National Cancer Institute

Digital cytology- Applications in cervical cancer screening

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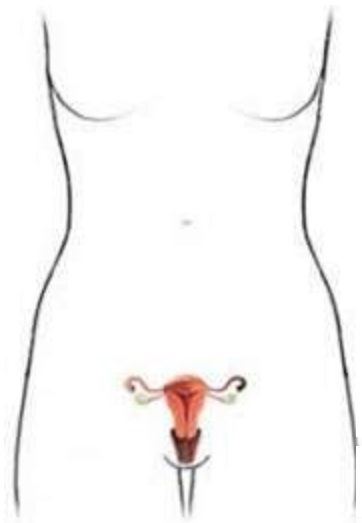
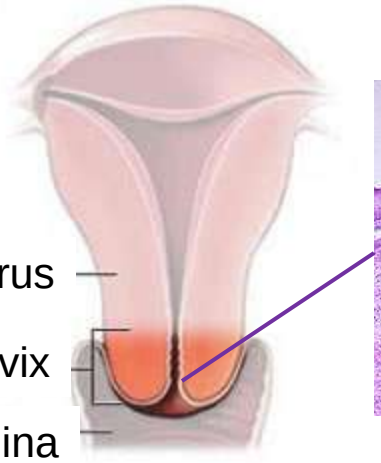
U.S. DEPARTMENT OF HEALTH AND HUMAN SERVICES
National Institutes of Health

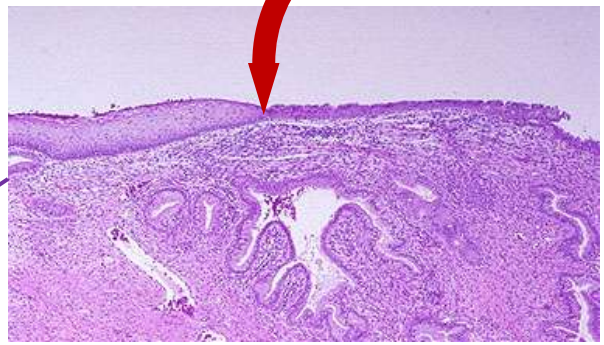
Statements

- I have no conflict of interest
- These are personal opinions and not official NCI statements

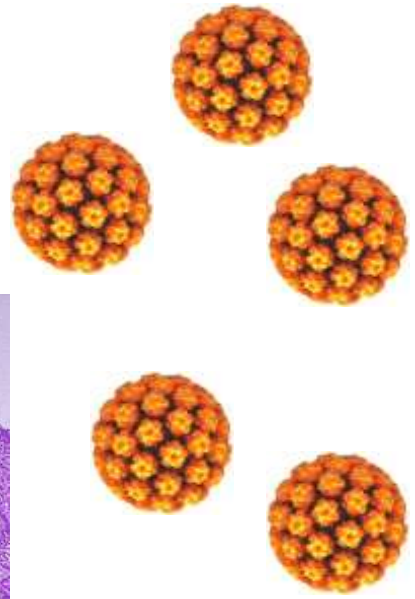
Cervical cancer







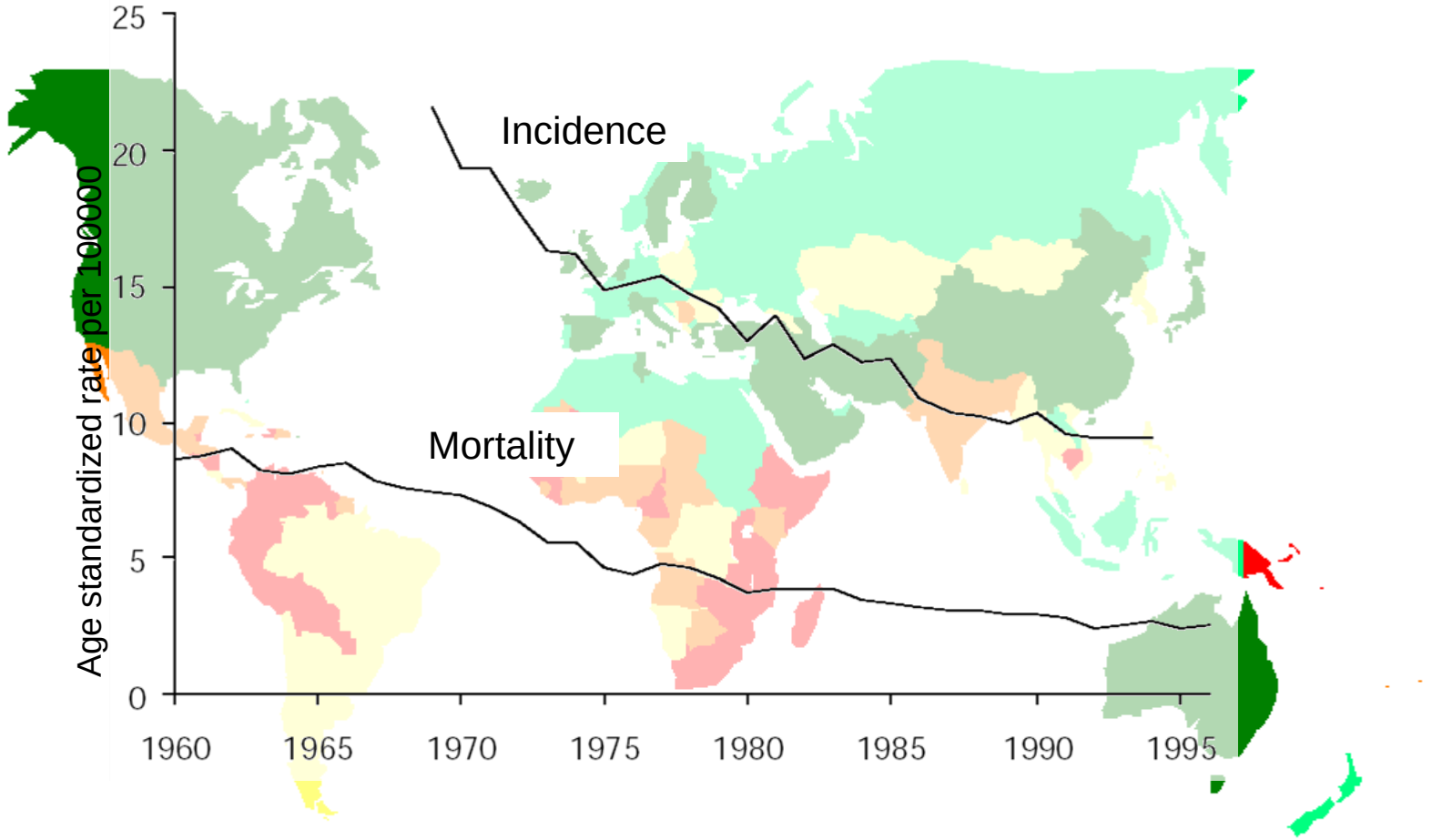
Transformation zone



Carcinogenic HPV

- Second most frequent cancer in women worldwide
- Caused by persistent infections with carcinogenic human papillomaviruses (most importantly 16,18)

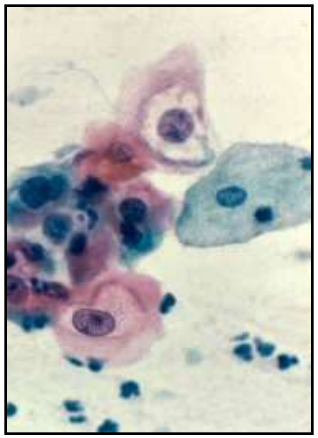
Cervical cancer incidence and Pap based screening



Reduction of cervical cancer incidence after introduction of screening (Canada)

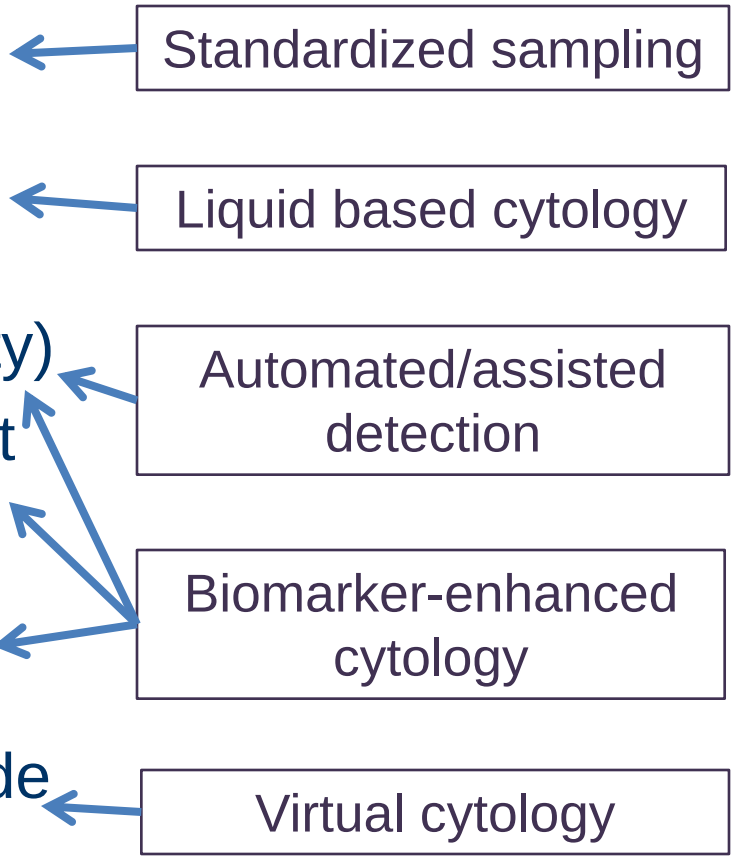
Characteristics of the Pap smear

- Introduced by Papanicolaou in the 1940s
- A smear is taken from the cervix and transferred to a glass slide, stained with several stains (nuclear, cytologic)
- Abnormal/precancerous cells are detected by morphology
- Presence of abnormal cells triggers further management and treatment



Limitations of current cytological screening

- Problems of (1) sampling and processing, (2) interpretation
- Heterogeneity of sample (mucus, blood, varying cellularity)
- Rare event detection (low sensitivity)
- Many unspecific changes (transient HPV infections)
- Intra- and interobserver variation (subjectivity, experience)
- Expert review, sample logistics, slide storage

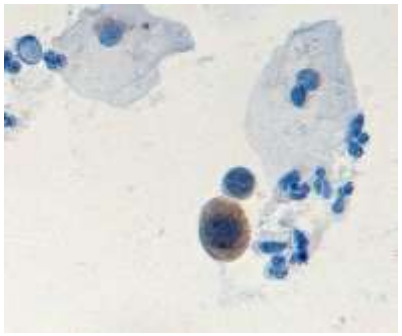


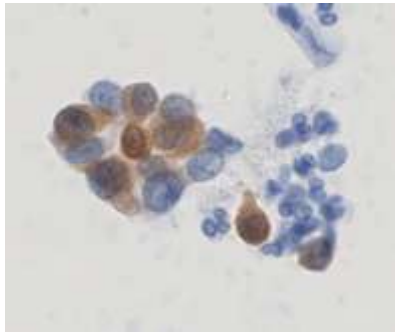
Large epidemiologic/interventional studies are necessary to demonstrate the efficiency of these approaches

Cytiga- a virtual cytology system

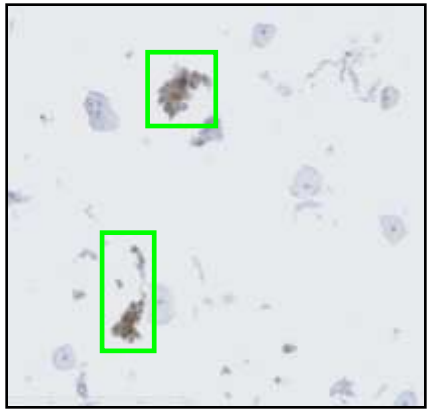
Virtual cytology

Biomarker enhanced cytology



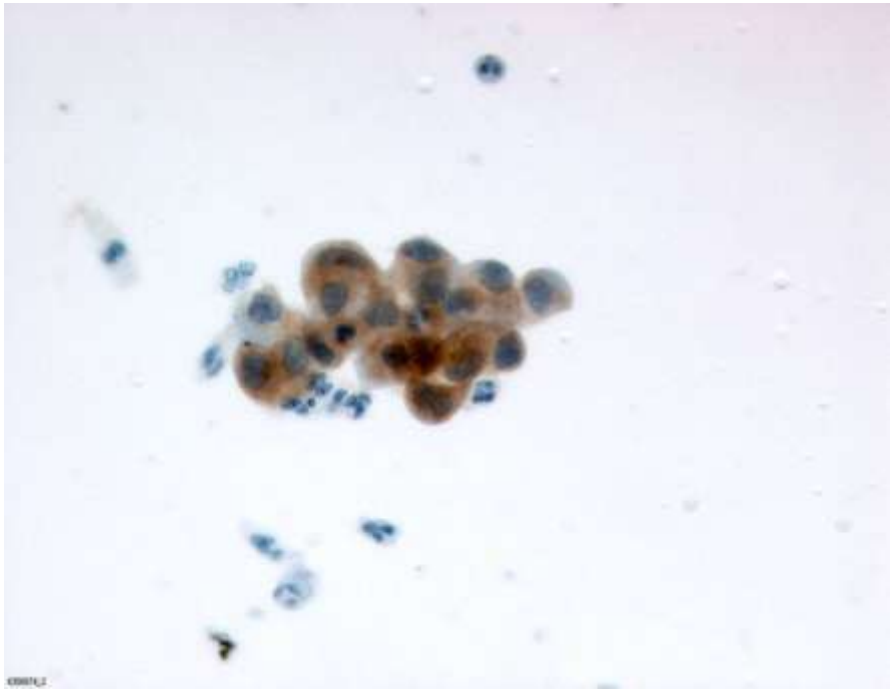


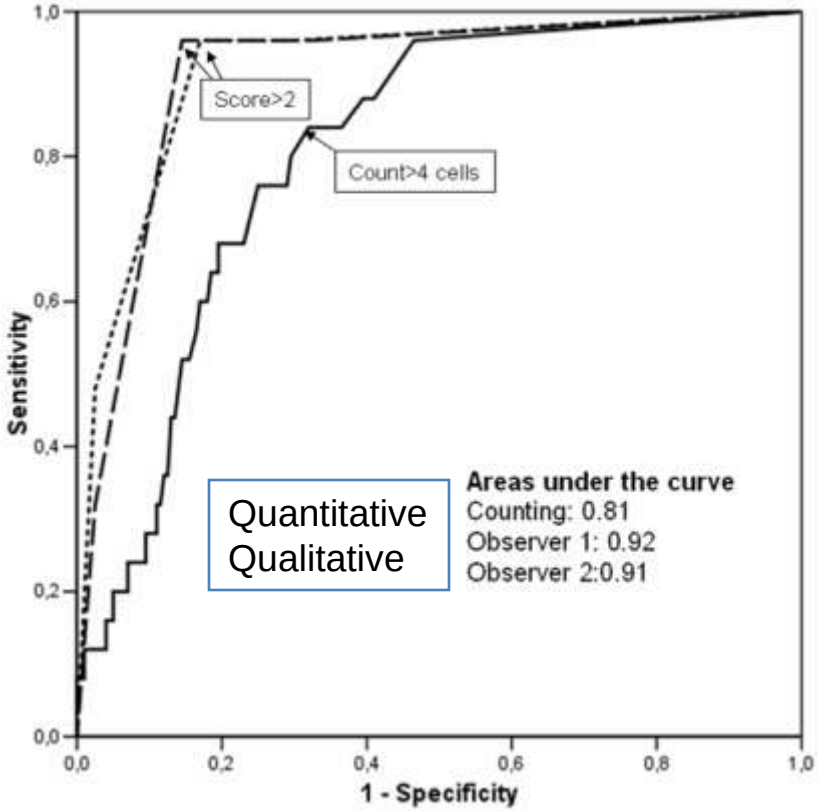
Automated/assisted detection



The biomarker example: p16 ICC

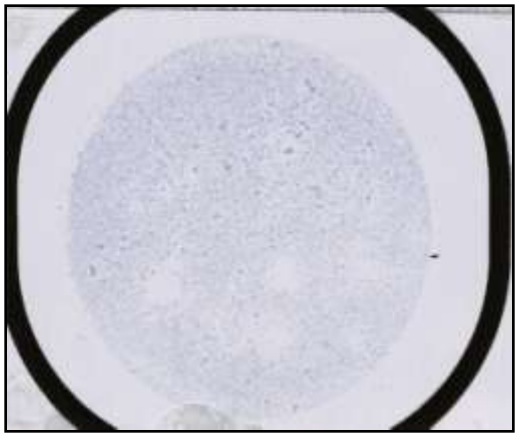
- Direct link to HPV carcinogenesis: high sensitivity and specificity for detecting cervical precancers
- Improved rare event detection (locator function)
- Qualitative (nuclear morphology) and quantitative (cell counting) evaluation possible

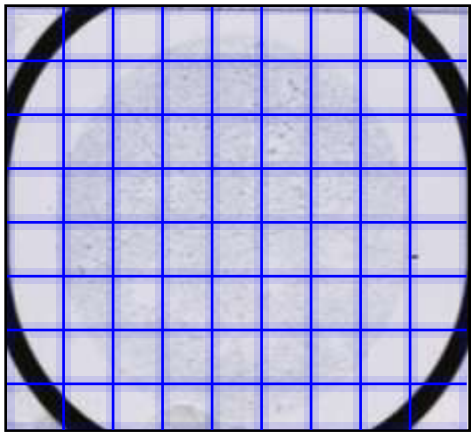


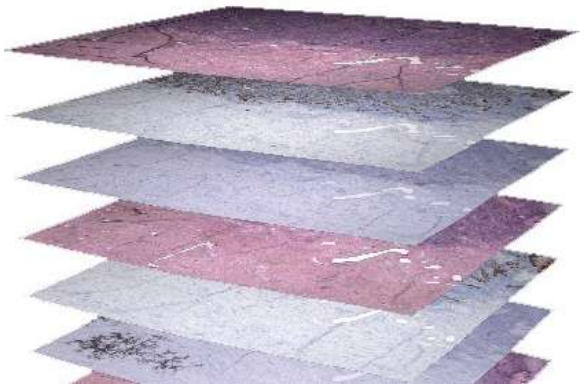


Virtual cytology

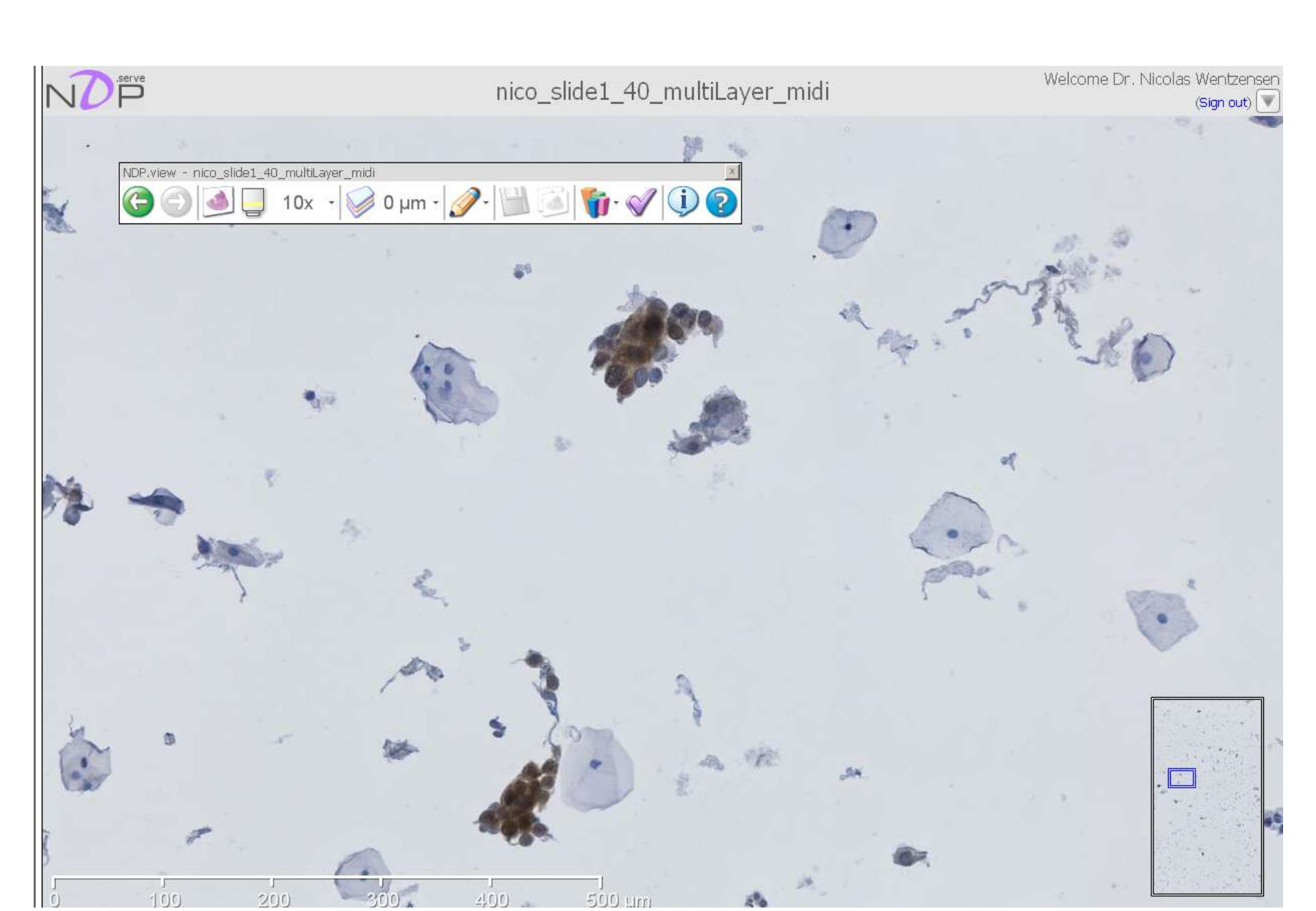
- Slide is scanned, image is stored in tiles
- Image can be accessed via internet through a web browser plugin, only the active area is transmitted
- Z-stack option: Multiple images are superimposed to analyze 3D cells







Using a virtual slide


nico_slide1_40_multiLayer_midi
Welcome Dr. Nicolas Wentzensen
(Sign out)
NDP.view - nico_slide1_40_multiLayer_midi
10x
0 µm
0
100
200
300
400
500 µm

NDP.serve - nico_slide1_40_multiLayer_midi - Microsoft Internet Explorer
File Edit View Favorites Tools Help
Back Search Favorites
Address http://tiga.bioquant.uni-heidelberg.de/NDPServe.dll?ViewItem?ItemID=108
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NDP.serve
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Welcome Dr. Nicolas Wentzensen
(Sign out)
NDP.view - nico_slide1_40_multiLayer_midi
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Internet

NDP.serve - nico_slide1_40_multiLayer_midi - Microsoft Internet Explorer
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Welcome Dr. Nicolas Wentzensen
(Sign out)
NDP.view - nico_slide1_40_multiLayer_midi
40x
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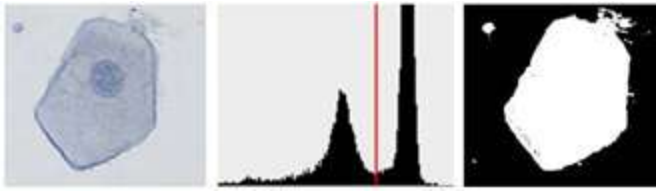
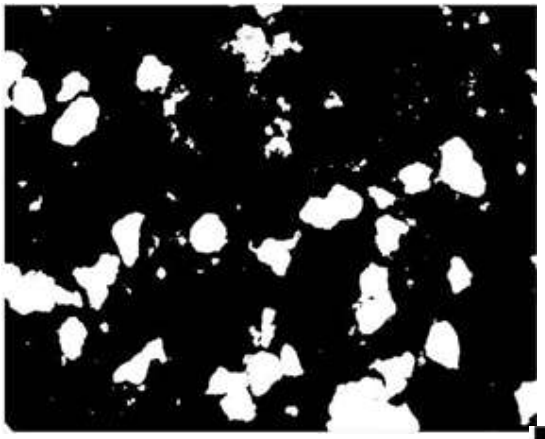
25
50
75
100 µm
Internet

Cell detection

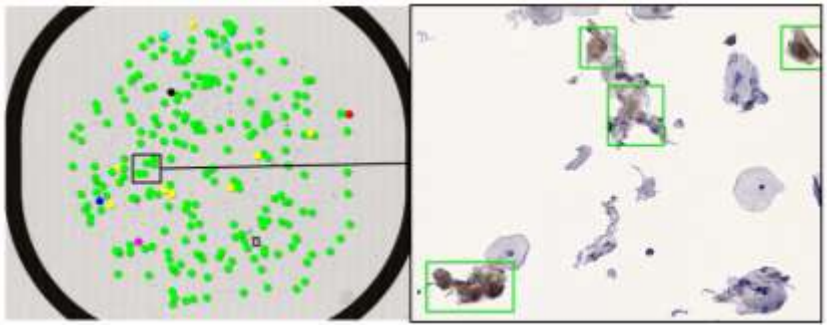


Set color-based cutoff between cells and background



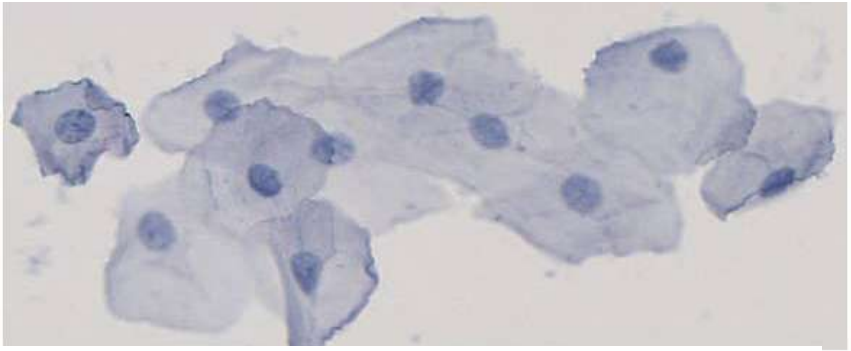
Apply cutoff on slide

Small object removal

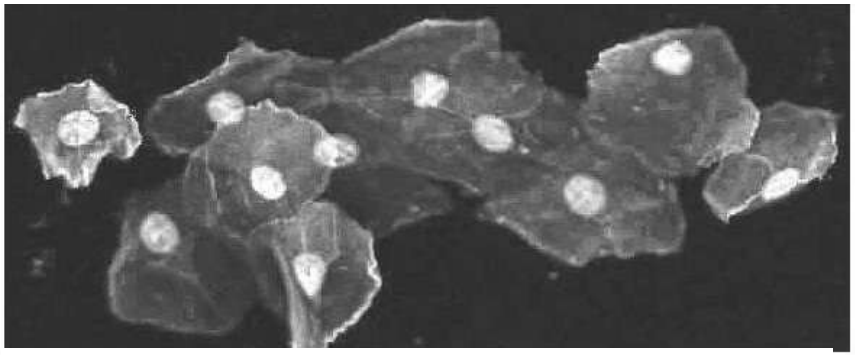


Overlay annotations on original image

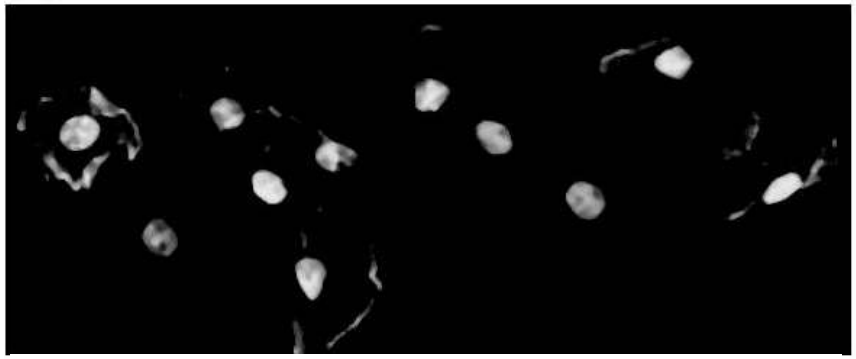
Nucleus detection



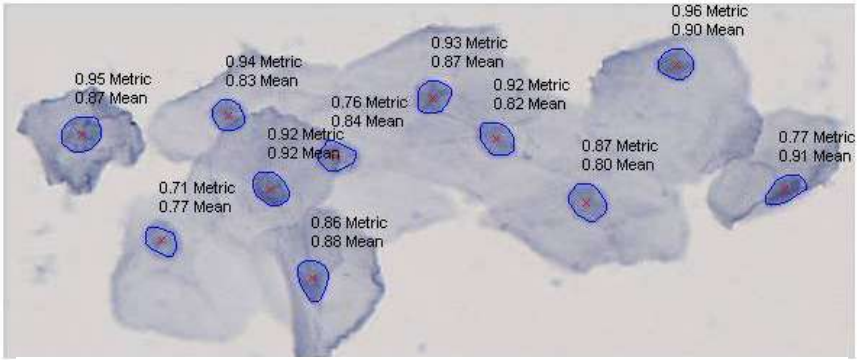
Original image



Probability function for nuclear color

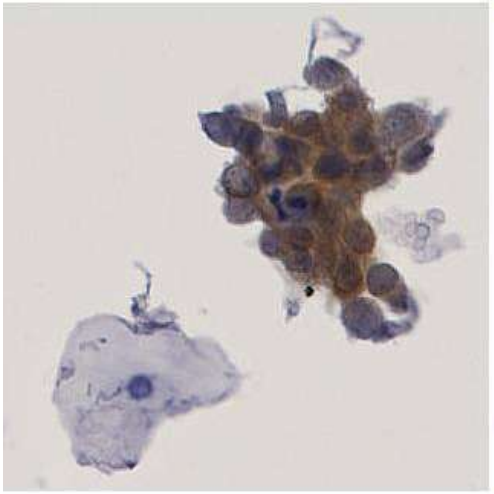


Subtraction of cytoplasm

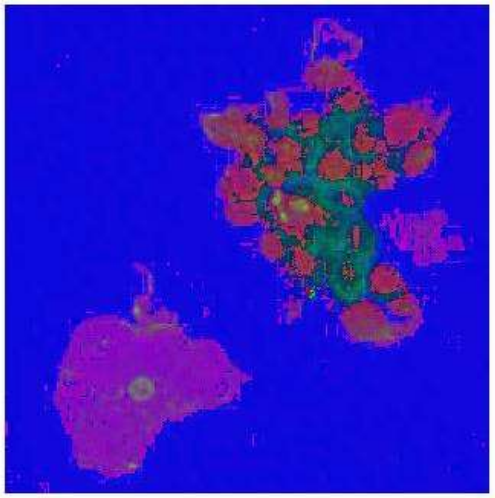


Nuclear annotation on original image
(probability and roundness)

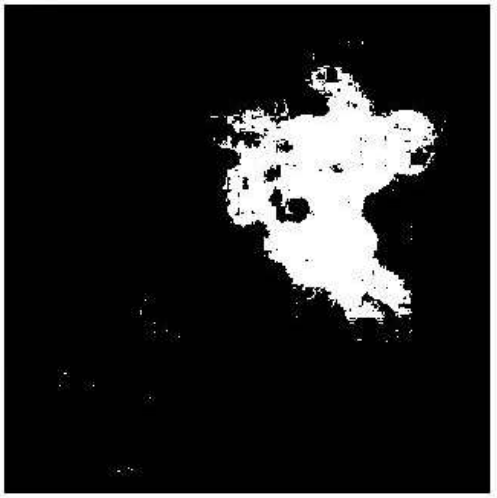
Biomarker detection



Original image



HSV color space



Applying threshold for HSV color of p16 stain

Detection of p16 - stained cells

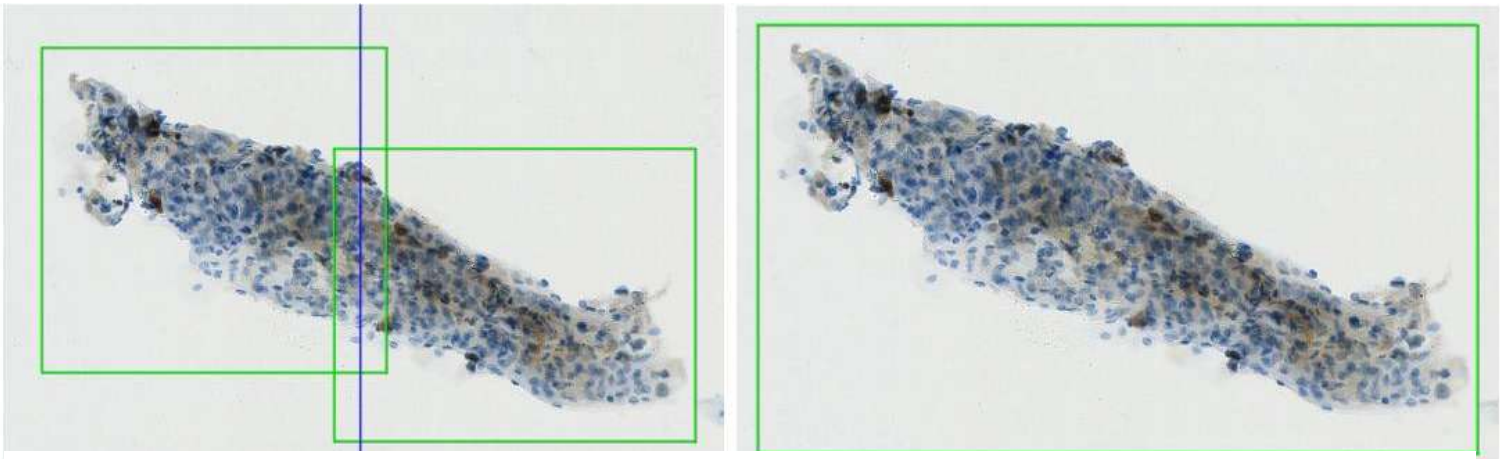
		Man		
		positive	negative	total
Machine	positive	98	22	120
	negative	11	~50,000	50,011
	total	109	50,022	50,131

McNemar's test: Not significant

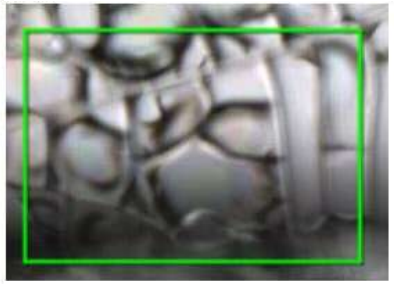
Man:	109/125 (87%)
Machine:	120/125 (96%)

- High agreement between automated and manual detection of p16 positive cells
- Few discrepancies related to missed cells in manual evaluation and faint staining / artifacts in automated evaluation

Challenges and artifacts



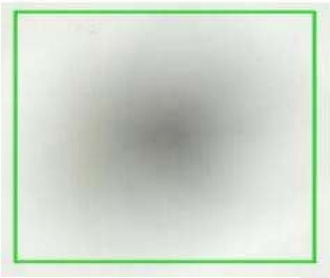
large object spanning multiple tiles



air

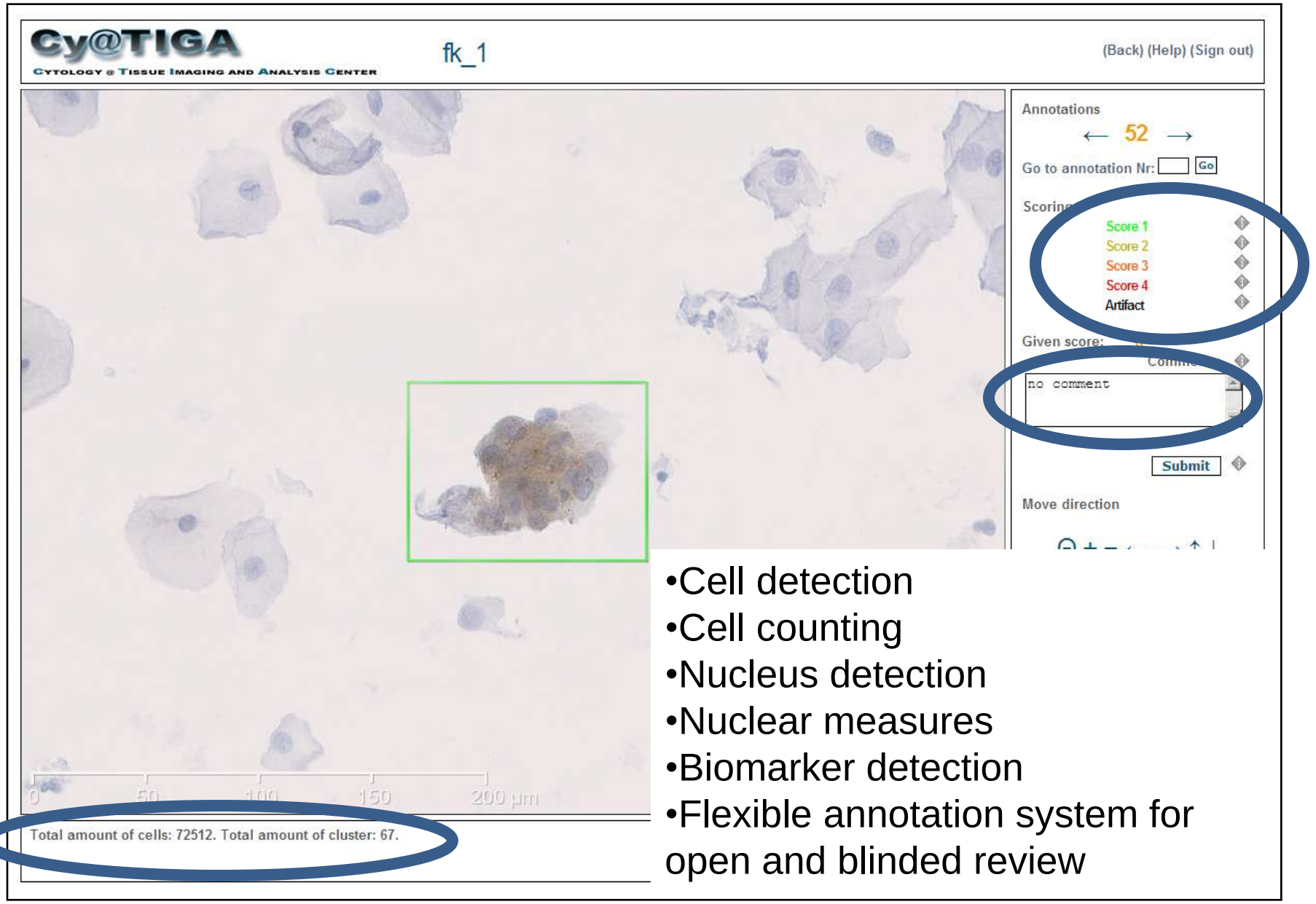
unfocused cell

dirt/precipitate



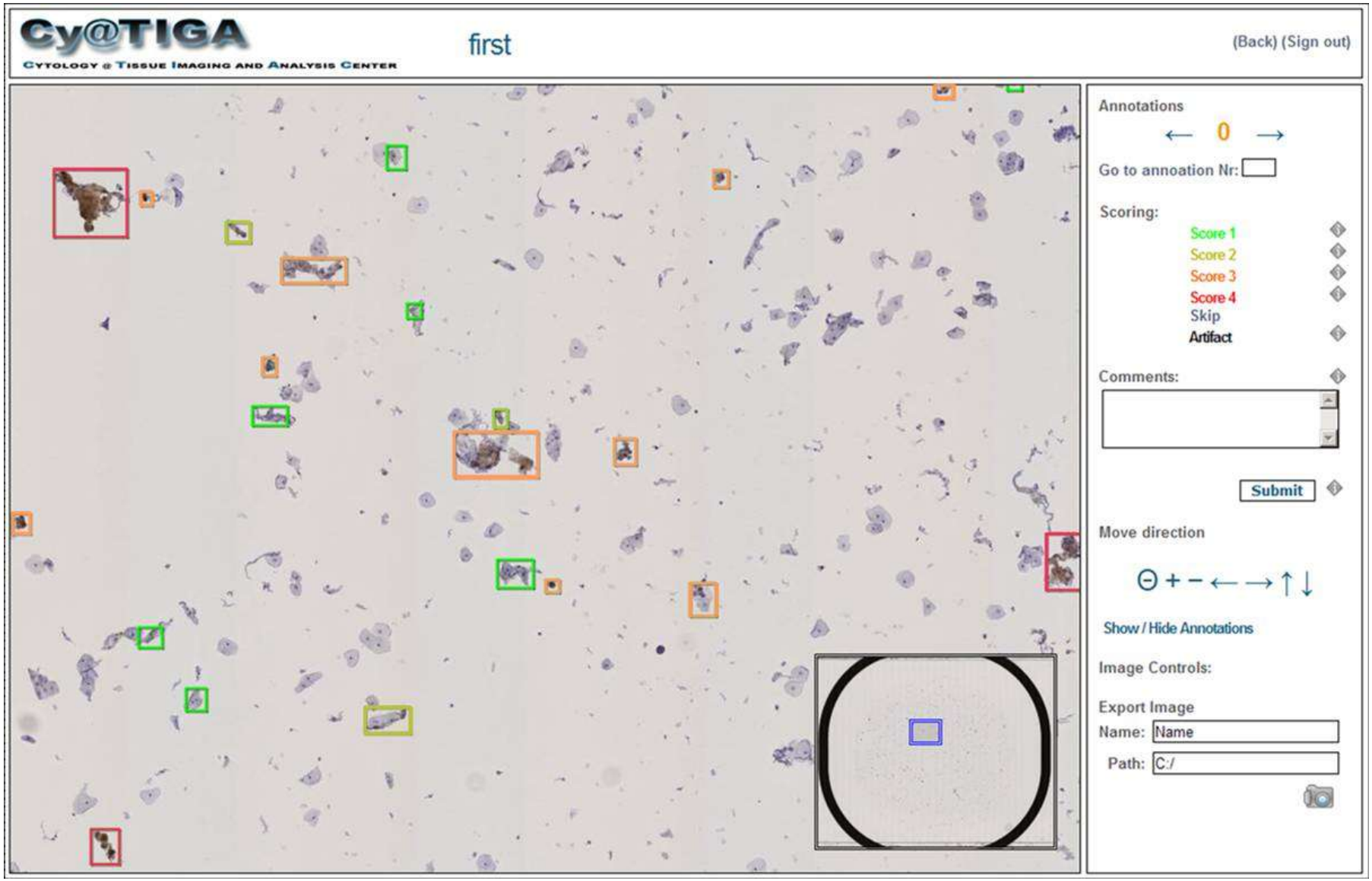
dust

Cell annotation



- Cell detection
- Cell counting
- Nucleus detection
- Nuclear measures
- Biomarker detection
- Flexible annotation system for open and blinded review

Database for slide evaluation



Next steps

- Fine-tuning of biomarker detection
- Expand automated evaluation
- Analyze large series of p16 stained slides, e.g. from NCI cervical cancer screening studies
- Challenges for cytology:
  - New QC regulations (case loads)
  - HPV vaccination against 16, 18
  - Primary HPV testing
- Many opportunities for biomarker enhanced virtual cytology
- **Goal: Create a system for biomarker research and for routine cytology use**

Team

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