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European Association of Urology

Bladder Cancer

Quantitative Cytology on Bladder Wash versus Voided Urine: A Comparison of Results

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Article info

Article history:
Accepted January 23, 2006
Published online ahead of print on February 13, 2006

Keywords:
Bladder cancer
Diagnosis
Marker
Quantitative cytology
Quanticyt

Abstract

Objectives: Quantitative cytology (Quanticyt®) is a valuable marker for the identification of high-risk superficial bladder cancer (SBC) patients and can be used to individualize surveillance of patients. A disadvantage is the necessity to perform an invasive procedure to obtain the required bladder wash sample. This study investigated whether quantitative cytology can be performed on voided urine with reliable results, consistent with the quantitative cytology performed on bladder wash samples.

Methods: Between June 2003 and May 2005, 288 voided urine samples in combination with bladder wash samples were obtained from patients with SBC who visited our urologic outpatient department. Quantitative cytology was performed on all samples. Corresponding clinical pathologic features and washed cytopathology results were collected. Linear regression analyses were performed for comparison of results from both types of samples.

Results: Ninety-one percent of the samples fell into the low or intermediate region on bladder wash. A clear deviation in the nuclear shape (MPASS) was seen in the voided urine samples, which led to more low-risk results. The clinical characteristics show that this shift is not the result of under-staging. The nuclear content (2c deviation index [DI]) did not change by performing the analysis on urine.

Conclusion: When urine is correctly processed after voiding, quantitative cytology can be done on these samples. Voided urine-based quantitative cytology can be implemented in daily practice.

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0302-2838/$ – see back matter doi:10.1016/j.eururo.2006.01.029

1. Introduction

The combination of urethrocystoscopy and cytology is currently used for surveillance of patients with superficial bladder cancer (SBC). The limitations of both are well known. Cystoscopy is an invasive procedure and carcinoma in situ lesions may easily be missed. To compensate, cytology is used, which has a high sensitivity for these high-grade lesions and a high overall specificity. Unfortunately, it lacks sensitivity for low-grade lesions. Moreover, it has a low interobserver and intraobserver reproducibility [1]. One of the goals in attempts to improve of surveillance in patients with SBC is prevention of unnecessary cystoscopies. To achieve this, urinary markers are being developed with a higher sensitivity than cytology and a specificity comparable to cystoscopy. Quantitative cytology is a method of cellular analysis that has already proven to be superior to urine cytology for the identification of patients at risk for tumor recurrence and progression [2,3]. Sensitivity and specificity are 95.2% and 65%, respectively, on the condition that presence of tumor is assumed when the result is in the intermediate- or high-risk region. The subsequent risk of a recurrence within 1 yr for the low-, intermediate-, and high-risk regions is 5.5%, 12.5%, and 20.5%, respectively. The risk of progression at 1 yr is 0.54%, 1.25%, and 6.34%, respectively [2] (Fig. 1). In addition it appeared that a 2c deviation index (2cDI) cut-off of 2.00c can be used to further stratify high-risk quantitative bladder wash cytologic findings. With a median time to progression of 20 mo, patients with a 2cDI $\geq$ 2.00c showed a risk of progression of 43% versus 13% for the patients with a 2cDI $<$ 2.00c [4]. Moreover, addition of consecutive quantitative cytology to urine cytopathology evaluations improves the detection of high-grade lesions and provides a more accurate prediction of tumor stage

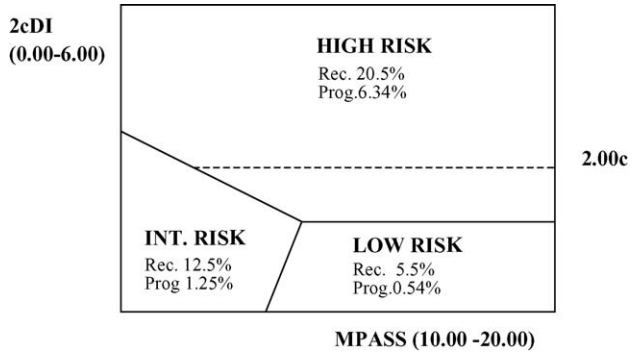

Fig. 1 – Report form of quantitative cytology (bladder wash) risk groups with recurrence (rec) and progression (prog) after 1 yr of follow-up [2].

[5]. A disadvantage of quantitative cytology is the need for an invasive procedure to obtain the required bladder wash sample. Earlier attempts to perform the procedure on voided urine samples did not succeed because of the vulnerability of the urine cells. Therefore, adaptations have been made in the method of the analysis, that is, the time factor, the method to increase the number of cells in the sample, and the fixative used. This study compared conventional quantitative cytology on bladder wash, with the new technique performed on samples of voided urine.

2. Materials and methods

2.1. *Patients*

Between June 2003 and May 2005, 288 samples were obtained from patients who visited our outpatient department. Patients who underwent cystoscopy for surveillance of SBC or for the evaluation of symptoms suspicious for bladder cancer were eligible for the study. If intravesical instillations were given, the last treatment was at least 6 wk prior. Both a voided urine sample and a bladder wash sample were obtained just before and during cystoscopy, respectively. Quantitative cytology was performed on all samples.

2.2. *Quantitative cytology*

Quantitative cytology is an automated karyometric analysis, using a personal computer-based image analysis system. On each slide, 50 randomly selected images containing at least 100 nuclei were analyzed. For each nucleus present in the images, the DNA content (i.e., the 2cDI according to Böcking et al. [6]) and the nuclear shape feature PASS (based on the smoothed Freeman difference chain code) were determined (Fig. 2). Based on these features patients are divided into risk groups. This division is based on correlation with histology data. Normal samples were found mainly in the right lower quadrant of the report form and the low-grade malignancies in the left lower quadrant; the grade increased with the 2cDI [7,8]. On the report form these groups are called, respectively, low, intermediate, and high risk.

2.3. *Voided urine sample*

The voided urine samples were obtained at the outpatient department prior to cystoscopy. The samples arrived at the laboratory within 30 min, where they were centrifuged (771 g for 10 min) and the pellet was fixed with Boonfix® (commercially available at Leiden Cytology and Pathology Laboratory, Leiden, The Netherlands). Further staining and karyometric analysis were performed as described by van der Poel et al. [2].

2.4. *Bladder wash sample*

The bladder wash material was obtained by rinsing the bladder at least twice with 25 cc saline solution through the