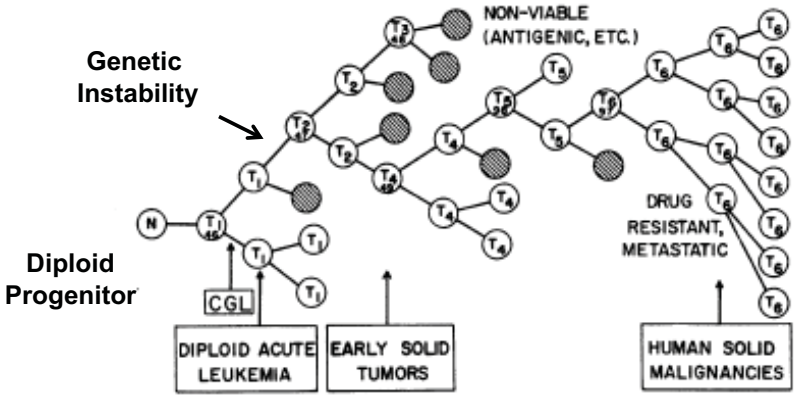
Application of Genomic Tools to Assist in Combination Therapy Development

Institute of Medicine
13 June 2011
Washington DC

TRANSLATIONAL GENOMICS RESEARCH INSTITUTE WWW.TGEN.ORG

Clonal Evolution of Human Neoplasia: Why are Cancers so Hard to Treat?

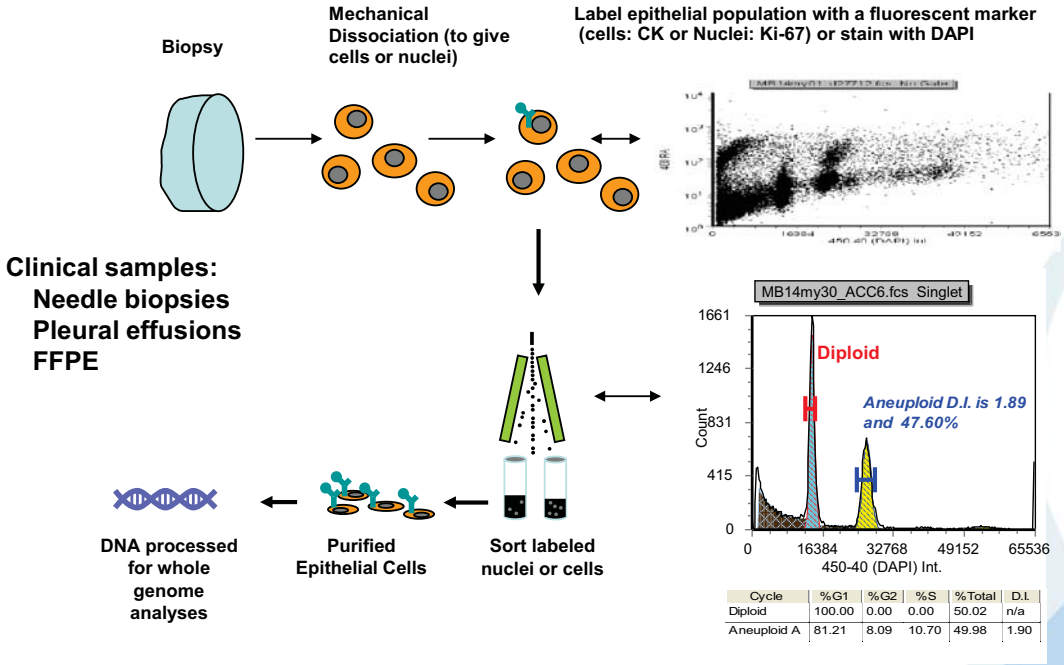


"The acquired genetic instability and associated selection process, most readily recognized cytogenetically, results in advanced human malignancies being highly individual karyotypically and biologically.

Hence, each patient's cancer may require individual specific therapy, and even this may be thwarted by emergence of a genetically variant subline resistant to the treatment."

Peter Nowell: Science 1976.

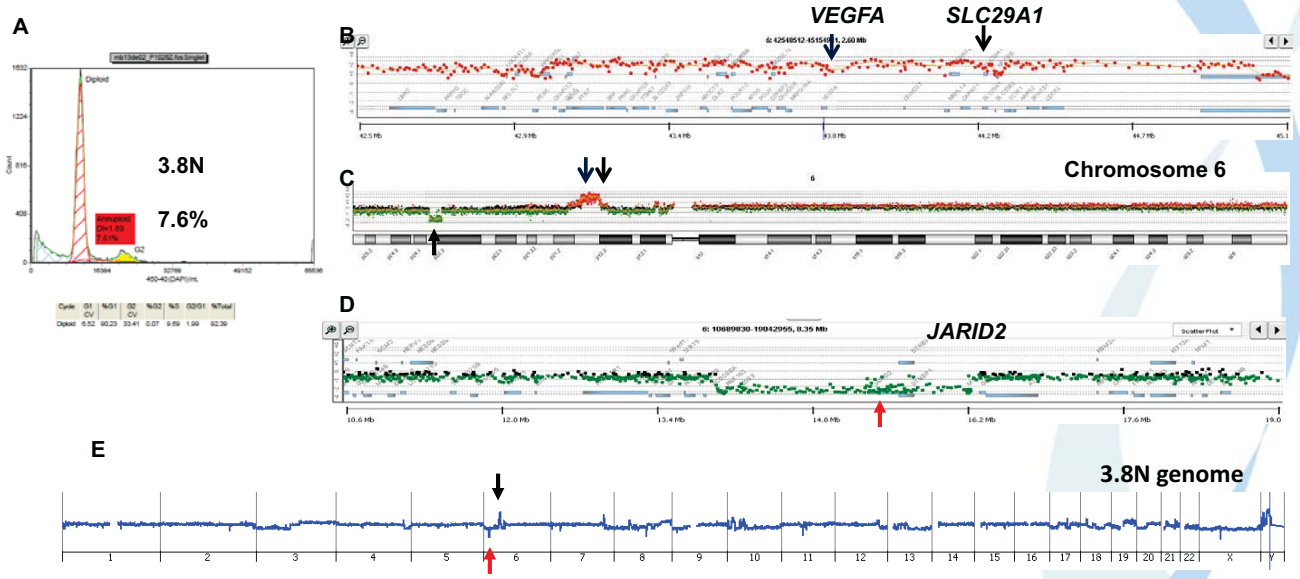
Clonal Analysis: Flow Sorting Neoplastic Cells from Biopsies



Predictions of Clonal Evolution

- Each genome has unique sets of selected aberrations and mutations.
- Multiple populations can be present in a clinical biopsy.
- Evolution of populations: Progression and metastases.
- Assymetric distribution of aberrations and mutations: Resistance to therapies.

Clonal Profiling of Clinical PDA Sample



Clinical Hypothesis: ***VEGFA*** **-> anti angiogenic therapy**
SLC29A1 **-> responsive to gemcitabine**
JARID2 **-> HDAC inhibitors?**

Patient-specific clonal aberrations "N's of 1"

P1026Z

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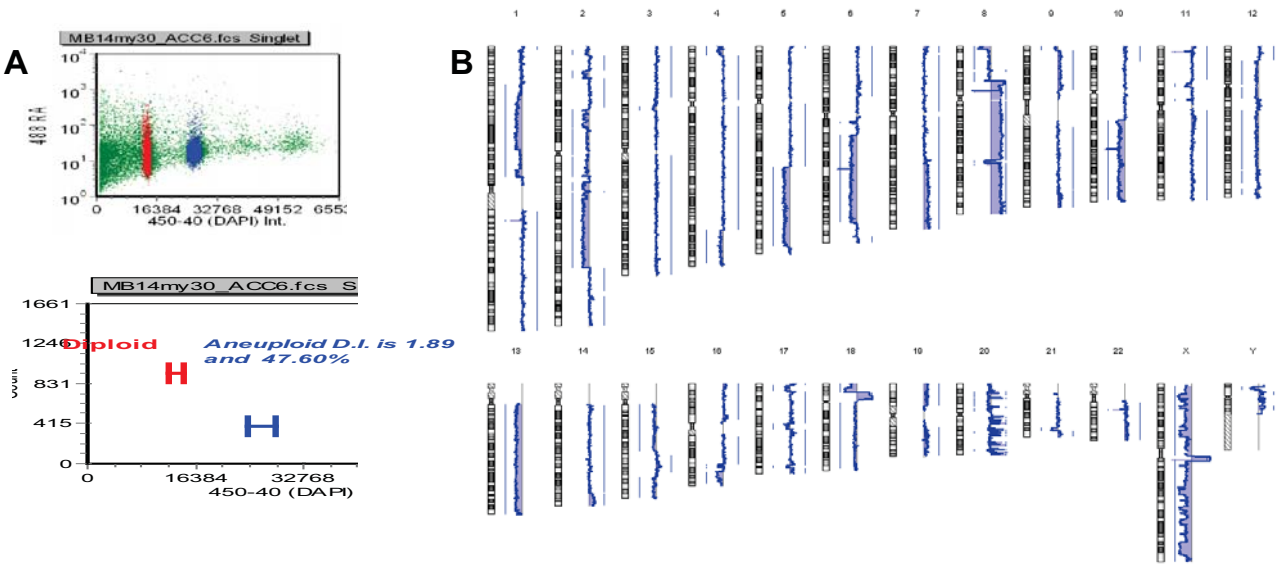
Interpreting Cancer Genomes

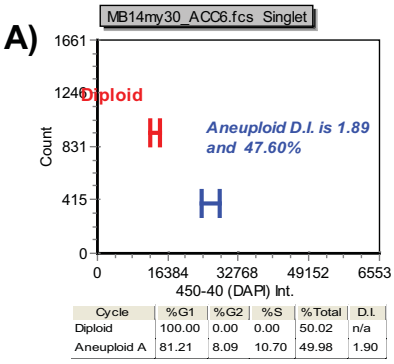


Dx: ACC

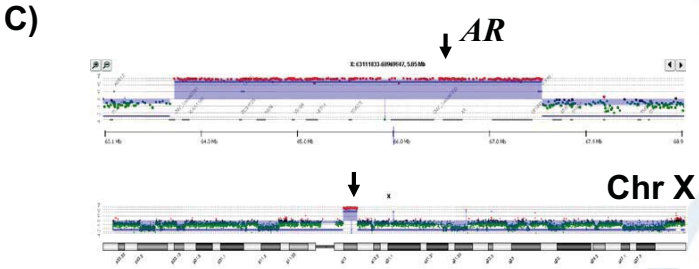
3.7N Genome

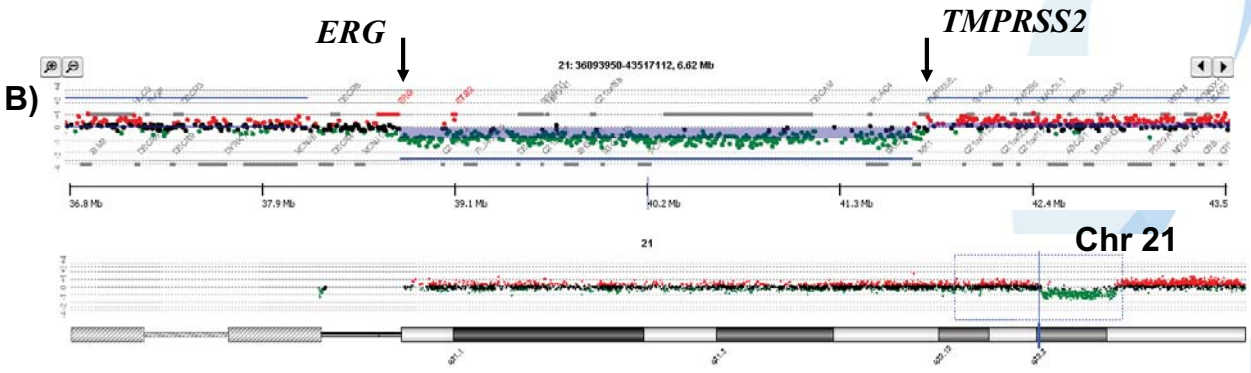
➢100 intrachromosomal copy number aberrations

Clonal Analysis of Cancer Genomes



Cycle	%G1	%G2	%S	%Total	D.I.
Diploid	100.00	0.00	0.00	50.02	n/a
Aneuploid A	81.21	8.09	10.70	49.98	1.90





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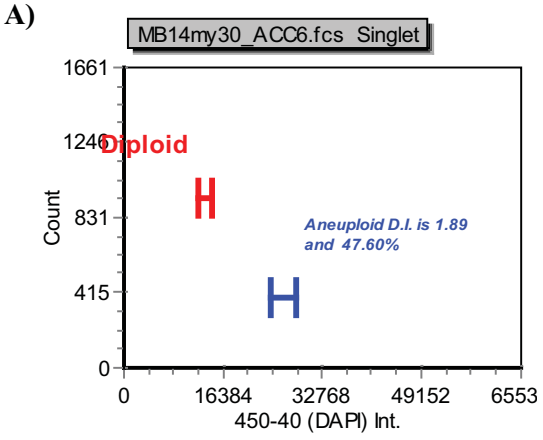
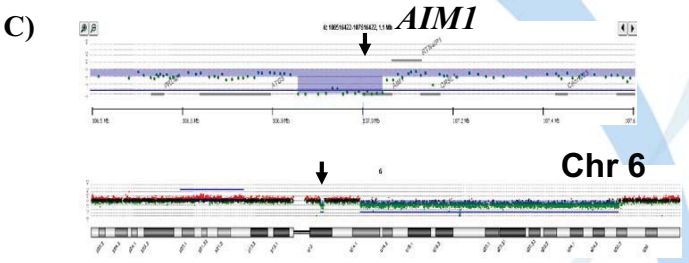
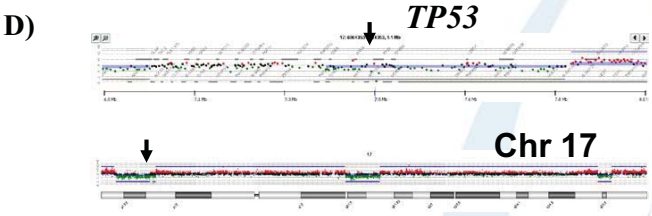
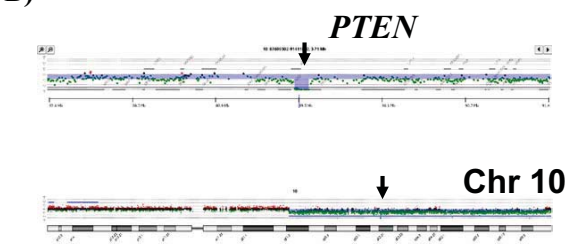
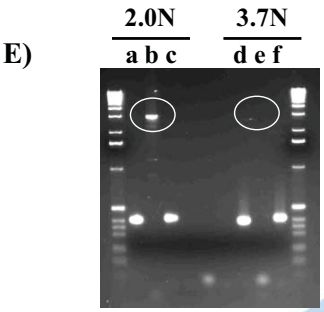
Clonal Analysis of Cancer Genomes





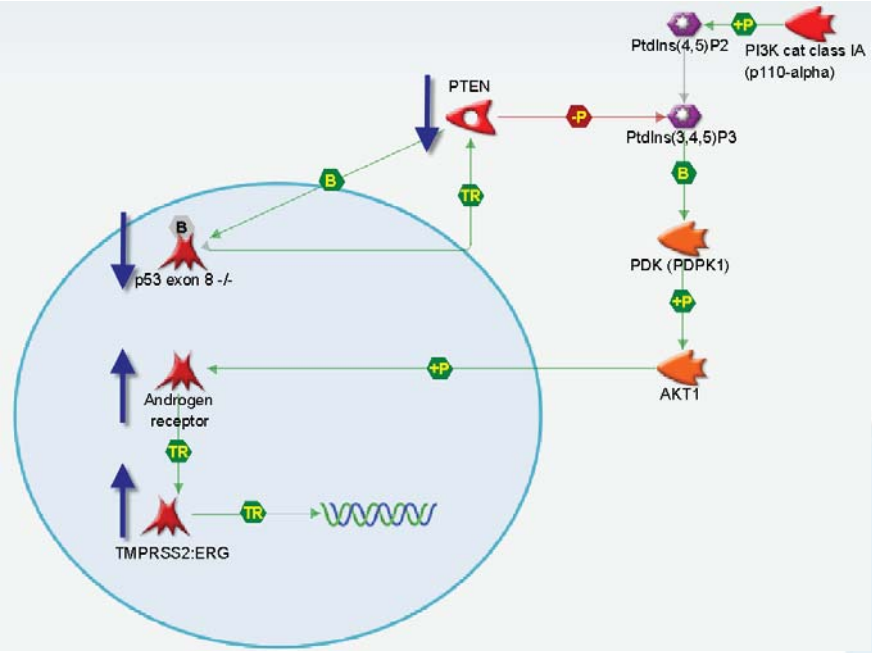






a, d: exon 1 *TP53*
b, e: exon 8 *TP53*
c, f: *FGFR2*

Knowledge Mining Cancer Genomes



New Dx: Prostate met to adrenal gland

Pathology review (2009): IHC of Tumor block PSA and prostatic acid phosphatase positive keratin 903 and p63 (basal cell markers) negative

MapEditor interface in the Metacore knowledge base (GeneGO).

Predictions of Clonal Evolution

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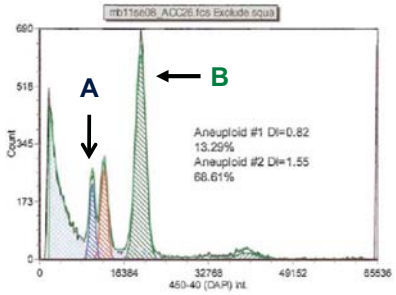
- **Multiple populations can be present in a clinical biopsy.**
- **Evolution of populations: Progression and metastases.**
- **Assymetric distribution of aberrations and mutations: Resistance to therapies.**

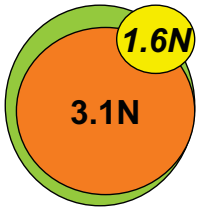
Clonal Analysis of Cancer Genomes



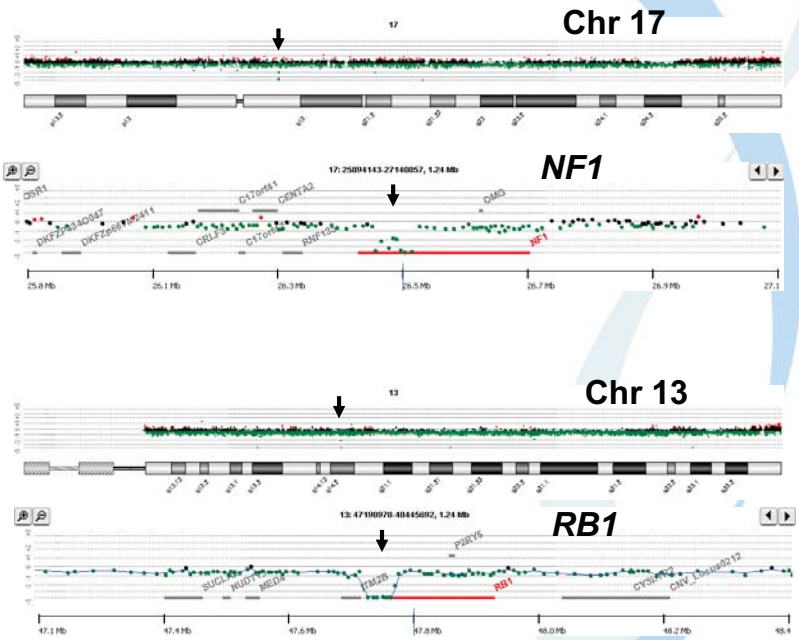
Cycle	G1 Mean	G1 CV	%G1	G2 Mean	G2 CV	%G2	%S	G2/G1	%Total	D.I.	B.A.D.
Diploid	12776.81	4.86	100.00	25026.43	4.87	0.00	0.00	1.96	18.10	n/a	15.96
Aneuploid A	10496.46	4.87	92.51	20734.90	4.87	0.00	7.49	1.98	13.29	0.82	15.53
Aneuploid B	19655.80	4.86	93.21	40421.96	4.86	6.50	0.29	2.04	68.61	1.55	22.24

Aneuploid clones

A (13% tumor cells)



B (68% tumor cells)



3.1N Clone (Aneuploid B)

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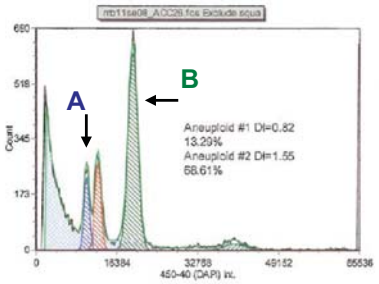
Clonal Analysis of ACC Genomes



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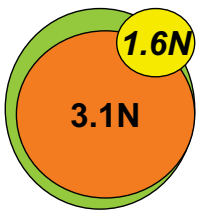
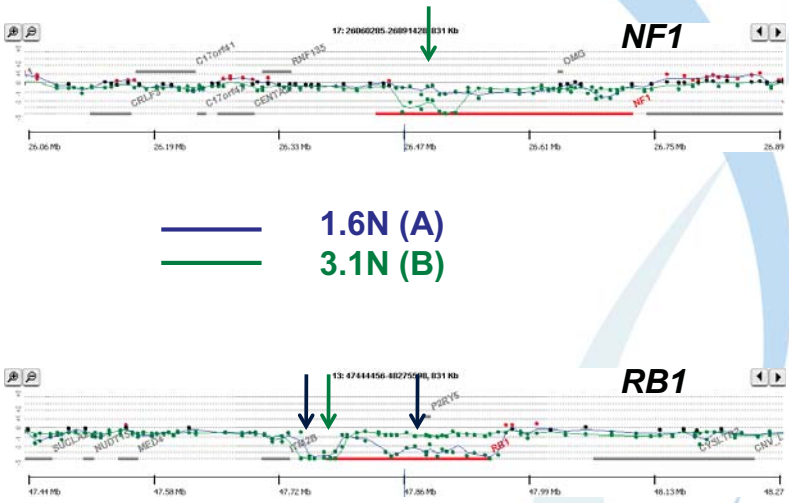
Aneuploid clones

A (13% tumor cells)



B (68% tumor cells)



1.6N (A)
3.1N (B)

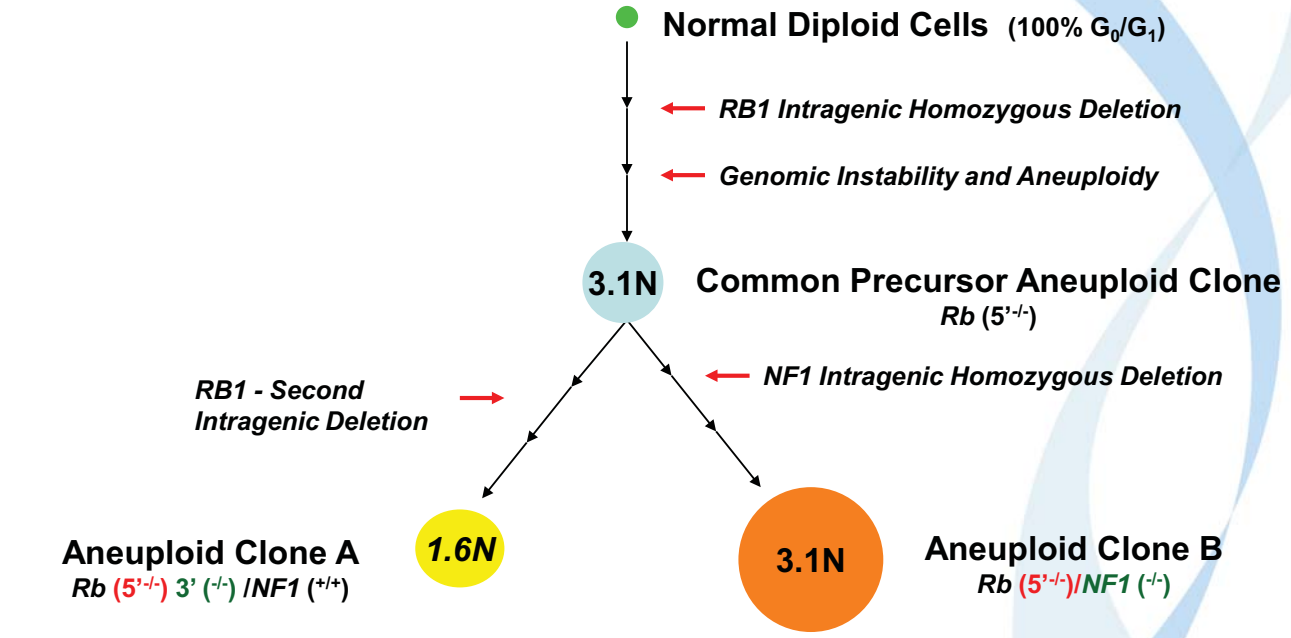
1.6N (A)

Rb **5' (-/-) 3' (-/-)**
NF1 **(+/+)**
S ↑

3.1N (B)

Rb **5' (-/-)**
NF1 **(-/-)**
G2 ↑

Modeling of Clonal Cancer Contexts



Aneuploid Clones:	*A (13% of Tumor Cells)*	*B (68.6 % of tumor cells)*
S Phase	*7.5%*	*0.3%*
G_2 Phase	*0.0%*	*6.5%*

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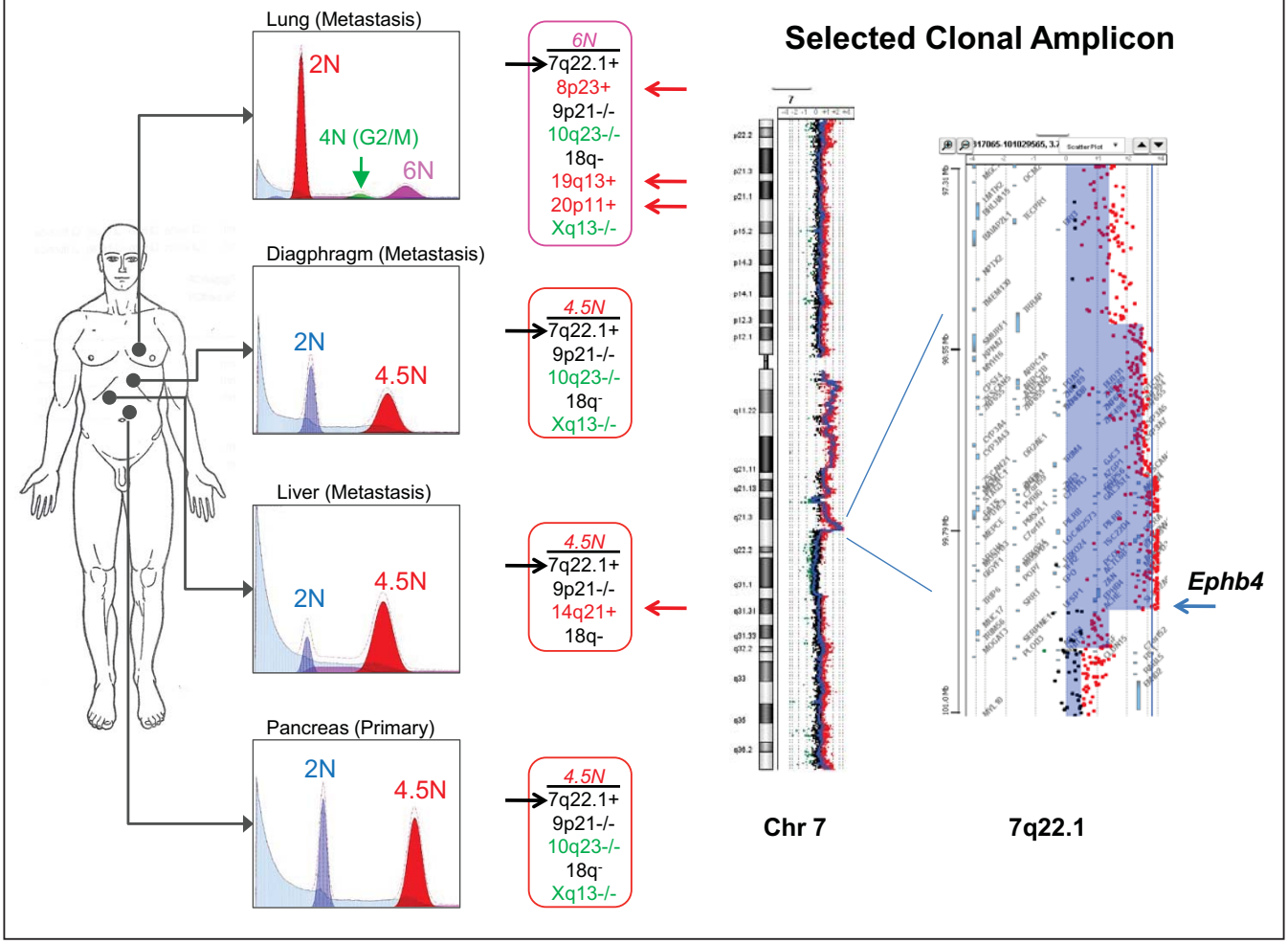

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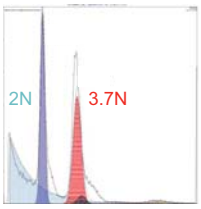
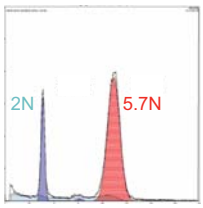
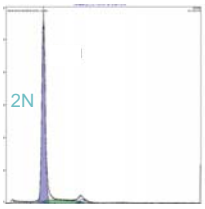
Evolution of Androgen-Independent Prostate Cancer

A

Year	Clinical history	Sorted populations	Genomic aberrations
2000:	PSA: 164ng/ml Gleason 7 (4+3) *Therapy:* First palliative TURP Bilateral orchiectomy		***2.0N*** → ***3.7N***
2007:	PSA: 19.6ng/ml Gleason 9 (5+4) *Therapy:* Second palliative TURP Bicalutamide 50mg daily.		***2.0N*** → ***5.7N***
2008:	PSA: 6.6ng/ml Gleason 9 (5+4) *Therapy:* Third palliative TURP		***2.0N***

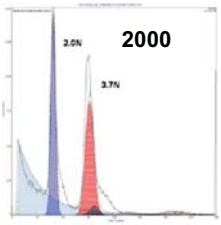
2.0N: 1p⁻/1q⁻, 12p⁻, 18q⁻

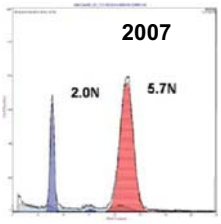
3.7N: 1p⁻/1q⁻, 12p⁻, 18q⁻

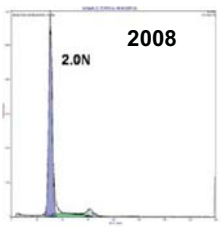
5.7N: 1p⁻/1q⁻, 5p⁺/5q⁺, 8q⁺, 12p⁻, 18q⁻, Xp⁺/Xq⁺, *AR*⁺⁺⁺

2.0N: 1p⁻/1q⁻, 5p⁺/5q⁺, 8q⁺, 12p⁻, 18q⁻, *AR*⁺, *GeneX*⁻/⁻

FISH Validation of *AR* Amplicons





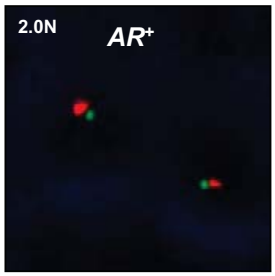


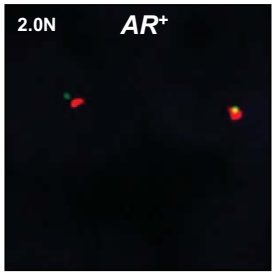
2.0N: 1p⁻/1q⁻, 12p⁻, 18q⁻

2.0N: 1p⁻/1q⁻, 5p⁺/5q⁺, 8q⁺, 12p⁻, 18q⁻, *AR*⁺

2.0N: 1p⁻/1q⁻, 5p⁺/5q⁺, 8q⁺, 12p⁻, 18q⁻, *AR*⁺, *GeneX*⁻/⁻

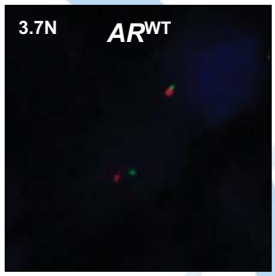
AR Xq12
Chr X centromere

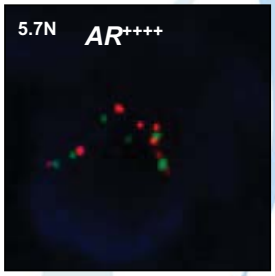




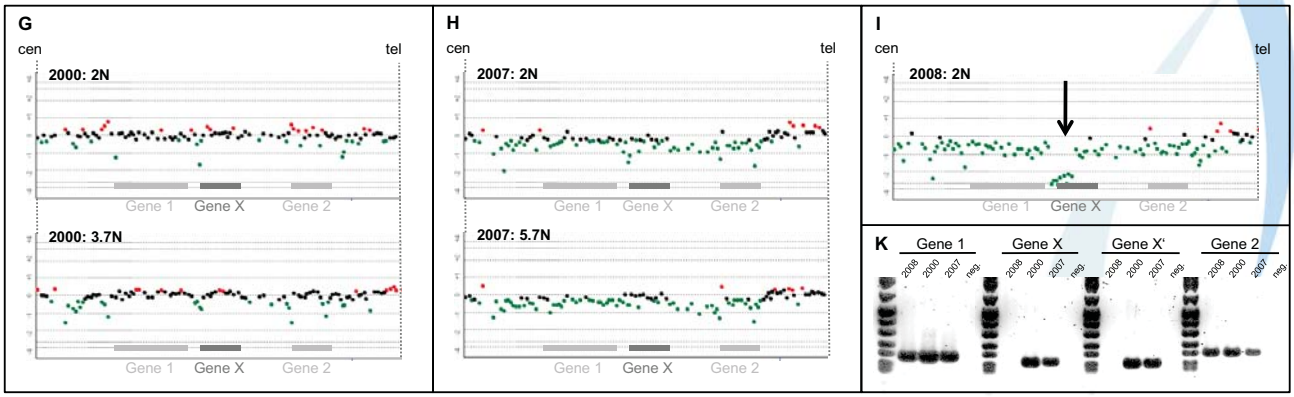
3.7N: 1p⁻/1q⁻, 12p⁻, 18q⁻

5.7N: 1p⁻/1q⁻, 5p⁺/5q⁺, 8q⁺, 12p⁻, 18q⁻, Xp⁺/Xq⁺, *AR*⁺⁺⁺

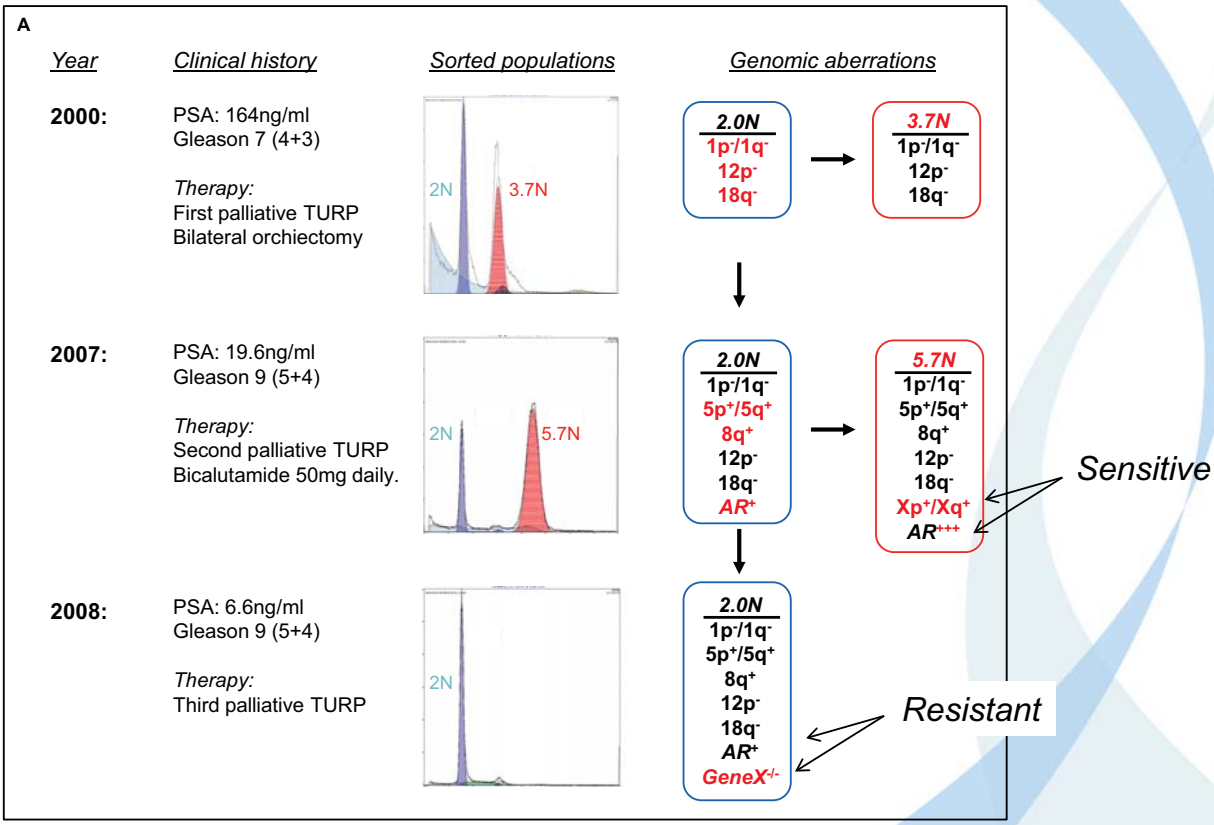




PCR Validation of Homozygous Deletion



Evolution of Androgen-Independent Prostate Cancer



Application of Genomic Tools to Assist in Combination Therapy Development

- **Unbiased profiling of patient biopsies.**
  - **Any % tumor cell content.**
  - **Any type of clinical specimen (resections, needle biopsies, FFPE, etc).**
  - **Maximize samples from clinical trials.**
- **Identify therapeutic vulnerabilities in each patient.**
  - **Highly objective quantitative measures.**
- **Targeted therapies:**
  - **Single targets are present in context of cancer genome.**
  - **Need to identify concurrent aberrations/mutations in each tumor cell population**

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Thank You