regulated by a network of proteins such as ATM or Chk2 and their activated forms (phosphorylated).

The aim of our study is to investigate the possible role of DNA damage response (DDR) related proteins in the recurrence of NMIBC.

METHODS: We performed a retrospective study of 172 consecutive patients (median age 69 years, 84% males) with primary NMIBC diagnosis within the period 1998-2003. Representative paraffin blocks from each patient were used to construct three TMA. Immunohistochemistry (IHC) staining for ATM, Chk-2, p53 and their active forms (phosphorylated) was performed. IHC results were analyzed in a qualitative and a semi-quantitative manner (histoscore). Hazard ratios (HR) were calculated using Cox proportional Hazards model.

RESULTS: With a median follow up of 72 months, the rate of recurrence was 50.6%. Median time to first recurrence was 16.2 months. We observed a higher risk of recurrence in patients who expressed pATM (HR 2.4, IC 95% 1.02-6.29) and p53 (HR 1.66, IC 95% 1.02-2.68). If we subdivide patients according to grade and tumor stage, we found that pTa tumors with pATM expression had 8.24 times more possibilities to recur than those which did not express it (IC 95% 1.14-59.6). In multivariate analysis, pATM expression was an independent predictor for recurrence in pTa tumors (HR 8.41, IC 95% 1.16-60.9). We did not find any relation between recurrence and expression of Chk2, Chk2 or ATM.

In Cox proportional Hazards model, using the p53 cut point of 11(which corresponds to p53 median value), recurrence risk of pTa with p53 < 11 and expression of pATM was 34.3%, 68.4% and 86.5% in 1, 2 and 5 years respectively, whereas recurrence risk of pTa tumors with p53<11 and negative expression of pATM was 2.22%, 5.95% and 10.17% in 1, 2 and 5 years.

CONCLUSIONS: The positive expression of p53 and pATM increases the risk of recurrence in NMIBC patients. The risk of recurrence is clearly increased in pTa tumors when positive staining of pATM is present, especially if there is also an overexpression of p53. Further studies are needed to corroborate the prognostic value of pATM expression in recurrence of NMIBC.

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MP45-13
CENTROSOME AMPLIFICATION AS A PUTATIVE PROGNOSTIC BIOMARKER FOR IMPROVED OBJECTIVE CLASSIFICATION OF UROTHELIAL CARCINOMA
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INTRODUCTION AND OBJECTIVES: Recent studies have reported that centrosome amplification (CA) is closely related to chromosomal instability (CIN) and patient prognosis in human malignancies. The relationship between DNA copy number aberrations (DCNAs) and CA remains to be elucidated. Therefore we examined the relationship between CA and genomic alterations in urothelial carcinomas. To our knowledge, this is the first report to confirm CA as a reliable biomarker for assessing DCNAs in urothelial carcinomas.

METHODS: Centrosomes were evaluated by immunohistochemistry using an anti-γ-tubulin antibody. Array-based comparative genomics hybridization technology using DNA chips was applied to 70 urothelial carcinomas to examine DCNAs in the entire genome. Studying aberrations in the number of chromosomes 7, 9 and 17 using fluorescence in situ hybridization allowed for the estimation of the degree of CIN. We compared DCNAs and CA with tumor grade. Further, the patient prognosis of these parameters was evaluated.

RESULTS: DNA copy number gains at 20p12.2, 5p15.2, 5p15.31, and 17q25.3 and losses at 17p12, 8p22, 2q37.3, 5q31.1, and 2q37.3 were more frequent in tumors with CA than in those without it. The total numbers of DCNAs and frequency of CIN were also larger in tumors with CA than in those without it (P<0.0001 and P<0.0001, respectively). These parameters were more closely associated with CA than with subjectively assigned tumor grade (Table). Recurrence-free survival rate was higher in tumors without CA than in those with it (P=0.0307) but was not statistically different between high and low grade tumors (P=0.5529). For the subgroup of patients with low grade tumors, CA and tumor recurrence were more frequent in tumors with CA than in those without it (P=0.0111 and P=0.0242, respectively).

CONCLUSIONS: These data suggest that CA may have great potential as a biomarker for improved objective classification of urothelial carcinoma and estimation of prognosis. Tumors with CA have large total numbers of DCNAs and CIN, and therefore, these tumors may have worse prognoses.

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MP45-14
EPIGENETIC REGULATION OF CANCER TUMOR SUPPRESSOR GENE ADAMTS18 IN HUMAN BLADDER CANCER ASSOCIATED WITH ITS PROGNOSIS
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INTRODUCTION AND OBJECTIVES: A disintegrin and metalloprotease with thrombospondin motifs 18 (ADAMTS18), located on 16p23.1, is involved in carcinogenesis of multiple cancers, but its role in bladder cancer (BCa) remains unclear. Here, we reported the epigenetic alterations of ADAMTS18 and its function in bladder carcinogenesis.

METHODS: We detected ADAMTS expression in normal BCA tissue, BCA cell lines and primary tumors by semi-quantitative RT-PCR and immunohistochemistry, and further assessed its tumor suppressive functions. Methylation-specific PCR and bisulfite genomic sequencing were used to examine the methylation status of ADAMTS18 in non-muscle invasive/muscle invasive bladder cancer tissues and urine sediments.

RESULTS: We found that ADAMTS18 was highly expressed in normal bladder cell line and tissue, but frequently downregulated in BCA cell lines and tissues. Moreover, pharmacologic demethylation could restore ADAMTS18 expression. We further detected ADAMTS18 methylation in our clinical specimens, and found that ADAMTS18 was methylated in 14/17 muscle invasive tumors primary tumors, but relatively lower in non-muscle invasive tissues (13/30). ADAMTS18 methylation was also associated with high pathologic grade (p=0.017). Then the tumor suppressor function of ADAMTS18 was detected in T24. Ectopic expression of T24 inhibited the colony formation and suppressed migration of bladder cancer cells.

CONCLUSIONS: Taken together, our data suggests that ADAMTS18 as a functional tumor suppressor is frequently methylated in BCa and associates with tumor stage and grade, indicating that it may serve as a tumor prognosis marker for BCa diagnosis.

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MP45-15
NON-INVASIVE UROTHELIAL CANCER BIOMARKERS
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INTRODUCTION AND OBJECTIVES: The current gold standard diagnostic method of urothelial cancers including bladder, ureter