Prostate biopsy
from diagnostic criteria to molecular markers

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Prostate Needle Biopsy

Cancer ?

Yes

No

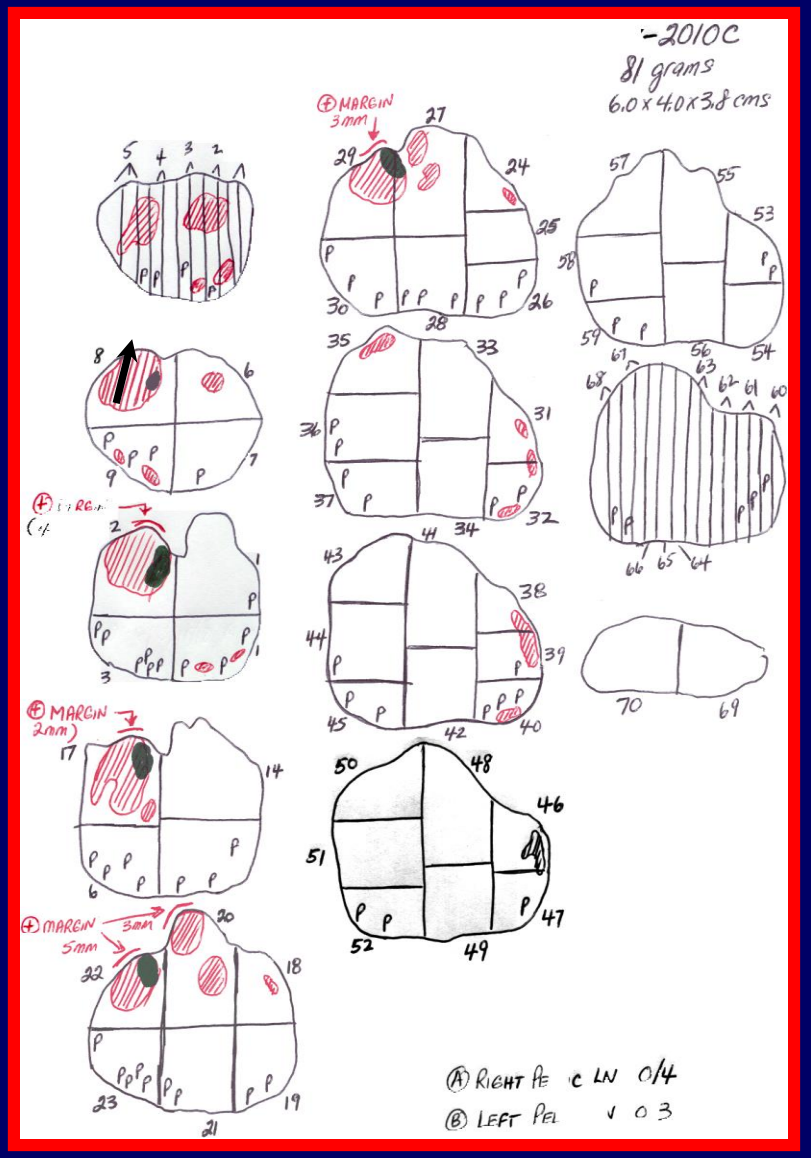
How much information can be gleaned from this limited sample to optimize management ?

Prostate biopsy: General considerations

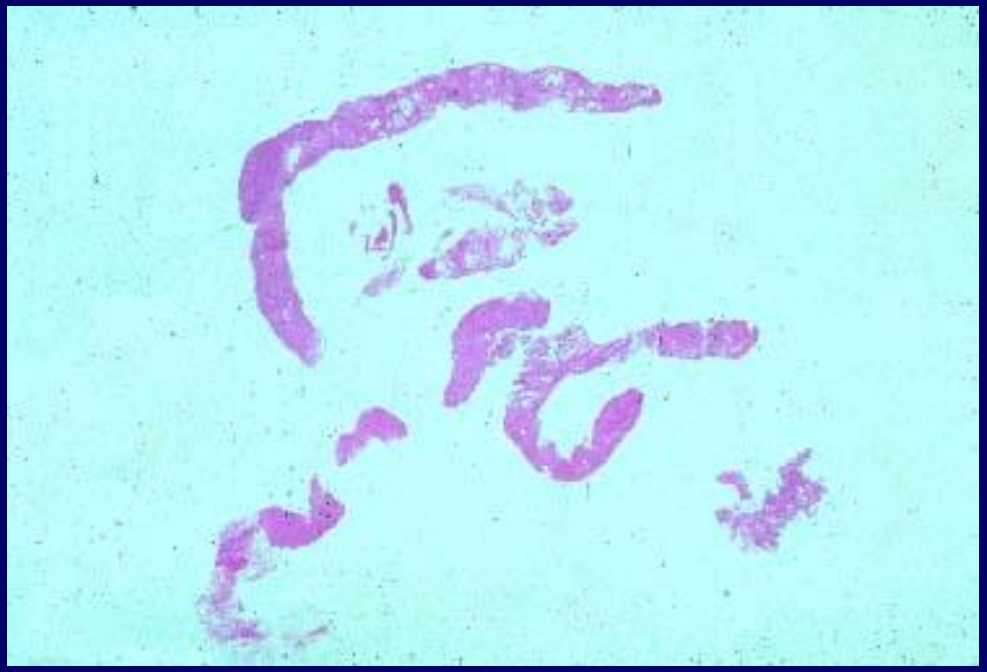
A biopsy set represents on average 1/500 –1/1000 of the total gland volume

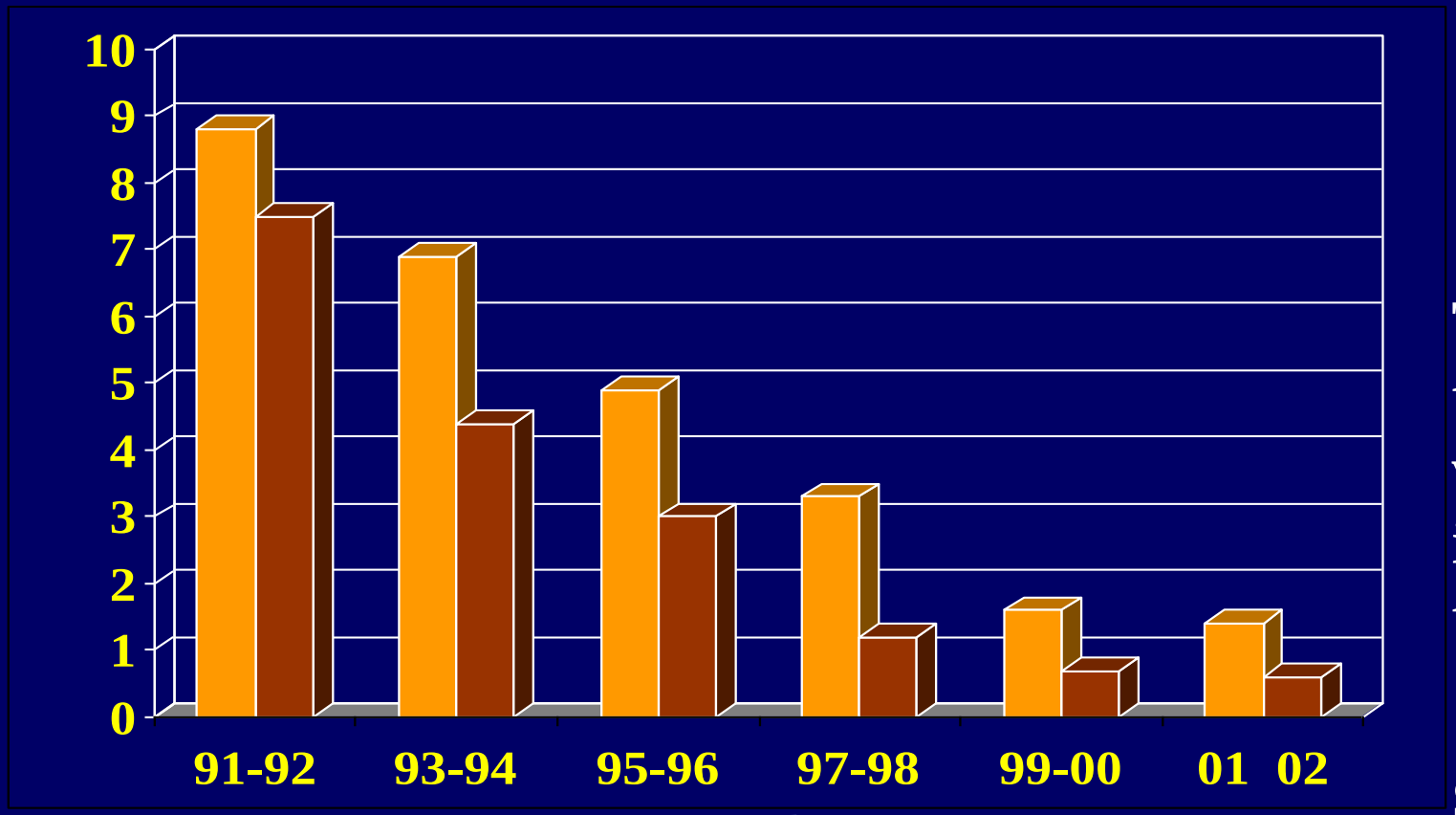
Biopsies are NOT obtained under direct vision

Prostate cancer is a multifocal disease with variably sized tumor foci, often of different grades



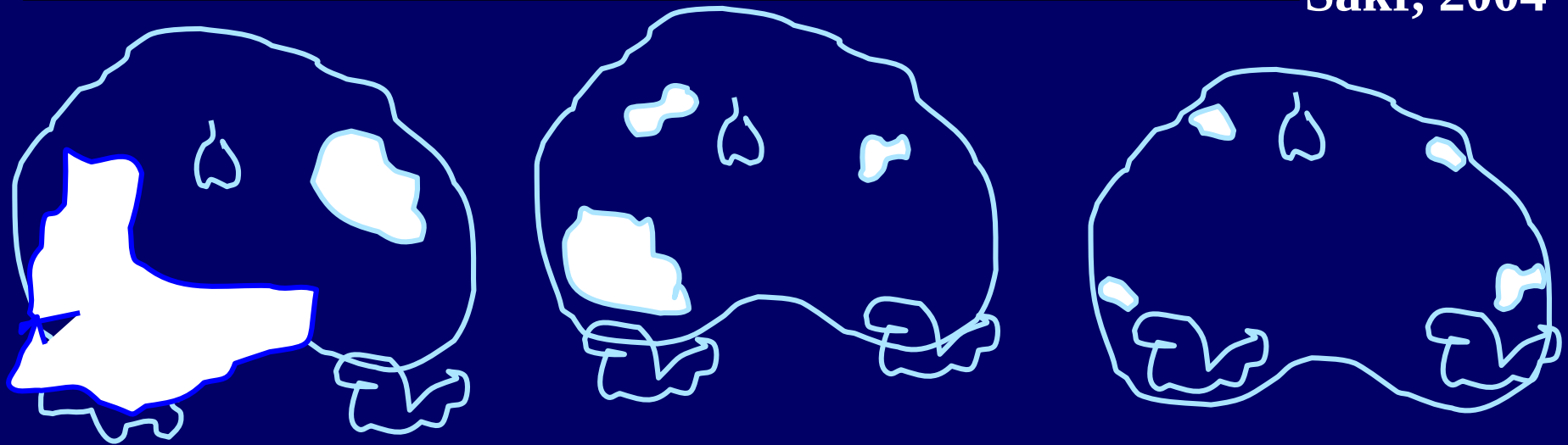
Prostate Needle Biopsy: Maximizing Important Information



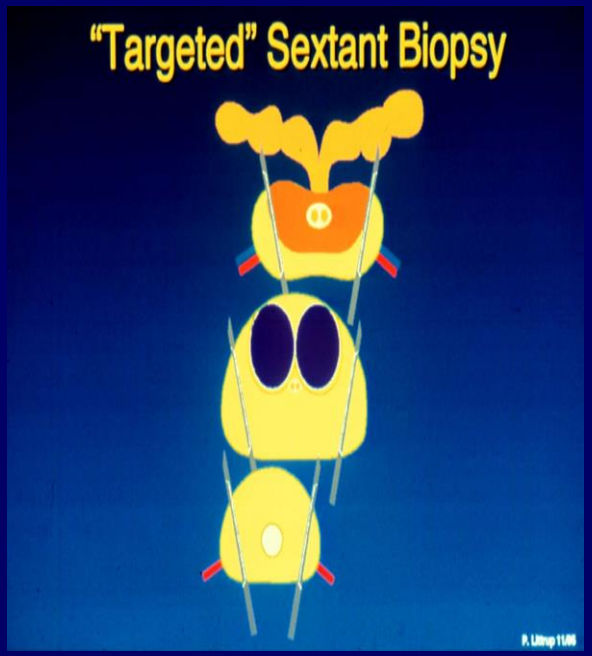


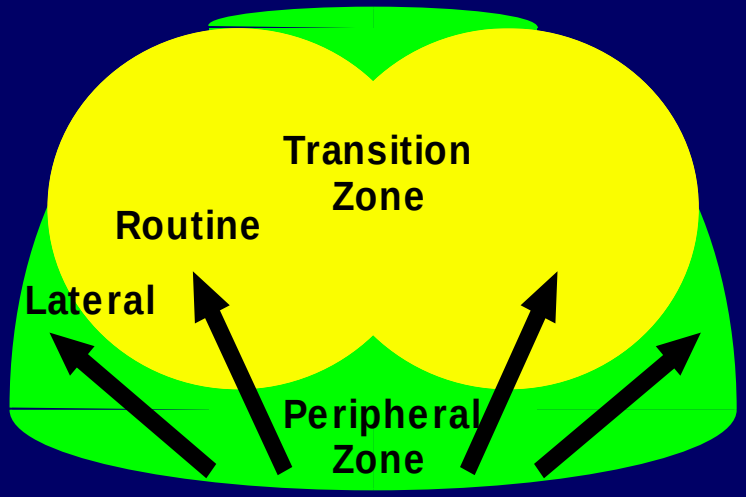
Total tumor volume & index tumor 1991-2002

Sakr, 2004



Evolving Bx approach: Improved cancer detection





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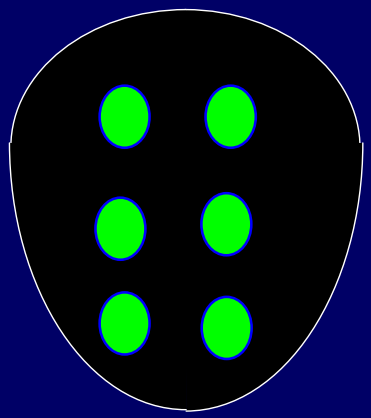
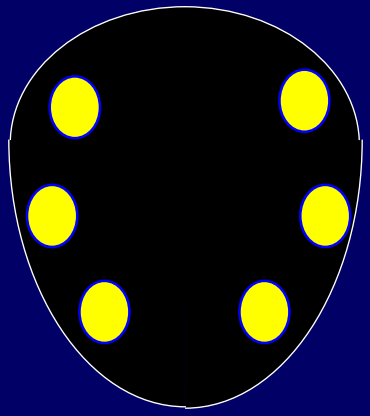
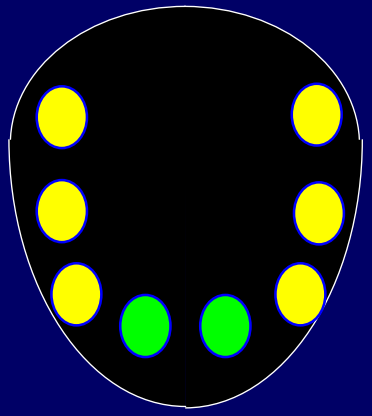


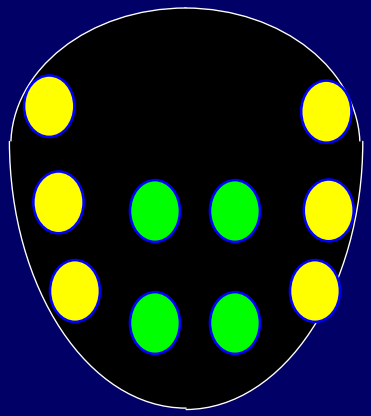
83%



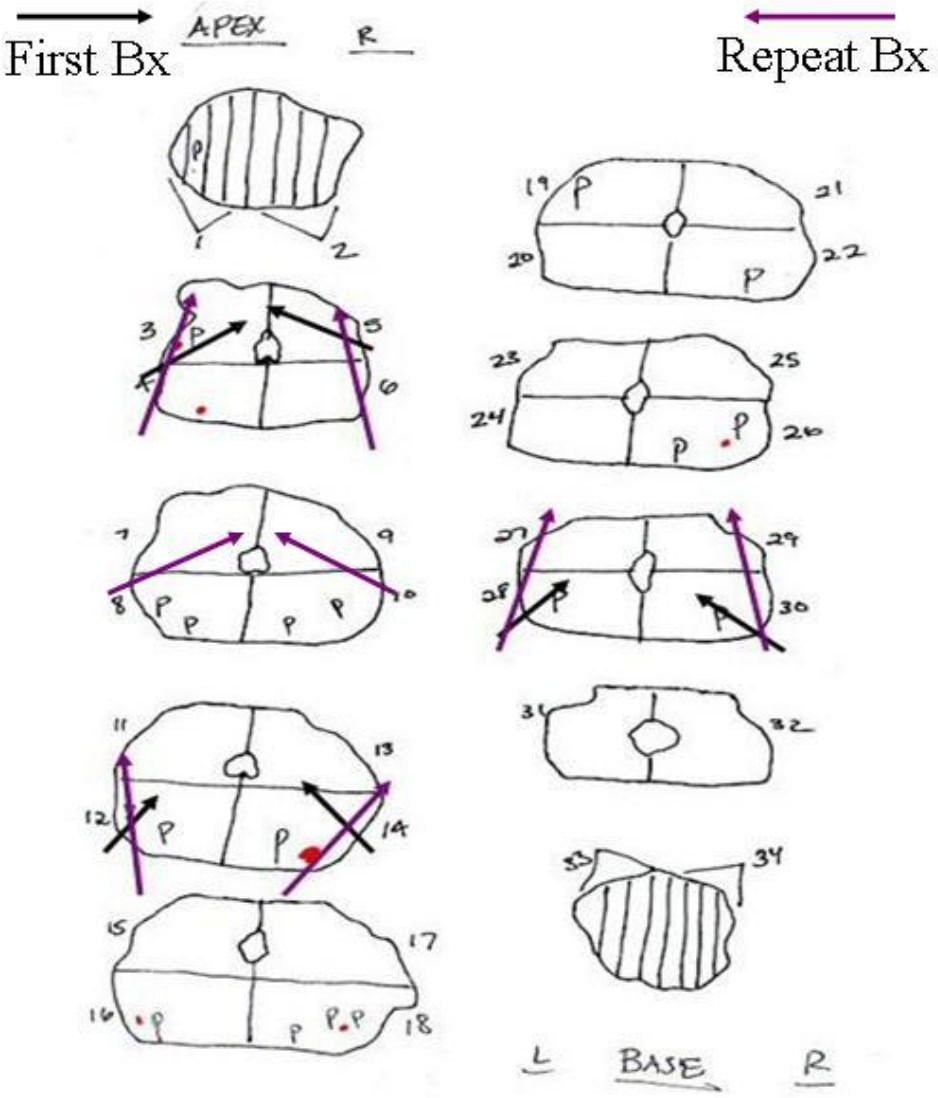
92%



96%



Presti JC Jr, et al. J Urol. 2003;169:125-9.


First Bx
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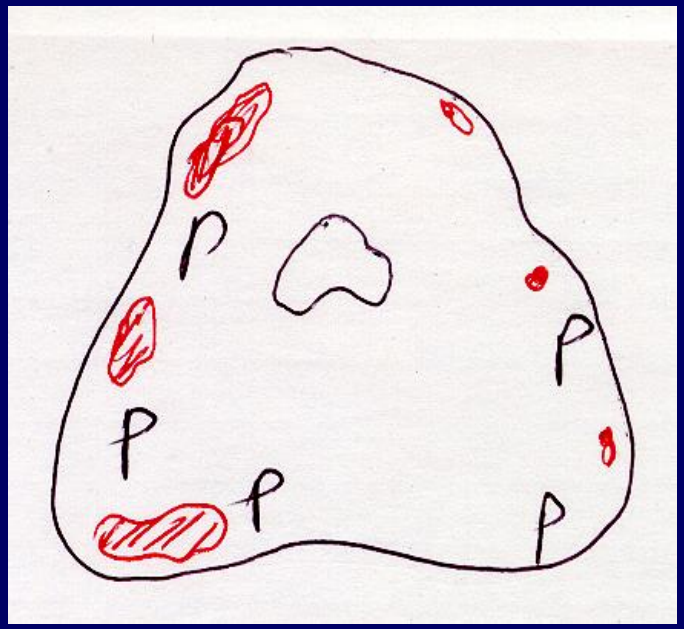
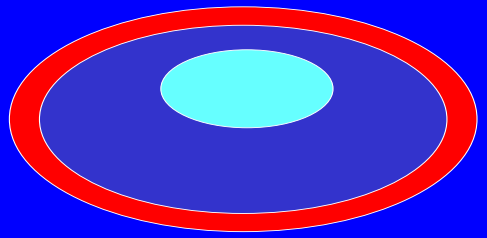
L
BASE
R

Early and small volume cancers are VERY peripheral !



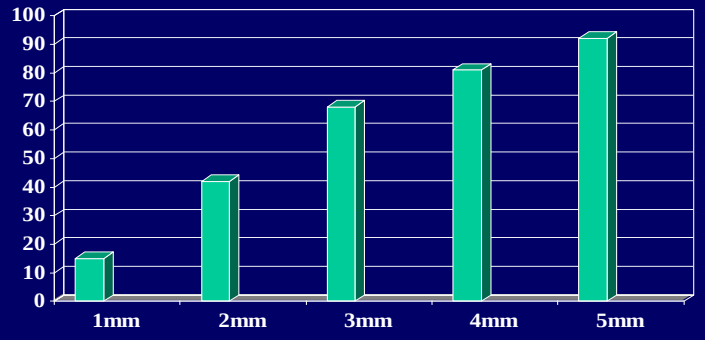
In 44% of patients, the ENTIRE Ca is located within the outer 5 mm



In 79%, the epicenter of the largest Ca focus is within the outer 5 mm.

Sub-clinical prostate cancer

Distance of small cancer foci from prostate capsule



Distribution of 149 tumor foci from 38 whole mount autopsy glands

Diagnostic Categories-Needle Biopsy

Benign Tissue

Atypical/Suspicious Foci

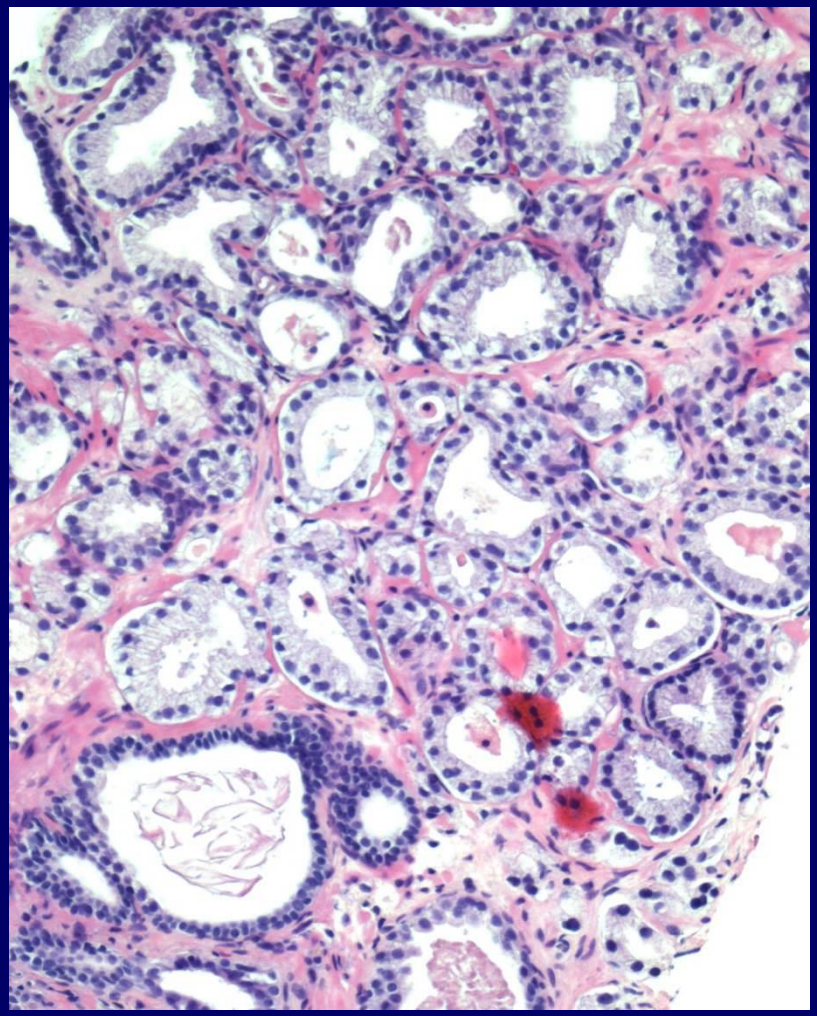
High Grade PIN

Prostatic Carcinoma

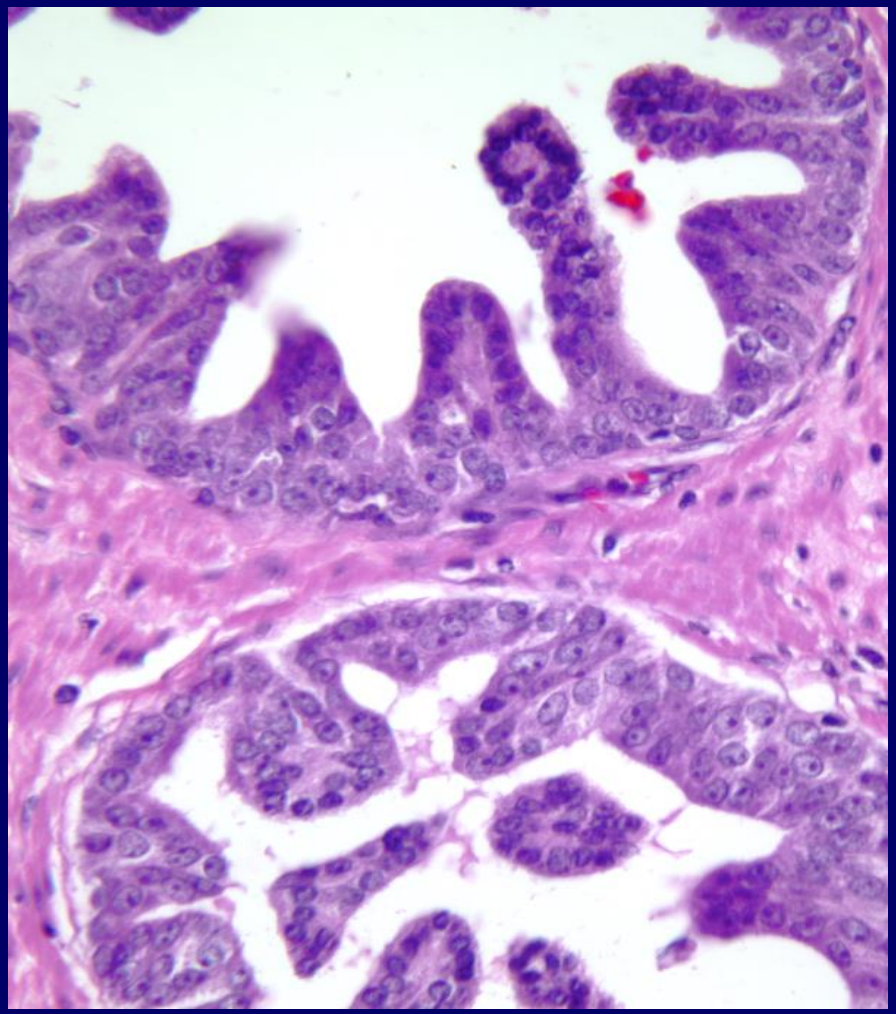
Atypical, Suspicious, and "Premalignant Lesions"

Histologic types and features of prostatic "dysplasia."

Atypical lesions of prostatic epithelium



Acinar Proliferation

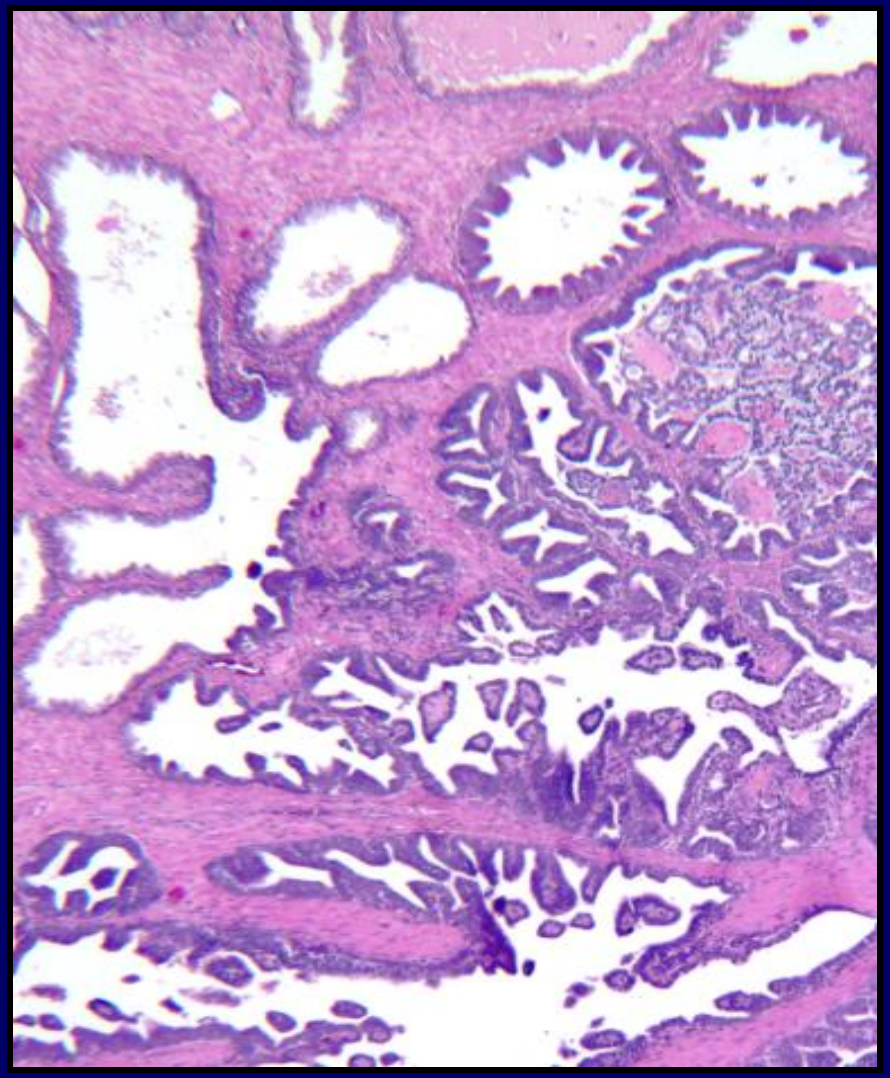


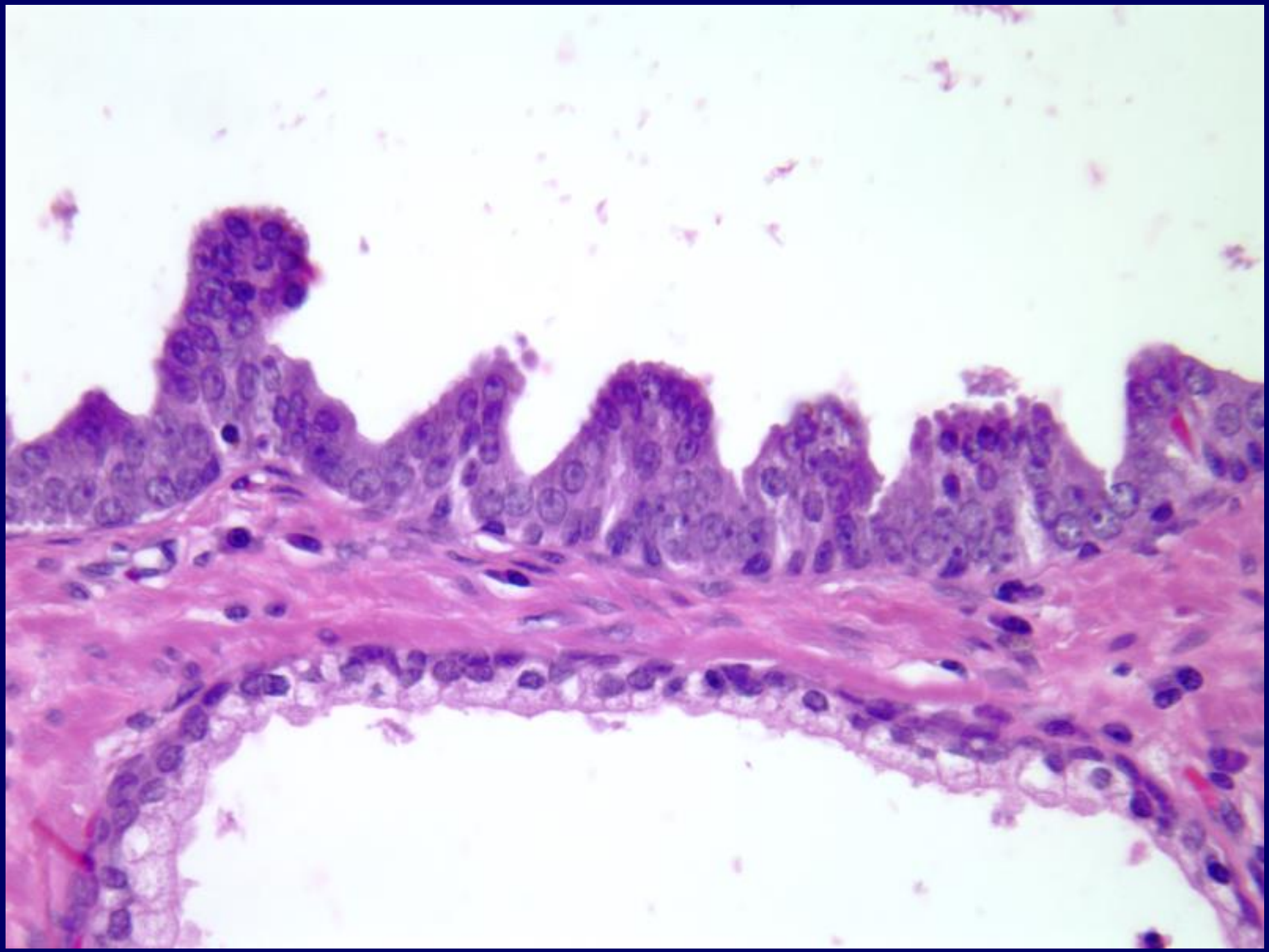
Cytologica Atypia

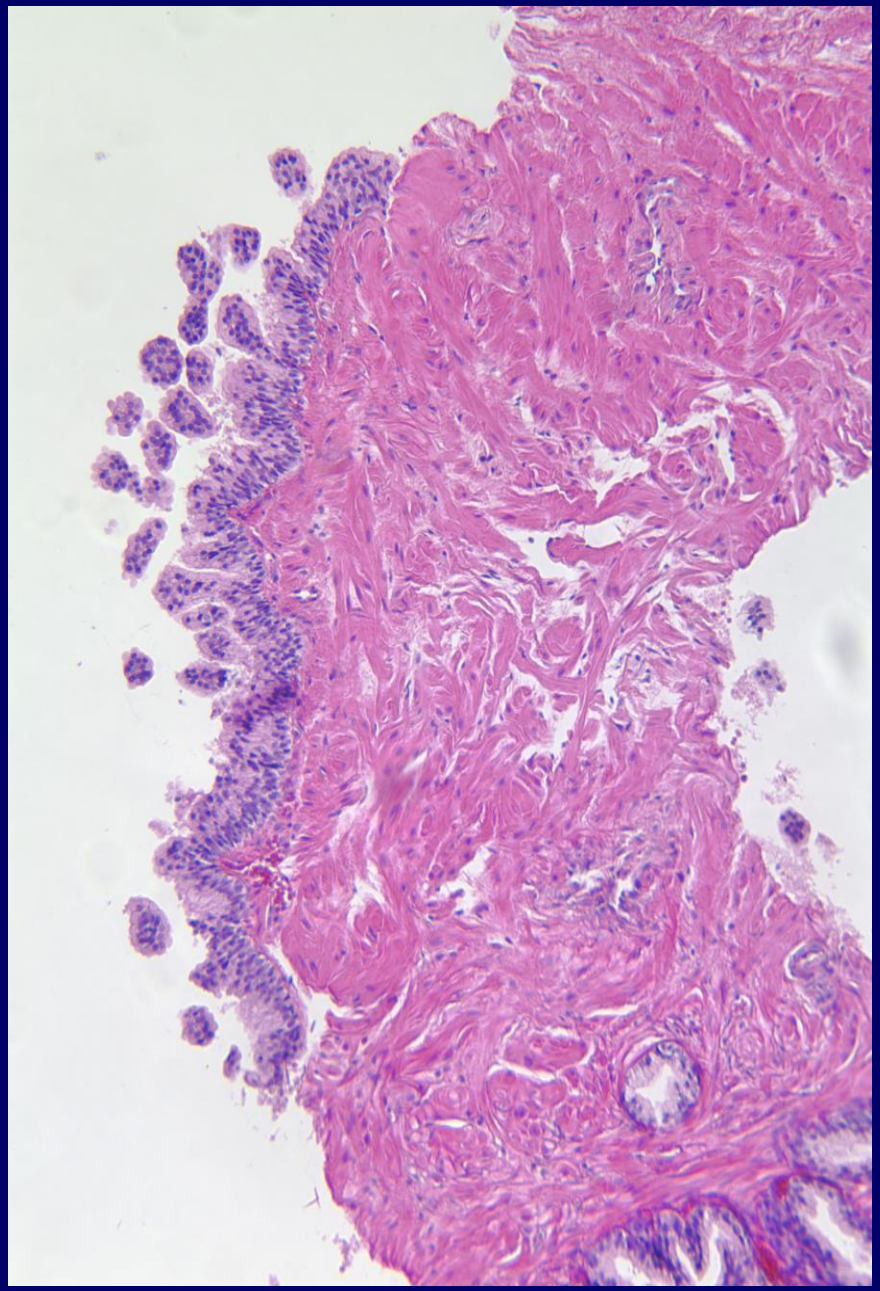
PROSTATIC INTRAEPITHELIAL NEOPLASIA

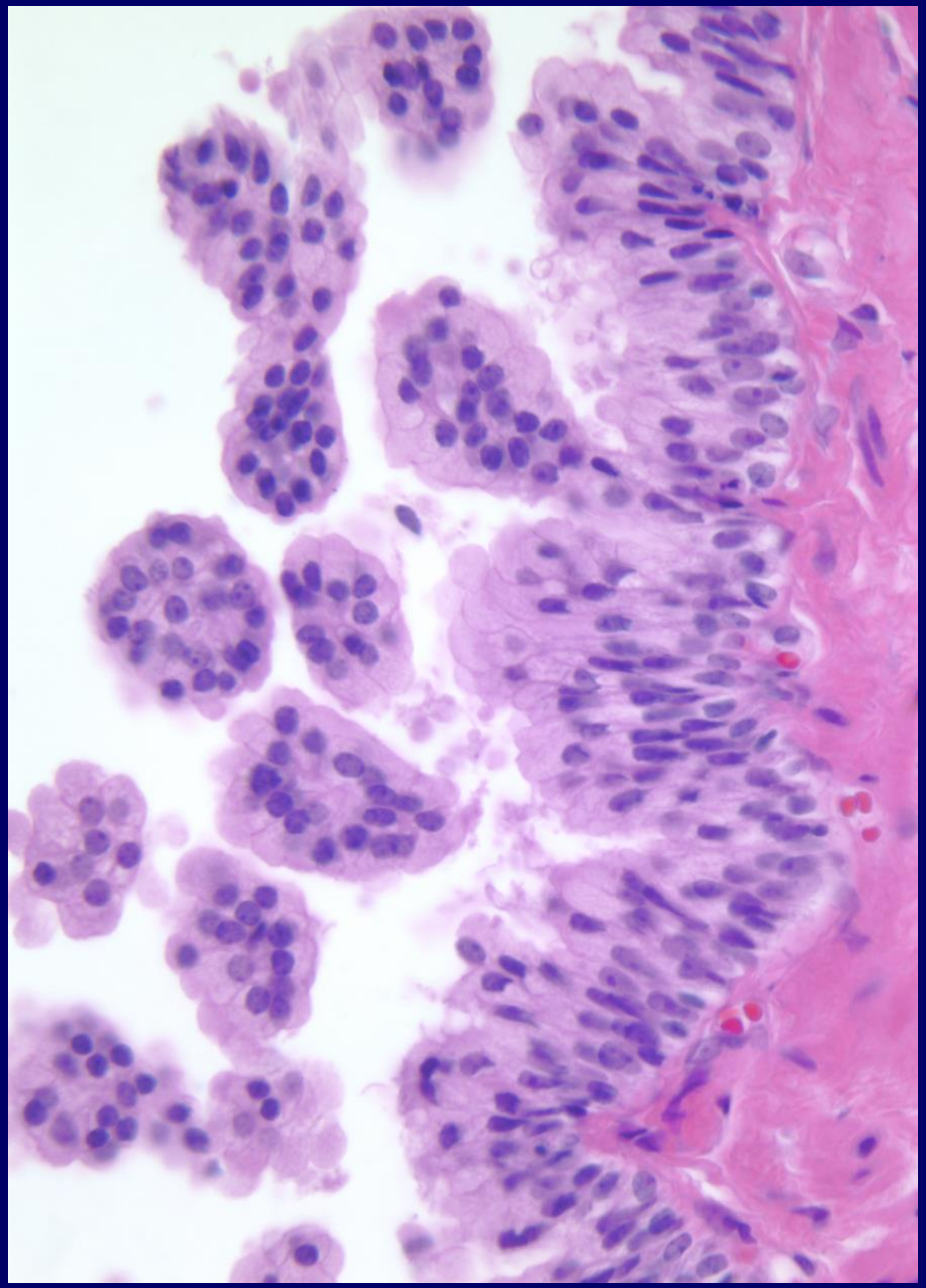
PATHOLOGY

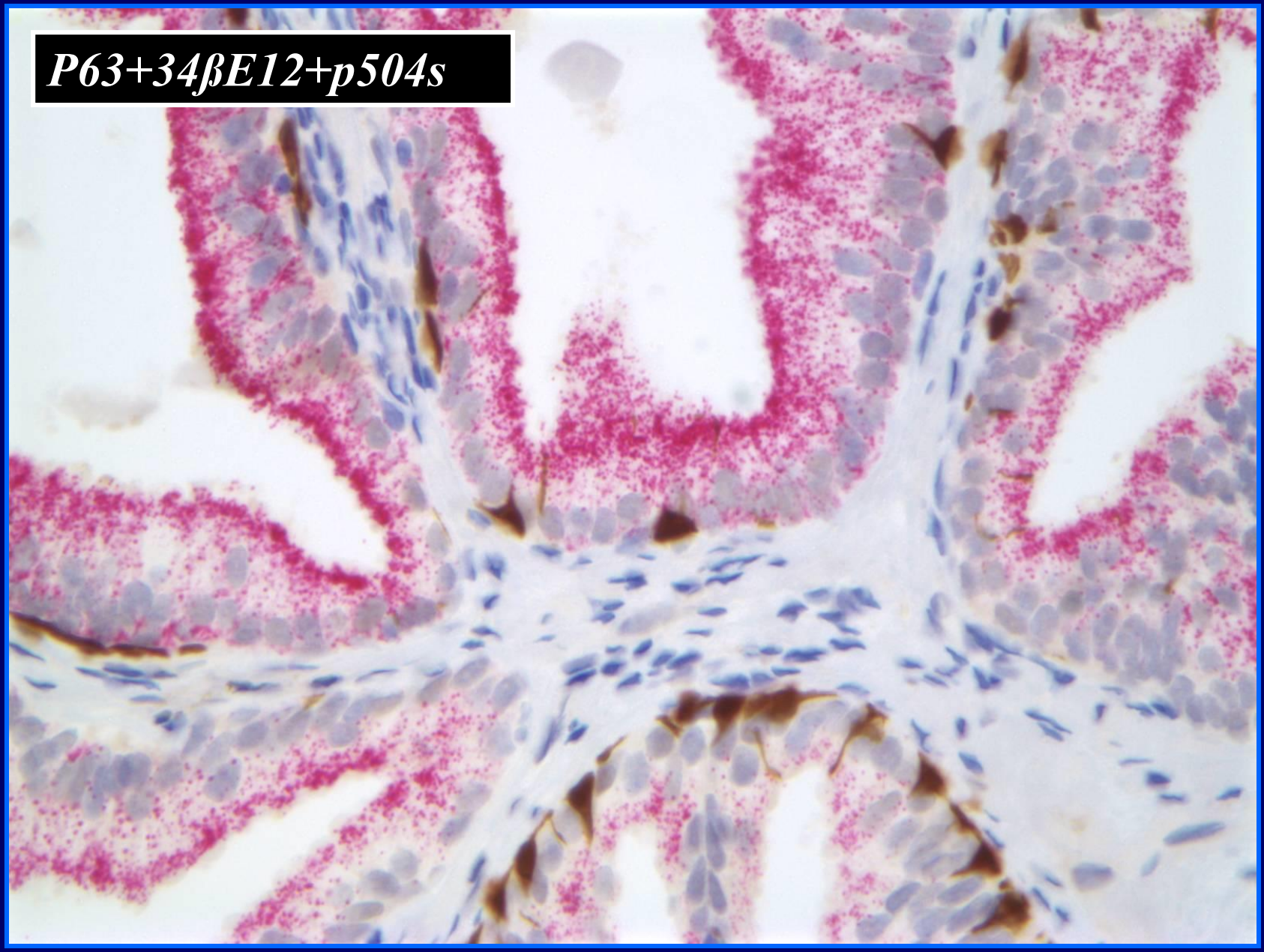
- Complex “blue” appearance on low power
- Cellular proliferation with tufted, papillary or cribriform architectures
- Cells have large nuclei with prominent nucleoli
- Amphophilic cytoplasm





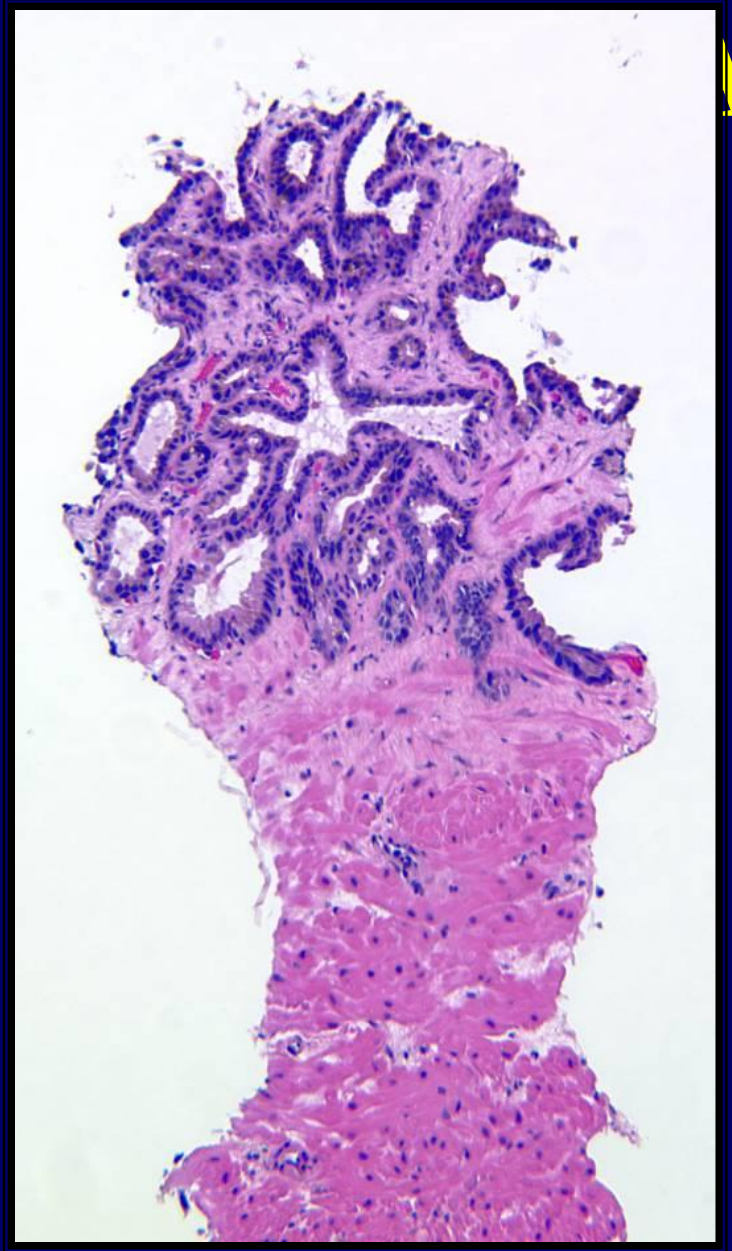


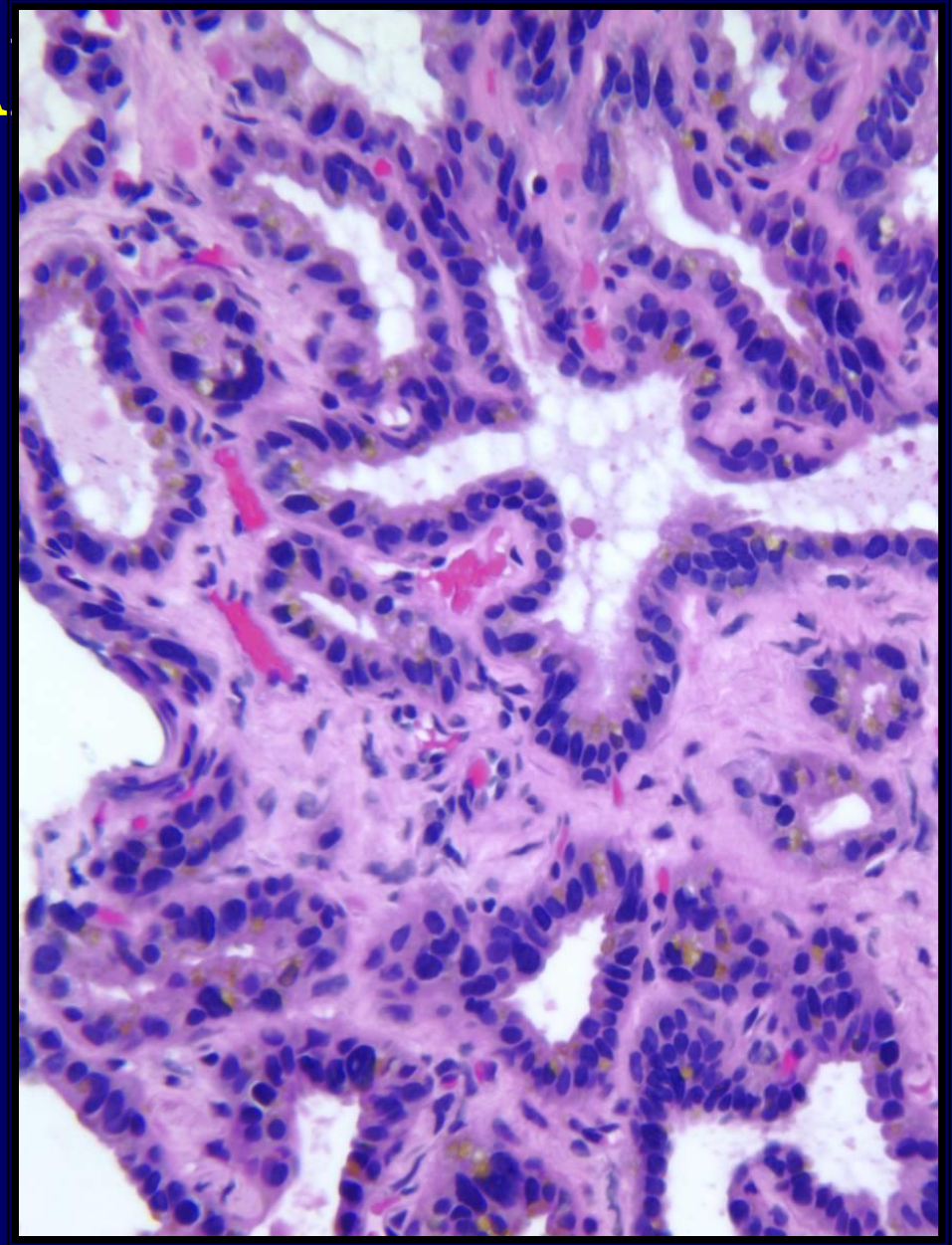



P63+34ßE12+p504s

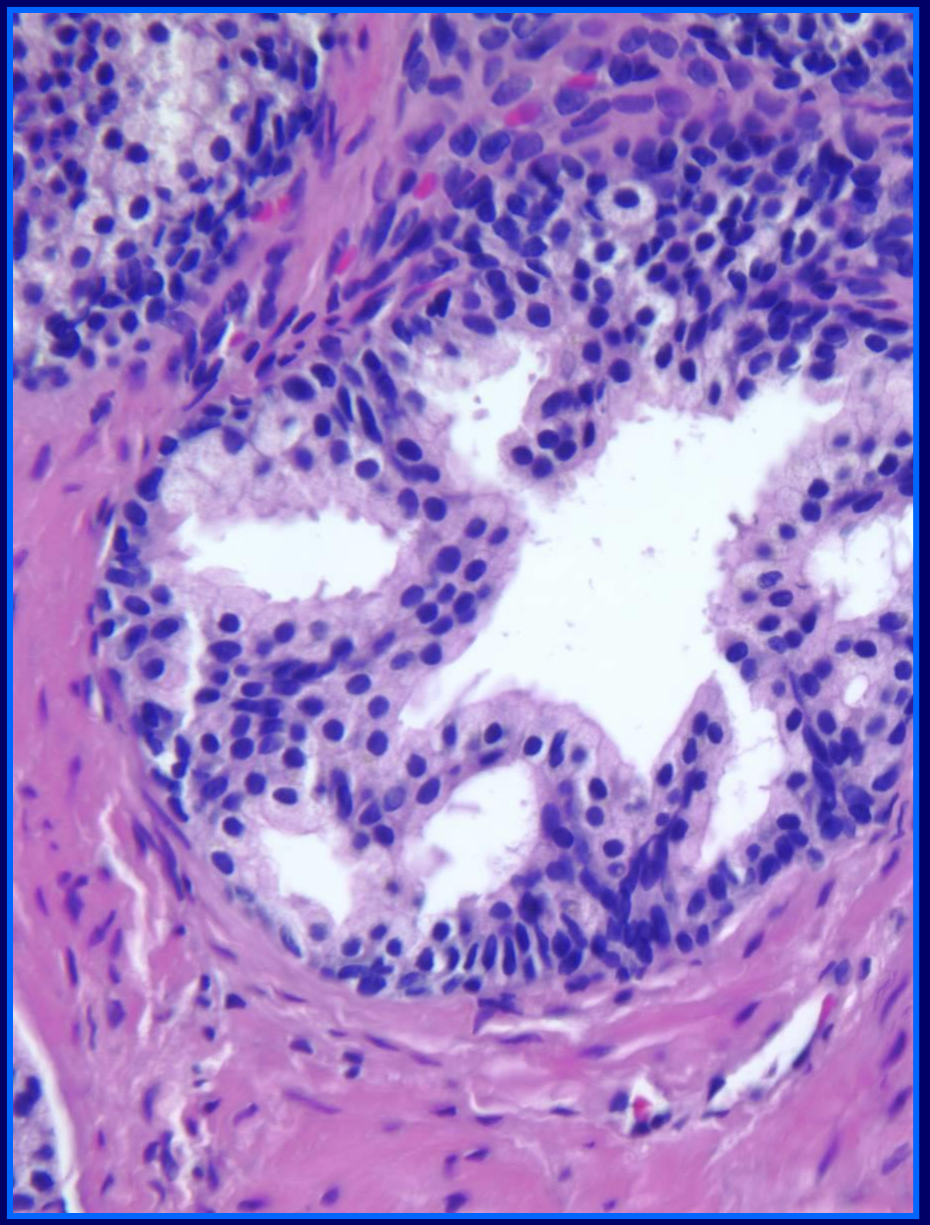
DIFFERENTIAL DIAGNOSIS

- Seminal Vesicle Epithelium
- Central zone
- Basal Cell Hyperplasia
- Transitional Cell Metaplasia
- Ductal Carcinoma
- Urothelial Carcinoma

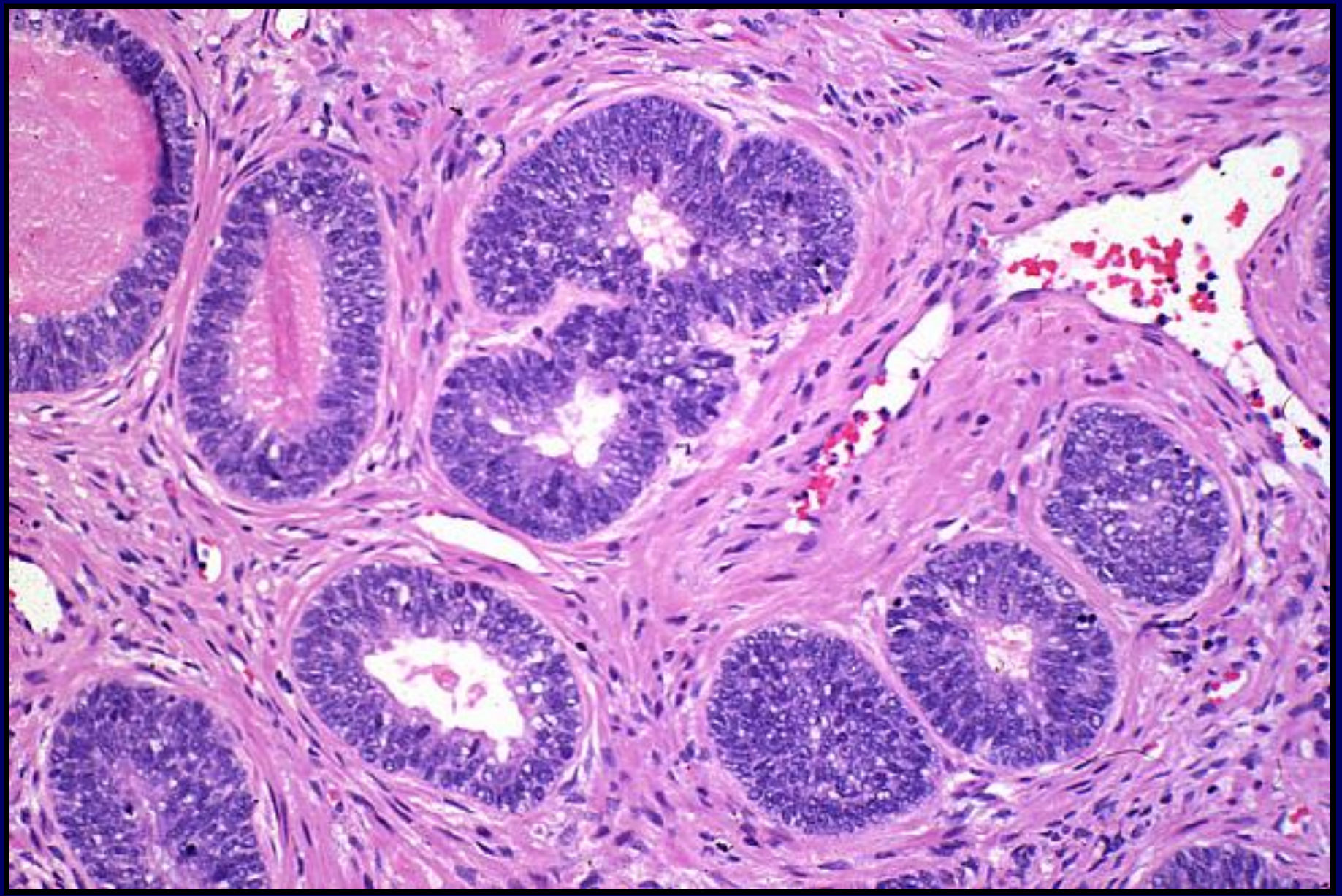




CENTRAL ZONE



BASAL CELL HYPERPLASIA



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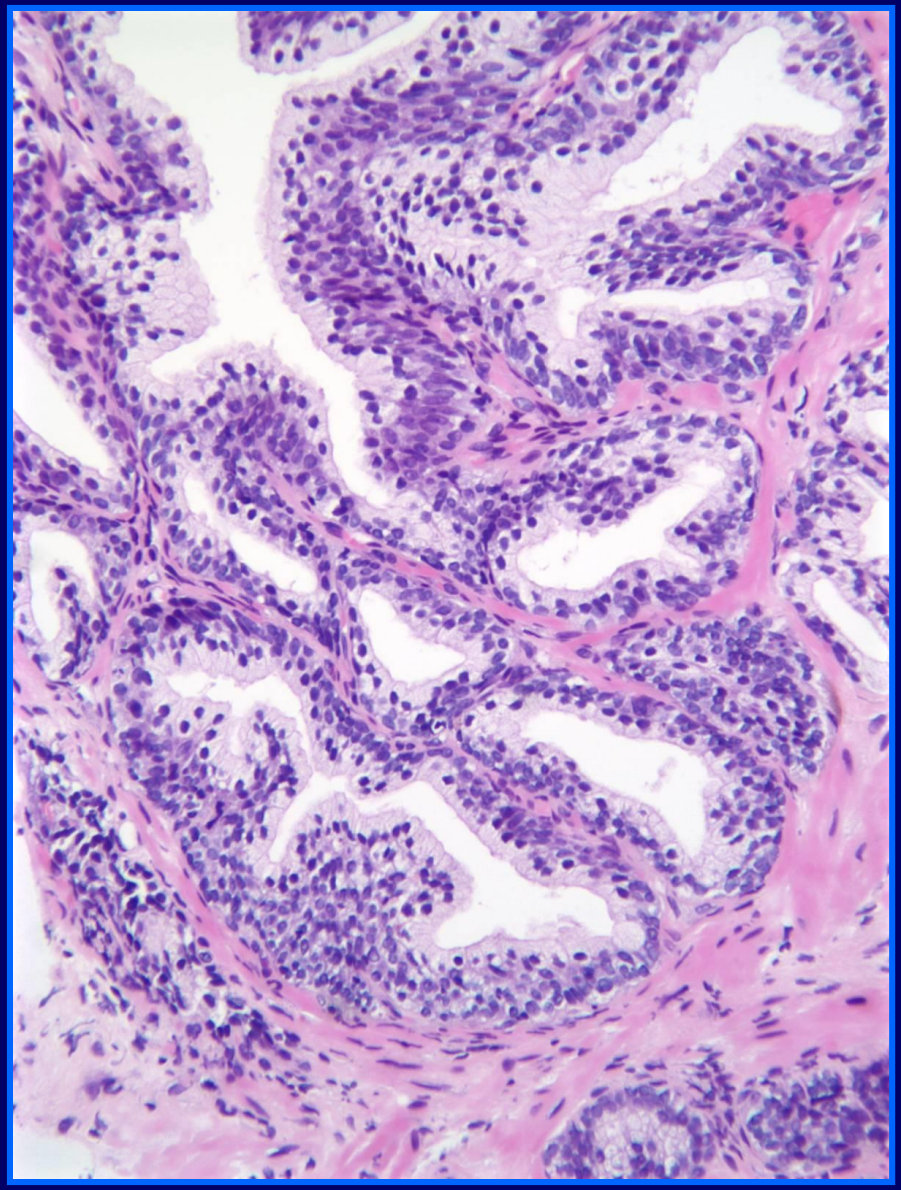
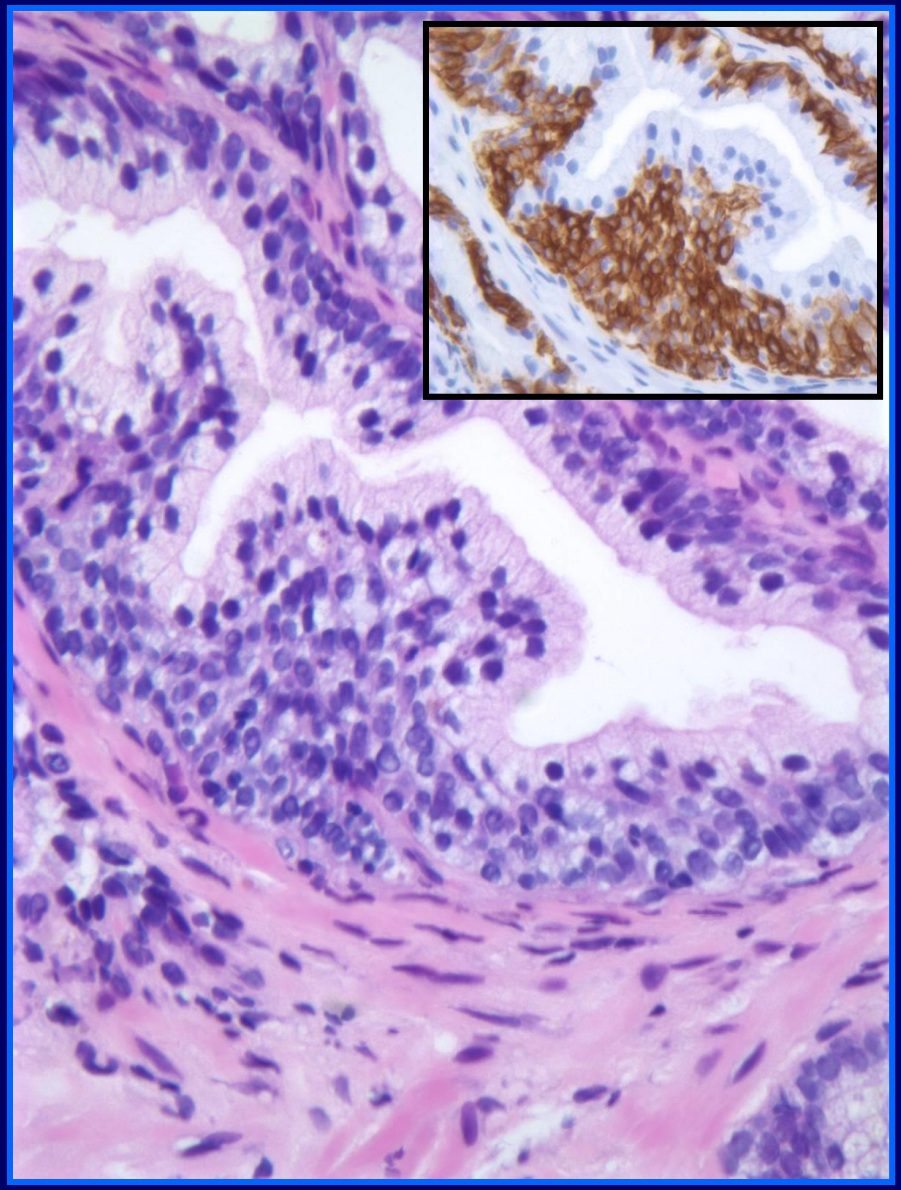
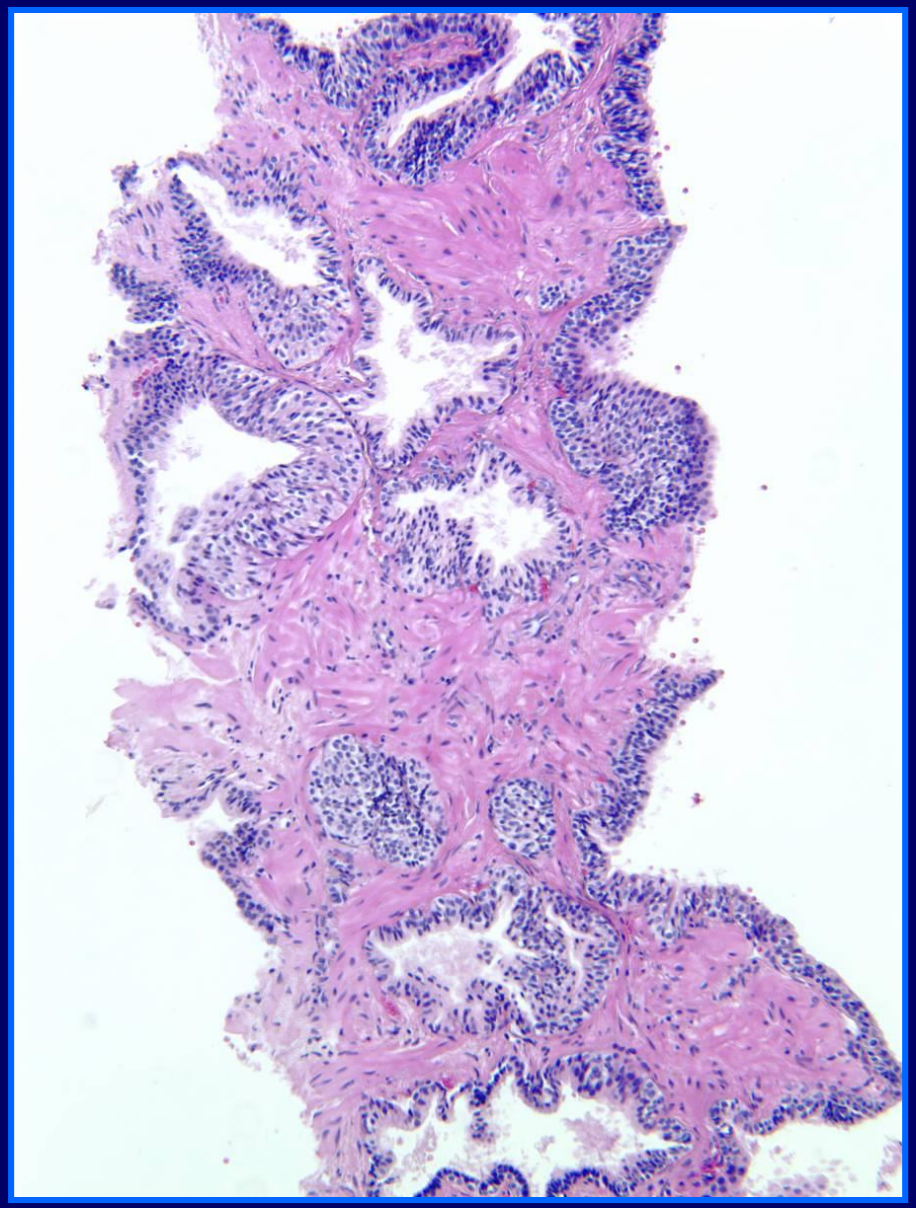
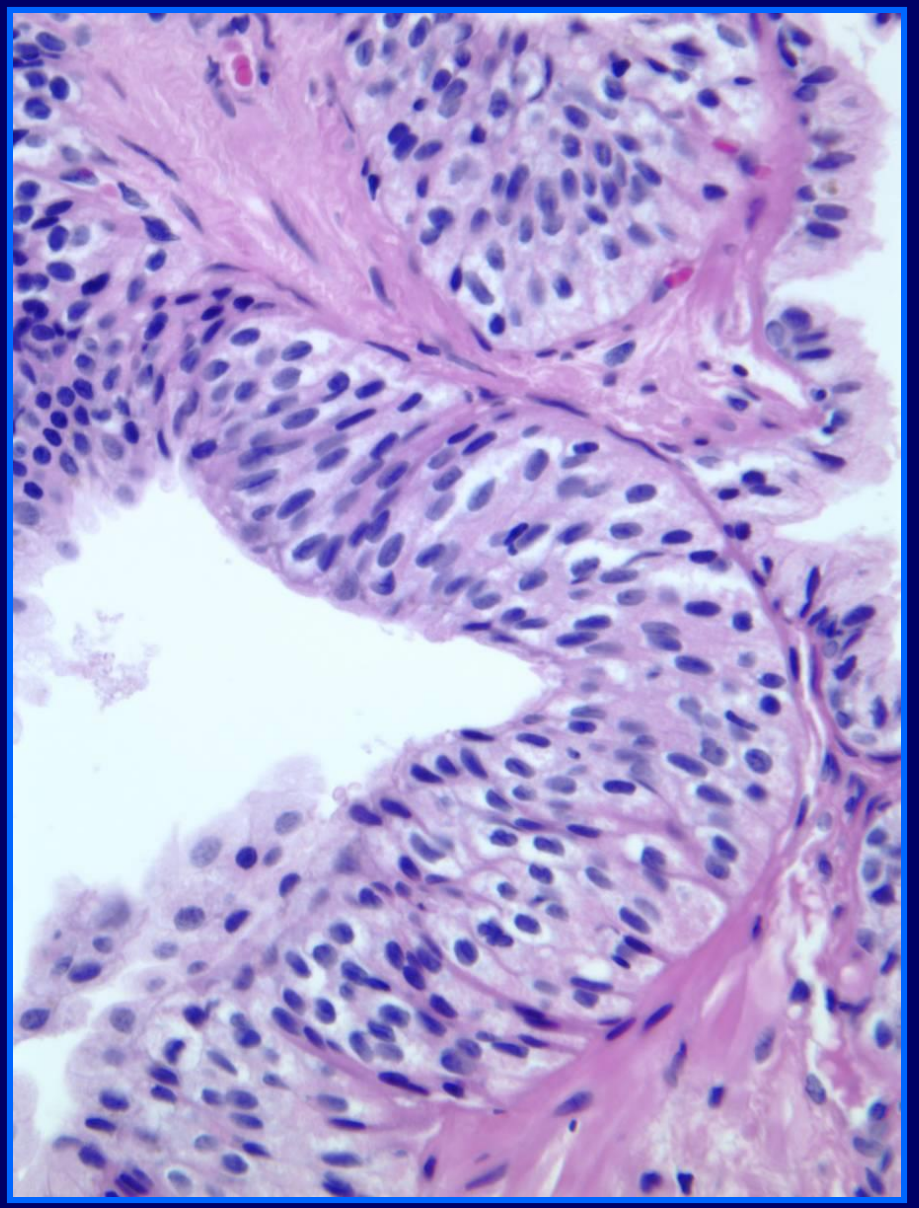


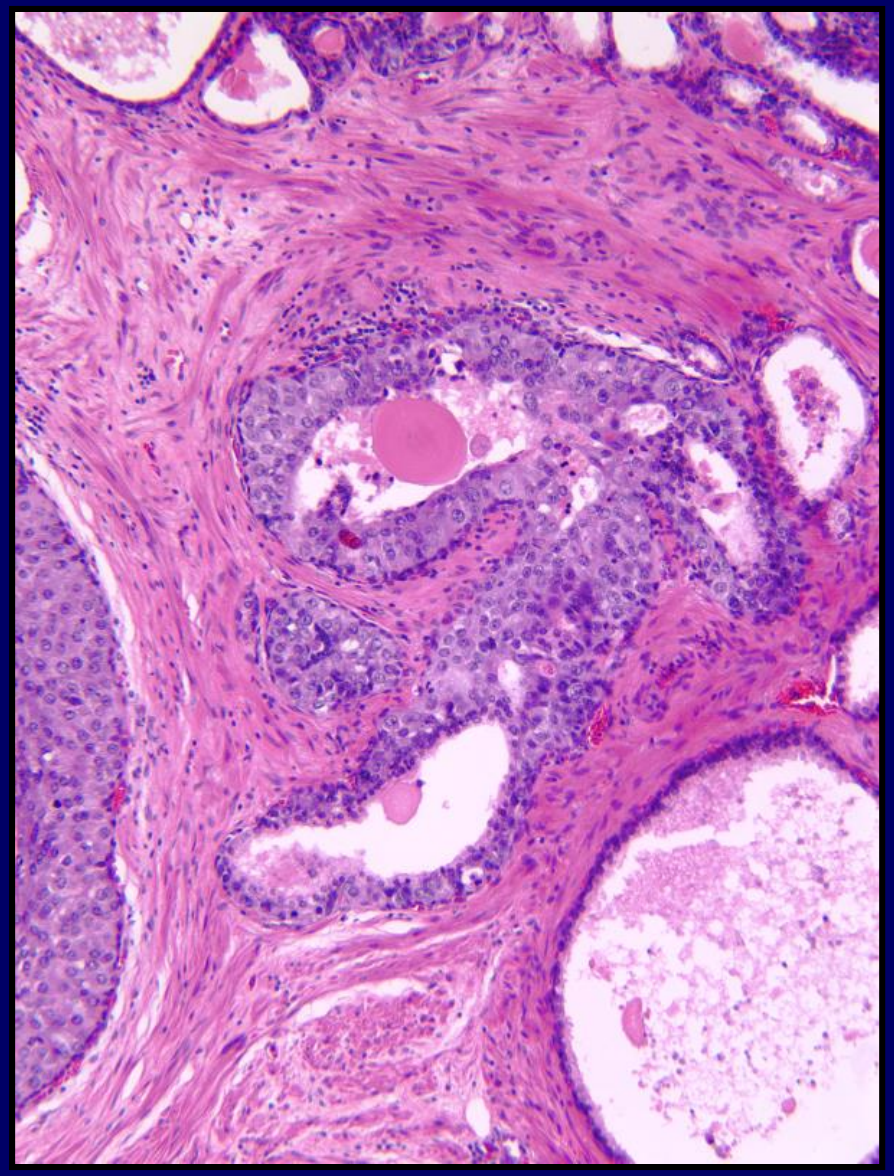


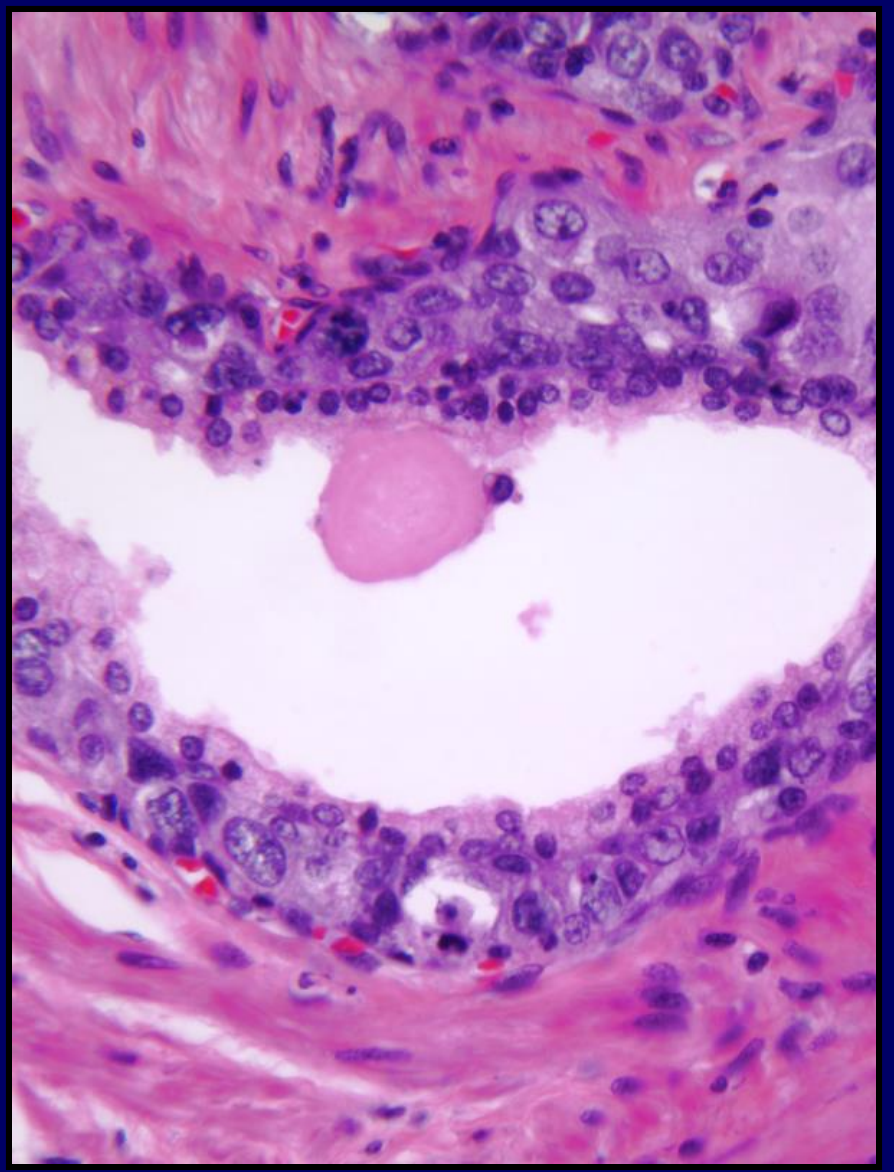
TRANSITIONAL CELL METAPLASIA





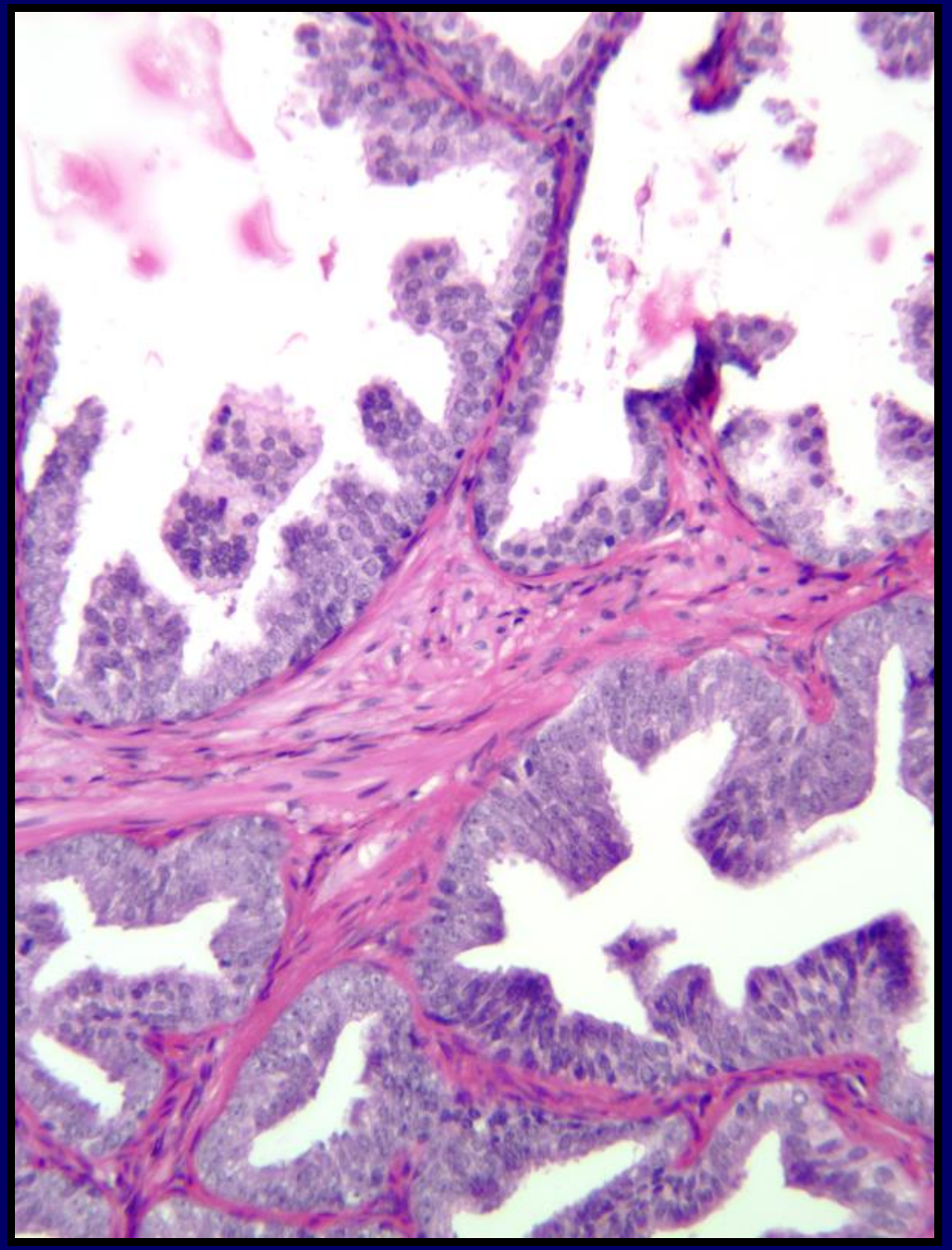
PROSTATE
UROTHELIAL CARCINOMA





PIN - *Clues to Diagnosis*

- **Pre-existing structures**
- **Complex blue glands**
- **Stratification of epithelium**
- **Cytologic atypia**



Trends in Predicting Cancer Following Diagnosis of HGPIN

Study	Patients with HGPIN in First Biopsy	PCa in Second Biopsy (%)
Kronz, 2001	245	32.2
Gokden, 2003	221	28.1
Sakr, 2003 (*time trend*)		
• 1993–1998	101	38
• 1999–2007	540	27
Hoedemaeker, 1999	1,824	21.1
Schlesinger, 2003	23	22

Average: 26%

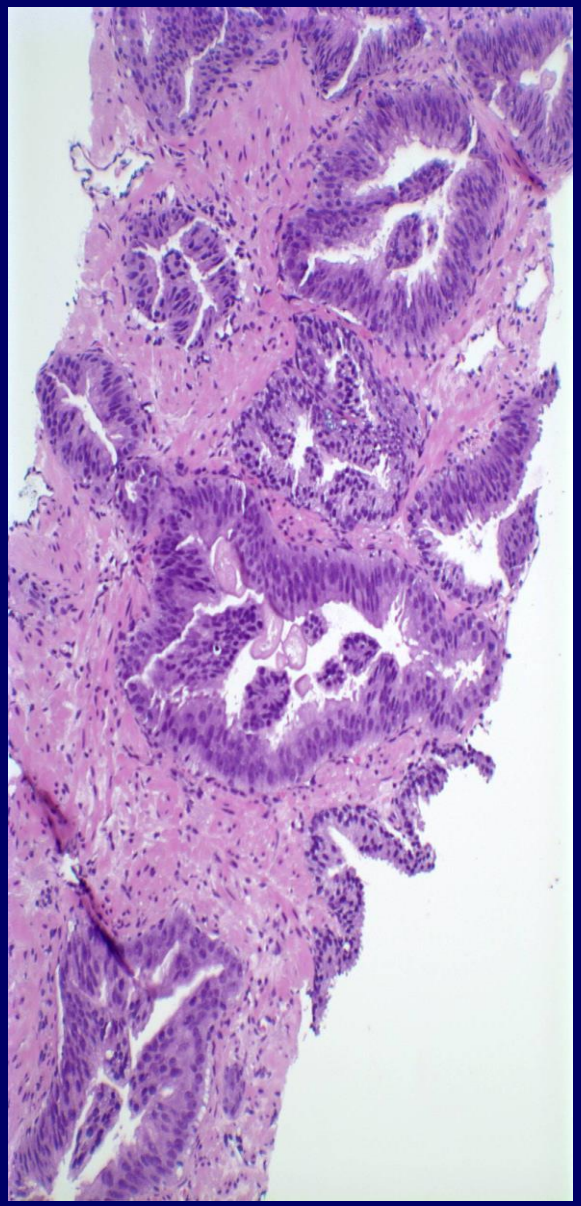
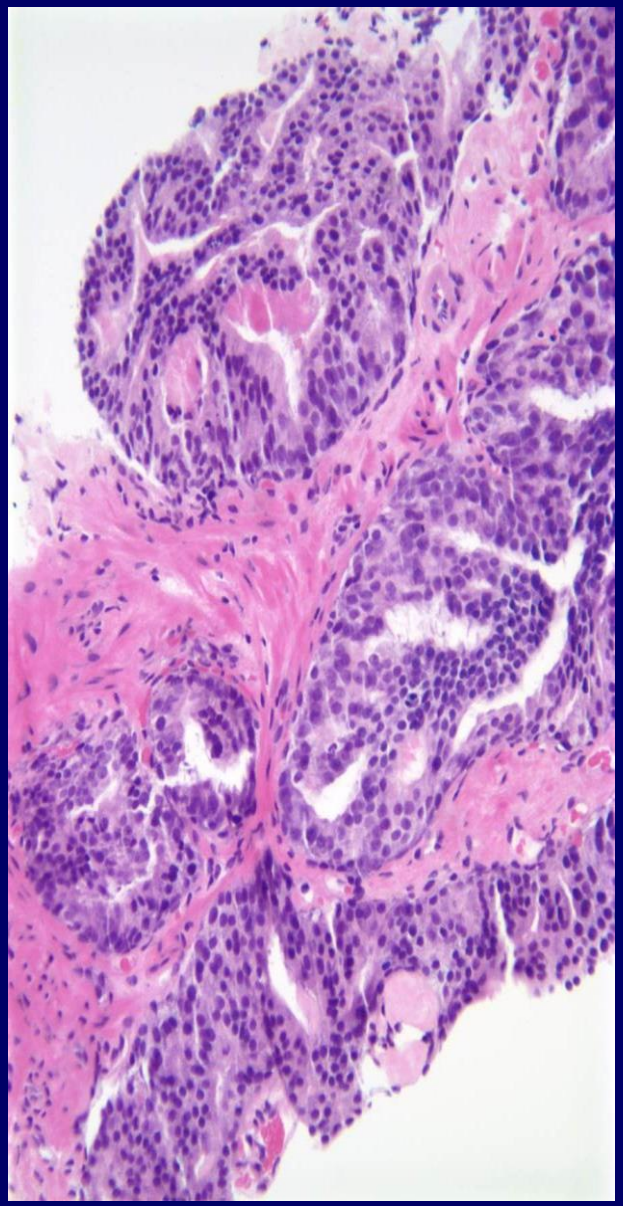
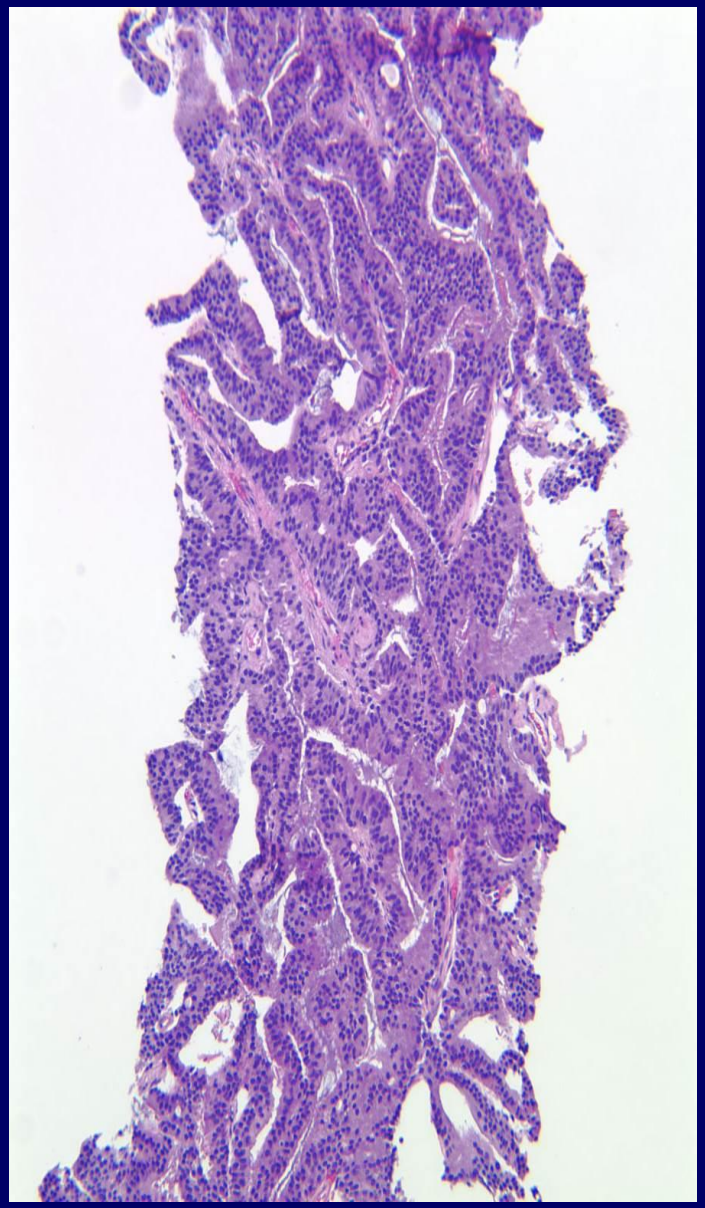
Widespread high-grade prostatic intraepithelial neoplasia on prostatic needle biopsy: a significant likelihood of subsequently diagnosed adenocarcinoma

39% Cancer

9.5% HGPIN with adjacent atypia

Netto & Epstein

Am J Surg Pathol. 2006; 30:1184-8.

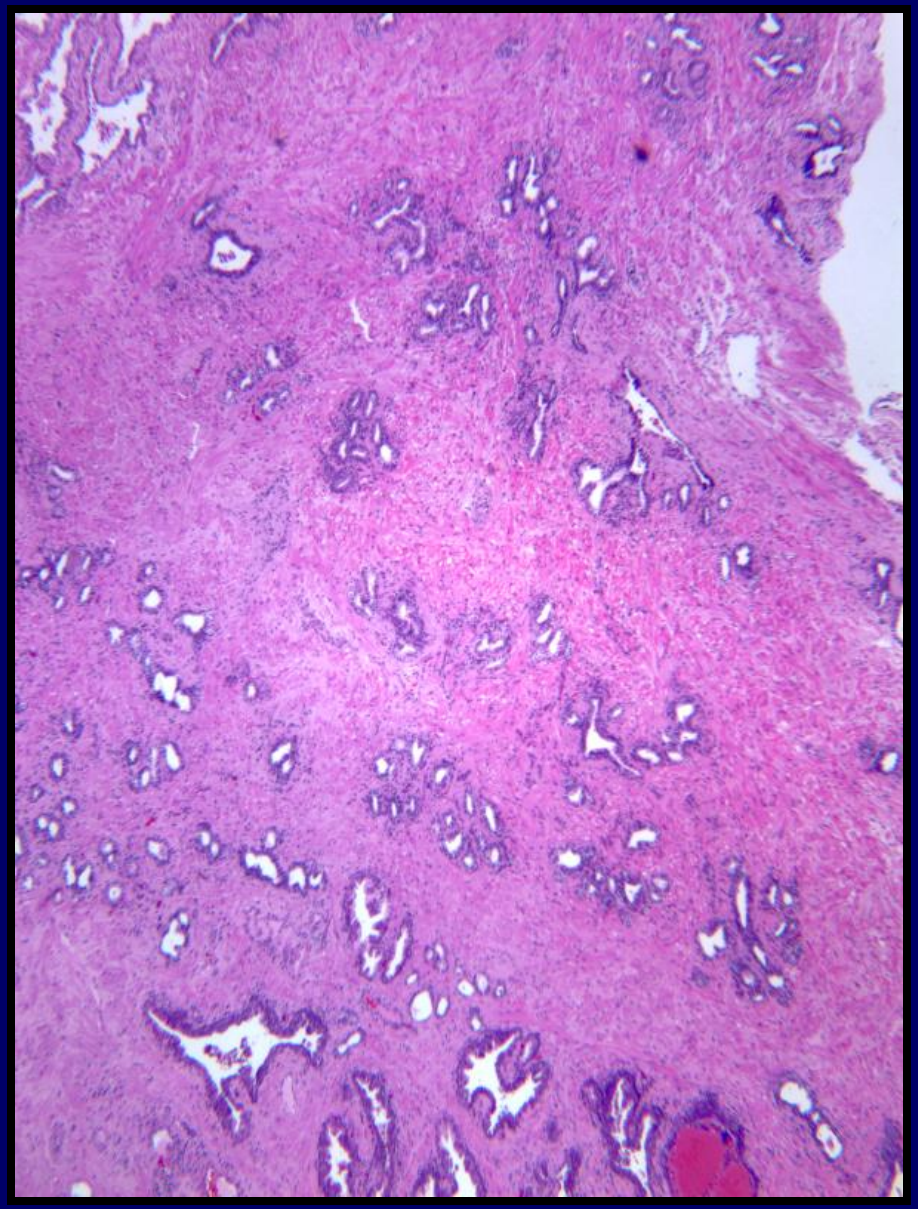
Small Acinar Lesions

PROSTATIC ATROPHY

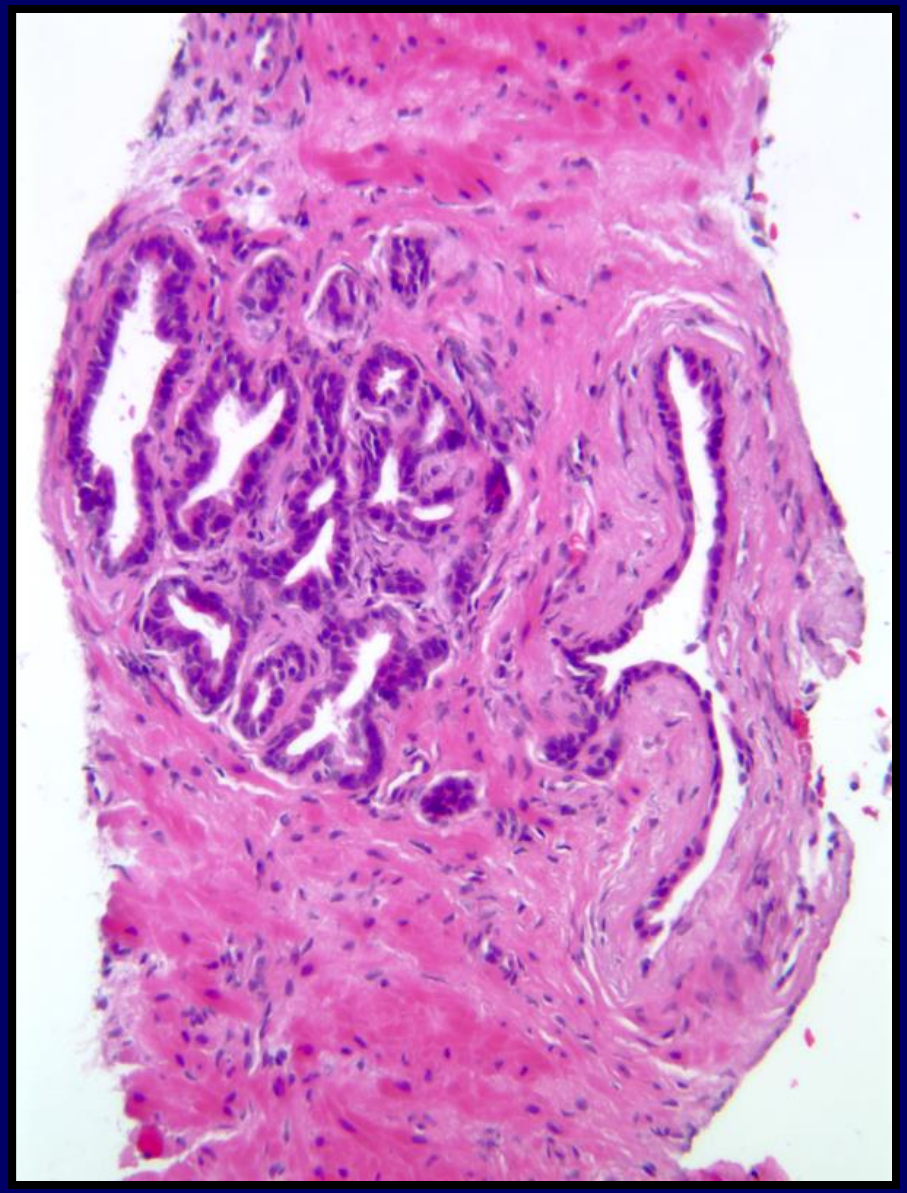
- Foci appear as early as 3rd decade
- Associated with prior inflammation
- May be side by side with BPH
- Clinical significance:
  - Can mimic CA on US (hypoechoic)
  - Can mimic CA on histology
- Three histologic patterns: lobular, sclerotic and postatrophic hyperplasia

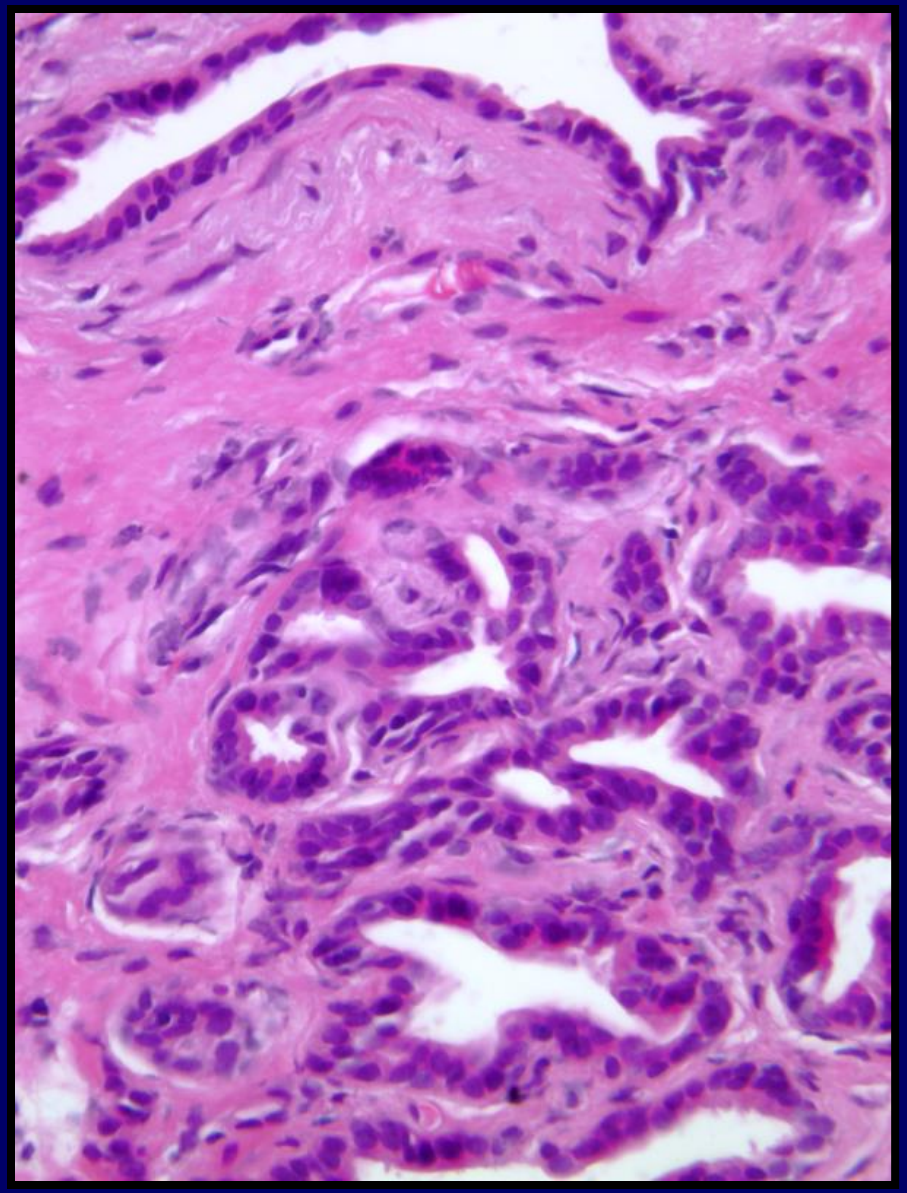
SIMPLE LOBULAR ATROPHY

- Circumscribed groups of glands
- Glands are small
- Some are angulated
- Central dilated duct
- Epithelium appears “blue”
- Stromal fibrosis
- +/- inflammation



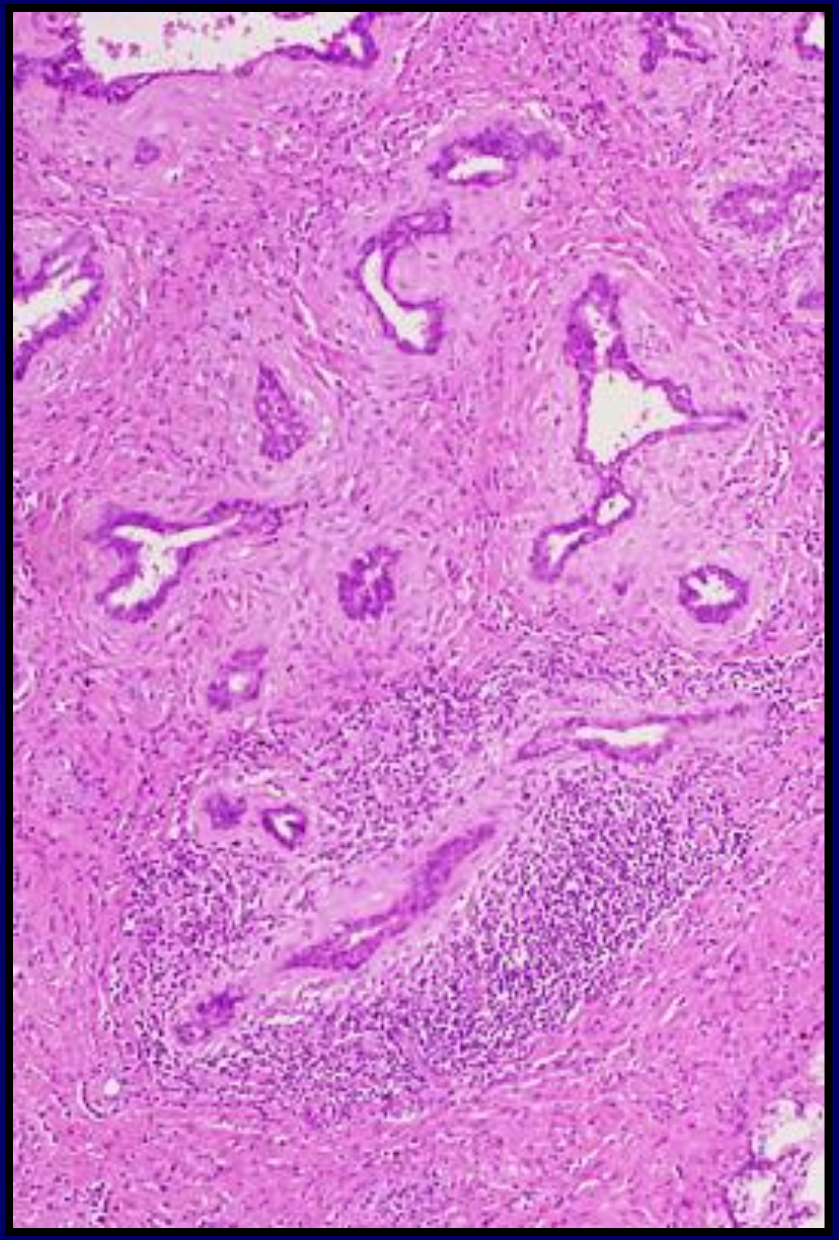
SIMPLE LOBULAR ATROPHY





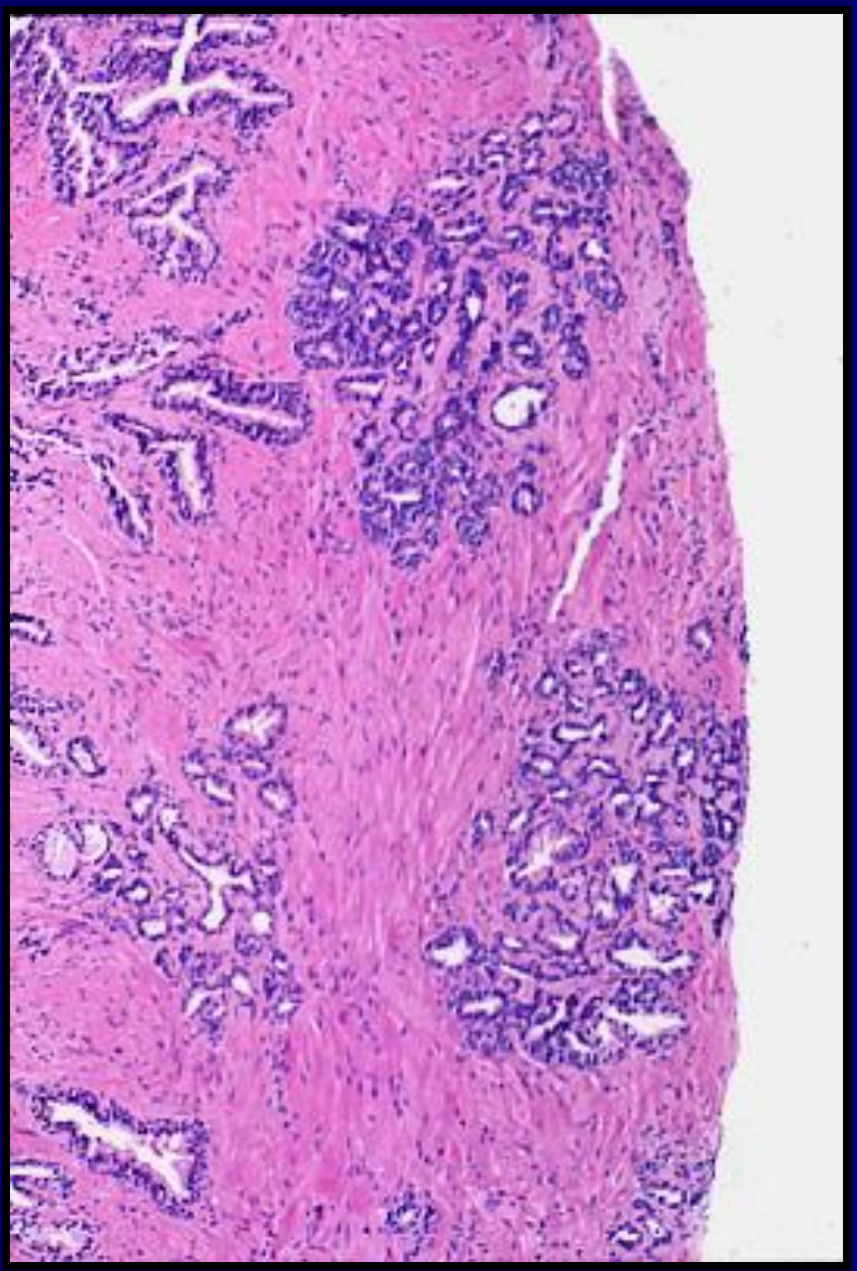
SCLEROTIC ATROPHY

- Lobular architecture
- Irregular angulated glands
- Dense, pink fibrotic stroma
- Central dilated ducts
- Atrophic epithelium (scant cytoplasm)
- +/- inflammation

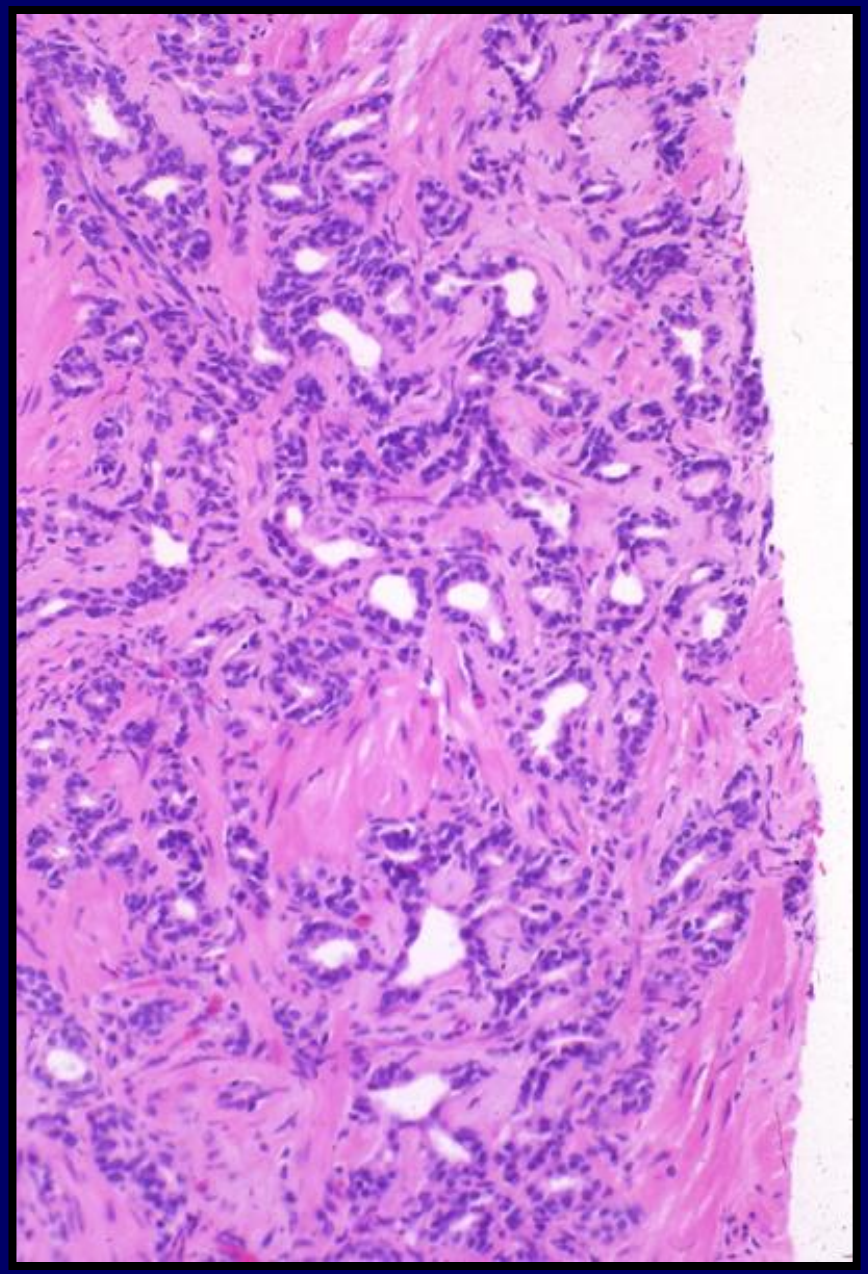


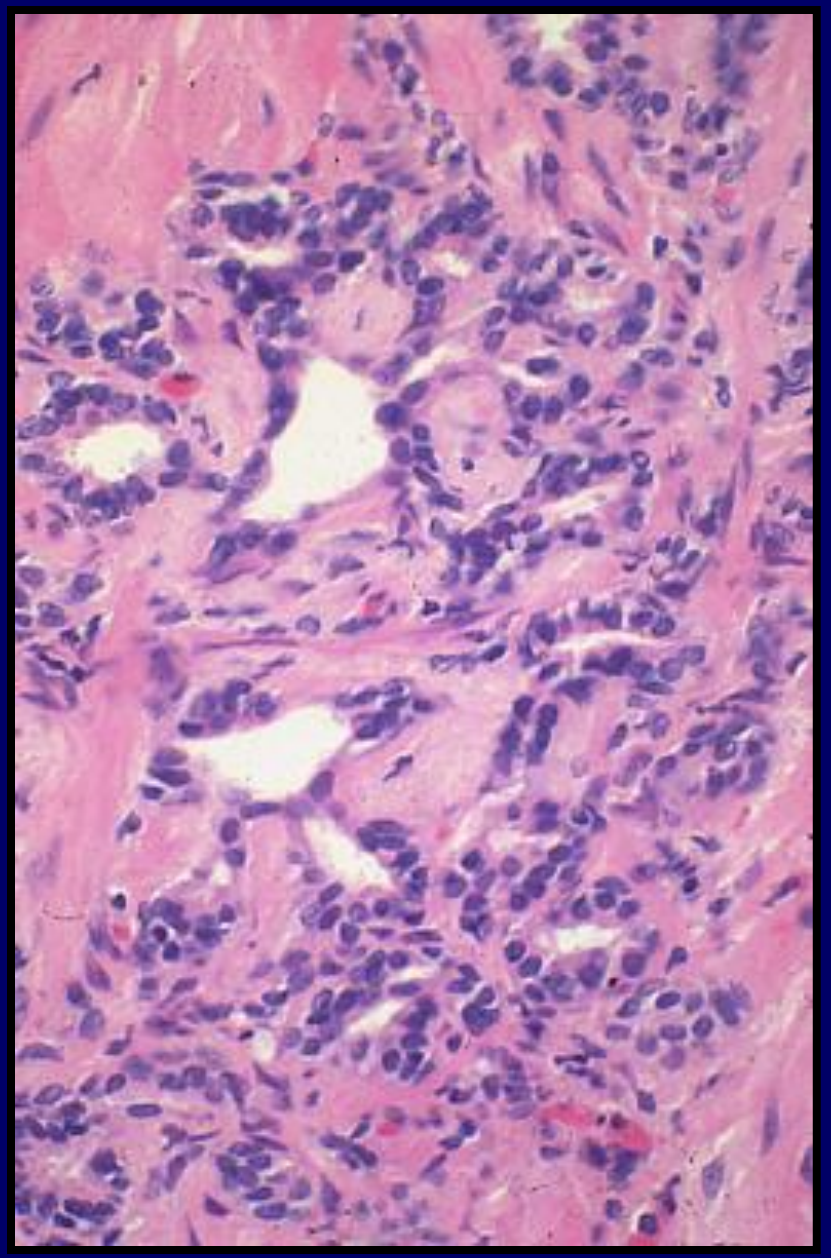
POSTATROPHIC HYPERPLASIA

- Lobular architecture
- Increased numbers of acini over usual
- Epithelium has moderate cytoplasm (not as much as usual for CA)
- Small nuclei; nucleoli generally inconspicuous
- +/- Inflammation

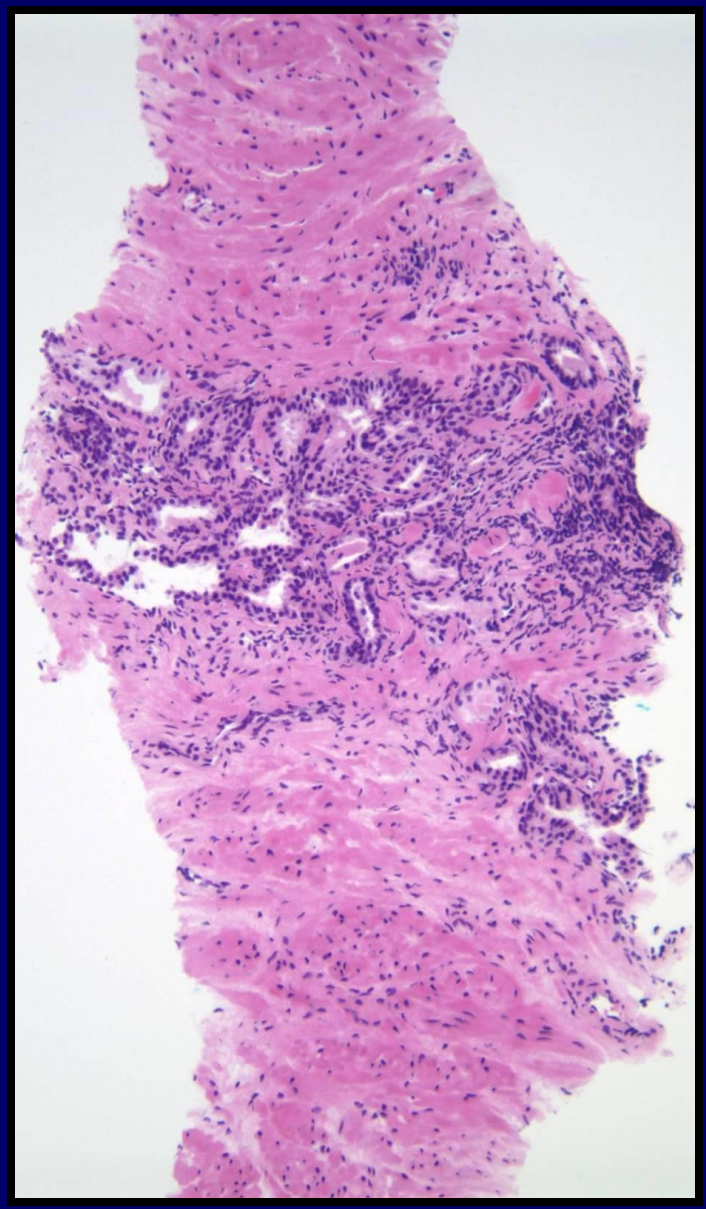


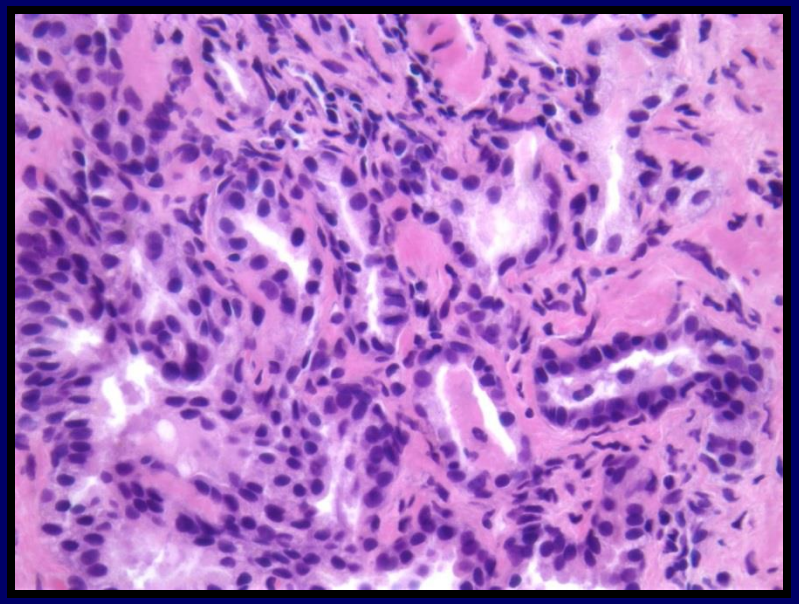
POSTATROPHIC HYPERPLASIA

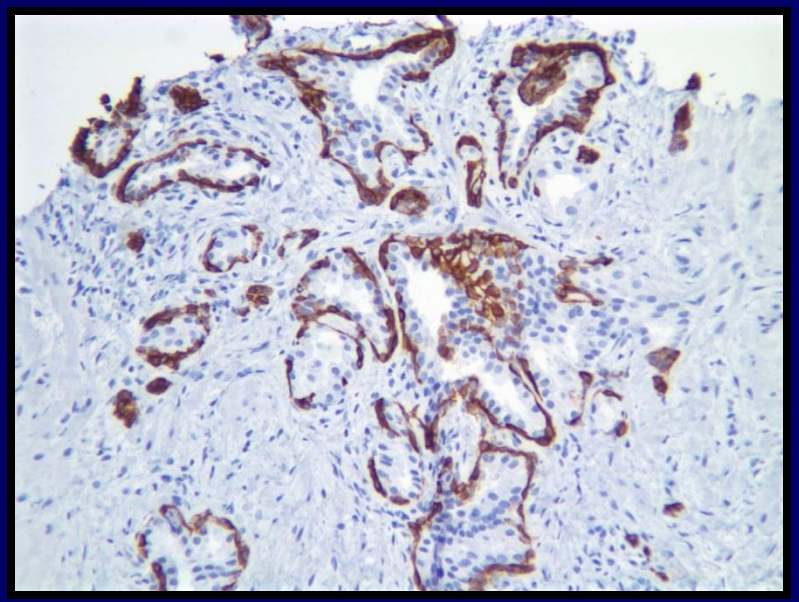


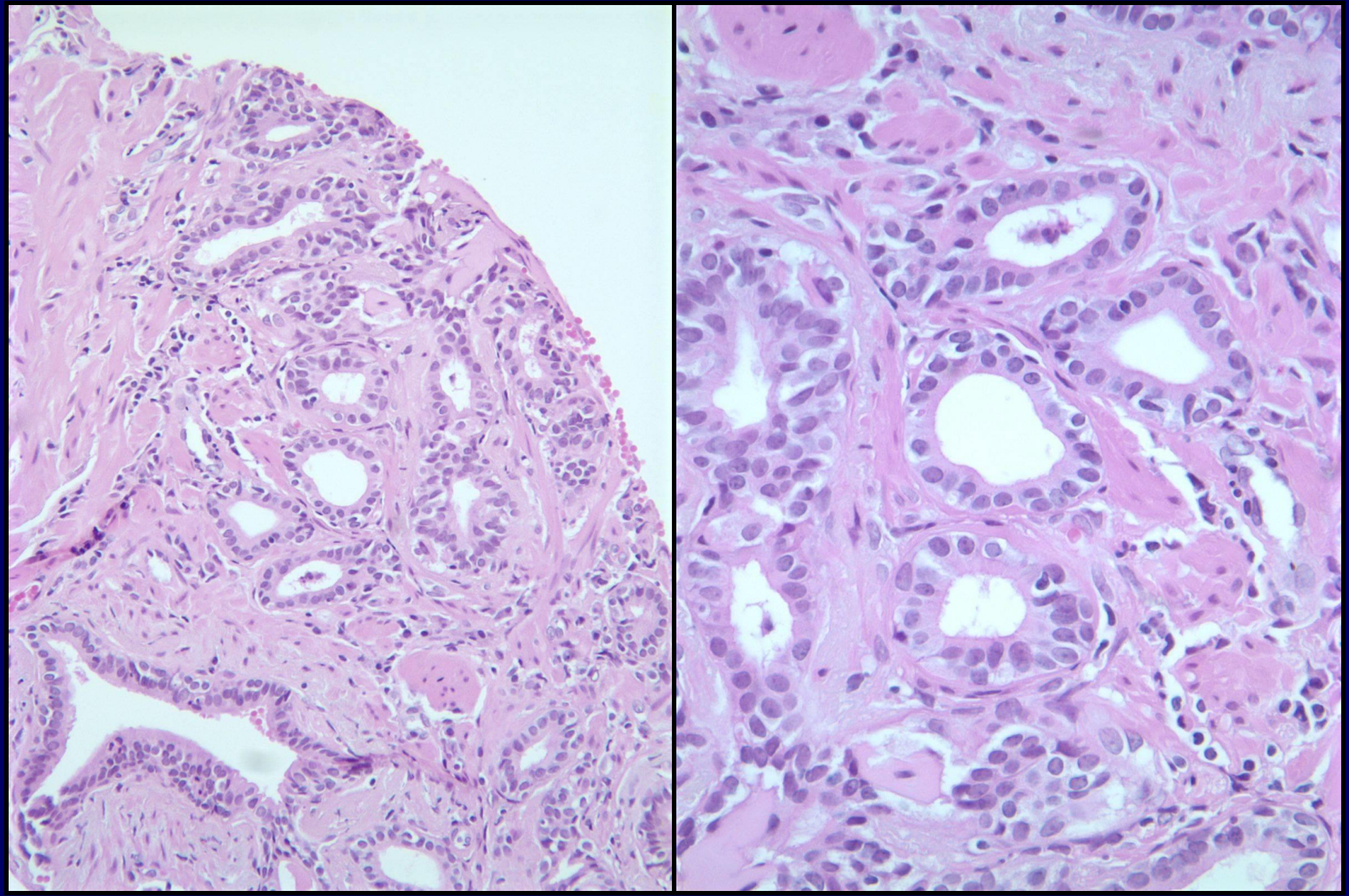


POSTATROPHIC HYPERPLASIA









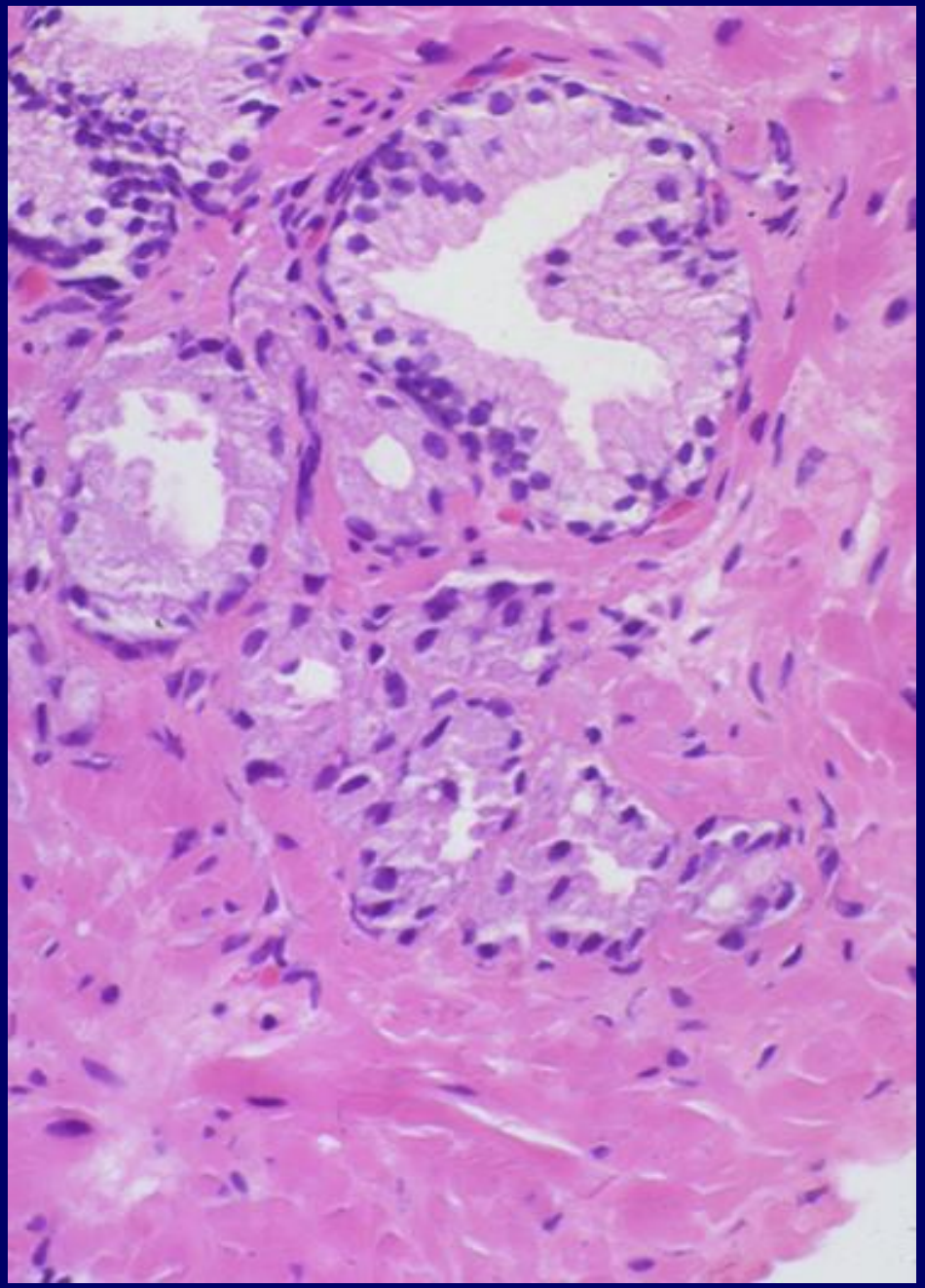
POSTATROPHIC HYPERPLASIA

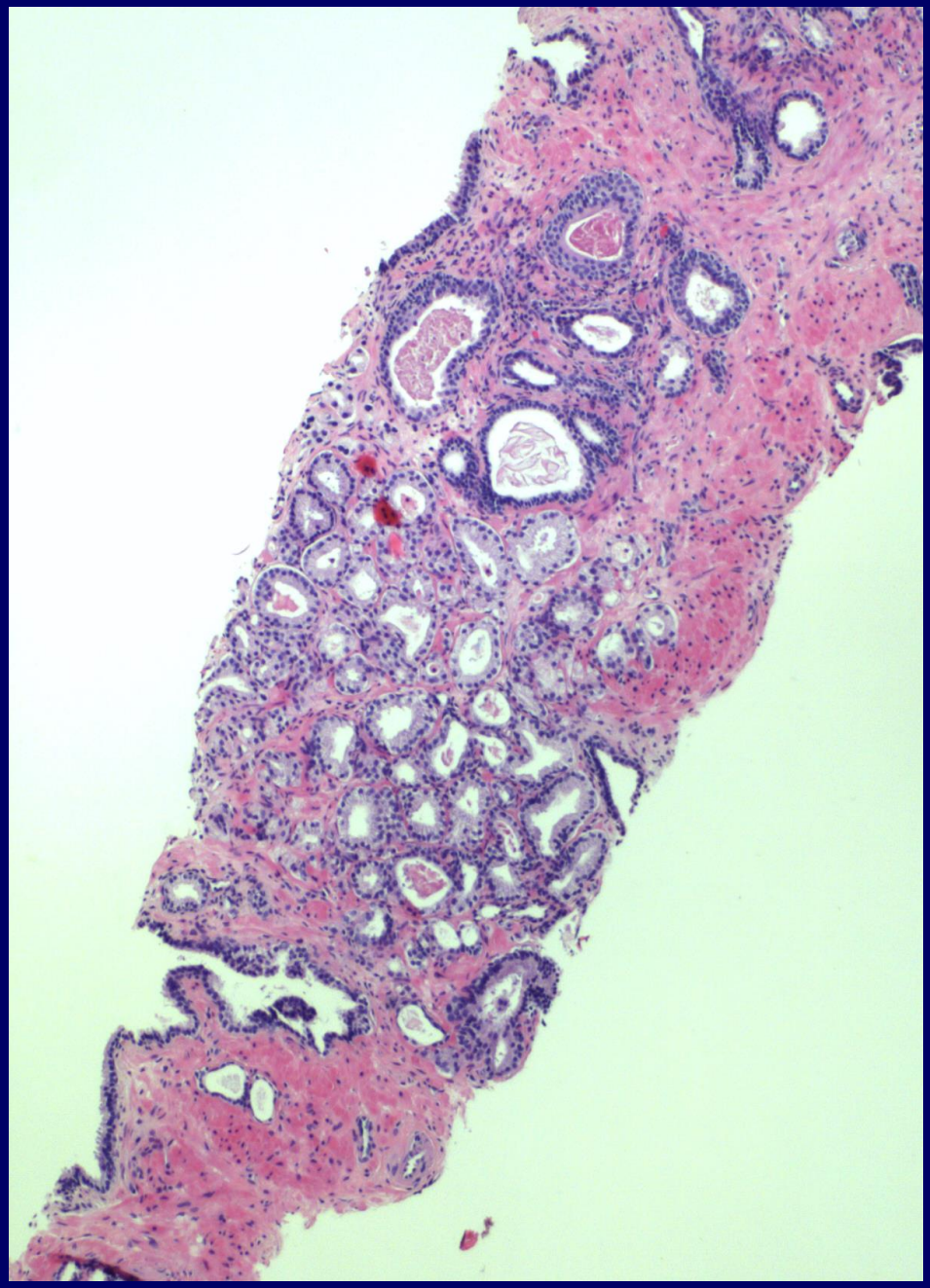
Atypical small acinar proliferation

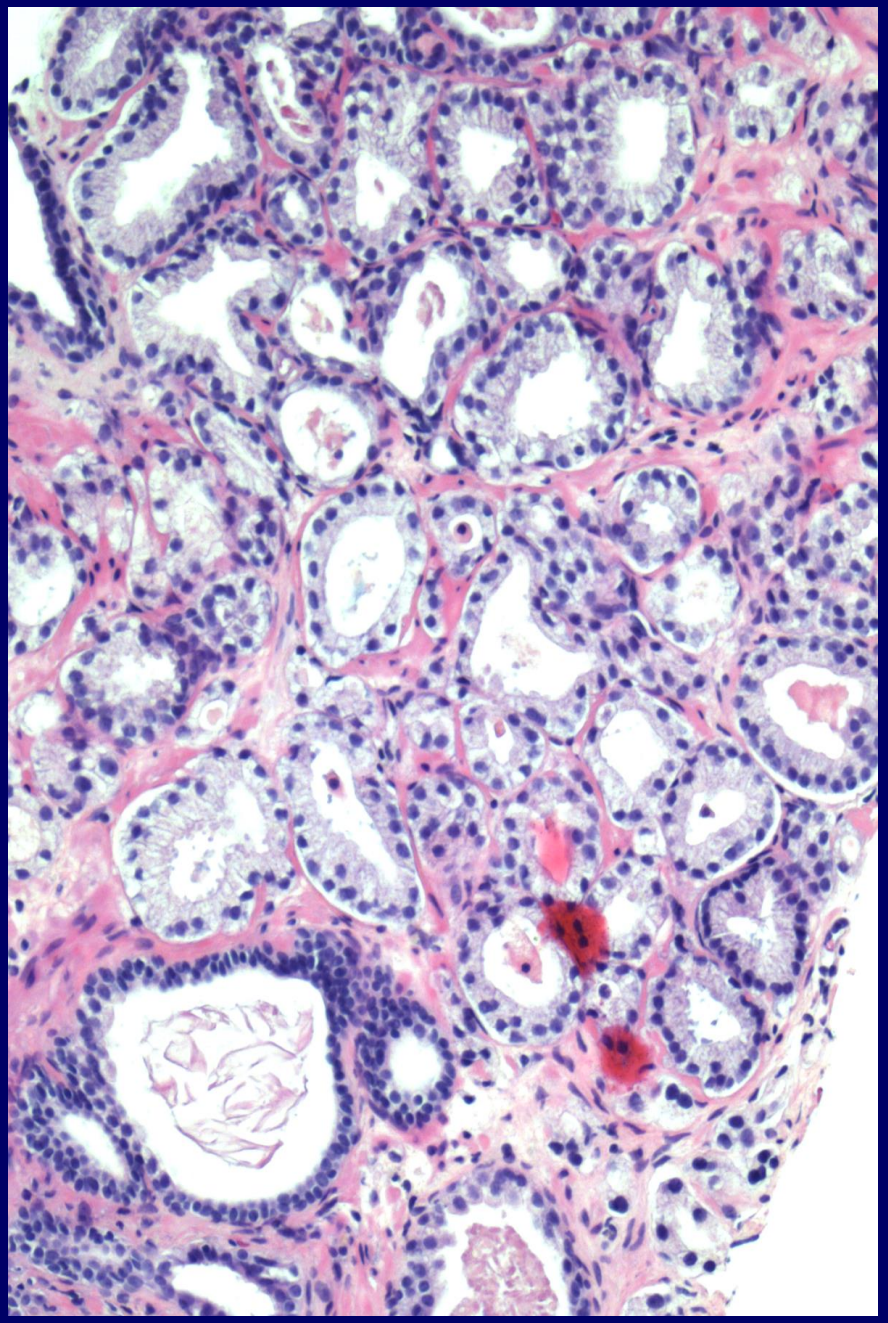
Fcous of atypical acini that does not fulfill minimal criteria for adenocarcinoma

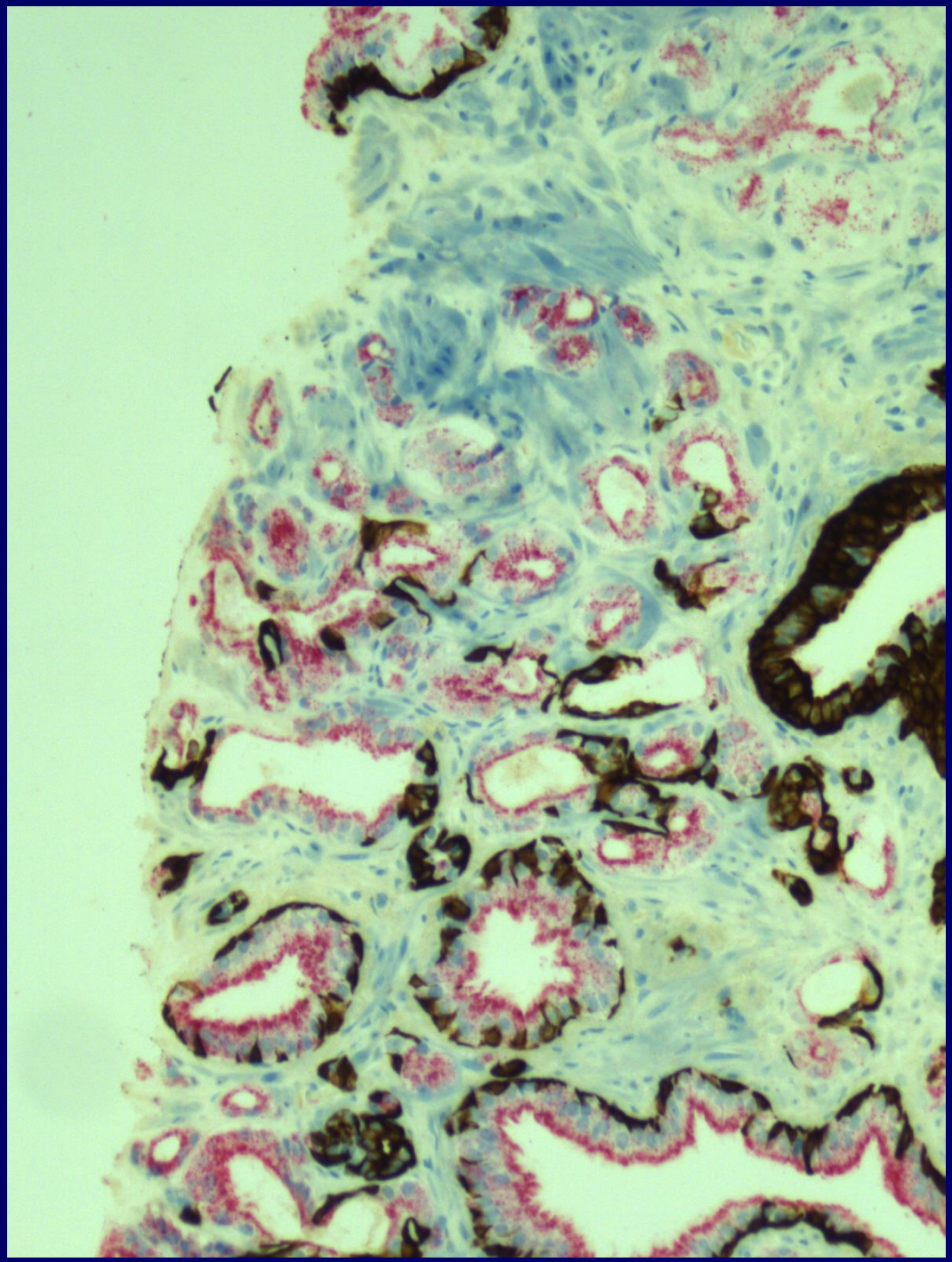
Not a diagnostic entity

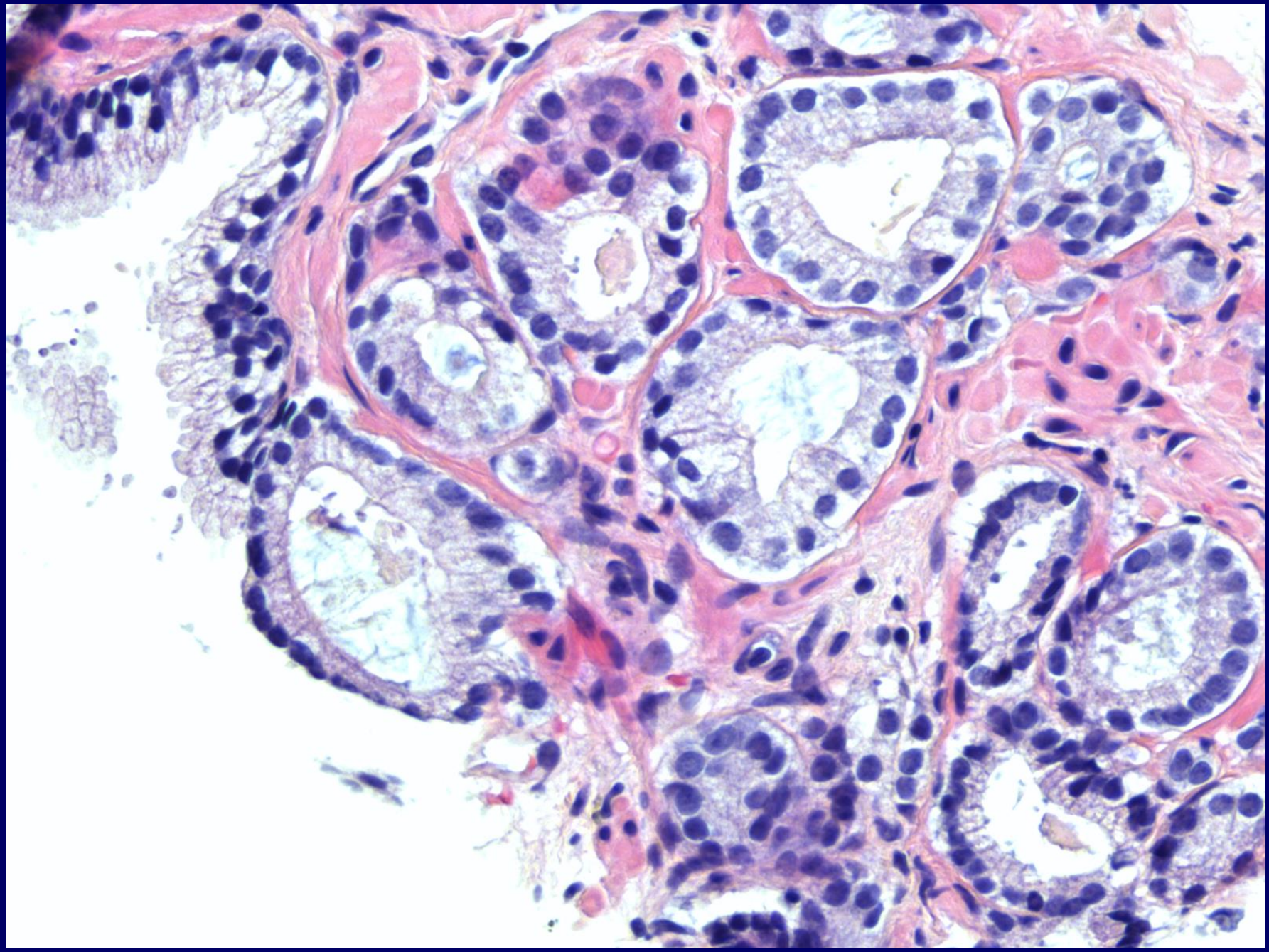
Clinically significant

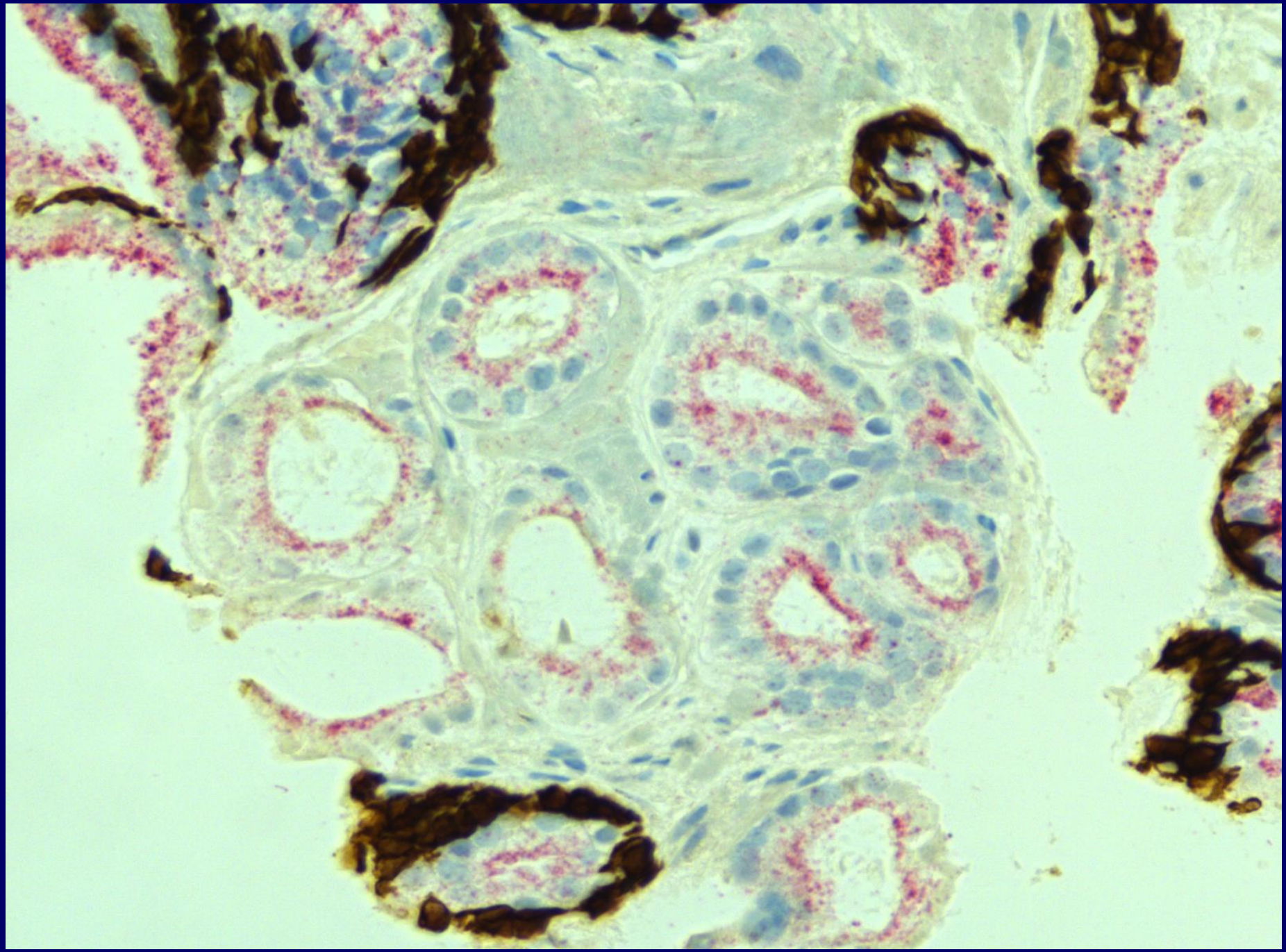


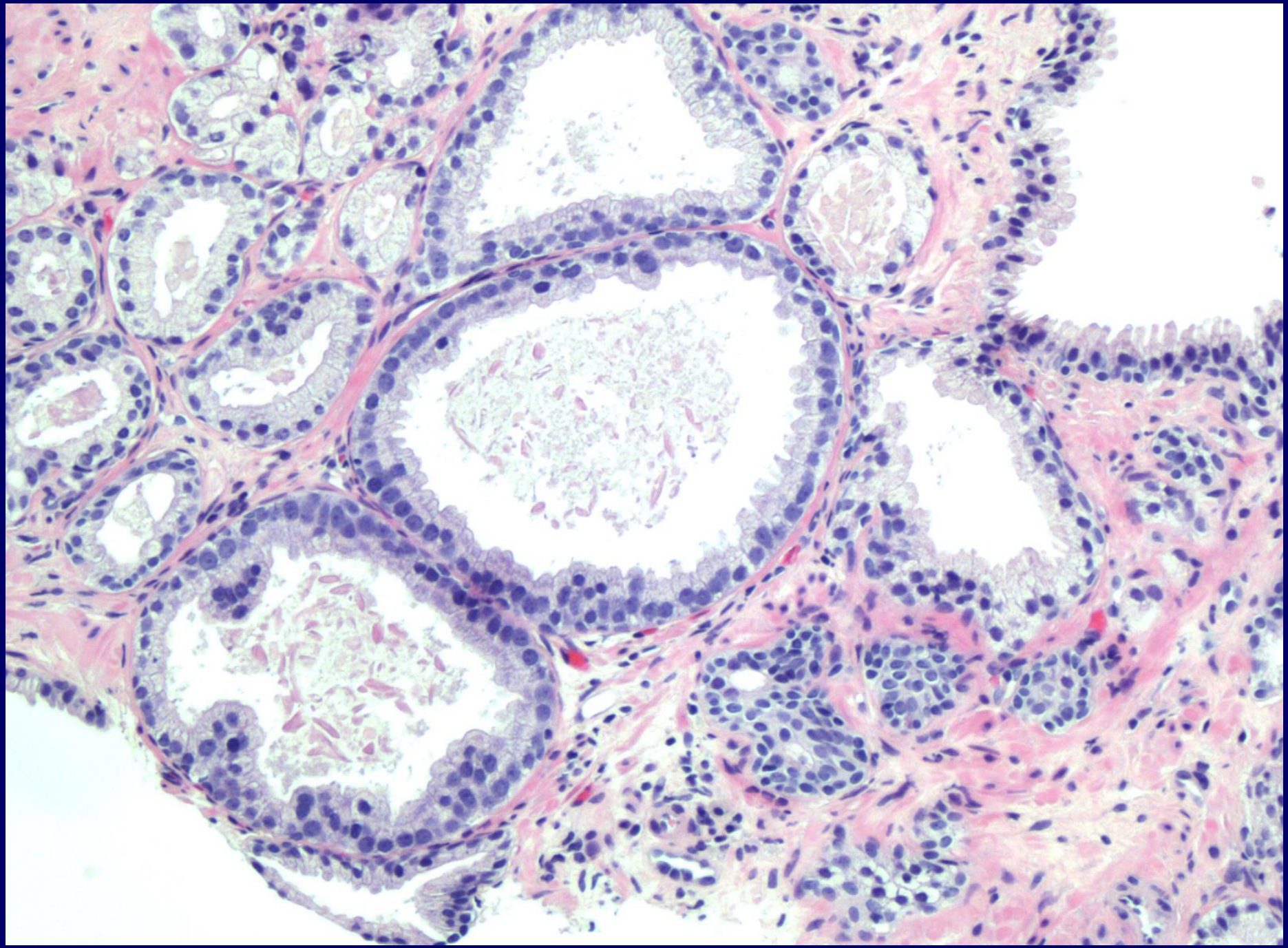


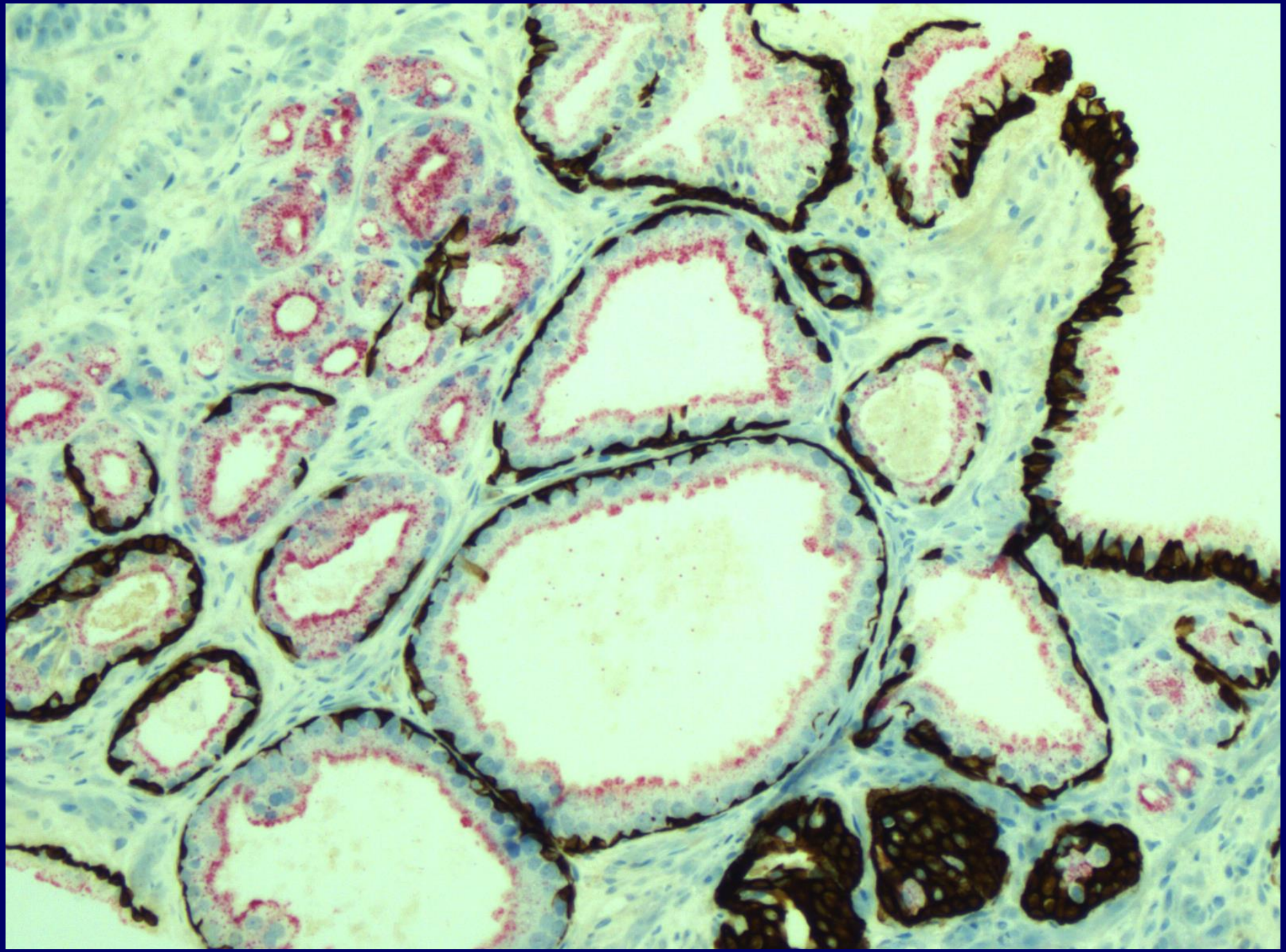


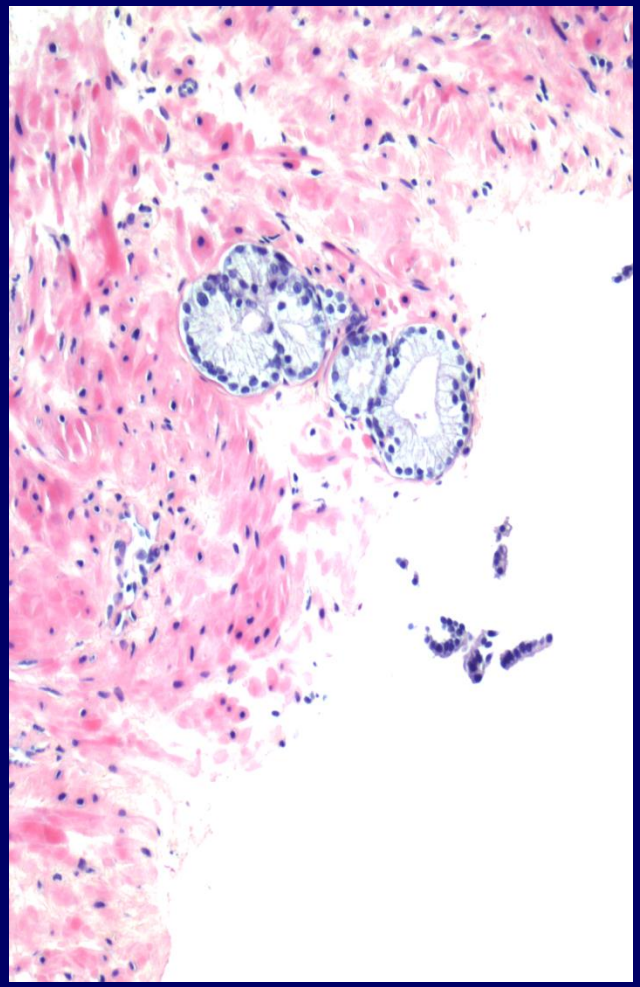
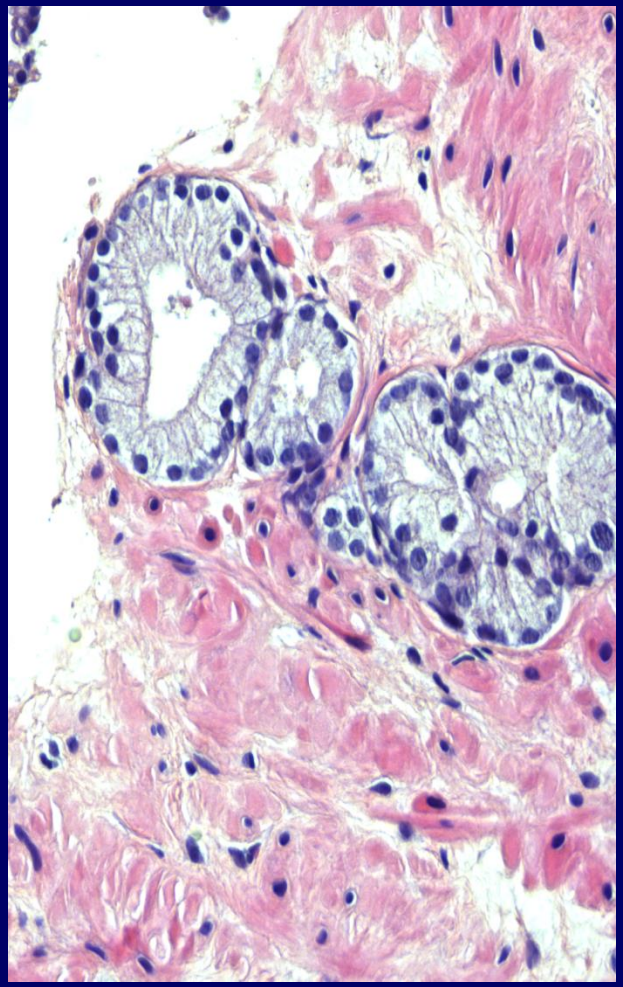
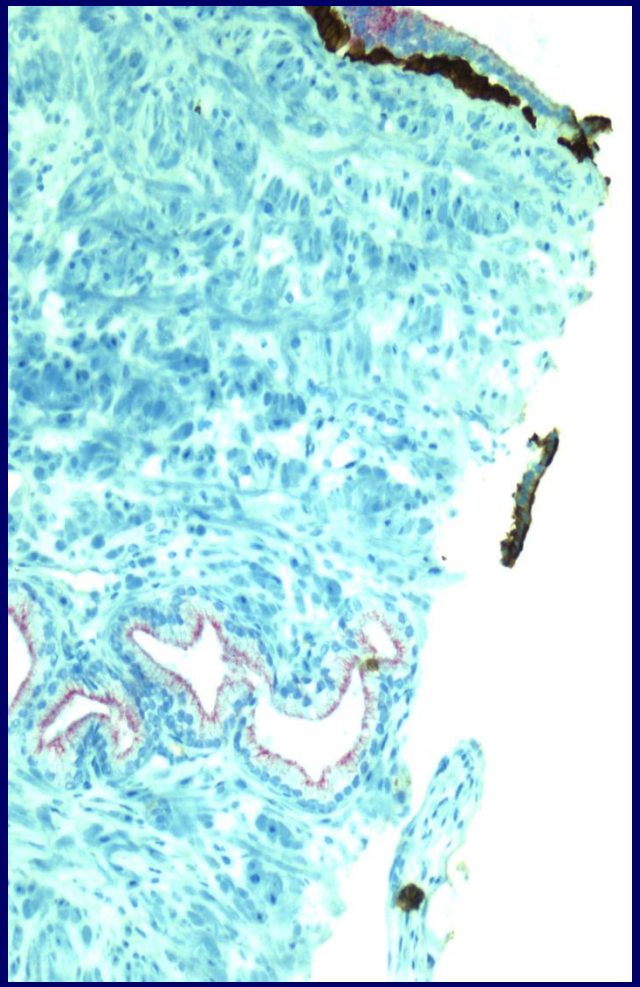




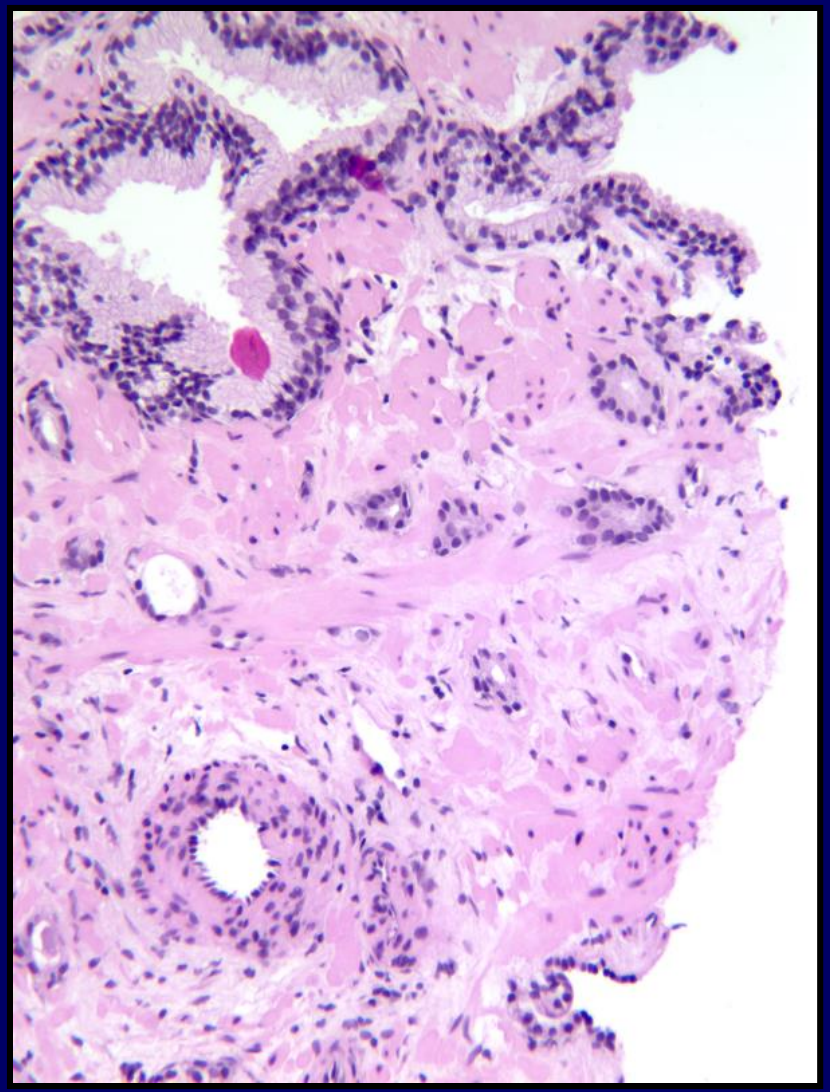


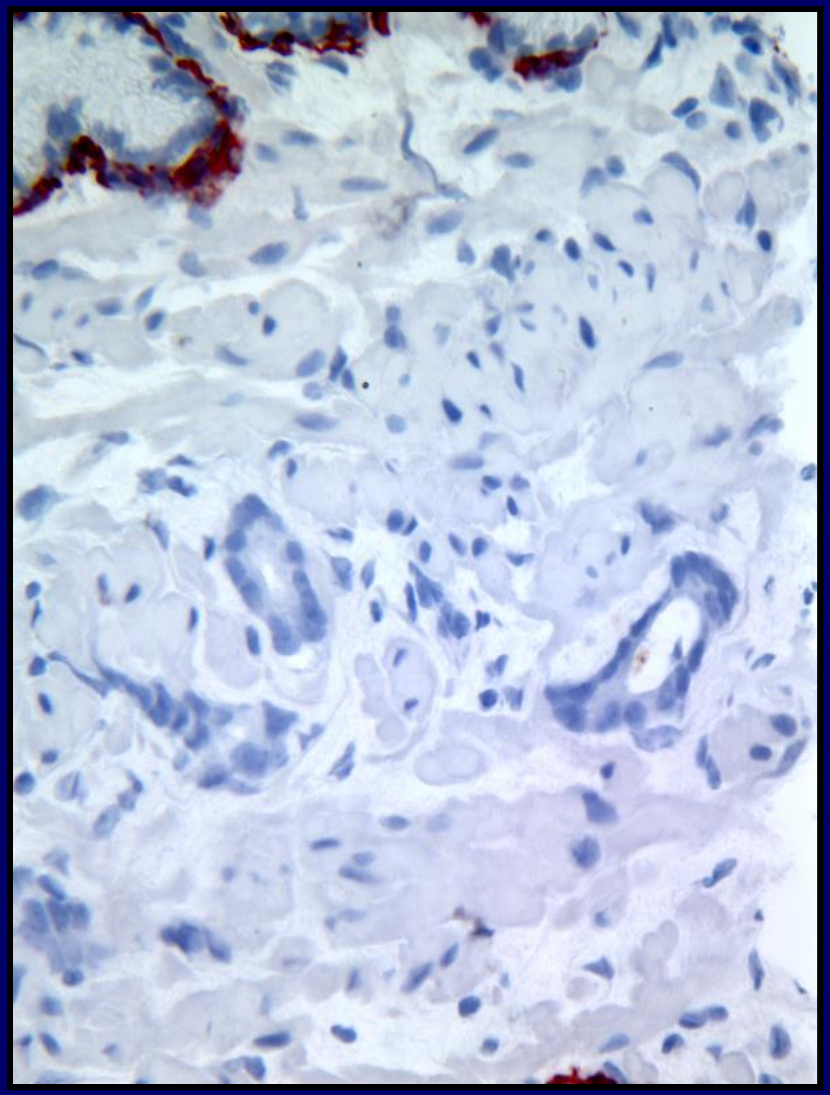


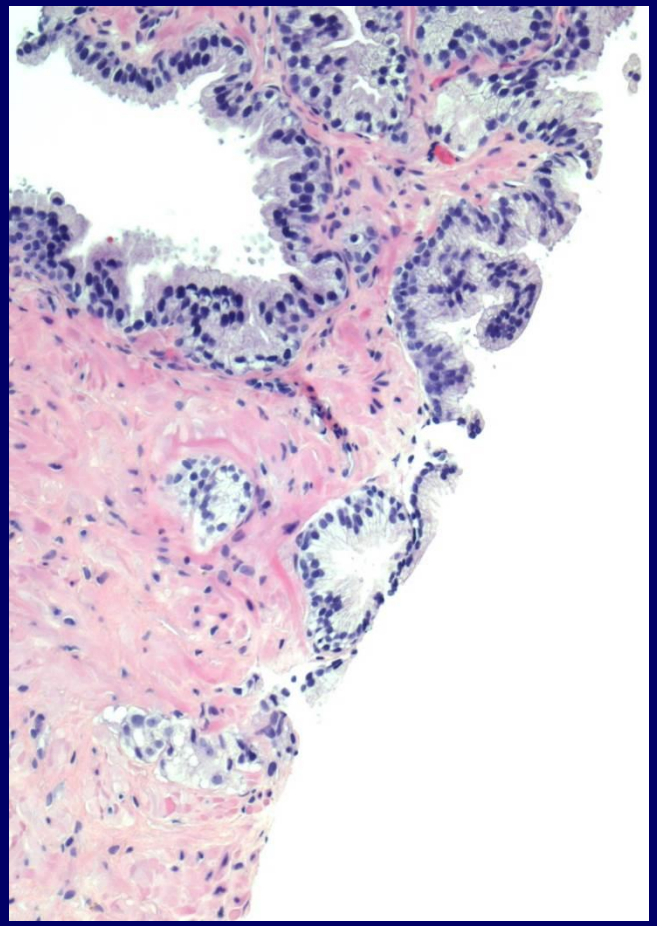
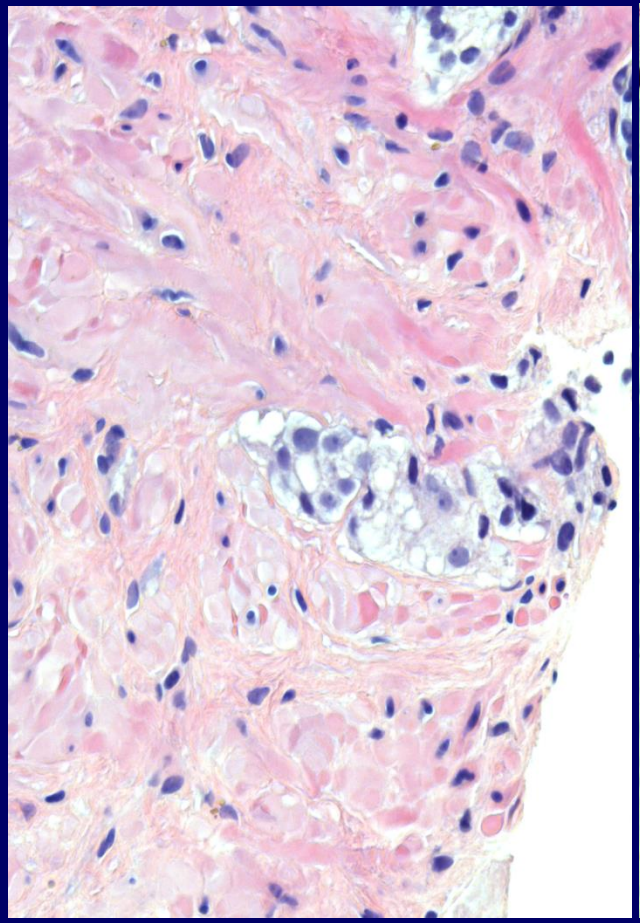
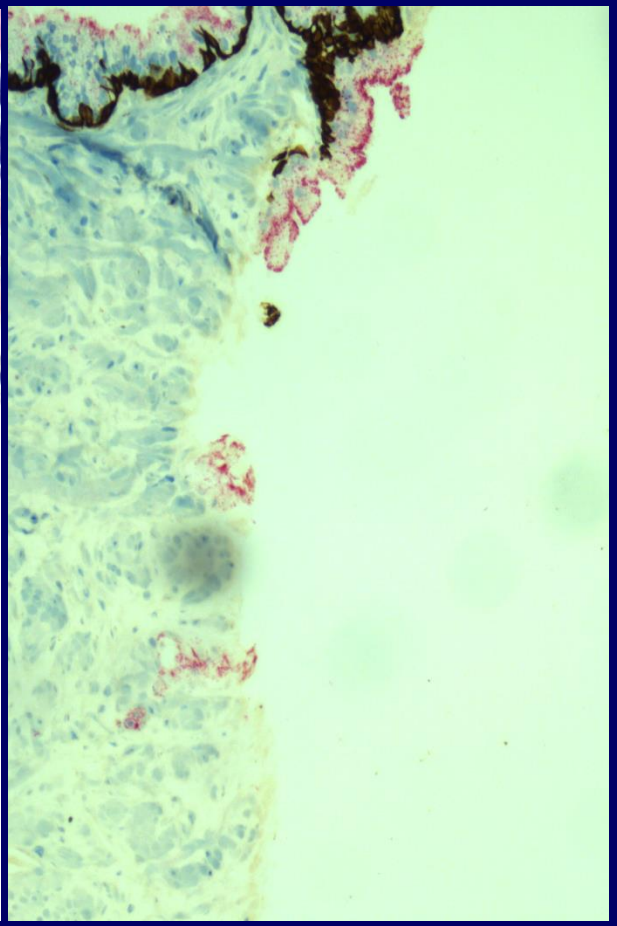


HIGH MOLECULAR WEIGHT CYTOKERATIN IHC





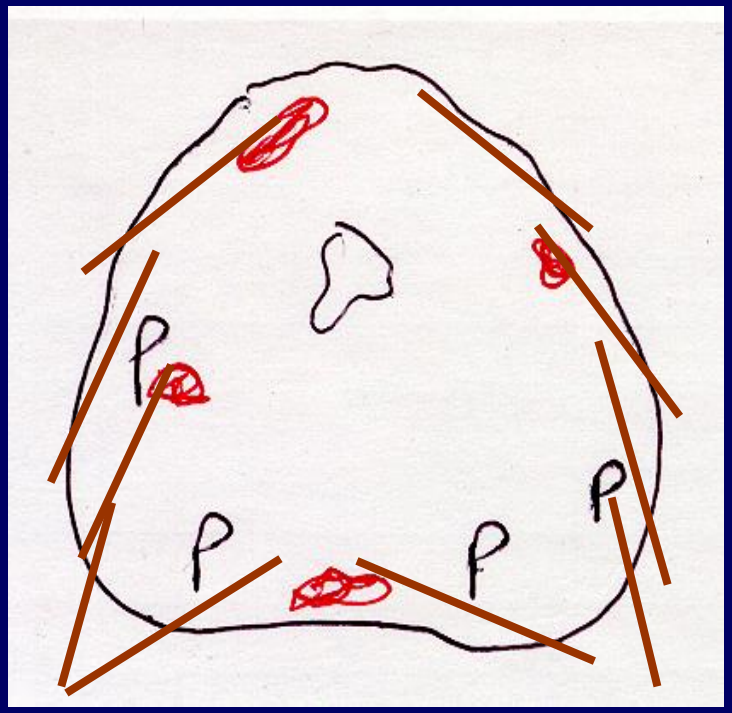
Two sequential diagnoses of atypical foci suspicious for carcinoma on prostate biopsy: a follow-up study of 179 cases

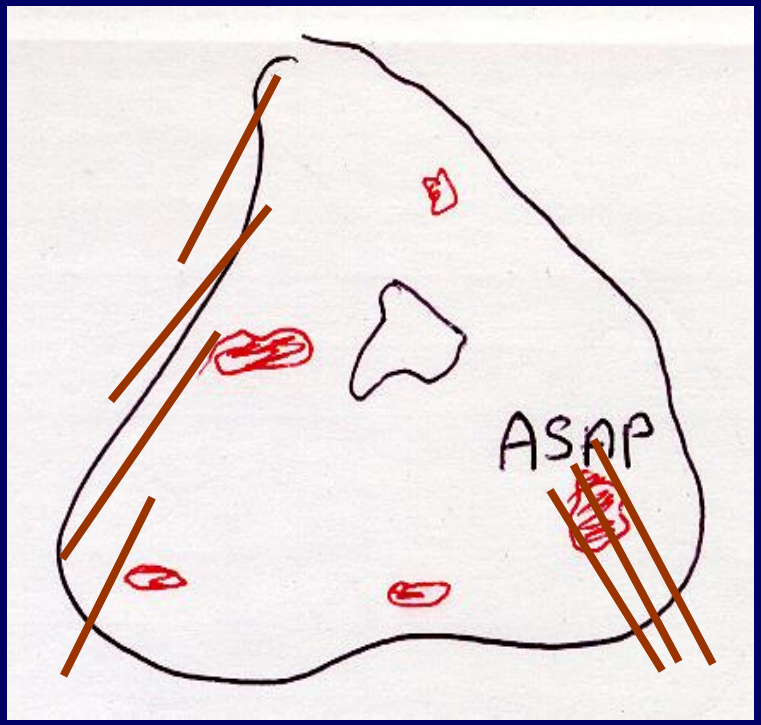
36%	**Cancer with one forth>Gleason 7**
53%	**Benign or with HGPIN**
10.6%	**Third time diagnosis of atypia**

Zhang et al

Urology. 2013;82: 861-4

Rebiopsy of HGPIN and ASAP





HGPIN: Risk of confirming CA on repeat Bx= 25-30%

Bx systemic to avoid 33% false -

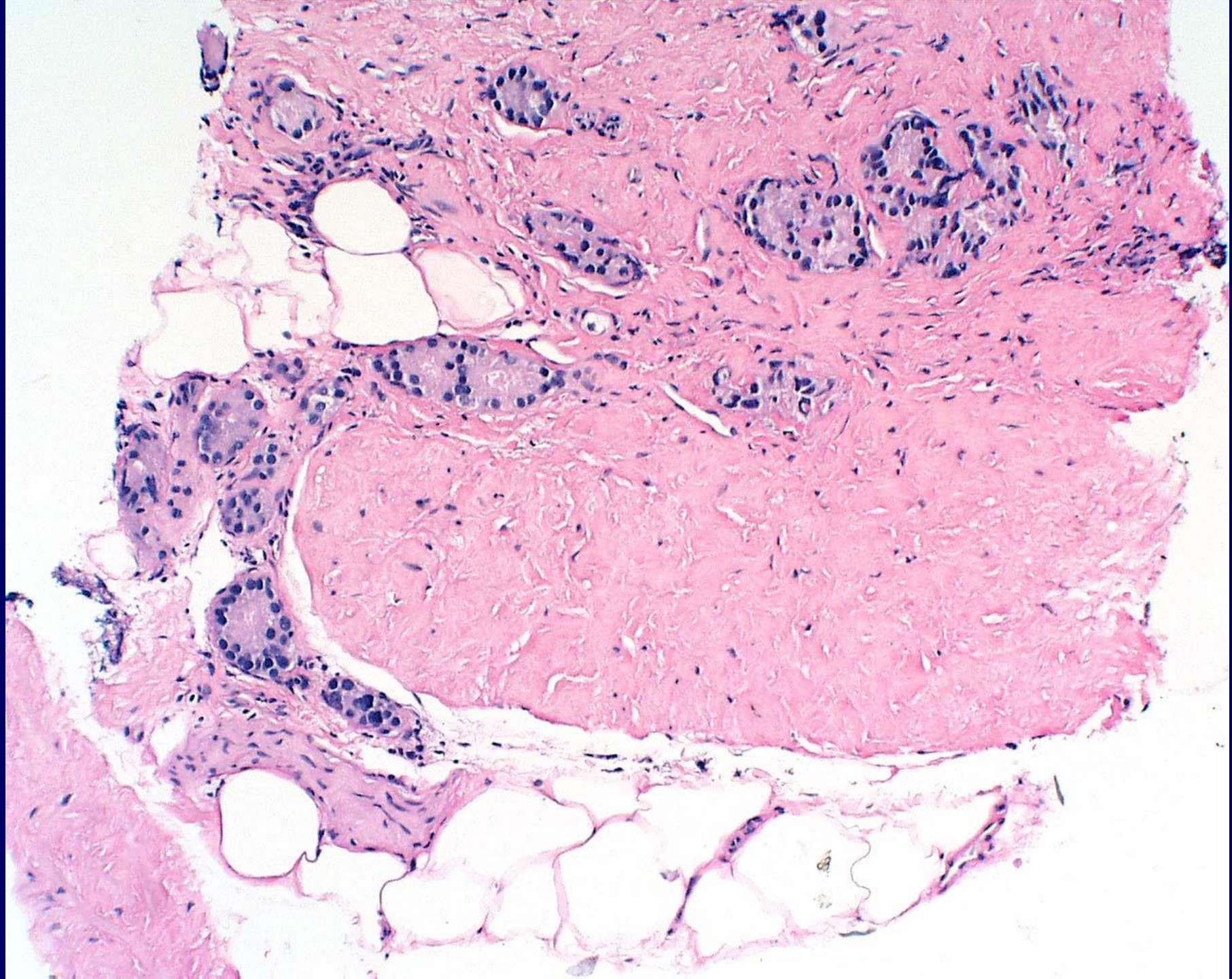
ASAP: Risk of confirming CA on repeat Bx = 45- 60%

Bx systemic but concentrate on site

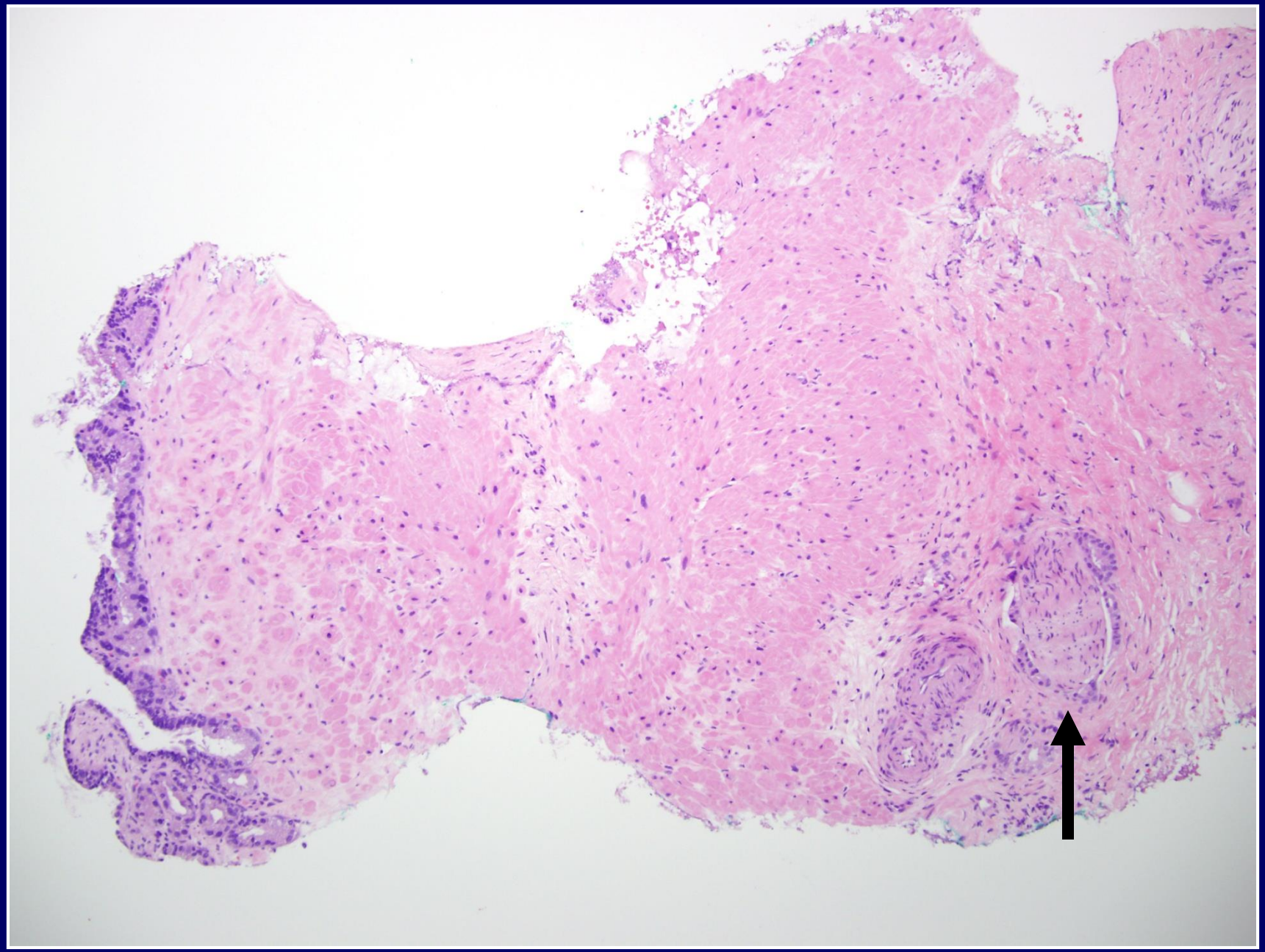
ACTIVE SURVEILLANCE IN PROSTATE CANCER: ELIGIBILITY

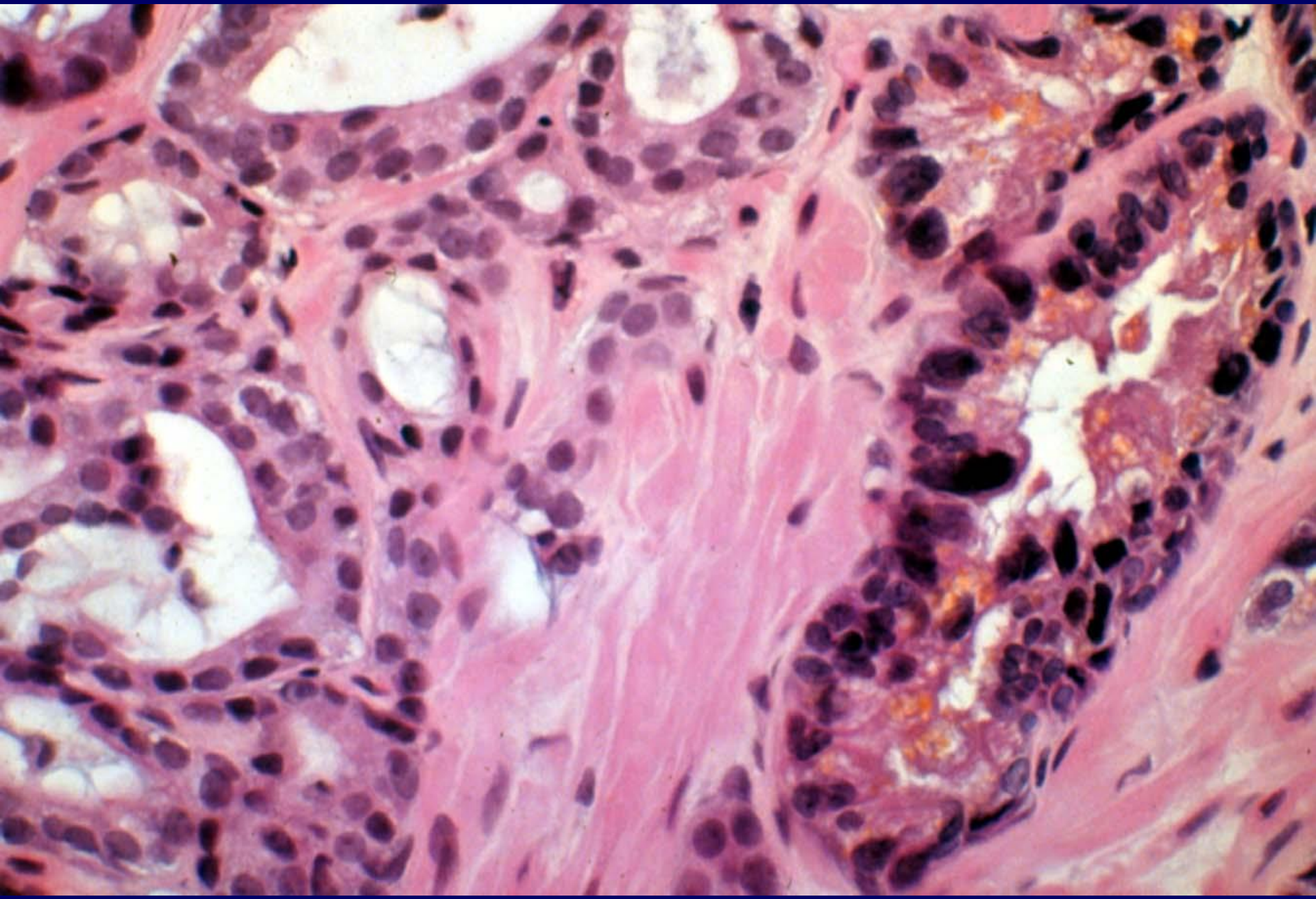
INSTITUTION	GLEASON SCORE	# POSITIVE CORES	% CORE POSITIVE
Hopkins	≤ 6	≤ 2	≤ 50%
MSKCC	≤ 6	≤ 3	≤ 50%
Schroder	≤ 6	≤ 2	--
UCSF	≤ 6	≤ 33% (min 6)	≤ 50%
U Miami	≤ 6	≤ 2	≤ 20%

Iremashvili et al Eur Urol 62:462, 2012



SEMINAL VESICLE BIOPSY



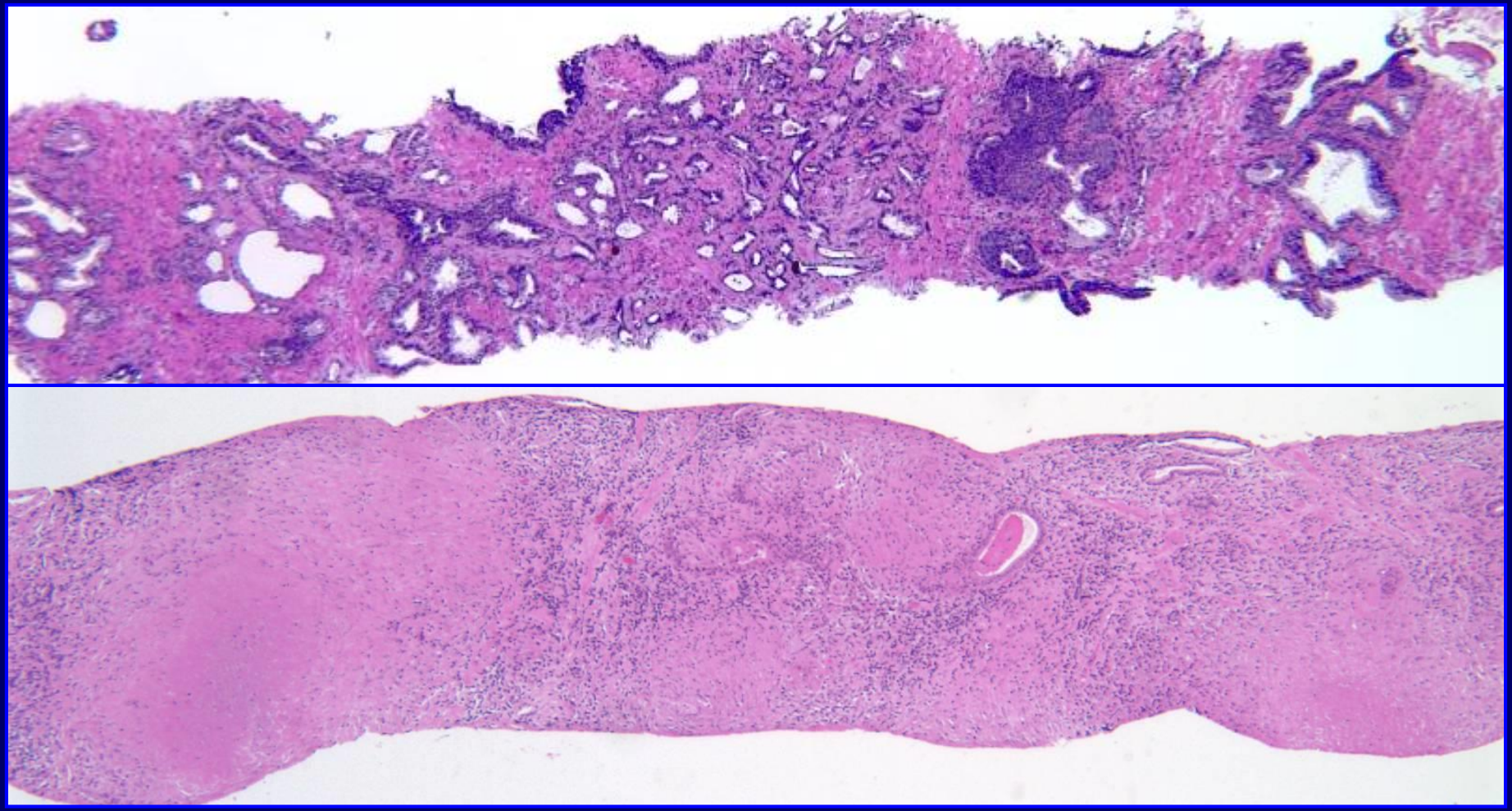


REPORTING OF "NEGATIVE" NEEDLE BIOPSIES

#4 - Left base prostate needle biopsy: Prostatic tissue, no evidence of malignancy.

#5 - Left mid prostate needle biopsy: Prostatic tissue, no evidence of malignancy.

#6 - Left apex prostate needle biopsy: Prostatic tissue, no evidence of malignancy.



REPORTING OF "NEGATIVE" NEEDLE BIOPSIES

Extent of Prostatic Atrophy in Needle Biopsies and Serum PSA Levels: Is There an Association?

Athanase Billis, Luciana R. Meirelles, Luis A. Magna, Jamal Baracat, Adilson Prando, and Ubirajara Ferreira

Association of Extent and Aggressiveness of Inflammation with Serum PSA Levels and PSA Density in Asymptomatic Patients

Engin Kandirali, Cetin Boran, Erdinc Serin, Atilla Semercioz, and Ahmet Metin

Inverse Association Between Histologic Inflammation in Needle Biopsy Specimens and Prostate Cancer in Men With Serum PSA of 10-50 ng/mL

Tomoaki Terakawa, Hideaki Miyake, Naoki Kanomata, Masafumi Kumano, Atsushi Takenaka, and Masato Fujisawao

Prostate Needle Biopsy

Cancer ?

Yes

Classify

Grade
Gleason Score of every + Core
Include
Primary
Secondary
Tertiary

Extent of involvement

- # of cores involved
- % of core involvement and/or mm measurement of Ca segment
- Perineural invasion

Staging Parameters

- Extraprostatic extension (EPE)
- Seminal vesicle invasion

No

Atypical
Suspicious

HGPIN

optional
# of cores
Extent

Benign Conds

Inflammation

Atrophy

Histopathology reports of prostate needle biopsies: Individual treatment

95 of 282 (33 %)

58 % took 10-12 cores, 34 % 6-8 cores and 6 % > 6 cores

Urologists expected:

- Primary and secondary Gleason patterns 38%
- Gleason score on single cancer focus, if not given, 48%
- Used worst Gleason score for multiple positive cores 88%
-They repeated bx in 44% after ASAP & 57% after HGPIN
- Treatment influenced by

    perineural invasion 60%, extraprostatic spread 57%

    percentage of core involvement 13%

Urologe A. 2013 Feb;52(2):226-39

Web-based survey of 266 pathologists in 22 countries
98% claimed to follow ISUP recommendations

Smoothly rounded cribriform glands were assigned Gleason pattern (GP) 3 by 51% and GP 4 by 49%

Necrosis was diagnosed as Gleason grade 5 by 62%

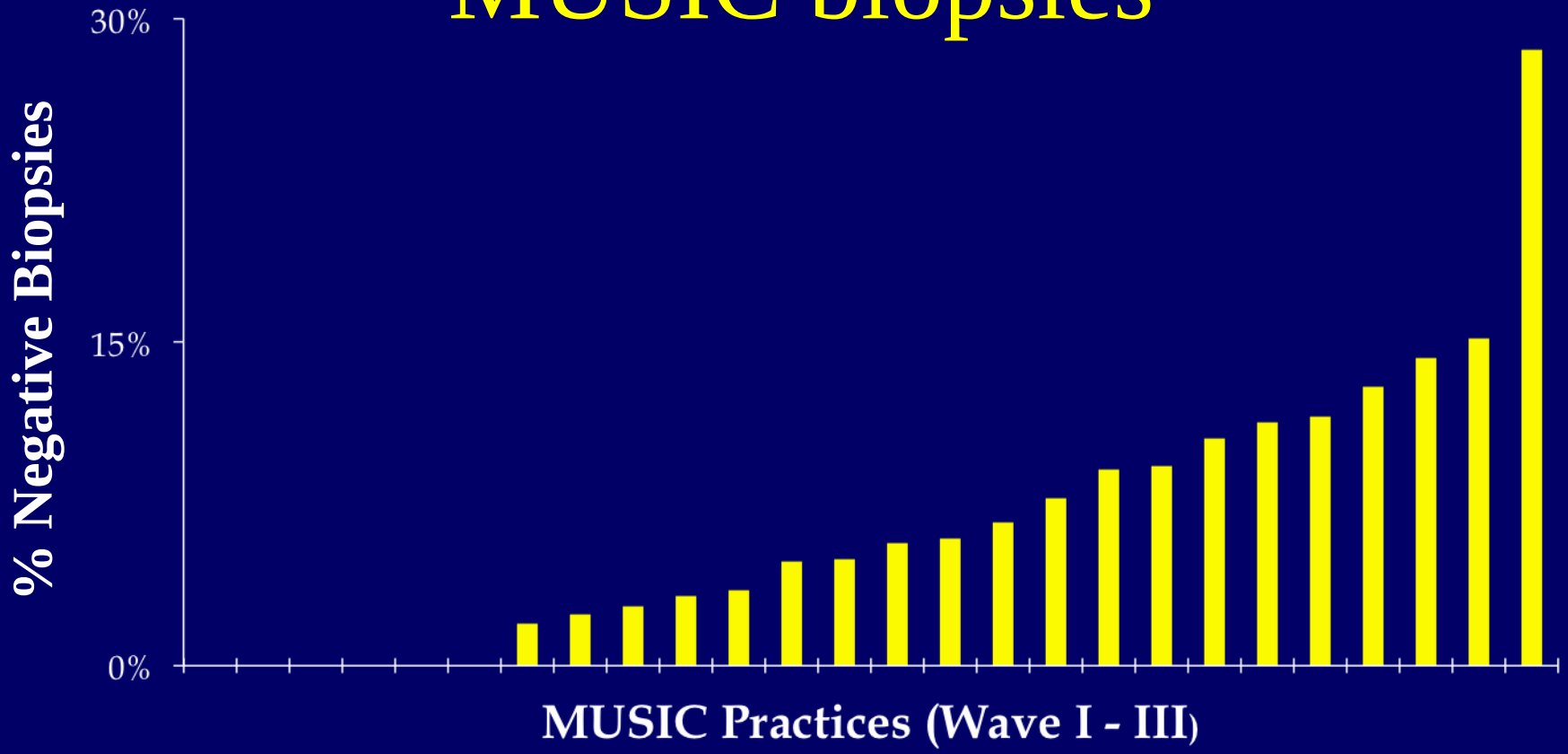
Any amount of secondary pattern of higher grade in needle biopsies was included in the Gleason score by 58%

Tertiary GP of higher grade included in score by only 58%

Gleason score assigned for each core if submitted separately 56%

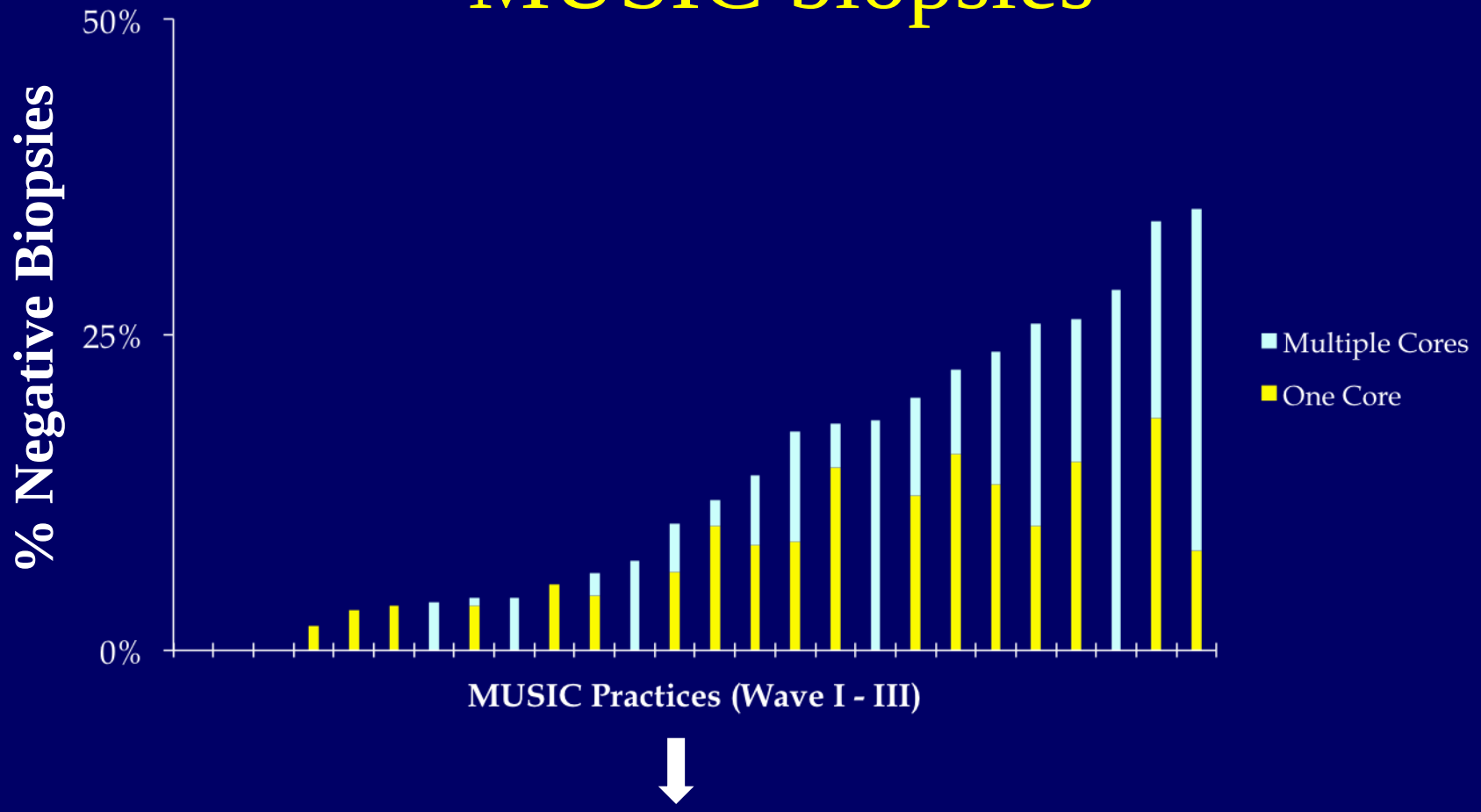
Histopathology 2013

Frequency of ASAP in 6,000 MUSIC biopsies



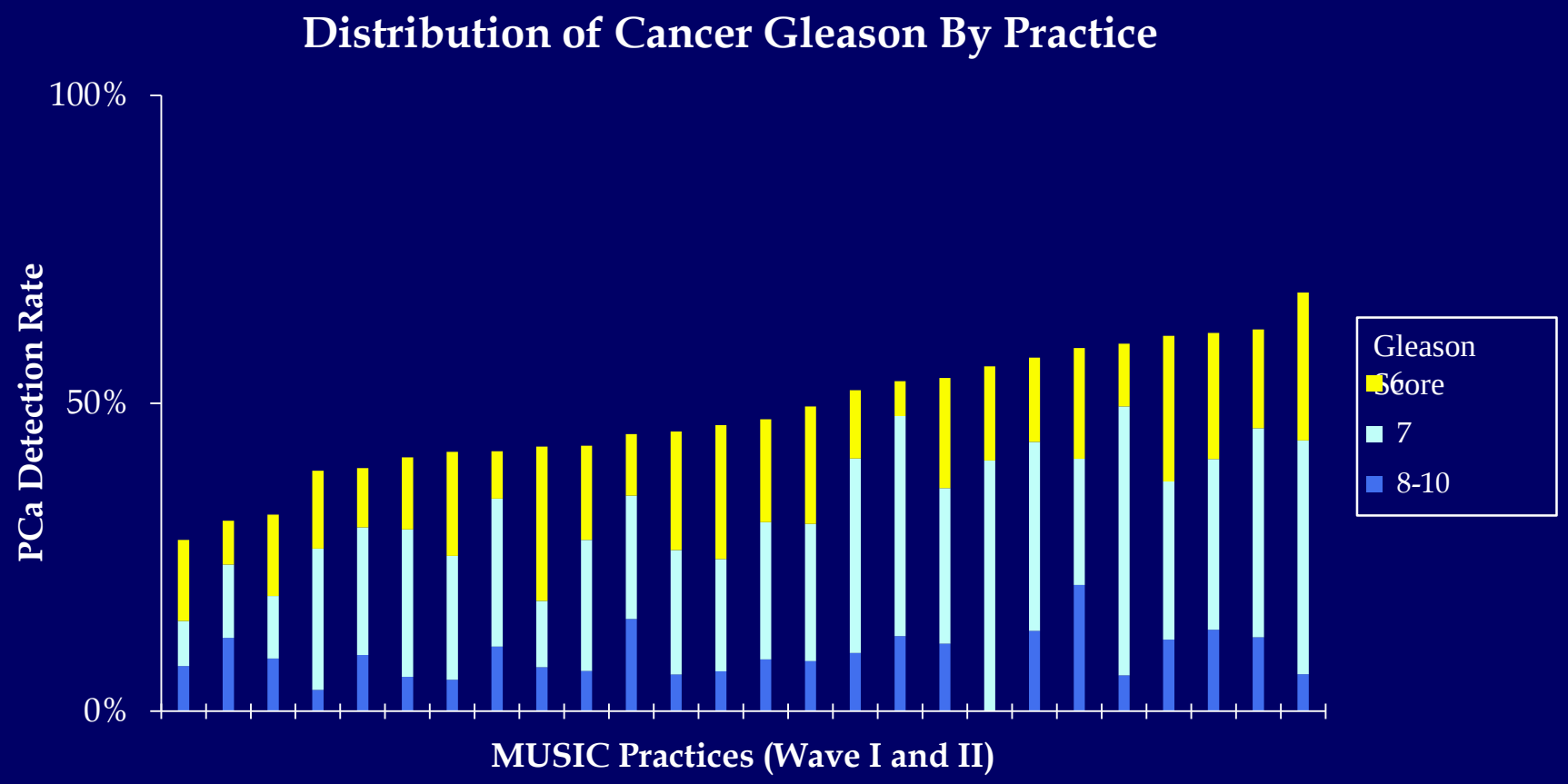
Range 0-30%

Frequency of HGPIN in 6,000 MUSIC biopsies



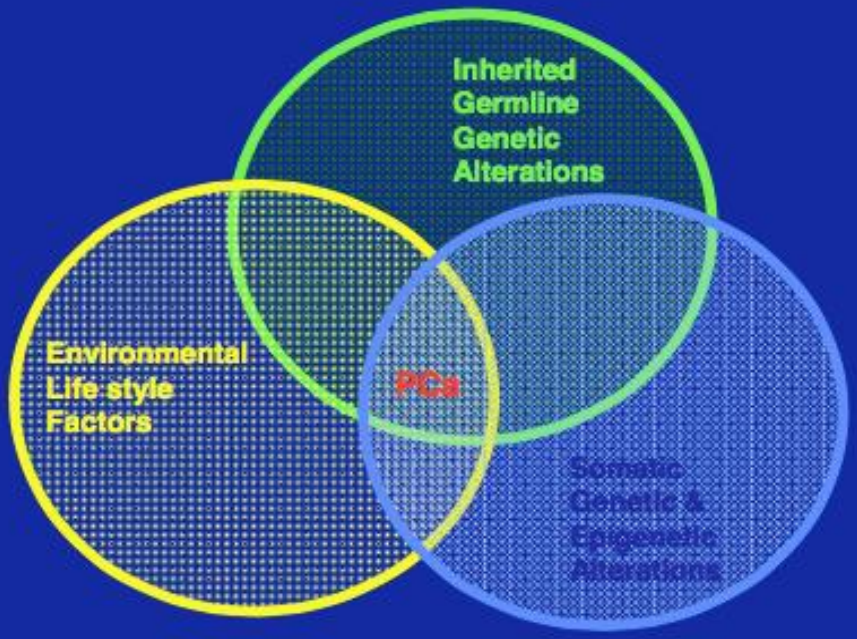
Multiple cores range 0-20%

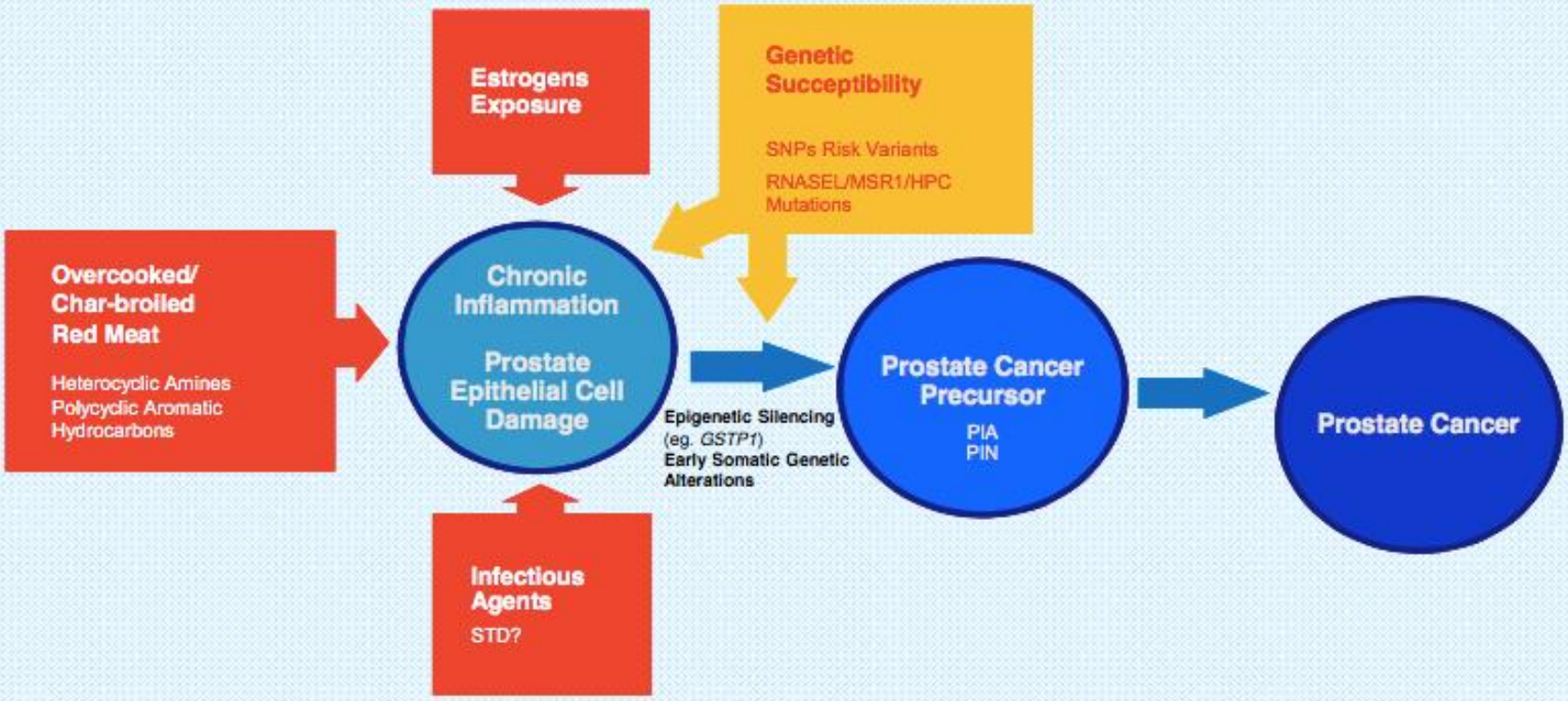
Cancer Detection Rate



Prostate Carcinoma

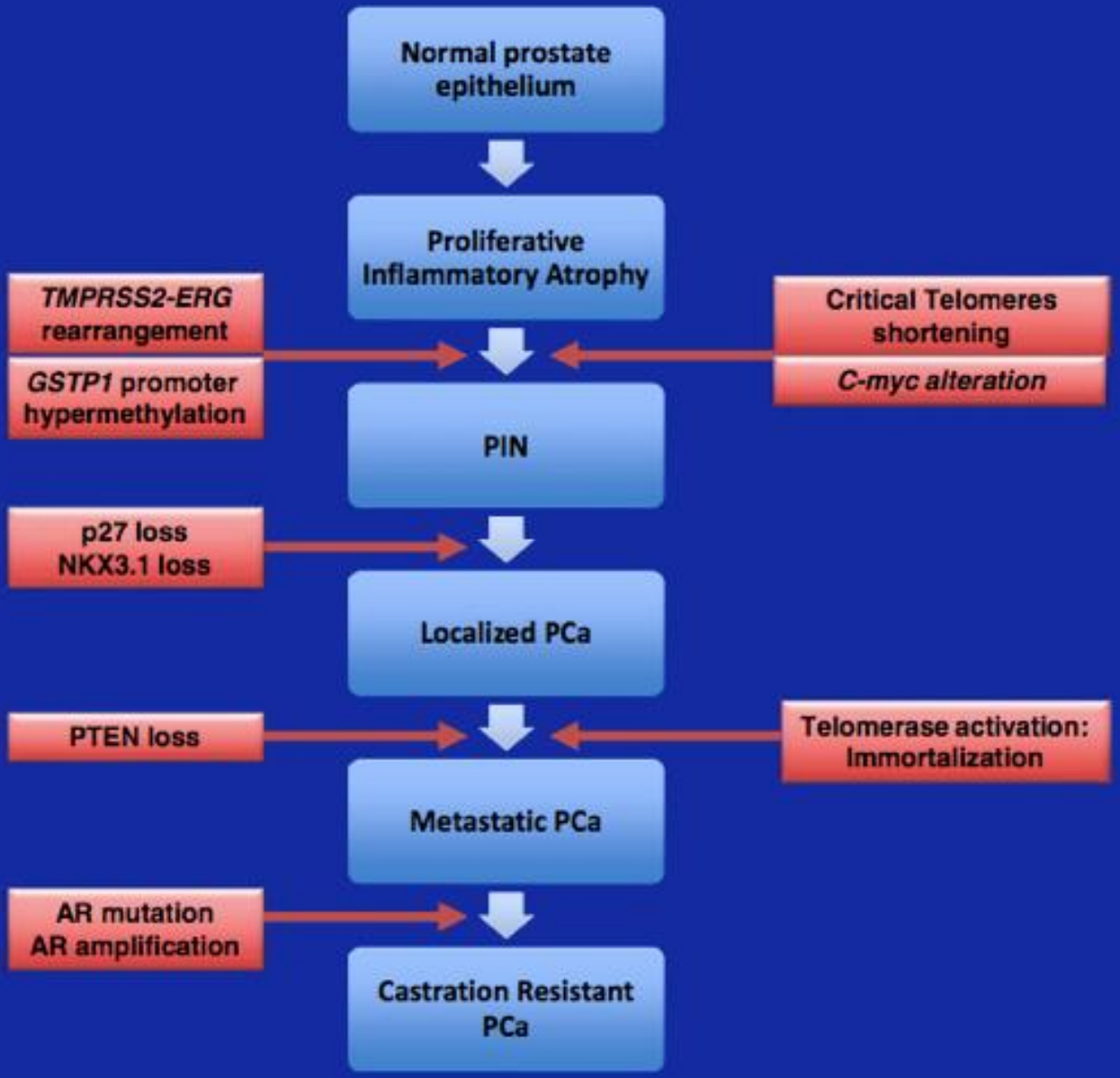
Risk Factors/Etiology





Etiological factors implicated in prostate cancer development

Somatic Alterations
Genetic and Epigenetic



Netto, G. Adv Anat Pathol 2013

ETS FUSIONS

RESEARCH ARTICLE

Recurrent Fusion of *TMPRSS2* and ETS Transcription Factor Genes in Prostate Cancer

Scott A. Tomlins,[1] Daniel R. Rhodes,[1,2] Sven Perner,[7,9] Saravana M. Dhanasekaran,[1] Rohit Mehra,[1] Xiao-Wei Sun,[7] Sooryanarayana Varambally,[1,6] Xuhong Cao,[1] Joelle Tchinda,[7] Rainer Kuefer,[10] Charles Lee,[7] James E. Montie,[3,5,6] Rajal B. Shah,[1,3,5,6] Kenneth J. Pienta,[3,4,5,6] Mark A. Rubin,[7,8] Arul M. Chinnaiyan[1,2,3,5,6*]

TMPRSS2 (Transmembrane Protease, Serine 2)

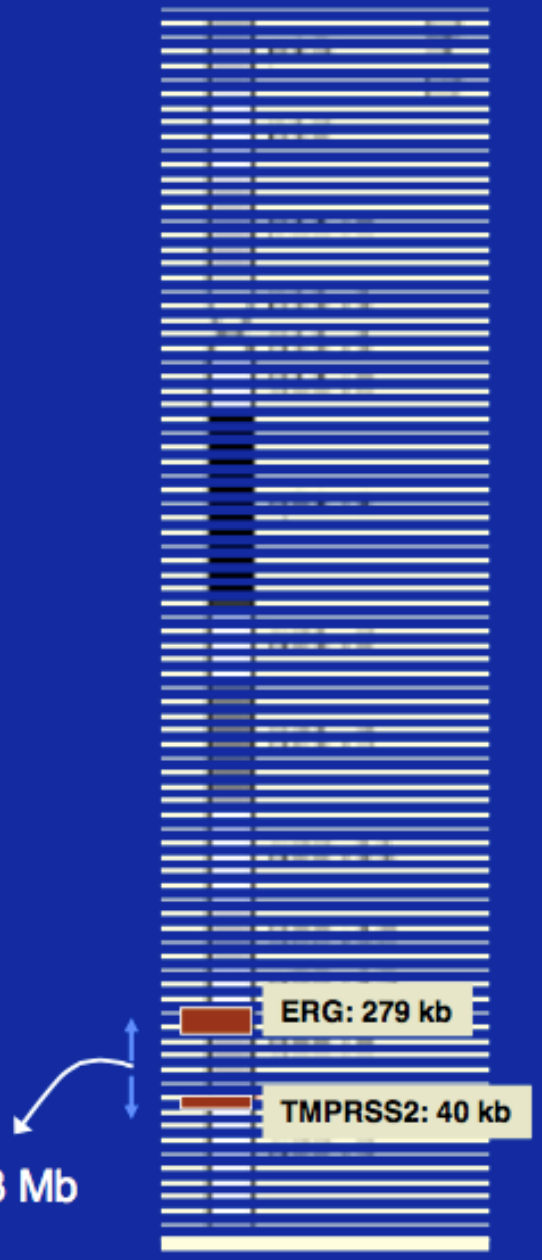
- Prostate specific serine protease family (PSA, hK2, prostate/PRSS18, TMPRSS2)
- *TMPRSS2* gene is located on chromosome 21q22.3, it encodes a protein of 492 amino acids
- *TMPRSS2* expression is regulated by androgen
- *TMPRSS2* is highly expressed in normal and neoplastic prostate epithelium.

ERG (ETS Related Gene)

- ERG gene is located on Chr 21q22.2
- Member of ETS transcription factor family

ERG (ets-related gene)	21q22.2
ETV1	7p21.2
ETV4	17q21

- Most consistently overexpressed oncogene in malignant epithelial cells of the prostate *Petrovics et al. Oncogene. 2005*



TMRSS2-ERG Fusion & Prognosis

Perner et al. Cancer Res 2006

- FISH: fusion in 49% of 118 localized and 41% of 18 LN Mets
- Significant correlation between rearrangement through deletion and stage and presence of mets

Attard et al Oncogene 2007

- FISH, 445 cases
- Tumors without fusion had favorable PGx (90% @ 8yr)
- Tumors with fusion by deletion but not by translocation had worse OS and DSS
- Tumors with duplication of "fusion by deletion" had extremely poor PGx (25%@ 8 yr) independent of Gleason score

ERG EXPRESSION Status
IHC vs FISH

Immunohistochemistry for ERG Expression as a Surrogate for TMPRSS2-ERG Fusion Detection in Prostatic Adenocarcinomas

Alcides Chaux, MD, Roula Albadine, MD,* Antoun Toubaji, MD,* Jessica Hicks, BS,* Alan Meeker, PhD,*† Elizabeth A. Platz, ScD, MPH,†‡ Angelo M. De Marzo, MD, PhD,*†‡ and George J. Netto, MD*†‡*

A Chaux et al. Am J Surg Path 2011

ERG expression status
IHC vs FISH

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A Chaux et al. Am J Surg Pathol 2011

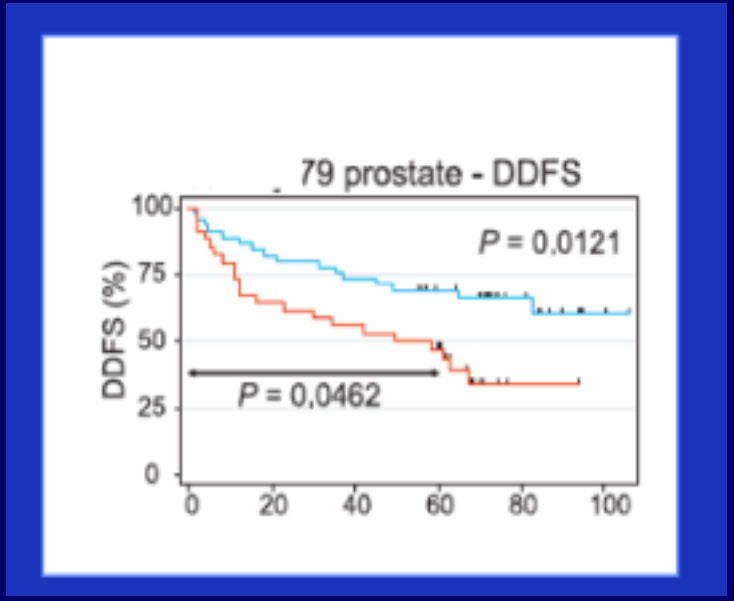
PTEN/PI3K/mTOR Pathway

- PTEN loss occurs in 20% of localized and over 60%
- of metastatic PCa and has been

associated with high Gleason score and pT stage and chemo resistance

Saal et al. PNAS 2007

Gene expression signature of aberrant PTEN activity is associated with poor prognosis in PCa



Loss of PTEN expression is associated with increased risk of recurrence after prostatectomy for clinically localized prostate cancer

Alcides Chaux[1,2], Sarah B Peskoe[3], Nilda Gonzalez-Roibon[1], Luciana Schultz[1], Roula Albadine[1], Jessica Hicks[1], Angelo M De Marzo[1,4,5], Elizabeth A Platz[3,4,5] and George J Netto[1,4,5]

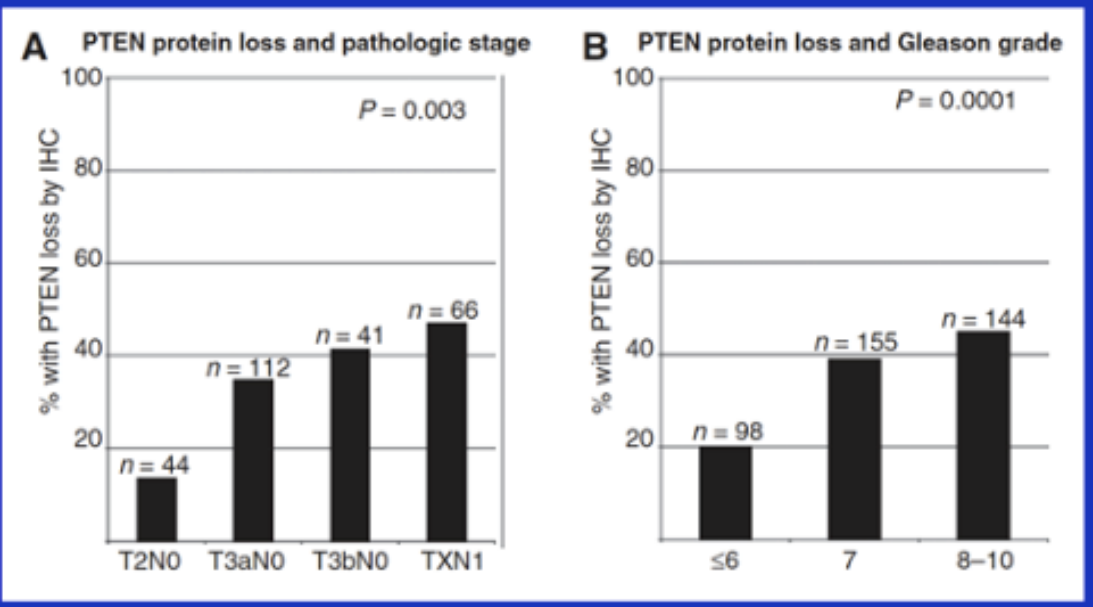
Modern Pathol 2012

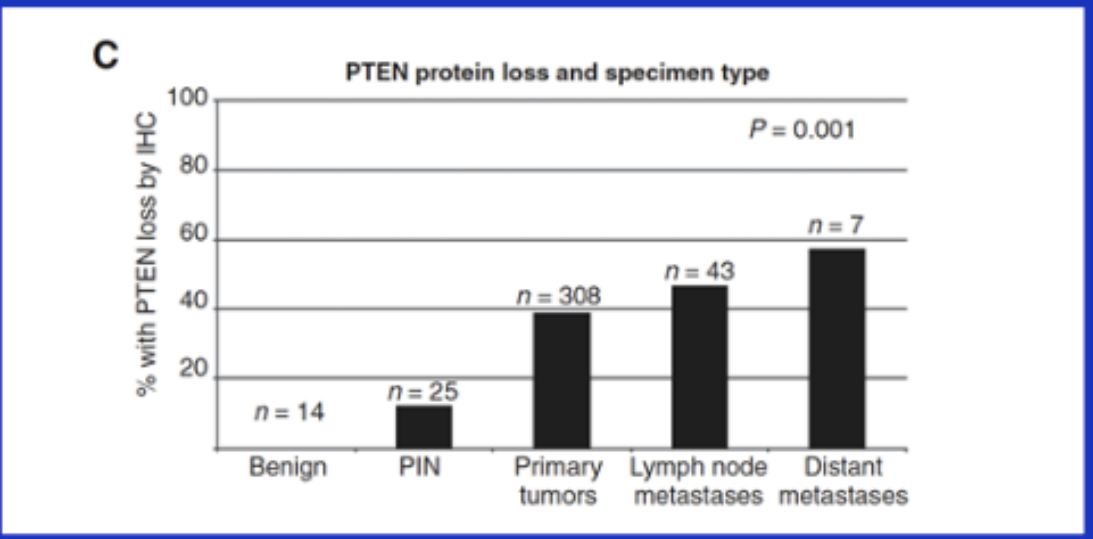
- 541 cases / 541 controls
- IHC nested case control study
- PTEN loss predicted recurrence (OR:2.2; p0.002)

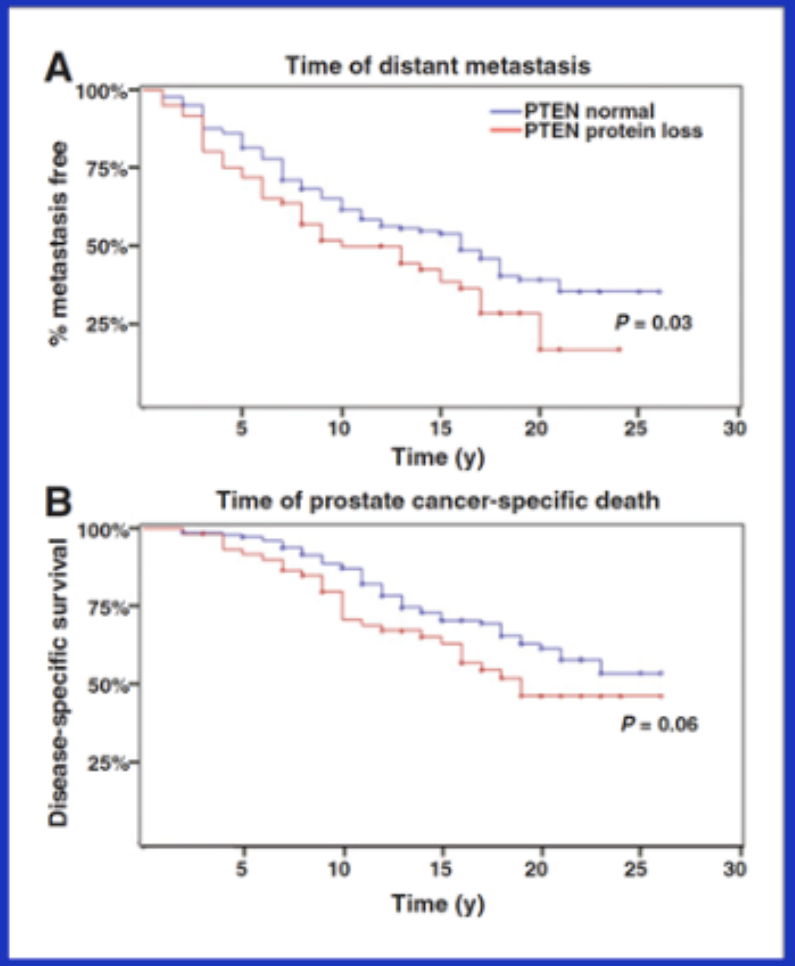
PTEN Protein Loss by Immunostaining: Analytic Validation and Prognostic Indicator for a High Risk Surgical Cohort of Prostate Cancer Patients

Tamara L. Lotan[1], Bora Gurel[1], Siobhan Sutcliffe[4], David Esopi[2], Wennuan Liu[5], Jianfeng Xu[5], Jessica L. Hicks[1], Ben H. Park[2], Elizabeth Humphreys[3], Alan W. Partin[3], Misop Han[3], George J. Netto[1,2,3], William B. Isaacs[2,3], and Angelo M. De Marzo[1,2,3]

Clin Cancer Research 2011


A
PTEN protein loss and pathologic stage
% with PTEN loss by IHC
100
80
60
40
20
P = 0.003
n = 44
n = 112
n = 41
n = 66
T2N0
T3aN0
T3bN0
TXN1
B
PTEN protein loss and Gleason grade
% with PTEN loss by IHC
100
80
60
40
20
P = 0.0001
n = 98
n = 155
n = 144
≤6
7
8–10


C
PTEN protein loss and specimen type
% with PTEN loss by IHC
100
80
60
40
20
P = 0.001
n = 14
n = 25
n = 308
n = 43
n = 7
Benign
PIN
Primary tumors
Lymph node metastases
Distant metastases


A
Time of distant metastasis
% metastasis free
100%
75%
50%
25%
PTEN normal
PTEN protein loss
P = 0.03
5
10
15
20
25
30
Time (y)
B
Time of prostate cancer-specific death
Disease-specific survival
100%
75%
50%
25%
P = 0.06
5
10
15
20
25
30
Time (y)

PTEN loss is associated with upgrading of prostate cancer from biopsy to radical prostatectomy

Lotan T et al. Mod Path 2014

- 71 cases with only Gleason score 6 on biopsy followed by a radical prostatectomy that contained Gleason score 7 or higher were compared to patients with Gleason score 6 on both biopsy and prostatectomy

- IHC for PTEN was performed, confirmed by FISH to detect PTEN gene deletion

- Tumors with PTEN protein loss were more likely to be upgraded (P=0.035) after adjusting for age, preoperative PSA, stage and race

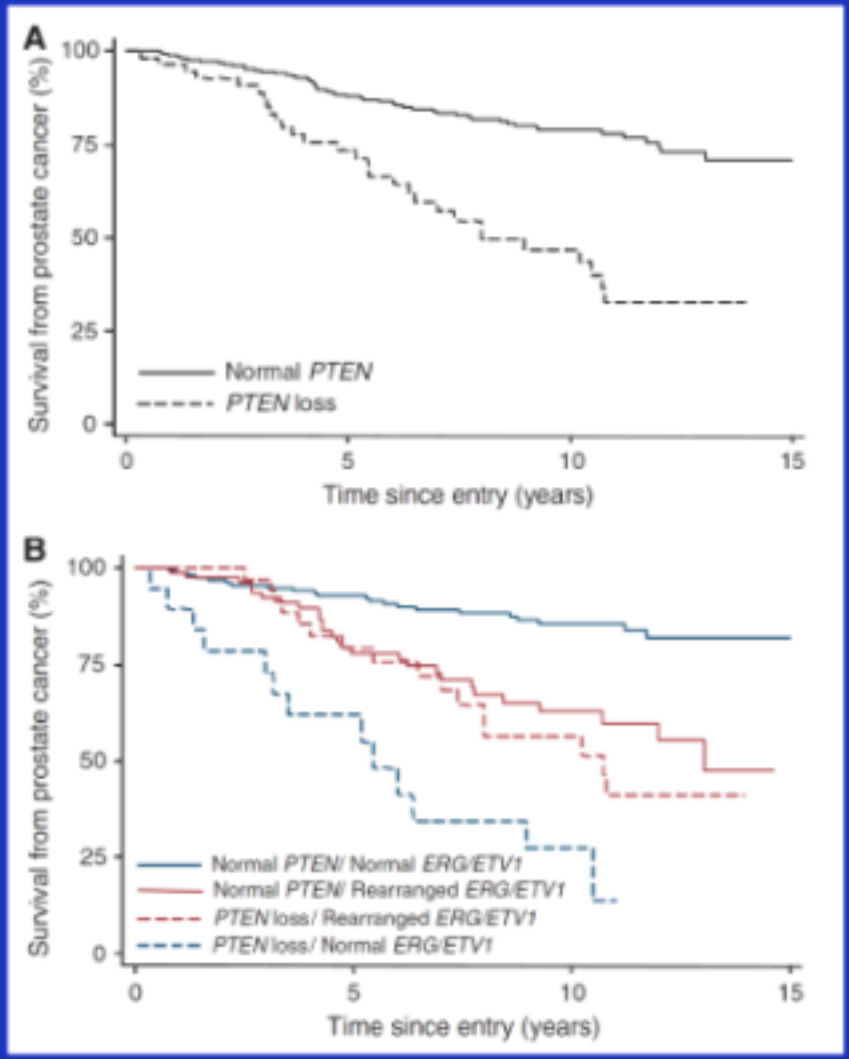
Genomic Deletion of *PTEN* Is Associated with Tumor Progression and Early PSA Recurrence in ERG Fusion-Positive and Fusion-Negative Prostate Cancer

- FISH on TMA study (4700 cases)
- PTEN mutations and methylation in subsets
- PTEN deletions in 20.2% (8.1% heterozyg ; 12.1% homozyg

Antje et al. Am J Pathol 2012

Molecular characterisation of *ERG*, *ETV1* and *PTEN* gene loci identifies patients at low and high risk of death from prostate cancer

Ried AHM et al. BJC 2010 Trans Atlantic Group



Molecular characterisation of *ERG*, *ETV1* and *PTEN* gene loci identifies patients at low and high risk of death from prostate cancer

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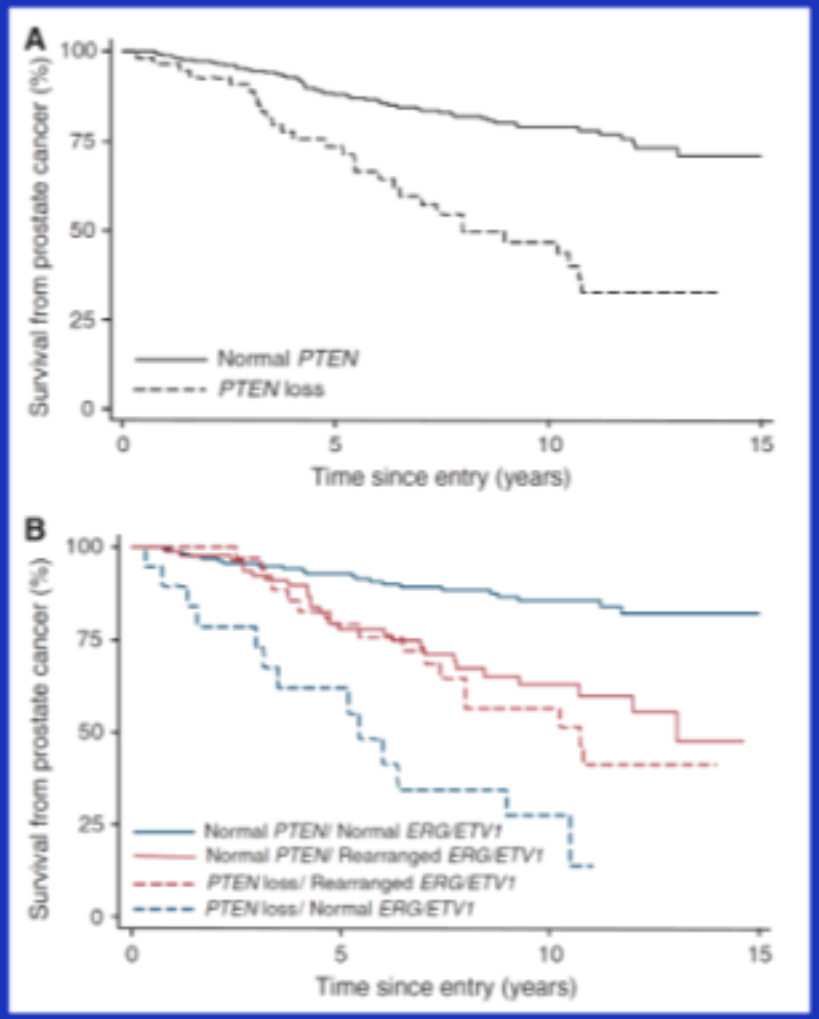
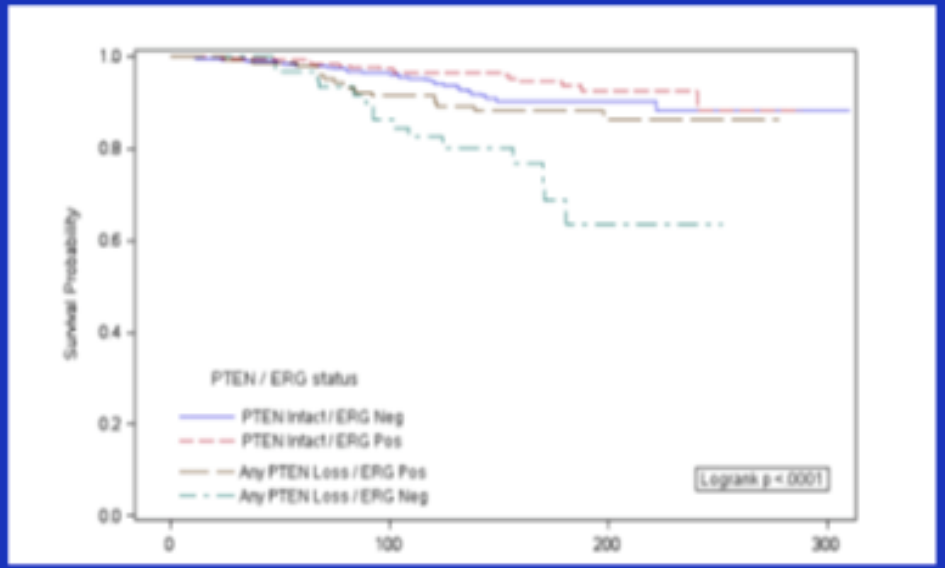


Aheam, Mucci and Lotan

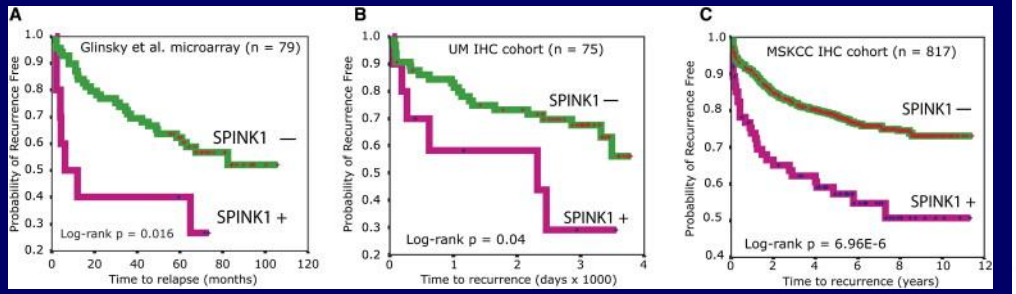
Health Professionals follow up and Ph

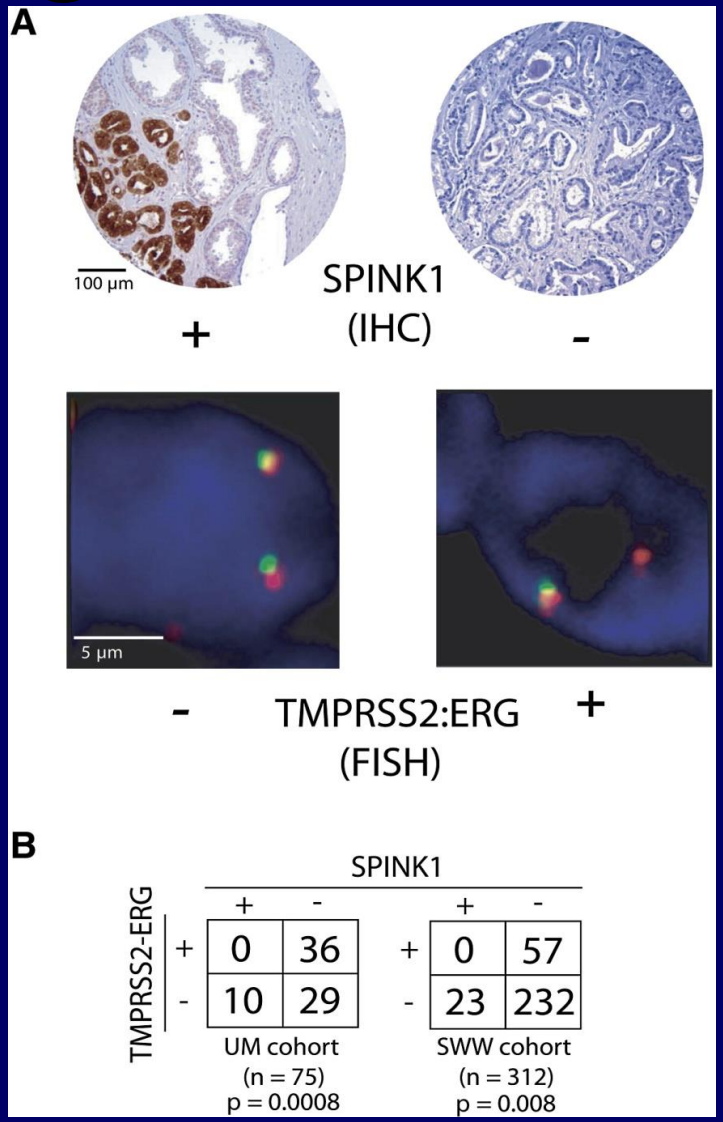
Health studies



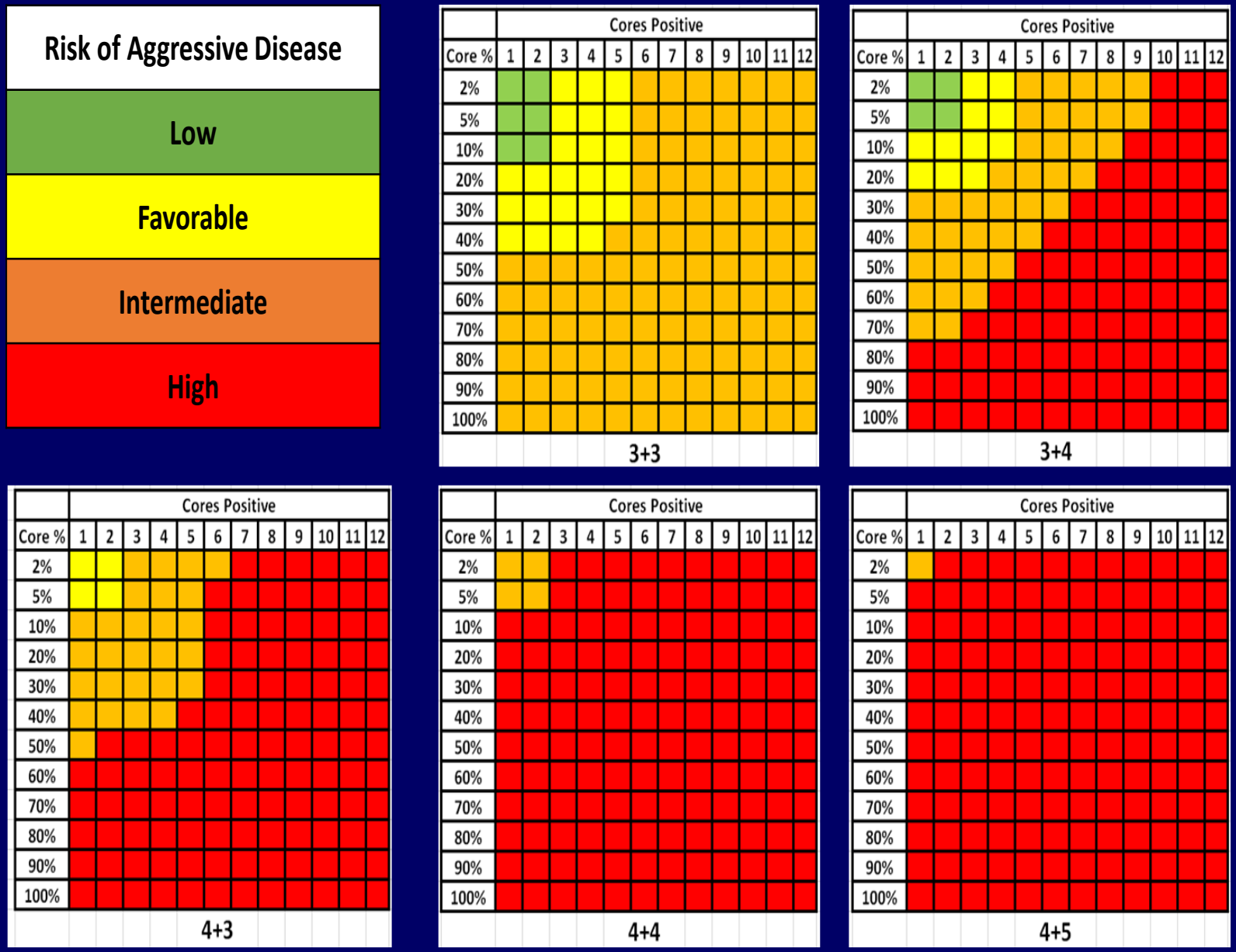
TMPRSS2-ETS negative PCA

- *Pancreatic secretory trypsin inhibitor (PSTI) also known as serine protease inhibitor Kazal-type 1 (SPINK1) or tumor-associated trypsin inhibitor (TATI) is a protein that in humans is encoded by the SPINK1*
- *SPINK1 (located on Chr5 5q32)* overexpression seem to be in the range of 10-15% of ERG negative tumors





S . Tomlins *et al.* Cancer Cell , Volume 13 , Issue 6 , Pages 519 - 528


Risk of Aggressive Disease
Low
Favorable
Intermediate
High
Cores Positive
Core %
1 2 3 4 5 6 7 8 9 10 11 12
2%
5%
10%
20%
30%
40%
50%
60%
70%
80%
90%
100%
3+3
3+4
4+3
4+4
4+5

Expression of **22 biomarkers** generates the Genomic Classifier score

Biomarkers correspond to RNA from both coding and non-protein coding regions of the genome

Marker	Nearest Gene/Locus	Type of Marker	Cytoband	Androgen Regulated[1]	Biological Process(es)	Reference(s) (PMID)
1	*LASP1*	CODING	17q12		Cell Proliferation, Differentiation	Grunewald et al, 2007 [17211471]; Traenka et al, 2010 [20924110]
2	*IQGAP3*	3′ UTR	1q23.1		Cell Proliferation, Differentiation	Nojima et al, 2008 [18604197]
3	*NFIB*	INTRONIC	9p23		Cell Proliferation, Differentiation	Qian et al, 1995 [7590749]; Dooley et al, 2011 [21764851]
4	*S1PR4*	3′ UTR	19p13.3		Cell Proliferation, Differentiation	Yamazaki et al, 2000 [10679247]
5	*THBS2*	3′ UTR	6q27		Cell Structure, Adhesion, Motility	Volpert et al, 1995 [8526929]; Kyriakides et al, 2001 [11583953]
6	*ANO7*	3′ UTR	2q37.3	Yes	Cell Structure, Adhesion, Motility	Das et al, 2008 [18676855]
7		NON-CODING TRANSCRIPT*				
8	*PCDH7*	INTRONIC	4p15.1	Yes	Cell Structure, Adhesion, Motility	Yoshida, 2003 [12949613]
9	*MYBPC1*	CODING	12q23.2	Yes	Cell Structure, Adhesion, Motility	Gregg et al, 2010 [20426842]
10		INTRONIC				
11	*EPPK1*	3′ UTR	8q24.3	Yes	Cell Structure, Adhesion, Motility	Yoshida et al, 2008 [18498355]
12	*TSBP*	INTRONIC	6p21.32		Immune Response	Liang et al, 1994 [7530381]
13	*PBX1*	CODING	1q23.3	Yes	Immune Response	Chung et al, 2007 [18093541]; Kikugawa et al, 2006 [16637071]; Qiu et al, 2007 [17200190]
14	*NUSAP1*	3′ UTR	15q15.1		Cell Cycle Progression, Mitosis	Raemaekers et al, 2003 [12963707]; Ribbeck et al, 2007 [17276916]
15	*ZWILCH*	3′ UTR	15q22.31		Cell Cycle Progression, Mitosis	Williams et al, 2003 [12686595]
16	*UBE2C*	3′UTR	20q13.12	Yes	Cell Cycle Progression, Mitosis	Rape and Kirschner, 2004 [15558010]
17		CODING ANTISENSE				
18	*CAMK2N1*	CODING ANTISENSE	1p36.12	Yes	Cell Cycle Progression, Mitosis	Wang et al, 2008 [18305109]
19	*RABGAP1*	EXON/INTRON JUNCTION ANTISENSE	9q33.2		Cell Cycle Progression, Mitosis	Culf et al, 1999 [10202141]
20	*PCAT-32*	NON-CODING TRANSCRIPT	5p15.2		Other, Unknown Function	Prensner et al, 2011 [21804560]
21	*GLYATL1P4/PCAT-80*	NON-CODING TRANSCRIPT	11q12.1		Other, Unknown Function	Prensner et al, 2011 [21804560]
22	*TNFRSF19*	INTRONIC	13q12.12		Other, Unknown Function	Eby et al, 2000 [10809768]

*Overlaps with an exon of a 'retained intron' category.
[1]Based on Jiang et al. Mol Endocrinol 23:1927-33, 2009; Massie et al. EMBO Rep 8:871-8, 2007.
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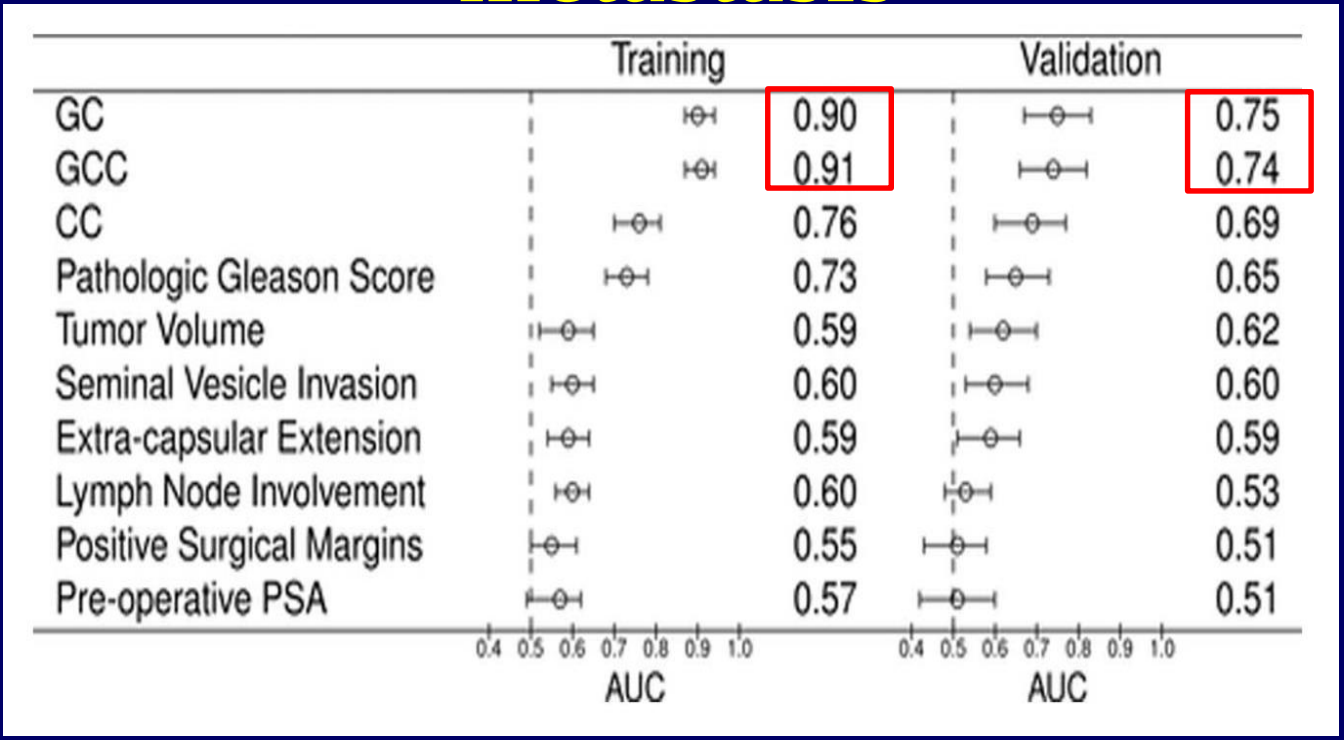
GC and GCC performed with highest accuracy to predict metastasis



- GCC added only marginal value to the prognosis of metastasis in the training set
- GC predicted metastasis with highest discriminatory performance in the validation set