# Normalization Protocol for Sørensen et al.

# James C Costello<sup>1,2</sup>, Mehmet M Dalkilic<sup>2,3</sup>, Justen R Andrews<sup>1,2,3</sup>

<sup>1</sup>Dept. of Biology, <sup>2</sup>School of Informatics, <sup>3</sup>Center for Genomics and Bioinformatics Indiana University, Bloomington, IN

## **Data Acquisition**

Data were downloaded from Gene Expression Omnibus (GEO) as raw Affymetrix .CEL files under the accession id, GSE5147:

http://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE5147

#### All data are referenced in:

Sørensen JG, Nielsen MM, Kruhøffer M, Justesen J et al. *Full genome gene expression analysis of the heat stress response in Drosophila melanogaster*. Cell Stress Chaperones. 2005.10(4):312-28.

## **Affymetrix Normalization Protocol**

Data for this dataset were normalized by the authors with GCRMA [1].

# **Affymetrix Presence/Absence calls**

No presence/absence call information or raw data files were supplied with the dataset and therefore presence/absence calls are not included.

### Mapping Affymetrix Probe Sets to FlyBase Annotated Genes

*Drosophila melanogaster* sequences were downloaded from FlyBase<sup>1</sup>. All sequences associated with the printed probes were searched against the version 5 genome assembly of the *D. melanogaster* genome using BLASTn (E-value < 10<sup>-3</sup>). BLAST results were processed with custom Perl scripts. The physical coordinates of transcripts associated with a FlyBase annotated gene were from version 5.10 of the *D. melanogaster* genome annotation. An Affymetrix probe set was mapped to a FlyBase gene ID if the BLAST results mapped to transcripts from a unique gene and only one gene. Strandedness was also considered.

#### **Merging Probe Sets Across Replicate Arrays**

There are multiple hybridizations done for the same condition and the intensity values are merged by simply taking the average for a particular probe set across the arrays under the same condition.

#### References

<sup>&</sup>lt;sup>1</sup> ftp://www.flybase.net/genomes/Drosophila melanogaster/

1. Wu, Z. and R.A. Irizarry, *Preprocessing of oligonucleotide array data*. Nature Biotechnol. 2004. **22**(6):656-658.