

Chromosomal Microarrays

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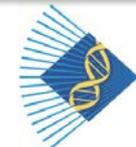
Associate Professor of Pathology and Immunology

Associate Professor of Pediatrics and Genetics

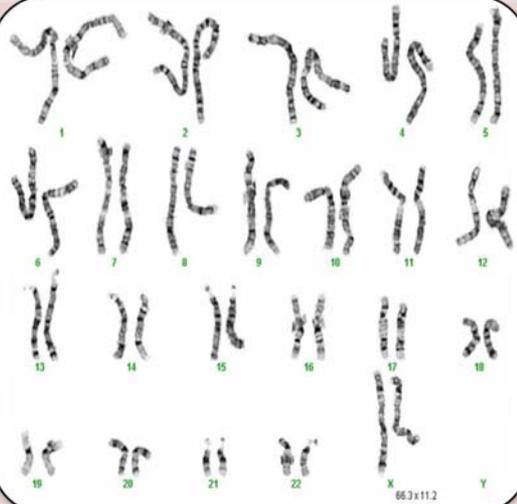


Washington University Physicians

Washington University School of Medicine in St. Louis



Cytogenomic Diagnostic Tools

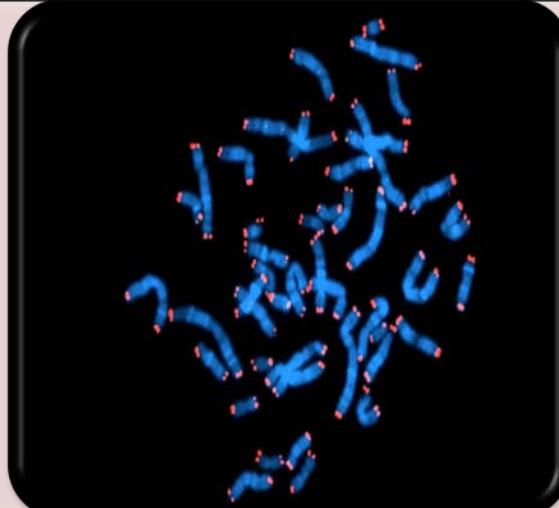


Classical Cytogenetics

~5 Mb

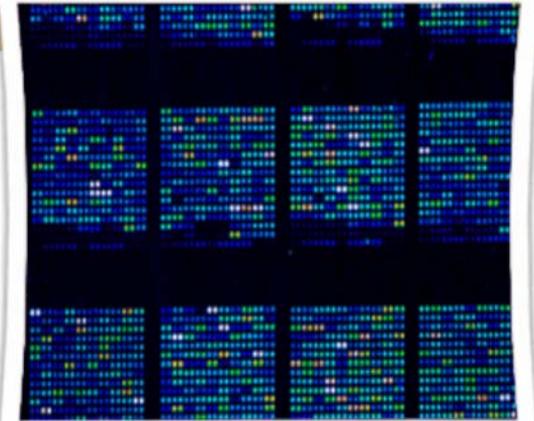
Global survey of
genome

Detects genomic
imbalances



FISH

Needs *a priori*
information



Cytogenomics

~ 50 Kb

Global survey of
genome

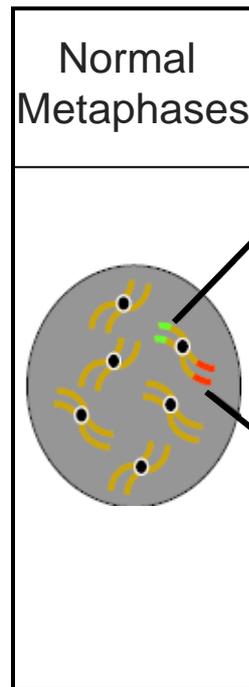
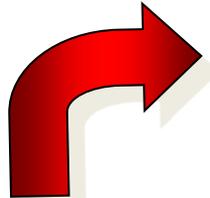
Detects genomic
imbalances

Comparative Genomic Hybridization (CGH) Chromosomal

Patient DNA



Control DNA



Gain

Resolution
 $\leq 10\text{Mb}$

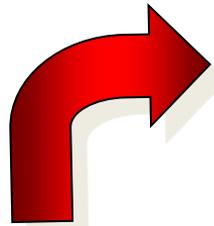
Loss

Comparative genomic hybridization (CGH)

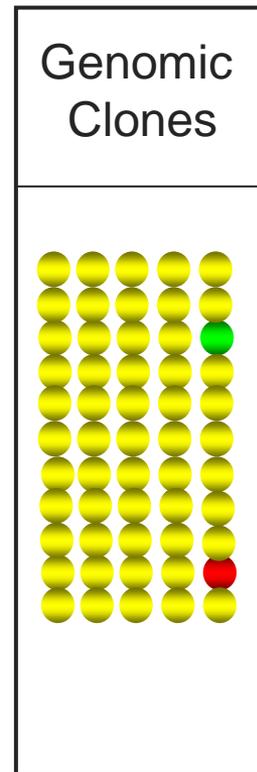
- Detects gains or losses of small chromosomal regions by DNA differences
- The patient's DNA is compared to a normal control in human metaphase chromosome spreads or arrays

Comparative Genomic Hybridization Arrays (aCGH)

Patient DNA



Control DNA



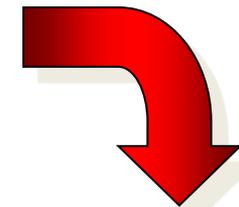
Gain

Resolution
=clone size

Loss

Single Nucleotide Polymorphism Arrays (SNP-A)

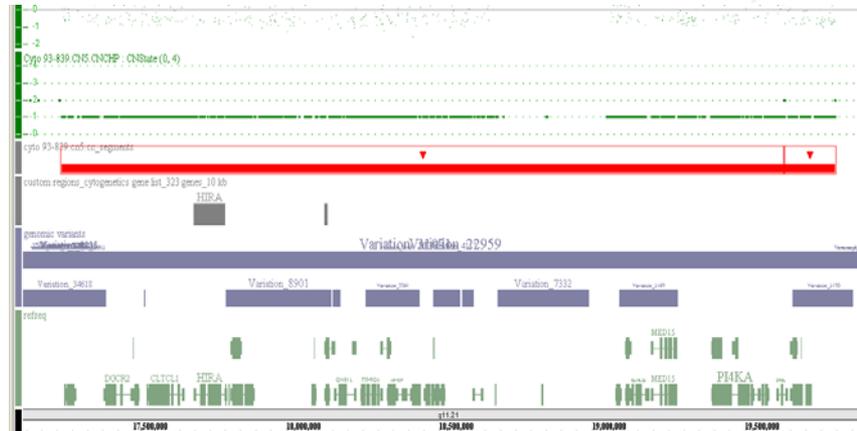
Patient DNA



In silico
Reference data
(hapmap)

 Gain

 Loss

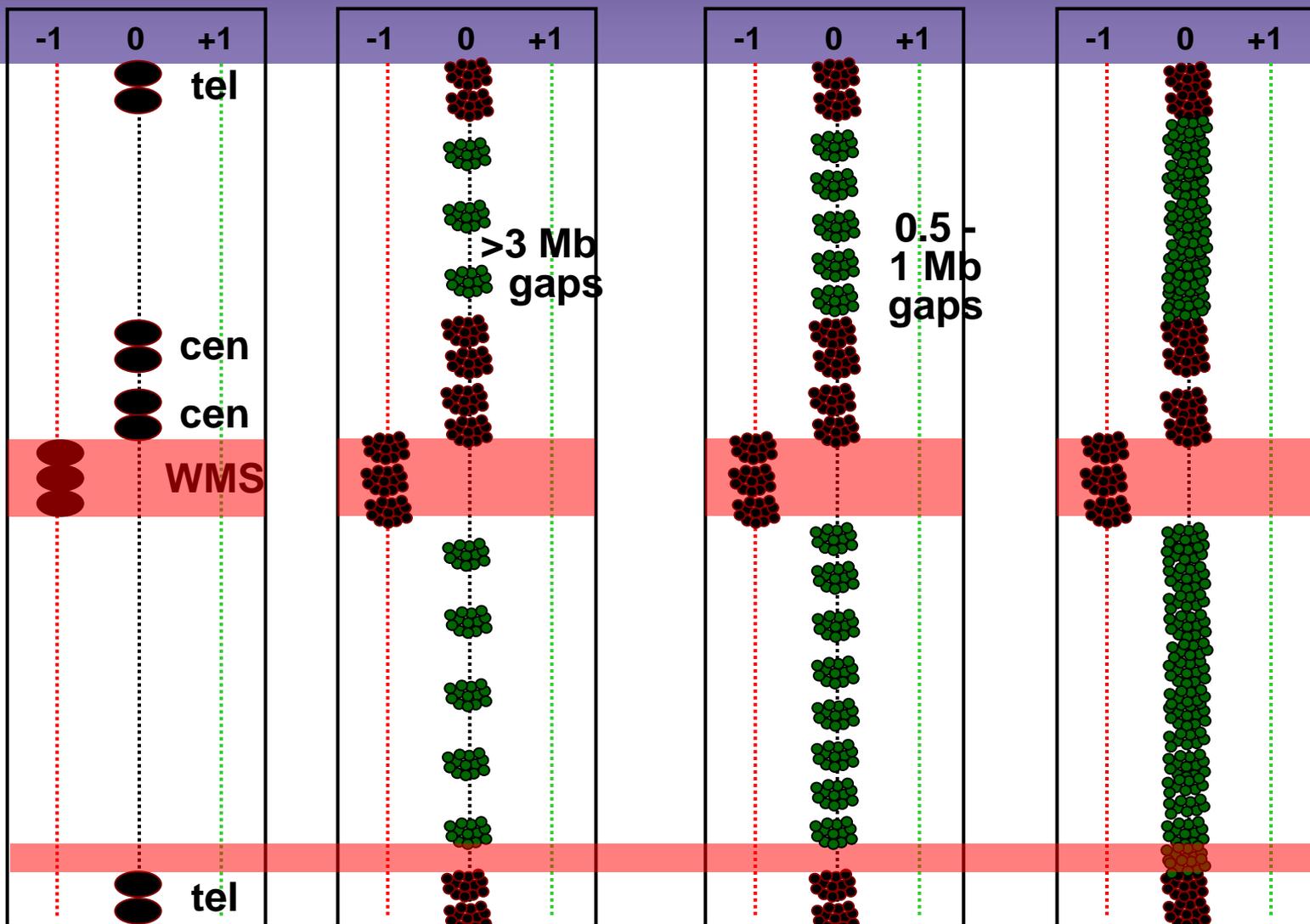
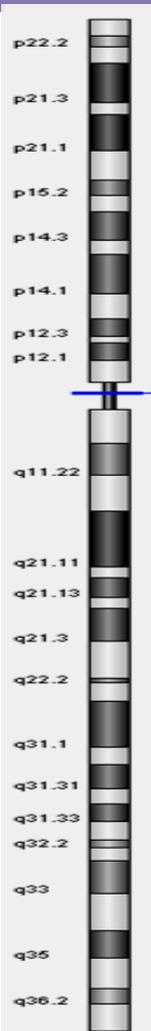


Various Platforms

BAC arrays	Oligo arrays	SNP+whole genome
BAC libraries make it easily customizable	Oligos are shorter 25-mer to 80-mer	Similar to Oligo array, but incorporate Bi-allelic probes
Tiling can make resolution better, but only to ~46Kb	Able to determine breakpoints more accurately	Able to determine breakpoints more accurately Can detect copy number changes and copy neutral changes (UPD, LOH)
BAC mis-localization Cross hybridization	“normal control” Interpretation can be difficult	With “correct reference data” noise can be reduced Interpretation can be difficult

Evolution of Array Designs

Chr 7



Targeted

Targeted + 850

Targeted +

Targeted +

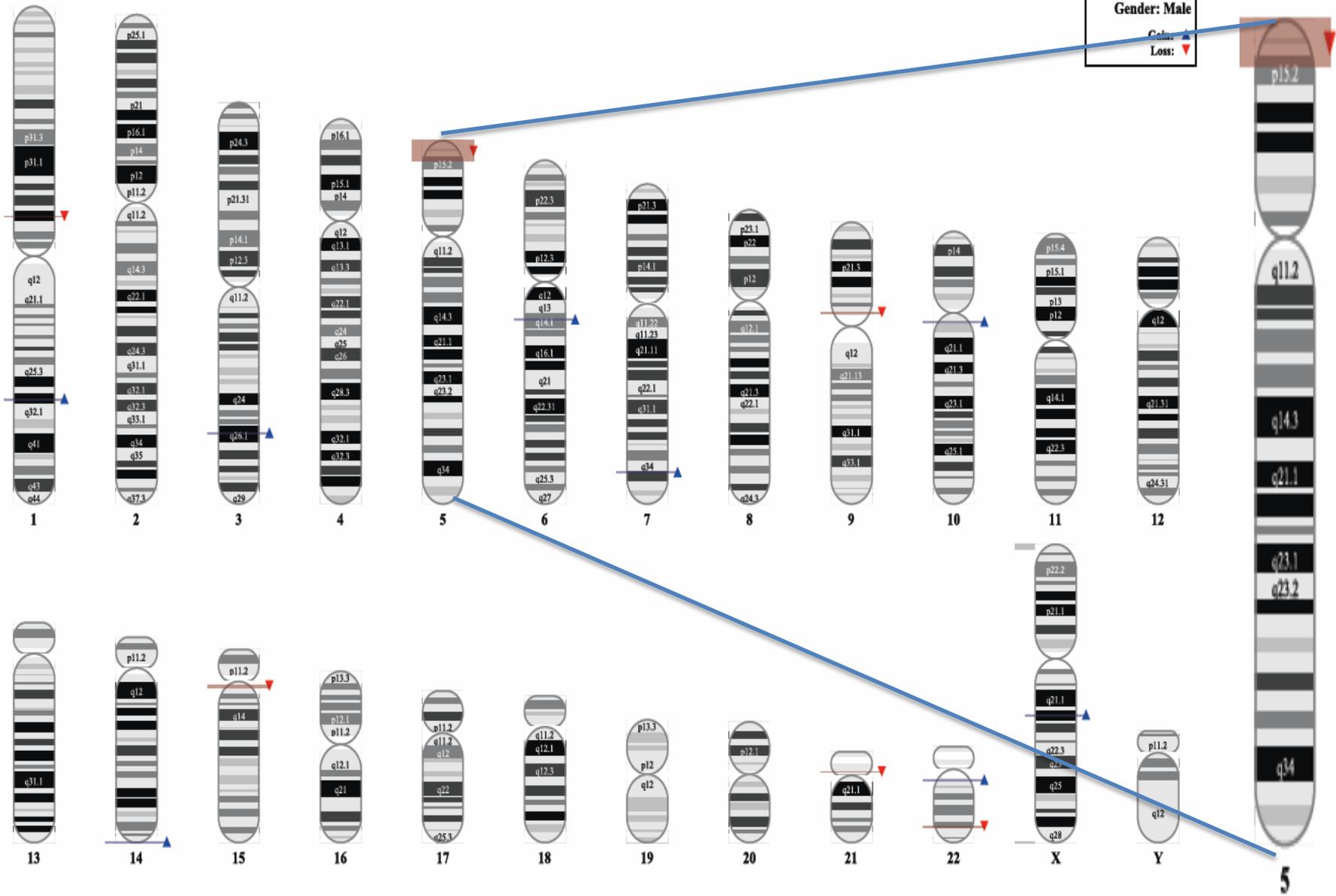
Slide courtesy of
Madhuri Hegde

BAC or oligo array

1 Mb or 500 kb

Whole Genome

G10-4309 BD.CN5
Gender: Male
Gain: ▲
Loss: ▼



Interpretation challenges

- **Copy Number Variations (CNV)**
 - DNA segment, longer than 1kb with a variable copy number compared to reference genome
 - Described both in disease and “normal” states
- Copy Number Alterations (CNA) preferred

Copy Number Variants (CNV)

- ~5-25% of human reference genome is copy number variable
- 1447 CNVs catalogued in 270 individuals (360 million bases, 12% of the genome)
Redon et al Nature 2006
- Each person has ~1500 common CNVs (inherited), average size of ~20 kb- 30 million bases of CNVs/person
- CNVs- interpretation challenges

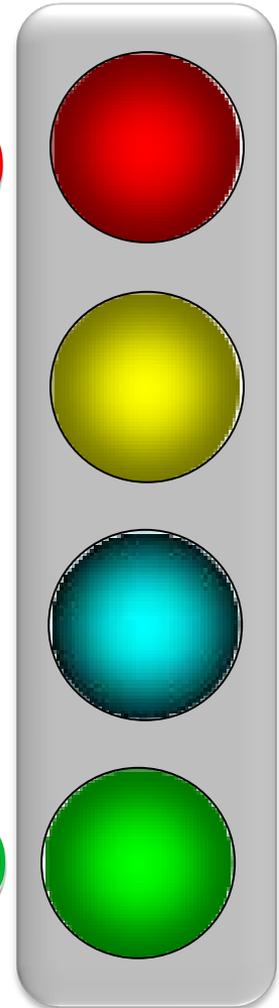
CNA Interpretation-four major criteria

Pathogenic (~ 10-15%)

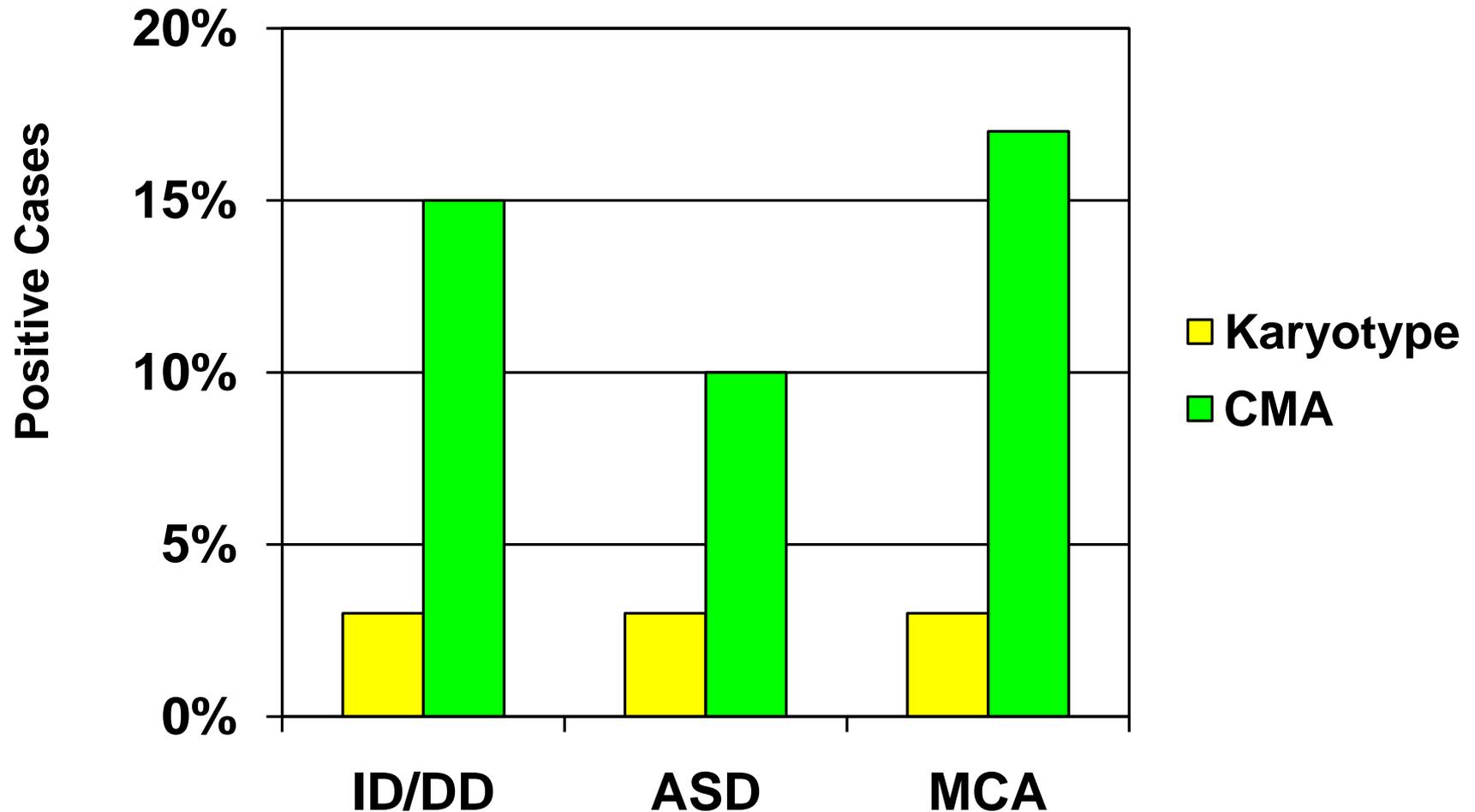
Variants of Uncertain Significance (VOUS)

Variant of Likely Pathologic Significance (VLPS)

Benign (~ 80%)



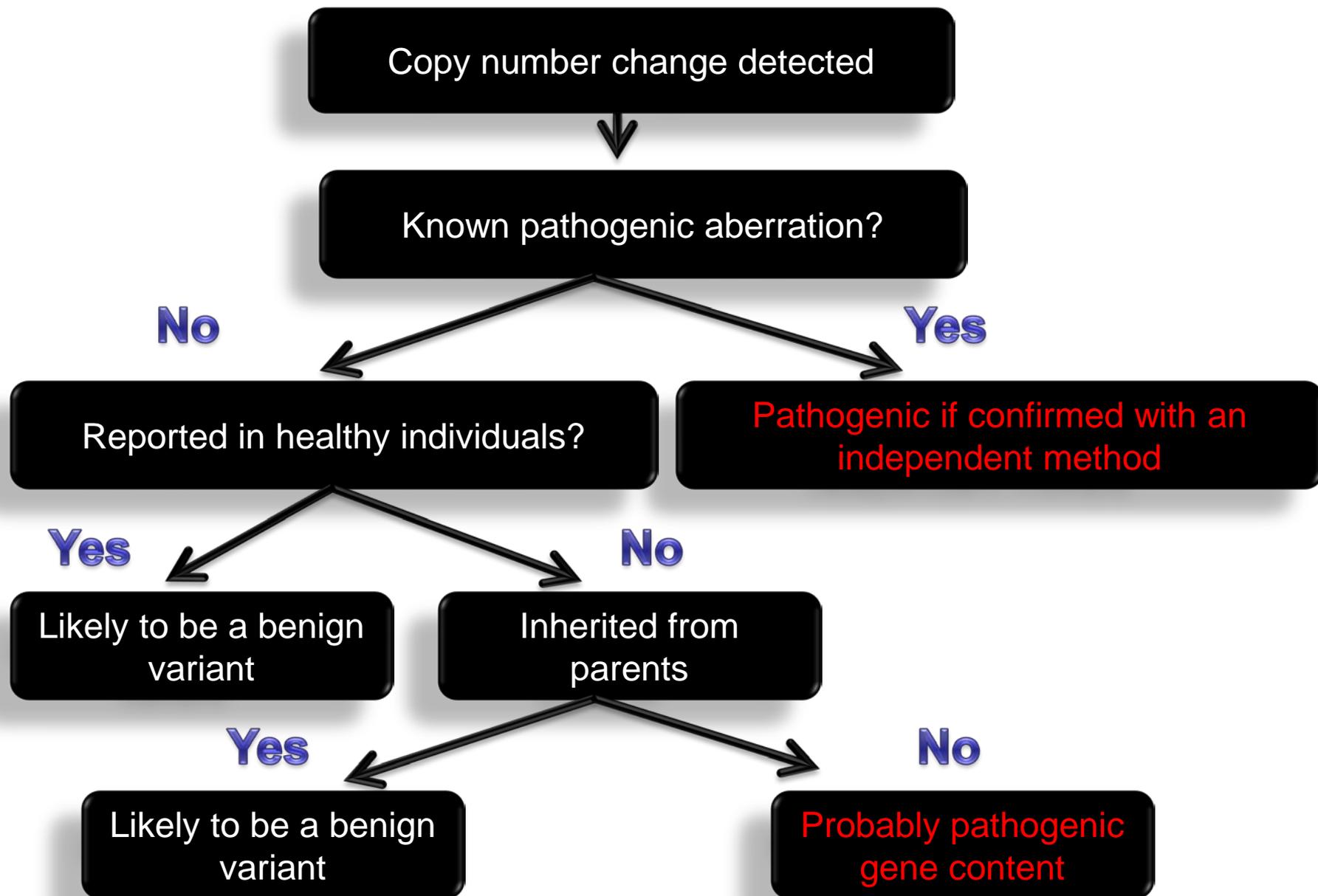
Clinical Utility: Better Diagnostic Yield



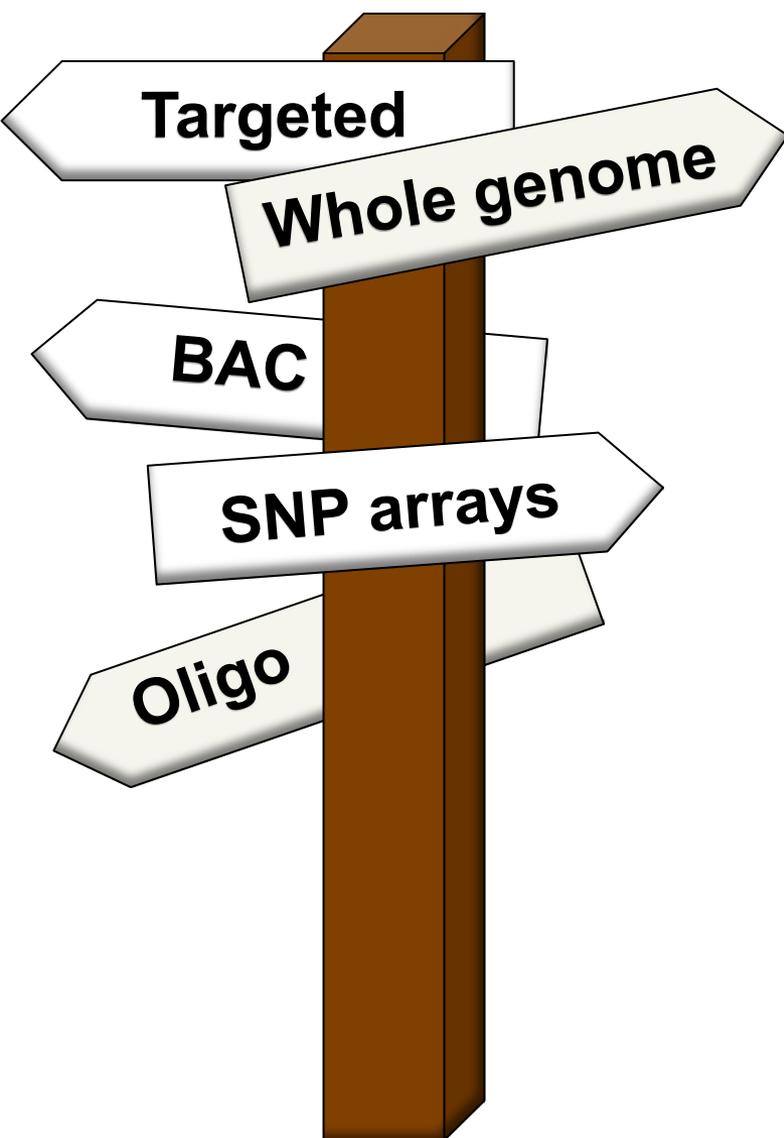
Which genomic imbalances are clinically significant?

- CNA that overlap critical regions of known pathogenic regions
- Parental and family studies
- Databases are very useful
- Nature of CNA important
 - Content, size, nature

Basic Algorithm for Clinical Chromosomal Microarray testing



Gaps in standards and Opportunities in CMA testing



- Incomplete and Inaccurate databases (benign and pathogenic)
- Lack of Uniformity (various platforms with different degrees of resolution, thresholds)
- Urgent need for education
- Lack of reference material