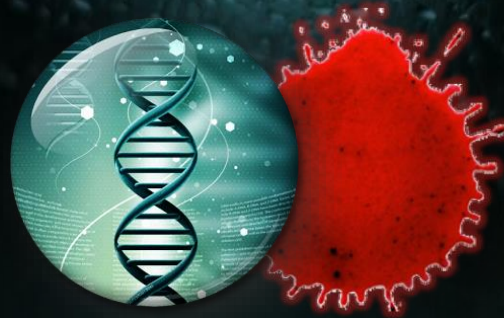
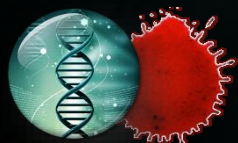


NUKLEOVÉ KYSELINY V ROLI SVĚDKA



Halina Šimková



o čem budeme mluvit



1. INDIVIDUALITA
A IDENTIFIKACE



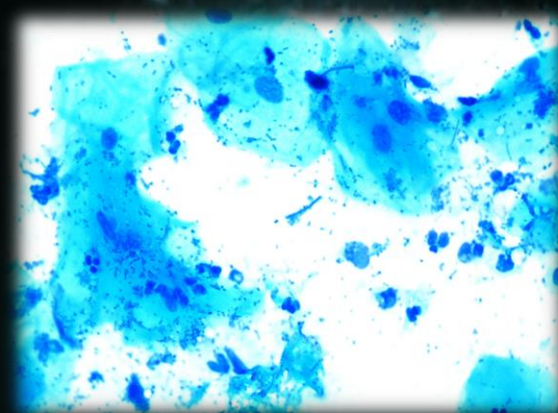
2. PŘÍBUZNOST A
RODOVOST



3. KRIMINALISTICKÉ
ANALÝZY



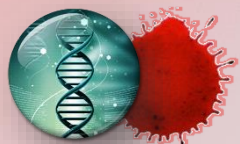
4. FORENZNÍ
PHENOTYPING



5. FORENZNÍ
EPIGENETIKA

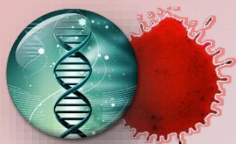


6. „NON-HUMAN“
ANALÝZY



1. INDIVIDUALITA A IDENTIFIKACE





PODSTATA INDIVIDUALITY DNA

1) polymorfismus DNA lokusů = existence alel

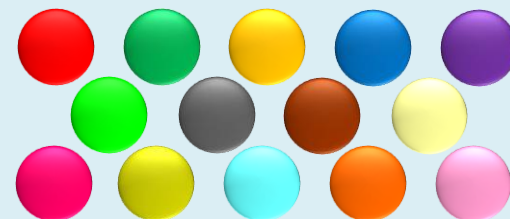
žádný nebo malý



střední

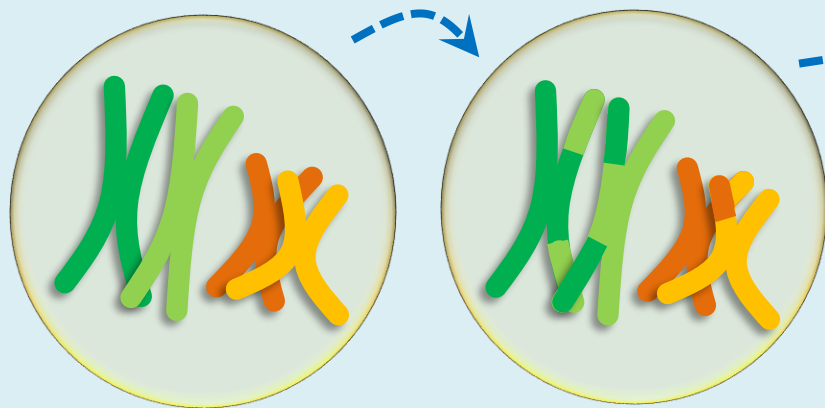


velký (mnohotný)



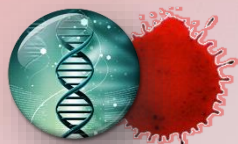
2) náhodné „prohazování“ DNA lokusů při meióze

2a) crossing-over



2b) segregace chromozómů





PODSTATA INDIVIDUALITY DNA

3) další specifické odchylky vzniklé mutacemi

drobného rozsahu

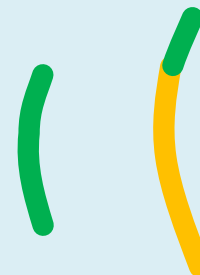
středního rozsahu

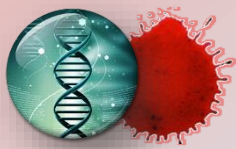
velkého rozsahu

CACCGTATTTA



CACGCGTAATTA

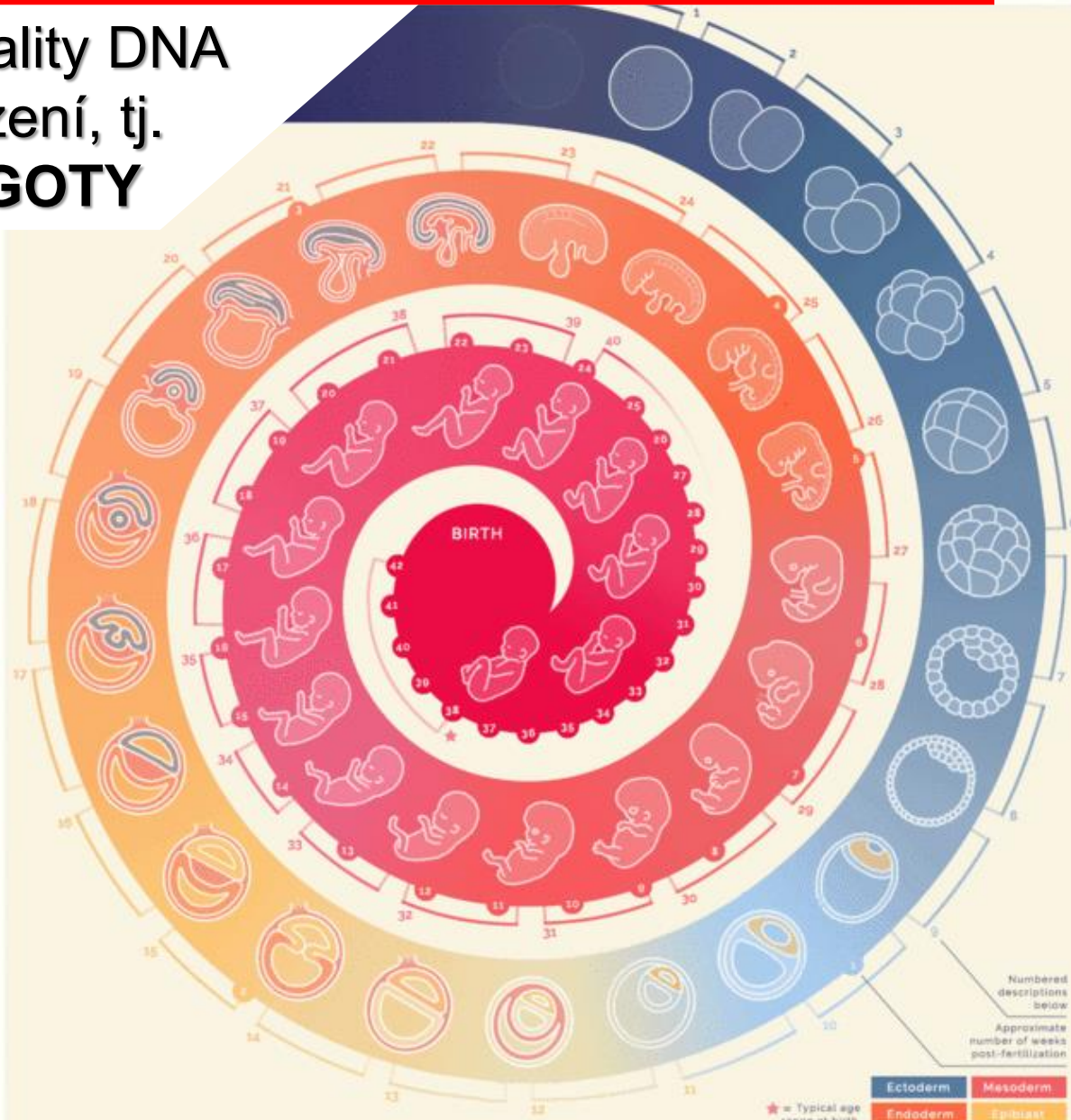


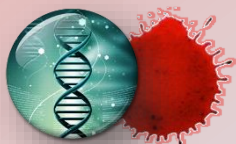


VZNIK DNA INDIVIDUA

➡ základní rysy individuality DNA dány v okamžiku oplození, tj. v momentě vzniku **ZYGOTY**

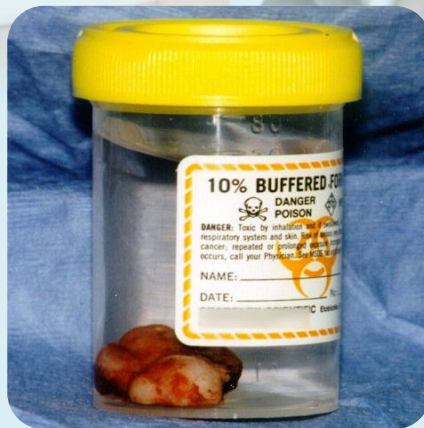
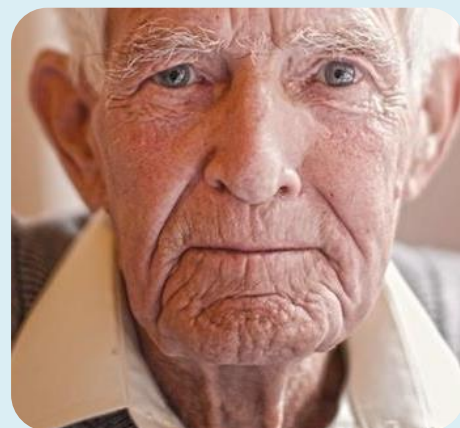
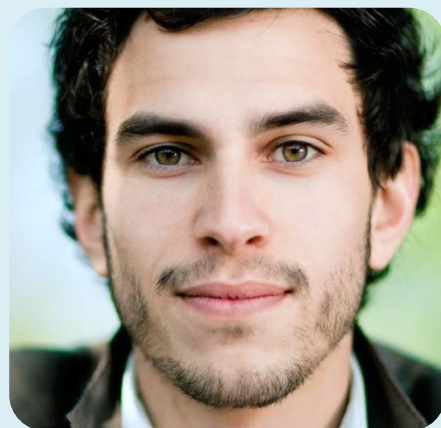
➡ všechny buňky vzniklé dělením **ZYGOTY** tvoří genetického jedince neboli **GENETU**

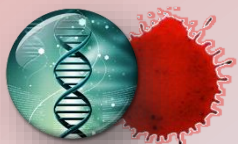




TESTOVÁNÍ INDIVIDUALITY DNA

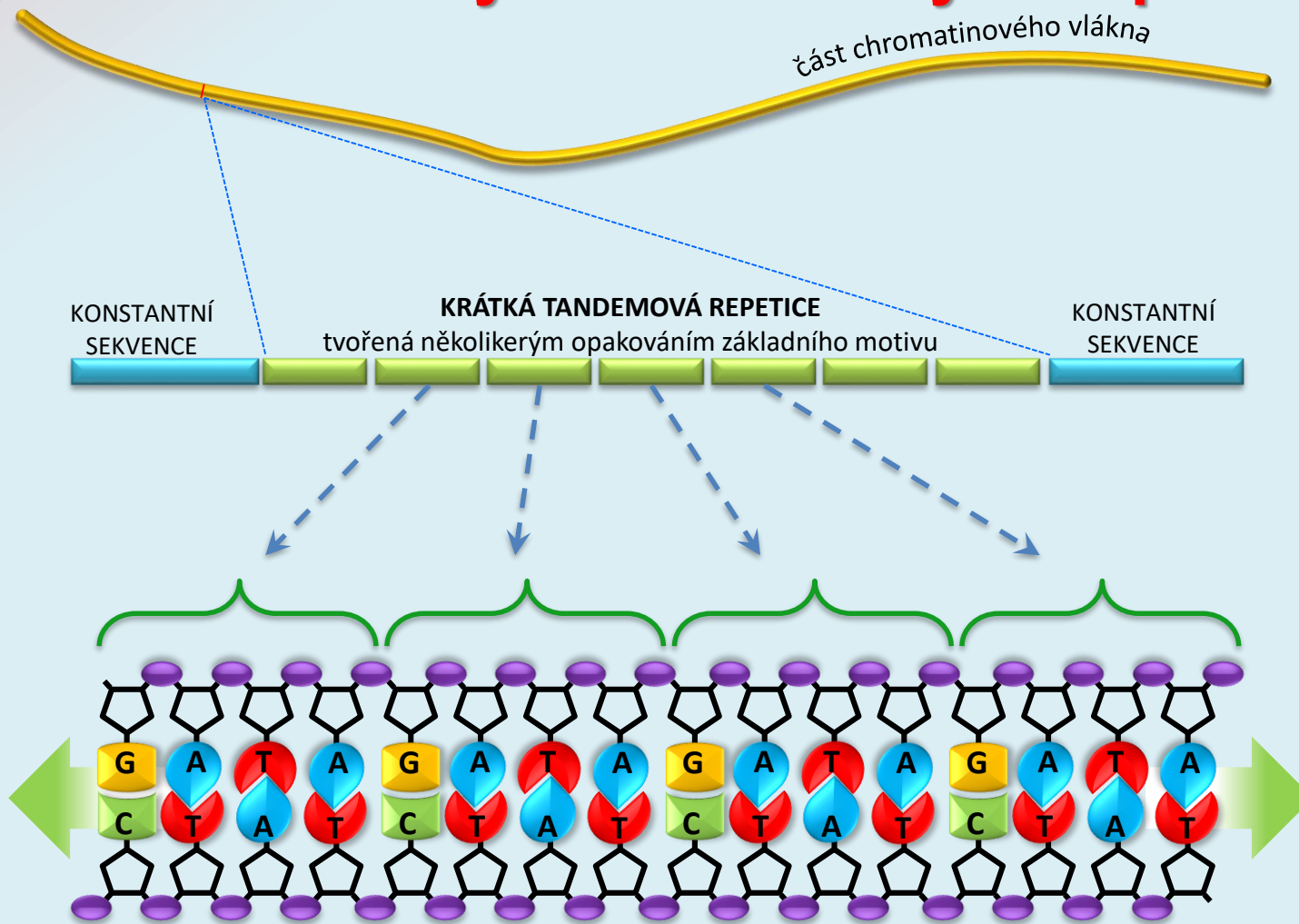
- ➡ forenzně genetickými metodami lze geneticky teoreticky identifikovat od okamžiku jejího vzniku a po celou dobu jejího trvání (ne nutně v živém stavu)

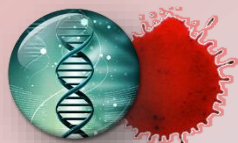




JAK NA TO?

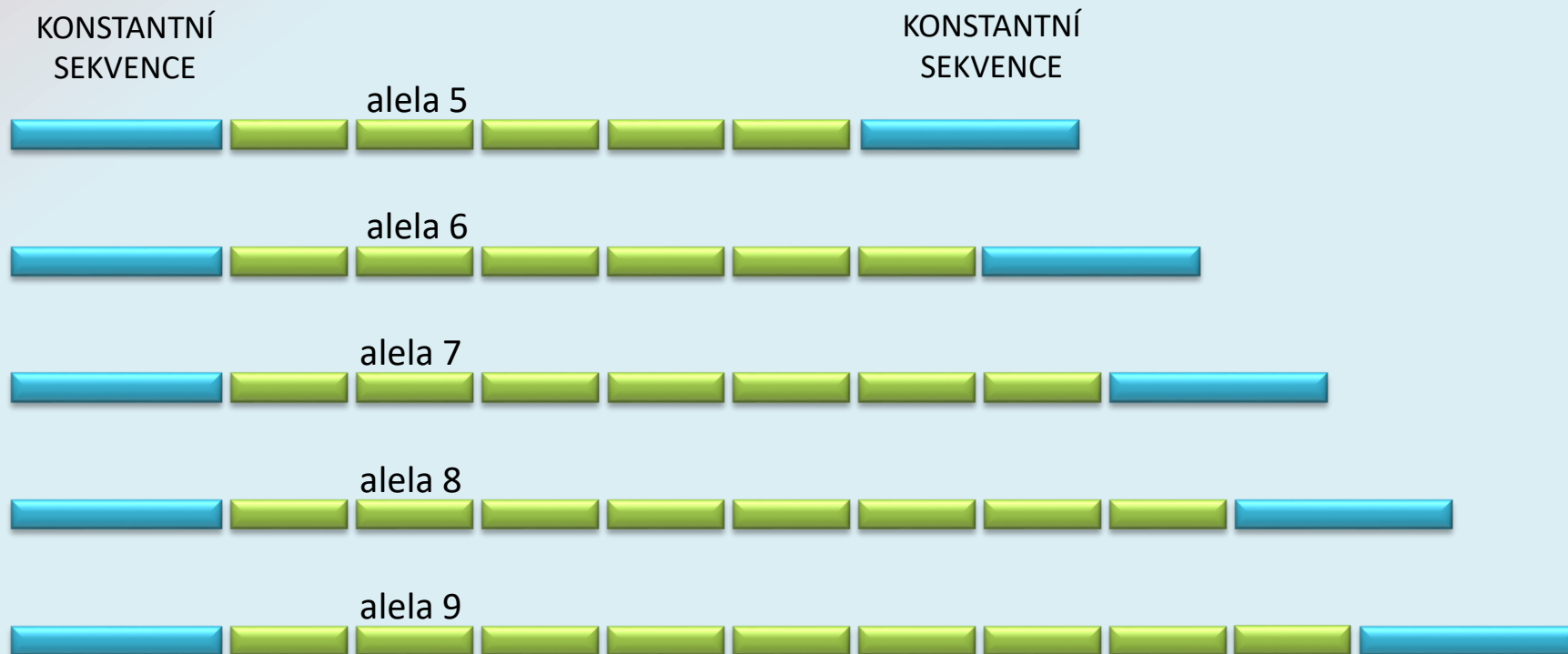
- ▶ hlavní identifikační metoda – analýza délkového polymorfismu **krátkých tandemových repetic (STR)**

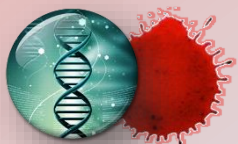




PROČ REPETICE?

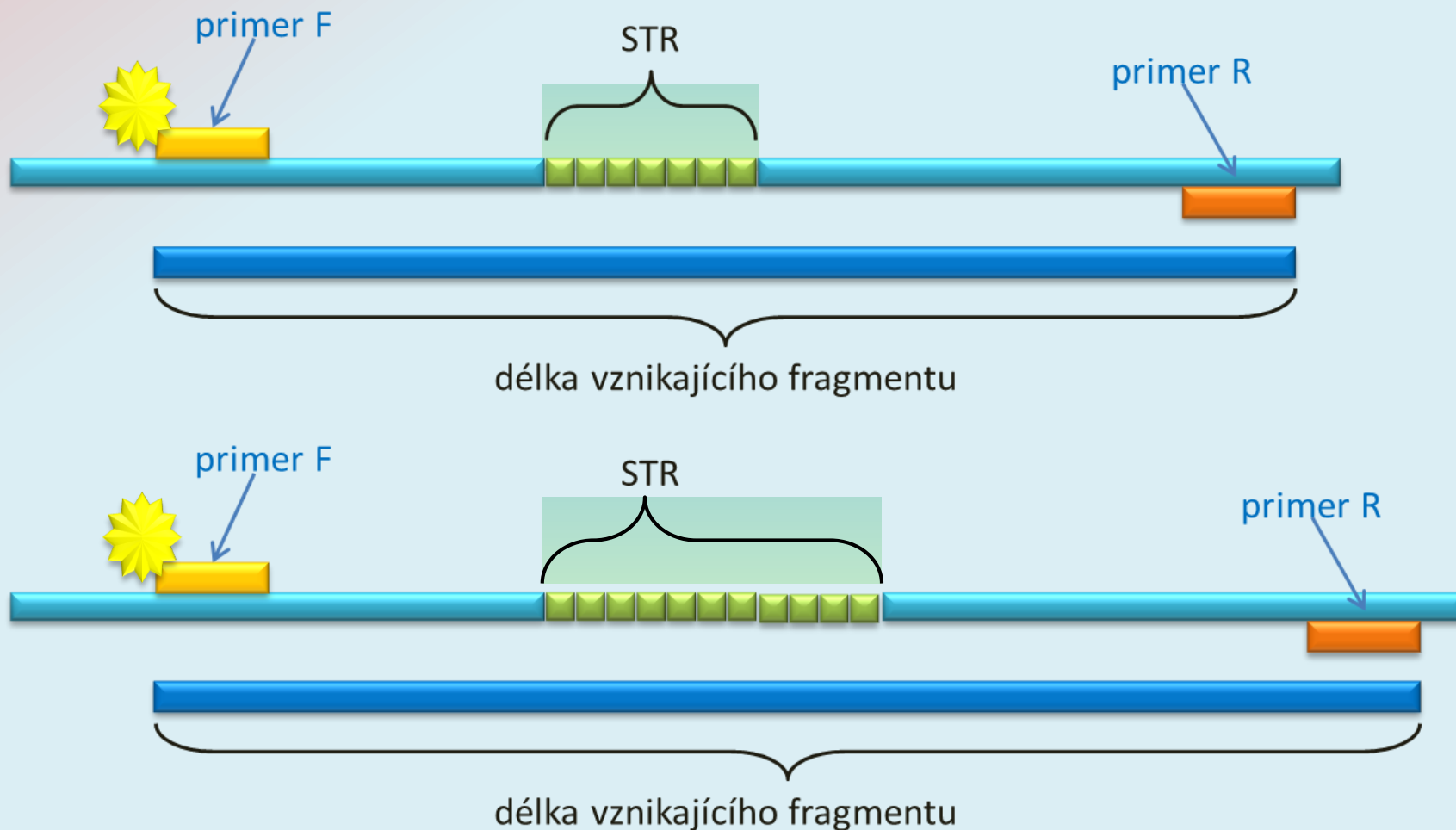
- ➡ lidé se liší tím, jak dlouhá u nich daná repetice je (tj. kolikrát se opakuje motiv)

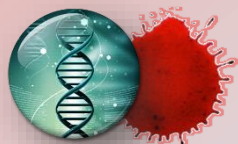




TECHNOLOGIE

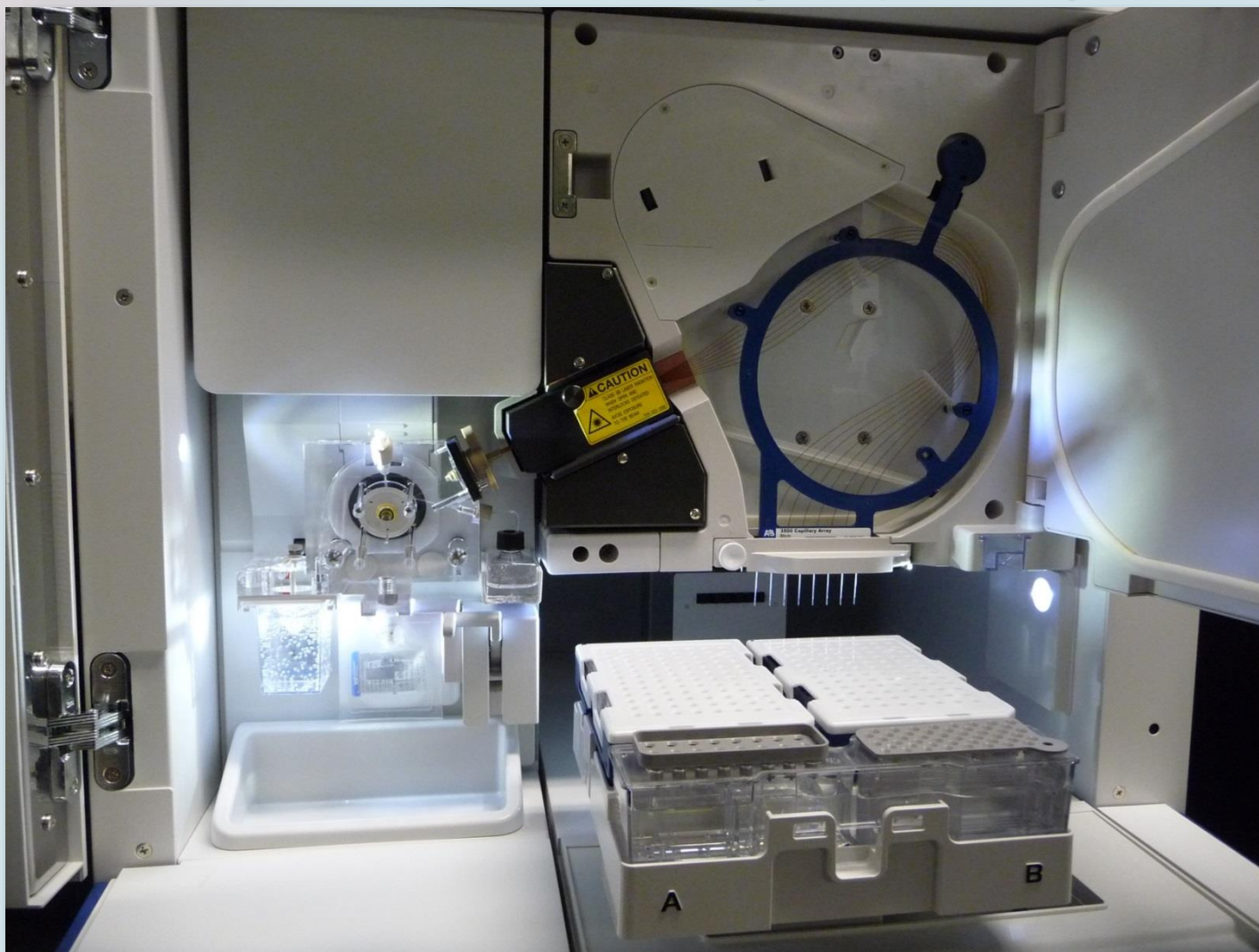
PCR s fluorescenčně značenými primery + CE

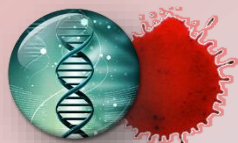




TECHNOLOGIE

- ➡ PCR s fluorescenčně značenými primery + CE





JAK ROZLIŠIT NA ÚROVEŇ INDIVIDUA

➡ genotypuje se set autozomálních STR lokusů

GENETICKÝ PROFIL

STR lokus 1



STR lokus 2

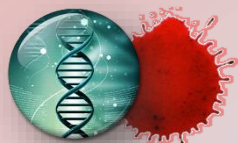


STR lokus 3



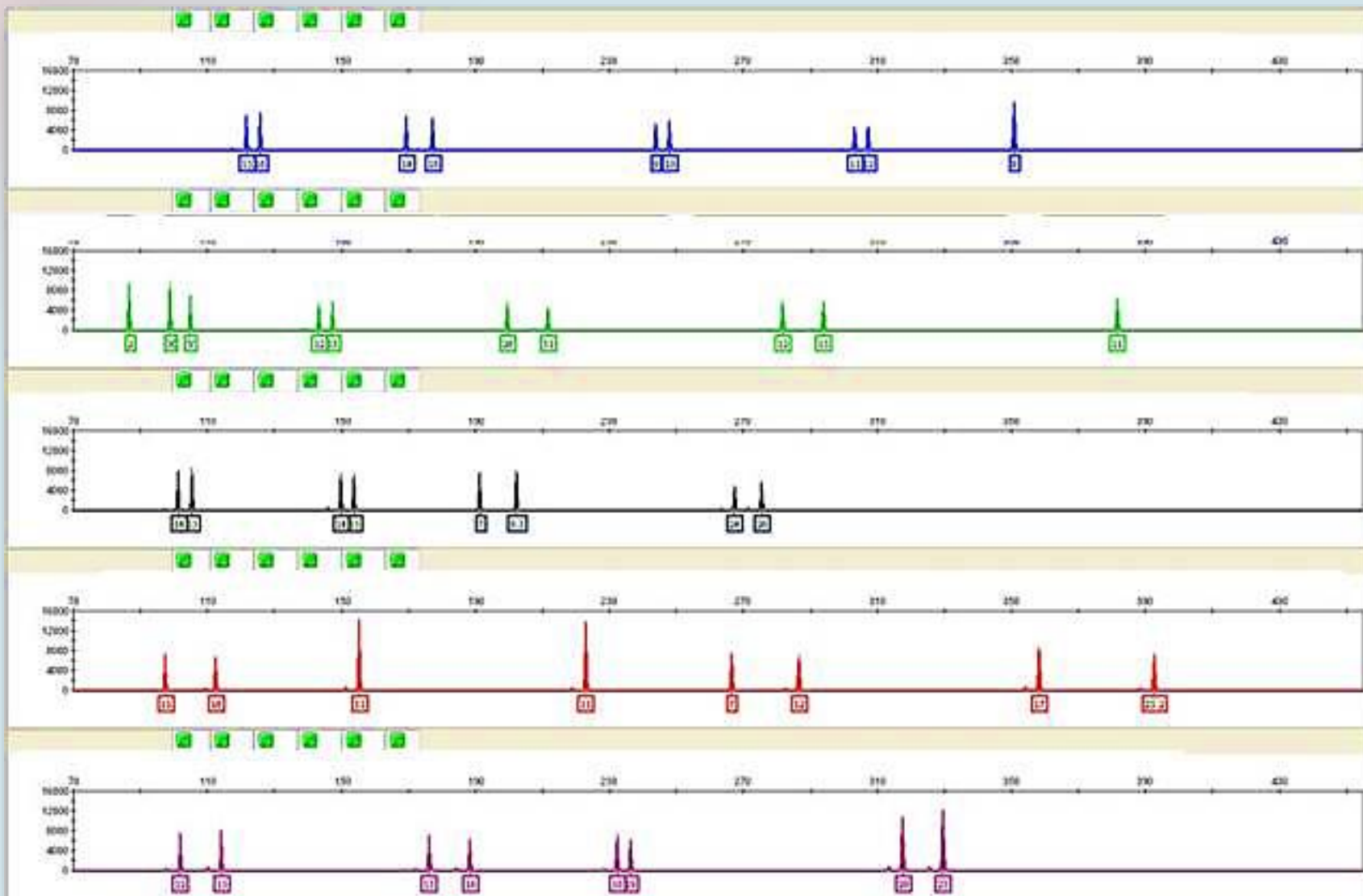
STR lokus 4

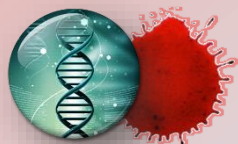




JAK ROZLIŠIT NA ÚROVEŇ INDIVIDUA

- ➔ současné metody analyzují řádově 25 STR lokusů



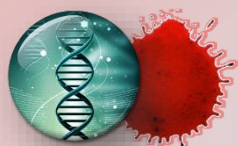


IDENTICKÁ VÍČERČATA

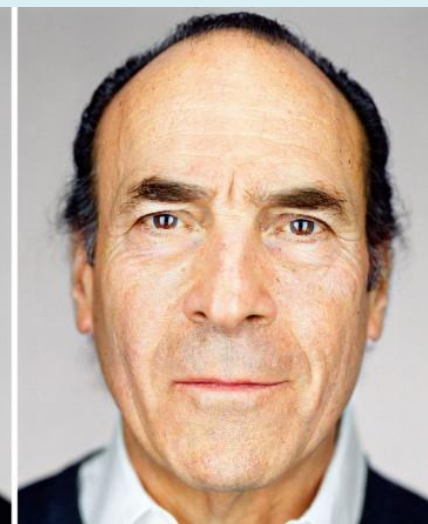
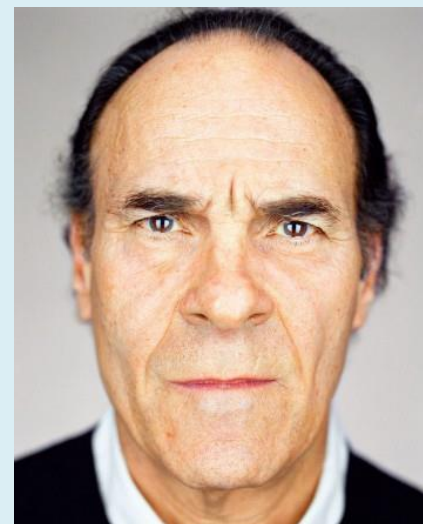
- ▶ jeden genetický jedinec, tj. **GENETA**, je vyjádřen více **RAMETAMI**, tj. fyzickými jedinci
- ▶ **problém!** analýza STR mezi nimi nerozliší



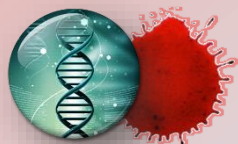
STR lokus 1	14	18	14	18
STR lokus 2	11	12	11	12
STR lokus 3	12	16	12	16
STR lokus 4	27	29	27	29



IDENTICKÁ VÍČERČATA



Martin Schoeller (photographer) – „IDENTICAL“



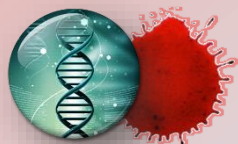
IDENTICKÁ VÍČERČATA? – NE!!!



➡ **nepříbuzné osoby!**

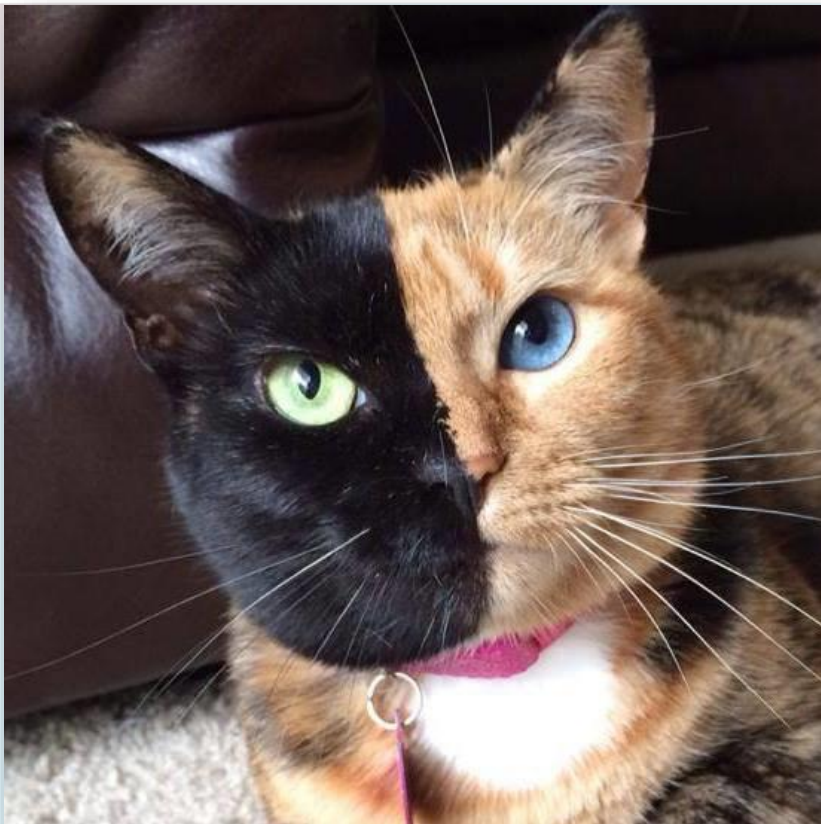
François Brunelle (photographer) – „I'M NOT A LOOK-LIKE!“





CHIMÉRY

► jeden fyzický jedinec, tzv. **RAMETA**,
je tvořen více **GENETAMI**

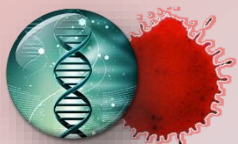


tetragametický chimérismus (tetragametic chimerism)

Lydia Fairchild # Karen Keegan

cat Venus



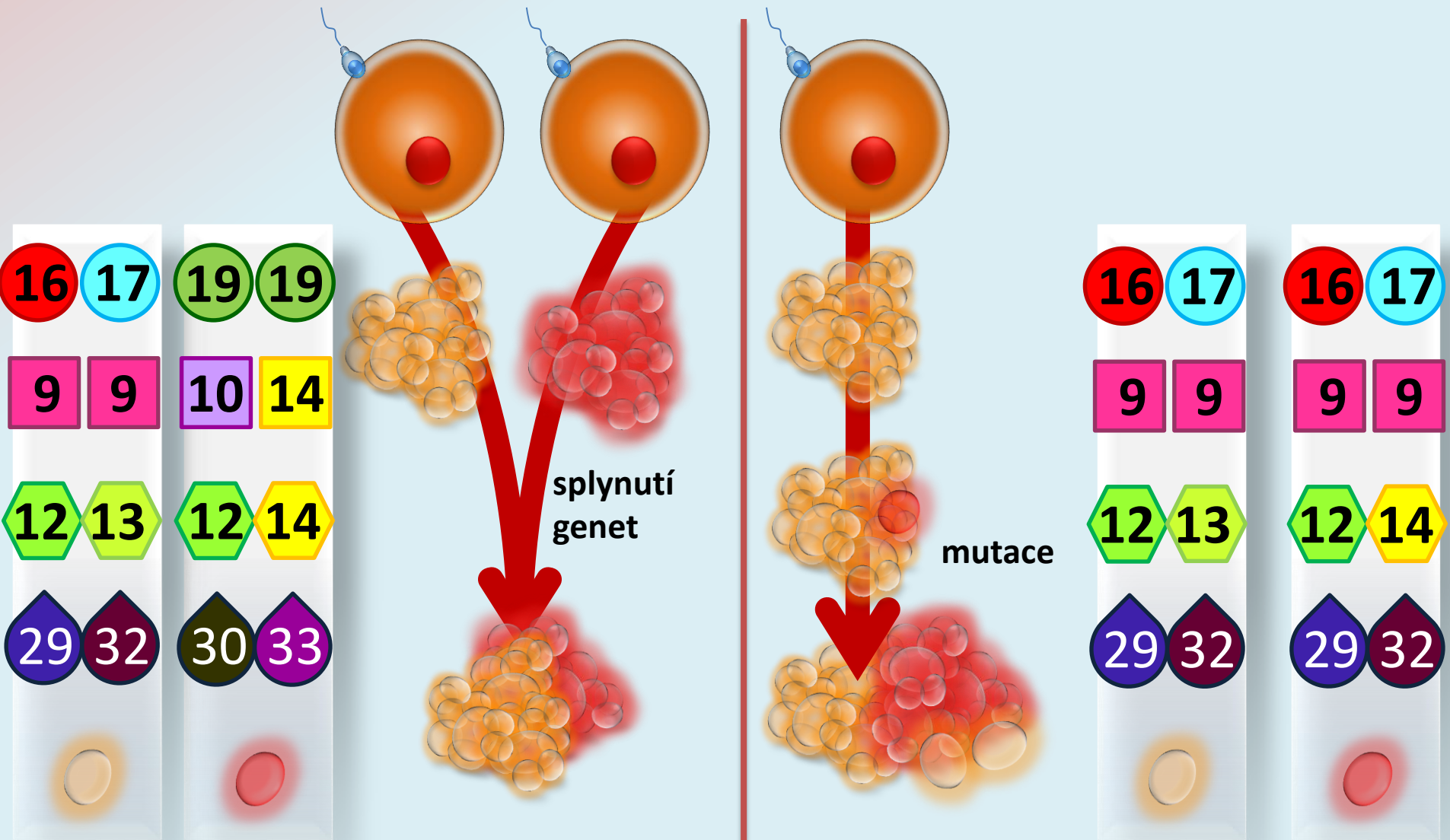


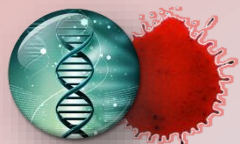
CHIMÉRY

chiméra

vs.

mozaika

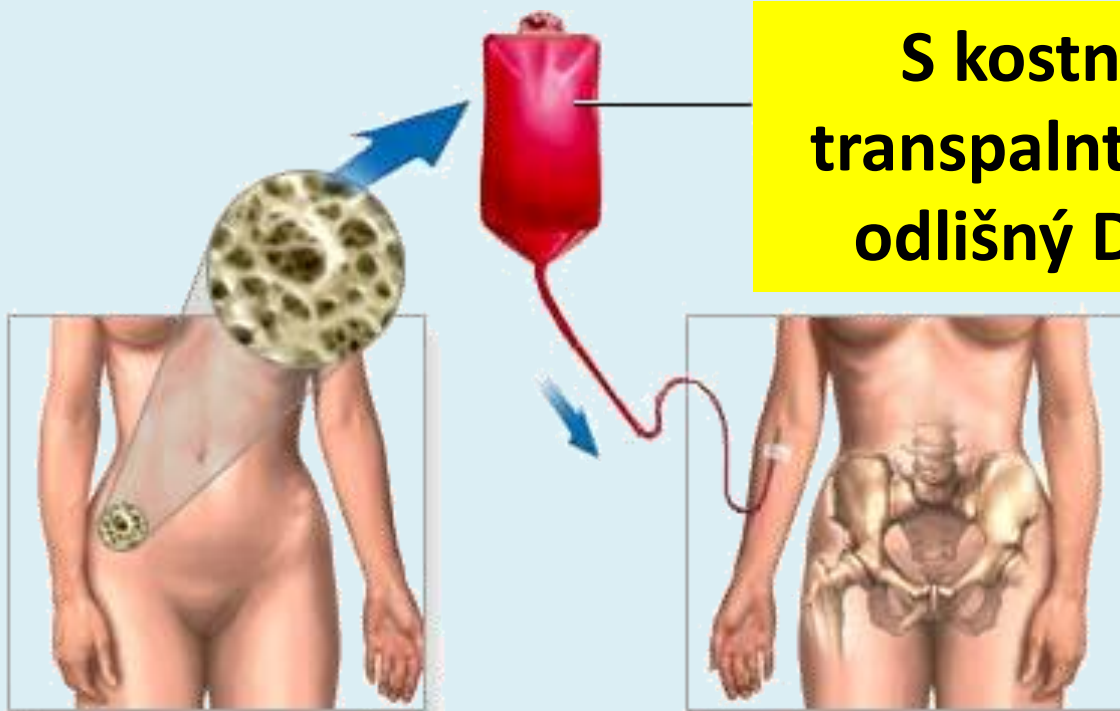


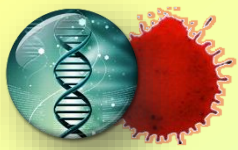


CHIMÉRY

➡ další typy chimérismu

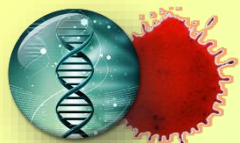
- ➡ fetální mikrochimérismus
- ➡ maternální mikrochimérismus
- ➡ krevní chimérismus fraternálních dvojčat
- ➡ **posttransplantační chimérismus**





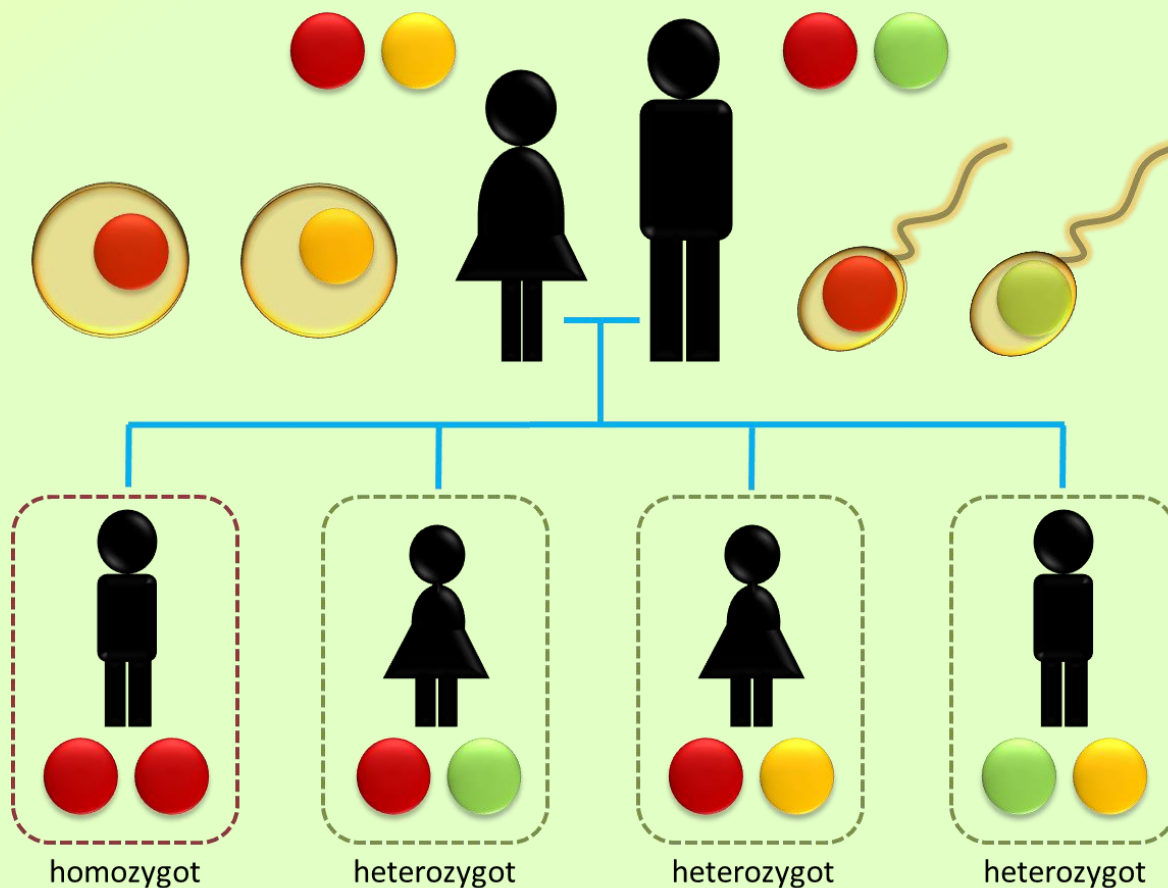
2. PŘÍBUZNOST A RODOVOST

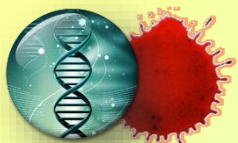




PATERNITA A MATERNITA

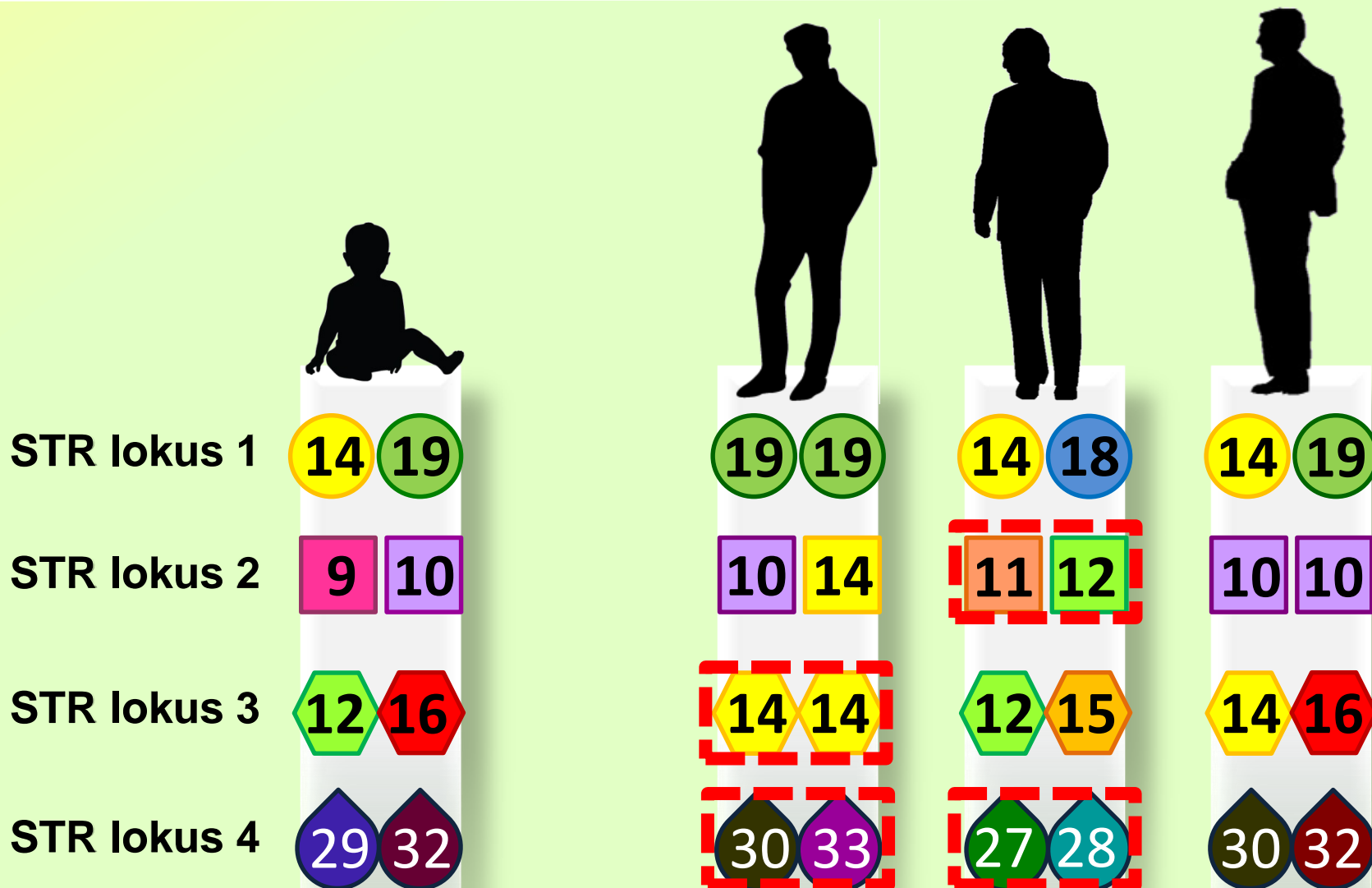
► blízkou příbuznost osob lze dobře hodnotit pomocí nám již známých autozomálních STR

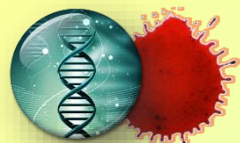




PATERNITA A MATERNITA

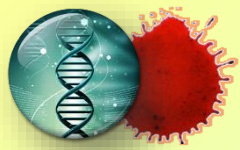
➡ rodič a dítě musejí v každém lokusu sdílet alespoň 1 alelu





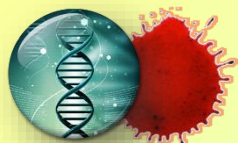
PROBLÉM: IDENTICKÁ DVOJČATA





PROBLÉM: IDENTICKÁ DVOJČATA





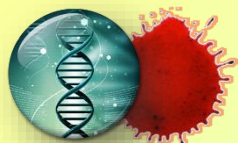
FRATERNÁLNÍ DVOJČATA

► geneticky úplní sourozenci, ale ne vždy...

► **heteropaternální superfekundace**



Koen & Tuen Stuart



FRATERNÁLNÍ DVOJČATA

➡ geneticky úplní sourozenci, ale ne vždy...

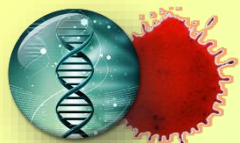
➡ **heteropaternální superfekundace? – ne!!!**



➡ geneticky úplné sestry

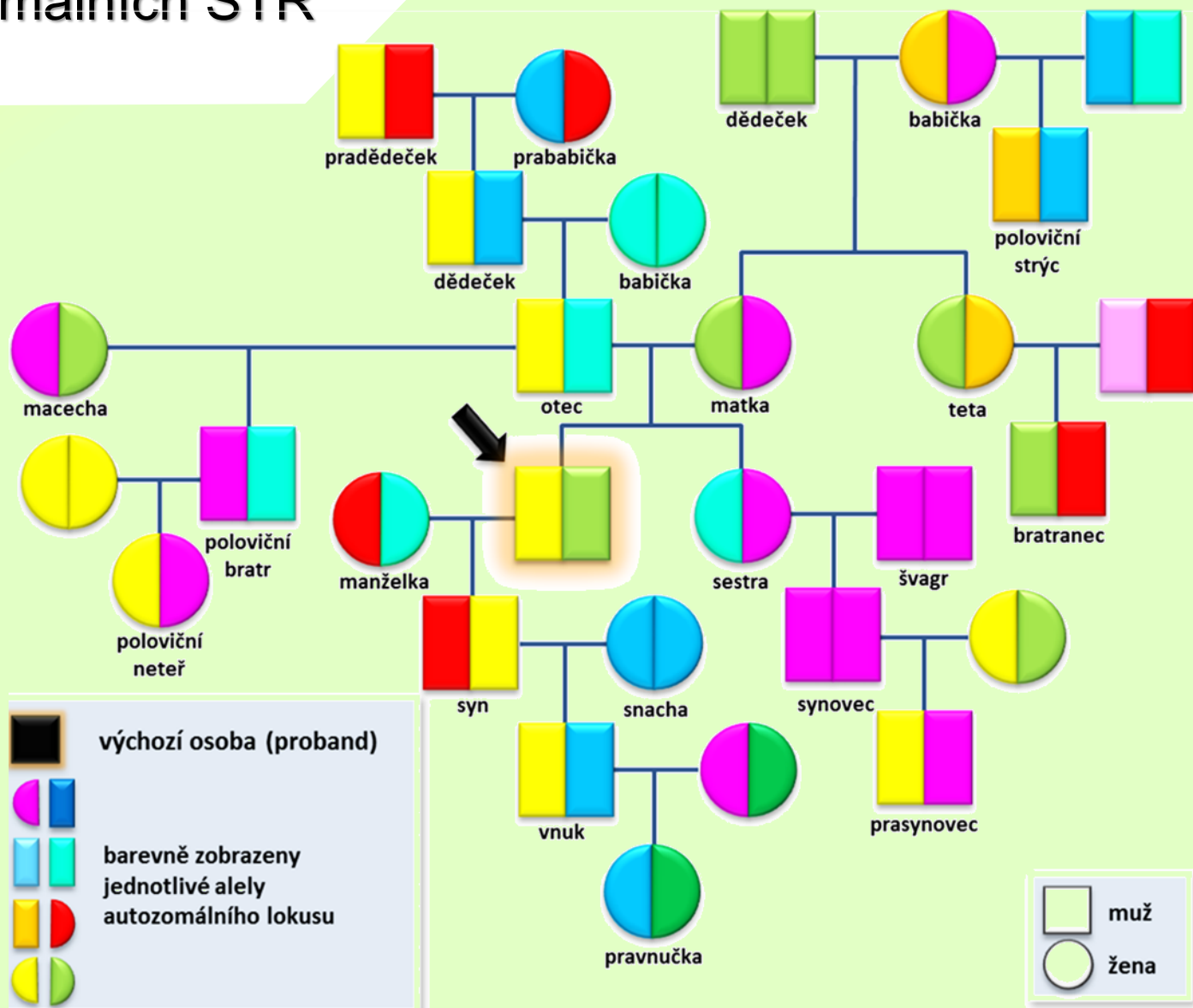


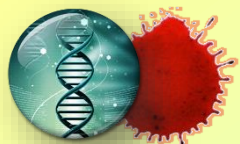
Kian & Remea Hodgson



VZDÁLENĚJŠÍ PŘÍBUZNOST

► pomocí autozomálních STR
neprůkazná

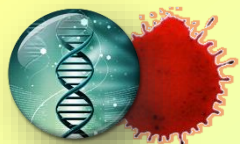




RODOVÉ MARKERY

maternální linie: mtDNA

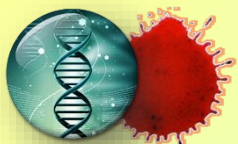




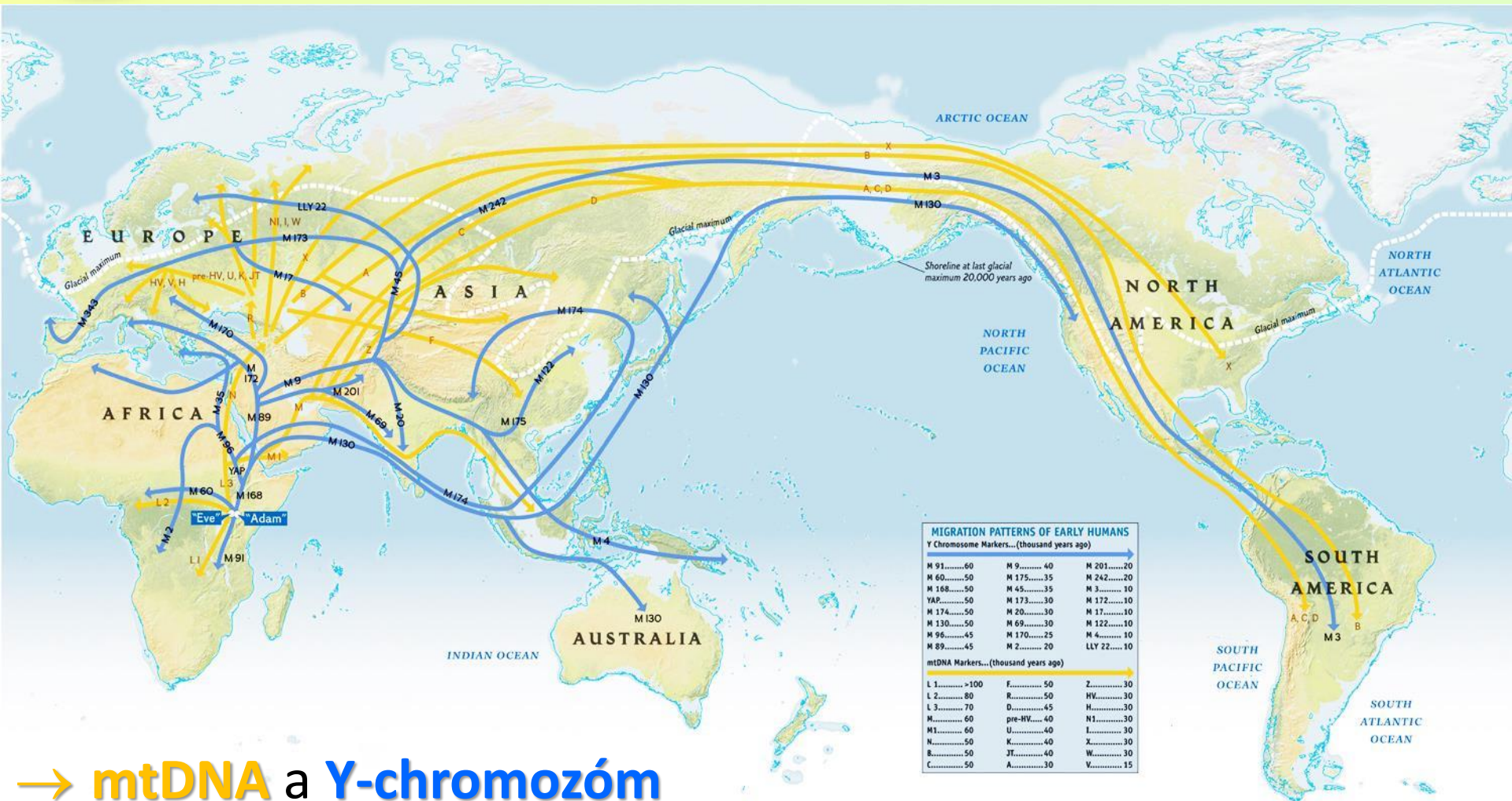
RODOVÉ MARKERY

paternální linie: Y-chromozóm



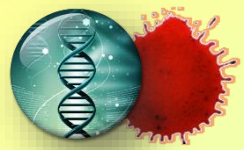


RODOVÉ MARKERY

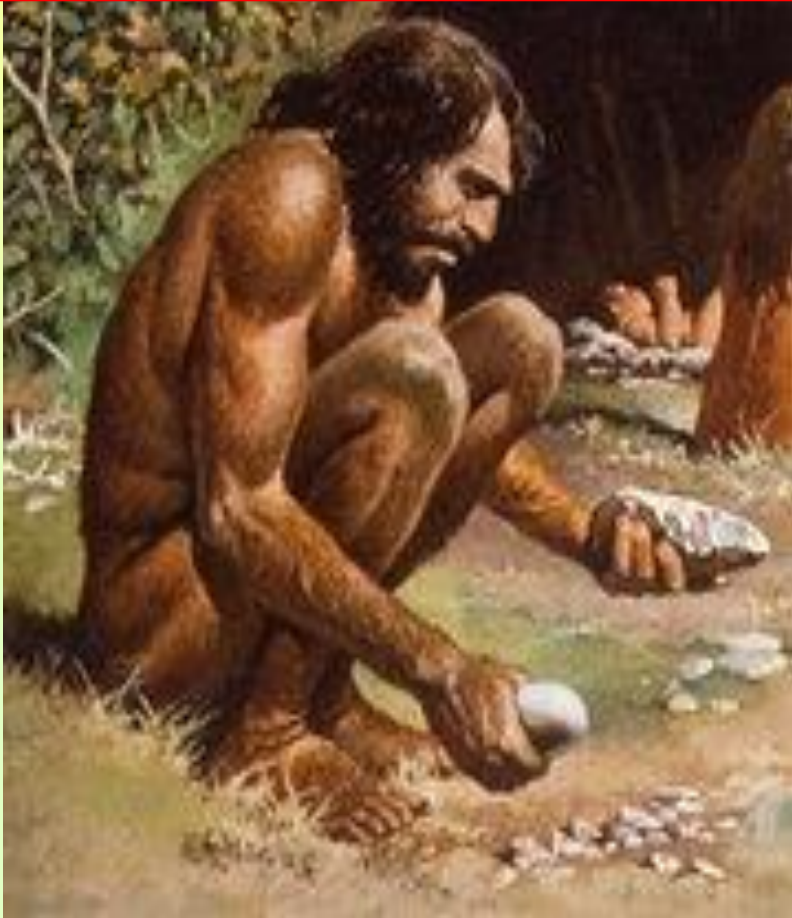


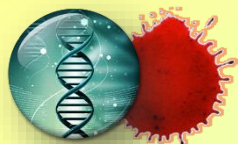
http://www.pro-classic.com/ethnicgv/vivi/ancestry5_medium.jpg

→ mtDNA a Y-chromozóm



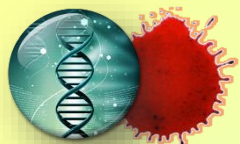
Y-chrz-Adam a mtDNA-Eva





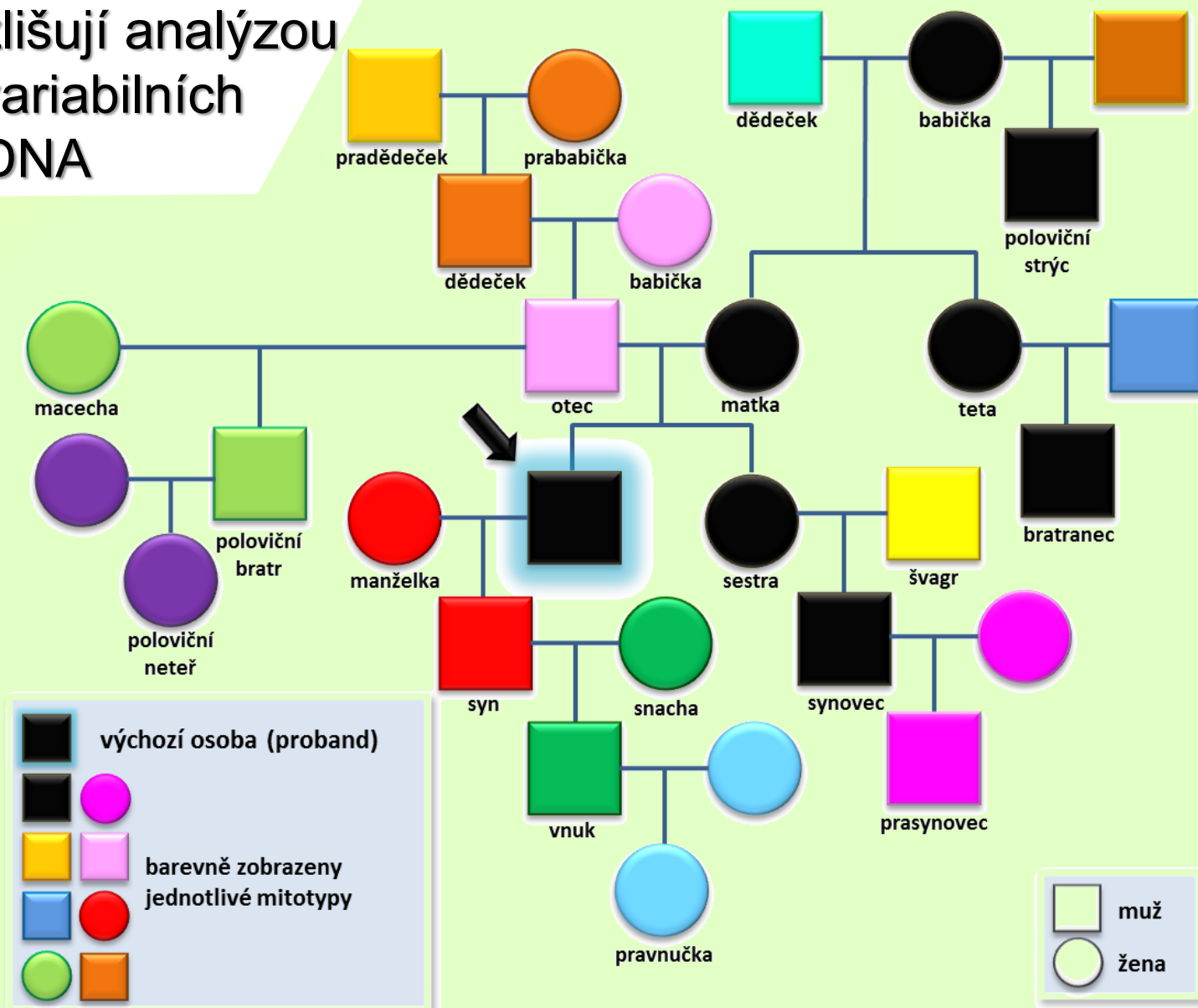
mtDNA LINIE

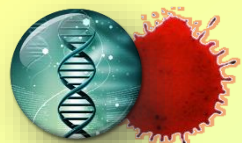




RODOVÉ MARKERY - mtDNA

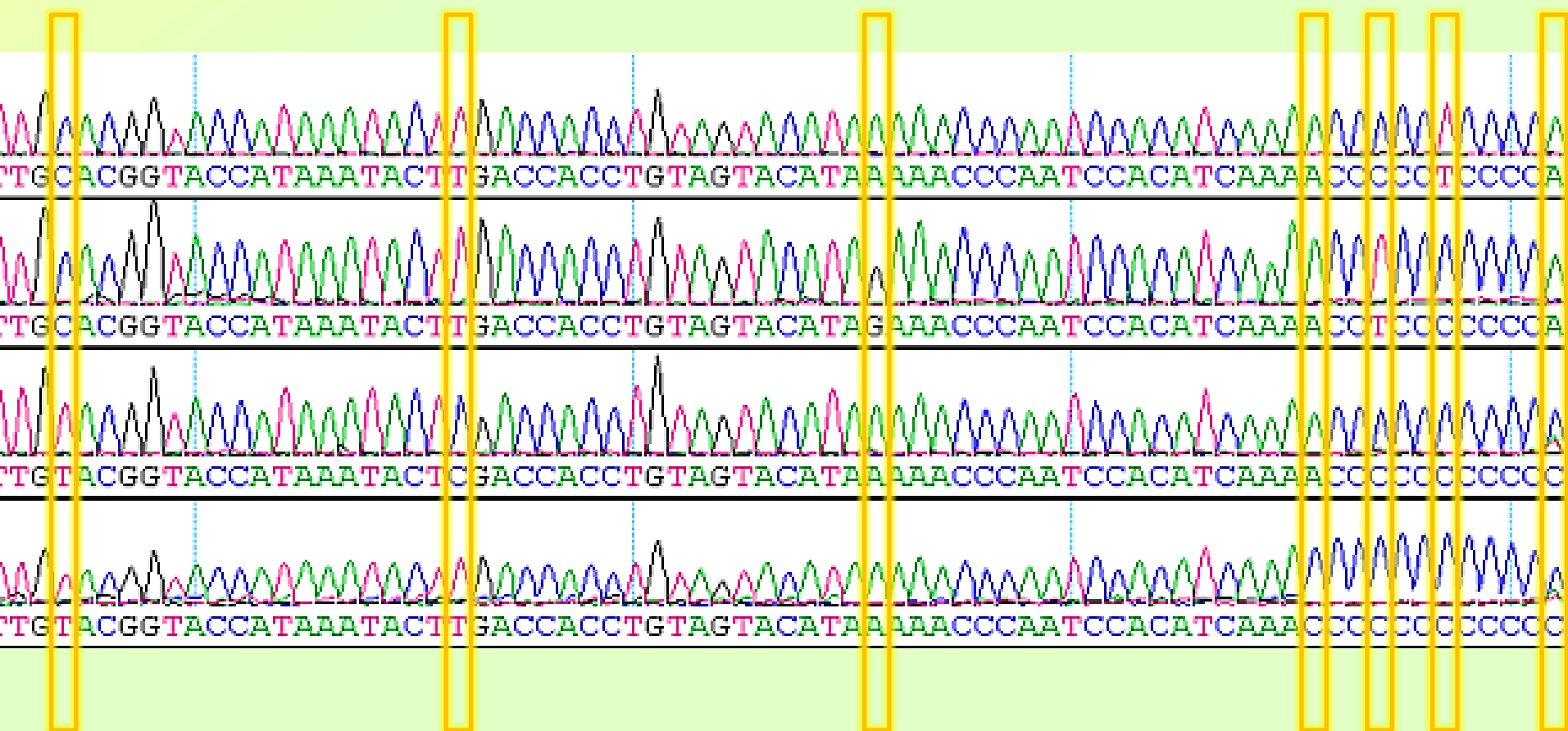
➔ linie se rozlišují analýzou tzv. hypervariabilních oblastí mtDNA

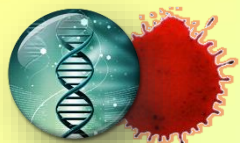




RODOVÉ MARKERY - mtDNA

- ➔ sekvenační analýza nebo analýza setu vybraných nukleotidů

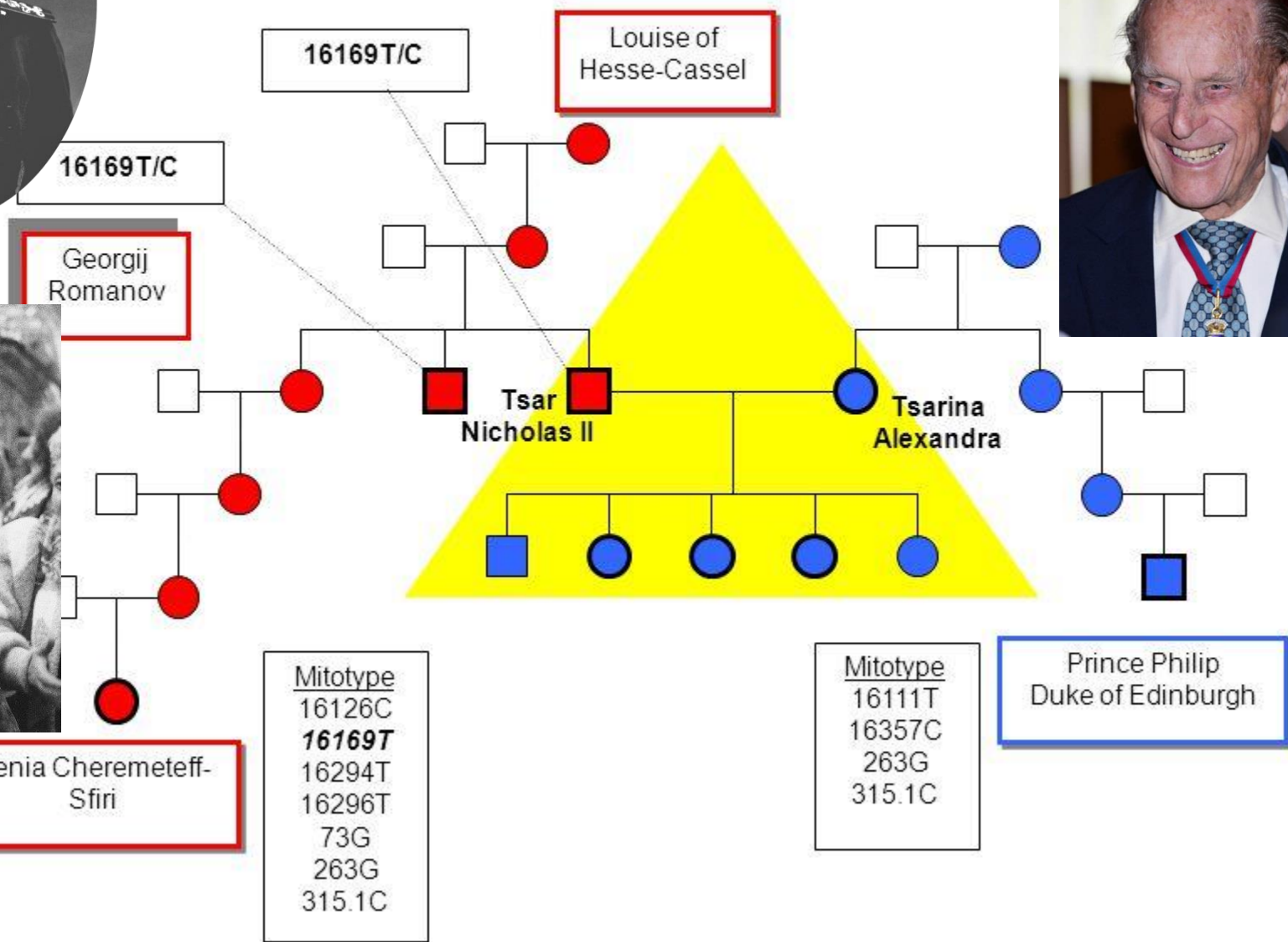




Identifikace carské rodiny

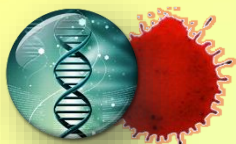


Lineage of Romanov Family



c DNA Typing

John M. Bu

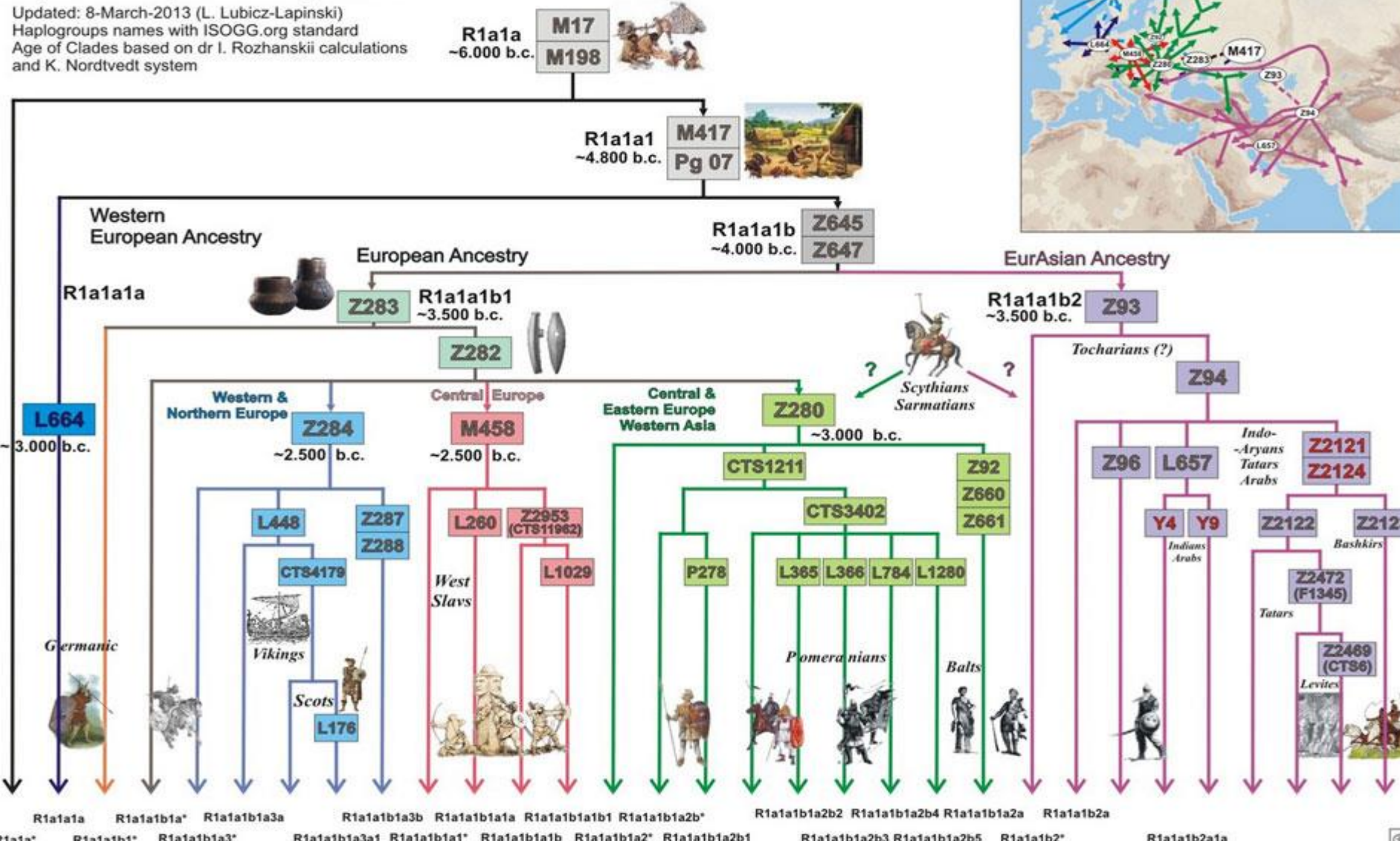
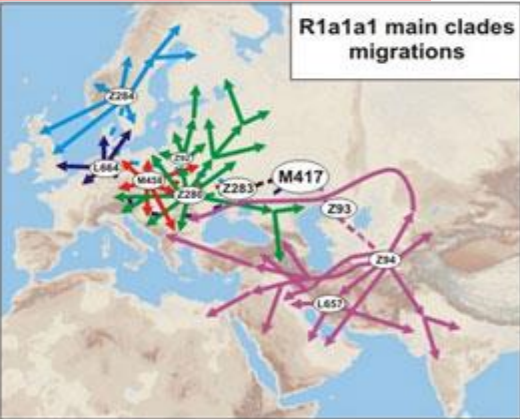


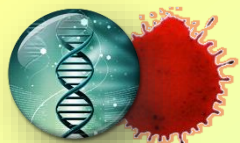
RODOVÉ MARKERY – Y

R1a1a Clades (by G. F. Underhill)

<http://www.familytreedna.com/public/R1a>

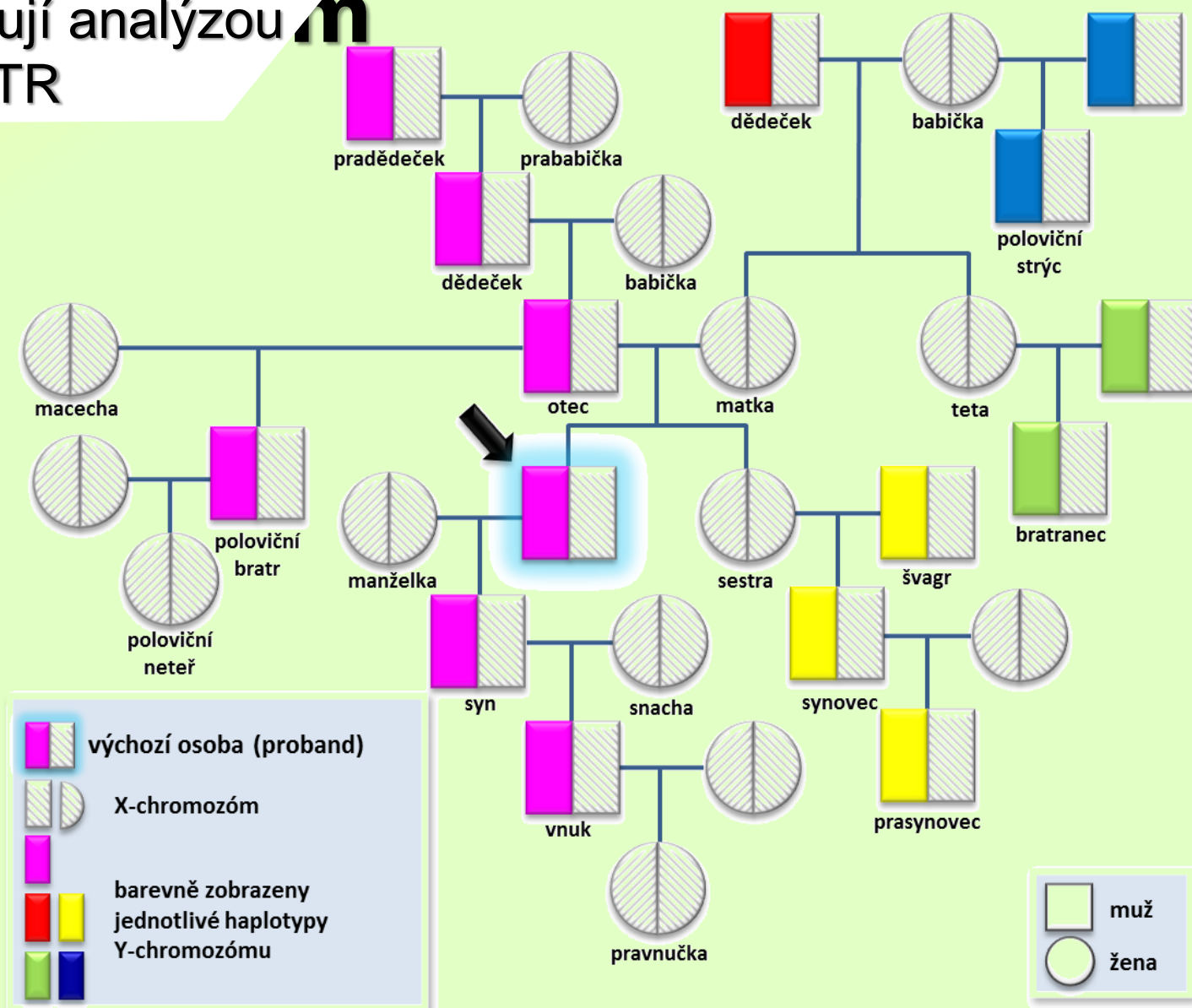
Updated: 8-March-2013 (L. Lubicz-Lapinski)
 Haplogroups names with ISOGG.org standard
 Age of Clades based on dr I. Rozhanskii calculations
 and K. Nordtvedt system

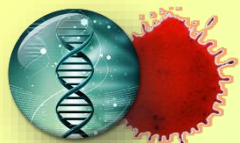




RODOVÉ MARKERY – Y

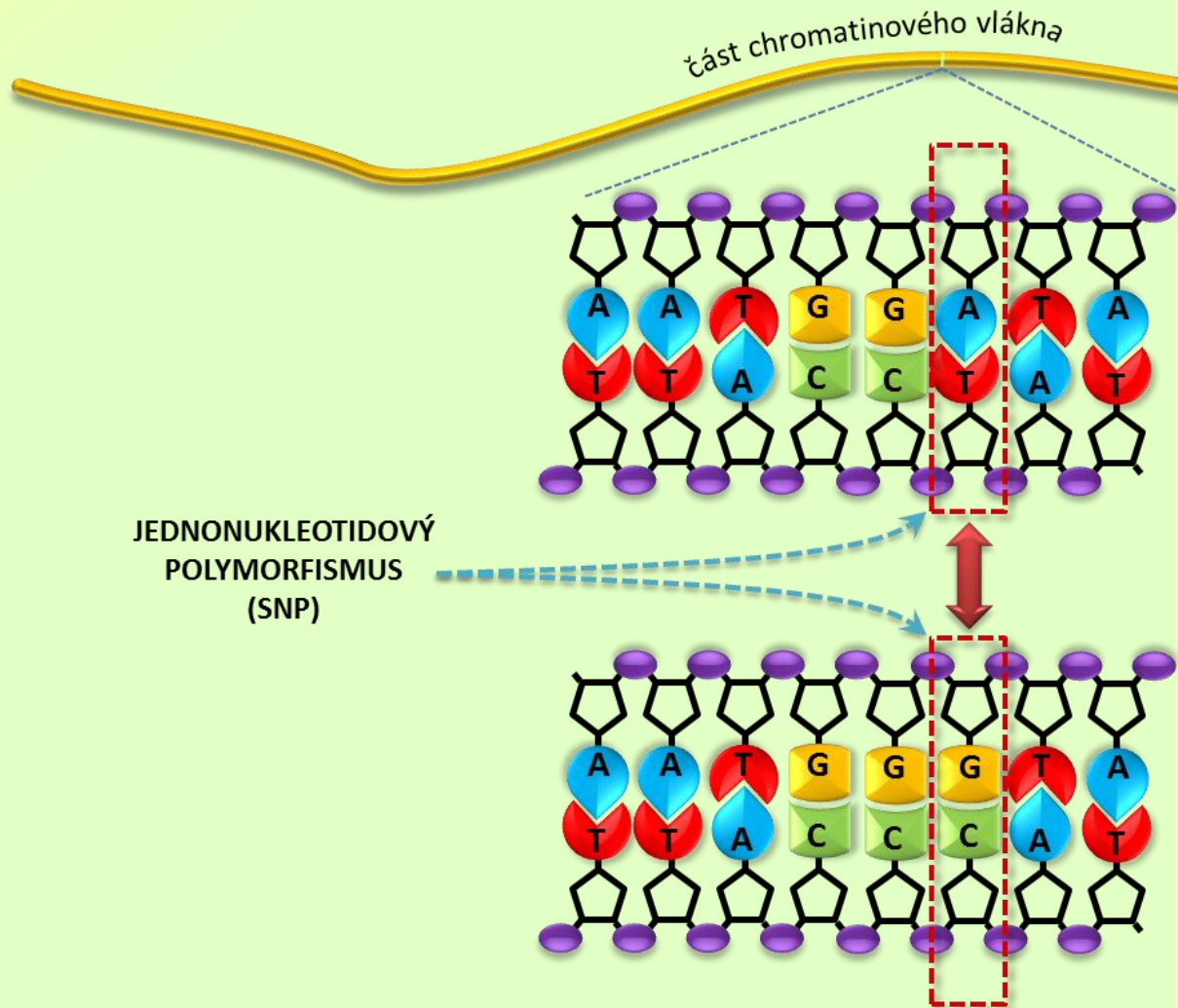
➔ linie se rozlišují analýzou **m**
Y-SNP a Y-STR

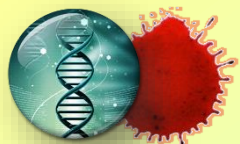




RODOVÉ MARKERY

➡ analýza bodového polymorfismu (SNP)

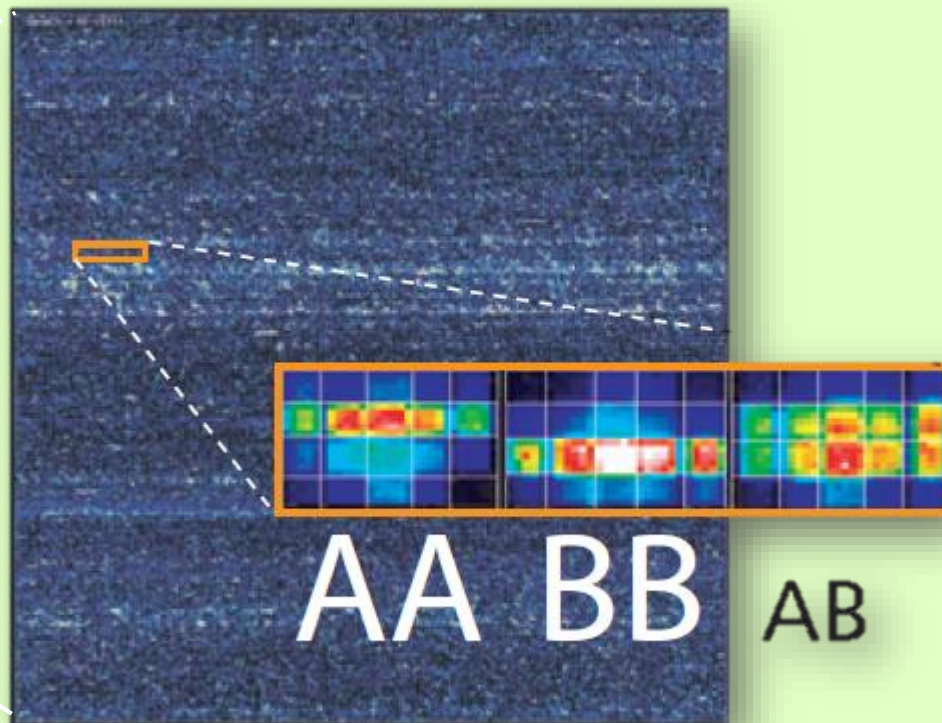
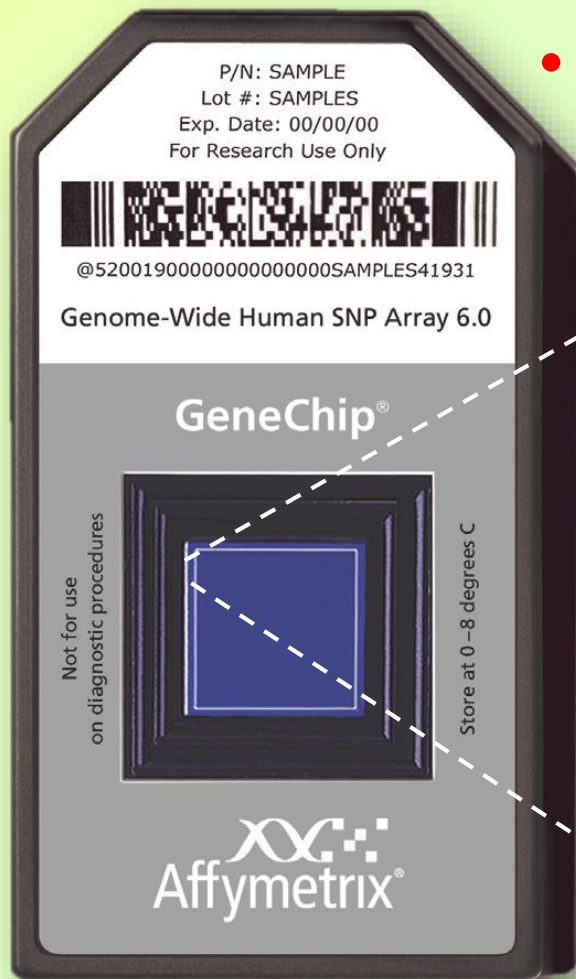


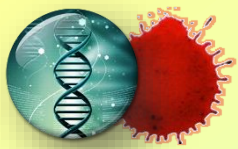


RODOVÉ MARKERY

➔ analýza bodového polymorfismu (SNP)

- velkokapacitní biočipy
- mnoho jiných technik (SNaPshot)





GAP – genetika a příjmení

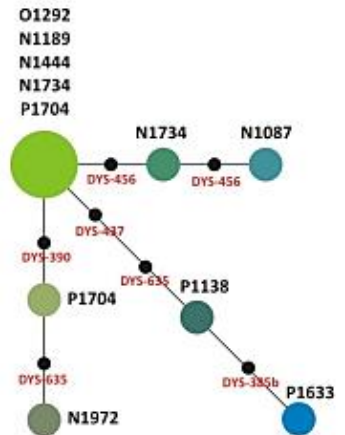
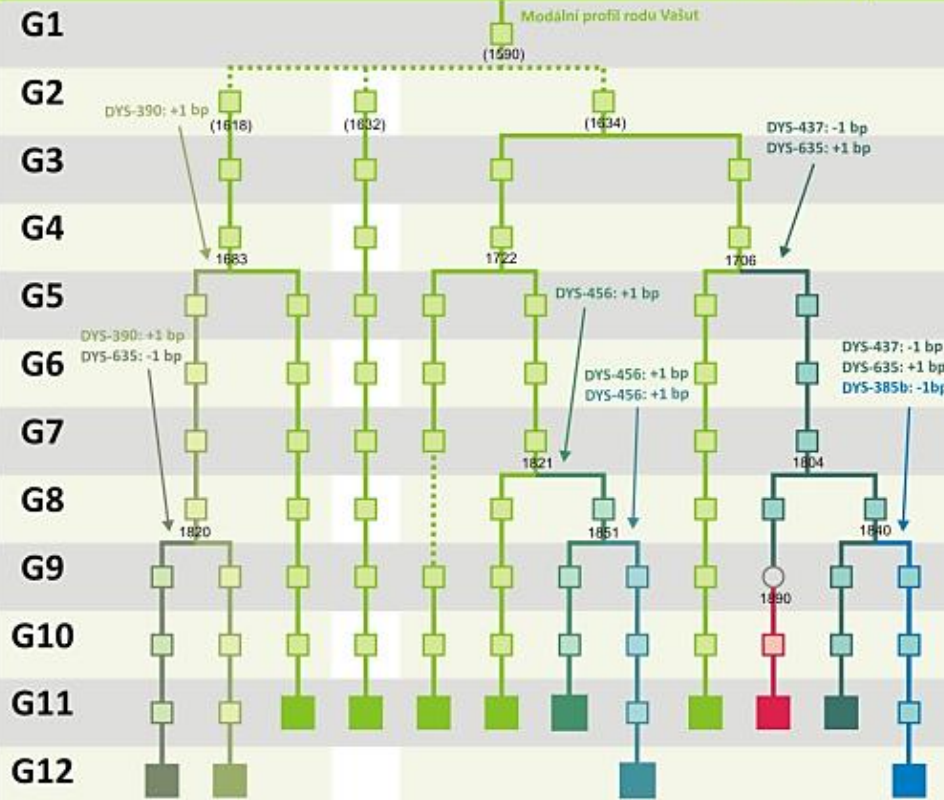
Rodokmenové schéma rodu VAŠUT (větev Rožnovská)

Genetika a Příjmení

Stav k 8.12.2009

I2a2

(ověřeno bodovou mutací v M423)



Zjednodušené schéma MJN grafu pro rod Vašut (rožnovská větev).

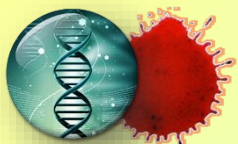
Černé body představují jednotlivé mutace. Barvy se shodují s rodovými větvemi v rodokmenovém schématu.

N1972 P1704 O1292 N1189 N1444 N1521 N1734 N1087 P1677 P1521 (R1a) P1138 P1633

Velké Karlovice

Dolní Bečva

Prostřední Bečva (Vašůtky)
Horní Bečva (Pod Vysokou)

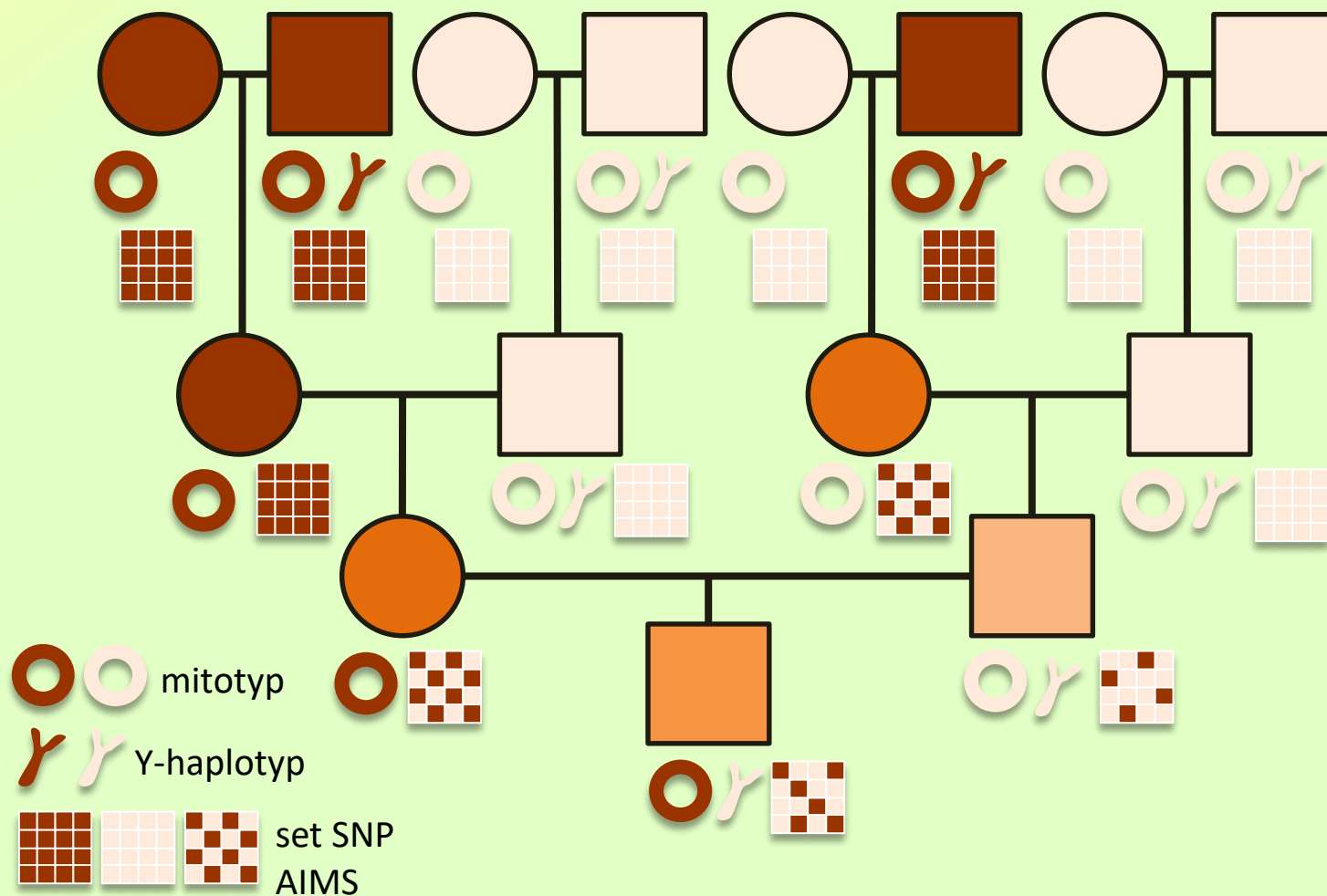


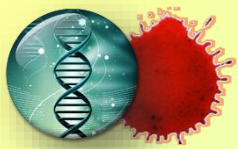
VELMI VZDÁLENÁ

PŘÍBUZNOST

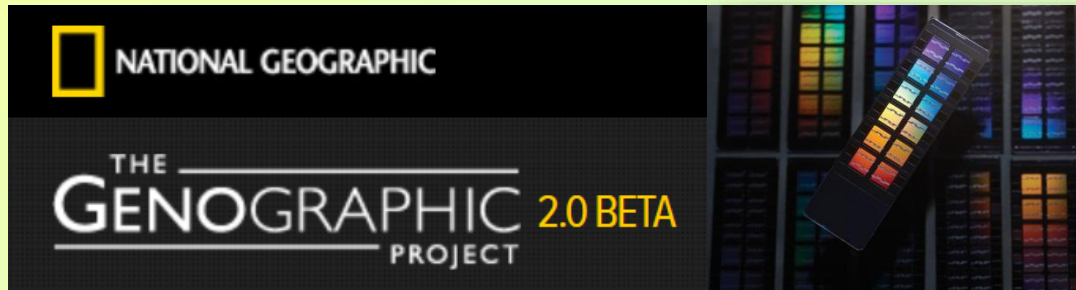
➔ komplexní biogeografický původ

- autozomální SNP





GENOGRAFIE



GenoChip

cca 150.000 SNP

autozómy, Y-SNP, mtDNA

1 YOUR RESULTS



NORTHERN EUROPEAN

This component of your ancestry is found at highest frequency in northern European populations—people from the UK, Denmark, Finland, Russia and Germany in our reference populations. While not limited to these groups, it is found at lower frequencies throughout the rest of Europe. This component is likely the signal of the earliest hunter-gatherer inhabitants of Europe, who were the last to make the transition to agriculture as it moved in from the Middle East during the Neolithic period around 8,000 years ago.



YOUR HOMINID ANCESTRY

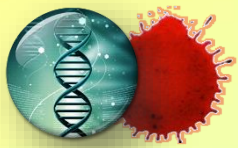
When our ancestors first migrated out of Africa around 60,000 years ago, they were not alone. At that time, at least two other species of hominid cousins walked the Eurasian landmass: Neanderthals and Denisovans. Most non-Africans are about 2% Neanderthal. Indigenous sub-Saharan Africans have no Neanderthal DNA because their ancestors did not migrate through Eurasia.



LEARN MORE >

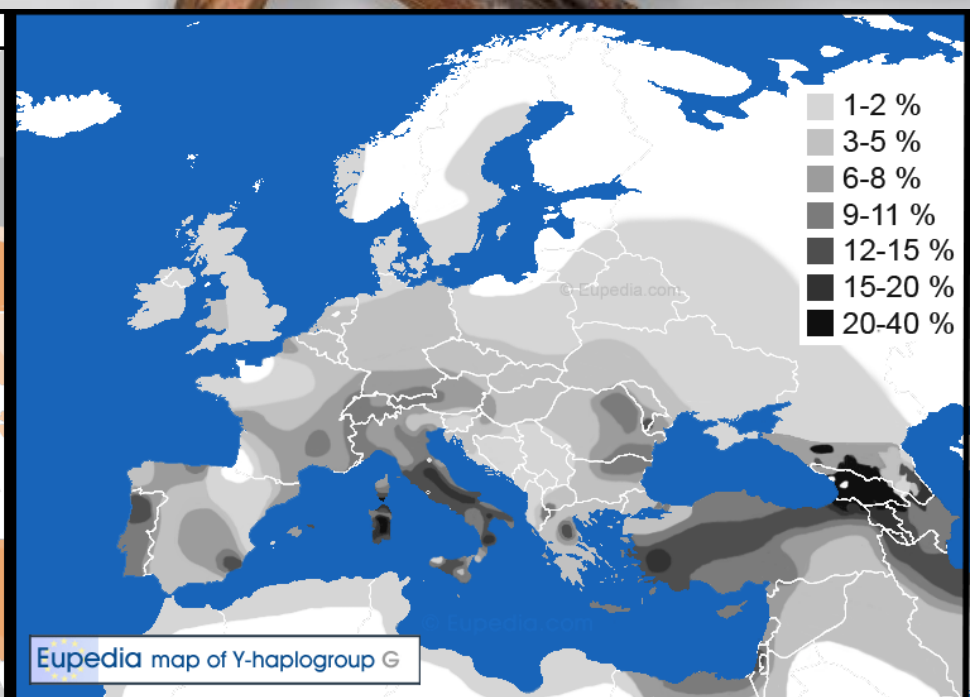
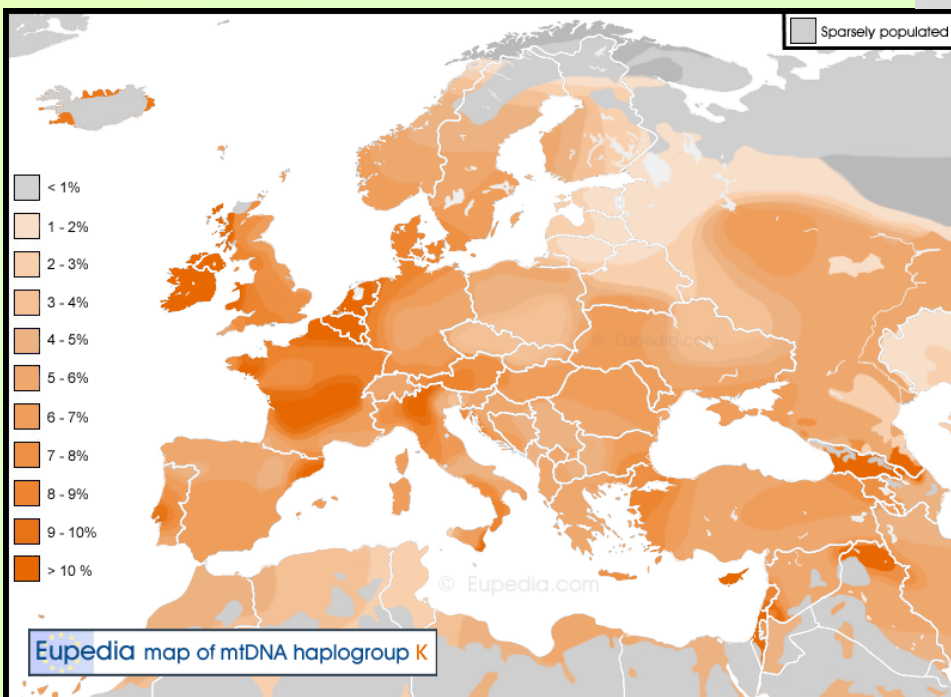
LEARN MORE >

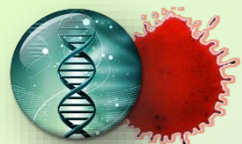
- paternální a maternální linie
- komplexní biogeografický původ
- příspěvek genofondu hominidů



ÖTZI

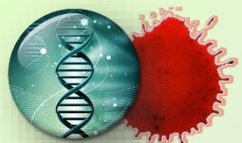
- mtDNA- haploskupina K1
- Y-haploskupina G2a2b





3. KRIMINALISTICKÉ ANALÝZY





3. KRIMINALISTICKÉ ANALÝZY

- ➡ především identifikace a určování příbuznosti pro potřeby objasnění trestných činů



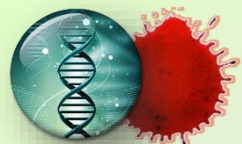
➡ BIOLOGICKÉ STOPY

- DNA různé kvality i kvantity
- často smíšené stopy
- unikátní, nelze je získat znovu

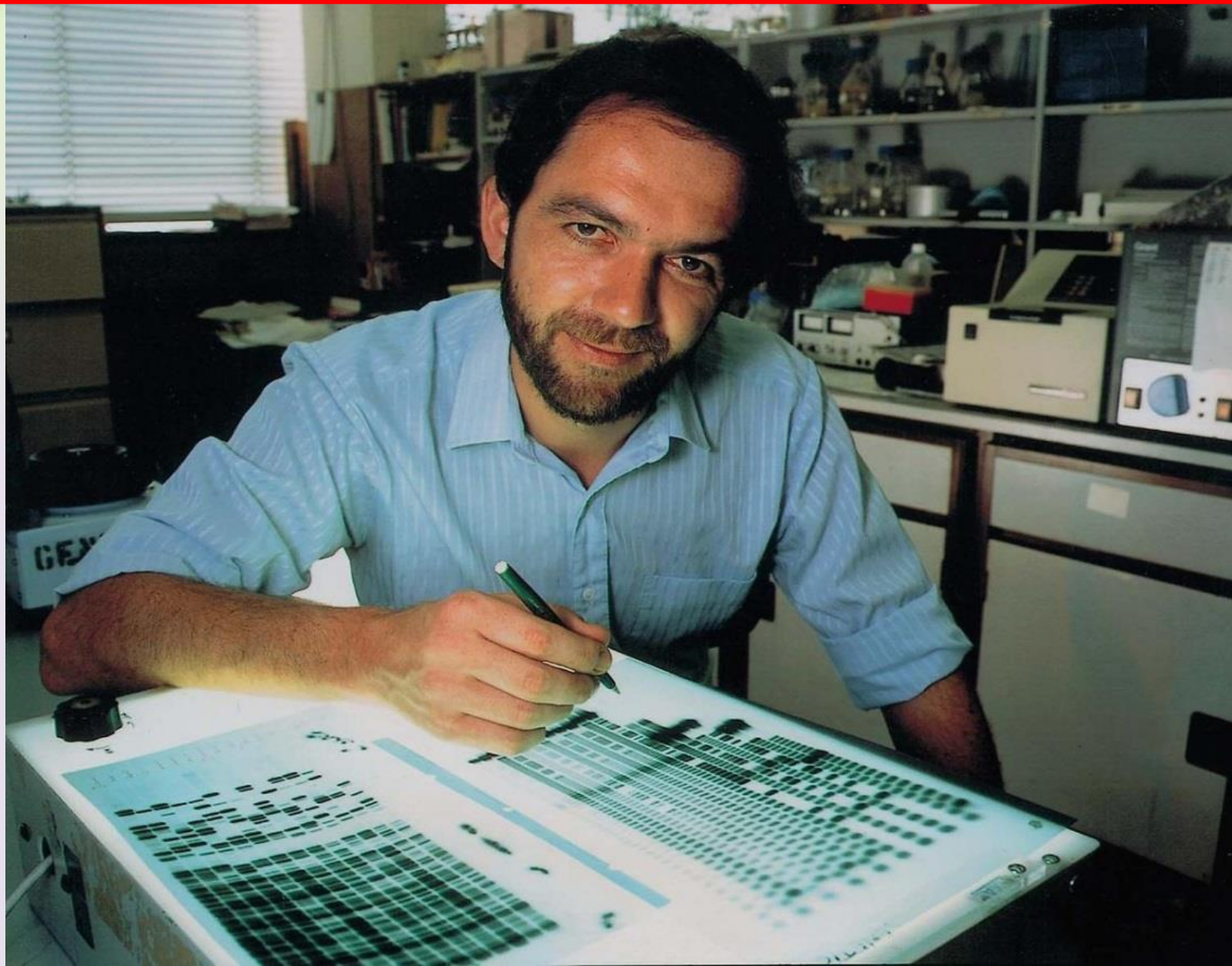


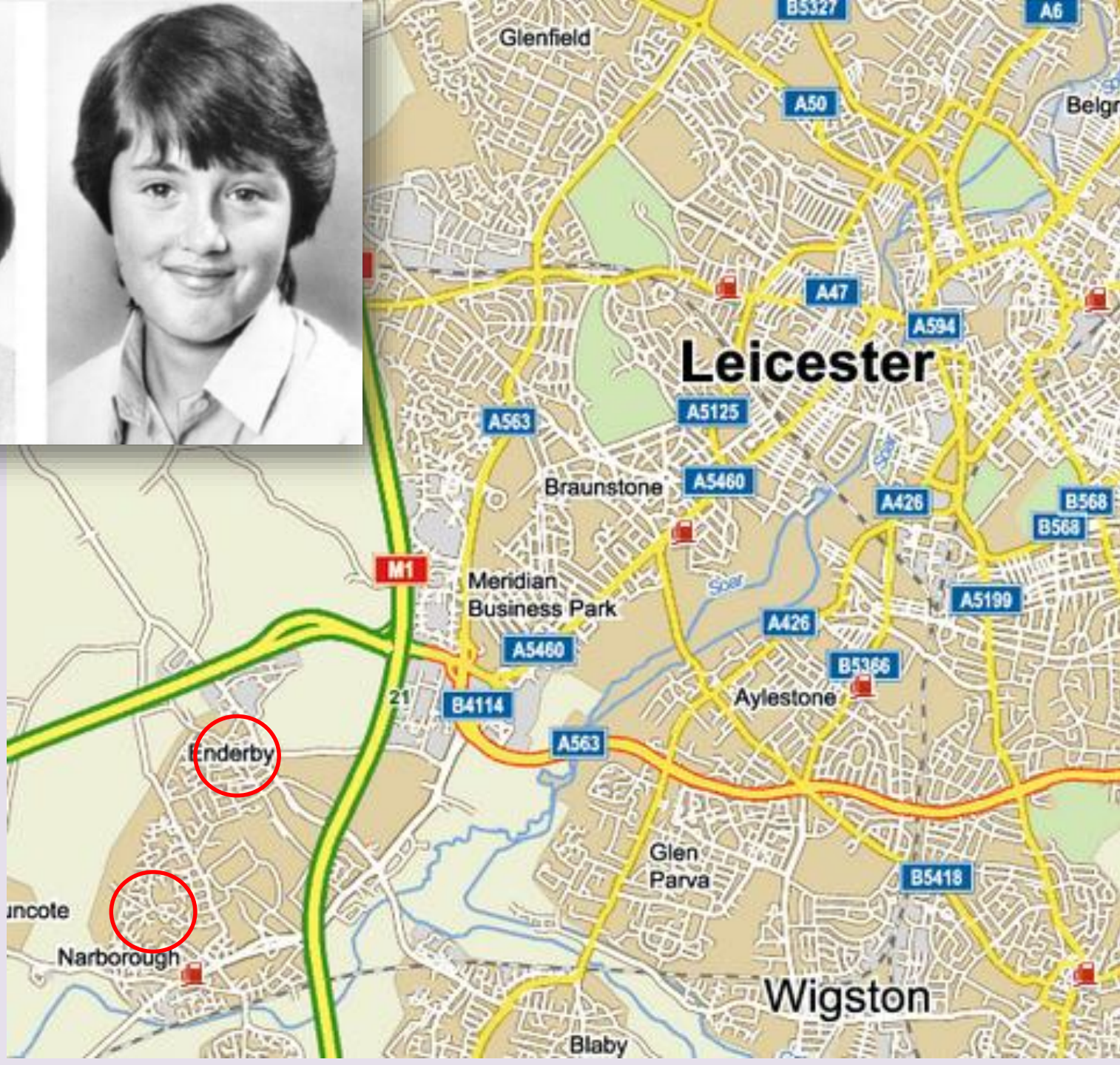
➡ SROVNÁVACÍ VZORKY

- DNA standardní kvality i kvantity
- různé názory na právo společnosti tyto vzorky získávat



3. KRIMINALISTICKÉ ANALÝZY

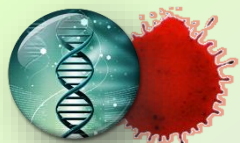






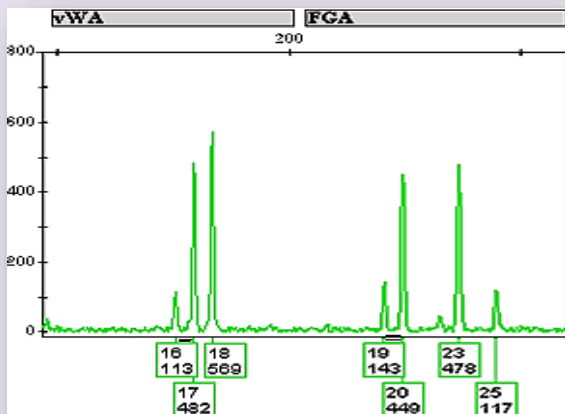
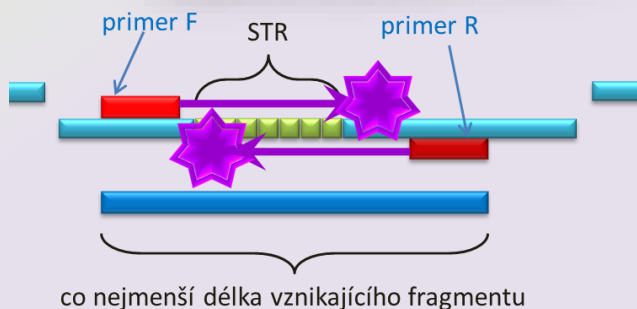
gettyimages
Mirrorpix



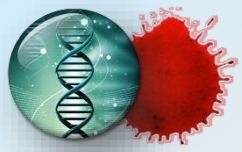


3. KRIMINALISTICKÉ ANALÝZY

► trendy

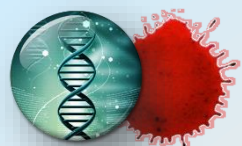


- **zrychlení a automatizace**
- cílem je identifikace do několika minut bez nutnosti být v laboratoři
- **zlepšení možností analýzy LCN (low-copy number = „kde nic tu nic“)**
- zkracování produktů
- zvyšování citlivosti
- **posun možností identifikace ze smíšených stop**
- mikrodisekční techniky
- digital PCR

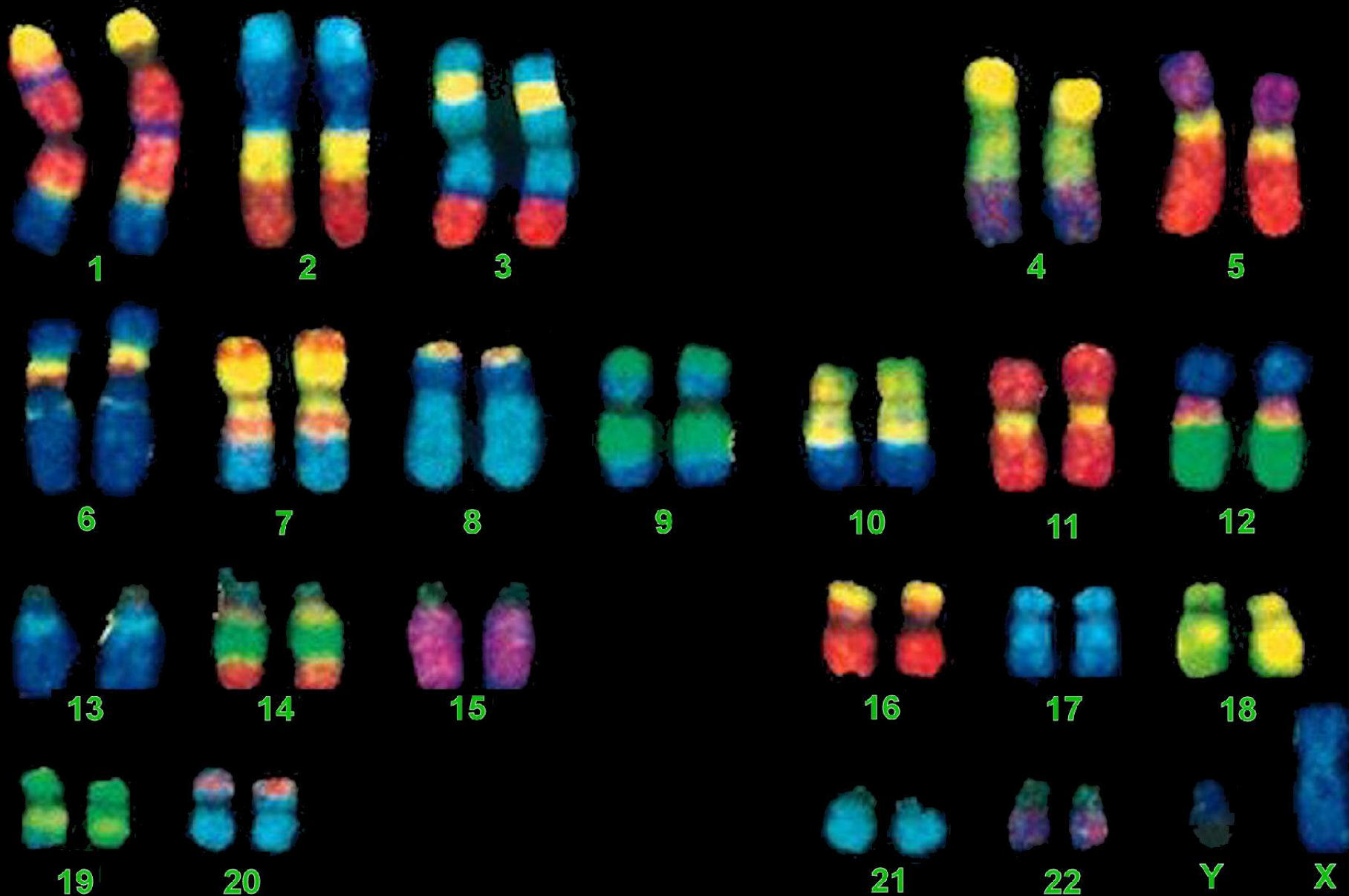


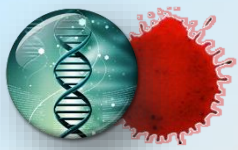
4. FORENZNÍ PHENOTYPING



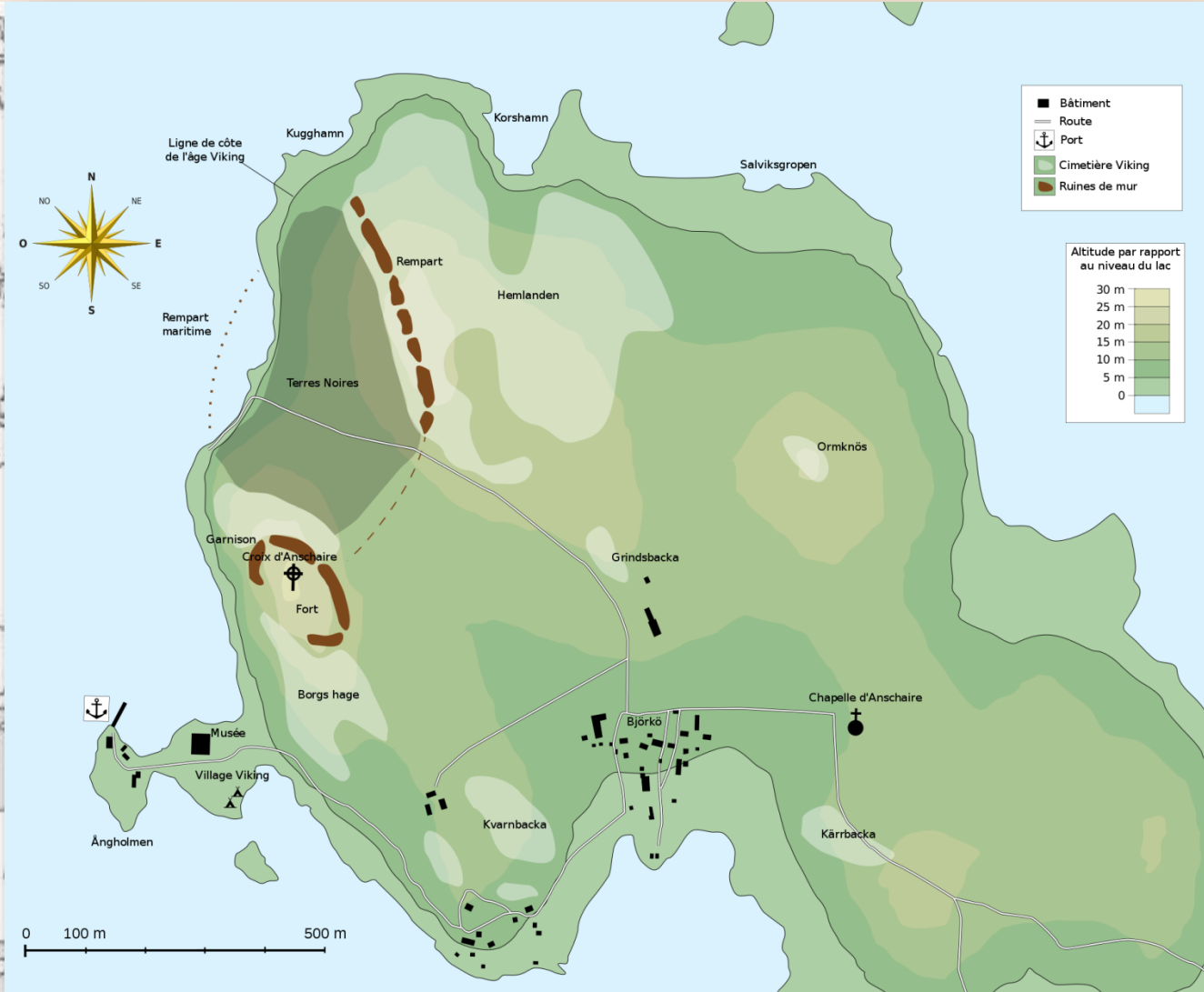


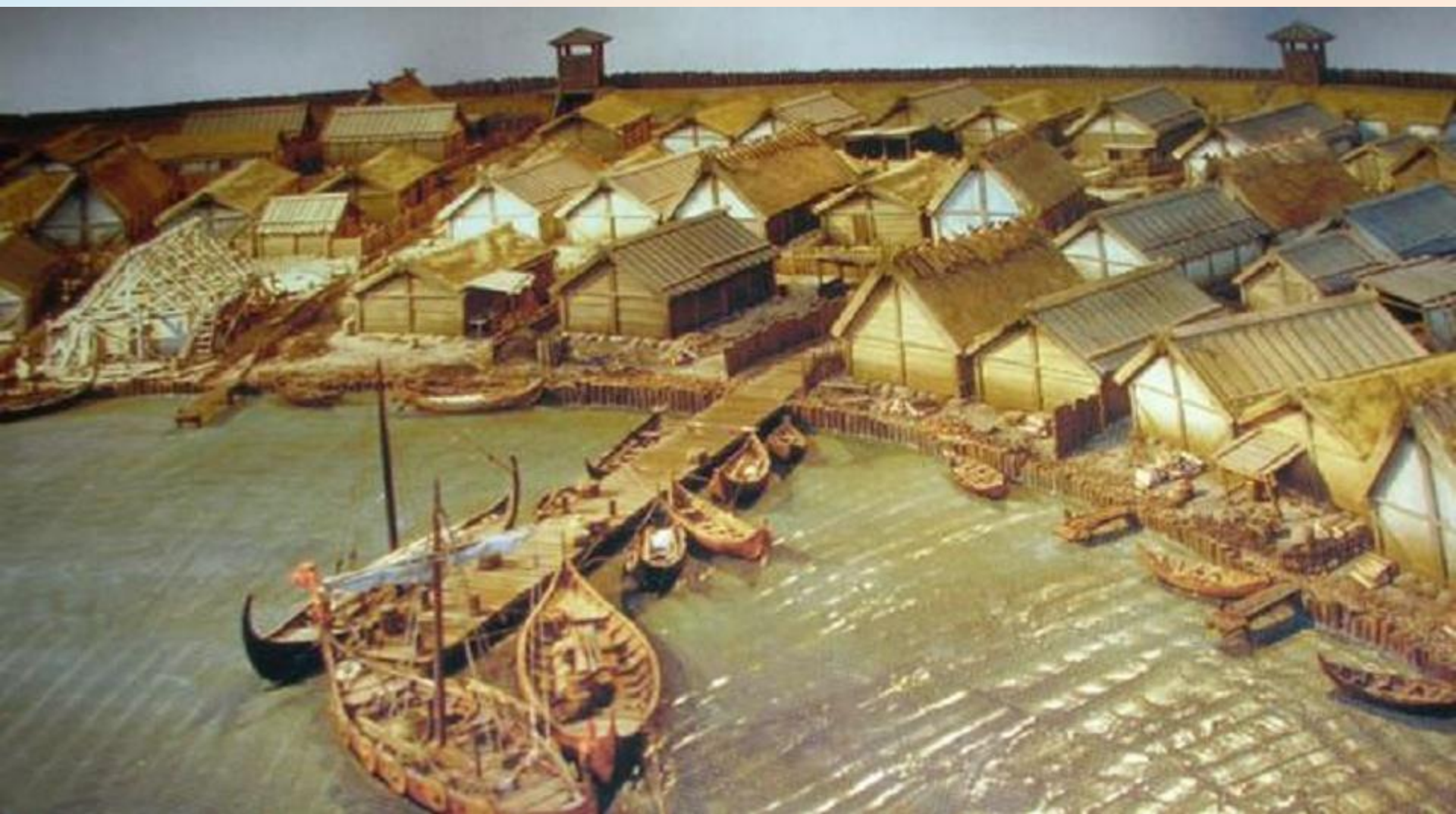
URČENÍ POHLAVÍ





VÁLEČNÍK Z BIRKY










Birka, hrob Bj581





BRIEF COMMUNICATION

A female Viking warrior confirmed by genomics

Charlotte Hedenstierna-Jonson , Anna Kjellström, Torun Zachrisson, Maja Krzewińska, Veronica Sobrado, Neil Price, Torsten Günther, Mattias Jakobsson, Anders Götherström, Jan Storå

First published: 8 September 2017 [Full publication history](#)

DOI: 10.1002/ajpa.23308 [View/save citation](#)

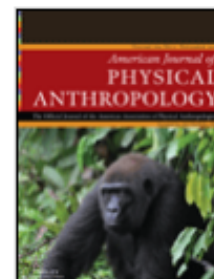
Cited by (CrossRef): 1 article  [Check for updates](#) |  [Citation tools](#) ▼

 score 2,604

[Funding Information](#)

Funding information Swedish Research Council (VR) & Riksbankens jubileumsfond (RJ).

Early View

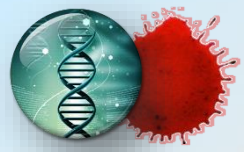


[Browse Early View Articles](#)
Online Version of Record
published before inclusion
in an issue

Abstract

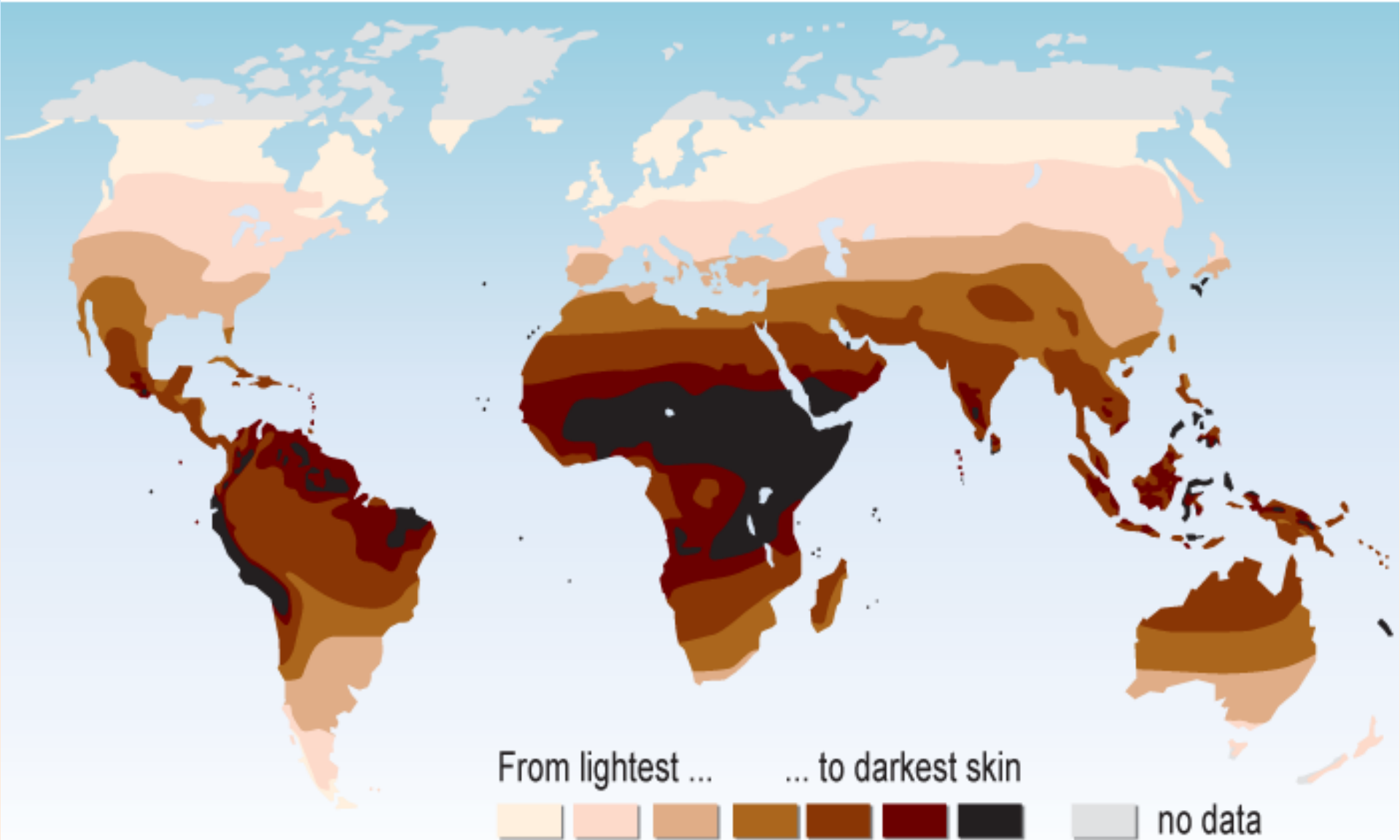
Objectives

The objective of this study has been to confirm the sex and the affinity of an individual buried in a well-furnished warrior grave (Bj 581) in the Viking Age town of Birka, Sweden. Previously, based on the material and historical records, the male sex has been associated with the gender of the warrior and such was the case with Bj 581. An earlier osteological classification of the individual as female was considered controversial in a historical and archaeological context. A genomic confirmation of the biological sex of the individual was considered necessary to solve the issue.

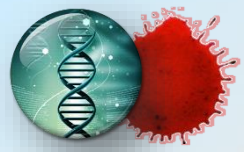


PIGMENTAČNÍ VARIABILITA



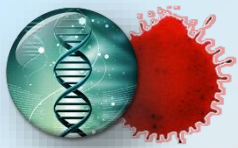


Source: Chaplin G.© , *Geographic Distribution of Environmental Factors Influencing Human Skin Coloration*, *American Journal of Physical Anthropology* 125:292–302, 2004; map updated in 2007.

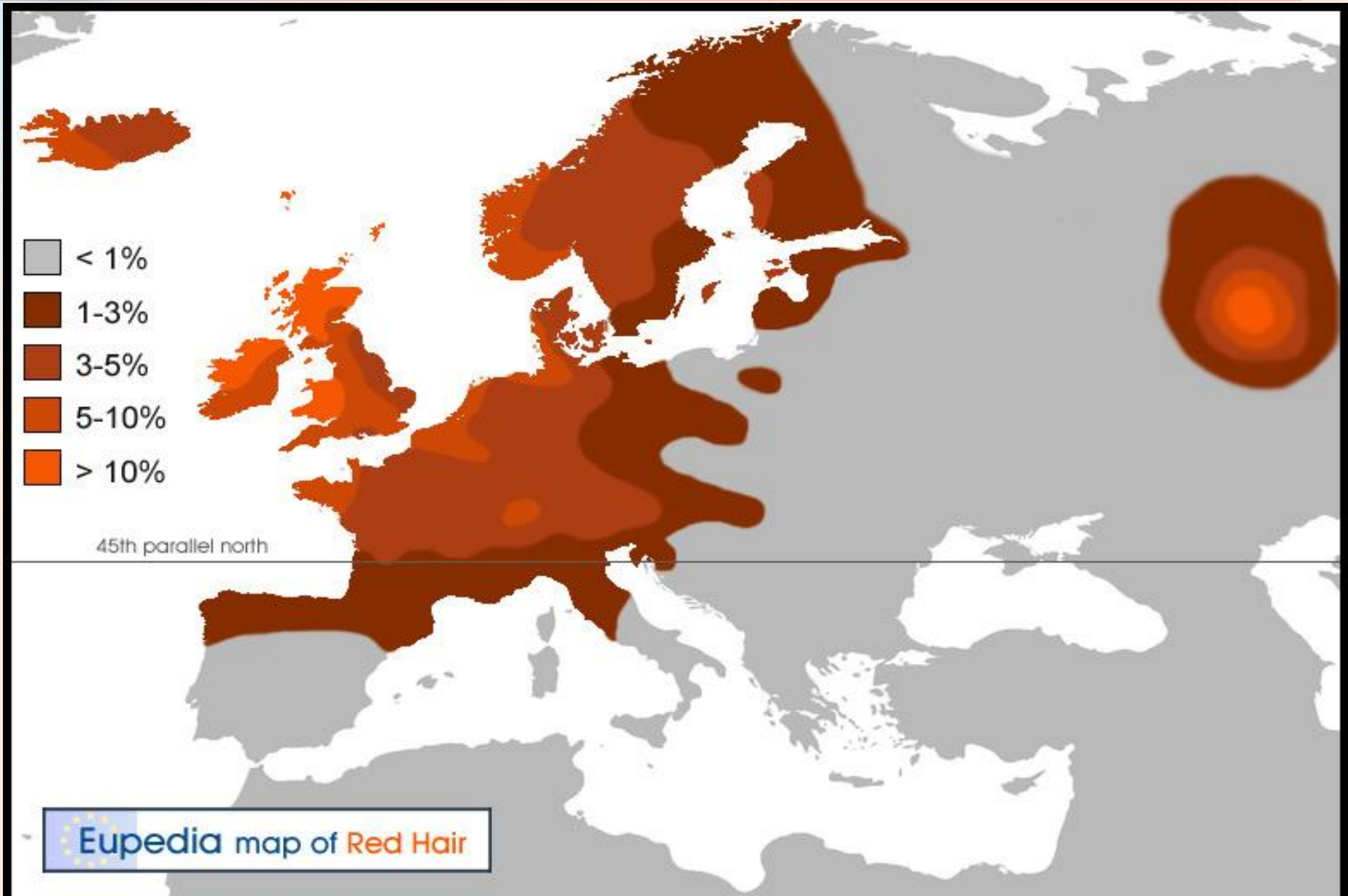


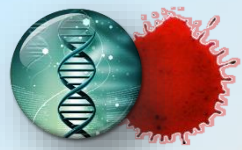
RUSOVLASOST





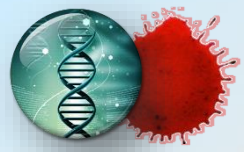
RUSOVLASOST





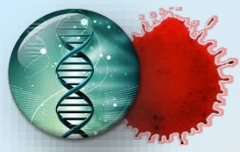
RUSOVLASOST





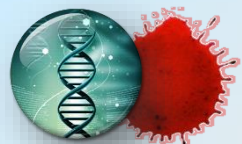
ALBINISMUS





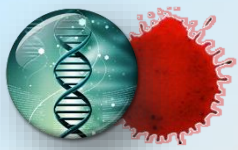
VITILIGO





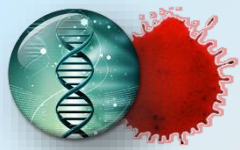
BARVA DUHOVKY





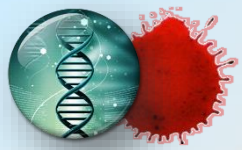
MODROOKOST





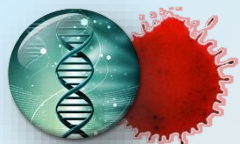
HETEROCHROMIE





HETEROCHROMIE



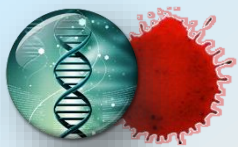


4. FORENZNÍ PHENOTYPING

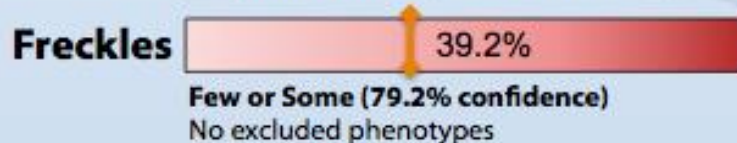
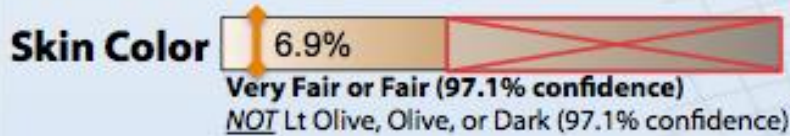
- ▶ určení vzhledových charakteristik osoby, která zanechala biologickou stopu
- ▶ analýza SNP, které jsou ve vazbě s geny ovlivňujícími daný fenotyp



Hair	Dark	Light	
	0.033	0.967	
Black	Brown	Red	Blond
0.007	0.077	0.816	0.1
Eye	Blue	Int.	Brown
	0.965	0.027	0.007

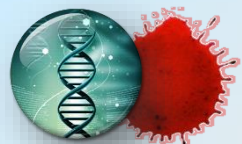


Predicted (▣) & Excluded (⊠) Phenotypes



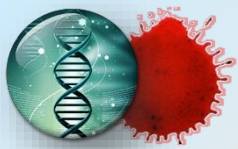
Sex: Female
Ancestry: 100% European
 (Northwest – British Isles)



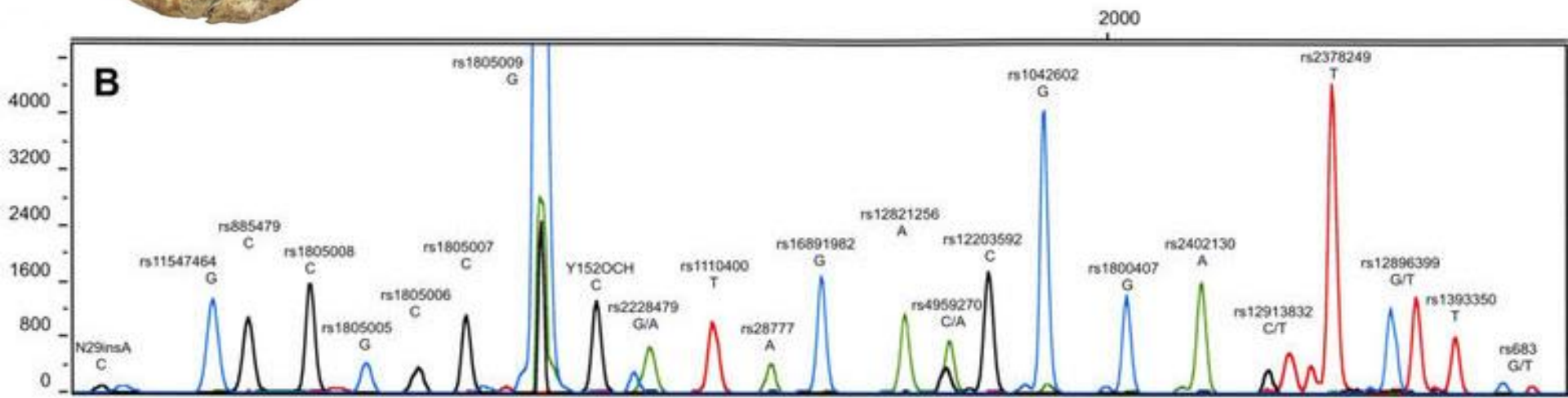


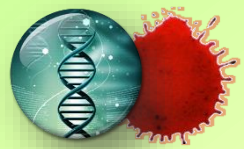
ŽENA Z TYNIECKÉHO KLÁŠTERA



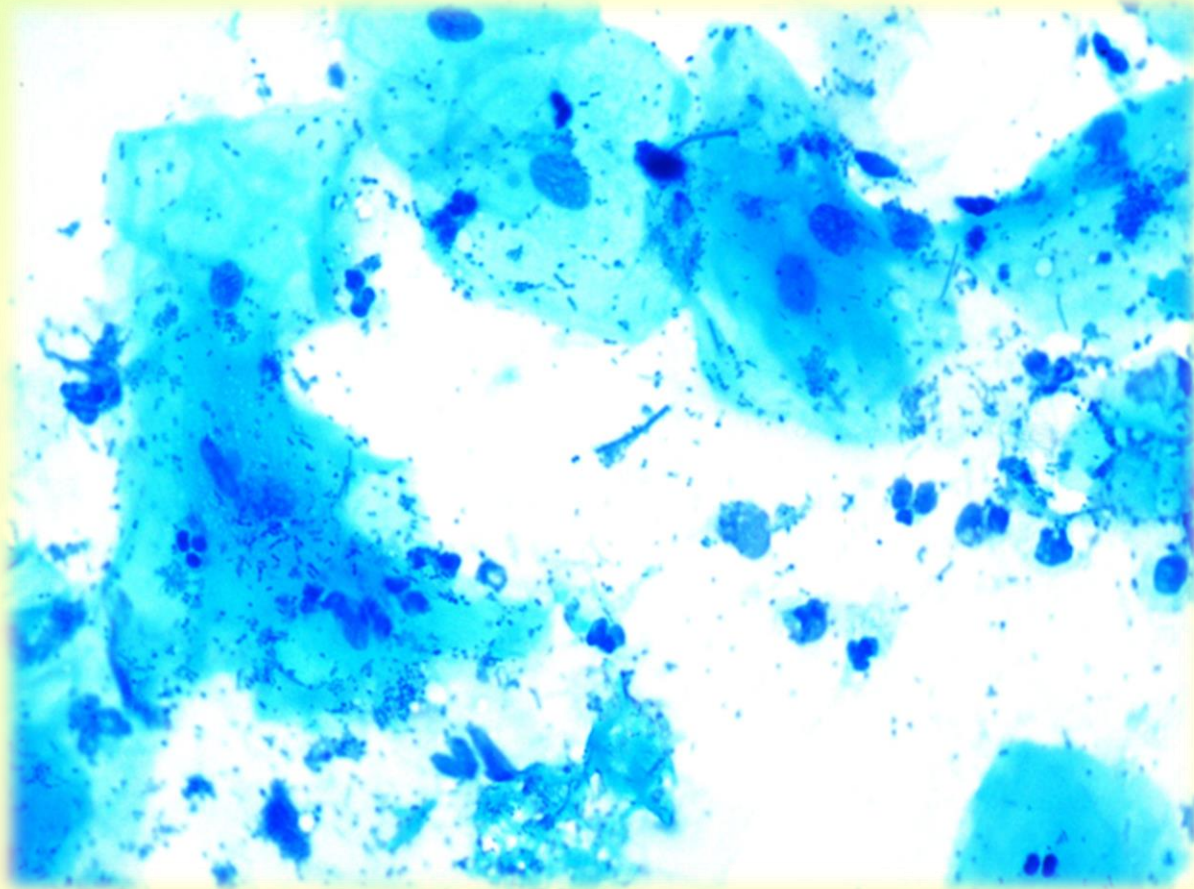


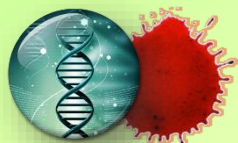
PHENOTYPING





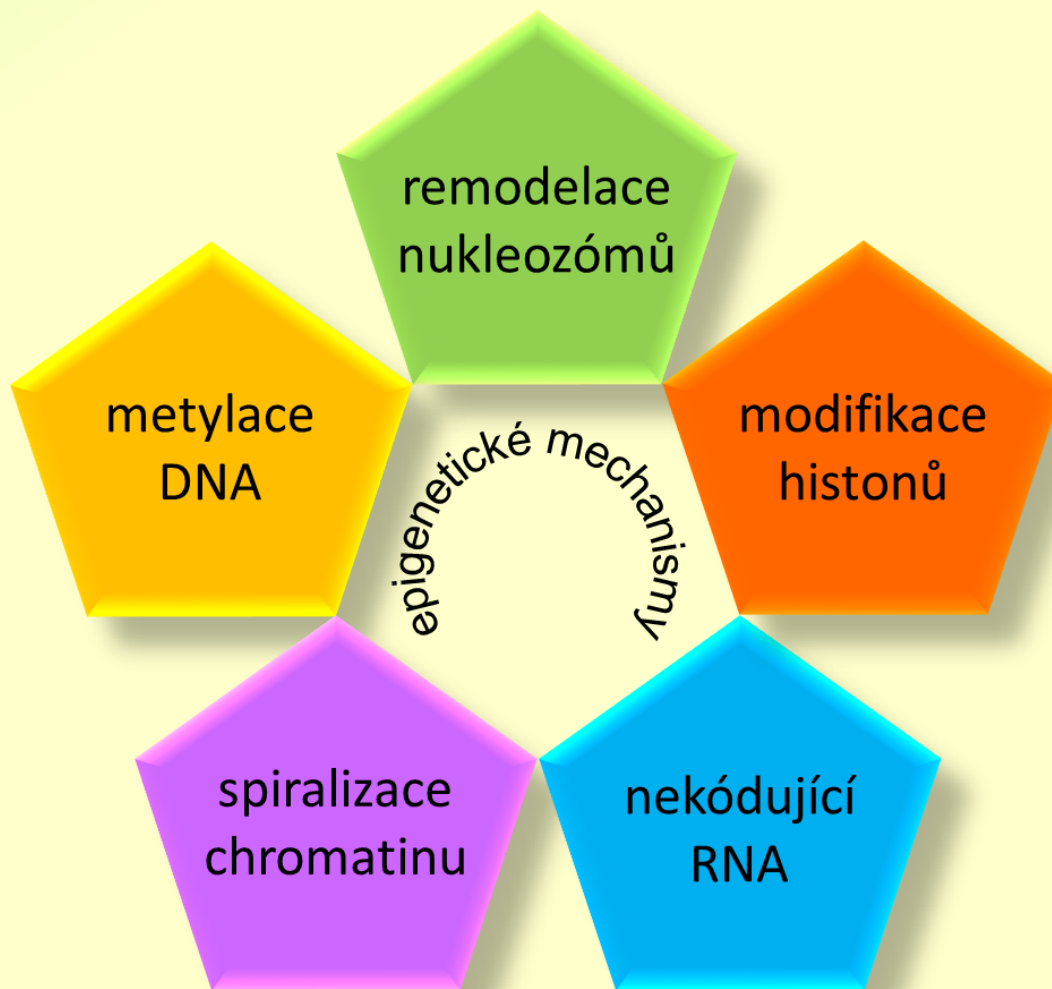
5. FORENZNÍ EPIGENETIKA

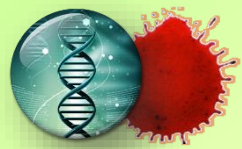




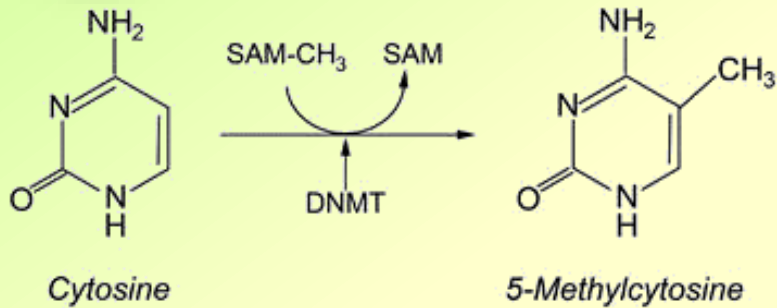
5. FORENZNÍ EPIGENETIKA

- ➡ epigenetika studuje reverzibilní změny funkce genů (tj. mění se fenotyp, ale nemění se genotyp)

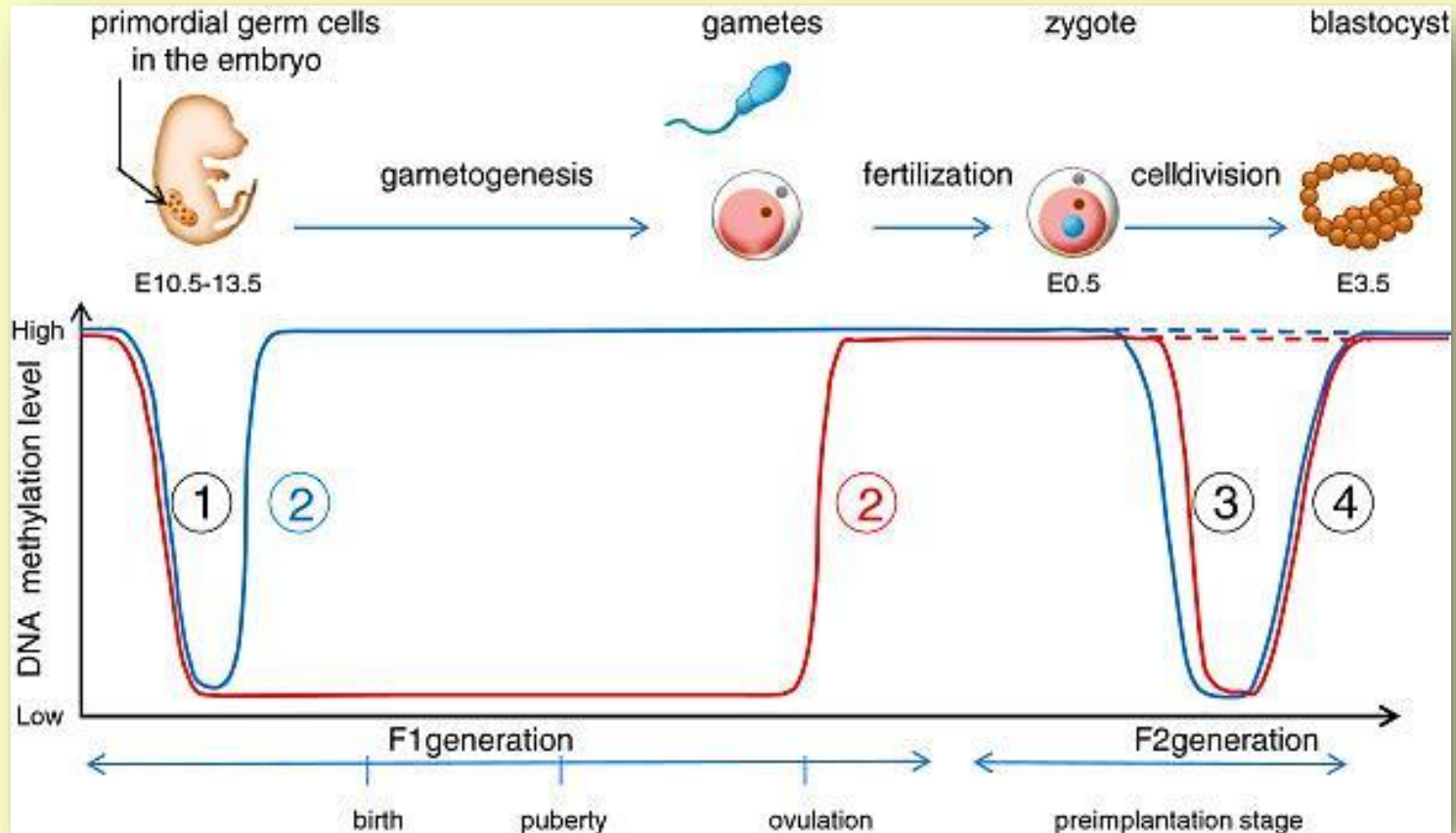


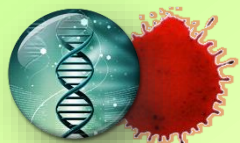


metylace DNA



- ➡ metylace = inaktivace genu
- ➡ základní mechanismus regulace exprese genu

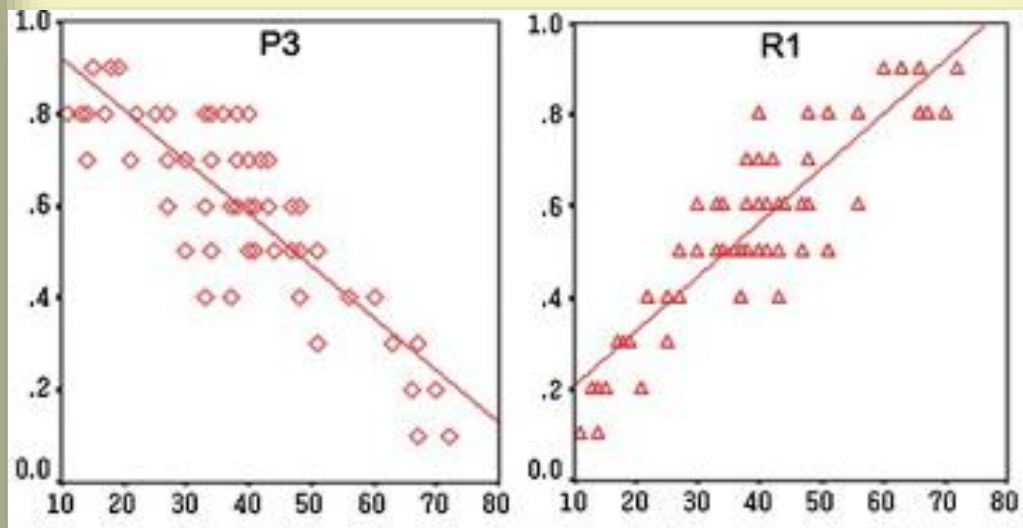


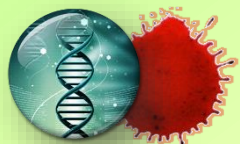


využití analýzy metylace DNA



► **určení stáří osoby**
(míra metylace určitých genů roste nebo naopak klesá s věkem)



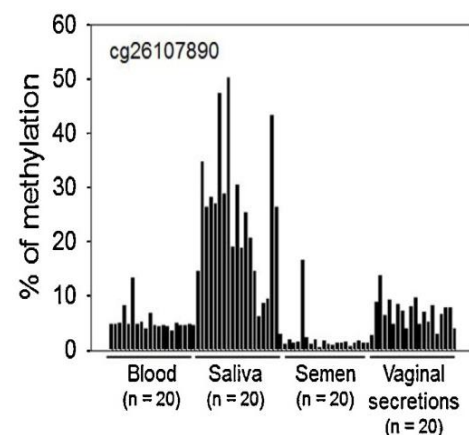
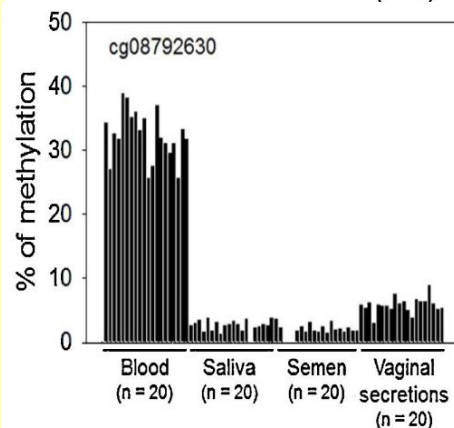
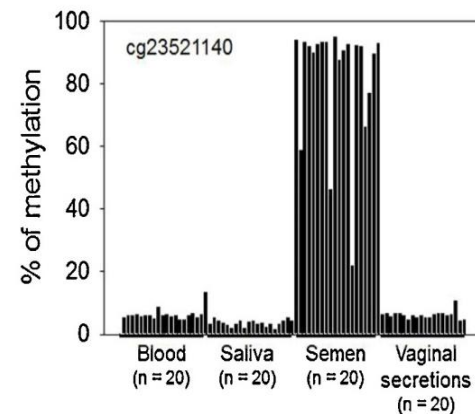
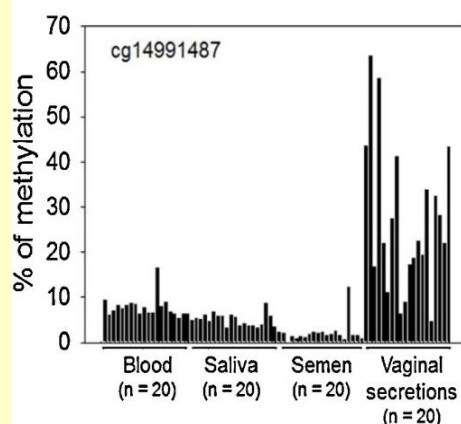


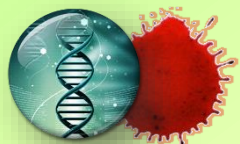
využití analýzy metylace DNA



určení typu biologického materiálu

(v každém typu tkáně exprimovány jiné geny → rozdíly v metylaci)

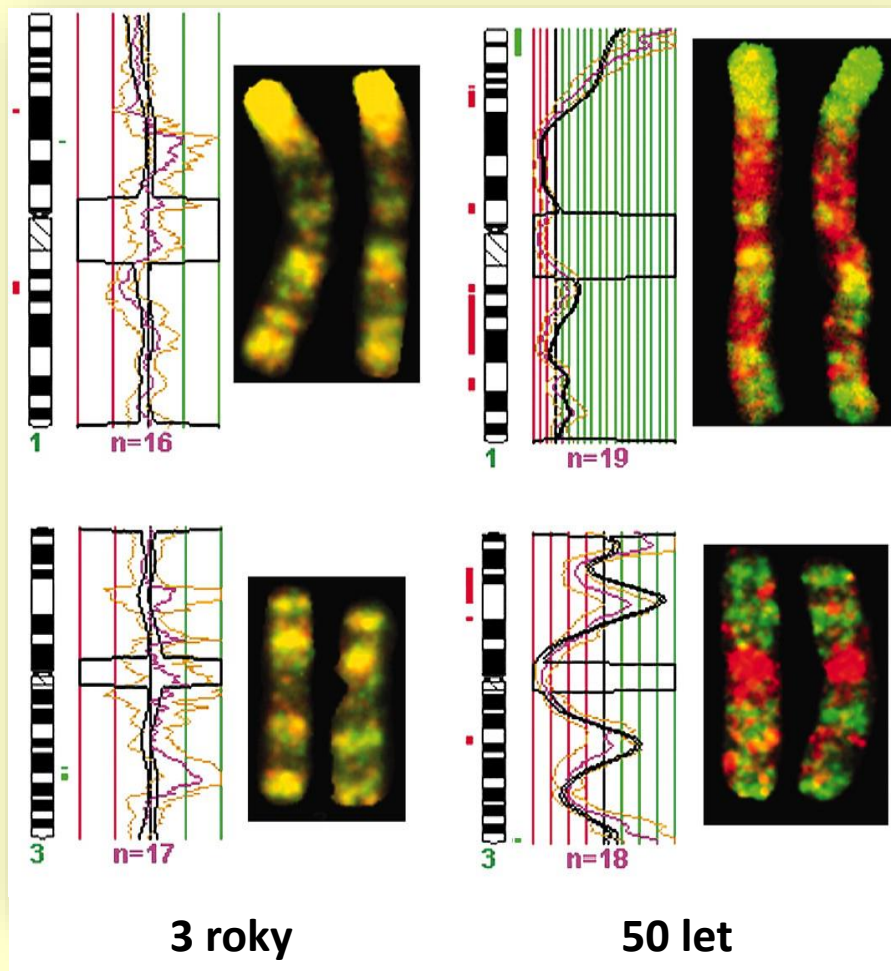


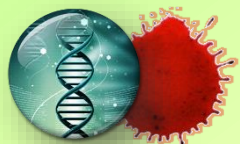


využití analýzy metylace DNA



➡ rozlišení jednovaječných dvojčat

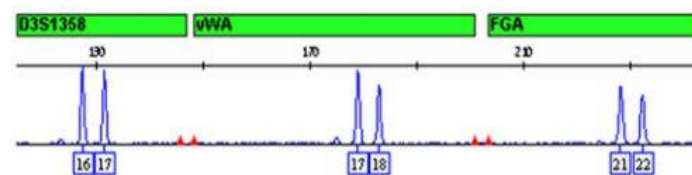
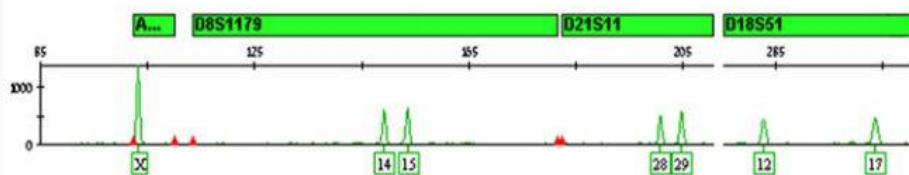




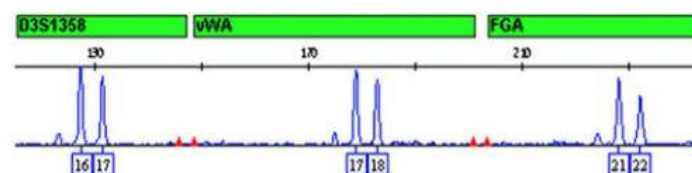
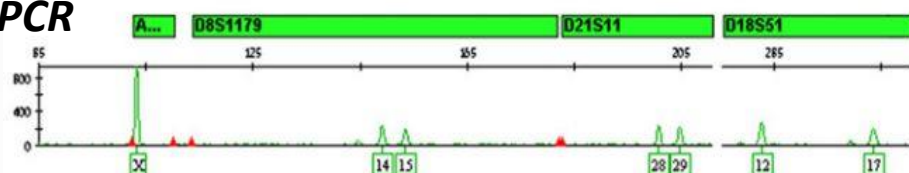
využití analýzy metylace DNA

➔ autentizace vzorku
(*in vivo* x *in vitro* DNA)

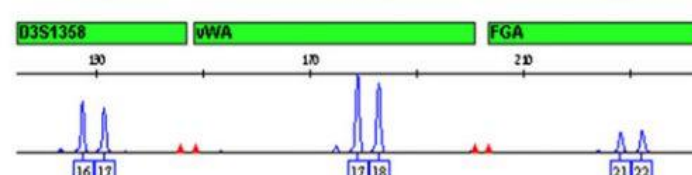
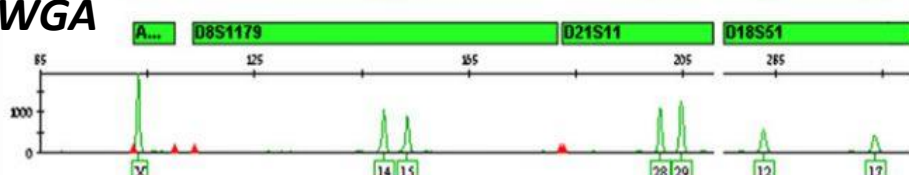
in vivo



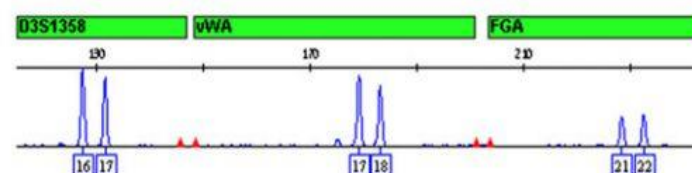
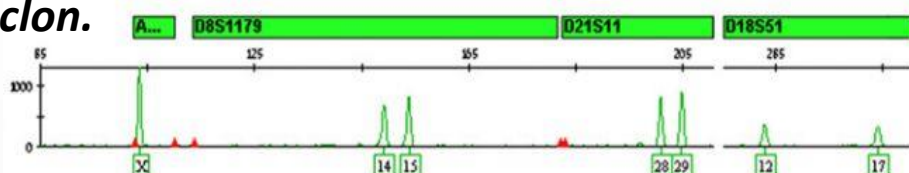
in vitro PCR



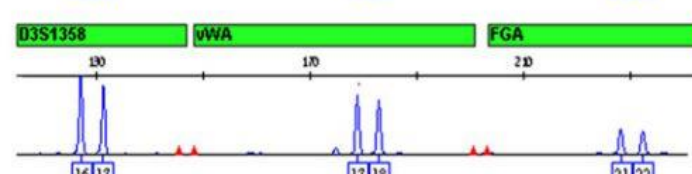
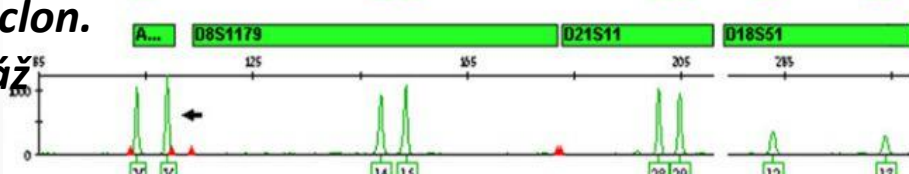
in vitro WGA

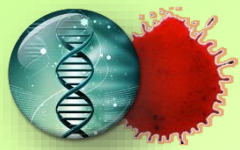


in vitro clon.



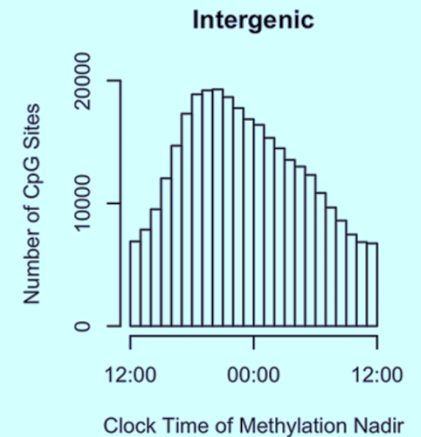
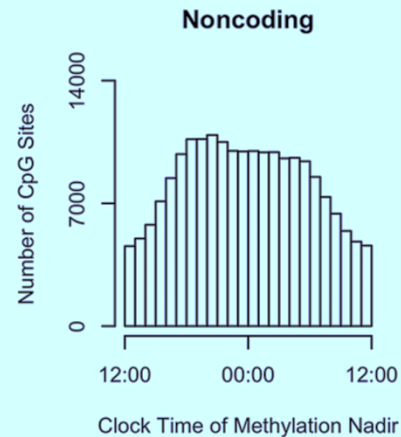
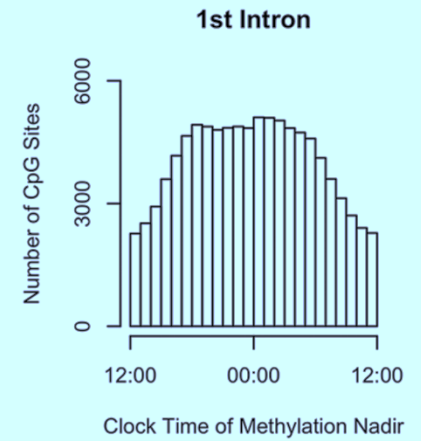
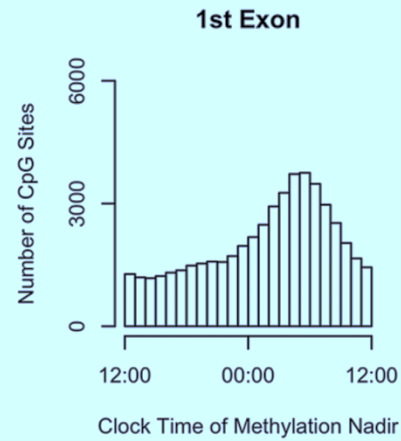
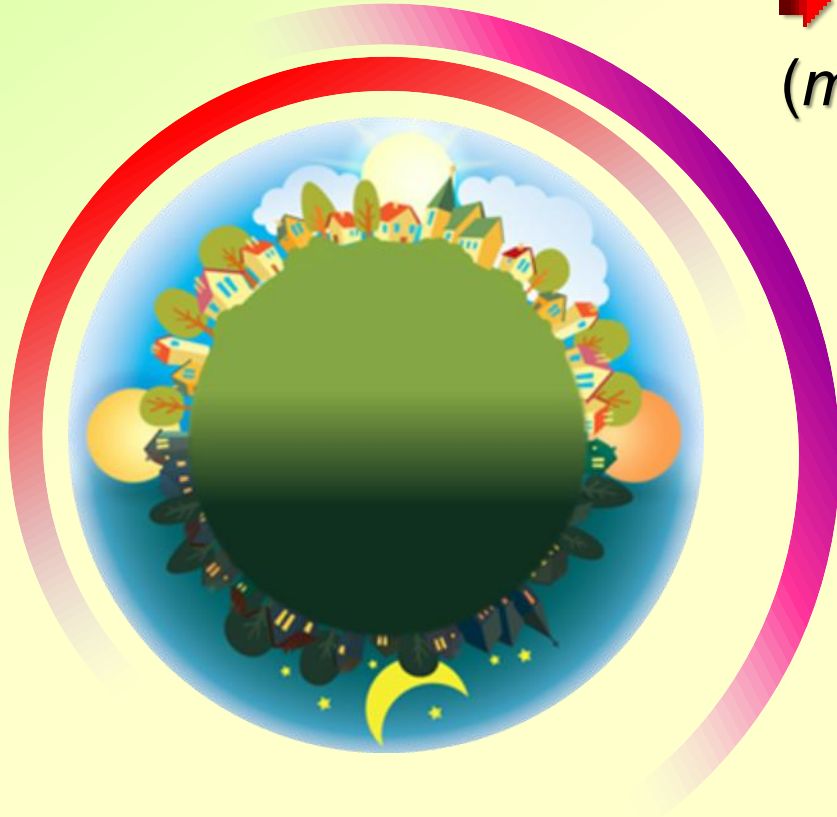
in vitro clon.
+ montáž

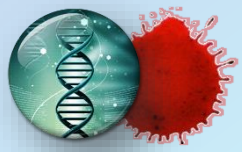




využití analýzy metylace DNA

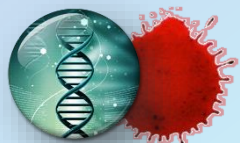
určení denní doby smrti
(methylace cirkadiálních genů)





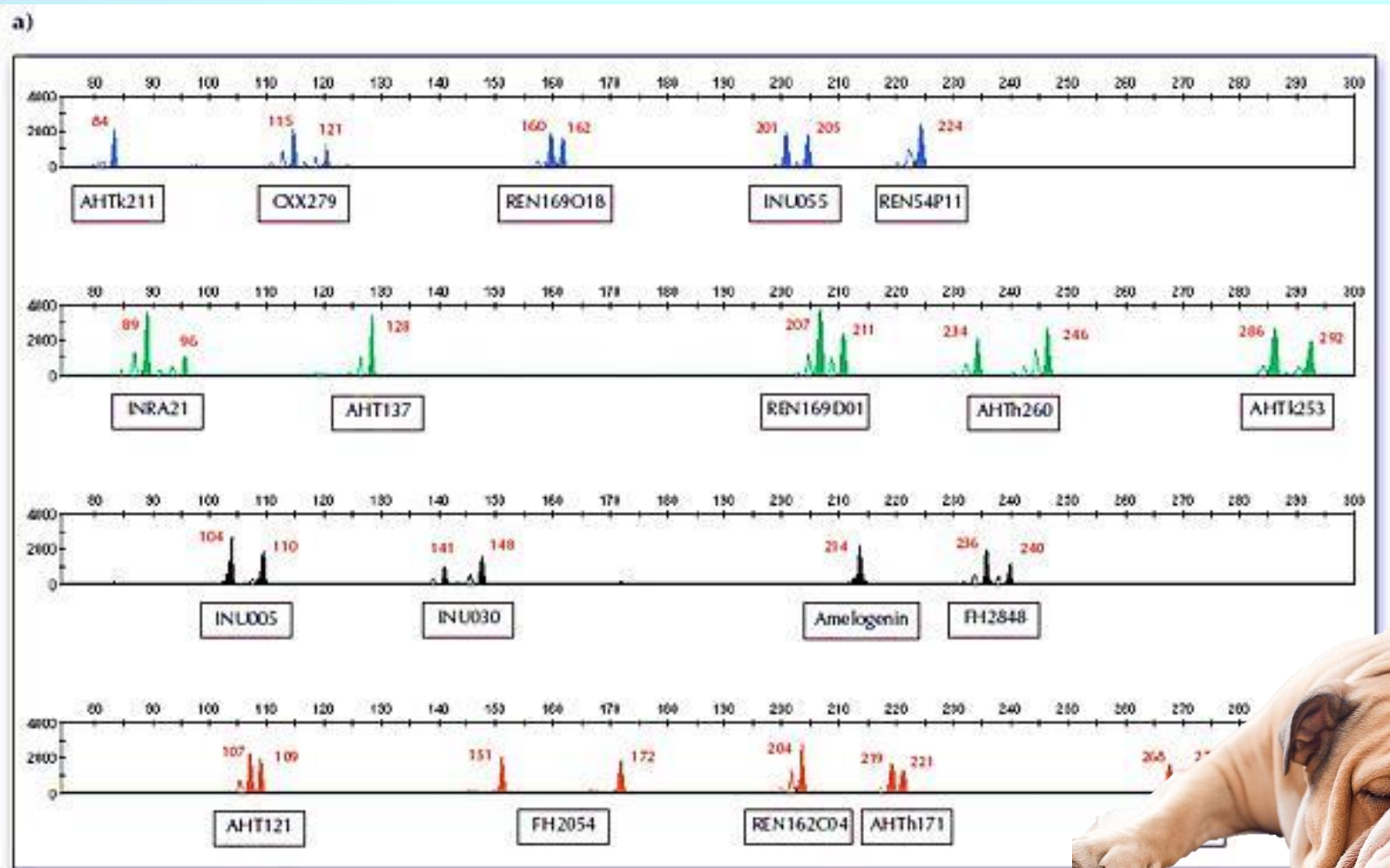
6. „NON-HUMAN“ ANALÝZY

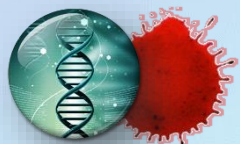




6. „NON-HUMAN“ ANALÝZY

- ➡ stejně jako lze identifikovat lidské jedince, lze identifikovat i zvířata a rostlinné genety *via* STR

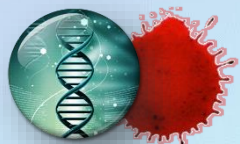




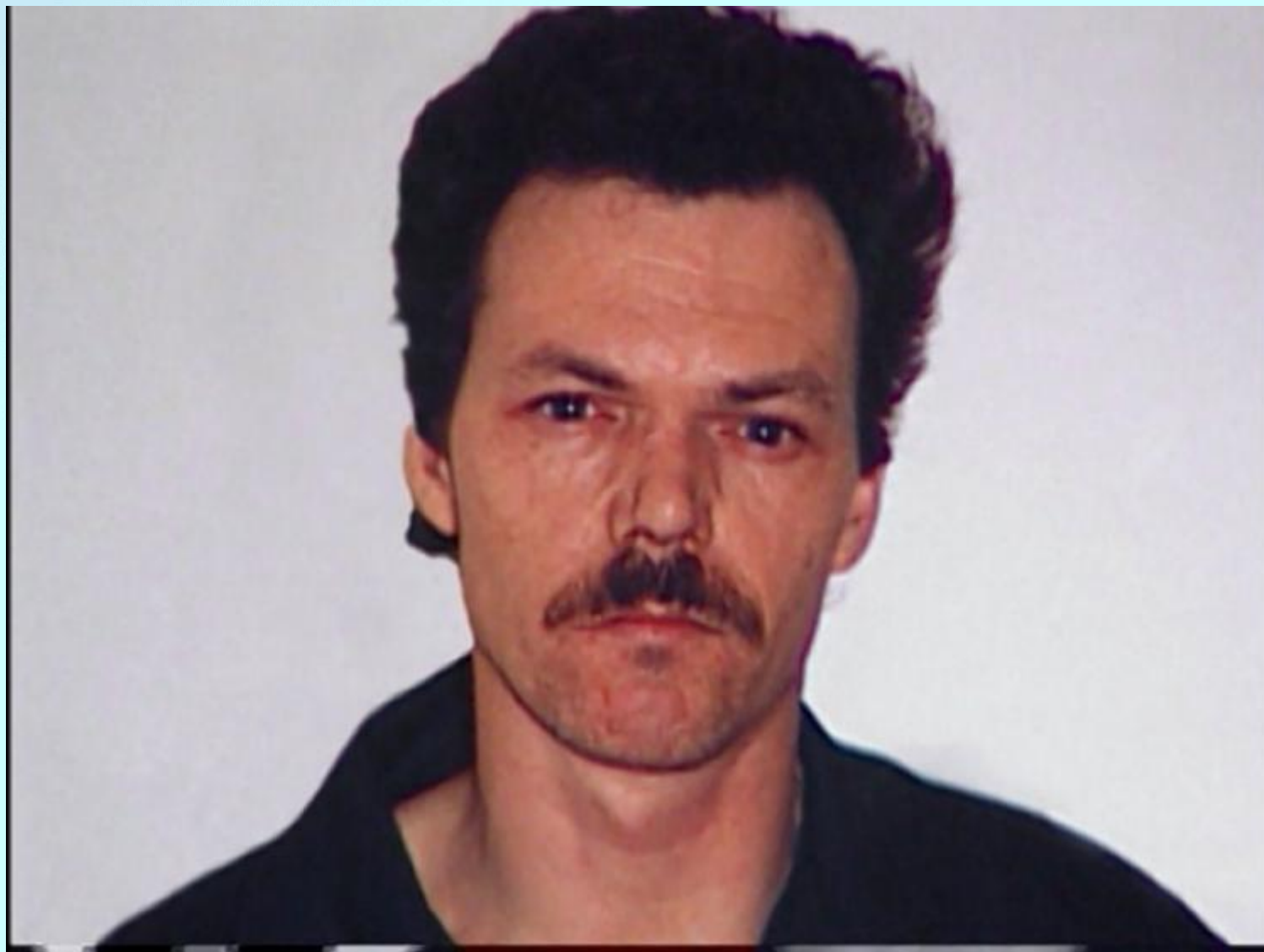
6. KAUZA „SNOWBALL“

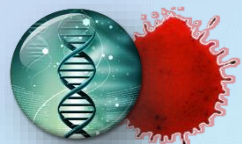




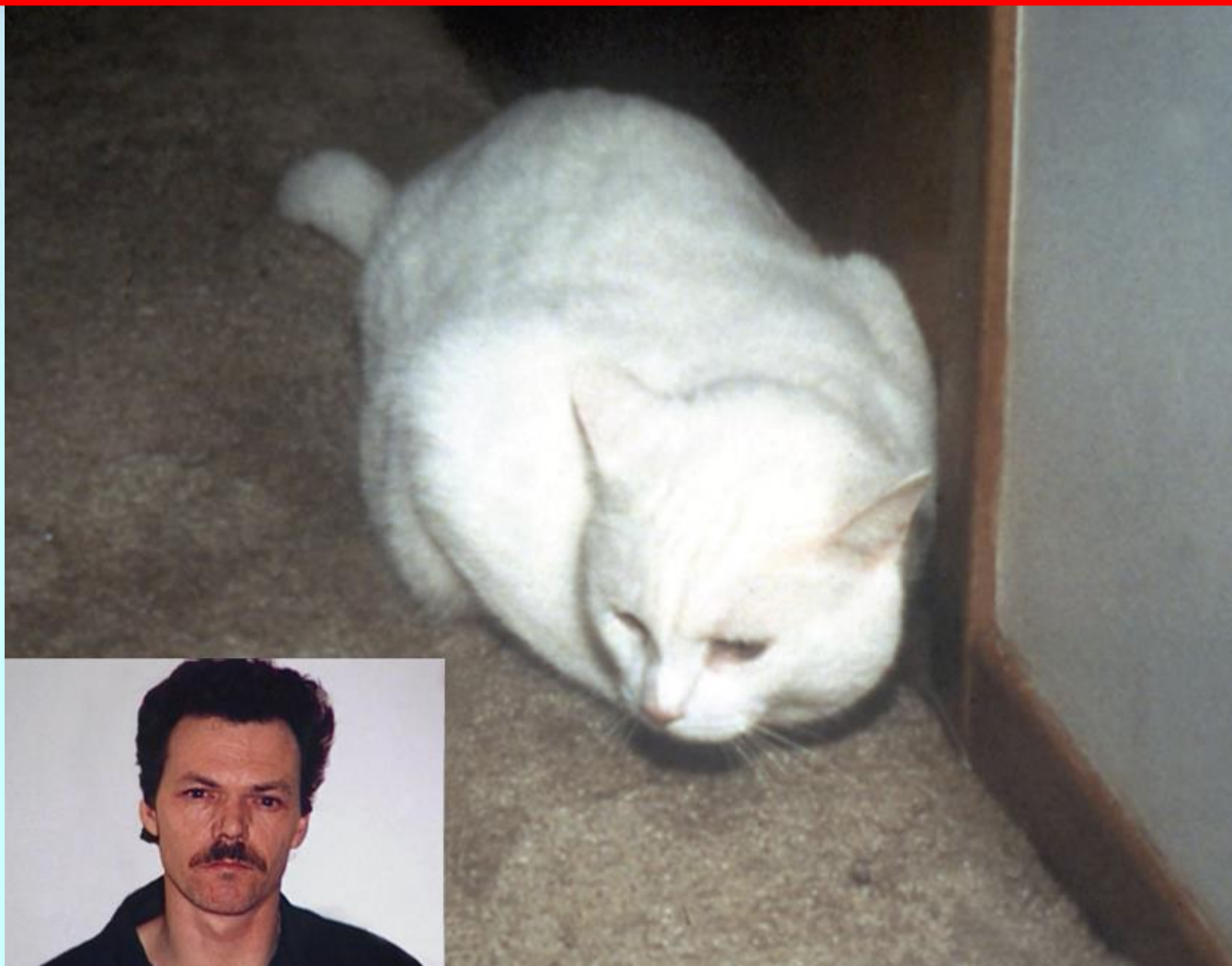


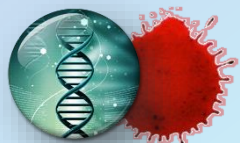
6. KAUZA „SNOWBALL“





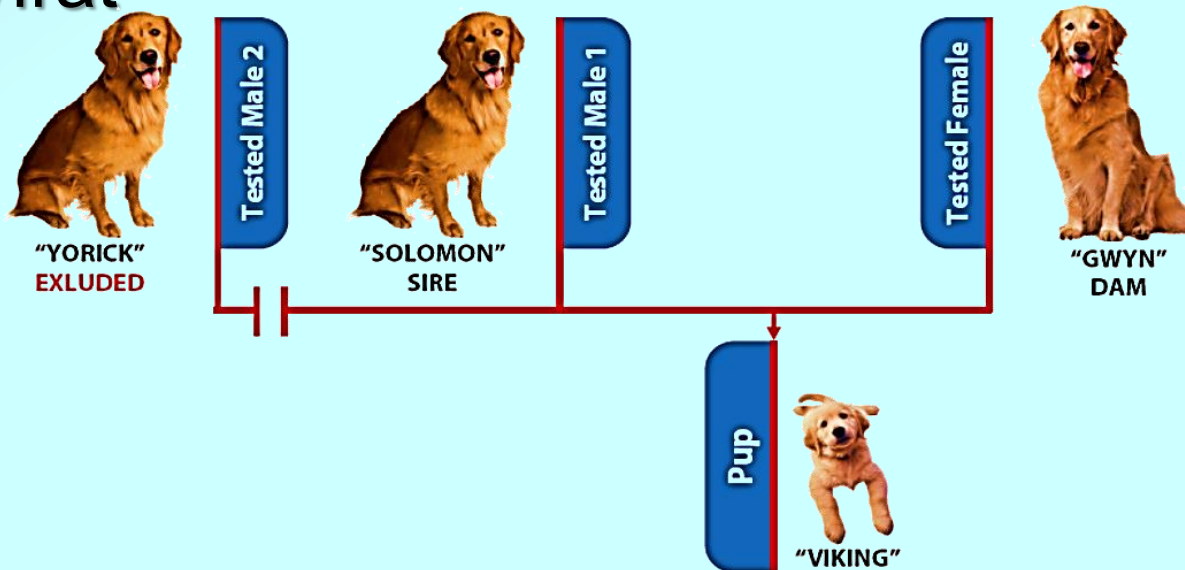
6. KAUZA „SNOWBALL“





6. „NON-HUMAN“ ANALÝZY

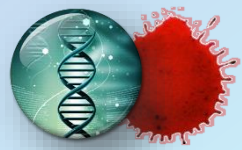
- stejně jako lze analyzovat příbuzenské vztahy u lidí, lze to i u zvířat



REGISTERED NAME	PEZ1	PEZ3	PEZ5	PEZ6	PEZ8	PEZ12	PEZ20	FHC2010	FHC2054	FHC2079	PARENTAGE
SOLOMON	AE	EF	AB	EE	BH	DI	DE	BD	EF	AB	PATERNITY INCLUDED
VIKING	CE	FJ	AA	EE	EH	IK	BE	BD	EF	AB	
GWYN	CD	GJ	AD	DE	EE	FK	BD	BB	EE	BC	MATERNITY INCLUDED

REGISTERED NAME	PEZ1	PEZ3	PEZ5	PEZ6	PEZ8	PEZ12	PEZ20	FHC2010	FHC2054	FHC2079	PARENTAGE
YORICK	BD	FJ	AC	BD	BC	DI	BD	AD	BF	BD	PATERNITY EXCLUDED
VIKING	CE	FJ	AA	EE	EH	IK	BE	BD	EF	AB	
GWYN	CD	GJ	AD	DE	EE	FK	BD	BB	EE	BC	MATERNITY INCLUDED

- Exclusion
- Letters in blue indicate obligate paternal type
- Letters in red indicate obligate maternal type

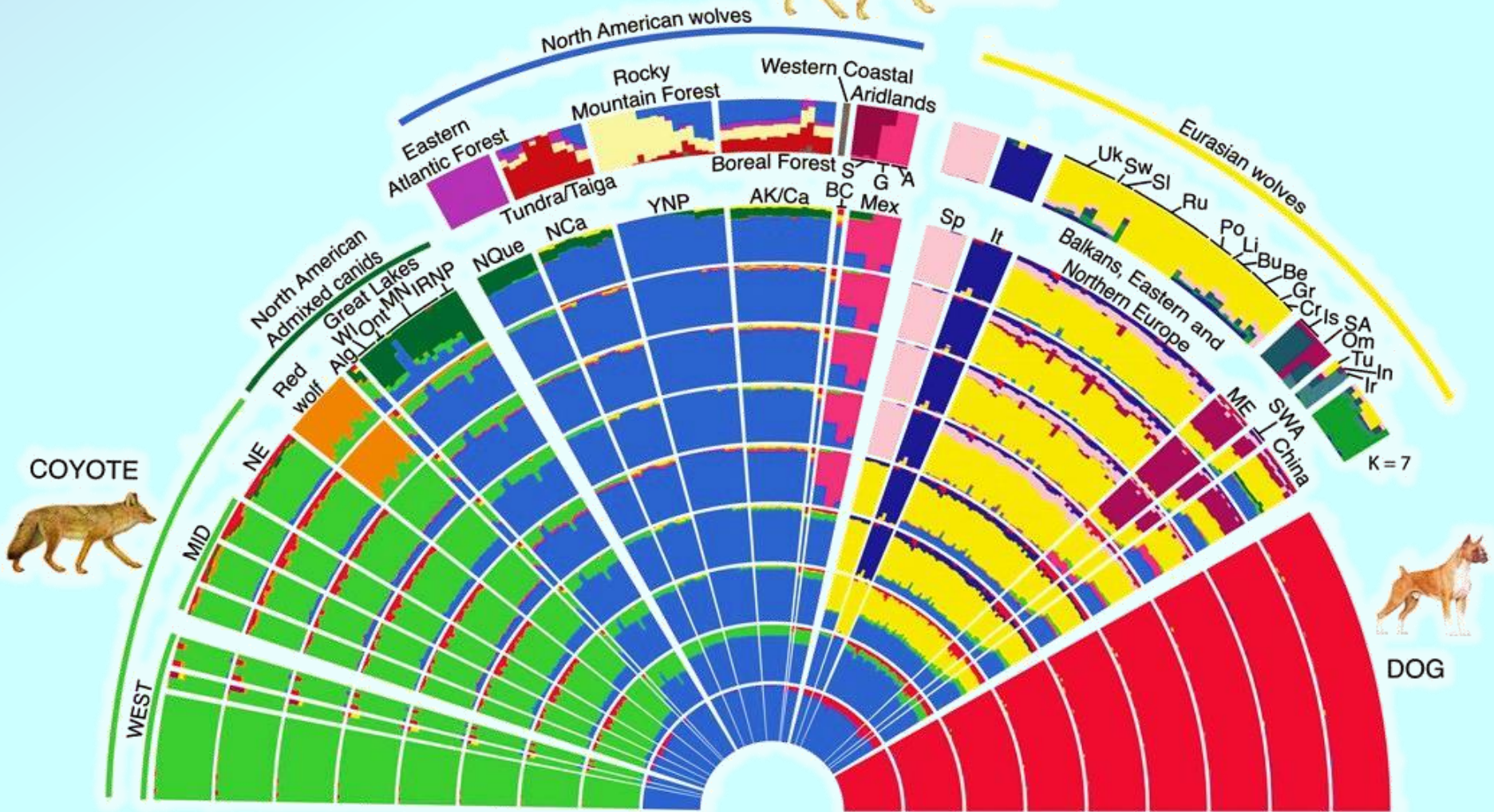


6. „NON-HUMAN“ ANALÝZY

- ➡ identifikace plemen, *subspecies* a příbuzných druhů *via* SNP

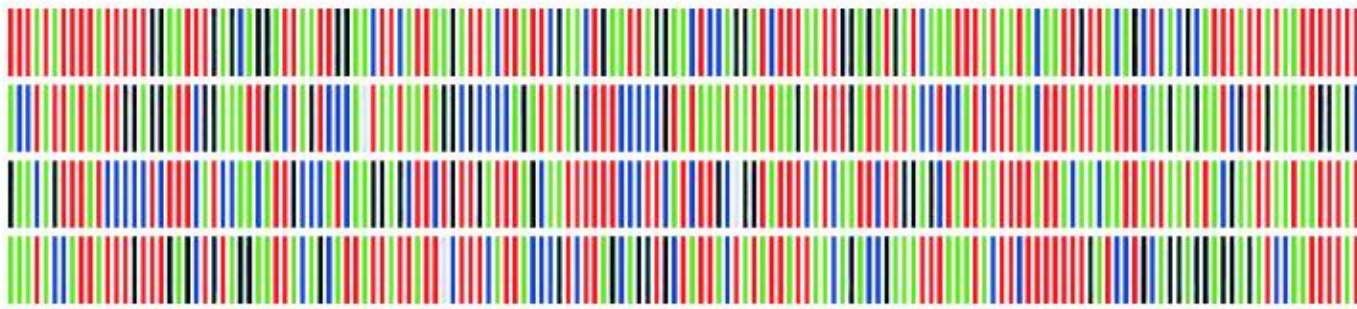


WOLVES

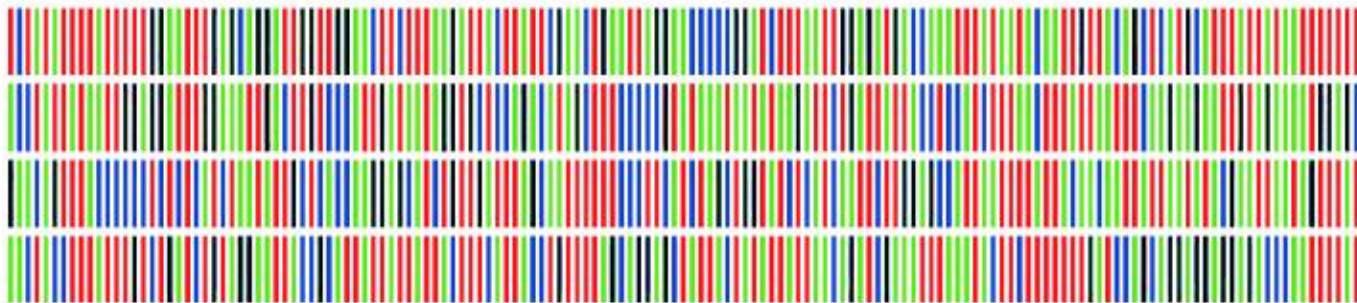




Astraptes fulgerator CELT



Astraptes fulgerator TRIGO

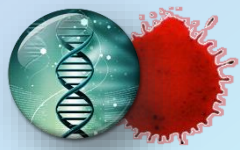


Bubo virginianus



Tyto alba

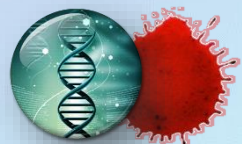




6. „NON-HUMAN“ ANALÝZY

- ➔ identifikace mikroorganismů *via* NG sekvenování 16S rRNA

DQ513019	277995	ACA	GC	AAG	-	CGAG	CGG	AAA	CGA	AGA	---	GG	GC	TGC	ACC	C	---	GGCG	CGA	GCGGGCG	
AB180385	161913	ACA	GC	AAG	-	CGAG	CGG	CAG	CGA	CAACA	---	GA	ACC	T	CGGGGG	A	T	G	GGCGG	CGA	GCGGGCG
DQ334351	273467	---	GC	AAG	-	CTAG	CGG	AACA	TT	---	---	CTAG	CT	GCT	AGAA	---	---	GA	T	ACGAGCGGGCG	
AF539776	105592	ACA	GC	AAG	-	CGAG	CGG	AACA	TT	---	---	CTAG	CT	GCT	AGAA	---	---	GA	T	GACGAGCGGGCG	
AF172988	24086	ACA	GC	AAG	-	CGAG	CGG	AACA	TT	---	---	CTAG	CT	GCT	AGAA	---	---	GA	T	GACGAGCGGGCG	
AY007201	92372	ACA	GC	AAG	-	CGAG	CGG	AACA	TT	---	---	CTAG	CT	GCT	AGAA	---	---	GA	T	GACGAGCGGGCG	
AY345481	119323	ACA	GC	AAG	-	CGAG	CGG	AACA	TT	---	---	CTAG	CT	GCT	AGAA	---	---	GA	T	GACGAGCGGGCG	
AY082666	77999	ACA	GC	AAG	-	CGAG	CGG	AACA	TT	---	---	CTAG	CT	GCT	AGAA	---	---	GA	T	GACGAGCGGGCG	
AY082667	78000	ACA	GC	AAG	-	CGAG	CGG	AACA	TT	---	---	CTAG	CT	GCT	AGAA	---	---	GA	T	GACGAGCGGGCG	
AM111082	161924	ACA	GC	AAG	-	CGAG	CGG	AAA	CGA	AGA	---	GG	GC	TGC	ACC	C	---	GGCG	CGA	GCGGGCG	
AM111083	161925	ACA	GC	AAG	-	CGAG	CGG	AAA	CGA	AGA	---	GG	GC	TGC	ACC	C	---	GGCG	CGA	GCGGGCG	
AM111084	161926	ACA	GC	AAG	-	CGAG	CGG	AAA	CGA	AGA	---	GG	GC	TGC	ACC	C	---	GGCG	CGA	GCGGGCG	
AM111085	161927	ACA	GC	AAG	-	CGAG	CGG	AAA	CGA	AGA	---	GG	GC	TGC	ACC	C	---	GGCG	CGA	GCGGGCG	
AM111086	161928	ACA	GC	AAG	-	CGAG	CGG	AAA	CGA	AG	---	AGG	AGC	TGC	CC	---	---	GGCG	CGA	GCGGGCG	
AM111088	161929	ACA	GC	AAG	-	CGAG	CGG	AAA	CGA	AGA	---	GG	GC	TGC	ACC	C	---	GGCG	CGA	GCGGGCG	

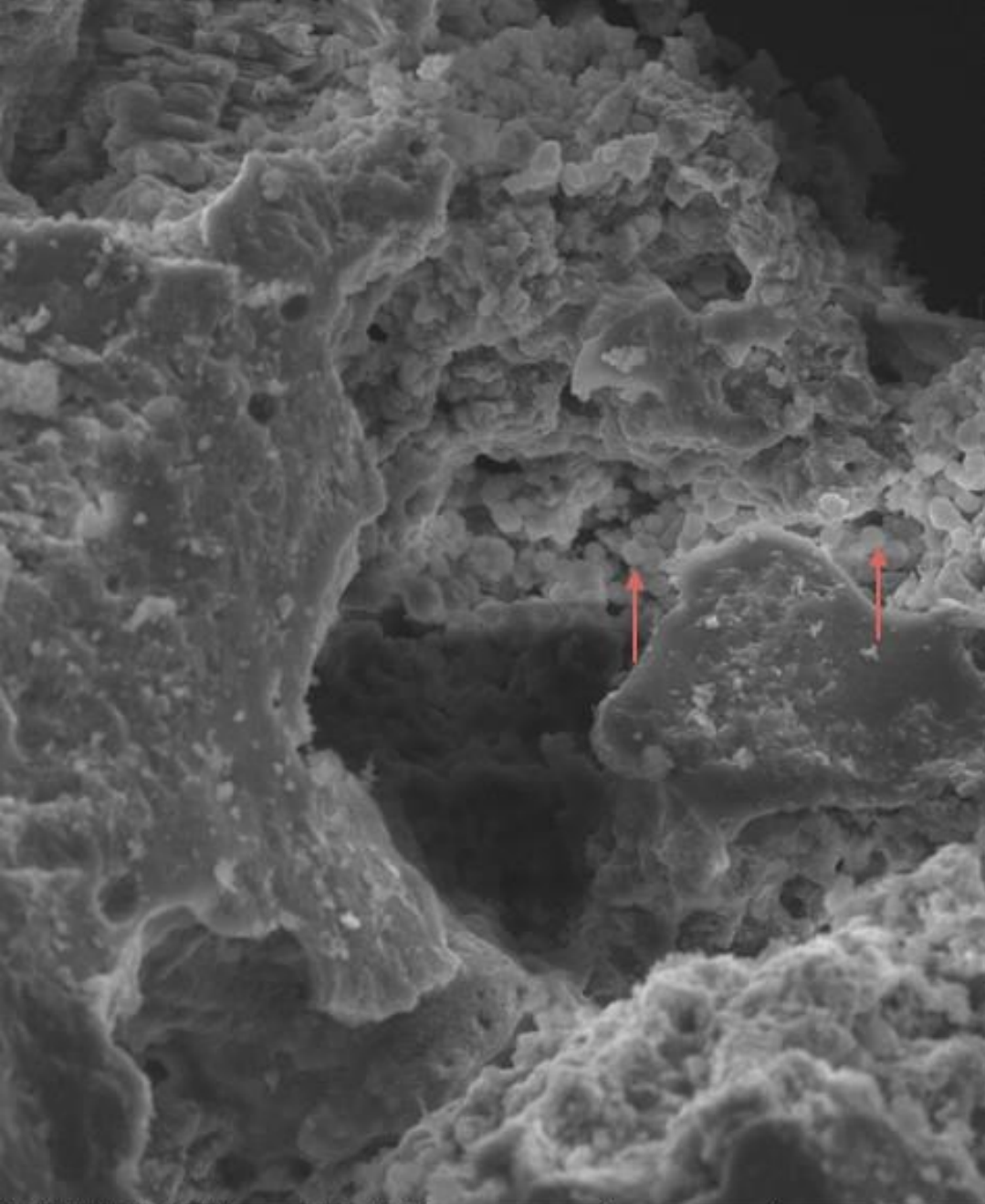


6. HROB TRÓJANKY



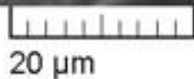


Gardrenella vaginalis
Staphylococcus saprophyticus



SEM HV: 20.00 kV
View field: 144.5 μm
SEM MAG: 2.00 kx

WD: 15.28 mm
Det: SE
Date(m/d/y): 06/02/14





Děkuji za pozornost!