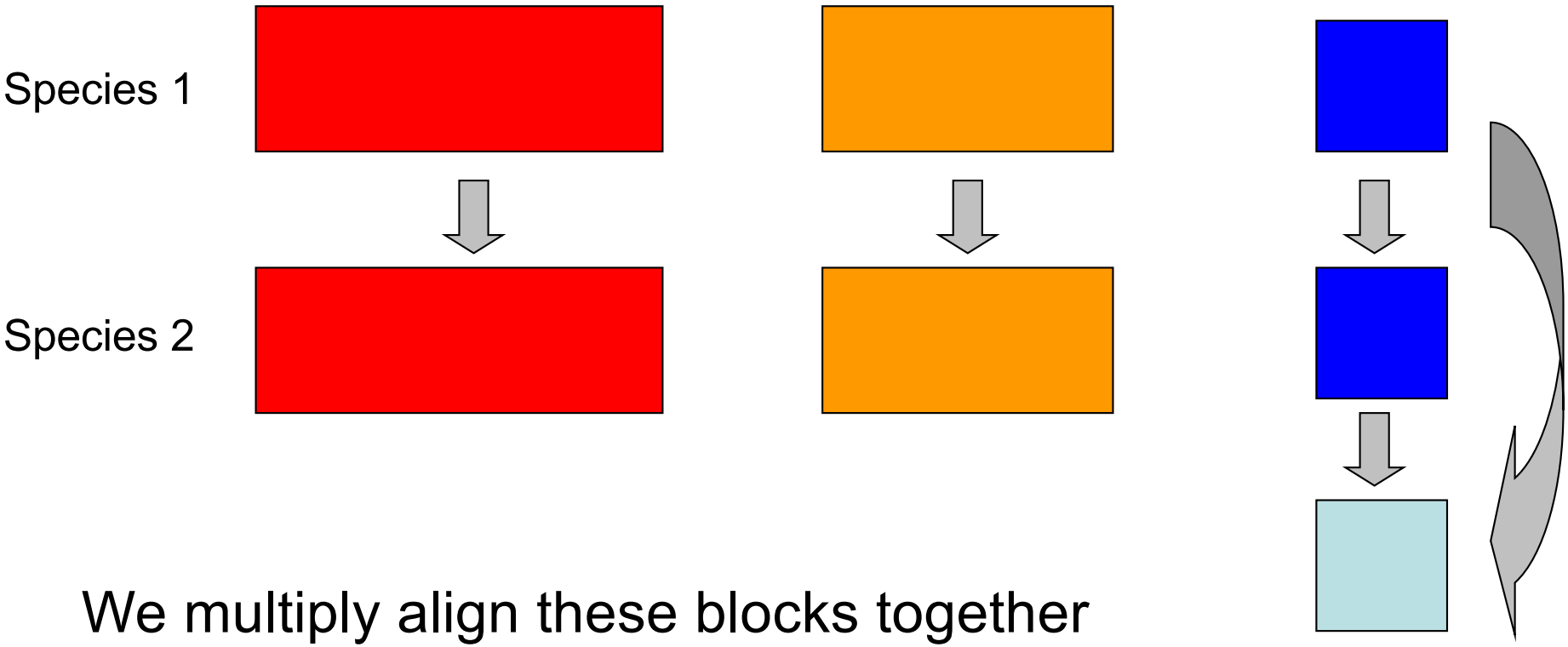
Whole genome alignment

Applications of genome alignment

- Comparing different genome assemblies
- Locating genome duplications and conserved segments
- Gene finding through comparative genomics
- Analyzing pathogenic bacteria against their harmless close relatives

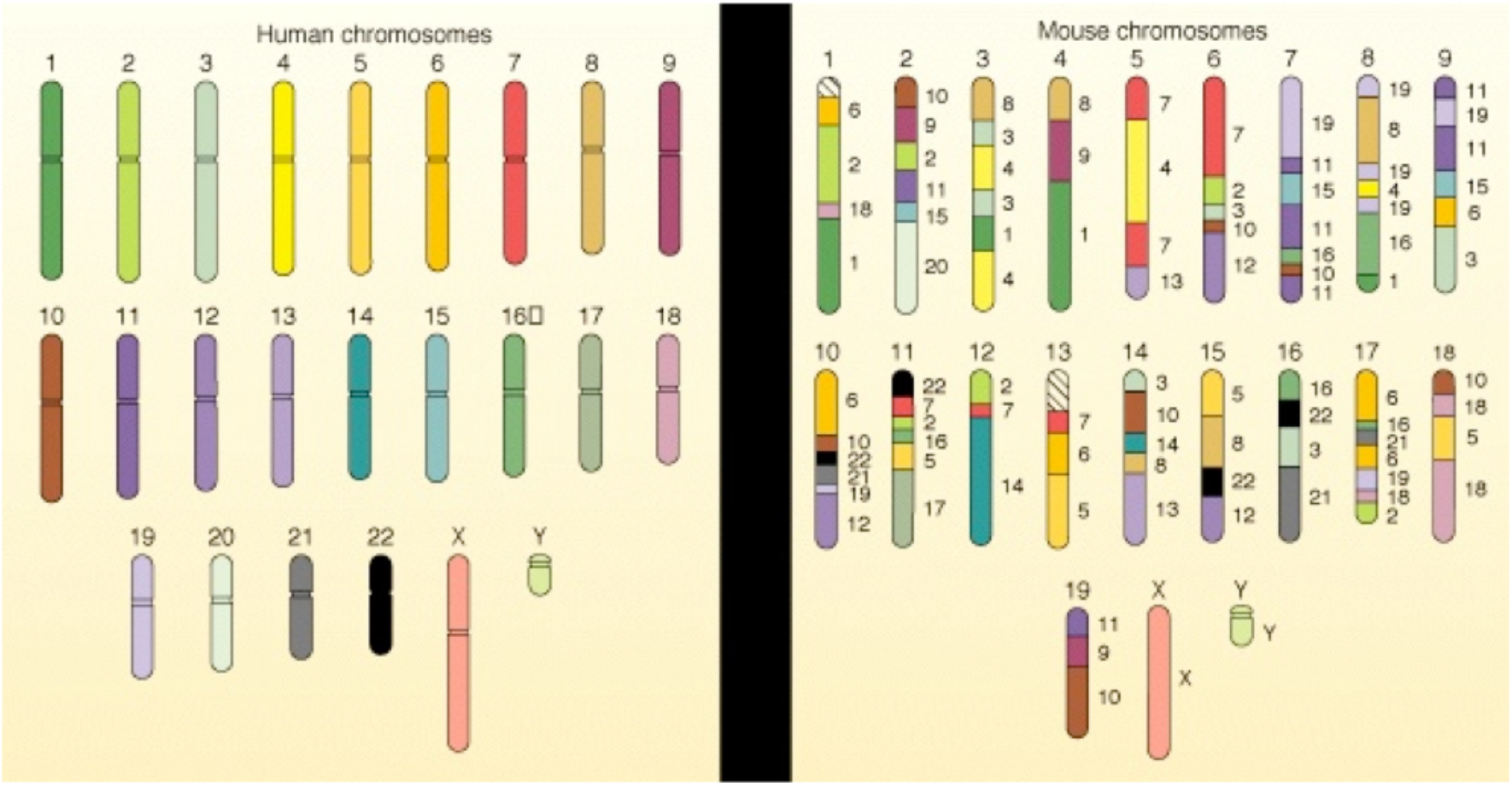
Homology map



We multiply align these blocks together

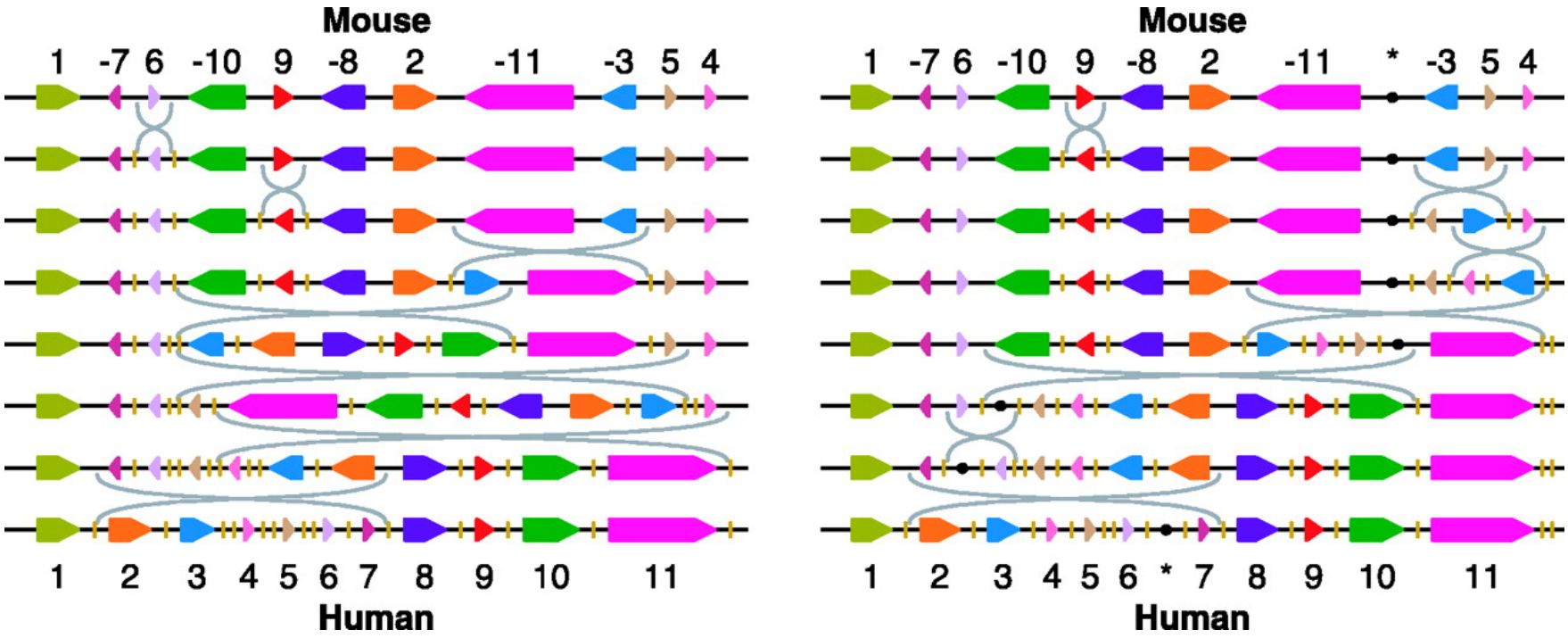
Overview/Goals

- Input:
  - Set of whole genomes, which may differ by substitutions, indels and rearrangements
  - Uses open reading frames or other gene predictions
- Output:
  - One alignment per region of genomes that has not been “shuffled”
    - Two genomes = global
    - > 2 genomes = multiple



http://fig.cox.miami.edu/Faculty/Dana/synteny.jpg

Two different most parsimonious scenarios that transform the order of the 11 synteny blocks on the mouse X chromosome into the order on the human X chromosome



Pevzner P., Tesler G. PNAS 2003;100:7672-7677

Whole-genome alignment

- Advanced data structures can also be used to efficiently speed up genomic alignments of closely-related organisms.

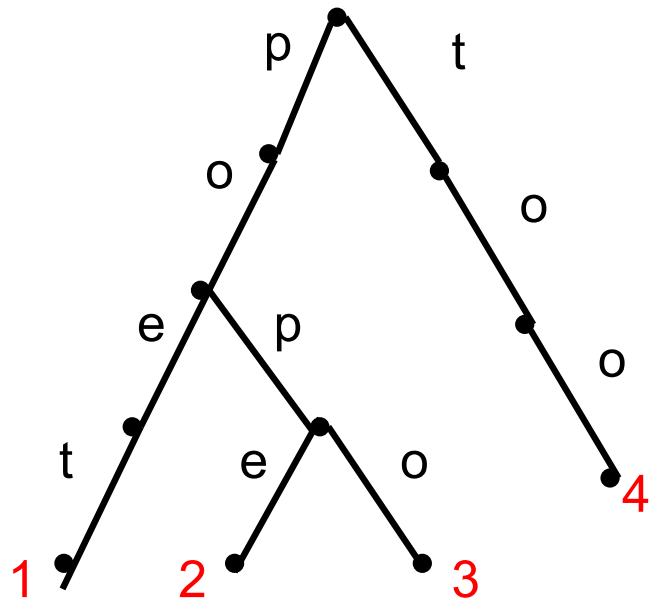
- We will introduce suffix trees and the MUMmer algorithm before going into detail next week.

Suffix trees

- Specialized form of keyword trees/tries
- Key idea:
  - preprocess text T, not pattern P
    - O(m) preprocess time
    - O(n+k) search time
      - k is number of occurrences of P in T

Keyword Tree

- P = {poet, pope, popo, too}



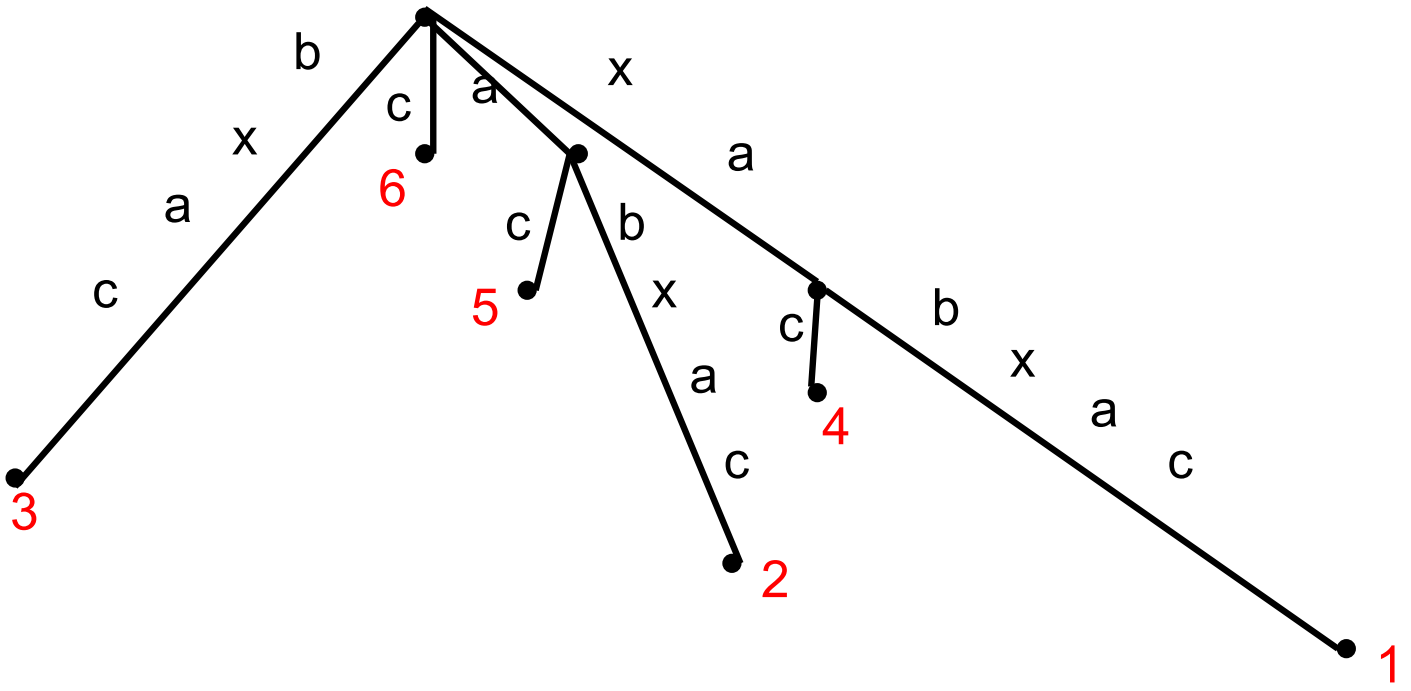
Suffix Tree

- Take any *m* character string S like xabxac
- Set of keywords is the set of suffixes of S
  - {xabxac, abxac, bxac, xac, ac, c}

- Changes relative to keyword trees:
  - Assumption: no suffix is a prefix of another suffix (can be a substring, but not a prefix)
    - Assure this by adding a character $ to end of S
  - Internal nodes except root must have at least 2 children

Example suffix tree

- {xabxac, abxac, bxac, xac, ac, c}



Notation to keep track of

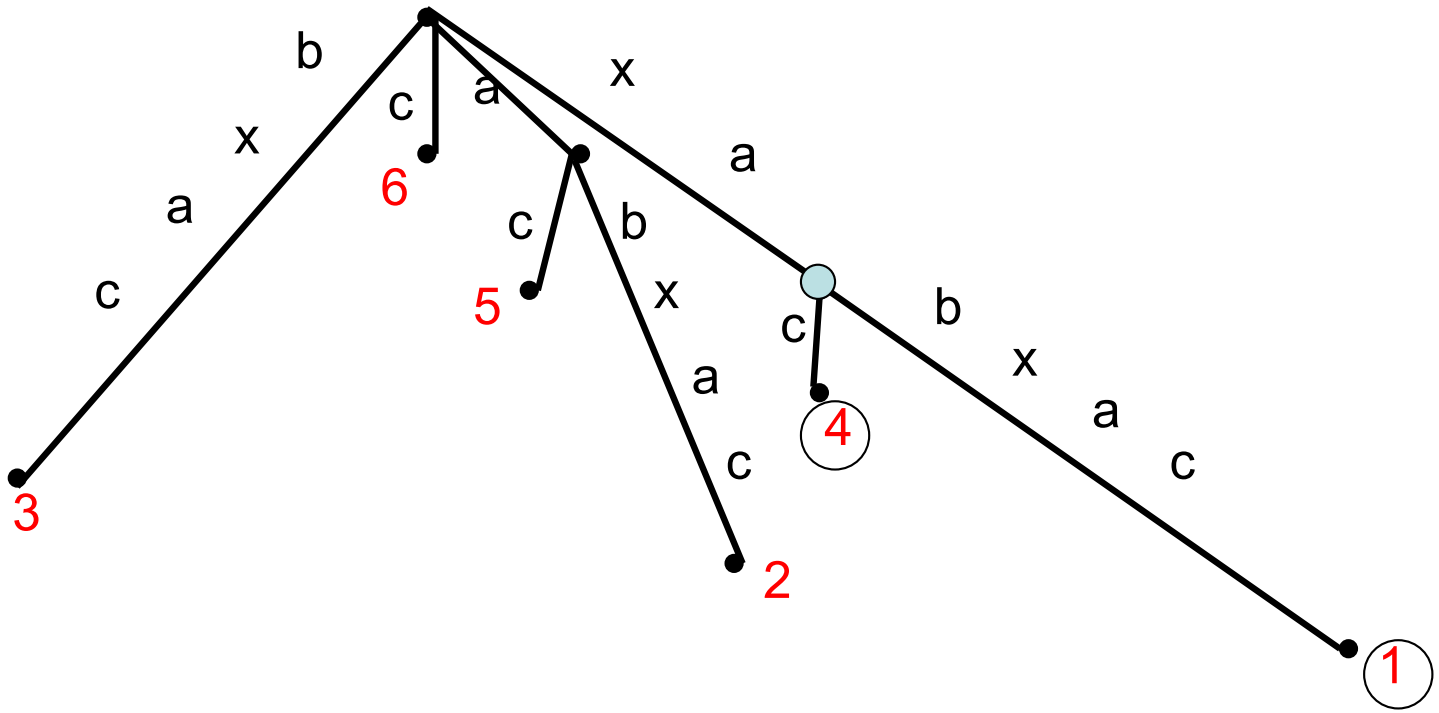
- Label of a path from root r to a node v is simply the concatenation of labels on edges from r to v
- label of a node v is L(v)
  - path label from r to v
- string-depth of v
  - number of characters in v's label L(v)

Using suffix trees in exact matching

- Build suffix tree for text T
- Match pattern P against tree starting at root until
  - Case 1, P is completely matched
    - Every leaf below this match point is the starting location of P in T
  - Case 2: No match is possible
    - P does not occur in T

Illustration

- T = xabxac
  - suffixes ={xabxac, abxac, bxac, xac, ac, c}
- Pattern P_1: xa
- Pattern P_2: xb



In-class example

- S = xabxabdeabhixab$
- xabxacdefghixab$
- abxacdefghixab$
- bxacdefghixab$
- xacdefghixab$
- …
- $

Building trees: $O(m^2)$ algorithm

- Initialize
  - One edge for the entire string S[1..m]$
- For i = 2 to m
  - Add suffix S[i..m] to suffix tree
    - Find match point for string S[i..m] in current tree
    - If in “middle” of edge, create new node w
    - Add remainder of S[i..m] as edge label to suffix i leaf
- Running Time
  - O(m-i) time to add suffix S[i..m]

Running Time Analysis

- Build suffix tree:
  - Will show this is O(m)
  - This is preprocessing
- Search time:
  - O(n+k) where k is the number of occurrences of P in T
  - O(n) to find match point if it exists
  - O(k) to find all leaves below match point

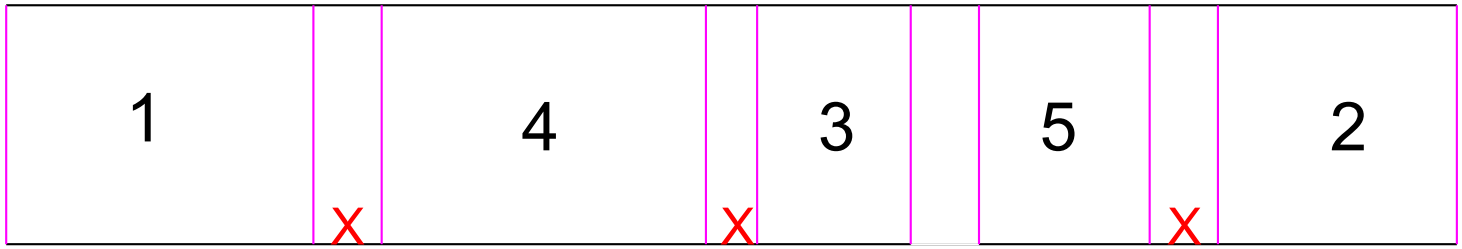
Why suffix trees are important in genome alignment

- Long unique matches have a high probability of being included in the final genomic alignment.

- We need to set the minimum length high-enough, however, to avoid random noise.
  - MUMs = maximal unique matches
  - MEMs = maximal exact matches

Overview

Genome A



Genome A’

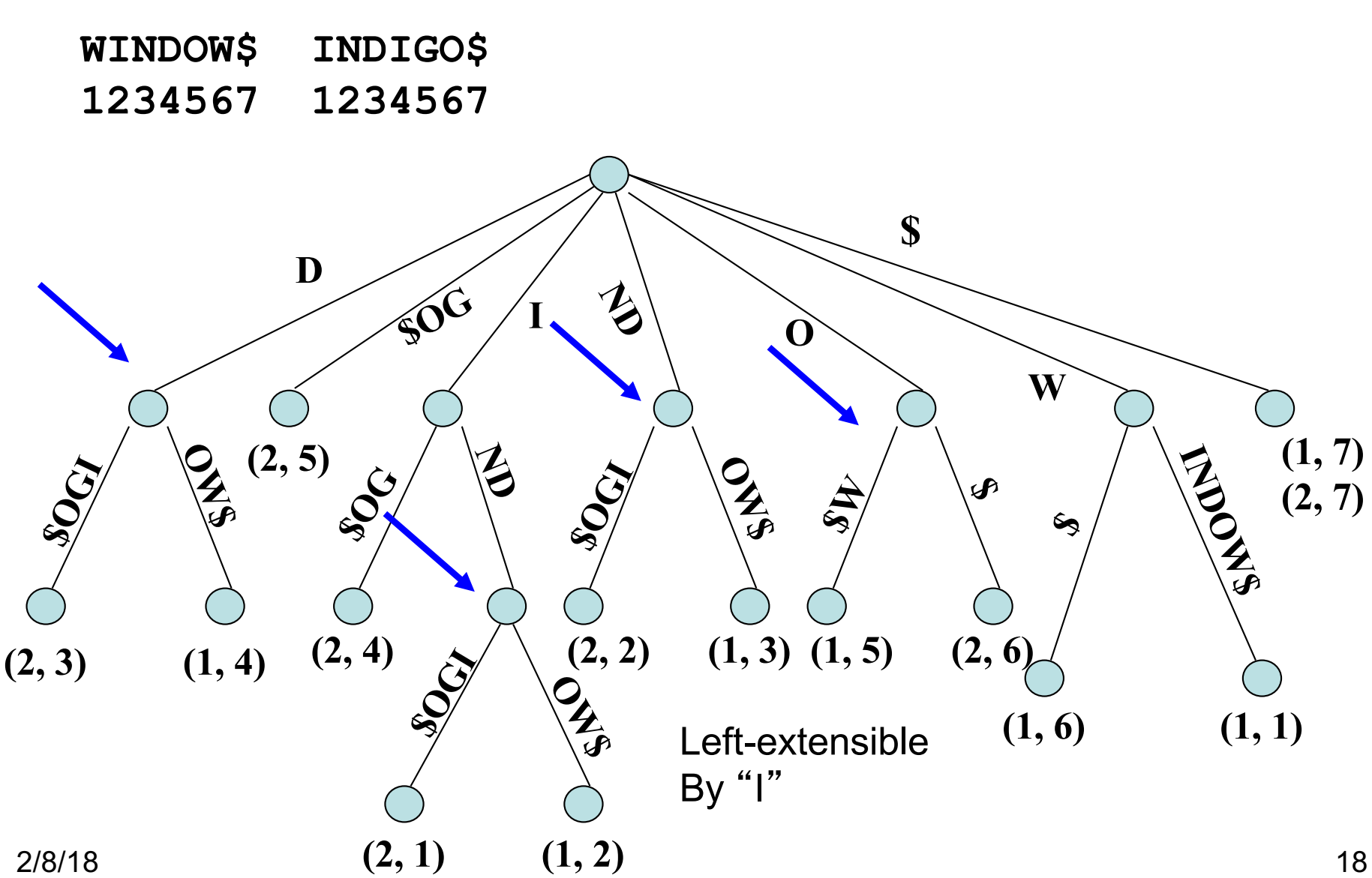
We have 5 matches that can not be extended to left or right

We have 4 gaps to fill between these matches

MUM-based alignments

- MUMs are by definition unique maximal matches in both sequences
  - Originally required building a generalized suffix tree of both genomes
  - Internal nodes w/ only two leaves, one from each input, are unique and not right-extensible
  - Check for left-extensibility, then go!

Maximal Unique Matches

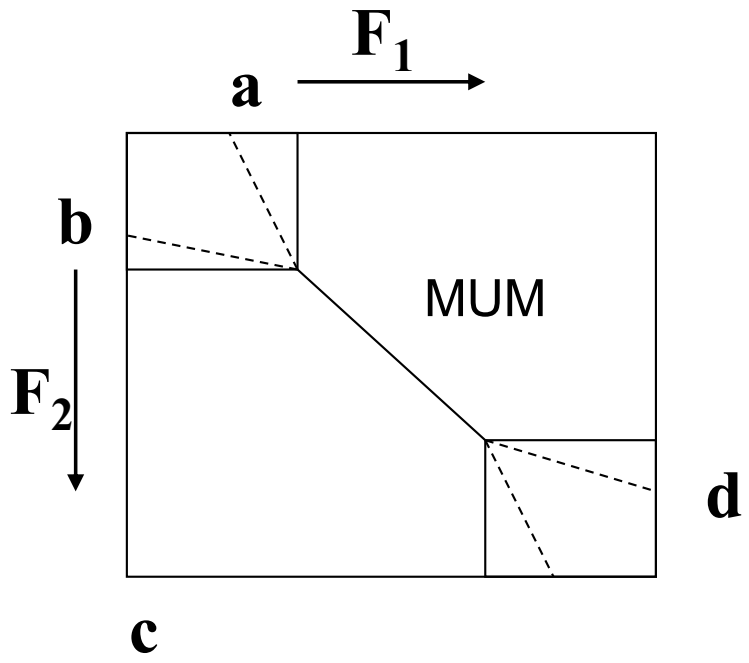


Early whole genome alignment algorithms

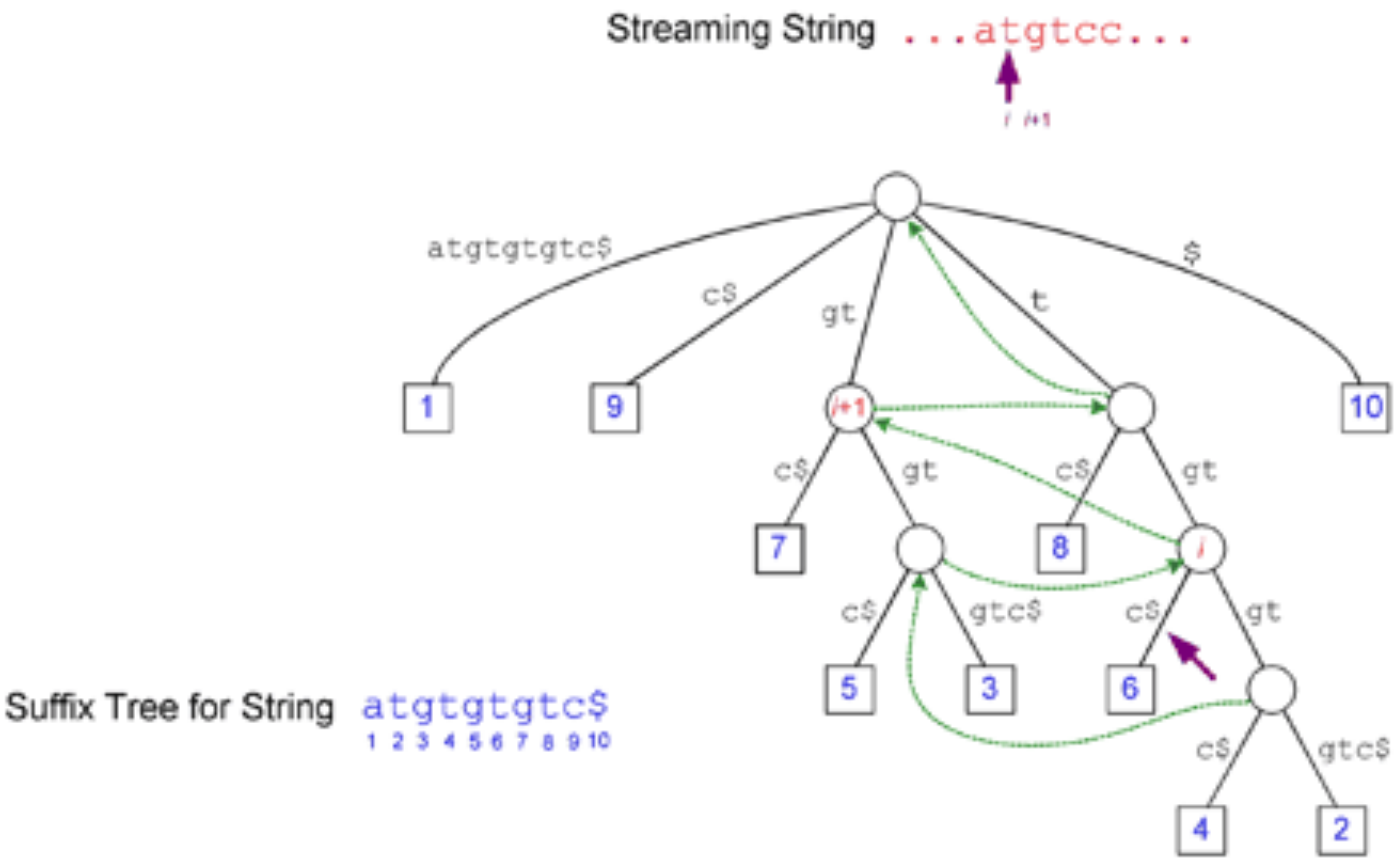
- Arranged MUMs relative to one genome using Longest Increasing Subsequence (LIS) algorithm

- Filled in small gaps using dynamic programming
  - Space inefficient for large gaps

Banded Dynamic Programming



– Compute only lower and upper rectangles based on desired percent similarity



Suffix links are in green

From Delcher et al., 2002, Nucleic Acids Res30(11):2478-83

Applications

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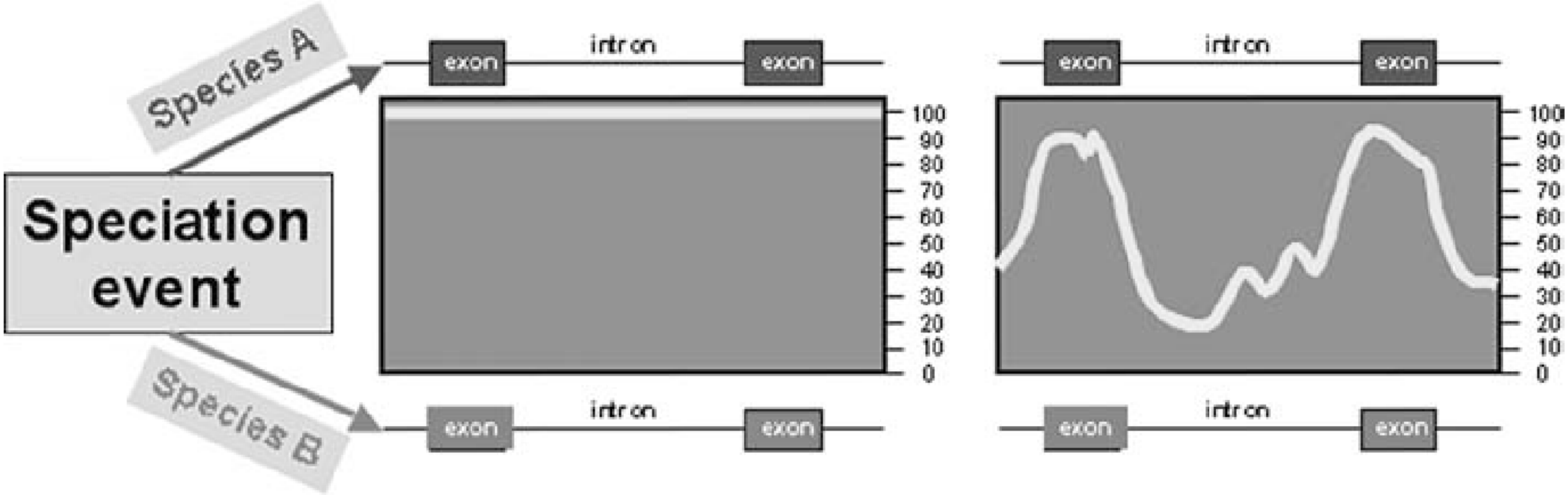


From: Miller et al. Annu. Rev. Genom. Human. Genet. 2004.5:15-56.

BLASTZ

- Modification of BLAST for whole-genome alignment of close species (i.e. human-mouse)
- Optimized for intron-exon discovery.
- Two differences with gapped BLAST:
  - Matching regions can be restricted to occur in same order and orientation.
  - Uses a special scoring matrix that limits false positive alignments in low complexity regions.

Optimization

- Two changes to BLASTZ significantly improved its execution speed.
- If the software realizes that many regions of the mouse genome align to the same human segment,that segment is marked so that it will be ignored in later steps
- Second,the idea of Ma et al. (2002) where for runs of 19 consecutive within which the 12 positions indicated by a 1 in the string 111010011100101011111 are identical.

Results

- Data:
  - human genome into ~3000 segments (1 MB each)
  - Divided mouse genome into 100 30MB segments
- Run time:
  - 481 CPU days
  - 0.5 days on a 1,024 processor cluster
  - 20 GB of output

MUMmer 2.0

- Improved space implementation of suffix tree using a few tricks (17 bytes/base)
- Introduced banded dynamic programming and advanced clustering to tackle larger gaps
- Used suffix tree “streaming” of multiple queries against a reference

MUMmer 2

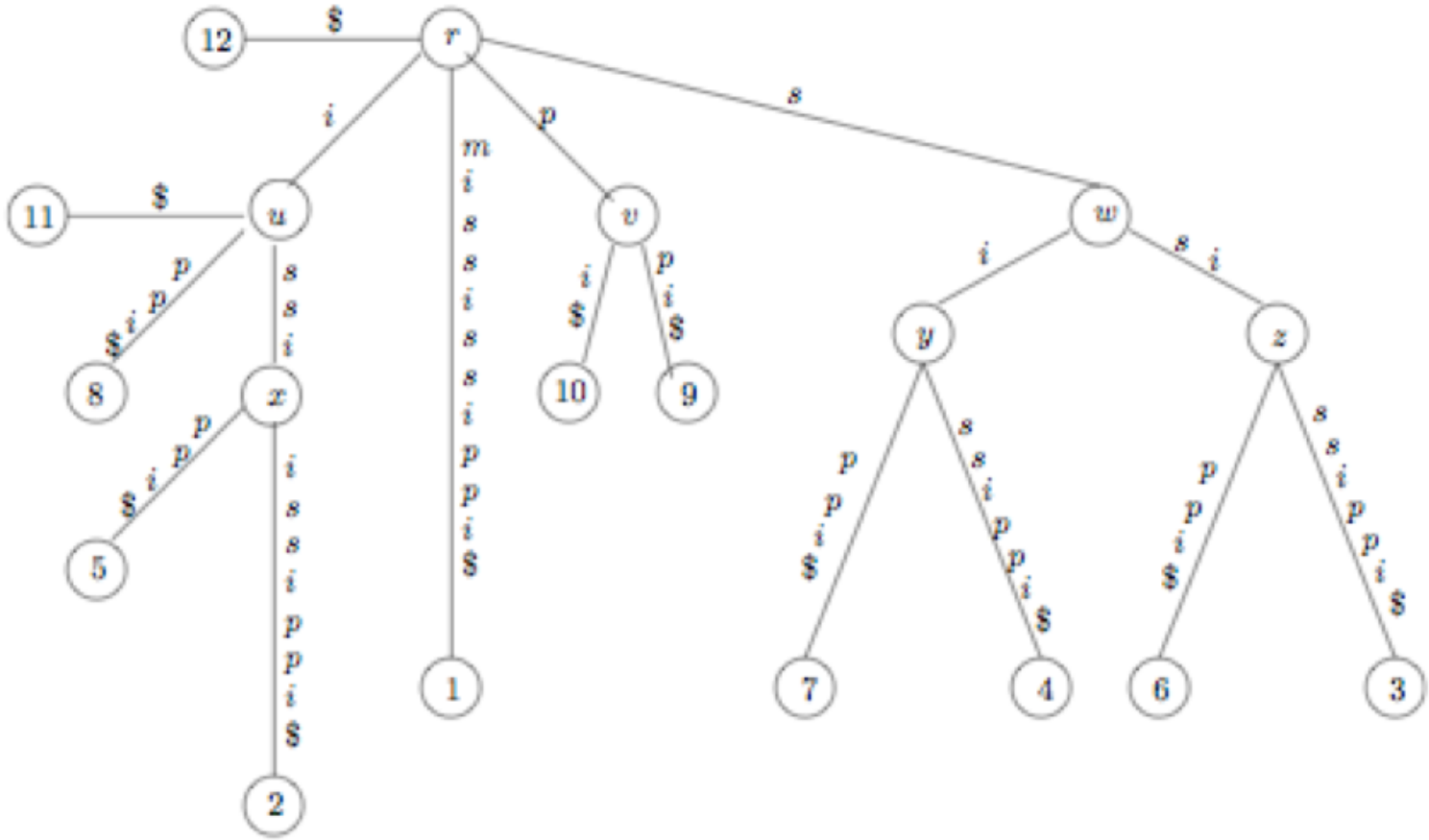
- Three times faster
- One-third memory usage
- Support protein sequence and multiple sequences.
- Entire human chromosomes

- Can align **millions** of nucleotides in **a few minutes** on **a desktop computer**.

Linear time of suffix arrays

- There were three papers in 2002 that solved the old problem of constructing suffix arrays in linear time.

- These were:
  - Ko and Aluru – very interesting, but hard to understand
  - Kim et al. – was based on older parallel suffix tree algorithms
  - Karakkanen and Sanders is the simplest and most elegant.



SA	12	11	8	5	2	1	10	9	7	4	6	3
Lcp	0	1	1	4	0	0	1	0	2	1	3	

FIGURE 1.1: Suffix tree, suffix array and Lcp array of the string *mississippi*. The suffix links in the tree are given by $x \to z \to y \to u \to r$, $v \to r$, and $w \to r$.