The Parasitic Plant *Phelipanche aegyptiaca* Adapts Gene Expression to Different Host Species

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Introduction

Phelipanche aegyptiaca (Egyptian broomrape) is an obligate holoparasite that lacks photosynthetic capacity and relies entirely on host plants for water and nutrients. Unlike more specialized parasites, P. aegyptiaca has a broad range of broadleaf hosts. Examples of its range of hosts include carrot, cucumber, tomato, sunflower, mustard and lentil. Because of this parasitic plants represent a significant agricultural threat, causing billions of dollars in crop losses annually.

Successful parasitism requires a precise coordination of molecular processes including host recognition, haustorium development, vascular connection establishment, and nutrient acquisition. Though morphological stages of parasitic plant infection have been well-characterized, molecular mechanisms underlying host compatibility remain an open area of research, particularly in generalist parasites capable of a wide range of phylogenetically distinct hosts.

Understanding host-specific transcriptional programs could reveal novel targets for parasitic control and provide further insights into the molecular basis of parasitic plant host range.

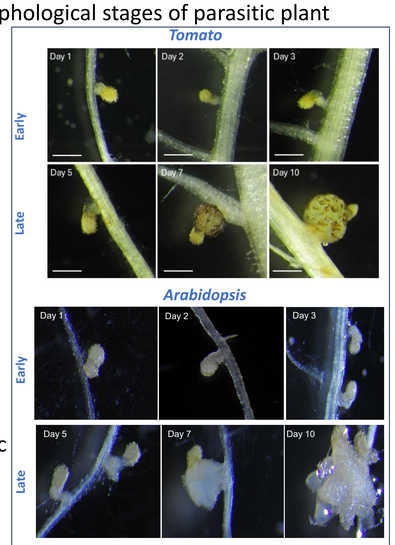

Figure 1. Developmental progression of *Phelipanche aegyptiaca* on *Arabidopsis* and tomato roots. Day 1: 5 d after inoculation; parasite radical stuck to host root. Image Credit: Sukhmanpreet Kaur

Objective

Does *Phelipanche aeygptiaca* exhibit host-specific gene expression patterns when parasitizing phylogenetically divergent host species?

Specific Objectives:

- Determine host defense responses to parasitism
  1. Identify host genes manipulated by the parasite
  2. Test function of specific genes I parasitic interactions
- Gain insight into the mechanisms of parasitism
  1. Identify important parasite genes
  2. Explore how parasites adapt to different hosts

Figure 2. Left. A closeup look of *Phelipanche* flowers. Right. *Phelipanche aegyptiaca* infecting tomato in a field setting.

Methods

Condition	Day1	Day2	Day3	Day5	Day7	Day10	Genes x Samples
Arabidopsis (control)	4	4	3	4	4	4	32,833 genes x 23 samples
Arabidopsis (parasitized)	4	4	4	4	4	4	29,864 genes x 24 samples
Tomato (control)	4	4	4	4	4	4	34,075 genes x 24 samples
Tomato (parasitized)	3	3	4	3	3	4	29,864 genes x 20 samples

Table 1. Experimental design across hosts, time, and treatment.

Plant Material & Infection Protocol

- Seeds for *Arabidopsis thaliana* and *Solanum lycopersicum* were selected based on susceptibility to *Phelipanche aegyptiaca* and genome availability
- *P. aegyptiaca* seeds were used to inoculate host plants
- *Arabidopsis* and tomato hosts were grown in pots for ~20 days.
- Plants were maintained in growth chambers until they were transplanted into a polyethylene (PE) bag system

RNA Extraction & Sequencing

- Host root sections were collected by removing parasite tissue under a dissecting microscope
- Control samples consisting of host root sections from uninoculated plants were collected simultaneously to serve as a reference
- RNA was extracted from host root sections across days 1-10

Bioinformatics Pipeline

- Dual RNA-seq alignment was performed using HISAT2 against combined reference genomes (*P. aegyptiaca* and respective host genomes)
- DESeq2 to identify host-specific parasite response
  - Genes with adjusted p-value < 0.05 and |log2FC| > 1 were considered significantly differentially expressed.
- Co-expression network analysis was performed using WGCNA.
- Functional enrichment was assessed using GO term analysis

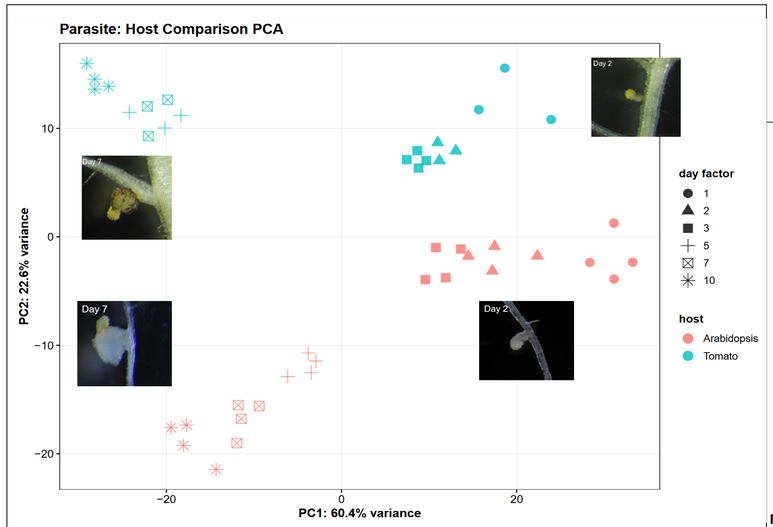

Figure 2. Principal component analysis of *Phelipanche aegyptiaca* transcriptomes across host species and infection timepoints.

Results

Global Transcriptional Patterns Reveal Host-Specific Responses

- Dual RNA-seq analysis successfully separated parasite and host transcriptomes, yielding expression data for 18,432 *P. aegyptiaca* genes across both hosts
- Principal component analysis revealed clear separation of samples by host species (PC1: 60% variance)

Extensive Differential Expression Between Host Infections

- 3,847 differentially expressed genes (DEGs) between *Arabidopsis* and tomato infections
- Of these, 1,923 genes showed higher expression during *Arabidopsis* infection while 1,924 were upregulated in tomato infections
  - GO enrichment analysis showed flavonoid metabolism enrichment in *Arabidopsis* and serine hydrolase enrichment in tomato.
- 2,156 genes were commonly upregulated during both infections (shared core parasitism program) while 1,691 genes showed host-specific expression

Co-expression Network Analysis Identifies Host-Responsive Gene Modules

- WGCNA identified 24 co-expression modules, with 6 modules showing significant correlation with host identity ($p < 0.001$).

Discussion

Host-Specific Transcriptional Plasticity Underlies Parasitic Success

- *P. aegyptiaca* exhibits transcriptional plasticity when infecting divergent hosts
- Nearly 4,000 host-responsive genes

Implications for Host Range and Parasitic Evolution

- Parasites maintain a core set of conserved parasitism genes supplemented by flexible, host-responsive expression programs
  - This may represent a key innovation enabling broad host range evolution in parasitic plants.

Future Directions and Applications

- Future work should focus on functionally validating candidate genes
- Genes showing consistent upregulation across all hosts represent targets for broad-spectrum parasite control
  - host-specific genes could enable crop-tailored resistance strategies.

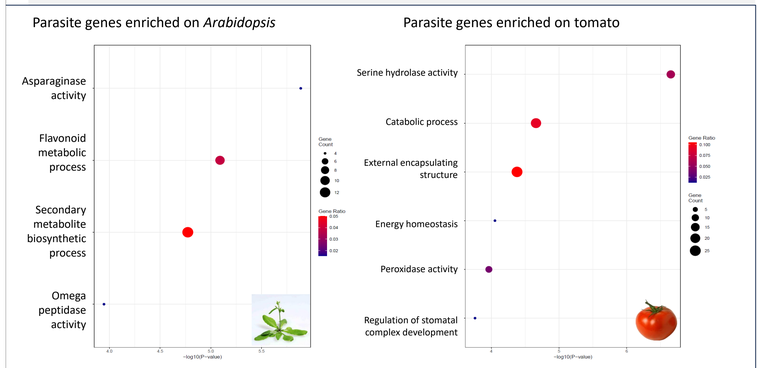

Figure 3. Gene Ontology enrichment of *Phelipanche aegyptiaca* genes differentially expressed in response to host species. Total terms have been simplified using rrvgo semantic simplification.

Sources

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Acknowledgements

I'd like to thank my advisors Drs. Wright and Haak as well as Dr. Westwood for their guidance. I also want to thank Sukhman for her help during rotation. Finally, I'd also like to thank Virginia tech SPES, BSE, TPSC for their support in addition to the funding provided by USDA NIFA.