GENETICS 101 & WHOLE GENOME SEQUENCING

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The Magic of a Cure

Dream It.
Hope It.

National Ataxia Foundation
Annual Ataxia Conference
April 1-3, 2016 ★ Orlando, Florida
Hosted by the Southeast Region

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PRESENTER DISCLOSURES

- Vikram Shakkottai
- No relationships to disclose

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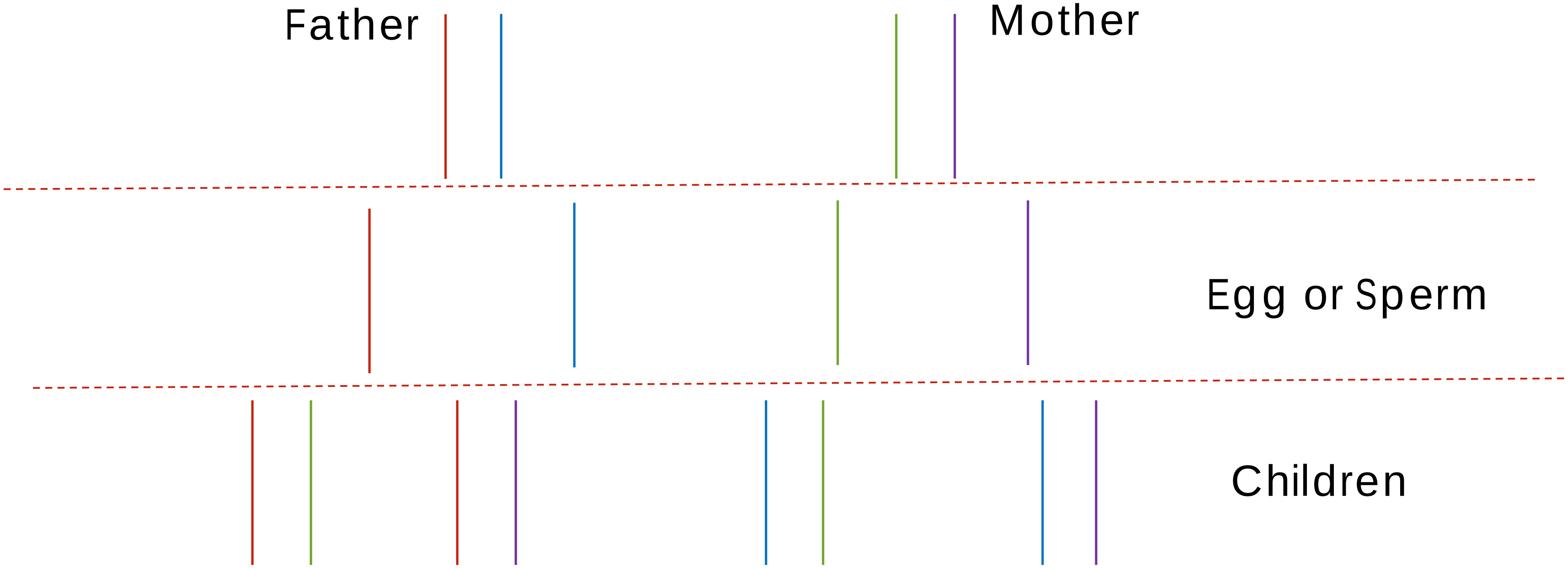
WHAT IS A GENE

- A gene is the basic physical and functional unit of heredity.
- Genes, which are made up of DNA, act as instructions to make molecules called proteins.
- Every person has two copies or alleles of each gene, one inherited from each parent.

INHERITANCE



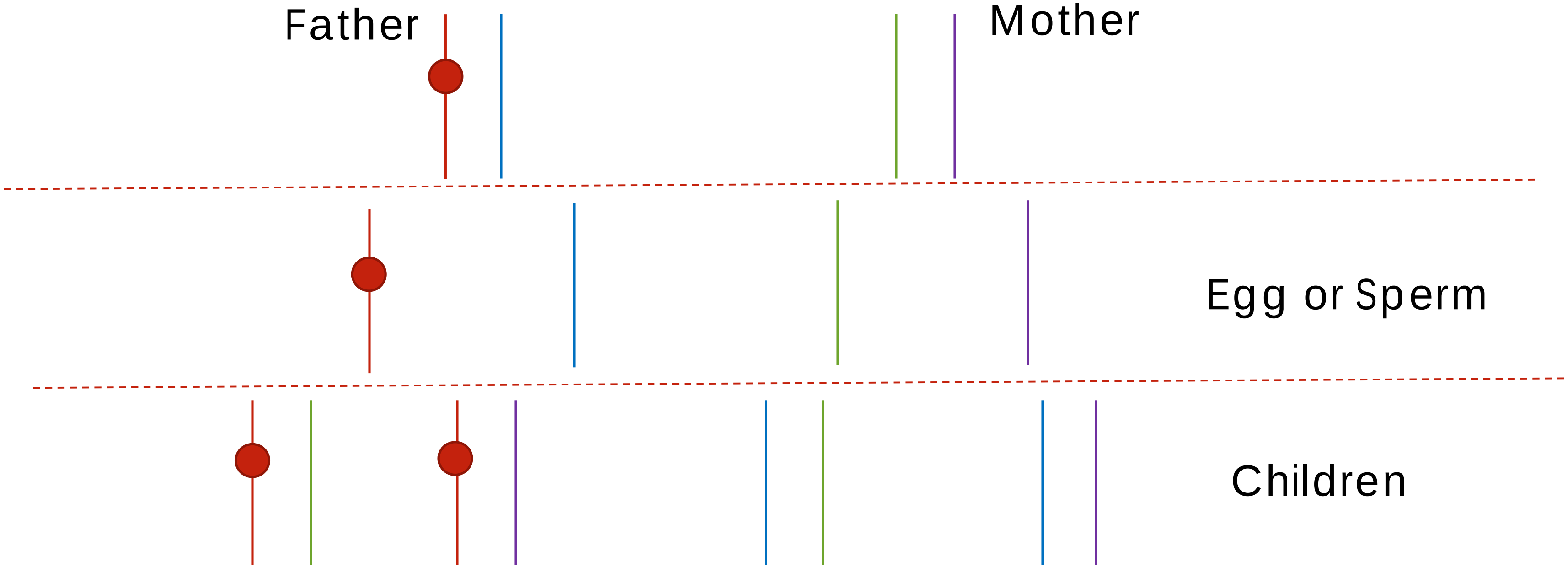
Father

Mother

Egg or Sperm

Children

DOMINANT DISEASE



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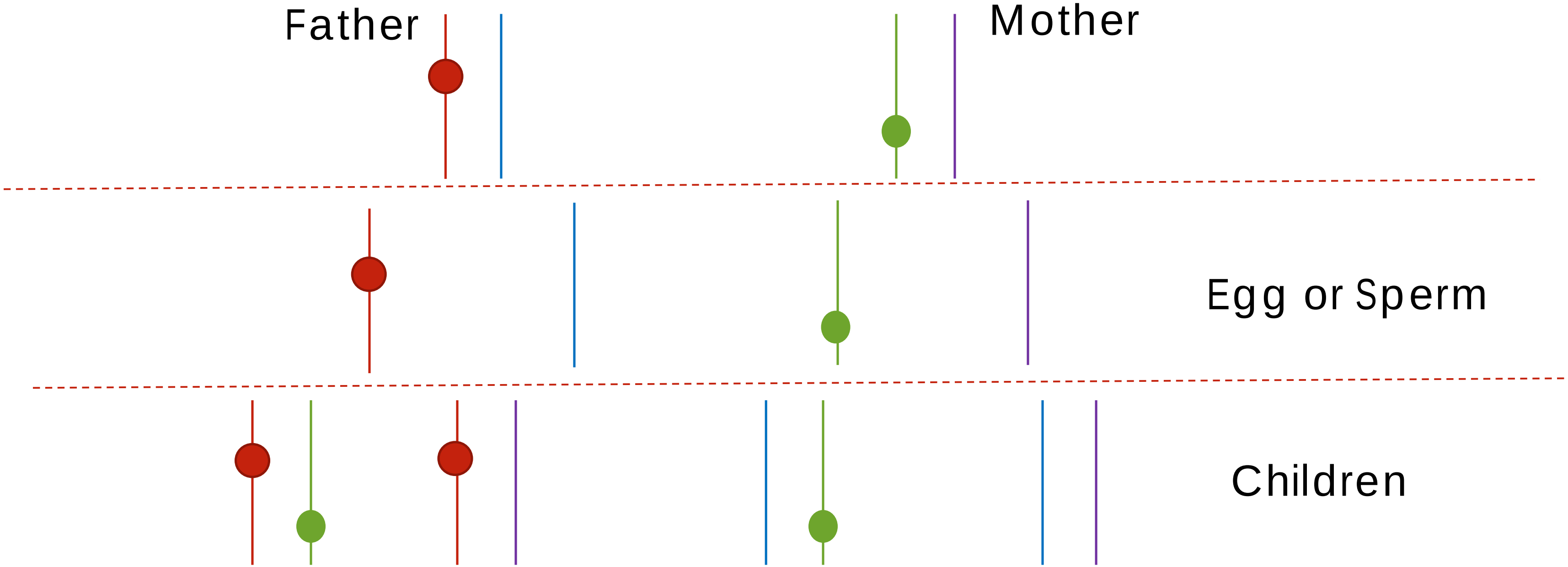
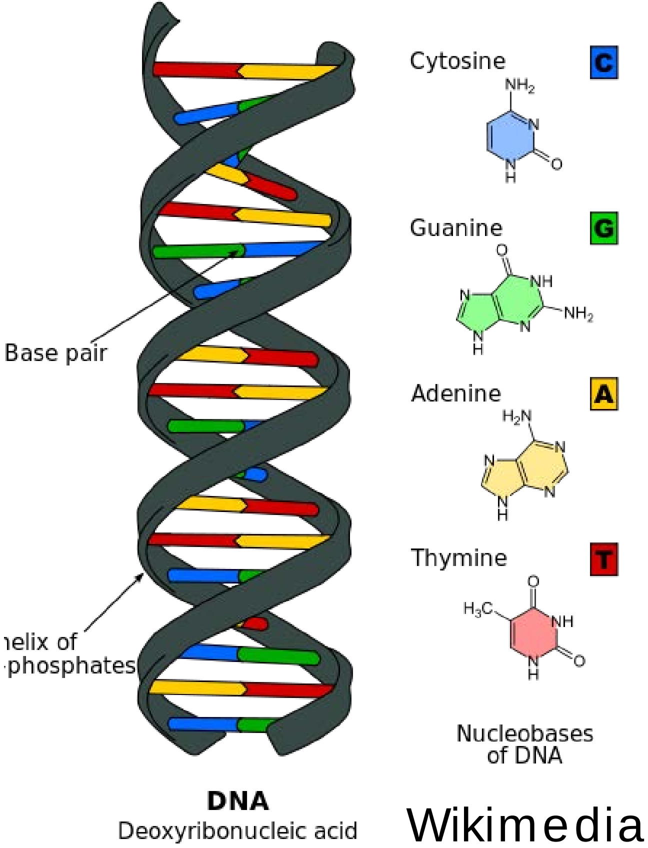
RECESSIVE DISEASE



WHAT IS A GENE MADE OF?

- DNA
- Letters-A,T,G,C
- 3 letters made a code for an amino acid



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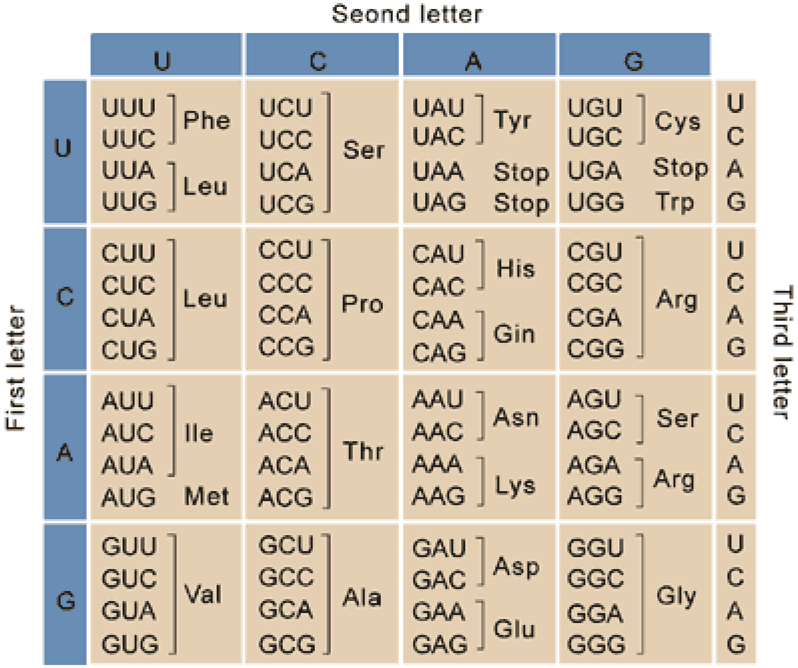
THE TRIPLET CODE

- A,T(U),G,C
- Examples: AAA-Lysine
- CAG-Glutamine



Seond letter

First letter

Third letter

	U		C		A		G		
U	UUU UUC UUA UUG	Phe Leu	UCU UCC UCA UCG	Ser	UAU UAC UAA UAG	Tyr Stop Stop	UGU UGC UGA UGG	Cys Stop Trp	U C A G
C	CUU CUC CUA CUG	Leu	CCU CCC CCA CCG	Pro	CAU CAC CAA CAG	His Gin	CGU CGC CGA CGG	Arg	U C A G
A	AUU AUC AUA AUG	Ile Met	ACU ACC ACA ACG	Thr	AAU AAC AAA AAG	Asn Lys	AGU AGC AGA AGG	Ser Arg	U C A G
G	GUU GUC GUA GUG	Val	GCU GCC GCA GCG	Ala	GAU GAC GAA GAG	Asp Glu	GGU GGC GGA GGG	Gly	U C A G

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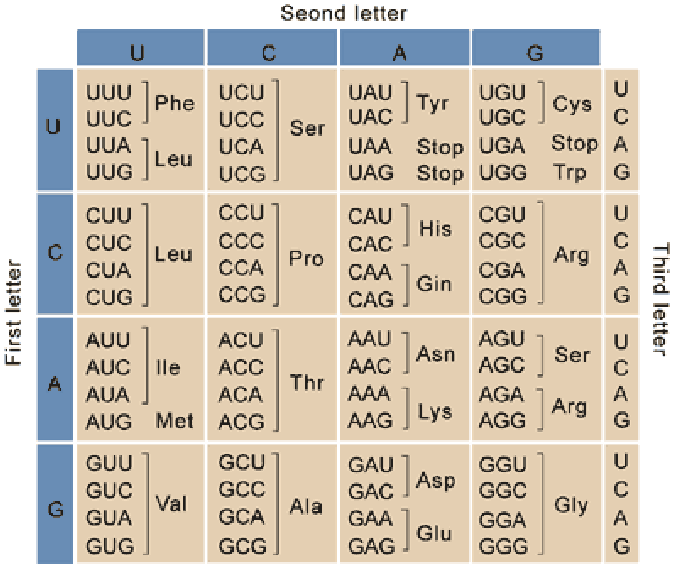
MUTATION

- A defect in a gene
- Can be inherited or arise *de novo*

MUTATION

- AUGCAGAUG
- AUG CAG AUG
- Methionine-Glutamine-Methionine

- AUGACAGAUG
- AUG AAG AUG
- Methionine-Lysine-Methionine



Seond letter

First letter	U	C	A	G	Third letter
U	UUU Phe UUC UUA Leu UUG	UCU Ser UCC UCA UCG	UAU Tyr UAC UAA Stop UAG Stop	UGU Cys UGC UGA Stop UGG Trp	U C A G
C	CUU Leu CUC CUA CUG	CCU Pro CCC CCA CCG	CAU His CAC CAA Gin CAG	CGU Arg CGC CGA CGG	U C A G
A	AUU Ile AUC AUA AUG Met	ACU Thr ACC ACA ACG	AAU Asn AAC AAA Lys AAG	AGU Ser AGC AGA Arg AGG	U C A G
G	GUU Val GUC GUA GUG	GCU Ala GCC GCA GCG	GAU Asp GAC GAA Glu GAG	GGU Gly GGC GGA GGG	U C A G

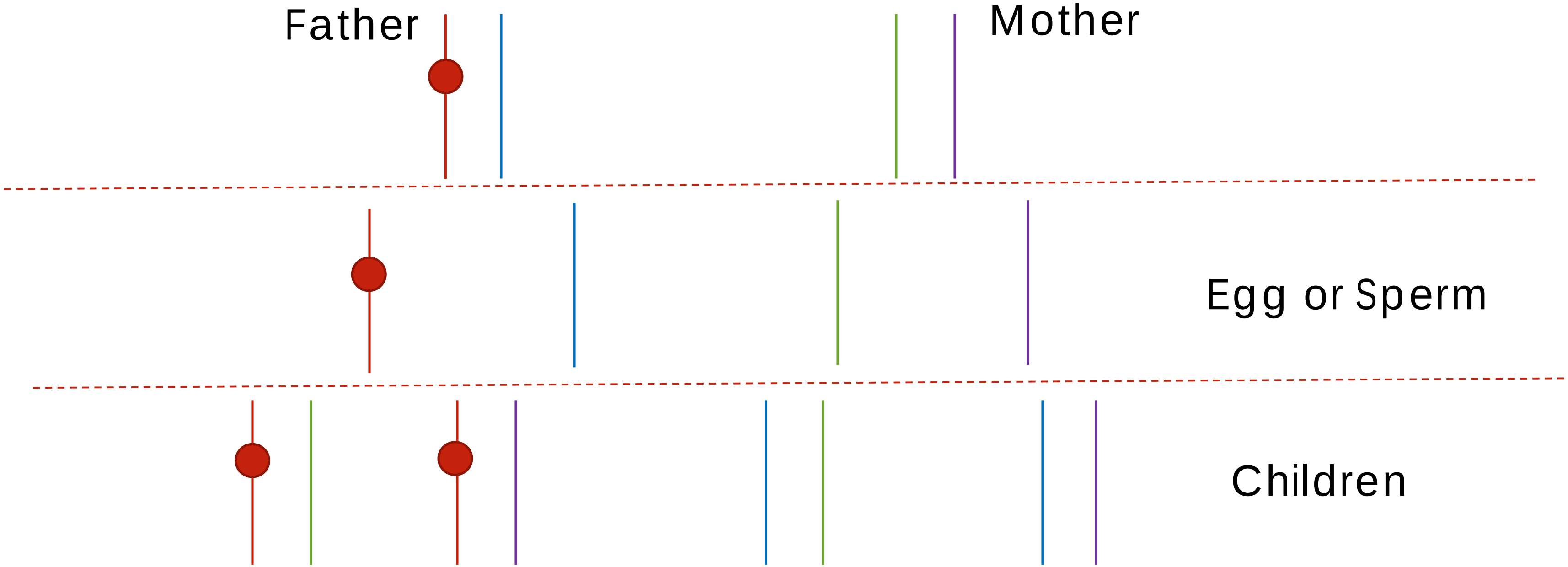
REPEAT DISORDERS

- Stretch of repeating sequence
- For example CAG CAG CAG CAG 4 repeats
- Mutation here-CAG CAG CAG CAG CAG CAG 6 repeats

SCA

- Dominantly inherited ataxia
- Numbering is based on order of discovery
- For example, SCA1 was the first to be identified

DOMINANT DISEASE



Father

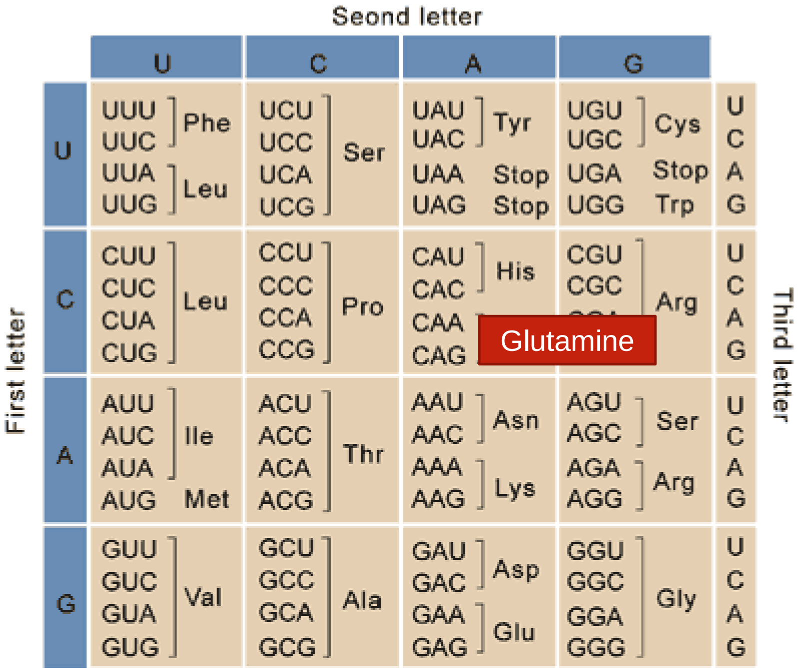
Mother

Egg or Sperm

Children

SCA

- Polyglutamine or
  CAG expansions
- SCA1, 2, 3, 6, 7, 17



Seond letter

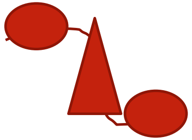
First letter	U	C	A	G	Third letter
U	UUU Phe UUC Phe UUA Leu UUG Leu	UCU Ser UCC Ser UCA Ser UCG Ser	UAU Tyr UAC Tyr UAA Stop UAG Stop	UGU Cys UGC Cys UGA Stop UGG Trp	U C A G
C	CUU Leu CUC Leu CUA Leu CUG Leu	CCU Pro CCC Pro CCA Pro CCG Pro	CAU His CAC His CAA Glutamine CAG Glutamine	CGU Arg CGC Arg CGA Arg Arg	U C A G
A	AUU Ile AUC Ile AUA Ile AUG Met	ACU Thr ACC Thr ACA Thr ACG Thr	AAU Asn AAC Asn AAA Lys AAG Lys	AGU Ser AGC Ser AGA Arg AGG Arg	U C A G
G	GUU Val GUC Val GUA Val GUG Val	GCU Ala GCC Ala GCA Ala GCG Ala	GAU Asp GAC Asp GAA Glu GAG Glu	GGU Gly GGC Gly GGA Gly GGG Gly	U C A G

REPEATS

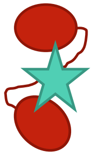
- Toxicity if repeats are too long

DOMINANT DISEASE

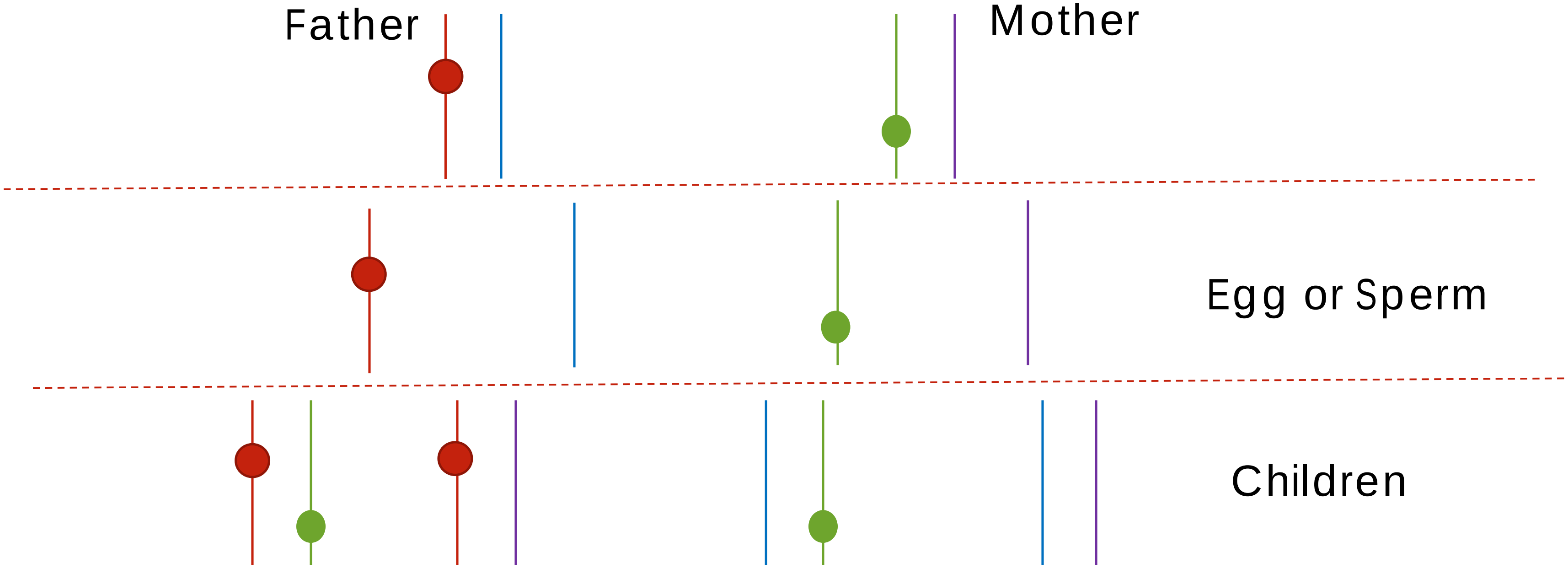
- Point mutations
  - AUGCAGAUG
  - AUG CAG AUG
  - Methionine-Glutamine-Methionine



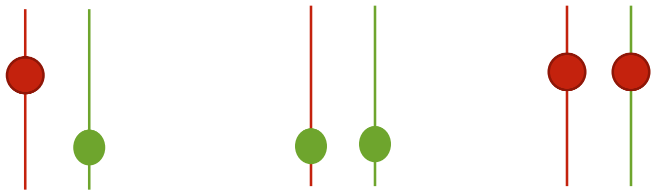
  - AUGACAGAUG
  - AUG AAG AUG
  - Methionine-Lysine-Methionine



RECESSIVE DISEASE



RECESSIVE DISEASE

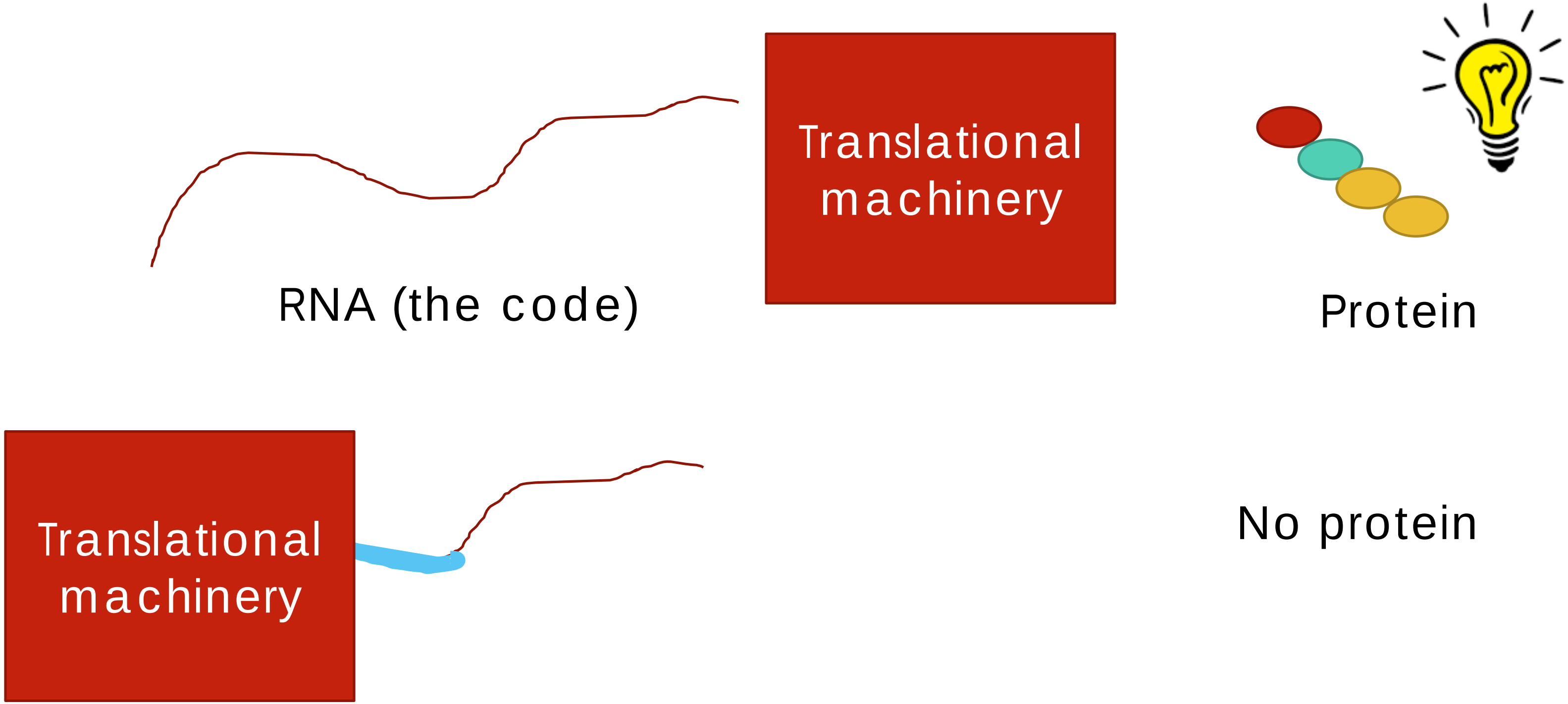


2 mutations-one on each copy of the gene

RECESSIVE ATAXIA

- Friedreich ataxia
- Also a repeat disorder-but non coding

NON-CODING REPEATS



Translational machinery

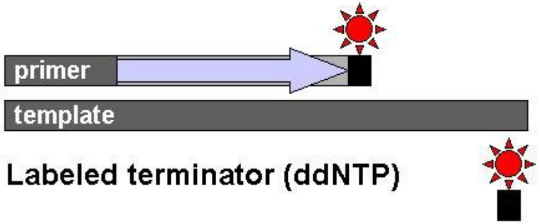
RNA (the code)

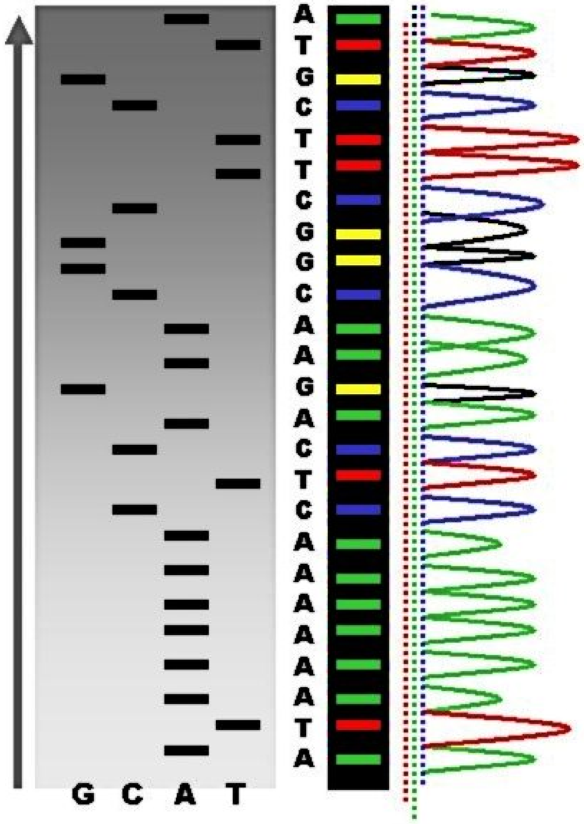
Protein

Translational machinery

No protein

GENE SEQUENCING





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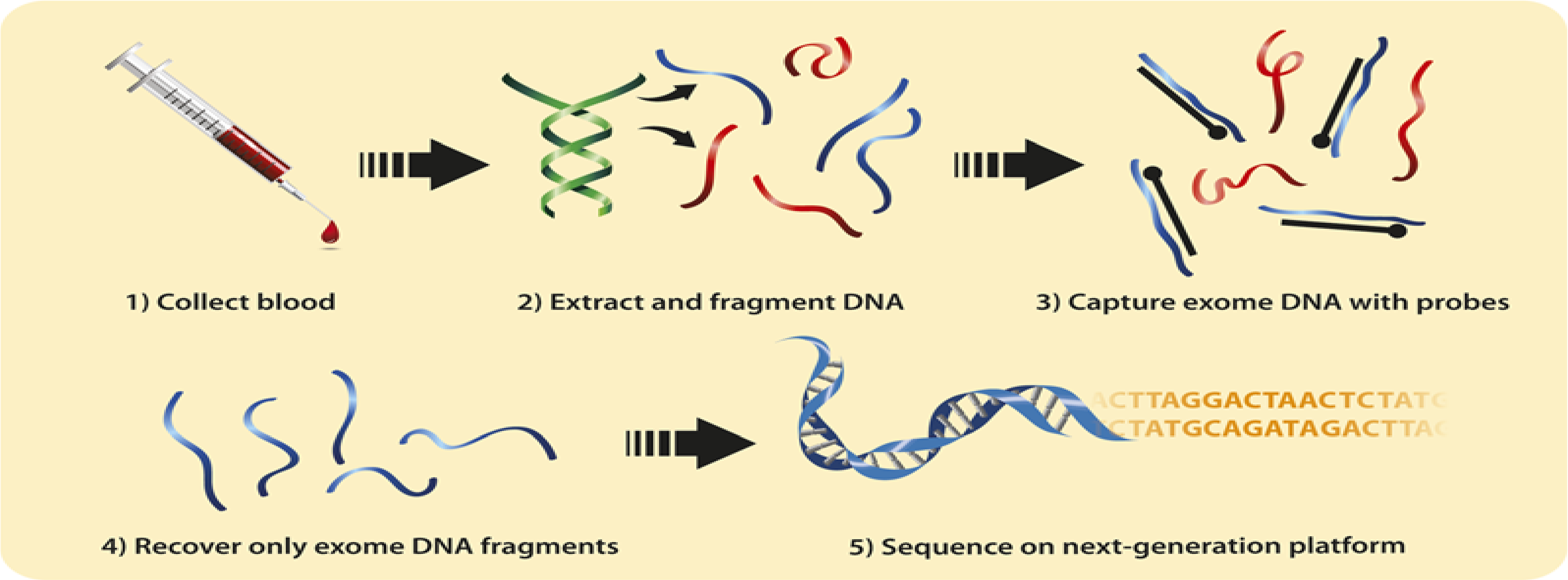
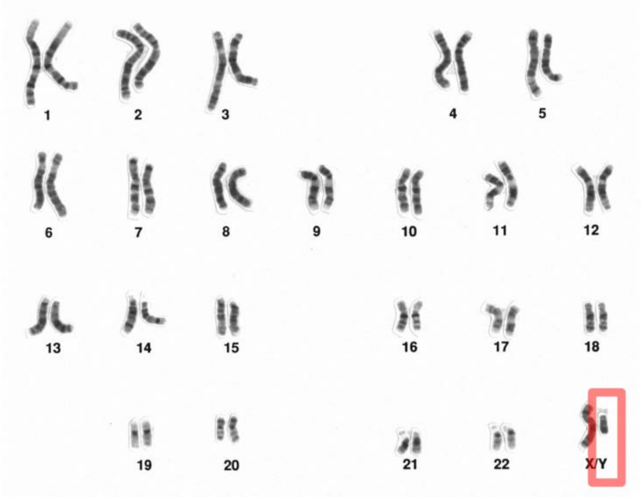
WHOLE EXOME SEQUENCING



Paria N, Copley LA, Herring JA, Kim HK, Richards BS, Sucato DJ, Wise CA, Rios JJ. (2013) **Whole-exome sequencing: discovering genetic causes of orthopaedic disorders.** *J Bone Joint Surg Am* 95(23), e1851-8

EXOME

- Simplified definition: The part of the genome that codes for protein



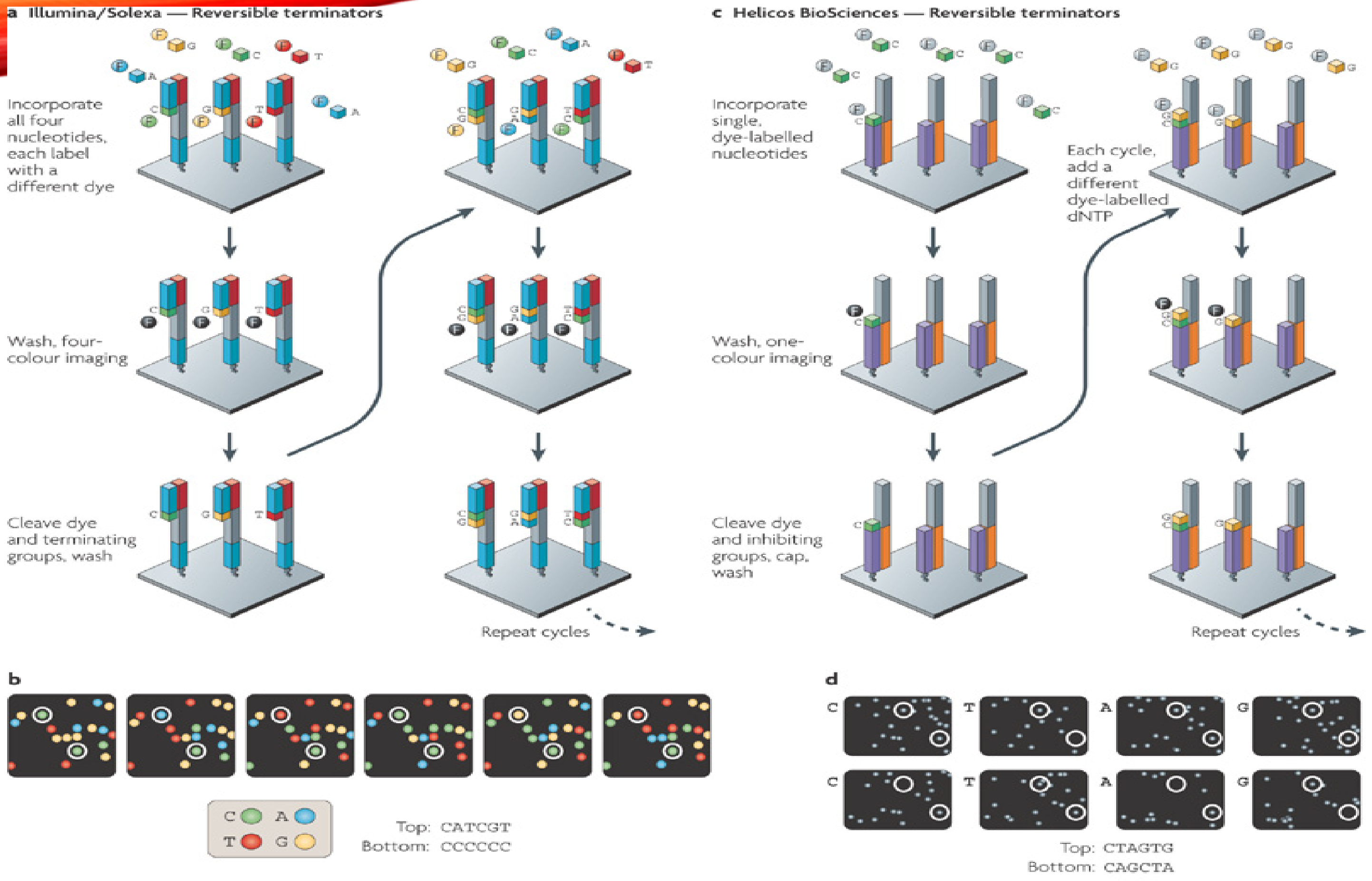
1% of the genome

WHY EXOME SEQUENCING?

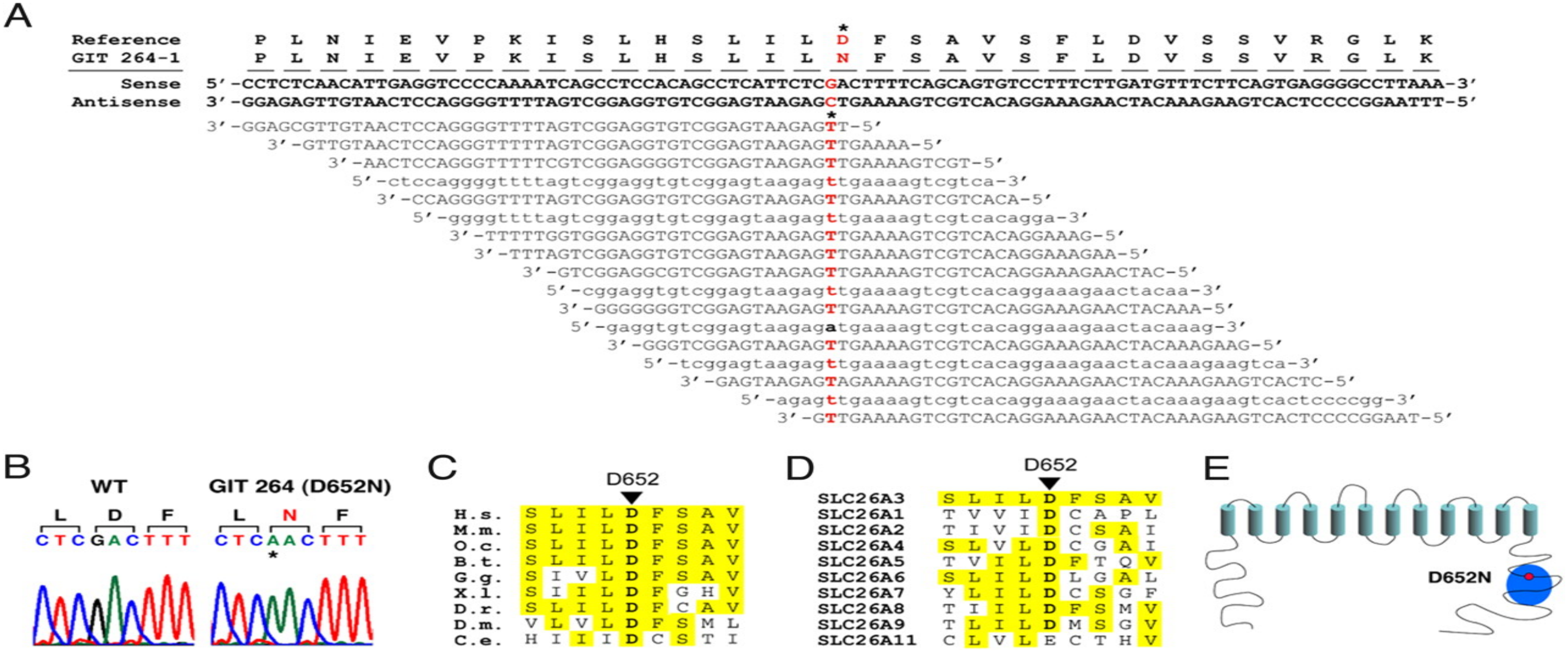
- A way to survey all the coding genes at the same time
- If you don’t know where to look, look at it all
- Over 300 gene mutations can cause ataxia
- Can be cost effective

NGS PLATFORMS

- 454 Life Sciences
- Illumina
- Applied BioSystems
- Helicos Biosciences
- Danaher Motion
- Oxford Nanopore Technologies
- Pacific Biosciences.

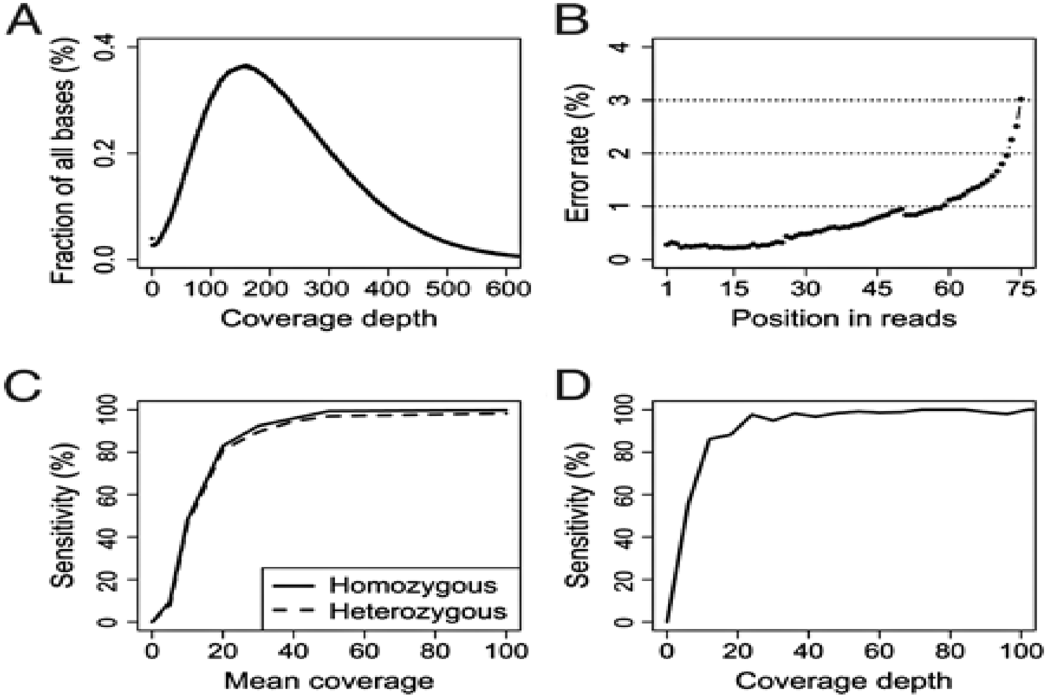


Nature Reviews | Genetics



Choi et al. Genetic diagnosis by whole exome capture and massively parallel DNA sequencing. PNAS 2009

DEPTH OF COVERAGE



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QUESTIONS?