SENSITIVITY AND SPECIFICITY OF URINARY BLADDER CANCER ANTIGEN FOR DIAGNOSIS OF BLADDER TUMOR; A COMPARATIVE STUDY WITH URINARY CYTOLOGY

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Abstract- Cystoscopy and urinary cytology are currently the basis for diagnosis and follow-up of bladder tumors. Research to find a sensitive and specific tumor marker for diagnosis of bladder tumor is actively underway, however, due to low sensitivity and high cost of cytology. This cross-sectional study was performed in 65 patients to evaluate whether urinary bladder cancer (UBC) antigen level can predict the presence of active bladder tumor. In patients with inactive tumor, UBC antigen level was determined in addition to standard cystoscopy and cytology for follow-up. Patients with active tumor were subjected to standard treatment and UBC antigen level determination. UBC antigen levels were measured by ELISA, using monoclonal antibodies specific for UBC antigen. As a control group, UBC antigen level was also determined in 65 persons who had been referred for urinalysis for other reasons. UBC antigen level more than 1 μg/L which was regarded as positive was found in 49.4% of the patients. In control group, 96.9% had UBC antigen $< 1\mu g/L$. Mean UBC antigen level in patients was 3.77 μg/L while it was 0.508 μg/L in controls ($P < 0.0001$). Sensitivity of UBC antigen was 53.3% and its specificity was 40%. Sensitivity and specificity of urinary cytology was 17.3% and 88.2%, respectively. This difference was statistically significant ($P < 0.001$). UBC antigen is more sensitive than urinary cytology, although cytology still retains its priority in specificity. It is not yet recommended to replace UBC antigen for cytology due to its low specificity and not favorable sensitivity.
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INTRODUCTION

Transitional cell carcinoma (TCC) of bladder is the second most common malignancy of the genitourinary tract. More than 90% of bladder cancers are TCC. Seventy-five percent of cases are superficial whereas 20% show muscle invasion and 5% have distant metastasis (1). Seventy percent of bladder cancers recur after treatment and 30% of cases experience progression (2). Early diagnosis and proper staging are the basis of cancer treatment but follow-up is of special importance due to a high incidence of recurrence.

Cystoscopy and urinary cytology are accepted as standard modalities for follow-up. Urinary cytology costs about 100 dollars and is not appropriate for mass screening due to its high expense and low sensitivity. Urinary cytology has a sensitivity of 20-40% in low-grade tumors, with false negative and false positive values of 20% and 1-12%, respectively (3). Efforts to substitute cytology with a more sensitive and specific test with lower cost are under way. Currently, bladder tumor markers are under extensive research and urinary bladder cancer (UBC) antigen is one of the tumor markers with an acceptable sensitivity and specificity in primary

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reports (4). UBC antigen test is available in the market with an acceptable price. It is a monoclonal test using enzyme-linked immunoassay against cytokeratin 8 and 18 epitopes. More than 20 cytokeratins have been detected in human and most of them are specific for epithelial cells and their expressions have different patterns in normal and malignant epithelium (5). The monoclonal antibody for UBC does not cross react with other urinary antigens. It is suspected that UBC measures tumor activity instead of tumor burden, which means better detection rate at primary stages. This study evaluates the sensitivity and specificity of this biomarker for diagnosis and follow-up of bladder tumor and compares this marker with conventional cytology.

MATERIALS AND METHODS

This is a cross-sectional, non-interventional study. From April 2002 to February 2003, 65 patients with bladder tumor, either with active tumor or with history of bladder tumor who returned for regular follow-up were enrolled in this study. We obtained informed consent from all patients.

The patients underwent standard cystoscopy and conventional cytology in addition to UBC antigen determination. The UBC antigen level was measured by ELISA technique. The monoclonal antibodies used in the test were specific for UBC antigen (cytokeratin 8 and 18) without detectable cross reactivity to other cytokeratins. UBC antigen levels lower than 1 µg/L was interpreted as negative. UBC antigen level was also determined in 65 other patients who had been referred randomly for urinalysis to the reference laboratory for other reasons. Cystoscopy was taken as the gold standard of tumor presence and was performed by a single urologist. UBC antigen level determination and cytology were performed in one laboratory under supervision of one cytopathologist. In case of active tumor, standard treatment plan was undertaken and tumor grade and stage were determined according to pathology and imaging. In case of inactive tumor in follow-up, the primary tumor grade and stage were adopted as reference. In addition, patients' age, gender, and history of previous treatments were asked.

RESLUTS

The mean age of patients was 63 years while the control group had a mean age of 34.54 years. Mean UBC antigen level was 3.77 ± 5.02 µg/L (0.1-15 µg/L) in patients; 50.6% of them had a UBC level below 1 µg/L which was interpreted as negative and 49.4% had a UBC antigen level more than 1 µg/L which considered positive. In control group, the mean UBC antigen level was only 0.508 ± 0.311 µg/L (0.1-1.3 µg/L); 96.9% had UBC level less than 1 µg/L and only 3.1% were found to have UBC antigen more than 1 µg/L. Mean UBC antigen level in patients and controls had statistically significant difference (independent t test, $P < 0.0001$).

Urinary cytology was reported positive in 9.4% of patients while 70.3% had negative cytology and 20.3% had a suspicious interpretation by the cytopathologist. On cystoscopy, 46.2% of the patients had active tumor while in 46.2% tumor activity was not found. A suspicious lesion was found in 7.7% on cystoscopy but biopsy results did not prove an active tumor. The pathology results showed TCC in 61 patients (93.8%) and squamous cell carcinoma in 4 patients (6.2%). In TCC group, low, intermediate and high-grade tumors were found in 55.7%, 18% and 26.1% of cases, respectively. All squamous cell carcinomas were high grade tumors with muscle invasion.

The number and percentage of the patients with different tumor grades and UBC antigen results are shown in table 1. No statistically significant difference was observed between different grades and positive UBC antigen.

Mean UBC antigen level was 3.67 ± 5.08 µg/L in low grade TCC group and 3.41 ± 4.7 µg/L in intermediate grade TCC group and no statistically

Table 1. UBC antigen in different tumor grades*

Grade	UBC-	UBC+	Total
Low	17 (26.1%)	17 (26.1%)	34 (52.3%)
Intermediate	5 (7.6%)	6 (9.2%)	11 (16.8%)
High	7 (10.7%)	13 (20%)	20 (30.7%)
Total	29 (44.6%)	36 (55.3%)	65 (100%)

Abbreviation: UBC, urinary bladder antigen.
* Data are given as number (percent).

significant difference was detected between these two groups (*t* test, *P* = 0.89). Mean UBC antigen level was 4.77±5.51 μg/L in high-grade group which showed no statistically significant difference with low-grade and intermediate-grade groups (*t* test, *P* = 0.46 for low-grade; *t* test, *P* = 0.66 for intermediate-grade). Table 2 shows UBC antigen values in different tumor stages and table 3 compares UBC antigen levels in different tumor activity states. No statistically significant correlation was observed between UBC antigen, tumor stage and tumor activity.

UBC antigen sensitivity was 53.3% and the sensitivity of cytology was 17.3% in our cases. The specificities of UBC antigen and cytology were 40% and 88.2%, respectively. Sensitivity and specificity of UBC antigen were, respectively, 40% and 40.4% in low-grade tumors, 40% and 25% in intermediate-grade tumors and 66.6% and 50% in high-grade tumors. The sensitivity and specificity of intermediate-grade tumors were not statistically reliable due to limited number of patients in this subgroup.

Sensitivity and specificity of UBC antigen were, respectively, 42% and 37.5% in tumors confined to mucosa, 0% and 40% in cases with lamina propria involvement, and 77.7% and 100% in tumor with muscle invasion; the latter two had limited number of cases and would not be statistically reliable. It seems that sensitivity was reduced significantly in patients with previous BCG treatment: while the sensitivity and specificity were, respectively, 75% and 33.3% in patients without such treatment, they were 0% and 47.3% in BCG treated patients. The limited number of patients with other intravesical or systemic treatments like mitomycin was not enough for comparison.

Table 2. UBC antigen in different tumor stages*

Stage	UBC-	UBC+	Total
Mucosa confined	23(35.3%)	25 (38.4%)	48(73.7%)
LP involvement	3 (4.6%)	3 (4.6%)	6 (9.2%)
Muscle invasion	3 (4.6%)	7 (10.7%)	10(15.3%)
Metastatic	0 (0%)	1 (1.5%)	1 (1.5%)
Total	29(44.6%)	36 (55.3%)	65 (100%)

Abbreviations: UBC, urinary bladder antigen; LP, lamina propria.
* Data are given as number (percent).

Table 3. UBC antigen in different tumors activities*

Tumor activity	UBC-	UBC+	Total
Not active	12(18.4%)	18(27.6%)	30(46.1%)
Active	14(21.5%)	16(24.6%)	30(46.1%)
Suspicious	3(4.6%)	2(3%)	5 (7.6%)
Total	29(44.6%)	36(55.3%)	65 (100%)

Abbreviation: UBC, urinary bladder antigen.
* Data are given as number (percent).

DISCUSSION

Finding a sensitive and specific tumor marker for reliable diagnosis and follow-up of a tumor will improve detection in lower stages and thus improving treatment results. It may help to monitor treatment results to detect probable recurrence early. The best example of the tumor marker is prostate-specific antigen (PSA), which has improved detection rate of prostate cancer in lower stage when definitive treatments are provided; PSA is also the best monitor of treatment.

Looking for such a tumor marker in bladder tumor is currently under investigation. Unfortunately, we are not equipped with such a marker up to now. Several tumor markers have been studied with different sensitivities and specificities but not with acceptable results. A few of these tumor markers like NMP22, BTA, telomerase, and UBC antigen have shown promising results (4). Giannopoulos *et al.* found sensitivity and specificity of 80.5% and 80.2% for UBC in 213 patients. The sensitivity of UBC antigen in stage Ta was 80.8%, which was significantly higher than NMP22 and BTA-Stat. In stage T1 the sensitivity of UBC antigen was the same with the other two tumor markers. They concluded that the sensitivity of UBC antigen in lower stages is higher than BTA-Stat and NMP22 (4). Sunchez *et al.* reported a sensitivity and specificity of 70% and 95%, respectively (6). However they found the sensitivity of cytology to be only 7%, significantly less than what they reported for UBC antigen, but cytology remained more specific in their study. Sumi *et al.* concluded that the sensitivity of UBC was higher than cytology (82% versus 60.7%); meanwhile the sensitivity of UBC antigen and cytology in low-grade TCC was 76.5% and 11.8%, respectively (7).

On the other hand, Mungan did not find UBC

valuable in follow-up of patients with bladder tumor (8). Heicappell and associates also concluded that UBC antigen play little role in diagnosis of bladder tumor; they reported the sensitivity of UBC antigen to be 21.6% in stage pTa and 75% in stage pT4, with a specificity of 76.6% (9).

The results of our study showed a lower sensitivity and specificity compared with other studies but still in parallel with cytology which was also found to be less sensitive and specific than similar studies. These could be due to the low standard of our laboratories, defect in delivery of washout cytology, or limited number of patients and short follow-up time in this study. The short time of follow-up in this study prevented us to study the predictive value of UBC antigen for tumor recurrence that needs long follow-ups and well-designed studies with adequate number of patients.

In conclusion, although UBC antigen shows better sensitivity than cytology we still do not recommend replacing UBC antigen for cytology due to its lower specificity and not still favorable sensitivity. It is not recommended to use UBC antigen for bladder tumor screening due to its limited sensitivity and specificity in high-risk groups. However, using UBC antigen in addition to cytology may give better sensitivity, considering the UBC antigen is much less costly than cytology.

REFERENCES

1. Messing EM, Young TB, Hunt VB, Gilchrist KW, Newton MA, Bram LL, Hisgen WJ, Greenberg EB, Kuglitsch ME, Wegenke JD. Comparison of bladder cancer outcome in men undergoing hematuria home screening versus those with standard clinical presentations. Urology. 1995 Mar; 45(3):387-396.

2. Burchardt M, Burchardt T, Shabsigh A, De La Taille A, Benson MC, Sawczuk I. Current concepts in biomarker technology for bladder cancers. Clin Chem. 2000 May; 46(5):595-605.

3. Badalament RA, Hermansen DK, Kimmel M, Gay H, Herr HW, Fair WR, Whitmore WF Jr, Melamed MR. The sensitivity of bladder wash flow cytometry, bladder wash cytology, and voided cytology in the detection of bladder carcinoma. Cancer. 1987 Oct 1; 60(7):1423-1427.

4. Giannopoulos A, Manousakas T, Gounari A, Constantinides C, Choremi-Papadopoulou H, Dimopoulos C. Comparative evaluation of the diagnostic performance of the BTA stat test, NMP22 and urinary bladder cancer antigen for primary and recurrent bladder tumors. J Urol. 2001 Aug; 166(2):470-475.

5. Fuchs E. Keratins as biochemical markers of epithelial differentiation. Trends Genet. 1988 Oct; 4(10):277-281.

6. Sanchez-Carbayo M, Urrutia M, Hernandez-Cerceno ML, Gonzalez de Buitrago JM, Navajo JA. [Cytokeratins (UBC and CYFRA 21-1) and nuclear matrix proteins (NMP22) as urine tumor markers in the diagnosis of bladder cancer]. Med Clin (Barc). 2000 Mar 18; 114(10):361-366.

7. Sumi S, Arai K, Kitahara S, Yoshida KI. Preliminary report of the clinical performance of a new urinary bladder cancer antigen test: comparison to voided urine cytology in the detection of transitional cell carcinoma of the bladder. Clin Chim Acta. 2000 Jun; 296(1-2):111-120.

8. Mungan NA, Vriesema JL, Thomas CM, Kiemeney LA, Witjes JA. Urinary bladder cancer test: a new urinary tumor marker in the follow-up of superficial bladder cancer. Urology. 2000 Nov 1; 56(5):787-792.

9. Heicappell R, Schostak M, Muller M, Miller K. Evaluation of urinary bladder cancer antigen as a marker for diagnosis of transitional cell carcinoma of the urinary bladder. Scand J Clin Lab Invest. 2000 Jul; 60(4):275-282.