



Histotyping ovarian cancers

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Key Issues

- Review morphology, immunophenotype, genotype and differential diagnosis*
- Emphasize endometrioid and clear cell tumors
- Suggest refinements to diagnostic criteria

Rationale for “histotyping”

- Distinct disease entities
 - BRCA1 and 2
 - Tumor progression from:
 - Endometriosis
 - Borderline tumor
- Diagnostic criteria for carcinoma
- Carcinoma grading
- Therapeutic relevance

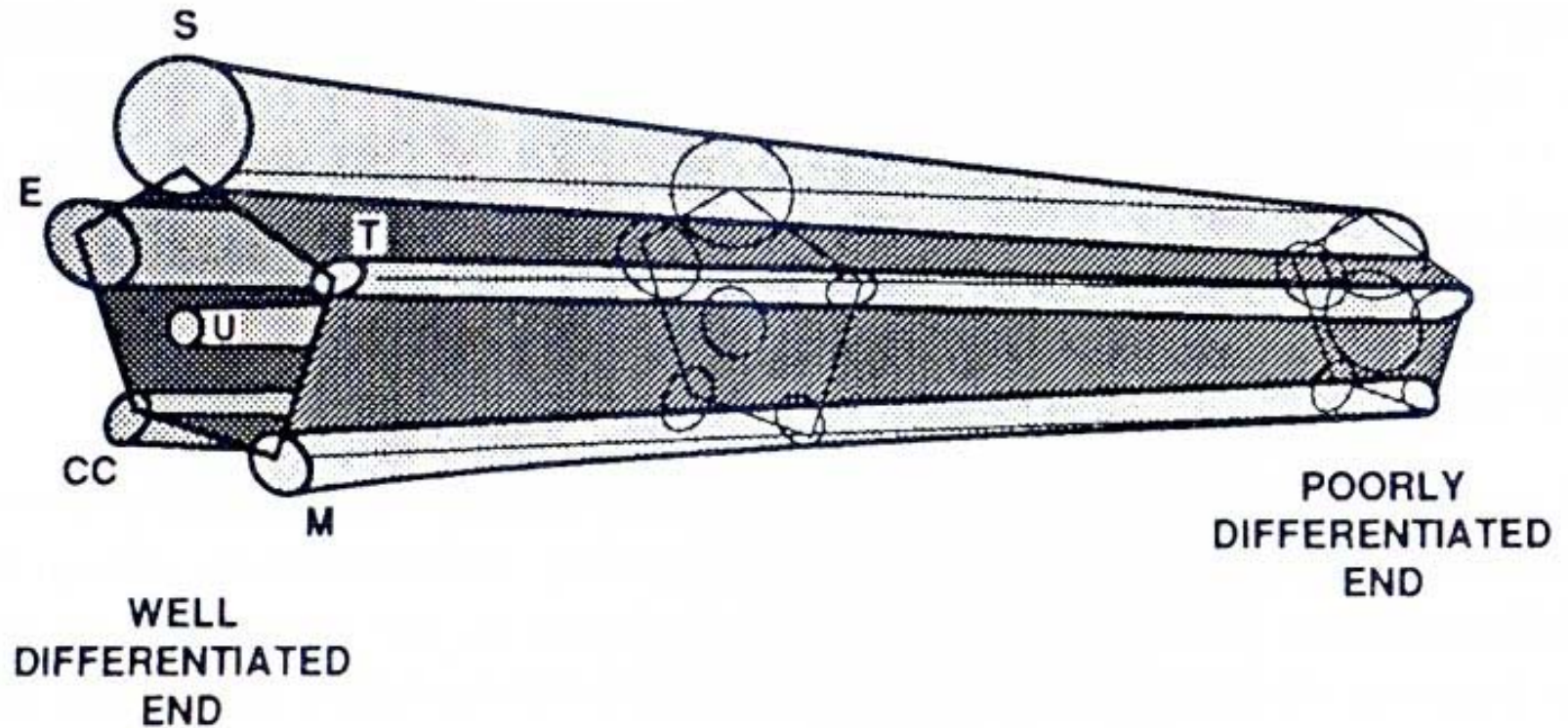
Introduction

- WHO classification
 - Serous
 - Mucinous
 - Endometrioid
 - Clear cell
 - Transitional
 - Squamous
 - Mixed epithelial
 - Undifferentiated

WHO critique

- WHO approach
 - Morphologically based, but not entirely objective
- Problems
 - Mucinous carcinoma
 - Poorly differentiated endometrioid carcinoma
 - Transitional cell carcinoma
 - Mixed epithelial carcinoma

DIAGRAM 5C. (below).



Hendrickson MR, Longacre TA.
Classification of surface epithelial neoplasms of the ovary
Pathology (Phila). 1:189-254, 1993

Bland cytological features
and/or
minimal epithelial stratification

SEROUS

- Psammoma bodies (more than rare)
- Tubal like or indeterminate epithelium
- Pink cells
- Papillary architecture with stromal cores
- Micropapillary structures embedded in collagen

TRANSITIONAL

- Transitional epithelium ± mucinous differentiation ± squamous differentiation
- Coffee bean shaped nuclei with grooves
- Sheets embedded in fibrous stroma
- Stromal Ca⁺⁺
- Papillary structures resembling low grade urothelial transitional cell carcinoma

MUCINOUS

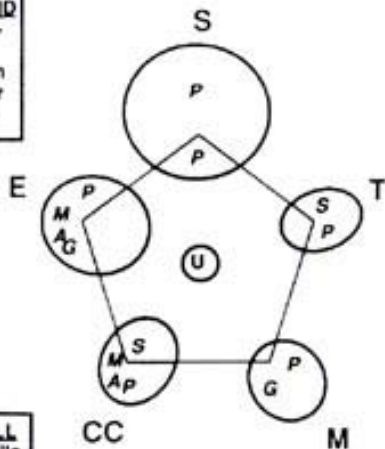
- Intracytoplasmic mucin (more than rare cells)
- Pseudomyxoma ovarii

ENDOMETRIOID

- Squamous-Morular differentiation
- Microacinar architecture

CLEAR CELL

- Hobnail cells
- Cleared or eosinophilic cytoplasm



A P = papillary MA = microacinar G = tubulo-cystic S = sheets

Considerations other than SENS

ENDOMETRIOID

- Sertoli cell tumor
- Sertoli-Leydig tumor*
- Granulosa cell tumor
- Carcinoid
- Struma ovarii
- Endometrioid-like yolk sac tumor*
- Ovarian tumor of probable Wolffian origin
- Metastasis (large bowel)
- Metastatic uterine corpus carcinoma (? simultaneous primary)

UNCLASSIFIED

- Metastasis

IF STROMA PROMINENT

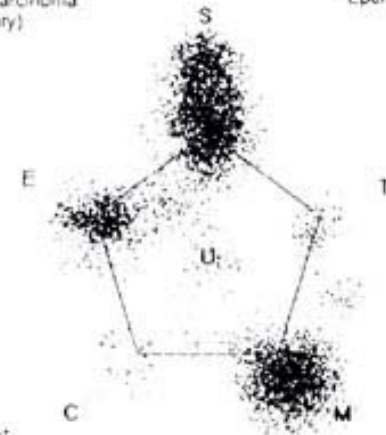
- Stromal prominence or luteinization in SEN
- Adenosarcoma
- Carcinosarcoma

CLEAR CELL

- Endodermal sinus tumor*
- Hepatoid EST*
- Struma ovarii
- Steroid cell tumor
- Juvenile granulosa cell tumor*
- Ovarian tumor of probable Wolffian origin

SEROUS

- Reiter's Sertoli-Leydig tumor*
- Mesothelioma
- Ependymoma



B * = occurs in children or young women EST=Endodermal sinus tumor

Hendrickson MR, Longacre TA.
Classification of surface epithelial neoplasms of the ovary
Pathology (Phila). 1:189-254, 1993

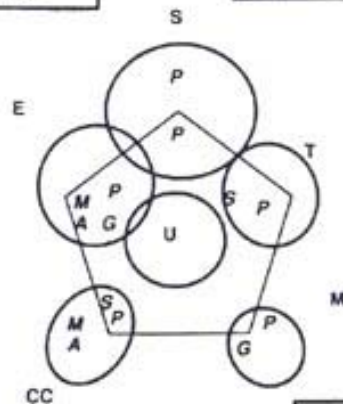
malignant cytological features and/or moderate epithelial stratification

- SEROUS**
- Psammoma bodies (more than rare)
 - Tubal-like or indeterminate epithelium
 - Pink cells
 - Papillary architecture with stromal cores
 - Slit-like spaces produced by closely approximated papillae
 - Micropapillary (lace-like) structures embedded in collagen
 - More uniform appearance than endometrioid or mucinous

- TRANSITIONAL**
- Abortive papillary structures resembling moderately differentiated urothelial transitional cell carcinoma
 - Sheets of moderately differentiated transitional cells embedded in fibrous stroma

- ENDOMETRIOID**
- Squamous-morular differentiation
 - Microglandular aggregates
 - More heterogeneous from area to area than serous

- CLEAR CELL**
- Hobnail cells
 - Cleared or Eosinophilic cytoplasm



- MUCINOUS**
- Intracytoplasmic mucin (more than rare cells)
 - Pseudomyxoma ovarii unusual
 - More heterogeneous from area to area than serous
 - Signet ring cells

A P = papillary G = tubulo-cystic MA = microacinar S = sheets

Considerations other than SENS

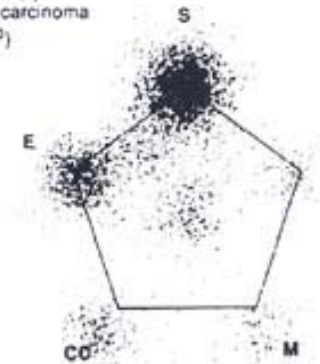
- ENDOMETRIOID**
- Sertoli cell tumor
 - Sertoli-Leydig tumor*
 - Granulosa cell tumor
 - Carcinoid
 - Struma ovarii
 - Endometrioid-like yolk sac tumor*
 - Ovarian tumor of probable Wolffian origin
 - Metastasis (large bowel)
 - Metastatic uterine corpus carcinoma (? simultaneous¹⁰)

- UNCLASSIFIED**
- Metastasis
- IF STROMA PROMINENT
- Adenosarcoma
 - Carcinosarcoma

- CLEAR CELL**
- Endodermal sinus tumor*
 - Dysgerminoma
 - Hepatoid EST*
 - Choriocarcinoma
 - Struma ovarii
 - Steroid cell tumor
 - Juvenile granulosa cell tumor*
 - Ovarian tumor of probable Wolffian origin
 - Metastatic renal cell carcinoma

- SEROUS**
- Retiform Sertoli-Leydig tumor*
 - Embryonal carcinoma
 - Mesothelioma
 - Metastatic uterine papillary serous carcinoma

- TRANSITIONAL**
- Metastasis of urothelial origin



B * = occurs in children or young women EST=Endodermal sinus tumor

Hendrickson MR, Longacre TA.
 Classification of surface epithelial neoplasms of the ovary
 Pathology (Phila). 1:189-254, 1993

Considerations other than SENS

Malignant cytological features and/or marked epithelial stratification

SEROUS

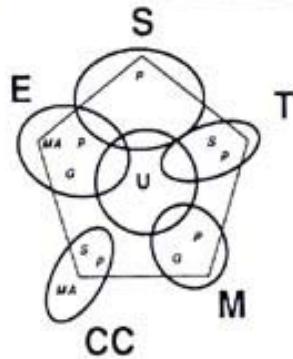
- Solid sheets of small, uniform, sometimes vaguely spindled cells
- Syncytial-like aggregates
 - Psammoma bodies (more than rare); usually not helpful
- Papillary architecture with stromal cores
- Slit-like spaces

TRANSITIONAL

- Abortive papillary structures resembling high grade urothelial transitional cell carcinoma
- Sheets of transitional cells embedded in fibrous stroma

ENDOMETRIOID

- Squamous-morular differentiation
- Microglandular aggregates
- More heterogeneous from area to area than serous



CLEAR CELL

- Hobnail cells
- Cleared or eosinophilic cytoplasm

MUCINOUS

- Intracytoplasmic mucin (more than rare cells)
- Pseudomyxoma ovarii rare
- More heterogeneous from area to area than serous
- Signet ring cells

P = papillary MA = microacinar G = tubulo-cystic S = sheets

A

ENDOMETRIOID

- Sex cord stromal tumor*

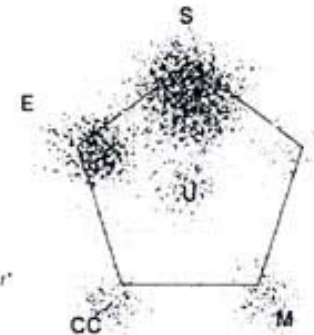
- Carcinoid
- Struma ovarii
- Component of carcinosarcoma

CLEAR CELL

- Endodermal sinus tumor*
- Metastatic renal cell carcinoma
- Component of carcinosarcoma

SEROUS

- Retiform Sertoli-Leydig tumor*
- Embryonal carcinoma
- Component of carcinosarcoma



TRANSITIONAL

- Metastasis of urothelial origin
- Component of carcinosarcoma

MUCINOUS

- Metastasis from GI tract

UNDIFFERENTIATED

- Endometrial stromal sarcoma
 - Fibrosarcoma
 - Carcinosarcoma
- Granulosa cell tumor
- High grade gonadal stromal tumor
 - Adult granulosa cell tumor
 - Juvenile granulosa cell tumor*
- Small cell carcinoma*
- Embryonal carcinoma*
 - Carcinoid
- Metastasis (e.g. breast, melanoma)
 - Hematolymphoid

* = occurs in children or young women

B

Hendrickson MR, Longacre TA.
 Classification of surface epithelial neoplasms of the ovary
 Pathology (Phila). 1:189-254, 1993

Serous carcinoma: morphology

- Overview
- Problems
 - Architecture
 - Glandular
 - Cribriform
 - Microcystic
 - Trabecular
 - Cytology
 - Clear cells
 - Signet ring cells
 - Spindle cells

Serous carcinoma: immunophenotype

- All serous
 - WT1 (>70%)
- Low-grade serous
 - ER/PR
- High-grade serous
 - P53 (overexpression, >70%)
 - P16
 - Variable ER/PR
 - Variable loss of BRCA1

Serous carcinoma: genotype

- Low grade serous
 - *BRAF* mutation
 - *K-ras* mutation
- High grade serous
 - *P53* mutation
 - BRCA1 or 2 abnormalities (sporadic)
 - *BRCA1 or 2* mutation (familial)

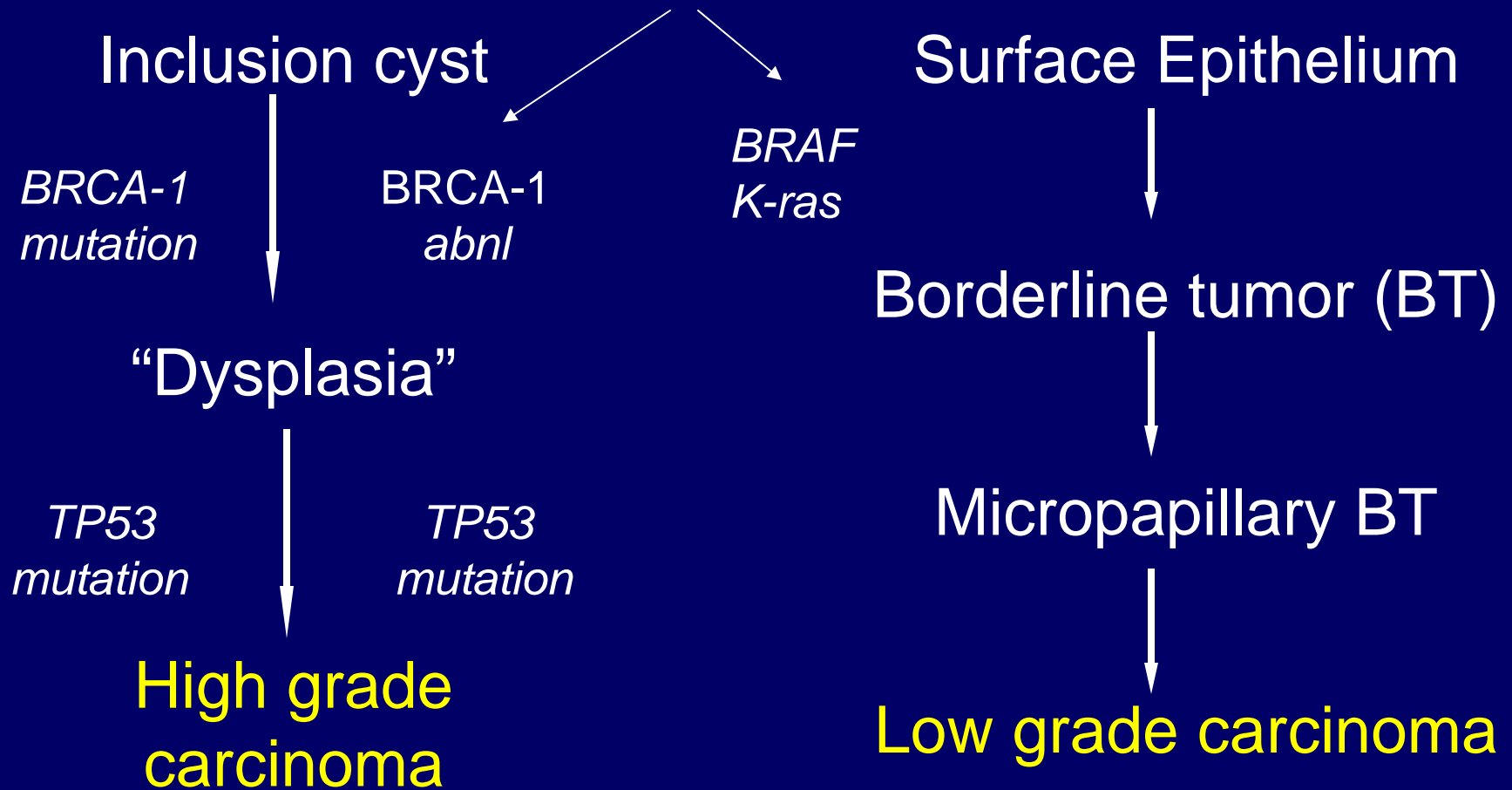
Singer G, et al. *Am J Surg Pathol* 29:218-24, 2005

Shaw PA, et al. *Int J Gynecol Pathol* 21:407-11, 2002

Serous carcinoma: pathogenesis

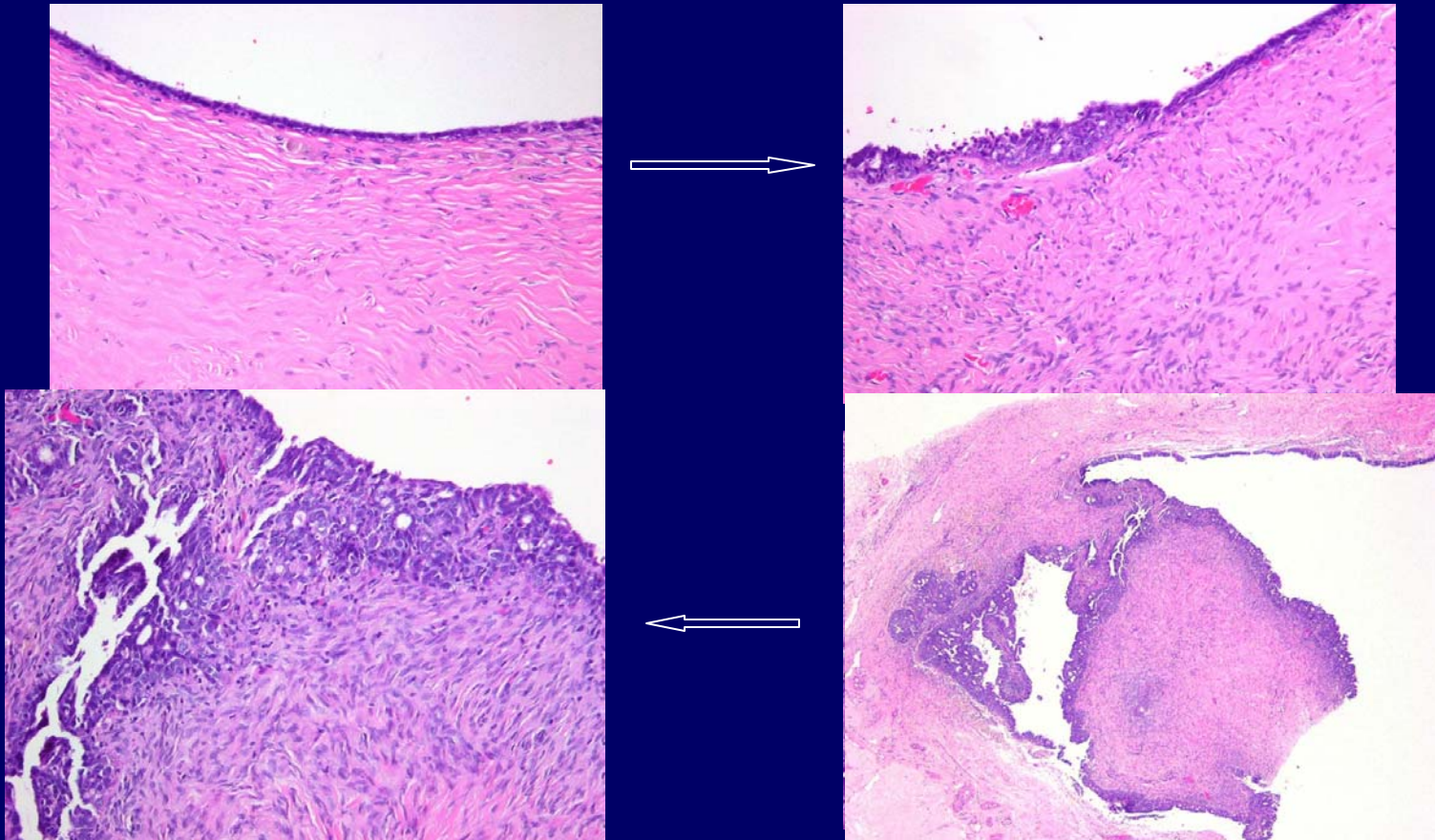
Familial

Sporadic

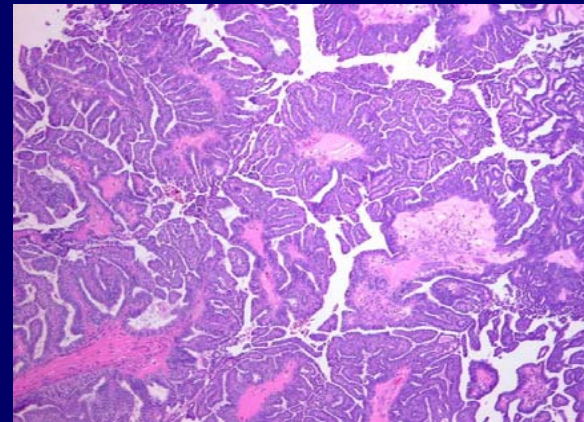
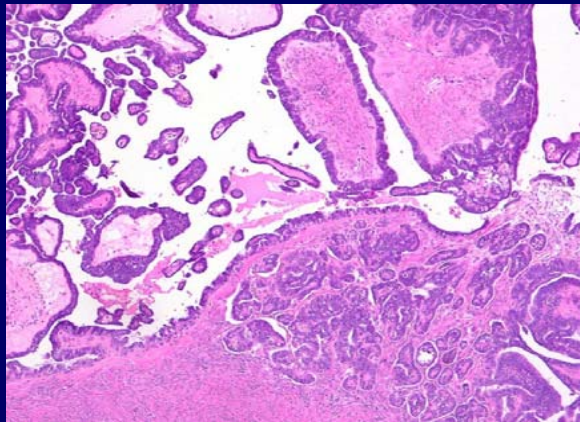
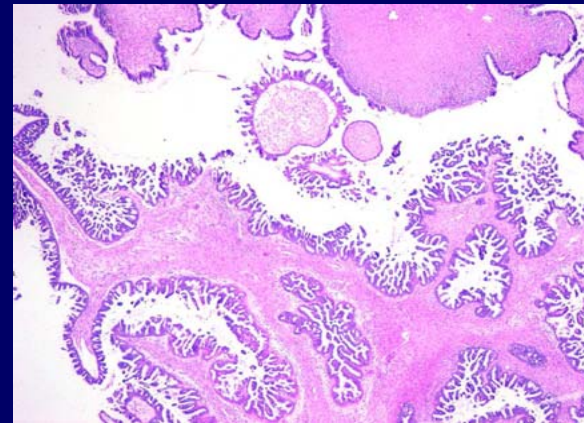
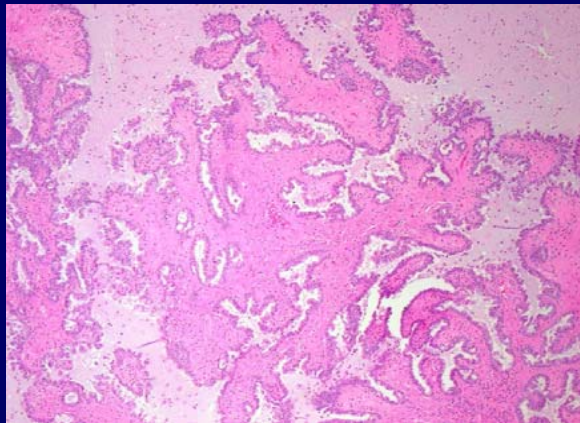


Singer G, et al. *Am J Surg Pathol* 29:218-24, 2005

High grade serous carcinoma: pathogenesis



Low grade serous carcinoma: pathogenesis



Serous carcinoma: differential diagnosis

- Glandular, cribriform, microcystic, trabecular:
 - Mucinous, endometrioid, clear cell, transitional
- Clear cells, signet ring cells:
 - Clear cell, endometrioid, mucinous

Ovarian carcinoma classification: serous

- Broad range of histologic features
 - Slit-like spaces, irregular luminal contours
- Frequent WT1
- Low-grade: serous borderline tumor, *BRAF/K-ras*, ER/PR
- High-grade: tubal intraepithelial carcinoma, *p53*, *p16*, loss of BRCA1, BRCA1 or 2 family
- Other entities are excluded

Serous tumors: prevalence

- 80-85% of ovarian carcinomas
- 95% of stage III-IV ovarian carcinoma
- Low stage serous carcinomas are rare
 - ~25% of stage I/II carcinomas are serous
 - ~25% of stage I/II serous carcinomas are stage I

Leitao MM, et al. *Am J Surg Pathol* 28:147-59, 2004

Seidman JD, et al. *Int J Gynecol Pathol* 23:41-4, 2004

(Intestinal) Mucinous tumors: morphology

- Overview
- Problems
 - Primary versus metastasis
 - Paucity of intracytoplasmic mucin/prevalence of extracellular mucin
 - Borderline tumor versus carcinoma

Mucinous tumors: immunophenotype

- Immunophenotype
 - CK7>20 (GI ddx)
 - Negative racemase and β -catenin (GI ddx)
 - Negative p16 (Endocervical ddx)
 - Negative ER (Endometrioid ddx)
 - Retained SMAD4/DPC4 (Pancreatic ddx)
 - Negative mesothelin and fascin (Pancreatic ddx)

Mucinous carcinoma: pathogenesis

Mucinous cystadenoma



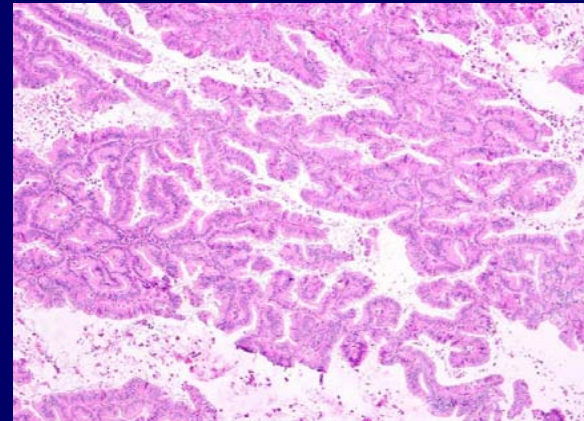
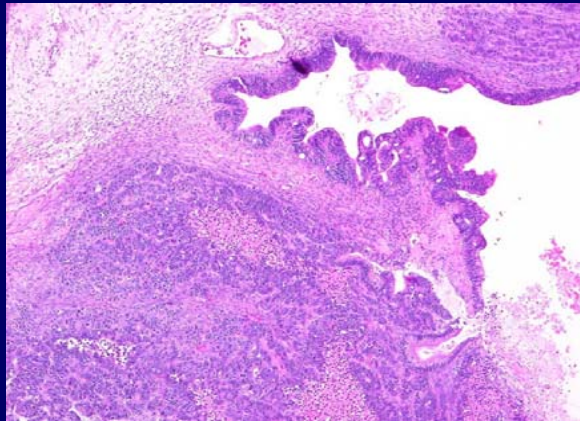
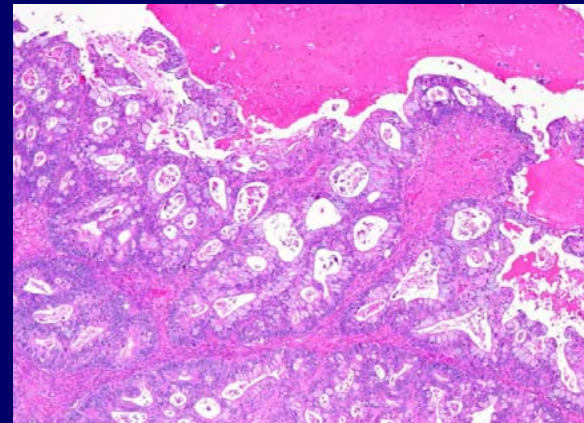
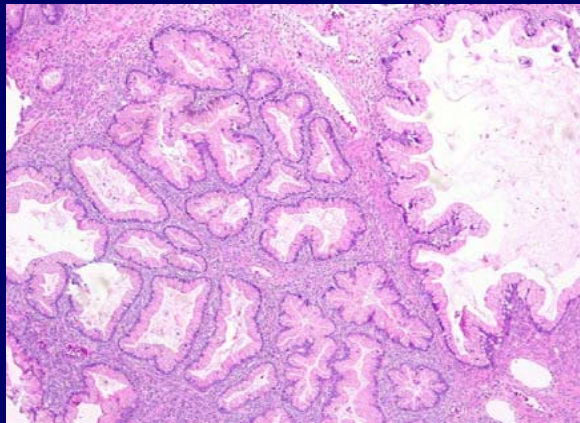
K-ras

Intestinal mucinous BT



Intestinal mucinous
carcinoma

Mucinous carcinoma: pathogenesis



Mucinous tumors: borderline versus carcinoma

- Expansile invasion
 - Large cribriform glands
 - Extensive gland fusion
 - Complex papillary architecture
- Destructive invasion

Am J Surg Pathol 23:617-35, 1999, 24:1447-64, 2000, 26:139-52, 2002, 22:1449-62, 1998

- Low stage ovarian carcinomas (n=140)
 - Diagnosis changed to borderline (30%)
 - 70% were mucinous or endometrioid

Leitao MM, et al. *Am J Surg Pathol* 28:147-59, 2004

Mucinous tumors: differential diagnosis

- Exclude metastasis
- Scant intracytoplasmic mucin
 - Endometrioid
- Extracellular mucin
 - Pseudomyxoma peritonei
 - Low grade serous carcinoma (cribriform)

Mucinous tumors: features favoring metastasis

- Bilateral disease
- Surface involvement
- Destructive stromal invasion
- Nodular growth pattern
- Single cells/signet ring cells
- Vascular invasion

Ovarian carcinoma classification: intestinal mucinous

- Intracytoplasmic mucin, expansile invasion
- Intestinal mucinous borderline tumor
- CK7>20, retained SMAD4
- Negative racemase, β -catenin, ER, p16, mesothelin, fascin
- *K-ras*
- Other entities are excluded: exclude metastasis

Mucinous tumors: prevalence

- Only <3% of all ovarian carcinomas
- >2/3 are stage I
- ~15% of all stage I tumors

Leitao MM, et al. *Am J Surg Pathol* 28:147-59, 2004

Seidman JD, et al. *Am J Surg Pathol* 27(7):985-93, 2003

Riopel MA, et al. *Am J Surg Pathol* 23(6):617-35, 1999

Seromucinous tumors

- Definition and synonyms
- Similarities with low grade serous tumors
 - Architecture
 - Clinical profile
- Differences with intestinal mucinous tumors
 - Morphology
 - Clinical profile
- Similarities with endometrioid tumors
 - Endometriosis

Endometrioid tumors: morphology

- Overview
- Problems
 - Architecture:
 - Cribriform
 - Trabecular
 - Papillary
 - Cytology:
 - Intracytoplasmic mucin
 - Clear cells
 - High nuclear grade
 - Borderline tumor versus carcinoma

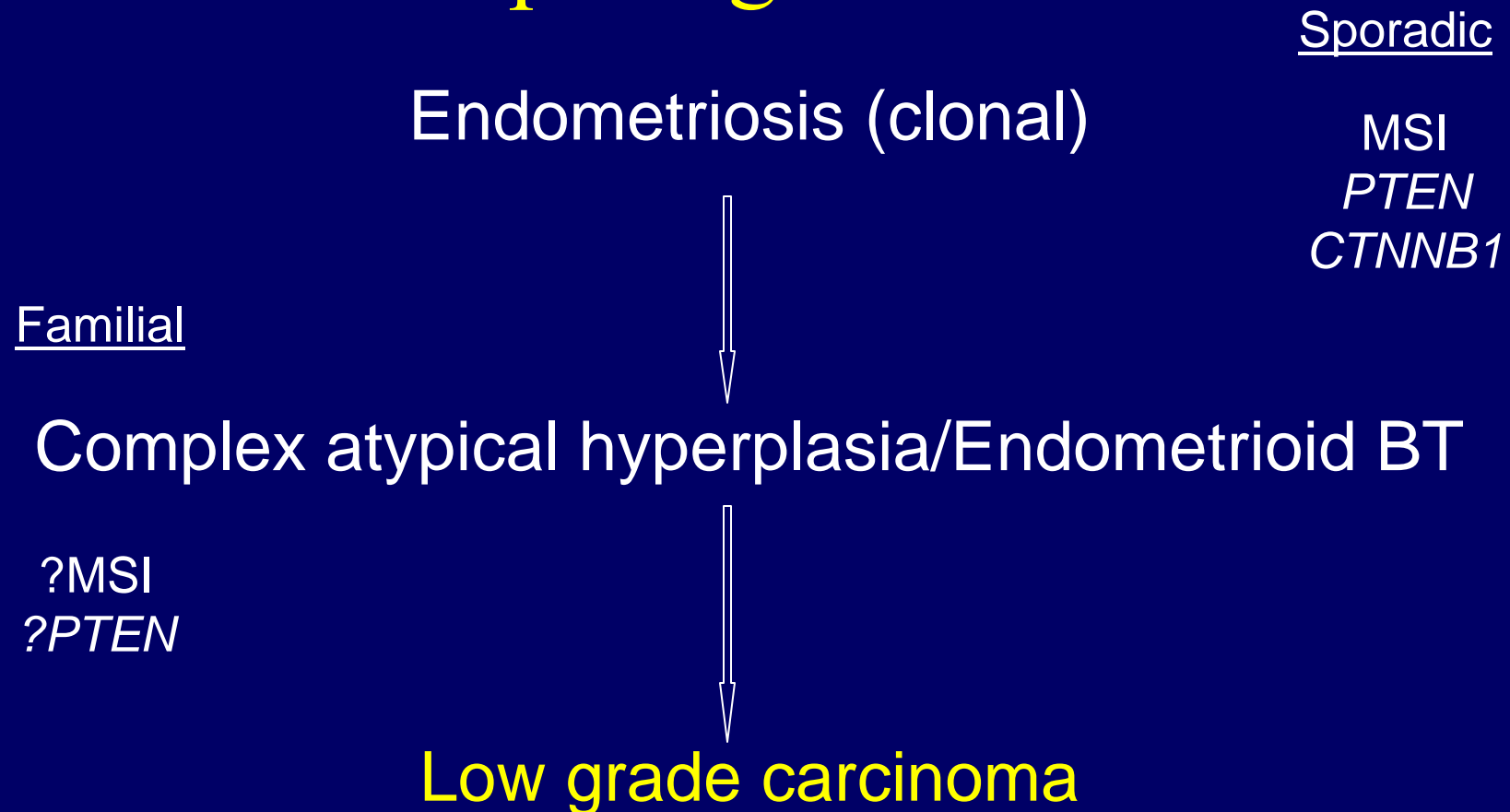
Endometrioid tumors: immunophenotype

- ER/PR
- β -catenin
- Not WT1
- P53 in “high grade” examples

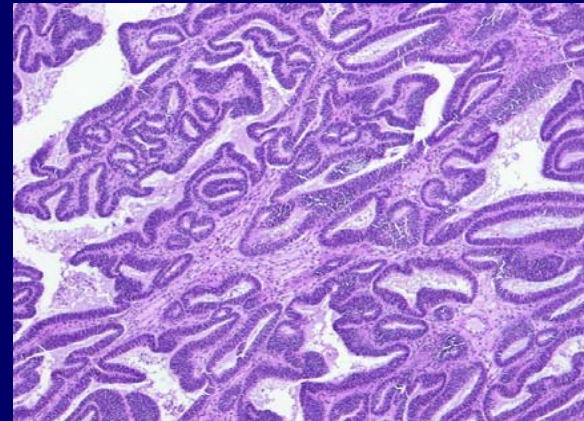
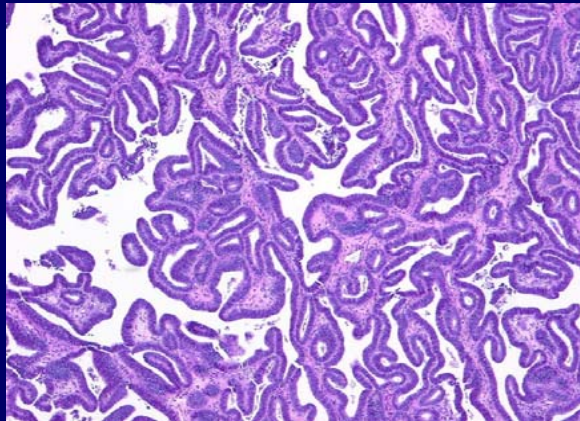
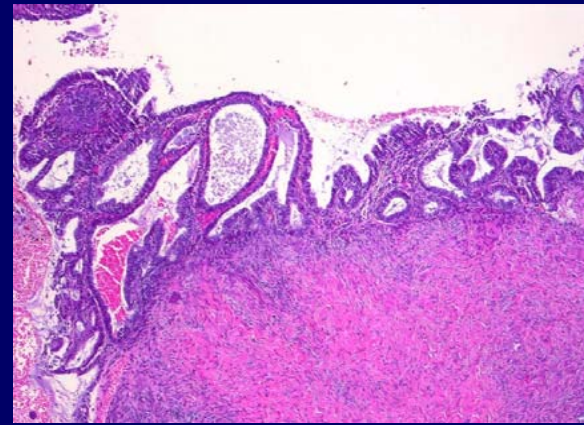
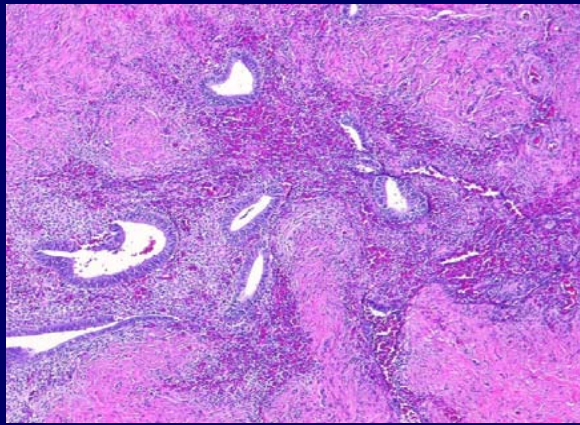
Endometrioid tumors: genotype

- *CTNNB-1* (β -catenin)
- *PTEN*
- Microsatellite instability (MSI)

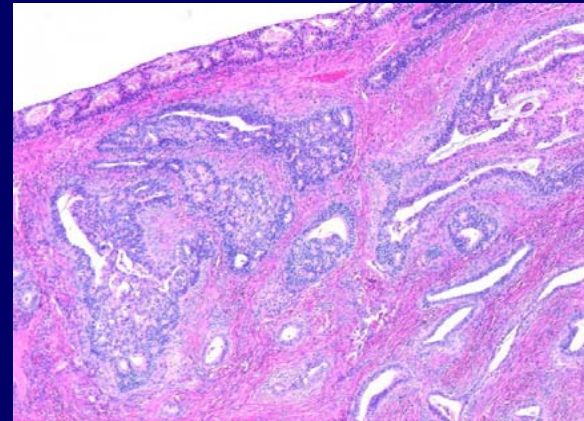
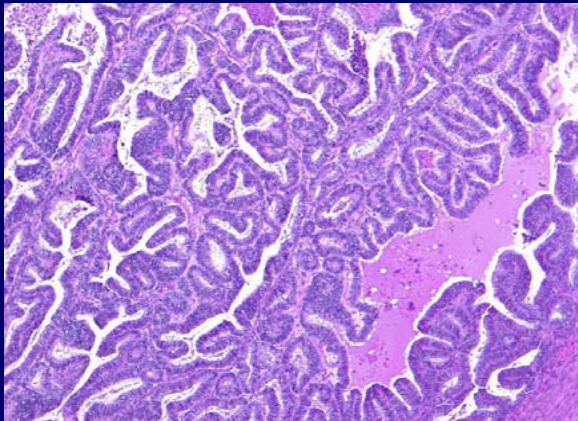
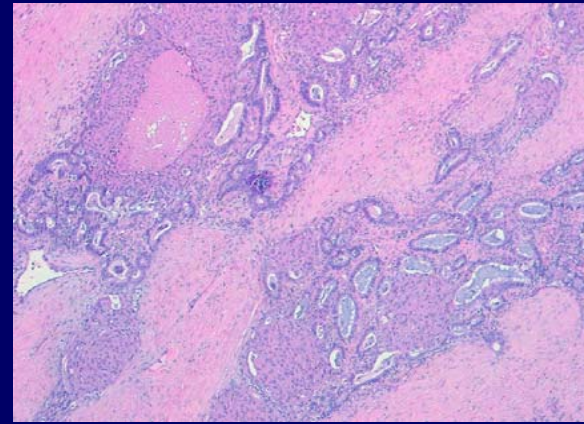
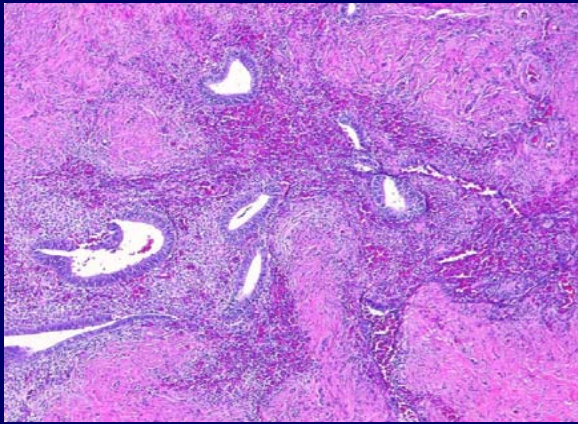
Endometrioid carcinoma: pathogenesis



Endometrioid carcinoma: pathogenesis



Endometrioid carcinoma: pathogenesis



Endometrioid tumors: borderline versus carcinoma

- Expansile invasion
 - Large cribriform glands
 - Extensive gland fusion
 - Complex papillary architecture
- Destructive invasion

Am J Surg Pathol 24:1465-79, 2000; *Am J Surg Pathol* 27:1253-9, 2003

- Low stage ovarian carcinomas (n=140)
 - Diagnosis changed to borderline (30%)
 - 70% were mucinous or endometrioid (70%)

Leitao MM, et al. *Am J Surg Pathol* 28:147-59, 2004

Endometrioid tumors: invasion patterns

- 13 grade 1 and 2 endometrioid carcinomas (8 IA, 5 IC); median F/U 81 mos:
 - 9 expansile (9/9 NED)
 - 4 expansile and destructive* (3/4 NED)
- Low grade, stage I endometrioid carcinoma with expansile invasion: **VERY** limited malignant potential

Endometrioid tumors: differential diagnosis

- Cribriform, papillary, trabecular:
 - Serous, transitional
- Intracytoplasmic mucin, clear cells, high nuclear grade:
 - Metastasis, mucinous, clear cell, serous

Ovarian carcinoma classification: endometrioid

- Endometrial-like, metaplasias, secretory change, expansile invasion
- Endometriosis, endometrioid borderline tumor, endometrioid uterine carcinoma
- ER/PR, β -catenin; not WT1
- *CTNNB-1* (β -catenin), *PTEN*, MSI-H
- Other entities are excluded

Endometrioid tumors: prevalence

- 10% of ovarian carcinomas
- Most common stage I carcinoma (~50%)
- Most endometrioid carcinomas are Stage I or II (>2/3)

Leitao MM, et al. *Am J Surg Pathol* 28:147-59, 2004

Seidman JD, et al. *Int J Gynecol Pathol* 23:41-4, 2004

Clear cell tumors: morphology

- Overview
- Problems
 - Architecture:
 - Papillary
 - Solid
 - Cytology:
 - Clear cytoplasm
 - Oxyphilic variant
 - Borderline tumor versus carcinoma

Clear cell tumors: diagnostic reproducibility, immunophenotype, lessons learned

- Clear cell carcinomas have a limited and distinctive architectural repertoire and immunophenotype
- Mixed epithelial tumors containing clear cells (MET-C) are not reproducibly diagnosed
- MET-Cs are seldom clear cell carcinomas—most are serous carcinomas

Interobserver agreement

Diagnosis	Kappa	Degree of agreement beyond chance
CCC	0.82	Almost perfect
SC	0.59	Moderate
MET-C	0.32	Fair
Overall	0.62	Substantial

Clear cell immunophenotype

	WT1	ER	BRCA1
CCC	10%	10%	100%
SC	90%	90%	64%
MET-C CC	90%	90%	60%
MET-C SC	90%	90%	40%

Han G, et al. USCAP 2007, abstract

Clear cell tumors: immunophenotype

- Immunophenotype
 - Paucity of ER/PR
 - Lack of WT1
 - Variable p53 expression
 - Low proliferation rate

Clear cell tumors: genotype

- Mutations:
 - *PTEN*
 - *TGF beta R2*
 - *K-RAS*
- MSI-H

Clear cell carcinoma: pathogenesis

Endometriosis



Atypical endometriosis or Endometrioid carcinoma

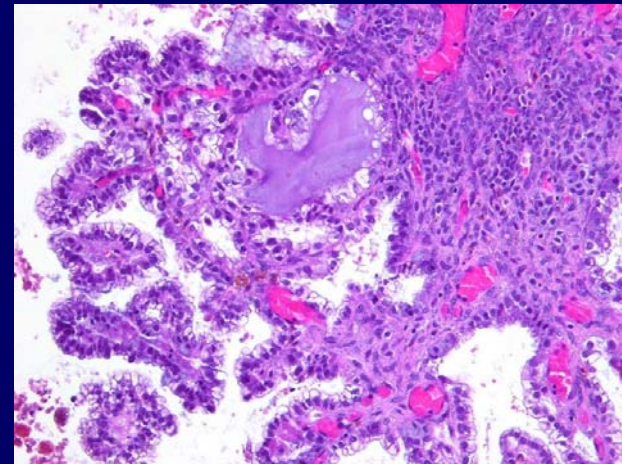
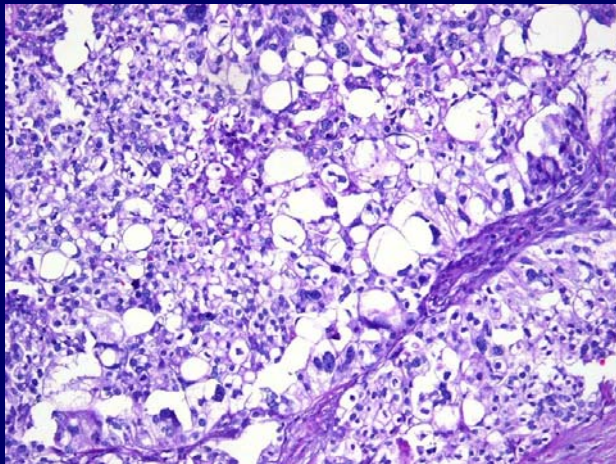
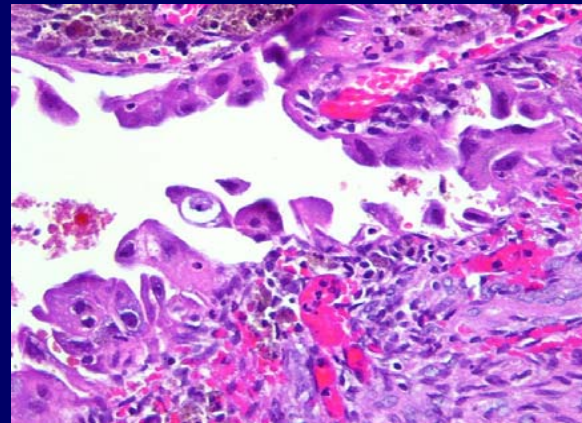
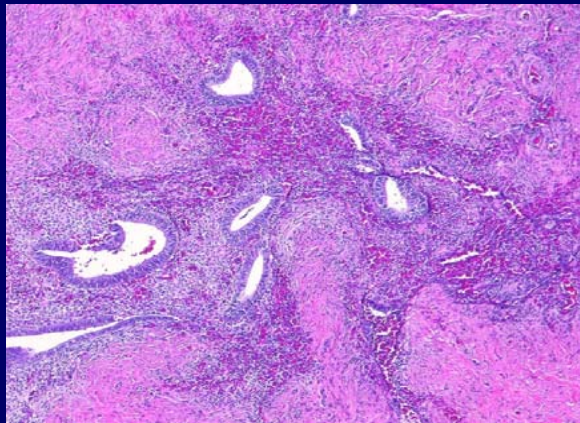
(Clear cell borderline tumor)



Clear cell carcinoma



Clear cell carcinoma: pathogenesis



Clear cell tumors: borderline versus carcinoma

- Adenofibromatous
 - Destructive invasion greater than microinvasion: carcinoma
 - What counts as invasion?
 - Pure borderline tumors are almost never encountered
- Papillary
 - Essentially always considered carcinoma
 - Importance of cytologic characteristics
 - Nuclear grade?
 - Ddx with papillary endometrioid carcinoma and serous borderline tumor

Clear cell tumors: differential diagnosis

- Papillary, tubulocystic, solid
 - Serous, seromucinous, endometrioid
- Hobnail cells, clear cells
 - Serous, seromucinous, endometrioid

Ovarian carcinoma classification: clear cell

- Papillary, tubulocystic, solid, hobnail, frequently clear cytoplasm
- Endometriosis, clear cell borderline tumor
- Low ER/PR, WT1, p53, mib-1
- MSI-H, *PTEN*
- Lack of features that define other entities
 - Metaplasias, secretory changes
 - Multilayering, serrated luminal profiles

Clear cell tumors: prevalence

- 5% of ovarian carcinomas
- Disproportionately represented in stages I and II
- 25% of stage I and II carcinomas are clear cell
- Most clear cell carcinomas are low stage at presentation (>2/3)

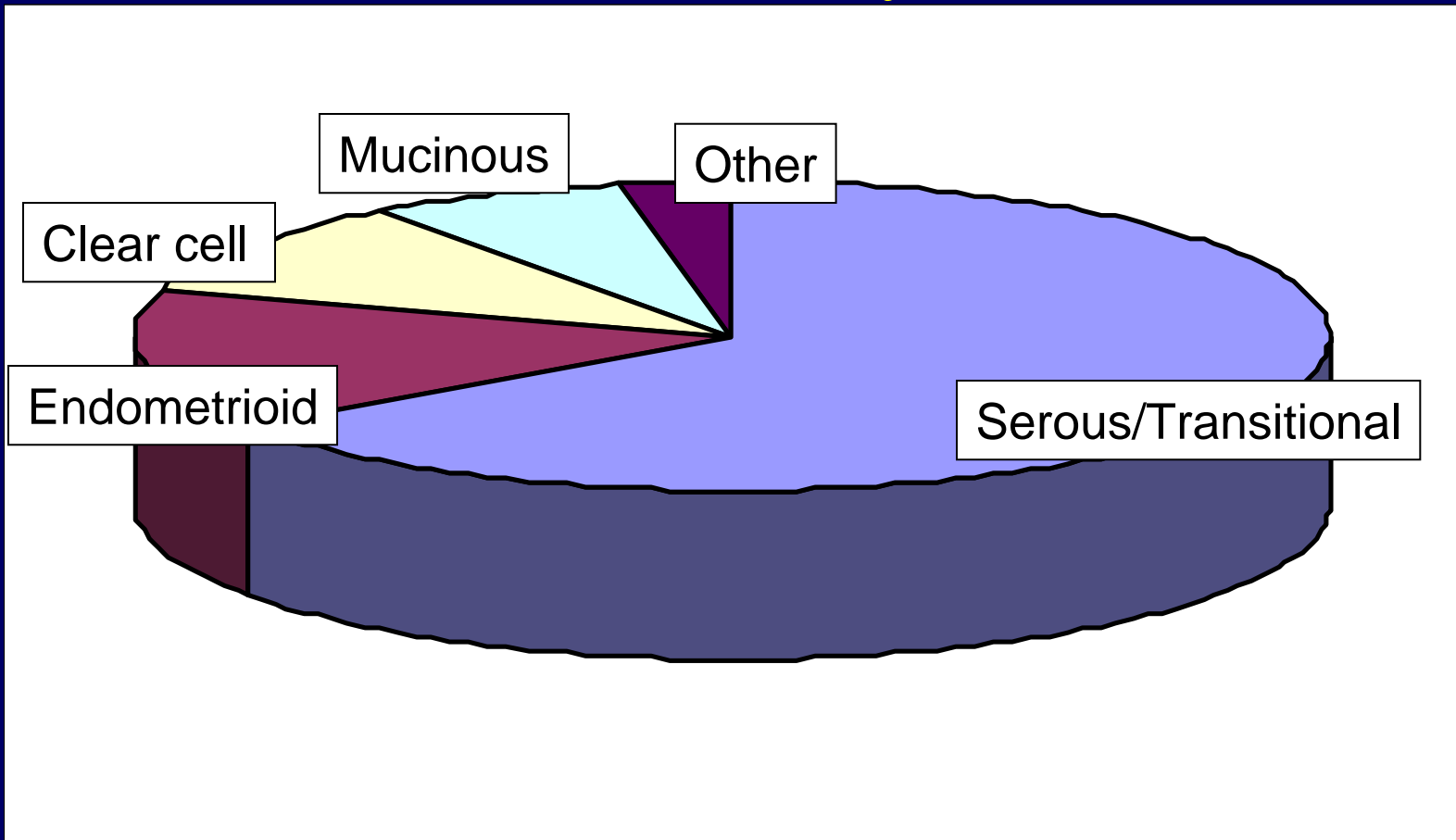
Leitao MM, et al. *Am J Surg Pathol* 28:147-59, 2004

Seidman JD, et al. *Int J Gynecol Pathol* 23:41-4, 2004

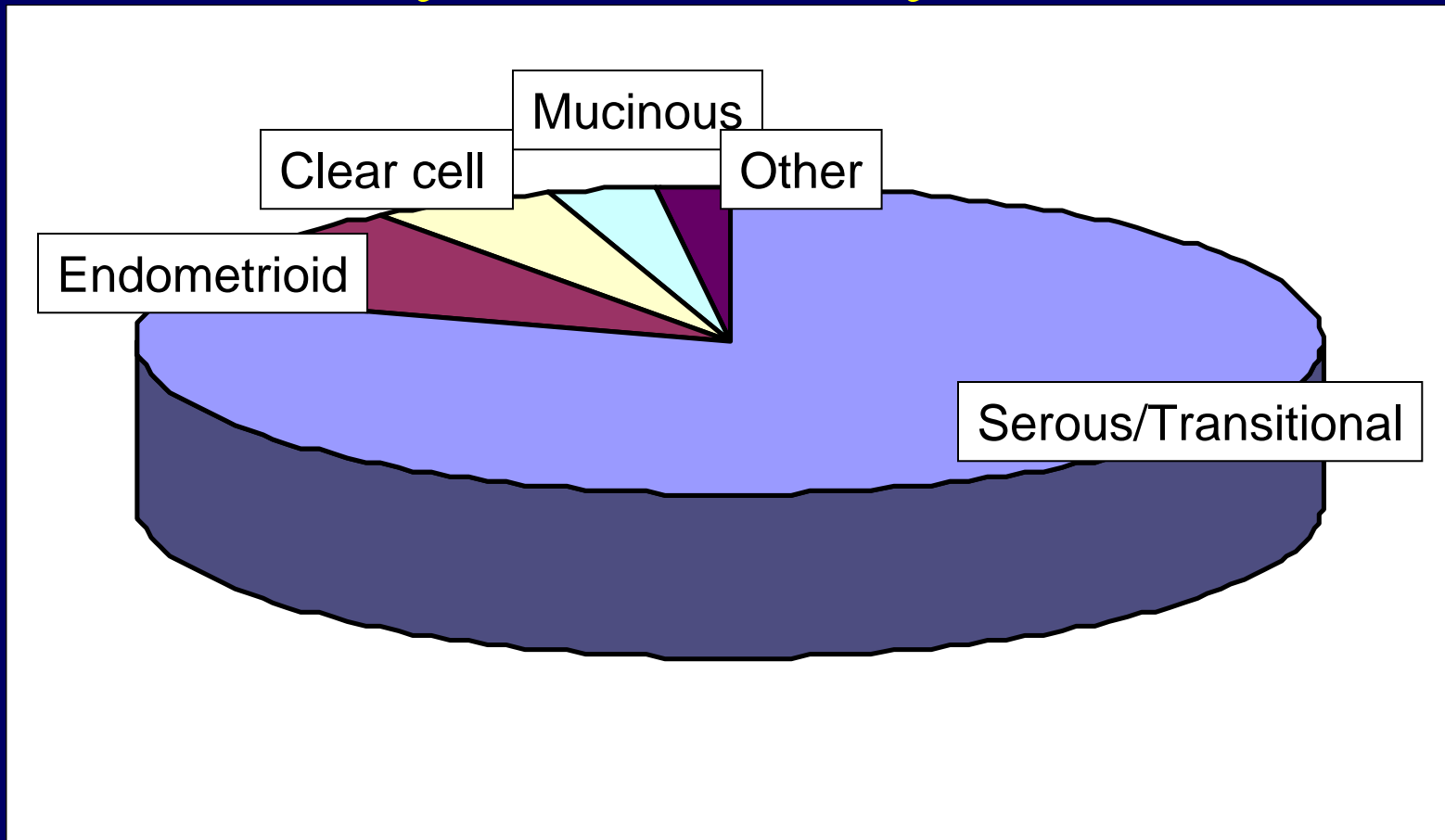
Transitional cell carcinoma

- Definition
- Similarities with high grade serous tumors
 - Morphology
 - Immunophenotype
 - ?Clinical profile
- Differences with urothelial carcinoma

Malignant surface epithelial tumors: late 20th century, West



Malignant surface epithelial tumors: early 21st century, West



Seidman JD, et al. *Int J Gynecol Pathol* 23:41-4, 2004