Fossil-calibrated phylogeny of Onagraceae: patterns of floral and chromosome evolution

William A. Freyman<sup>a,\*</sup>

 $^a Jepson\ Herbarium\ and\ Department\ of\ Integrative\ Biology,\ University\ of\ California,\\ Berkeley$ 

Abstract

blah blah

1. Introduction

blah blah

2. Methods

 $Supermatrix\ assembly.\ I$  downloaded all available DNA sequences from Gen-

Bank release 200 PLN division and performed an exhaustive all-by-all BLASTn

(Camacho et al., 2009) comparison for sequences in Onagraceae and Lythraceae.

Using a BLASTn e-value of  $1.0e^{-10}$  threshold and a sequence length percent sim-

ilarity cutoff of 0.5, I constructed clusters of putative homologs using a single-

linkage hierarchical clustering algorithm. Subspecies names were removed from

all sequences, and all but one sequence of each species was pruned from each cluster. Clusters that were not phylogenetically informative (< 4 taxa) were

discarded, and each cluster was aligned using MUSCLE (Edgar, 2004). The

alignments were concatenated by species, and any species that was not present

in at least two clusters was removed from the supermatrix. Code written to

assemble the supermatrix is availabe as the Python module Supermatrix Constructor (SUMAC) (Freyman, 2014, https://github.com/wf8/sumac).

\*Corresponding author

Email address: freyman@berkeley.edu (William A. Freyman)

Group	Age (Mya)	Prior Distribution	Mean	SD	Offset	Reference
Circaea (Onagraceae)	12	lognormal	0.0	2.0	12	(Grímsson et al., 2012)
$Epilobium \ ({\it Onagraceae})$	12	lognormal	0.0	2.0	12	(Grímsson et al., 2012)
S. Pacific Fuschia (Onagraceae)	23	lognormal	0.0	1.0	23	(Lee et al., $2013$ )
$Ludwigia \ ({\it Onagraceae})$	Paleocene	normal	60.0	3.0	-	(Zhi-Chen et al., $2004$ )
Lythraceae	82	lognormal	0.0	2.0	82	$({\rm Graham},2013)$

Table 1: Fossil priors used in BEAST analyses.

Phylogenetic analyses. Maximum likelihood analyses were performed with RAxML-HPC (Stamatakis, 2014) on the CIPRES Scientific Gateway (Miller et al., 2010) using the rapid bootstrap heuristic and the GTRCAT nucleotide substitution model. I used the ML tree to select 15 species phylogenetically widely distributed in Lythraceae to act as outgroup; all other members of Lythraceae were subsequently removed from the supermatrix before estimating divergence times. Bayesian estimates of divergence times were inferred using BEAST (Drummond and Rambaut, 2007; Suchard and Rambaut, 2009) on CIPRES and calibrated with five fossils identified with morphological synapomorphies (Table 1). The Ludwiqia fossil pollen was dated broadly to the Paleocene (Grímsson et al., 2012), so I set the prior to a normal distribution with a wide standard deviation to cover the entire time period. For all other calibration points I used a lognormal prior distribution with the offset (the minimum age of the node) corresponding to the fossil age. The BEAST analysis utilized the GTR+ $\Gamma$ nucleotide substitution model with a relaxed molecular clock (uncorrelated lognormal model) and a Yule process tree prior. The Markov Chain Monte Carlo (MCMC) was run for 100 million generations, sampling every 10 thousand generations. Tracer v1.6 (Rambaut et al., 2013) was used to assess the MCMC output for parameter convergence and ensure that the effective sample size for all parameters was above 200. The first 1000 trees were discarded as burn-in, and the remaining 9000 trees were summarized as a maximum clade credibility tree with mean divergence times.

Character evolution. blah blah

## 40 3. Results

Supermatrix assembly. blah blah

Divergence time estimates. Here is a table with the mean divergence time estimates including %95 confidence intervals....

Character evolution. blah blah

## 4. Conclusion

blah blah

## References

Camacho, C., Coulouris, G., Avagyan, V., Ma, N., Papadopoulos, J., Bealer, K., Madden, T.L., 2009. BLAST+: architecture and applications. BMC bioinformatics 10, 421.

Drummond, A.J., Rambaut, A., 2007. Beast: Bayesian evolutionary analysis by sampling trees. BMC evolutionary biology 7, 214.

Edgar, R.C., 2004. MUSCLE: multiple sequence alignment with high accuracy and high throughput. Nucleic acids research 32, 1792–1797.

Freyman, W.A., 2014. Supermatrix Constructor (SUMAC). https://github.com/wf8/sumac.

Graham, S.A., 2013. Fossil records in the lythraceae. The Botanical Review 79, 48–145.

Grímsson, F., Zetter, R., Leng, Q., 2012. Diverse fossil onagraceae pollen from a miocene palynoflora of north-east china: early steps in resolving the phytogeographic history of the family. Plant Systematics and Evolution 298, 671–687.

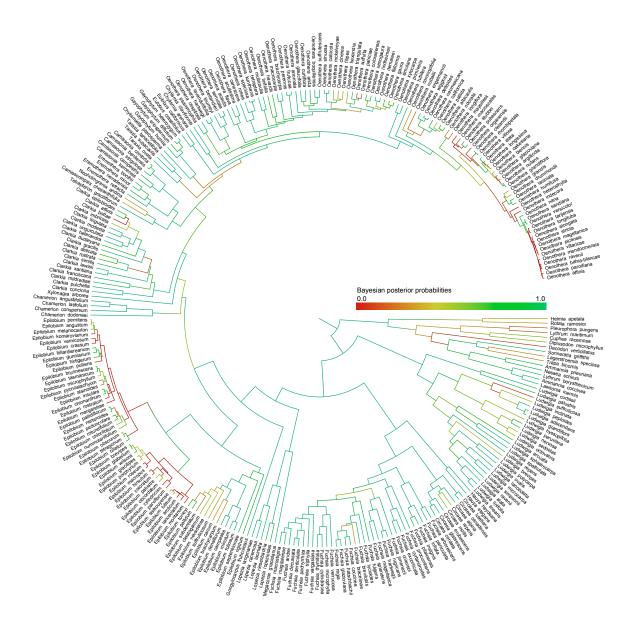


Figure 1: Phylogeny blah blah blah

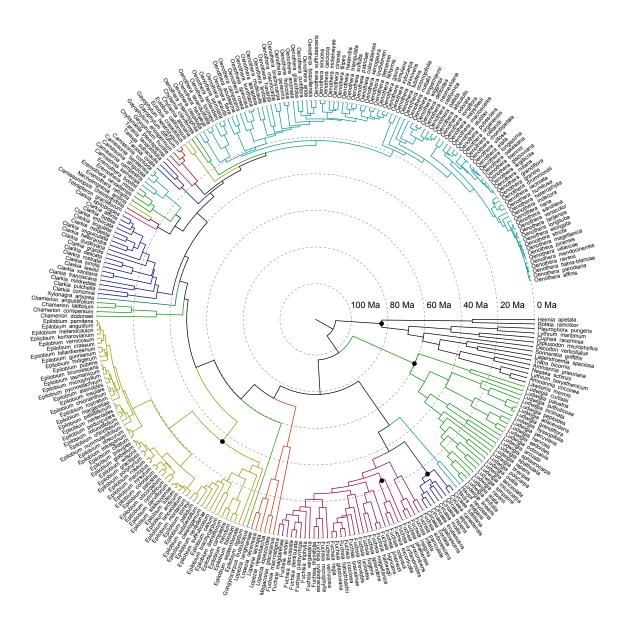


Figure 2: Sequence diagram

- Lee, D.E., Conran, J.G., Bannister, J.M., Kaulfuss, U., Mildenhall, D.C., 2013.

  A fossil fuchsia (onagraceae) flower and an anther mass with in situ pollen from the early miocene of new zealand. American journal of botany 100, 2052–2065.
- Miller, M.A., Pfeiffer, W., Schwartz, T., 2010. Creating the CIPRES science gateway for inference of large phylogenetic trees, in: Gateway Computing Environments Workshop (GCE), 2010, IEEE. pp. 1–8.
- Rambaut, A., Suchard, M., Drummond, A.J., 2013. Tracer v1.6. http://tree.bio.ed.ac.uk/software/tracer/.
  - Stamatakis, A., 2014. RAxML Version 8: A tool for phylogenetic analysis and post-analysis of large phylogenies. Bioinformatics, btu033.
  - Suchard, M.A., Rambaut, A., 2009. Many-core algorithms for statistical phylogenetics. Bioinformatics 25, 1370–1376.
    - Zhi-Chen, S., Wei-Ming, W., Fei, H., 2004. Fossil pollen records of extant angiosperms in china. The Botanical Review 70, 425–458.