

Datasheet for ABIN705056  
**anti-TNFSF9 antibody (AA 101-200)**[Go to Product page](#)

## 1 Validation

## 3 Images

## Overview

Quantity:	100 µL
Target:	TNFSF9
Binding Specificity:	AA 101-200
Reactivity:	Human, Mouse, Rat
Host:	Rabbit
Clonality:	Polyclonal
Conjugate:	This TNFSF9 antibody is un-conjugated
Application:	Western Blotting (WB), ELISA, Immunocytochemistry (ICC), Immunofluorescence (Cultured Cells) (IF (cc)), Immunofluorescence (Paraffin-embedded Sections) (IF (p)), Immunohistochemistry (Paraffin-embedded Sections) (IHC (p)), Immunohistochemistry (Frozen Sections) (IHC (fro))

## Product Details

Immunogen:	KLH conjugated synthetic peptide derived from human TNFSF9
Isotype:	IgG
Cross-Reactivity:	Human, Mouse, Rat
Purification:	Purified by Protein A.

## Target Details

Target:	TNFSF9
Alternative Name:	TNFSF9/CD137L ( <a href="#">TNFSF9 Products</a> )

## Target Details

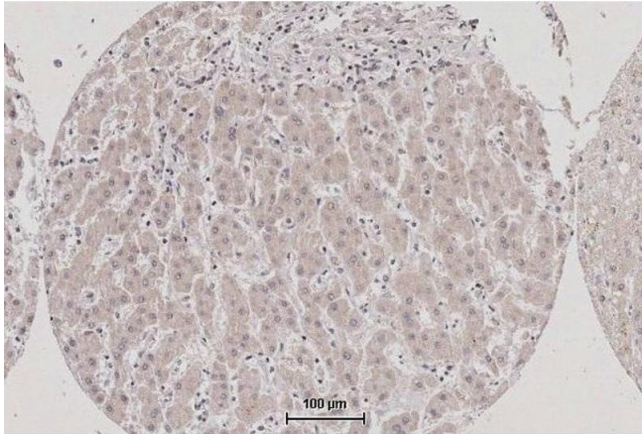
Background:	Synonyms: CD137L, 4-1BB-L, Tumor necrosis factor ligand superfamily member 9, 4-1BB ligand, 4-1BBL, TNFSF9 Background: Cytokine that binds to TNFRSF9. Induces the proliferation of activated peripheral blood T-cells. May have a role in activation-induced cell death (AICD). May play a role in cognate interactions between T-cells and B-cells/macrophages.
Gene ID:	8744
UniProt:	<a href="#">P41273</a>
Pathways:	<a href="#">Activated T Cell Proliferation</a> , <a href="#">Cancer Immune Checkpoints</a>

## Application Details

Application Notes:	WB 1:300-5000 ELISA 1:500-1000 IHC-P 1:200-400 IHC-F 1:100-500 IF(IHC-P) 1:50-200 IF(IHC-F) 1:50-200 IF(ICC) 1:50-200 ICC 1:100-500
Restrictions:	For Research Use only

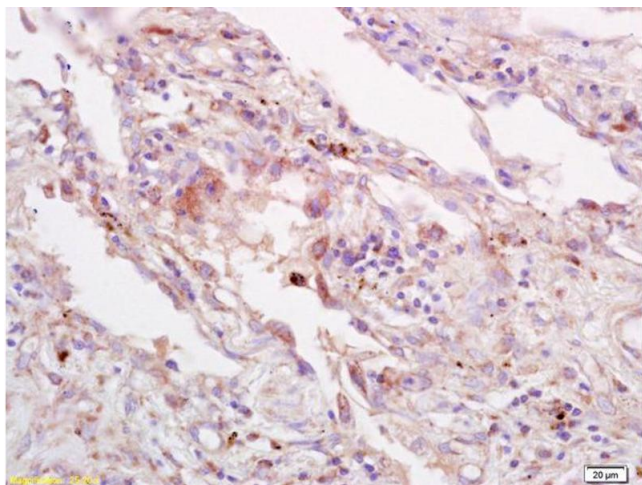
## Handling

Format:	Liquid
Concentration:	1 µg/µL
Buffer:	0.01M TBS( pH 7.4) with 1 % BSA, 0.02 % Proclin300 and 50 % Glycerol.
Preservative:	ProClin
Precaution of Use:	This product contains ProClin: a POISONOUS AND HAZARDOUS SUBSTANCE, which should be handled by trained staff only.
Storage:	4 °C, -20 °C
Storage Comment:	Shipped at 4°C. Store at -20°C for one year. Avoid repeated freeze/thaw cycles.
Expiry Date:	12 months



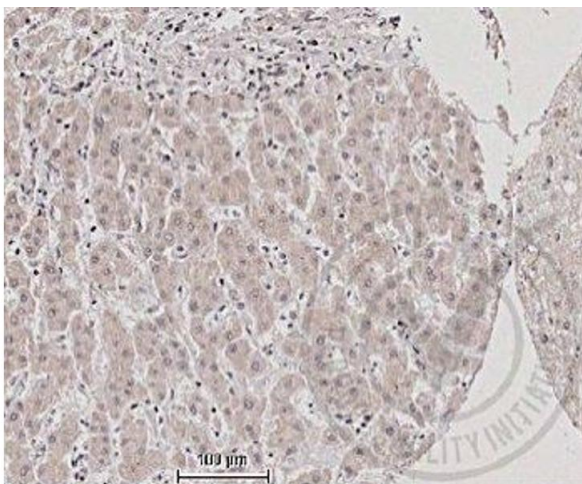
#### Immunohistochemistry

**Image 1.** Independently Validated Antibody, image provided by Science Direct, badge number 029578:Formalin-fixed and paraffin embedded human liver labeled with Anti-TNFSF9/CD137L Polyclonal Antibody, Unconjugated (ABIN705056) at 1:200 followed by conjugation to the secondary antibody and DAB staining



#### Immunohistochemistry

**Image 2.** Formalin-fixed and paraffin embedded human lung carcinoma labeled with Anti-TNFSF9/CD137L Polyclonal Antibody, Unconjugated (ABIN705056) at 1:200 followed by conjugation to the secondary antibody and DAB staining



#### Immunohistochemistry (Paraffin-embedded Sections)

**Image 3.** Images provided the Independent Validation Program, badge number 029578:Formalin-fixed and paraffin embedded human liver labeled with Anti-TNFSF9/CD137L Polyclonal Antibody, Unconjugated at 1:200 followed by conjugation to the secondary antibody and DAB staining



## Successfully validated (Immunohistochemistry (IHC))

by [Immunohistochemistry Core, NYU Langone](#)

Report Number: 029578

Date: Jan 18 2014

Lot Number:	999892W
Method validated:	Immunohistochemistry (IHC)
Positive Control:	<a href="#">Human liver</a>
Negative Control:	<a href="#">Human placental decidual cells</a>
Notes:	Signal was detected in positive control tissue and not in negative control tissue.
Primary Antibody:	- Antibody: Tumor necrosis factor ligand superfamily member 9 (TNFSF9) - Catalog number: ABIN705056 - Supplier: Bioss - Supplier number: bs-3851r - Lot number: 999892W
Secondary Antibody:	- Antibody: Biotinylated goat anti-rabbit/anti-mouse (Kit) - Catalog number: 760-091 - Supplier: Ventana Medical Systems - Lot number: D05923BA
Isotype:	- Antibody: Rabbit IgG isotype control - Catalog number: 790-4795 - Supplier: Ventana Medical Systems - Lot number: C11487
Controls:	<ul style="list-style-type: none"><li>• Tissues stained came from a human FFPE tissue microarray (12-003d):</li><li>• Positive control: human liver (specimen known to contain the target protein).</li><li>• Negative Control: human placental decidual cells (specimen known to not contain the target protein).</li><li>• Primary antibody isotype control: Human placenta and human liver (specimen known to contain the target protein) treated with primary antibody isotype control instead of the primary antibody.</li><li>• Secondary antibody only control: Human placenta and human liver (specimen known to contain the target protein) treated with secondary antibody only (no primary antibody).</li></ul>
Protocol:	<ul style="list-style-type: none"><li>• Immunohistochemistry was performed on a Ventana NexES automated platform, instrument manufacturer specific reagents are italicized.</li><li>• 1. Slides were preheated in convection oven at 60°C for 30 minutes</li><li>• 2. Deparaffinization procedure: - 3 changes of Xylene, 5 minutes each - 3 changes of 100% Ethanol, 3 minutes each - 3 changes of 95% Ethanol, 3 minutes each - Rinsed in distilled water, 3 changes</li><li>• 1. Heat retrieval procedure - Slides retrieved in 10.0 mM Citrate, pH6.0 in a 1000W microwave oven (~100°C) for 15 minutes. - Slides were allowed to cool (in citrate) for 30 minutes. -</li></ul>

Slides were washed x 3 in Distilled water

- 1. NexES instrument procedure, iVIEW DAB paraffin protocol (\*abridged\*): - Slide chamber warmed to 37°C
- 1. Slides rinsed with \*reaction buffer\* x 3
- 1. \*iVIEW Inhibitor (H2O2)\* applied and incubated for 4 minutes
- 1. Slides rinsed with \*reaction buffer\*
- 1. Antibody Application - Primary antibody diluted 1:250 in PBS (100 microliters applied/slide) - Ventana Isotype control applied neat - Slides incubated overnight at room temperature (~12 hours ~25°C)
- 1. Slides rinsed with \*reaction buffer\* x3
- 1. \*iVIEW Biotinylated IgG\* applied and incubated for 8 minutes
- 1. Slides rinsed with \*reaction buffer\*
- 1. \*iVIEW Streptavidin-Horseradish Peroxidase\* applied and incubated for 8 minutes
- 1. Slides rinsed with \*reaction buffer\*
- 1. \*iVIEW DAB/H2O2\* applied and incubated for 8 minutes
- 1. Slides rinsed with \*reaction buffer\*
- 1. \*iVIEW Copper\* applied and incubated for 4 minutes
- 1. Slides rinsed with \*reaction buffer\*
- 1. Slides washed in Dawn Detergent/tap water
- 1. Counterstain Procedure - Hematoxylin (Leica 560 MX) 30 seconds - Slides washed in tap water, 1 minute - Decolorized (10% Acetic Acid in 70% ethanol), 1 minute - Slides washed in tap water, 1 minute - Bluing (Austin Clear Ammonia), 1 minute - Slides washed in tap water, 1 minute
- 1. Dehydration/coverslipping procedure: - 3 changes of 95% Ethanol, 3 minutes each - 3 changes of 100% Ethanol, 3 minutes each - 3 changes of Xylene, 5 minutes each - Mounted with Permount
- 1. Imaging: Leica SCN 400F Whole Slide Scanner with Digital Image Hub and Leica Slidepath software

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Experimental Notes:

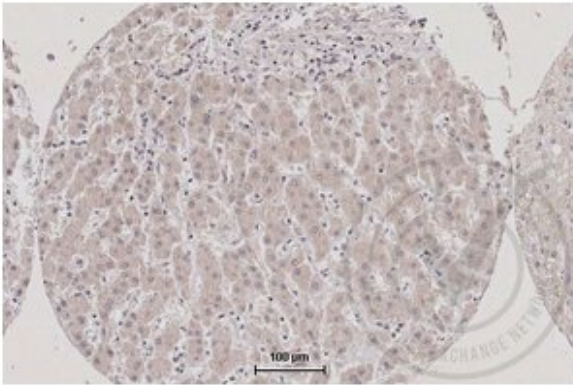
- Deviations from protocol/procedure supplied by manufacturer:
- Step 1: Heated tissue 60°C for 30 minutes; manufacturer heats for 45 minutes.
- Step 2: No ethanol wash was performed during deparaffinization; manufacturer includes 1 wash of 80% ethanol for 3 minutes.
- Step 3.1: Slides heated for 15 minutes; manufacturer provides a range of 15-20 minutes.
- Step 3.2: Slides cooled for 30 minutes; manufacturer cools for 20 minutes.
- Step 4: Italicized reagents and incubation time are fixed instrument parameters.
- Step 5: Secondary species-specific serum block not used; manufacturer blocks with 5% normal goat serum for 2 hours.
- Step 8.1: Antibody diluted in PBS at 1:250; manufacture recommends dilution range (1:100-1:500). No primary antibody diluent recommended.
- Step 8.2.1: Primary antibody incubated at room temperature overnight; manufacturer incubates overnight 4°C with agitation.
- Tissue Interpretation (limited): • TNFSF9: Under the staining parameters described above, placenta was weakly cytoplasmic positive (trophoblastic and decidua cells). Liver



hepatocytes were weakly stained. Substantial signal detected in limited number of other tissues, including kidney, prostate, and skin.

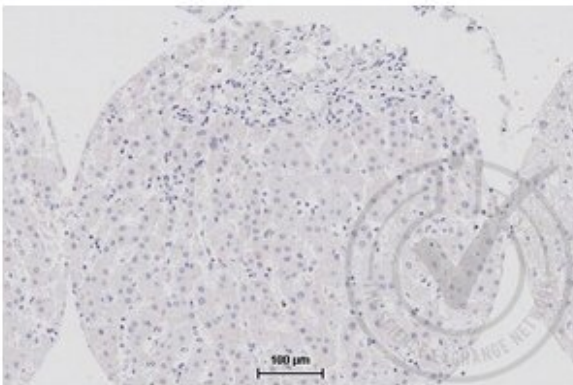
- I-NC (Isotype negative control): No signal detected
- B-NC (Blank negative control): No signal detected
- Signal Localization: - Signal to noise was adequate with predominately cytoplasmic subcellular localization observed. Nuclear staining was rare and no distinct membrane signal was detected.

## Images for Validation report #029578



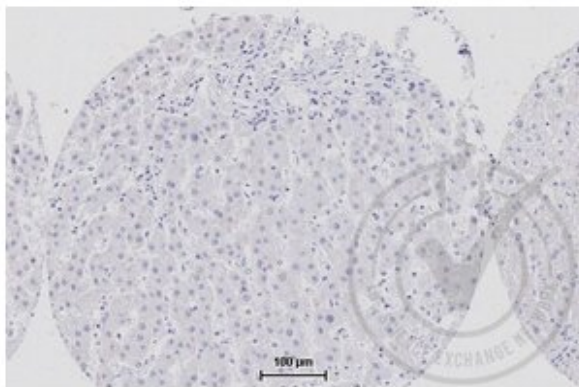
**Validation image no. 1 for anti-Tumor Necrosis Factor (Ligand) Superfamily, Member 9 (TNFSF9) (AA 101-200) antibody (ABIN705056)**

Figure 1: TNFSF9 staining of human liver (brown). Counterstain in blue.



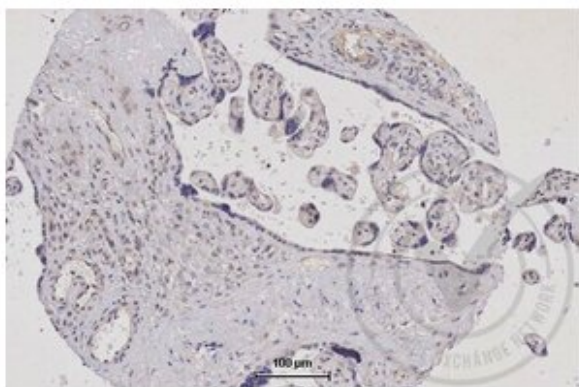
**Validation image no. 2 for anti-Tumor Necrosis Factor (Ligand) Superfamily, Member 9 (TNFSF9) (AA 101-200) antibody (ABIN705056)**

Figure 2: Isotype control staining of human liver (brown). Counterstain in blue.



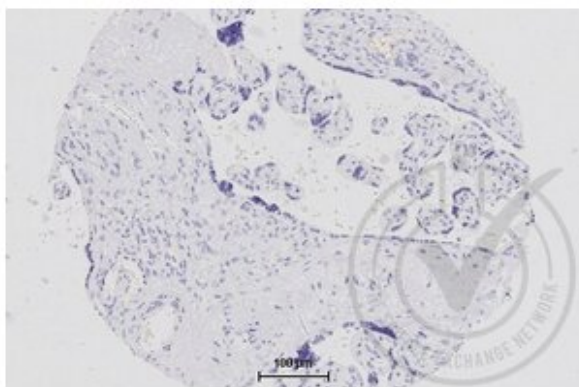
**Validation image no. 3 for anti-Tumor Necrosis Factor (Ligand) Superfamily, Member 9 (TNFSF9) (AA 101-200) antibody (ABIN705056)**

Figure 3: Secondary only staining of human liver (brown). Counterstain in blue.



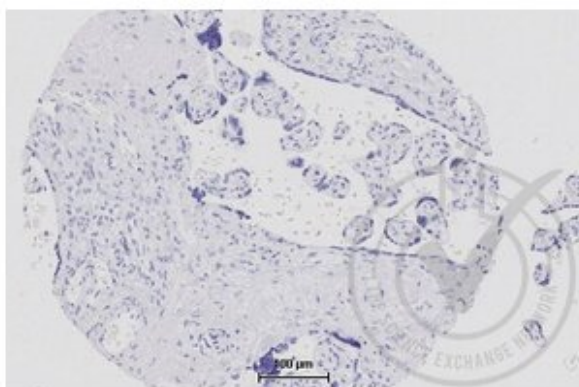
**Validation image no. 4 for anti-Tumor Necrosis Factor (Ligand) Superfamily, Member 9 (TNFSF9) (AA 101-200) antibody (ABIN705056)**

Figure 4: TNFSF9 staining of human placenta (brown). Counterstain in blue.



**Validation image no. 5 for anti-Tumor Necrosis Factor (Ligand) Superfamily, Member 9 (TNFSF9) (AA 101-200) antibody (ABIN705056)**

Figure 5: Isotype control staining of human placenta (brown). Counterstain in blue.



**Validation image no. 6 for anti-Tumor Necrosis Factor (Ligand) Superfamily, Member 9 (TNFSF9) (AA 101-200) antibody (ABIN705056)**

Figure 6: Secondary only staining of human placenta (brown). Counterstain in blue.