Future developments

Warning

- I am born optimist
- I am a researcher
- I stress possibilities
  esp. the positive sides
- I do understand choices made so far
  do not criticize current choices,
  but do think we can do even better

Your genome

who knows his genome sequence ?

who had a DNA test ?

do you have the DRD4 7R gene ?

...future !?

...your grand children will not believe you dared to live without knowing your genome,

...nor your partner's genome

maybe good to start trying to understand what info your DNA contains

first a DNA test, then sex

nrc.nl

...future !?

nowadays nobody would start surgery without an X-ray,

why do we start treatment without knowing the genome ?

Olaf Rieß

... for the hospital

...a patient will not be treated when the basics, the DNA, is not known

...why risk undesired effects from treatment, when these can be determined beforehand ?

...why risk treating a problem for which the origin lies elsewhere (has a genetic component) ?

Your genome

"preventive medicine"

European Journal of Human Genetics

Article | Published: 05 October 2018

1 in 38 individuals at risk of a dominant medically actionable disease

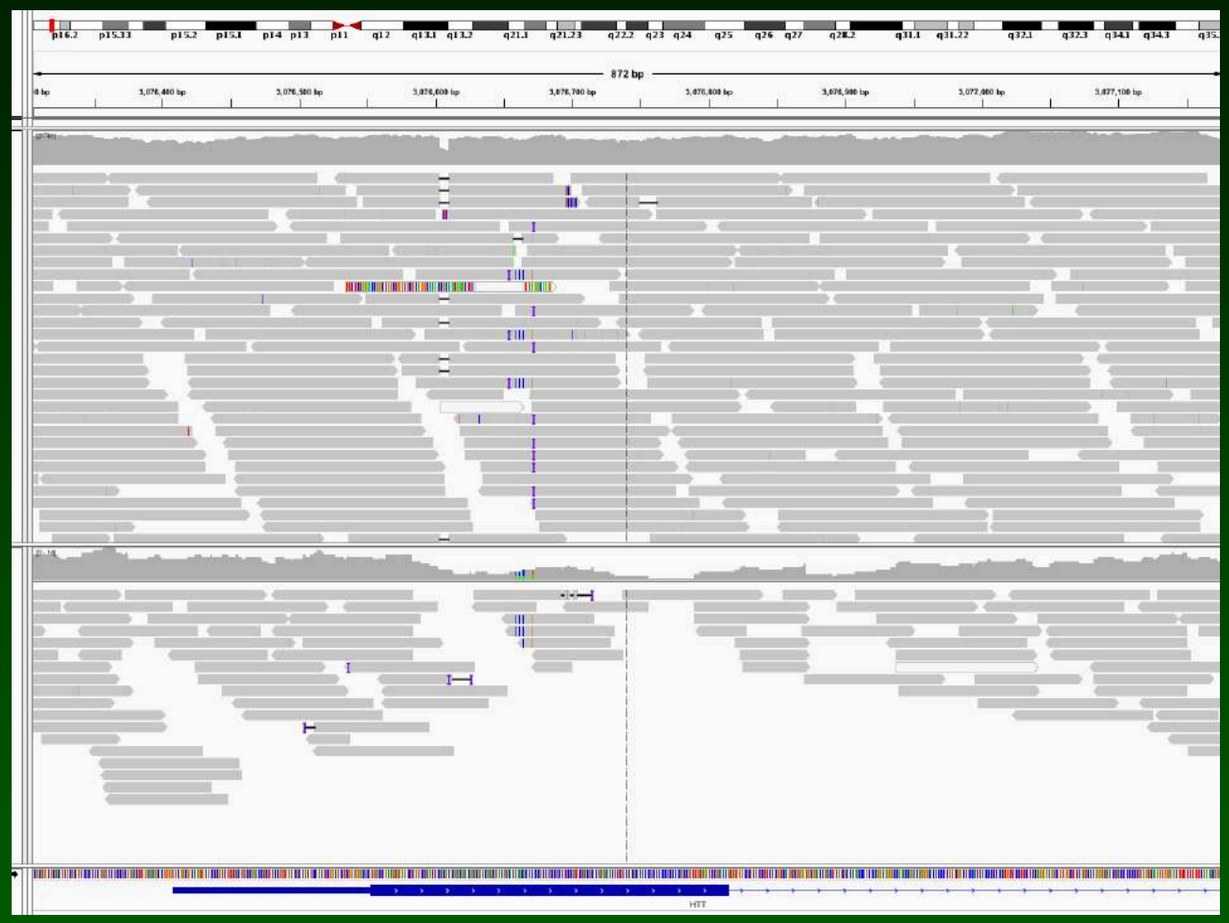
Lonneke Haer-Wigman, Vyne van der Schoot, Ilse Feenstra, Anneke T. Vulto-van Silfhout, Christian Gilissen, Han G. Brunner, Lisenka E. L. M. Vissers & Helger G. Yntema

...and from pharmacogenetic information to fun (bitter taste, m/paternal origin, ...)

My genome

sequenced it twice

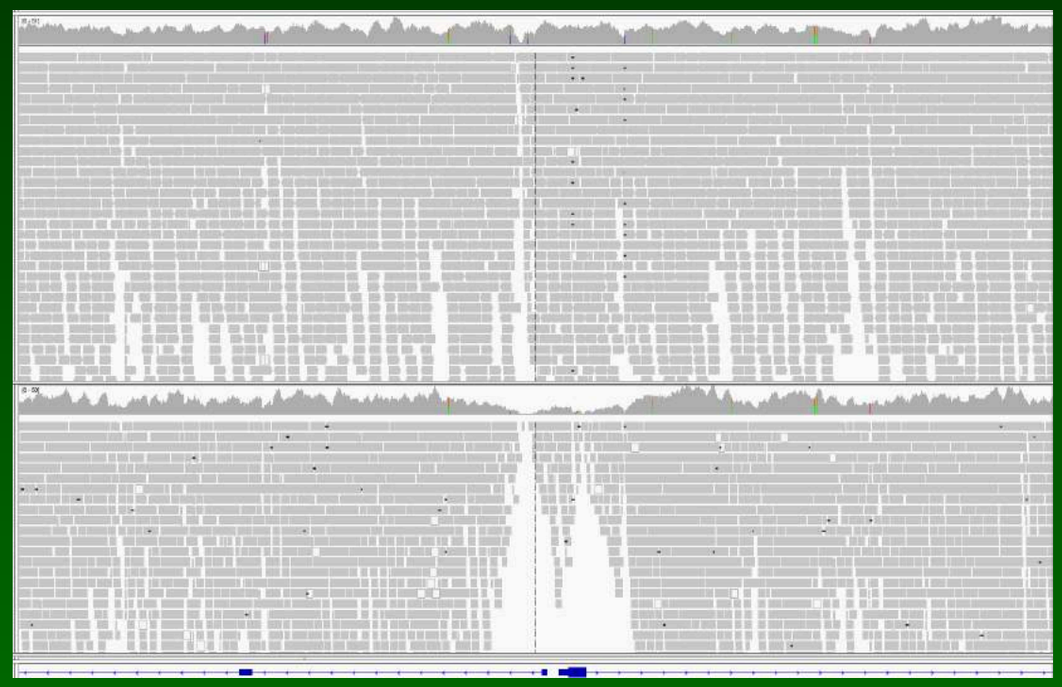
(best way to determine quality)



HTT gene

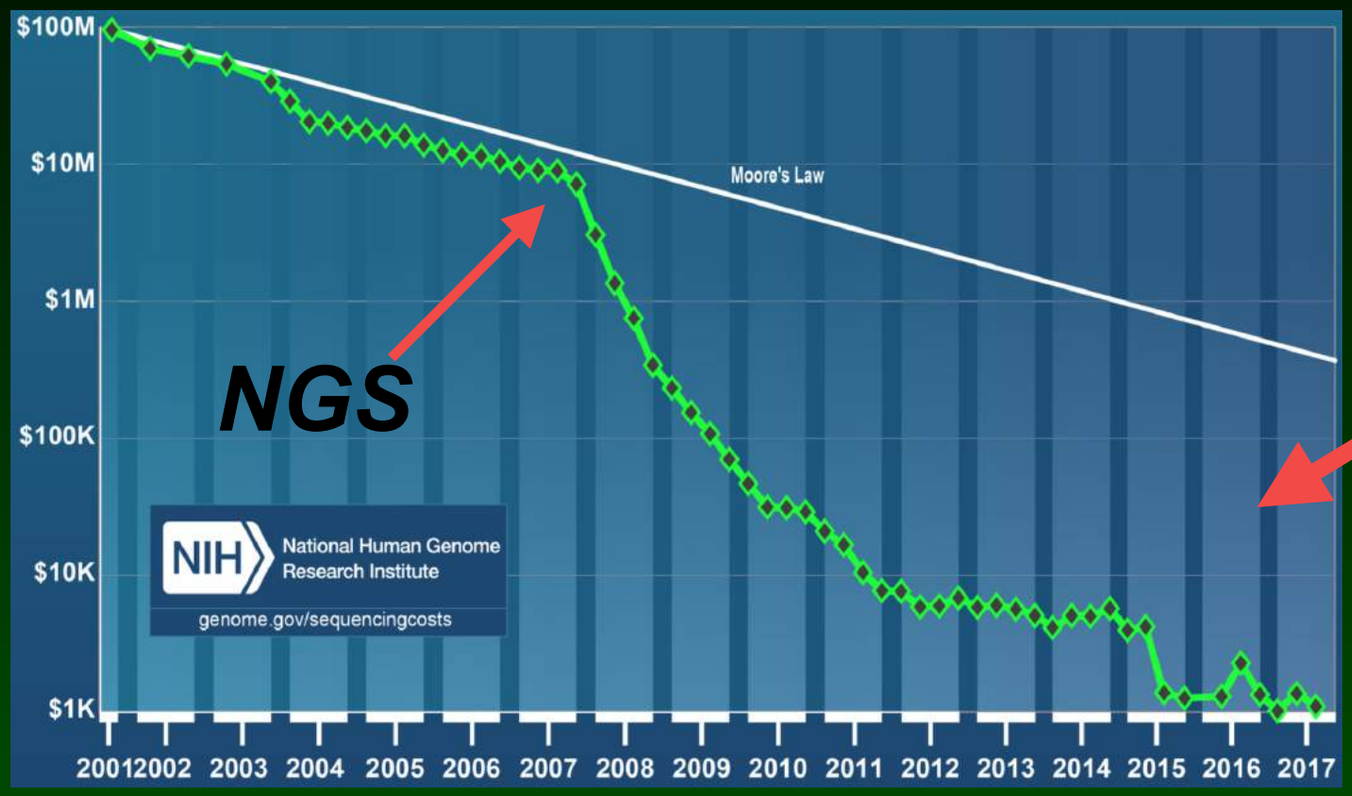
top: no PCR (2013)
bottom: few cycles (2017)

FMR1 gene



GC-bias (WGS)

Sequencing revolution



to a €1000 genome

now a reality !

Roche

454 LIFE SCIENCES

Solexa

illumina

Helicos BioSciences Corporation

Complete genomics

PACIFIC BIOSCIENCES

gnubio A Bio-Rad Company

Applied Biosystems

SOLiD™

ion torrent by life technologies

Oxford NANOPORE Technologies

more sequence
longer reads
faster
cheaper

...single molecule
...label-free

World record

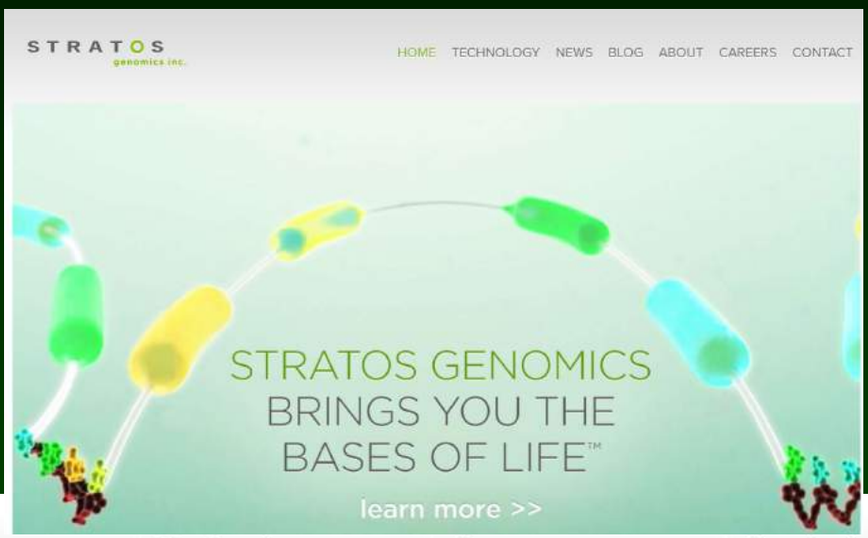
Kingsmore Has Done it Again: Rady Children's Set Guinness World Record for Genetic Diagnosis

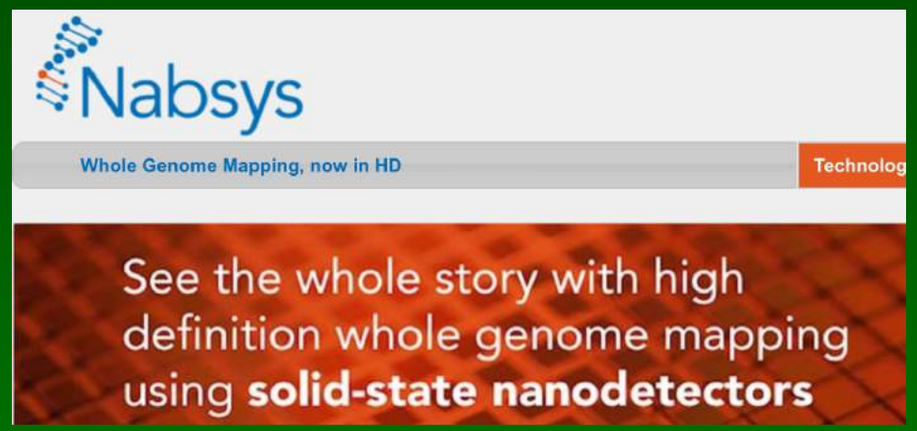
Front Line Genomics
Unzipping genes for the good of humanity

19.5 h
incl. diagnosis

Single molecule sequencing

nanopore technology

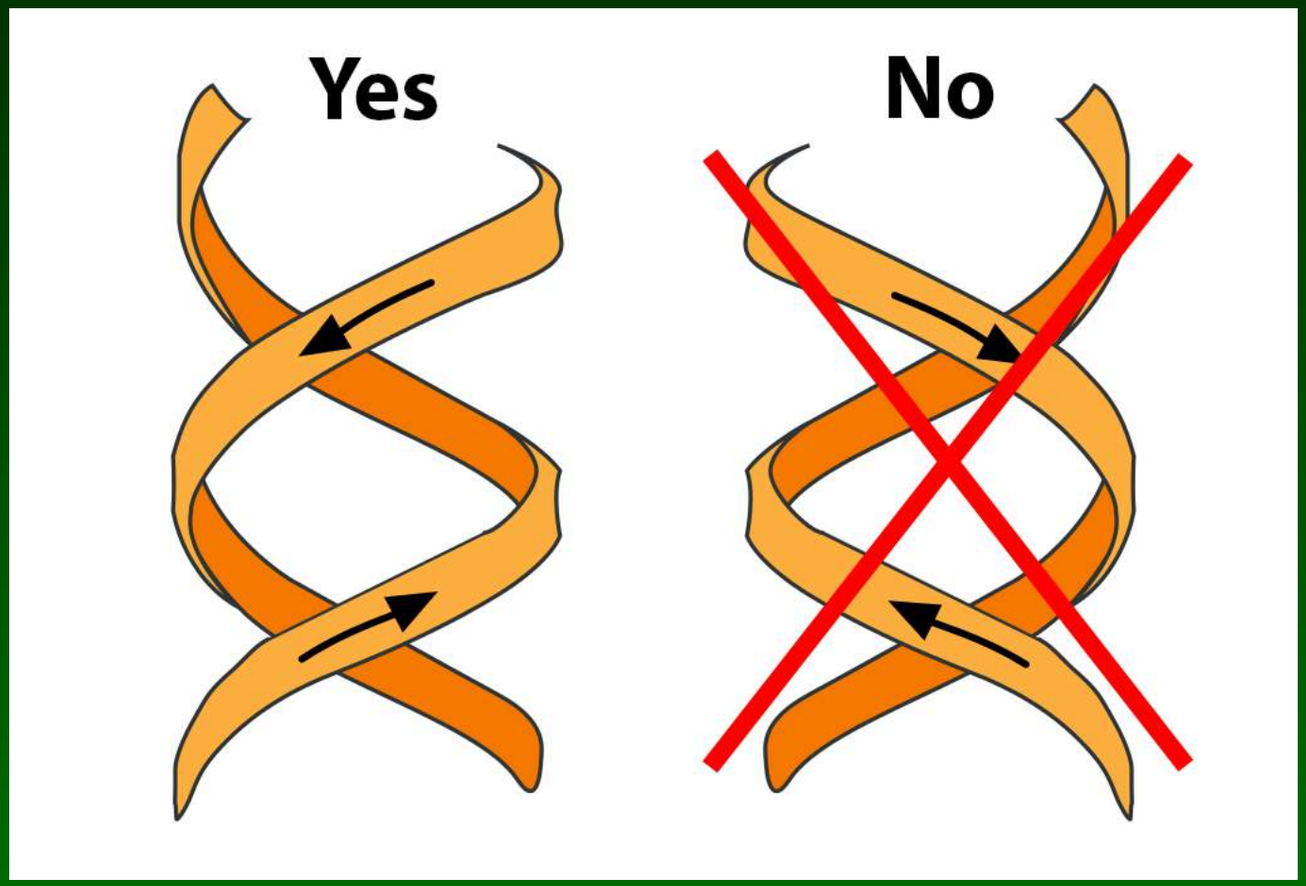




DNA

will the system work?

...YOU did pay attention at school



ESHG 2019

Future technology

twoporeguys

..and then

Oxford NANOPORE Technologies

SmidgION

sequence @home

Long-read seq

nature COMMUNICATIONS

Article | OPEN | Published: 30 June 2016

Long-read sequencing and *de novo* assembly of a Chinese genome

Lingling Shi, Yunfei Guo […] Kai Wang

Nature Communications **7**, Article number: 12065 (2016) | Download Citation

Journal of Human Genetics

Article | Published: 13 February 2019

A 12-kb structural variation in progressive myoclonic epilepsy was newly identified by long-read whole-genome sequencing

Takeshi Mizuguchi, Takeshi Suzuki, Chihiro Abe, Ayako Umemura, Katsushi Tokunaga, Yosuke Kawai, Minoru Nakamura, Masao Nagasaki, Kengo Kinoshita, Yasunobu Okamura, Satoko Miyatake, Naomichi Matsumoto

Genome Biology

Menu

Research | Open Access

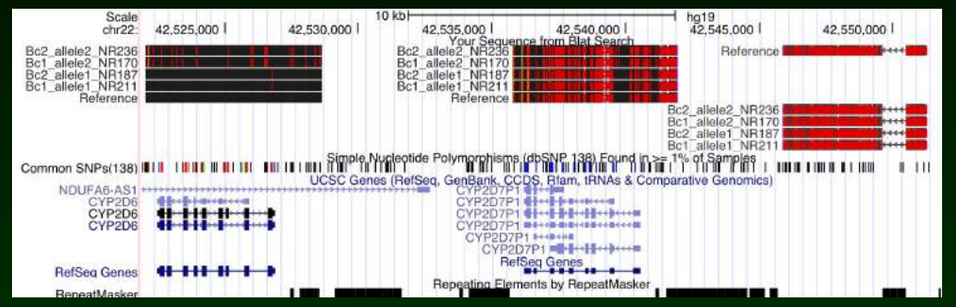
Systematic analysis of dark and camouflaged genes reveals disease-relevant genes hiding in plain sight

Mark T. W. Ebbert †, Tanner D. Jensen †, Karen Jansen-West, Jonathon P. Sens, Joseph S. Reddy, Perry G. Ridge,

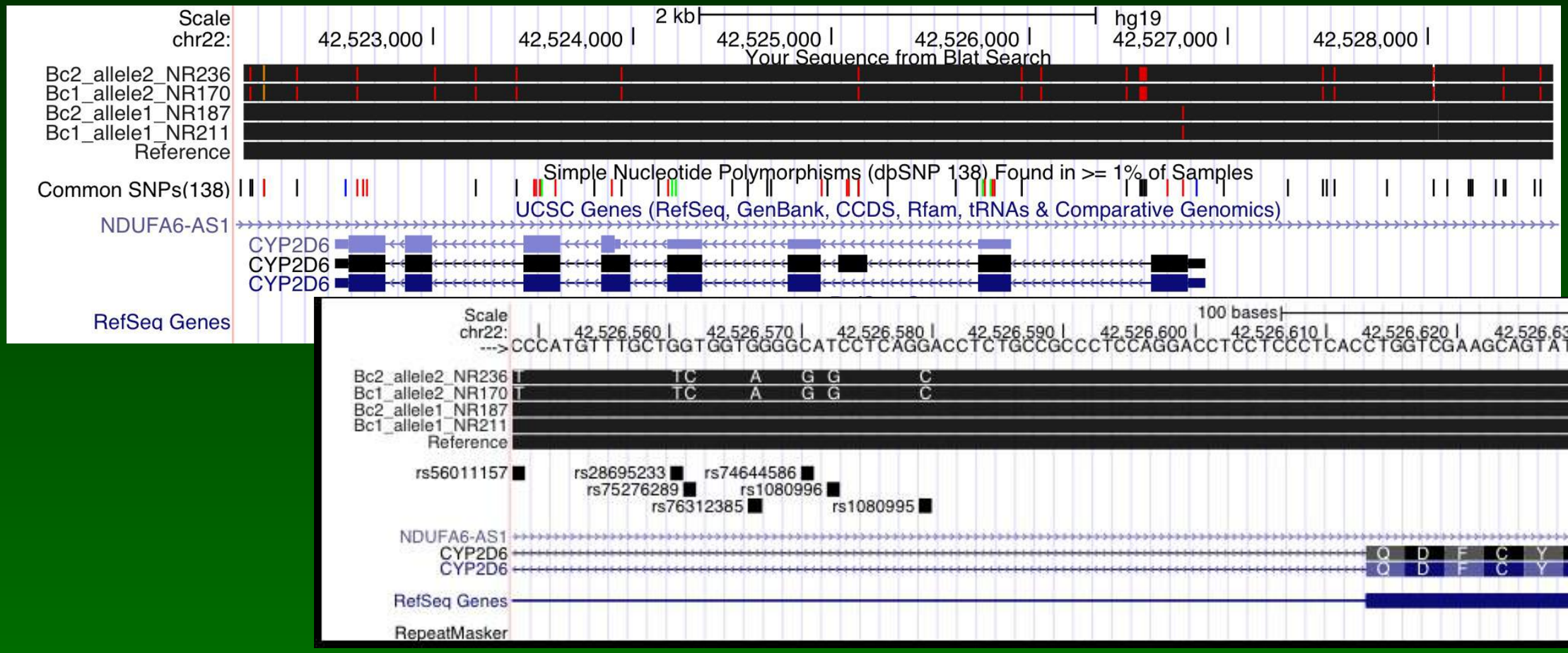
Long-read seq

(phasing)

- **CYP2D6**
  - ***long-range PCR > PacBio seq***
  - ***2 alleles clearly separated***



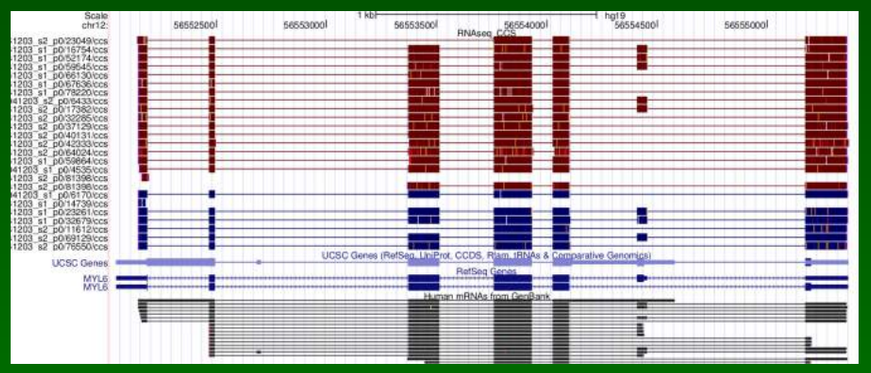
CYP2D6 *CYP2D6-like*



PacBio allele-seq

- **PKD1 gene**
  unique 3' end, repetitive 5' end
  long-range PCR far into repetitive region
- **PMS2 gene**
  several pseudogenes
  specific long-range PCR and/or
  discriminate based on sequence
  get & load long molecules
- **...other genes**
  CYP-genes
  discriminate maternal/paternal
  long-range cDNA-seq

MYL6 cDNA



Rare cases ?

- **many mono-genic diseases solved**

where are the di-genic diseases ??
I would expect many more

NATURE GENETICS VOLUME 44 | NUMBER 12 | DECEMBER 2012

Digenic inheritance of an *SMCHD1* mutation and an FSHD-permissive D4Z4 allele causes facioscapulohumeral muscular dystrophy type 2

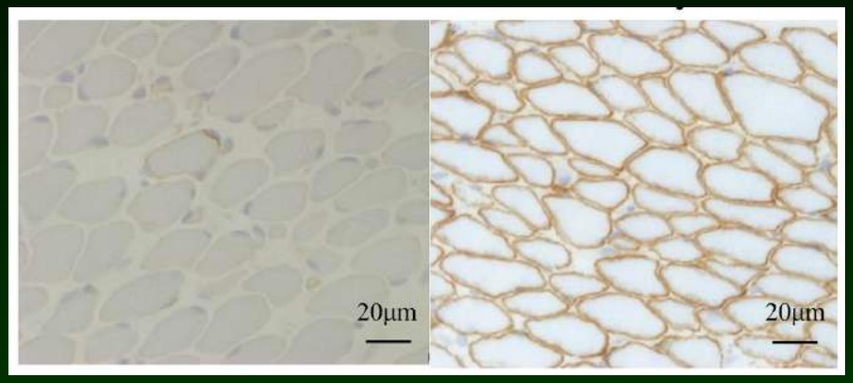
Richard J L F Lemmers[1,13], Rabi Tawil[2,13], Lisa M Petek[3], Judit Balog[1], Gregory J Block[3], Gijs W E Santen[4], Amanda M Amell[3], Patrick J van der Vliet[1], Rowida Almomani[4], Kirsten R Straasheijm[1], Yvonne D Krom[1], Rinse Klooster[1], Yu Sun[1], Johan T den Dunnen[1,4], Quinta Helmer[5], Colleen M Donlin-Smith[2], George W Padberg[6], Baziel G M van Engelen[6], Jessica C de Greef[1,12], Annemieke M Aartsma-Rus[1], Rune R Frants[1], Marianne de Visser[7], Claude Desnuelle[8,9], Sabrina Sacconi[8,9], Galina N Filippova[10], Bert Bakker[4], Michael J Bamshad[3,11], Stephen J Tapscott[10], Daniel G Miller[3,11] & Silvère M van der Maarel[1]

clear phenotype
unsolved FSHD cases

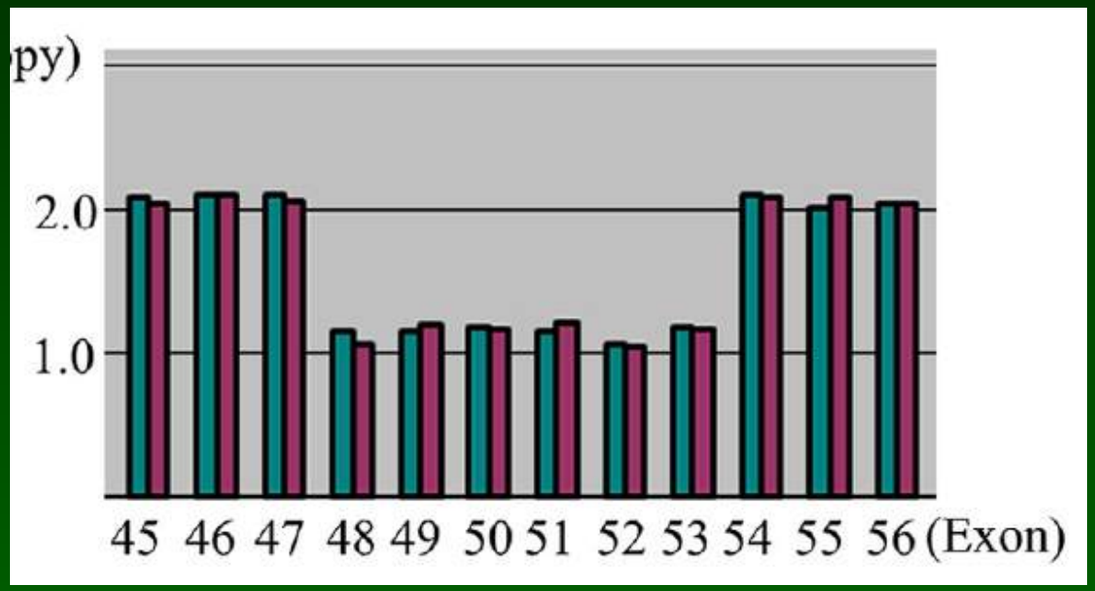
WES analysis several families
shared SMCHD1 variants

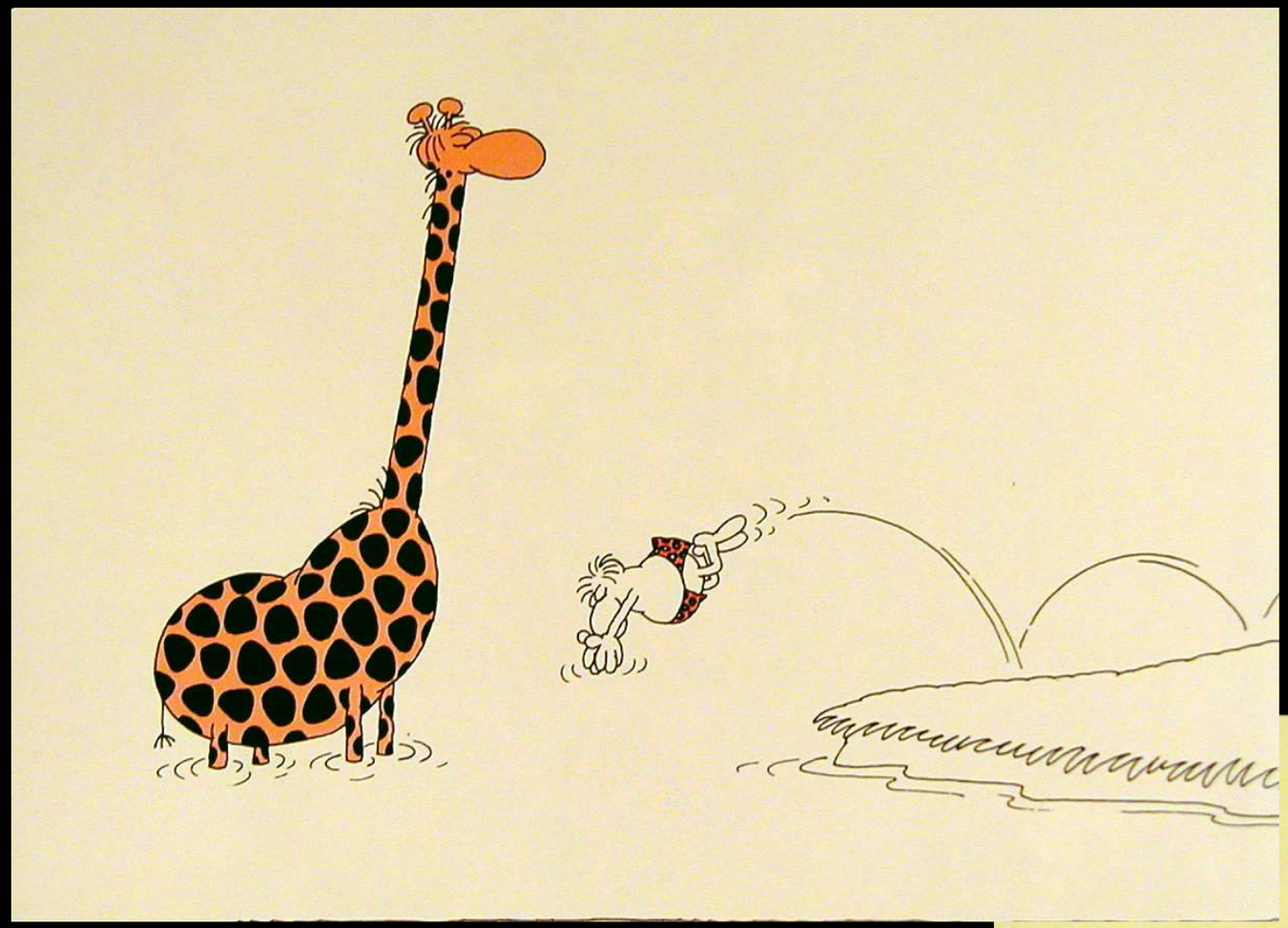
Female DMD

- **muscle biopsy**
  no dystrophin staining
- **MLPA**
  deletion exons 48-53
  in-frame
- **sequencing**
  no deleterious variants
- **X-inactivation**
  random

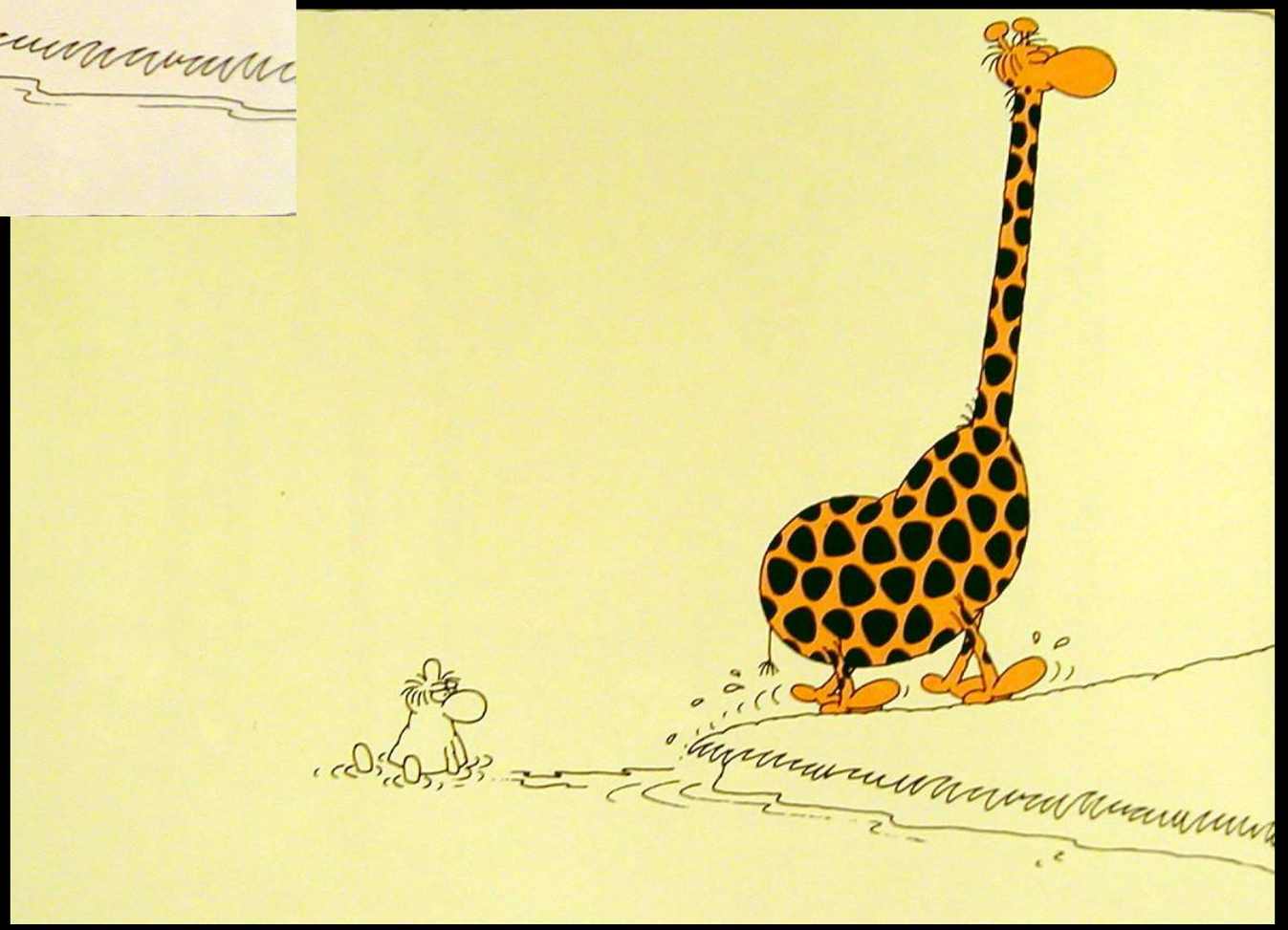


patient *control*





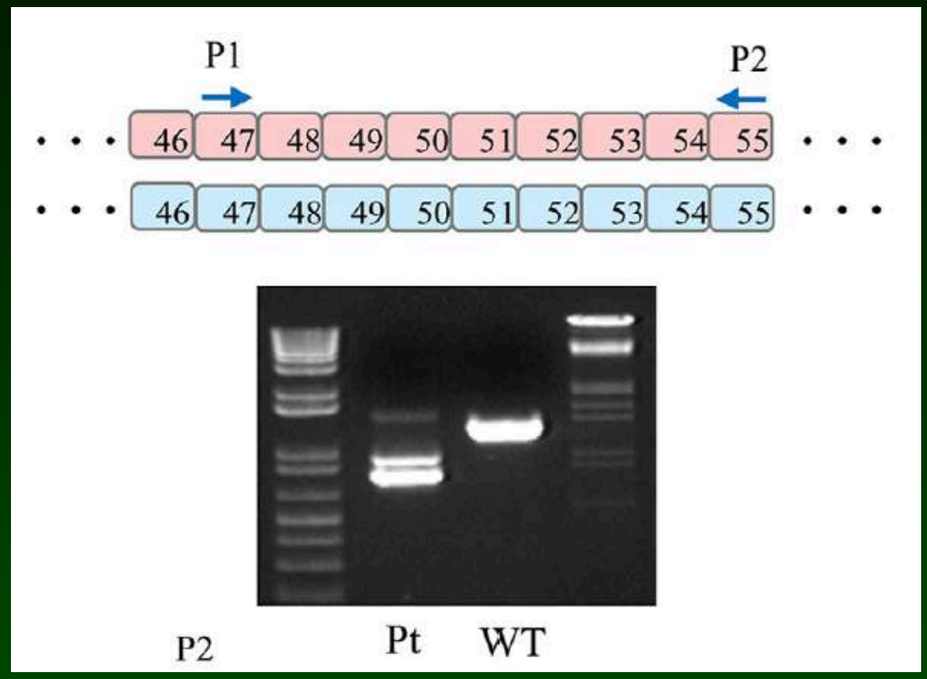
Is my conclusion right ?



Female DMD

- **RNA analysis**

RNA analysis



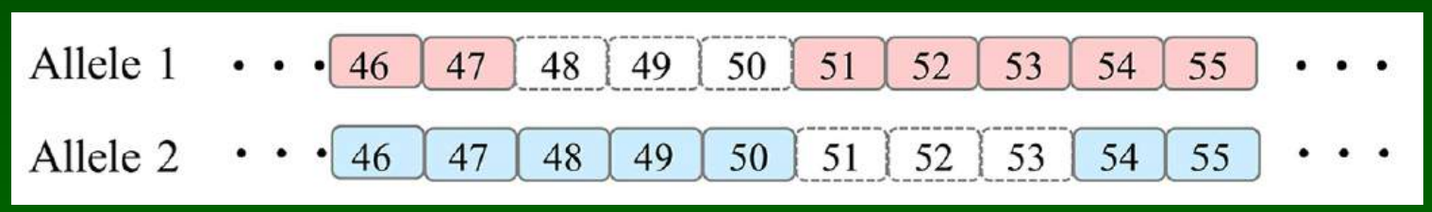
Available online at www.sciencedirect.com
ScienceDirect
Neuromuscular Disorders 27 (2017) 569–573
www.elsevier.com/locate/nmd

Case report

Duchenne muscular dystrophy in a female with compound heterozygous contiguous exon deletions

Eri Takeshita [a,*], Narihiro Minami [b,c], Kumiko Minami [c], Mikiya Suzuki [d], Takeya Awashima [a], Akihiko Ishiyama [a], Hirofumi Komaki [a], Ichizo Nishino [c,e], Masayuki Sasaki [a]

two different deletions, both frame shifting



RNA, it exists !

..the neglected molecule

under-appreciated

most go blindly DNA > protein

..there is much more

Describe ?

NOTE: reference sequences can be assumed to be known, spaces in descriptions used for clarity only.

MLPA shows all exons are present except exons 3 to 6 (nucleotide c.411 to 1428). Which description(s) is/are correct ?

- **c.411-?_1428+?del**
- **c.(411-?_1428+?)del**
- **c.(410+1_411-1)_(1428+1_1429-1)del** ***correct***
- **c.411_?_1428del**
- **none of these**

NOTE: using probe-based positions would be even better

GLOBAL VARIOME

HGVS HUMAN GENOME VARIATION SOCIETY

MLPA variants

LOVD 3 — Leiden Open Variation Database

THE HUMAN VARIOME PROJECT

Global Variome shared LOVD — LOVD is supp

CAPN3 (calpain 3, (p94))

Curators: Johan den Dunnen and Jacqui Beckmann

Genes | Transcripts | Variants | Individuals | Diseases | Screenings | Submit

CAPN3 gene homepage

This database is one of the gene variant databases from the Leiden Muscular Dystrophy pages

General information	
Gene symbol	CAPN3
Gene name	calpain 3, (p94)
Chromosome	15
Chromosomal band	q15.1
Imprinted	Unknown
Genomic reference	NG_008660.1
Transcript reference	NM_000070.2
Exon/intron information	NM_000070.2 exon/intron table
Associated with diseases	LGMD-2, LGMD-2A
Citation reference(s)	-
Refseq URL	Genomic reference sequence
Curators (2)	**Johan den Dunnen** and **Jacqui Beckmann**
Total number of public variants reported	2791
Unique public DNA variants reported	635
Individuals with public variants	2263
Hidden variants	445

exon	c.startExon	c.endExon	g.startExon	g.endExon	lengthExon	lengthIntron
1	-306	309	16398	17012	615	24368
2	310	379	41381	41450	70	1614
3	380	498	43065	43183	119	1467
4	499	632	44651	44784	134	1041
5	633	801	45826	45994	169	856
6	[illegible]02	945	46851	46994	144	2542
[illegible]	946	1029	49537	49620	84	1533
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]

calpain 3, (p94) (CAPN3) - 313 nt intron 11

(intronic numbering for coding DNA Reference Sequenc

```
                                                          g.58768
gtgtgcagtcctgattggctccagcccaggaaacatactttcccagggaggacgcttcca  c.1524+60

                                                          g.58828
ggggcttctagaggggccctctggcttcctcaatacccagtgacccacagagctcctggt  c.1524+120

                                  g.58865
atcaggaccacttgtgtttgtaacaagcaaaaaatac                     c.1524+157

-------------------- middle of intron ----------------------
                  g.58866                                 g.58901
                  c.1525-156  cagggggggcattagagaggcagtggagcgggcctg  c.1525-121

                                                          g.58961
gcagaacaggtgcctgggggtcaggcttccgcatgcgggctgcagttgctggcattgcct  c.1525-61

                                                          g.59021
tccgcaggctcctcatcctcattcacatctgaagcatcttcctttctgtttcttctcaag  c.1525-1
```

Graphical displays and utilities	
Graphs	Graphs displaying summary information of all variants in the database »
Reading frame checker	The Reading-frame checker generates a prediction of the effect of whole-exon changes. Active for: NM_000070.2.
UCSC Genome Browser	Show variants in the UCSC Genome Browser (full view, compact view)
Ensembl Genome Browser	Show variants in the Ensembl Genome Browser (full view, compact view)
NCBI Sequence Viewer	Show distribution histogram of variants in the NCBI Sequence Viewer

LOVD Leiden Open Variation Database

LUMC

MLPA variants

Graphical displays and utilities	
Graphs	Graphs displaying summary information of all variants in the database »
Reading frame checker	The Reading-frame checker generates a prediction of the effect of whole-exon changes. Active for: NM_000070.2.
UCSC Genome Browser	Show variants in the UCSC Genome Browser (full view, compact view)
Ensembl Genome Browser	Show variants in the Ensembl Genome Browser (full view, compact view)
NCBI Sequence Viewer	Show distribution histogram of variants in the NCBI Sequence Viewer

Currently viewing gene/transcript: **CAPN3 / NM_000070.2**

Deletion or Duplication	Deletion
From exon	6
To exon	11
	Check

Deleting exon 6 to exon 11 leads to ... an IN-FRAME deletion.

According to the CAPN3_NM_000070.2 reference sequence in the LOVD database, ex06ex11del -> c.802-?_1524+?del

...will be modified to report HGVS correct

c.(801+1_802-1)_(1524+1_1525-1)del

LOVD Leiden Open Variation Database

LUMC

LOVD queries

queries

- | = or
  A>T |T>A
- ! = not
  > !>T

4997 entries on 50 pages. Showing entries 1001 - 1100.

100 per page | Legend | « First | ‹ Prev | 1 | 2 | 3 | 4 | 5

sort

Effect	Reported	Exon	DNA change (cDNA)
+/.	1	6	c.484dup
+/.	1	6	c.484_493delinsTGGCTTTGAAT
+/.	1	6	c.485_486dup
+/.	1	6	c.487T>G
+/.	2	6	c.488G>A
+?/.	1	6	c.488G>C
+/.	1	6	c.489G>A
-?/., +?/., ?/.	3	6	c.494A>T

Search terms	*Field*	*Result*
>	Variant/DNA	Show only substitutions
A>T|T>A	Variant/DNA	Show only A to T or T to A substitutions
c.328 >	Variant/DNA	Show only substitutions at position c.328
p.(Arg X)	Variant/Protein	Show only arginine to stopcodon changes
Asian	Patient/Origin/Ethnic	Shows "Asian", but also "Caucasian" entries
Asian !Caucasian	Patient/Origin/Ethnic	Shows "Asian", but no "Caucasian" entries
Asian|African !Caucasian	Patient/Origin/Ethnic	Shows "Asian" or "African", but no "Caucasian" entries
"South Asian"	Patient/Origin/Ethnic	Shows "South Asian", but no "South East Asian" entries

Arg *|Ter

LOVD Leiden Open Variation Database

LUMC

Web Images Video Maps News Shopping Gmail more ▾

Google

c.62G>A lovd

Search | Advanced Search | Preferences

Web Show options...

Did you mean: c.62G>A ***loved***

Search unique variants - **LOVD** - Leiden Open Variation Database ...
02, **c.62G**>A, -, r.(?) p.(Arg21Gln), ARG1_00001, -, -, Mitchell 2009, DNA, HRMA ... Powered by **LOVD** v.2.0 Build 18. Enabled modules: showmaxdbid,
chromium.liacs.nl/**LOVD**2/variants.php?select_db=ARG1&a
unique&search_pathogenic_=- - 28k - Cached - Similar pag

View unique variants - **LOVD** - Leiden Oper
01, 1-**62G**>A (Reported 10 times), -, -, GCK_00037,
Leu20Pro, GCK_00063, -. 02, 106C>T (Reported 2 ti
chromium.liacs.nl/**LOVD**2/variants.php?action=view_
Cached - Similar pages
More results from chromium.liacs.nl »

Variants - NGRL, Manchester **LOVD** - Leiden Ope
NGRL, Manchester **LOVD**. ubiquitin protein ligase E3A (UE
RNA change. Protein, p.Cys21Tyr (predicted) ...
ngrl.man.ac.uk/**lovd**2/variants.php?select_db=UBE3A&acti
0000082%2C0000082%2C21 - Similar pages

LOVD
Leiden Open Variation Database

Essayez avec cette orthographe : c.62G>A ***love***

Google

c.1A>G LOVD

ALL IMAGES VIDEOS SHOPPING NEWS MAPS BOOKS

Did you mean: c.1A>G ***LOVE***

HBB:c.1A>G - bx.psu.edu).
https://lovd.bx.psu.edu › variants › DNA...

HBB homepage View unique variants Public list of submitters Submit new data
View unique variants · Search unique ... LOVD - Variant listings for HBB. Unhide

All transcript variants in gene FANCA - Global Variome shared LOVD
https://databases.lovd.nl › shared › FANCA

Web Images Video Maps News Shopping Gmail more ▾

Google

c.62G>A lovd

Search Advanced Search Preferences

Web Show options...

Did you mean: c.62G>A ***loved***

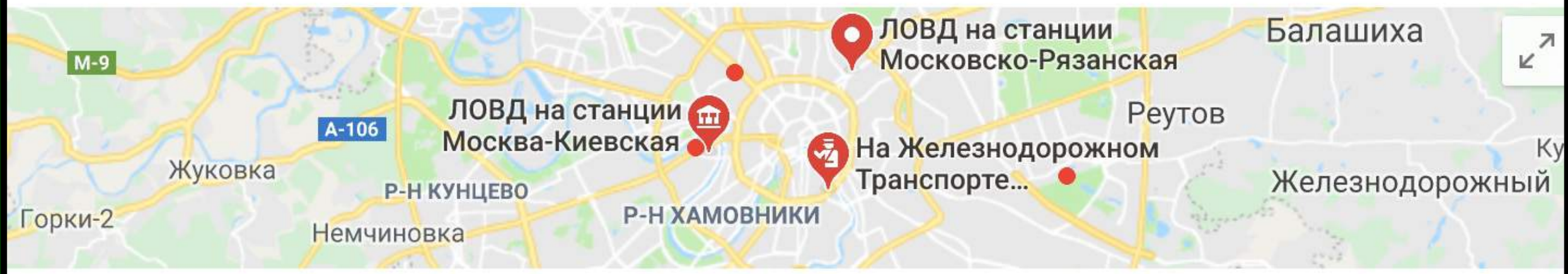
Search unique variants - **LOVD** - Leiden Open Variation Database ...
02, **c.62G**>A, -, r.(?) p.(Arg21Gln), ARG1_00001, -, -, Mitchell 2009, DNA, HRMA ... Powered

LOVD
Leiden Open Variation Database

Essayez avec cette orthographe : c.62G>A ***love***

Lovd

Москва Обновить местоположение



https://lovd.bx.psu.edu › variants › DNA...

HBB homepage View unique variants Public list of submitters Submit new data View unique variants · Search unique ... LOVD - Variant listings for HBB. Unhide

All transcript variants in gene FANCA - Global Variome shared LOVD
https://databases.lovd.nl › shared › FANCA

Standards

- **annoying, ...but**
  we need them
  ...and use without errors

- **variants**
  HGVS nomenclature

- **phenotypes**
  Human Phenotype Ontology (HPO)

- **classification**
  ACMG

Variant classification

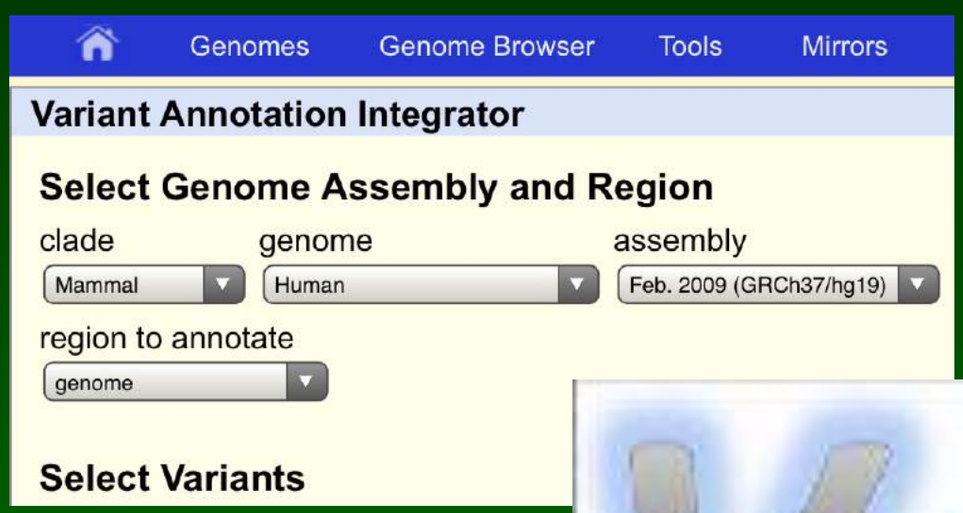
standards for classification

ACMG recommendations

labs start sharing classifications

ACMG:

"beware of variants that may impact splicing"



Gene panels

- **20,000 protein coding**
  60,000 total

- **which gene should be in a specific panel**

- **which transcript to use**
  preferred reference transcript

Genomics england

New rare disease gene tool launched
PanelApp

standards for analysis, agreement on what to analyse

Databases

...all these databases

DNA diagnostics is based on:

SHARING *what we know on the* ***relation***

between ***variants in genes*** *&* ***phenotypes***

without sharing, no DNA diagnostics

Variant of Unsufficient Sharing

(VUS)

A good idea ?

you are only allowed to perform clinical diagnosis when you have sequenced your own genome

(what about a VUS in your genome ?)

LOVD (Global Variome sha... 08:48
LOVD submission (TASP1)
Curator: LOVD-team, but with Curator vacancy Managers: Johan den Dunnen...

LOVD (Global Variome sha... 08:47
LOVD submission update (TASP1)
Curator: LOVD-team, but with Curator vacancy Dear Curator, Ayman El-Hatta...

LOVD (Global Variome sha... 08:40
LOVD submission update (TASP1)
Curator: LOVD-team, but with Curator vacancy Dear Curator, Ayman El-Hatta...

LOVD (Global Variome s... 08:38
LOVD submission (THOC6)
Curator: LOVD-team, but with Curator vacancy Dear Curator, MedGenome_d...

LOVD (Global Variome sha... 08:37
LOVD submission update (TASP1)
Curator: LOVD-team, but with Curator vacancy Dear Curator, Ayman El-Hatta...

LOVD (Global Variome sha... 08:29
LOVD submission (TASP1)
Curator: LOVD-team, but with Curator vacancy Managers: Johan den Dunnen...

LOVD (Global Variome shar... 08:16
LOVD submission

The DNA bank

you in control of your data

INVITED COMMENTARY

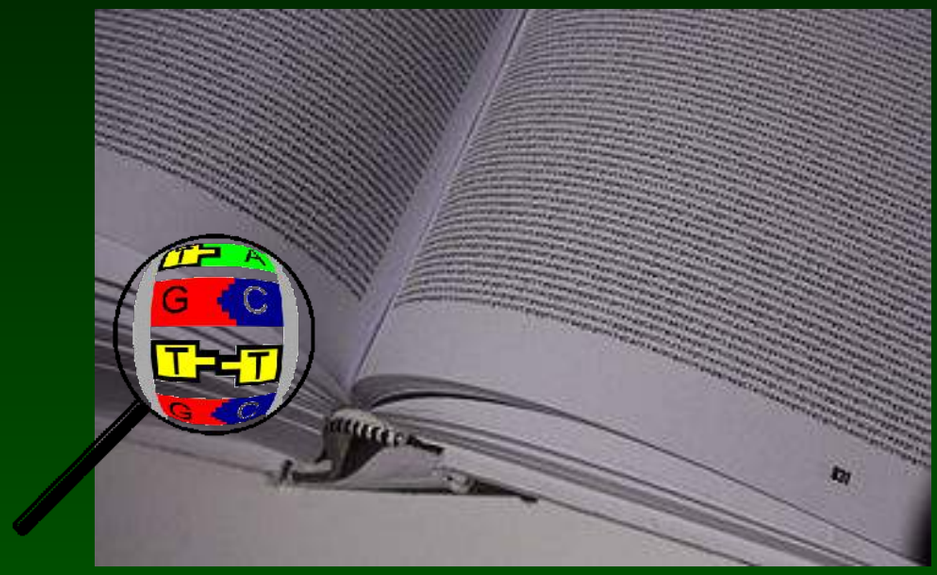
Human Mutation

OFFICIAL JOURNAL
HGVS
HUMAN GENOME VARIATION SOCIETY
www.hgvs.org

The DNA Bank: High-Security Bank Accounts to Protect and Share Your Genetic Identity

Johan T. den Dunnen*

Human Mutation (2015) 36: 657



everybody should own all their genetic data

when you do not agree, who should?

(the state, hospital, GP, insurance company, ..)

Adopt a gene !

become a
foster parent
database curator

THE
HUMAN VARIOME
PROJECT
an NGO official partner of UNESCO
sharing data • reducing disease

HGV
HUMAN GENOME
VARIATION SOCIETY

LOVD
Leiden Open Variation Database

claim your child at
gene.LOVD.nl

essential on your CV
...only ~15,000 available

with 7,000,000,000 people
chance 1/400,000

Your country !?

ru.LOVD.org

Russian Federation DNA Variant Database

Based on: LOVD Leiden Open Variation Database

RU

Home | Variants in individuals from Russian Federation | Variants by submitters from Russian Federation

This resource automatically retriev … d installation. The information retrieved is based on the geog … or **Russian Federation**).

Variants shown are either linked to … (patient), or to the country of origin of the *submitter* of the data. Note the differenc … can be submitted by a *submitter* from Belgium.

Queries … boxes in the column's header.

1	Andrey Marakhonov
2	Andrey Marakhonov
1	Andrey Marakhonov
1	Andrey Marakhonov
1	Anna Sokolenko
1	Anna Sokolenko
1	Anna Sokolenko
1	Anna Sokolenko
1	Evgeny Suspitsin
1	Grigorij Yanus
1	Grigorij Yanus
1	Grigorij Yanus
1	Grigorij Yanus
1	Grigorij Yanus

Home | Variants in individuals from Russian Federation | Var … ussian Federation

126 entries on 6 pages. Showing entries 1 - 25.

25 per page | **Legend** | « First | ‹ Prev | 1 | 2 | 3 | 4 | 5 | 6

scroll

Gene	Transcript	Effect	DNA change (cDNA)	Protein
APC	NM_000038.5	+/.	c.218_219insTA	p.(Lys73Asnfs*6)
APC	NM_000038.5	+/.	c.694C>T	p.(Arg232*)
APC	NM_000038.5	+/.	c.712C>T	p.(Gln238*)

Country node

GV shared LOVD offers access to country home page

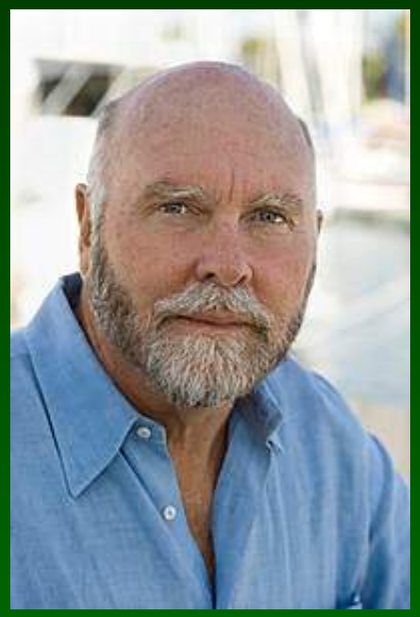
2008

Focus on disease

(individual genomes sequenced)

James Watson

JLupsky, Kim,
GChurch, DTutu, JFlattery,
MSnyder,
….



Craig Venter

Marjolein Kriek

A rumour

female DNA finally sequenced

"here the defective gene for parking a car backwards"

AdelaideNow
adelaidenow.com.au

From: **Pastafarian ®**
Subject: **re: Scientists claim to understand women**
Nobel Prize for them

27/05/2008 3:15:32 PM
post id: 3604572

Scientists crack women's DNA code

FINALLY, men may be able to understand women, it seems. Dutch scientists said they have mapped the full genetic sequence of an individual woman's DNA for the first time.

Researchers at Leiden University Medical Centre said they had sequenced the genome of one of their researchers, geneticist Marjolein Kriek, and plan to publish it after review.

2008

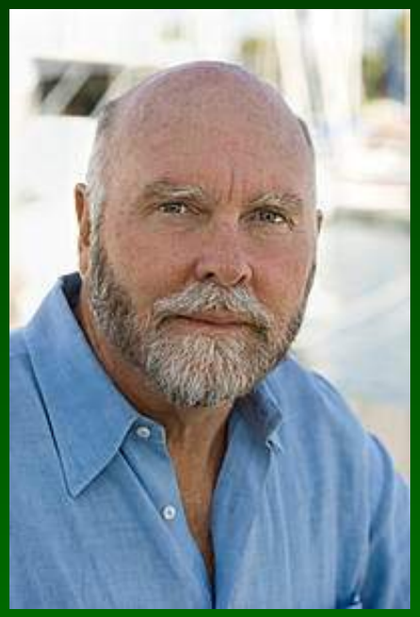
Focus on disease

(individual genomes sequenced)

James Watson

JLupsky, Kim,
GChurch, DTutu, JFlattery,
MSnyder,
….

Marjolein Kriek

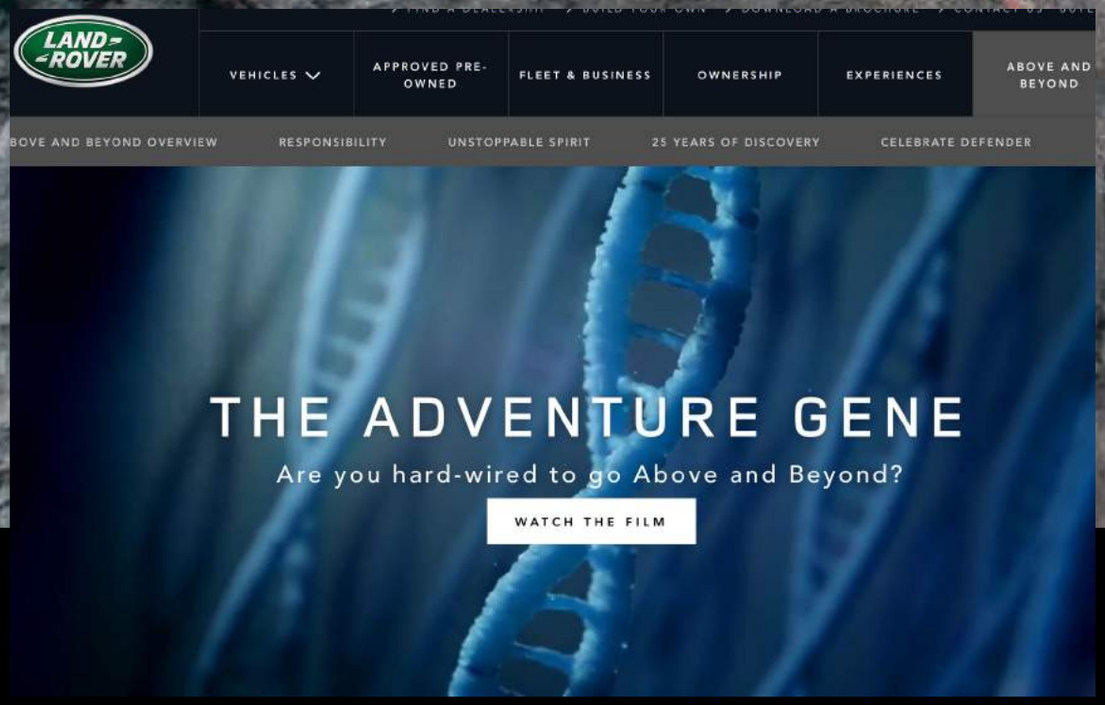


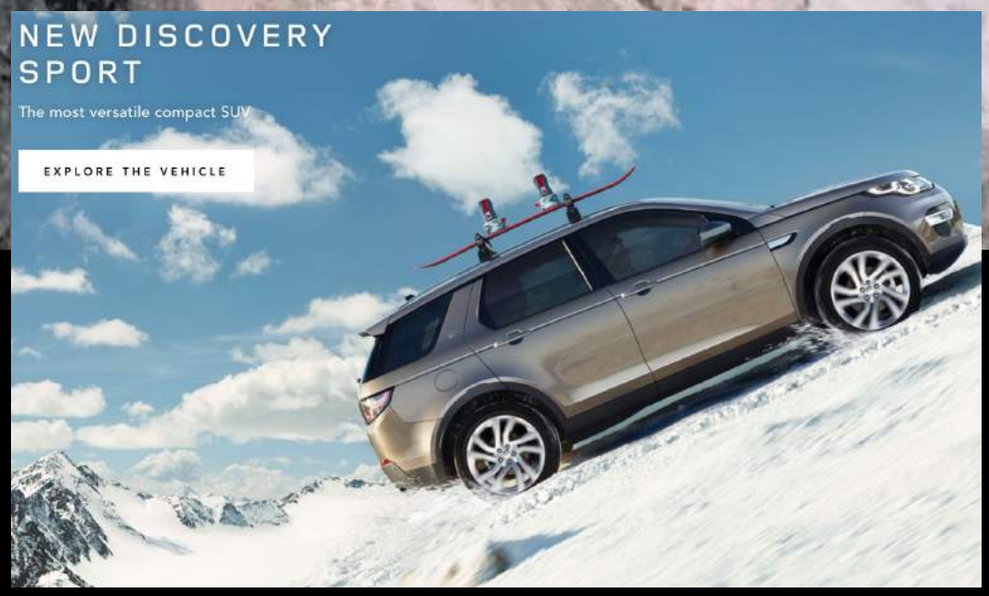
Craig Venter

conclusion 'sick' much easier then 'healthy'

Do you have the DRD4 7R gene ?

1 in 4 has the
DRD4 7R gene

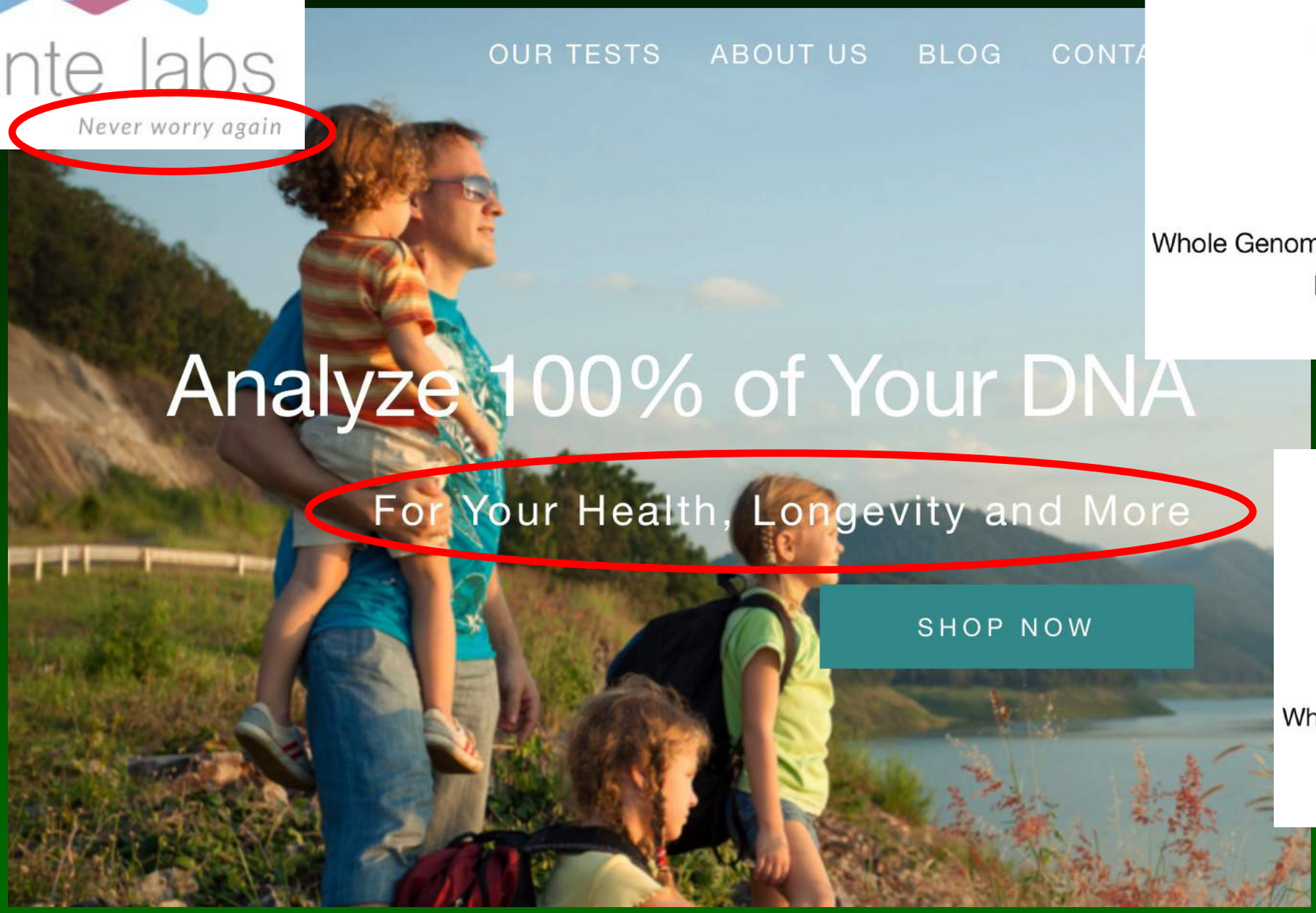




Commercial DNA test

dante labs

Never worry again



Whole Genome Sequencing (WGS) - Full DNA Analysis

£399

Whole Exome Sequencing (WES) - Sequence all Your Genes

£299

2017

Special offer

dante labs
Never worry again

maybe offer on Halloween
because it is a scary thing to do ?

Test the baby

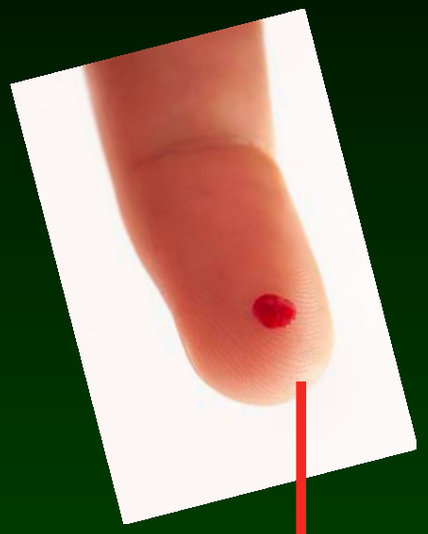
the nicest and most original present

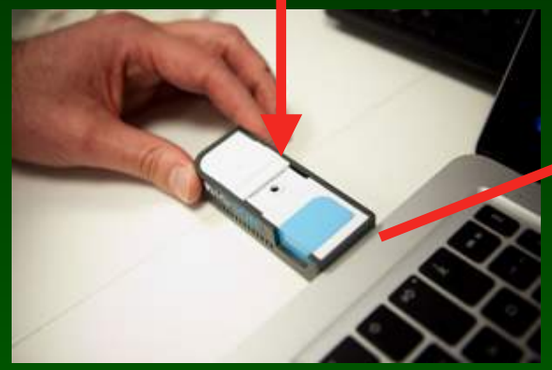
Predictions

...it is good we can not yet trust predictions

(dangerous tools, eventually they will take over your job)

Future VEP





LOVD
Leiden Open Variation Database

FINAL

VeIP

CE

GENOME REPORT

(complete error free)