

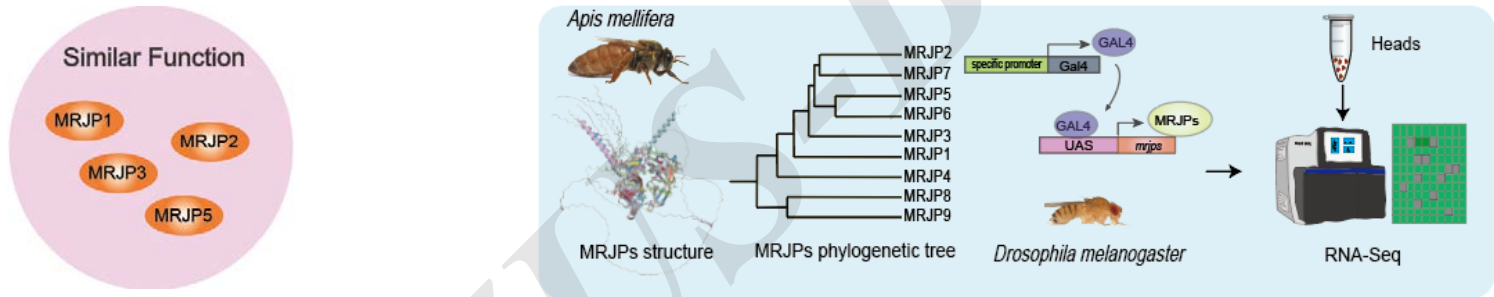
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Ectopic expression of structurally similar major royal jelly proteins reveals their distinct functions in *Drosophila*

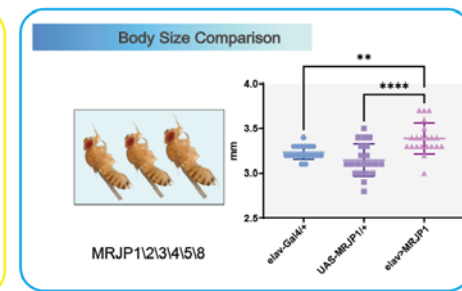
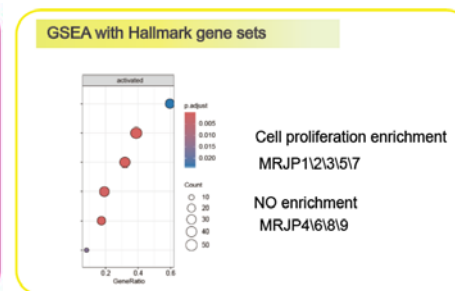
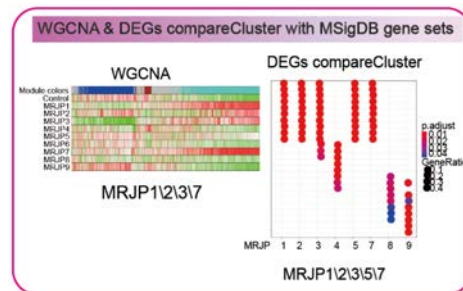
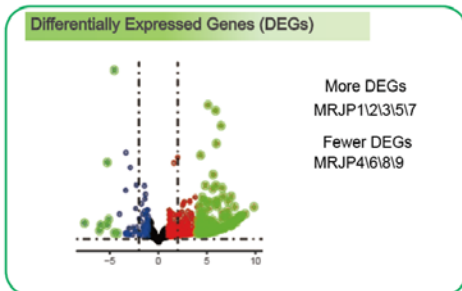
Key words: *Drosophila melanogaster*; Major royal jelly protein (MRJP); Ectopic expression

Research Summary

This study expressed structurally similar MRJPs 1-9 in *Drosophila* neurons, revealing that MRJP1/2/3/5 activate proliferation pathways and increase body size, while others show minimal effects, demonstrating their functional diversity.

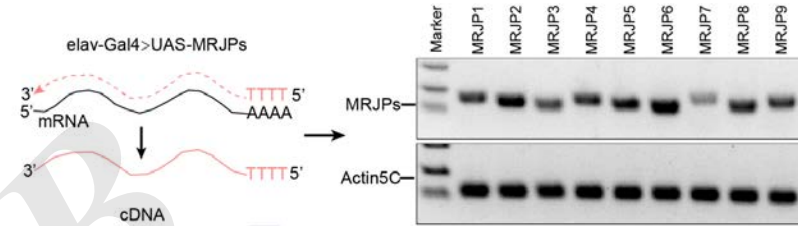


- Comparative analysis of protein structures
- Construction and expression analysis of transgenic *Drosophila*
- DEGs of different MRJPs overexpression *Drosophila*
- Gene set enrichment comparison analysis
- Body size of *Drosophila* following neuronal overexpression of different MRJPs

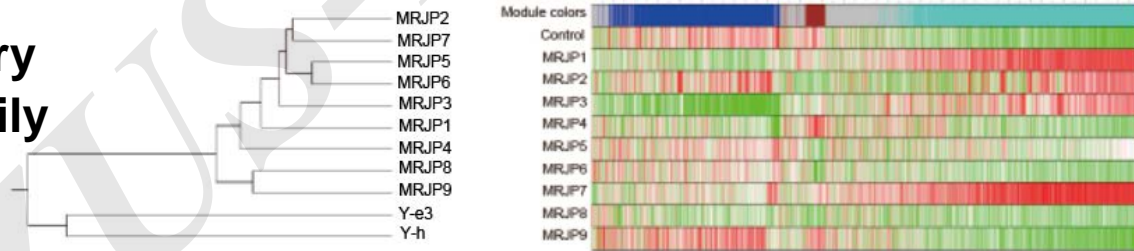


Innovation points

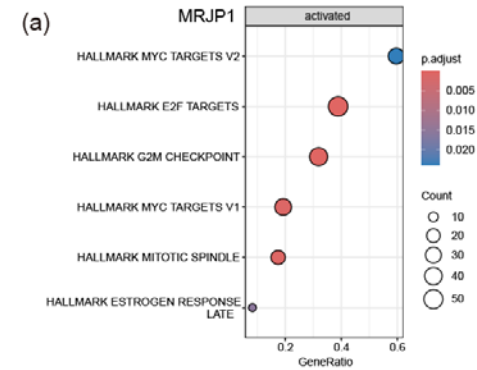
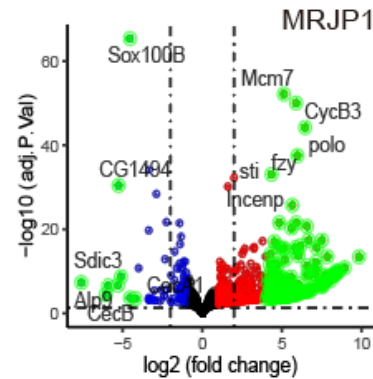
- **Constructed** nine *Drosophila* models for ectopic expression of distinct MRJPs.



- **Confirmed** the evolutionary relationships among MRJP family members.



- **Revealed** distinct gene expression profiles and signaling pathway disruptions caused by overexpression of specific MRJPs.



Innovation points

A series of comprehensive figures and tables were generated to elucidate the potential functional variations of MRJPs in the brain.

Figure 1 | Comparative analysis of protein sequences and tertiary structures of different MRJPs.

Figure 2 | Construction and expression analysis of transgenic *Drosophila* with different UAS-MRJPs.

Figure 3 | Differentially expressed genes following the overexpression of different MRJPs in neurons of *Drosophila* heads

Figure 4 | Co-expression network analysis and gene enrichment comparison analysis of head transcriptomes in different MRJP overexpression and control groups.

Figure 5 | Gene set enrichment analysis of Hallmark gene sets in different MRJP overexpression groups.

Figure 6 | Changes in the body size of *Drosophila* following neuronal overexpression of different MRJPs.

Innovation points

A series of comprehensive figures and tables were generated to elucidate the potential functional variations of MRJPs in the brain.

Figure S1 | Comparative analysis of genomic structures for different MRJPs.

Figure S2 | Comparison of the tertiary structure of monomeric MRJP1 predicted by AlphaFold3 with the reported tertiary structure of multimeric MRJP1.

Figure S3 | Principal component analysis of transcriptomic data comparing overexpression of different MRJPs in the brain to the control group.

Figure S4 | Changes in body size of *Drosophila* following whole-body overexpression of different MRJPs.

Figure S5 | Changes in body size of *Drosophila* following fat body overexpression of different MRJPs

Table S1 | Primers used for genes amplification

Table S2 | Gene and protein sequences of MRJPs