

MONOCLONAL ANTIBODY

Anti-SynCAM (TSLC1/CADM1) (Human) mAb

Code No.	Clone	Subclass	Quantity	Concentration
CM005-3	9D2	Chicken IgY	100 μ L	1 mg/mL

BACKGROUND: SynCAM (Synaptic Cell Adhesion Molecule), also known as TSLC1 (tumor suppressor in lung cancer-1), SgIGSF, or CADM1, is a homophilic, transmembrane Ig-domain containing protein with intracellular PDZ protein-binding motifs. Although originally identified as a tumor suppressor of small lung cell carcinomas, SynCAM appears to be primarily involved in intracellular adhesion and synapse formation. The majority of SynCAM is localized to synaptic sites where it initiates synaptic assembly and synapse differentiation throughout the central nervous system. SynCAM also mediates the cellular adhesion of spermatogenic cells to Sertoli cells and mast cells to fibroblasts.

SOURCE: This antibody was purified from hybridoma (clone 9D2) supernatant using anti-IgY affinity column. This hybridoma was established by fusion of chicken B cell line MUH1 with chicken splenocyte immunized with the recombinant SynCAM/Fc fusion protein.

FORMULATION: 100 μ g IgG in 100 μ 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 extracellular domain of SynCAM on Flow cytometry.

APPLICATIONS:

Western blotting; Not recommended
Immunoprecipitation; Not recommended
Immunohistochemistry; Not tested
Immunocytochemistry; Not tested
Flow cytometry; 1 μ g/mL (final concentration)

*It is reported that clone 9D2 can be used in adhesion blocking experiments in the reference number 2) and 3).

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

INTENDED USE:

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

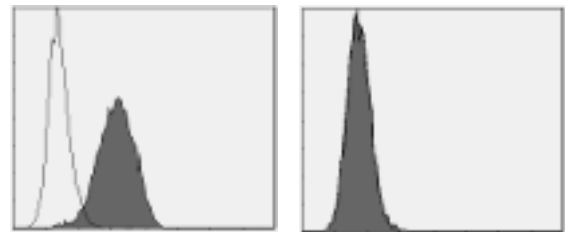
SPECIES CROSS REACTIVITY:

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

REFERENCES:

- 1) Hollins, F., *et al.*, *J. Immunol.* **181**, 2772-2780 (2008)
- 2) Yang, W., *et al.*, *J. Immunol.* **176**, 1238-1243 (2006)
- 3) Furuno, T., *et al.*, *J. Immunol.* **174**, 6934-6942 (2005)
- 4) Sara, Y., *et al.*, *J. Neuroscience* **25**, 260-270 (2005)
- 5) Biederer, T., *et al.*, *Science* **297**, 1525-1531 (2002)

Clone 9D2 is used in the reference number 1) - 3).



Flow cytometric analysis of SynCAM expression on L cells (right) and SynCAM transfected L cells (left). Open histogram indicates the reaction of isotypic control to the cells. Shaded histograms indicate the reaction of CM005-3 to the cells.

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) and 0.09% NaN₃].
*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 (5x10⁶ cells/mL).
- 3) Add 50 μ 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 20 μ 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 40 μ L of the primary antibody at the concentration as suggested in the **APPLICATIONS** 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 30 μ L of 1:100 FITC conjugated anti-chicken IgY (CHEMICON; code no. AP162F) 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 μ L of the washing buffer and analyze by a flow cytometer.

(Positive control for Flow cytometry; Transfectant)

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