

Monoclonal Antibody CD45RA

PN IM1143 – Pre-diluted – Liquid – 6 mL – Clone DBB42

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

SPECIFICITY

The CD45 molecule (also named LCA for Leucocyte Common Antigen) regroups single type I transmembrane glycoproteins with a molecular weight (Mr) ranging from 180 to 220 kDa (1).

The CD45 proteins are all coded by a single gene composed of 33 exons (1). Differential splicing of exons 4, 5 and 6 (which encode A, B, and C determinant respectively) generates at least five isoforms of the CD45 protein (i. e. ABC, AB, BC, B and 0) identified by relevant antibodies (2,3). Antibodies reactive with all five isoforms are clustered as CD45 (CD45 "non-restricted" or pan-CD45) (2). Antibodies reactive with restricted epitope are clustered as CD45R. CD45RA, CD45RB, CD45RC antibodies recognize isoforms which include the expression of A, B and C exon respectively. CD45R0 antibodies react with CD45 isoforms lacking the exon A-, B- and C-encoded regions (1).

The CD45 family of protein is expressed on the surface of all nucleated hematopoietic cells (2, 3).

In contrast to T lymphocytes, most peripheral B cells express the CD45RA isoform (2).

Monocytes and dendritic cells express predominantly low molecular weight isoform (i. e. CD45R0, CD45RB) with a subset expressing CD45RA and CD45RC. Granulocytes principally express only the lower molecular weight isoform (i. e. CD45R0, CD45RB) (2). The most immature thymocytes express only the 180 kDa isoform (i. e. pan-CD45 and CD45R0 positive phenotype). During the maturation of T cells, higher Mr isoforms are expressed (i. e. CD45RA and CD45RC) (1).

On reactive lymph nodes, DBB42 stains both germinal center and mantle zone cells. A subpopulation of T cells may be stained in the paracortical areas. The numbers vary greatly from case to case.

In normal lymph nodes, a subpopulation of macrophages is also strongly positive. In normal spleen, B-lymphocytes in the marginal zone are strongly positive. DBB42 also reacts with macrophages in the red pulp of the spleen.

In the thymus, a few medullary lymphocytes are stained.

Cross reactivity: Kidney tubules, islet cells in pancreas, sweat glands of the skin and gastro-intestinal adenocarcinomas may show a weak to moderate reactivity.

DBB42 stains a subpopulation of megakaryocytes and osteoblasts.

DBB42 monoclonal antibody was evaluated during the 6th International workshop on Human Leukocyte Differentiation in Kobe (1996), (section Non Lineage CD antigens, WS 6, WS code: NL-005) (2).

REAGENT

Monoclonal Antibody CD45RA
PN IM1143 – Pre-diluted – Liquid

CLONE

DBB42

ISOTYPE

IgG1

IMMUNOGEN

Deau cell line, established from a large cell lymphoma (centroblastic type) (3)

HYBRIDOMA

Myeloma
P3XAg8.653 x Balb/c spleen cells

SPECIES

Mouse

SOURCE

ascites fluid

PURIFICATION

Ion exchange or affinity chromatography

REAGENT CONTENTS

Purified Ig in 50 mM Tris-HCl, 0.15 M NaCl, pH 7.2 containing 1 mg/ml bovine serum albumin and 0.1% sodium azide. The buffer contains a green dye.

STATEMENT OF WARNINGS

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 capable of transmitting 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 reagent beyond the expiration date on the vial label.
5. Avoid microbial contamination of reagent or erroneous results may occur.
6. Use general good laboratory practices when handling this reagent.

STORAGE CONDITIONS AND STABILITY

This reagent is stable up to the expiration date on the vial label when stored at 2 – 8°C.

REAGENT PREPARATION

DBB42 6 ml-form is ready for use on routinely fixed paraffin-embedded tissue sections.

PROCEDURE

For research studies using immunohistochemical and cytochemical staining on frozen or routinely-fixed, paraffin-embedded tissue sections (4, 5).

Recommended incubation: at room temperature for 60 minutes on tissue sections.

Paraffin section:

Optimal fixatives are as follows: B5, Bouin's, Dubosq-Brasil (ethanol based Bouin's), Zenkers, formalin.

Positive control:

normal, human lymphoid tissues (e.g. lymph node, spleen, tonsils).

SELECTED RESEARCH REFERENCES

1. Weiss, L.M., Arber, D.A., Chang, K.L., "CD45 : a review", 1993, Applied Immunohistochemistry, 3, 1, 166-181.
2. Sewell, W.A., Cooley, M.A., Hegen, M., "CD45 Workshop Panel Report", 1997, Leucocyte Typing VI, White Cell Differentiation Antigens, 499-502.
3. Al Saati, T., Caspar, S., Brousset, P., Chittal, S., Caveriviere, P., Hounieu, H., Dastuge, N., Idoipe, J-B., Icart, J., Mazerolles, C., Delsol, G., "Production of anti-B monoclonal antibodies (DBB42, DBA44, DNA7, and DND53) reactive on paraffin embedded tissues with a new B-lymphoma cell line grafted into athymic nude mice", 1989, Blood, 74, 2476-2485.
4. Gaulier, A., Fourcade, C., Szekeres, G., Pulik, M., "Bone marrow one step fixation-decalcification in Lowy FMA solution: an immunohistological and in situ hybridization study", 1994, Path. Res. Pract., 190, 1149-1161.
5. Leong, ASY., "Immunohistochemistry: theoretical and practical aspects", 1993, In Leong ASY Ed, Applied Immunohistochemistry for the Surgical Pathologist, Edward Arnold, London, pp.2-22.

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