

Color and Morphology Combination for Detection of Low-Grade Urothelial Cell Cancer: A Multi-Center Validation Study

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Introduction

Urothelial cell carcinoma (UCC) is the most common malignancy in the urinary system. With up to 80% recurrence rate, UCC requires lifelong surveillance and is thus considered one of the most costly cancers in terms of lifetime expenditure per patient. CellDetect[®] is a novel histochemical platform allowing for color and morphological discrimination between normal and neoplastic cells. The technology has been validated in several preclinical and clinical studies in multiple types of cancers. Recently, the technology was implemented in a preliminary study of bladder cancer, demonstrating high sensitivity in detecting both low and high grade tumors. The present multicenter trial was designed to validate these findings and further explore its utility in patients undergoing routine bladder cancer monitoring.

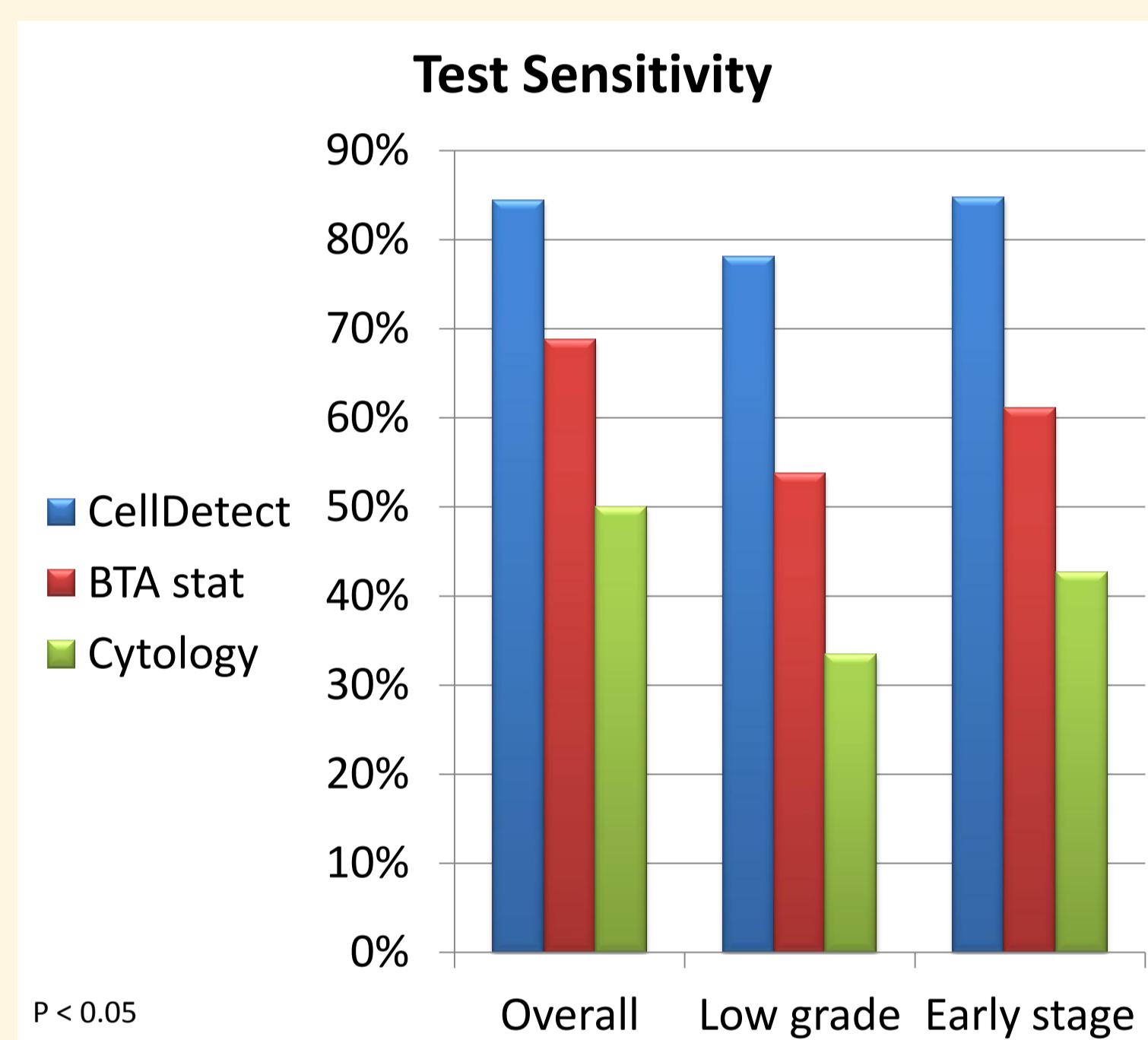
Methods

The study was designed as a multi-center, prospective, longitudinal trial involving 9 medical centers. Voided urine samples were collected from subjects previously diagnosed with UCC who were undergoing routine outpatient surveillance or scheduled for transurethral resection or radical cystectomy. Urine samples were tested for BTA stat[®] test and standard urine cytology, and the remaining urine volume was processed to cytospin smears. Smears were stained by CellDetect[®] and assessed under a light microscope by two independent cytologists. Gold standard for negative cases was cystoscopy and biopsy for positive cases.

Results

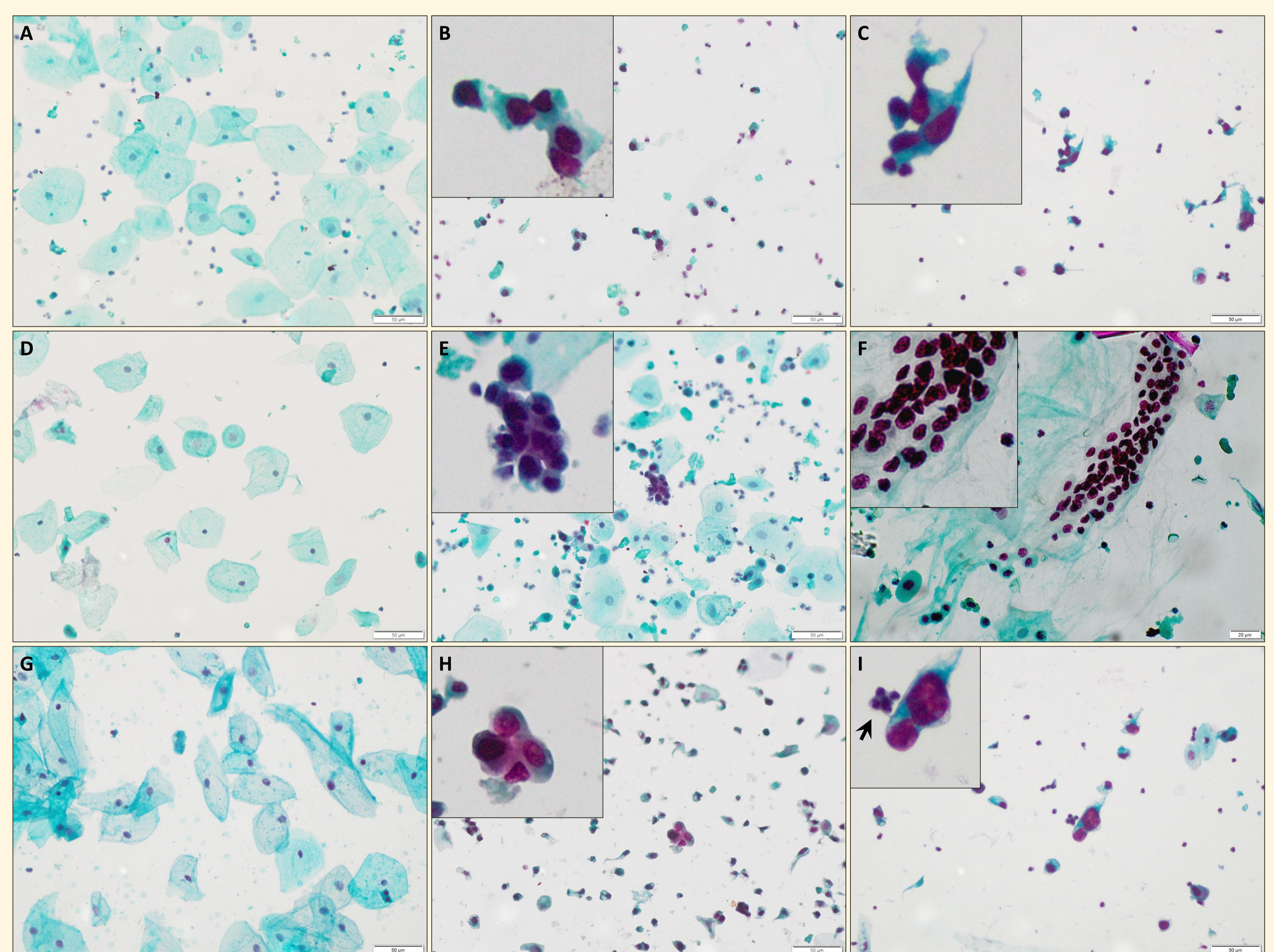
CellDetect[®] demonstrates high sensitivity for the detection of early stage, low grade tumors

Diagnosis	No. of Subjects
Negative	121
Positive	96
Total	217
Cancer grade	
Low grade	41
High grade	54
Undetermined	1
Total	96
Cancer stage	
Ta, T1, Tis	72
T2 and up	22
Undetermined	2
Total	96



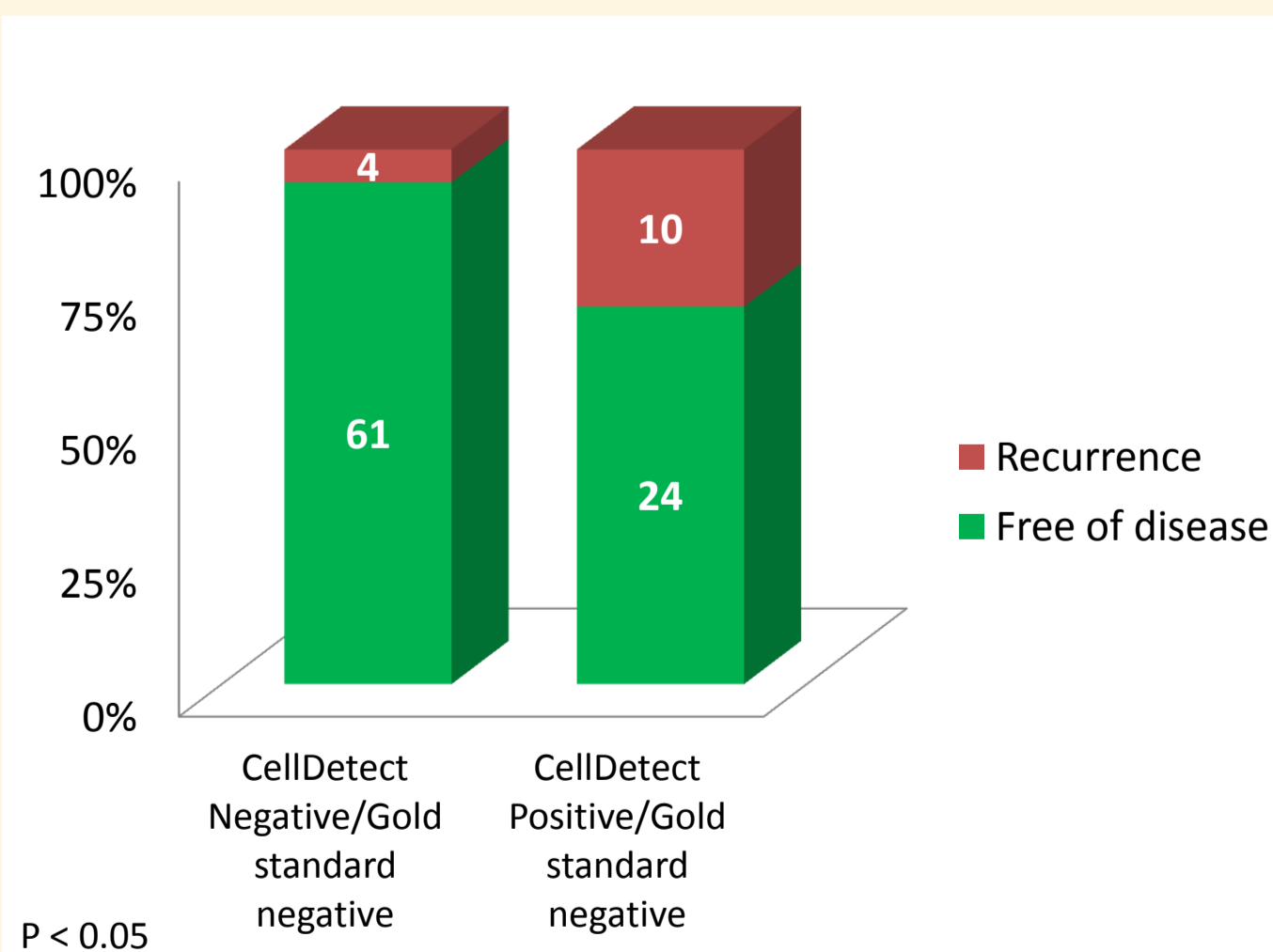
Diagnosis	CellDetect [®]	BTA stat [®]	Cytology
Specificity			
Negative cystoscopy group	84%	76%	89%
Sensitivity			
Biopsy positive group	84%	69%	50%
Low grade	78%	54%	33%
High grade	89%	82%	68%
Stages Ta, T1, Tis	85%	61%	43%
Stage T2 and up	82%	91%	77%

Images of urine smears stained by CellDetect[®]



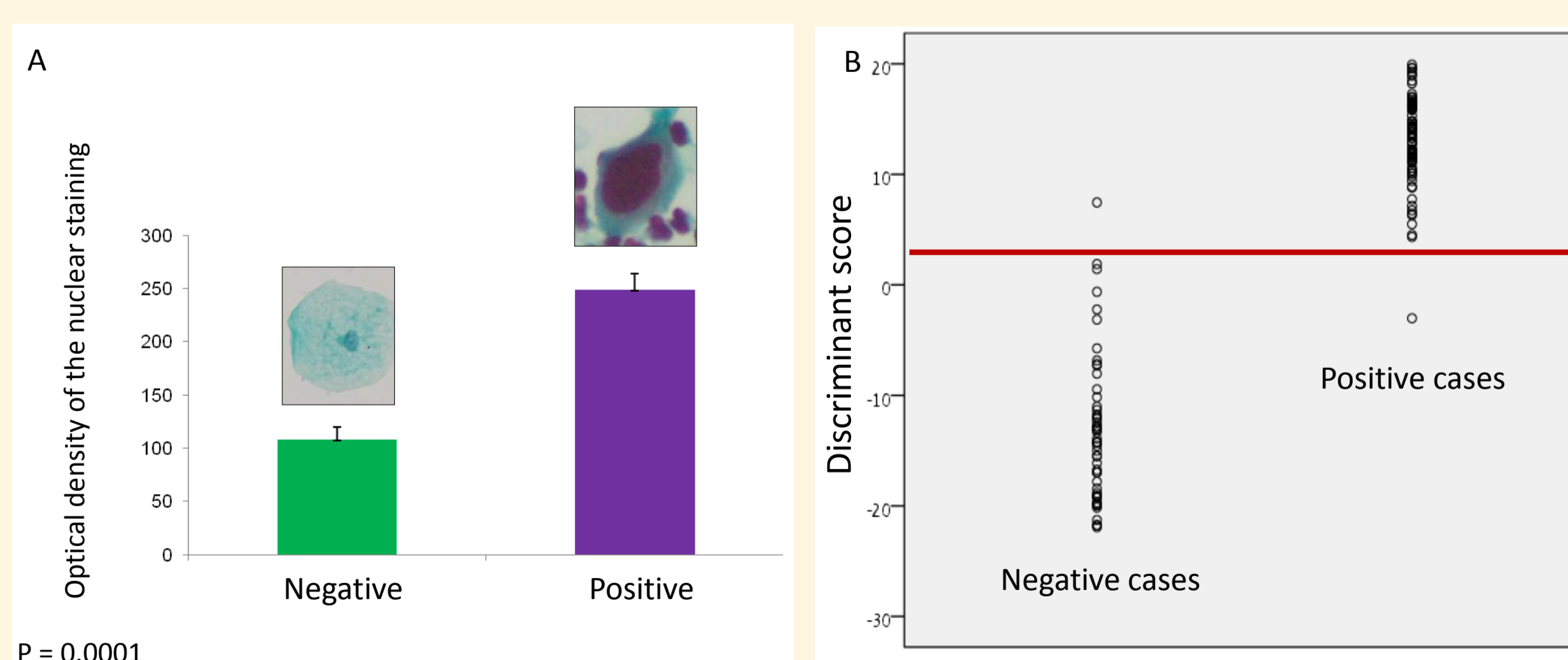
A-I are images of urine cytospin smears of normal subjects (A,D,G), low grade UCC (B,E,H) or high grade UCC (C,F,I). Epithelial cells are stained in green while dysplastic cells exhibiting reddish-purple nuclei (digital zoom in the boxes). Inflammatory cells are occasionally stained in purple (I, marked with arrows). Magnification: x40

Follow up on patients with positive CellDetect[®] results and negative gold standard shows a significant increase in recurrence within the following 14 months



Follow up on patients with a positive CellDetect[®] result and negative gold standard revealed a significant elevation (p value < 0.05) in the recurrence rate (positive cystoscopy and/or cytology) among this group (30%) in comparison to true-negative cases (5%), providing a supportive evidence that CellDetect[®] is capable of early detection of UCC recurrence.

Digital analysis of slides stained by CellDetect[®] accurately identified UCC



A digital analysis of 142 images (37 cases) was performed using the Image Pro-Plus software. The variables that were extracted from the morphometric analysis included morphology and tinctorial parameters (see example in A). The result showed nearly complete separation between positive and negative images (B). Area under the curve was 99.6% (high grade tumors) and 99% (low grade tumors).

Conclusions

This study validates the capability of CellDetect[®] in accurately identifying UCC in urine smears. Of special importance is that the high sensitivity was evident in low grade and early stage tumors, often missed by other non-invasive tests. Employing an on-the-shelf digital algorithm showed that automated analysis of the smears provides excellent results in detection of both high grade and low grade tumors. These data demonstrates that the CellDetect[®] stain can be utilized as a non-invasive test for monitoring patients for UCC recurrence.