

The slide to be stained for CD23 comprised:

1: tonsil, 2: marginal zone lymphoma, 3: B-CLL, 4: follicular cell lymphoma, 5: mantle cell lymphoma.

Criteria for assessing a CD23 staining as optimal included: A moderate to strong, distinct membranous staining of normal, activated B-cells in the mantle zone of the tonsil, a strong staining of the follicular dendritic reticulum cells in the germinal centres and a distinct staining of the neoplastic cells of the B-CLL. The neoplastic cells of the mantle cell lymphoma should be negative.



59 laboratories submitted stainings. At the assessment 35 achieved optimal staining (59 %), 10 good (17 %), 10 borderline (17 %) and 4 poor staining (7%).

46 laboratories used mAb 1B12 (Novocastra, n=39; Ventana, n=3; NeoMarkers, n=3; Maixin Bio, n=1). 8 used mAb MHM6 (DakoCytomation), 5 used mAb BU38 (Binding Site, n=4; Ancell, n=1).

In this assessment optimal staining could be obtained only with mAb **1B12** (33/42) and **MHM6** (2/8). Almost all laboratories were able to detect CD23 in the follicular dendritic reticulum cells, but the staining of CD23 in normal mantle zone B-cells and the neoplastic cells of the B-CLL could only be obtained in optimized protocols.

All laboratories achieving an optimal staining used HIER, most frequently (31/34) with Tris-EDTA/EGTA pH 9 as the heating buffer. MWO, pressure cooker and water bath could be used as the heating device. The optimal dilution of mAb 1B12 was in the range of 1:20 – 200, that of mAb MHM6 was 1:50.

The most frequent causes of insufficient stains were:

- Insufficient HIER: too short efficient heating time (MWO < 15 min., water bath < 40 min.)
- Usage of proteolytic pre-treatment
- Too dilute concentration of the primary antibody
- Inappropriate choice of mAb.

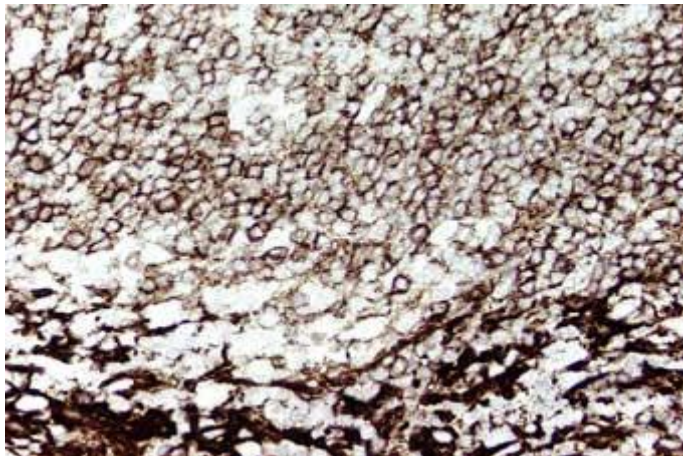


Fig. 1a
Optimal staining for CD23 using mAb 1B12. Normal tonsil. The dendritic reticulum cells (below) are intensely stained, while the mantle zone lymphocytes show a moderate membranous staining.

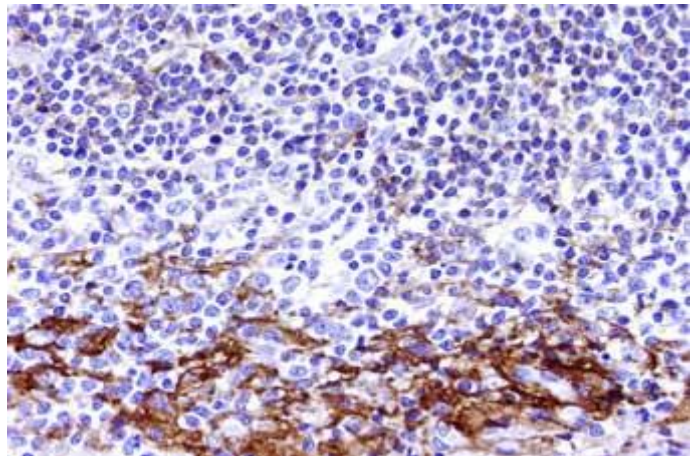


Fig. 1b.
Insufficient staining for CD23 using mAb 1B12. Normal tonsil. The dendritic reticulum cells (below) are moderately stained, while the mantle zone lymphocytes show almost no staining.

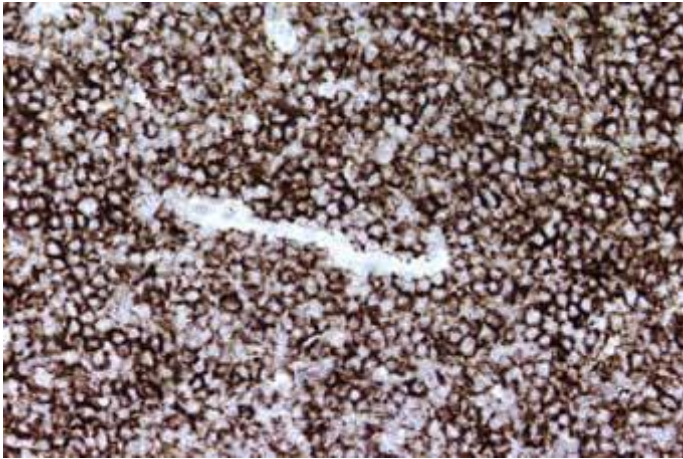


Fig. 2a
Optimal staining for CD23 using mAb 1B12. B-CLL. All neoplastic cells show an intense membranous staining.

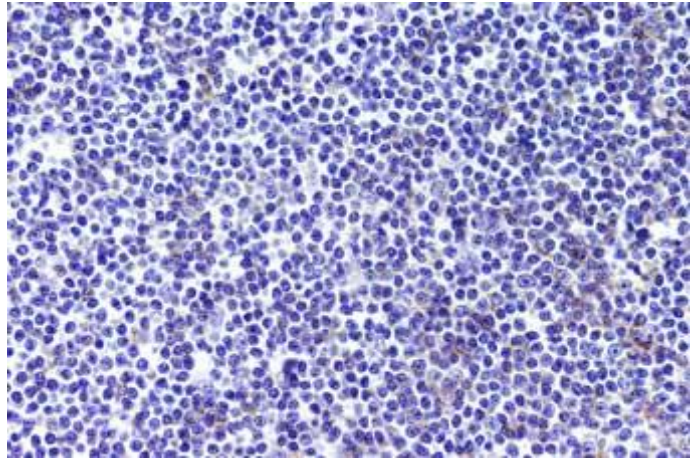


Fig. 2b
Insufficient staining for CD23 using mAb 1B12. B-CLL. The neoplastic cells show almost no staining.

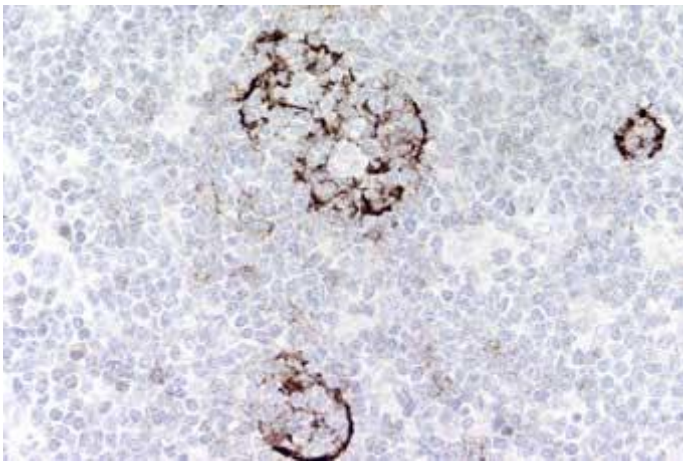


Fig. 3a
Optimal staining for CD23 using mAb 1B12. Mantle cell lymphoma. The dendritic reticulum cells representing remnants of germinal centres are intensely stained, while the neoplastic cells are negative.

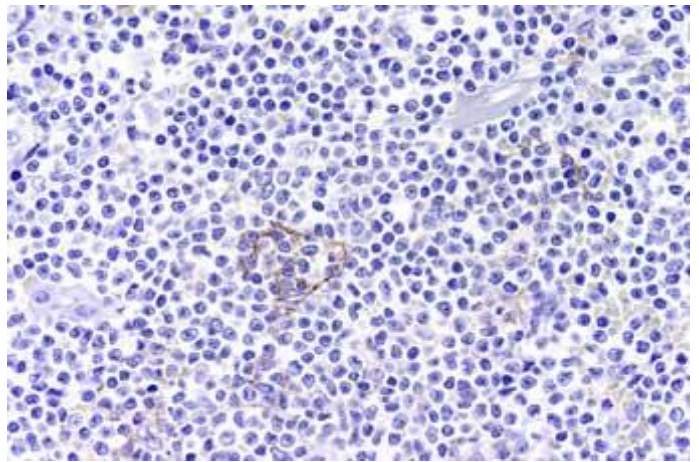


Fig. 3b
Insufficient staining for CD23 using mAb 1B12. Mantle cell lymphoma. The dendritic reticulum cells representing remnants of germinal centres are weakly stained.

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