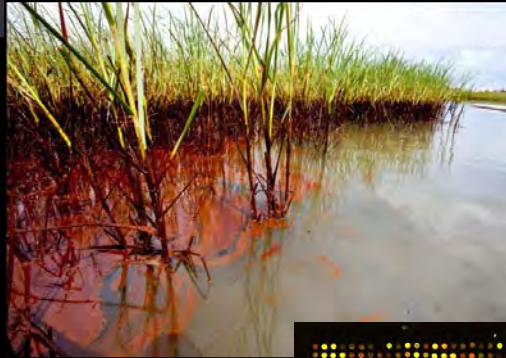


Genome Expression Response of Resident Killifish Impacted by the DWH Oil Spill



Andrew Whitehead

Fernando Galvez

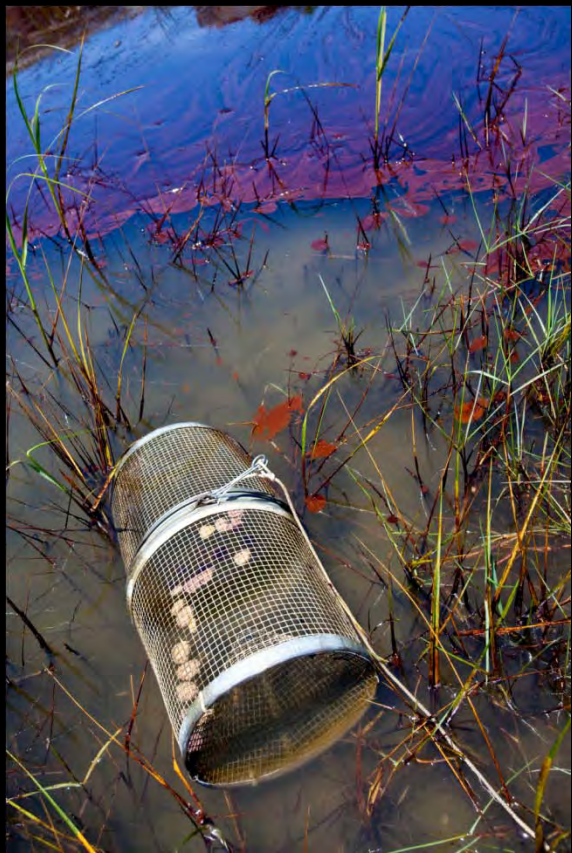
*Dept. of Biological Sciences,
Louisiana State University*





April 20, 2010





THE MODEL:



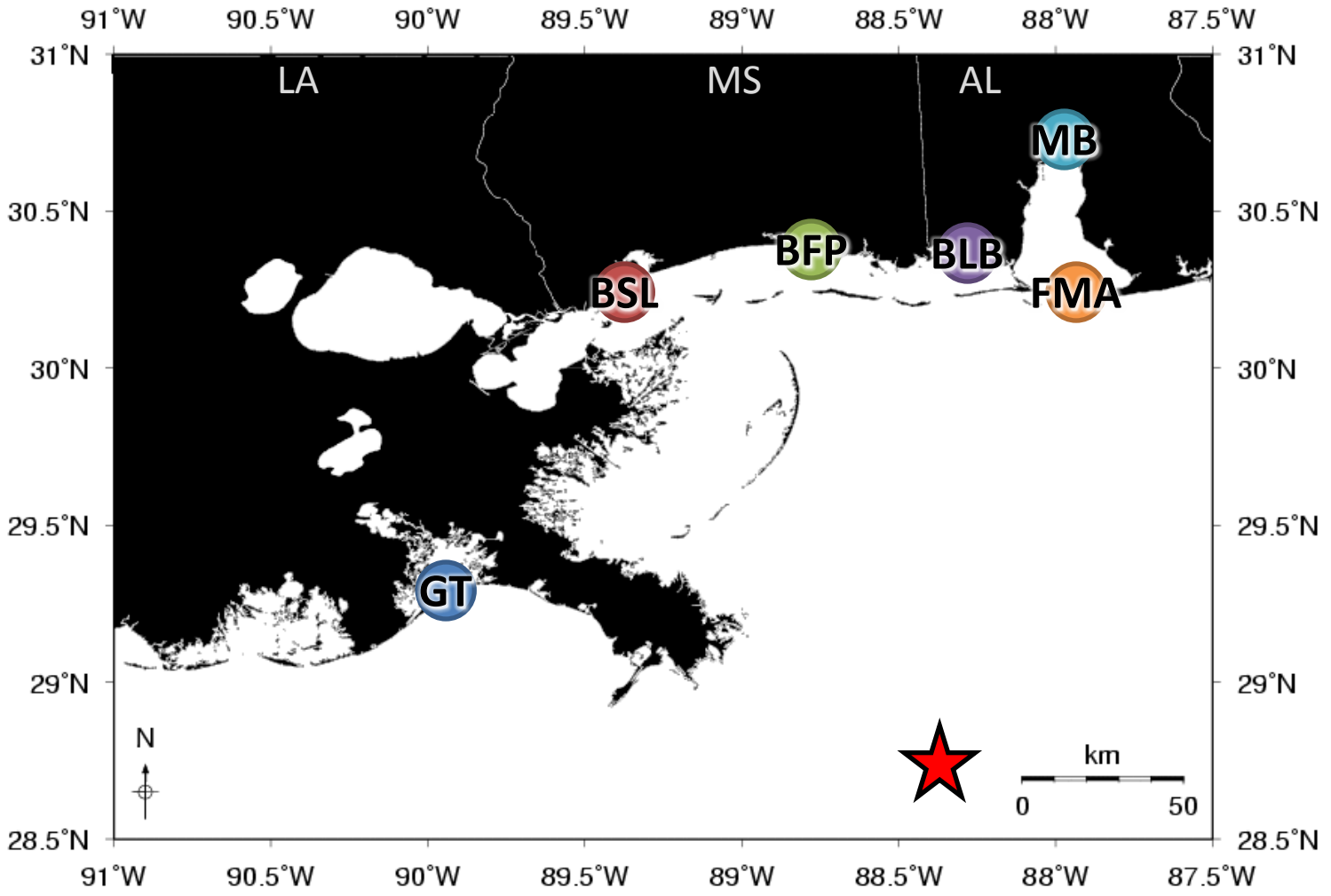
Gulf Killifish

(Fundulus grandis)

- large population sizes
- non-migratory
- high site fidelity
- relatively sensitive (among fishes) to toxicity from organic pollutants



Deepwater Horizon oil spill – field study



Physiological
Genomics

Andrew Whitehead



Jen Roach



Eve McCullough



David Roberts

Tissue
Histochemistry

Fern Galvez



Ben Dubansky



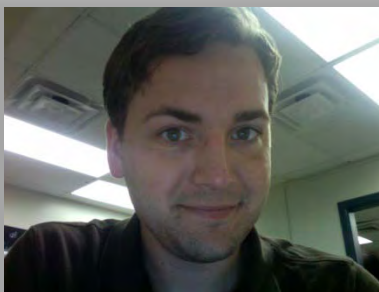
Charlotte Bodinier



Charlie Rice

Remote
Sensing

Nan Walker



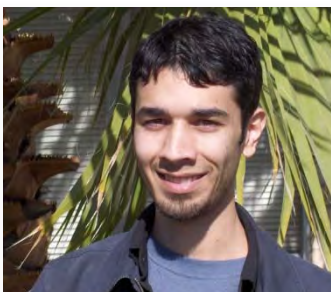
Chet Pilley



Vandana Raghunathan

RNaseq

Ron Walter



Tzintzuni Garcia

Analytical
Chemistry

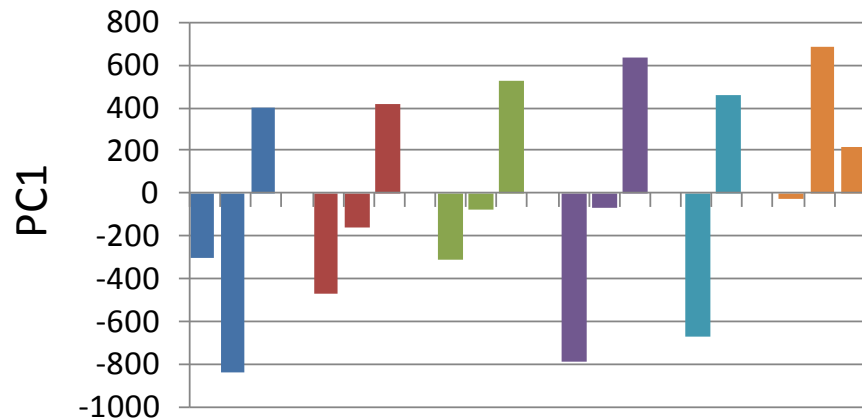
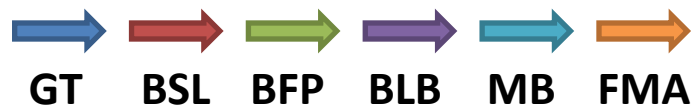
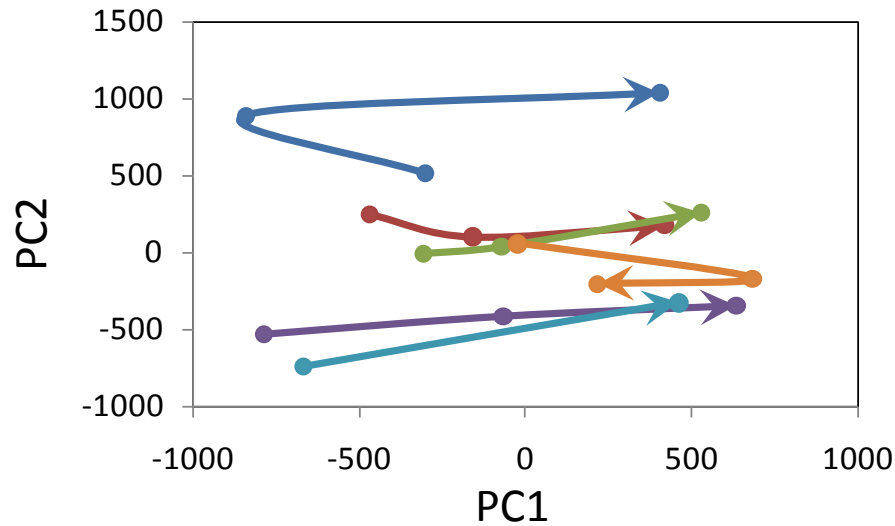


Scott Miles

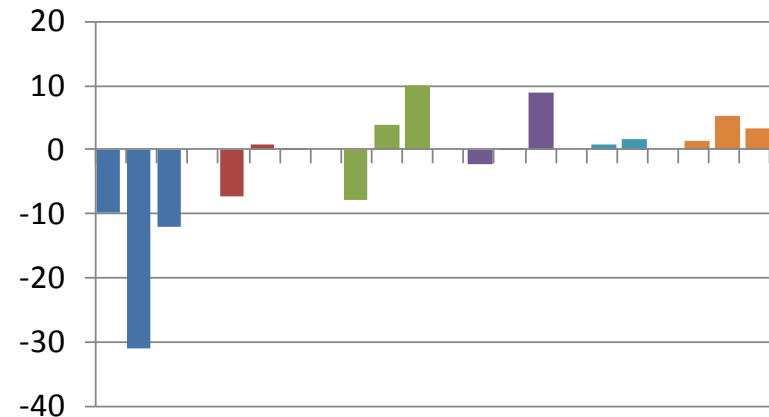
Genome expression: Microarray



Genome expression response trajectories through time (liver)



Subset of genes that are PCB-induced in killifish



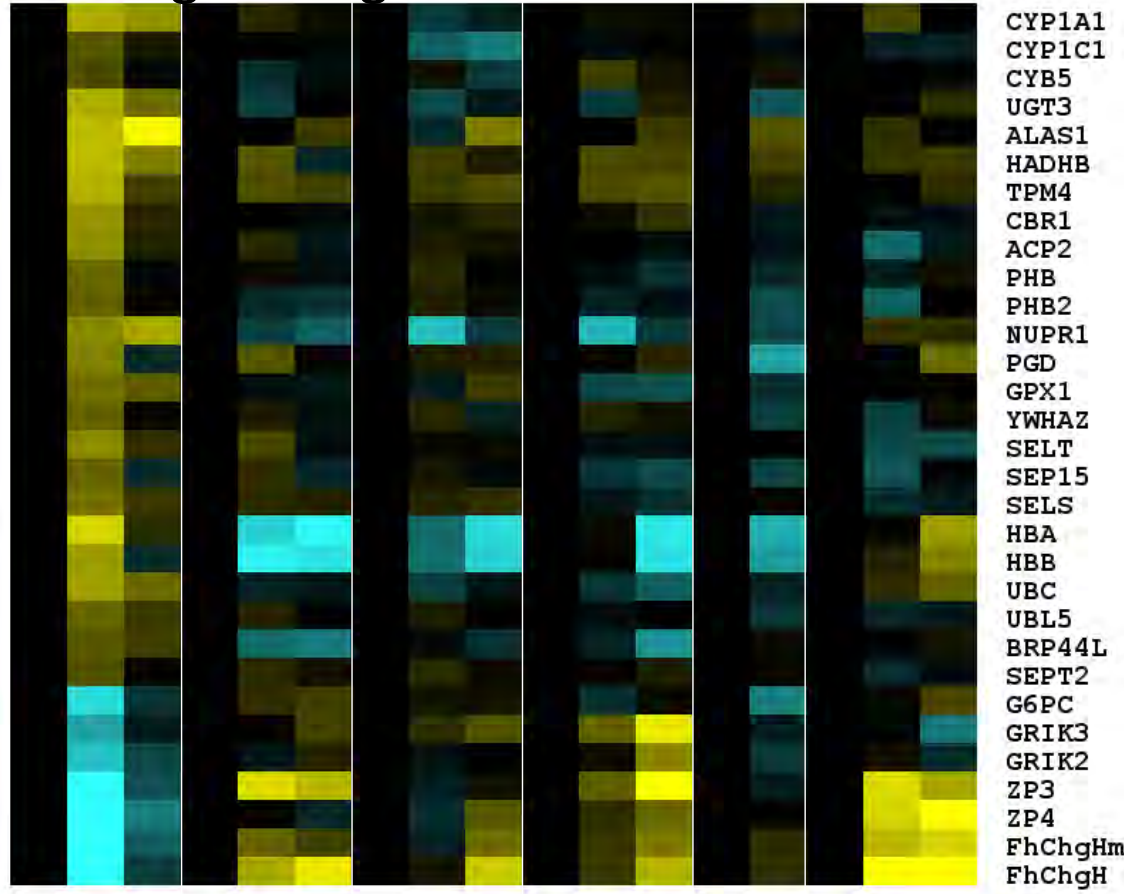
Genome expression response

→ GT outlier



AHR ligand regulated

-4 +4



- CYP1A1
- CYP1C1
- CYB5
- UGT3
- ALAS1
- HADHB
- TPM4
- CBR1
- ACP2
- PHB
- PHB2
- NUPR1
- PGD
- GPX1
- YWHAZ
- SELT
- SEP15
- SELS
- HBA
- HBB
- UBC
- UBL5
- BRP44L
- SEPT2
- G6PC
- GRIK3
- GRIK2
- ZP3
- ZP4
- FhChgHm
- FhChgH



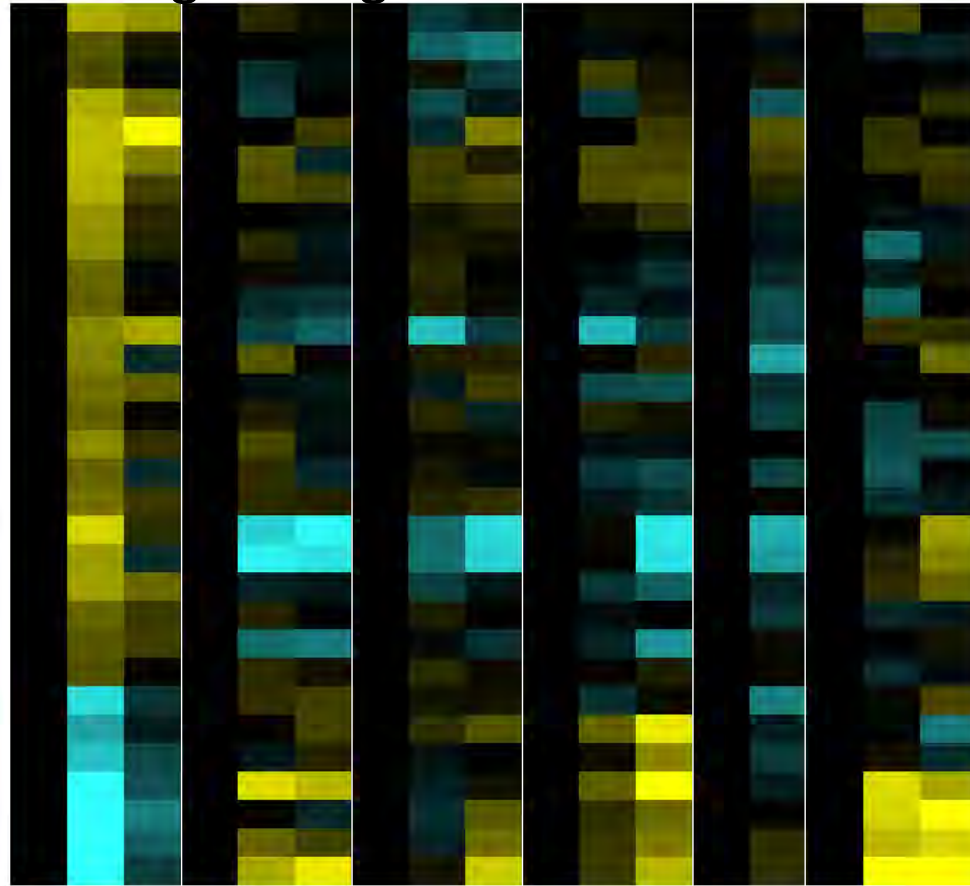
Genome expression response

→ GT outlier



AHR ligand regulated

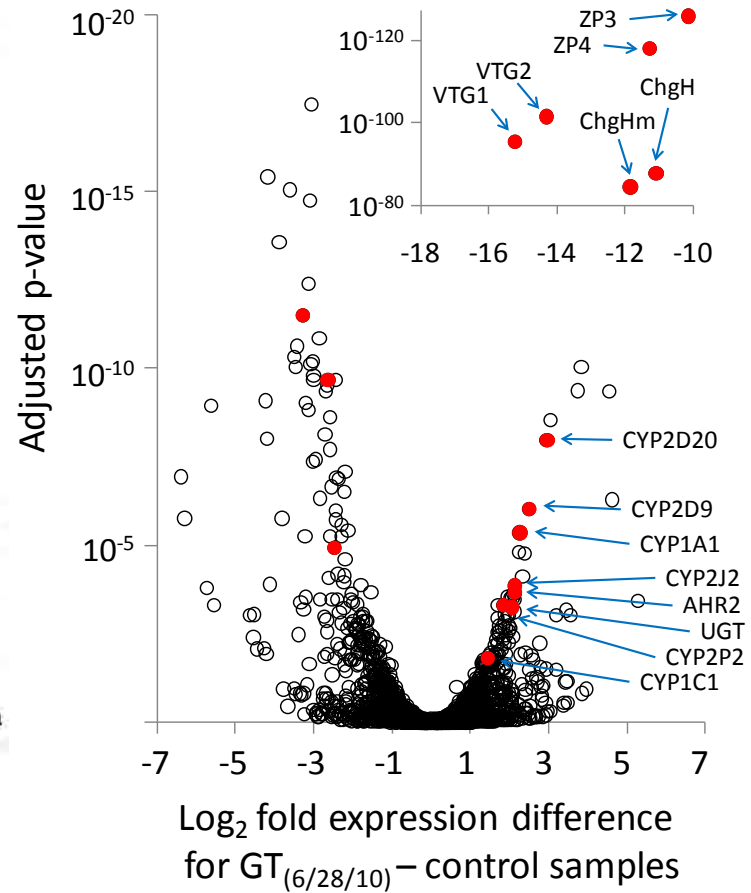
-4 +4



- CYP1A1
- CYP1C1
- CYB5
- UGT3
- ALAS1
- HADHB
- TPM4
- CBR1
- ACP2
- PHB
- PHB2
- NUPR1
- PGD
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- UBL5
- BRP44L
- SEPT2
- G6PC
- GRIK3
- GRIK2
- ZP3
- ZP4
- FhChgHm
- FhChgH

GT BSL BFP BLB MB FMA

Collaborator: Ron Walter, Post-doc: Tzintzuni Garcia



Genome expression response

→ GT outlier



Proteolysis

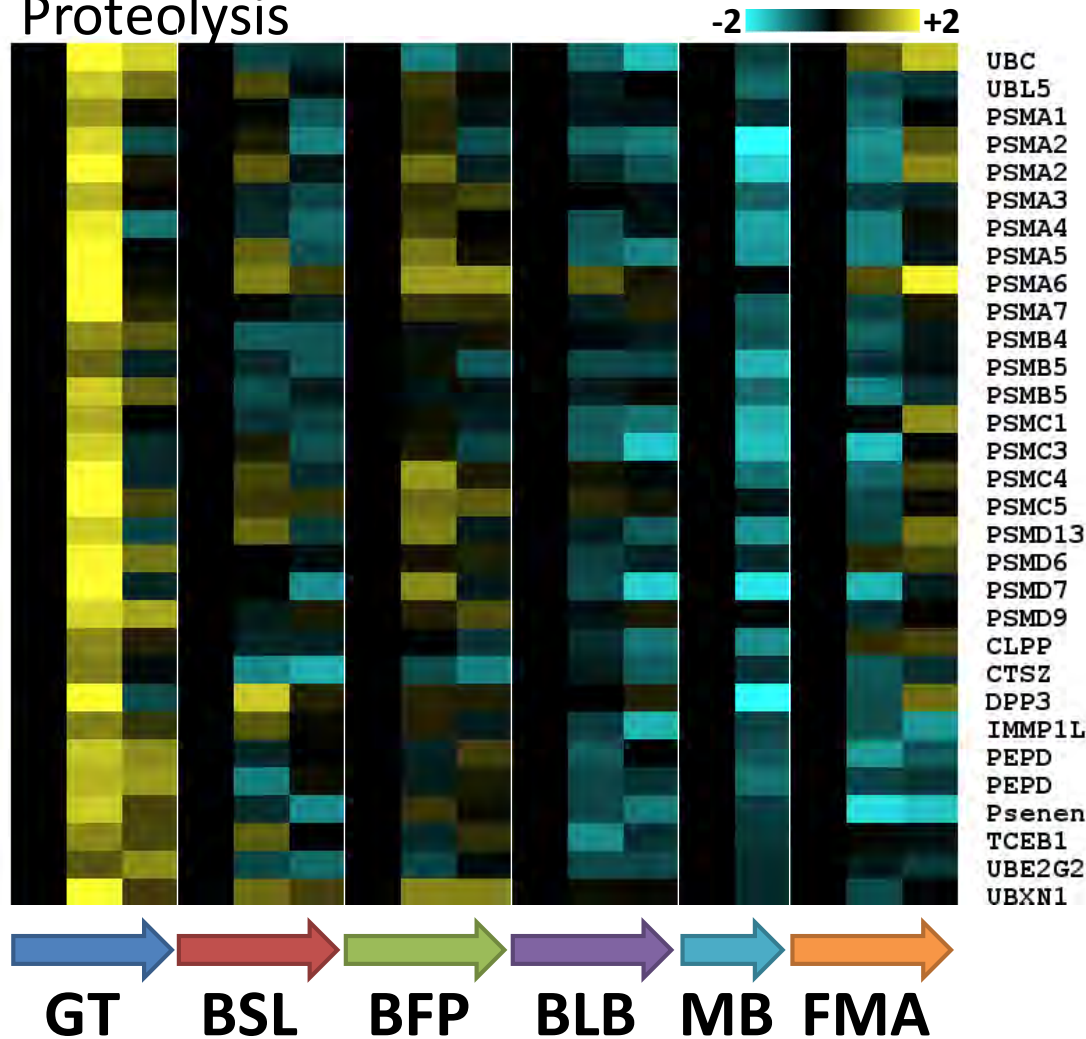


Figure Vol 446/29 March 2007 doi:10.1038/nature05663

LETTERS

Dioxin receptor is a ligand-dependent E3 ubiquitin ligase

Fumiaki Ohtake^{1,2}, Atsushi Baba², Ichiro Takada², Maiko Okada², Kei Iwasaki¹, Hiromi Miki², Sayuri Takahashi^{2,3}, Alexander Kouzmenko^{1,2}, Keiko Nohara¹, Tomoki Chiba¹, Yoshiaki Fujii-Kuriyama^{1,2} & Shigeaki Kato^{1,2}

Ligand (PAH)-activated AHR

↓
Ubiquitin-ligase activity

↓
Proteolytic degradation of sex steroid receptors

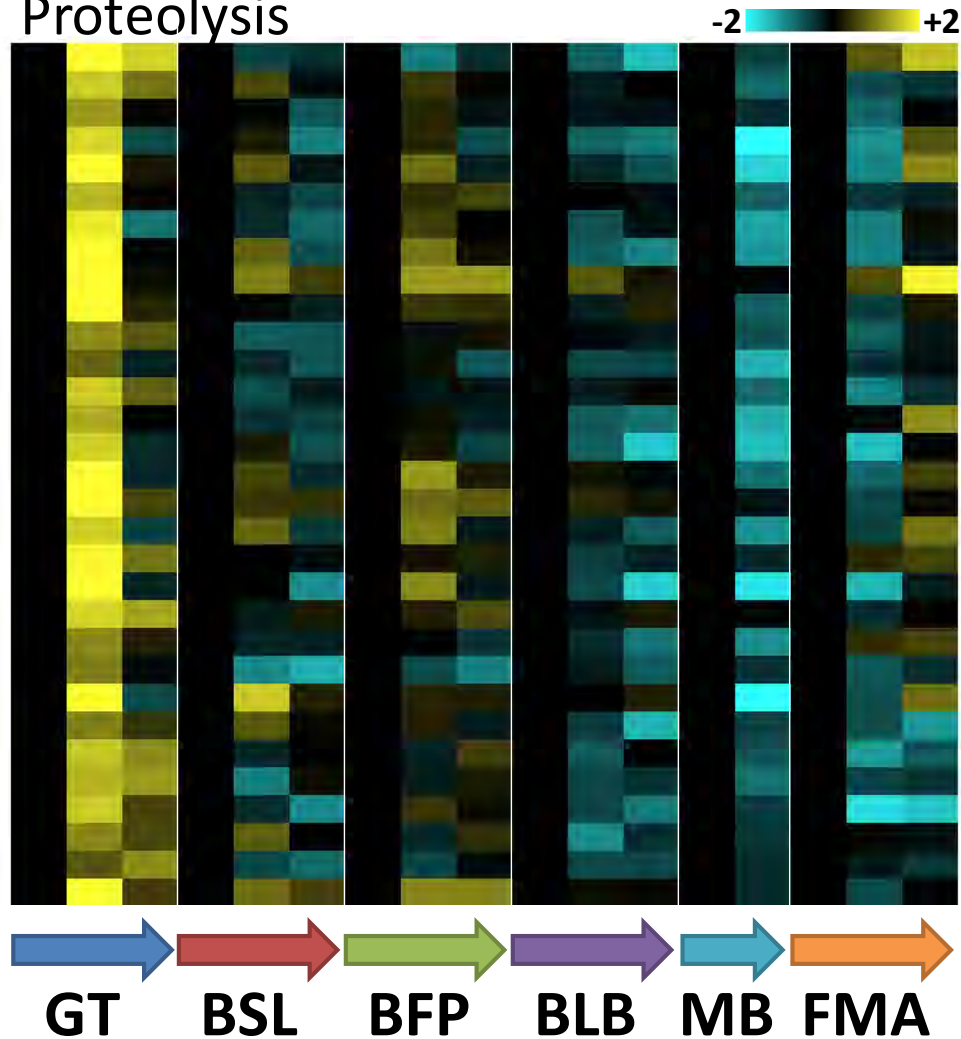
↓
Suppression of sex hormone induced gene expression

Genome expression response

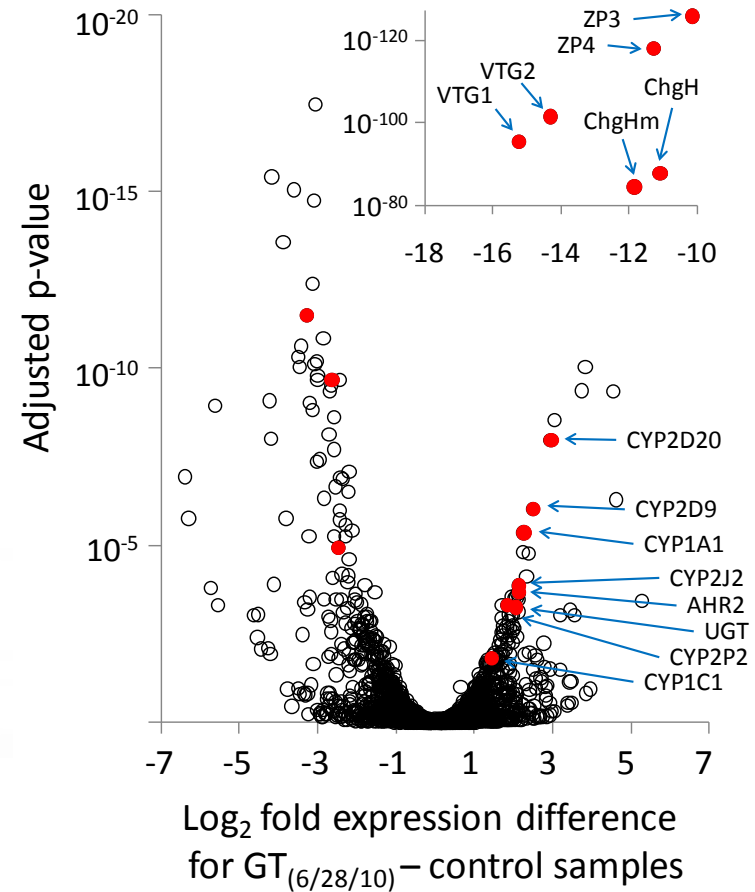
→ GT outlier



Proteolysis



- UBC
- UBL5
- PSMA1
- PSMA2
- PSMA2
- PSMA3
- PSMA4
- PSMA5
- PSMA6
- PSMA7
- PSMB4
- PSMB5
- PSMB5
- PSMC1
- PSMC3
- PSMC4
- PSMC5
- PSMD13
- PSMD6
- PSMD7
- PSMD9
- CLPP
- CTSZ
- DPP3
- IMMP1L
- PEPD
- PEPD
- Psenen
- TCEB1
- UBE2G2
- UBXN1



Genome expression response

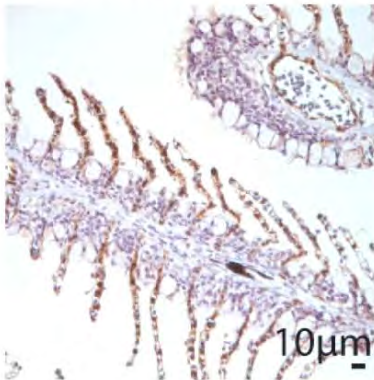
→ GT outlier

Collaborator: Fern Galvez
Student: Ben Dubansky



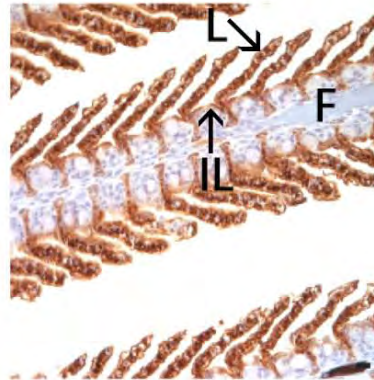
Trip 1

(5/1 to 5/9/2010)



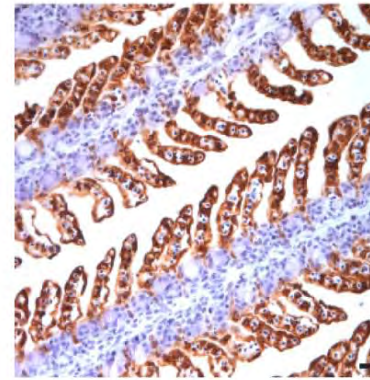
Trip 2

(6/28 to 6/30/2010)



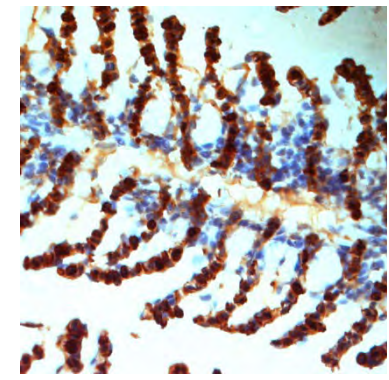
Trip 3

(8/30 to 9/1/2010)



Trip 4

(August 2011)



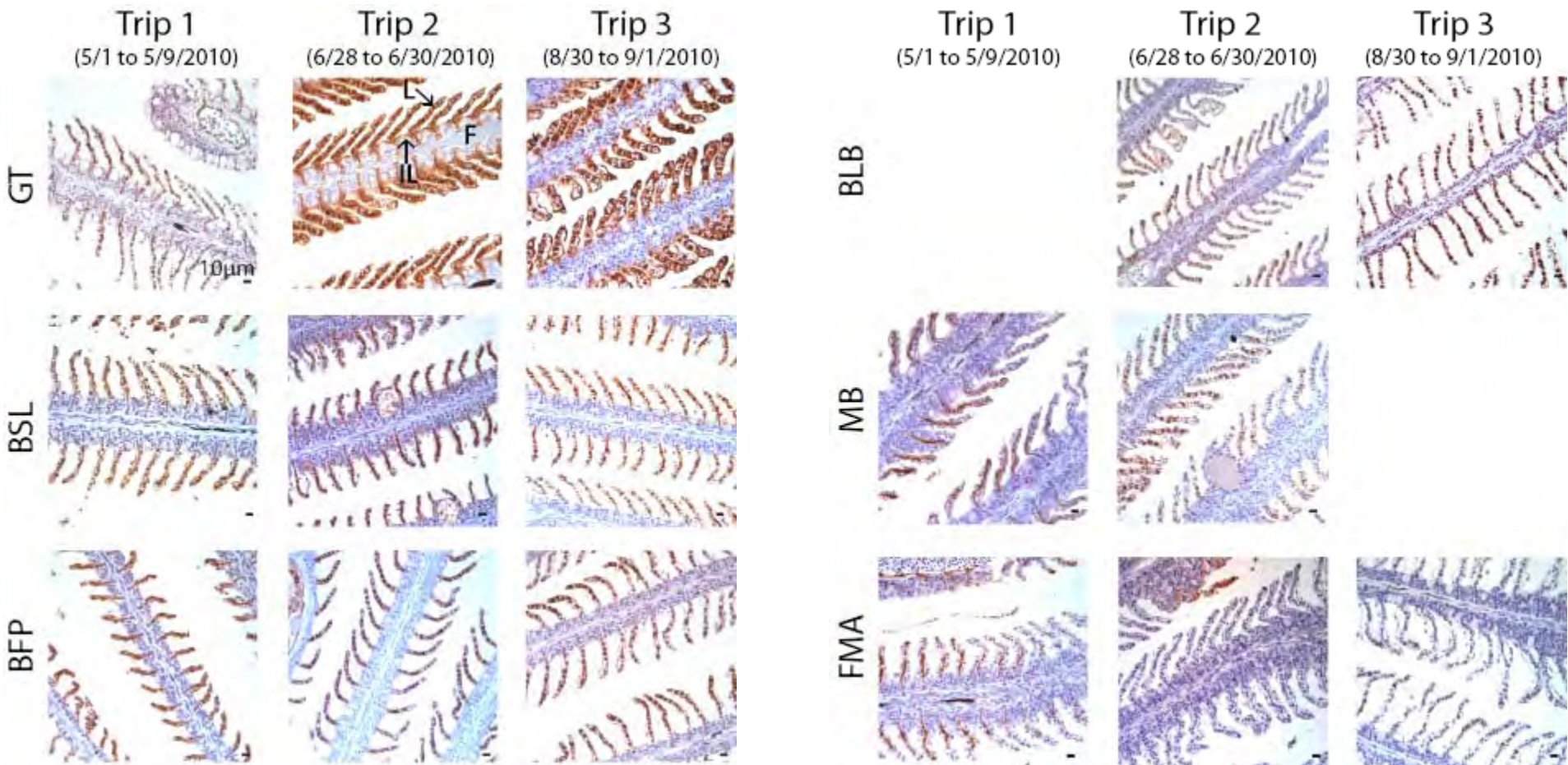
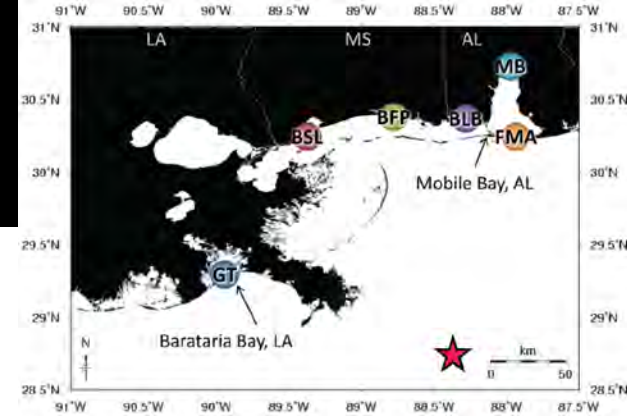
GT

Dark red stain = CYP1A protein in gills from fish collected *in situ*
Blue stain = hematoxylin (nuclei)

Genome expression response

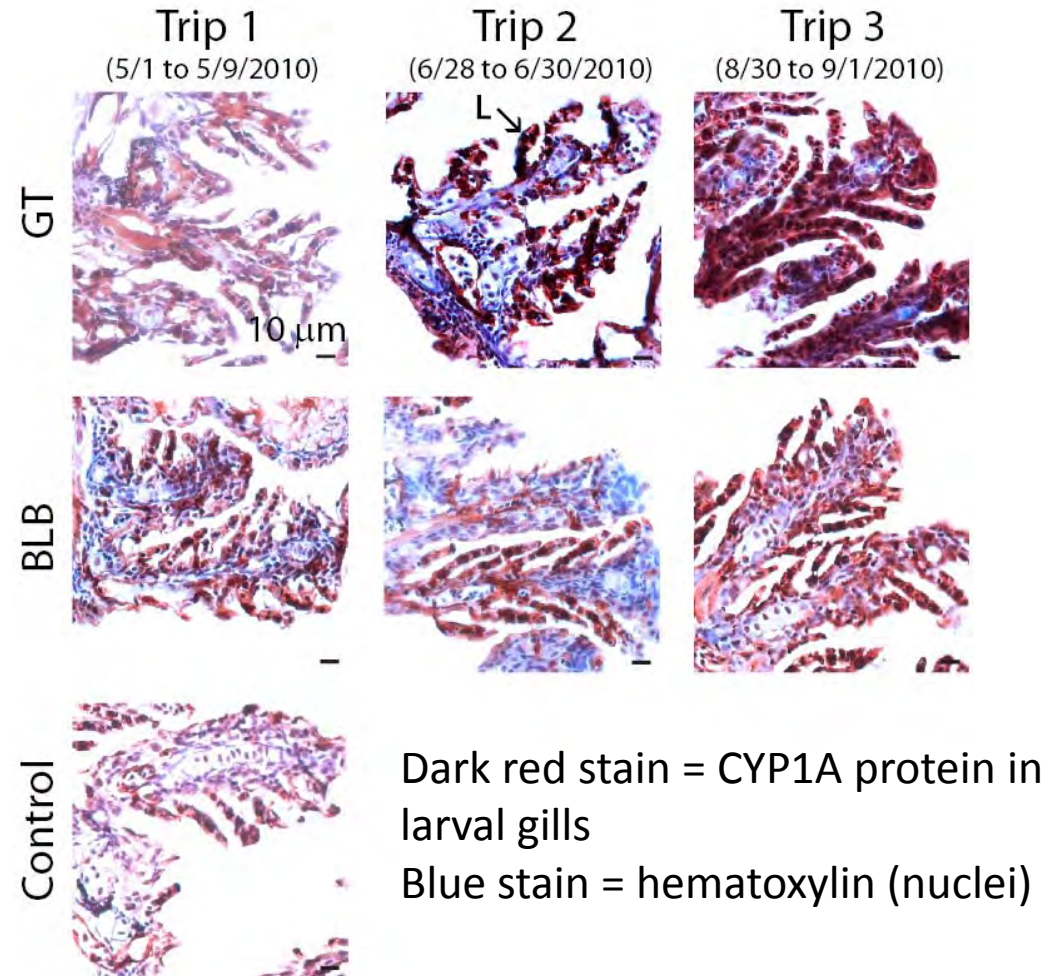
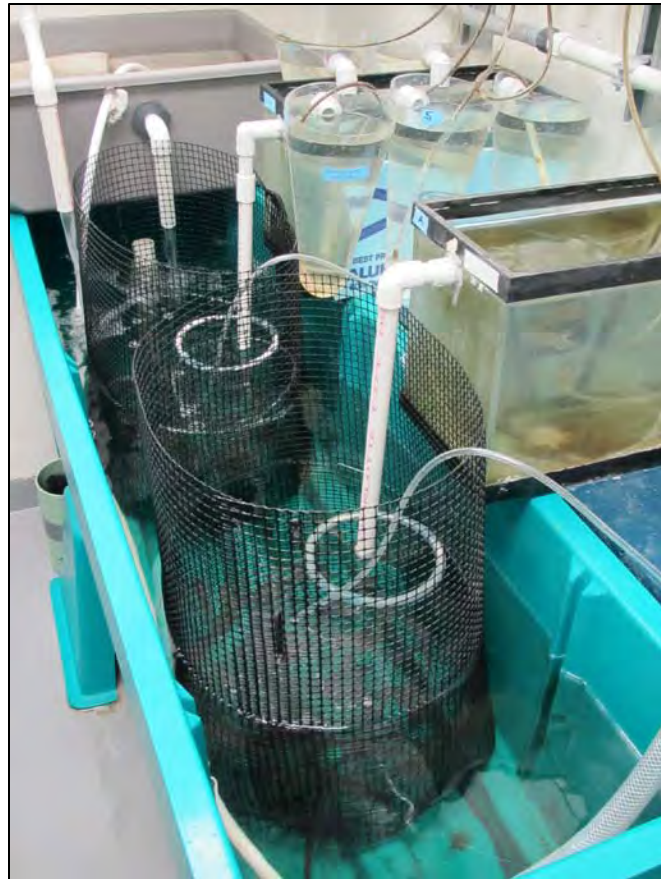
→ GT outlier

Collaborator: Fern Galvez
Student: Ben Dubansky



Early life-stage exposures to field-collected waters

Collaborator: Fern Galvez
Post-doc: Charlotte Bodinier



Early life-stage exposures to field-collected waters

No developmental abnormalities observed in killifish exposed to field-collected waters

BUT...

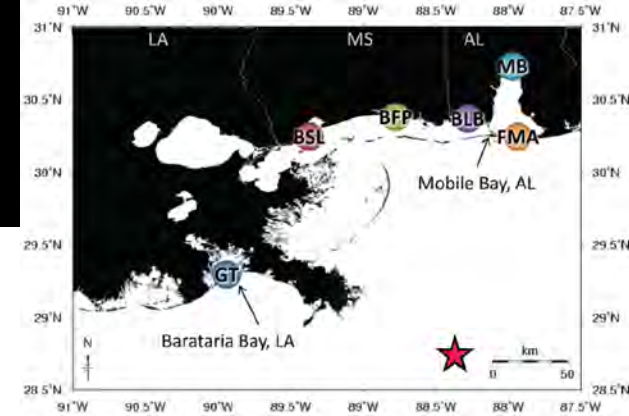


Sublethal exposure to crude oil during embryonic development alters cardiac morphology and reduces aerobic capacity in adult fish

Corinne E. Hicken^a, Tiffany L. Linbo^b, David H. Baldwin^b, Maryjean L. Willis^b, Mark S. Myers^b, Larry Holland^c, Marie Larsen^c, Michael S. Stekoll^a, Stanley D. Rice^c, Tracy K. Collier^{b,1}, Nathaniel L. Scholz^b, and John P. Incardona^{b,2}

PNAS Early Edition

→ very low-concentration exposures during development, insufficient to induce cardiovascular abnormalities in embryos, can impair cardiac performance in adulthood



DWH Oil Spill: NEXT?...

→ Consequences of multiple-stressor exposures?

→ Impacts on FITNESS

→ Consequences of low-level early life stage exposures (cardiac performance)?

→ How does evolutionary history and exposure history affect sensitivity?

Genomic and physiological footprint of the *Deepwater Horizon* oil spill on resident marsh fishes

Andrew Whitehead^{a,1}, Benjamin Dubansky^a, Charlotte Bodinier^a, Tzintzuni I. Garcia^b, Scott Miles^c, Chet Pilley^d, Vandana Raghunathan^e, Jennifer L. Roach^a, Nan Walker^e, Ronald B. Walter^b, Charles D. Rice^f, and Fernando Galvez^a

Departments of ^aBiological Sciences, ^cEnvironmental Sciences, and ^eOceanography and Coastal Sciences, and ^dCoastal Studies Institute, Louisiana State University, Baton Rouge, LA 70803; ^bDepartment of Chemistry and Biochemistry, Texas State University, San Marcos, TX 78666; and ^fDepartment of Biological Sciences, Clemson University, Clemson, SC 29634

Edited by Paul G. Falkowski, Rutgers, The State University of New Jersey, New Brunswick, NJ, and approved September 1, 2011 (received for review June 13, 2011)

The biological consequences of the *Deepwater Horizon* oil spill are unknown, especially for resident organisms. Here, we report results from a field study tracking the effects of contaminating oil across space and time in resident killifish during the first 4 mo of the spill event. Remote sensing and analytical chemistry identified exposures, which were linked to effects in fish characterized by genome expression and associated gill immunohistochemistry, despite very low concentrations of hydrocarbons remaining in water and tissues. Divergence in genome expression coincides with contaminating oil and is consistent with genome responses that are predictive of exposure to hydrocarbon-like chemicals and indicative of physiological and reproductive impairment. Oil-contaminated waters are also associated with aberrant protein expression in gill tissues of larval and adult fish. These data suggest that heavily weathered crude oil from the spill imparts significant biological impacts in sensitive Louisiana marshes, some of which remain for over 2 mo following initial exposures.

ecological genomics | ecotoxicology | microarray | RNA-seq |
toxicogenomics

Acknowledgements

Whitehead Lab:

- **Jen Roach** (Research Associate)
- **Eve McCullough** (Graduate student)
- **Whitney Pilcher** (Graduate student)
- **Reid Brennan** (Graduate student)
- **David Roberts** (undergraduate)

Funding:



• **DEB:**
*Evolutionary
Genetics*



Collaborators:

- **Fernando Galvez, Ben Dubansky, Charlotte Bodinier** (LSU-Biol.Sci.)
- **Nan Walker, Chet Pilley, Vandana Raghunathan** (LSU-EarthScan Lab)
- **Scott Miles** (LSU-Environ.Sci.)
- **Diane Nacci** (US EPA)
- **Ron Walter, Tzintzuni Garcia** (TX State U)
- **Charles Rice** (Clemson U.)