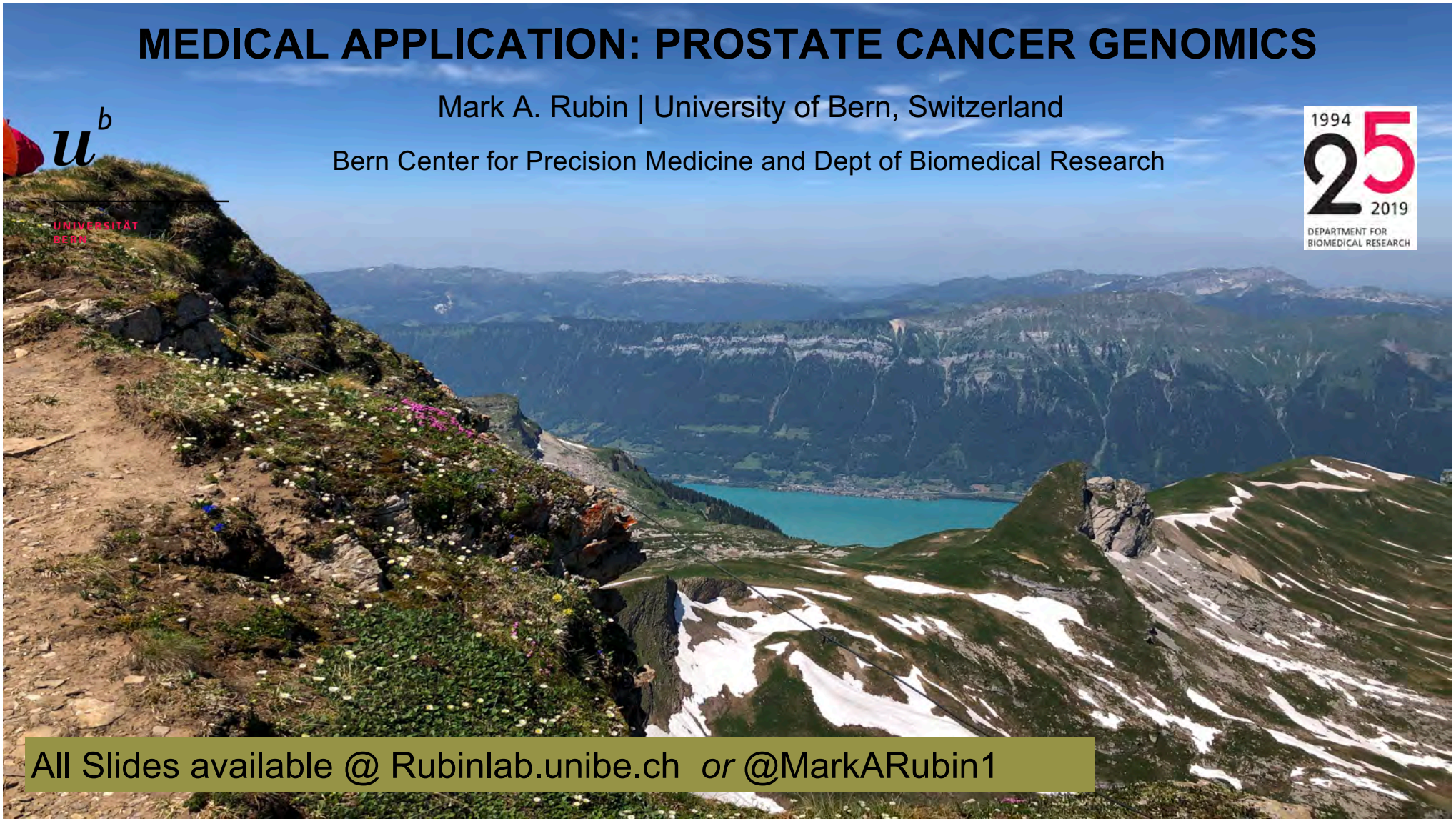
MEDICAL APPLICATION: PROSTATE CANCER GENOMICS

Mark A. Rubin | University of Bern, Switzerland

Bern Center for Precision Medicine and Dept of Biomedical Research



All Slides available @ Rubinlab.unibe.ch *or* @MarkARubin1

DISCLOSURES

FUNDING:

NCI, EDRN,PCF, SU2C/PCF, Starr Cancer Consortium, DOD, SNF, Krebsliga, SPHN
Sanofi-Aventis, Millennium Pharma, Eli-Lilly, and Janssen

PATENTS:

Listed as co-inventor on patents in the diagnostic and treatment fields for ETS fusions (Harvard/Michigan), EZh2 (Michigan), SPOP (Cornell), and AURKA (Cornell)

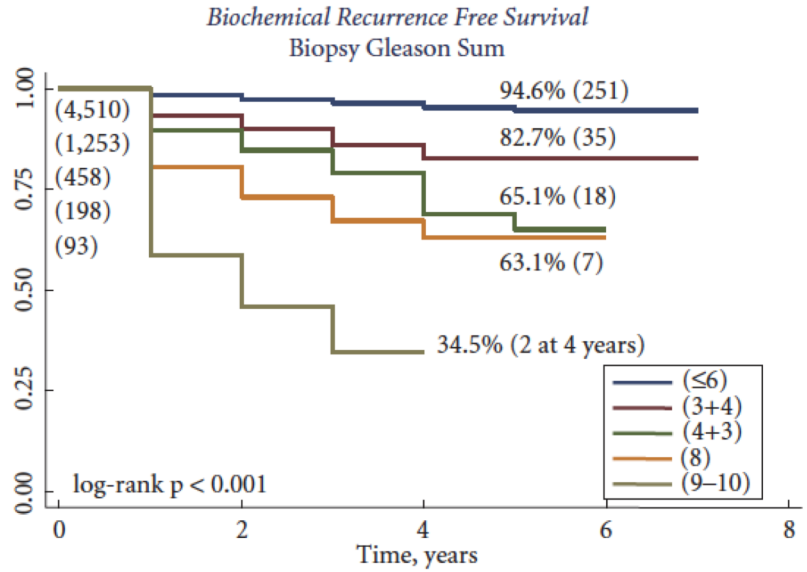
OFF-LABEL USE OF DRUGS WILL BE DISCUSSED

SAB NeoGenomics Laboratories, Inc.

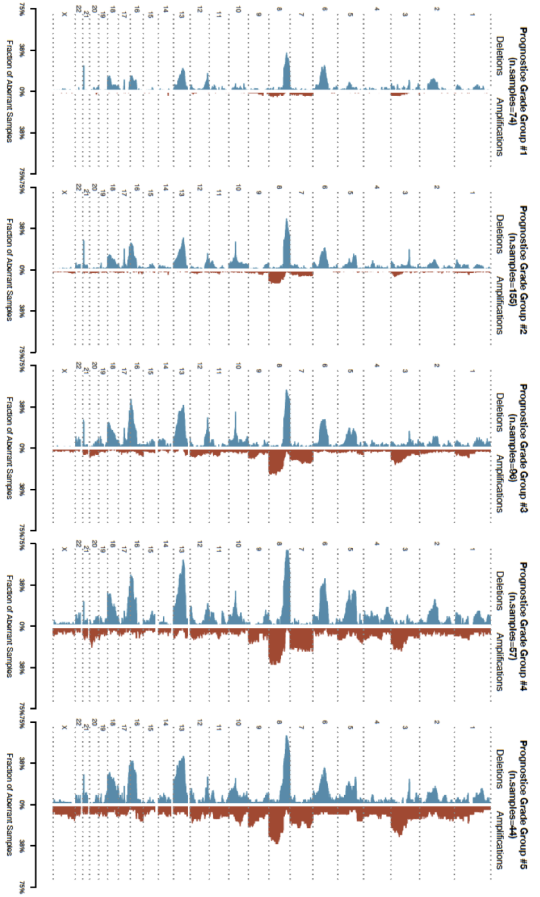
Primary Prostate Cancer

Prognostic Gleason grade grouping: data based on the modified Gleason scoring system

Phillip M. Pierorazio*, Patrick C. Walsh*, Alan W. Partin* and Jonathan I. Epstein*†‡

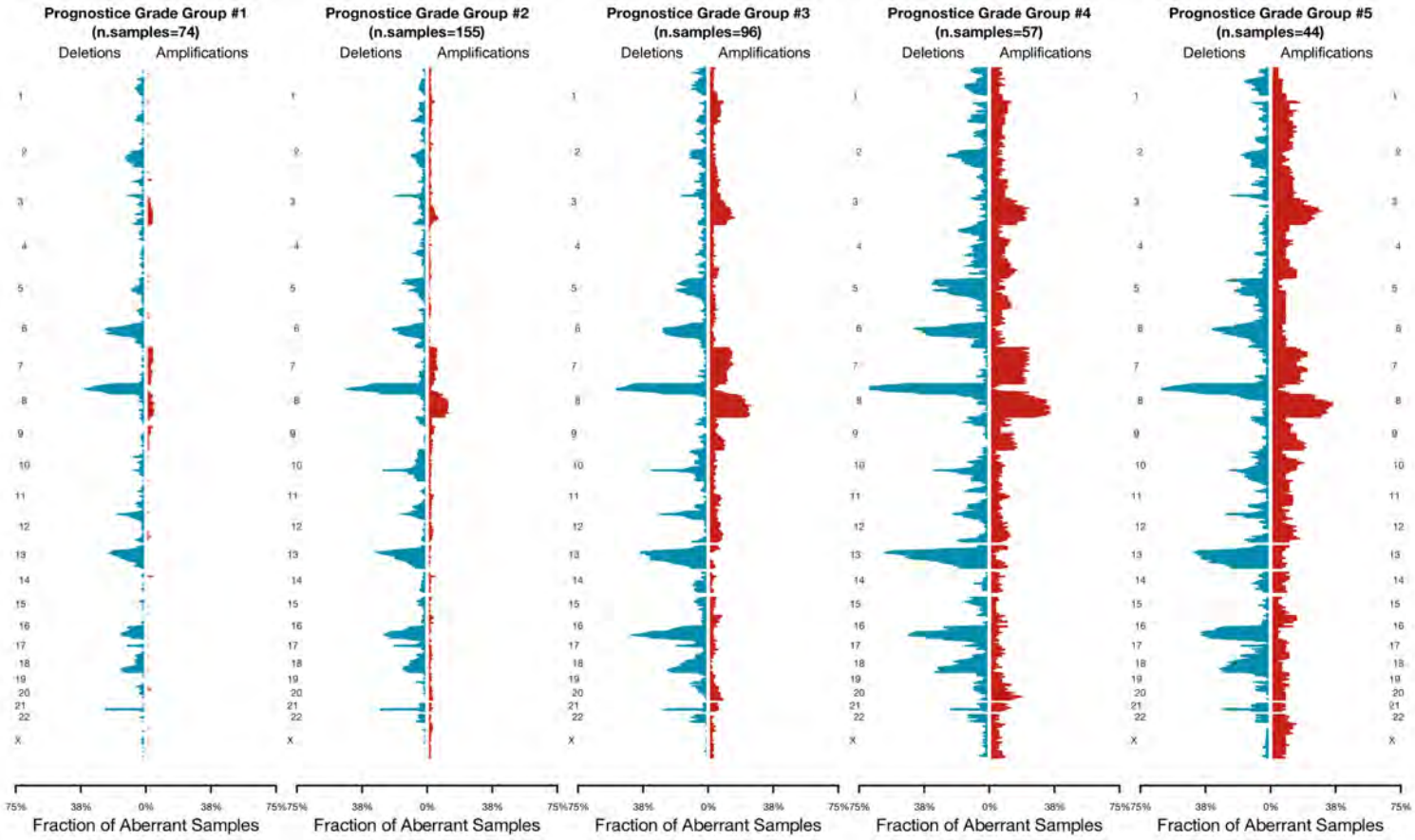


2013 BJU International



Rubin and Demichelis, Eur Urol 2016

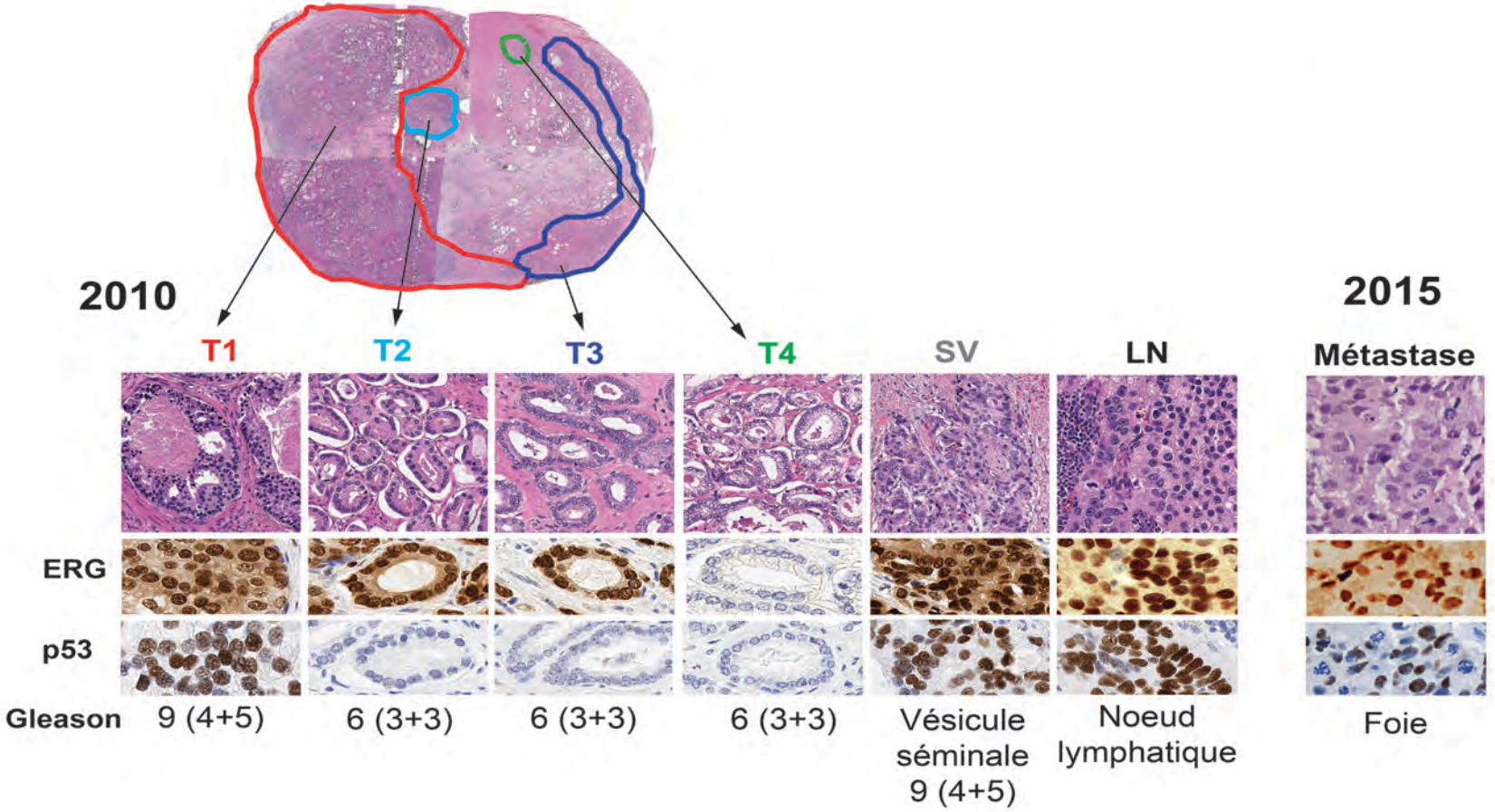
SCNA in Prostate Cancer with Increasing Risk Groups



6 3/4 4/3 4/4 4/5+

Rubin and Demichelis, Eur Urol 2016

The Problem: Heterogeneity in **Gleason Scores** & **Molecular Alterations**



Cyrta, Prandi, unpublished

Ten Minutes

1) ***Quick Summary*** of ISUP Report on molecular pathology prostate cancer (Localized Prostate Cancer)

2) ***Quick Summary*** of molecular pathology from the APCCC2019 (Advanced Prostate Cancer)

Report from the International Society of Urological Pathology (ISUP) Consultation Conference On Molecular Pathology Of Urogenital Cancers.

I. Molecular Biomarkers in Prostate Cancer

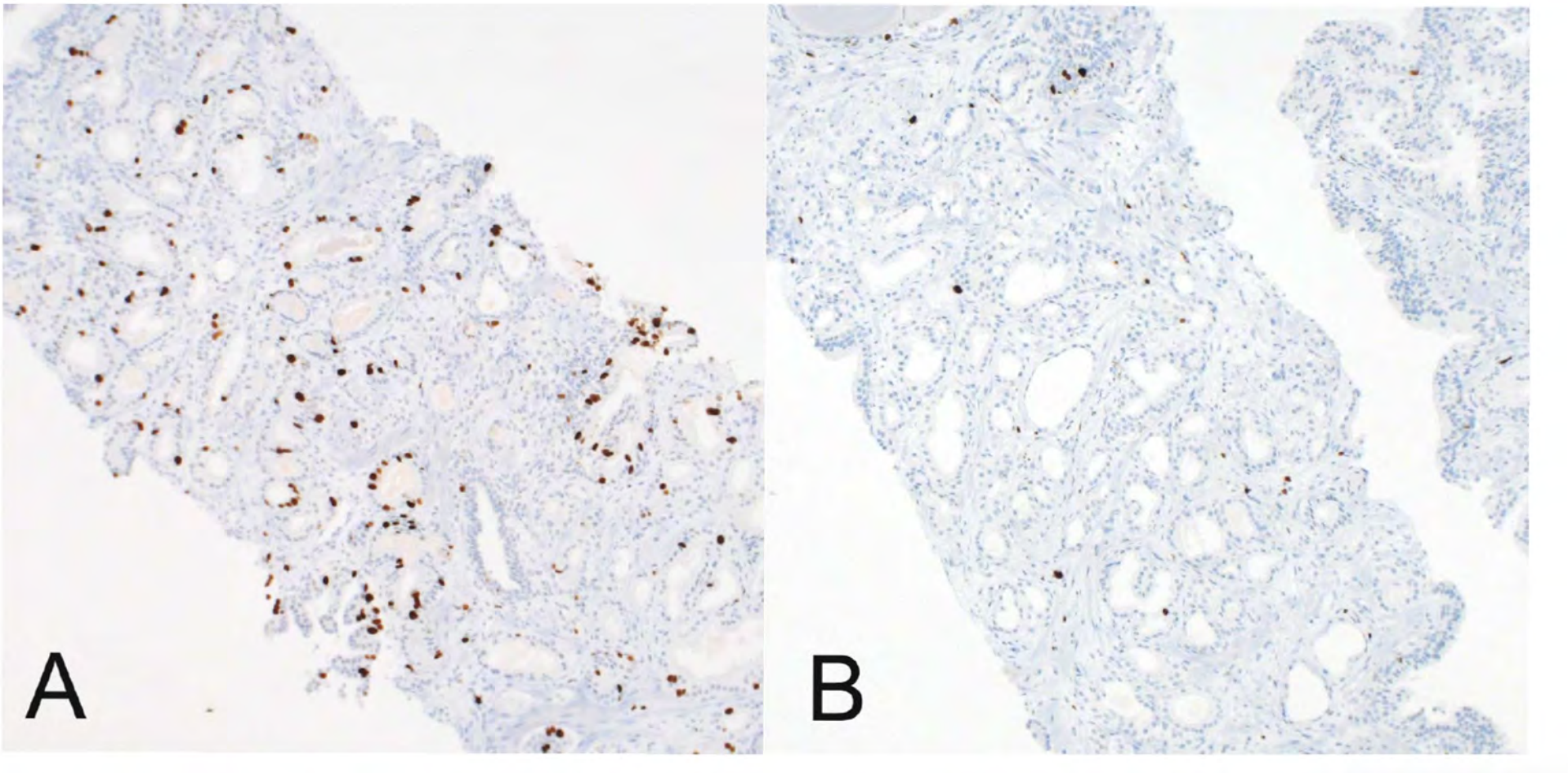
Tamara L. Lotan[1], Scott A. Tomlins[2], Tarek A. Bismar[3], Theodorus. H. Van der Kwast[4] , M.D., Ph.D., David Grignon[5] , M.D., Lars Egevad[6], M.D., Glen Kristiansen[7] **Colin C. Pritchard[8], Mark A. Rubin[9], Lukas Bubendorf[10]**

Sub-committee members in bold

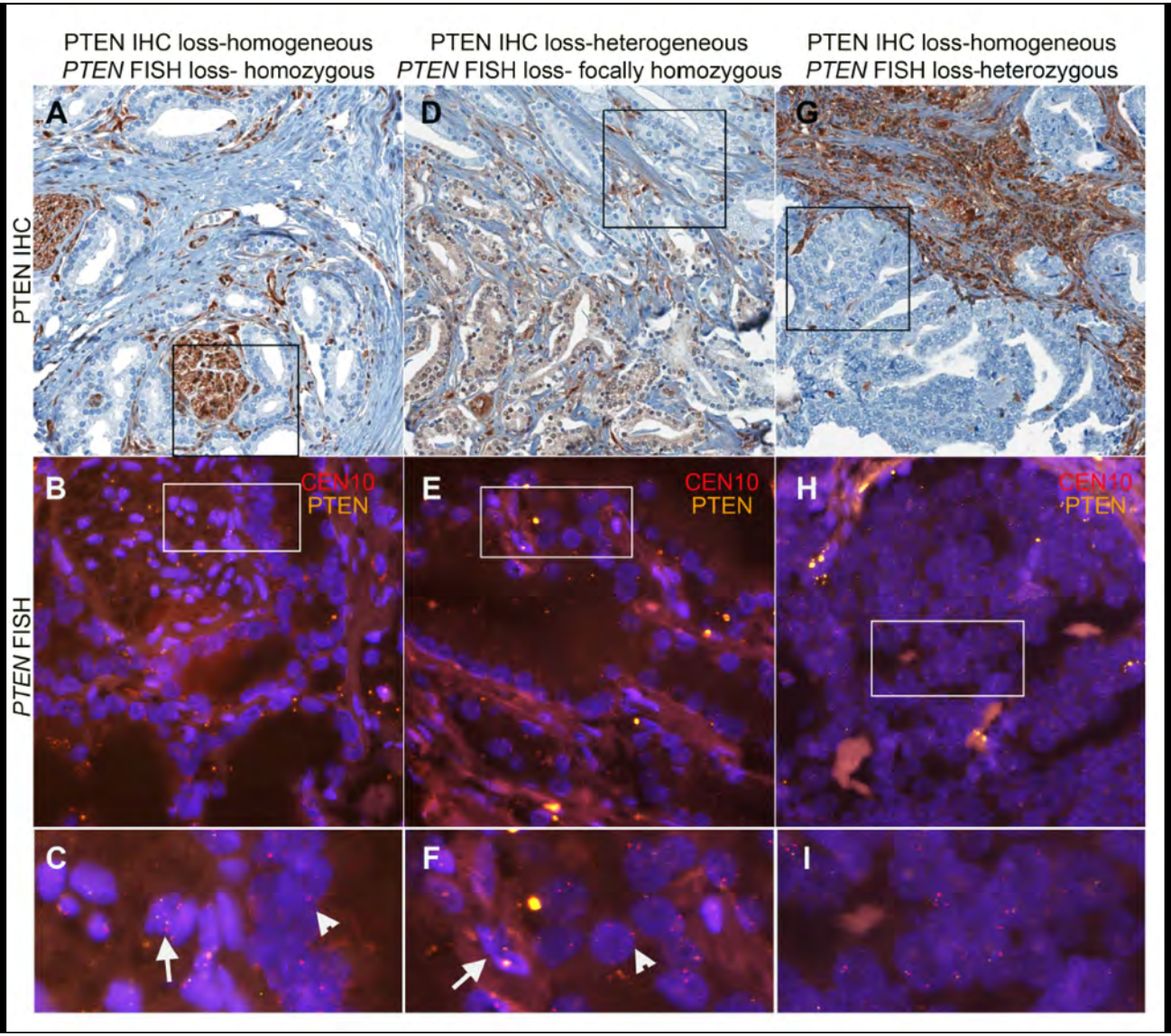
1Departments of Pathology, Oncology and Urology, Johns Hopkins University, Baltimore, MD, USA; 2Department of Pathology, Urology and Rogel Cancer Center, University of Michigan Medical School, Ann Arbor, MI, USA; 3Department of Pathology and Laboratory Medicine, University of Calgary Cumming School of Medicine and Alberta Public Labs, Calgary, AB, Canada; 4Laboratory Medicine Program, University Health Network, Toronto, Canada; 5Department of Pathology and Laboratory Medicine, Indiana University School of Medicine, Indiana, USA; 6Department of Oncology and Pathology, Karolinska Institutet Sweden; 7Department of Pathology, University Hospital Bonn, Germany 8Department of Laboratory Medicine, University of Washington, Seattle, WA, USA; 9Department for Biomedical Research, University of Bern and Bern Center for Precision Medicine, Bern, Switzerland; 10Institute of Pathology, University Hospital Basel, Basel, Switzerland.

Manuscript in Review

Ki-67 IHC Expression in two Prostate Cancer Biopsies



Report from ISUP Consultation Conference: Mol. Path Subgroup


PTEN IHC loss-homogeneous
PTEN FISH loss- homozygous
PTEN IHC loss-heterogeneous
PTEN FISH loss- focally homozygous
PTEN IHC loss-homogeneous
PTEN FISH loss-heterozygous
PTEN IHC
PTEN FISH
A
D
G
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H
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I
CEN10
PTEN

PTEN and Ki-67

Recommendations:

1) Ki-67 LI and PTEN are potentially useful prognostic biomarkers in the subset of Grade Group 1 (and/or Grade Group 2) prostate cancer biopsies where patient is eligible for active surveillance.

2) **High Ki-67 LI or PTEN loss** would be one factor (among several) suggesting that the patient should seek definitive treatment, <u>while intact PTEN would not be an informative result in this regard.</u>

Ki-67 and PTEN remain among the most promising prognostic molecular biomarkers studied to date, the Committee agreed that additional dedicated studies of active surveillance populations are warranted before widespread adoption.

Report from ISUP Consultation Conference: Mol. Path Subgroup

Table 1: Common Example of Commercial mRNA Based Tests Available Clinically

Test	Indication	Science	Results	Cost
Oncotype DX® Genomic Health	• Biopsy-based • NCCN Very Low, Low & Intermediate Risk • Provides personalized risk assessment	Assay looks at expression of 17 genes by RT-PCR within 4 pathways (androgen signaling, stromal response, cellular organization, proliferation) to assess tumor aggressiveness	• Genomic Prostate Score (GPS) from 0 to 100 • Likelihood of freedom from Dominant 4 or high-GS and/or non-organ confined disease • GPS is reflective of the biology of the tumor at the time of biopsy	• $3,820 • Medicare = No ABN required* • Other insurance: If estimated out-of-pocket cost>$100, company will contact the patient to offer financial assistance program.
Prolaris (CCP) Myriad Genetics	• Biopsy tissue based test for patients who are active surveillance candidates -or- • Post-prostatectomy tissue-based test to determine relative risk of BCR	46-gene expression signature by RT-PCR includes cell cycle progression genes (CCP) selected based upon correlation with prostate tumor cell proliferation	• Prolaris score • Biopsy is < or = or > than AUA risk group & estimates 10y mortality risk • Post-surgical is similar but 10y risk for BCR	• $3,400 • Medicare = No ABN required* • Other insurance: If estimated out-of-pocket cost>$375, company will contact patient to make arrangements...they have a financial assistance program.
Decipher® GenomeDx Biosciences	• Post-prostatectomy tissue-based test used for patients who are candidates for secondary therapy post surgery (pT2 with positive margins or any pT3 or BCR) • More recent studies in needle biopsy cohorts to predict adverse pathology and oncologic outcomes after RP	Analyzes the expression of 22 genes in multiple pathways using Affymetrix microarrays to measure the tumor's biological potential for metastasis	• Decipher reports the probability of metastasis at 5y after surgery and 3y after PSA recurrence.	• $4250 • Medicare =No ABN required*; CMS draft coverage policy published • Other ins: Company contacts all patients to offer financial assistance program. • Majority of patients have out-of-pocket cost no more than $395

Report from ISUP Consultation Conference: Mol. Path Subgroup

RNA-based genomic signatures are of potential benefit

However, the potential for under-sampling remains a major concern for all tissue-based tests.

1) Genomic signatures are of potential benefit but only if adequately sampled.

2) The improvement in such signatures should be compared to implementing robust pathological assessment and potential use of IHC biomarkers which needs further validation

3) Studies are needed to assess signatures performance in relation to heterogeneity

Report from ISUP Consultation Conference: Mol. Path Subgroup

Predictive Biomarkers:

Predictive biomarkers estimate the chances of **response** to a specific therapy

To date, predictive biomarkers have largely been studied in the context of **metastatic disease.** Despite their importance for precision medicine, relatively few tissue-based predictive biomarkers have been validated.

DNA Repair Deficiency Markers

NCCN guidelines for prostate cancer recommend germline testing in high risk subsets of patients with clinically localized prostate cancer

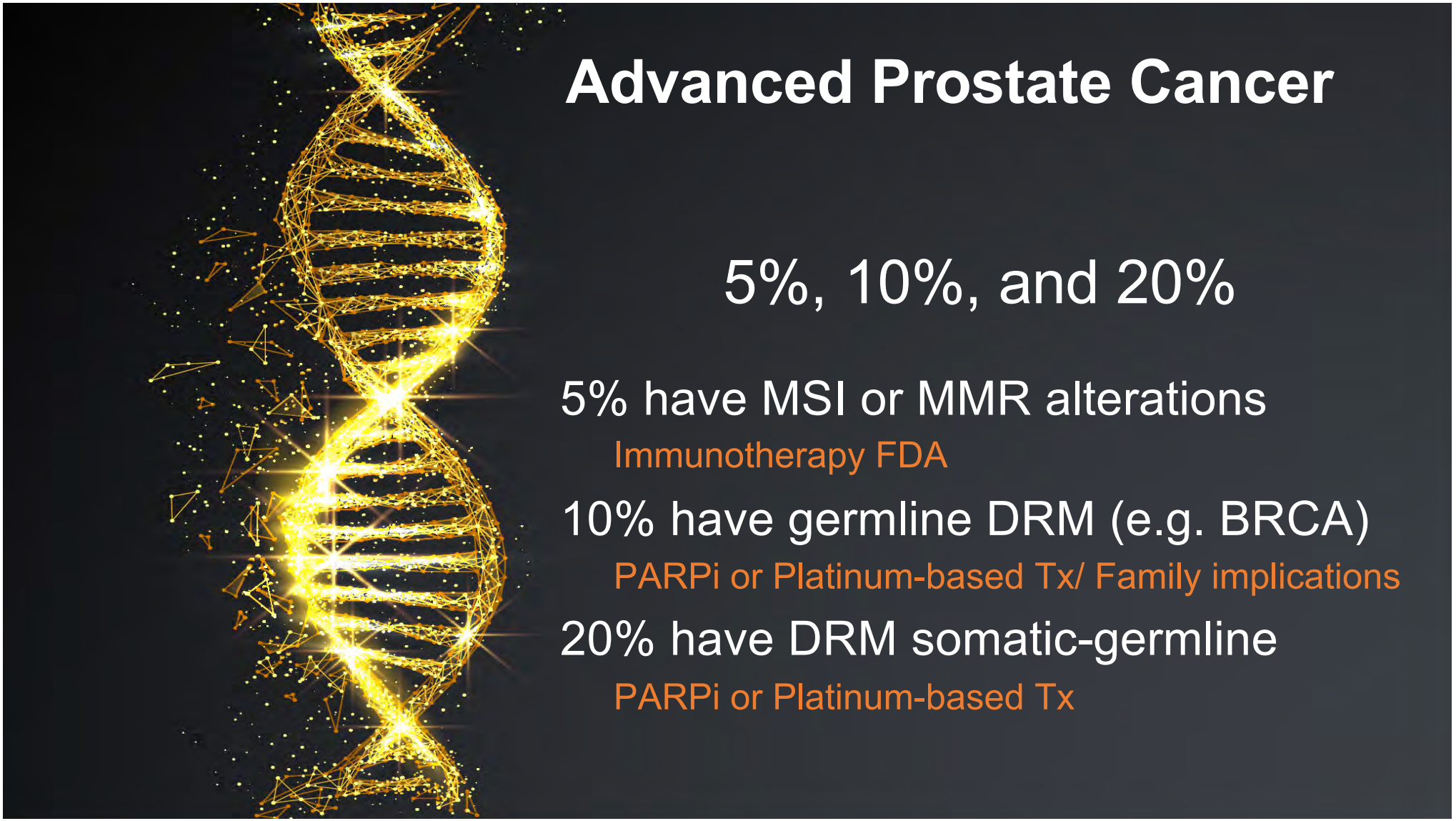
Grade Group 4 or higher tumors or

patients with PSA of 20 ng/mL or higher.

In addition, **germline testing** should be performed in all patients with metastatic prostate cancer if clinically indicated, **with appropriate genetic counseling.**

Currently, all metastatic patients should also be offered somatic genomic testing of tumor tissue for HRD and MMR defects if clinically indicated.

Report from ISUP Consultation Conference: Mol. Path Subgroup

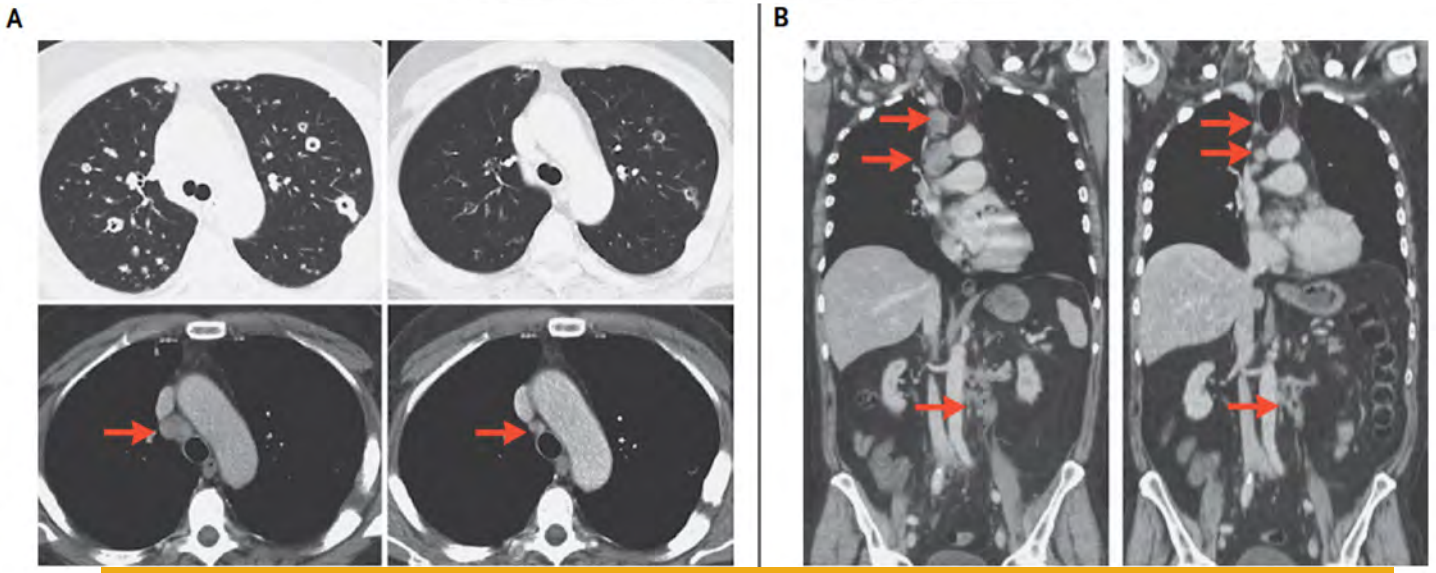

Advanced Prostate Cancer
5%, 10%, and 20%
5% have MSI or MMR alterations
Immunotherapy FDA
10% have germline DRM (e.g. BRCA)
PARPi or Platinum-based Tx/ Family implications
20% have DRM somatic-germline
PARPi or Platinum-based Tx

The NEW ENGLAND JOURNAL of MEDICINE

ESTABLISHED IN 1812 OCTOBER 29, 2015 VOL. 373 NO. 18

DNA-Repair Defects and Olaparib in Metastatic Prostate Cancer

J. Mateo, S. Carreira, S. Sandhu, S. Miranda, H. Mossop, R. Perez-Lopez, D. Nava Rodrigues, D. Robinson, A. Omlin, N. Tunariu, G. Boysen, N. Porta, P. Flohr, A. Gillman, I. Figueiredo, C. Paulding, G. Seed, S. Jain, C. Ralph, A. Protheroe, S. Hussain, R. Jones, T. Elliott, U. McGovern, D. Bianchini, J. Goodall, Z. Zafeiriou, C.T. Williamson, R. Ferraldeschi, R. Riisnaes, B. Ebbs, G. Fowler, D. Roda, W. Yuan, Y.-M. Wu, X. Cao, R. Brough, H. Pemberton, R. A'Hern, A. Swain, L.P. Kunju, R. Eeles, G. Attard, C.J. Lord, A. Ashworth, M.A. Rubin, K.E. Knudsen, F.Y. Feng, A.M. Chinnaiyan, E. Hall, and J.S. de Bono



TOPARP Trial shows 30% Long Term Responders

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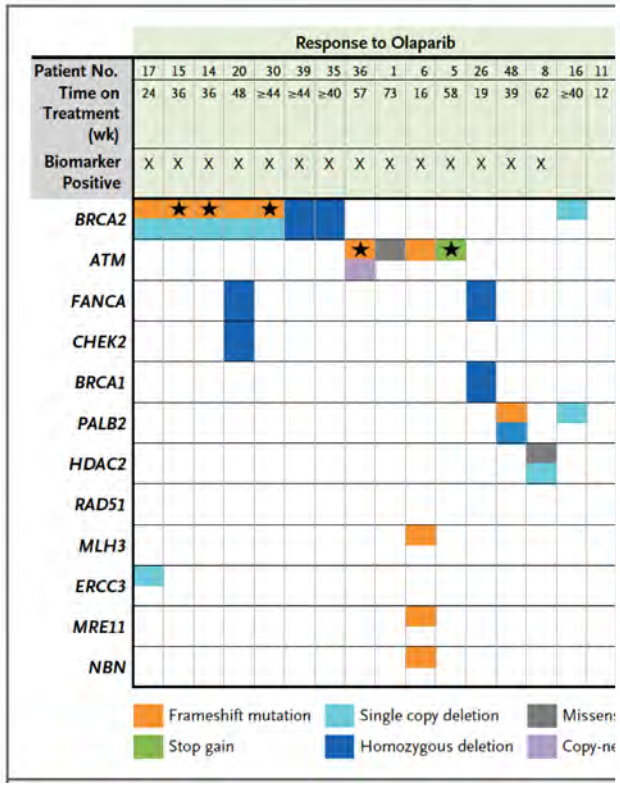
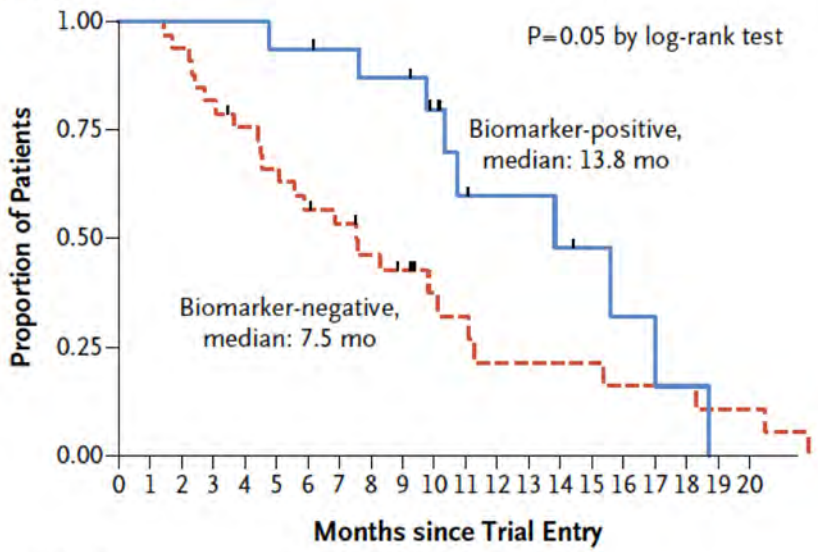
NEJM, Oct 29 2015



B Overall Survival



No. at Risk

Biomarker-negative	33	33	31	27	24	21	18	16	13	11	7	6	4	4	4	4	3	3	3	2	2
Biomarker-positive	16	16	16	16	16	15	15	14	13	13	10	6	5	5	4	3	2	2	1	0	0

No. of Events

Biomarker-negative	0	2	4	2	3	3	1	2	1	1	1	2	0	0	0	1	0	0	1	0	–
Biomarker-positive	0	0	0	0	1	0	0	1	0	1	2	0	0	1	0	1	0	2	0	0	–

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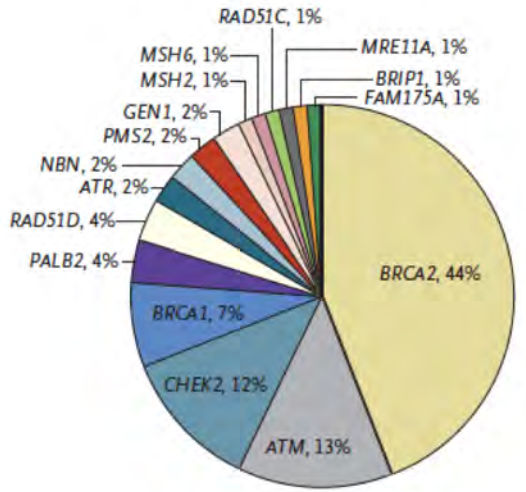
The NEW ENGLAND JOURNAL of MEDICINE

ORIGINAL ARTICLE

Inherited DNA-Repair Gene Mutations in Men with Metastatic Prostate Cancer

Table 2. Germline Mutations in Metastatic Cases as Compared with the General Population and Primary Cases.

Gene	Metastatic Prostate Cancer (N=692)*	Exome Aggregation Consortium (N=53,105)†	TCGA Cohort with Primary Prostate Cancer (N=499)	Metastatic Prostate Cancer vs. Exome Aggregation Consortium		Metastatic Prostate Cancer vs. TCGA Cohort	
	No. of Mutations (% of Men)			Relative Risk (95% CI)	P Value	Relative Risk (95% CI)	P Value
ATM	11 (1.59)	133 (0.25)	5 (1.00)	6.3 (3.2–11.3)	<0.001	1.6 (0.8–2.8)	0.12
ATR	2 (0.29)	43 (0.08)	0	3.6 (0.4–12.8)	0.11	—	—
BAP1‡	0	1	0	—	—	—	—
BARD1‡	0	38 (0.07)	1 (0.20)	—	—	—	—
BRCA1	6 (0.87)	104 (0.22)	3 (0.60)	3.9 (1.4–8.5)	0.005	1.4 (0.5–3.1)	0.32
BRCA2	37 (5.35)	153 (0.29)	1 (0.20)	18.6 (13.2–25.3)	<0.001	26.7 (18.9–36.4)	<0.001
BRIP1‡	1 (0.18)	100 (0.19)	1 (0.20)	0.9 (0.02–5.3)	1.0	0.9 (0.0–4.9)	1.0
CHEK2‡	10 (1.87)	314 (0.61)	2 (0.40)	3.1 (1.5–5.6)	0.002	4.7 (2.2–8.5)	<0.001
FAM175A‡	1 (0.18)	52 (0.10)	0	1.8 (0.05–10.1)	0.42	—	—
GEN1‡	2 (0.46)	42 (0.08)	0	5.8 (0.7–20.8)	0.048	—	—
MLH1	0	11 (0.02)	0	—	—	—	—
MRE11A	1 (0.14)	36 (0.07)	1 (0.20)	2.1 (0.1–11.8)	0.38	0.7 (0.0–4.0)	1.0
MSH2	1 (0.14)	23 (0.04)	1 (0.20)	3.3 (0.1–18.5)	0.26	0.7 (0.0–4.0)	1.0
MSH6	1 (0.14)	41 (0.08)	1 (0.20)	1.9 (0.05–10.4)	0.41	0.7 (0.0–4.0)	1.0
NBN	2 (0.29)	61 (0.11)	1 (0.20)	2.5 (0.3–9.1)	0.19	1.4 (0.2–5.2)	0.40
PALB2	3 (0.43)	65 (0.12)	2 (0.40)	3.5 (0.7–10.3)	0.05	1.1 (0.2–3.1)	0.76
PMS2	2 (0.29)	56 (0.11)	1 (0.20)	2.7 (0.3–9.8)	0.17	1.4 (0.2–5.2)	0.40
RAD51C	1 (0.14)	59 (0.11)	2 (0.40)	1.3 (0.03–7.2)	0.54	0.4 (0.0–2.0)	0.54
RAD51D	3 (0.43)	40 (0.08)	1 (0.20)	5.7 (1.2–16.7)	0.02	2.2 (0.4–6.3)	0.16
XRCC2	0	23 (0.04)	0	—	—	—	—



Selected *DNA repair germline mutations from targeted panel and WES reveal 10-20% frequency* (Pritchard and Nelson, 2016)

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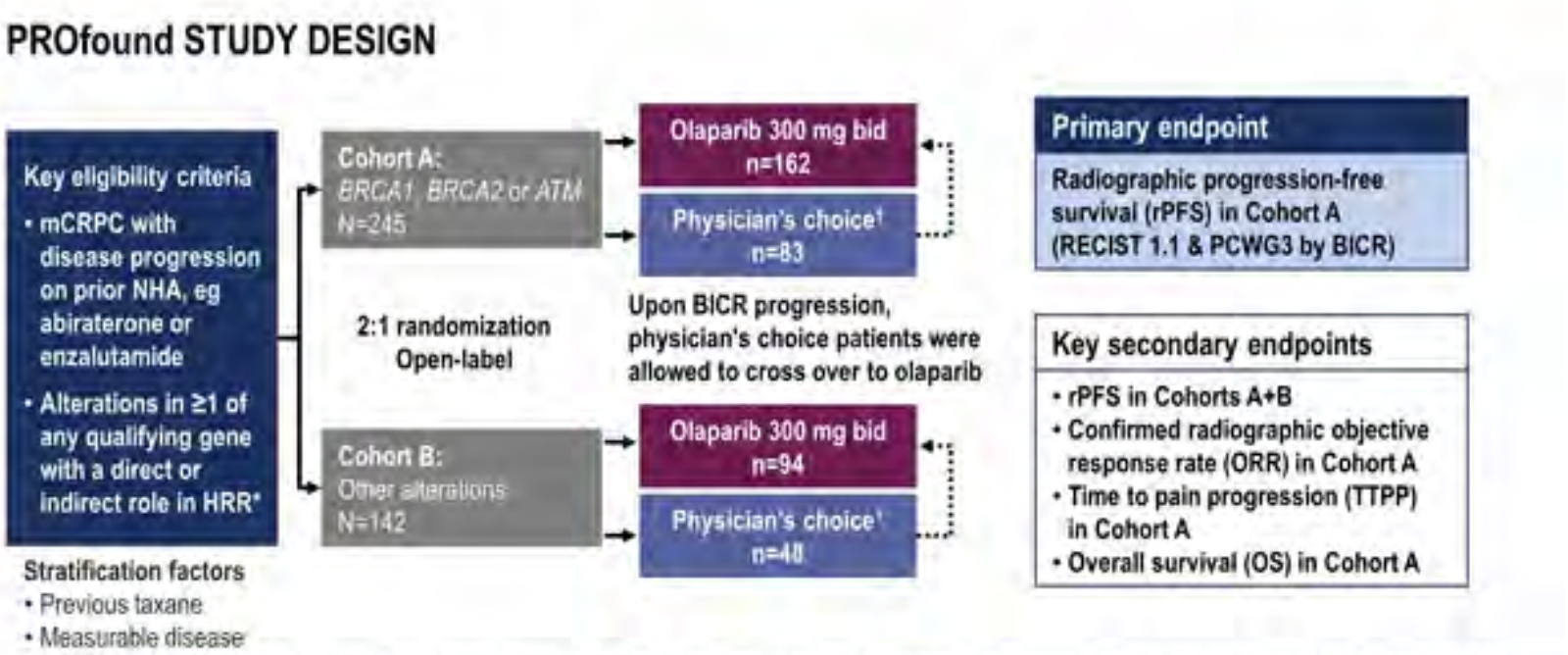
ESMO 2019: PROfound: Phase 3 Study of Olaparib vs. Enzalutamide or Abiraterone for Metastatic Castration-Resistant Prostate Cancer with Homologous Recombination Repair Gene Alterations



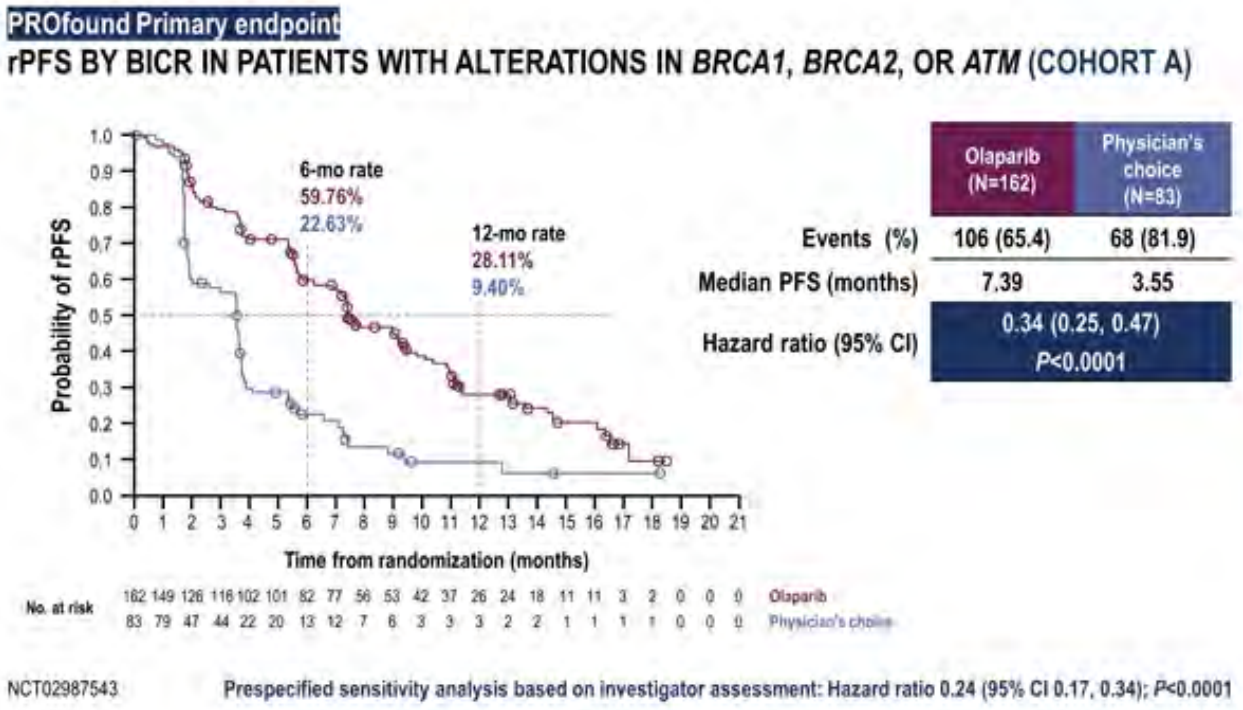
*An investigational Clinical Trial Assay, based on the FoundationOne CDx next-generation sequencing test, and developed in partnership with Foundation Medicine Inc, was used to prospectively select patients harboring alterations in the following genes in their tumor tissue: *BRCA1, BRCA2, ATM, BARD1, BRIP1, CDK12, CHEK1, CHEK2, FANCL, PALB2, PPP2R2A, RAD51B, RAD51C, RAD51D* or *RAD54L*

†Physician's choice of either enzalutamide (160 mg qd) or abiraterone (1000 mg qd plus prednisone [5 mg bid])

BICR, blinded independent central review; PCWG3, Prostate Cancer Working Group 3; RECIST, Response Evaluation Criteria in Solid Tumors

NCT02987543

ESMO 2019: PROfound: Phase 3 Study of Olaparib vs. Enzalutamide or Abiraterone for Metastatic Castration-Resistant Prostate Cancer with Homologous Recombination Repair Gene Alterations



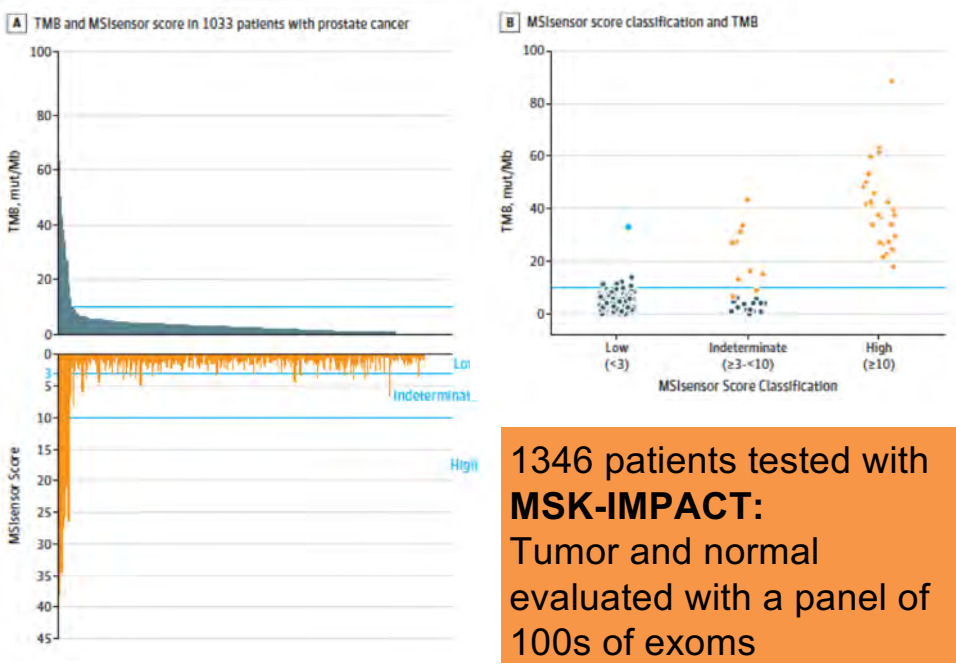
Among the men who underwent screening, 4047 had samples that were tested, among which 2792 (69%) were successfully sequenced and yielded biomarker status. In screened patients, samples were mainly derived from archived tissue (89.9%); most archived samples (79.7%) were from the primary tumour and 10.1% were derived from metastatic tissue. ESMO 2019

JAMA Oncology | Original Investigation

Analysis of the Prevalence of Microsatellite Instability in Prostate Cancer and Response to Immune Checkpoint Blockade

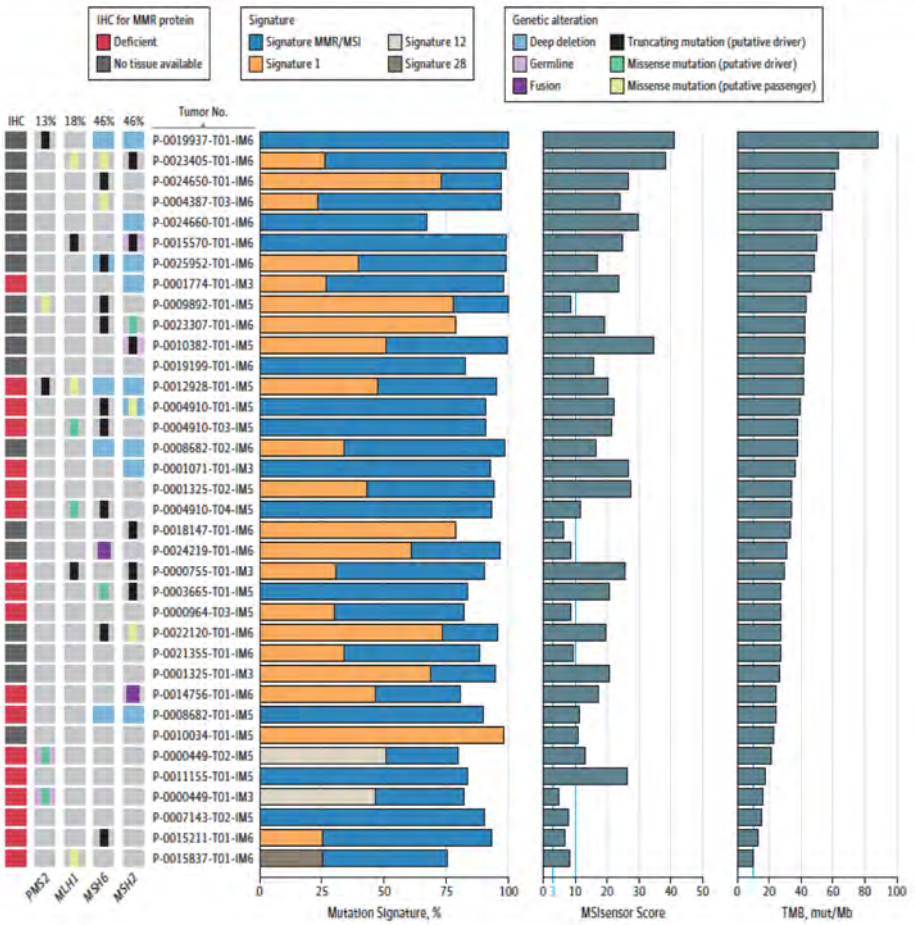
Wassim Abida, MD, PhD; Michael L. Cheng, MD; Joshua Armenia, PhD; Sumit Middha, PhD; Karen A. Autio, MD; Hebert Alberto Vargas, MD; Dana Rathkopf, MD; Michael J. Morris, MD; Daniel C. Danila, MD; Susan F. Slovin, MD, PhD; Emily Carbone, BA; Ethan S. Barnett, MS; Melanie Hullings, BA; Jaclyn F. Hechtman, MD; Ahmet Zehir, PhD; Jinru Shia, MD; Philip Jonsson, PhD; Zsofia K. Stadler, MD; Preethi Srinivasan, BA; Vincent P. Laudone, MD; Victor Reuter, MD; Jedd D. Wolchok, MD, PhD; Nicholas D. Socci, PhD; Barry S. Taylor, PhD; Michael F. Berger, PhD; Philip W. Kantoff, MD; Charles L. Sawyers, MD; Nikolaus Schultz, PhD; David B. Solit, MD; Anuradha Gopalan, MD; Howard I. Scher, MD

Figure 1. Tumor Mutation Burden (TMB) and Microsatellite Instability (MSI) in Prostate Cancer



1346 patients tested with **MSK-IMPACT:**
Tumor and normal evaluated with a panel of 100s of exoms

Figure 2. Integrative Analysis of Microsatellite Instability (MSI), Tumor Mutation Burden (TMB), Mutational Signature Decomposition, and Mismatch Repair (MMR) Gene and Protein Status



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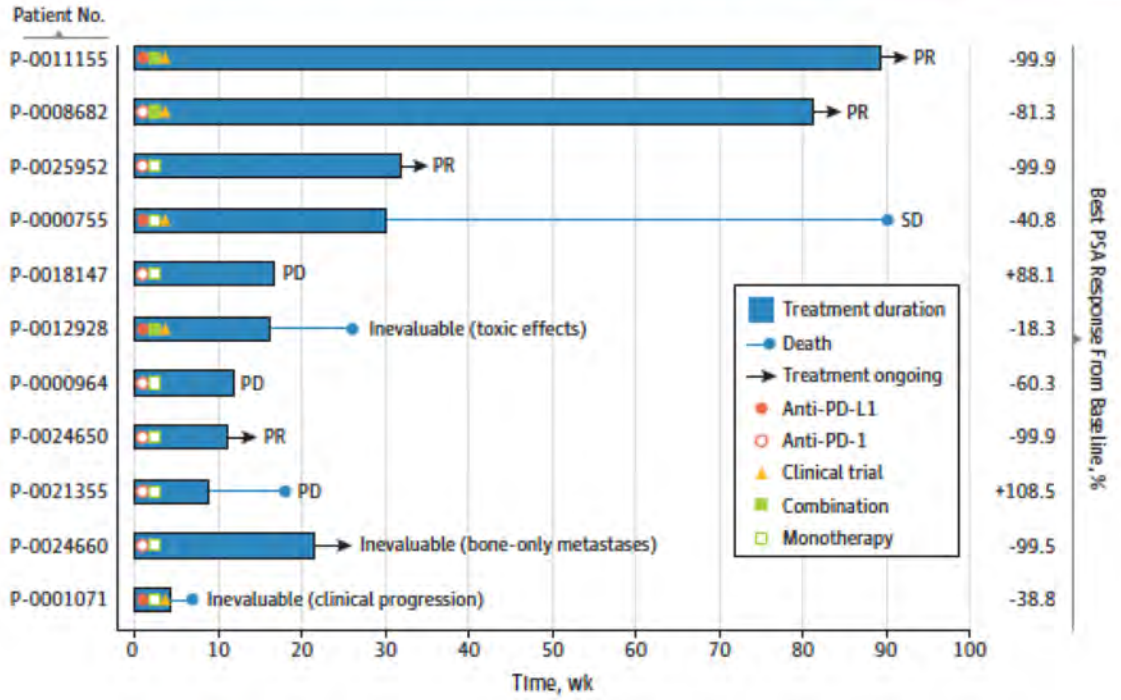
JAMA Oncology Published online December 27, 2018

Figure 4. Responses to Immune Checkpoint Blockade in Microsatellite Instability–High and Mismatch Repair Deficient (MSI-H/dMMR) Prostate Cancer



M.A.Rubin Copyright

JAMA Oncology Published online December 27, 2018

Recommendations of the Working Group were the following:

In combination with appropriate genetic counseling, germline panel testing for DNA repair gene alterations should be offered (if clinically indicated) to patients with:

Localized Grade Group ≥4 tumors

Any Grade Group with PSA ≥20

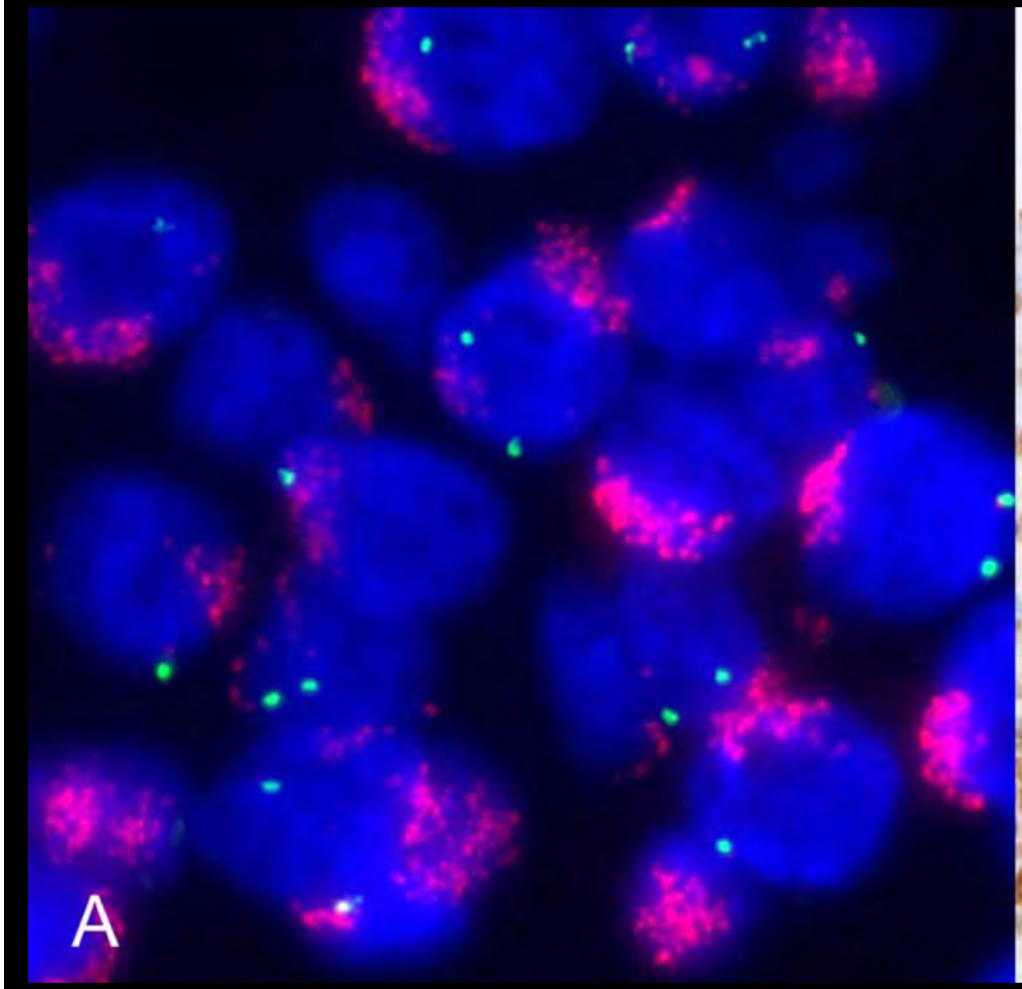
Known metastatic disease

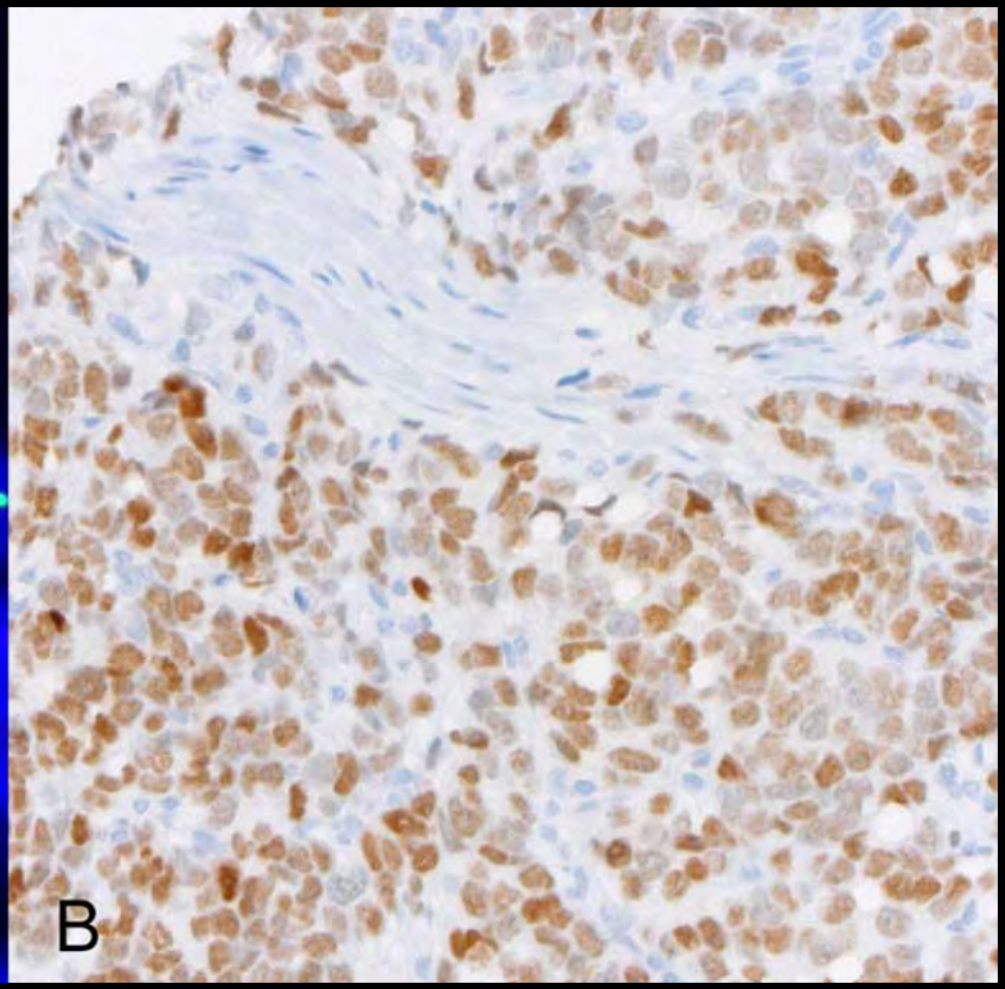
Testing should include:

1) Defective MMR assessment via MMR IHC for MSH2, MSH6, MLH-1, PMS2 with or without MSI testing and/or sequencing of MMR genes (and tumor mutation burden estimate)

AND

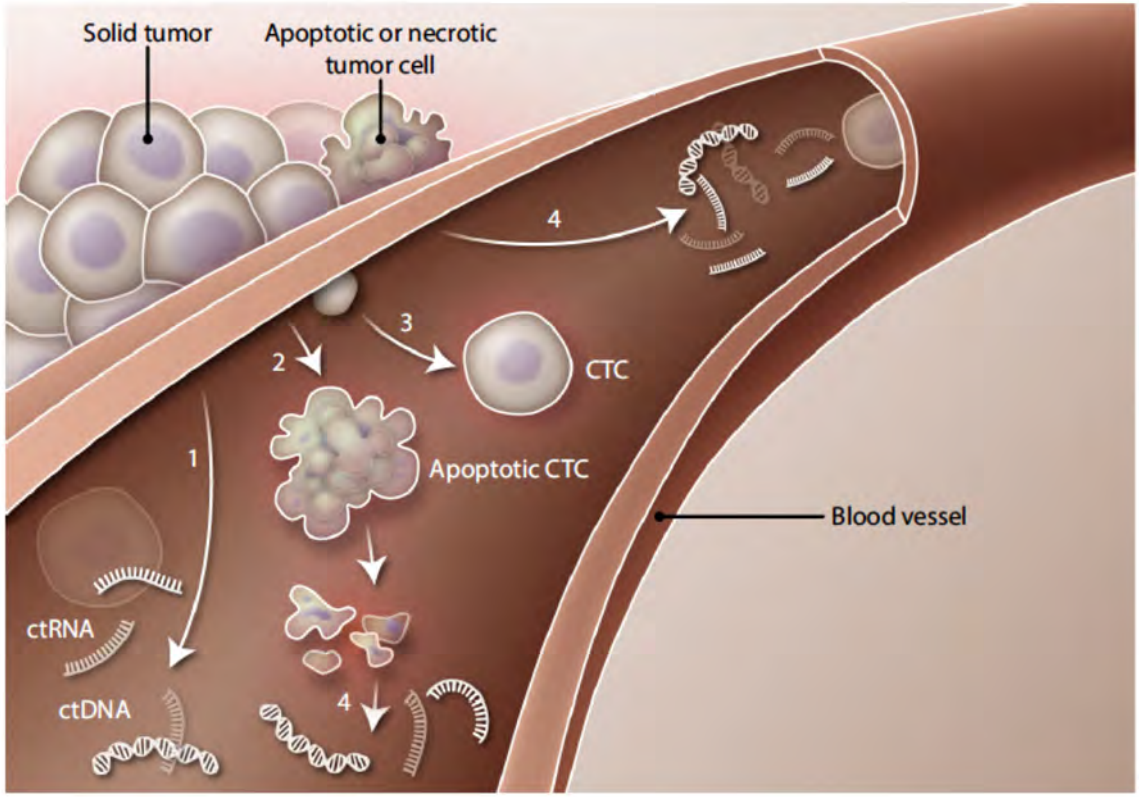
2) Defective HR assessment via sequencing for: BRCA1, BRCA2 at a minimum, with ability to detect copy number alterations





Report from ISUP Consultation Conference: Mol. Path Subgroup

What is next for CRPC Diagnostics



Liquid biopsy to overcome limits of multiple metastasis biopsies to capture heterogeneity and/or serial biopsies

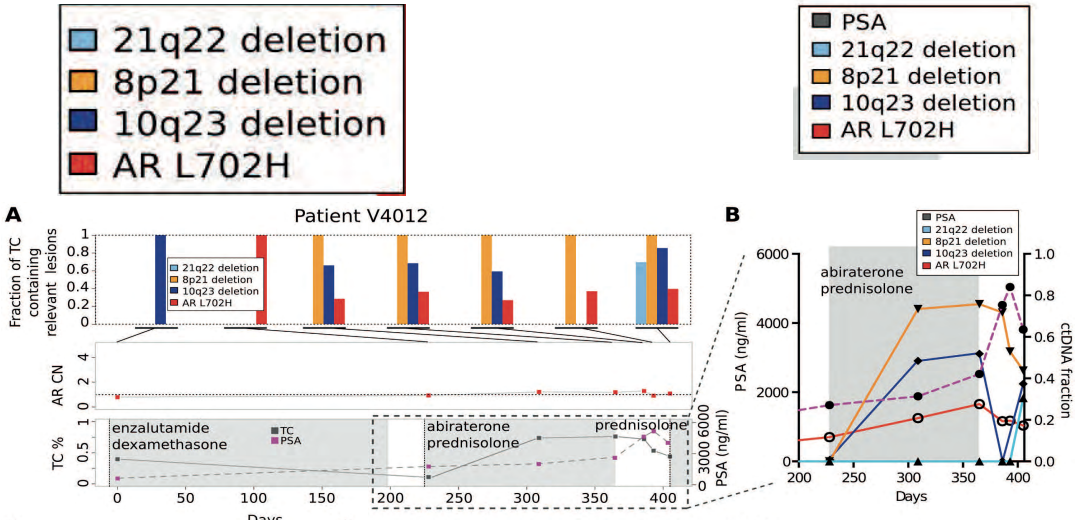
from Schweizer MT and Antonarakis ES, Sci Transl Med. 2015 Nov 4;7(312)

RESEARCH ARTICLE

CANCER

Tumor clone dynamics in lethal prostate cancer

Suzanne Carreira,[1]* Alessandro Romanel,[2]* Jane Goodall,[1]* Emily Grist,[1,3] Roberta Ferraldeschi,[1,3] Susana Miranda,[1] Davide Prandi,[2] David Lorente,[1,3] Jean-Sebastien Frenel,[1] Carmel Pezaro,[1,3] Aurelius Omlin,[1,3] Daniel Nava Rodrigues,[1] Penelope Flohr,[1] Nina Tunariu,[1,3] Johann S. de Bono,[1,3] Francesca Demichelis,[2,4,5†‡] Gerhardt Attard[1,3†‡]



Emergence of *AR-L702H* on treatment

Sci Transl Med **6**, *254ra125 (2014)*

REPORT

CANCER

Plasma *AR* and abiraterone-resistant prostate cancer

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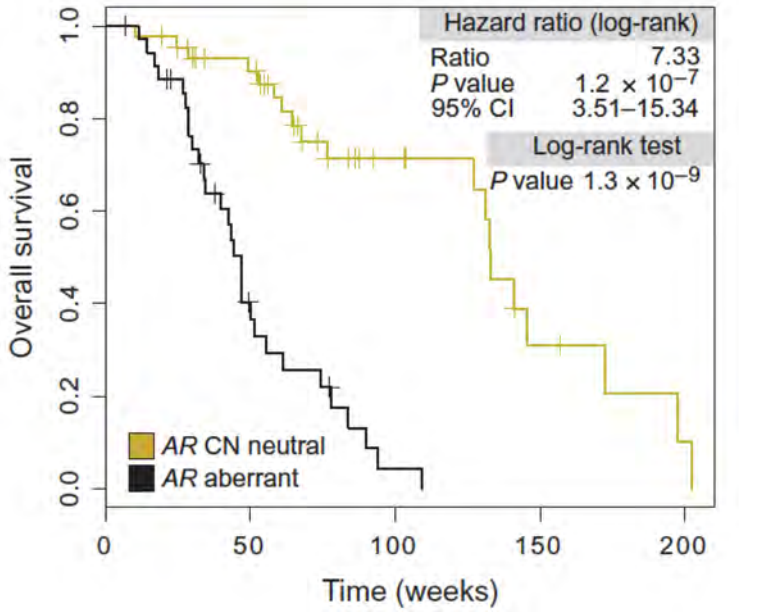


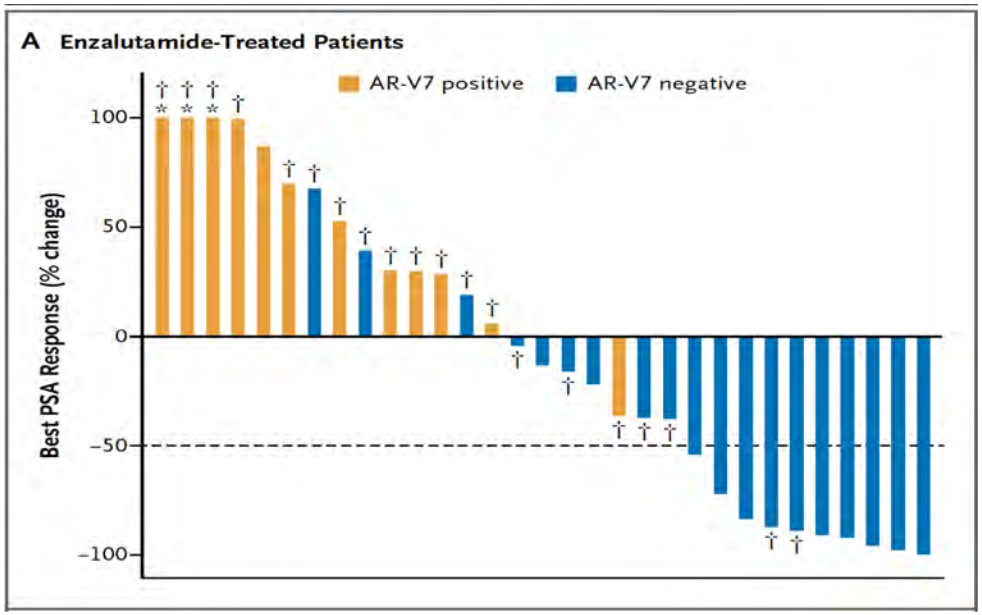
Plasma AR and abiraterone-resistant PCa

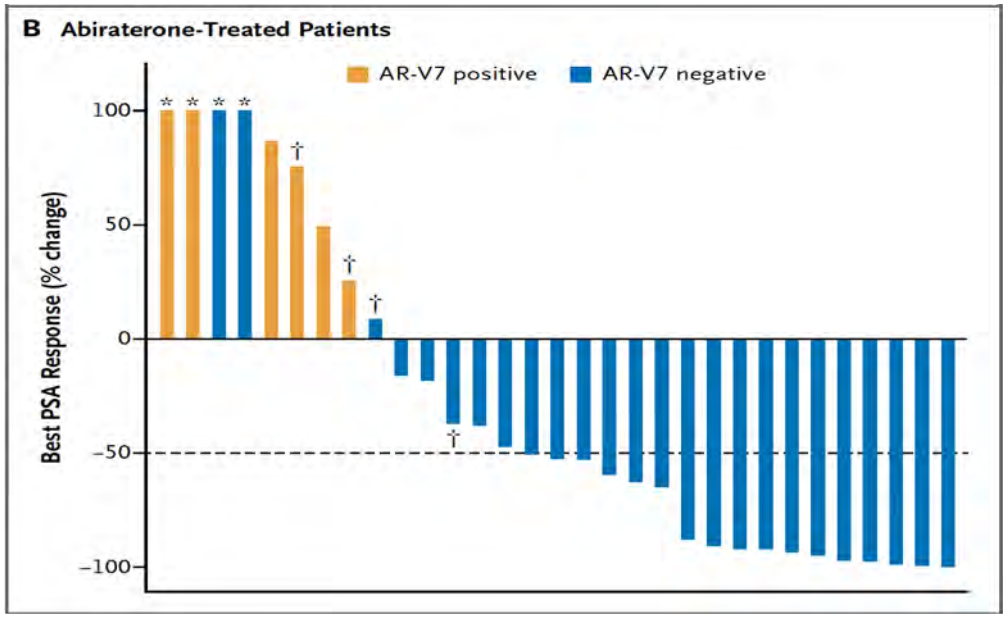
Sci Transl Med, 2015 Vol 7 Issue 312 312re10

The NEW ENGLAND JOURNAL of MEDICINE

ORIGINAL ARTICLE

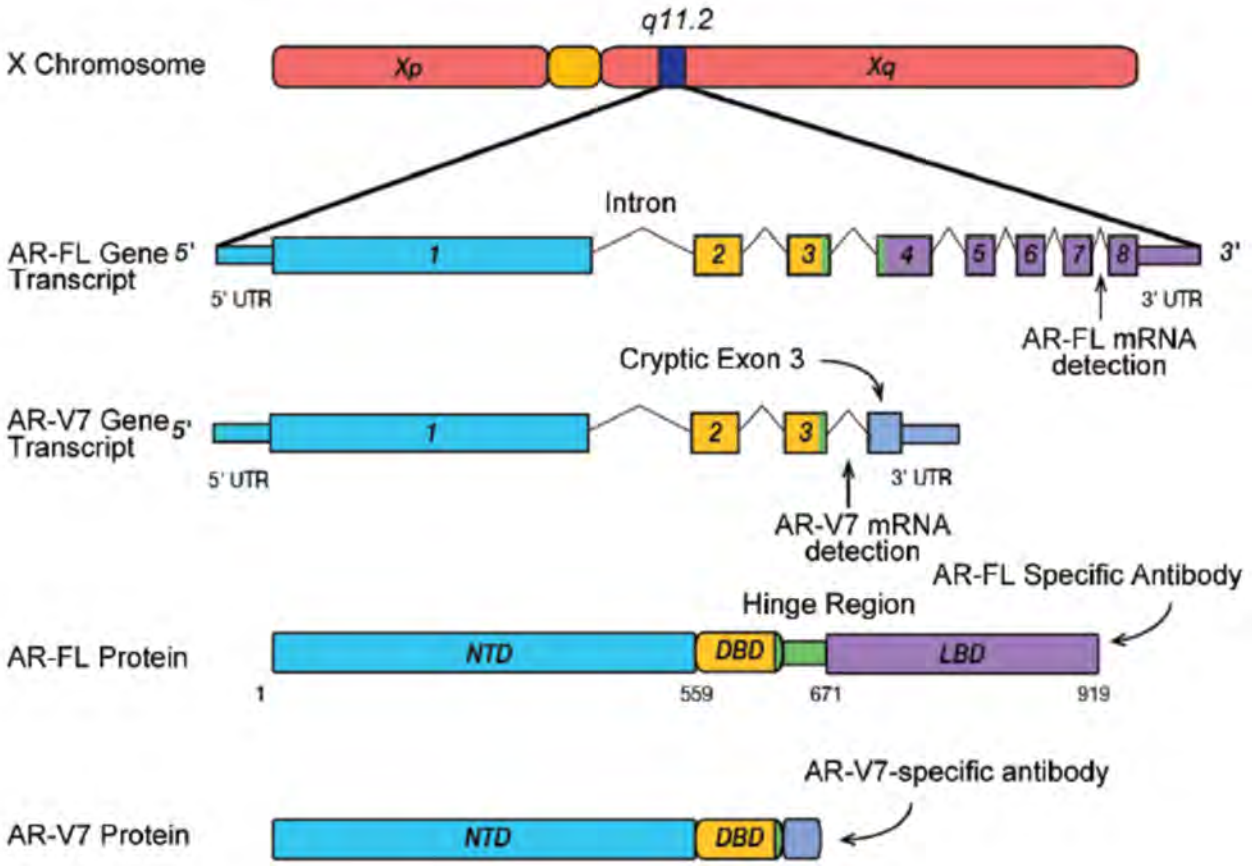
AR-V7 and Resistance to Enzalutamide and Abiraterone in Prostate Cancer





Antonarakis ES et al, NEJM 2014

The Androgen Receptor and associated ligand-indepedent variant, AR-V7



From the blood:
What is predictive? Prognostics? Reproducible?

cfDNA (tumor DNA)

AR-V7

AR gain

AR mutations

Other (neuroendocrine differentiation)

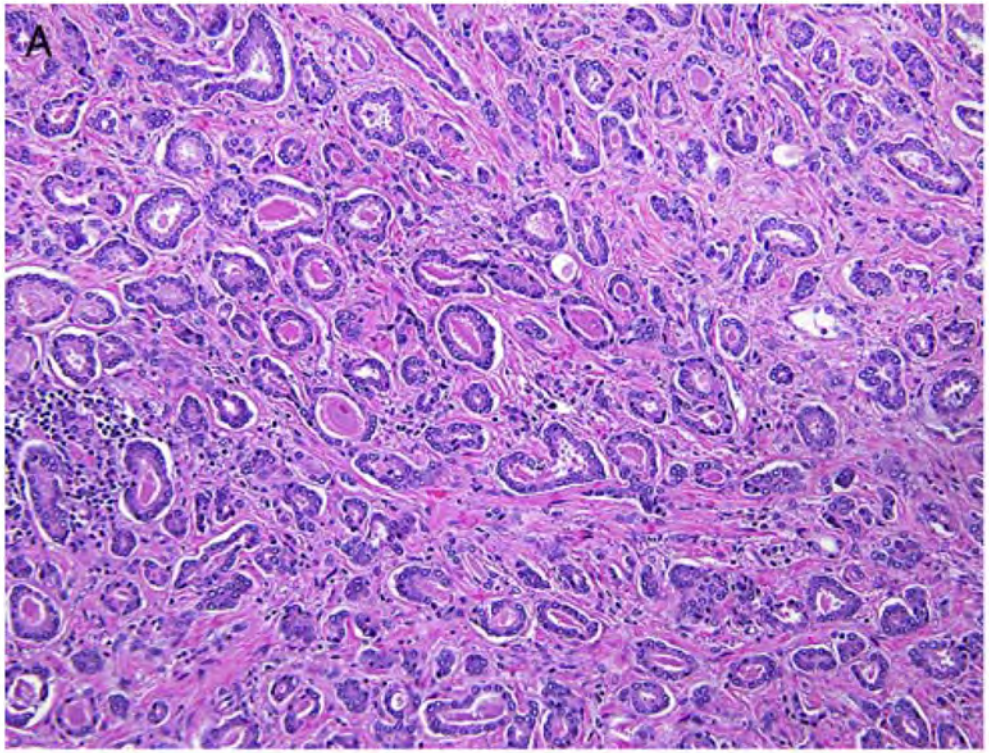
Most studies are not exploring these parameters together

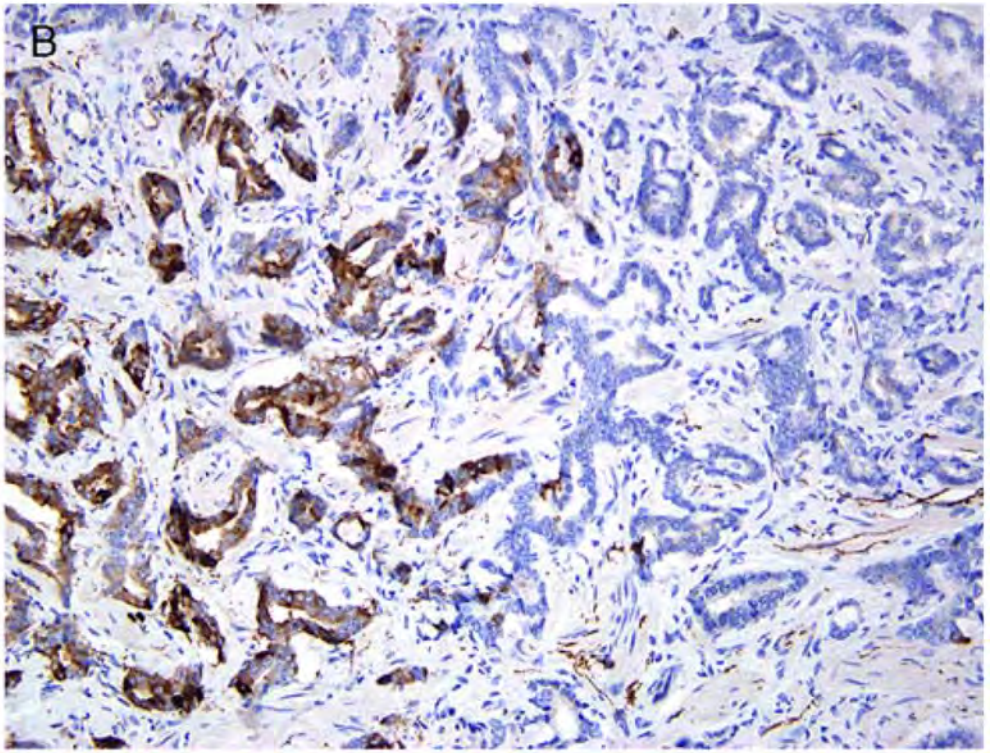
AR Testing:

At present, tissue based androgen receptor (*AR*) alteration assessment (amplifications, mutations, expression, splice variant expression) has no clear clinical utility

AR amplification and *ARv7* expression are prognostic in CRPC; emerging evidence suggests that ARv7 and AR amplification may be predictive, however, the evidence is not yet sufficient to justify systematic ARv7 or *AR* amplification testing.

Report from ISUP Consultation Conference: Mol. Path Subgroup





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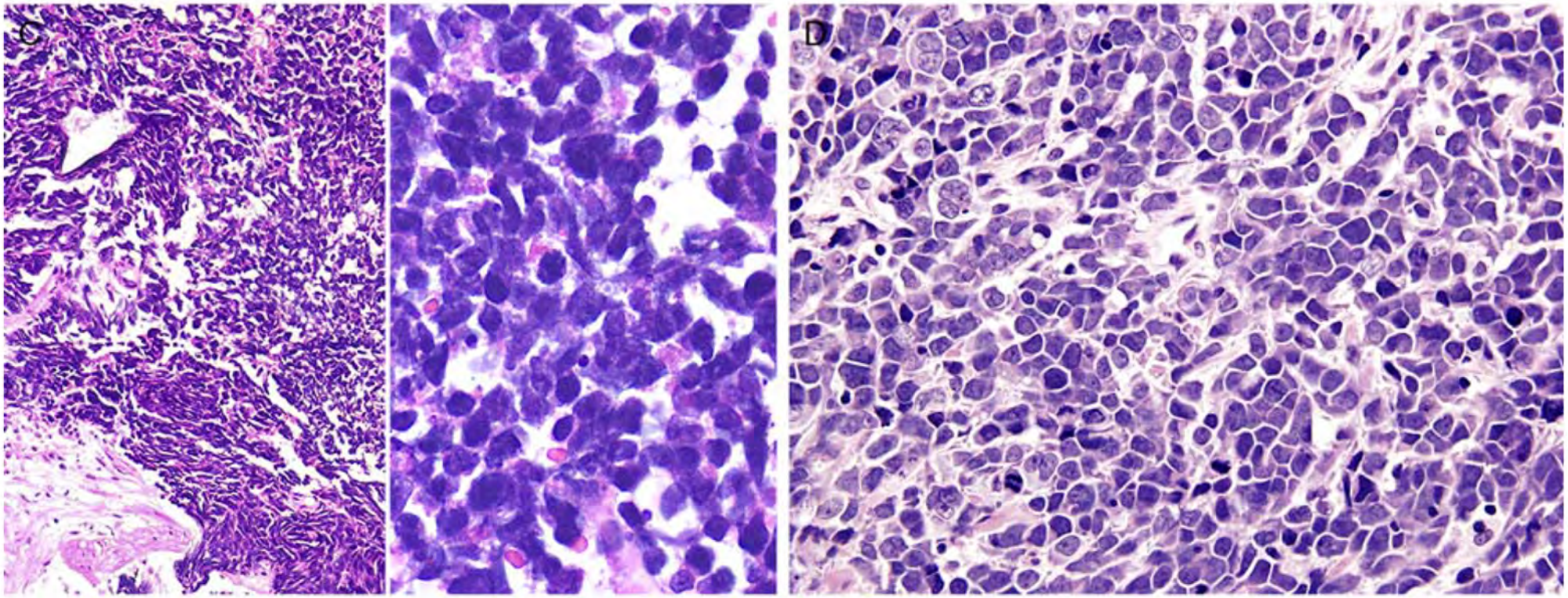

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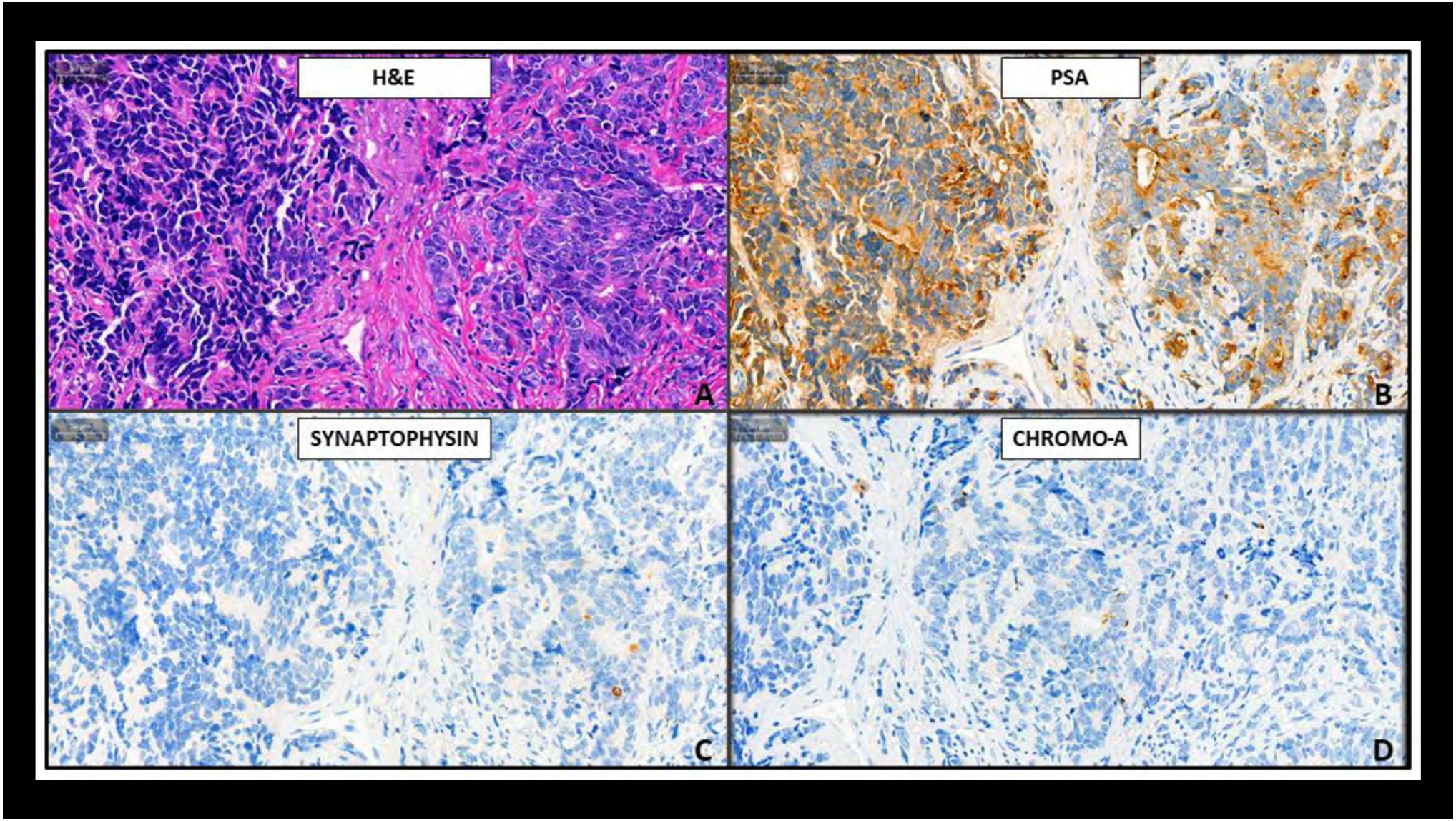
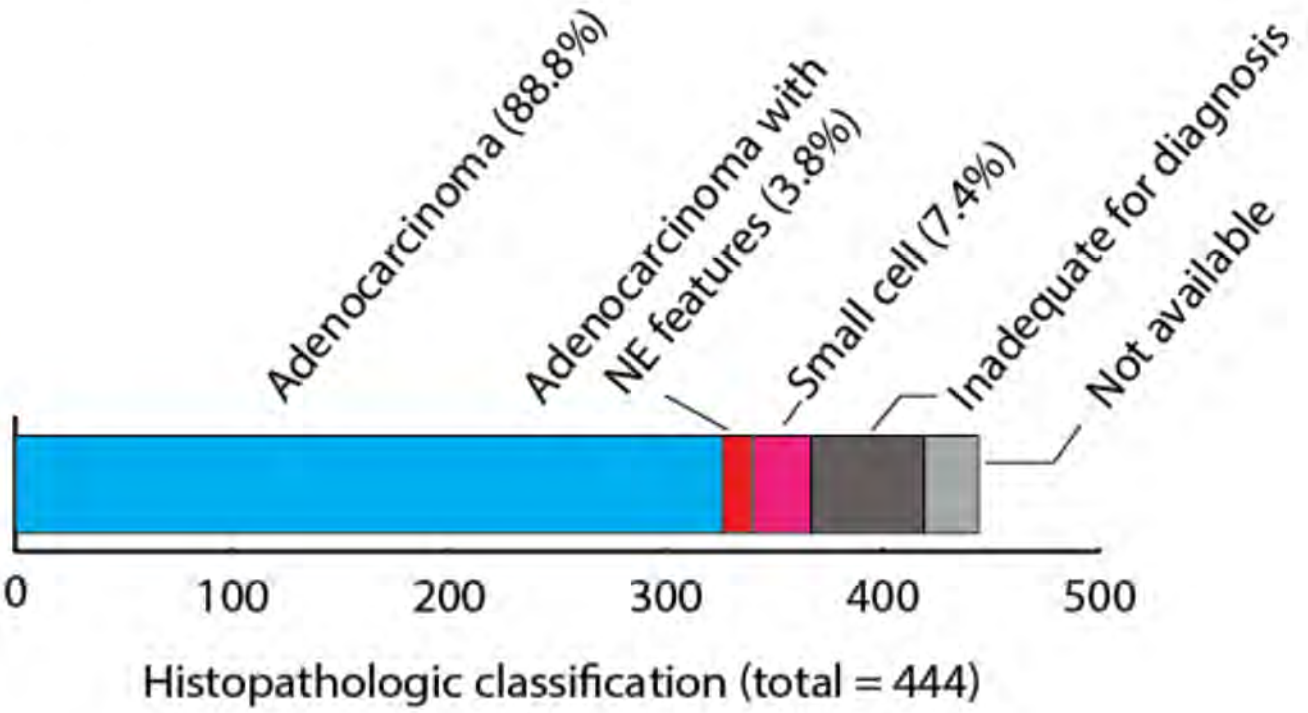
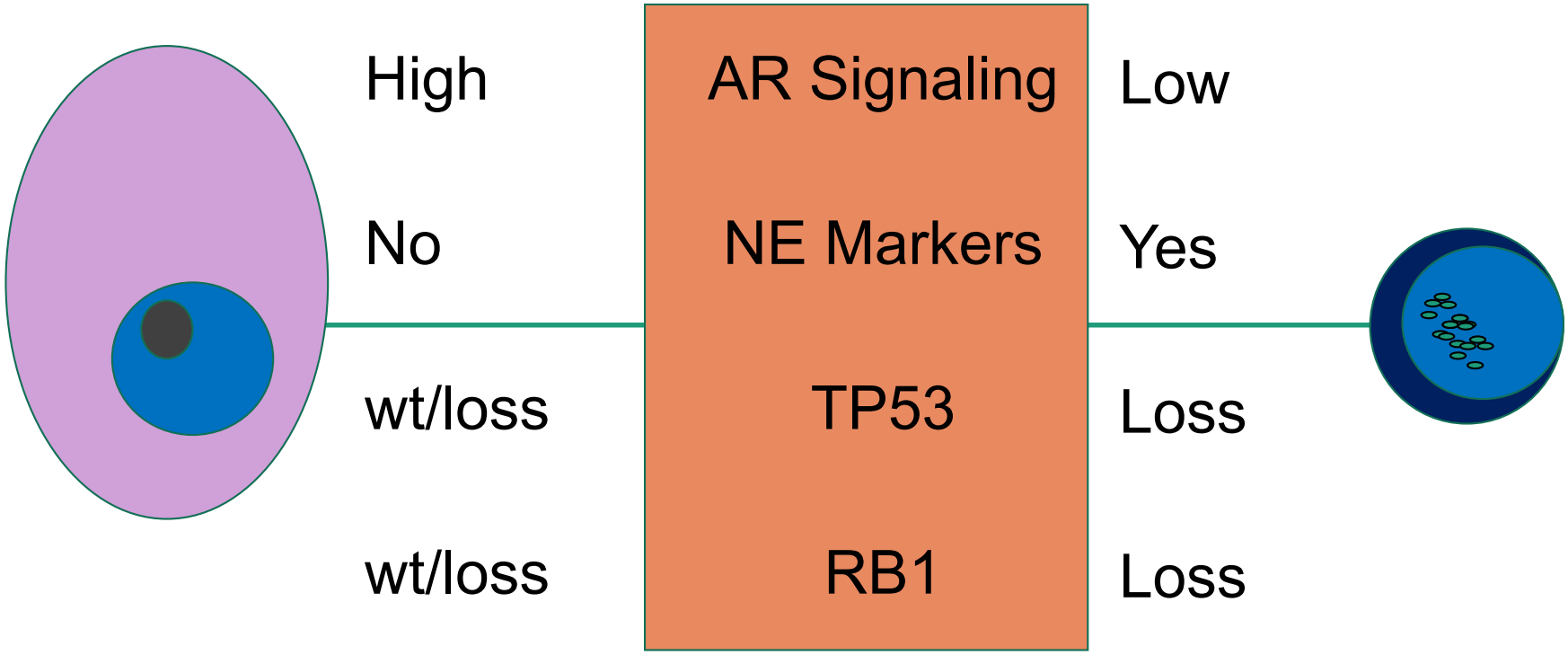

H&E
A
PSA
B
SYNAPTOPHYSIN
C
CHROMO-A
D

11% mCRPC have some neuroendocrine features



Abida et al, PNAS 2019

Characteristics that help define NEPC



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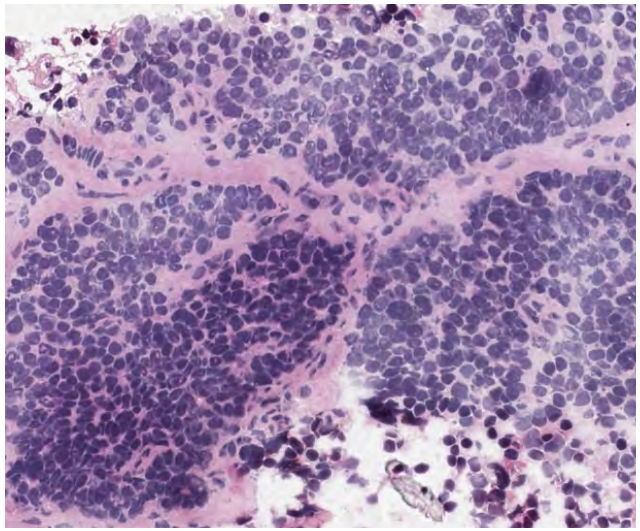
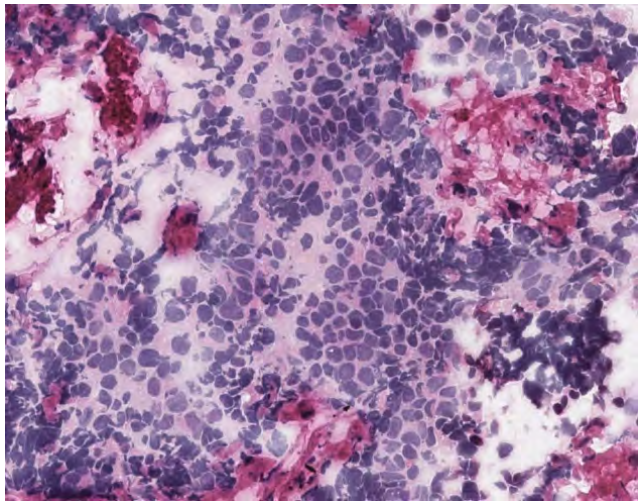
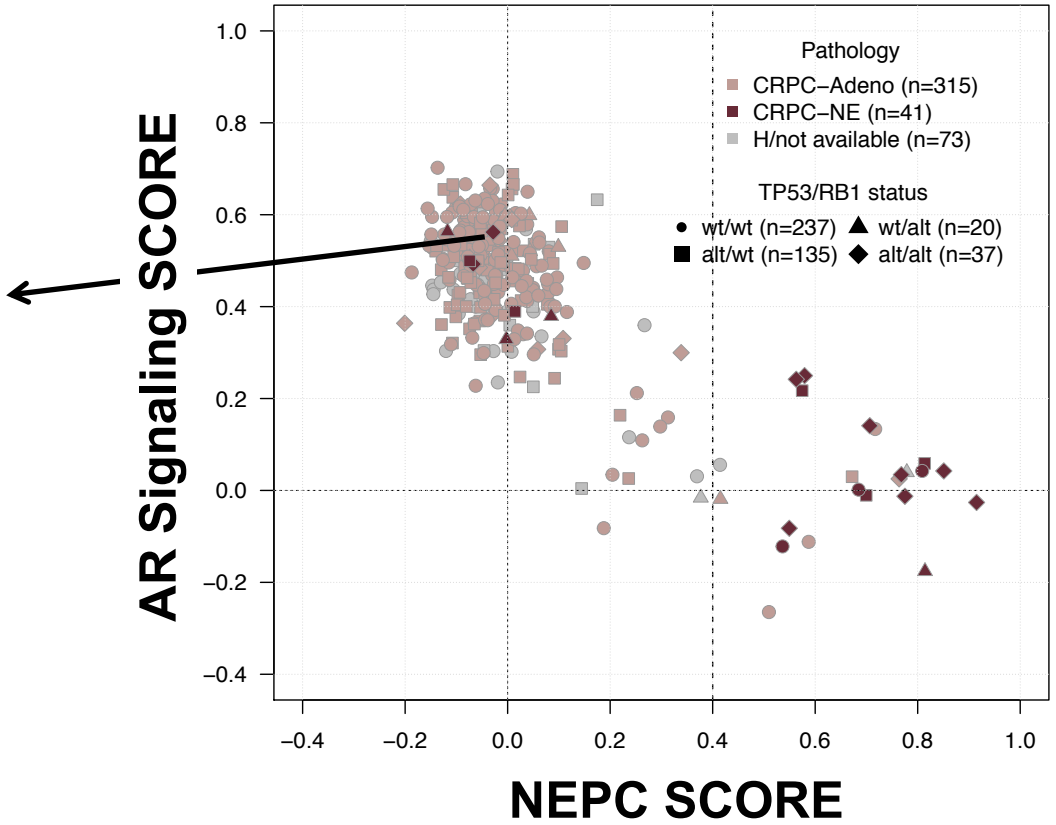




Small cell morphology, but low NEPC score, high AR, and mut TD53 and RB1



Abida et al, PNAS 2019

Neuroendocrine Prostate Cancer:

1) For clinically localized prostate cancer, unless there are clear morphologic neuroendocrine features, immunostaining for neuroendocrine expression (e.g., synaptophysin, chromogranin, or CD56) is **NOT** recommend.

2) Given its clinical implications, the term neuroendocrine differentiation is best reserved for high-grade cancers and not usual-type adenocarcinomas or well-differentiated neuroendocrine tumors.

3) Advanced metastatic CRPC may manifest a range of morphologic features of neuroendocrine differentiation and a combination of molecular evaluation and morphologic features may be required in future definitions of CRPC, guided by biomarker-driven clinical trials.

Report from ISUP Consultation Conference: Mol. Path Subgroup

Conclusions for Localized PCA

1) Only few pathologists are applying molecular markers in prostate cancer in routine practice due to insufficient evidence

2) Lack of standardization and the complexity of the diverse disease stages, treatment modalities and clinical endpoints.

3) Ki-67 and PTEN are emerging as potentially useful and widely available prognostic markers to support treatment decisions at an early stage.

4) mRNA based commercial genomic signatures can help stratifying the risk of progression in individual patients, although more studies are needed before widespread use of prognostic markers can be recommended.

Conclusion for mCRPC Promising Test*

a. **MSI testing**
b. **DNA repair status** ("BRCAness"-assay for BRCA1/2/ATM,PALB2) for mutation/loss or HR signature useful for for platinum therapy or PARPi

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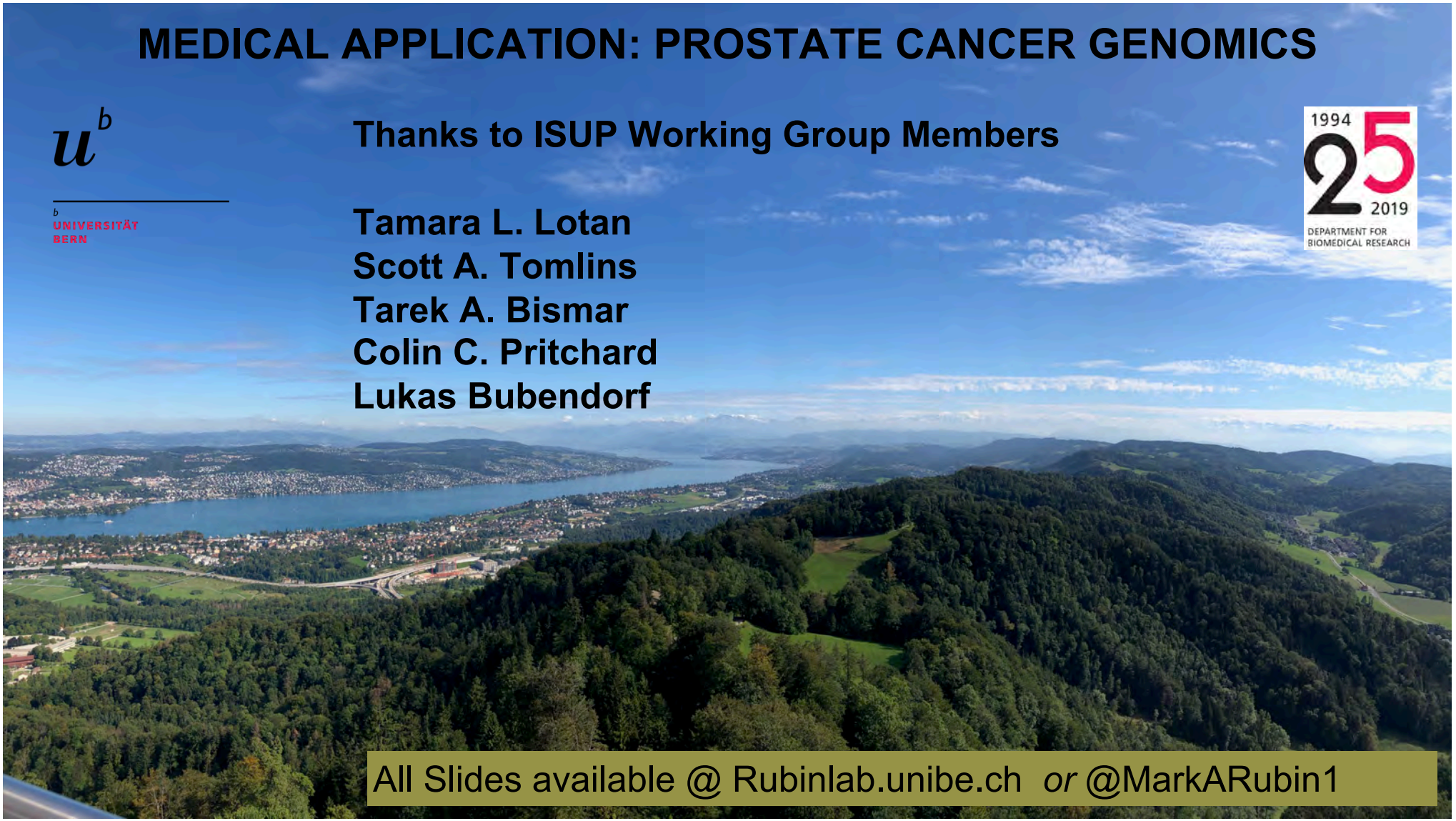
c. **Loss of AR** lack of response to AR therapy (AR-V7, mutations)
d. **cfDNA** amount associated with prognosis
e. **PTEN loss** - possibly response to AKT inhibitor (de Bono CCR 2018)
f. **CDK12 loss** - possibly response to checkpoint blockade
g. **Loss of TP53/RB1** - short duration of response to AR-therapy--possibly predictive response to platinum
h. **CTC heterogeneity** ("clusters") response to docetaxel vs AR therapy
i. **Pathology** phenotype for NEPC response to platinum
j. **Double negative (AR- and NE-)** response to FGFRi
k. **PSMA expression response** to PSMA-drug therapies
l. **DLL3** expression response to chemoconjugate

*Thanks Pete Nelson

MEDICAL APPLICATION: PROSTATE CANCER GENOMICS



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