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Datasheet for ABIN5692886

## anti-TFF3 antibody (AA 23-81)

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### Overview

Quantity:	100 µg
Target:	TFF3
Binding Specificity:	AA 23-81
Reactivity:	Mouse, Rat
Host:	Rabbit
Clonality:	Polyclonal
Conjugate:	This TFF3 antibody is un-conjugated
Application:	Western Blotting (WB), ELISA, Immunohistochemistry (IHC)

### Product Details

Brand:	Picoband™
Immunogen:	E. coli-derived mouse TFF3 recombinant protein (Position: A23-F81).
Cross-Reactivity (Details):	No cross reactivity with other proteins.
Characteristics:	Rabbit IgG polyclonal antibody for TFF3 detection. Tested with WB, IHC-P, Direct ELISA in Mouse,Rat.

### Target Details

Target:	TFF3
Alternative Name:	Tff3 ( <a href="#">TFF3 Products</a> )
Background:	Synonyms: Trefoil factor 3, Intestinal trefoil factor, mITF, Tff3, Itf

## Target Details

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Tissue Specificity: Expressed in goblet cells of the intestines and colon (at protein level). Expressed abundantly in goblet cells of intestine and colon, and at low levels in stomach. No expression in brain, lung, spleen, kidney, uterus, pancreas, liver, heart or thymus.

Background: Trefoil factor 3 is a protein that in humans is encoded by the TFF3 gene. It is mapped to 21q22.3. Members of the trefoil family can interact with mucins and have an influence on mucus viscosity. They also promote migration of epithelial cells, are linked to antiapoptosis, induce cell scattering, trigger chemotaxis, and participate in immune responses. This gene is a marker of columnar epithelium and is expressed in a variety of tissues including goblet cells of the intestines and colon. It has been found that TFF3 is the most highly upregulated gene in 19 poorly differentiated endometrioid endometrial carcinomas (G3-EECs) compared with normal endometrium biopsies.

UniProt: [Q62395](#)

## Application Details

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Application Notes: Recommended Detection Systems: Enhanced Chemiluminescent Kit with anti-Rabbit IgG (ABIN921124) for Western blot, and HRP Conjugated anti-Rabbit IgG Super Vision Assay Kit (SV0002-1) for IHC(P).

Application Details: Western blot, 0.1-0.5 µg/mL  
Immunohistochemistry(Paraffin-embedded Section), 0.5-1 µg/mL  
Direct ELISA, 0.1-0.5 µg/mL

Restrictions: For Research Use only

## Handling

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Format: Lyophilized

Reconstitution: Add 0.2 mL of distilled water will yield a concentration of 500 µg/mL.

Buffer: Each vial contains 4 mg Trehalose, 0.9 mg NaCl, 0.2 mg Na<sub>2</sub>HPO<sub>4</sub>, 0.05 mg NaN<sub>3</sub>.

Preservative: Sodium azide

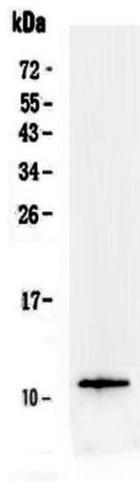
Precaution of Use: This product contains Sodium azide: a POISONOUS AND HAZARDOUS SUBSTANCE which should be handled by trained staff only.

Storage: 4 °C, -20 °C

Storage Comment: At -20°C for one year. After reconstitution, at 4°C for one month.  
It can also be aliquotted and stored frozen at -20°C for a longer time. Avoid repeated freezing

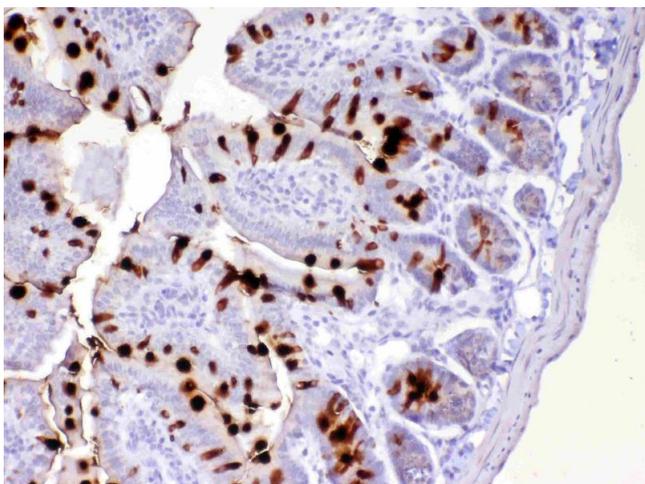
and thawing.

## Images



### Western Blotting

**Image 1.** Western blot analysis of TFF3 using anti-TFF3 antibody. Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each Lane was loaded with 50µg of sample under reducing conditions. Lane 1: mouse small intestine tissue lysates. After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-TFF3 antigen affinity purified polyclonal antibody (Catalog # ) at 0.5 µg/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for TFF3 at approximately 12KD. The expected band size for TFF3 is at 9KD.



### Immunohistochemistry

**Image 2.** IHC analysis of TFF3 using anti-TFF3 antibody. TFF3 was detected in paraffin-embedded section of mouse small intestine tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1µg/ml rabbit anti-TFF3 Antibody overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC)(Catalog

# SA1022) with DAB as the chromogen.



### Successfully validated (Western Blotting (WB))

by [Laboratory for neurodegenerative disease research, Department of Molecular Medicine, Ruđer Bošković Institute, Zagreb, Croatia](#)

Report Number: 103596

Date: Feb 18 2019

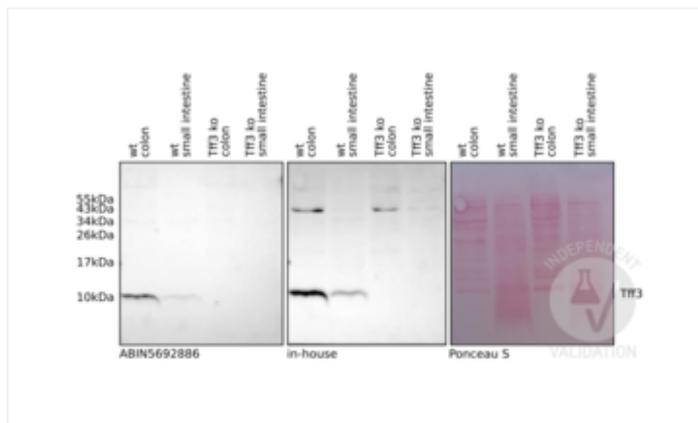
Target:	Tff3
Lot Number:	0011712Da513858
Method validated:	Western Blotting (WB)
Positive Control:	colon from WT C57BL/6 mice
Negative Control:	small intestine from Tff3 deficient mice
Notes:	ABIN5692886 specifically recognizes Tff3 in colon and small intestine tissue of wild type C57BL/6 mice.
Primary Antibody:	ABIN5692886
Secondary Antibody:	goat anti-rabbit IgG-HRP antibody (Bio-Rad, 170-6515, L005679 A)
Protocol:	<ul style="list-style-type: none"><li>• Isolate colon and small intestine tissue of wild type C57BL/6 mice and small intestine of Tff3 deficient mice using RIPA buffer (50mM TRIS HCL, pH8, 150mM NaCl, 1mM EDTA, 1% NP40 (Sigma-Aldrich, I-3021), 1% sodium deoxycholate (Sigma-Aldrich, D6750-25g), 0.1% SDS) containing PhosSTOP phosphatase inhibitor (Roche, 04906845001, lot 27423300) and cOmplete Mini protease inhibitor (Roche, lot 25765800).</li><li>• Determine total protein concentration using Pierce BCA protein assay kit (ThermoFisher Scientific, 23227, lot Tf268093).</li><li>• Denature 30µg total protein for 5min at 95°C in 6µl 5x Laemmli SDS sample buffer, 3µl 3M DTT and 1.5ul β-mercaptoethanol and Milli-Q H<sub>2</sub>O in 30µl total volume per sample.</li><li>• Separate proteins on a freshly cast reducing 15% SDS-PAGE gel at 100V for the stacking gel and 150V for the running gel for 2-3 hours in a Mini-PROTEAN Tetra Vertical Electrophoresis Cell chamber (Bio-Rad).</li><li>• Transfer proteins onto a nitrocellulose membrane using Trans-Blot Transfer Medium (Bio-Rad, 162-0112, lot BR 8158009) on a Trans-Blot Turbo Transfer System (Bio-Rad, 1704150) at 150mA for 90min.</li><li>• Stain membrane using Ponceau S to verify protein transfer.</li><li>• Wash membrane with TBS.</li><li>• Block the membrane with 1x TBS, 5% non-fat Milk, 0.1% Tween) for 1h at RT.</li><li>• Incubate membrane with primary<ul style="list-style-type: none"><li>◦ polyclonal rabbit anti-TFF3 antibody (antibodies-online, ABIN5692886, lot 0011712Da513858) diluted 1:1000 in 1x TBS, 5% BSA, 0.1% Tween ON at 4°C.</li></ul></li></ul>

## Validation report #103596 for Western Blotting (WB)

- polyclonal rabbit anti-TFF3 antibody (in-house) diluted in 1x TBS, 5% BSA, 0.1% Tween ON at 4°C.
- Wash membrane 3x for 10min with TBST (1x TBS, 0.1% Tween).
- Incubate membrane with secondary goat anti-rabbit IgG-HRP antibody (Bio-Rad, 170-6515, L005679 A) diluted 1: 5000 in TBST for 1.5h at RT.
- Wash membrane 3x for 10min with TBST.
- Reveal proteins bands using Pierce ECL Western Blotting Substrate (ThermoFisher Scientific, 32106, lot NJ175924) on an Imager Alliance 4.7 western blot imaging system (Uvitec).

Experimental Notes: ABIN5692886 reveals a protein of the expected molecular weight of the antigen in mice tissue. The protein bands is only visible in the positive but not the negative control. The same protein band was also detected by an in-house anti-TFF3 antibody hat was used as a reference.

## Image for Validation report #103596



### Validation image no. 1 for anti-Trefoil Factor 3 (Intestinal) (TFF3) (AA 23-81) antibody (ABIN5692886)

ABIN5692886 specifically recognizes Tff3 in colon and small intestine tissue of wild type C57BL/6 mice.