COMPARATIVE ANALYSIS OF THE DELTOCEPHALINE LEAFHOPPER TRANSCRIPTOMES FOR THE ELUCIDATION OF VECTOR COMPETENCE GENES

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BACKGROUND

- The subfamily Deltocephalinae (Hemiptera: Cicadellidae) is the largest and more diverse subfamily of Deltocephalinae [1].
- Most of the leafhopper species that are vectors of economically important diseases are included within this subfamily [2].
- Even though insect-borne plant pathogens cause serious damage to economically important crops, the mechanisms that regulate their transmission process are not completely understood [3]
- It is believed that the ability to effectively acquire and transmit a pathogen may be genetically regulated in the insect vectors [5].
- Therefore, the comparison of the transcriptomes of vector and non-vector leafhoppers can produce new insights regarding this process.

OBJECTIVE

- Assemble, annotate and compare the transcriptomes of the six leafhopper species to identify candidate genes involved in vector competence

MATERIALS AND METHODS

- **RNA extraction**

Total RNA of the six leafhoppers was extracted using the E.Z.N.A. Mollusk RNA kit (Omega Bio-Tek, Inc., Norcross, GA). Some insects were recollected from the field during late summer and early fall, while others come from colonies maintained at Oklahoma State University.

- **Transcriptome sequencing and assembly**

Transcriptomes were sequenced using an Illumina HiSeq 4000 platform. Transcriptome assembly was made using the Trinity *de novo* pipeline [6]. Duplicates with more than 95% similarity were removed to obtain “unigene” sets for each species.

- **Annotation and clustering analysis**

Peptide sequences were obtained with TransDecoder [7]. CD-HIT [8] was used to perform a clustering analysis of the peptides.

RESULTS

Table 1. Trinity assembly and unigene set statistics

Species	Trinity assembly # contigs	Trinity assembly N50 (bp)	Unigene set # contigs	Unigene set N50 (bp)
B. neglecta	542000	1365	344753	1323
B. rubrostriata	415639	1564	285542	1416
D. maidis	221586	2020	133542	2000
E. exitiosus	336694	1599	178231	1578
G. nigrifrons	287140	1696	178157	1647
M. quadrilineatus	391269	1452	253294	1375

Table 2. Percentage of transcripts annotated to different biological databases

	Swiss-Prot	KEGG	GO	eggNOG
B. neglecta	44.96%	30.02%	33.80%	28.95%
B. rubrostriata	30.03%	17.34%	20.26%	16.74%
D. maidis	47.25%	28.98%	32.91%	28.25%
E. exitiosus	28.32%	16.54%	19.58%	15.91%
G. nigrifrons	41.90%	23.55%	27.46%	22.88%
M. quadrilineatus	26.54%	17.12%	20.23%	16.50%

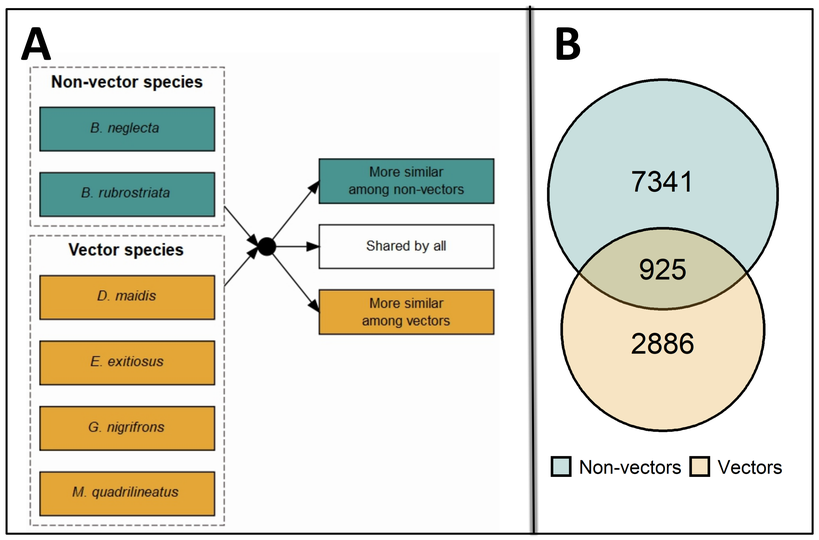

Figure1. Clustering analysis of the peptide sequences. (A) Diagram of how the comparison was made. (B) Number of peptide sequences obtained in each group.

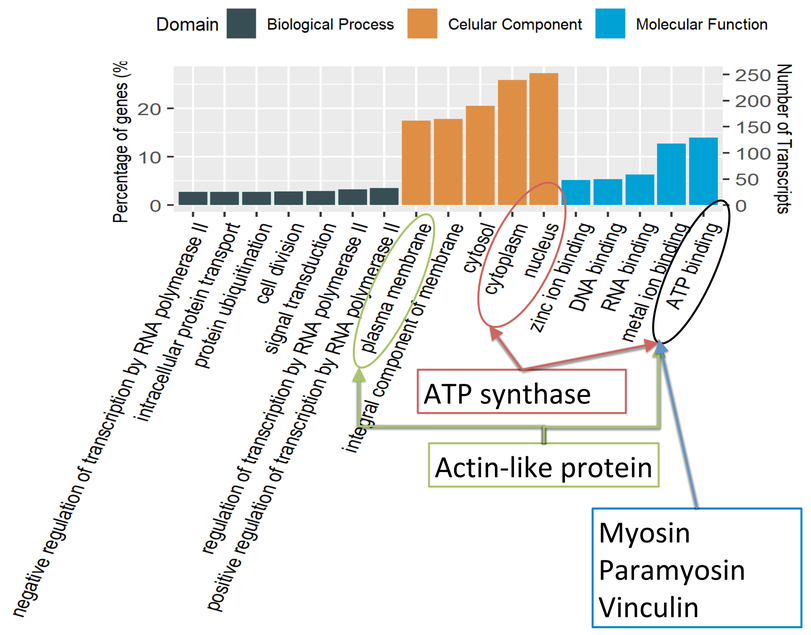

Figure 2. Five most represented GO terms of each domain in the vector peptide dataset and some associated insect vector transmission proteins.

CONCLUSIONS

- Between 29.95% to 47.78% of all the transcripts produced a match to at least one of the tested functional databases.
- A total of 925 peptide sequences shared among vector species have less than 85% similarity to sequences obtained from non-vector leafhoppers.
- Several of those peptide sequences seems to be related with insect vector transmission and need to be further studied to have a better understanding of vector competence

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