



Objective

- Withering syndrome is a fatal disease of abalone caused by a rickettsial bacterium (WS-RLO)
- WS-RLO transmission experiments demonstrate progeny from disease selected populations are more resistant to WS-RLO than naïve populations
- GOAL: Characterize and compare the transcriptomes of WS-resistant and naïve abalone to elucidate mechanisms associated with increased disease tolerance

Approach

1. WS-Resistant Exposed
2. WS-Resistant Control
3. Naïve Exposed
4. Naïve Control

De novo assemble & annotate

RNA-Seq & DESeq

Galaxy – join annotation & GO

Enrichment Analysis - DAVID

Visualize data - REVIGO

Discovery: QC

1 Trim summary

Name	Number of reads	Avg.length	Number of reads after trim	Percentage trimmed	Avg.length after trim
Roberts_201 00712_CC_F3	34,887,723	49.6	11,946,848	34.24%	39.7
Roberts_201 00712_CE_F3	32,011,762	49.6	11,540,752	36.05%	40.1
solid0078_20 110412_FRA_G_BC_WHITE_E_WHITE_F3_QV_SC	43,703,122	49.6	30,322,522	69.38%	32.4
solid0078_20 110412_FRA_G_BC_WHITE_E_WHITE_F3_QV_SE	27,927,346	49.6	18,567,800	66.49%	37.4

Losing large # of reads after trimming – trim on length only

1 Trim summary

Name	Number of reads	Avg.length	Number of reads after trim	Percentage trimmed	Avg.length after trim
solid0078_201104112_FRAG_BC_WHITE_WHITE_F3_QV_SE	27,927,346	49.6	27,730,188	99.29%	50.0
solid0078_201104112_FRAG_BC_WHITE_WHITE_F3_QV_SC	43,703,122	49.6	43,367,666	99.23%	50.0
Roberts_20100712_CE_F3	32,011,762	49.6	31,750,064	99.18%	50.0
Roberts_20100712_CC_F3	34,887,723	49.6	34,601,857	99.18%	50.0

Assessing different RNA-Seq software – CLC vs Tophat

Working – Annotating 3 assemblies (blastx)

Lisa Blk Ab Assemblies

	#Contigs	Length #sum	mean	stdev	min (100bp)	25%	50%	75%	Max
CLC Version 6	5490	1622880	295.606	105.578	200	230	262	323	1378
SOAP	27065	4016880	148.416	56.1419	100	111	130	165	896
Minia	9428	1458860	154.737	49.9967	108	121	139	171	613
					(200bp)				
CLC Version 6	5490	1622880	295.606	105.578	200	230	262	323	1378
SOAP	3621	948635	261.981	65.9723	200	217	241	285	896
Minia	1310	332406	253.745	54.9737	200	215	238	273	613

Discovery

(enrichment only on 2 naïve libraries)

Less DEG in naïve WS-RLO exposed animals

Shift from homeostatic processes to catabolic processes (autophagy, cytosis, autolysis)

Consistent with characteristics of withering syndrome so pretty cool 😊

Next Step

Pick “best” assembly & RNA-Seq for DESeq and enrichment analysis on all 4 libraries

EXPOSED

CONTROL

