


Introduction to Bioinformatics


Bioinformatics

GUEST LECTURE :

Phylogenetic Analysis

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Overview

1. Introduction

2. On trees and evolution

3. Inferring trees

4. Combining multiple trees

5. Case study : the phylogenetic analysis of SARS

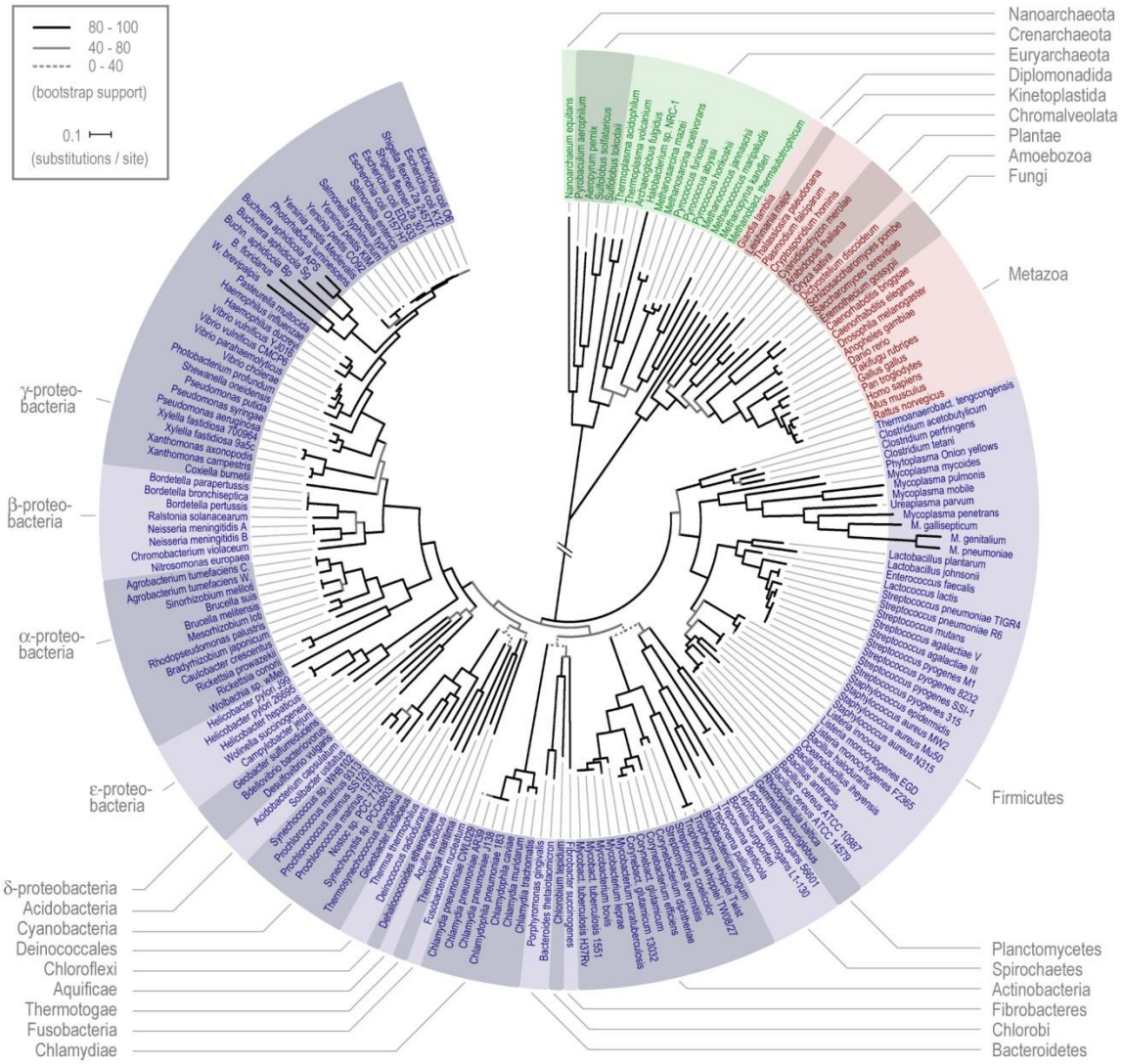
6. References and recommended reading

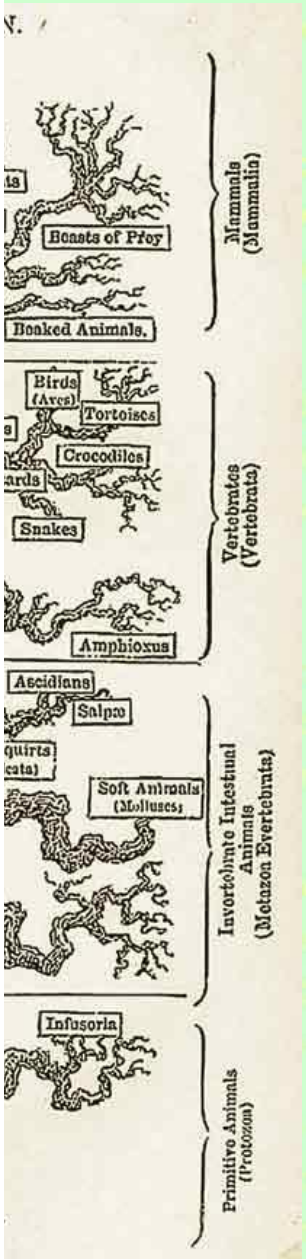
On trees and evolution

* Traditionally, the evolutionary history connecting any group of (related) species has been represented by an **evolutionary tree**

* The analysis of the evolutionary history involving evolutionary trees is called **Phylogenetic Analysis**

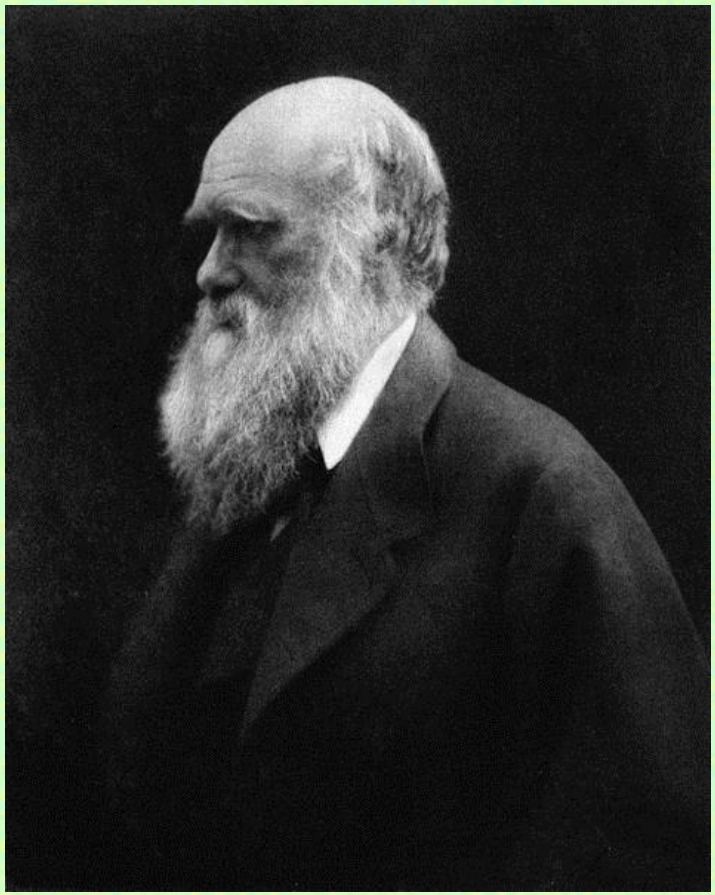
PHYLOGENETIC TREES

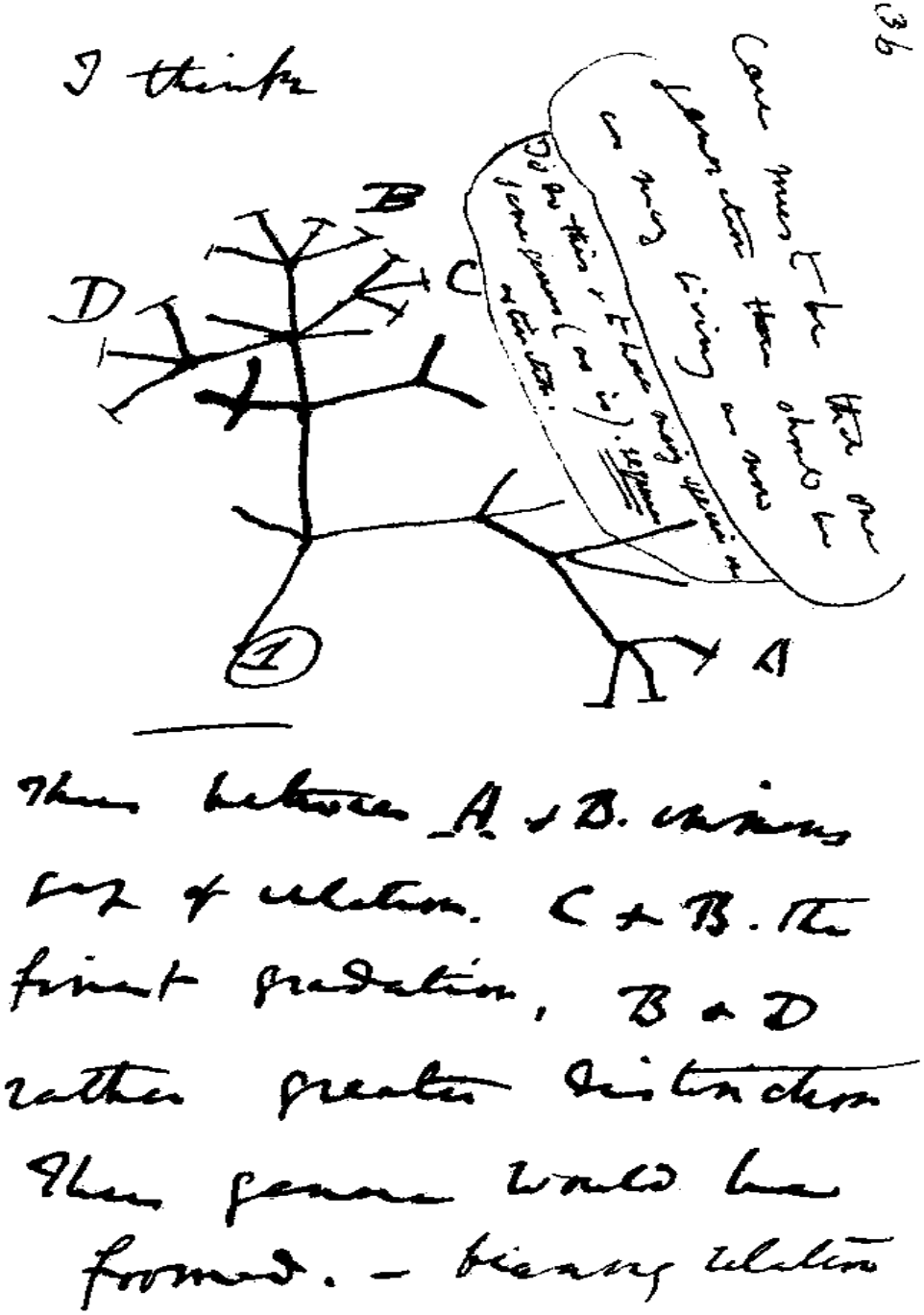




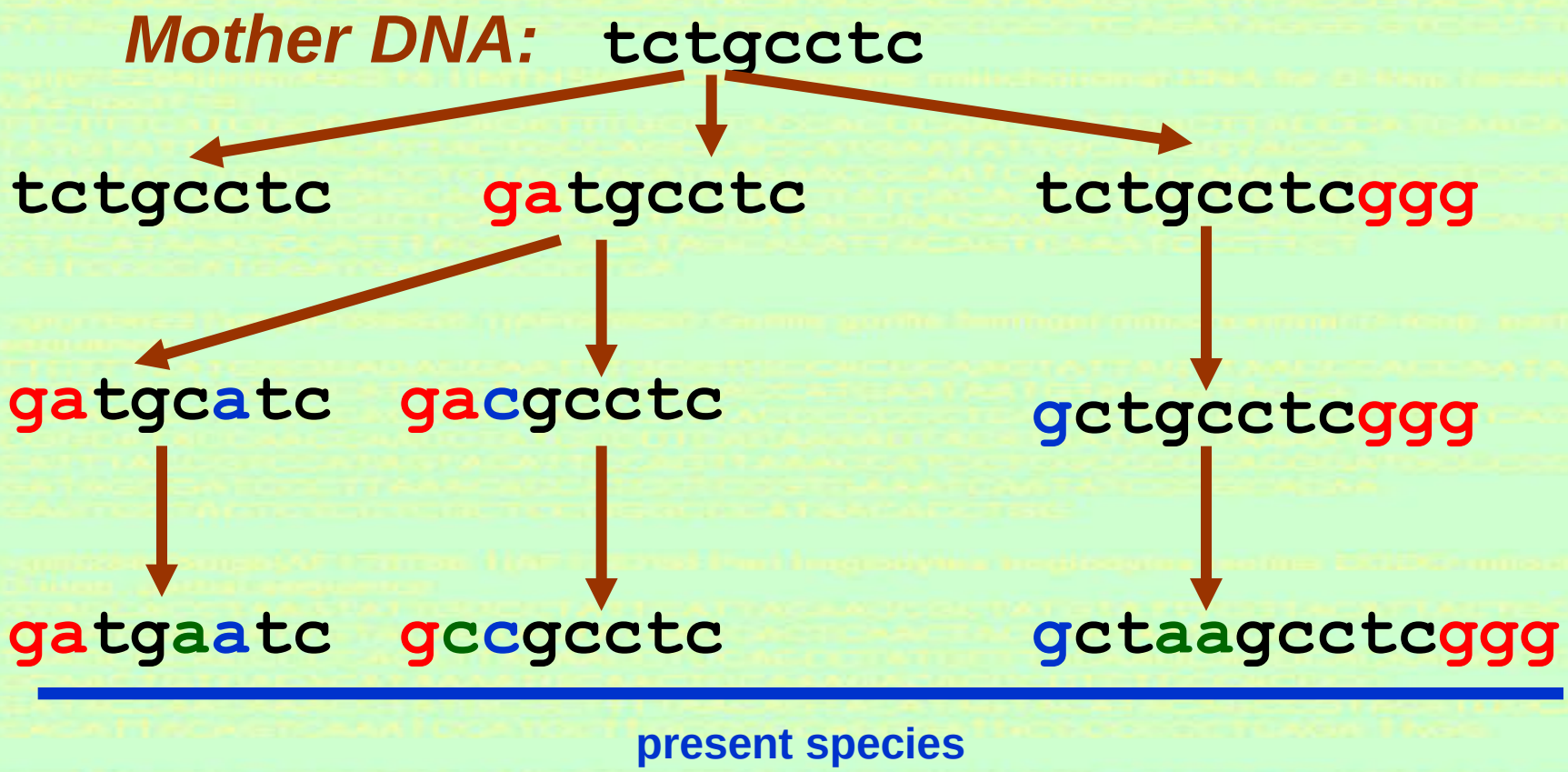
PHYLOGENETIC TREES

The only *figure* in Darwin's "On the origin of species" is a tree.





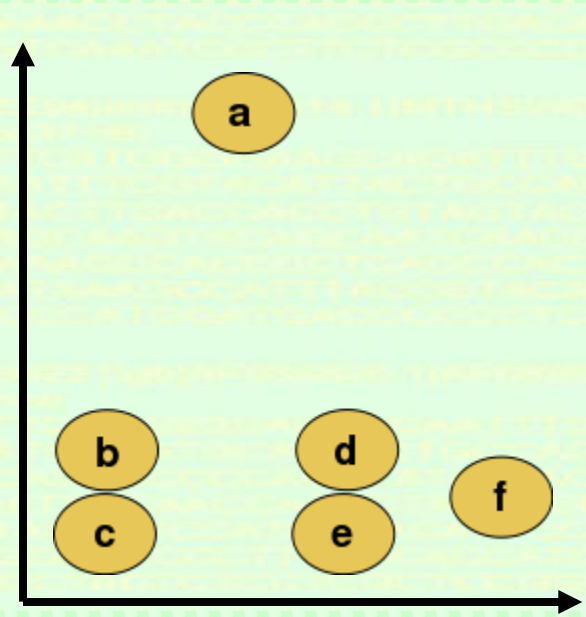
The biological basis of evolution



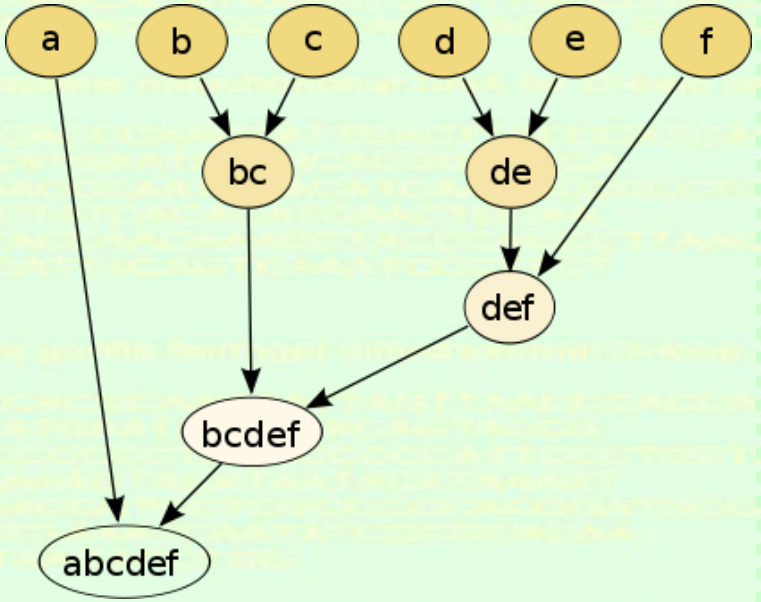
Phylogenetics

phylogenetics is the study of evolutionary relatedness among various groups of organisms (e.g., species, populations).

Visualizing phylogenetic relations

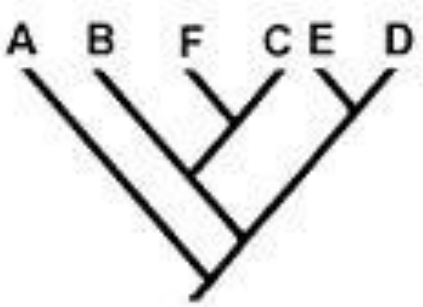


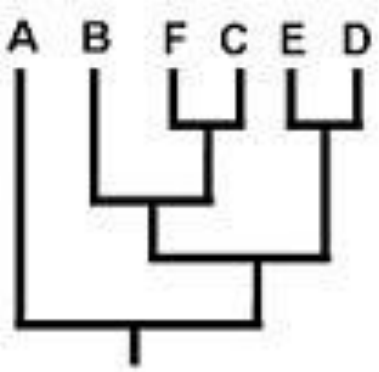
Multi-Dimensional Scaling
(MDS map)

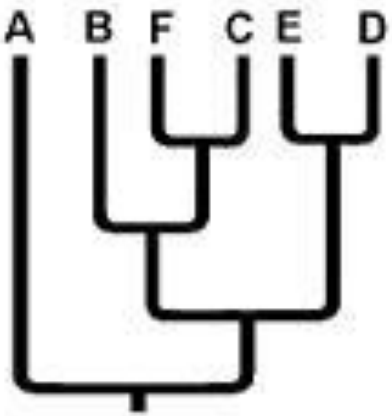


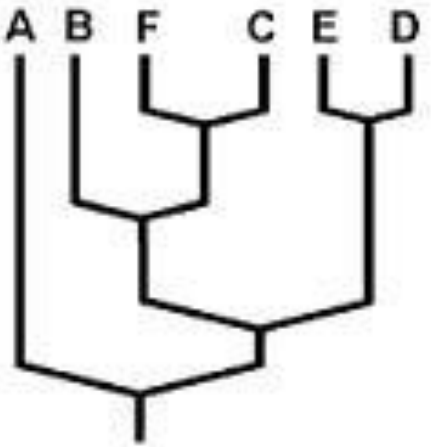
Dendrogram

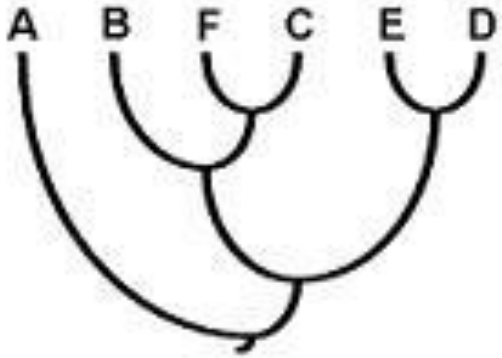
Visualizing phylogenetic relations

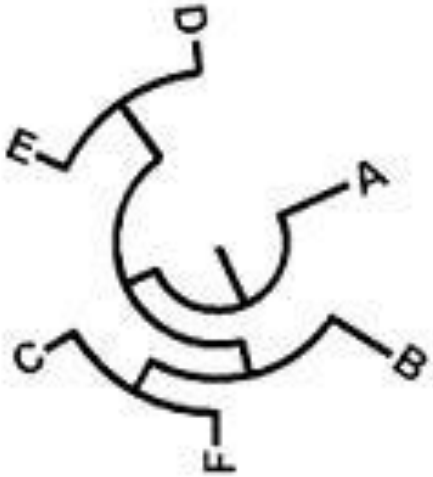








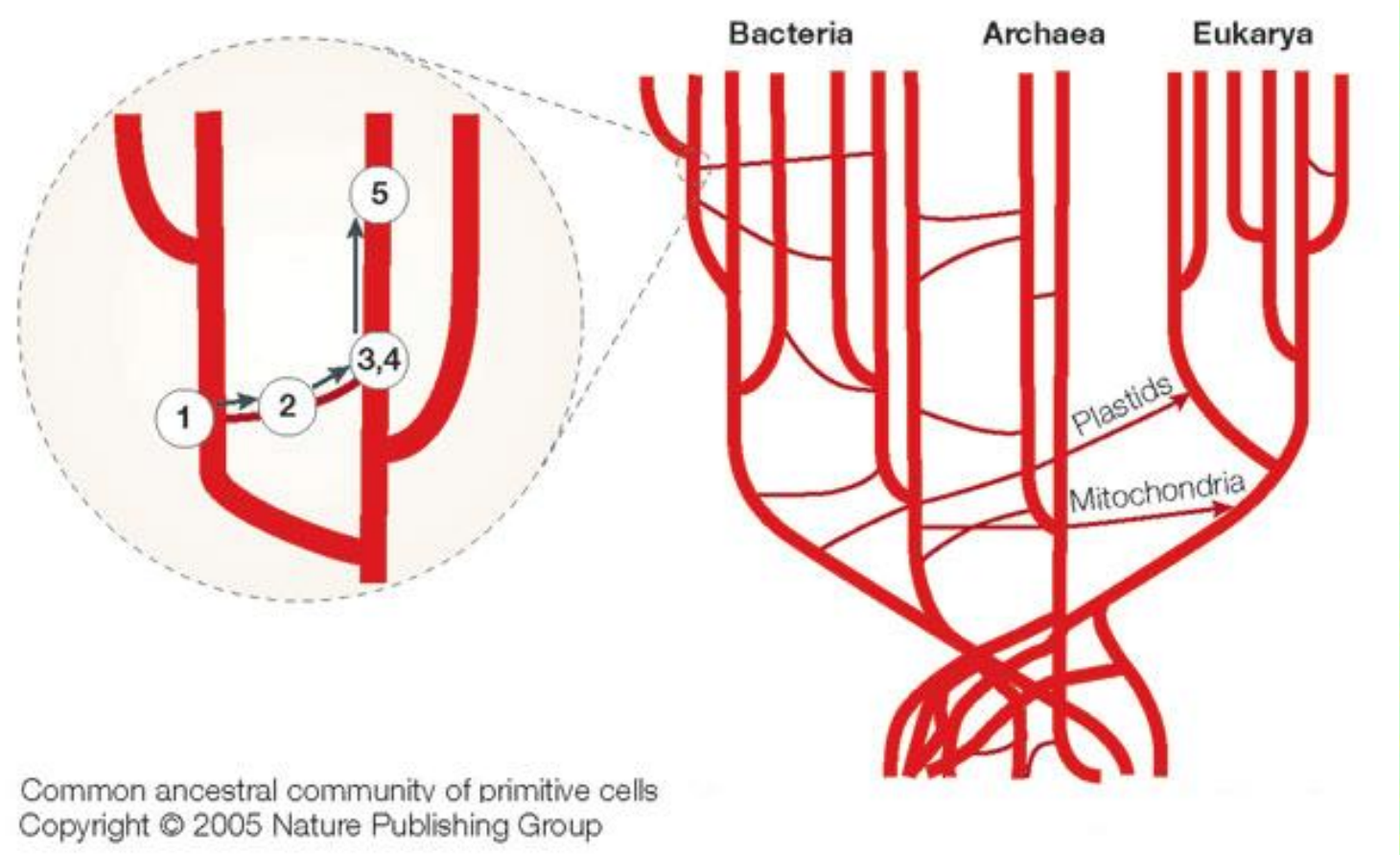




On trees and evolution

* Normal procreation of individuals is via a tree

* In case of horizontal gene transfer a **phylogenetic network** is more appropriate → Presentation of Steven Kelk



From phylogenetic data to a phylogenetic tree

1. Homology vs homoplasy, and orthologous vs paralogous

2. Sequence alignment (weights)

3. Multiple substitutions: corrections

4. (In)dependence and uniformity of substitutions

5. Phylogenetic analysis: tree, timing, reconstruction of ancestors

Character and Distance

A phylogenetic tree can be based on

1. based on **qualitative aspects** like **common characters**, or

2. **quantitative measures** like the **distance** or **similarity** between species or number of acquired mutations from last common ancestor (LCA).

Character based comparison

character 1

character 2

character 3

Non-numerical data:

has/has not

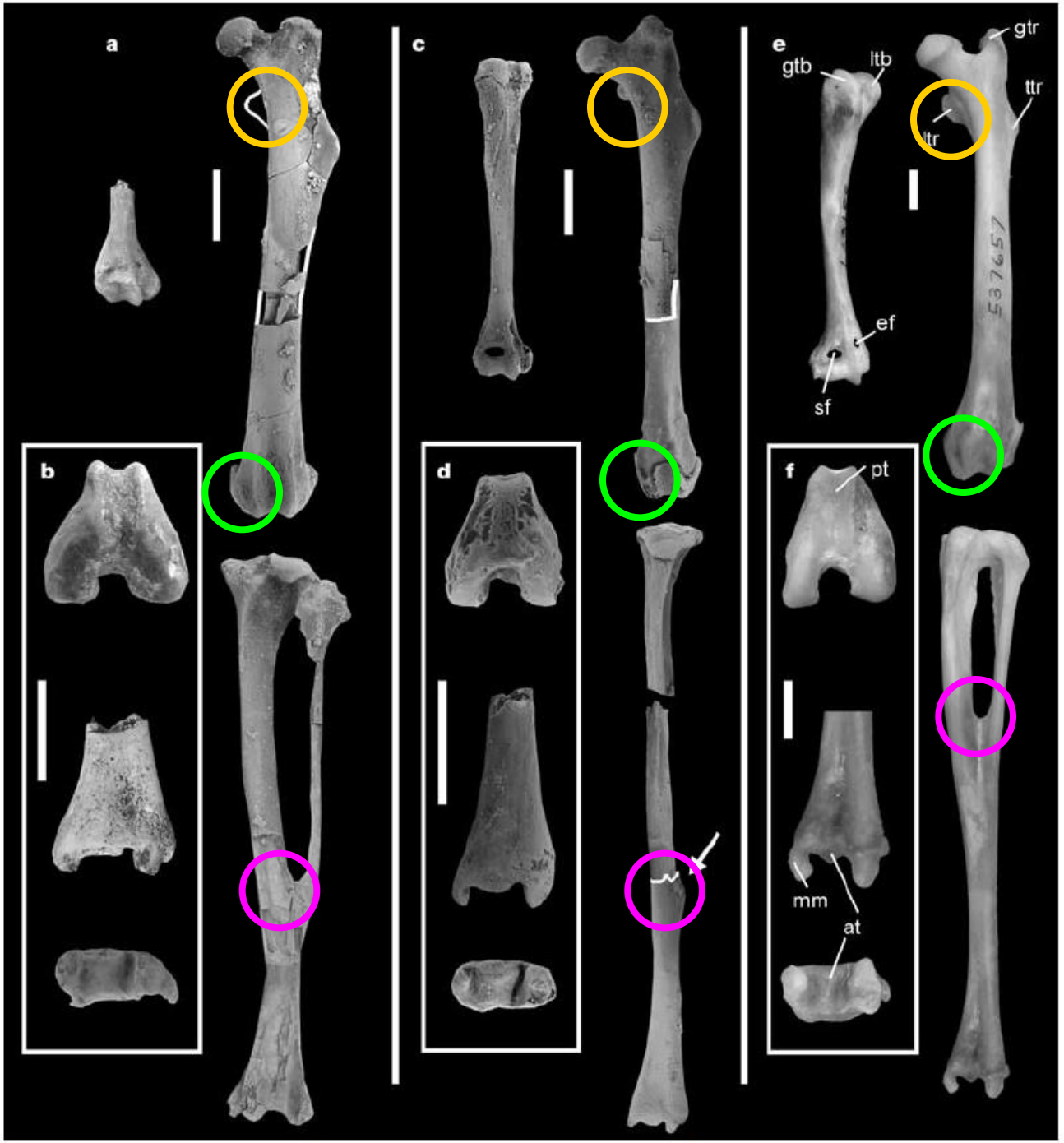


Figure 1 Comparison of apheliscine and macroscelidean long bones.

Constructing Phylogenetic Trees

There are three main methods of constructing phylogenetic trees:

* **character-based methods** such as maximum likelihood or Bayesian inference,

* **distance-based methods** such as UPGMA and neighbour-joining, and

* **parsimony-based methods** such as maximum parsimony.

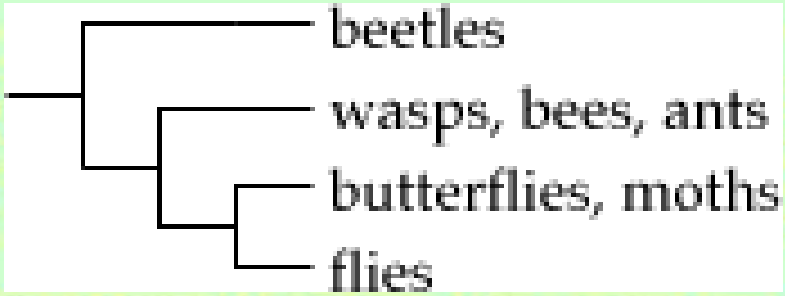
Parsimony is a 'less is better' concept of frugality, economy, stinginess or caution in arriving at a hypothesis or course of action. The word derives from Latin *parsimonia*, from *parcere*: **to spare**.

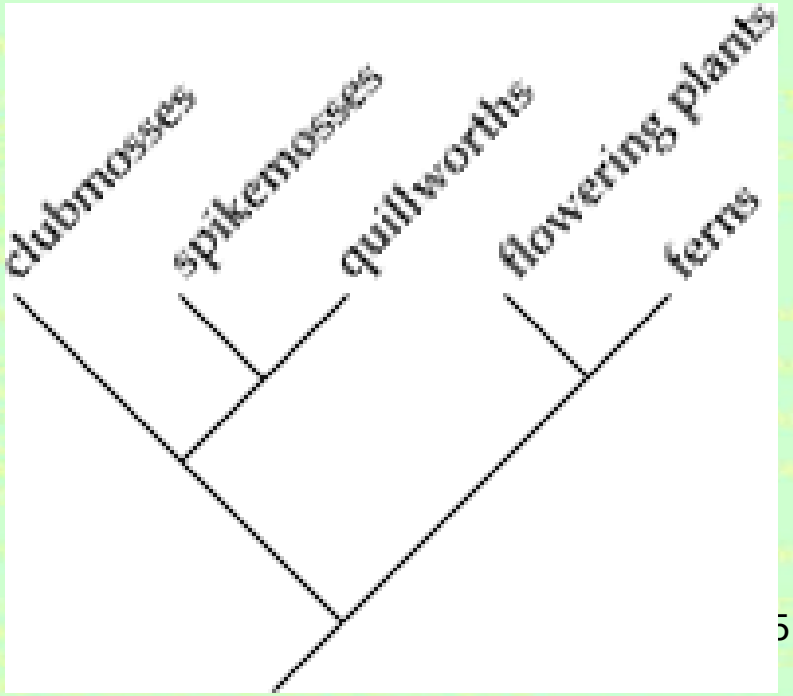
Cladistics

As treelike relationship-diagrams called "cladogram" is drawn up to show different hypotheses of relationships.

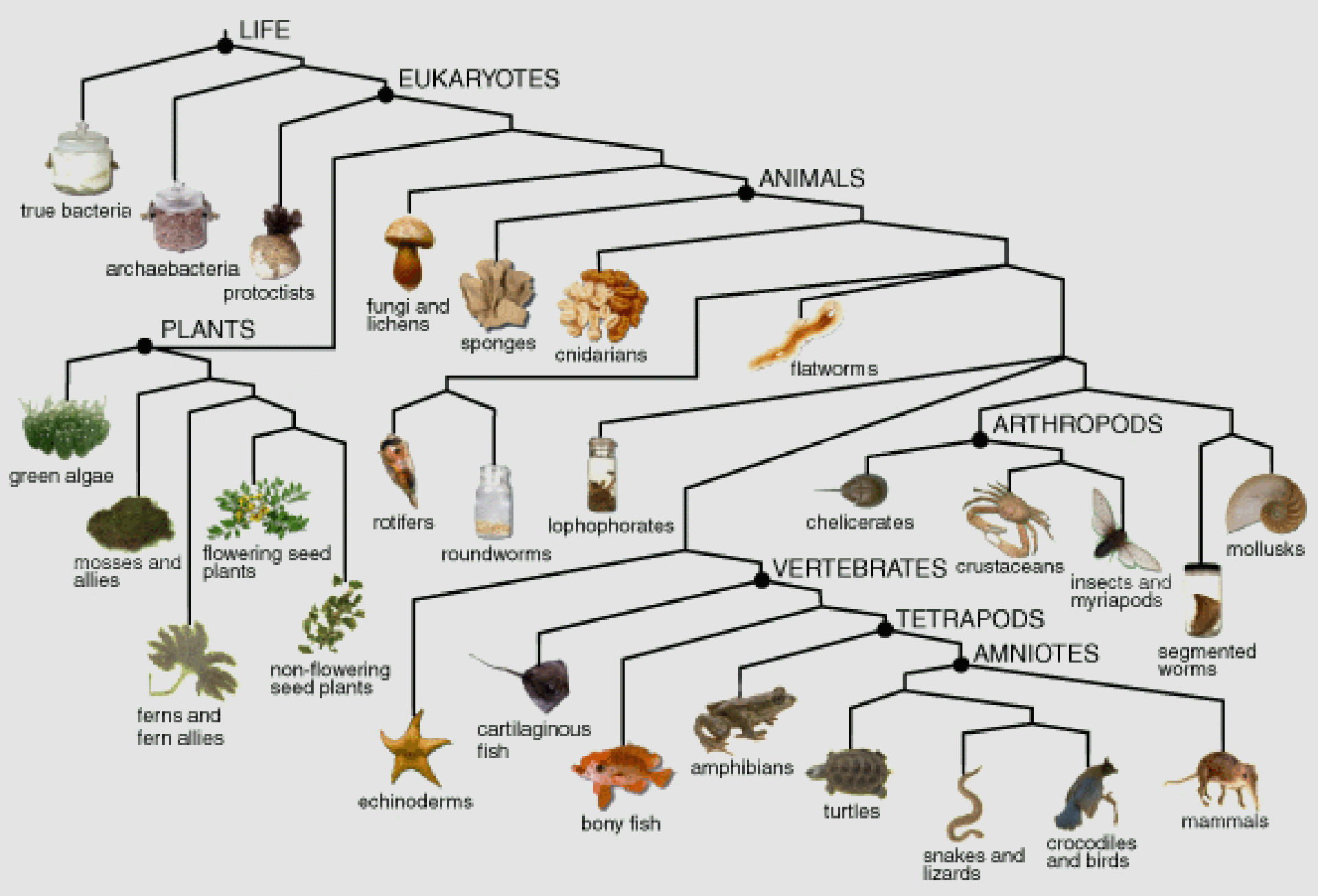
A cladistic analysis is typically based on morphological data.

This traditionally is *character based*





Cladistics : *tree of life*



Phylogenetic Trees

A phylogenetic tree is a tree showing the evolutionary interrelationships among various species or other entities that are believed to have a common ancestor. A phylogenetic tree is a form of a cladogram. In a phylogenetic tree, each node with descendants represents the most recent common ancestor of the descendants, and edge lengths correspond to time estimates.

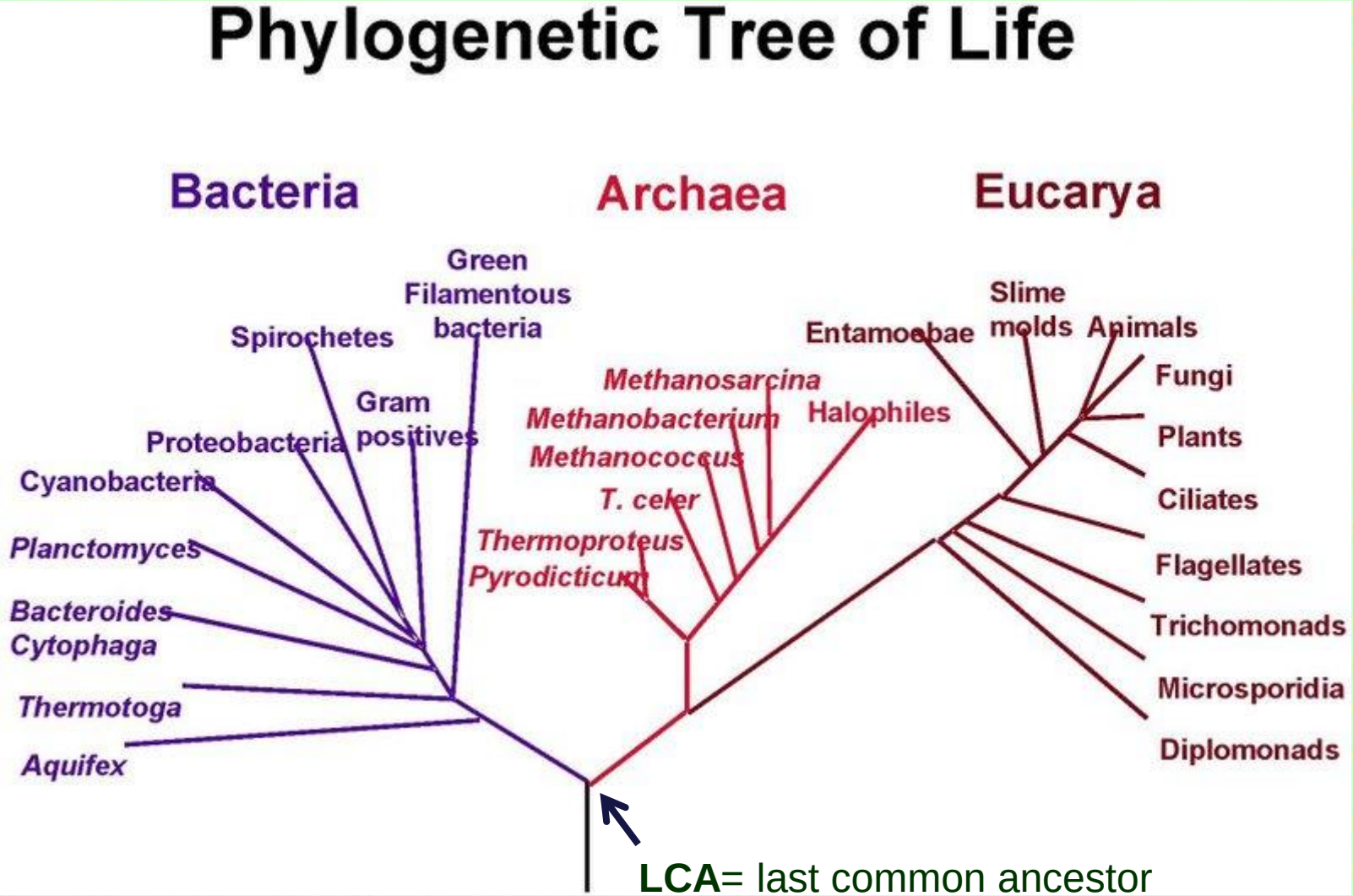
Each node in a phylogenetic tree is called a taxonomic unit. Internal nodes are generally referred to as Hypothetical Taxonomic Units (HTUs) as they cannot be directly observed.

Rooted and Unrooted Trees

A **rooted phylogenetic tree** is a directed tree with a unique node corresponding to the (usually imputed) **most recent common ancestor** of all the entities at the leaves of the tree.

PHYLOGENETIC TREES

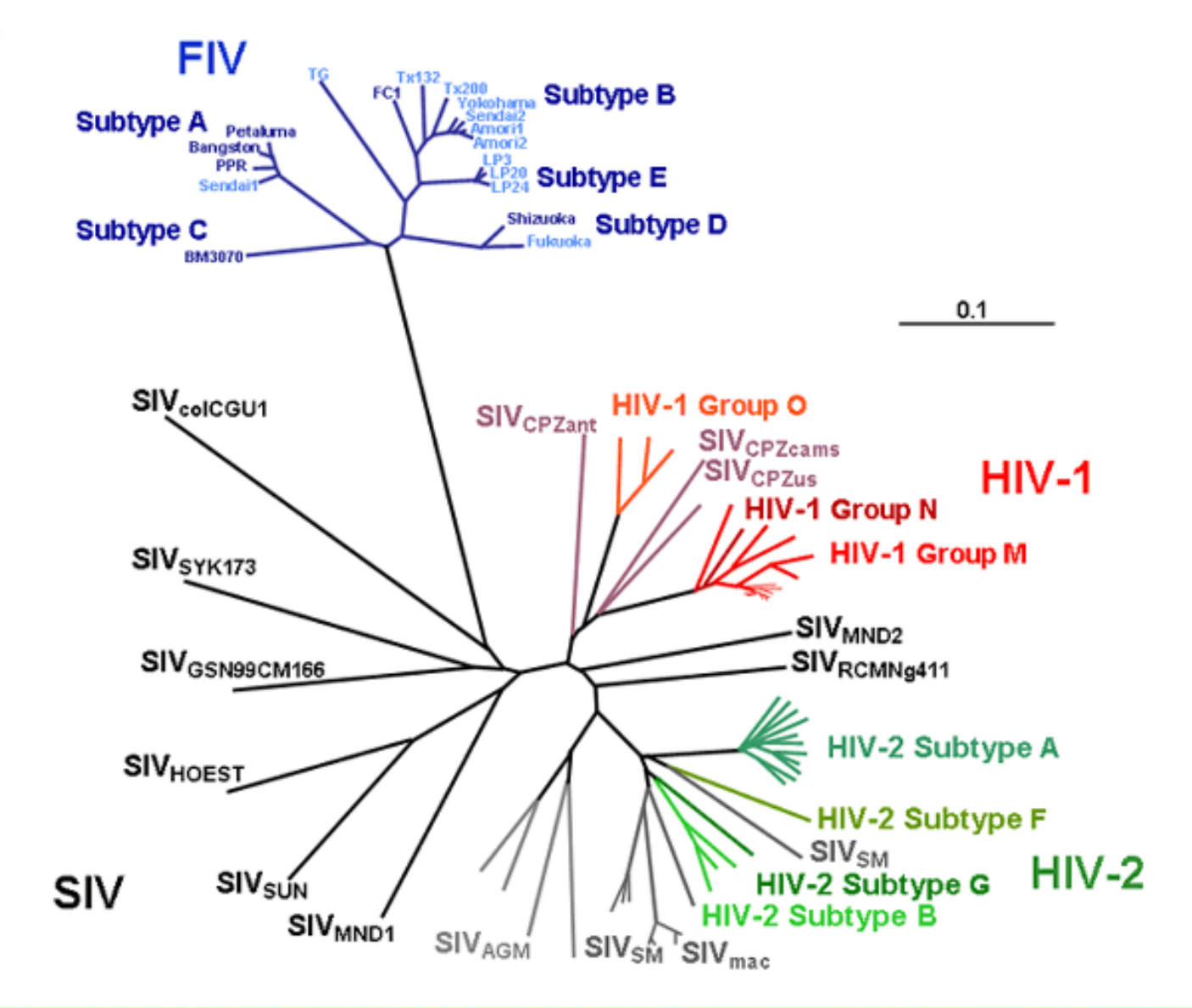
Rooted Phylogenetic Tree



Rooted and Unrooted Trees

Unrooted phylogenetic trees can be generated from rooted trees by omitting the root from a rooted tree, a root cannot be inferred on an unrooted tree without either an outgroup or additional assumptions.

Unrooted Phylogenetic Tree

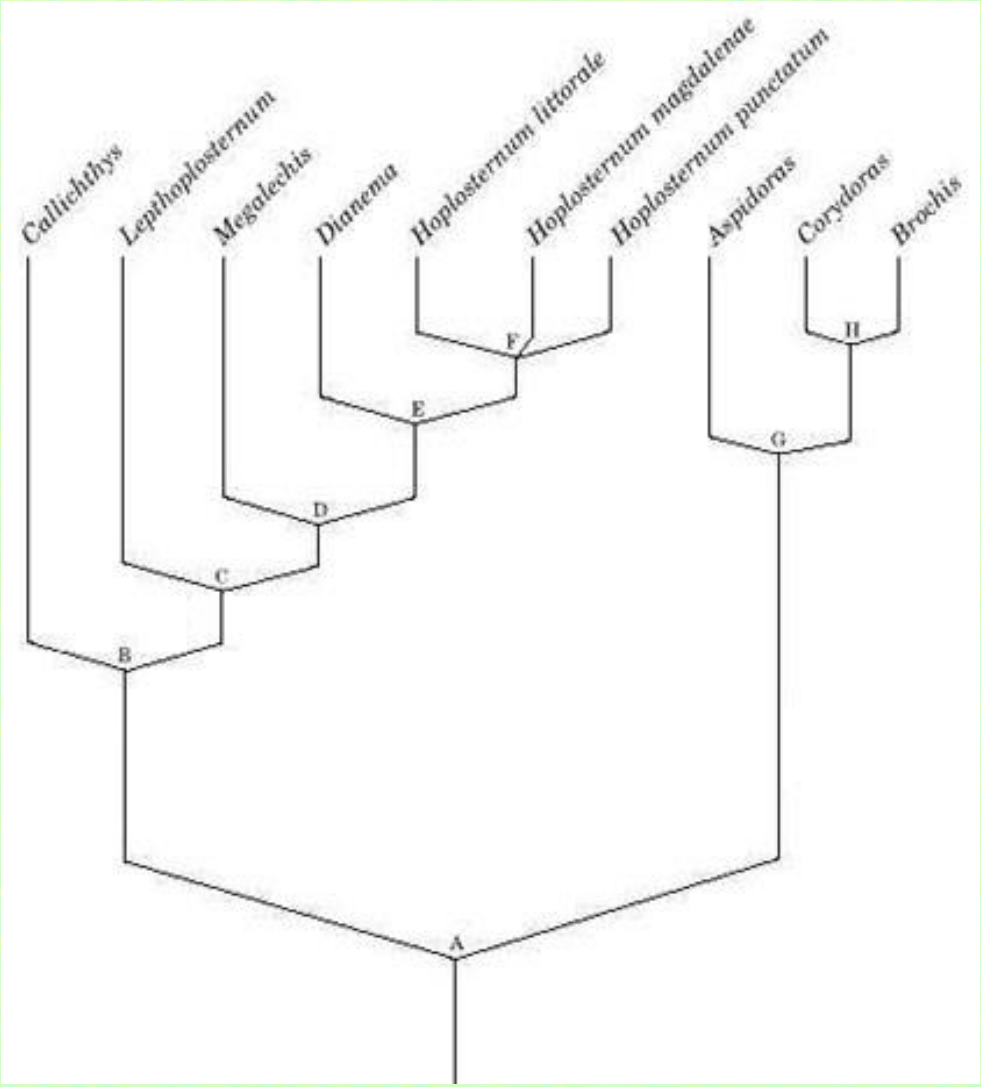


Trees and Branch Length

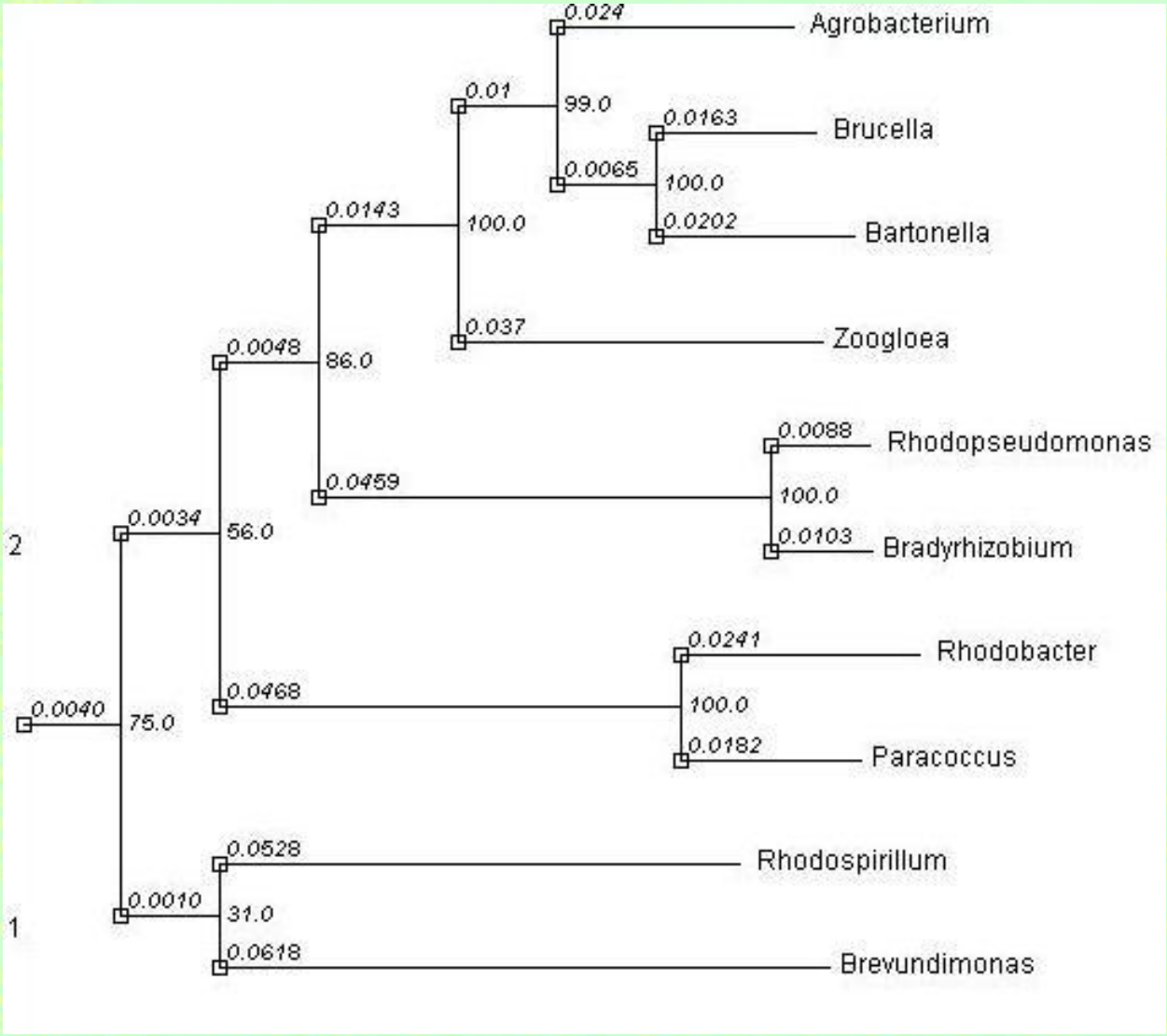
A tree can be a branching tree-graph where branches indicate close phylogenetic relations.

Alternatively, branches can have length that indicate the phylogenic closeness.

Tree without Branch Length



Tree with Branch Length



On trees and evolution

* Relation between “taxa”

* Internal nodes and external nodes (leafs)

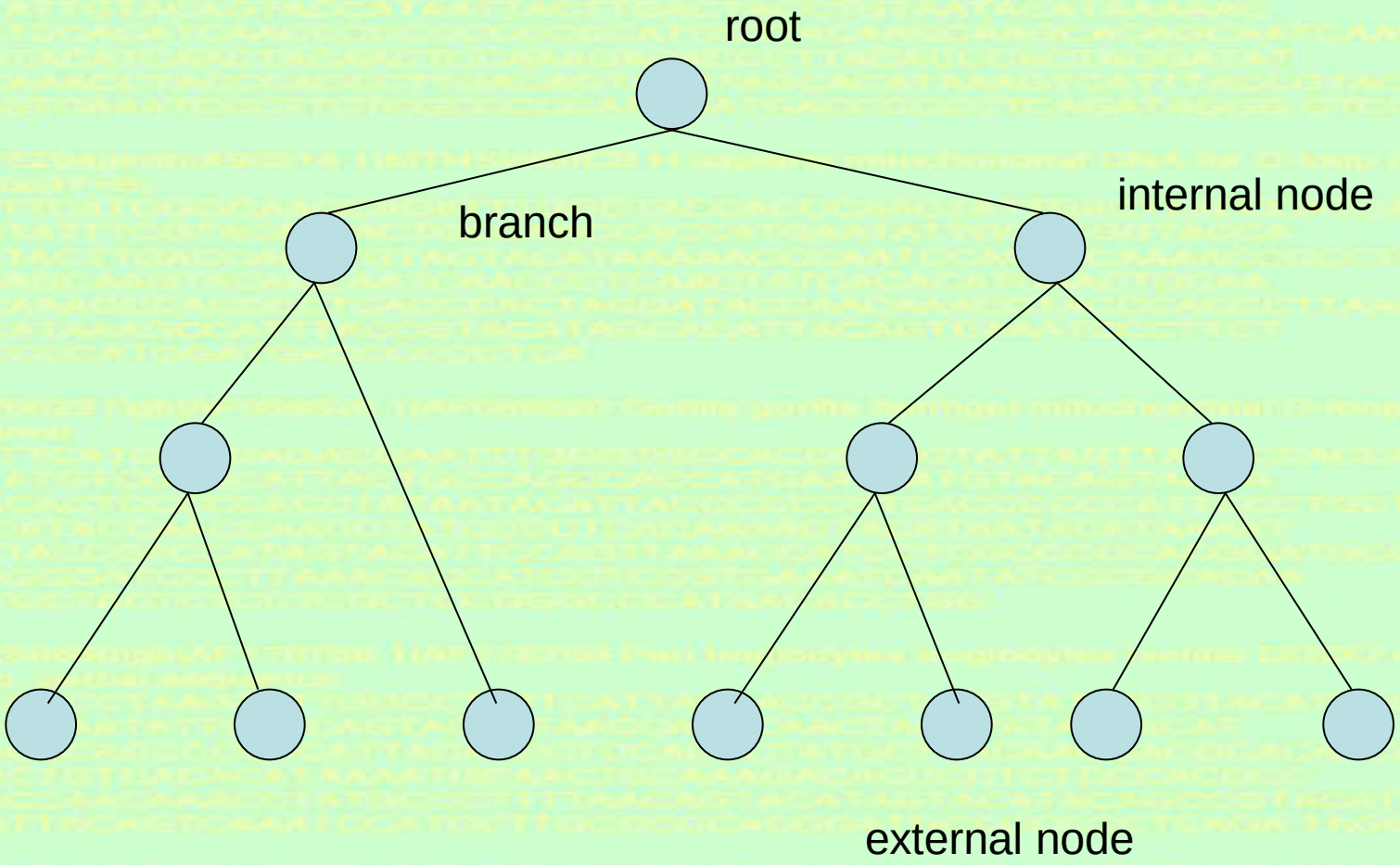
* Branches connects nodes

* Bifurcating tree: **internal** nodes have **degree**: **3**, **external** nodes degree: **1**, root **degree**: **2.**

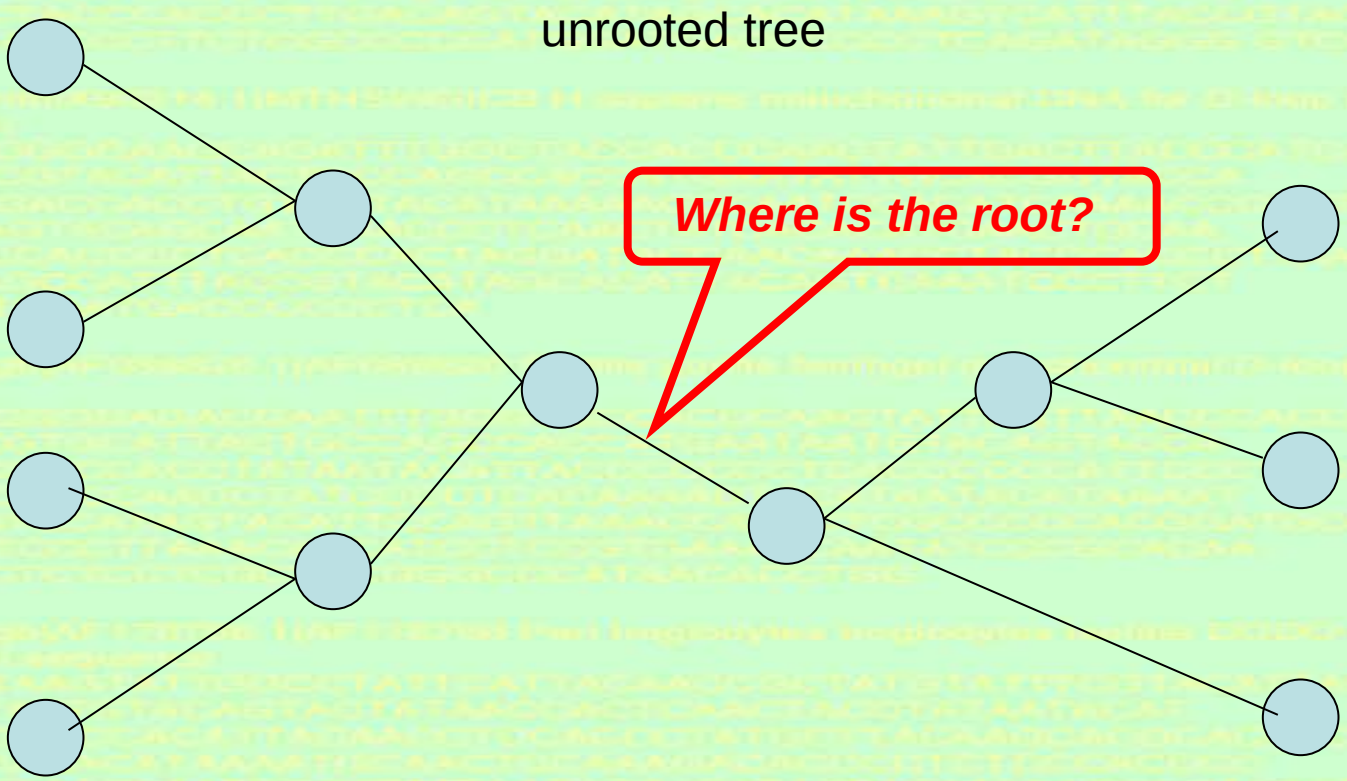
* Root connects to ‘outgroup’

* Multifurcating trees

ON TREES AND EVOLUTION



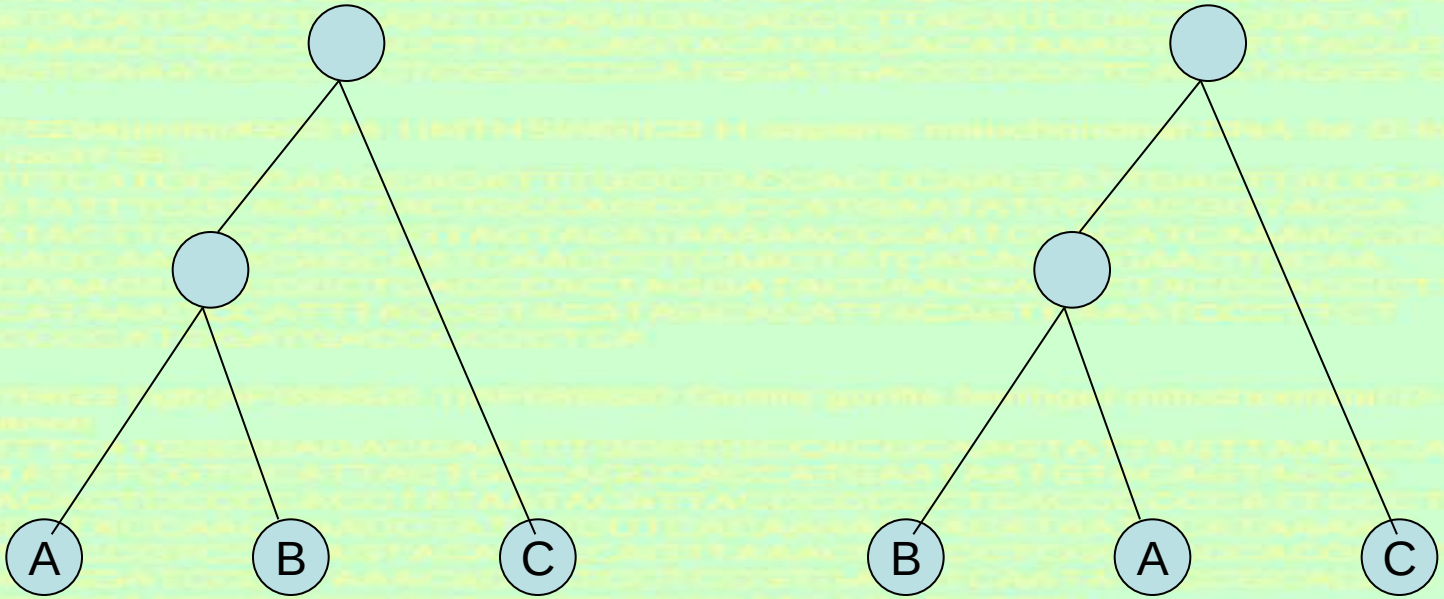
ON TREES AND EVOLUTION



ON TREES AND EVOLUTION

* Any rotation of the internal branches of a tree keeps the the phylogenetic relations intact

ON TREES AND EVOLUTION



rotation invariant

Number of possible trees

* n is number of taxa

* # unrooted trees for $n > 2$: $(2n - 5)!/(2^{n-3}(n-3)!)$

* # rooted trees for $n > 1$: $(2n - 3)!/(2^{n-2}(n-2)!)$

* $n = 5$: #rooted trees = 105

* $n = 10$: #rooted trees = 34,459,425

Representing trees

* Various possibilities

* Listing of nodes

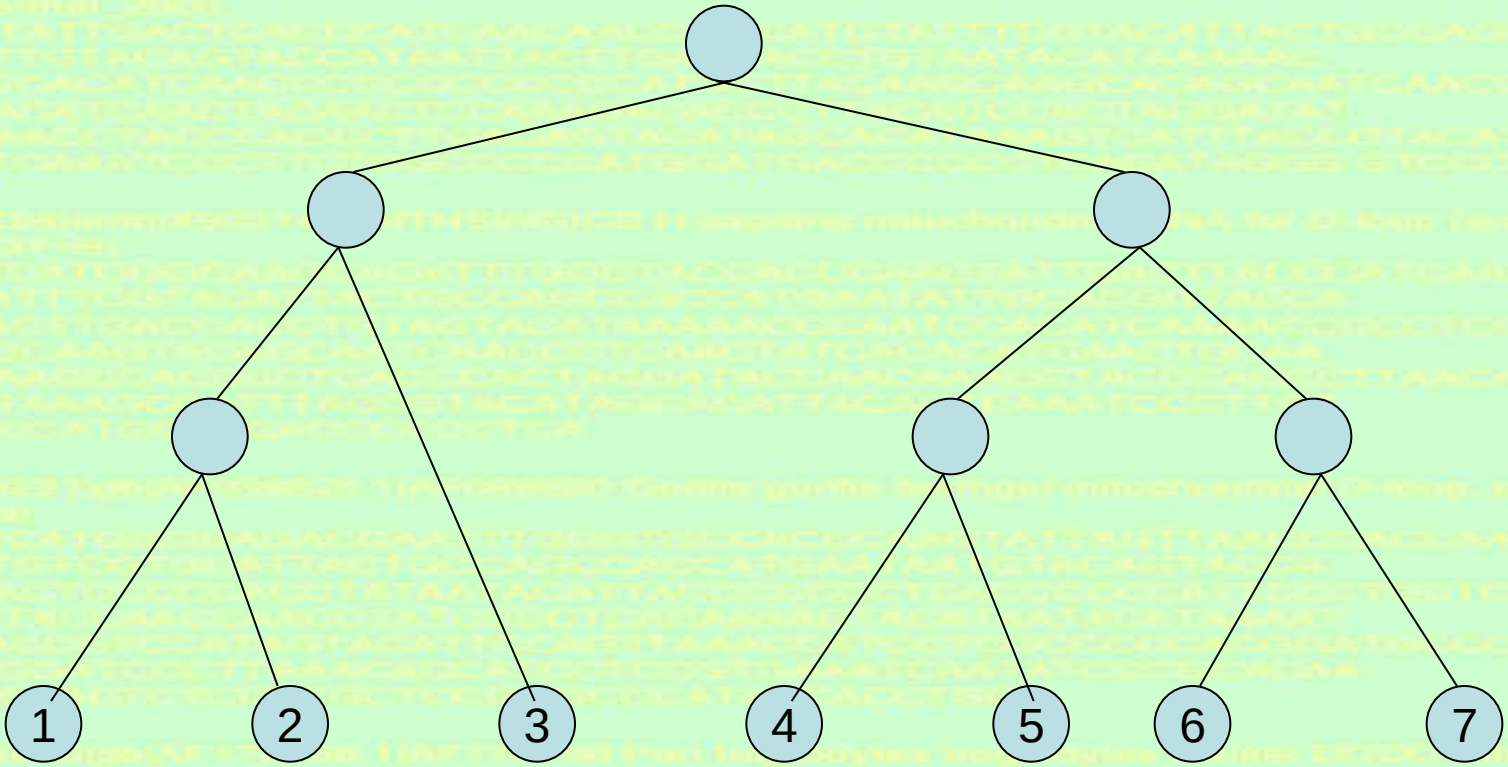
* *n* taxa = *n* external nodes: (*n* -1) internal nodes

* internal nodes with childeren: (*n* – 1)x3 matrix

* (internal node, daughter_1, daughter_2)

* Newick format: see next slide for example

ON TREES AND EVOLUTION



Newick format: (((1,2),3),((4,5),(6,7)))

PARSIMONY

Under **parsimony**, the preferred phylogenetic tree is the tree that requires the **least evolutionary change** to explain some observed data.

Given a family of trees $T(\theta)$ with minimum substitutions $n(i,j|\theta)$ between branches i and j:

$$\theta^* = min\ \Sigma\ n(i,j|\theta)$$

The obtained result is the **maximum parsimonous tree**

PARSIMONY

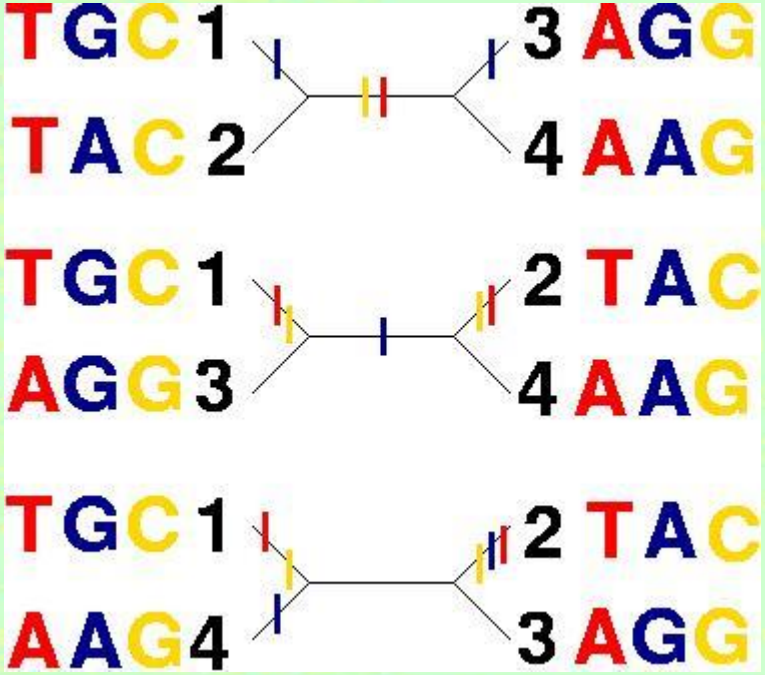
The aim of maximum parsimony is to find the shortest tree, that is the tree with the smallest number of changes that explains the observed data.

Example:

Position	1	2	3
Sequence1	T	G	C
Sequence2	T	A	C
Sequence3	A	G	G
Sequence4	A	A	G

1. draw all the possible trees
2. consider each position separately
3. find tree with fewest changes to explain data

(1,2): 4
(1,3): 5
(1,4): 6
So: shortest tree : ((1,2)(3,4))



PARSIMONY

* Real evolution may have more substitutions!
* So maximum parsimonous tree is a lower bound on the evolution

Inferring distance based trees

* input: distance table

QUESTION: *which* distances ?!

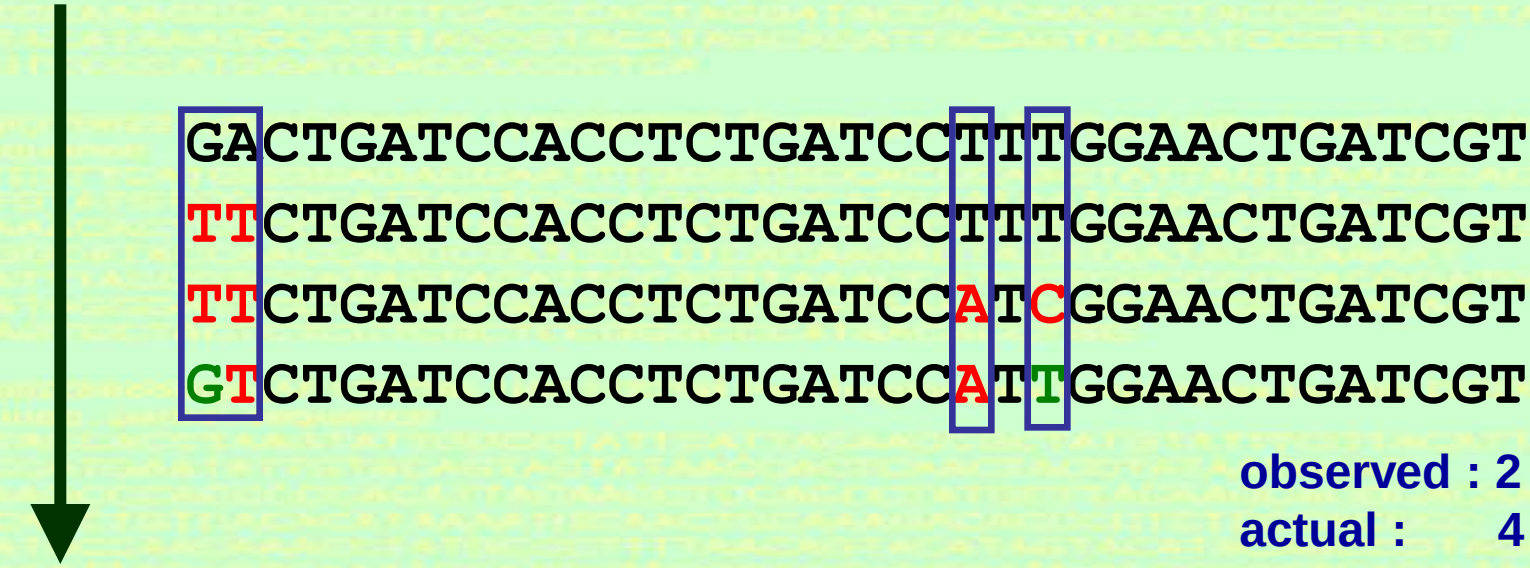
Estimating genetic distance

* Substitutions are independent (?)
* Substitutions are random
* Multiple substitutions may occur
* Back-mutations mutate a nucleotide back to an earlier value

PHYLOGENETIC ANALYSIS

Multiple substitutions and Back-mutations

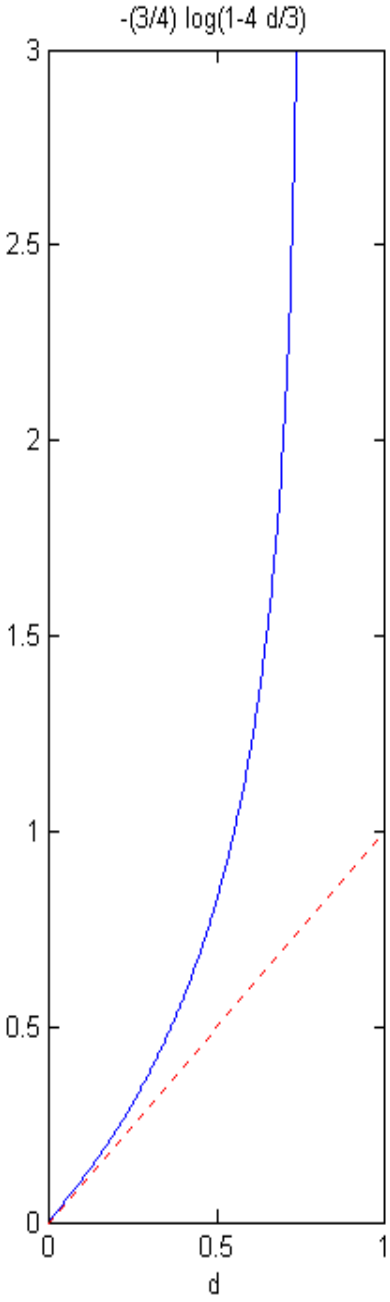
conceal the real genetic distance



The **actual** genetic distance *K* for an **observed** gene-gene dissimilarity *d* is the **Jukes-Cantor formula :**

$$K \approx -\frac{3}{4}\ln\left(1-\frac{4}{3}d\right)$$

Jukes-Cantor



Inferring trees

* n taxa $\{t_1,\ldots,t_n\}$

* D matrix of pairwise genetic distances + JC-correction

* **Additive** distances: distance over path from $i \rightarrow j$ is: $d(i,j)$

* (total) length of a tree: sum of all branch lengths.

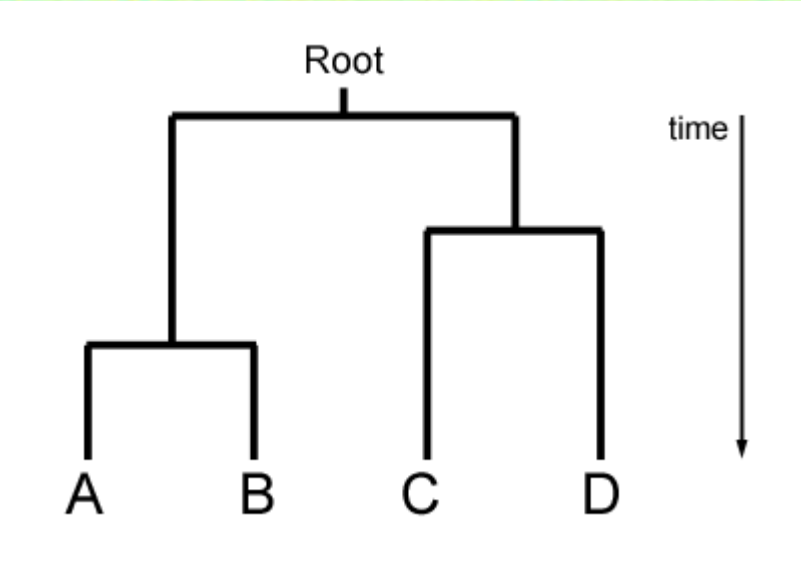
Ultrametric trees:

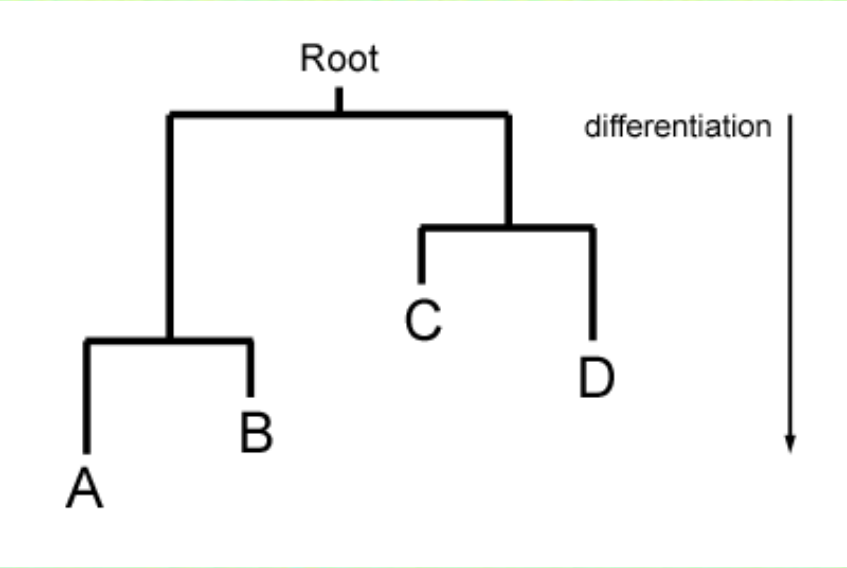
If the distance from the root to all leafs is equal the tree is **ultrametric**

Ultrametricity must be valid for the real tee, but due to noise this condition will in practice generate erroneous trees.

INFERRING TREES

Ultrametric - Minimum length





MINIMUM LENGTH TREE

Find phylogenetic tree with minimum total length of the branches

Given a family of trees $T(\theta)$ with branch length $\lambda(i,j|\theta)$ between nodes i and j – and genetic distance $d(i,j)$

$$L^* = min\ \Sigma\ \lambda(i,j|\theta) \text{ subject to } \lambda(i,j|\theta) \geq d(i,j|\theta) \geq 0$$

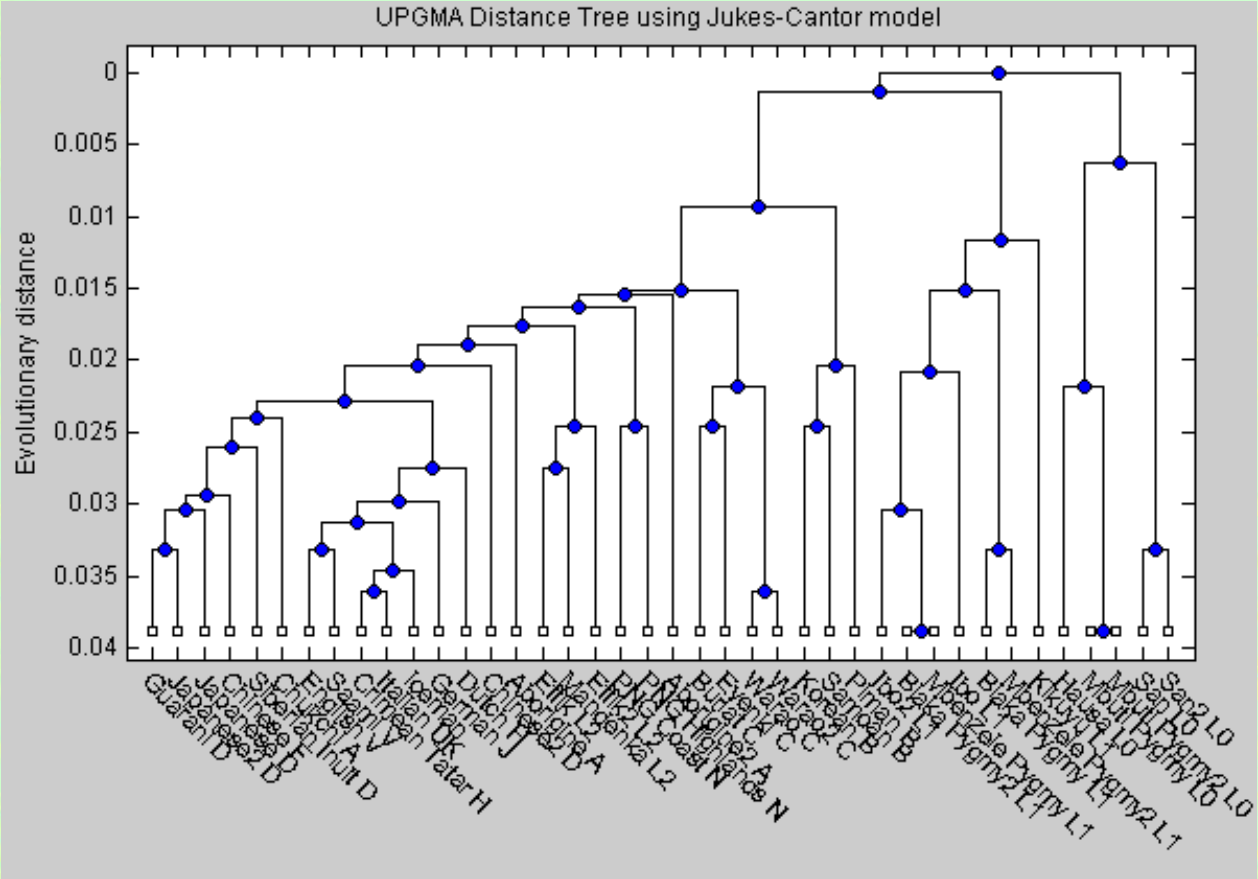
The obtained result is the **minimum length tree**

This looks much like the maximum parsimonous tree

NEIGHBOR JOINING algorithm:

Popular distance-based clustering method

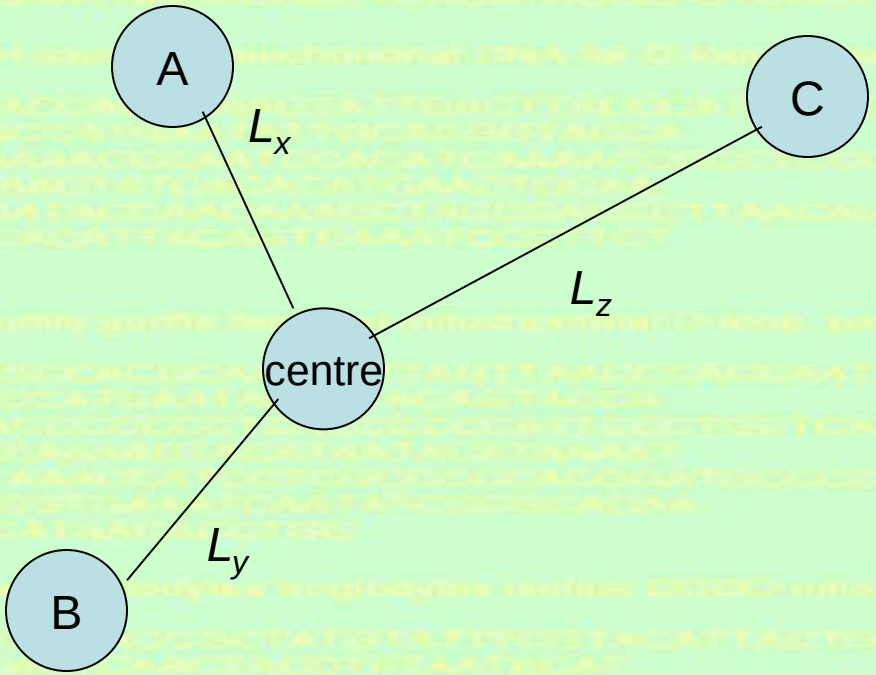
Iteratively combine closest nodes



Finding Branche lengths:

Three-point formula:

$L_x + L_y = d_{AB}$
$L_x + L_z = d_{AC}$
$L_y + L_z = d_{BC}$



$L_x = (d_{AB}+d_{AC}-d_{BC})/2$
$L_y = (d_{AB}+d_{BC}-d_{AC})/2$
$L_z = (d_{AC}+d_{BC}-d_{AB})/2$

INFERRING TREES

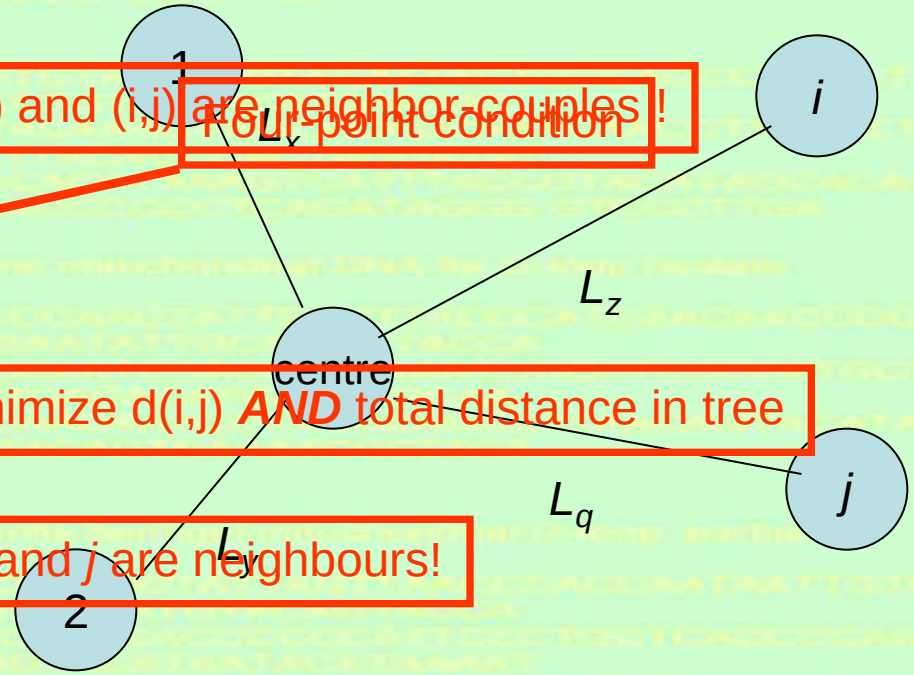
Four-point formula:

$d(1,2) + d(i,j) < d(i,1) + d(2,j)$

$R_i = \sum_j d(t_i, t_j)$

$M(i,j) = (n\text{-}2)d(i,j) - R_i - R_j$

$M(i,j) < M(i,k)$ for all k not equal to j



NEIGHBOR JOINING algorithm:

Input: *nxn* distance matrix *D* and an outgroup
Output: rooted phylogenetic tree *T*

Step 1: Compute new table *M* using *D* – select smallest value of *M* to select two taxa to join

Step 2: Join the two taxa t_i and t_j to a new vertex *V* - use 3-point formula to calculate the updates distance matrix *D'* where t_i and t_j are replaced by *V* .

Step 3: Compute branch lengths from t_k to *V* using 3-point formula, $T(V,1) = t_i$ and $T(V,2) = t_j$ and $TD(t_i) = L(t_i,V)$ and $TD(t_i) = L(t_i,V)$.

Step 4: The distance matrix *D*' now contains *n – 1* taxa. If there are more than 2 taxa left go to step 1. If two taxa are left join them by an branch of length $d(t_i,t_j)$.

Step 5: Define the root node as the branch connecting the outgroup to the rest of the tree. (Alternatively, determine the so-called "**mid-point**")

UPGMA and ultrametric trees:

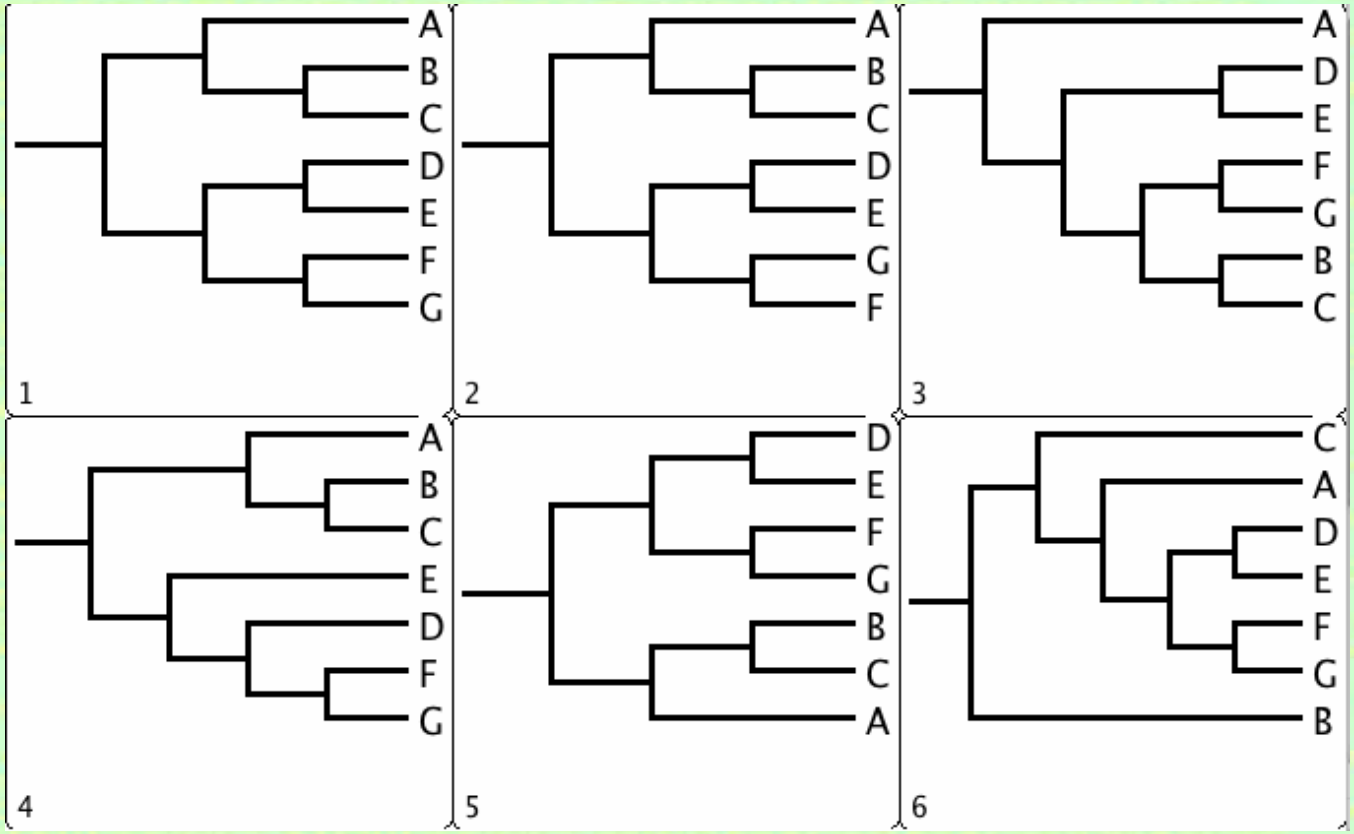
For **ultrametric** trees use *D* instead of *M* and the algorithm is called UPGMA (**Unweighted Pair Group Method**)
.

EVALUATING TREES

- **(un)decidability**
- **Hypothesis testing: models of evolution**
- **Using numerical simulation**

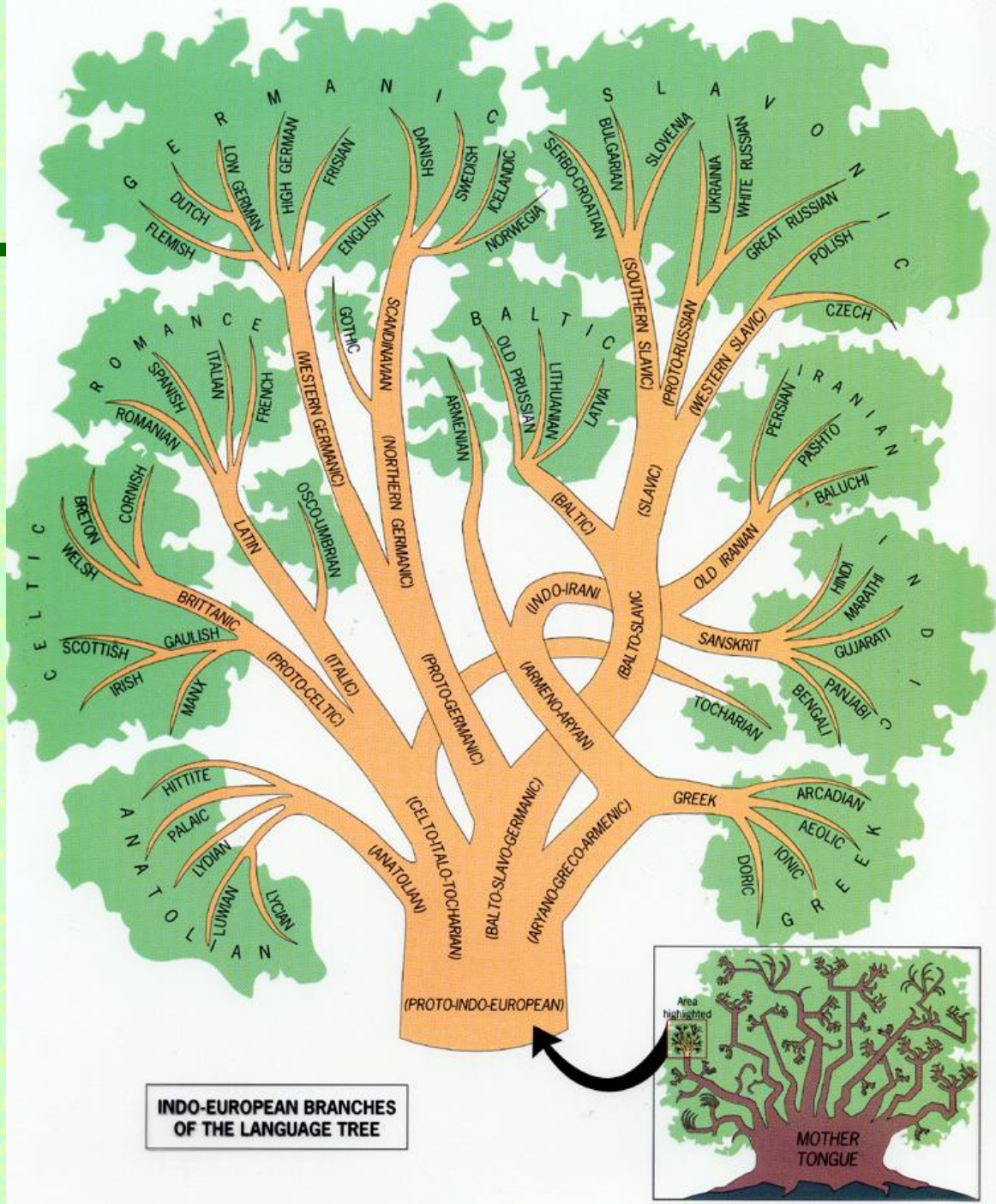
CONSENSUS TREES

Different genes/proteins can/will give different trees



OTHER APPLICATIONS

Language families



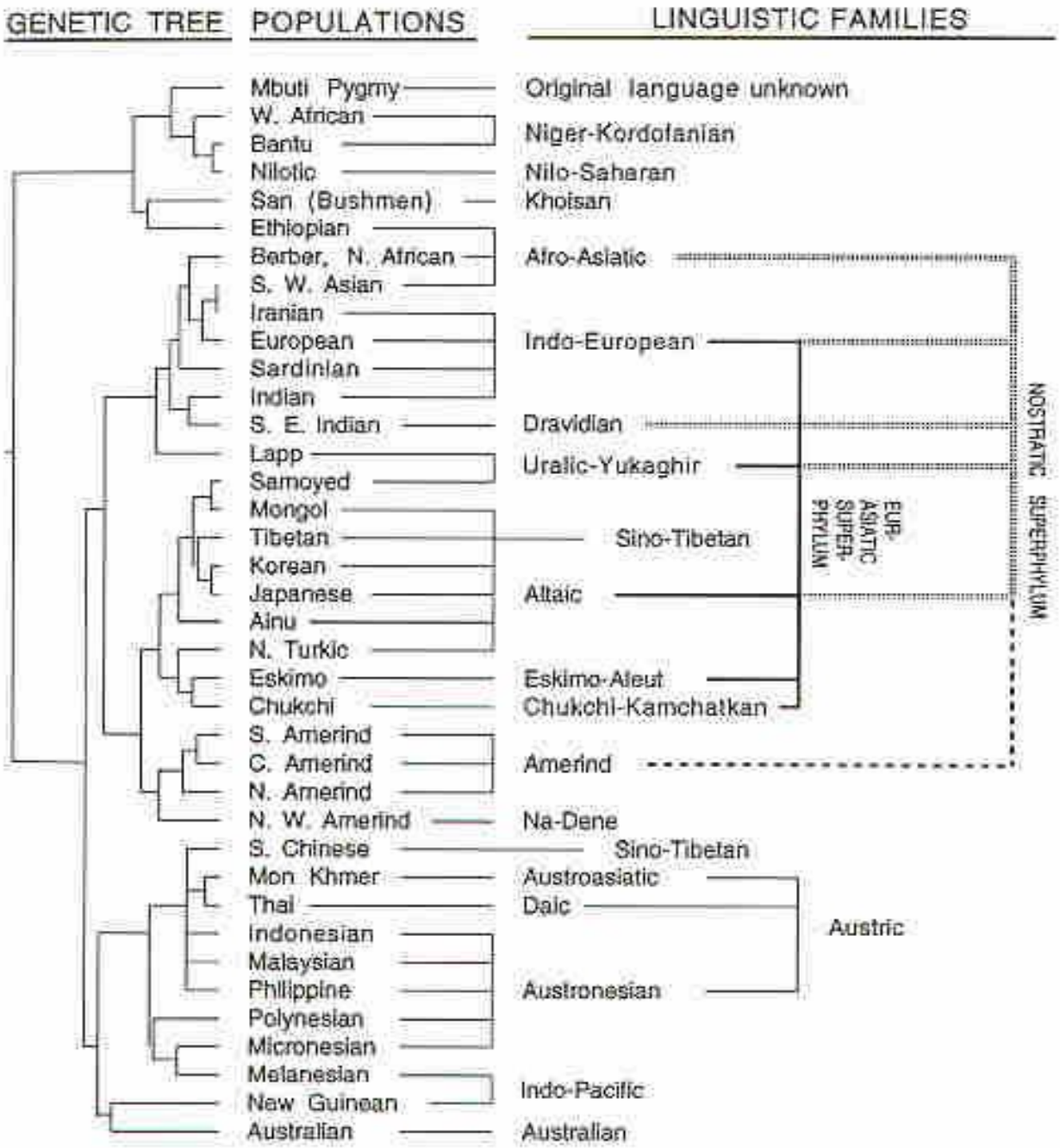


Fig. 2.6.2 The genetic tree comparing linguistic families and superfamilies published in Cavalli-Sforza et al. (1988). Populations pooled on the basis of linguistic classification belong to the following groups: Bantu, Niger-Kordofanian family; Nilotic, Nilo-Saharan family; Southeast Indian, Dravidian family; Samoyeds, Uralic family from Russia; North Turkic, branch of Altaic family; Northwest Amerind, Na-Dene family. The genetic tree was constructed by average linkage analysis of Nei's genetic distances and is the same as that of figure 2.3.2A.

Stone, Linda; Lurquin, Paul F.; ***Cavalli-Sforza, L. Luca Genes, Culture, and Human Evolution: A Synthesis****. Malden (MA): Wiley-Blackwell (2007).*

CASE STUDY:

Phylogenetic Analysis of the 2003 SARS epidemic

SARS: the outbreak

* February 28, 2003, Hanoi, the Vietnam French hospital called the WHO with a report of an influenza-like infection.

* Dr. Carlo Urbani (WHO) came and concluded that this was a new and unusual pathogen.

* Next few days Dr. Urbani collected samples, worked through the hospital documenting findings, and organized patient quarantine.

* Fever, dry cough, short breath, progressively worsening respiratory failure, death through respiratory failure.

SARS: the outbreak

* Dr. Carlo Urbani was the first to identify *Severe Acute Respiratory Syndrome: SARS*.

* In three weeks Dr. Urbani and five other healthcare professionals from the hospital died from the effects of *SARS*.

* By March 15, 2003, the WHO issued a global alert, calling *SARS* a worldwide health threat.

PHYLOGENETIC TREES

Dr. Carlo Urbani (1956-2003)
WHO

Hanoi, the Vietnam French hospital, March 2003

Origin of the SARS epidemic

* Earliest cases of what now is called *SARS* occurred in November 2002 in Guangong (P.R. of China)

* Guangzhou hospital spread 106 new cases

* A doctor from this hospital visited Hong Kong, on Feb 21, 2003, and stayed in the 9th floor of the Metropole Hotel

* The doctor became ill and died, diagnozed pneumonia

* Many of the visitors of the 9th floor of the Metropole Hotel now became disease carriers themselves

Origin of the SARS epidemic

* One of the visitors of the 9th floor of the Metropole Hotel was an American business man who went to Hanoi, and was the first patient to bring *SARS* to the Vietnam French hospital of Hanoi.

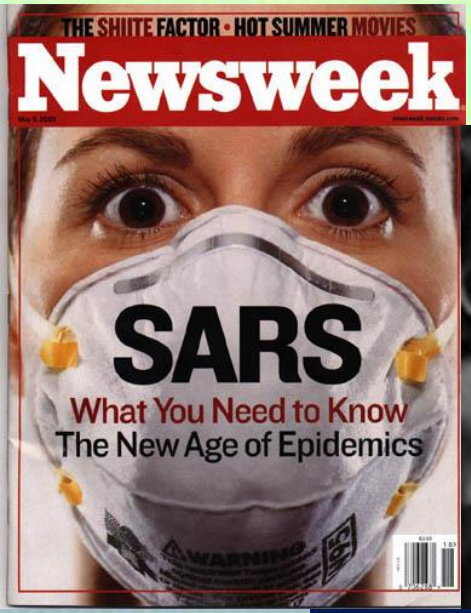
* He infected 80 people before dying

* Other visitors of the 9th floor of the Metropole Hotel brought the diesease to Canada, Singapore and the USA.

* By end April 2003, the disease was reported in 25 countries over the world, on 4300 cases and 250 deaths.

PHYLOGENETIC TREES

SARS panic & Mediahype, April-June 2003



The SARS corona virus

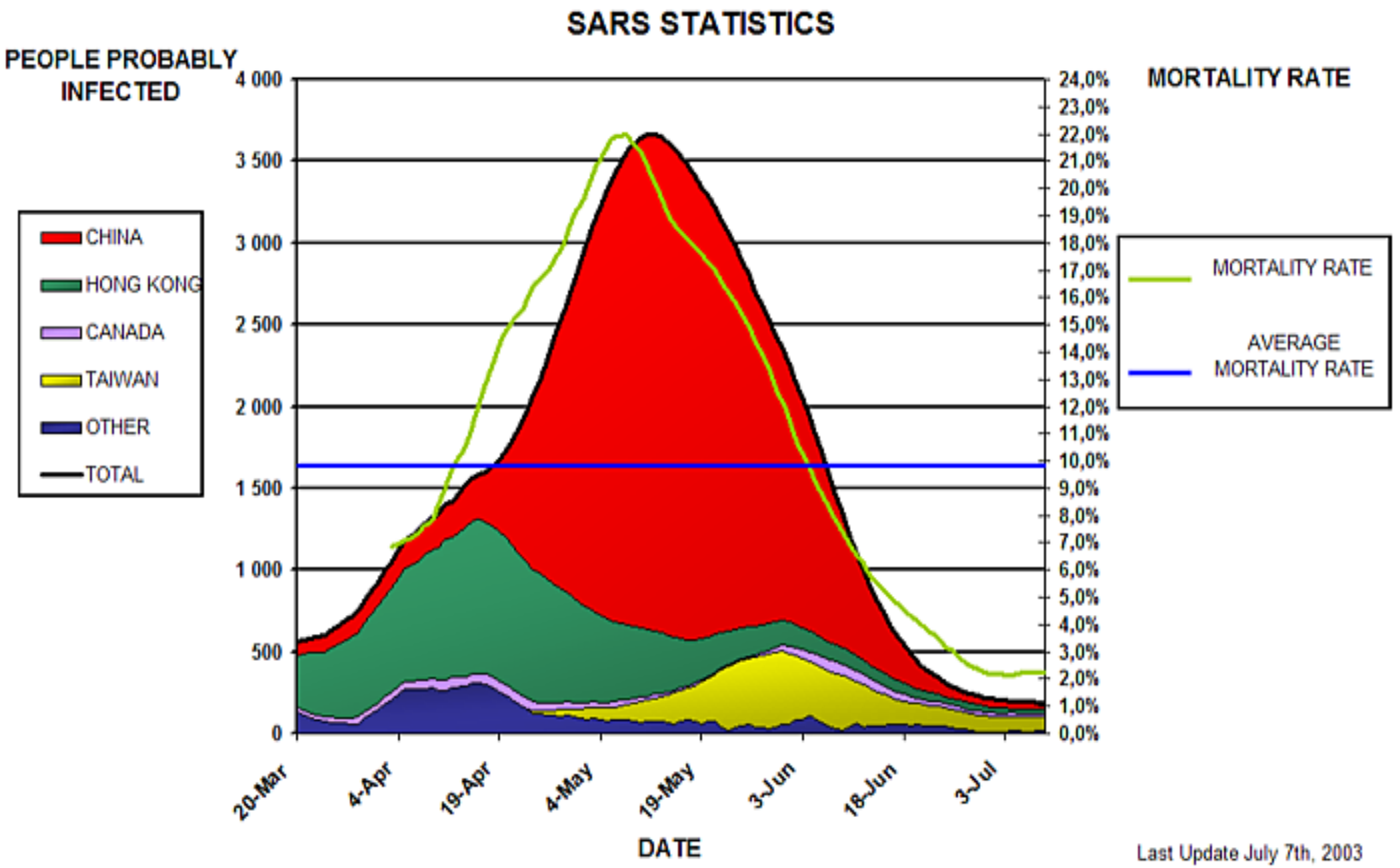
* Early March 2003, the WHO coordinated an international research .

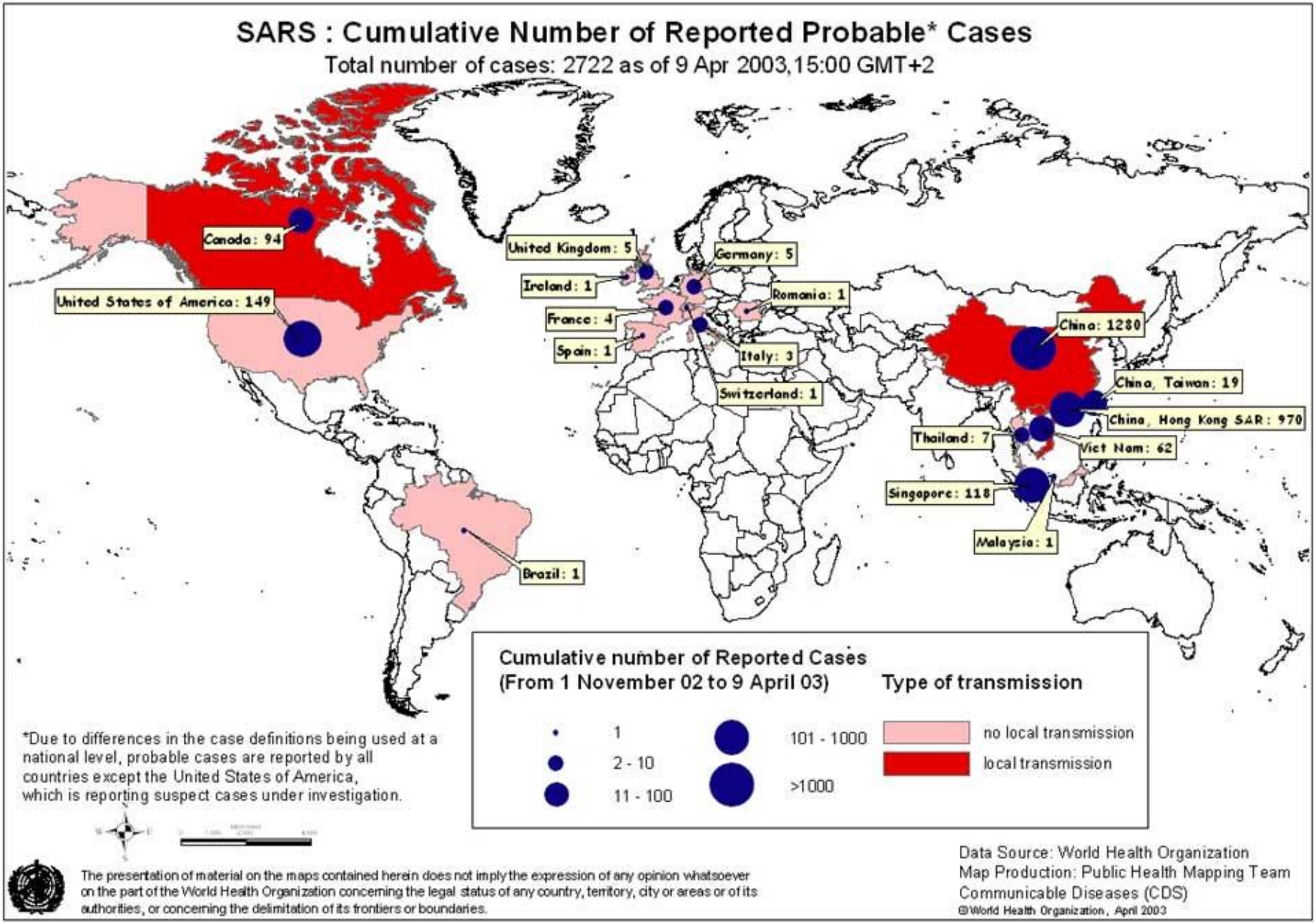
* End March 2003, laboratories in Germany, Canada, United Staes, and Hong Kong independently identified a novel virus that caused *SARS*.

* The *SARS* corona virus (SARS-CoV) is an RNA virus (like HIV).

* Corona viruses are common in humans and animals, causing ~25% of all upper respiratory tract infections (e.g. common cold) .

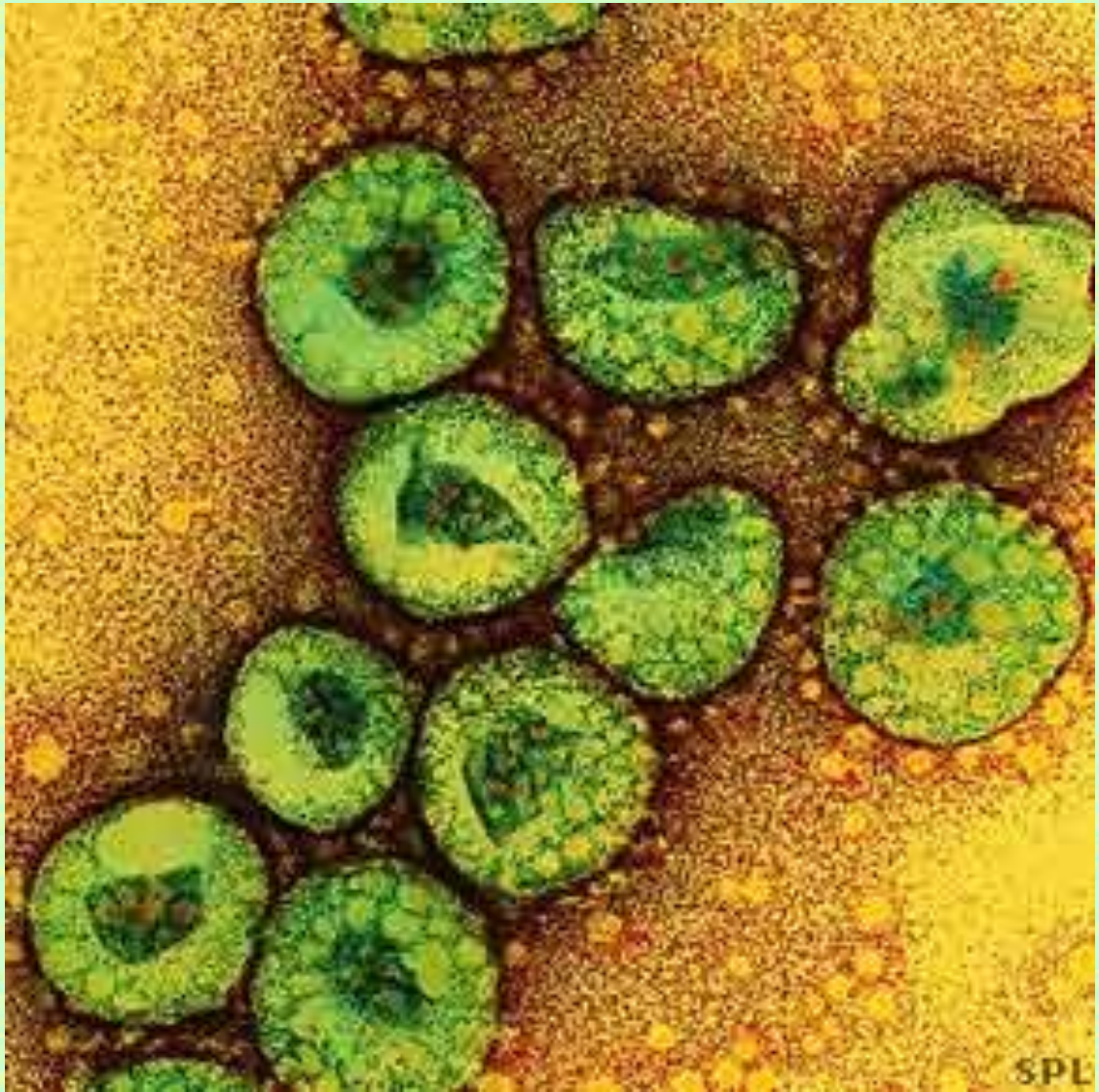
SARS: the outbreak



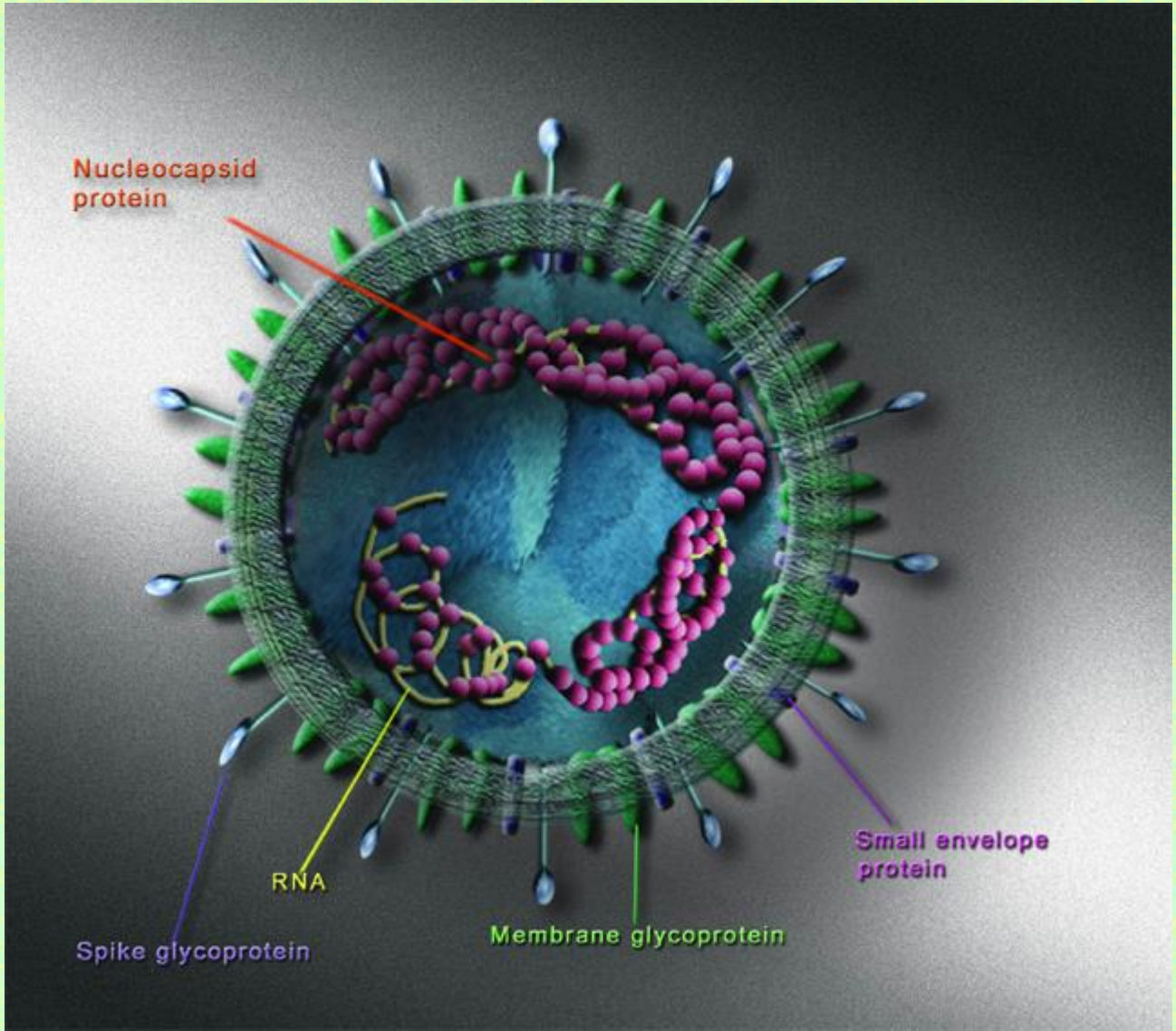

SARS : Cumulative Number of Reported Probable* Cases
Total number of cases: 2722 as of 9 Apr 2003,15:00 GMT+2
Canada: 94
United States of America: 149
Brazil: 1
United Kingdom: 5
Ireland: 1
France: 4
Spain: 1
Germany: 5
Romania: 1
Italy: 3
Switzerland: 1
China: 1280
China, Taiwan: 19
China, Hong Kong SAR: 970
Thailand: 7
Viet Nam: 62
Singapore: 118
Malaysia: 1
Cumulative number of Reported Cases
(From 1 November 02 to 9 April 03)
1
2 - 10
11 - 100
101 - 1000
>1000
Type of transmission
no local transmission
local transmission
*Due to differences in the case definitions being used at a national level, probable cases are reported by all countries except the United States of America, which is reporting suspect cases under investigation.
The presentation of material on the maps contained herein does not imply the expression of any opinion whatsoever on the part of the World Health Organization concerning the legal status of any country, territory, city or areas or of its authorities, or concerning the delimitation of its frontiers or boundaries.
Data Source: World Health Organization
Map Production: Public Health Mapping Team
Communicable Diseases (CDS)
©World Health Organization, April 2003

SARS: the outbreak

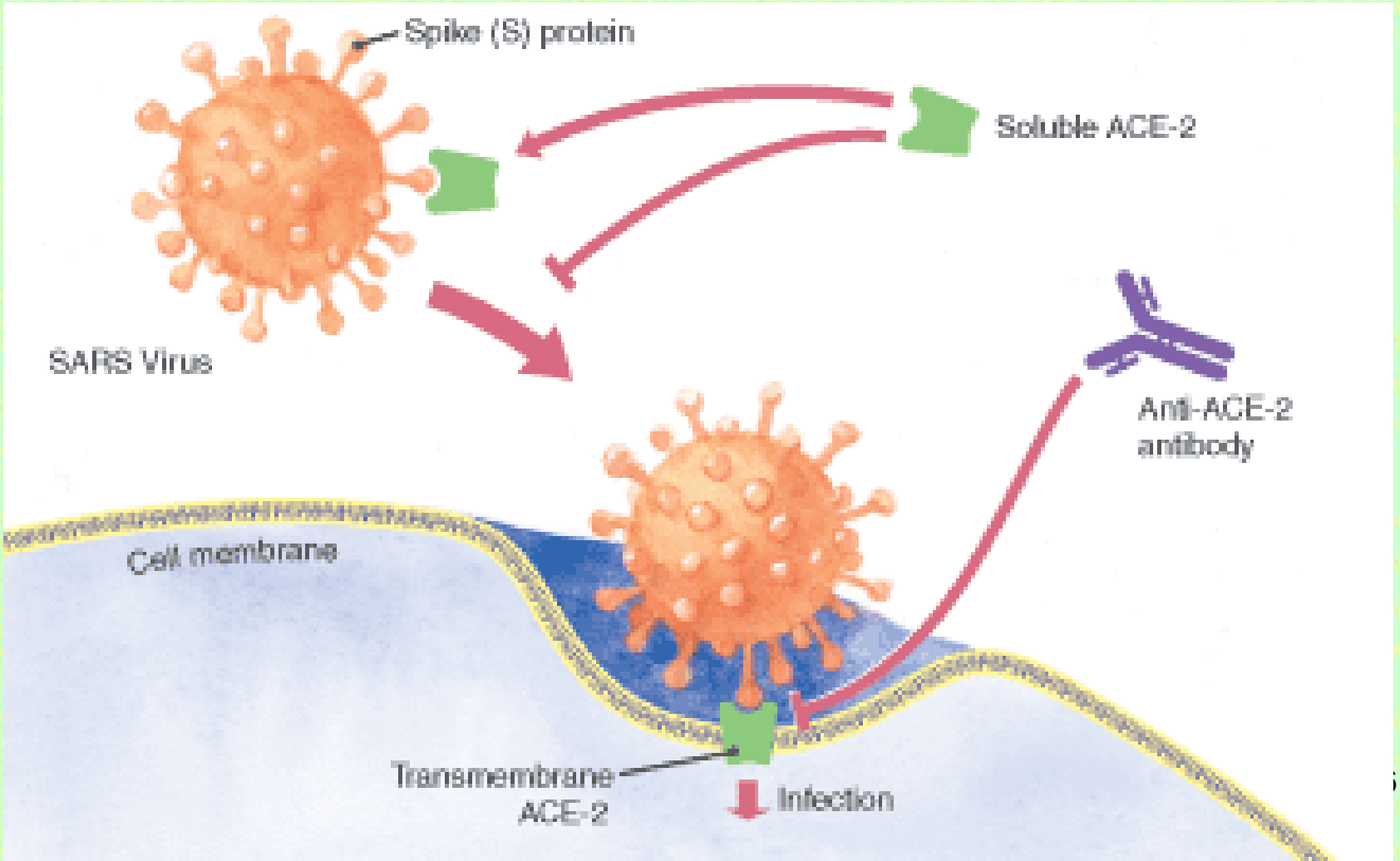
The SARS corona virus



The SARS corona virus



The SARS corona virus



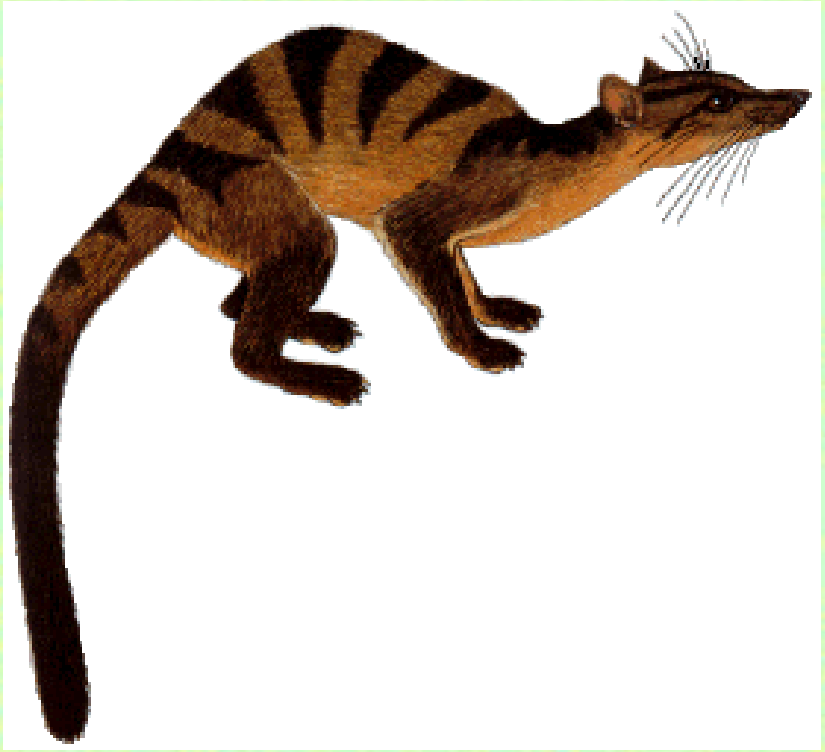
The SARS corona virus

* April 2003, a laboratory in Canada announced the entire RNA genome sequence of the *SARS* CoV virus.

* Phylogenetic analysis of the *SARS* corona virus showed that the most closely related CoV is the *palm civet*.

* The palm civet is a popular food item in the Guangdong province of China.

SARS: the outbreak



Palm civet *alive*

Palm civet *as Chinese food item*

Phylogenetic analysis of SARS CoV

* May 2003, 2 papers in Science reported the full genome of *SARS* CoV.

* Genome of *SARS* CoV contains 29,751 bp.

* Substantially different from all human CoVs.

* Also different from bird CoVs – so no relation to bird flue.

* End 2003 *SARS* had spread over the entire world

Phylogenetic analysis of SARS CoV

Phylogenetic analysis helps to answer:

* What kind of virus caused the original infection?

* What is the source of the infection?

* When and where did the virus cross the species border?

* What are the key mutations that enabled this switch?

* What was the trajectory of the spread of the virus?

Case study: phylogenetic analysis of the SARS epdemic

* Genome of SARS-CoV: 6 genes

* Identify host: Himalayan Palm Civet

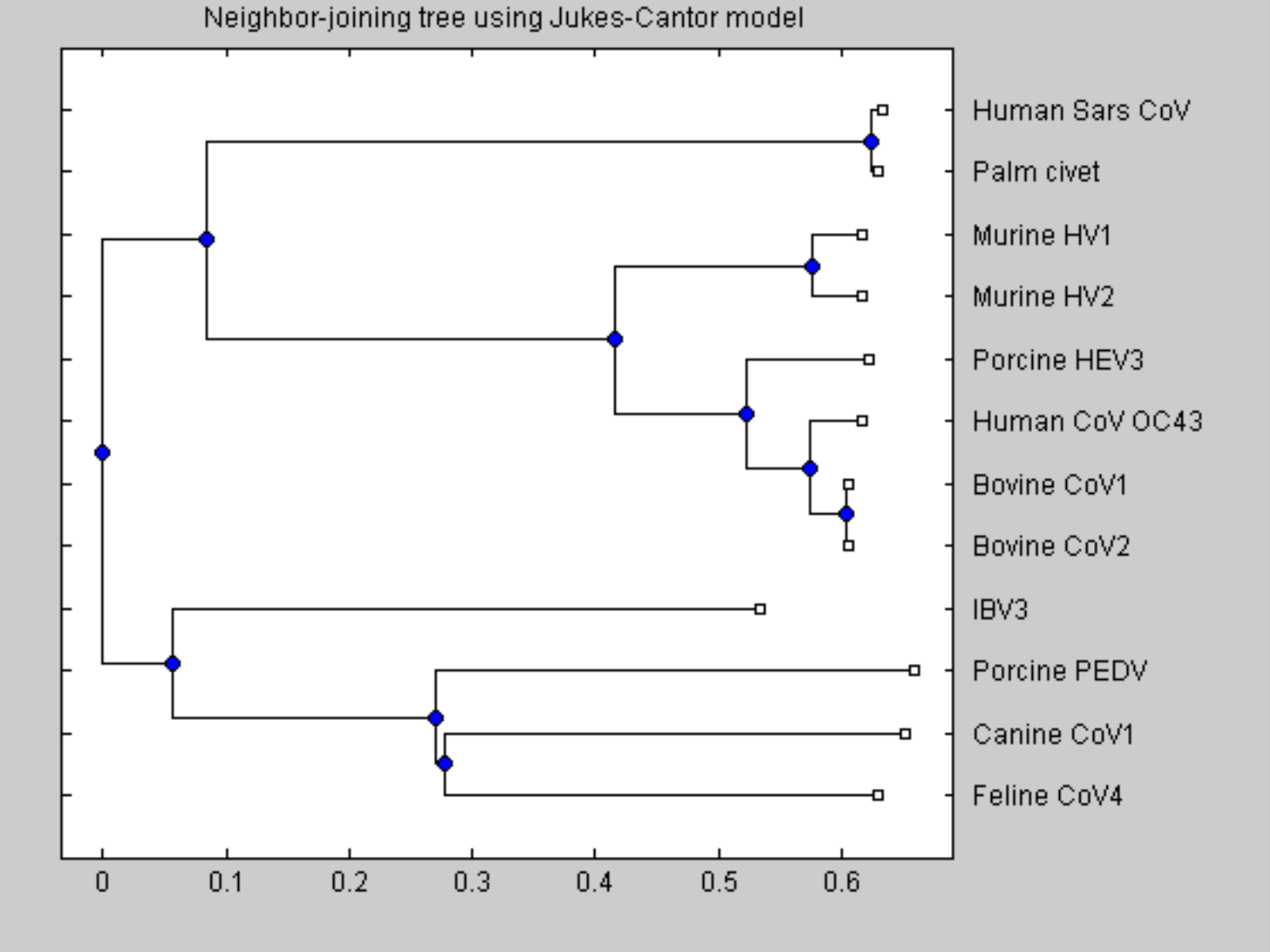
* The epidemic tree

* The date of origin

* Area of Origin

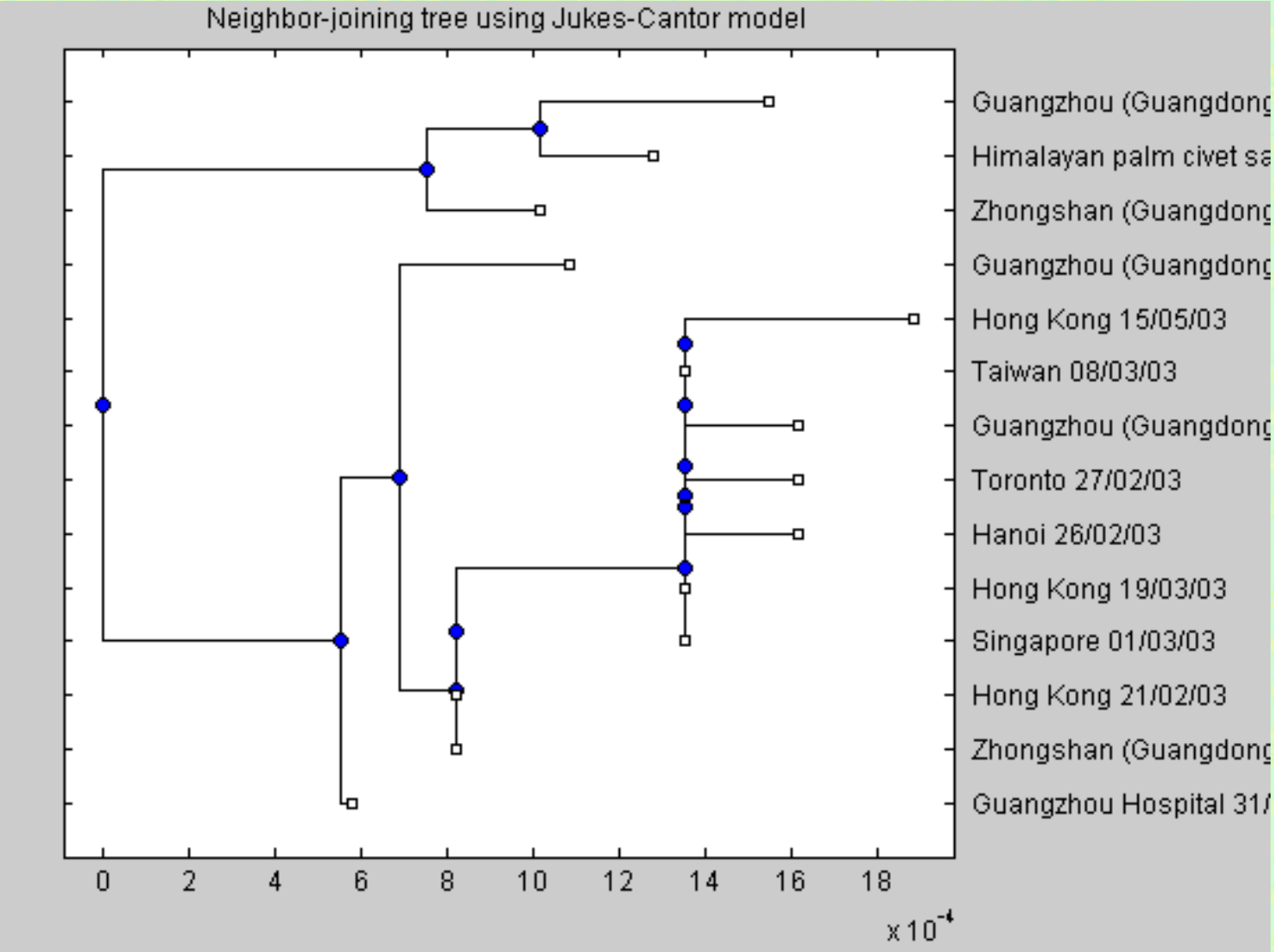
PHYLOGENETIC TREES

phylogenetic analysis of SARS : Identifying the Host



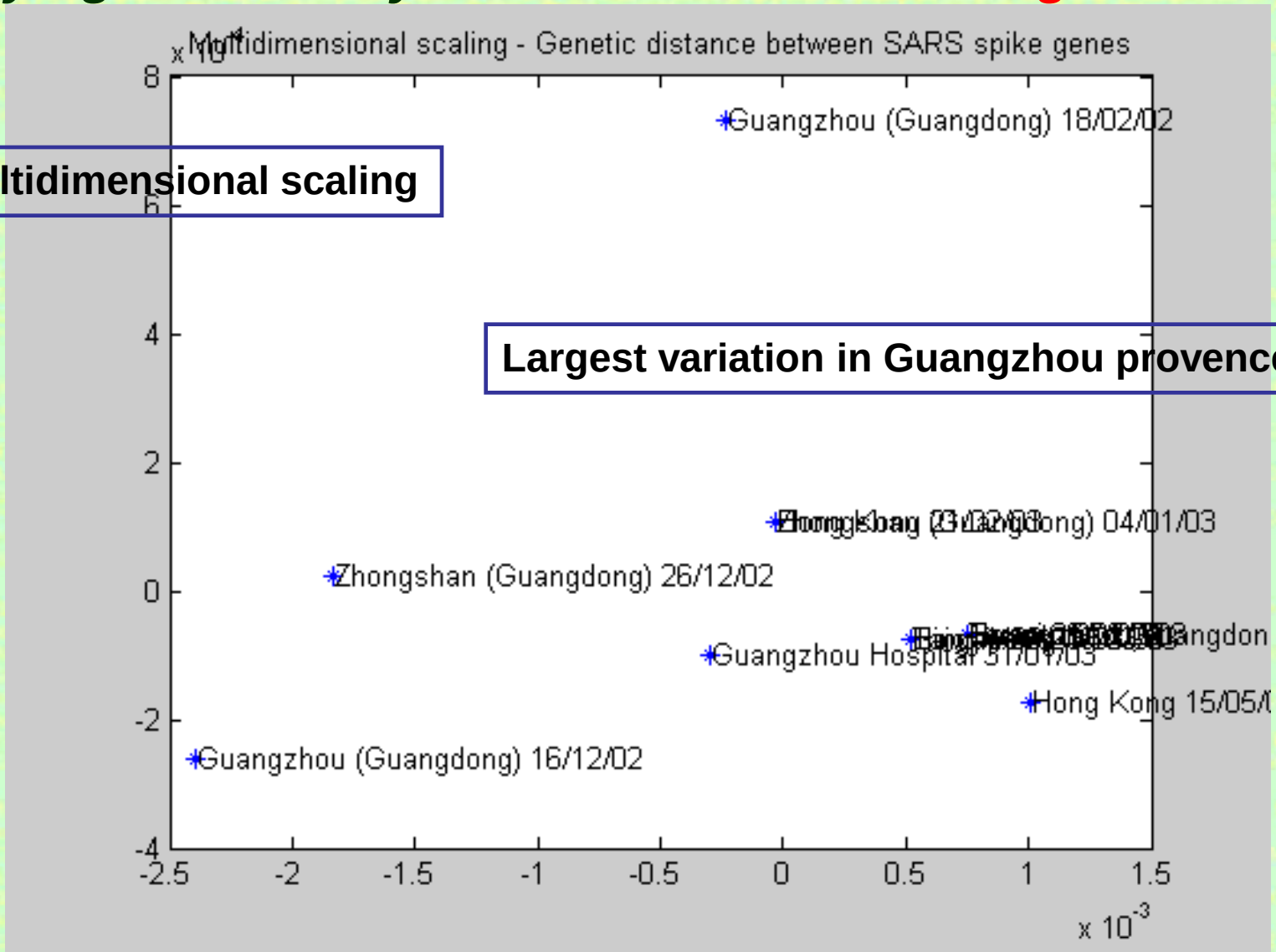
PHYLOGENETIC TREES

phylogenetic analysis of SARS : The epidemic tree



PHYLOGENETIC TREES

phylogenetic analysis of SARS : Area of origin

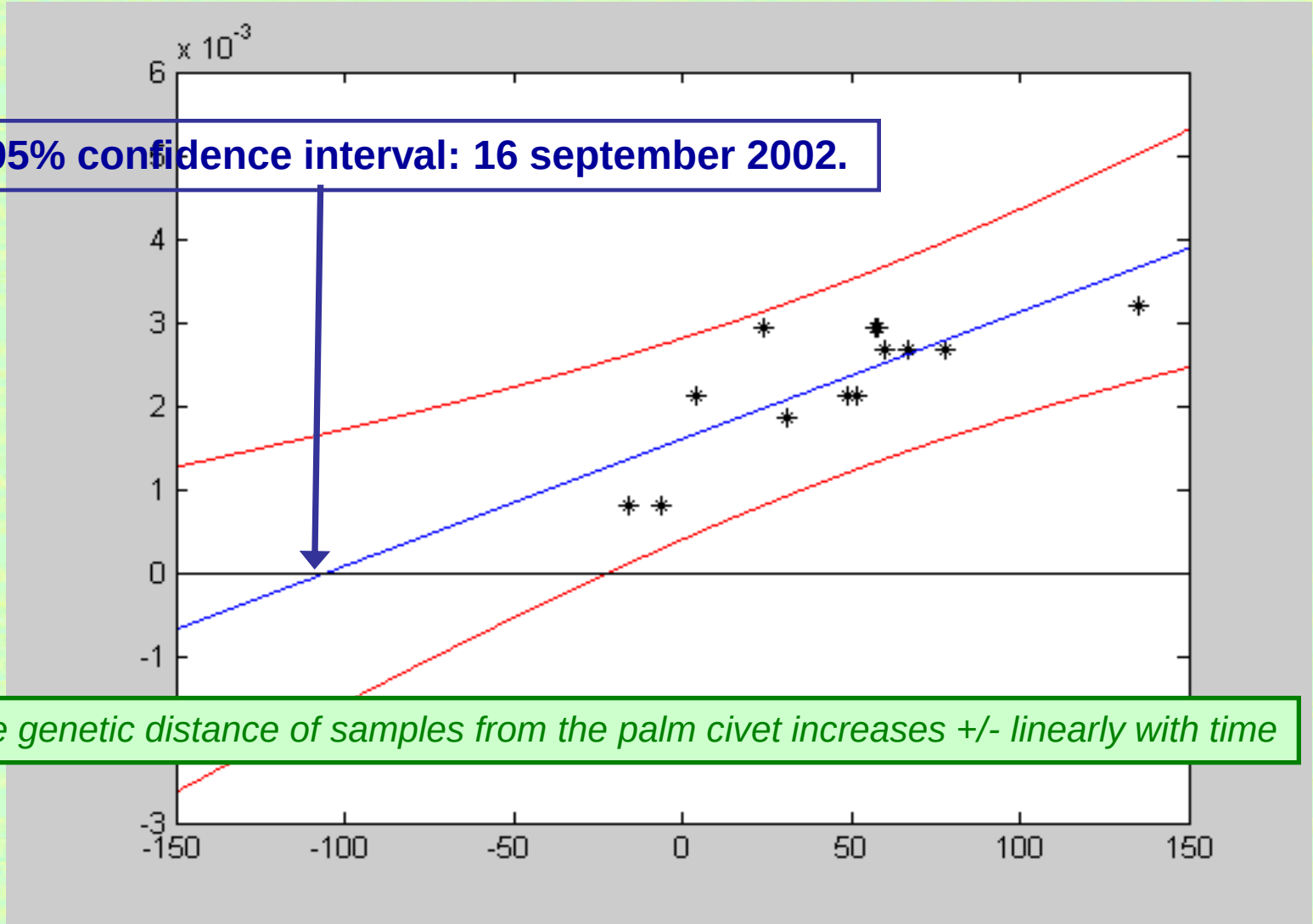


multidimensional scaling

Largest variation in Guangzhou provence

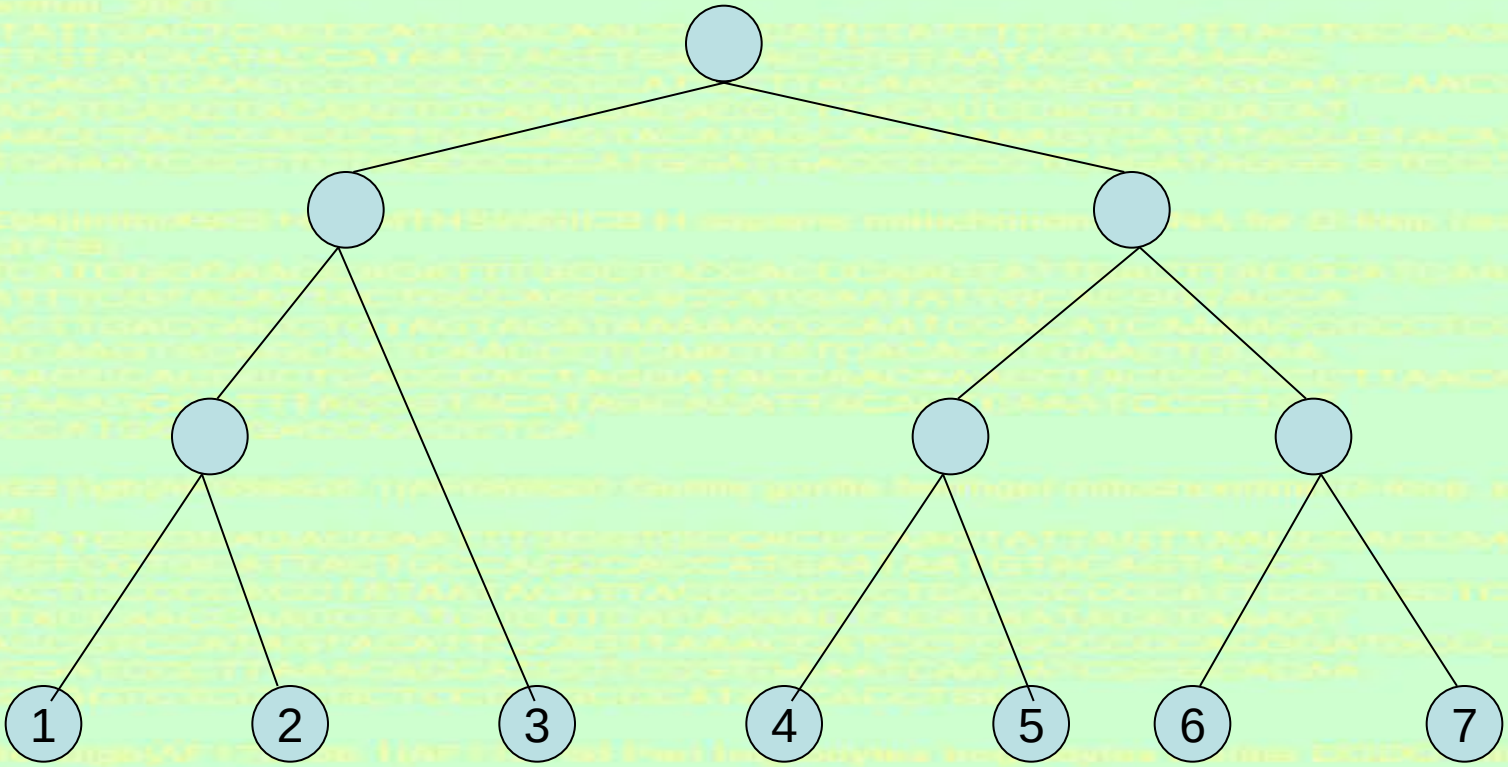
PHYLOGENETIC TREES

phylogenetic analysis of SARS : Date of origin



The genetic distance of samples from the palm civet increases +/- linearly with time

THE NEWICK FORMAT



Newick format: (((1,2),3),((4,5),(6,7)))

REFERENCES AND RECOMMENDED READING

GENERAL:

Molecular evolution, a phylogenetic approach, Roderic Page, Edward Holmes; Blackwell Science, Oxford, UK, 3d Edition, 2001

Computational Genomics, a case study approach, Nello Christianini, Matthew Hahn, Cambridge University press, Cambridge UK, 2007

APPLY AND USE:

A Practical Approach to Phylogenetic Analysis and Hypothesis Testing, Philippe Lemey, Marco Salemi, Anne-Mieke Vandamme, Cambridge University Press, Cambridge UK, 2007

MATHEMATICAL BACKGROUND:

T-theory: An Overview, A. Dress, V. Moulton, W. Terhalle**,** *European Journal of Combinatorics* **17** (2–3): 161–175.

END of LECTURE

Appendix *