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The Final thesis

Follow-up of Superficial Urothelial Carcinoma with non-Invasive Diagnostic Tools

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Table of contents

I. INTRODUCTION

1. EPIDEMEOLOGY

Bladder cancer makes up 3% of all cancer cases with around 614,000 new diagnoses and 220,000 deaths recorded in 2022 as, per GLOBOCAN data. It mainly impacts individuals aged 55 and older showing a prevalence among men compared to women (1). In cases of upper tract urothelial carcinoma (UTUC) where the male to female ratio ranges from 1.5 to 2.1 (2). Factors like smoking for half to two thirds of cases and exposure to toxins are significant risk factors. In regions like Southern Europe such as Spain there are rates of bladder cancer incidence observed; meanwhile UTUC prevalence stands out among women in Bosnia and Croatia (3). While there has been a decrease or stabilization in bladder cancer rates among men since the '90s in some countries due to smoking rates (4) there is an upward trend in cases among women attributed to increased smoking habits (5). Mortality rates have seen a decline in countries with human development indexes (HDI) due to smoking rates and better healthcare services. However, Egypt has reported an increase, in deaths related to bladder cancer (4).

Bladder cancer is mostly classified as non-muscle invasive cancer making up, around 75% of cases. On the hand UTUC accounts for 5-10% and urethral carcinoma is rare representing less than 1% of all genitourinary cancers (2). Interestingly about two thirds of individuals diagnosed with UTUC have muscle disease from the start, which's higher compared to the 15-25% prevalence seen in newly diagnosed bladder cancer cases (6).

2. OVERVIEW OF SUPERFICIAL UROTHELIAL CARCINOMA

Any neoplastic growth that arises from the urothelial lining in the urinary tract from the calyces to the renal pelvis, ureter and bladder, falls under the term “urothelial carcinoma” (7). Superficial bladder cancer includes papillary tumors confined to the mucosa (stage Ta) and those invading the lamina propria (stage T1) along with flat high-grade tumors confined to the mucosa (CIS) or (Tis) (12). Histologically, urothelial carcinoma can present itself in a various morphologies due to its

heterogeneous nature. Some of the variants include nested, microcystic, sarcomatoid, glandular, clear cell, plasmacytoid, small cell carcinoma, and adenocarcinoma (8). Tumorigenesis in urothelial carcinoma involves a series of molecular processes, such as the activation of oncogenes like HRAS and FGFR3 the inactivation of tumor suppressor genes such as TP53 and RB1 and the disturbance of signaling pathways like PI3K/AKT and MAPK (9) (10). Additional factors contributing to tumor growth include DNA repair mechanisms, abnormal epigenetic changes like DNA methylation and histone modifications and disruptions, in cell cycle regulation involving CDKN2A (11).

TNM STAGING:

Table 1

AJCC TNM Staging System for Bladder Cancer 8th ed., 2017	Histologic Grade (G)
LG Low-grade	Low grade
HG High-grade	High grade
GX	Grade cannot be assessed
G1 Well differentiated	Well differentiated
G2 Moderately differentiated	Moderately differentiated
G3 Poorly differentiated	Poorly differentiated

AJCC Prognostic Groups	T	N	M
Stage 0a	Ta	N0	M0
Stage 0is	Tis	N0	M0
Stage I	T1	N0	M0
Stage II	T2a, T2b	N0	M0

Stage IIIA	T3a, T3b, T4a	N0	M0
AJCC Prognostic Groups	T	N	M
	T1-T4a	N1	M0
Stage IIIB	T1-T4a	N2, N3	M0
Stage IVA	T4b	Any N	M0
	Any T	Any N	M1a
Stage IVB	Any T	Any N	M1b

Abbreviations: Ta - Noninvasive papillary carcinoma, Tis - Urothelial carcinoma in situ, T1 - Tumor invades lamina propria, T2 - Tumor invades muscularis propria, T3 - Tumor invades perivesical tissue, T4 - Extravesical tumor directly invades adjacent organs or structures, N0 - No regional lymph node metastasis, N1 - Single regional lymph node metastasis, N2 - Multiple regional lymph node metastases, N3 - Lymph node metastasis to common iliac lymph nodes, M0 - No distant metastasis, M1a - Distant metastasis limited to lymph nodes beyond the common iliacs, M1b - Non-lymphnode distant metastases. (NCCN guidelines 2024) (12) (The original source for this information is the AJCC Cancer Staging Manual, Eighth Edition (2017) published by Springer International Publishing)

TREATMENT:

Transurethral resection of bladder tumor (TURBT) is the cornerstone in diagnosing and treating non-muscle-invasive bladder cancer (NMIBC). It aims to achieve radical tumor resection while providing specimens for accurate pathological diagnosis and follow-up (13).

Bacillus Calmette–Guérin (BCG) stands as an option for therapy in high risk NMIBC cases (14), Derived from Mycobacterium bovis despite its mechanism of action remaining unknown after more than four decades of clinical use. It is considered essential for the treatment, effectively lowering the chances of progression to muscle-invasive (15). Nevertheless, BCG is ineffective in around 40% of cases leading to the need for alternative treatments like cystectomy or bladder sparing options (16).

Patients who do not respond well to BCG treatment are typically categorized as refractory relapsing or intolerant with options for further intravesical therapy (16). In cases of bladder cancer radical cystectomy is still considered the treatment to halt disease progression according to guidelines, from the European Association of Urology (17).

3. CURRENT DIAGNOSTIC AND FOLLOW-UP GUIDLINES

Current Diagnostic guidelines:

Bladder cancer often manifests with painless hematuria, requiring a multi directional investigation approach. Common symptoms like dysuria, increased frequency, and urgency (18). Imaging techniques such as bladder ultrasonography or cross-sectional imaging aid in detecting intraluminal bladder masses. However, the definitive diagnosis relies on cystoscopic examination, and histological evaluation through cold-cup biopsy or transurethral resection of the bladder tumor (TURBT) (18). The complete resection of all tumor tissue including the lamina propria and detrusor muscle is essential for accurate staging in most cases (18).

Concurrent carcinoma in situ (CIS) signifies an adverse prognosis, requiring bladder biopsies from suspicious urothelium or mapping biopsies from normal-looking mucosa, especially in patients with positive urine cytology or a history of high-grade non-muscle-invasive bladder cancer (18). High-risk NMIBC patients, especially those with CIS, should undergo upper tract imaging to screen for synchronous upper urinary tract urothelial carcinoma using computed tomography (CT) urography or magnetic resonance imaging (MRI) urography (18).

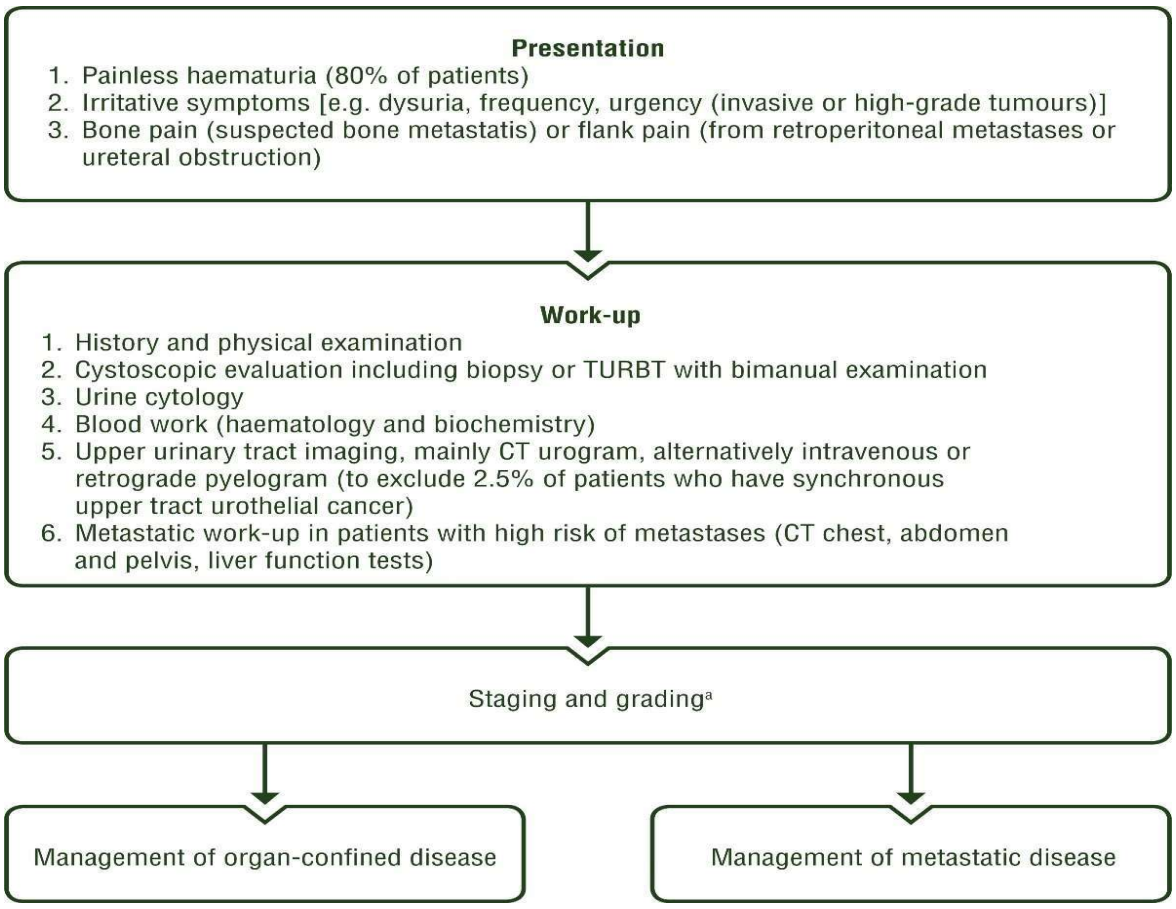

Figure 1. (19)

Current Follow-up guidelines:

For patients with low-risk non-muscle invasive bladder cancer, which is generally represented by Ta stage and grade 1 (LG/G1) tumors, the risk of progression is typically low, especially for those with small, papillary Ta LG/G1 recurrences. Given that recurrences more than five years post-diagnosis are infrequent and account for less than 2% after the first TURB (20), consideration for ceasing cystoscopy or replacing it with less invasive surveillance methods is warranted after a five-year follow-up period (21).

Patients categorized with intermediate-risk NMIBC occupy a risk spectrum that resides between low and high-risk categories. Evidence from a small, randomized controlled trial suggests it is safe to

pursue a follow-up protocol of reduced intensity for individuals with multiple and/or recurrent low-grade tumors. This approach includes a three-month post-diagnosis cystoscopy, followed by semiannual examinations for two years and annual assessments for up to ten years (19). Nevertheless, in the absence of sufficient data to support a reduced follow-up regimen for intermediate risk high grade NMIBC, similar surveillance protocols as recommended for high-risk NMIBC are advisable.

In the context of high or very high-risk NMIBC, early identification of muscle-invasive and high-grade recurrences is important due to the potential for life-threatening progression. Continuous surveillance with frequent cystoscopy and cytological analyses is essential, as tumor recurrences may occur even after a decade of no evidence of disease (19). Accordingly, a strict surveillance strategy involving regular cystoscopies and cytological studies, potentially extending throughout the patient's lifetime, is considered optimal (19).

Surveillance of the urothelium lining outside of the bladder should address the possibility of recurrence in the prostatic urethra for males and the upper urinary tract for both genders. High-risk tumors present a substantial recurrence risk in these locations, with the ten-year recurrence rate ranging from 2.8% for carcinoma in situ to 25% for those with recurrent and multifocal high-risk NMIBC(19). Urinary cytology, cystoscopy, and computed tomography urography stand as primary modalities for the early detection of extravesical recurrence(19).

4. FOLLOW-UP CHALLENGES IN EVERYDAY PRACTICE

Bladder cancer presents itself in many types, ranging from minimally invasive to highly aggressive variants. The patient's overall quality of life is tied to effective disease management. Urologists are challenged to evade the malignant potential of cancer and tailor treatment strategies accordingly. While low-risk cases may only need minimal surveillance to avoid overtreatment, high-risk cases require strict monitoring to ensure timely surgical intervention (22).

The current follow-up protocol for NMIBC is invasive and it causes significant physical and psychological discomfort for patients (23). It carries risks of side effects and increased costs. Due to the absence of robust scientific evidence, recommendations for NMIBC follow-up heavily rely on expert

opinion. The authors suggest that overdiagnosis may be occurring, particularly among patients with an intermediate risk profile (24).

Over a span of five years, the cumulative expenses of care amounted to $52,125 for low-risk, $146,250 for intermediate-risk, and $366,143 for high-risk NMIBC cases. The main factor influencing costs was the progression to muscle-invasive disease necessitating definitive treatment making up 81% and 92% of the total expenses for intermediate- and high-risk disease, respectively. Despite low-risk tumors showing a likelihood of recurrence within five years, the financial impact of recurrence constituted only 8% of the overall costs, with disease progression accounting for 71% (25).

II. METHODOLOGY

1. OBJECTIVES:

-To evaluate availability and efficacy of various non-invasive diagnostic techniques in detecting disease recurrences or progression of superficial urothelial carcinoma and provide an outlook on future directions.

2. LITERATURE SEARCH

To accomplish the objectives outlined in this study, I conducted my search using PubMed and Google scholar databases. The search strategy involved keywords such as: <Urothelial carcinoma> and < nmibc> and <upper urothelial carcinoma> and<epidemiology bladder cancer> < epidemiology upper urothelial carcinoma> <diagnostic tools urothelial carcionoma> and < diagnostic tools bladder cancer> and < Urinary biomarkers> and < Financial burden bladder cancer> and < DNA myhtelation bladder cancer> and <blood markers bladder cancer> and <Future diagnostic tools bladder cancer> and <combination of diagnostic tools bladder cancer> and < liquid biopsie> and looking up the citations

and manually selecting related articles. the focus was on articles written in English in the time frame between 2014 and 2024. The references were managed using Zotero.

3. PRISMA DIAGRAM

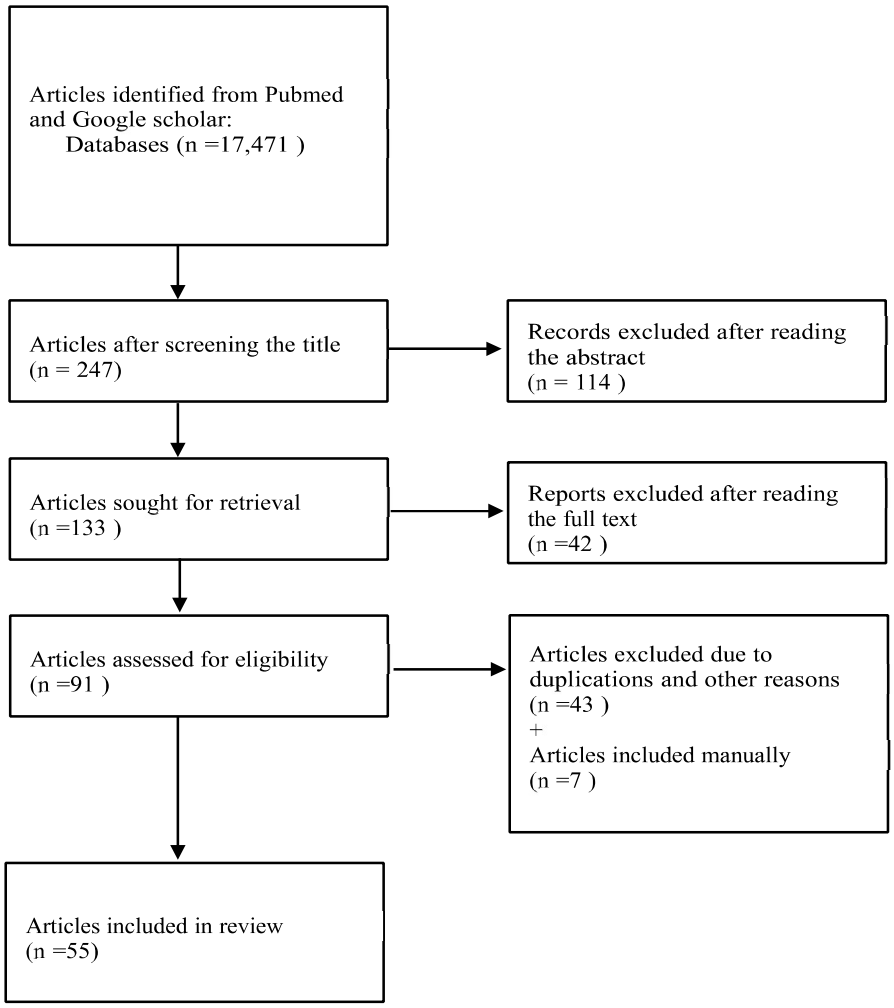

Figure 2

4. ETHICAL CONSIDERATION

This literature review adhered to the academic and ethical guidelines of Vilnius university.

It is not possible to confirm the given consent of all subjects included in the reviewed studies.

There are no conflicts of interests or sponsors causing any sort of bias.

III. RESULTS

1. Evidence Synthesis

After screening 17,478 articles, 55 were included in this literature review as stated in the PRISMA diagram.

A. Cytology

Urinary cytology has long been the gold standard for surveillance of bladder cancer paired with cystoscopy. Despite its historical significance and ease of access, urinary cytology has notable limitations, particularly in its sensitivity for low-grade tumors, with sensitivity reported as low as 16% for low-grade tumors and 84% for high-grade tumors. Recent data suggest that its initially reported high specificity rates around 95% may not always be reproducible, with specificity reported at 86%. Moreover, interpretation can be challenging in the presence of inflammation or infection. Combining cytology with urinary biomarkers, most notably NMP22 BladderChek, increased the sensitivity for high-grade tumors to 94% and for low-grade tumors to 31% (26).

B. Ultrasound

When it comes to planning follow-up strategies, ultrasound plays a crucial role in identifying bladder tumors. Ultrasound is effective in detecting tumors larger than 0.5 cm on the lateral and/or posterior

bladder walls, with an accuracy rate of 95%. This non-invasive imaging technique allows for visualizing intraluminal nonmobile masses, or areas where the bladder walls are thickened, indicating the presence of a tumor. Moreover, color flow Doppler ultrasonography helps in examining blood flow within the mass, which assists in distinguishing a tumor from conditions like blood clots. It is important to understand that while ultrasound can spot a tumor, it doesn't provide information on the stage or aggressiveness of the tumor. Therefore, it should be combined with other diagnostic methods for a thorough follow up assessment. (27)

Contrast enhanced ultrasound (CEUS) is an advancement in ultrasound imaging using ultrasonographic contrast agents (UCAs) that are made up of microbubbles containing gases in a lipid shell. These microbubbles move through the body's pathways and their detection via ultrasound happens when they interact with waves creating distinct reverberations. The uptake of UCAs enables real time staging and grading of carcinoma of the bladder (CAUB).(28) (29)

The utilization of CEUS has proven to be crucial in differentiating between low grade carcinoma. Notably CEUS shows enhancement patterns, wash in and slow wash out for high grade tumors and rapid wash in and rapid wash out for low grade tumors. The ability to distinguish bladder tumors using CEUS prospectively demonstrates sensitivity and specificity with an 86% sensitivity and 90% specificity for high grade tumors well as an 85% sensitivity and 89% specificity for low grade tumors. These results highlight the potential of CEUS in assessing and following-up bladder tumors. With an accuracy rate of 88% for both low grade cases along with its cost effectiveness and non-invasive nature CEUS emerges as a reliable tool for following-up patients with bladder tumors. (29)

C. MRI

Imaging techniques like MRI play a crucial role in the diagnosis, staging, and follow-up of bladder cancer, including superficial bladder carcinoma (27). MRI has shown promising results demonstrates moderate sensitivity ranging from 67% to 73% and specificity from 62 to 81% in detecting recurrent bladder tumors post-transurethral resection, with high accuracy particularly in identifying multifocal

abnormalities and papillary masses; 83-100% of detected multifocal abnormalities and 88-100% of detected papillary masses were tumor recurrences, but it overlooked many low grade stage Ta lesions in a study done by et al. Rosenkratz (30). It can accurately distinguish between muscle-invasive and non-muscle-invasive tumors, aiding in local staging (27). The use of multiparametric MRI and the Vesical Imaging-Reporting and Data System (VI-RADS) score further enhance its diagnostic capabilities (31) (32).

D. COMPUTER TOMOGRAPHY

CT urography stands out as the most utilized imaging method for detecting and staging bladder cancer, boasting a diagnostic accuracy estimated at 91% for detection and 35–55% for staging (27). In a study done by et al. Helenius (33) CT urography showed similar sensitivity 87% and specificity 99% in comparison to flexible cystoscopy sensitivity 87% and specificity 100%. Typically, this examination employs a three-phase protocol, encompassing noncontrast, nephrographic, and delayed phases. Bladder cancer typically manifests as an intraluminal polypoidal or nodular mass, or as localized wall thickening. During the nephrographic phase, occurring 100–120 seconds after contrast medium administration, the bladder mass demonstrates varying degrees of postcontrast enhancement. Subsequently, on the delayed phase of CT urography, the tumoral mass exhibits washout and may be identified as a soft tissue filling defect within the contrast material-filled hyperattenuating lumen (27). CT urography could be incorporated in the follow-up regime as it demonstrates it demonstrates high accuracy around 91.7% for detecting recurrent bladder tumors in the nephrographic phase compared to 83.2% in the excretory phase (34).

Virtual cystoscopy (VC) is a novel technique used for detecting and assessing bladder cancer. It involves creating virtual reality images of the bladder through 3D volume rendering methods. Two main techniques are used: noncontrast CT with air/CO2 filling or delayed phase MDCT urography. VC allows for 360-degree navigation of the interior of the bladder to evaluate its wall and lumen aiding in the detection of abnormal lesions. While VC has advantages like being minimally invasive and providing precise tumor measurements, it cannot detect mucosal color changes or obtain tissue biopsies. Despite its limitations, VC can be valuable when conventional cystoscopy is not possible or contraindicated (27).

Et al. Elawady. (35) conducted a study to compare virtual cystoscopy to conventional cystoscopy. Over three years, 30 patients with NMIBC underwent CT virtual cystoscopy. It detected 20 lesions in 18 patients, while conventional cystoscopy found 23 lesions in 19 patients. Virtual cystoscopy showed 87% sensitivity, 100% specificity, and 100% positive predictive value, but only 78.5% negative predictive value. It's promising for lesion detection but not yet a replacement for conventional cystoscopy (35).

E. BLOOD MARKERS

Several blood markers have emerged as potential indicators of urothelial carcinoma. Paner et al. (36) demonstrated the utility of GATA3, S-100P, CK7, CK20, HMCK, and p63 in supporting the urothelial lineage across different variants of urothelial carcinoma. Fang et al. (37) shed light on the presence of cancer stem cells in urothelial carcinoma, identifying a diverse panel of markers relevant for simulation studies and treatment strategies. Soria et al. (38) uncovered the potential of p53 overexpression as a marker for tumor invasion in urothelial bladder carcinoma. Circulating tumor cells (CTCs) test commercially available and FDA-approved as The CellSearch CTC Test™ (39), detects CTCs in peripheral blood which indicate early metastatic progression and correlate with shorter time to disease recurrence and Cancer specific survival (CSS) post-radical cystectomy (38). CellSearch Technology shows around 50% positive detection for metastatic BC and approximately 15% for clinically localized BC. Recurrent tumor detection is 20-44% in progressing patients. Meta-analysis reveals CTC presence correlates with tumor stage, histological grade, metastasis, and regional lymph node involvement. CTC detection assays exhibit 35% sensitivity and 97% specificity. (40) Together, these findings indicate a promising role for blood markers in the diagnosis, prognosis, and treatment of urothelial carcinoma.

F. URINARY MARKERS

Urinary markers have gained massive attention due to the low cost and ease of collection, with no related disadvantages or discomfort experienced with cystoscopy (41).

Despite the countless available urinary biomarkers, only a few tests have been FDA approved for diagnosing and surveilling bladder cancer (42).

Comparison table of some FDA- approved urinary markers

Table 2

Test	Mechanism	Sensitivity (%)	Specificity (%)	PPV	NPV	Actual Role in Clinical Guidelines
BTA Stat (POC)	Detect human complement factor H-related protein	40–72	29–96	40–88	38–76.9	-
BTA TRAK	Detect human complement factor H-related protein	50–62	68–87	45.4	88.4	-
NMP22 BladderChek (POC)	Detect NMP22 protein	11–85.7	77–100	18.2–100	61.9–93.9	-
NMP22 Bladder Cancer Test	Detect NMP22 protein	24–81	49–100	31–100	60–91	-
UroVysion	FISH chromosome abnormalities	13–100	63–100	21–83	67.9–100	Might serve as a reflex test following unremarkable cystoscopy findings and inconclusive or ambiguous cytology results
ImmunoCyt	Immunofluorescence antigen detection	50–85	62–86	26–72	81–93	Might serve as a reflex test following unremarkable cystoscopy findings and inconclusive or ambiguous cytology results

Test	Mechanism	Sensitivity (%)	Specificity (%)	PPV	NPV	Actual Role in Clinical Guidelines
CxBladder	mRNA biomarkers (IGF, HOXA, MDK, CDC, IL8R)	90	91	-	-	-
Uromonitor	Detection of specific genetic mutations in TERT, FGFR3, and KRAS	93.1	85.4	79.4	95.3	-

Abbreviations: POC, point of care; PPV, positive predictive value; NPV, negative predictive value.(43,44)

Et al. sciarra. (45) compared sensitivity in low grade tumors UroVysion and immunoCyt to cytology concluding the superiority of the two urinary biomarkers. Combining urinary biomarkers with cytology enhanced sensitivity in but still missing 10% of lower grade tumors and are susceptible to producing false-positives results according to a study done by et al. Chou. (46).

It is imperative to discuss these 3 promising biomarkers tests, namely: Bladder Epicheck, CxBladder, Uromonitor, as they show superior sensitivity and specificity in comparison to other markers.

F.1. Bladder Epicheck: The Bladder EpiCheck test, widely employed, relies on analyzing the methylation profile of urothelial cells to identify bladder neoplasms (47).

In a research study carried out by et al. Witjes (48) that involved 440 patients 353 individuals were considered for analysis. The Bladder EpiCheck (BE) test displayed a sensitivity of 68.2% and a specificity of 88.0%. When excluding grade (LG) Ta recurrences the sensitivity rose to 91.7% with a negative predictive value (NPV) of 99.3%. The ROC curve areas, both including and excluding LG Ta lesions were measured at 0.82 and 0.94 respectively.

In general, the BE test exhibited a NPV of 95.1% which increased to an impressive 99.3% when LG Ta recurrences were not included alongside maintaining a specificity of 88.0%. These results indicate the potential of incorporating this test to follow-up NMIBC and lessen the need for repeat cystoscopies and cytology tests.

F.2. CxBladder: The Cxbladder Monitor (CxM) is a urine test that utilizes mRNA and is created for home use to identify NMIBC. It evaluates the quantities of five mRNA biomarkers associated with bladder cancer (IGF, HOXA, MDK, CDC, IL8R) well as two clinical factors, whether the previous tumor was initial or recurrent and the duration since the last tumor removal (49).

A research study done by et al. Kavalieris (50) involving 763 patients the Cxbladder Monitor test showed a sensitivity of 93% and an impressive negative predictive value of 97% in detecting recurrent urothelial carcinoma. It displayed a sensitivity rate of 95% for identifying high risk disease and 86% for low grade Ta disease. The analysis of subgroups revealed consistent accuracy levels across various factors with false negative results observed in less than 1.5% of cases. The findings indicate that the test could be used as an additional tool to complement or replace cystoscopy by potentially reducing or postponing some of them.

F.3. Uromonitor: Uromonitor V2® is a test that uses urine to identify specific genetic mutations in three genes (TERT, FGFR3 and KRAS), for evaluating the likelihood of recurrence. Among the 97 patients who underwent testing with Uromonitor V2® 37.1% received results successfully detecting 27 out of 29 cases where the disease recurred. The presence of confirmed recurrences in examinations matched with positive outcomes from Uromonitor V2® tests. However, there were instances of positives in patients who did not have a history of bladder cancer or any signs of recurrence. The test showed a sensitivity rate of 93.1% specificity rate of 86.8% positive predictive value (PPV) of 75.0% and negative predictive value (NPV) of 96.7%. In patients with a history of non-muscle invasive bladder cancer (NMIBC) the sensitivity was found to be at 93.1% specificity, at 85.4% PPV at 79.4% and NPV at 95.3% (51).

G. MRI + URINE CYTOLOGY

Et al. Lee. (41) performed a study aimed to assess noninvasive methods for monitoring NMIBC. Data from cases of transurethral resection of bladder tumor (TURBT) were analyzed. The combination of urine cytology and bladder MRI exhibited impressive sensitivity 92% but lower specificity 43%. Instances of false-negative results stemmed from missed radiological interpretations, an empty bladder during MRI, or gross hematuria. Encouragingly, none of the false-negative cases displayed rapid progression necessitating surgery. While the combined MRI and cytology approach demonstrated sensitivity akin to cystoscopy, specificity was lacking. Nevertheless, it provides patients with alternative follow-up options and potential reduction in cystoscopy reliance. Moreover, it holds promise in minimizing additional imaging tests for kidney, ureter, and peri-vesical lesions (41).

H. FUTURE

-MicroRNAs: Urinary miRNAs have been considered as markers for diagnosis, but there are variations in findings across different research studies. A comprehensive analysis showed that urinary miRNAs have a sensitivity of 83% and specificity of 81% in detecting bladder cancer. Additional investigations suggest that using multiple miRNAs together provides more accurate diagnostic results compared to using a single miRNA which resulted in sensitivity of 0.69% and specificity of 0.74%. However extensive research studies are required to validate their diagnostic effectiveness particularly for identifying low grade bladder cancer cases (52).

-Exosomes: New types of agents, like exosomes and micro vesicles are currently being investigated for identifying cancer in cases of bladder tumors. These structures, with their protective lipid bilayer play a role in shielding tumor components from breaking down and supporting the advancement of tumors. Exosomes that are specifically released by bladder tumors could potentially be more abundant in muscle invasive bladder cancer (NMIBC) making them promising indicators for detecting NMIBC using ELISA tests. Exosomes have an impact on the progression of tumors influencing factors such as the development of new blood vessels, invasion and spread to other parts of the body. Some specific miRNAs found in exosomes in urine are associated with bladder cancer show promise as potential indicators. Research has shown that examining CD9 levels on exosomes in urine and blood samples has demonstrated specificity (100% in blood samples 83.3% in urine) and sensitivity (82.4% in blood

samples 92.6% in urine). Additional studies are required to determine how tumor exosomes and micro vesicles are, at detecting NMIBC (44).

-SERS: discovered in 1928 and modified and enhanced in 1974 a molecular vibration spectra technique that gained interest in the oncological and biomedical field recently with its utilization in liquid biopsie (53) a Serum surface-enhanced Raman's study done by et al. Li. (54) employs genetic algorithms (GAs) combined with linear discriminant analysis (LDA) to characterize and classify serum surface enhanced Raman spectroscopy (SERS) spectra between bladder cancer patients and normal volunteers. Serum SERS spectra from healthy volunteers (n = 36) and bladder cancer patients (n = 55) are analyzed. By examining the Raman bands associated with proteins nucleic acids and lipids the diagnostic models show enhanced sensitivity (90.9%). Specificity (100%) in distinguishing bladder cancer patients from those with normal serum SERS spectra compared to other methods like principal component analysis. The effectiveness of the algorithm is further confirmed by ROC curves based on the GA LDA technique indicating potential, for using serum SERS as a non-invasive method to detect bladder cancer through blood samples (54).

-Combining MiRna and SERS: Et al. Moisoiu. (55) collected samples from 15 BC patients and 16 control subjects, using next-generation sequencing for miRNA, and performed SERS profiling. Differentially expressed miRNAs were validated in a larger retrospective cohort of 116 individuals via RT-qPCR.Machine learning algorithms were applied to assess diagnostic accuracy, with input from miRNA, SERS, or both. The combined use of SERS data with three miRNAs (miR-34a-5p, miR-2053p, miR-210-3p) achieved an Area Under the Curve of 0.92, outperforming individual miRNA profiling and individual SERS data For distinguishing between luminal and basal types of BC, the combination averaged an AUC of 0.95, surpasses using both individually (55).

The study concluded that combining miRNA and SERS profiling in urine can accurately diagnose and molecularly stratify BC, potentially serving as an effective point-of-care tool (55).

IV. DISCUSSION

1. Key Findings

-Imaging: Ultrasound, MRI and CT scans play a role in the detection and staging of bladder cancer. Contrast enhanced ultrasound (CEUS) and MRI are known for their accuracy in providing information on tumor characteristics and response to treatment.

-Urine and blood Markers: Various markers found in blood and urine such as BTA Stat, NMP22 BladderChek and ImmunoCyt hold promise in diagnosing bladder cancer. When used alongside cytology these markers improve sensitivity levels. However, challenges exist in achieving specificity with low grade tumors.

-Novel Biomarker Tests: New biomarker tests like Bladder EpiCheck, Cxbladder and Uromonitor show effectiveness in detecting bladder cancer with sensitivity and specificity. These tests offer alternatives to traditional cystoscopy procedures potentially reducing the need for repetitive invasive interventions.

-Combining tools: The combination of MRI with urine cytology shows potential in monitoring cases of muscle invasive bladder cancer. This integrated approach offers methods for follow up care while possibly reducing the reliance on cystoscopy. Nonetheless maintaining specificity poses a challenge that calls for refinement of strategies.

- Future Trends: Urinary exosomes are emerging as promising tools for diagnosing bladder cancer. MicroRNAs and SERS have shown both sensitivity and specificity in this regard. exosomes released by bladder tumors could serve as markers for detecting the disease through ELISA tests.

2. Clinical implications and practical recommendations

As for currently, cystoscopy remains the “gold standard”. However, its invasive nature leads to many complications including discomfort, embarrassment and even infections. Integrating non-invasive

diagnostic methods is crucial for effectively monitoring superficial urothelial carcinoma. Urinary biomarkers tests and advanced imaging techniques may help identify recurrences early on. By reducing the frequency of cystoscopies, patients experience less discomfort, anxiety and costs making it easier to stick to the follow-up plan. Tailoring follow-up approaches according to individual risk factors ensures that surveillance intervals are optimized, and any disease progression is detected timely. It is also important to educate healthcare providers on how to interpret and incorporate non-invasive diagnostic findings into their clinical decision-making processes. Ongoing research and validation studies are essential to continuously assess the accuracy and effectiveness of these tools in real world scenarios.

In short words, the adoption of non-invasive diagnostic methods provides clinical advantages such as early detection, decreased invasiveness and personalized monitoring of superficial urothelial carcinoma.

3. Drawbacks

Heterogeneity of the results, many reviewed articles had varying and small sample sizes, different conditions of the subjects, some of them lacked control groups. Only two of the reviewed authors considered ethnicity as a factor in their research (52) (48).

V. Conclusion

The included literature discusses the evolving landscape of tools and methods used for detecting and following-up non-muscle-invasive bladder cancer. The combination of tests, like Bladder EpiCheck and Cxbladder with imaging methods and urine cytology shows potential for improving accuracy in follow-up and patient care. Despite progress, there are obstacles to achieving sensitivity and specificity in detecting low grade tumors and non-invasive diagnostic tools are still not capable enough to replace cystoscopy. Ongoing research is crucial to tune methods and improve outcomes for patients with bladder cancer.

Contribution to the field:

My literature review offers a summarization of the latest literature and meta-analysis concerning numerous diagnostic measures and discusses their implications.

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