



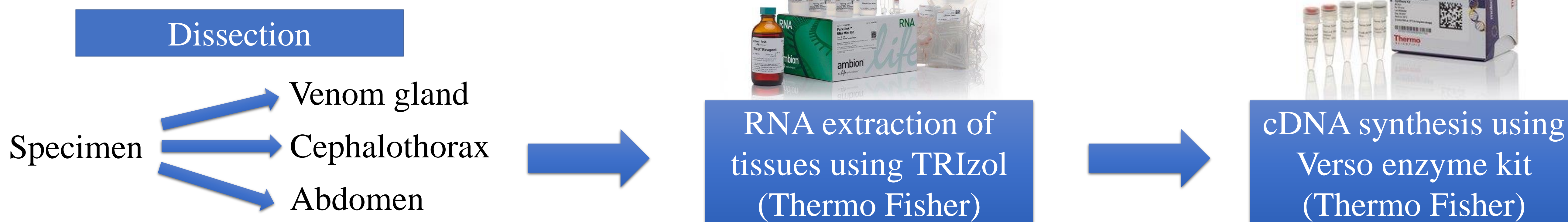
Comparative Analysis of Black Widow Toxins to Evaluate Biomosquitocide Potential

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Background

Widow spiders such as *Latrodectus mactans*, or more commonly known as the black widow, have unique venom profiles characterized by their taxa-specificity. However, few toxin sequences have been resolved to allow inter-specific comparisons and is a lack of studies exploring the expression profiles of these toxins. In this study, we bioinformatically compared two black widow delta-latrodectotoxin (δ -LIT) sequences from *Latrodectus mactans* and *Latrodectus geometricus*, that were resolved in our lab, with a publicly available δ -LIT from the Mediterranean widow, *Latrodectus tredecimguttatus*. Additionally, toxin expression profiles were resolved for *L. mactans* female, male, and spiderling tissues.

Methods and Results

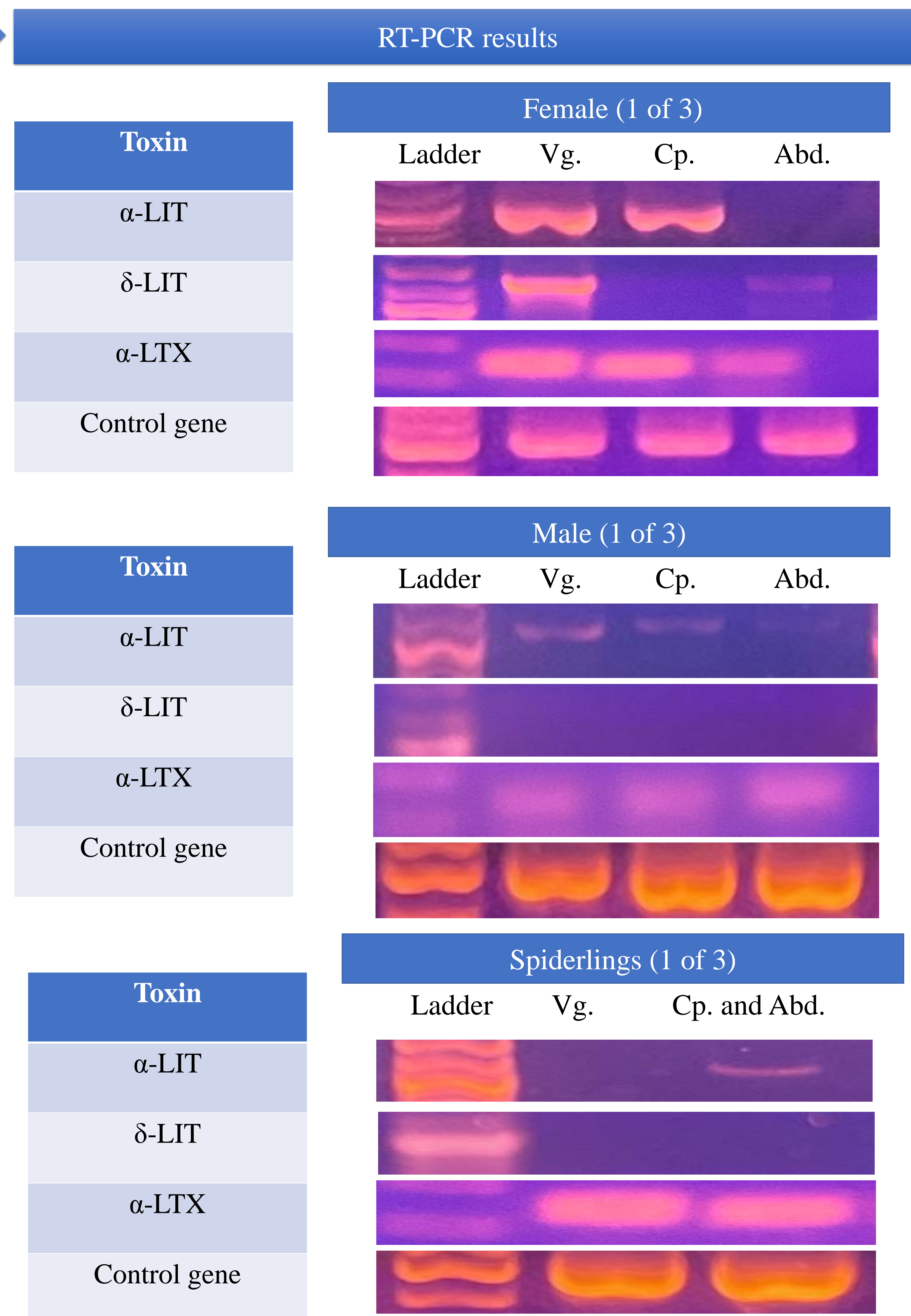


Pairwise alignments of our resolved δ -LITs (*L. mactans* & *L. geometricus*) with other LITs

Toxin comparison	Percent Identity	Query coverage
Lmac α -LIT vs δ -LIT	33.33 %	7%
Lmac α -LIT vs Lgeo α -LIT	93.91%	65%
Lmac δ -LIT vs Lgeo δ -LIT	99.27%	29%
Lmac α -LIT vs Ltre α -LIT	64.27%	99%
Lmac δ -LIT vs Ltre δ -LIT	88.0%	88%

Amino Acid Sequence Alignments of δ -LITs from *L. mactans*, *L. geometricus*, and *L. tredecimguttatus* using Clustal Omega

LtreDLIT	VDDNKAFAQALYALFYGTQTYAAVMFFLLEQHSYLADYQYKGGDDVFNFAEFNNVAIIFD	300
LmacDLIT	VDDNKAFAQALYALFYGTQTYAAVMFFLLEQHSYLADYQYKGGDDVFNFAEFNNVAIIFD	100
LgeoDLIT	VDDNKAFAQALYALFYGTQTYAAVMFFLLEQHSYLADYQYKGGDDVFNFAEFNNVAIIFD	257
LtreDLIT	DFKSSLTG-DGGLIDNVIEVLTNPKALPFKINADSKLYRELVRTKALETLKNTIKQTDL	360
LmacDLIT	DFKSSLTG-DGGLIDNVIEVLTNPKALPFKINADSKLYRELVRTKALETLKNTIKQTDL	159
LgeoDLIT	DFKSSLTG-DGGLIDNVIEVLTNPKALPFKINADSKLYRELVRTKALETLKNTIKQTDL	316
LtreDLIT	PLIDDIPETLSQVNFNDENQLPTPIGNWVGVVRYAVQYENKGMYSKFSEWSDPFTVQ	420
LmacDLIT	PLIDDIPQNLVQINFPD-ENQLSTPIGNWVGVVRYAVQYENKGMYSKFSEWSDPFTVQ	218
LgeoDLIT	PLIDDIPQNLVQINFPD-ENQLSTPIGNWVGVVRYAVQYENKGMYSKFSEWSDPFTVQ	375
LtreDLIT	GNACPTIKVRVDPKRNRLIFRKFNSGKPFAGTMTHTSQTNFKDIDRDLYDAALNINLKLK	480
LmacDLIT	GKGSPIKVPVDPKRNRLIFRKFNSGKPFAGTMTHTSQTNFKDIDRDLYDAALNINLKLK	278
LgeoDLIT	GKGSPIKVPVDPKRNRLIFRKFNSGKPFAGTMTHTSQTNFKDIDRDLYDAALNINLKLK	435



Discussion

Toxin profiling revealed a very limited expression pattern of δ -LIT specifically in females and was absent in males and spiderlings. Since females display cannibalistic behavior against males and spiderlings against each other, the lack of δ -LIT in males and spiderlings implies toxicity is substantial against other widows. Previous studies of the *L. tredecimguttatus* δ -LIT revealed that δ -LIT is synthesized as an inactive precursor, further supporting this hypothesis and is extremely potent against invertebrates in its active form. Pairwise and multiple sequence alignment comparisons of δ -LITs across species shows high conservation levels suggesting that characteristics of *L. tredecimguttatus* δ -LIT, related to functionality, are also conserved. Moreover, substantially less conservation was observed in the pairwise alignment of *L. mactans* α -LIT to *L. mactans* δ -LIT suggesting differential functions for these two toxin classes. In conclusion, δ -LIT appears likely to be a highly potent invertebrate-specific toxin and therefore an intriguing candidate to further pursue as a biomosquitocides.

Acknowledgements

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Key:
Vg. = Venom gland
Cp. = Cephalothorax
Abd. = Abdomen

