\ (Cat.\ No.\ LF102)\ at$ 

1:5,000 dilution

Predicted band size: 52 kDa

Observed band size: 58 kDa, 62 kDa

Developed using the ECL technique (Cat. No. SQ201).



Western Blot - Anti-Smad2/3 Rabbit mAb [14L86C22]

All lanes: R013762 at 1:1,000 dilution

Lane 1: MCF7 (human breast adenocarcinoma epithelial cell) whole cell lysates

Lane 2: T24 (human bladder cancer epithelial cell) whole cell lysates

Lane 3: HepG2 (human hepatocellular carcinoma epithelial cell) whole cell lysates

Lane 4: SW620 (human colorectal carcinoma epithelial cell) whole cell lysates

Lane 5: Rat brain whole tissue lysates

Lane 6: Rat liver whole tissue lysates

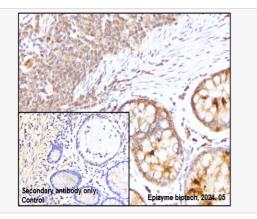
Lysates/proteins at 10 µg per lane.

Secondary antibody: Goat Anti-Rabbit IgG(H+L), HRP Conjugated (Cat. No. LF102) at

1:5,000 dilution

Predicted band size: 52 kDa Observed band size: 58-62 kDa

Developed using the ECL technique (Cat. No. SQ201).



Immunohistochemistry - Anti-Smad2/3 Rabbit mAb [14L86C22]

Sample: Paraformaldehyde-fixed, paraffin embedded human colorectal carcinoma tissue Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins.

Primary antibody: R013762 at 1:200 dilution

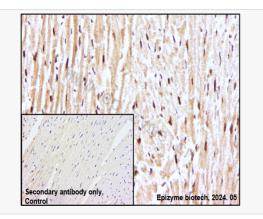
Secondary antibody: Goat Anti-Rabbit IgG (H+L), HRP conjugated at 1:1,000 dilution

DAB was used as the chromogen.

Counter stained with hematoxylin.

Positive/negative staining were presented.

Only the secondary antibody was used as the negative control.



Immunohistochemistry - Anti-Smad2/3 Rabbit mAb [14L86C22]

Sample: Paraformaldehyde-fixed, paraffin embedded mouse heart tissue

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins.

Primary antibody: R013762 at 1:200 dilution

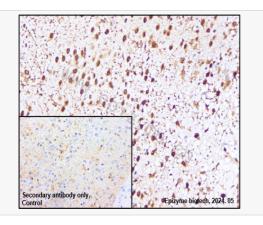
Secondary antibody: Goat Anti-Rabbit IgG (H+L), HRP conjugated at 1:1,000 dilution

DAB was used as the chromogen.

Counter stained with hematoxylin.

Positive/negative staining were presented.

Only the secondary antibody was used as the negative control.



Immunohistochemistry - Anti-Smad2/3 Rabbit mAb [14L86C22]

Sample: Paraformaldehyde-fixed, paraffin embedded mouse brain tissue

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins.

Primary antibody: R013762 at 1:200 dilution

Secondary antibody: Goat Anti-Rabbit IgG (H+L), HRP conjugated at 1:1,000 dilution

DAB was used as the chromogen.

Counter stained with hematoxylin.

Positive/negative staining were presented.

Only the secondary antibody was used as the negative control.