Urinary Cytology: Ancillary Diagnostic Testing
Ancillary Diagnostic Testing

• Why?

• When?

• How?
Urothelial Carcinoma

false neg. in -30%
invasive
long-term surveillance:
expensive

&

cheap
high spec. for high-grade UC
non-visible high-grade UC
equivocal urinary cytology
low sens. for low-grade UC
Urinary Tumor Markers

• DNA-based
  – UroVysion® FISH Test
  – Microsatellite/LOH detection

• RNA- and protein-based
  – Cxbladder (uRNA-2)
  – uCyt+/ ImmunoCyt
  – BTA stat/ TRAK
  – NMP22

• Epigene-based
  – DNA methylathion

Add significant benefit?
Cost-effectiveness?
Standardization?
Validation?

Sapre N et al. Urol Oncol 2014
Urinary Tumor Markers

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Add significant benefit? Cost-effectiveness? Standardization? Validation?

Sapre N et al. Urol Oncol 2014
nuclear / cytologic atypia

mild

moderate

severe (G3)
Nuclear / cytologic atypia

Probability of high grade UC

- Low
- Moderate/high
- Certain
Nuclear/cytologic atypia

Probability of high grade UC
High grade UC
Spec./PPV >90%
Malignant
Diagn. work-up
Treatment
Atypia (8-31%) → Risk of malignancy 21-71%

negative or equivocal cystoscopy → ?

Ubago JM et al. Cancer Cytopathol. 2013
High grade UC

Atypia (8-31%)

Risk of malignancy 21-71%

UroVysion as ancillary test upon request

Negative
Follow-up

Positive
Follow-up/diagn. work-up
Treatment

Risk of malignancy 21-71%
UroVysion FISH Test

• High-grade UC
  – no additional benefit compared to cytology
    (sens. & spec. 70-100%)

• Low-grade UC
  – can increase sens. of cytology from 25% to 60-75%
  – usually visible by cystoscopy

• Ancillary test for clarification of atypical urothelial cells
  – detection of non-visible high-grade UC
  – high-grade UC with AUC: 48/52 (92%) FISH +

Dimashkieh H et al. Cancer Cytopathol. 2013
FISH positive result:

- ≥4/25 cells with gains in ≥2 chr.
- ≥12/25 cells with homozyg. del 9p21

Sokolova IA et al. J Mol Diagn 2000
UroVysion FISH Test

- Sensitivity: range 8-100%
- Specificity: range 29-100%

→ lack of standardization

*Dimashkieh H et al. Cancer Cytopathol 2013*
UroVysion FISH Test

Performance depends on:

• UC: Low-grade – high-grade

• Urinary specimen (cellularity)

• Technical procedures
  – FISH on PAP stained slide – on remaining cellular material

• Test evaluation
  – automated relocation
  – definition of a positive result
  – pitfalls (false positivity): Tetraploidy, radiation
1:1 50% ethanol

coated glass slide
UroVysion FISH on cytological specimens

Automated cross table with relocation
Chromosomal aberrations in benign urinary cytology: False positive FISH

- Reactive umbrella cells
- Polyploidy

- Tetraploidy common in benign urothelial cells
- Radiation induced chromosomal aberrations
- No 9p21 deletions

Tapia C et al. Cancer Cytopathol 2011
Wojcik EM et al. Annal Quant Cytol Histol 2000
FISH positive result:
≥4/25 cells with gains in ≥2 chr. or
≥12/25 cells with
homozyg. or heteroz. del 9p21

Tetraploidy >10/25 cells

Savic S et al. Int J Cancer 2009
Zellweger T et al. Int J Cancer 2006
80y, renal pelvic washing
History of pTaG2 (low grade) bladder UC
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History of pTaG2 (low grade) bladder UC

FISH pos
80y, renal pelvic washing
History of pTaG2 (low grade) bladder UC

\[ \rightarrow \text{pTa G2 (low grade), 5mm} \]
70y, renal pelvic washing
Renal pelvis with suspicious lesion
AUC-US

Negative cytology
pT3, G2 urothelial carcinoma
82y, bladder washing
History of pT1G2 bladder UC, post-BCG
82y, bladder washing
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→ pT1 G2
Why?

• to clarify atypical urothelial cells
  – non-visible high-grade UC
  – avoid unnecessary invasive procedures

→ Communication with urologist
When?

- Atypical urothelial cells:
  - after intravesical therapy for high-grade UC (BCG)
  - in upper urinary tract cytology
    - Cytology with low sensitivity
    - 60% invasive (urinary bladder: 15%)

- Not in “benign” and “malignant” cytology
  - Predicting recurrence of UC with negative cytology and cystoscopy (anticipatory positive)
    → no impact on clinical management

UroVysion FISH Test

How?

• On diagnostic PAP-stained slides
• With automated relocation of target cells
• By cytomorphologists trained in FISH analysis
Thanks!

Lukas Bubendorf
Audrey Barascud
Bruno Grilli
Michelle Herzog
Cytopathology team