Assessment Run 9 2003

Thyroid Transcription Factor-1 (TTF1)

The slide to be stained for Thyroid Transcription Factor-1 (TTF1) comprised:
1: liver, 2: Thyroid papillary carcinoma, 3: Lung adenocarcinoma, 4: Lung large cell carcinoma, 5: Lung carcinoid tumour.

Criteria for assessing a TTF1 staining as optimal included: A moderate to strong distinct nuclear staining of all alveolar cells and thyroid epithelial cells (when present) and all or almost all neoplastic cells in the lung tumours as well as both the thyroid papillary carcinoma. No other cells should show nuclear staining. No cytoplasmic staining should be seen except for staining of hepatocytes with mAb 8G7G3/1.

63 laboratories submitted stainings. At the assessment 14 achieved optimal staining (22 %), 24 good (38 %), 20 borderline (32 %) and 5 (8 %) poor staining.

52 laboratories used mAb 8G7G3/1 (DakoCytomation, n=49, NeoMarkers, n=2, Ventana, n=1). 11 laboratories used mAb SPT24 (Novocastra).

Both mAbs could be used for obtaining an optimal staining, giving identical staining patterns (Fig. 1a and 2a) with the exception of staining of liver cell cytoplasm seen with mAb 8G7G3/1 only. The optimal dilution of mAb 8G7G3/1 was in the range of 1:50 – 200 and of mAb SPT24 in the range of 1:30 - 200.

The majority of the laboratories was able to demonstrate TTF1 in both the normal and neoplastic epithelial cells in the thyroid papillary carcinoma and the lung adenocarcinoma, whereas the demonstration of TTF1 in the lung large cell carcinoma and in particular the lung carcinoid caused difficulties for several laboratories. For instance, TTF1 could be detected in the lung adenocarcinoma using a dilution of the mAb 8G7G3/1 of 1:1,000 – 5,000 (using HIER in Tris/EDTA pH 9 and EnVision™+/DAB+), but with these dilutions the carcinoid were weakly stained or unstained.

All laboratories achieving an optimal staining used HIER, most frequently with Tris-EDTA/EGTA pH 9 as the heating buffer (10/14), in few cases EDTA pH 8 (2/14) or Citrate pH 6 (2/14).

The most frequent causes of insufficient stainings (often in combination) were:
- Insufficient HIER (too short efficient heating time [MWO < 15 min., water bath <40 min.] typically in combination with Citrate buffer pH 6)
- Too low or too high concentration of the primary Ab.

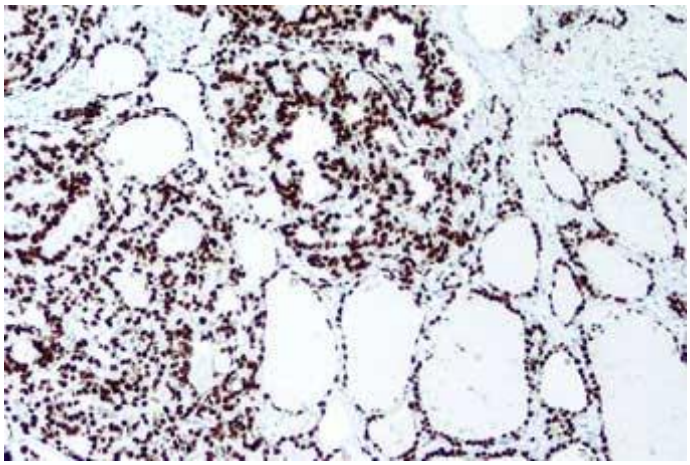

Fig. 1a
Optimal staining for TTF1 using mAb 8G7G3/1. Thyroid papillary carcinoma. All nuclei of normal thyroid cells and neoplastic cells are strongly stained.

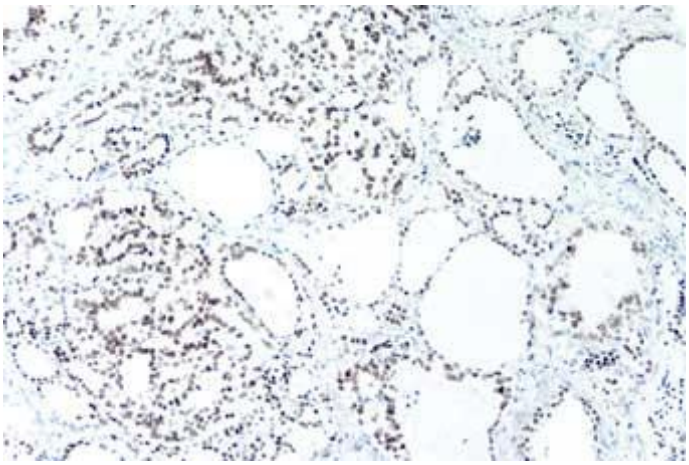

Fig. 1b
Staining for TTF1 using mAb 8G7G3/1 with an insufficient protocol. Thyroid papillary carcinoma (same field as in Fig. 1a). The nuclei of normal thyroid cells and neoplastic cells are weakly or moderately stained. However, compare with Fig. 2b.

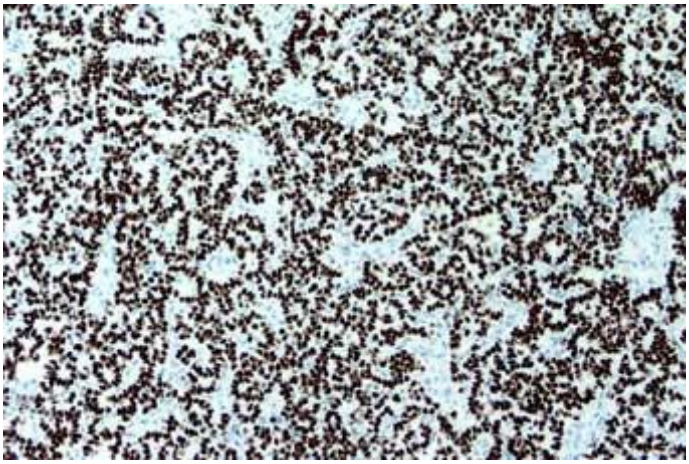

Fig. 2a
Optimal staining for TTF1 using mAb 8G7G3/1. Lung carcinoid tumour. All nuclei of the neoplastic cells are strongly stained.

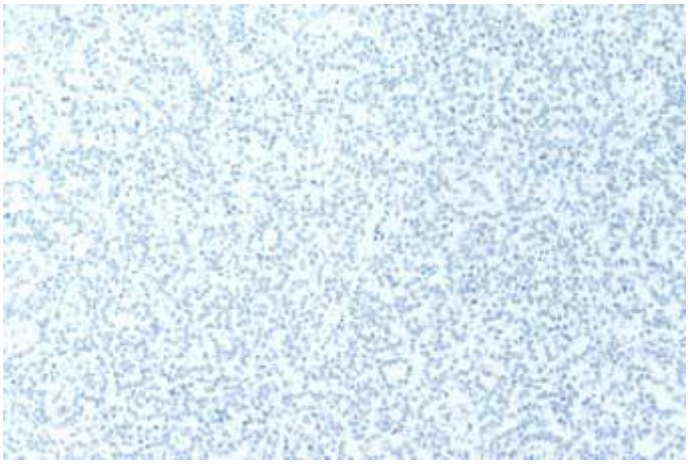

Fig. 2b
Insufficient staining for TTF1 using mAb 8G7G3/1 with the same protocol as in Fig. 1b. Lung carcinoid tumour (same field as in Fig. 2a). The nuclei are unstained.

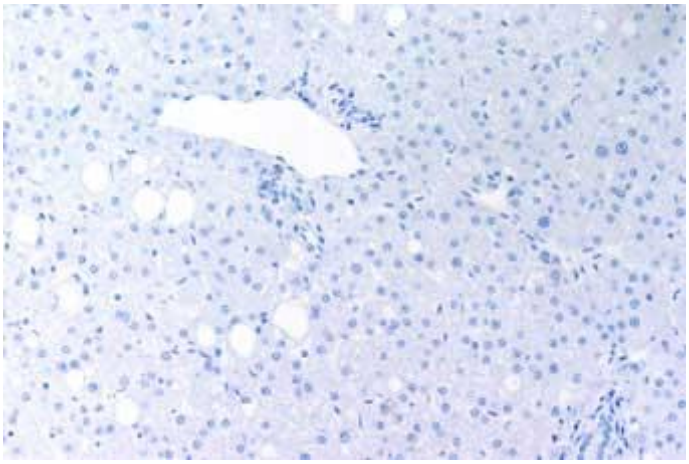

Fig. 3a
Staining for TTF1 using mAb SPT24. Normal liver. No staining is seen.

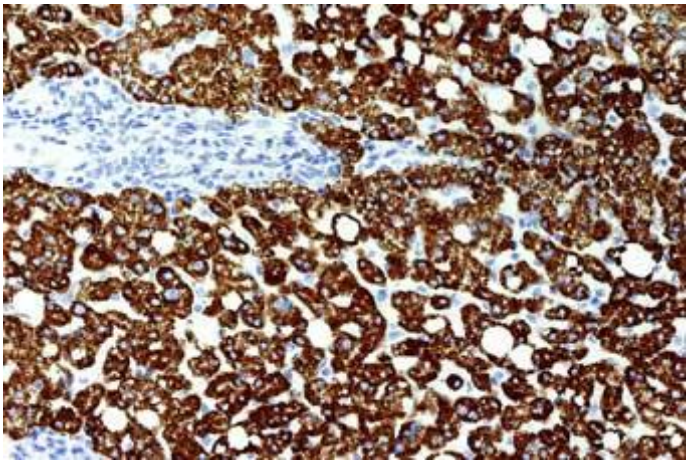

Fig. 3b
Staining for TTF1 using mAb 8G7G3/1. Normal liver. Strong cytoplasmic staining is seen. The reaction has some resemblance to an endogenous biotin reaction. However, the staining was carried out in a biotin free system.

SN/MV/LE 1-12-2003