

MONOCLONAL ANTIBODY

# Anti-CPM (Mouse)

Code No.	Clone	Subclass	Quantity	Concentration
D293-3	40-1	Rat IgG2a	100 $\mu$ L	1 mg/mL

**BACKGROUND:** Carboxypeptidase M (CPM) is a 62 kDa of glycosylphosphatidylinositol (GPI)-anchored plasma membrane enzyme catalyses the removal of carboxy-terminal basic amino acids, such as arginine and lysine. CPM is different from pancreatic carboxypeptidase A and B, human plasma carboxypeptidase N and carboxypeptidase H in its enzyme structurally, catalytically and immunologically. CPM is widely distributed in human tissues and often highly expressed in epithelial cells.

**SOURCE:** This antibody was purified from hybridoma (clone 40-1) supernatant using protein G agarose. This hybridoma was established by fusion of mouse myeloma cell P3X with Wistar rat lymphocyte immunized with mouse fetal hepatic cells.

**FORMULATION:** 100  $\mu$ g IgG in 100  $\mu$ L volume of PBS containing 50% glycerol, pH 7.2. No preservative is contained.

**STORAGE:** This antibody solution is stable for one year from the date of purchase when stored at -20°C.

**REACTIVITY:** This antibody reacts with mouse CPM on Flow cytometry.

**APPLICATION:**

Flow cytometry: 5  $\mu$ g/mL (final concentration)

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

**SPECIES CROSS REACTIVITY:**

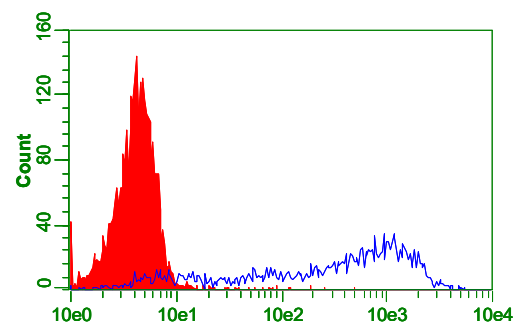
Species	Human	Mouse	Rat
Cell	Not Tested	Transfectant	Not Tested
Reactivity on FCM		+	

**INTENDED USE:**

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

**REFERENCES:**

- 1) McGwire, G. B., *et al.*, *J. Biol. Chem* **274**, 31632-31640 (1999)
- 2) Deddish, P. A., *et al.*, *J. Biol. Chem* **265**, 15083-15089 (1990)



**Flow cytometric detection of CPM (Mouse) on Ba/F3 transfectant**

Open: D293-3

Closed: isotype control

The descriptions of the following protocols are examples. Each user should determine the appropriate condition.

**PROTOCOL:**

**Flow cytometric analysis for floating cells**

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

- 1) Wash the cells 3 times with washing buffer [PBS containing 2% fetal calf serum (FCS)].
- 2) Resuspend the cells with washing buffer ( $5 \times 10^6$  cells/mL).
- 3) Add 100  $\mu$ L 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  $\mu$ L of normal goat serum to the cell pellet after tapping. Mix well and incubate for 5 minutes at room temperature.
- 5) Add 20  $\mu$ L of the primary antibody at the concentration as suggested in the **APPLICATION** diluted in 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) Add 40  $\mu$ L of 1:50 PE conjugated anti-rat IgG (MBL; code no. IM-1623) diluted with the washing buffer. Mix

well and incubate for 15 minutes at room temperature.

- 8) 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.
- 9) Resuspend the cells with 500  $\mu$ L of the washing buffer and analyze by a flow cytometer.

(Positive control for Flow cytometry; transfectant)

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