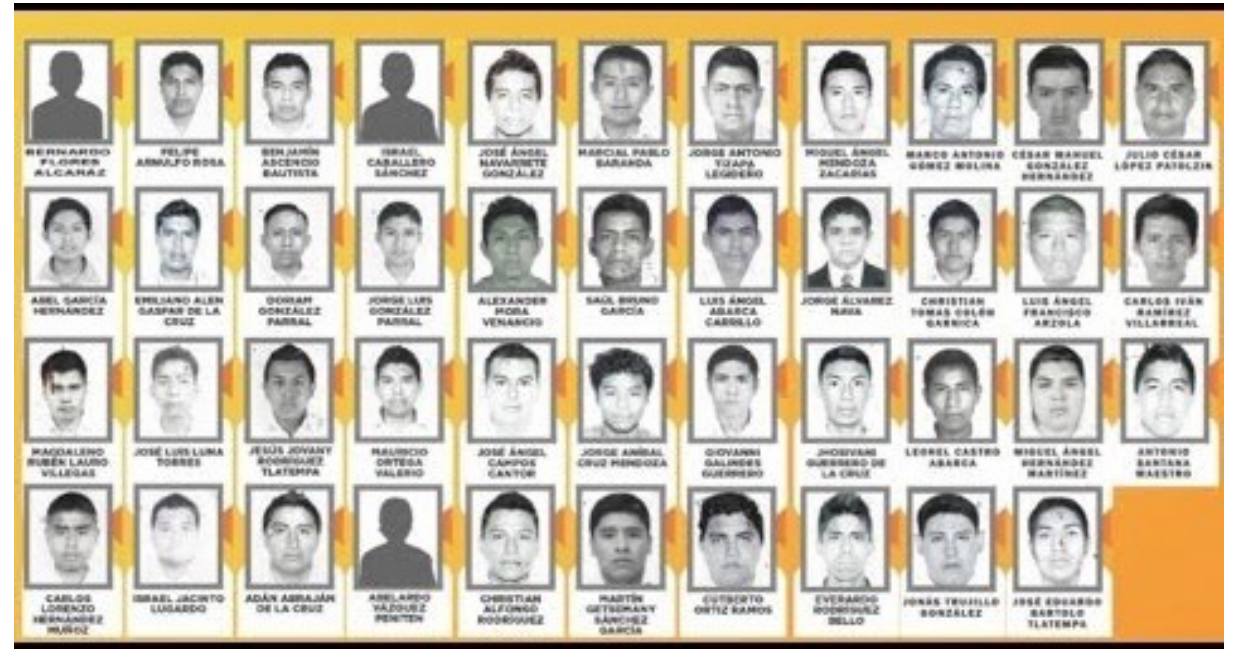


Identificación de personas desaparecidas por medio de análisis genéticos

De los métodos estándar de ADN a las recientes tecnologías de genética molecular para casos complejos



(Conventional) Forensic Genetic Workflow



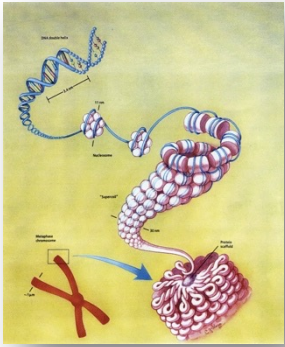
< 150 pg DNA ≈ DNA in 15 cells



Detection



Human Identification

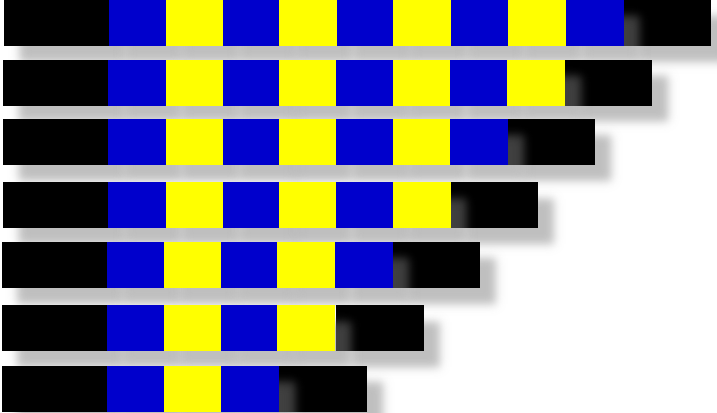


DNA extraction



DNA quantification

Amplification (PCR)



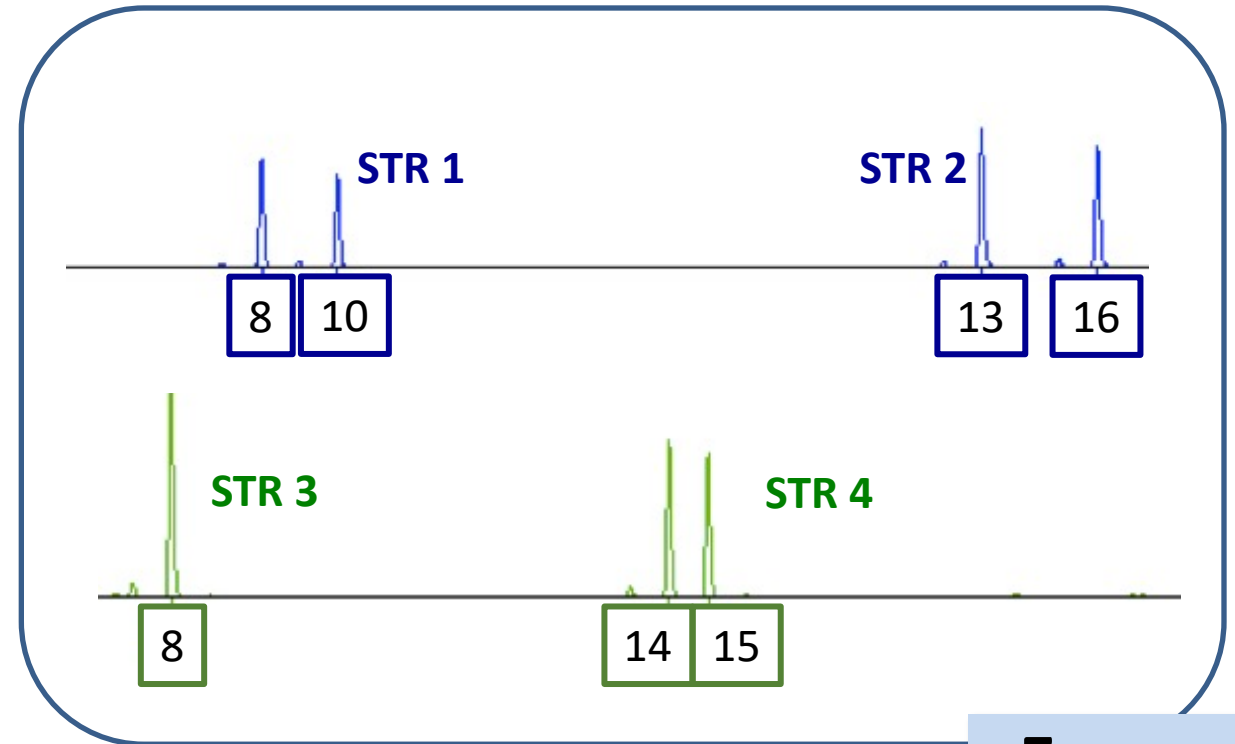
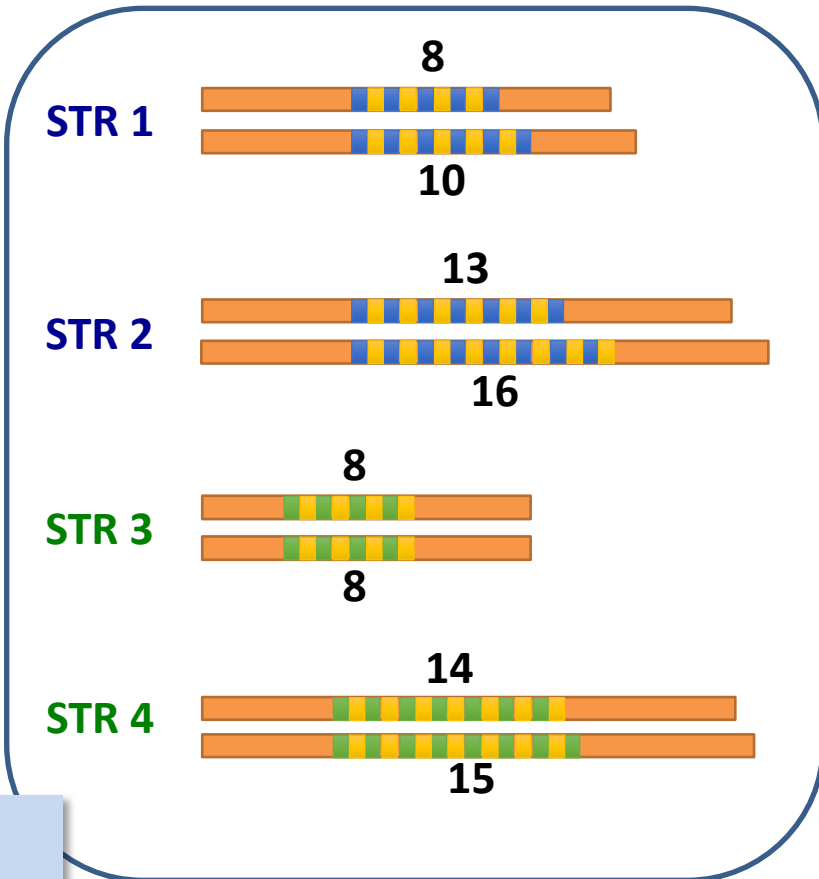
Size (Short Tandem Repeats, Indels)



Sequence



(Conventional) Short Tandem Repeat Analysis



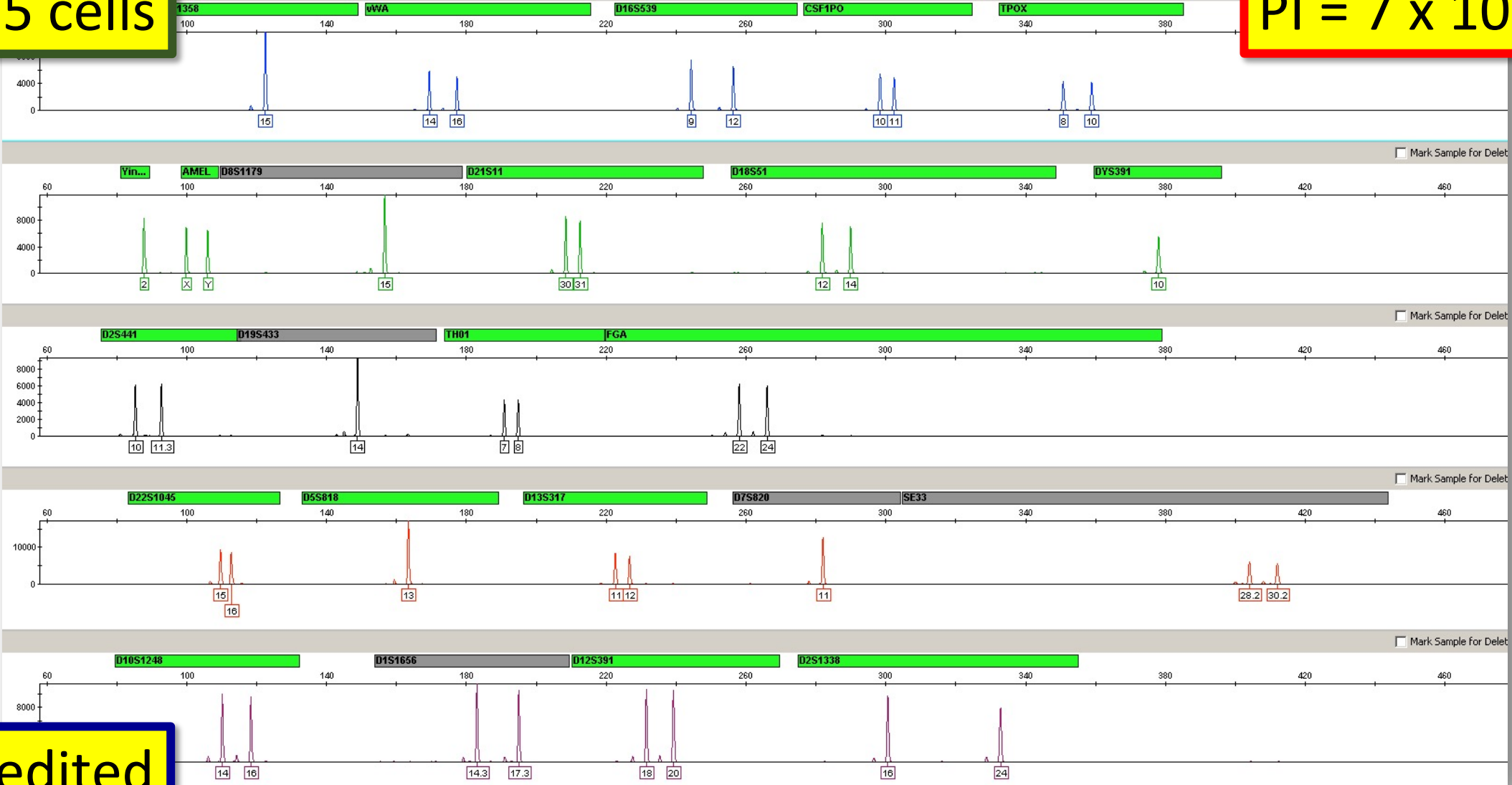
E-gram

8/10 13/16 8/8 14/15

STR-Multiplexes

ca. 15 cells

PI = 7×10^{-26}



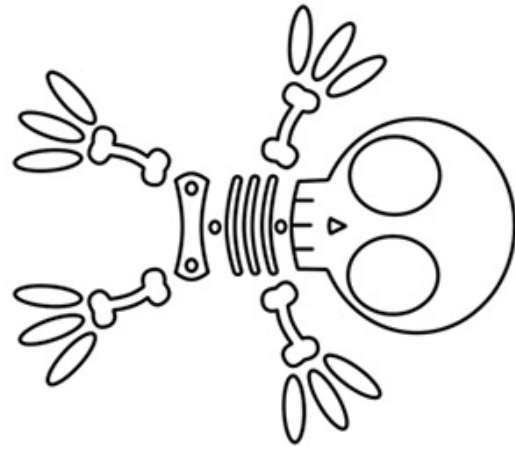
accredited



Applications



Crime casework
reference sample



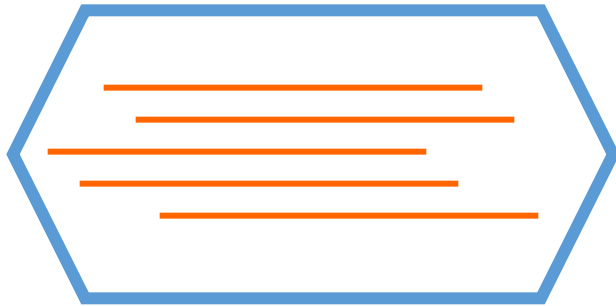
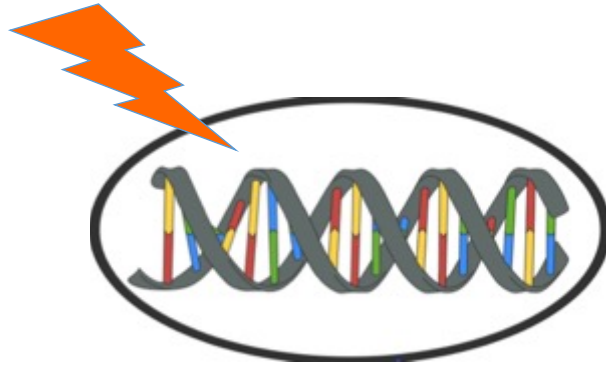
Human identification
ante mortem sample
or
related person(s)



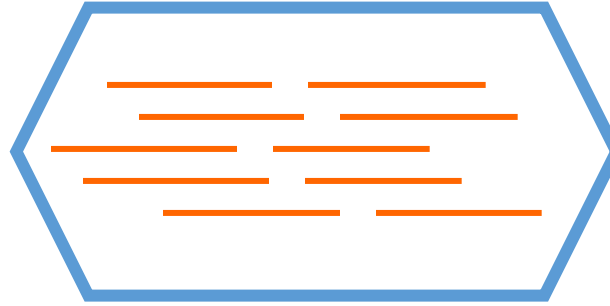
Paternity casework
related person(s)



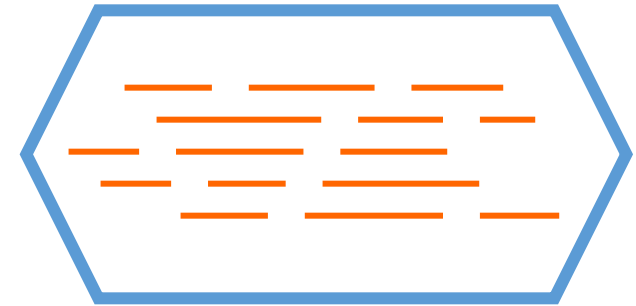
UV



> 400 bp
'standard' protocol

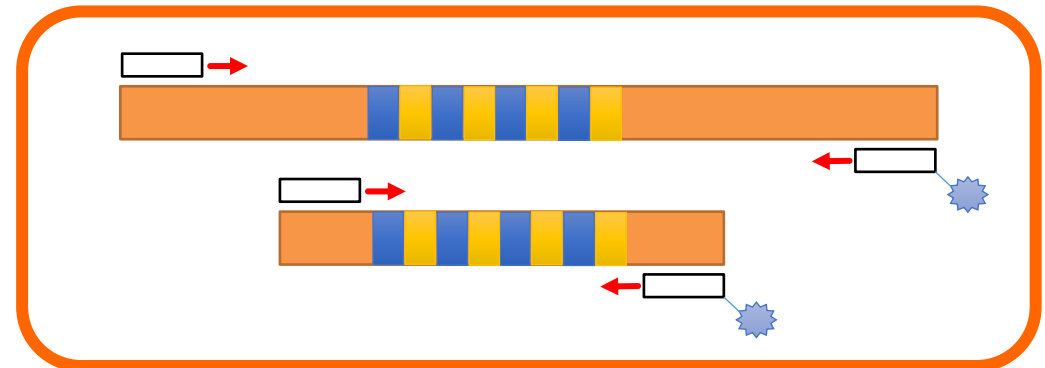


200-400 bp
'midi' protocol



150-200 bp
'mini' protocol

DNA is degrading
(shortening in size)



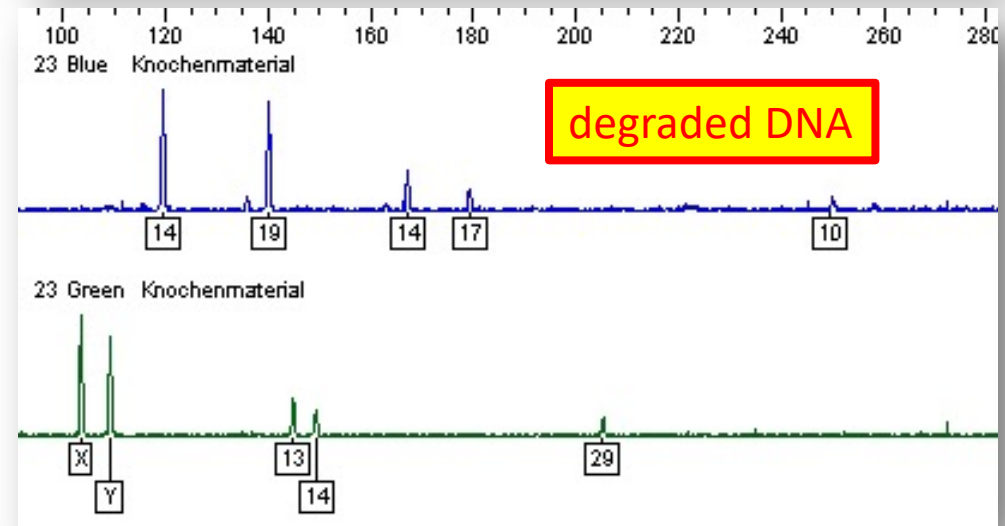
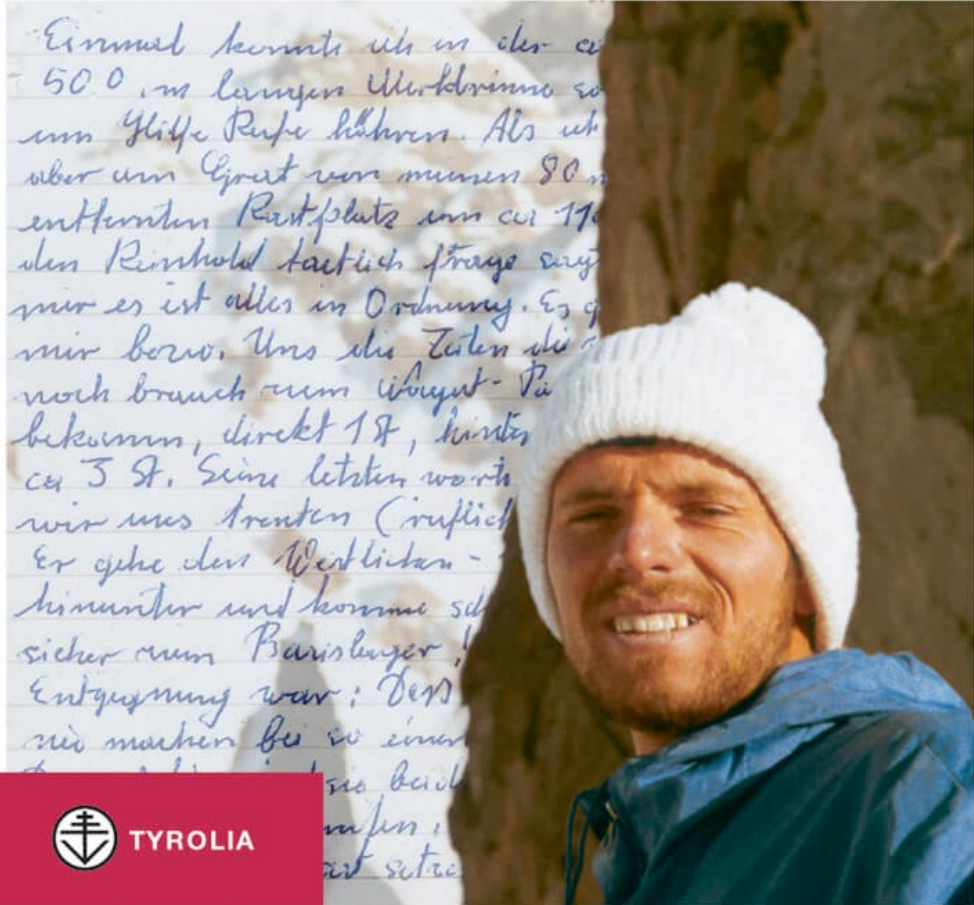
NANGA PARBAT

DAS DRAMA 1970 UND DIE KONTROVERSE



Jochen Hemmleb

WIE DIE MESSNER-TRAGÖDIE ZUM GRÖSSTEN STREITFALL IN DER ALPINGESCHICHTE WURDE



Parson et al (2006) *IJLM*



CASE REPORT

Walther Parson · Anita Brandstätter ·
Harald Niederstätter · Petra Grubwieser ·
Richard Scheithauer

Unravelling the mystery of Nanga Parbat

Received: 23 December 2005 / Accepted: 10 April 2006
© Springer-Verlag 2006

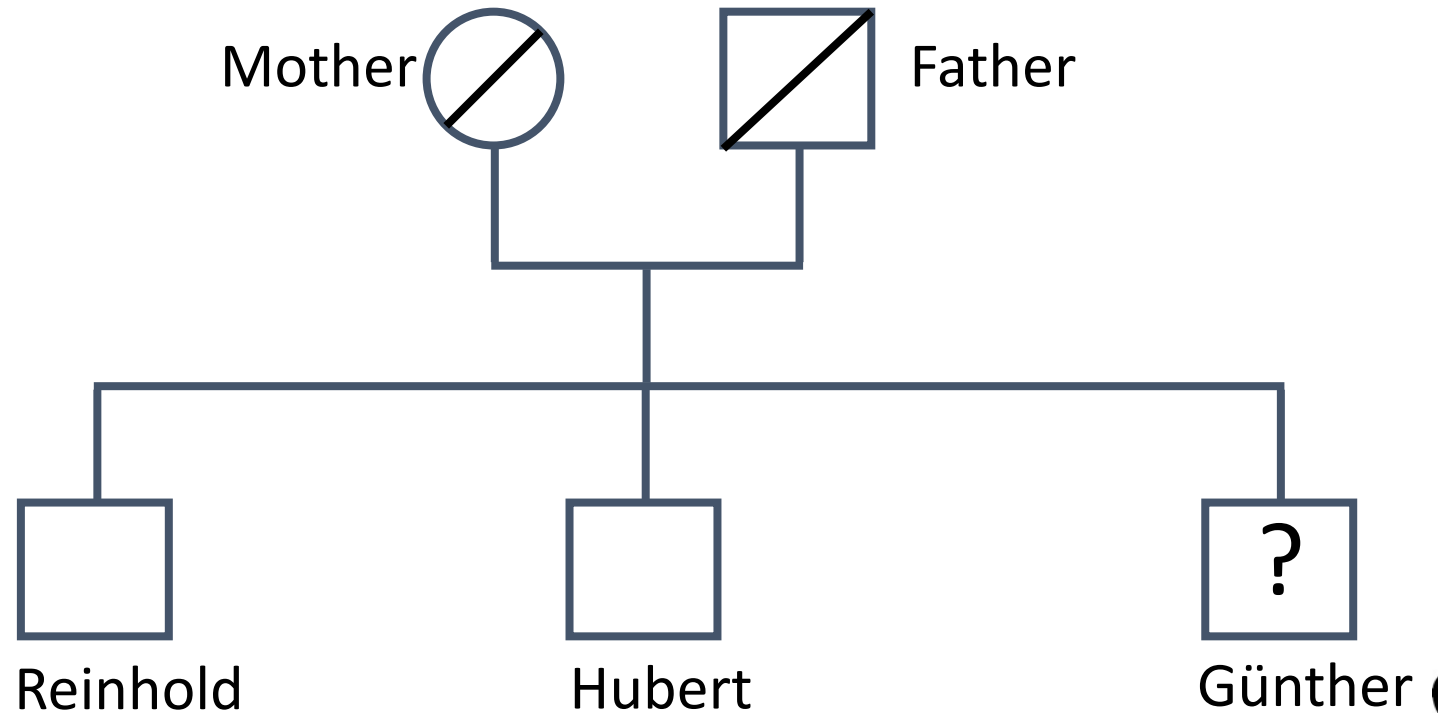


Table 1 Results of autosomal STR profiling

Sample	Amelogenin	D3S1358	vWA	D16S539	D2S1338	D8S1179	D21S11	D18S51
Fibula	X/Y	14/19	14/17	10/12	20/23	13/14	n.r.	12/15
Proximal phalanx	X/Y	14/19	14/17	10/12	20/23	13/14	29/34	12/15
Reinhold Messner	X/Y	14/19	17	10/11	20/25	14	29/32.2	13/15
Hubert Messner	X/Y	16/19	14/17	11/12	25	13/14	30/32.2	12
Sample	D19S433	TH01	FGA	D7S820	CSF1PO	D13S317	TPOX	D5S818
Fibula	14/15	6/9	21/23	n.r.	n.r.	n.r.	n.r.	n.r.
Proximal phalanx	14/15	6/9	21/23	11/12	10/11	11	8/9	11/13
Reinhold Messner	14/16.2	6	22/24	10/12	10/12	11	8/9	12/13
Hubert Messner	14/16.2	6	21/23	11/12	10/11	11/12	8/9	11/13

The STR loci were typed with AmpFLSTR Identifiler and SGM Plus PCR Amplification Kits (both from Applied Biosystems) and a homemade mini-STR multiplex [3]

n.r.: no result

LR=81,900

Confirmed and extended by phalanx (LR=18x10⁹)

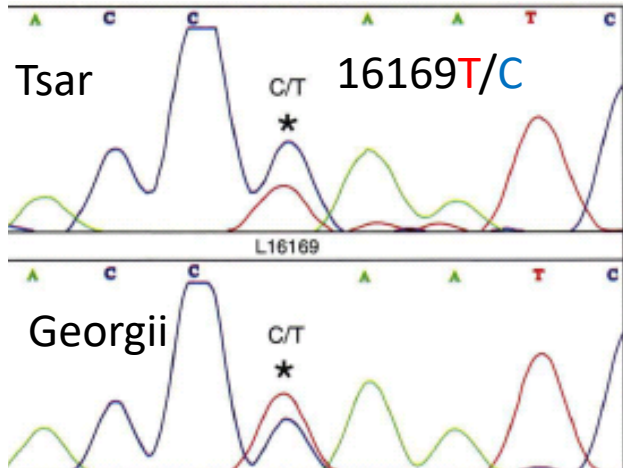
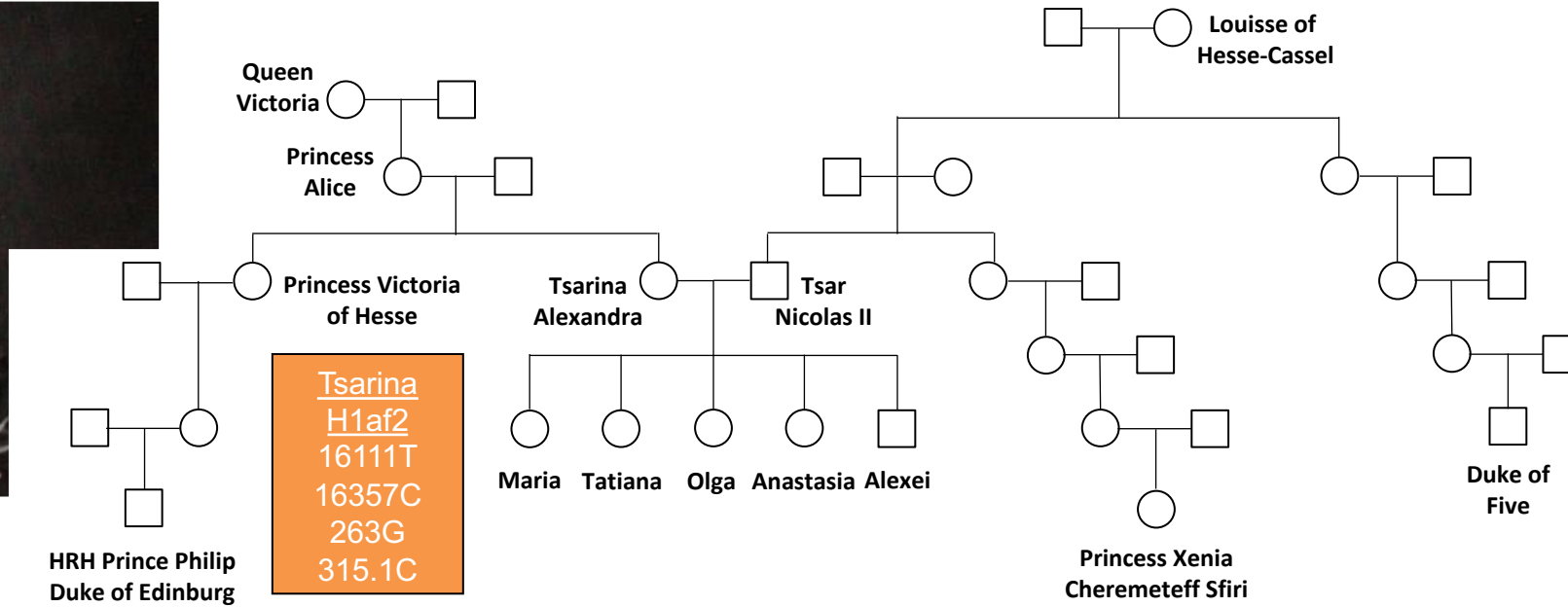
The Identification of the Romanov Family



wikipedia.org

Tsar family (1913) 300th Anniversary of the Romanov Dynasty

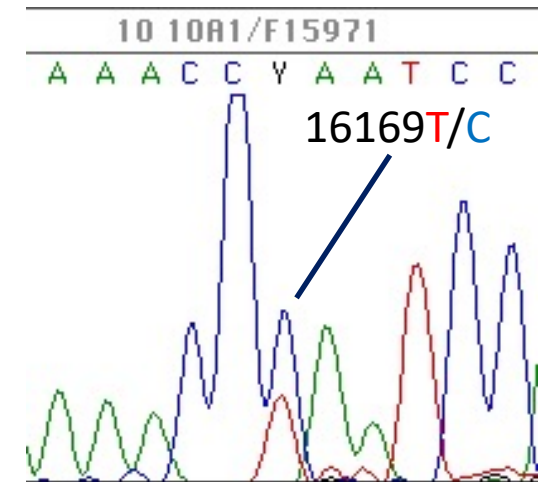
Mitochondrial lineages of the Romanov Family



Georgii
T2a1a
16126C
16169T/C
16294T
16296T
73G
263G
315.1C

Tsar
T2a1a
16126C
16169T/C
16294T
16296T
73G
263G
315.1C

Sfiri/Duke
T2a1a
16126C
16169T
16294T
16296T
73G
263G
315.1C



1991

9 bodies

Tsar Nicolas II
Tsarina Alexandra
Olga
Tatiana
Anastasia
Anna Demidova
Alexei Trupp
Dr. Botkin
Ivan Kharitinov

~ 70 meters away

2007

2 sets of remains

Alexei and
Maria

???

The Identification of the Romanov Family - part 2



autosomal
STRs

Table 3. Autosomal STR Genotypes for the Romanov Family.

Marker	Sample 4.3 Tsar Nicholas II	Sample 7.4 Tsarina Alexandra	Sample 3.46 Olga	Sample 5.21 Tatiana	Sample 6.14 Maria or Anastasia	Sample 147 Anastasia or Maria	Sample 146.1 Alexei
Amelog	X, Y	X, X	X, X	X, X	X, X	X, X	X, Y
D3S1358	14, 17	16, 18	17, 18	17, 18	16, 17	17, 18	14, 18
TH01	7, 9.3	8, 8	8, 9.3	7, 8	8, 9.3	7, 8	8, 9.3
D21S11	32.2, 33.2	30, 32.2	30, 33.2	32.2, 33.2	30, 33.2	30, 33.2	32.2, 33.2
D18S51	12, 17	12, 13	12, 12	12, 12	13, 17	12, 17	12, 17
D5S818	12, 12	12, 12	12, 12	12, 12	12, 12	12, 12	12, 12
D13S317	11, 12	11, 11	11, 11	11, 11	11, 11	11, 11	11, 12
D7S820	12, 12	10, 12	12, 12	10, 12	12, 12	10, 12	12, 12
D16S539	11, 14	9, 11	11, 11	11, 11	11, 14	9, 11	11, 14
CSF1PO	10, 12	11, 12	11, 12	11, 12	10, 11	10, 12	10, 12
D2S1338	17, 25	19, 23	17, 19	23, 25	17, 19	17, 23	23, 25
VWA	15, 16	15, 16	15, 16	15, 16	15, 16	15, 16	15, 16
D8S1179	13, 15	16, 16	13, 16	15, 16	13, 16	15, 16	15, 16
TPOX	8, 8	8, 8	8, 8	8, 8	8, 8	8, 8	8, 8
FGA	20, 22	20, 20	20, 22	20, 20	20, 22	20, 22	20, 22
D19S433	13, 13.2	13, 16.2	13.2, 16.2	13.2, 16.2	13, 16.2	13, 13	13, 13.2

Table 1. Sequences of the samples recovered from "Grave #2" in August 2007 and tested in this study.

Bone	Russian #	Region Sequenced	Sequence
Right humerus	141	16024-16391 and 35-369	16111T, 16357C, 263G, 315.1C
Occipital fragment	139	no results	-
Occipital fragment	144.1	16024-576	16111T, 16357C, 16519C, 263G, 315.1C, 524.1A; 524.2C
Right os coxae-♀	145	16024-16391 and 35-369	16111T, 16357C, 263G, 315.1C
Left femur	146.1*	16024-576	16111T, 16357C, 16519C, 263G, 315.1C, 524.1A; 524.2C
Right femur - ♀	147*	16024-576	16111T, 16357C, 16519C, 263G, 315.1C, 524.1A; 524.2C
Right scapula	140	16024-16391 and 35-369	16111T, 16357C, 263G, 315.1C
Cranial fragment	143	16024-16391 and 35-369	16111T, 16357C, 263G, 315.1C
Left ilium	142	no results	-

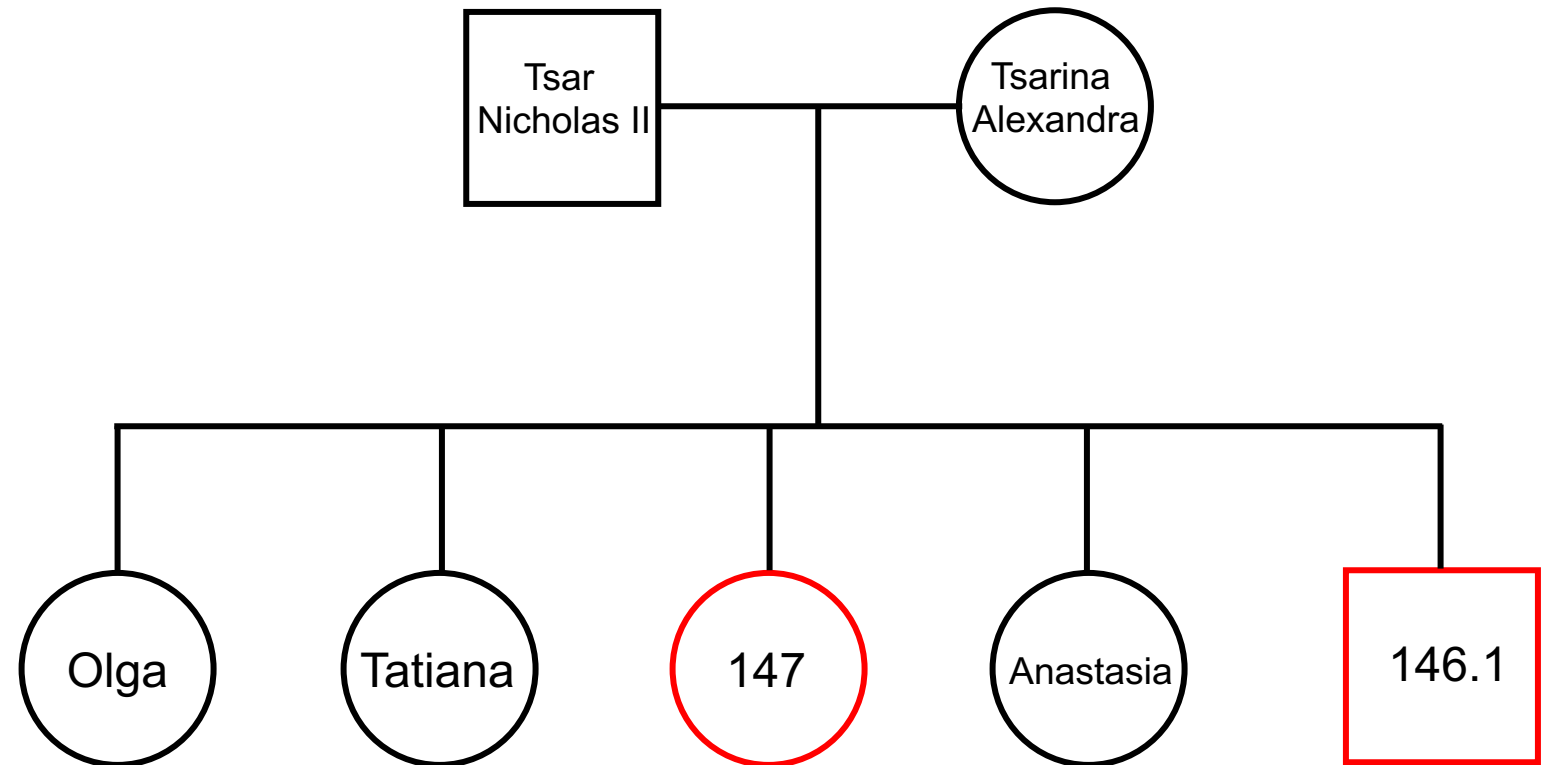
Y-STRs

Table 4. Y-STR haplotype for Nicholas, Alexei and Andrew Romanov.

DYS19	DYS389I	DYS389II	DYS390	DYS391	DYS392	DYS393	DYS385a/b
14	13	29	24	10	13	13	11, 14
DYS438	DYS439	DYS437	DYS448	DYS456	DYS458	DYS635	YGATAH4
12	11	15	19	16	17	24	12

mtDNA

The Identification of the Romanov Family - part 2



Scenario: Samples 146 and 147 are members of the Romanov Family

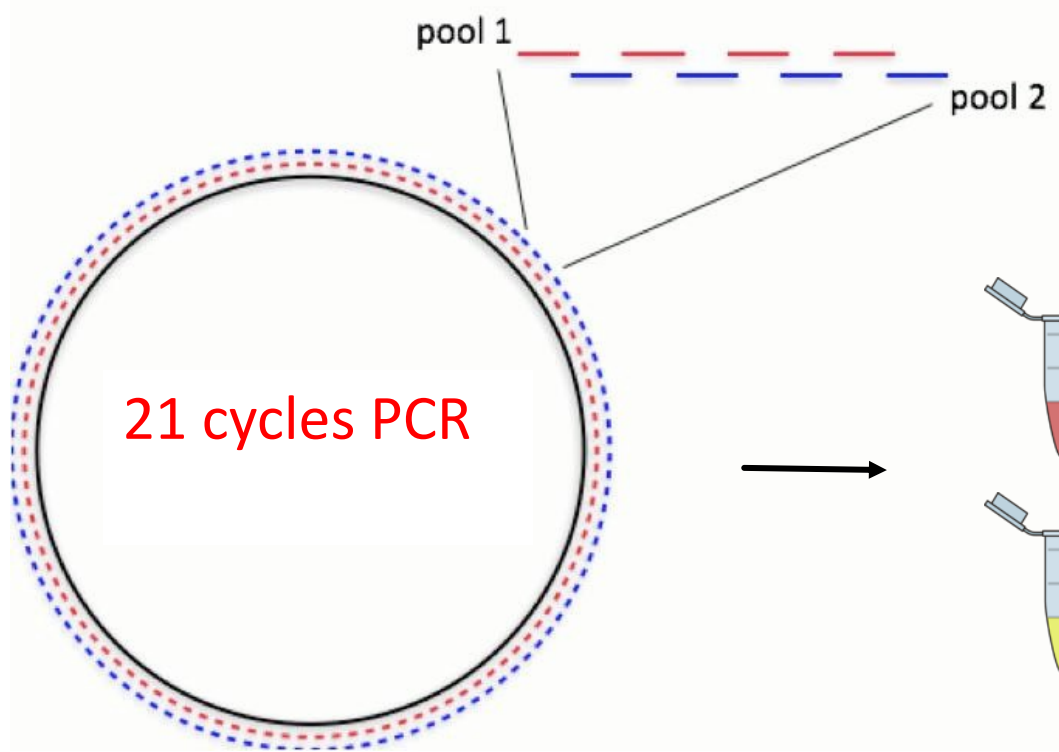
Cumulative LR = **4.36 Trillion to 80 Trillion**

Massively Parallel Sequencing in Forensics

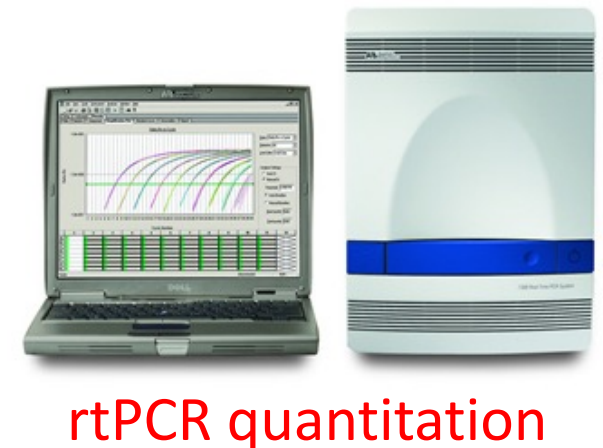
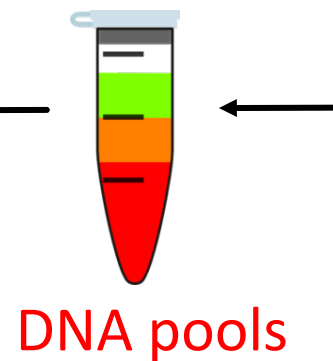
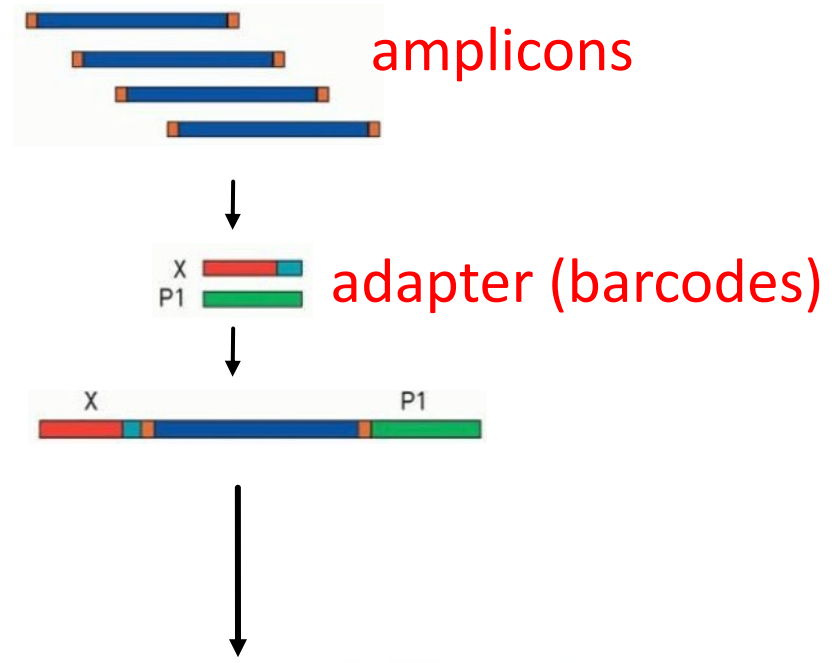
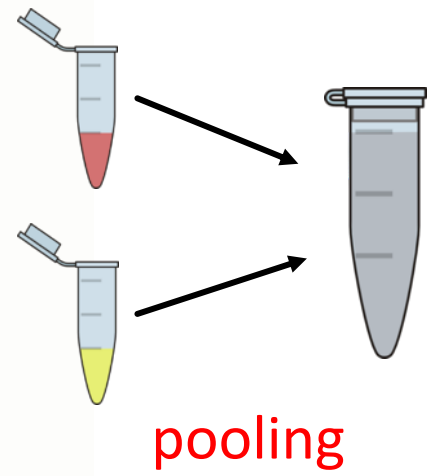


Illumina

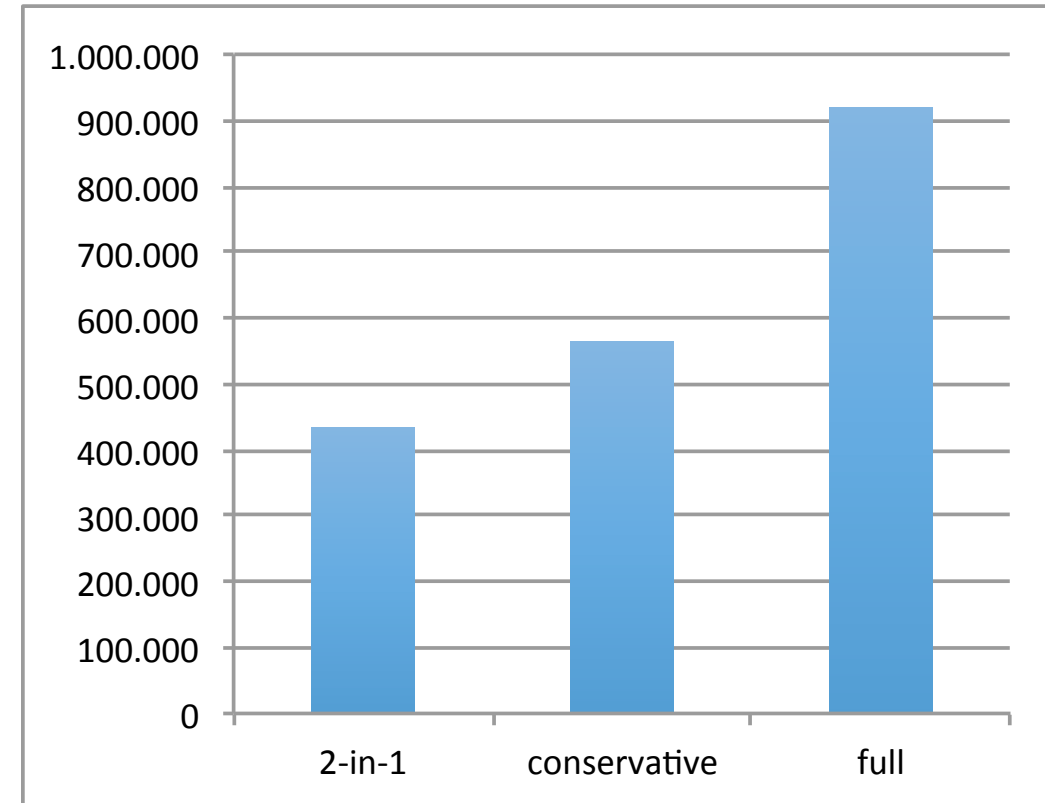
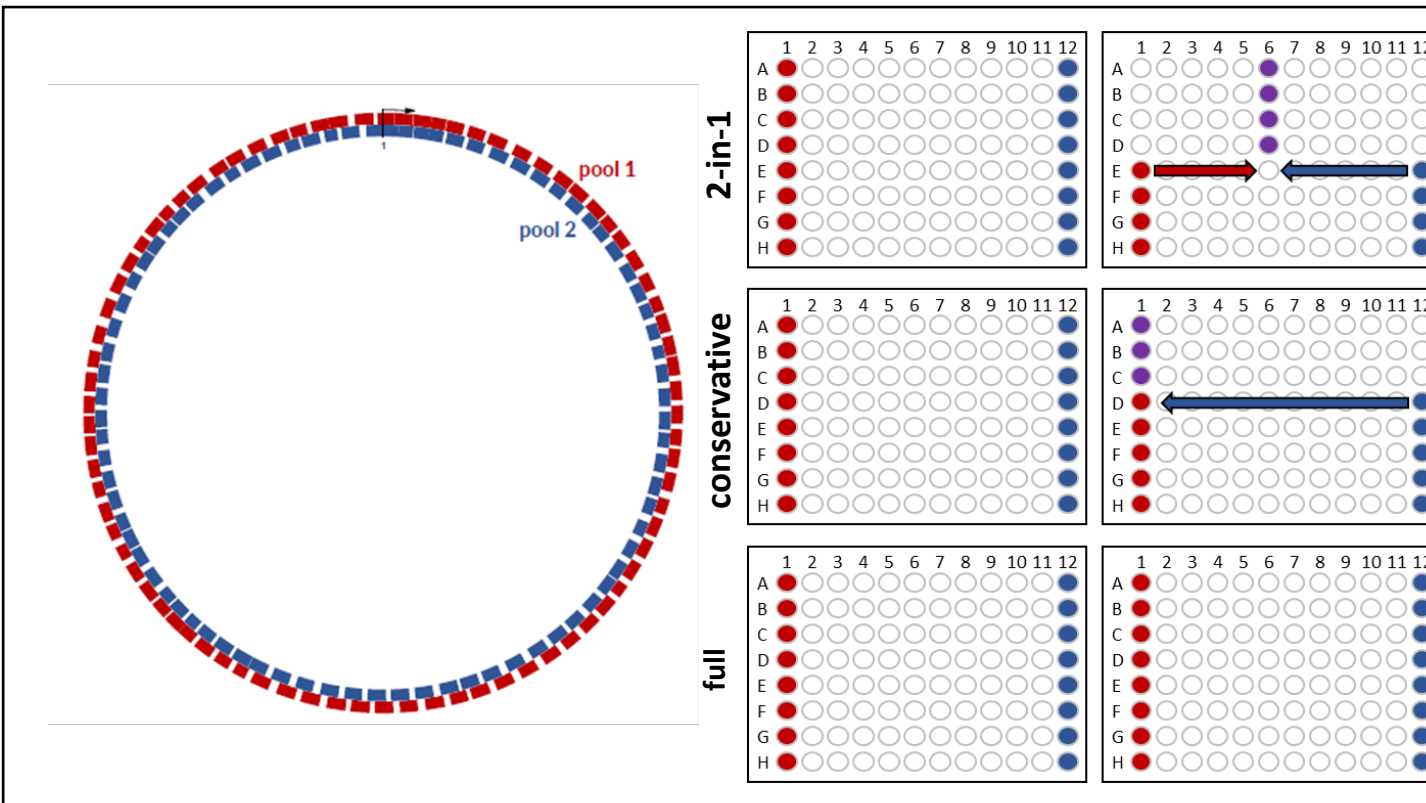
Ion Torrent



Workflow



Ion Torrent (TFS) protocol for low-quality mtDNA



Three library pooling strategies (DNA quality)

Solving Cold Cases with mitogenomes

Cold case 1			
Hair shaft	2-in-1	V1a1	1-16569
Hair shaft	Sanger	HV0	16024-425 428-543
Hair end	2-in-1	V1a1	1-16569
Hair end	Sanger	HV0	16024-460 484-576
Cold case 2			
Swab	conservative	U5a1a1b	1-16569
Swab	Sanger	R	16128-16348
Victim	conservative	U5a2c4	1-16569
Victim	Sanger	U5a	45-287 16128-16348
Suspect	conservative	H1	1-16569
Suspect	Sanger	R0	45-287 16128-16348

Validation of IonChef/S5 mitogenome MPS

- Population Studies
- Contamination Assessment
- Concordance Studies
- Sensitivity and Stochastic Effects
- Mock Casework Samples
- Mixtures



Validation of IonChef/S5 mitogenome MPS

Forensic Science International: Genetics 42 (2019) 244–251



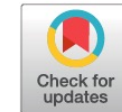
Contents lists available at [ScienceDirect](#)

Forensic Science International: Genetics

journal homepage: www.elsevier.com/locate/fsigen



Evaluation of mitogenome sequence concordance, heteroplasmy detection, and haplogrouping in a worldwide lineage study using the Precision ID mtDNA Whole Genome Panel






Christina Strobl^a, Jennifer Churchill Cihlar^b, Robert Lagacé^c, Sharon Wootton^c, Chantal Roth^c, Nicole Huber^a, Lisa Schnaller^a, Bettina Zimmermann^a, Gabriela Huber^a, Seah Lay Hong^d, Rodrigo Moura-Neto^e, Rosane Silva^f, Farida Alshamali^g, Luis Souto^h, Katja Anslingerⁱ, Balazs Egyed^j, Renata Jankova-Ajanovska^k, Andrea Casas-Vargas^l, Wiliam Usaquén^l, Dayse Silva^m, Claudia Barletta-Carrilloⁿ, Dean Herman Tineo^o, Carlos Vullo^p, Reinhard Würzner^q, Catarina Xavier^a, Leonor Gusmão^m, Harald Niederstätter^a, Martin Bodner^a, Bruce Budowle^b, Walther Parson^{a,r,*}

Validation of IonChef/S5 mitogenome MPS



Article

Developmental Validation of a MPS Workflow with a PCR-Based Short Amplicon Whole Mitochondrial Genome Panel

Jennifer Churchill Cihlar ^{1,2,*} , Christina Amory ³ , Robert Lagacé ⁴, Chantal Roth ⁴, Walther Parson ^{3,5}  and Bruce Budowle ^{1,2}

¹ Center for Human Identification, University of North Texas Health Science Center, 3500 Camp Bowie Boulevard, Fort Worth, TX 76107, USA; Bruce.Budowle@unthsc.edu

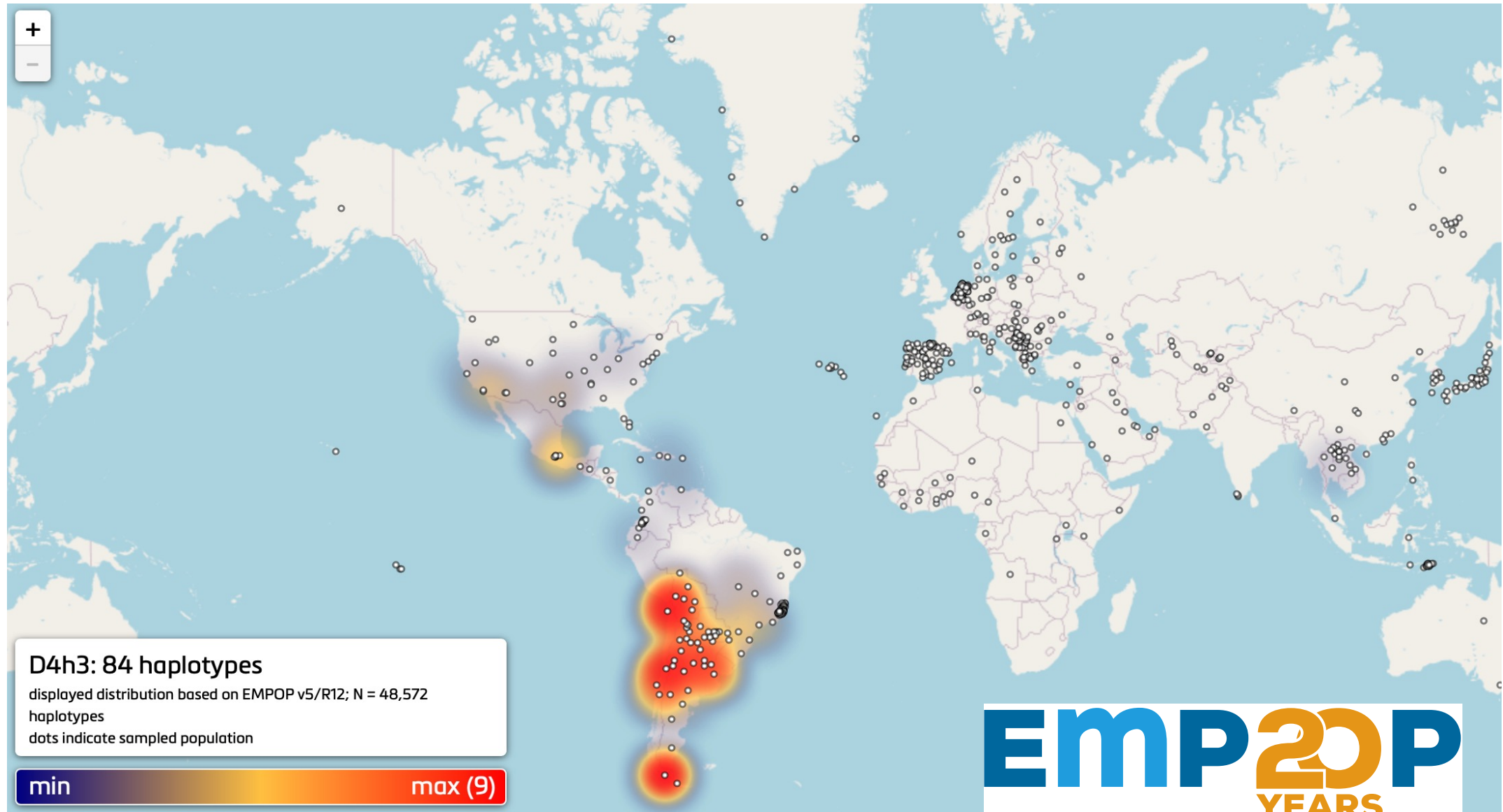
² Department of Microbiology, Immunology and Genetics, University of North Texas Health Science Center, 3500 Camp Bowie Boulevard, Fort Worth, TX 76107, USA

³ Institute of Legal Medicine, Medical University of Innsbruck, 6020 Innsbruck, Austria; christina.amory@i-med.ac.at (C.A.); walther.parson@i-med.ac.at (W.P.)

⁴ Human Identification Group, Thermo Fisher Scientific, South San Francisco, CA 94080, USA; Robert.Lagace@thermofisher.com (R.L.); chantal.roth@thermofisher.com (C.R.)






⁵ Forensic Science Program, The Pennsylvania State University, University Park, PA 16802, USA

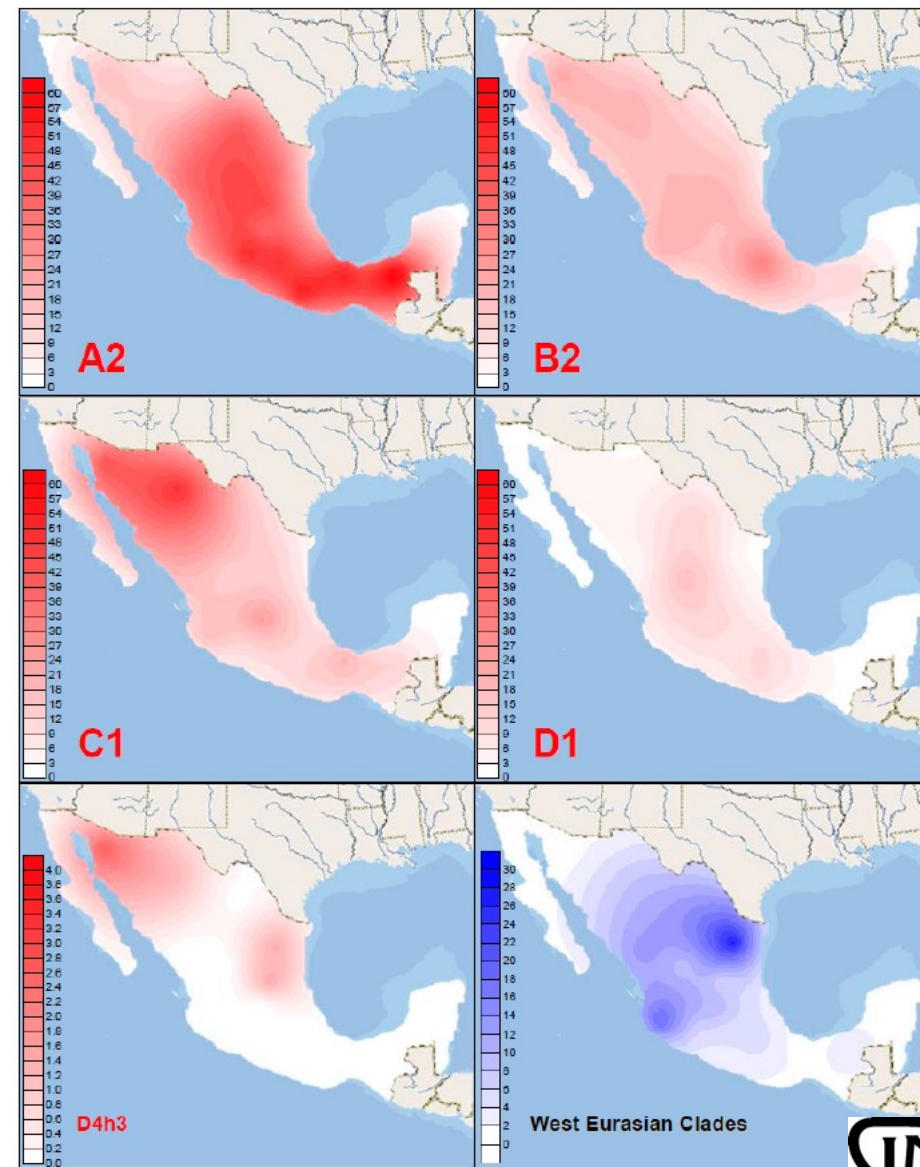
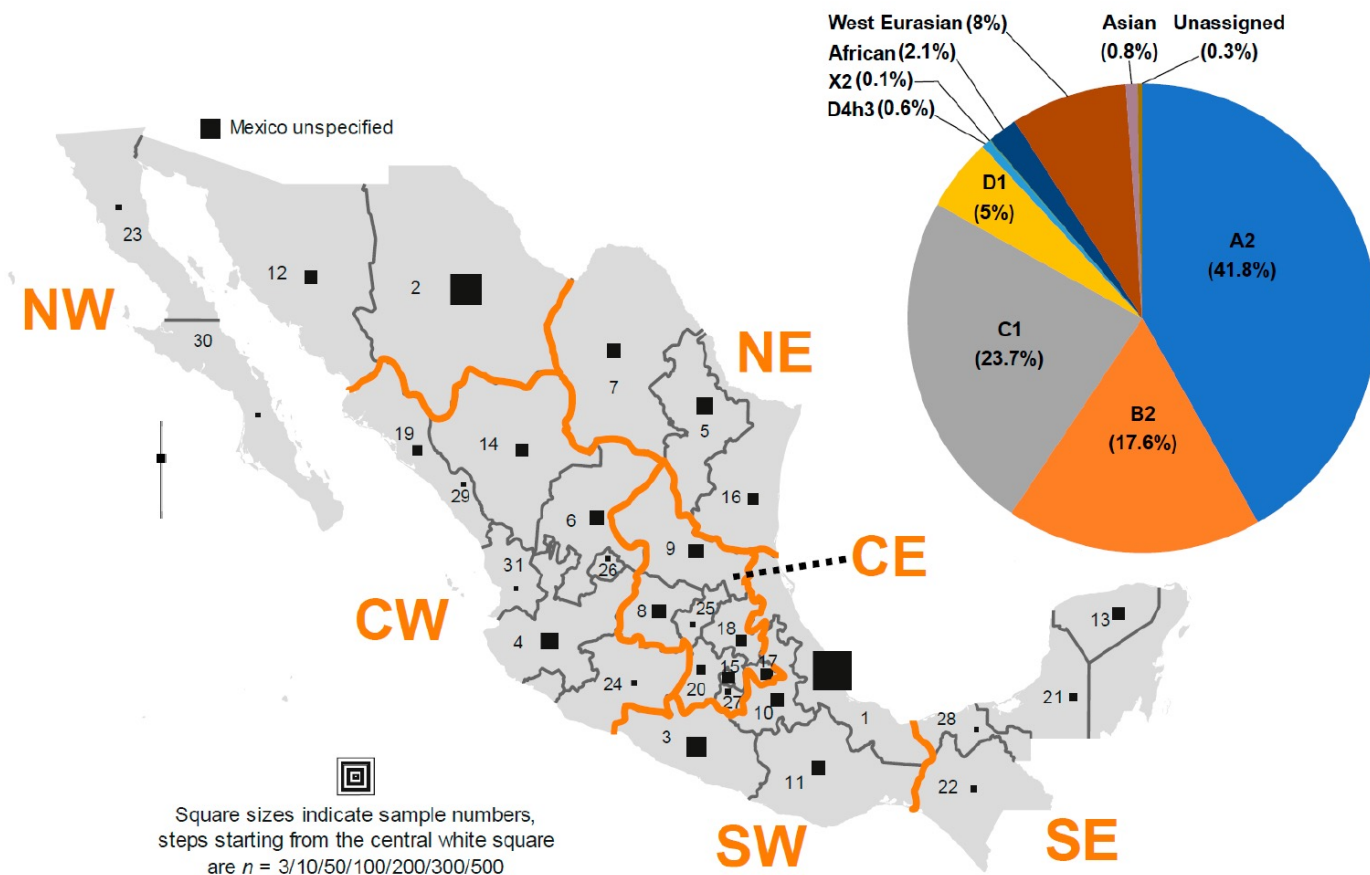
<https://empop.online>



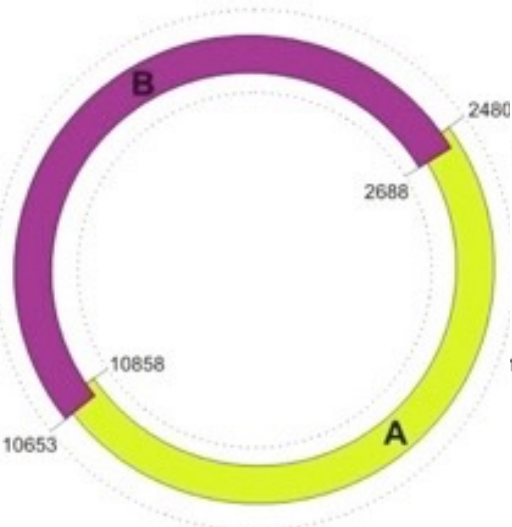
Article

The Mitochondrial DNA Landscape of Modern Mexico

Martin Bodner ^{1,†} , Ugo A. Perego ^{2,3,4,†} , J. Edgar Gomez ^{3,5}, Ricardo M. Cerda-Flores ⁶ , Nicola Rambaldi Migliore ², Scott R. Woodward ³, Walther Parson ^{1,7,*}  and Alessandro Achilli ^{2,*} 



mtDNA enrichment strategies for MPS

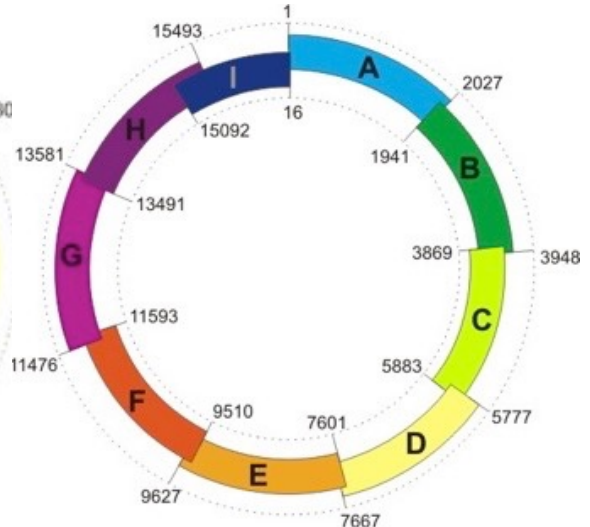


"maxi"

Fendt et al 2009
mtG ca. 8.5 kbp



PGM

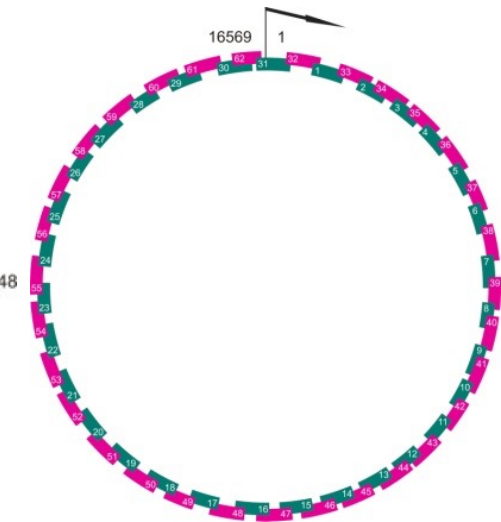


"midi"

GMI modified
mtG ca.2 kbp



Ion Chef/Ion S5

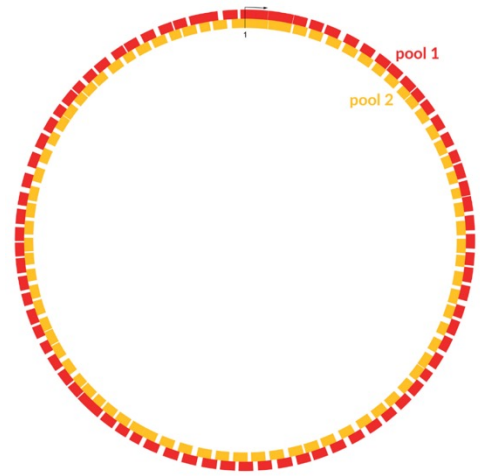


"mini"

Precision-ID (TFS)
CR/mtG ca. 175 bp



MiSeq FGx[®]

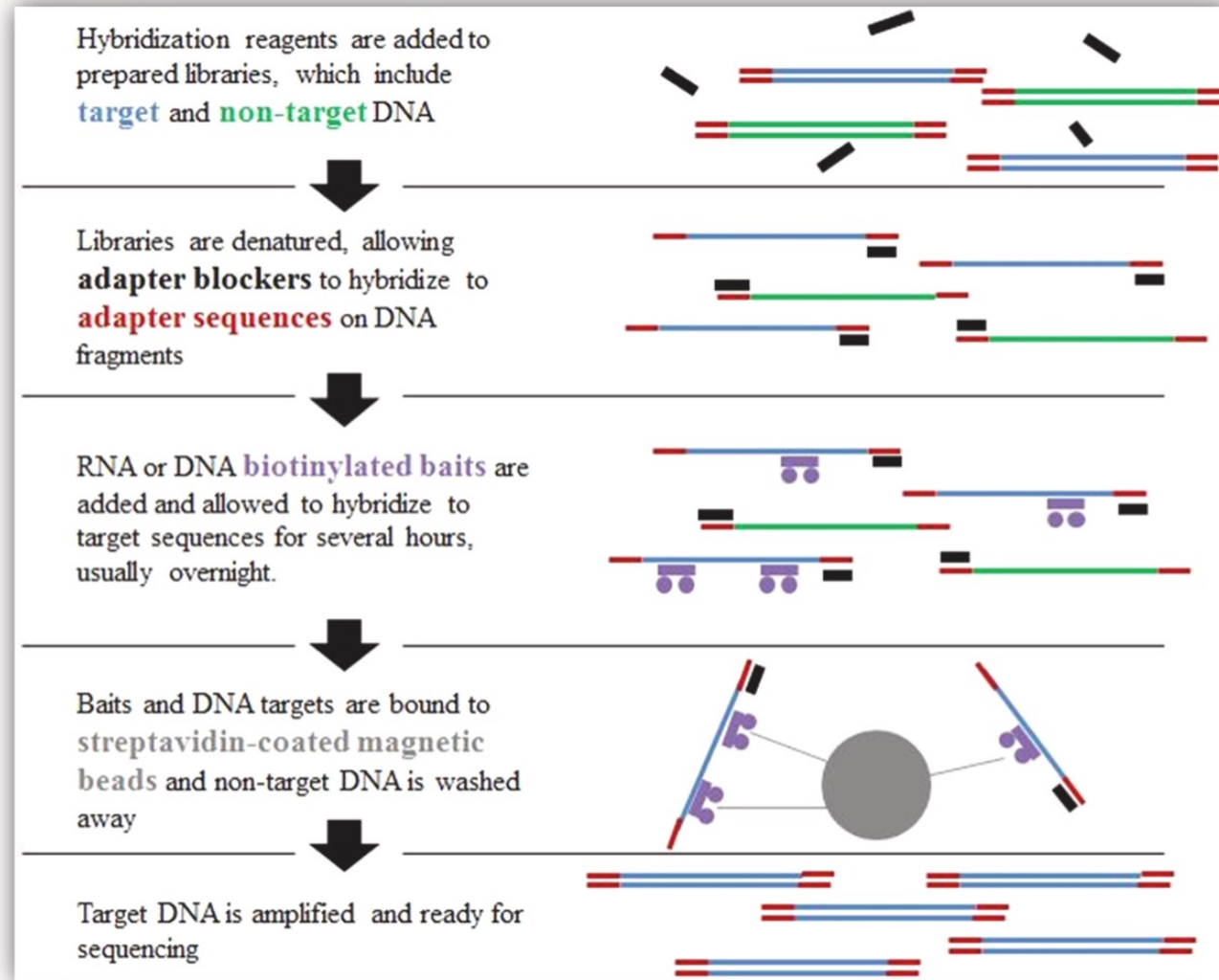


"micro"

Eduardoff et al 2017
PEC CR/mtG < 70 bp



Hybridization Capture - MPS



Application of different library methods for MPS of mtDNA

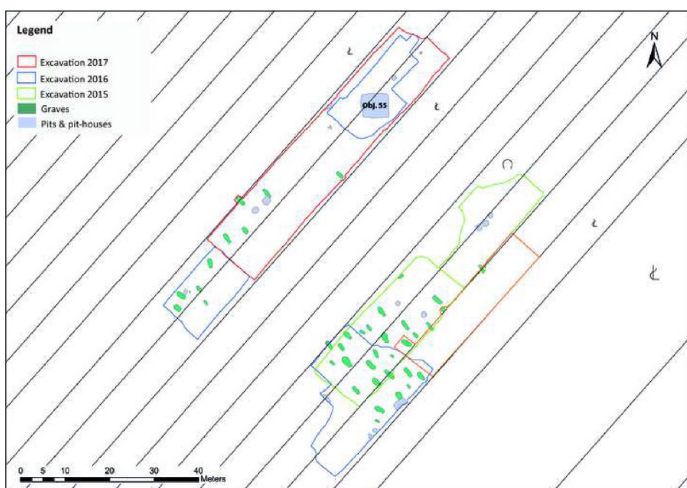
Archaeological and Anthropological Sciences (2020) 12: 171
<https://doi.org/10.1007/s12520-020-01102-5>

ORIGINAL PAPER



House of the dead-exceptional burials of the Avar period (seventh century AD) in Podersdorf am See (Burgenland/A)

Bendeguz Tobias¹  • Thomas Koch Waldner²  • Christina Strobl³  • Harald Niederstätter⁴  • Christina Musalek⁵ • Konstantina Saliari⁶  • Ulrike Töchterle⁷  • Walther Parson^{8,9} 



Hybridization Capture of nuclear SNPs

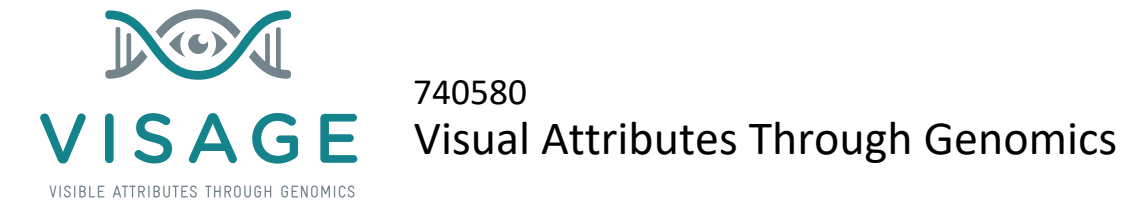
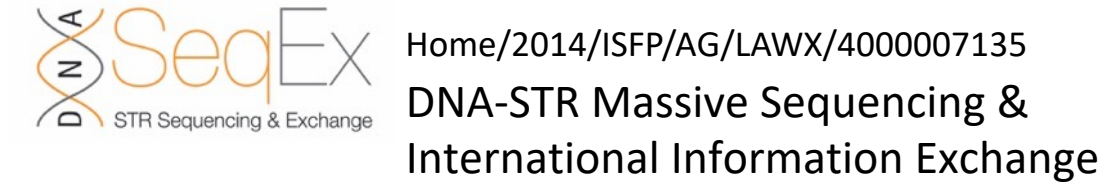
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1 **Extended kinship analysis of historical remains using SNP capture**

2 Erin M. Gorden^{1,2}, Ellen M. Greytak³, Kimberly Sturk-Andreaggi^{1,2,4}, Janet Cady³, Timothy P.

3 McMahan¹, Steven Armentrout³, Charla Marshall^{1,2,5}

Acknowledgements



2011-MU-MU-K402

Maximizing mtDNA Testing Potential with the
Generation of High-Quality mtGenome Reference
Data



Der Wissenschaftsfonds.

Translational Research project L397

EMPOP—an innovative human mtDNA database

Research project P22880-B12

Genetic discovery of an early medieval Alpine
population

dna.bases



Co-funded by the Internal
Security Fund of the
European Union

Monopoly 2016, STEFA, 779485

Steps Towards a European Forensic Science
Area; WP7; Empowering Forensic Genetic
DNA Databases for the Interpretation of Next
Generation Sequencing Profiles (dna.bases)

EMPOP

STRidER

