The slides to be stained for CD3 contained four T-cell lymphomas and one B-cell lymphoma in a spleen 'matrix'.

45 laboratories submitted the staining. Of these, 35 used Dako's pAb, 8 used mAb PS1, 1 used mAb F7.2.38, and 1 used mAb UCHL-1.

At the assessment optimal staining was achieved in 10, acceptable in 21, borderline in 12 and poor staining in 2 of the laboratories. Optimal results were in one or more cases achieved with all of the Abs Dako's pAb, PS1 and UCHL-1.

Mandatory for an optimal staining was an efficient HIER and appropriate dilution of the primary antibody. The main reasons for a borderline or poor staining was insufficient HIER (too short efficient heating time and/or inappropriate pH) and a too dilute primary Ab (in one too concentrated Ab) in relation to the overall sensitivity of the protocol.

Fig. 1A
Optimal staining of a T-cell lymphoma, using mAb PS1 (good protocol 1). All T-cells are strongly stained, while residual B-cells (upper left corner) are unstained. Exactly the same staining result were obtained using Dako's pAb (good protocol 2).

Fig. 1B (same field as in Fig. 1A, same Ab)
Acceptable staining of a T-cell lymphoma. The reaction can be interpreted, but the signal is weak.
Fig. 1C (same field as in Fig. 1A, same Ab)
Insufficient staining. The T-cells is very faintly stained, the nuclear counterstaining predominate.

Fig. 2
Staining of spleen. Insufficient staining using DAKO’s polyclonal Ab. Due to high concentration of the Ab, a considerable cross reaction with B-cells is seen.

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