

# Gene expression responses to crowding in *Drosophila*

## 1. Background

- ➔ Density-dependent effects limit the population development of all organisms, although the effects of density dependence upon gene expression are poorly understood.
- ➔ Microarrays offer the opportunity to quantify ecological effects on global gene expression.
- ➔ *Drosophila melanogaster* is frequently used as a model organism for the innate immune system and is reared in crowded conditions.



## 2. Objectives

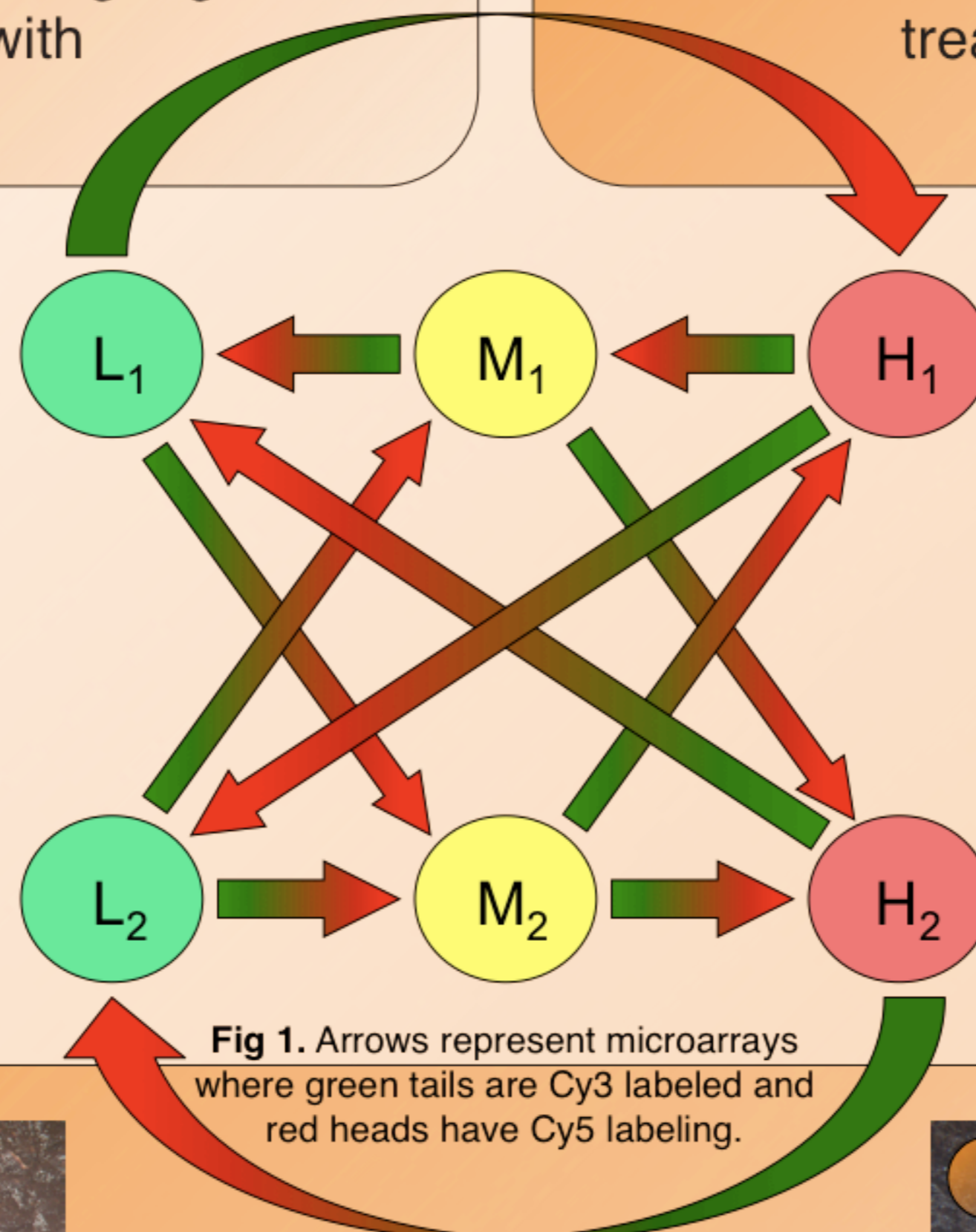
- ➔ Induce stress in *Drosophila melanogaster* through larval crowding before using microarrays to determine any subsequent changes in gene expression.
- ➔ Perform quantitative PCR on selected target genes to study the effect of pathogen challenge, with focus on the innate immune system.

## 3. Lab Work

- ➔ High, medium and low densities of breeding adult pairs were given 24 hours to lay eggs.
- ➔ These eggs counted prior to being left to incubate.
- ➔ The numbers of individuals at each life stage in each treatment were recorded at several time points for growth rate comparisons.

## 4. Experimental Design

- ➔ Two biological replicates used in microarray experiments.
- ➔ Pooled samples of ten third instar larvae used for analysis on 12 microarrays. (Fig. 1)
- ➔ Agilent 60-mer two-colour oligo arrays used for higher quality results than cDNA arrays.



## 5. Results

- ➔ Crowding the larvae resulted in differential growth rates. (Fig. 2)
- ➔ qPCR was performed on target genes including *Actin 79B*, *PPO*, *HSP70*, *Drosomycin* and *DCV1*.
- ➔ *Actin 79B* and *HSP70* were up-regulated in newly emerged (48hr) against week old adults.

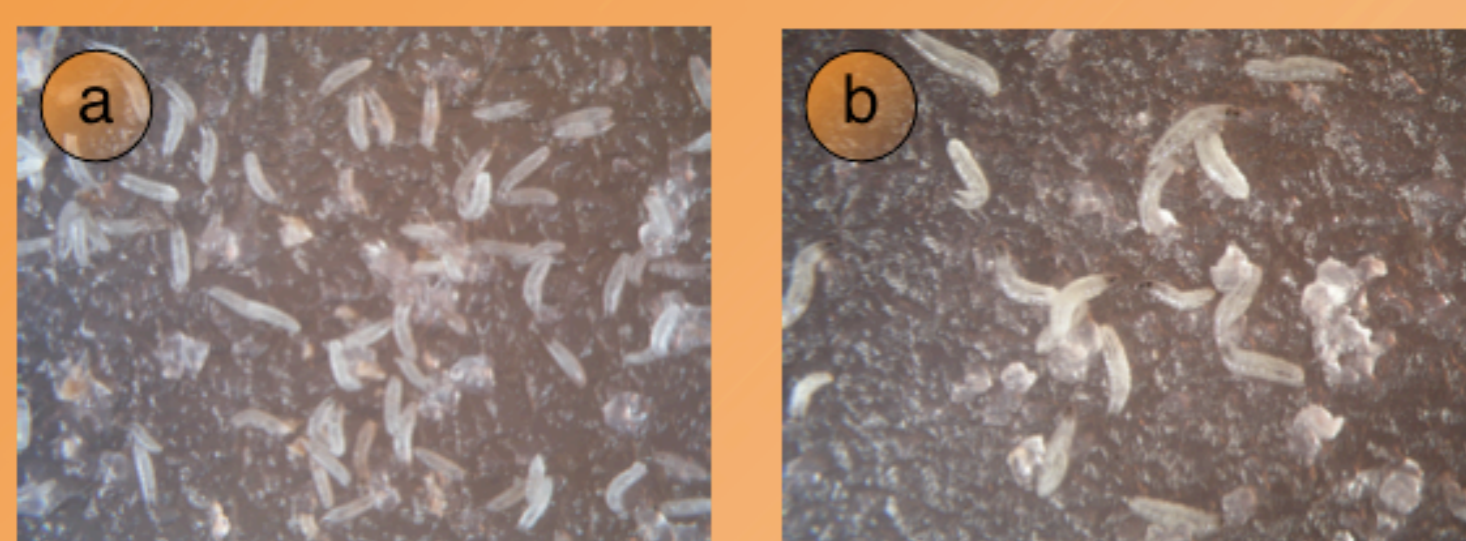
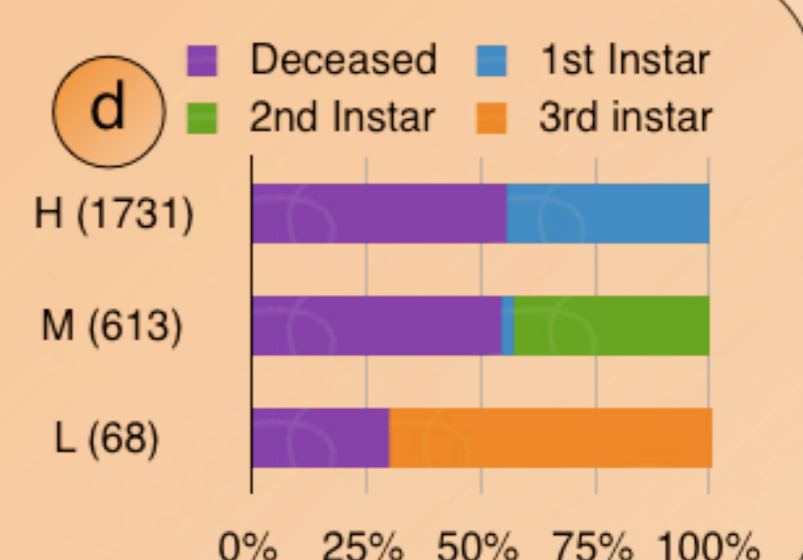


Fig 2. Larvae six days after the removal of adults from cultures - all at the same magnification (a) high; (b) medium; (c) low density treatments. (d) chart showing survival of eggs laid (number in parentheses) to various stages after 1 week.

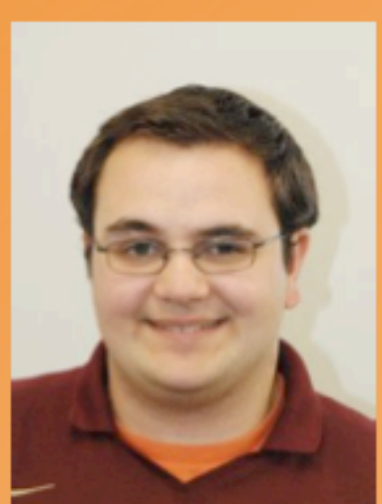


## 6. Future Work

- ➔ A time course of gene expression for each of the samples would show variation in expression from the first larvae through to the adult flies across densities.
- ➔ Intra-specific competitions within life-stages can be developed further, leading to Leslie Matrix models.
- ➔ An extension to work on the honey bee *Apis mellifera* is underway, relating to colony collapse disorder.

## 7. References

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- ➔ Sorensen, J.G. and Loeschke, V. (2001) Larval crowding in *Drosophila melanogaster* induces HSP70 expression, and leads to increased adult longevity and adult thermal stress resistance. *Journal of Insect Physiology*, 47, 1301-1307.
- ➔ Barker, J.S.F. (1972) Adult Population Density, Fecundity and Productivity in *Drosophila melanogaster* and *Drosophila simulans*. *Oecologia*, 11, 83-92.
- ➔ Yang, Y.H. and Speed, T. (2002) Design issues for cDNA Microarray Experiments. *Nature - Genetics Reviews*, 3, 579-588.



Poster and project work by:  
Robert Mark Gardner  
r.m.gardner@warwick.ac.uk  
Systems Biology MSc Student



Project supervised by:

Dave Chandler, Kevin Moffat, Jim Bull,  
Eugene Ryabov and Cunjin Zhang

