In silico methods for genome rearrangement analysis: from identification of common markers to ancestral reconstruction

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Sous la direction de :
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Comparative genomics

What ?

Study of genome structure by the comparison of different species

Why ?

- Understanding function processes
- Understanding evolution processes

How ?

Huge amount of data in contemporary genomes

- Automation of analyses
- Development of *in silico* methods

Interests ?

- Short-term : scientific answers
- On the long run : medicinal or therapeutic solutions

Comparative genomics

Genetic material comparison : DNA

- **First approach : study of local mutations in genes**
  Gene comparison by sequence alignment

```
sequence 1 : CAGCA-CGTGGATTCTCGG
               | ||  ||||| ||  |
sequence 2 : TATCAGCGTGG-CACTAGC
```

Observation (Palmer & Herbon 1988)

The major part of *Bassica oleracea*'s and *Bassica canpestris*'s genes is quasi identical (99%), but only the gene order differ significantly.

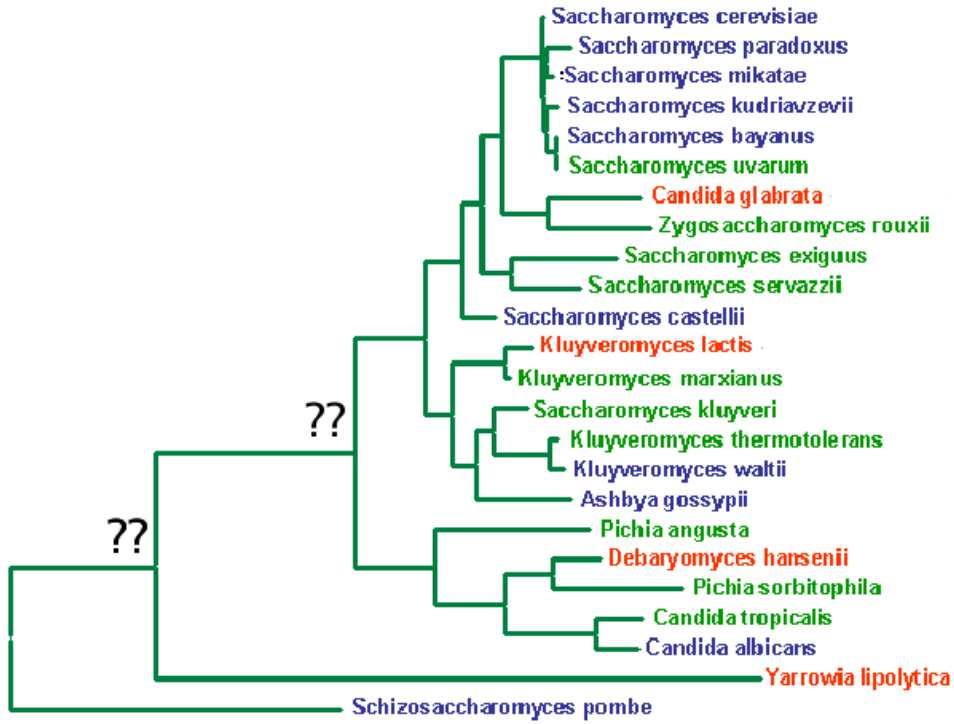
- **Second approach : study of genomic rearrangements**
  Whole genome architecture comparison by gene order and content study

Comparative genomics

- **Challenge** : emitting hypotheses on the history of contemporary genomes and the general mechanisms of their formation

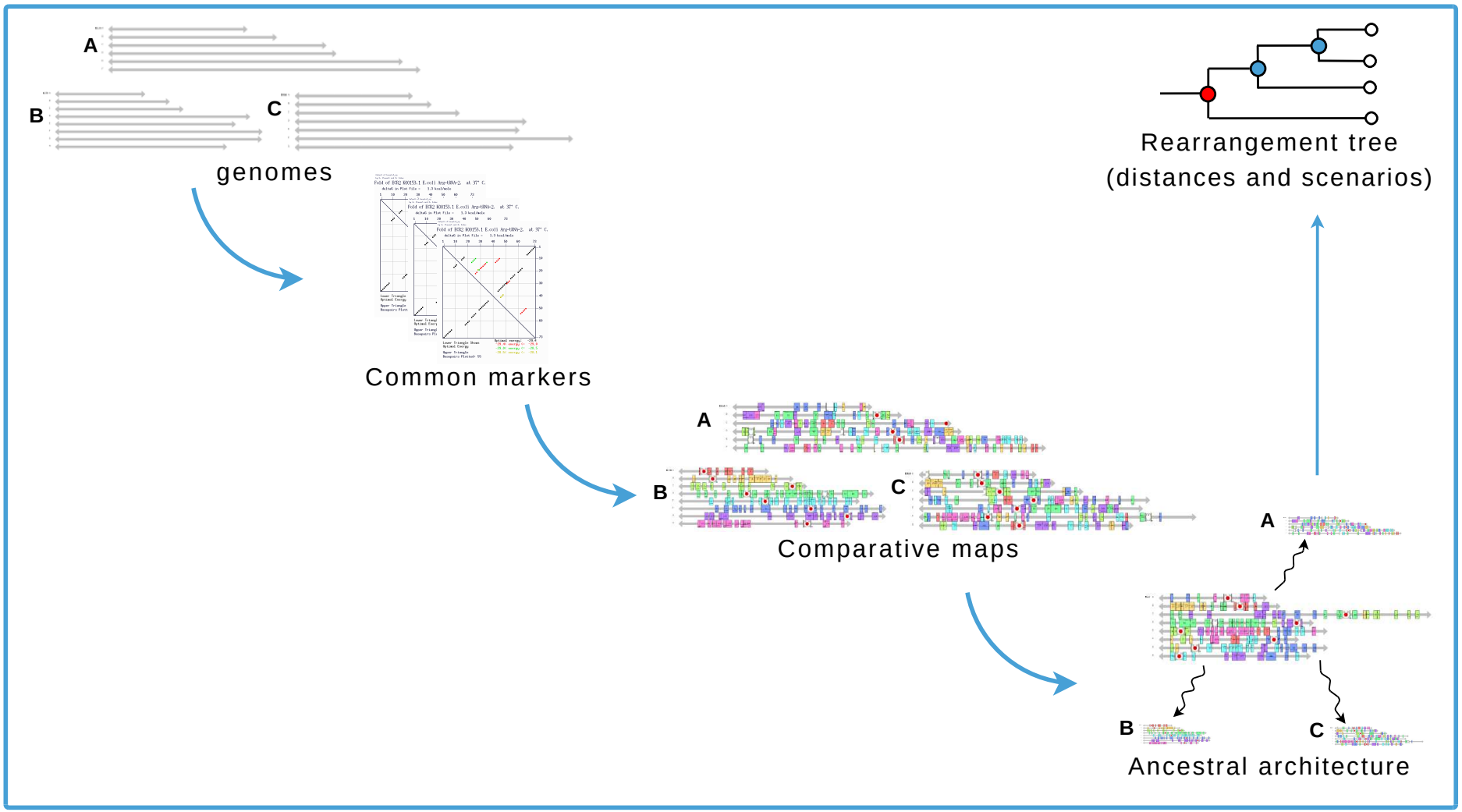
- **Problem** : impossibility of knowing with certainty the architecture of the common ancestral genomes

- **Solution** : developing methods for constructing a plausible architecture of ancestral genomes



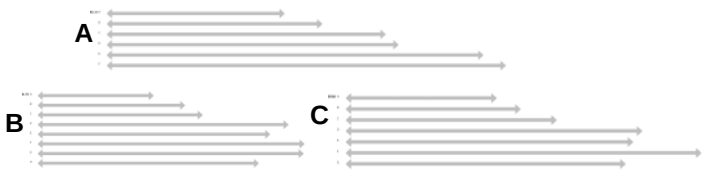
From common markers to ancestral reconstruction

Constructing ancestral architecture from the comparison of contemporary genomes requires 3 basic steps :

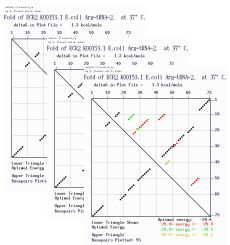


In this thesis

Combinatorial and algorithmic approach

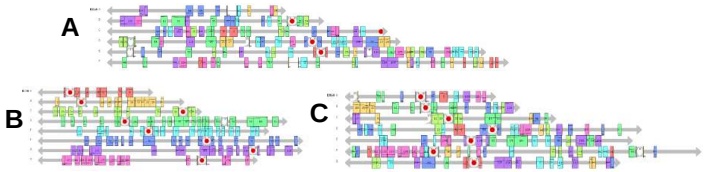


genomes



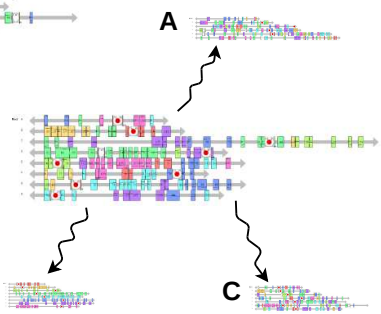
Common markers

Orthology and synteny study:
SyDiG algorithm (in prep)

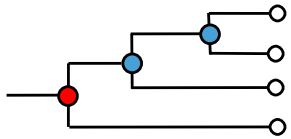


Comparative maps

Partial reconstructions of a median genome:
Super-block method (Jean et al., JCB, revision)



Ancestral architecture



Rearrangement tree
(distances and scenarios)

- Computation of a parsimonious scenario (Jean et al., IPL, 2007)
- Interative vizualization of scenarios: VIRAGE tool
(Jean et al. , ESF EMBO, 2007)

Chromosomal rearrangements in Yeasts

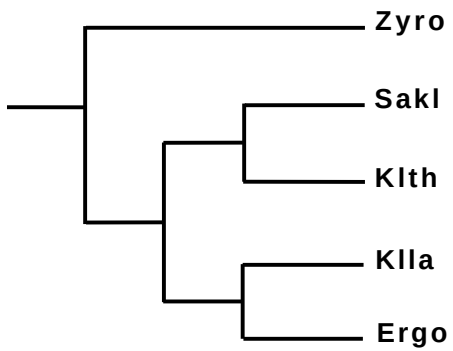
Is it **possible** to study ?

- Uniqueness of Génolevures data : complete genome sequences, availability of protein families
- Hemiascomycete yeasts : weak redundancy, synteny
- Additional information : positions of centromeres

⇒ We can specify markers for Hemiascomycetes

Hemiascomycete genomes

Species	Mnemonic
Kluyveromyces thermotolerans	Klth
Ashbya gossypii	Ergo
Kluyveromyces lactis	Klla
Saccharomyces kluyveri	Sakl
Zygosaccharomyces rouxii	Zyro



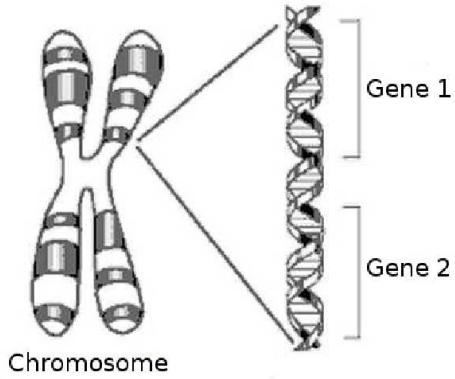
Cladogram

Outline

- Modeling a genome and evolutionary mechanisms
- Partial ancestral reconstructions : super-block method
- Constructing and visualizing parsimonious scenarios
- Conclusion and perspectives

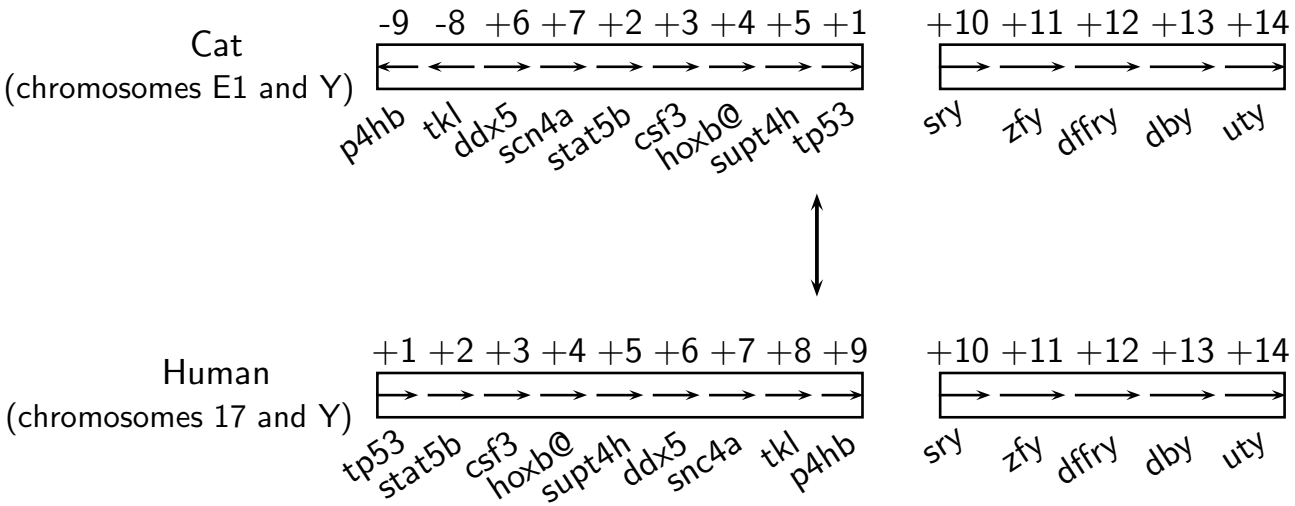
Signed permutation model

- **Gene** : functional unity composed of DNA
- **Chromosome** : sequence of genes
- **Genome** : set of chromosomes



Model :

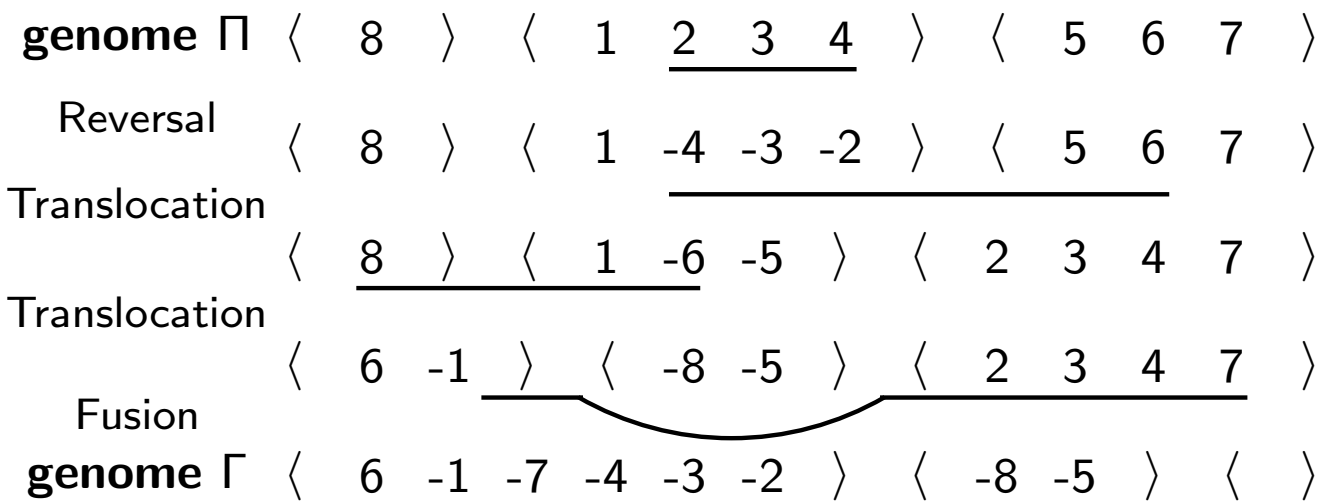
- **Marker (a gene or a set of genes) :** signed ordinal
- **Chromosome :** signed permutation
- **Genome :** set of signed permutations



Rearrangement operations

- Evolution mechanisms :

- Mathematical operations on permutations :



Distance and scenario

Two closely related problems :

- **Rearrangement distance** $d(\Pi, \Gamma)$:
  minimum number of rearrangements that transform Π into Γ
- **Parsimonious rearrangement scenario** :
  sequence of rearrangements respecting rearrangement distance

Distance and scenario

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Π : $\langle 8 \rangle \langle 1 \ \ 2 \ \ 3 \ \ 4 \rangle \langle 5 \ \ 6 \ \ 7 \rangle$

$d(\Pi, \Gamma) = 4$

Γ : $\langle 6 \ \ \text{-}1 \ \ \text{-}7 \ \ \text{-}4 \ \ \text{-}3 \ \ \text{-}2 \rangle \langle 5 \ \ 8 \rangle \langle \rangle$

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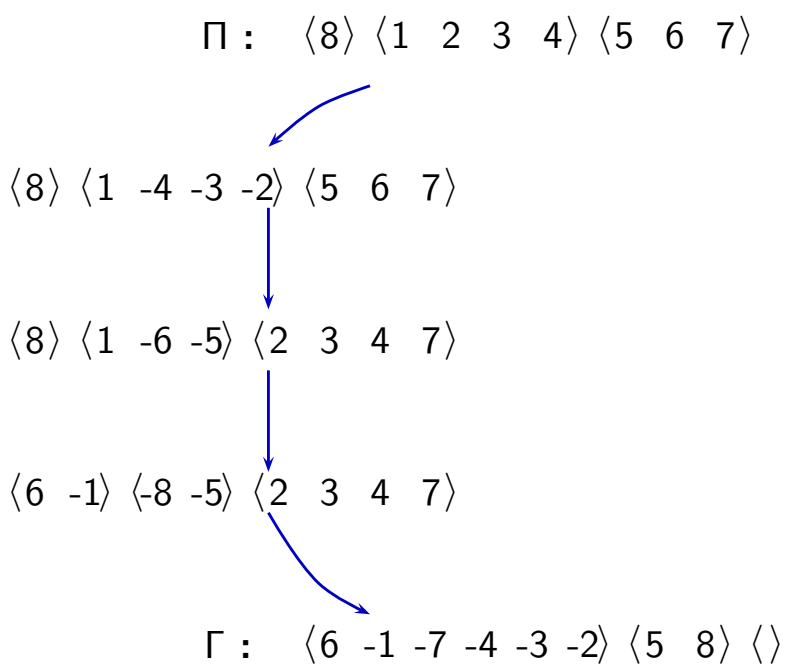

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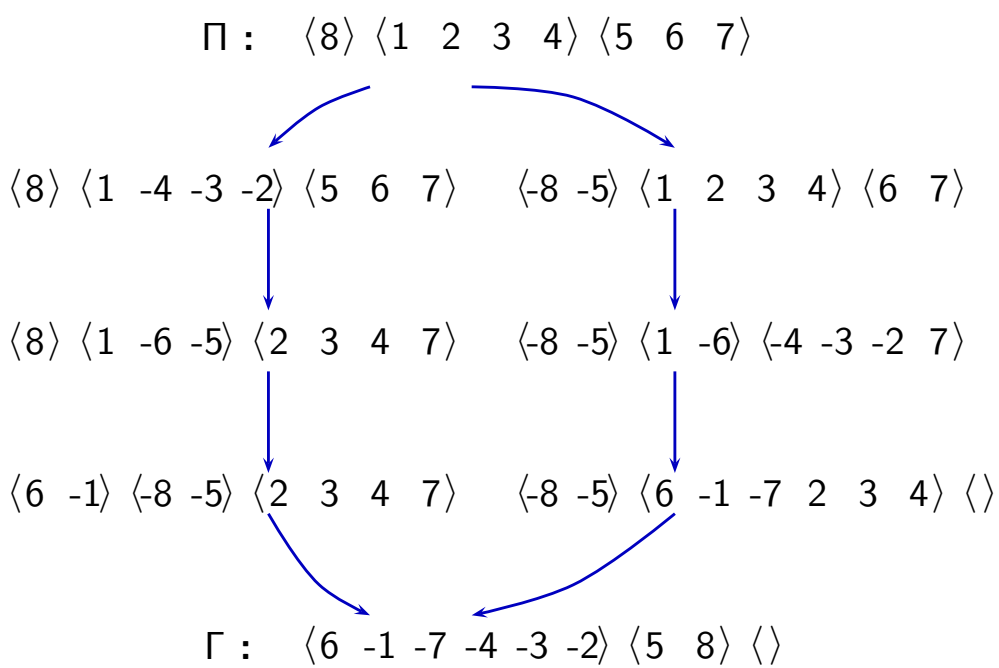

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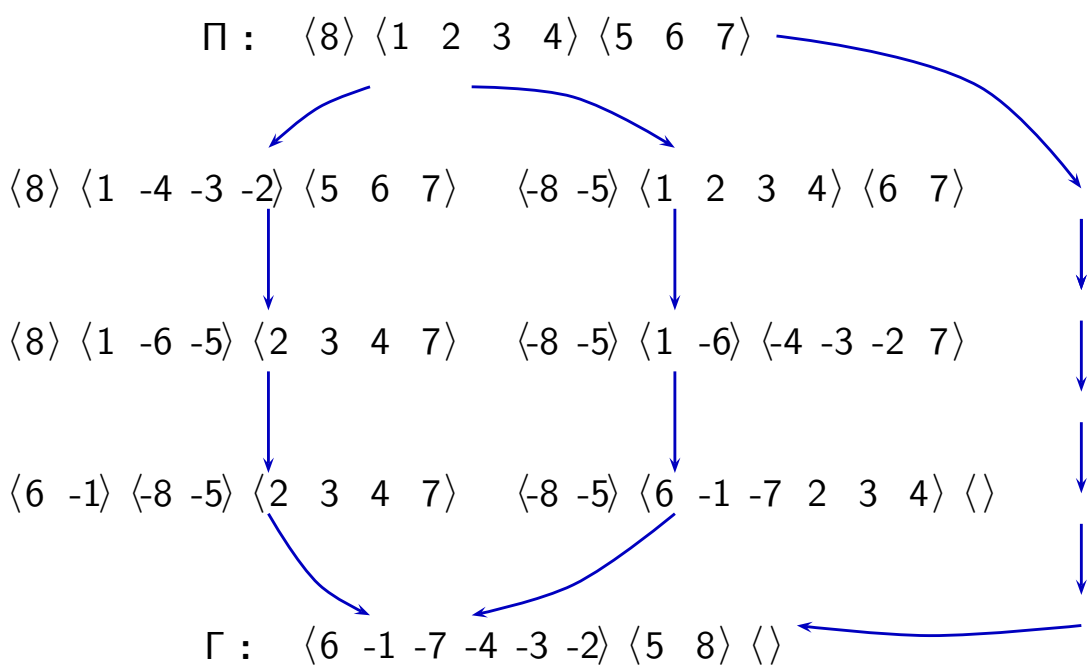

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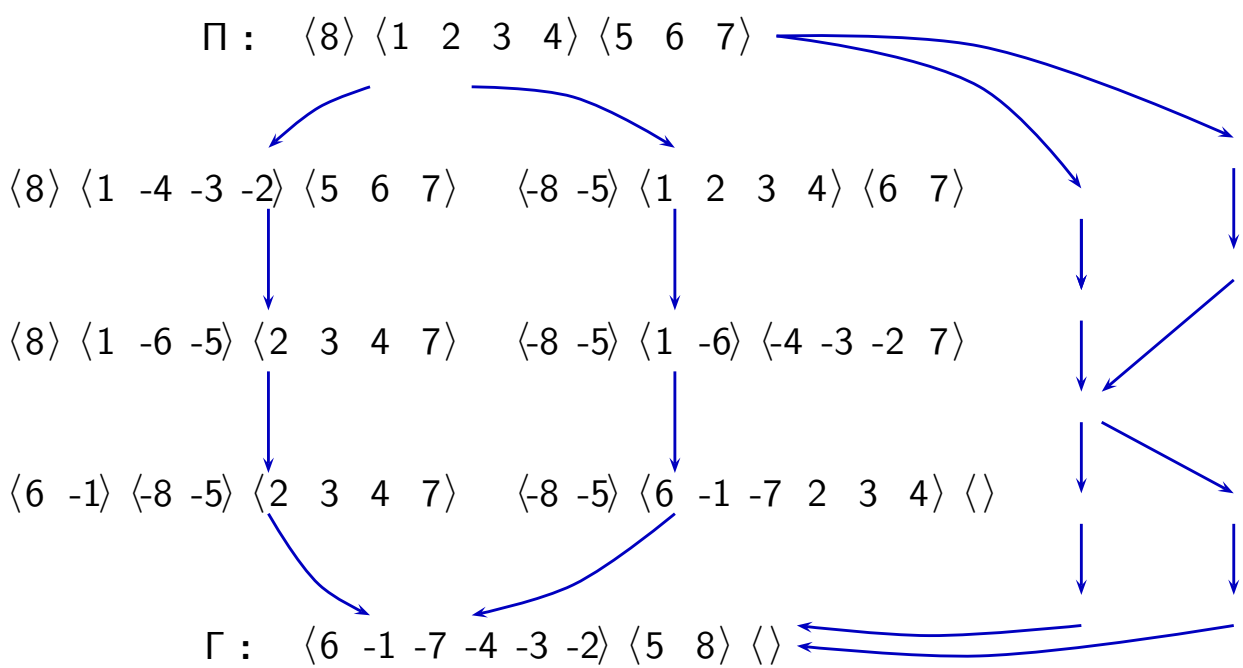

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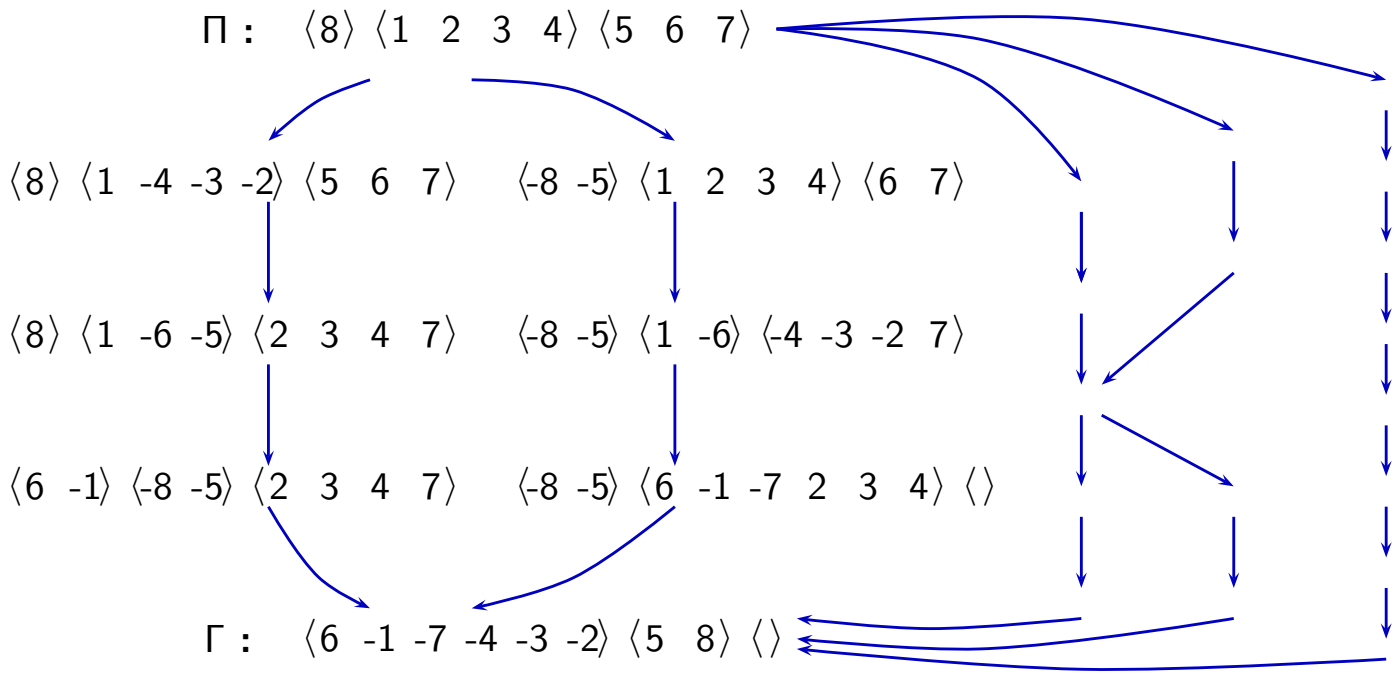

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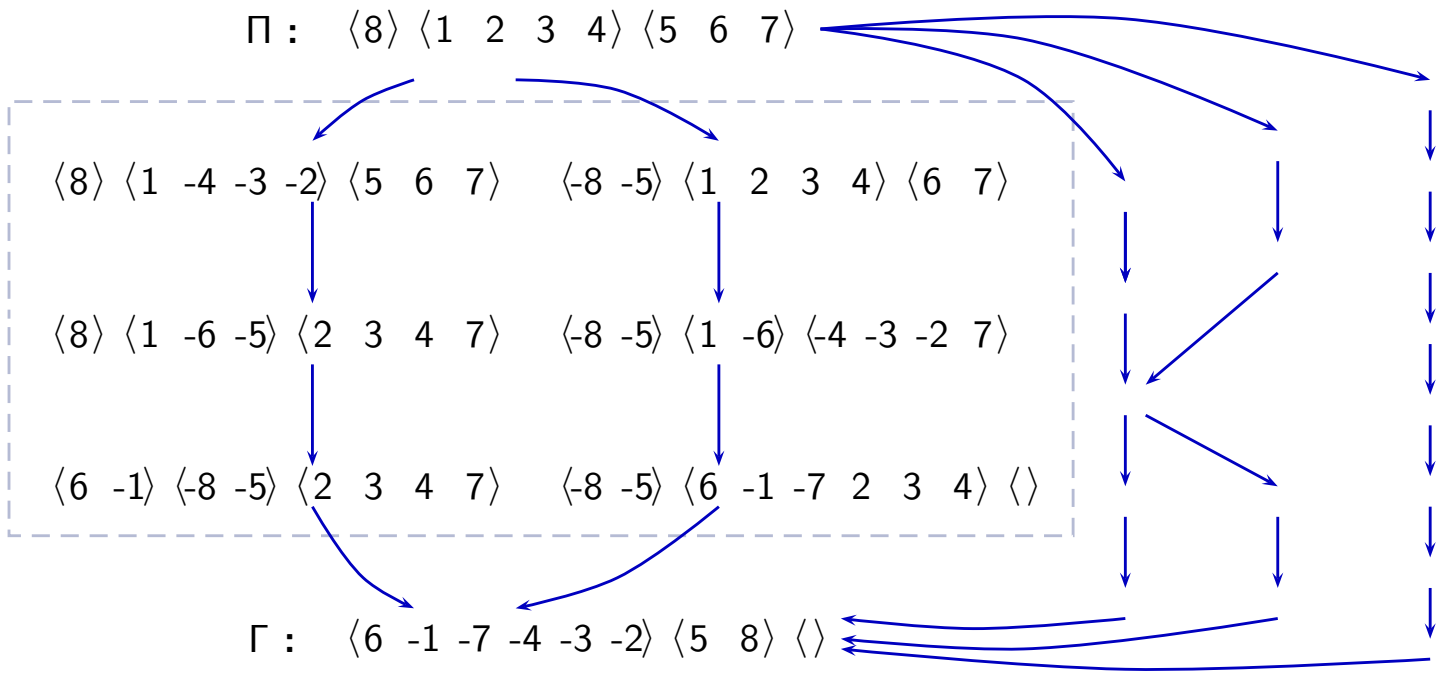
Two parsimonious scenarios



Breakpoints

Definition (Nadeau and Taylor 1984)

Two consecutive elements π_i and π_{i+1} of a chromosome π are said to be **adjacent** in the genome Π.
If two elements π_i and π_{i+1} are adjacent in Π but neither $\pi_i.\pi_{i+1}$ nor $-\pi_{i+1}.-\pi_i$ are present in Γ, then $\pi_i.\pi_{i+1}$ forms a **breakpoint** in Π.

$$\Pi = \langle \text{-9} \quad \text{-8} \quad +6 \quad +7 \quad +2 \quad +3 \quad +4 \quad +5 \quad +1 \rangle$$
$$\Gamma = \langle +1 \quad +2 \quad +3 \quad +4 \quad +5 \quad +6 \quad +7 \quad +8 \quad +9 \rangle$$

Breakpoints in Π are :

$$\Pi = \langle \text{-9} \quad \text{-8} \bullet +6 \quad +7 \bullet +2 \quad +3 \quad +4 \quad +5 \bullet +1 \rangle \bullet$$

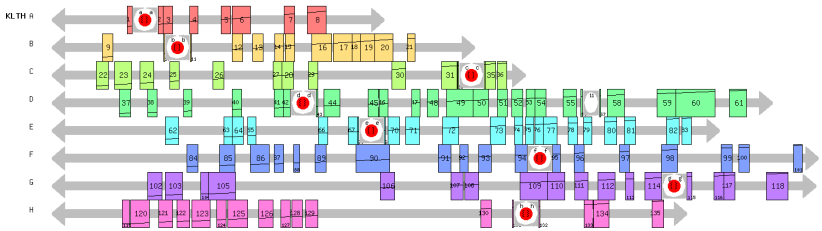
- Breakpoint distance $b(\Pi, \Gamma)$: Number of breakpoints between Π and Γ
- $d(\Pi, \Gamma)$ and $b(\Pi, \Gamma)$ closely related : finding rearrangements within Π that eliminate breakpoints between Π and Γ

Signed permutations for Yeasts

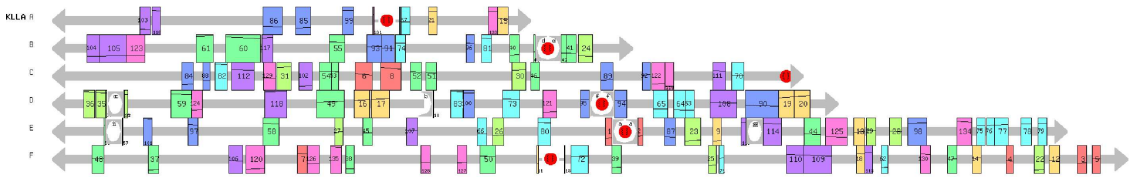
640 common markers computed with **SyDiG algorithm** :

- Same marker content without duplication, centromere positions
- Conservation of the 120 longest markers for study

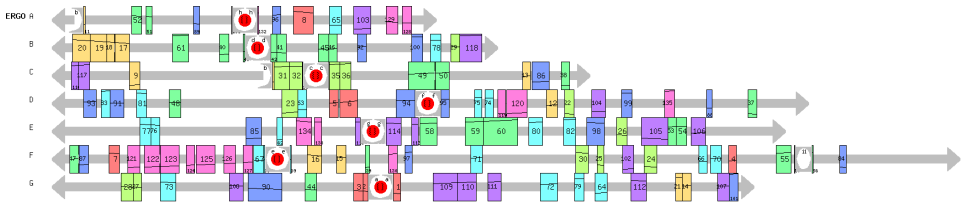
Klth



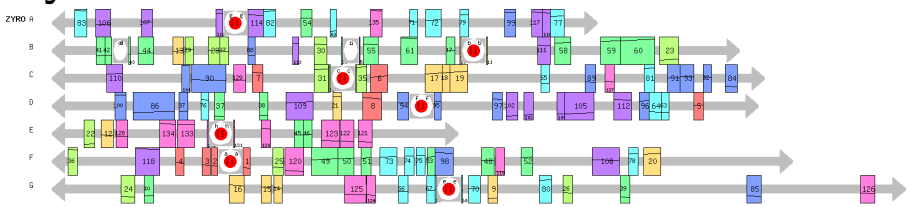
Klla



Ergo



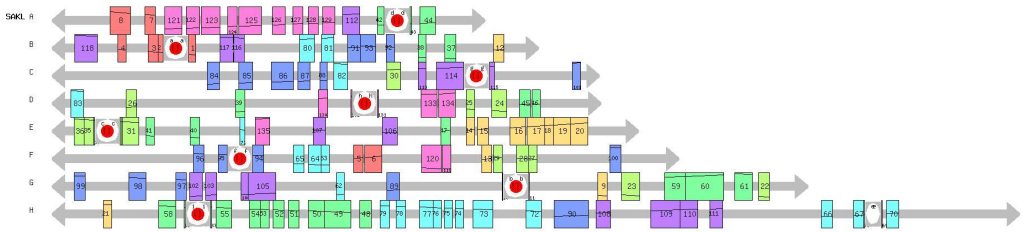
Zyro



	Klth	Ergo	Klla	Sakl	Zyro
Klth	0	88	105	45	84
Ergo		0	109	85	101
Klla			0	98	115
Sakl				0	79
Zyro					0

Pairwise distances

Sakl



Partial ancestral reconstructions : super-block method

Interest of ancestral reconstruction

Discovery of biological clues by the analysis of computed results :

- History of contemporary genomes
- General mechanisms of their formation

But results from two techniques do not necessarily agree

In Silico	**In Vitro**
Rearrangement distance	Chromosomal painting
(Bourque & Pevzner 2002), (Bourque, Tesler & Pevzner 2004)	(Wienberg et al. 1990)
Complete genomes ($\approx$ 5 species)	Eutherian clade ($\approx$ 80 species)
$\approx$ 3 Kb ($\approx$ yeast gene length)	$\approx$ 4 Mb ($>$ yeast chromosome length)

Possible solution : integrate more **biological knowledge** into the mathematical approach (Rocchi 2006)

Ancestors as median genomes

Definition

Median genome problem : given $G_1, ..., G_N$, find M such that for a distance d

$$\sum_{i=1}^{N} d(M, G_i) \text{ is minimal}$$

Different distances : **rearrangement, breakpoint**, double cut and join

- NP-complete even for $N = 3$
  breakpoint distance (Bryant 1998, Pe'er & Shamir 1998)
  rearrangement distance (Caprara 1999 and 2003)
- High number of equivalent solutions (Eriksen 2007)
- Minimal solutions can be highly divergent (Eriksen 2007)

$\Rightarrow$ Giving a unique global solution is biologically misleading

Piece-wise ancestral reconstruction

Looking for **common features** present in ancestral genome architecture

CARs (Contiguous Ancestral Regions) method (Ma et al. 2006) :

- Based on phylogenetic tree
- Analogous to Fitch's parsimony method (Fitch 1971)

Phylogeny relationships	**Chromosome dynamics**
Rate of punctual mutations in genomic sequences	Rearrangement of breakpoint distance
Temporal notion	No time-scale of the rearrangement events

⇒ We propose a new piece-wise method without phylogenetic consideration

A new method for reconstructing ancestral architecture

Breakpoint-based method (Sankoff & Blanchette 1997) : more an adjacency is frequent in contemporary genomes, more it should appear in the ancestral genome

An adjacency which appears in only one genome is not informative !

New method :

- Piece-wise reconstruction
- Based on breakpoints and rearrangement distance
- Adding biological constraints is possible

Adjacency frequencies

Definition

The frequency of an adjacency a**,** $u(a)$, is the number of genomes where a is present

Particular adjacencies in $\pi = \{\pi_1, .., \pi_n\}$: telomeres $0.\pi_1$ and $\pi_n.0$

Example

$$G_1 = \{1\ 2\ 3\ 4,\ 5\ 6\} \qquad G_2 = \{1\ 2\ 3\ 4,\ -5\ 6\}$$
$$G_3 = \{3\ 1\ 4\ 2\ -5,\ 6\} \qquad G_4 = \{2\ 1\ 3\ 4,\ 5\ 6\}$$

frequency	adjacencies
4	6.0
3	3.4, 0.5, 4.0
2	5.6, 2.3, 1.2, 0.1
1	−5.6, 2.-5, 4.2, 1.4, 1.3, 3.1, 2.1, 0.6, 5.0, 0.3, 0.2

Adjacency graph

Unsigned representation of signed permutation (Hannenhalli & Pevzner, 1995) :

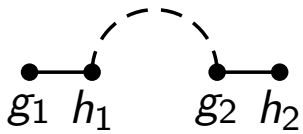
$g = 2\pi_i - 1 \quad h = 2\pi_i$ (a) π_i is positive

$h = 2\pi_i \quad g = 2\pi_i - 1$ (b) π_i is negative

Denoted : $\pi_i.\pi_j$ by $(g_i\ h_i).(g_j\ h_j)$ and $\pi_i.-\pi_j$ by $(g_i\ h_i).(h_j\ g_j)$

Example

The **adjacency graph** for a set $A = \{(g_1\ h_1).(g_2\ h_2)\}$:



- 4 vertices g_1, h_1, g_2 and h_2
- two edges stand for markers $e_1 = \{g_1, h_1\}$ and $e_2 = \{g_2, h_2\}$.
- one edge stands for the adjacency $e_3 = \{h_1, g_2\}$

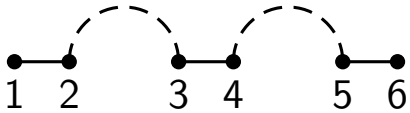
Intuition

Hypothesis

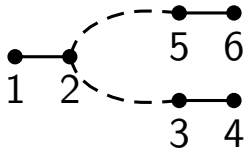
For a set of genomes $\{G_i\}$, the higher is the frequency of an adjacency, the higher is the probability that it should be present in a median genome.

Build **partial assemblies** of median genomes

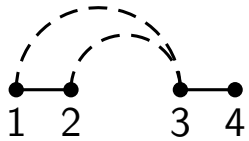
1. Build a partition $\mathcal{P}$ of adjacencies where each part is composed of inter-dependent adjacencies.



complementary adjacencies



vertex contradiction



cycle contradiction

2. Inspect $\mathcal{P}$ in decreasing frequency of its parts, and construct the partial assemblies by favoring adjacencies with higher frequency.

Assemble these partial assemblies into **potential medians**

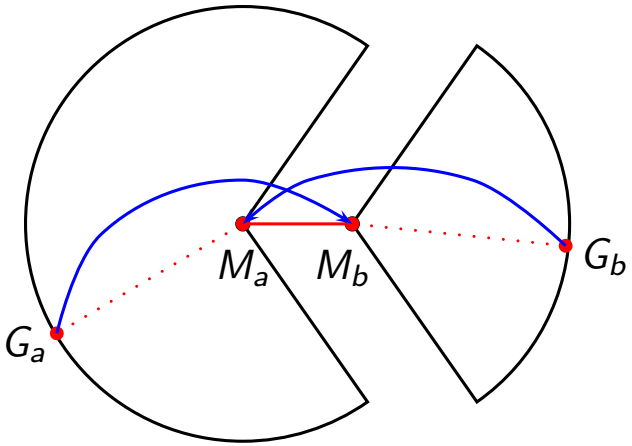
Adjacencies and distances

N contemporary genomes $\{G_i\}$, d rearrangement distance
C the set of all pairs of contradictory adjacencies

Theorem (Jean et al.)

For any pair $\{a, b\} \in C$ and two genomes M_a and M_b identical up to 2 adjacencies with $a \in M_a$ and $b \in M_b$, it holds that

$$\left| \sum_i^N d(M_a, G_i) - \sum_i^N d(M_b, G_i) \right| \leq N.$$



If $u(a) > u(b)$

$$\sum_i^N d(M_a, G_i) - \sum_i^N d(M_b, G_i) \ll N$$

Similarly for the breakpoint distance

Groups of adjacencies

$\mathcal{P}(A)$ be a partition of $\mathcal{A}$, set of all adjacencies.
$\mathcal{P}_0(\mathcal{A})$: adjacencies in cycle contradiction and singletons

Merging of parts : $\sqcup$ defines a partition of $\mathcal{A}$ such that for any $p \in \sqcup(\mathcal{P}(\mathcal{A}))$

- $\exists\ p_1 \in \mathcal{P}(A)$ s.t. $p = p_1$ or
- $\exists\ p_1, p_2 \in \mathcal{P}(\mathcal{A})$ s.t. $p = p_1 \cup p_2$ and moreover $\exists\ a \in p_1$ and $\exists\ b \in p_2$ s.t. $u(a) = u(b) = u(p_1) = u(p_2)$ and either a and b are dependent or a and b participate in a cycle $c \in G$ without vertex $v = 0$ s.t. $\forall v \in c$ we have $u(v) \geq u(a)$.

Definition

A **group** g is a part of $\sqcup^n(\mathcal{P}_0(A))$, where $\sqcup^n(\mathcal{P}_0(A))$ is the fixed point of $\sqcup$.

Groups of adjacencies, continued

Example

$G_1 = \{$ **1 2 3 4, 5 6** $\}$ $G_2 = \{$ **1 2 3 4, -5 6** $\}$

$G_3 = \{$ **3 1 4 2 -5, 6** $\}$ $G_4 = \{$ **2 1 3 4, 5 6** $\}$

Groups of adjacencies, continued

Example

$G_1 = \{ \underset{1\ 2}{\mathbf{1}} \quad \underset{3\ 4}{\mathbf{2}} \quad \underset{5\ 6}{\mathbf{3}} \quad \underset{7\ 8,}{\mathbf{4,}} \quad \underset{9\ 10}{\mathbf{5}} \quad \underset{11\ 12}{\mathbf{6}} \}$

$G_2 = \{ \underset{1\ 2}{\mathbf{1}} \quad \underset{3\ 4}{\mathbf{2}} \quad \underset{5\ 6}{\mathbf{3}} \quad \underset{7\ 8,}{\mathbf{4,}} \quad \underset{10\ 9}{\mathbf{-5}} \quad \underset{11\ 12}{\mathbf{6}} \}$

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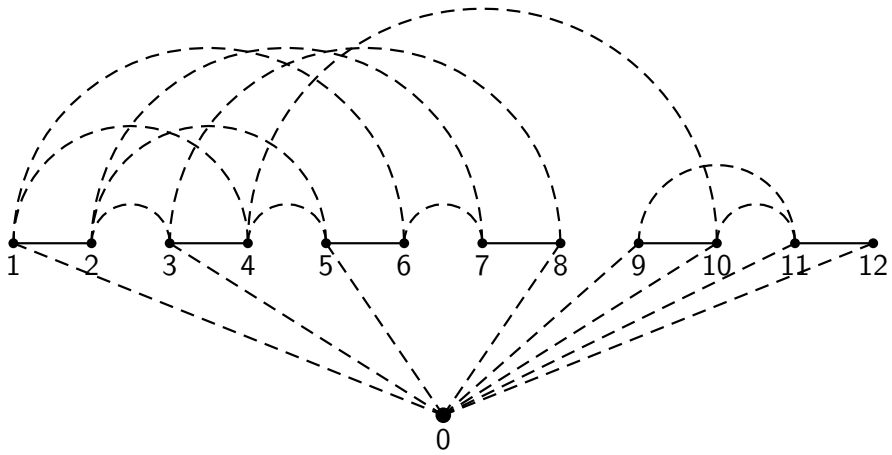
$G_4 = \{ \underset{3\ 4}{\mathbf{2}} \quad \underset{1\ 2}{\mathbf{1}} \quad \underset{5\ 6}{\mathbf{3}} \quad \underset{7\ 8,}{\mathbf{4,}} \quad \underset{9\ 10}{\mathbf{5}} \quad \underset{11\ 12}{\mathbf{6}} \}$



Adjacency graph

Groups of adjacencies, continued

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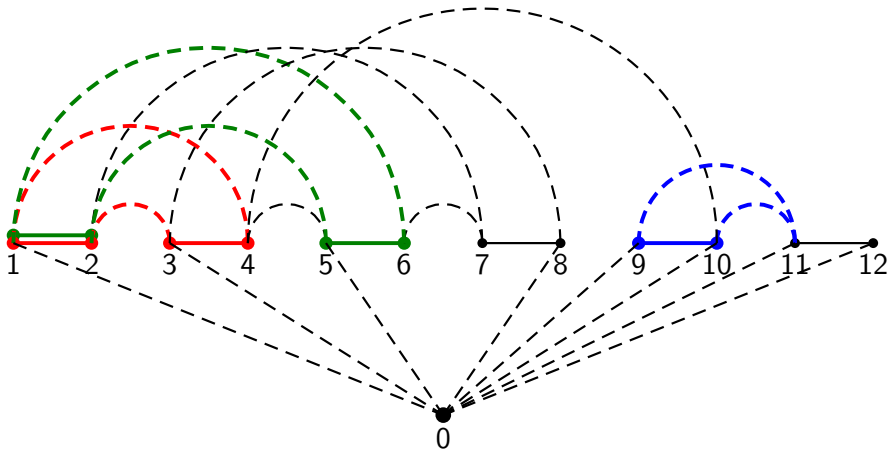
$G_1 = \{ \underset{1\ 2}{\mathbf{1}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{9\ 10}{\mathbf{5}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_2 = \{ \underset{1\ 2}{\mathbf{1}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{10\ 9}{\mathbf{-5}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_3 = \{ \underset{5\ 6}{\mathbf{3}} \ \underset{1\ 2}{\mathbf{1}} \ \underset{7\ 8}{\mathbf{4}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{10\ 9,}{\mathbf{-5,}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_4 = \{ \underset{3\ 4}{\mathbf{2}} \ \underset{1\ 2}{\mathbf{1}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{9\ 10}{\mathbf{5}} \ \underset{11\ 12}{\mathbf{6}} \}$



Partition $\mathcal{P}_0$: 3 cycle contradictions and singletons

Part freq.	Adjacencies
4	6.0(4)
3	3.4(3)
3	4.0(3)
3	0.5(3)
2	**-5.6(1), 5.6(2)**
2	2.3(2)
2	0.1(2)
2	**1.2(2), 2.1(1)**
1	**1.3(1), 3.1(1)**
1	0.3(1)
1	1.4(1)
1	4.2(1)
1	2.-5(1)
1	0.6(1)
1	0.-5(1)
1	0.2(1)

Groups of adjacencies, continued

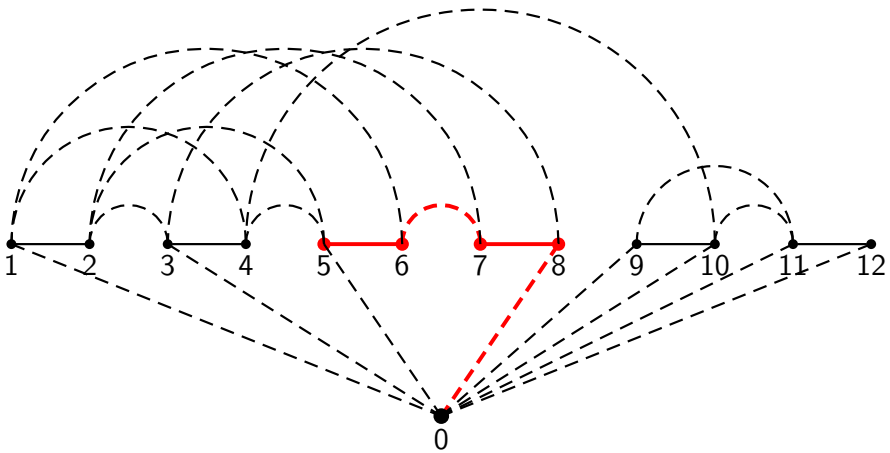
Example

$G_1 = \{ \underset{1\ 2}{1} \ \underset{3\ 4}{2} \ \underset{5\ 6}{3} \ \underset{7\ 8,}{4,} \ \underset{9\ 10}{5} \ \underset{11\ 12}{6} \}$

$G_2 = \{ \underset{1\ 2}{1} \ \underset{3\ 4}{2} \ \underset{5\ 6}{3} \ \underset{7\ 8,}{4,} \ \underset{10\ 9}{-5} \ \underset{11\ 12}{6} \}$

$G_3 = \{ \underset{5\ 6}{3} \ \underset{1\ 2}{1} \ \underset{7\ 8}{4} \ \underset{3\ 4}{2} \ \underset{10\ 9,}{-5,} \ \underset{11\ 12}{6} \}$

$G_4 = \{ \underset{3\ 4}{2} \ \underset{1\ 2}{1} \ \underset{5\ 6}{3} \ \underset{7\ 8,}{4,} \ \underset{9\ 10}{5} \ \underset{11\ 12}{6} \}$



Complementary adjacencies 3.4 and 4.0 (same part frequencies)

Part freq.	Adjacencies
4	6.0(4)
3	**3.4(3)**
3	**4.0(3)**
3	0.5(3)
2	-5.6(1), 5.6(2)
2	2.3(2)
2	0.1(2)
2	1.2(2), 2.1(1)
1	1.3(1), 3.1(1)
1	0.3(1)
1	1.4(1)
1	4.2(1)
1	2.-5(1)
1	0.6(1)
1	0.-5(1)
1	0.2(1)

Groups of adjacencies, continued

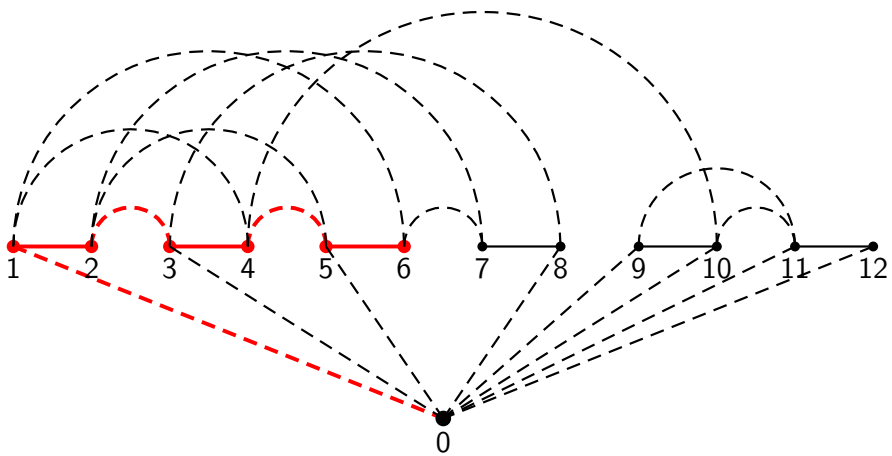
Example

$G_1 = \{ \underset{1\ 2}{\mathbf{1}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{9\ 10}{\mathbf{5}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_2 = \{ \underset{1\ 2}{\mathbf{1}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{10\ 9}{\mathbf{-5}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_3 = \{ \underset{5\ 6}{\mathbf{3}} \ \underset{1\ 2}{\mathbf{1}} \ \underset{7\ 8}{\mathbf{4}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{10\ 9,}{\mathbf{-5,}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_4 = \{ \underset{3\ 4}{\mathbf{2}} \ \underset{1\ 2}{\mathbf{1}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{9\ 10}{\mathbf{5}} \ \underset{11\ 12}{\mathbf{6}} \}$



Complementary adjacencies 0.1, 1.2 and 2.3 (same part frequencies)

Part freq.	Adjacencies
4	6.0(4)
3	3.4(3), 4.0(3)
3	0.5(3)
2	-5.6(1), 5.6(2)
2	**2.3(2)**
2	**0.1(2)**
2	**1.2(2)**, 2.1(1)
1	1.3(1), 3.1(1)
1	0.3(1)
1	1.4(1)
1	4.2(1)
1	2.-5(1)
1	0.6(1)
1	0.-5(1)
1	0.2(1)

Groups of adjacencies, continued

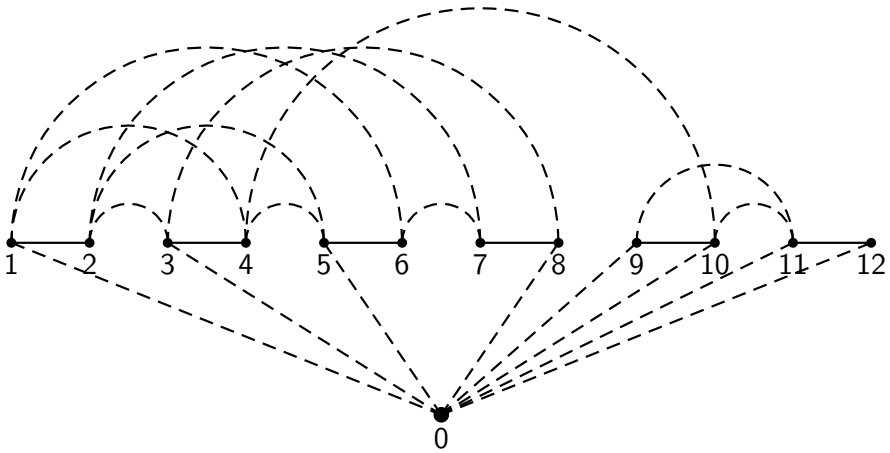
Example

$G_1 = \{ \underset{1\ 2}{\mathbf{1}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{9\ 10}{\mathbf{5}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_2 = \{ \underset{1\ 2}{\mathbf{1}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{10\ 9}{\mathbf{-5}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_3 = \{ \underset{5\ 6}{\mathbf{3}} \ \underset{1\ 2}{\mathbf{1}} \ \underset{7\ 8}{\mathbf{4}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{10\ 9,}{\mathbf{-5,}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_4 = \{ \underset{3\ 4}{\mathbf{2}} \ \underset{1\ 2}{\mathbf{1}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{9\ 10}{\mathbf{5}} \ \underset{11\ 12}{\mathbf{6}} \}$



Other complementary adjacencies and adjacencies in contradiction...

Part freq.	Adjacencies
4	6.0(4)
3	3.4(3), 4.0(3)
3	0.5(3)
2	-5.6(1), 5.6(2)
2	1.2(2), 2.1(1), 2.3(2), 0.1(2)
1	**1.3(1), 3.1(1)**
1	**0.3(1)**
1	**1.4(1)**
1	**4.2(1)**
1	**2.-5(1)**
1	0.6(1)
1	**0.-5(1)**
1	**0.2(1)**

Groups of adjacencies, continued

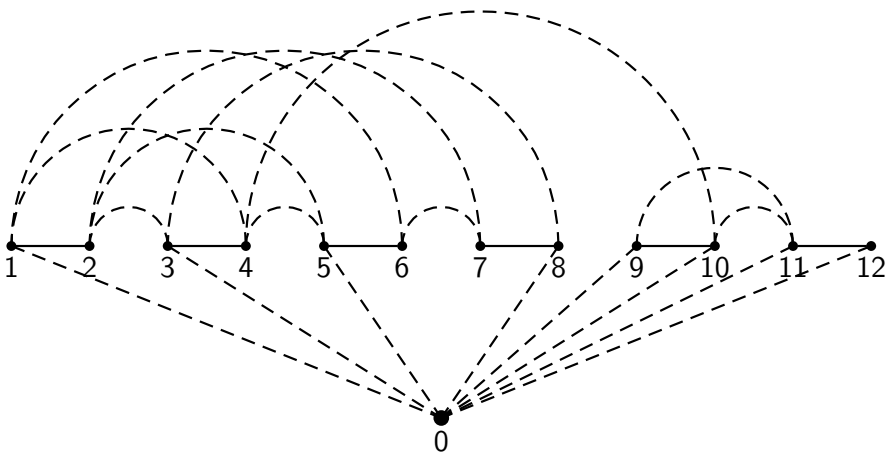
Example

$G_1 = \{ \underset{1\ 2}{\mathbf{1}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{9\ 10}{\mathbf{5}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_2 = \{ \underset{1\ 2}{\mathbf{1}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{10\ 9}{\mathbf{-5}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_3 = \{ \underset{5\ 6}{\mathbf{3}} \ \underset{1\ 2}{\mathbf{1}} \ \underset{7\ 8}{\mathbf{4}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{10\ 9,}{\mathbf{-5,}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_4 = \{ \underset{3\ 4}{\mathbf{2}} \ \underset{1\ 2}{\mathbf{1}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{9\ 10}{\mathbf{5}} \ \underset{11\ 12}{\mathbf{6}} \}$



Group freq.	Adjacencies
4	6.0(4)
3	3.4(3), 4.0(3)
3	0.5(3)
2	-5.6(1), 5.6(2)
2	1.2(2), 2.1(1), 2.3(2), 0.1(2)
1	1.3(1), 3.1(1), 0.3(1), 1.4(1), 4.2(1), 2.-5(1), 0.-5(1), 0.2(1)
1	0.6(1)

Final groups of $\sqcup^n(\mathcal{P}_0)$

Super-blocks (intuition part 2)

Definition

A **super-block** is a set S of $n \geq 1$ adjacencies s.t. $\forall a, b \in S$, a does not contradict b, and there exists an order over S such that $\forall i \in [1, n)$, a_i complements a_{i+1}, and a_1, a_n are either independent or $a_1 = a_n = 0$. A **partial assembly** $\mathcal{P} = \{S_k\}$ is a set of superblocks such that $\forall k \neq l$ if $S_k \cap S_l \neq \emptyset \Rightarrow S_k \cap S_l = \{0\}$.

Lemma

The adjacency graph $G = (V, E)$ of a partial assembly $\mathcal{P}$ is a graph such that (1) $\forall v \in V$, $d(v) \leq 2$, except for $v = 0$, and (2) any cycle in G contains 0.

Super-block construction

Example

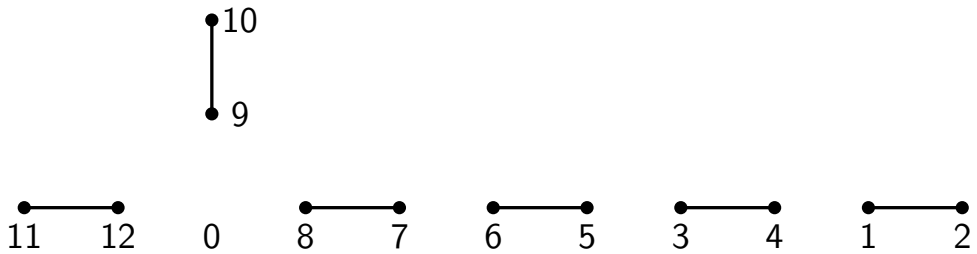
$G_1 = \{$ **1** **2** **3** **4,** **5** **6** $\}$ (1 2 3 4 5 6 7 8, 9 10 11 12)

$G_2 = \{$ **1** **2** **3** **4,** **-5** **6** $\}$ (1 2 3 4 5 6 7 8, 10 9 11 12)

$G_3 = \{$ **3** **1** **4** **2** **-5,** **6** $\}$ (5 6 1 2 7 8 3 4 10 9, 11 12)

$G_4 = \{$ **2** **1** **3** **4,** **5** **6** $\}$ (3 4 1 2 5 6 7 8, 9 10 11 12)

Group freq.	Adjacencies
4	6.0(4)
3	3.4(3), 4.0(3)
3	0.5(3)
2	-5.6(1), 5.6(2)
2	1.2(2), 2.1(1), 2.3(2), 0.1(2)
1	1.3(1), 3.1(1), 0.3(1), 1.4(1), 4.2(1), 2.-5(1), 0.-5(1), 0.2(1)
1	0.6(1)



Initial graph

Super-block construction

Example

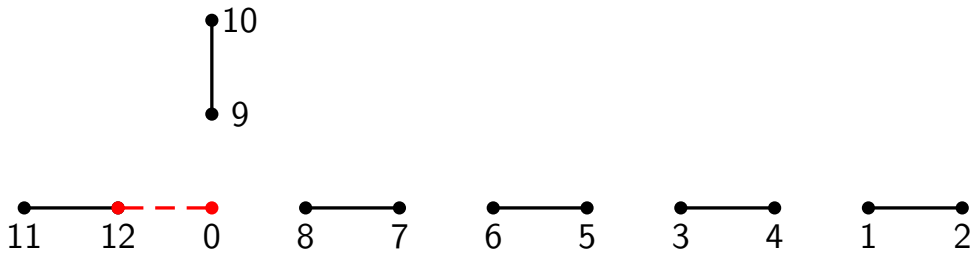
$G_1 = \{ \underset{1\ 2}{\mathbf{1}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{9\ 10}{\mathbf{5}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_2 = \{ \underset{1\ 2}{\mathbf{1}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{10\ 9}{\mathbf{-5}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_3 = \{ \underset{5\ 6}{\mathbf{3}} \ \underset{1\ 2}{\mathbf{1}} \ \underset{7\ 8}{\mathbf{4}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{10\ 9,}{\mathbf{-5,}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_4 = \{ \underset{3\ 4}{\mathbf{2}} \ \underset{1\ 2}{\mathbf{1}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{9\ 10}{\mathbf{5}} \ \underset{11\ 12}{\mathbf{6}} \}$

Group freq.	Adjacencies
4	**6.0(4)**
3	3.4(3), 4.0(3)
3	0.5(3)
2	-5.6(1), 5.6(2)
2	1.2(2), 2.1(1), 2.3(2), 0.1(2)
1	1.3(1), 3.1(1), 0.3(1), 1.4(1), 4.2(1), 2.-5(1), 0.-5(1), 0.2(1)
1	0.6(1)



Adding groups by decreasing frequency

$6.0 = (11\ 12).0$

Super-block construction

Example

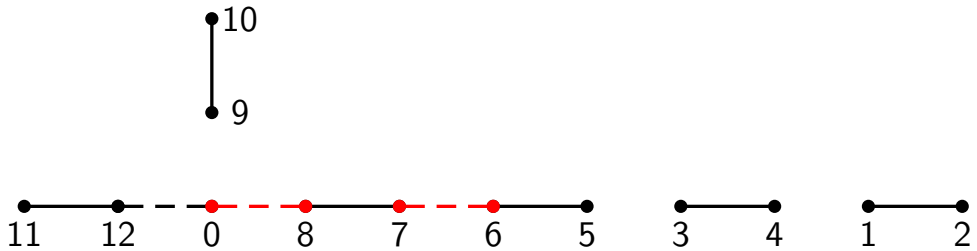
$G_1 = \{$ **1** **2** **3** **4,** **5** **6** $\}$
1 2 3 4 5 6 7 8, 9 10 11 12

$G_2 = \{$ **1** **2** **3** **4,** **-5** **6** $\}$
1 2 3 4 5 6 7 8, 10 9 11 12

$G_3 = \{$ **3** **1** **4** **2** **-5,** **6** $\}$
5 6 1 2 7 8 3 4 10 9, 11 12

$G_4 = \{$ **2** **1** **3** **4,** **5** **6** $\}$
3 4 1 2 5 6 7 8, 9 10 11 12

Group freq.	Adjacencies
4	6.0(4)
3	**3.4(3), 4.0(3)**
3	0.5(3)
2	-5.6(1), 5.6(2)
2	1.2(2), 2.1(1), 2.3(2), 0.1(2)
1	1.3(1), 3.1(1), 0.3(1), 1.4(1), 4.2(1), 2.-5(1), 0.-5(1), 0.2(1)
1	0.6(1)



$3.4 = (5\ 6).(7\ 8)$ and $4.0 = (7\ 8).0$

Super-block construction

Example

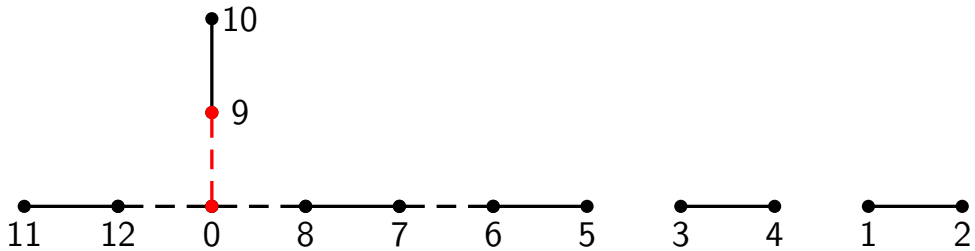
$G_1 = \{$ **1** **2** **3** **4,** **5** **6** $\}$ (1 2 3 4 5 6 7 8, 9 10 11 12)

$G_2 = \{$ **1** **2** **3** **4,** **-5** **6** $\}$ (1 2 3 4 5 6 7 8, 10 9 11 12)

$G_3 = \{$ **3** **1** **4** **2** **-5,** **6** $\}$ (5 6 1 2 7 8 3 4 10 9, 11 12)

$G_4 = \{$ **2** **1** **3** **4,** **5** **6** $\}$ (3 4 1 2 5 6 7 8, 9 10 11 12)

Group freq.	Adjacencies
4	6.0(4)
3	3.4(3), 4.0(3)
3	**0.5(3)**
2	-5.6(1), 5.6(2)
2	1.2(2), 2.1(1), 2.3(2), 0.1(2)
1	1.3(1), 3.1(1), 0.3(1), 1.4(1), 4.2(1), 2.-5(1), 0.-5(1), 0.2(1)
1	0.6(1)



$0.5 = 0.(9\ 10)$

Super-block construction

Example

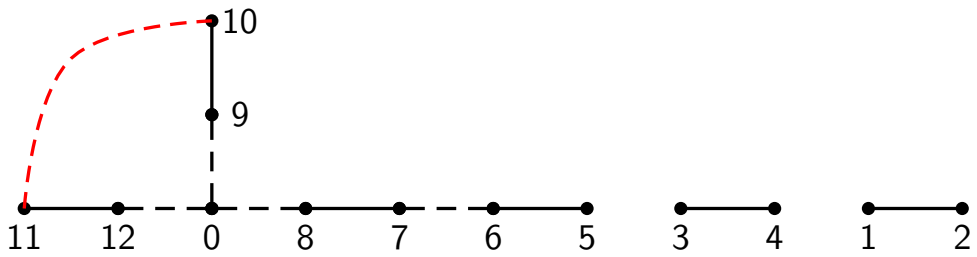
$G_1 = \{ \underset{1\ 2}{\mathbf{1}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{9\ 10}{\mathbf{5}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_2 = \{ \underset{1\ 2}{\mathbf{1}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{10\ 9}{\mathbf{-5}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_3 = \{ \underset{5\ 6}{\mathbf{3}} \ \underset{1\ 2}{\mathbf{1}} \ \underset{7\ 8}{\mathbf{4}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{10\ 9,}{\mathbf{-5,}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_4 = \{ \underset{3\ 4}{\mathbf{2}} \ \underset{1\ 2}{\mathbf{1}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{9\ 10}{\mathbf{5}} \ \underset{11\ 12}{\mathbf{6}} \}$

Group freq.	Adjacencies
4	6.0(4)
3	3.4(3), 4.0(3)
3	0.5(3)
2	**-5.6(1), 5.6(2)**
2	1.2(2), 2.1(1), 2.3(2), 0.1(2)
1	1.3(1), 3.1(1), 0.3(1), 1.4(1), 4.2(1), 2.-5(1), 0.-5(1), 0.2(1)
1	0.6(1)



$-5.6 = (10\ 9).(11\ 12)$ in conflict with $0.5 = 0.(9\ 10)$

$\Rightarrow$ Only addition of $5.6 = (9\ 10).(11\ 12)$

Super-block construction

Example

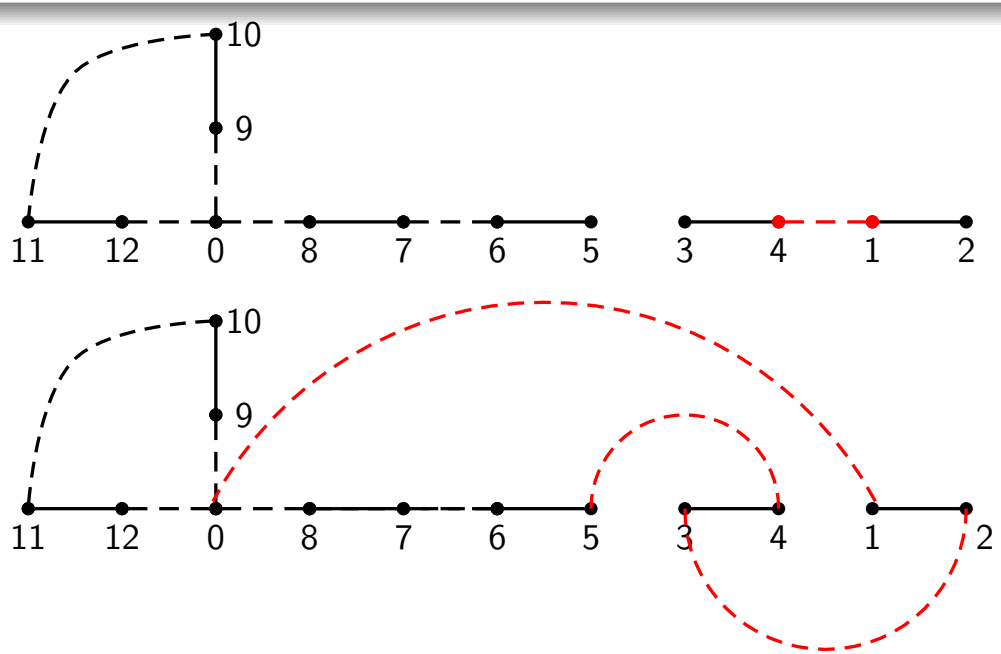
$G_1 = \{$ 1 2 3 4, 5 6 $\}$ (1 2 3 4 5 6 7 8, 9 10 11 12)

$G_2 = \{$ 1 2 3 4, -5 6 $\}$ (1 2 3 4 5 6 7 8, 10 9 11 12)

$G_3 = \{$ 3 1 4 2 -5, 6 $\}$ (5 6 1 2 7 8 3 4 10 9, 11 12)

$G_4 = \{$ 2 1 3 4, 5 6 $\}$ (3 4 1 2 5 6 7 8, 9 10 11 12)

Group freq.	Adjacencies
4	6.0(4)
3	3.4(3), 4.0(3)
3	0.5(3)
2	-5.6(1), 5.6(2)
2	**1.2(2), 2.1(1), 2.3(2), 0.1(2)**
1	1.3(1), 3.1(1), 0.3(1), 1.4(1), 4.2(1), 2.-5(1), 0.-5(1), 0.2(1)
1	0.6(1)



2 possible sets :
$\{2.1\}$ or $\{0.1, 1.2, 2.3\}$

Super-block construction

Example

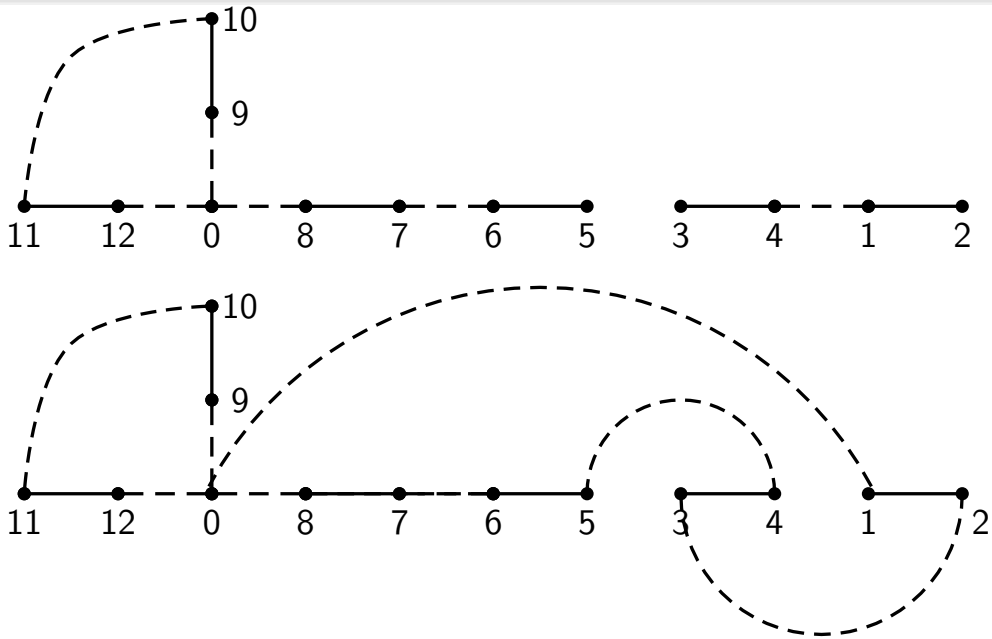
$G_1 = \{ \underset{1\ 2}{\mathbf{1}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{9\ 10}{\mathbf{5}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_2 = \{ \underset{1\ 2}{\mathbf{1}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{10\ 9}{\mathbf{-5}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_3 = \{ \underset{5\ 6}{\mathbf{3}} \ \underset{1\ 2}{\mathbf{1}} \ \underset{7\ 8}{\mathbf{4}} \ \underset{3\ 4}{\mathbf{2}} \ \underset{10\ 9,}{\mathbf{-5,}} \ \underset{11\ 12}{\mathbf{6}} \}$

$G_4 = \{ \underset{3\ 4}{\mathbf{2}} \ \underset{1\ 2}{\mathbf{1}} \ \underset{5\ 6}{\mathbf{3}} \ \underset{7\ 8,}{\mathbf{4,}} \ \underset{9\ 10}{\mathbf{5}} \ \underset{11\ 12}{\mathbf{6}} \}$

Group freq.	Adjacencies
4	6.0(4)
3	3.4(3), 4.0(3)
3	0.5(3)
2	-5.6(1), 5.6(2)
2	1.2(2), 2.1(1), 2.3(2), 0.1(2)
1	**1.3(1), 3.1(1), 0.3(1), 1.4(1), 4.2(1), 2.-5(1), 0.-5(1), 0.2(1)**
1	**0.6(1)**



Not enough support in groups of frequency 1

Super-block construction

Example

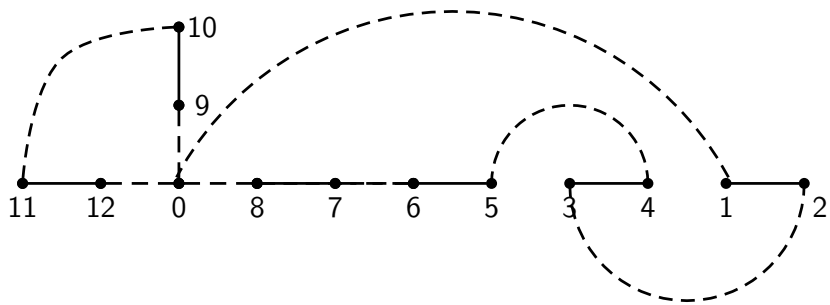
$G_1 = \{$ **1 2 3 4, 5 6** $\}$ (1 2 3 4 5 6 7 8, 9 10 11 12)

$G_2 = \{$ **1 2 3 4, -5 6** $\}$ (1 2 3 4 5 6 7 8, 10 9 11 12)

$G_3 = \{$ **3 1 4 2 -5, 6** $\}$ (5 6 1 2 7 8 3 4 10 9, 11 12)

$G_4 = \{$ **2 1 3 4, 5 6** $\}$ (3 4 1 2 5 6 7 8, 9 10 11 12)

Two partial assemblies :

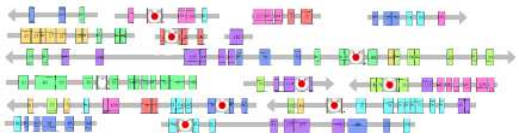


$M_1 = \{2\ 1,\ 3\ 4,\ 5\ 6\}$ having 3 super-blocks and $\sum d(M_1, G_i) = 10$

$M_2 = \{1\ 2\ 3\ 4,\ 5\ 6\}$ having 2 super-blocks and $\sum d(M_2, G_i) = 9$

Final assembly

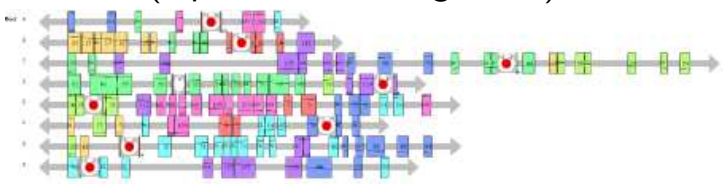
A partial assembly
(a set of super-blocks)



Fusions of super-blocks
=
solving extremity adjacencies
of super-blocks

biological constraint :
one and only one centromere
by chromosome

A final assembly
(a possible median genome)



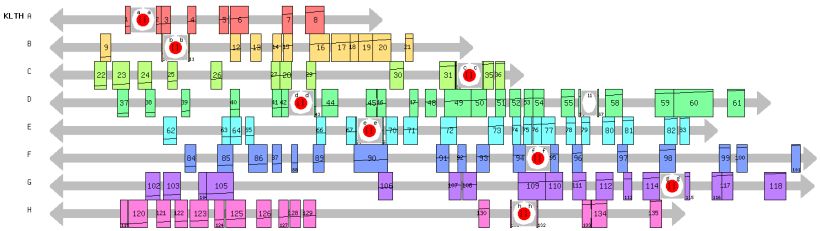
Theorem (Jean et al.)

For any $\mathcal{P} \in \{\mathcal{P}\}$ of $G_1, ..., G_N$ such that $\mathcal{P} = \{S_k\}$, there exists a genome M such that for any chromosome π of M either $\exists\, S_k \in \mathcal{P}$ such that $\pi = S_k$, or $\exists\, \{S_k\} \subseteq \mathcal{P}$ such that π is formed by a series of fusions $\pi = S_1 ... S_k$. Moreover,

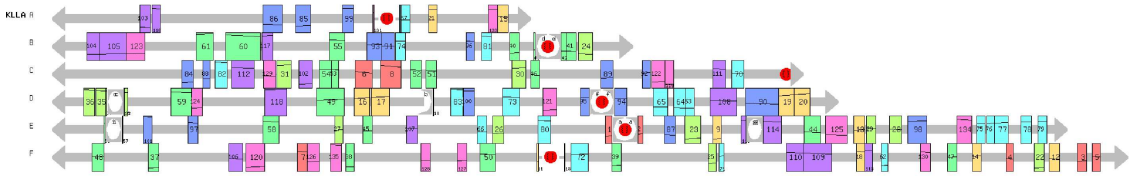
$$\sum_i^N d(M, G_i) - \sum_i^N d(P, G_i) \leq 0 \text{ and } \sum_i^N b(M, G_i) - \sum_i^N b(P, G_i) \leq 0.$$

Comparative maps

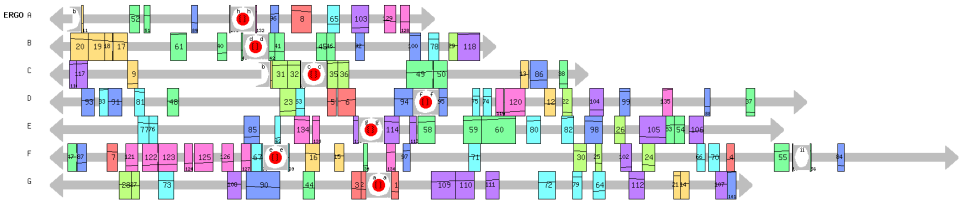
Klth



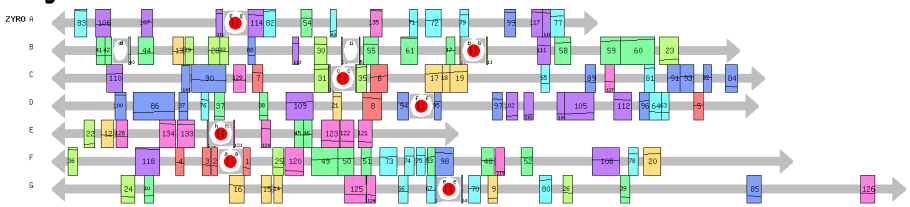
Klla



Ergo



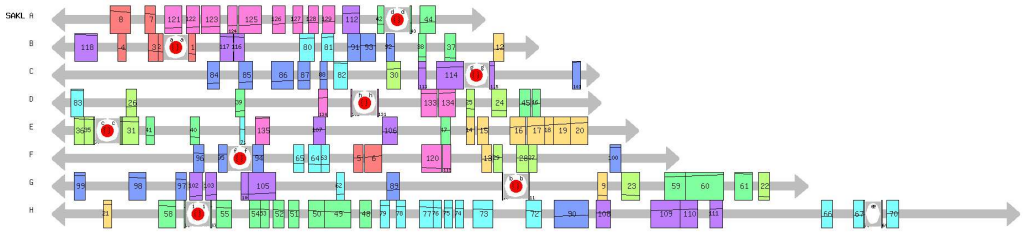
Zyro



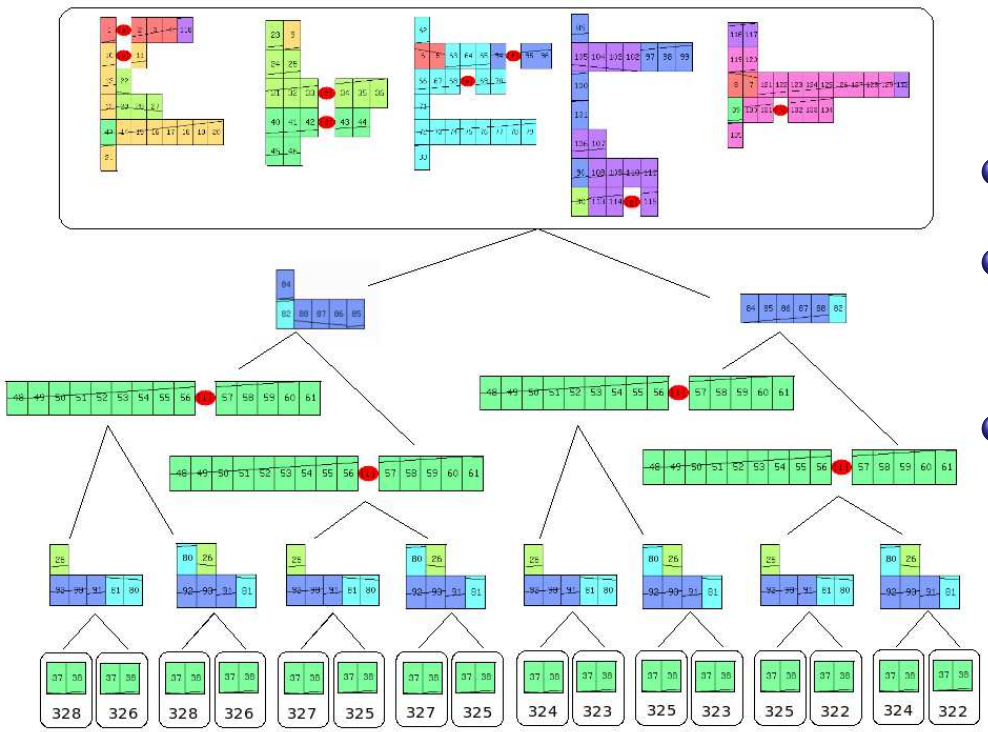
	Klth	Ergo	Klla	Sakl	Zyro
Klth	0	88	105	45	84
Ergo		0	109	85	101
Klla			0	98	115
Sakl				0	79
Zyro					0

Pairwise distances

Sakl

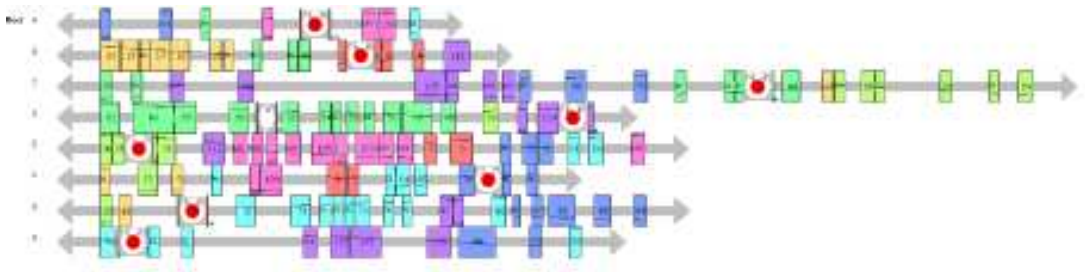


Sharing tree of super-blocks



- 1 branch = 1 partial assembly
- 16 partial assemblies $\longrightarrow$ 4 unsolved conflicts
- 29 common super-blocks on 34 or 35

- A possible final assembly
- $\sum_{i=1}^{5} d(M, G_i) = 284$



Constructing and visualizing parsimonious scenarios

HP theory (Hannenhalli & Pevzner 1995)

Definition

A parsimonious scenario is a sequence of rearrangements respecting rearrangement distance

- Unichromosomal case : exact reversal distance and parsimonious scenario in polynomial time
- Multichromosomal case : using unichromosomal theory by mimicking multichromosomal rearrangements by reversals on a single permutation

$$\Pi = \{< -1\ 2 >, < 3\ 4 >, < 5\ 8\ 7\ 6 >\}$$
$$\Gamma = \{< 1\ 2\ 3\ 4 >, < 5\ 6\ 7\ 8 >\}$$

Capping (caps)

Concatenate

$$\hat{\pi} = 9\ -1\ 2\ 10\ 11\ 3\ 4\ 12\ 13\ 5\ 8\ 7\ 6\ 14$$
$$\hat{\gamma} = 9\ 1\ 2\ 3\ 4\ 10\ 11\ 5\ 6\ 7\ 8\ 12\ 13\ 14$$

- Errors in distance and scenario : corrected by (Tesler, 2002) and by (Ozery-Flato & Shamir, 2003)
- Still one error to delineate chromosomes

Breakpoint graph

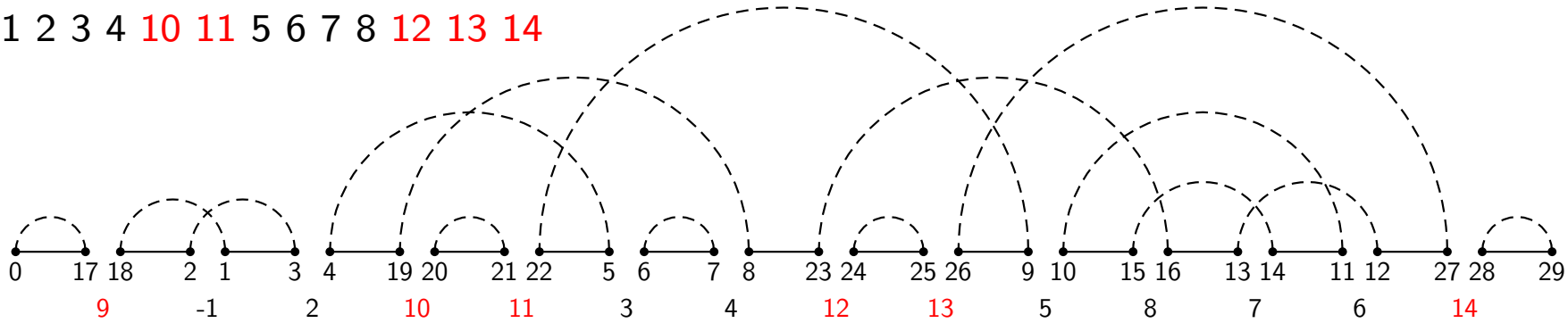
HP theory based on the **breakpoint graph** $G(\Pi, \Gamma)$

- Two vertices per signed identifier :

$2\pi_i - 1$ $+\pi_i$ $2\pi_i$ $\qquad$ $2\pi_i$ $-\pi_i$ $2\pi_i - 1$

- edges : adjacencies in Π (black edges) or Γ (dashed edges)

$\hat{\pi} = 9\ -1\ 2\ 10\ 11\ 3\ 4\ 12\ 13\ 5\ 8\ 7\ 6\ 14$
$\hat{\gamma} = 9\ 1\ 2\ 3\ 4\ 10\ 11\ 5\ 6\ 7\ 8\ 12\ 13\ 14$



Aim : increase the number of cycles

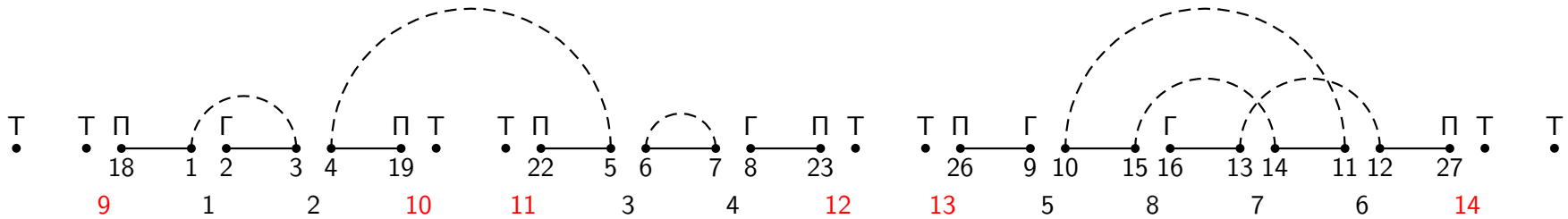
Breakpoint graph

HP theory based on the **breakpoint graph** $G(\Pi, \Gamma)$

- Two vertices per signed identifier :

- edges : adjacencies in Π (black edges) or Γ (dashed edges)

$\hat{\pi} = 9\ -1\ 2\ 10\ 11\ 3\ 4\ 12\ 13\ 5\ 8\ 7\ 6\ 14$
$\hat{\gamma} = 9\ 1\ 2\ 3\ 4\ 10\ 11\ 5\ 6\ 7\ 8\ 12\ 13\ 14$



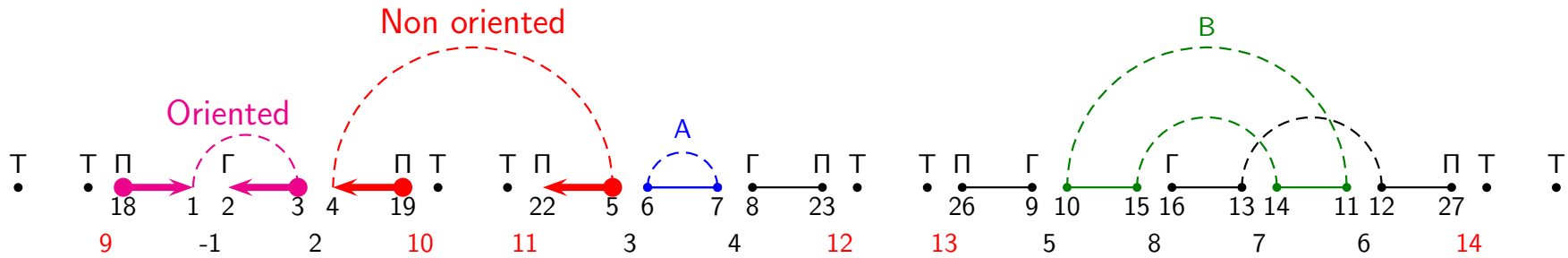
Distance computation independent of capping and concatenate
Particular vertices : Tails (T), Π-caps (Π) and Γ-tails (Γ)

Breakpoint graph

Cycle and path decomposition :

- ΠΠ-path, ΠΓ-path, ΓΓ-path.
- trivial cycles or paths (A) vs proper cycles or paths (B)

Orientation : arbitrary orientation of dark edges within a cycle or path



Number of rearrangements compared to eliminated breakpoints

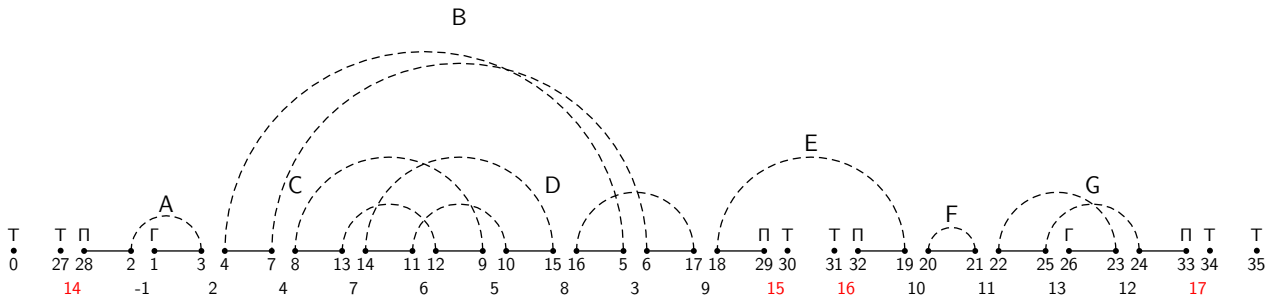
- 1 rearrangement for 1 or 2 breakpoints in oriented cycle
- 2 rearrangements for 1 or 2 breakpoints in non oriented cycle

Interleaving graph component classifications

Distance based on the breakpoint graph decomposition

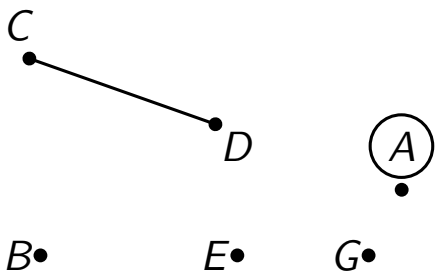
Incoherences and new notions in the literature

Clarification into a double classification (Jean & Nikolski 2007)



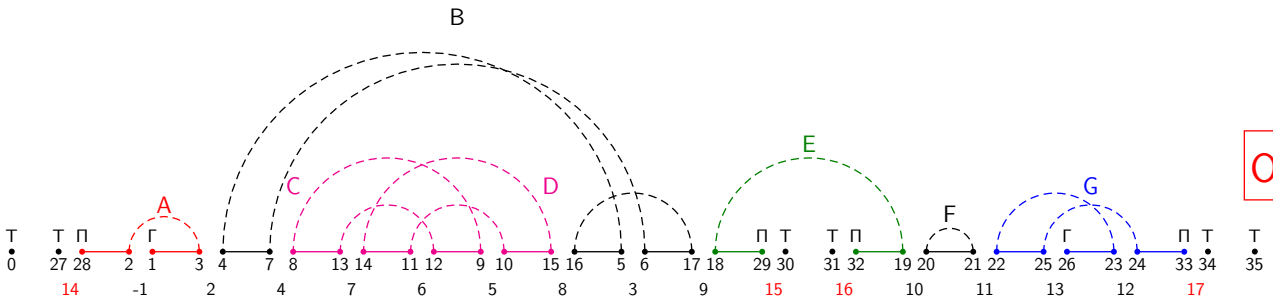
Connected components

Interleaving graph

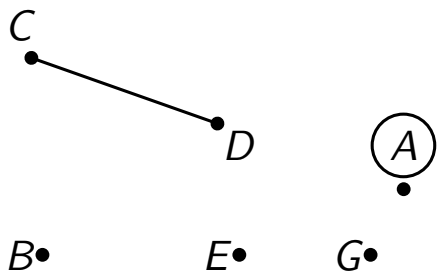


Interleaving graph component classifications

Proposition of a double classification

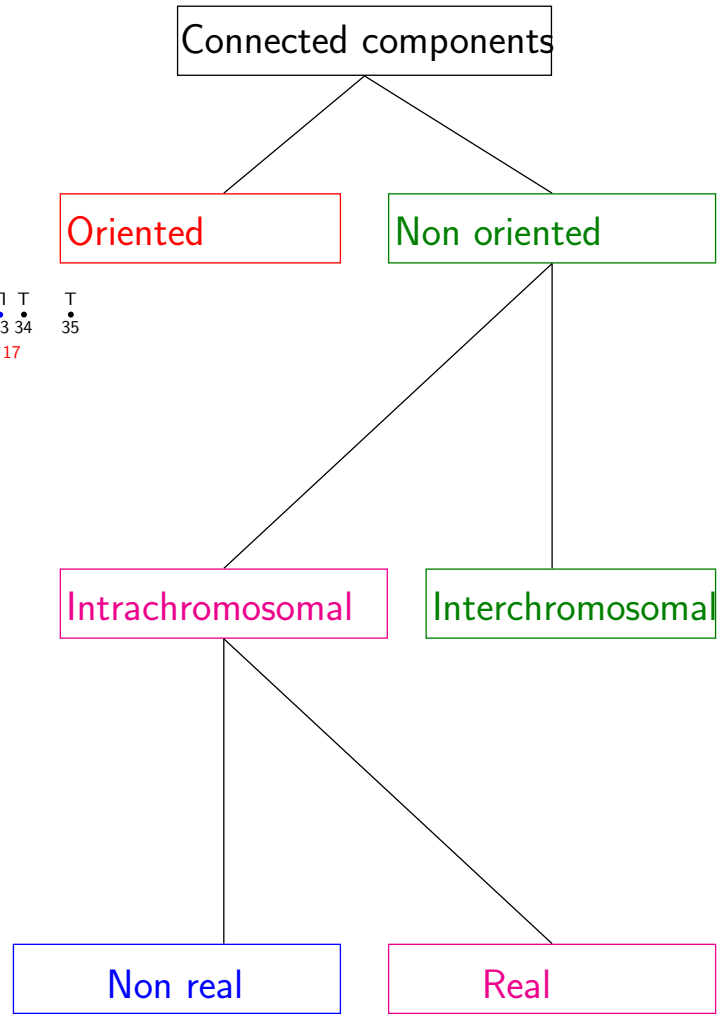


Interleaving graph



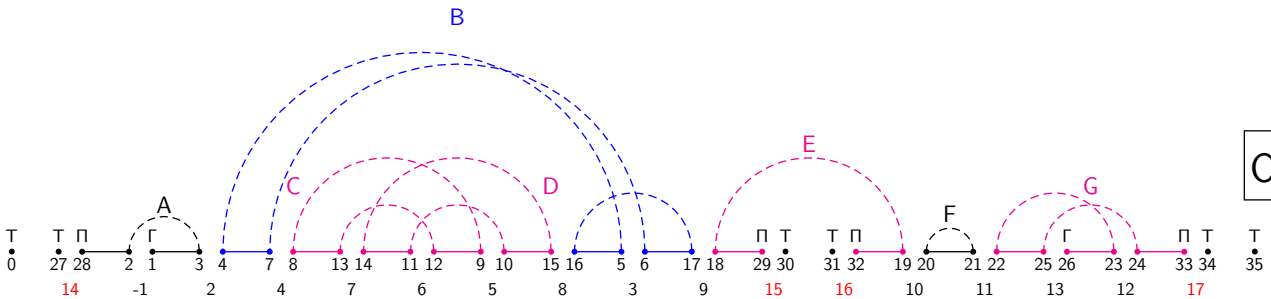
A component is real if it does not cover a Π-cap or a Γ-tail.

Intrinsic classification



Interleaving graph component classifications

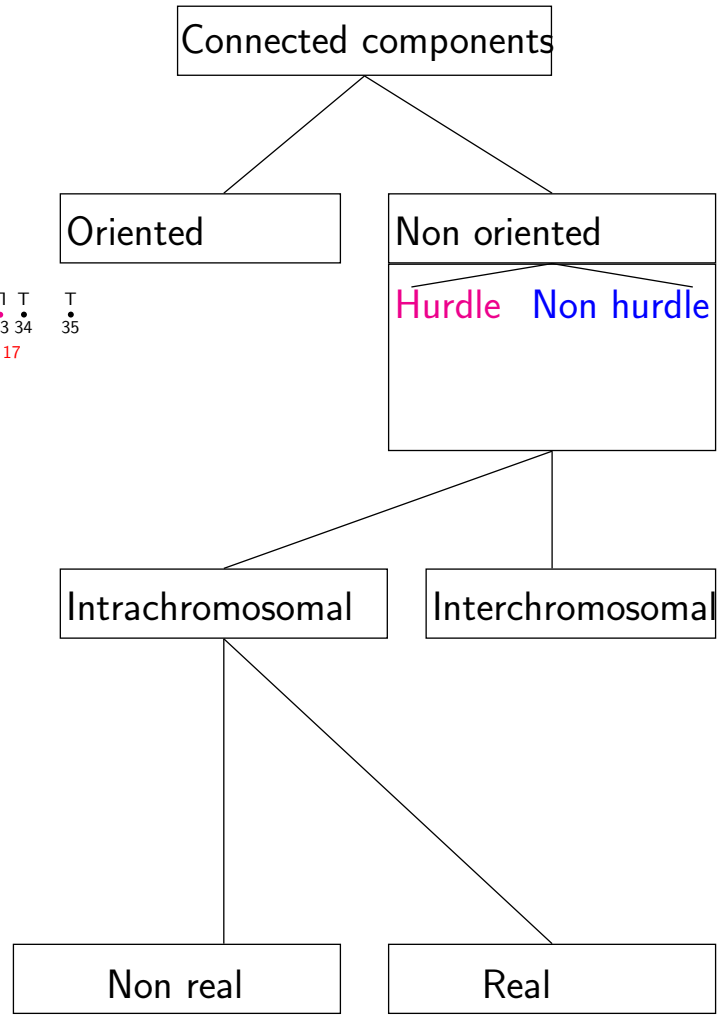
Proposition of a double classification



B separates $\{C, D\}$ from E or G

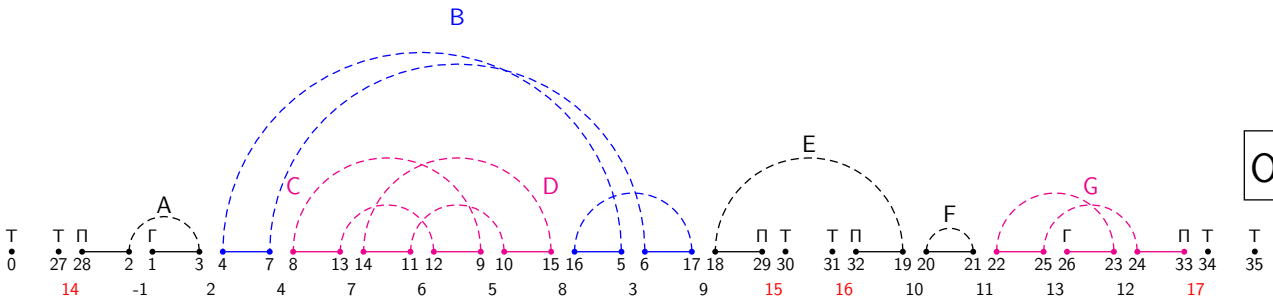
$\{C, D\}$ is a super hurdle
E and G are simple hurdles

Extrinsic classification



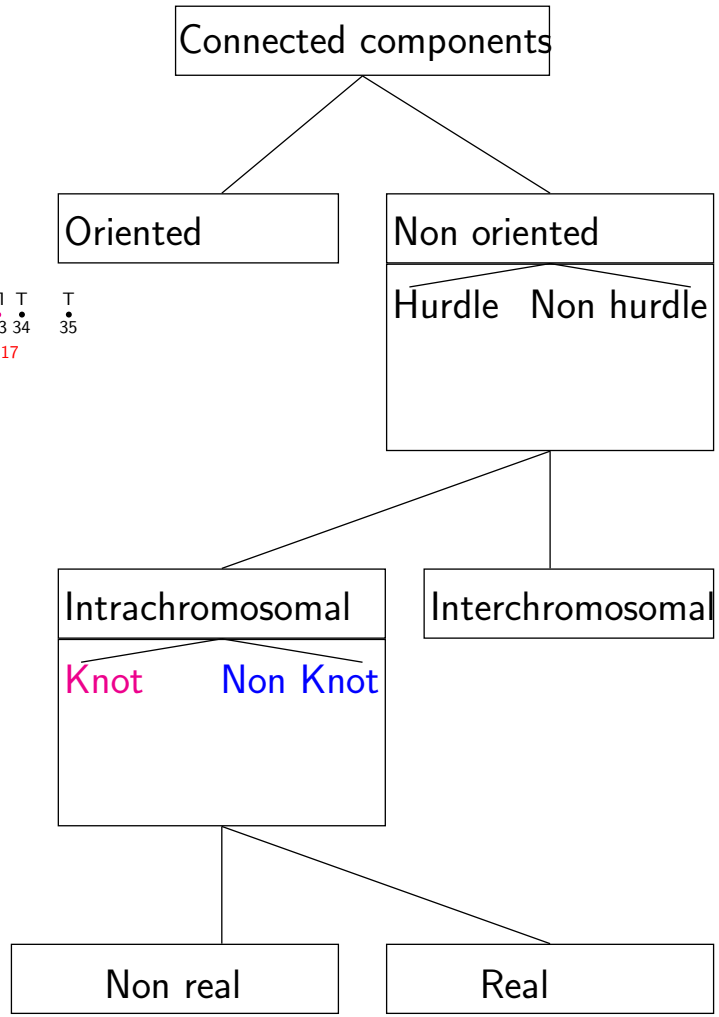
Interleaving graph component classifications

Proposition of a double classification



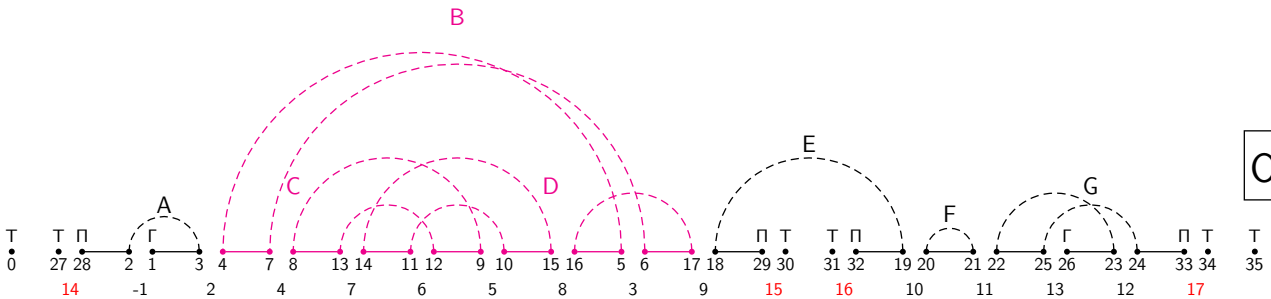
B separates $\{C, D\}$ from G

Extrinsic classification



Interleaving graph component classifications

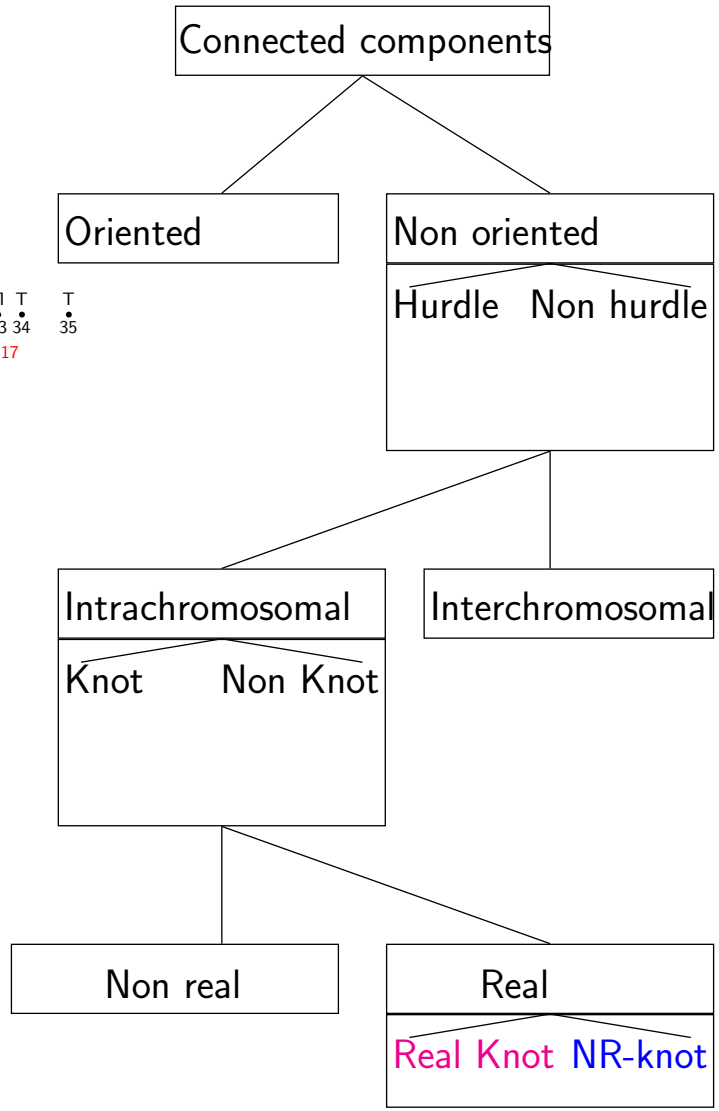
Proposition of a double classification



B is the greatest real-knot

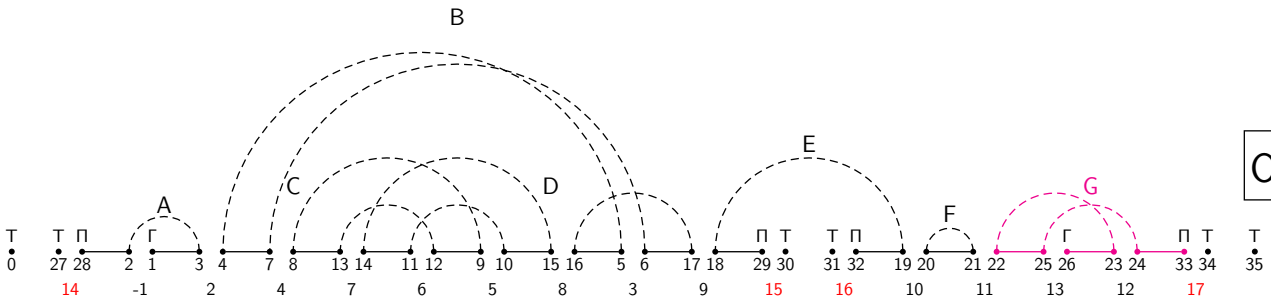
$\{C, D\}$ is a minimal real-knot

Extrinsic classification



Interleaving graph component classifications

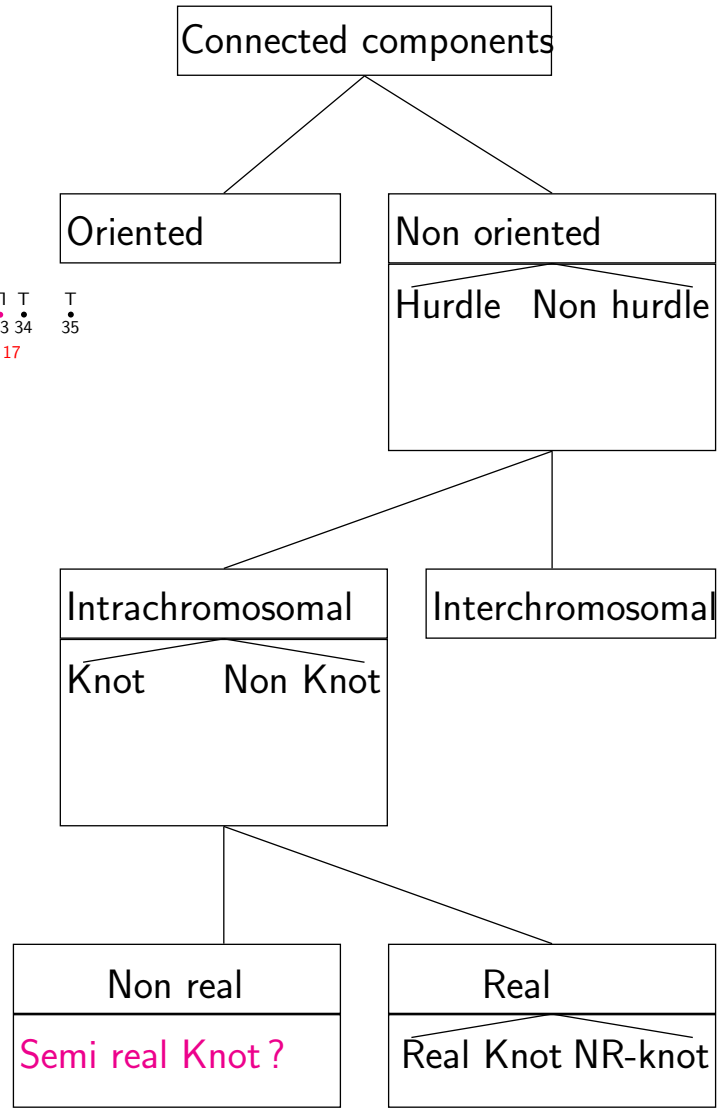
Proposition of a double classification



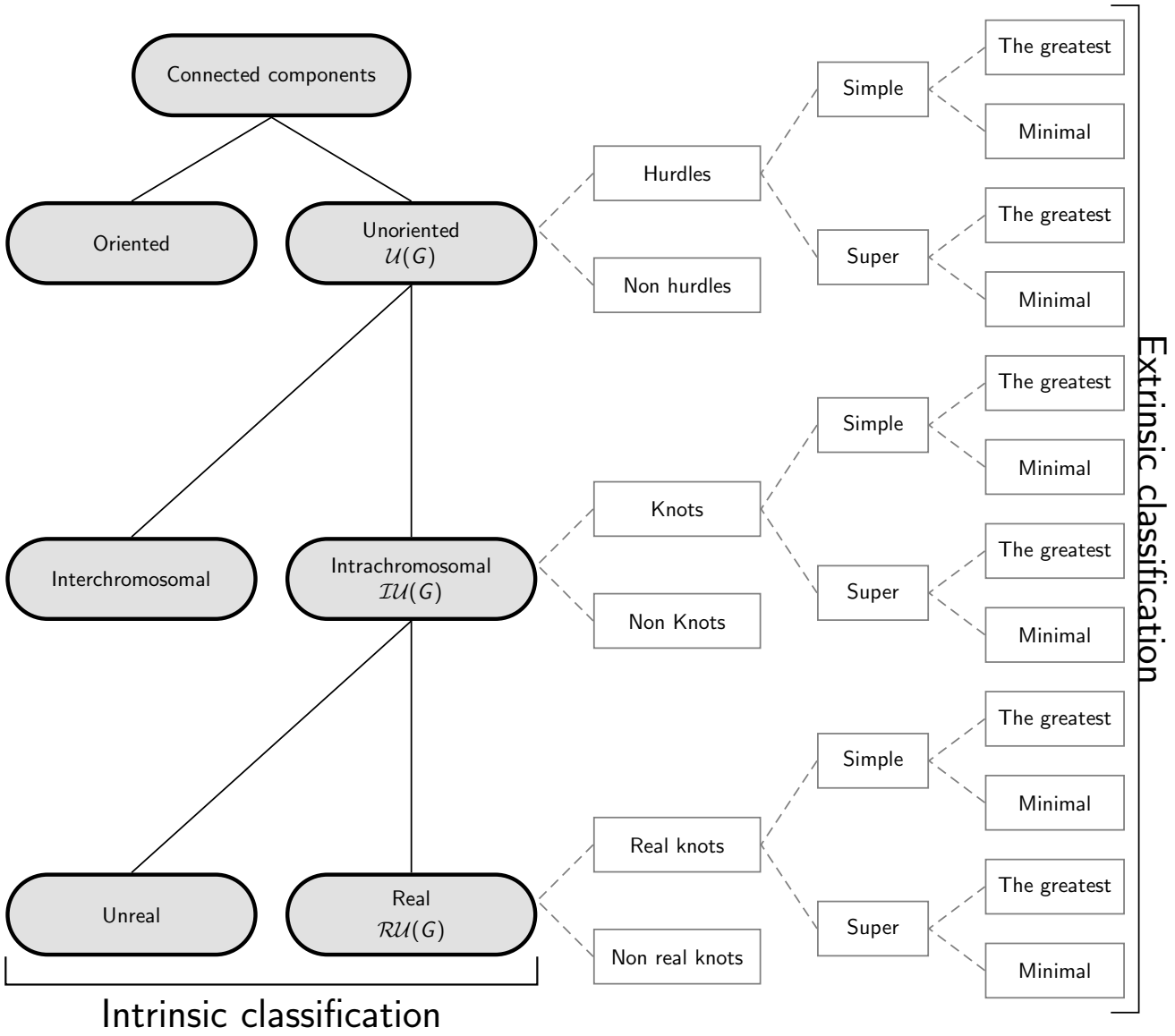
A **semi-real-knot** does not cover a ΓΓ-path and becomes a minimal real-knot or the simple greatest real knot after closing its ΠΓ-paths.

A **simple component** contains at least one ΠΓ-path and is not a semi-real-knot.

Extrinsic classification



Double classification



All of the configurations are encountered in distance and scenario computations

(Jean & Nikolski 2007)

Why optimal capping and concatenation are needed ?

Distance

arbitrary capping and concatenation

Scenario

sequence of rearrangements respecting minimal distance

optimal capping and concatenation

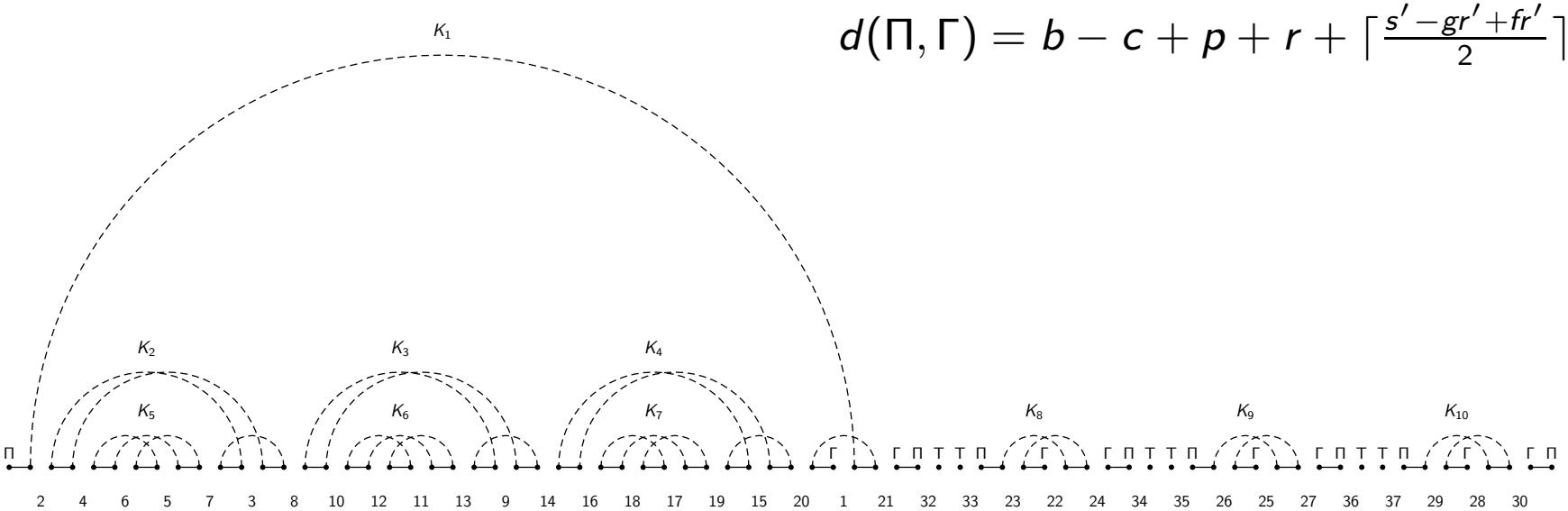
Reduction to the unichromosomal problem

- Reversals only
- Nonsense operations : chromosome flipping and cap reposition

Counterexample to the Ozery-Flato's and Shamir's algorithm (2003)

(Ozery-Flato & Shamir 2003)

$$d(\Pi,\Gamma) = b - c + p + r + \lceil \frac{s' - gr' + fr'}{2} \rceil$$

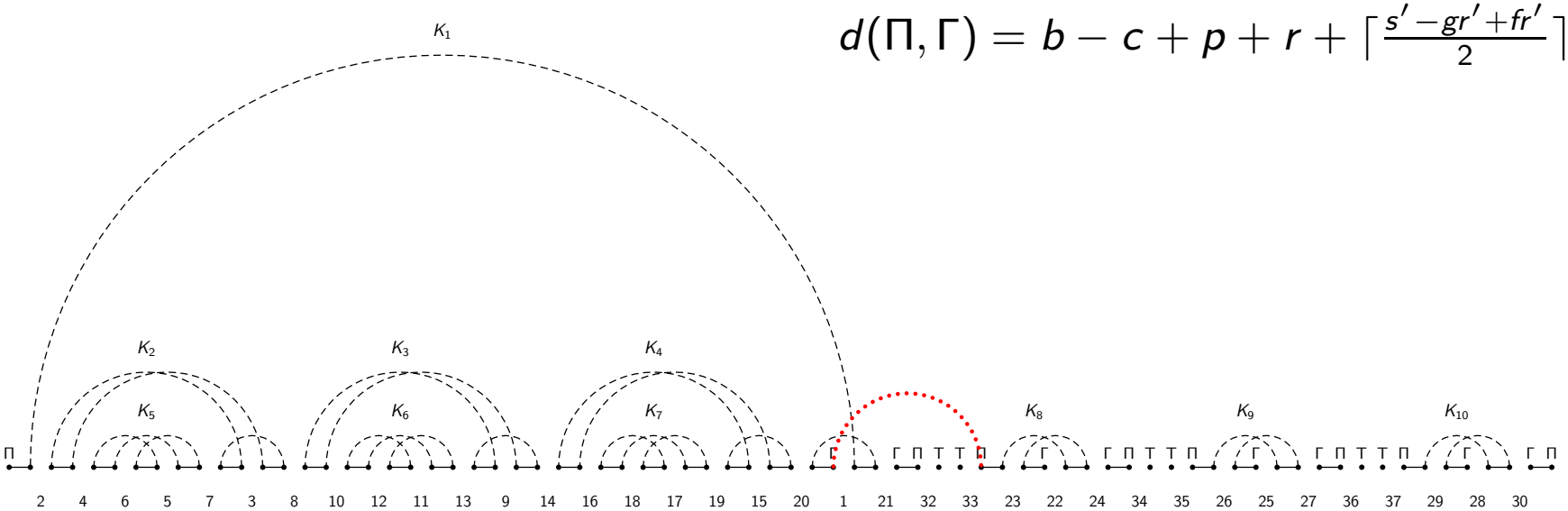


$$d(\Pi,\Gamma) = 34 - 14 + 0 + 3 + \lceil \frac{4-0+0}{2} \rceil = 25$$

Counterexample to the Ozery-Flato's and Shamir's algorithm (2003)

(Ozery-Flato & Shamir 2003)

$$d(\Pi,\Gamma) = b - c + p + r + \lceil \frac{s' - gr' + fr'}{2} \rceil$$



$$d(\Pi,\Gamma) = 34 - 14 + 0 + 3 + \lceil \frac{4-0+0}{2} \rceil = 25$$

$\Rightarrow$ cycle closure modifies the rearrangement distance so that it is no longer optimal

$$d = 34 - 13 + 0 + 3 + \lceil \frac{2-0+1}{2} \rceil = 26$$

A correct algorithm for optimal capping

Correct_Optimal_Capping

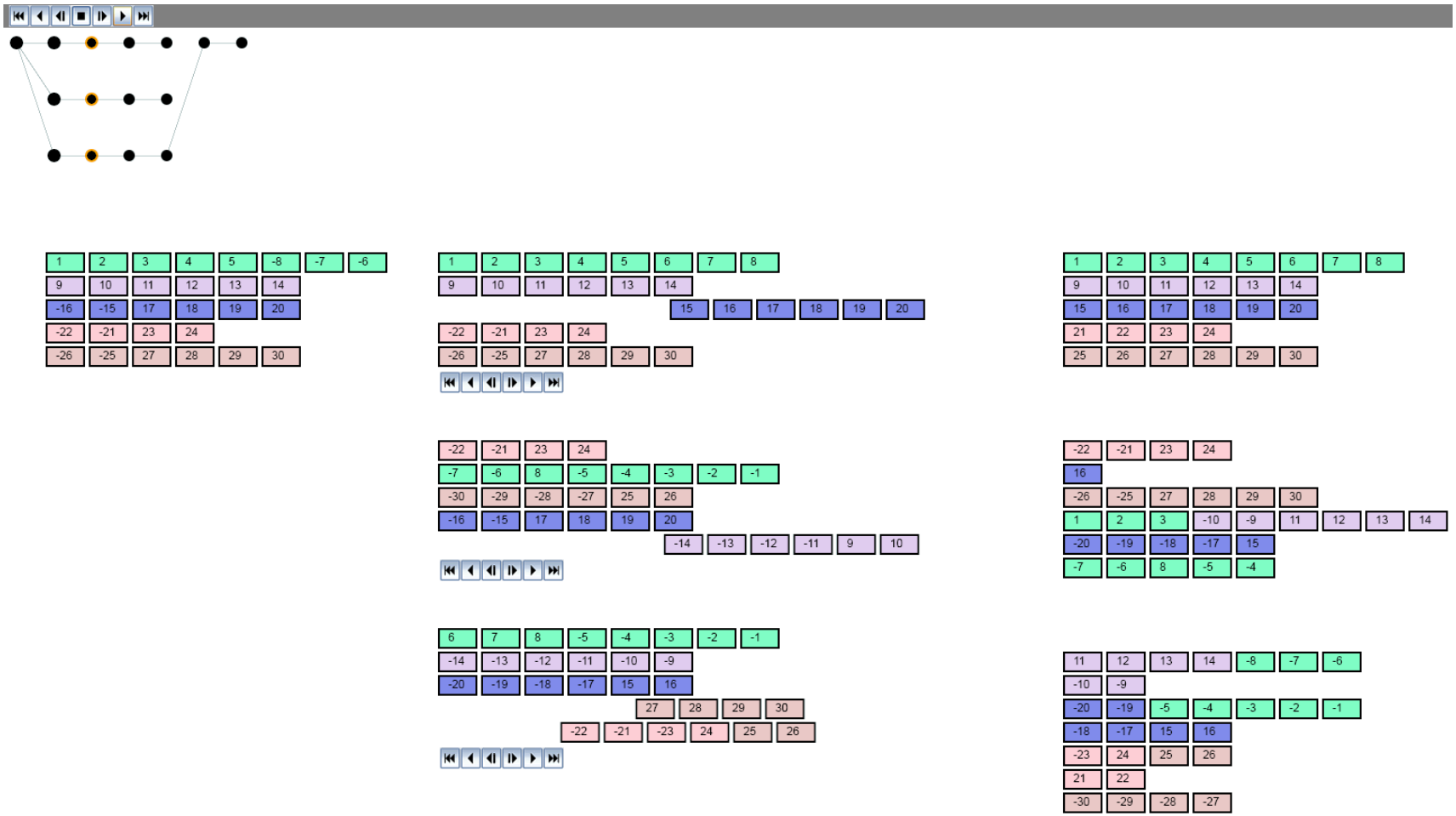
Construct the graph $G = G(\Pi, \Gamma)$
while there is a $\Gamma\Gamma$-path in G **do**
    Find an interchromosomal or oriented edge joining this $\Gamma\Gamma$-path with a $\Pi\Pi$-path and add it to G
end while
Close all remaining $\Pi\Pi$-paths in G
Close all $\Pi\Gamma$-paths in simple components in G
if s' is even and $s' \geq 2$ and G is a fortress-of-real-knots **then**
    if G has the semi-greatest-real-knot **then**
        Close all $\Pi\Gamma$-paths in the semi-greatest-real-knot
    else
        Close all $\Pi\Gamma$-paths in any one semi-real-knot
    end if
end if
while G has more than one semi-real-knot **do**
    Find an interchromosomal or oriented edge joining $\Pi\Gamma$-paths in any two semi-real-knot and add it to G
end while
Close all remaining $\Pi\Gamma$-paths in G
Find a capping $\hat{\Gamma}$ defined by the graph $G(\hat{\Pi}, \hat{\Gamma})$

Theorem (Jean & Nikolski 2007)

Let $d = d(\Pi, \Gamma)$ be the distance between two multichromosomal genomes Π and Γ. Algorithm Correct_Optimal_Capping constructs an optimal capping $\hat{\Gamma}$ for any arbitrary capping $\hat{\Pi}$, such that $d(\hat{\Pi}, \hat{\Gamma}) = d$.

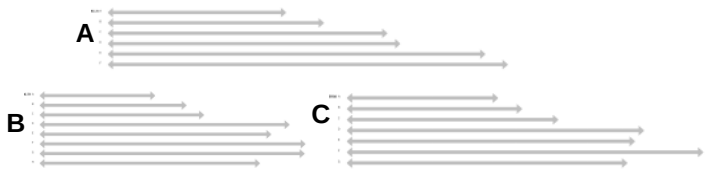
VIRAGE

A new interactive tool for the visualization of rearrangements from scenarios

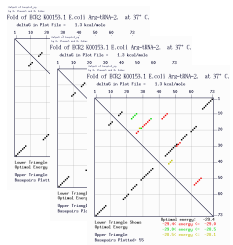


Conclusions and perspectives

Conclusion

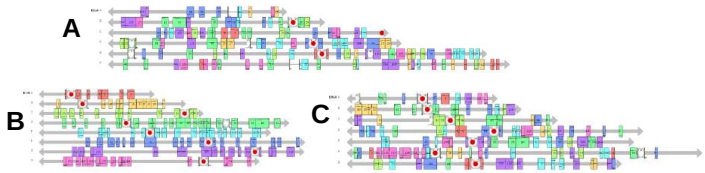


genomes



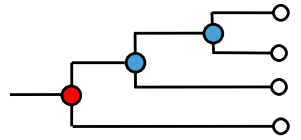
Common markers

SyDiG algorithm:
Identification of common markers in distant genomes



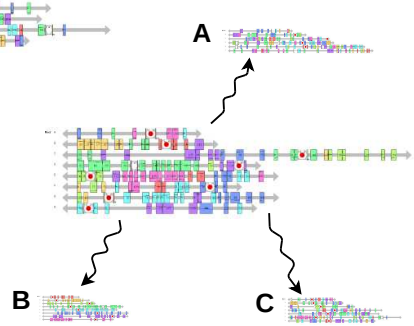
Comparative maps

Parsimonious scenario:
Suggestion of a synthesis for component classification
Introduction of a correct algorithm for optimal capping with its proof
Development of a visualization tool: VIRAGE



Rearrangement tree

Super-block method:
Based on an adjacency analysis and a rearrangement study
Results: all the sets of possible partial assemblies
Centromeres positions can be take into the account



Ancestral architecture

Conclusion

Development and validation of the methods on real data :

Complete framework for Hemiascomycetes yeasts

- Identification of common markers
- Ancestral hypothesis from comparative maps
- Interactive visualization of possible scenarios

Perspectives

Theoretical perspectives :

- Method extension to genomes with duplicates and different gene content
- Adding centromere position constraint in parsimonious scenarios
- Whole reconstruction of the species tree by recovering the root and internal nodes

Biological perspectives :

- Application to the *Drosophilia* twelve (Stark et al. 2007)
- Medical interest : application to 5 species phylogenetically close to *Candida glabrata* (available soon)

"In Amelie's rearrangement world"

Acknowledgments





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CNRS

ACI IMPBIO

Special thanks to ENSEIRB students for the first version of VIRAGE
And thank you for your attention !

Distance formula

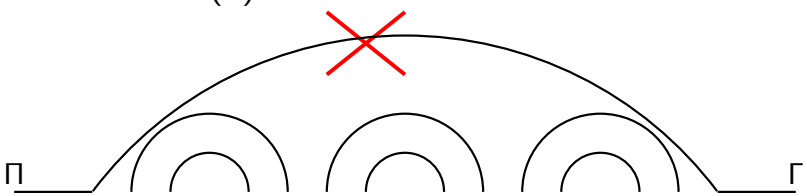
Theorem (Ozery-Flato & Shamir 2003)

$$d(\Pi, \Gamma) = b - c + p + r + \lceil \frac{s' - gr' + fr'}{2} \rceil$$

b is the number of genes plus the number of chromosomes (max)
c is the number of cycles and paths
p is the number of ΓΓ-paths
r is the number of real-knots
s' is the number of semi-real-knots

gr' $= 1$ if the greatest-real-knot exists and $s' > 0$, $gr' = 0$ else.
fr'$= 1$ if (a) or (b), fr'$=0$ else.

(a) fortress of real-knots



(b) weak fortress of real-knots