Naturally Occurring Toxicants as Etiologic Agents of Foodborne Disease

Food Toxicology
Instructor: Gregory Möller, Ph.D.
University of Idaho

Learning Objectives

• Examine the etiology (causation) of human disease related to naturally-occurring foodborne toxicants.
• Understand the chemical complexity of foods.
• Explore goiter, tropical ataxia neuropathy (TAN), tropical amblyopia, lathyism, and their linkage to foodborne toxicants.
• Review a range of natural food toxicants that are involved in human disease.

Complexity of Food

Nutrients
• Carbohydrates
• Proteins
• Lipids
• Minerals
• Vitamins

Non-nutrients
• Food Additives
• Naturally Occurring Secondary Chemicals
• Contaminants
• Processing Chemicals

Non-nutrient Chemicals in Different Foods
• Cheddar Cheese 160
• Orange juice 250
• Banana 325
• Tomato 350
• Wine 475
Natural Toxicants and Human Disease

- Goitrogens
- Cyanogenic glycosides
- Lathyrism
- Lectins
- Alkaloids
- Protease inhibitors
- Vasoactive amines

Goitrogens

- Contribute to growth of goiters
- Compounds in Cruciferae
  - *Brassica* species (cabbage, kale, turnips)
  - Seeds only - not leaves
- Combined with iodine deficiency

**Cruciferae → Glucosinolates**

- A class of about 100 naturally occurring thioglucosides that are characteristic of the Cruciferae and related families.
- Diets of people in many parts of the world include considerable amounts of Cruciferous crops and plants.
  - Processed radish and wasabi in the Far East
  - Cabbage and traditional root vegetables in Europe and North America.
  - Rapeseed, kale, swede and turnip may also contribute since they are extensively used as animal feed stuffs.
  - Glucosinolates in crops, such as oilseed rape (*Brassica napus*) and Brassica vegetables, is undesirable because of the toxicological effects of their breakdown products.
  - Breakdown products include nitriles, isothiocyanates, thiocyanates, epithionitriles and vinyl oxazolidinethiones.

Goitrogens

- Goitrogenic compounds (goitrin) are formed from breakdown of glucosinolates by thioglucosidase
- Goitrogenic metabolites
  - Nitriles; Thiocyanates; Oxazolidine

Goitrogens

- Thyroid gland secretes thyroxine (TY), triiodothyronine (TII), thyroglobulin (TH)
- Controlled by hypothalamus and pituitary
- Hypothalamus produces thyrotropin-releasing hormone (TRH)
• Stimulates pituitary to release thyroid-stimulating hormone (TSH)

**Goitrogens**

• TSH promotes uptake of iodine, synthesis of TH and release of TY and TII which feed back to reduce TSH
• TY and TII hormones also affect
  – Oxygen consumption, cardiovascular function, neuromuscular activity, cholesterol metabolism, cerebral function
  – Growth and development

**Goitrogens**

• Goitrin and thiourea inhibit TY synthesis
• Thiocyanates, oxazolidine and nitriles inhibit uptake of iodine by thyroid
• Lack of iodine causes thyroid to enlarge and hypervascularate to trap more iodine = goiter

**Goitrogens - Testing**

• Weight and histology of thyroid
• Growth rate of patient/animal
• Iodine content of blood and thyroid
• Feed compound - measure uptake of radioactive iodine

**Cyanogenic Glycosides**

**Plant**
• Bitter almond
• Cassava root
• Sorghum
• Lima bean

**Glycoside**
• Amygdalin
• Linamarin
• Dhurrin
• Linamarin

**Cassava Root**

• The cassava (manioc, yucca; *Manihot esculenta*) is a woody shrub of the Euphorbiaceae (spurge family) that is extensively cultivated as an annual crop in tropical and subtropical regions for its edible starchy tuberous root, a major source of carbohydrate.
Cyanogenic Glycosides

- Cultivars contain varying amounts of CGs
- Cultivar, drought, and food preparation significant for CGs

**Cyanogenic Glycosides**

- Toxic chemical = hydrogen cyanide
- HCN released when plant is chewed or chopped
- Releases 2 enzymes normally separate
  - Beta-glucuronidase
  - Hydroxynitrile lyase
- Act synergistically to release HCN

**Cyanogenic Glycosides**

- Can reduce CN by chopping or grinding in water
- Cassava flour is made from boiled or fermented root
- Intestinal bacteria may also be able to break down cyanogenic glycosides to HCN

**Cyanogenic Glycosides**

**Amygdaline (Laetrile)**

- Anticancer compound
- Apricot pits
- Not approved in US
- Several deaths in foreign countries

**Acute Cyanide Poisoning: Mechanism**

- Shuts down cellular respiration - energy metabolism in mitochondria
- Binds ferric ion on cytochrome oxidase in Krebs cycle
- Min lethal dose
  0.5-3.5 mg HCN/kg bw

**Acute Cyanide Poisoning**

**Symptoms**

- Muscular paralysis
- Mental confusion
- Respiratory distress
- Rapid onset

**Acute Cyanide Poisoning**

**Treatment**

- Nitrite or amyl nitrite
• Converts hemoglobin (Fe$^{+2}$) to methemoglobin (Fe$^{+3}$)
  – Draws CN$^-$ away from cytochrome oxidase
• Add thiosulfate to form thiocyanate

**Chronic Cyanide Poisoning**

• Occurs in areas of cassava in diet
• Not well understood
• Two disorders
  – Tropical Ataxia Neuropathy (TAN)
  – Tropical Amblyopia

**Chronic Cyanide Poisoning**

  **Tropical Ataxia Neuropathy (Konzo)**
  • Atrophy of optic nerve, ataxia, mental disorders
  • More prevalent - West Africa
  • High prevalence of goiter
  • Low levels of S-containing aa
  • Elevated plasma thiocyanate (goitrogen)

**Chronic Cyanide Poisoning**

  **Dietary Modifiers**
  • No goiter with adequate iodine
  • Malnutrition increases neural effects
  • Protein-deficient diets
    – Lack S-containing amino acid to convert CN$^-$ to thiocyanate

**Chronic Cyanide Poisoning**

  **Tropical Amblyopia**
  • Atrophy of optic nerve
    – Blurred vision, blindness
  • Africa and South America where cassava is staple in diet
  • Reproducible in lab animals

**Lathyrisim**

• Consumption of peas – esp. *Lathyrus sativus*
  – Grass pea, blue sweet pea, chickling vetch, Indian pea, Indian vetch, white vetch, almorta or alverjón (Spain), *guaya* (Ethiopia), and *khésari* (India).
  – Primarily restricted to areas in Asia/Africa
• Well-known neurodegenerative disease
  – Enzyme inhibitor (BAPN)
  – Neurotoxic amino acid (ODAP)
• Hardy plant, drought resistant

Lathyism
Two forms disease
• Osteolathyism - animals
• Neurolathyism - humans

Osteolathyism
Animals
• Bone deformations
• Weakness in artery wall and connective tissues
• Beta-L-glutamylaminopropionitrile (BAPN)

Osteolathyism
Mechanism
• BAPN inhibits lysyl oxidase enzyme
• Lysyl oxidase is needed to crosslink collagen strands for strength
• Collagen is main component of connective tissue and bones

Neurolathyism
Humans
• Chronic consumption of L. sativus
• Paralysis of legs followed by general weakness and muscle rigidity
• Young man disease
• Sudden onset
  – Calf muscle spasms
• No animal model

Neurolathyism
Humans
• B-N-oxalyl-L-alpha,B-diaminopropionic acid (ODAP)

Neurolathyism
Humans
• Etiologic agent may be ODAP
• Only found with L. sativus species
• OPAP interferes with normal function of nerve synapse
  – Inhibits uptake of glutamic acid
• No animal model to study

Cholinesterase Inhibitors
• Found in a variety of plants - potato, tomato, eggplant
• Western African calibar bean (prototype)
  – Physostigmine
  – Natural carbamate

**Cholinesterase Inhibitors**

• Solanine - most studied - potato
• Glycoalkyloid - 20-100 mg/kg wet wt
• >200 mg/kg banned by FDA
• Lenape potato variety incident
  – > 300 mg/kg

**Cholinesterase Inhibitors**

  **Solanine**

• Greatest concentrations
  – Peel, around sprouts
• Natural/artificial light increases levels
  – Russet Burbank - 250-700 mg/kg – 5 days
  – Green due to chlorophyll
    • Toxicity marker

**Cholinesterase Inhibitors**

  **Solanine**

• Documented human toxicity and death
• Potato sprouts, sprouted potatoes, greened potatoes
• Gastric pain, nausea, vomiting, hyperesthesia, increased & accelerated respiration
  – 2/6 deaths
• Concentration unknown

**Cholinesterase Inhibitors**

  **Solanine - Case Study**

• 420 mg/kg total alkaloid content
• Approx 50% solanine = 200 mg/kg
• Need to consume 1 kg potato for toxicity due to only solanine
• May act with other glycoalkyloids
  – Chaconine
• Animal LD50
  500-1000 mg/kg
**Vasoactive Amines**

- Highest in cheese (aged), beer, wine
- Lower levels in banana, tomato, avocado, spinach, orange
- Spoiled meat (see food allergy)
- “Pressor Amines” (catecholamines)
  - Cause vasoconstriction, hypertension

**Vasoactive Amines**

- Meat/fish - bacterial action
  - Putrescine, cadaverine
- Banana/avocado
  - Dopamine, tyramine
- Catecholamine neurotransmitters
  - Norepinephrine, dopamine, serotonin

**Vasoactive Amines: Problems**

- Monoamine oxidase (MAO)
  - Widely distributed in body
  - Breaks down vasopressive amines
- MAO inhibitors - treat clinical depression
- Co-exposure to vasopressive amines in food
  - Mostly tyramine

**Vasoactive Amines: Tyramine Mechanisms**

- Indirect action
- Displaces normal catecholamines from granules in nerves
- Leads to hypertension
- Can be severe in presence of MAO inhibitors

**Vasoactive Amines**

*Levels in Food - mg/kg*

- **Tyramine*****
  - Cheese 20-2000
  - Avocado 25
- **Seratonin**
  - Banana pulp 30
– Avocado 10
• All others < 10, all sources

**Vasoactive Amines**

**Symptoms**

• Hypertension
  – Mild to severe
• Migraine headache
• Rare - intracranial bleeding/death

**Pyrrolizidine Alkaloids**

• A problem in food animal forage and human subpopulations exposed acutely in a toxic incident or chronically via cultural foods.
• High levels in some plants >5% dry wt
• 100 different compounds

**Pyrrolizidine Alkaloids**

• Most human exposure from herbal tea or crop contamination
  – Low level exposure from milk/meat
  – Bush tea in Jamaica
• Decreased use as herbal medicine
  – Comfrey - wound dressing (+other)
• Carcinogenic and liver toxin in animals
  – Epoxidation

**PA Hepatotoxicity: Veno-Occlusive Disease**

• Occlusive lesions are produced in the centrolobular hepatic veins (obliterating endophlebitis).
• Subendothelial edema, narrowing and occlusion of the lumina, atrophy, and necrosis of liver cells with portal hypertension are the results.

**Tajikistan, 1992**

• Due to a military blockade there was a late wheat harvest.
• Weeds were able to thrive in wheat fields.
• The harvest contained large amounts of the seed of *Heliotropium lasocarpium*.
  – The first cases occurred 6 weeks after contaminated bread was consumed.
• By the spring of 1993 there had already been more than 3900 cases.
• Patients at Stage I had abdominal pain, nausea, vomiting and asthenia.
• In Stage II hepatomegaly followed.
• Stage III was characterized by ascites.
• In the final stage there is hepatic encephalopathy.
• Case fatality ratio was 1.3% and increased with age.

Protease Inhibitors
• Anti-nutritional compounds
  – Legumes, grains, potatoes, eggplant, onion
• Inhibit gastric enzymes that break down proteins
  – e.g. trypsin (protease)
• All toxicity studies in animals
  – Human relevance unclear

Protease Inhibitors
• Pancreatic hypertrophy
  – Pancreatic hypersecretion of amino acid-rich proteins
    • aa deficiency
    • Growth retardation
• Similar mechanism of lathryism

Other Possible Natural Toxicants
• Caffeine
• Spices
• Licorice, nutmeg, sassafras
• Phytoalexins

Vicine and Convicine
• Fava bean alkaloids
• Causes favism in people who have an inherited absence of the enzyme glucose-6-phosphate dehydrogenase (G6PD) in their red blood cells
  – Headaches, dizziness, nausea, yawning, then
  – Vomiting, abdominal pain, and fever.
  – At this point, symptoms either spontaneously subside,
  – Or acute hemolytic anemia via oxidative stress occurs

Vicine and Convicine
Lectins
• Proteins or glycoproteins that bind carbohydrates
• Cause cells to agglutinate
• Basis for blood-type assays (hemagglutination)
• Decrease nutrient absorption from intestinal cells

**Lectins: Sources**
• Plant (800 species) and animal tissues
• Black beans, soybeans, lima beans, kidney beans, peas, lentils

**Lectins: Problem**
• Cause growth retardation from consumption of raw product
• Some very toxic (legume-specific)
  – Ricin - castor bean - rat LD50 0.05 mg/kg
  – Kidney bean - 0.5% rat diet two wks
• Growth retardation
  – 0.5-1% of diet black bean and soybean
• Heating destroys toxicity

**Lectins: Mechanism of Action**
• Unknown; complex
• Prevents nutrient uptake
• Gut flora involved
  – Germ-free animals show less effects
• Immune component?