

New Developments in Genetic
Genealogy
Roots Users Group
Nov 8, 2025

Stephen Ullrich, PhD.

What is the true extent of Extra Pair Paternity in Western societies?

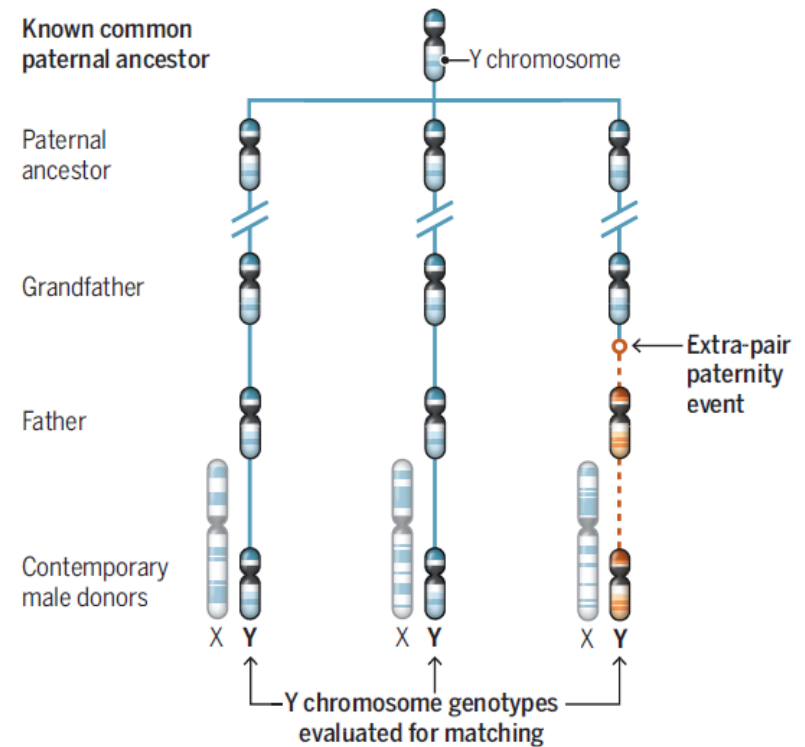
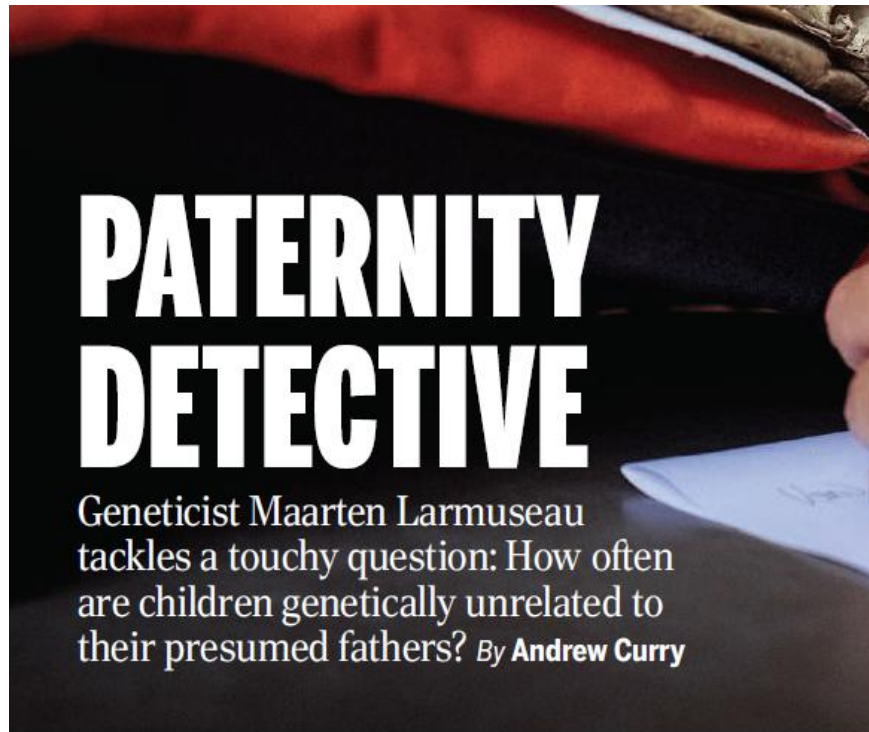


Jan Steen – Celebrating the Birth (1664)

Famous Dutch painting from 1664 with hidden message, “depicting childbirth celebrations by focusing on the father rather than the mother, and by suggesting the child’s illegitimacy. His subversive treatment of the subject was not apparent until the painting was cleaned in 1983, and the removal of 19th-century paint revealed a hand making the sign of the cuckold above the baby’s head.”

"Mommy's baby, daddy's maybe": Genetic Genealogical Reconstruction of Extra-pair Paternity (EPP) Behaviour, Maarten Larmuseau PhD

Head of the Laboratory of Human Genetic Genealogy, KU (Katholieke Universiteit) Leuven, Belgium



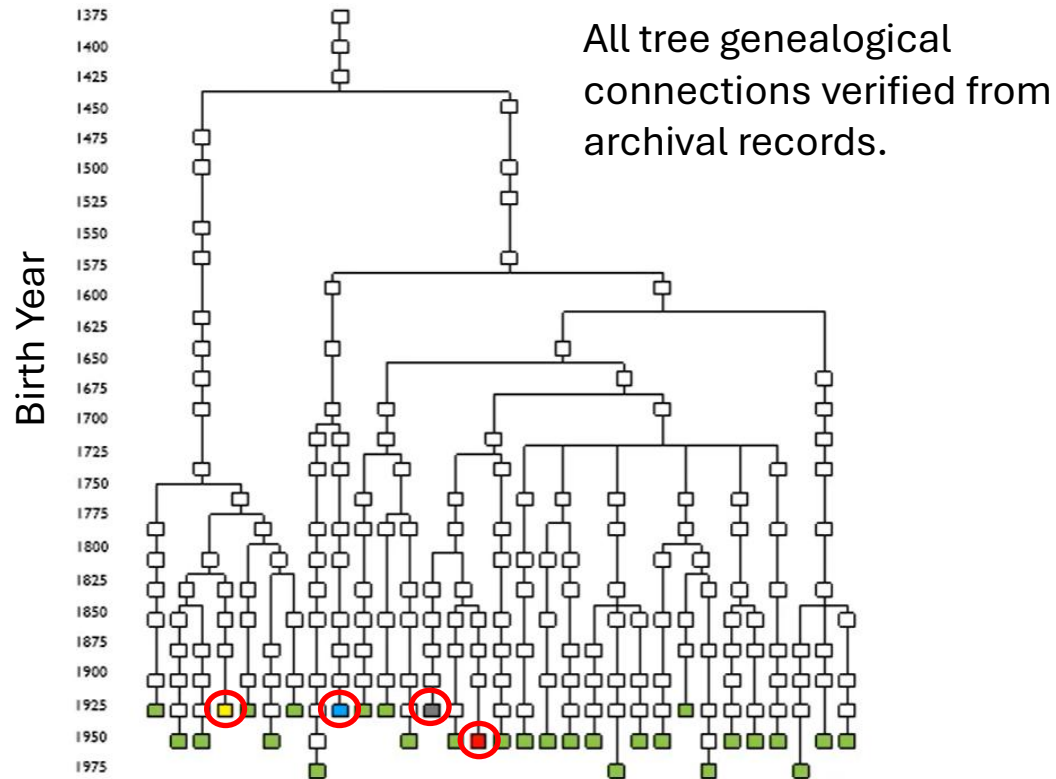
Source: Science: [https://www.cell.com/current-biology/fulltext/S0960-9822\(19\)31305-3](https://www.cell.com/current-biology/fulltext/S0960-9822(19)31305-3)

Genetic Genealogical Reconstruction of Extra-pair Paternity (EPP) Behaviour – Maarten Larmuseau

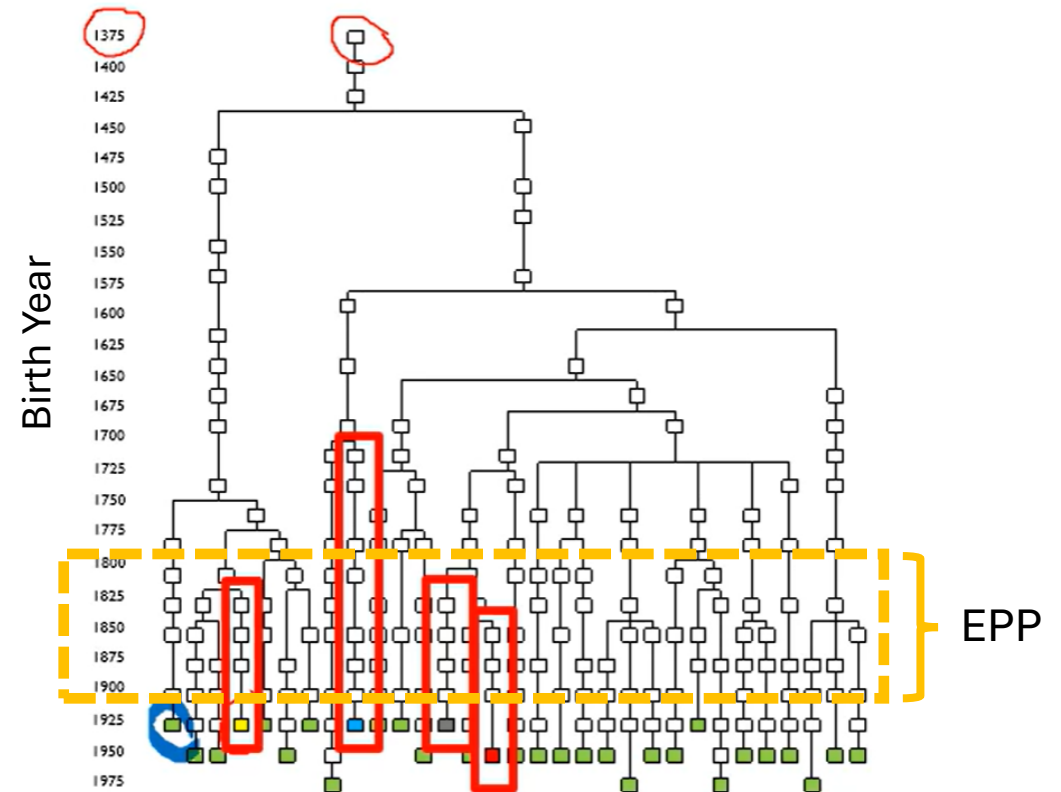
- An oft-repeated zombie statistic posits that as many as 10% of children are fathered outside of marriage
- In the 1991 book, *The Third Chimpanzee: The Evolution and Future of the Human Animal*, biologist Jared Diamond claimed the adultery rate among humans was between 5% and 30%.
- In a widely cited 1997 paper, University of Reading evolutionary biologist Mark Pagel argued EPP was so common in humans that babies evolved to be indistinguishable at birth, concealing their true paternity as a protective mechanism.
- Existing evidence was questionable. Estimates based on paternity tests are biased, as people paying for a test often already suspect Not Parent Expected (NPE) or Extra-Pair Paternity (EPP).

Source: Science: [https://www.cell.com/current-biology/fulltext/S0960-9822\(19\)31305-3](https://www.cell.com/current-biology/fulltext/S0960-9822(19)31305-3)

Large Single Patrilineal Tree – Founder born in the 14th Century



The colored boxes are living male descendants of 14th century their patriarch. **Green** matching Y-haplogroup. Other color don't match.



EPP occurred in the 19th Century or just before

How to estimate when the EPP/NPP Happened?

Different Y-DNA haplogroup verifies unrelated male ancestor.

- Male individuals who share the same Y-SNP (terminal SNP) also must share a common male lineage since the point when the SNP originated.

What proof can we use to prove when these EPP/NPP occurred?

- Autosomal DNA from descendants. Amount of shared cM can predict the range of possible relationships which narrows down when the EPP event occurred.

Private SNP becomes Public when found in at least two or more unrelated or distantly related individuals.

Testing Methodology - Expanded Study

- Collected family trees going back to the 1400s with the help of Belgian and Dutch genealogy enthusiasts. Then independently verified the trees.
- Identified thousands of men living today who, according to genealogical records, should all be distantly related on their father's side. Local history societies helped him contact candidates.
- The men were informed that he was looking for NPE in their ancestry, and any hesitation was enough to exclude them from the study. Didn't test siblings, or people with parents, grandparents, or great-grandparents in common, because the consequences of uncovering a painful family secret in the recent past are too high.
- When a family tree is accurate, the Y chromosomes will match.

Article: DOI: [10.1016/j.cub.2019.09.075](https://doi.org/10.1016/j.cub.2019.09.075)

Expand Study to Gather Statistically Significant Sample Size

- Expand study to of **513 genealogical pairs of males** sharing a male ancestor obtain in-depth genealogical **details of all 6,818 male *ancestors*** most were born *before* the introduction of modern contraception (median year of birth: 1840, quartiles (25 / 75%: 1762–1896, range 1315 - 1974),
- Each Y-chromosome mismatch provided genetic evidence of at least one case of **NPE/EPP**.
- Examined archival records to obtain **in-depth genealogical details** of all 6,818 male ancestors that occurred in our genealogies, including place and date of birth and, for the males born between 1750 and 1950, the occupation of the legal father.

Study Data – Analysis and Verification

- Verified in the genealogical records that the birth of each child in the patrilineages happened within wedlock and was declared officially by the father himself
 - The high quality of both the historical demographic data and the genealogical records available for our study area meant that we could not only reconstruct the mean historical EPP rate from these mismatches but also estimate EPP as a function of socio-demographic factors that are expected to influence its incidence.
 - For children born after the start of the civil records after 1800 or that there was clear genealogical evidence that the father was alive when the child was baptized
- Occupation of the father used to infer the socioeconomic status of the ancestors and linked place and date of birth to recorded (or estimated) historical population sizes and densities.

Y-Haplogroup Distribution

- Y DNA analysis: genotyped all of our contemporary male DNA donors at a panel of 191 Y chromosome **SNP** (Y-SNPs) and 38 Y chromosome **STR** (Y-STR) loci and used mismatches in the Y chromosomal haplotype of genealogically paternally related pairs of men as evidence for the occurrence of one or more EPP events within their genealogy.
- There were 299 **haplogroups** in the study along with sub-haplogroups.

Major Haplogroups

Haplogroup	Unbiased sample	
	<i>N</i>	<i>Freq</i>
A1	0	0.0
E1b1b	17	5.7
G2a	12	4.0
I1	39	13.0
I2	23	7.7
J1	5	1.7
J2	14	4.7
Q	1	0.3
R1a	10	3.3
R1b	175	58.5
L	2	0.7
T	1	0.3
Total	299	

EPP/NEP Results

- Historical EPP rates, while low overall, were strongly impacted by socioeconomic and demographic factors.
 - Specifically, we observe that estimated **EPP rates (1A)** among married couples varied by more than an order of magnitude, from 0.4% to 5.9%, and peaked among families with a low **socioeconomic background (1B)** living in **densely populated (1C)** cities of the late 19th century

Red dot = low income

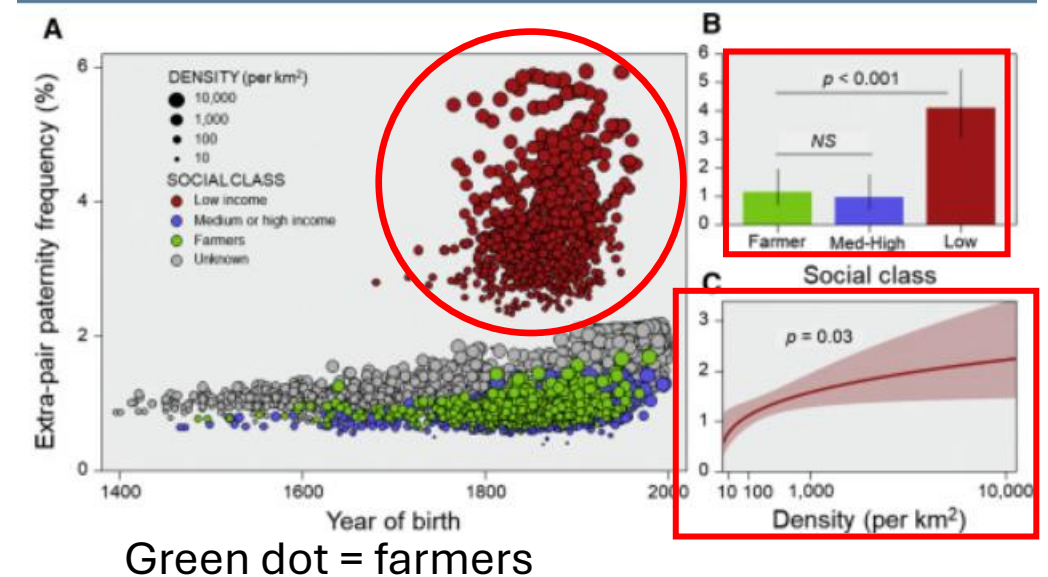
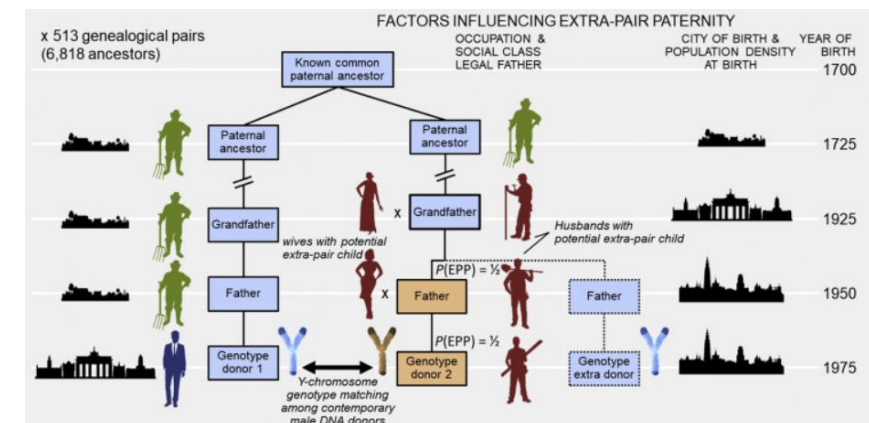


Figure 2 Context-Specific Variation in Human Extra-Pair Paternity



EPP/NPE Plotted on a Map vs. Year

A) Historical rate and (B) location of historical EPP rates plotted on map of study area

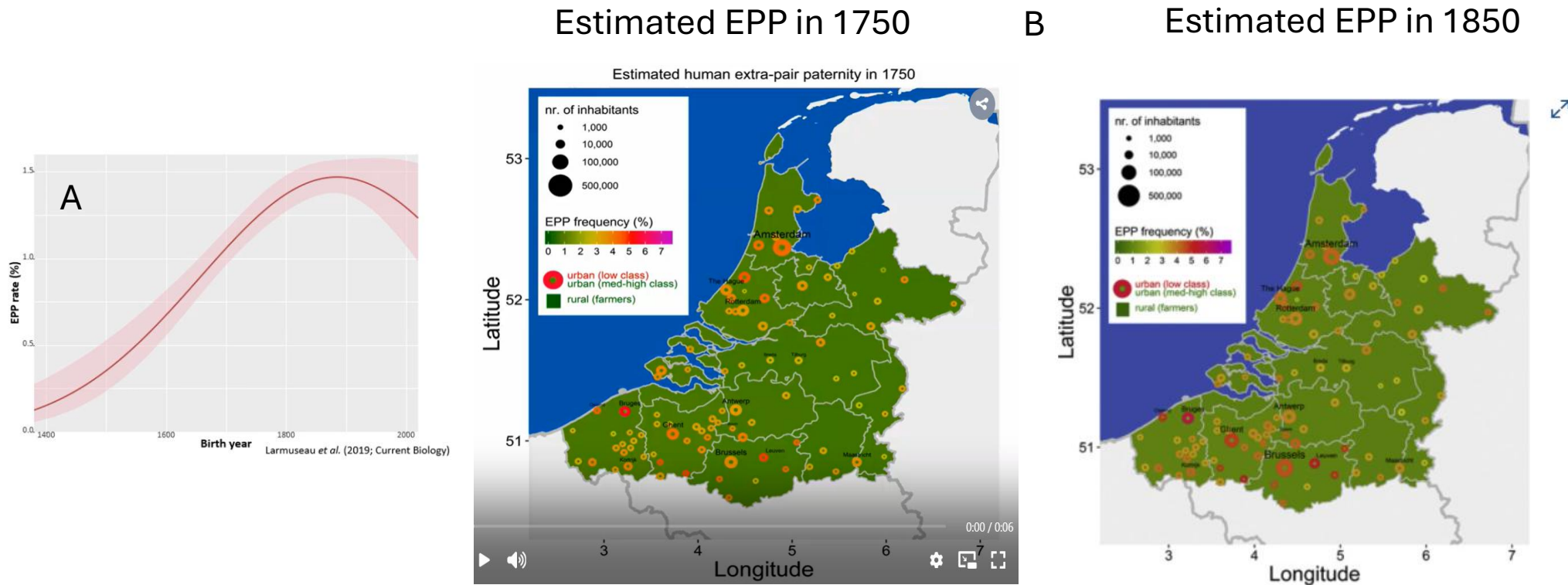


Figure 3 Estimated Extra-Pair Paternity Rates in the Low Countries around 1850

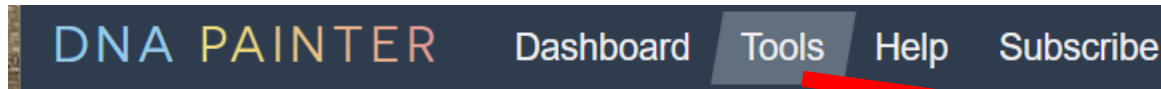
Video: <https://www.cell.com/cms/10.1016/j.cub.2019.09.075/attachment/c926f70f-06eb-48ad-a3f0-2b83552f83be/mmc3.mp4>

DNA Painter Recent Updates

Johnny Perl

- Matrix tool *NEW*
- Ancestral Trees - Updated
- WATO Plus *NEW*
- Chromosome Map features – Updated

DNA Painter – Matrix Tool



Untitled matrix: click here to add/edit title

Click to add notes

Welcome

Use this tool to create, view, and share a grid of autosomal testers. You can also add **relationships** to compare shared DNA with expected amounts for that relationship.

Adding data

- Click **Add tester** to add manually, or **Load** to import from a CSV or paste a Gedmatch/FTDNA matrix
- Once you've added more than one person, enter **shared DNA amounts** by clicking **Edit mode** or by clicking a name on the left and editing **Relatives and shared DNA**
- Relationships can be added manually or via CSV import

Sharing

- Click **Share** to generate a link; viewers will see initials by default for privacy
- You can also click **Export** to save the matrix as an image, CSV, or data file

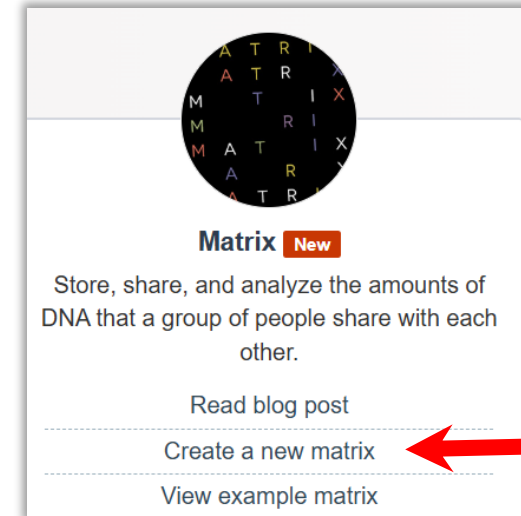
[See an example](#)

[Read blog post](#)

[Read new features blog post](#)

	EN	JN	RG	DG	WN	NH	DD	RD	FR	SD	IR
Edith	--	2802.9	875	781.1	2502	331.6	724.3	689.4	282.3	695.1	677.7
Jerry	2802.9	--	805.2	736.2	2871	396.8	772.5	625.5	396.5	776.3	736.5
Rudy	875	835.2	--	2669.4	834.3	418.5	747.6	639.9	443	894.6	724.4
Dan	781.1	736.2	2869.4	--	716.8	444.6	1131.6	913.8	424.3	1092.1	940.1
Wendy	2502	2871	834.3	716.8	--	355.7	640.3	752.6	347.3	809.2	676.8
Nathan	331.6	396.8	418.5	444.6	355.7	--	2098	1744.2	901.9	3567	1840.3
Don	724.3	772.5	747.6	1131.6	640.3	2098	--	2572.7	1755.3	2886.2	2712.8
Becky	689.4	825.5	639.9	913.8	752.6	1744.2	2572.7	--	1835.4	2561.4	2825
Pilon	282.3	346.5	443	424.3	347.3	901.9	1735.3	1835.4	--	1825.6	3377.8
Sally	695.1	776.3	894.6	1092.1	809.2	3567	2886.2	2581.4	1825.6	--	2781.5
Imogen	577.7	739.5	724.4	940.1	676.8	1840.3	2712.8	2825	3377.8	2781.5	--

DNA shared vs expected
Less More



- Use to create permanent matrix of cM shared among matching autosomal testers that you can share with others.
- Can import from a csv file
- Linked to cM tool

DNA Painter – Matrix Tool

EDIT MODE Click Save to confirm edits or cancel to revert them. Cancel Save

DNA Matrix tool beta [About this tool](#) [Webinar](#)

Click to add notes

Load Export Share Clear Edit mode Settings

- Click a name to edit a tester
- Add/Edit shared cM and relationships directly in the grid
- Drag :: to re-order testers
- Click ✕ to delete a match

	Robert	Stephen Ullrich	Kathy W	Mara F
Robert	--	enter shared cM + add relationship	enter shared cM + add relationship	enter shared cM + add relationship
Stephen Ullrich	enter shared cM + add relationship	--	enter shared cM + add relationship	enter shared cM + add relationship
Kathy W	enter shared cM + add relationship	enter shared cM + add relationship	--	enter shared cM + add relationship
Mara F	enter shared cM + add relationship	enter shared cM + add relationship	enter shared cM + add relationship	--

+ Add tester Cancel Save

Select 'Edit'.

- ① Add New tester
- ② Add amount of cM
- ③ Add relationship
- ④ Can Load a matrix

DNA Painter – Matrix Tool Features

Show **Shading**
 cM % shared DNA shared vs. expected SD from average

Sort: Shared DNA with Robert Ullrich ▼

	RU	SU	KW	MF	KL	WWD
R [redacted]	--	2945.6	2719.9	588.1	271.3	29.2
Stephen [redacted]		--	2698	779.4	223.8	32.9
K [redacted]			--	588.1	206.4	0
Mara [redacted]				--	289.7	12.4
Karl [redacted]					--	33
WHD194 [redacted]						--

+ Add tester

⑥ See next slide

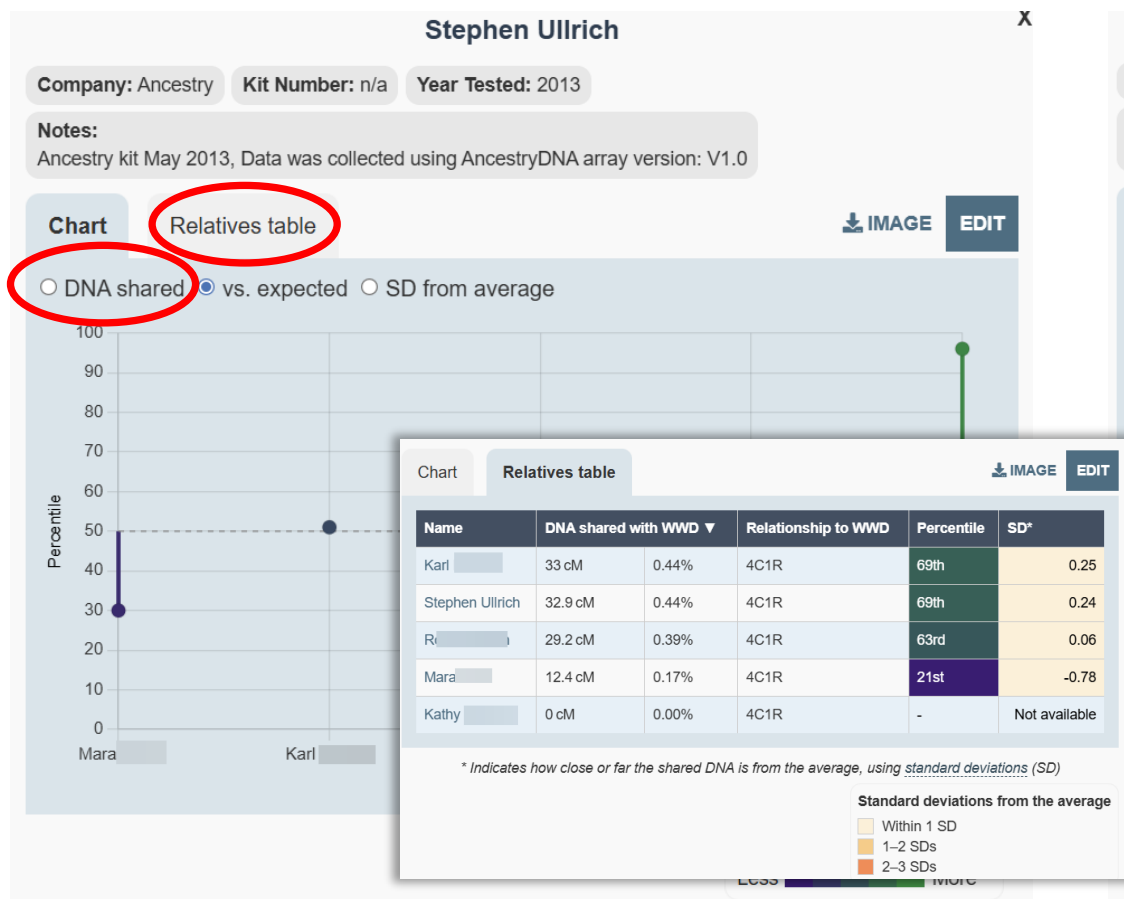
RU and KL share 271.3 cM
 (3.65%)
 ~ 72nd percentile for 2C
 • Within one sd of the average
[View in Shared cM Project](#)

Features

- ① Show ½ matrix
- ② Show initials
- ③ Options to display
- ④ Color intensity shows amount DNA shared
- ⑤ Click on box
- ⑥ Click on name: stats

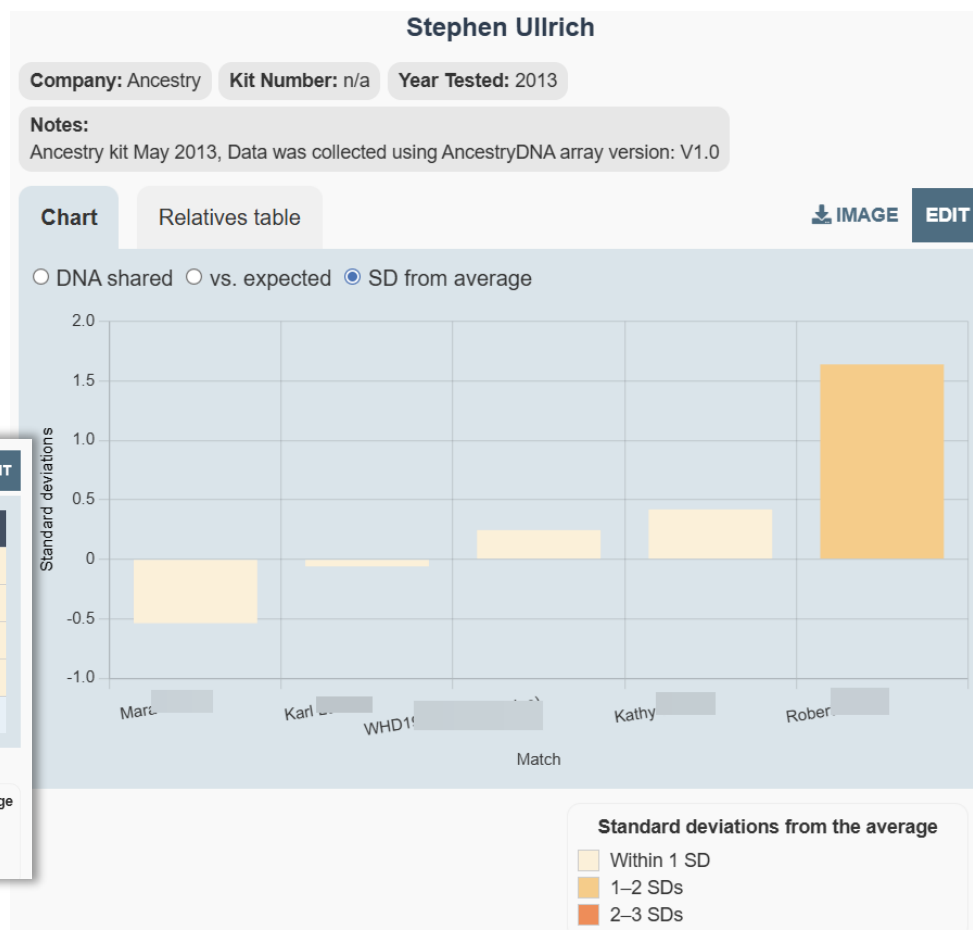
Matrix Tool Features: Match Statistics

① Percentile rank vs expected



DNA Shared option = cM shared; not shown.

② SD* from average



*±2 SD 95% Confidence Interval (normal distribution).
Relationships >>± 2SD might be a suspect relationship.

Chromosome Maps – Add Y- and mito DNA Information

DNA PAINTER Dashboard Tools Help Subscribe

ANCESTRAL TREES TOOLS AND WATO CHROMOSOME MAPS

22 X Y mt

Add Y-DNA information

Add mitochondrial DNA information

I-S8522 mitoYDNA Edit

HV1c5 mitoYDNA Edit

Scroll down chromosome map below Chrm 22 and X CHRM until you reach Y-DNA and mt (mitochondrial) DNA chromosome information. Click to enter your info.

Y AND MITOCHONDRIAL DNA X

All fields are optional [Blog post about this feature](#)

Y-DNA haplogroup

Please use same format as FTDNA (e.g. R-M269 as opposed to R1b1a1b)

Y-DNA kit number at MitoYDNA.org

Normally in the form T12345

Y-DNA Ancestor
Please select

Mitochondrial DNA haplogroup

Please use same format as FTDNA (e.g. a string beginning with a letter)

mtDNA kit number at MitoYDNA.org

Normally in the form T12345

Mitochondrial DNA ancestor
Please select

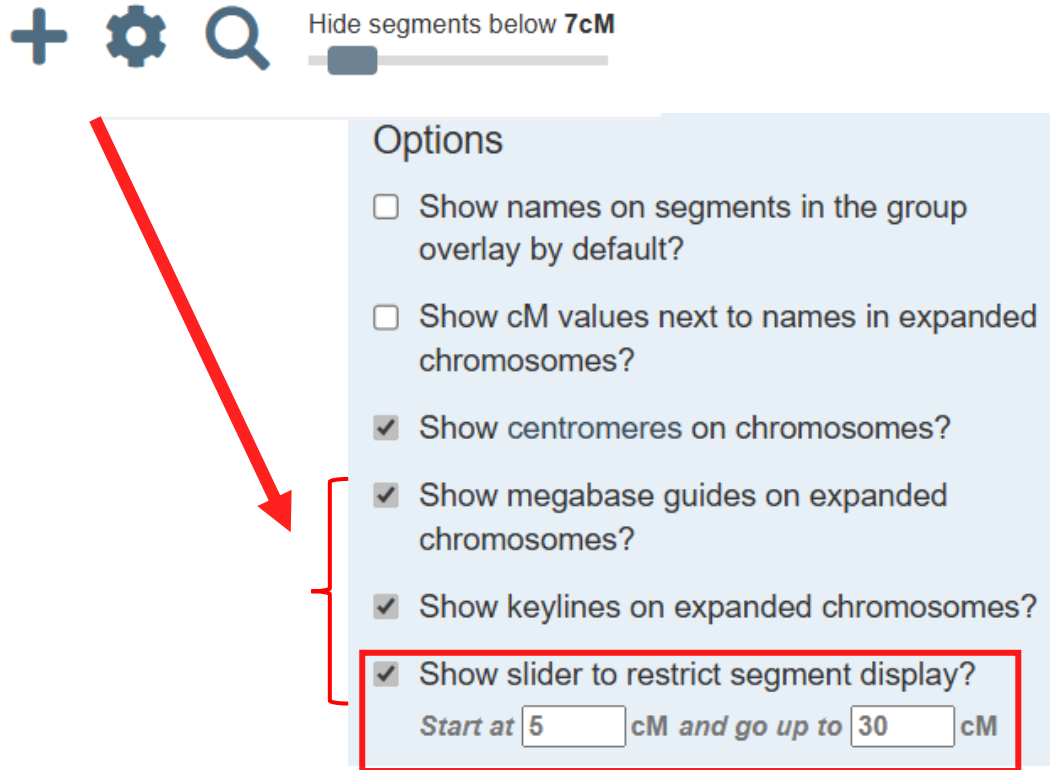
SAVE

Link to mitoYDNA kit Add from tree or add new.

DNA Painter Chromosome Maps Updates

Three new Options in controls on right side

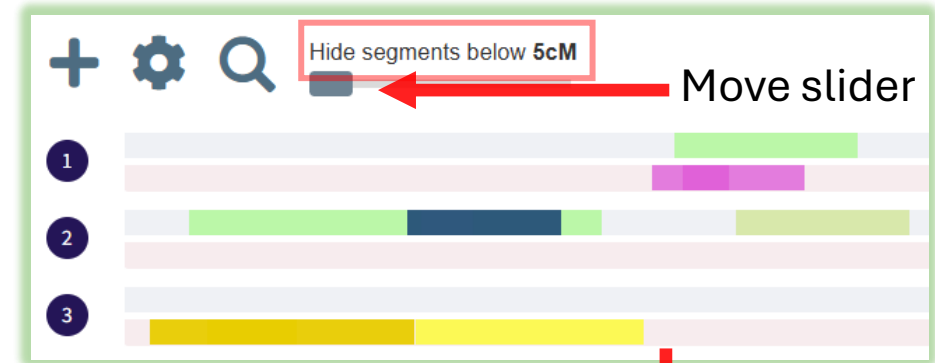
Slider allows removal of smaller segments from 5 to 30 cm.



Hide segments below 7cM

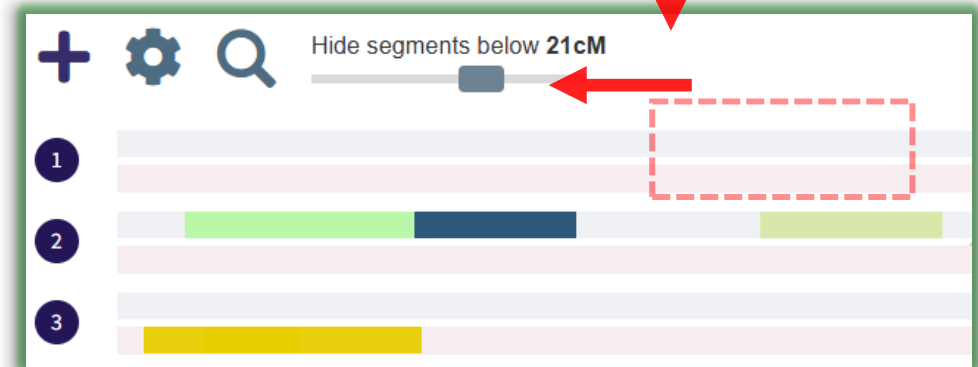
Options

- Show names on segments in the group overlay by default?
- Show cM values next to names in expanded chromosomes?
- Show centromeres on chromosomes?
- Show megabase guides on expanded chromosomes?
- Show keylines on expanded chromosomes?
- Show slider to restrict segment display?
Start at cM and go up to cM



Hide segments below 5cM

Move slider



Hide segments below 21cM

Gridlines and chrm bp position see next slide

DNA Painter Updates for Chromosome Maps

Next new Options in controls

Options

- Show names on segment overlay by default?
- Show cM values next to chromosomes?
- Show centromeres on chromosomes?
- Show megabase guides on expanded chromosomes?
- Show keylines on expanded chromosomes?
- Show slider to restrict segment display?
Start at cM and go up to cM

Expand/collapse all chromosomes. Click on a segment to

Hide segments below 5cM

Select (+) and the chromosomes expand showing gridlines and Mbp scale

SHARED OR BOTH Show match names Mass edit mode

PATERNAL

MATERNAL

24,283,768

Mara 7

Florren

Steven

110mb

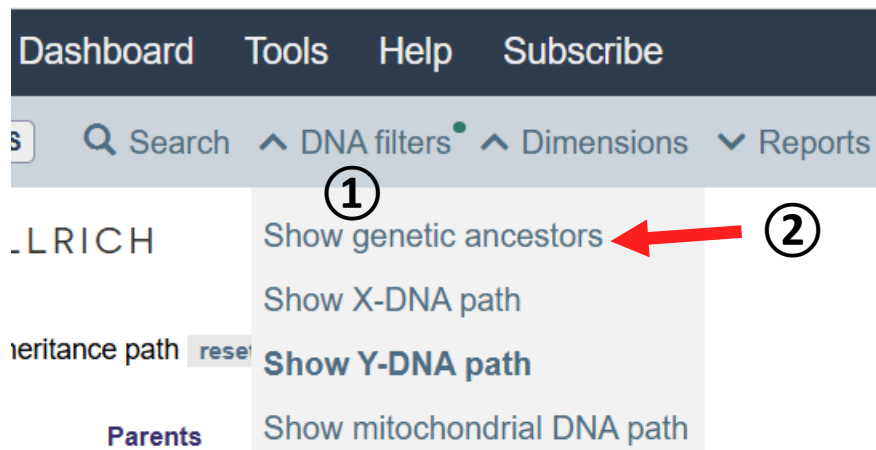
Hovering over a segment shows segment info

Option requires you to open chromosomes up

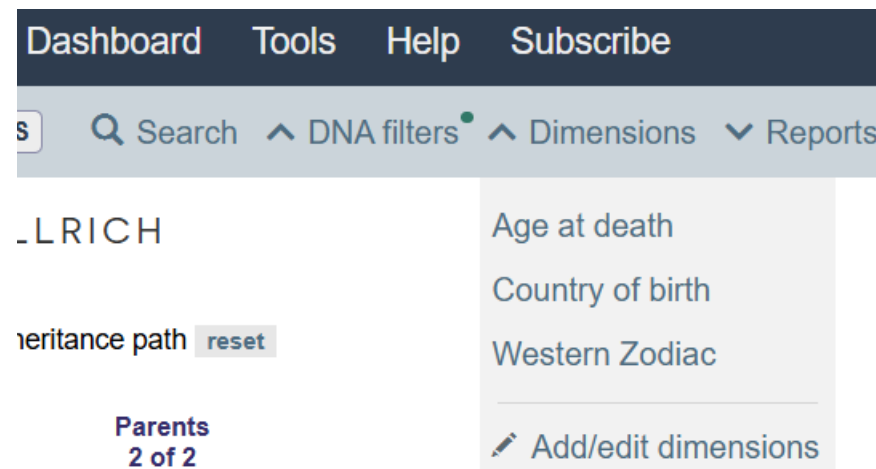
Keylines show exact bp position on the chromosome where the mouse is over.

DNA Painter Updates - Family Trees

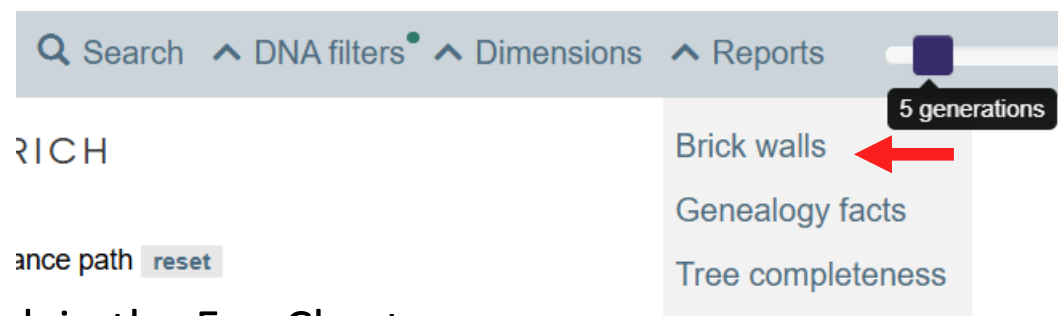
1. Select DNA filter



2. Select dimension (age at death, country of birth etc.)



3. Select type of report

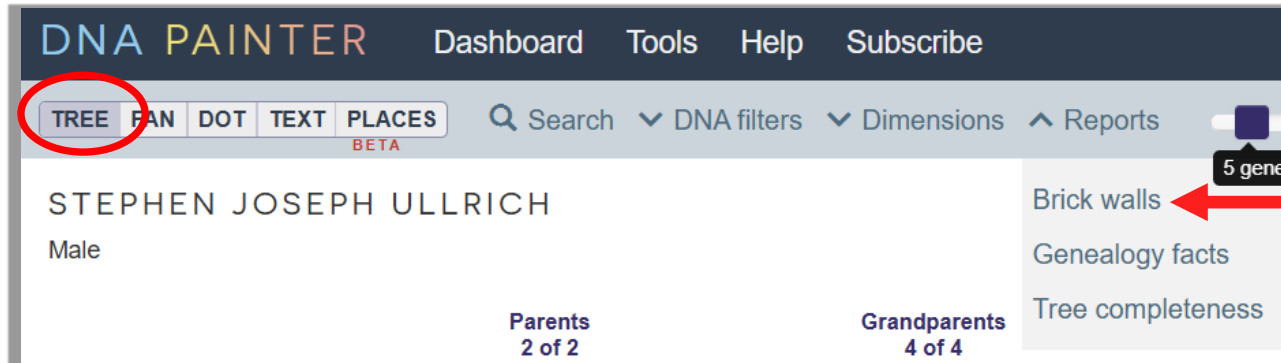


① Filters work in the Fan Charts

② Genetic ancestors: those painted/mapped onto a chromosome location

DNA Painter Updates

Brick Wall Report Listing



DNA PAINTER Dashboard Tools Help Subscribe

TREE FAN DOT TEXT PLACES BETA

Search DNA filters Dimensions Reports

STEPHEN JOSEPH ULLRICH Male

Parents 2 of 2 Grandparents 4 of 4

Brick walls
Genealogy facts
Tree completeness

In Tree view, select 'Reports'
From dropdown menu select 'Brick Walls'
and a list of four Brickwall.



Tools Help Subscribe

CSV file BRICK WALLS

The table below shows unknown parents of ancestors in this tree (most recent first).

Two most recent generations All brick walls

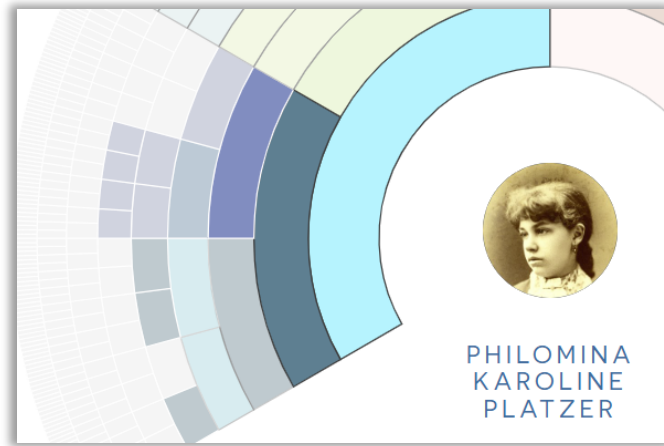
Generation	Brick wall
3rd-Great-Grandparent	The mother of Frederick H. Ullrich b. 1816 in Saxe-Coburg, Deutschland
3rd-Great-Grandparent	The parents of Johanna Wernly b. 1843 in Budweis, Böhmen, Kaisertum Österreich (Czech Republic)
3rd-Great-Grandparent	The mother of George Joseph Walker b. 1824 in Ireland
3rd-Great-Grandparent	The parents of Adalbert Biniak b. 1800 in Zempelburg, Westpreußen, Königreich Preußen (now Poland)
3rd-Great-Grandparent	The parents of Agnes Saydak b. 1805 in Zempelburg, Westpreußen, Königreich Preußen (now Poland)

DNA Painter Updates

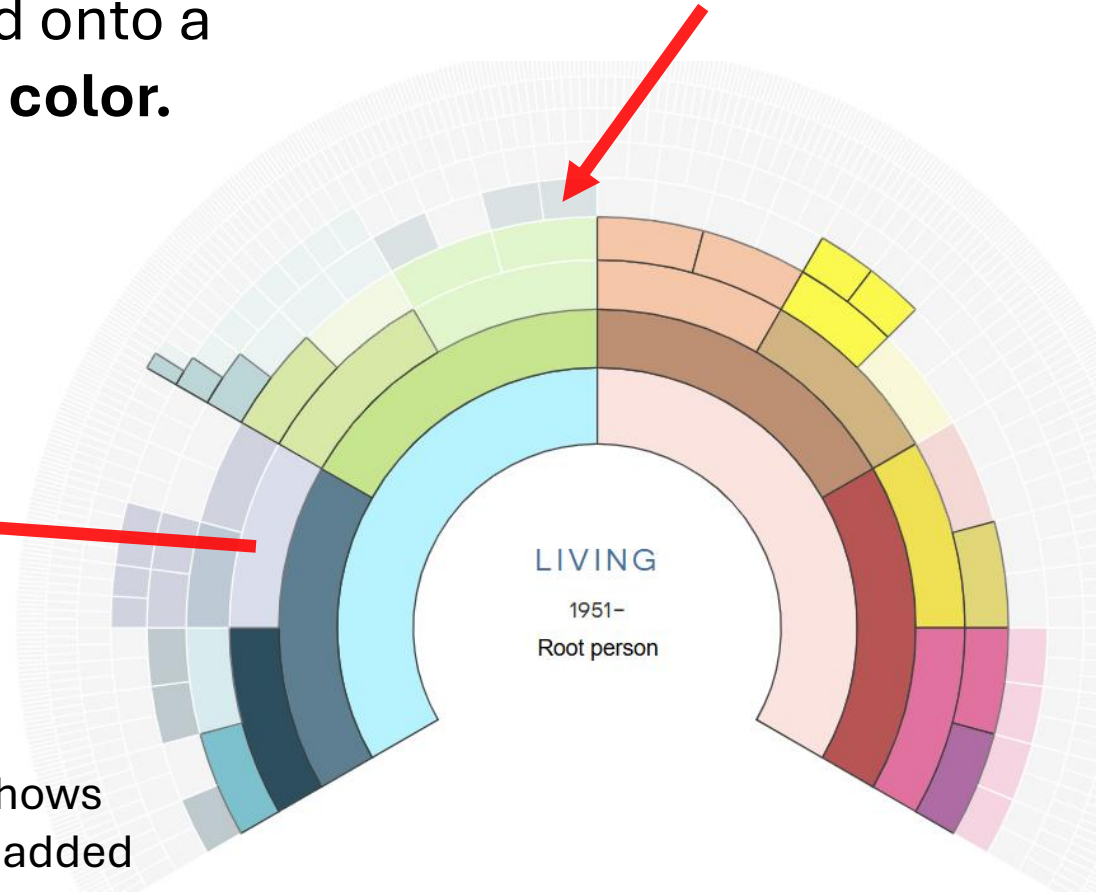
Genetic Ancestors View

Genetic ancestors are those painted onto a chromosome location are shown in **color**.

Darker grayed out areas are ancestors not yet Chromosome painted



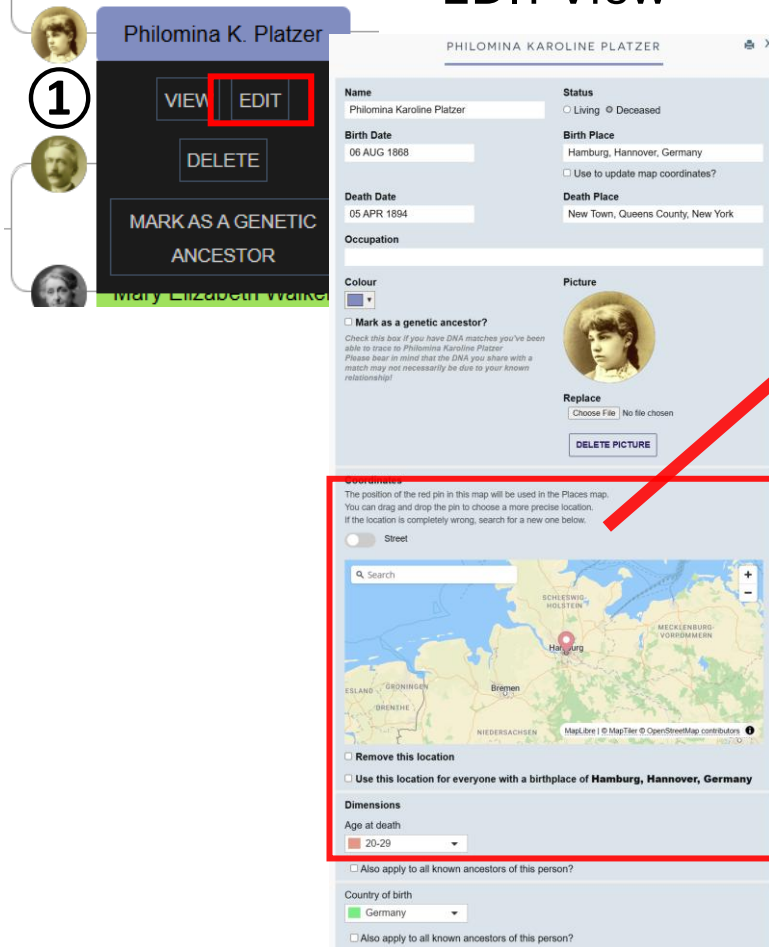
Hover over a person shows their name and image if added



DNA Painter Updates - Maps

EDIT View

From tree view hover over name of ancestor and click EDIT (1).
Scroll down to 'Coordinates'.



Philomina K. Platzer

VIEW EDIT

DELETE

MARK AS A GENETIC ANCESTOR

PHILOMINA KAROLINE PLATZER


Name: Philomina Karoline Platzer Status: Living Deceased

Birth Date: 06 AUG 1868 Birth Place: Hamburg, Hannover, Germany

Death Date: 05 APR 1894 Death Place: New Town, Queens County, New York

Occupation:

Colour:

Picture: 

Replace: Choose File | No file chosen

DELETE PICTURE

Mark as a genetic ancestor?

Coordinates

The position of the red pin in this map will be used in the Places map. You can drag and drop the pin to choose a more precise location. If the location is completely wrong, search for a new one below.

Street

Search:

Remove this location

Use this location for everyone with a birthplace of **Hamburg, Hannover, Germany**

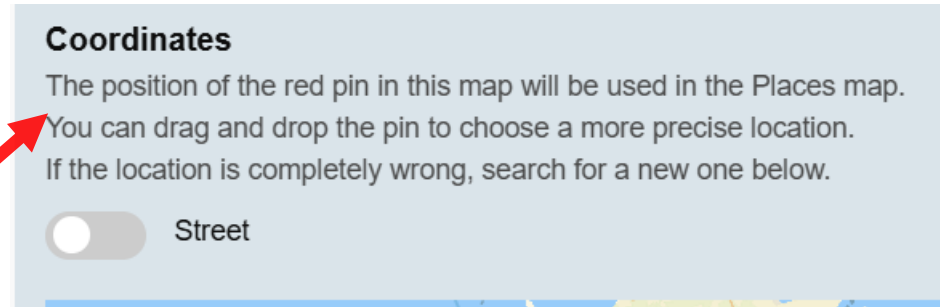
Dimensions

Age at death:

Also apply to all known ancestors of this person?

Country of birth:

Also apply to all known ancestors of this person?

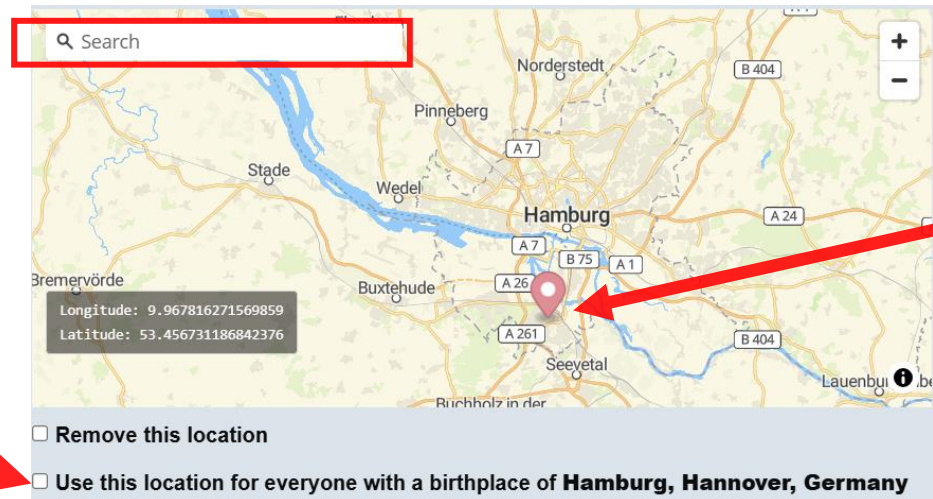


Coordinates

The position of the red pin in this map will be used in the Places map. You can drag and drop the pin to choose a more precise location. If the location is completely wrong, search for a new one below.

Street

Select street icon to map to a specific street. Move the red pin to the street address location or 'Search'. 'Save'.



Search:

Longitude: 9.967816271569859
Latitude: 53.456731186842376

Remove this location

Use this location for everyone with a birthplace of **Hamburg, Hannover, Germany**

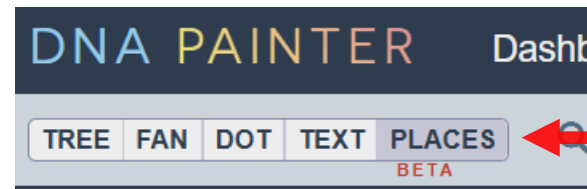
Marienstraße

Click here to apply location ALL people with same location.

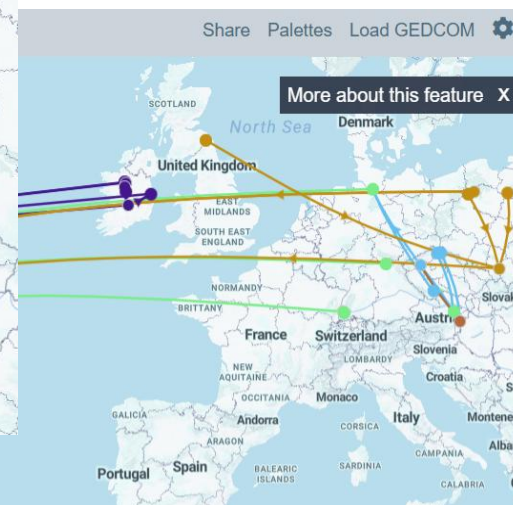
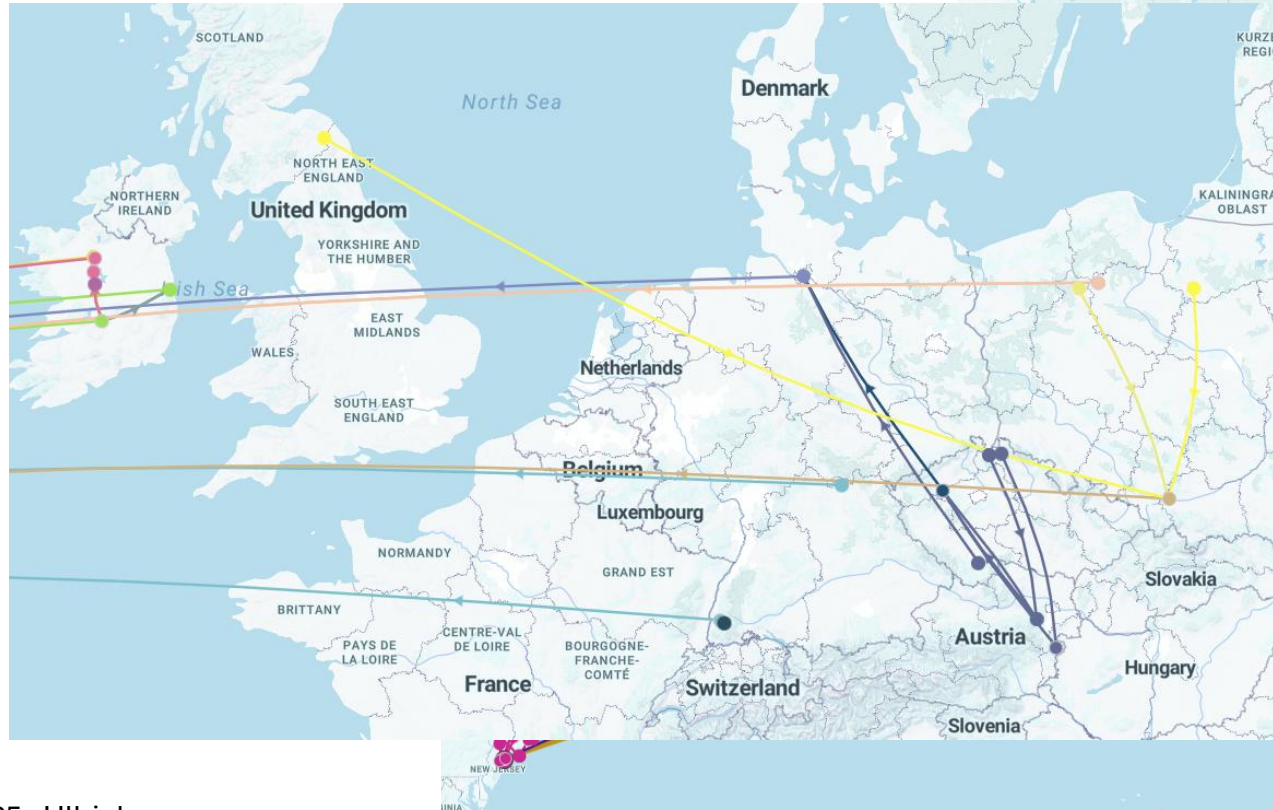
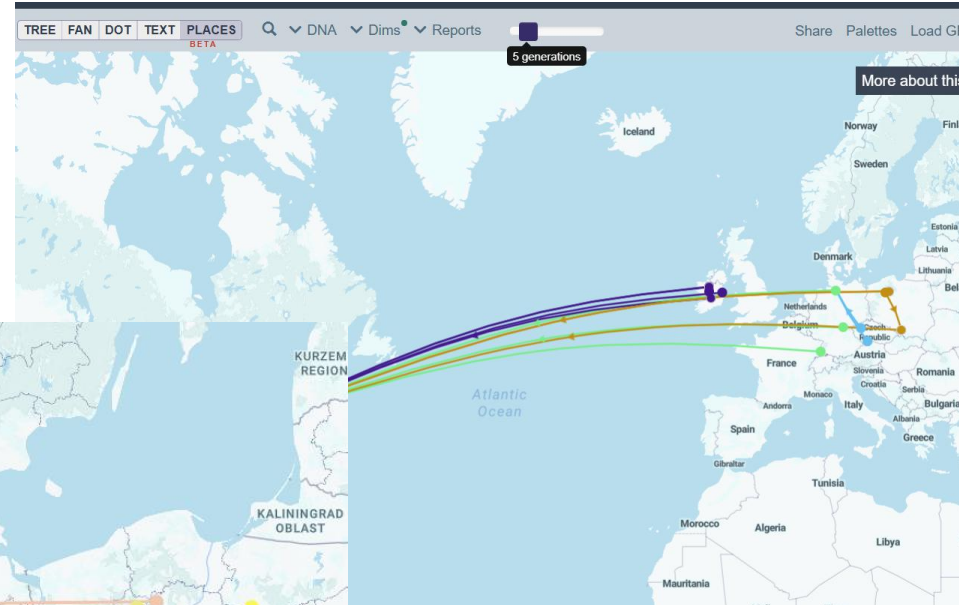
DNA Painter Updates

Map Ancestral Migrations

- Maps different ancestral lines with one location per generation



Open 'Places' to view



7 generations

DNA Painter Updates

New WATO Updates WATO Plus

WATO Plus - Easier to use
(requires subscription)

- New easier to use structured research question form. Input used in analysis.
- Lets you factor in the age of the unknown parent.

Research Question Form

WATO *plus* lets you use DNA matches to help figure out where you might fit into their tree [read more](#)

I would like to identify the biological **Father** of **Paul's mother**

born in the year **1948**

using the DNA Matches of **Paul's mother's child**

whose name is **Paul**

► **Optional info**

SAVE

Screenshot of the research question form

WATO Plus Guide: <https://dnapainter.com/help/user-guide/wato-plus>

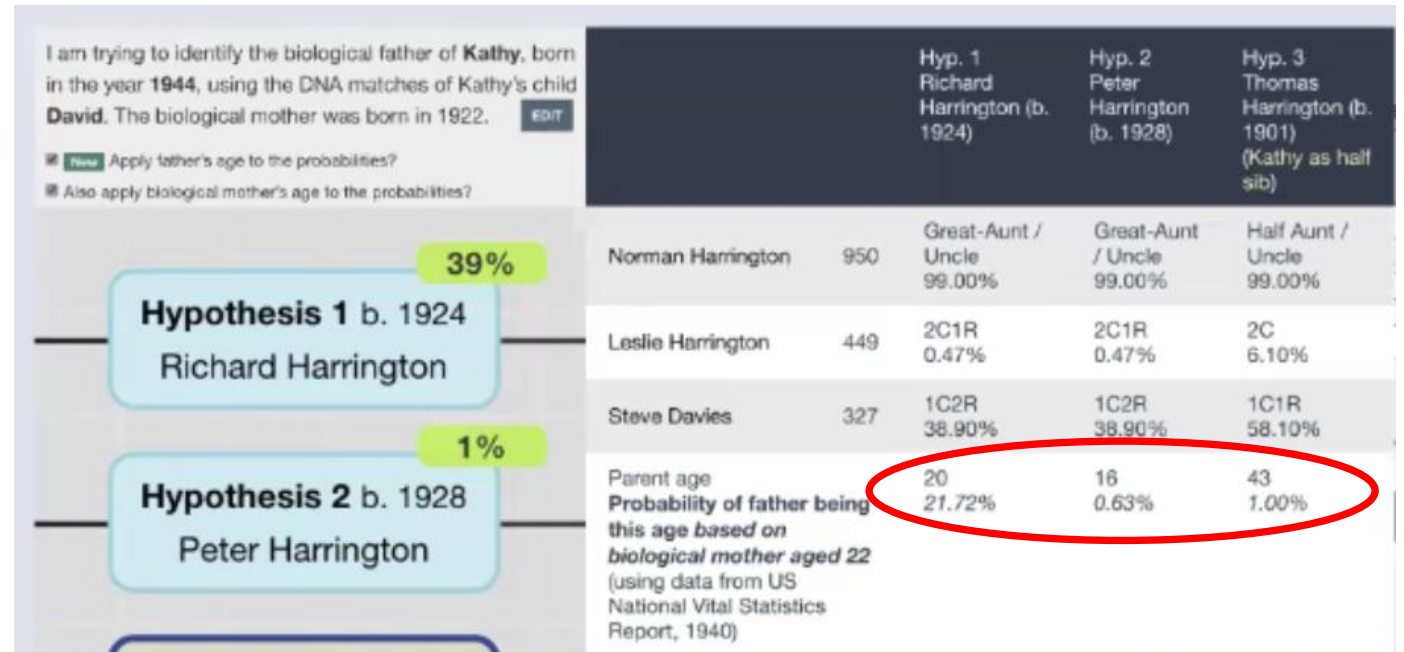
WATO Facebook Group: <https://www.facebook.com/groups/244344539465504>

DNA Painter WATO Plus

Research Question: find father for Kathy born 1944 using DNA matches of Kathy's child, David.

Uses aged based probabilities derived from birth statistics.

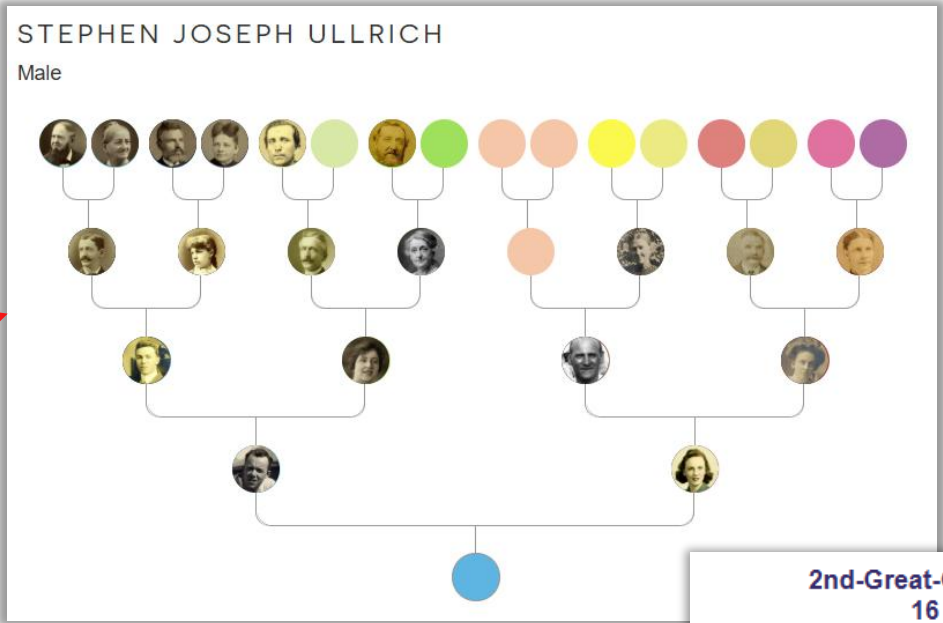
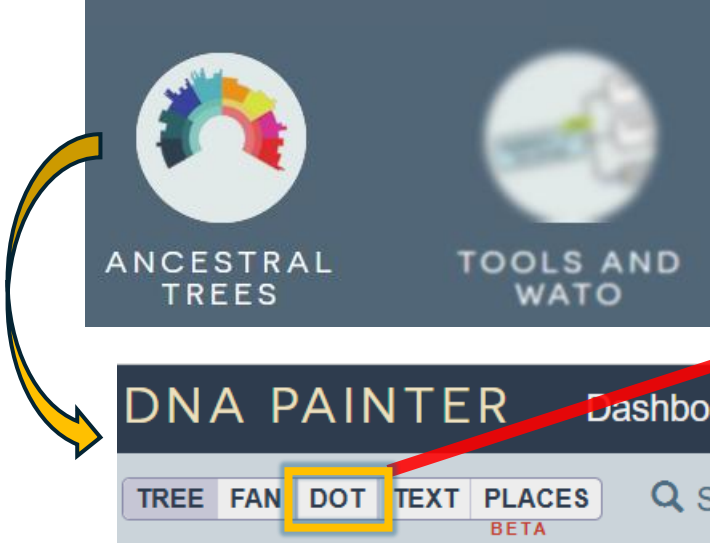
Good for ruling people out and finding who to test.



Uses 1940 USA Birth Data to calculate probabilities.

DNA Painter: New Tree Tool to Display Photos

Open, Ancestral trees; select 'DOT'



2nd-Great-Grandparents
16 of 16

Frederick H. Ullrich
Anna Maria Burger

A screenshot of a pop-up window showing the 2nd-great-grandparents of the subject. It displays two photos: a man in a suit (Frederick H. Ullrich) and a woman in a light-colored dress (Anna Maria Burger).

Need to upload ancestor's photo. From Tree view hover over name of ancestor and click EDIT. Find 'Picture' option and upload a file. After upload and select the face within the picture to display.

Picture

Choose File No file chosen

A screenshot of the 'Picture' upload interface. It features a 'Choose File' button and the text 'No file chosen'.

Can also have photos showing in trees

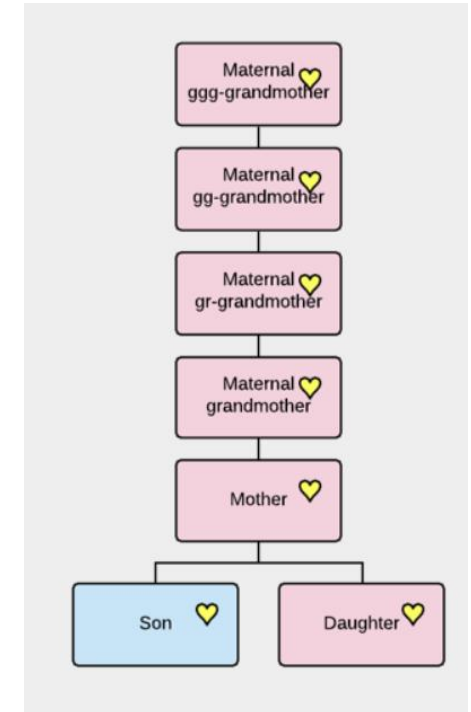
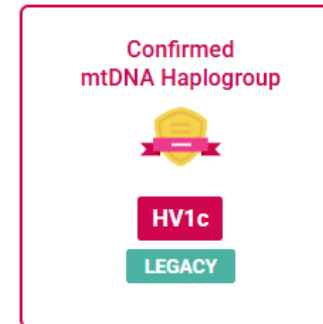
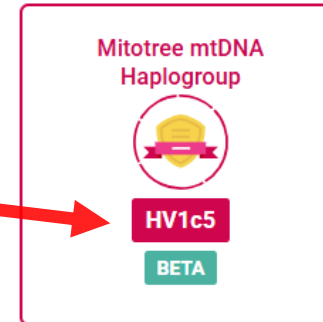
DISPLAY OPTIONS

Names and pictures
 Names and dates

A screenshot of the 'DISPLAY OPTIONS' menu. It has two radio button options: 'Names and pictures' (which is selected) and 'Names and dates'.

FTDNA Mitochondrial DNA – Revised tree and new features

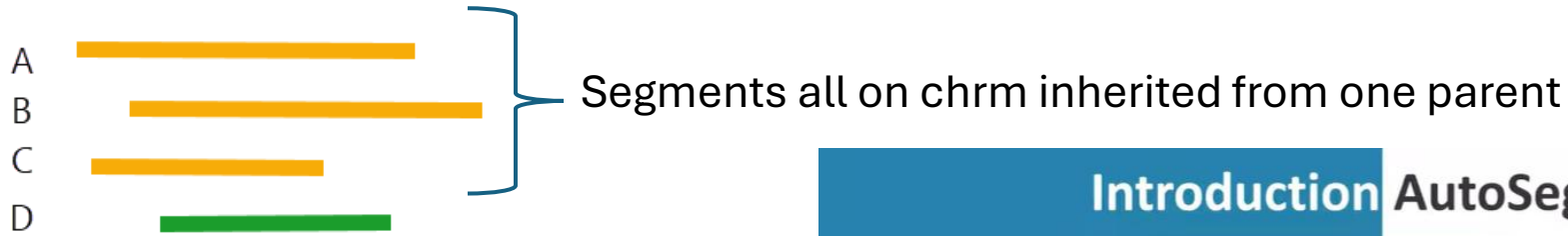
- Newly updated mito tree
- Mitochondrial DNA's strengths is that it can reach far back in time
- MitoDNA Beta:
 - Over 40,000 branches
 - Over 250,000 mtFull Sequences from FTDNA
 - Over 10,000 third-party full sequences from GenBank, 1000 Genomes, etc.
 - Over 1000 Ancient Connections
 - Over 100 Notable Connections



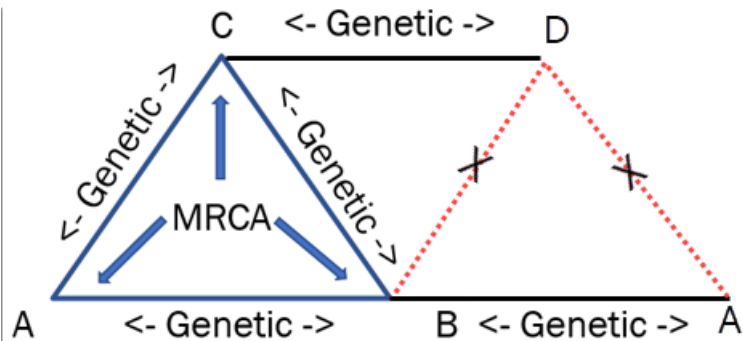
Blog: <https://dna-explained.com/2025/09/02/mitochondrial-dna-a-z-a-step-by-step-guide-to-matches-mitotree-and-mtdna-discover/>
<https://dna-explained.com/2025/02/25/mitotree-is-born/>

AutoSegment Split - GEDmatch's Latest Tier 1 Tool

Based on non-triangulating segments



Cluster A, B, C, D match on same Chrm segment but D only matches C. D is non-triangulating with A, B & C.



Introduction AutoSegment Split

- **Triangulating segments:** overlapping segments on the same chromosome on the same side
 - are shared by all DNA Matches (at least three or more individuals, e.g., yourself and two DNA matches)
 - triangulated segments are likely to be inherited from a common ancestor
- **Non-triangulating segments:** overlapping segments on the same chromosome but on opposite sides
- Example: the DNA segments from **John** and **Robert**
- The DNA segments from **John** & **Robert** do not match:
 - John and Robert are related on *opposite parent sides*
 - Not known if maternal or paternal side

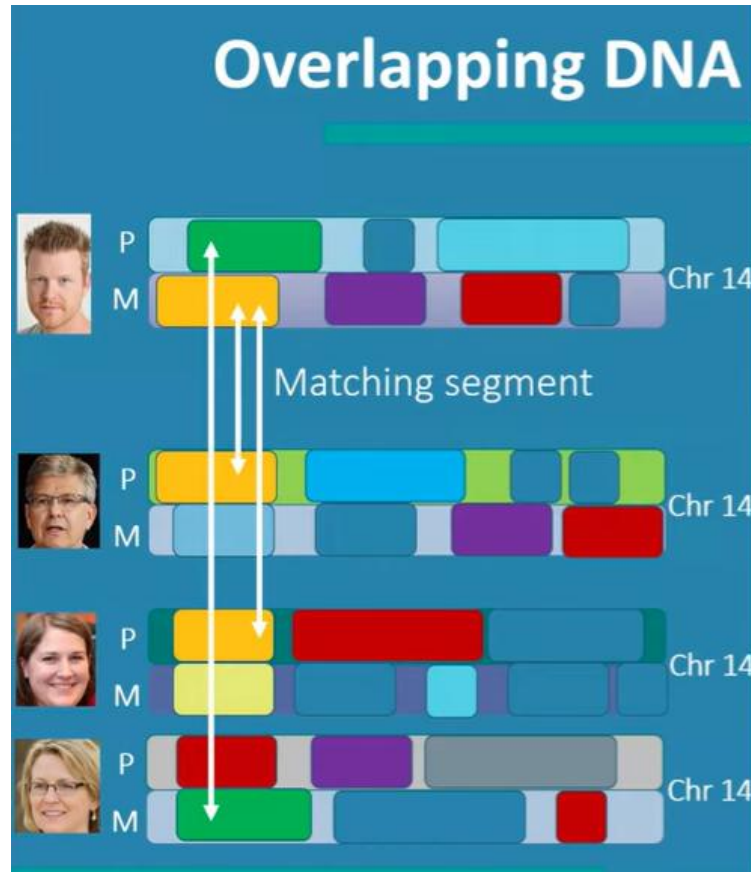
2

For same overlapping segments, non-triangulating and triangulating segments much match opposite parents.

Link to presentation by Evert-Jan Blom: [AutoSegment Split - GEDmatch's Latest Tier 1 Tool](#)

AutoSegment Split - GEDmatch's Latest Tier 1 Tool

Based on non-triangulating segments



- Chromosome 14 representation

- Matching segment with match:



- Matching segment with match:



- Matching segment with match:



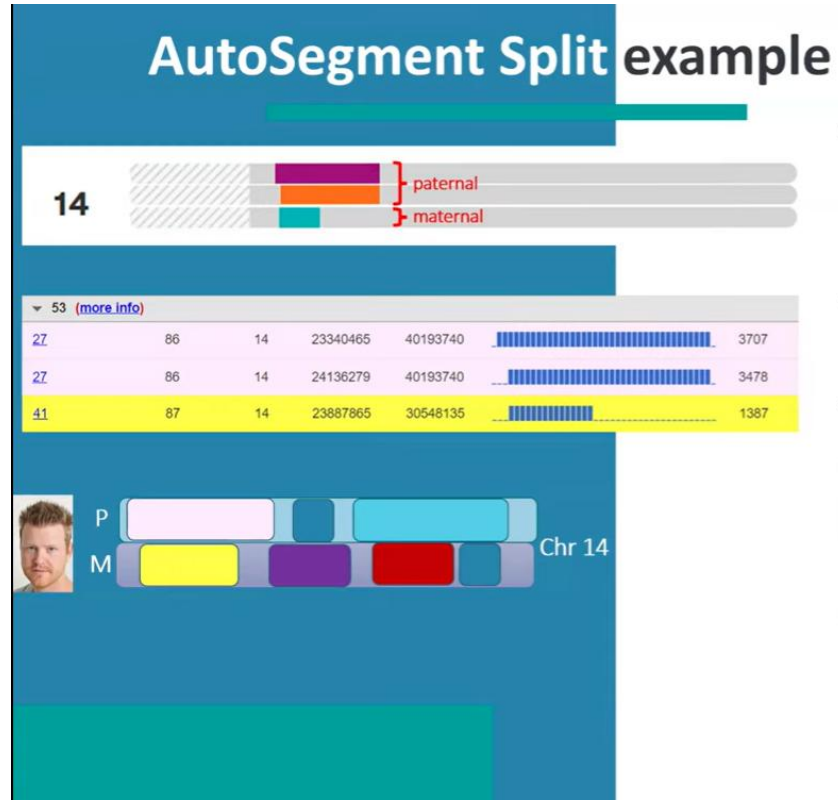
- Triangulating segments on one side:



Link to presentation by Evert-Jan Blom: [AutoSegment Split - GEDmatch's Latest Tier 1 Tool](#)

AutoSegment Split - GEDmatch's Latest Tier 1 Tool

Triangulation matrix



- Overlapping segments on chr 14
 - 2 of 3 paternal
 - 1 of 3 maternal
- Segments linked to both parents
- Triangulation matrix



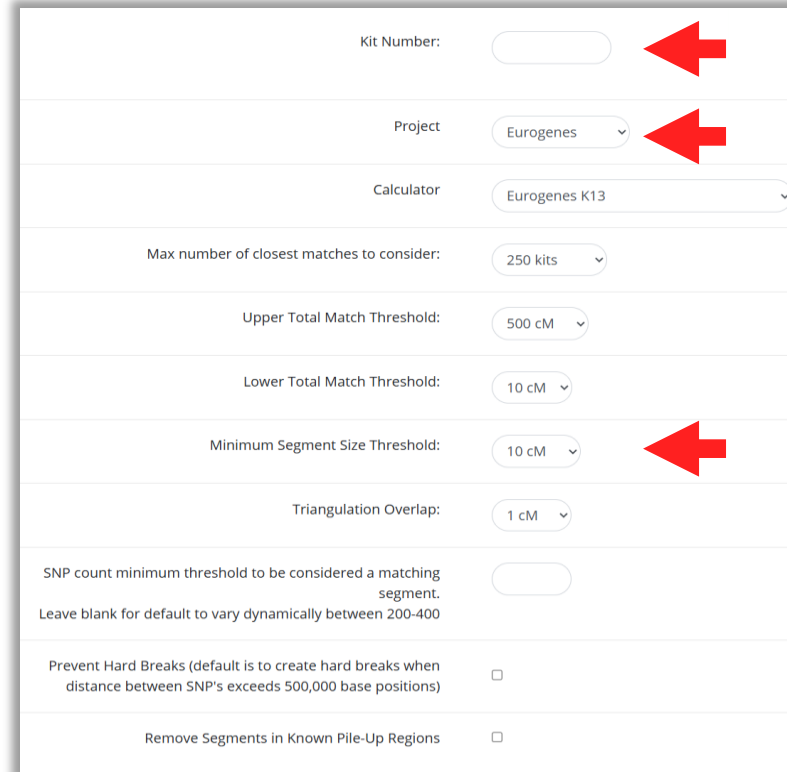
Link to presentation by Evert-Jan Blom: [AutoSegment Split - GEDmatch's Latest Tier 1 Tool](#)

GEDmatch Autosegment Split

➤ Tier 1 tool



Input kit and other parameters



A screenshot of the GEDmatch Autosegment Split configuration form. The form contains the following fields and options:

- Kit Number: (Red arrow pointing to the input field)
- Project: Eurogenes (Red arrow pointing to the dropdown menu)
- Calculator: Eurogenes K13 (dropdown menu)
- Max number of closest matches to consider: 250 kits (dropdown menu)
- Upper Total Match Threshold: 500 cM (dropdown menu)
- Lower Total Match Threshold: 10 cM (dropdown menu)
- Minimum Segment Size Threshold: 10 cM (dropdown menu) (Red arrow pointing to the dropdown menu)
- Triangulation Overlap: 1 cM (dropdown menu)
- SNP count minimum threshold to be considered a matching segment. Leave blank for default to vary dynamically between 200-400 (input field)
- Prevent Hard Breaks (default is to create hard breaks when distance between SNP's exceeds 500,000 base positions)
- Remove Segments in Known Pile-Up Regions

Insert kit number

Eurogenes Admixture
Select other
depending on
ethnicity

Partitions your DNA segments into parental sides AND provides admixture results for each segment on every chromosome. allows users to understand the genetic contributions from different ancestral populations across distinct regions of their genome, revealing how diverse ancestries are distributed along their chromosomes.

AutoSegment Split - Results

Results for cluster on Chr 5

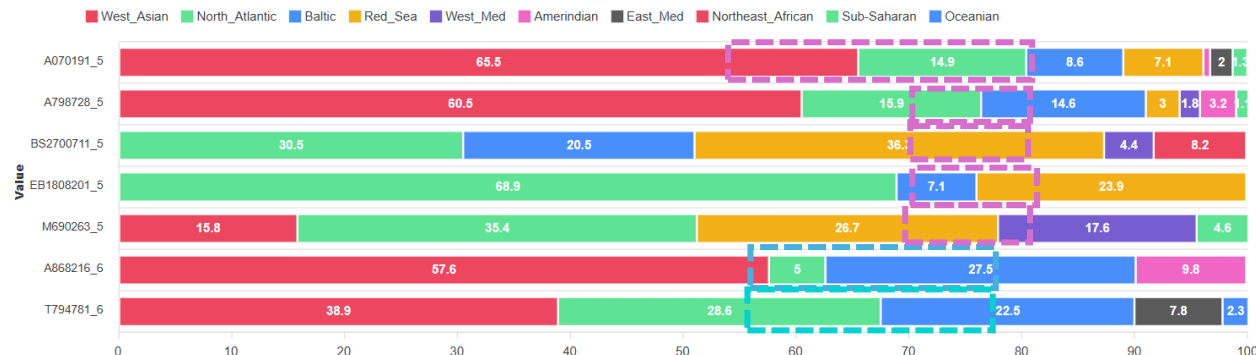
Chr	Start	Stop	Segment representation	SN...	Ad...	Match
5	52978722	82127629		4139	1828	*Bull
5	53068791	82127629		4132	1824	*7
5	75226673	82955163		1427	223	George f
5	74817394	82886028		1472	226	Sean n
5	75302307	82372665		979	514	Teena ,,,
5	55444040	78150147		3081	1371	Marc
5	54324622	78106062		4796	1478	*Jim

If you know how related to someone in the cluster can assign whether paternal or maternal side

One side

Opposite side

Admixture results using Eurogenes+K13 and 13 populations



Admixture analysis Shows different ancestral populations.



AutoSegment Split - GEDmatch's Latest Tier 1 Tool

Options and Considerations

- Admixture file to use
- Overlap segment threshold 5 cM best
- Number of matches to analyze top 100 to 150
- Endogamy may not work as often related from both sides

Link to presentation by Evert-Jan Blom: [AutoSegment Split - GEDmatch's Latest Tier 1 Tool](#)

Whole Genome Sequencing

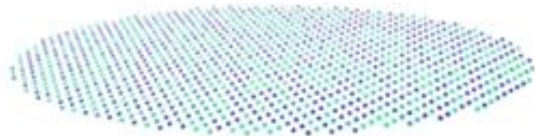
“Generations” of DNA sequencing



1986

GEN 1 Clonal, single reaction chain termination, fragment separation

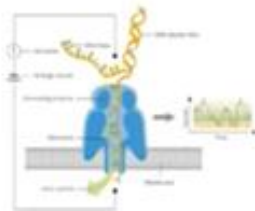
1st Generation Scale:
96 DNA clones per run



2005

GEN 2 Array SBX (Clonal → Stepwise)
aka Next Generation Sequencing
>90% of sequencing market

2nd Generation Scale:
multiplex runs



2010

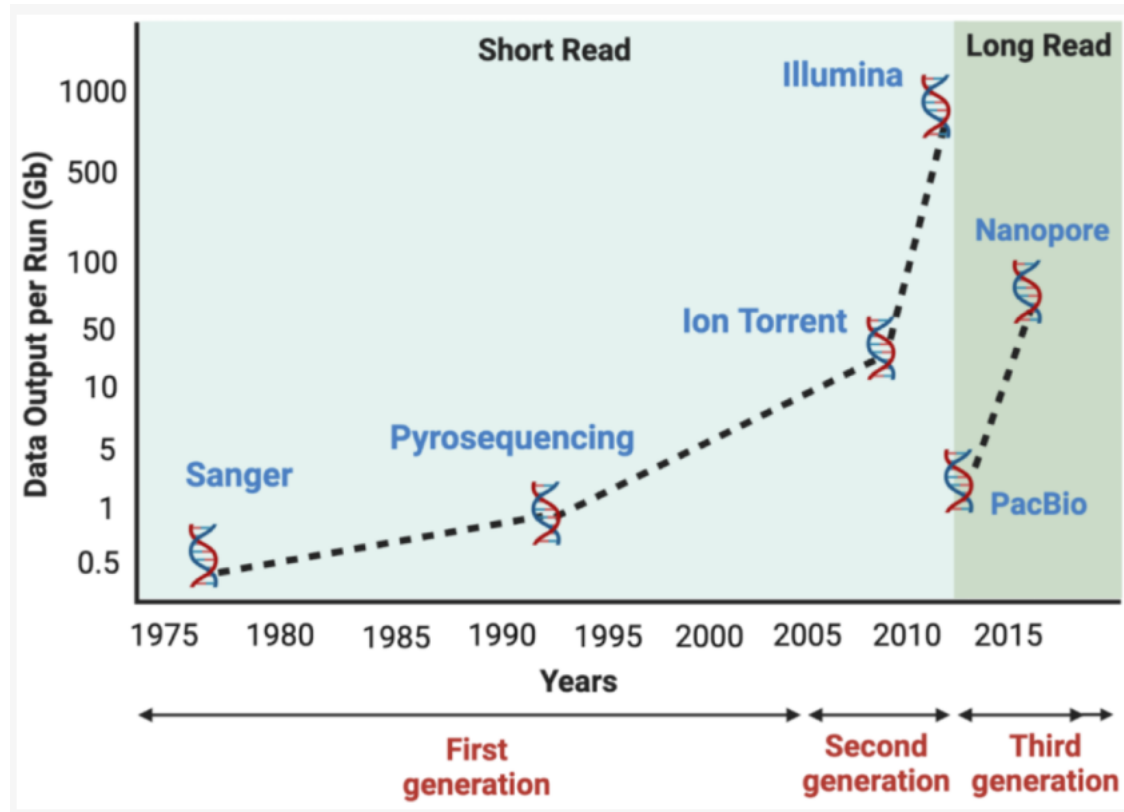
GEN 3 Single molecule, stochastic

3rd Generation Scale:
single DNA molecule
reads, longer length
reads

ULTIMA GENOMICS

6

Evolution of DNA Sequencing

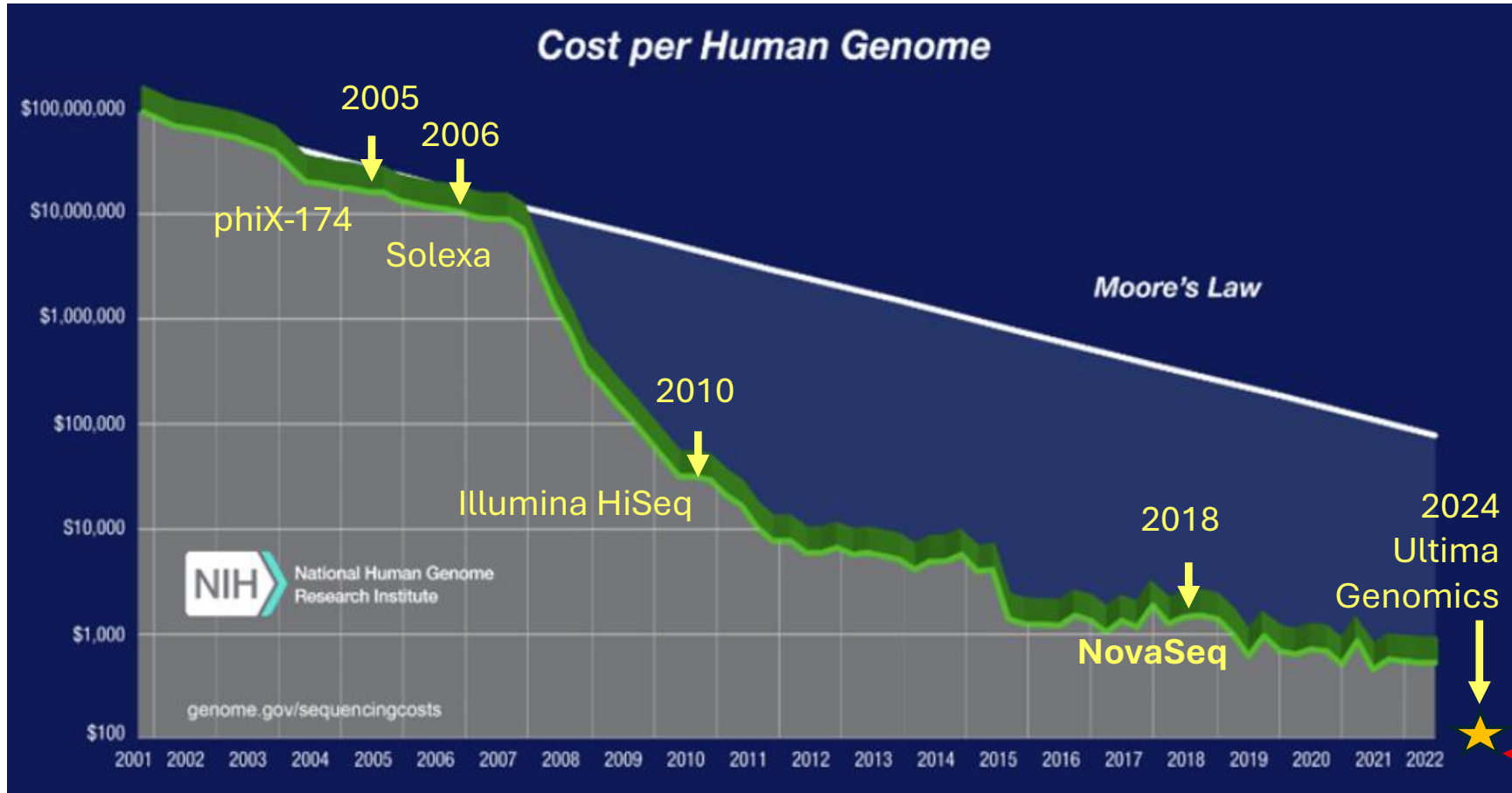


Pyrosequencing:

<https://geneticeducation.co.in/pyrosequencing-principle-process-advantages-and-limitations/#Limitations>

Next-Generation Sequencing Technology, 2023. <https://www.mdpi.com/2386720>

Cost of Genome per year vs. Moore's Law



Moore's Law number of transistors in an integrated circuit (IC) doubles about every two years.

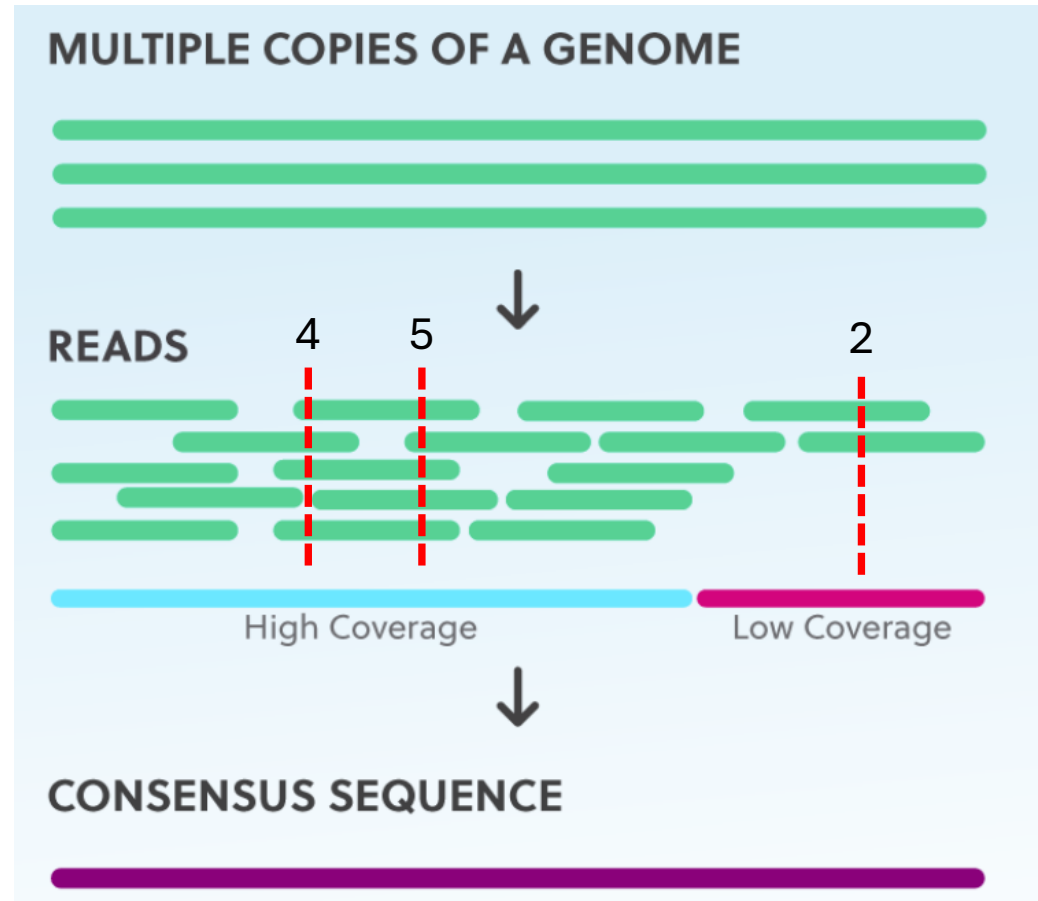
Solex GA: 10^6 sequences and NovaSeq: 20×10^9 reads, 6Tb of data simultaneously

NIH Genome Research Institute info: <https://www.genome.gov/about-genomics/fact-sheets/DNA-Sequencing-Costs-Data>

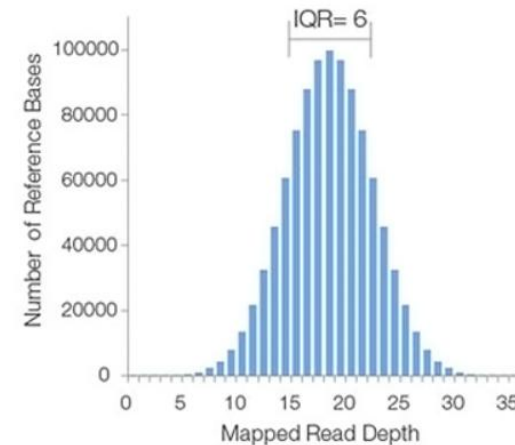
What is DNA Coverage?

- Coverage refers to how many times the genome was sequenced. Average number of reads that align to a known reference at a particular location within the target transcript or genome
- Best 30X to 50X for human WGS (for Illumina sequencer)
- Some NGS requires more coverage or over-sequencing to overcome low capture rates of duplex molecules mandating a reliance on multiple supporting fragments for a variant to be confidently called as a mutation (i.e., multiple independent sequencing of the genome). Error rate 10^{-2} and 10^{-3}
- ppmSeq™ (new MyHeritage WGS) allows very low coverage to get reliable data because it has accuracy of one part-per-million or better for calling SNVs (SNPs)

WGS – NGS Coverage



Coverage is an average number for the total genome



General equation is: $C = LN / G$
C stands for coverage
G is the haploid genome length
L is the read length
N is the number of reads

Coverage: www.illumina.com/

<https://irepertoire.com/ngs-considerations-coverage-read-length-multiplexing/>
https://support.illumina.com/downloads/sequencing_coverage_calculator.html

MyHeritage DNA test Upgrades to Whole-genome Sequencing (WGS)

- Sequencing is being done in the Gene by Gene laboratories using technology powered by Ultima Genomics.
 - Cost down to \$80/genome \approx to chip technology.
- Low-coverage 2x sequencing. Coverage refers to the number of times a base is read by the sequencing machine.
- Clinical-grade sequencing, where the goal is to detect rare variants, is normally done at 30 x or higher. FamilyTreeDNA's [BigY test](#) has a [read-depth of 70 x](#).
- **“Paired Plus-Minus Sequencing”**, aka ppmSeq, and approach that takes advantage of the system’s clonal amplification chemistry to identify and exclude sequencing errors due to DNA damage. With ppmSeq, sequencing errors are reduced to one **part per million** or better.

“You can buy the kit now, but only those arriving at the lab in Jan 2026 will be warranted to have WGS.” If you order a kit, I would therefore recommend delaying its return to ensure you get the WGS test.

Results downloaded as: Compressed Reference-oriented Alignment Map (CRAM)

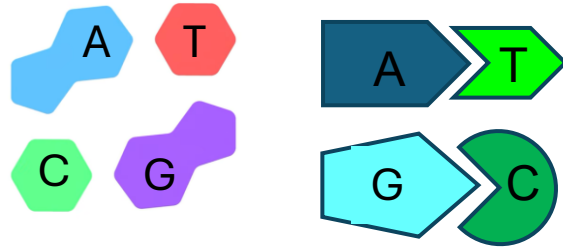
Blog: <https://cruwys.blogspot.com/2025/10/myheritage-upgrades-its-consumer-dna.html>

CRAM file tutorial: <https://www.youtube.com/watch?v=mMr26JG6SCI>

DNA consists of 4 Nucleotide Bases: A, T, C, G

Sequence conveys genetic information

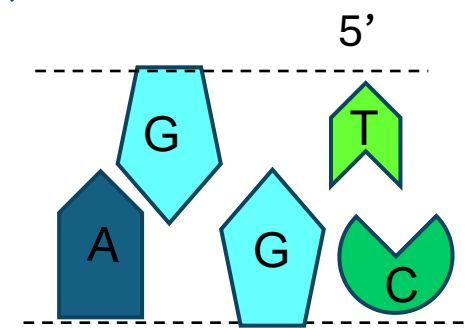
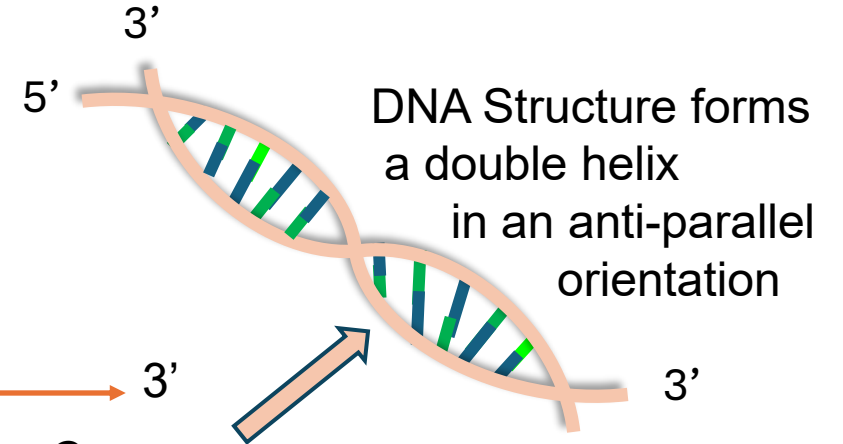
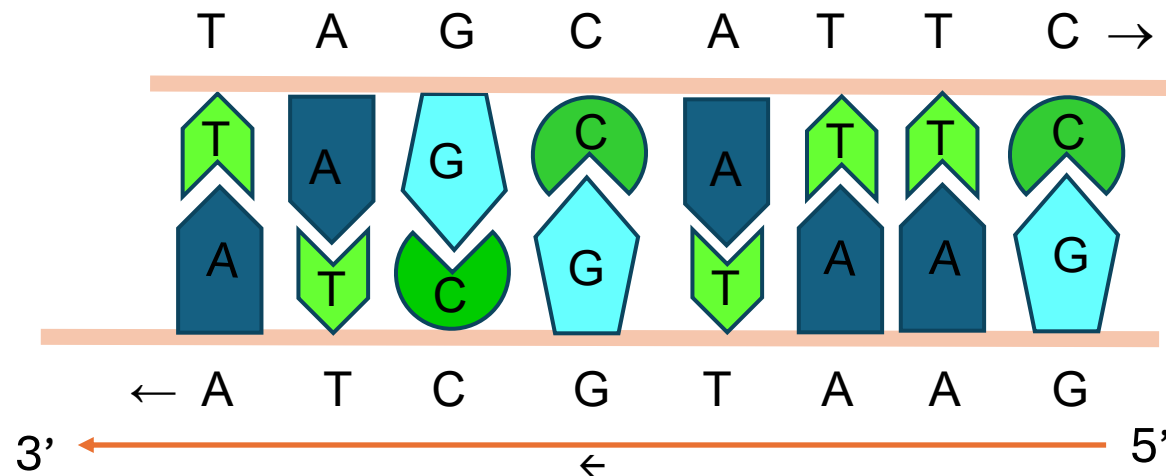
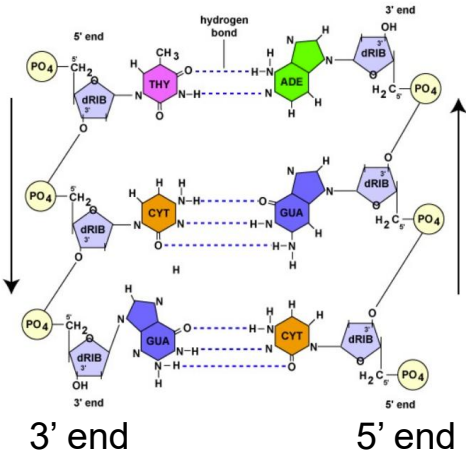
Bases A:T and G:C are paired in DNA



Complimentary

Ribo sugar backbone

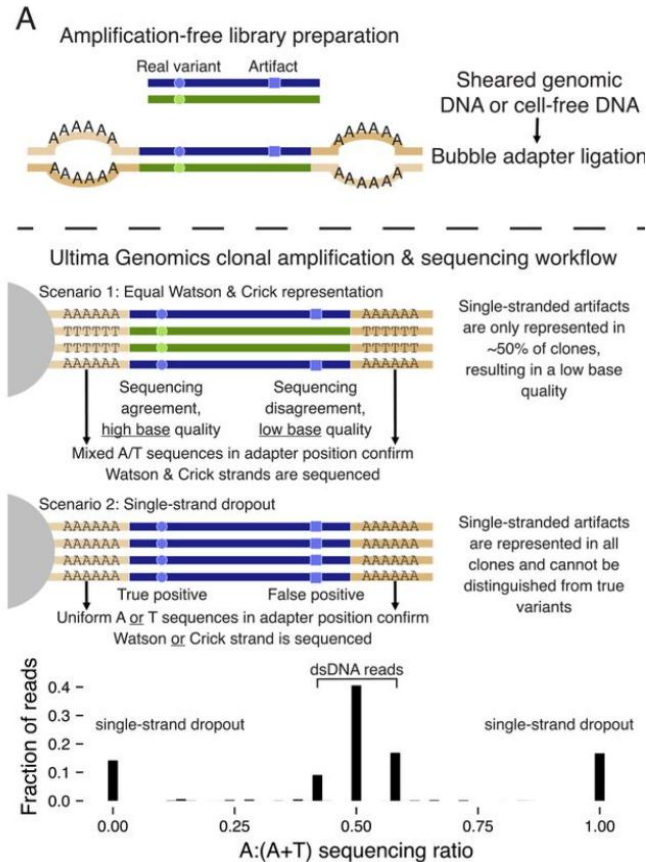
Polymer → 5' → 3'



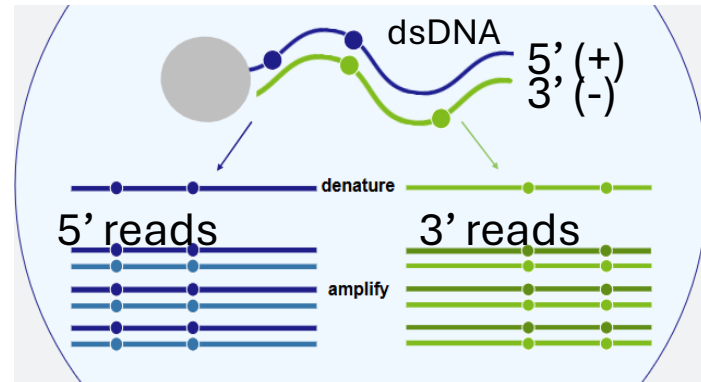
DNA structure

- - - hydrogen bonds

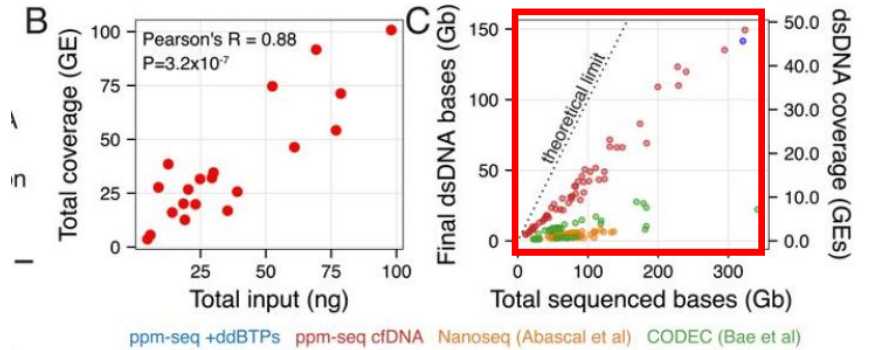
Chip based vs Whole Genome Sequencing (WGS with Next Generation Sequencing (NGS)



5' CTG**C**AGT 3'
3' GAC**G**TCA 5'



Duplex Sequencing filters out false variants showing up in one but not the other strand of the original dsDNA fragment, and gives further confidence of variants showing up in both strands of the original fragment. 5'CTG**C**AGT (+ strand), 3'ACT**G**CAG (-strand) doi: <https://doi.org/10.1101/2020.09.09.289322>

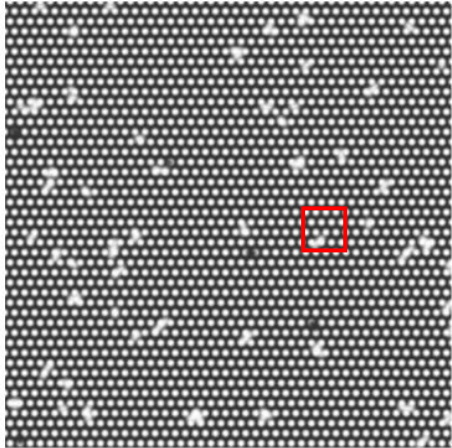


PPM Sequencing: <https://www.biorxiv.org/content/10.1101/2025.08.11.669689v1>

Ultima Genomics: <https://www.biorxiv.org/content/10.1101/2022.05.29.493900v1.full>

Ultima Genomics Sequencing

Silicon Disk

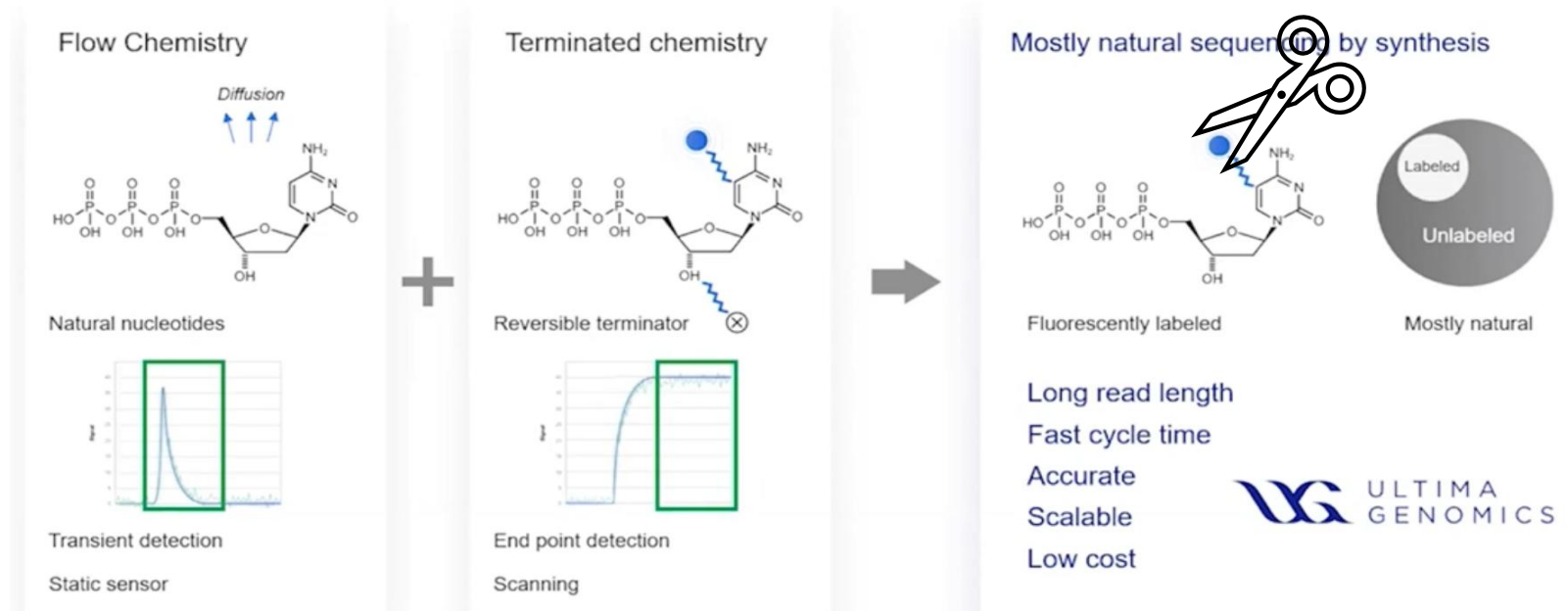


DNA fragments attached one per bead, amplified then electrostatically bound to a silicon disk

A novel spin on SBS sequencing chemistry



SBS = Sequencing By Synthesis



High-speed cameras capture the DNA being synthesized in "real-time." 10 to 20 x 10⁹ reads per wafer over 20 hrs.

Datafile: CRAM File Format

Results downloaded: Compressed Reference-oriented Alignment Map (CRAM), encrypted file for security.

Humans are ~99% identical to the human genome reference sequence the file only stores where we differ from the reference sequence.

A whole human genome CRAM file is typically **less than 1 GB**, often ranging from **150 MB to 1 GB**, though the size depends on the specific sequencing technology, coverage depth, and software used for compression. In contrast to FASTQ files, which contain the raw sequence reads, are much larger than CRAM files, sometimes reaching 100's of GB for a whole genome.

Note you don't have to download the file

YouTube video from founder of Ultima Genomics: <https://www.youtube.com/watch?v=npOfuo3lL4I>



Thank You
Questions?