Figure 3. Cytology and GS_D2 results based on risk groups on tumour samples. Abbreviations: NMIBC, Non-muscle invasive bladder cancer; MIBC, Muscle invasive bladder cancer.

Clinical validation of a gene expression test for the non-invasive diagnosis of bladder cancer: A prospective, blinded, international and multicenter study.

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OBJECTIVE

Current standard methods used to detect and monitor bladder cancer (BC) are invasive or have low sensitivity. We have previously reported four non-invasive tests for BC diagnosis based on the gene expression patterns of urine[1]. Here, we present the clinical validation of our test in a prospective, blinded, international and multicenter study.

METHODS

•Consecutive voided urine samples from BC patients and controls were collected in five European centres (n=789). Finally, 525 samples were successfully analyzed (216 tumour and 309 control urines).

•Gene expression values were quantified using TaqMan Arrays. The same cut-off as previously reported for discrimination between tumour and controls was used in this study.

•Results from the most accurate gene signature were correlated to clinical parameters, such as cytology results, tumour multiplicity and tumour size using ANOVA test.

	HOSPITAL CLINIC	FUNDACIO PUIGVERT	VIRGEN DEL ROCIO	UNIVERSITY OF VIENNA	RADBOUD UNIVERSITY NIJMEGEN
N	59	115	22	14	6
Median age (range)	72 (51-88)	74 (38-90)	65 (45-83)	72 (59-94)	65 (30-83)
Stage					
Tx	1	2	1	-	-
Cis	2	-	-	-	-
Ta	20	70	8	7	4
T1	26	20	11	4	2
≥T2	10	23	2	3	-
Grade					
Gx	-	-	1	-	-
LG	21	44	14	8	2
HG	38	71	7	6	4

Table 1. Pathological features of tumour samples included in each participating centre **(LG: Low-grade; HG: High grade).**

RESULTS

Table 2. Gene expression signatures for BC diagnosis

Gene symbol	GS_D12	GS_D10	GS_D5	GS_D2
IGF2	■	■	■	■
MAGEA3	■	■	■	■
KLF9	■	■	■	
CRH	■	■	■	
SLC1A6	■	■	■	
POSTN	■	■		
EBF1		■		
CFH		■		
MCM10		■		
MMP12		■		
TERT	■			
AHNAK2	■			
ANXA10	■			
CTSE	■			
KRT20	■			
PPP1R14D	■			

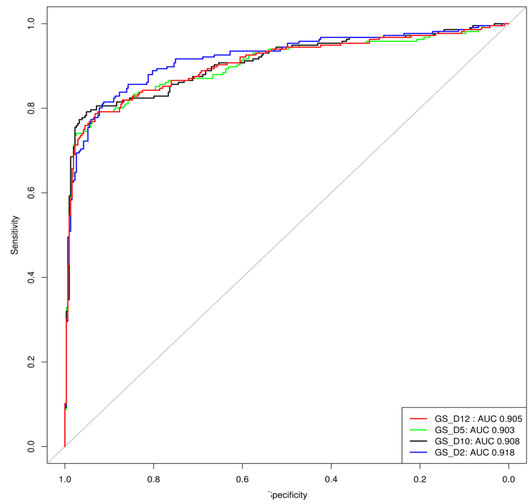

Figure 1. ROC curves of the 4 diagnostic gene expression signatures in the multicenter validation set of 525 samples.

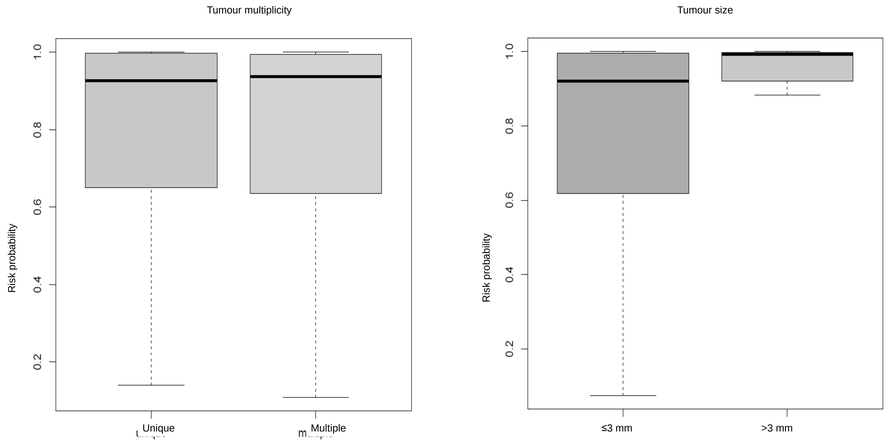

Figure 2. Correlation between the performance of GS_D2 and tumour multiplicity and size.

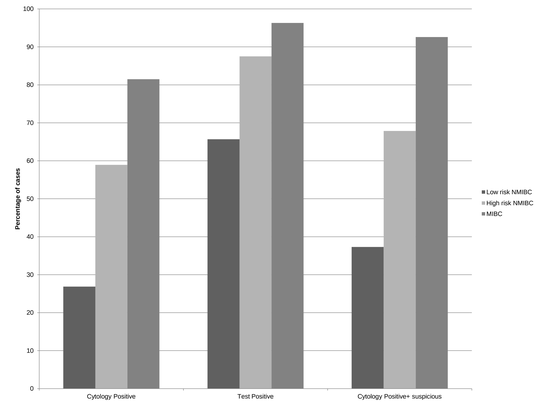

Figure 3. Cytology and GS_D2 results based on risk groups on tumour samples. Abbreviations: NMIBC, Non-muscle invasive bladder cancer; MIBC, Muscle invasive bladder cancer.

CONCLUSIONS

✓Our GS_D2 test is non-invasive, non-observer dependent, non labour-intensive and requires no special technology. So it is a low cost technique, and has demonstrated diagnostic accuracy in an independent, international and multicenter study, equal or superior to the current gold standard. Consequently, our two gene signature meets the requirements to be considered a molecular test applicable to clinical practice in the management of BC.

(1) Mengual L, et al. Validation study of a non-invasive urine test for diagnosis and prognosis assessment of bladder cancer. Evidence for improved models. ***J Urol****, 2014 Jan;191(1):261-9.*