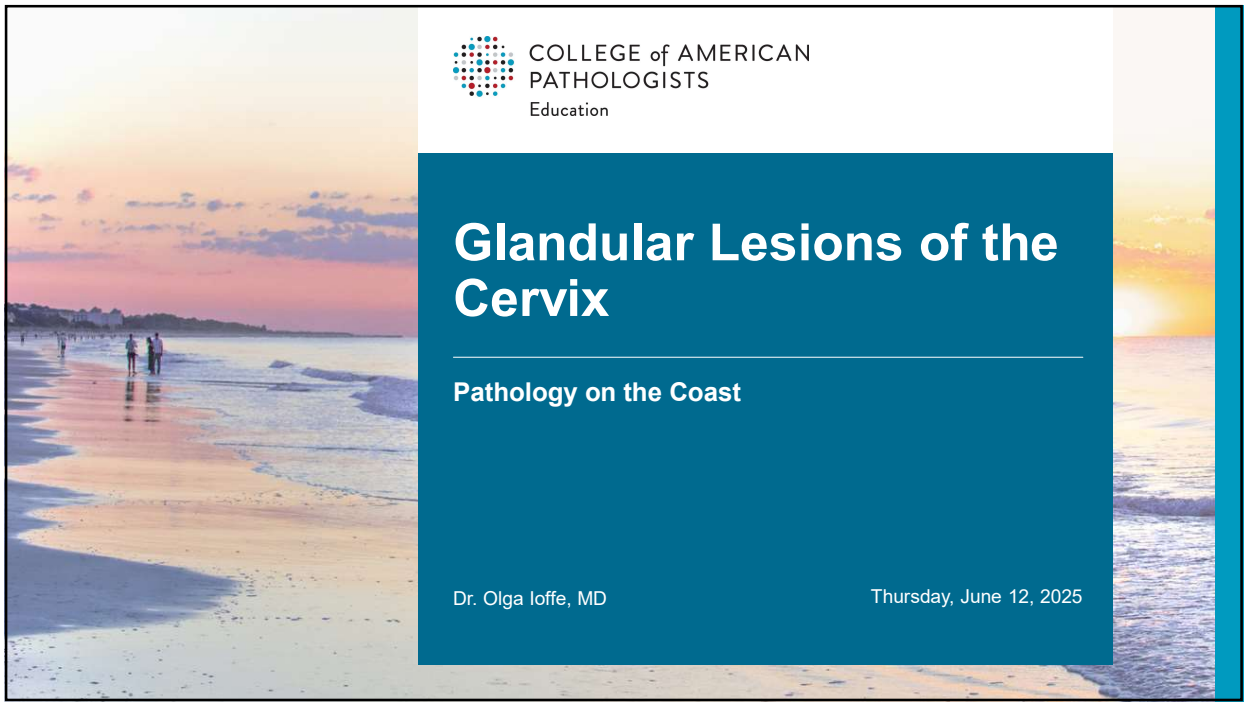


1

Objectives

- **Use diagnostic criteria for the diagnosis of endocervical adenocarcinoma in situ and its distinction from benign glandular lesions and invasive adenocarcinoma.**
- **Recognize morphologic types of HPV-associated and HPV-independent types of adenocarcinoma using the diagnostic algorithm.**
- **Discuss indications and interpretation of ancillary testing in the diagnosis of endocervical glandular lesions.**

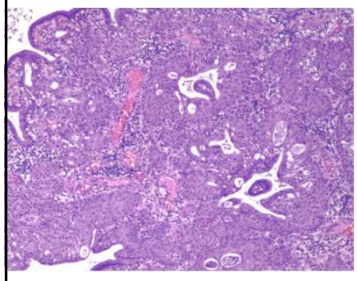
2

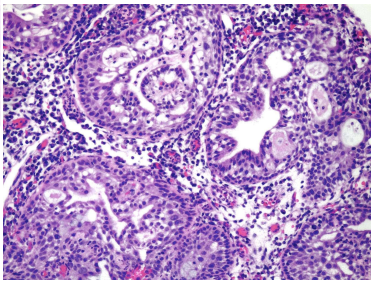
Outline

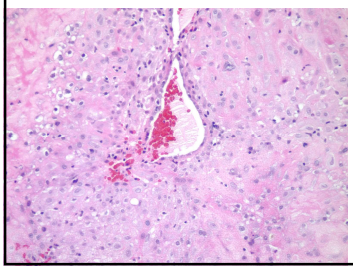
- Benign endocervical lesions
- Adenocarcinoma
  - HPV-associated
    - AIS
    - Invasive adenocarcinoma
  - HPV-independent adenocarcinomas

3

Endocervical Polyp





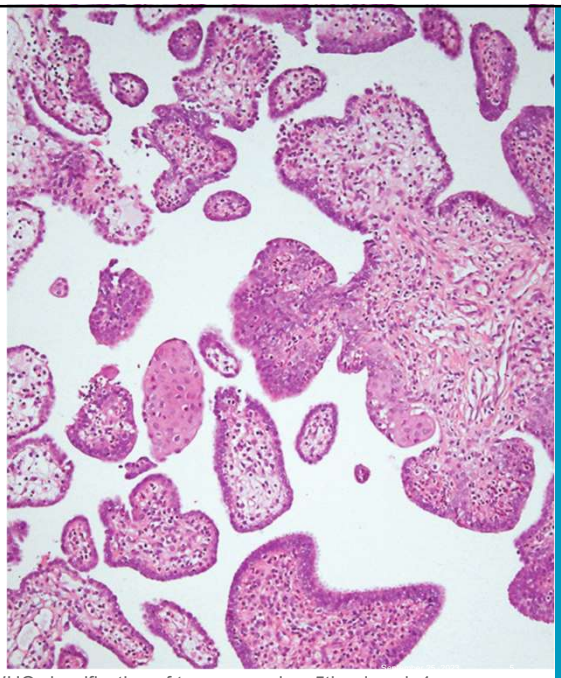


- Usually asymptomatic but may be associated with vaginal bleeding (especially postcoital) and/or discharge
- Fibrovascular tissue lined by benign endocervical-type epithelium, with glands permeating the central stroma
- Common squamous metaplasia or microglandular hyperplasia
- Rarely, surface papillary proliferation or stromal decidual change

4

Mullerian papilloma of the uterine cervix

- Upper vagina or cervix, usually posteriorly
- Associated with vaginal bleeding or discharge
- Almost exclusively in children, typically aged 2–5 years
- Finely branching fibrous papillae lined by a single layer of attenuated, cuboidal to columnar epithelial cells
- Squamous, tubal metaplasia or hobnail-type cells may be present

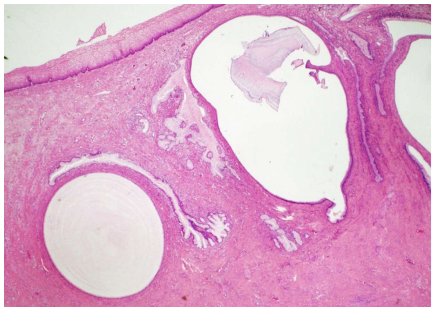


WHO classification of tumours series, 5th ed.; vol. 4

5

Nabothian cysts

- May be deep in the cervical wall – concern for gastric type adenocarcinoma (former minimal deviation adenocarcinoma)
- Debris/histiocytes in ECC may mimic necrosis – and vice versa

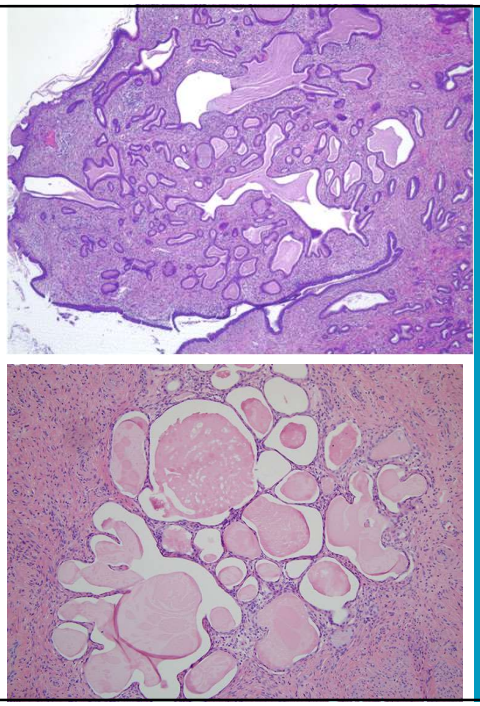


WHO classification of tumours series, 5th ed.; vol. 4

6

Tunnel clusters

- **Tight lobular aggregates of cystically dilated glands**
- **Superficial 1/3 of cervix**
- **10% of women, increases with age, associated with previous pregnancies**
- **Occasionally, deep in the cervical stroma – deep tunnel clusters**



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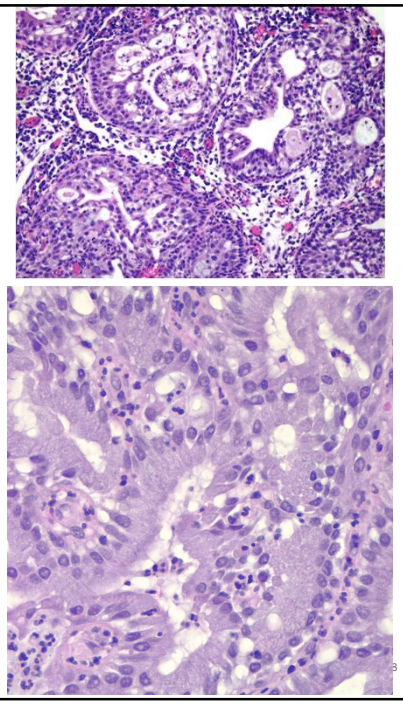
Microglandular hyperplasia

- **Closely packed, small glands lined by mucinous epithelium**
- **Neutrophils**
- **Subnuclear vacuolation and reserve cell hyperplasia**



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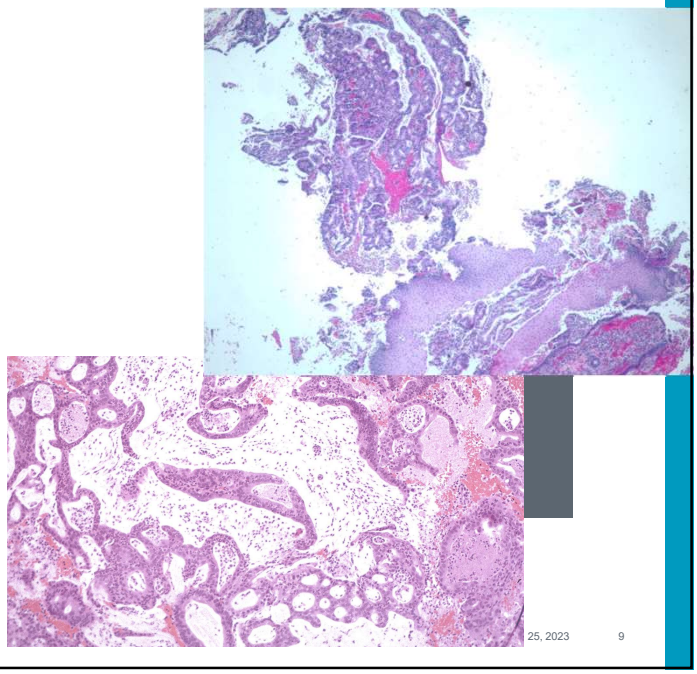
Microglandular pattern in endometrial endometrioid carcinoma

- **May mimic MGH especially if seen in ECC**



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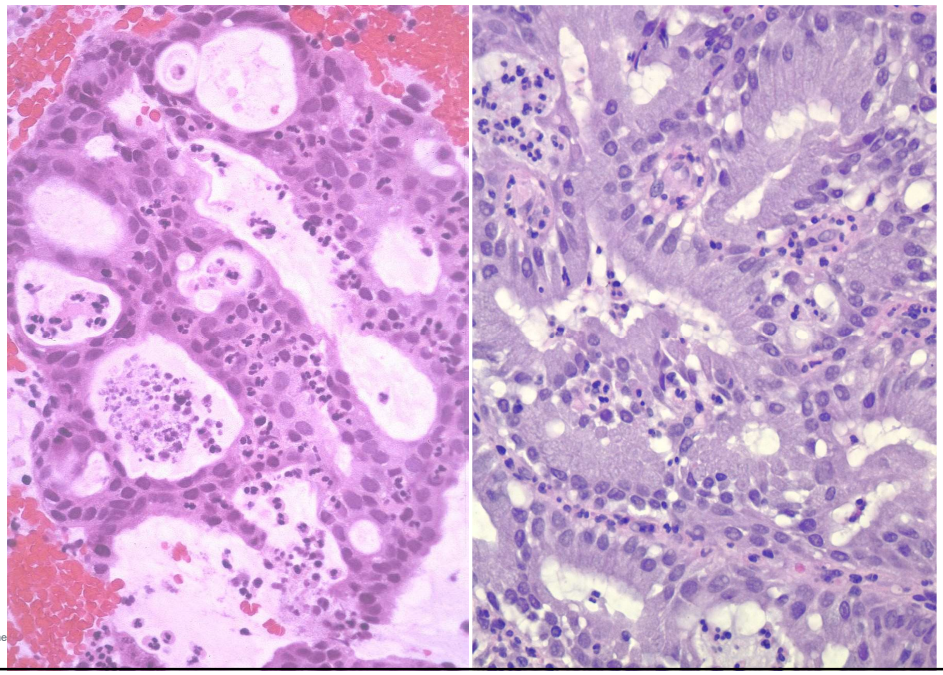

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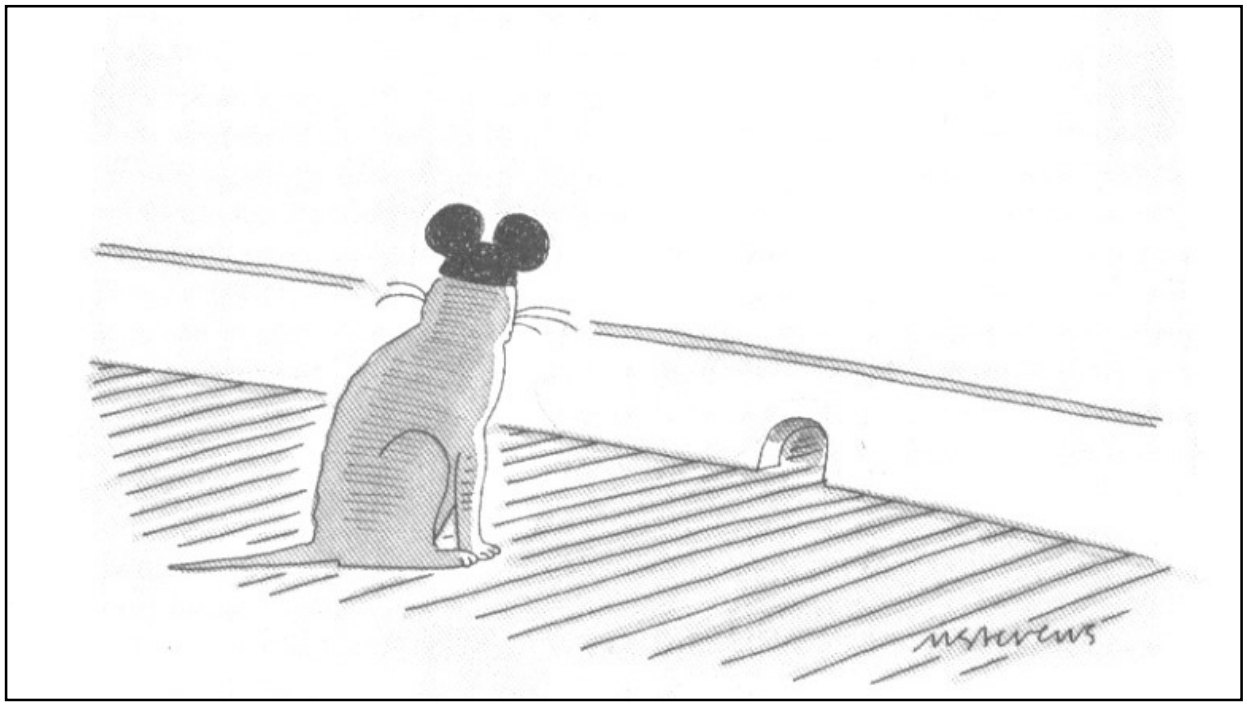
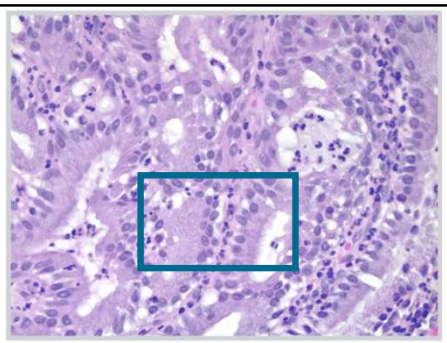


11

Features Favoring Microglandular-like Endometrial Carcinoma over MGH

- **Postmenopausal status**
- **Large amount of biopsy material, large cribriform structures**
- **Endometrial tissue in the same specimen (stroma)**
- **Mitotic figures**
- **Luminal squamous metaplasia vs. basal**
- **"Shared party walls" (cribriforming)**
- **Other patterns of endometrial carcinoma**



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Microglandular hyperplasia

- **May have reactive atypia but no or very rare mitoses**
- **No true cribriforming**
- **No need for IHC but**
  - **Negative vimentin, and CEA (endometrial carcinoma positive)**
  - **Ki-67 proliferation index of <<< 10%**

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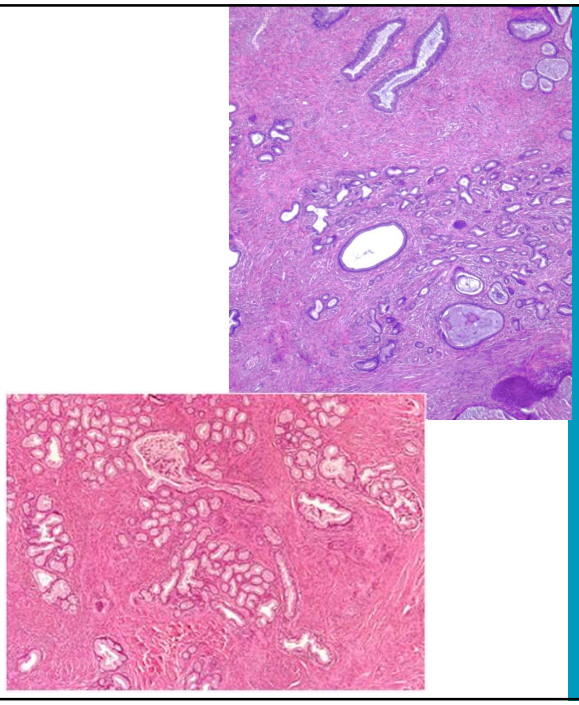
Lobular endocervical glandular hyperplasia

- **Lobular proliferation of small glands around a larger central gland**
- **Inner half of the cervical wall**
- **Lining – benign endocervical epithelium**
- **HK1083 and MUC6 positive**
- **Sporadic or in Peutz–Jeghers syndrome (germline STK11 mutation)**
- **If cytologic atypia - "atypical LEGH", may be precursor of gastric-type adenocarcinoma**



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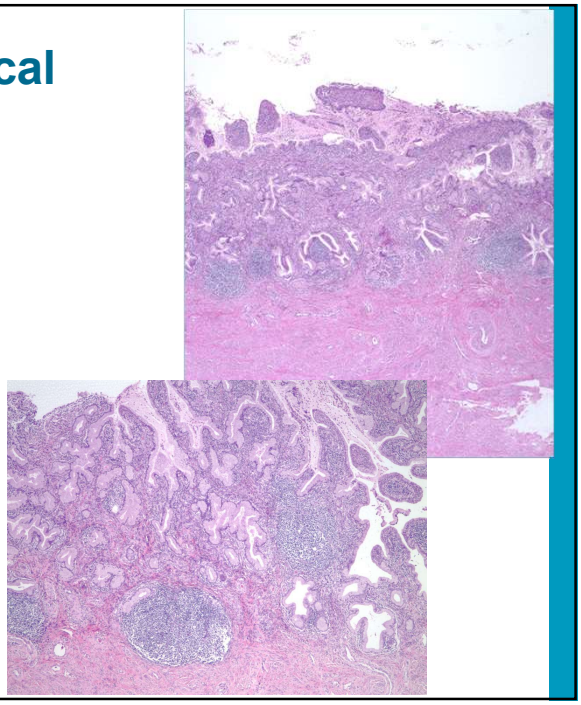
Diffuse laminar endocervical hyperplasia

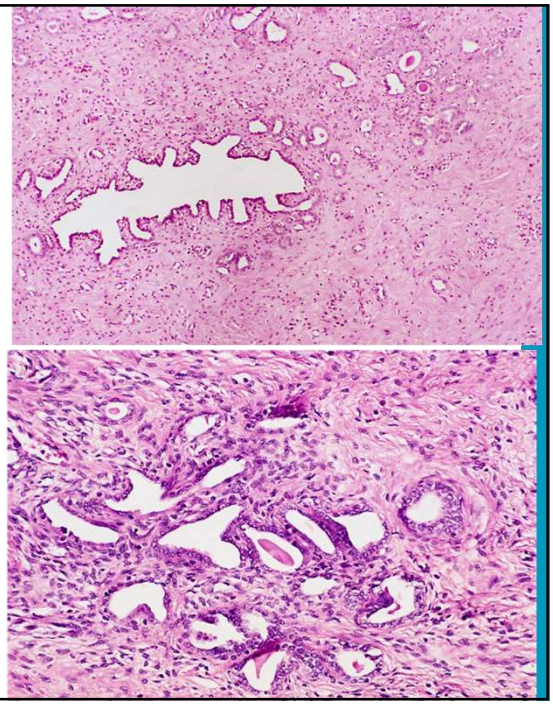
- **Well-demarcated laminar proliferation of endocervical glands**
- **Dense acute and chronic inflammatory infiltrate**
- **No/reactive cytologic atypia, no or rare mitoses**



15

Mesonephric remnants/hyperplasia

- **Embryonic remnants of mesonephric duct**
- **Located deep in the lateral cervical wall**
- **Detected in as many as 20% of cervices**
- **Lobular arrangement, some around long central ducts**
- **Medium-sized tubules**
- **Cuboidal cells with scant eosinophilic cytoplasm**
- **Dense eosinophilic PAS-positive mucin positive secretions**



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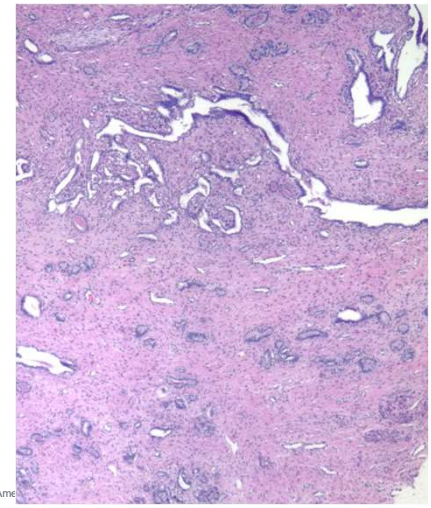
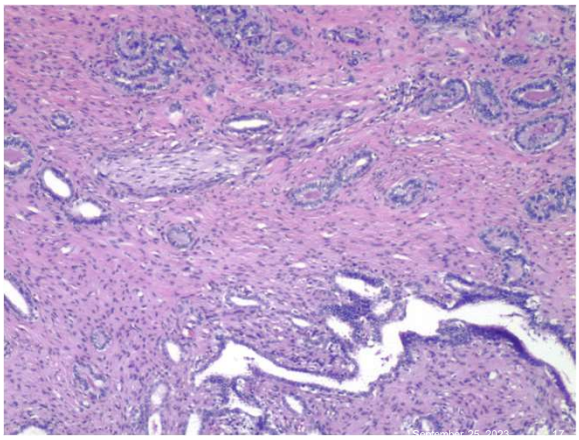
Mesonephric remnant hyperplasia





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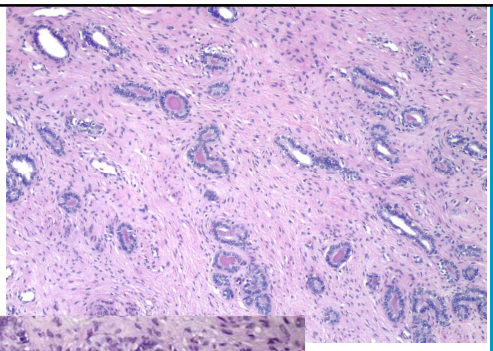
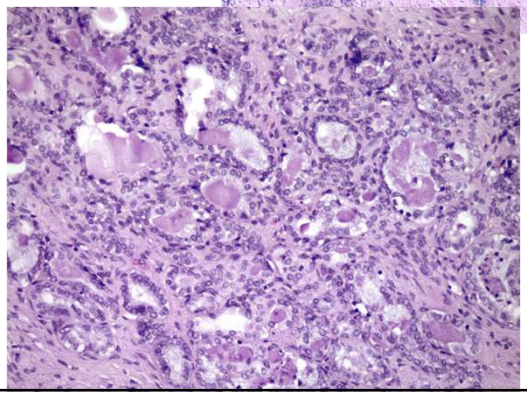
Mesonephric remnant hyperplasia

- **Occasionally can come up to the cervical surface – concern for neoplasia**
- **No atypia/mitoses**
- **CD10, TTF-1, calretinin positive**
- **ER negative**





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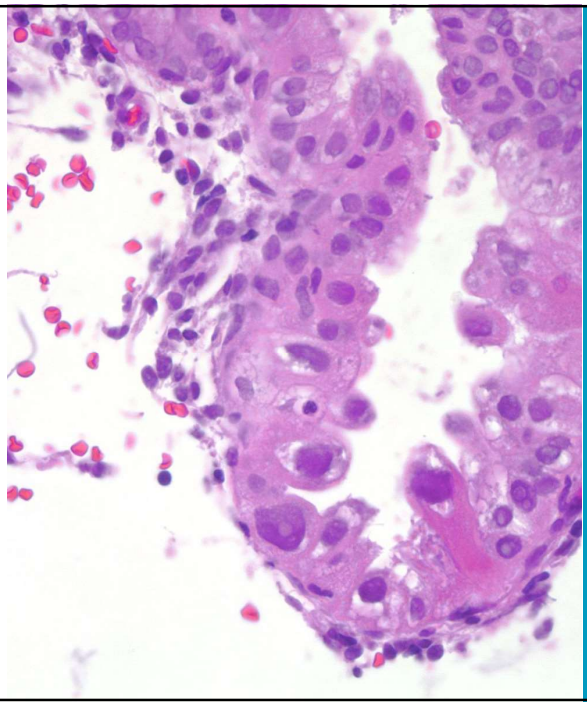
Arias Stella reaction

- **Associated with pregnancy, gestational trophoblastic disease, or high doses of progestins**
- **Enlarged hobnail cells with a maintained N:C ratio**
- **Smudged chromatin, no mitoses, no invasion**
- **Focal change**



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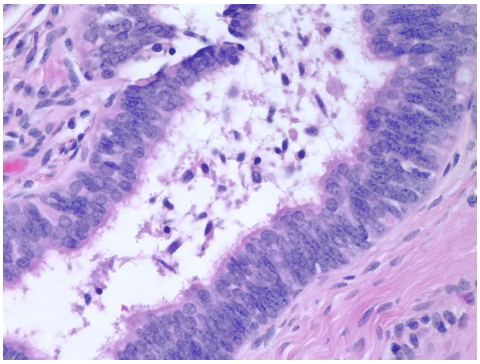
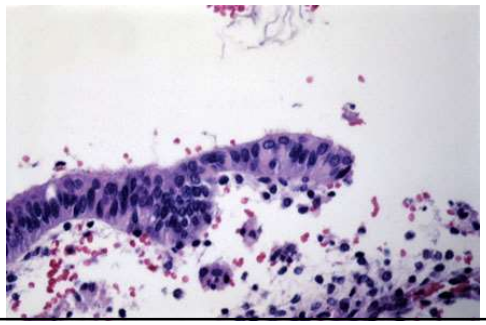
Tuboendometrioid metaplasia

- **Architecturally normal endocervical glands with ciliated cells, pseudostratified non-ciliated cells, and intercalated/peg cells**
- **May be associated with previous instrumentation**





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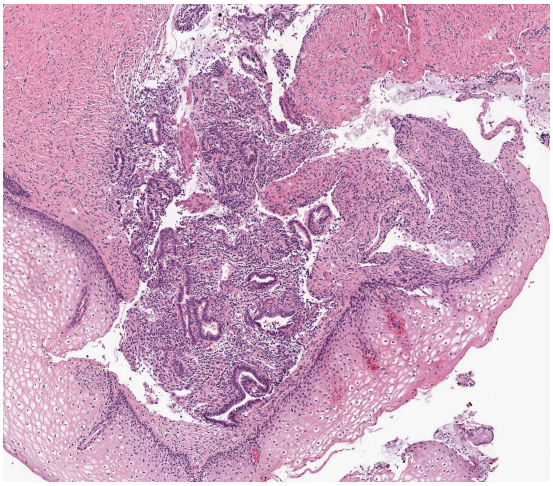
Cervical Endometriosis

- **Both tuboendometrioid metaplasia and endometriosis can mimic AIS**
- **No apical mitoses, apoptosis or atypia**
- **p16 negative (patchy)**
- **ER positive**



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Outline

Benign endocervical lesions

Adenocarcinoma

- **HPV-associated**
  - **AIS**
  - **Invasive adenocarcinoma**
- **HPV-independent**
  - **AIS**
  - **Invasive adenocarcinoma**

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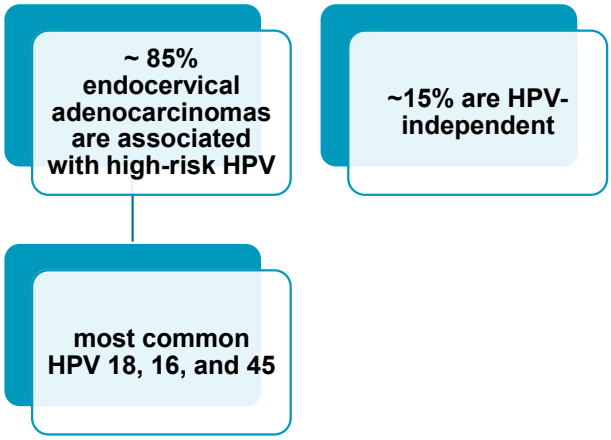
Endocervical adenocarcinoma



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HPV-associated Adenocarcinoma in situ (AIS)

- Precursor of invasive HPV-associated adenocarcinoma
- SEER age-adjusted incidence rate of AIS 0.61 cases per 100,000
- Ratio of AIS to HSIL 1:26 to 1:234
- 1% of all CIS
- Mean patient age is 35 but getting younger

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AIS

- **Located at the transformation zone, may spread up to 3 cm upward in the endocervical canal - both the surface and underlying glands**
- **Discontinuous spread**

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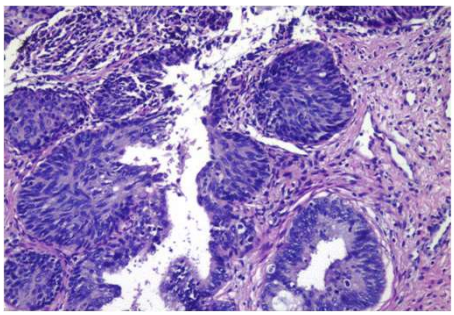
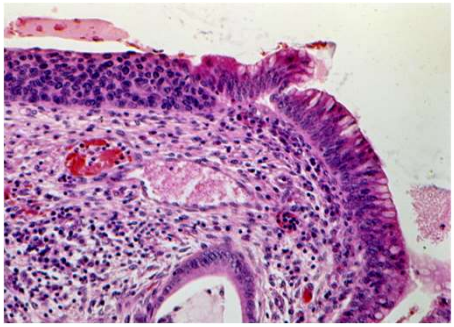
HPV-associated Adenocarcinoma in situ (AIS)

- **50% or more of cases coexist with SIL**
- **HPV (18>16)**





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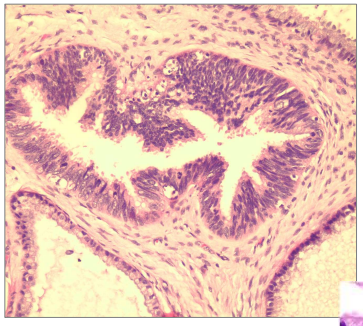
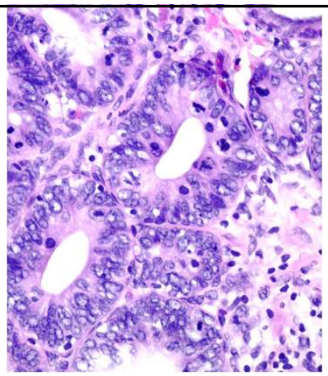
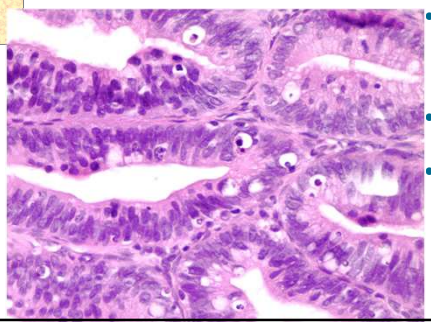
AIS







- Marked cytologic atypia in architecturally normal glands
- Nuclear hyperchromasia, enlargement, pseudostratification and crowding
- Mitotic activity: apical/"floating" mitoses
- Frequent apoptosis
- Usually mucin depletion

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Mucin depletion

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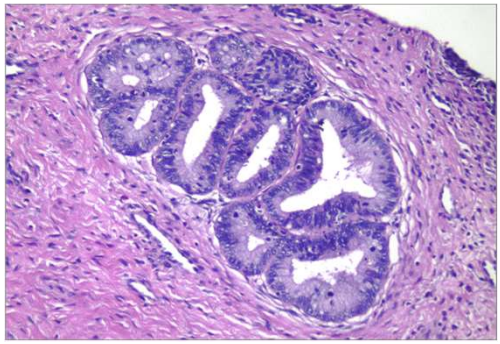
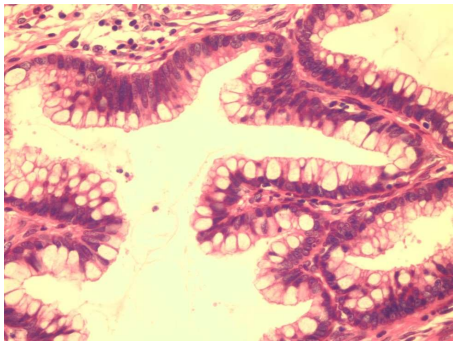
AIS





- **Obvious intracytoplasmic mucin with or without goblet cells**
- **Goblet cells are not diagnostic of AIS without other features**

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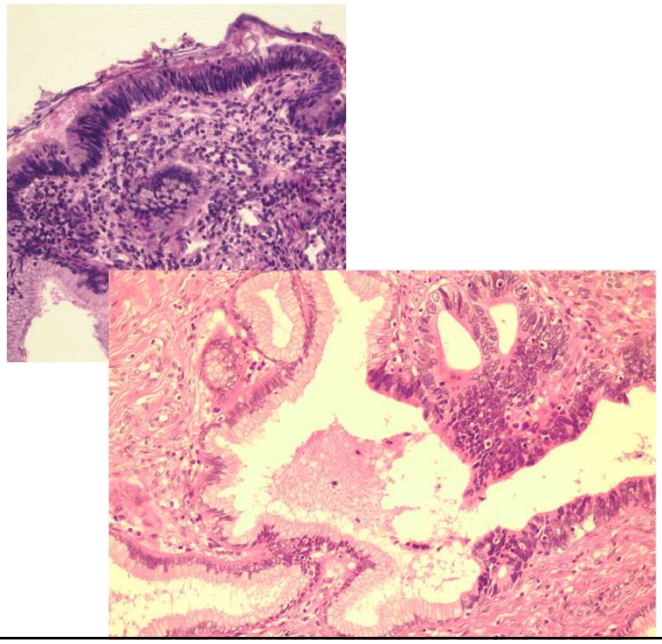
AIS

- **Surface or underlying glands**
- **Abrupt transition between normal and AIS**



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AIS Scoring System

- **Based on mitoses/apoptoses, pseudostratification and nuclear atypia**
- **Each scored 0-3**
- **Average per gland in two most active/atypical glands**
- **Total score**
  - **0-3: benign**
  - **4-5: uncertain atypia (reactive or dysplasia)**
  - **>6: AIS**

O. Ioffe, …S.G. Silverberg
AJSP, 2000

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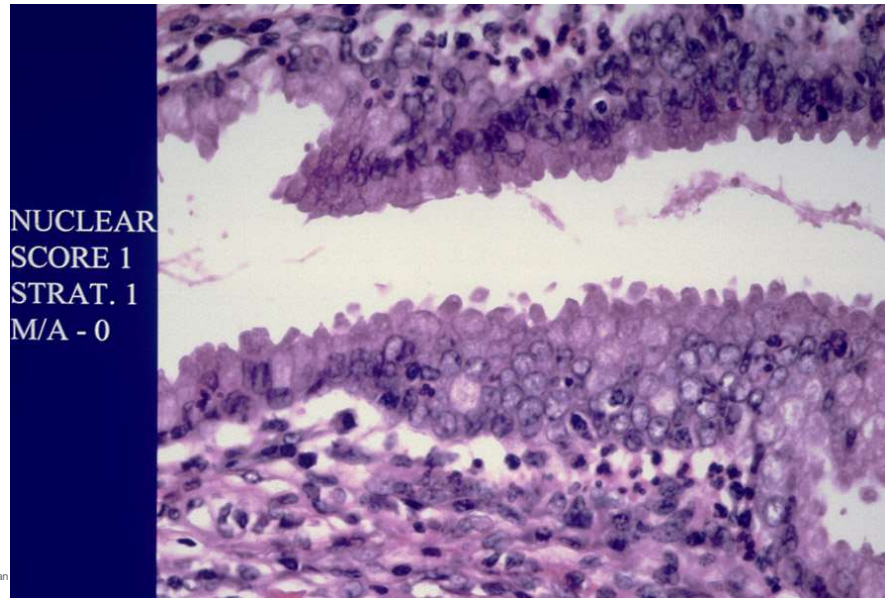
AIS Scoring System: Score = 2



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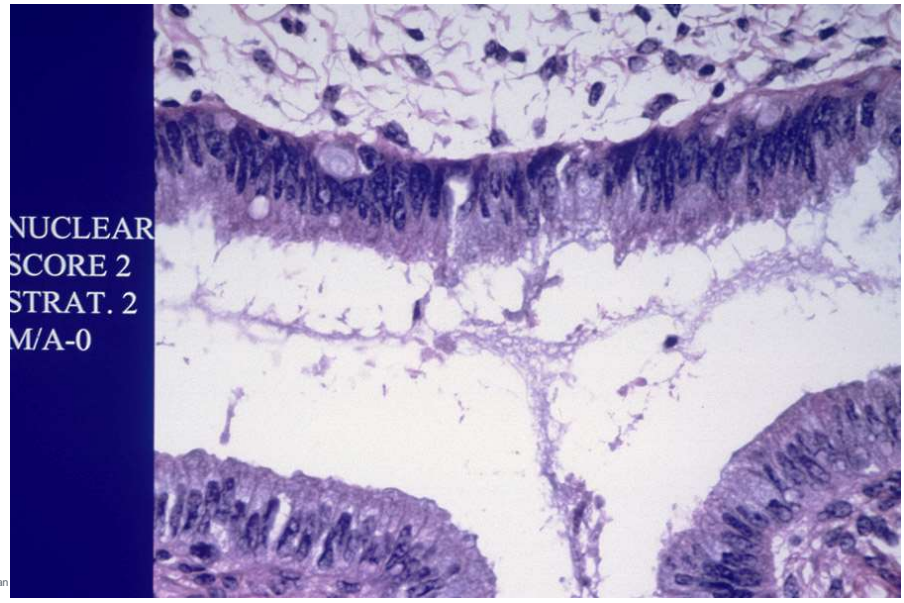
AIS Scoring System: Score = 4



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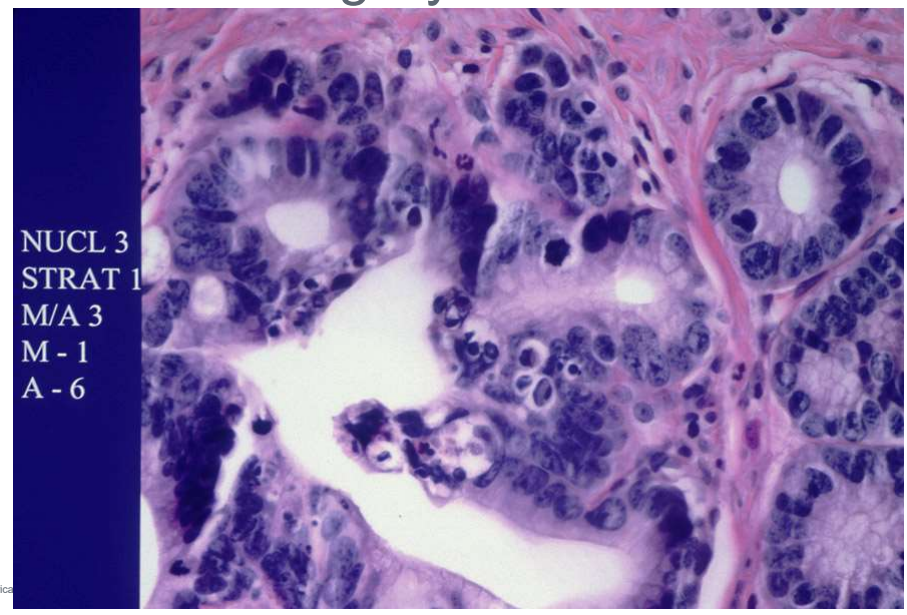
AIS Scoring System: Score = 7



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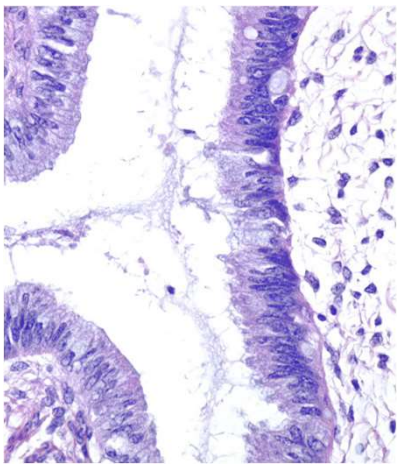
Endocervical glandular atypia/dysplasia



- Some but not all features of AIS; not associated with inflammation
- Has been proposed to represent a precursor lesion of AIS on the basis of reports demonstrating it adjacent to AIS or invasive adenocarcinoma
- Overlap with reactive conditions
- Probably a mixed bag – some have HPV DNA

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"This symposium has gotten completely out of hand!"

36

Endocervical glandular dysplasia

Not reproducible

No agreement on diagnostic features, significance and management

Not recommended for use in clinical practice

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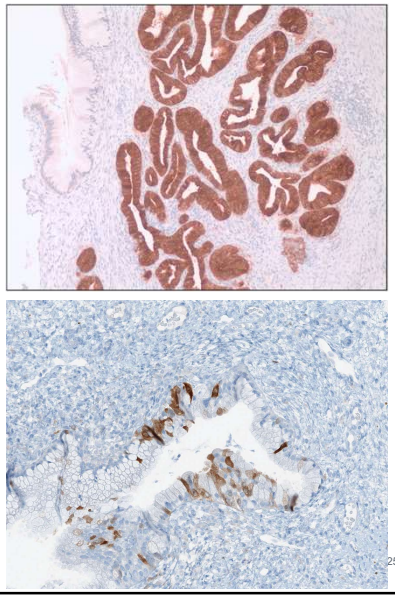
Ancillary studies: p16(INK4a)

- **Strong block-type staining**
- **Not required - morphology is usually sufficient**
- **Use in hyperplastic, reactive, and metaplastic lesions - patchy (negative) p16 staining**



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Ancillary studies

- **HR-HPV RNA in situ hybridization in difficult cases**
- **ER/PR negative (vs. tuboendometrioid metaplasia, endometrioid carcinoma)**

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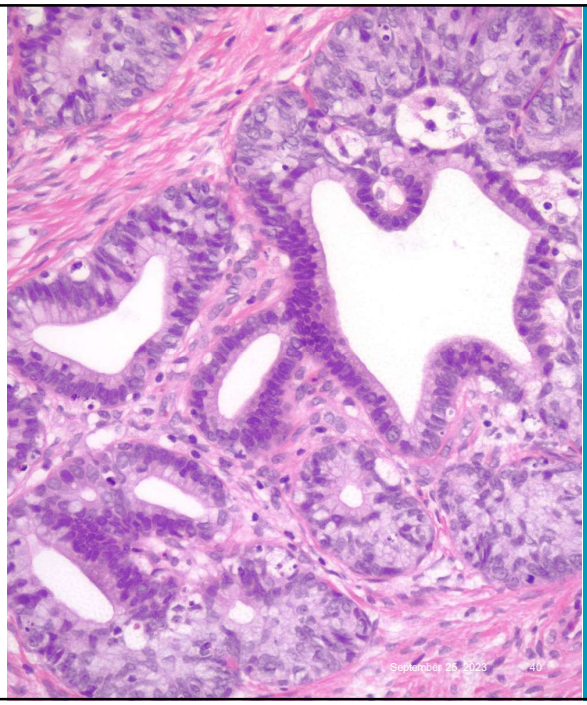
Stratified mucin-producing intraepithelial lesion (SMILE) – subtype of AIS

- **Stratified epithelium with mucin throughout all cell layers and (frequently) a peripheral cuff of basaloid/reserve cells**
- **Resembles HSIL involving endocervical glands**



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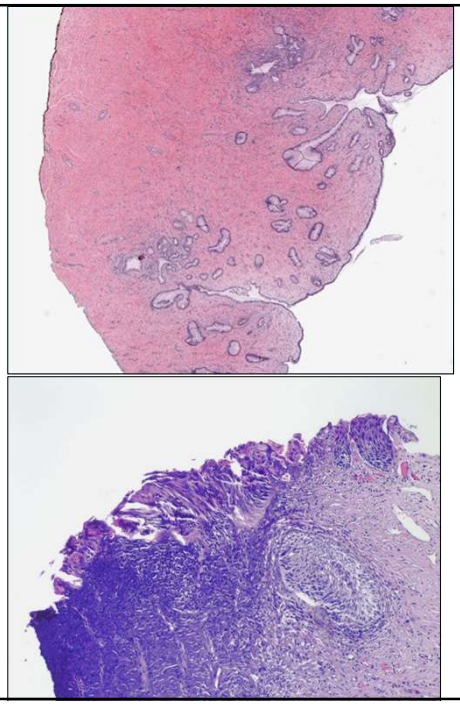
Diagnosis and Treatment of AIS

- If AGC/AIS on Pap – colpo with bx/ECC
- Cold knife conization for better assessment of surgical margins and to rule out invasion
  - Positive margins in 75% of LEEP, 24% CKC



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Management of AIS

- Conization with negative margins and close follow up – ECC, Pap and HPV testing
- Hysterectomy is the definitive treatment, even with negative cone margins
- In patient desiring preservation of fertility, cold cone then close follow-up with ECC, then hysterectomy after childbearing
- Recurrence in 14% of cases with negative margins (Etherington, 2001)

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Outline

Benign endocervical lesions

Adenocarcinoma

- HPV-associated
  - AIS
  - Invasive adenocarcinoma
- HPV-independent adenocarcinomas

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Pattern-based classification (Silva system) of HPV-associated endocervical adenocarcinoma – strongly associated with LN mets and survival

- **Pattern A (non-destructive invasion)**
  - **Well-demarcated glands with rounded contours**
  - **No lymphovascular invasion**
  - **Complex intraglandular growth acceptable (i.e. cribriform growth, papillae)**
  - **Lack of solid growth**

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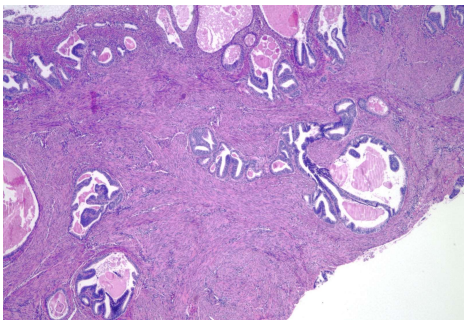
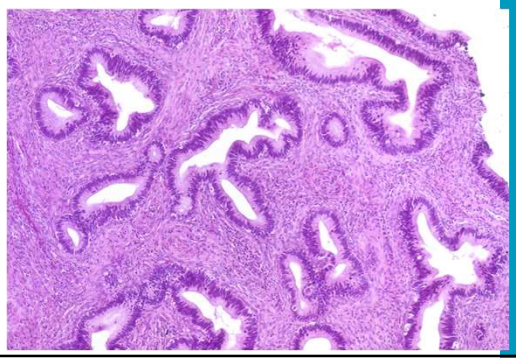
- **Distinction between AIS and pattern A is difficult and poorly reproducible**
- **May report concern for invasion and the deepest point of possible invasion**





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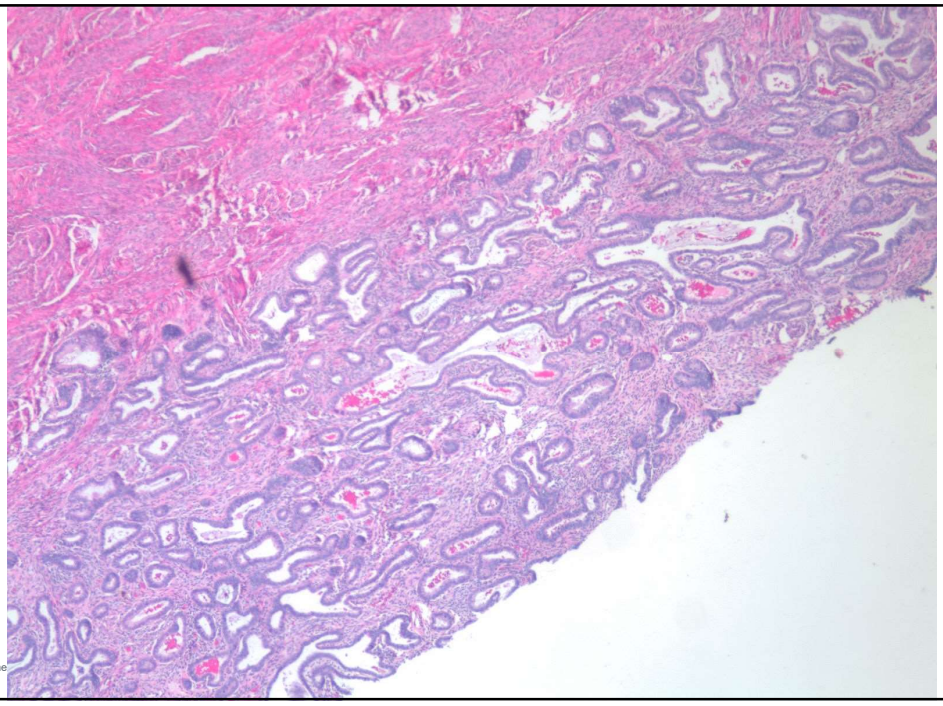

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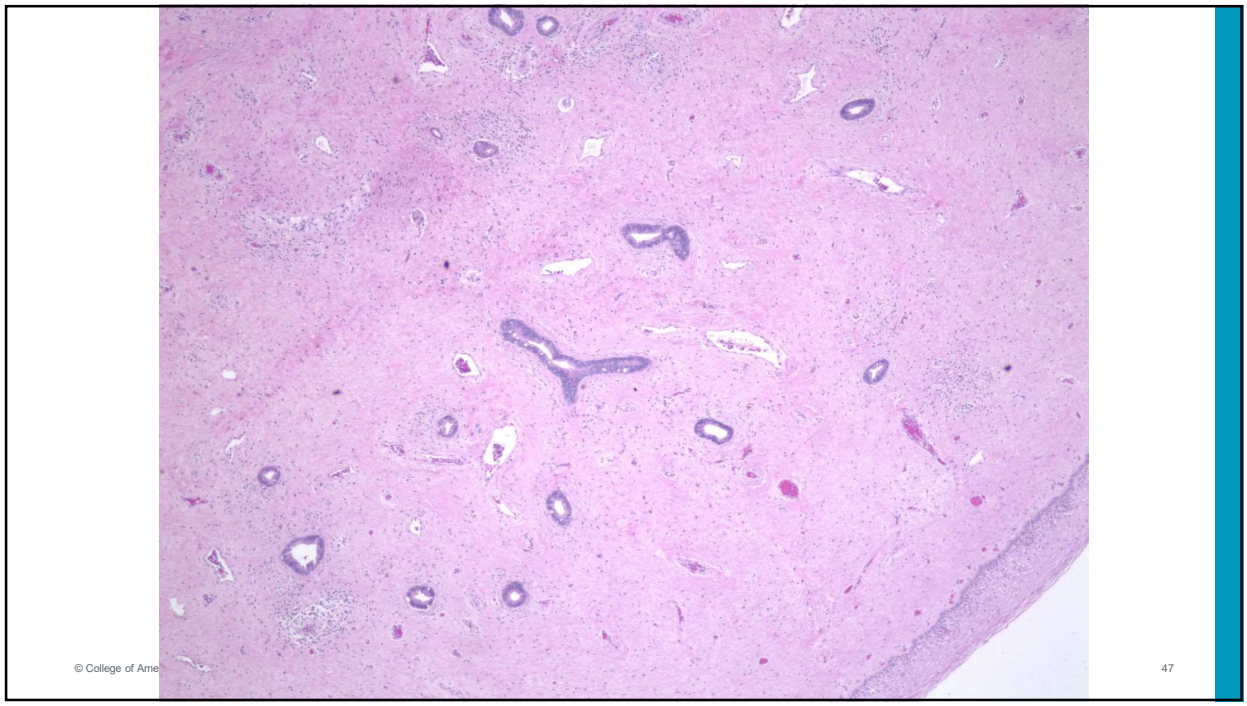

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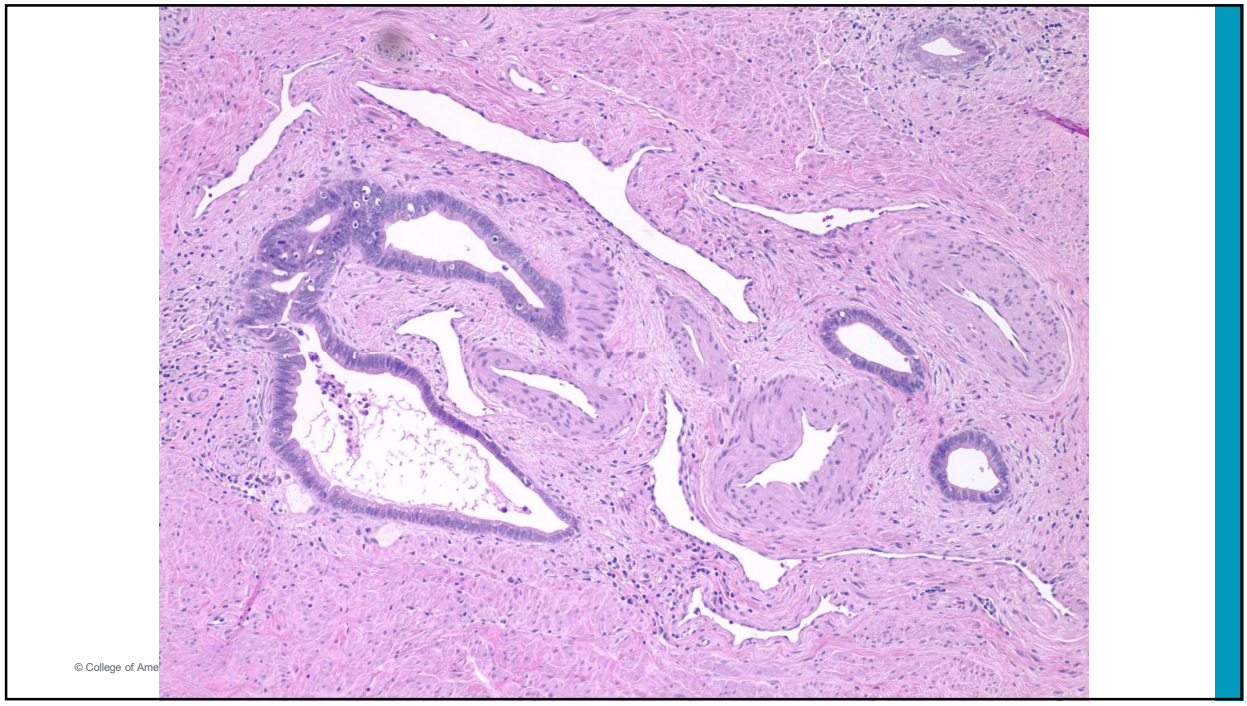

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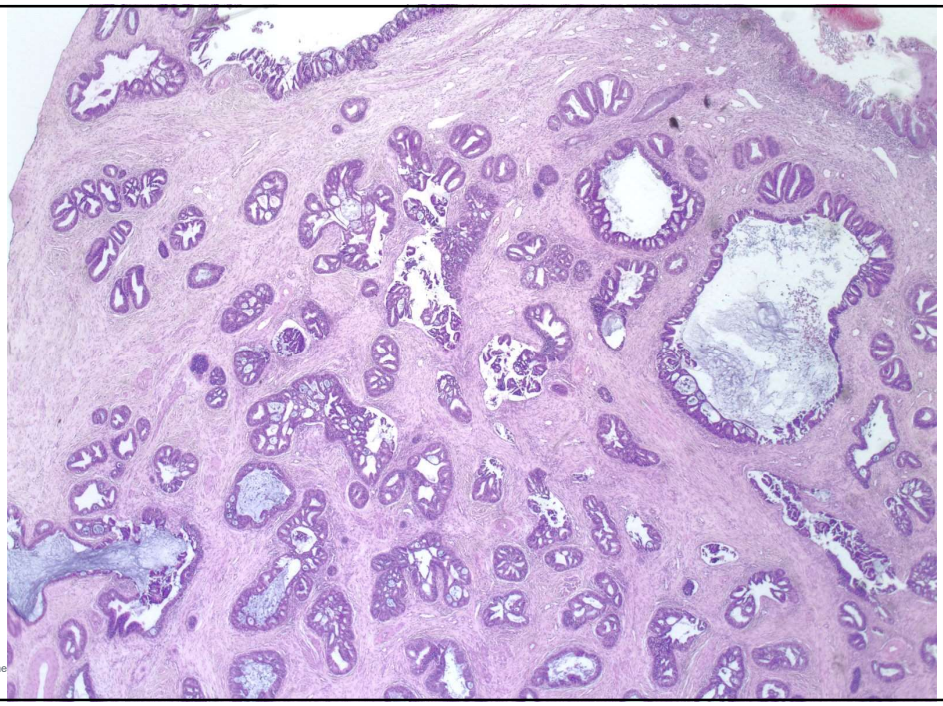

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Pattern-based classification (Silva system) of HPV-associated endocervical adenocarcinoma

- **Pattern B (early / focally destructive invasion)**
  - **Individual or small groups of tumor cells, separated from the rounded glands; focally desmoplastic or inflamed stroma**
  - **Foci may be single, multiple, or linear at base of tumor**
  - **Lymphovascular invasion +/−**
  - **Lack of solid growth (i.e. architecturally well to moderately differentiated)**

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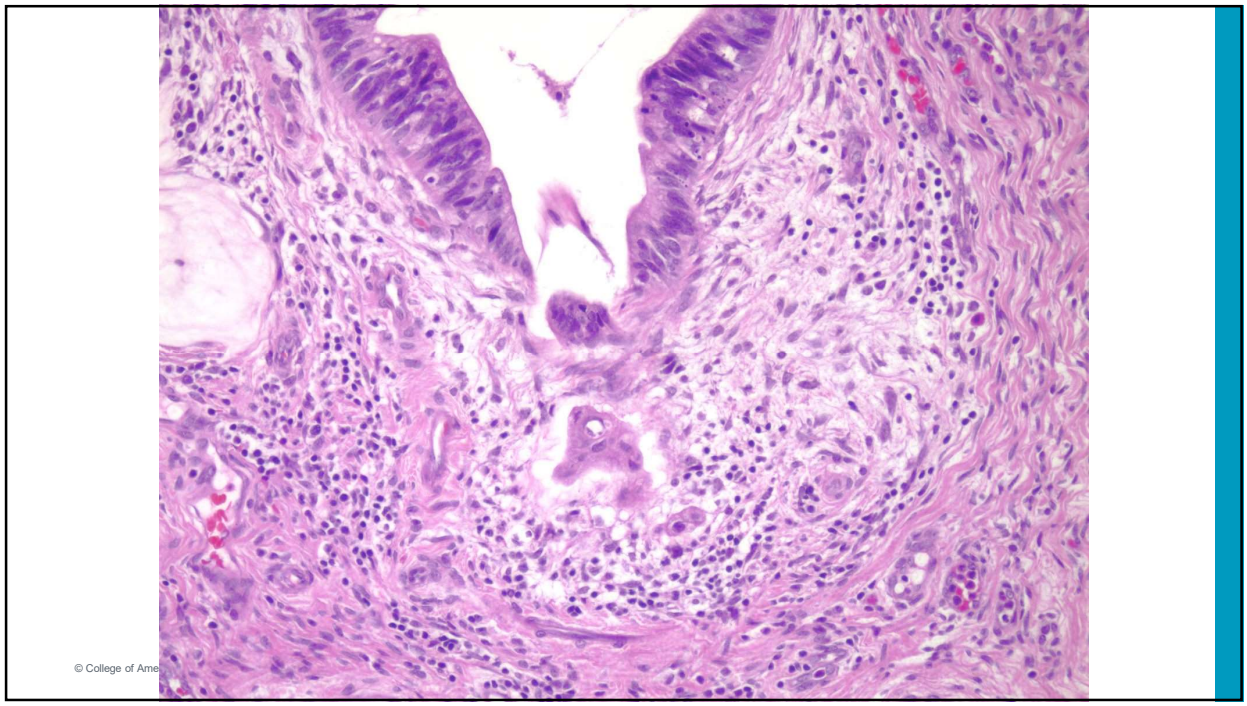

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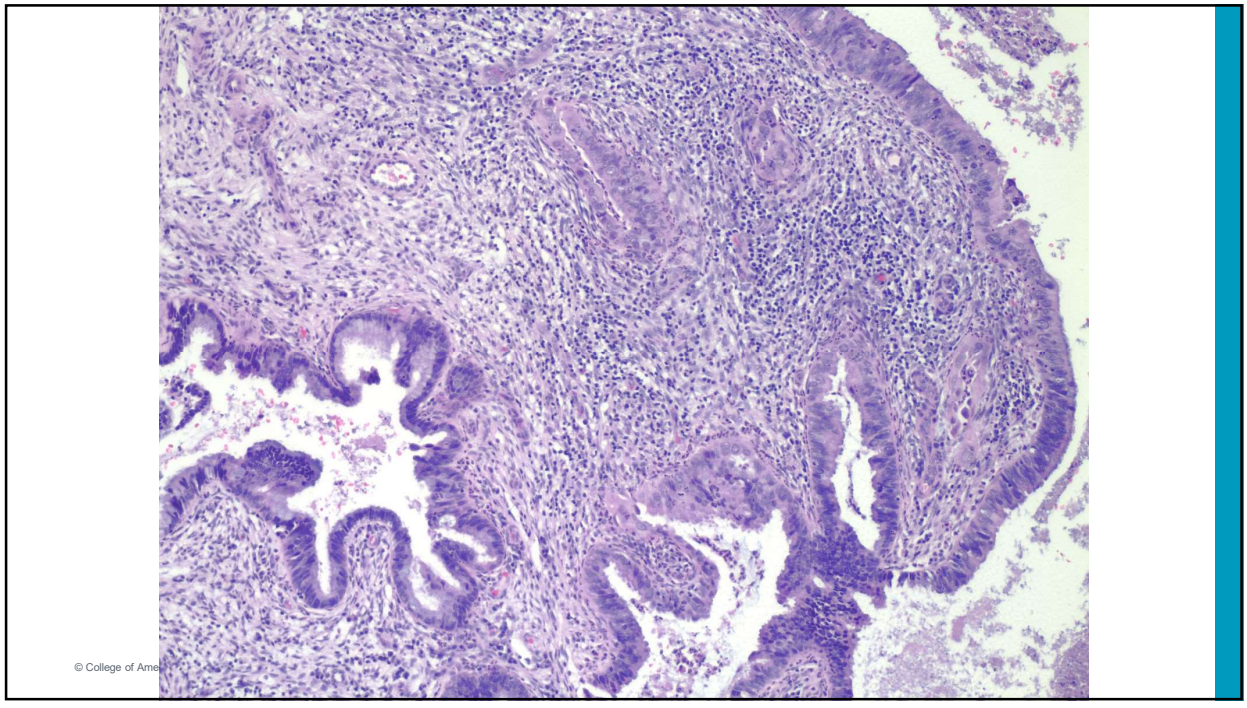

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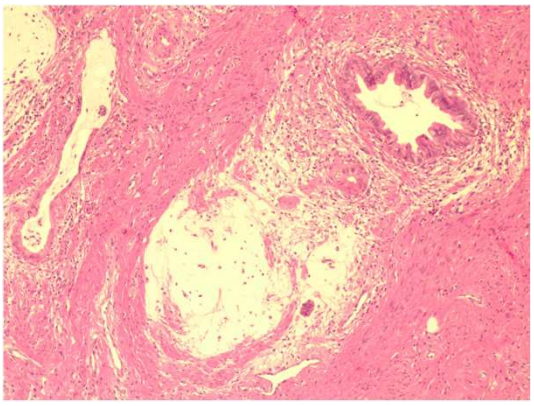
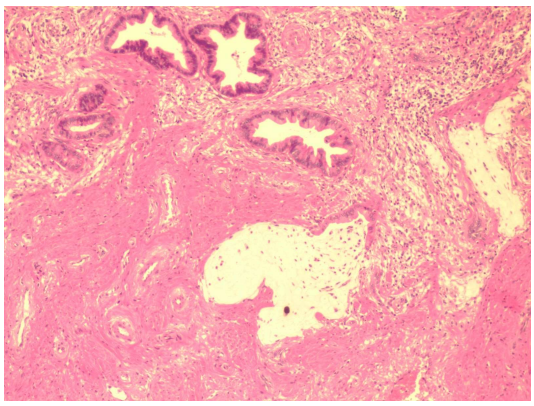
Pattern-based classification (Silva system) of HPV-associated endocervical adenocarcinoma

- **Pattern C (diffusely destructive invasion)**
  - **Diffusely infiltrative glands with associated extensive desmoplastic response**
  - **Glands often angulated or with canalicular pattern, with some open/incomplete glands**
  - **Confluent growth filling a 4× field (5 mm): glands, papillae (stroma only within papillae), or mucin lakes**
  - **Solid, poorly differentiated component (architecturally high-grade); nuclear grade is disregarded**
  - **Lymphovascular invasion +/−**

53

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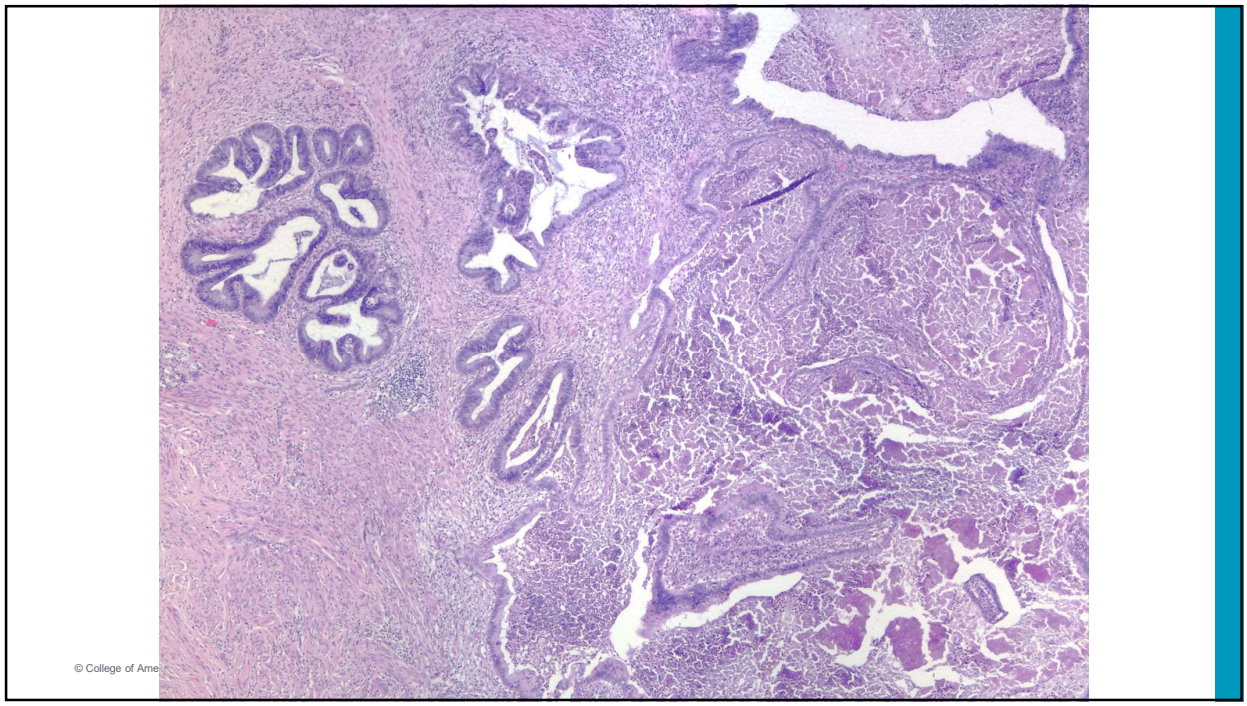

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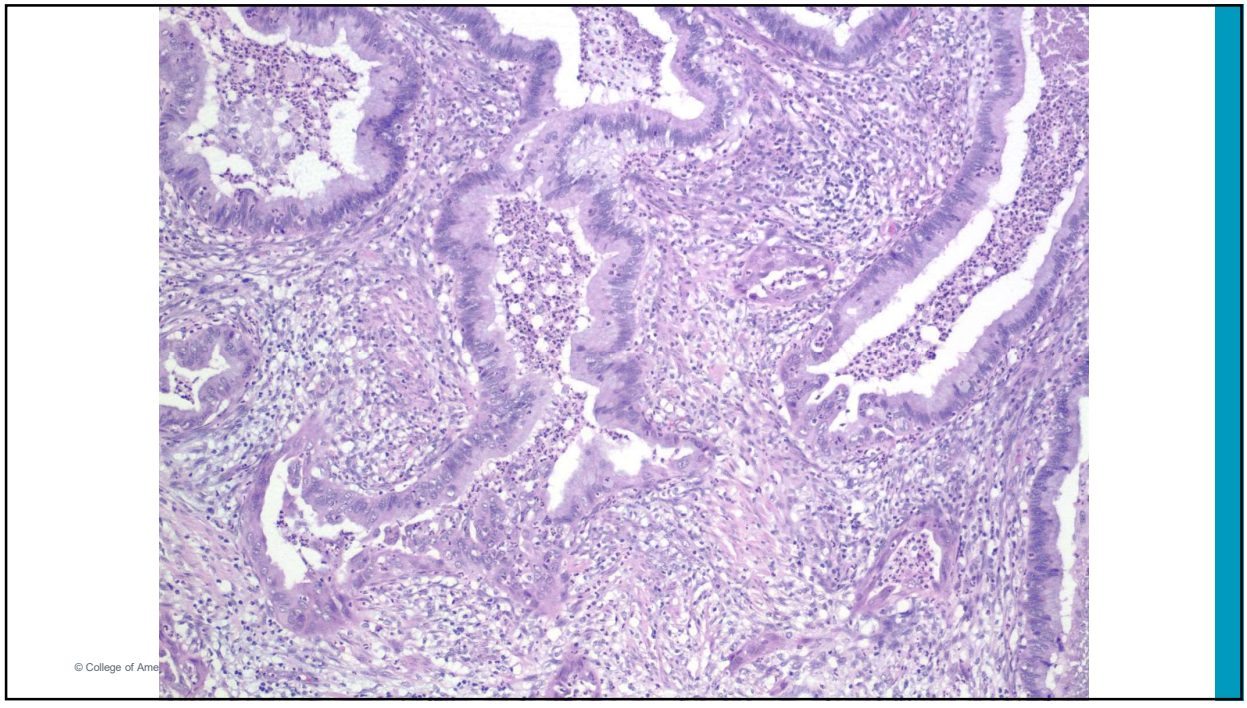

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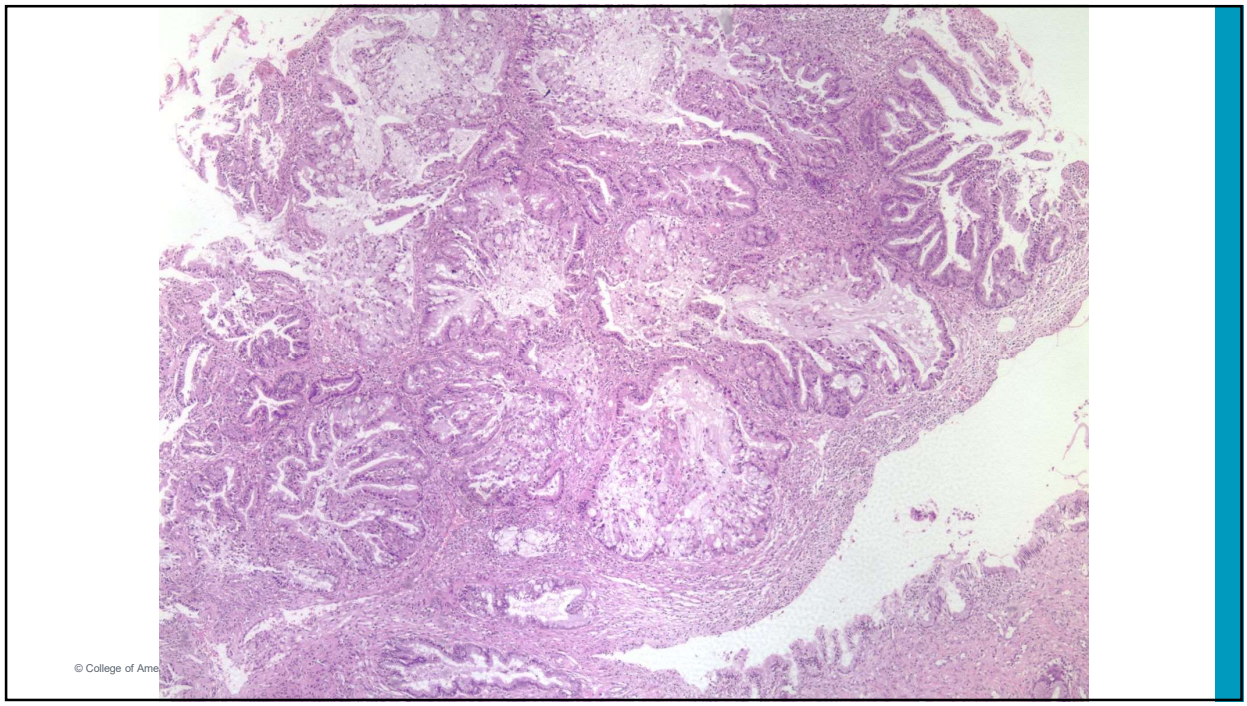

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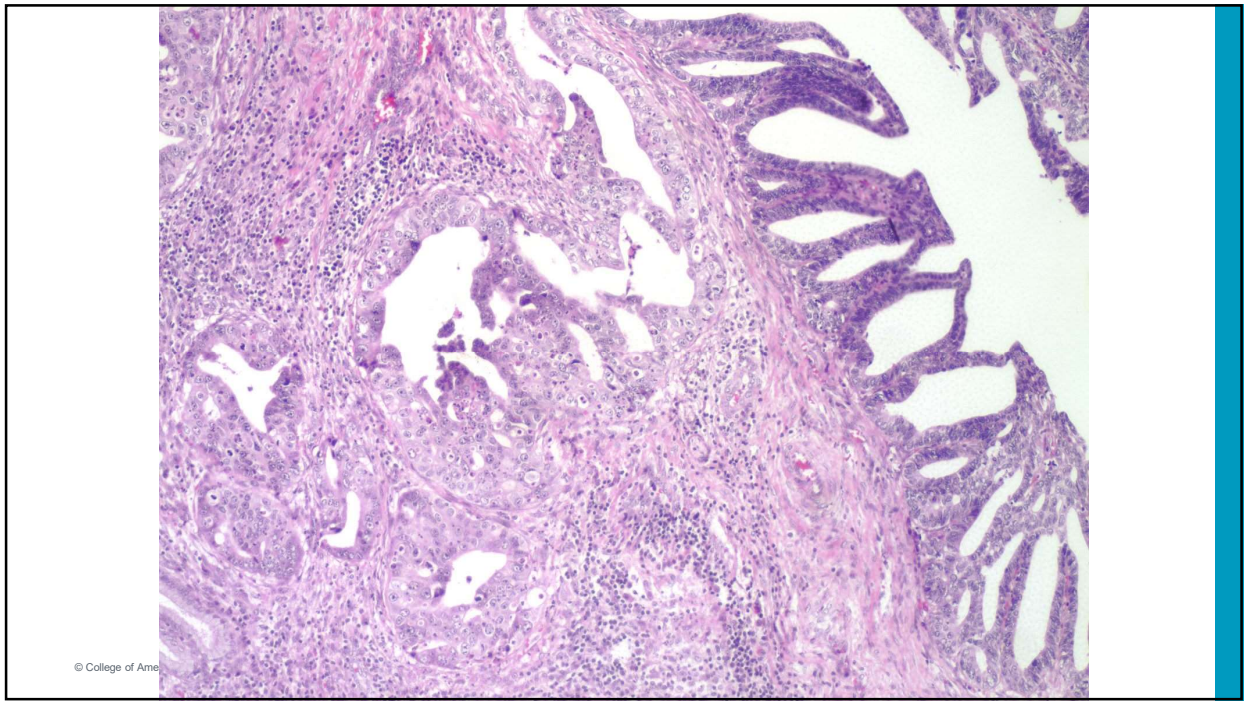

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Histologic types of HPV-associated adenocarcinoma

- **Usual type (less than 50% mucinous)**
  - **Villoglandular variant**
- **Mucinous type (>50% mucinous)**
  - **Mucinous NOS (endocervical)**
  - **Intestinal (>50% goblet cells, may have neuroendocrine differentiation)**
  - **Signet-ring cell (>50% of tumor)**
  - **Stratified mucin-producing**

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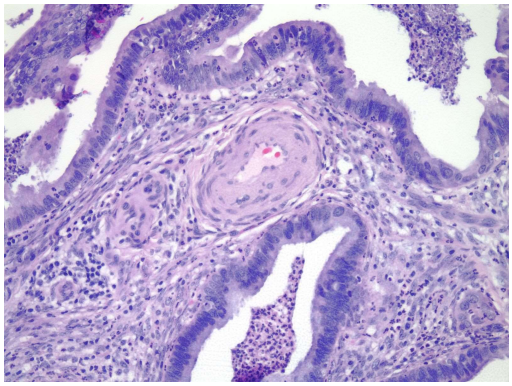
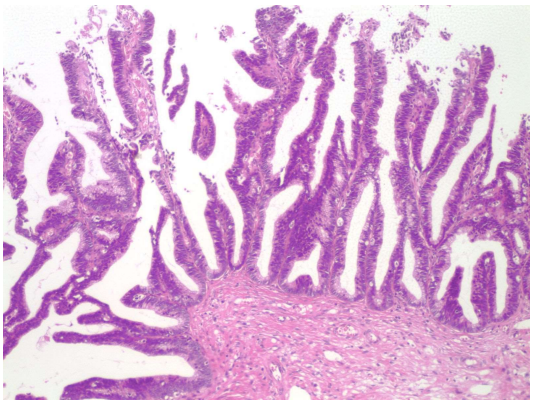


Usual type



Villoglandular

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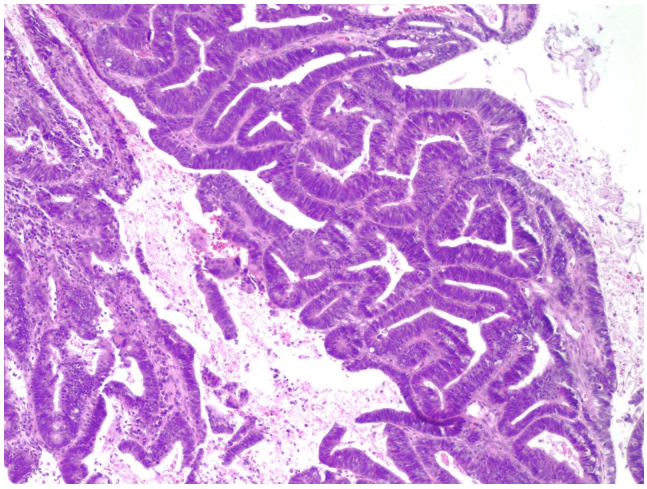
Usual type with mucin depletion: mimics endometrioid carcinoma



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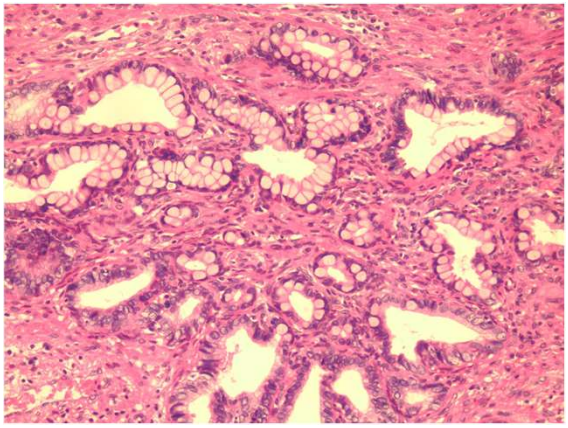
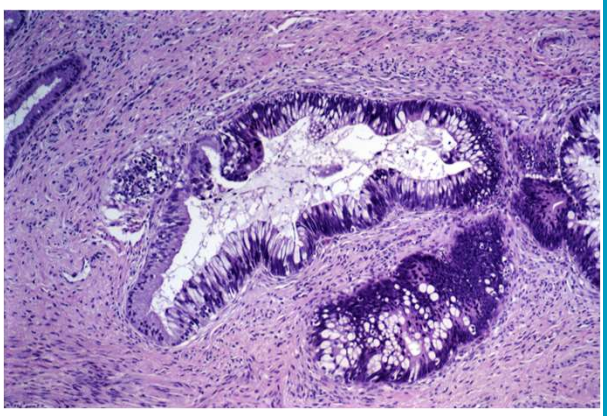
Mucinous intestinal type





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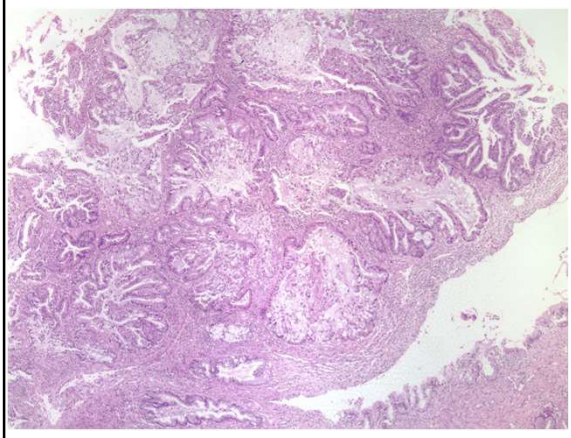
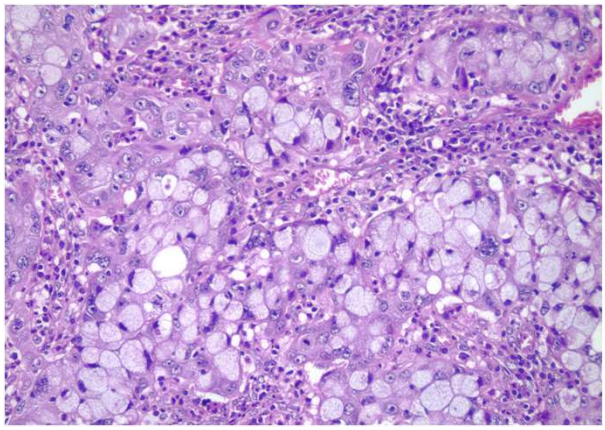
Mucinous type



Mucinous NOS (endocervical)



Signet ring cell type

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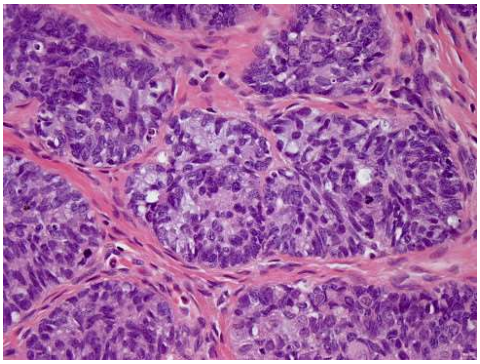
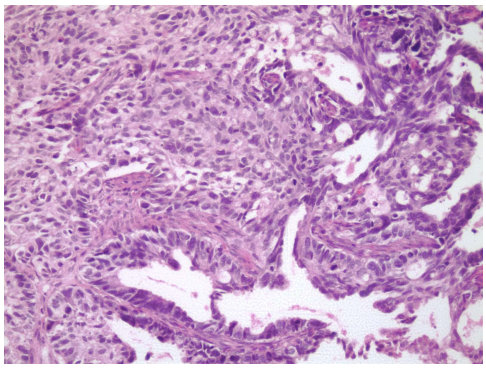
Invasive stratified mucin-producing carcinoma

- **Nested pattern with peripheral palisading**



- **Lumen formation**



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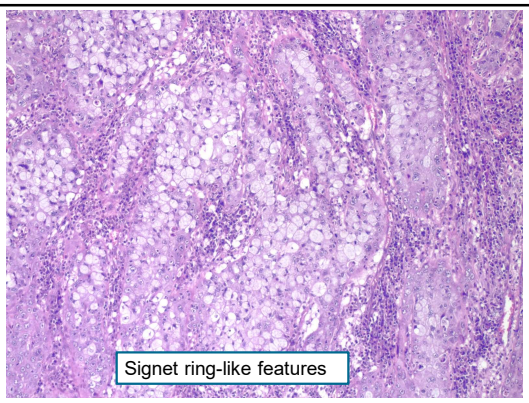
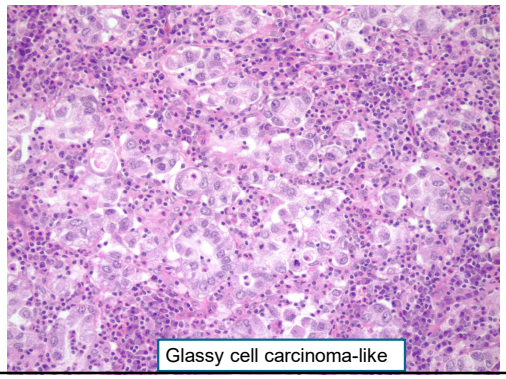


Invasive stratified mucin-producing carcinoma



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IHC in HPV-associated adenocarcinoma

Positive	Negative	p53
p16(block-like) and/or HPV but fixation-dependent CK7 PAX8 (except SMILE) p40 in 30% SMILE	ER, PR – 70-80% Vimentin MUC6 HNF1b Napsin A CK20	Usually wild-type

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Outline

- Benign endocervical lesions
- Adenocarcinoma
  - HPV-associated
    - AIS
    - Invasive adenocarcinoma
  - HPV-independent adenocarcinomas

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HPV-independent adenocarcinomas

- Gastric type
- Mesonephric
- Clear cell
- Endometrioid

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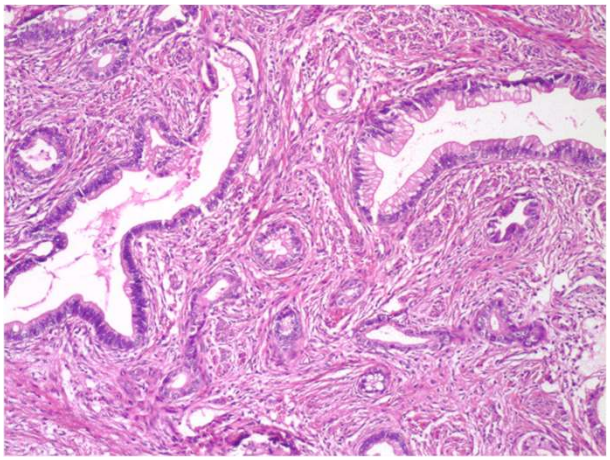
Gastric type adenocarcinoma



- Morphology more like pancreatic/biliary adenocarcinoma
- Abundant cytoplasm, prominent cell borders/plant-like

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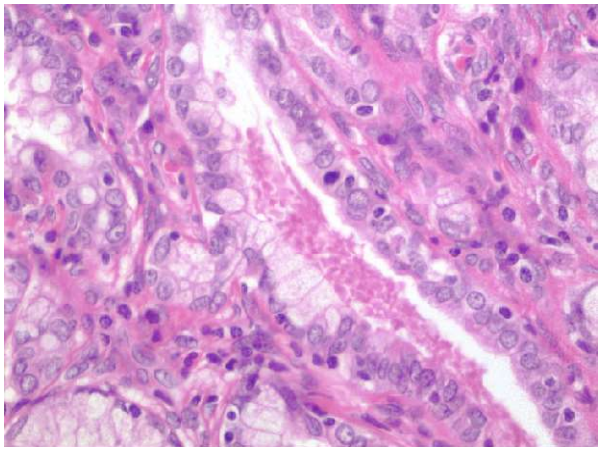
Gastric type adenocarcinoma



- Named gastric due to expression of gastric markers such as HIK1083 and MUC6
- neutral-type mucin (pinkish-red on Alcian blue / PAS)
- Can have goblet cells

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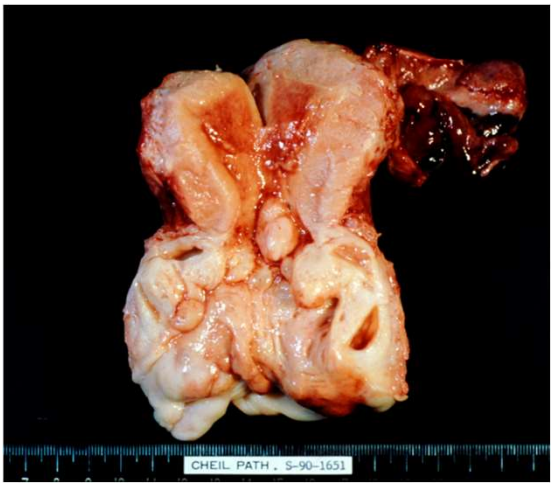
Well differentiated gastric type adenocarcinoma: formerly known as minimal deviation adenocarcinoma ("adenoma malignum") of the cervix

- **Abundant watery/mucoid discharge, abnormal bleeding**
- **May be part of Peutz-Jeghers syndrome but most are sporadic**
  - **LKB (SKT11) mutations may be seen in patients without the syndrome**



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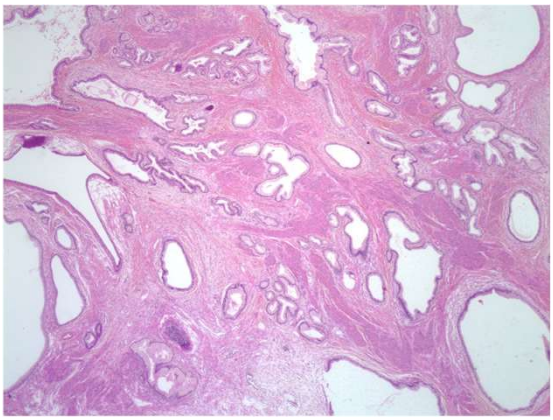
Well differentiated gastric type adenocarcinoma: formerly known as minimal deviation adenocarcinoma ("adenoma malignum") of the cervix

- **Architecturally atypical but cytologically benign glands (pincushion, claw, crab, cystic etc)**
- **Increased number of glands, haphazard**
- **Glands in abnormal locations**
  - **Deep (>5 mm)**
  - **Adjacent to arteries**
  - **Infiltrating between benign glands and stroma**



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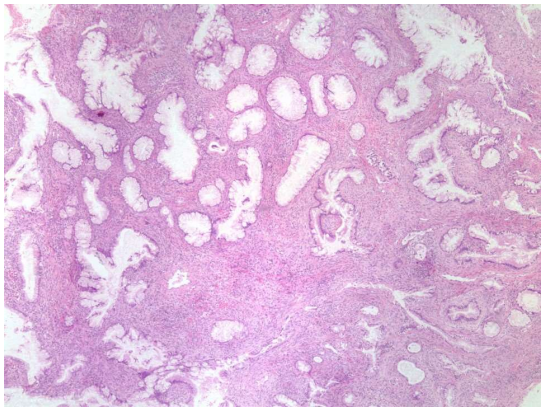
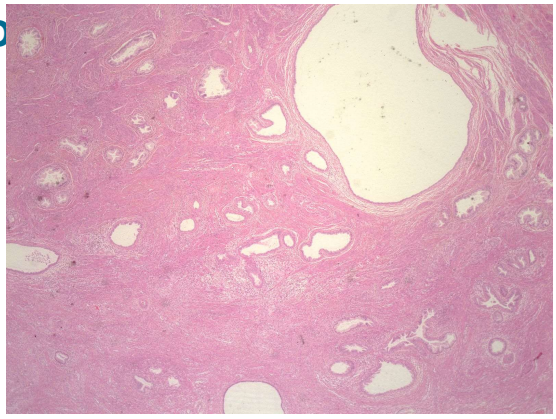
Well differentiated gastric type adenocarcinoma: formerly known as minimal deviation





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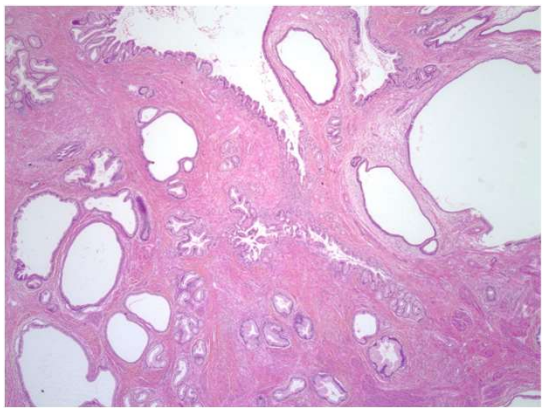
Well differentiated gastric type adenocarcinoma: formerly known as minimal deviation adenocarcinoma ("adenoma malignum") of the cervix



- Periglandular stromal reaction (usually focal), inflammation
- Cytologically atypical glands (by definition focal) –may merge into obvious gastric-type adenoca

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Adenoma malignum vs gastric type adenocarcinoma

Adenoma malignum = well-differentiated form of gastric-type adenocarcinoma

Spectrum with overlapping morphology, spatially adjacent

Term gastric-type adenocarcinoma is used to encompass this spectrum of mucinous adenocarcinomas

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IHC in gastric type adenocarcinoma

Positive	Negative	p53
MUC6 PAX8 (60-80%) CK7 CEA CAIX CK20/CDX2 ~50% HNF1b	• p16 patchy • HPV • ER, PR • Vimentin • p63 • P40	Mutated pattern in 50%

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Prognosis of gastric type adenocarcinoma

Worse than ordinary well-differentiated adenocarcinoma

50% survival for stage I

30% overall survival at 2 years

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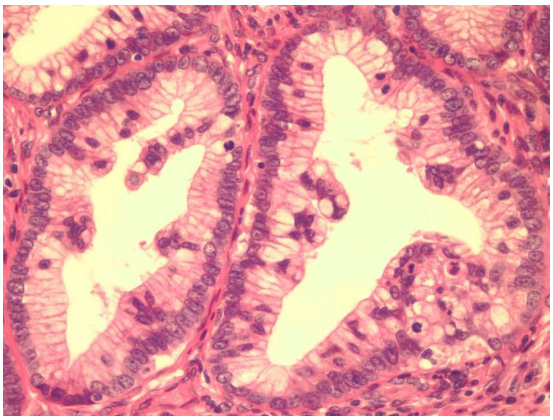
Precursor of gastric type adenocarcinoma?

- **Gastric type AIS**
- **Possibly from lobular endocervical glandular hyperplasia (LEGH)**



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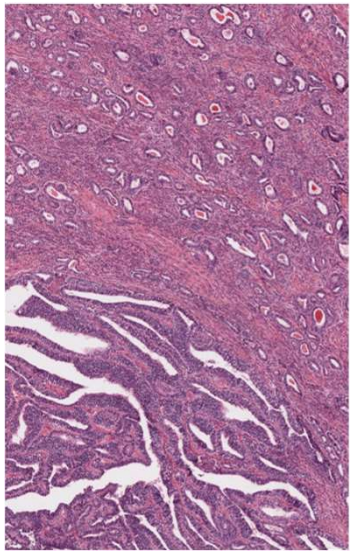
Mesonephric carcinoma

- **Malignant neoplasm with mesonephric (Wolffian) differentiation**
- **From mesonephric remnants along the course of the embryological mesonephric duct - deep in the lateral cervical wall**
- **Can be transmural and exophytic**
- **Differential diagnosis with mesonephric remnant hyperplasia:**
  - **architectural crowding**
  - **haphazard infiltrative growth**
  - **elevated mitotic activity**
  - **intraluminal necrosis**
  - **nuclear atypia**



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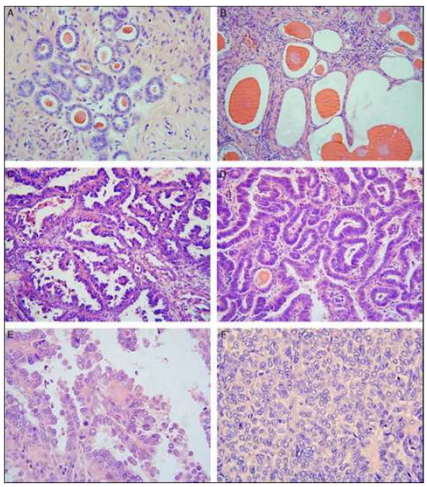
Mesonephric carcinoma

Classic pattern - tubular
Cuboidal cells
Lumina filled with dense eosinophilic secretions (PAS-positive and mucicarmine-positive)

NO squamous differentiation or cytoplasmic mucin!

Ductal (pseudoendometrioid) pattern: angulated glands lined by columnar cells

Other patterns: papillary, retiform, sex cord–like, hobnail, glomeruloid, sieve-like, spindled, and solid



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IHC in Mesonephric adenocarcinoma

CD10 pos	ER, CEA, p16 negative	PAX8, HNF-beta, HMGA2, GATA3, TTF-1 positive

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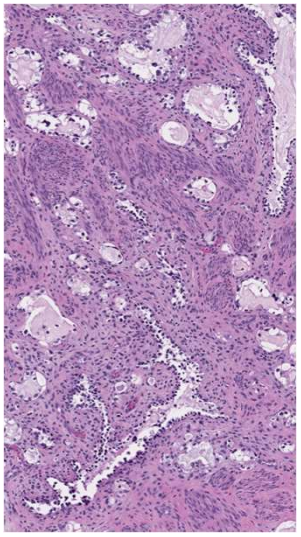
Adenocarcinoma, Clear cell type, HPV independent

- Sporadic or after in utero diethylstilbestrol (DES) exposure
- 3–4% of cervical adenocarcinomas
- Uniform, clear or eosinophilic, flat or cuboidal cells
  Tubulocystic, papillary, solid patterns



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Adenocarcinoma, Clear cell type, HPV independent

Can be diffusely positive for p16 but no HPV

HNF1β +
Napsin A+
AMACR +

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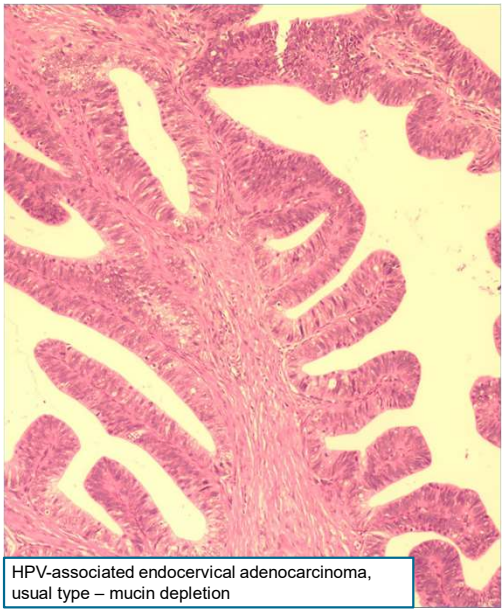
Endometrioid adenocarcinoma

- **Primary in the cervix very rare**
  - **May arise in endometriosis???**
  - **p16 negative**
  - **No apical mitoses/ apoptotic bodies**
- **Endometrial carcinoma involving cervix – much more common**
- **If it is cervical – likely mucin-depleted usual type HPV-associated adenocarcinoma**
- **Diagnosis of exclusion**



HPV-associated endocervical adenocarcinoma, usual type – mucin depletion

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Serous carcinoma???

- **Almost never primary in cervix**
- **Not included in the WHO 2020**

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HPV-associated adenocarcinomas

- **Usual type**
- **Mucinous**
  - **NOS (endocervical)**
  - **Intestinal**
  - Signet ring
  - Invasive SMILE

HPV-independent adenocarcinomas

- **Gastric type**
- **Mesonephric**
- **Clear cell**
- **Endometrioid**

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Simple rules to classify endocervical adenocarcinoma

- **HPV/p16 are not necessary: morphology is tightly linked to HPV status**
- **If difficult to classify, use HPV status and intracytoplasmic mucin:**
  - **HPV-associated or HPV-independent adenocarcinoma NOS**
    - **mRNA-based high-risk HPV in situ hybridization > DNA PCR**
    - **Indirect assay – p16**

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Caveats of p16 and HPV testing

- **p16 can be positive in HPV-independent adenocarcinoma (up to 30% gastric type), and negative in up to 30% SMILE**
- **HPV is more sensitive, specific, better PPV and NPV but not widely available**

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IHC algorithm for endocervical adenocarcinomas

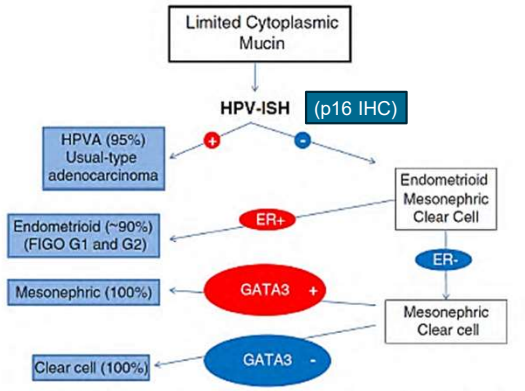

FIGURE 6. IHC algorithm for ECAs with limited cytoplasmic mucin. FIGO indicates International Federation of Gynaecology and Obstetrics.

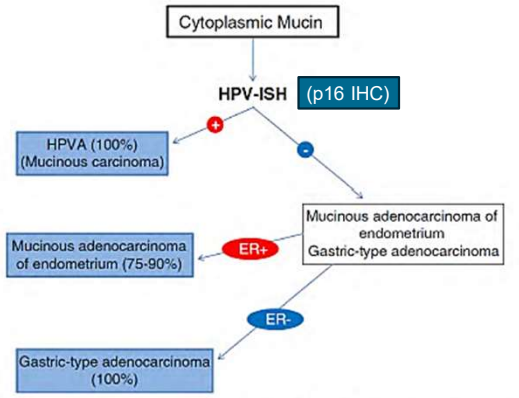

FIGURE 7. IHC algorithm for ECAs containing obvious cytoplasmic mucin.

Stolnicu et al, AJSP 2018; 42:989

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Questions?

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