Understanding your genetic test results

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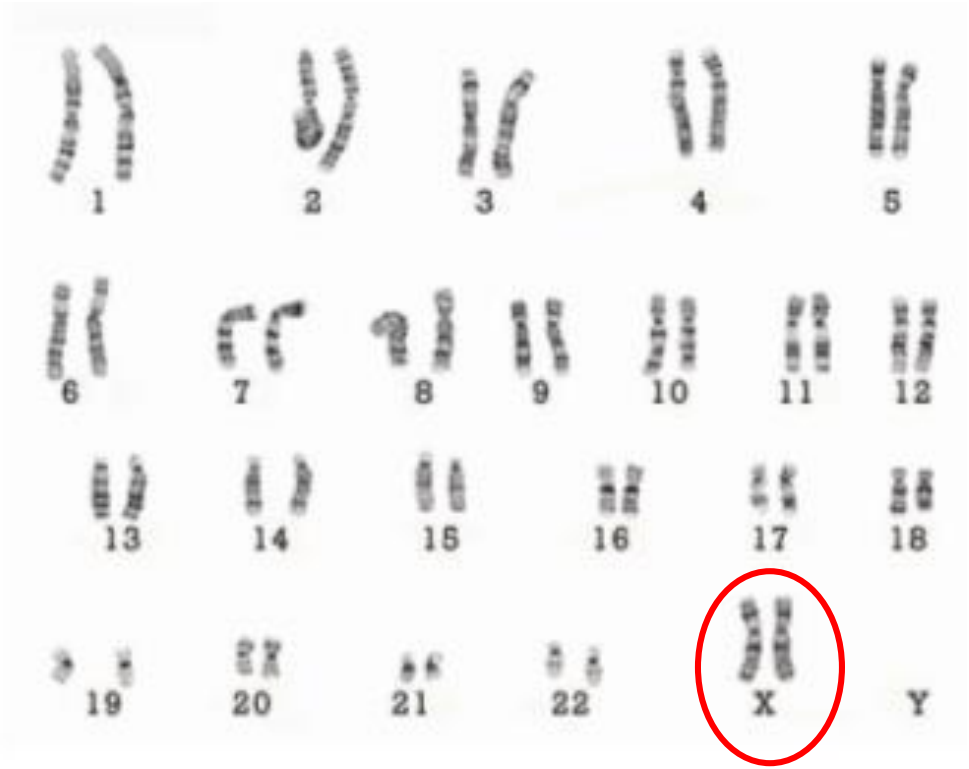
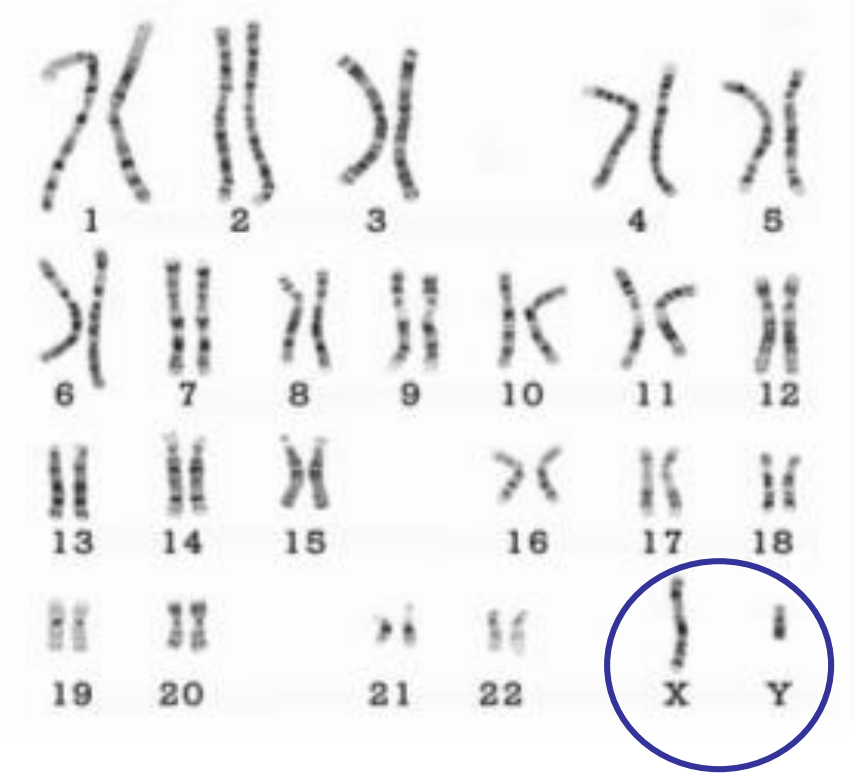
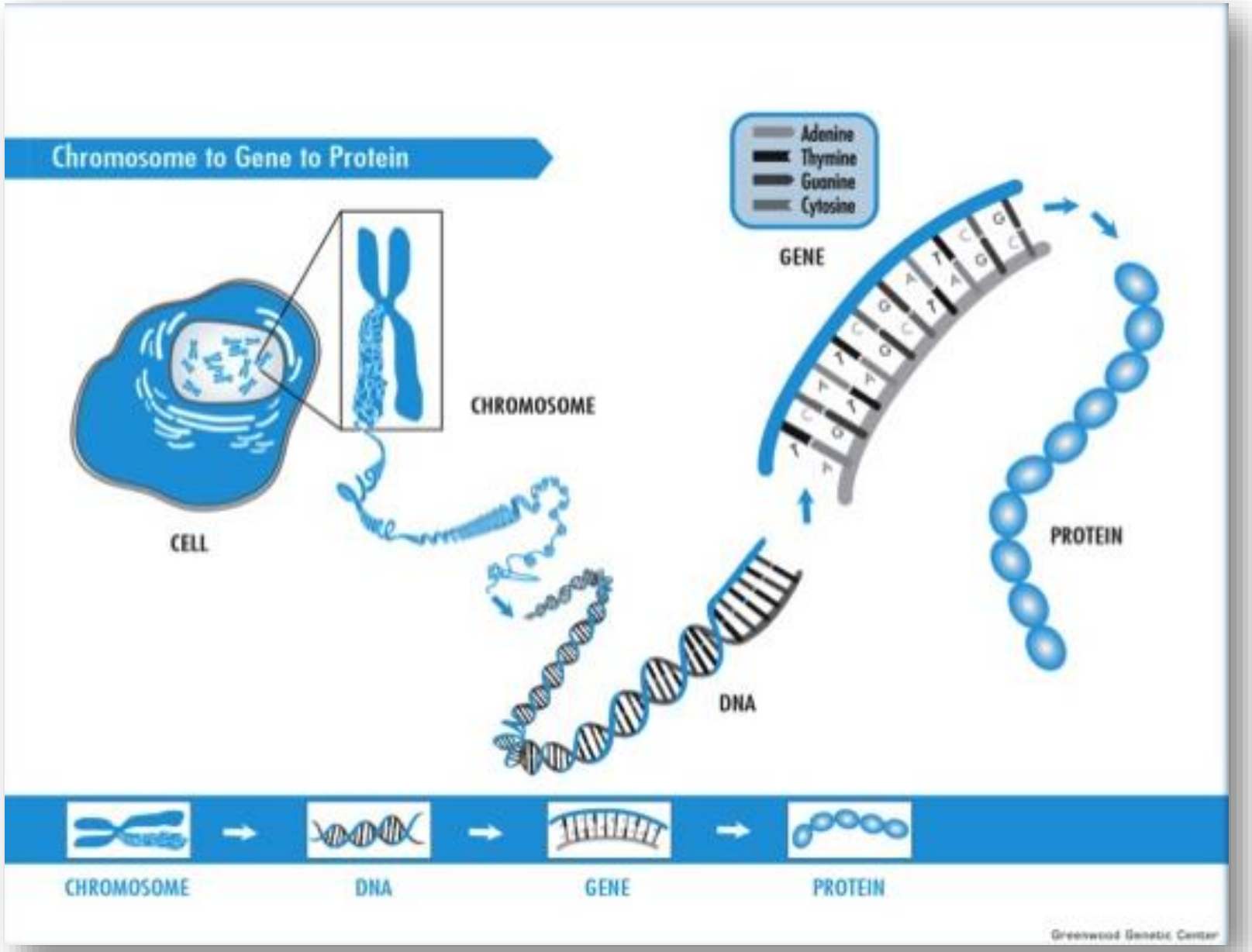
Chromosomes

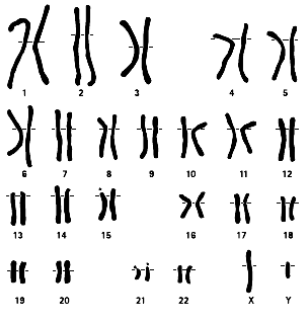





Chromosome to Gene to Protein
Adenine
Thymine
Guanine
Cytosine
GENE
CHROMOSOME
CELL
PROTEIN
DNA
CHROMOSOME
DNA
GENE
PROTEIN
Greenwood Genetic Center

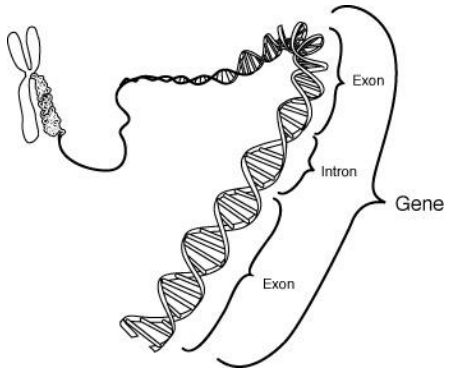
Genetics

Genome



Chromosome

Gene



DNA – A, C, T, G

Enclyclopedia

Volume

Sentence

Alphabet- A, B, C, D…

Genetic variants

- Variants are changes in a gene.
- We ALL have genetic variants in our genes – they are what make us unique!
- Some changes are “benign” and do not cause any problems.
- Some changes are more serious and can cause problems with what that particular gene is supposed to do.
  - These are often called “Pathogenic” variants.

Types of variants

Original

THE CAT HAD RED FUR AND RAN FAR.

Deletion

THE CAT HAD RED FUR AND RAN FAR.

THE CAT HAR EDF URA NDR ANF AR.

Missense

THE CAT HAD RED FER AND RAN FAR.

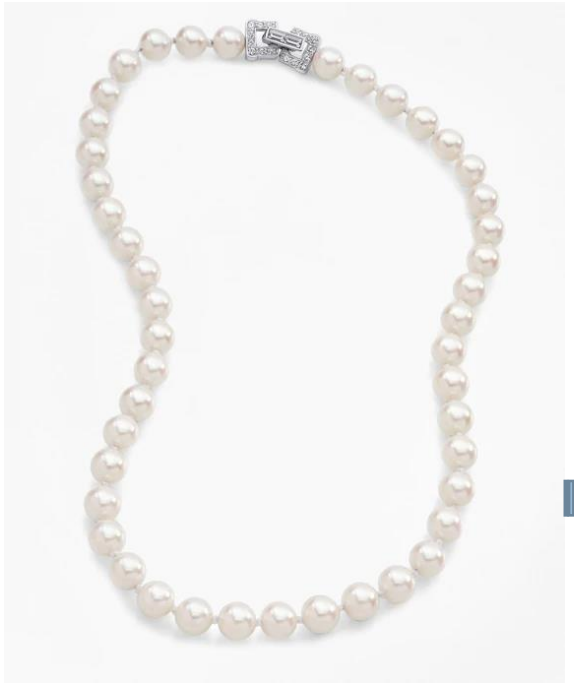
THE MAT HAD RED FUR AND RAN FAR.

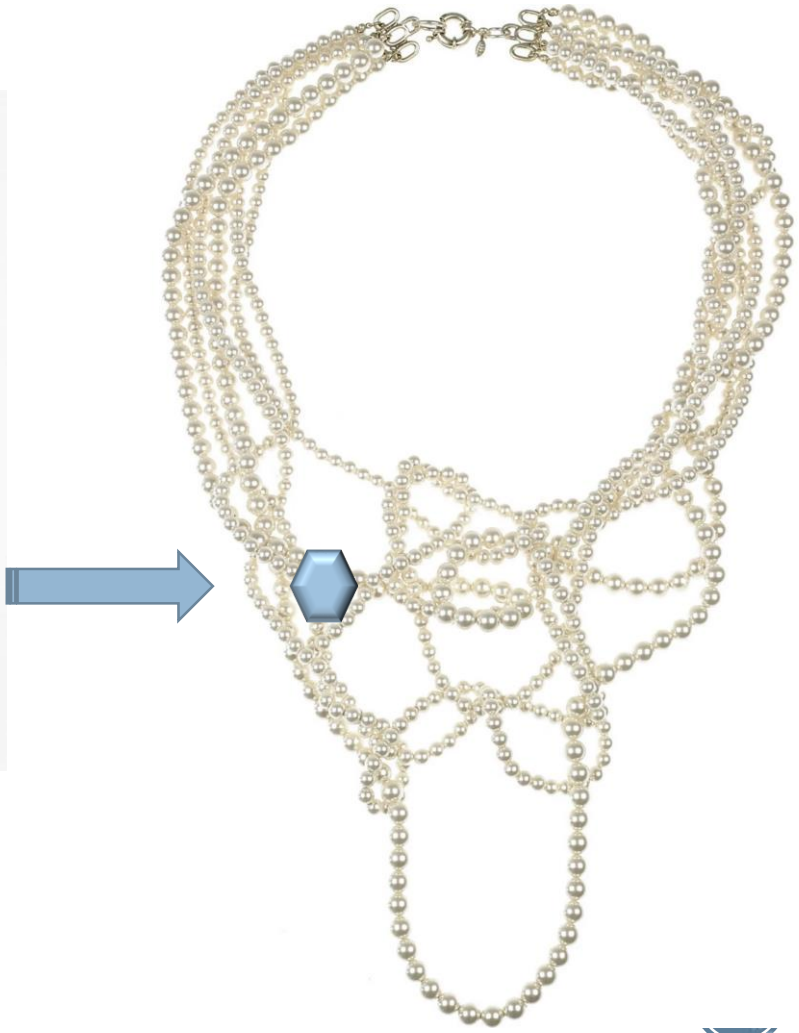
Nonsense

THE CAT HAD RED. 

Types of variants in Noonan syndrome

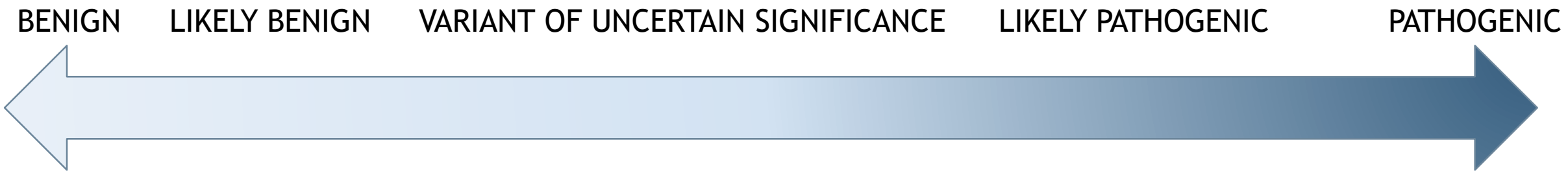
- Many of the variants that cause NS are misspellings in the genes.
- The variant changes an amino acid (bead).
- This change causes the gene to code for a protein that is different than the original protein.
- This change affects the ability of the protein to work correctly.





Variant classification

- Genetic testing laboratories will classify variants along the following spectrum:



Benign vs. Pathogenic

- Often times, the lab may be able to predict if a change in a gene is likely to be benign or pathogenic IF:

- The specific change has been seen before in other children with the condition OR

- If the specific change has been seen before in healthy, unaffected individuals or their parents.

Variants of unknown significance (VUS)

- If a gene change is found but has NOT been seen before or reported in the medical literature, then it can make it very hard to interpret the results.
- These are called "variants of uncertain significance" (VUS)

- Testing other family members may help us interpret the results.
- If a parent or unaffected sibling has the same change, then it is probably benign.
- If neither parent has the same change, then it might be pathogenic.

How to interpret VUS

- Familial testing – does the variant segregate with the phenotype/condition?
- Software prediction programs – SIFT and PolyPhen; can give predictions about the effect of the variant on the gene function/protein
- Conservation studies – Is the location of the variant highly conserved across different species?
- Population databases – ExAC – has this specific variant been seen in “healthy” populations and if so, with what frequency?

Genotype-Phenotype correlations

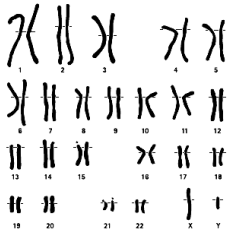
- Sometimes knowing the genetic variant or size and location of the chromosome abnormality can help predict the clinical features that may be more likely to occur in an individual.
- But there remains significant variability both between and within families/individuals who may carry the same genetic change, so we need to be cautious with making assumptions and predictions.

- Genotype = Genetic cause (specific gene or variant)
- Phenotype = Clinical features of the person (heart problems, learning difficulties, etc)

Genetic Testing Options

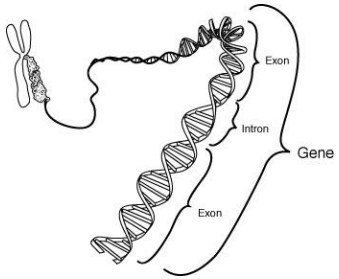
Genetics

Genome



Chromosome

Gene



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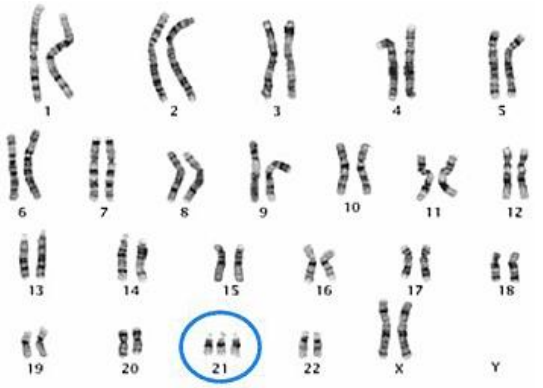
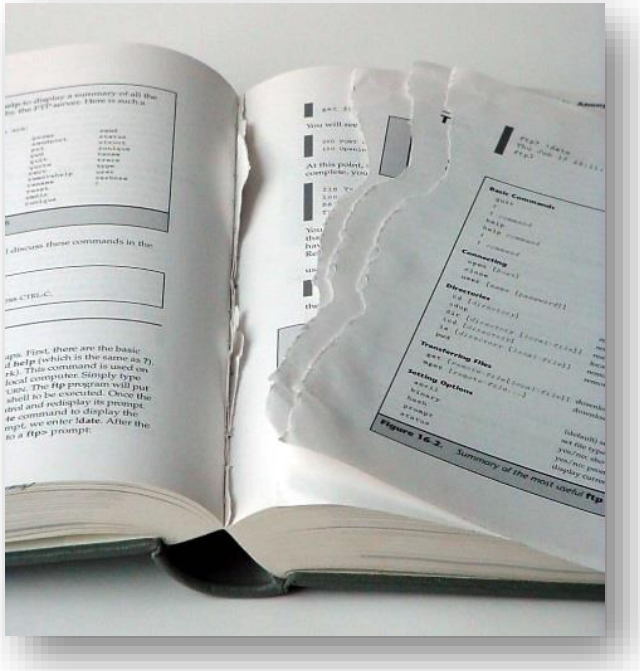
Chromosome Analysis

- Chromosome analysis involves looking at all of the chromosomes under a microscope.
- Looking for extra or missing entire chromosomes or large pieces of chromosomes.
- Example: Down syndrome (trisomy 21)
- Limitations: Can only detect relatively LARGE chromosome abnormalities, but will miss smaller ones.



Chromosome microarray analysis (CMA)

- Chromosome microarray analysis is a computer technique that analyzes a person's DNA to a reference DNA.
- It is able to detect very small missing or extra pieces of chromosome material.
- We are able to see much more detail than a routine chromosome study.



Single Gene Testing

- Single gene testing involves sequencing/reading a single gene and looking for variants (i.e. misspellings) in any of the letters.
- You have to know your gene of interest
- Results are usually available in 2-4 weeks

Human brian is still evolving, says scientist

THE human brain is still evolving, scientists said today.

Researchers at the University of Chicago have identified two genes linked to brain size which are rapidly evolving in humans.

man evolution — the growth of brain size and complexity — is likely still going on.

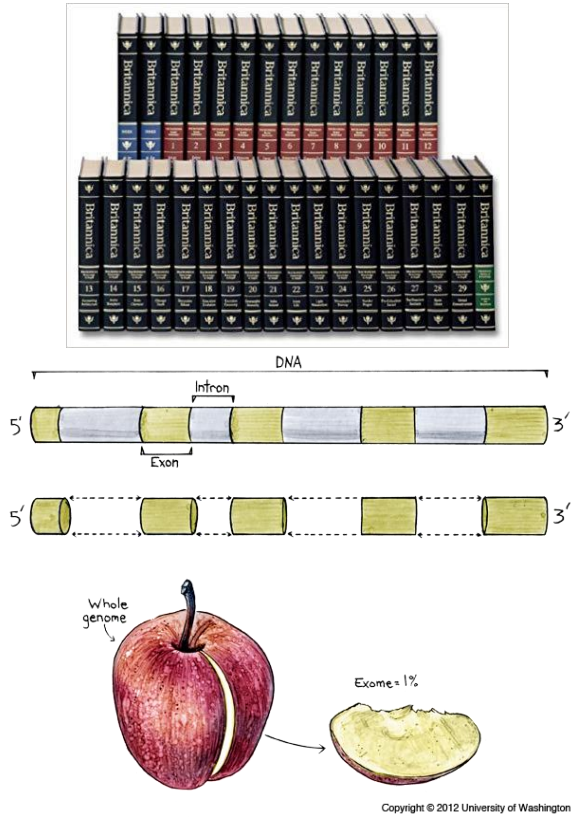
"Meanwhile, our environment and the skills we need to survive in it are changing faster then we ever

Multi-Gene Panel Testing

- Often times, genetic conditions are caused by more than one gene.
- Multi-gene panels involve looking at 3-100+ genes
- Identify misspellings in any of those genes that may cause the condition.
- Results are usually available in 4-6 weeks

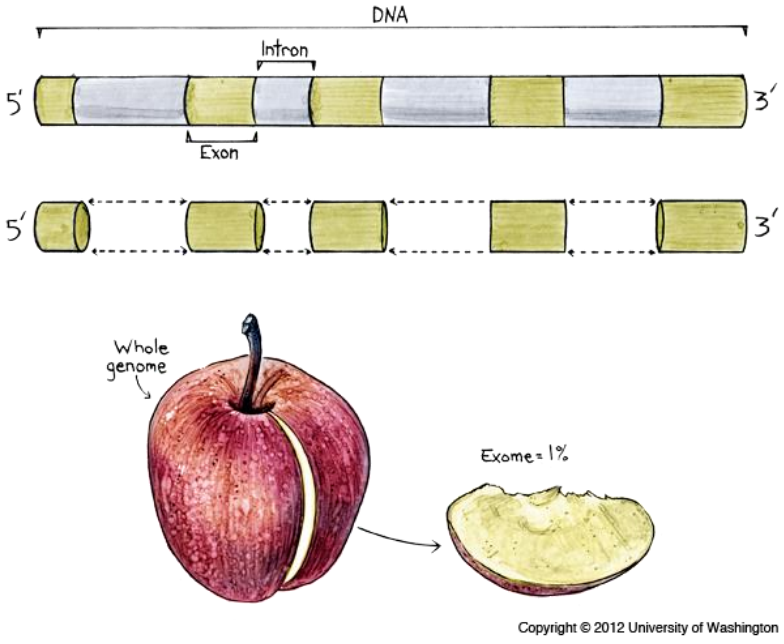
Whole exome sequencing (WES)

- Whole exome sequencing examines the coding parts (exons) of all of our genes.
- We have ~20,000, however, we only know the function of about 4,000 of those genes.
- Interpretation can be difficult if we identify variants with unknown/uncertain significance.
- The likelihood of getting an “answer” is 30-50% depending on the patient population and indication



Whole genome sequencing

- Whole genome sequencing examines the coding AND non-coding regions of our entire genome.
  - 3.2 ***billion*** base pairs
- It also has the ability to detect chromosome deletions and duplications, similar to a chromosome microarray analysis.
- Clinical availability is still limited
- Interpretation can be difficult given the uncertainty of the types of results that may be uncovered.
- Great potential as a first tier test for individuals with suspected genetic conditions.



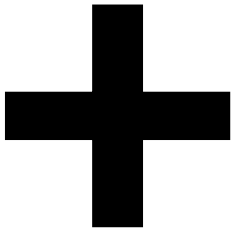
Genetic testing for Noonan syndrome

- Individuals may be diagnosed by:
- Panel testing for Rasopathies, developmental delay, congenital heart defects, cardiomyopathy
  - Would need to make sure that the genes for NS are on the list of genes being tested
- Whole exome/genome sequencing
- Known familial variant testing
  - This is used *only if* the variant in the family is already known and a family member wants to get testing for that condition only.

Understanding your genetic test result

Genetic test results may come back:

- Positive - meaning that a known disease-causing (pathogenic) variant was identified



- Negative – no disease-causing variants were identified at this time

- Variant of uncertain significance

Non-diagnostic test results

- If results are NEGATIVE or with a VUS this does NOT necessarily mean that you/your child does not have Noonan syndrome.
- They could still have NS or a Rasopathy, but we just have not identified the specific genetic cause yet. The field of genetics is constantly evolving and we are continuing to learn about new genes and variants that cause NS and other Rasopathies.
- In addition, the genetic test result may come back with a variant that has not been previously reported in the literature or in databases. This could mean that the change is rare or unique in your family OR there may be other individuals with the same change, who have just not been reported in the medical literature.
- If more than one variant is identified on the genetic test, further discussion with a genetics provider is needed to help interpret the significance of each finding.

Sample Exome report

PRIMARY FINDINGS: **Heterozygous for a Pathogenic Variant in *PPP2R5D***

SECONDARY FINDINGS: **None Detected**

CARRIER STATUS: **Heterozygous for Pathogenic Variants in *CYP21A2* and *GALT***

PG DISCOVERY FINDINGS: **One Variant Detected**

INDICATION FOR TESTING: Intellectual disability, mild, specific learning disability, delayed speech and language development, long face, high, narrow palate

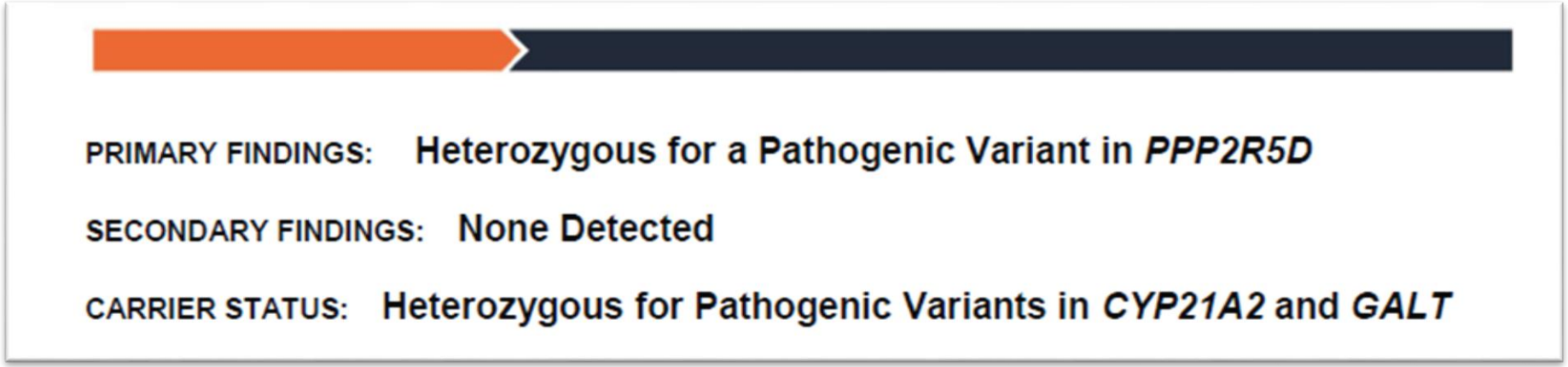
Variants in genes known to be associated with phenotype:

Gene, Transcript	Mode of Inheritance, Gene OMIM	DNA Variations, Predicted Effects, Zygosity	ClinVar ID	Highest Allele Frequency in a gnomAD Population	In Silico Missense Predictions	Interpretation
PPP2R5D, NM_006245.3	AD, 601646	c.598G>A, p.Glu200Lys, Heterozygous (*De novo*)	217456	Not Present	Conflicting	PATHOGENIC

Mode of Inheritance: Autosomal Dominant=AD, Autosomal Recessive=AR, X-Linked=XL
ClinVar ID: Variant accession (www.ncbi.nlm.nih.gov/clinvar)
GnomAD: Allele Frequency registered in a large population database (gnomad.broadinstitute.org). Value listed is the highest allele frequency reported within one of seven population categories recognized in gnomAD v.2.0 (The "Other" population is excluded).
Missense Predictions: Summarized output (Damaging, Conflicting, or Tolerated) via PolyPhen-2, SIFT, MutationTaster, and FATHMM (PMID: 26555599).

RESULTS AND INTERPRETATIONS: This patient is heterozygous in the *PPP2R5D* gene for a *de novo* variant designated c.598G>A, which is predicted to result in the amino acid substitution p.Glu200Lys. This is a recurrent *de novo* variant that has been reported to be causative for autosomal dominant intellectual disability (Table 1, Houge et al. 2015. PubMed ID: 26168268; Table 1, Loveday et al. 2015. PubMed ID: 25972378; Supp. Tables 6 and 10, Lelieveld et al. 2017. PubMed ID: 28867141; Table 1, Reijnders et al. 2017. PubMed ID: 29051493). In summary, we interpret this variant as pathogenic.

Understanding genetic test results



- Heterozygous = one variant was identified on one copy of the gene
  - Homozygous = two of the *same* variants were identified in the same gene – one on each copy of the gene
  - Compound heterozygous = two *different* variants were identified in the same gene – one on each copy of the gene
- Pathogenic = causes the condition/disease
  - Likely pathogenic
  - Variant of uncertain significance
  - Likely benign
  - Benign

Understanding genetic test results

Gene, Transcript	Mode of Inheritance, Gene OMIM	DNA Variations, Predicted Effects, Zygosity	ClinVar ID	Highest Allele Frequency in a gnomAD Population	In Silico Missense Predictions	Interpretation
PPP2R5D, NM_006245.3	AD, 601646	c.598G>A, p.Glu200Lys, Heterozygous (*De novo*)	217456	Not Present	Conflicting	PATHOGENIC

Nucleotide # 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 … 598

Gene: THE MAT HAD RED FUR AND RAN FAR.

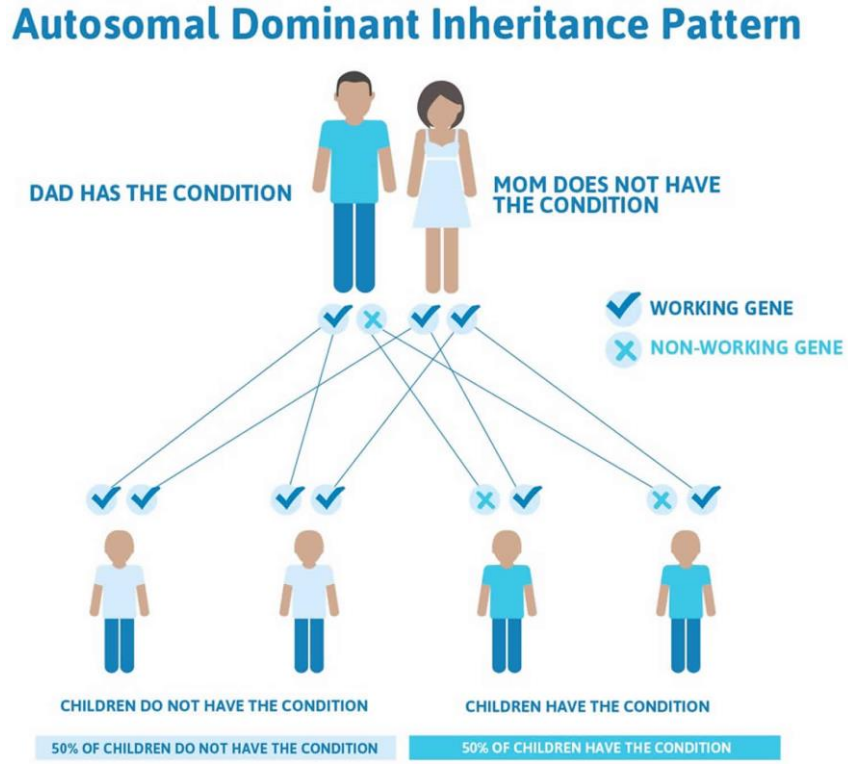
Amnio acid # 1 2 3 4 5 6 7 8 … 200

- "c.598G>A" means at position number 598 in the DNA code (c.DNA), the nucleotide "G" was changed to an "A"
- "p.Glu200Lys" means that amino acid number 200 in the protein chain, the amino acid Glutamic acid was change to Lysine.

Frequently asked questions

What are the chances of having another child with NS?

- Noonan syndrome is inherited in an autosomal dominant manner.
- If a parent has NS, there is a 50% chance of having a child with NS. passing it on *with each pregnancy*
- If the parent does NOT have NS, the chances of having another child with NS very low (less than 1%)
- Rarely, the parent may carry the genetic change in a small percentage of their cells. This is called mosaicism.
  - If the egg or sperm cells carry the genetic change, there is an increased chance of having a child with NS, even if the parent is unaffected.
  - This is difficult to detect, but also very rare.



Can you predict the severity or how the child will be affected?

- Sometimes knowing the genetic variant or size and location of the chromosome abnormality ("genotype") can help predict the clinical features that may be more likely to occur in an individual ("phenotype").

- But there remains significant variability both between and within families/individuals who may carry the same genetic change, so we need to be cautious with making assumptions and predictions.

- Genotype = Genetic cause (deletion, mutation)
- Phenotype = Clinical features of the person (heart problems, learning difficulties, etc)

Are other family members at an increased risk of having a child with NS?

- No, unless other family members also have NS.

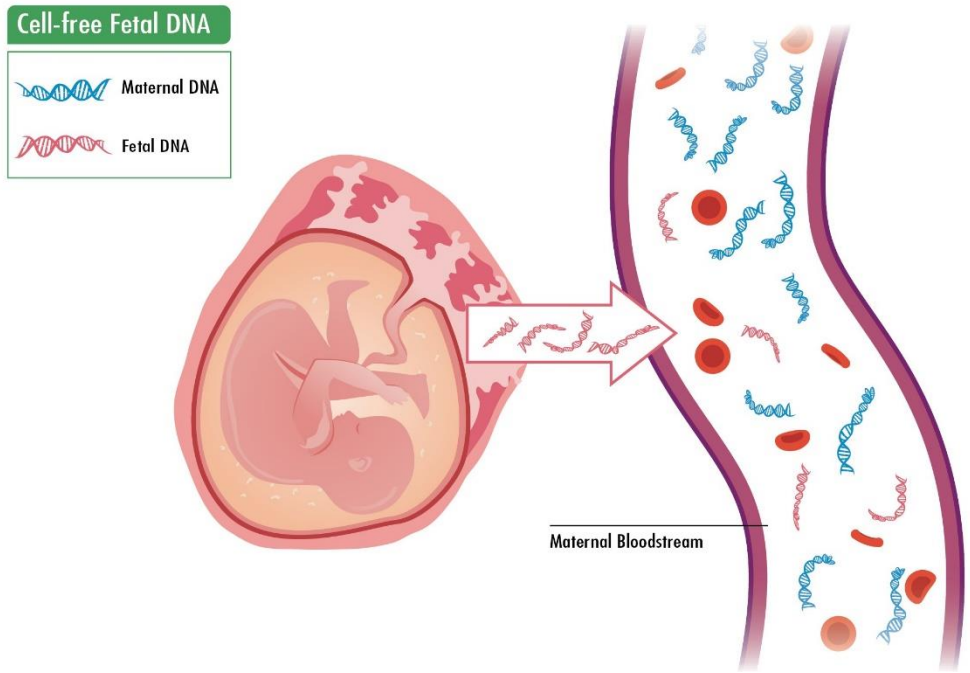
- If so, then the risk to their children would be 50%.

What reproductive options are available in future pregnancies?

- Recommend genetic counseling *before* getting pregnant to discuss options (preconception counseling)
- The recurrence risk is low, but some couples may want to pursue genetic testing.
- In-vitro fertilization (IVF) with preimplantation genetic testing (PGT)
  - This involves the couple going through IVF, testing the embryos for the genetic condition, and implanting the embryos that do not carry the genetic change.
- Prenatal Diagnosis
  - CVS (chorionic villi sampling) –first trimester
  - Amniocentesis – second trimester
- Need to know the specific genetic variant you are testing first!

Non-invasive Prenatal Testing

- NIPT is a blood test that is done on the mother that tests the fetal DNA.
- This is most commonly used to screen for chromosome abnormalities, such as Down syndrome, but can now also screen for conditions such as Noonan syndrome.
- This test is not useful if the MOTHER has Noonan syndrome though since the test cannot differentiate between the maternal DNA and fetal DNA if a variant is detected.
  - It CAN be used if the father has Noonan syndrome or if neither parent has Noonan syndrome.



When in doubt…

- Ask your genetics providers!

- National Society of Genetic Counselors
  - www.nsgc.org

National Society of Genetic Counselors

Thank you!

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Noonan syndrome Walk (2014)