Bayesian Modeling of Numt Evolution with Application to the Estimation of Gorilla Divergence Times

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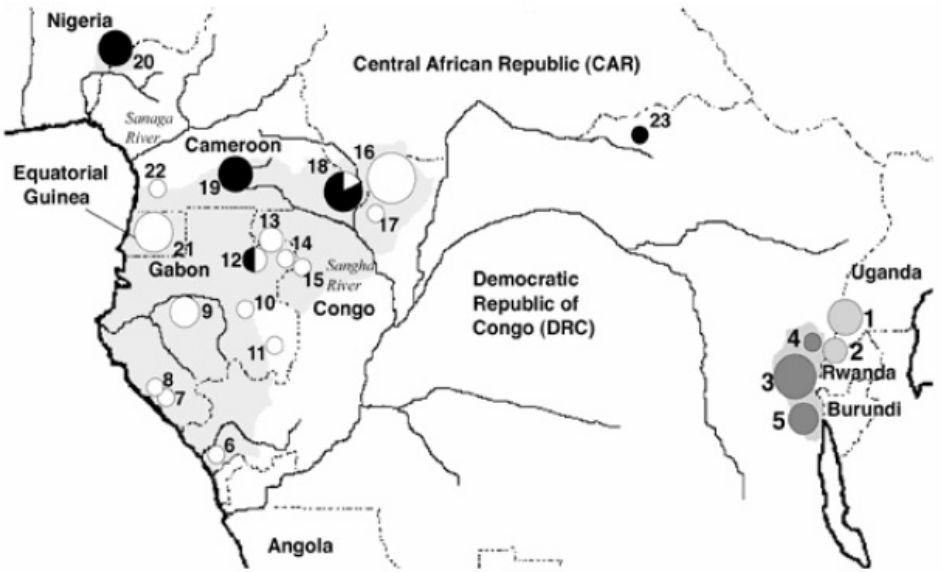
The Modern Distribution of Gorilla Populations



- There are three subspecies of gorillas:
  - Mountain Gorillas (MTG, sites 1–2)
  - Eastern Lowland Gorillas (ELG, sites 3–5)
  - Western Lowland Gorillas (WLG, sites 6–23)
- Eastern gorillas (ELG and MTG) are physically separated from western gorillas (WLG) by at least 850 km.
- Gorillas live within forests and are not observed crossing open savannah.

Gorilla Phylogeopgraphy

- How did the modern distribution of gorillas arise?
- The *Pleistocene refugia theory*:
  - The Pleistocene was a period of frequent glaciation from about 1.8 million to about 11,000 years ago.
  - During times of maximal glaciation, central Africa would have been arid and forests would have fragmented.
  - Gorilla populations may have been restricted to small refugia.
- Did the current split between eastern and western gorillas originate during the Pleistocene?

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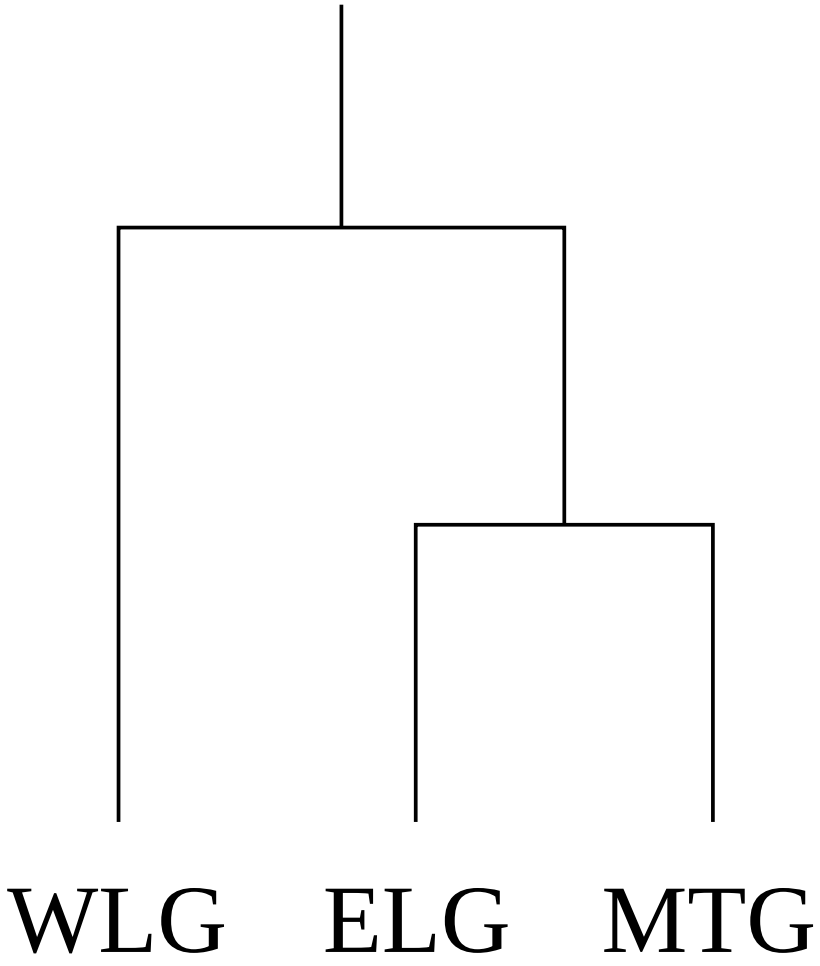
The Phylogeographic Problem



- We wish to address questions related to the phylogeographic history of gorilla populations on the basis of genetic sequence data sampled today.

HV1 sequences

- Many population studies of humans and other primates are based on DNA sequence data from the *first hyper-variable region (HV1)* of the control region of the mitochondrial genome.
- The rate of substitution in HV1 is relatively high.
- HV1 is thought to evolve neutrally.
- The mitochondrial genome is inherited maternally and there is not thought to be any recombination.
- Within the last few years, scientists have developed the technology to extract mtDNA from hairs that gorillas shed in night nests.
- This means that the DNA of gorillas *can be sampled noninvasively from wild populations*.

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Numt Sequences

- Sometimes, the PCR reaction to sequence HV1 (or other mitochondrial DNA) produces multiple sequences from one individual.
- This can be caused by the existence of *Numts*, or *nuclear DNA of mitochondrial origin*.
- Numts are thought to have arisen from the introgression of mitochondrial DNA into the nuclear genome in the past.
- From analysis of complete human and chimpanzee genomes, more than 400 Numts have been discovered. (“A Comparative Analysis of Numt Evolution in Human and Chimpanzee”, Hazkani-Covo and Graur, 2006).
- Only a small fraction of these would amplify using primers for HV1.
- Most of these Numts are shared between human and chimps, but several dozen have originated in each species after their split.
- Gorilla HV1 samples include putative Numts.

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Objectives

- We wish to develop a model *based on population genetics* for the relationship among HV1 and Numt sequences sampled from gorilla populations.
- Within the framework of this model, we wish to:
  - estimate divergence times of African gorilla populations;
  - estimate how many introgressions occurred and when they occurred in the history of the sampled data;
  - estimate effective population sizes, and evolutionary parameters such as substitution rates and transition-transversion ratio.

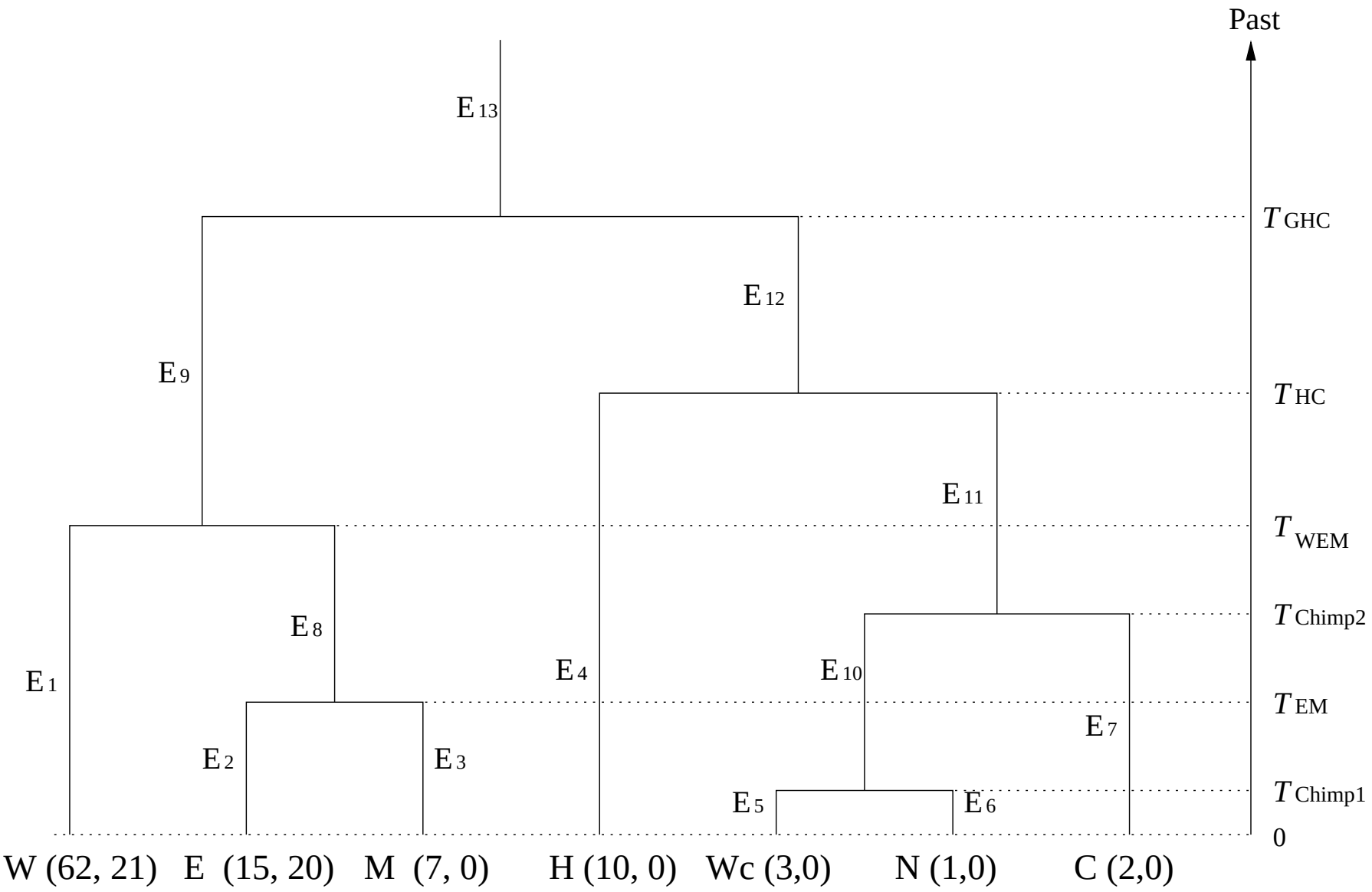
Data

- We analyze an alignment of 141 sequences with 236 sites.
- We include human and chimpanzee sequences in order to calibrate divergence times.
- There are:
  - 125 Gorilla sequences (Anthony *et al.*, 2006);
  - 10 Human sequences (sampled from Ingman *et al.*, 2000);
  - 6 Chimpanzee sequences (from Hu *et al.*, 2001; Thalmann *et al.*, 2004);

Data Table

Group	Symbol	HV1	Numt
Western lowland gorilla	W	62	21
Eastern lowland gorilla	E	15	20
Mountain gorilla	M	7	0
Western common chimpanzee	$\mathrm{W_C}$	3	0
Central common chimpanzee	C	2	0
Nigerian chimpanzee	N	1	0
Human	H	10	0
Total		100	41

The Population Tree



The Hybrid Coalescent Process

- We have a population tree relating populations.
- There are sequences of two types at the tips of the population tree.
- We model three separate processes in each edge of the tree:
  - an HV1 coalescent process with rate θ_{HV1};
  - a Numt coalescent process with rate θ_{Numt};
  - a Numt transfer process (introgression in reverse time) with rate η.
- This provides a likelihood model for a sequence tree within a population tree framework.
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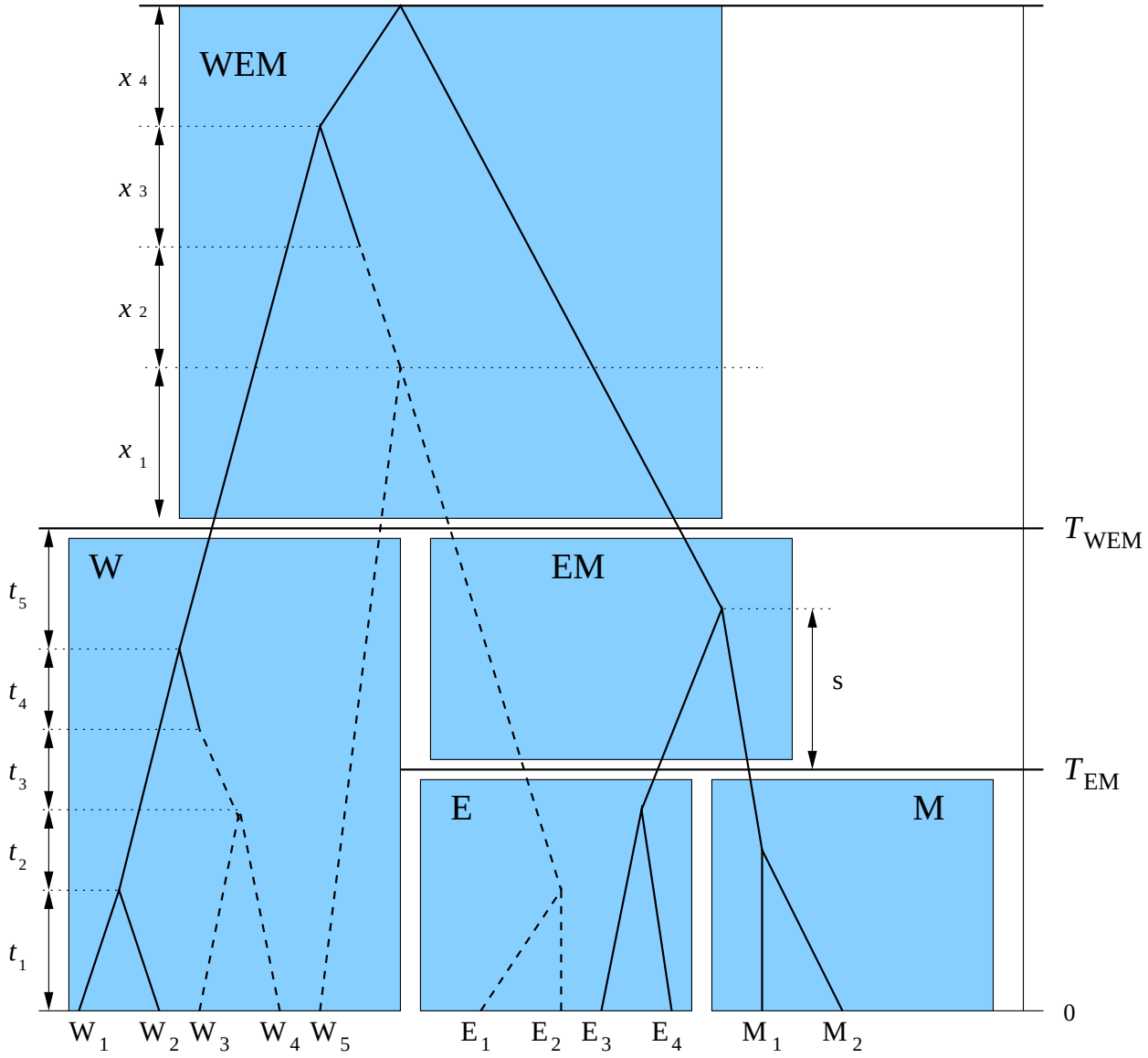
Example


WEM
W
EM
E
M
x_4
x_3
x_2
x_1
t_5
t_4
t_3
t_2
t_1
s
T_{WEM}
T_{EM}
0
W_1 W_2 W_3 W_4 W_5
E_1 E_2 E_3 E_4
M_1 M_2

Substitution Model

- Separate HKY Models for HV1 and Numts

$$Q = \mu\phi \begin{pmatrix} - & \pi_C & \kappa\pi_G & \pi_T \\ \pi_A & - & \pi_G & \kappa\pi_T \\ \kappa\pi_A & \pi_C & - & \pi_T \\ \pi_A & \kappa\pi_C & \pi_G & - \end{pmatrix}$$

where

- π_{A}, π_{C}, π_{G}, π_{T}: relative frequencies for each nucleotide base.
- κ is the transition-transversion parameter.
- $\phi = 1/(2(\kappa(\pi_{\mathrm{A}}\pi_{\mathrm{G}} + \pi_{\mathrm{C}}\pi_{\mathrm{T}}) + \pi_{\mathrm{R}}\pi_{\mathrm{Y}}))$, a scaling parameter.
- μ is the number of substitutions per site per million years.

Bayesian Estimation of Parameters

- State space ($\boldsymbol{T}$, Θ, G)
  - $\boldsymbol{T}$=(T_{EM}, T_{WEM}, T_{Chimp1}, T_{Chimp2}, T_{HC}, T_{GHC}).
  - Θ=(θ_1,..., θ_{13},λ_θ, η, μ_{HV1}, μ_{Numt}, κ_{HV1}, κ_{Numt}).
  - G= Gene genealogy determined by the hybrid coalescent process.
- Target distribution

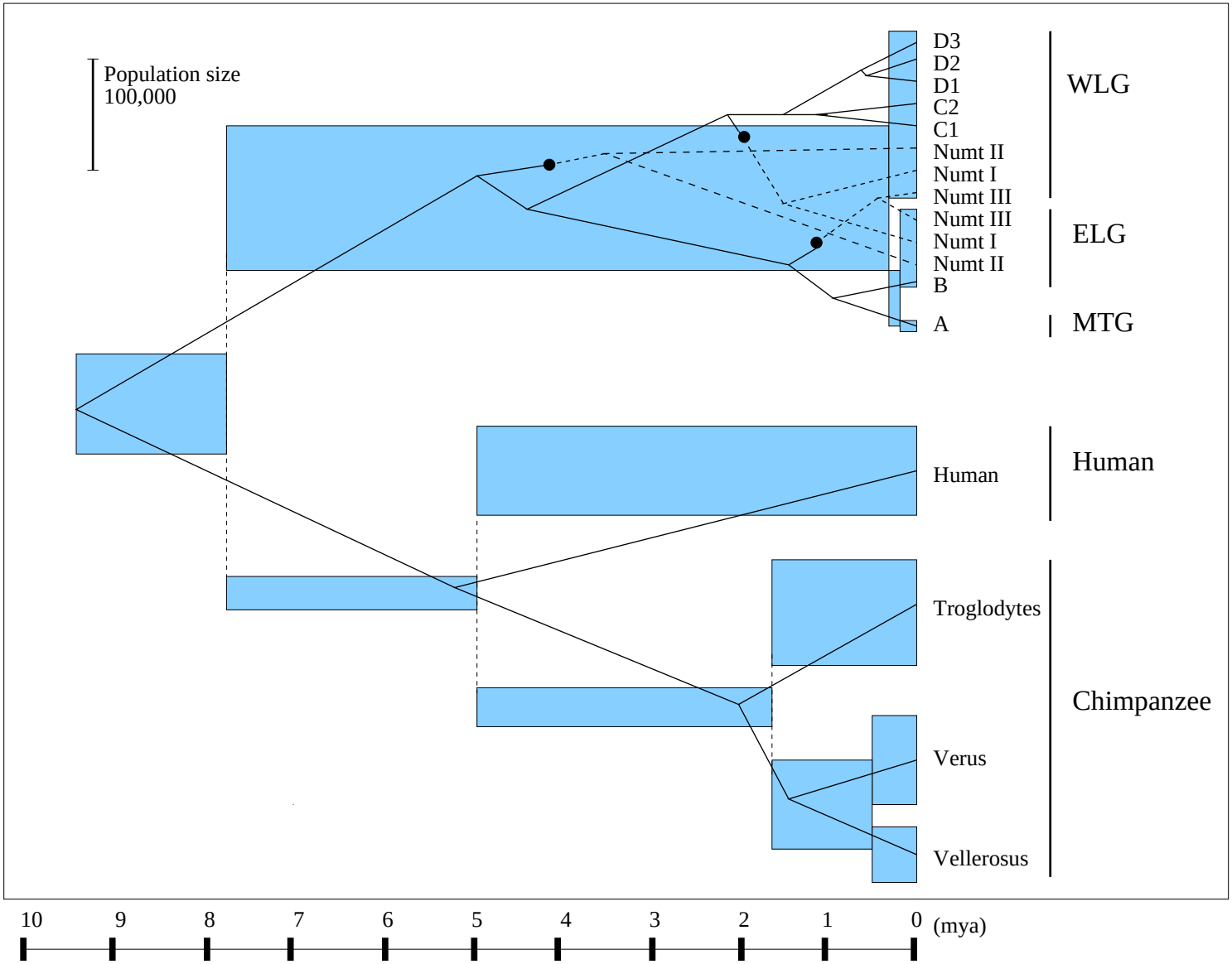
$$f(\boldsymbol{T}, \Theta, G \mid D) = \frac{f(D \mid \boldsymbol{T}, \Theta, G) f(G \mid \boldsymbol{T}, \Theta) f(\boldsymbol{T}) f(\Theta)}{\int_{(\boldsymbol{T}, \Theta, G)} f(D \mid \boldsymbol{T}, \Theta, G) f(G \mid \boldsymbol{T}, \Theta) f(\boldsymbol{T}) f(\Theta)}$$

- Use MCMC over this space.

MCMC Approaches

- Update population divergence time $\boldsymbol{T}$ and gene genealogy G:
  - Multiply a constant to the whole trees.
  - Update population divergence times.
  - Update histories in a population edge.
    - Generate new event times in a population edge.
    - Relocate an event in a population edge.
    - Change a pair of coalescent events.
  - Update the total number of transfer events
- Update parameters in Θ:
  - Propose a new value by multiplying a scale factor from Gamma(2,2) distribution to the current value.

Results (100 HV1 + 41 Numt Sequences)



Robustness?

- The analysis of the full data set implies a fairly recent divergence time between eastern and western gorillas (95% credible region 110,000 to 610,000 million years ago).
- However, the earliest east/west coalescents are among Numt sequences.
- How robust are the estimated times if the Numt data is removed?

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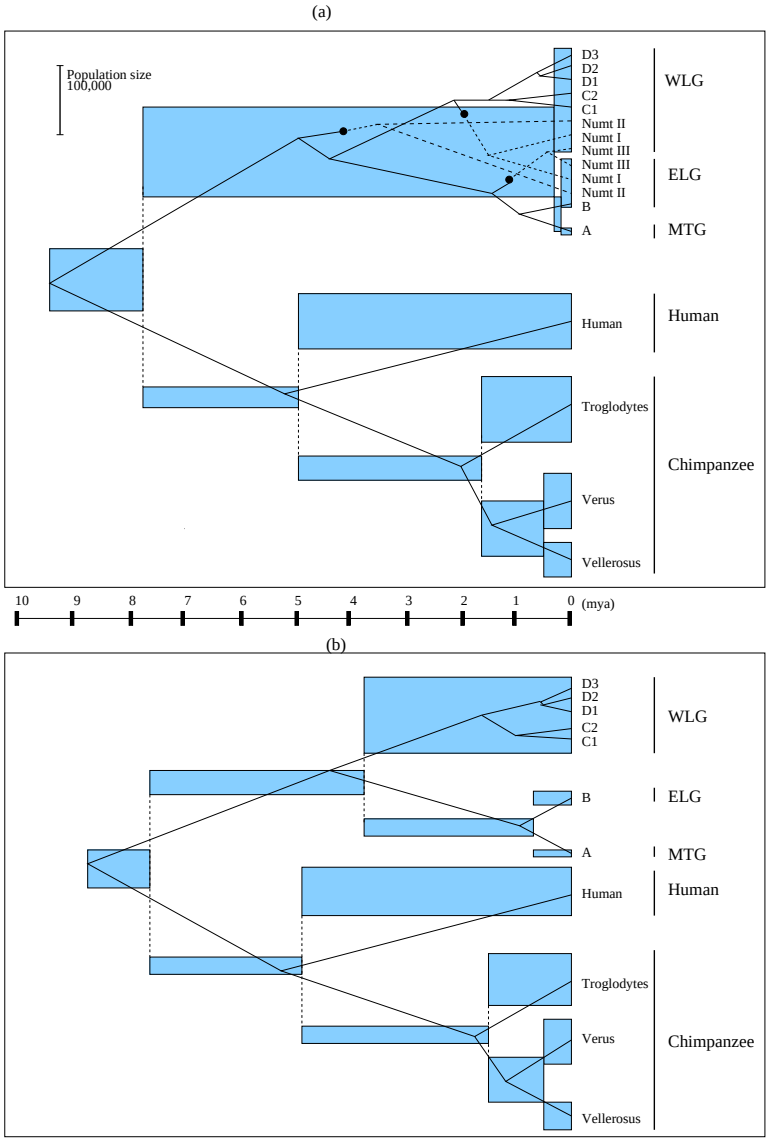
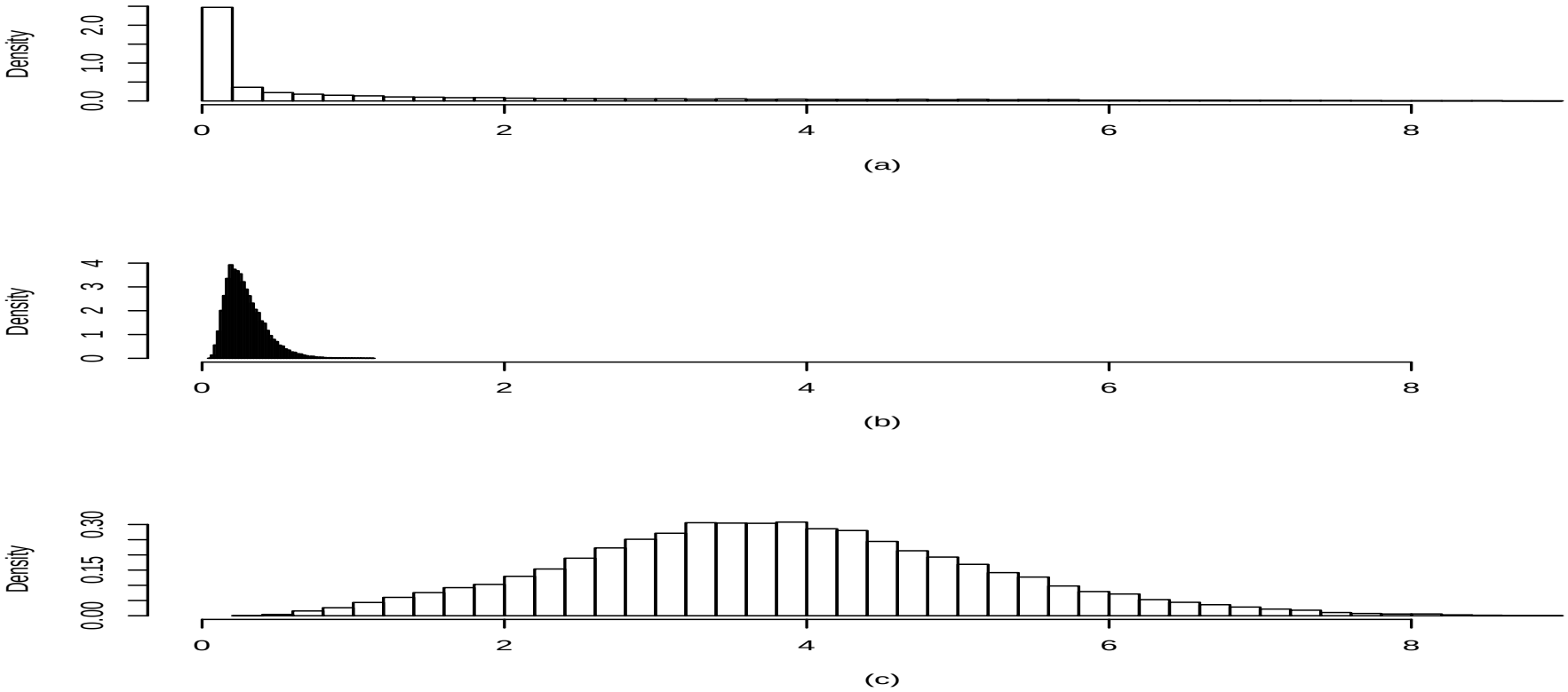
HV1 + Numt (top) versus HV1 only (bottom)



Estimated Population Divergence Times

Time	Numt-HV1 case			only-HV1 case		
	mean	median	95% C.R.	mean	median	95% C.R.
T_{EM}	0.190	0.170	(0.045, 0.430)	0.67	0.63	(0.14, 1.50)
T_{WEM}	0.29	0.26	(0.11, 0.61)	3.8	3.8	(1.3, 6.7)
T_{Chimp1}	0.53	0.38	(0, 1.8)	0.52	0.38	(0, 1.7)
T_{Chimp2}	1.60	1.60	(0.26, 2.90)	1.50	1.50	(0.36, 2.70)
T_{HC}	4.9	4.8	(4.0, 5.9)	4.9	4.9	(4.0, 5.9)
T_{GHC}	7.7	7.9	(5.4, 9.0)	7.6	7.8	(5.3, 8.9)

East-West Split Time Posterior Distributions



Conjecture

- What is a biologically plausible reason for the discordance in the results?
- We conjecture that *differences in male and female gorilla migratory behavior* could be the reason for the difference.
- If there is some male migration between east and west long after female migration ends, then we would expect that nuclear genes would coalesce much more recently than mitochondrial genes.
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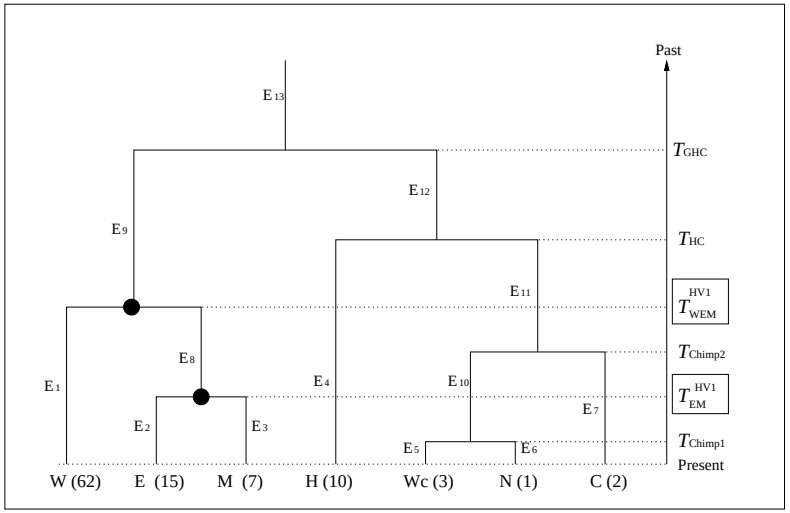
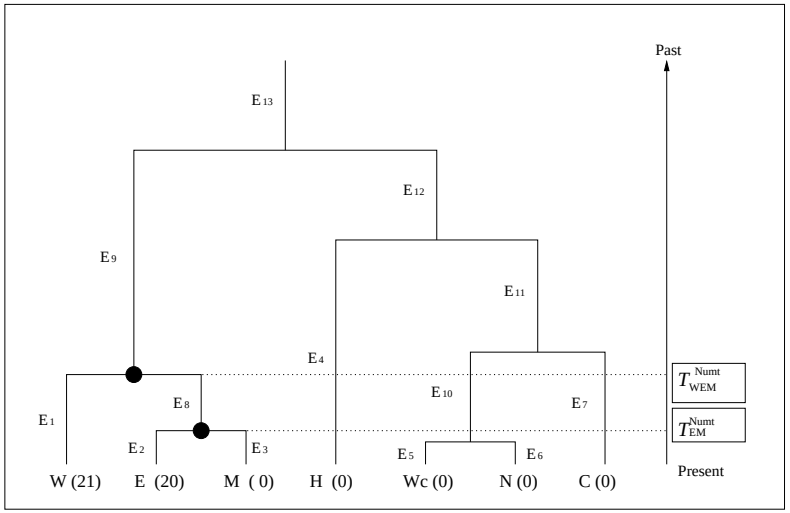
A Genome-differentiated Population Tree Model

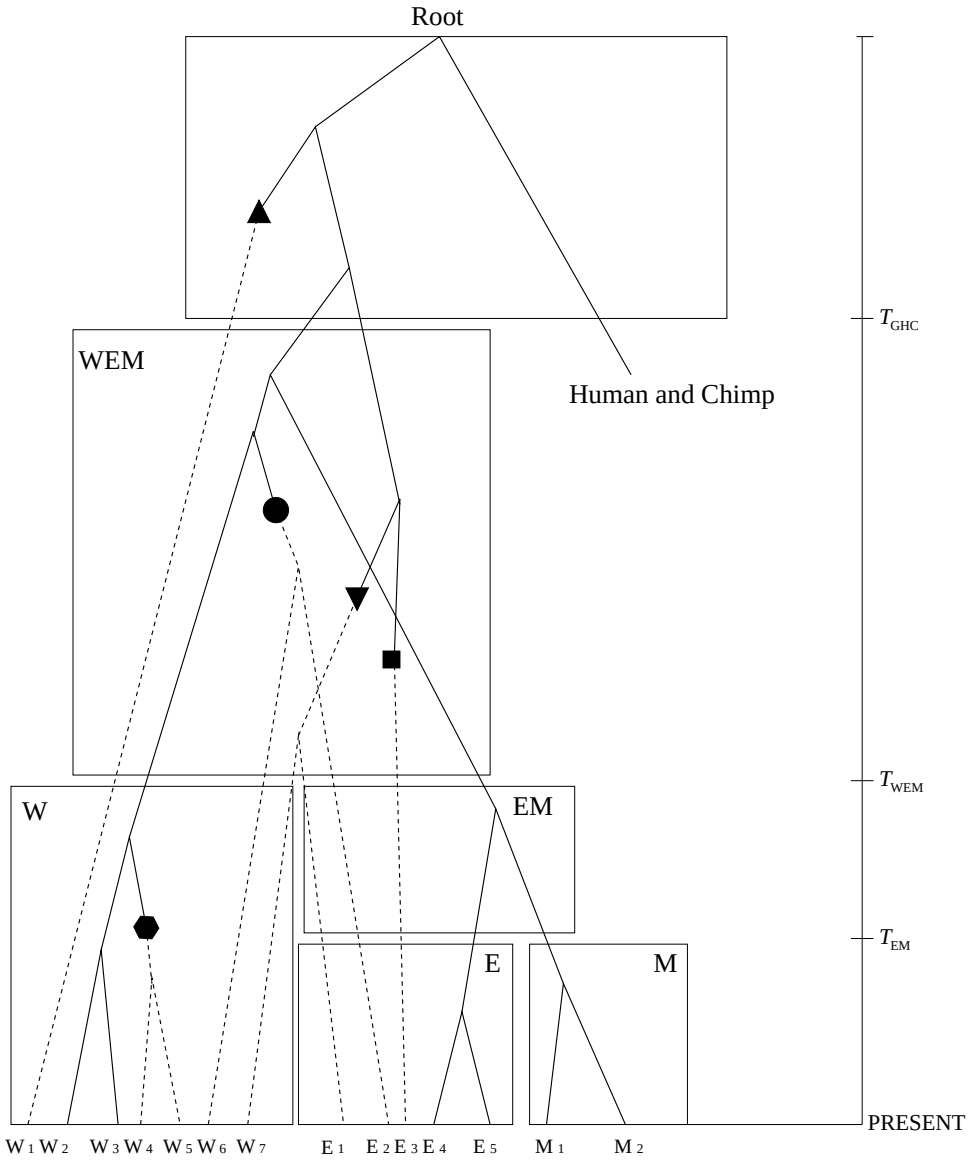
(a) HV1 Population Tree



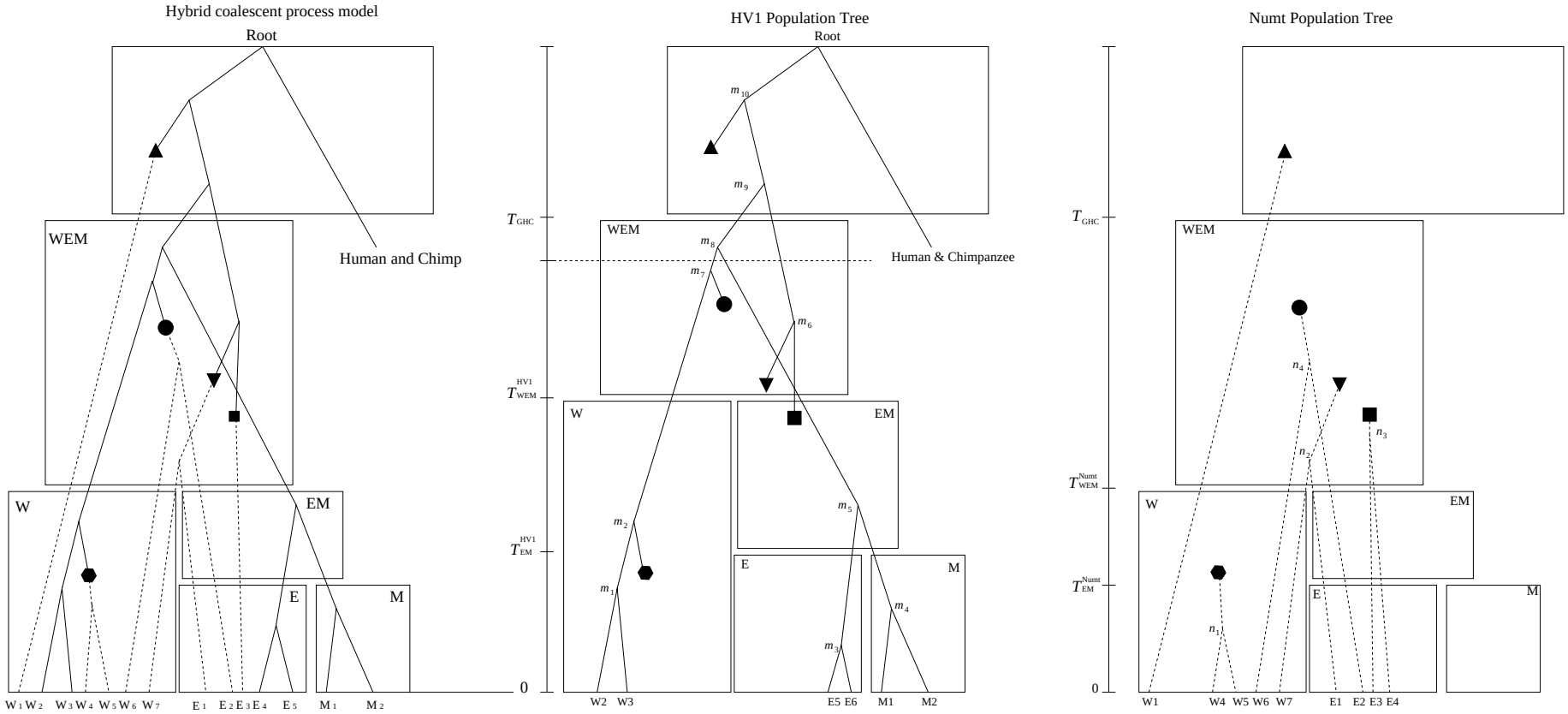
(b) Numt Population Tree



Example Gene Genealogy (old)



Example Gene Genealogy (old and new)



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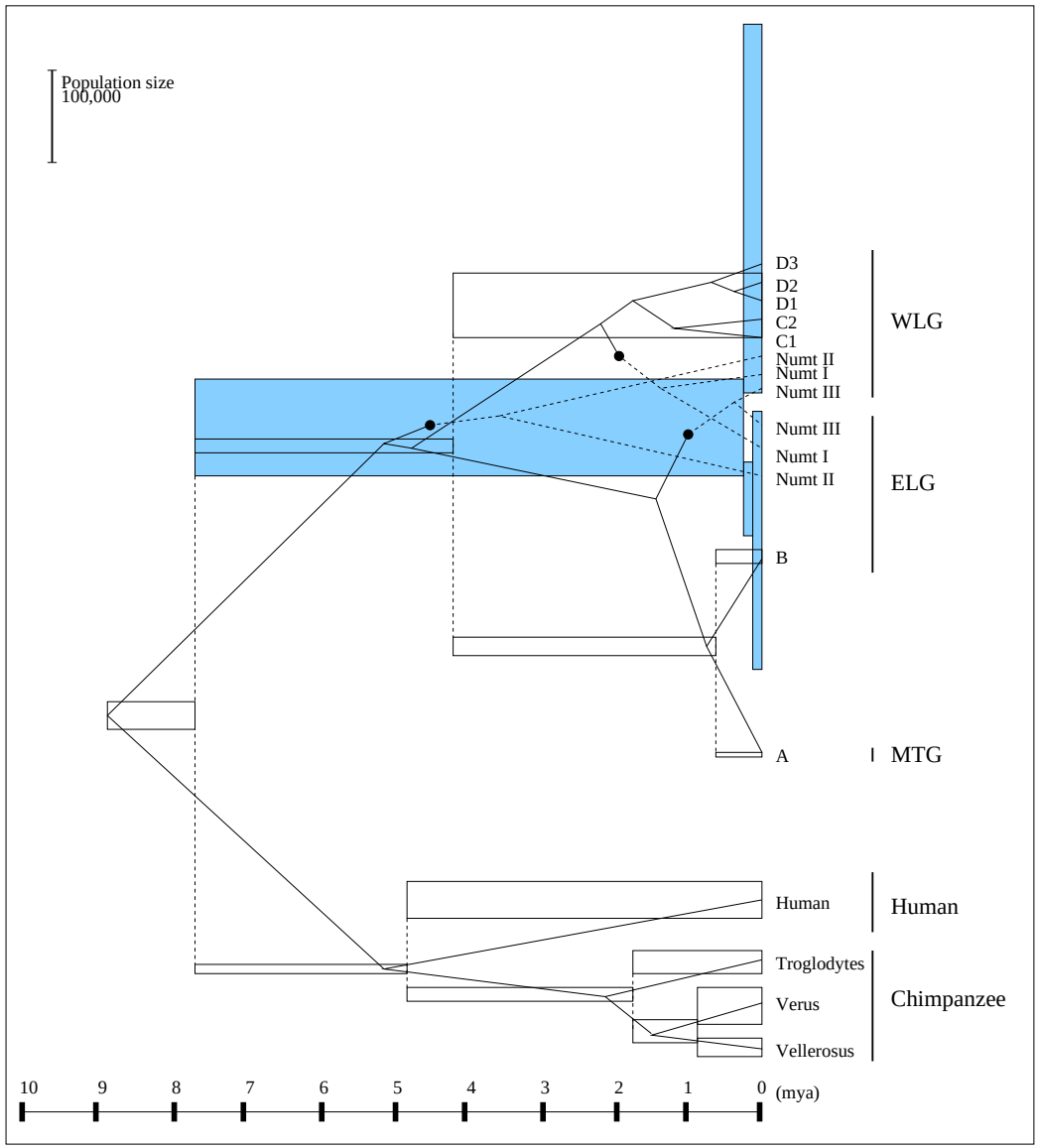
  - Θ=($\left\{\theta_i^{\mathrm{HV1}}\right\}$, $\left\{\theta_j^{\mathrm{Numt}}\right\}$, λ_{HV1}, λ_{Numt}, η, μ_{HV1}, μ_{Numt}, κ_{HV1}, κ_{Numt})
  - G= Gene genealogy over the genome-differentiated population trees.
- Target distribution

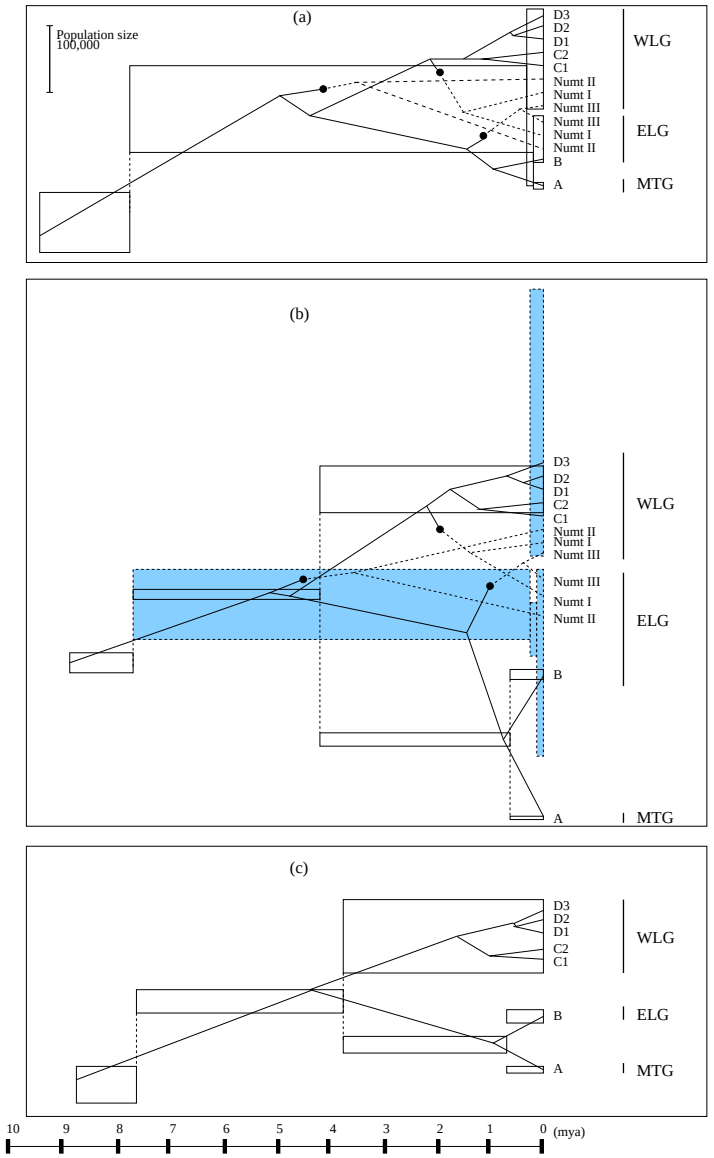
$$f(\boldsymbol{T}, \Theta, G \mid D) = \frac{f(D \mid \boldsymbol{T}, \Theta, G) f(G \mid \boldsymbol{T}, \Theta) f(\boldsymbol{T}) f(\Theta)}{\int_{(\boldsymbol{T}, \Theta, G)} f(D \mid \boldsymbol{T}, \Theta, G) f(G \mid \boldsymbol{T}, \Theta) f(\boldsymbol{T}) f(\Theta)}$$

- MCMC approaches: *more complex than the hybrid coalescent process model*

Summary


Population size
100,000
D3
D2
D1
C2
C1
Numt II
Numt I
Numt III
WLG
Numt III
Numt I
Numt II
ELG
B
A
MTG
Human
Human
Troglodytes
Verus
Vellerosus
Chimpanzee
10
9
8
7
6
5
4
3
2
1
0
(mya)

Comparison


(a)
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D3
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B
A
WLG
ELG
MTG
(b)
D3
D2
D1
C2
C1
Numt II
Numt I
Numt III
Numt III
Numt I
Numt II
B
A
WLG
ELG
MTG
(c)
D3
D2
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10 9 8 7 6 5 4 3 2 1 0 (mya)

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T_{Chimp1}	0.86	0.83	(0.0034, 2.0)
T_{Chimp2}	1.8	1.8	(0.75, 3.00)
T_{HC}	4.8	4.8	(4.0, 5.9)
T_{GHC}	7.7	7.9	(5.4, 9.0)
T_{EM}^{Numt}	0.15	0.12	(0.0024, 0.45)
T_{WEM}^{Numt}	0.25	0.23	(0.053, 0.61)

Summary

- Discordant east-west split times for Numt and HV1 sequences.
- HV1 east-west split time may predate the Pleistocene.
- Numt east-west split time probably falls within the Pleistocene.
- Male-mediated gene-flow may have persisted much longer after female east/west migration stopped.
- There are likely three separate Numt loci in this data set.

Future work

- Use more human data to better estimate HV1 substitution rates and parameters.
- Extend the current population tree to incorporate real gorilla populations.
- Topological uncertainty in the population trees.
- Changes in population sizes over time.
- Explicit migration in a population tree.
- Multiple loci DNA data.
- Subdivisions in each Numt population.
- Recombination in Numt sequences.

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  - She and co-workers collected many hair samples from the field;
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