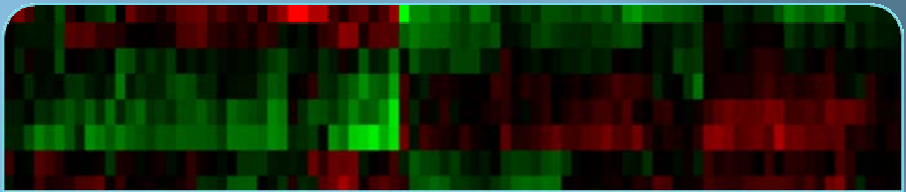
Variation in the genetic response to high temperature in *Montastraea faveolata* from the Florida Keys & Mexico



Nicholas R. Polato[1], Christian R. Voolstra[2], Julia Schnetzer[3] , Michael K. DeSalvo[4] , Carly J. Randall[5], Alina M. Szmant[5], Mónica Medina[4], and Iliana B. Baums[1]

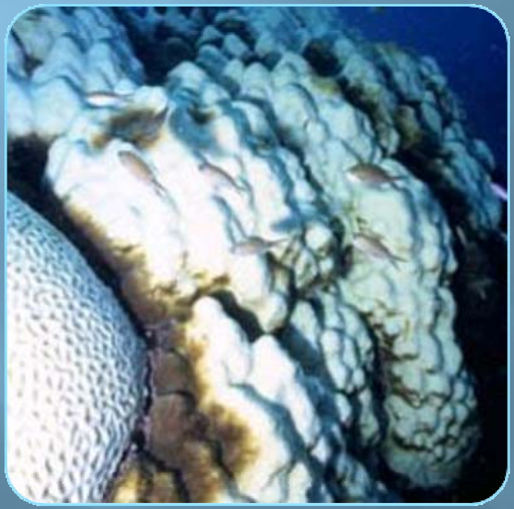
[1] Department of Biology, Penn State University, 208 Mueller Laboratory, University Park, PA 16802, USA
[2] Red Sea Research Center, King Abdullah University of Science and Technology (KAUST), 23955-6900 Thuwal, Saudi Arabia
[3] Max Planck Institute for Marine Microbiology, Celsiusstr. 1, 28359 Bremen, Germany.
[4] School of Natural Sciences, University of California Merced, 5200 North Lake Road, Merced, CA 95343, USA
[5] Center for Marine Sciences, University of North Carolina Wilmington, 5600 MarvinK. Moss Lane, Wilmington, NC 28409, USA

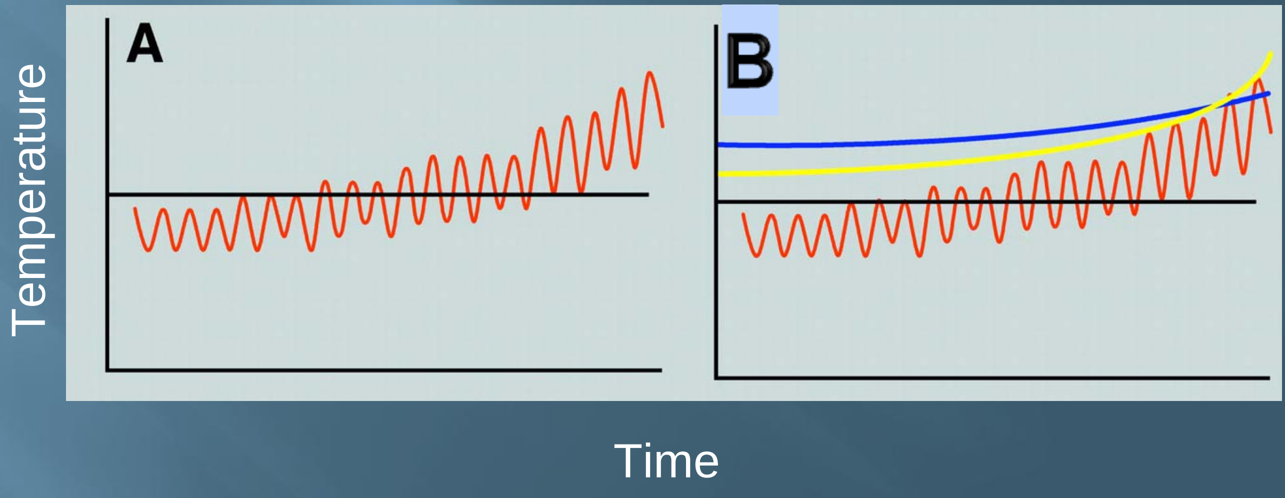
Many threats currently face coral reefs

Overfishing & Extractive resource use

Pollution & Coastal Development

Climate Change:
- •Rising sea level
- •Ocean Acidification
- •Rising Sea Surface Temperatures (SST)





Graphs: T. P. Hughes et al., Science 301, 929 -933 (2003)
Image: http://www.cdnn.info/news/eco/coral_bleaching_250373.jpg

This situation is exacerbated in Caribbean reefs by declining juvenile recruitment

- Combined with reduced growth, and increased mortality this is leading to population declines

- Successful sexual reproduction is necessary for recovery and persistence of these ecosystems

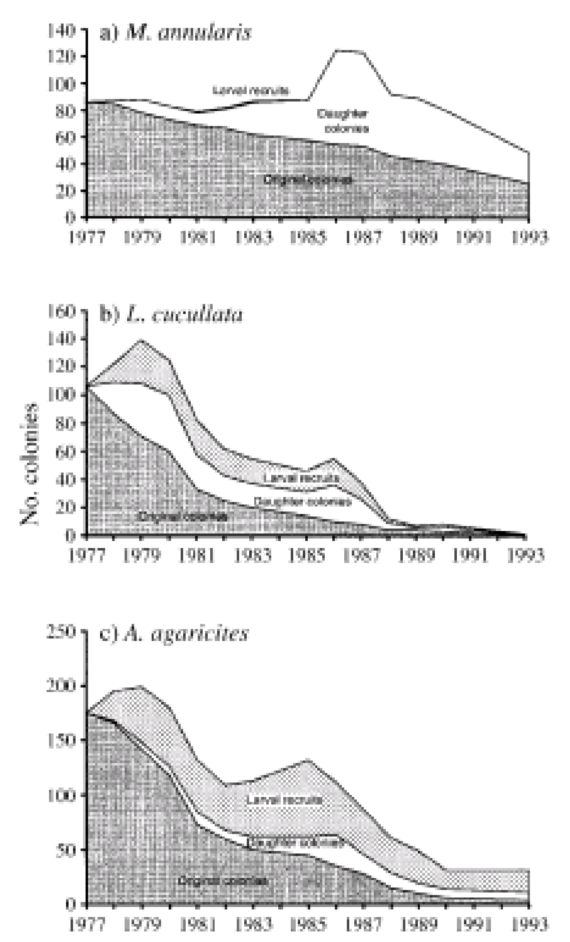


FIG. 2. Number of colonies of *Montastrea annularis*, *Agaricia agaricites*, and *Leptoseris cucullata* over time. Colonies at each census are divided into three types: those originally present in 1977, daughter colonies formed by fission, and new larval recruits.

Hughes TP, Tanner JE (2000) Recruitment failure, life histories, and long-term decline of Caribbean corals. Ecology 81: 2250-2263.

Can corals adapt to rapid climate change?

The answer depends upon the adaptive potential inherent to coral populations!

Con:

- Corals currently live at or near their thermal maximum throughout much of their range
- Local adaptation will be hindered by long distance migration

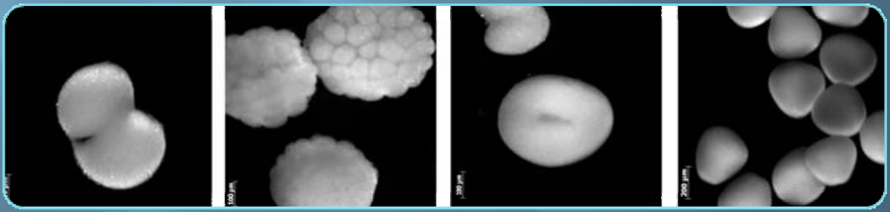
Pro:

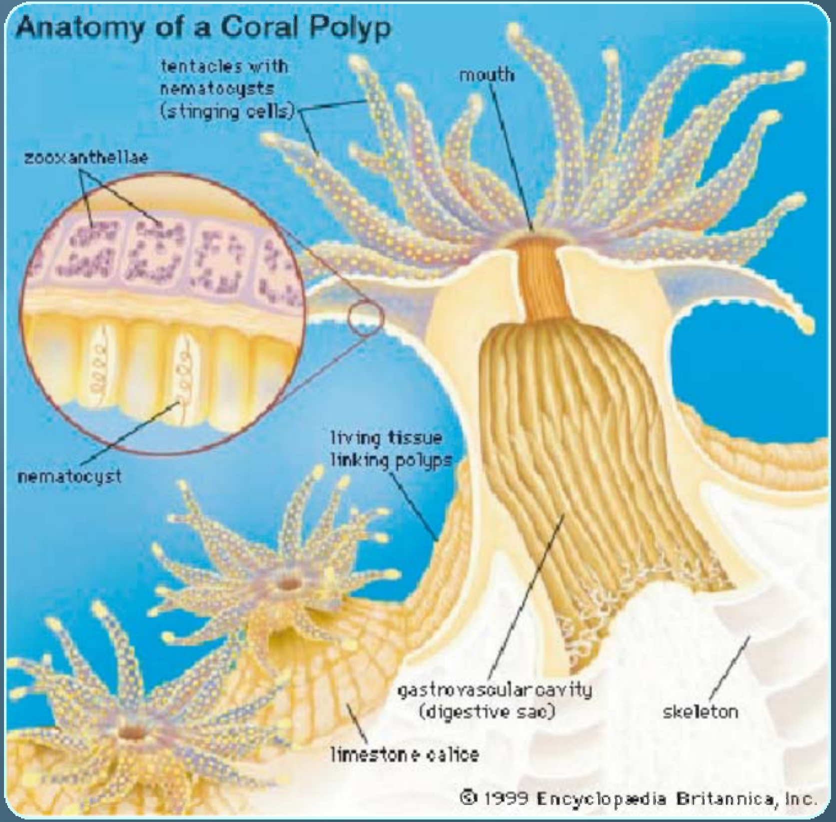
- Corals have the ability to switch symbiont types
- There is Evidence for local adaptation in several species:
    - small scale population structure has been observed multiple coral species

Each member of the holobiont contributes to fitness

• It is important to understand the contribution of different symbiont types

• Coral larvae allow investigation of the host response in isolation

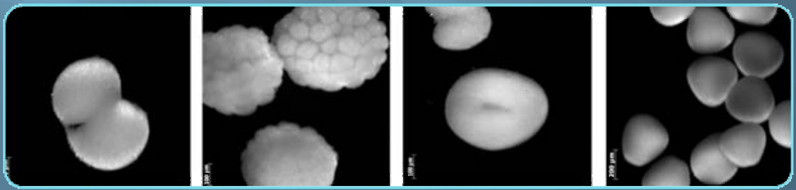


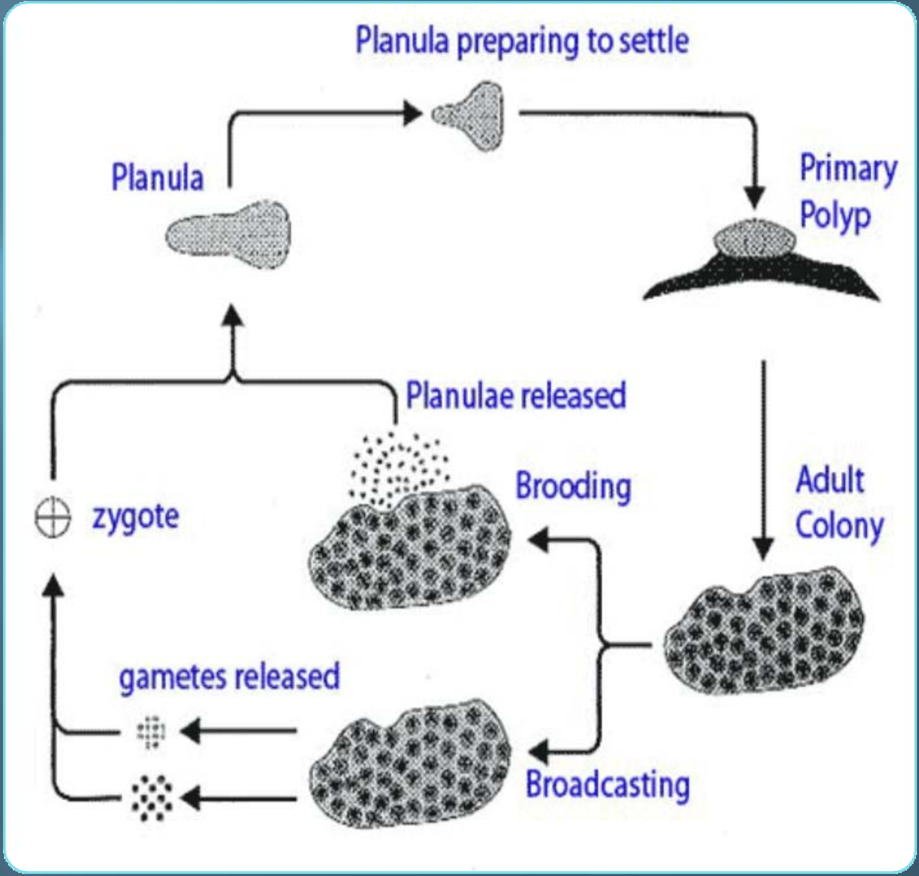


Larvae are critical for coral survival:

- By maintaining genetic connectivity among populations

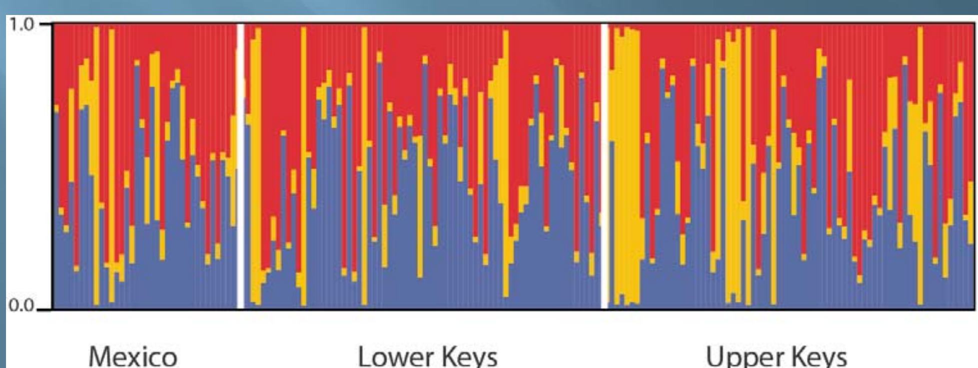
- Dispersing larvae are the only way corals can escape unsuitable habitat and exploit new ones





http://www.bios.edu/research/images/lifecycle.png

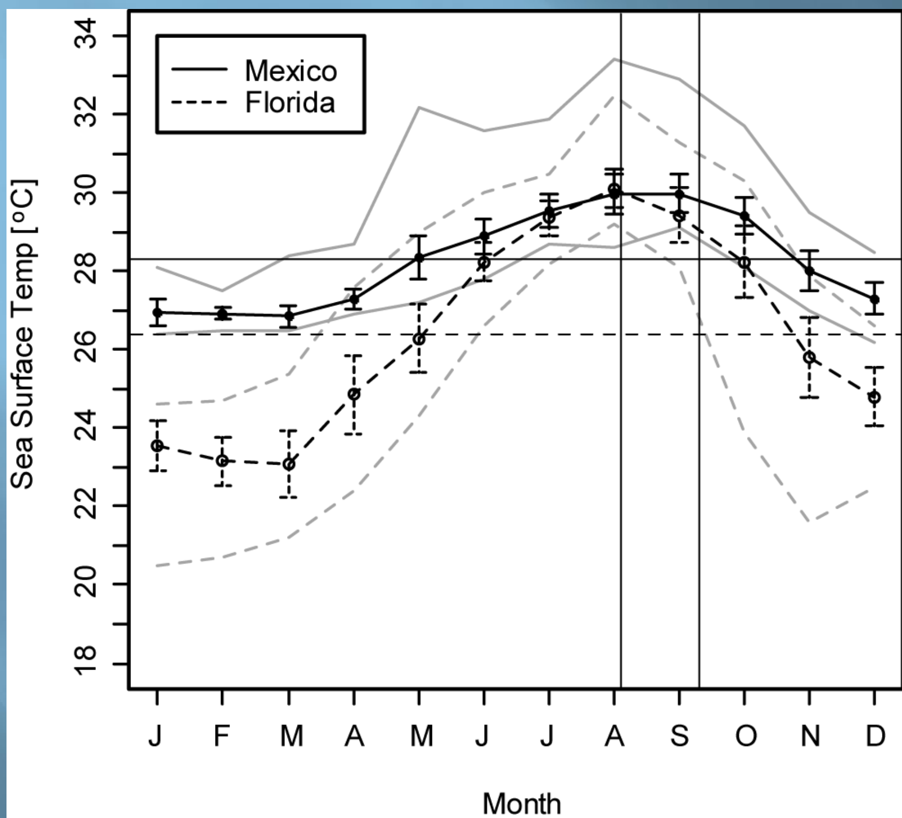
Gametes from multiple parents from two locations were collected:



Study sites are linked by regional currents

No geographical populations structure was detected

Temperatures in Mexico reach higher extremes than Florida

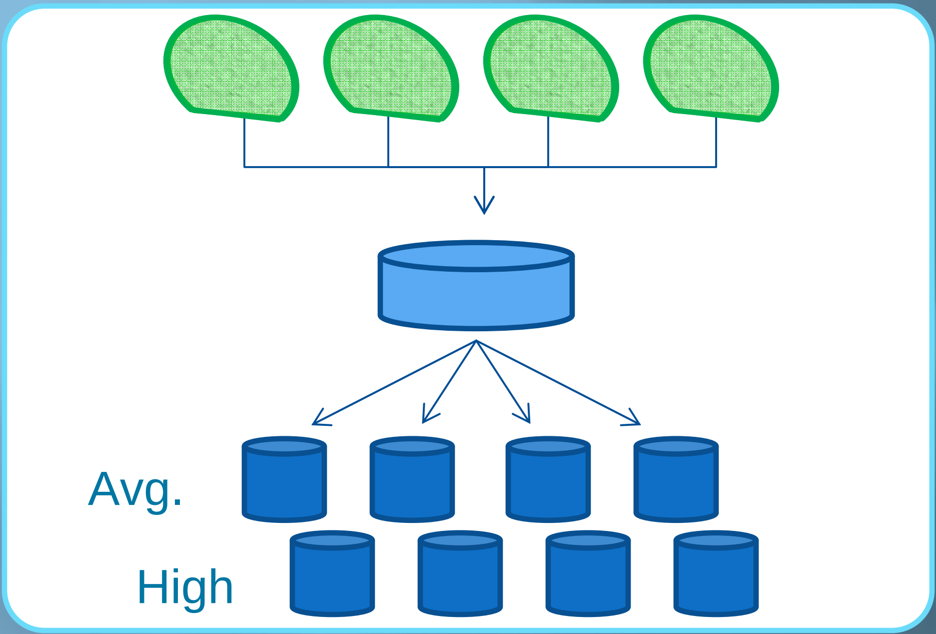


Annual means (05 - 08) differ by ~2° C

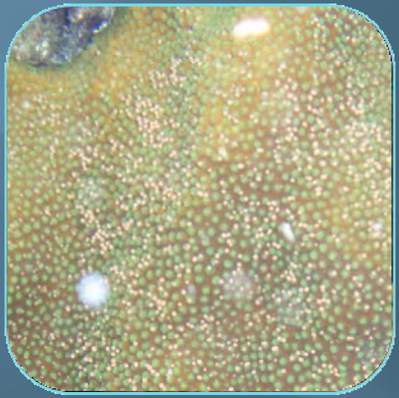
Summer highs average 33 in Mexico and 31 in Florida

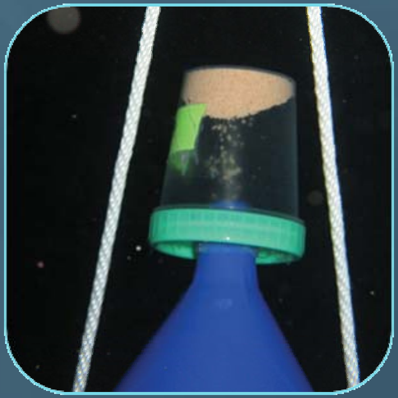
Florida has a greater thermal range with lower winter lows

Crosses included offspring from a minimum of 3 parents from each site



- Collect gametes from parents at both populations
- Pool sperm and eggs to generate batch crosses
- Raise larvae at 2 temperatures
    - 27 and 30 in Florida
    - 27.5 and 32.5 in Mexico
- Need larval tissue to avoid genetic material from the algal symbiont !



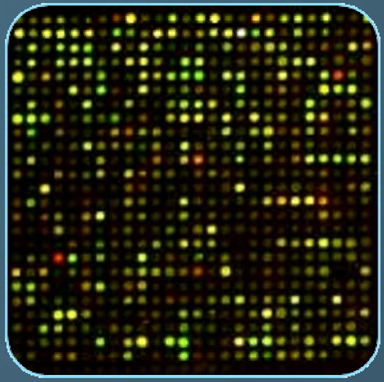


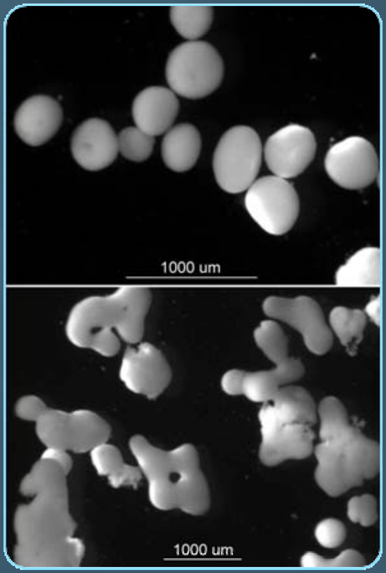
Do corals differ in their response to thermal stress depending on the location where they live?

1. Transcription profiles will reflect location specific variation in thermal stress response

2. Thermal stress response will include differential expression of genes for previously identified stress markers
   1. Heat shock proteins
   2. Oxidative stress genes

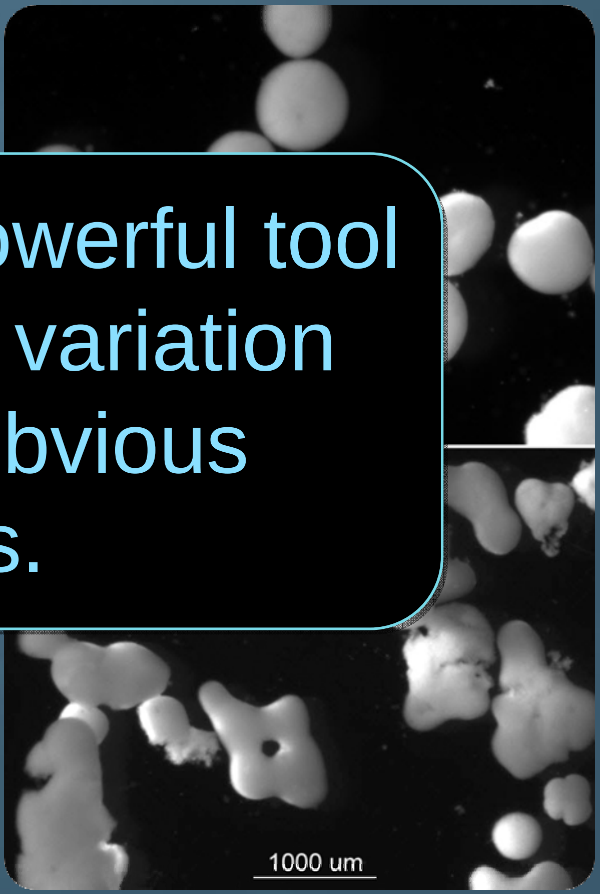
3. DEGs relating to cell structuring and development will be observed





Higher temperatures resulted in more malformed larvae

Locatio	Age	Temperature	Irregular Embryos	Normal Embryos as Invaginated	Normal Embryos as Gatrula
Floric					
Mexic					
	50.5	27.5	11	0	99
	50.5	31.5	4	0	100

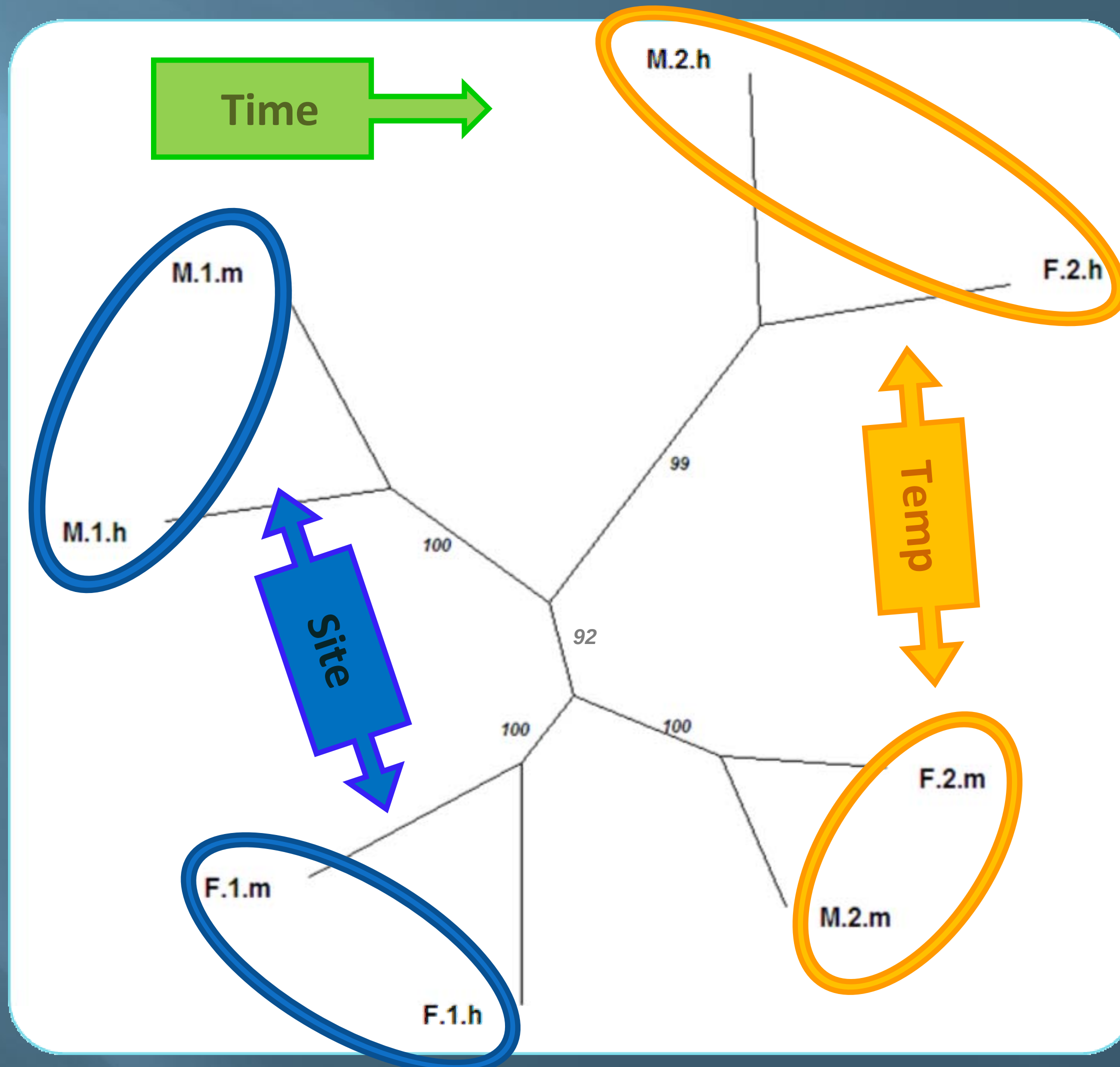


Transcription profiling is a powerful tool for observing physiological variation even in the absence of obvious external differences.

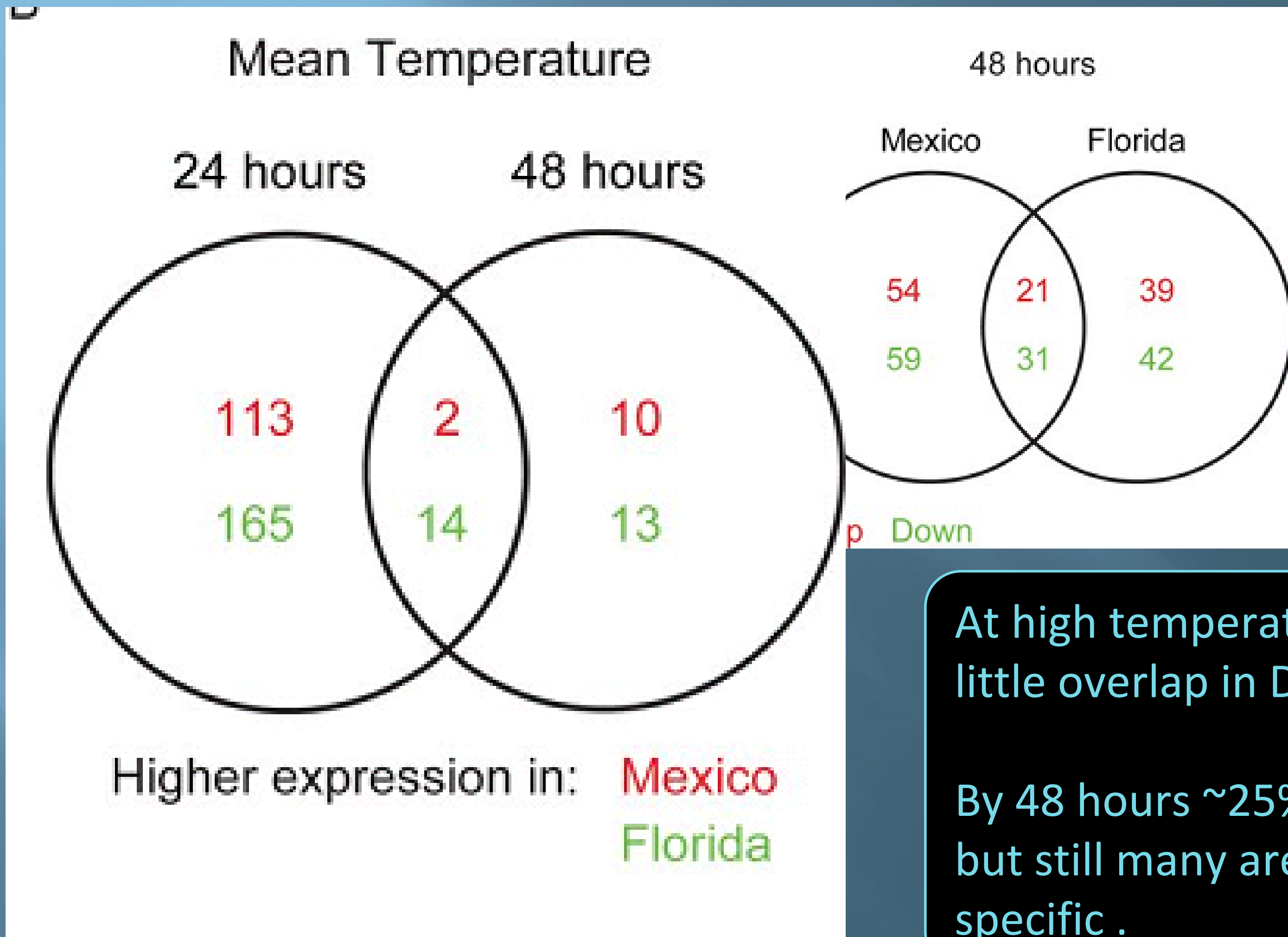
Developmental differences between temperatures were not observed at 24 or 48 hours

There is a strong geographical component to the response of larvae to thermal stress

M – Mexico
F – Florida
1 – day 1
2 – day 2
m – avg temp.
h – high temp.



Response to thermal stress has both conserved and site specific components



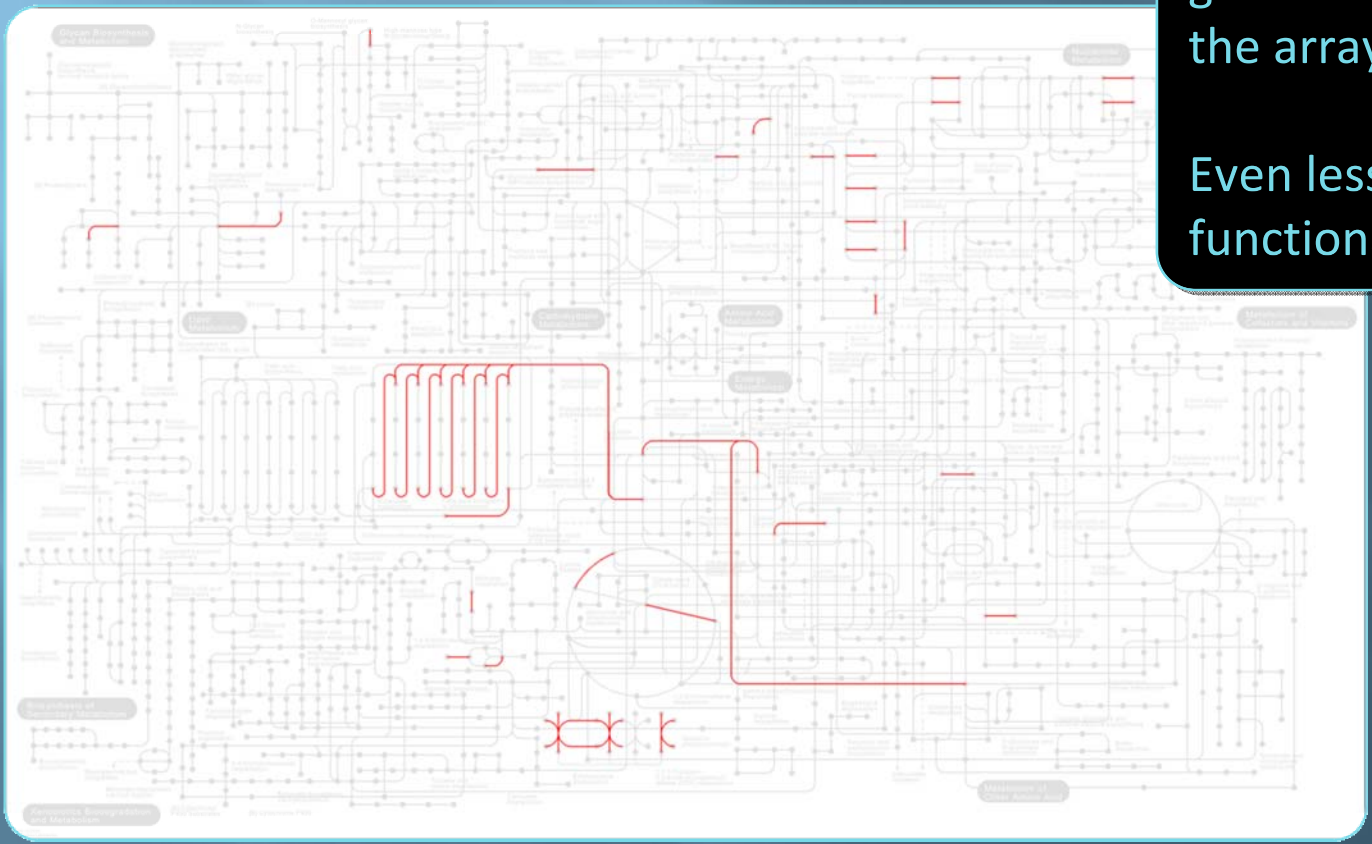
At high temperatures there is little overlap in DEGs

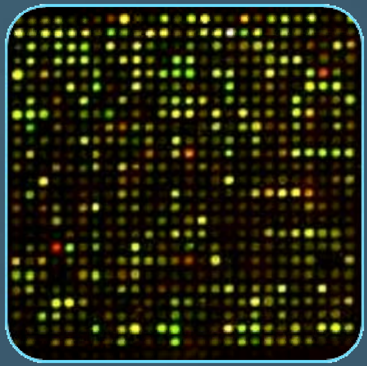
By 48 hours ~25% are shared, but still many are population specific .

Our ability to interpret the function of theses differences is limited by a lack of annotation

Only a small fraction of the genome is represented on the array

Even less (~20%) is functionally annotated





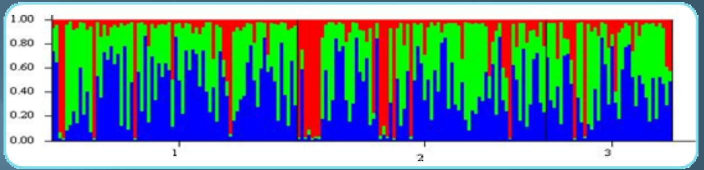
Conclusions:

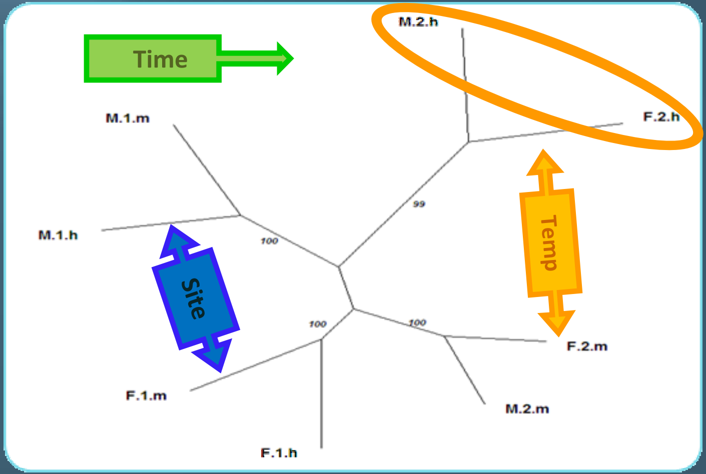
1. There is a strong **geographical component** to the response of coral larvae to thermal stress

    1. Management efforts at one location may not give the same results in the other

2. Application of thermal stress leads to a **conserved response** across populations

3. Understanding the function of DEG's requires better annotation of cnidarian genomes and consideration of gene function at specific life stages

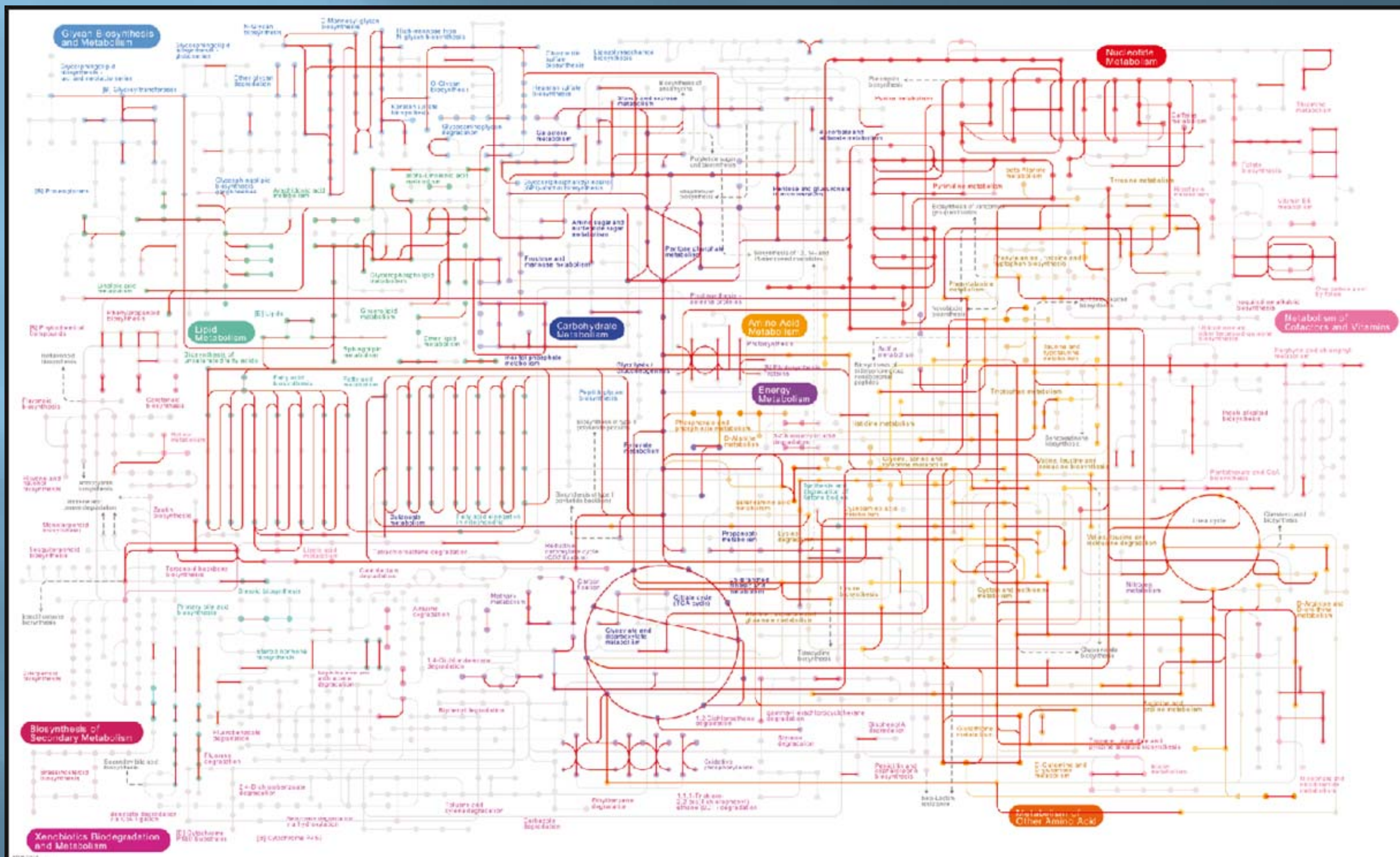




The *A. palmata* transcriptome provides a comprehensive set of ESTs with which to survey gene expression

		A.palmata			
		N sequences	total length [Mb]	Avg length (sd) [bp]	Depth of coverage (max)
raw reads		964,519	384	398 (118)	
trimmed reads		741,271	320.5	432 (64)	
contigs		42,630	44	1034 (624)	5.6 (315)
	> l kb	16,274			
	# annotated	29,413			
Singletons		45,390	20	441 (95)	
	# annotated	16,848			

Coverage of the *A. palmata* transcriptome is comparable to the *N. vectensis* genome



A 135K feature microarray will enable more detailed surveys of gene expression

Nimblegen 12-plex slides will be used to profile gene expression patterns in *A. palmata* adults and larvae

Tests for interspecific hybridization will be performed using *A. cervicornis*

•2 probes per contig

•1 probe per singleton

•Enriched for stress & calcification related transcripts

http://ddlab.sci.univr.it/FunctionalGenomics/facility.html

Thank You!

Baums Lab : Iliana Baums, Meghann Devlin-Durante, Katey Glunt, Jennifer Boulay, Dannise Ruiz, John Parkinson, Dennis Xu

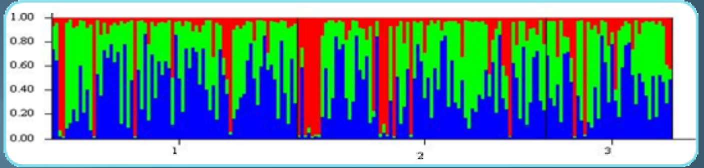
Medina Lab: Michael De Salvo, Chris Voolstra, Julia Schnetzer Erika Diaz, Collin Closek, Shini Sunagawa, Monica Medina

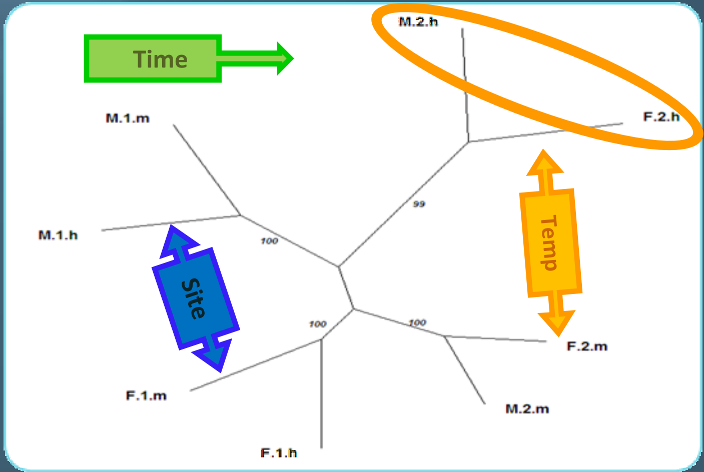
NOAA : Margaret Miller, Abel Valdivia

Secore, CARMABI

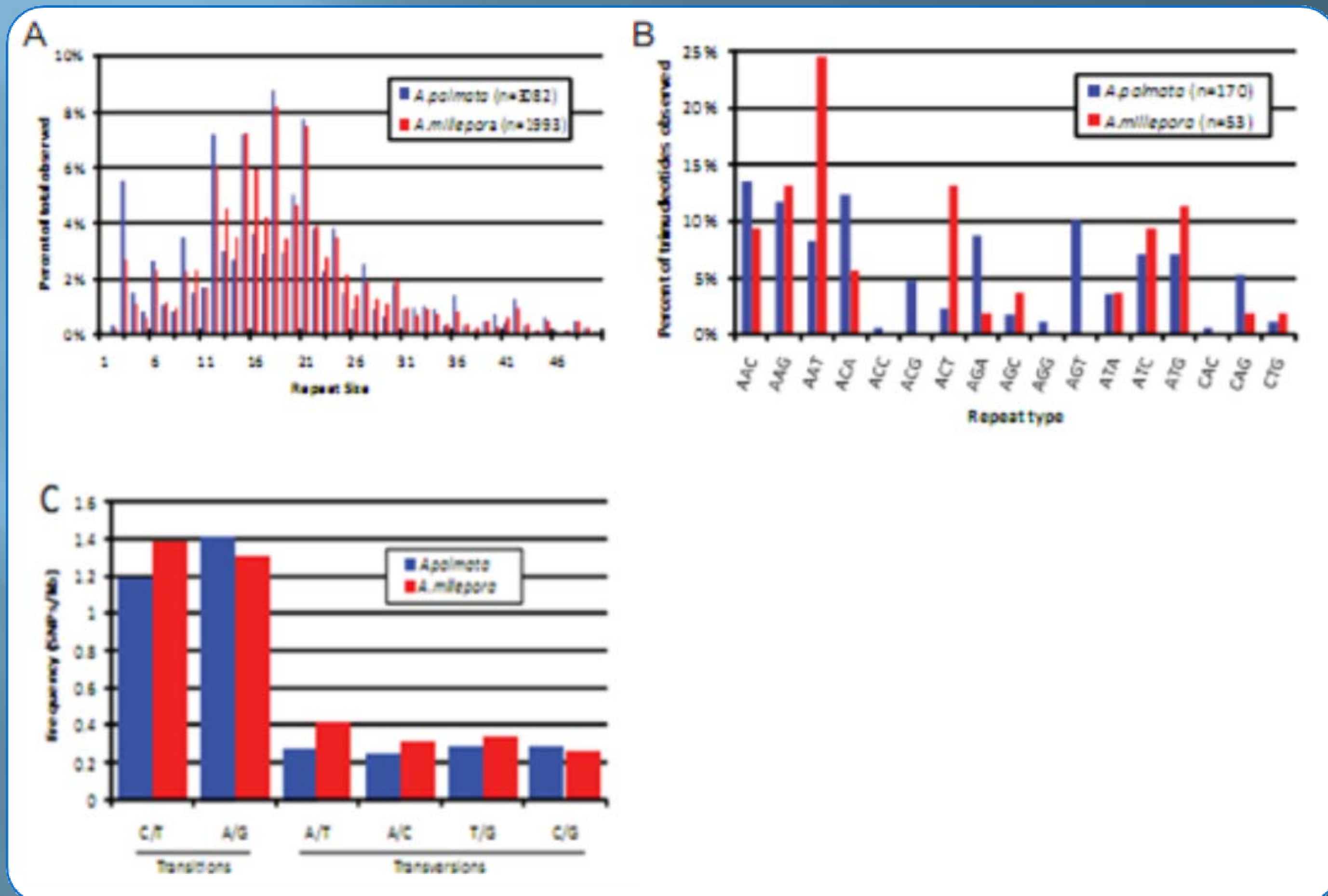
NSF

NSF Graduate Research Fellowship Program





Sequencing results also identify a wealth of new molecular markers



Even with limited annotation, enrichment of key functions is observed:

	Common to both populations	Unique to Florida	Unique to Mexico
Up			
	cell proliferation, growth, development	autophagy, protein degredation	degradation
	cell structure, motility	cytoskeleton, cell adhesion	DNA
	cytoskeleton, cell adhesion	DNA repair	ER, ion binding , transport
	lipid binding/metabolism	iron transport	lipid binding
	response to stress	lipid binding/metabolism	metabolism
	transcription, transcription regulation	metabolism	protein binding
		ribosome, translation	response to oxidative stress
		transcription regulation, development	transcription regulation
Down			
	Apoptosis	cell adhesion, development	cell growth, development
	cell proliferation, growth, development	cell growth, development	cell structure
	cytoskeleton, cell adhesion	cell structure, motility	degradation
	DNA	degradation	energy metabolism
	electron transport, oxidative phophorylation	protein biosynthesis	ER, iron ion binding , transport
	metabolism	protein degradation	metabolism
	response to oxidative stress	response to stress	protein biosynthesis/folding/transport
	response to stress	RNA binding	protein degradation
	DNA, mRNA modification		response to oxidative stress
	signaling		response to stress
	translation, ribosomes, protein biosynthesis		ribosome, translation
			RNA binding
			transcription, apoptosis

Several genes related to heat and oxidative stress response are downregulated:

M – Mexico
F – Florida
1 – day 1
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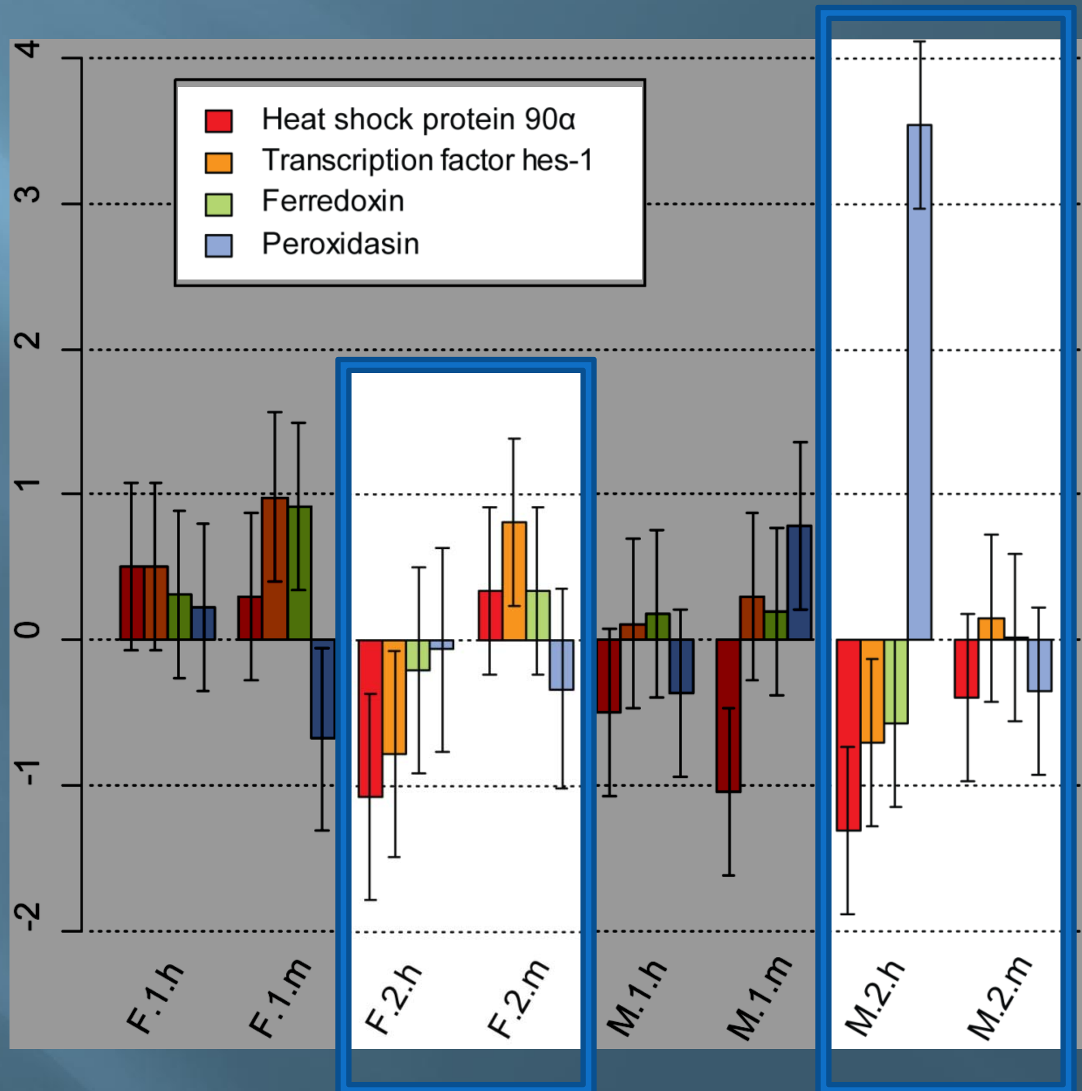
h – high temp.



Heat shock proteins blast to *N. vectensis* genome with high homology:

Hsp 90 a: AOSC617

PO Box 2345, Beijing 100023, China
Fax: +86-10-85381893
E-mail: wjg@wjgnet.com www.wjgnet.com

WJG, 1999 June; 5(3):199-208
World Journal of Gastroenterology

ISSN 1007-9327

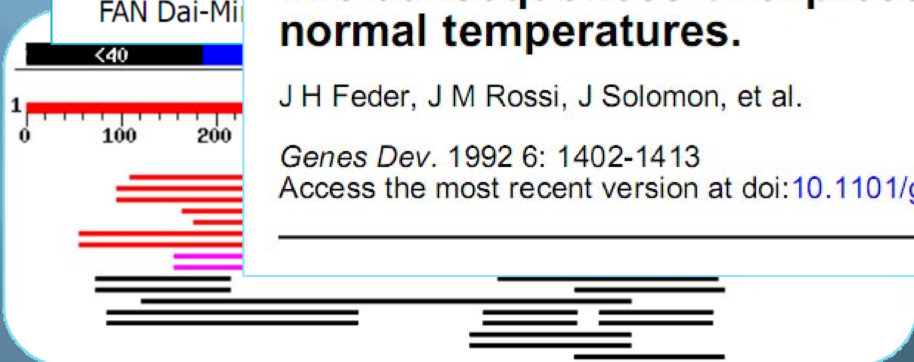
Original Articles

Down-regulation of Hsp90 could change cell cycle distribution and increase drug sensitivity of tumor cells

LIU Xian-Li
FAN Dai-Mi

a

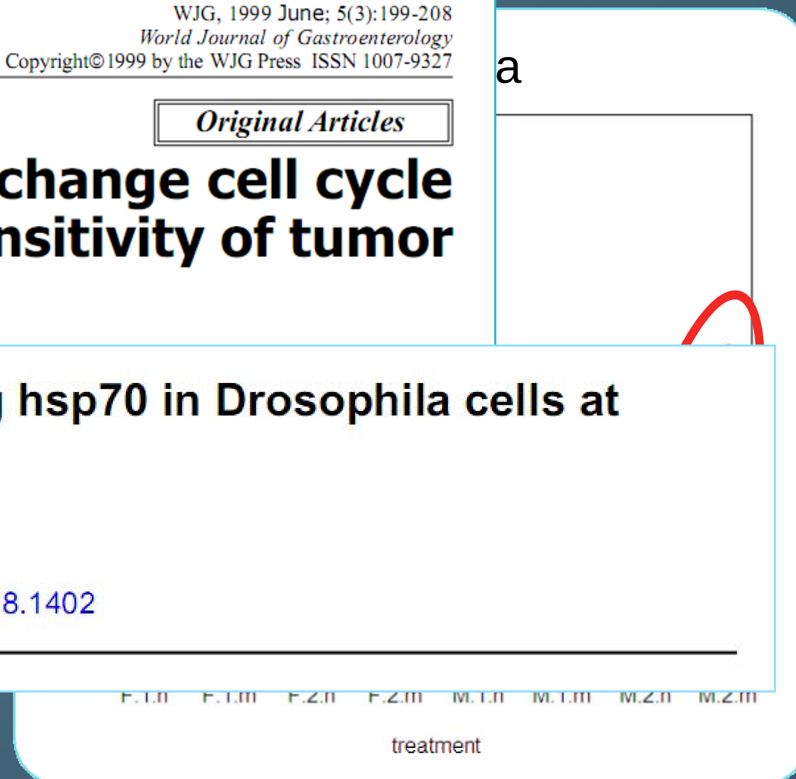
H



The consequences of expressing hsp70 in Drosophila cells at normal temperatures.

J H Feder, J M Rossi, J Solomon, et al.

Genes Dev. 1992 6: 1402-1413
Access the most recent version at doi:10.1101/gad.6.8.1402



Many threats currently face coral reefs:

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Climate Change:
- Rising Sea Surface Temperatures (SST)
- Rising sea level
- Ocean Acidification

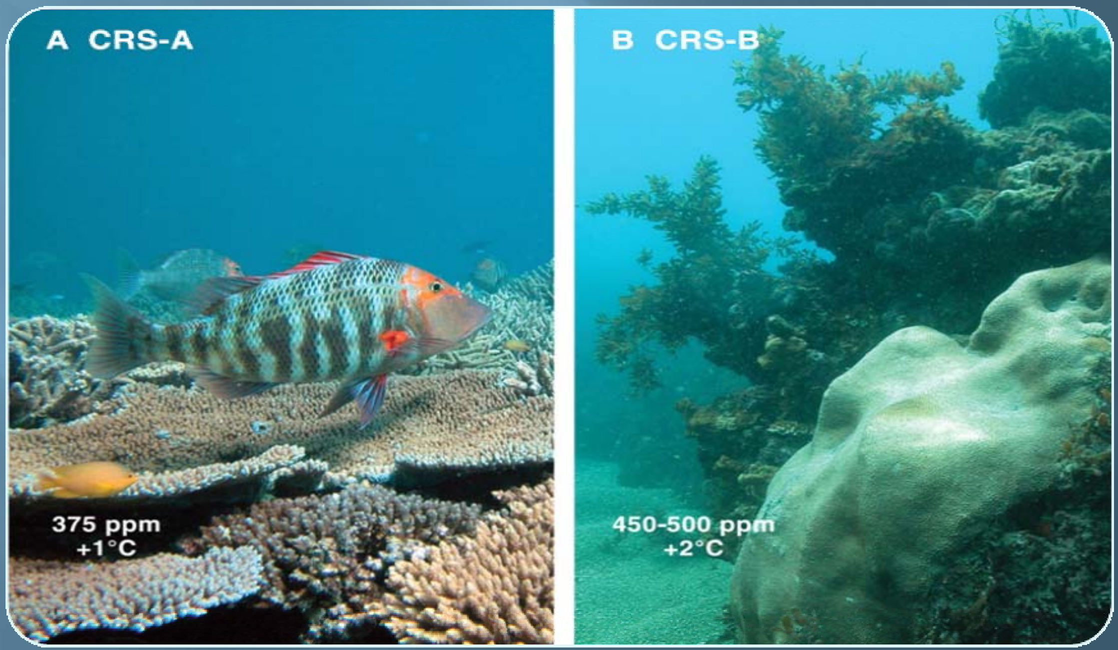

Image source: O. Hoegh-Guldberg et al., Science 318, 1737 -1742 (2007)

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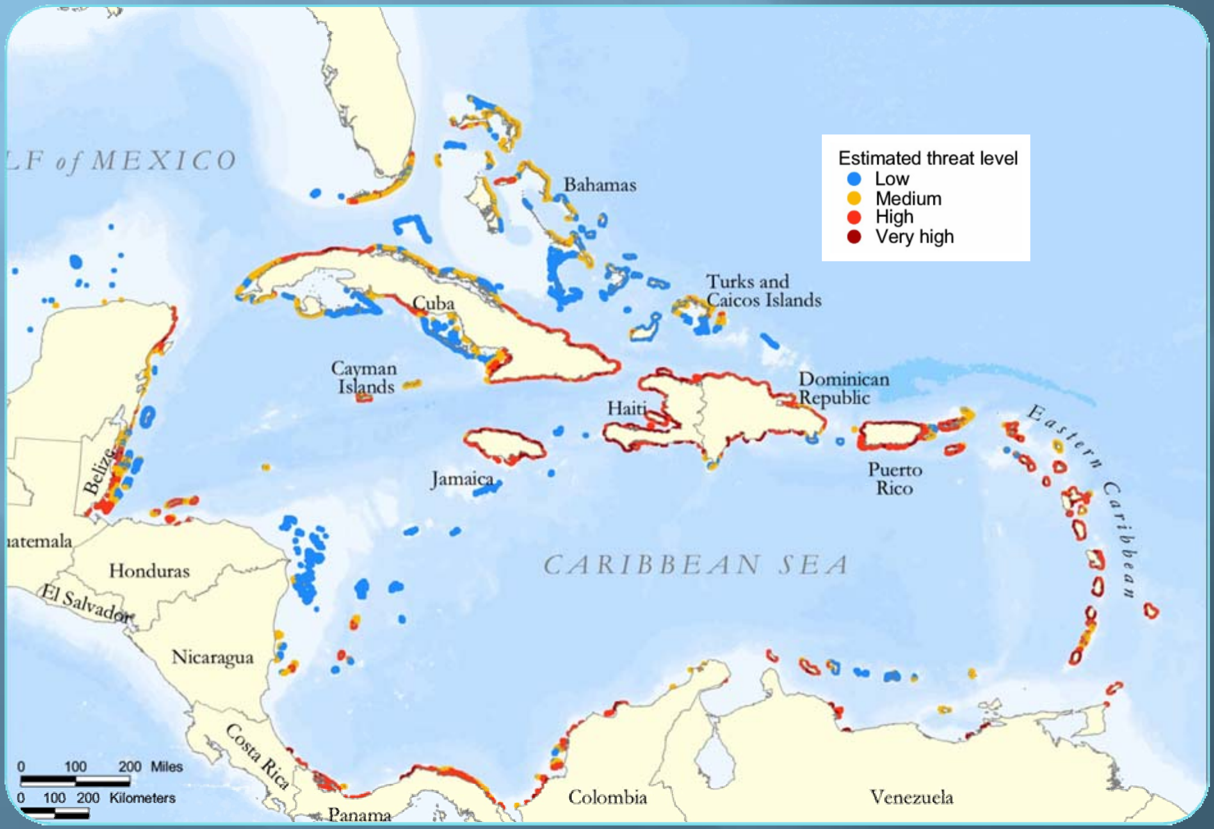
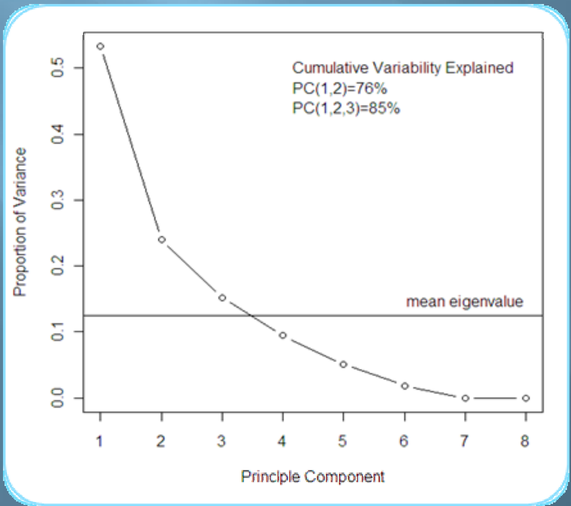
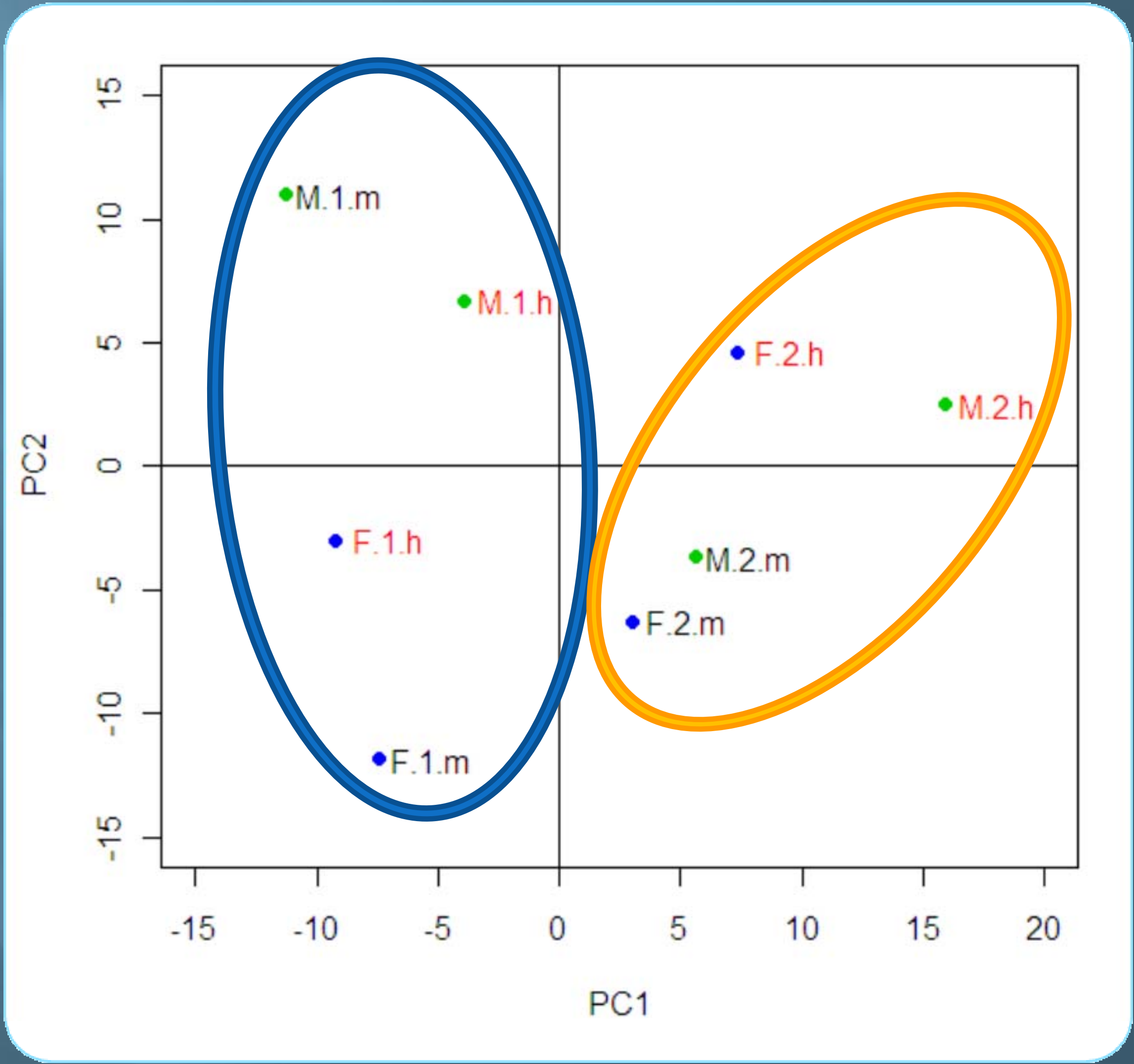


Image Source: The World Resources Institute

All 3 main effects influence gene expression profiles:

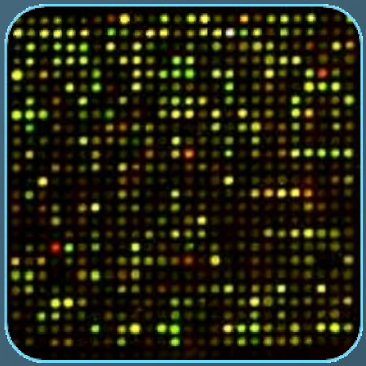
M – Mexico (green)
F – Florida (blue)
1 – day 1
2 – day 2
m – avg temp. (black)
h – high temp. (red)



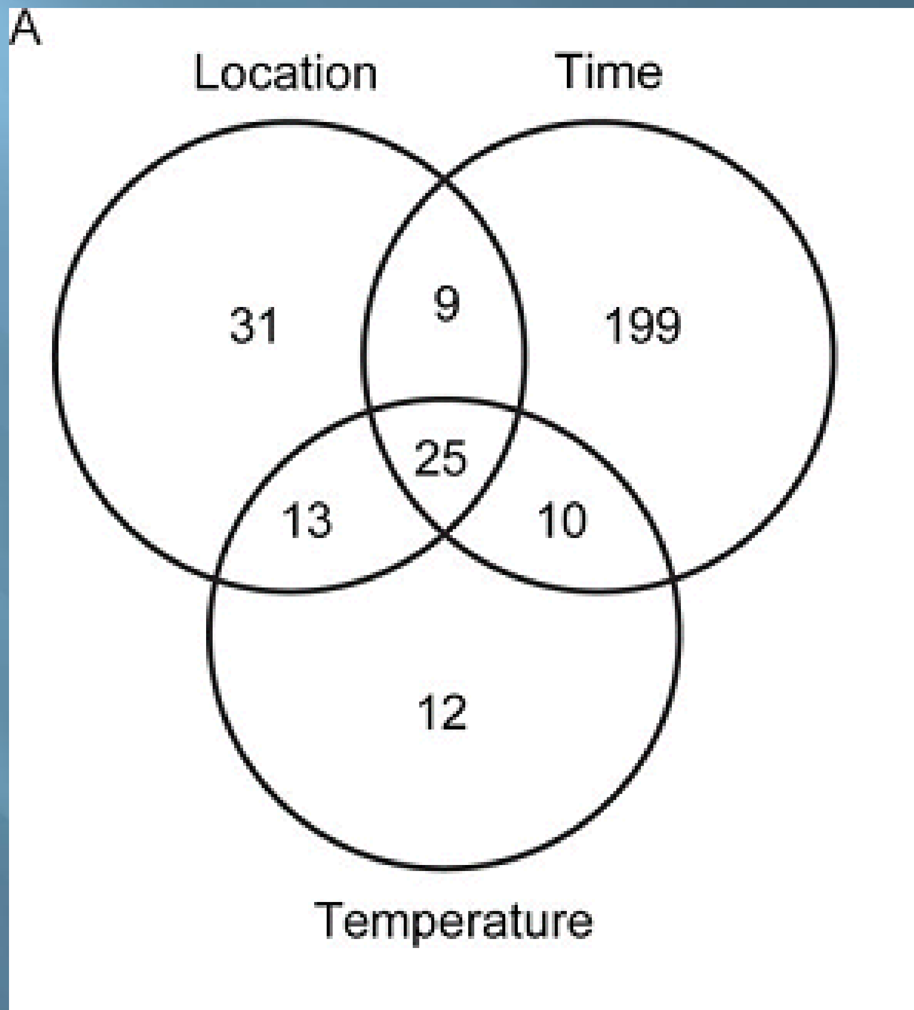


Do corals differ in their response to thermal stress depending on the location where they live?

- Gamete bundles collected from Mexico and Florida
- Gametes pooled in batches and allowed to fertilize 1 hour
- Fertilized eggs distributed into aquaria at 2 treatment temperatures (mean & high)
- RNA was extracted from samples at 24 & 48 hours of development
- 3 replicates of each sample used to interrogate 1300 feature microarray



Microarrays were run for 2 time-points, at 2 temperatures, from both locations.


A
Location
Time
31
9
199
25
13
10
12
Temperature