Understanding the evolution of spiders through novel genetic markers

Maya Wolf, Jesus A. Ballesteros & Gustavo Hormiga
Department of Biological Sciences, The George Washington University

THE GEORGE WASHINGTON UNIVERSITY
WASHINGTON, DC

1. *Dipoena kuyuwini*

2. *Tetragnatha straminea*

3. *Araneus marmoreus*

4. *Micrathena gracilis*

Introduction

- Spiders are one of the most ubiquitous predatory organisms on Earth with over 45,000 species described.[1]
- Much information regarding this lineage's history remains unknown.[2]
- This is in part due to a limited amount of genetic markers.[3]
- We selected ten protein coding fragments to test and optimize their amplifications on a variety of lineages within the orb-weaver clade as well as on a few outgroups.
- We aim to test the utility of these new markers in providing information on spider phylogenies and illuminating evolutionary hypotheses.

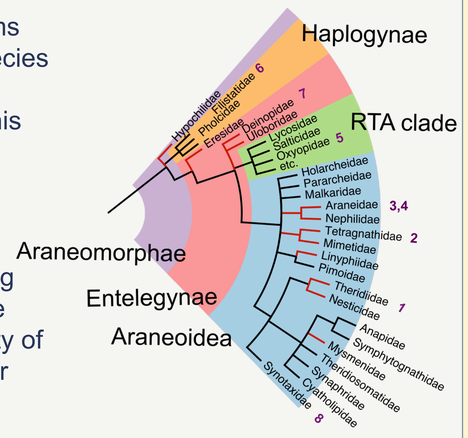
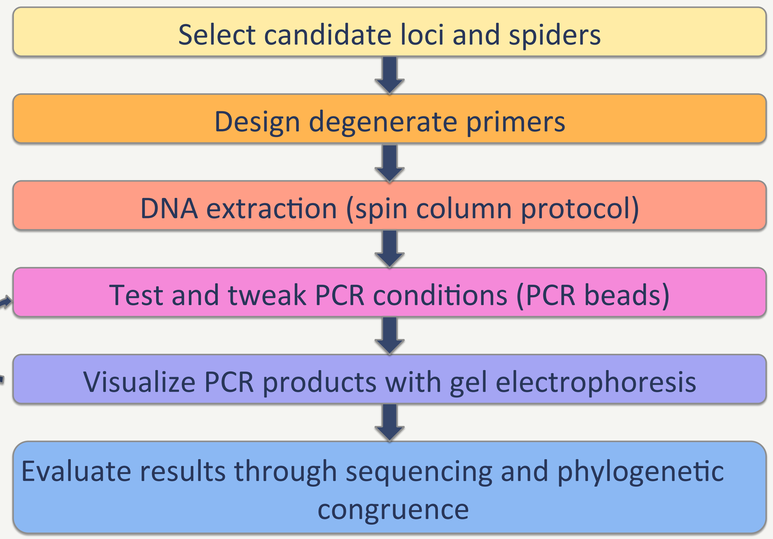


Figure 1. Summary phylogenetic tree of the spider lineages included in this study. Numbers next to a family indicate spiders we tested on. Highlighted in red are the spider lineages used as reference in the primer design.

Materials and Methods



Results

- So far, when visualizing the agarose gels after amplifying our genetic markers, we have obtained mostly smears and unspecific bands.
- Using PCR beads has given us mixed results, and is difficult to troubleshoot.

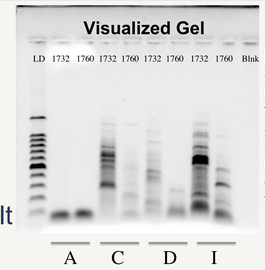


Figure 2. Example of an agarose gel showing Multiple bands when testing primers A,C, D, and I, on two different spiders. Next rounds of PCR on this set should be done at a higher annealing temperature.

Detail of the MSA (nt) of Sarcomeric protein

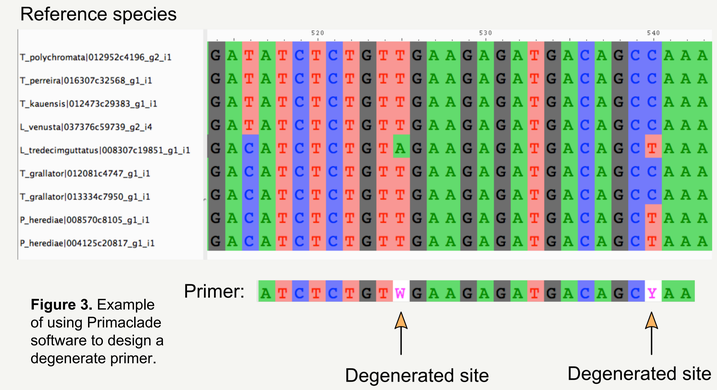


Figure 3. Example of using Primaclade software to design a degenerate primer.

Discussion & Future Directions

- We are still working on optimizing PCR conditions for each of these new primers, and have made progress in understanding which factors to manipulate in order to make the reactions more successful.
- We are moving towards using a custom PCR kit, as it will give us more control over the factors involved in PCR optimization.
- While we might not succeed in amplifying the 10 chosen primer pairs, we can continue choosing from a large pool of candidate genes until we are successful.

Acknowledgements & References

Robert Kallal provided insights on content and imaging. This research was supported by the Harlan Scholarship Fund and a grant from the US NSF to GH.

1. World Spider Catalog. 2015. Natural History Museum Bern, version 16.5 http://wsc.nmbe.ch **2.** Wunderlich J. 1986. *Spinnenfauna Gestern und Heute: Fossile Spinnen in Bernstein und ihre Heute Leben-denVerwandten*. Wiesbaden, Ger.: Erich Bauer Verlag bei Quelle und Meyer. **3.**Dimitrov D, Lopardo L, Giribet G, Arnedo M, Alvarez-Padilla F, Hormiga G. 2011.Tangled in a sparse spider web: single origin of orb weavers and their spinning work unravelled by denser taxonomic sampling. *Proceedings of the Royal Society B: Biological Sciences,* 279(1732), p.1341-1350.

2. *Peucetia viridans*

6. *Kukulcania hibernalis*

7. *Deinopis* sp.

8. *Paratupua grayi*