

The slide to be stained for CD30 comprised:

1. Tonsil, 2. Hodgkin's lymphoma NS, 3. Anaplastic large cell lymphoma,
4. Embryonal carcinoma, 5. Choriocarcinoma.

Criteria for assessing a CD30 staining as optimal included:

A distinct membranous staining of activated B- and T-cells in the tonsil, the anaplastic large cell lymphoma, the embryonal carcinoma and the choriocarcinoma as well as a strong and a distinct membranous and dot-like (Golgi) staining of the Reed-Sternberg and Hodgkin's cells in the Hodgkin's lymphoma.



74 laboratories submitted stainings. At the assessment 32 achieved optimal staining (43 %), 36 good (49 %), 3 borderline (4 %) and 3 (4 %) poor staining.

The following Abs were used:

- clone Ber-H2 (DakoCytomation, n=66; Ventana, n=4; NeoMarkers, n=1; Zymed, n=1)
- clone 1G12 (Novocastra, n=1)
- clone HRS-4 (Immunotech, n=1)

In this assessment optimal stainings could only be obtained with clone Ber-H2. However, this was used by all but two of the laboratories.

In the optimal protocols all used HIER. With Tris-EDTA/EGTA pH 9 as the heating buffer, 26 out of 54 obtained an optimal result, with Citrate pH 6 it was 2 out of 10, with Target Retrieval Solution (DakoCytomation) it was 3 out of 3, and with CC1 (Ventana Benchmark) it was 1 out of 2 labs.

None of the 5 laboratories using proteolytic pretreatment obtained an optimal staining, and 3 were insufficient. Ber-H2 was used in the range of 1:20 – 200 depending on the total sensitivity of the protocol used.

The majority of laboratories were able to detect CD30 in the anaplastic large cell lymphoma and the two carcinomas, whereas the demonstration of CD30 in the activated B and T lymphocytes and Hodgkin's lymphoma was only achieved with sensitive protocols.

The most frequent causes of insufficient stainings were:

- Inappropriate choice of epitope retrieval (i.e., proteolytic pre-treatment)
- Too low concentration of the primary antibody

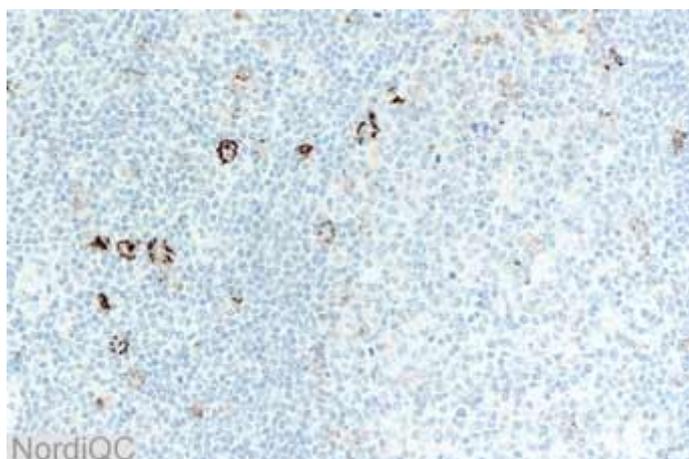


Fig. 1a
Optimal CD30 staining of the tonsil. Intense membranous staining is seen in the activated lymphocytes.

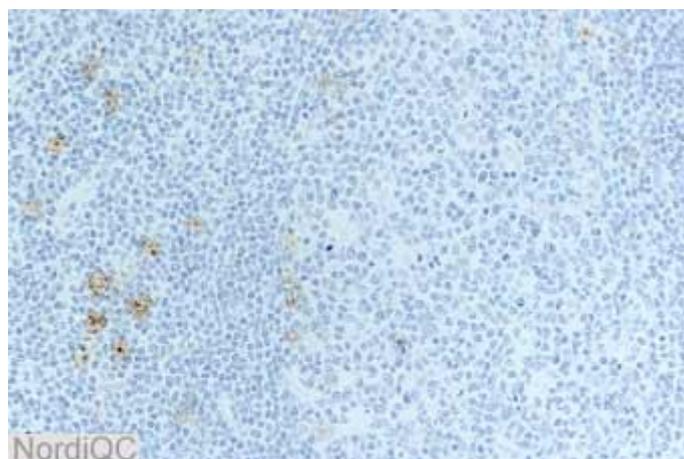


Fig. 1b
Insufficient CD30 staining of the tonsil. The activated lymphocytes are only weakly demonstrated (same field as Fig. 1a). Compare with Figs. 2a and 2b.

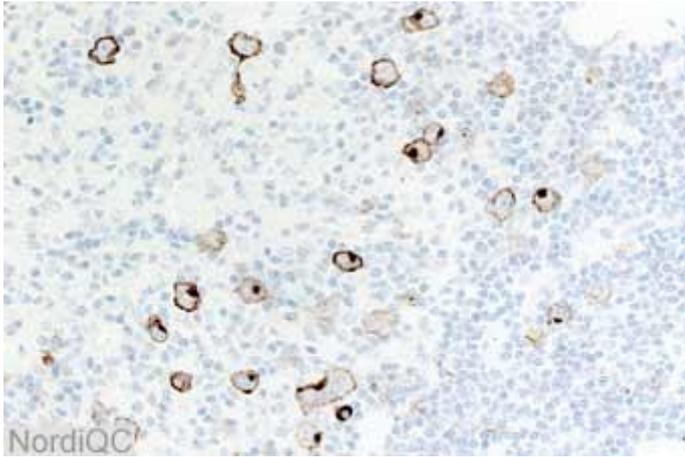


Fig. 2a
Optimal CD30 staining of the Hodgkin's lymphoma. Intense membranous staining and a dot-like reaction is seen in the Hodgkin's and Reed-Sternberg cells.

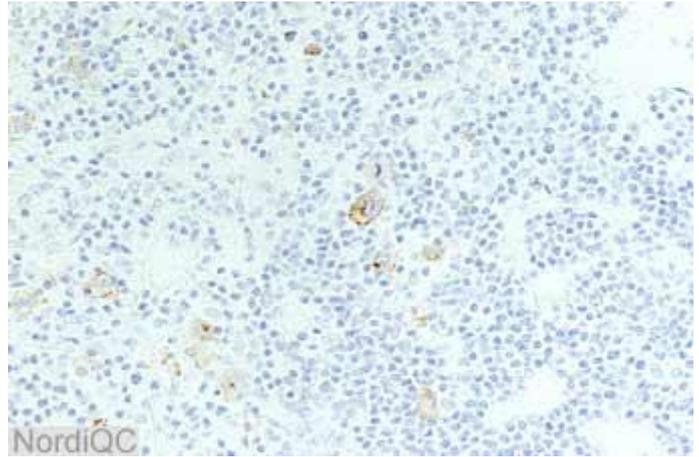


Fig. 2b
Insufficient CD30 staining of the Hodgkin's lymphoma (same protocol as Fig. 1b). The Hodgkin's and Reed-Sternberg cells are only weakly stained or unstained (same field as Fig. 2a).

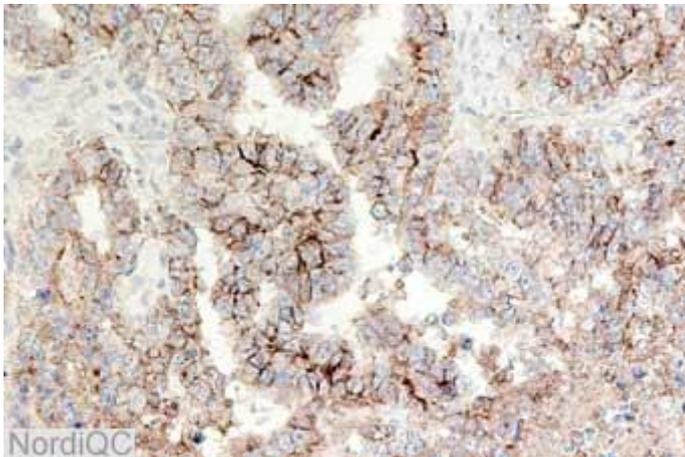


Fig. 3a
Optimal CD30 staining of the embryonal carcinoma. Intense membranous staining is seen.

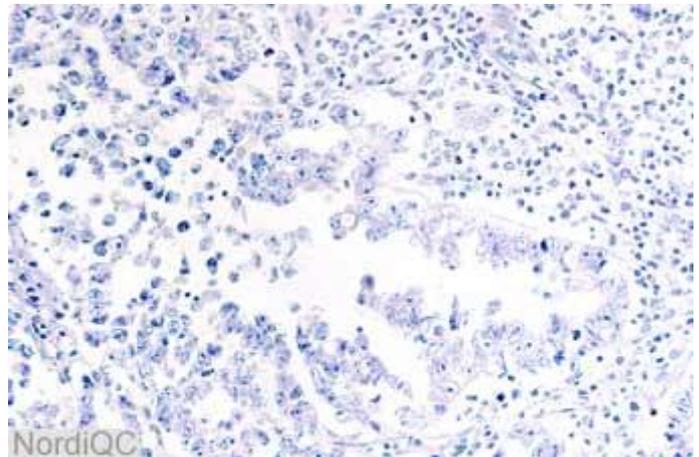


Fig. 3b
Insufficient CD30 staining of the embryonal carcinoma. The tumour is false negative with a poor morphology due to excessive proteolysis.

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