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Schillerstr. 5

# AM31836PU-N Monoclonal Antibody to Myeloid Lineage - Purified

Quantity: 0.25 mg
Concentration: 1.0 mg/ml

Background: Hematopoietic stem cells (HSC) are the precursor cells found in the bone marrow

which give rise to all the blood cell types of both the Myeloid and lymphoid lineages, which include monocytes and macrophages, neutrophils, basophils, eosinophils, T cells, B cells, NK cells, microglia, erythrocytes, megakaryocytes and dendritic cells. During the process of hematopoiesis, Myeloid lineage cells originate from the bone marrow, while Lymphoid lineage cells originate from the lymph tissue. Blimp-1 is a key regulator of the differentiation of the separate hematopoietic myeloid and lymphoid

lineages.

The distinction between myeloid and lymphoid lineages is essential to diagnose and treat certain cancers. Myeloid lineage cells induce inflammatory cytokine production upon activation by Kaposi's sarcoma-associated herpesvirus OX2 glycoprotein. At the stage of myelocytes, Myeloid lineage cells express a substantial number of IL-8

receptor homologs.

Host / Isotype: Mouse / IgG1

**Recommended Isotype** 

**Controls:** 

SM20P (for use in rat samples), AM03095PU-N

Clone: OX-82

Immunogen: Anemic Rat bone marrow.

**Format:** State: Liquid purified lg fraction.

Purification: Protein G Chromatography of Ascites fluid.

**Buffer System:** PBS containing 0.02% Sodium Azide as preservative.

**Applications:** Flow Cytometry (See Protocols).

This clone OX-82 has been reported for use in Western Blotting and

Immunohistochemistry on Frozen Sections (Ref. 2).

Other applications not tested. Optimal dilutions are dependent on conditions and

should be determined by the user.

Specificity: This Myeloid Lineage monoclonal antibody recognizes a 35 kDa antigen found on

myeloid cells and stromal elements from a variety of tissues in the adult Rat.

**Species Reactivity:** Tested: Rat.

Storage: Store the antibody undiluted at 2-8°C for one month or (in algiuots) at -20°C for

longer.

Avoid repeated freezing and thawing. Shelf life: one year from despatch.

General Readings: 1. Crook K, Hunt SV. Enrichment of early fetal-liver hemopoietic stem cells of the rat

using monoclonal antibodies against the transferrin receptor, Thy-1, and MRC-OX82.

Dev Immunol. 1996;4(4):235-46. PubMed PMID: 8924759.

2. Hoffmann JC, Herklotz C, Zeitz M, Bayer B, Zeidler H, Westermann J. Effects of the



anti-CD2 mAb OX34 on in vivo proliferation. Ann N Y Acad Sci. 1998 Nov 17;859:216-8. PubMed PMID: 9928391.

#### **Protocols:**

### 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.
- 2. Wash 2 times.
- 3. Resuspend the cells to a concentration of 2x10e7 cells/ml in media A. Add 50 µl of this suspension to each tube (each tube will then contain 1x10e6 cells, representing 1
- 4. To each tube, add 0.1 μg of AM31836PU-N.
- 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 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:**

Rat Strain: Wistar

Cell Concentration: 1x10e6 cells per test

Antibody Concentration Used: 0.1 ug /10e6 cells

Isotypic Control: Mouse IgG1



Pictures:

Cell Source: Bone Marrow Percentage of cells stained above control: 21.5%

