Introducing Metabolomics: the Biochemical Balancing Act

Teresa Cassel
University of Louisville
Molly Woofter
Jefferson County Public Schools

Outline

- High School at U of L
- Metabolomics
- Selenium in the Environment
  - Chemical and biological importance
  - How metabolism effects eco-toxicity
- Selenium in the Human Diet
  - Medical significance
  - How metabolism effects cancer protection

Introducing CREAM

- Center for Regulatory and Environmental Analytical Metabolomics - 2005
- Sponsors:
  - National Science Foundation
  - State of Kentucky
  - University of Louisville

REAM
Conference on Regulatory and Environmental Analytical Metabolomics

- 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

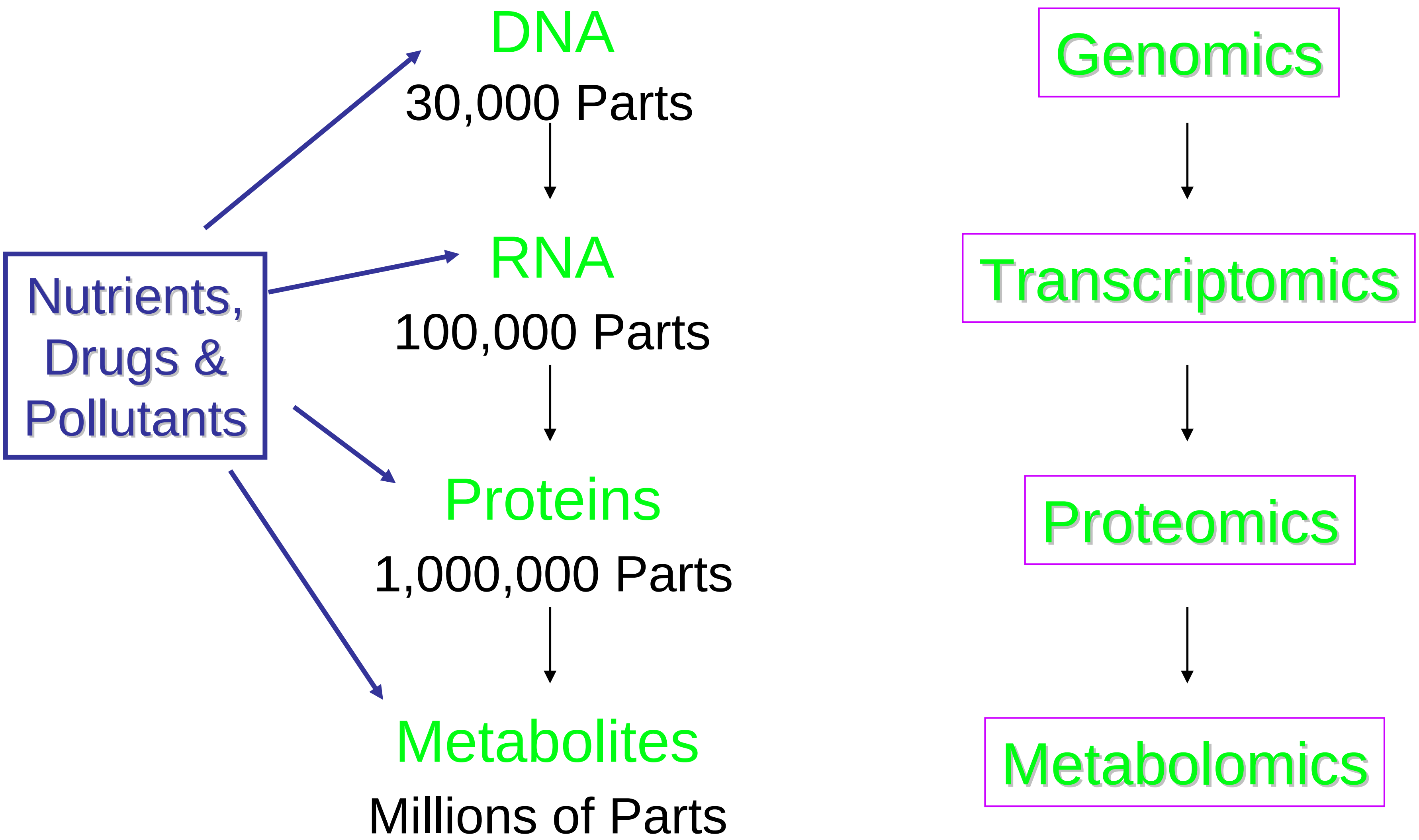
LTQ FT-MS

- 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

What is Metabolomics?

- *The systematic study of the unique chemical fingerprints that specific cellular processes leave behind* – Wikipedia
  - Basically, the collection of all metabolites.
  - Produces a ‘snapshot’ of cellular activity.
- Used to describe the metabolic changes caused by a biological perturbation (e.g. disease, poisoning, genetic modification)
  - Assess therapeutic intervention
  - Discover and develop new drugs

The New World Order of “Omics”



From Peter James (ed), Proteome Research, 2001.

What is Metabolomics good for?

- Assessing **bioavailability** (uptake and incorporation)
  - How and if an organism will respond to a pollutant, nutrient, or drug.
- Discovering toxic and/or therapeutic **mechanisms** (how it works)
  - by tracing metabolic pathways responsible for cell death or health
- Producing target **biomarkers**
  - Biochemicals we can measure that indicate health.

What is Selenium?

- Found in fossil fuels, shales, alkaline soils
  - High spatial variability in soils concentrations
  - High solubility in water
- A potent teratogenic poison
  - produces birth defects
  - Birds and fish are particularly sensitive
- An essential micronutrient for all species

Periodic Table of the Elements

Alkali metals	Actinide series	C Solid
Alkaline earth metals	Poor metals	Br Liquid
Transition metals	Nonmetals	H Gas
Lanthanide series	Noble gases	Tc Synthetic

New / Original	1 IA	2 IIA	3 IIIB	4 IVB	5 VB	6 VIB	7 VIIB	8	9 VIIIB	10	11 IB	12 IIB	13 IIIA	14 IVA	15 VA	16 VIA	17 VIIA	18 VIIIA
1	1 H Hydrogen 1.00794																	2 He Helium 4.002602
2	3 Li Lithium 6.941	4 Be Beryllium 9.012182											5 B Boron 10.811	6 C Carbon 12.0107	7 N Nitrogen 14.00674	8 O Oxygen 15.9994	9 F Fluorine 18.9984032	10 Ne Neon 20.1797
3	11 Na Sodium 22.989770	12 Mg Magnesium 24.3050											13 Al Aluminum 26.981538	14 Si Silicon 28.0855	15 P Phosphorus 30.973761	16 S Sulfur 32.066	17 Cl Chlorine 35.453	18 Ar Argon 39.948
4	19 K Potassium 39.0983	20 Ca Calcium 40.078	21 Sc Scandium 44.955910	22 Ti Titanium 47.867	23 V Vanadium 50.9415	24 Cr Chromium 51.9961	25 Mn Manganese 54.938049	26 Fe Iron 55.8457	27 Co Cobalt 58.933200	28 Ni Nickel 58.6934	29 Cu Copper 63.546	30 Zn Zinc 65.409	31 Ga Gallium 69.723	32 Ge Germanium 72.64	33 As Arsenic 74.92160	34 Se Selenium 78.96	35 Br Bromine 79.904	36 Kr Krypton 83.798
5	37 Rb Rubidium 85.4678	38 Sr Strontium 87.62	39 Y Yttrium 88.90585	40 Zr Zirconium 91.224	41 Nb Niobium 92.90638	42 Mo Molybdenum 95.94	43 Tc Technetium (98)	44 Ru Ruthenium 101.07	45 Rh Rhodium 102.90550	46 Pd Palladium 106.42	47 Ag Silver 107.8682	48 Cd Cadmium 112.411	49 In Indium 114.818	50 Sn Tin 118.710	51 Sb Antimony 121.760	52 Te Tellurium 127.60	53 I Iodine 126.90447	54 Xe Xenon 131.293
6	55 Cs Cesium 132.90545	56 Ba Barium 137.327	57 to 71	72 Hf Hafnium 178.49	73 Ta Tantalum 180.9479	74 W Tungsten 183.84	75 Re Rhenium 186.207	76 Os Osmium 190.23	77 Ir Iridium 192.217	78 Pt Platinum 195.078	79 Au Gold 196.96655	80 Hg Mercury 200.59	81 Tl Thallium 204.3833	82 Pb Lead 207.2	83 Bi Bismuth 208.98038	84 Po Polonium (209)	85 At Astatine (210)	86 Rn Radon (222)
7	87 Fr Francium (223)	88 Ra Radium (226)	89 to 103	104 Rf Rutherfordium (261)	105 Db Dubnium (262)	106 Sg Seaborgium (266)	107 Bh Bohrium (264)	108 Hs Hassium (269)	109 Mt Meitnerium (268)	110 Ds Darmstadtium (271)	111 Rg Roentgenium (272)	112 Uub Ununbium (285)	113 Uut Ununtrium (284)	114 Uuq Ununquadium (289)	115 Uup Ununpentium (288)	116 Uuh Ununhexium (292)	117 Uus Ununseptium	118 Uuo Ununoctium

Atomic masses in parentheses are those of the most stable or common isotope.

57 La Lanthanum 138.9055	58 Ce Cerium 140.116	59 Pr Praseodymium 140.90765	60 Nd Neodymium 144.24	61 Pm Promethium (145)	62 Sm Samarium 150.36	63 Eu Europium 151.964	64 Gd Gadolinium 157.25	65 Tb Terbium 158.92534	66 Dy Dysprosium 162.500	67 Ho Holmium 164.93032	68 Er Erbium 167.259	69 Tm Thulium 168.93421	70 Yb Ytterbium 173.04	71 Lu Lutetium 174.967
89 Ac Actinium (227)	90 Th Thorium 232.0381	91 Pa Protactinium 231.03588	92 U Uranium 238.02891	93 Np Neptunium (237)	94 Pu Plutonium (244)	95 Am Americium (243)	96 Cm Curium (247)	97 Bk Berkelium (247)	98 Cf Californium (251)	99 Es Einsteinium (252)	100 Fm Fermium (257)	101 Md Mendelevium (258)	102 No Nobelium (259)	103 Lr Lawrencium (262)

Note: The subgroup numbers 1-18 were adopted in 1984 by the International Union of Pure and Applied Chemistry. The names of elements 112-118 are the Latin equivalents of those numbers.

Chemical Characteristics of Se

- Similar to Sulfur
  - Found in place of S in amino acids
- Five oxidation levels:
  - Insoluble
    - Elemental Se
  - Water Soluble
    - Selenite Se^{4+} or SeO_3^-
    - Selenate Se^{6+} or SeO_4^{2-}
  - Selenols: powerful reducing agents (MeSeH)
  - Proteinaceous/Organic Se
- Six non-radioactive, natural isotopes

16 $^3P_2$

S

Sulfur

32.066

$[Ne]3s^2 3p^4$

34 $^3P_2$

Se

Selenium

78.96

$[Ar]3d^{10} 3s^2 3p^4$

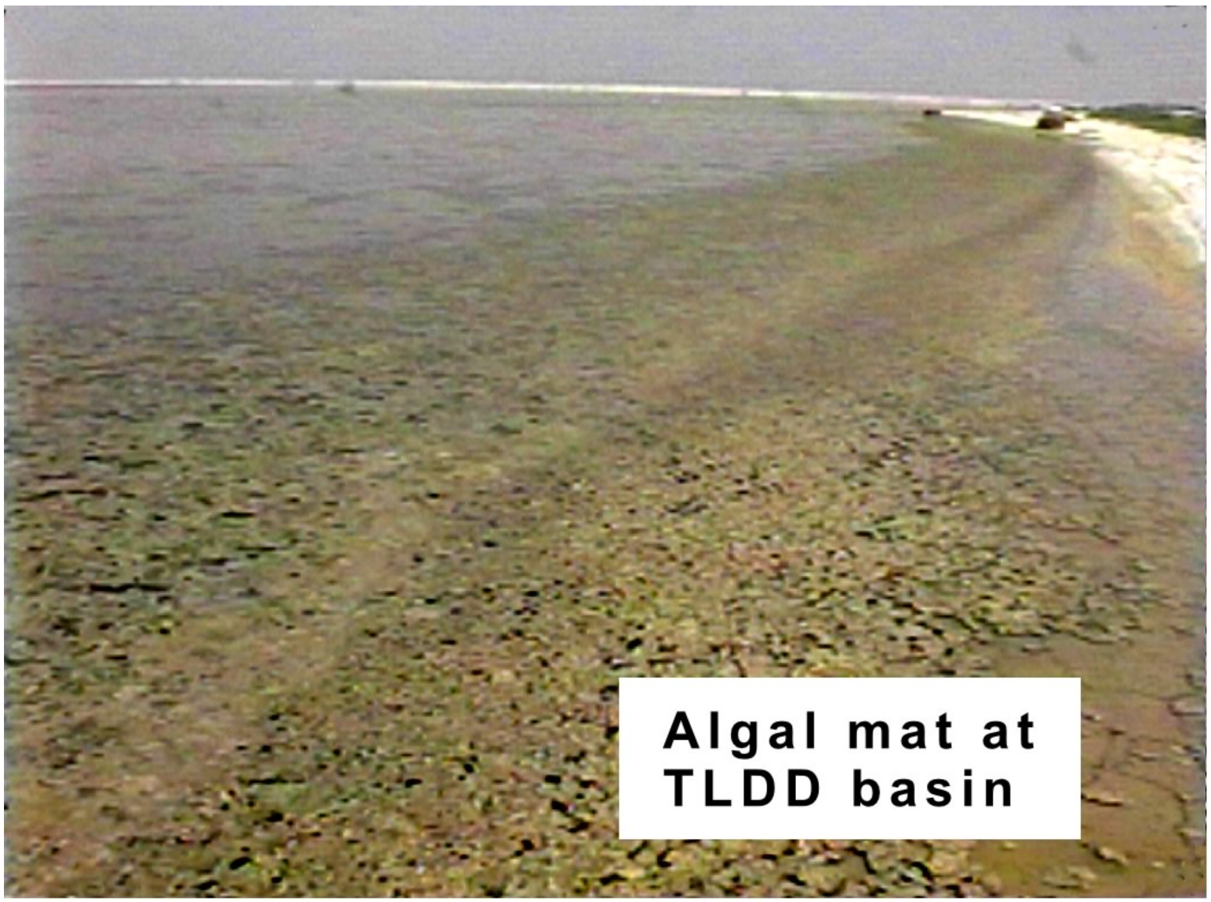
Human impact on Se distribution

- Irrigated agriculture leaches soluble forms from soils. Drainage waters can be highly enriched.
  - Kesterson Wildlife Refuge
- Surface mining also leaches soluble Se
- Power plant discharges contain Se
  - Tennessee Valley Authority; Belews Lake

Kennecott Copper Mine pit in Utah.

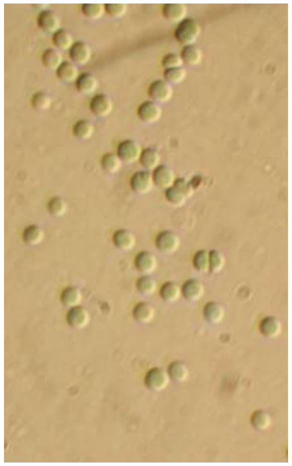
Photographer: Bruce Molnia US Geological Survey

Agricultural drainage pond



Uptake and Incorporation of Se

- The metabolism of an organism determines whether and how:
  - Se is transferred from the surroundings into the organism
  - Se is detoxified and depurated back to the surroundings
  - What form the Se is in when the organism is predated or decomposed
- Selenium must be "eaten" or actively transferred into an organism
  - Ingestion is the main route of exposure for selenium



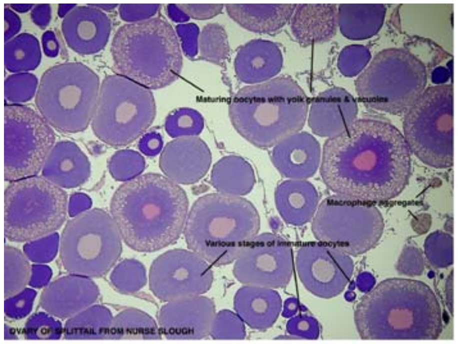
Algae

Cattails

- Some halophyte algae detoxify by sending to the air
  - *Volatilization* – metabolic methylation (adding CH_3)
  - Vascular plants metabolism produces less methylation
- Incorporation into vascular plants
  - incorporated into plant proteins
  - plant decomposition can make toxic forms more available than in plant-free environments

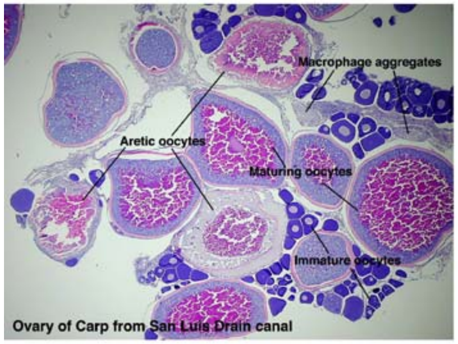
Bioaccumulation of Se

- From the Primary Producers Se is passed up the food web
  - High water [Se] ≠ High tissue [Se]
  - Bioaccumulation of total Se and proteinaceous Se
- Toxic form is identified as that bound to protein
  - High tissue [Se] ≠ High toxicity
  - Sensitive species have metabolism that tends to incorporate more Se into the proteins.
  - Biomarker example



Splittail ovary

Total Se (µg/g)	%Protein-bound Se
50.2	3.2



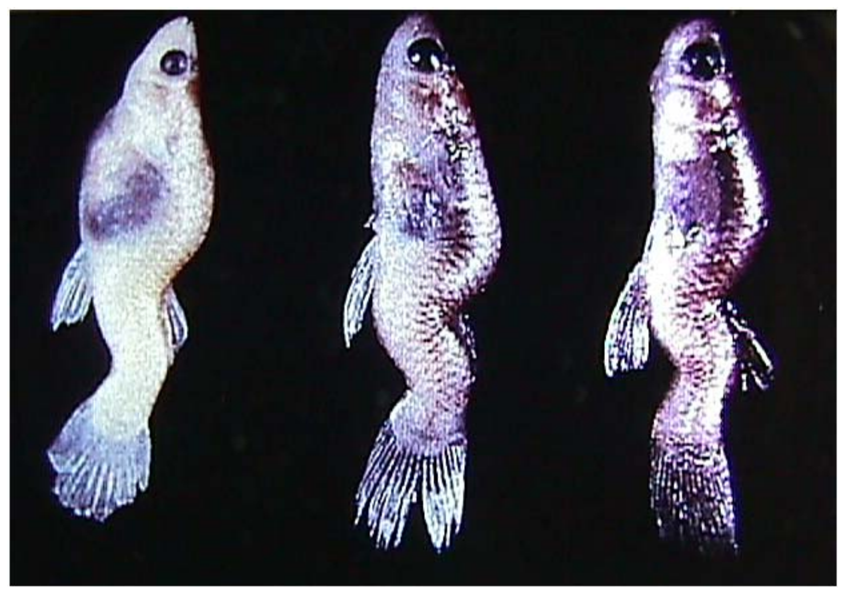
Carp ovary

Total Se (µg/g)	%Protein-bound Se
10.6	47.4

Toxic Mechanism of Se

- Toxic currency – selenomethionine concentration is correlated to teratogenesis
  - measuring tissue concentrations of Se does not predict teratogenesis
  - Selenomethionine concentration can indicate health (biomarker)
  - Looking for Se metabolite that is directly causing deformities

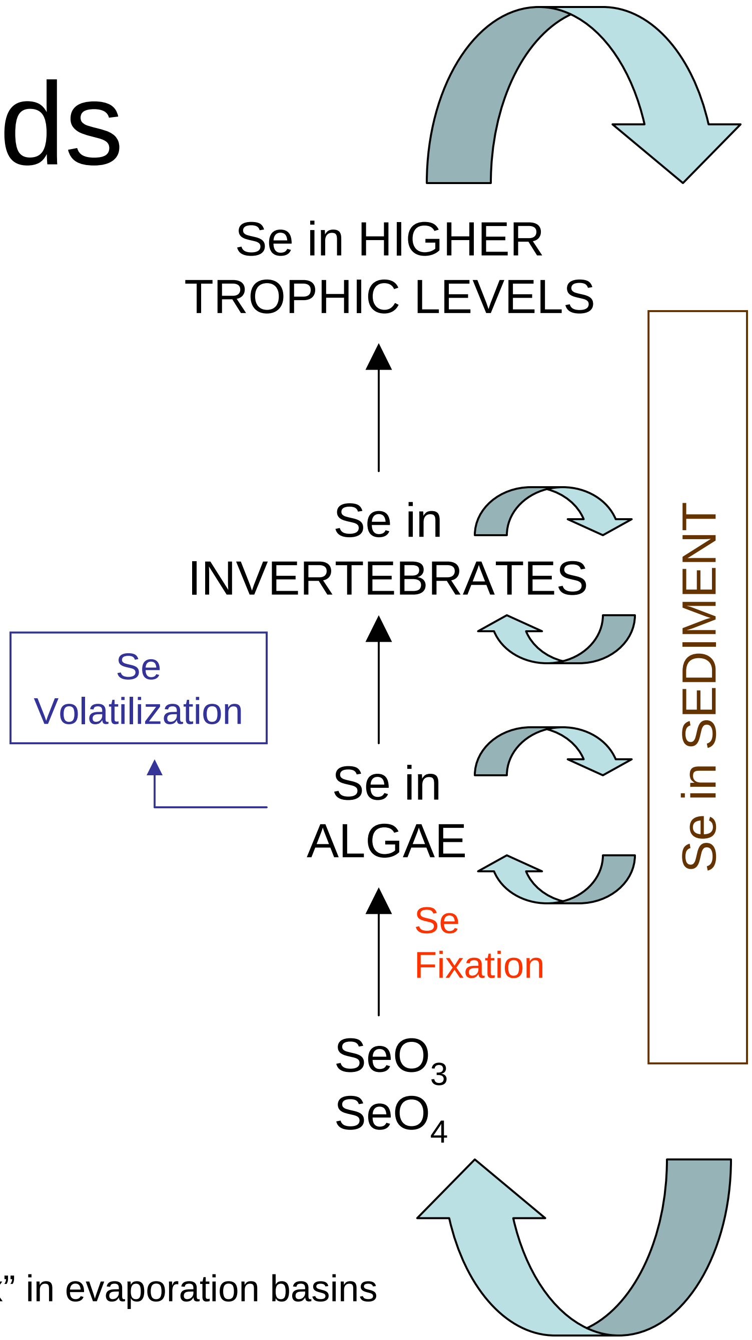
Avocet From Kesterson, c/o Dr. Joseph Skorupa, US F&W Service



From Belews Lake, c/o Dr. Allen Knight, UC Davis

Se Cycling in ponds

- The ultimate fate of all living organisms is:
  - Consumption or
  - Decomposition
- Se is fixed by primary producers
  - Some Se is removed by volatilization
- Se is bioaccumulated in next trophic level (biomagnification)
- Death and decomposition release organic Se to sediment organisms



Biogeochemical “Reflux” in evaporation basins

How Se affects Human Health

- Recommended dietary allowance is 55 µg/d
  - Based on intake required to optimize plasma glutathione peroxidase activity, a metabolic function.
  - Deficiency can lead to compromised immune function; muscle wasting in livestock
- Selenocysteine, the 21st amino acid, is a powerful antioxidant acting through Selenoproteins
  - Essential for maintaining redox balance (prevent cellular damage from free radicals natural by-products of oxygen metabolism that may contribute to the development of chronic diseases such as cancer and heart disease)
  - Other selenoproteins help regulate thyroid function and play a role in the immune system
- Mechanisms for anti-cancer activity are being investigated using metabolomics at CREAM
  - Treating cancer cells with various forms of Se
  - Dietary Se supplementation in mice with cancer tumors

Observational Studies of Se Effects on Overall Cancer Risk

Investigator	Year	Population	Size
Willett et al.	1983	USA	111
Salonen et al.	1984	Finland	128
Fex et al.	1987	Sweden	35
Kok et al.	1987	Netherlands	69
Knekt et al.	1990	Finland	1096
Virtamo et al.	1987	Finland	109
Ringstad et al.	1988	Norway	60
Peleg et al.	1985	USA	154
Coates et al.	1988	USA	154
Avanzini et al.	1995	Italy	95
Garland et al.	1995	USA	934

Meuillet et al. *J. Cellular Biochemistry* (**2003**)91:3 pp. 443-458

SELECT trial

- The Selenium and Vitamin E Cancer Prevention Trial
- Determine if Se and Vitamin E can help prevent Prostate cancer
- Funded by National Cancer Institute
- Over 35,000 men from over 400 sites in US, Puerto Rico, and Canada

http://www.crab.org/select/about.asp

Foods rich in Se

Summary

- Ingestion is critical in Se toxicity and nutrition.
  - Form (e.g. oxidation state) determines absorption.
- Metabolism determines effects of Se on the organism or tissue.
  - Ovarian tissue can be effected when other systems are not – teratogenesis.
  - Cancer tumors have different metabolism from normal cells and can be effected by plasma Se concentrations that may not effect healthy tissue.

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