

The 3D Organization of Chromatin Explains Evolutionary Fragile Genomic Regions by

Camille Berthelot, Matthieu Muffato,
Judith Abecassis and Hugues Roest Crollius

Speaker: Ilia Minkin

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Two types of genome alterations

1. Small point mutations:

ACTTG
AGT-G

Two types of genome alterations

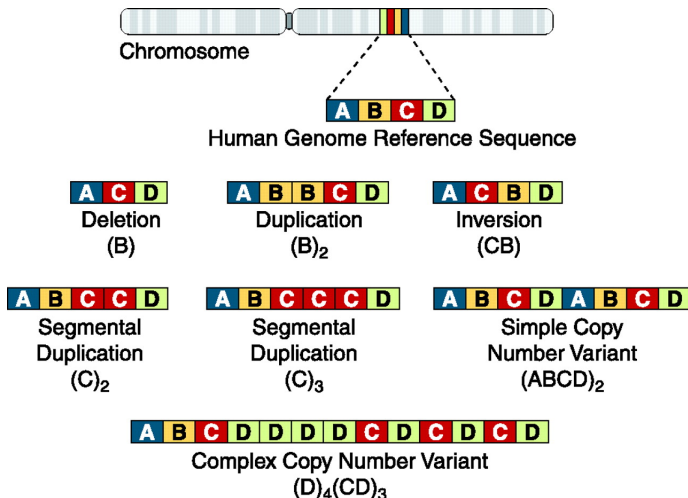
1. Small point mutations:

ACTTG
AGT-G

2. Large rearrangements:

- ▶ Inversions
- ▶ Transpositions
- ▶ Fusions
- ▶ ...

Genome Rearrangements



Source: *Dierssen et al, 2009*

Motivation

Rearrangements:

- ▶ Are a major driving force in evolution
- ▶ Play large role in diseases (e.g. cancer)

Known mechanisms:

- ▶ Non-homologous end joining
- ▶ Non-allelic homologous recombination
- ▶ Replication fork stalling
- ▶ ...

The Big Question

Are rearrangements more likely to happen in one parts of a genome than the others?

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Two hypotheses:

1. Rearrangements are distributed uniformly
2. Some regions are more likely to be disrupted

A Short Survey

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The story is to be continued...

The Study

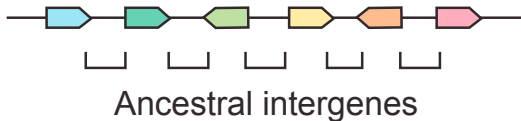
Two questions:

- ▶ Do fragile regions exist?
- ▶ If they do, what is cause of fragility?

A note: fragility is not "physical", it only means higher possibility of rearrangements

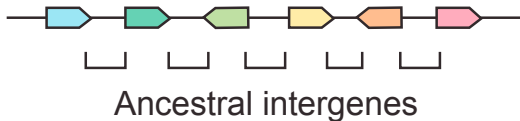
Genomes Representations

Genomes are sequences of gene markers that are unbreakable:

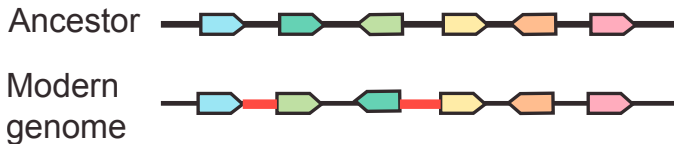


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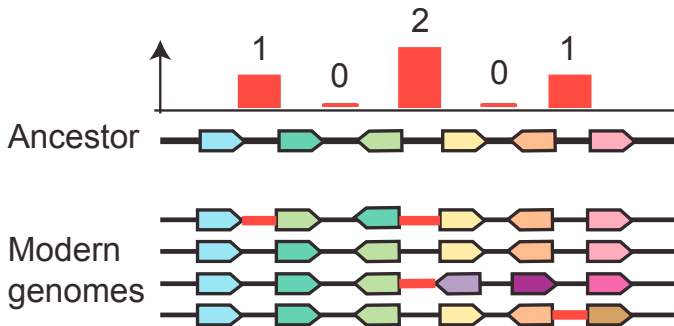


Here red dashes are breakpoints:



Methodology

Suppose that we have an ancestral genome and its successors



How does ancestral intergene length affects its breakage rate?

Methodology

Null hypothesis: breakpoint density is uniform

As intergene length \uparrow , # of breakpoints \uparrow as well

It yields Poisson distribution of breakage rate

Methodology

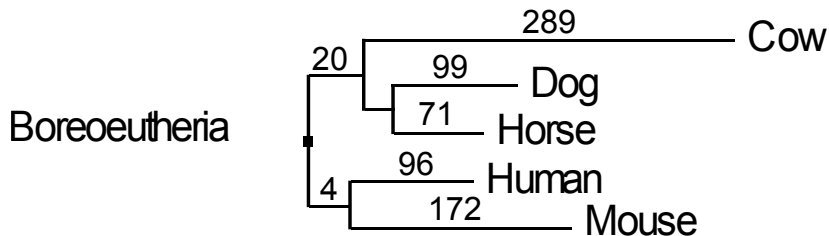
Boreoeutheria: the last common ancestor of primates, rodents, and laurasiatherians

Stages of the study:

- ▶ Reconstruct gene order of Boreoeutheria
- ▶ Annotate ancestral intergenes
- ▶ Identify breakpoints w.r.t. human, mouse, dog, cow and horse
- ▶ Do Poisson regression of "breakage rate"

Expect linear law if the null hypothesis is true






The Phylogenetic Tree



Intergene Annotation

CNEs – conservative non-coding elements

Characteristics of
ancestral intergenes

	Length	%GC	%CNE
	X_1	Y_1	Z_1
	X_2	Y_2	Z_2
	X_3	Y_3	Z_3
	X_4	Y_4	Z_4
	X_5	Y_5	Z_5

The Result

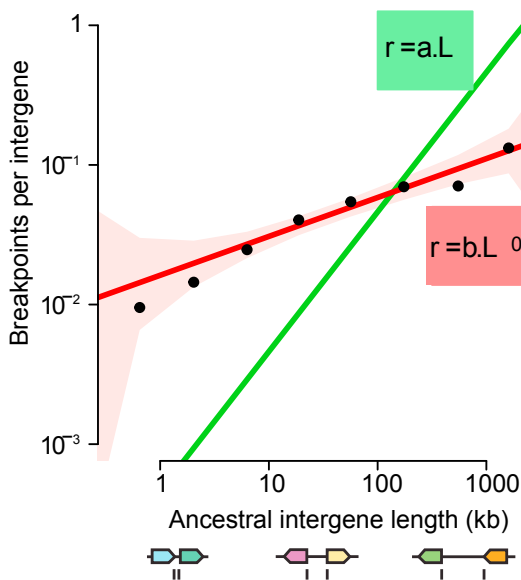


Table 1. Coefficients and Statistics of Poisson Regression Models Describing the Average Number of Breakpoints per Intergene as a Function of Intergene Length, %GC, and %CNE

	Coefficients		P(> z)	Null Deviance (df)	Residual Deviance (df)	Goodness of Fit		Pseudo R ²
	Simple Regression	Stepwise Regression				c ² p value	Stepwise c ² p value	
Model 1: length only								
Intergene length	0.28	—	< 2.10 ^{À16a}	167.3 (10)	12.4 (9)	0.19 ^a	—	0.93 ^a
Model 2: length + %GC								
Intergene length	0.26	0.27	< 2.10 ^{À16a}	137.8 (28)	25.7 (27)	0.53 ^a	—	0.81 ^a
%GC	—	0.003	0.44	137.8 (28)	25.1 (26)	0.52	0.42	0.82
Model 3: length + %CNE								
Intergene length	0.28	0.30	< 2.10 ^{À16a}	179.2 (19)	26.3 (18)	0.09 ^a	—	0.85 ^a
%CNE	—	À4.55	0.01 ^a	179.2 (19)	20.7 (17)	0.24 ^a	0.02 ^a	0.88 ^a
Simulation: 3D contacts in open chromatin								
Intergene length	0.28	—	< 2.10 ^{À16}	253.8 (14)	29.6 (13)	0.005	—	0.88

A parameter significantly affecting the breakage rate has a regression coefficient statistically different from 0 (P(> |z|) < 0.05). The goodness of fit of each model is assessed by a c² test on the residual deviance and degrees of freedom (i.e., likelihood ratio test): a non-significant p value means that the residual deviance may be attributed to statistical noise. The effect of an additional parameter on the fit is assessed by a c² test on the difference in residual deviances and degrees of freedom with and without the parameter: a significant p value means that the fit is significantly better with the additional parameter. The pseudo R² corresponds to McFadden's pseudo R² (proportion of null deviance explained by the model).

For methods, see the [Supplemental Information](#).

^aValues indicative of an improvement in the model.

How to Explain the Equation?

$$r = 2.410^{-3} \times L^{0.28}$$

Intergene length explains 93% of variation in breakpoint occurrence

Short intergenes are more breakable than under pure random model, while longer ones are less

Why?

Is GC Content has any Influence?

GC content strongly correlates with gene density

Maybe GC content can be a physical explanation?

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Added GC content in regression — got a non-significant coefficient

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Are CNEs Affect Fragility?

CNEs – conservative non-coding elements

Logic: regulative elements may affect genes that are nearby

Disrupting synteny between CNEs and genes may have impact on rearrangements that we observe

Do they?

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Do they?

Not that much

Added CNE rate in regression — got a significant coefficient, improved explanation rate only by 3%

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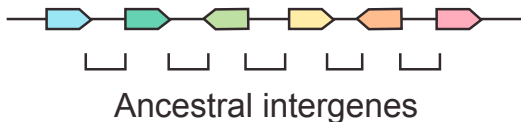
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Inversions within Intergenes

A reminder: we work with gene markers \Rightarrow see only rearrangements disrupting their order



Regression showed that longer intergenes have smaller breaks than expected

What if really missing breakpoints within long intergenes?

Inversions within Intergenes

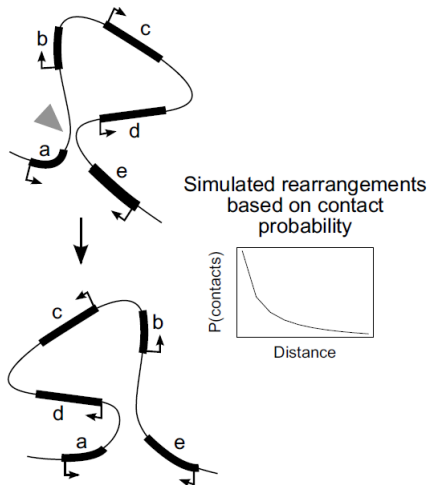
Solution: simulate rearrangements, select detectable ones and compare with the real data

If bias exists, the results should be very close

Rearrangements have been shown to occur between regions in close 3D proximity in the nucleus

Contact probability is a good proxy for rearrangement probability

Simulation

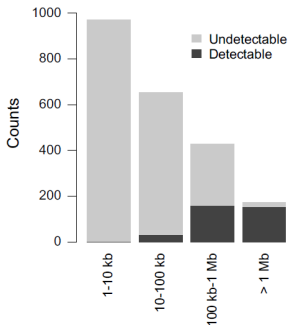


Inversions are simulated in the human genome (gray arrow) based on the probability of 3D DNA contacts experimentally derived from Hi-C studies (right inset).

Simulation Result

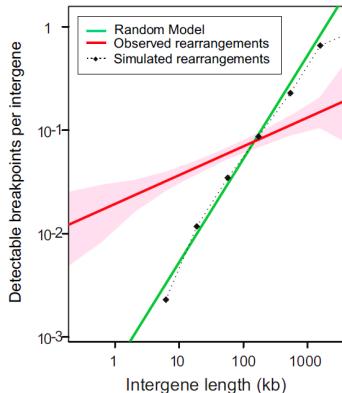
B

Lengths of simulated inversion segments



C

Rearrangements between regions in contact



Conclusion: non-detected rearrangements do not introduce enough bias

Open Chromatin is the Culprit

Stick with the simulation – restrict rearrangements to only **open chromatin** regions

ENCODE published chromatin state profiles

The study used four different cell types

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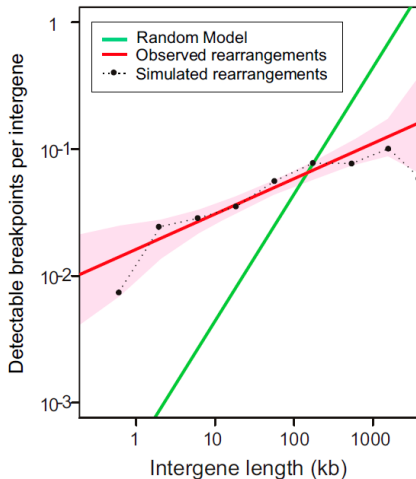
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Voilà – simulation coincides with the model!

Simulation Result

F

Rearrangements between
open chromatin regions in contact



Conclusion

This slide is under construction, please visit later

Thank you!