

- **MOLECULAR BIOLOGY OF THE GENE**
DNA, RNA, AND PROTEIN SYNTHESIS

In virtually all organisms, **DNA (deoxyribonucleic acid)** is the genetic material that is inherited from one generation to the next.

In a few viruses, **RNA (ribonucleic acid)** is the genetic material that is inherited from one generation of viruses to the next generation of viruses.

nucleotide = a monomer of a nucleic acid (either DNA or RNA). Each nucleotide consists of a five carbon sugar (ribose or deoxyribose) covalently bonded to one of four different nitrogenous bases (a compound that contains nitrogen atoms) and a phosphate group. Nucleotides are the building blocks of DNA and RNA.

DNA = the genetic material that organisms inherit from their parents; a double stranded helical molecule that consists of nucleotides linked together. The nucleotides that make up DNA are made of a deoxyribose sugar molecule attached to a phosphate group and a nitrogenous base. The four nitrogenous bases in DNA are adenine (A), cytosine (C), guanine (G), and thymine (T). DNA is most often double stranded.

RNA = a type of nucleic acid consisting of nucleotides bonded together. The nucleotides that make up RNA are made of a ribose sugar molecule attached to a phosphate group and a nitrogenous base. The four nitrogenous bases in RNA are adenine (A), cytosine (C), guanine (G), and uracil (U). RNA is usually a single stranded molecule and plays important roles in protein synthesis. RNA is most often single stranded.

sugar-phosphate backbone = refers to the "sides of the ladder" in a molecule of DNA. The sugar (deoxyribose) and phosphate groups of each nucleotide in a strand of DNA are linked together with their nitrogenous bases (A, C, G, T) sticking out as the "rungs of the ladder".

complementary bases = nitrogenous bases in nucleic acids that will form hydrogen bonds with each other. To form a double stranded DNA molecule, certain bases will only form hydrogen bonds with certain other bases. Adenine will only bond to thymine (A to T) and guanine will only bond to cytosine (G to C). Thus, one strand of DNA that had the sequence ATGCTCGTA would only bind to a complementary strand that had the sequence TACGAGCAT to form a double stranded DNA molecule.

DNA double helix = refers to the three dimensional structure of DNA. The two strands of DNA are held together by hydrogen bonds and the two strands are wound around each other into a helical formation.

gene = a discrete unit of hereditary information consisting of a specific nucleotide sequence in DNA. Genes are made of DNA so they are found on the chromosomes in the nucleus of all cells (or the nucleoid region of bacteria).

transcription = the synthesis of mRNA from a DNA template. mRNA is formed from a DNA template due to the complementary base pairing of RNA nucleotides to a specific sequence of one strand of DNA (a gene). During mRNA synthesis by the enzyme **RNA polymerase**, a sequence of DNA such as ATGCTCGTA is used to form a complementary mRNA molecule (UACGAGCAU). RNA uses the base uracil instead of thymine. **Uracil is complementary to (binds to) adenine on a strand of DNA. RNA molecules do not contain the thymine.**

translation = the synthesis of a protein using the genetic information encoded in an mRNA molecule. The nucleic acid sequenced is "translated" into an amino acid sequence. Ribosomes (on the rough endoplasmic reticulum and in the free floating in the cytoplasm) are the site of translation in a cell.

RNA polymerase = the enzyme that links together the growing chain of RNA nucleotides to form a molecule of mRNA during transcription.

mRNA (messenger RNA) = the type of RNA that encodes genetic information from DNA and conveys it to ribosomes, where the information is translated into an amino acid sequence and a protein is made.

codon = a three nucleotide sequence of mRNA that specifies a particular amino acid and functions as the basic unit of the genetic code. Codons on mRNA bind to anticodons on tRNA during protein synthesis.

tRNA (transfer RNA) = the type of RNA that functions as an interpreter in translation. Each tRNA molecule carries a specific anticodon, picks up a specific amino acid, and conveys the amino acid to the appropriate codon on mRNA. tRNA's work together with ribosomes to make proteins.

anticodon = on a tRNA molecule, a specific sequence of three nucleotides that is complementary to a codon triplet on an mRNA molecule.

rRNA (ribosomal RNA) = the type of RNA that, together with proteins, makes up ribosomes.

genetic (or triplet) code = the set of rules that causes a specific codon (three nucleotides) of mRNA to cause a specific amino acid to become part of a protein which is synthesized from the mRNA molecule. Each codon codes for one of twenty different amino acids that make up proteins. The genetic code is redundant meaning that sometime more than one codon will code for the same amino acid.

start codon = on an mRNA molecule, the codon (AUG) that signals where the first tRNA should bind to start the translation of a protein.

stop codon = on an mRNA molecule, one of three codons (UAG, UAA, or UGA) that signals translation to stop.

ribosome = a cell organelle consisting of tRNA and protein organized into two subunits. Ribosomes are where translation (protein synthesis) occurs either in the cytoplasm or in association with the rough endoplasmic reticulum.

mutation = a change in the nucleotide sequence of DNA. If a mutation changes the sequence of DNA in a gene, then the mRNA and protein made from that gene will be different than it was before the mutation. Many mutations are harmful and lead to the synthesis of abnormal or non-functional proteins which could cause cell death or cancer. Rarely, a mutation occurs that is beneficial to an organism that allows it to make new proteins capable of new functions. These new "mutated" proteins may cause the organisms to show different traits and possibly have different abilities. Mutations are caused by mutagens.

mutagen = a chemical or physical agent that interacts with DNA to cause a mutation. The most common physical mutagen is high-energy radiation such as X-rays or UV (ultraviolet) light. There are thousands of chemical mutagens such as benzene, cigarette smoke, some steroids, and alcohol. Some mutagens are also carcinogens.

carcinogen = a chemical or physical agent that causes cells to become cancerous. Many carcinogens interact with DNA to cause mutations that lead to cancer. UV light from the sun and cigarette smoke are leading causes of skin and lung cancer worldwide.

• BIOTECHNOLOGY, GENETIC ENGINEERING AND CLONING

genetic expression = the overall production of the various proteins of an organism from its genes.

cellular differentiation = specialization in the structure and function of cells. Different cell types in a multicellular organism express different genes and thus different cells have different structures and functions. All cells in humans (animals) contain the same DNA/chromosomes, but different cells “turn-on” and “turn off” various genes to become different cell types.

stem cell = cells found in most, if not all, multi-cellular organisms. They are characterized by the ability to renew themselves through mitotic cell division and differentiating into a diverse range of specialized cell types. Stem cells are pluripotent cells that can differentiate and become many different cell types.

regulatory gene = a gene that codes for a protein, such as a repressor, that controls the transcription of another gene or group of genes.

transcription factor = in a eukaryotic cell, a protein that functions in initiating or regulating transcription. Transcription factors bind to DNA or to other proteins that bind DNA.

Steroids such as testosterone and estradiol (estrogen) bind to protein receptors that are transcription factors.

plasmid = in a bacterial cell, a small ring of DNA that is separate from the bacterial chromosome. **Using restriction enzymes, plasmids can be modified to contain genes from other organisms. When these genetically modified plasmids are put back into bacteria (transformation) the bacteria will produce proteins from the newly introduced genes. These proteins can be isolated and used by humans for a variety of purposes from agriculture to medicine and beyond.**

biotechnology = the use of living organisms (often microbes) to perform useful tasks important for human technology, food production or industry.

restriction enzyme = a bacterial enzyme that cuts up DNA at a specific nucleotide sequence in the DNA.

recombinant DNA = a DNA molecule carrying genes derived from two or more sources.

gene cloning = the production of multiple copies of a gene.

restriction fragments = molecules of DNA produced from a longer DNA molecule cut up by a restriction enzyme; used in RFLP analysis.

RFLP's = Restriction Fragment Length Polymorphisms ("rif-lips"). The differences in homologous DNA sequences are reflected in different lengths of restriction fragments produced when DNA is cut apart with restriction enzymes.

electrophoresis = a technique for separating and purifying macromolecules (such as DNA or proteins). A mixture of molecules is placed on a gel between a positively charged electrode and a negatively charged electrode. Negative charges on the molecules are attracted to the positive electrode and the molecules migrate toward the negative electrode. The molecules separate in the gel according to their rate of migration (which is largely determined by their size). Larger molecules move slower through the gel than smaller molecules.

DNA fingerprint = an individual's unique collection of DNA restriction fragments that can be detected by electrophoresis.

transgenic organism = an organism that contains genes from another species.

- **CLONING**

gene cloning = the production of multiple copies of a gene.

clone = a group of genetically identical cells or organisms derived from a single common cell or single organism; a copy of a specific gene.

Cloning in biology is the process of producing populations of genetically-identical individuals that occurs in nature when organisms such as bacteria, insects or plants reproduce asexually. Cloning in biotechnology refers to processes used to create copies of DNA fragments (molecular cloning), cells (cell cloning), or organisms

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Nuclear Transfer/Transplantation = a form of cloning. The steps involve removing the DNA from an oocyte (unfertilized egg), and injecting the nucleus which contains the DNA to be cloned. In rare instances, the newly constructed cell will divide normally, replicating the new DNA while remaining in a pluripotent state. If the cloned cells are placed in the uterus of a female mammal, a cloned organism develops to term in rare instances. This is how Dolly the Sheep and many other species were cloned.

The nucleus, which contains the organism's DNA, of a somatic cell (a body cell other than a sperm or egg cell) is removed and the rest of the cell discarded. At the same time, the nucleus of an egg cell is removed. The nucleus of the somatic cell is then inserted into the enucleated egg cell. After being inserted into the egg, the somatic cell nucleus is reprogrammed by the host cell. The egg, now containing the nucleus of a somatic cell, is stimulated with a shock and will begin to divide. After many mitotic divisions in culture, this single cell forms a blastocyst (an early stage embryo with about 100 cells) with almost identical DNA to the original organism.

Refer to your lecture notes and the websites below for a description of how Dolly the sheep was cloned. Importantly, Dolly was cloned from a **differentiated mammalian cell**. Previous cloning work involved the use of amphibian (frog) eggs that were not yet differentiated.

Cloning Dolly at:

<http://www.luc.edu/faculty/wwasser/dev/shclone.htm>

<http://www.animalresearch.info/en/medical/timeline/Dolly>

- **CHROMOSOMAL ABNORMALITIES AND SOME GENETIC DISORDERS**

Hemophilia = a sex linked recessive trait. Hemophiliacs bleed excessively when injured because they have inherited an abnormal allele for a protein factor that is required for normal blood clotting.

gene therapy = the replacement or supplementation of a defective gene with a normal gene in an individual who is suffering from a disorder that is traceable to a single defective gene. Also, the use of manipulated genes in the treatment of genetically based diseases such as cancer or AIDS.

eugenics = a socially rejected practice, among humans, of attempting to eliminate genetic disorders (or races) and "undesirable" inherited traits by selective breeding.

karyotype = a display of micrographs of the metaphase chromosomes of a cell arranged by size and shape. The 23 different chromosomes in human cells can be identified by their appearance using this technique. Karyotypes can be used to detect chromosomal abnormalities such as Down's syndrome.

nondisjunction = an accident of meiosis or mitosis in which both members of a pair of homologous chromosomes or both sister chromatids fail to separate. Nondisjunction during meiosis ends with one gamete that contains zero copies of a particular chromosome and another gamete that contains two copies of the same chromosome.

Down syndrome = a human genetic disease resulting from having an extra chromosome 21 (trisomy of chromosome 21). Down's is characterized by heart and respiratory defects, facial malformations and varying degrees of mental retardation. Incidence of Down's syndrome increases with the age of the mother (about 1% at the age of 40 and greater in older women).

Some Recessive Genetic Disorders = albinism, cystic fibrosis, phenylketonuria, sickle cell anemia (homozygous form), Tay-Sachs disease

Some Dominant Genetic Disorders = achondroplasia (a type of dwarfism), Alzheimer's disease (one type, not all types), Huntington's disease, Hypercholesterolemia

Huntington's disease = a human genetic disease caused by a dominant allele. If you have one copy of the Huntington's gene you will get Huntington's disease. Symptoms of the disease do not usually appear until after the age of 40. Huntington's is characterized by uncontrollable body movements, degeneration of the nervous system and individuals usually die within 10-20 years after the onset of symptoms.

Sex-Linked Traits/Disorders = a disorder resulting from alleles of genes located on sex chromosomes. Most of the known sex-linked disorders involve genes on the X chromosome (the Y chromosome is small and has relatively few genes on it). More males than females display sex-linked disorders because males only have one copy of the X chromosome. If a male receives the allele for a sex-linked disorder, he will get it regardless of whether the allele is dominant or recessive. Females will only get recessive sex-linked disorders if they are homozygous for the allele (if the female has two copies of the allele, one on each X chromosome).

Sex-Linked Disorders = red-green color blindness and hemophilia.