

Genetic Testing for TSC in 2010

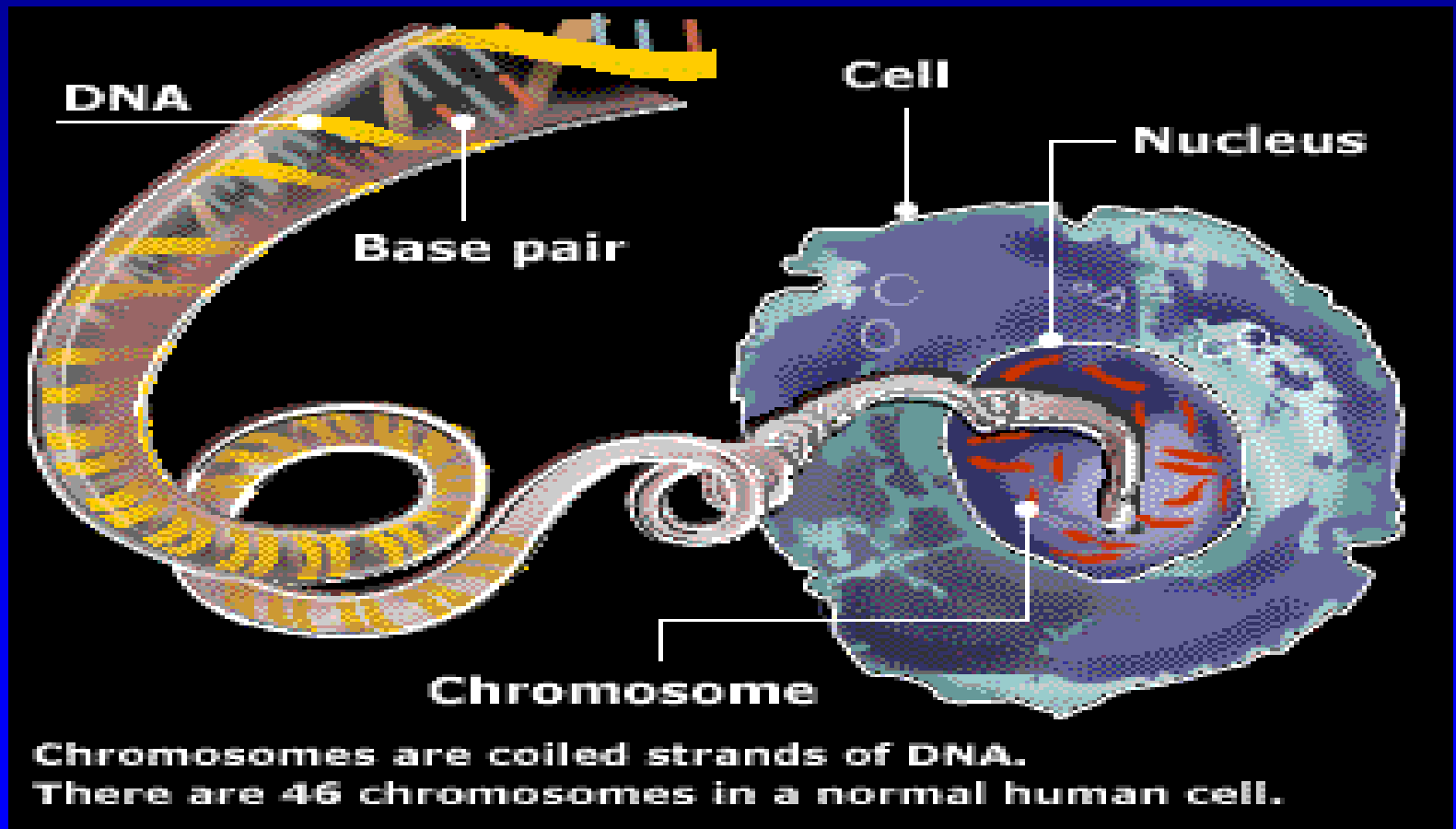
Hope Northrup, MD
The University of Texas Medical
School at Houston

Genetics 101: The Basics

- Our bodies contain millions and millions of cells
- The instructions within each cell are the **genes**
- Human beings have approximately 25,000 different genes
- The genes are in packages called **chromosomes**

Analogy: If the human genome is the “book of life” then the chromosomes are the chapters, and the genes are the paragraphs

Genetics 101: The Basics



Genetics 101: Genes

- Genes have DNA that is divided into specific sections: exons and introns
- Exons are the important sections of the gene, producing the gene's product (proteins)
- Introns are the sections “in between” the exons, they do not produce proteins

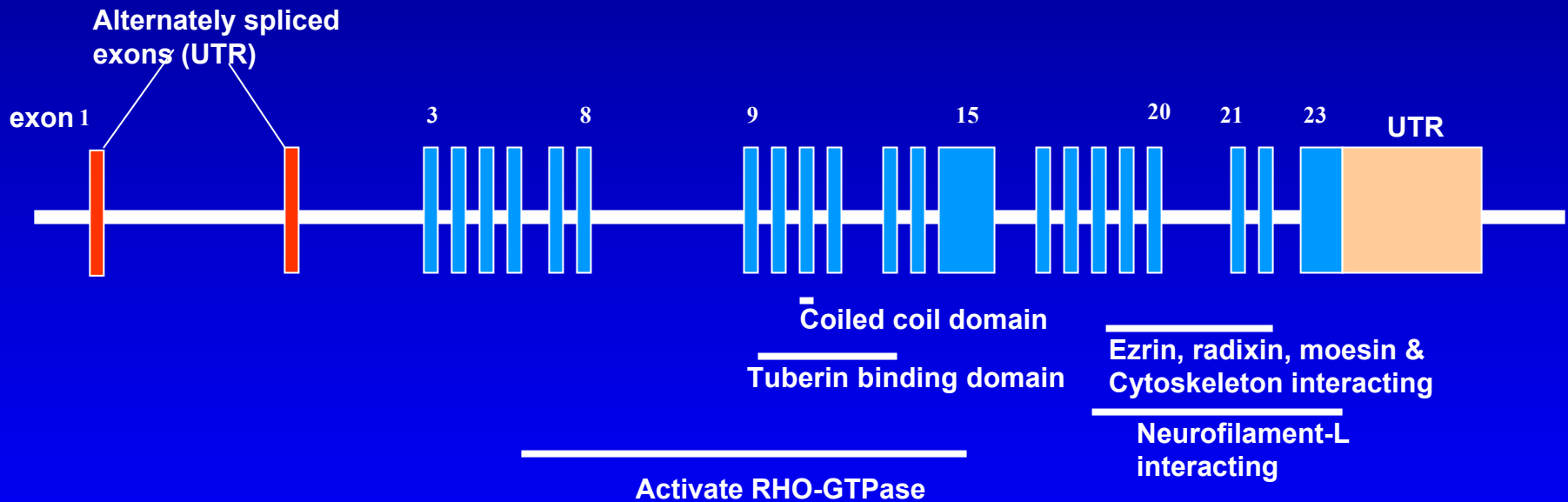
Analogy: Exons are the sentences, introns are sentences that the “editor” removes from the final version

Genetics 101: DNA

- Genes are made of DNA
- The DNA has building blocks called “bases” (A=adenine, T=thymine, C=cytosine, G=guanine)
- Three bases make up one “codon”
- These codons specify amino acids, directing the production of proteins
- Three special codons “stop” the production of proteins

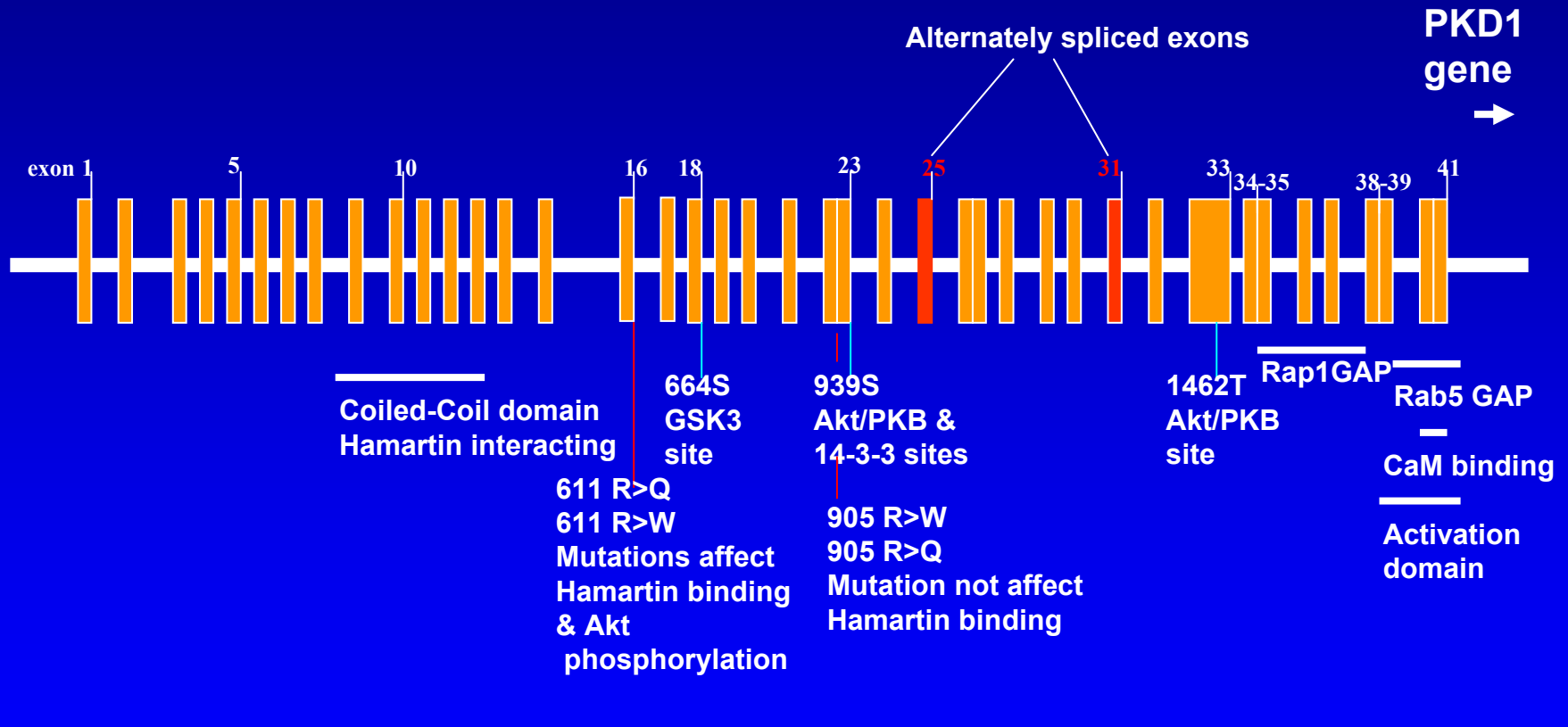
Analogy: Bases are the letters and codons are the words

TSC1 Gene Structure and Functions



Ref. Lamb et al. 2000. Nat. Cell. Biol. 2:281-6. Hodges et al. 2001. HMG 10:2899-2905. Haddad et a. 2002. JBC 277:44180-6.

TSC2 Gene Structure and Functions



C-terminal fragment of tuberlin can rescue Eker rat mutation

An Overview of TSC Genetics

- Tuberous sclerosis complex (TSC) is a dominant genetic condition
- TSC can occur as a result of an inherited *TSC1* or *TSC2* gene mutation
- TSC can also occur as a result of a sporadic or new *TSC1* or *TSC2* gene mutation

Genetics 101: Mutations

The Easy Ones

- A **deletion** mutation is when a piece of the gene is missing. A deletion may include parts of exons and/or introns, it may be large or small, and may include the entire gene

Analogy:

Our dog ate the cat toy



Deletion

Our dog ate the cat



Four Biotech Park • 377 Plantation Street
Worcester, Massachusetts 01605
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Patient

Requesting Physician

Hope Northrup, MD

Date of Birth

06/18/1994

Sex

M

Social Security Number

Report to

Hope Northrup, MD

Specimen Type

Whole Blood

Address

6431 Fannin, Msb 3.

Test Category

Not Available

Houston, TX 77030

Test Requested

Complete Tuberous Sclerosis Evaluation

Additional Reports to:

Interpretation

POSITIVE

This individual possesses a DNA variant in the *TSC1* and/or *TSC2* gene(s) that is **predicted** to be a **disease-associated mutation**. This test result is consistent with a diagnosis of, or a predisposition to developing, Tuberous Sclerosis.¹⁻⁹ Please refer to the Technical Results and Comments sections of this report for further information.

Technical Results

TSC2 Deletion Result: Deletion of exons detected

Exons Deleted: 1-14

TSC1 DNA Sequence Variants:

No abnormal DNA sequence variants identified.

TSC2 DNA Sequence Variants:

DNA variant 1: Transversion C > A

Nucleotide position: 2476

Codon position: 826

Amino acid change: Leucine > Methionine

Variant type: Variant of unknown clinical significance

Genetics 101: Mutations

The Easy Ones

- An **insertion** mutation is when there are extra bases present. There may be an insertion into an exon and/or intron, and may also be large or small

Analogy:

Our dog ate the cat toy

Insertion red

Our dog ate red the
cat toy



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Patient			Requesting Physician
Date of Birth	Sex	Social Security Number	Report in
12/28/2009	M		Mayo Medical Labo
Specimen Type	Address		
Whole Blood	P.O. Box 4100		
Test Category	Rochester, MN 5590		
Not Available			
Test Requested	Additional Reports for		
Complete Tuberous Sclerosis Evaluation			

Interpretation

POSITIVE

This individual possesses a DNA variant in the *TSC1* and/or *TSC2* gene(s) that is predicted to be a disease-associated mutation. This test result is consistent with a diagnosis of, or a predisposition to developing, Tuberous Sclerosis.¹⁻⁹ Please refer to the Technical Results and Comments sections of this report for further information.

Technical Results

TSC2 Deletion Result: Inconclusive

TSC1 DNA Sequence Variants:
No abnormal DNA sequence variants identified.

TSC2 DNA Sequence Variants:

DNA variant 1: 1 bp Duplication of C
Nucleotide position: 4351_4352
Codon position: 1451
Amino acid change: Frame Shift
Variant type: Predicted disease-associated mutation

Genetics 101: Mutations

The Easy Ones

- A **missense** mutation is when one base has been substituted for another base (an “A” instead of a “T”) causing the codon to make a different amino acid

Analogy:

Our dog ate the cat toy



Substitute

b

Our dog ate the cat
boy

Patient mutation report

Patient's name: _____

Putative Mutation:

TSC1 gene--exon ___X___

nucleotide changes ___X___

forward primer _____X_____

reverse primer _____X_____

product size _____X_____

sequencing primers _____X_____

TSC2 gene--exon ___40___

nucleotide changes (amino acid change)___5228G->A (1743R->Q)___

forward primer TGG CCA AGA TCG TGT CTG AC

reverse primer TGA CAG GCA ATA CCG TCC AA

product size _____585 bp_____

sequencing primers___ TGG GTC CAG GCG TGA GCT G

Genetics 101: Mutations

The Easy Ones

- A **nonsense** mutation is when one base has been substituted for another, changing the codon from one that makes an amino acid, to one that tells the gene to “stop” making the protein before it’s finished (protein truncation)

Analogy:

Our dog ate the cat toy

↑
Nonsense

Our dog ate the

57452100

Athena Diagnostics, Inc.

Genetic Testing
Pharmaceutical Companies

Four Birkbeck Park • 331 Pleasanton Street

Woburn, Massachusetts 01803

(800) 394-4493 • (508) 756-2866

Patient			Reporting Physician
Date of Birth 12/18/2003	Sex M	Social Security Number 000000000	Report to American Medical L
Specimen Type Whole Blood			Address Attn: JACK C. Brigg
Test Category Not Available			14225 Newbrook Dr
Test Requested Complete Tuberos Sclerosis Evaluation			Chantilly, VA 20151
			Additional Reports to:

Interpretation

This individual possesses a DNA sequence variant in the *TSC2* gene that is a previously reported disease-associated mutation. Therefore, it is predicted that this individual is affected with, or predisposed to developing, tuberous sclerosis. Please refer to the Technical Results and Comments sections of this report for further information.

Technical Results

TSC2 DNA Sequence Variants:

DNA variant 1:	Transition C > T
Nucleotide position:	1372
Codon position:	458
Amino Acid change:	Arginine > OPA (Stop codon)
Variant type:	Disease-associated mutation

Comments

Analysis of this individual's *TSC1* and *TSC2* genes identified a DNA sequence variant in the *TSC2* gene which has been reported in the literature to be associated with tuberous sclerosis (Disease-associated mutation). Therefore, this individual is likely to be affected with or predisposed to developing, tuberous sclerosis. In addition to this result, other DNA sequence variants, differing from the *TSC1* and *TSC2* reference sequences, may have been revealed by this analysis. However, in the context of the results reported here, the presence of additional sequence variants is generally of reduced significance. Please consult the

How Genes Work

Gene's message: The color gray is beautiful.

Mutations that interfere with the message.

- Large deletion: The gray is beautiful.
- Large insertion: The color gray **gray** is beautiful.
- Protein truncation: The color gray.



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Worcester, Massachusetts 01605
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Patient			Requesting Physician	Hope Northrup, MD
Date of Birth	Sex	Social Security Number	Report to	Hope Northrup, MD
01/05/2009	M		Address	Uth Medical School
Specimen Type				P.O.
Whole Blood				Houston, TX 77225
Test Category			Additional Reports to:	Hope Northrup, MD
Not Available				
Test Requested				
Complete Tuberous Sclerosis Evaluation				

Interpretation

This individual possesses a DNA variant or combination of variants in the *TSC1* and/or *TSC2* gene(s), whose significance is unknown **amino acid variant of unknown significance**). The significance of this new variant may be clarified by careful reconciliation of this molecular data with this patient's clinical symptoms and family member testing if appropriate. Please refer to the Technical Results and Comments sections of this report for further information.

Technical Results

TSC2 Deletion Result: No deletion detected

TSC1 DNA Sequence Variants:
No abnormal DNA sequence variants identified.

TSC2 DNA Sequence Variants:

DNA variant 1: Transition G > A
Nucleotide position: 5228
Codon position: 1743
Amino acid change: Arginine > Glutamine
Variant type: Variant of unknown clinical significance

Genetics 101: Polymorphisms

- Polymorphisms are changes that don't cause a problem with the protein
- Polymorphisms cause the bases to change, without affecting the production of the protein
- Polymorphisms can fool us!

Analogy:

Our dog ate the cat toy



bad (Polymorphism)

Our bad dog ate the
cat toy

Genetics 101: Polymorphisms

The word GRAY can also be spelled GREY
The meaning is unchanged, therefore it is a
polymorphism

An example of a mutation of GRAY would be
GROY (missense mutation)

It no longer means Gray, therefore it is a
mutation

With genes, we cannot always tell if a change is
a polymorphism or a mutation

How Genes Work

Gene's message: The color gray is beautiful.

More subtle mutations

- Small deletion: The coor gray is beautiful.
- Small insertion: The coldor gray is beautiful.
- Missense: The color gray is beautiful.

Polymorphisms

- The colour gray is beautiful.
- The color grey is beautiful.

Athena Diagnostics, Inc.Formerly Genica
Pharmaceuticals CorporationFour Biotech Park * 377 Plantation Street
Worcester, Massachusetts 01605

(800) 394-4493 * (508) 756-2886

Patient

Date of Birth
11/19/2005Sex
F

Social Security Number

Specimen Type
Whole BloodTest Category
Not AvailableTest Required
Complete Tuberous Sclerosis EvaluationRequesting Physician
Hope NorthrupReport to
Hope NorthrupAddress
6431 Fannin, Msb 3

Houston, TX 77030

Additional Reports to:

Interpretation**NEGATIVE**

Analysis of this individual's *TSC1* and *TSC2* genes did not identify any abnormal DNA variants in the coding region and intron/exon junctions for this gene and is thus interpreted to be **normal**. Therefore, this individual is unlikely to be affected with, or predisposed to developing, Tuberous Sclerosis due to mutations in the *TSC1* and *TSC2* genes. However, the clinical symptoms of your patient could likely be due to sequence alterations in genes not included in this assay. Please refer to the Comments section of this report for further information.

Technical Results

TSC2 Deletion Result: No deletion detected

No DNA sequence variants have been detected.

No Mutation Identified (NMI)

- Mutation that we can't identify
- Mosaicism
- Additional gene(s) that cause TSC

Genetics 101: Genetic Testing for TSC

Athena Diagnostics, Inc

- Complete TSC Evaluation-----\$4,140
- TSC 1 Sequencing Test-----\$1,730
- TSC 2 Sequencing Test-----\$2,345
- TSC Familial Evaluation-----\$690
- TSC deletion Test-----\$660

Genetics 101: Genetic Testing for TSC

- sequencing all exons of *TSC1* and *TSC2* genes
- testing for large deletions in *TSC2* gene
- detects mutation in 85% of cases when patient has a Definite diagnosis

Genetics 101: Genetic Testing for TSC

- In persons with a definite diagnosis of TSC, mutations can be found in approximately 85%
- The remaining persons with a definite diagnosis may have mutations in *TSC1* or *TSC2* we cannot find, may have mosaicism for a mutation or there may be additional TSC-causing genes

Questions?