CH 17
Transcription & Translation
Basic Principles of Transcription & Translation
RNA is the bridge between genes and the proteins for which they code.
Transcription is the synthesis of RNA under the direction of DNA.
Transcription produces messenger RNA (mRNA).
Translation is the synthesis of a polypeptide, using information in the mRNA.
Ribosomes are the sites of translation.

In prokaryotes, translation of mRNA can begin before transcription has finished.
In a eukaryotic cell, the nuclear envelope separates transcription from translation.
Eukaryotic RNA transcripts are modified through RNA processing to yield finished mRNA.
A primary transcript is the initial RNA transcript from any gene prior to processing.
The central dogma is the concept that cells are governed by a cellular chain of command:

The Genetic Code
There are 20 amino acids, but there are only four nucleotide bases in DNA.
How many nucleotides correspond to an amino acid?

The flow of information from gene to protein is based on a triplet code: a series of
nonoverlapping, three-nucleotide words.
The words of a gene are transcribed into complementary nonoverlapping three-nucleotide words
of mRNA.
These words are then translated into a chain of amino acids, forming a polypeptide.

During transcription, one of the two DNA strands, called the template strand, provides a template
for ordering the sequence of complementary nucleotides in an RNA transcript.
The template strand is always the same strand for a given gene.
During translation, the mRNA base triplets, called codons, are read in the 5' to 3' direction.
Codons along an mRNA molecule are read by translation machinery in the 5' to 3' direction.
Each codon specifies the amino acid (one of 20) to be placed at the corresponding position along
a polypeptide.

Genetic Code is Universal. Genes can be transcribed and translated after being transplanted from
one species to another.
Transcription is the DNA directed synthesis of RNA
Transcription is the first stage of gene expression.
RNA synthesis is catalyzed by RNA polymerase, which pries the DNA strands apart and hooks
together the RNA nucleotides.
The RNA is complementary to the DNA template strand. RNA synthesis follows the same base-pairing rules as DNA, except that uracil substitutes for thymine.

The DNA sequence where RNA polymerase attaches is called the promoter; in bacteria, the sequence signaling the end of transcription is called the terminator. The stretch of DNA that is transcribed is called a transcription unit.

Synthesis of RNA transcripts
3 steps:
Initiation
Elongation
Termination
RNA polymerase binding
Promoters signal the transcriptional start point and usually extend several dozen nucleotide pairs upstream of the start point.
Transcription factors mediate the binding of RNA polymerase and the initiation of transcription. The completed assembly of transcription factors and RNA polymerase II bound to a promoter is called a transcription initiation complex.
A promoter called a TATA box is crucial in forming the initiation complex in eukaryotes.

Elongation of the RNA strand
As RNA polymerase moves along the DNA, it untwists the double helix, 10 to 20 bases at a time.
Transcription progresses at a rate of 40 nucleotides per second in eukaryotes.
A gene can be transcribed simultaneously by several RNA polymerases.
Nucleotides are added to the 3' end of the growing RNA molecule.

Termination of Transcription
The mechanisms of termination are different in bacteria and eukaryotes.
In bacteria, the polymerase stops transcription at the end of the terminator and the mRNA can be translated without further modification.
In eukaryotes, RNA polymerase II transcribes the polyadenylation signal sequence; the RNA transcript is released 10–35 nucleotides past this polyadenylation sequence.

Modification of RNA after transcription by Eukaryotic cells
Enzymes in the eukaryotic nucleus modify pre-mRNA (RNA processing) before the genetic messages are dispatched to the cytoplasm.
During RNA processing, both ends of the primary transcript are usually altered.
Also, usually some interior parts of the molecule are cut out, and the other parts spliced together.

Alteration of mRNA ends
Each end of a pre-mRNA molecule is modified in a particular way;
The 5' end receives a modified nucleotide 5' cap
The 3' end gets a poly-A tail
These modifications share several functions
They seem to facilitate the export of mRNA
They protect mRNA from hydrolytic enzymes
They help ribosomes attach to the 5' end

Split genes & RNA splicing

Noncoding regions are called intervening sequences, or introns.
The other regions are called exons because they are eventually expressed, usually translated into amino acid sequences.
RNA splicing removes introns and joins exons, creating an mRNA molecule with a continuous coding sequence.

In some cases, RNA splicing is carried out by spliceosomes.
Spliceosomes consist of a variety of proteins and several small nuclear ribonucleoproteins (snRNPs) that recognize the splice sites.

Ribozymes
Ribozymes are catalytic RNA molecules that function as enzymes and can splice RNA.
The discovery of ribozymes rendered obsolete the belief that all biological catalysts were proteins.

What properties allow RNA to behave like an enzyme?
It can form a three-dimensional structure because of its ability to base-pair with itself.
Some bases in RNA contain functional groups that may participate in catalysis.
RNA may hydrogen-bond with other nucleic acid molecules.

Translation = mRNA       Protein
A cell translates an mRNA message into protein with the help of transfer RNA (tRNA).
tRNA transfer amino acids to the growing polypeptide in a ribosome.
Translation is a complex process in terms of its biochemistry and mechanics.

Structure & Fx of tRNA
Each carries a specific amino acid on one end.
Each has an anticodon on the other end; the anticodon base-pairs with a complementary codon on mRNA.
A tRNA molecule consists of a single RNA strand that is only about 80 nucleotides long.
Flattened into one plane to reveal its base pairing, a tRNA molecule looks like a cloverleaf.
Accurate translation requires two steps
First: a correct match between a tRNA and an amino acid, done by the enzyme aminoacyl-tRNA synthetase
Second: a correct match between the tRNA anticodon and an mRNA codon
Flexible pairing at the third base of a codon is called wobble and allows some tRNAs to bind to more than one codon

Ribosomes
Ribosomes facilitate specific coupling of tRNA anticodons with mRNA codons in protein synthesis.
The two ribosomal subunits (large and small) are made of proteins and ribosomal RNA (rRNA). Bacterial and eukaryotic ribosomes are somewhat similar but have significant differences: some antibiotic drugs specifically target bacterial ribosomes without harming eukaryotic ribosomes.

A ribosome has three binding sites for tRNA
The P site holds the tRNA that carries the growing polypeptide chain
The A site holds the tRNA that carries the next amino acid to be added to the chain
The E site is the exit site, where discharged tRNAs leave the ribosome

Building a Polypeptide
The three stages of translation
Initiation
Elongation
Termination
All three require protein “factors” that aid in the translation process

The initiation stage of translation brings together mRNA, a tRNA with the first amino acid, and the two ribosomal subunits.
First, a small ribosomal subunit binds with mRNA and a special initiator tRNA.
Then the small subunit moves along the mRNA until it reaches the start codon (AUG). Proteins called initiation factors bring in the large subunit that completes the translation initiation complex.

Elongation of the Polypeptide chain
During the elongation stage, amino acids are added one by one to the preceding amino acid at the C-terminus of the growing chain.
Each addition involves proteins called elongation factors and occurs in three steps: codon recognition, peptide bond formation, and translocation.
Translation proceeds along the mRNA in a 5' to 3' direction.
Termination of Translation
Termination occurs when a stop codon in the mRNA reaches the A site of the ribosome. The A site accepts a protein called a release factor. The release factor causes the addition of a water molecule instead of an amino acid. This reaction releases the polypeptide, and the translation assembly then comes apart.

Polysomes
A number of ribosomes can translate a single mRNA simultaneously, forming a polyribosome (or polysome). Polyribosomes enable a cell to make many copies of a polypeptide very quickly.

Mutations
Mutations are changes in the genetic material of a cell or virus. Point mutations are chemical changes in just one base pair of a gene.
Nucleotide-pair substitutions
One or more nucleotide-pair insertions or deletions
The change of a single nucleotide in a DNA template strand can lead to the production of an abnormal protein.

Substitutions
A nucleotide-pair substitution replaces one nucleotide and its partner with another pair of nucleotides.
Silent mutations have no effect on the amino acid produced by a codon because of redundancy in the genetic code.
Missense mutations still code for an amino acid, but not the correct amino acid.
Nonsense mutations change an amino acid codon into a stop codon, nearly always leading to a nonfunctional protein.

Insertions & Deletions
Insertions and deletions are additions or losses of nucleotide pairs in a gene. These mutations have a disastrous effect on the resulting protein more often than substitutions do. Insertion or deletion of nucleotides may alter the reading frame, producing a frameshift mutation.

Mutagens
Spontaneous mutations can occur during DNA replication, recombination, or repair. Mutagens are physical or chemical agents that can cause mutations.