Chapter 11
Mutation:
The Source of Genetic Variation
Somatic mutations

Germline mutations
Chromosomal mutations

Gene mutations
DNA

Normal or WT gene

Normal protein

WT phenotype

Mutated gene

Mutant protein

Mutant phenotype
“...the possibility that genes were...subject to the hurly-burly of both insult and clumsy efforts to reverse the insults, were unthinkable.”

Frank Stahl
Mutations

• Heritable changes in the nucleotide sequence or chromosome

• Mutations may be:
  – Spontaneous as a result of errors in DNA replication or
  – Induced by exposure to radiation, chemicals, viruses, or other mutagenic agents
Detecting Original Mutations

- Dominant mutations are the easiest to detect
- Can be identified by pedigree analysis
- X-linked mutations can sometimes be identified by an examination of male progeny
- If the mutation is autosomal recessive, it is extremely difficult to identify the original mutant individual
A Dominant Trait: Foot Blistering

The mutation first appeared in II-5
An X-linked Mutation: Hemophilia

Fig. 11.2
Spontaneous Mutation Rates

• Studies suggest that mutations are rare
• 1/1,000,000 copies of a human gene
• Impact on the population of mutation is less severe because
  – Nature of genetic code
  – Recessive mutations are not expressed in the heterozygotes
  – Lower reproductive success or early death associated with many mutations
Rates

- Mutation rate = the number of mutated alleles per gene per generation
- For accurate measurement, the mutant phenotype must be
  - Never produced by recessive alleles
  - Fully expressed
  - With clear paternity
  - Never produced by nongenetic agents
  - Produced by the dominant alleles of only one gene
<table>
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<tr>
<th>Trait</th>
<th>Mutants /Million</th>
<th>Mutation Rate</th>
<th>OMIM Number</th>
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<td>Gametes</td>
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<td>$1.4 \times 10^{-5}$</td>
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<td>180200</td>
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<td>Osteogenesis imperfecta</td>
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<td>Neurofibromatosis</td>
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<tr>
<td>Duchenne muscular dystrophy</td>
<td>50–100</td>
<td>$0.5–1 \times 10^{-4}$</td>
<td>310200</td>
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Mutation Rates Vary Between Genes

Factors that influence mutation rates

• Size of gene (increased risk for mutation in large genes)

• Nucleotide sequence - presence of nucleotide repeats may increase risk of mutation

• Spontaneous chemical change

• Genes rich in G/C pairs have increased risk
Trinucleotide Repeats

• Class of mutations associated with a number of genetic disorders
• Caused by an expansion of nucleotide triplets
• Process is **allelic expansion** when the gene size is increased by an increase in trinucleotide repeats
Allelic Expansion in the $FMR-1$ Gene at the Fragile-X Locus

Fig. 11.13
Fragile-X Syndrome

- Approximately 1% of males institutionalized for mental retardation have fragile-X syndrome
- Heterozygous females have a normal phenotype
- In 20–50% of all cases, the mutant allele has a low degree of penetrance in males (transmitter males)
- Daughters of transmitter males have a high risk of producing affected sons
Inheritance of Fragile-X syndrome

Fig. 11.12

Unaffected females

Transmitter male

Affected males

Gene Expansion and Anticipation

- Progressive degeneration of nervous system
- Inherited as an autosomal dominant trait
- All have expanded CAG repeats
- Show correlation between the increasing number of repeats and age at onset
- The appearance of increasing symptoms in succeeding generations is called anticipation
Spontaneous:

**DNA replication errors** - can lead to additions or deletions point mutations

Spontaneous **chemical** changes:

- depurination
- deamination
DNA replication error → Additions and deletions
Spontaneous chemical changes → point mutations
Induced

Radiation - X-Ray ionizing radiation - ds breaks
UV rays - leads to T-T dimers

Chemical mutagens:

- Base analogs
- Base modifiers
- Intercalating agents
Environmental Factors: Radiation

- Process by which energy travels through space
- Exposure to radiation is unavoidable
- Exposure comes from a wide variety of sources both natural and due to human activity

![Pie chart showing sources of radiation exposure to the U.S. population](Fig. 11.5)
Radiation Exposure May Damage Cells

- Causes biological damage at several levels
- Some radiation forms highly reactive ionized molecules that can cause mutations in DNA
- Repair is possible, but if too many mutations form, the system is overwhelmed and cell death or cancer may occur
UV irradiation $\rightarrow$ Thymine dimers
UV Light irradiation Produces Thymine Dimers

They distort the DNA molecule and may cause errors in replication

Fig. 11.15
Effect of Chemicals

Chemicals can cause mutations in a number of ways

- **Base analog** – may change pairing
- **Chemical modification** – mutagens change one base into another
- **Intercalating agents** – alter the shape may cause deletion or addition
Base Analog

A structurally similar chemical bonds to DNA or RNA

Fig. 11.6
Some mutagens attack bases in DNA and change one base into another.

Fig. 11.7
Base modifiers

Original base | Mutagen | Modified base | Pairing partner | Predicted transition
---|---|---|---|---
1) Guanine | Nitrous acid (HNO₂) | Xanthine | Cytosine | None
2) Cytosine | Nitrous acid (HNO₂) | Uracil | Adenine | CG → TA
3) Adenine | Nitrous acid (HNO₂) | Hypoxanthine | Cytosine | AT → GC
Base modifiers

Original base | Mutagen | Modified base | Pairing partner | Predicted transition
---|---|---|---|---
Cytosine | Hydroxylamine (NH₂OH) | Hydroxylaminocytosine | Adenine | CG → TA
Guanine | Methylmethane sulfonate (MMS) (alkylating agent) | O⁶-Methylguanine | Thymine | GC → AT
The Ames test

Test for mutagenicity

Solution of his\(^{-}\) mutant bacteria
Rat liver enzymes
Solution of potential mutagen/carcinogen
Mixture is plated onto medium without histidine
Growth of bacteria his\(^{-}\)→ his\(^{+}\) revertants

Control: no mutagen

Solution of his\(^{-}\) mutant bacteria
Rat liver enzymes
Solution of potential mutagen/carcinogen
Mixture is plated onto medium without histidine
No growth
No his\(^{-}\)→ his\(^{+}\) revertants
Intercalating agents

- **Intercalating agents** insert themselves into the DNA and distort its shape
- Replication of distorted region a cause a deletion or insertion
- Breakdown products of common pesticides are intercalating agents

Acridine Orange

Fig. 11.8
Ethidium bromide used to visualize DNA is an intercalating agent.
Nucleotide Substitutions

- **Missense mutations**
  - Single nucleotide change that changes one amino acid for another

- **Sense mutations**
  - Produce longer or shorter proteins by changing a termination codon into one that codes for an amino acid

- **Nonsense mutations**
  - Change of a codon for an amino acid into a termination codon shortens the protein product
Base-pair substitution mutations

Transition

<table>
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<th>Pur</th>
<th>Other Pur</th>
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<tr>
<td>Pyr</td>
<td>Pyr</td>
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</tbody>
</table>

- A → G
- T → C

Transversion

<table>
<thead>
<tr>
<th>Pur</th>
<th>Pyr</th>
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</thead>
<tbody>
<tr>
<td>Pyr</td>
<td>Pur</td>
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</tbody>
</table>

- A → T
- T → A
Effect on protein encoded

**Missense mutation**

bp mutation in DNA results in change in mRNA codon, so that a different amino acid is inserted at that site in the protein.

<table>
<thead>
<tr>
<th>DNA</th>
<th>AAA</th>
<th>→</th>
<th>GAA</th>
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<tbody>
<tr>
<td>RNA</td>
<td>AAA</td>
<td>→</td>
<td>GAA</td>
</tr>
<tr>
<td>Protein</td>
<td>lysine</td>
<td>→</td>
<td>glutamic acid</td>
</tr>
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</table>

Single nucleotide change in codon 6 (glu to val) of β-globin gene leads to mutant form of hemoglobin and sickle cell anemia.
**Nonsense mutation**

bp change in DNA results in change in mRNA codon to a STOP codon, so that translation is terminated.

<table>
<thead>
<tr>
<th>DNA</th>
<th>AAA → TAA</th>
</tr>
</thead>
<tbody>
<tr>
<td>RNA</td>
<td>AAA → UAA</td>
</tr>
<tr>
<td>Protein</td>
<td>lysine → STOP</td>
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</table>

Truncated polypeptide
**Neutral mutation**

bp change in DNA results in change in mRNA codon, so that an equivalent aa is inserted.

DNA

RNA

Protein

Protein function and/or structure not significantly altered.
**Silent mutation**

bp change in DNA results in change in mRNA codon, so that the SAME aa is inserted.

<table>
<thead>
<tr>
<th>DNA</th>
<th>AAA</th>
<th>→</th>
<th>AAG</th>
</tr>
</thead>
<tbody>
<tr>
<td>RNA</td>
<td>AAA</td>
<td>→</td>
<td>AAG</td>
</tr>
<tr>
<td>Protein</td>
<td>lysine</td>
<td>→</td>
<td>lysine</td>
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</table>

No effect.
Frameshift mutation

bp addition or deletion in DNA results in change in mRNA sequence, so that protein sequence changes.

DNA

RNA

Protein

Change in protein sequence.
Triplet code

WT

THE BIG BOY HIT THE CAT WHO ATE THE FAT RAT

+   

THE BIG BOY HIT TTH ECA TWH OAT ETH EFA TRA T

-   

THE BIG BOH ITT HEC ATW HOA TET HEF ATR AT

+     -   

THE BIG BOY HIT TTH ECA TWH ATE THE FAT RAT

+     +   +   +   

THE BIG BAO YHA ITT HEC CAT WHO ATE THE FAT RAT
Phenotypic level

Morphological

Lethal

Biochemical

Conditional

Resistance
### Types of Mutations

A sentence comprised of three-letter words can provide an analogy to the effect of mutations on a gene's DNA sequence:

<table>
<thead>
<tr>
<th>Type</th>
<th>Normal</th>
<th>Missense</th>
<th>Nonsense</th>
<th>Frameshift</th>
<th>Deletion</th>
<th>Insertion</th>
<th>Duplication</th>
<th>Expanding mutation</th>
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<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>THE ONE BIG FLY HAD ONE RED EYE</td>
</tr>
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<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<td>generation 2</td>
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<td>THE ONE BIG FLY[FLY FLY] HAD ONE RED EYE</td>
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<td></td>
<td>THE ONE BIG FLY[FLY FLY FLY FLY] HAD ONE RED EYE</td>
</tr>
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</table>
Nucleotide Substitutions in the Hemoglobin Gene

There are several hundred variants of the alpha and beta globins with single amino acid substitutions.
Mutations in Cystic Fibrosis Gene

Fig. 11.17
DNA Repair Systems

- Cells have enzyme systems to repair damaged DNA
- There are several categories of repair systems and they function during different parts of the cell cycle
- The repair systems are under genetic control and they too can undergo mutation
# Table 11.4 Rates of DNA Damage in a Mammalian Cell

<table>
<thead>
<tr>
<th>Damage</th>
<th>Events/Hour</th>
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<tbody>
<tr>
<td>Depurination</td>
<td>580</td>
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<tr>
<td>Depyrimidation</td>
<td>29</td>
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<tr>
<td>Deamination of cytosine</td>
<td>8</td>
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<tr>
<td>Single-stranded breaks</td>
<td>2,300</td>
</tr>
<tr>
<td>Single-stranded breaks after depurination</td>
<td>580</td>
</tr>
<tr>
<td>Methylation of guanine</td>
<td>130</td>
</tr>
<tr>
<td>Pyrimidine (thymine) dimers in skin (noon Texas sun)</td>
<td>$5 \times 10^4$</td>
</tr>
<tr>
<td>Single-stranded breaks from background ionizing radiation</td>
<td>$10^{-4}$</td>
</tr>
<tr>
<td>Damage</td>
<td>Repairs / Hour</td>
</tr>
<tr>
<td>--------------------------------</td>
<td>----------------</td>
</tr>
<tr>
<td>Single-stranded breaks</td>
<td>$2 \times 10^5$</td>
</tr>
<tr>
<td>Pyrimidine dimers</td>
<td>$5 \times 10^4$</td>
</tr>
<tr>
<td>Guanine methylation</td>
<td>$10^4$–$10^5$</td>
</tr>
</tbody>
</table>
p53: an important cell cycle gene

In order for DNA repair to occur - cell cycle must slow down

p53 protein is a transcription factor:
When activated it induces transcription of DNA repair genes and genes that slow down the cell cycle
Failure of DNA Repair

- Fewer mutations are corrected
- Increase in mutations in the genome
- The protein p53 monitors repair of DNA
- If damage is too severe, the p53 protein promotes programmed cell death or apoptosis
- Mutations may occur in genes encoding DNA repair proteins
- Lead to overall increase in mutations

- p53 - tumor suppressor gene. Loss of function implicated in multiple cancers
Repair System Disorders

- Xeroderma pigmentosum (XP)
- 1/250,000
- Damage from UV light
- 1000X increase in cancer risk
- Mutations of at least 8 different genes may cause XP
Other Examples of DNA Repair Disorders

- Fanconi anemia
- Ataxia telangiectasia
- Bloom syndrome

- Indicate DNA repair is a complex system
- Many genes
Genomic Imprinting -

• Expression of the gene depends on whether it is inherited from the mother or the father
• Genes are marked during gamete formation or early embryonic development
• The mechanism is not clearly understood
• Does not affect all genes
• Not a mutation, but a modification of the DNA affects the gene expression
• Example
  – Prader-Willi syndrome
  – Angelman syndrome
Imprinting on Chromosome 7

maternal alleles - red
paternal alleles - blue

Prevention of parthenogenesis
Conflict hypothesis

All female genome - abnormal placenta
All male genome - abnormal embryonic structures

Wood and Oakey, 2006
<table>
<thead>
<tr>
<th>Stem cells</th>
<th>Prenatal genetic testing</th>
<th>Cloning of animals/humans</th>
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<tr>
<td>Saunders</td>
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