Introduction to Molecular Biology and Genomics

BMI/CS 576

www.biostat.wisc.edu/bmi576/

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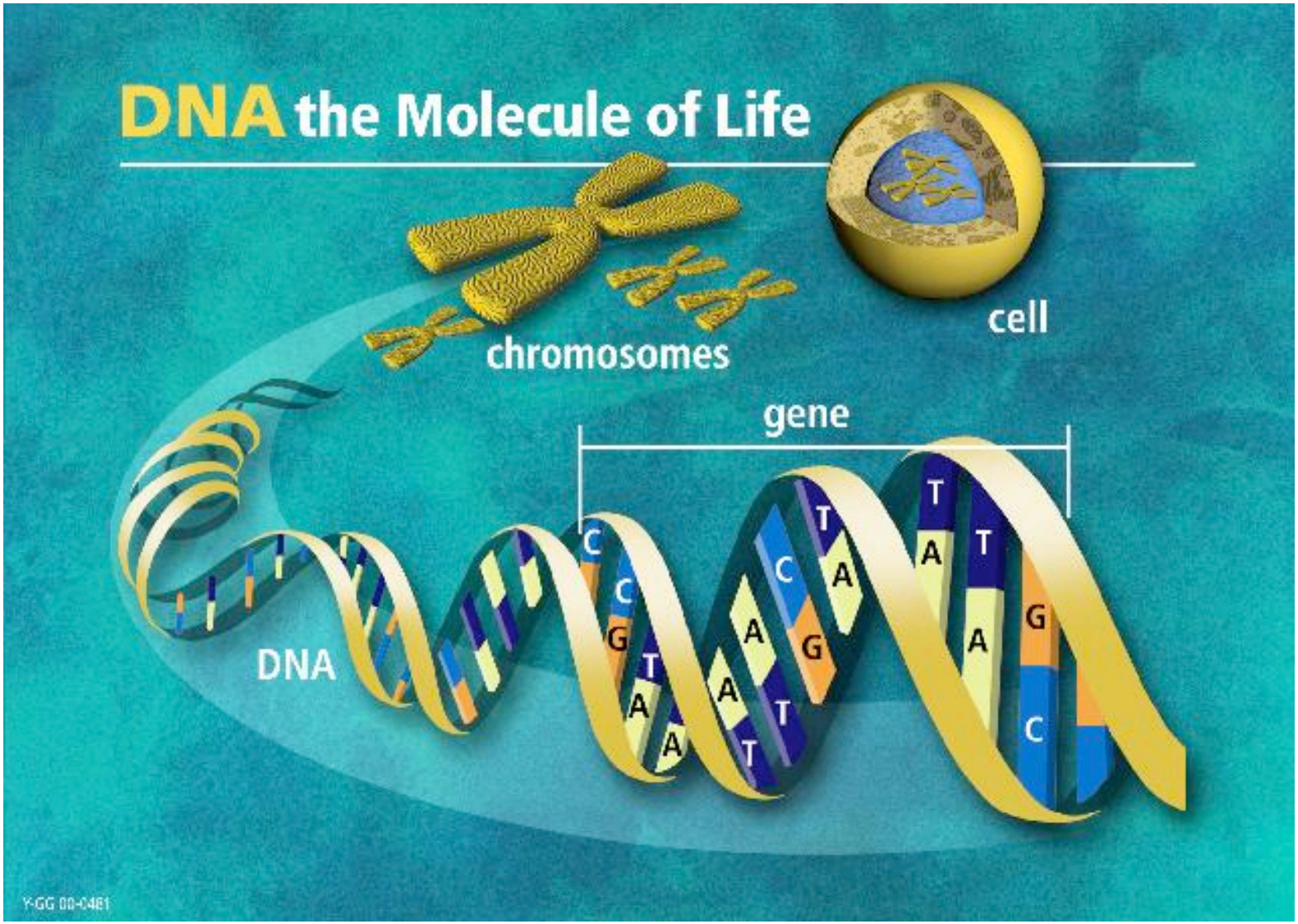


image from the DOE Human Genome Program
http://www.ornl.gov/hgmis

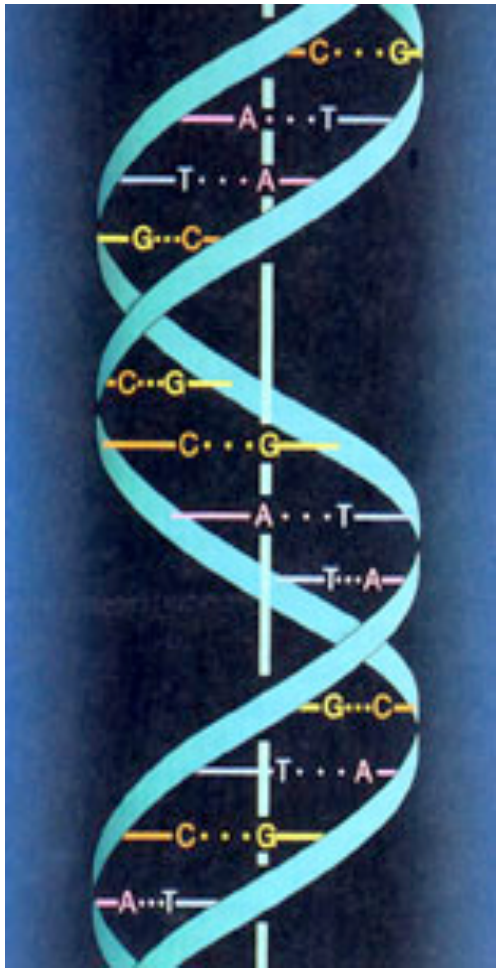
DNA

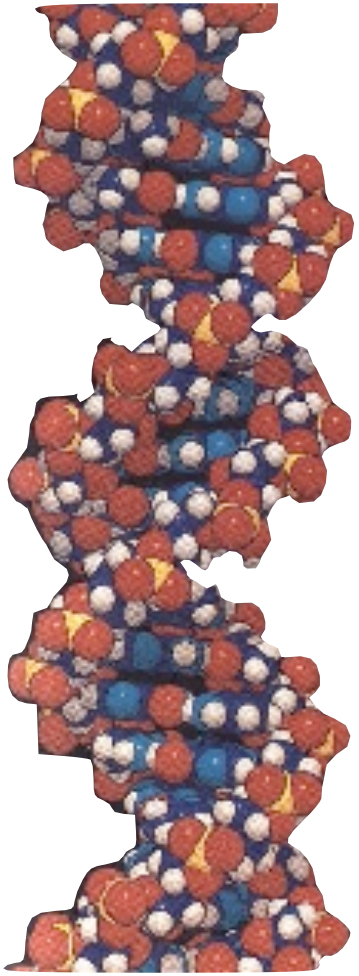
- can be thought of as the "blueprint" for an organism
- a linear chain of small molecules called *nucleotides*
  - four different nucleotides distinguished by the four *bases*: adenine (A), cytosine (C), guanine (G) and thymine (T)
- is a *polymer*: large molecule consisting of similar units (nucleotides in this case)
- a single strand of DNA can be thought of as a string composed of the four letters: A, C, G, T

CTGCTGGACCGGGTGCTAGGACCCTGACTGCCCGGGG
CCGGGGGTGCGGGGCCCGCTGAG...

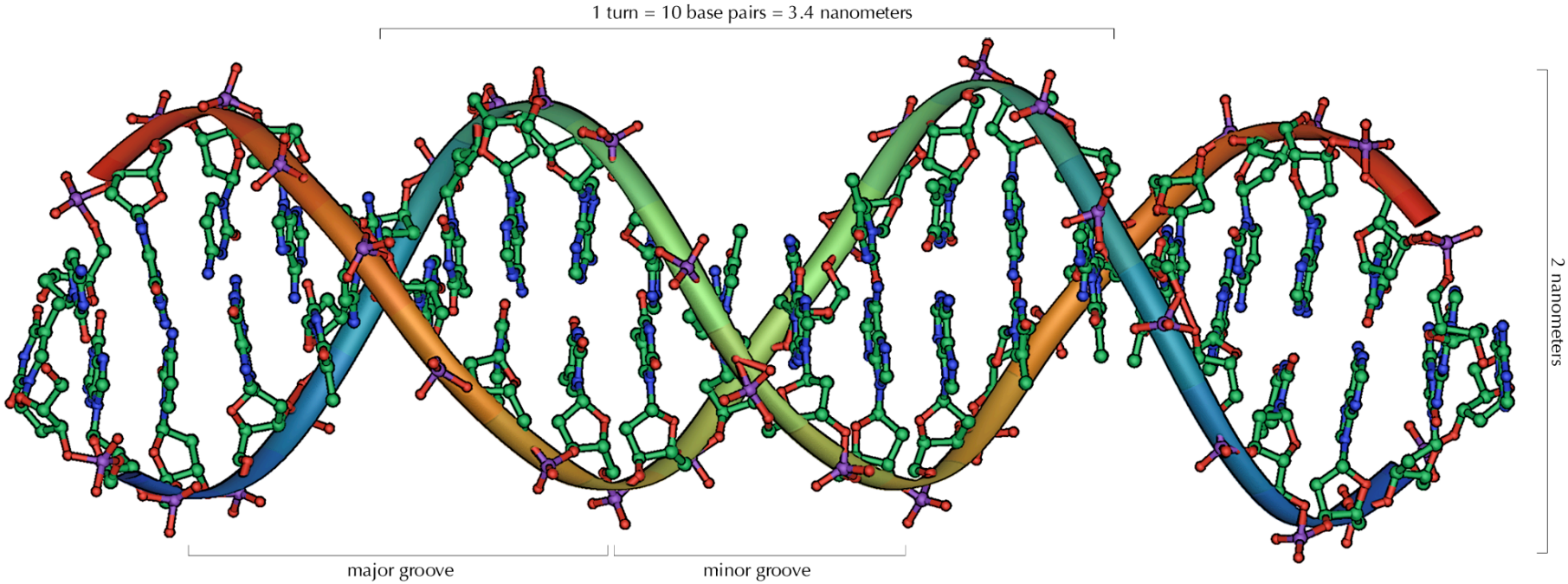
The Double Helix

- DNA molecules usually consist of two strands arranged in the famous double helix

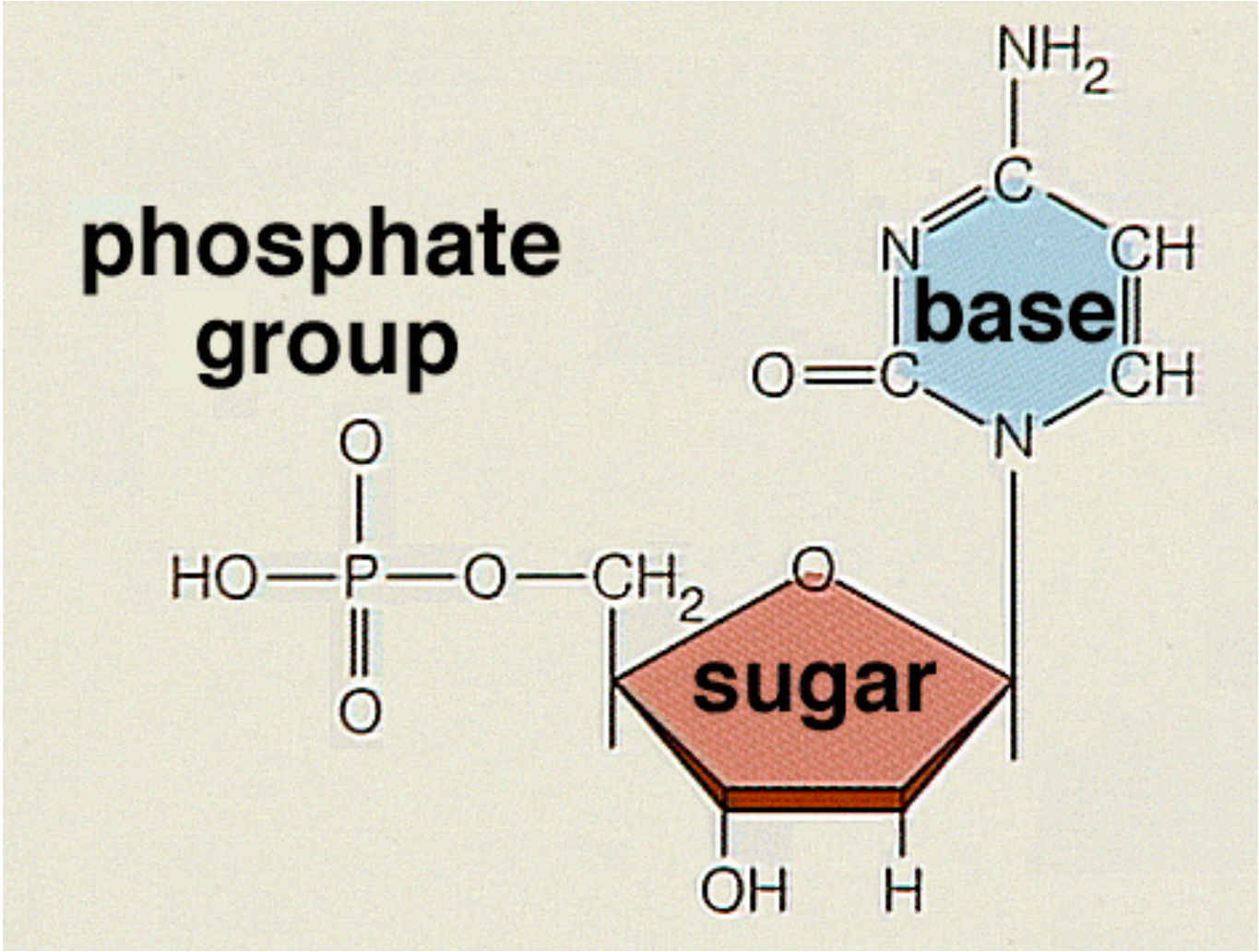




DNA Dimensions



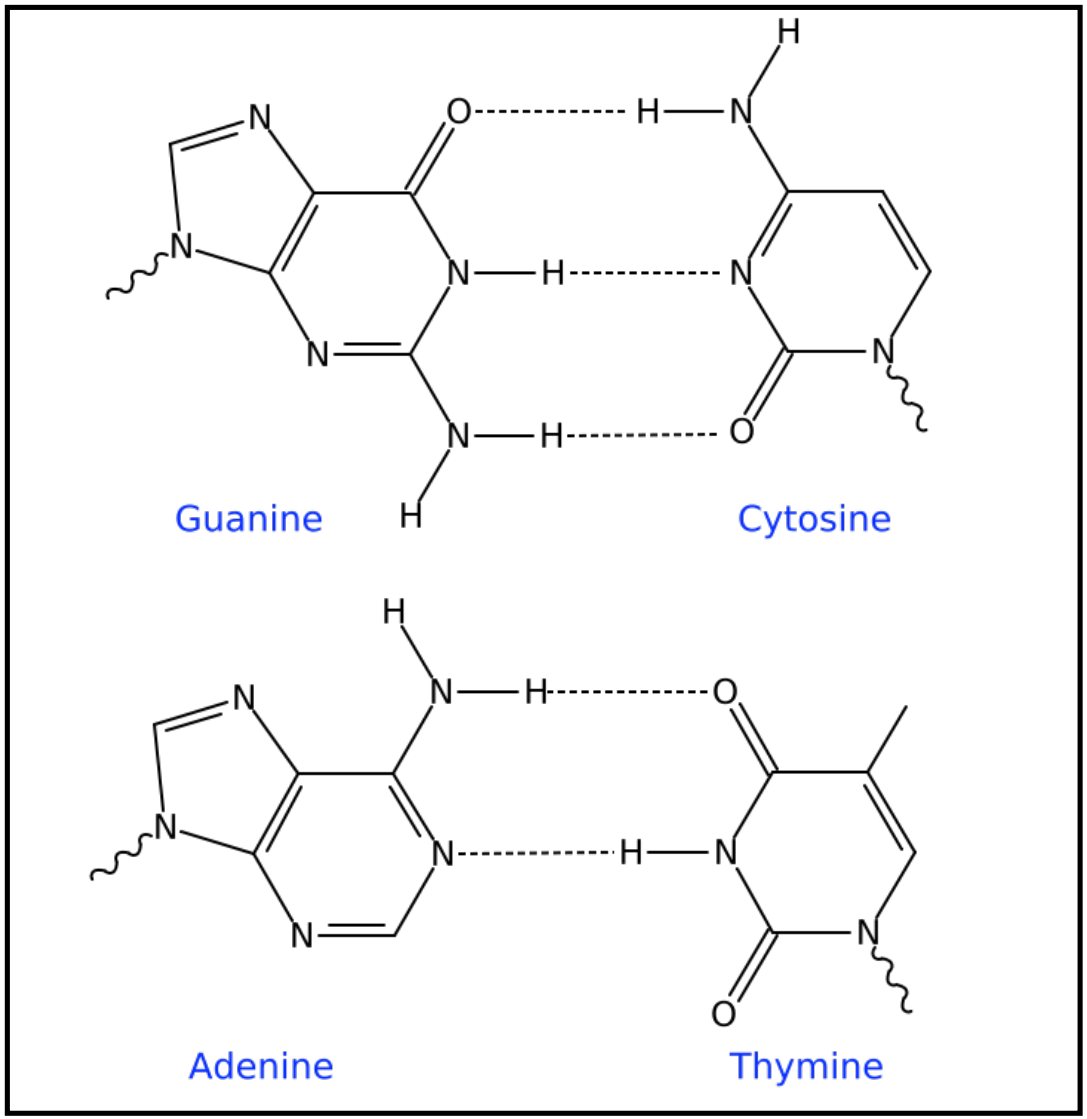
Nucleotides: the subunits of DNA



The four DNA bases

G

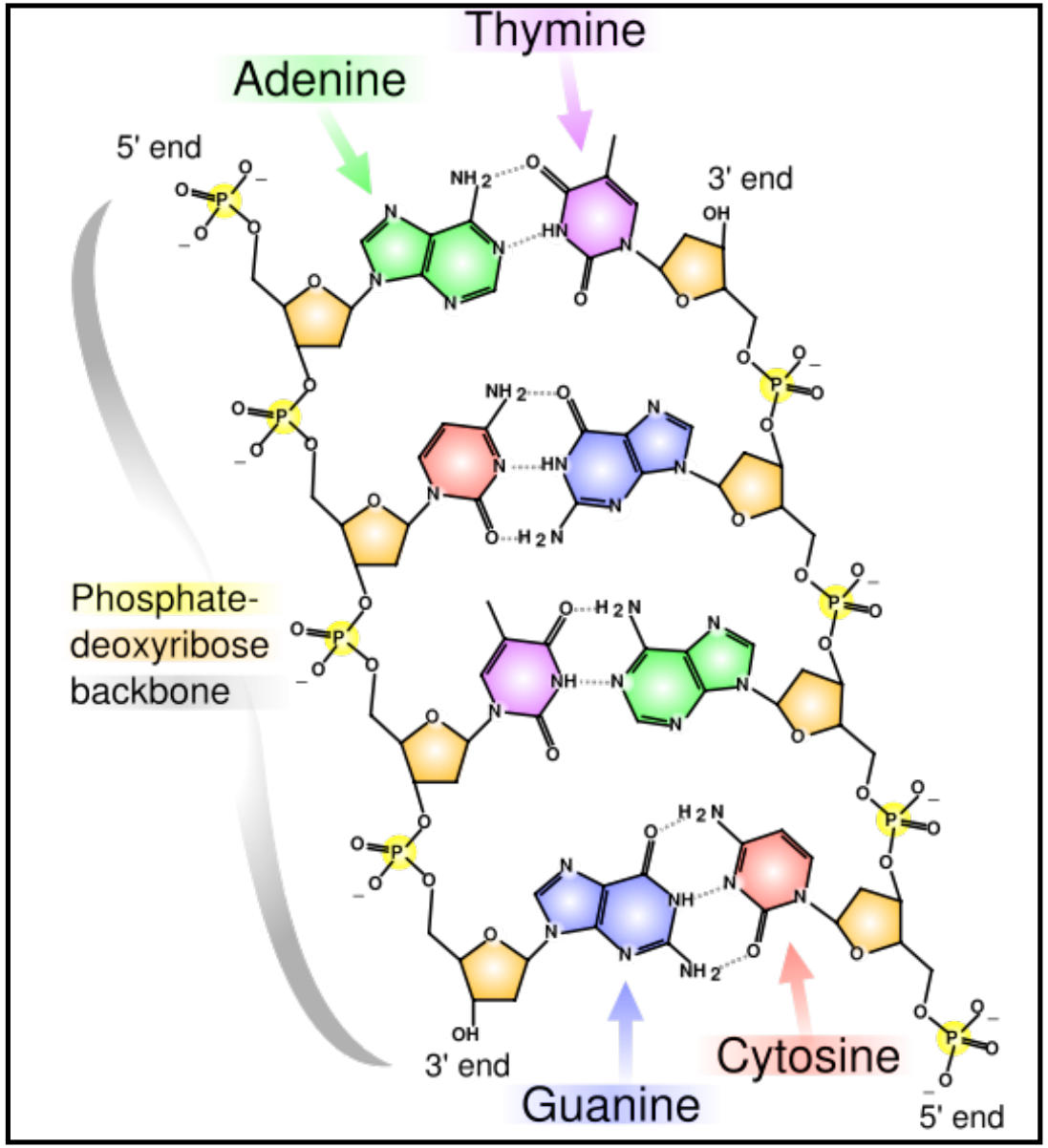
C



A

T

DNA strand: polymer of nucleotides



- in double-stranded DNA

A always bonds to T

C always bonds to G

The Double Helix

- each strand of DNA has a "direction"
  - at one end, the terminal carbon atom in the backbone is the 5′ carbon atom of the terminal sugar
  - at the other end, the terminal carbon atom is the 3′ carbon atom of the terminal sugar
- therefore we can talk about the *5'* and the *3'* ends of a DNA strand
- in a double helix, the strands are *antiparallel* (arrows drawn from the 5′ end to the 3′ end go in opposite directions)

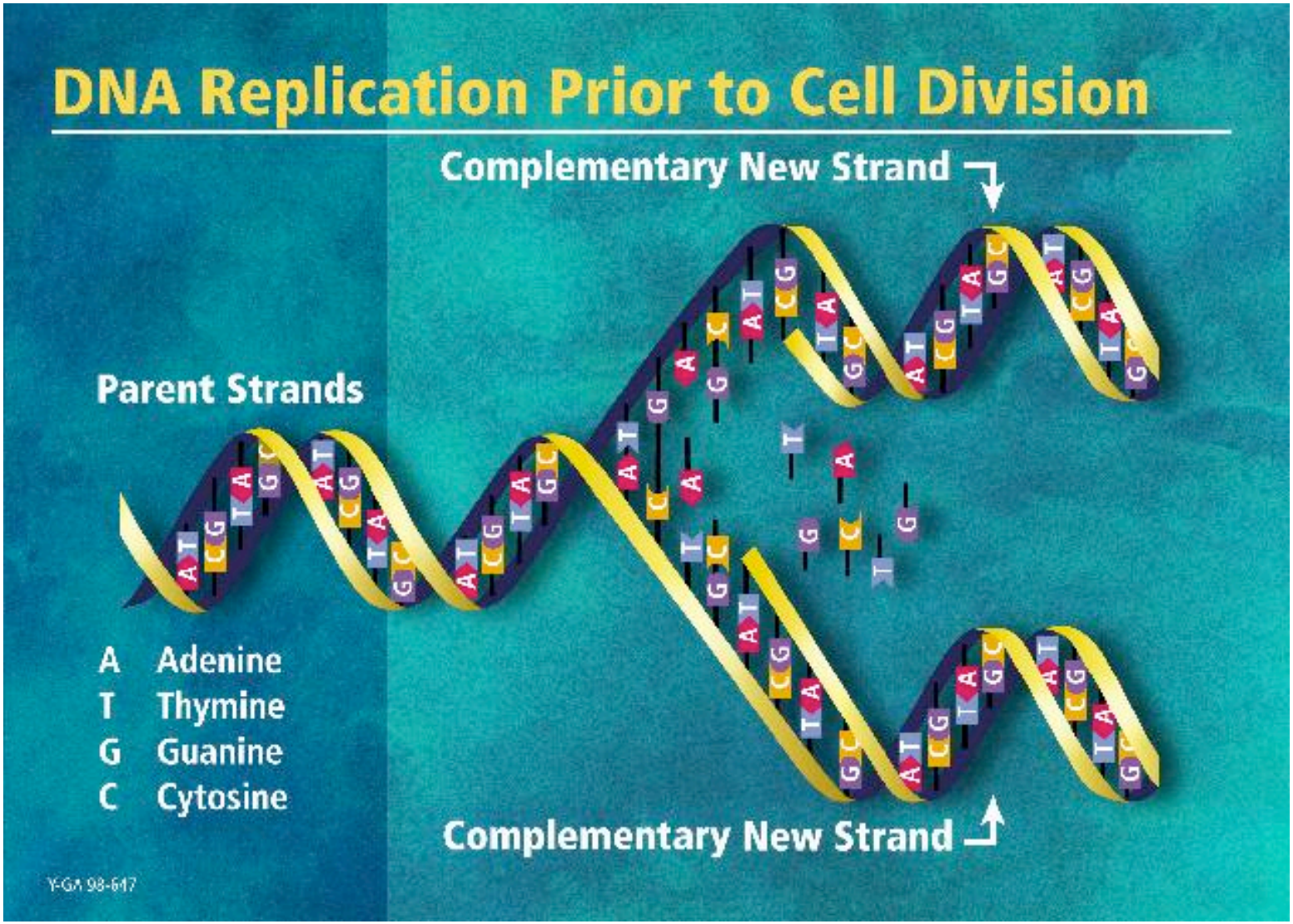


image from the DOE Human Genome Program
http://www.ornl.gov/hgmis

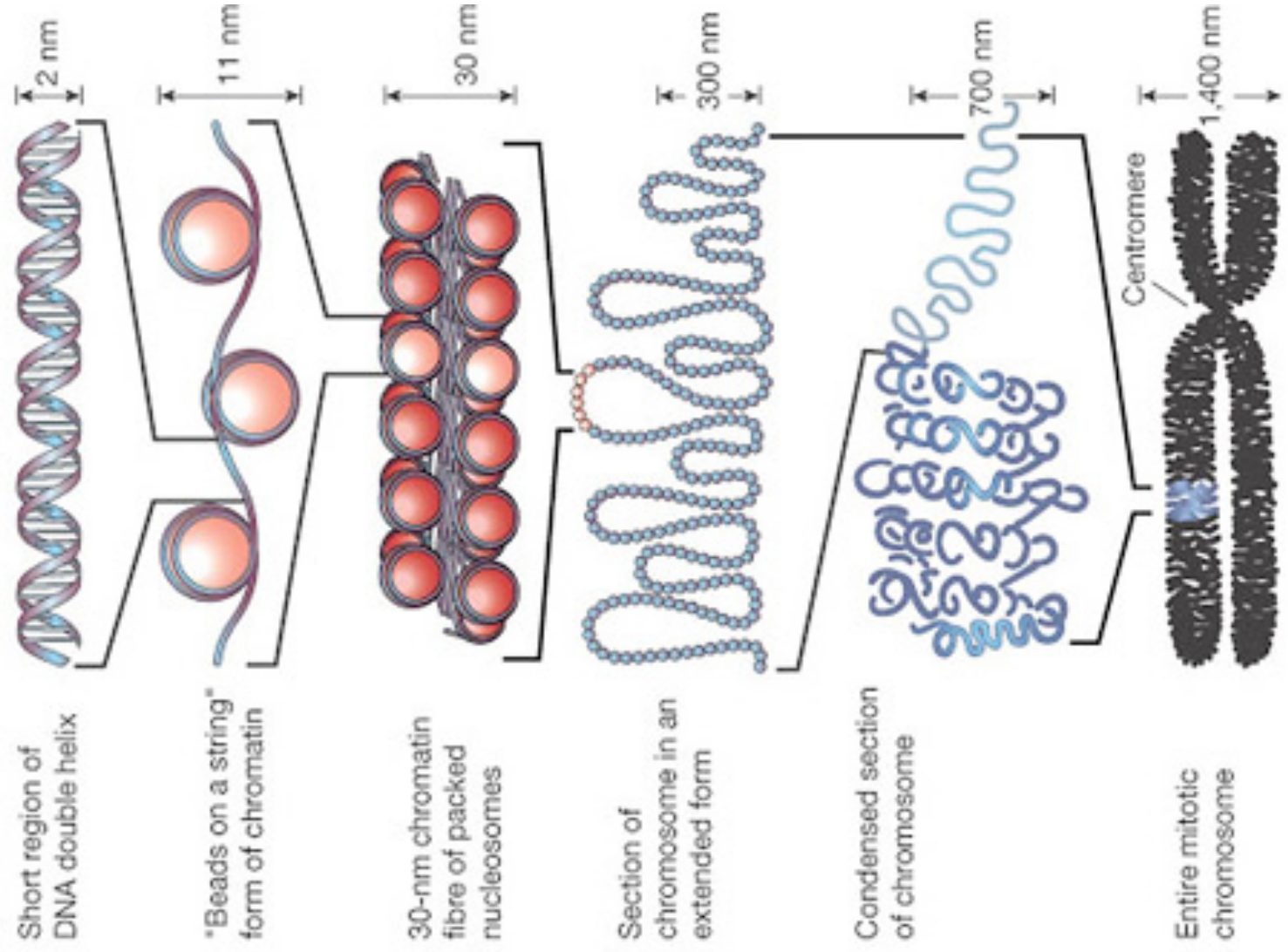
Chromosomes

- DNA is packaged into individual *chromosomes* (along with proteins)

- *prokaryotes* (single-celled organisms lacking nuclei) typically have a single circular chromosome

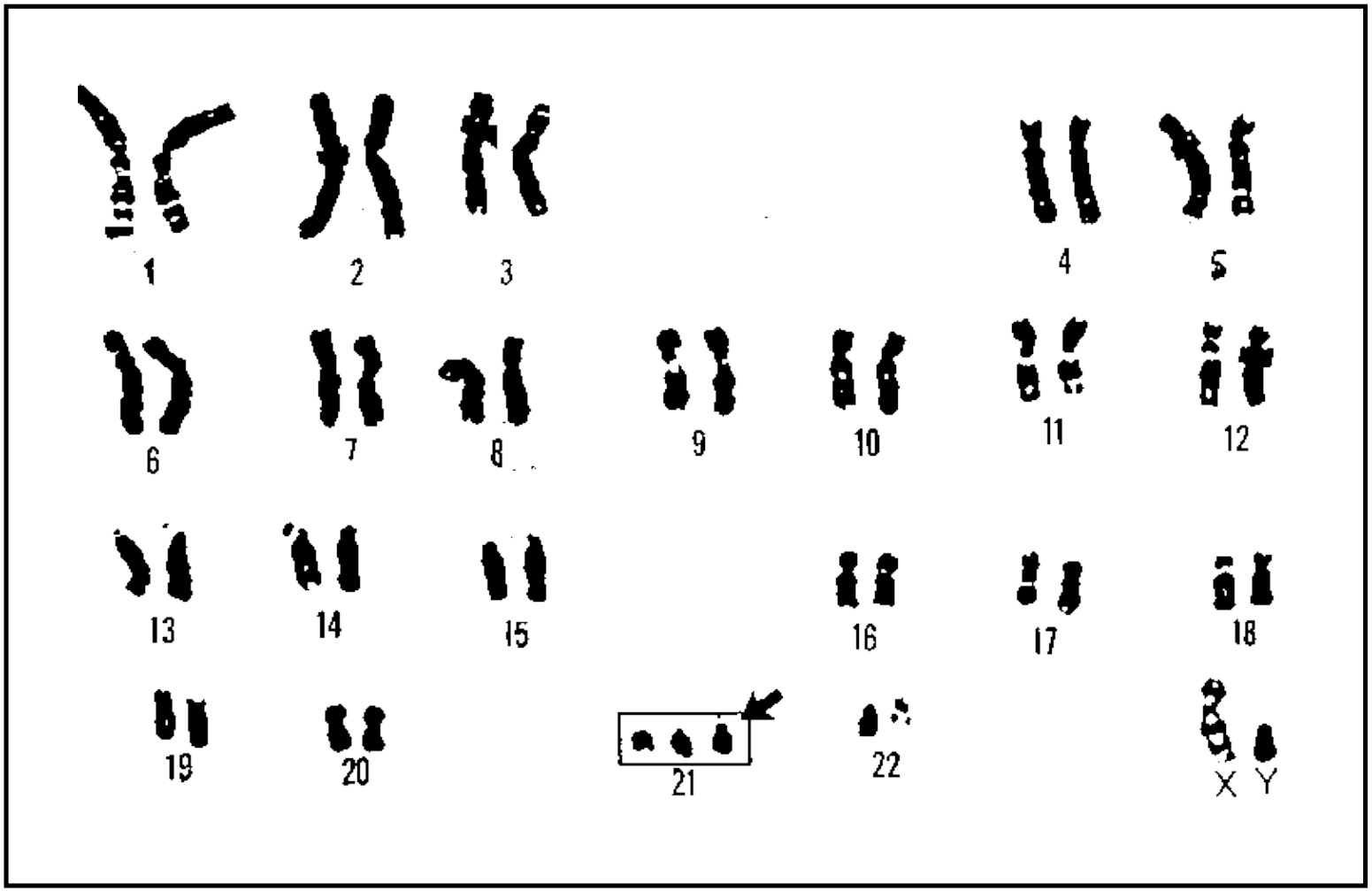
- *eukaryotes* (organisms with nuclei) have a species-specific number of linear chromosomes

The inaccessible code



DNA is tightly packed!

Human Chromosomes



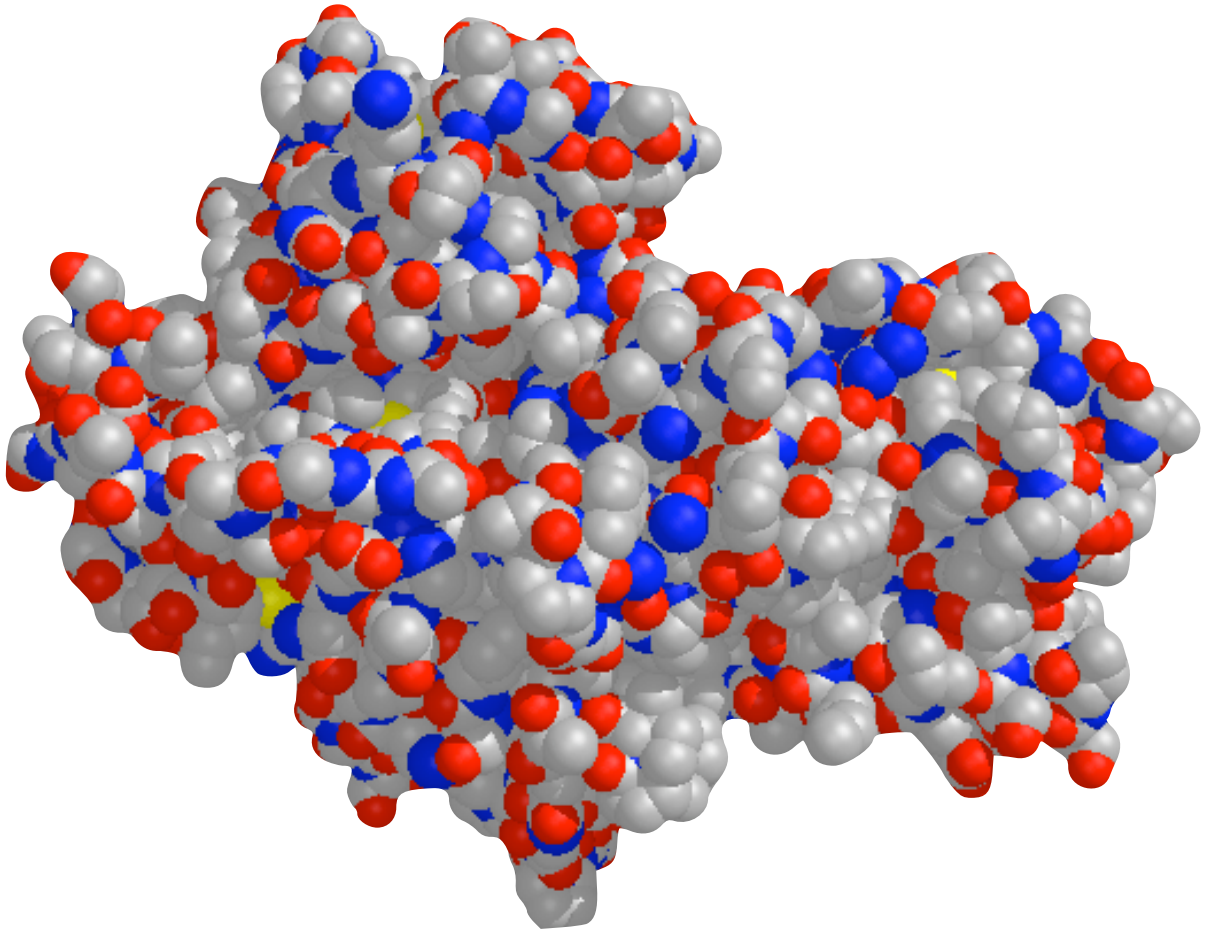
Genomes

- the term *genome* refers to the complete complement of DNA for a given species
- the human genome consists of 46 chromosomes (23 pairs)
- every cell (except sex cells and mature red blood cells) contains the complete genome of an organism

Proteins

- proteins are molecules composed of one or more *polypeptides*
- a polypeptide is a polymer composed of *amino acids*
- cells build their proteins from 20 different amino acids
- a polypeptide can be thought of as a string composed from a 20-character alphabet

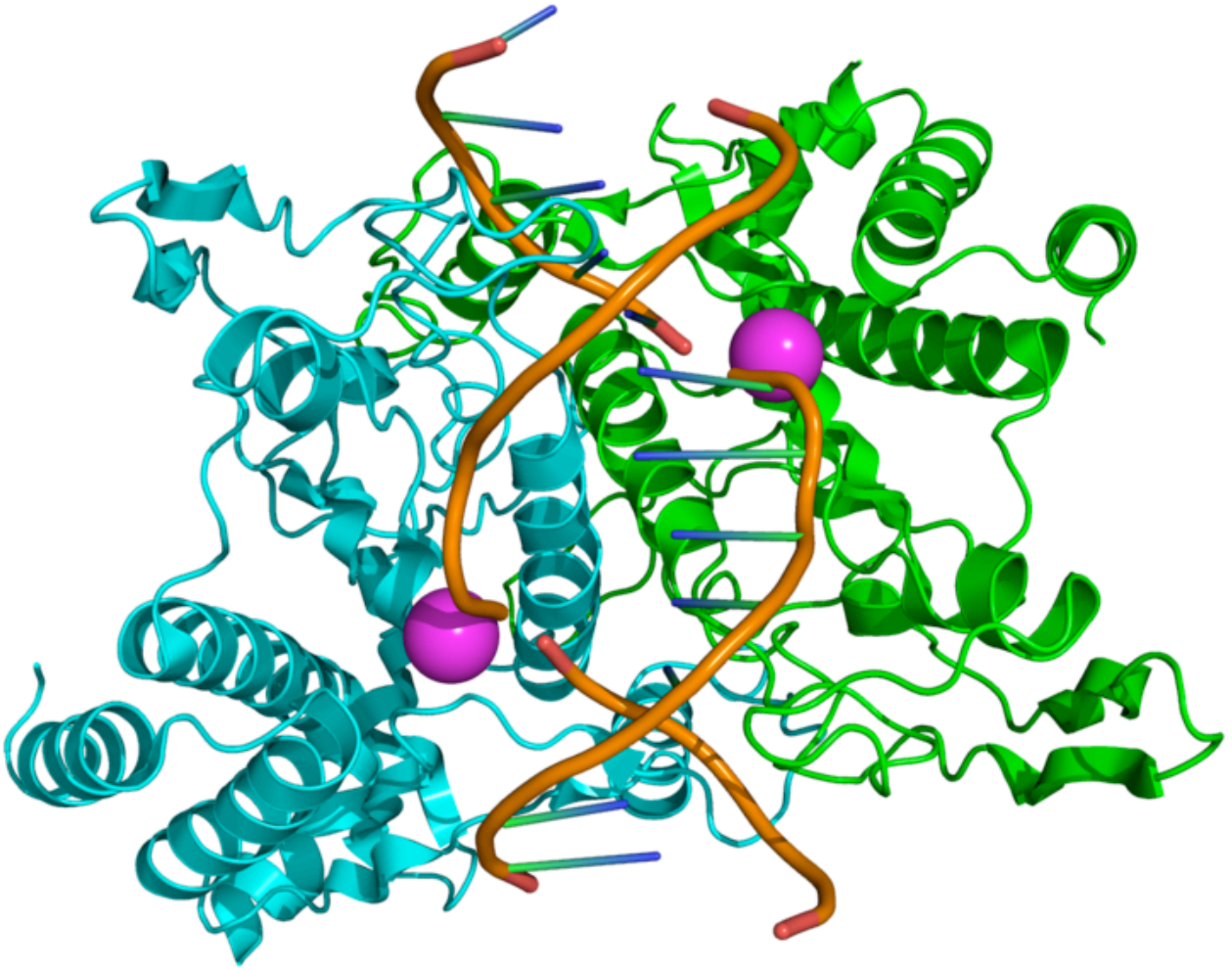
Space-Filling Model of Hexokinase



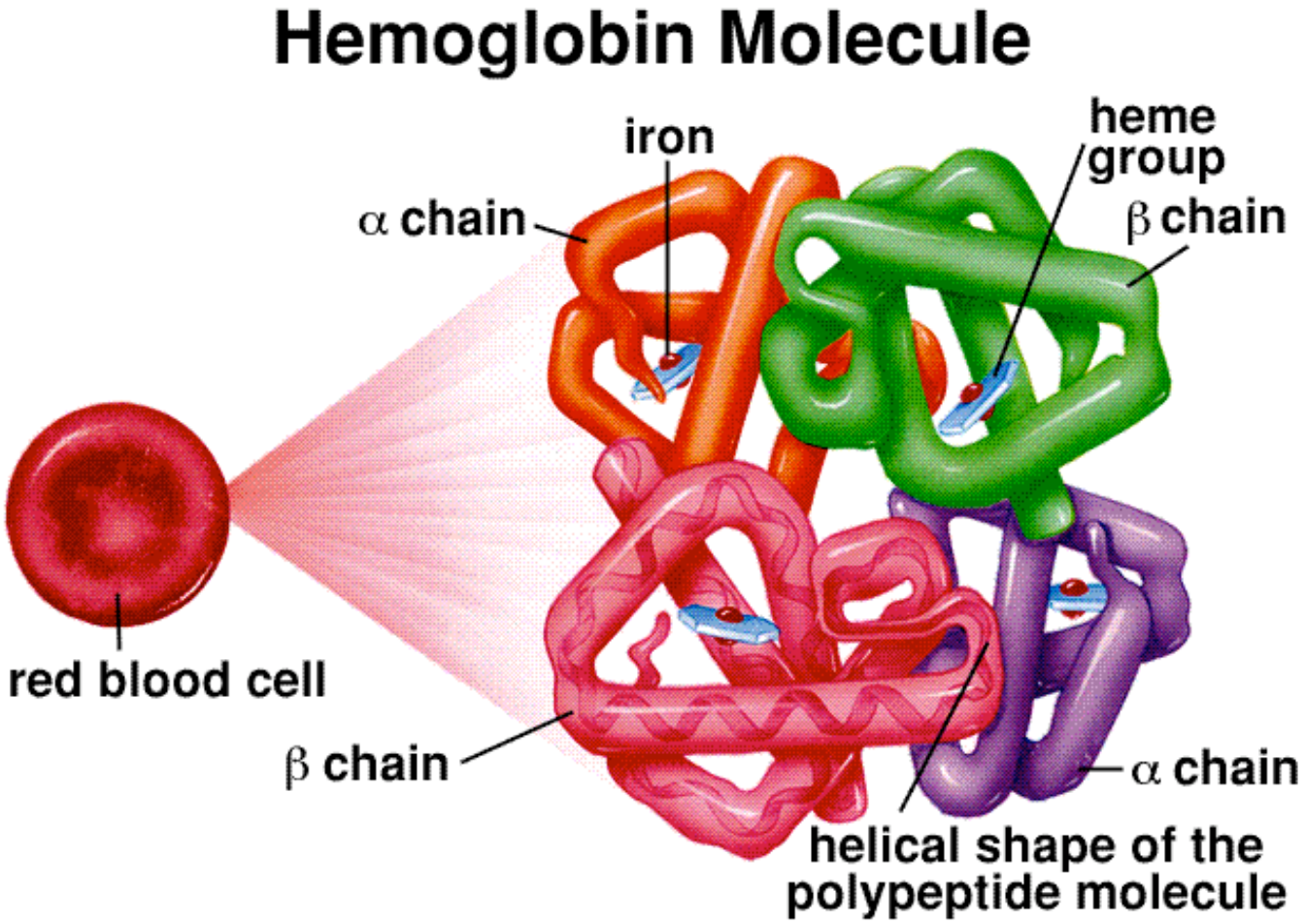
Examples of proteins

Protein	Role
alpha-keratin	component of hair
beta-keratin	component of scales
insulin	regulates blood glucose level
actin & myosin	muscle contraction
DNA polymerase	synthesis of DNA
ATP synthase	makes ATP
hemoglobin	transport of oxygen
endonuclease	cuts DNA (restriction enzyme)

EcoRI - restriction enzyme



Hemoglobin: carrier of oxygen

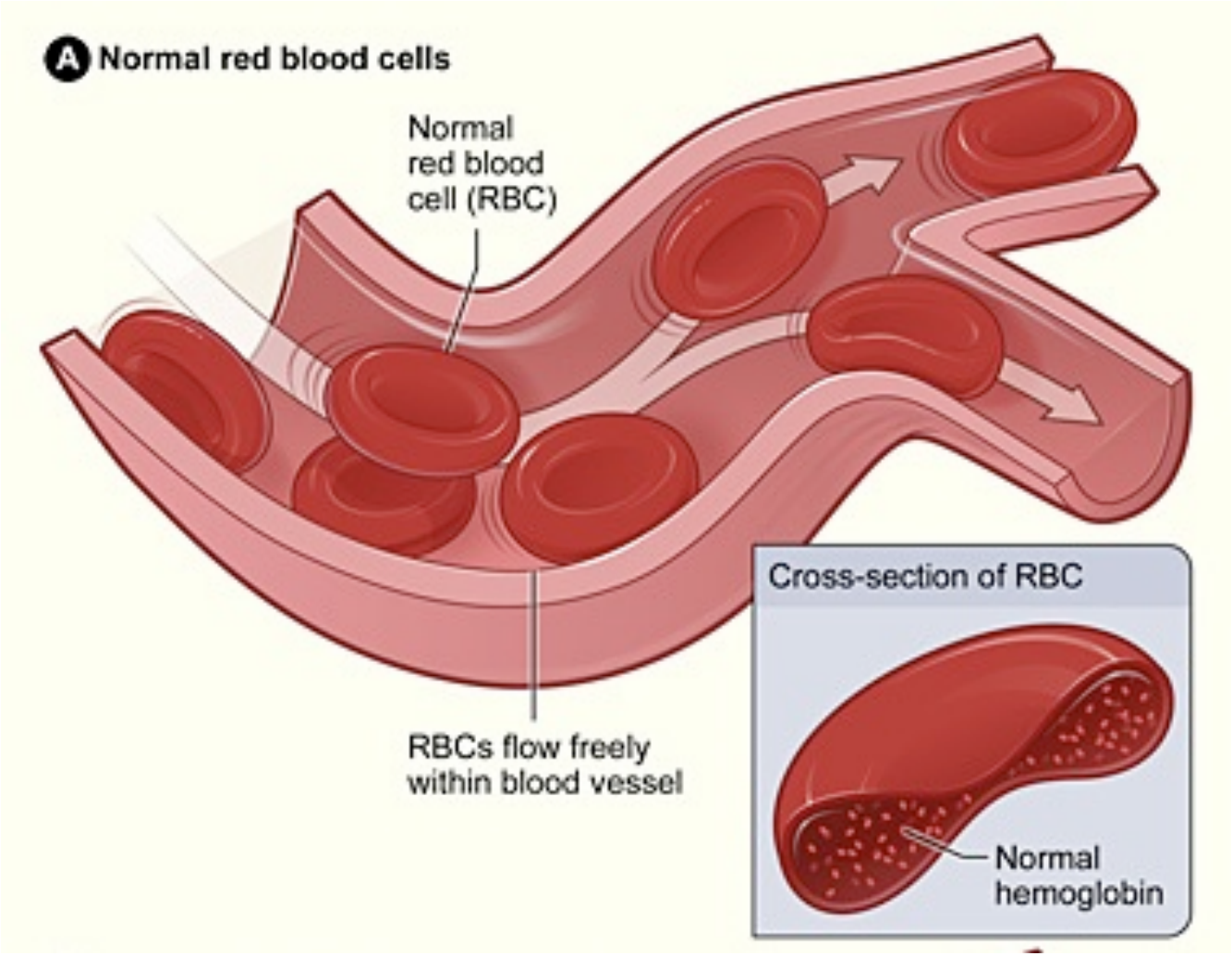


Mutant β-globin → Sickle blood cells

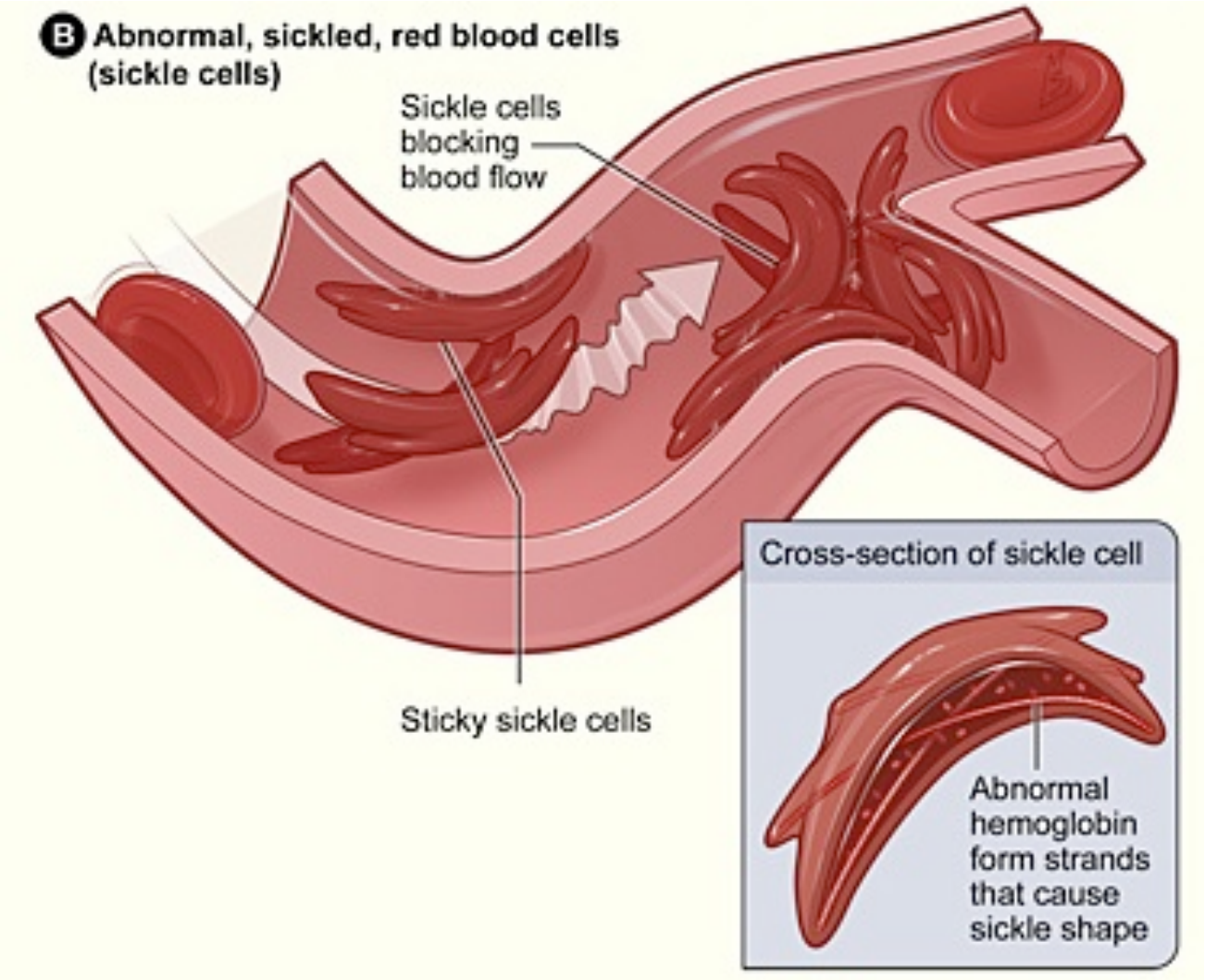
Fiber of sickle hemoglobin

Sickle and normal blood cells

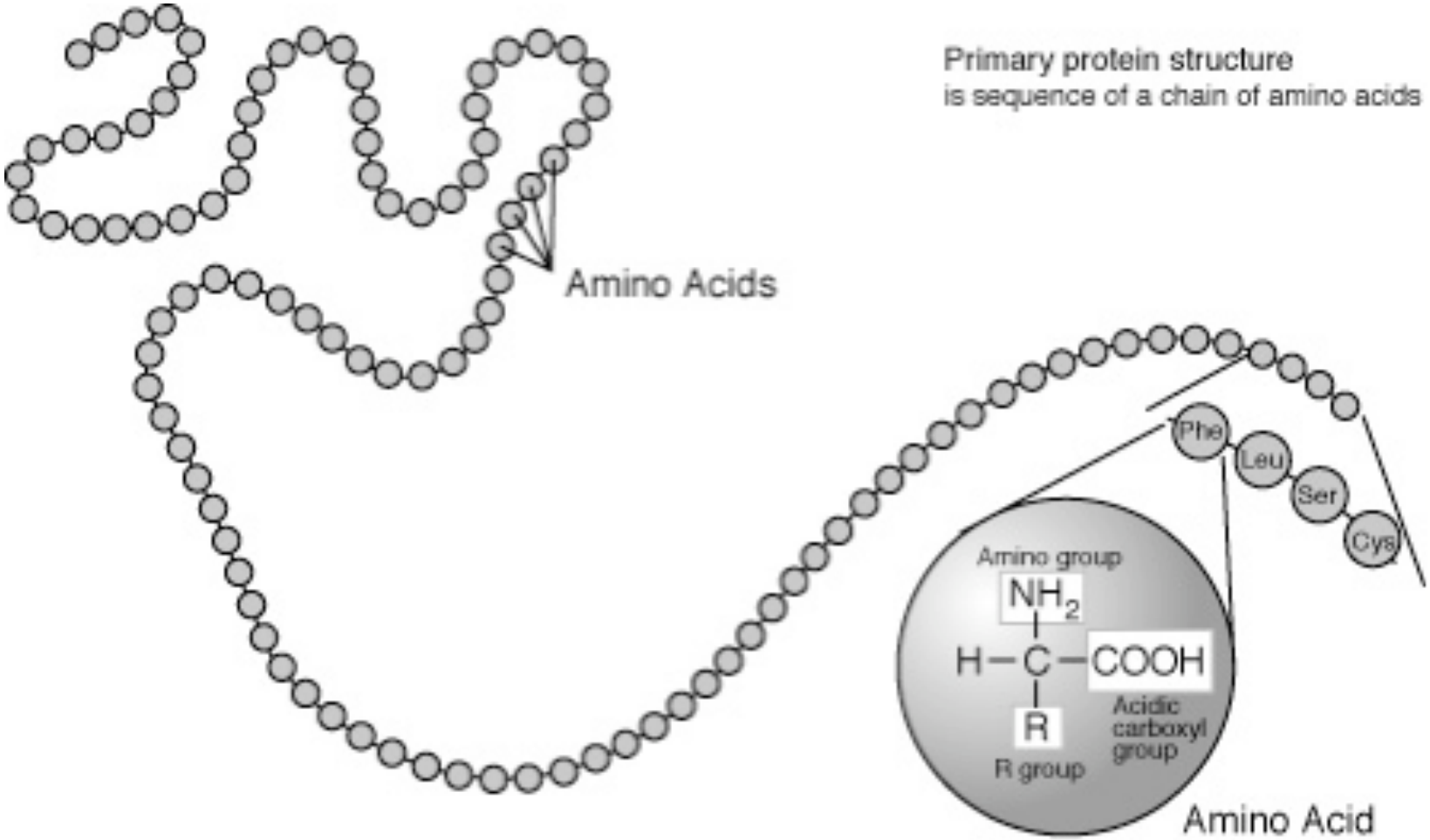
Normal blood flow



Sickle cell complications



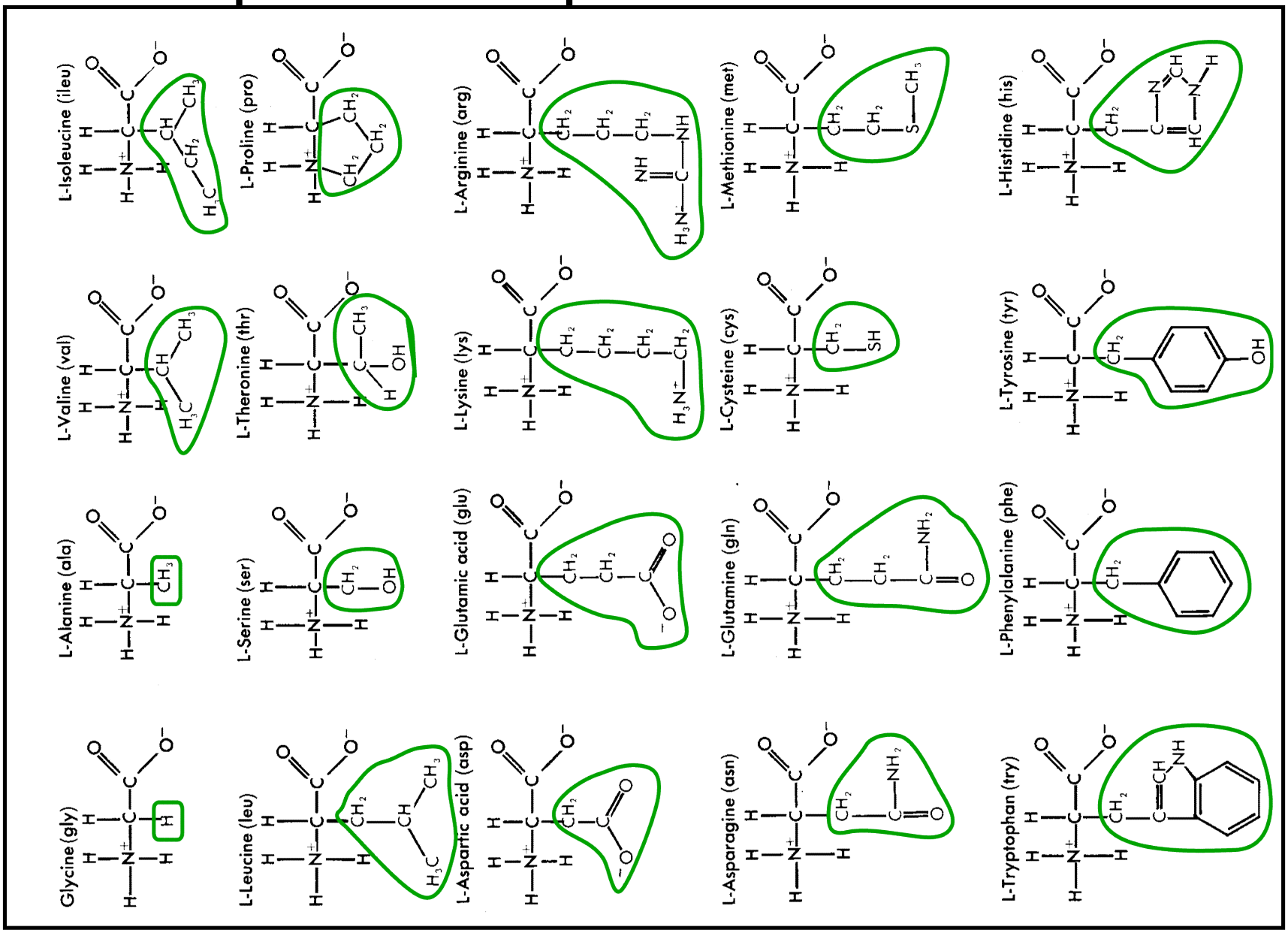
Protein: polymer of amino acids



Amino Acids

Alanine	**Ala**	**A**
Arginine	**Arg**	**R**
Aspartic Acid	**Asp**	**D**
Asparagine	**Asn**	**N**
Cysteine	**Cys**	**C**
Glutamic Acid	**Glu**	**E**
Glutamine	**Gln**	**Q**
Glycine	**Gly**	**G**
Histidine	**His**	**H**
Isoleucine	**Ile**	**I**
Leucine	**Leu**	**L**
Lysine	**Lys**	**K**
Methionine	**Met**	**M**
Phenylalanine	**Phe**	**F**
Proline	**Pro**	**P**
Serine	**Ser**	**S**
Threonine	**Thr**	**T**
Tryptophan	**Trp**	**W**
Tyrosine	**Tyr**	**Y**
Valine	**Val**	**V**

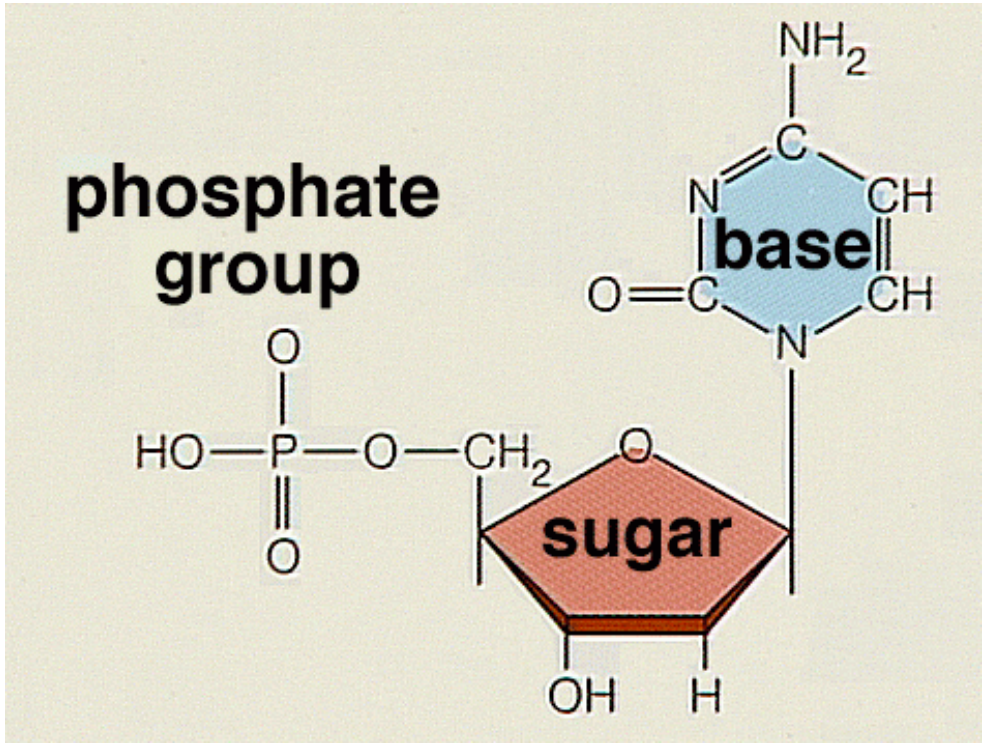
The alphabet of protein: 20 amino acids



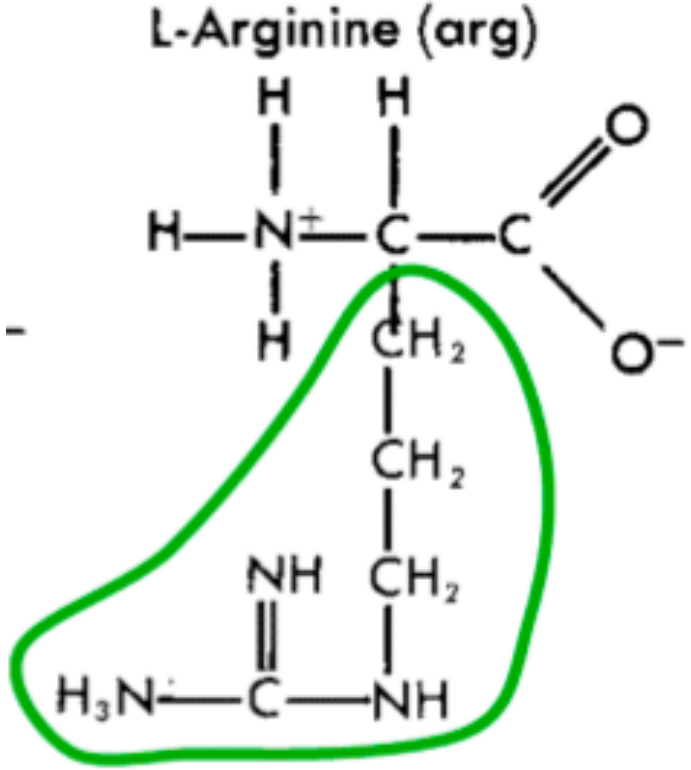
Amino Acid Sequence of Hexokinase

```
          5        10        15        20        25        30
  1 A A S X D X S L V E V H X X V F I V P P X I L Q A V V S I A
 31 T T R X D D X D S A A A S I P M V P G W V L K Q V X G S Q A
 61 G S F L A I V M G G G D L E V I L I X L A G Y Q E S S I X A
 91 S R S L A A S M X T T A I P S D L W G N X A X S N A A F S S
121 X E F S S X A G S V P L G F T F X E A G A K E X V I K G Q I
151 T X Q A X A F S L A X L X K L I S A M X N A X F P A G D X X
181 X X V A D I X D S H G I L X X V N Y T D A X I K M G I I F G
211 S G V N A A Y W C D S T X I A D A A D A G X X G G A G X M X
241 V C C X Q D S F R K A F P S L P Q I X Y X X T L N X X S P X
271 A X K T F E K N S X A K N X G Q S L R D V L M X Y K X X G Q
301 X H X X X A X D F X A A N V E N S S Y P A K I Q K L P H F D
331 L R X X X D L F X G D Q G I A X K T X M K X V V R R X L F L
361 I A A Y A F R L V V C X I X A I C Q K K G Y S S G H I A A X
391 G S X R D Y S G F S X N S A T X N X N I Y G W P Q S A X X S
421 K P I X I T P A I D G E G A A X X V I X S I A S S Q X X X A
451 X X S A X X A
```

Nucleotides vs. Amino Acids



Nucleotide



Amino Acid

Both made up of "backbone" and "residue" parts

Genes

- genes are the basic units of heredity
- a gene is a sequence of bases that carries the information required for constructing a particular protein (polypeptide really)
- such a gene is said to *encode* a protein
- the human genome comprises ~25,000 protein-coding genes

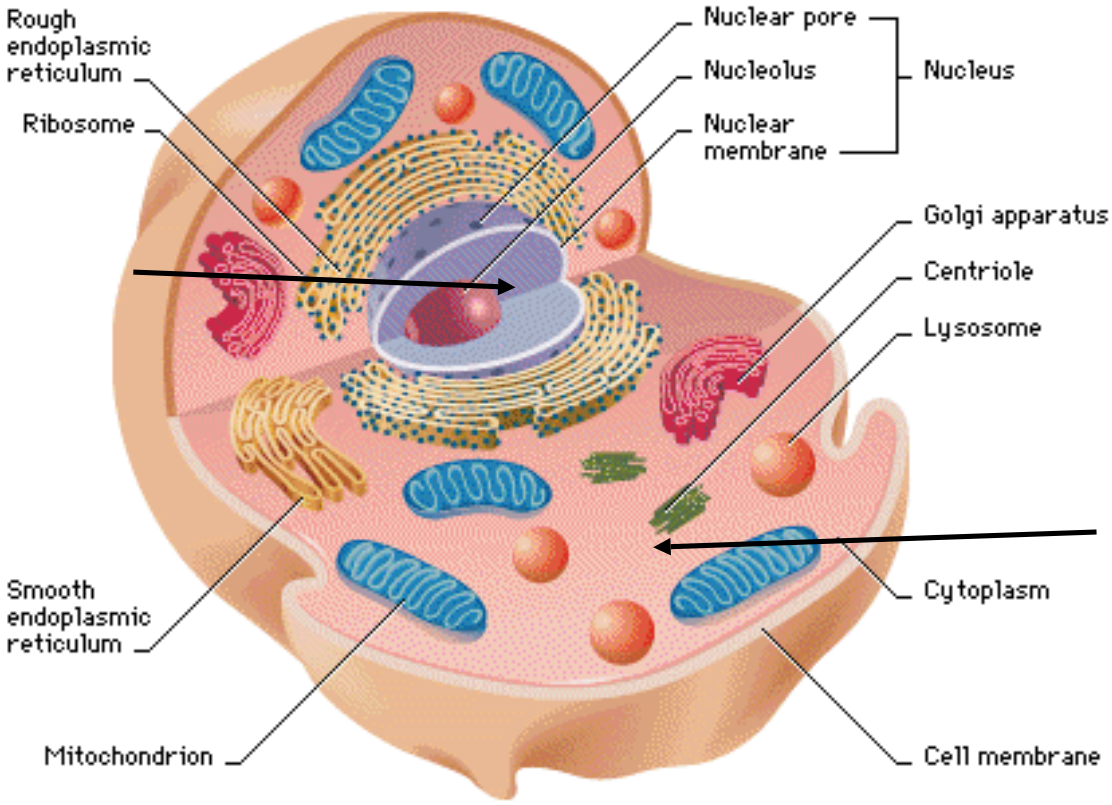
Gene Density

- not all of the DNA in a genome encodes protein:

bacteria	~90% coding gene/kb
human	~1.5% coding gene/35kb

The inaccessible code

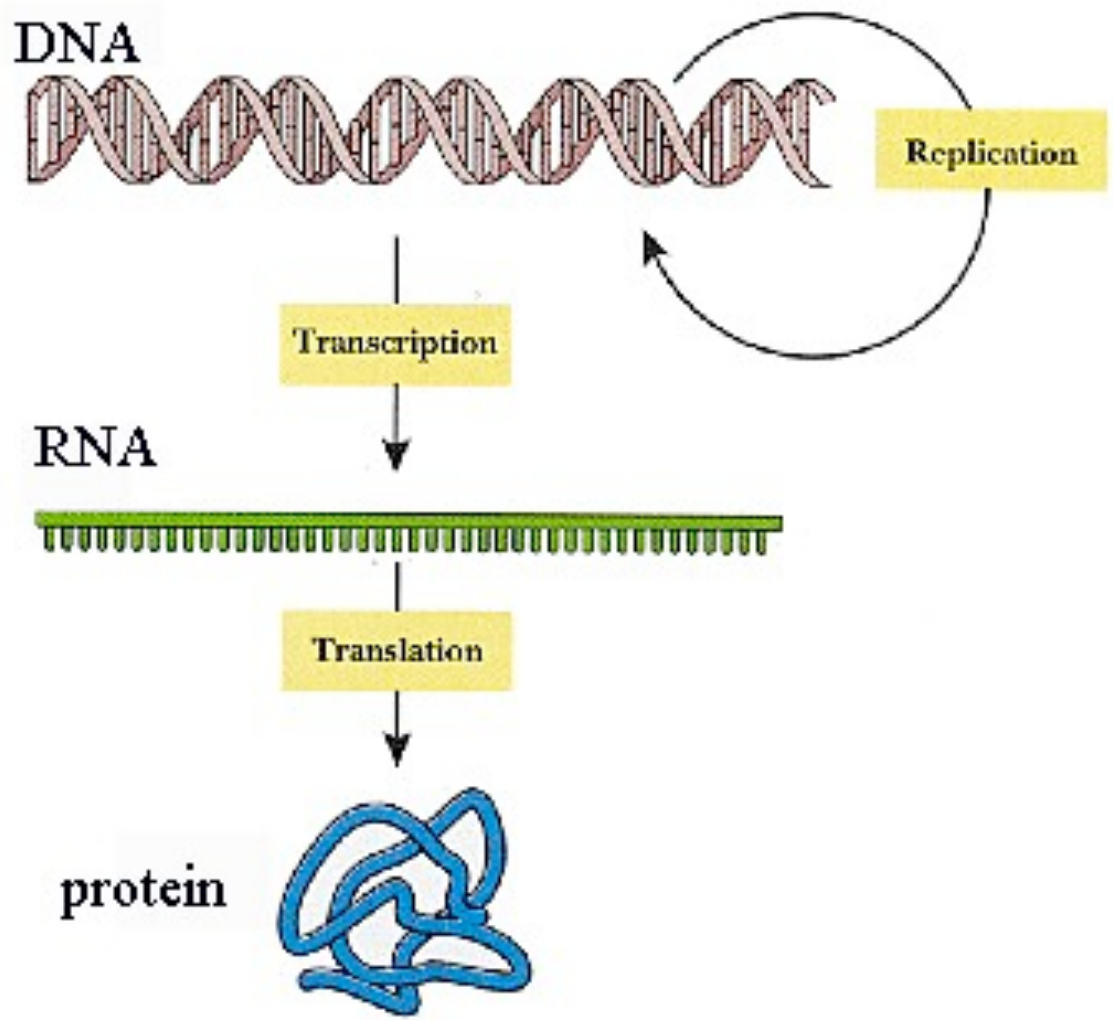
DNA is in the nucleus



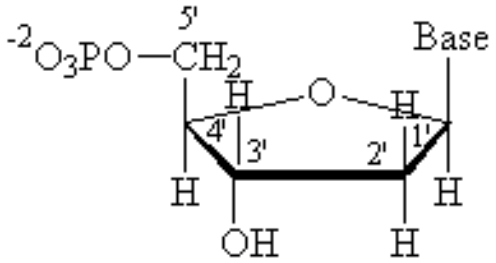
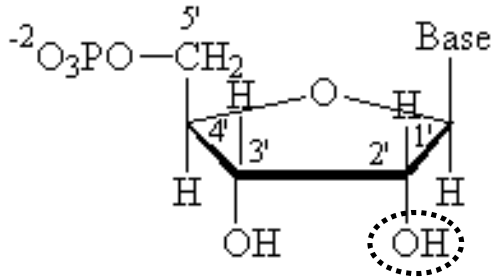
Proteins are (mostly made in the cytoplasm

(eukaryotic cell)

The Central Dogma



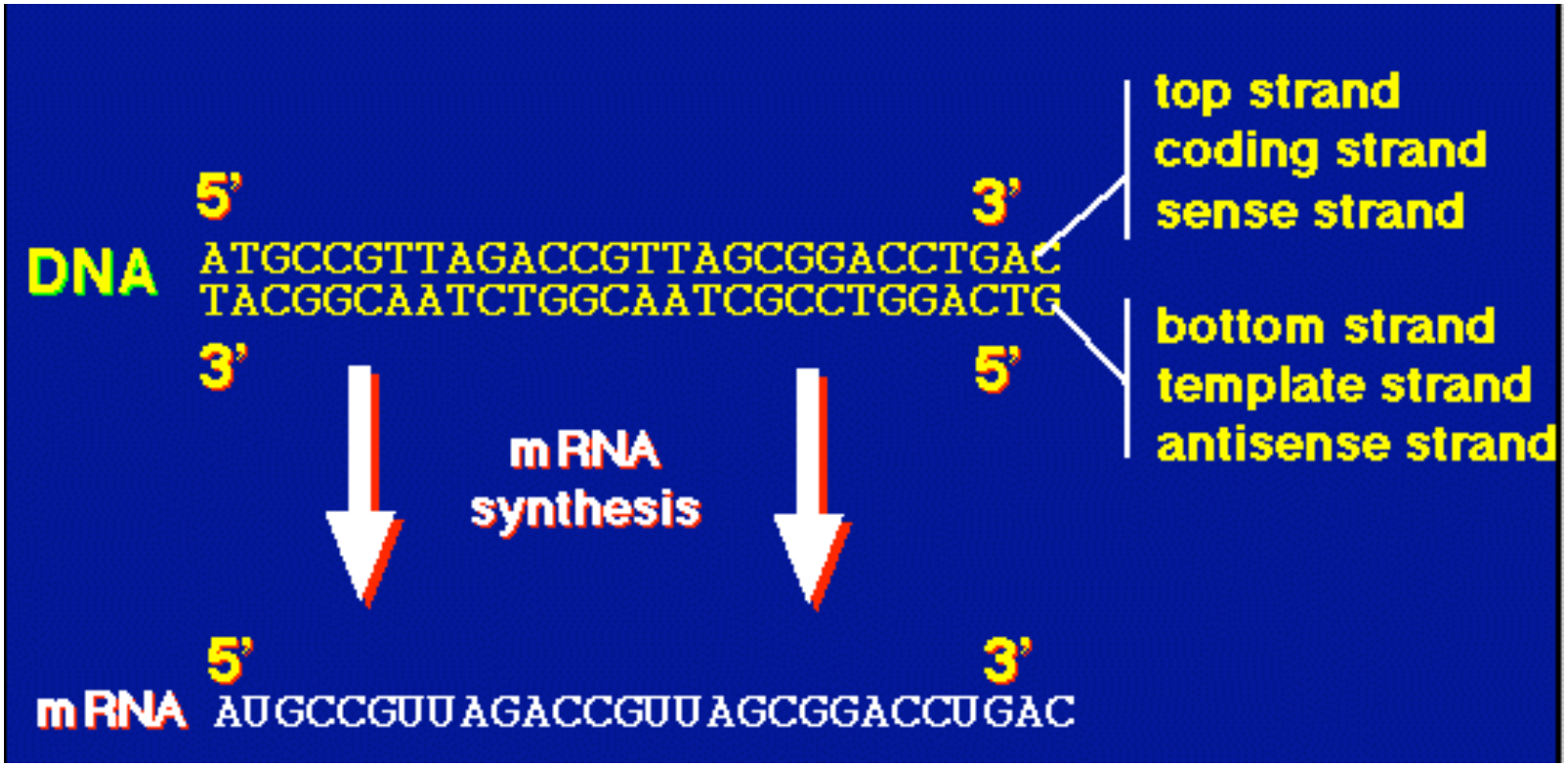
RNA vs. DNA structure

DNA	RNA
linear polymer	linear polymer
double-stranded	single-stranded
deoxyribonucleotide monomer 	ribonucleotide monomer 
A,C,G,T bases	A,C,G,U bases

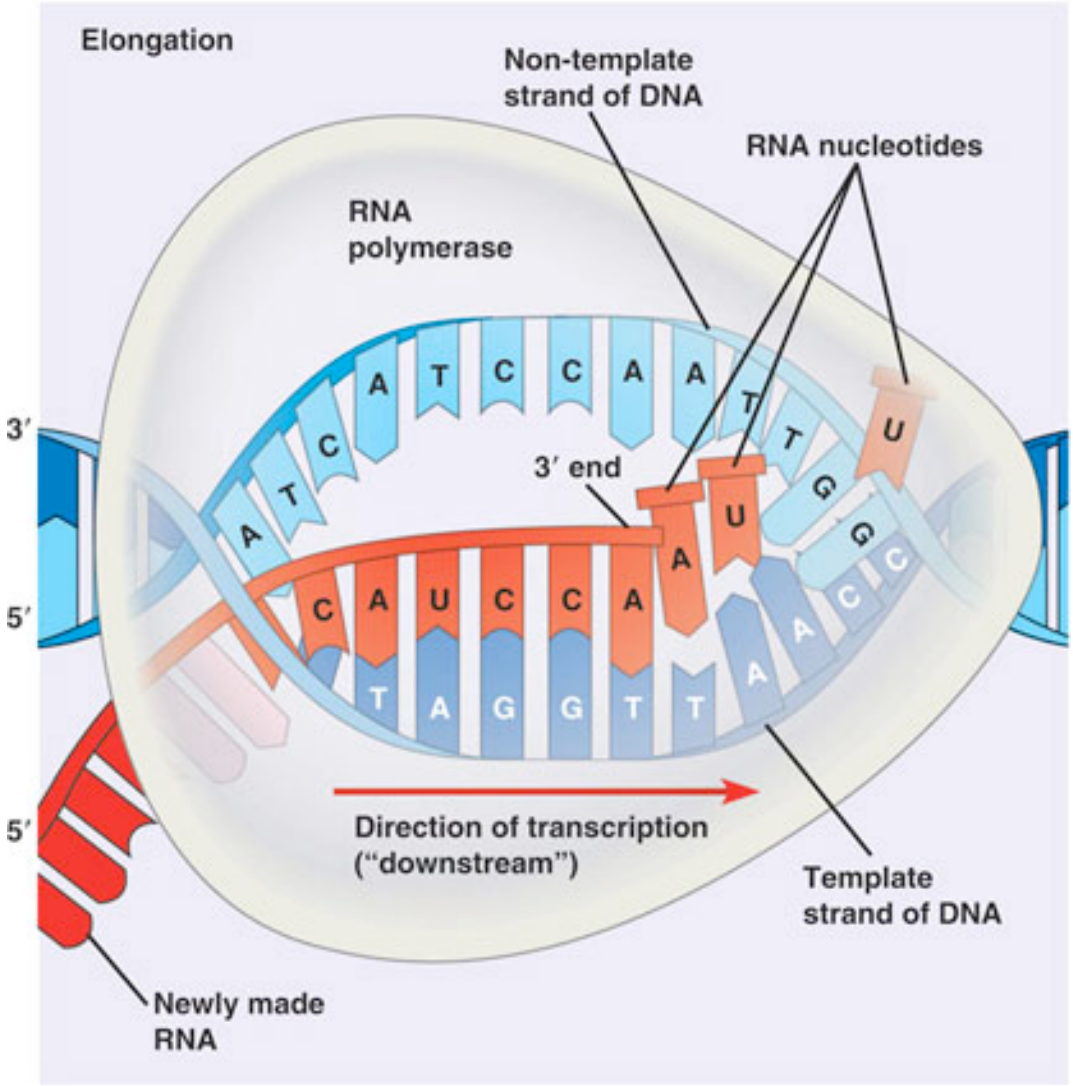
RNA

- RNA is like DNA except:
  - backbone is a little different
  - often single stranded
  - the base uracil (U) is used in place of thymine (T)
- a strand of RNA can be thought of as a string composed of the four letters: A, C, G, U

Transcription



Transcription: DNA→RNA

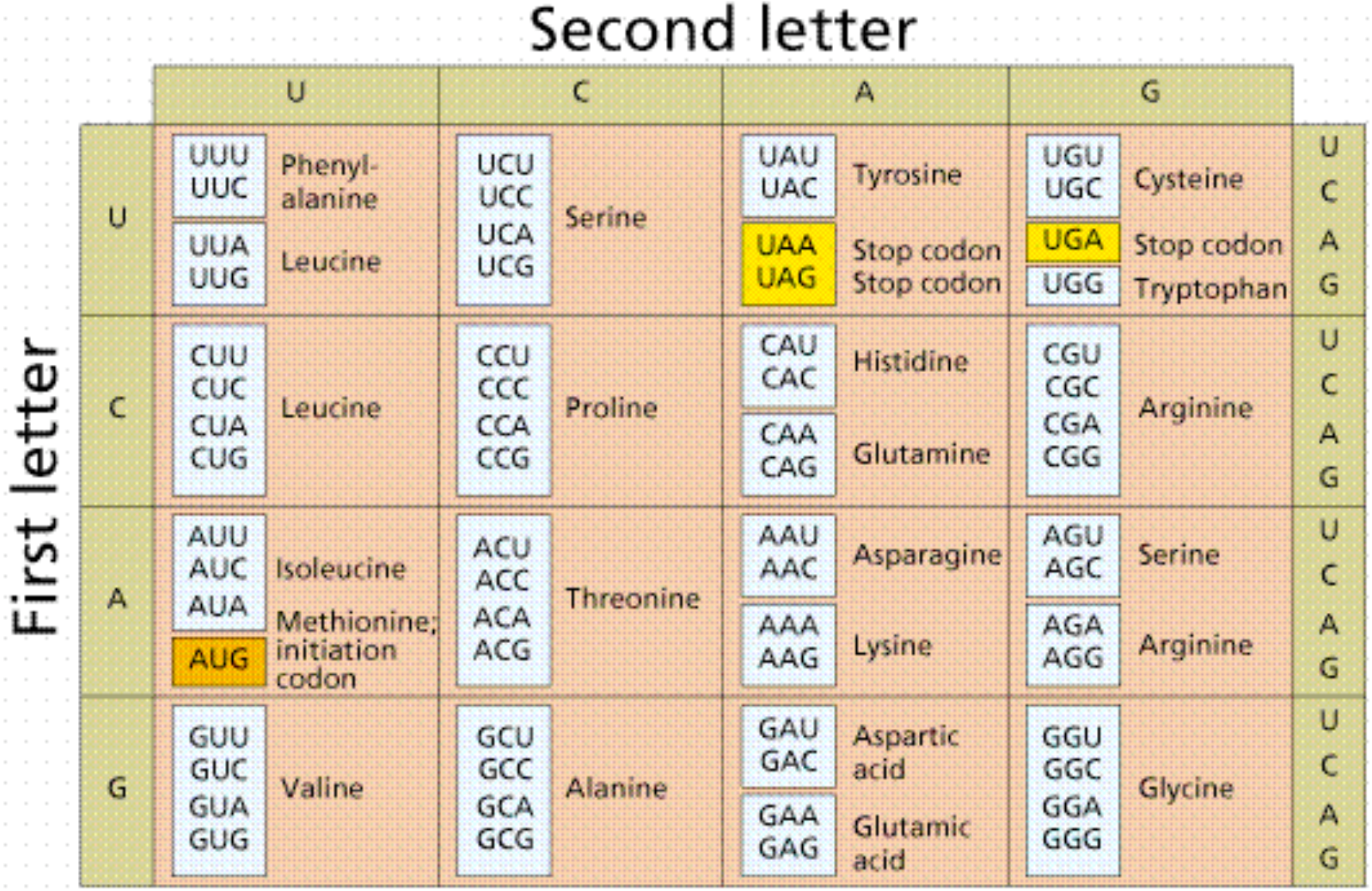


Transcription

- *RNA polymerase* is the enzyme that builds an RNA strand from a gene

  - RNA that is transcribed from a gene is called *messenger RNA* (mRNA)

The Genetic Code



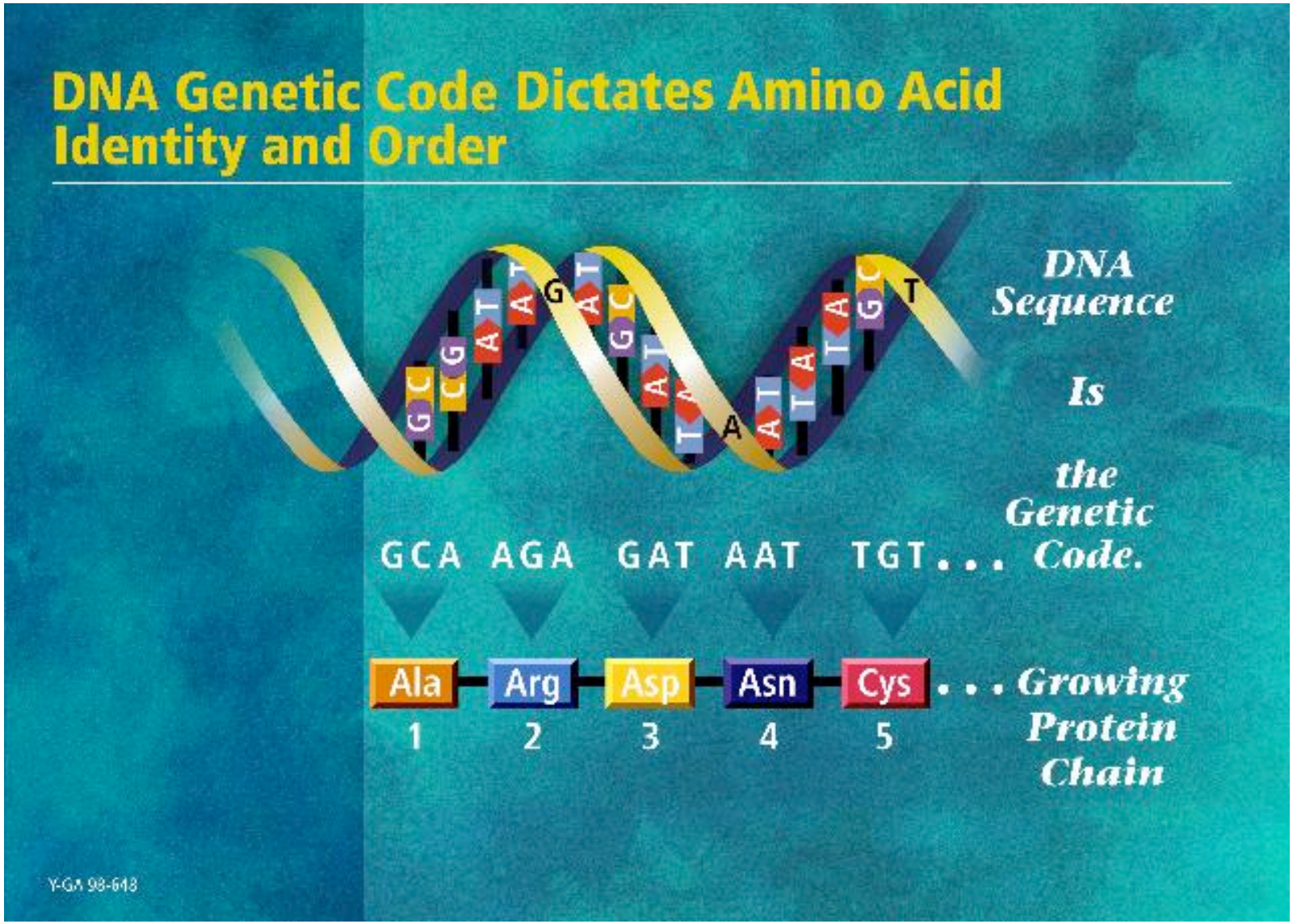
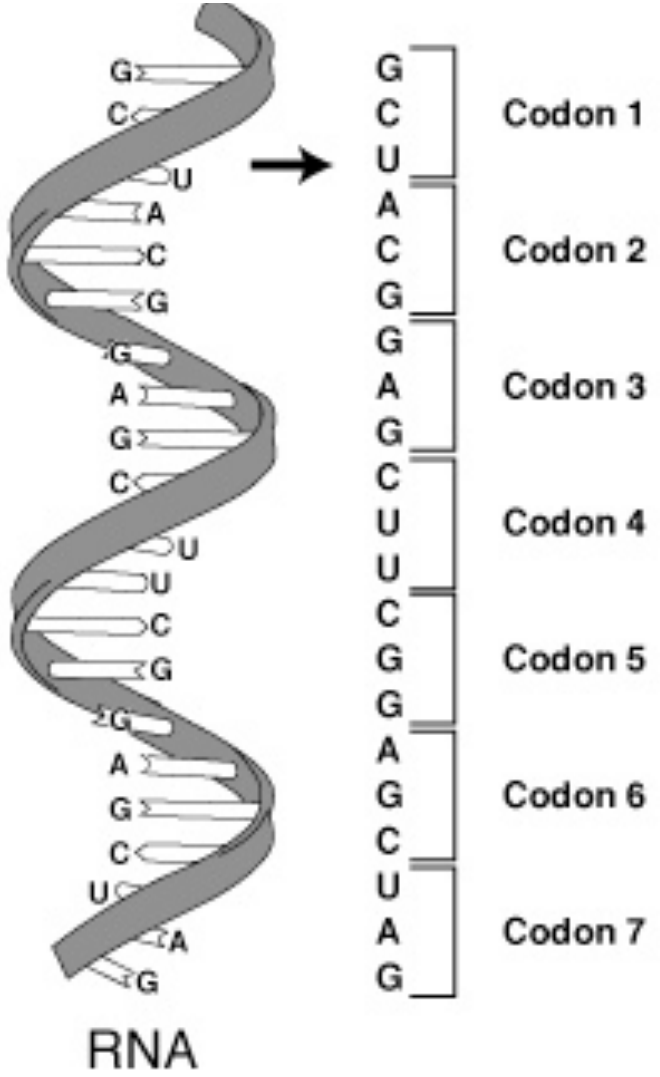


image from the DOE Human Genome Program
http://www.ornl.gov/hgmis

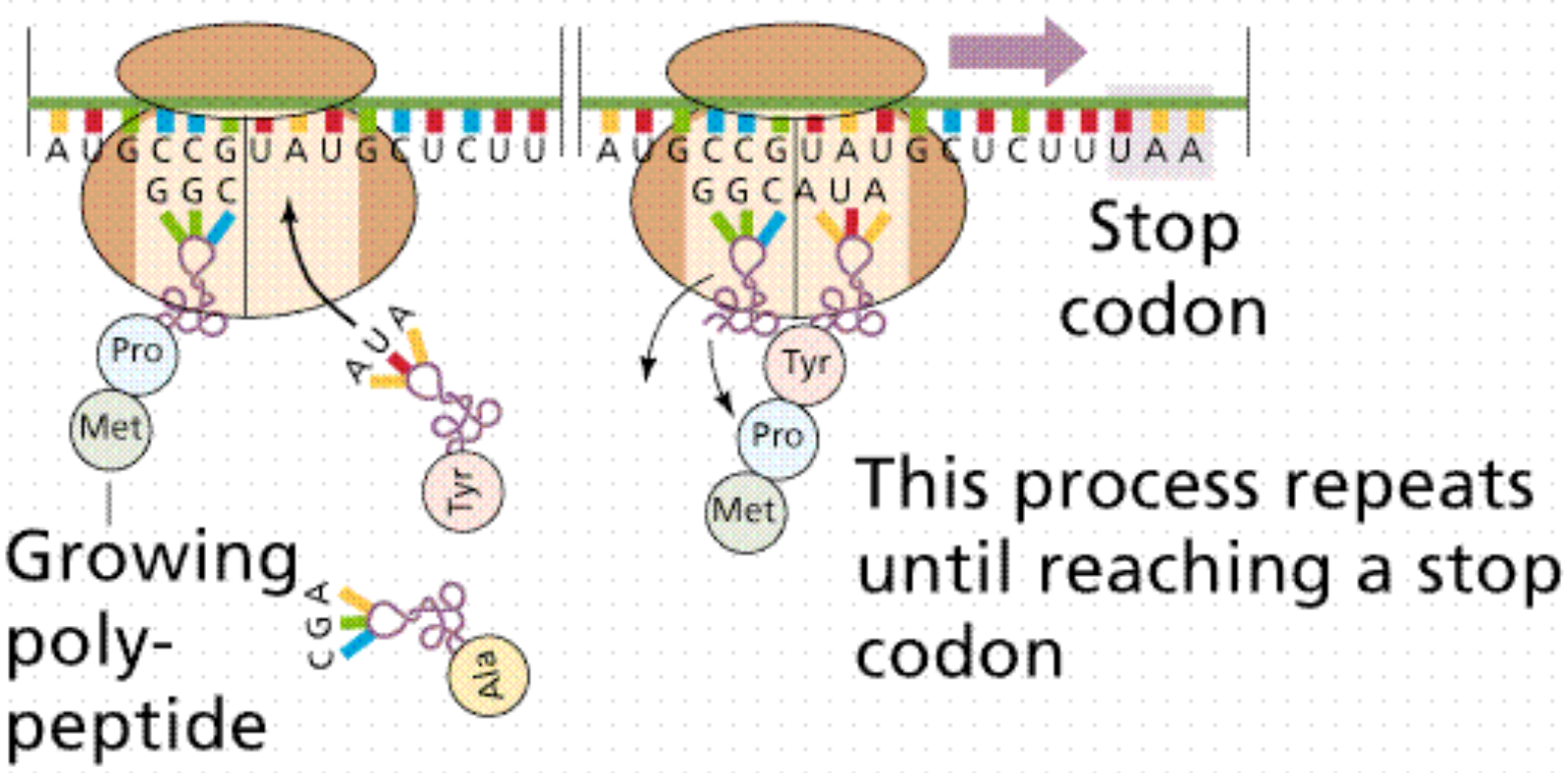
Translation

- *ribosomes* are the machines that synthesize proteins from mRNA
  - the grouping of codons is called the *reading frame*
  - translation begins with the *start codon*
  - translation ends with the *stop codon*

Codons and Reading Frames



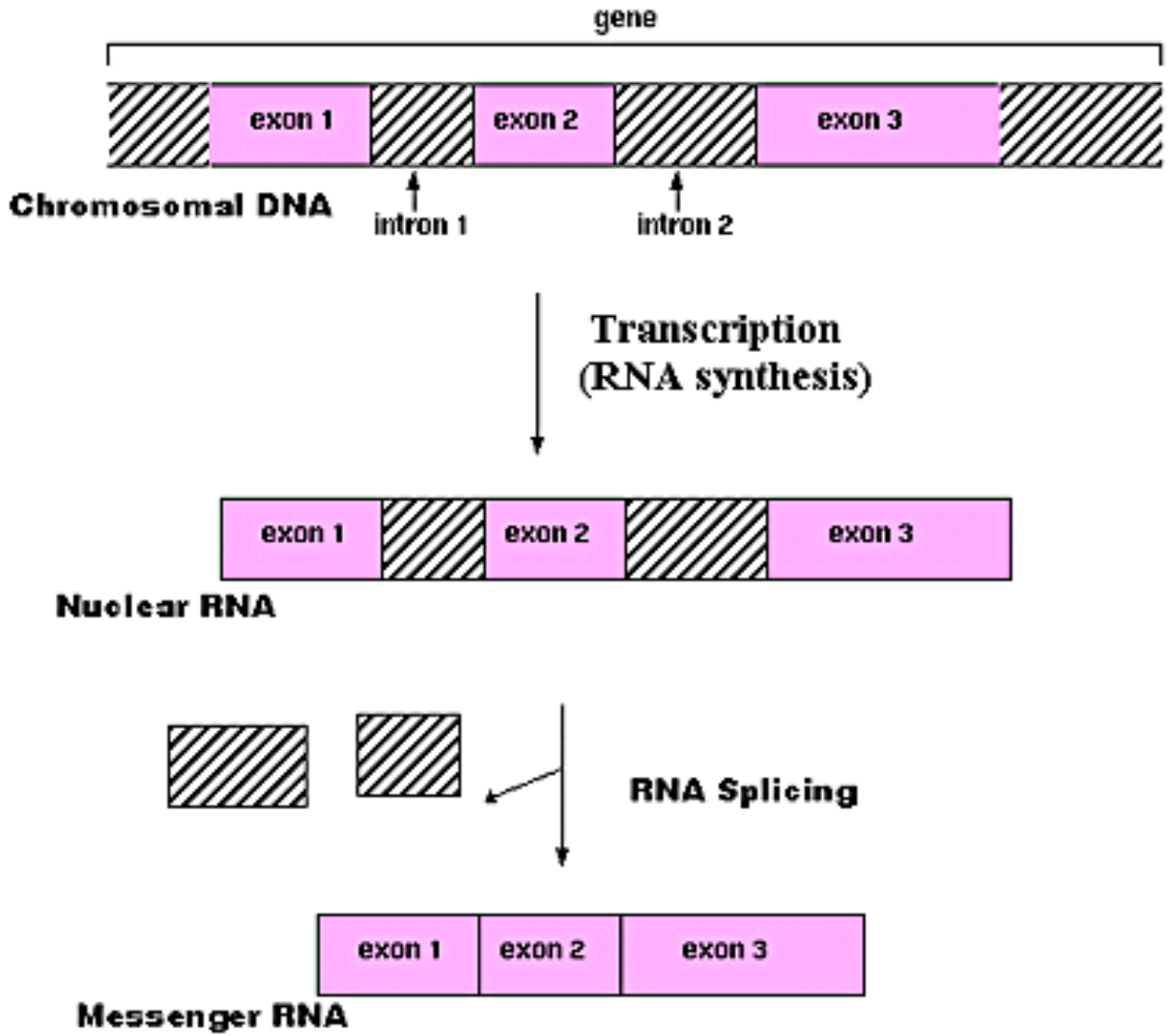
Translation



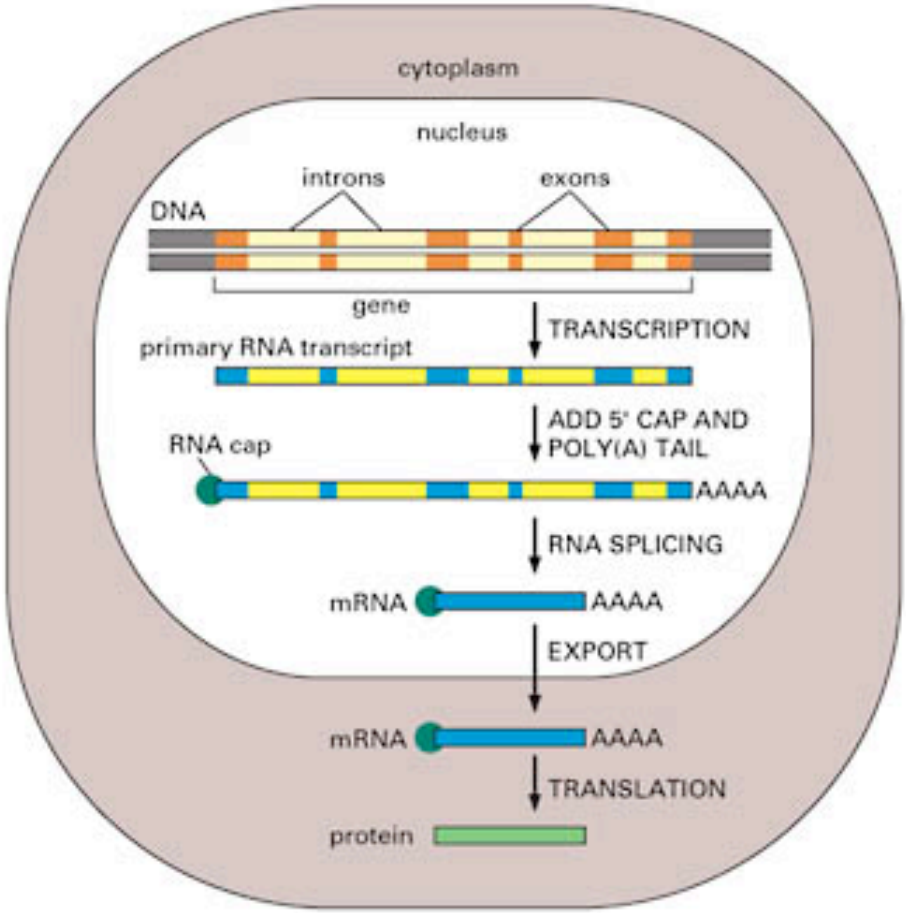
RNA Processing in Eukaryotes

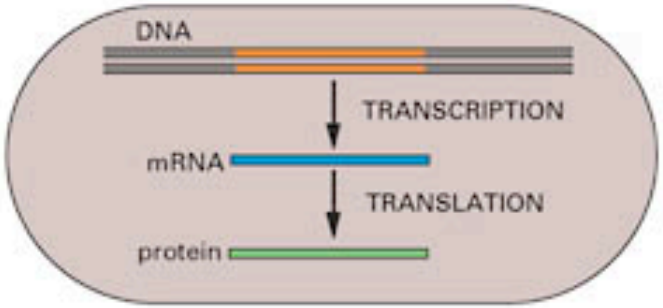
- *eukaryotes* are organisms that have enclosed nuclei in their cells
  - in many eukaryotes, genes/mRNAs consist of alternating *exon/intron* segments
  - exons are the coding parts
  - introns are spliced out before translation

RNA Splicing



Protein Synthesis in Eukaryotes vs. Prokaryotes





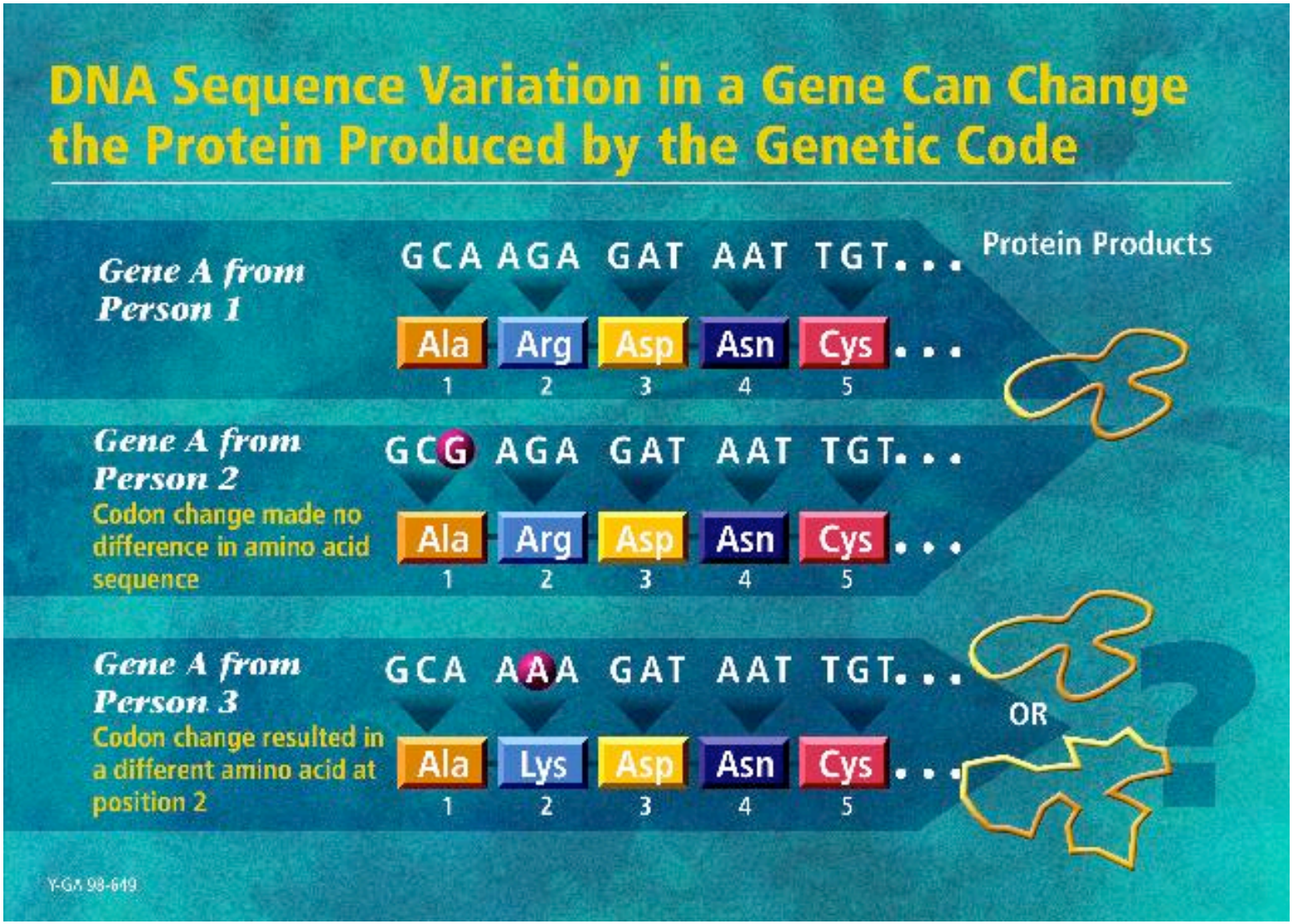


image from the DOE Human Genome Program
http://www.ornl.gov/hgmis

RNA Genes

- not all genes encode proteins
- for some genes the end product is RNA
  - *ribosomal RNA* (rRNA), which includes major constituents of ribosomes
  - *transfer RNAs* (tRNAs), which carry amino acids to ribosomes
  - *micro RNAs* (miRNAs), which play an important regulatory role in various plants and animals
    - etc.

The Dynamics of Cells

- all cells in an organism have the same genomic data, but the genes expressed in each vary according to cell type, time, and environmental factors
- there are networks of interactions among various biochemical entities in a cell (DNA, RNA, protein, small molecules) that carry out processes such as
  - metabolism
  - intra-cellular and inter-cellular signaling
  - regulation of gene expression

Overview of the E. coli Metabolic Pathway Map

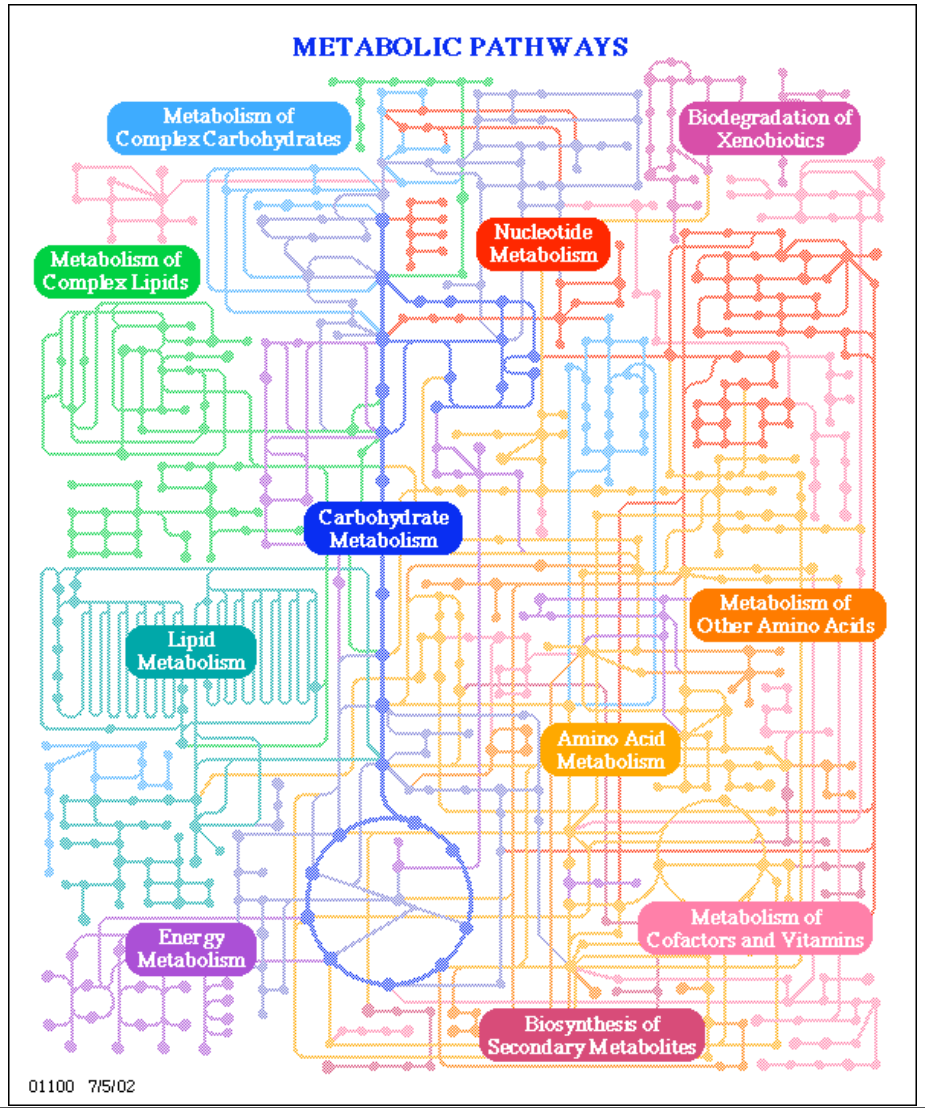
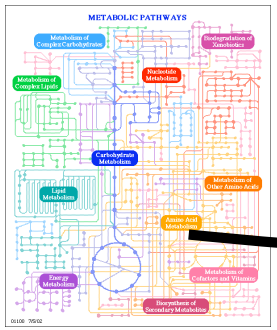


image from the KEGG database

The Metabolic Pathway for Synthesizing the Amino Acid Alanine



→ reactions

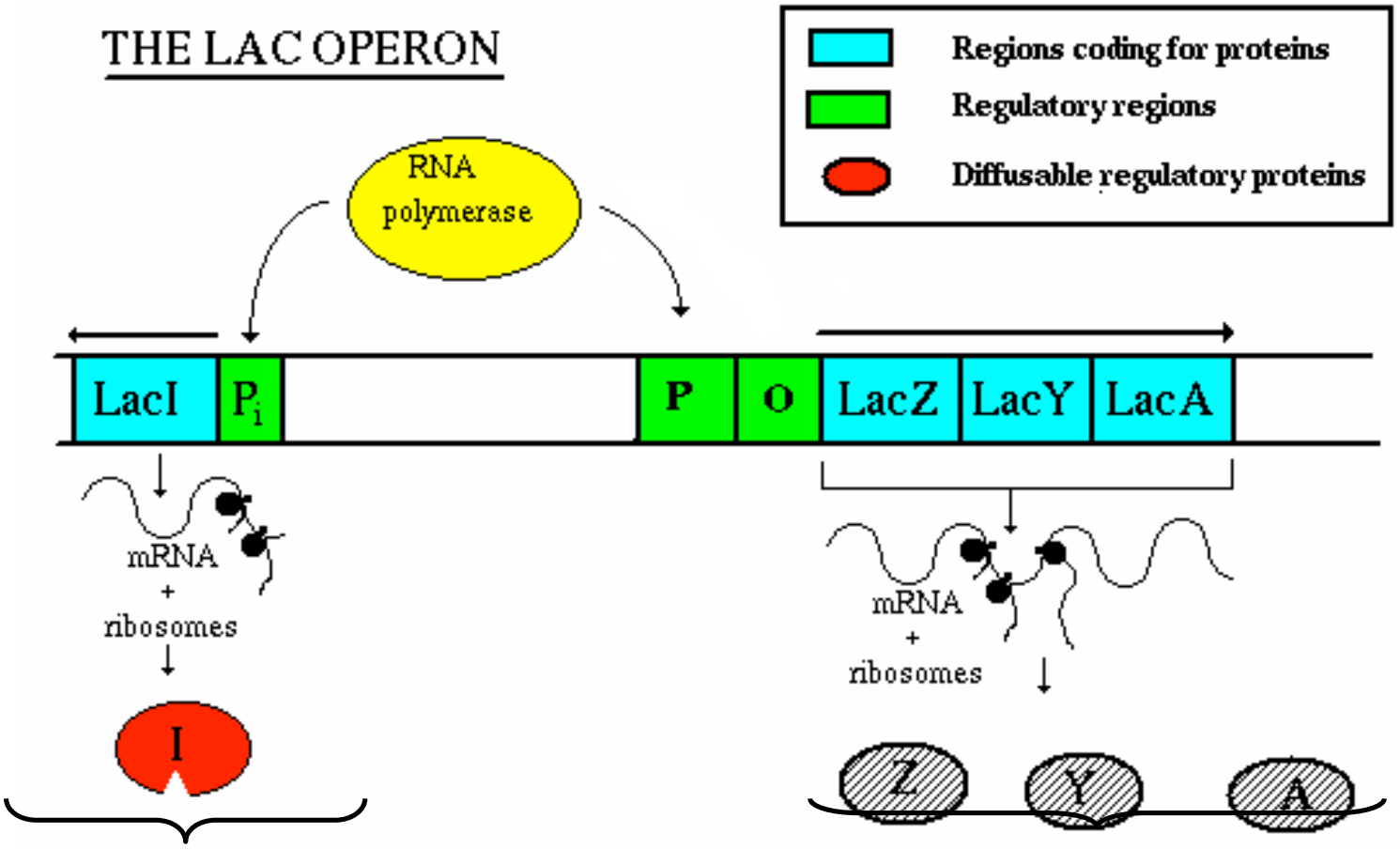
metabolites

enzymes (proteins that catalyze reactions)

genes encoding the enzymes

image from the Ecocyc database
www.biocyc.org

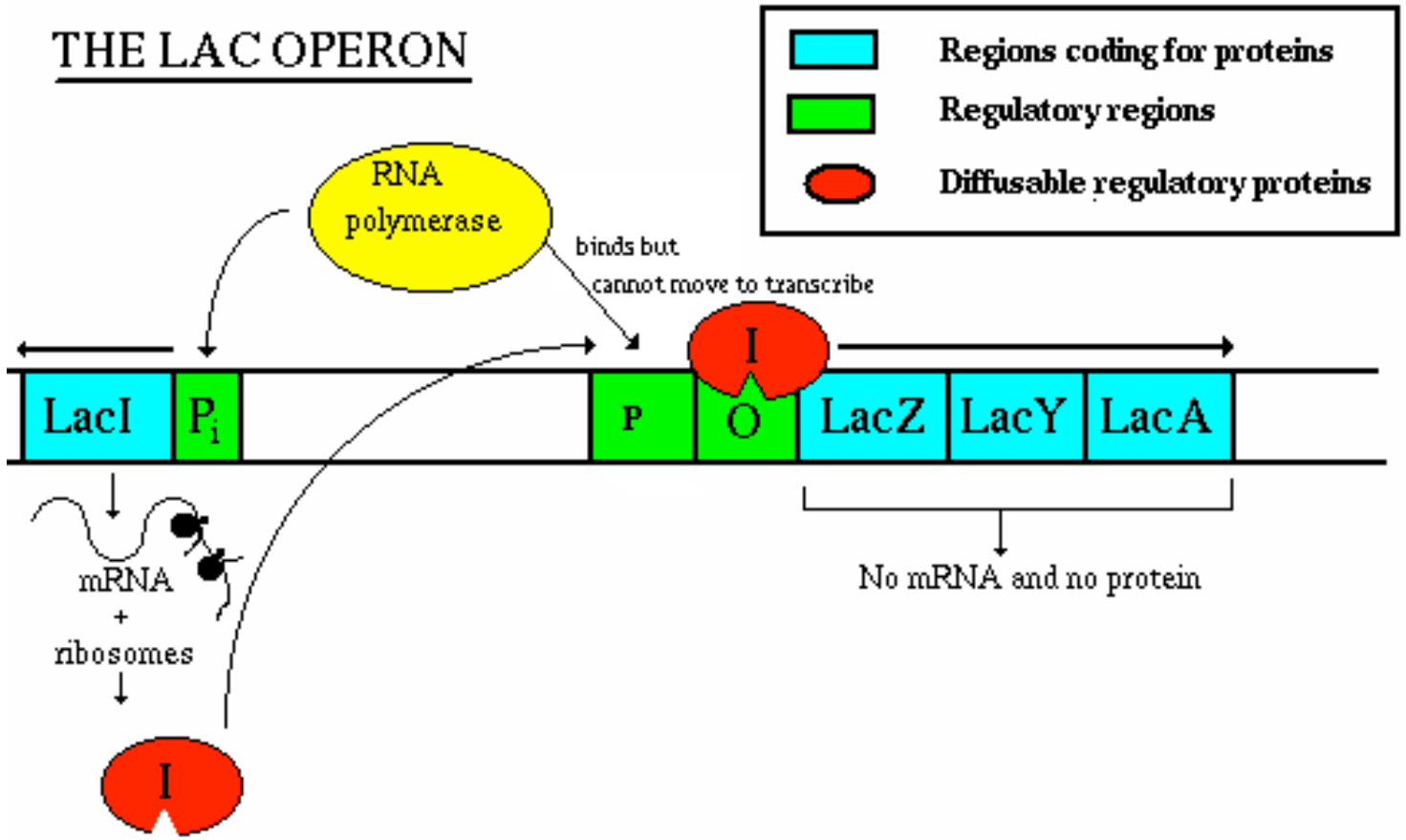
Gene Regulation Example: the lac Operon



this protein regulates the transcription of LacZ, LacY, LacA

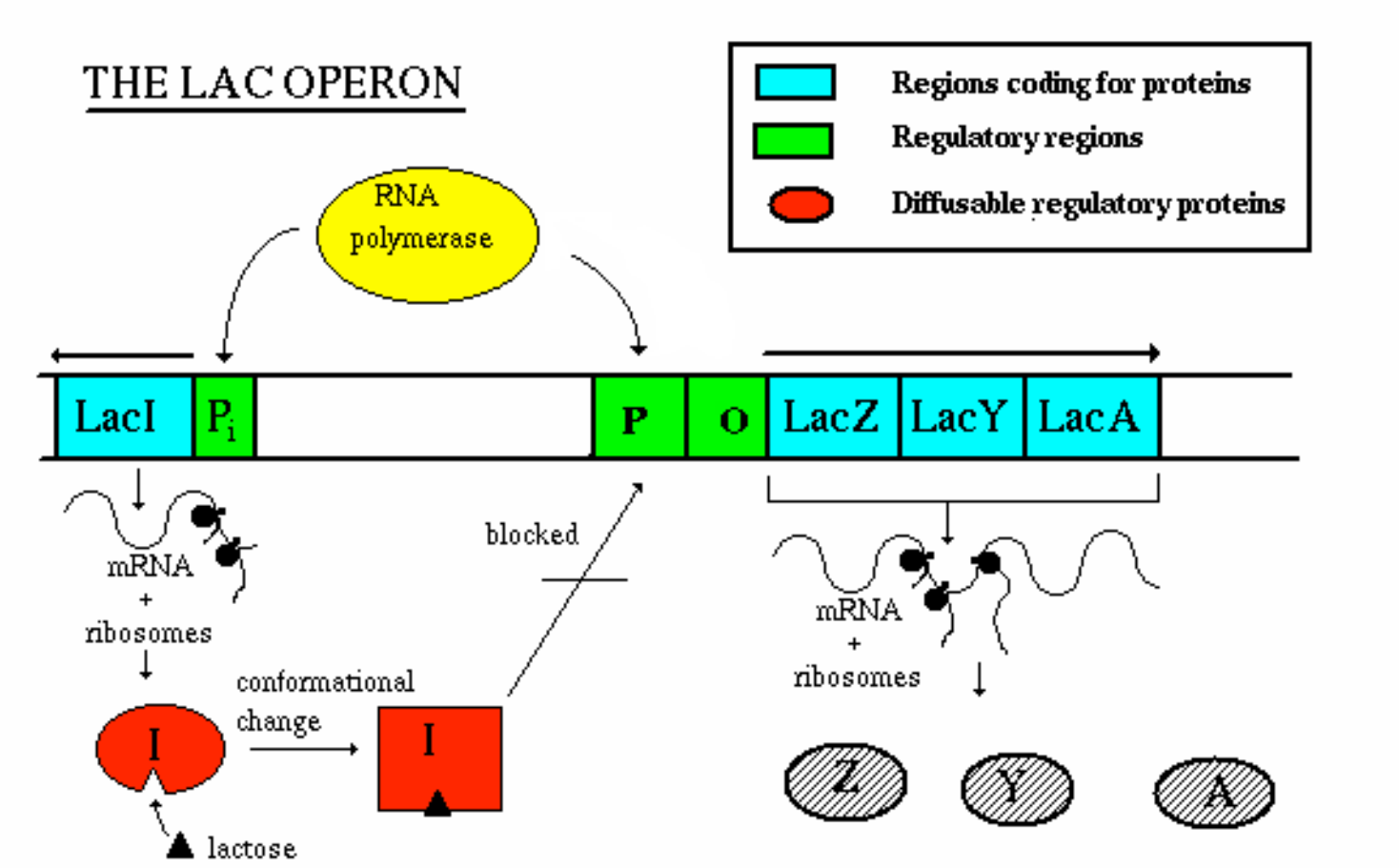
these proteins metabolize lactose

Gene Regulation Example: the lac Operon



lactose is absent ⇒ the protein encoded by lacI represses transcription of the lac operon

Gene Regulation Example: the lac Operon



lactose is present ⇒ it binds to the protein encoded by lacI changing its shape; in this state, the protein doesn't bind upstream from the lac operon; therefore the lac operon can be transcribed

Gene Regulation Example: the lac Operon

- this example provides a simple illustration of how a cell can regulate (turn on/off) certain genes in response to the state of its environment
  - an *operon* is a sequence of genes transcribed as a unit
  - the lac operon is involved in metabolizing lactose
    - it is "turned on" when lactose is present in the cell
    - the lac operon is regulated at the transcription level
- the depiction here is incomplete; for example, the level of glucose in the cell also influences transcription of the lac operon

Completed Genomes

Type	Approximate # Completed
Archaea	53
Bacteria	700
Eukaryota	94
metagenomes	130
Organelles, Phages, Plasmids, Viroids, Viruses	too many to keep track!

* Genomes OnLine Database (9/08)

Selected milestones in genome sequencing

	Year	Common Name	Species	# of Chromosomes	Size (base pairs)
	1995	Bacterium	Haemophilus influenzae	1	1.8×10^6
	1996	Yeast	Saccharomyces cerevisiae	16	1.2×10^7
	1998	Worm	Caenorhabditis elegans	6	1.0×10^8
	1999	Fruit Fly	Drosophila melanogaster	4	1.3×10^8
	2000	Human	Homo sapiens	23	3.4×10^9
	2002	Mouse	Mus musculus	20	3.5×10^9
	2004	Rat	Rattus norvegicus	21	2.9×10^9
	2005	Chimpanzee	Pan troglodytes	24	3.6×10^9

Sequence is freely available

NCBI - http://www.ncbi.nlm.nih.gov UCSC - http://genome.ucsc.edu

But Wait, There's More...

- > 1000 other publicly available databases pertaining to molecular biology (see pointer to *Nucleic Acids Research* directory on course home page)
- GenBank

> 82 million sequence entries

> 85 billion bases

- UniProtKB / Swis-Prot

> 6 million protein sequence entries

> 2 billion amino acids

- Protein Data Bank

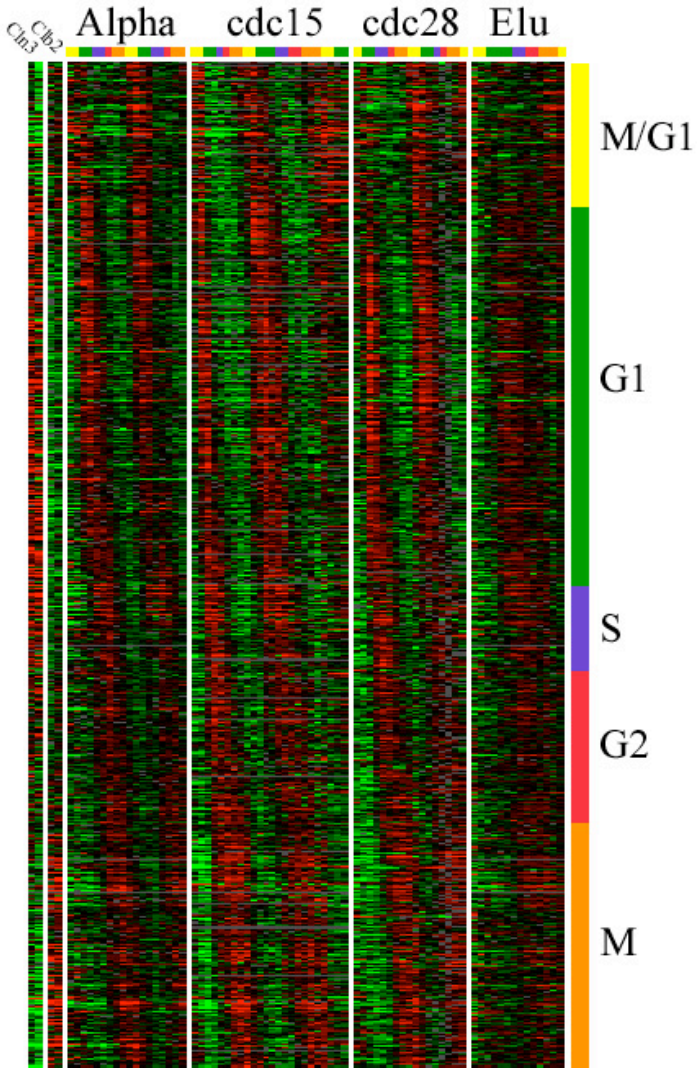
52,821 protein (and related) structures

* all numbers current about 9/08

More Data: High-Throughput Experiments

- RNA abundances
- protein abundances
- small molecule abundances
- protein-protein interactions
- protein-DNA interactions
- protein-small molecule interactions
- genetic variants of an individual (e.g. which DNA base does the individual have at a few thousand selected positions)
- something (e.g. viral replication) measured across thousands of genetic variants
- etc.

Example HT Experiment



- this figure depicts one yeast gene-expression data set
  - each row represents a gene
  - each column represents a measurement of gene expression (mRNA abundance) at some time point
- red indicates that a gene is being expressed more than some baseline; green means less

Figure from Spellman et al., *Molecular Biology of the Cell,* 9:3273-3297, 1998

More Data: Interactions

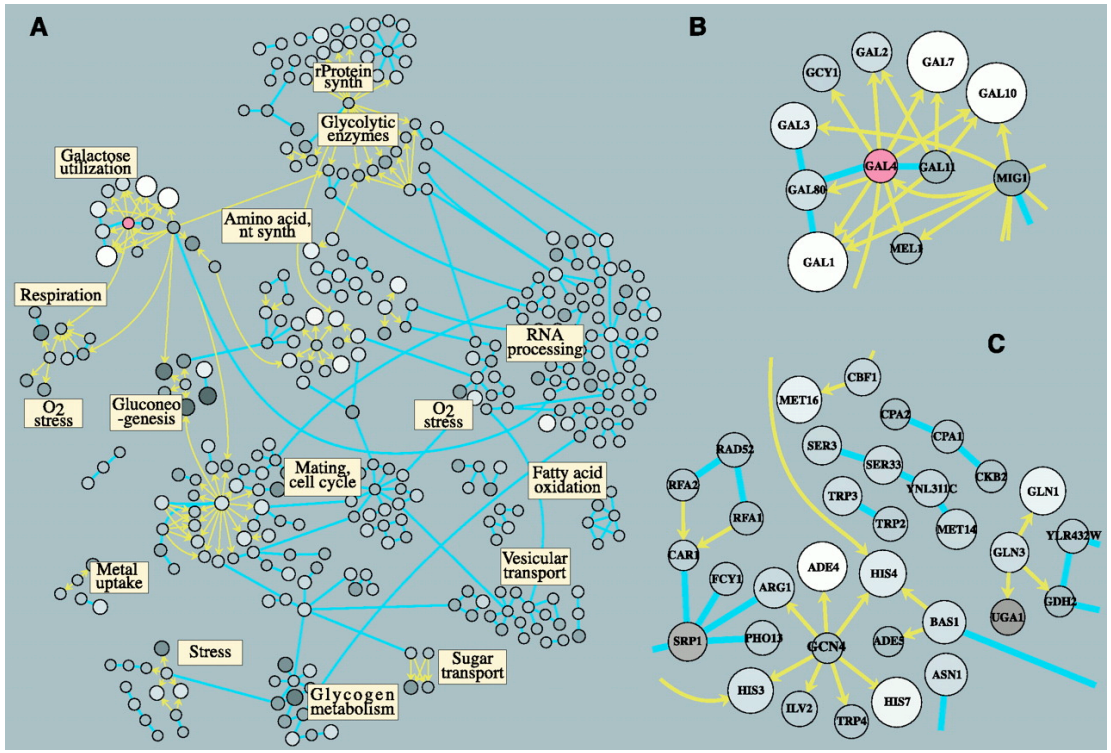


Figure from Ideker et al., *Science* 292(5518):929-934, 2001

- each node represents a gene product (protein)
- blue edges show direct protein-protein interactions
- yellow edges show interactions in which one protein binds to DNA and affects the expression of another

Significance of the Genomics Revolution

- data driven biology
  - functional genomics
  - comparative genomics
  - systems biology
- molecular medicine
  - identification of genetic components of various maladies
  - diagnosis/prognosis from sequence/expression
  - gene therapy
- pharmacogenomics
  - developing highly targeted drugs
- toxicogenomics
  - elucidating which genes are affected by various chemicals

Bioinformatics Revisited

Representation/storage/retrieval/ analysis of biological data concerning

- sequences (DNA, protein, RNA)
- structures (protein, RNA)
- functions (protein, sequence *signals*)
- activity levels (mRNA, protein, metabolites)
- networks of interactions (metabolic pathways, regulatory pathways, signaling pathways)

of/among biomolecules