Anti-Lambda Light Chain Antibody [A8G5]

HA601055

Product Type:	Mouse monoclonal IgG2b, primary antibodies
Species reactivity:	Human
Applications:	IHC-P, WB, IF-Cell, FC
Molecular Wt:	Predicted band size: 11 kDa
Clone number:	A8G5

Description: While benign (reactive) B-lymphocytic populations produce Ig-molecules containing an almost equal amount of kappa and lambda light chains, i.e. the number of cells producing kappa is more or less equal to the number producing lambda, neoplastic B-lymphocytic populations has light chain restriction (i.e., are monoclonal) producing either kappa or lambda. Consequently, demonstration of light chains is the most important procedure in the diagnosis of neoplasms of B-lymphocytes (lymphomas and leukemias). The amount of Ig produced in individual types of B-lymphocytic neoplasia vary and is often small, demanding a sensitive technique for detection. About 80% of non-Hodgkin lymphomas are B-cell lymphomas, of which the large majority express surface (s) IgM, although the expression in small cell lymphoma/chronic lymphocytic leukaemia is weak. Plasmacytoma/multiple myeloma do not show sIgM. Lymphoplasmacytic lymphoma most often shows sIgM, while plasmacytoma/multiple myeloma show sIgG in 50% and sIgA in 20%. Light chain restriction is the single most important marker for neoplasms of B-lymphocytic origin. Demonstration of heavy chains can sometime be of aid in the study of malignant lymphomas, as lymphoplasmacytic lymphomas are usually focally IgM positive, while plasmacytoma/multiple myeloma in most cases express strong cytoplasmic IgG or IgA. Precursor B-cell neoplasms are are Ig negative. Normal tonsil is appropriate control tissue: approximately 50% of the mantle zone B-cells should show a distinct membrane staining reaction, while the rest should be unstained.

Immunogen:	Constant region of natural protein (constant region of light chain lambda chain).
Positive control:	Human small intestine tissue lysates, human plasma lysates, human tonsil tissue, Ramos.
Subcellular location:	Secreted, Cell membrane.
Database links:	SwissProt: P0CG04 Human

Recommended Dilutions:

IHC-P	1:4,000
WB	1:1,000
IF-Cell	1:100
FC	1:1,000

Storage Buffer:	PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% SodiumAzide.
Storage Instruction:	Store at +4℃ after thawing. Aliquot store at -20℃. Avoid repeated freeze / thaw cycles.
Purity:	Protein A affinity purified.

Images

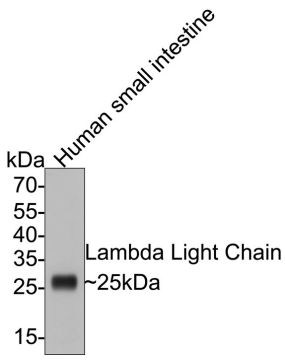

Fig1: Western blot analysis of Lambda Light Chain on human small intestine tissue lysates with Mouse anti-Lambda Light Chain antibody (HA601055) at 1/1,000 dilution.

Lysates/proteins at 20 µg/Lane.

Predicted band size: 11 kDa
Observed band size: 25 kDa

Exposure time: 30 seconds;

12% 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 (HA601055) at 1/1,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1:100,000 dilution was used for 1 hour at room temperature.

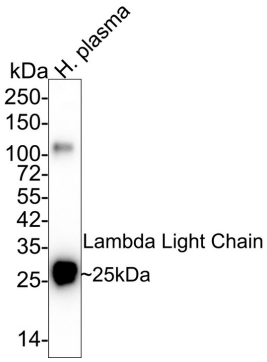

Fig2: Western blot analysis of Lambda Light Chain on human plasma lysates with Mouse anti-Lambda Light Chain antibody (HA601055) at 1/1,000 dilution.

Lysates/proteins at 30 µg/Lane.

Predicted band size: 11 kDa
Observed band size: 25 kDa

Exposure time: 43 seconds;

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 (HA601055) at 1/1,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Anti-Mouse IgG for IP Nano-secondary antibody (NBI02H) at 1/5,000 dilution was used for 1 hour at room temperature.

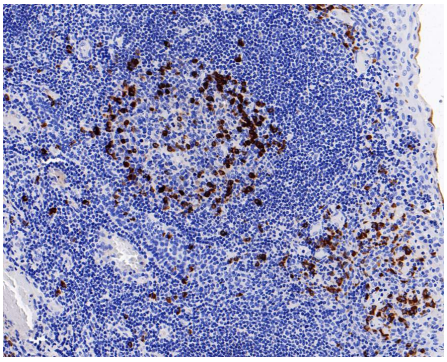

Fig3: Immunohistochemical analysis of paraffin-embedded human tonsil tissue with Mouse anti-Lambda Light Chain antibody (HA601055) at 1/4,000 dilution.

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_2O and PBS, and then probed with the primary antibody (HA601055) at 1/4,000 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.

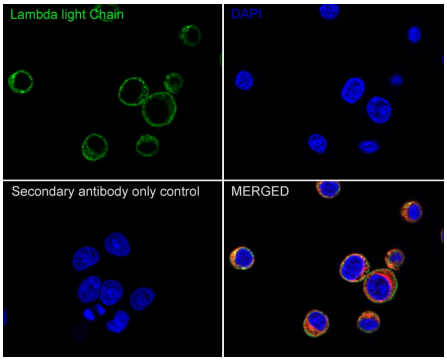

Fig4: Immunocytochemistry analysis of Ramos cells labeling Lambda Light Chain with Mouse anti-Lambda Light Chain antibody (HA601055) at 1/100 dilution.

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 Mouse anti-Lambda Light Chain antibody (HA601055) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) 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.

beta Tubulin (ET1602-4, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) were used as the secondary antibody at 1/1,000 dilution.

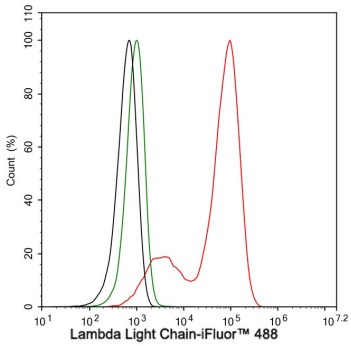

Fig5: Flow cytometric analysis of Ramos cells labeling Lambda Light Chain.

Cells were washed twice with cold PBS and resuspend. Then stained with the primary antibody (HA601055, 1μg/mL) (red) compared with Mouse IgG1 Isotype Control (green). After incubation of the primary antibody at +4℃ for 30 minutes, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Mouse IgG Secondary antibody (HA1125) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Background References

1. Kato A. et. al. An expanded genetic code facilitates antibody chemical conjugation involving the lambda light chain. Biochem Biophys Res Commun. 2021 Mar
2. van der Kant R. et. al. Adaption of human antibody λ and κ light chain architectures to CDR repertoires. Protein Eng Des Sel. 2019 Dec