

## Xenobiotic compounds and physiology

- (from the Greek *xenos* "stranger" and *biotic* "related to living beings")
- **Absorption**
- **Distribution**
- **Metabolism**
- **Excretion**

## Each of these steps poses problems

- Metabolism - metabolites
- Excretion - transformation products
- Bioconcentration – food chain events

## Compounds of Concern

- Pharmaceuticals
- Disinfectants
- Pesticides
- Flame retardants
- De-icing fluids
- Newly manufactured particles (electronics)

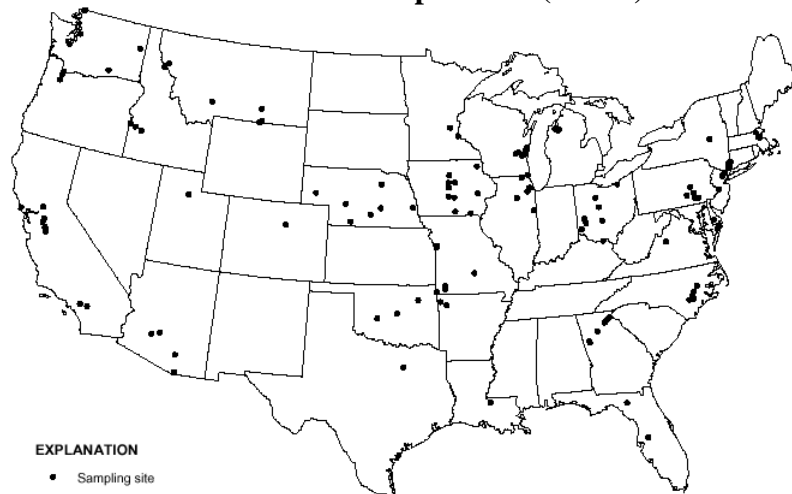
## Clean Water Act on 1972

- Did not consider these in detail or at all
- Conserved with metals and nutrients more than anything else in the design of treatment and mitigation facilities
- Agricultural pesticides part but little awareness

## The Wake Up Call

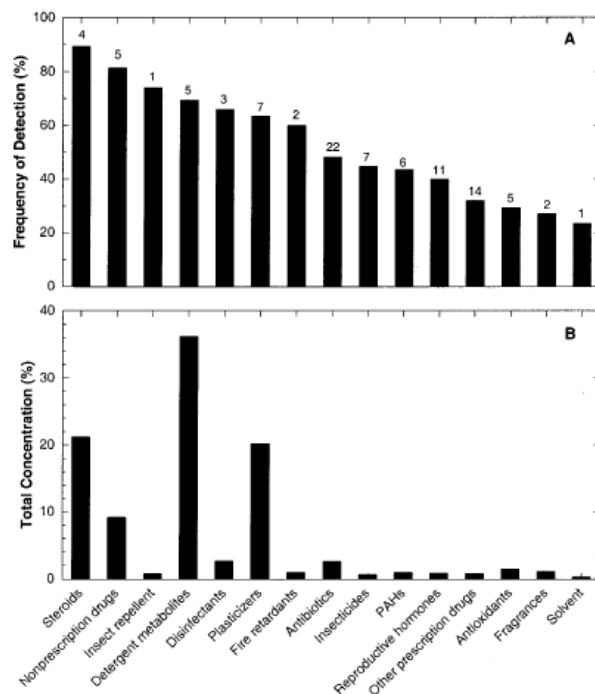
- Study of rivers across the nation
- Identified the suite of non-natural or unexpected (Xenobiotics) in the water ways.

### Water-Quality Data for Pharmaceuticals, Hormones, and Other Organic Wastewater Contaminants in US Streams, 1999 – 2000. Kolpin et al. (USGS)



## Kolpin et al. (USGS)

- ~ 50% of samples collected across 30 different states had at least one antibiotic or a breakdown product of an antibiotic
- Some samples from the 139 streams and rivers contained 4 or 5 of the 22 antibiotics assayed



Frequency and number of compounds

Percent total measured concentrations

## EPA response

- Convened scientists to look at emerging compounds, how to regulate and deal with them.

### **WHITE PAPER**

### **AQUATIC LIFE CRITERIA FOR CONTAMINANTS OF EMERGING CONCERN – on web**

## Who is in Charge – USA?

- EPA – pesticides, herbicides, waste discharge, emissions.
- FDA – drugs for animals and humans to protect user, target and environment.

## The Concerns

- Are they talking to one another
- What about cumulative risks?

- **Persistent organic pollutants** (POPs) such as polybrominated diphenyl ethers (PBDEs; used in flame retardants, furniture foam, plastics, etc.) and other global organic contaminants such as perfluorinated organic acids;

- **Pharmaceuticals and personal care products** (PPCPs), including a wide suite of human prescribed drugs (e.g., antidepressants, blood pressure), over-the-counter medications (e.g., ibuprofen), bactericides (e.g., triclosan), sunscreens, synthetic musks;

- **Veterinary medicines** such as antimicrobials, antibiotics, anti-fungals, growth promoters and hormones;•

### **Endocrine-disrupting chemicals (EDCs),**

including synthetic estrogens (e.g., 17 $\alpha$ -ethynylestradiol, which also is a PCPP) and androgens (e.g., trenbolone, a veterinary drug), naturally occurring estrogens (e.g., 17 $\beta$ -estradiol, testosterone), as well as many others (e.g., organochlorine pesticides, alkylphenols) capable of modulating normal hormonal functions and steroidal synthesis in aquatic organisms;

- **Nanomaterials** such as carbon nanotubes or nano-scale particulate titanium dioxide, of which little is known about either their environmental fate or effects.

## Pharmaceuticals

- Use is increasing – drug dependent populations
- Human populations - \$ and access - China
- Agricultural industry use
  - Meat production Land and Aquaculture - FDA
  - Fruit and other plant production systems - EPA

## Orchard use for blight control

- In the United States, streptomycin is registered for use on twelve fruit, vegetable, and ornamental plant species; oxytetracycline is registered for use on four fruit crops.
- Antibiotic use on crops and ornamental plants in the U.S. is regulated by the Environmental Protection Agency
- Estimated annual use up to 65,227 kg – data not easy to find

## Occurrence and Fate of Pharmaceuticals in Aquatic Systems

- **Recent plethora of studies in the US and elsewhere revealed a number of compounds in waste waters and waterways**
- **What are the sources?**
  - **Human and Animal**



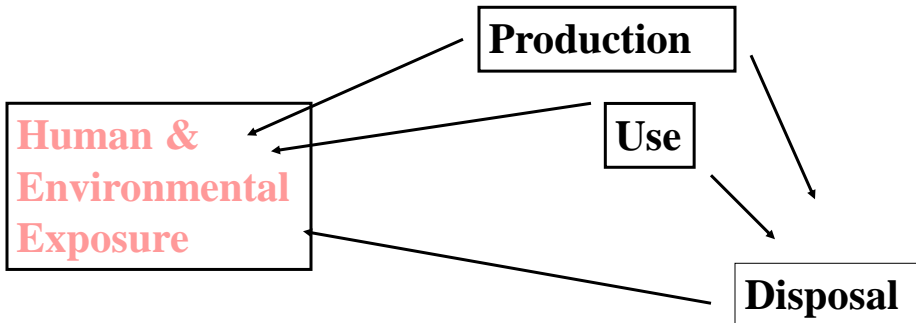
## **Types of Compounds Human and Vet Medicine**

- **Analgesics**
- **Diuretics**
- **Anti-asmatics**
- **Psychleptics**
- **Cancer drugs**
- **Endocrine disruptors**

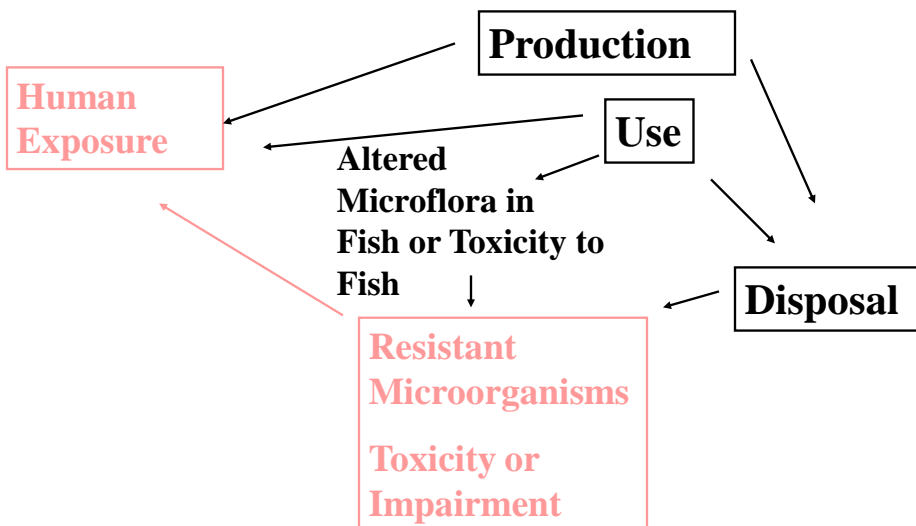
## **Studies in the Literature**

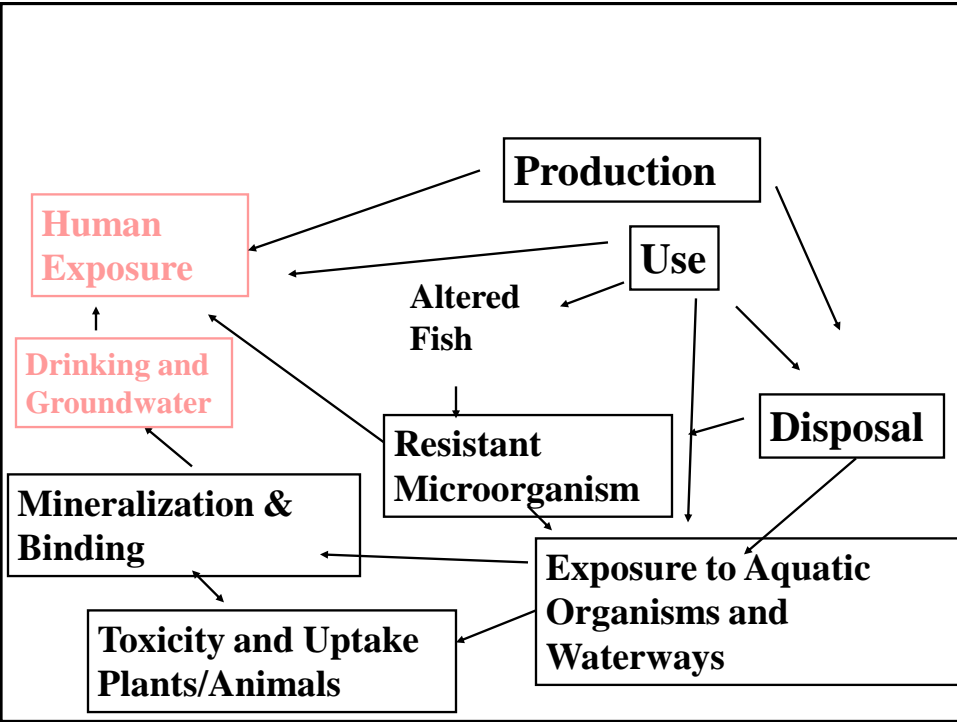
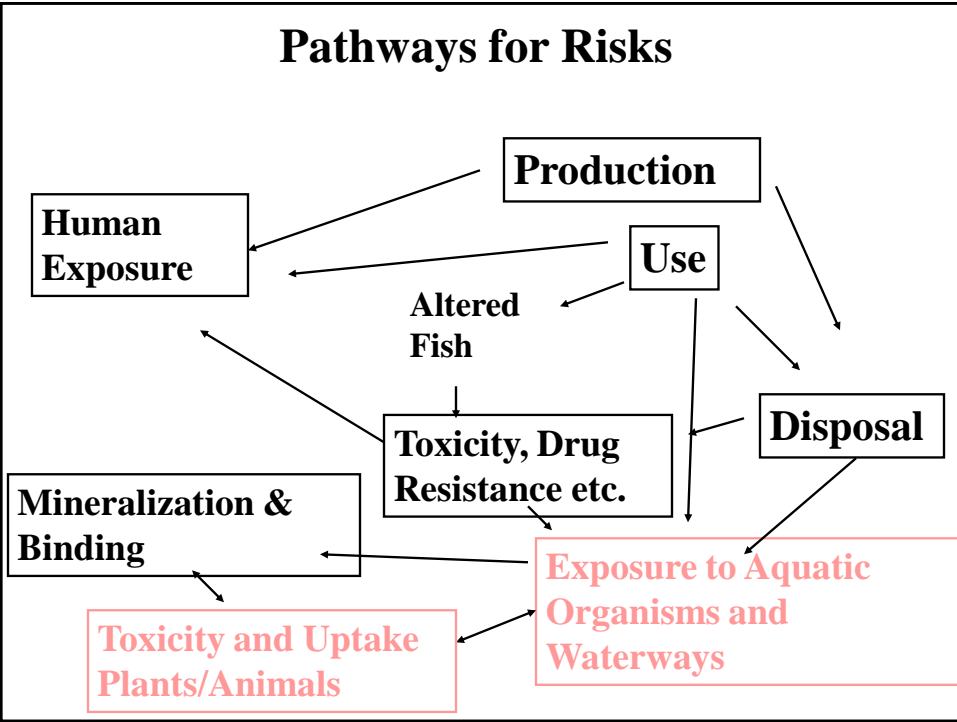
- Localized monitoring
- Meta analyses
- Streams, lakes, rivers, some marine but difficult

## Environmental and Human Risks Pharmaceuticals



## Pathways for Risks





## **Antimicrobial Resistance & Acute and Chronic Toxicity**

- **Resistance = Indirect and more focused on humans from zoonotic bacteria/microbial communities**
- **Toxicity = Direct effect on the environment and humans through ingestion or exposure**

## **Antimicrobial Resistance**

- **Linked to frequent and improper use of antibiotics for humans**
- **Global public health problem**
- **Concern of Center for Disease Control (CDC)**
- **CDC goal to keep *Salmonella*, *Shigella* and the strains of *E. coli* and other enterococci from getting out of control.**

## **Risks and Decision Analysis**

- **Precautionary Principle – Europe**
- **Risk Assessment Approach – USA**  
– **FDA and EPA**

## **Effects of Compounds**

- Similar to bioenergetic models, the compounds can accumulate if the intake exceeds the metabolism and excretion

## Models of Distribution

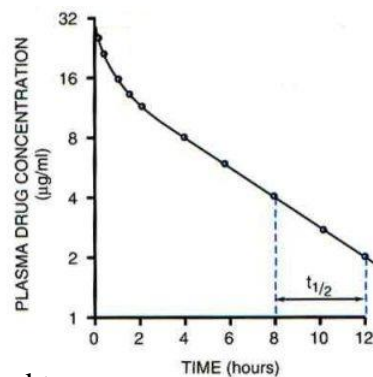
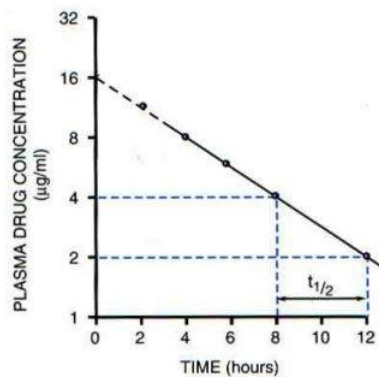
- The distribution can be one phase or several phases.
- As a consequence, the elimination will be related to the distributions and properties of the compounds.

## Compartmental model

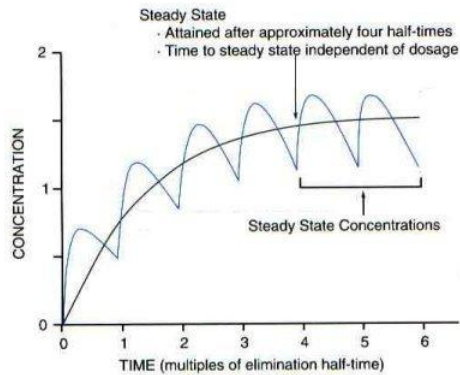
- Single compartment – metabolism and depletion can keep pace and remain consistent over time
- Two compartment or more is one that has different rates for different dosages, and more complex elimination and depletion

## Methods of Entry

- Waterborne exposure – uptake across gills and skin in freshwater
- Feeding or ingestion – uptake from the GI
- Uptake from injection (drugs- non normal routes of exposure)



Elimination Models, one and two phase



## Considerations

- Absorption - Chemical speciation and membrane permeability
- Physicochemical properties
- Lipophilicity – or hydrophobicity
- Stability or Degradation, inside and in environment



## Parameters that Affect Clearance

- **Filtration**
- **Secretion**
- **Re-absorption**

## Traditional Calculations

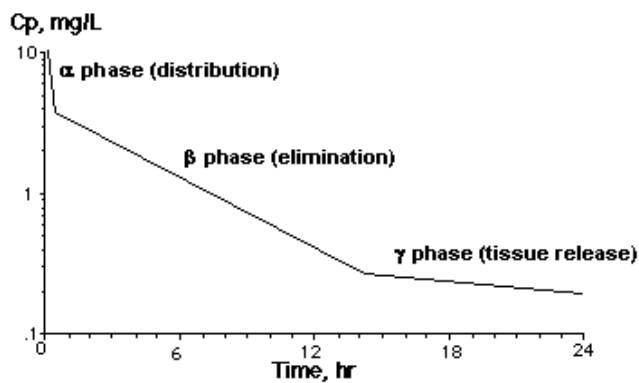
- **Volume of Distribution  $V_d$  ( mL/kg) =**
- **Loading dose to get the target needed in plasma**

$$\frac{\text{Total amt drug in body}}{\text{Drug blood plasma concentration}}$$

# Bio-availability

- Fraction absorbed unchanged that reaches target relative to dose administered
- If IV applied, it is all entered into system, but if oral the absorption is key feature
- Integrated area under the curve to obtain entire distribution

Three Compartment Models with tissue storage  
Area under the curve is plotted or modeled



## Exposure & Dosing

- MIC – pathogen or some effect
- Concentration independent
- Concentration dependent

## Detoxification Pathways

- The most common reaction catalysed by cytochrome P450 is a monooxygenase reaction, e.g. insertion of one atom of oxygen into an organic substrate (RH) while the other oxygen atom is reduced to water:
- $\text{RH} + \text{O}_2 + 2\text{H}^+ + 2\text{e}^- \rightarrow \text{ROH} + \text{H}_2\text{O}$

## Cytochrome p450

- CYP enzymes have been identified from all lineages of life, including mammals, birds, fish, insects, worms, sea squirts, sea urchins, plants, fungi, slime molds, bacteria and archaea.

- Cytochrome P450 naming comes from pigmented ('chrome') cellular ('cyto') proteins. The pigment absorbs at 450 nm.
- This happens when the heme iron is reduced (often with sodium dithionite) and complexed to carbon monoxide.

## Drug Metabolism

- CYPs are the major enzymes involved in drug metabolism, accounting for ~75% of the total metabolism.
- Cytochrome P450 is the most important element of oxidative metabolism (also known as phase I metabolism). (Metabolism in this context is the chemical modification or degradation of drugs.)

## Drug Interactions

- Some compounds reduce or bind this system and can cause changes in bio-availability of drugs
- Many drugs may increase or decrease the activity of various CYP isozymes in a phenomenon known as enzyme induction and inhibition.

## BioAvailability

- For example, bioactive compounds found in grapefruit juice and some other fruit juices, including bergamottin, dihydroxybergamottin, and paradisin-A, have been found to inhibit CYP3A4-mediated metabolism of certain medications, leading to increased bioavailability and thus the strong possibility of overdosing.

## Example from my own research

- Erythromycin dosages
  - Low dosages are eliminated and single compartment
  - Higher dosages, the elimination is fixed rate and the substance accumulates in certain organs and is two and three compartmental model.

## Different affinities and depletions

- Kidney, Spleen, Liver – perfused tissues
- White and Red muscle
- Blood plasma

## Endocrine Disrupting Chemicals EDC - and other Xenobiotics

- EDCs have the potential to modulate or disrupt the synthesis, secretion, transport, binding, action, or elimination of endogenous hormones in the body and consequently to affect homeostasis, development, reproduction, and behavior of organisms

- Steroid substances are a group of phylogenetically conserved hormones that act through a common mechanism; i.e., they bind to cytoplasmic and nuclear receptors and the ligand–receptor complex subsequently activates transcription of steroid-responsive genes.

## Contraceptives and estrogenic hormones

- 17 $\alpha$ -ethinylestradiol (EE2), a major constituent of common contraceptive pills
- Estrogenic hormones, estrone (E1) and 17 $\beta$ -estradiol (E2)



## Xeno estrogens

- Bisphenol A, BPA, is an organic compound with two phenol functional groups used to make polycarbonate plastic and epoxy resins, along with other applications. In type 3 and type 7 plastics, type 6 is supposed to be free.
- Bisphenol A is also a precursor to the flame retardant tetrabromobisphenol A, and was formerly used as a fungicide. Bisphenol A is a preferred color developer in carbonless copy paper and thermal paper, with the most common public exposure coming from some thermal point of sale receipt paper. BPA-based products are also used in foundry castings and for lining water pipes.
- The current U.S. human exposure limit set by the EPA is 50  $\mu\text{g}/\text{kg}/\text{day}$

- octylphenol (OP) The main use of octylphenol (80% of the total quantity) is in the production of Para-Tert-Octylphenol (PTOP) based resins, which are used as tackifiers in tire manufacture. Additionally, these resins are also used for metals to rubber bonding applications in the technical rubber goods

## Detoxification Process Vertebrates

- Liver – eliminate exogenous and endogenous toxins
- Phase I and Phase II systems
- Phase I reactions involve blood filtration, bile excretion, and interaction of enzymatic processes acting on the toxin.
- Bile excretion is efficient regarding detox. Process
- Toxin is escorted with fibers from intestines

## Phase I detoxification

- Produces free radicals, as toxins are transformed – for each molecule of toxin metabolized by phase I, one molecule of free radical is generated.
- Without adequate free radical defenses, every time the liver neutralizes toxin, it is damaged by free radicals produced.