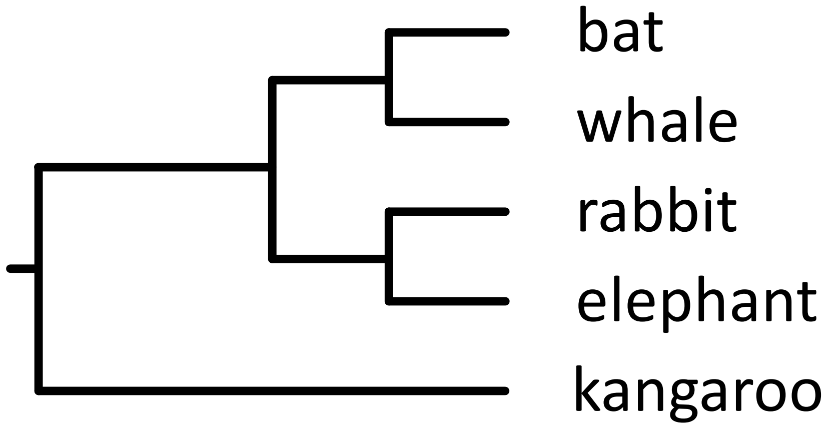
Lecture 1.4

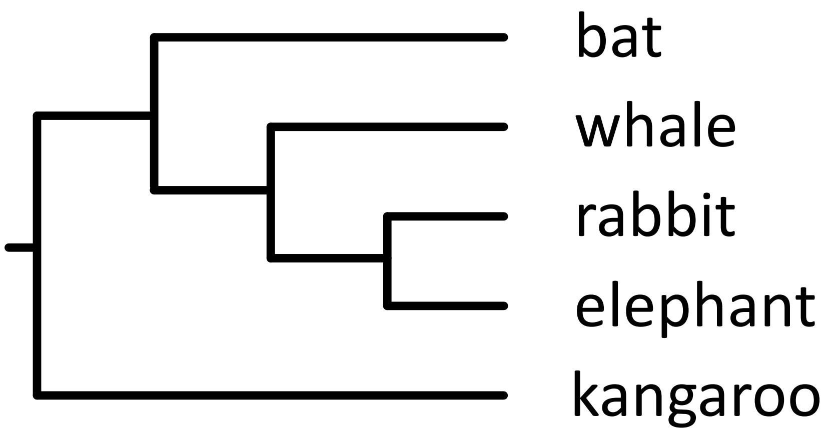
# Phylogenetic Methods

# Maximum parsimony

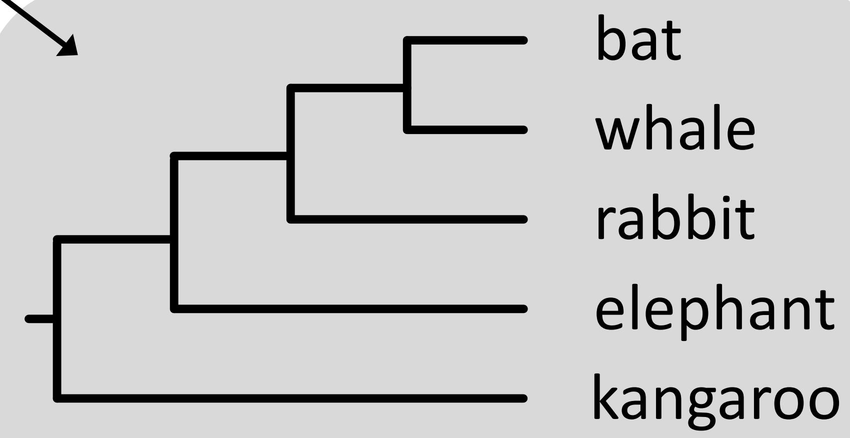
| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |



**7 steps**



**8 steps**



**6 steps**

# Popular phylogenetic methods

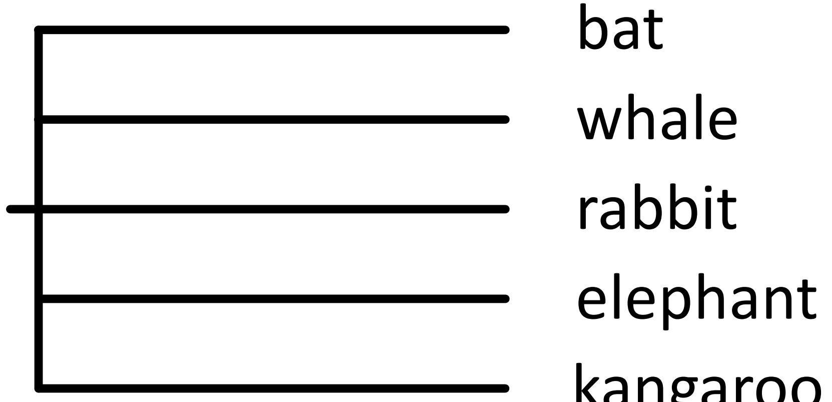
1. Maximum parsimony
2. Distance-based methods
3. Maximum likelihood
4. Bayesian inference

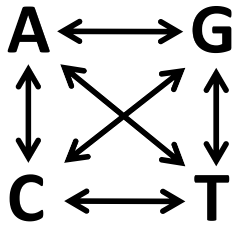
Model-based methods

# Distance-Based Methods

# Neighbour joining

| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |



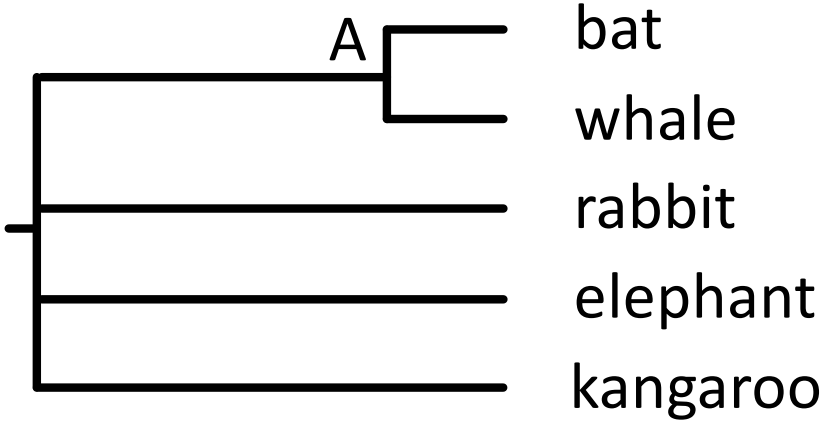


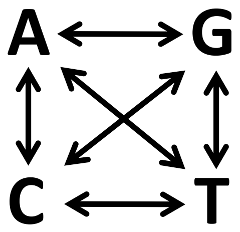
| | bat | whale | rabbit | elephant | kangaroo |
|---|---|---|---|---|---|
| bat | – | | | | |
| whale | .15 | – | | | |
| rabbit | .20 | .25 | – | | |
| elephant | .35 | .40 | .35 | – | |
| kangaroo | .55 | .60 | .55 | .55 | – |

**Clustering algorithm**

# Neighbour joining

| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |



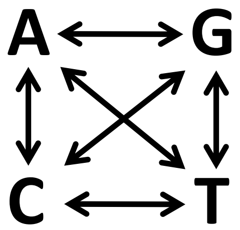


| | bat | whale | rabbit | elephant | kangaroo |
|---|---|---|---|---|---|
| bat | – | | | | |
| whale | .15 | – | | | |
| rabbit | .20 | .25 | – | | |
| elephant | .35 | .40 | .35 | – | |
| kangaroo | .55 | .60 | .55 | .55 | – |

**Clustering algorithm**

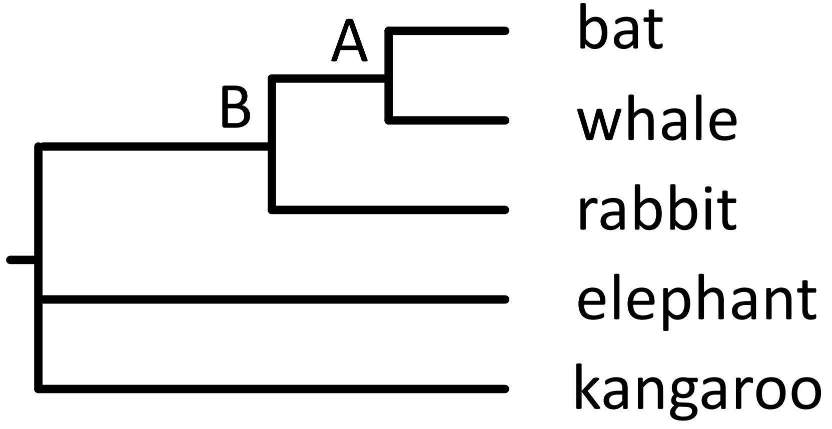
# Neighbour joining

| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |



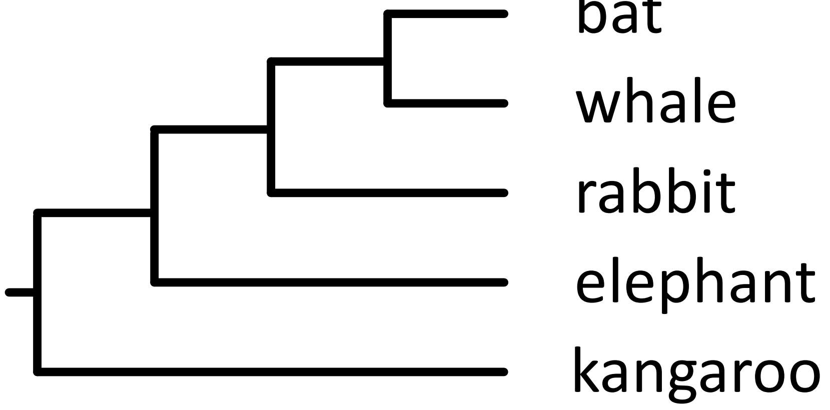
| | A | rabbit | elephant | kangaroo |
|---|---|---|---|---|
| A | – | | | |
| rabbit | .15 | – | | |
| elephant | .30 | .35 | – | |
| kangaroo | .50 | .55 | .60 | – |

**Clustering algorithm**



# Neighbour joining

| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |



# Distance-based methods

- **Clustering algorithms**
  - Unweighted pair group method with arithmetic mean (UPGMA)
  - Neighbour joining

- **Tree searching using optimality criteria**
  - Minimum evolution
  - Least-squares inference

# Strengths and weaknesses

- **Strengths**
  - Very quick
  - Deals with multiple substitutions and long-branch attraction

- **Weaknesses**
  - Loss of information in pairwise comparisons
  - Unable to implement sophisticated evolutionary models

# Maximum Likelihood

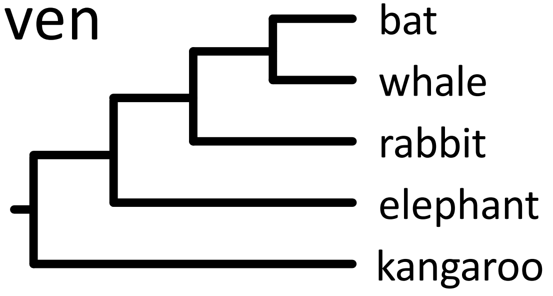
# Maximum likelihood

Likelihood of hypothesis *H* =

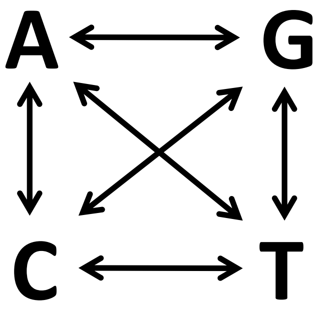
$$\boxed{P(D|H)}$$

Probability of the data, given the hypothesis

Given



+

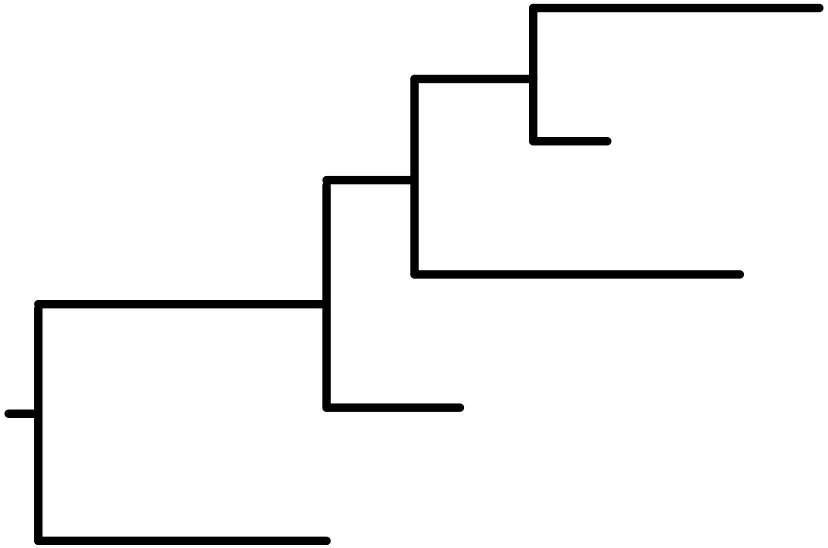


→

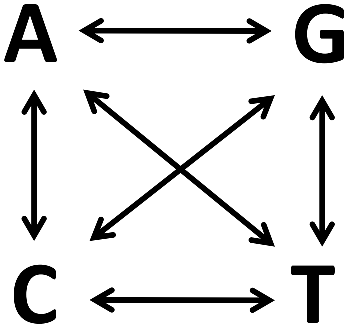
Probability of?

| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |

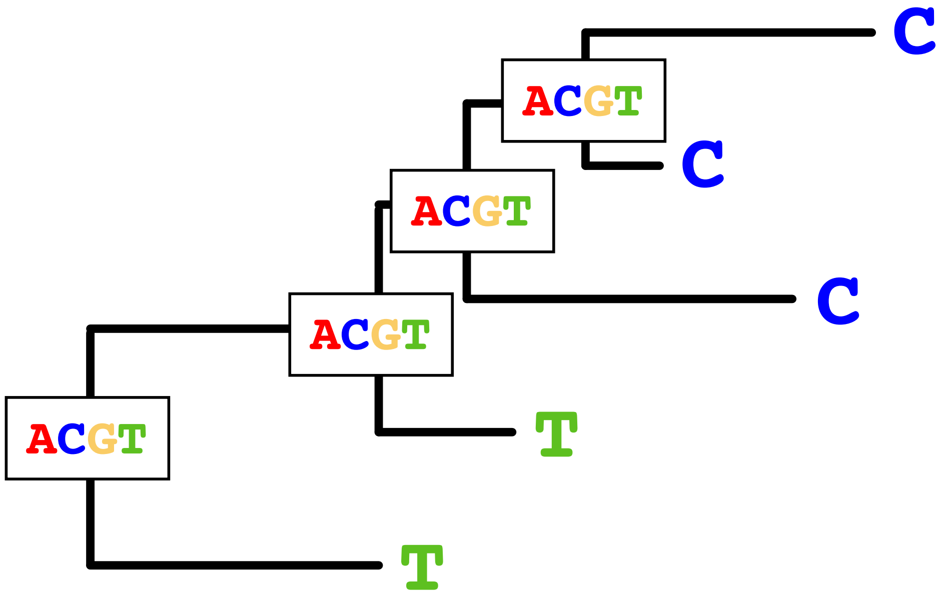
# Maximum likelihood



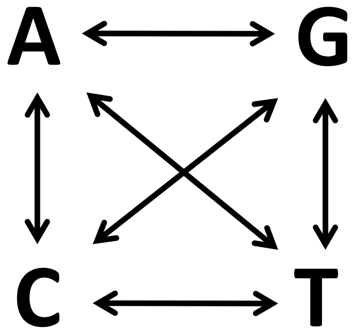
| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |



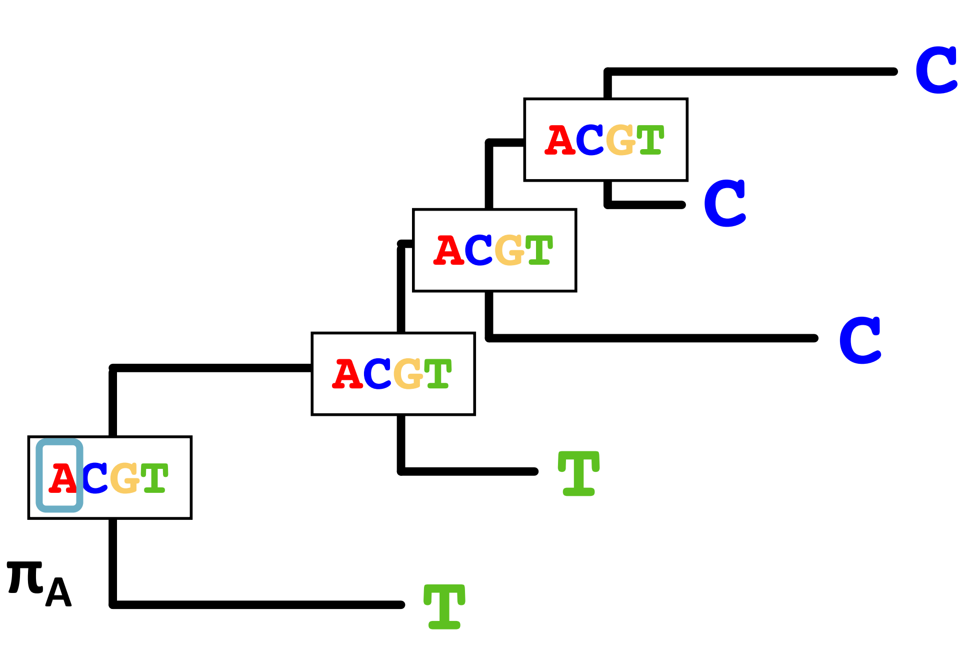
# Maximum likelihood



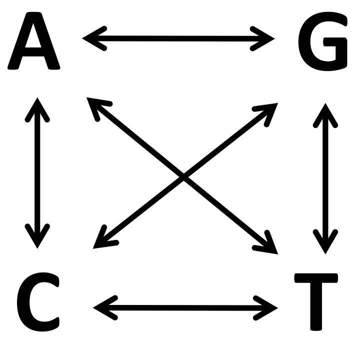
| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |



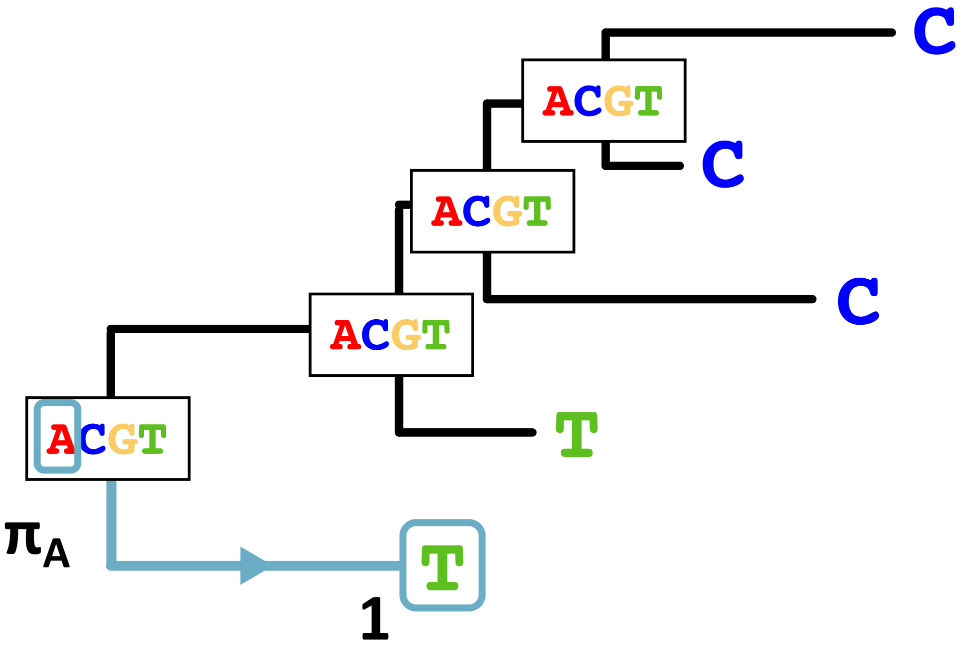
# Maximum likelihood



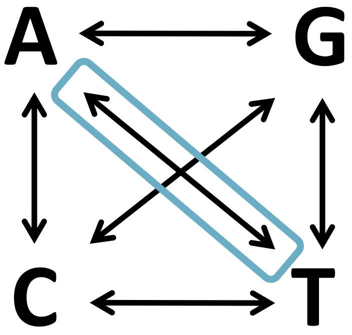
| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |



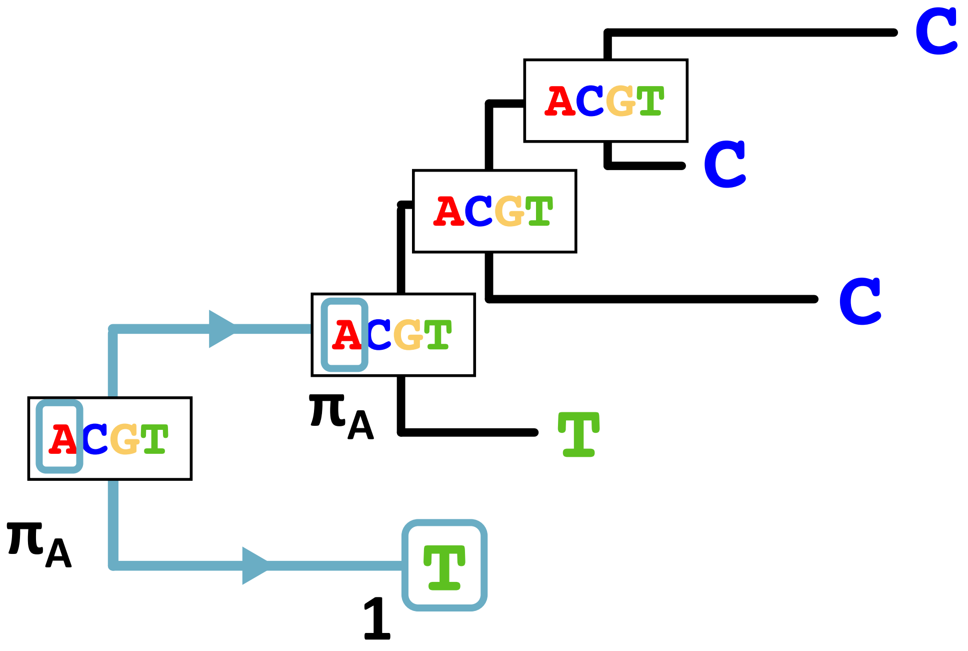
# Maximum likelihood



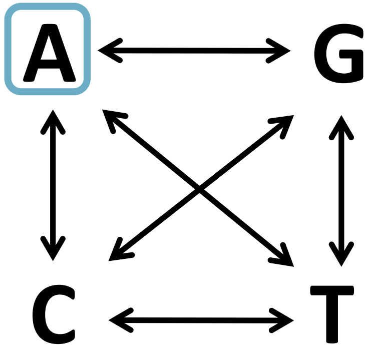
| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |



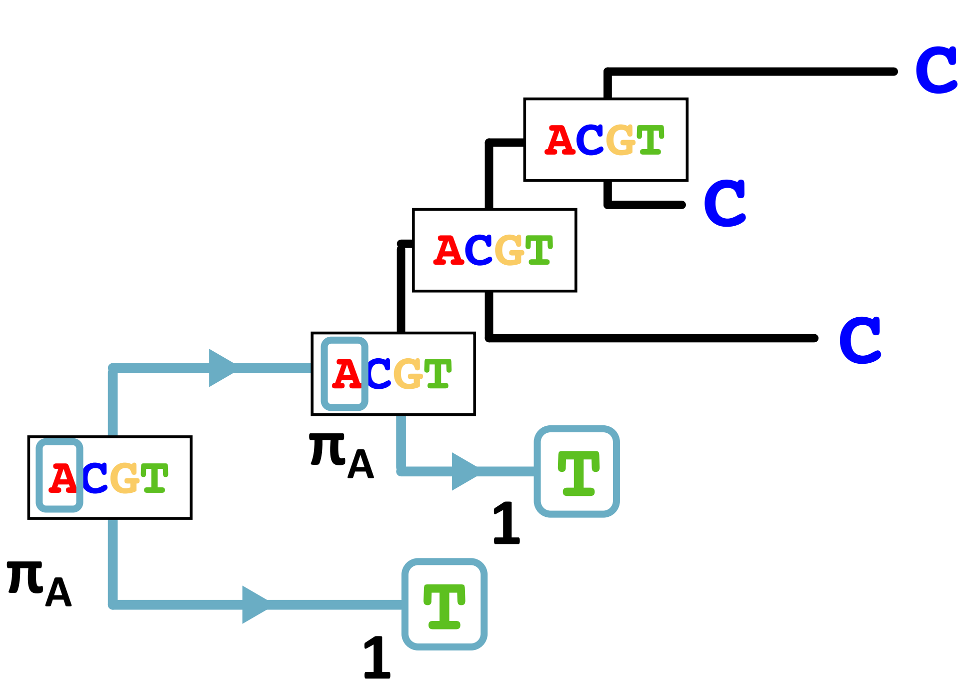
# Maximum likelihood



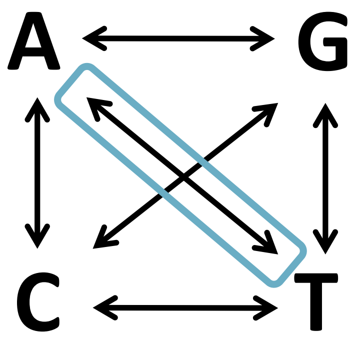
| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |



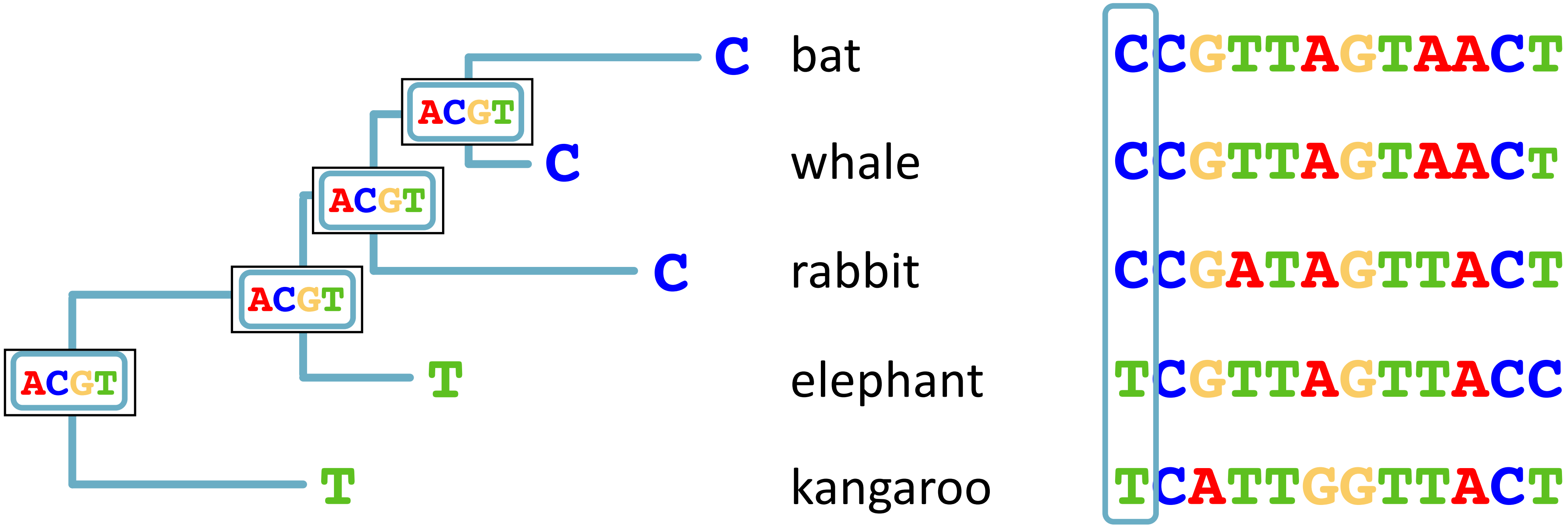
# Maximum likelihood



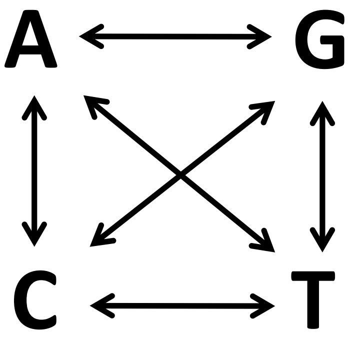
| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |



# Maximum likelihood

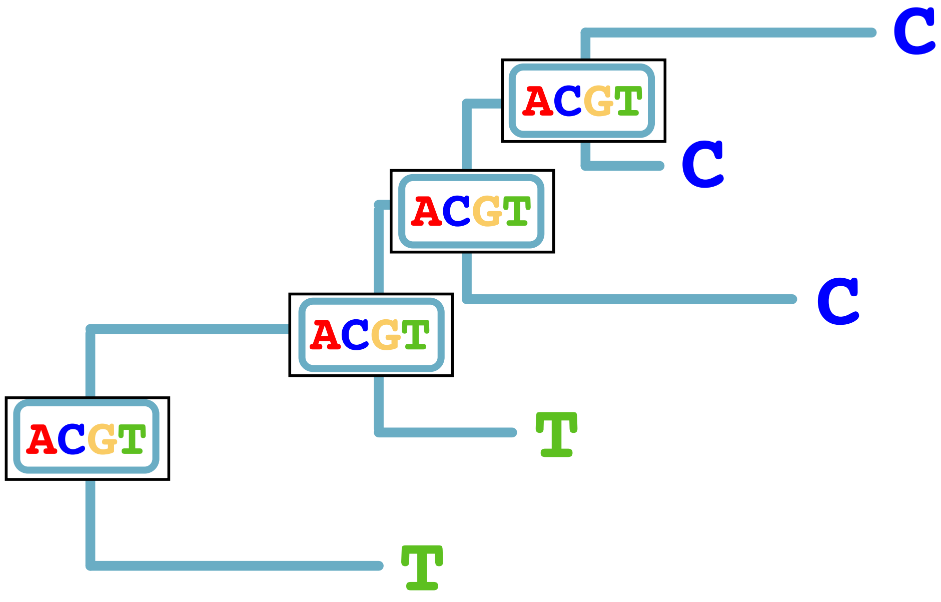


| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |

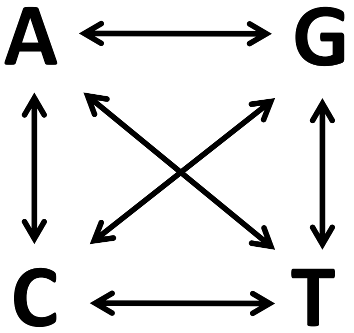


Likelihood is summed over all possibilities

# Maximum likelihood



| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |

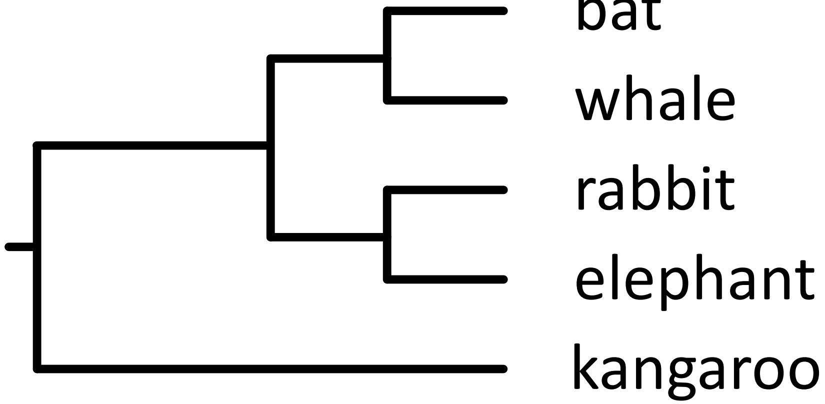


Likelihood is multiplied across all sites

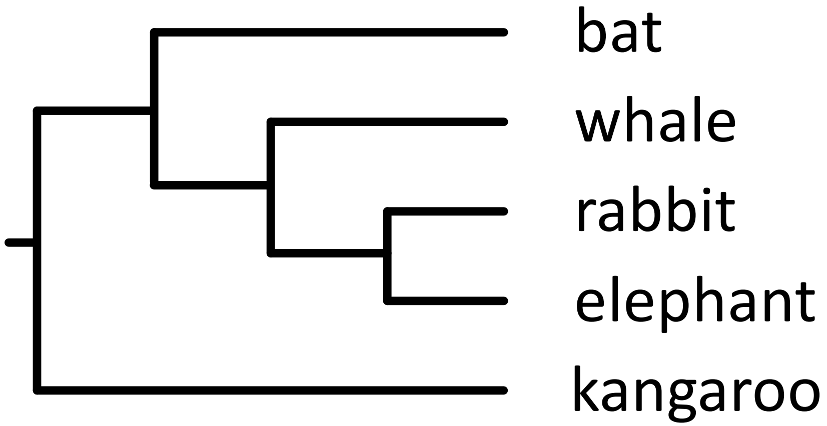
Very low probability of observing
any particular alignment

# Maximum likelihood

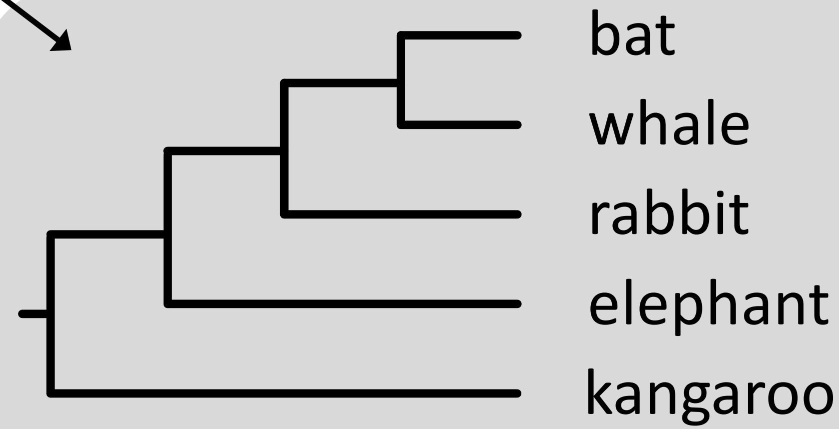
| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |



**lnL = -1203.83**



**lnL = -1241.47**



**lnL = -1008.58**

# Likelihood optimisation

- Search through the space of possible trees (including branch lengths) and model parameter values
- Calculate the likelihood for these
- Find best tree and model parameter values
- Multivariate optimisation

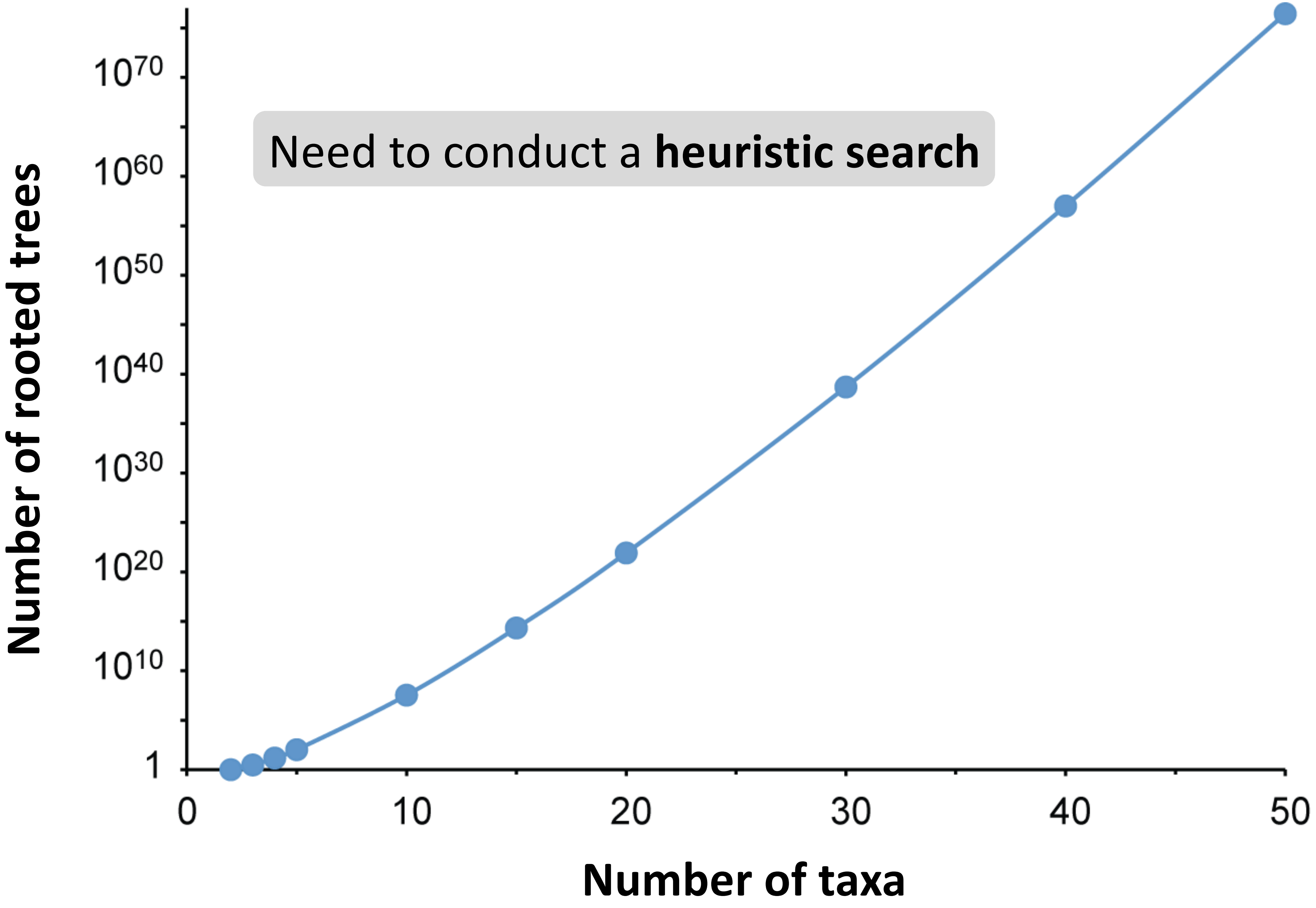
# Finding the best tree

- For *n* taxa, the number of possible unrooted trees ($B_n$) is:

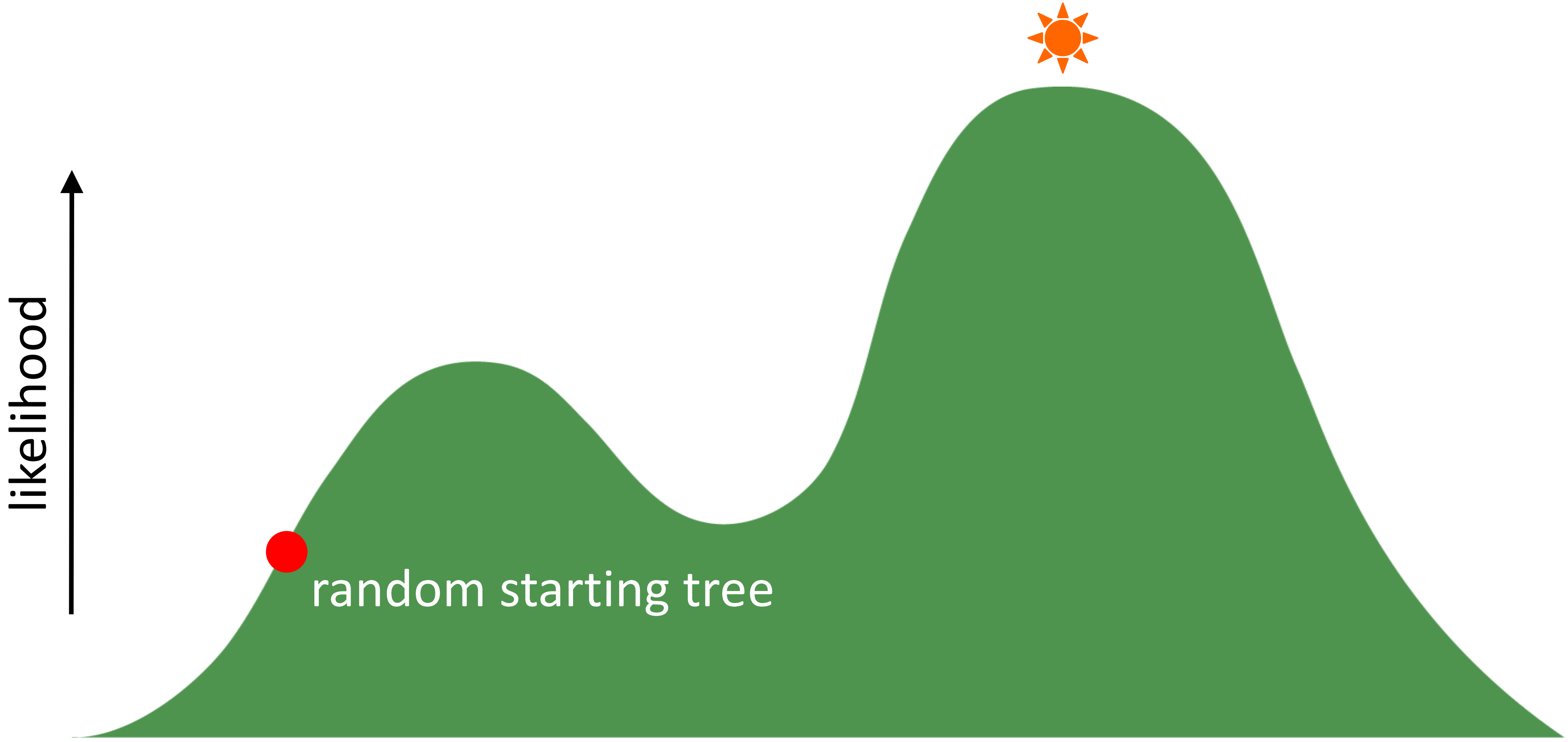
$$B_n = 1 \times 3 \times 5 \times \ldots \times (2n-5) = \prod_{i=3}^{n} (2i-5)$$

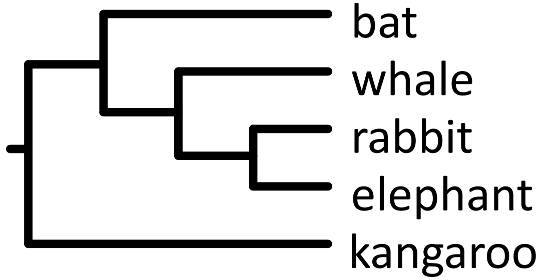
- For example:
    - 4 taxa → 3 trees
    - 5 taxa → 15 trees
    - 10 taxa → 2,027,025 trees

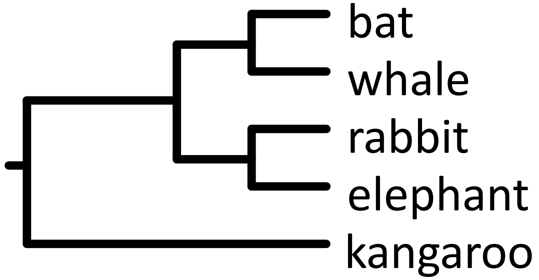
# Finding the best tree

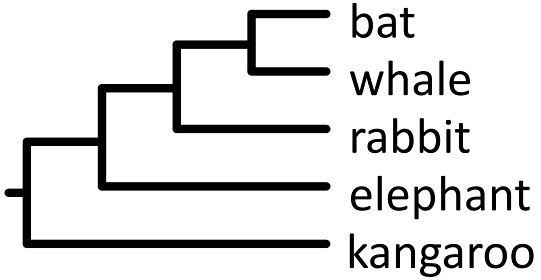

Need to conduct a **heuristic search**

# Heuristic search

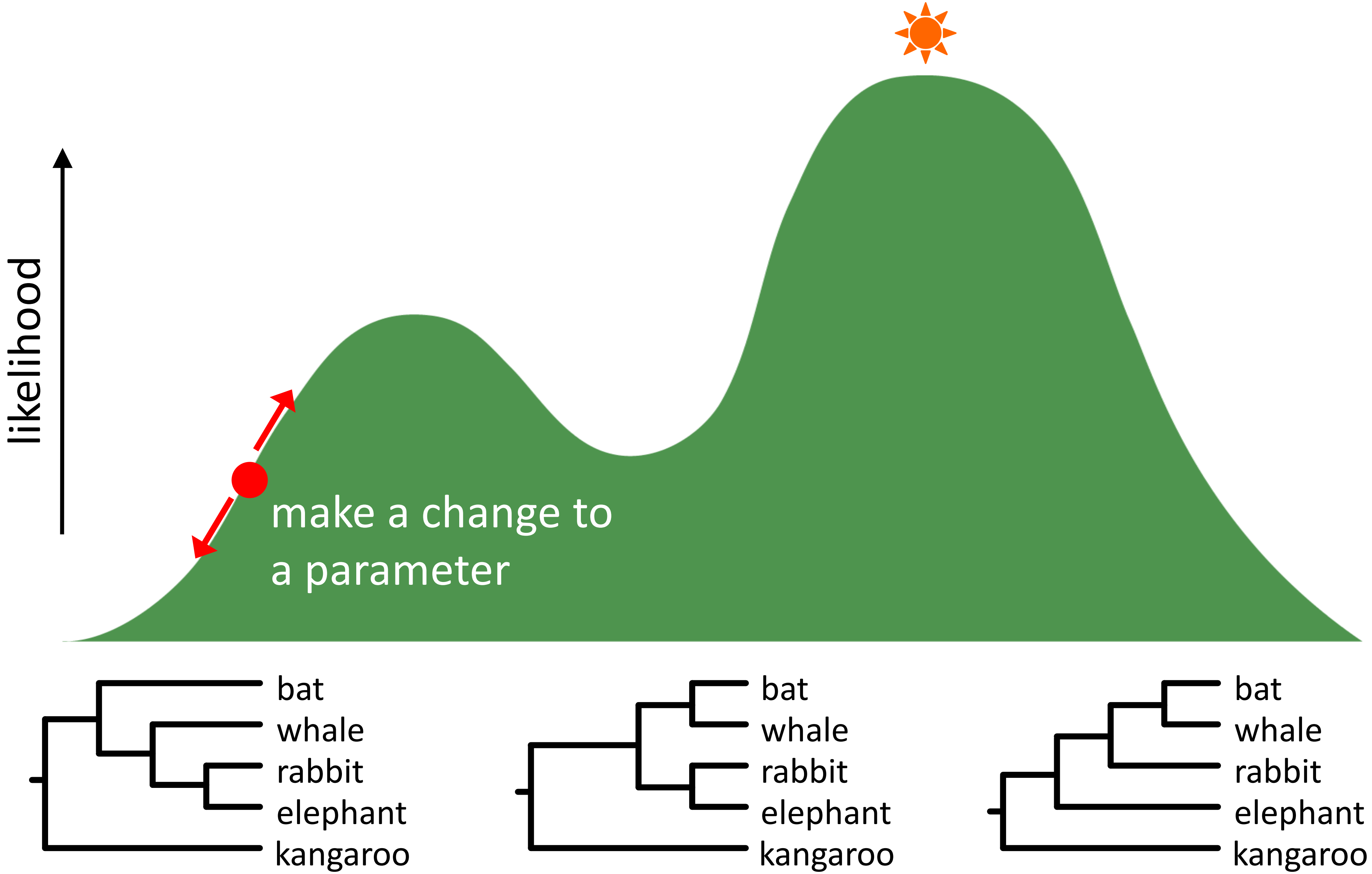






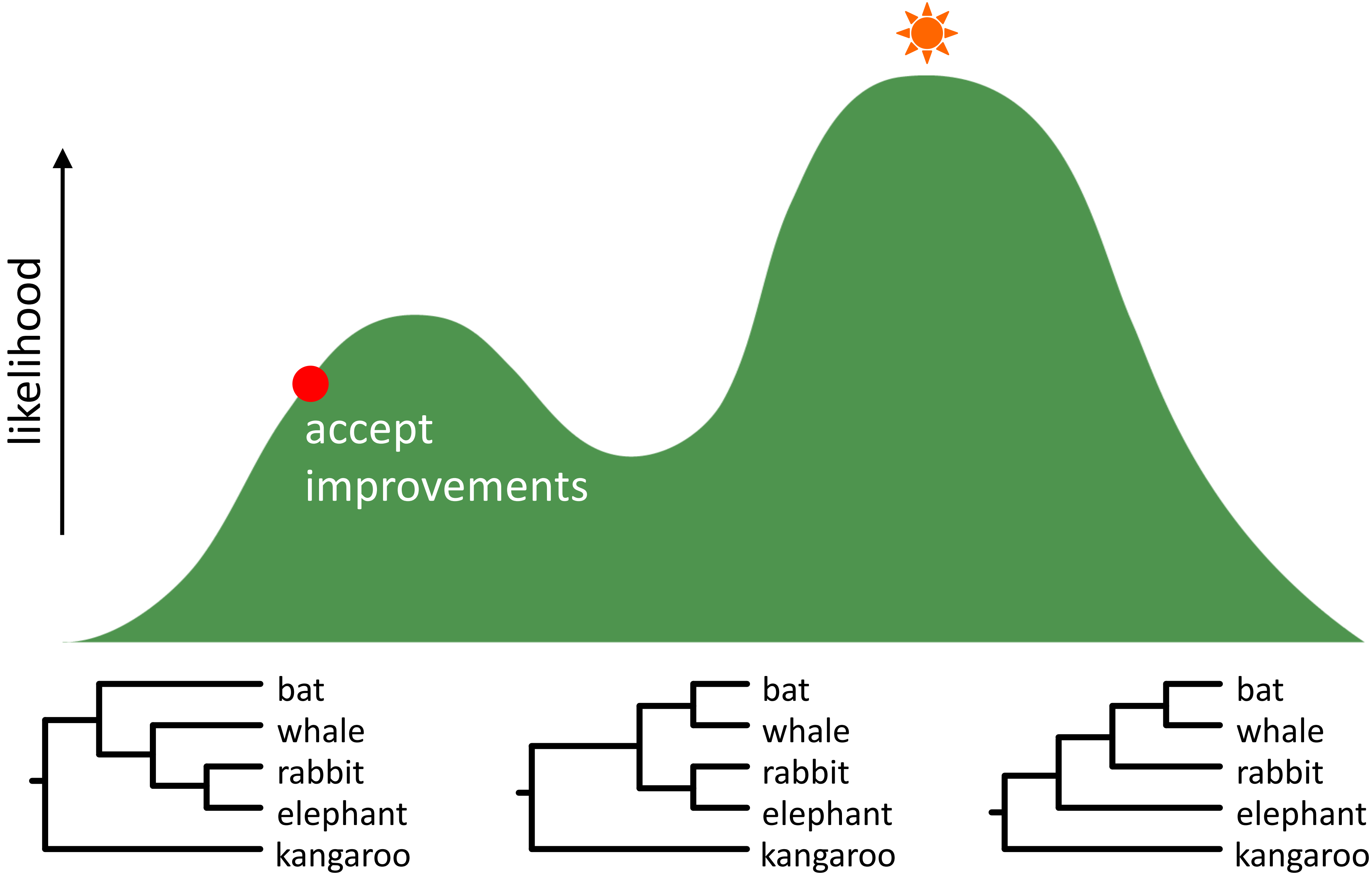


# Heuristic search



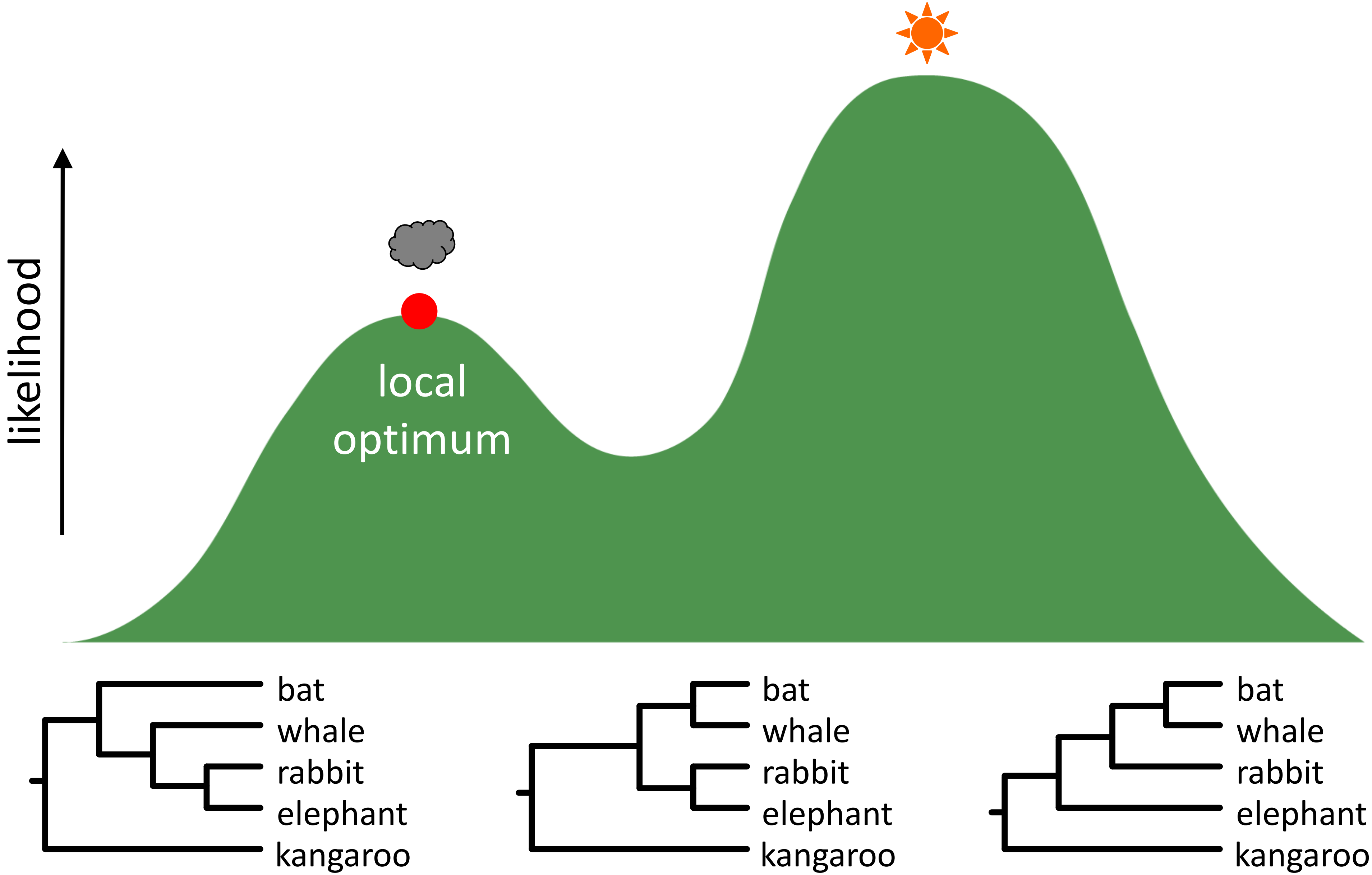
make a change to a parameter

# Heuristic search

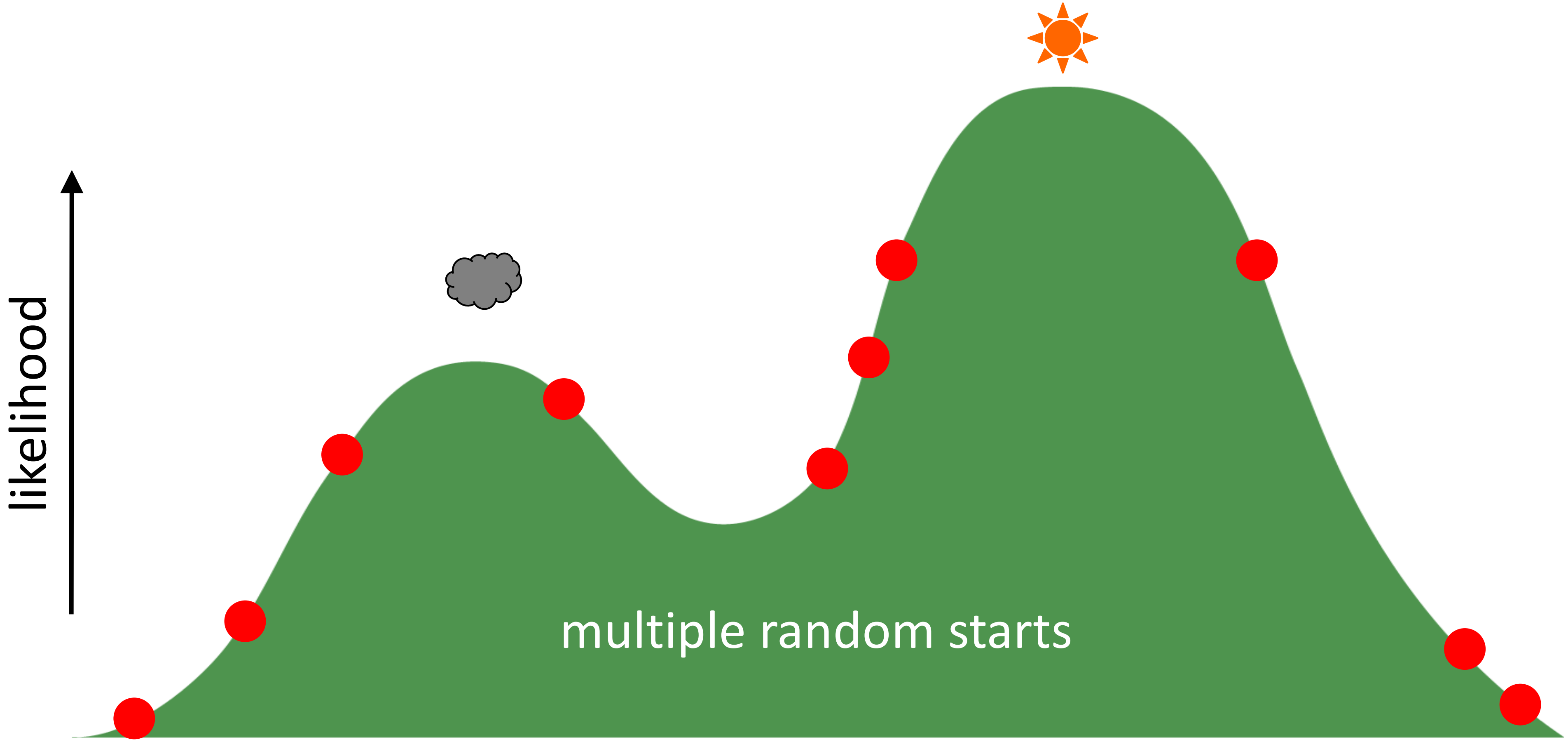


accept improvements

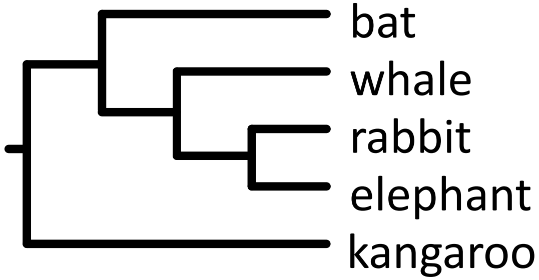
# Heuristic search

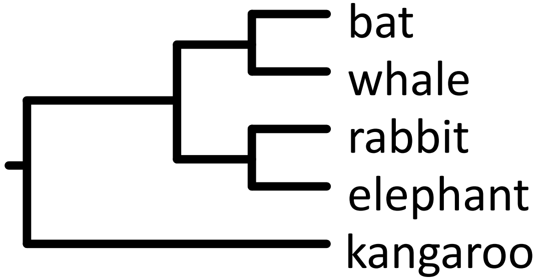


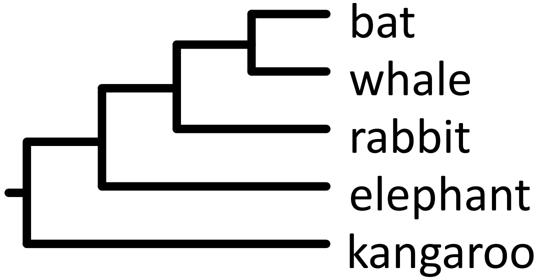
# Heuristic search



multiple random starts

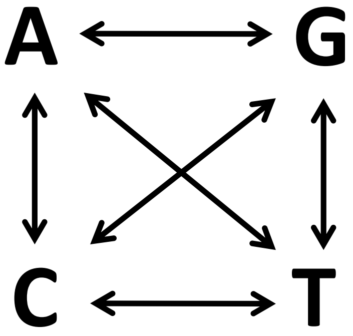






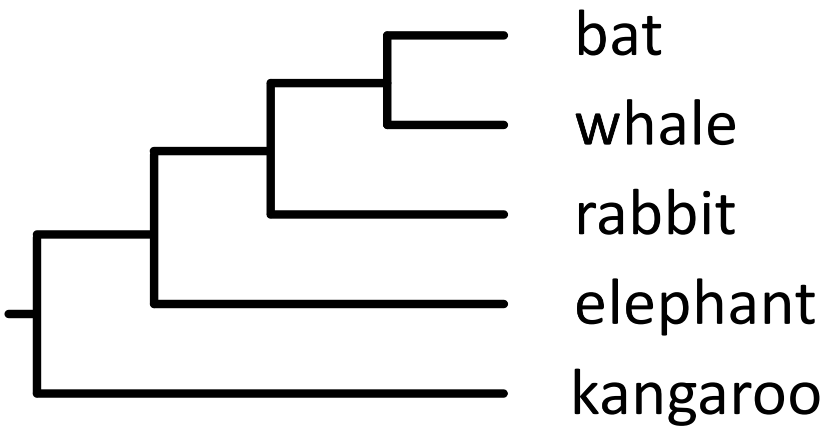
# Maximum-likelihood estimates

A single set of maximum-likelihood estimates of model parameters



$\boldsymbol{\pi_A, \pi_C, \pi_G, \pi_T}$

A single maximum-likelihood tree



# Strengths and weaknesses

- **Strengths**
  - Rigorous statistical method
  - Deals with multiple substitutions and long-branch attraction
  - Robust to violations of assumptions
- **Weaknesses**
  - Generally not feasible to implement very parameter-rich models
  - Searching tree space can be difficult

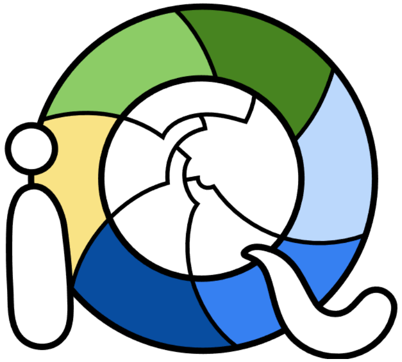
# Software

**RAxML**

**PhyML**

**MEGA**

**PAML**



**IQ-TREE**

# Bootstrapping

# Nonparametric bootstrap

- Uncertainty in the estimate of the tree is inferred indirectly using **bootstrapping analysis**
- “pull oneself up by one's bootstraps”
- Bootstrapping analysis can be used in various phylogenetic methods:
  - Maximum parsimony
  - Distance-based methods
  - Maximum likelihood

# Bootstrapping

| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |

**Randomly sample sites (with replacement)**

| | |
|---|---|
| bat | T |
| whale | T |
| rabbit | A |
| elephant | T |
| kangaroo | T |

# Bootstrapping

| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |

| | |
|---|---|
| bat | TG |
| whale | TG |
| rabbit | AG |
| elephant | TG |
| kangaroo | TG |

# Bootstrapping

| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |

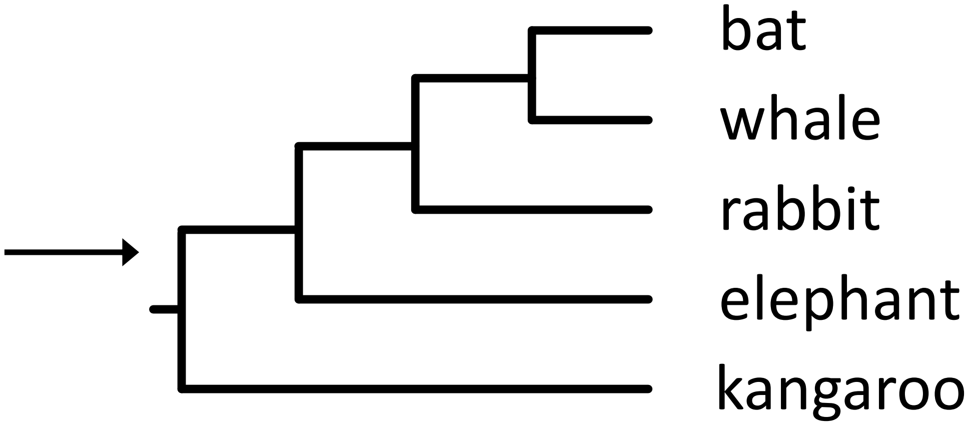
| | |
|---|---|
| bat | TGCCCTTAGCAC |
| whale | TGCCCTTAGCAC |
| rabbit | AGCCCATAGCAC |
| elephant | TGCTCTCAGCAT |
| kangaroo | TGCTCTTAACGT |

# Bootstrapping

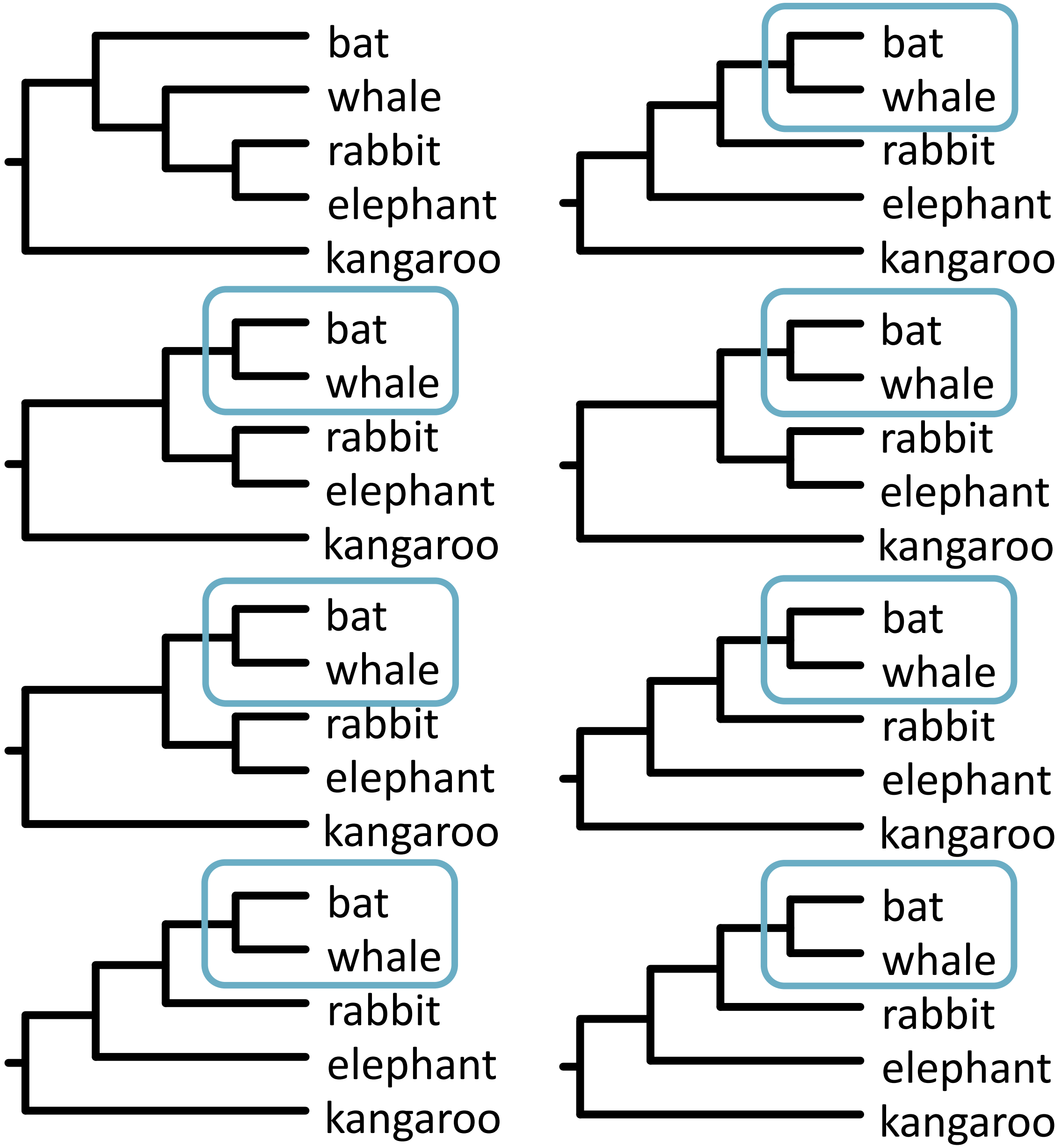
| | |
|---|---|
| bat | CCGTTAGTAACT |
| whale | CCGTTAGTAACT |
| rabbit | CCGATAGTTACT |
| elephant | TCGTTAGTTACC |
| kangaroo | TCATTGGTTACT |

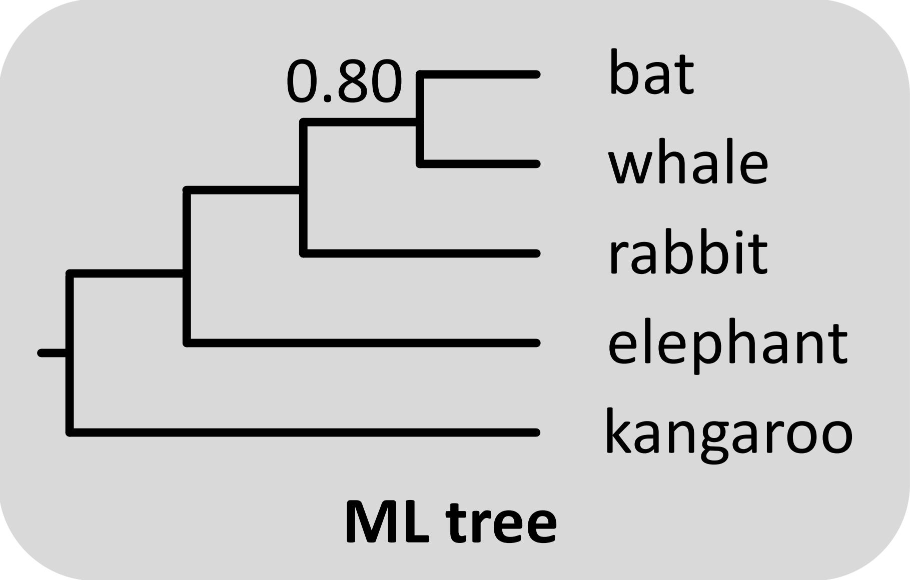
**Repeat 1,000 times**

| | |
|---|---|
| bat | TGCCCTTAGCAC |
| whale | TGCCCTTAGCAC |
| rabbit | AGCCCATAGCAC |
| elephant | TGCTCTCAGCAT |
| kangaroo | TGCTCTTAACGT |



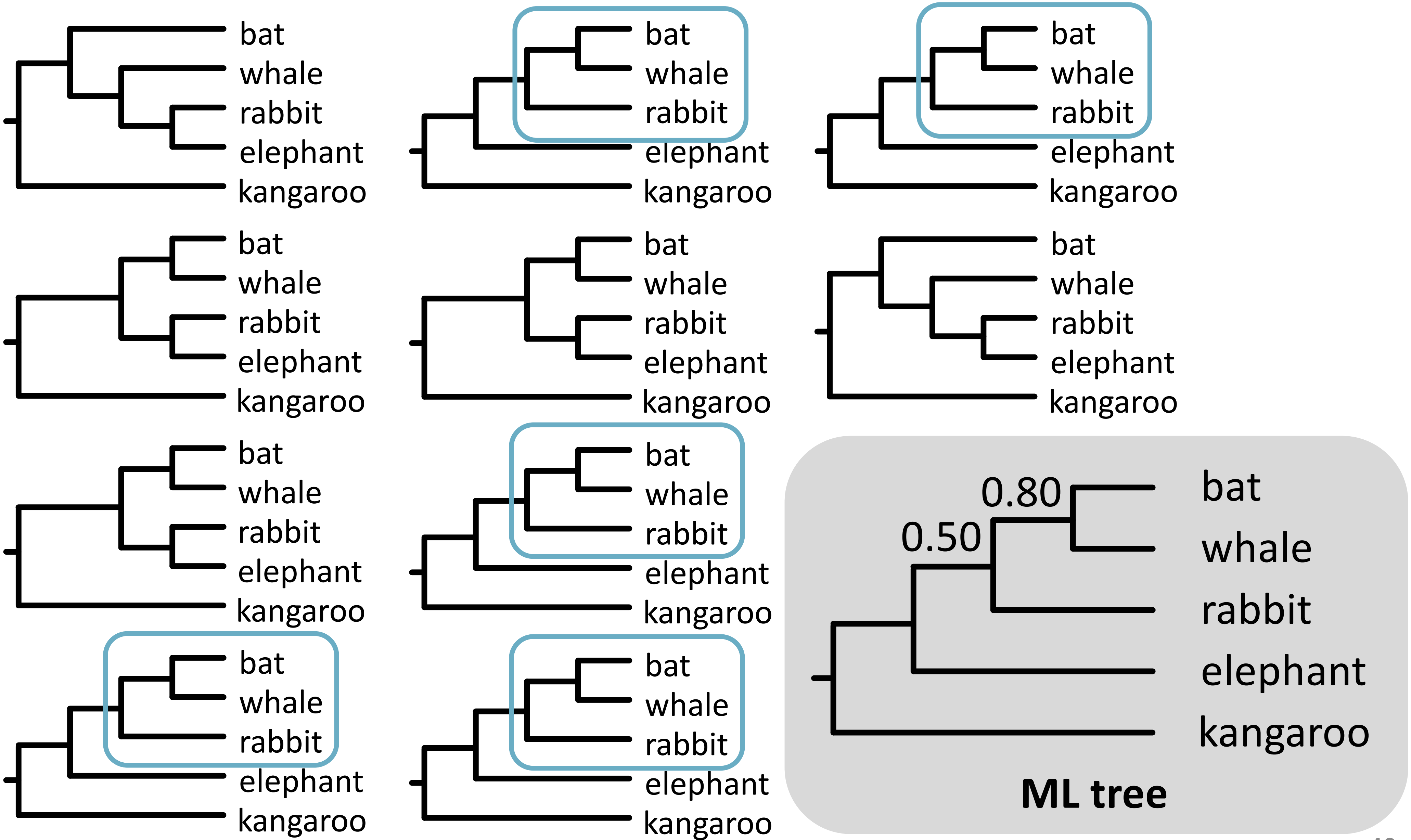
# Bootstrapping





**ML tree**

# Bootstrapping



# Interpreting bootstrap values

- **Felsenstein (1985)**
  bootstrapping provides a confidence interval that contains the *phylogeny that would be estimated from repeated sampling of many characters from the underlying set of all characters*

- Bootstrap values are **measures of repeatability**
  - High when the data set is large
  - Not meaningful when analysing genome-scale data

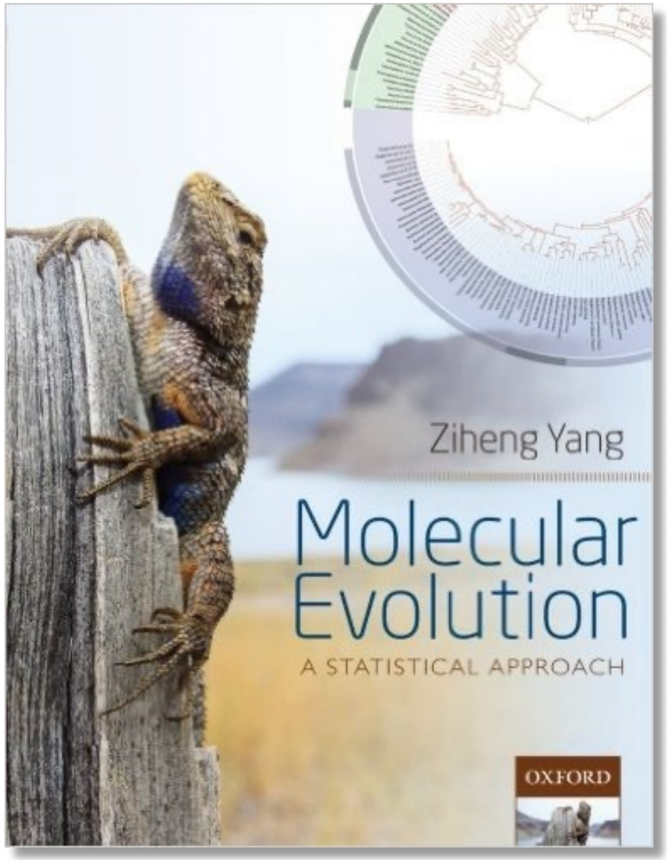
Soltis & Soltis (2003) *Stat Sci*

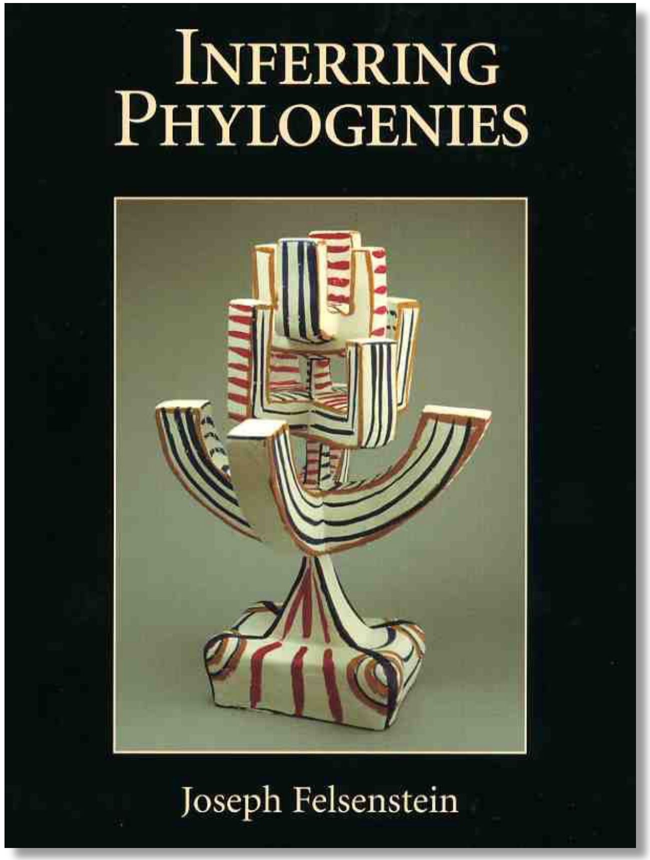
# Methods in practice

- **Maximum parsimony**
  - Commonly used to analyse morphological data
  - Rarely used to analyse molecular data
- **Distance-based methods**
  - Popular in some fields of research
  - Used to analyse very large data sets with many taxa
- **Maximum likelihood**
  - Widely used, lost some ground to Bayesian methods but is experiencing a resurgence (thanks to rapid ML methods)

# Useful references

- **Molecular phylogenetics: principles and practice**
  Yang & Rannala (2012) *Nature Reviews Genetics* 13: 303–314.


Ziheng Yang
Molecular Evolution
A STATISTICAL APPROACH
OXFORD


INFERRING PHYLOGENIES
Joseph Felsenstein