

MONOCLONAL ANTIBODY

# Anti-CD52 (CAMPATH-1) (Mouse) mAb-PE

Code No.	Clone	Subclass	Quantity
D204-5	BTG-2G	Rat IgG2a $\kappa$	50 tests

**BACKGROUND:** CAMPATH-1, also known as CD52, is a heavily glycosylated, GPI-anchored protein expressed at high levels on almost all thymocytes, lymphocytes, monocytes, and macrophages. CAMPATH-1 is an exceptionally good target for complement-mediated cell lysis and antibody-mediated cellular cytotoxicity. Humanized CAMPATH-1 antibodies have been used therapeutically to effectively deplete lymphocytes in allogeneic bone marrow transplants, hematologic malignancies, and autoimmune diseases. Clinical trials suggest CAMPATH-1 antibodies are especially promising in the treatment of leukemia, non-Hodgkin lymphomas, and rheumatoid arthritis. The apparent size of CAMPATH-1 by SDS-PAGE is 25-29 kDa; however, the actual molecule is much smaller, ~ 8-9 kDa as confirmed by total structure analysis and mass spectrometry.

**SOURCE:** This antibody was purified from hybridoma (clone BTG-2G) supernatant using protein G agarose. This hybridoma was established by fusion of mouse myeloma cell P3U1 with Wister rat splenocyte immunized with IL-2R $\alpha$ -transgenic mouse splenocyte.

**FORMULATION:** 50 tests in 1 mL volume of PBS containing 1% BSA and 0.1% ProClin 150.

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

**REACTIVITY:** This antibody reacts with mouse CD52 (CAMPATH-1) on Flow cytometry.

**APPLICATION:**

Flow cytometry; 20  $\mu$ L

\*Please refer to the data sheet (MBL; code no. D204-3) for other applications.

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

**SPECIES CROSS REACTIVITY:**

Species	Human	Mouse	Rat
Cell	Not tested	Splenocyte	Not tested
Reactivity on FCM		+	

**INTENDED USE:**

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

**REFERENCES:**

- 1) Kubota, H., *et al.*, *J. Immunol.* **145**, 3924-3931 (1990)

This antibody is used in this reference.

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

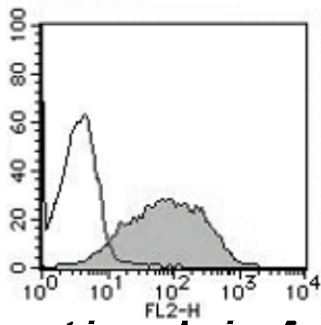
**PROTOCOL:**

**Flow cytometric analysis for floating cells**

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

- 1) Wash the cells 3 times with washing buffer [PBS containing 2% fetal calf serum (FCS) and 0.09% NaN<sub>3</sub>].  
\*Azide may react with copper or lead in plumbing system to form explosive metal azides. Therefore, always flush plenty of water when disposing materials containing azide into drain.
- 2) Resuspend the cells with washing buffer (5 x 10<sup>6</sup> cells/mL).
- 3) Add 50  $\mu$ 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 Clear Back (human Fc receptor blocking reagent, MBL; code no. MTG-001) to the cell pellet after tapping. Mix well and incubate for 5 minutes at room temperature.
- 5) Add the antibody as suggested in the **APPLICATION**. Mix well and incubate for 20 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  $\mu$ L of the washing buffer and analyze by a flow cytometer.

(Positive control for Flow cytometry; Mouse splenocyte)



**Flow cytometric analysis of mouse CD52 expression on mouse splenocyte.** Open histogram indicates the reaction of isotypic control to the cells. Shaded histogram indicates the reaction of D204-5 to the cells.

**RELATED PRODUCTS:**

Please visit our website at <https://ruo.mbl.co.jp/>.