

Teratogenesis, Mutagenesis, and Carcinogenesis

Food Toxicology
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Learning Objectives

- Define teratogenesis, mutagenesis, and carcinogenesis.
- Describe the relevance of replication, transcription, and translation to teratogenesis, mutagenesis, and carcinogenesis.
- Summarize the mechanism of action for teratogenesis, mutagenesis, and carcinogenesis.
- Discuss examples of known teratogens, mutagens, and carcinogens.

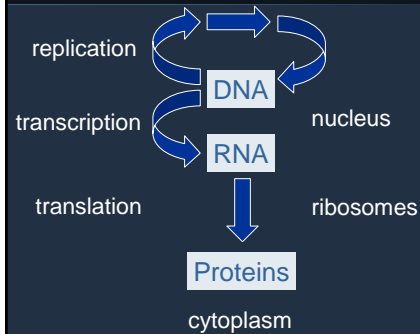
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Molecules of Life

- Toxicants can react with or modify DNA or RNA.
 - Can lead to heritable change in offspring or changes in cellular growth and development.
- Replication → perpetuate genetic information.
- Transcription and translation → express genetic information.

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The Cell



4

Protein Functions

- Antibodies.
 - Recognize molecules of invading organisms.
- Receptors.
 - Part of the cell membrane; recognize other proteins, or chemicals, and inform the cell.
- Enzymes
 - Assemble or digest.
- Neurotransmitters, hormones
 - Trigger receptors.
- Channels and pores.

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Endpoints

- Teratogenesis.
 - Origin or production of malformed fetuses or offspring.
- Mutagenesis.
 - Production of a mutation or change in the genetic code of an organism.
- Carcinogenesis.
 - Cancer formation including carcinoma and malignant neoplasms.

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DNA Replication

- Structure implies replication
- Occurs via multiple enzyme action
- Helix unravels, strands part, DNA replicates
- Mitosis, meiosis
- Not always perfect
 - Repair enzymes

7

Replication

- Duplicates cell DNA.
- Mitosis - one somatic cell with $2n$ chromosomes divides to create two cells with $2n$ chromosomes (humans, $n = 23$).
- Number, quality and quantity of chromosomes per cell is conserved.
 - Triggers for mitosis (receptors + proteins).
 - External signals.
 - Hormones.
 - Internal factors.
 - Growth factors.

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Replication, 2

- Meiosis - germ cells are cells that divide into gametes.
 - 2 cell divisions.
 - Four daughter cells.
 - Each with a different set of chromosomes.
 - Each with 1 set that will be joined by another in fertilization.

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DNA Transcription

- DNA is copied via expendable mRNA
- mRNA codes for specific proteins
- Occurs in nucleus of cell

10

DNA Translation

- Occurs in the cytosol
- Interaction of mRNA, tRNA, amino acids and enzymes
- tRNA has three-base codons which correspond to different aa
- AA are added one at a time to form chain - polypeptide
- Polypeptide corresponds to protein with a specific aa sequence

11

Transcription and Translation

- DNA: double strand of nucleotides.
 - Nucleotide = nucleic acid, sugar and phosphate.
 - **C**ytosine, **T**hymine, **U**racil; **A**denine, **G**uanine.
 - Base pairing = A-T, G-C.
 - Gene: sequence of bases that code for a specific sequence of amino acids (protein).
 - Codon: sequence of 3 bases that code for a single amino acid, i.e.
 - AGC → Serine.
 - AAA → Lysine.

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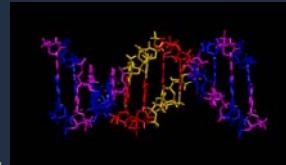
Transcription and Translation, 2

- Transcription = copying.
 - DNA unzips and enzymes make RNA “copy”.
 - Differences:
 - T → U (UA not TA).
 - Deoxyribose → ribose.
 - mRNA formation; transport to cytoplasm.
- Translation = protein formation.
 - mRNA (blueprint).
 - rRNA (support).
 - tRNA (a.a. transport).

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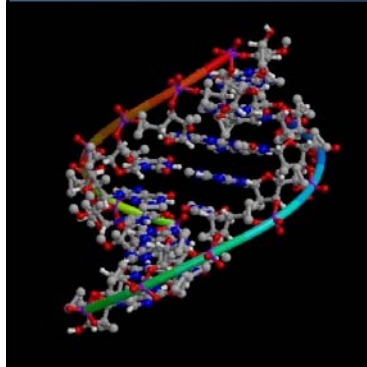
DNA Structure - Function

- Nucleotides form chains
- 3 nucleotides form a codon
- Multiple codons form genes
- Multiple genes form chromosomes
- Multiple chromosomes form DNA

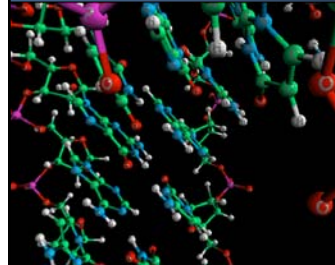


14

DNA/RNA Complex



DNA/RNA Complex, 2



16

Errors in DNA Replication, Transcription and Translation

- Base pairing
- Repair enzymes and other enzymes
- Regulatory genes, operons, termination sequences
- Methylation patterns
- Post transcriptional/translational processing

17

DNA/Chemical Interactions

- Alkylation - covalent adduct between DNA and chemical
- Intercalation - noncovalent binding of chemical between two adjacent base pairs
- Cross-linkage - Inter or intrastrand covalent binding of chemical
- Breakage - scission of one or both strands of DNA

18

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Aflatoxin B₁ – DNA Adduct

19

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Teratogenesis

- Teratology: the study of the frequency, causation, and development of congenital malformations.
- Complex mechanisms and timing of disruptive interaction during embryogenesis.
- Some natural “bad path” spontaneous abortion.
 - Humans: critical 1st 8 wks gestation.
 - Embryonic stage.
 - Morphological defects in specialized tissues and organs.
 - Fetal stage exposure.
 - Developmental or neoplastic endpoints.
- Known human teratogens.

20

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Example: Teratogenesis

Five Legged Frog

21

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Example: Teratogenesis, 2

Ovine Cyclopia
Veratrum Californicum
W. False Helebores

22

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Example: Teratogenesis, 3

Crooked Calf Disease

Lupine

OCC1CN2CCCC2C1

Quinolizidine alkaloids
Anagyrene

23

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Case Study: Lupine Alkaloid Birth Defects


- In September 1980, a baby boy born in the mountainous back-country of northwestern California (Trinity County) was brought to the UC Medical Center in Sacramento with severe bone deformities in his arms and hands, including a partial absence of forearm bones (radial aplasia) and absent thumbs.
- Extensive medical histories and genetic analyses of his parents indicated that the probable cause was environmental rather than hereditary.
- His mother feared that somehow exposure to herbicide spraying was responsible. Association of forest spraying and a reportedly high incidence of birth defects in northwestern California and southern Oregon has been highly publicized in recent years and has become controversial. Indeed, it appears likely that this herbicide had been applied to a forested ridge several miles distant from the mother's home more than a year before the child's conception.

24

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Case Study: Lupine Alkaloid Birth Defects

- The mother provided the evidence that her goats also gave birth to kids stillborn or with deformed legs during and after the period of her pregnancy, and that puppies born to a dog fed the goat's milk during pregnancy were likewise deformed.
- Local goat's milk has become a common food item in the area, and the child's mother drank it regularly herself throughout pregnancy.
- A thorough survey of nearby areas where the goats had regularly browsed at the time of the mother's early pregnancy showed that a perennial lupine, identified as the widely distributed *Lupinus latifolius*, often formed the principal low-growing forage as well as wild tobacco (*Nicotiana*), poison hemlock (*Conium*), and skunk cabbage (*Veratrum*).
- Circumstantial evidence.



25 UC Davis Env Tox Newsletter
23 November 5, 1981

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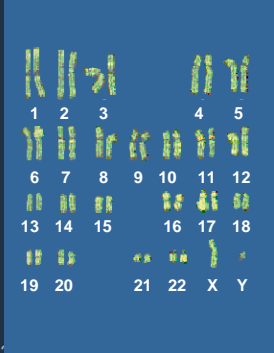
Mutagenesis

- Somatic cell mutations → metabolic dysfunction; carcinogenesis.
- Germ cell mutation → heritable change.
- Point mutation.
 - Base substitution (including analogues).
 - Frame shift.
- Chromosomal aberration.
 - Structural anomaly.
 - Numerical anomaly.

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Karyotypes

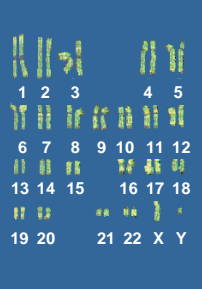


Patterns photographed during metaphase help examine for chromosomal defects.

27

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Abberations



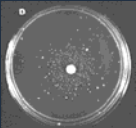
- Aneuploidy/Polyploidy
- Loss or gain of complete chromosomes
 - Microscopically visible
 - Down's Syndrome - (47,21+) trisomy
 - Klinefelter's Syndrome (47,XXY) trisomy
 - Turner's Syndrome (45,XO) monosomy

28

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Ames Test for Chemical Mutagenicity

- Salmonella bacteria strain with histidine coding defect.
- Mutagenic chemicals can change the defect to allow cell division and growth.
- Add salmonella + test chemical + rat hepatocytes (for biotransformation).
 - Growth indicates mutagenic effect.



29

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Carcinogenesis

Multi-step, multi-factorial disease

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    graph TD
      A[Procarcinogen (nonreactive)] --> B[Carcinogen]
      B --> C[Carcinogen + DNA]
      C --> D[Mutation]
      C --> E[Mutation/Initiation]
      D --> F[Cancer]
      E --> G[Promoting agent]
      G --> H[Promotion]
      H --> I[Cancer]
  
```

30

Cancer Definitions

- **Cancer.** NLM
 - A malignant tumor that has the ability to metastasize or invade into surrounding tissues.
- **Tumor (Neoplasm).**
 - A general term for the uncontrolled growth of cells that becomes progressively worse with time.
- **Neoplasia.**
 - The growth of new tissue with abnormal and unregulated cell proliferation.

31

Cancer Definitions, 2

- **Benign tumor.**
 - A tumor that does not metastasize.
- **Metastasis.**
 - Ability to establish secondary tumor growth at a new location.

NLM
32

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Cancer Definitions, 3

- **Carcinoma.** NLM
 - Malignant tumor arising in the epithelium.
 - Most common form of cancer.
 - Usually spreads in the lymphatic system.
- **Sarcoma.**
 - Malignant tumor in muscle or connective tissue.
 - Usually spread in the blood stream.
 - Frequently metastasizes to the lung.

33

Multistage Carcinogenesis: Initiation

- Chemical-virus-spontaneous causes DNA lesion
- Cell division perpetuates DNA lesion
- **No outcome if not promoted**
 - Some chemicals can initiate and promote
 - May remain indefinitely if not promoted
- **One hit**
 - No threshold; irreversible



34

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Properties of Initiated Cells

- No phenotypic differences
- Excess/deficiency of enzymes
 - e.g. δ -GT, G-6-P, Fe exclusion, ATPase
- Resistance to cytotoxic chemicals
 - Faster or slower metabolism
- Impaired cellular communication
- Enhanced response to growth factors
- Resistance to terminal differentiation

35

Multistage Carcinogenesis: Promotion

- Change in micro-environment of cells
- Chemical, viral, spontaneous-induced clonal proliferation of initiated cells
- Growth control factors; receptors; immune function; endocrine control; communication; metabolic; apoptosis
- Multi-hit, high dose
 - Reversible
 - Threshold

36

Multistage Carcinogenesis: Progression

- Complete loss of growth control
- Karyotype instability
- Loss/gain of chromosomal fragments
- DNA demethylation/deregulation
- Gene amplification
- Error prone DNA repair
- Irreversible
- Same mechanisms as promotion

37

Classification of Carcinogens

- **Genotoxic.**
 - Act directly on DNA or expression of DNA during translation.
 - DNA replication errors.
 - Point mutations.
 - Chromosomal aberration.
- **Epigenetic.**
 - Non-DNA reactive.
 - Potentiators.
 - Cell, hormone, immune function modifiers.

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38

Genotoxic Carcinogens

- Chemical capable of producing cancer by directly altering the genetic material of target cells.
- Direct carcinogens (no metabolic activation).
 - Alkylating agents.
- Indirect carcinogens (metabolic activation).
 - Polycyclic aromatic hydrocarbons.
 - Aromatic amines.
 - Nitrosamines.
 - Natural substances.
 - Mycotoxins.
 - Inorganic carcinogens.
 - Ni, Cr, Cd, As.

Marquardt

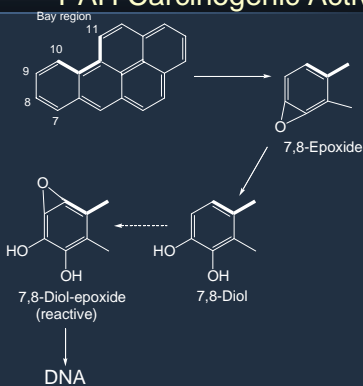
Epigenetic Carcinogens

- **Cytotoxic carcinogens.**
 - Nitrotriacetate, BHA, BHT.
- **Tumor promoters.**
 - DDT, Dioxin
- **Hormones.**
 - Estradiol, DES
- **Immunosuppressants.**
 - Cyclosporin A
- **Particulates.**
 - Asbestos.

Marquardt

40

PAH Carcinogenic Activation



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41

Proven Human Carcinogens

- **Chemicals.**
 - Aflatoxins, 4-aminobiphenyl, As, benzene, benzidine, Be, bis-chloroethylether, Cd, Cr(VI), soot, mineral oils, mustard gas, 2-naphthylamine, Ni, vinyl chloride.
- **Substance abuse.**
 - Alcohol, betel nuts, cigarettes.
- **Dust and fiber.**
 - Asbestos, silica, soots, talcum, wood dust.
- **Chronic infection.**
 - *H pylori*, Hepatitis B/C, HIV, liverfluke, papilloma virus, schistosomes.

Marquardt

42

Initiator Chemicals in Food

- Most genotoxic chemicals
- PAHs
- Aromatic amines
- Heterocyclic amines
- Mycotoxins
- Nitrosamines
- Nitrosamides

43

Promoting Agents in Food

- Butylated hydroxy toluene (BHT)
- Saccharin
- Cholic acid
- Tetrachloro-dibenzo-dioxin (TCDD)
- Alcohol

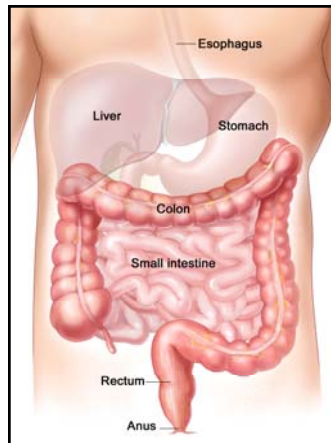
44

Chemical Cancer Assessment

Group A	Human carcinogen	Sufficient human evidence	NLM
Group B1	Probable human	Limited human evidence	
Group B2	Probable human	Inadequate human evidence	
Group C	Possible human	Limited animal evidence	
Group D	Not classifiable	Inadequate animal evidence	
Group F	No evidence	2 animal tests or epidem. and animal	

45

Colorectal Cancer



Colon Polyps

Risk Factors

- Over 50
- Previous occurrence
- Family history
- High fat/low fiber diet
- Smoking
- Alcohol consumption
- Sedentary
- Over weight

National Cancer Institute

Stages of Colorectal Cancer

Spread to other organs

Normal Blood vessel

Lymph node

Normal

Serosa

Muscle layers

Submucosa

Mucosa

National Cancer Institute


48

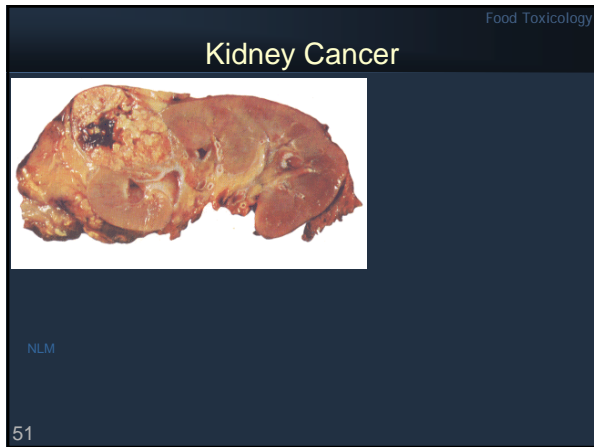
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Common Cancers

Men		Women		
Prostate	32%	Breast	32%	NLM
Lung	16%	Lung	13%	
Colorectal	12%	Colorectal	13%	
Urinary tract	9%	Uterus	8%	
Leukemia/ lymphoma	7%	Leukemia/ lymphoma	6%	

49

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- ### Causes of Cancer
- Diet, 35%
 - Tobacco, 30%
 - Sexual behavior, 7%
 - Alcohol, 3%
 - Infection, 10%
 - Occupational exposure, 4%
 - UV/radiation, 3%
 - Pollution, 2%
- 
- 50 NLM



- Food Toxicology
- ### Acrylamide in Food
- In April 2002, the Swedish National Food Administration released results of its analysis of acrylamide, showing elevated levels in potato chips, French fries, cereals, biscuits, and other starchy foods that are fried or baked.
 - Acrylamide appears to form as a result of a reaction between specific amino acids and sugars found in foods when heated above 100 °C.
 - Listed as a probable human carcinogen.
 - No epidemiological link with increased colorectal, bladder, and kidney cancer with total dietary acrylamide intake in humans.
- 
- 54 Tom Reynolds, FOCUS 2003