Postnatal Screening and Testing Options

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Learning Objectives

- Understand Newborn Screening process and purpose
- Become familiar with SNP Microarray, Single Gene, Multi-Gene Panel, and Exome genetic testing
- Recognize different considerations that go into genetic test selection

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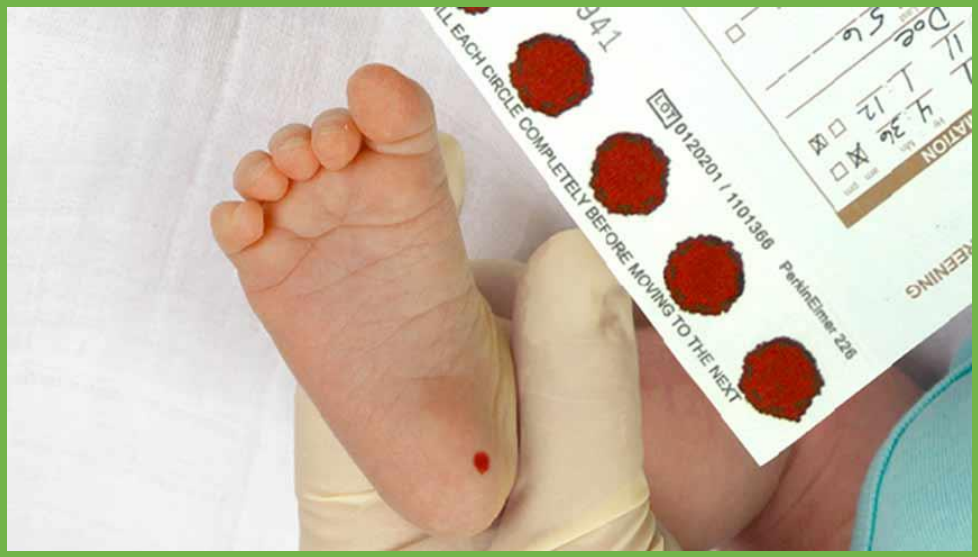
Newborn screening



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Newborn Screening (NBS)

- Began in 1960's with Dr. Robert Guthrie's test for PKU in newborns
- State-run program, Health Resources and Services Administration (HRSA) recommends screening for 35 specific conditions
- Goal is to diagnosis individuals who would benefit from early intervention

http://www.healthy.arkansas.gov

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NBS Roadmap

Blood (from heel-prick) sent to a state-run laboratory
(within 24-48 hours of life)

Physician is notified if any results are abnormal
(no news = good news)

Physician informs parents of the results and discusses a follow-up plan

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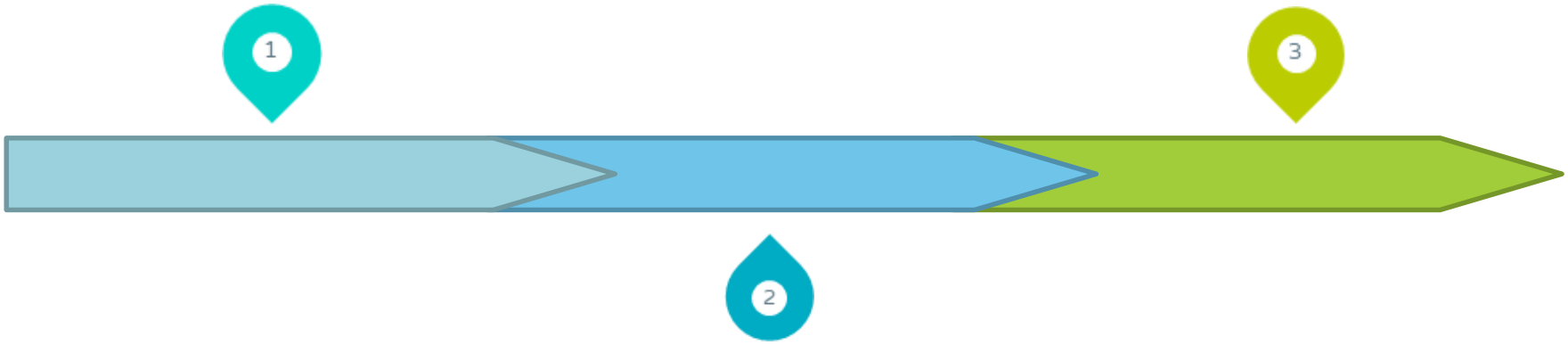
NBS Roadmap Example: Cystic Fibrosis

Newborn flags for elevated IRT levels in blood spot test



Newborn is brought in at 4 weeks old for a sweat chloride test

Confirmatory genetic testing of the *CFTR* gene is ordered for the newborn

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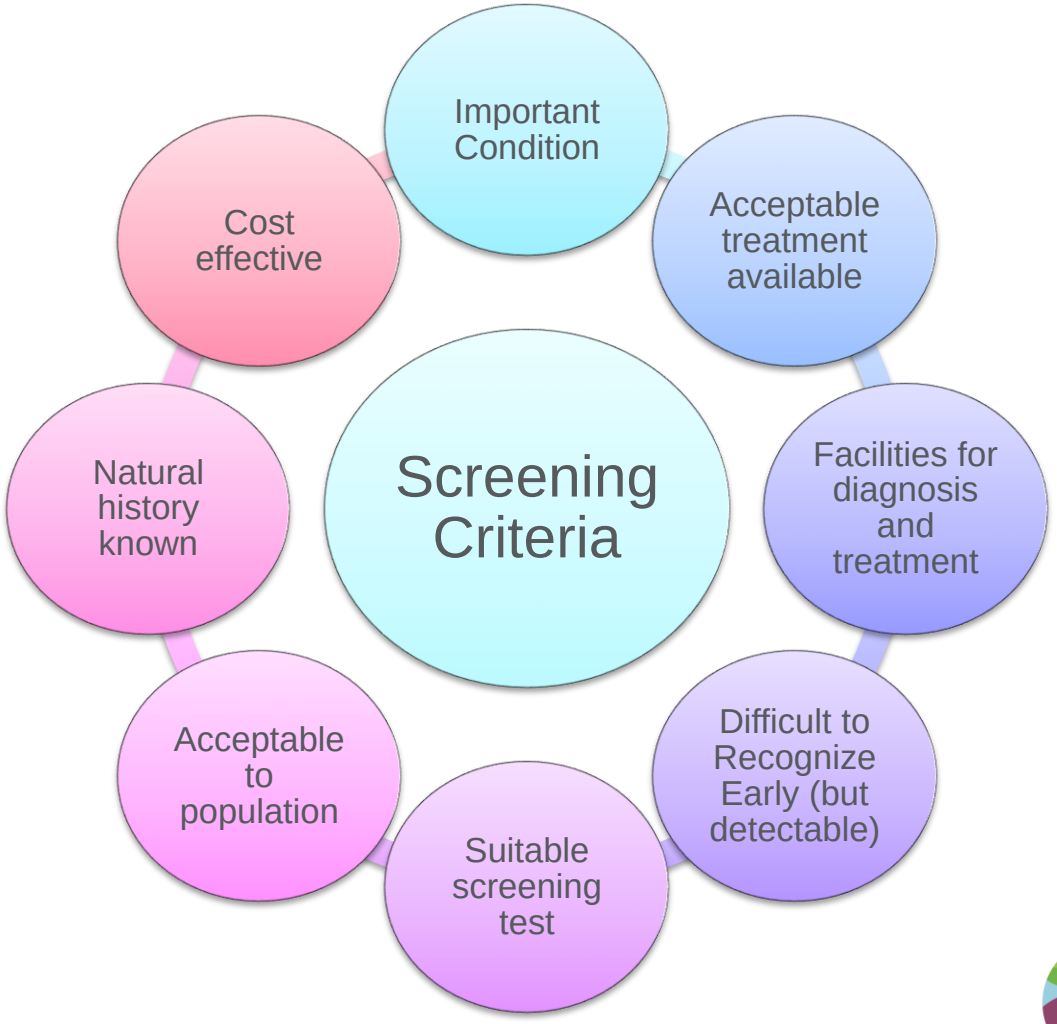
What conditions should we screen for?

WHO Screening Principles



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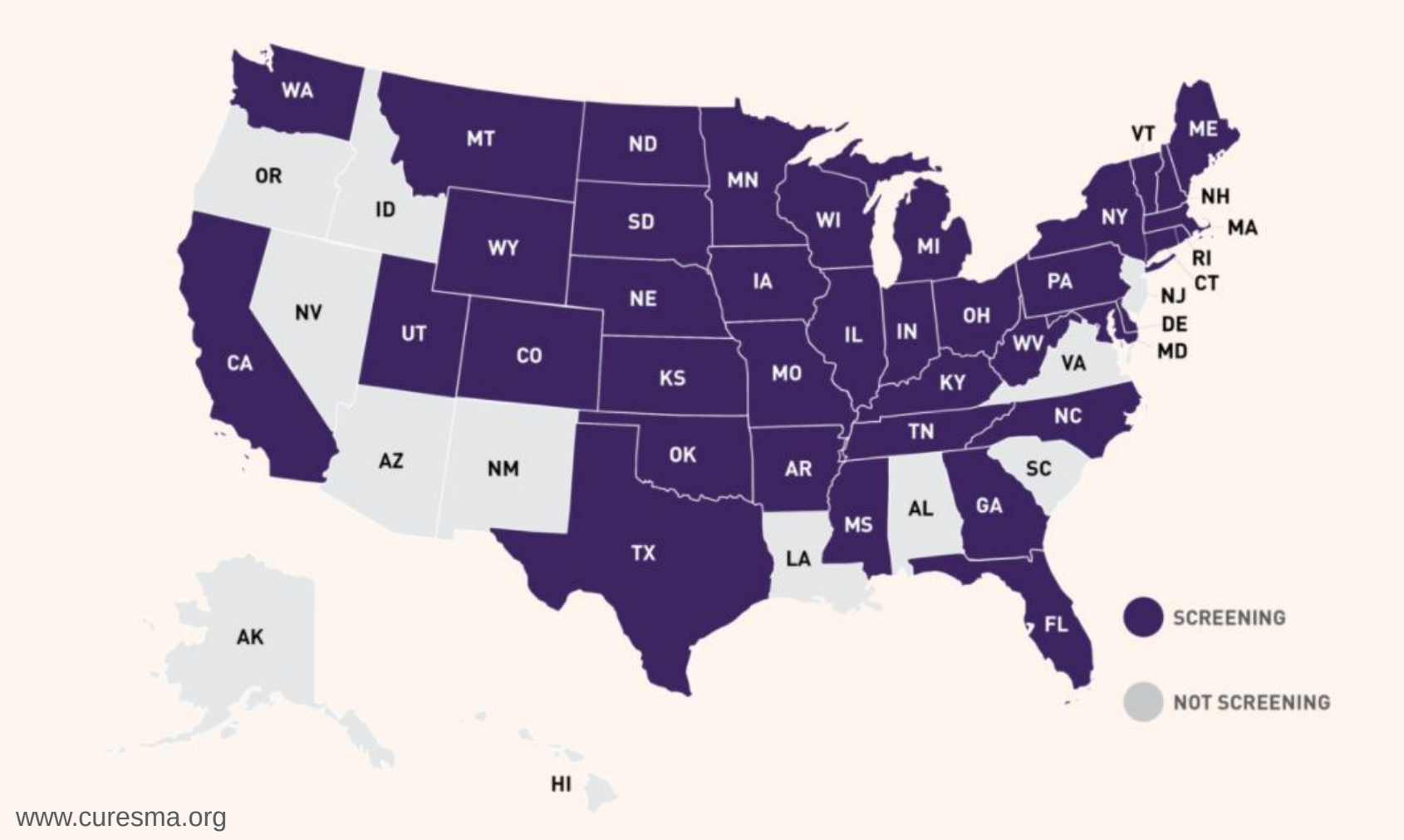
NBS Advancements: SMA

Spinal Muscular Atrophy (SMA)

- AR disease affecting the motor neurons in the spinal cord and brainstem, resulting in progressive motor weakness and atrophy
- FDA-approved treatment
  - Spinraza (Nusinersen) FDA-approved 12/23/2016 - first disease-modifying therapy for SMA patients, all types and ages.
  - Gene therapy (Zolgensma recently approved)
    - SMN1 gene replacement therapy

States screening for SMA on NBS



www.curesma.org

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OH NBS

AA Disorders

Argininemia (ARG)
Argininosuccinic Acidemia (ASA)
Citrullinemia Type I (CIT) and Citrullinemia Type II (CIT II)
Homocystinuria (HCY)
Hypermethioninemia (MET)
Maple Syrup Urine Disease (MSUD)
Phenylketonuria (PKU)
Tyrosinemia Type I,II,III

Endo

Congenital adrenal Hyperplasia
Primary Congenital Hypothyroidism

FA Disorders

Carnitine Acylcarnitine Translocase Deficiency (CACT)
Carnitine Palmitoyl Transferase Deficiency Type II (CPT-II)
Carnitine Uptake Defect (CUD)
Glutaric Acidemia Type II (GA-2)
Long-Chain Hydroxyacyl-CoA Dehydrogenase Deficiency (LCHAD)
Medium-Chain Acyl-CoA Dehydrogenase Deficiency (MCAD)
Trifunctional Protein Deficiency (TFP)
Very Long-Chain Acyl-CoA Dehydrogenase Deficiency (VLCAD)

LSD

Krabbe Leukodystrophy
Glycogen Storage Disease Type II (Pompe Disease)
Mucopolysaccharidosis type I (MPS I)

OA Disorders

2-Methylbutyryl-CoA Dehydrogenase Deficiency (2MBG)
3-Hydroxy-3-Methylglutaryl-CoA Lyase Deficiency (HMG)
3-Ketothiolase Deficiency (BKT)
3-Methylcrotonyl-CoA Carboxylase Deficiency (3-MCC)
Glutaric Acidemia Type I (GA-1)
Isobutyryl-CoA Dehydrogenase Deficiency (IBG)
Isovaleric Acidemia (IVA)
Methylmalonic Acidemia
- Cobalamin Disorders A and B (Cbl A,B)
- Methylmalonyl-CoA Mutase Deficiency (MUT)
- Methylmalonic Acidemia with Homocystinuria (Cbl C, D, F)

Multiple CoA Carboxylase Deficiency (MCD)
Propionic Acidemia (PROP)

Other

Biotinidase Deficiency (BIOT)
Cystic Fibrosis (CF)
Galactosemia (GALT)
Severe Combined Immunodeficiency (SCID)
Sickle Cell Disease (Hb SS)
Other Hemoglobinopathies (e.g. beta-Thalassemia)

NBS Challenges/Limitations

- Limitations to informed consent and pre-test counseling for mandatory test
- False positive results
- Patients lost to follow-up
- Some conditions screened don’t have great therapy options
  - Ex: Krabbe
- Each state is different!

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Postnatal Clinical Diagnostic Testing

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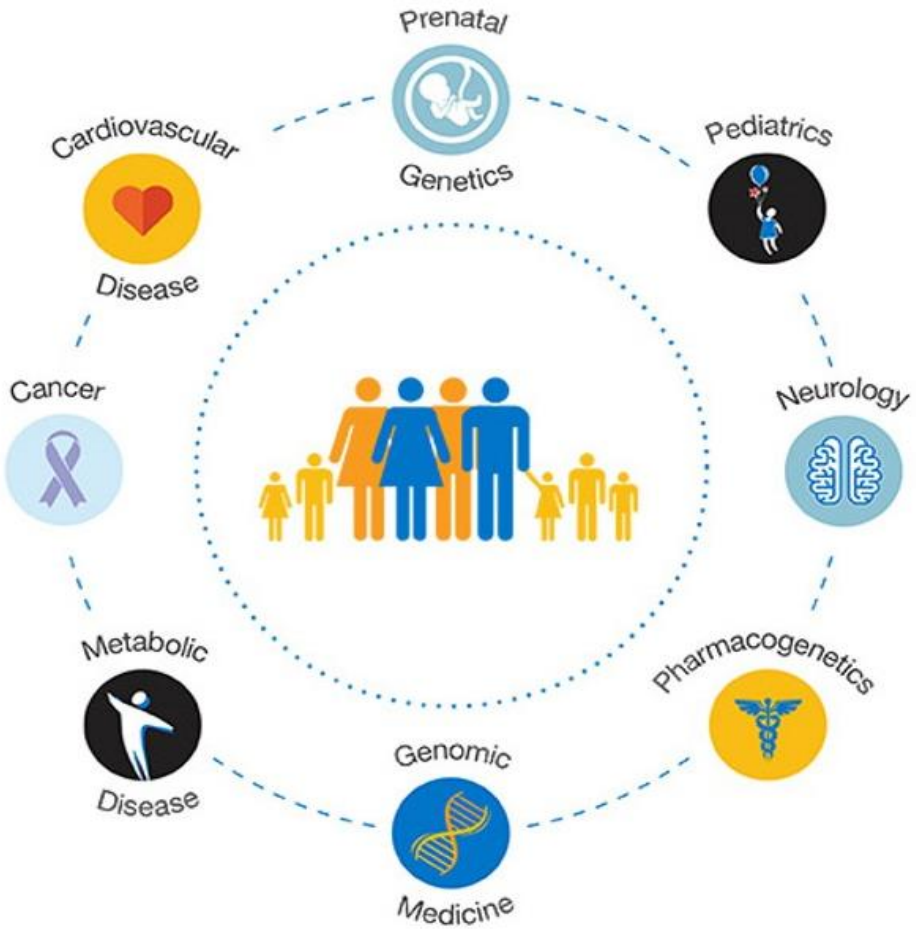
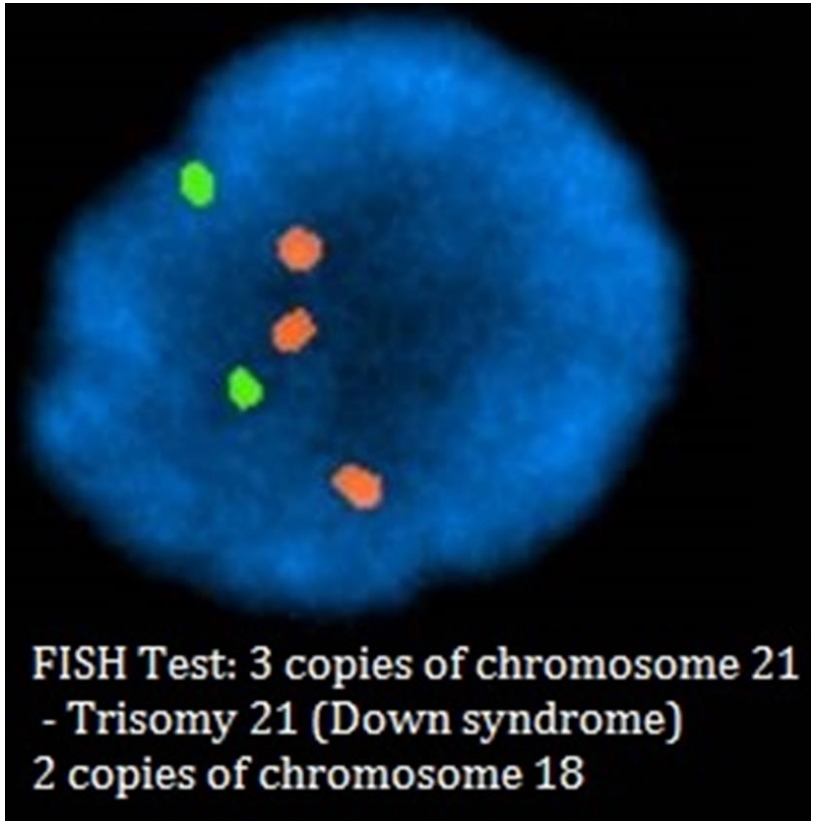
Purpose of Clinical Testing

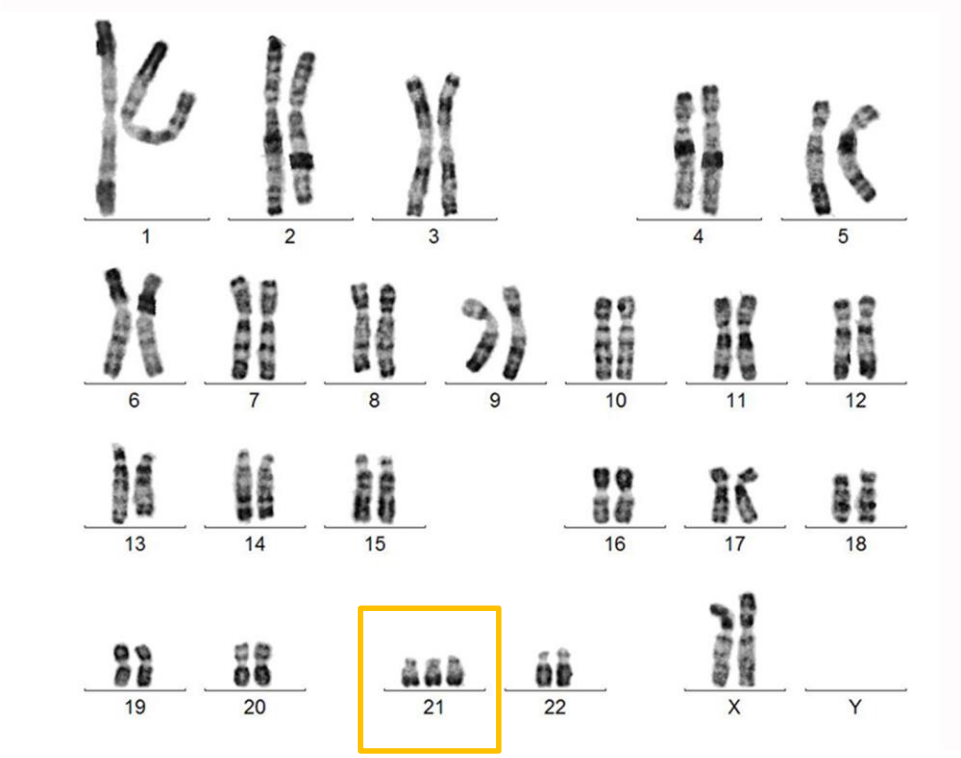


- Find a diagnosis
- Family planning or pregnancy decisions
- Treatment decisions
- Risk for future health concerns
- Risks for family members

Review/Shoutout!



FISH



Karyotype

SNP Microarray

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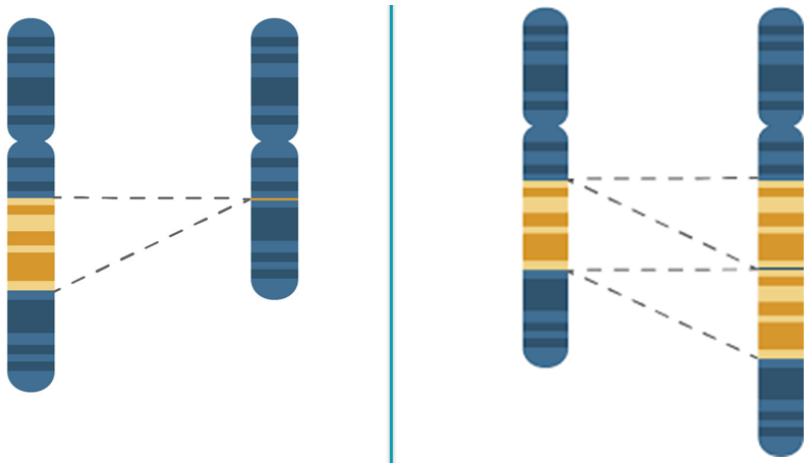
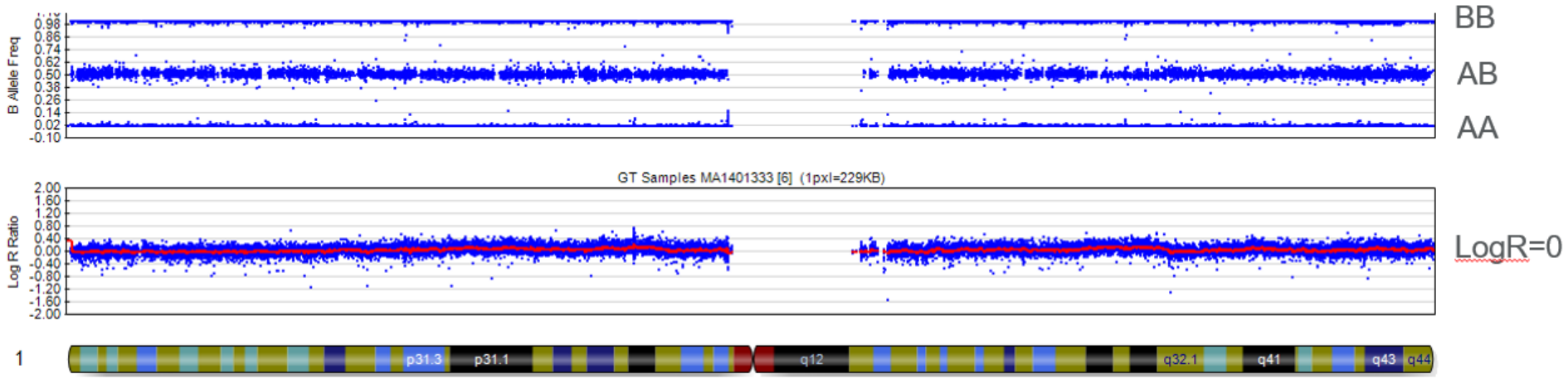
SNP Microarray

- Usually a front-line test in Genetics
- Detects deletions or duplications of DNA
- Can analyze which genes are in deletion or duplication
- Can identify Regions of Homozygosity (ROH)
- Limitations:
  - Cannot tell if a piece of DNA is moved to the wrong location (such as a translocation)
  - Cannot see if there is a small mutation, like a sequence change



SNP Microarray Plot

Typical chromosome



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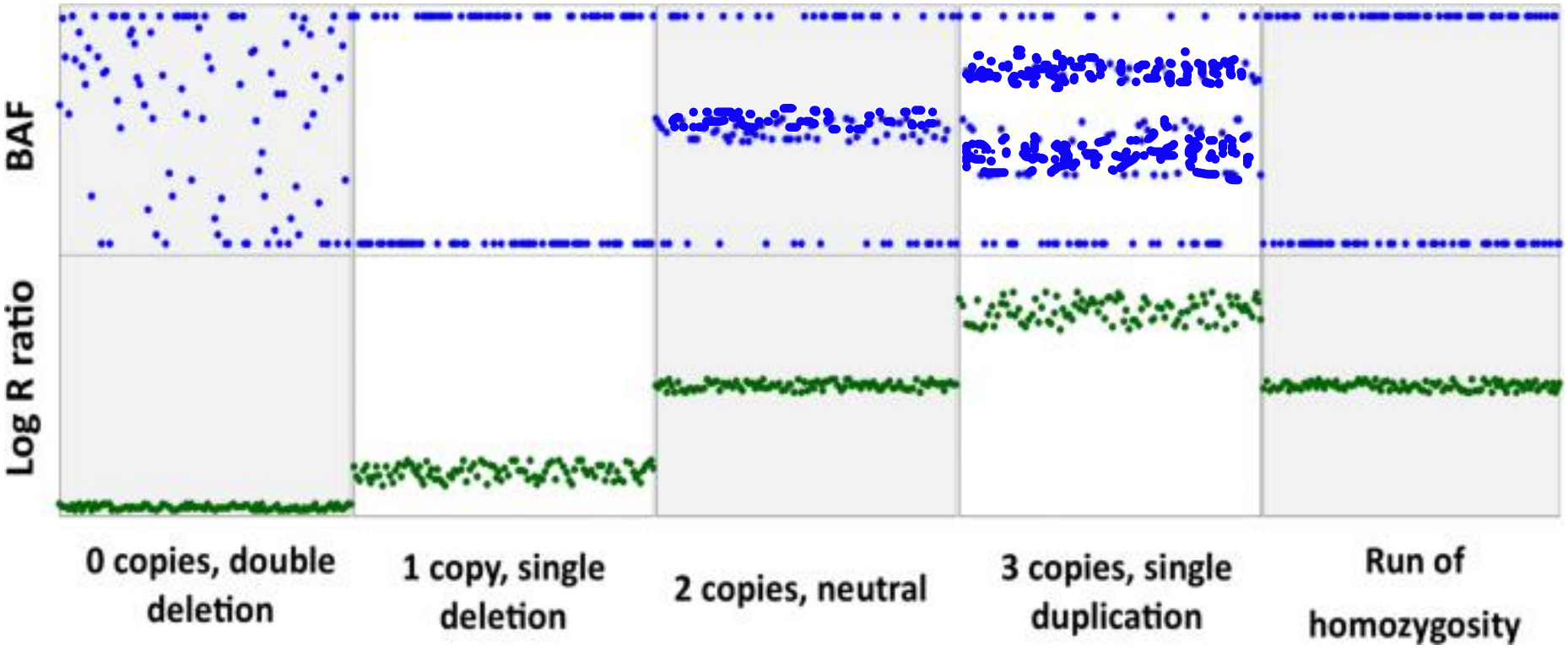
SNP Microarray Plot



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Case Presentation

- 6 year old child presents with global developmental delay, autism, and behavioral issues

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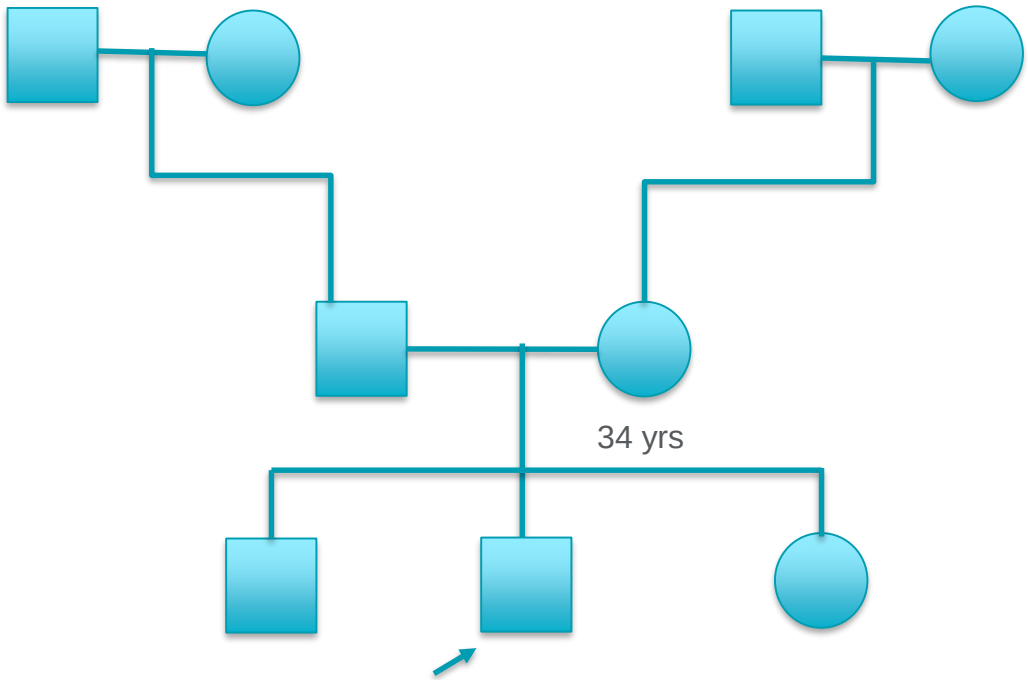
Pedigree



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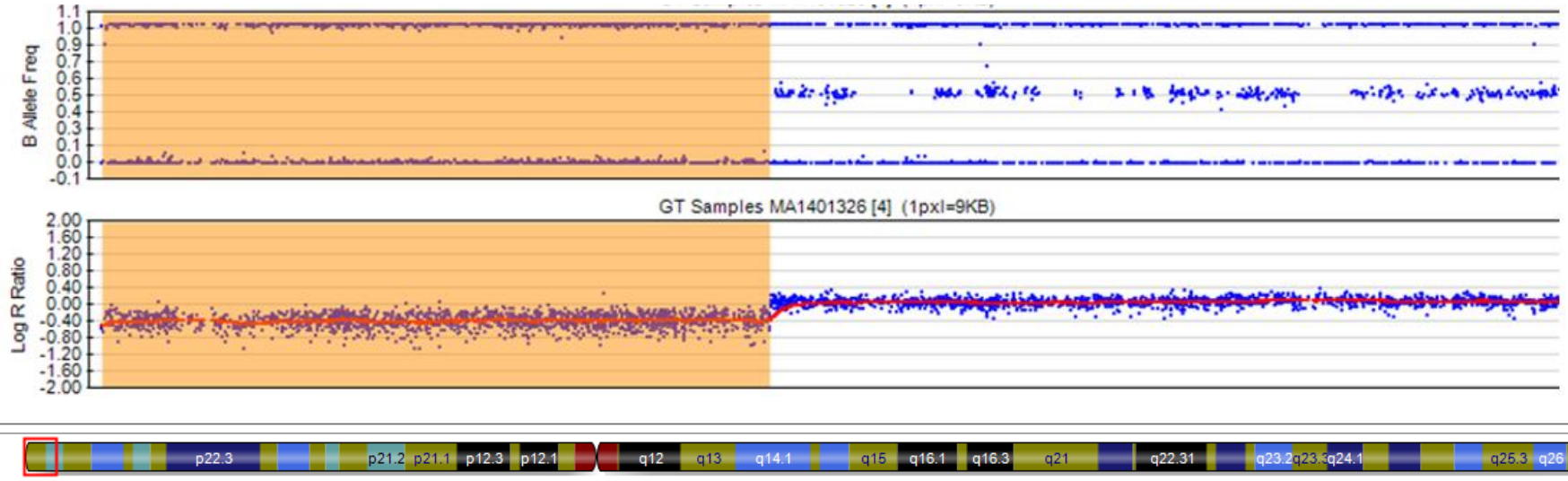
SNP Microarray Plot

Result:
arr[hg19] 6p25.3(108,666-1,505,511)X1

Terminal deletion of 1.4 Mb from the short arm of chromosome 6 (6p25.3)



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Sequencing

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What is Sequencing?

- Looks for smaller changes in our DNA

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Summary of Sequencing

Single Gene Test

One test looks for one gene/condition

- Ex: Cystic fibrosis

Multi-Gene Panel

One test looks at many genes based on:

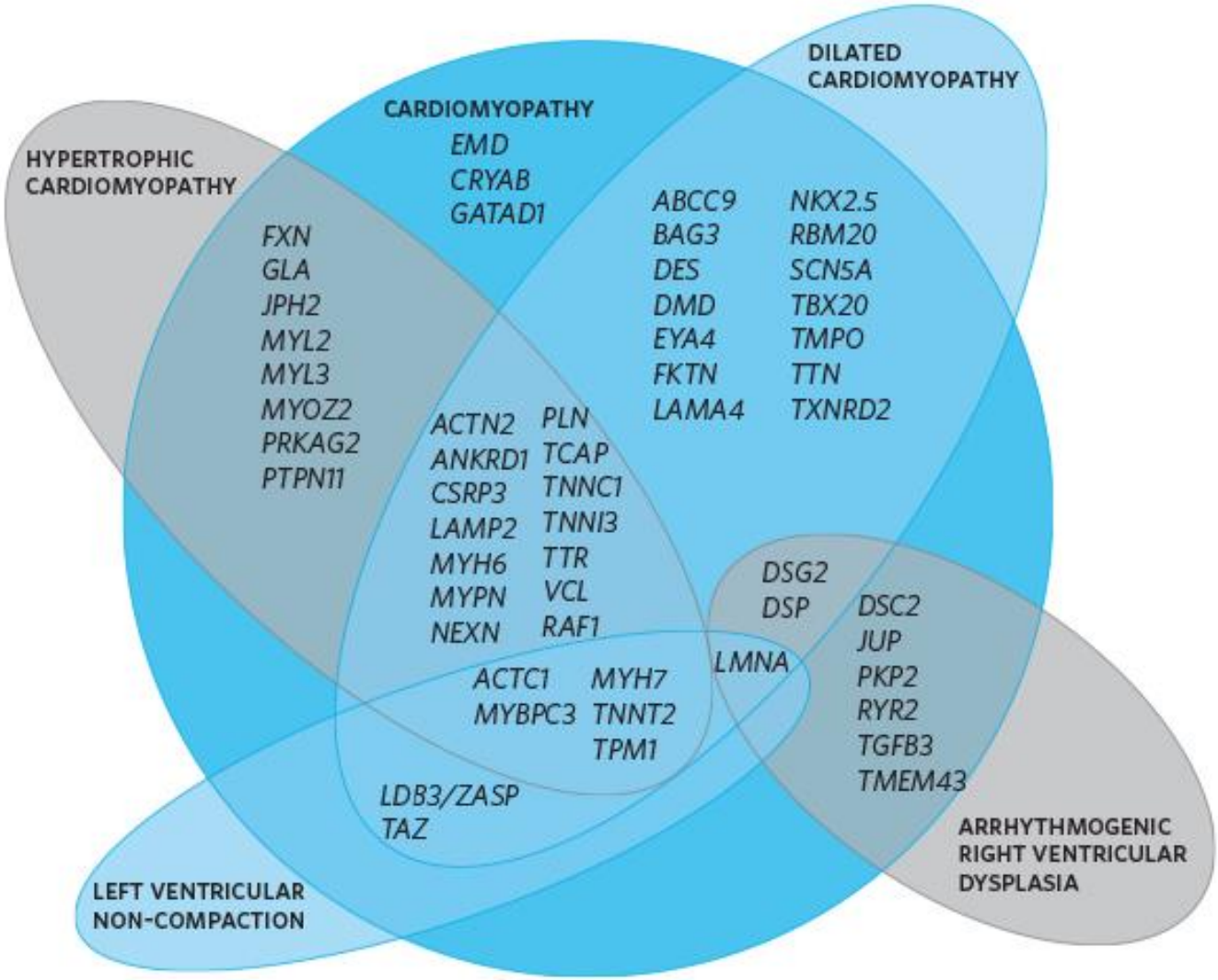
- Single condition/Dx (ex: breast cancer, Fanconi anemia)
- Group of conditions (ex: multiple cancers, autism/ID)
- Symptom (ex: epilepsy, immunodeficiency)

Exome

Reads through the regions of DNA (exons) that directly code for proteins

- One test looks for thousands of disorders!

Single Gene vs. Panel Testing



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Not every panel is equal

Product Comparison

Test Name	Breast/Gyn Cancer Panel	Invitae Breast and Gyn Cancers Panel - Primary + Preliminary-Evidence Genes	Hereditary Breast and Gynecological Cancer Panel Updated (Sequencing)
Actions	Remove From Compare · Add to Order	Remove From Compare · Add to Order	Remove From Compare · Add to Order
Lab Name	✓ GeneDx	✓ Invitae	✓ Blueprint Genetics
Category	Hereditary Breast and Ovarian Cancer Panel Tests	Hereditary Breast and Ovarian Cancer Panel Tests	Hereditary Breast and Ovarian Cancer Panel Tests
Test Code	B273	01201-1	ON1801-SEQ
Source			
Price	$3,272.50	$950.00	$990.00
TAT	14 days	10-21 days	21-28 days
Techniques	Deletion/Duplication · Sequencing	Deletion/Duplication · Sequencing	Sequencing
Mechanisms	MLPA · Next Generation Sequencing · aCGH (Array Comparative Genomic Hybridization)	Next Generation Sequencing	Next Generation Sequencing
Overlapping Genes	ATM · BARD1 · BRCA1 · BRCA2 · BRIP1 · CDH1 · CHEK2 · EPCAM · MLH1 · MSH2 · MSH6 · NBN · NF1 · PALB2 · PMS2 · PTEN · RAD51C · RAD51D · TP53		
Unique Genes	FANCC · MUTYH · POLD1 · RECQL	AKT1 · CDC73 · DICER1 · FAM175A · FANCC · MRE11 · MUTYH · PIK3CA · POLD1 · RAD50 · RINT1 · SDHB · SDHD · SMARCA4 · STK11 · XRCC2	BLM · DICER1 · FANCM · MRE11 · RAD50 · SMARCA4 · STK11 · XRCC2

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INVITAE

INVITAE DIAGNOSTIC TESTING RESULTS

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Patient name:	Jane Doe	**Sample type:**	Blood	**Report date:**	
DOB:		**Sample collection date:**		**Invitae #:**	
Sex:	Female	**Sample accession date:**		**Clinical team:**	
MRN:					

Reason for testing

Diagnostic test for personal history of disease

Test performed

Sequence analysis of the 207 genes listed in the Genes Analyzed section.

- Invitae Primary Immunodeficiency Panel

RESULT: POSITIVE

One homozygous Pathogenic variant identified in CTPS1. CTPS1 is associated with autosomal recessive CTPS1 deficiency.

One Pathogenic variant identified in PMM2. PMM2 is associated with an autosomal recessive congenital disorder of glycosylation.

Additional Variant(s) of Uncertain Significance identified.

GENE	VARIANT	ZYGOSITY	VARIANT CLASSIFICATION
CTPS1	c.1692-1G>C (Splice acceptor)	homozygous	PATHOGENIC
PMM2	c.422G>A (p.Arg141His)	heterozygous	PATHOGENIC
MOGS	c.721C>T (p.Arg241Cys)	heterozygous	Uncertain Significance

About this test

This diagnostic test evaluates 207 gene(s) for variants (genetic changes) that are associated with genetic disorders. Diagnostic genetic testing, when combined with family history and other medical results, may provide information to clarify individual risk, support a clinical diagnosis, and assist with the development of a personalized treatment and management strategy.

Laboratory Director: Kristin Gibson, PhD, FACMG, NYCQ, CGMB, ErCLG
Invitae 1400 16th Street, San Francisco, CA 94103 · clientservices@invitae.com · 800.436.3037

Report Example

CCHMC Approach

- If child presents with global developmental delay and autism, at CCHMC our standard of care is to order:
  1. SNP Microarray
  2. Fragile X
  3. Autism/ID panel*

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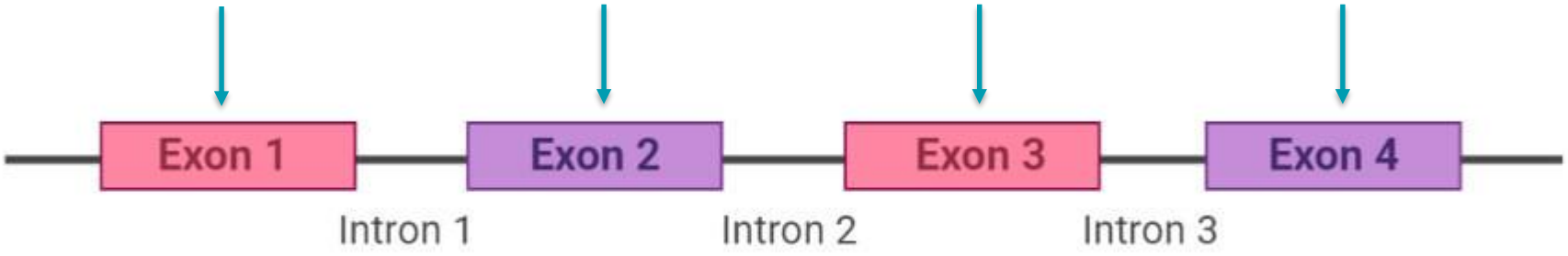
Whole Exome Sequencing



- Sequencing of Exons
- Proband Only vs. Trio approach
- Answer found only ~25% for Trios
- Primary vs. Secondary Findings

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Whole Genome vs. Whole Exome

Genome vs. Exome

Genome

A genome is like watching a football game from beginning to end.

Vs.

Exome

An exome is like reading about the game highlights the next day.

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Exome Result Example

CHARACTERIZED GENETIC ETIOLOGIES: **LIKELY POSITIVE** **ALTERATION(S) WITH LIKELY CLINICAL RELEVANCE DETECTED**

Results Summary

Gene Symbol	Gene Inheritance	Characterized/ Uncharacterized Gene*	Protein Change	Nucleotide Change	Genotype	Alteration Type	Alteration Classification	Clinical Correlation
PMPCA	Autosomal recessive	Characterized	p.A377V	c.1130C>T	Homozygous, biparental	Missense	Likely pathogenic	Positive

Patient's likely diagnosis based on molecular results:

Spinocerebellar ataxia, autosomal recessive 2 (OMIM_213200)

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Case Examples:
Single gene vs. panel vs. Exome

1. 15-year-old adolescent presents with hypertrophic cardiomyopathy

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Case Examples:
Single gene vs. panel vs. Exome

1. 15-year-old adolescent presents with hypertrophic cardiomyopathy

2. 3-year-old presents with a family history of cardiomyopathy, and his mother has a known mutation in *EMD* gene

3. 5-day-old presents with dysmorphic facial features, failure to thrive, multiple congenital anomalies, and ataxia with no recognizable condition. All frontline testing is normal

Predictive or Predisposition Testing

- Used to determine a healthy person's predisposition to develop disease
- Testing usually sought based on family or medical history
- Should only be done in minors if there are medical interventions that would be recommended as children, if the testing is positive

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Predictive or Predisposition Testing

- Treatments/Screening available
  - Cardiac Genetic Panels
  - Cancer Genetic Panels
- **Treatment not available**
  - AD Alzheimer's Disease
  - Huntington's Disease

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Proactive Genetic Testing

- Marketed to healthy adults who want to understand their DNA and focus on prevention
- For example: Three testing options that analyze up to 147 genes that are well-established indicators of a significantly increased risk of developing hereditary cancers, cardiovascular conditions, and other medically important disorders

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Testing Considerations

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Test Interpretation

- Our technology is improving faster than our ability to correctly interpret results
- Types of Results:

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  - Negative
    - Can rule out some conditions
    - Can make other conditions less likely
    - Doesn't take away a clinical diagnosis for some conditions

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  - Negative
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    - Doesn't take away a clinical diagnosis for some conditions
  - Variant of Uncertain Clinical Significance (VUS)
    - Does not change medical management
    - Could be reclassified in the future

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Other Considerations

- Picking the best test for your patient
  - Not every test is equal!
- Insurance
  - Will obtaining a diagnosis change management?
  - Will insurance company cover cost of testing?
  - GINA (Genetic Information Nondiscrimination Act)
- Incidental findings
  - Find Dx you weren't looking for
  - Parental relationships that weren't disclosed

Questions?

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