Ben Allen Supplementary Files

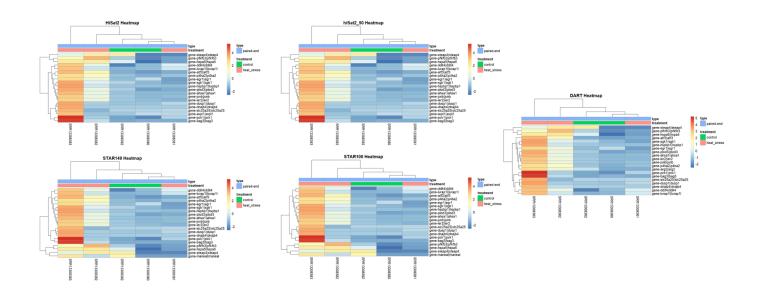


Figure 1. Heatmaps for each modified mapping process displaying genes with the greatest variance of transcript count between the control and heat stress treatments. Experimental trial ID is shown at the bottom of each column with each having its respective treatment group above the column. Expressed genes are shown to the right of each chart, genes were filtered so that only named genes appear in this analysis.

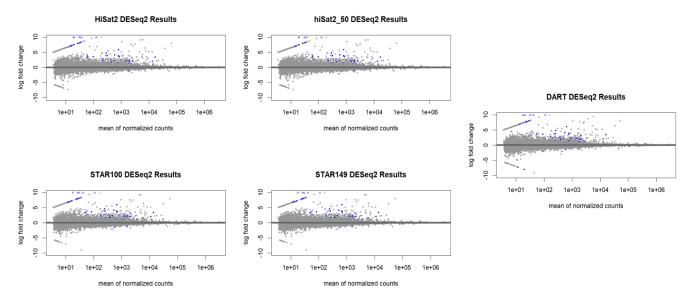


Figure 2. MA plots for each of the modified mapping processes displaying the log2 fold change from each of the respective DDSeq 2 results. Those dots in blue represent genes that significantly (p-value <0.1) up/downregulated. Triangles represent points located out of frame.

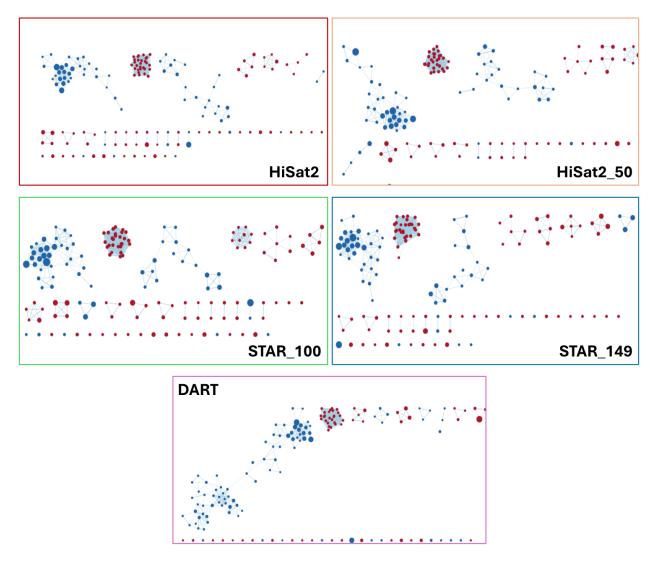


Figure 3. Enrichment Maps for each mapping process generated with Cytoscape. With red nodes representing upregulated genes and blue node downregulated genes. Gene labels have been removed for clarity, fully labeled and interactive enrichment maps can be found in the project GitHub.

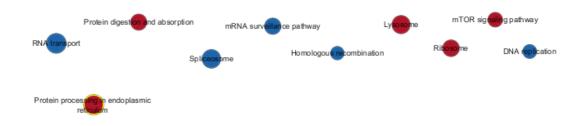


Figure 4. Enrichment Map of KEGG Pathways generated using Cytoscape. With red nodes representing upregulated pathways and blue nodes downregulated pathways. No pathways relating to metabolism were recovered.

Mapping Process	HiSat2	HiSat2_50	STAR_100	STAR_149	DART
Genes Read	17,817	17,817	17,630	17,630	17,514
LFC Upregulated	53	32	27	27	31
LFC Downregulated	2	1	1	1	3
Outliers	15	15	14	14	13
Low counts	346	691	0	0	0

Table 1. Summary data of differentially expressed genes, at a p-value of < 0.05, for each of the mapping processes.

Mapping Process	Top 10 differentially expressed genes by rank								
HiSat2	ULK2	KCNE1	BCO2	ANGPT*	CUNH1*	NEURO*	CXCR4	P4HA2	ELL
	/5.72	/5.71	/5.83	/4.96	/4.51	/4.45	/4.26	/4.24	/4.15
HiSat2_50	ULK2	BCO2	KCNE1	ANGPT*	CUNH1*	NEURO*	CXCR4	P4HA2	ELL
	/5.72	/5.82	/5.14	/4.96	/4.51	/4.45	/4.45	/4.24	/4.14
STAR_100	LIN28A	BCO2	ULK2	KCNE1	ANGPT*	CXCR4	NEURO*	P4HA2	CUNH1*
	/5.89	/5.70	/5.74	/5.62	/4.77	/4.56	/4.36	/4.28	/4.26
STAR_149	LIN28A	BCO2	ULK2	KCNE1	ANGPT*	CXCR4	NEURO*	P4HA2	CUNH1*
	/5.91	/5.70	/5.74	/5.62	/4.77	/4.56	/4.36	/4.28	/4.26
DART	MEGF6	BCO2	ULK2	KCNE1	ANGPT*	CUNH1*	CXCR4	ELL	PDP1
	/9.07	/5.72	/5.51	/5.44	/4.99	/4.38	/4.35	/4.25	/4.20

Table 2. List of top 10 ranked differentially expressed genes for each mapping process, filtered so that only named genes are displayed. Shown as gene name/ rank value. Genes marked with * are abbreviated, full names are as follows: ANGPTL3, CUNH14ORF180, NEUROD2.

Dataset	SRR13308392	SRR13308380	SRR13308381	SRR13308382	SRR13308383
Aligned 0 Times	99.32%	99.22%	99.20%	99.25%	99.16%
Aligned 1 Time	0.68%	0.78%	0.80%	0.75%	0.84%
Aligned >1 Times	0.00%	0.00%	0.00%	0.00%	0.00%
Overall Alignment Rate	0.68%	0.78%	0.80%	0.75%	0.84%

Table 3. Alignment of summary data for each dataset's alignment to the *Paralichthys olivaceus* mitochondrial reference genome.