Results of UroVysion test versus results of LOH analysis in bladder cancer patients

Borkowska E.M.¹, Constantinou A.², Jedrzejczyk A.², Traczyk M.¹, Rozniecki M.³, Sosnowski M.⁴
Kaluzewski B.¹

Introduction and objectives

The multiprobe fluorescence in situ hybridization (FISH) test UroVysion (Vyysis, Inc., U.S.A.) identifies specific abnormalities in chromosomes 3:7:17 and in 9p21-specific centromeric blocks, which are frequently present in urinary UC (UC). Most authors have reported that the FISH test demonstrates higher sensitivity for all grades and stages of UC than conventional cytology alone. Also the US Food and Drug Administration has approved its use in UC diagnosis. According to the manufacturer’s guidelines, a positive FISH result is defined as the presence of 4 or morphologically abnormal (large nuclear size, irregular shape) of 25 analyzed cells that demonstrate either polyploidy of 2 chromosomes 3 and 17 in the same cell or homologous deletions of 2p11 in 12 of 25 cells.

Loss of heterozygosity (LOH) is frequently observed in urine bladder neoplasms. In the reported study, an attempt was undertaken to determine the loss of heterozygosity of 9p21(3p21), 17p13(6p21), CDKN2A/ARF(p16) genes in DNA from metastatic tumors, collected from patients with diagnosed UC-bladder cancer patients, and to compare the results with those of UH evaluation in DNA isolated from urine sediment cells.

Two hundred cases of urinary carcinoma in various stages and grades, collected during the years 2005-2011. The loss of heterozygosity and UroVysion test were evaluated in 205 patients (178 male and 27 female). The mean age of the patients was 69 years. The average observation period of the patients was 32 months. See Table 1 for histopathological results.

<table>
<thead>
<tr>
<th>Category</th>
<th>Tumour grade</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stage</td>
<td>T4</td>
</tr>
<tr>
<td></td>
<td>T3N0M0</td>
</tr>
<tr>
<td>Grade</td>
<td>G1</td>
</tr>
<tr>
<td></td>
<td>G2</td>
</tr>
</tbody>
</table>

Table 1.

Results

The loss of heterozygosity (LOH) at the least, one marker was identified in 50% (225) of the studied tumours. LOH was found in 14.3% of the informative cases for TP53 and RB1 genes, as well as in 34.2% of the informative cases for CDKN2A/ARF genes. See Diagram 4 for LOH frequency in the particular studied markers. The highest LOH percentage was recorded for O6R1348 marker (26.3%). The baseline expression of all evaluated genes in the positive reaction was 9%, while in 0%, it was 93%. The average rate of loss of heterozygosity and UroVysion test was 10.5% in DNA isolated from metastatic tumors, collected from patients with diagnosed UC.

Loss of heterozygosity (LOH) was tested in 9p21(3p21) and 17p13(6p21) genes by UroVysion test. The results showed that the frequency of loss of heterozygosity in various stages and grades of bladder cancer patients was 7% and 26% in DNA isolated from metastatic tumors and urine sediment cells, respectively. In 13 cases, the results of the study showed that DNA isolated from urine sediment did not correspond to the results of LOH analysis in DNA isolated from metastatic tumors. We identified 2, 1, and 10 as the positive results of LOH analysis in metastatic tumors, DNA isolated from urine sediment, and DNA isolated from metastatic tumors, respectively. The values of the parameters were 72% and 98%, respectively.

Conclusions

The analysis of the loss of heterozygosity of DNA, isolated from urine sediment cells, is probably a more reliable method than the cytological study of urine; however, the sensitivity of the presented test, which was obtained in the course of the reported study, 25% of tumours with informative results for studied markers is not satisfactory. A selection of a more appropriate set of markers may be of key importance to regard the results of microsatellite sequences as a reliable, non-invasive method of metastatic identification in urine sediment.

References