

# Introducing Metabolomics: the Biochemical Balancing Act

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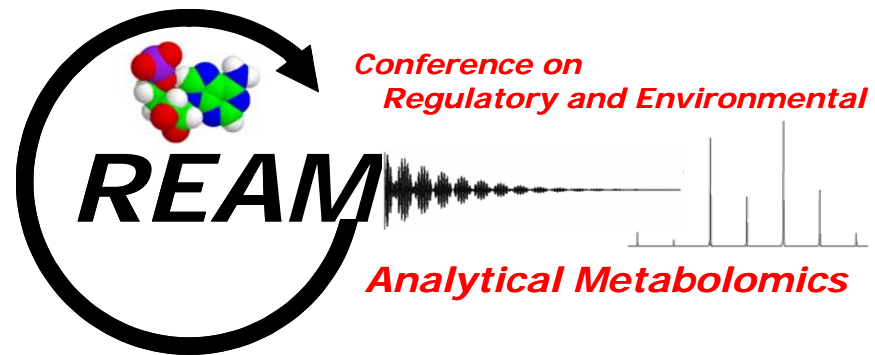
Jefferson County Public Schools

# Outline

- High School at U of L
- Metabolomics
- Universality of many chemical reactions
  - Ubiquitous Oxidation reactions
  - Identical reactions and/or products can have very different impacts on biological systems
- Telling one reaction from another
  - Isotopomer analysis as a tool
  - Using metabolomics to find answers to cancer

# Introducing CREAM

- Center for Regulatory and Environmental Analytical Metabolomics - 2005
- Sponsors:
  - National Science Foundation
  - State of Kentucky
  - University of Louisville
- High School Outreach, so far
  - Cancer cell and lipid metabolism labs
  - Four High School, one Junior High, students
  - Six posters at Regional Science Fairs, one Navy Research Institute award winner.



# Supporting Teachers' Science Education

- Survey
  - How can CREAM provide information and support?
  - Form a cooperative to share resources, support, and experience.
- Provide resources to add to your toolbox
  - CD-ROM to take home today
  - Help answer students' questions about relevance of science today

# Future Outreach

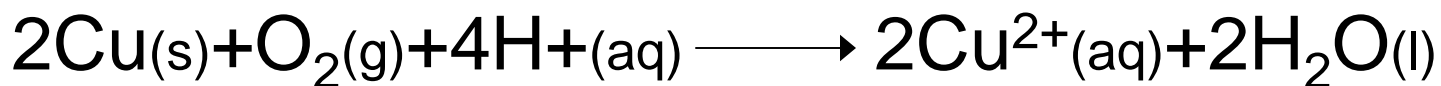
LTQ FT-MS

- Planning a Teacher Day at U of L
  - Professors from several departments
  - Tour laboratories
  - See cutting-edge research in action
- “Science immersion day”
  - Give you a chance to explore and indulge your science curiosity
  - Enhance your current science knowledge, giving you more to take into the classroom



# Oxidation-Reduction

- Reactions in which electrons are transferred between atoms.
  - Electron gain = reduction
  - Electron loss = oxidation
- Green surface on Copper roofs



- Hydrated copper (II) carbonate and sulfate formed from oxidation
- in the presence of atmospheric  $\text{CO}_2$  and  $\text{SO}_2$

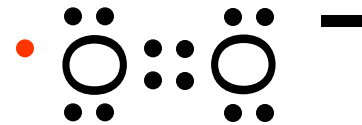


# Reactive Oxygen Species

- Ozone ( $O_3$ ), hydroxyl radicals (OH), atomic oxygen (O), hydroperoxy radicals ( $HO_2$ ).
- Both Photochemically and Enzymatically derived
- “Radical” is atom with an **unpaired electron**



Hydroxyl  
radical



Superoxide  
anion

# Ozone in the Environment

- Stratosphere
  - 10-50 km from surface
  - Absorbs 98% UV light
- Troposphere
  - $\text{NO}_x$  & VOC w/sunlight
  - Affects biotic health

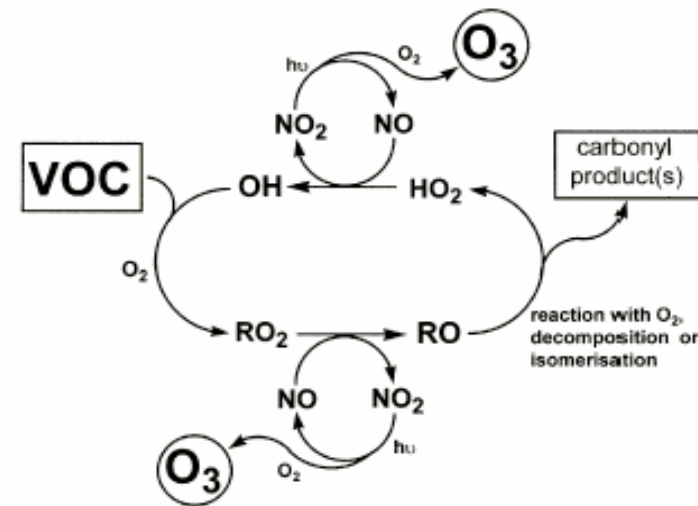
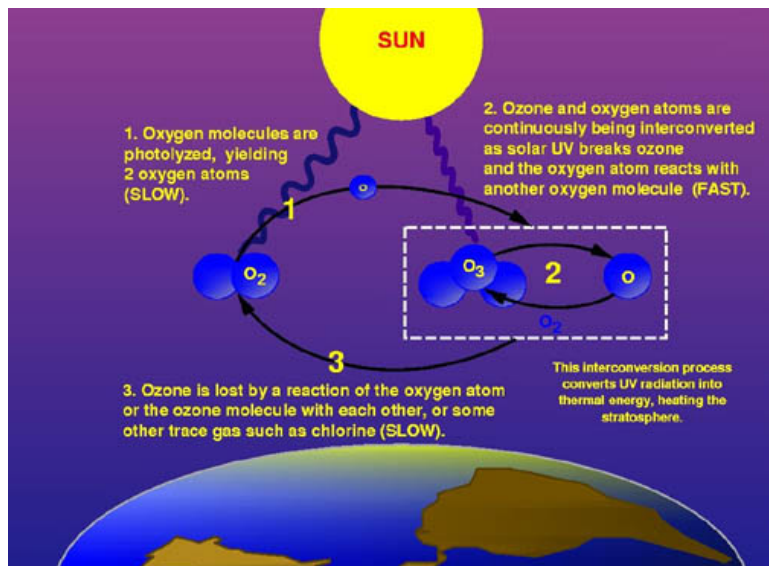


Fig. 1. Schematic representation of the free radical catalysed oxidation of a VOC into its first generation carbonyl product(s) in the presence of  $\text{NO}_x$ , and the associated generation of ozone ( $\text{O}_3$ ).



# ROS formation in the cell

- Ionizing radiation
- Byproduct of cellular respiration
  - Unavoidable
  - Electron transport chain leakage
  - Produce the superoxide anion
- Synthesized by enzymes in phagocytic cells
  - Neutrophils
  - macrophages

# ROS activity in the cell

## Damages cell structures

- Hydroxyl radical acts on lipids in mitochondrial membranes
- Crosslinkages are formed
- Deformed lipids damage the membrane

Mitochondria Structural Features

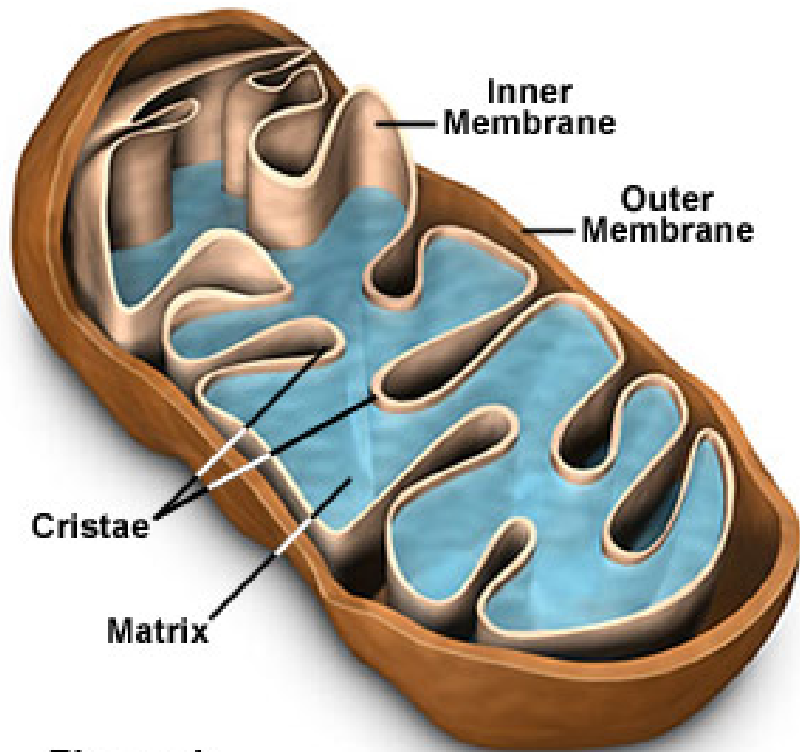
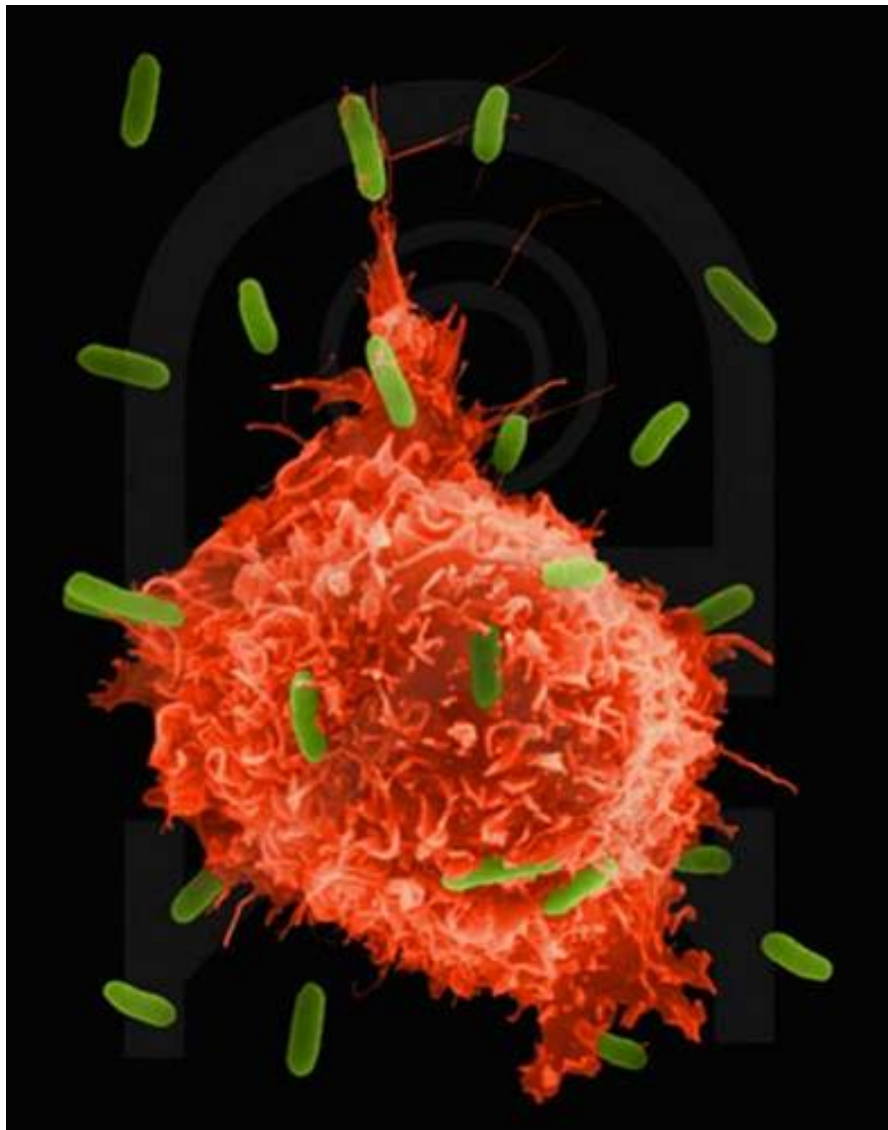


Figure 1

# ROS activity in the cell



## Essential to cell health

- Thyroid gland uses  $\text{H}_2\text{O}_2$  to attach Iodine atoms
- Macrophages use the superoxide anion for phagocytosis
- Neutrophils use  $\text{H}_2\text{O}_2$  to produce antiseptic hypochlorite ion



## Life without ROS?

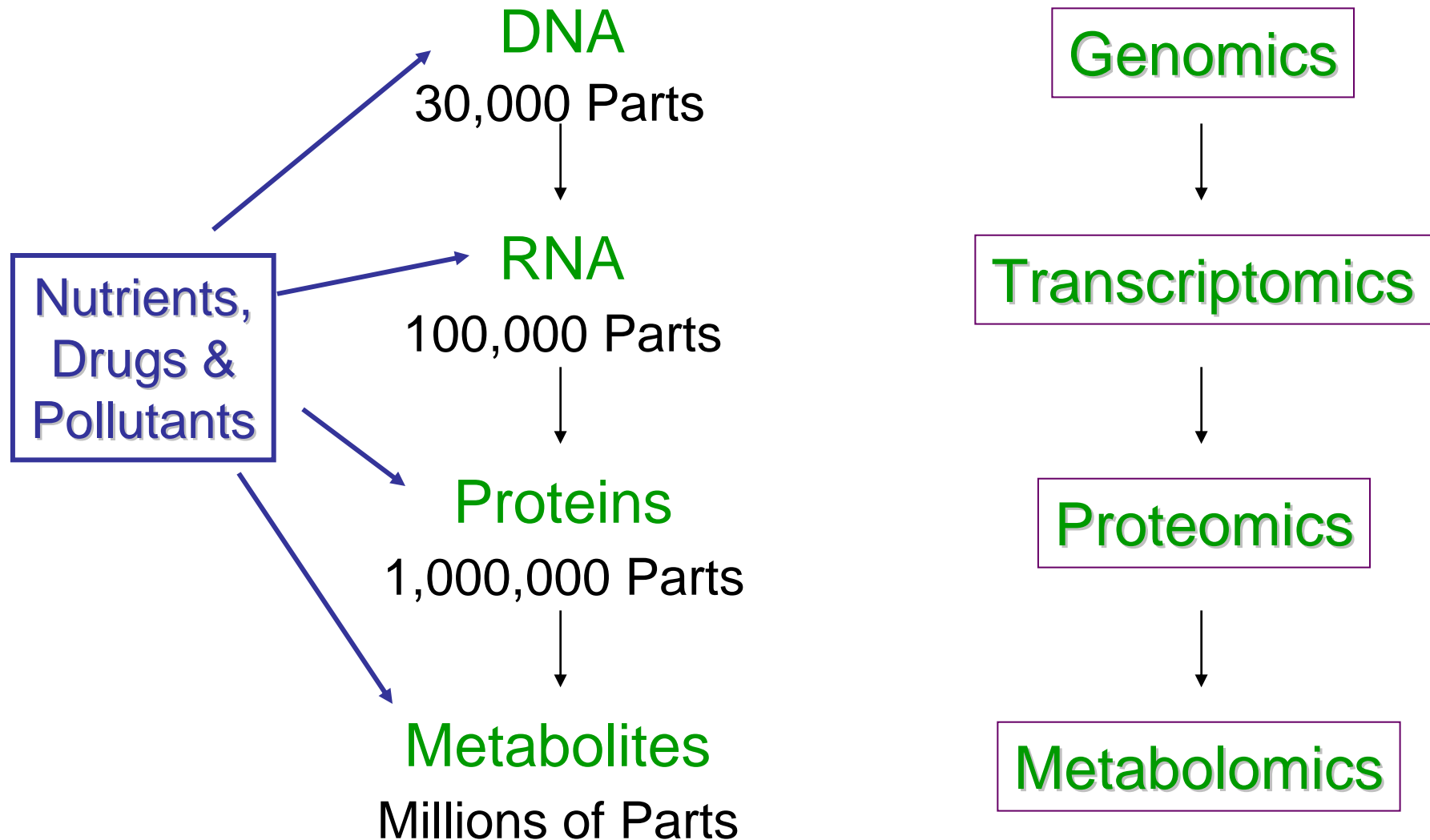
- Oxidation underpins most energy used by humans
  - Combustion
  - Metabolism
- Managing oxidative products allows life to continue
  - In the cell and
  - In the environment

# Metabolomics

- Metabolic pathways are a sequence of reactions
  - Take molecules apart (catabolism)
  - Build molecules – usually from “scraps” (anabolism)
- The same reaction can be either destructive or supportive depending on the pathway it is associated with
- Many pathways produce the same products (metabolites)

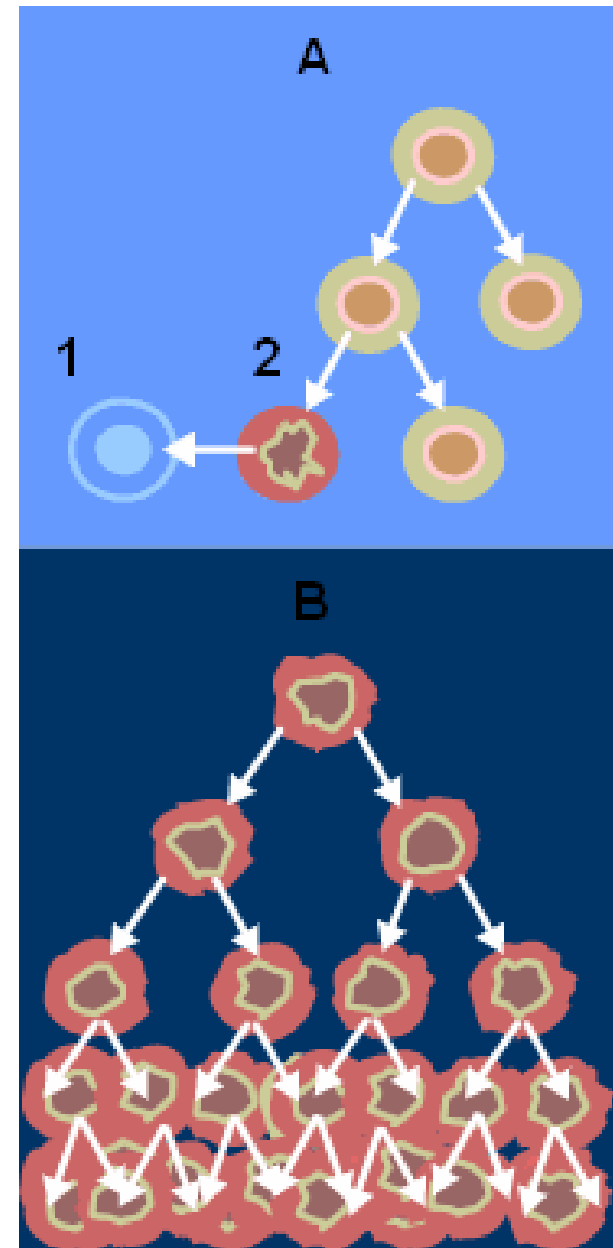
**A tool to identify the metabolic sources of chemically identical molecules.**

# The New World Order of “Omics”



# What is Cancer?

- Caused by abnormal genetic material
  - They exist side-by-side with “normal” cells
- Cells are aggressive, invasive, metastatic
  - They exhibit uncontrolled growth
- Identify cancer cells by characteristic metabolism



# Metabolomics in Cancer Research

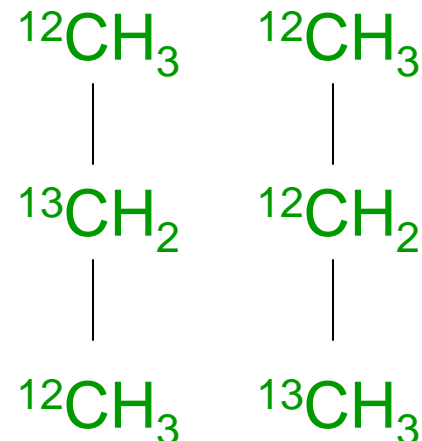
- Diagnostic **biomarkers**: compounds only formed by cancer cells that can be detected in biofluids (blood, urine, etc.)
- Test proposed treatments: does the metabolism of cancer cells slow down?
- Discover new treatments: Block metabolism used exclusively by cancer cells



# Isotopomers (isotopic isomers)

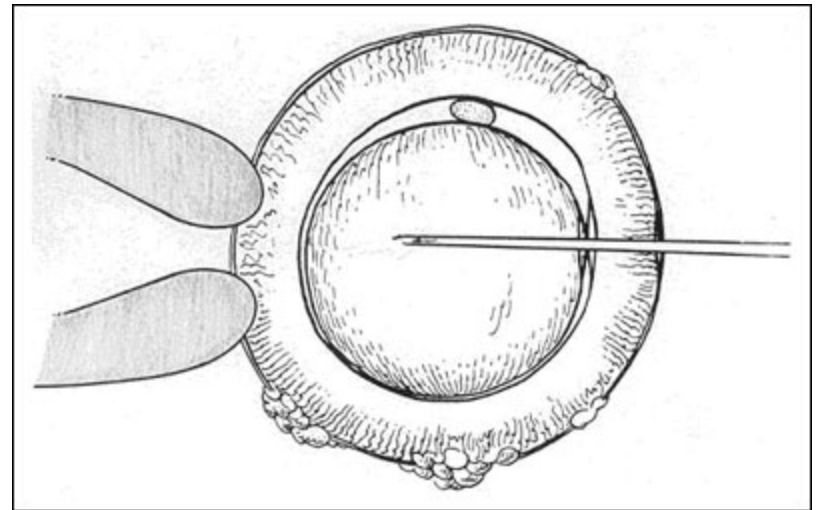
- **Isomers** with the same number of each **isotopic** atom but differing in their positions
  - Number of neutrons + protons - Mass number
  - molecules with the same chemical formula but in which the atoms are arranged differently
- Radiocarbon dating uses radioisotope  $^{14}\text{C}$
- Labeling uses stable isotope  $^{13}\text{C}$

Symbol	$^{12}\text{C}$	$^{13}\text{C}$	$^{14}\text{C}$
Neutrons	6	7	8
Protons	6	6	6
abundance	99%	1 %	1ppt



# How are isotopomers created?

- Inject uniformly labeled glucose (all C are  $^{13}\text{C}$ )
- Leave the cell to “do it's thing” for 24 hours
- Find that the cell now has fatty acids with  $^{13}\text{C}$  carbons.
- How did the  $^{13}\text{C}$  get into the fatty acids?
  - Magic?
  - Osmosis?
- **ONLY** Metabolism can take the molecule apart and build another one.





National Geographic News Jan. 12, '06

## Dogs detect cancer

- in human breath
- 88-97% accuracy

# Biomarkers

- Metabolite in biofluid (e.g. blood)
- Need high
  - Sensitivity
  - Specificity
- Isotopomer analysis sorts metabolites by pathway
- Those from cancer cells can be identified

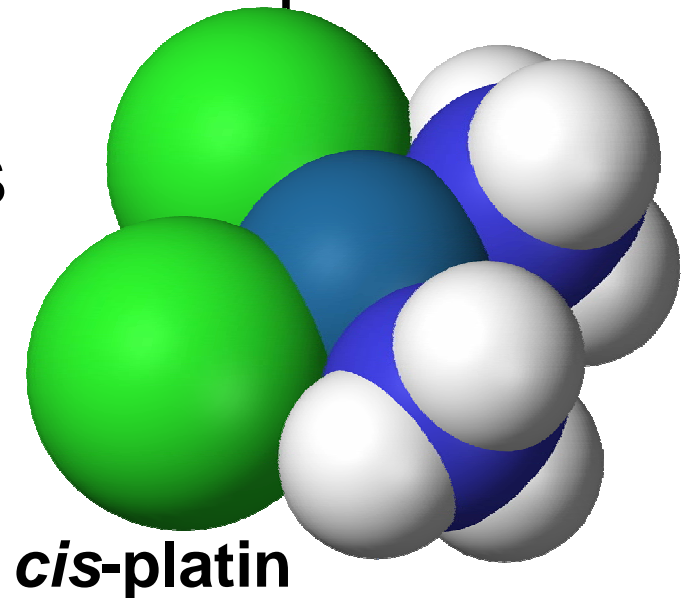
# Tissue resection studies

- Infuse  $^{13}\text{C}$  glucose
- Remove normal and cancer tissue
- Compare pathways used by different cell types
- Helpful to many projects:
  - Biomarkers
  - Treatment evaluation
  - Drug discovery

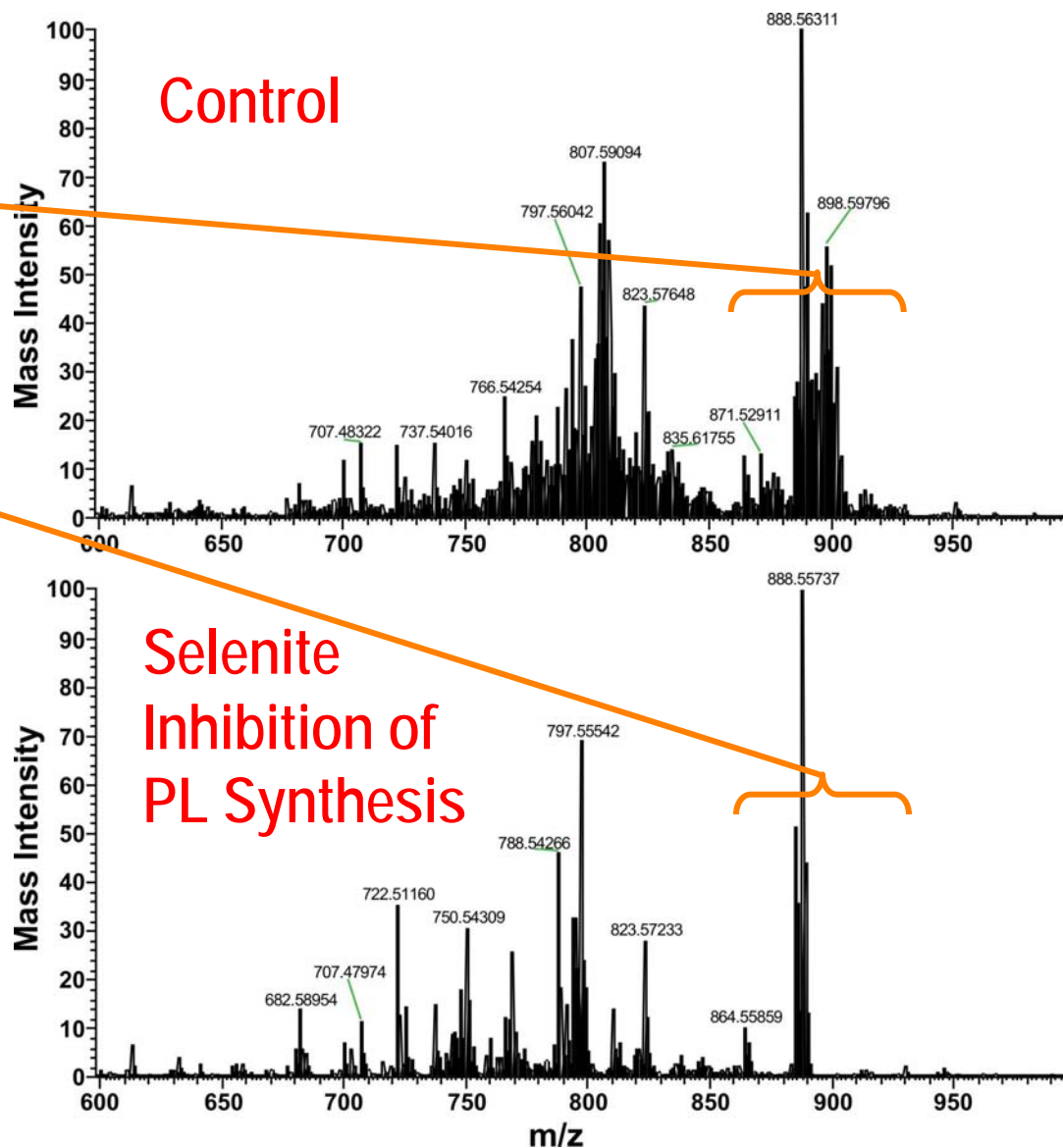
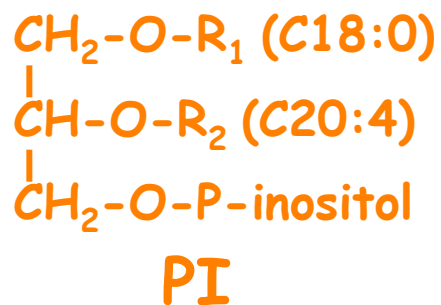
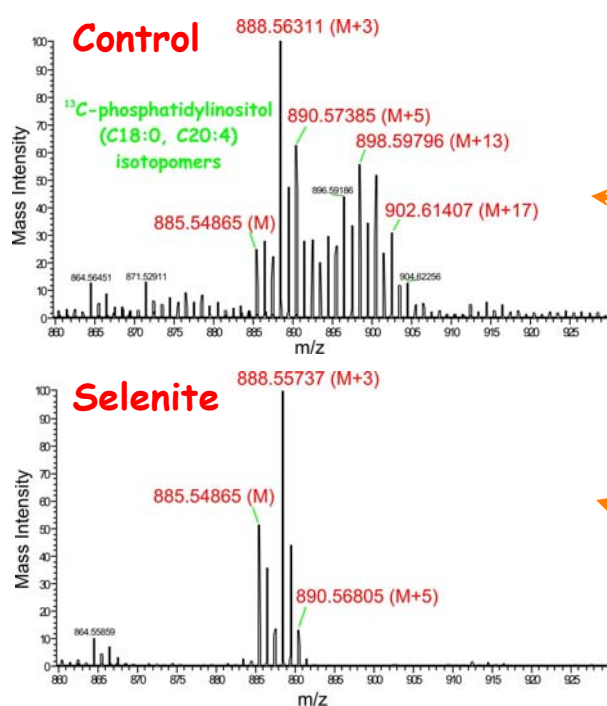


# Evaluation of cancer treatment

- Chemotherapy = side effects
  - Normal cells will be killed by cancer poisons
  - Drug companies test the drug in isolation
- Ovarian cancer cells can be “made” more sensitive to platinum-based therapies
  - Potential gene therapy
- How do multiple therapies work together?
  - Effects on cancer cells
  - Effects on normal cells



# Cancer drug development

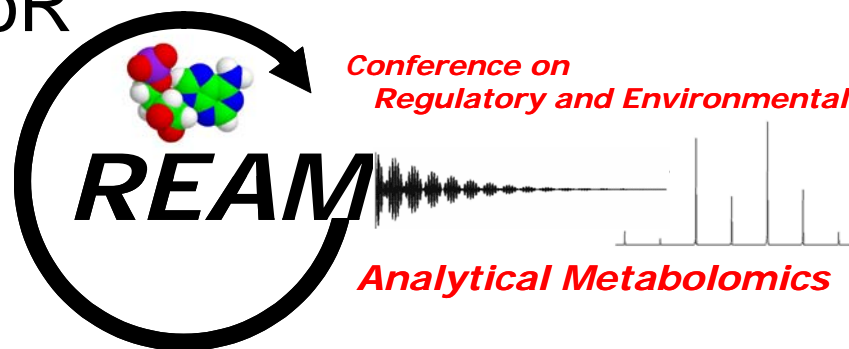


# Summary

- One reaction can be either destructive or supportive depending upon the pathway.
  - Sometimes differentiated in space (organelle, atmospheric level) or time
- Metabolism is a series of reactions that take apart and build molecules (metabolites).
- Cancer is primarily distinguished by metabolism
- The use of isotopomer metabolomics
  - Distinguishes normal cell from cancer cell activities
  - Identify and target cancer while supporting normal cell

# Acknowledgements

- Dr. Teresa Fan – Director
- Drs. Andrew Lane and Richard Higashi – Associate Directors
- Dr. Michael Bousamra II, MD – Cardiothoracic surgeon
- Ricky Woofter – Graduate Student in Chemistry
- Funded by NSF-EPSCoR grant EPS-0447479





# CD Summary

- Contains over 30 ready to use activities for your classroom
- List of frequently used web pages and addresses
- 6 folders
  - Air Pollution
  - Cancer
  - Genomics
  - Metabolomics
  - Periodic Table and Basic Chemistry
  - Toxicology

# Air Pollution Highlights

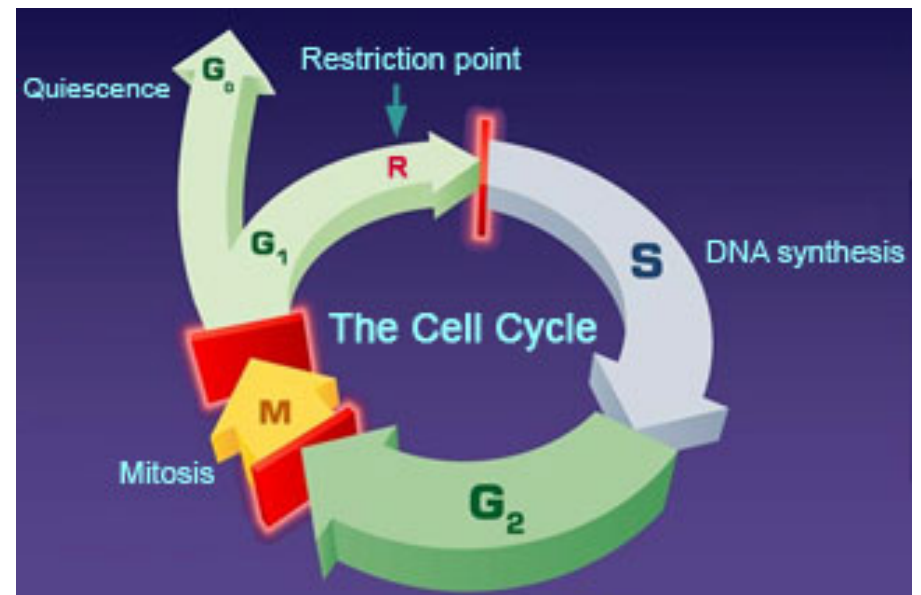
- Ground Ozone Lab
  - Uses basic materials and the Schoenbien Color Scale to determine ground level ozone

Colors on the scale below indicate the amount of ozone in the area



# Cancer Highlights

- Mitosis Run Amok and Cancer Brochure
  - Ideal for a KY portfolio piece in a Life Science class



# Genomics Overview

- Various Activities ranging from webquests to transcription/translation practice
- Lots of practice problems

# Metabolomics Highlights

- Articles discussing this up and coming science
- Can be used during CATS review time to help with reading comprehension skills

# Periodic Table and Basic Biochemistry Highlights

- Lots of review sheets covering information ranging from unit conversions to organic chemistry
- Half Life Labs
  - Can be used in various science classes
  - Cheap and easy to do!

# Toxicology Highlights

- Energy in the Food Web
  - Biomagnification
  - Energy Transfer
- Basic Toxicology Lab Stations
  - LD 50
  - Dose/response

# Web Pages

- This information is on the document labeled “Basic Summary of Items on CD”
  - Lists the main websites I have used in teaching my AP Biology and AP Environmental Science classes