# Anti-IP10 Antibody [JA10-82]

## ET1704-27



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human
Applications:	WB, IF-Cell, IF-Tissue, IHC-P
Molecular Wt:	Predicted band size: 11 kDa
Clone number:	JA10-82
Description:	Chemokines are members of a superfamily of inducible, secreted, pro-inflammatory cytokines. Members of the chemokine family exhibit 20% to 50% homology in their predicted amino acid sequences and are divided into four subfamilies: C-C, C-X-C, C and C-X3-C. In the C-X-C or $\alpha$ subfamily, the first two of four cysteine motifs are separated by another amino acid residue. In the second subfamily, designated C-C or $\beta$ , the first cysteines are adjacent. C subfamily members, also designated $\gamma$ chemokines, lack the first and third cysteine residues of the conserved motif. In the C-X3-C, or $\delta$ subfamily, members have three amino acids between the two cysteines. The C-X-C chemokine subfamily includes IL-8, GRO $\alpha/\beta/\gamma$ (and the murine homologs KC, MIP-2 $\alpha$ and MIP-2 $\beta$ ), platelet basic protein, ENA-78, GCP-2, PF4, IP-10 (and its murine homolog, CRG) and MIG.
lmmunogen:	Synthetic peptide within Human IP10 aa 49-98 / 98.
Positive control:	THP-1 treated with 100ng/mL IFN- $\gamma$ for 24 hours then 300ng/mL Brefeldin A for 20 hours cell lysate, THP-1 treated with 200ng/mL IFN- $\gamma$ and 50ng/mL LPS for 24 hours cell lysate, HepG2, SH-SY5Y, human skin tissue, human tonsil tissue.
Subcellular location:	Secreted.
Database links:	SwissProt: P02778 Human
Recommended Dilutions: WB IF-Cell IF-Tissue IHC-P	1:1,000 1:50-1:100 1:50-1:1,000
Storage Buffer:	1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Store at +4 $^\circ\!C$ after thawing. Aliquot store at -20 $^\circ\!C$ or -80 $^\circ\!C$ . Avoid repeated freeze / thaw cycles.
Purity:	Protein A affinity purified.

# Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880

Technical:0086-571-89986345

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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

#### Images



**Fig1:** Western blot analysis of IP10 on different lysates with Rabbit anti-IP10 antibody (ET1704-27) at 1/1,000 dilution.

Lane 1: THP-1 cell lysate

Lane 2: THP-1 treated with 100ng/mL IFN- $\gamma$  for 24 hours then 300ng/mL Brefeldin A for 20 hours cell lysate Lane 3: THP-1 cell lysate Lane 4: THP-1 treated with 200ng/mL IFN- $\gamma$  and 50ng/mL LPS for 24 hours cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 11 kDa Observed band size: 11 kDa

Exposure time: 3 minutes;

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 (ET1704-27) at 1/1,000 dilution was used in 5% NFDM/TBST at  $4^{\circ}$ C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.



**Fig2:** ICC staining of IP10 in HepG2 cells (red). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 10% negative goat serum for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1704-27, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®594 conjugate-Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).



**Fig3:** ICC staining of IP10 in SH-SY5Y cells (red). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 10% negative goat serum for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1704-27, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®594 conjugate-Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

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**Fig4:** Immunohistochemical analysis of paraffin-embedded human skin tissue using anti-IP10 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes.The tissues were blocked in 1% BSA for 30 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1704-27, 1/50) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

**Fig5:** Immunohistochemical analysis of paraffin-embedded human tonsil tissue with Rabbit anti-IP10 antibody (ET1704-27) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1704-27) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

**Fig6:** Immunocytochemistry analysis of THP-1 cells treated with 200ng/mL IFN- $\gamma$  then treated with 50ng/mL LPS for 24 hours labeling IP10 with Rabbit anti-IP10 antibody (ET1704-27) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-IP10 antibody (ET1704-27) at 1/100 dilution in 1% BSA in PBST overnight at 4  $^{\circ}$ C. Goat Anti-Rabbit IgG H&L (iFluor<sup>TM</sup> 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

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

#### **Background References**

- 1. Sindhu S et al. Obesity Is a Positive Modulator of IL-6R and IL-6 Expression in the Subcutaneous Adipose Tissue: Significance for Metabolic Inflammation. PLoS One 10:e0133494 (2015).
- 2. Muthuswamy R et al. Combination of IFNa and poly-I:C reprograms bladder cancer microenvironment for enhanced CTL attraction. J Immunother Cancer 3:6 (2015).



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