

**In order to save computational time, this short README just guides you to reproduce a small case (248 genes) I played before.**

- 1) Find constrained-based ML tree for t2 ((birds,crocodiles),turtles) on the supermatrix using RAxML or IQ-TREE, which is incongruent with unconstrained ML  
t1((turtles,crocodiles),birds)

raxml:

```
raxmlHPC-PTHREADS-SSE3 -T 6 -p 748962402 -# 5 -g t2.tre -s 284genes.fas -n t2_ML -m GTRGAMMA --no-bfgs
```

iq-tree: (the option "-g" was released recently in IQ-TREE 1.5.X, I would recommend you IQ-TREE since it runs much faster than RAxML if your data is pretty huge)

```
iqtree-omp1.5.1 -nt 6 -st DNA -s 284genes.fas -m GTR+G4 -g t2.tre -pre t2_ML
```

- 2) Estimate site-wise log-likelihood for each of alternative hypotheses (-z trees.tre) using raxml with option (-f G)

```
raxmlHPC-PTHREADS-SSE3 -T 6 -f G -m GTRGAMMA -z trees.tre -s 284genes.fas -n turtle_site_1k --no-bfgs
```

- 3) Passer sits-wise log-likelihood file by Phylogenetic\_signal\_parser.pl (please make sure PERL has been installed on your computer)

```
perl Phylogenetic_signal_parser.pl RAxML_perSiteLLs.turtle_site_1k turtle_genes_order.txt No
```

##### **Option Explanations** #####

**RAxML\_perSiteLLs.turtle\_site\_1k:** Site-wise log likelihood file estimated from RAxML (step 2)

**turtle\_genes\_order.txt:** Gene-based partition file in RAxML-style

**Yes/No:** Do you want to print out site-wise log likelihood for each gene?

#####

Note that:

- a) Characters in red are command lines
- b) Phylogenetic\_signal\_parser.pl only accepts no more than three alternative hypotheses (i.e., T1 Vs T2 or T1 Vs T2 VS T3).
- c) If you type "Yes" to print out site-wise log likelihood for each gene, the script Sitewise\_statistics.pl can help you do the statistics about the proportion of sites supporting each hypothesis at three levels (please see supplementary Fig. 4 in our paper).
- d) If you use this small case in your paper, please cite the original study (Chiari et al., 2012 in BMC Biology; doi: 10.1186/1741-7007-10-65).

Please let me know if you have any questions about them.

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