

The effects of crude oil on disease susceptibility and physiological responses of Gulf of Mexico fishes

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Disease susceptibility

- Immune impacts
 - Exxon Valdez oil spill
 - Increased occurrence of fish disease
 - Suppressed immune system
 - Limited understanding of oil effects

 - Gulf of Mexico
 - Different climate
 - Different water temperature
 - Different species
 - Different crude oil

- Research question:
 - How does crude oil affect the immune system and result in increased disease susceptibility?



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Disease susceptibility

- Physiological impacts
 - ▣ Ionoregulation compromised
 - ▣ Respiration effected
 - ▣ Focus on larval fishes

- ▣ Limited understanding of fishes, particularly Gulf of Mexico species

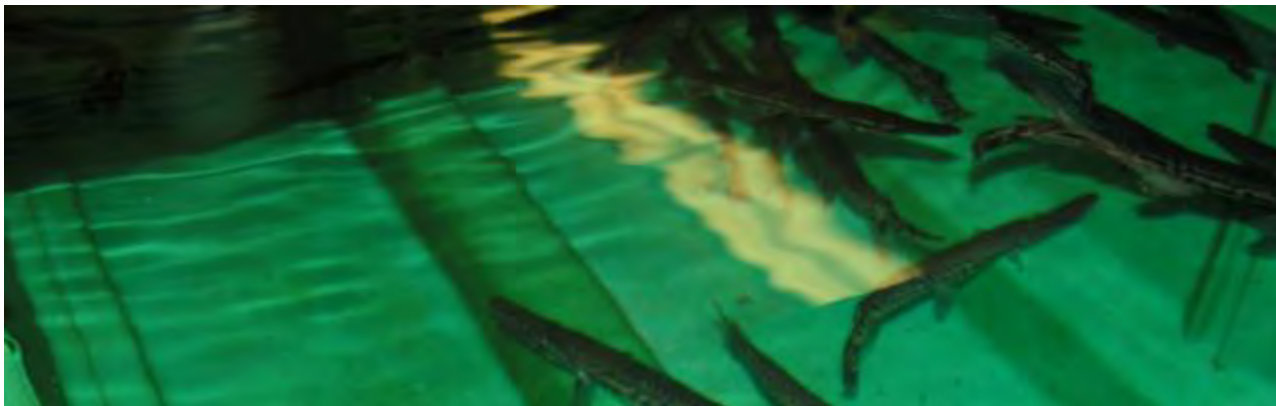
- Research question:
 - ▣ How does exposure to crude oil affect the physiology of different Gulf of Mexico fishes and relate to immune function?



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Objectives

- Collect samples from Gulf fishes representing a range of habitats and trophic levels
- Measure immunological and physiological impacts
- Compare with a controlled oil-exposure study



Field sampling

- Techniques
 - ▣ Blood and tissue samples

- Species
 - ▣ Alligator gar
 - Marshes near Terrebonne Bay, LA
 - ▣ Gulf killifish
 - Golden Meadow, LA
 - ▣ Red snapper
 - NOAA Oregon II



Estuarine fishes: alligator gar

- Wide distribution
- Movement between FW, estuarine and ocean habitats
- Top predator
- Bimodal breather

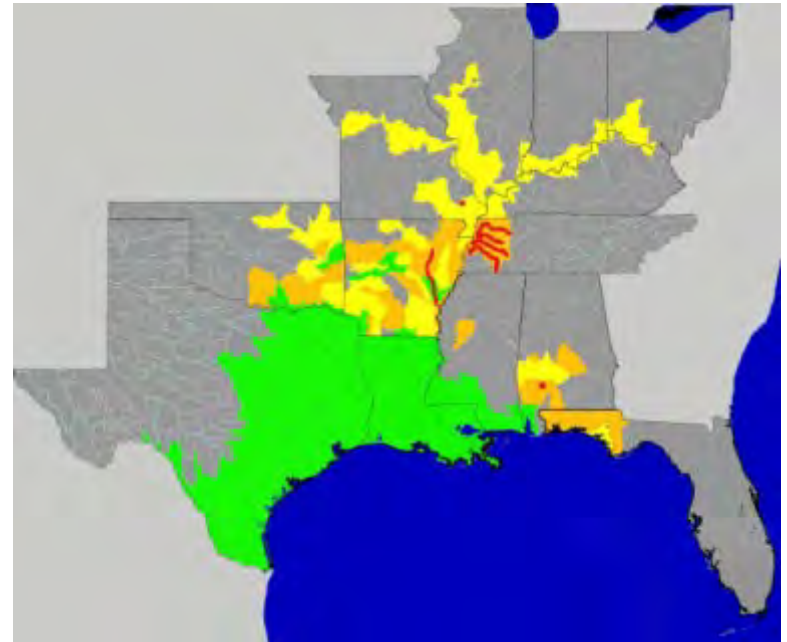


Image by: http://www.fws.gov/arkansas-es/A_Gar/AGar_Maps.html

Gulf fishes: Gulf killifish and red snapper

- Different trophic levels
- Near shore and off-shore





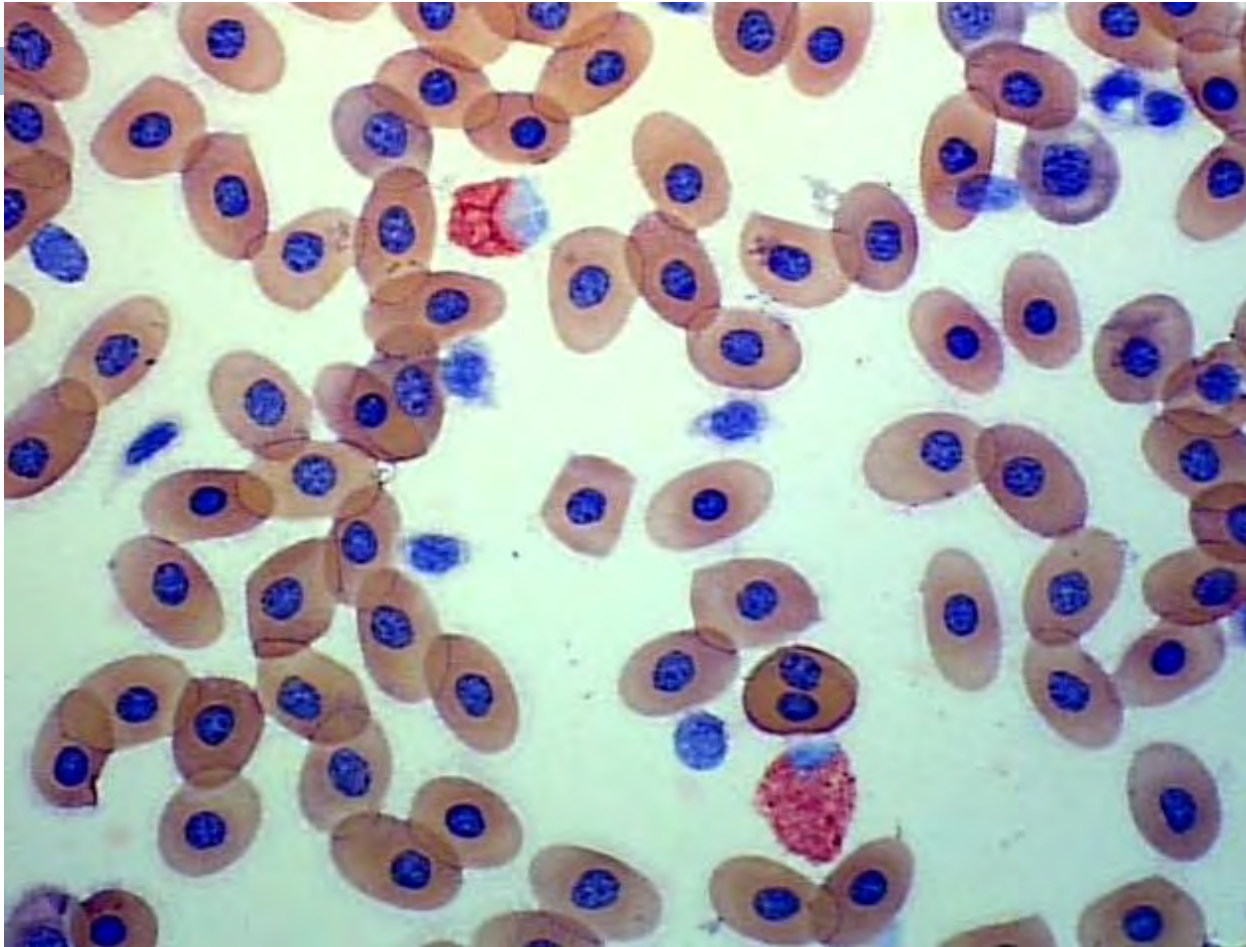
STEERING ROOM
NATURAL VENT



Analyses

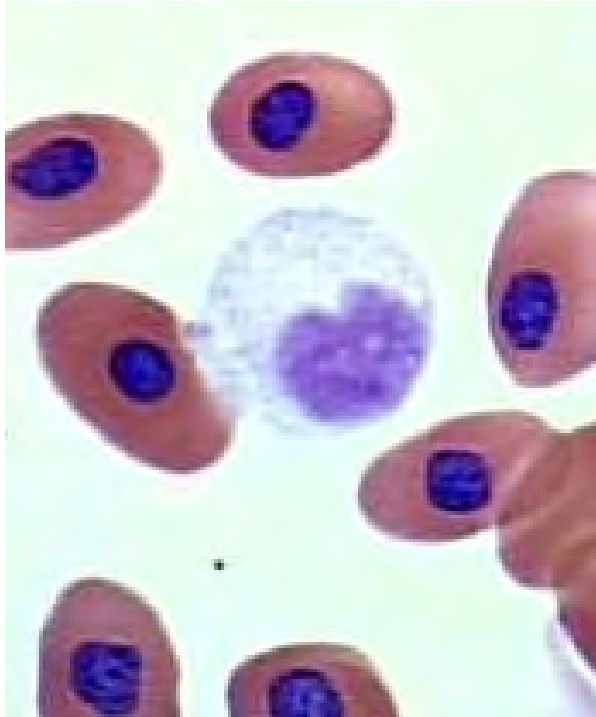
- Analyzed blood
 - Flow cytometry
 - Differentiating cell types
 - Erythrocytes
 - Leukocytes (immune response)
 - Granulocytes (eosinophils and neutrophils)
 - Monocytes
 - Lymphocytes
 - Histology

Results

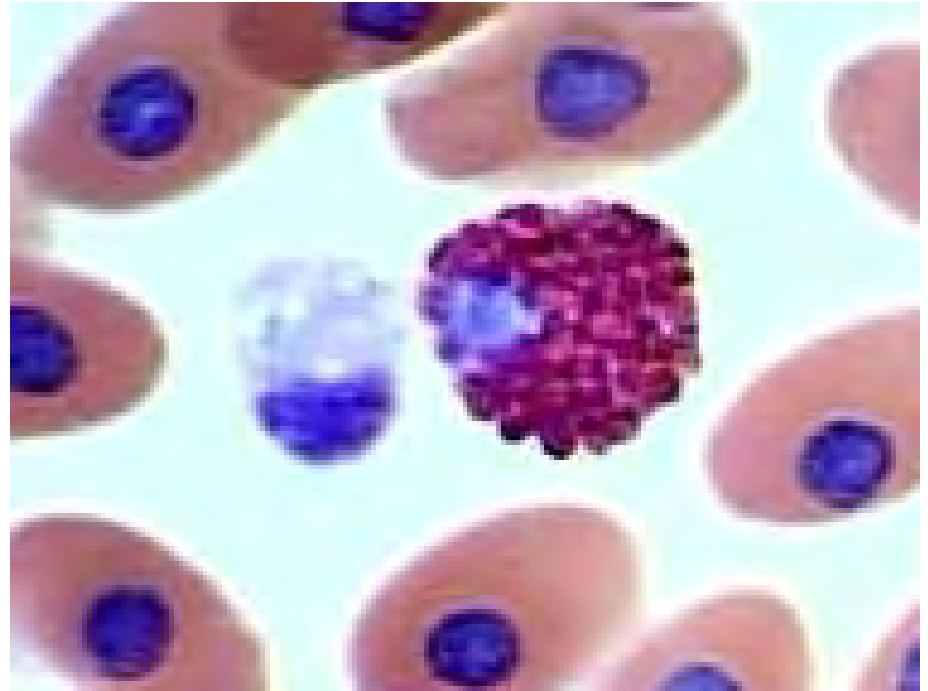


Gar erythrocytes (RBCs), leukocytes: eosinophils (bright red granules), small lymphocytes, thrombocytes

Results

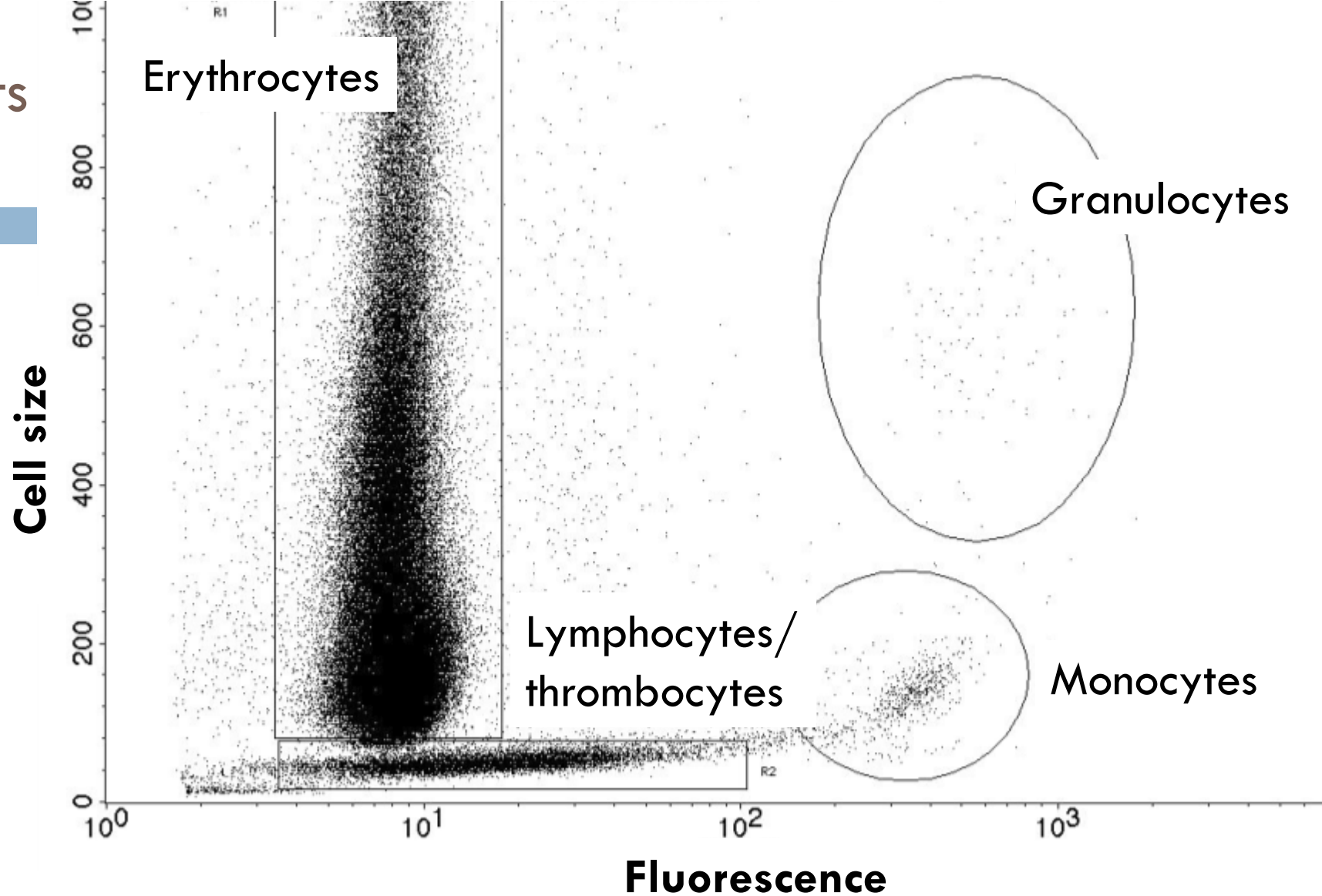


Leukocyte: monocyte



Leukocytes: neutrophil and eosinophil (red) – both granulocytes

Results



Alligator gar leukocyte populations (stained with DiOC₆)

FACSAria III Flow Cytometer

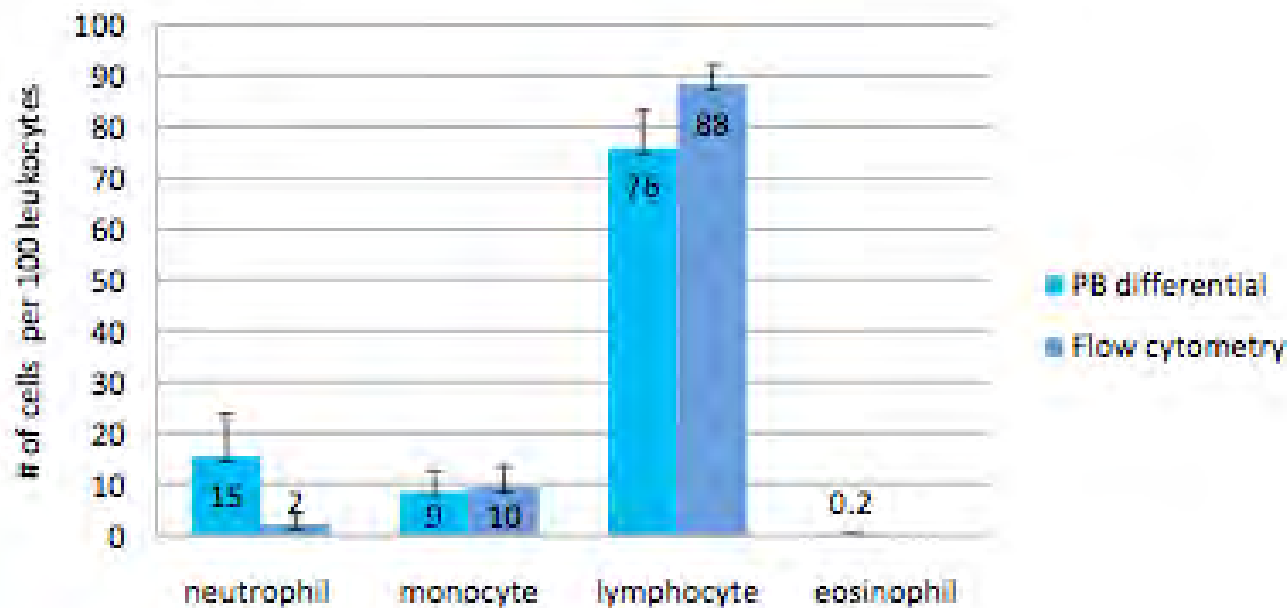
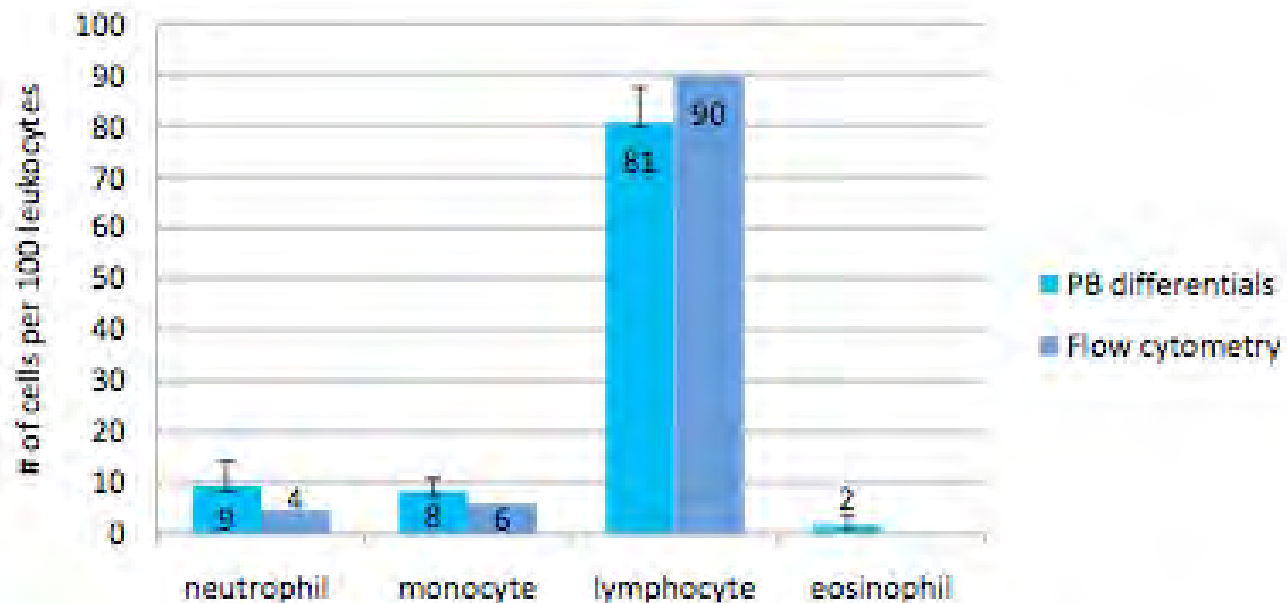
Results:

flow cytometry

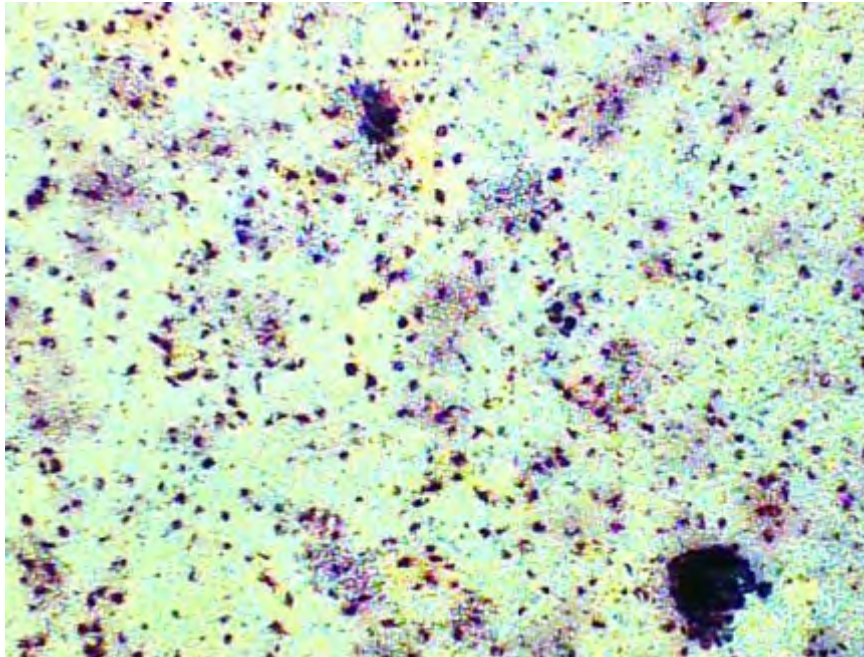
Alligator gar:

□ Control

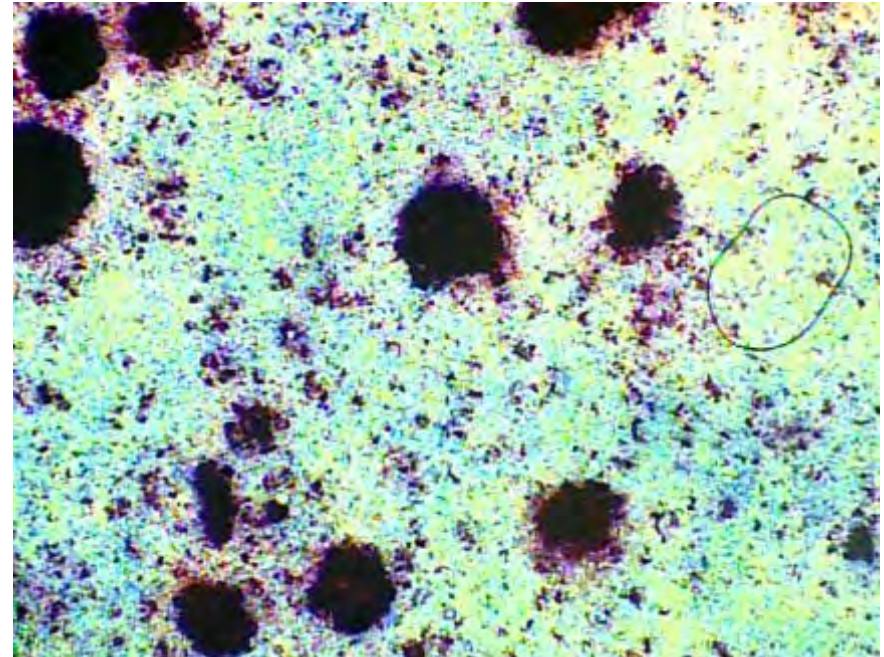
□ Gulf coast



Results: histology

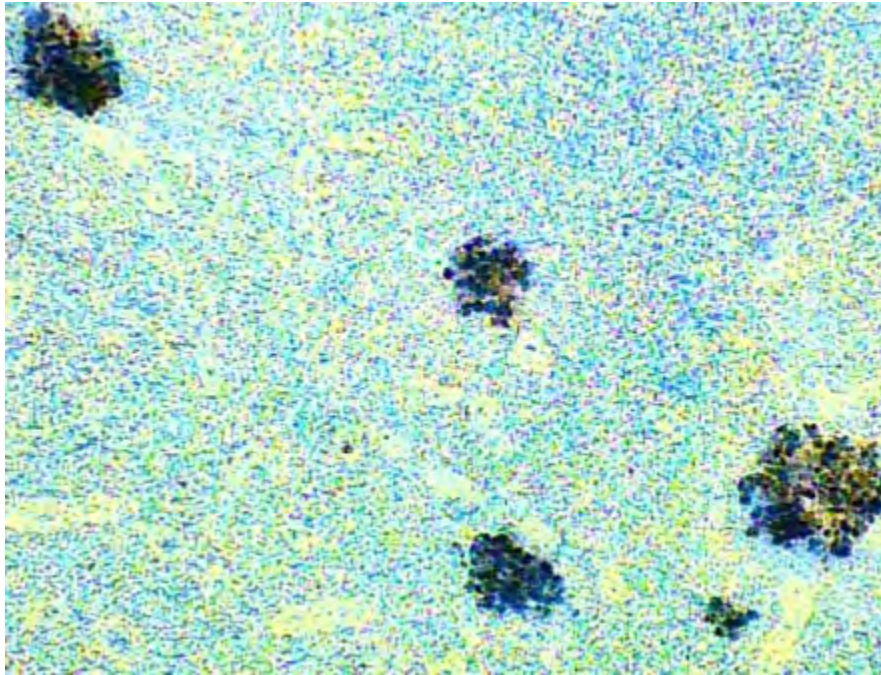


Control *F. grandis* spleen: AP positive cells demonstrating leukocytes

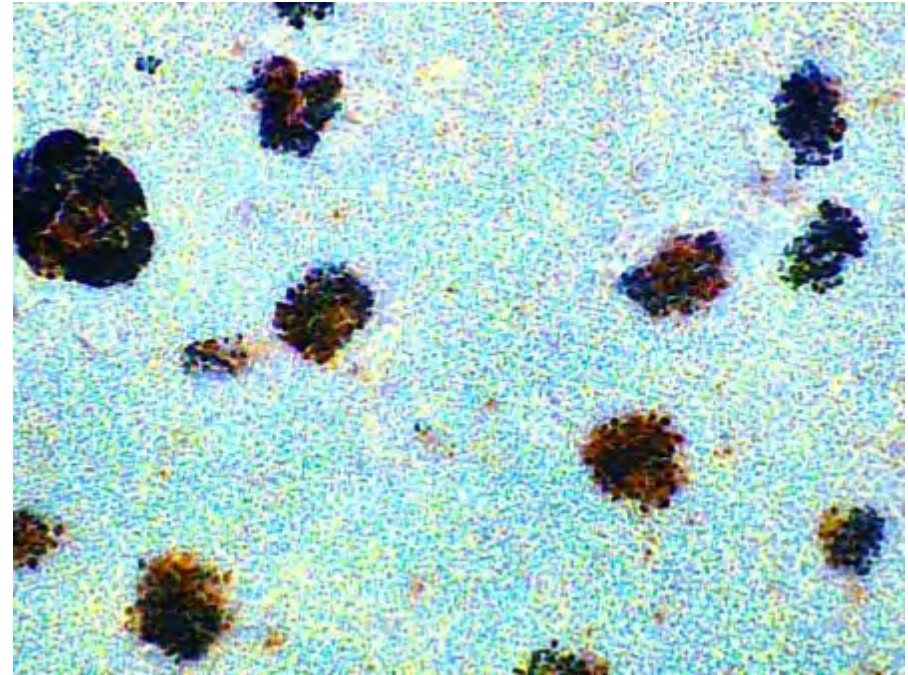


Oil-exposed *F. grandis* spleen: AP positive cells
- Increased # and size of melanomacrophage centers.

Results: histology



Control *F. grandis* spleen: BG positive cells demonstrating lymphocytes



Oil-exposed *F. grandis* spleen: BG positive cells

- Increased # and intensity
- Increased # and size of melano-macrophage centers.

Analyses

- Currently analyzing tissues
 - EROD activity (liver)
 - Metabolomics (liver and muscle)
- Controlled oil-exposure studies

Laboratory experiments

- Controlled WAF oil exposure
- Alligator gar
- Serial sampling





Measurements

- Ionoregulation
 - ▣ Salinity challenge
 - ▣ Ability to move between habitats

- Metabolomics
 - ▣ Study of metabolite profiles of cellular processes

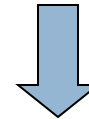
 - ▣ Metabolites: small MW compounds involved in metabolism
 - Amino acids, fatty acids, lipids, carbohydrates



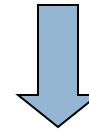
Information flow in biology

Genomics

DNA



RNA



Transcriptomics

Proteins



Proteomics

Metabolites

Metabolomics

Role of metabolites

- Specific combinations of metabolites reflect:
 - Normal body processes
 - Organ function
 - Stressful state
 - Diet / food quality
 - Allergenicity
 - Toxicity



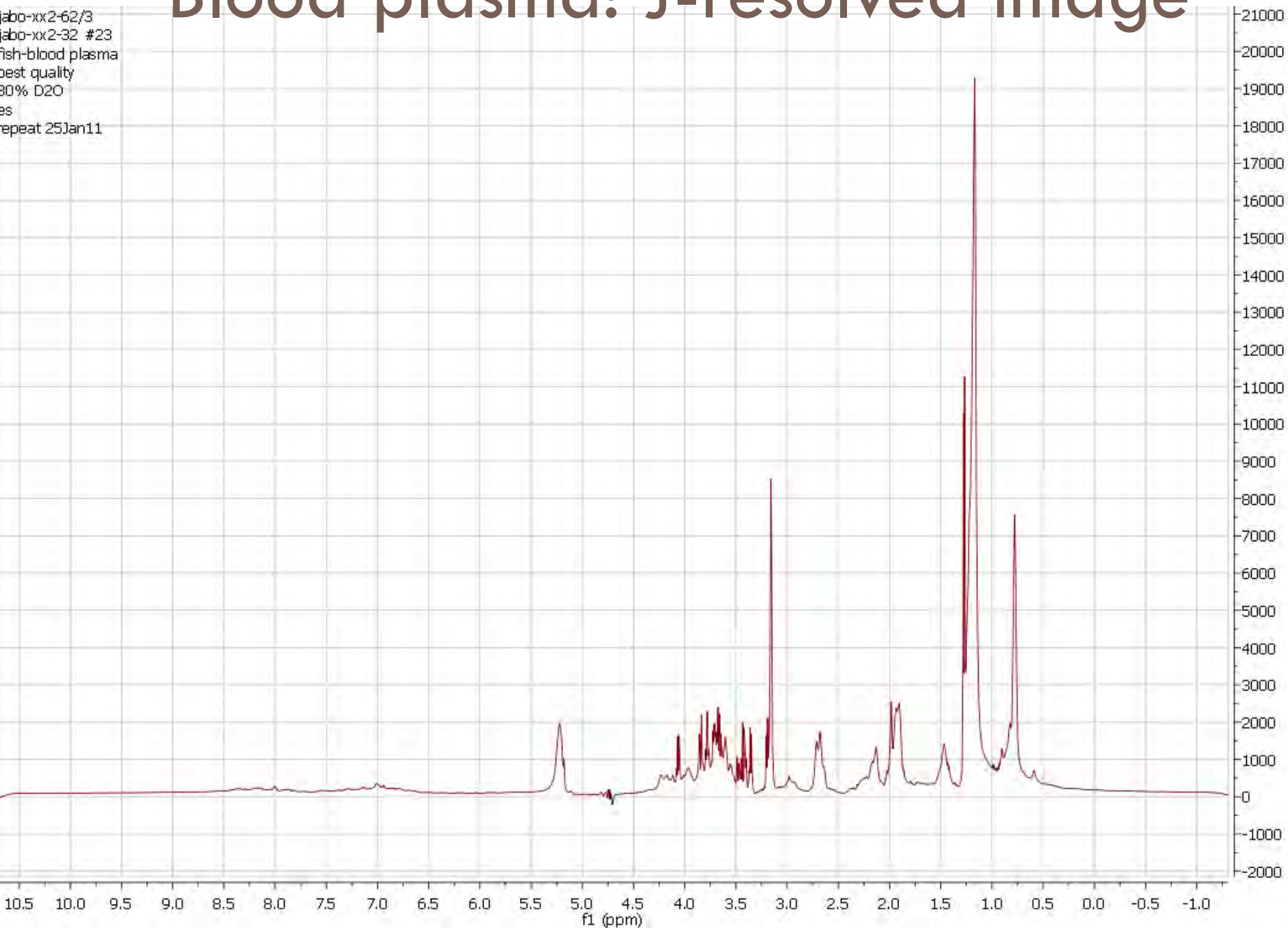
NMR FACILITY - UAB

- **Nuclear magnetic resonance (NMR) spectroscopy**
 - Cryoprobe



Blood plasma: J-resolved image

labo-xx2-62/3
labo-xx2-32 #23
fish-blood plasma
best quality
30% D2O
es
repeat 25Jan11

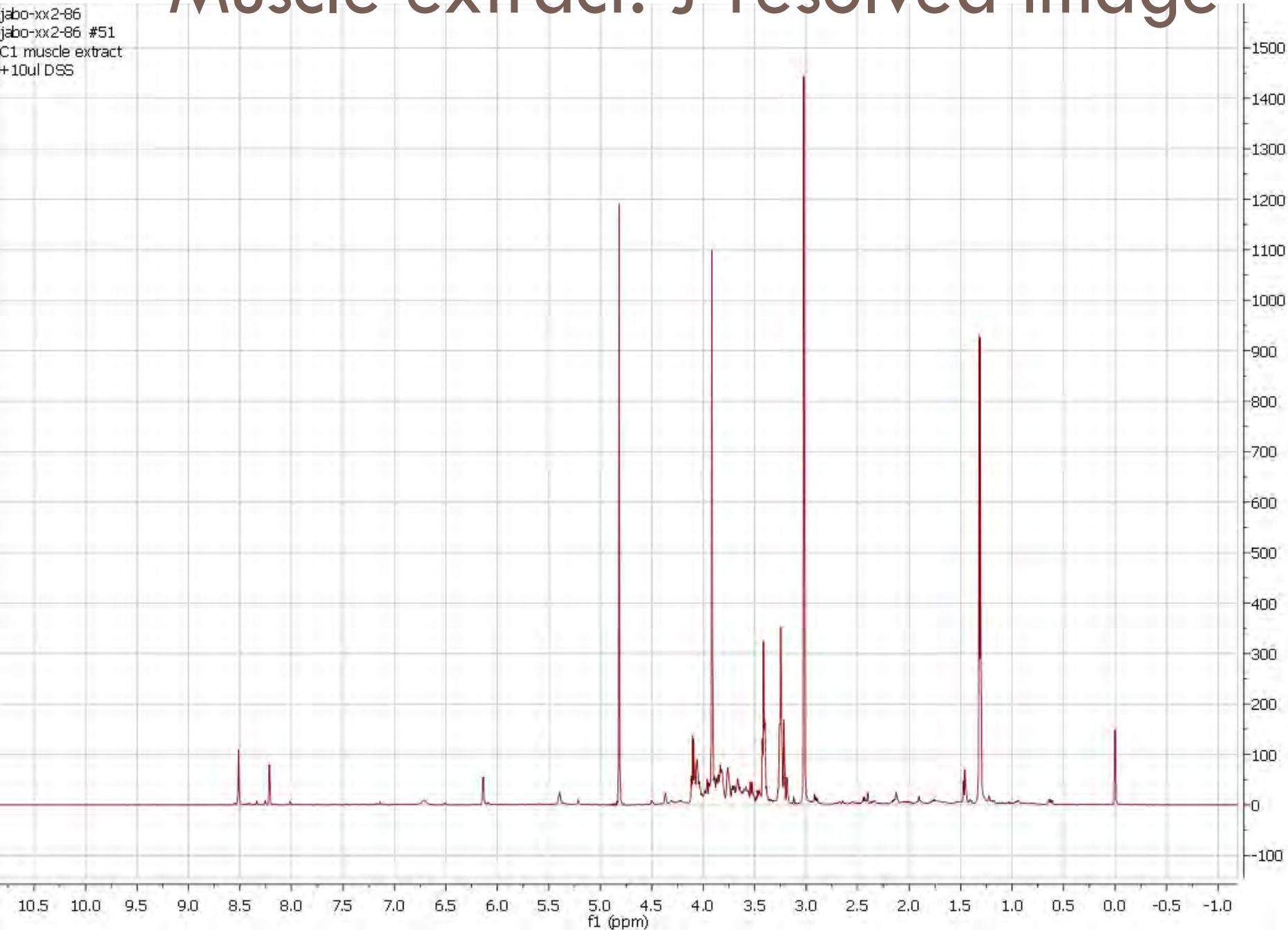


Liver extract: J-resolved image

jabo-xx2-86
jabo-xx2-86 #31
C2 liver extract
+ 10ul DSS



Muscle extract: J-resolved image



Summary: early results

- Disease susceptibility
 - ▣ Crude oil suppresses immune function
 - ▣ Analyses of samples underway
- Controlled-exposure studies underway
 - ▣ Understand nature of immune impacts
 - ▣ Understand time-course of impacts
 - ▣ Determine physiological impacts

Acknowledgments

■ People

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■ Crude oil - BP

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