Expression of p53 as a potential marker of muscle invasiveness in bladder cancer

S.M. Warli
Department of Urology, Faculty of Medicine, Universitas Sumatera Utara, Medan, North Sumatera, Indonesia

L.I. Laksmi
Department of Pathology, Faculty of Medicine, Universitas Sumatera Utara, Medan, North Sumatera, Indonesia

F. Safriadi
Department of Urology, Faculty of Medicine, Padjajaran University, Bandung, Indonesia

R. Umbas
Department of Urology, Faculty of Medicine, University of Indonesia, Jakarta, Indonesia

ABSTRACT: To analyse the association of p53 and muscle invasiveness in bladder cancer. The samples of this study are patients with bladder cancer. The samples were then classified into Non-Muscle Invasive Bladder Cancer (NMIBC) and Muscle Invasive Bladder Cancer (MIBC). All samples then underwent immunohistochemistry assay for p53, and then group analysed. p53 was expressed in 29 (96.7%) patients with MIBC and 23 (76.7%) patients with NMIBC. There was a significant difference in the expression of p53 between both groups (OR 8.8; 95% CI: 1.012- 76.96). The expression of p53 was significantly associated with muscle invasiveness of bladder cancer. Prospective study is needed to determine its potential in predicting muscle invasiveness of bladder cancer.

Keywords: Bladder cancer, predictive factors, p53

1 INTRODUCTION

Bladder cancer is the ninth most common malignancy in the world, with more than 380,000 cases and 150,000 deaths annually (Witjes et al., 2014). More than 90% of bladder cancers are Urothelial Cell Carcinoma (UCC). Most of all new cases (75–80%) from UCC are classified as non-muscle-invasive or superficial. This tumour has a recurrence rate of 50–70% and 10–15% progress to muscle invasion over a five year period (Shariat et al., 2008).

The high incidence causes special attention became necessary after the initial management. Progression and metastasis are the main problems after initial therapy (Cheng et al., 2009). Staging, grading, size, and multifocality, which are conventional prognostic factors, could not predict clinical outcome in the majority of patients with bladder cancer. Marking could predict recurrence, progression, therapeutic response, and survival (Malats et al., 2005; Schrier et al., 2004).

UCC has various biological and functional characteristics. Recurrence, progression into a higher grade and stage tumours and metastasis are the most common risks in patients with clinically diagnosed UCC. Bladder cancer prognostic could not be predicted by current available prognostic markers that to newly discovered molecular markers. Molecular changes
caused phenotype alterations, thus immunohistochemistry could be used as an early detection tool for bladder cancer.

2 METHODS

The expression of p53, which indicates the muscle invasion in bladder cancer, were analysed in this case-control study, which included patients with muscle-invasive bladder cancer while patients with non-muscle invasive bladder cancer were used as controls. The study was conducted in Haji Adam Malik Hospital – Faculty of Medicine, Sumatera Utara University in conjunction with the Pathology Department, Haji Adam Malik Hospital in Medan, Indonesia from January 2012 to December 2015. Histopathologically diagnosed bladder cancer and good condition of paraffin block. Meanwhile, the exclusion criteria patients with bladder cancer who chemotherapy or radiotherapy and other malignancy.

Changes in brownish colour in epithelial cell cytoplasm or stroma for p53, which is seen the light microscope and will be assessed as intensity positivity and classified into four levels: Negative = 0; Weak = +1, Moderate = +2, Strong = +3. The positive value of p53 immunohistochemistry is defined as a quantitative value in the brown intensity distribution percentage per one field of view of the light microscope with a magnitude of 400 times. Positivity of value is classified into three levels: Negative (0); IHC negative; Focal (1): coloured cells < 50%; Diffuse (2): coloured cells > 50%. These two positivity value combined nH-score value. H-score value is determined based on McCarty criteria, which are: intensity positivity value with the addition of 1 multiplied with the quantity positivity value; [HS = (i+1) x k] (McCarty, 1986) thus the cut-off point is determined at 3. Therefore, the samples were divided into two groups, with H-score 3 or greater, and below 3.

The negative value is acquired by determining the cut-off point involving cases with colour intensity and negative control, cases with weak, moderate, and strong positive colour, with focal quantity positivity. Further analyses were done using SPSS version 20.0. Any statistical test which has p-value < 0.05 is considered statistically significant.

3 RESULT

A total of 60 subjects were involved. This study consisted of 30 cases and 30 controls. Characteristics of the subjects are presented in Table 1.

The mean age in the cases and controls group were 57.07 years and 55.9 years respectively then. ladder cancer occur in men. Both age and gender were not significantly correlated with muscle invasiveness of bladder cancer (p > 0.05). The example result of IHC of p53 is shown in Figure 1.

p53 was significantly correlated with muscle invasiveness of bladder cancer. The probability of positive expression of p53 was 8.8 times higher in MIBC (p = 0.026; OR 8.8; 95% CI: 1.012–76.96).

<table>
<thead>
<tr>
<th>Variable</th>
<th>Cases n = 30</th>
<th>Controls n = 30</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age Mean</td>
<td>5.9 ± 10.37</td>
<td>55.9 ± 11.91</td>
<td>0.687</td>
</tr>
<tr>
<td>Gender</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>26 (86.7%)</td>
<td>26 (86.7%)</td>
<td>0.647</td>
</tr>
<tr>
<td>Female</td>
<td>4 (13.3%)</td>
<td>4 (13.3%)</td>
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</tr>
</tbody>
</table>

Table 1. Frequency of age and sex results based on p53 expression.
A total of 96.7% of MIBC patients in this study showed a positive p53 expression. To find the relationship between the expression of IHC p53 with the occurrence of muscle invasion in bladder cancer, we performed bivariate analysis using a chi-square test. The chance of IHC p53 positive appearing in MIBC is 8.8 times higher than the negative p53 IHC. This indicates that p53 contributes to muscle invasion in bladder cancer.

The result in this study is in accordance with some previous studies (Schrier et al., 2004; Esrig et al., 1994; Serdar et al., 2005), which also concluded that overexpression of p53 was higher in group invasive tumours. This study also discovered that overexpression of p53 with tumour progression does not depend on the grading, the presence or absence of vascular invasion, or the presence of carcinoma in situ (Sarkis et al., 1993).

Grosmann Barton et al. (1998) recommend allocating bladder cancer patients stratification of pT1 based on the status of p53 and RB.10 as results suggest that patients whose normal expression of these two genes can be managed conservatively. However, if there is a change in any of the two genes, aggressive management to prevent disease progression into invasive is recommended (Grossman et al., 1998; Van Rhijn et al., 2001).

A meta-analysis that was performed by Malats et al. (2005) in 117 studies had shown different results. Four showed that changes in p53 are a weak predictor factor of recurrence, progression, and mortality in bladder cancer. This result might be caused by the long period of patient recruitment, thus causing a heterogeneity, variability of immunohistochemistry, either in the antibody or the scoring system used, or the absence of a uniform definition to determine the limits of positive staining so that it can give different results due to differences in the cut-off (Malats et al., 2005). p53 has an important role for cell response to various stress and to maintain the stability of the gene. Because of its critical role in tissue homeostasis, p53 is the most commonly mutated gene in malignancy. p53 mutation causes the accumulation of p53 in the cell nucleus. Generally, overexpression in cell nuclei associates with p53 inactivation. In addition, overexpression can also be caused by the physiological response to DNA damage. The instability of these genes can also be caused by their bond

<table>
<thead>
<tr>
<th>Variable p53</th>
<th>MIBC n = 30</th>
<th>NMIBC N = 30</th>
<th>p-Value</th>
<th>OR</th>
<th>95%CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>29 (96.7%)</td>
<td>23 (76.7%)</td>
<td>p = 0.026</td>
<td>8.8</td>
<td>1.012–76.96</td>
</tr>
<tr>
<td>Negative</td>
<td>1 (3.33%)</td>
<td>7 (23.3%)</td>
<td></td>
<td></td>
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</tr>
</tbody>
</table>

4 DISCUSSION

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with other molecules, such as MDM2 oncogenes, and viral genes, or even mutations that cause damage to the protein due to the insertion of a stop codon prematurely.

5 CONCLUSION

Expression of p53 was significantly associated with muscle invasiveness of bladder cancer. Prospective study is needed to investigate further its potential in predicting muscle invasiveness of bladder cancer.

REFERENCES


