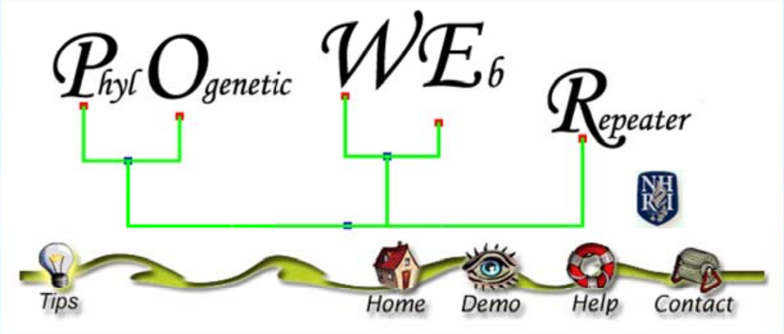
POWER: PhylOgenetic WEb Repeater

- An integrated and user-optimized framework for biomolecular phylogenetic analysis



Speaker: Chieh –Hua Lin (林介華)

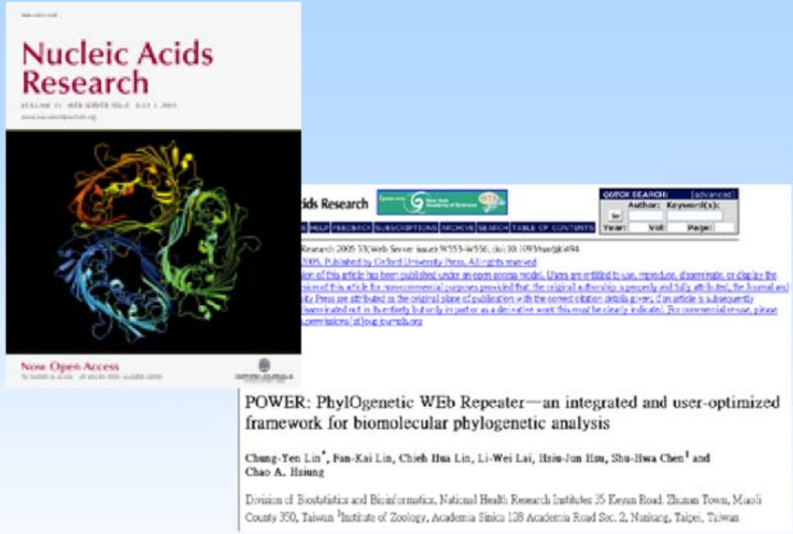
mammer@nhri.org.tw

Phylogenetic Analysis

- Based on observable evidence to estimate or infer their evolution relationship
- **Phylogenetic tree** can been used to represent the historical relationships of groups of organisms, not similarities
  - a graphical representation of the evolution relations among organisms or taxa. The taxa can be species, populations, individuals or genes.

POWER: PhylOgenetic WEb Repeater - An integrated and user-optimized framework for biomolecular phylogenetic analysis

http://power.nhri.org.tw

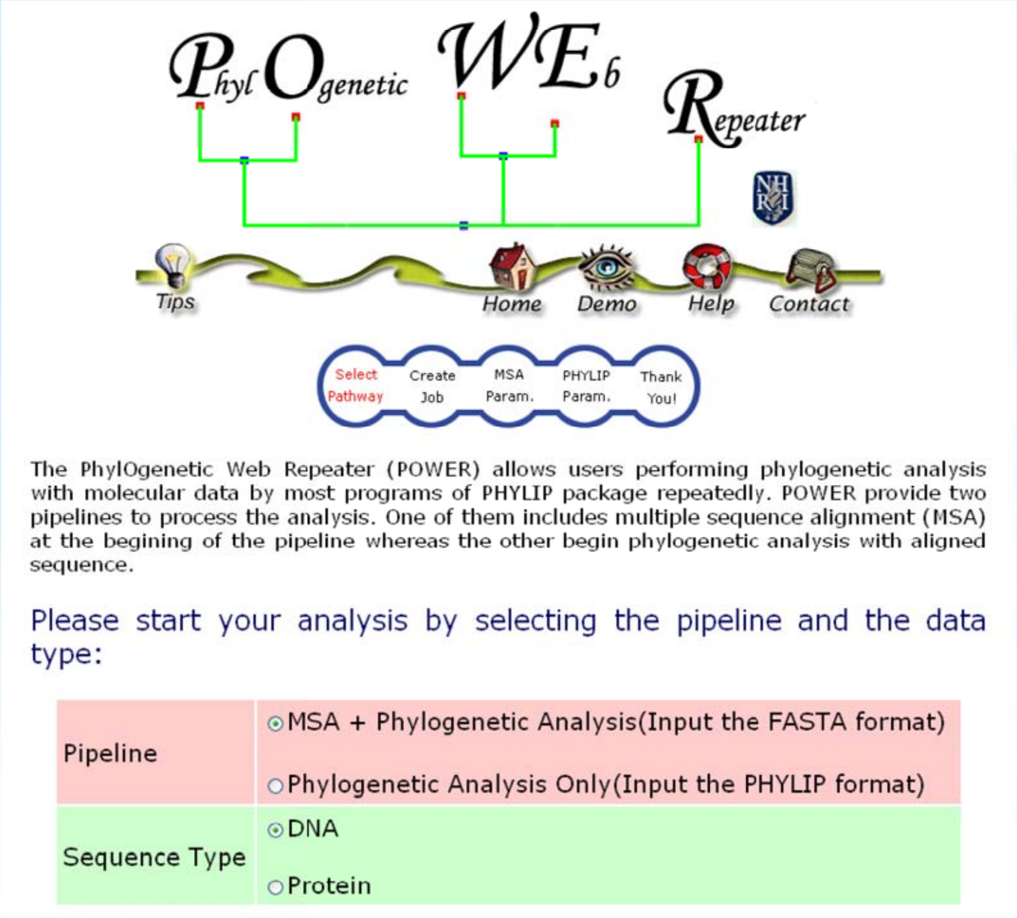


POWER: PhylOgenetic WEb Repeater—an integrated and user-optimized framework for biomolecular phylogenetic analysis

Chung-Yen Lin*, Fan-Kai Lin, Chieh Hua Lin, Li-Wei Lai, Hsiu-Jun Hsu, Shu-Hwa Chen[1] and Chao A. Hsiung

Division of Biostatistics and Bioinformatics, National Health Research Institutes 35 Keyan Road, Zhunan Town, Miaoli County 350, Taiwan [1]Institute of Zoology, Academia Sinica 128 Academia Road Sec. 2, Nankang, Taipei, Taiwan

Nucl. Acids Res. 2005 33: W553-W556



The PhylOgenetic Web Repeater (POWER) allows users performing phylogenetic analysis with molecular data by most programs of PHYLIP package repeatedly. POWER provide two pipelines to process the analysis. One of them includes multiple sequence alignment (MSA) at the begining of the pipeline whereas the other begin phylogenetic analysis with aligned sequence.

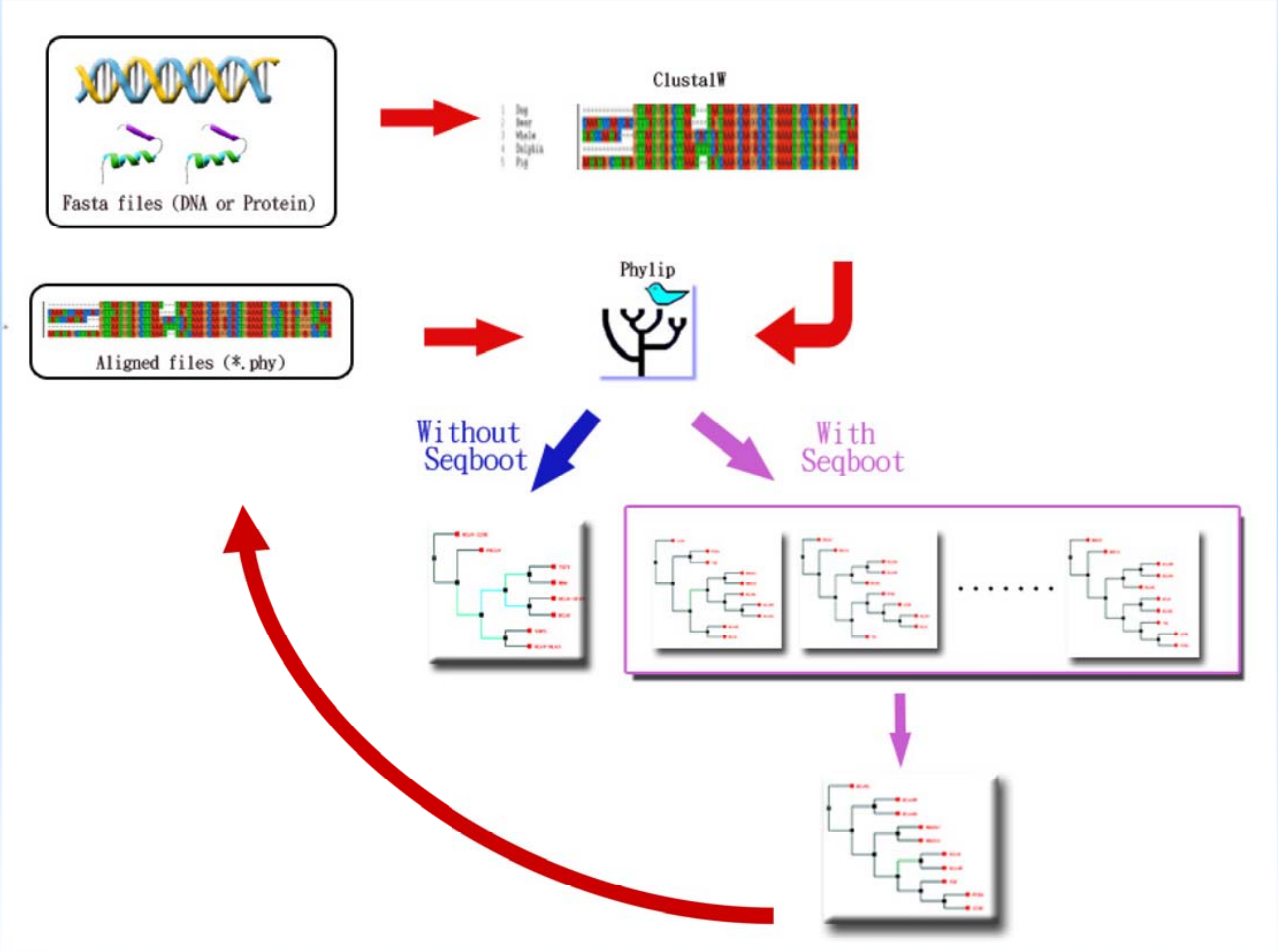
Please start your analysis by selecting the pipeline and the data type:

Pipeline	● MSA + Phylogenetic Analysis(Input the FASTA format) ○ Phylogenetic Analysis Only(Input the PHYLIP format)
Sequence Type	● DNA ○ Protein

Phylogenetic Analysis Steps

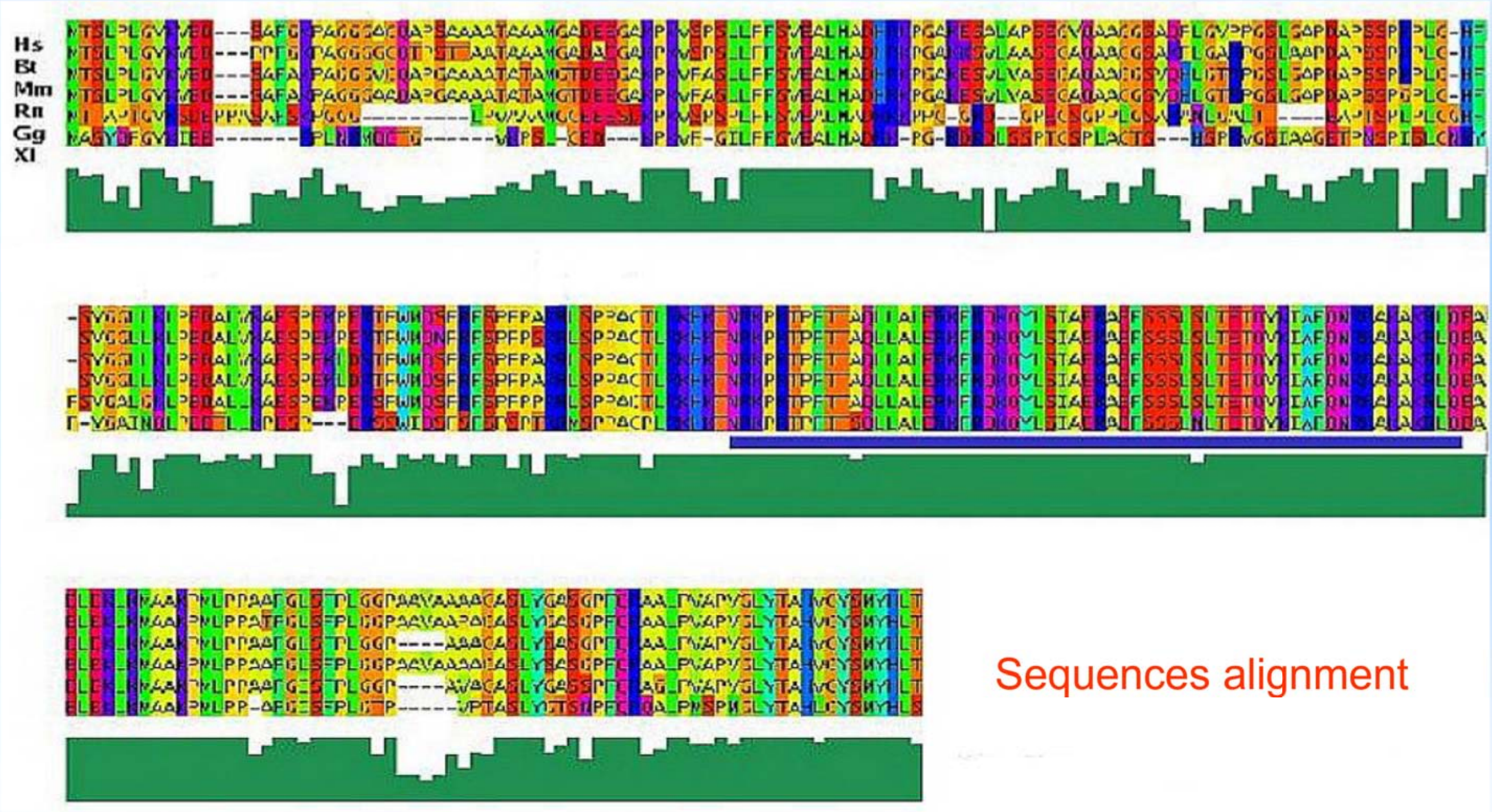
- Sequences alignment
- Build substitution model
- Construct phylogenetic tree
- Evaluate phylogenetic tree

PhylOgenetic Web Repeater (POWER)



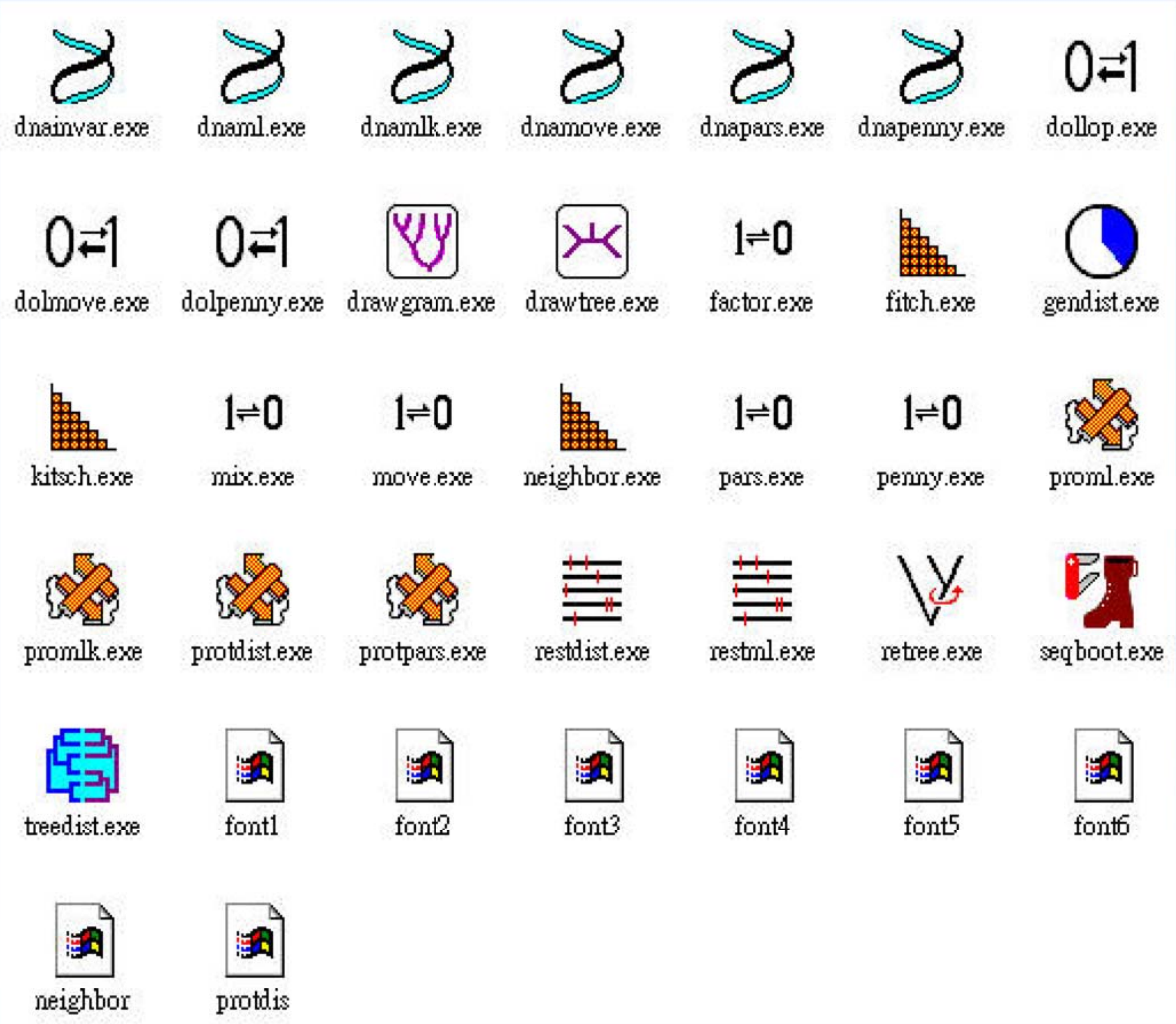
Add/ delete sequences to invoke new job

Schema of POWER

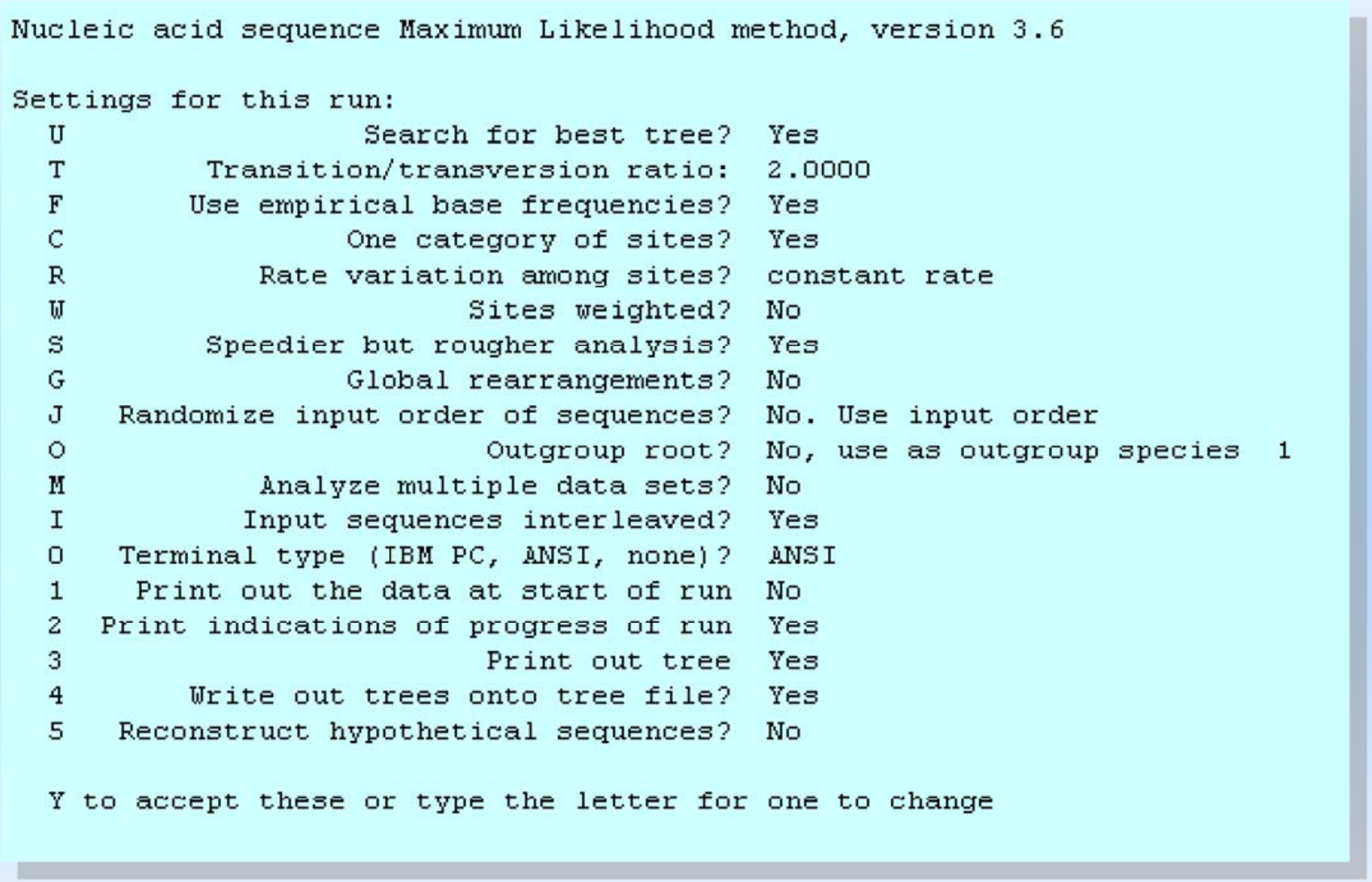


Sequences alignment

Phylip


dnainvar.exe
dnaml.exe
dnamlk.exe
dnamove.exe
dnapars.exe
dnapenny.exe
dollop.exe
dolmove.exe
dolpenny.exe
drawgram.exe
drawtree.exe
factor.exe
fitch.exe
gendist.exe
kitsch.exe
mix.exe
move.exe
neighbor.exe
pars.exe
penny.exe
proml.exe
promlk.exe
protdist.exe
protpars.exe
restdist.exe
restml.exe
retree.exe
seqboot.exe
treedist.exe
font1
font2
font3
font4
font5
font6
neighbor
protdis

Interactive Interface for Phylip

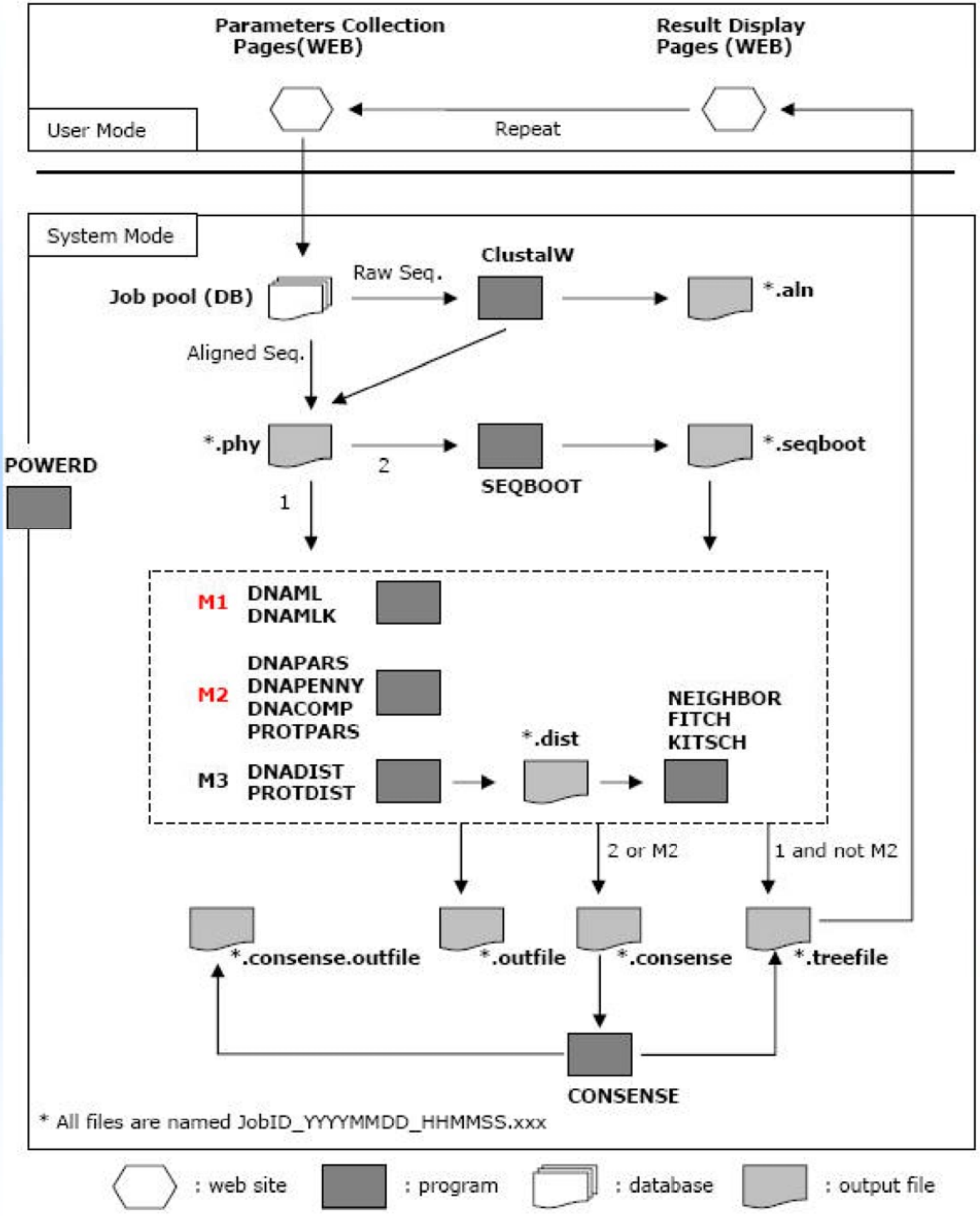


```
Nucleic acid sequence Maximum Likelihood method, version 3.6

Settings for this run:
  U                 Search for best tree?  Yes
  T        Transition/transversion ratio:  2.0000
  F       Use empirical base frequencies?  Yes
  C             One category of sites?  Yes
  R          Rate variation among sites?  constant rate
  W                    Sites weighted?  No
  S        Speedier but rougher analysis?  Yes
  G             Global rearrangements?  No
  J   Randomize input order of sequences?  No. Use input order
  O                     Outgroup root?  No, use as outgroup species  1
  M          Analyze multiple data sets?  No
  I         Input sequences interleaved?  Yes
  0   Terminal type (IBM PC, ANSI, none)?  ANSI
  1    Print out the data at start of run  No
  2  Print indications of progress of run  Yes
  3                     Print out tree  Yes
  4       Write out trees onto tree file?  Yes
  5   Reconstruct hypothetical sequences?  No

  Y to accept these or type the letter for one to change
```

At this stage they do not have a mouse-windows interface for PHYLIP



Parameters Collection Pages(WEB)

Result Display Pages (WEB)

User Mode

Repeat

System Mode

Job pool (DB)

Raw Seq.

ClustalW

*.aln

Aligned Seq.

*.phy

2

SEQBOOT

*.seqboot

POWERD

1

M1 DNAML DNAMLK

M2 DNAPARS DNAPENNY DNACOMP PROTPARS

M3 DNADIST PROTDIST

*.dist

NEIGHBOR FITCH KITSCH

2 or M2

1 and not M2

*.consense.outfile

*.outfile

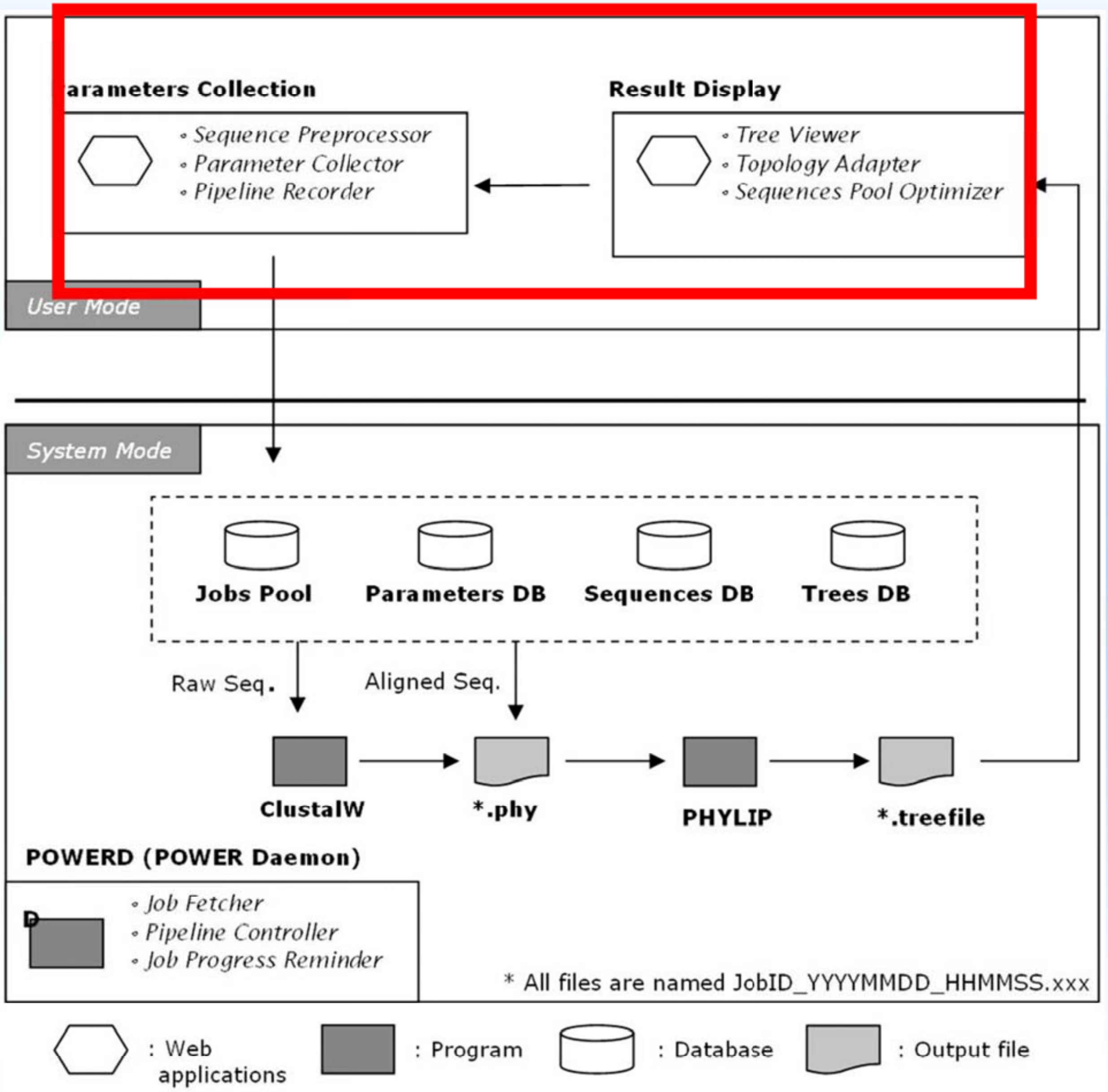
*.consense

*.treefile

CONSENSE

* All files are named JobID_YYYYMMDD_HHMMSS.xxx

: web site : program : database : output file

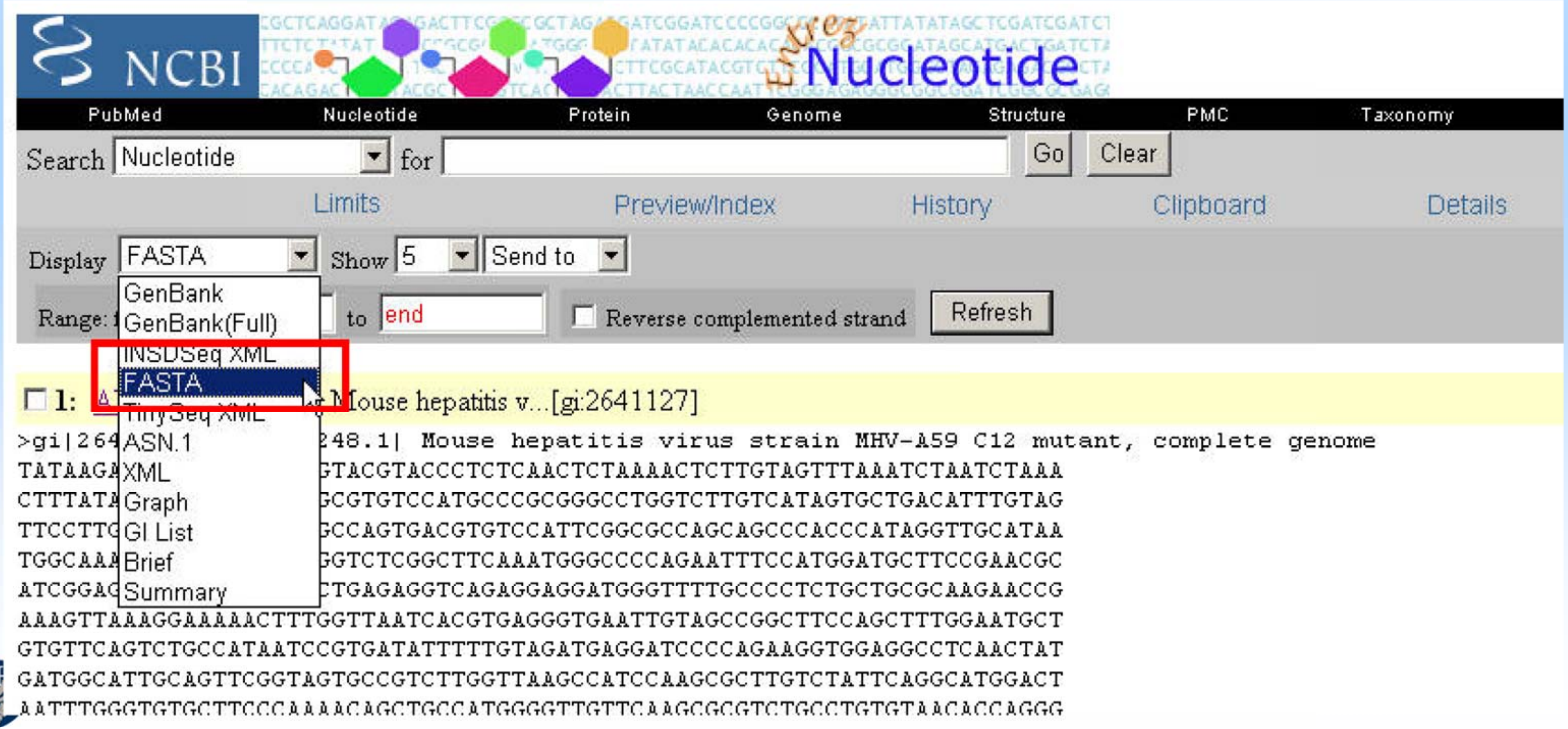

arameters Collection
• Sequence Preprocessor
• Parameter Collector
• Pipeline Recorder
Result Display
• Tree Viewer
• Topology Adapter
• Sequences Pool Optimizer
User Mode
System Mode
Jobs Pool
Parameters DB
Sequences DB
Trees DB
Raw Seq.
Aligned Seq.
ClustalW
*.phy
PHYLIP
*.treefile
POWERD (POWER Daemon)
D
• Job Fetcher
• Pipeline Controller
• Job Progress Reminder
* All files are named JobID_YYYYMMDD_HHMMSS.xxx
: Web applications
: Program
: Database
: Output file

Phylogenetic Analysis Step 1

- Sequences alignment
  - ClustalW, Jalview

To get the sequence

- The resource of sequence database: NCBI, EMBL, DDBJ
- Data format

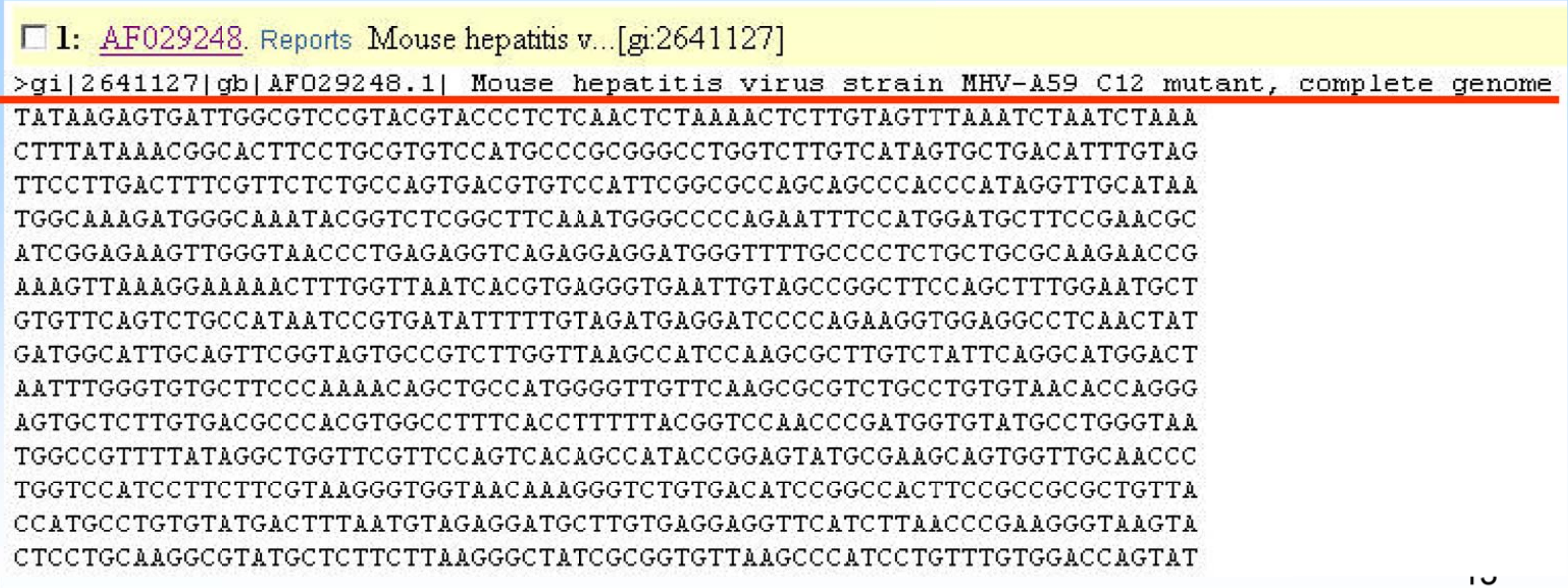


☐ 1: Mouse hepatitis v...[gi:2641127]

```
>gi|264           248.1| Mouse hepatitis virus strain MHV-A59 C12 mutant, complete genome
TATAAGA           GTACGTACCCTCTCAACTCTAAAACTCTTGTAGTTTAAATCTAATCTAAA
CTTTATA           GCGTGTCCATGCCCGCGGGCCTGGTCTTGTCATAGTGCTGACATTTGTAG
TTCCTTG           GCCAGTGACGTGTCCATTCGGCGCCAGCAGCCCACCCATAGGTTGCATAA
TGGCAAA           GGTCTCGGCTTCAAATGGGCCCCAGAATTTCCATGGATGCTTCCGAACGC
ATCGGAG           CTGAGAGGTCAGAGGAGGATGGGTTTTGCCCCTCTGCTGCGCAAGAACCG
AAAGTTAAAGGAAAAACTTTGGTTAATCACGTGAGGGTGAATTGTAGCCGGCTTCCAGCTTTGGAATGCT
GTGTTCAGTCTGCCATAATCCGTGATATTTTTGTAGATGAGGATCCCCAGAAGGTGGAGGCCTCAACTAT
GATGGCATTGCAGTTCGGTAGTGCCGTCTTGGTTAAGCCATCCAAGCGCTTGTCTATTCAGGCATGGACT
AATTTGGGTGTGCTTCCCAAAACAGCTGCCATGGGGTTGTTCAAGCGCGTCTGCCTGTGTAACACCAGGG
```

Input file

- FASTA format: start with a symbol “ › “
- The names of each sequence are not more than 10 characters.



☐ **1:** AF029248. Reports Mouse hepatitis v...[gi:2641127]

```
>gi|2641127|gb|AF029248.1| Mouse hepatitis virus strain MHV-A59 C12 mutant, complete genome
TATAAGAGTGATTGGCGTCCGTACGTACCCTCTCAACTCTAAAACTCTTGTAGTTTAAATCTAATCTAAA
CTTTATAAACGGCACTTCCTGCGTGTCCATGCCCGCGGGCCTGGTCTTGTCATAGTGCTGACATTTGTAG
TTCCTTGACTTTCGTTCTCTGCCAGTGACGTGTCCATTCGGCGCCAGCAGCCCACCCATAGGTTGCATAA
TGGCAAAGATGGGCAAATACGGTCTCGGCTTCAAATGGGCCCCAGAATTTCCATGGATGCTTCCGAACGC
ATCGGAGAAGTTGGGTAACCCTGAGAGGTCAGAGGAGGATGGGTTTTGCCCCTCTGCTGCGCAAGAACCG
AAAGTTAAAGGAAAAACTTTGGTTAATCACGTGAGGGTGAATTGTAGCCGGCTTCCAGCTTTGGAATGCT
GTGTTCAGTCTGCCATAATCCGTGATATTTTTGTAGATGAGGATCCCCAGAAGGTGGAGGCCTCAACTAT
GATGGCATTGCAGTTCGGTAGTGCCGTCTTGGTTAAGCCATCCAAGCGCTTGTCTATTCAGGCATGGACT
AATTTGGGTGTGCTTCCCAAAACAGCTGCCATGGGGTTGTTCAAGCGCGTCTGCCTGTGTAACACCAGGG
AGTGCTCTTGTGACGCCCACGTGGCCTTTCACCTTTTTACGGTCCAACCCGATGGTGTATGCCTGGGTAA
TGGCCGTTTTATAGGCTGGTTCGTTCCAGTCACAGCCATACCGGAGTATGCGAAGCAGTGGTTGCAACCC
TGGTCCATCCTTCTTCGTAAGGGTGGTAACAAAGGGTCTGTGACATCCGGCCACTTCCGCCGCGCTGTTA
CCATGCCTGTGTATGACTTTAATGTAGAGGATGCTTGTGAGGAGGTTCATCTTAACCCGAAGGGTAAGTA
CTCCTGCAAGGCGTATGCTCTTCTTAAGGGCTATCGCGGTGTTAAGCCCATCCTGTTTGTGGACCAGTAT
```

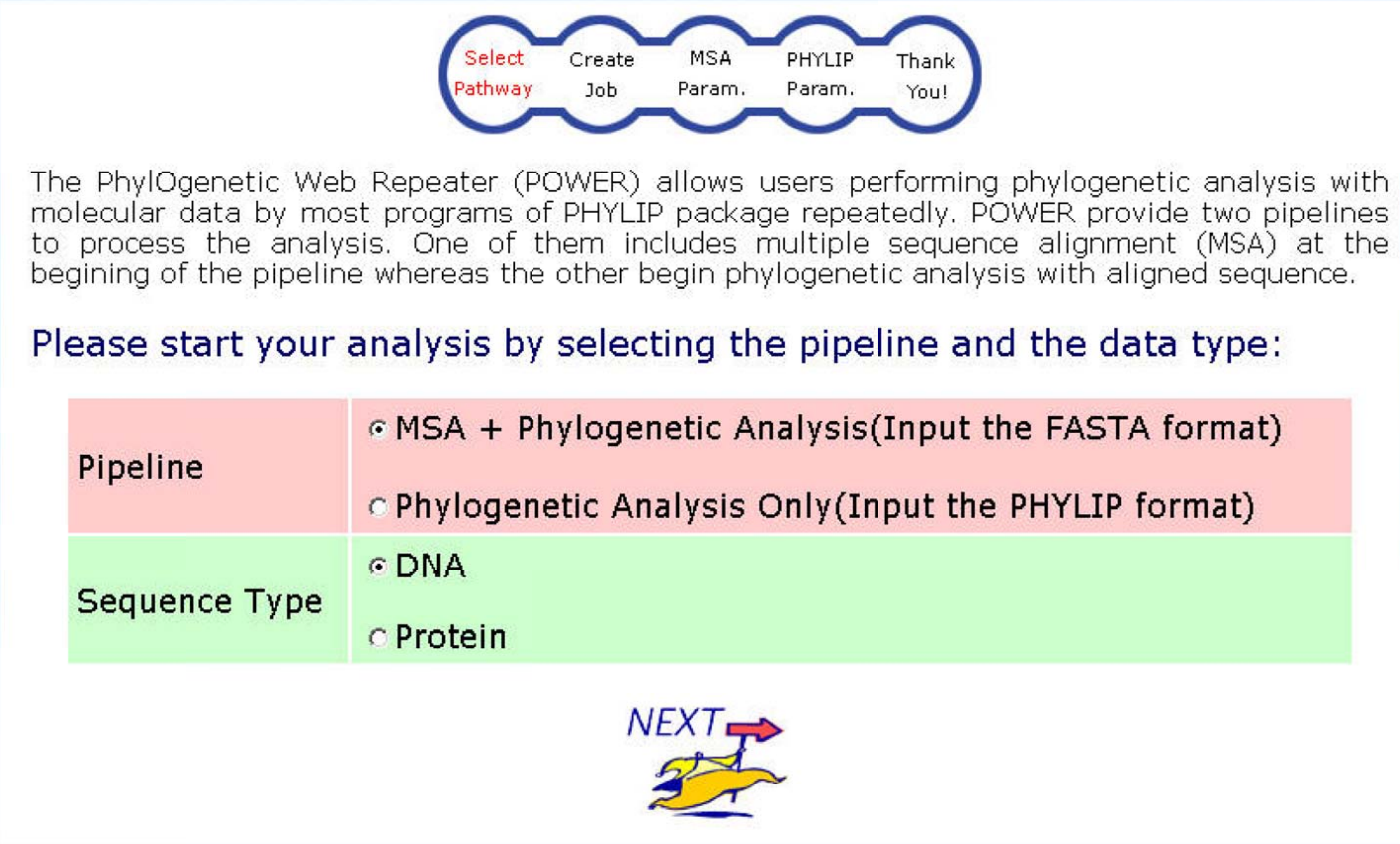
Choose the sequences

- DNA or protein sequence: far related can use protein sequence
  - Nucleotide
  - Amino acid
    - nature selection act on protein.
    - different translation table: species, organelle

$$\text{AUA} \begin{cases} \text{Ile (N)} \\ \text{Met (M)} \end{cases}$$

AUG Met

Start with POWER



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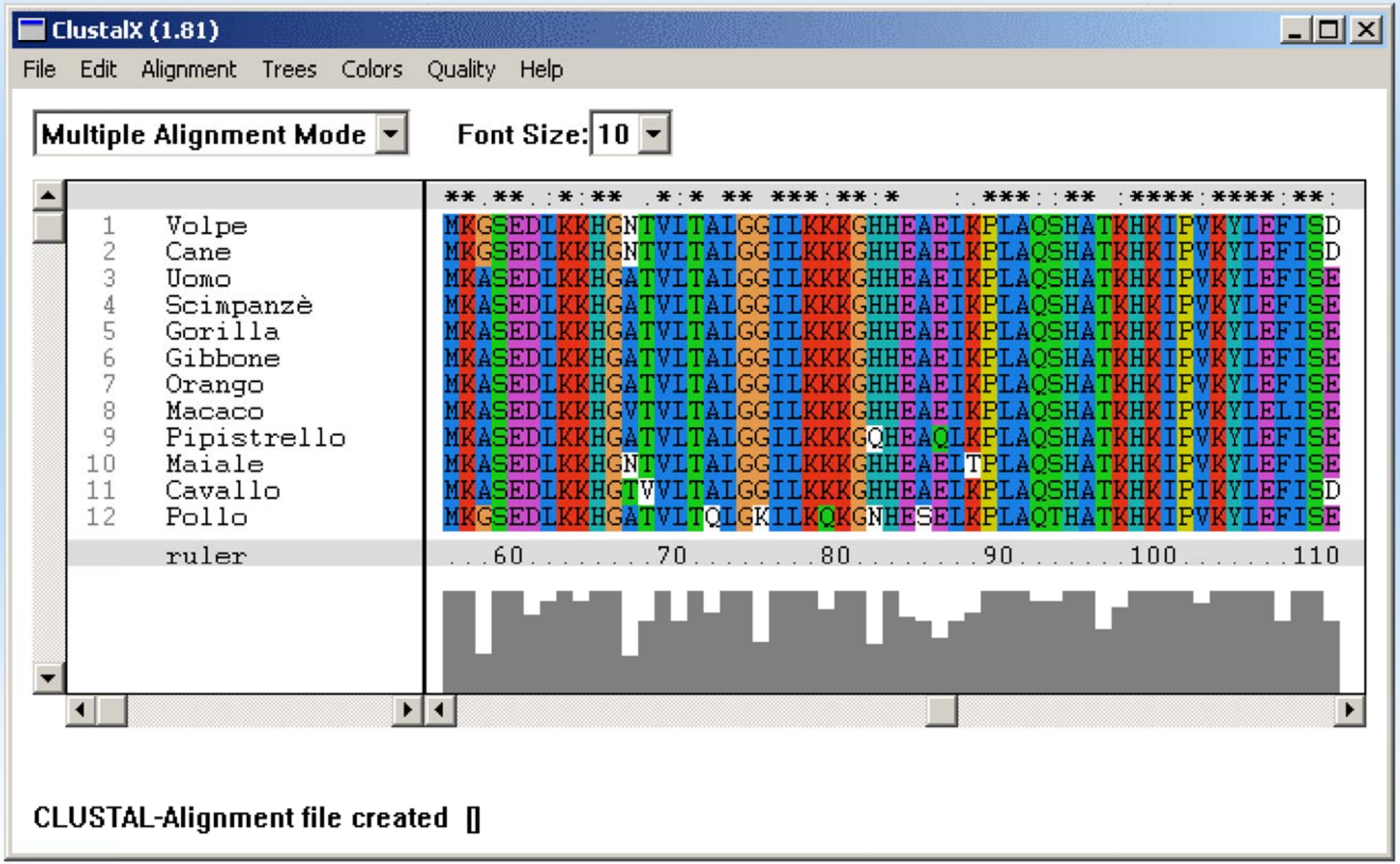
Sequence Type	● DNA ○ Protein

Alignment program-ClustalX (for Windows)



Multiple Alignment Mode

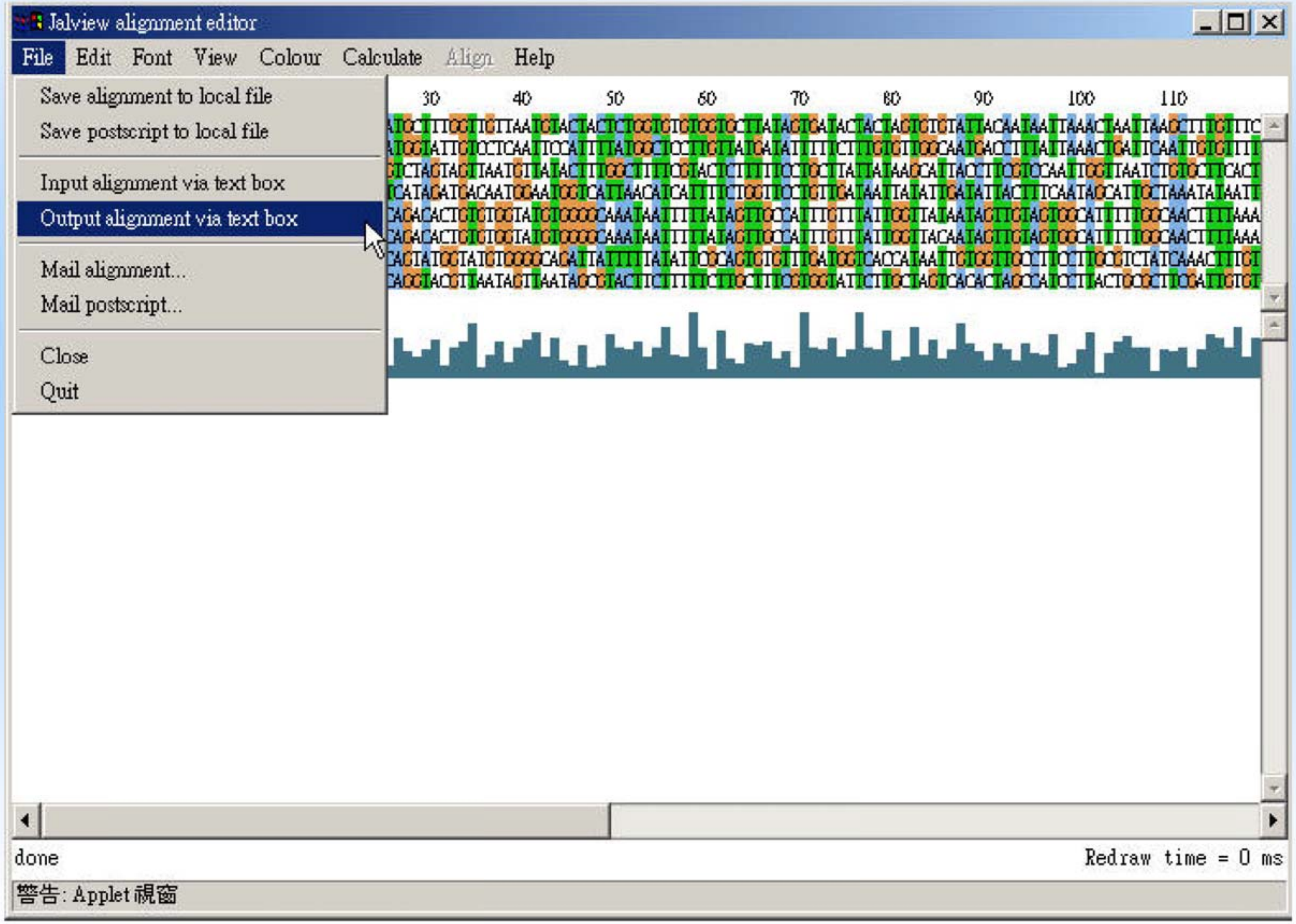
Font Size: 10

```
                      **.**. :*:**  .*:* ** ***:**:*      : .***: :**  :****:****:**:
 1   Volpe            MKGSEDLKKHGNTVLTALGGILKKKGHHEAELKPLAQSHATKHKIPVKYLEFISD
 2   Cane             MKGSEDLKKHGNTVLTALGGILKKKGHHEAELKPLAQSHATKHKIPVKYLEFISD
 3   Uomo             MKASEDLKKHGATVLTALGGILKKKGHHEAEIKPLAQSHATKHKIPVKYLEFISE
 4   Scimpanzè        MKASEDLKKHGATVLTALGGILKKKGHHEAEIKPLAQSHATKHKIPVKYLEFISE
 5   Gorilla          MKASEDLKKHGATVLTALGGILKKKGHHEAEIKPLAQSHATKHKIPVKYLEFISE
 6   Gibbone          MKASEDLKKHGATVLTALGGILKKKGHHEAEIKPLAQSHATKHKIPVKYLEFISE
 7   Orango           MKASEDLKKHGATVLTALGGILKKKGHHEAEIKPLAQSHATKHKIPVKYLEFISE
 8   Macaco           MKASEDLKKHGVTVLTALGGILKKKGHHEAEIKPLAQSHATKHKIPVKYLELISE
 9   Pipistrello      MKASEDLKKHGATVLTALGGILKKKGQHEAQLKPLAQSHATKHKIPVKYLEFISE
10   Maiale           MKASEDLKKHGNTVLTALGGILKKKGHHEAELTPLAQSHATKHKIPVKYLEFISE
11   Cavallo          MKASEDLKKHGTVVLTALGGILKKKGHHEAELKPLAQSHATKHKIPIKYLEFISD
12   Pollo            MKGSEDLKKHGATVLTQLGKILKQKGNHESELKPLAQTHATKHKIPVKYLEFISE
     ruler            ...60.......70.......80.......90......100......110
```

CLUSTAL-Alignment file created []

http://bips.u-strasbg.fr/fr/Documentation/ClustalX/

Jalview for Repeated Job (in POWER)



done

Redraw time = 0 ms

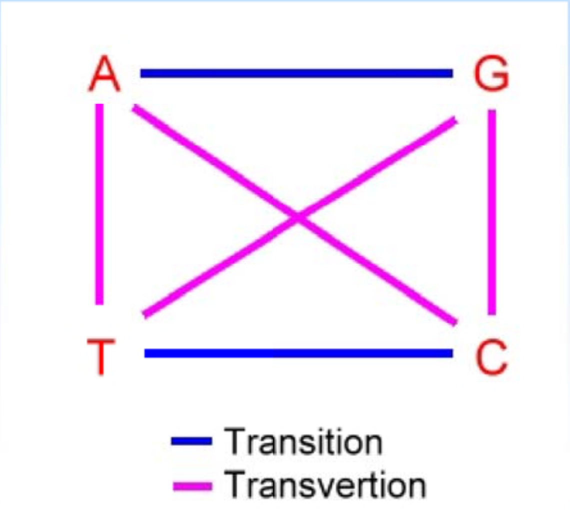
警告: Applet 視窗

Phylogenetic Analysis Step 2

- Sequences alignment
  - ClustalW, Jalview
- Build substitution model
  - Parameters determined

Substitution model

- Transition: purine to purine, pyrimidine to pyrimidine
- Transversion: purine to pyrimidine (and pyrimidine to purine)
- Base frequencies
- Among-site rate variation: Wobble hypothesis
  - Invariable, Variable sites
- Outgroup: to root a tree

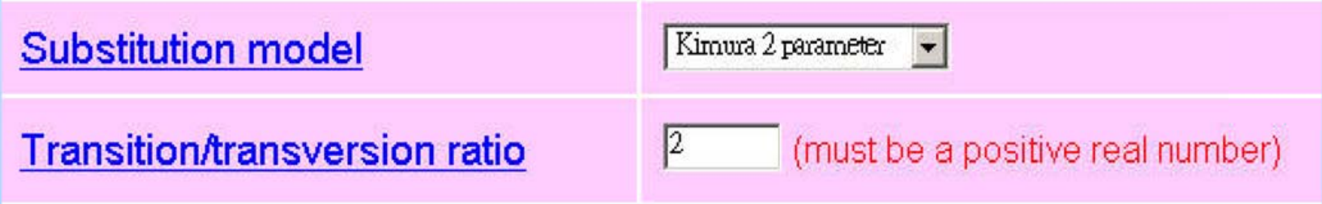


Substitution model of Distance methods

Jukes-Cantor

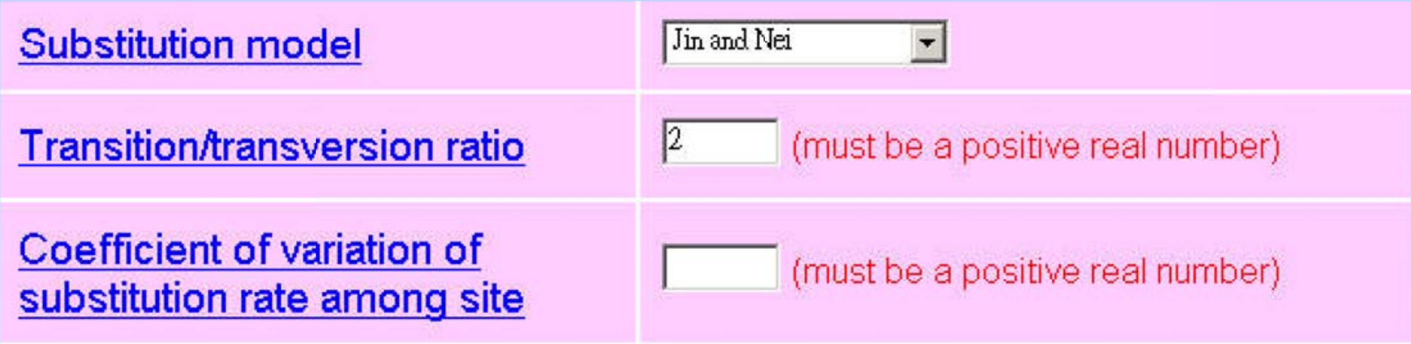
Substitution model	Jukes-Cantor

Kimura 2 parameter



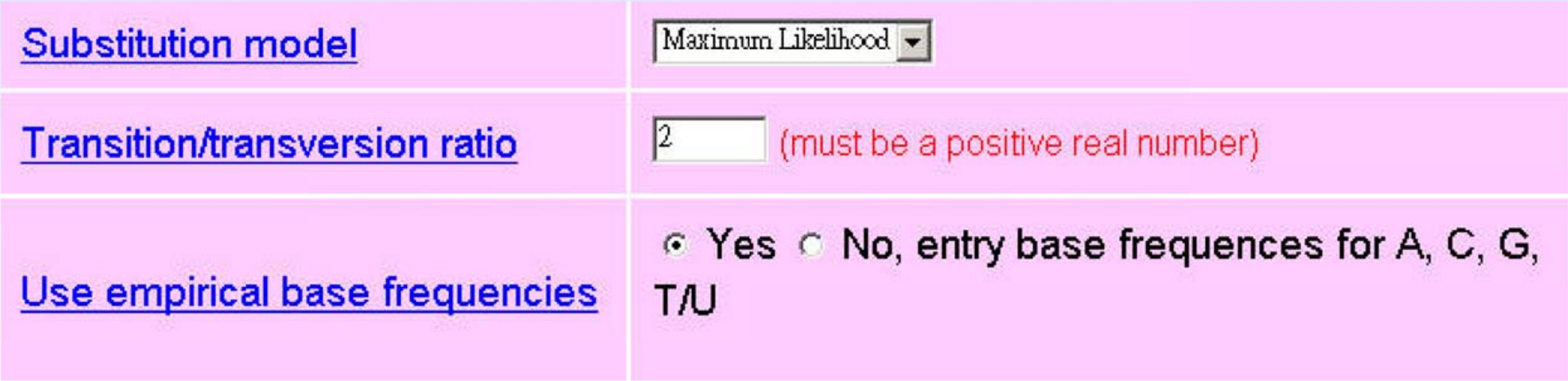
Substitution model	Kimura 2 parameter
Transition/transversion ratio	2 (must be a positive real number)

Jin and Nei



Substitution model	Jin and Nei
Transition/transversion ratio	2 (must be a positive real number)
Coefficient of variation of substitution rate among site	(must be a positive real number)

ML

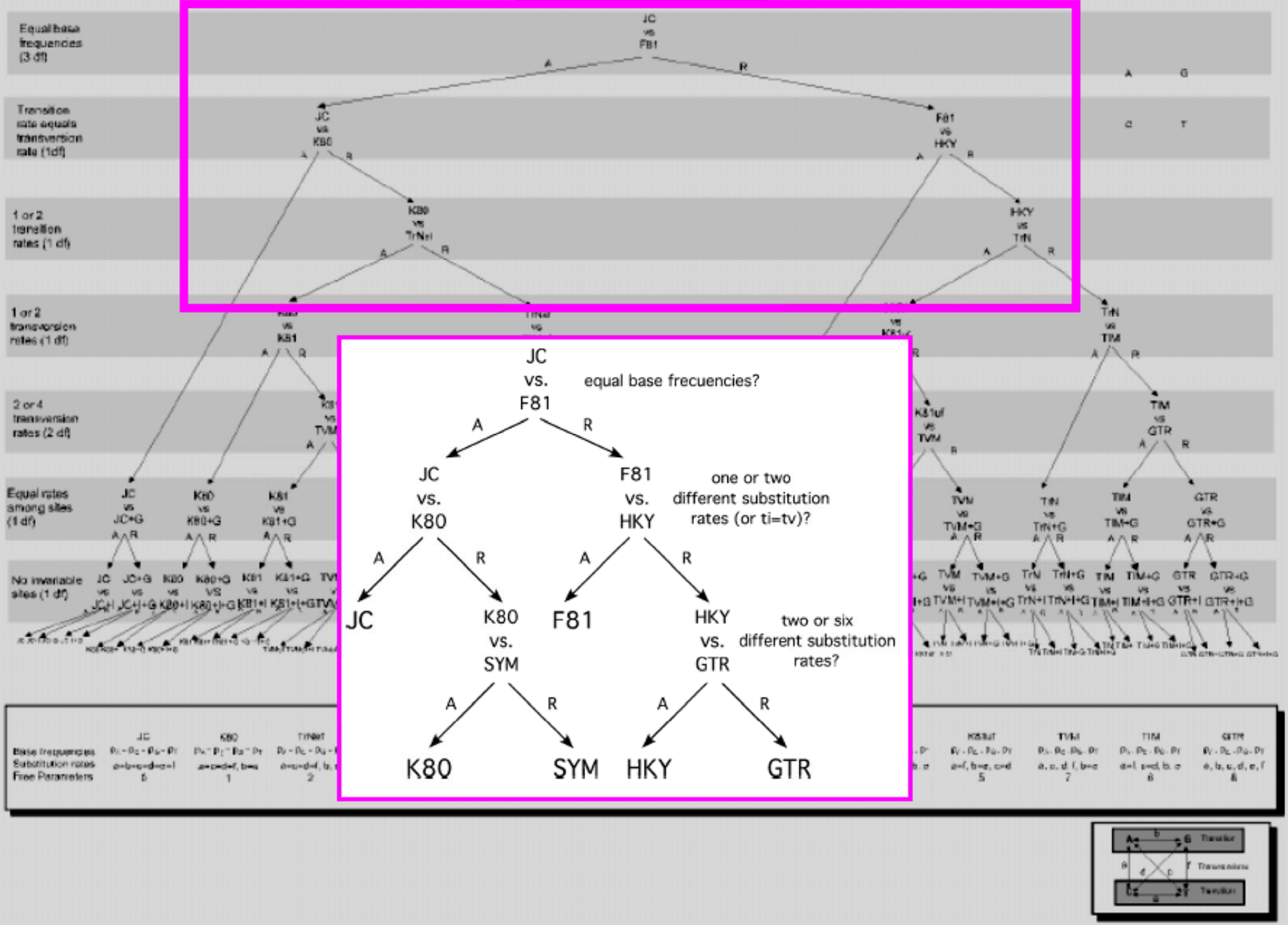


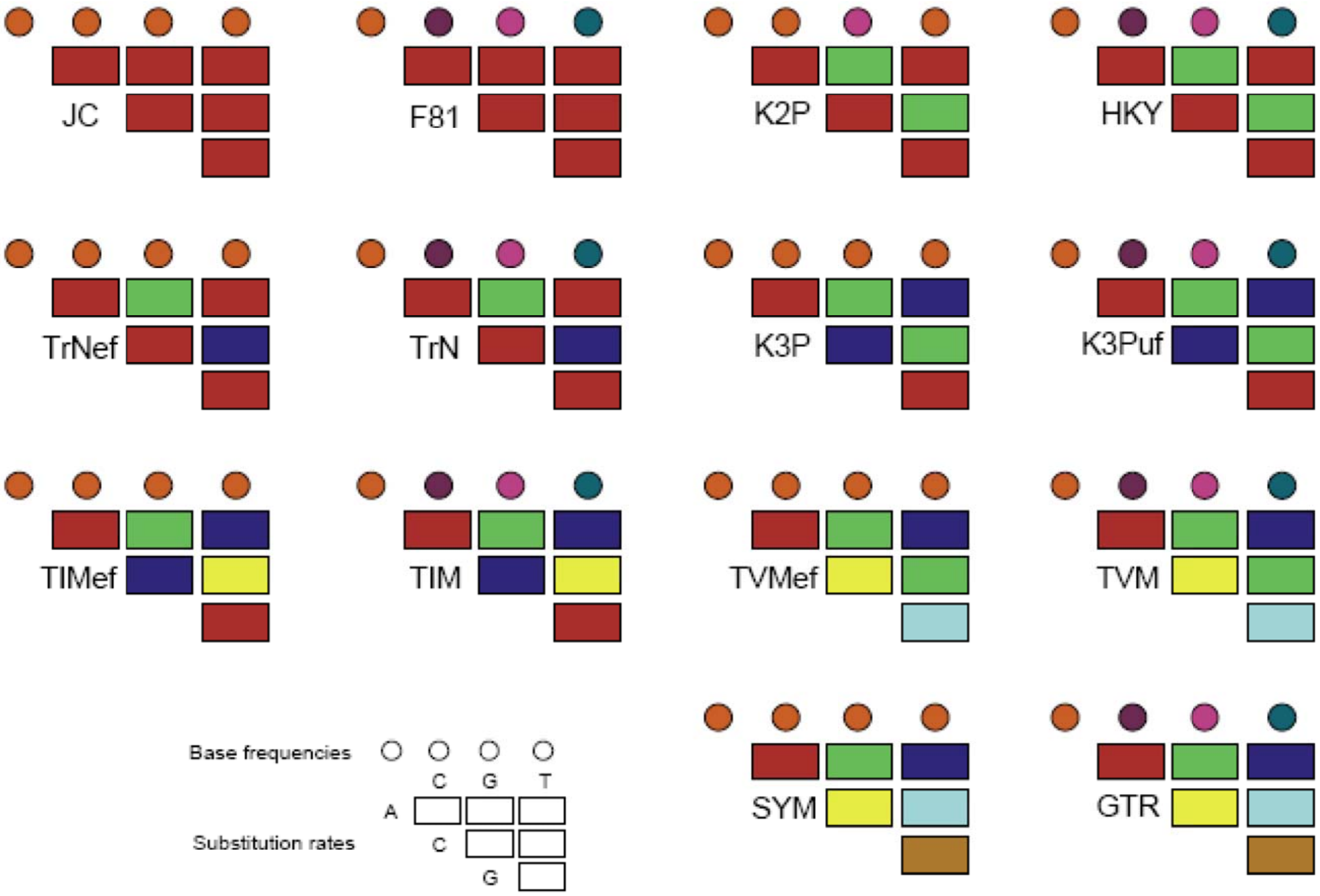
Substitution model	Maximum Likelihood
Transition/transversion ratio	2 (must be a positive real number)
Use empirical base frequencies	● Yes ○ No, entry base frequences for A, C, G, T/U

Choose the suitable model : Model selection

- Which method would be better for our data matrix?
- Using a variety of method to analysis the data matrix
- Software: Modeltest 3.7
  - http://darwin.uvigo.es/software/modeltest.html

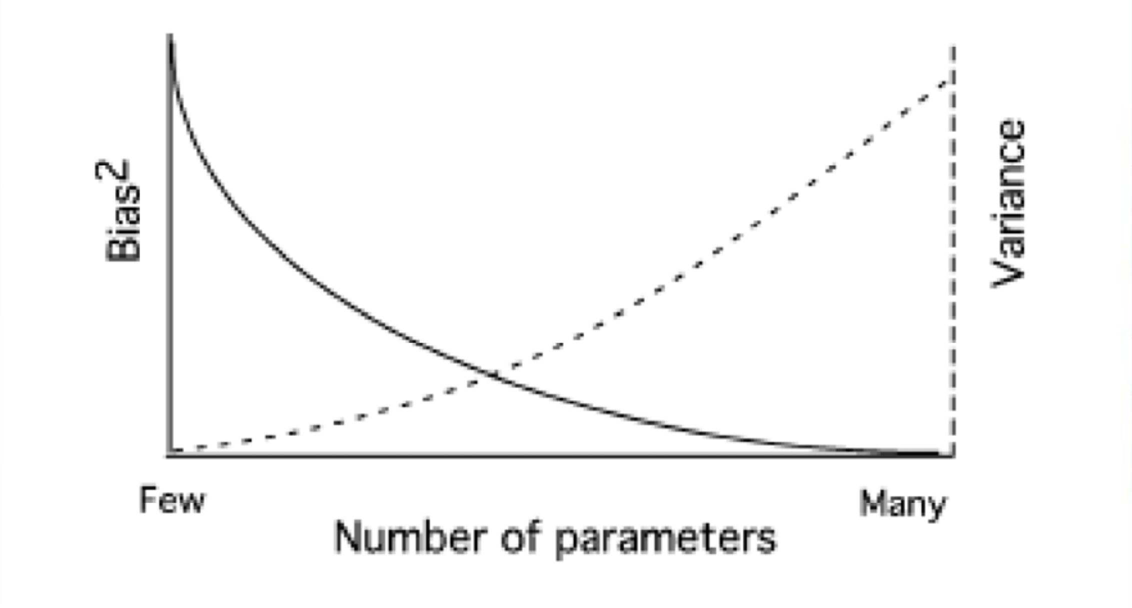
Modeltest 3.0 hierarchy




JC
F81
K2P
HKY
TrNef
TrN
K3P
K3Puf
TIMef
TIM
TVMef
TVM
SYM
GTR
Base frequencies
C
G
T
A
Substitution rates
C
G

Parameter in the model

- More parameters the model uses, more powerful it does?



Bias: distance between the average estimate and truth

Variance: spread of the estimates around the truth

Phylogenetic Analysis Step 3

- Sequences alignment
  - ClustalW, Jalview
- Build substitution model
  - Parameters determined
- Construct phylogenetic tree
  - Quantitatively determine the relationship between input sequences

Phylogenetic Analysis Step 3

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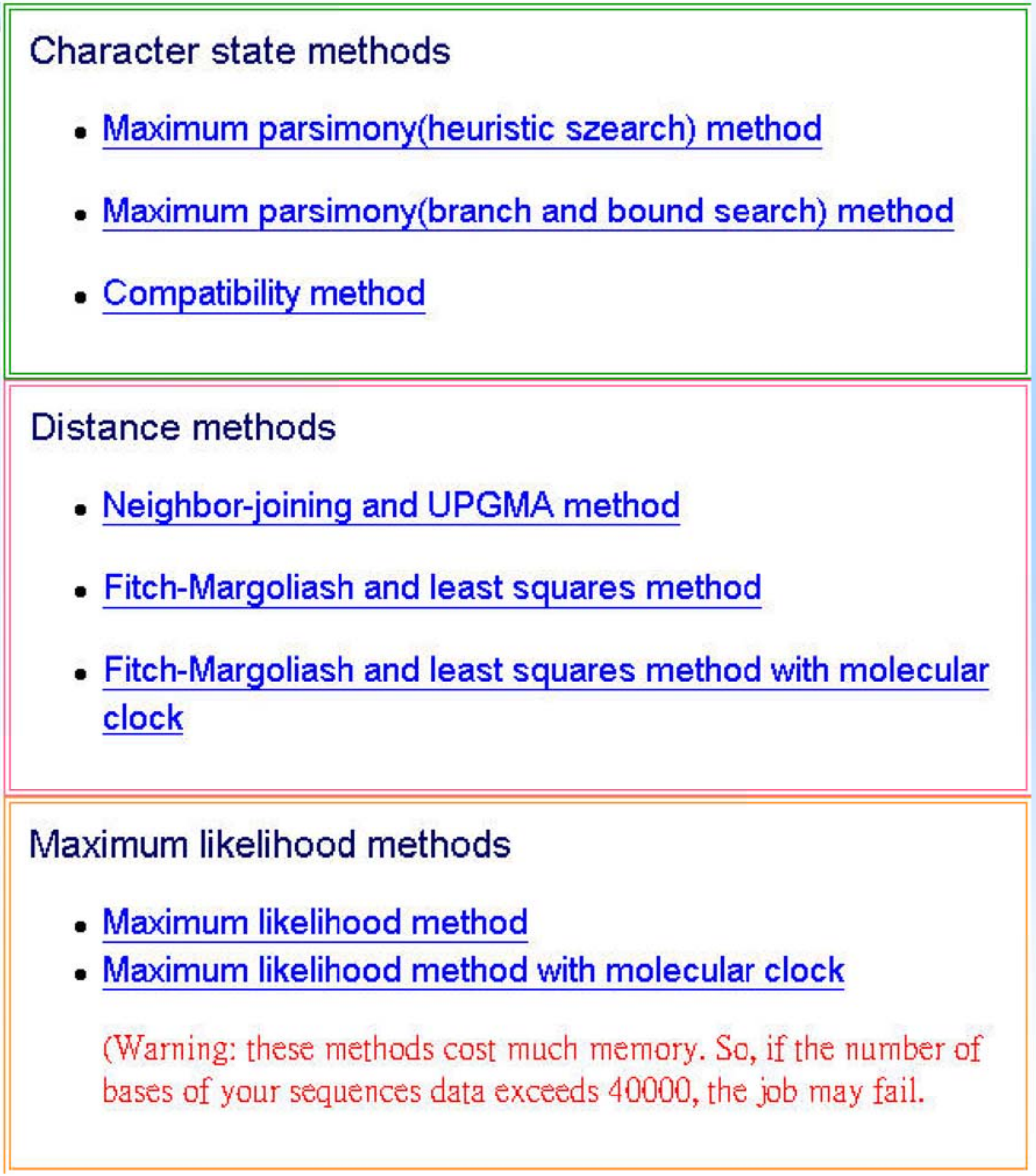
  - Quantitatively determine the relationship between input sequences
- Evaluate phylogenetic tree

Pylogenetic analysis for DNA



Character state methods

- Maximum parsimony(heuristic szearch) method
- Maximum parsimony(branch and bound search) method
- Compatibility method

Distance methods

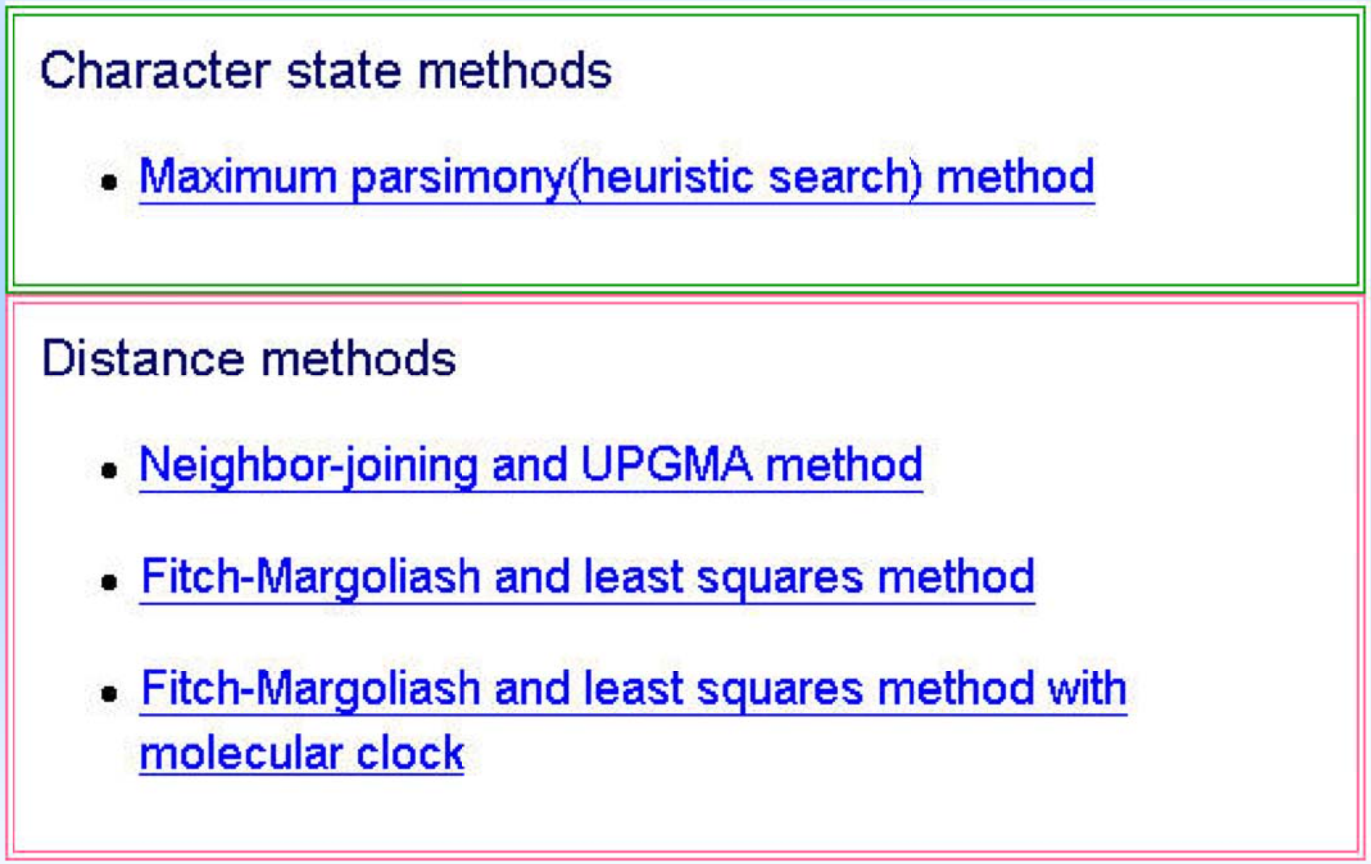
- Neighbor-joining and UPGMA method
- Fitch-Margoliash and least squares method
- Fitch-Margoliash and least squares method with molecular clock

Maximum likelihood methods

- Maximum likelihood method
- Maximum likelihood method with molecular clock

(Warning: these methods cost much memory. So, if the number of bases of your sequences data exceeds 40000, the job may fail.

Pylogenetic analysis for Protein



Character state methods

- Maximum parsimony(heuristic search) method

Distance methods

- Neighbor-joining and UPGMA method
- Fitch-Margoliash and least squares method
- Fitch-Margoliash and least squares method with molecular clock

Phylogenetic Analysis Step 4

- Sequences alignment
  - ClustalW, Jalview
- Build substitution model
  - Parameters determined
- Construct phylogenetic tree
  - Quantitatively determine the relationship between input sequences
- Evaluate phylogenetic tree

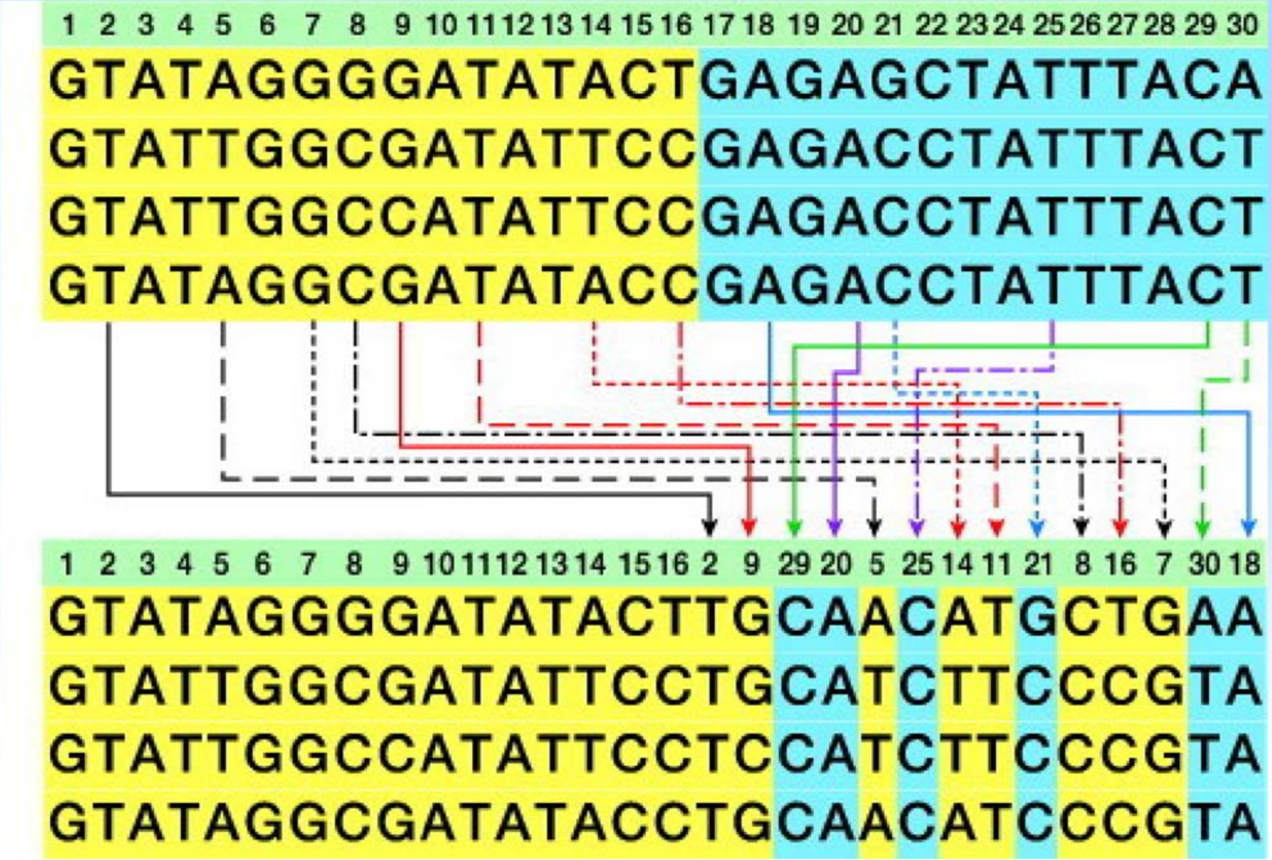
Evaluate reliability of tree

- Sequences resampling
    - Bootstrap
    - Delete-half jackknife
    - Permute species for each character

- Get the same results with different methods

Bootstrapping

- Generate pseudo-sequence alignment randomly
- Compare with original result to get bootstrap value

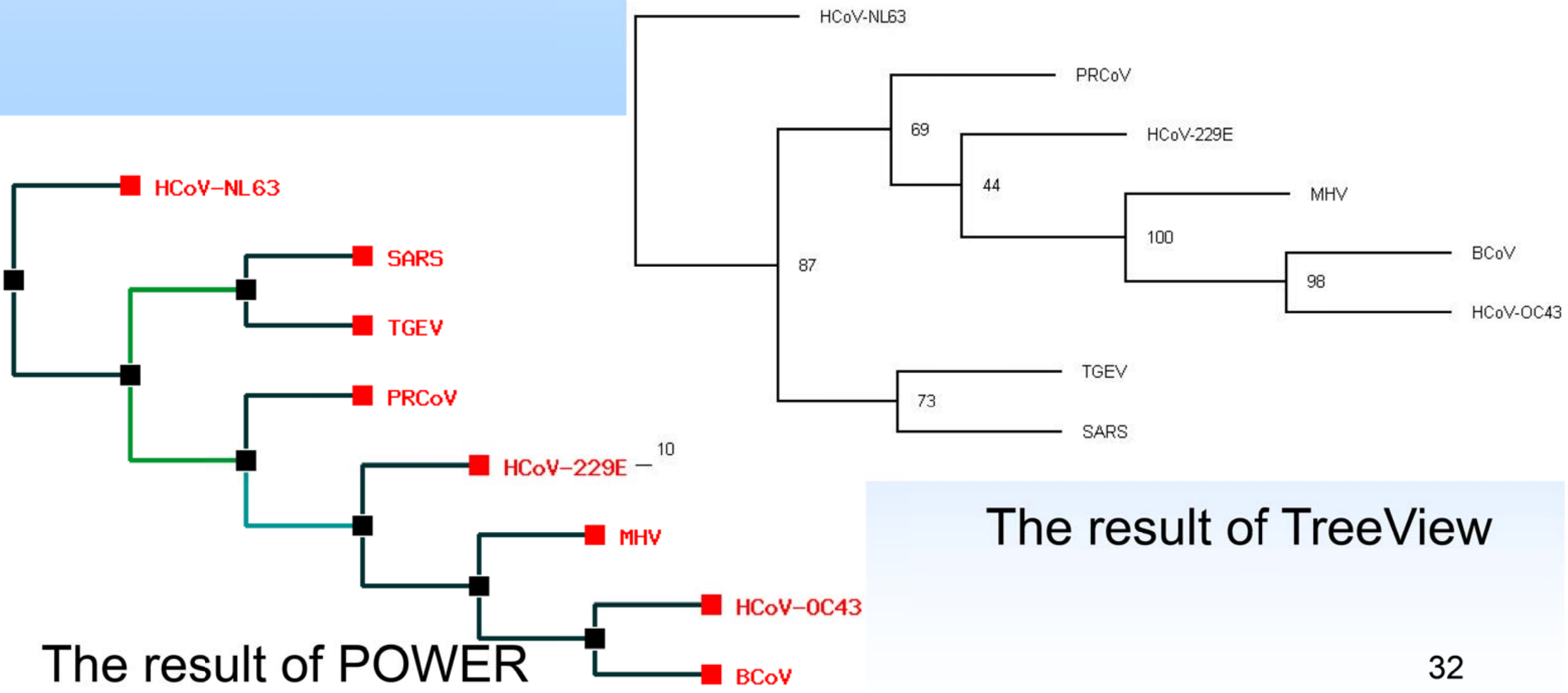


1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30
G	T	A	T	A	G	G	G	G	A	T	A	T	A	C	T	G	A	G	A	G	C	T	A	T	T	T	A	C	A
G	T	A	T	T	G	G	C	G	A	T	A	T	T	C	C	G	A	G	A	C	C	T	A	T	T	T	A	C	T
G	T	A	T	T	G	G	C	C	A	T	A	T	T	C	C	G	A	G	A	C	C	T	A	T	T	T	A	C	T
G	T	A	T	A	G	G	C	G	A	T	A	T	A	C	C	G	A	G	A	C	C	T	A	T	T	T	A	C	T

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	2	9	29	20	5	25	14	11	21	8	16	7	30	18
G	T	A	T	A	G	G	G	G	A	T	A	T	A	C	T	T	G	C	A	A	C	A	T	G	C	T	G	A	A
G	T	A	T	T	G	G	C	G	A	T	A	T	T	C	C	T	G	C	A	T	C	T	T	C	C	C	G	T	A
G	T	A	T	T	G	G	C	C	A	T	A	T	T	C	C	T	C	C	A	T	C	T	T	C	C	C	G	T	A
G	T	A	T	A	G	G	C	G	A	T	A	T	A	C	C	T	G	C	A	A	C	A	T	C	C	C	G	T	A

Construct trees with Bootstraping

Reliability measurement: The probability that the nodes of a given clade are always nodes of that clade

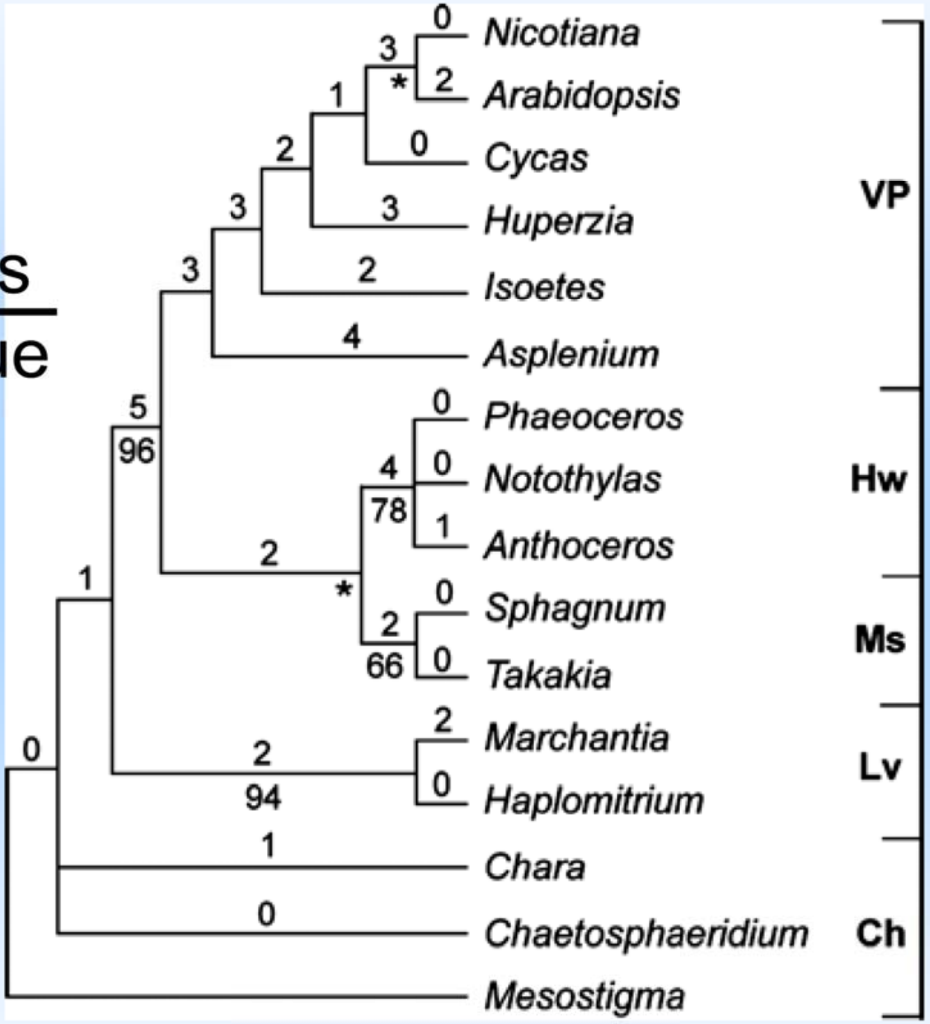


The result of TreeView

The result of POWER

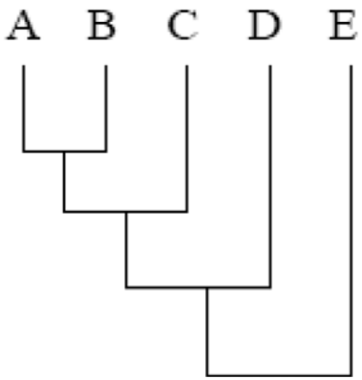
Branch lengths

Bootstrap value

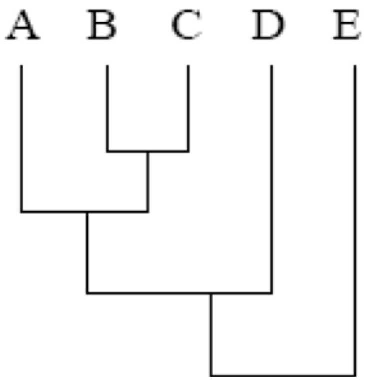


(*Yin-Long Qiua et al. 2006 PNAS*)

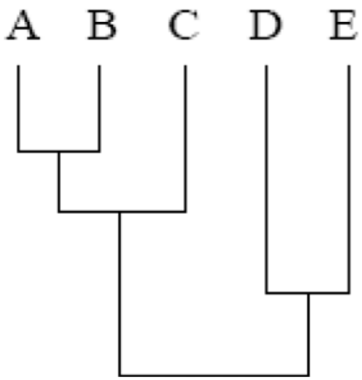
Consensus trees



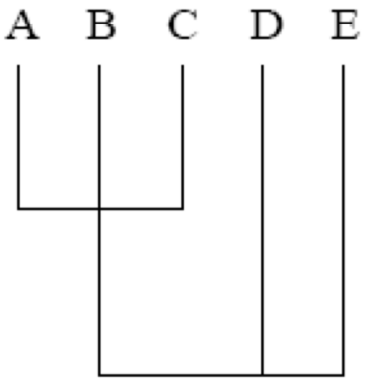
Tree 1



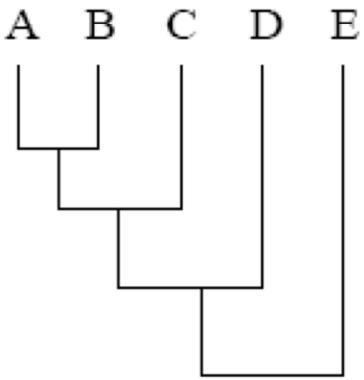
Tree 2



Tree 3



Strict
consensus tree

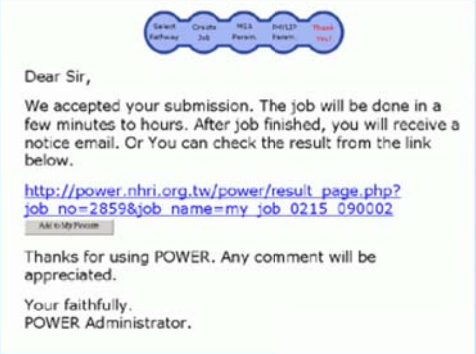


Majority-rule
consensus tree

PhylOgenetic Web Repeater (POWER)

Result and Logs

Online or as bookmark



Dear Sir,

We accepted your submission. The job will be done in a few minutes to hours. After job finished, you will receive a notice email. Or You can check the result from the link below.

http://power.nhri.org.tw/power/result_page.php?job_no=2859&job_name=my_job_0215_090002

Thanks for using POWER. Any comment will be appreciated.

Your faithfully.
POWER Administrator.

Or E-mail notification

Subject [POWER]Job 'comonavirus0720' Finished at 2004-07-20 18:06:35

Dear Sir or Madam:

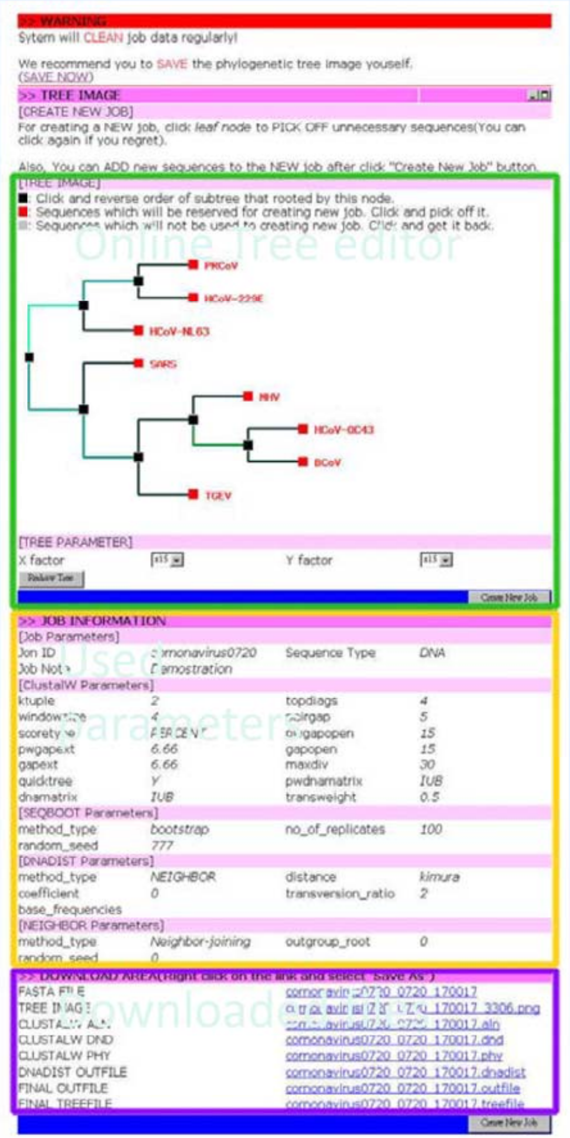
The job 'comonavirus0720' you sent at 2004-07-20 18:00:37 has finished!
The whole process that started at 2004-07-20 18:06:13 and finished at 2004-07-20 18:06:35 cost 00:00:22.
You can check the result from the link below.
Thank you for using POWER.

Your faithfully.
POWER Administrator.

--
Job ID:comonavirus0720
Job Note:
Demonstration
http://211.76.166.77/power/result_page.php?job_no=2041&job_name=comonavirus0720_0720_170017

POWER version 1.0
PHYLIP package version 3.5
ClustalW version 1.82

May the POWER with you.



>> WARNING

Sytem will CLEAN job data regularly!

We recommend you to SAVE the phylogenetic tree image youself.
(SAVE NOW)

>> TREE IMAGE

[CREATE NEW JOB]

For creating a NEW job, click *leaf node* to PICK OFF unnecessary sequences(You can click again if you regret).

Also, You can ADD new sequences to the NEW job after click "Create New Job" button.

[TREE IMAGE]

■: Click and reverse order of subtree that rooted by this node.
■: Sequences which will be reserved for creating new job. Click and pick off it.
▨: Sequences which will not be used to creating new job. Click and get it back.

[TREE PARAMETER]

X factor	x15	Y factor	x15

>> JOB INFORMATION

[Job Parameters]			
Jon ID	comonavirus0720	Sequence Type	DNA
Job Note	Demostration		
[ClustalW Parameters]			
ktuple	2	topdiags	4
windowsize	4	pairgap	5
scoretype	PERCENT	pwgapopen	15
pwgapext	6.66	gapopen	15
gapext	6.66	maxdiv	30
quicktree	Y	pwdnamatrix	IUB
dnamatrix	IUB	transweight	0.5
[SEQBOOT Parameters]			
method_type	bootstrap	no_of_replicates	100
random_seed	777		
[DNADIST Parameters]			
method_type	NEIGHBOR	distance	kimura
coefficient	0	transversion_ratio	2
base_frequencies			
[NEIGHBOR Parameters]			
method_type	Neighbor-joining	outgroup_root	0
random_seed	0		

>> DOWNLOAD AREA(Right click on the link and select "Save As")

FASTA FILE	comonavirus0720_0720_170017
TREE IMAGE	comonavirus0720_0720_170017_3306.png
CLUSTALW ALN	comonavirus0720_0720_170017.aln
CLUSTALW DND	comonavirus0720_0720_170017.dnd
CLUSTALW PHY	comonavirus0720_0720_170017.phy
DNADIST OUTFILE	comonavirus0720_0720_170017.dnadist
FINAL OUTFILE	comonavirus0720_0720_170017.outfile
FINAL TREEFILE	comonavirus0720_0720_170017.treefile

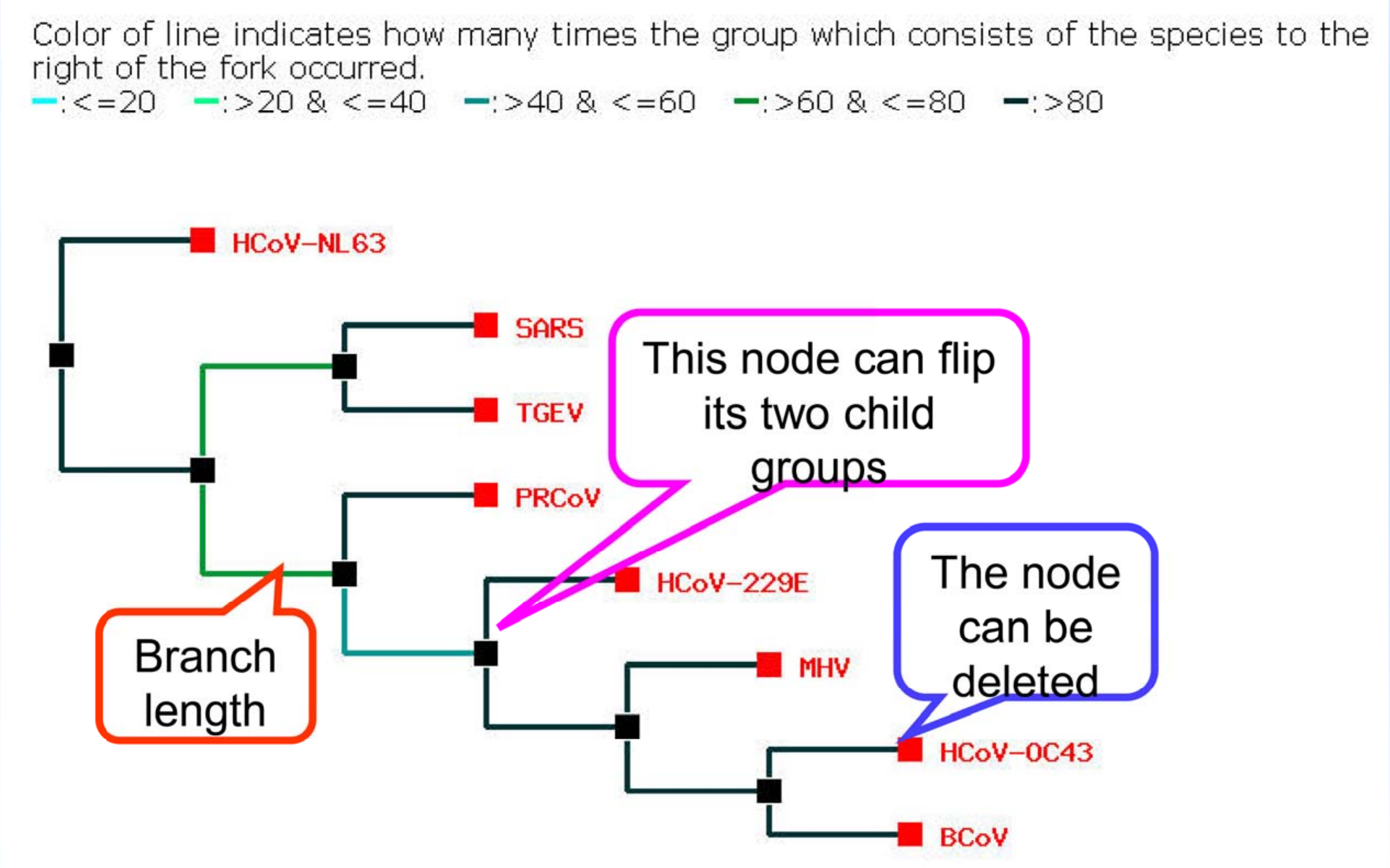
Your data type is **DNA Sequences.**

Please input your data and other related information.

Job ID*	my_job (string with character 0~9 a~z A~Z _ -)
Input sequences in FASTA format* Example Note: - Length of sequence ID should be less then 10! - Only 'A~Z', 'a~z', '0~9', '_', '-' are valid for Sequence ID!	>HCoV-229E …
Or load it from disk	
Job Note	
E-Mail If you are submitting a long job and would like to be informed by email when it finishes, please enter your email address in the space below	(please use ',' to separate multiple email addresses)

Re-perform the process
by items added or deleted

The Result of POWER



Color of line indicates how many times the group which consists of the species to the right of the fork occurred.

—:<=20 —:>20 & <=40 —:>40 & <=60 —:>60 & <=80 —:>80

This node can flip its two child groups

The node can be deleted

Branch length

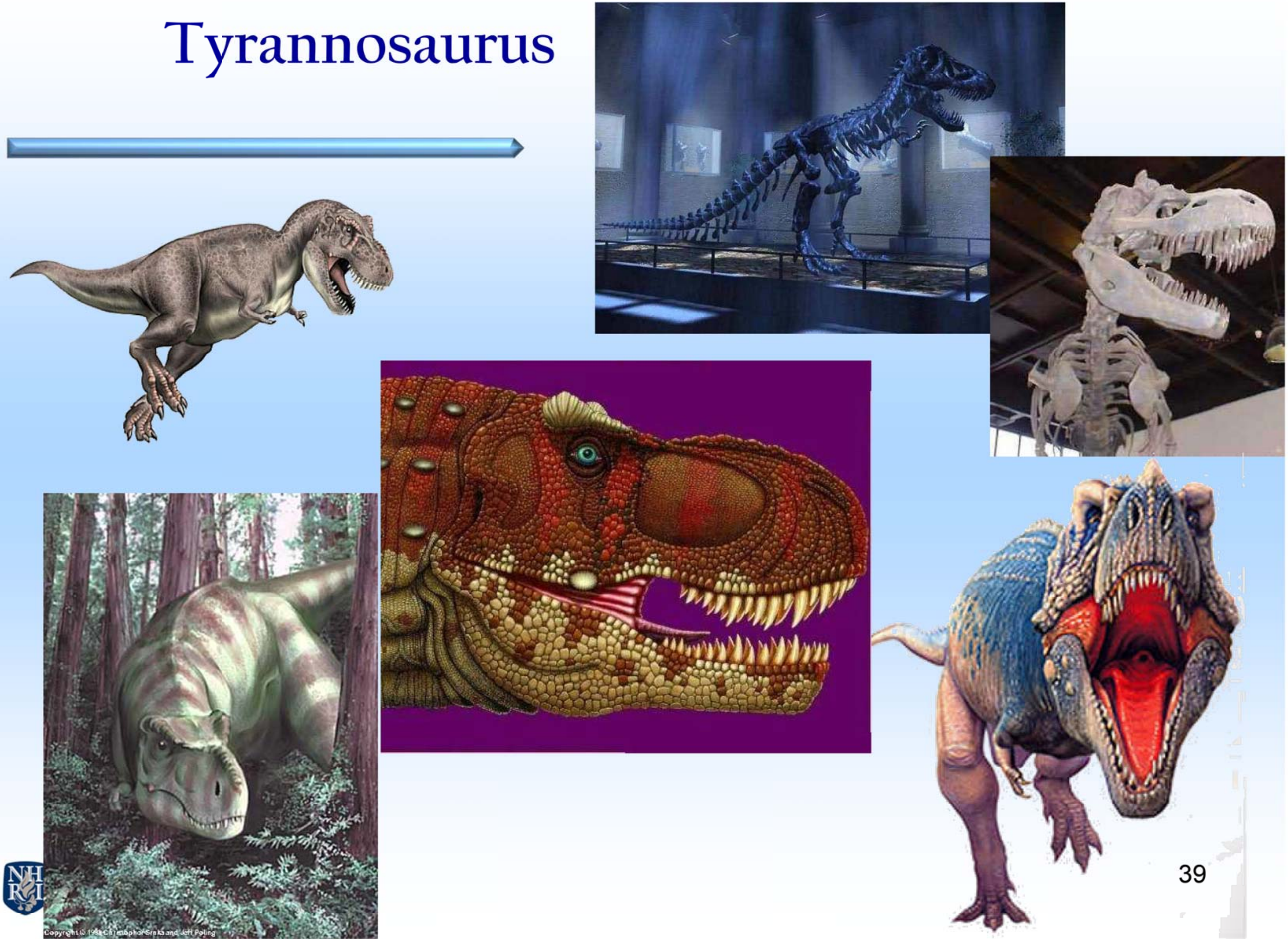
The files can downloaded

- Original fasta file
- .phy (generated by clustalw)
- Tree Image
- outfile(generated by phylip programs)
  - Consensus file if bootstrap had be made
- treefile

Reconstruct tree completely??

- Species evolution is a history, which can't be reconstructed fully
- All models are questionable but some are useful
- Narrow down difference between gene tree and species tree

Tyrannosaurus



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Reconstruct tree completely??

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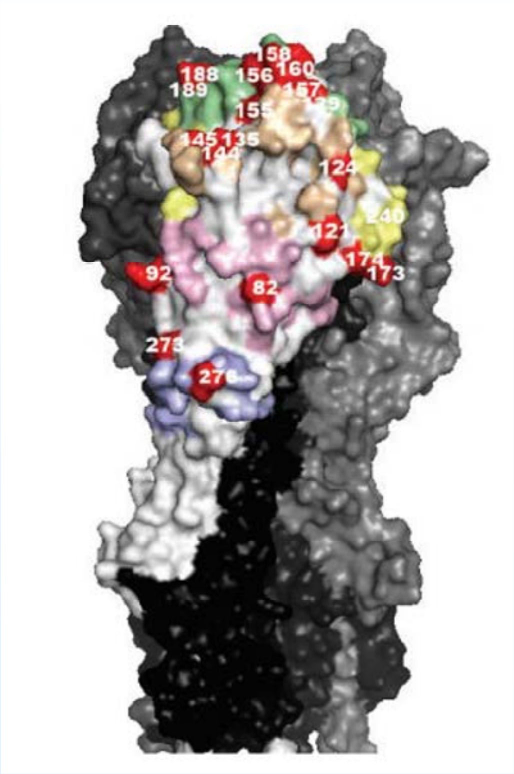

BIOINFORMATICS **ORIGINAL PAPER**

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doi:10.1093/bioinformatics/btm638

Sequence analysis

Bioinformatics models for predicting antigenic variants of influenza A/H3N2 virus

Yu-Chieh Liao[1,†], Min-Shi Lee[2,†], Chin-Yu Ko[1] and Chao A. Hsiung[1,*]

[1]Division of Biostatistics and Bioinformatics and [2]Vaccine R&D Center, National Health Research Institutes, Zhunan 350, Taiwan

Division of Biostatistics and Bioinformatics,
Institute of Population Health Sciences

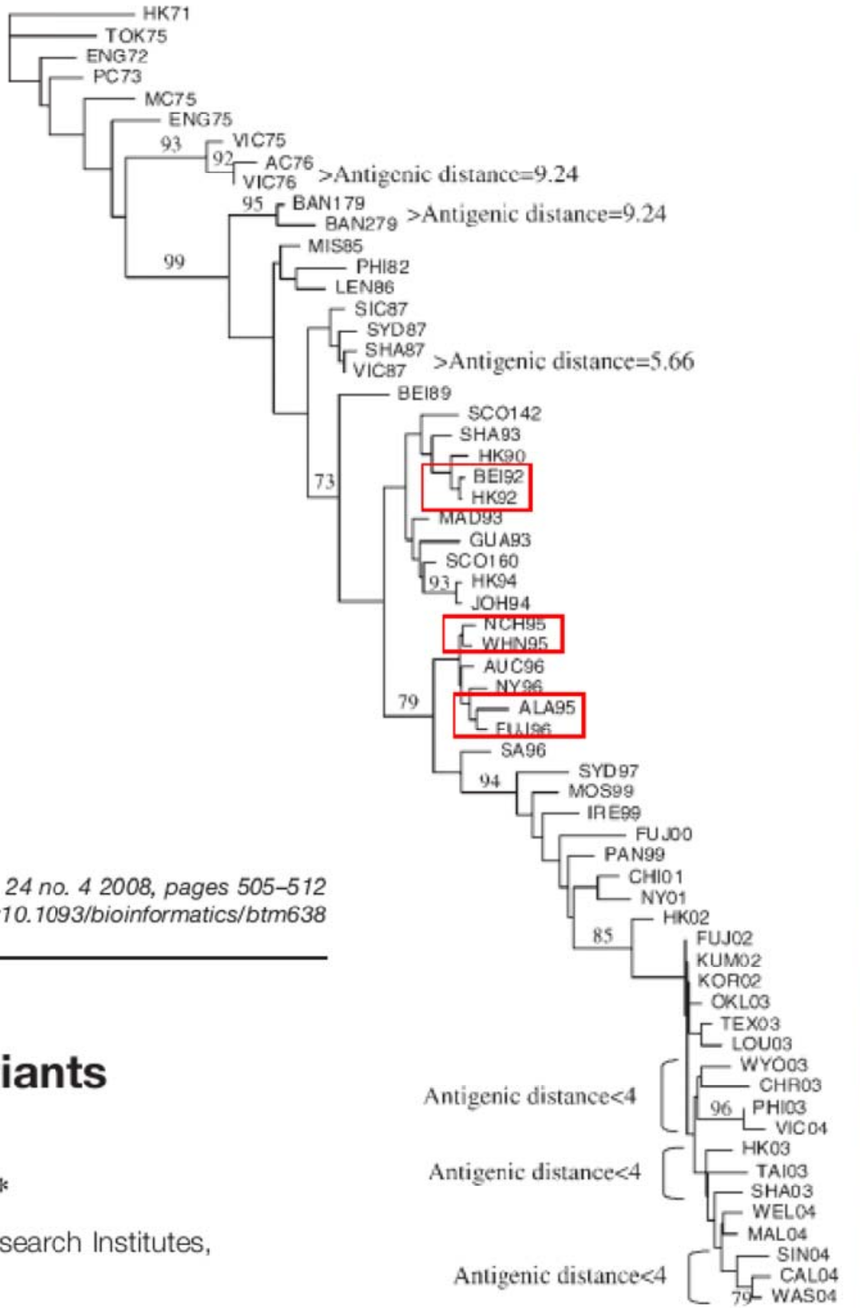
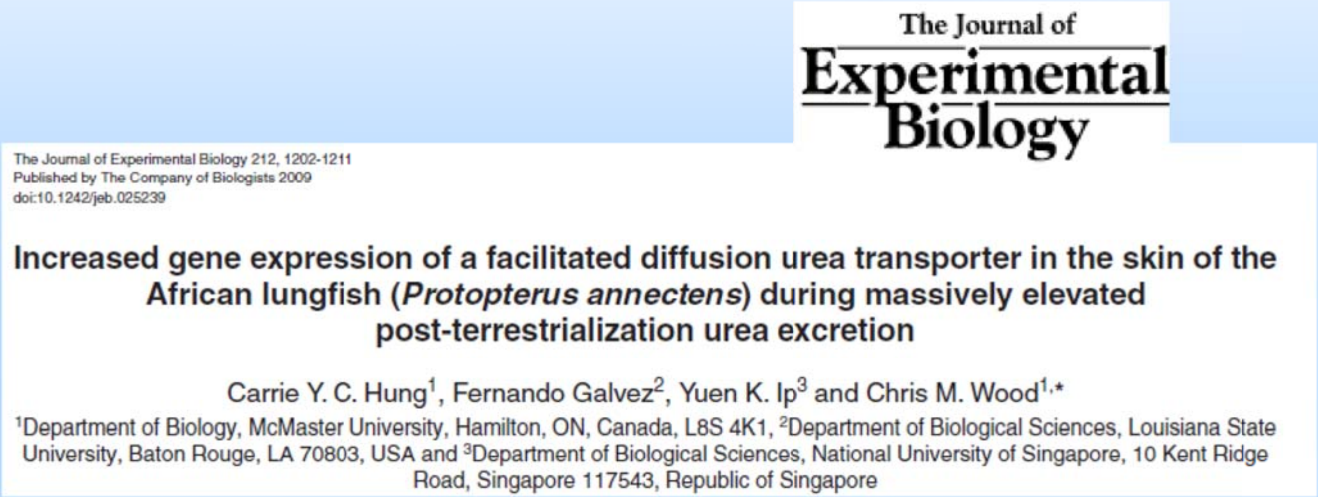


Fig. 3. Neighbor-joining phylogenetic tree of the 62 influenza H3N2 viruses (HA1 protein with 329 amino acid residues). The full names of viruses are in the Supplementary Data. Numbers at branch nodes refer to the percentage of 1000 bootstrap repetitions, and only those occurring at a percentage >70% are shown.

NJ with 1000 Replicates in POWER



The Journal of Experimental Biology

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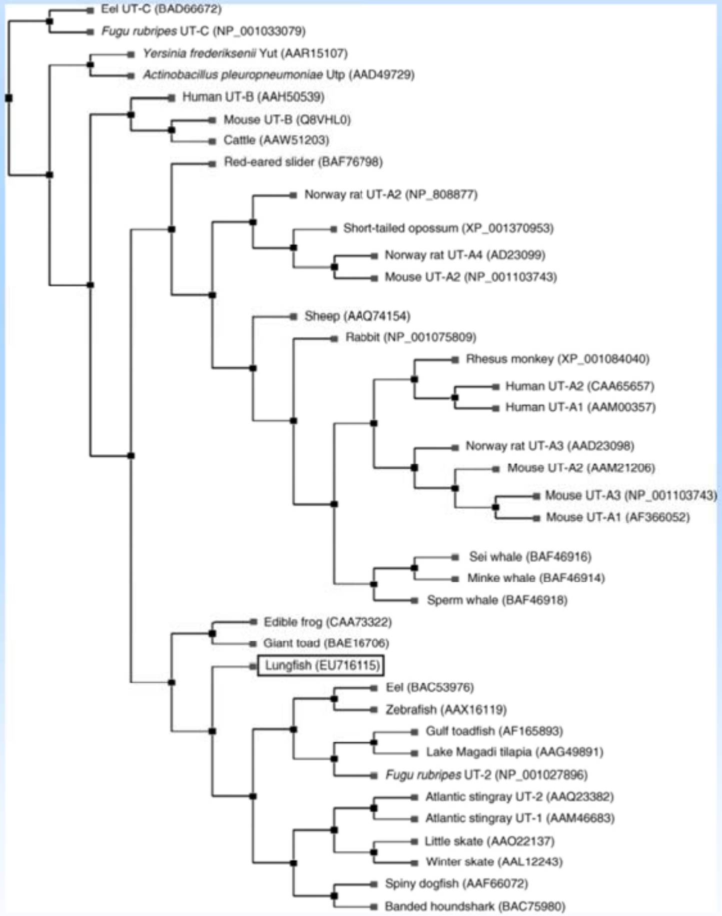
doi:10.1242/jeb.025239

Increased gene expression of a facilitated diffusion urea transporter in the skin of the African lungfish (*Protopterus annectens*) during massively elevated post-terrestrialization urea excretion

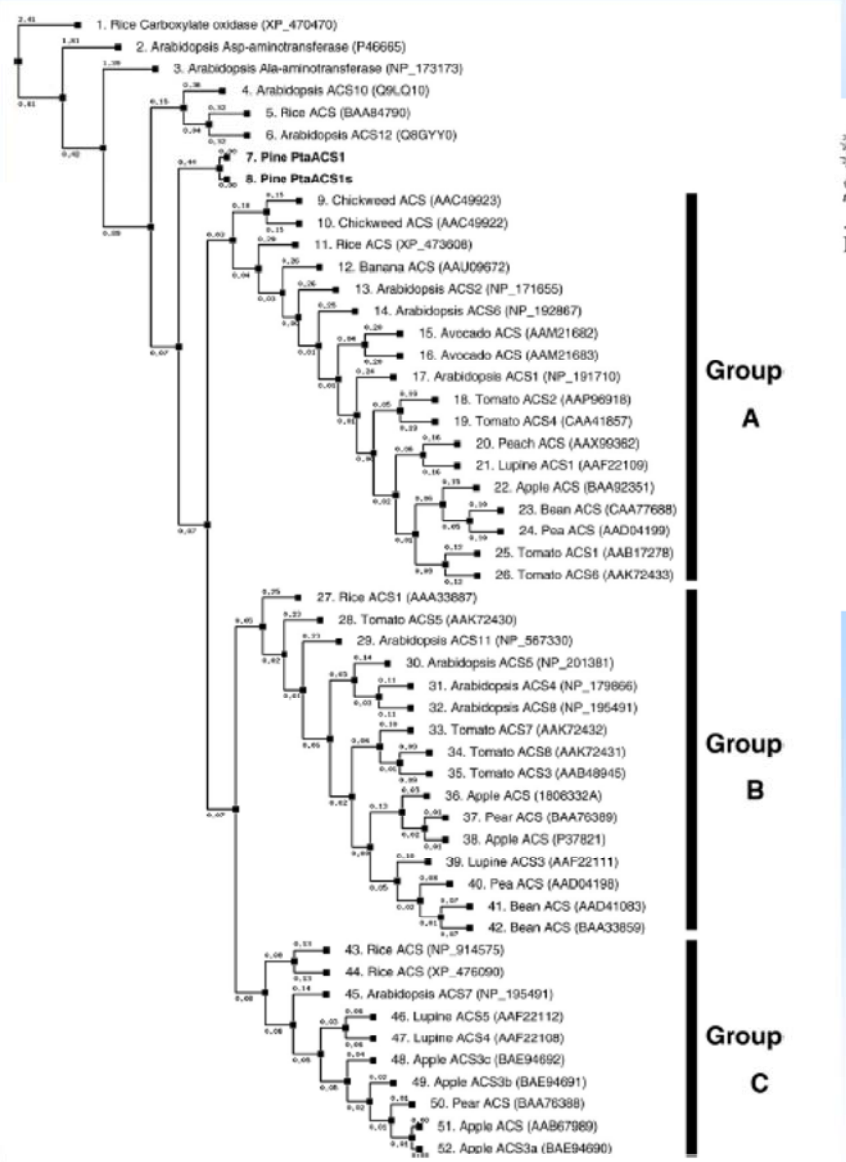
Carrie Y. C. Hung[1], Fernando Galvez[2], Yuen K. Ip[3] and Chris M. Wood[1,*]

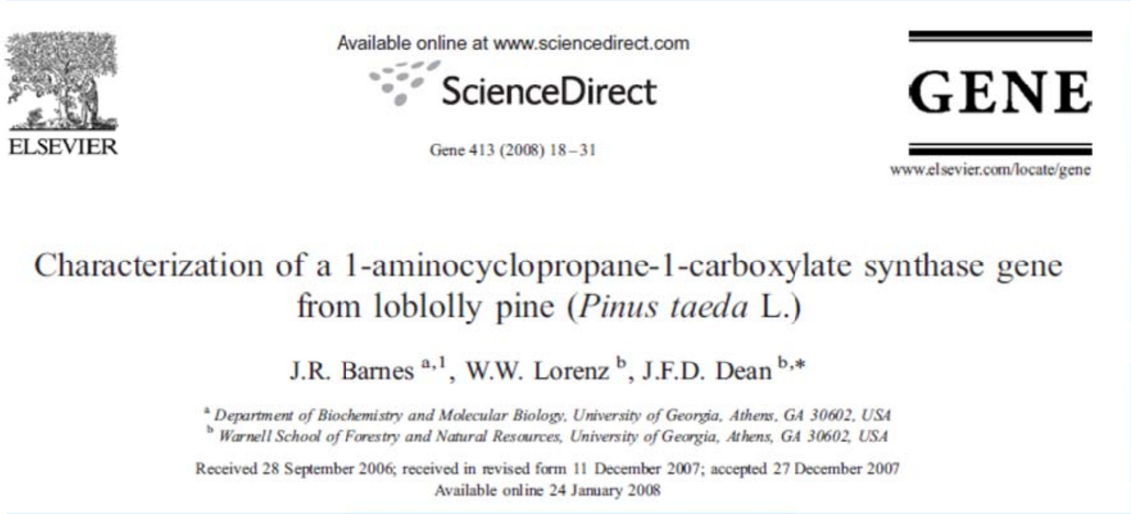
[1]Department of Biology, McMaster University, Hamilton, ON, Canada, L8S 4K1, [2]Department of Biological Sciences, Louisiana State University, Baton Rouge, LA 70803, USA and [3]Department of Biological Sciences, National University of Singapore, 10 Kent Ridge Road, Singapore 117543, Republic of Singapore



Phylogenetic tree of urea transporters across taxa. The protein sequences were aligned using ClustalW software, followed by neighbour-joining (NJ) matrix for tree reconstruction and evaluated by means of a bootstrap of 1000 replicates at http://power.nhri.org.tw

Perform ML in POWER





Available online at www.sciencedirect.com
ScienceDirect
Gene 413 (2008) 18 – 31
GENE
www.elsevier.com/locate/gene

Characterization of a 1-aminocyclopropane-1-carboxylate synthase gene from loblolly pine (*Pinus taeda* L.)

J.R. Barnes [a,1], W.W. Lorenz [b], J.F.D. Dean [b,*]

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Available online 24 January 2008

Phylogenetic tree depicting the relatedness of ACC synthase and aminotransferase protein sequences in GenBank.The phylogenetic tree was generated using the **POWER server (http://power.nhri.org.tw/)** with default parameters for the maximum likelihood method (ML) and molecular clock, but without bootstrapping or random input of sequences.

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Chieh-Hua Lin
Li-Wei Lai
Pao-Yang Chen