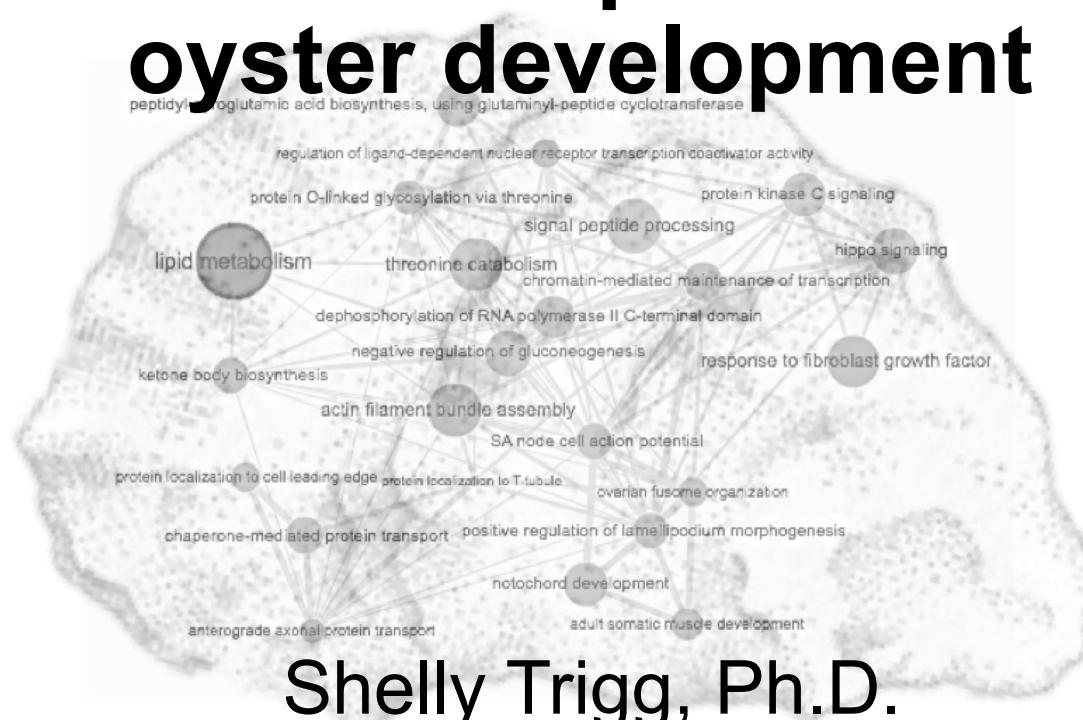


A protein inventory reveals mechanisms of temperature impact on oyster development



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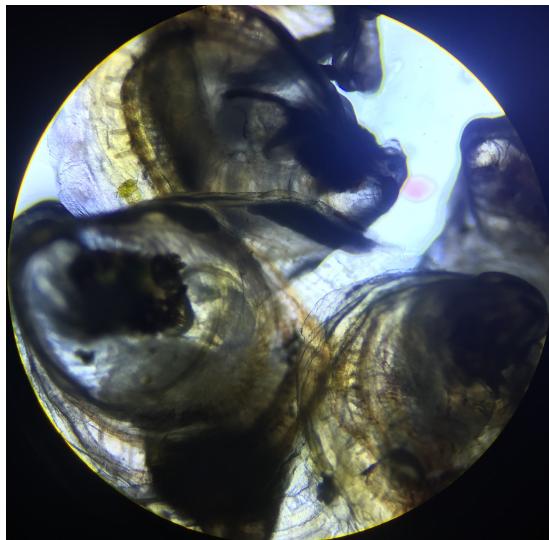
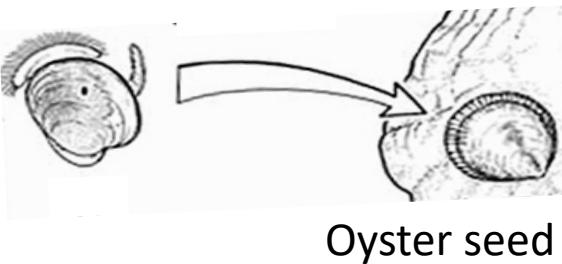
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Oyster (*C.gigas*) seed are delicate



- Pacific oyster are **sensitive** to environmental conditions from larval transition → early stage juveniles
- High mortality typically happens 10-14 days post-set @23C
- Increased rearing temperature (27-29°C) has lead to increased growth rates and survival
- We don't fully understand oyster physiology, why high mortality occurs, or why increased rearing temperature helps
- A thorough molecular baseline of oyster development during this life stage could help:
 1. identify candidate biomarkers for monitoring seed health
 2. mitigate current challenges

Questions

- What fundamental physiology occurs during oyster development?
- How do warmer temperatures affect juvenile development on a global molecular level?

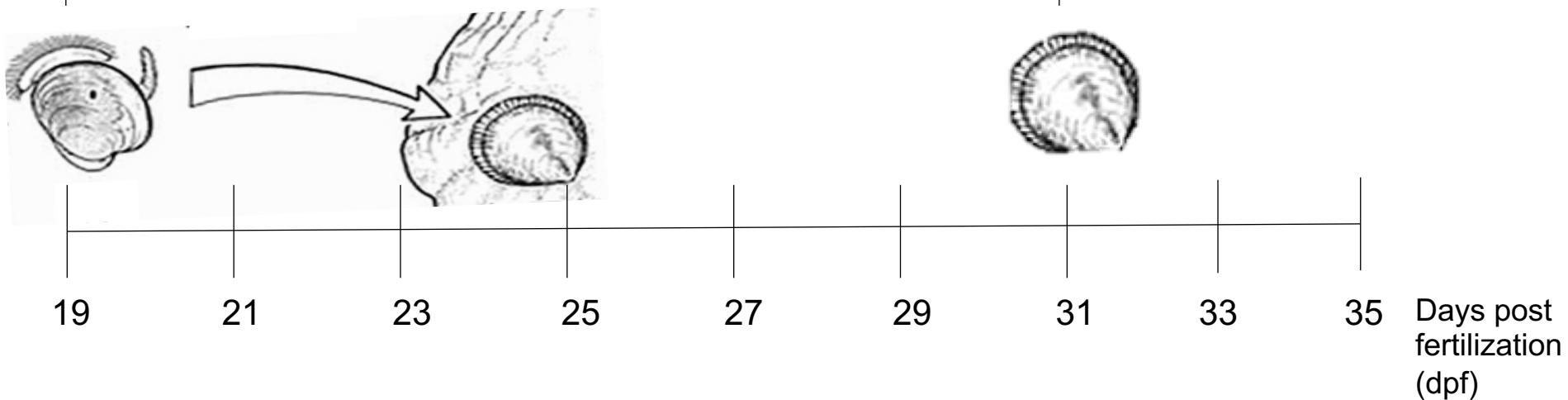
Experimental Design

temperature treatment:

23°C

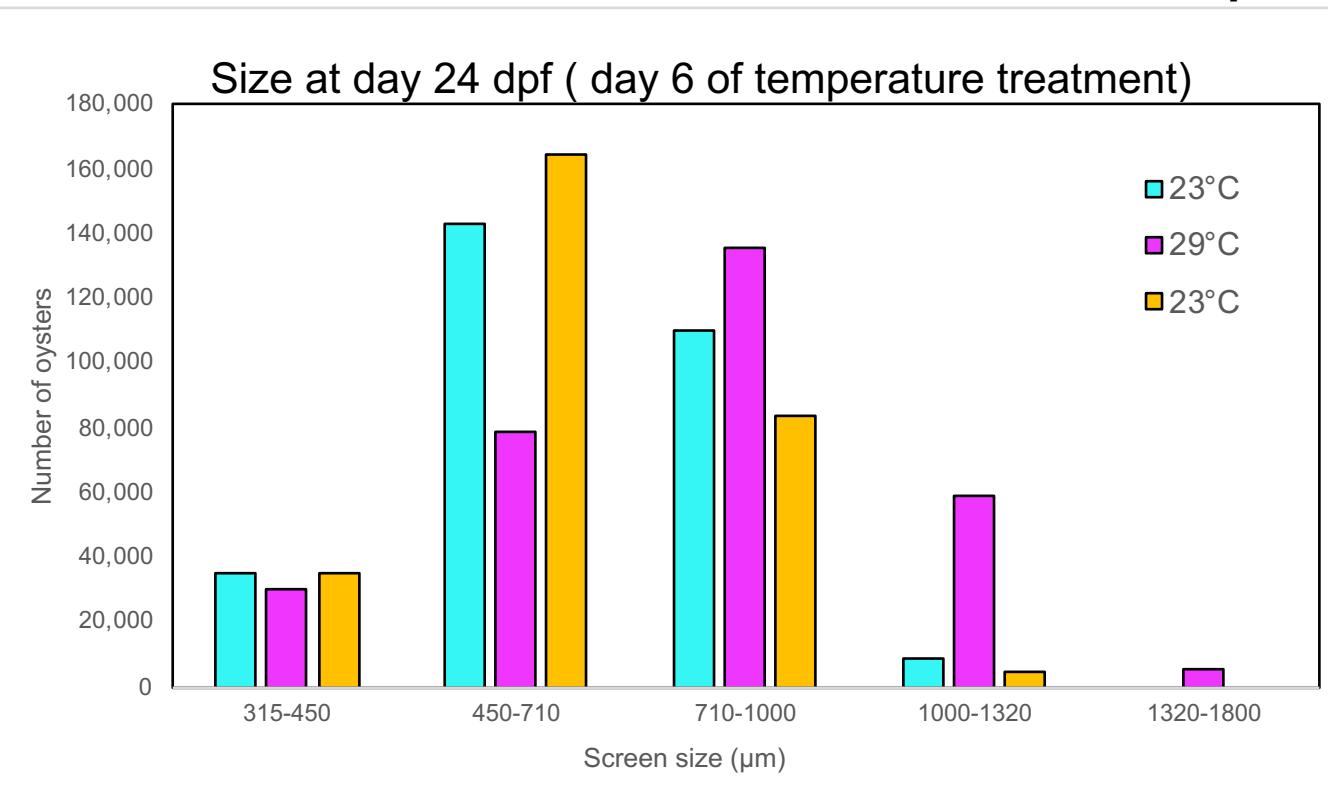
29°C

13 days

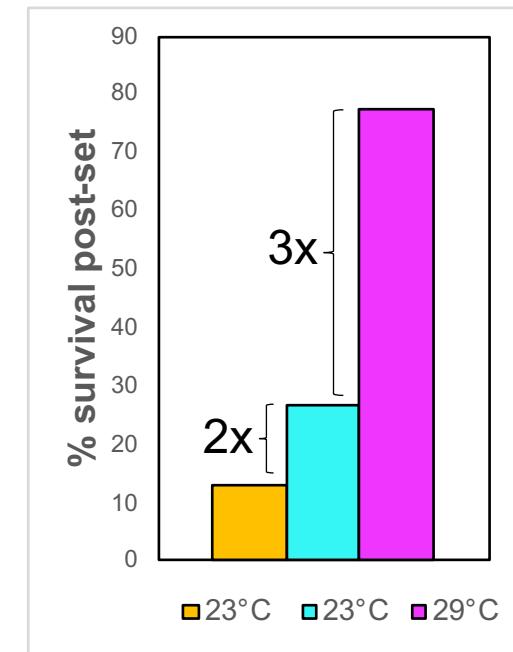


Size and survival differences between temperature groups

Difference in size distribution but not survival at 24 dpf



Difference in survival at 35 dpf



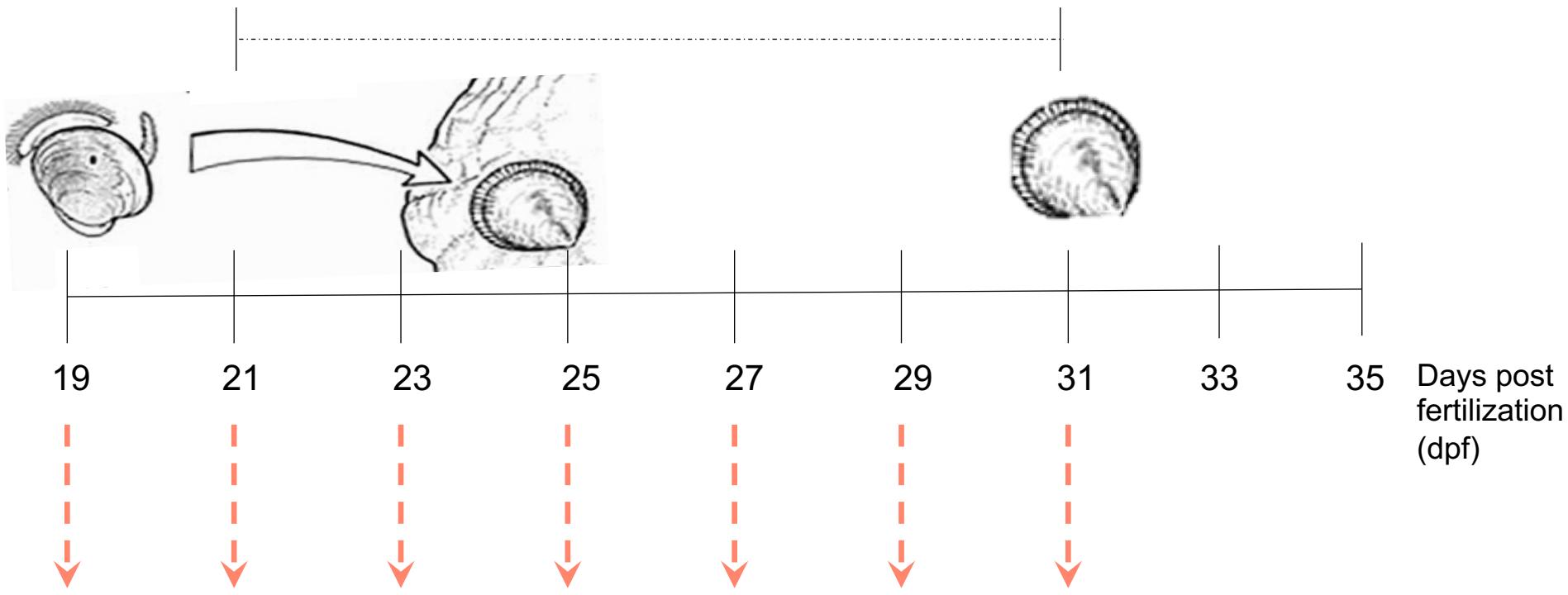
- 29°C has more animals of larger size

Experimental Design

temperature treatment:

23°C

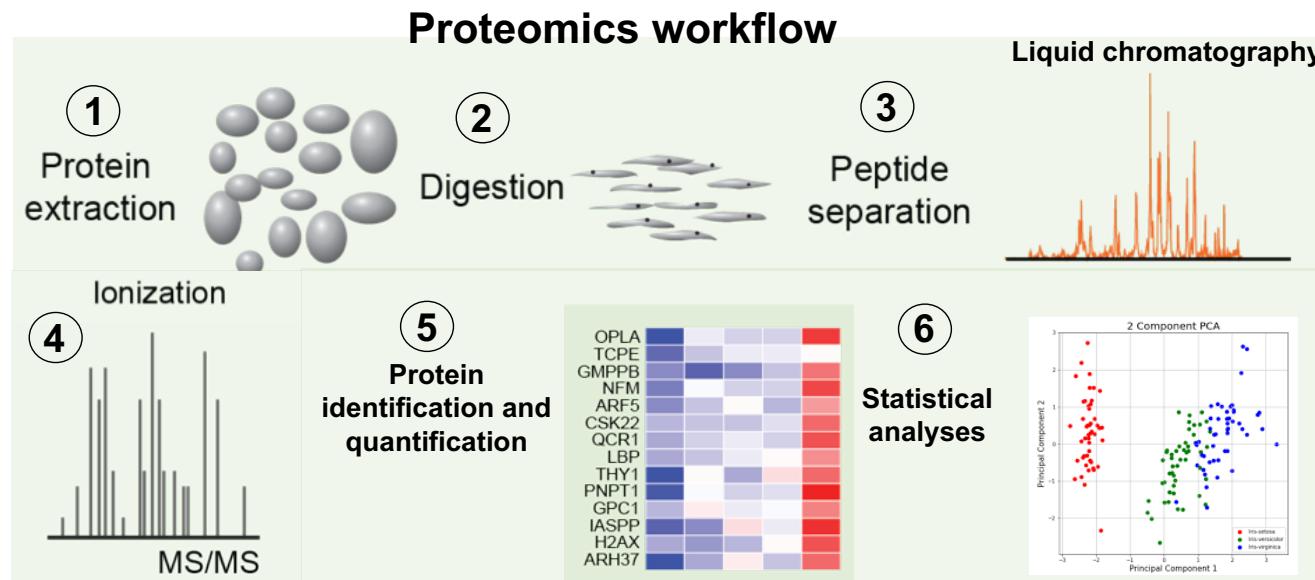
29°C



PROTEOMICS

Proteomics = global protein inventory

- Proteins are the active players in the cell that carry out biological processes
- Non-biased birds eye view of all proteins present and their abundances
 - We can infer what processes are going on
- We can make comparisons between individuals and see what processes may underlie different outcomes



Proteomics comparisons

1. Fundamental development:

Proteins abundances that
commonly change over time

23°C

29°C

2. Temperature effect:
Proteins abundance differences
driven by temperature

23°C

29°C

3. Silo effect:
Protein abundance differences
driven by silo

23°C

23°C

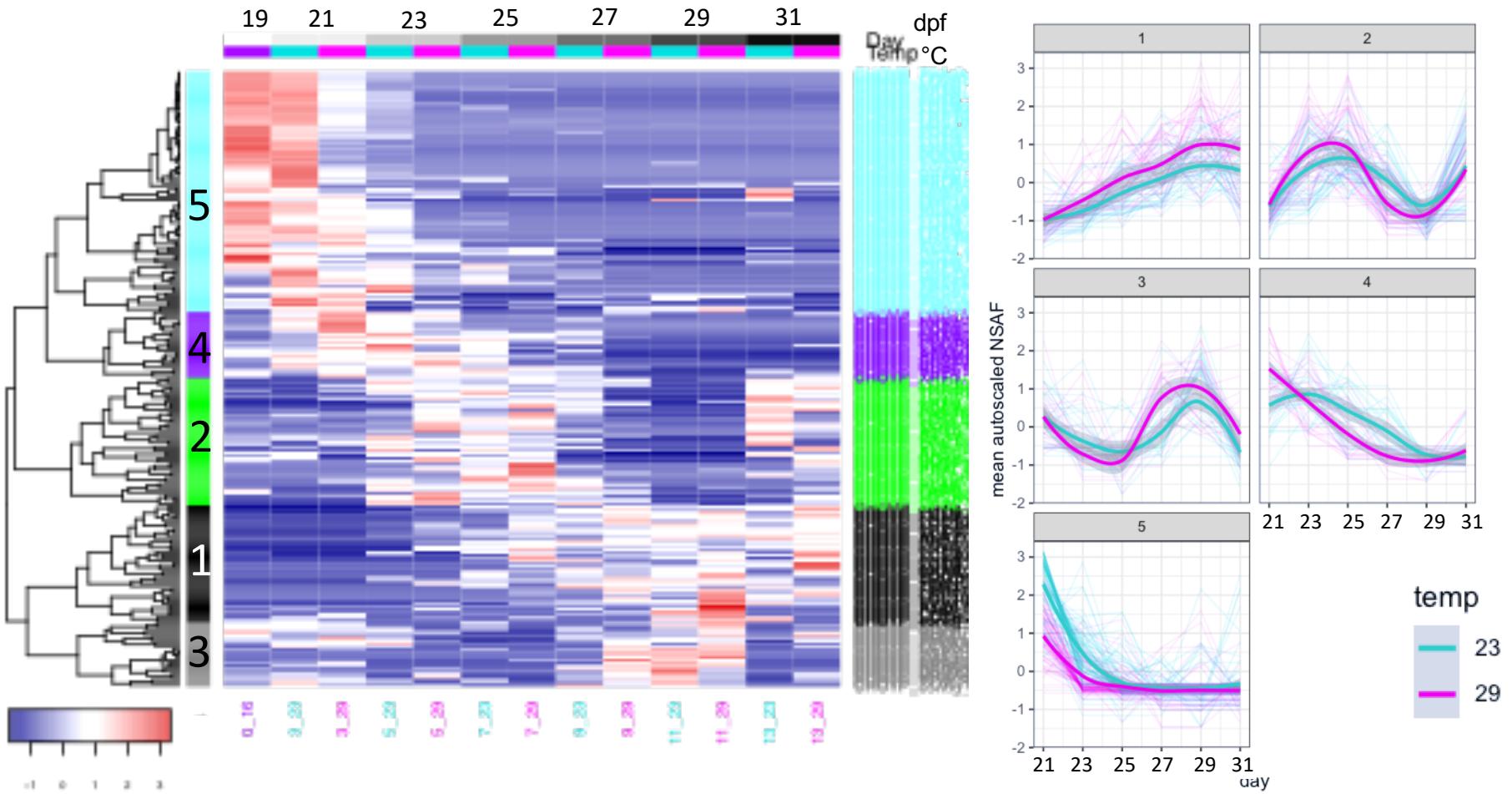
Clustering differentially abundant proteins reveals 5 distinct patterns over time

1. Fundamental development:

Proteins abundances that commonly change over time

23°C

29°C

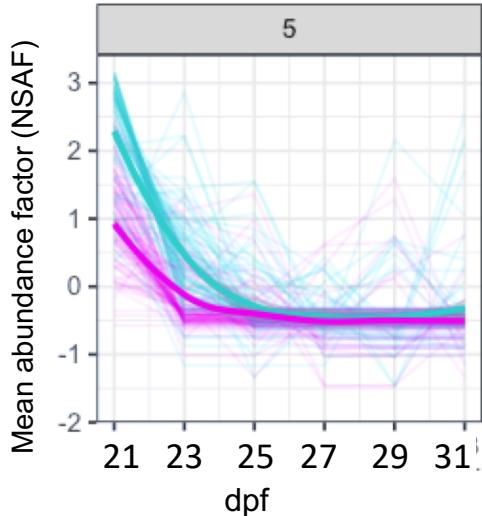


Early developmental processes that sharply decrease over two days

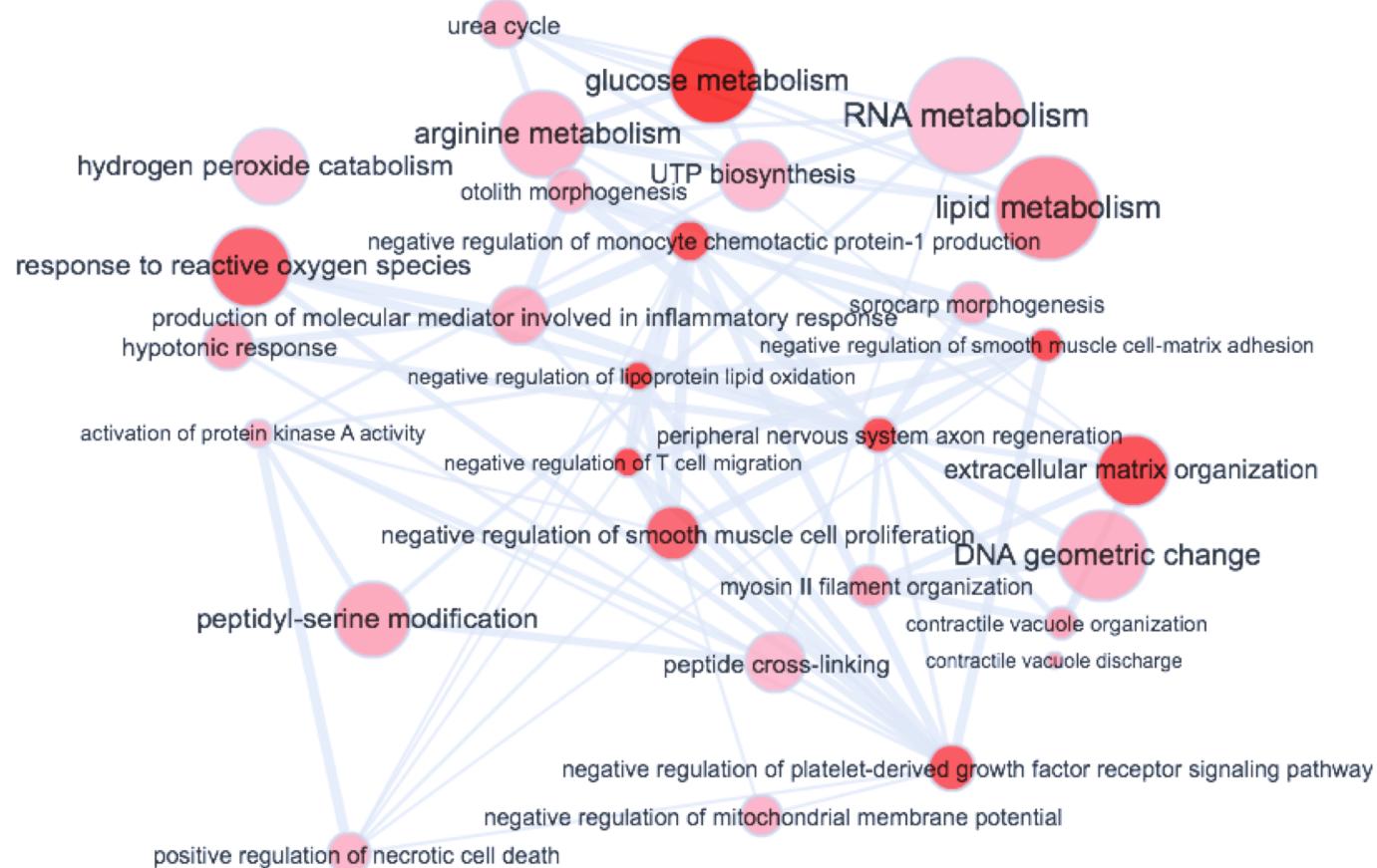
1. Fundamental development:

Proteins abundances that commonly change over time

23°C 29°C



Clade 5

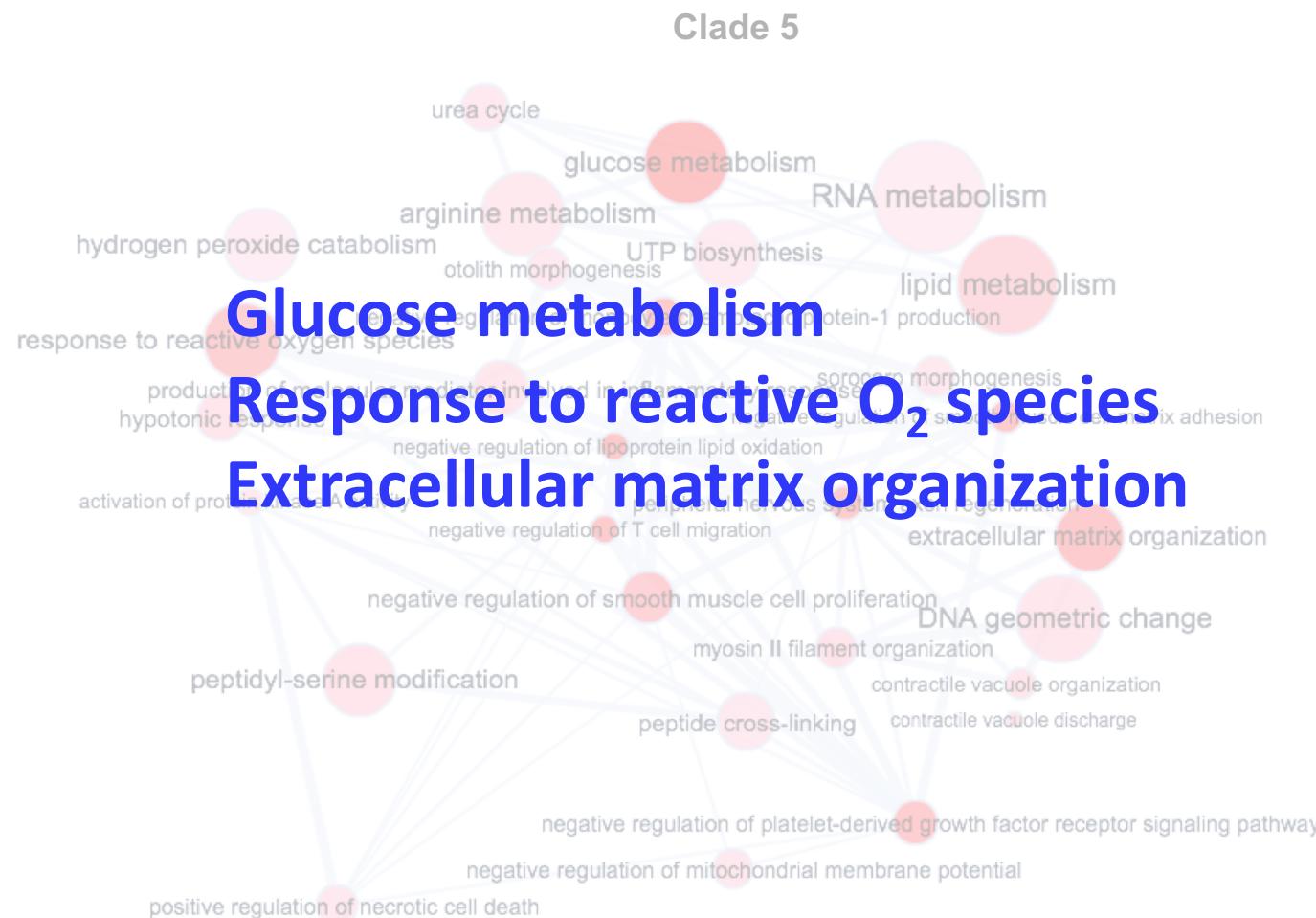
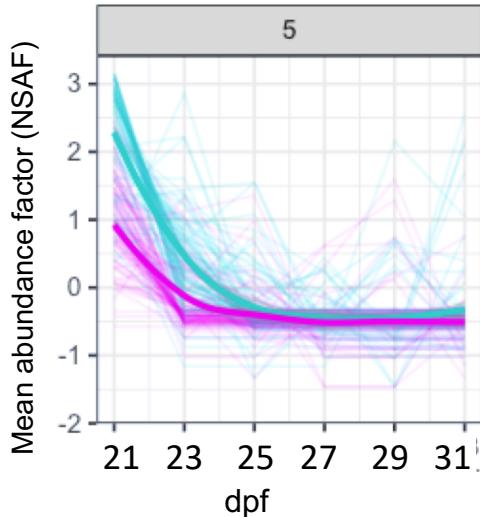


Early developmental processes that sharply decrease over two days

1. Fundamental development:

Proteins abundances that commonly change over time

23°C 29°C



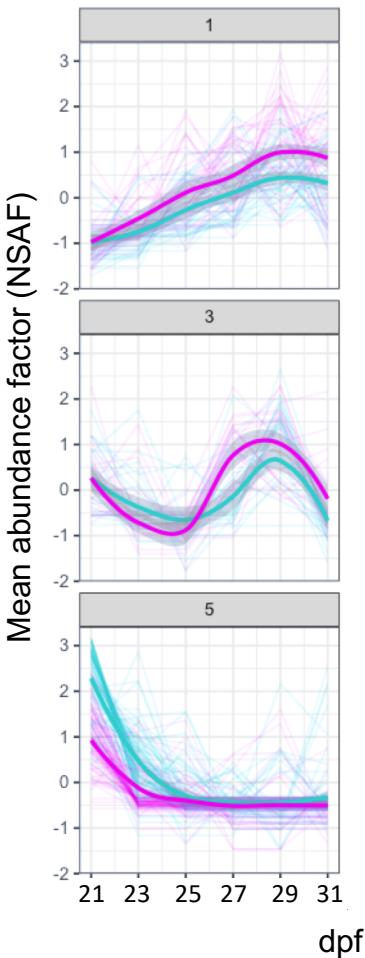
Protein patterns and processes we could expect to see in normal development

1. Fundamental development:

Proteins abundances that commonly change over time

23°C

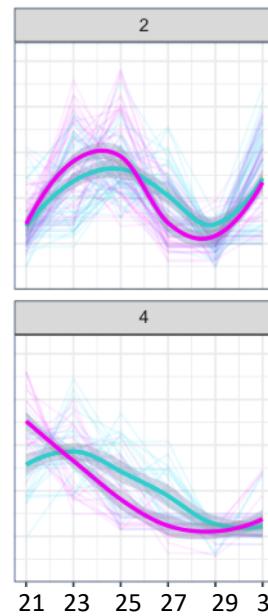
29°C



- Glycolytic process and glucose homeostasis
- Cortical cytoskeleton organization

- Actin cytoskeleton organization
- Synaptic vesicle exocytosis

- Glucose metabolism
- Response to reactive O₂ species
- Extracellular matrix organization



- Regulation of cilium assembly
- Extracellular matrix organization

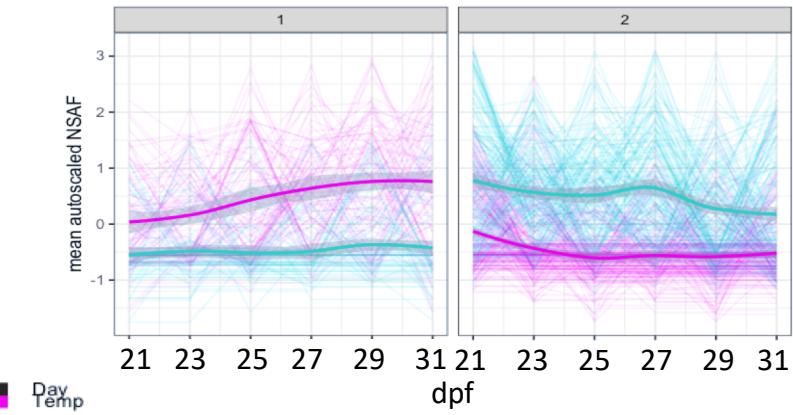
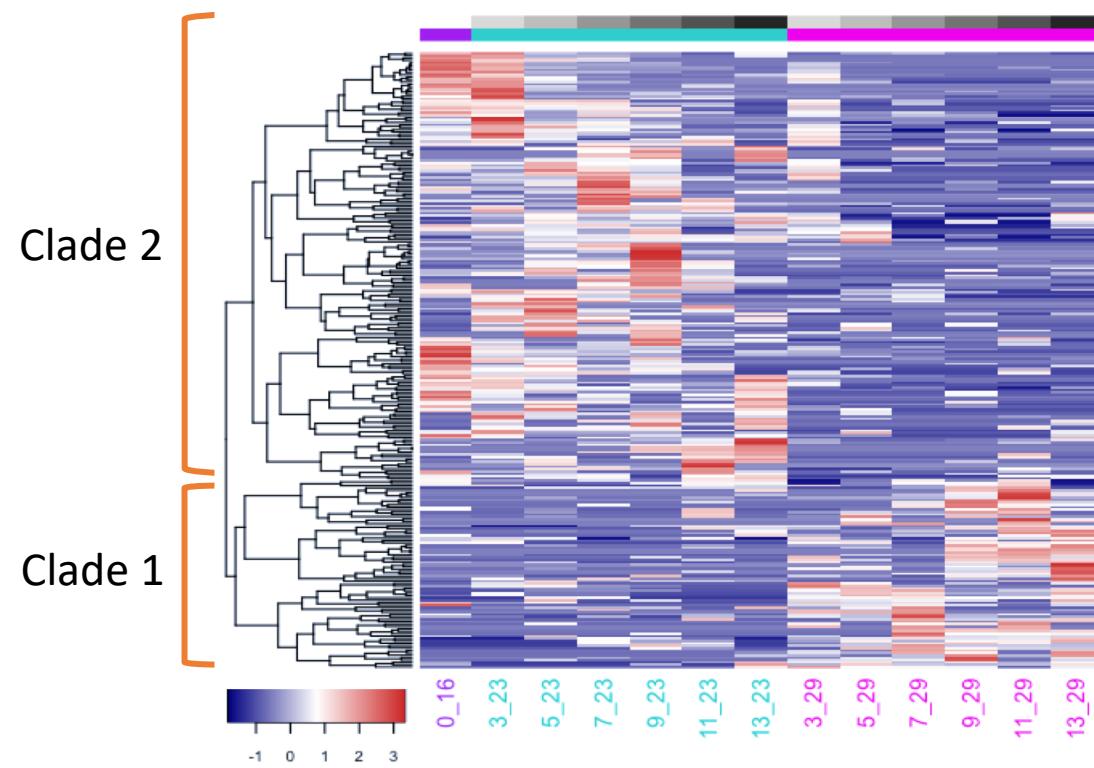
- Stress signaling
- Apoptotic process
- Amino and nucleic acid metabolism

Proteins and processes underlying increased growth and survival at 29°C

2. Temperature effect:

Proteins abundance differences driven by temperature

23°C 29°C



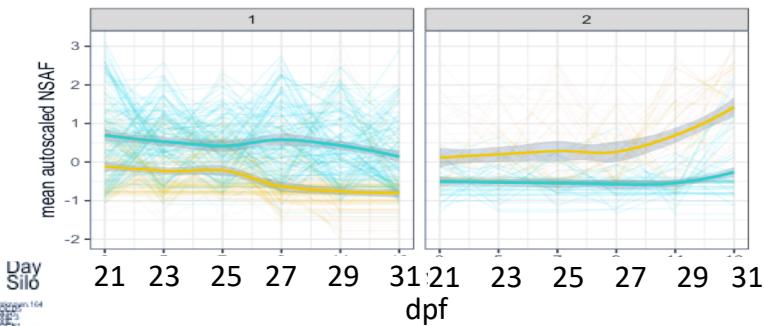
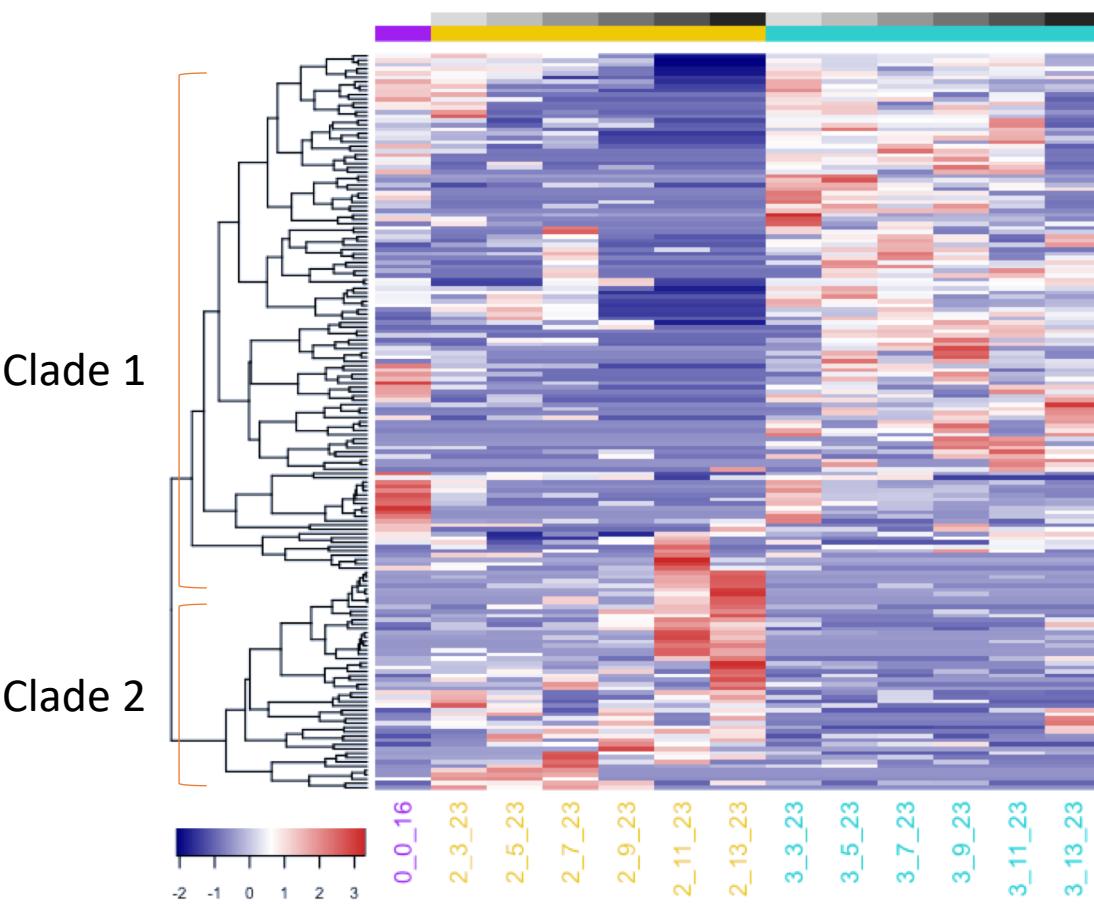
- Lipid metabolism
- Response to fibroblast growth factor
- Actin filament bundle
- Microtubule-based process
- Positive regulation of cell death
- Protein homotetramerization

Proteins and processes underlying decreased survival at 23°C

3. Silo effect:

Protein abundance differences
driven by silo

23°C 23°C



- Pyrimidine nucleoside salvage
- Cellular amino acid metabolism
- Endosome transport via multivesicular body sorting pathway
- Regulation of exocytosis
- Regulation of cholesterol import and esterification
- Rab protein signal transduction

Conclusions

- 29°C rearing led to increased size and survival
- Proteomics data show clear physiological differences between animals reared at 29°C and 23°C
- Established a baseline protein and biological process inventory for fundamental oyster development
- Identified set of proteins and potential processes underlying decreased survival
- Identified set of proteins and potential processes underlying increased growth and survival

Significance and the future

- Proteins in these sets have potential to be biomarkers for determining the health status of seed and predicting outcome before mass mortality events happen
- Need to measure top ranked proteins and/or cell processes on a larger scale