



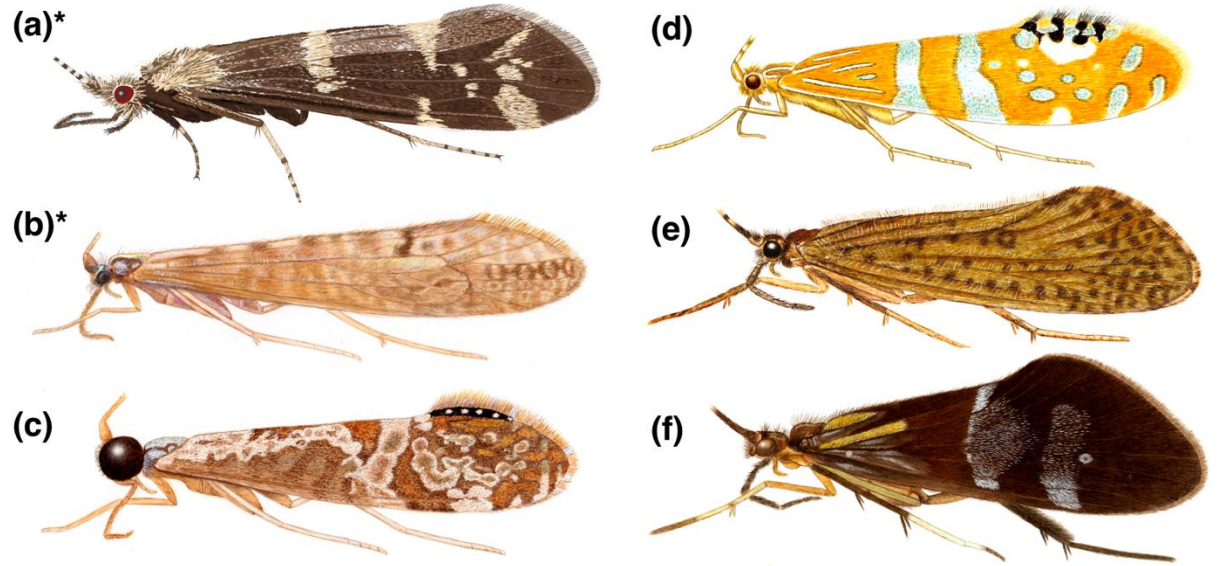
## Evolution of Opsin Genes in Caddisflies (Insecta: Trichoptera)

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Ernesto Razuri-Gonzales, Seth Bybee, Paul B. Frandsen



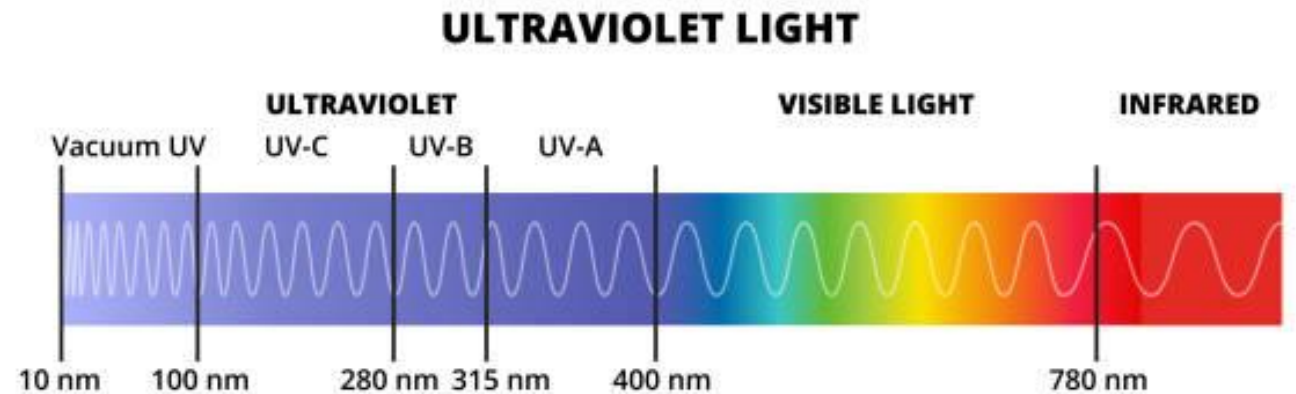
# Background

- Caddisflies are aquatic insects that spend most of their life underwater, in varying light intensities by environment.
- They use vision for finding food, avoiding predation, and in selecting a mate.
- Evolution of visual genes (opsins) in this group is poorly understood.



# Opsin Genes

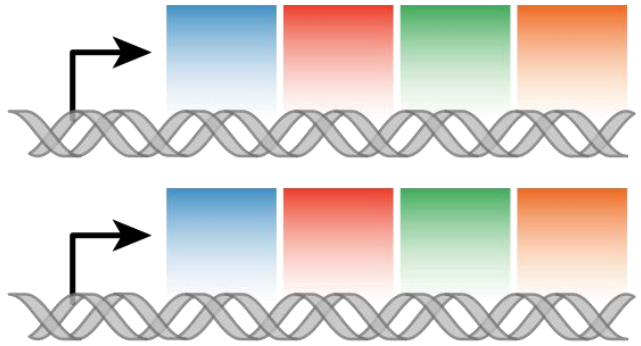
- Eyes absorb light, activate photoreceptors, opsins/chromophores change to signaling state.
- Insects have opsins from 4 gene families:
  - LW; 500-600 nm (Long Wavelength)
  - SW; 400-500 nm (Short Wavelength)
  - UV; 300-400 nm (Ultraviolet Wavelength)
  - RH7; unknown (Rhodopsin 7; diel activity)
- Typically show one or more copies of each opsin type, with gene duplications and losses.



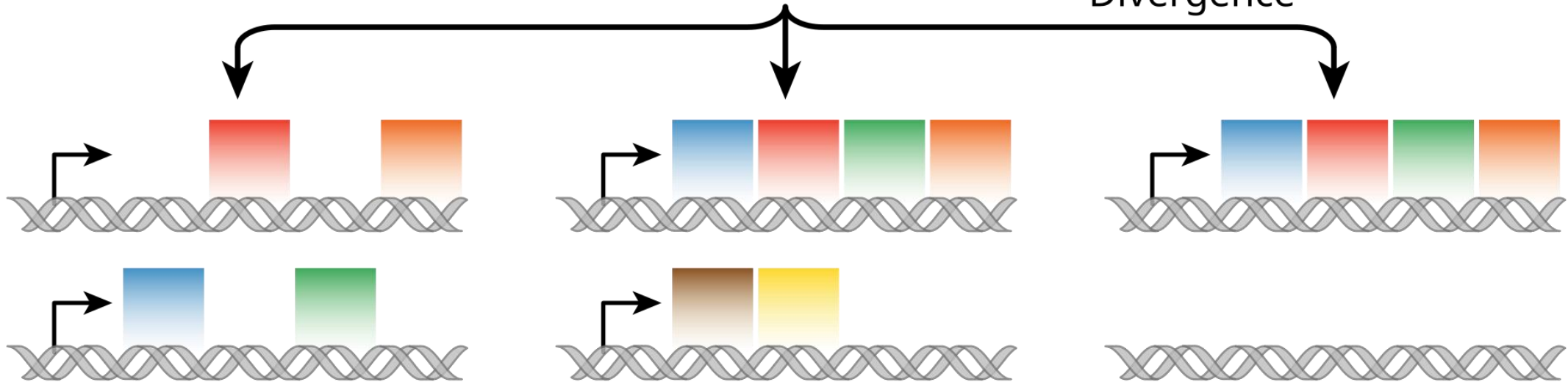
# Gene Duplication



Duplication



Divergence



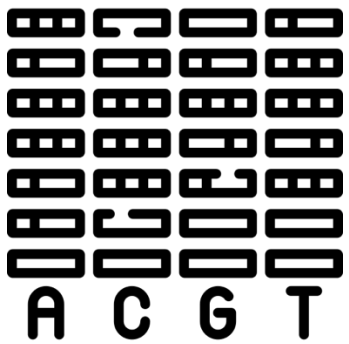
Subfunctionalization

Neofunctionalization

Degeneration/Gene loss



# Bioinformatic Methods



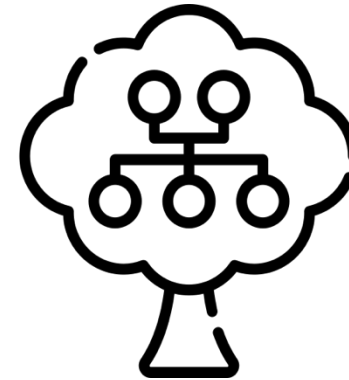
Data Acquisition  
(NCBI)



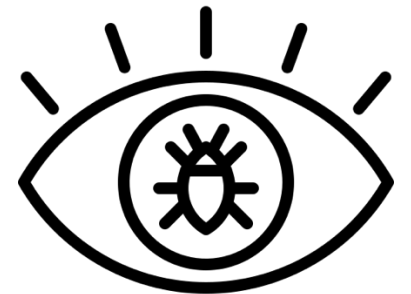
Sequence ID  
(tBLASTn)



Gene Annotation  
(AUGUSTUS)

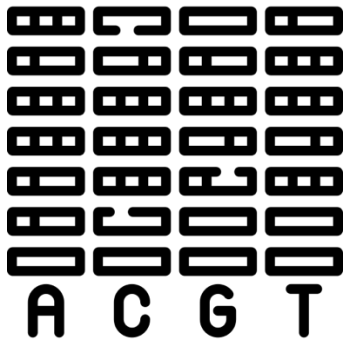


Tree Search  
(IQTREE)



Selection Tests  
(BUSTED)

# Bioinformatic Methods



Data Acquisition  
(NCBI)

# Bioinformatic Methods



Sequence ID  
(tBLASTn)

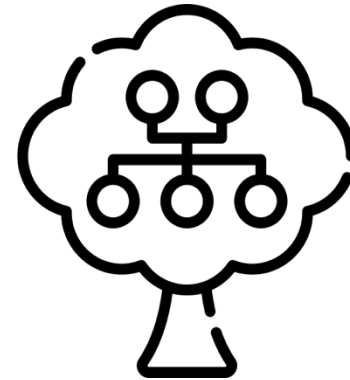
# Bioinformatic Methods



Gene Annotation  
(AUGUSTUS)

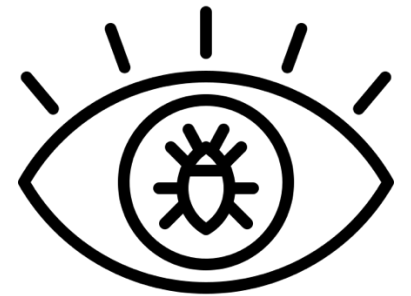


# Bioinformatic Methods

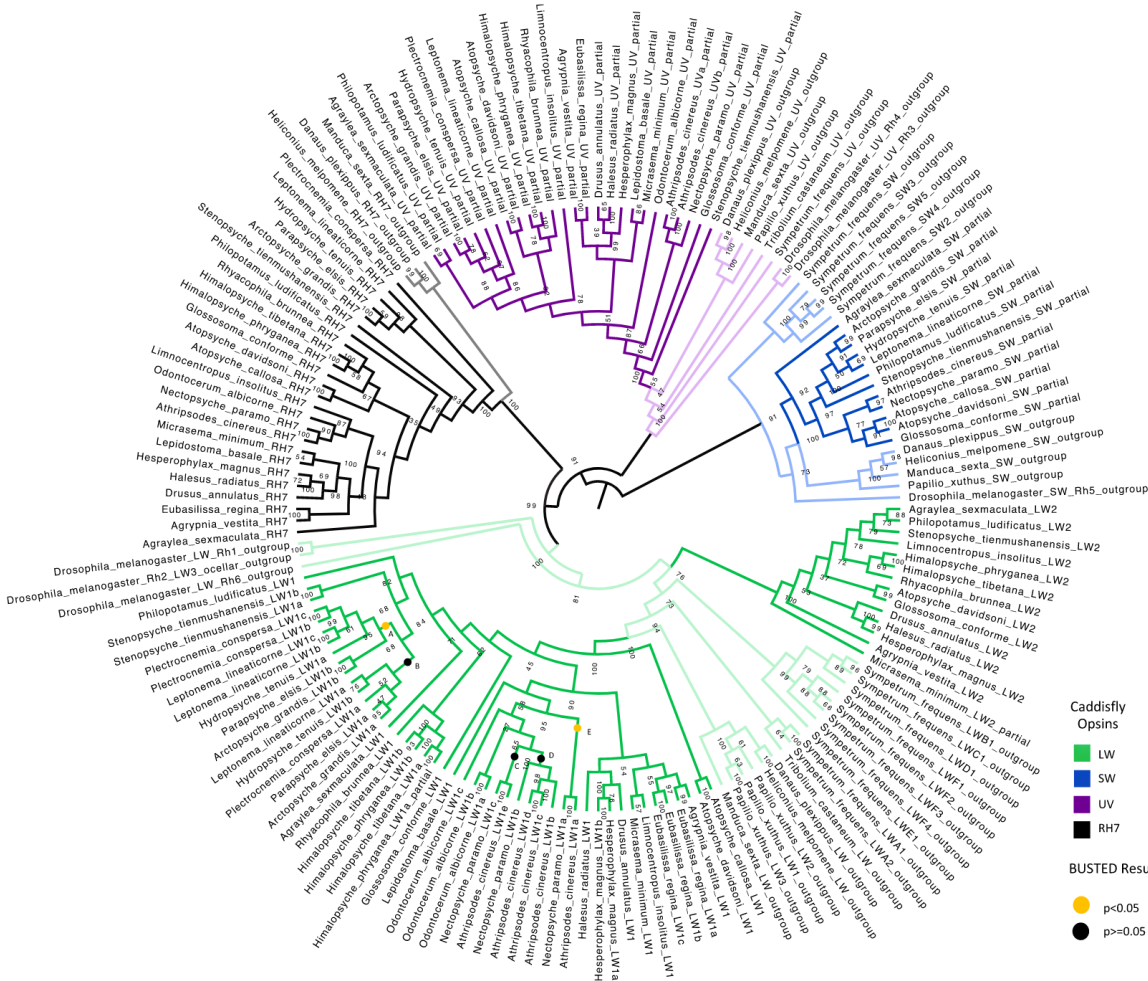


Tree Search  
(IQTREE)

# Bioinformatic Methods



Selection Tests  
(BUSTED)



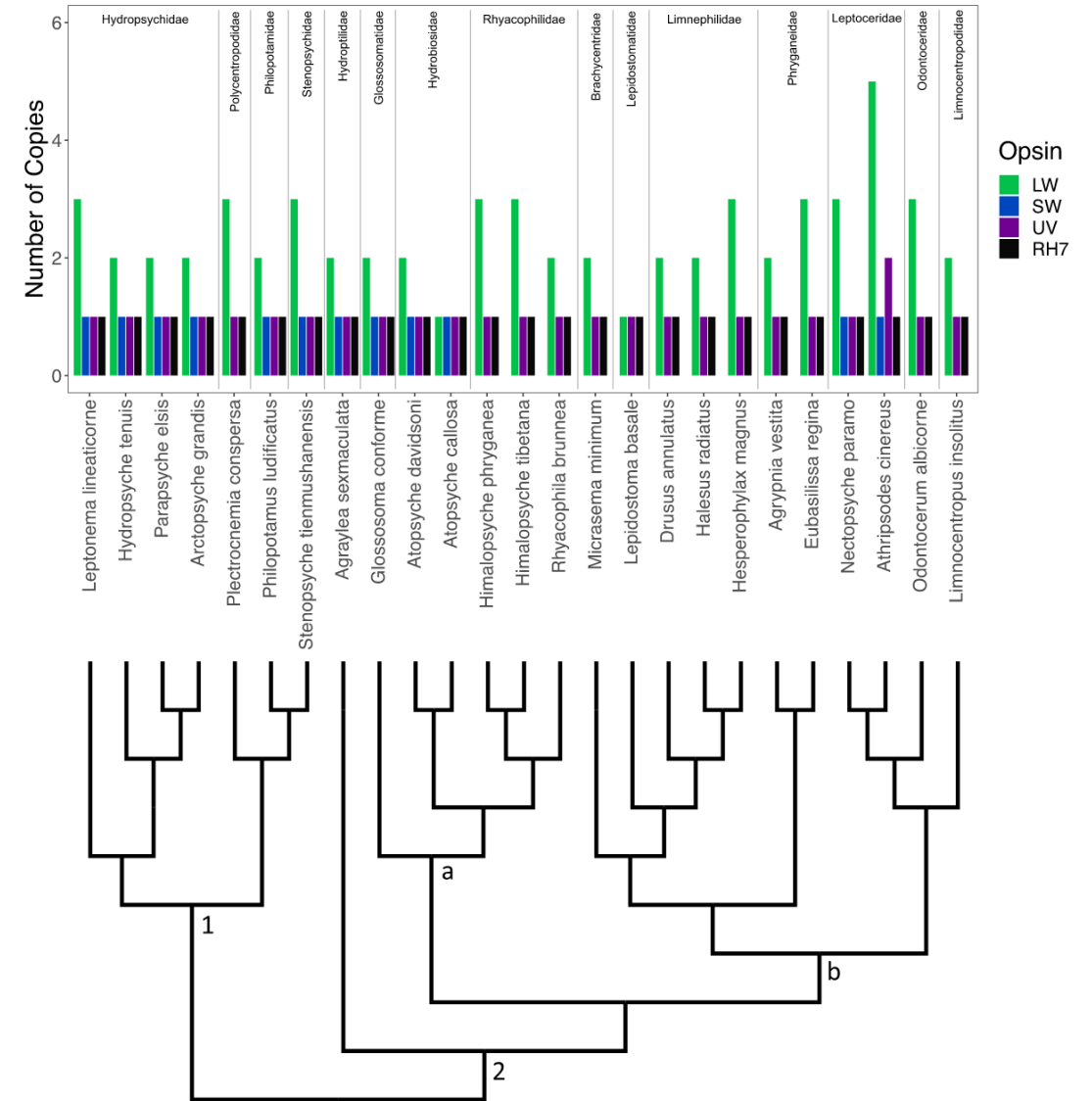
**Fig. 2. Opsin gene tree.** Maximum likelihood tree from caddisfly and outgroup opsin DNA sequences. Nodes are labeled with bootstrap values and branches are colored by opsin type. The letters at the end of the node labels (e.g. LW1a, LW1b, etc.) denote **multiple copies of that opsin type in a species**. Clades marked with a dot (A to E) were tested for episodic diversifying selection with BUSTED ([supplementary table S3, Supplementary Material online](#)).

## Results: Opsin Gene Tree

- Maximum likelihood tree of opsin genes of caddisflies and outgroups
- Number of opsin paralogs found in each specie ranged from three to nine
- LW opsins form 2 clades, consistent w/other insects
- SW opsins were lost once
- Episodic diversifying selection identified in 3 paralogs

# Results: Copy Number

- Opsin count by species
- Bar plot shows copy number of each opsin gene within the species' genome
- Subfamilies with varying life history strategies annotated
  - 1) Annulipalpia – retreat makers
  - 2) Intergripalpia – cocoon/tubes
  - a) Basal Intergripalpia – cocoon
  - b) Basal Intergripalpia - tube/case
  - *N. paramo* and *A. cinereus* regained UV opsin usage?



**Fig. 3.** Opsin counts by species. A bar plot of the number of opsin copies in each caddisfly genome. The bars are colored by opsin type. The species are ordered by the species phylogeny indicated below the bar plot, which is based on a recent study that examined the caddisfly phylogeny in depth (Frandsen et al. 2024). Families are labeled above the bar plot. Suborders are labeled within the phylogeny as follows: (1) Annulipalpia—retreat makers; (2) Intergripalpia—cocoon- and tube-case makers; (a) and *Agraylea sexmaculata* basal Intergripalpia—cocoon makers; (b) tube-case-making Intergripalpia.





## Discussion & Conclusions

- Caddisfly opsin evolution likely driven by life-history strategies
- Species inhabit fast moving streams as larvae and live shortly in riversides as adults.
- Caddisflies primarily crepuscular, most active low-light conditions, may be related to SW opsin loss
- High # of opsins, and presence of SW opsins could be related to wing coloration patterns.
- Some caddisflies exhibit sexual eye dimorphism, which also contain patterns of the most colorful wings (Leptoceridae).