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# TECHNICALLY Speaking

Place your order with CEDARLANE® or your local distributor.

*Please contact CEDARLANE® for lot specific information.*

## **Anti-Rat SIRP (Signal-Regulatory Protein) Monoclonal Antibody-Ascites**

**CL041A  
LOT: 4111**

### **DESCRIPTION:**

CL041A (MRC OX-41) monoclonal antibody recognizes rat SIRP (Signal-Regulatory Protein), a surface protein of about 110 kDa. SIRP is selectively expressed by myeloid cells (macrophages, monocytes, granulocytes, dendritic cells) and neurons. The SIRP antigen is a transmembrane glycoprotein with 3 immunoglobulin-like extracellular domains: an N-terminal V-set domain and two C1-set domains. The SIRP Ig domains are closely related to those of the antigen receptors, Ig, TCR and MHC. The selective expression of this antigen by myeloid and neuronal cells suggests that SIRP is involved in the modulation of myeloid and neuronal cell function.

This clone can be used in FACS analysis, immunofluorescence, immunocytochemistry, and indirect radioimmunoassays.

**PRESENTATION:** 0.5 ml, lyophilized ascites

### **STORAGE/STABILITY:**

Lyophilized form stable at 4°C or -20°C. Reconstitute with 0.5 ml of cold distilled water. After reconstitution, aliquot and freeze unused portions in volumes appropriate for single usage. Avoid repeated freeze/thaw cycles.

For more information or to place an order please contact...

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**SPECIFICATIONS:**

Clone: MRC OX-41

Hybridoma Production:

Immunization: Immunogen: Rat Peritoneal Macrophages  
Donor: BALB/c Spleen

Fusion Partner: NSO/U

Specificity: Rat SIRP (Signal-Regulatory Protein)

Ig Class: Mouse IgG<sub>2a</sub>

Format: Ascitic Fluid, filtered to 0.45μm and lyophilized.

**FLOW CYTOMETRY ANALYSIS:**

Method:

1. Prepare a cell suspension in media A. For cell preparations, deplete the red blood cell population with Lympholyte®-Rat cell separation medium (CL5040).
2. Wash 2 times.
3. Resuspend the cells to a concentration of 2x10<sup>7</sup> cells/ml in media A. Add 50 μl of this suspension to each tube (each tube will then contain 1x10<sup>6</sup> cells, representing 1 test).
4. To each tube, add 50 μl of a 1:5000-1:10000 dilution\* of **CL041A**.
5. Vortex the tubes to ensure thorough mixing of antibody and cells.
6. Incubate the tubes for 30 minutes at 4°C.
7. Wash 2 times at 4°C.
8. Add 100 μl of secondary antibody **CLCC30201** (FITC Goat anti-mouse IgG (H+L)) at 1:500 dilution.
9. Incubate the tubes at 4°C for 30-60 minutes.  
(It is recommended that the tubes are protected from light since most fluorochromes are light sensitive).
10. Wash 2 times at 4°C in media B.
11. Resuspend the cell pellet in 50 μl ice cold media B.
12. Transfer to suitable tubes for flow cytometric analysis containing 15 μl of propidium iodide at 0.5 mg/ml in PBS. This stains dead cells by intercalating in DNA.

Media:

- A. Phosphate buffered saline (pH 7.2) + 5% normal serum of host species + sodium azide (100 µl of 2M sodium azide in 100 mls).
- B. Phosphate buffered saline (pH 7.2) + 0.5% Bovine serum albumin + sodium azide (100 µl of 2M sodium azide in 100 mls).

Results:Tissue Distribution by Flow Cytometry Analysis:

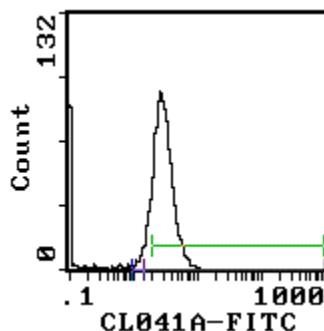
Mouse Strain: Fisher

Cell Concentration :  $1 \times 10^6$  cells per tests

Antibody Concentration: 1:10000 in 50 µl / $10^6$  cells

Isotypic Control: Purified Mouse IgG<sub>2a</sub>

| <u>Cell Source</u>     | <u>Percentage of cells stained above control:</u> |
|------------------------|---|
| Peritoneal Macrophages | 78.6%   |



Cell Source: Peritoneal Macrophages  
 Percentage of cells stained above control: 78.6%

**N.B. Appropriate control samples should always be included in any labelling studies.**

\* For optimal results in various applications, it is recommended that each investigator determine dilutions appropriate for individual use.

**REFERENCES:**

1. Robinson, A. P., White, T. M. and Mason, D. W. (1986) Macrophage heterogeneity in the rat as delineated by two monoclonal antibodies MRC OX-41 and MRC OX-42, the latter recognizing complement receptor type 3. *Immunology* **57**: 239-247.
2. Adams, S., van der Laan, L. J. W., Vernon-Wilson, E., Renardel de Lavalette, C., Dopp, E. A., Dijkstra, C. D., Simmons, D. L. and van der Berg, T. K. (1998) Signal-Regulatory Protein is Selectively Expressed by Myeloid and Neuronal Cells. *The Journal of Immunology* **161**:1853-1859.

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