**Introduction and Objectives**

- EpiCheck is a novel DNA methylation-based assay for detection of bladder cancer (BC) in voided urine samples.
- The test is based on identification of DNA methylation changes associated with BC in a panel of 15 genomic biomarkers.
- The assay generates a numerical EpiScore (0-100) reflecting the overall methylation level in the urine sample.
- An EpiScore ≥ 60 is considered positive for BC.
- The aim of this study was to compare the sensitivity and specificity of the assay to that of cytology in a population of patients with history of BC.

**Methods**

- Voided urine samples from 222 patients with history of BC undergoing routine surveillance were tested with Bladder EpiCheck. 40/222 patients (18%) had biopsy-proven recurrent BC.
- The distribution of stages was: 16-Ta, 13-T1, 3-T2 and 12-CIS.
- The distribution of grades was 19-LG, 26-HG.
- Cytology results were available in 173 of these samples.
- Statistical analysis was performed using the Kolmogorov-Smirnov test.

**Results**

**Overall sensitivity and specificity**

- EpiCheck showed an overall sensitivity of 90% over specificity of 83%.

**Sensitivity by Stage and Grade**

- EpiCheck sensitivity of the high grade tumors was 95% and of the low grade tumors 84%.
- The sensitivity of the different stages and grades is shown below:

**Negative Predictive Value**

- The negative predictive value for this cohort was 97.4%.

**Comparison to Cytology**

- Cytology results were available in 173 of these cases. Cytology performance was 40% sensitivity and 96% specificity.
- 15 of these 173 samples were from patients with tumors. All 6 tumors that were detected by cytology were detected also by Bladder EpiCheck and the 2 tumors that were missed by Bladder EpiCheck were also missed by cytology. 7 tumors were detected only by Bladder EpiCheck.

**EpiScore analysis**

- The average EpiScore was 69 and 85 for patients with LG and HG tumors, respectively, hence significantly correlated with tumor grade (p = 0.006).
- The average EpiScore per stage was:

<table>
<thead>
<tr>
<th>Stage</th>
<th>Average EpiScore</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ta</td>
<td>68.6</td>
</tr>
<tr>
<td>T1</td>
<td>84.4</td>
</tr>
<tr>
<td>T2</td>
<td>90.7</td>
</tr>
<tr>
<td>CIS</td>
<td>84.4</td>
</tr>
</tbody>
</table>

- Statistical analysis using the Kolmogorov-Smirnov test showed a statistical significant difference between the Ta and T2 EpiScores (p=0.0077) and between Ta and CIS (p=0.0155).

**Conclusion**

- Bladder EpiCheck is a sensitive and specific operator-independent assay for the detection of BC in voided urine samples.
- In comparison with cytology, Bladder EpiCheck has significantly higher sensitivity with slightly lower specificity.
- Our results suggest that Bladder EpiCheck can be used instead of urine cytology in monitoring of BC patients without the risk of missing recurrences.