SMP30 (Senescence Marker Protein 30)
Regucalcin
Gluconolactonase (GNL)
WesternBlot・ImmunoStain Kit

Catalog Number: RDIK01-EX

Kit component

Antibody :	Rabbit anti SMP30・GNL antibody 0.1 mL
	10 mM Tris (pH 7.4), 0.14 M NaCl
	This kit does not contain NaN_3
Specimen :	SMP30・GNL Knockout Mouse Liver 2 slides
	Wild type Mouse Liver 2 slides
Tissue extract :	SMP30・GNL Knockout Mouse Liver 30 μL
	(Protein concentration 0.4 mg/mL)
	Wild type Mouse Liver 30 μL
	(Protein concentration 0.4 mg/mL)
Storage and Stability :	-20 ℃, 2 years

【Antibody】

SMP30 (Senescence Marker Protein 30) IgG

Gluconolactonase (GNL) IgG

Regucalcin IgG

★ SMP30, Gluconolactonase and Regucalcin are all identical protein.

Rabbit Polyclonal Antibody (Purified IgG Fraction)

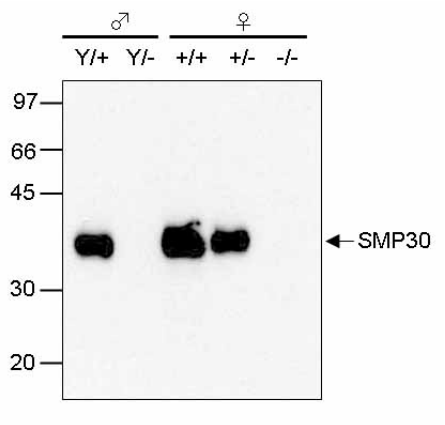
Volume :	0.1 mL
Antigen :	Rat SMP30 purified from rat liver, Molecular weight 34 kDa
Host :	Rabbit
Supplied As :	IgG fraction purified from rabbit serum.
	Prepared in 10 mM Tris (pH 7.4), 0.14 M NaCl.
Storage and Stability :	-20 ℃, 2 years
Tested applications :	• Immunofluorescence (1:100-1:500 dilution)
	• immunohistochemistry (1:100-1:500 dilution)
	• Western Blot (1:1,000-1:3,000 dilution)
Cross Reactivity :	Cross reacts with Human, Mouse and Rat SMP30.
	Not yet tested in other species.

【Sample Preparation for Western blot】

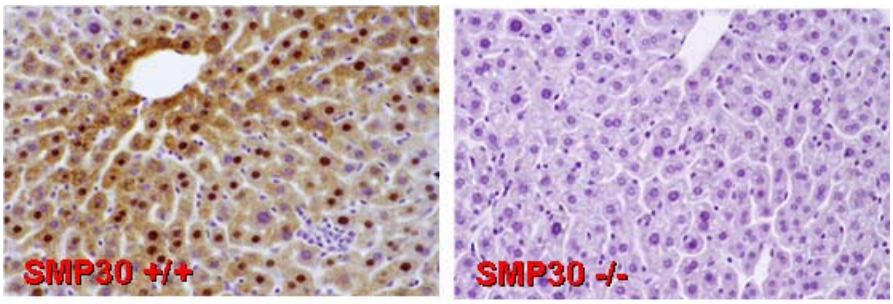
SMP30・GNL Knockout Mouse Liver　30 μL (Protein concentration 0.4 mg/mL)
Wild type Mouse Liver　30 μL (Protein concentration 0.4 mg/mL)

1. Add 30 μl SDS-PAGE Lysis Buffer.
2. Boil at 95ºC for 5 minutes and cool on ice.
3. Centrifuge at 10,000 rpm for 5 minutes and transfer the supernatant to a fresh tube.
4. Load the sample 10 μL (Protein 2 μg) per lane.

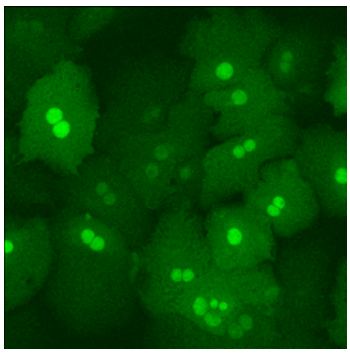
2X SDS-PAGE Lysis Buffer
125 mM Tris-HCl, pH 6.8
4% SDS
10% 2-mercaptoethanol
20% glycerol
0.01% bromophenol blue (BPB)



[Western Blot Analysis]
Each lanes：Mouse Liver Extract
• Wild type Mouse：SMP30Y/+ and SMP30+/+
• SMP30 Knockout Mouse：SMP30Y/- and SMP30-/-
• Heterozygous Mouse：SMP30+/-
SMP30・GNL antibody at 1:1,000 dilution used.



[Immunohistochemical staining]
Mouse liver stained with SMP30・GNL antibody at 1:300 dilution and developed by 3,3'-diaminobenzidine. Nucleus and cytoplasm of wild type (SMP30+/+) mice stained, but not stained in liver from SMP30 knockout (SMP30-/-) mice.



[Immunofluorescence staining]

Primary cultured mouse hepatocytes stained with SMP30・GNL antibody at 1:200 dilution. Nucleus and cytoplasm stained in green.

References :

1. Ishigami, A. et al., Senescence marker protein-30 knockout mouse liver are highly susceptible to TNF-alpha- and Fas-mediated apoptosis. *Am. J. Pathol.* 161 1273-1281 (2002)
2. Ishigami, A. et al., Nuclear localization of senescence marker protein-30 (SMP30) in cultured mouse hepatocytes and its homology to RNA polymerase. *Biosci. Biotechnol. Biochem.* 67 158-160 (2003)
3. Kondo, Y. et al., Senescence Marker Protein 30 Functions as Gluconolactonase in L-Ascorbic Acid Biosynthesis and Its Knockout Mice Are Prone to Scurvy. *Proc. Nat. Acad. Sci. USA* 103 5723-5728 (2006)
4. Sato, T. et al., Senescence Marker Protein-30 Protects Mice Lungs from Oxidative Stress, Aging and Smoking. *Am. J. Respir. Crit. Care Med.* 174 530-537 (2006)

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