

Anti-CD66b Antibody

HA500100



Product Type:	Rabbit polyclonal IgG, primary antibodies
Species reactivity:	Human
Applications:	WB, IHC-P, mIHC, IF-Tissue
Molecular Wt:	Predicted band size: 38 kDa.

Description:	Carcinoembryonic antigen-related cell adhesion molecule 8, also designated CD67, CD66b or nonspecific cross-reacting antigen (NCA-95), belongs to the human carcinoembryonic antigen (CEA) family. The CD67 antigen is encoded by the CEACAM8 (CGM6) gene, which is exclusively expressed in neutrophils and eosinophils. In neutrophils, the CEACAM8 gene is primarily detected in the secondary granules within the cytoplasm, but it can also be found in lower amounts on the plasma membrane. The amount of CD67 on the plasma membrane is up-regulated upon granulocyte activation. CD67 has been located on the surface of neutrophilic and eosinophilic granulocytes at late stages of differentiation. It exhibits heterophilic cell adhesion properties with CD66c, which is coexpressed with CD67 in granulocytes. CD67, which is attached to the membrane by a GPI-anchor, is expressed in leukocytes of chronic myeloid leukemia patients and bone marrow and in granulocytes in the spleen, thymus and lungs.
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Immunogen: Recombinant protein within human CD66b aa 1-250.

Positive control: Human cervical cancer, human colon cancer tissue, human breast cancer tissue, human spleen tissue, human colon tissue, U937 cell lysate, A431 cell lysate, Daudi cell lysate.

Subcellular location: Cell membrane, Cell surface.

Database links: SwissProt: P31997 Human

Recommended Dilutions:

IHC-P	1:1,000
mIHC	1:1,000
IF-Tissue	1:200

Storage Buffer: 1*TBS (pH7.4), 0.2% BSA, 50% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Shipped at 4°C. Store at +4°C short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20°C long term.

Purity: Immunogen affinity purified.

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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

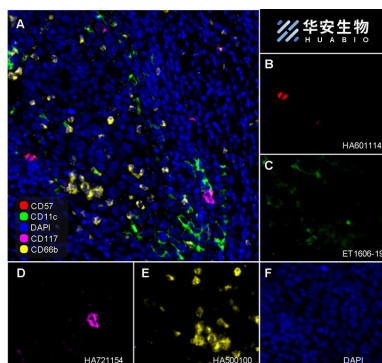


Fig1: Fluorescence multiplex immunohistochemical analysis of the human cervical cancer (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-CD57 (HA601114, red), anti-CD11c (ET1606-19, green), anti-CD117 (HA21154, magenta) and anti-CD66b (HA500100, yellow) on human cervical cancer. Panel B: anti- CD57 stained on NKT cells. Panel C: anti-CD11c stained on dendritic cells. Panel D: anti-CD117 stained on mast cells. Panel E: anti-CD66b stained on neutrophils. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with the Sequential Immuno-staining Kit (IRISKit™MH010101, www.luminiris.cn). The section was incubated in four rounds of staining: in the order of HA601114 (1/500 dilution), ET1606-19 (1/1,000 dilution), HA721154 (1/1,000 dilution), and HA500100 (1/1,000 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95°C. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Olympus VS200 Slide Scanner.

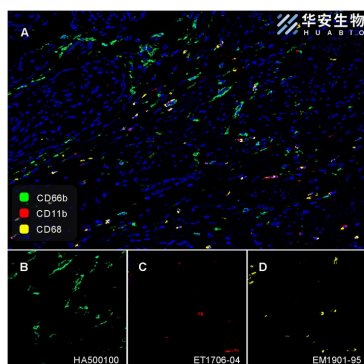


Fig2: Fluorescence multiplex immunohistochemical analysis of human cervical carcinoma (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-CD66b (HA500100, Green), anti-CD11b (ET1706-04, Red) and anti-CD68 (EM1901-95, Yellow) on human cervical carcinoma. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with the Sequential Immuno-staining Kit (IRISKit™MH010101, www.luminiris.cn). The section was incubated in three rounds of staining: in the order of HA500100 (1/1,000 dilution), ET1706-04 (1/1,000 dilution) and EM1901-95 (1/3,000 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95°C. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Zeiss Observer 7 Inverted Fluorescence Microscope.

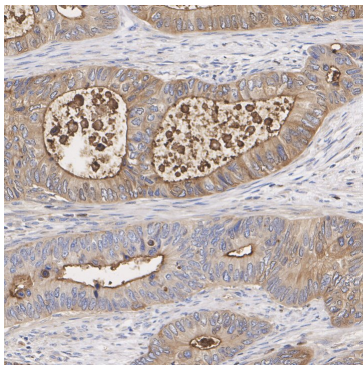


Fig3: Immunohistochemical analysis of paraffin-embedded human colon cancer tissue with Rabbit anti-CD66b antibody (HA500100) at 1/1,000 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (HA500100) 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.

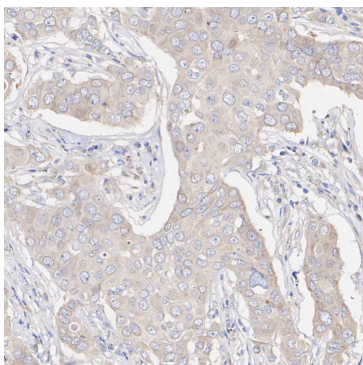


Fig4: Immunohistochemical analysis of paraffin-embedded human breast cancer tissue with Rabbit anti-CD66b antibody (HA500100) at 1/1,000 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (HA500100) 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.

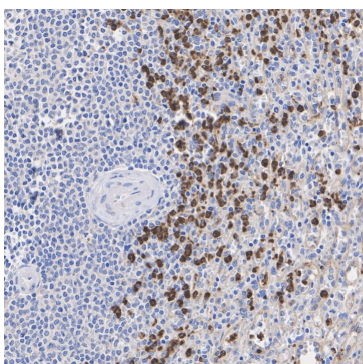


Fig5: Immunohistochemical analysis of paraffin-embedded human spleen tissue with Rabbit anti-CD66b antibody (HA500100) at 1/1,000 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (HA500100) 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.

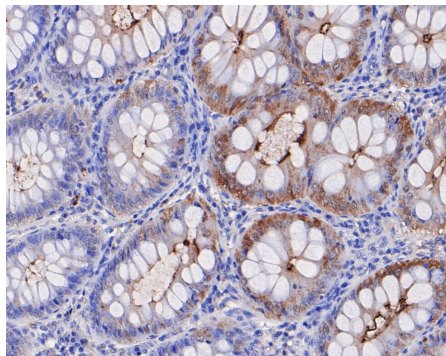


Fig6: Immunohistochemical analysis of paraffin-embedded human colon tissue using anti-CD66b antibody. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA500100, 1/400) 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.

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

Background References

1. Posabella A. et. al. High density of CD66b in primary high-grade ovarian cancer independently predicts response to chemotherapy. J Cancer Res Clin Oncol. 2020 Jan
2. Huang X. et. al. Prognostic significance of the infiltration of CD163(+) macrophages combined with CD66b(+) neutrophils in gastric cancer. Cancer Med. 2018 May

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