

Testing for molecular convergence and selection on gecko opsin genes

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Introduction and Objectives

Though geckos are ancestrally nocturnal (active at night), diurnality (active during the day) has independently evolved multiple times (1, 2, 3). To adapt to changes in light conditions, diurnal geckos may have experienced changes in visual rods and cones or their opsin proteins, which initiate phototransduction at specific light wavelengths (4). Geckos have long-wavelength (LWS), short-wavelength (SWS1), and mid-wavelength (RH2) sensitive opsins (4, 5). I previously found evidence of molecular convergence in the LWS and SWS1 opsins. Here, I gather additional sequence data and examine all three gecko opsins for 1. further evidence of convergence and 2. evidence of differing rates of evolution throughout the gecko lineage, which may indicate selection. I expect to see convergence on diurnal geckos (Fig. 1 (A)), and if so, evidence of selection as well. Alternatively, I may see no evidence of convergence (Fig. 1 (B)) in RH2 and no differing evolutionary rates in any opsins.

Methods

- Sequences for all three opsins were gathered from whole genomes, eye transcriptomes, and Sanger Sequenced cDNA
- To obtain additional sequences, Sanger Sequencing was done with cDNA obtained from gecko eye mRNA
- Gene identifies were verified using BLAST
- Geneious software was used to align sequences and create gene trees from amino acid alignments
- To test for convergence, amino acid gene trees were compared to the species tree (pruned from (1))
- To test for differing rates of evolution, gene trees were used to run molecular clock tests for each opsin in MEGA (6)

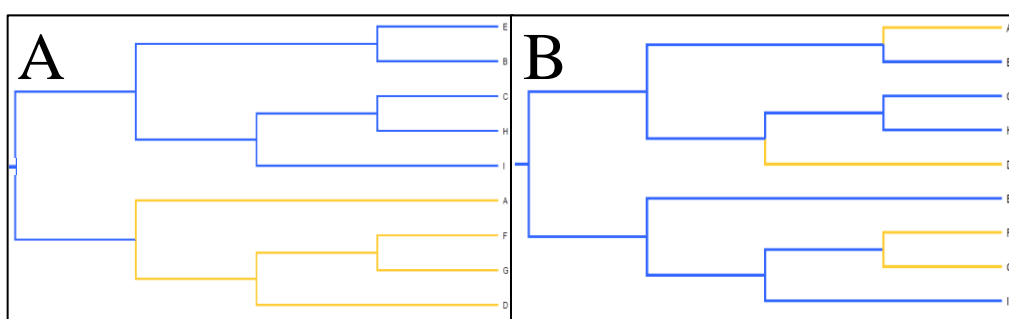


Figure 1. Amino acid tree examples where image (A) depicts convergence, while (B) depicts lack of convergence.

Results

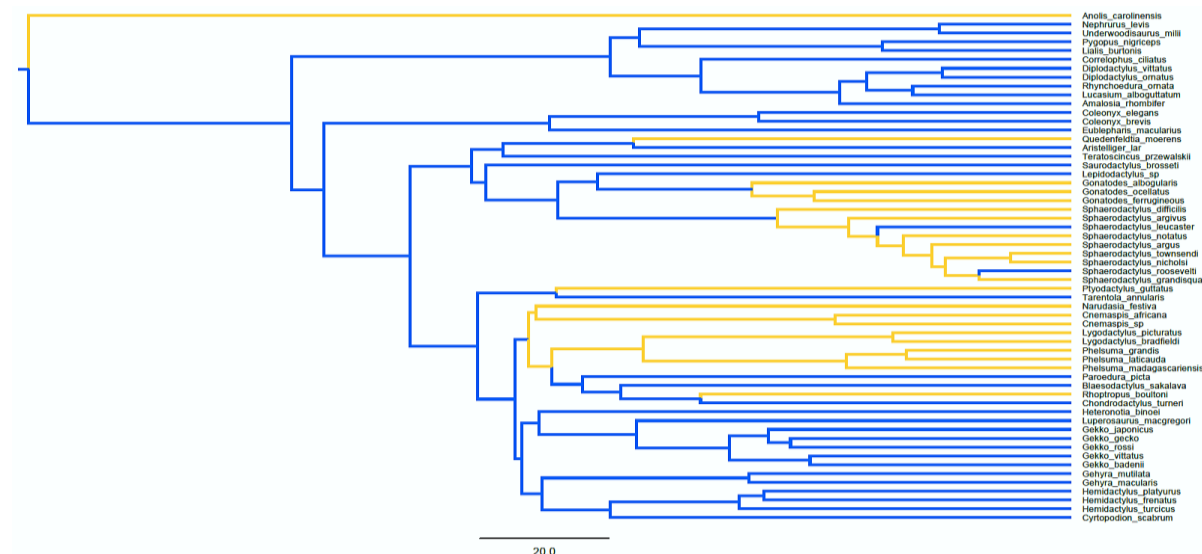


Figure 2. Accepted species tree where yellow branches are diurnal and blue branches are nocturnal (Pruned from Gamble et al. 2015).

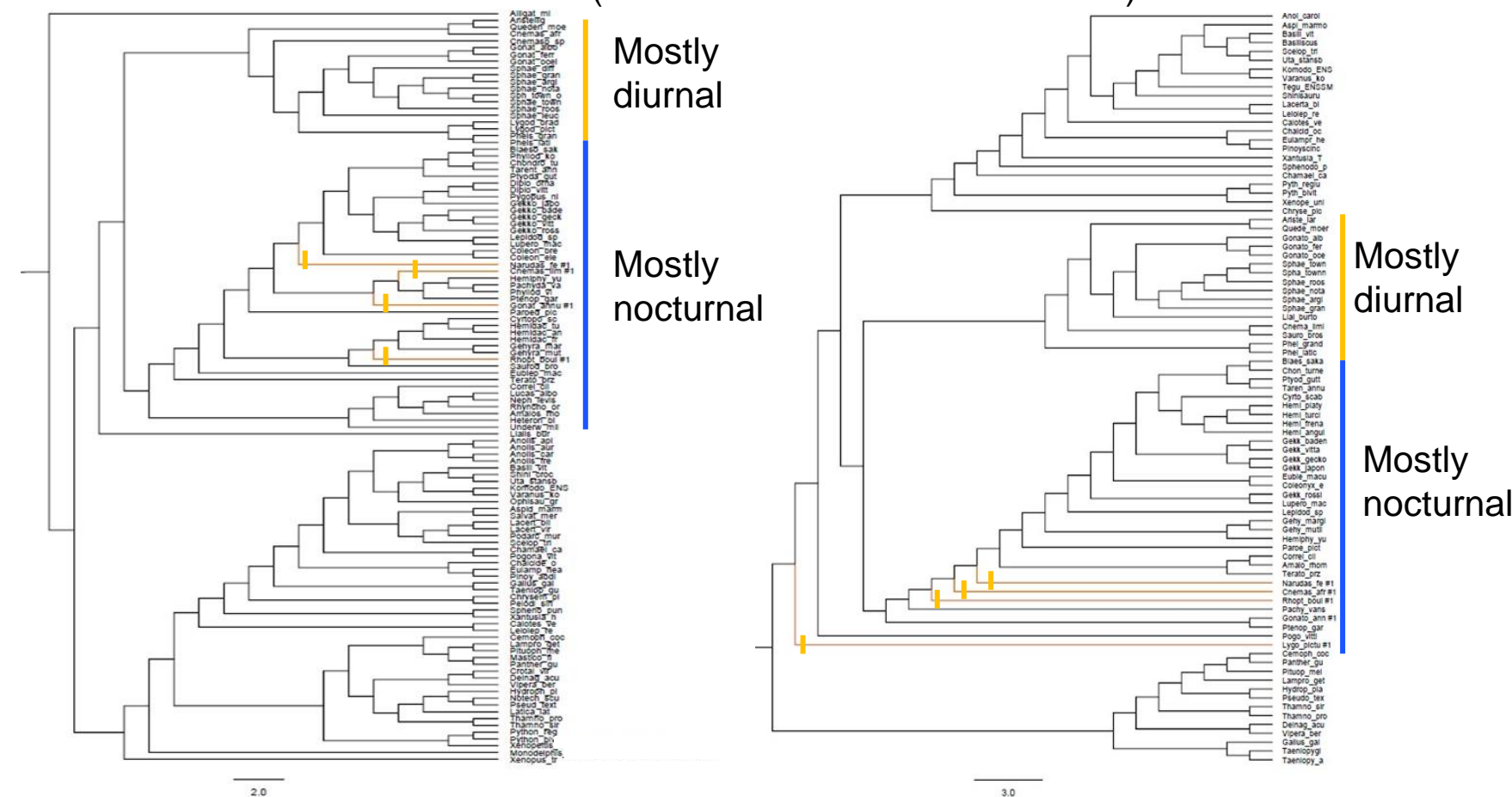


Figure 3. LWS amino acid tree where diurnal geckos are colored yellow, with yellow bands, when they show up in the mostly nocturnal clade

Figure 3. SWS1 amino acid tree where diurnal geckos are colored yellow, with yellow bands, when they show up in the mostly nocturnal clade

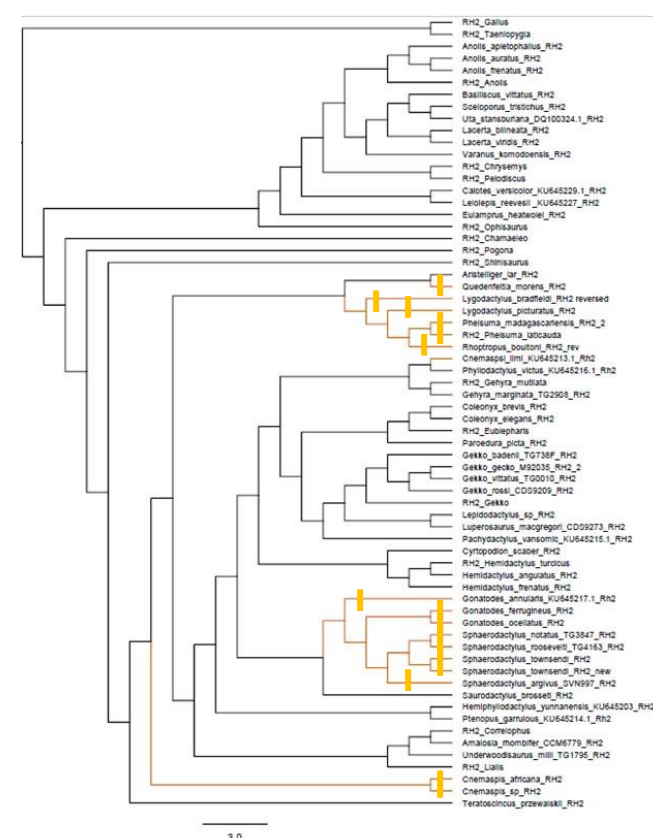


Figure 3. RH2 amino acid tree where diurnal geckos are colored yellow with yellow bands

Conclusion

With more taxa added to the LWS and SWS1 datasets, the previous result of one mostly diurnal gecko clade and one mostly nocturnal clade is seen again. In both opsins, results are more similar to Fig. 1 (A) than Fig. 1 (B), providing evidence for molecular convergence in diurnal geckos. However, the RH2 amino acid tree shows results more similar to Fig. 1 (B) than Fig. 1 (A), which does not provide evidence of molecular convergence. It is possible that molecular convergence does occur on this opsin but is not visible in the tree due to the smaller taxon sampling of RH2.

The molecular clock tests rejected the null hypothesis of equal evolutionary rates throughout geckos in all three opsins. This indicates that there are different rates of molecular change throughout the tree, which may provide evidence for selection on the opsins of diurnal geckos. Further selection tests are needed to confirm the specific branches experiencing selection.

The combination of molecular convergence and differing evolutionary rates supports the hypothesis that diurnal geckos have undergone spectral tuning of their opsins to the daytime light environment (8). I hope to do further tests of selection and ancestral state reconstruction to uncover the evolutionary history of gecko opsins and determine how molecular evolution has contributed to the visual adaptations of diurnal geckos.

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	LWS	SWS1	RH2
InL with clock	-6734.582	-9587.592	-5986.475
InL without clock	-6156.258	-9279.396	-5367.134
Result at 5% significance	Null rejected	Null rejected	Null rejected
P-value	4.580E-202	1.038E-099	9.137E-231

Figure 6. Molecular clock test was performed in MEGA (6) by comparing the ML value for the given topology with and without the molecular clock constraints under Jones-Taylor-Thornton (1992) model (7). The null hypothesis of equal evolutionary rate throughout the tree was rejected at a 5% significance level for all three opsins.