THE UNIVERSITY OF CHICAGO MEDICINE & BIOLOGICAL SCIENCES

"Abnormal thyroid tests and NGS: a nightmare to the clinician?"

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ENDORAMA
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Dr. Refetoff does not have any relevant financial relationships with any commercial interests.

Abnormal thyroid tests and NGS: a nightmare to the clinician?

Learning Objectives:

1. How to interpret VUS (variants of unknown significance)
2. Clinical decision making in face of VUS

Endorama

Sept 24,2020

Case History:

A 49-year-old woman diagnosed as having Hashimoto's thyroiditis in her 20s, treated with levothyroxine (L-T4).
Information regarding TFTs prior to starting L-T4 treatment is not available

Thyroid US showed diffusely atrophic gland with heterogeneous architecture consistent with Hashimoto's thyroiditis.

During L-T4 treatment, she had normal to slightly elevated FT4 and TSH levels and had a mixtur of non specific hypothyroid and hyperthyroid symptoms.

Fist encounter (referral):

Her Physician, Chrysoula Dosiou,
Endocrinologist at Stanford University School of Medicine

wrote:

I am seeing a patient for whom I have a high index of suspicion for thyroid hormone resistance syndrome. After dynamic testing with high dose T3 (100 mcg for 5 days) resulting in TSH suppression (from 1.98 to 0.05), I have sent her serum to test for TRbeta mutations. Below is the report I received from Quest. Could you please share your thoughts re whether I can do additional testing to identify if this is a clinically relevant mutation or if you have observed this mutation before, as a cause of thyroid hormone resistance? Thank you in advance!

Verbatim extracts of the genetic report:

INTERPRETATION: DNA testing indicates that this individual is positive for one copy of the c.1154G>A (p.Gly385Glu) variant in exon 10 of the THRB gene. This variant has not been reported in online databases, and to the best of our knowledge, has not been described in the published literature. ….. bioinformatics tools (e.g. SIFT, PolyPhen-2, Mutation Taster and Align-GVGD) …..yielded predictions that this variant is deleterious and neutral. ….. we are unable to determine the clinical significance of the c.1154G>A (p.Gly385Glu) variant.

a VUS

Suggestions

Should have the referring physician sent for THRB sequencing?
What should I do? Answer? Recommend?

12/2000 TSH 2.41, TT4 **14.1** (upper normal 12.5)

4/2001 TSH 2.28, TT4 **13.1** (upper normal 12.5), on LT4

8/2001 TSH 1.95, TT4 **15.2** (upper normal 12.5) on LT4 125 mcg daily

11/2001 TSH **5.54**, TT4 **13.7**

3/2002 TSH **7.86**, TT4 **13.3**; increase Synthroid to 150 mcg

6/2002 TSH 2.49, FT4 1.8 (upper normal 1.8), TT3 160

8/2002 TSH 0.64, TT4 **15.8**

8/2003 TSH 1.41, TT4 12.7

10/2004 TSH 4.17, FT4 2.2 (upper normal 1.8)

11/2004 TSH 1.56, TT4 **14.1** -- decrease Synthroid to 137 mcg

4/2005 TSH 3.65, FT4 1.8

Additional Laboratory Information:

TPO anytobodies, Pos

T4 antibodies, Neg

Heterophile antibodies, Neg

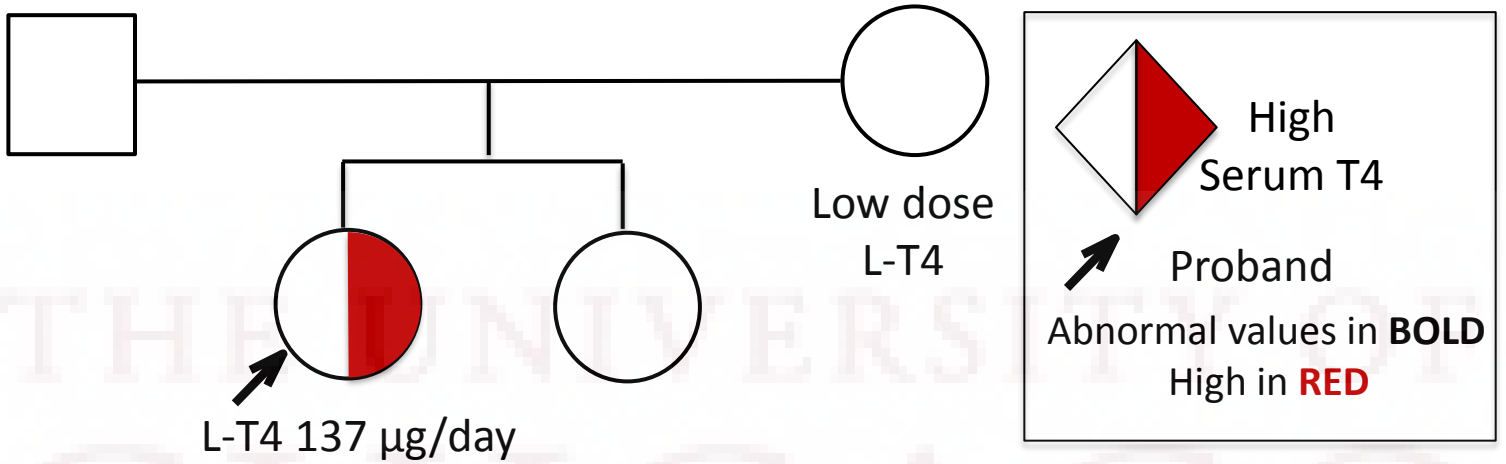
Pituitary MRI, 3mm lesion (likely microadenoma)

Alpha subunit, 0.3 ng/ml

Suggestions

What Next?

Family Msih



Saple #	R6977	R6844	R6978	R6976	
Age (y)	81	49	48	73	**Normal range**
TT4 (µg/dL)	7.7	**12.6**	7.9	10.5	5 - 12
TT3 (ng/dL)	100	101	114	120	80 - 190
TrT3 (ng/dL)	27.6	**61.8**	29.9	**46.9**	16 - 36
FT4I	7.3	**11.2**	8.4	10.2	6 - 10.5
TSH (µIU/mL)	**5.1**	**10.5**	2.7	3.2	0.4 - 3.6
TG (ng/mL)	18	3	**78**	33	2 - 38
TGab/TPOab	<0.4 / <0.4	<0.4 / **10.3**	<0.4 / <0.4	<0.4 / <0.4	<0.4 / <0.4

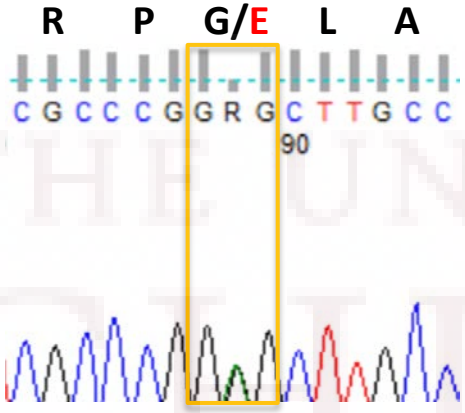
Sequencing identified a mutation in exon 10 of THRB gene

R6844 proband

G**G**G → G**A**G

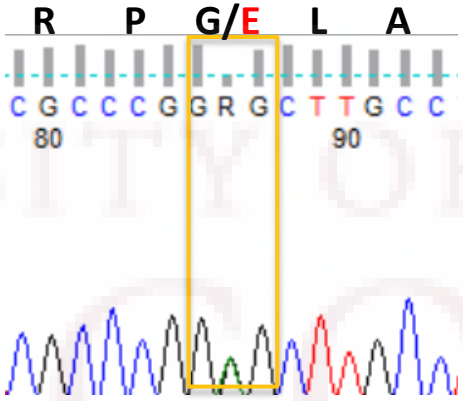
Gly → Glu

G385E



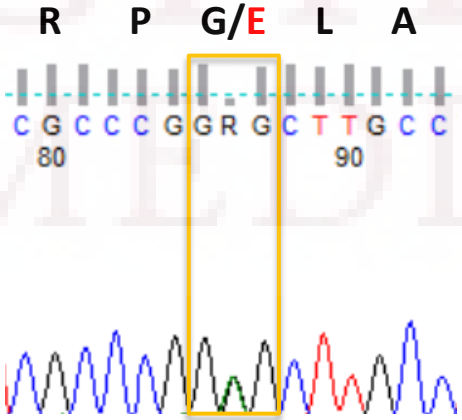
R6978 sister

G**G**G → G**A**G



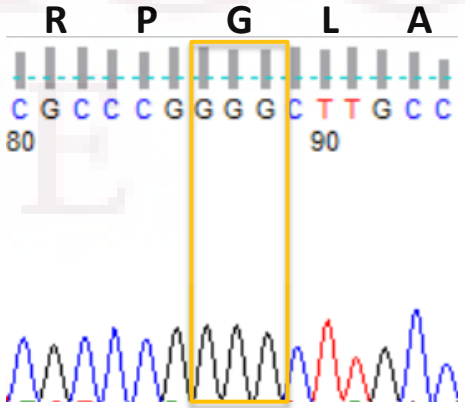
R6976 mother

G**G**G → G**A**G



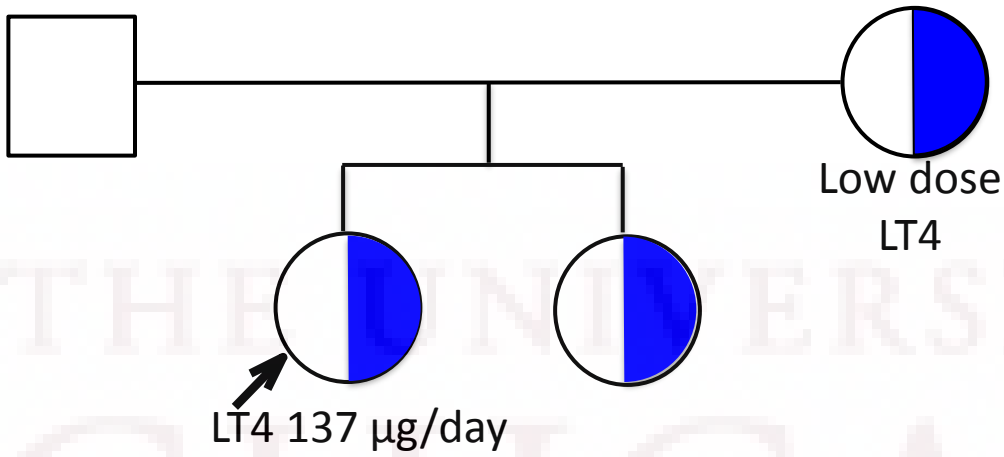
R6977 father

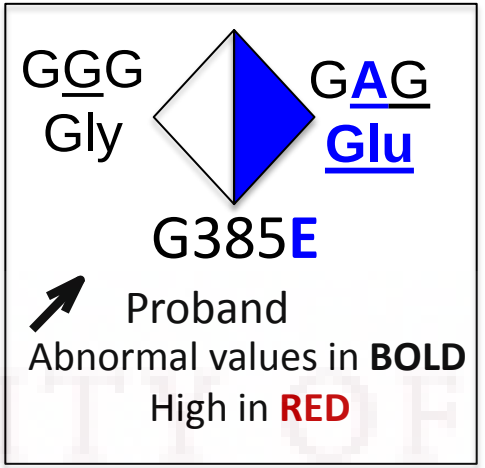
No mutation



- This variant has never been reported (Ensembl, gnomAD, dbSNP)

RTHß Family Msih





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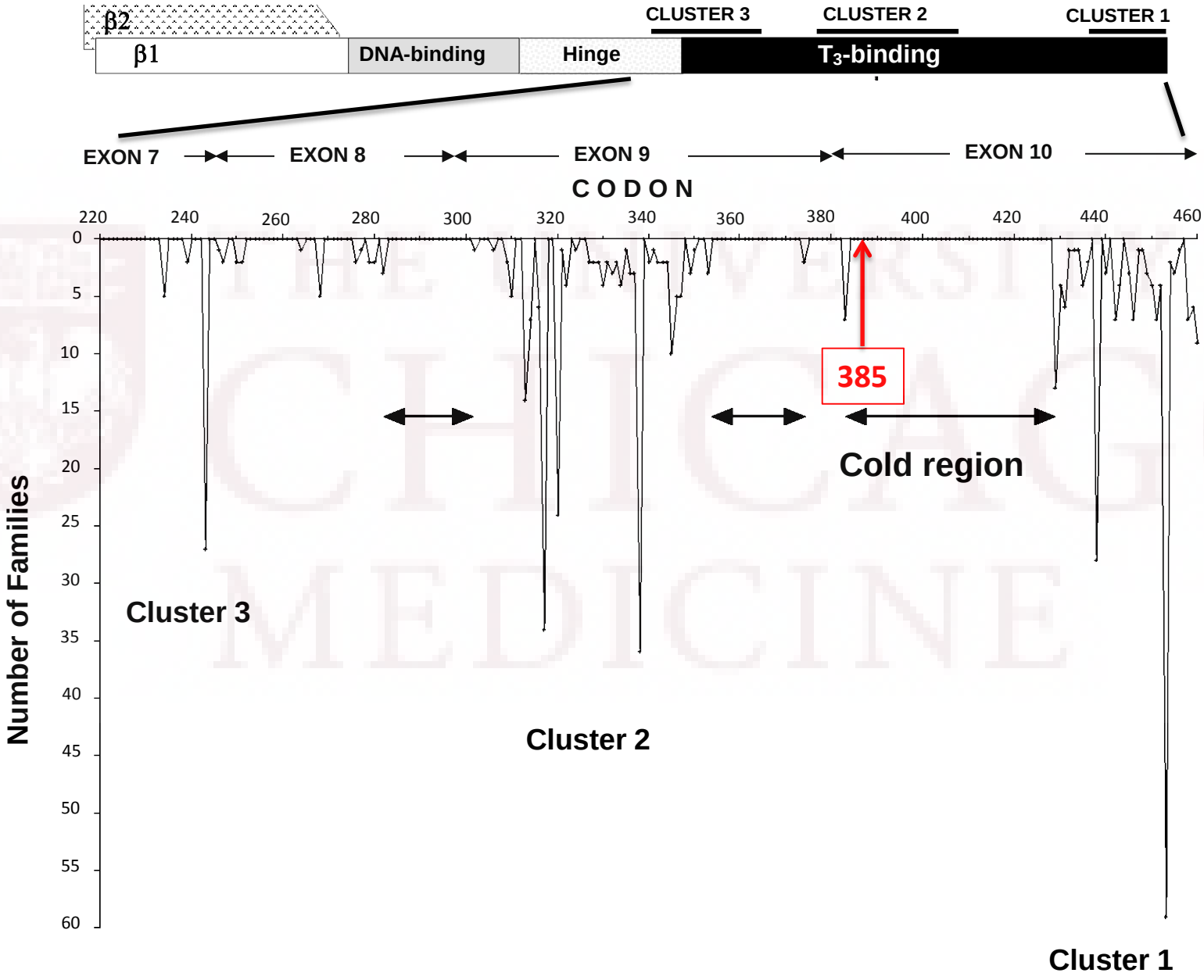
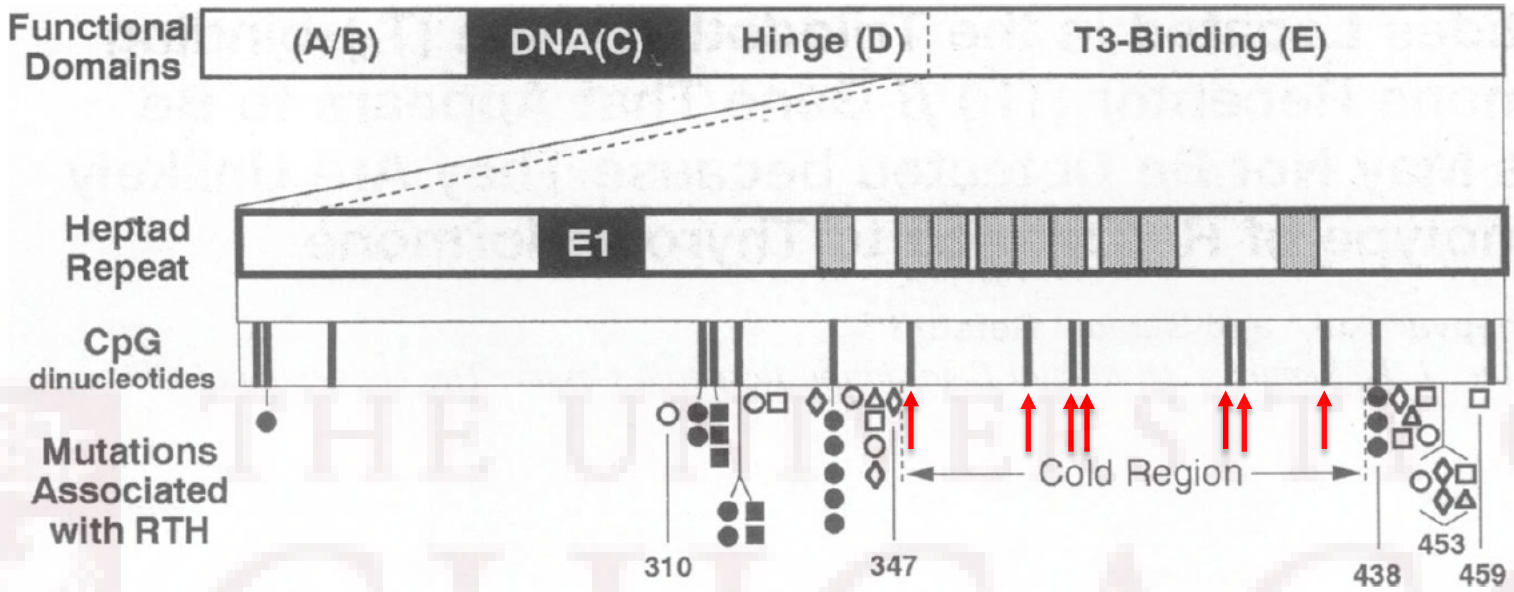
Suggestions

How does this help?


β2
β1
DNA-binding
Hinge
T3-binding
CLUSTER 3
CLUSTER 2
CLUSTER 1
EXON 7
EXON 8
EXON 9
EXON 10
C O D O N
220
240
260
280
300
320
340
360
380
400
420
440
460
0
5
10
15
20
25
30
35
40
45
50
55
60
Number of Families
385
Cold region
Cluster 3
Cluster 2
Cluster 1



R320H	0.42±0.02[‖]	F64(CL), F95
R338W	0.21±0.04[‖]	F29, F106, F112(KT) FE, LT
G345R	<0.03[‖]	F44(Mf)
A352T	**0.81±0.09**[‖]	**Artifical mutations**
V376I	**0.94±0.19**	
R383C	**0.62±0.06**[‖]	
R383H	**0.72±0.12**[‖]	
P384L	**1.05±0.16**	
R410Q	**0.99±0.07**	
R410X	**<0.03**[‖]	
V414M	**0.95±0.11**	
R429Q	**1.00±0.00**	
R429W	**0.91±0.23**	
R438H	0.25±0.07[‖]	F45, F68, F111

Hayashi, Sunthornthepvarakul, Refetoff, JCI, 94:607,1994

Follow up:

L-T4 dose was increased from 137 to 150 µg daily.
This increased her FT4I from 11.2 to 13.8 (6 – 10.5)
Normalized TSH to 0.6 µIU/mL
TT3 remained in the normal range, at 129 ng/dL.

Interpretation:
The high total and free serum T4 with clearly normal T3 may represent:

1. Irregular intake of L-T4 or
2. Increased inactivation of T4 (see high rT3) seen in some individuals on L-T4 replacement.

Further follow up:

Sequencing of deiodinase 1 was negative
The proband did not harbor the deiodinase 2 variant Thr92Ala