

Nischalke HD, Schmitz V, Luda C, Aldenhoff K, Berger C, Feldmann G, Sauerbruch T, Spengler U, Nattermann J (2012). Detection of IGF2BP3, HoxB7, and NEK2 mRNA expression in brush cytology specimens as a new diagnostic tool in patients with biliary strictures. *PLOS one* 2012;7(8):e42141. Epub 2012 Aug 7.

**INTRODUCTION:** It is a challenging task to distinguish between benign and malignant lesions in patients with biliary strictures. Here we analyze whether determination of target gene mRNA levels in intraductal brush cytology specimens may be used to improve the diagnosis of bile duct carcinoma.

**MATERIALS AND METHODS:** Brush cytology specimens from 119 patients with biliary strictures (malignant: n = 72; benign: n = 47) were analyzed in a retrospective cohort study. mRNA of IGF-II mRNA-binding protein 3 (IGF2BP3), homeobox B7 (HOXB7), Forkhead box M1 (FOXM1), kinesin family member 2C (KIF2C) and serine/threonine kinase NEK2 was determined by semi-quantitative RT-PCR using the  $\Delta$ Ct method.

**RESULTS:** IGF2BP3 ( $p < 0.0001$ ), HOXB7 ( $p < 0.0001$ ), and NEK2 ( $p < 0.0001$ ) mRNA expression levels were significantly increased in patients with cholangiocarcinoma or pancreatic cancer. Median  $\Delta$ Ct values differed by 3.5 cycles (IGF2BP3), 2.8 cycles (HOXB7) and 1.3 cycles (NEK2) corresponding to 11-fold, 7-fold and 2.5-fold increased mRNA levels in malignant versus benign samples.

Sensitivity to detect biliary cancer was 76.4% for IGF2BP3 (80.9% specificity); 72.2% for HOXB7 (78.7% specificity) and 65.3% for NEK2 (72.3% specificity), whereas routine cytology reached only 43.1% sensitivity (85.4% specificity).

Diagnostic precision was further improved, when all three molecular markers were assessed in combination (77.8% sensitivity, 87.2% specificity) and achieved 87.5% sensitivity and 87.2% specificity when molecular markers were combined with routine cytology.

**CONCLUSIONS:** Our data suggest that measuring IGF2BP3, HOXB7 and NEK2 mRNA levels by RT-PCR in addition to cytology has the potential to improve detection of malignant biliary disorders from brush cytology specimens.