Supplementary Material

Gene expression in diapausing rotifer eggs in response to divergent environmental predictability regimes

Eva Tarazona^{1§}, J. Ignacio Lucas-Lledó^{1§}, María José Carmona¹ & Eduardo M. García-Roger¹*

¹ Institut Cavanilles de Biodiversitat i Biologia Evolutiva, Universitat de València, Valencia, Spain.

- * Corresponding author: eduardo.garcia@uv.es
- [§] Both authors contributed equally to this work.

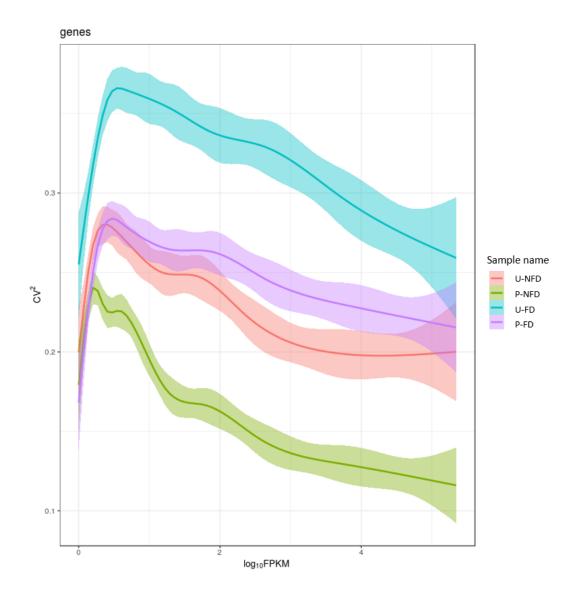


Figure S1. RPKM density distribution plot (the squared coefficient of variation (CV^2) vs log₁₀FPKM) per selective regime and diapause condition.

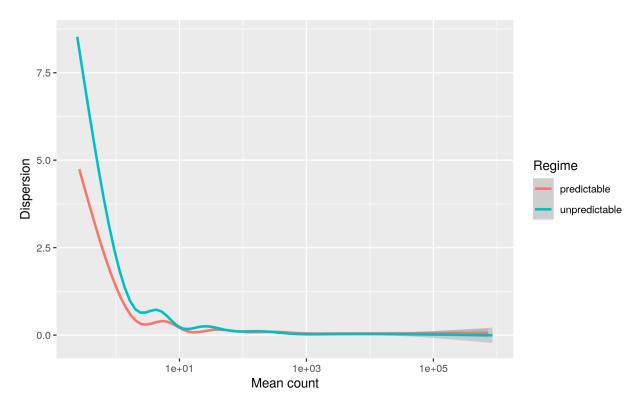


Figure S2. Estimated relationship between biological variation in gene expression between replicates (dispersion) and average level of expression among replicates, measured by a normalized number of counts. Estimates of gene-wise dispersions were obtained using the *estimateDispersion()* function of the *DESeq2* package, separately for samples from the two selective regimes.

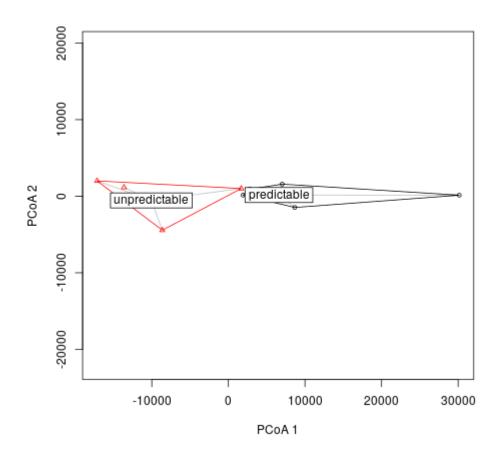
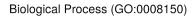
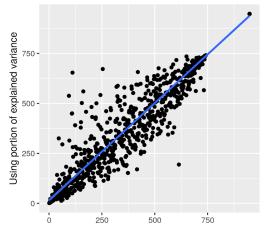


Figure S3. Principal coordinates of gene expression profiles based on Poisson distances. Distances of samples to their regime-specific centroid are not significantly different between selective regimes (p-value = 0.883).





Molecular Function (GO:0003674)

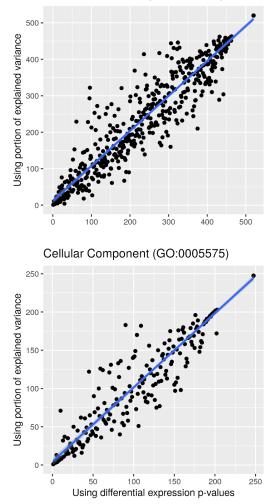


Figure S4. Ordination of gene ontology (GO) terms according to their significance in the enrichment analyses based on either the differential expression *p*-values or the portion of variance explained by regime.

Table S1. Gene Ontology (GO) specific terms for biological process (GO:0008150) significantly associated with selective regime according to all three algorithms used in enrichment analysis. *p*-values are derived from Kolmogorov-Smirnov tests.

GO ID	GO Term	<i>p</i> -value
GO:0055085	transmembrane transport	< 0.001
<u>GO:0006508</u>	proteolysis	< 0.001
<u>GO:0055114</u>	oxidation-reduction process	< 0.001
<u>GO:0007186</u>	G protein-coupled receptor signalling pathway	< 0.001
<u>GO:0006468</u>	protein phosphorylation	< 0.001
<u>GO:0006813</u>	potassium ion transport	< 0.001
<u>GO:0006355</u>	regulation of transcription, DNA-templated	< 0.001
GO:0006289	nucleotide-excision repair	< 0.001
<u>GO:0034220</u>	ion transmembrane transport	0.001
<u>GO:0006470</u>	protein dephosphorylation	0.001
<u>GO:0006812</u>	cation transport	0.002
<u>GO:0003341</u>	cilium movement	0.002
<u>GO:0060271</u>	cilium assembly	0.002
<u>GO:0005992</u>	trehalose biosynthetic process	0.003
<u>GO:0007018</u>	microtubule-based movement	0.003
<u>GO:0006367</u>	transcription initiation from RNA polymerase II promoter	0.004
<u>GO:0046039</u>	GTP metabolic process	0.004
<u>GO:0046129</u>	purine ribonucleoside biosynthetic process	0.004
<u>GO:0007160</u>	cell-matrix adhesion	0.005
<u>GO:0070836</u>	caveola assembly	0.006
<u>GO:0006821</u>	chloride transport	0.007
<u>GO:0006979</u>	response to oxidative stress	0.007
GO:0043161	proteasome-mediated ubiquitin-dependent protein catabolic process	0.008
<u>GO:0051260</u>	protein homooligomerization	0.008
<u>GO:0008272</u>	sulphate transport	0.009

Table S2. Gene ontology (GO) specific terms for molecular function (GO:0003674) significantly associated with selective regime according to all three algorithms used in enrichment analysis. *p*-values are derived from Kolmogorov-Smirnov tests.

GO ID	GO Term	<i>p</i> -value
<u>GO:0005515</u>	protein binding	< 0.001
<u>GO:0005509</u>	calcium ion binding	< 0.001
<u>GO:0003774</u>	motor activity	< 0.001
<u>GO:0004930</u>	G protein-coupled receptor activity	< 0.001
<u>GO:0004252</u>	serine-type endopeptidase activity	< 0.001
<u>GO:0005524</u>	ATP binding	< 0.001
<u>GO:0004672</u>	protein kinase activity	< 0.001
GO:0022857	transmembrane transporter activity	< 0.001
GO:0042626	ATPase activity, coupled to transmembrane movement of substances	0.001
<u>GO:0004198</u>	calcium-dependent cysteine-type endopeptidase activity	0.001
<u>GO:0005525</u>	GTP binding	0.001
<u>GO:0061630</u>	ubiquitin protein ligase activity	0.001
<u>GO:0004181</u>	metallocarboxypeptidase activity	0.001
<u>GO:0005230</u>	extracellular ligand-gated ion channel activity	0.002
<u>GO:0005249</u>	voltage-gated potassium channel activity	0.002
<u>GO:0005507</u>	copper ion binding	0.003
<u>GO:0051015</u>	actin filament binding	0.003
<u>GO:0016840</u>	carbon-nitrogen lyase activity	0.004
<u>GO:0004550</u>	nucleoside diphosphate kinase activity	0.005
GO:0004601	peroxidase activity	0.005
<u>GO:0030246</u>	carbohydrate binding	0.005
<u>GO:0005200</u>	structural constituent of cytoskeleton	0.006
<u>GO:0016491</u>	oxidoreductase activity	0.007
<u>GO:0003777</u>	microtubule motor activity	0.008
<u>GO:0047631</u>	ADP-ribose diphosphatase activity	0.008

Table S3. Gene ontology (GO) specific terms for cellular component (GO:0005575) significantly associated with selective regime according to all three algorithms used in enrichment analysis. *p*-values are derived from Kolmogorov-Smirnov tests.

GO ID	GO Term	<i>p</i> -value
GO:0016020	membrane	< 0.001
GO:0016459	myosin complex	< 0.001
GO:0008076	voltage-gated potassium channel complex	< 0.001
GO:0005887	integral component of plasma membrane	0.001
GO:0005576	extracellular region	0.001
GO:0090575	RNA polymerase II transcription factor complex	0.001
GO:0005634	nucleus	0.003
GO:0005886	plasma membrane	0.003
GO:0005743	mitochondrial inner membrane	0.004
GO:0005929	cilium	0.004
GO:0005741	mitochondrial outer membrane	0.009