More Applications of Pharmacology & Toxicology

Week 3

Environmental Toxicology

- Study of the hazardous effects that poisons in the environment have on human health
- 4 billion tons of waste are produced annually from mining, agriculture, industry, municipal sewage
- Daily 2.5 kg (4 lb) domestic solid waste per capita
  - Only 10% of this waste is disposed of in environmentally safe manner
- 5 million natural & man-made chemicals; 80,000 synthetic chemicals
- WHO epidemiological data: 90-95% cancers are "environmentally related"
- Environmental toxicology can be divided into two subcategories:
- **Environmental health toxicology**: Study of the adverse effects of environmental chemicals on human health.
- **Ecotoxicology** which focuses upon the effects of environmental contaminants upon ecosystems & constituents thereof (fish, wildlife, etc).

Toxicant Distribution

- Lymph: capillaries, nodes, special lymphoid structures (tonsils, spleen, thymus), peripheral lymphocytes
- Cardiovascular: heart, arterial vessels, venous vessels, capillaries
- Nervous system: brain/CNS, PNS
- Organs handling xenobiotic biotransformation, excretion: liver, kidney

Target Organ Toxicity Examples

Organ	Toxicant	Mechanism	Toxicity
heart	fluorocarbons (Freon)	sensitizes heart to epinephrine?	decreased heart rate, contractility, and conduction
	carbon monoxide (CO)	interferes with energy metabolism	myocardial infarction. increase or decrease in heart rate
	cobalt (Co)	competes with Ca^{2+}	heart failure
testis	lead (Pb)	mutations in sperm?	decreased male fertility, increased spontaneous abortions
	carbon disulfide (CS_2)	CNS effect on ejaculation	reduced sperm counts
ovary/ uterus	solder fumes	unknown	increased spontaneous abortions
	polycyclic aromatic hydrocarbons (PAHs)	unknown	damaged oocytes
eye	Busulfan (chemotherapeutic agent)	alters mitosis in lens cells?	formation of cataracts
	methanol (CH_3OH)	produces optic atrophy	permanent visual impairment, blindness

Environmental Toxicants

- Pesticides
  - insecticides
  - herbicides
  - fungicides
  - rodenticides
- Plastics
- Metals

  As, Cr, Be, Hg, Pb, Ni, Cd
- Organic Solvents

  methanol, benzene + alkylbenzenes (toluene, xylenes), carbon disulfide, ethanol,
  ink/paint solvents (methyl ethyl ketone)

Pesticides: Insecticides

- most are neurotoxicants to insects sometimes to humans
- entry routes include skin, lung, oral (GI tract)

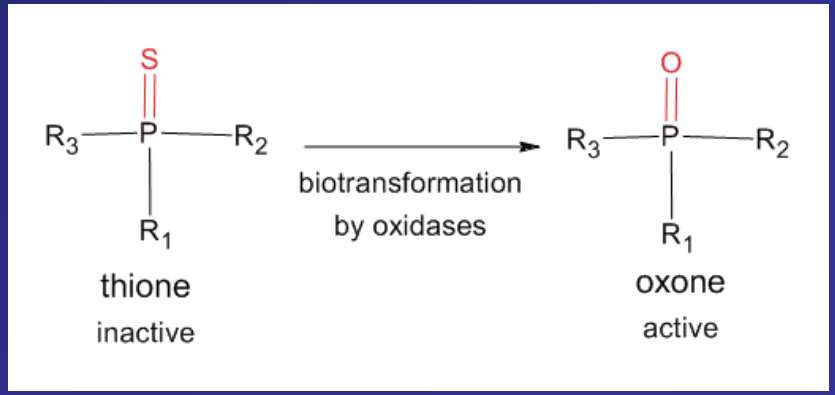
Organophosphates

- parathion, diazinon, malathion: specific targets for arthropod acetylcholinesterase

  California Mediterranean fruit fly malathion spraying

Carbamates

- aldicarb, carbaryl, propoxur
- also anti-cholinesterases



Pesticides: Insecticides

Acute toxicity

- OPs: DUMBELS=diarrhea, urination, miosis (pinpoint pupils), bronchospasm, emesis (vomiting), lacrimation (tearing), salivation
- recovery periods vary from a few days to weeks for mild/moderate exposure to a few months for severe
- death can occur usually within 24 h from untreated exposure
- Carbamate exposures resolve more quickly because cholinesterase inhibition is reversible

Chronic toxicity

- Organophosphate-induced delayed neuropathy (OPDIN) for select organophosphate: predominantly motor paralysis

Pesticides: Insecticides

Organochlorines

- dichlorodiphenyltrichloroethane (DDT)
- neurotoxic: either stimulatory or inhibitory: interfere with K^+ or Ca^{2+}; poorly absorbed in skin
- very stable (long-lived) chemicals: reason for ban
- accumulates in lipid stores (biological magnification)
- phase I: dechlorinations, demethylation
- phase II: glutathione
- acute toxicity: CNS—headaches, dizziness, tremors, convulsions
- chronic toxicity: memorly loss, personality changes, low sperm count

Pesticides: Herbicides

- mechanism of toxicity is by interfering with hormonal systems regulating growth or promoting dehydration/dessication
- weak *acute* toxicity to humans

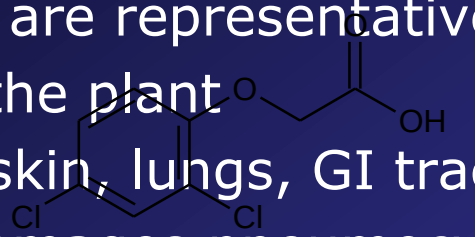
  chronic exposure?
- entry routes include skin, lung, oral (GI tract)

- Bipyridyls
- Chlorophenoxy compounds
- DNP

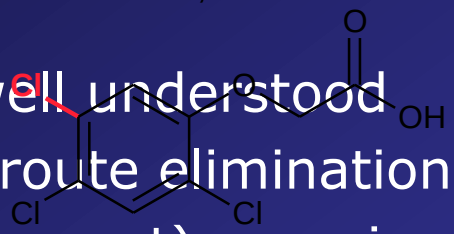
Pesticides: Herbicides

Bipyridyls

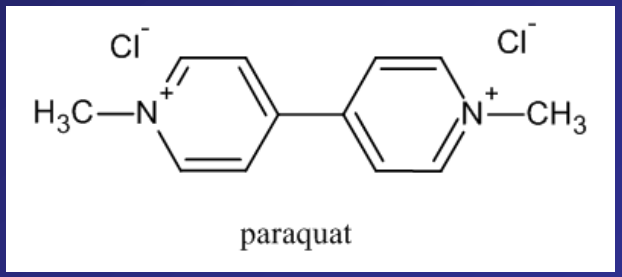
- diquat, paraquat are representative
- act to desiccate the plant
- routes of entry: skin, lungs, GI tract
- acute toxicity: damages pneumocytes to decrease O_2/CO_2 gas exchange
- metabolism not well understood
- urinary and fecal route elimination
- acute toxicity (paraquat): anoxia, coma, organ (liver, kidney, lung) damage; ingestion of concentrated product causes death



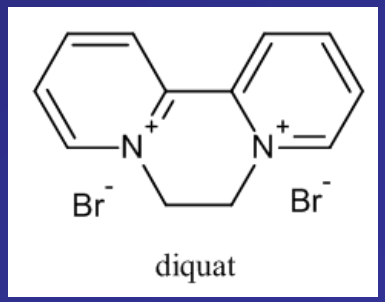
2,4-D



2,4,5-T



paraquat

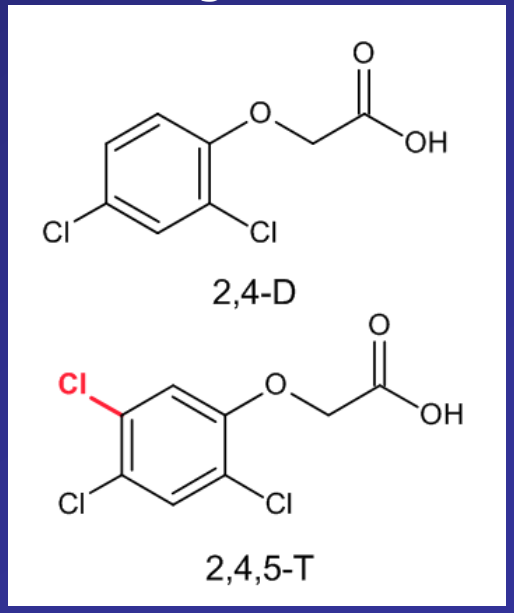


diquat

Pesticides: Herbicides

Chlorophenoxy Compounds

- 2,4-dichlorophenoxyacetic acid (2,4-D), 2,4,5-trichlorophenoxyacetic acid (2,4,5-T)
- Agent Orange is a 50:50 mix of 2,4-D & 2,4,5-T
- Herbicidal action through uncontrolled plant growth
- these compounds alone are weakly toxic to humans, but the presence of TCDD* in trace amounts in manufacture gives it (controversial) high toxicity
- routes of entry: skin, lungs, GI tract
- metabolism and excretion data wanting
- acute toxicity: sweating, oliguria, peripheral neuropathies, muscle weakness, dizziness, headache, vomiting, fatigue
- TD_{50} ~ 50-60 mg/kg; LD_{50} ~ 300 mg/kg

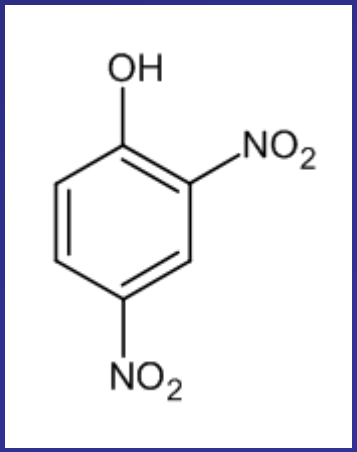


* TCDD is addressed in later slide

Pesticides: Herbicides

2,4-Dinitrophenol

- DNP absorbed by skin, lungs, GI tract
- toxicity is by inhibition of ATP synthesis: symptoms include tachypnea, tachycardia, sweating, coma, development of cataracts
- carcinogenic: feature of aromatic nitro and amine compounds
- It had been used as an anti-obesity drug in the 1930s but no data on toxicokinetics is available at all
- US EPA now considers it environmental contaminant



Pesticides: Fungicides

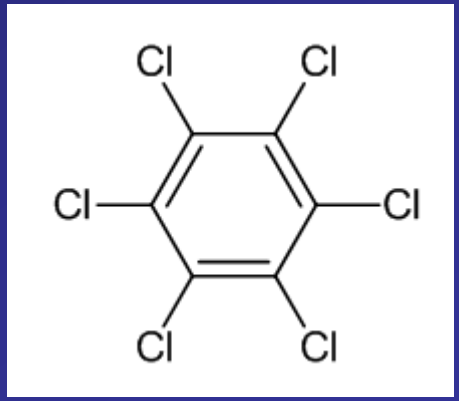
- Many of these chemical forms are no longer in use but could still be encountered in the environment
- Fungicides in general are still widely used because fungi themselves produce deadly fungitoxins such as phallotoxins (*Amanita*) and aflatoxins (*Aspergillus*)

- Hexachlorobenzene
- Organomercurials
- Phthalimides
- Dithiocarbamates

Pesticides: Fungicides

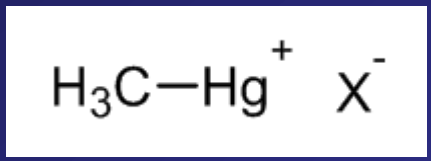
Hexachlorobenzene

- Prior to 1960 used on seed grain before planting to prevent fungal infestation
- Toxic responses: skin blister, hepatomegaly, thyroidmegaly, arthritis, osteomyelitis, osteoporosis
- HCB toxicokinetics unknown
- Persistence in environment similar to other organochlorines (DDT)
- Biomagnification quite likely



Pesticides: Fungicides

Organomercurials

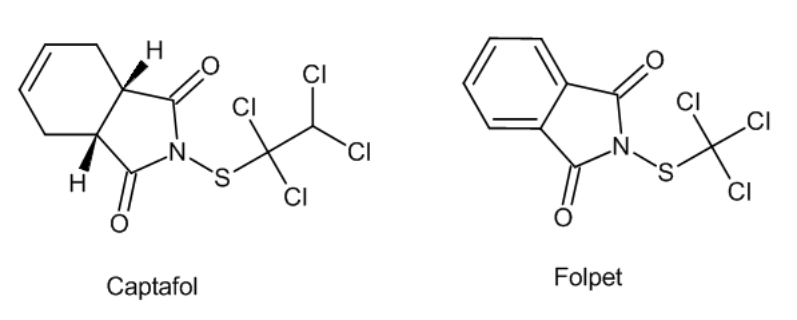


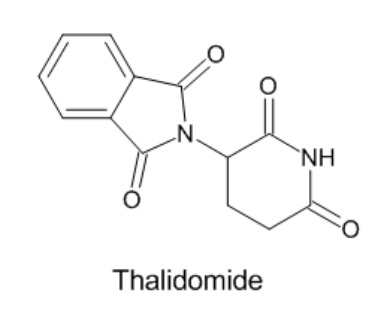
- Methylmercury is representative
- Methylmercury had been used to treat seed grains
- In past poisonings showed severe neurotoxic effects; crosses blood-brain barrier
- Linked to severe effects in fetal development and to autoimmune disease
- Strong evidence of bioaccumulation in food chains

Pesticides: Fungicides

Phthalimides

- Captafol, Folpet are representative
- Strong structural similarity to thalidomide*
- Percutaneous & respiratory routes of entry
- Biotransformation rapid, elimination in urine & feces
- Acute toxicity: irritant, allergic contact dermatitis
- Chronic toxicity: mutagenesis, carcinogenesis, teratogenesis (all lab animals)
- LD_{50}: 10,000 mg/kg (rats)
- * Thalidomide causes severe and fatal birth defects but apparently has limited medical uses



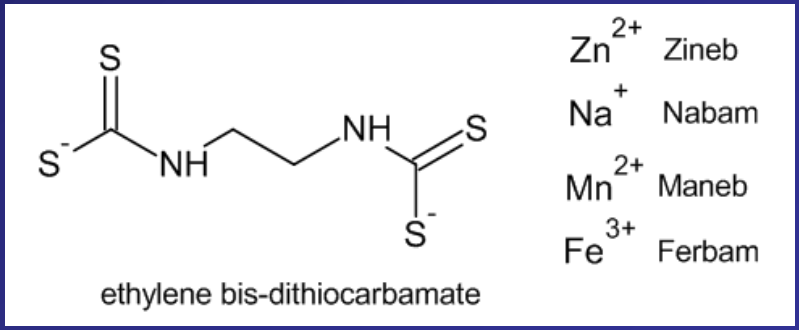


Pesticides: Fungicides

Dithiocarbamates

- ethylene-bis-dithiocarbamate (EBDC) representative

  Ferbam (Fe^{3+}), Maneb (Mn^{2+}), Nabam (Na^{+}), Zineb (Zn^{2+}) are metal cation forms
- dithiocarbamate derivatives also used as insecticides (AChase inhibitors) & in chemical/rubber industries
- routes of entry: skin, lungs, GI tract
- distribution, biotransformation, excretion all "rapid"
- toxicity: contact dermatitis, irritant, CNS depression
  animal studies show mutagenesis/carcinogenesis potential



Pesticides: Rodenticides

- Insecticides, herbicides and fungicides are chemicals synthesized so as generally not to have toxicity to humans when "applied using manufacturer's recommendations
- But many of the toxic chemicals used to control pests that are mammalian (rats, mice) have moderate to strong toxicity to humans
- Adult exposures are accidental percutaneous and respiratory entry routes
- But exposure to children can include accidental ingestion
- Anticoagulants
- Cell Respiration Inhibitors/Uncouplers
- Vasoconstrictors
- Diabetogenics

Pesticides: Rodenticides

Anticoagulants

- Warfarin (coumadin) is representative
- Anticoagulants interfere with vitamin K recycling: vitamin K is used to synthesize proteins required for blood clotting (prothrombin)
- half-life directly dependent on plasma protein binding
  warfarin shows 97% binding, $t_{1/2}$ = 40 h; heparin has little binding, $t_{1/2}$ = 1 h
- Acute toxicity: nosebleeds, bruising, internal hemorrhaging, GI tract bleeding, stroke

Pesticides: Rodenticides

Cell Respiration Inhibitors

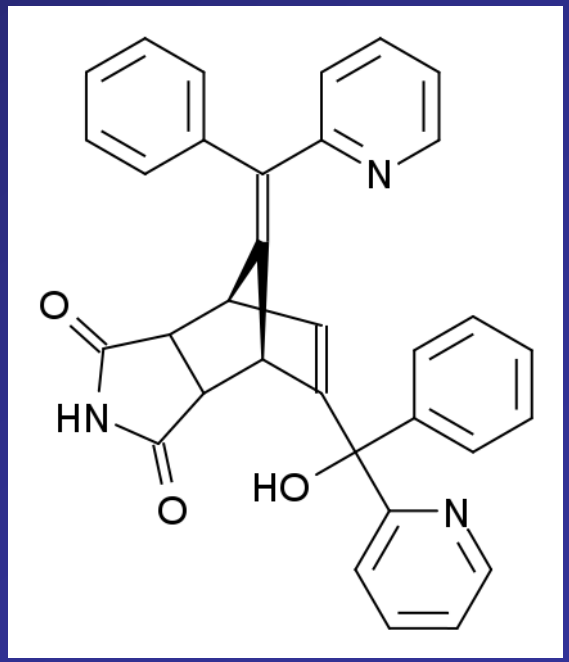
- sodium fluoroacetate (CF_3COONa) [SFA], fluoroacetamide (CF_3COONH_2) representative
- These block enzymes in Krebs (TCA) cycle, which block ATP production
- Acute toxicity: nausea, vomiting, abdominal pain, increased heart rate, kidney failure, coma, death ($LD_{50} \leq 10$ mg/kg SFA)

Pesticides: Rodenticides

Vasoconstrictors

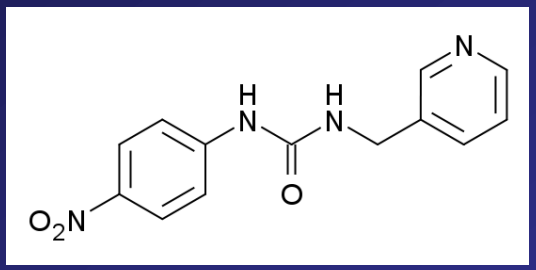
- norbormide representative
- rings of smooth muscle at the entryways of bifurcating arterial vessels contract without the normal autonomic control

  chemical and nervous signals typically control smooth muscle in arteries and arterioles to direct blood flow to tissues/organs in a prioritized way; poisons can override this control
- poisons can cause irreversible vasoconstriction, causing ischemia leading to necrosis, then death
- 5-15 mg/kg rats
  up to 300 mg/kg exposure not toxic in humans



- mechanism of action appears to be
  it keeps myocyte Ca^{2+} channels open, not allowing muscle to relax

Pesticides: Rodenticides



Diabetogenics

- pyriminil representative
- Mammalian pancreases produce insulin, which regulates the entry (transport) of glucose into many cell types

  Note that cell types/tissues/organs such as the brain do NOT depend on insulin to obtain glucose, a vital physiological characteristic for tissues that depend on a constant, high supply of glucose
- Pyriminil apparently targets and kills the beta cells of the pancreas which produce insulin: if insulin can no longer be produced, cells dependent on insulin (skeletal muscle, adipose tissue, etc) for transport of glucose into the cells no longer get glucose, so the cells and their tissues die
- Pyriminil is no longer approved for use in US (EPA); it and other diabetogenic rodenticide with toxicity to humans (same mechanism) will likely never be approved
- Acute toxicity would manifest with neurological disorders caused by transient hypoglycemia

Plastics

- Plastics are any polymeric substance that can be shaped by heat or pressure to the form of a cavity or mold
- Thermoplastics make 80% of all plastics and can be remelted or remolded (polyethylene, polypropylene, polyvinylchloride, polystyrene); thermosetting plastics cannot be remelted or remolded
- Most are not degradable, biologically or by UV radiation (under sunlight)
- Only 9% (2.8 million tons) of all plastic was recycled in 2012
- The burning of plastic as a means of disposal, which happens in many developing countries, produces worse toxicants, such as dioxane, a known carcinogen and absorbed by inhalation

Metals

- 80 metals (e.g., Fe, Mg, Zn) are required in human biochemistry, but others (e.g., Pb, Hg, Cd) are toxic
- Routes of entry are either by ingestion or inhalation
- Elimination routes typically in urine, but can be excreted back into intestinal lumen

- Arsenic (As)
- Chromium (Cr)
- Mercury (Hg)
- Lead (Pb)
- Nickel (Ni)

Metals: Arsenic (1 of 2)

- ubiquitous: used paints/dyes, metals, drugs, soaps, semiconductors, weed killers (used heavily in golf courses until 2013), animal medicines (!)
- soil levels: 10 mg/kg, groundwater: < 1 µg/L
- volatile forms in air: ~20-30 ng/m^3 in urban areas
- dietary intake: 1-20 µg/day; grains, fruit, vegetables
- inorganic arsenic
  - As(III)/As^{3+}: As_2O_3, $As(OH)_3$
  - As(V)/As^{5+}: As_2O_5, $AsO(OH)_3$
- organic arsenic
  - monomethylarsonic acid (CH_3AsO_2OH) [MMA]
  - dimethylarsonic acid [(CH3)2AsO–OH) [DMA]

Metals: Arsenic (2 of 2)

- absorption is usually through the GI tract, but inhalation also possible
- biotransformation: inorganic forms reduced & methylated to organic forms
- elimination: usually via urine ($t_{1/2}$ about a few days) 70-100% organic form, 0-30% inorganic
- acute toxicity: anorexia, hepatomegaly, cardiovascular failure, death (LD threshold > 70 mg)
- chronic toxicity: CNS/PNS neuropathologies, muscle weakness, typically shows up in horizontal white bands in nail beds (Mee's lines)
- carcinogenesis: oral ingestion of drinking water when several hundred micrograms per liter induces bladder, liver, skin cancers

Metals: Beryllium

- metal used in alloy production, released in coal combustion, volcanic ash
- if inhaled, acute toxicity produces lungs that are fibrotic, smaller in size, with cyst development ("honeycomb lung"); skin contact causes a dermatitis
  oxides in H_2O insoluble, but Cl, F, NO3, SO4 forms soluble
  poorly absorbed in skin and GI tract
- carcinogenic: beryllium particles can reside in particles for years

Metals: Cadmium

- Used in both manufacturing processes and household products
- typically inhaled and produce pulmonary edema
- increased absorption in GI tract may be associated with low iron levels
- acute toxicity of ingested Cd shows as nausea/vomiting, abdominal pain
- chronic exposure leads to nephrotoxicity (to tubules)
- binds to plasma proteins, concentrates in the kidneys, and with a half-life of 30 years (!) (0.001% excreted per day)
- Ni-Cd (ni-cad) battery workers in UK and Sweden found to be at increased risk for prostate and lung cancers

Metals: Chromium

- Most exposure is to workers manipulating Cr in manufacture: stainless steel, paint pigments, wood preservatives, leather tanning
- Cr^{3+} involved in metabolism of glucose, fat, protein
- Cr^{6+} can be reduced to Cr^{3+} but if it enters cells it may exert its most toxic effects
- routes of entry: oral, inhalation
- distributes to all tissues
- urinary elimination
- acute toxicity: damage to GI tract, immune system, hematopathies
- carcinogenesis
  - no reliable results by oral route
  - respiratory system cancers for workers by inhalation
- skin exposure: contact dermatitis, skin ulcers

Metals: Lead

- ubiquitously found ceramics, paints, automobile exhausts
- exists as Pb^0, Pb^{2+} (typical inorganic) and Pb^{4+} (usually organic)
- absorbed by inhalation or ingestion
- in GI tract:
  - passive diffusion in trans- and paracellular pathways
  - active transport for systems to absorb calcium & iron
- binds to RBCs in blood
- accumulates in bone (Ca^{2+} replacement) where it has a half-life for 20 years
- acute toxicity (0.1 mg/dl) produces pathologies in blood, kidneys, and CNS
- children particularly susceptible: Pb-based encephalopathy, appetite loss, ataxia, coma, death
- known carcinogen in animals, suspected in human

Metals: Mercury

- Hg in all forms (Hg^0 liquid metal, inorganic salt, organic forms) is toxic
- Uses: industrial catalysts, batteries, pigments, preservatives, switches, CFLs, fungicides
- Organic-Hg absorbed by GI tract and accumulates in brain
- Inorganic Hg salts accumulate in kidneys
- Excretion is in urine or feces (depends on form)
- Acute toxicity: Hg^0, organic-Hg: CNS, kidney
- Mechanisms of toxicity range from complexing with thiols, induction of oxidative stress, perturbing Ca^{2+} mobilization

Metals: Nickel

- common in alloys, batteries, coins, electronics, food processing
- 70-150 µg/day ingestion from food & water
- absorption in lungs, but less so in GI tract
- binds to plasma proteins
- absorbed Ni eliminated in urine
- some individuals sensitive to metal and get dermatitis ("nickel itch") in handling coins or from costume jewelry
- studies report cancer risk for lungs and nasal cavities for workers of nickel

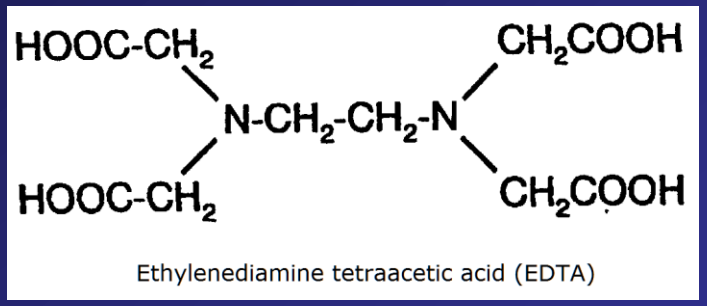
Mechanisms of Metal Toxicity

Metal cations are toxic in several ways

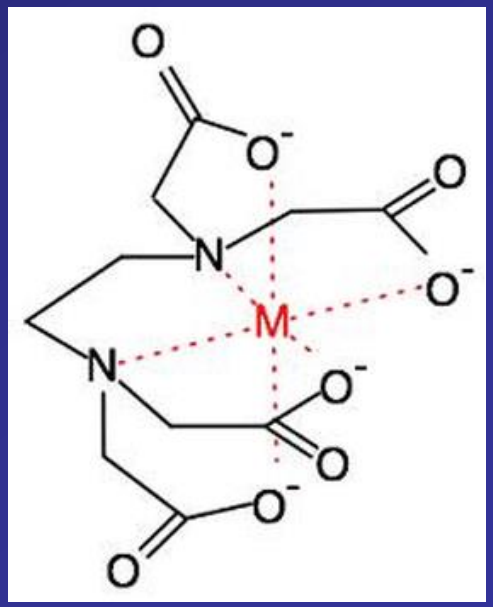
- For metal-dependent enzymes (e.g., superoxide dismutase requires Cu and Zn), another metal can place itself within the Cu and/or Zn-binding site of the enzyme: the catalytic effect is lost because the correct metal ion is not in place, so the vital biochemical reaction does not occur
- In the case of arsenic, the AsO_4 (arsenate) substitutes for PO_4 (phosphate) in the formation of ATP: but arsenate forms of the vital molecule are not stable and instantly hydrolyze, causing the cell to lose energy quickly, leading to cell death

Chelation Chemistry

- Chelation: process of chemically sequestering a metal cation for the purpose of rendering it (relatively) non-toxic or at least excretable
- Chelators: chemicals effectively forming coordinate bonds using negatively charged atoms or atoms with unbonded electron pairs in their valence shell (e.g., O^- and N: atoms in EDTA)
- Thiols bind metals also, and dithiols used in chelation



Ethylenediamine tetraacetic acid (EDTA)

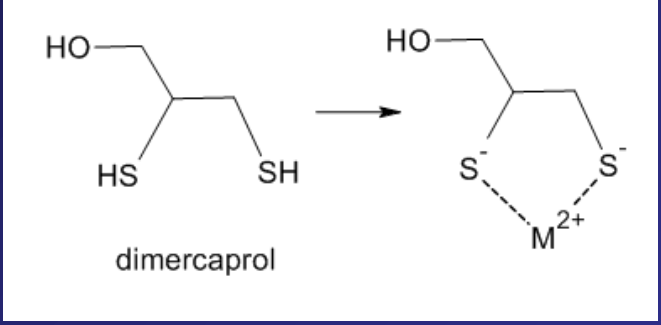


Chelators

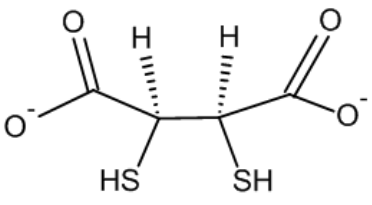
- Dimercaprol (British anti-Lewisite, BAL)
- meso-2,3-Dimercaptosuccinic acid (DMSA)
- 2,3-dimercapto-propane sulfonate (DMPS)
- Penicillamine
- EDTA

Generally used for As, Hg, and Pb acute exposures

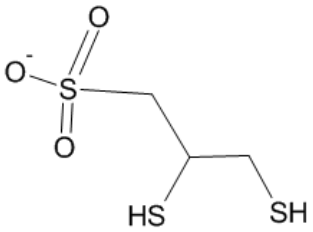
Chelators include anionic moiety (carboxylate or sulfonate) to promote excretion



dimercaprol



DMSA



DMPS

Chelation Therapy

- Term does not strictly refer to treatments for life-threatening acute (over)exposure to metals
- Includes controversial application of chelators to treat heart disease (atherosclerosis) and autism
- Chelators quite nonspecific: target & eliminate essential divalent metal cations (Ca, Mg, Fe) which are vital in all cell processes, particularly myocardial function
- Systemic therapy to be used only in life-threatening metal-caused exposures

Organic Solvents

- aliphatic alcohols
  - ethyl alcohol
  - methyl alcohol
- chlorinated aliphatics
- carbon disulfide (CS_2)
- glycols
- aromatic hydrocarbons
- benzne
- alkylbenzenes: toluene, xylenes

Organic Solvents

Aliphatic Alcohols

- effects of acute and chronic exposure to ethyl alcohol covered elsewhere
- methyl alcohol (methanol), or "wood alcohol," can be absorbed through skin, lungs, and GI tract and can cause blindess even in small doses as retinal neurons are particularly sensitive

Chlorinated Aliphatics

- chloroform ($CHCl_3$) had been used as an anesthetic until early 1900s, but it is toxic to kidneys, liver and heart
- carbon tetrachloride (CCl_4) is a dry cleaning solvent and highly toxic to liver, probably because it depletes CYP enzymes and leaves liver vulnerable to other toxicants

Organic Solvents

Carbon Disulfide

- CS_2 is used in cellophane & semiconductor production (sometimes pesticides as well)
- It is inhaled, biotransformed into sulfur-containing metabolites excreted in urine
- In brain, CS_2 can cause severe toxicity with brain damage, sleep disturbances, memory loss, Parkinson's disease-like symptoms

Glycols

- ethylene, propylene, and diethylene glycols are ingredients in automobile anti-freeze

Air Pollution

Nitrogen oxides (*NO_x*)

NO (nitric oxide), NO_2, N_2O (nitrous oxide), N_2O_2 (dinitrogen dioxide), NO_3 (nitrogen trioxide), N_2O_3 (diN triO), N_2O_4 (diN tetraO), N_2O_5 (diN pentO)

- route of entry by inhalation (all gases)
- NO_2 may reduce lymphocyte counts, increase susceptibility to infection, sensitize lung tissue with damaging inflammatory response, alter cellular architecture in alveoli
- NO is produced in body as signal mediator: action of nitroglycerine to cause strong vasodilation is via NO

what effect is there as an air pollutant?

Air Pollution

Ozone (*O_3*)

- Produced in photochemical smog
- Strongly reactive with C=C: unsaturated fatty acids a target, forming lipid peroxides
- Forms reactive oxygen species: singlet oxygen, OH radicals, H_2O_2, which attack DNA, other biomolecules
- Acute exposure cause injury/death upper respiratory epithelial cells with exudates & inflammation, which resolves in about a week
- Poorly absorbed in lungs, so does not distribute well throughout body

Air Pollution

Sulfur dioxide (SO_2)

- Highly water soluble: inhalation exposures generally confined to upper airways
- Irritation caused by forming acids when reacted with H_2O
- Acute overexposure may induce a bronchoconstriction and also diminish pulmonary defense (mucociliary transport, alveolar clearance of deposited particles, and macrophage function)

Air Pollution

Particulates

- Many airborne particles carry toxic substances on their surface
- Toxic metal exposure by inhalation results from metals covering surface of particles

Environmental Persistence

- "Persistent" toxicants distribute quickly to "low metabolic turnover" storage depots

  lipophilic substances to adipose tissue
- Become part of food chain or web
- Bioamplification

  increase in stored mass of toxicant in organism at higher level in food chain or web
- Biomagnification

  synonym for bioamplification
- Bioaccumulation

  toxicant shows increase in levels in tissue faster than is removed
- Bioconcentration

  A toxic solute bioaccumulated from an aqueous solution

Sources

- WW Hughes, *Essentials of Environmental Toxicology: The Effects of Environmentally Hazardous Substances on Human Health*, (Philadelphia: Taylor & Francis), 1996
- SM Roberts, RC James, PL Williams, eds., Principles of Toxicology: Environmental and Industrial Applications, 3rd Ed, (Wiley), 2015
- DL Costa, "Air Pollution," Chap 28 in CD Klaassen, ed., *Casarett and Doull's Toxicology: The Basic Science of Poisons*, 7th Ed (New York: McGraw-Hill), 2008

Group Work

Research access to assessment protocols, laboratories and specific analyses for management of toxic exposure

Review these with instructor

Take-home midterm to be posted to MOODLE