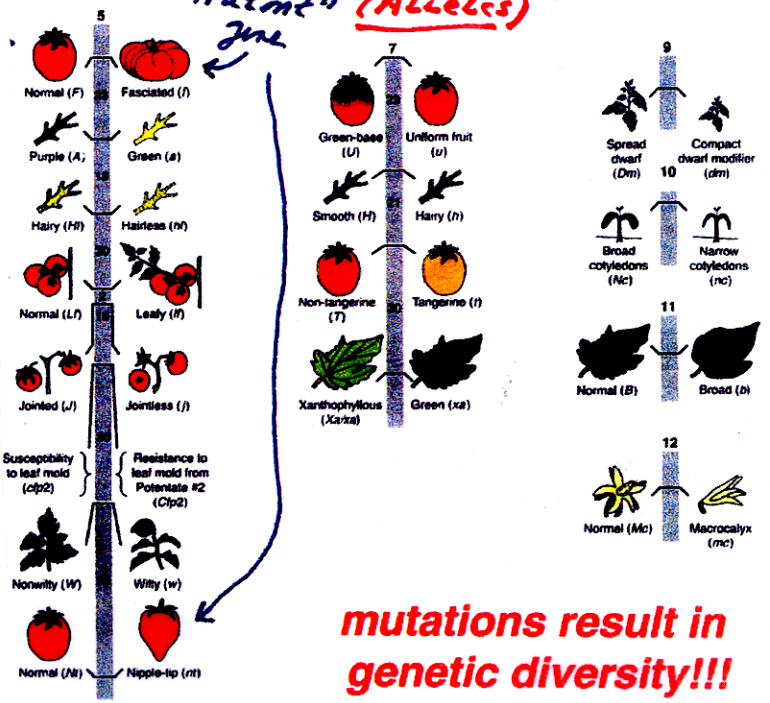
MUTATIONS GIVE RISE TO GENETIC DIVERSITY

Alternative Forms of the Same Gene Lead to Genetic Diversity

"Mutant" (Alleles)



mutations result in genetic diversity!!!

Change in DNA Sequence!

How know Sequence changed?

VARIABILITY ACTED ON & USED BY "Nature" & BY our ANCESTORS / EARLY Gene Engineers!

SAME PROCESS → DIVERSITY OF HUMAN GENES!

HOW KNOW MUTATION? ARE MUTATIONS GOOD, BAD, NEUTRAL?

HOW CAN IT BE SHOWN THAT MUTATIONS OCCUR RANDOMLY?

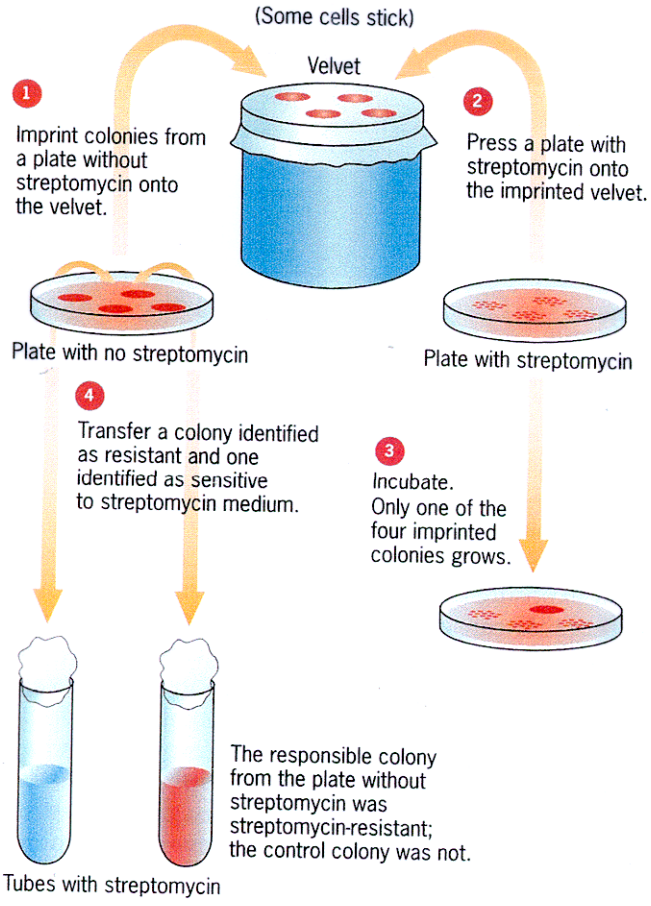


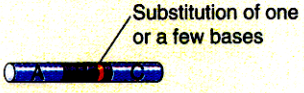
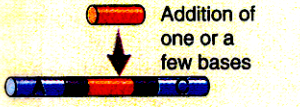
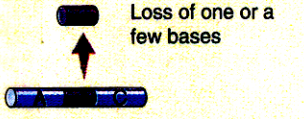
Figure 14.3 Joshua and Esther Lederberg's use of replica plating to demonstrate the random or nondirected nature of mutation. For simplicity, only four colonies are shown on each plate, and only two are tested for streptomycin resistance in step 4. Actually, each plate contains about 200 colonies, and many plates must be used to find an adequate number of mutant colonies.

1) Hypothesis?
2) Predictions?
3) Experiment to test?
4) Results?
5) Conclusions?

LEDERBERG EXPERIMENT!

MUTATIONS CAN OCCUR DIFFERENT WAYS

Table 18.1 Types of Mutation

Mutation	Example result
NO MUTATION	
	Normal B protein is produced by the *B* gene.
POINT MUTATION	
Base substitution	B protein is inactive because changed amino acid disrupts function.
	
Insertion	B protein is inactive because inserted material disrupts proper shape.
	
Deletion	B protein is inactive because portion of protein is missing.
	
CHANGES IN GENE POSITION	
Transposition	*B* gene or B protein may be regulated differently because of change in gene position.
	
Chromosomal rearrangement	*B* gene may be inactivated or regulated differently in its new location on chromosome.
	

(1) BASE-PAIR CHANGE

(2) ADD/DELETE BASE PAIRS

BASE SEQUENCE OF GENE CHANGES!

(3) MOVE GENE OR PART OF GENE TO NEW LOCATION!

SWITCHES CHANGE!

HUMAN GENETIC DISORDERS OCCUR AS A RESULT OF MUTATIONS

Table 13.2 Some Important Genetic Disorders

Disorder	Symptom	Defect	Dominant/ Recessive	Frequency among Human Births
Cystic fibrosis	Mucus clogs lungs, liver, and pancreas	Failure of chloride ion transport mechanism	Recessive	1/2500 (Caucasians)
Sickle cell anemia	Poor blood circulation	Abnormal hemoglobin molecules	Recessive	1/625 (African Americans)
Tay-Sachs disease	Deterioration of central nervous system in infancy	Defective enzyme (hexosaminidase A)	Recessive	1/3500 (Ashkenazi Jews)
Phenylketonuria	Brain fails to develop in infancy	Defective enzyme (phenylalanine hydroxylase)	Recessive	1/12,000
Hemophilia	Blood fails to clot	Defective blood clotting factor VIII	Sex-linked recessive	1/10,000 (Caucasian males)
Huntington's disease	Brain tissue gradually deteriorates in middle age	Production of an inhibitor of brain cell metabolism	Dominant	1/24,000
Muscular dystrophy (Duchenne)	Muscles waste away	Degradation of myelin coating of nerves stimulating muscles	Sex-linked recessive	1/3700 (males)
Hypercholesterolemia	Excessive cholesterol levels in blood, leading to heart disease	Abnormal form of cholesterol cell surface receptor	Dominant	1/500

HUMAN DISEASE GENES AFFECT MANY CELL PROCESSES AND MANIFEST THEMSELVES AT DIFFERENT TIMES

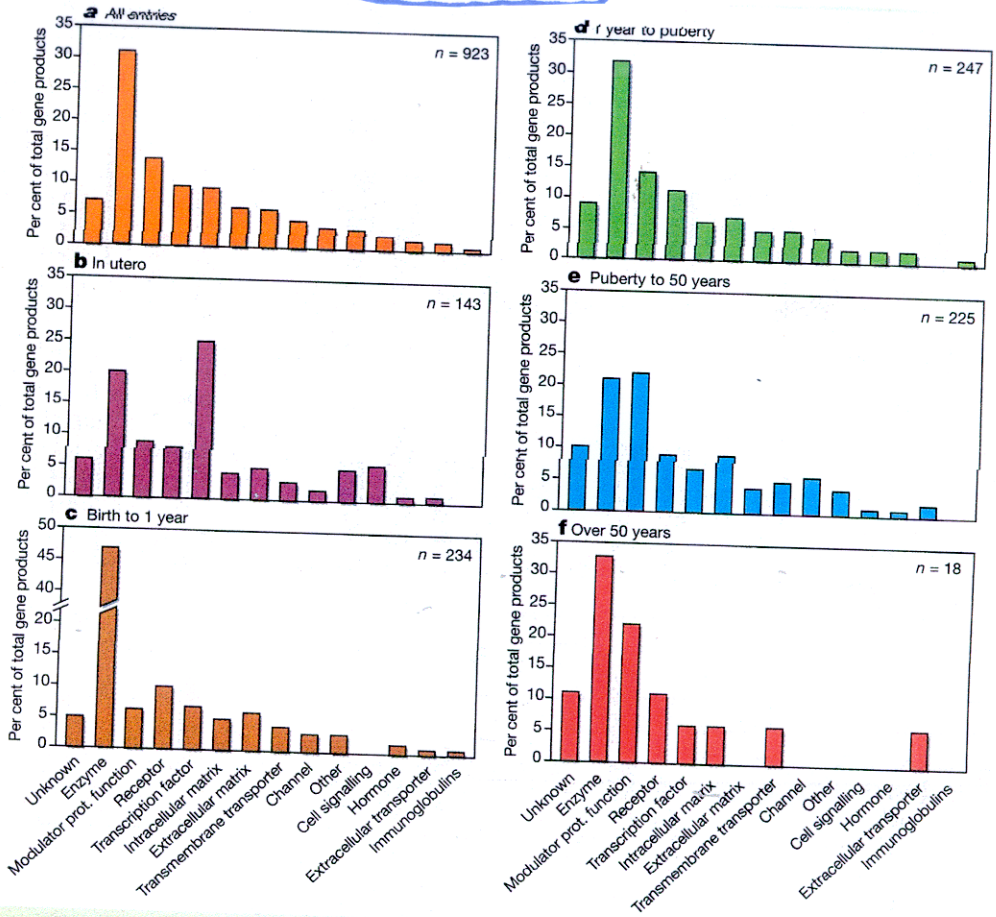
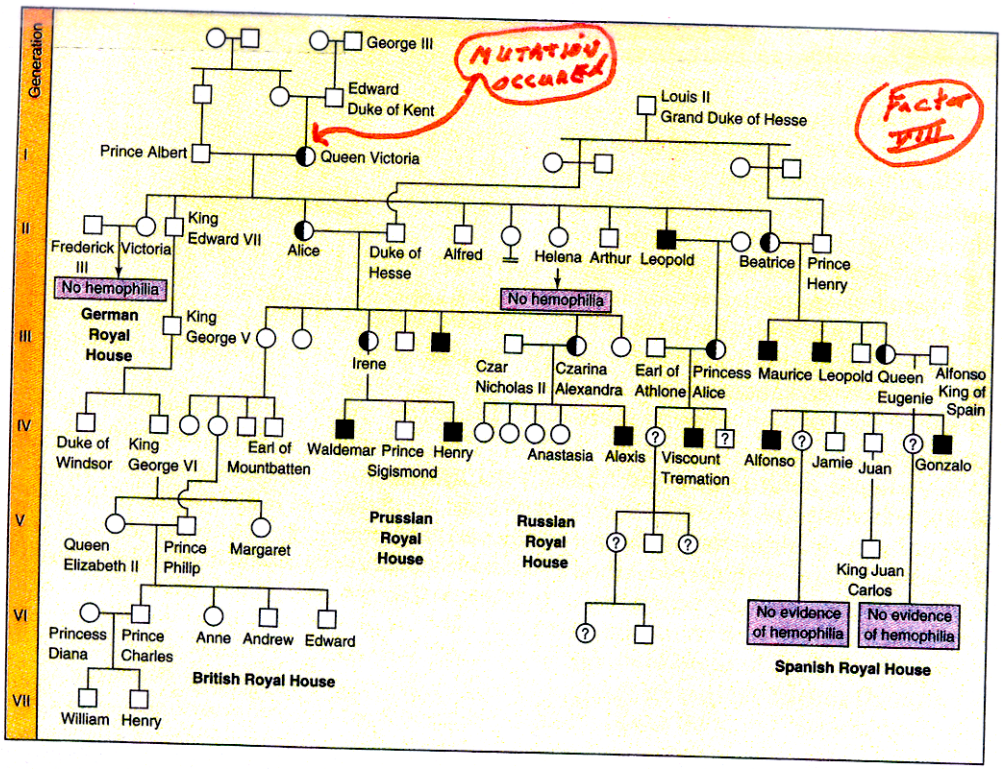


Figure 1 The functions of the protein products of disease genes. **a**, The entire disease gene set. **b–f**, Disease genes stratified according to the typical age of onset of the disease phenotype. The fraction of disease genes encoding transcription factors in the *in utero* onset disorders (25%) differs from the fraction encoding transcription factors for disorders with onset after birth (6%; $\chi^2 = 49.4$, $P < 0.001$). Similarly, the fraction of disease genes encoding enzymes causing a disorder with onset in the first year of life (47%) is different from the fraction encoding enzymes causing disorders with other ages of onset (25.8%; $\chi^2 = 35.8$, $P < 0.001$).

KNOW ABOUT THESE BECAUSE OF HUMAN GENOME PROJECT

PEDIGREES CAN BE USED TO FOLLOW DISEASE Genes in HUMAN FAMILIES



Hemophilia

Followed by Bleeding Phenotype

FIGURE 13.26

The Royal hemophilia pedigree. Queen Victoria's daughter Alice introduced hemophilia into the Russian and Austrian royal houses, and Victoria's daughter Beatrice introduced it into the Spanish royal house. Victoria's son Leopold, himself a victim, also transmitted the disorder in a third line of descent. Half-shaded symbols represent carriers with one normal allele and one defective allele; fully shaded symbols represent affected individuals.

OR FOLLOWED BY DNA TESTS USING MOLECULAR METHODS

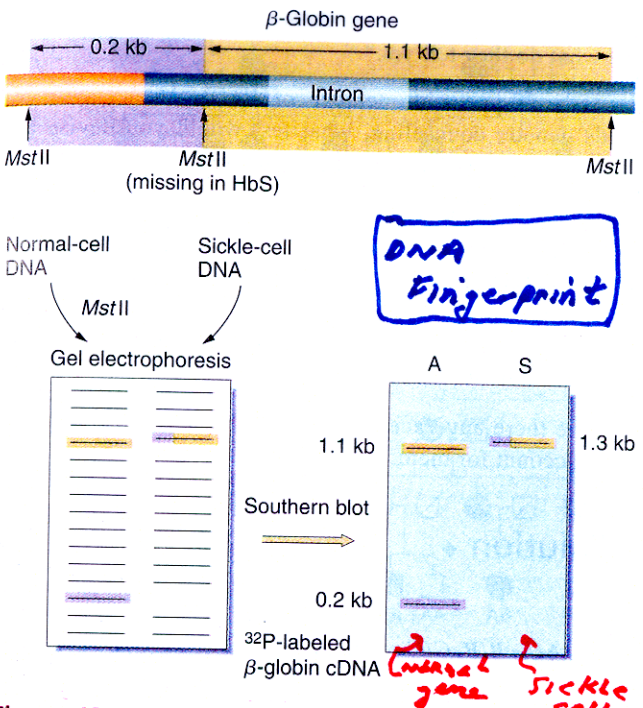


Figure 13-29 Detection of the sickle-cell globin gene by Southern blotting. The base change (A → T) that causes sickle-cell anemia destroys a *Mst*II target site that is present in the normal β-globin gene. This difference can be detected by Southern blotting. (After *Recombinant DNA*, 2d ed. Scientific American Books. Copyright © 1992 by J. D. Watson, M. Gilman, J. Witkowski, and M. Zoller.)

DNA Fingerprint

SICKLE-CELL VS. "NORMAL" GLOBIN GENE/PROTEIN

Type of Hb	Amino acid sequence Nucleotide sequence
A	—Pro—Glu—Glu— —CCT—GAG—GAG— *Mst*II
S	—Pro—Val—Glu— —CCT—GTG—GAG

BASIS OF DNA TESTING USING CLONED GENES OR PCR

IMPLICATIONS? COMBINED WITH PCR & TESTING OF EMBRYOS? CONFINED TO FATAL DISEASE GENES?

MUTATIONS in GENES ARE RARE
BUT ARE INHERITED

① one gene per gamete

② 2 genes per somatic cell

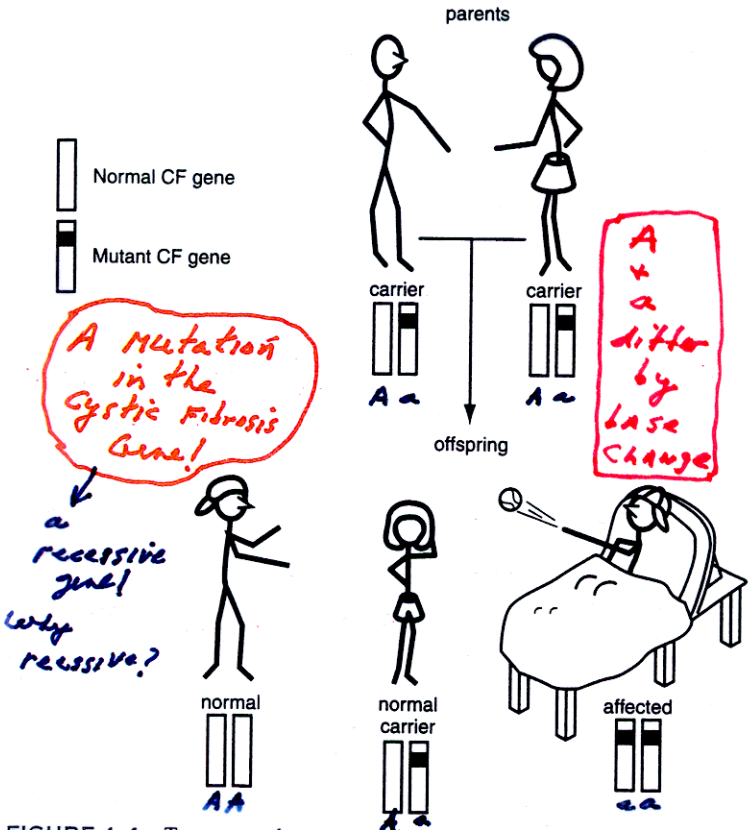


FIGURE 1.4 Two parents heterozygous for CF and producing normal or affected kids.

FOLLOWS Mendelian Rules

Eugenics

How FOLLOW Inheritance?
What Allows Disease to be Followed?

HOW DOES A GENE LEAD TO A PHENOTYPE?

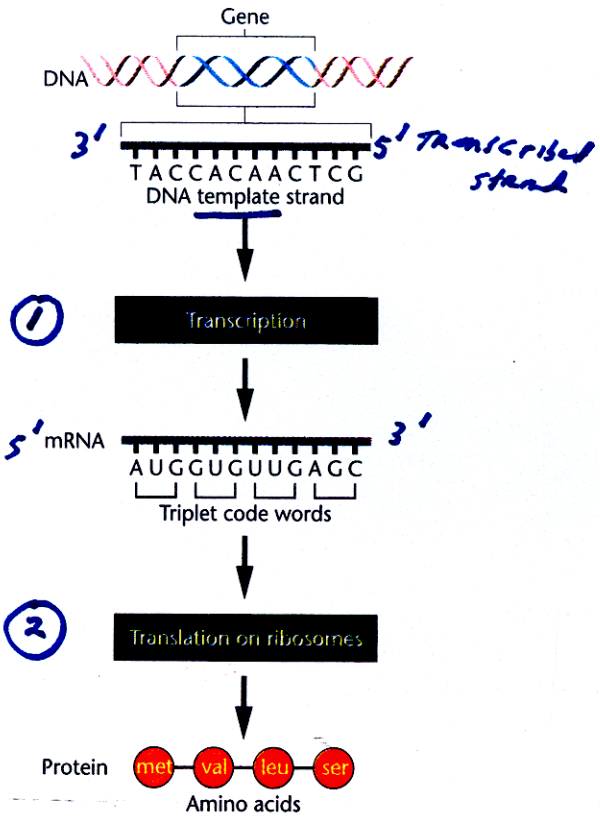


FIGURE 13.1 An overview of the concept of the flow of genetic information encoded in DNA to messenger RNA to protein.

(1) mRNA synthesized by TRANSCRIPTION

COMPLEMENTARY to TRANSCRIBED, NONSENSE STRAND

SAME SEQUENCE as SENSE STRAND

(2) mRNA TRANSLATED into PROTEIN by TRANSLATION of the Genetic Code

Genetic Code on mRNA TRANSLATED to protein sequence

∴ Sequence of Gene
↳ Sequence of mRNA
↳ Sequence of protein

KNOW SEQUENCE
KNOW PROTEIN
↳ ENGINEER NEW PROTEINS

GENETIC CODE ALLOWS THE SEQUENCE of NUCLEOTIDES in mRNA / Sense STRAND of Gene to be TRANSLATED into Sequence of AMINO ACIDS in PROTEINS

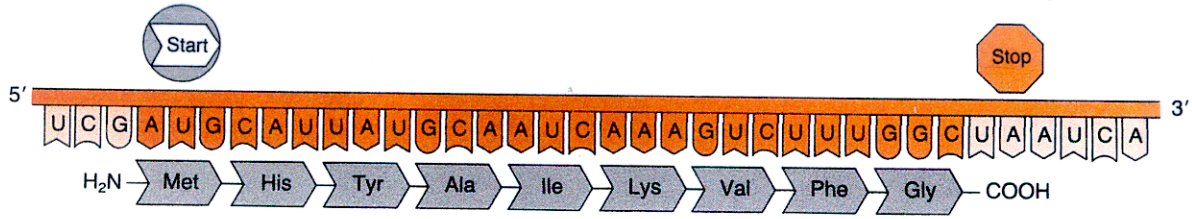


Figure 3.4 Decoding a messenger RNA sequence into a polypeptide.

NOTE: SEQUENCE in mRNA (= Sense Gene STRAND) IS TRANSLATED 5′→3′ (= beginning of sense STRAND to end) & protein made in N→C direction ∴ order nts in gene = order aa in protein!

The Genetic Code is Universal!

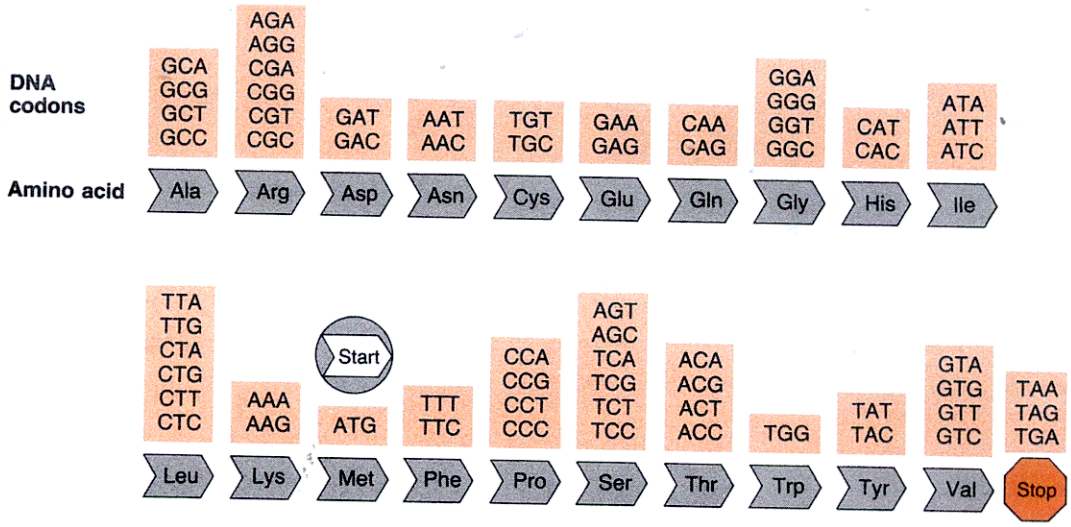


Figure 3.3 The genetic code. The codons shown for each amino acid are those for DNA. For RNA, the Ts are replaced by Us.

(1) Universal (2) Triplet (3) Punctuation
(4) Degenerate

KNOW SEQUENCE OF GENE - KNOW SEQUENCE OF PROTEIN USING GENETIC CODE

Big Implication For Genetic Engineering! CAN MAKE Genes, Genomes & Specify Proteins Wanted! CAN Express Genes FROM ONE Organism in Another!

ANOTHER VIEW OF THE GENETIC CODE

Second letter

First letter	U	C	A	G	Third letter
U	UUU, UUC: Phenylalanine	UCU, UCC, UCA, UCG: Serine	UAU, UAC: Tyrosine	UGU, UGC: Cysteine	U C A G
	UUA, UUG: Leucine		UAA: Stop codon; UAG: Stop codon	UGA: Stop codon; UGG: Tryptophan	
C	CUU, CUC, CUA, CUG: Leucine	CCU, CCC, CCA, CCG: Proline	CAU, CAC: Histidine; CAA, CAG: Glutamine	CGU, CGC, CGA, CGG: Arginine	U C A G
A	AUU, AUC, AUA: Isoleucine	ACU, ACC, ACA, ACG: Threonine	AAU, AAC: Asparagine; AAA, AAG: Lysine	AGU, AGC: Serine; AGA, AGG: Arginine	U C A G
	AUG: Methionine; start codon				
G	GUU, GUC, GUA, GUG: Valine	GCU, GCC, GCA, GCG: Alanine	GAU, GAC: Aspartic acid; GAA, GAG: Glutamic acid	GGU, GGC, GGA, GGG: Glycine	U C A G

12.5 The Universal Genetic Code

Genetic information is encoded in mRNA in three-letter units—codons—made up of the bases uracil (U), cytosine (C), adenine (A), and guanine (G). To decode a codon, find its first letter in the left column, then read across the top to its second letter, then read down the right column to its third letter. The amino acid the codon specifies is given in the corresponding row. For example, AUG codes for methionine, and GUA codes for valine.

Design Experiment to Show Universal!

There is a colinearity between nucleotide sequence of a gene & amino acid sequence of protein

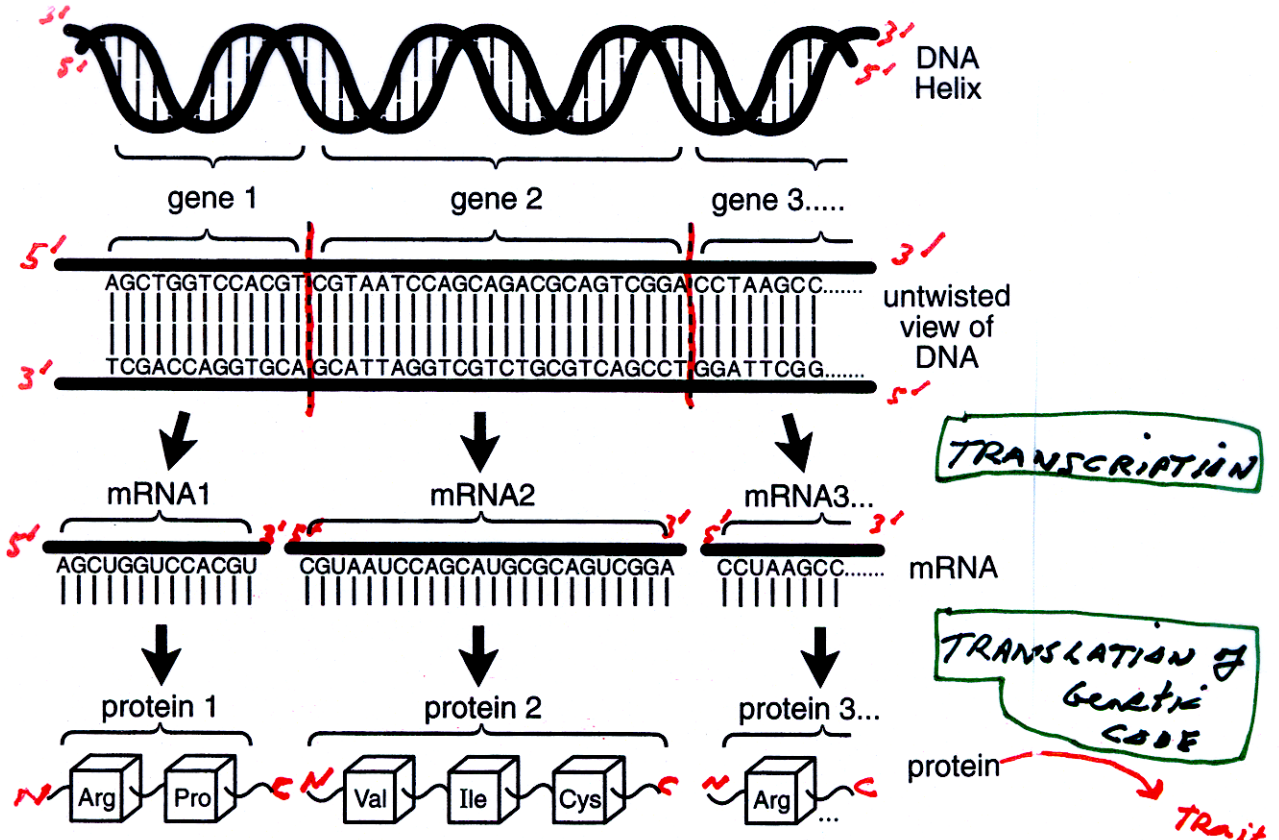


FIGURE 2.6 Adjacent sets of base pairs comprising different genes.

Specific trait 1 → Function 1

Specific trait 2 → Function 2

Specific trait 3 → Function 3

ONE GENE → ONE PROTEIN → ONE FUNCTION

TRANSCRIPTION OF A GENE INTO Complementary RNA

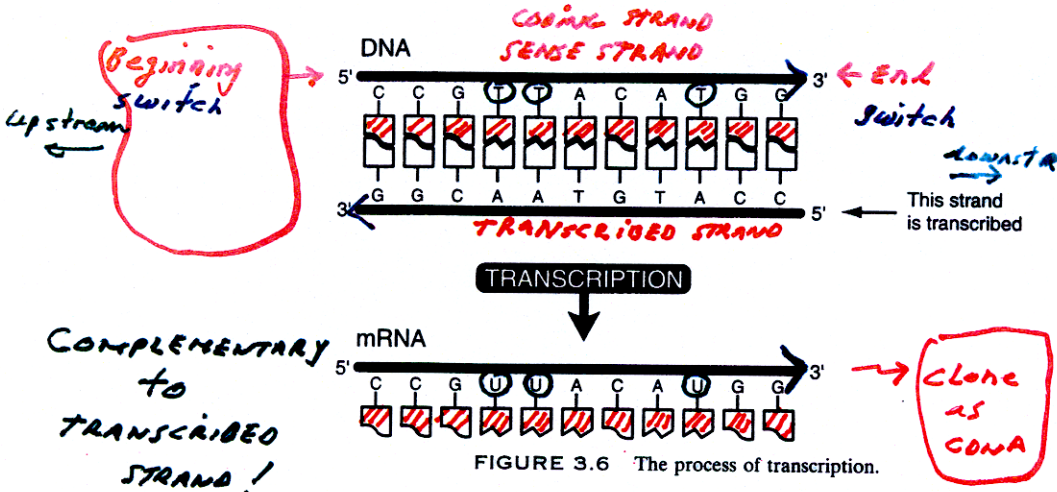


FIGURE 3.6 The process of transcription.

NOTE!

1. one gene strand TRANSCRIBED
2. RNA has same sequence as SENSE STRAND
3. RNA is complementary to TRANSCRIBED Strand
4. Uracil takes the place of Thymine in RNA!

PROCESS OF TRANSCRIPTION

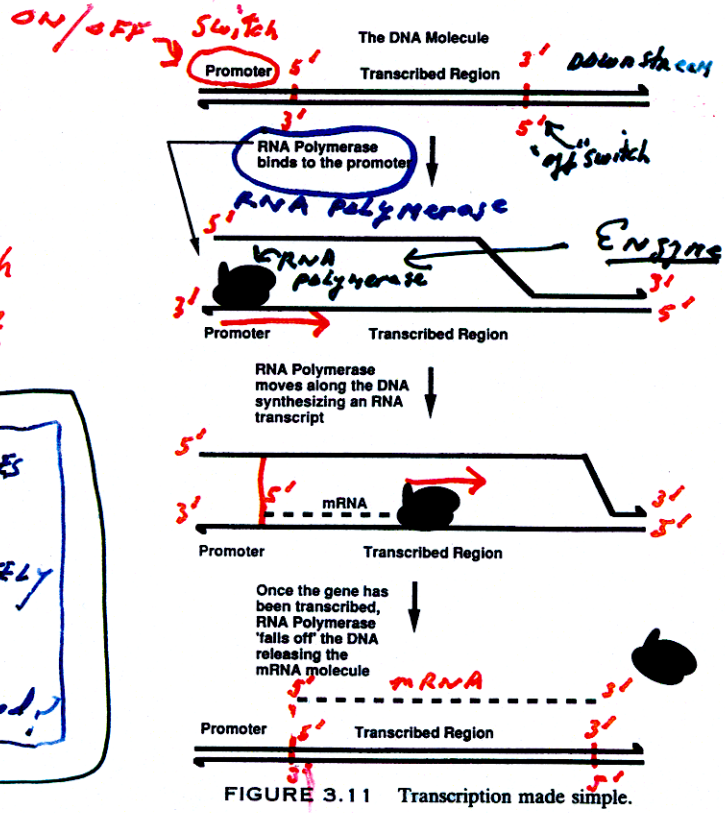


FIGURE 3.11 Transcription made simple.

NOTE!

The Switch TURNS the Gene on!

CAN SWITCHES + GENES BE SEPARATELY CLONED & ENGINEERED!

QUESTIONS?!

ARE SWITCHES Gene Specific?

ARE switches ORGANISM or KINGDOM Specific?

TRANSCRIPTION OCCURS ON the Anti-Sense STRAND

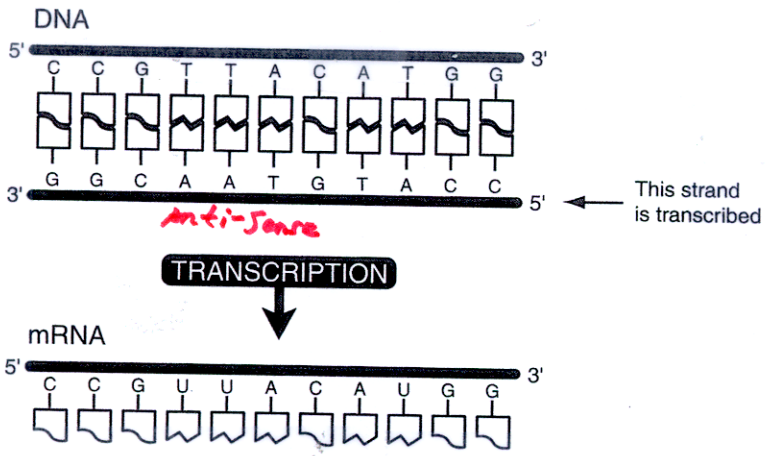


FIGURE 3.6 The process of transcription.

And Results in a RNA Complementary to Anti-Sense STRAND

But the SAME Sequence as Sense Strand

TRANSCRIPtion Starts at the Switch!

TRANSCRIPTION BEGINS at the Switch

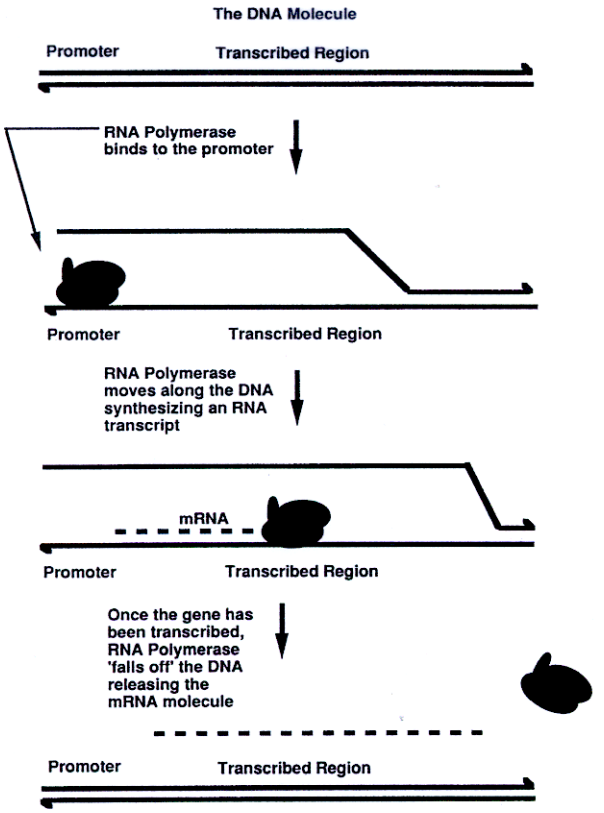


FIGURE 3.11 Transcription made simple.

Recall - Switches are Gene & Kingdom Specific! They can be cloned & used in Synthetic Genes!

VISUALIZING TRANSCRIPTION

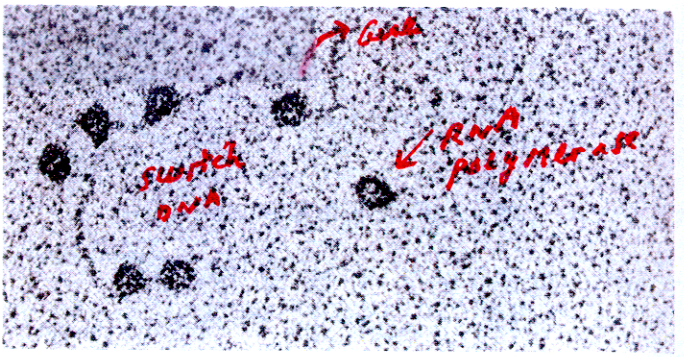


FIGURE 15.7
RNA polymerase. In this electron micrograph, the dark circles are RNA polymerase molecules bound to several promoter sites on bacterial virus DNA.

RNA Polymerase Binds to Switch & TRANSCRIBES Nonsense Strand of Gene (3'→5' strand) in a 5'→3' direction!

VISUALIZING TRANSCRIPTION CARTOON

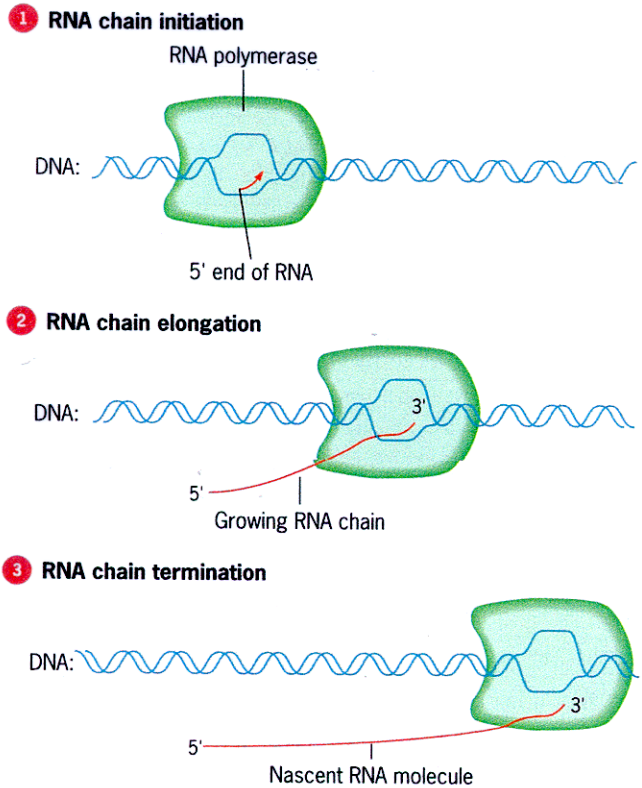


Figure 12.10 The three stages of transcription: initiation, elongation, and termination.

A Gene is TRANSCRIBED MANY TIMES into Complementary RNAs

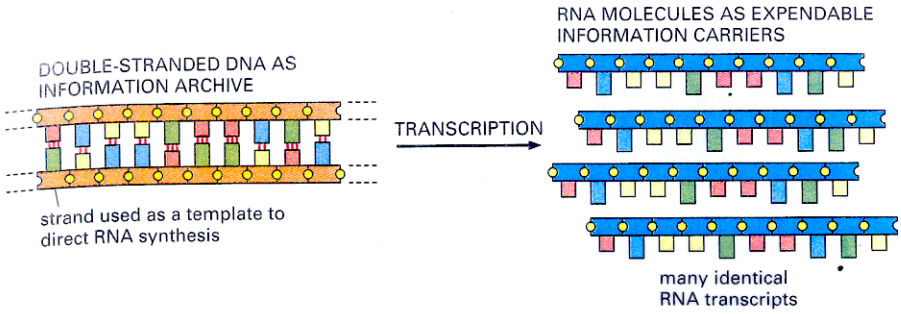
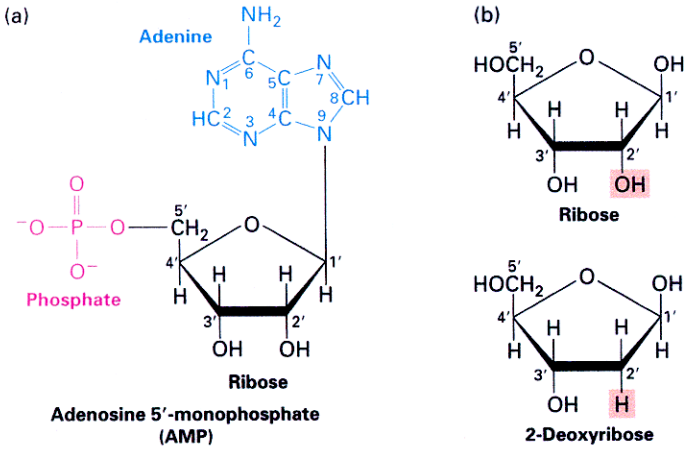


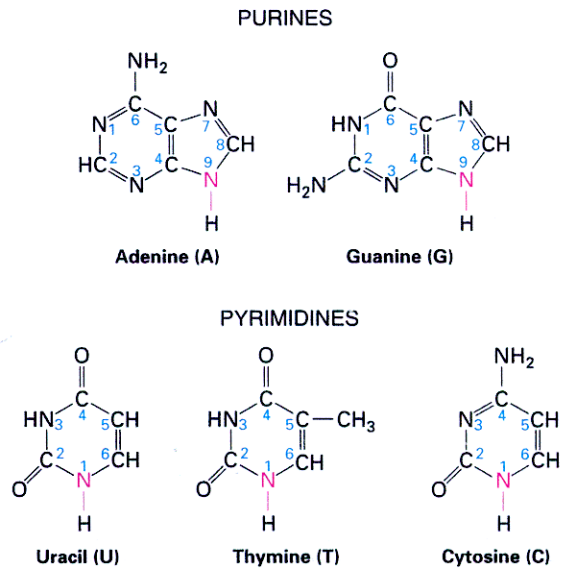
Figure 1–5 How genetic information is broadcast for use inside the cell. Each cell contains a fixed set of DNA molecules—its archive of genetic information. A given segment of this DNA serves to guide the synthesis of many identical RNA transcripts, which serve as working copies of the information stored in the archive. Many different sets of RNA molecules can be made by transcribing selected parts of a long DNA sequence, allowing each cell to use its information store differently.

Significance to Biology?
to Genetic Engineering?

RNA or Ribonucleic Acid contains Ribose Sugar and Uracil



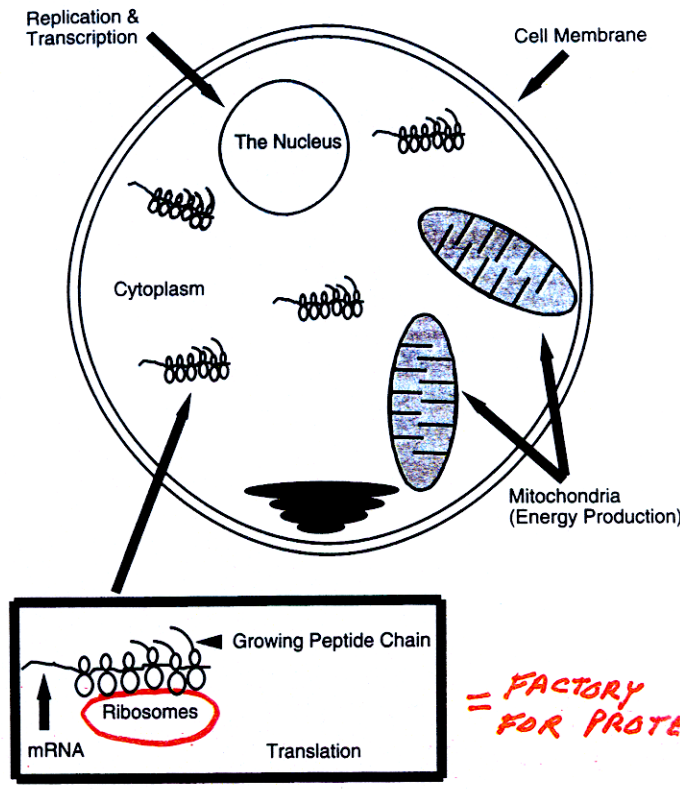
▲ **FIGURE 2-14 Common structure of nucleotides.** (a) Adenosine 5′-monophosphate (AMP), a nucleotide present in RNA. By convention, the carbon atoms of the pentose sugar in nucleotides are numbered with primes. In natural nucleotides, the 1′ carbon is joined by a β linkage to the base (in this case adenine); both the base (blue) and the phosphate on the 5′ hydroxyl (red) extend above the plane of the furanose ring. (b) Ribose and deoxyribose, the pentoses in RNA and DNA, respectively.



▲ **FIGURE 2-15 Chemical structures of the principal bases in nucleic acids.** In nucleic acids and nucleotides, nitrogen 9 of purines and nitrogen 1 of pyrimidines (red) are bonded to the 1′ carbon of ribose or deoxyribose. U is only in RNA, and T is only in DNA. Both RNA and DNA contain A, G, and C.

IN PLACE of Deoxyribose Sugar and Thymine!

PROTEINS ARE synthesized in the cytoplasm of ALL cells.....



= FACTORY FOR PROTEIN synthesis

FIGURE 3.5 Structures and events in the cytoplasm of a cell.

Using factories called ribosomes .....

And 3 RNAs called.....

① Genetic code

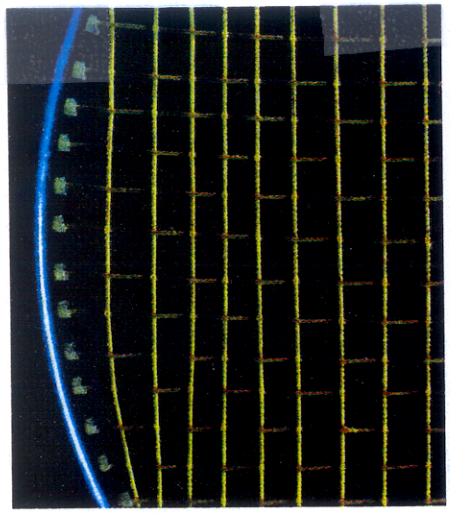
② Ribosome structure → Peptide bond formation

③ CARRY amino acids

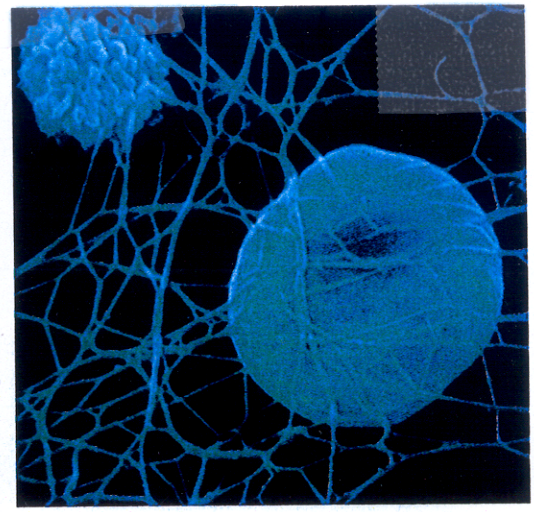
Table 7–1	Types of RNA Produced in Cells
Type of RNA	**Function**
mRNAs	codes for proteins
rRNAs	forms part of the structure of the ribosome and participates in protein synthesis
tRNAs	used in protein synthesis as an adaptor between mRNA and amino acids

UNIQUE PROTEINS CARRY OUT UNIQUE CELL FUNCTIONS

collagen

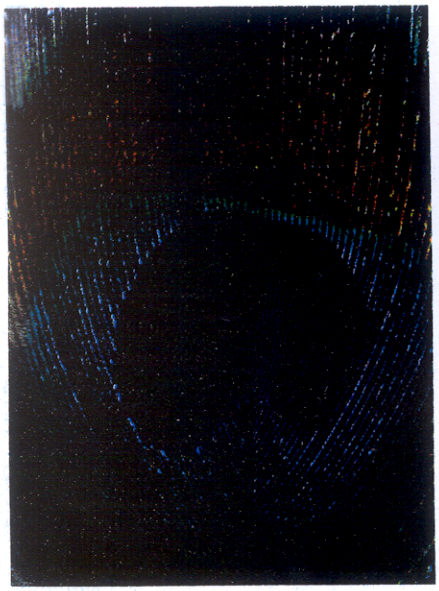


(a)



(b)

blood clot (protein) etc.



(c) keratin (feather)

(d) e.g. Spider Silk Protein in plants / Goat milk!

(e) keratin (hair)

FIGURE 3.4
Some of the more common structural proteins. (*a*) Collagen: strings of a tennis racket from gut tissue; (*b*) fibrin: scanning electron micrograph of a blood clot (3000×); (*c*) keratin: a peacock feather; (*d*) silk: a spider's web; (*e*) keratin: human hair.

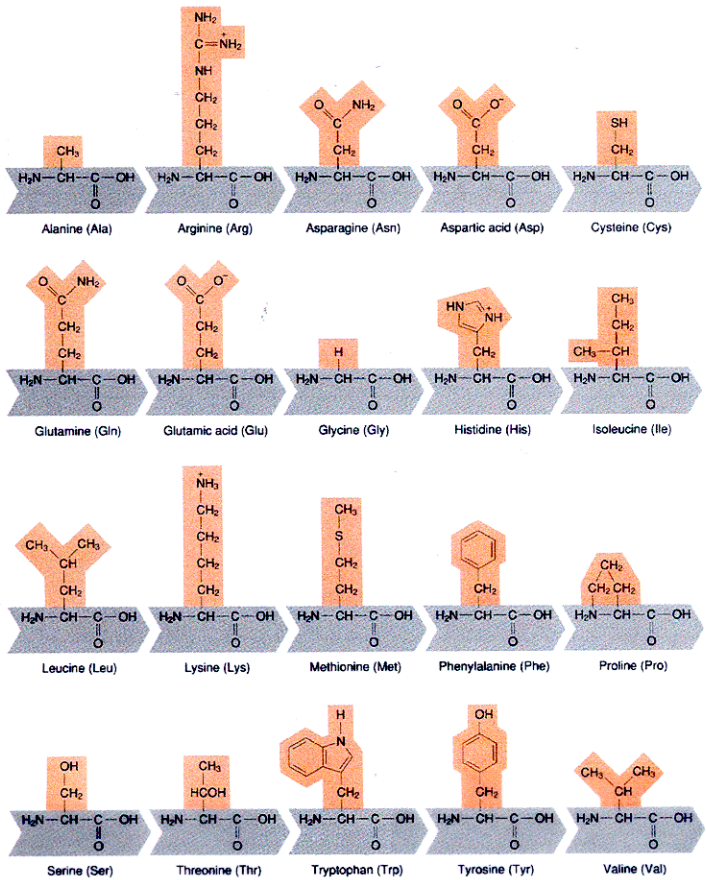
PROTEINS CARRY OUT DIVERSE CELL FUNCTIONS AND ARE UNIQUE BECAUSE OF SEQUENCE!

Table 3.2 The Many Functions of Proteins

Function	Class of Protein	Examples	Use
Metabolism (Catalysis)	Enzymes	Hydrolytic enzymes	Cleave polysaccharides
		Proteases	Break down proteins
For Gene Engineer →	DNA Polymerase, Reverse Tpase, Terminal Transferase, Restriction Enzymes	Polymerases	Produce nucleic acids
		Kinases	Phosphorylate sugars and proteins
Defense	Immunoglobulins	Antibodies	Mark foreign proteins for elimination
	Toxins	Snake venom	Block nerve function
Cell recognition	Cell surface antigens	MHC proteins	"Self" recognition
Transport throughout body	Globins	Hemoglobin	Carries O_2 and CO_2 in blood
		Myoglobin	Carries O_2 and CO_2 in muscle
		Cytochromes	Electron transport
Membrane transport	Transporters	Sodium-potassium pump	Excitable membranes
		Proton pump	Chemiosmosis
		Anion channels	Transport Cl^- ions
Structure/Support	Fibers	Collagen	Cartilage
		Keratin	Hair, nails
		Fibrin	Blood clot
Motion	Muscle	Actin	Contraction of muscle fibers
		Myosin	Contraction of muscle fibers
Osmotic regulation	Albumin	Serum albumin	Maintains osmotic concentration of blood
Regulation of gene action	Repressors (regulate switches!)	*lac* repressor	Regulates transcription
Regulation of body functions	Hormones	Insulin	Controls blood glucose levels
		Vasopressin	Increases water retention by kidneys
		Oxytocin	Regulates uterine contractions and milk production
Storage	Ion binding	Ferritin	Stores iron, especially in spleen
		Casein	Stores ions in milk
		Calmodulin	Binds calcium ions

Mutate Gene → Mutate Protein → Defective function

PROTEINS ARE MADE OF AMINO ACIDS



20 Amino Acids Differ By Chemistry

Chemistry of Proteins
↳ Biology

AMINO ACIDS ARE JOINED BY Peptide Bonds

Corresponds to 5' → 3' end of mRNA / sense strand of gene!

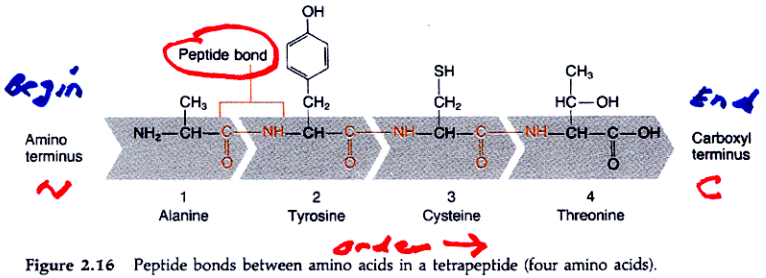
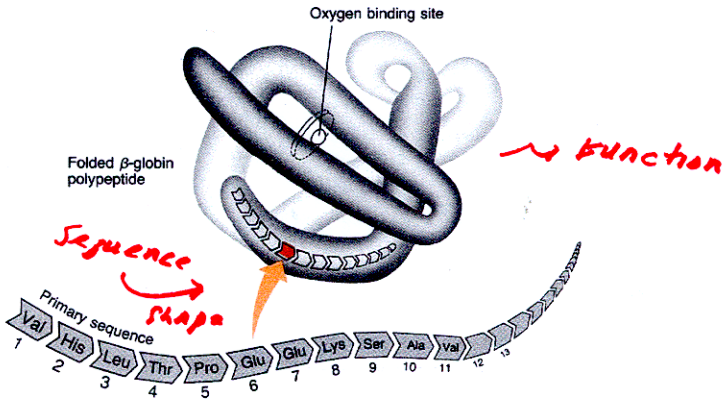


Figure 2.16 Peptide bonds between amino acids in a tetrapeptide (four amino acids).

Change Shape
Change Function
Change Phenotype



In sickle-cell hemoglobin, the Glu at position 6 is replaced by Val

Figure 2.17 A portion of the primary structure of the β-globin polypeptide and its location in the folded, complete polypeptide. Also shown is the amino acid that is altered in the β-globin polypeptide in sickle-cell disease.

ORDER OF AMINO ACIDS → Specific Protein Shape & Function

→ Phenotype