

## Introduction to Toxicology

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## Toxicology

1. It is a branch of science that deals with the study of the adverse effects of chemical agents on biologic systems. Can include other agents such as radiation, microbial toxins etc.
2. A chemical that interacts with an organism that is not found in the normal metabolic pathway of that organism is called a xenobiotic.

## LD50

- It is a dose that kills 50% of the exposed animals.
- Can vary among chemicals
- Examples:

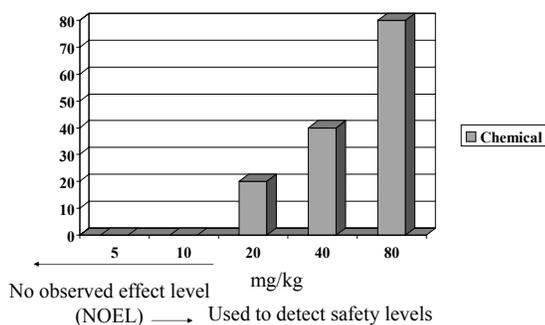
	<u>LD50 (mg/kg body weight)</u>
• Ethyl alcohol	10,000
• Morphine	900
• Dioxin (2,3,7,8-tetrachloro dibenzo-p-dioxin:TCDD)	0.001
• Botulinum toxin	0.00001

## Toxicity Testing in Lab Animals

Exposure:

- Acute: Usually a single dose within 24 hours.
- Subacute: repeated exposure to a chemical for one month or less.
- Subchronic: 1-3 months
- Chronic: > 3 months

## Dose response relationship



## Testing Lab animals

- Commonly used:
  - Rats, mice, dogs, monkeys etc
- Safety in experimental animals does not necessarily indicate the same in humans.
  - Example: Thalidomide a human teratogen shows toxicity at doses as low as 0.5-1 mg/kg and has little or no effect in mice or rats at doses as high as 4000 mg/kg.

## Branches of Toxicology

- Biochemical toxicology: Deals with how a chemical may induce biochemical and molecular events leading to elimination of the chemical as well as induction of toxicity.

## Environmental contaminants

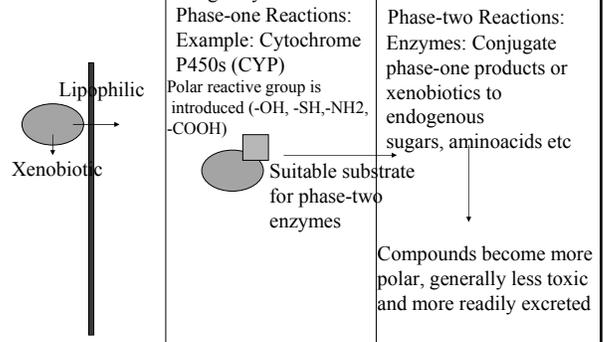
- Halogenated aromatic hydrocarbons (HAH) belong to a chemical family that includes polychlorinated and polybrominated biphenyls (PCBs and PBBs).
- HAHs are found in herbicides, automobile exhaust, combustion of municipal and industrial waste, pulp and paper manufacturing etc.
- They persist in environment.
- Dioxin (TCDD) is the most potent HAH compound that is considered to be human carcinogen.

## How does body detoxify?

- When exposed to foreign hydrophobic chemicals, the body produces cytochrome P450s (CYPs) which are heme-containing enzymes involved in oxidative metabolism of compounds.
- Humans have 17 known CYP gene families.
- CYP 1A1 (Cytochrome P450 1A1) plays a critical role in detoxification of polycyclic aromatic hydrocarbons.

## How are xenobiotics metabolized?

Through a large number of phase-one and phase-two xenobiotic-metabolizing enzymes

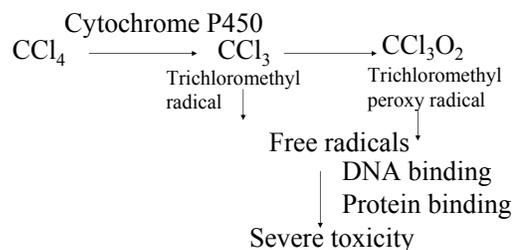


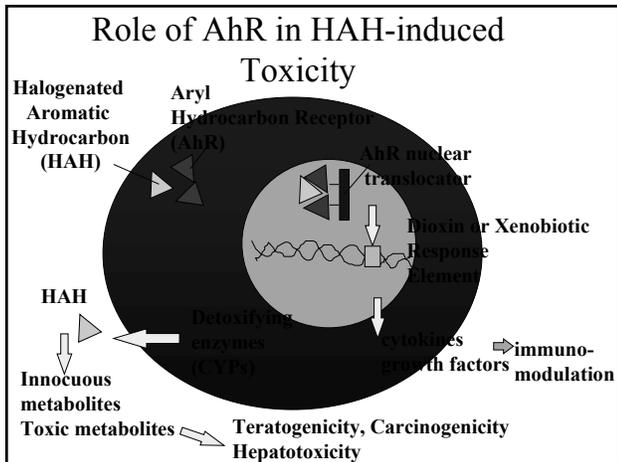
## How are toxicants metabolized?

Summary:

- By a wide array of enzymes: Convert lipophilic to hydrophilic.
- Phase-one reactions in which a polar reactive group is introduced into the xenobiotic molecule. Example: cytochrome P450s.
- Phase-two reactions in which the enzymes conjugate the xenobiotic to sugars, aminoacids etc, forming water-soluble products that are readily excreted.
- Sometimes the metabolites are more toxic than the parent molecule.

## Carbon Tetrachloride

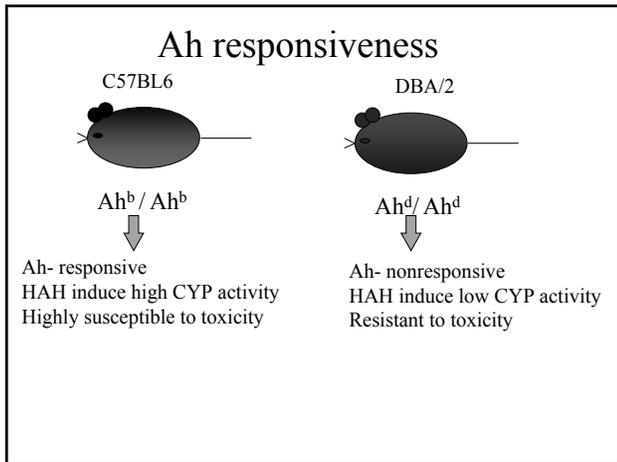




## AhR

**Summary:**

- Halogenated Aromatic Hydrocarbons such as dioxins, react with the Aryl hydrocarbon receptor (AhR).
- This is a prerequisite to induce the production of xenobiotic metabolizing/detoxifying enzymes.



## Carcinogenesis

- **Carcinogenesis:** Process by which chemicals cause cancer.
- Many environmental chemicals/agents are known to cause human cancer:
  - Tobacco, cadmium, coal tar, cigarette smoke, UV light, X-rays etc
  - A large number of chemicals are classified as carcinogens based on their ability to induce tumors in animal models.
- Chemicals can induce tumors by directly damaging or modifying DNA (genotoxic) or indirectly by suppressing the immune system, changing hormone balance etc

## Mutagenesis

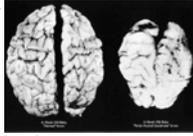
- **Mutagenesis:** Toxicity on genetic material and its inheritance.
- Mutagens include ionizing radiation, alkylating agents, most carcinogens

## Teratogenesis

- **Teratogenesis:** Deleterious effects on developmental process.
- Thalidomide is a well known teratogen for human species—was introduced as a sedative. It was teratogenic even when taken once during 3-7<sup>th</sup> week of pregnancy.
- It caused shortening or complete absence of limbs.
- Methyl mercury
- Alcohol
- Diethyl stilbestrol (DES)—a synthetic estrogen

## Organ toxicity

- Neurotoxicity:
- Neurons can not normally regenerate.
- Thus toxic effects can be permanent.
- Neurotoxic chemicals can cross blood brain barrier easily.
- Examples:
  - Lead, mercury, certain food additives, pesticides, industrial and/or cleaning solvents, etc
  - Mercury: In Japan, in 1950s, a chemical plant released large amounts of mercury into the bay. People who ate contaminated fish suffered from severe neurotoxicity.

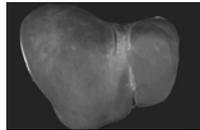


## Neurotoxicity

- Pesticides: Organophosphorus (OP) and carbamate insecticides inhibit acetyl choline esterase, an enzyme that breaks down acetyl choline.
- In humans, exposure to OP compounds causes muscle weakness, paralysis of respiratory muscles and death.
- Property of OP compounds used to develop “nerve gas”.
- A contaminant of “synthetic heroin”, MPTP (1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine), causes irreversible brain damage and Parkinson’s-like symptoms.

## Hepatotoxicity

- Liver is highly susceptible to xenobiotic-induced toxicity.
- It is the first organ to encounter the xenobiotic when ingested orally.
- Carbon tetrachloride is the most widely studied. Cytochrome P450 converts it into a highly reactive free radical which causes toxicity.



## Nephrotoxicity

- Kidneys: Heavy metals are potent nephrotoxins: Cadmium, mercury, lead etc
- Effects:
  - Glucosuria, proteinurea, renal necrosis and death.



## Pulmonary Toxicity

- Inhalation of silica (mining and quarrying area) causes fibrosis or formation of collagenous tissue.
- If large, can impair respiration.
- Asbestosis: may cause lung cancer.
- Cigarette smoke has polycyclic aromatic hydrocarbons such as benzo(a)pyrene which is metabolized by cytochrome P450. The metabolites may trigger cancer.

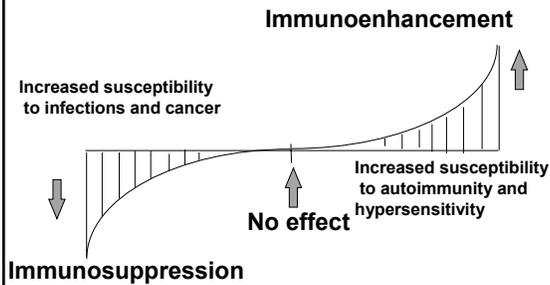
Silicosis



## Reproductive toxicity

- A large number of chemicals are toxic to the male or female reproductive system.
- Can cause decreased sperm count.
- Some environmental chemicals such as dichlorodiphenyltrichloroethane (DDT), DDE etc can mimic human estrogen. They are called “environmental estrogens”.
- In wildlife they cause hermaphroditic fish and other reproductive anomalies (sex reversal) in alligators.

## Effect of Chemicals on the Immune Response



## Immunotoxicology

- Late 1970-early 1980s--->Chemicals can alter functions of the immune system.
- Such toxicity is called Immunotoxicity.
- Several of these chemicals are environmental pollutants: organo metals, halogenated hydrocarbons, carbamates, organophosphates.
- Chemicals can alter the immune functions at very low concentrations--doses that do not produce organ toxicity.
- Example: 0.25 ng of TCDD(dioxin) can increase the susceptibility of mice to influenza virus.

## Immunotoxic Compounds

- ❑ **Environmental Contaminants:** Insecticides, pesticides, herbicides and heavy metals. Specifically, halogenated biphenyls(PCB, PBB), TCDD, vinyl chloride, hexachlorobenzene.
- ❑ **Pesticides:** DDT, parathion, Lindane, Methylnitrofos, Hepatochlor, Carbamate, paraquat, chlorophos.
- ❑ **Dusts:** Silica, carbon.
- ❑ **Heavy metals:** Lead, Cadmium, mercury, selenium, arsenic, cobalt, nickel, silica, chromium, platinum
- ❑ **Addictive substances:** Alcohol, heroin, cigarette smoke
- ❑ **Food additives:** pyrogallol, carrageenan etc

## Role of Ah receptor in HAH immunotoxicity

- Immunotoxicity of HAH(TCDD, PCBs and PBBs) is also regulated by Ah receptor.
- Polychlorinated biphenyls (PCBs) are used in plasticizers and other industrial applications.
- Polybrominated biphenyls (PBBs) are used in Firemaster, a flame retardant. In Michigan, Firemaster was accidentally substituted for magnesium oxide food supplement for livestock.

## Dioxin (TCDD) Toxicity in Humans

### Agent Orange

- Dioxin was the primary toxic component of Agent Orange.
- US troops during the Vietnam War released around 19 million gallons of defoliant from 1962-1971.
- Conditions recognized as service-connected for Vietnam veterans based on exposure to Agent Orange or other herbicides:
  - Chloracne
  - Non-Hodgkin's Lymphoma
  - Soft Tissue Sarcoma
  - Hodgkin's Disease
  - Porphyria Cutanea Tarda
  - Multiple Myeloma
  - Respiratory Cancers (lung, larynx, trachea and bronchus)
  - Prostate Cancer
  - Peripheral Neuropathy (acute or subacute)
- Conditions recognized in the children of Vietnam Veterans:
  - Spina bifida
- High levels of dioxin found at ground zero of WTC



## Industrial Accidents

- Seveso, Italy: Industrial accident led to release of dioxin in air. Exposed over 37,000 people to toxicity.
- Bhopal, India: Union Carbide factory released methyl isocyanate—killed thousands of people.



## Immunotoxicity of Metals

- Chromium, nickel, lead, cadmium etc. suppress humoral immunity
- Platinum, gallium, thorium etc. suppress cell-mediated immunity

## Mercury

Mercury (Hg) is a toxic metal that can exist as a pure element or in a variety of inorganic and organic forms and can cause immune, sensory, neurological, motor, and behavioral dysfunctions similar to traits defining or associated with autism.

Mercury compounds (thimerosal) are used as preservatives in nasal solutions, ophthalmic drugs, vaccines etc which has generated controversy on the possible toxicity.

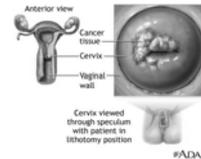


## Environmental estrogens and immunotoxicity

- Estrogen: Female sex hormone.
- Administration of estrogens is known to induce rapid thymic atrophy in rodents.
- Estrogens are also known to suppress humoral and cell-mediated immune responses.
- Females are more susceptible to lupus and other autoimmune diseases when compared to the males.
- Diethylstilbestrol (DES) is a synthetic estrogen used in humans, known to cause thymic atrophy and immunosuppression.
- Several pesticides of the chlorinated hydrocarbon class have estrogenic activity. Such compounds called environmental estrogens also cause immunosuppression.

## DES-mediated toxicity

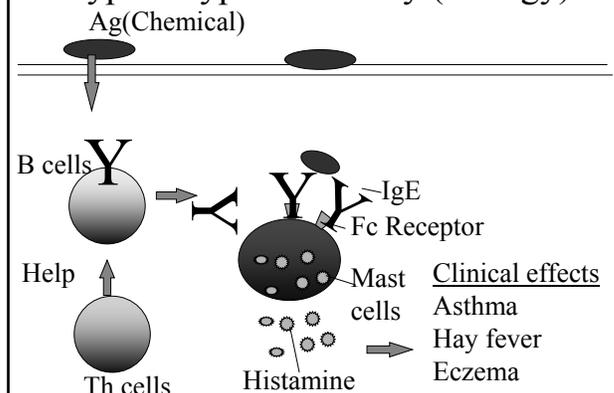
- In USA and Europe, between 1940-1980s, >10 million women were treated with DES during pregnancy.
- “DES mothers” have a higher risk for development of breast cancer and “DES daughters” have a higher risk for developing cervicovaginal cancer and autoimmunity.

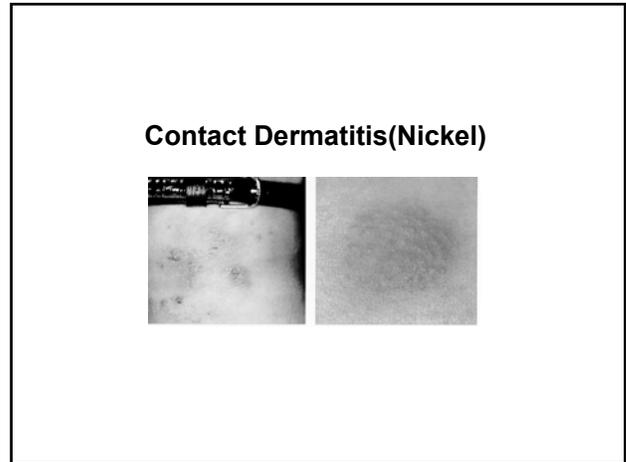
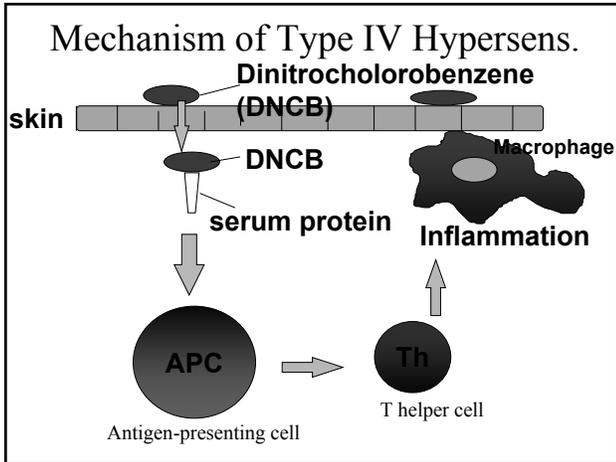


## Hypersensitivity induced by chemicals

- Two types:
  - Type I hypersensitivity (allergy): IgE Abs, Neutrophils
  - Type IV hypersensitivity (contact dermatitis): T cells, macrophages

## Type I Hypersensitivity (Allergy)





- ### Agents that cause allergies
- Asthma: Major type of hypersensitivity seen in industries.
  - Ex: 10% of people exposed to toluene diisocyanate develop Asthma.
  - Large number of chemicals cause contact dermatitis: Formaldehyde, antimicrobials in cosmetics, shampoos etc, heavy metals, drugs
  - Allergies to plastic and resins: contain trimellitic anhydride which binds to a self-protein and triggers allergies.

- ### Agents that cause allergies
- Beryllium: Used to coat fluorescent lamps causes berylliosis, hypersensitivity in lungs--often fatal.
  - Acts as a hapten, combines with tissue proteins and triggers Type IV hypersensitivity reaction. Chronic inflammation can lead to lung damage.

