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

SPECIFICITY
Basophils and mast cells are hematopoietic effector cells involved in allergic and inflammatory reactions (1 – 3). Both cell types highly express the high affinity IgE receptor. Mature basophils express IL-3Rα (CD123) but not c-kit (CD117), whereas tissue mast cells express high levels of CD117, but not CD123 (4 – 6). Only a few markers are specific for human mature basophils such as Bsp-1 (7, 8) and 2D7 (9), or mast cells (CD117 (10)), but none recognizes mature as well as immature cells in the basophil lineage.

The monoclonal antibody (mAb) 97A6 recognizes a novel surface antigen expressed on human peripheral blood basophils, but not on other blood cells (11). It also reacts with mature mast cells, and with CD34+ bone marrow progenitors of basophils and mast cells. Moreover the 97A6 antigen is up-regulated after activation of basophils by anti-IgE antibodies and acarid allergens.

The 97A6 mAb is also reactive with some mature megakaryocytic cell lines but does not react with platelets (12). The 97A6 antigen seems to appear during megakaryocytic cell differentiation and to disappear during platelet formation (11, 12).

It was shown that the basophil activation marker defined by the 97A6 mAb is identical to the ecto-nucleotide pyrophosphatase / phosphodiesterase 3 (E-NPP3) (13). This molecule is a type II transmembrane protein that belongs to a family of ectoenzymes involved in hydrolysis of extracellular nucleotides. When analyzing KU-812 basophil precursor cells, the 97A6 mAb immunoprecipitates two proteins of 270 and 150 kDa at reducing conditions and one protein of 270 kDa at nonreducing conditions, suggesting that the 97A6 antigen is a disulfide-linked dimer of two proteins of 150 kDa each (11).

The 97A6 mAb was assigned to the CD203c cluster of differentiation at the 7th International Workshop on Human Leucocyte Differentiation Antigens (HLDA) in Harrogate, England, in 2000 (13).

REAGENT CONTENTS
This antibody is provided in phosphate-buffered saline pH 7.4, containing 0.1% sodium azide and 2 mg/mL bovine serum albumin.

APPLICATION
Flow cytometry.

STATMENTS OF WARNING
1. This reagent contains 0.1% sodium azide. Sodium azide under acid conditions yields hydrazoic acid, an extremely toxic compound. Azide compounds should be flushed with running water while being discarded. These precautions are recommended to avoid deposits in metal piping in which explosive conditions can develop. If skin or eye contact occurs, wash excessively with water.

2. Specimens, samples and all material coming in contact with them should be handled as if they might transmit infection and disposed of with proper precautions.

3. Never pipet by mouth and avoid contact of samples with skin and mucous membranes.

4. Do not use antibody beyond the expiration date on the label.

5. Do not expose reagents to strong light or eye contact occurs, wash excessively with water.

6. Avoid microbial contamination of reagents or incorrect results might occur.

7. Use good laboratory practices when handling this reagent.

STORAGE CONDITIONS AND STABILITY
This reagent is stable up to the expiration date when stored at 2 – 8°C in the dark. Do not freeze.

REAGENT PREPARATION
No reconstitution is necessary. This monoclonal antibody may be used directly from the vial. Bring reagent to 18 – 25°C prior to use.

PROCEDURE
This reagent is designed for flow cytometry. A wash is required to yield optimal results. Assay volume: 20 µL per 5 x 10⁶ cells in one test, or per 100 µL whole blood.

Follow the procedure recommandée dans la version à mettre à jour.

EXAMPLE DATA
The 2 histograms below are biparametric representations (Side Scatter versus Fluorescence Intensity) of a normal heparinized whole blood sample, before (figure 1) and after activation (figure 2) by 1 µg/mL of antiIgGE antibody (See catalog for PN). Staining is with CD203c-PE monoclonal antibody. Isotopic control labelling(See catalog for PN) is not shown. Lysis and fixation are performed with the Whole Blood Lysing Reagent Kit (See catalog for PN). All events acquired are shown (gating on leukocytes).

SELECTED RESEARCH REFERENCES


