

## PRODUCT DATA SHEET

**Product:** Anti-Fas Ligand mAb, clone H11

**Cat. No.:** MC-106 (100 µg)

**Background:**

Fas Ligand (FasL) is a type II membrane protein and a member of the tumor necrosis factor family. FasL is released by cleavage by metalloproteinases. When interacting with the Fas receptor, the trimeric FasL induces apoptosis. It is responsible for the activation-induced cell death of peripheral T cells and functions as an effector molecule for cytotoxic T cells. FasL is expressed in activated lymphocytes, Sertoli cells, and in certain tumor cells.

**Species Reactivity:**

Mouse. Others not tested.

**Ig Isotype:**

Rat IgG<sub>2a</sub>

**Immunogen:**

Synthetic peptide corresponding to aa 196-220 of the mouse Fas Ligand protein (CD95L; APO-1L; CD178).

**Format:**

100 µL of 1 mg/mL antibody in PBS with 0.02% sodium azide.

**Purity:**

≥ 95% by SDS-PAGE.

**Storage:**

Store undiluted at 4°C. Do not freeze/thaw.

**Applications and Suggested Dilutions:**

- Immunocytochemistry: Use anti-Fas Ligand, clone H11 unlabeled (Cat. No. MC-106), biotin-labeled (Cat. No. MC-126), or FITC-labeled (Cat. No. MC-136).
- Flow Cytometry: Use Anti-Fas Ligand, clone H11 unlabeled (Cat. No. MC-106), biotin-labeled (Cat. No. MC-126), or FITC-labeled (Cat. No. MC-136). [Note: Activated T cells do not express high levels of surface Fas Ligand.]
- Western Blot: *Excellent for this application.* Use 2 µg/mL. Use unlabeled (Cat. No. MC-106) or biotin-labeled (Cat. No. MC-126).

The optimal dilution for a specific application should be determined by the researcher.

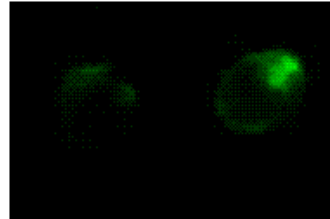


Figure 1. Immunocytochemistry: Detection of FasL in 293T cells transfected with a FasL expression vector.

Method: Transfected cells were plated onto polylysine treated glass slides, fixed and permeabilized in methanol at -20°C for 5 min, then in acetone at -20°C for 30 sec. After 3 washes in PBS, 0.1% BSA, slides were incubated for 1 hr at RT with 20 µg/mL of biotinylated H11 antibody (MC-126) in PBS, 0.1% BSA. After rinsing in PBS, FITC-conjugated streptavidin was added for 30 min. Slides were washed again in PBS and visualized using a fluorescence microscope.

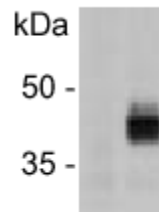


Figure 2. Western blot: Detection of FasL in 293T cells transfected with a FasL expression plasmid (right panel). Mock-transfected cells (left panel).

Method: Cell extracts from cells (2x10<sup>6</sup>) transfected with a FasL-expression plasmid were resolved by SDS-PAGE under reducing conditions, transferred to nitrocellulose and probed with the H11 antibody at 2 µg/mL. Proteins were visualized using a peroxidase-conjugated antibody to rat IgG and a chemiluminescence detection system.

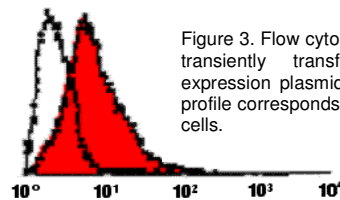


Figure 3. Flow cytometric profile of 293T cells transiently transfected with a FasL-expression plasmid (filled profile). The open profile corresponds to mock-transfected 293T cells.

Method: FasL transfected 293T cells (5x10<sup>5</sup>) were incubated on ice for 30 min in 50 µL FACS buffer (PBS, 5% fetal calf serum, 0.02% sodium azide) containing 1 µg of FITC-labeled H11 antibody (MC-136). After washing in FACS buffer, cells were analyzed by flow cytometry. Do not exclude pre-apoptotic cells during data acquisition.

**Limitations:**

For *in vitro* research use only. Not for use in diagnostics or in humans.

**Warranty:**

No warranties, expressed or implied, are made regarding the use of this product. KAMIYA BIOMEDICAL COMPANY is not liable for any damage, personal injury, or economic loss caused by this product.