

MONOCLONAL ANTIBODY

# FITC labeled CD120b/Anti-TNF-R2

Code No.	Clone	Subclass	Quantity	Concentration
K0040-4	80M2	Mouse IgG1	1 mL	50 µg/mL

**SOURCE:** This antibody was purified from hybridoma (clone 80M2) supernatant using protein A agarose. This hybridoma was established by fusion of mouse myeloma cell NSO with Balb/c mouse splenocyte immunized with the recombinant TR80 receptor material.

**FORMULATION:** 50 µg IgG in 1 mL volume of PBS containing 1% BSA and 0.09% NaN<sub>3</sub>.

\*Azide may react with copper lead in plumbing system to form explosive metal azides. Therefore, always flush plenty of water when disposing materials containing azide into drain.

**STORAGE:** This antibody solution is stable for one year from the date of purchase when stored at 4°C.

**REACTIVITY:** This antibody reacts with CD120b/human TNF-R2 antigen on Flow cytometry.

**APPLICATIONS:**

- Western blotting; Not tested
- Immunoprecipitation; Not tested
- Immunohistochemistry; Not tested
- Immunocytochemistry; Not tested
- Flow cytometry; 10-20 µg/mL (final concentration)

Detailed procedure is provided in the following **PROTOCOL**.

**SPECIES CROSS REACTIVITY:**

Species	Human	Mouse	Rat
Cell	U937	Not Tested	Not Tested
Reactivity on FCM	+		

**INTENDED USE:**

For Research Use Only. Not for use in diagnostic procedures.

**REFERENCES:**

- 1) Rae, C., *et al.*, *Cell Death Differ.* **11**, S162-S171 (2004)
- 2) Grell, M., *et al.*, *Cell* **83**, 793-802 (1995)
- 3) Grell, M., *et al.*, *Lymphokine Cytokine Res.* **12**, 143-148 (1993)

Clone 80M2 is used in the reference 2) and 3).

**RELATED PRODUCTS:**

- K0039-3 Anti-TNF-R1 (H398)
- K0039-4 FITC labeled Anti-TNF-R1 (H398)
- K0040-3 Anti-TNF-R2 (80M2)

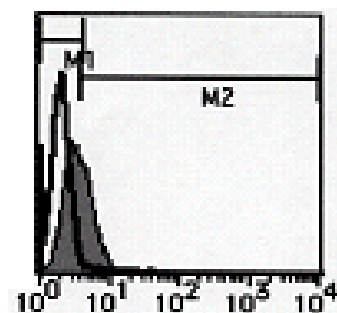
**PROTOCOL:**

**Flow cytometric analysis for floating cells**

We usually use Fisher tubes or equivalents as reaction tubes for all step described below.

- 1) Wash the cells 3 times with washing buffer [PBS containing 2% fetal calf serum (FCS) and 0.1% NaN<sub>3</sub>].
- 2) Resuspend the cells with washing buffer (5x10<sup>6</sup> cells/mL).
- 3) Add 50 µL of the cell suspension into each tube, and centrifuge at 500 x g for 1 minute at room temperature (20~25°C). Remove supernatant by careful aspiration.
- 4) Add 10 µL of normal goat serum containing 1 mg/mL normal human IgG and 0.1% NaN<sub>3</sub> to the cell pellet after tapping. Mix well and incubate for 5 minutes at room temperature.
- 5) Add 40 µL of the FITC labeled CD120b/anti-TNF-R2 (80M2) (25-50 µg/mL) diluted with the washing buffer. Mix well and incubate for 30 minutes at room temperature.
- 6) Add 1 mL of the washing buffer followed by centrifugation at 500 x g for 1 minute at room temperature. Remove supernatant by careful aspiration.
- 7) Resuspend the cells with 500 µL of the washing buffer and analyze by a flow cytometer.

(Positive control for flow cytometry; U937)



**Flow cytometric analysis of TNF-R2 expression on U937 cells.** Open histogram indicates the reaction of Isotypic control to the cells. Shaded histogram indicates the reaction of K0040-4 to the cells.

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