

РОЛЬ P16 В ПОСТАНОВКЕ ДИАГНОЗА

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кандидат медицинских наук
врач-патологоанатом

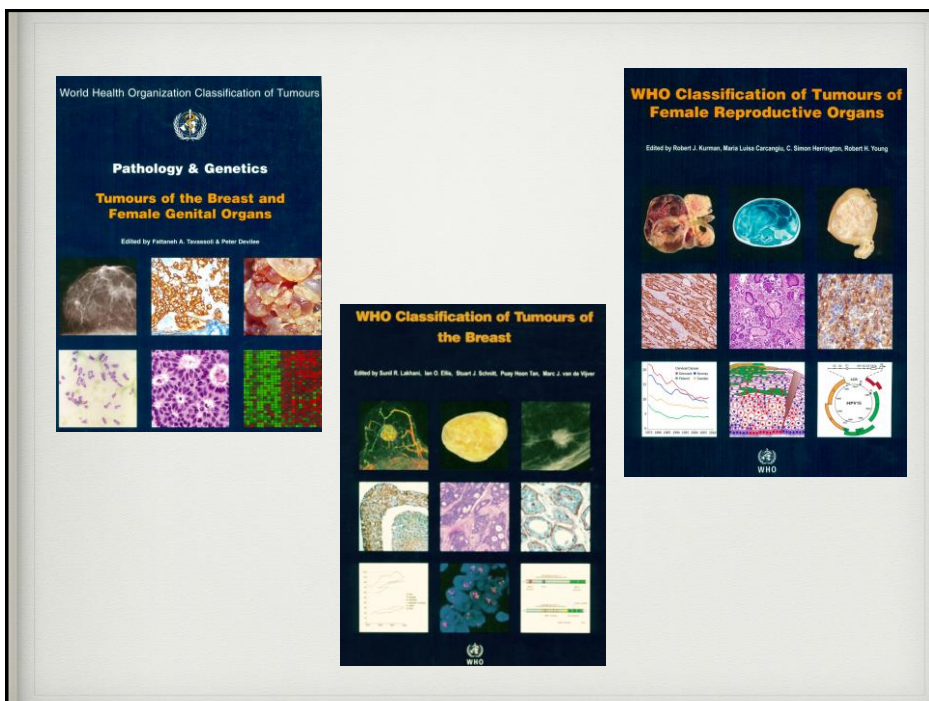
International fellow of United States & Canadian Academy of Pathology (USCAP)

ЕВРОПЕЙСКИЙ МЕДИЦИНСКИЙ ЦЕНТР

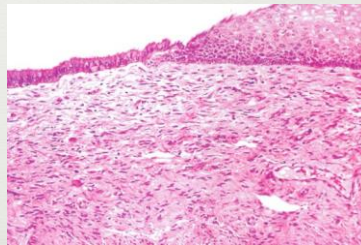
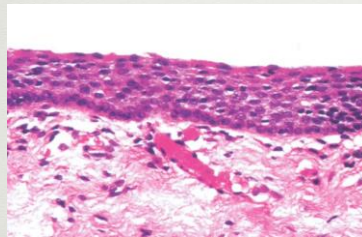
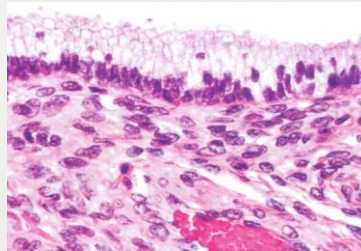
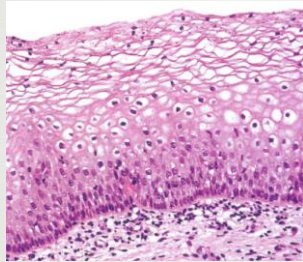
г. Москва

- Современная классификация
- Методы морфологической диагностики
- Молекулярно-биологические аспекты канцерогенеза, прогностические факторы

- Цитологическая диагностика
- Патоморфологическая диагностика (гистология, иммуногистохимия)
- Молекулярно-биологическая диагностика (CISH; FISH)



Гистологическое строение шейки матки



WHO Classification of tumours of the uterine cervix^{a,b}

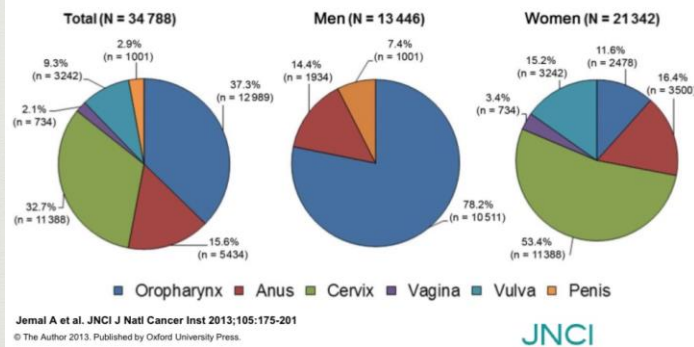


Epithelial tumours		Other epithelial tumours	
Squamous cell tumours and precursors		Adenocarcinoma	
Squamous intraepithelial lesions		Glassy cell carcinoma	
Low-grade squamous intraepithelial lesion	8077/0	Adenoid basal carcinoma	
High-grade squamous intraepithelial lesion	8077/2	Adenoid cystic carcinoma	
Squamous cell carcinoma, NOS	8070/3	Undifferentiated carcinoma	
Keratinizing	8071/3	Neuroendocrine tumours	
Non-keratinizing	8072/3	Low-grade neuroendocrine tumour	
Papillary	8052/3	Carcinoid tumour	
Basaloid	8063/3	Atypical carcinoid tumour	
Warty	8051/3	High-grade neuroendocrine carcinoma	
Verrucous	8051/3	Small cell neuroendocrine carcinoma	
Squamotransitional	8120/3	Large cell neuroendocrine carcinoma	
Lymphoepithelioma-like	8062/3		
Benign squamous cell lesions		Mesenchymal tumours and tumour-like lesions	
Squamous metaplasia		Benign	
Condyloma acuminatum		Leiomyoma	
Squamous papilloma	8062/0	Rhabdomyoma	
Transitional metaplasia		Others	
Glandular tumours and precursors		Malignant	
Adenocarcinoma in situ	8140/2	Leiomyosarcoma	
Adenocarcinoma	8140/3	Rhabdomyosarcoma	
Endocervical adenocarcinoma, usual type	8140/3	Alveolar soft-part sarcoma	
Mucinous carcinoma, NOS	8480/3	Angiosarcoma	
Gastric type	8482/3	Malignant peripheral nerve sheath tumour	
Intestinal type	8144/3	Other sarcomas	
Signet-ring cell type	8490/3	Liposarcoma	
Villoglandular carcinoma	8263/3	Undifferentiated endocervical sarcoma	
Endometrioid carcinoma	8360/3	Ewing sarcoma	
Clear cell carcinoma	8310/3	Tumour-like lesions	
Serous carcinoma	8441/3	Postoperative spindle-cell nodule	
Mesenchymal carcinoma	8110/3	Lymphoma-like lesion	
Adenocarcinomas admixed with neuroendocrine carcinoma	8574/3		
Benign glandular tumours and tumour-like lesions		Mixed epithelial and mesenchymal tumours	
Endocervical polyp		Adenosarcoma	
Müllerian papilloma		Adenosarcoma	
Nabothian cyst		Carcinosarcoma	
Tumour clusters			
Microglandular hyperplasia		Melanocytic tumours	
Lobular endocervical glandular hyperplasia		Blue naevus	
Diffuse laminar endocervical hyperplasia		Malignant melanoma	
Mesonephric remnants and hyperplasia			
Arias-Stella reaction		Germ cell tumours	
Endocervicosis		Yolk sac tumour	
Endometriosis			
Tuboendometrioid metaplasia		Lymphoid and myeloid tumours	
Ectopic prostate tissue		Lymphomas	
		Myeloid neoplasms	
		Secondary tumours	

Эпителиальные опухоли и предраковые поражения шейки МАТКИ

10 HPV types (high-risk): 16; 18; 31; 33; 35; 39; 45; 51; 56; 68

HPV-Associated Cancers United States, 2009



Transforming HPV Infection: Oncogenesis

- Since pRb is deactivated by HPV's E7 → p16 is overexpressed

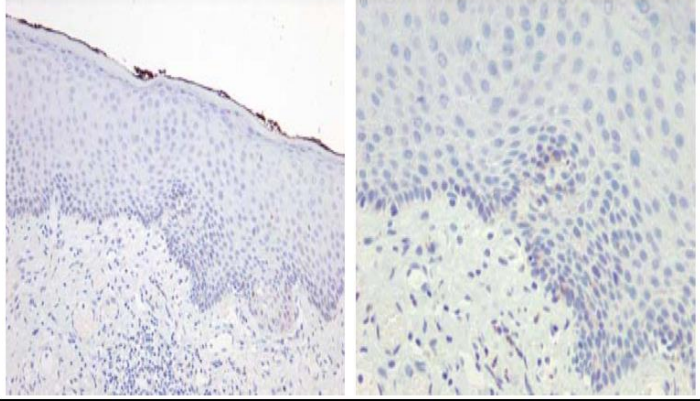


- In cells with transforming HPV infections, HPV viral oncoprotein E7 impairs the function of pRb, disrupting its ability to bind to E2F
- This leads to deregulated cell proliferation, genetic instability and p16 over-expression detectible by immunohistochemistry staining

JOURNAL OF LOWER GENITAL TRACT DISEASE

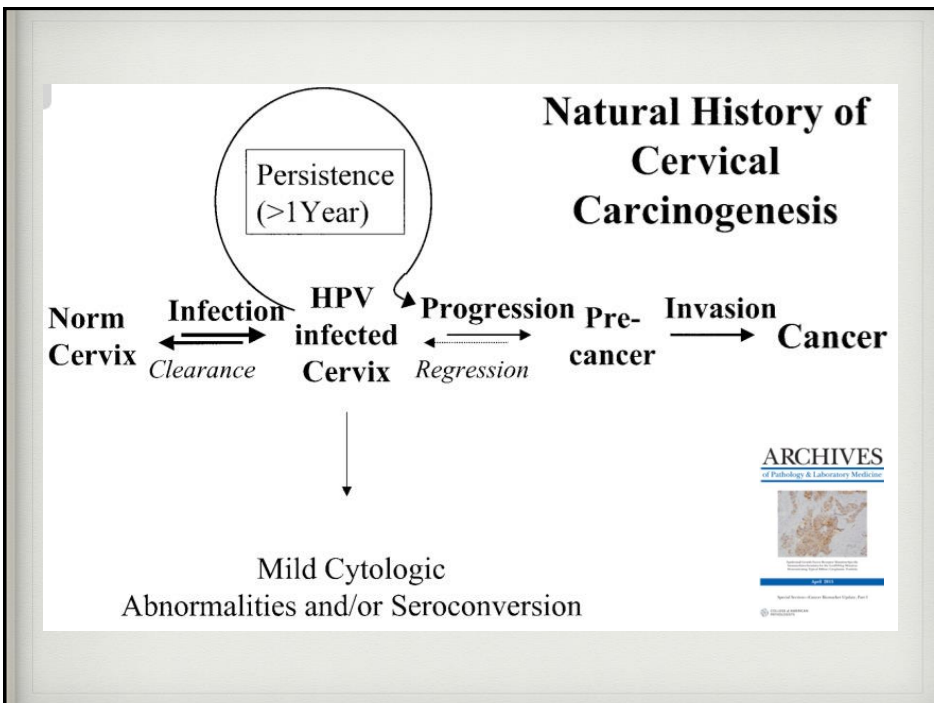
ASCP

P16 in Normal Epithelium: negative or very focally positive



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CAP



New Terminology and new treatment guidelines

- New terminology for HPV related squamous lesions of lower anogenital tract sponsored by the CAP and American Society for Colposcopy and Cervical Pathology

Arch Pathol Lab Med 2012; 136:1-32

J Low Genit Tract Dis 2012;16:205-242

ARCHIVES
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Consensus guidelines for the management of abnormal cervical screening tests and cancer precursors.

J Low Genit Tract Dis 2013; 17: S1-27

JOURNAL OF
LOWER GENITAL
TRACT DISEASE

Унифицированная Терминология

- LSIL, HSIL
- CIN1- 3, VIN 1-3, VAIN 1- 3, AIN 1- 3, PAIN 1 - 3
- Mild, moderate, severe dysplasia
- Vulvar dysplasia "usual" type
- Vulvar intraepithelial carcinoma Bowen's type
- Bowenoid papulosis (clinical diagnosis)
- Erythroplasia of Queyrat
- Bowen disease
- Condyloma
- Carcinoma in situ

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Sites Included in New Terminology

- Cervix
- Vagina
- Vulva
- Peri-anus
- Anus
- Penis
- Scrotum



Sites not included in the new terminology

- Larynx
- Mouth
- Tonsil
- Conjunctiva



Цитологическая диагностика

The Bethesda System 2001



WHO Classification of Tumours of Female Reproductive Organs

Edited by Robert J. Kurman, Maria Luisa Carcangiu, C. Simon Herrington, Robert H. Young

WHO Classification of Tumours of Female Reproductive Organs.

Fourth Edition
June 2014

WHO Classification of tumours of the uterine cervix^{a,b}

Epithelial tumours		Other epithelial tumours	
Squamous cell tumours and precursors		Adenosquamous carcinoma	85
Squamous intraepithelial lesions		Glassy cell carcinoma	80
Low-grade squamous intraepithelial lesion	8077/0	Adenoid basal carcinoma	80
High-grade squamous intraepithelial lesion	8077/2	Adenoid cystic carcinoma	80
Squamous cell carcinoma, NOS	8070/3	Undifferentiated carcinoma	80
Keratinizing	8071/3	Neuroendocrine tumours	
Non-keratinizing	8072/3	Low-grade neuroendocrine tumour	

WHO Classification of Tumours of Female Reproductive
Organs.

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**Low-grade squamous
intraepithelial lesion**

Definition

An intraepithelial lesion of squamous epithelium that represents the clinical and morphological manifestation of a productive HPV infection. Low-grade refers to the associated low risk of concurrent or future cancer.

ICD-O code 8077/0

Synonyms

Cervical intraepithelial neoplasia, grade 1 (CIN 1); mild squamous dysplasia; flat condyloma; koilocytotic atypia; koilocytosis

**High-grade squamous
intraepithelial lesion**

Definition

A squamous intraepithelial lesion that carries a significant risk of invasive cancer development if not treated {412,1586}.

ICD-O code 8077/2

Synonyms

Cervical intraepithelial neoplasia, grade 2 (CIN 2); cervical intraepithelial neoplasia, grade 3 (CIN 3); moderate squamous dysplasia; severe squamous dysplasia; squamous carcinoma in situ (CIS)

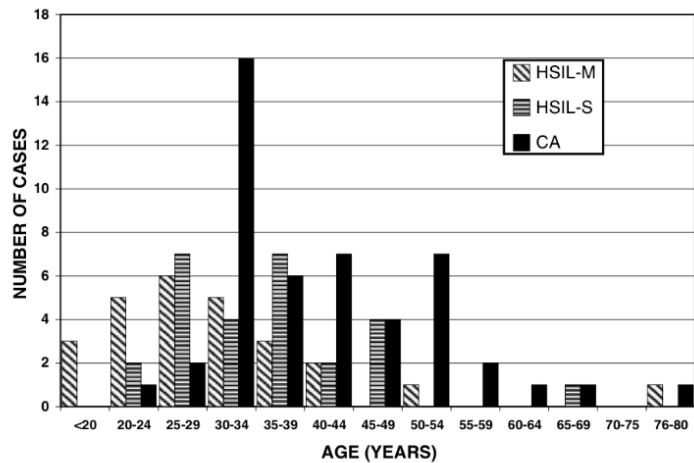
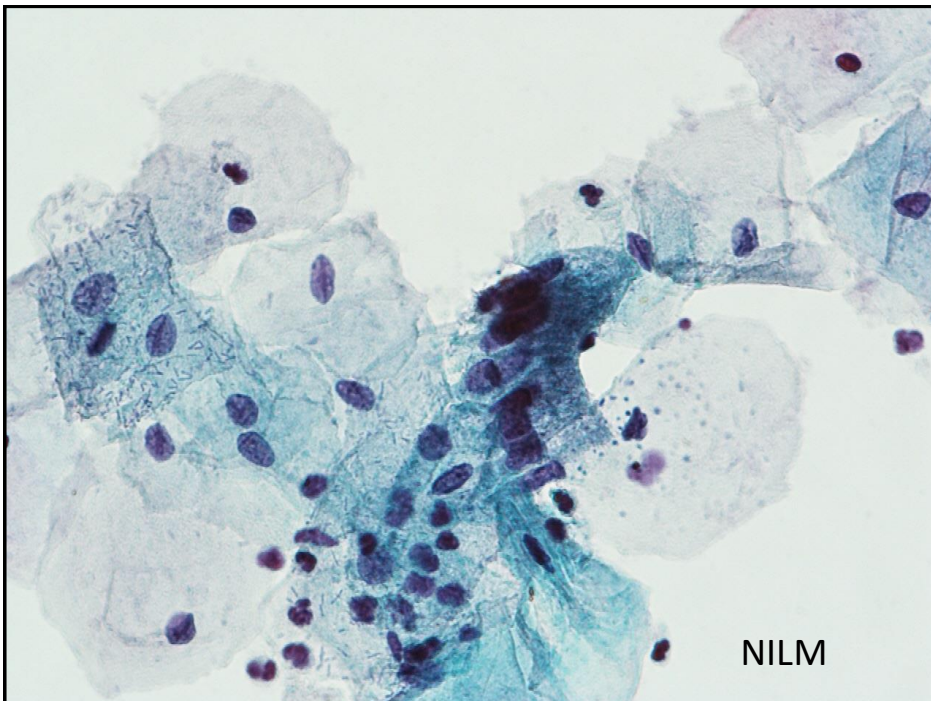
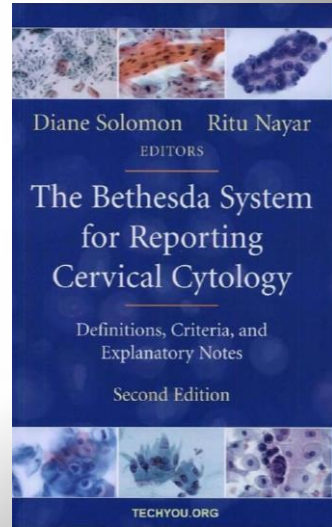


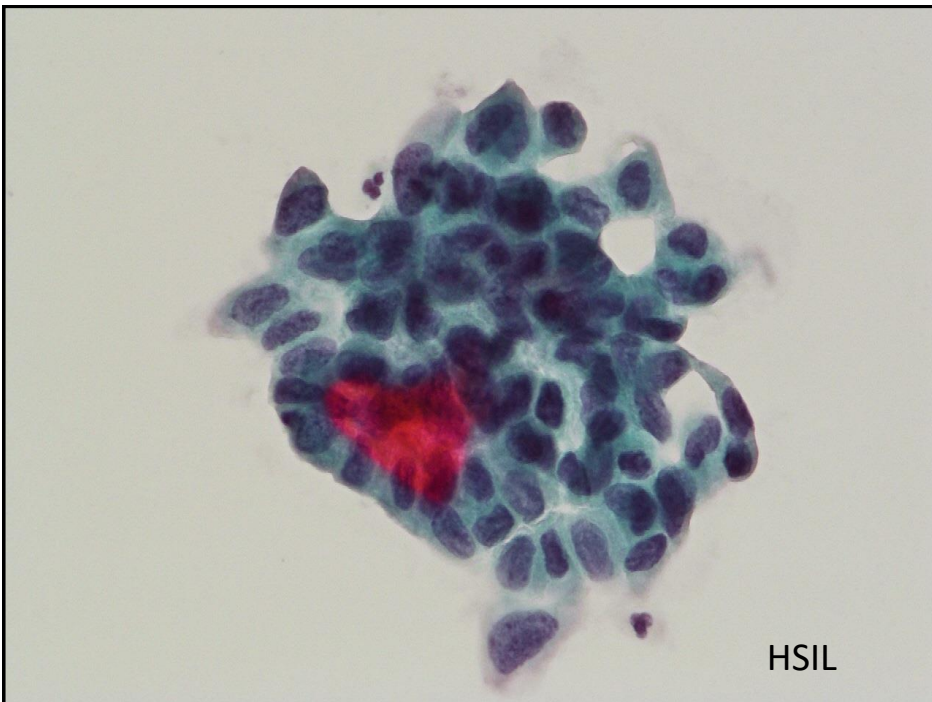
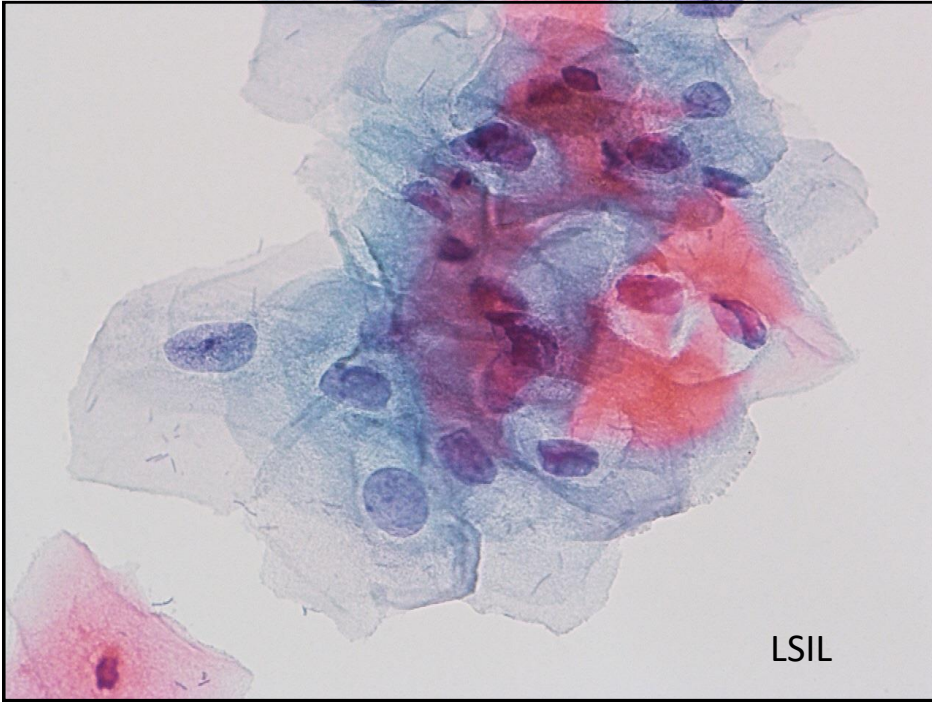
Figure 1 Age distribution of cases with HSIL-M (moderate dysplasia), HSIL-S (severe dysplasia) and invasive carcinoma (CA).

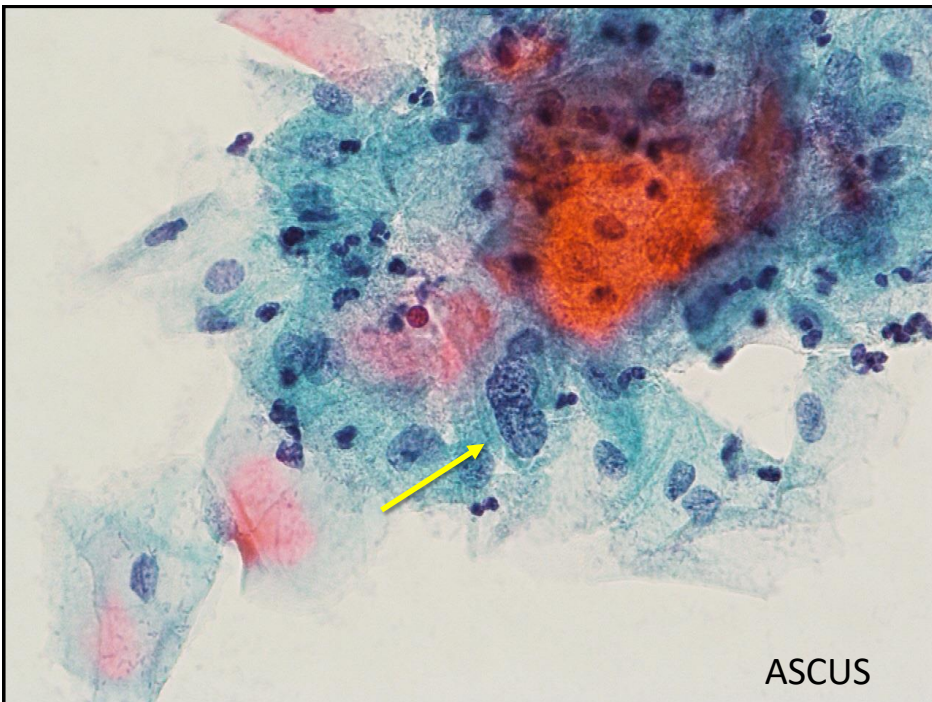
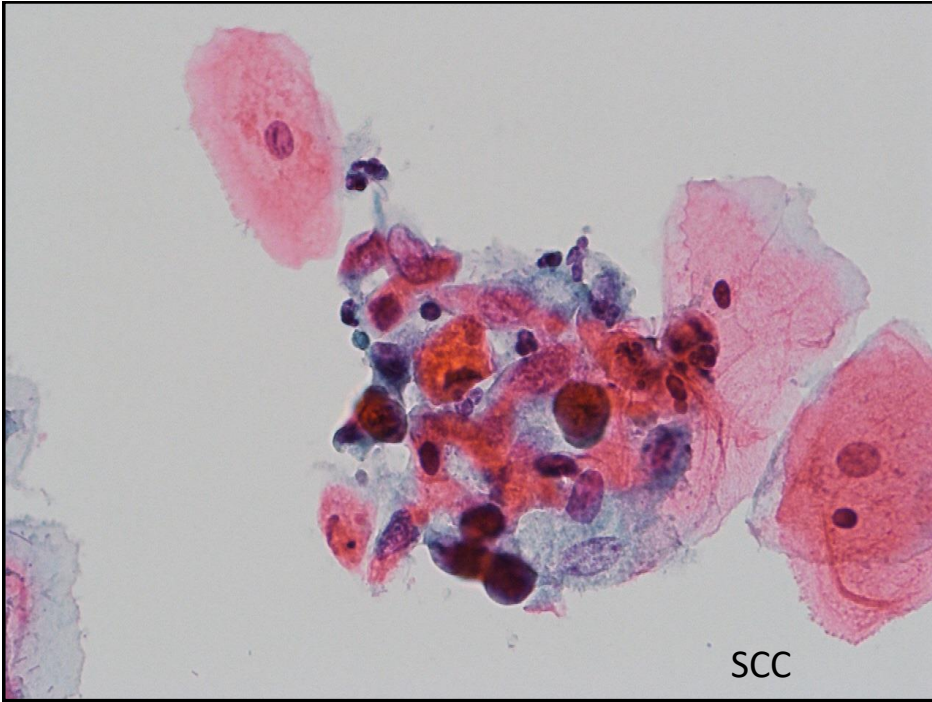
Modern Pathology (2004) 17, 1314–1322

Классификация Bethesda (The Bethesda System 2001)

1. Адекватность препарата
2. Основные категории
3. Интерпретация









МАТЕРИАЛ	Пап тест.	
Клинический диагноз:	Синдром предменструального напряжения. N86 .	
ЦИТОЛОГИЧЕСКОЕ ЗАКЛЮЧЕНИЕ	ТИП ОБРАЗЦА ДЛЯ ИССЛЕДОВАНИЯ	SPECIMEN TYPE
	Тонкослойный препарат на основе метода жидкостной цитологии (NovaPrep®).	Liquid-based thin layer method (NovaPrep®).
	КАЧЕСТВО ПРЕПАРАТА	SPECIMEN ADEQUACY
	Удовлетворительное для исследования:	Satisfactory for evaluation:
	Эндоцервикальные клетки отсутствуют.	Endocervical cells absent.
	ОСНОВНЫЕ КАТЕГОРИИ	GENERAL CATEGORIZATION
	Обнаружены изменения эпителиальных клеток.	Epithelial cell abnormality.
	ИНТЕРПРЕТАЦИЯ/РЕЗУЛЬТАТ	INTERPRETATION/RESULT
	Изменения клеток плоского эпителия:	Squamous cell Abnormalities:
	Клетки плоского эпителия с атипией неясного значения (ASC-US).	Atypical squamous cells of undetermined significance (ASC-US).
Организм/патоген:	Organism:	
Элементы гриба типа Candida.	Fungal organisms morphologically consistent with Candida spp.	
РЕКОМЕНДАЦИИ*	RECOMMENDATION*	
Показана кольпоскопия. Report according to 2001 Bethesda terminology.	Refer for colposcopy.	

*Согласно рекомендациям ASCCP (2013) .



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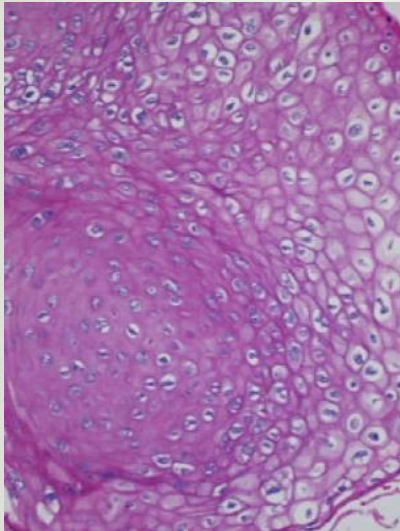
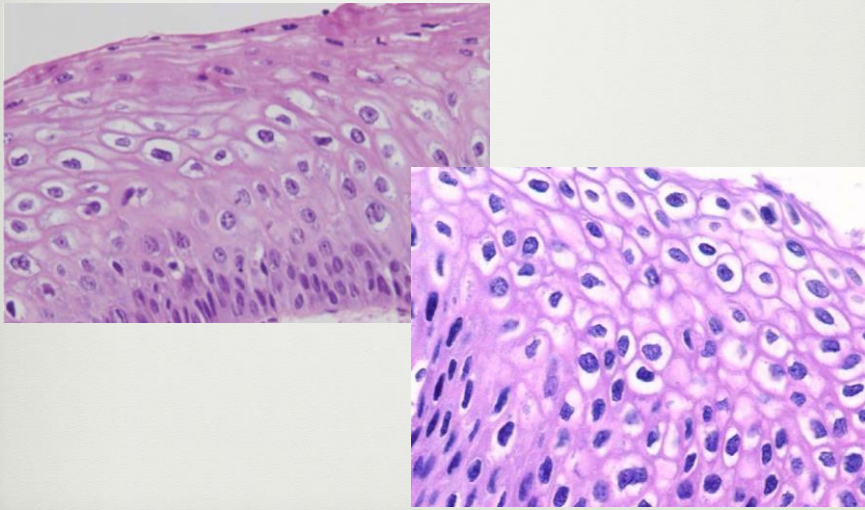
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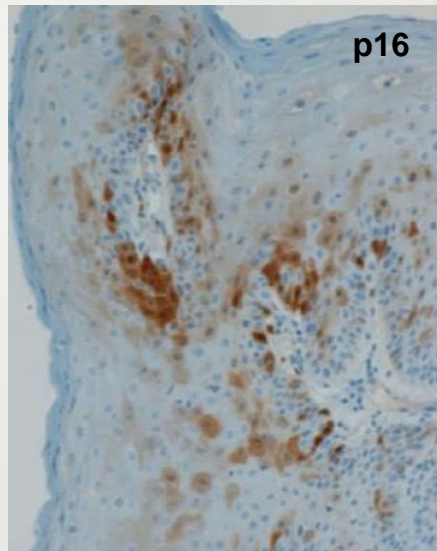
ICD-O code 8077/2

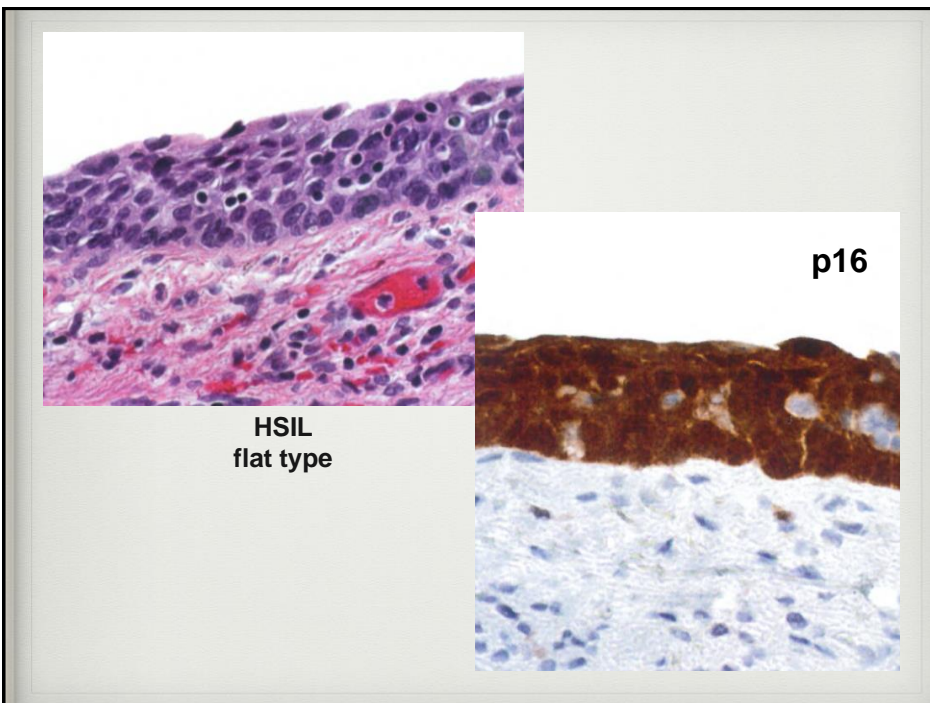
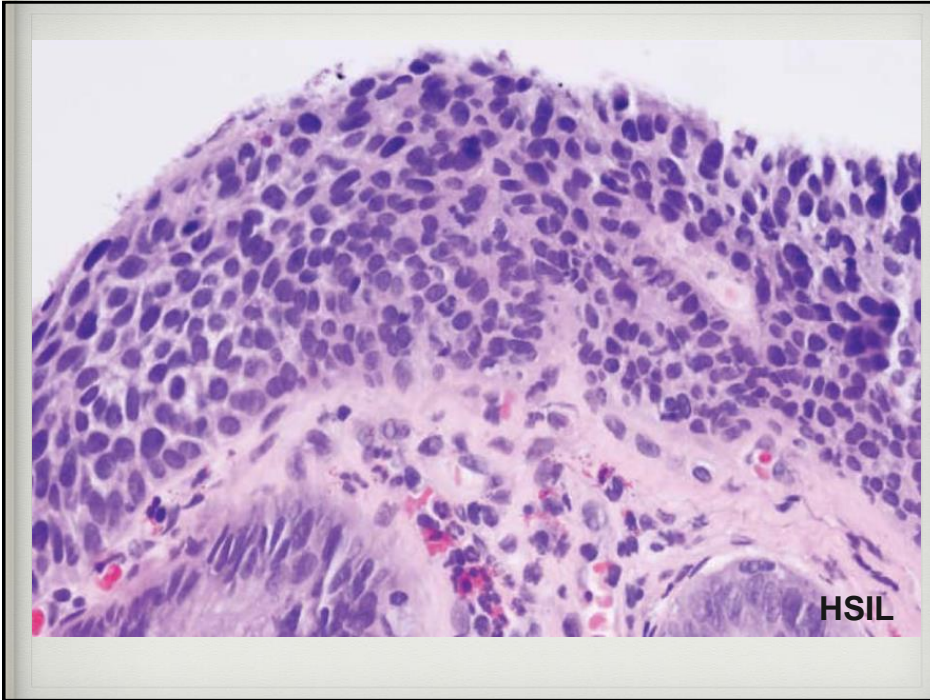
Synonyms
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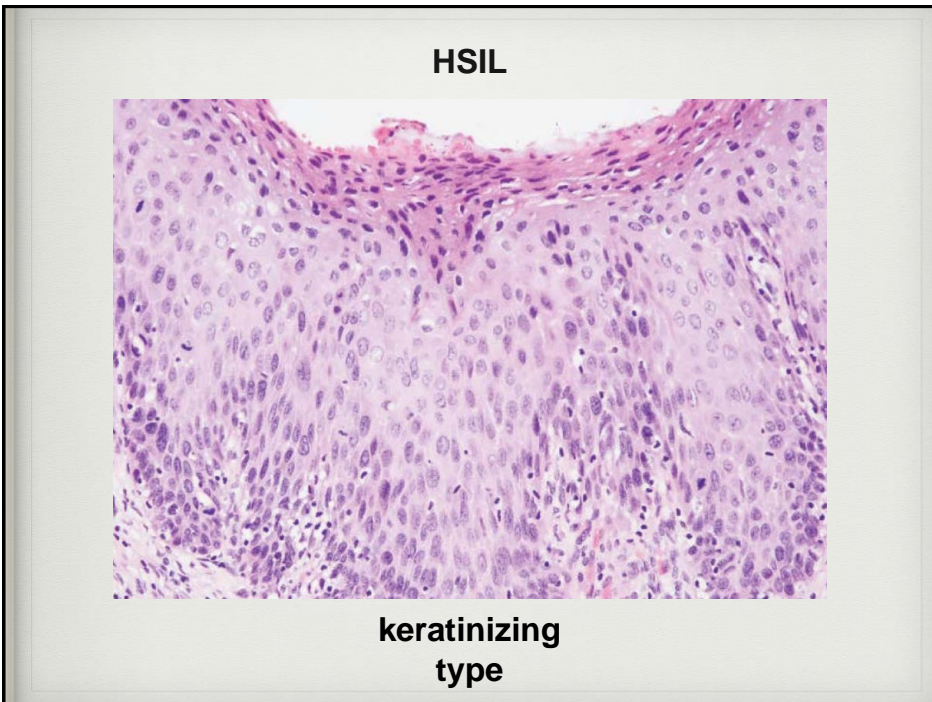
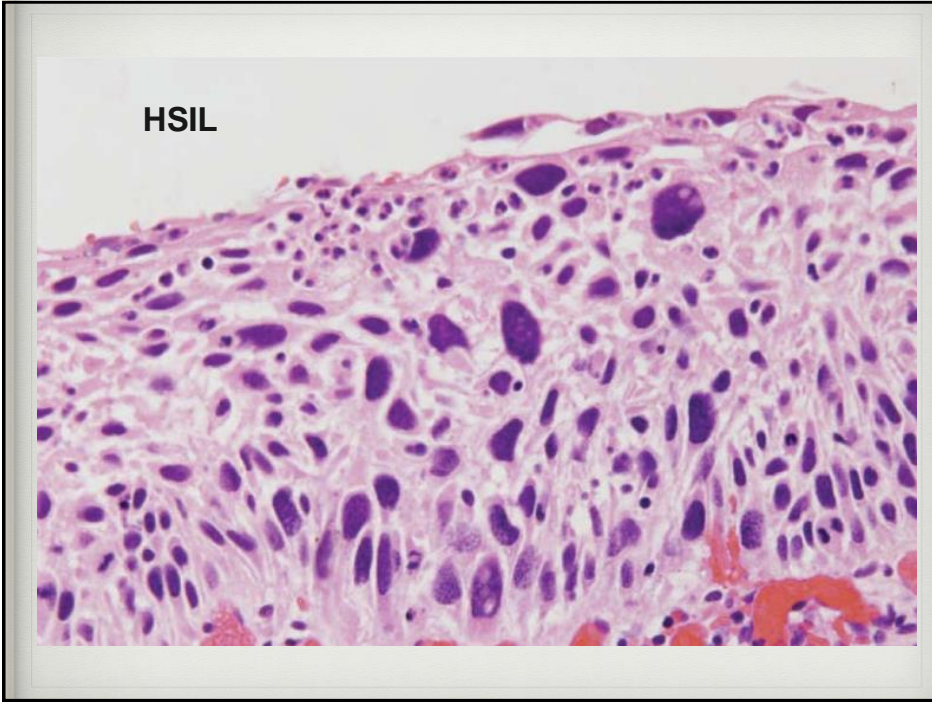
LSIL

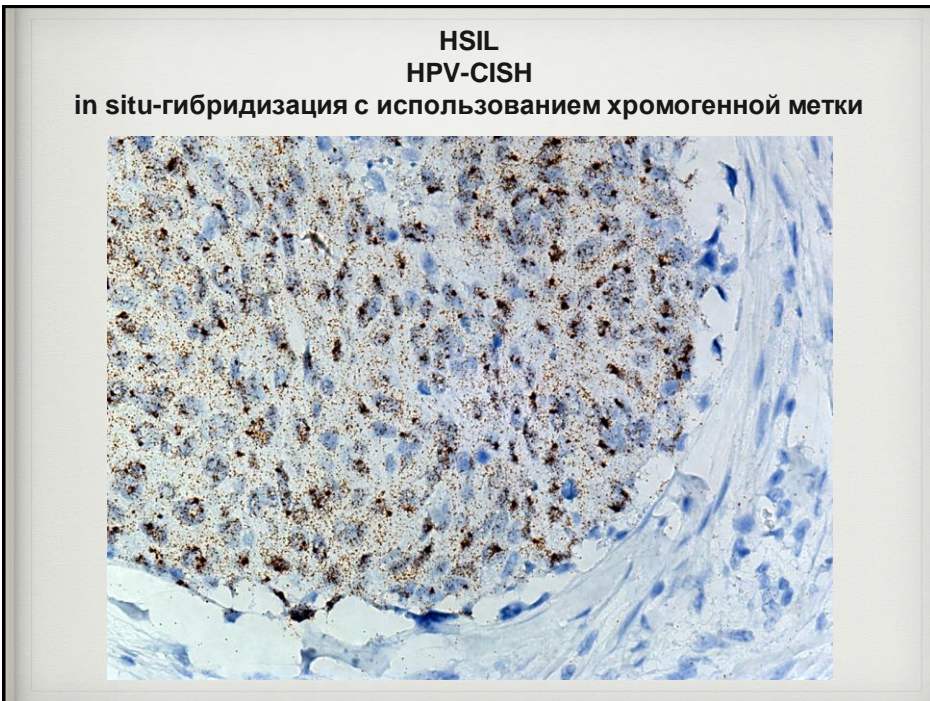
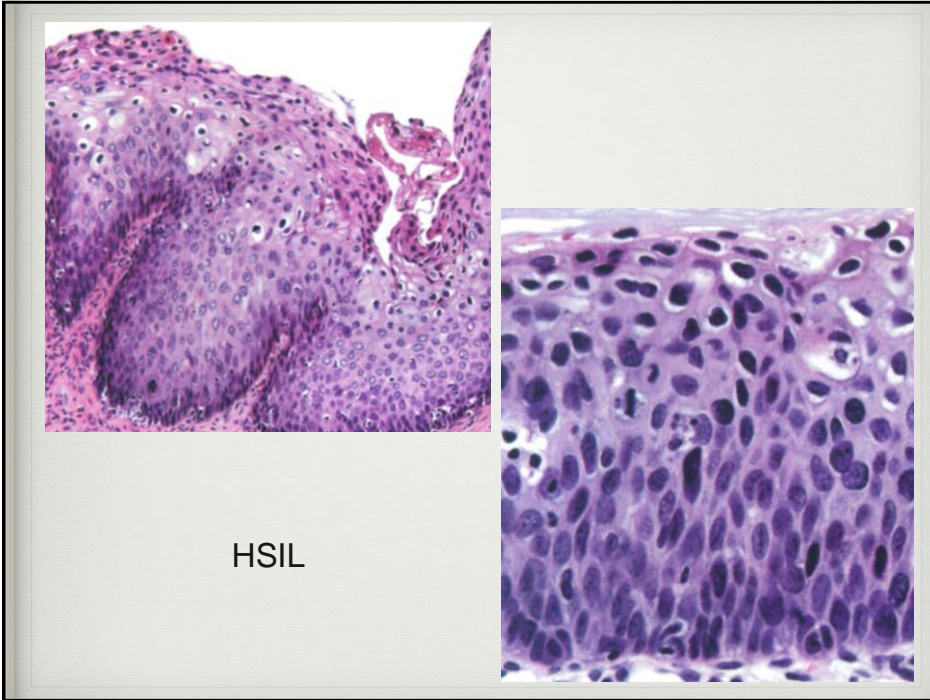


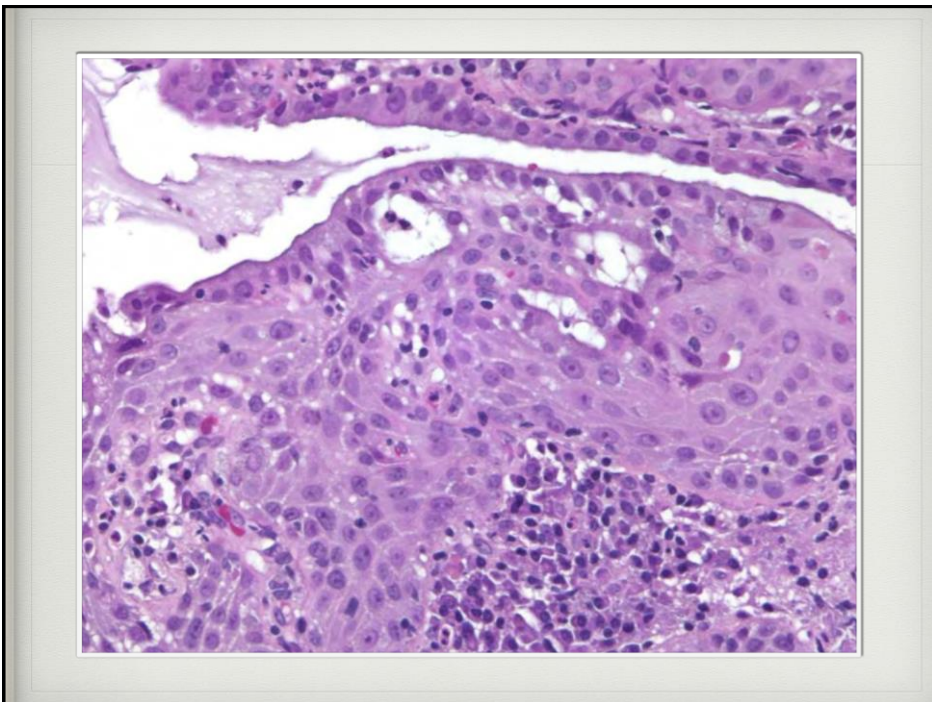
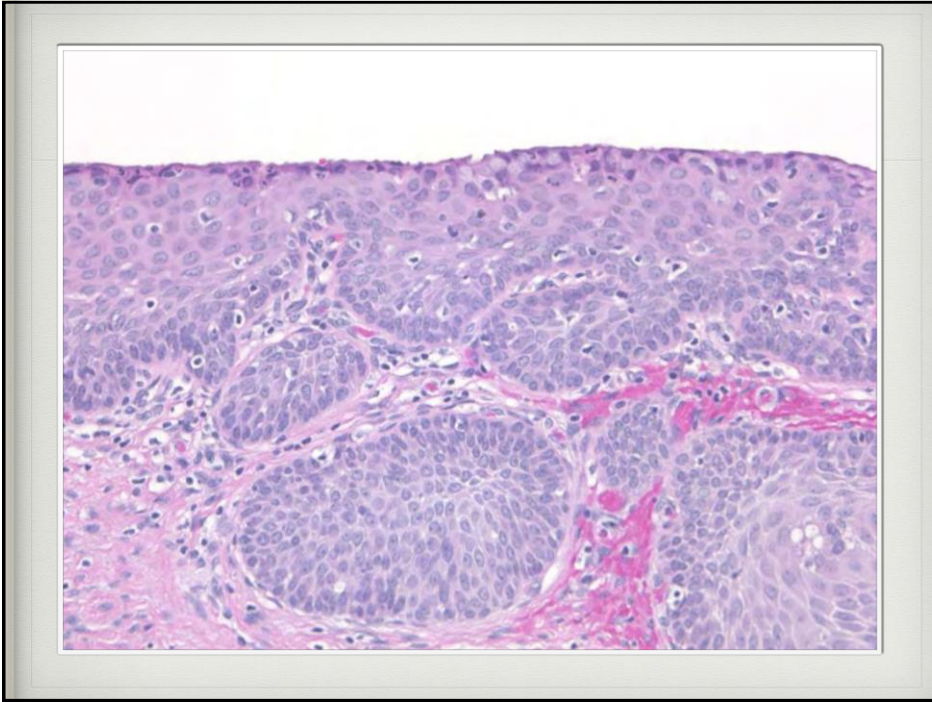
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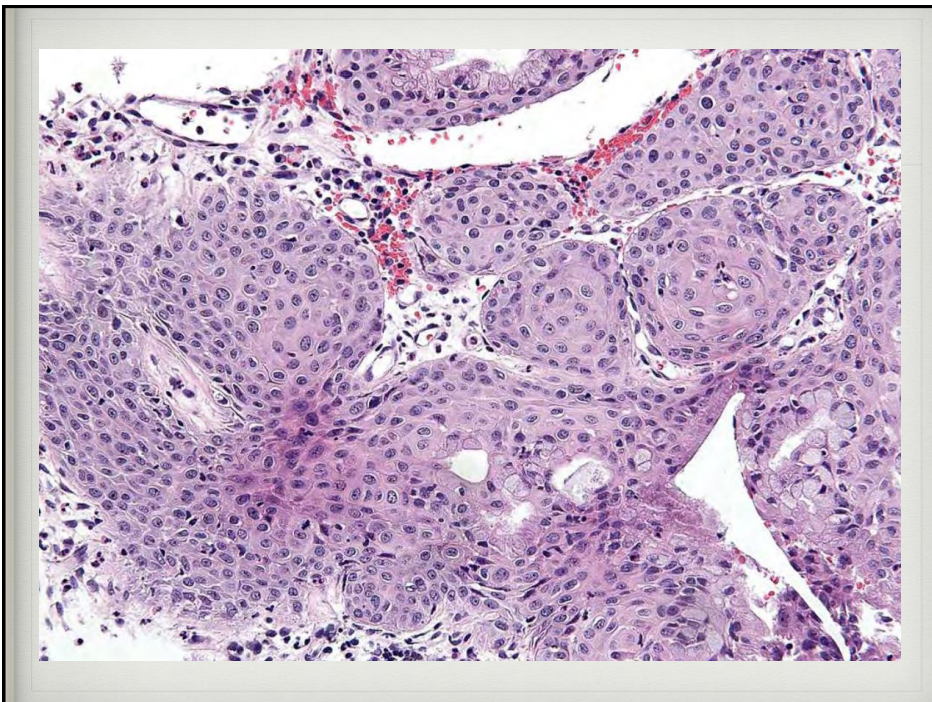
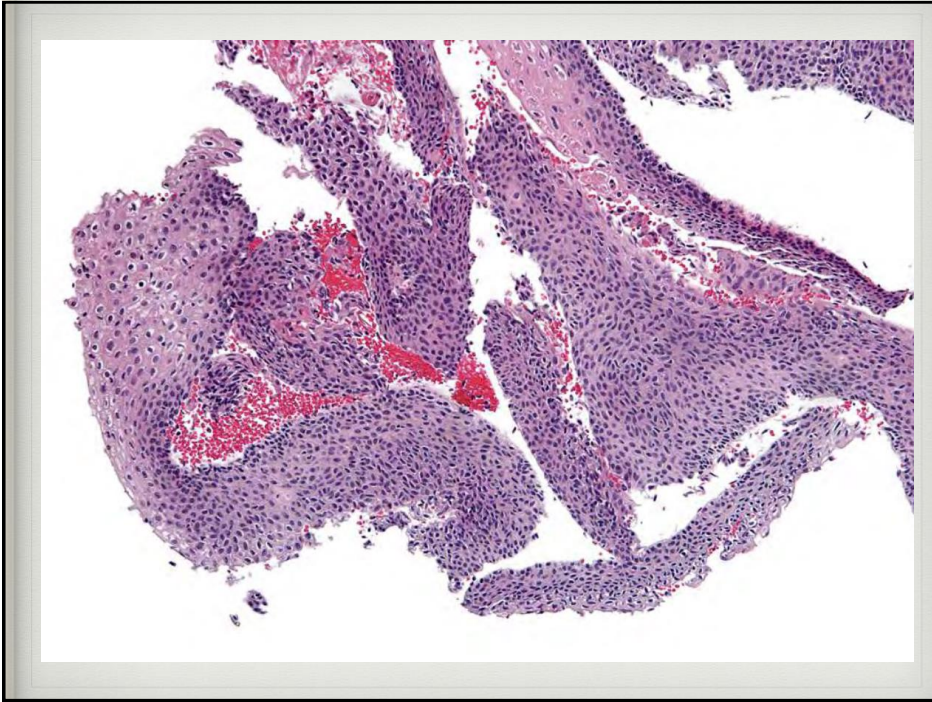


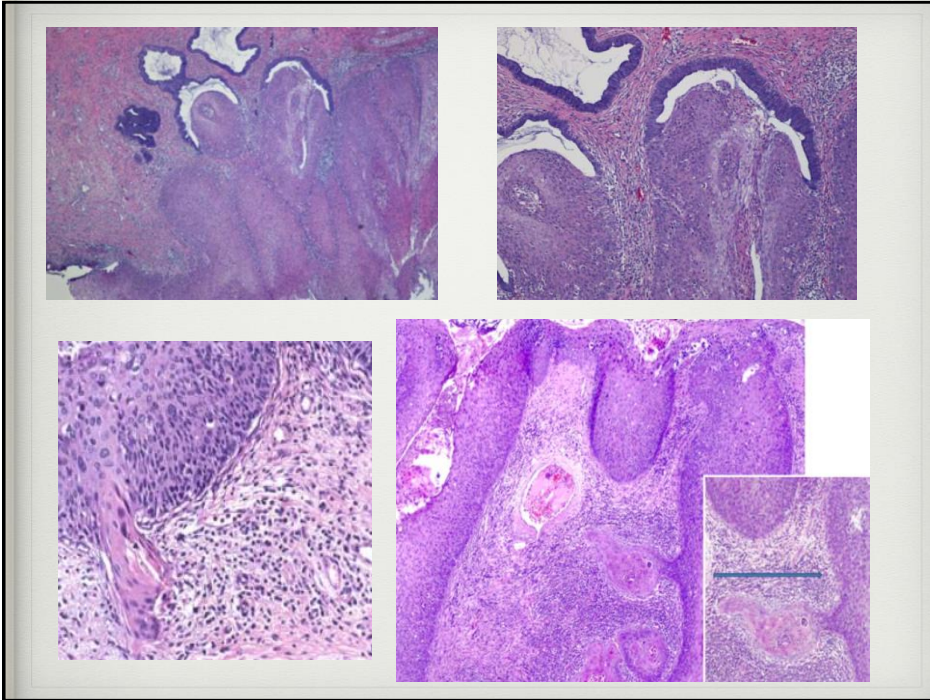












~~minimal invasive carcinoma~~

~~early invasive carcinoma~~

CARCINOMA with SUPERFICIAL
STROMAL INVASION

**ТОЛЬКО ДЛЯ
SCC**



Squamous cell carcinoma, NOS	8070/3
Keratinizing	8071/3
Non-keratinizing	8072/3
Papillary	8052/3
Basaloid	8083/3
Warty	8051/3
Verrucous	8051/3
Squamotransitional	8120/3
Lymphoepithelioma-like	8082/3

Cervical carcinoma: FIGO staging of carcinomas that can be seen only microscopically

- IA1 depth invasion ≤ 3 mm, horizontal extension ≤ 7 mm
- IA2 depth invasion > 3 mm and ≤ 5 mm, horizontal extension ≤ 7 mm
- IB1 depth > 5 mm or horizontal extension > 7 mm

Treatment of Cervical cancer

IA1 without LVI:

- Cone biopsy with negative margins or
- Extradiscal hysterectomy

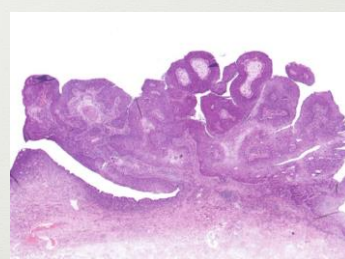
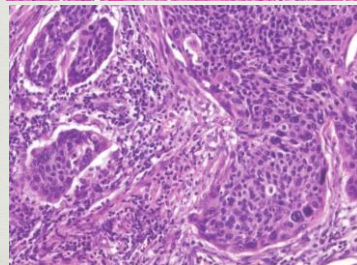
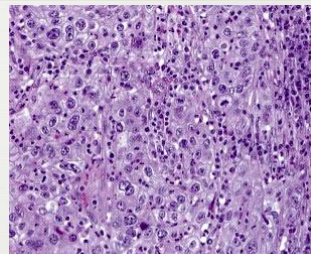
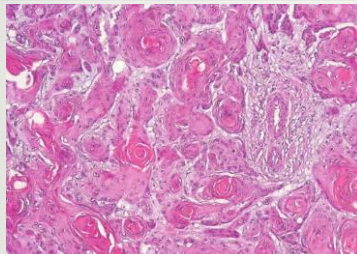
IA1 with LVI or IA2:

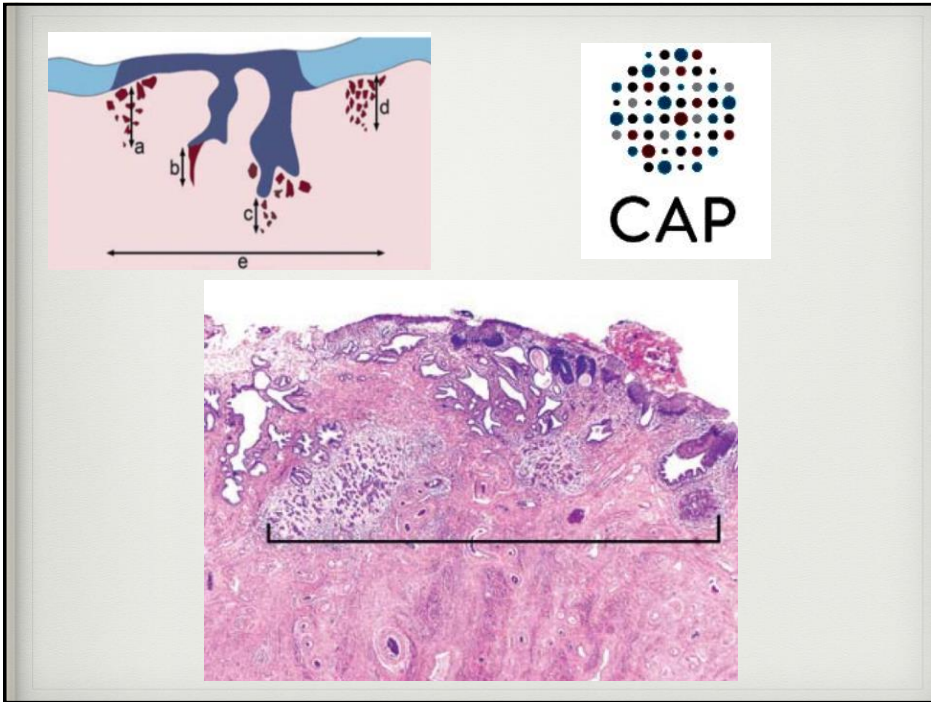
- Cone biopsy with negative margins+ pelvic nodes or radical trachelectomy + pelvic nodes or
- Modified radical hysterectomy + pelvic nodes

IB1:

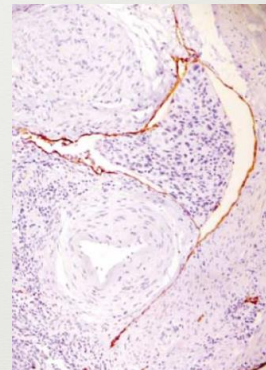
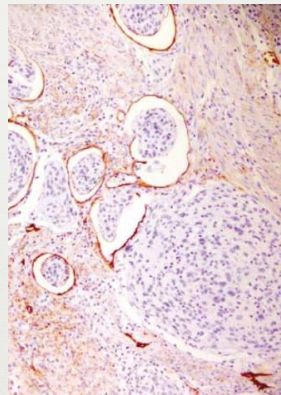
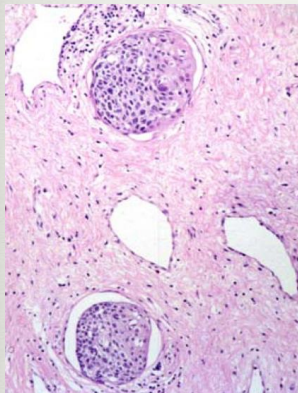
- Radical trachelectomy + pelvic nodes or
- Radical hysterectomy + pelvic nodes

Гистологические варианты плоскоклеточной карциномы шейки матки





Invasion of lymphovascular space (LVI)

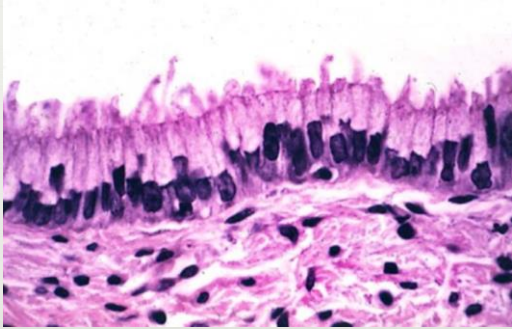


Podoplanin (D2-40)

Glandular tumours and precursors		
Adenocarcinoma in situ		8140/2
Adenocarcinoma		8140/3
Endocervical adenocarcinoma, usual type		8140/3
Mucinous carcinoma, NOS		8480/3
Gastric type		8482/3
Intestinal type		8144/3
Signet-ring cell type		8490/3
Villoglandular carcinoma		8263/3
Endometrioid carcinoma		8380/3
Clear cell carcinoma		8310/3
Serous carcinoma		8441/3
Mesonephric carcinoma		9110/3
Adenocarcinoma admixed with neuroendocrine carcinoma		8574/3

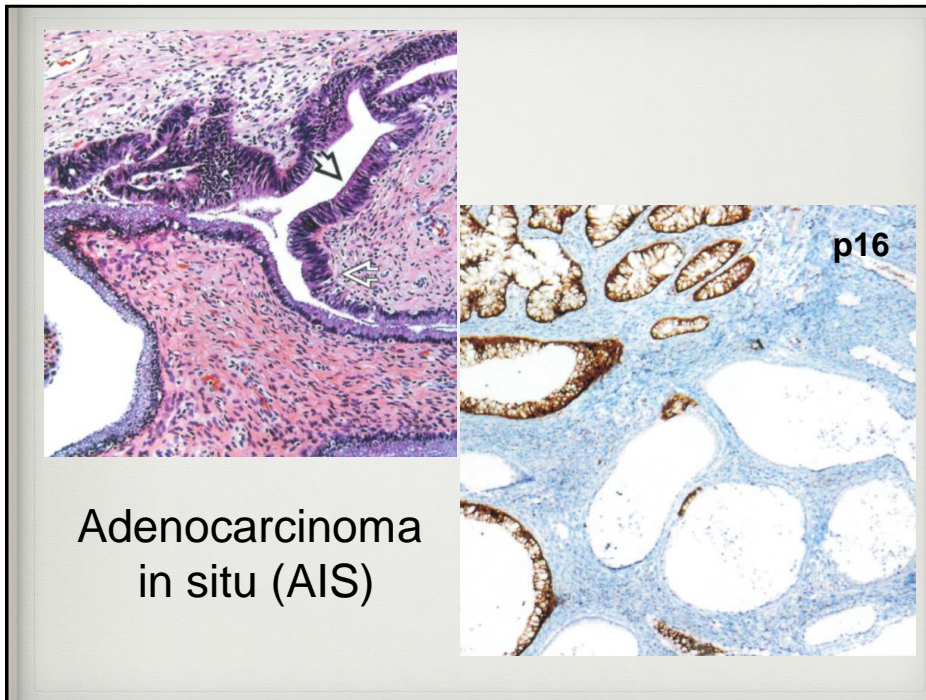
Adenocarcinoma in situ (AIS)

Adenocarcinoma



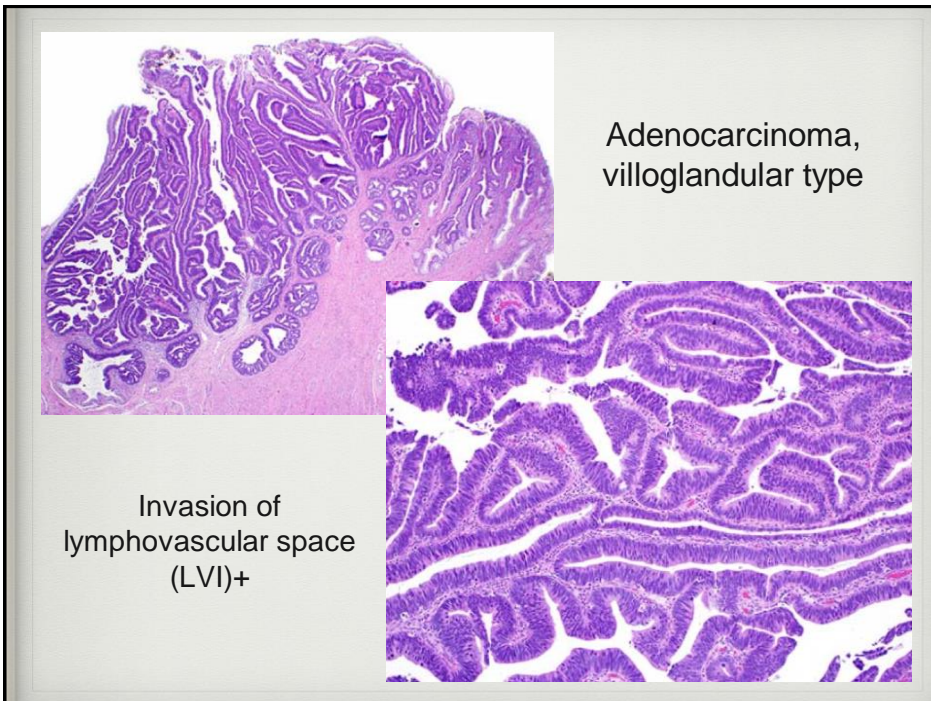
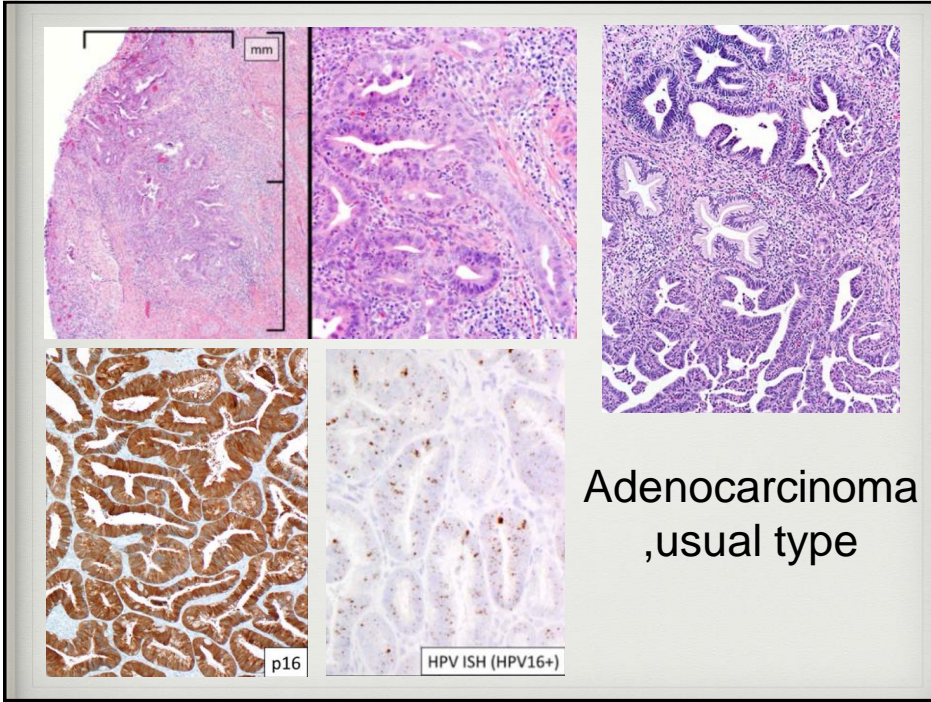
Adenocarcinoma in situ (AIS)

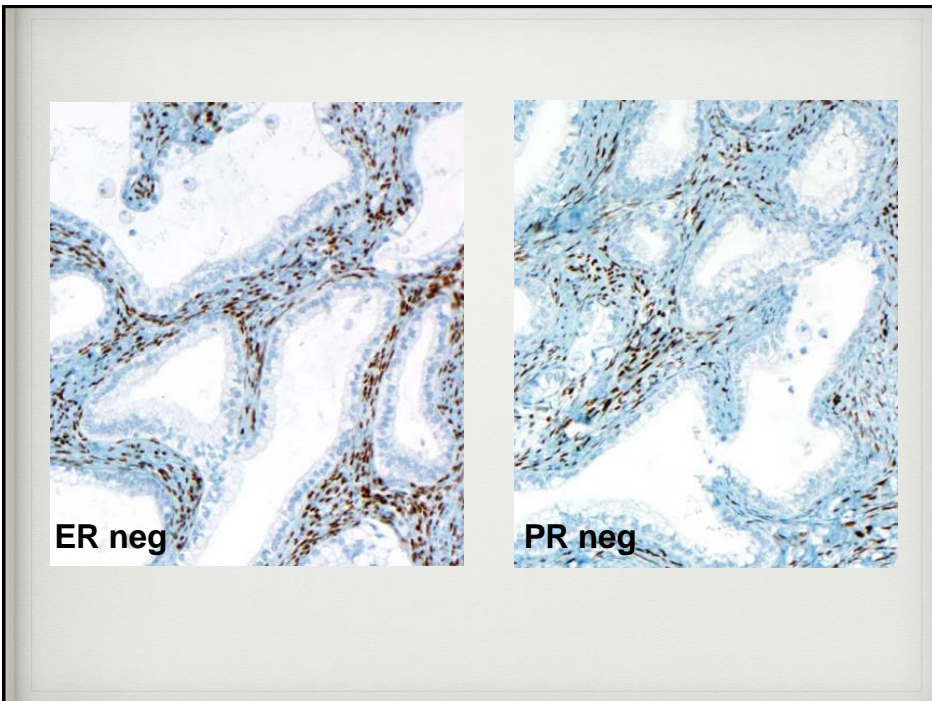
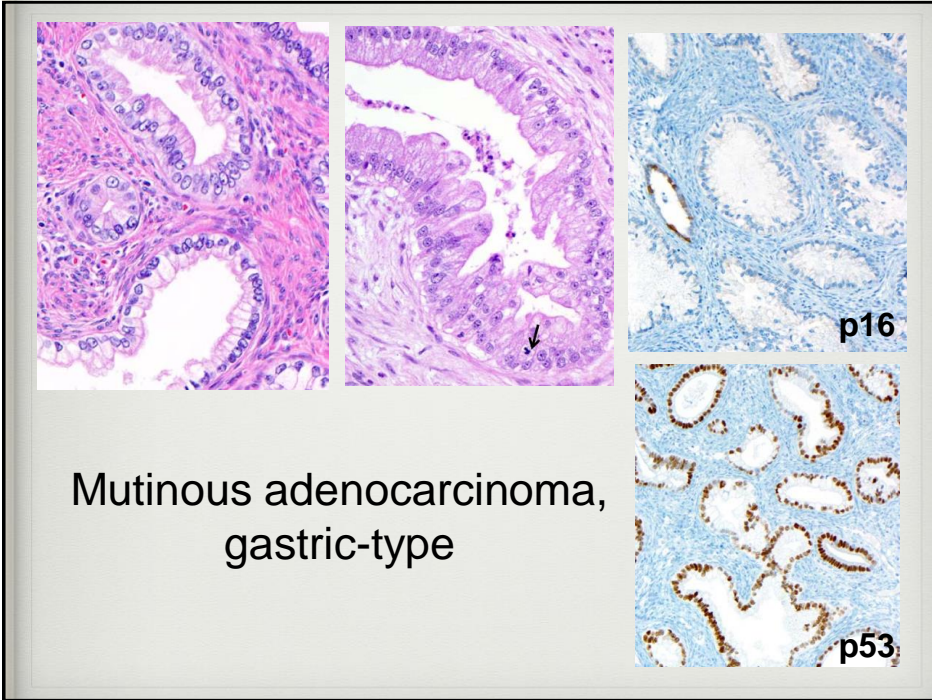


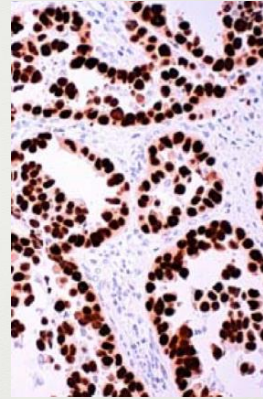
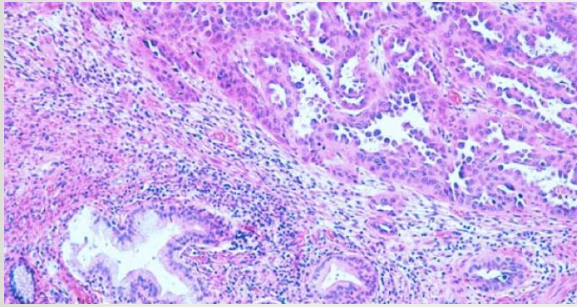


Adenocarcinoma

Adenocarcinoma	8140/3
Endocervical adenocarcinoma, usual type	8140/3
Mucinous carcinoma, NOS	8480/3
Gastric type	8482/3
Intestinal type	8144/3
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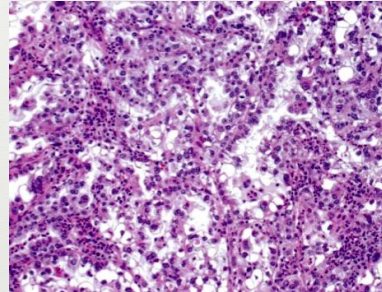
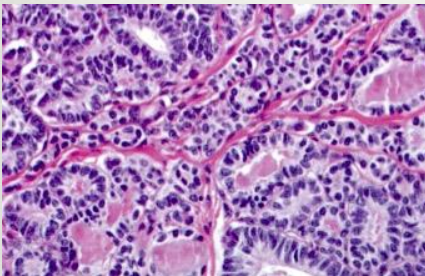
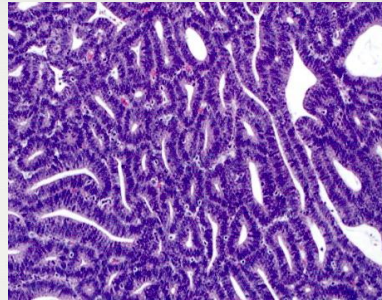
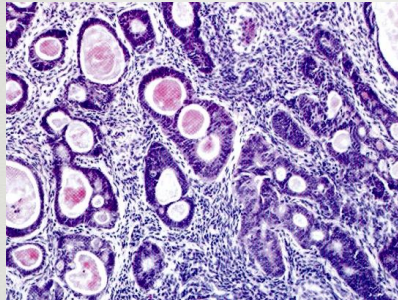




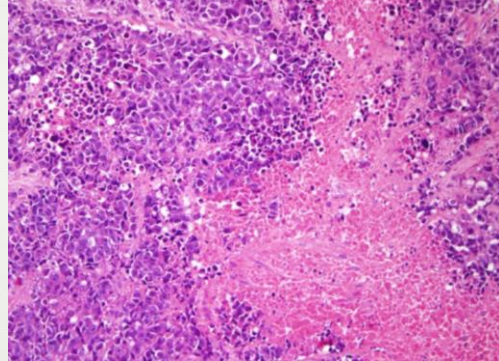
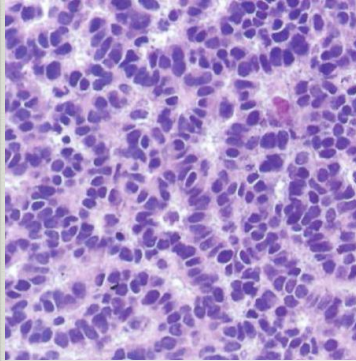


Serous adenocarcinoma

p53
+++



Neuroendocrine tumor & carcinoma



NET (G1-2) low grade

NEC (G3) high grade

Tumor	Growth Patterns	Cytologic Atypia	Mitotic Activity	Necrosis
Carcinoid	Organoid, insular, trabecular, cord-like	None to mild	Very rare	No
Atypical carcinoid	Organoid, insular, trabecular, cord-like	Mild to moderate	5-10 per 10 HPF	Focal
Small cell neuroendocrine carcinoma	Diffuse, nested, trabecular, cord-like	Moderate to severe	> 10 per 10 HPF	Extensive
Large cell neuroendocrine carcinoma	Diffuse, trabecular, and cord-like with peripheral palisading	Severe	> 10 per 10 HPF	Geographic

HPV-related & non-HPV-related tumor types

DOI: 10.1111/1471-0528.13071
www.bjog.org

Gynaecological oncology

HPV-negative carcinoma of the uterine cervix: a distinct type of cervical cancer with poor prognosis

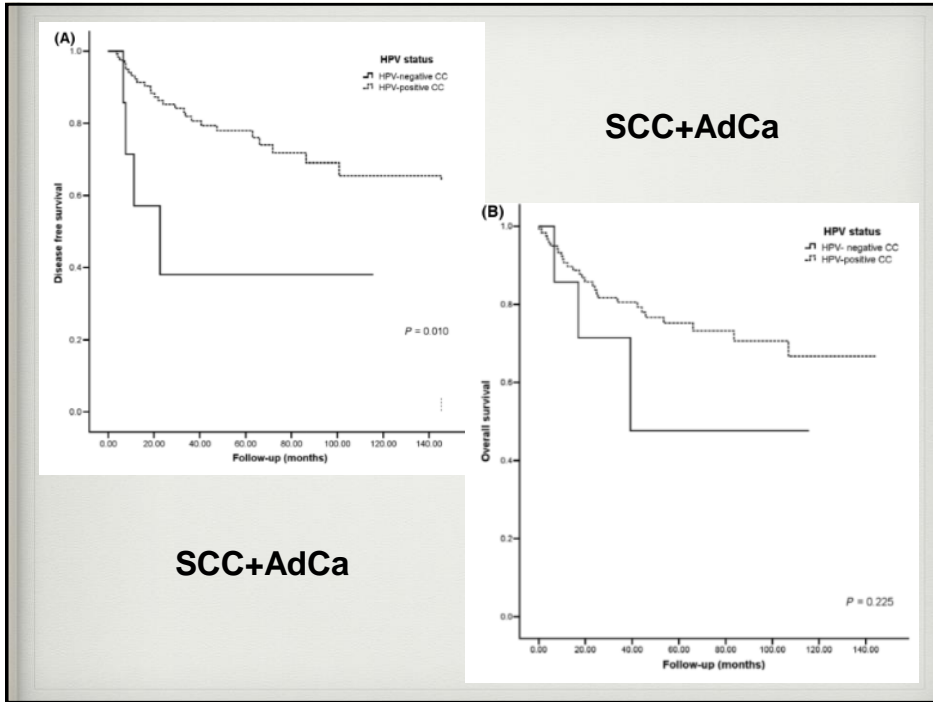
L Rodríguez-Carunchio,^a I Soveral,^b RDM Steenbergen,^c A Torné,^b S Martínez,^b P Fusté,^b J Pahisa,^b L Marimon,^a J Ordi,^a M del Pino^b

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Pathology Excellence Through Education



Patterns of Invasion of Cervical Adenocarcinoma as Predictors of Outcome

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2015 ANNUAL MEETING



Cervical carcinoma: FIGO staging of carcinomas that can be seen only microscopically

- IA1 depth invasion \leq 3 mm, horizontal extension \leq 7 mm
- IA2 depth invasion $>$ 3 mm and \leq 5mm, horizontal extension \leq 7 mm
- IB1 depth $>$ 5 mm or horizontal extension $>$ 7 mm

Treatment of Cervical cancer

IA1 without LVI:

- Cone biopsy with negative margins or
- Extradiscal hysterectomy

IA1 with LVI or IA2:

- Cone biopsy with negative margins+ pelvic nodes or radical trachelectomy + pelvic nodes or
- Modified radical hysterectomy + pelvic nodes

IB1:

- Radical trachelectomy + pelvic nodes or
- Radical hysterectomy + pelvic nodes

Stage IA EAC literature review

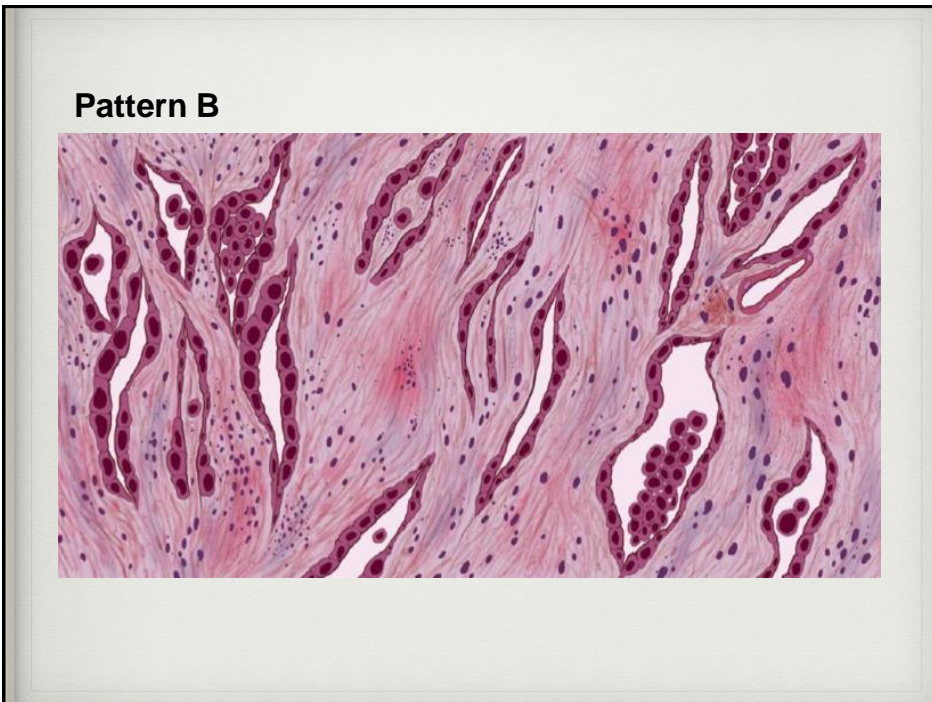
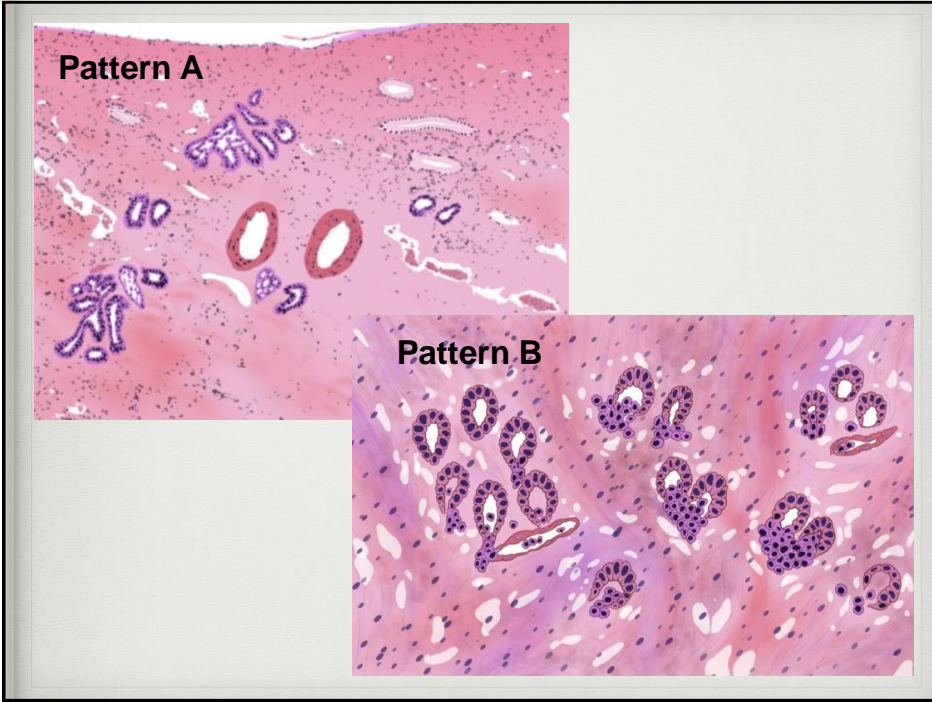
Lymph node metastasis

- IA1 2/228 (0.9%)
- IA2 3/179 (1.7%)

Recurrence

- IA1 4/330 (0.9%)
- IA2 7/346 (2%)

Poynor EA, et al. Gynecologic oncology 2006



Results

	Patients	Pts with pos LN	Total LN	# Pos LN	Stage I	Stage II-IV
Standard	352	49 (14%)	7993	83 (1%)	320 (91%)	32 (9%)
A	73 (20.7%)	0	1516	0	73 (100%)	0
B	90 (25.6%)	4 (4.4%)	2059	5 (0.2%)	90 (100%)	0
C	189 (53.7%)	45 (24%)	4418	76 (1.7%)	157 (83%)	32 (17%)

LN metastases

$p < 0.0001$ comparing Pattern A to B/C

$p = 0.0153$ comparing Pattern A to B

$p < 0.0001$ comparing Pattern A to C

21% of patient – spared
lymphadenectomy



MODERN PATHOLOGY (2011) 24, 726–728
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Molecular mechanisms of epidermal growth factor receptor overexpression in patients with cervical cancer

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The epidermal growth factor receptor is overexpressed in 70–90% of cervical cancers. Previously, we have shown that epidermal growth factor receptor overexpression independently predicts poor prognosis in cervical cancer patients, which makes it a potential therapeutic target. The aim of this study was to systematically analyze the molecular mechanism leading to epidermal growth factor receptor overexpression in cervical cancer. All experiments were performed on archival paraffin-embedded material. In 166 cervical cancer patients, cytoplasmic, membrane and phosphorylated epidermal growth factor receptor protein expression were studied in association with patient survival. Membrane epidermal growth factor receptor overexpression was associated with poor disease-specific survival ($P = 0.027$). This association was particularly present in human papillomavirus 16-positive patients ($P = 0.029$). We analyzed whether epidermal growth factor receptor overexpression was caused by gene amplification using fluorescence *in situ* hybridization. Epidermal growth factor receptor gene copy number was linked to chromosome 7 ploidy, as no gene amplification could be detected when corrected for chromosome 7 centromeric signals. Chromosome 7 aneuploidy was associated with membrane epidermal growth factor receptor overexpression ($P = 0.013$). Additional mutation analysis was performed by sequencing pure, flow-sorted tumor cells, but no mutations were detected. Furthermore, human papillomavirus 16 E5 and E6 oncogene mRNA expression was measured, using quantitative real-time polymerase chain reaction, to determine the association between the human papillomavirus and epidermal growth factor receptor overexpression. High human papillomavirus 16 E5 and E6 mRNA expression were associated with decreased survival ($P = 0.045$ and 0.047, respectively). High human papillomavirus 16 E6 mRNA expression was associated with membrane epidermal growth factor receptor overexpression ($P = 0.013$). This is the first study performed on cancer patient material showing that chromosome 7 aneuploidy and high human papillomavirus 16 E6 mRNA expression lead to membrane epidermal growth factor receptor overexpression in cervical cancer.

Modern Pathology (2011) 24, 726–728; doi:10.1038/modpathol.2010.239; published online 21 January 2011

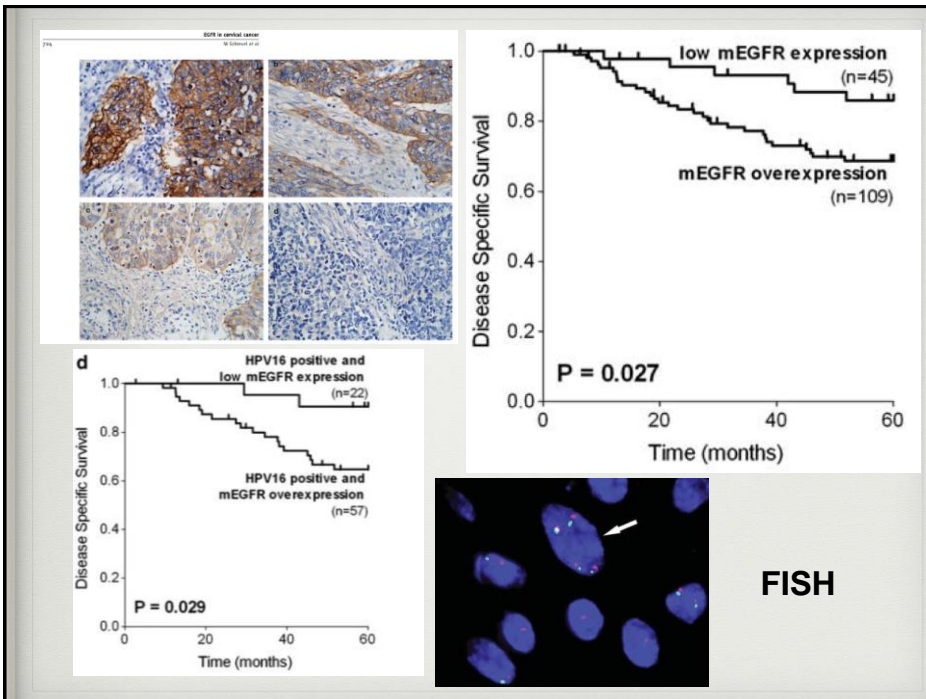
Keywords: aneuploidy; cervical cancer; E5; E6; epidermal growth factor receptor; HPV

Table 1 Patient characteristics

Clinicopathological parameter	N (%) ^a
<i>Age at diagnosis (years)</i>	
Median	45.0
Range	24–87
<i>FIGO stage^b</i>	
Ib1	62 (40)
Ib2	45 (29)
Ila	23 (15)
Ilb	21 (14)
IIIb	3 (2)
IV	1 (1)
<i>HPV type</i>	
16	82 (54)
18	34 (22)
Other	25 (16)
Negative	12 (8)
<i>Histopathology</i>	
Squamous cell carcinoma	100 (60)
Adenosquamous carcinoma	49 (30)
Adeno carcinoma	17 (10)
Lymph nodes positive	52 (31)
Tumor size ≥ 40 mm	75 (49)
Vaso-invasion	93 (57)
Infiltration depth ≥ 15 mm	69 (43)
Parametrial infiltrated	21 (13)
Postoperative radiotherapy	96 (58)

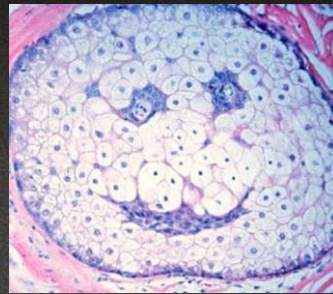
^aTotal number of cases = 166.

^bFIGO, International Federation of Gynaecologists & Obstetricians.



home message:

- FIGO stage не в полной мере отражает агрессивный биологический потенциал и не учитывает гистологический тип злокачественной опухоли.
- non-HPV-related cervical cancer (около 10 %) - агрессивный биологический потенциал, плохой прогноз.
- неороговевающие плоскоклеточные раки (special type), муцинозные и серозные аденокарциномы (p53 mut.) - агрессивный биологический потенциал, плохой прогноз.



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