Beyond the molecular clock

Kelley Harris
University of Washington

Workshop on Population and Speciation Genomics
January 31, 2018

Forces that shape genomic diversity

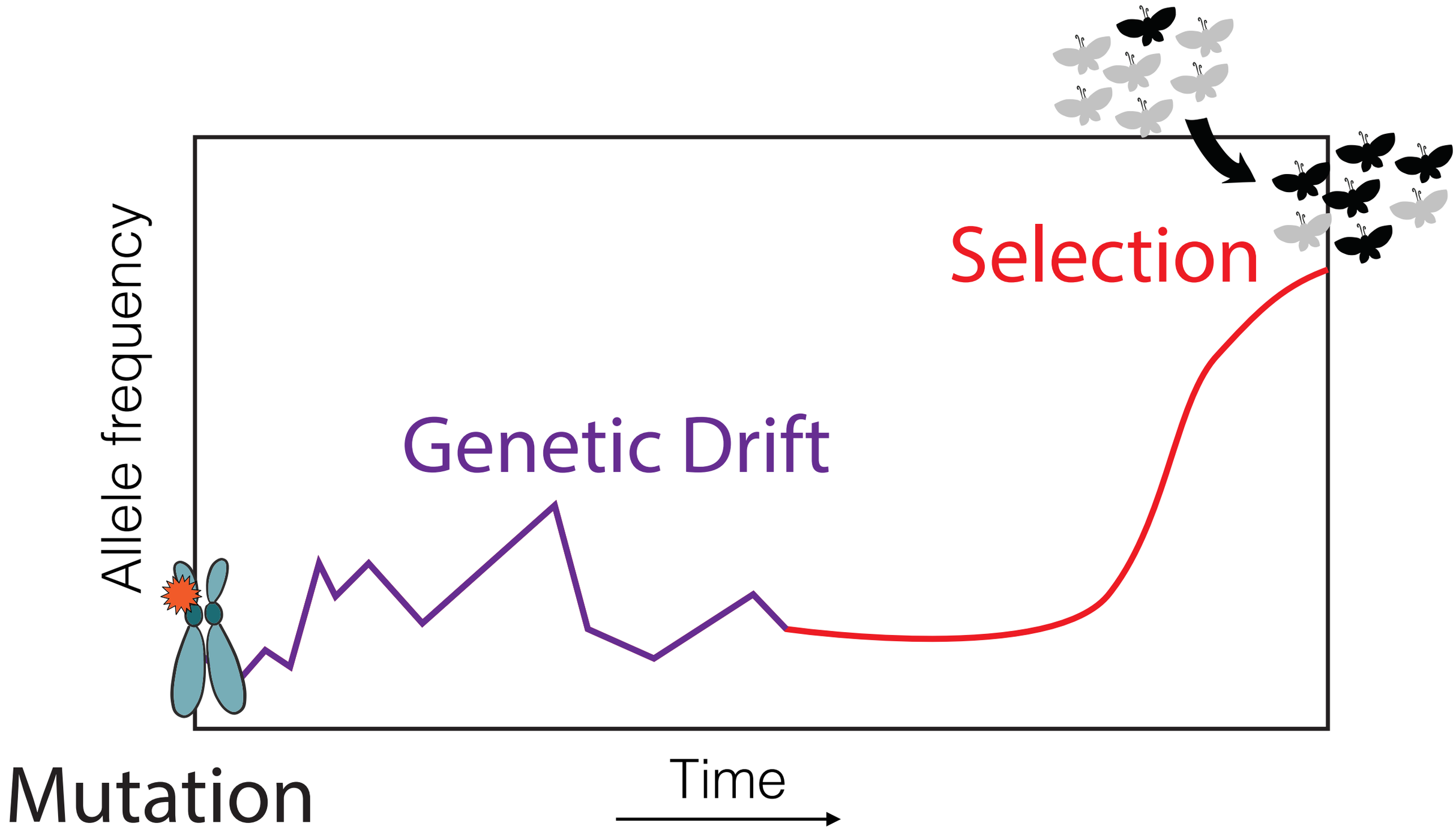
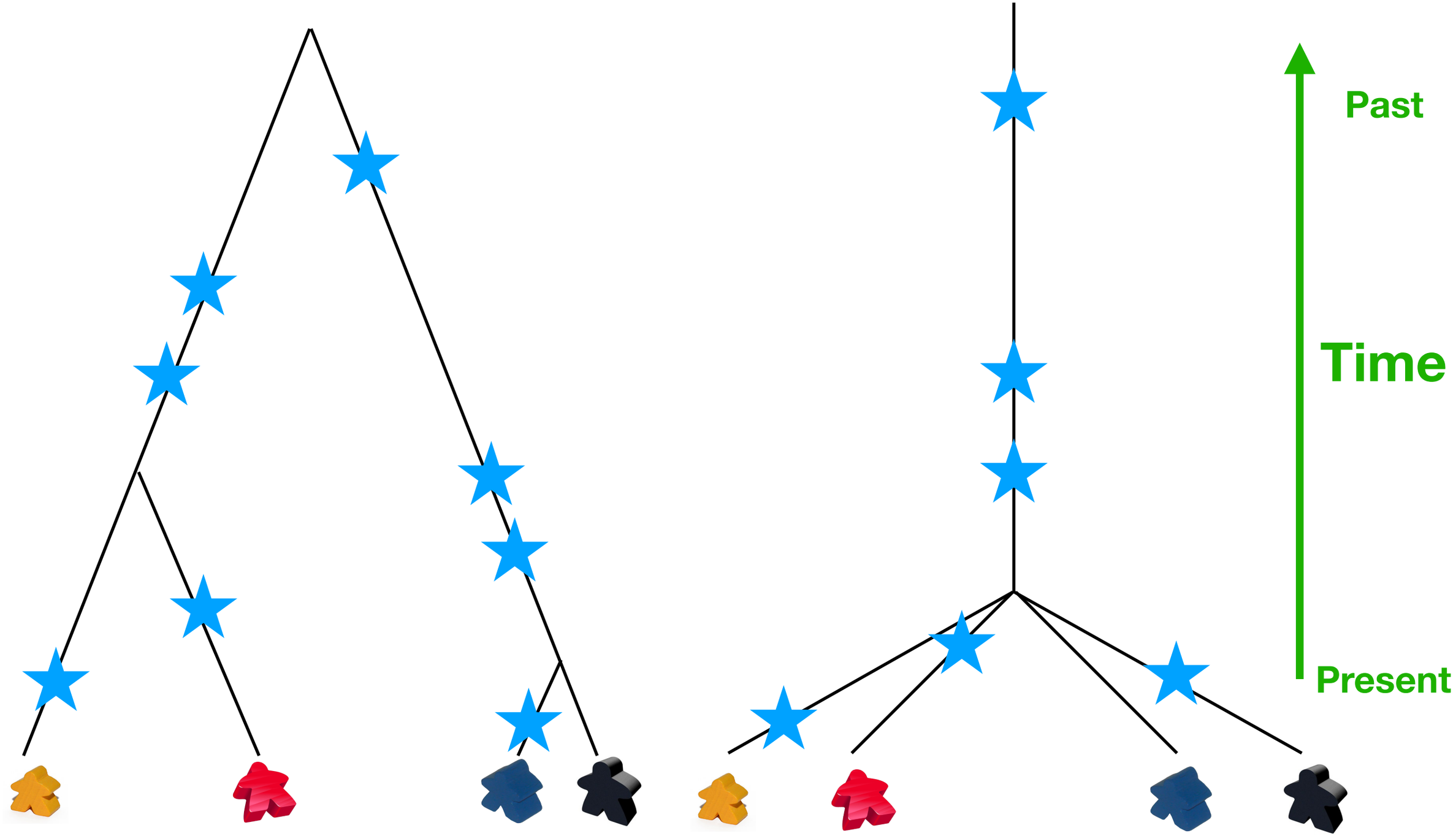


Image co-artist: Natalie Telis

Mutations as a molecular clock



When clock breaks down (runs out of batteries?)

- Almost every population genetic method assumes that mutagenesis = a super boring clock like process

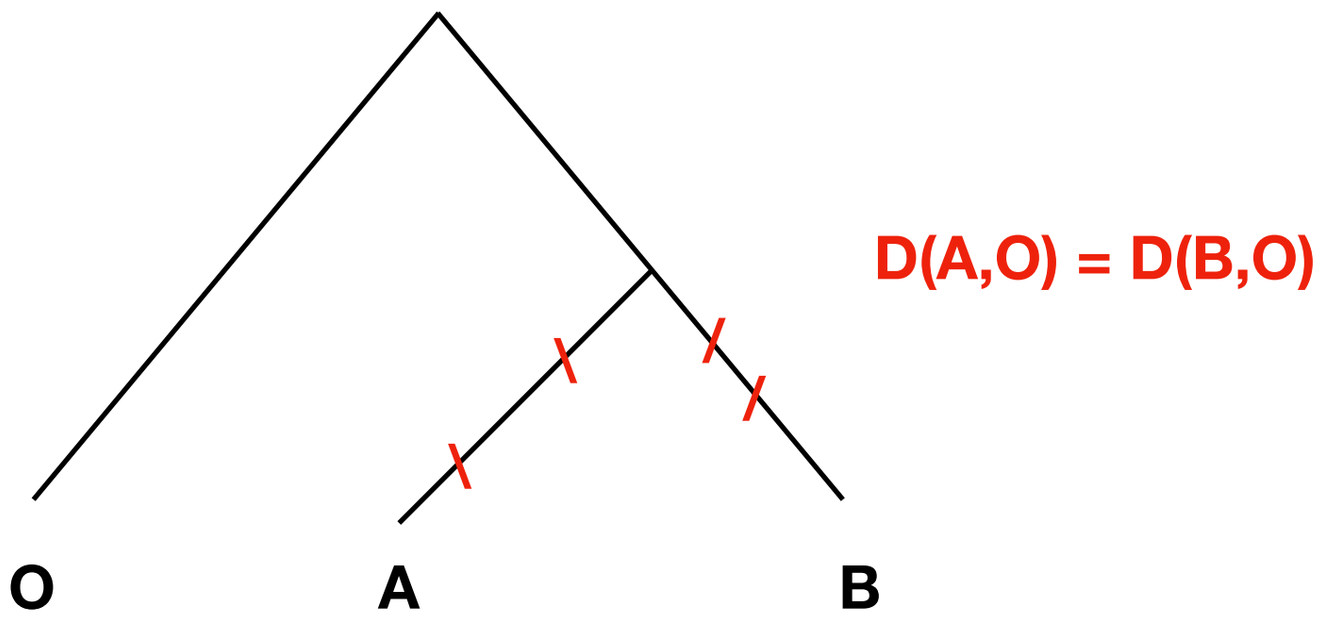
- This assumption works fine until it doesn’t

- The mutation process has cool, complex features that can trip you up if you aren’t looking out for them

Molecular clock 101

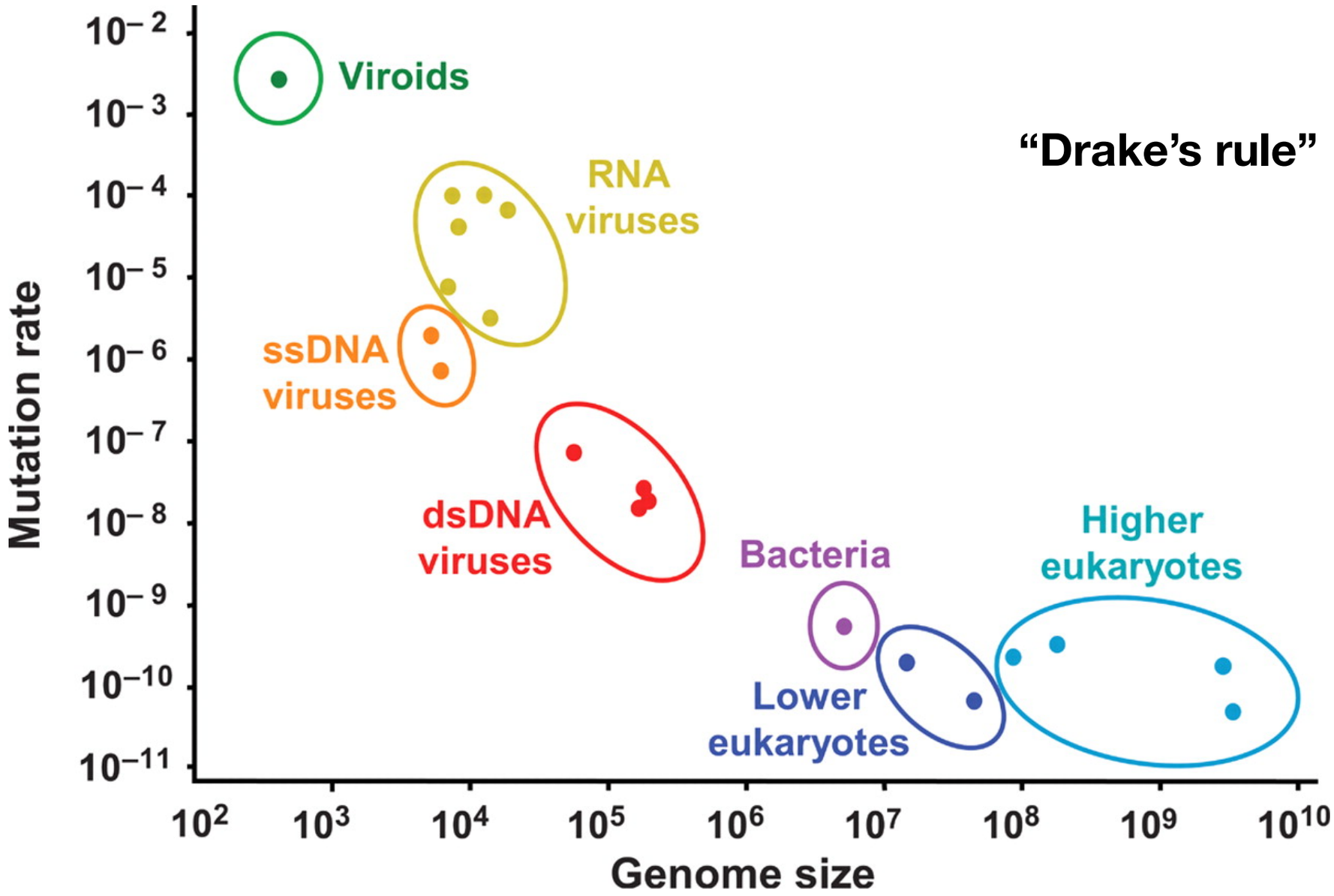
- Mutagenesis is more clock-like over short timescales compared to long time scales

- A simple branch length test can reveal whether mutagenesis is clock-ish in your data:



Data can fail this test due to mutation rate variation, selection, or introgression

Violation of molecular clock over very long timescales



“Drake’s rule”

Drake 1991

Gago, et al. *Science* 2009

The error threshold

- A simple model by Eigen & Schuster (1979) justifies Drake's rule

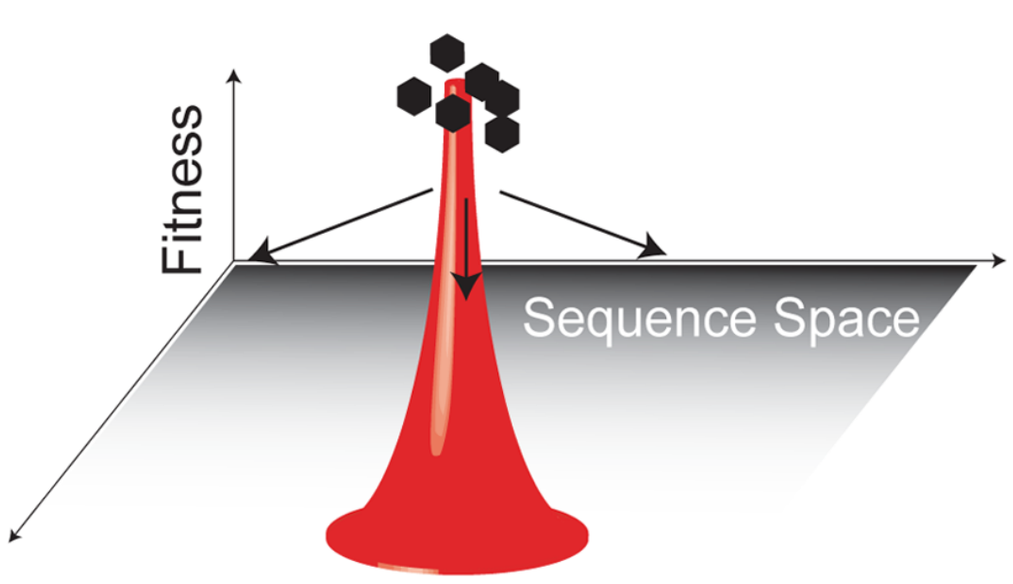
- Consider a "master" virus with fitness 1+*s* and genome length *L*

- All mutant viruses have fitness 1

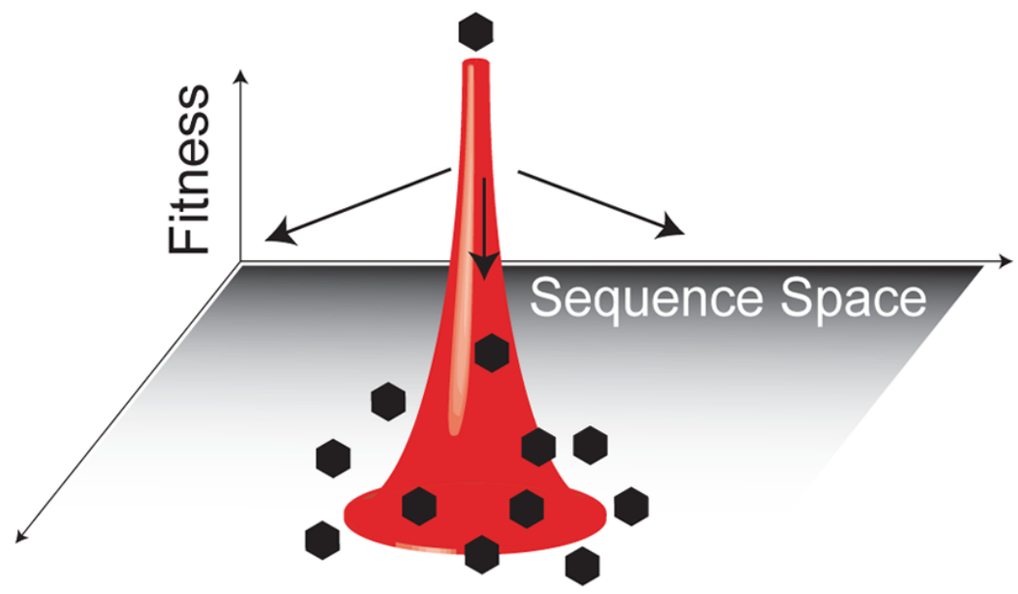
- The master sequence will die out due to Muller's rachet/"error catastrophe" if and only if the mutation rate mu is below a threshold:

- *mu* < log(s)/L

Stable quasispecies vs error catastrophe



mu **< error threshold**

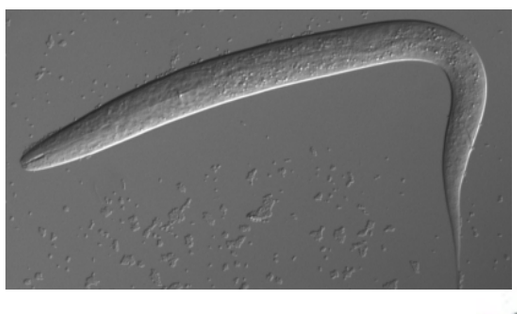


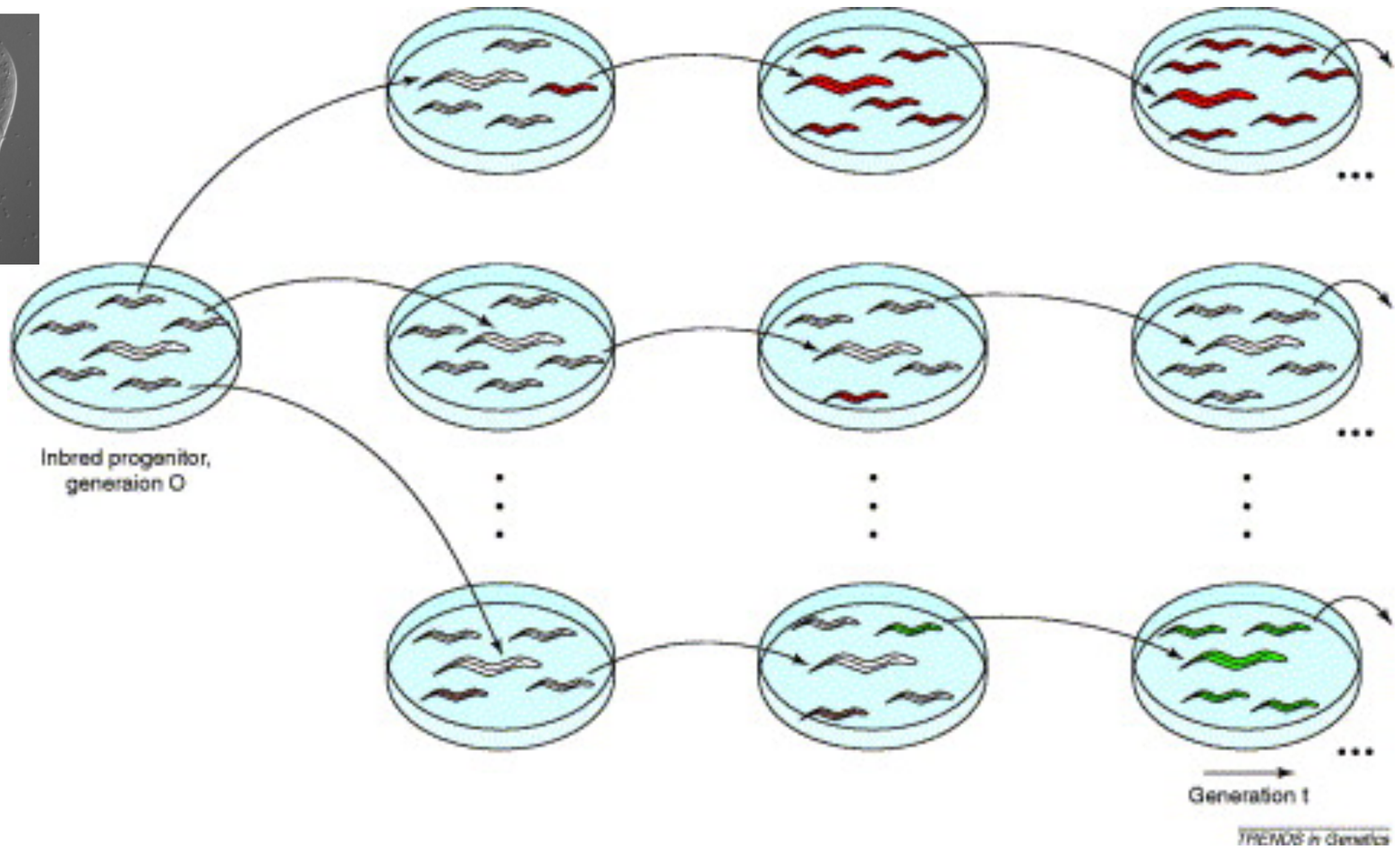
mu **> error threshold**

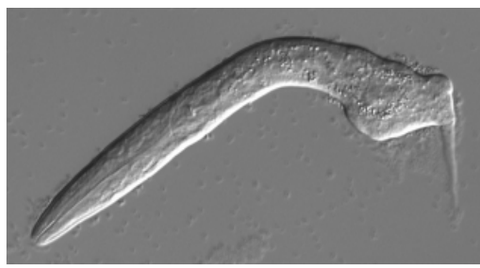
Lauring and Andino 2010

How might I gather some mutation rate data to test this weird theory?

Measuring mutation rates with mutation accumulation (MA) lines







Keightly and Charlesworth 2005

MA with a reporter gene

Reporter gene
(e.g. encoding GFP or
luciferase)

Inactive/ broken promoter

Point mutations can restore promoter function

A reporter protein
Amount is easily measured
(e.g. GFP by fluorescence)

Mutation rate estimates vary enormously in quality

- Your PSMC results depend heavily upon a mutation rate number. Where might that number come from?

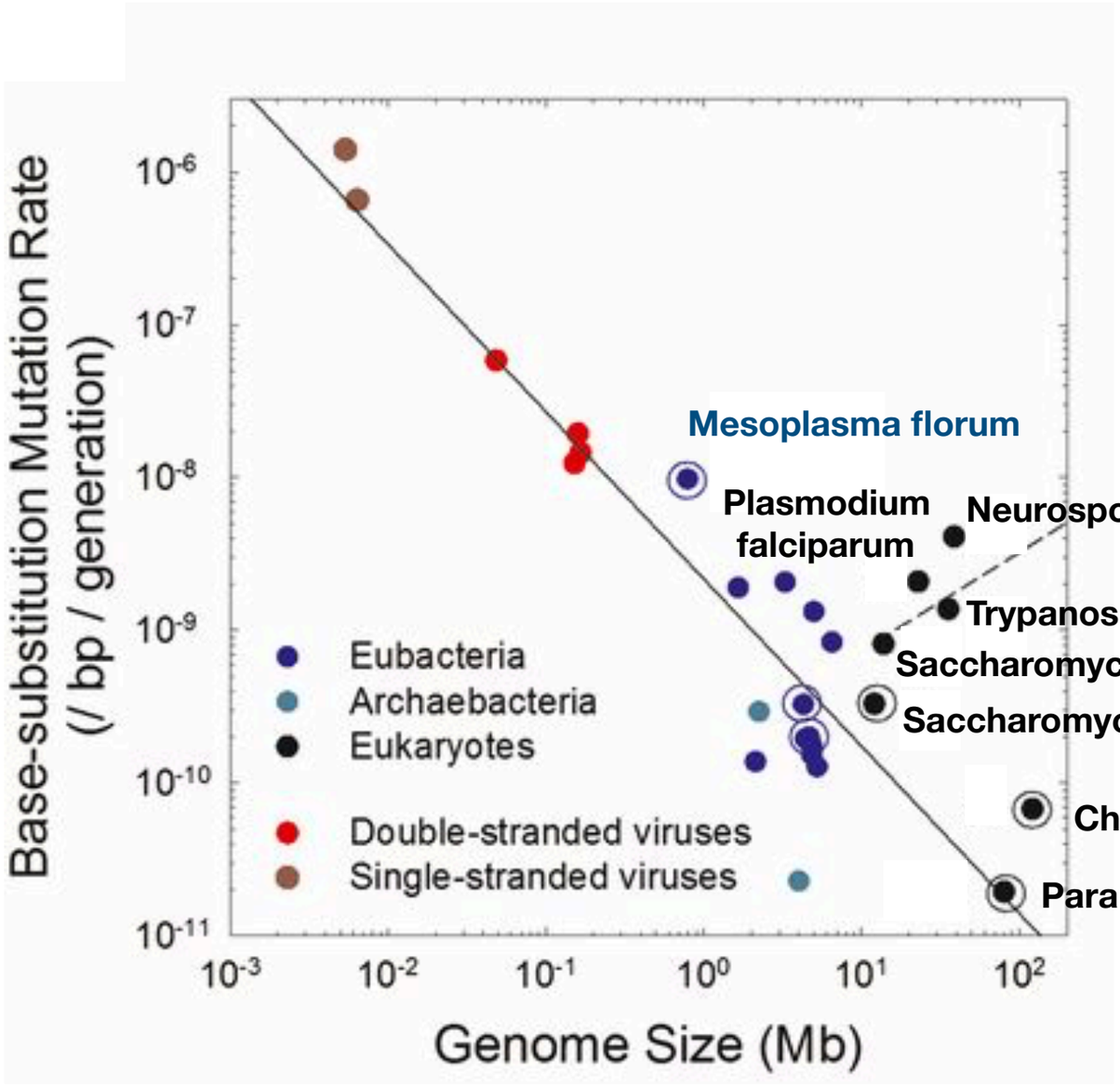
- MA experiment + whole genome sequencing ($$-$$$$)

- MA experiment + reporter gene sequencing (cheap today, only game in town 10 years ago)

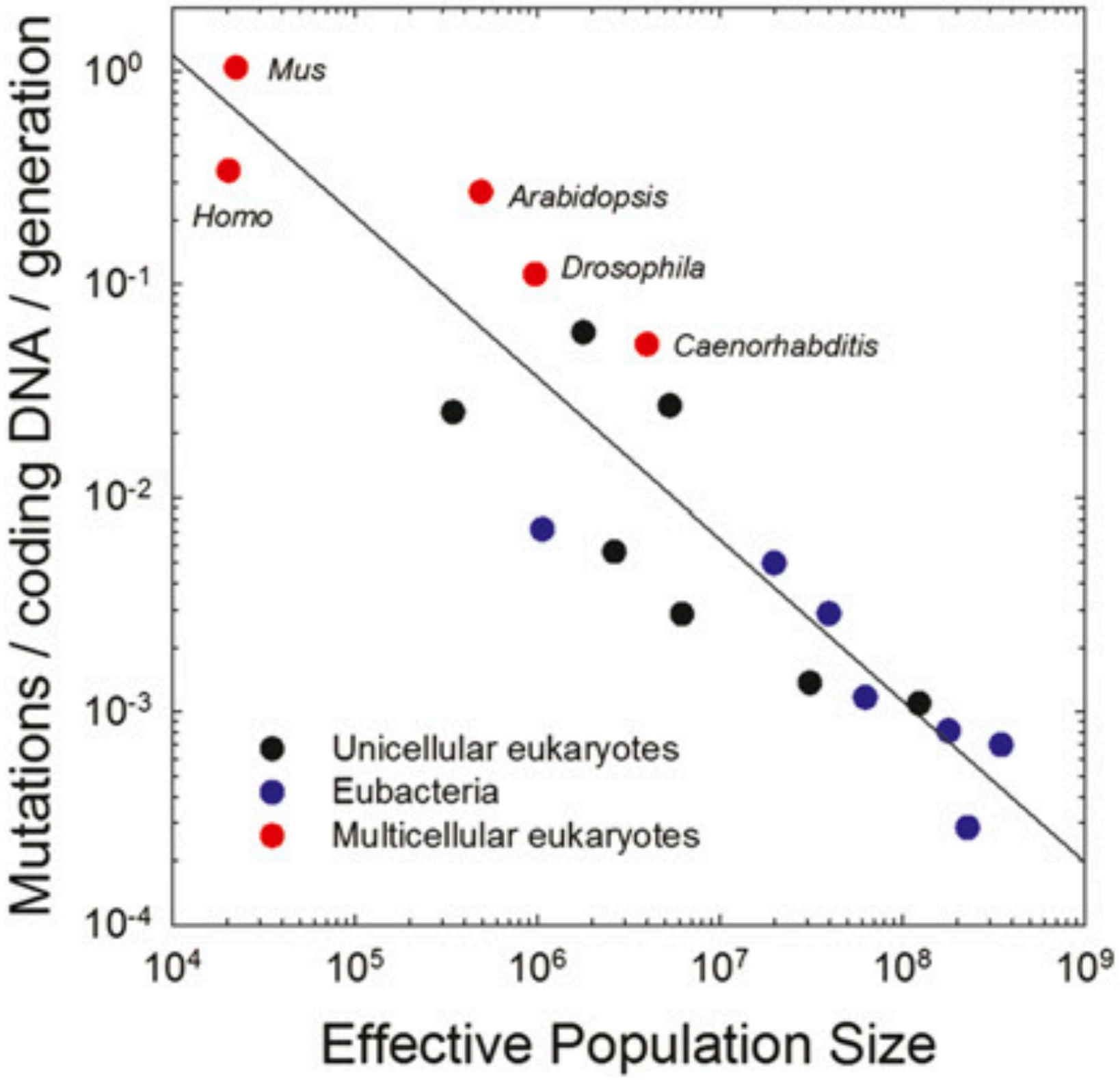
- Back-of-the-envelope calculation (substitutions / estimated divergence time)

- Whole-genome trio sequencing ($$$$$$$$$$)

Drake's rule driven mostly by viruses and bacteria



Sung, et al. 2012



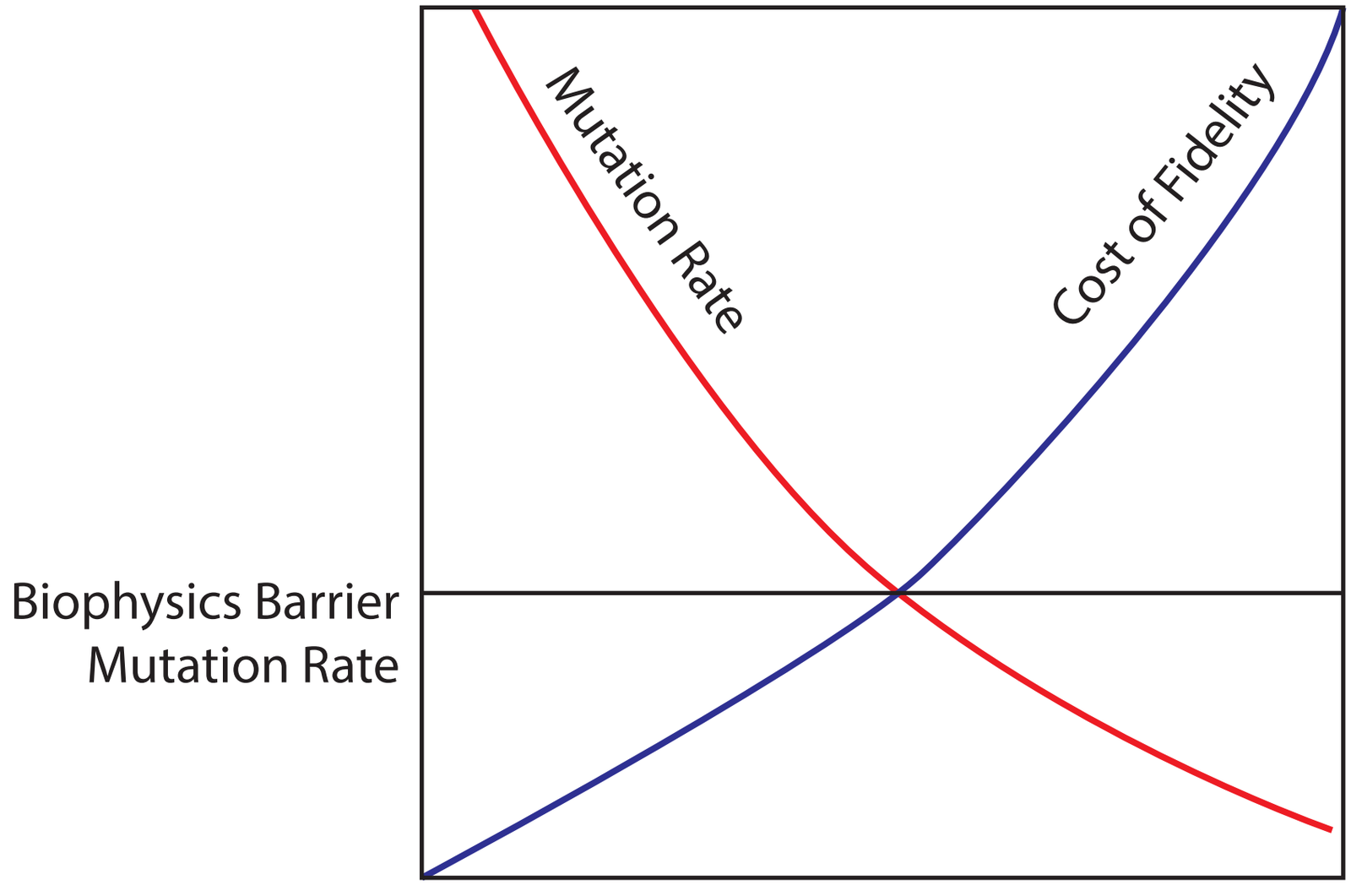
Lynch *Trends in Genetics* 2010

Sung, *et al*. *PNAS* 2012

Why should effective population size affect mutation rate?

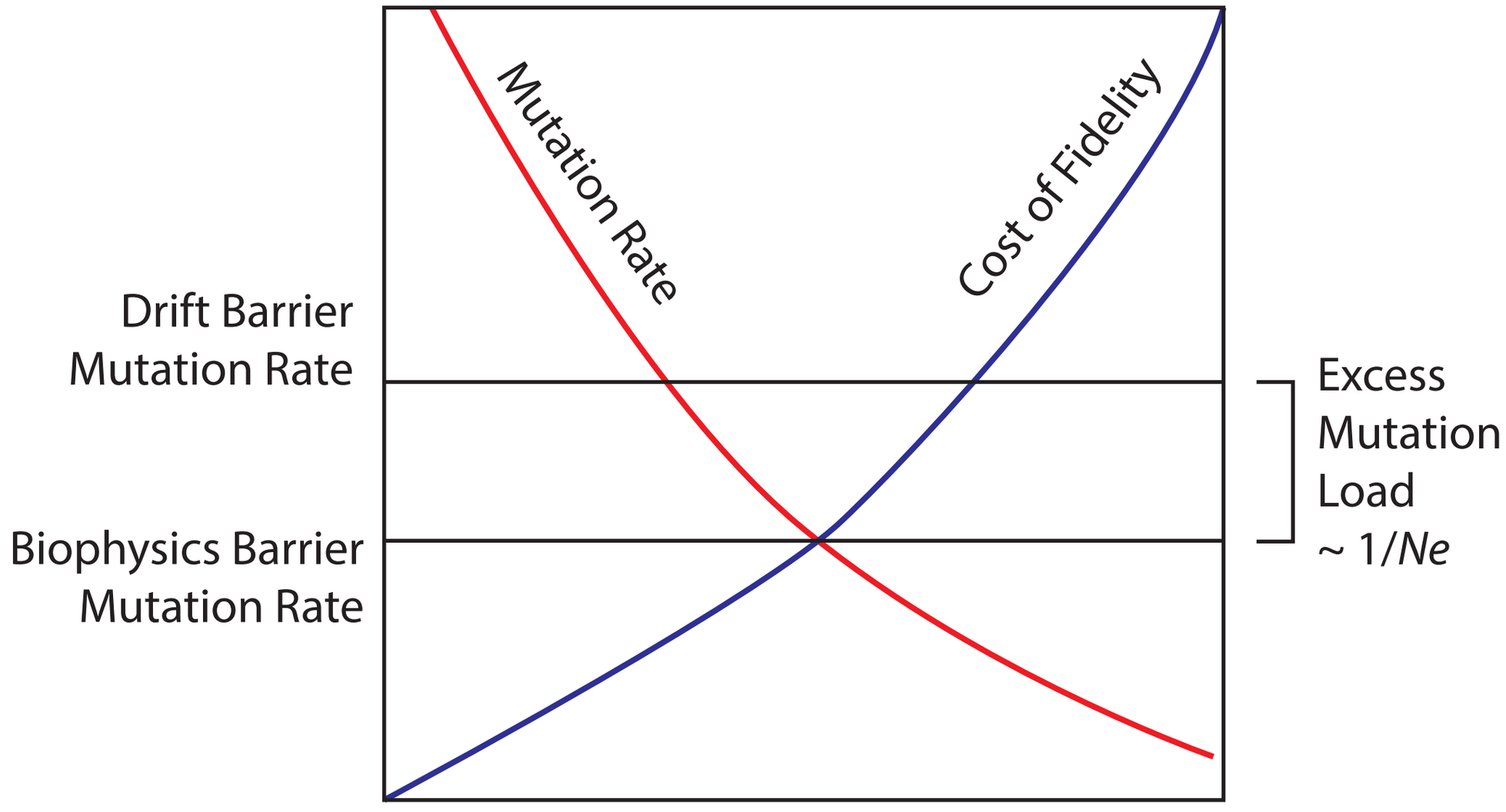
Why is the mutation rate what it is?

1.The Cost-of-Fidelity Model



Lynch *Trends in Genetics* 2010 Sung, *et al*. *PNAS* 2012

2.The Drift-Barrier Hypothesis



Lynch *Trends in Genetics* 2010 Sung, *et al*. *PNAS* 2012

Mutators can be favored in asexual organisms

- Expected extra load of deleterious mutations must not exceed the expected benefit of beneficial mutations
- Robustness to environmental change
- Stress-induced mutagenesis?

Stress-Induced Mutagenesis in Bacteria

Ivana Bjedov[1,*], Olivier Tenaillon[2,*], Bénédicte Gérard[2,*], Valeria Souza[3], Erick Denamur…

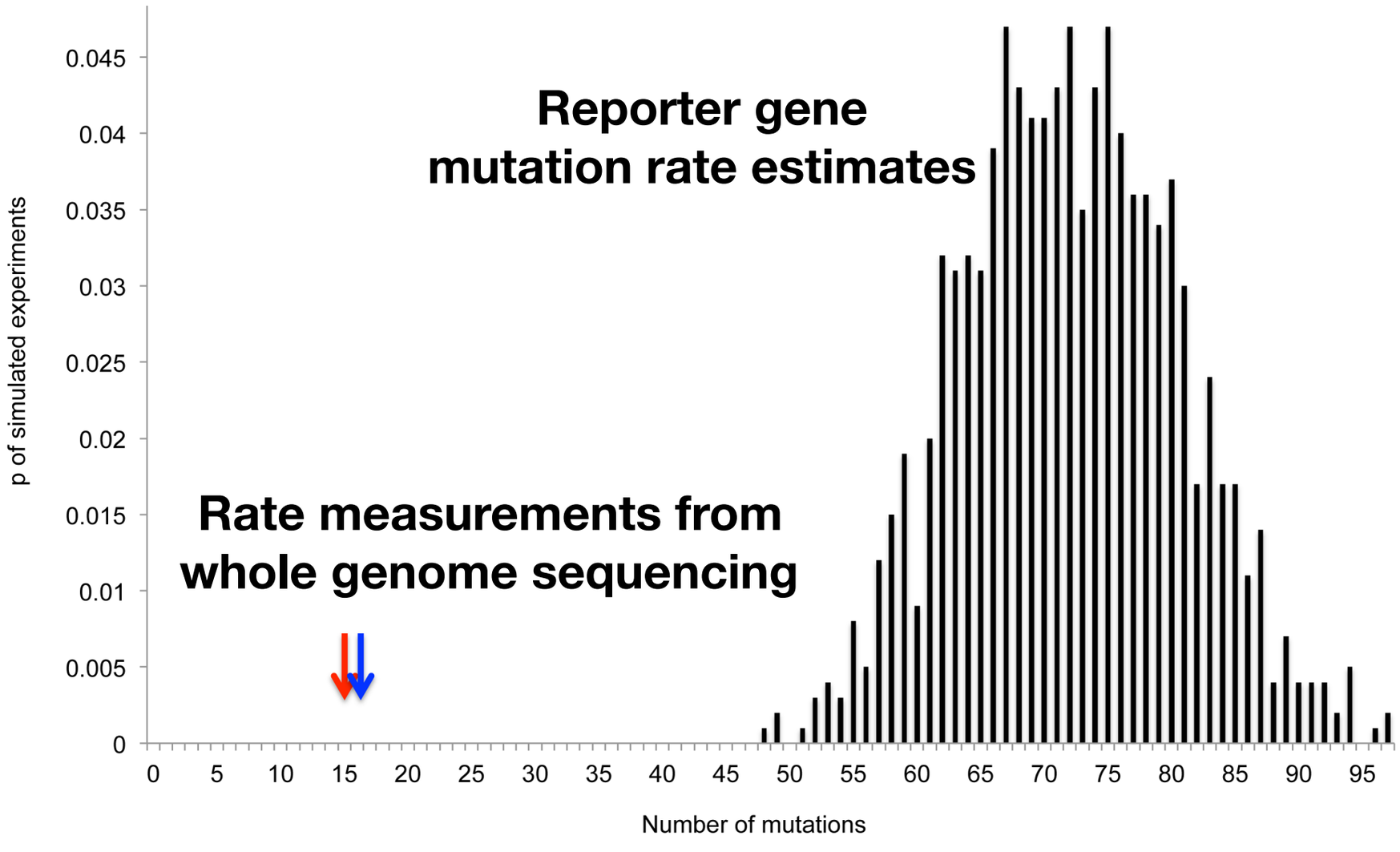
+ See all authors and affiliations

Science 30 May 2003:
Vol. 300, Issue 5624, pp. 1404-1409
DOI: 10.1126/science.1082240

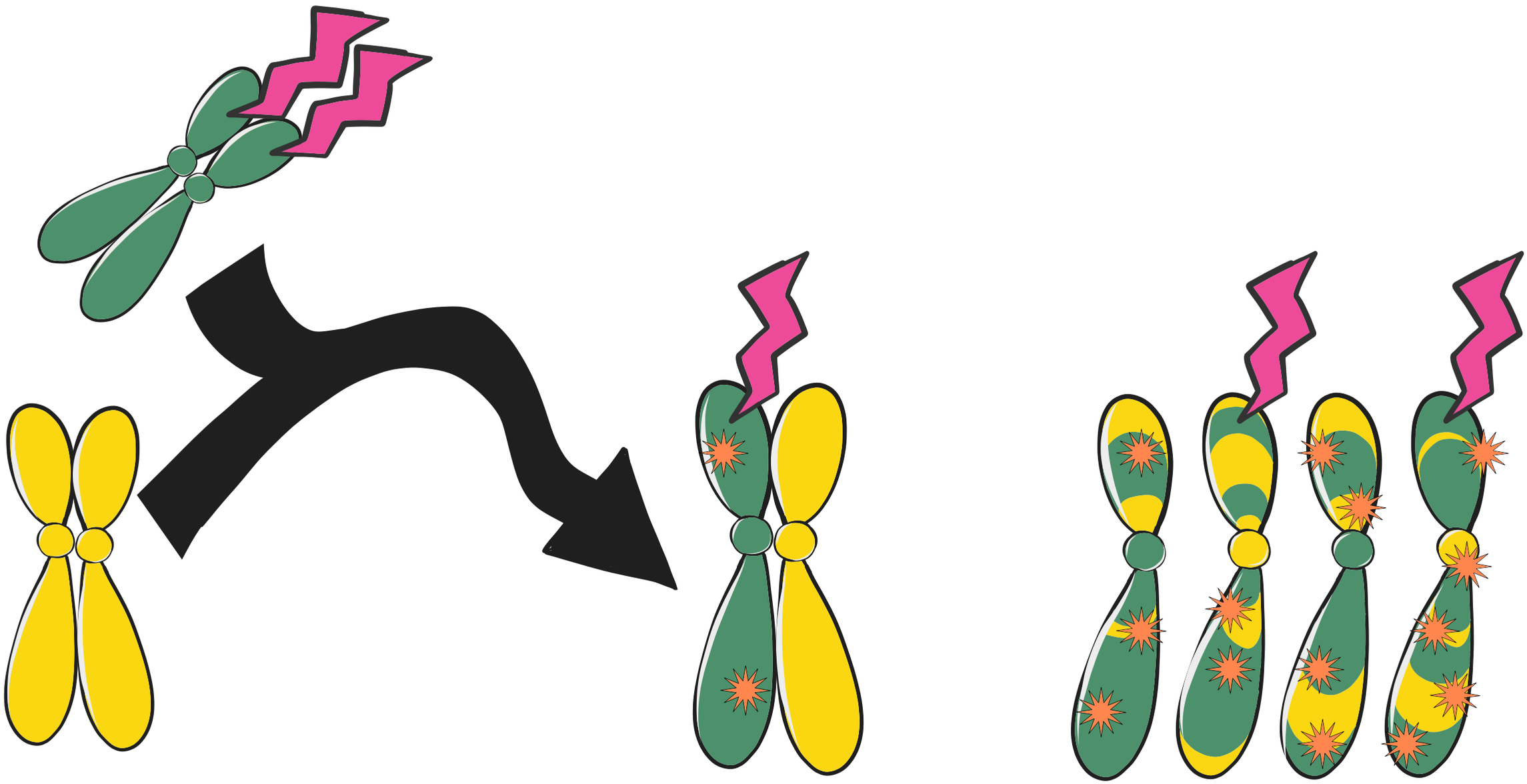
Elevated Mutagenesis Does Not Explain the Increased Frequency of Antibiotic Resistant Mutants in Starved Aging Colonies

Sophia Katz, Ruth Hershberg

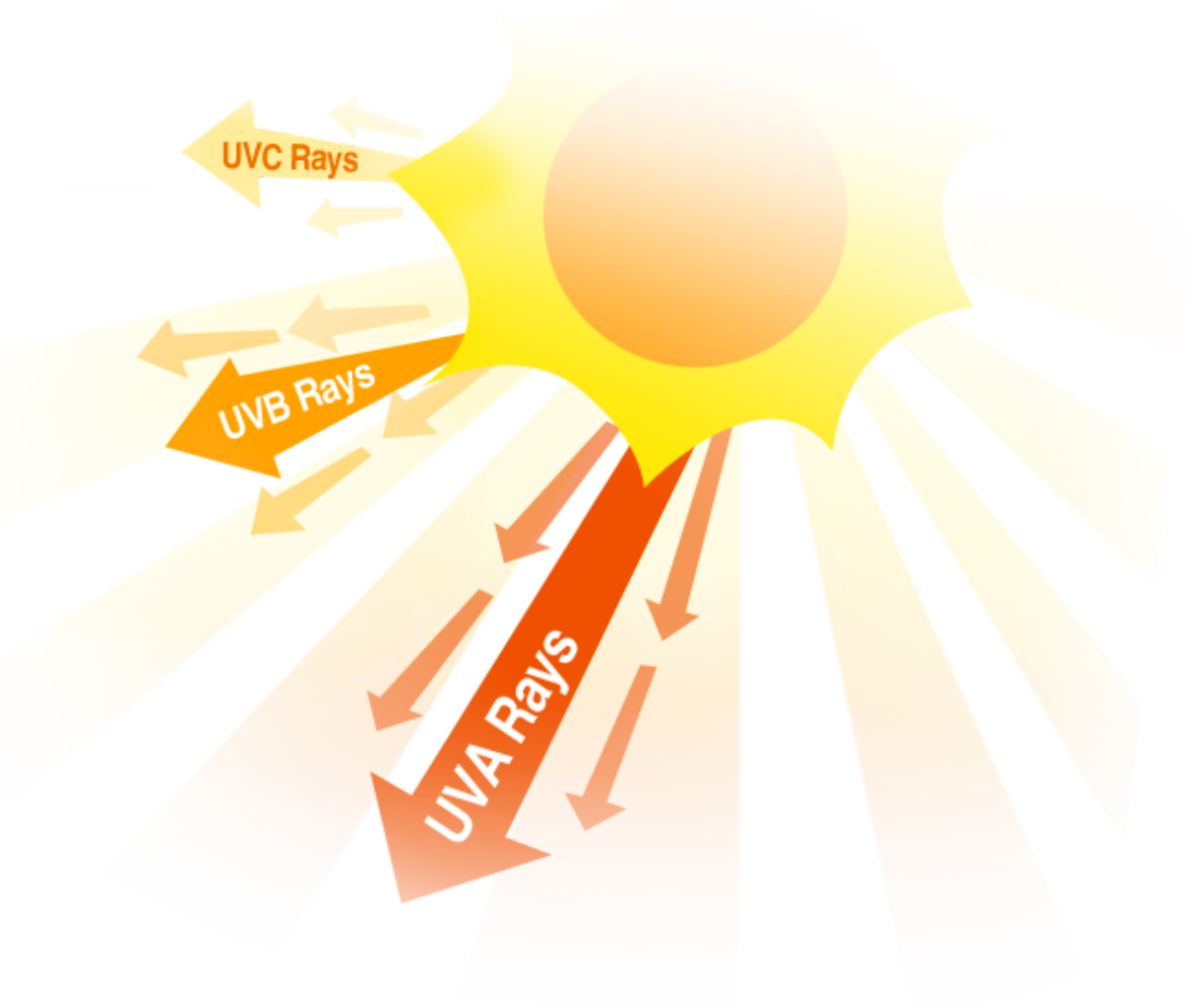
Published: November 14, 2013 • https://doi.org/10.1371/journal.pgen.1003968



Selection against mutator alleles is weak in sexual organisms



Other factors affecting the mutation rate

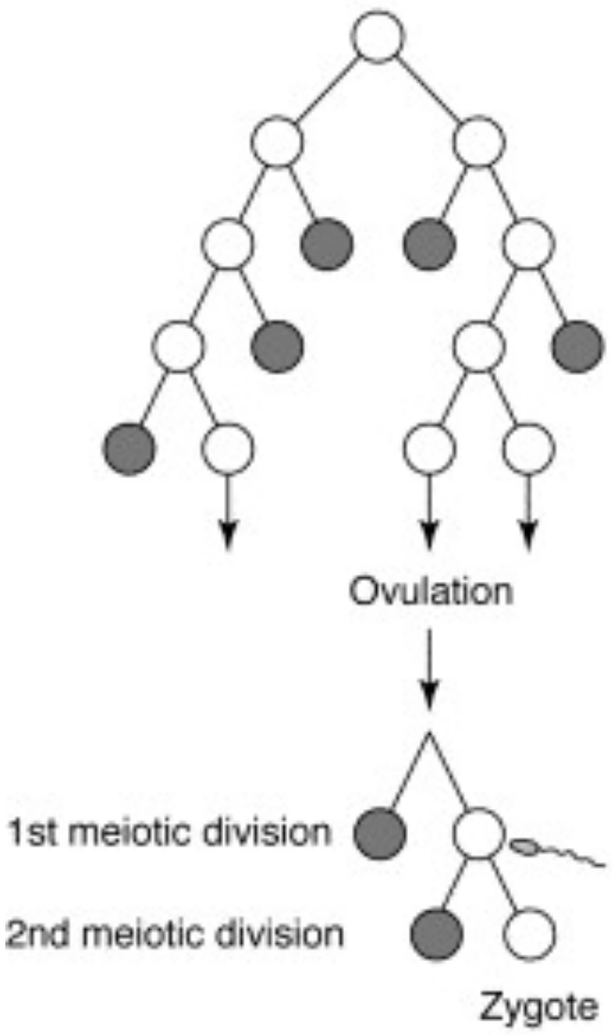


Environmental Mutagens

Life history

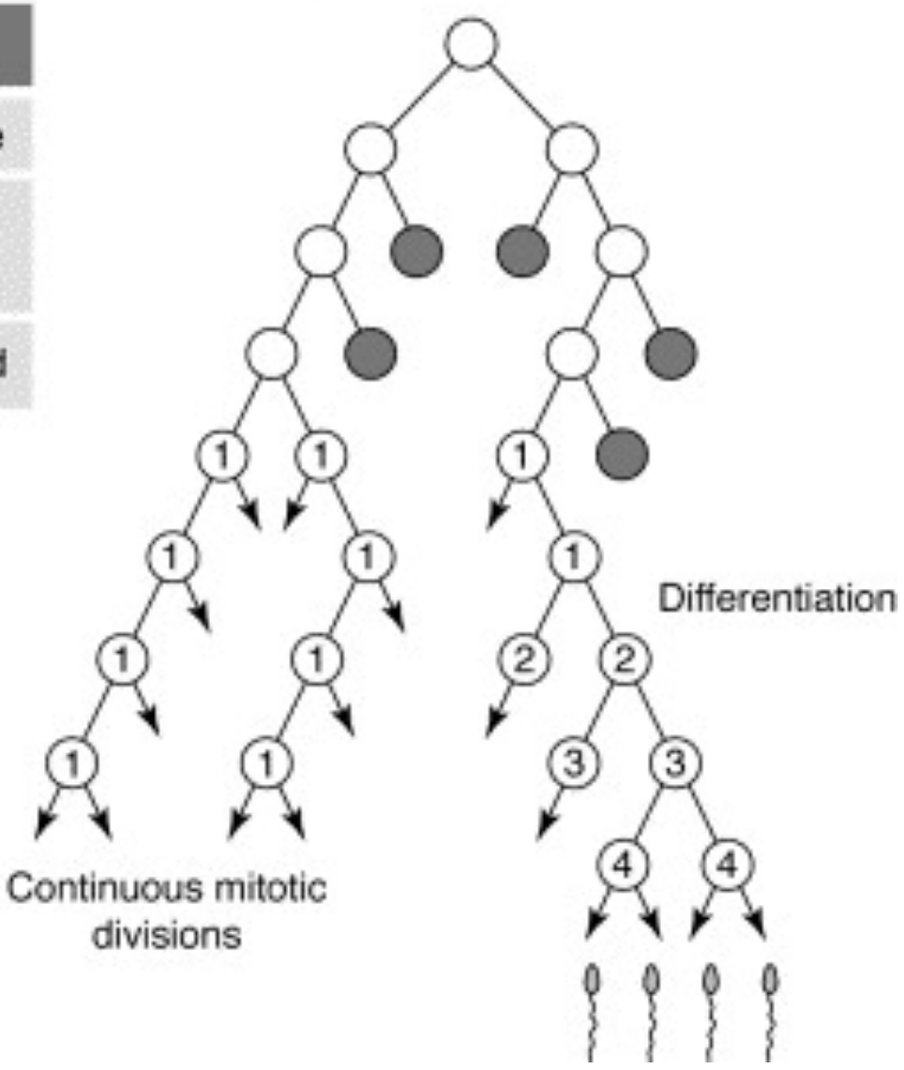
Male mutation bias

Oogenesis



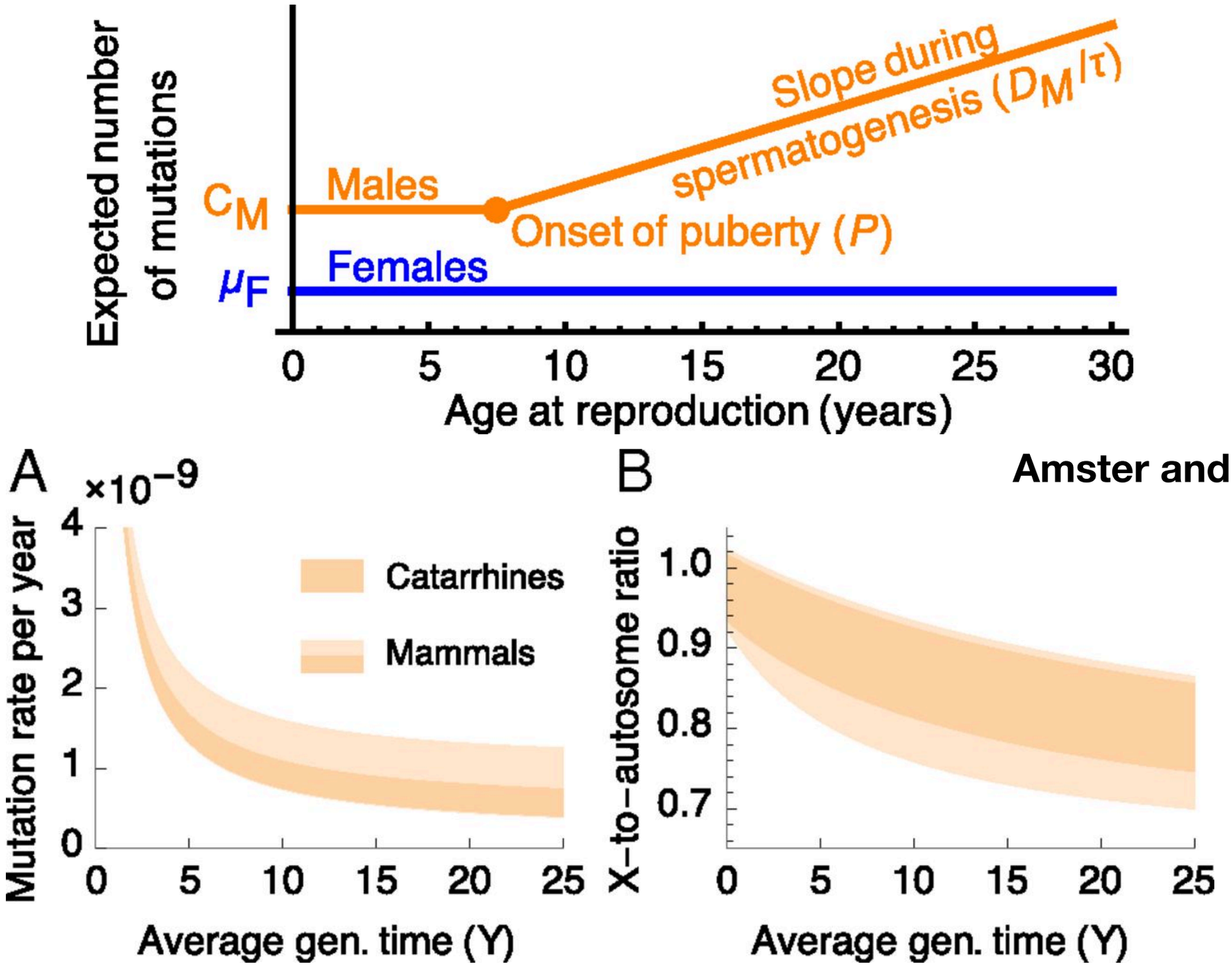
♀		♂	
Timetable	No. of cell divisions		Timetable
5th month of gestation	22	30	Puberty
Sexual maturity	2	23 per year	Adulthood
Total:	24	150 at 20 yr 380 at 30 yr 610 at 40 yr	

Spermatogenesis



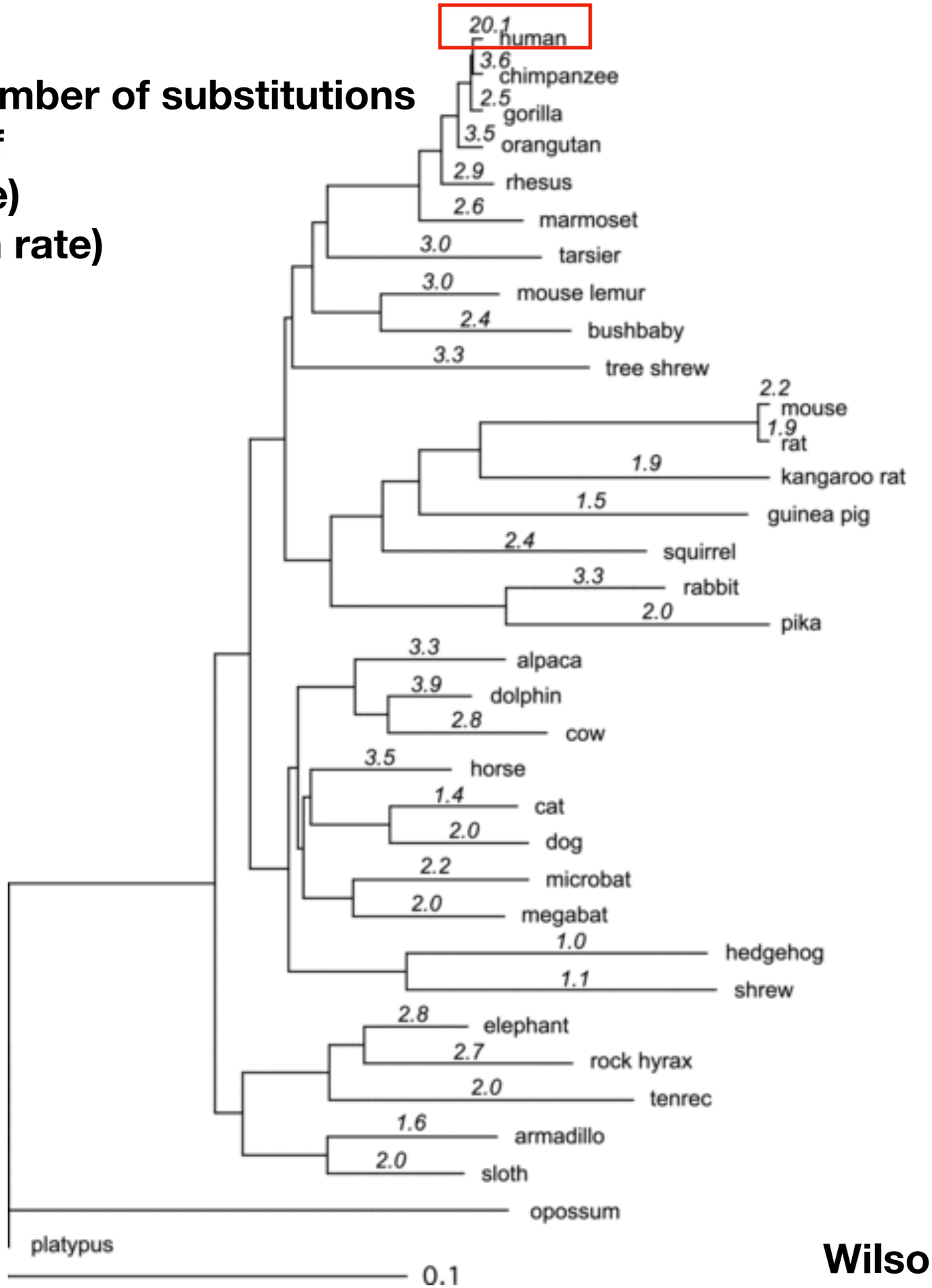
Hurst and Ellegren 1998

Paternal age effect



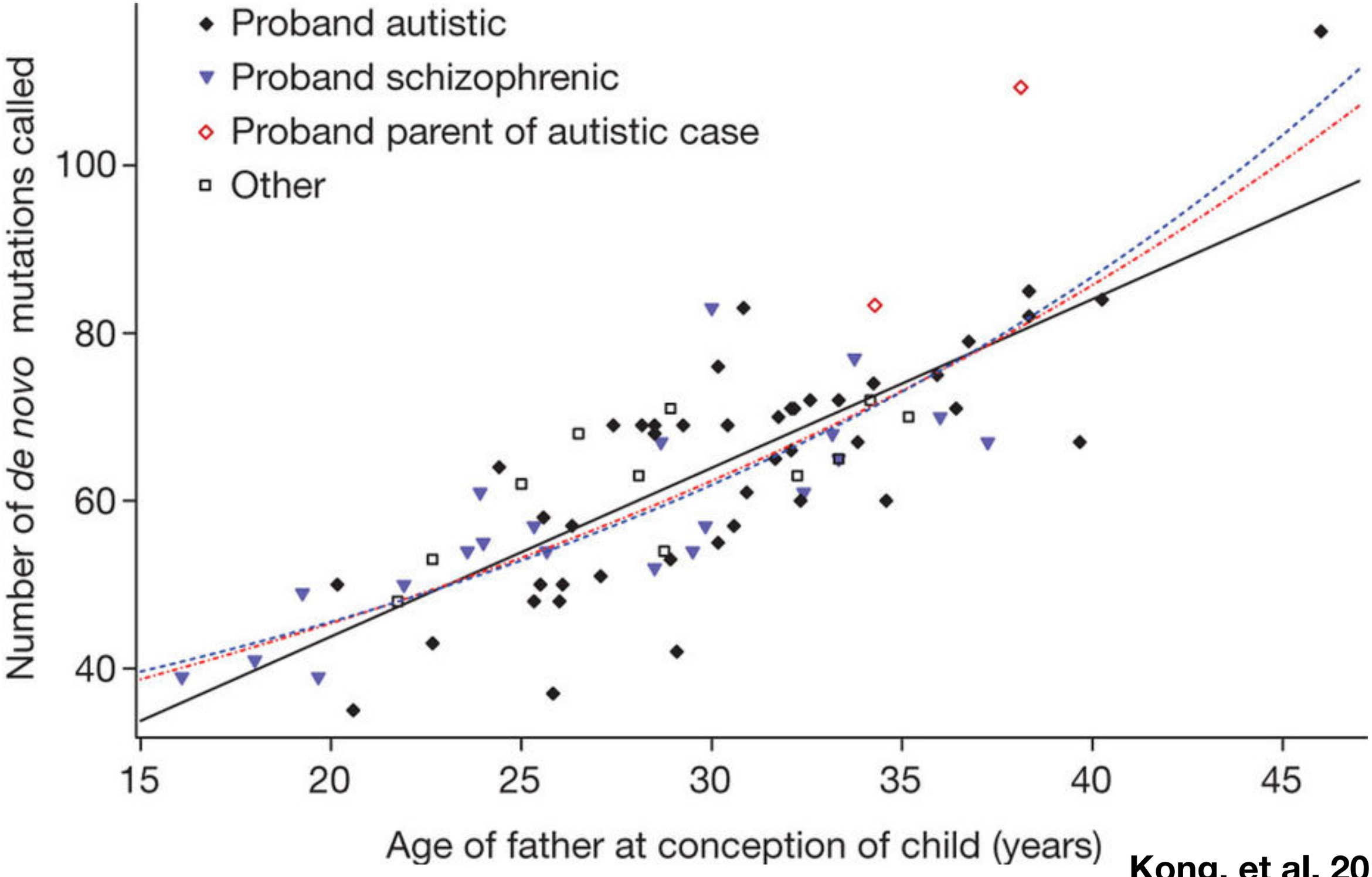
Amster and Sella 2016

Branch length ~ number of substitutions
Label = Estimate of
(male mutation rate)
/(female mutation rate)



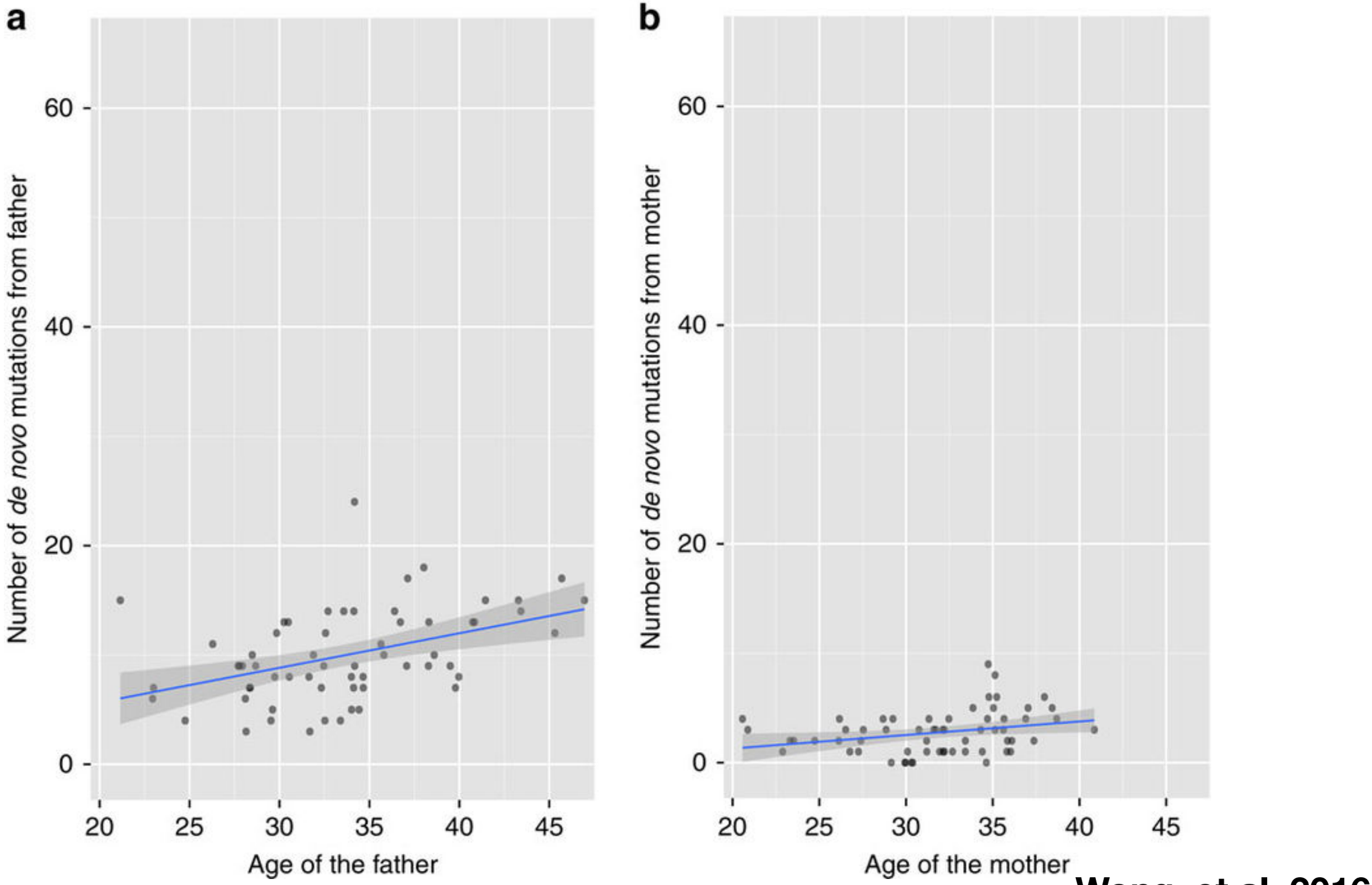
Wilson Sayres, et al. 2011

Two additional *de novo* mutations per year of paternal age



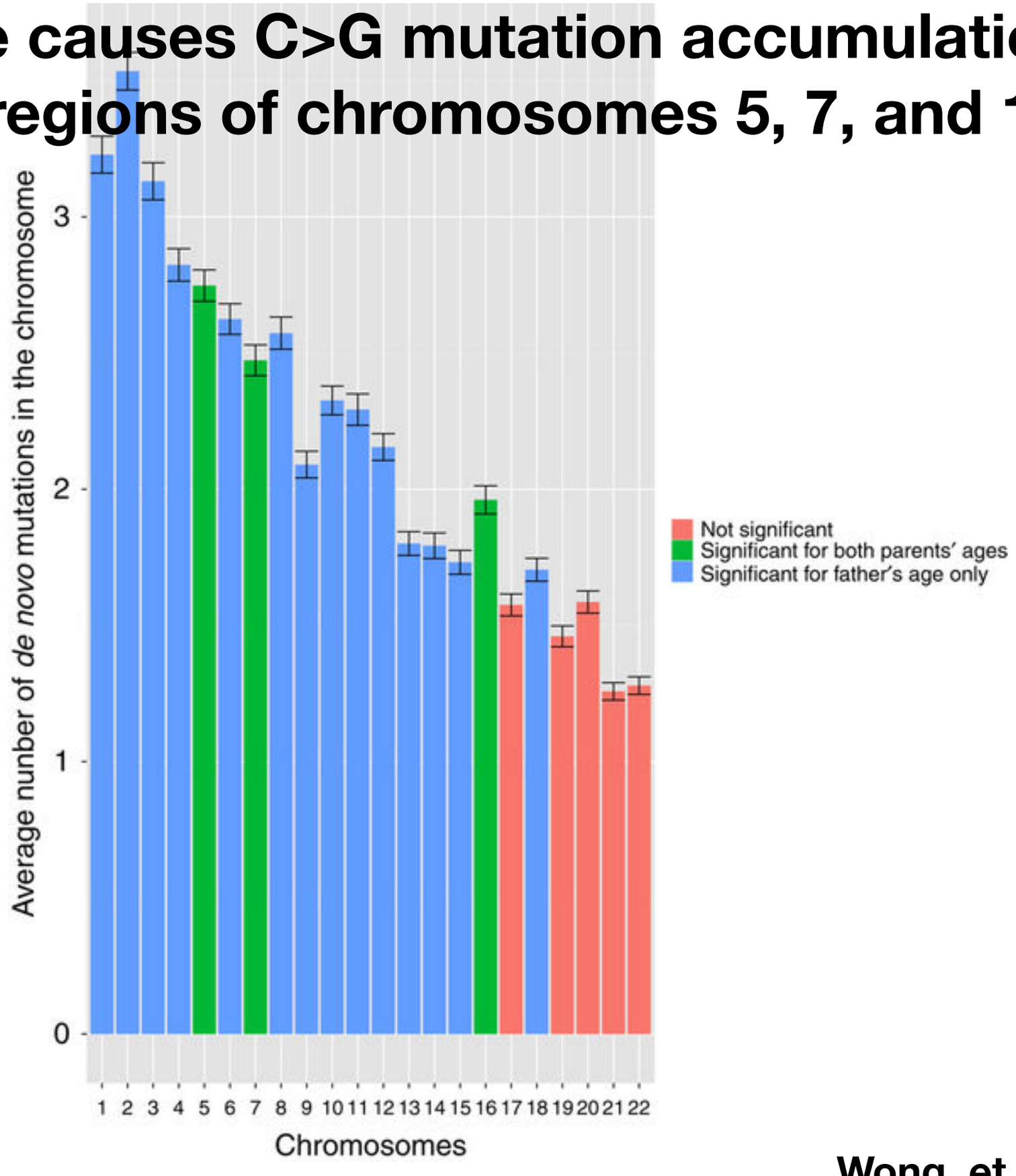
Kong, et al. 2012

A small but significant maternal age effect (0.5 muts/year)



Wong, et al. 2016

Maternal age causes C>G mutation accumulation in localized regions of chromosomes 5, 7, and 16



Wong, et al. 2016

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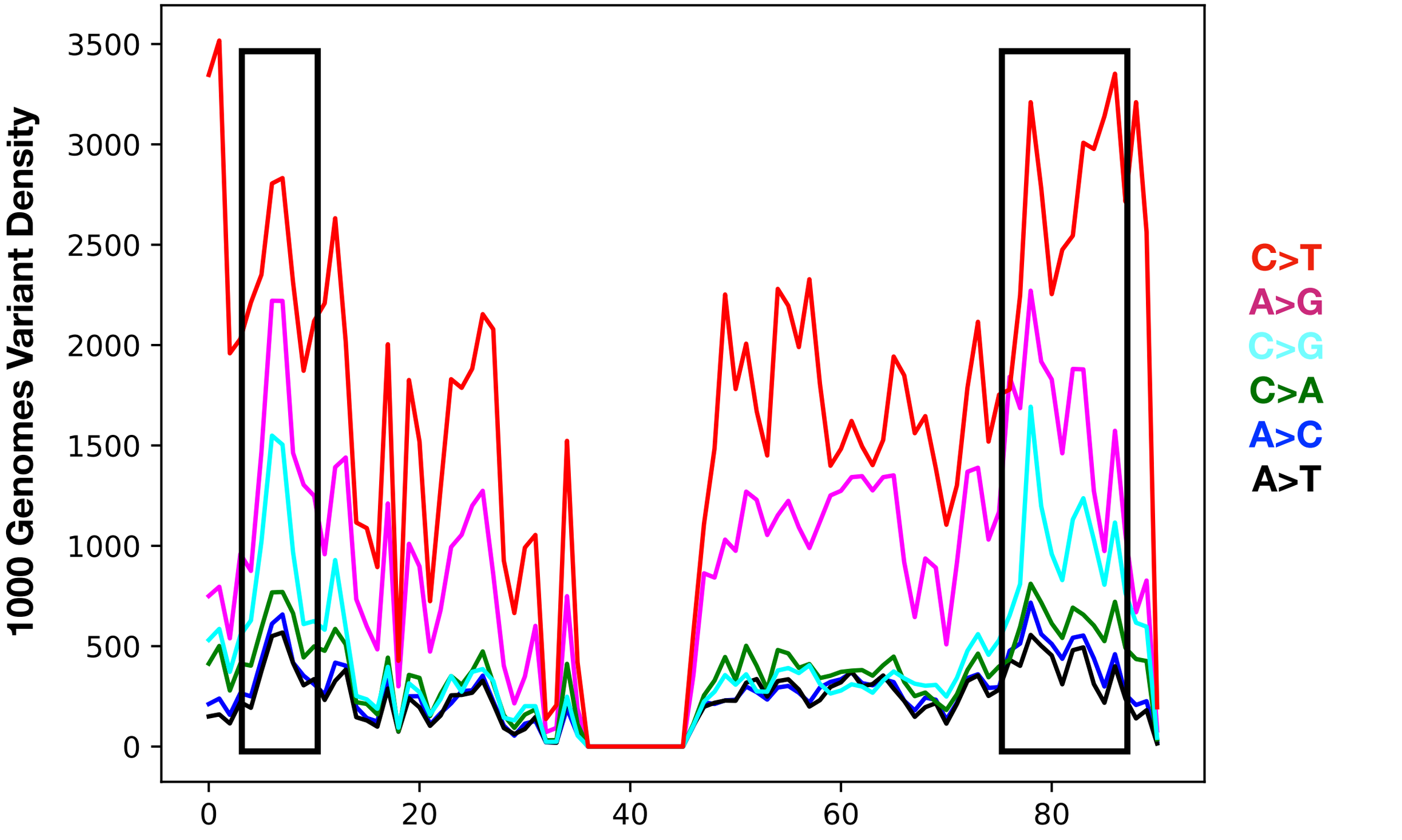
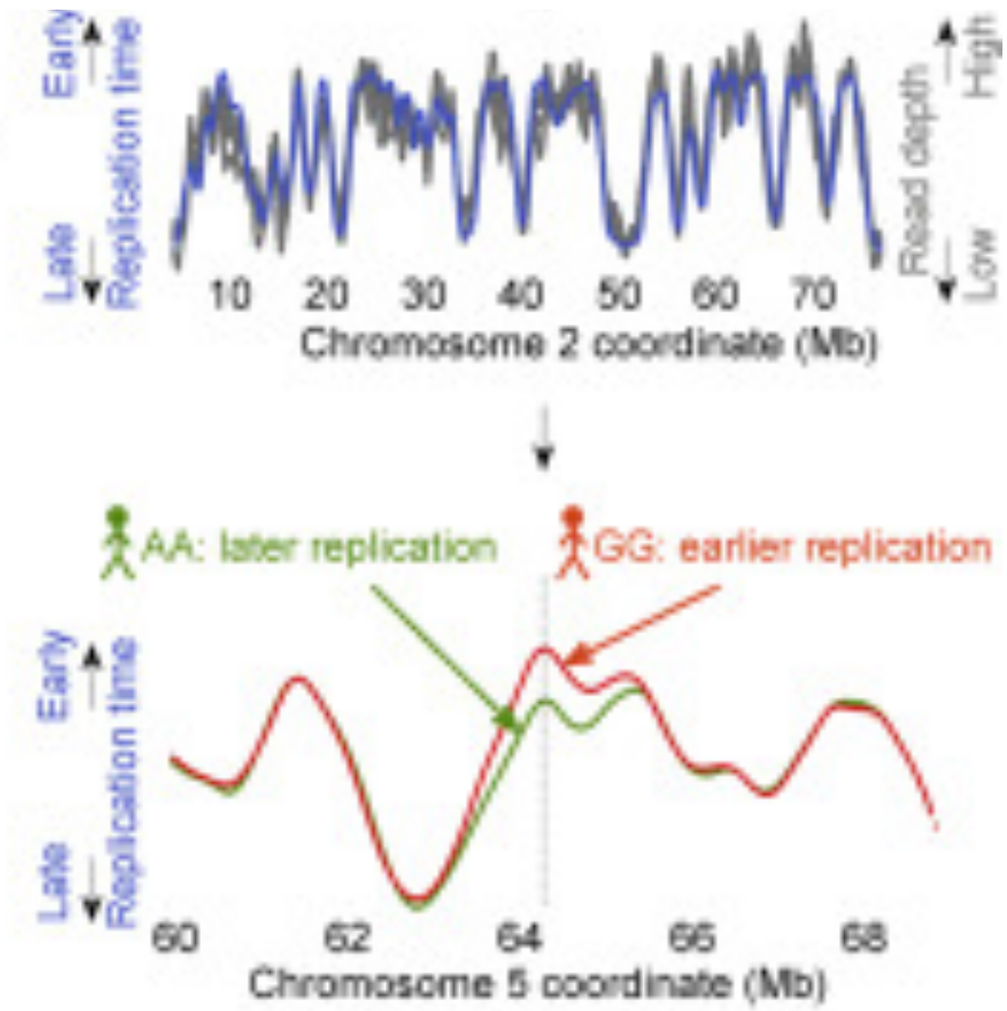


Wong, et al. 2016

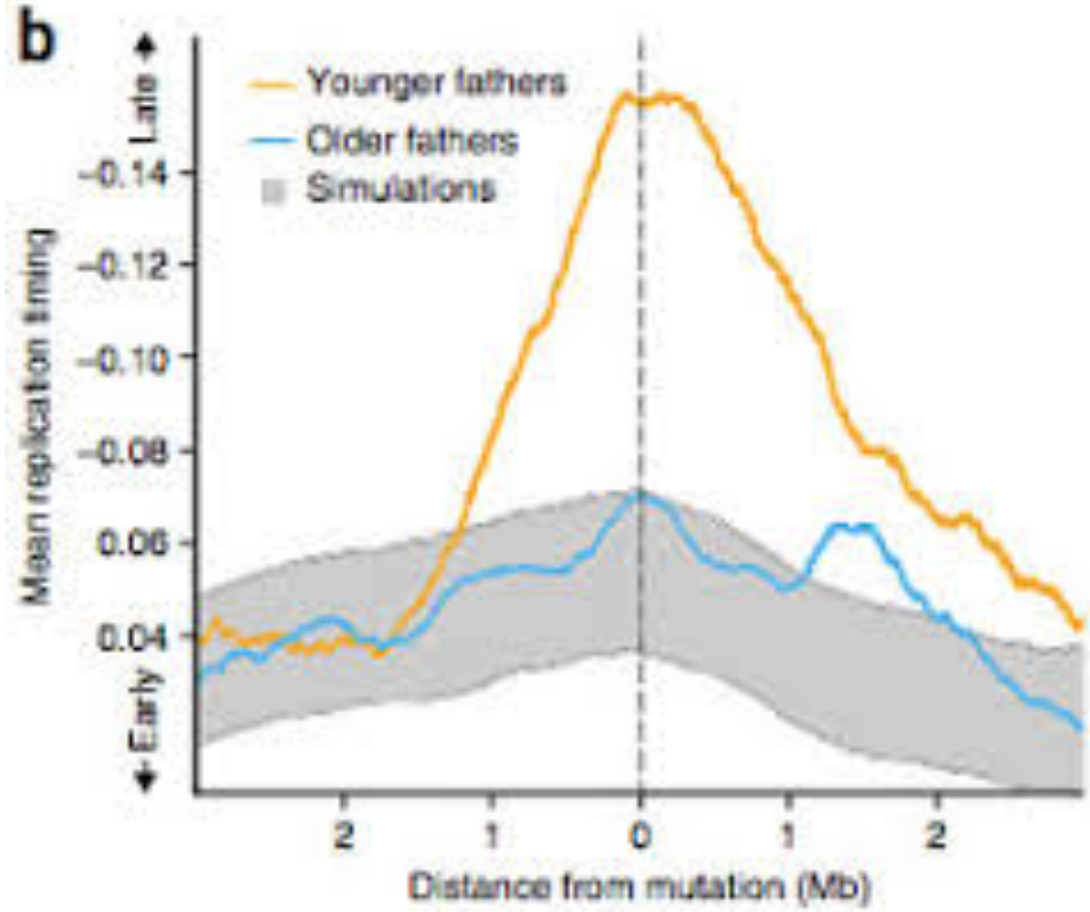
Other causes of mutation rate variation along the genome

- Replication timing
- Transcription-associated-mutagenesis (TAM) and transcription-coupled-repair (TCR)
- Non-B-DNA structures and other DNA repeats
- Chromatin state

Replication timing

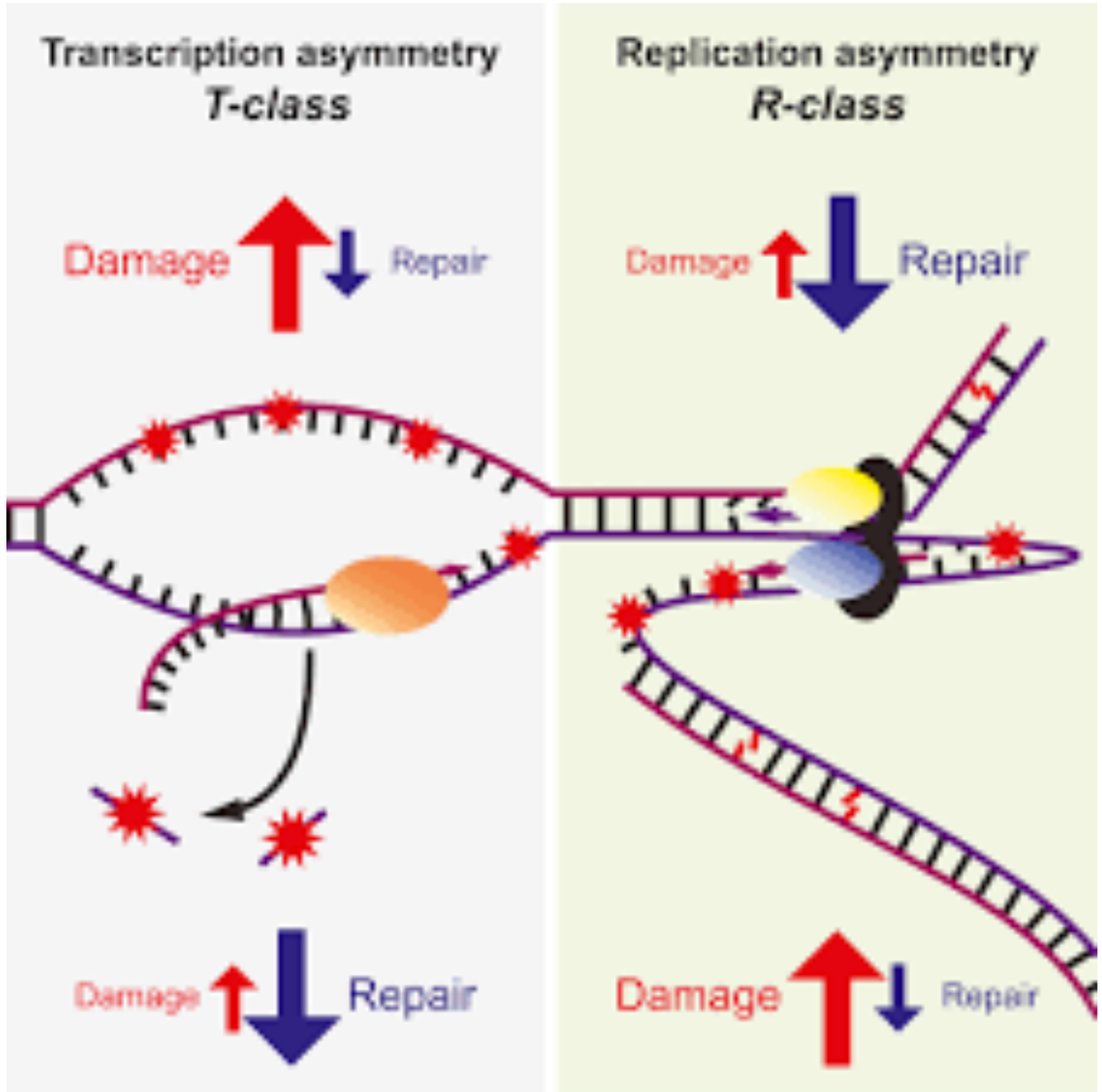


Koren, et al. 2012



Francioli, et al. 2015

Replication and transcription induce strand asymmetry

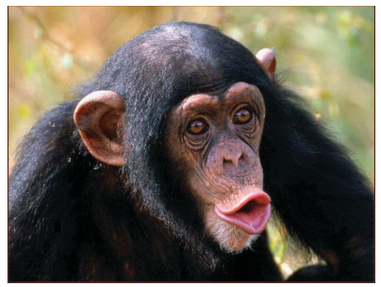


Excess of G+T over A+C on coding strand of most genes

Haradhvala, et al. 2016
Green, et al. 2003

Measuring the human mutation rate

Human



Chimpanzee

Nachman and Crowell 2001

2.5*e*-8 mutations per site per gen

mgr.com.my

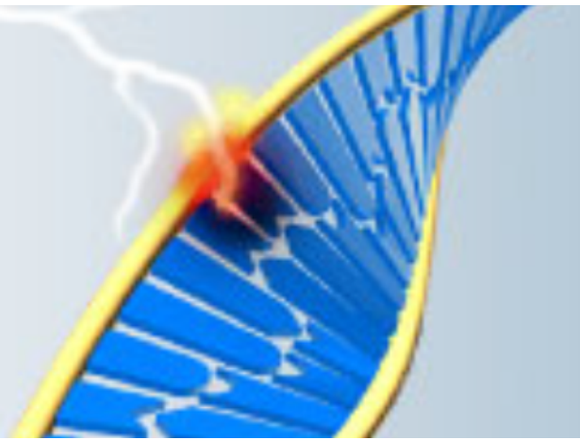
Parent-child trios

1000 Genomes Consortium 2010

1.0*e*-8 mutations per site per gen

The Human Mutation Rate Meeting

Leipzig, 25th - 27th February 2015



NATURE | NEWS

DNA mutation clock proves tough to set

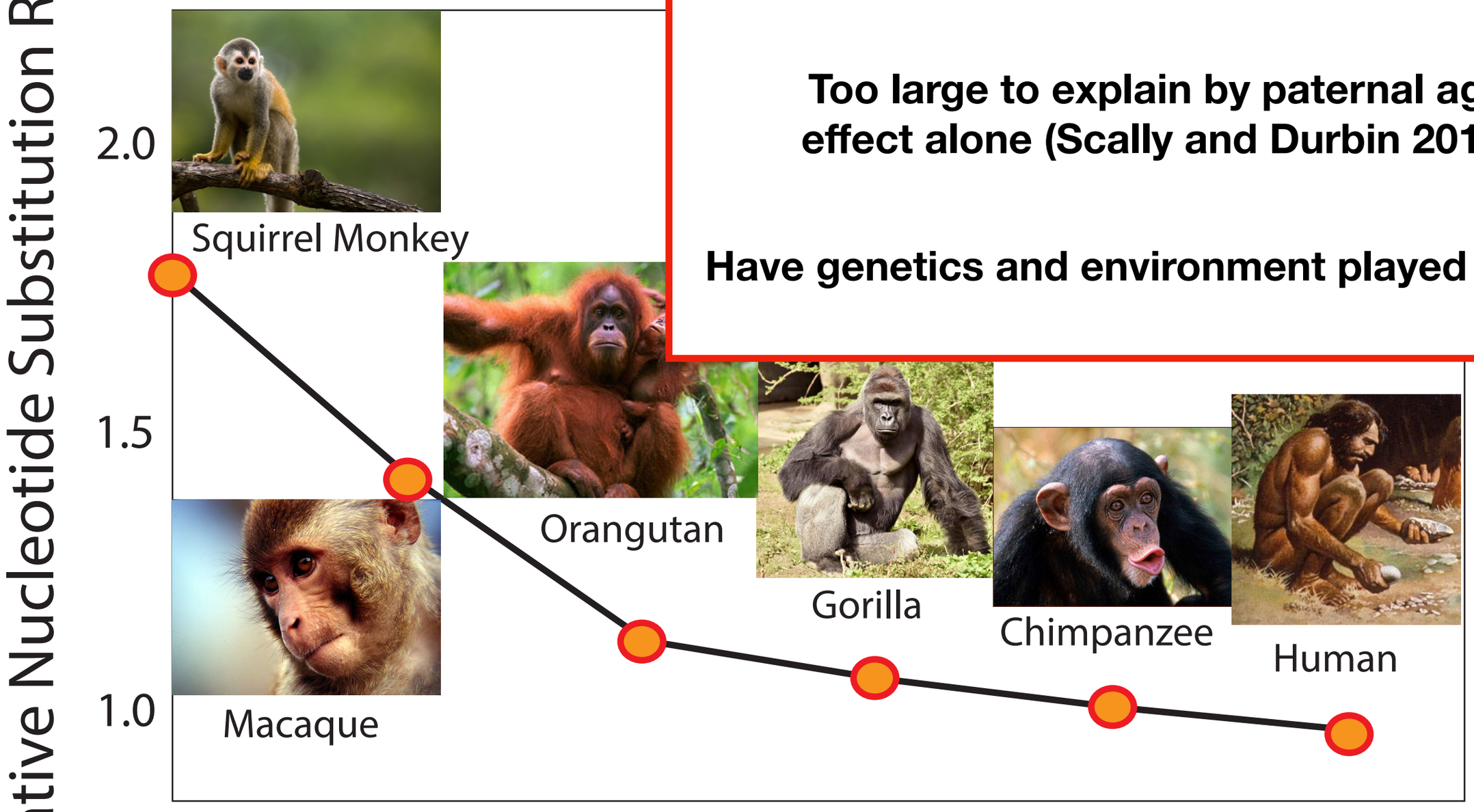
Geneticists meet to work out why the rate of change in the genome is so hard to pin down.

Ewen Callaway

10 March 2015

- What is the real human mutation rate?
- Has the mutation rate slowed down during recent human history?

The Hominoid Mutation Rate Slowdown



Too large to explain by paternal age effect alone (Scally and Durbin 2012)

Have genetics and environment played a role?

Adapted from http://www.bio.indiana.edu/graduate/multidisciplinary/GCMS/trainees/thomas_gregg.php

Goodman *BioEssays* 1985 Moorjani, et al. *PNAS 2016*

“The” mutation rate encompasses a menagerie of mutation types

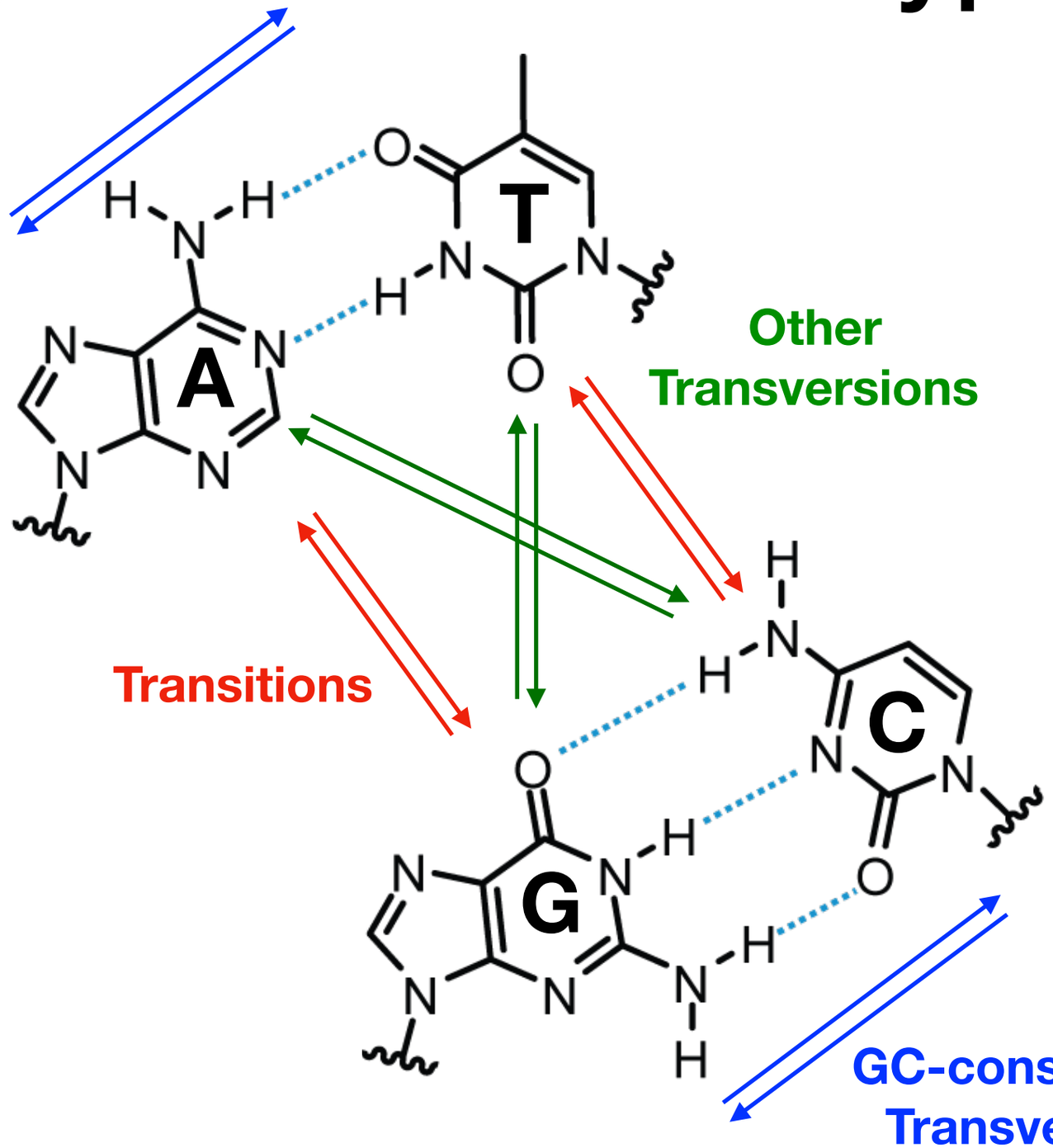
Point Mutations

Multinucleotide Mutations

CC ⟶ TT

Small indels

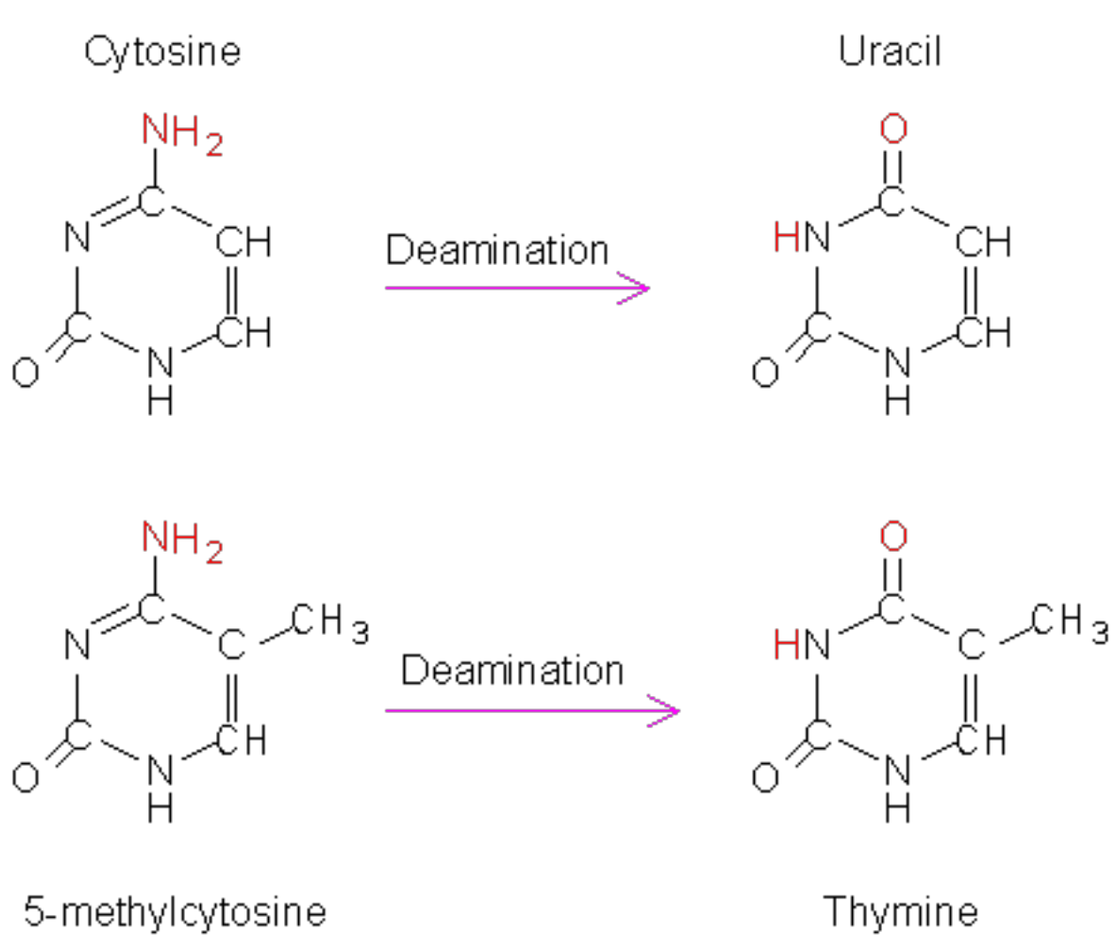
Large Copy Number Changes



CpG Mutations

- Many species (incl humans, not incl *Drosophila*) methylate C when it's next to G (C-phosphate-G)

- CpG methylation regulates gene expression



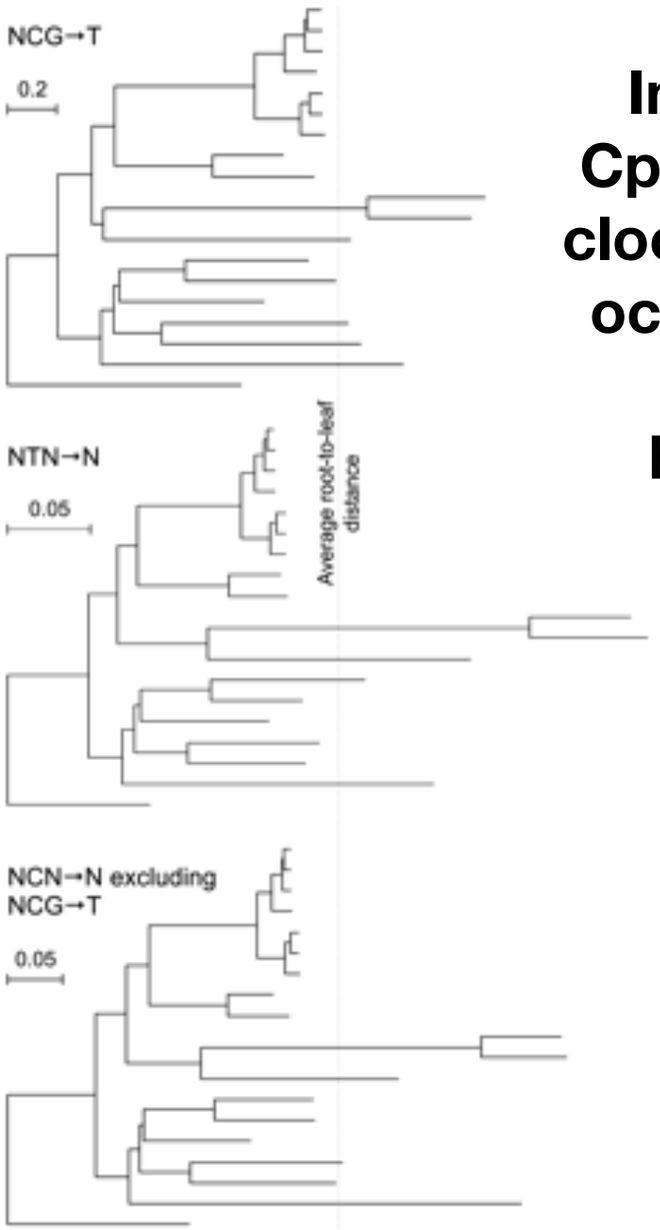
CpG sites are hypermutable

- On average, CpG sites have a 30-fold higher mutation rate than other C's in the human genome

- 70-80% of CpGs are methylated in mammals; most unmethylated CpGs are part of CpG islands

- Fewer than 1% of dinucleotides in the human genome are CpGs, although the expected frequency is 0.21*0.21=4.41%

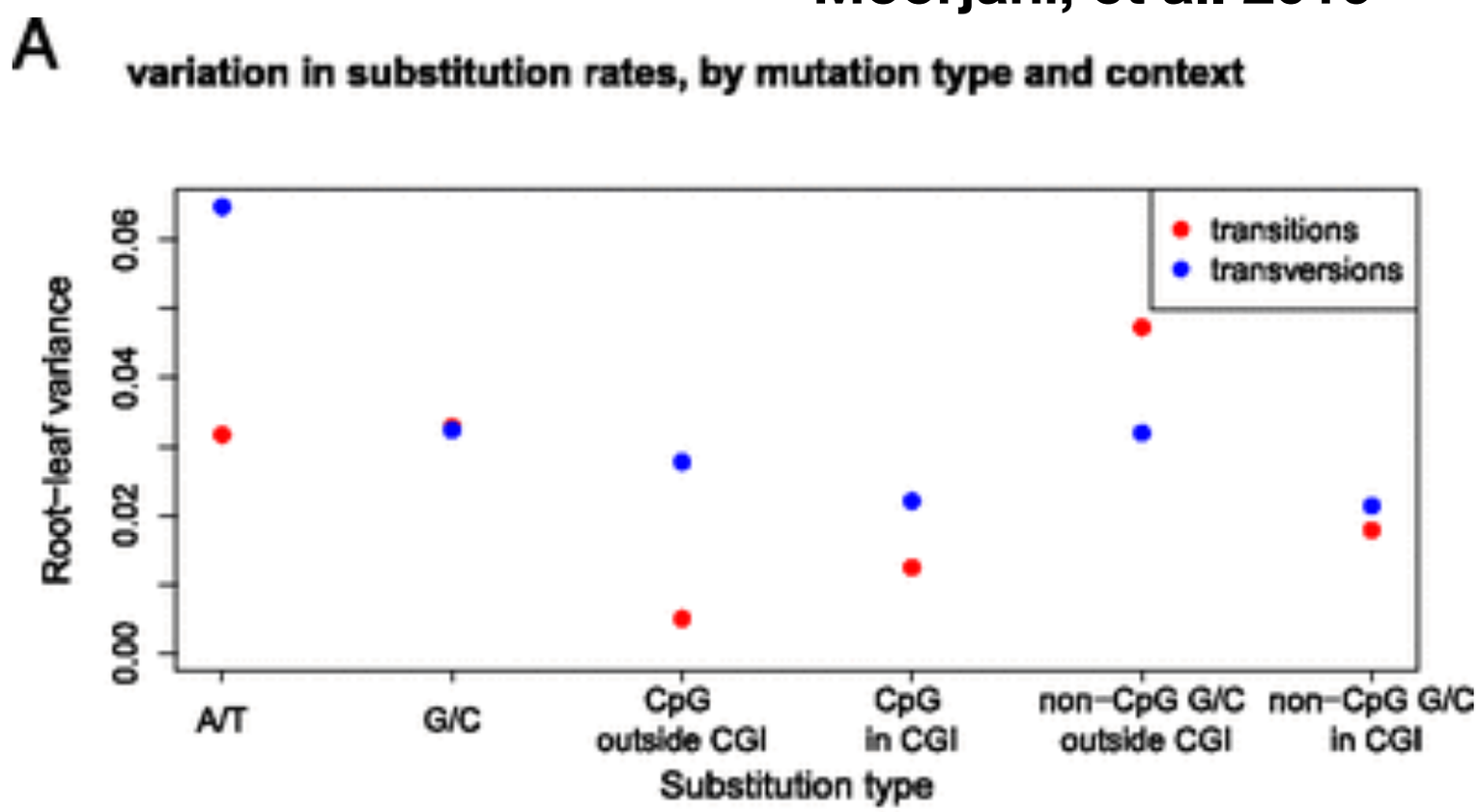
CpG transitions are somewhat more clocklike than other mutations



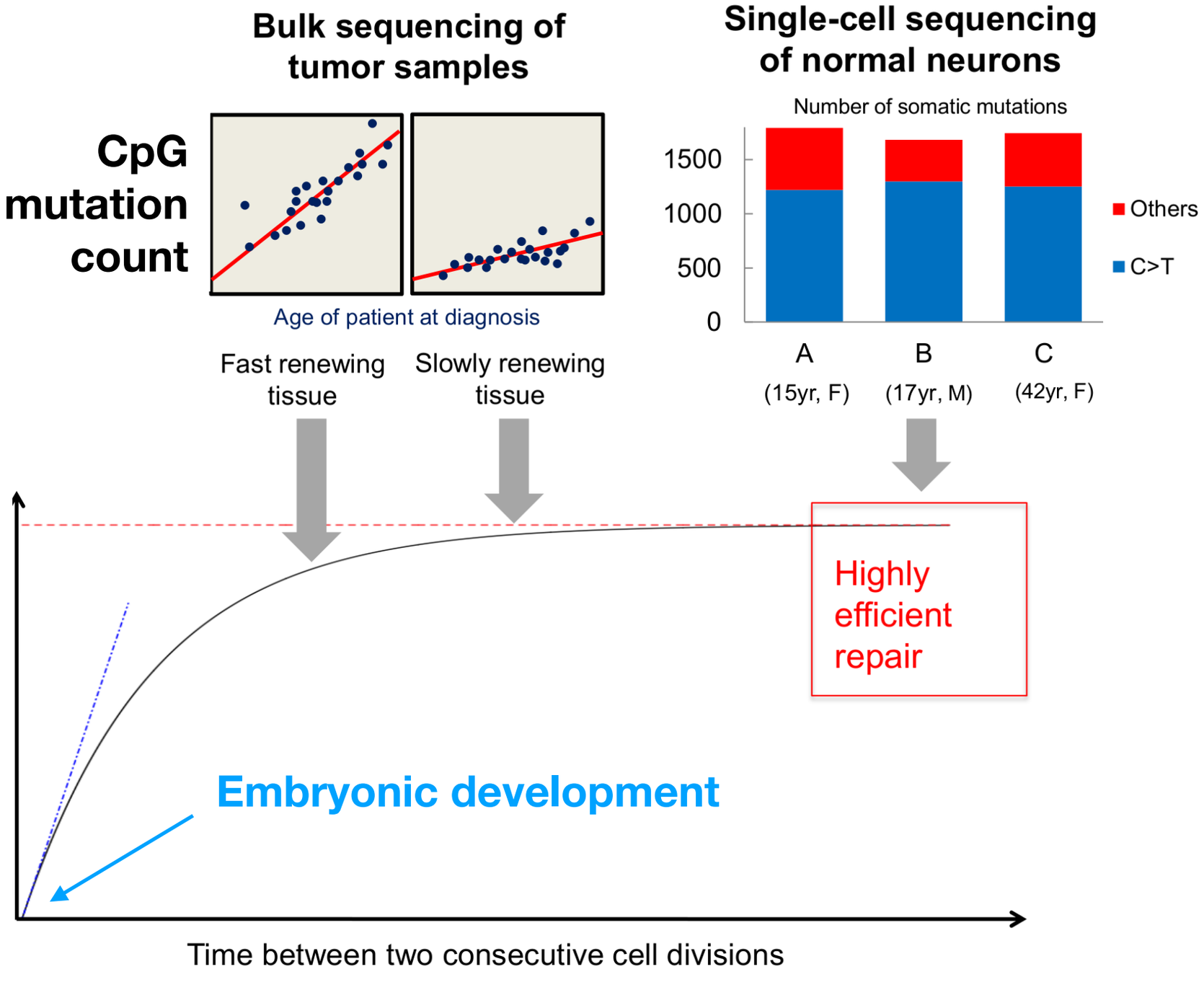
In a tree of 19 mammals, CpG mutations yield a more clocklike tree than mutations occurring in other contexts

Hwang and Green 2004

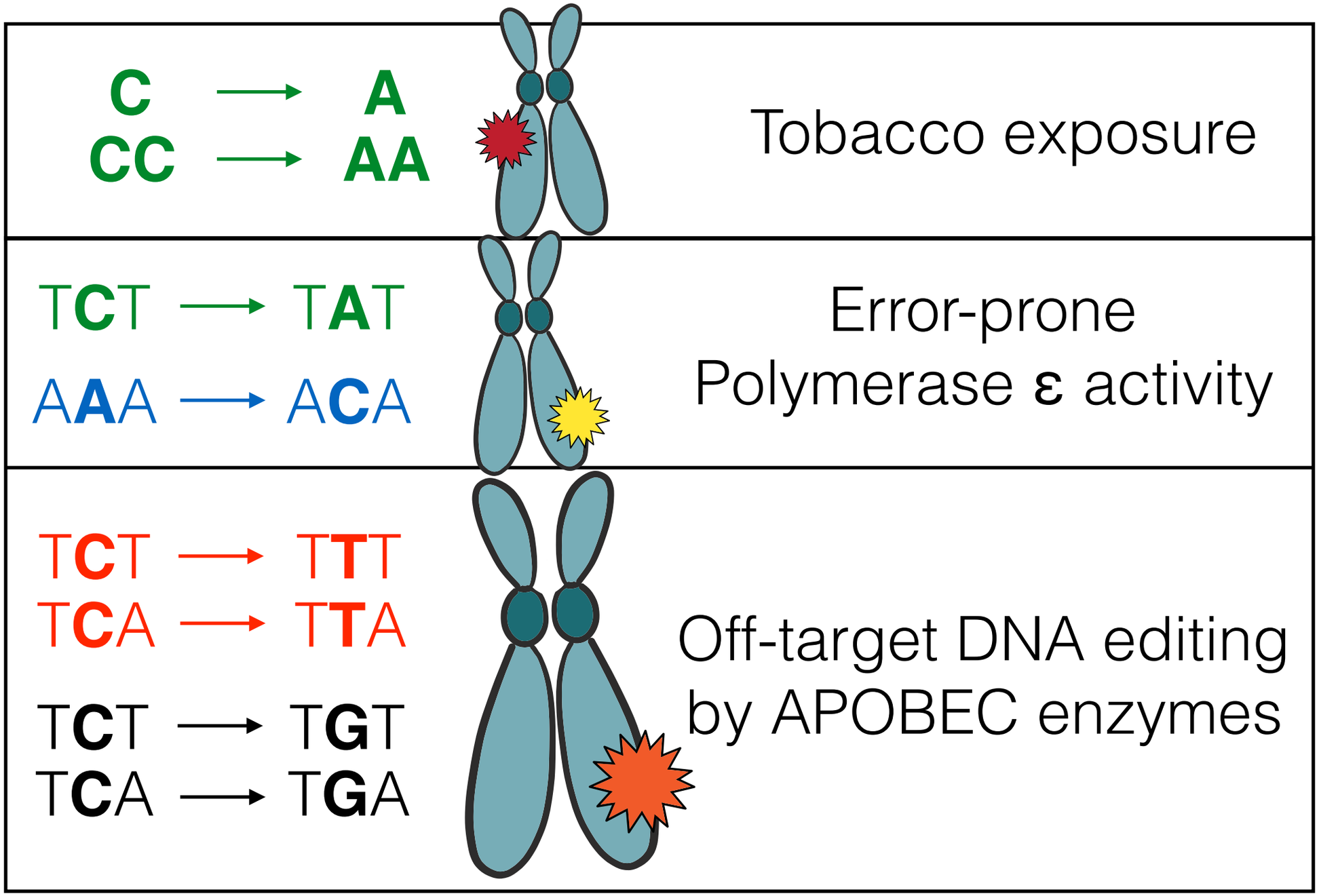
CpG mutations also appear more clocklike than other mutations in great ape tree
Moorjani, et al. 2016



Limits to clock-like behavior of CpGs



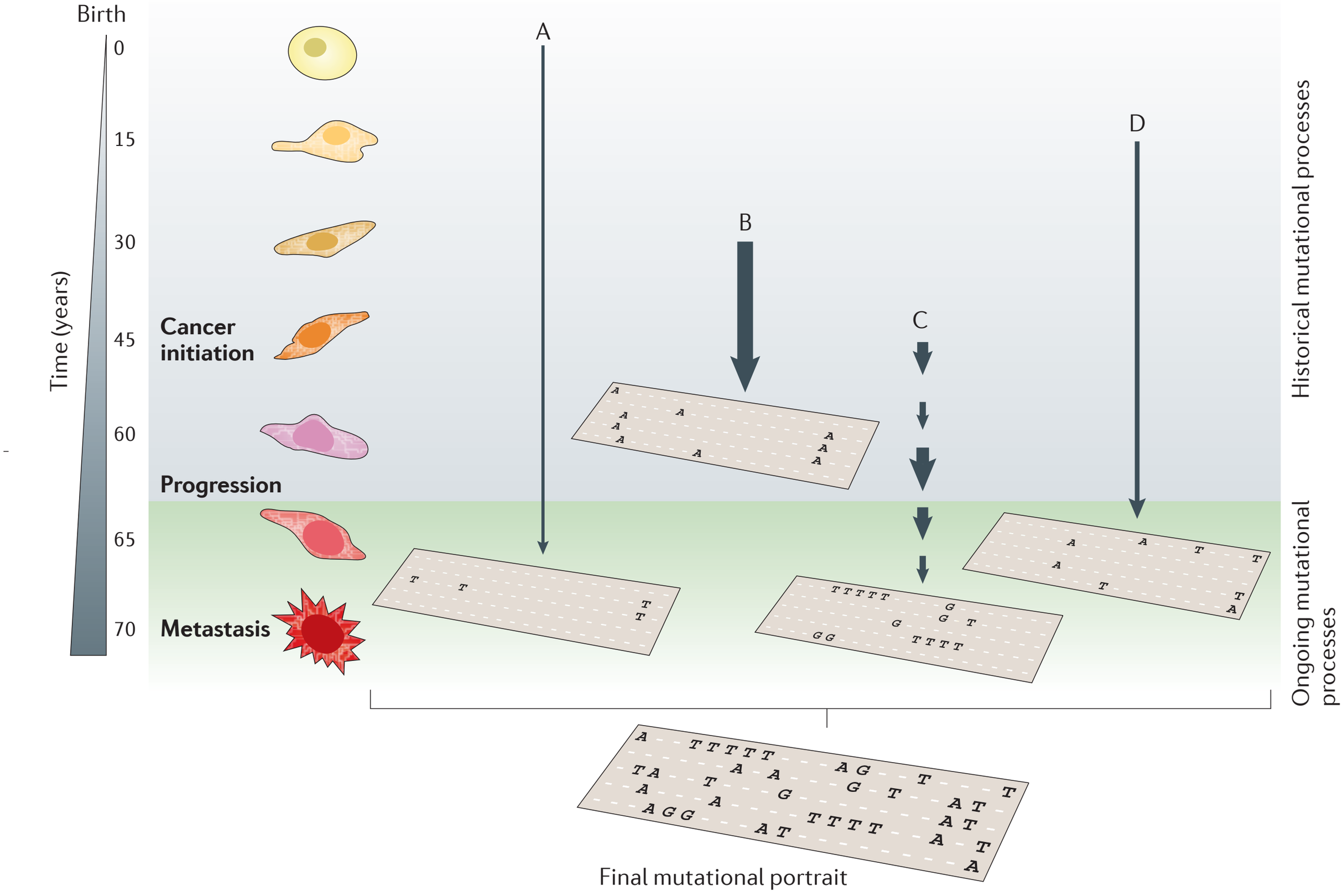
“Mutational signatures” of types of DNA damage in cancer



Alexandrov, *et al. Nature* 2013

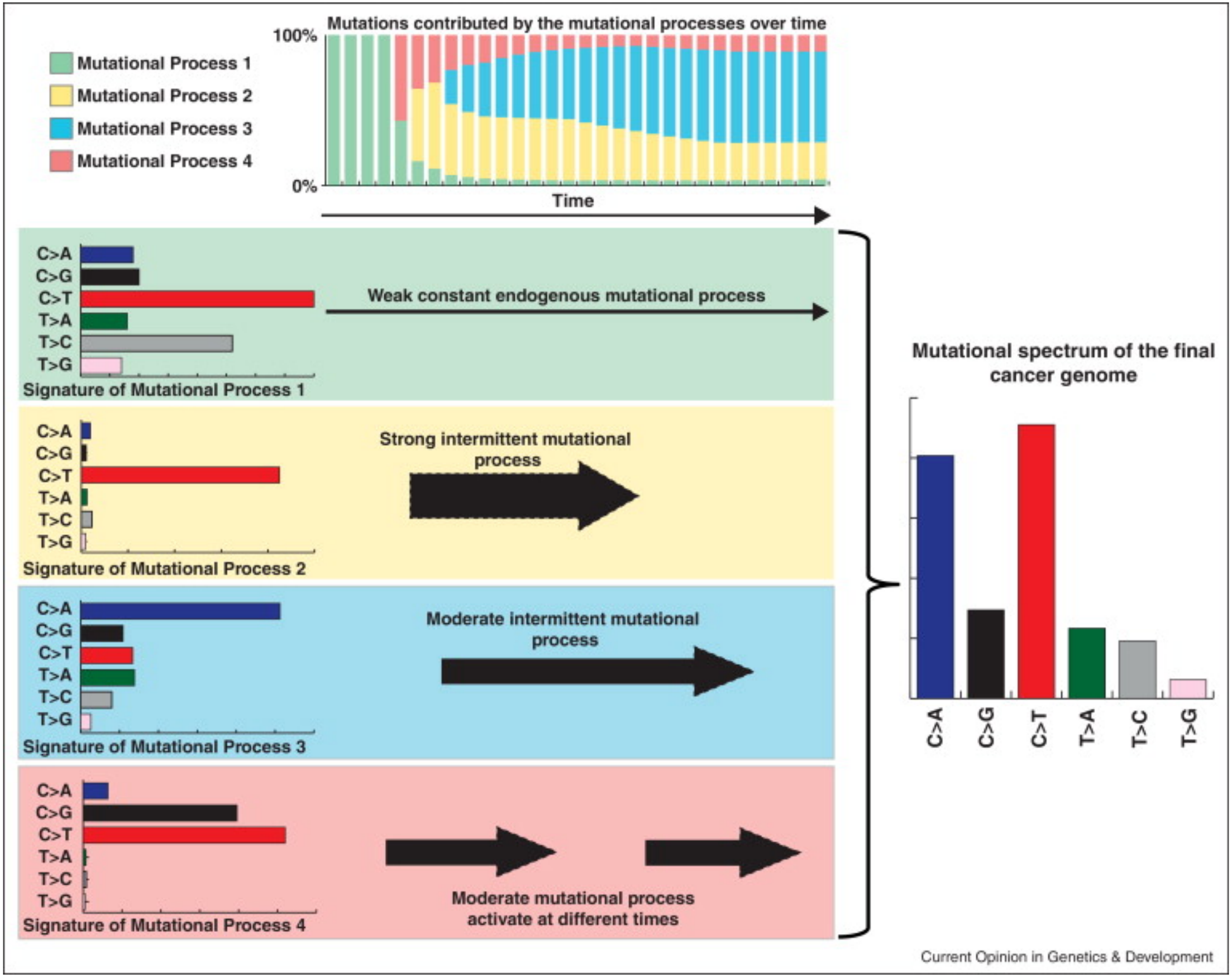
APOBEC / AID deaminases

- APOBEC attacks RNA viruses, mutating TCA and TCT by deamination
- Its homologue AID hypermutates T cell receptors for proper immune function
- Both cause off-target germline mutations, especially in endogenous retroviral sequences
- APOBEC is erroneously switched on in many cancers (esp cervical), associated with poorer outcomes



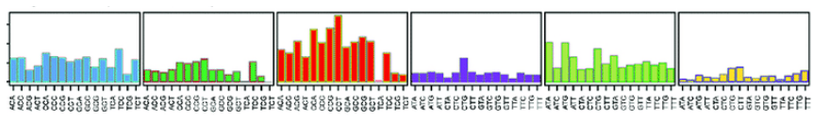
Helleday, *et al. Nature Review Genetics* 2014

Alexandrov, *et al. Nature* 2013



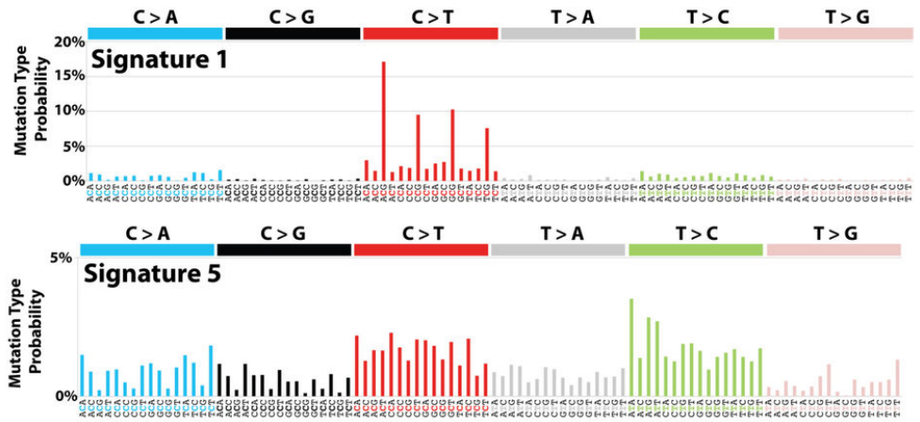
Alexandrov and Stratton 2014

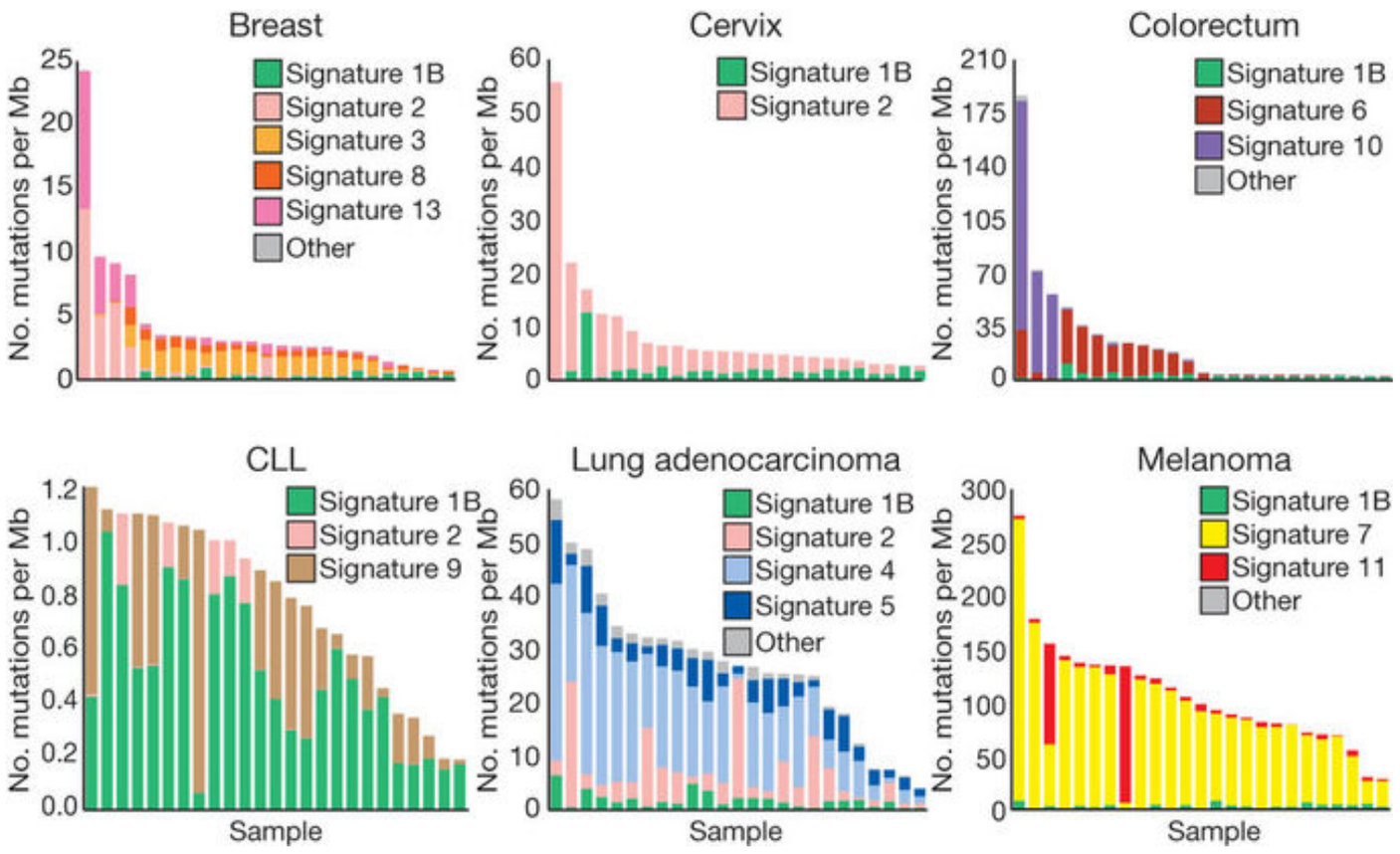
Mutation signature analysis



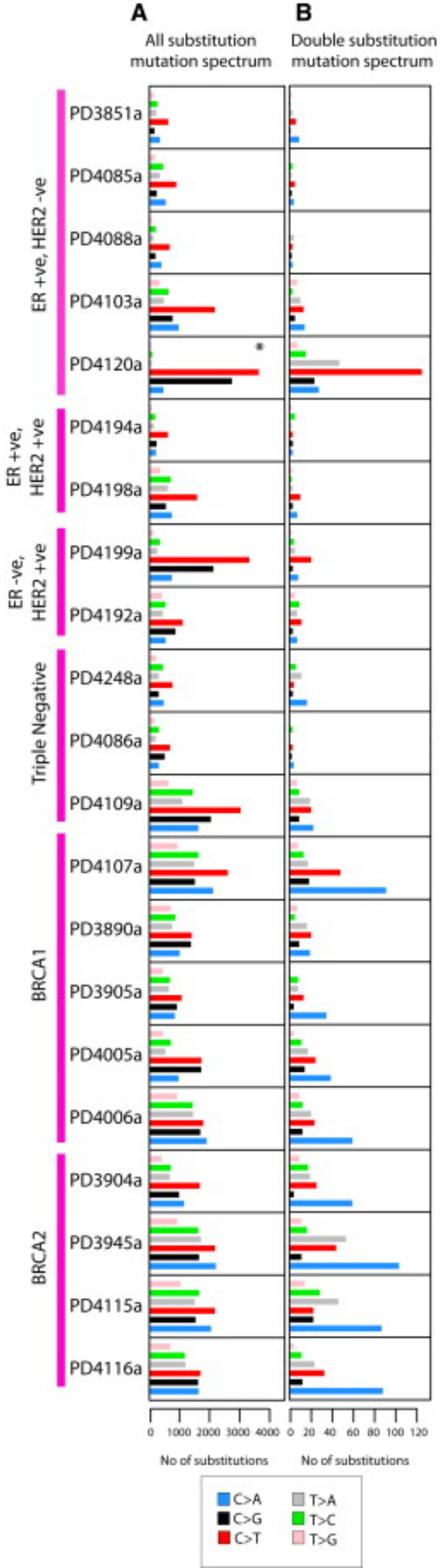
Mutation counts in 96 triplet contexts across cancers

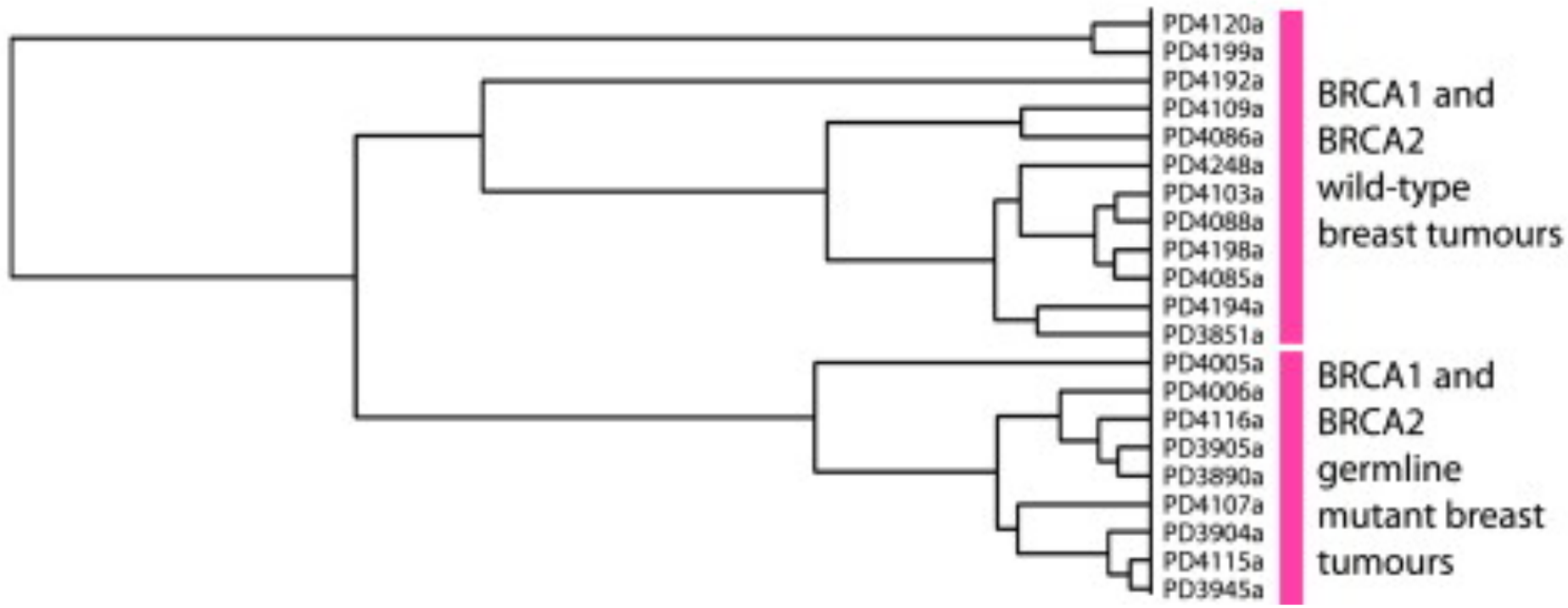
Nonnegative matrix factorization





Effect of BRCA germline mutations on breast cancer mutation distribution





Mutational signatures in the germline?

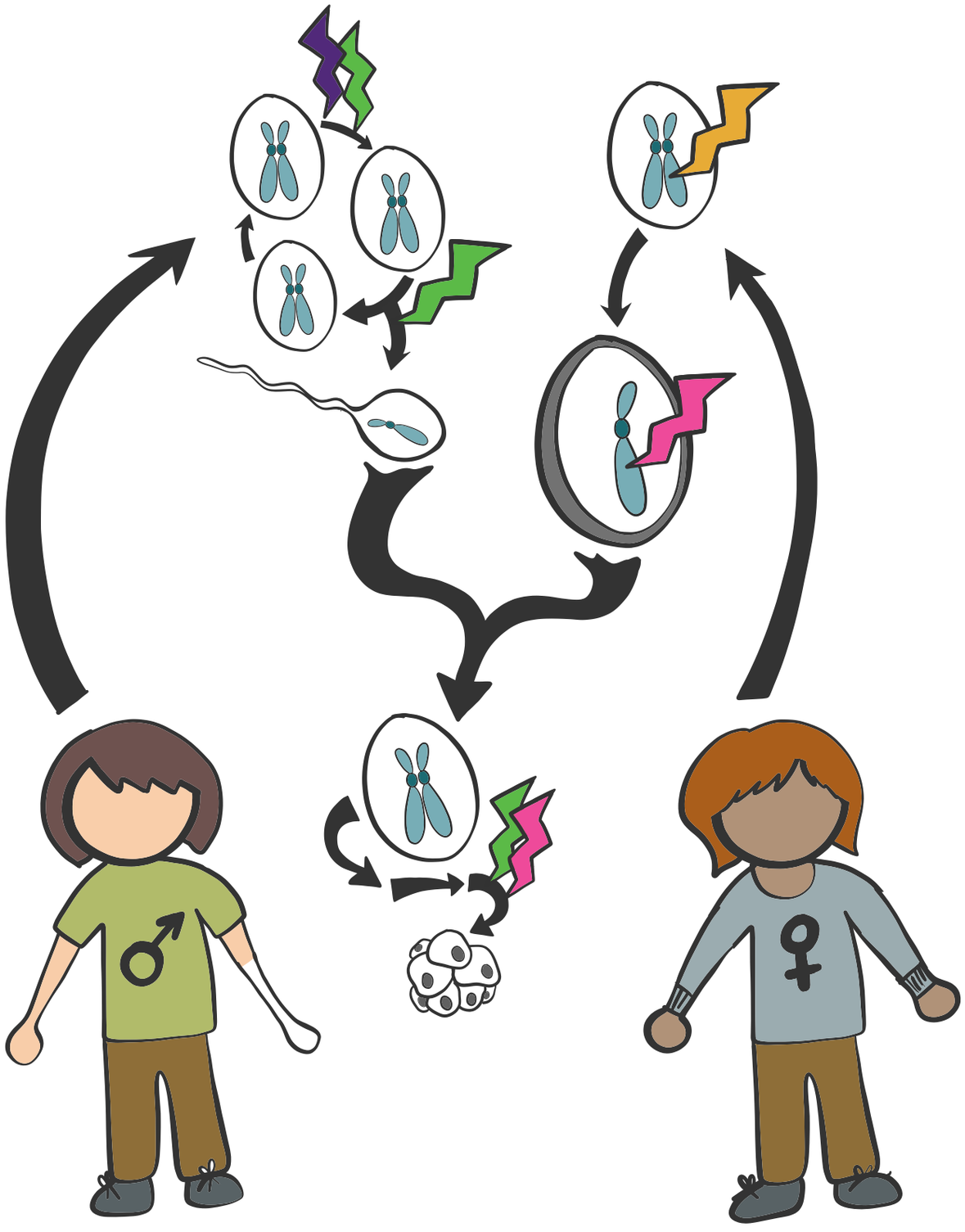
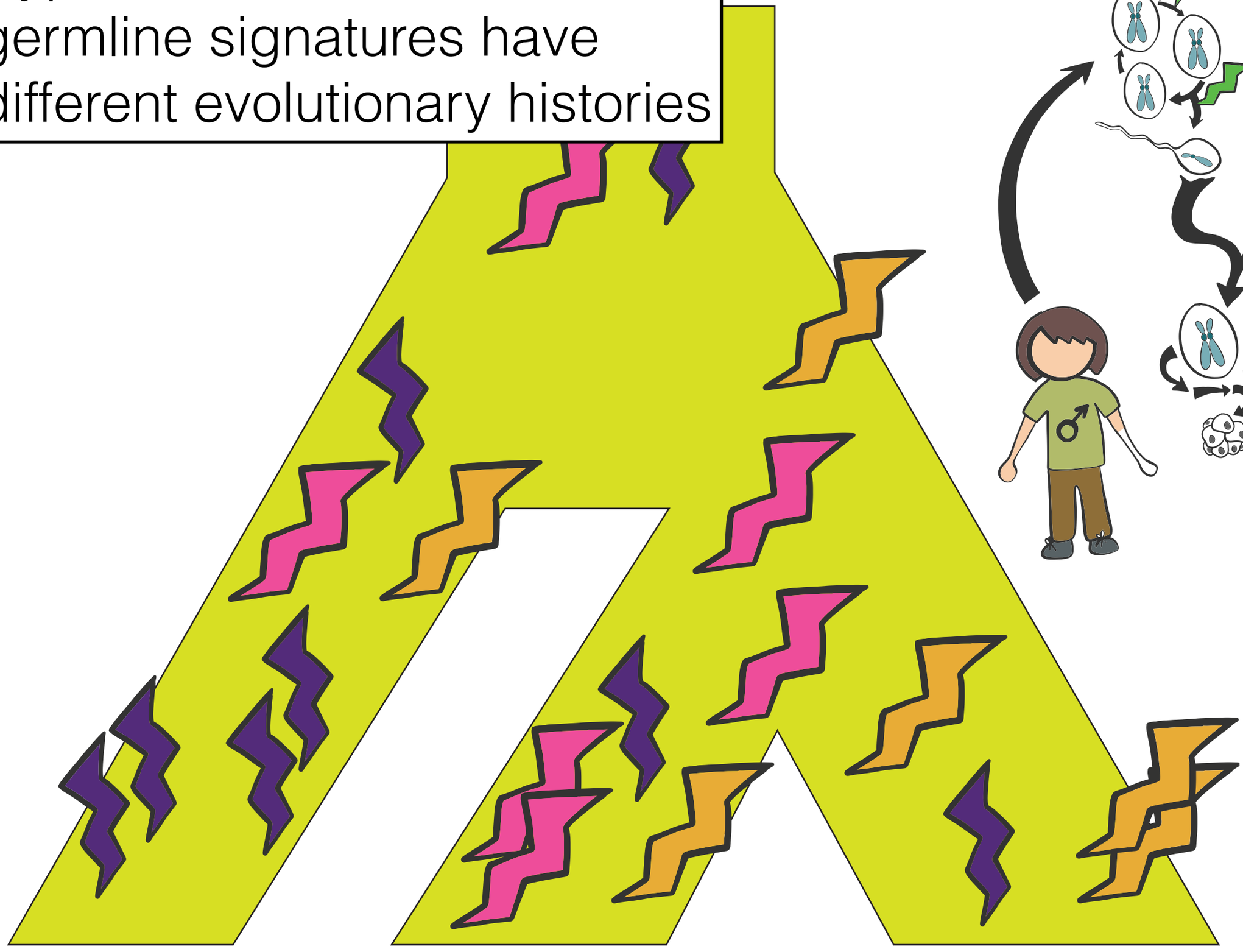
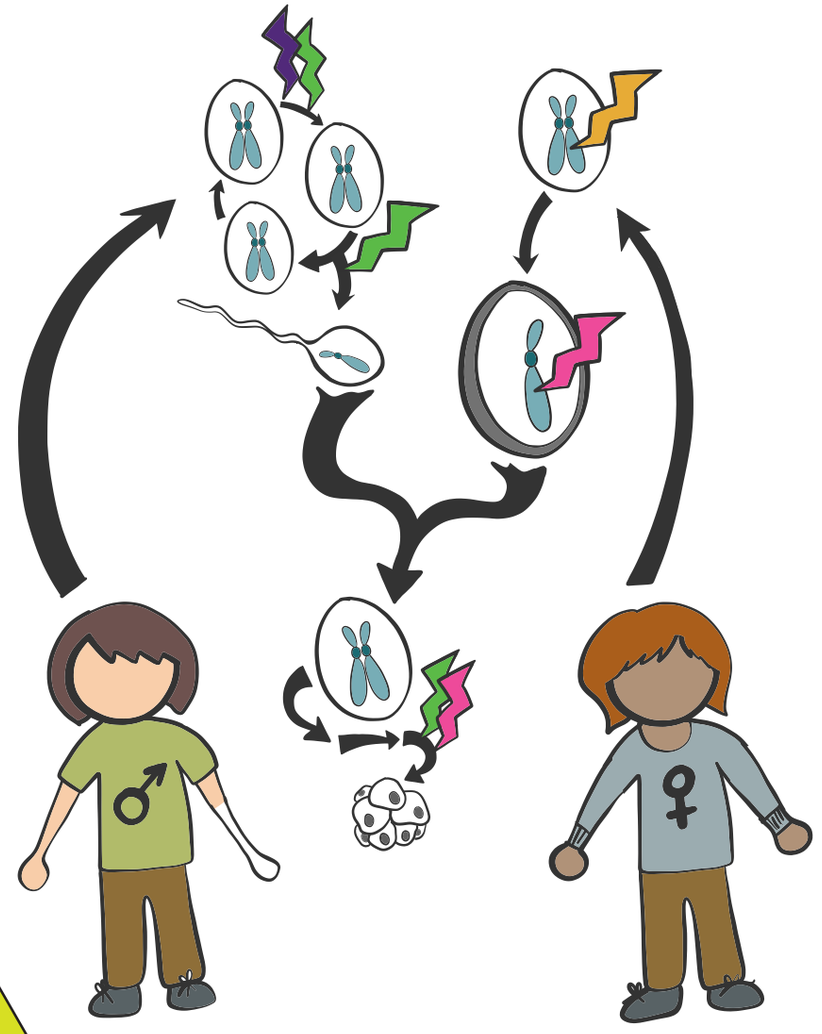


Image co-artist: Natalie Telis

Hypothesis: different germline signatures have different evolutionary histories



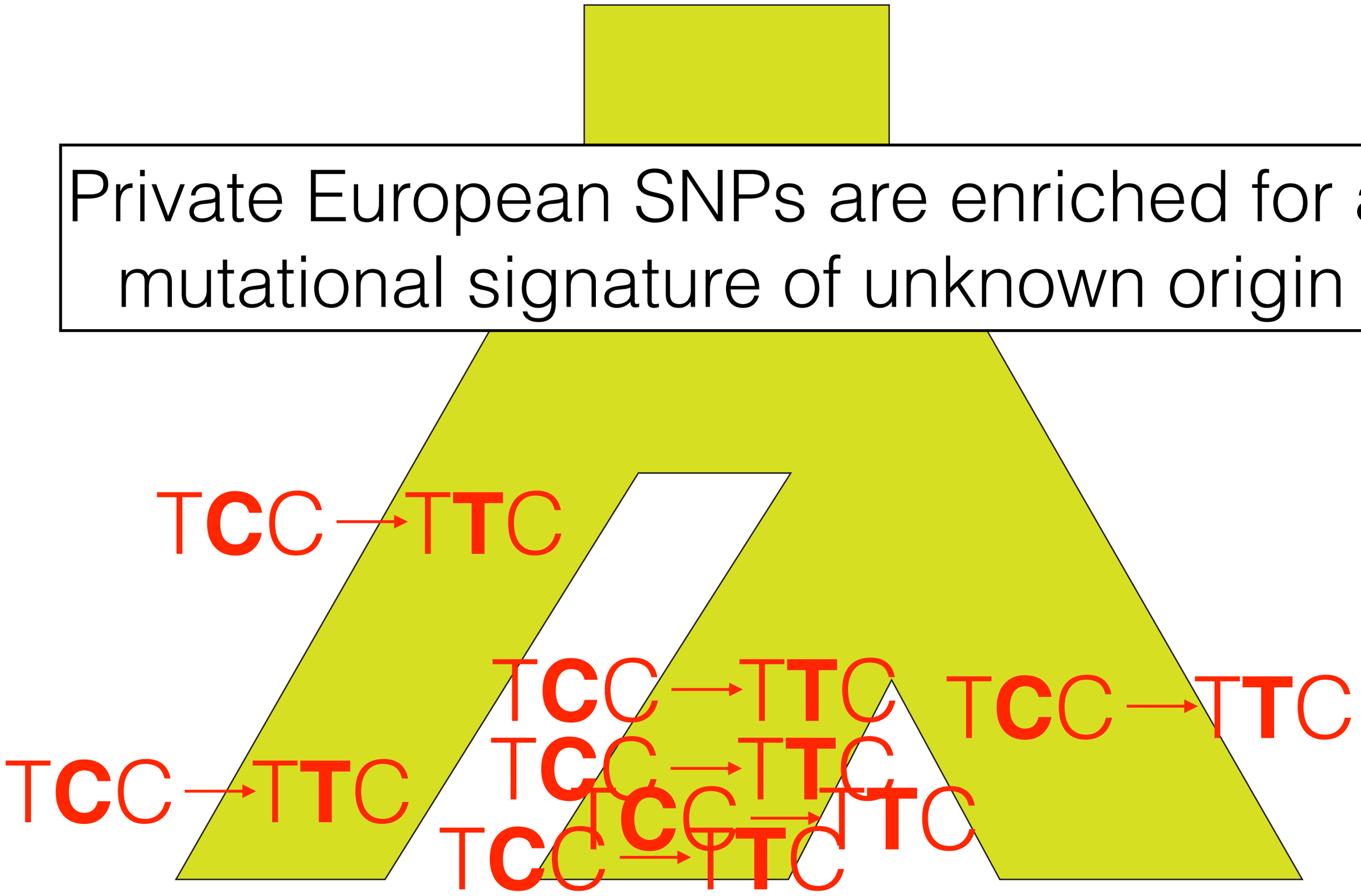


Africans

Europeans

East Asians

Private European SNPs are enriched for a mutational signature of unknown origin



Africans Europeans East Asians

Harris *PNAS* 2015

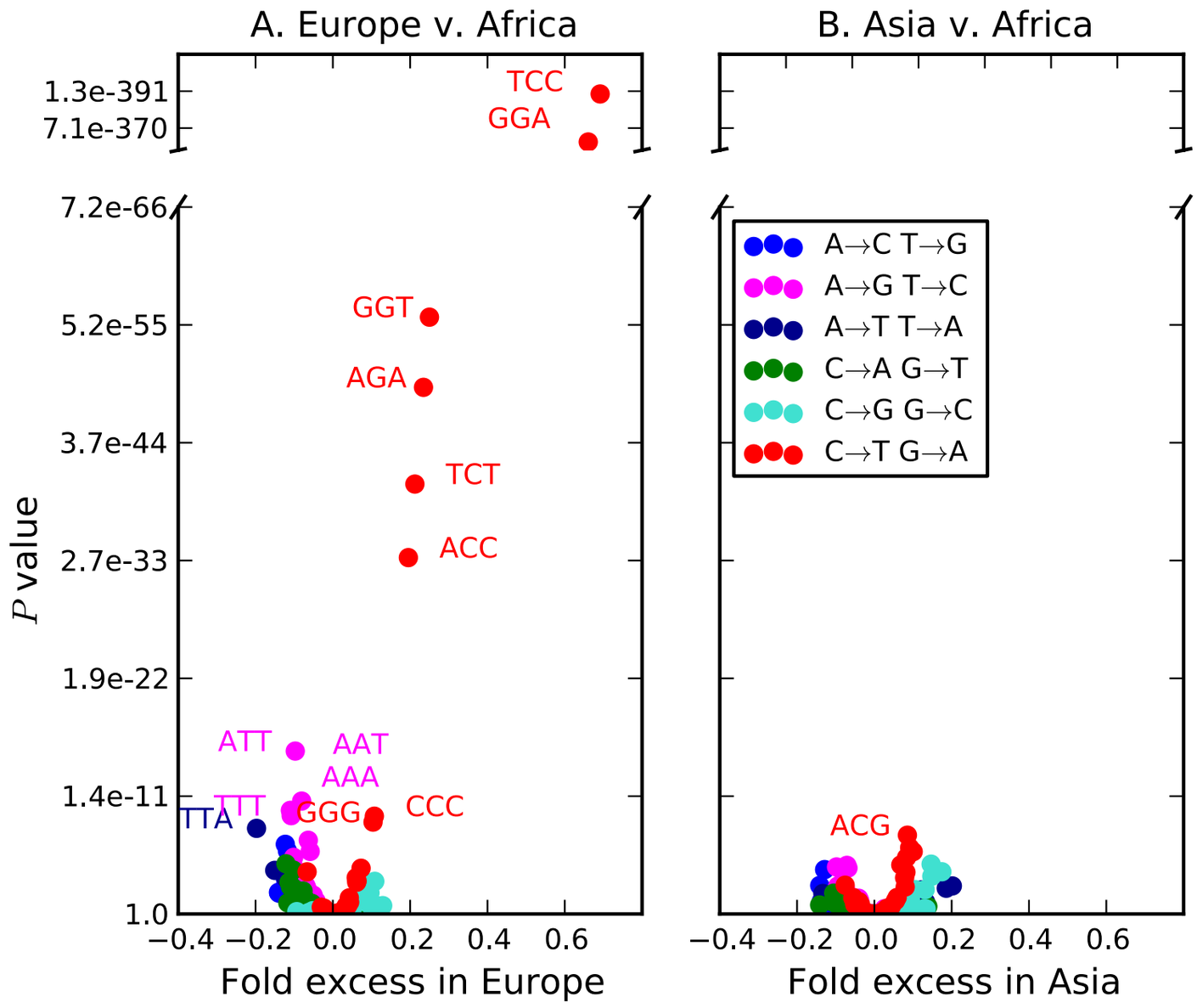
A signature of elevated mutagenesis in the European germline

T**C**C → T**T**C

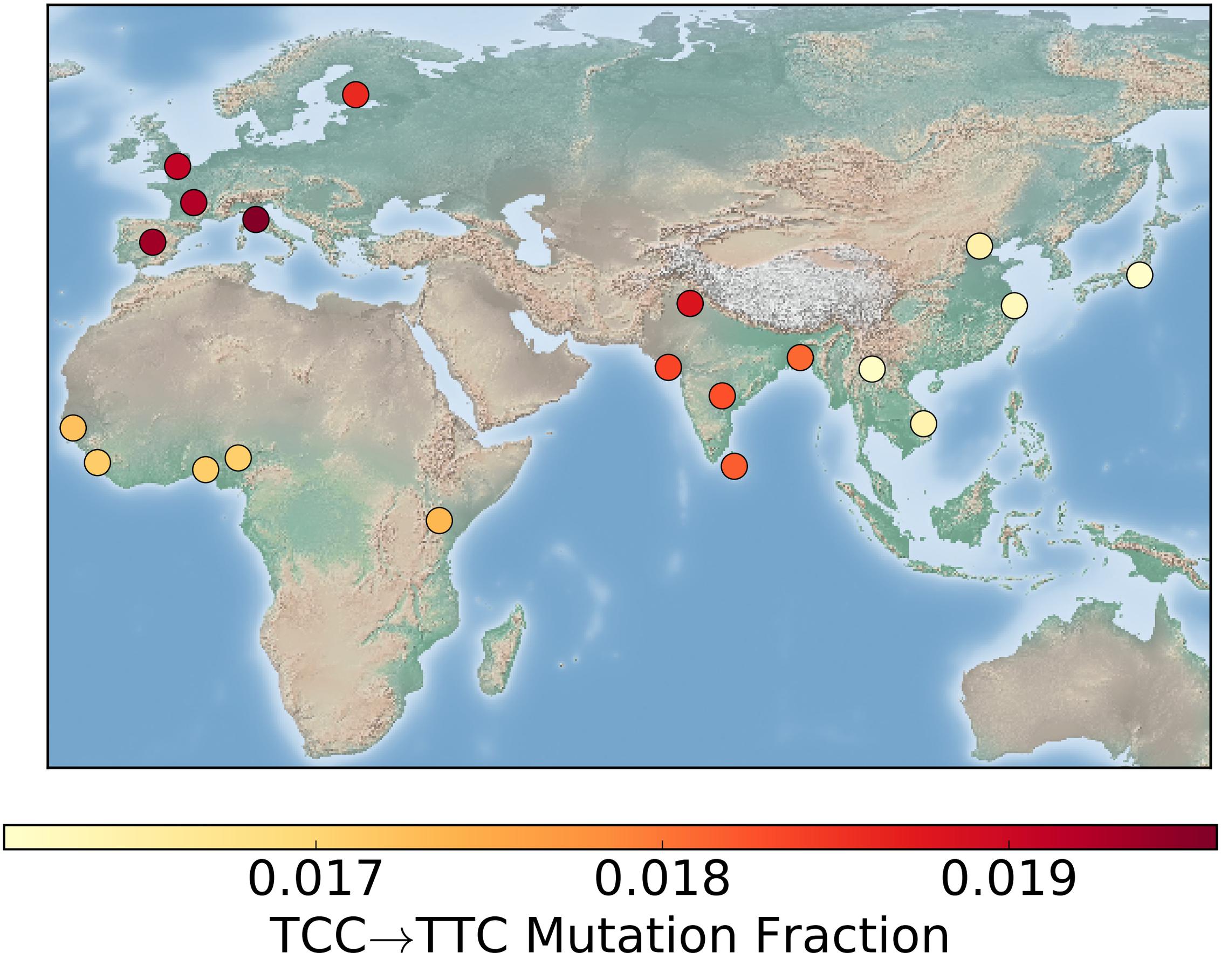
T**C**T → T**T**T

C**C**C → C**T**C

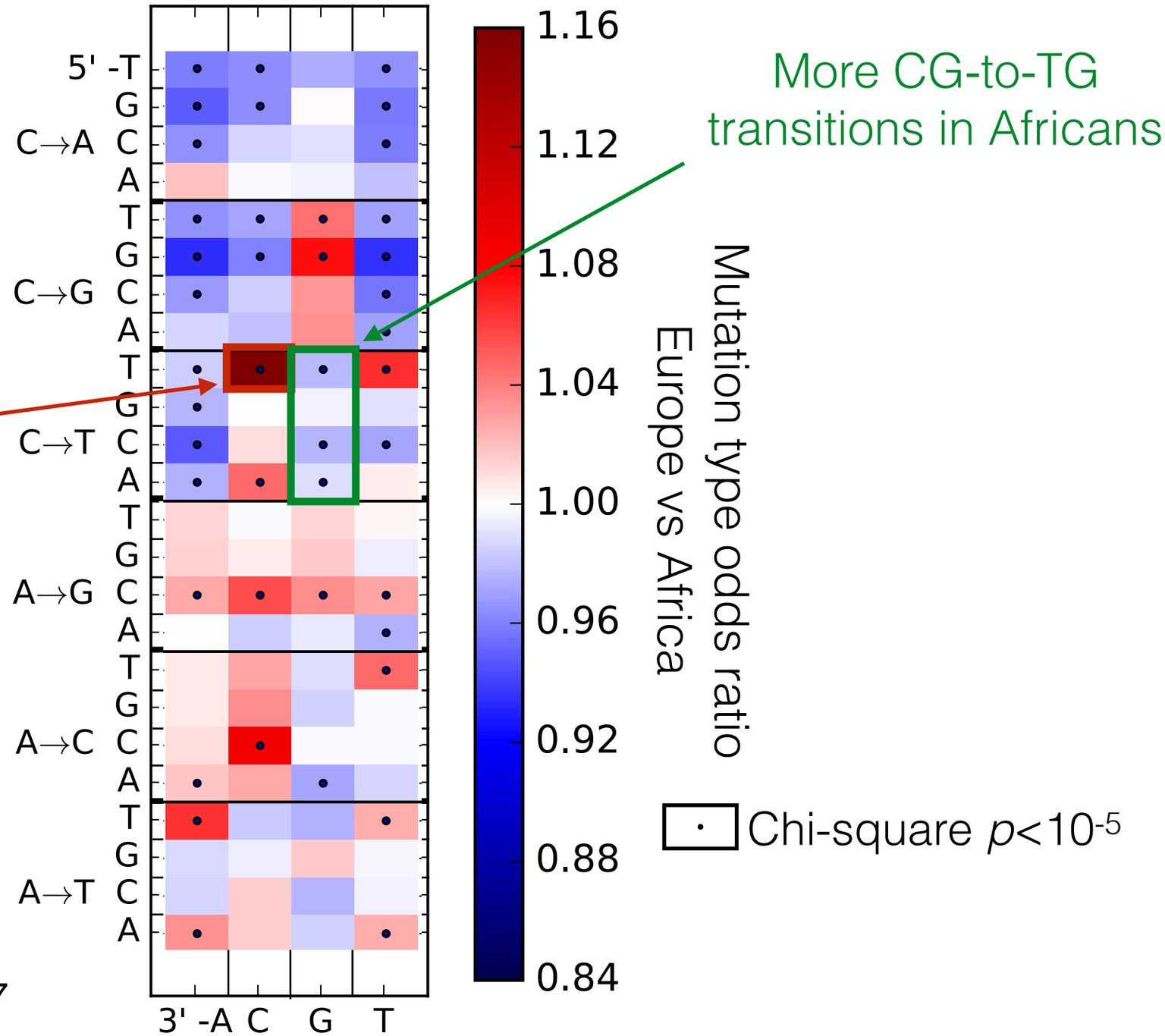
A**C**C → A**T**C



Harris *PNAS* 2015

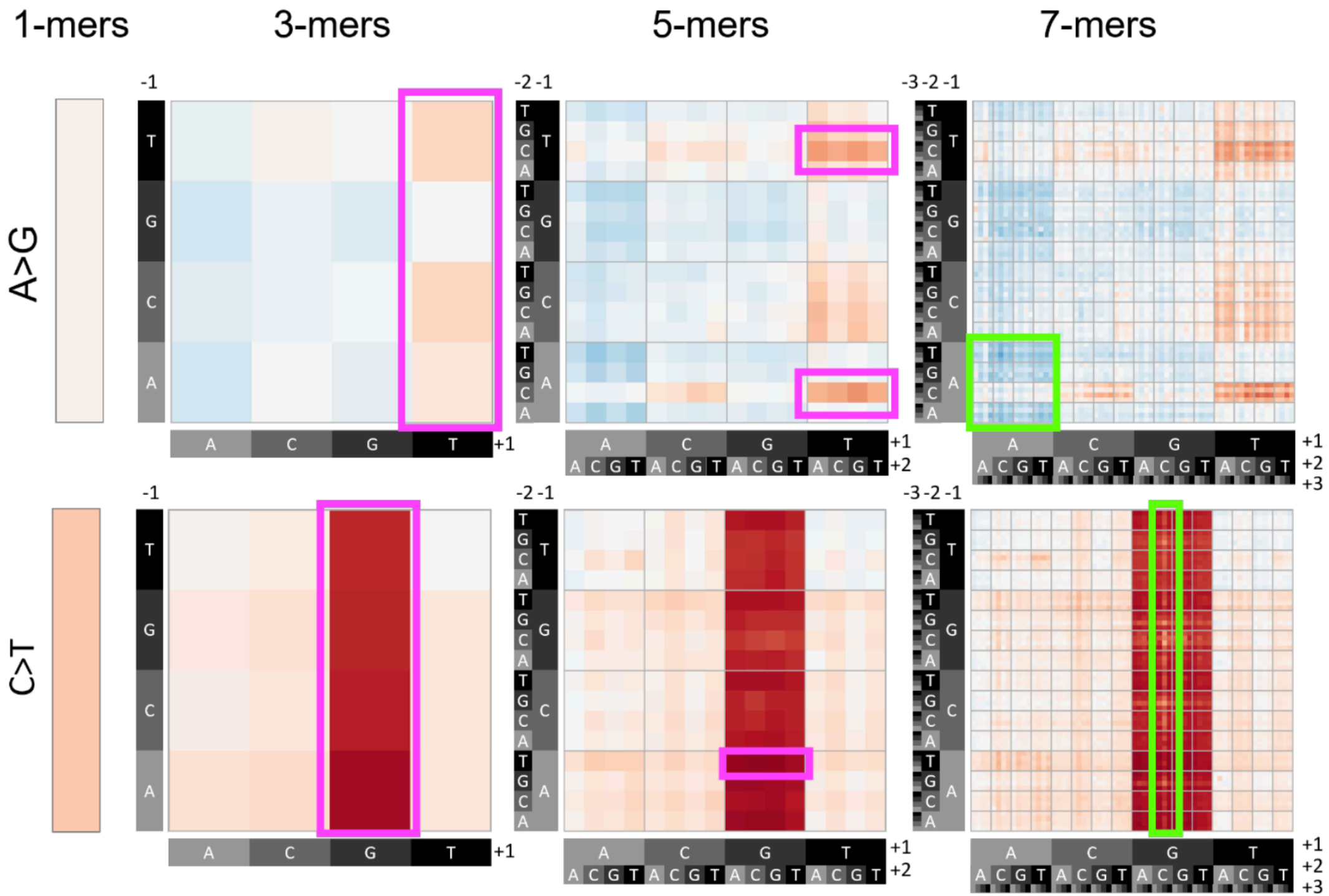

0.017
0.018
0.019
TCC→TTC Mutation Fraction

Visualizing differences between mutation spectra



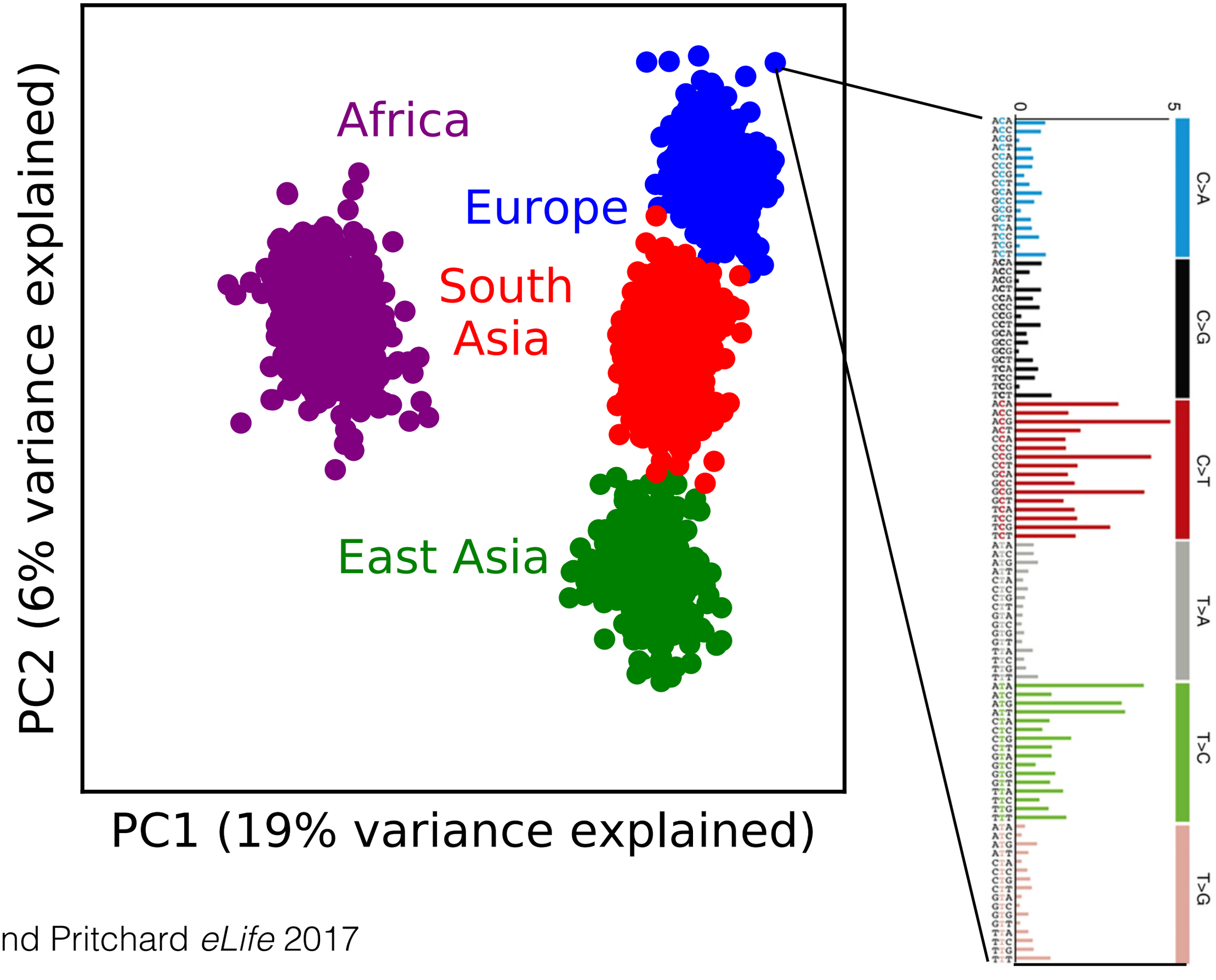
Harris and Pritchard *eLife* 2017

Beyond 3-mers to 7-mers



Aggarwala and Voight 2016

Carlson, et al. 2017



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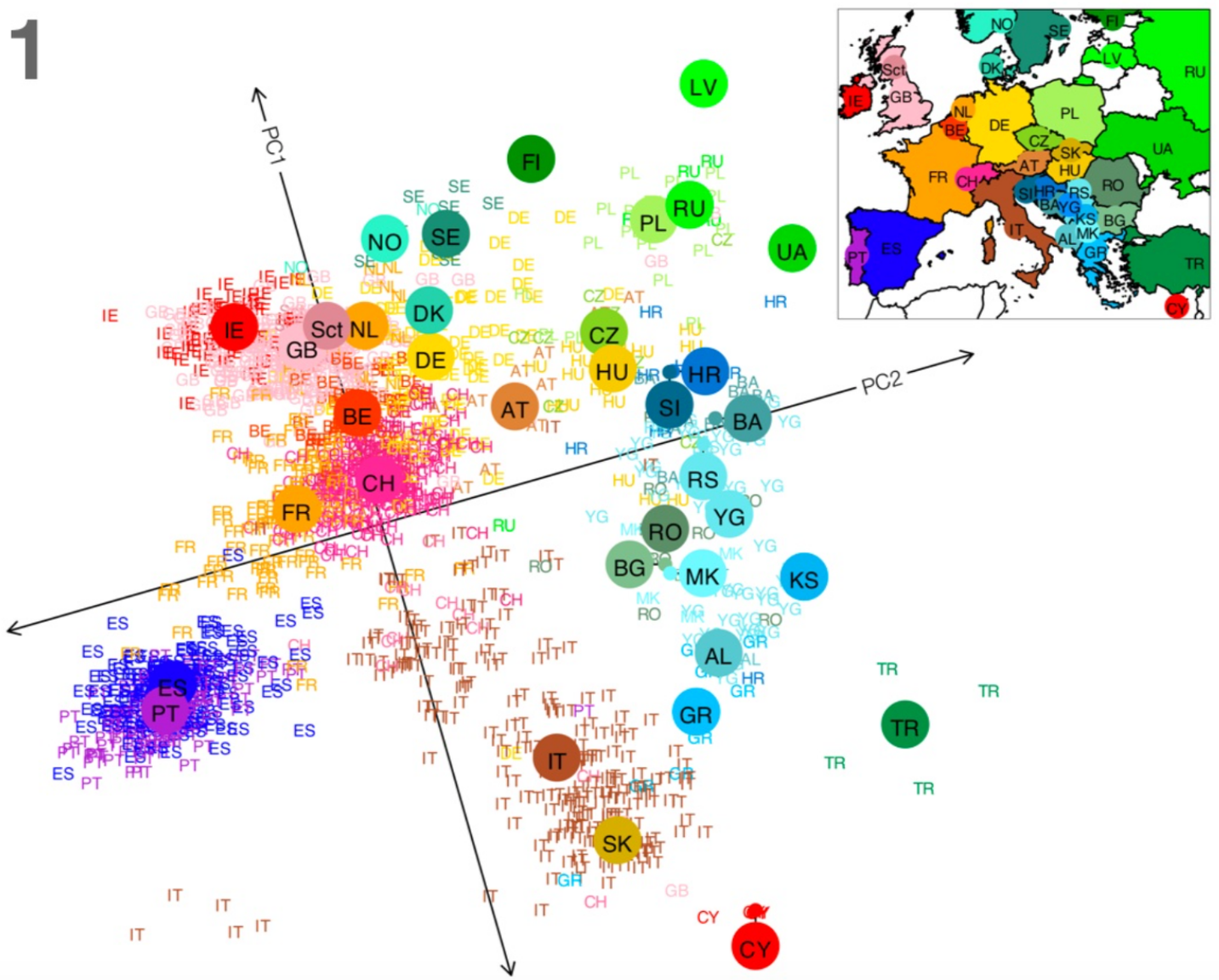
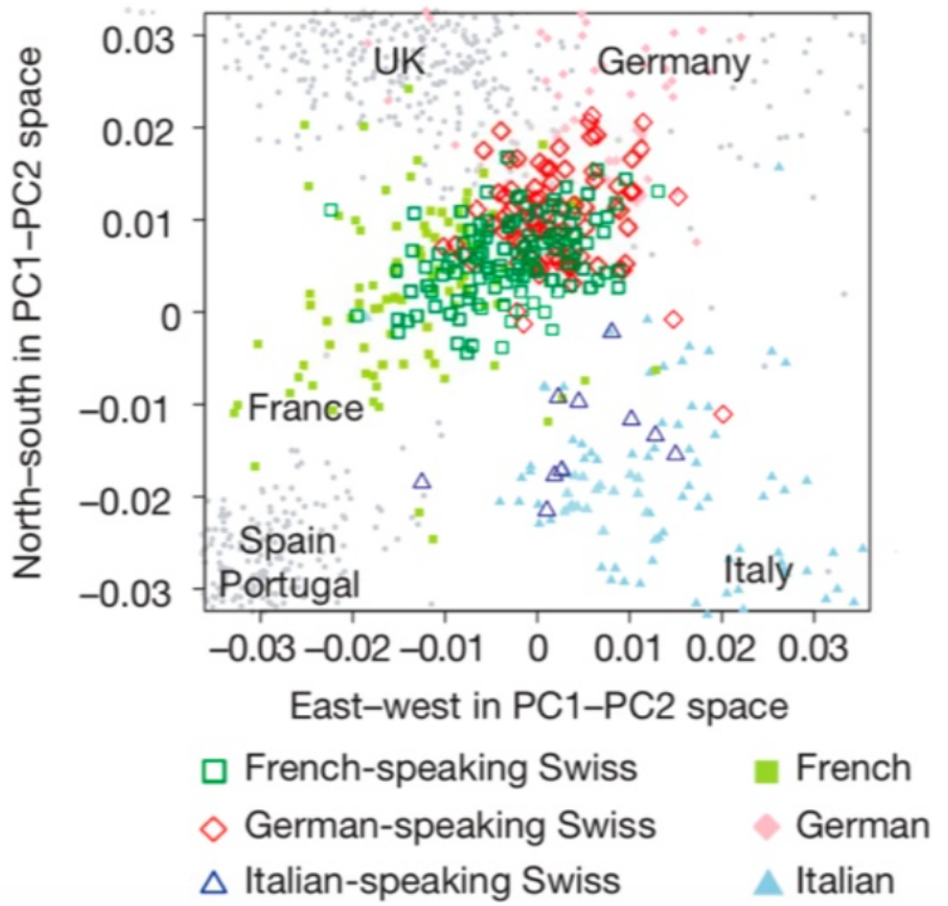
Genes mirror geography within Europe

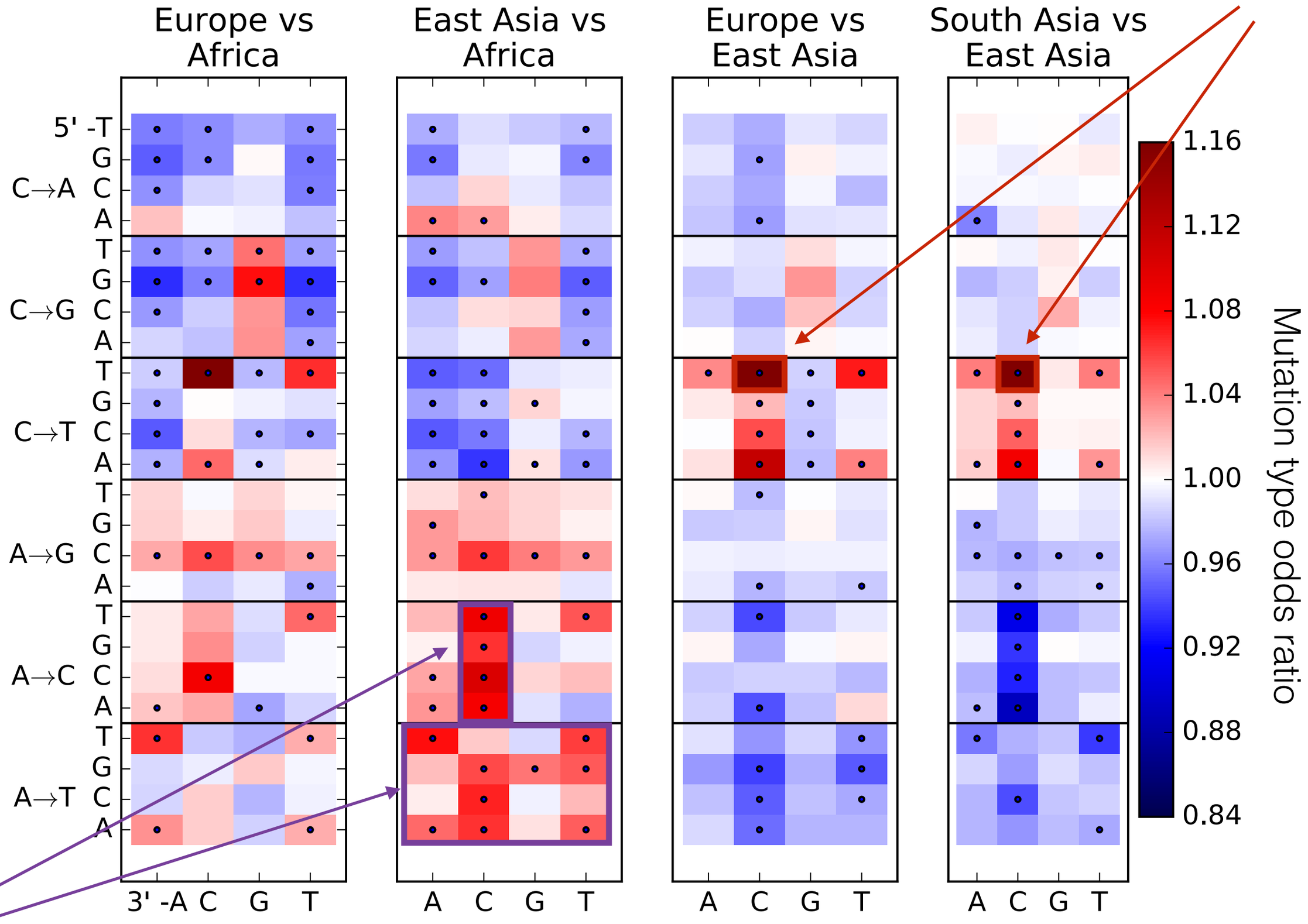
John Novembre[1,2], Toby Johnson[4,5,6], Katarzyna Bryc[7], Zoltán Kutalik[4,6], Adam R. Boyko[7], Adam Auton[7], Amit Indap[7], Karen S. King[8], Sven Bergmann[4,6], Matthew R. Nelson[8], Matthew Stephens[2,3] & Carlos D. Bustamante[7]

1



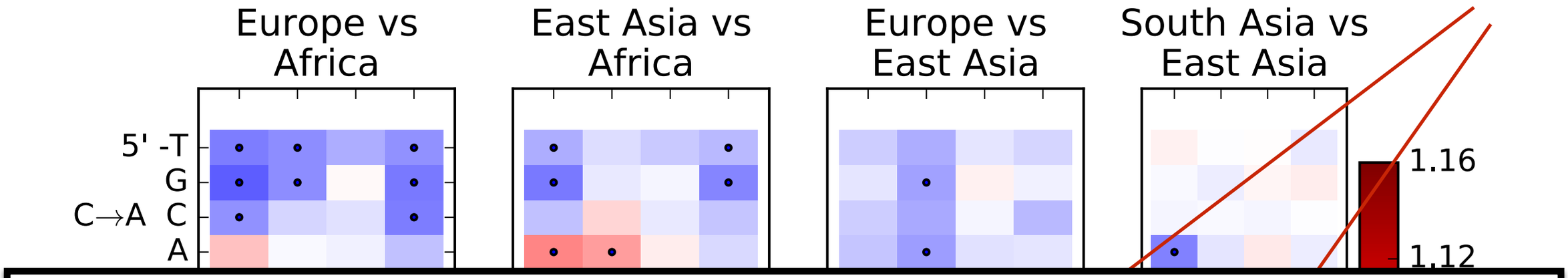


TCC-to-TTC transitions are enriched in South Asia as well as Europe

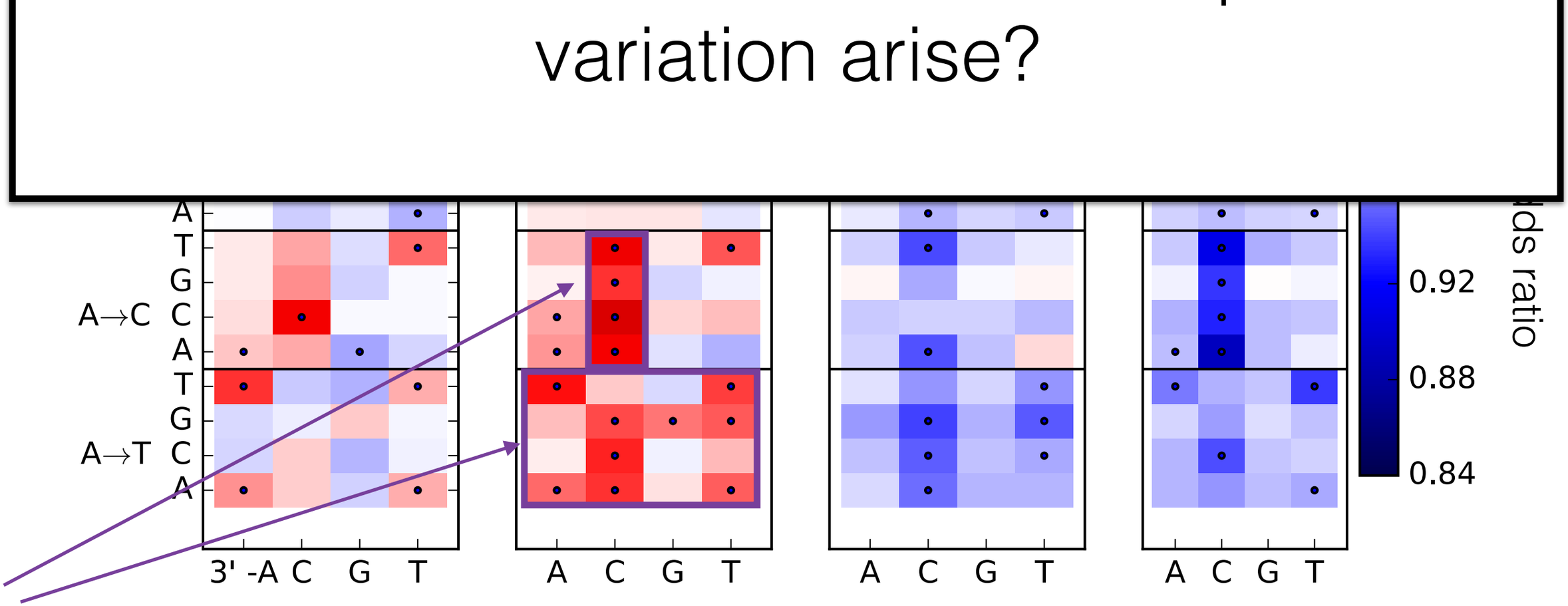


A-to-T and AC-to-CC transversions are enriched in East Asia

TCC-to-TTC transitions are enriched in South Asia as well as Europe

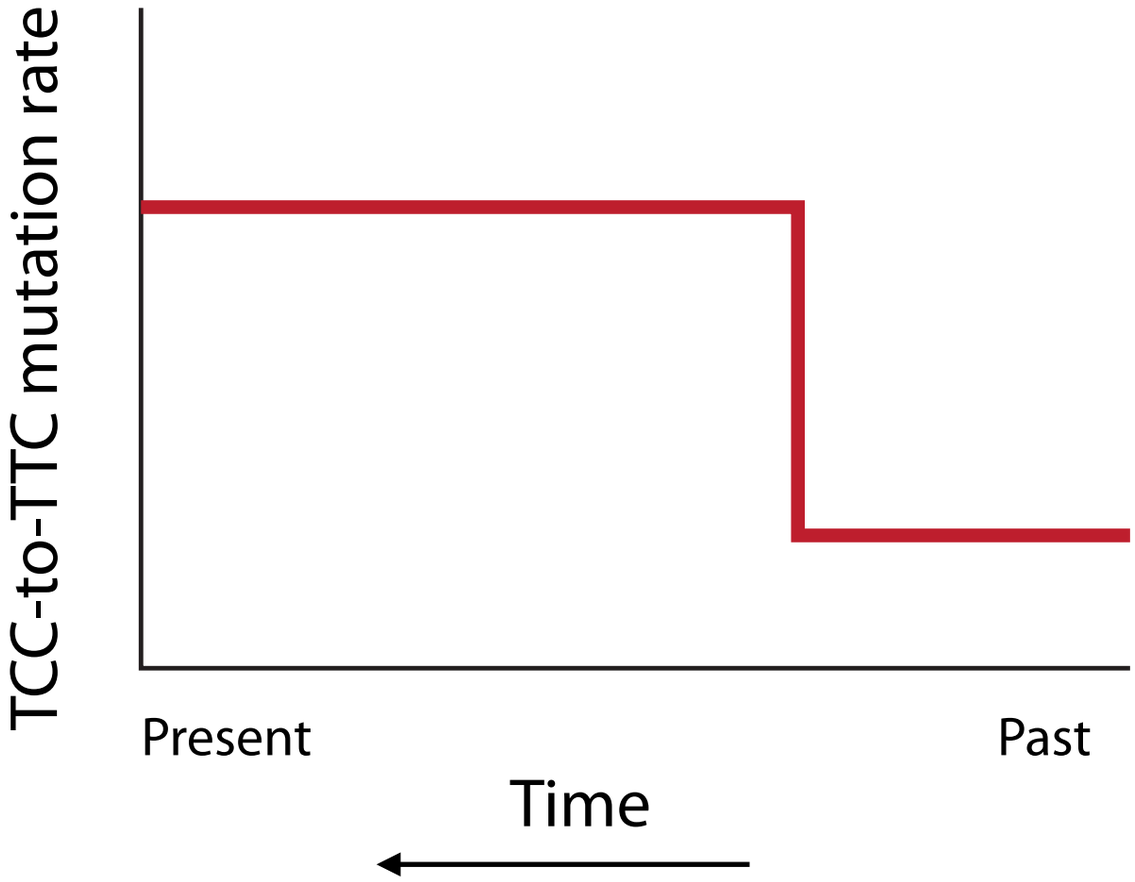


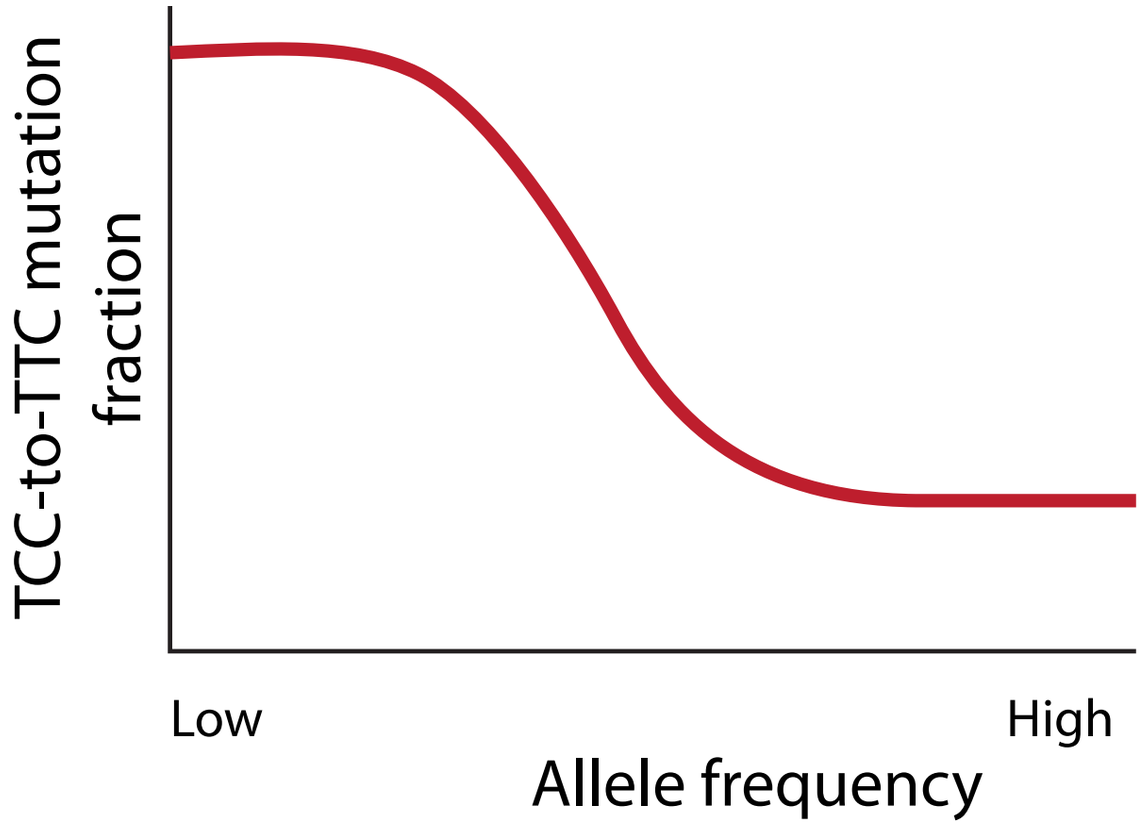
How and when did this mutation spectrum variation arise?

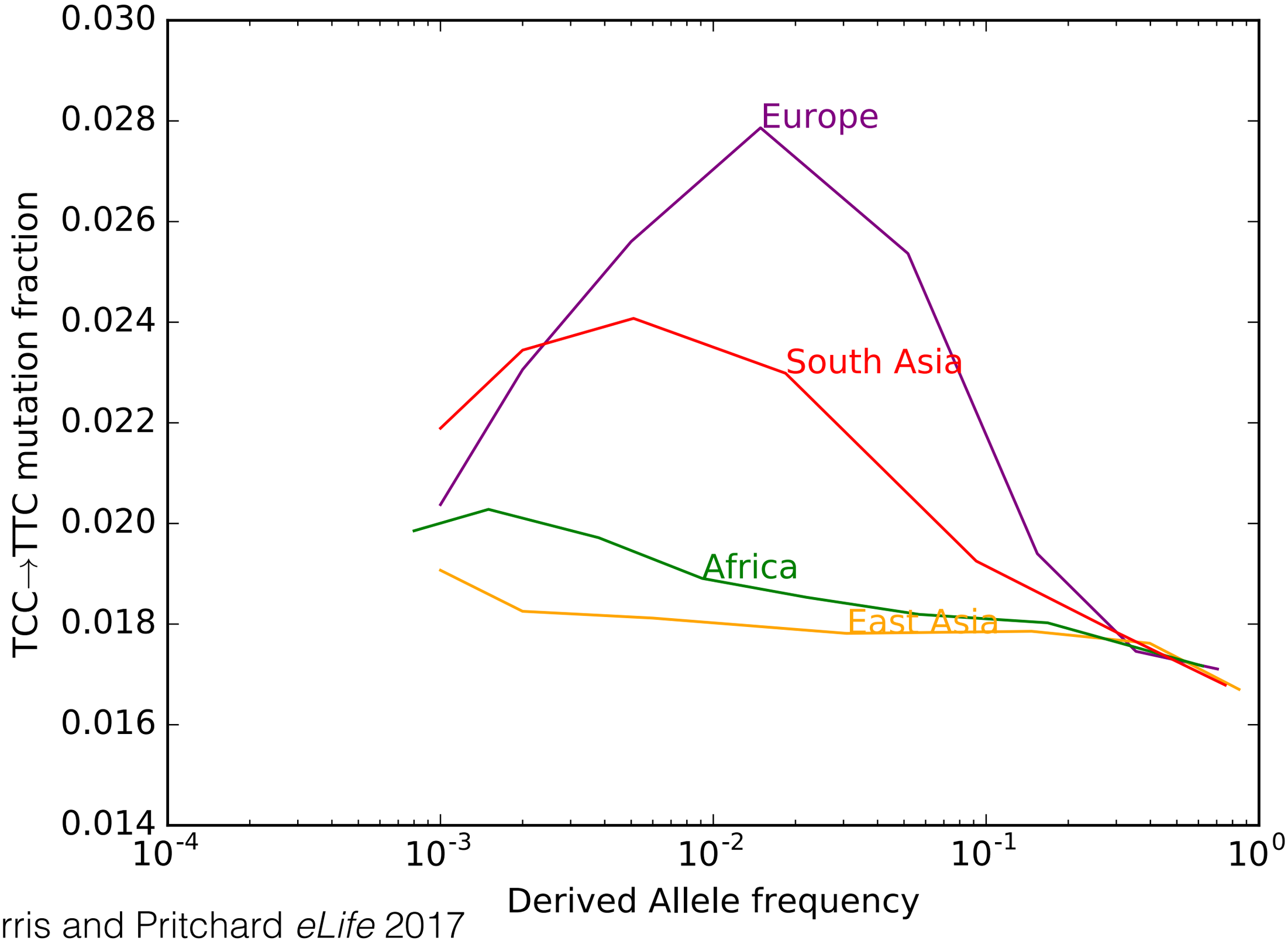


A-to-T and AC-to-CC transversions are enriched in East Asia

Hypothetical Signature of a TCC-to-TTC mutation rate increase







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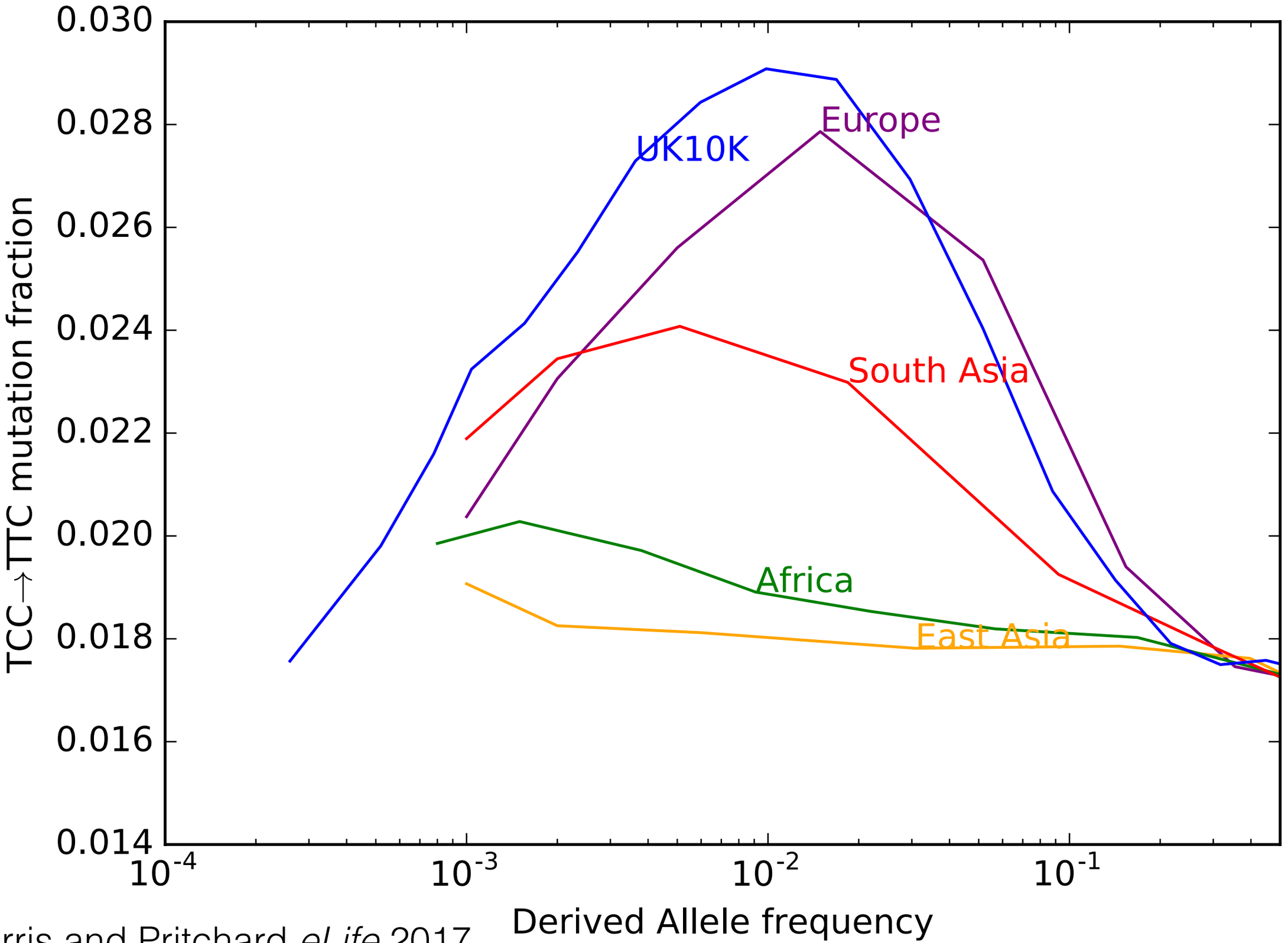
Pulse replicates in the UK10K data



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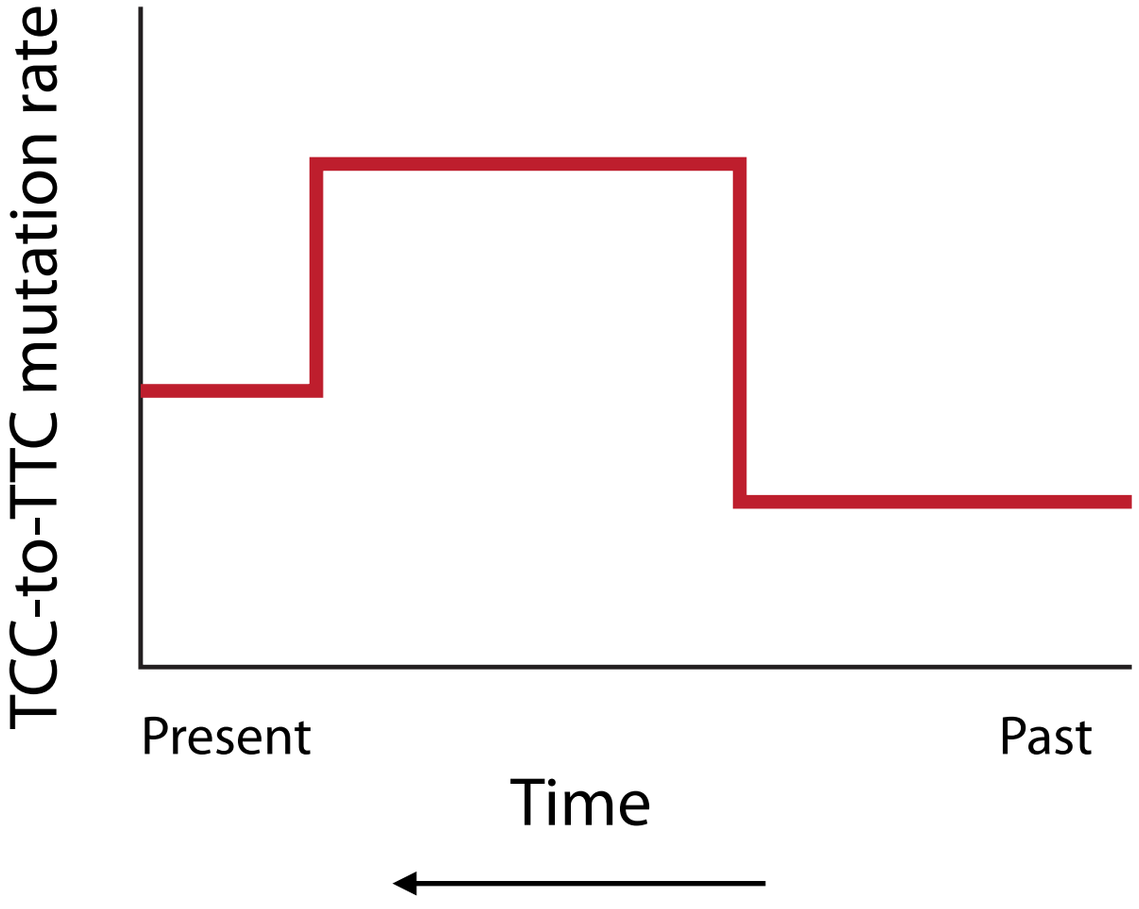
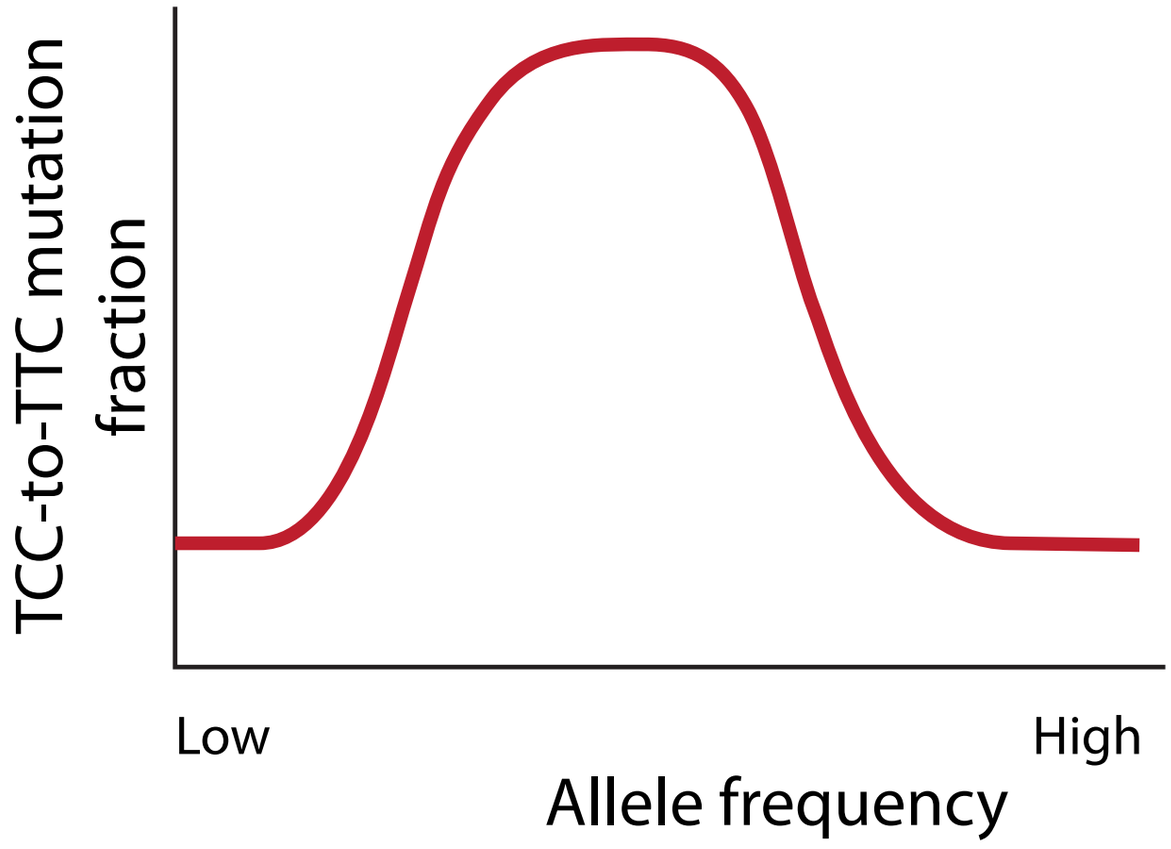
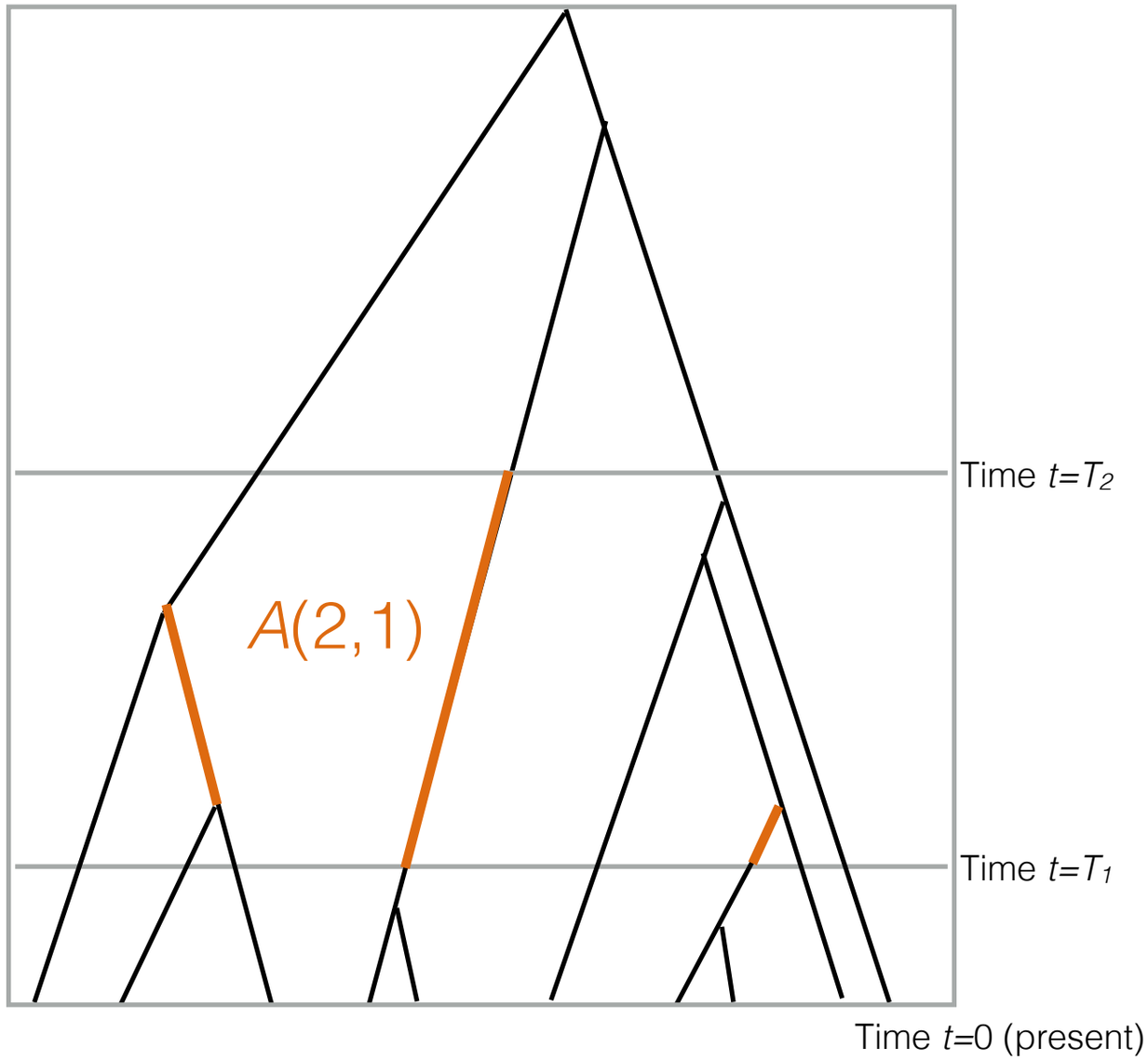
A pulse of TCC-to-TTC mutations in Europe and South Asia?





Expected TCC fraction as a function of allele frequency

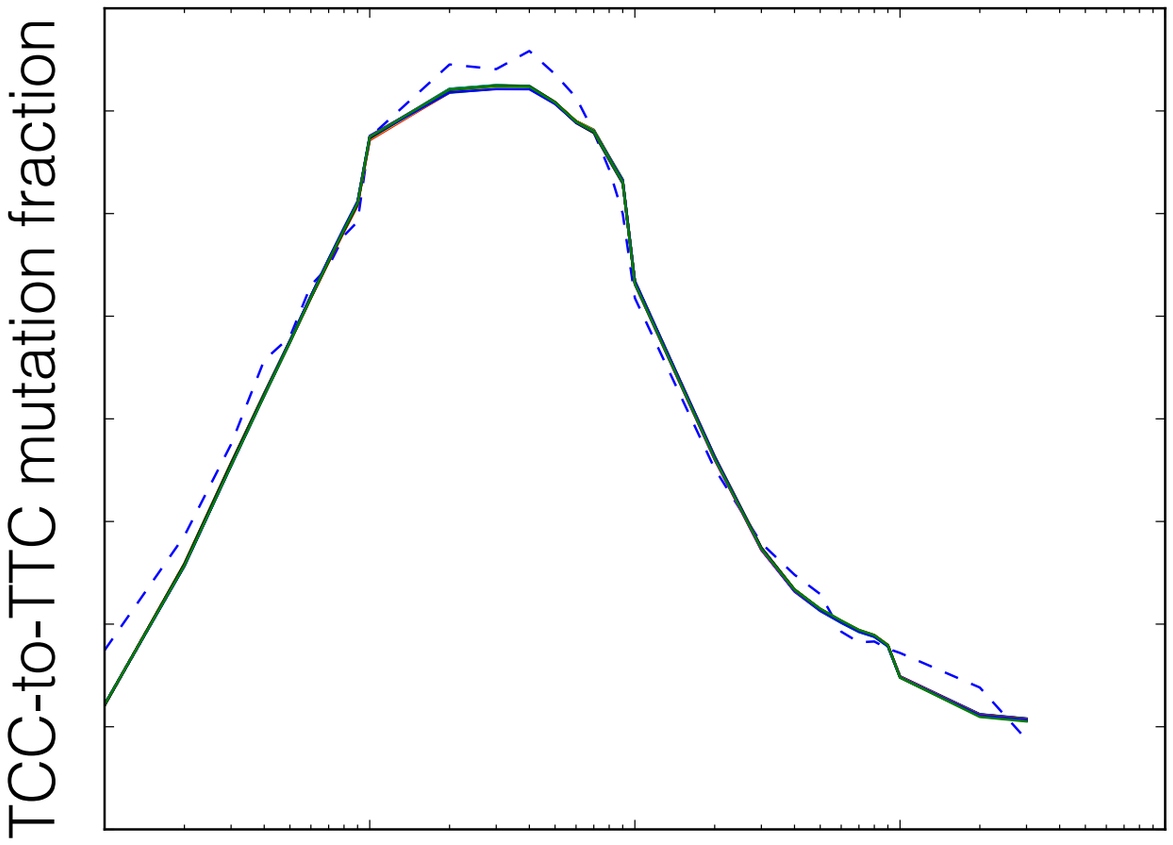
- Partition time into discrete intervals
- $A(k,i)$ = the total branch length subtending k lineages between times T_i and T_{i-1}
- r_i ~ the rate of TCC mutations between T_i and T_{i-1}



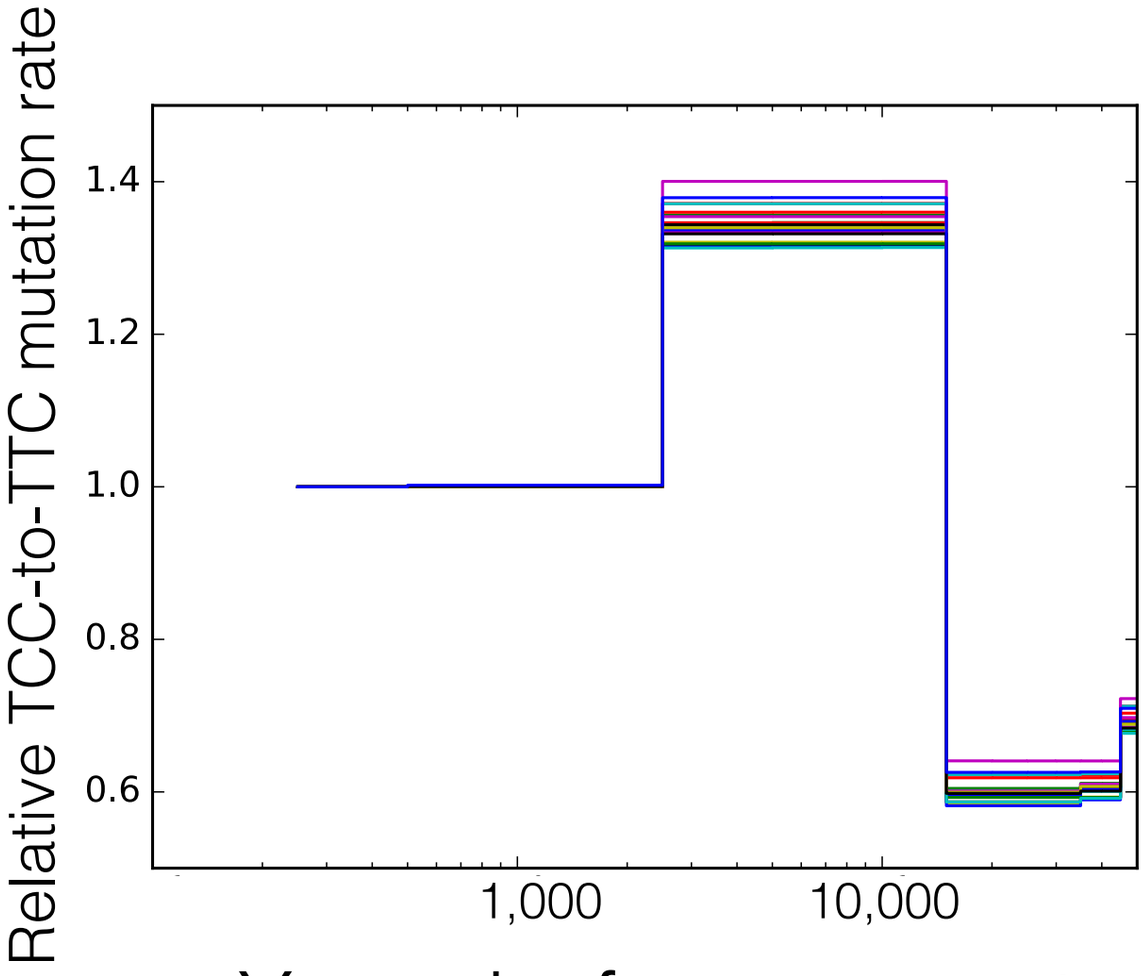
Expected TCC fraction as a function of allele frequency is

$$E[f(k)] \sim (\sum_i A(k,i)\ r_i) / \sum_i A(k,i)$$

Inference of a mutation pulse lasting from 15,000 to 2,000 years ago

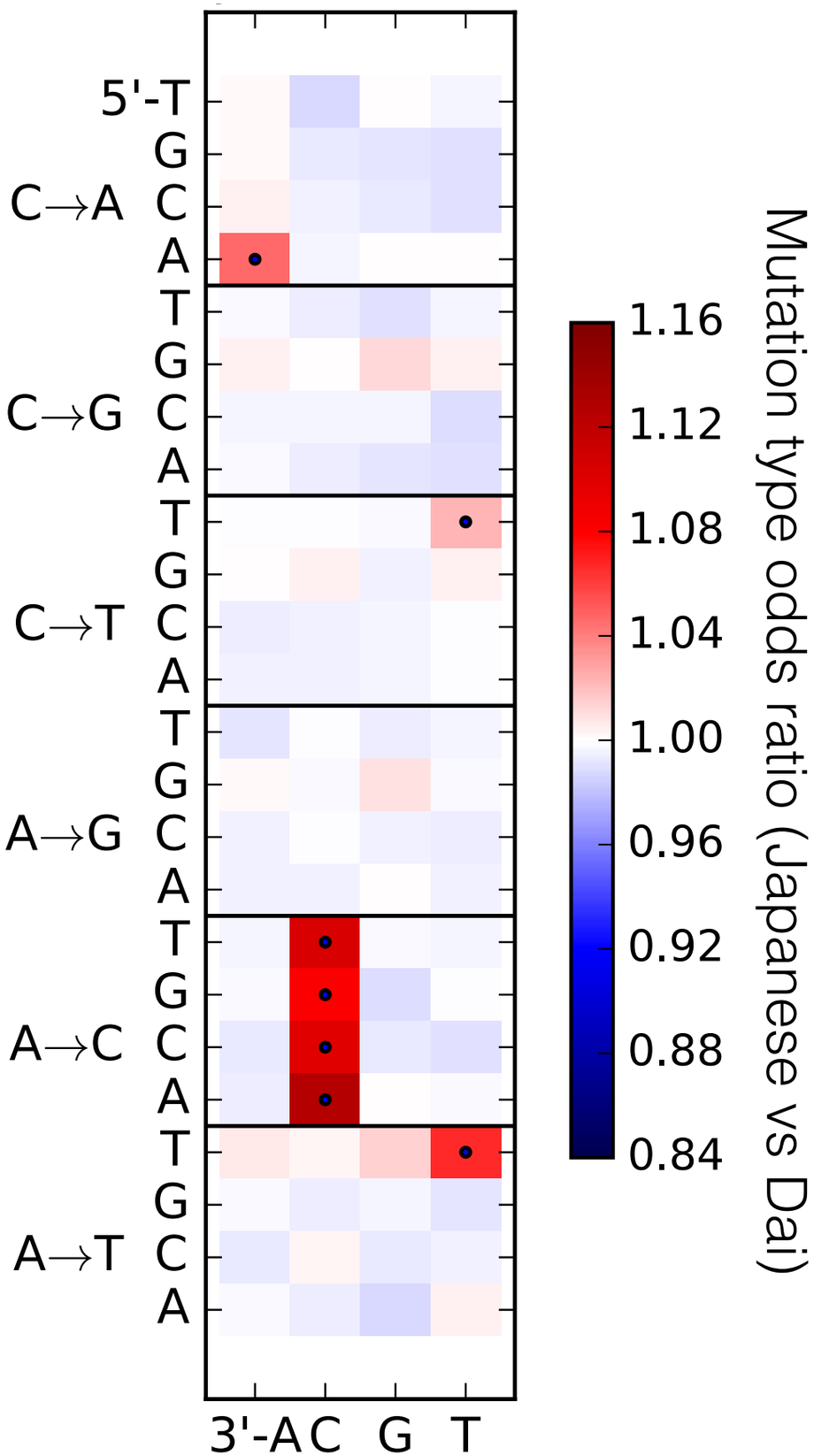


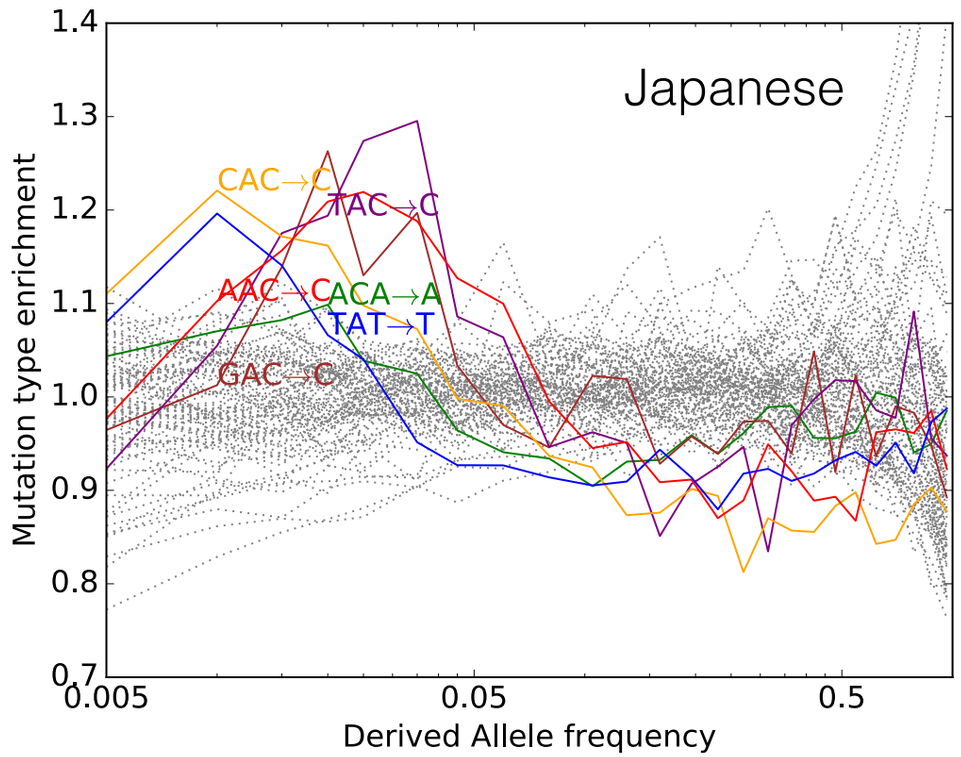
Allele frequency

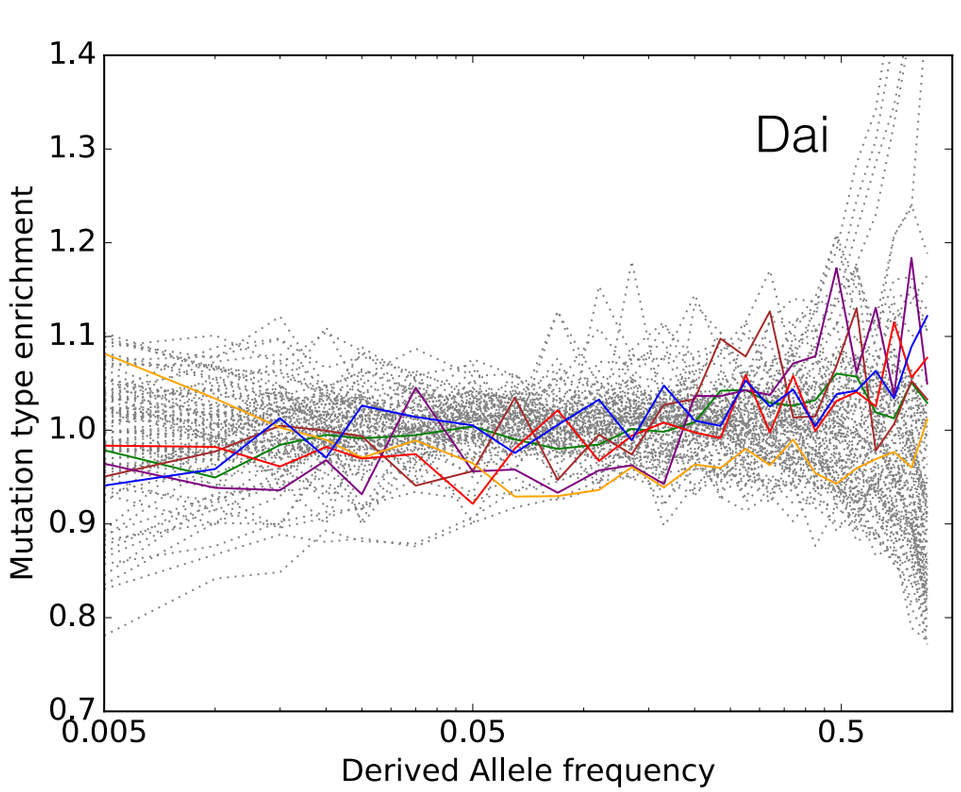


Years before present

A younger Japanese mutation pulse







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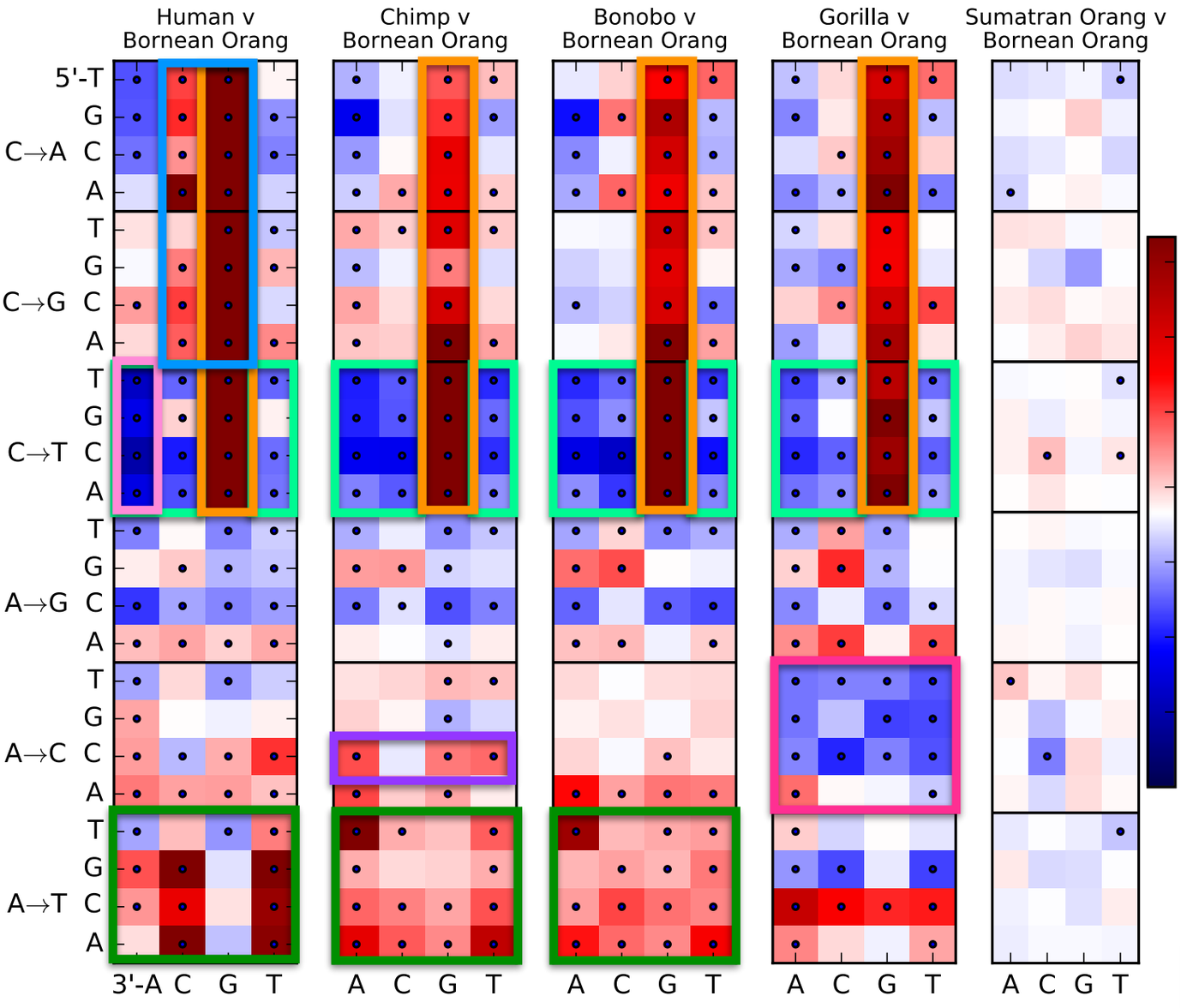
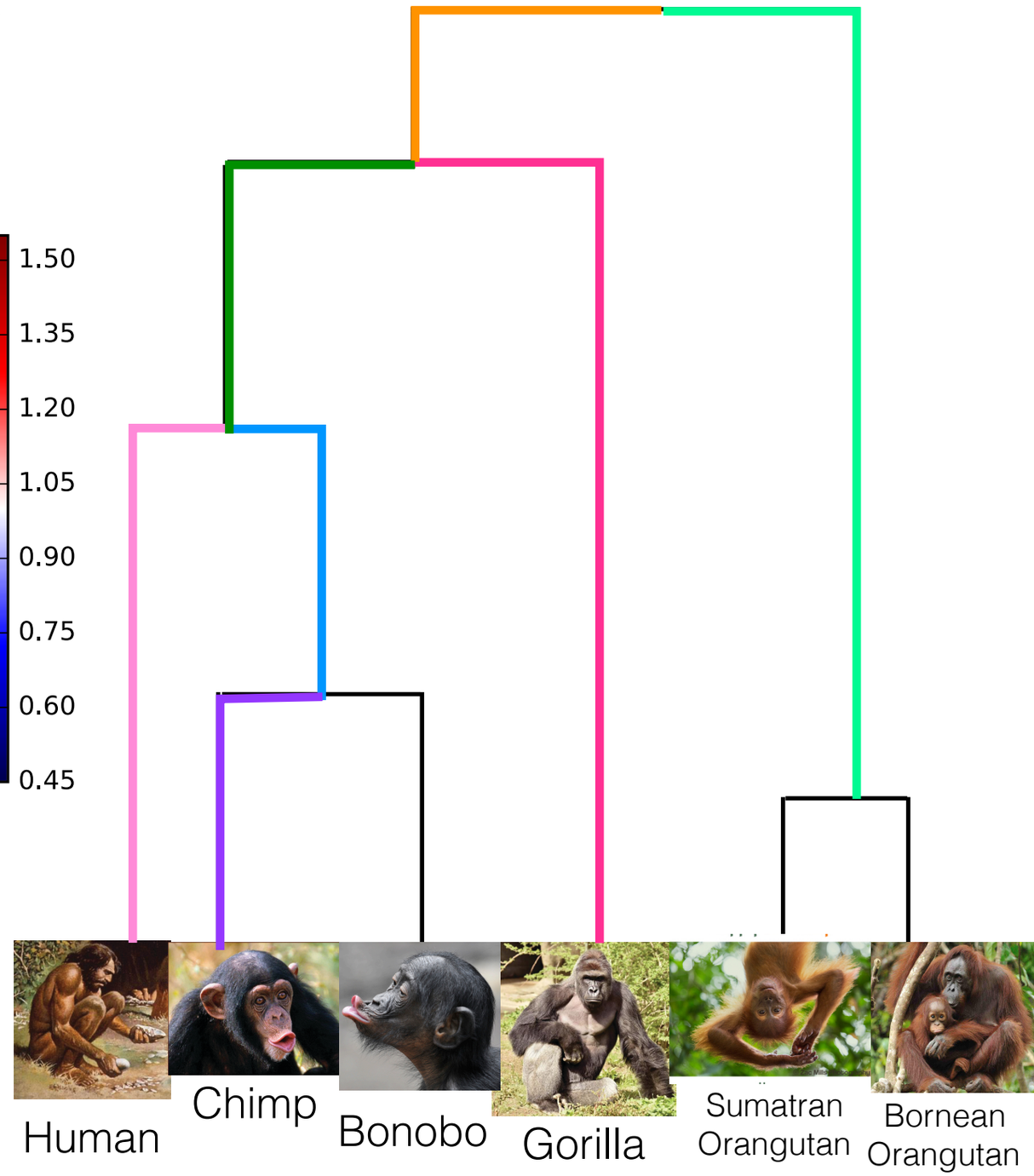
Great ape mutation spectrum evolution

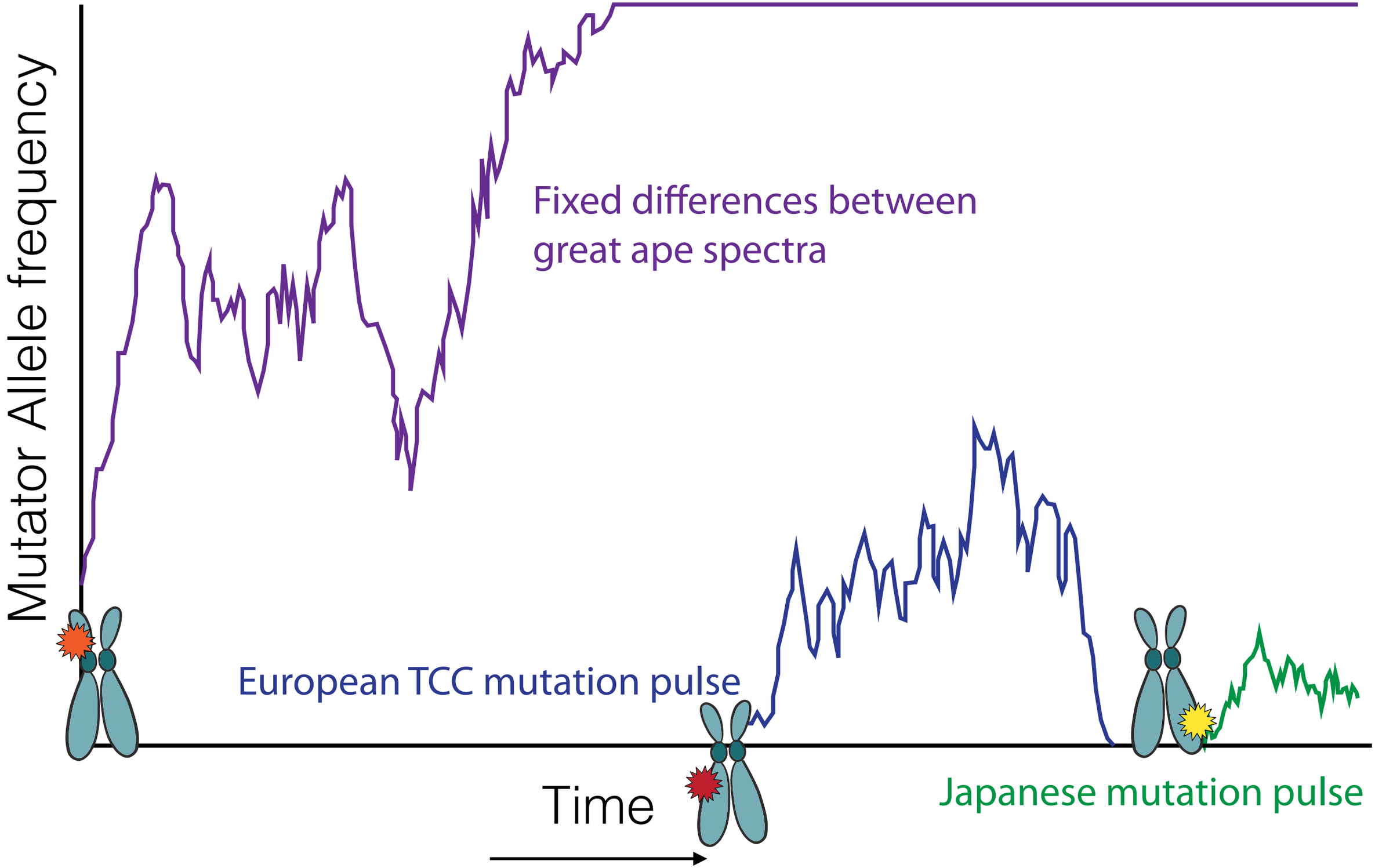




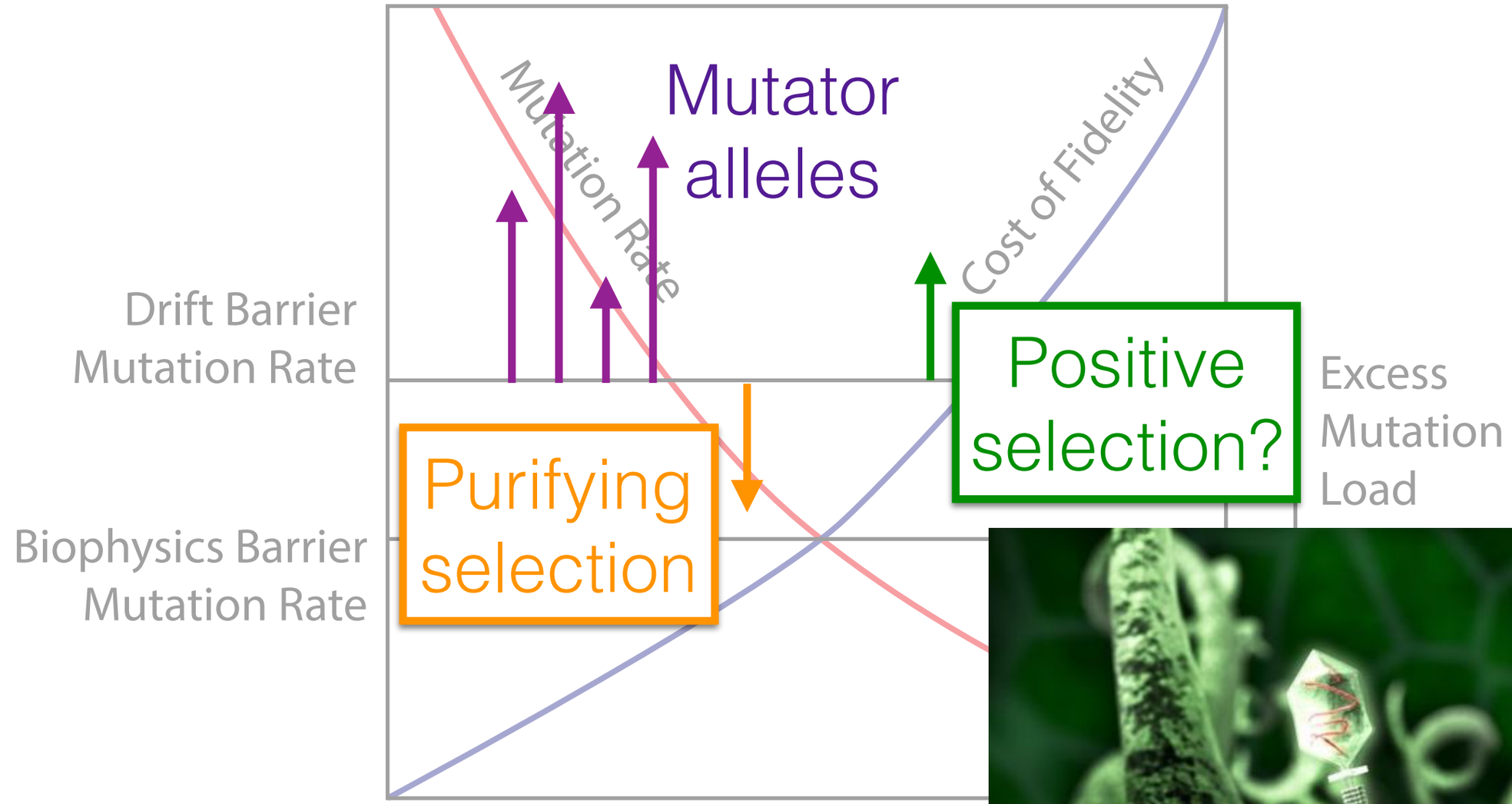
Within-species SNPs from 79 great ape whole genomes (Prado-Martinez, et al. 2013)

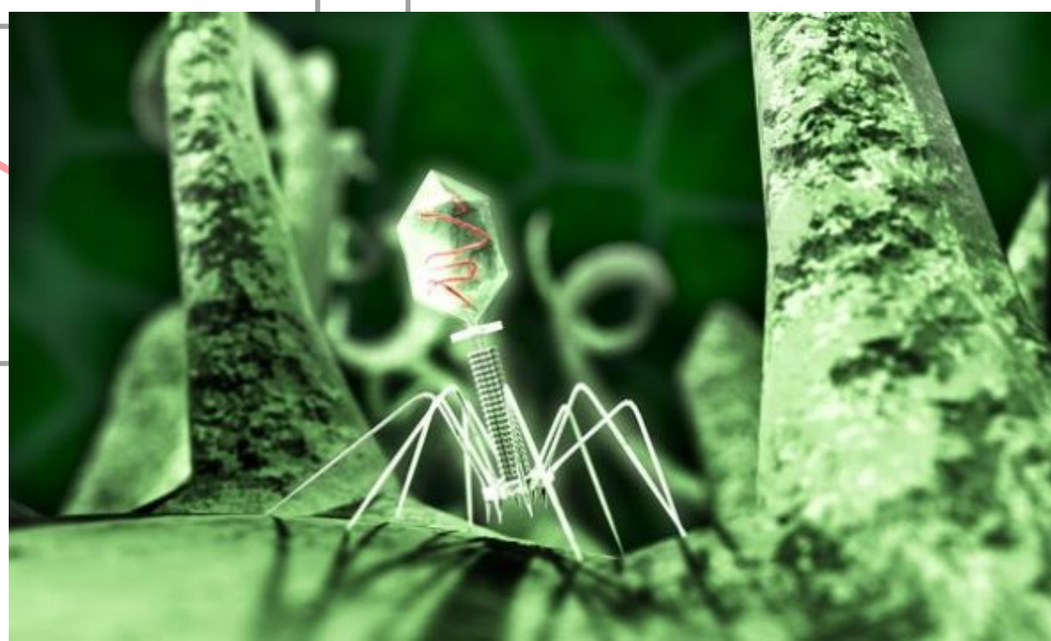
Harris and Pritchard *eLife* 2017

Future direction: are mutation pulses the relics of lost mutator alleles?



How mutator alleles could promote rapid mutation spectrum turnover

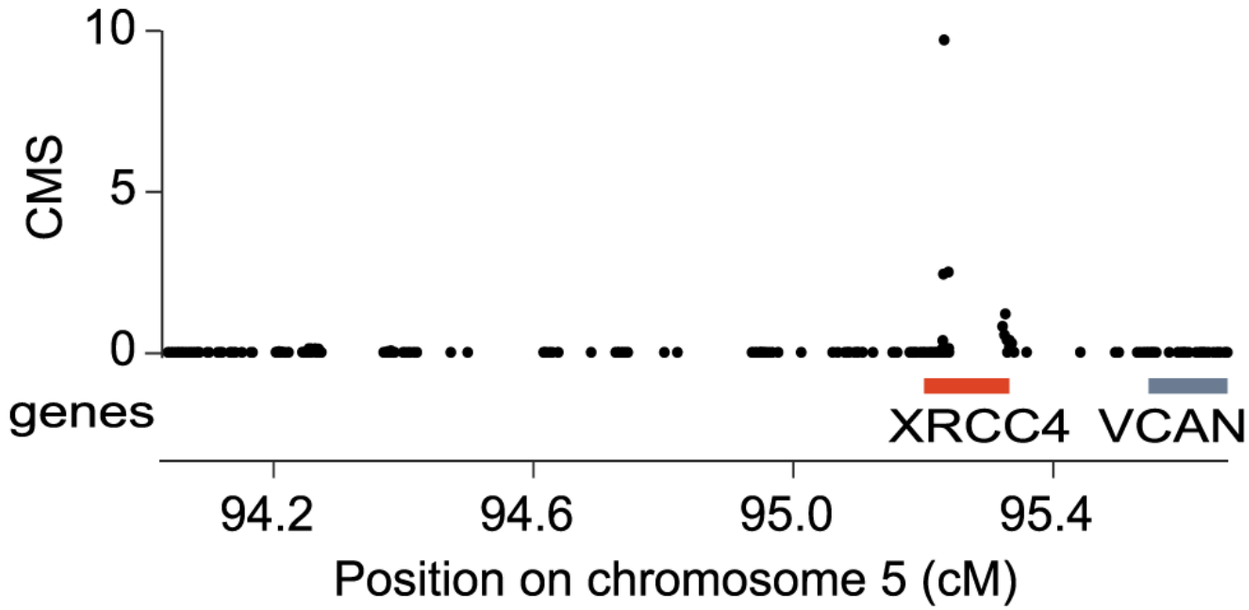




Sawyer and Malik *PNAS* 2006

Positive selection in DNA repair genes and other housekeeping genes

- BRCA1 & BRCA2 are under positive selection in primates
- 5 Nonhomologous end joining genes experienced positive selection during primate evolution, incl XRCC4 which has been under selection in Europeans
- Iron-uptake receptor TfR1 evolves under positive selection to avoid facilitating viral entry



Demogines, et al. 2010
Demogines, et al. 2013

A case study of a mutational process that complicates population genetics

Multinucleotide mutations (MNM) are nearby SNPs that appear in the same generation

AAAGTTAGCCGACAC

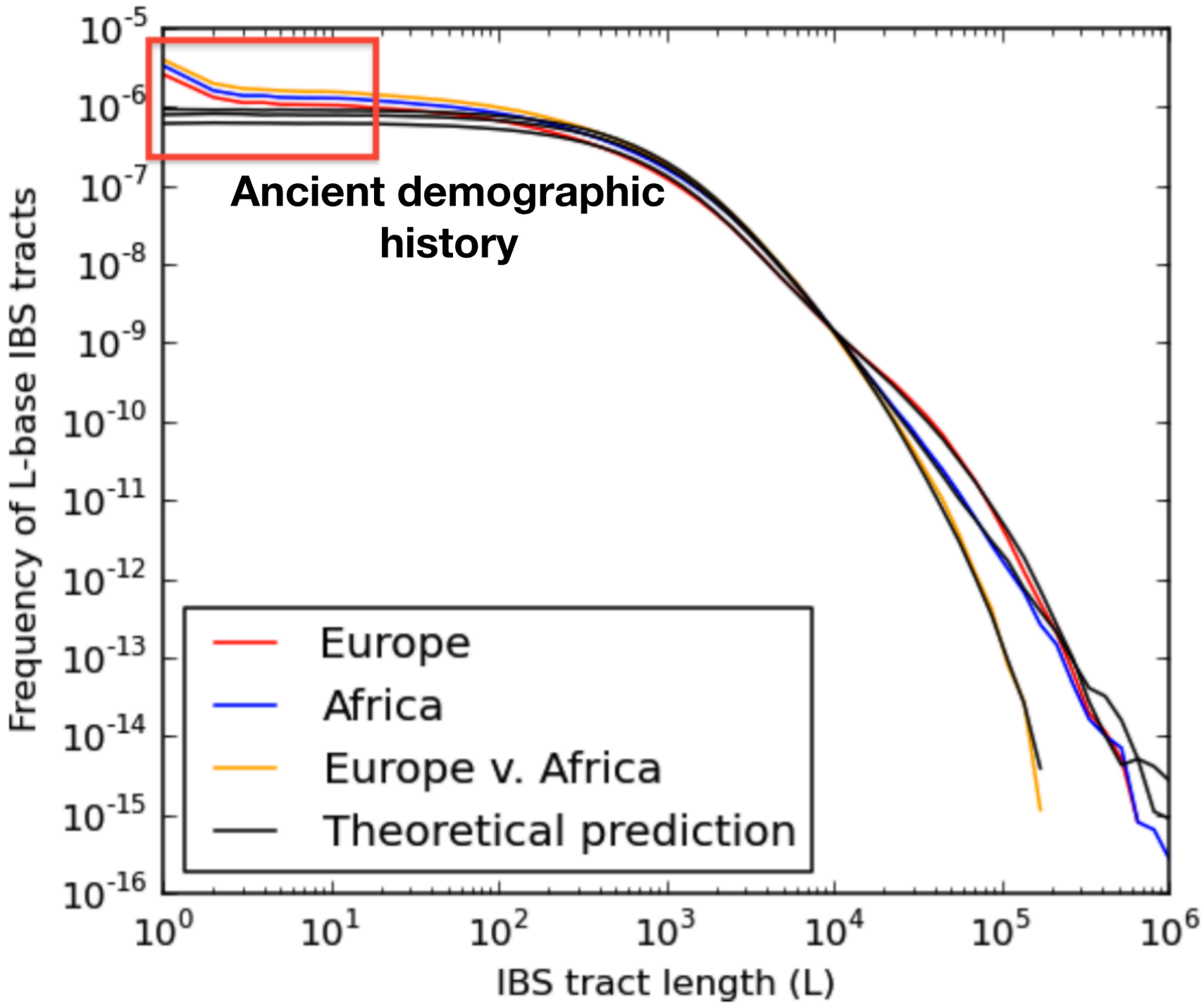
↓

AAAGATAACCGACAC

Schrider, et al. 2011

Harris and Nielsen 2014

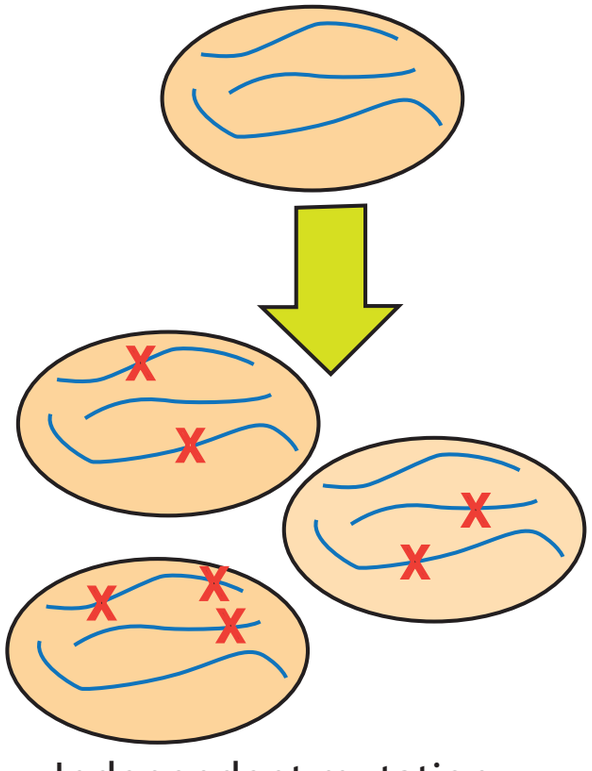
Effect of MNMs in the distribution of tracts of identity by state



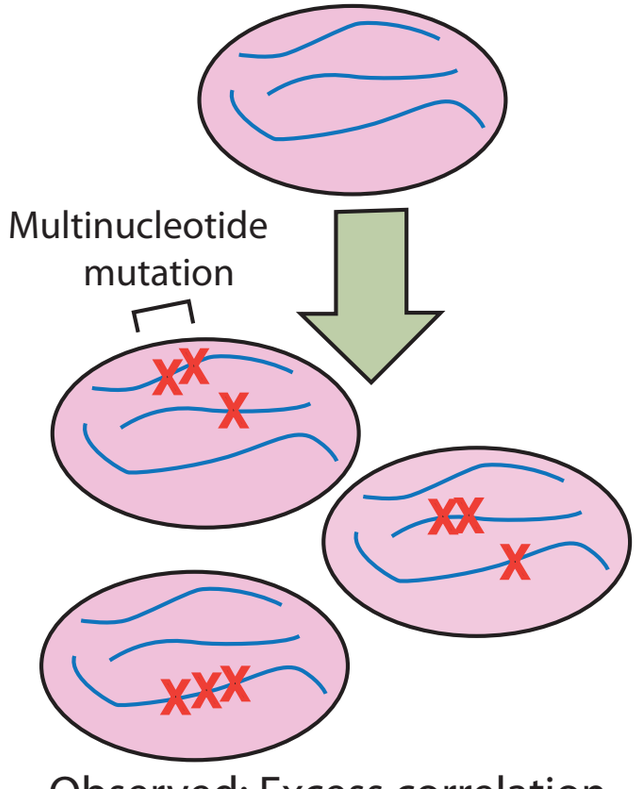
Direct evidence for MNMs

- Most methods assume that all SNPs arise from rare, independent mutation events

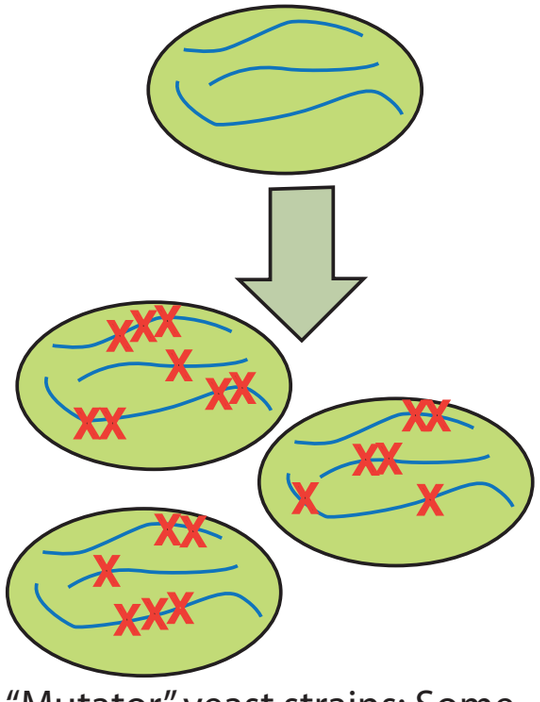
- MA experiments and trio sequences show that *de novo* mutations are too clustered for this to be true



Independent mutation hypothesis

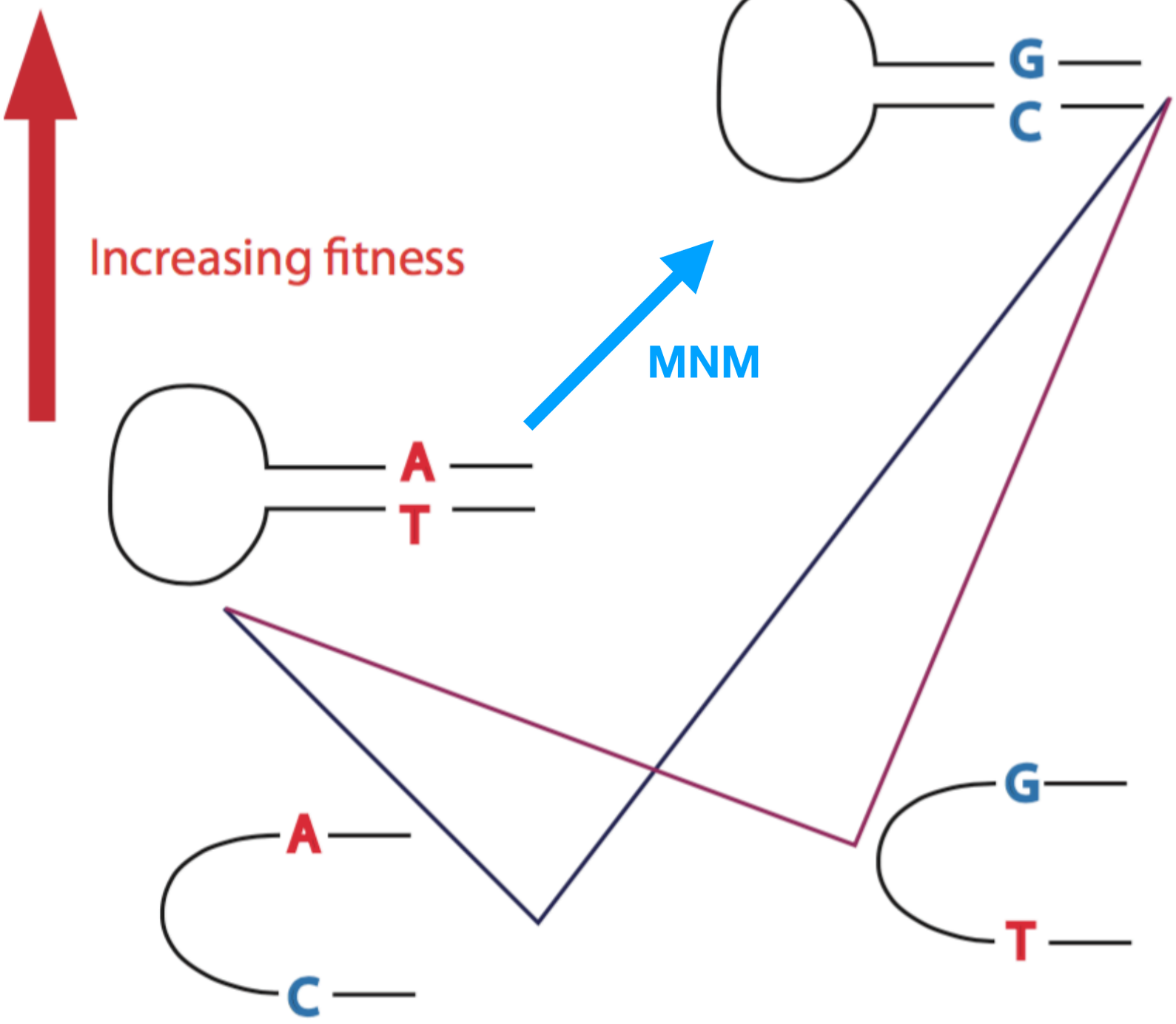


Observed: Excess correlation between *de novo* mutations

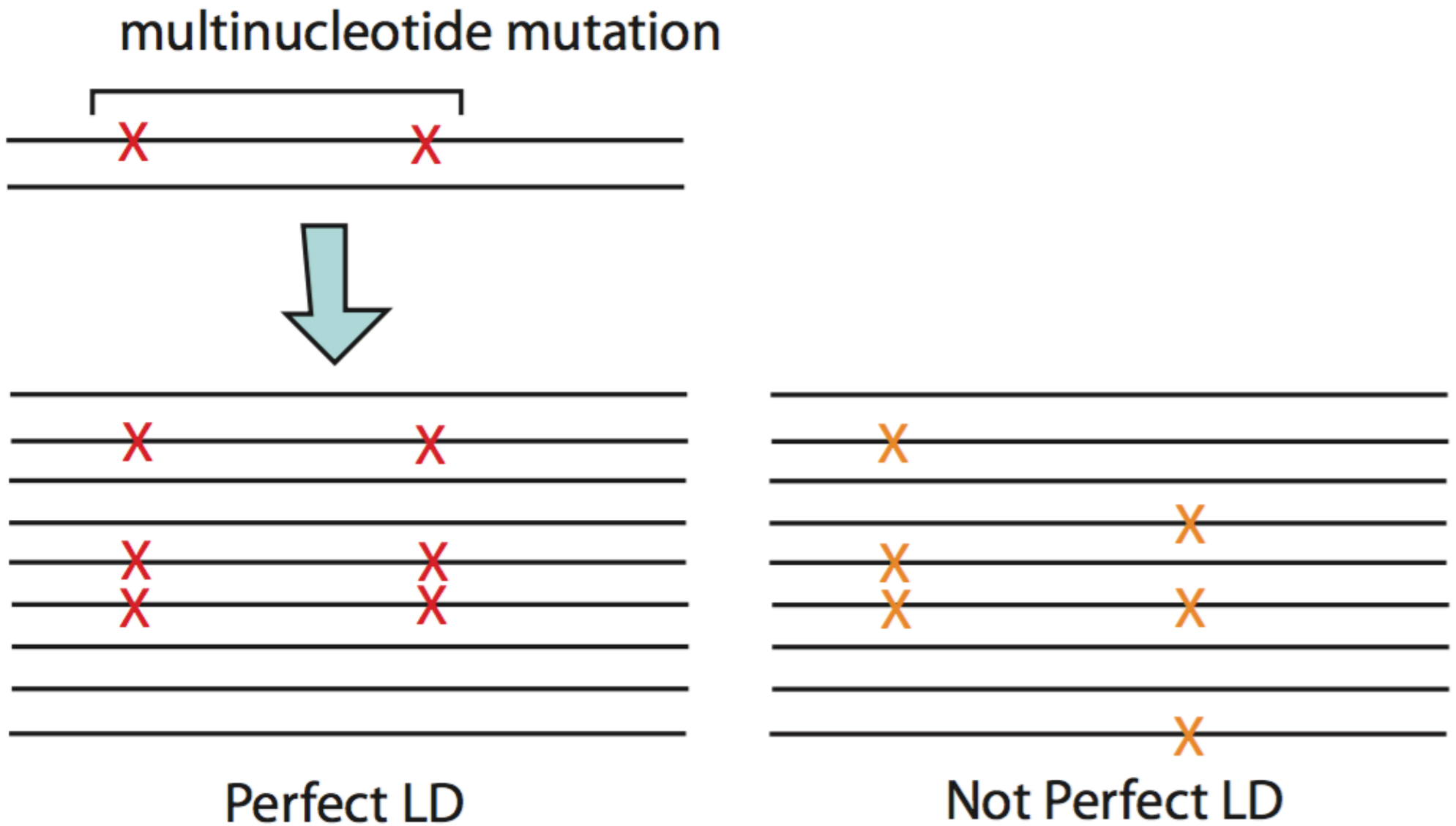


"Mutator" yeast strains: Some abnormal polymerases generate clustered mutations at a higher rate

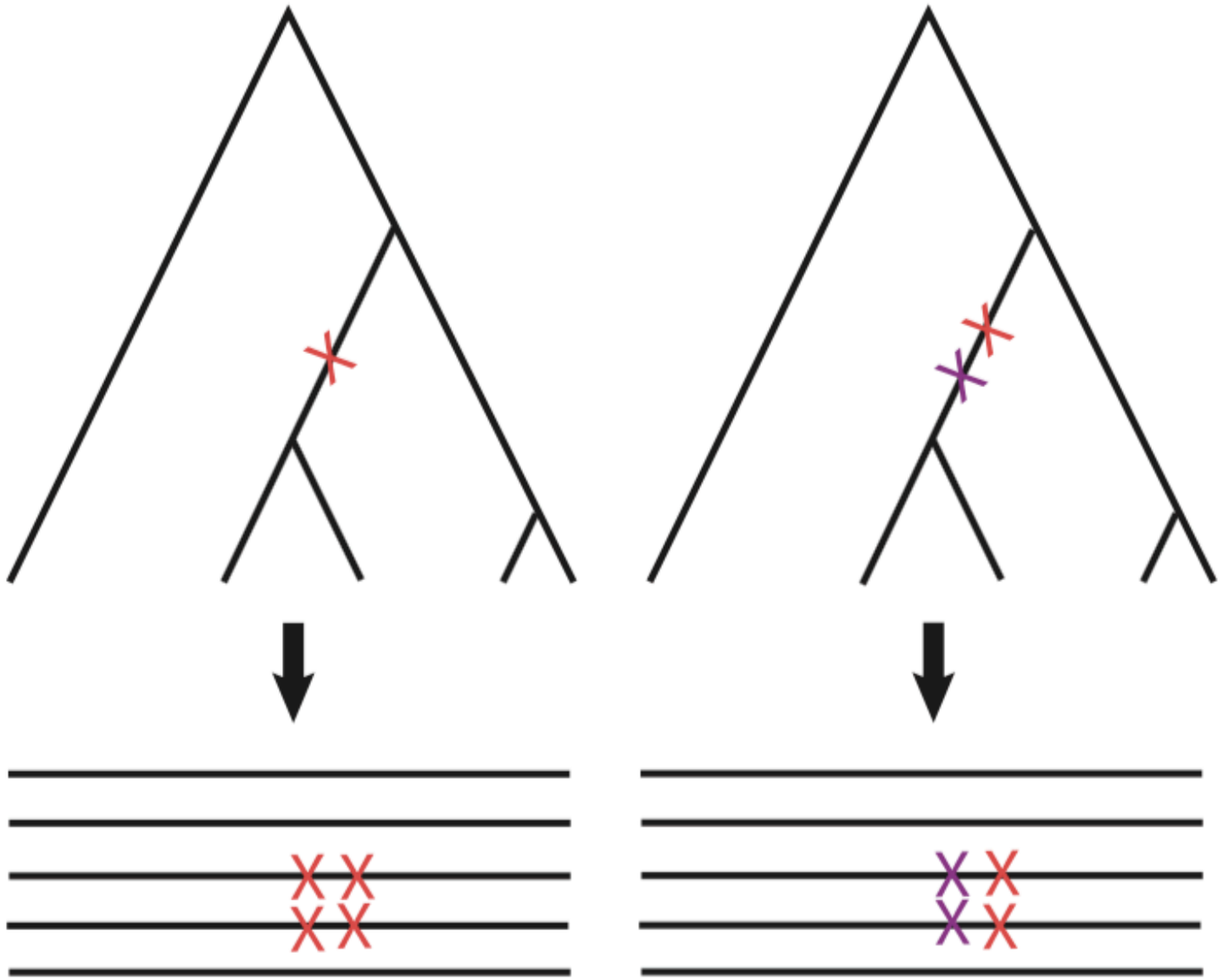
MNMs could accelerate evolution across fitness valleys



Multinucleotide mutation should create pairs of SNPs in *perfect linkage disequilibrium (LD)*
(derived alleles occur in the same set of individuals)

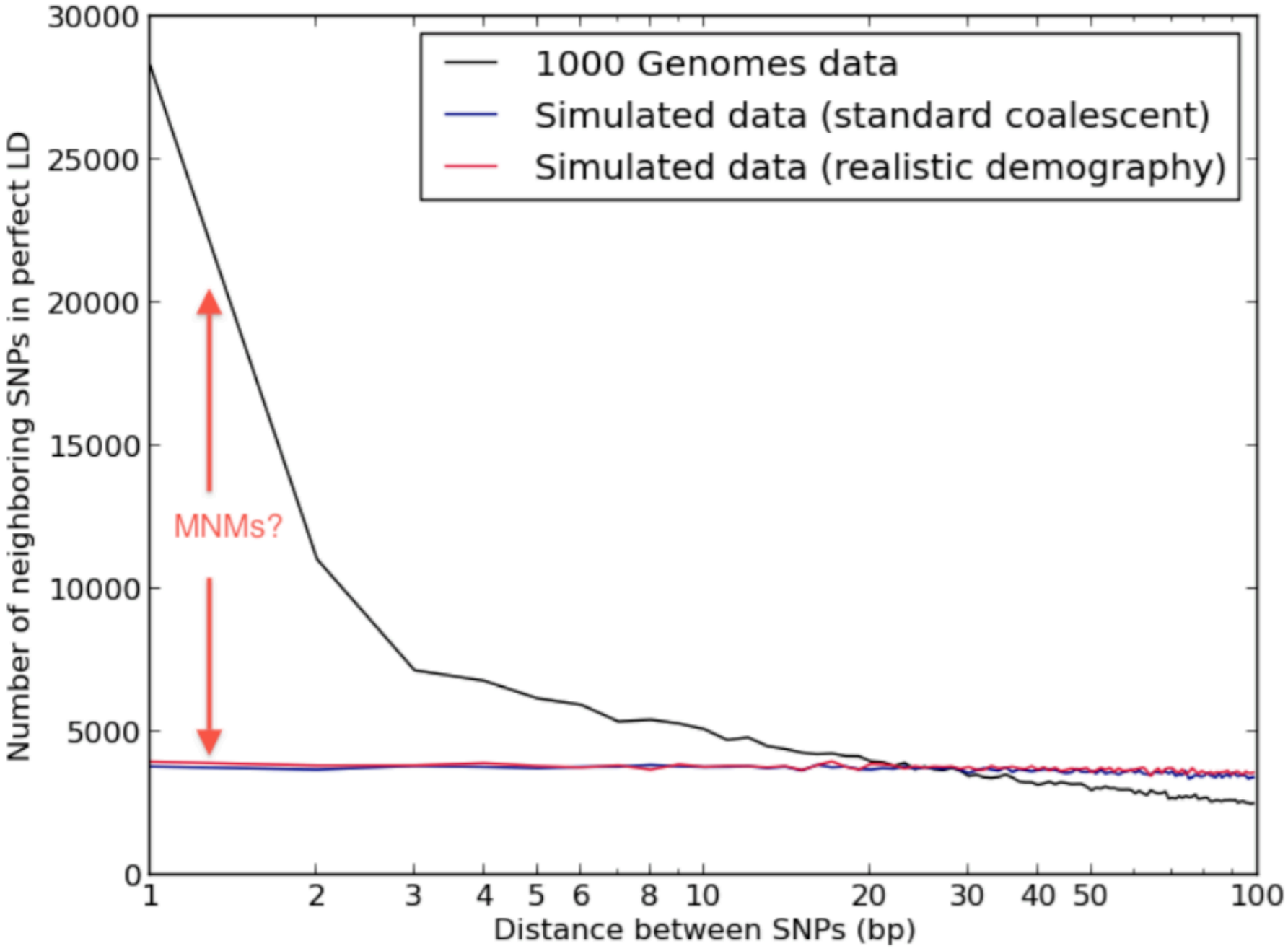


Independent mutations at neighboring sites can also create SNPs in perfect LD



One MNM

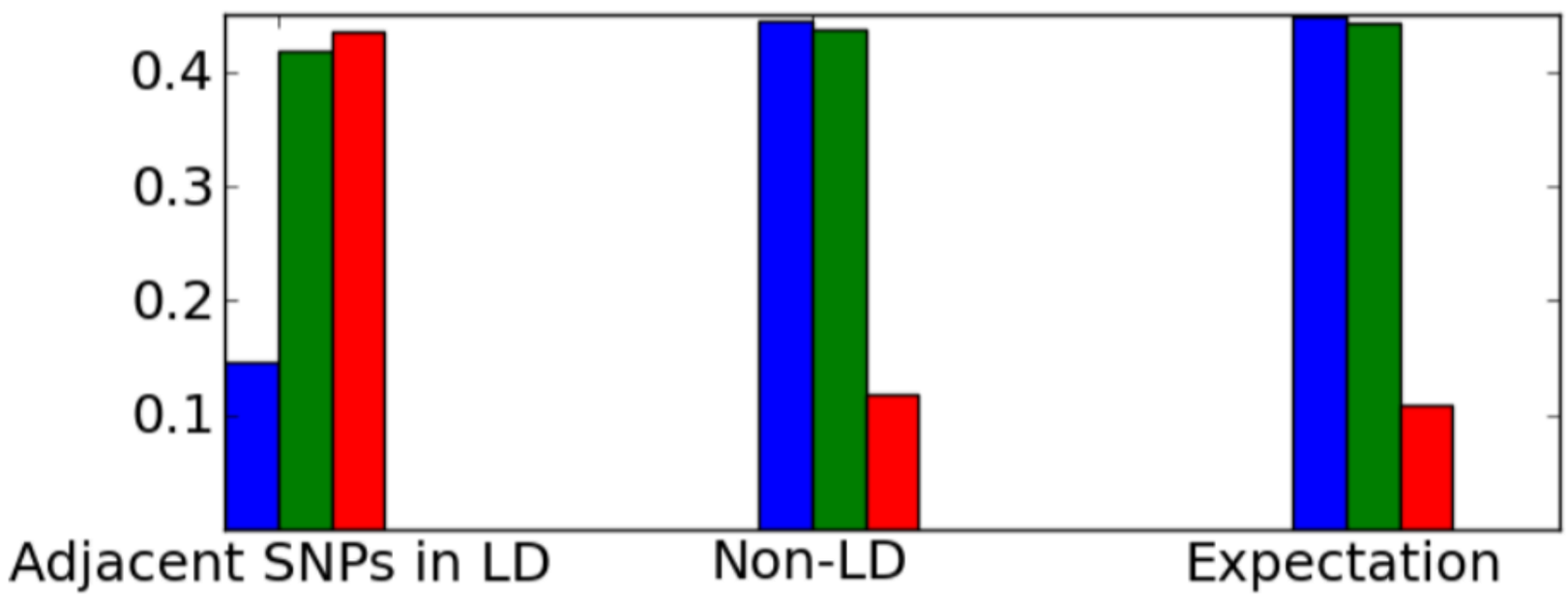
Two independent mutations



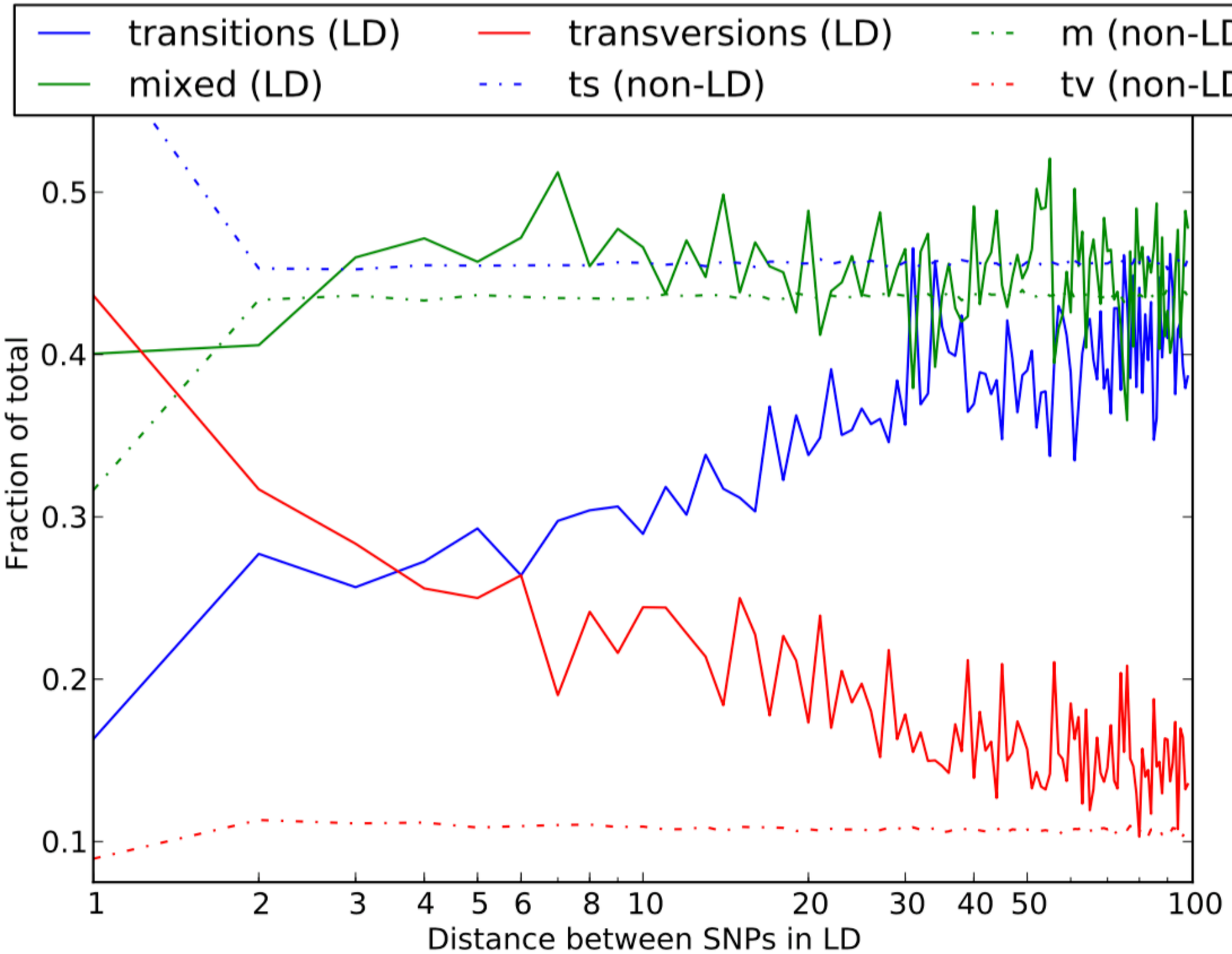
Compared to theoretical predictions, the 1000 Genomes Phase I data (1,092 humans from Africa, Europe, Asia, and the Americas) has excess close-together SNPs in perfect LD

SNPs in perfect LD are enriched for transversions

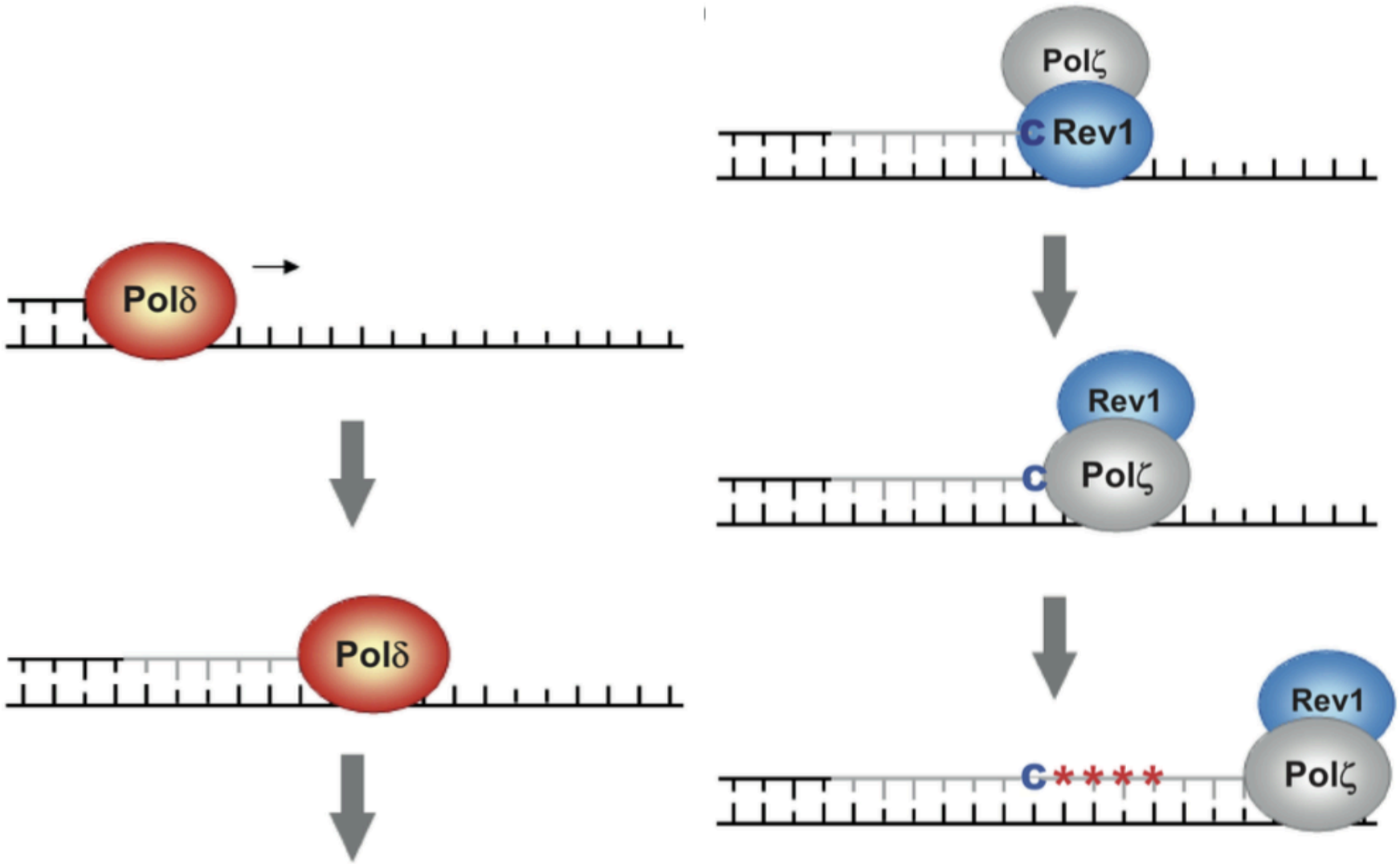
- 66% of human mutations are transitions (A>G, G>A, C>T, T>C)
- Pairs of SNPs in perfect LD are enriched for transversions, suggesting a different balance of mutational signatures



Transversion-enrichment as a function of the distance between linked SNPs



A candidate mechanism: error-prone translesion synthesis



Northam *et al.*, *Nucleic Acids Res.* 2014

Matching mutational signatures between human variation and laboratory yeast

Environmental and Molecular Mutagenesis 53:777–786 (2012)

Research Article

DNA Polymerase zeta Generates Clustered Mutations During Bypass of Endogenous DNA Lesions in *Saccharomyces cerevisiae*

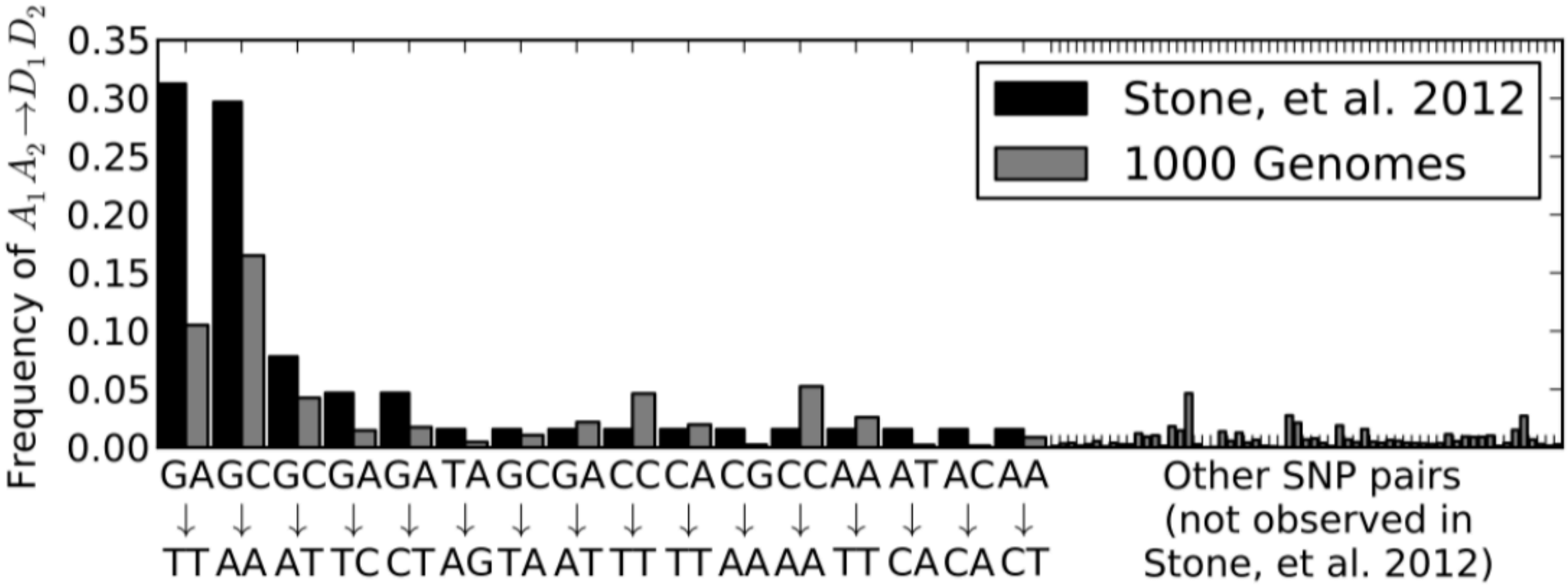
Jana E. Stone, Scott A. Lujan, and Thomas A. Kunkel*

Laboratory of Molecular Genetics and Laboratory of Structural Biology, National Institute of Environmental Health Sciences, NIH, DHHS, North Carolina

- Stone, et al. created yeast deficient in nucleotide excision repair machinery and observed a high MNM rate

- Mechanism: increased translesion synthesis by Pol Zeta

A matching dinucleotide mutational signature



Further characterization of the Pol zeta mutational signature

- GC>AA mutations are concentrated in late-replicating regions of the genome

- Usually occur in GCG context, triggered by CpG deamination followed by polymerase stalling

- CpG deamination is triggered by transcription; usually occurs on transcribed strand

- Some genes contain GC>TT mutation hotspots, including HRAS where the mutation causes the Mendelian disorder Costello Syndrome

Seplyarskiy, et al. 2015

More on the weirdness of Costello Syndrome

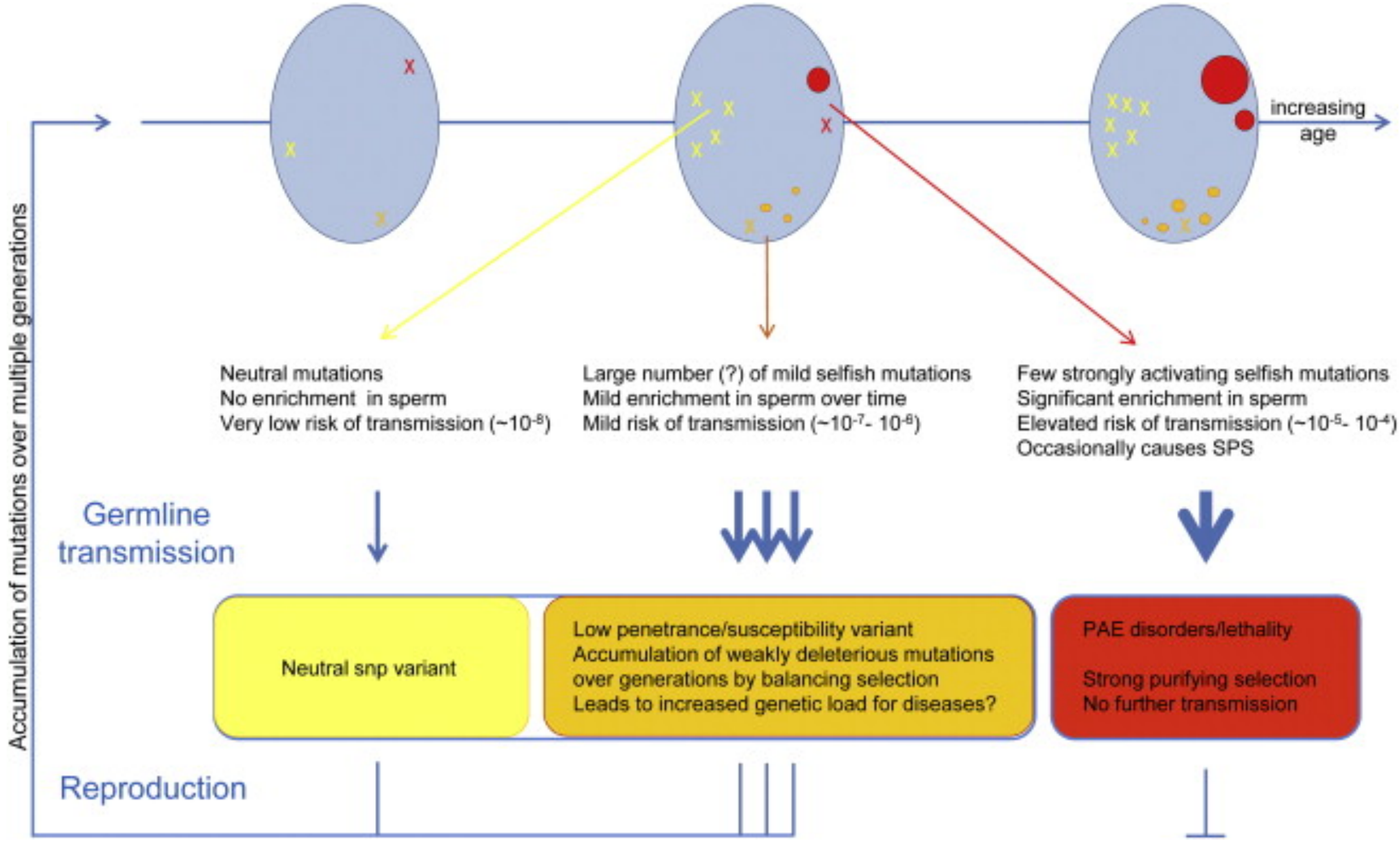
- A high penetrance Mendelian disease caused by a nonsynonymous point mutation in the HRAS oncogene

- Causes developmental delay and early childhood tumors

- Most commonly caused by a GC>TT mutation with a mutation rate of 10^{-5} per generation (normal mutation rate is 10^{-8} per site per generation)

- Biggest risk factor is paternal age

HRAS mutations experience selfish selection within the testis



Goriely and Wilkie 2012

UNIVERSITY of WASHINGTON

Genome Sciences
UNIVERSITY OF WASHINGTON

The Harris Lab is recruiting