

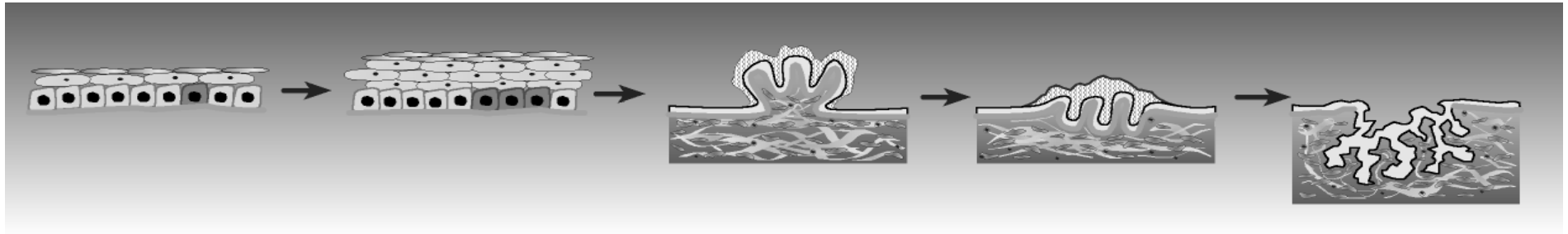
Chemical Carcinogenesis

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MD ANDERSON
CANCER CENTER

Review



**Cell DNA
damage**

**Proliferation
of damaged
cell**

**Inappropriate
cell
signaling/cell
cycling**

**Invasion of
surrounding
tissue**

**Metastasis to
other organs**

Historical Observations

- ⌚ Percival Pott (1775) Increased incidence of scrotal cancer in chimney sweeps.
- ⌚ Yamagiwa and Ichikawa (1918) Multiple topical applications of coal tar to rabbit ears produced skin carcinomas.
 - First demonstration that a chemical could produce cancer in animals.
 - Confirmed Pott's observations
- ⌚ Kennaway (1930's) Isolated benzo[a]pyrene from coal tar.

Causes of Cancer

- ⌞ **Genetics-dramatic examples but few cancers are due solely to genetic causes**
- ⌞ **Environment-includes anything not controlled genetically**
 - i.e. chemicals, habits, diet, occupation
- ⌞ **Gene X Environment Interaction**
 - Individual susceptibility to environmental causes may be genetically determined

Carcinogen-basically, an agent that causes cancer

- ⌞ **Increase over normal tumor incidence or earlier onset**

Types of Human Carcinogens

☞ Physical Carcinogens

☞ UV exposure, Ionizing radiation

☞ Biological carcinogens

☞ Viruses - HIV, HPV, EBV

☞ Bacteria - *H. pylori*

☞ Parasites - liver flukes, *Schistosoma*

☞ Chemical carcinogens

Chemical Carcinogens

☞ Organic

Examples:

- ☞ Polyaromatic hydrocarbons (PAHs)-tobacco smoke, combustion products/coal tar
- ☞ Nitrosamines-tobacco, dietary component
- ☞ Aflatoxin B1--produced by *Aspergillus flavus*

☞ Inorganic

- ☞ i.e. Arsenic, cadmium, chromium, beryllium, lead

☞ Particles

- ☞ i.e. Asbestos

Chemicals with Proven Carcinogenic Activity in Humans

Exposure	Degree of Human	Evidence Animal
4-Aminobiphenyl	S	S
Analgesic mixtures containing phenacetin	S	L
Arsenic and certain arsenic compounds	S	I
Asbestos	S	S
Azathioprine	S	L
Benzene	S	L
Benzidine	S	S
Betal quid containing tobacco (chewing)	S	L
N,N'-Bis-(2-chloroethyl)-2-naphthylamine	S	L
Bis(chloromethyl)ether	S	S
1,3-Butanediol dimethylsulphonate (myleran)	S	L
Chemotherapy for lymphomas (including MOPP, procarbazine, nitrogen mustard, vincristine and prednisone)	S	ND
Chlorambucil	S	S

Chemicals with Proven Carcinogenic Activity in Humans

Chromium and certain chromium compounds	S	S
Coal tars	S	S
Coal tar pitches	S	S
Conjugated estrogens	S	I
Cyclophosphamide	S	S
Diethylstilbestrol	S	S
Melphalan	S	S
Methoxsalen with UVA therapy (PUVA)	S	S
Mineral oils	S	S
Mustard gas	S	L
2-Naphthylamine	S	S
Shale oils	S	S
Smokeless tobacco products (oral use)	S	I
Soots and soot extracts	S	S
Tobacco smoke	S	S
Treosulphan	S	ND
Vinyl chloride	S	S

Chemicals with Proven Carcinogenic Activity in Humans

Industrial processes and occupational exposures

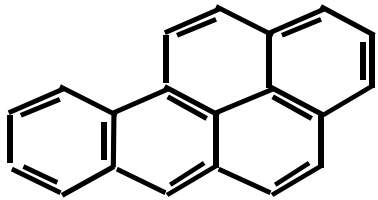
Auramine manufacturing	S
Boot and shoe manufacturing and repair	S
Coal gasification (older processes)	S
Coke production	S
Furniture manufacturing (wood dusts)	S
Isopropyl alcohol mfg (strong acid process)	S
Nickel refining	S
Rubber industry (certain occupations)	S
Underground hematite mining (radon)	S

S, sufficient; L, limited; I, inadequate; ND, no data

Source: IARC Monographs

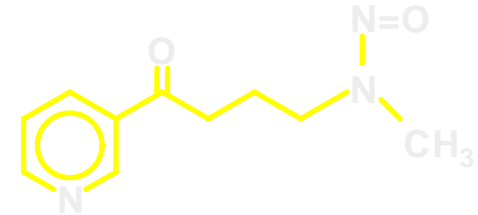
Structures of some carcinogens

Polycyclic aromatic hydrocarbons (PAHs)



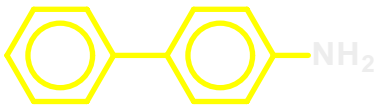
Benzo[a]pyrene

N-Nitrosamines



N-Nitrosodiethylamine 4-(Methylnitrosamino)-1-(3-pyridyl)-1-butanone (NNK)

Aromatic amines

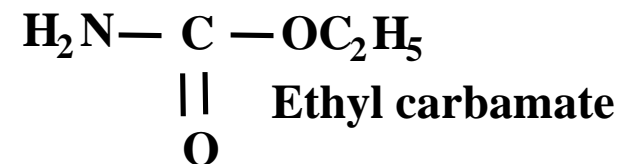
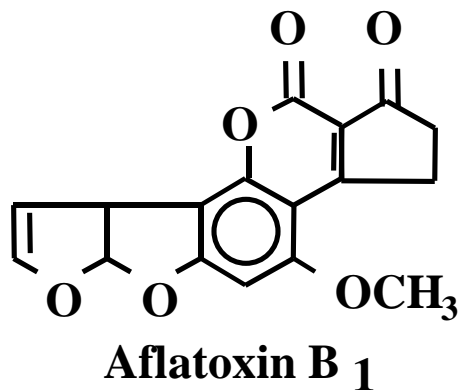
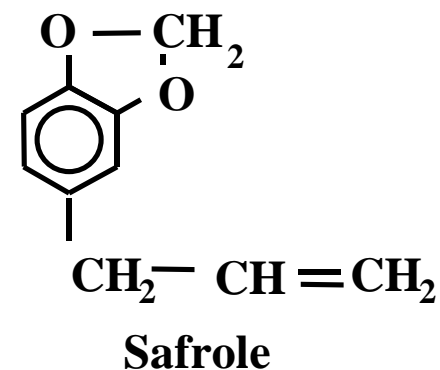
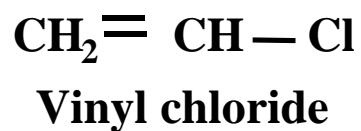
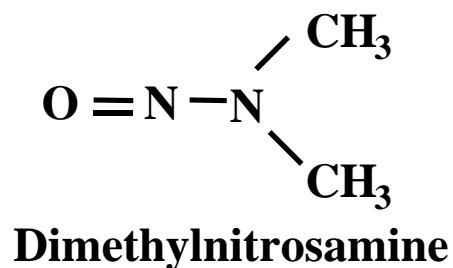


4-Aminobiphenyl

Free radicals

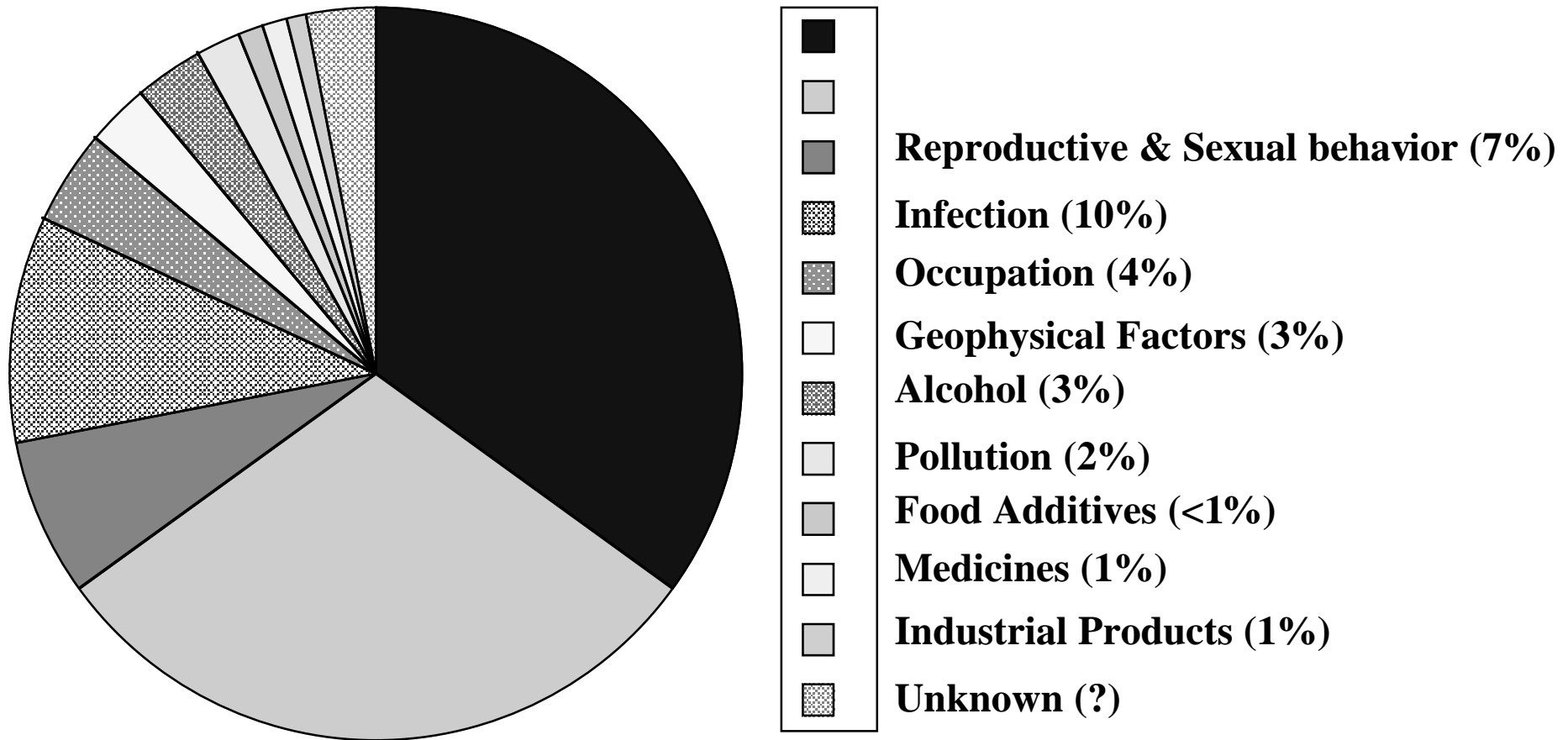
(- O₂⁻, H₂O₂, OH·)

Structures of Some Chemical Carcinogens



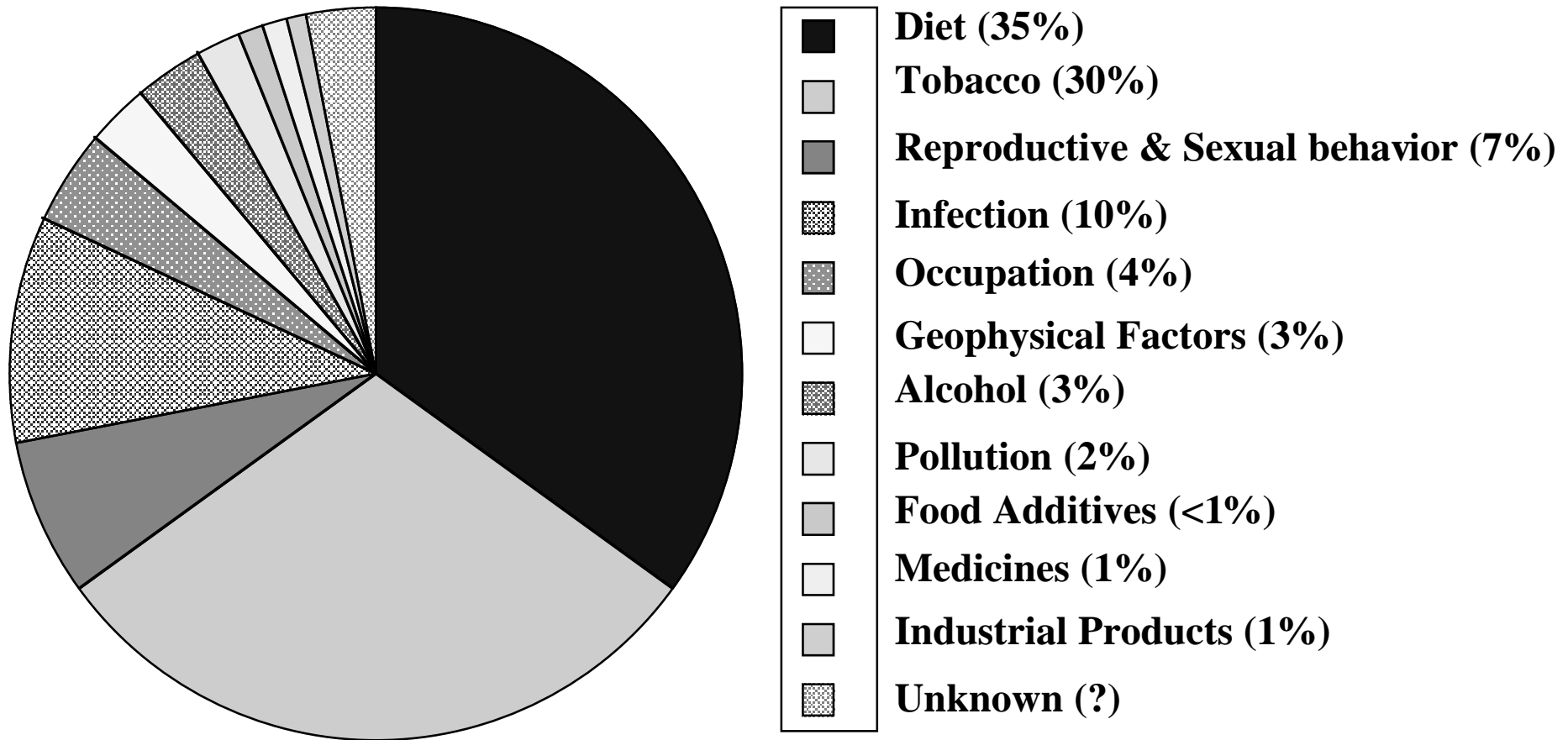
>3,000 chemicals have been shown to cause cancer in laboratory animals, may be suspect human carcinogens

Factors Contributing to the Incidence of Cancer Deaths



Adapted from Doll, R. and Peto, R.(1981) *The Causes of Cancer*. Oxford Medical Publications.

Factors Contributing to the Incidence of Human Cancer

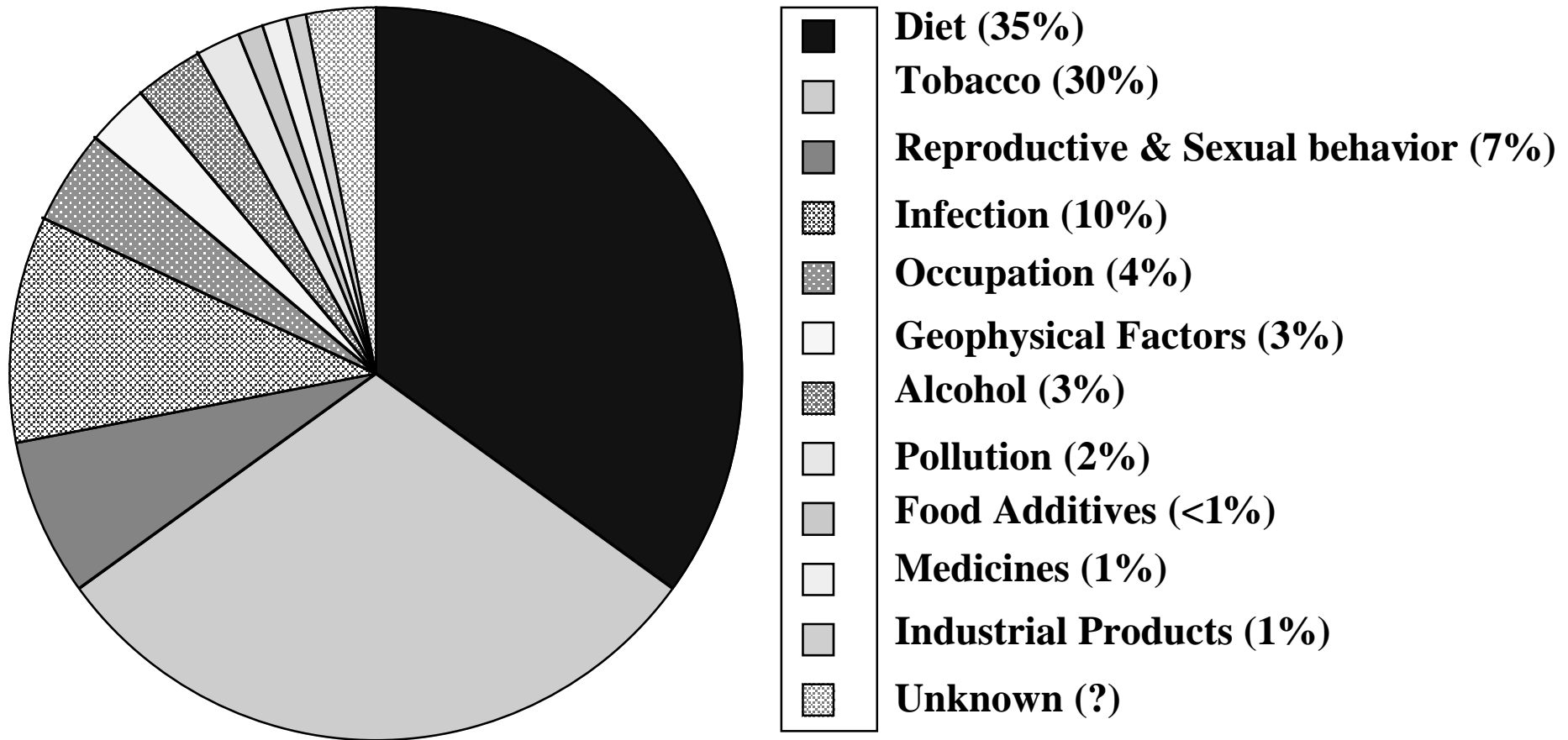


Adapted from Doll, R. and Peto, R.(1981) *The Causes of Cancer*. Oxford Medical Publications.

Summary of Carcinogens in Cigarette Smoke

Carcinogen Type	No. of Compounds
Polycyclic aromatic hydrocarbons	10
Aza-arenes	3
N-Nitrosamines	7
Aromatic amines	3
Heterocyclic aromatic amines	8
Aldehydes	2
Miscellaneous organic compounds	15
Inorganic compounds	7
Total	55

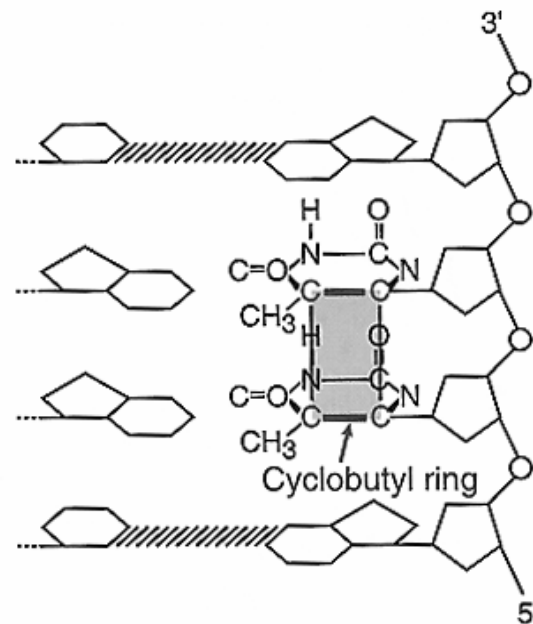
Factors Contributing to the Incidence of Human Cancer



Adapted from Doll, R. and Peto, R.(1981) *The Causes of Cancer*. Oxford Medical Publications.

Geophysical Factors: Sun and UV Exposure

Cyclobutane pyrimidine dimer (CPD)

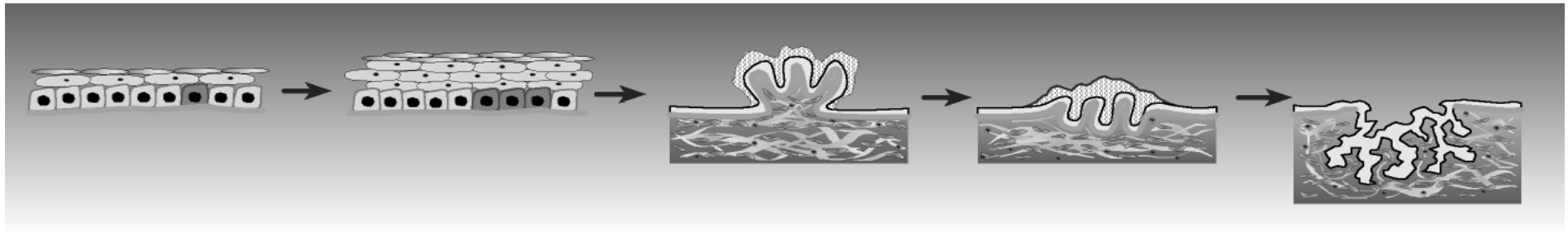


mouse skin model

INITIATION

PROMOTION

PROGRESSION



1. Covalent binding of carcinogen to DNA, cell replication, and fixation of mutation.
2. Mutation induction in critical target genes of stem cells, e.g. H-ras
3. Phenotypically "normal" epidermis

1. Expansion of initiated stem cells through epigenetic mechanisms
2. Altered gene expression/ enzyme activities
3. Angiogenesis

1. Production and maintenance of chronic cell proliferation
2. Development of clonal outgrowths; benign papillomas
3. Altered differentiation
4. Diploid stem line

1. Additional genetic events occurring stochastically
2. Aneuploidy e.g. nonrandom trisomies of chromosomes 6 & 7
3. LOH
4. Further alteration in differentiation
5. Dysplasia

1. Invasion
2. Metastasis
3. Loss of tumor suppressor activity e.g. p53 mutation
4. Gene amplification e.g. mutated Hras allele

GENETIC SUSCEPTIBILITY

Characteristics of Tumor Initiators and Promoters

∪ Initiators (genetic)

- ∪ Chemically reactive or require metabolic activation to chemically reactive intermediates.
- ∪ React covalently with cellular macromolecules such as DNA, RNA, protein.
- ∪ Produce an essentially irreversible event after a single application.
- ∪ Mutagenic

∪ Promoters (epigenetic)

- ∪ Do not require metabolism, i.e., are active in their parent form.
- ∪ Require prolonged and repeated exposure.
- ∪ Actions are essentially reversible.
- ∪ May alter gene expression

Chemical Carcinogenesis in Experimental Animals

Complete Carcinogenesis:

-exposure to a single agent either repeatedly or as a large single dose with the development of tumors in a particular organ.

Multistage Carcinogenesis:

-exposure to a subcarcinogenic or “initiating” dose of one agent followed by continued exposure to a non-carcinogenic agent that is co-carcinogenic (usually referred to as a tumor promoting agent).

Initiation and the Mutational Theory of Carcinogenesis

- ⌞ Agents that damage DNA are often carcinogenic.
- ⌞ Most carcinogenic agents are mutagens in *in vitro systems*
- ⌞ Individuals with DNA repair deficiencies have higher incidences of cancer
 - Xeroderma pigmentosum (XP)
 - Ataxia telangiectasia (AT)
 - Fanconi's anemia (FA)
 - Bloom's syndrome (BS)

4-7 mutations in key genes are usually necessary to produce most human cancers

Carcinogen-DNA Adducts, Mutations and Tumor Initiation

Species	Tumor	Carcinogen	Mutation	Oncogene
Human	Lung cancer	B[a]P	G --> T	<i>p53</i>
		NNK	G --> A	<i>p53</i>
Human	Hepatocellular carcinoma	AFB	G --> A	<i>p53</i>
Human	Skin cancer	UV-B	CC --> TT	<i>p53</i>
			C --> T	<i>p53</i>
Rat	Mammary carcinoma	MNU	G ³⁵ --> A	H-ras (12)
		DMBA	A ¹⁸² --> T	H-ras (61)
Mouse	Skin carcinoma	DMBA	A ¹⁸² --> T	H- <i>ras</i>
		MNNG	G ³⁵ --> A	

Overview of Mutations

- **Mutation**

A heritable change in the sequence of an organism's genome
Caused by a “mutagen”

- **Types of mutations**

- Base-pair substitutions (A-->G or G--->C)

- Frame-shift mutations

- Small deletions

- Small insertions

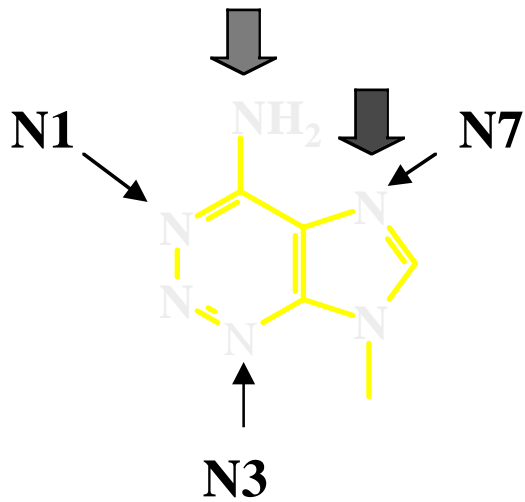
- Small inversions

- **Also, gross structural changes to the chromosome**

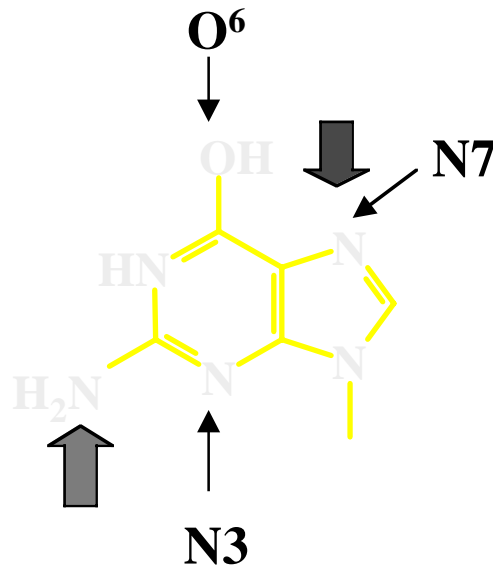
- Deletions, duplications, inversions, translocations, aneuploidy, polyploidy

Sites of DNA Adduct Formation

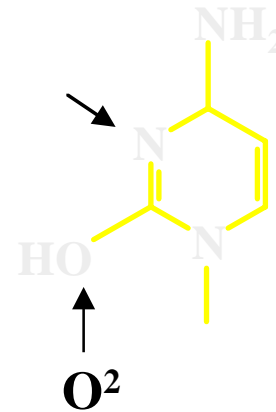
Adenine



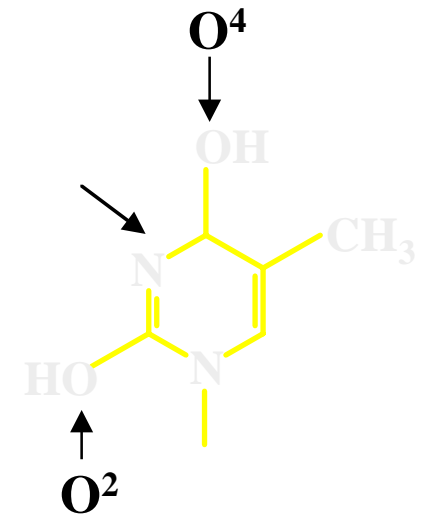
Guanine



Cytosine



Thymine



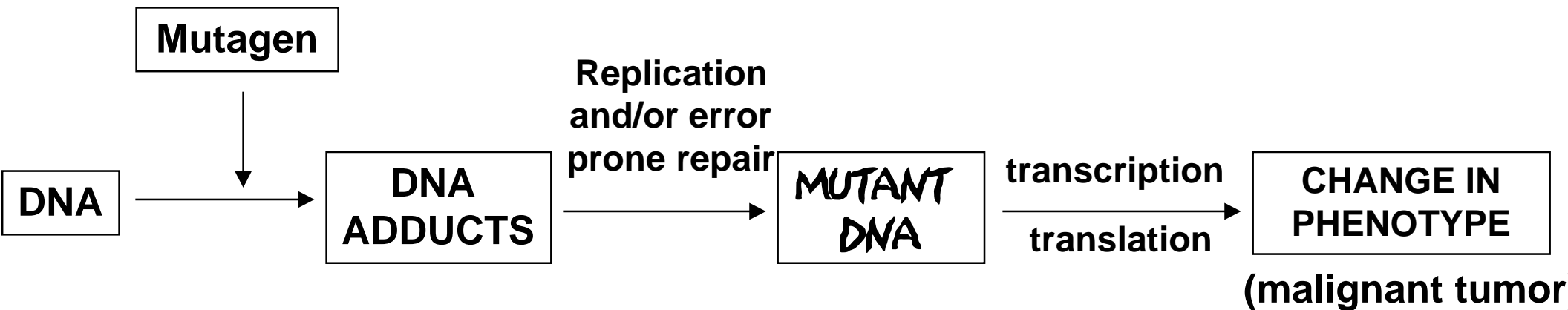
↓ PAHs (e.g., B[a]P, DMBA)

↓ Mycotoxins (e.g., AFB₁)

Molecular effects of substitution mutations

Location of mutation	Effect of mutation
1. Coding regions	Mutant protein : no detectable effect, partial loss, gain of function, an alteration of function, change protein stability, complete loss of function. Premature termination: STOP codons
2. Promoter	Reduced or increased gene expression
3. 3' of protein-coding region	Defective transcription termination or alteration of mRNA stability
4. Introns	Defective mRNA splicing
5. Origin of replication	Defect in initiation of DNA replication

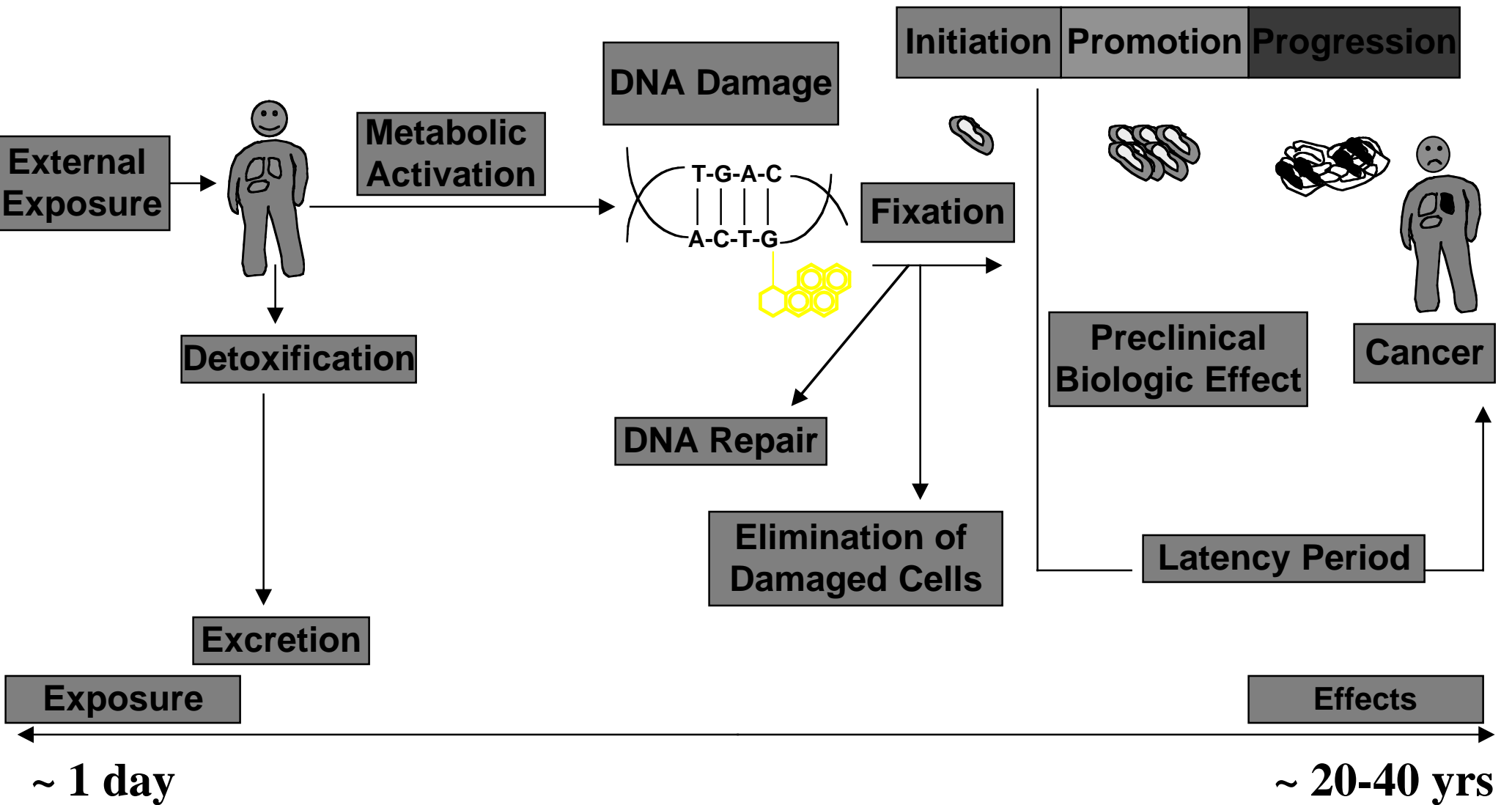
Classical Model of Carcinogenesis Based on Exposure to a Genotoxic Carcinogen



Change in Phenotype

- ⌞ Inappropriate cell division
- ⌞ Or lack of apoptosis
- ⌞ Loss of 2 copies of tumor suppressor
- ⌞ Activation of 1 copy of proto-oncogene

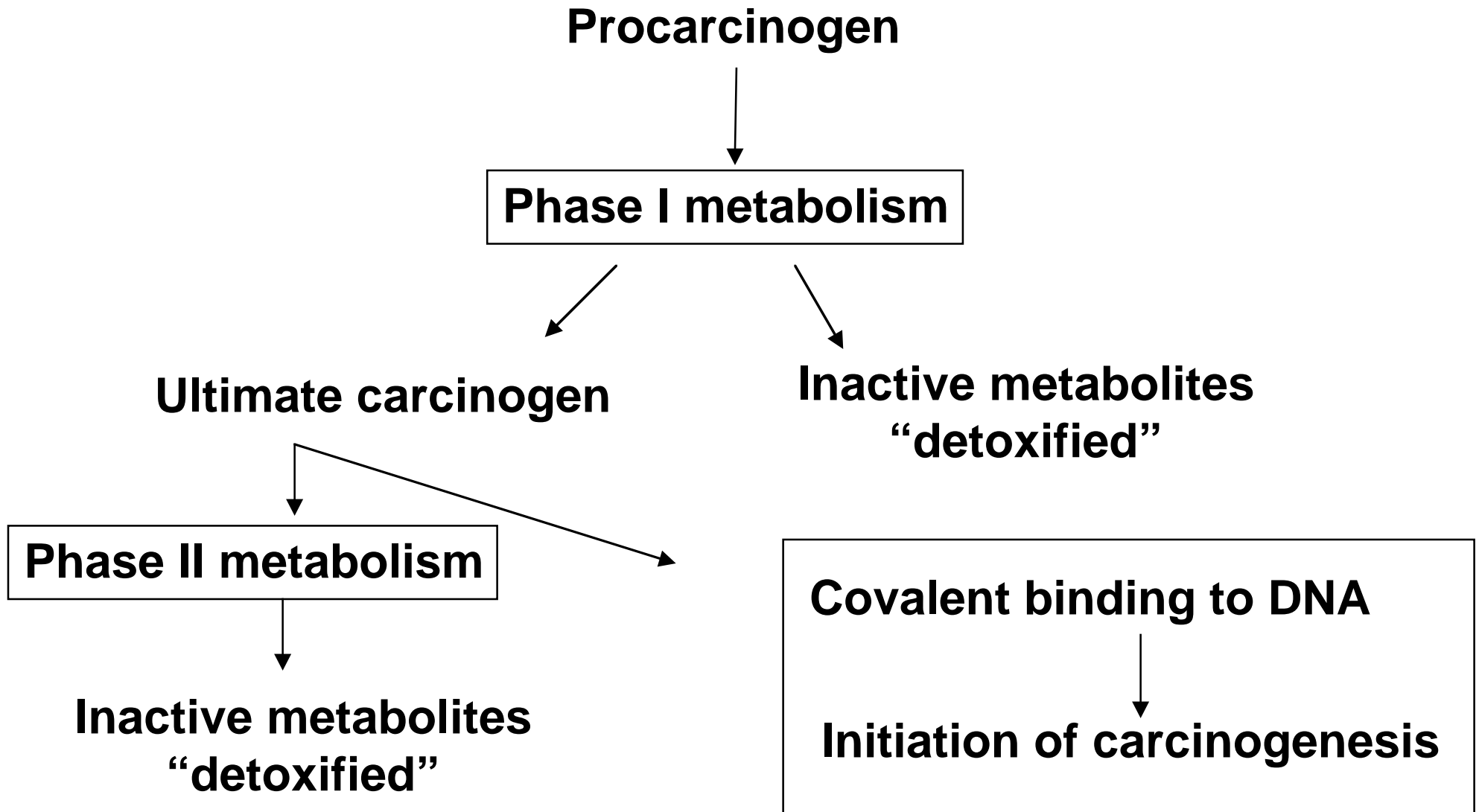
Overview of Chemical Carcinogenesis



Biotransformation

- 〉 **Once absorbed, chemicals are subject to metabolism by biotransformation enzymes**
 - **Phase I --often oxidative reaction**
 - **Phase II --often conjugation reaction**

- 〉 **Biotransformation may lead to activation, detoxification, or (if multiple enzymes are involved) both**



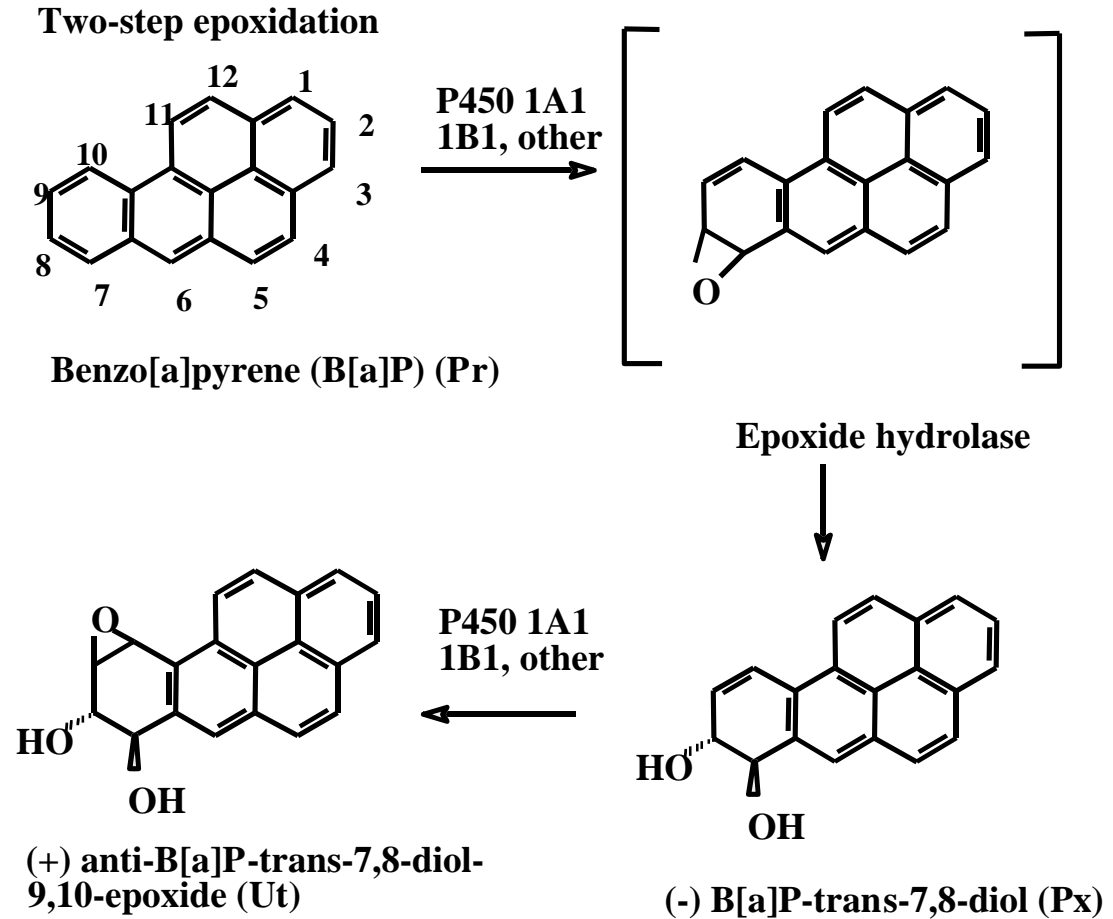
Phase I metabolism can either activate or detoxify. Phase II metabolism makes the metabolite more polar so it can be excreted, and usually detoxifies (with some exceptions).

Phase I Biotransformation

2 Examples:

-Cytochrome P450 enzymes-oxidation

-Epoxide hydrolases--hydrolysis of reactive epoxides



Phase II Metabolism

Examples:

Sulfate conjugation

sulfotransferases

Glutathione conjugation

Glutathione *S*-transferase (GST)

Amino acid conjugation

Acyl-CoA-synthetase

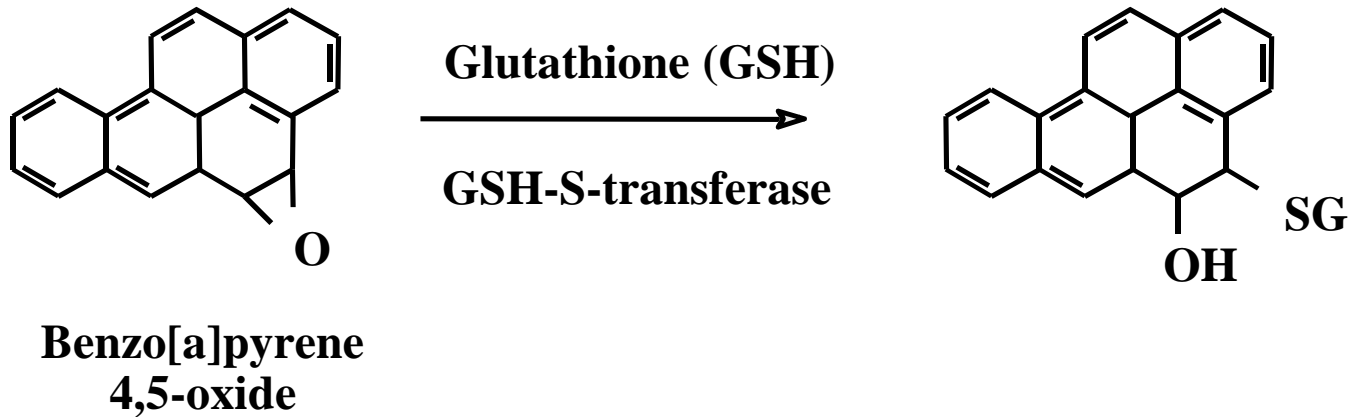
Acetylation

N-acetyltransferase (NAT)

Methylation

Catechol-*O*-methyltransferase
(COMT)

Usually elimination (detoxification) reactions



-this reaction prevents formation of the ultimate carcinogen

Metabolic Activation of Cigarette Smoke Carcinogens

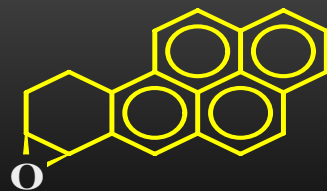
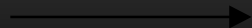
Parent Carcinogen

DNA Reactive Intermediate(s)

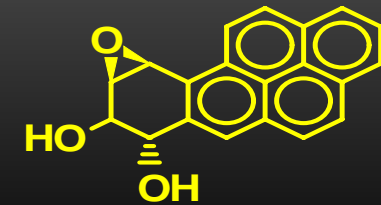
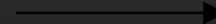
Polycyclic Aromatic Hydrocarbons (PAHs)



Benzo[a]pyrene



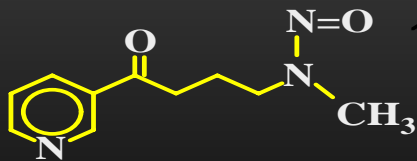
Benzo[a]pyrene-7,8-diol



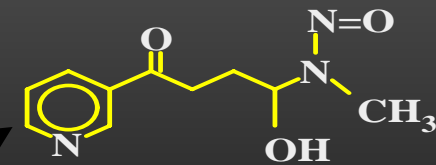
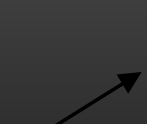
Benzo[a]pyrene-7,8-diol-9,10-epoxide

DNA

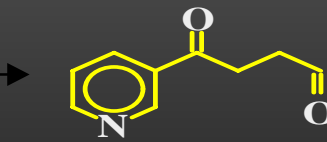
Nitrosamines



4-(Methylnitrosamino)-1-(3-pyridyl)-1-butanone (NNK)



α -methylene hydroxylation

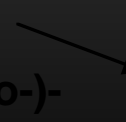


4-oxo-4-(3-pyridyl)-butanal

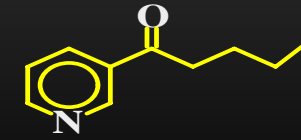


$[\text{CH}_3\text{N}=\text{NOH}]$
methanediazo-
hydroxide

DNA



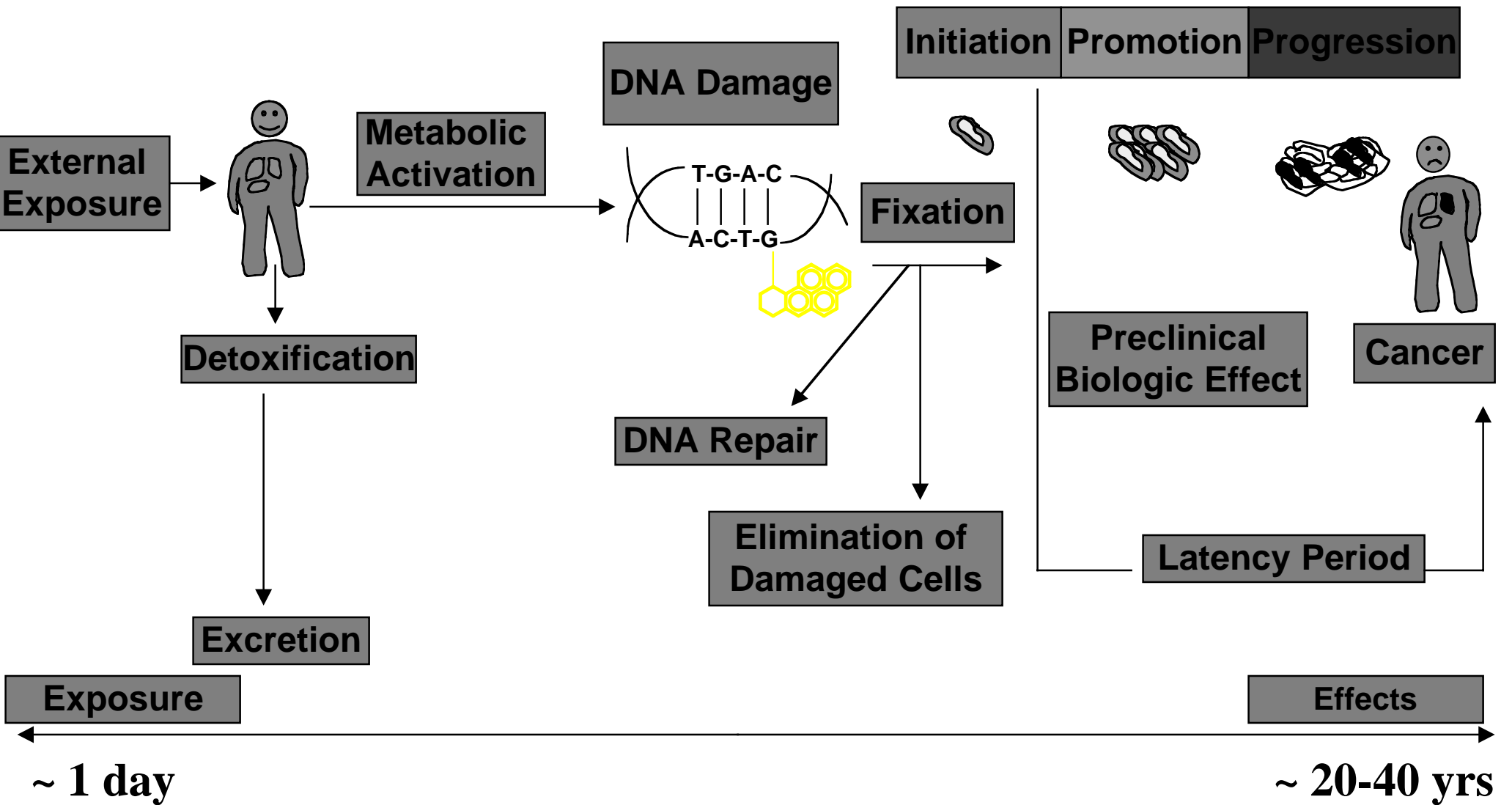
α -methyl hydroxylation



4-(3-pyridyl)-4-oxobutane-diazohydroxide

DNA

Overview of Chemical Carcinogenesis



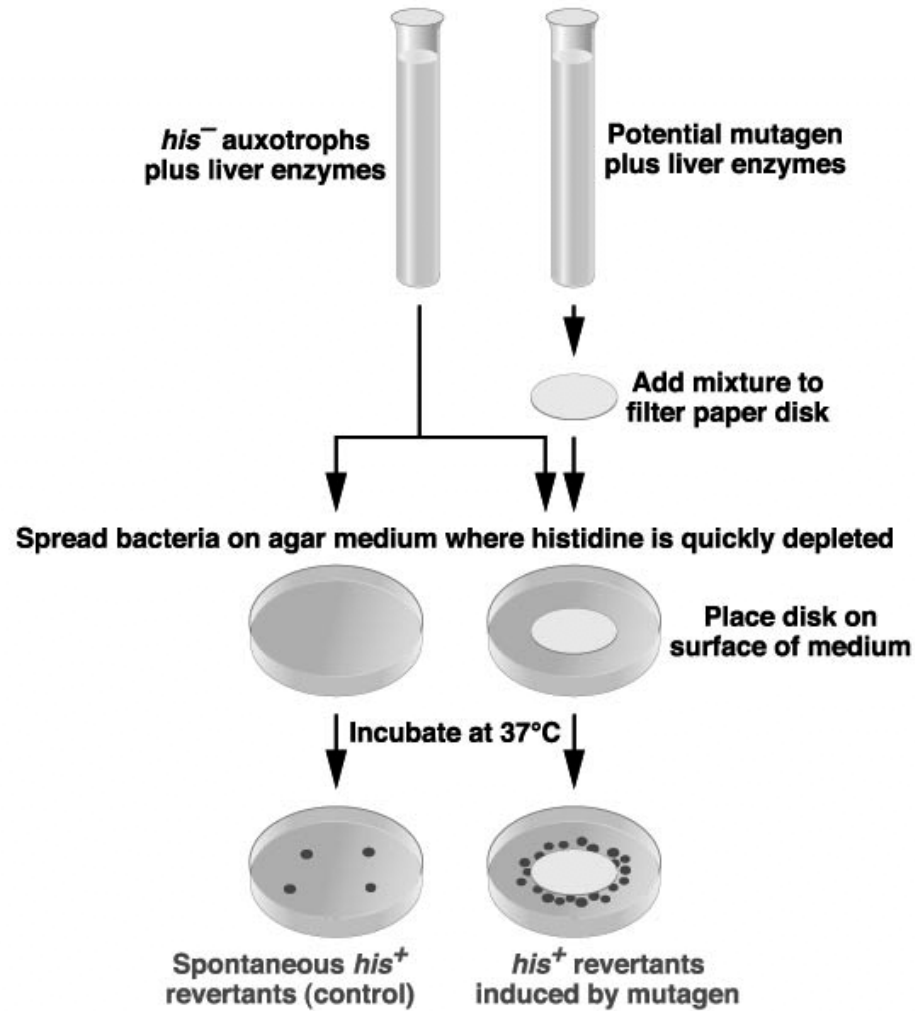
Examples of Metabolic Susceptibility genes

Gene	Metabolic pathway	Cancer sites
<i>CYP1A1</i>	Metabolism of polycyclic aromatic hydrocarbons, TCDD and estrogens	Lung, stomach, colon, breast
<i>CYP2E1</i>	Oxidation of N-nitrosamines, alcohol	Lung, bladder, colon
<i>CYP2D6</i>	Hydroxylation of lipophilic xenobiotics, possibly NNK	Lung, bladder, breast
<i>GSTM1</i>	Conjugation of organic epoxides with reduced glutathione	Lung, bladder, colon, stomach, breast, liver
<i>NAT2</i>	N-acetylation of arylamines and N-hydroxylated heterocyclic amines	Bladder, lung, colorectal, breast

How are carcinogenes identified?

- u Unfortunately, many examples by epidemiological association
- u Laboratory animals
 - Administer MTD over majority of life span
- u *In vitro* assays
 - Ex. Ames mutagenicity assay

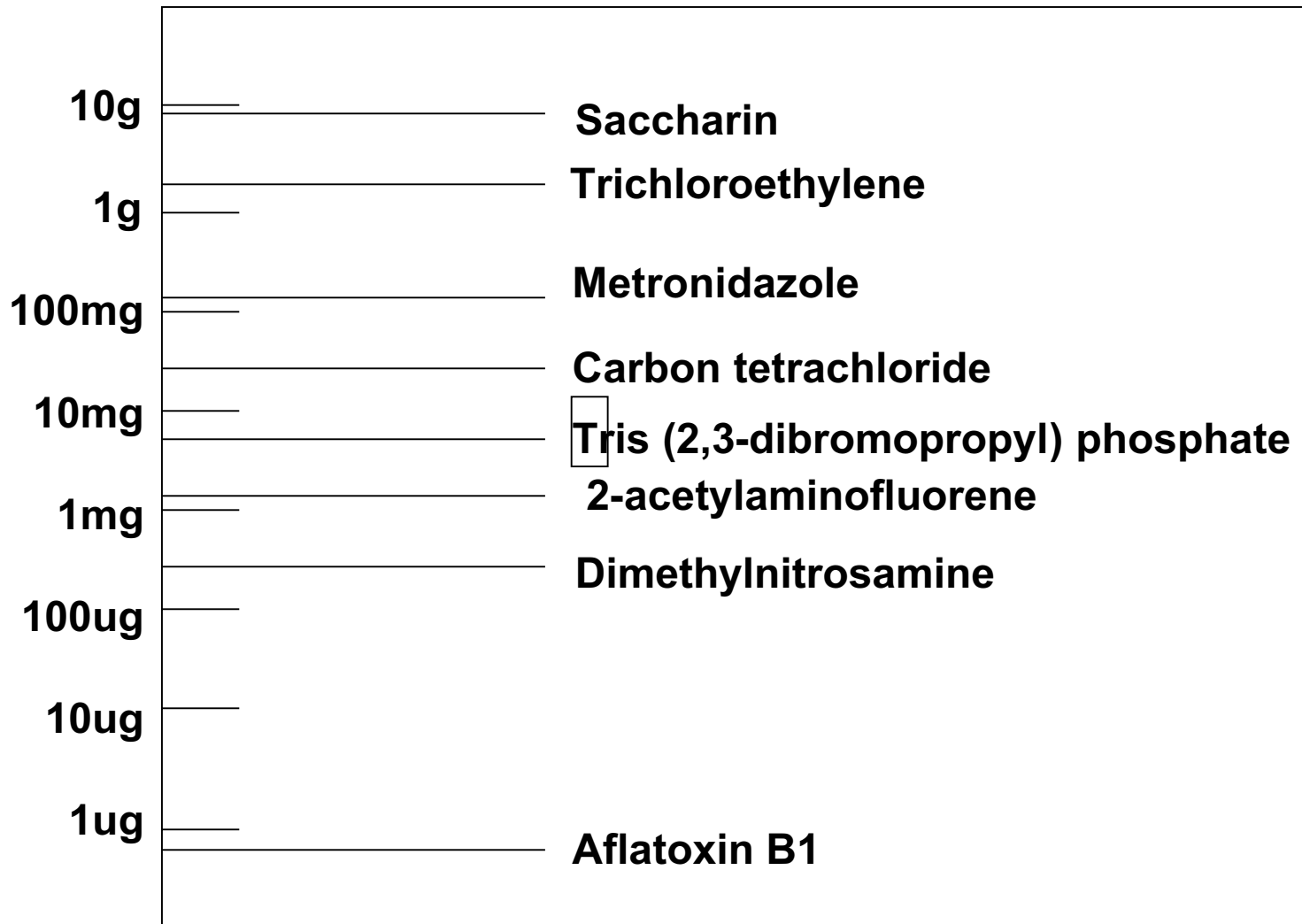
Ames Assay



Potency

- ⌞ Amount required to produce tumors when administered daily for a lifetime
- ⌞ Even the most potent carcinogens usually require repeated, long term administration to produce cancer

Relative Carcinogenic potency



Daily dose to give 50% of animals cancer in “life time ” (per kg body weight)

Reducing the rates of cancer

- ⤵ **Ames and Gold have suggested that traces of synthetic chemicals are not major contributors to cancer incidence**

- ⤵ **Further suggest that factors most likely to have effect on reducing cancer rate include:**
 - **Reduction of smoking**
 - **Increased consumption of fruits and vegetables**
 - **Control of infections**
 - **Avoidance of intense sun exposure**
 - **Increased physical activity**
 - **Reduced consumption of alcohol and possibly red meat**

*adapted from Ames and Gold, Environ Health Perspect 1997; 105:865-73.