



# Transcriptomic meta-analysis to elucidate the presence and expression of neuropeptides in insect embryos

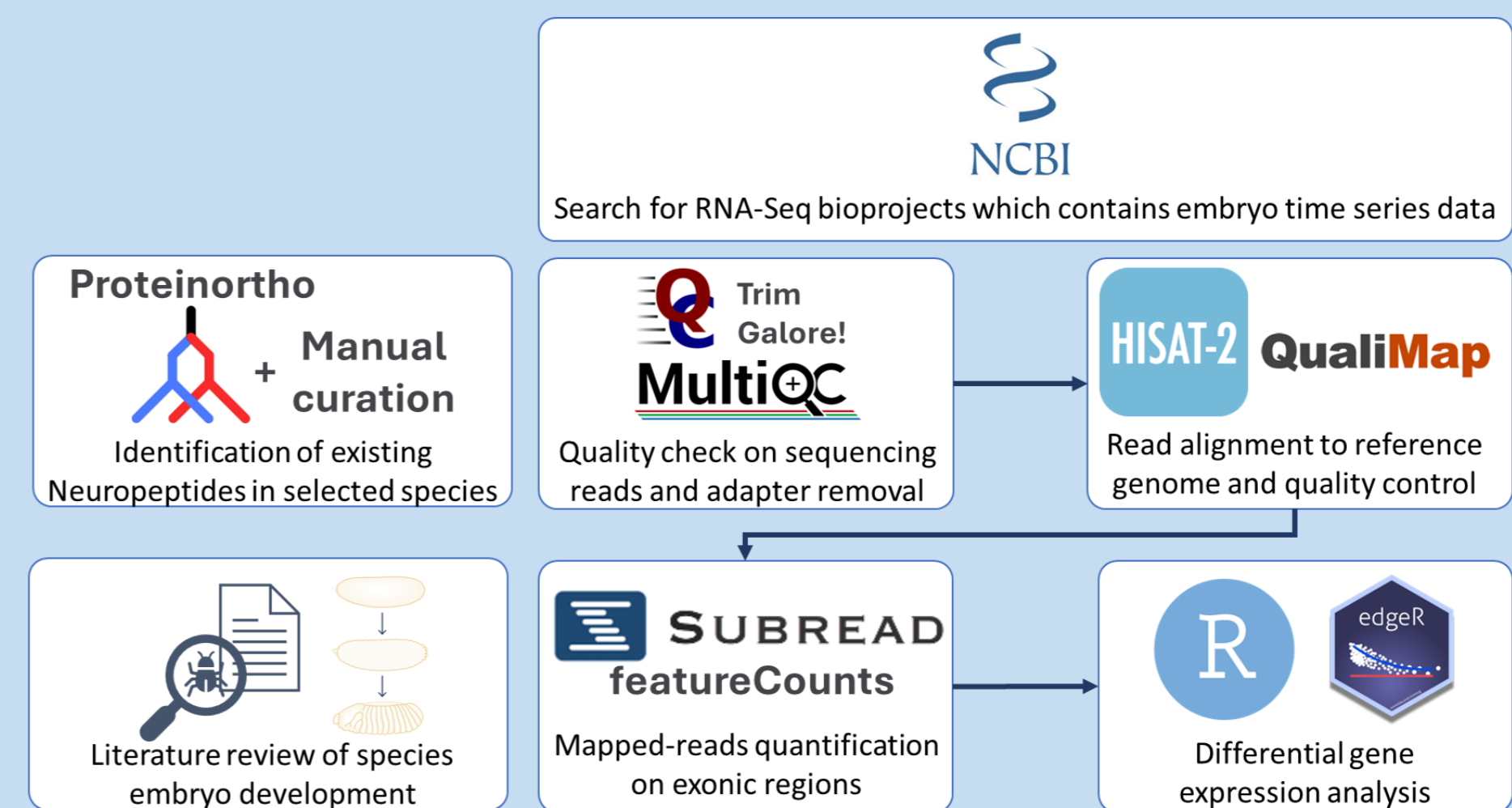
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## INTRODUCTION

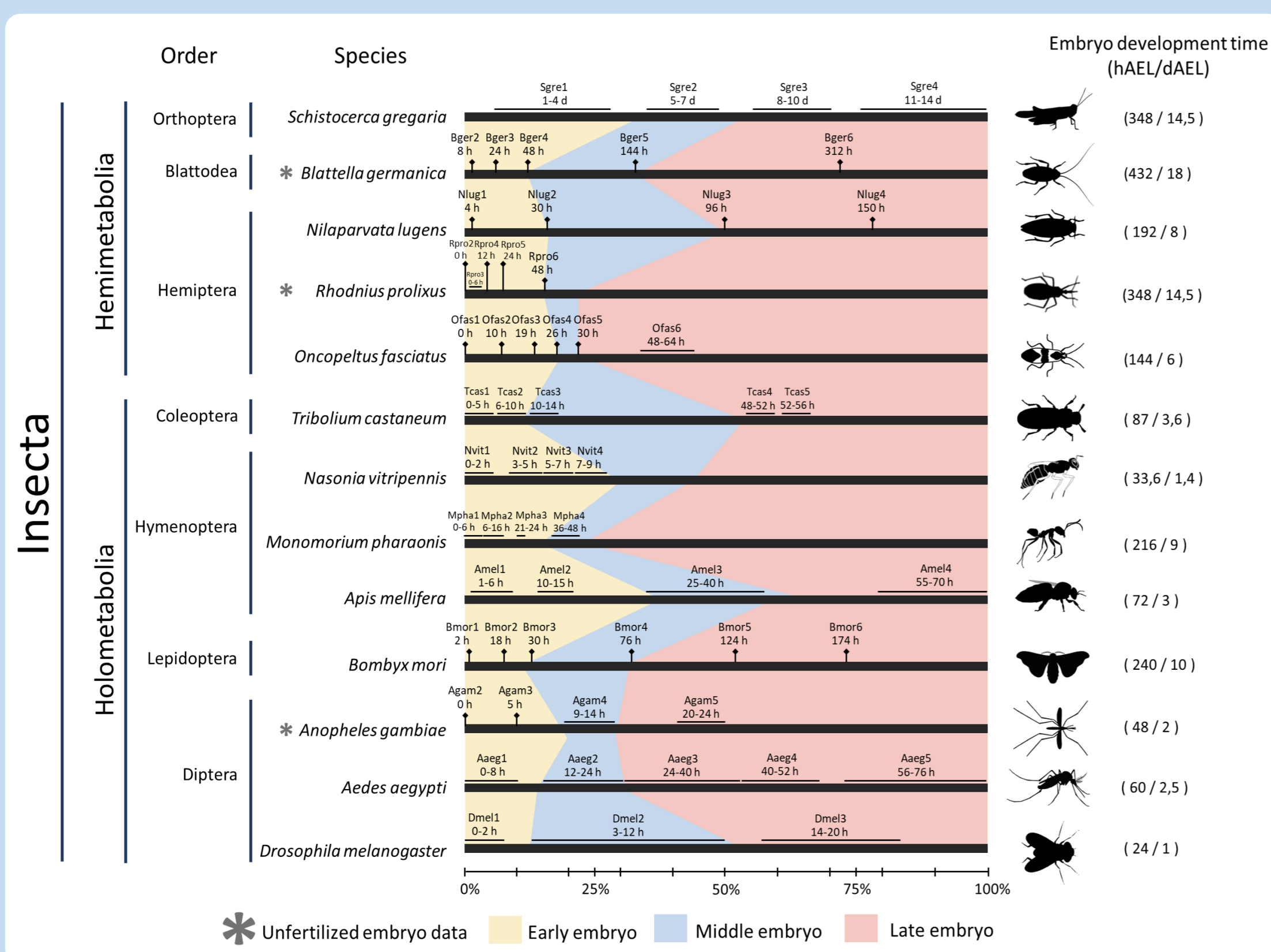
- **Neuropeptides (NPs)** are small peptides that result of the enzymatic processing of larger proteins precursors. They act as neurotransmitters, neuromodulators and growth factors, and participate in feeding, courtship, urination and circadian cycle, among others.
- Examples of NPs in early development are Trunk (*Drosophila melanogaster*) and PTH (*Bombyx mori*), with their receptor Torso.
- Little is known about the expression and function of NP genes during the embryonic development.
- The present work aims to elucidate the presence of NP precursor genes through embryonic development across different species of class Insecta.

## METHODS

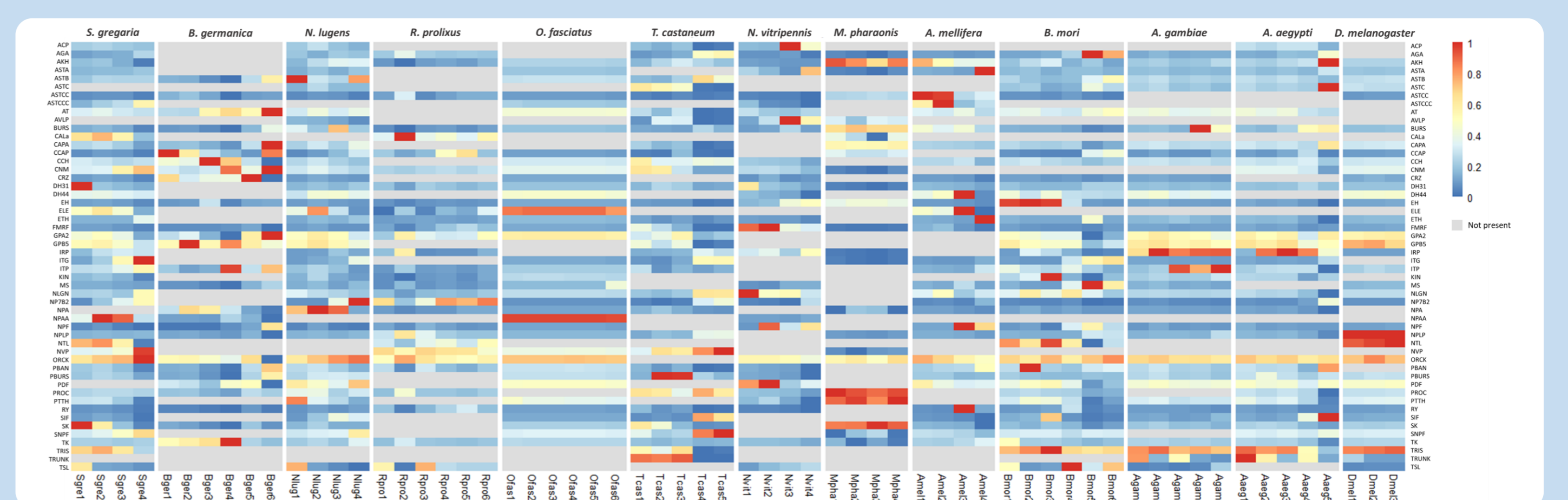
- **Literature review** to align the embryo stages with datasets and allow interspecies comparison.
- **Identification of existing NPs** in each species.
- **RNA-Seq embryo data search** in public databases.
- **DGE assays** to compare NP expression.
- **RT-qPCR** to experimentally detect NPs in *O. fasciatus*.



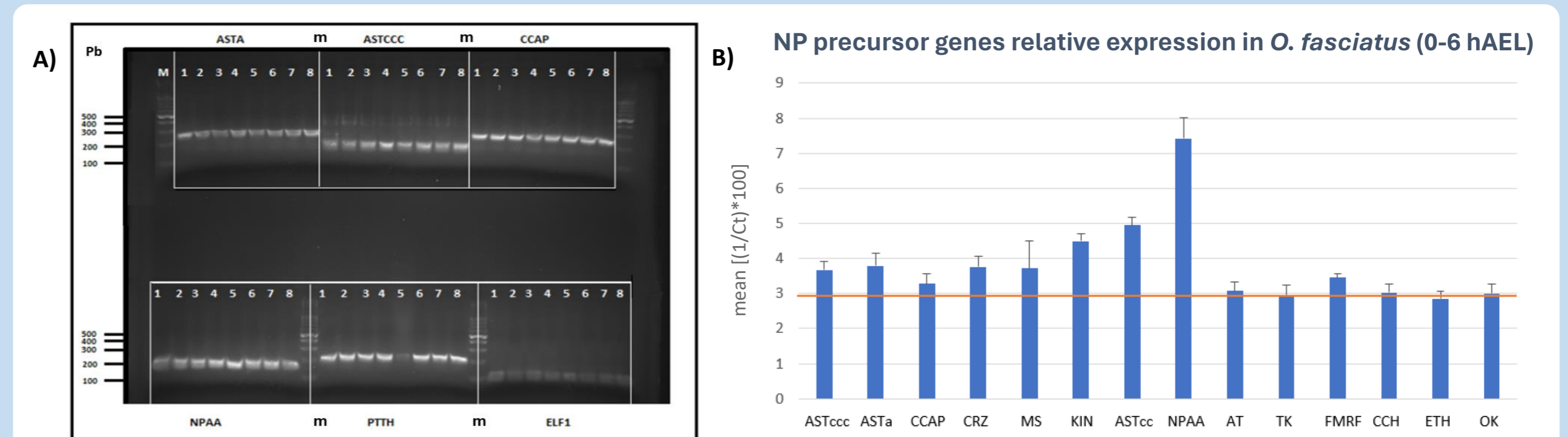
## RESULTS



**Fig. 1.** Alignment of embryo stages of development. The relative position (0-100%) of each RNA-Seq dataset is visualized. The colored regions mark the three main comparable stages across species, early (yellow), middle (light-blue) and late (red) embryo. These stages were established given the blastoderm formation, beginning of gastrulation, and subsequent events, respectively. Unfertilized embryo data in certain species is also showed. To the right, total time of embryo development is indicated in hours/days after egg laying (hAEL/dAEL).



**Fig. 2.** Heatmap of RNA-Seq analysis for 52 selected NP precursor genes. Expression values were measured in transcript per million (TPM) normalized by species (columns), and then scaled by NPs (rows). Colors indicate low (blue), mid (light-yellow) and high (orange-red) expression values, while absent NPs are showed in grey.



**Fig. 3.** NP expression in *Oncopeltus fasciatus* embryo. **A)** gel electrophoresis analysis of RT-qPCR products. 1: 0-24; 2: 0-6; 3: 6-24; 4: 24-48; 5: 48-72; 6: 72-96; 7: 96-120; 8: 120-144 hAEL. **B)** barplot indicating the relative expression levels of NPs at 0-6 hAEL. The horizontal line stands for the transformed cutoff value ( $Ct=35$  or  $[(1/Ct) * 100] = 2.8557$ ).

## CONCLUSIONS AND FUTURE PERSPECTIVES

- We observed neuropeptide expression *in silico* through all the compared embryo stages of development.
- We have experimentally proved the presence of several NPs in early embryo stages of *O. fasciatus*.
- We look forward to search for expression patterns that could infer the existence of totally or partially shared NPs within Orders or presented in all Insecta Class.

## ACKNOWLEDGMENTS

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- To the CCAD for the public availability of the computational clusters used to process all the data.