Study Materials for II B. Sc Zoology – III Semester

Subject – CELL AND MOLEULAR BIOLOGY

Subject Code: 18K3Z04

(Unit III to V)

CHROMOSOMES

Structure and Organization in Prokaryotic and Eukaryotic

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Prokaryotic and Eukaryotic Chromosomes

- The genome of an organism encompasses all of the genes of that organism.
- **Gene** is a distinct sequence of nucleotides forming part of a chromosome, the order of which determines the order of monomers in a polypeptide or nucleic acid molecule.
- Thus a protein-coding gene is defined as a region of DNA that encodes a single polypeptide or a set of closely related polypeptides.
- Genes are contained in chromosomes.
- Chromosomes are thus structures within cells that contain hundreds to thousands of genes.

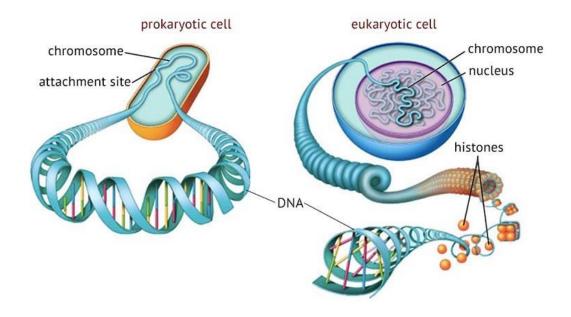


Image Source: https://image.slidesharecdn.com/3-150517031851-lva1-app6892/95/ib-biology-32-slides-chromosomes-3-638.jpg **and** http://bio1100.nicerweb.com/Locked/media/SAVE/ch06/chromosome.html

Prokaryotic Chromosomes

- The DNA of a bacterial cell, such as *Escherichia coli*, is a circular double-stranded molecule often referred to as the bacterial chromosome.
- The circular DNA is packaged into a region of the cell called the nucleoid where it is
 organized into 50 or so loops or domains that are bound to a central protein scaffold,
 attached to the cell membrane.
- The DNA is negatively supercoiled, that is, it is twisted upon itself.
- It is complexed with several DNA-binding proteins, the most common of which are proteins HU, HLP-1 and H-NS. These are histone-like proteins.

Eukaryotic Chromosomes

- The large amount of genomic DNA in a eukaryotic cell is tightly packaged in chromosomes contained within a specialized organelle, the nucleus.
- With the exception of the sex chromosomes, diploid eukaryotic organisms such as humans have two copies of each chromosome, one inherited from the father and one from the mother.

- Chromosomes contain both DNA and protein.
- Most of the protein on a weight basis is histones, but there are also many thousands
 of other proteins found in far less abundance and these are collectively called nonhistone proteins (NHP).
- This nuclear DNA–protein complex is called chromatin.
- In the nucleus, each chromosome contains a single linear double-stranded DNA molecule.
- The length of the packaged DNA molecule varies. In humans, the shortest DNA molecule in a chromosome is about 1.6 cm and the longest is about 8.4 cm.
- The extensive packaging of DNA in chromosomes results from three levels of folding involving nucleosomes, 30 nm filaments and radial loops.

1. Nucleosomes

- The first level of packaging involves the binding of the chromosomal DNA to histones.
- Overall, in chromosomes, the ratio of DNA to histones on a weight basis is approximately 1:1.
- There are five main types of histones called H1, H2A, H2B, H3 and H4.
- Histones are very basic proteins; about 25% of their amino acids are lysine or arginine so histones have a large number of positively charged amino acid sidechains.
- These positively charged groups therefore bind to the negatively charged phosphate groups of DNA

2. **30** nm fiber

- If nuclei are lysed very gently, the chromatin is seen to exist as a 30 nm diameter fiber.
- The fiber is formed by a histone H1 molecule binding to the linker DNA of each nucleosome at the point where it enters and leaves the nucleosome.
- The histone H1 molecules interact with each other, pulling the nucleosomes together.

3. Radial loops

- When chromosomes are depleted of histones, they are seen to have a central fibrous 'protein scaffold' (or nuclear matrix) to which the DNA is attached in loops.
- Therefore, in vivo it seems likely that the next order of packaging involves the attachment of the 30 nm fiber to multiple locations on this central protein scaffold in a series of radial loops.
- The mitochondria and chloroplasts of eukaryotic cells also contain DNA but, unlike the nuclear DNA, this consists of double-stranded circular molecules resembling bacterial chromosomes.

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GAINT CHROMOSOMES

Polytene chromosomes are large chromosomes which have thousands of DNA strands. They provide a high level of function in certain tissues such as salivary glands of insects.

Polytene chromosomes were first reported by E.G.Balbiani in 1881. Polytene the best found in dipteran flies: understood are of Drosophila, Chironomus and Rhynchosciara. They are present in another group of arthropods of the class Collembola, protozoan group Ciliophora, a mammalian trophoblasts and antipodal, and suspensor cells in plants. In insects, they are commonly found in the salivary glands when the cells are not dividing.

They are produced when repeated rounds of DNA replication without cell division forms a giant chromosome. Thus polytene chromosomes form when multiple rounds of replication produce many sister chromatids *which stay fused together*.

Polytene chromosomes, at interphase, are seen to have distinct thick and thin banding patterns. These patterns were originally used to help map chromosomes, identify small chromosome mutations, and in taxonomic identification. They are now used to study the function of genes in transcription.

Structure

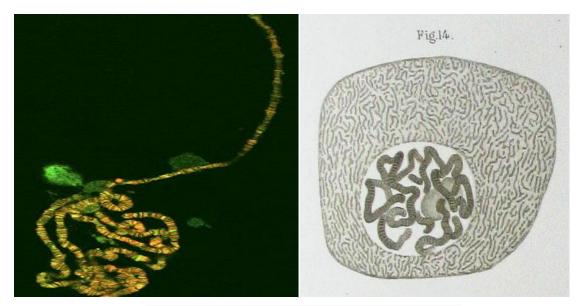
In insects, polytene chromosomes are commonly found in the salivary glands; they are also referred to as "salivary gland chromosomes". The large size of the chromosome is due to the presence of many longitudinal strands called chromonemata; hence the name polytene (many stranded). They are about 0.5 mm in length and 20 μ m in diameter. The chromosomal strands are formed after repeated division of the chromosome in the absence of cytoplasmic division.

This type of division is called endomitosis. The polytene chromosome contains two types of bands, dark bands and interbands. The dark bands are darkly stained and the inter bands are lightly stained with nuclear stains. The dark bands contain more DNA and less RNA. The interbands contain more RNA and less DNA. The amount of DNA in interbands ranges from 0.8 - 25%.

The bands of polytene chromosomes become enlarged at certain times to form swellings called puffs. The formation of puffs is called puffing. In the regions of puffs, the chromonemata uncoil and open out to form many loops. The puffing is caused by the uncoiling of individual chromomeres in a band. The puffs indicate the site of active genes where mRNA synthesis takes place.

The chromonemata of puffs give out a series of many loops laterally. As these loops appear as rings, they are called Balbiani rings after the name of the researcher who discovered them. They are formed of DNA, RNA and a few proteins. As they are the site of transcription, transcription mechanisms such as RNA polymerase and ribonucleoproteins are present.

In protozoans, there is no transcription, since the puff consists only of DNA.



Polytene chromosomes in a Chironomus salivary gland cell

Polytene chromosomes were originally observed in the larval salivary glands of *Chironomus* midges by Édouard-Gérard Balbiani in 1881. Balbiani described the chromosomal puffs among the tangled thread inside the nucleus, and named it "permanent spireme". In 1890, he observed similar spireme in a ciliated protozoan *Loxophyllum meleagris*.

The existence of such spireme in *Drosophila melanogaster* was reported by Bulgarian geneticist Dontcho Kostoff in 1930. Kostoff predicted that the discs (bands) which he observed were "the actual packets in which inherited characters are passed from generation to generation."

The hereditary nature of these structures was not confirmed until they were studied in *Drosophila melanogaster* in the early 1930s by German biologists Emil Heitz and Hans Bauer. In 1930, Heitz studied different species of *Drosophila* (*D. melanogaster*, *D. simulans*, *D. hydei*, and *D. virilis*) and found that all their interphase chromatins in certain cells were swollen and messy.

In 1932, he collaborated with Karl Heinrich Bauer with whom he discovered that the tangled chromosomes having distinct bands are unique to the cells of the salivary glands, midgut, Malphigian tubules, and brain of the flies *Bibio hurtulunus* and *Drosophila funebris*. The two papers were published in the early 1933. Unaware of these papers, an American geneticist Theophilus Shickel Painter reported in

December 1933 the existence of giant chromosome in *D. melanogaster* (followed by a series of papers the following year).

Learning of this, Heitz accused Painter of deliberately ignoring their original publication to claim priority of discovery.^[13] In 1935, Hermann J. Muller and A.A. Prokofyeva established that the individual band or part of a band corresponds with a gene in *Drosophila*. The same year, P.C. Koller hesitantly introduced the name "polytene" to describe the giant chromosome, writing:

It seems that we can regard these chromosomes as corresponding with paired pachytene chromosomes at meiosis in which the intercalary parts between chromomeres have been stretched and separated into smaller units, and in which, instead of two threads lying side by side, we have 16 or even more. Hence they are "polytene" rather than pachytene; I do not, however, propose to use this term; I shall refer to them as "multiple threads."

CELL DIVISION

Cell division is the process cells go through to divide. There are several types of cell division, depending upon what type of organism is dividing. Organisms have evolved over time to have different and more complex forms of cell division. Most prokaryotes, or bacteria, use binary fission to divide the cell. Eukaryotes of all sizes use *mitosis* to divide.

Sexually-reproducing eukaryotes use a special form of cell division called *meiosis* to reduce the genetic content in the cell. This is necessary in sexual reproduction because each parent must give only half of the required genetic material, otherwise the offspring would have too much DNA, which can be a problem. These different types of cell division are discussed below.

Types of Cell Division

Prokaryotic Cell Division

Prokaryotes replicate through a type of cell division known as *binary fission*. Prokaryotes are simple organism, with only one membrane and no division internally. Thus, when a prokaryote divides, it simply replicates the DNA and splits in half. The process is a little more complicated than this, as DNA must first be unwound by special proteins. Although the DNA in prokaryotes usually exists in a ring, it can get quite tangled

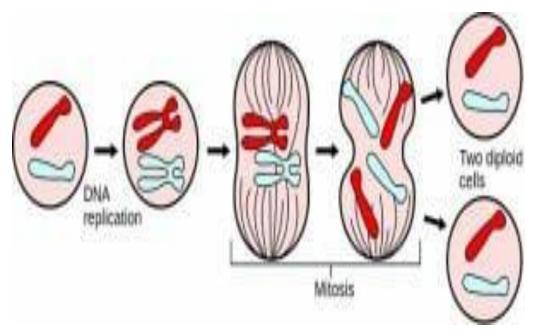
when it is being used by the cell. To copy the DNA efficiently, it must be stretched out. This also allows the two new rings of DNA created to be separated after they are produced. The two strands of DNA separate into two different sides of the prokaryote cell. The cell then gets longer, and divides in the middle. The process can be seen in the image below.

The DNA is the tangled line. The other components are labeled. Plasmids are small rings of DNA that also get copied during *binary fission* and can be picked up in the environment, from dead cells that break apart. These plasmids can then be further replicated. If a plasmid is beneficial, it will increase in a population. This is in part how antibiotic resistance in bacteria happens. The ribosomes are small protein structures that help produce proteins. They are also replicated so each cell can have enough to function.

Eukaryotic Cell Division: Mitosis

Eukaryotic organisms have membrane bound organelles and DNA that exists on chromosomes, which makes cell division harder. Eukaryotes must replicate their DNA, organelles, and cell mechanisms before dividing. Many of the organelles divide using a process that is essentially *binary fission*, leading scientist to believe that eukaryotes were formed by prokaryotes living inside of other prokaryotes.

After the DNA and organelles are replicated during *interphase* of the cell cycle, the eukaryote can begin the process of mitosis. The process begins during prophase, when the chromosomes condense. If mitosis proceeded without the chromosomes condensing, the DNA would become tangled and break. Eukaryotic DNA is associated with many proteins which can fold it into complex structures. As mitosis proceeds to *metaphase* the chromosomes are lined up in the middle of the cell. Each half of a chromosome, known as *sister chromatids* because they are replicated copies of each other, gets separated into each half of the cell as mitosis proceeds. At the end of mitosis, another process called *cytokinesis* divides the cell into two new daughter cells.



All eukaryotic organisms use mitosis to divide their cells. However, only single-celled organisms use mitosis as a form of reproduction. Most multicellular organisms are sexually reproducing and combine their DNA with that of another organism to reproduce. In these cases, organisms need a different method of cell division. Mitosis yields identical cells, but meiosis produces cells with half the genetic information of a regular cell, allowing two cells from different organisms of the same species to combine.

Eukaryotic Cell Division: Meiosis

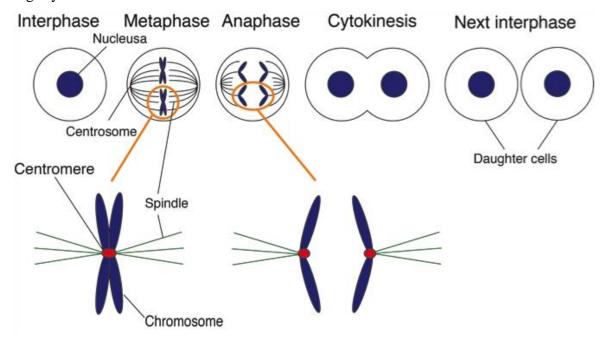
In sexually reproducing animals, it is usually necessary to reduce the genetic information before fertilization. Some plants can exist with too many copies of the genetic code, but in most organisms it is highly detrimental to have too many copies. Humans with even one extra copy of one chromosome can experience detrimental changes to their body. To counteract this, sexually reproducing organisms undergo a type of cell division known as *meiosis*. As before mitosis, the DNA and organelles are replicated. The process of meiosis contains two different cell divisions, which happen back-to-back. The first meiosis, *meiosis I*, separates homologous chromosomes. The homologous chromosomes present in a cell represent the two alleles of each gene an organism has. These alleles are recombined and separated, so the resulting daughter cells have only one allele for each gene, and no homologous pairs of chromosomes. The second division, *meiosis II*,

separated the two copies of DNA, much like in mitosis. The end result of meiosis in one cell is 4 cells, each with only one copy of the genome, which is half the normal number.

Organisms typically package these cells into *gametes*, which can travel into the environment to find other gametes. When two gametes of the right type meet, one will fertilize the other and produce a *zygote*. The zygote is a single cell that will undergo mitosis to produce the millions of cells necessary for a large organism. Thus, most eukaryotes use both mitosis and meiosis, but at different stages of their lifecycle.

Cell Division Stages

Depending upon which type of cell division an organism uses, the stages can be slightly different.



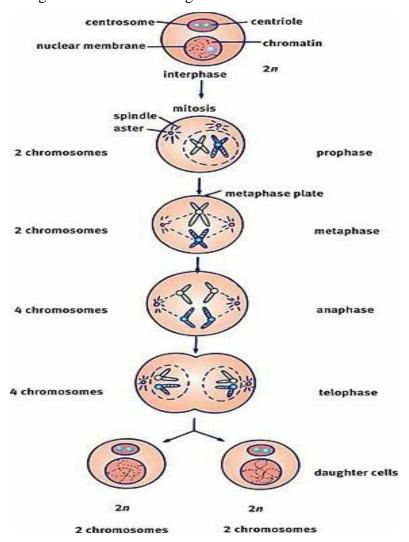
Mitosis Stages

Mitosis starts with *prophase* in which the chromosome is condensed. The cell proceeds to *metaphase* where the chromosomes are aligned on the metaphase plate. Then the chromosomes are separated in *anaphase* and the cell's cytoplasm is pinched apart during *telophase*. *Cytokinesis* is the final process that breaks the cell membrane and divides the cell into two.

Meiosis Stages

The stages of meiosis are similar to mitosis, but the chromosomes act differently. Meiosis has two phases, which include two separate cell divisions without the DNA replicating between them. *Meiosis I* and *meiosis II* have the same 4 stages as mitosis: prophase, metaphase, anaphase, and telophase. Cytokinesis concludes both rounds of meiosis.

prophase I, the chromosomes are condensed. In metaphase I, the chromosomes line up across from their homologous pairs. When they are separated in anaphase I and telophase I, there is only one form of each gene in each cell, known as a reduction division. Meiosis II proceeds in the same manner as mitosis, which sister chromatids dividing on the metaphase plate. By telophase II, there are 4 cells, each with half of the alleles as the parent cell and only a single copy of the genome. The cells can now become gametes and fuse together to create new organisms.

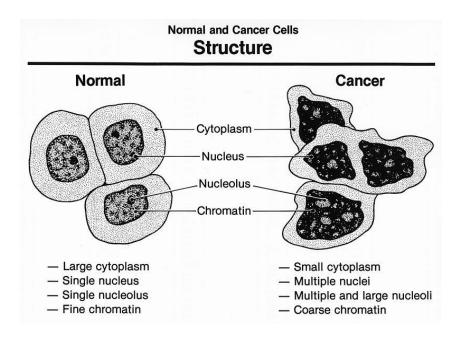


CYTOLOGY OF CANCER CELL

Cancer cells are cells that divide relentlessly, forming solid tumors or flooding the blood with abnormal cells. Cell division is a normal process used by the body for growth and repair. A parent cell divides to form two daughter cells, and these daughter cells are used to build new tissue or to replace cells that have died because of aging or damage. Healthy cells stop dividing when there is no longer a need for more daughter cells, but cancer cells continue to produce copies. They are also able to spread from one part of the body to another in a process known as metastasis.

There are different categories of cancer cell, defined according to the cell type from which they originate.

- Carcinoma, the majority of cancer cells are epithelial in origin, beginning in the membranous tissues that line the surfaces of the body.
- Leukaemia, originate in the tissues responsible for producing new blood cells, most commonly in the bone marrow.
- Lymphoma and myeloma, derived from cells of the immune system.
- Sarcoma, originating in connective tissue, including fat, muscle and bone.
- Central nervous system, derived from cells of the brain and spinal cord.
- Mesothelioma, originating in the mesothelium; the lining of body cavities.



Cancer cells have distinguishing histological features visible under the microscope. The nucleus is often large and irregular, and the cytoplasm may also display abnormalities.

The shape, size, protein composition, and texture of the nucleus are often altered in malignant cells. The nucleus may acquire grooves, folds or indentations, chromatin may aggregate or disperse, and the nucleolus can become enlarged. In normal cells, the nucleus is often round or ellipsoid in shape, but in cancer cells the outline is often irregular. Different combinations of abnormalities are characteristic of different cancer types, to the extent that nuclear appearance can be used as a marker in cancer diagnostics and staging.

Cancer cells are created when the genes responsible for regulating cell division are damaged. Carcinogenesis is caused by mutation and epimutation of the genetic material of normal cells, which upsets the normal balance between proliferation and cell death. This results in uncontrolled cell division in the body. The uncontrolled and often rapid proliferation of cells can lead to benign or malignant tumours (cancer). Benign tumors do not spread to other parts of the body or invade other tissues. Malignant tumors can invade other organs, spread to distant locations (metastasis) and become life-threatening.

More than one mutation is necessary for carcinogenesis. In fact, a series of several mutations to certain classes of genes is usually required before a normal cell will transform into a cancer cell.

Damage to DNA can be caused by exposure to radiation, chemicals, and other environmental sources, but mutations also accumulate naturally over time through uncorrected errors in DNA transcription, making age another risk factor. Oncoviruses can cause certain types of cancer, and genetics are also known to play a role.

Stem cell research suggests that excess SP2 protein may turn stem cells into cancer cells. However, a lack of particular co-stimulated molecules that aid in the way antigens react with lymphocytes can impair the natural killer cells' function, ultimately leading to cancer

Aging changes in organs, tissues, and cells

All vital organs begin to lose some function as you age during adulthood. Aging changes occur in all of the body's cells, tissues, and organs, and these changes affect the functioning of all body systems.

Living tissue is made up of cells. There are many different types of cells, but all have the same basic structure. Tissues are layers of similar cells that perform a specific function. The different kinds of tissues group together to form organs.

There are four basic types of tissue:

Connective tissue supports other tissues and binds them together. This includes bone, blood, and lymph tissues, as well as the tissues that give support and structure to the skin and internal organs.

Epithelial tissue provides a covering for superficial and deeper body layers. The skin and the linings of the passages inside the body, such as the gastrointestinal system, are made of epithelial tissue.

Muscle tissue includes three types of tissue:

- Striated muscles, such as those that move the skeleton (also called voluntary muscle)
- Smooth muscles (also called involuntary muscle), such as the muscles contained in the stomach and other internal organs
- Cardiac muscle, which makes up most of the heart wall (also an involuntary muscle)

Nerve tissue is made up of nerve cells (neurons) and is used to carry messages to and from various parts of the body. The brain, spinal cord, and peripheral nerves are made of nerve tissue.

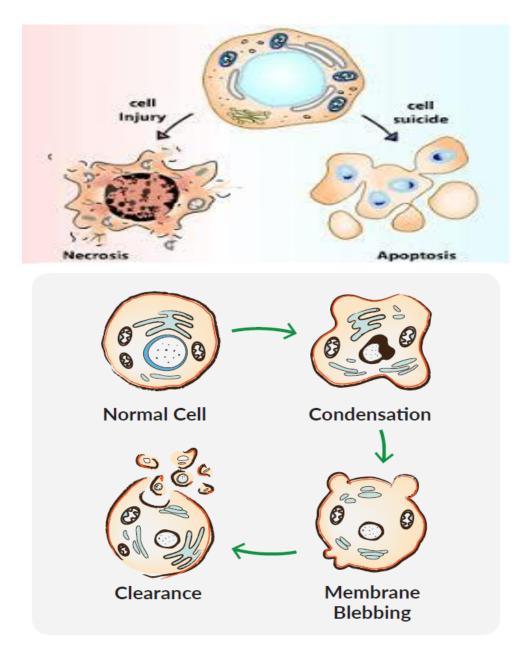
AGING CHANGES

Cells are the basic building blocks of tissues. All cells experience changes with aging. They become larger and are less able to divide and multiply. Among other changes, there is an increase in pigments and fatty substances inside the cell (lipids). Many cells lose their ability to function, or they begin to function abnormally.

As aging continues, waste products build up in tissue. A fatty brown pigment called lipofuscin collects in many tissues, as do other fatty substances.

Connective tissue changes, becoming more stiff. This makes the organs, blood vessels, and airways more rigid. Cell membranes change, so many tissues have more trouble getting oxygen and nutrients, and removing carbon dioxide and other wastes.

Many tissues lose mass. This process is called atrophy. Some tissues become lumpy (nodular) or more rigid.



Because of cell and tissue changes, your organs also change as you age. Aging organs slowly lose function. Most people do not notice this loss immediately, because you rarely need to use your organs to their fullest ability.

Organs have a reserve ability to function beyond the usual needs. For example, the heart of a 20-year-old is capable of pumping about 10 times the amount of blood that is actually needed to keep the body alive. After age 30, an average of 1% of this reserve is lost each year.

The biggest changes in organ reserve occur in the heart, lungs, and kidneys. The amount of reserve lost varies between people and between different organs in a single person.

These changes appear slowly and over a long period. When an organ is worked harder than usual, it may not be able to increase function. Sudden heart failure or other problems can develop when the body is worked harder than usual. Things that produce an extra workload (body stressors) include the following:

- Illness
- Medicines
- Significant life changes
- Sudden increased physical demands on the body, such as a change in activity or exposure to a higher altitude

Loss of reserve also makes it harder to restore balance (equilibrium) in the body. Drugs are removed from the body by the kidneys and liver at a slower rate. Lower doses of medicines may be needed, and side effects become more common. Recovery from illnesses is seldom 100%, leading to more and more disability.

Side effects of medicine can mimic the symptoms of many diseases, so it is easy to mistake a drug reaction for an illness. Some medicines have entirely different side effects in the elderly than in younger people.

AGING THEORY

No one knows how and why people change as they get older. Some theories claim that aging is caused by injuries from ultraviolet light over time, wear and tear on the body, or byproducts of metabolism. Other theories view aging as a predetermined process controlled by genes.

No single process can explain all the changes of aging. Aging is a complex process that varies as to how it affects different people and even different organs. Most gerontologists (people who study aging) feel that aging is due to the interaction of many

lifelong influences. These influences include heredity, environment, culture, diet, exercise and leisure, past illnesses, and many other factors.

Unlike the changes of adolescence, which are predictable to within a few years, each person ages at a unique rate. Some systems begin aging as early as age 30. Other aging processes are not common until much later in life.

Although some changes always occur with aging, they occur at different rates and to different extents. There is no way to predict exactly how you will age.

TERMS TO DESCRIBE TYPES OF CELL CHANGES

Atrophy:

- Cells shrink. If enough cells decrease in size, the entire organ atrophies. This is often a normal aging change and can occur in any tissue. It is most common in skeletal muscle, the heart, the brain, and the sex organs (such as the breasts and ovaries). Bones become thinner and more likely to break with minor trauma.
- The cause of atrophy is unknown, but may include reduced use, decreased workload, decreased blood supply or nutrition to the cells, and reduced stimulation by nerves or hormones.

Hypertrophy:

- Cells enlarge. This is caused by an increase of proteins in the cell membrane and cell structures, not an increase in the cell's fluid.
- When some cells atrophy, others may hypertrophy to make up for the loss of cell mass.
 Hyperplasia:
- The number of cells increases. There is an increased rate of cell division.
- Hyperplasia usually occurs to compensate for a loss of cells. It allows some organs and tissues to regenerate, including the skin, lining of the intestines, liver, and bone marrow.
 The liver is especially good at regeneration. It can replace up to 70% of its structure within 2 weeks after an injury.
- Tissues that have limited ability to regenerate include bone, cartilage, and smooth muscle (such as the muscles around the intestines). Tissues that rarely or never regenerate include the nerves, skeletal muscle, heart muscle, and the lens of the eye. When injured, these tissues are replaced with scar tissue.

Dysplasia:

- The size, shape, or organization of mature cells becomes abnormal. This is also called atypical hyperplasia.
- Dysplasia is fairly common in the cells of the cervix and the lining of the respiratory tract.
 Neoplasia:
- The formation of tumors, either cancerous (malignant) or noncancerous (benign).
- Neoplastic cells often reproduce quickly. They may have unusual shapes and abnormal function.

As you grow older, you will have changes throughout your body, including changes in:

- Hormone production
- Immunity
- The skin
- Sleep
- Bones, muscles, and joints
- The breasts
- The face
- The female reproductive system
- The heart and blood vessels
- The kidneys
- The lungs
- The male reproductive system
- The nervous system

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46Jn6ww : Binh อาการ หนุ่มอย หลงผู้ มุลท เอาเกบทูมลา (i) மூல் வட்ட வடி வமாகவும் வூலில் கரியை திவும் திறுனுமை திருத்திர் நாவிவியில்.

(11) 2 CAGO ULNIE & ORAWA OBALNESMAS.

(111) BESTREONLY LAM BLONLWASY 6, BOOT GARY 6 CONZED SON MAY

UN DEAG GOOD COMPUT SIZINGAMY.

(IV) Him Lism By noot of on Moon you Dongsom 2 Dunish Smy

(i) orginaria (BESABLA CANGO ORLUZ CARLON MALINE ன்பண்புள் இழைகள்ள புறுப்படுகியுள் அது அமைப்புள்

Home Dising of.

ப்ற டு தொடுமாகோம்ஸ் இறைகுகள் உடுவாகின்றனு.

(111) அதாடர்த்தியான கிரைகள் காணப்படுகிறது.

grooteun:

ப்) வுற்றவாடு இதாகமாகோமின் சென்ட்கோமியகும் Dami MAD Wy Day Mg.

(11) ARIN RONC GERM DIRING DO BESTELLE CON BANGAMAN May.

111) GBJBUF CANG DOOG SM STERNO 21/12 BOW GBJACON துடு உண்களை கோவக்கி கிழுக்கப்படுகிறது.

LABNUBUM:-

(i) உட்டு மணி மீண்கம் சேற்றன்றுகிறது.

(11) Floring LINNOWAS STON YOU 2 CAGO LLANGING

- 2. みららいのを強に付え、動かかるのは 風ををコカカのにものか、あめん DADEL GRUDING OF COMMENTO Browning - Doming 68378648876 GLONLWIND OSG MARNUYLE BOOKL LOMONNE கையையும் கொண்டுக்கன்றன.
- 3. 4ம்வக்டாலுண்டாக்: இளவ J வடிவ அவ்வது ட வடிவ BBONBLABBRUAM BDAMMIN ODDING BINGWIN SIMBERMYW BEUVERWIN Stones Die ODBAM Deuny DIE Browniu GANY.
- இளவ V வடிவ இஞாமோக்காம்கள் கியற்றில் 4. ODULTOBONICITÀ! அன்ட்டுராமியர் கைய பக்தியுவ் அமைந்துள்ளது. அனவே இரு Sound south obnown Lynn Brown US Rosingon.
- த். வாபுதன்ர யுழ்; இஇ விருன்ரவிரையுள்ள வயவைற்றவைற்ற வூழ்.
- 6. பாவிலைக்கார்: வசன்ட்டோயியர் வுடூ கிடர்தீல் வதனிவாக JANUWIUN LIDDERTS ENEWINDERMY.
 - Bener Bandon Downship;
- CANCIANT, DAN CERTACROA CONCIAN CLON DULY SOMERWAS.

Sombiy:

BBJABUH BANGAM BEJABUNOTILAONDUY; OHONICBJAGO

களையும் கொண்டிக்கின்றன. EBITBUTON 69: 046 BIERINL DOMQUENLUBTE GARDINAMY. And Allmir Dong BESTEUTERTUT OFORTUGENYS - Day മിട്ടത്ത്ര, ജനത്തി, அപ്പു அച്ച് Вமற்பட்ட இழைகளை OBNOON LAND BROWN OURDAY. @BITELETON WIND 2 mont இரைகள் சுன்கோவடான்று இடு அடிகள் அற்றிக் செல்லைகு

Brood UUGBMZ. (i) பாராணியிக் அடுள்கள் (Paranemic coils): இறைகள் व्यंकाठा मार्का मेड्सीकों मी ठार्का मार्ग मार्ग मार्थ Honor Sangna Homas Know this AGMAN OfonisuGEngs.

(11) Dorio & Lnoof was A Ginaria (Plectonemic coils): Dongain என்தில் மிரிக்க இயலாத அன்றல் சிமைந்து காணம்பட்டால் July Doring Connelling Hamanin dooriversmay.

Cho Styld Gray) Bour undergray To nught Charlen Garage of conditor () Trug to one of the conditor of the cond

Bomben foung in வர்க் இதுபகும் தேன்கு விலார் கிறார்க்குவர்.

மிக குறுகி உறைந்து உடக்குறுள் சிறைய கிறுக்காகுளம்கள் உடக்கு கணி மறைந்து உடக்குறுள் சிறை பற்புகிறுகி: உடக்கு கணி மறைந்து அக்கிறுகு.

BRITER BOWI: WIDER DAME BOWN BOY OF BODD WIDER BORNER OF STORED BORNER B

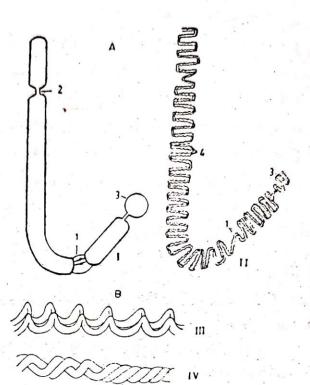
ECNAGUM I: DEGAGUA GANGAM AGM ANGIDAM ON DESING COMPLETON SOME CONTROL OF MONTH DE CONTROL OF MONTH DE CONTROL OF MONTH DE CONTROL DE CAMBON DANAM 2000 DENAMENTON 2000 DENAMENTON.

Down Love Down my Many (Second Merotic division)

திரண்டாம் டூன்று பிரிஅ கிடு கூம் ஏற்றையச் செக்களில் நடைவெறும் மறைடுகப் பிரிஅ ஆடும். கிதன் விறையச் வு வ்வவாகி ஏழ்ற்று பிக்கின்றன் -கேராற்று பிக்கின்றன் -

48778 ULNGO 2 CAG Grandy ம் மறைய்கு உடுவாகின்றன.

DECLE BUNII: BEINBUT EDEL WORD ON COM DE COM



படம் 56 மெட்டா சென்ட்ரிக் குரோமோசோமின் அமைப்பு

1.	புற அமைப்பு	19		
1.	சென்ட்ரோமியர்	II.	உட்புற அமைப்பு	
٥.	FILL MOVE	2.	ரெண்டாம் நிலை ஒடு	1881
В.	Durana are	4.	இருகுரோமோனிமேட்	LI
III.	இருவித குரோமோ பாரானிமிக் சுருள்க	TO STORE OF		
	[பெலிக்கி	611	~	[3 0]
கூற	ப்படுவது இ	இடையீட் புப்	பொருள் கொப்பு	370

45 BESTENDER BONGAM (Guant chaomosomes)

Bu Oberson Auginer Boursey, Objorge Hussing 43 @BJABUA CANGON OBNOOIL 21: BBBBBBB OUNTY ONING பு இப் நாகுருவ் நல்கிரை வயர்றியில் இடுவடுவர்கள்

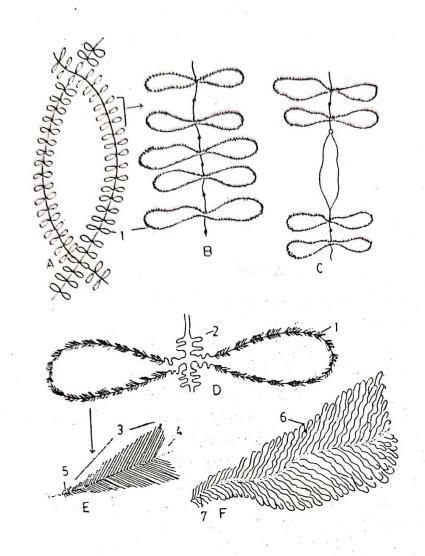
1. Trugger DesugenBeng By:-

இவ் வகை இதோகுமாகுகாம்கள் டிப் டீறா புத்திகளின் உடிக்கும் சுரர்வுல் மெய்கும் இம்வுகள் அது வைற்றின் Sousaniel Oberamin Brownin Obendo. Mun of 1861- or Unalle a Clarent Concerned Sout Logingoni. Chronin Di OUWACLAND. LIMON BON BERNBUM BANGAMA GE BOMAS FOR ERENGED ALON LANGE OFFENDEN CERTAINS FORM) กลุก เภาม Brossin บอริกาศ. มีณ ปนพามองกัน อาบบะลาเมลา of mid (Besnewa Genie Homeiyam dinay knowl) அளையவிகள்ளு அகாண் முடுக்கின் அன .

வாம்ப்பிரன் இதோகமாகோம்கள்:-

பினம்மில் 1882 - வ் இடு உரம்வகரின் பிண்டிரெவ்கரில் omicionson BESTEUTCHNG Amort Chyonia Boom Lini. Dona มหางาริสา Begrebog Concessor DILi กมกับควา Ama 100H Bongio 20H HANGE 2001 LUBRIO 46516 DON Bonnin EnoutiveExmes.

சுவ்வைற்கு எரம்பிரத்த இவோகமாகோகும் கூடு Honliy: வடு அத்து மற்றும் பக்க வளைஅகளை கொண்டுள்ளது. BO HEA BOOK BESTERLY CAMON ASOSTANON PO அச்சின் கேய் அடு உரிசை இதோகமாகியாகள் கோணுப் பகிறுவு. BETTEUT CONDIN JOH DILMLE JOB BETTEUT CONGING 15 mondo @Burkoncy &mnn 250018. Dis @Burloncy com இதிரும் வரிமாக்கள் பமல்லிய வளையாக்களில் பக்கு வக்கில் இதாகுப்பதினால் லாம்பர்த் ககோகமாகோம்கள் டிரேற் உடிறல் Broodi U Ga mas.



படம் 58 – லாம்ப்பிரஷ் குரோமோசோம்

A அமைப்பட

のあがいかり cell division

ത്യമാവന്ത്ര 2 un A ത്യാഴ്ച് ചാണ് ർ 8 മ്പ് ക് ചുത് വഴക്ക് ONTINA THE MIELLINE ON DIRESERED TO SOME SURVEY SULLENDER DLNN ZMM OHNO OHNBANG (FINGU ZMM OHNBM பிரிவடையதன் டூவம் கேரான்றுகிறது. உட்கடு சுகாண்ட கெவ் களின் பிறிவு இடு ஆன்கேறாவடான்று இணைக்கு றசமவக்களான 21:00 Drain (Karyokinesis) Lomposio ondi Confinatio பிரிதல் Ccytokinesis) அதலையற்றின் சேலம் கடைபெறுகிறது. താക് വസ്ചുക്കിൽ ചത്തുക്ക് !

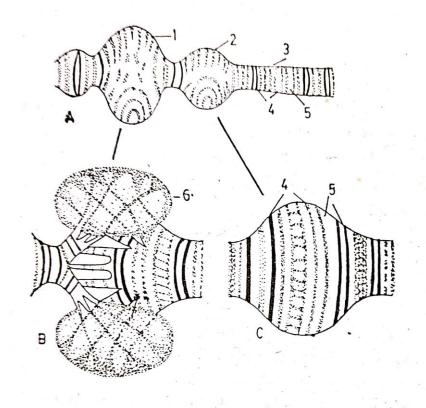
தாவர மற்றும் விலங்டு வசுக்களில் டுன்று அமைசுமான அத்த நிதிவுக்கு காணப்பகோது. 1. BBy @20 My Any Denro (Amitosia) 2. possinter Dest Digas onler ruly (untosis) 3. (B) sing mindy sindy would (Meiosis)

கேச்சேல் பிரிவு

Crowsi Day Line Comunisting Longing Hesincela Can มกลัลที่ Eu กลักที่ Oy @ อาล อ เกลือ คลั้ง มกลาคา ADDIONOBASE BOOLOUPED ODENGES BOOLOUPED MAS. SON (4 grownier 2 word ord misson of BEGUL NISSENT ON ചെഷ്ട്രുക്കാര് വാനമും ഒടുന്നും വാളവും വായിച്ചായി உட்கடு நெண்டு மையர்தில் இழிந்து திரண்டாகப் நடிகிறது. 2 - 200 Donasia Oburinas ont Ernambe During 20 oyes unglowned Bom orange Baroning alm of.

முறை முல் புயுவு (Mitosis)

LOON MOND WITH Concers Openionalin Bon Lours my முறைகேல் பிரிலன் டூவர் கேறான்றும் இடு கேல் தெரு ஆக ஆர் வீடு அன்றாகவும் சூகர் டுகுராகுமாககாம் சின்னிக்கை உடையதாகவும் Donalormont companional Do 2 mgi vombi y & smorts Osnoving Gradat my



படம் 58 – பாலிடீன் குரோமோசோம்கள்

- ^{ந் பெரிதாக்கப்பட்ட குரோமோசோம் பகுதி.}
- ^இ _{பெரிதாக்க}ப்பட்ட புடைப்பு (Puff)
- ^{(பெரிதாக்கப்பட்ட குமிழ் (Knob)}
- ! புடைப்பு
- ் புடைப்பற்ற பகுதி இடைப்பட்டைகள்
- குமிழ்
- பட்டைகள்
- பால்பியானி வளையம்

இ இது நடு மா இசாம் தன் முற்று ம் உடு அமுமனியை அற்றி தோற்று அத்தின்று வூ

முறை முகப் மரிவன் முக்கயத்துவம்;

1) இரோகமா சேசும்களின் பிண்ணிக்கை தாம் செய்வும் கிருப்புர் போன்றே கிடு சேய் சைய்களிலும் கிருக்கின்று?

11) DNA, RNA Am your de Homasin unganasio பக்கிறது.

Bringing Mry:

குள்ளுள் பிரிவு நடைவபறும் வபாதே இதோகமாகோம்கள் வுடு சுறையும் உடக்கு கைட்டே பிளிசம் இடு இரம்பைட் விடு சுறையும் உடக்கு கைட்டே பிளிசம் இடு இரம்பைட் கேன்று பிரிவு நடைவபறும் வபாதே இதோகமாகோம்கள்

(First Mew Hic Division)

Door Loome: Due Decidina romas, Age Amal and ingerin

HJAi 26 20 20 10 6 8 Mg

(P) God ON LELD STOR & mond i

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LID COM ON LELD STOR (N) ON LELD STOR (N) ON LELD STOR

(O) MOND COM (N) ON LIED STOR (N) ON LUE COM

(N) MOND COM (N) ON LUE COM

(N) MOND COM

(N)

- (1) முன் லாப் டோனி மா: இதோமோசோப்கள் முக லமல்லியராத நீன்டு அடுள்களற்று ஒற்றையாத நூலிழைகள் போன்று கோணப் மக்கிறது.
- (11) ONLIGHTON : BEDINGUT GANGEM BOOM BENDER DISTONERS DISTONERS OF STREET OF STREET BEDINGS STEPS BOOM BENDER BANDERS.
- Bennishon May. Bango as and wood of Shooksis) around of Bennishon of an envisor Bennish of an envisor Bennish of an envisor Bennish of an envisor Bennish of an envisor of

இதாகமானியா இழைகள் அடுள் வது அவற்றின் நீனர்ளதப் தாறுத்து அமைகிறது. துவ் பிரித்வன் போது (மாறுடுக் மற்றும் இண்றுவ் பிருவல்) மேண்று வது அடுள்கள் காணப்படுகிறது. (1) பெரிய அடுன்கள் (major ceils): இவை 10 சுத்வ 30 வைகிகள் தொண்ட அடுள்கள்.

(i) Infu Hamam (minor coils): ODUNG Hamamas OD-niss

(iii) Benuncian Aginam (Somatic coils): Binging Wiresing Oberwalin Complete Oberwaling Bungon By Connician Obersamin Complete Alaman Connician Haman Connician

BEJOBUTUNUS: LOSSINGES LON 1866 BONTY DAYS THOSE

DUNGES ON WHOM BEJOBUT CON WORD DESTOR CONTROP

BONG CONT CONT DEC HINDS CONTROL DESTOR OF MEDICAL DESTOR CONTROL BONGE DESTOR SON BOOM DUEST MAS DESTOR CONTROL BONTY OF MEDICAL DESTOR BEJOR CONTROL BONTY DESTOR BEJOR CONTROL BONTY DESTOR BEJOR CONTROL BONTY TONIONEST DESTOR DESTOR OF TONIONEST DESTOR CONTROL DESTOR DESTOR OF TONIONEST DESTOR OF TONIONES

குராகமாகியர்கள் அந்திய பகுதி சென்ட்கோகியல் சின்பகு அடு சுன்டகோகியர் கணைப்பட்டுள் கோகனர் சென்டாக், திரண்டு சிர்வல் கேர்வட்ட சென் உதோ மியர் காணப்பட்டால் டைசென்ட்டிக் டிர்மும் பாலி சென்ட்டிக் சினப் பகிறுக்

கிகாகமா கொல்களின் LeJunan குகியர்குவம்;

- 1. DE GONGON GONNAMM DOMONTAGE DOMON OUNA ANOMICOLINA
- 2. DNA Hindy RNA 25DW LOTY GUNGCAM Quingin
- 3. BBINBUN GANGAM, AND MONTON DNA-WIN ZMONT LIYZ OAWSAMM GONGOMM GONOGOMMANSO FINA KNANANSO NOSMASO.

איסחופטות ובשנה בו בשנים וחבו שובש חבו שובשות שונים והים שונים ובשנים ו Grand ABINGENERIA Dongomin AGSAJfonni Danio ucocaminal.

ICENTEUN []: 2 CAG GOOD 2 GONDAMB. 2 CAGULAGO Serverel, entrer Tours Della Berrand Del

இன்றுற்பிரிவின் முக்கியர்வுவம்;

(1) Aringy Dainy or Bingsunson, ALLunson @BINBUNGONG புண்ணிக்கையை உயாரினாவுகளில் நிலைபடுத்துகிறது.

(11) 2 Withom Birmon Coo Burgun Grin Bon Louring Unfrancis தீன் டூலம் புதிய உயர்கினால்களைத் தோற்ற உலக்கான்றன.

08 में सदा के की

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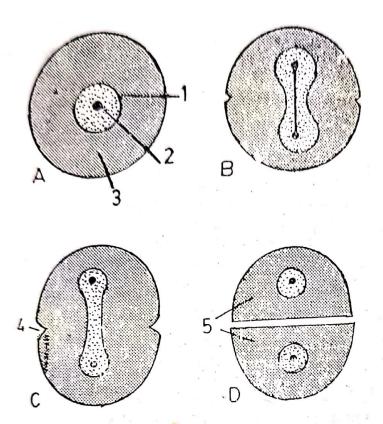
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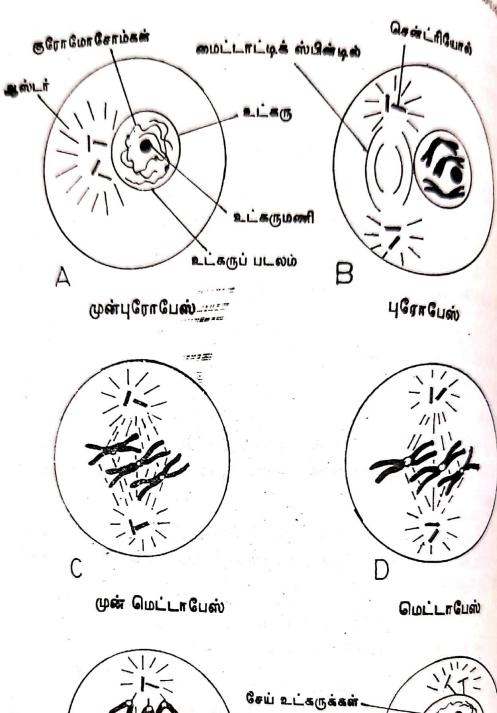
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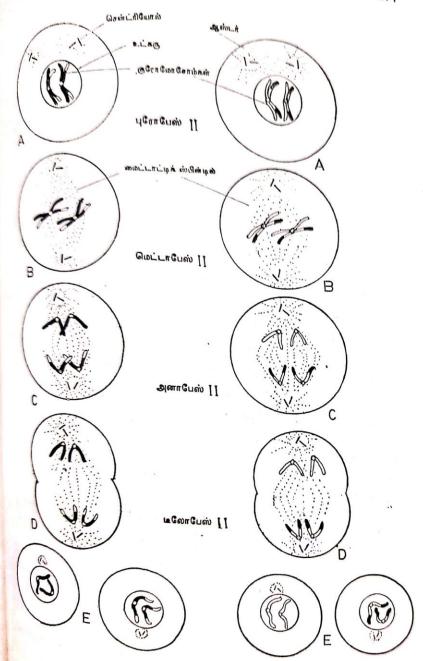
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படம் 97 – குன்றற் பிரிவு II-ன் படிநிலைகள்

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UNIT: IV

DNA: MOLECULAR STRUCTURE

- I Primary structure of the molecule: covalent backbone and bases aside
- I-1 Phosphoric acid
- I-2 Sugar
- I-3 Nitrogenous bases
- II Secondary and tertiary structures of the molecule -Three-dimentional conformation of DNA
- II.1 Dinucleotides
- II.2 DNA molecule
- II.2.1 Hydrogen bonds: bases pairing
- **II.2.2** Major groove and minor groove
- II.3 Non-B DNA
- **II.3.1 Z-DNA**
- II.3.2 Cruciform DNA and hairpin DNA
- II.3.3 H-DNA or triplex DNA
- **II.3.4 G4-DNA**
- **III** Quaternary structure of the molecule Chromatin
- IV Various
- IV.1 DNA and mitochondria
- **IV.2 DNA denaturation**

Deoxyribonucleic acid (DNA) IS the genetic information of most living organisms (a contrario, some viruses, called retroviruses, use ribonucleic acid as geneticinformation).

- -DNA can be copied over generations of cells: DNA replication
- DNA can be translated into proteins: DNA transcription into RNA, further ranslated into proteins, DNA can be repaired when needed: DNA repair.

Ribonucleic acids (RNAs) are described in another chapter (mRNA, r-RNA, t-RNA...)

- -DNA is a polymere, made of units called nucleotides (or mononucleotides).
- Nucleotides also have other functions: (energy carriers: ATP, GTP; cellular respiration:
- NAD, FAD; signal transduction: cyclic AMP; coenzymes: CoA, UDP; vitamins: nicotinamide mononucleotide, Vit B2).

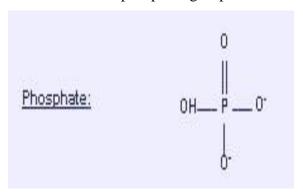
Using the protein nomenclature, we could speak in terms of primary, secondary, tertiary and quaternary structures of the molecule:

I Primary structure of the molecule: covalent backbone and bases aside

A nucleoside is made of a sugar + a nitrogenous base. A nucleotide is made of a phosphate + a sugar + a nitrogenous base. In DNA, the nucleotide is a deoxyribonucleotide (in RNA, the nucleotide is a ribonucleotide).

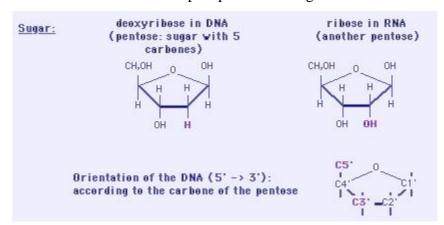
I-1 Phosphoric acid

Gives a phosphate group.



I-2 Sugar:

Deoxyribose, which is a cyclic pentose (5-carbon sugar). Note: the sugar in RNA is a ribose. Carbons in the sugar are noted from 1' to 5'. A nitrogen atom from the nitrogenous base links to C1' (glycosidic link), and the phosphate links to C5' (ester link) to make the nucleotide. The nucleotide is therefore: phosphate - C5' sugar C1' - base.

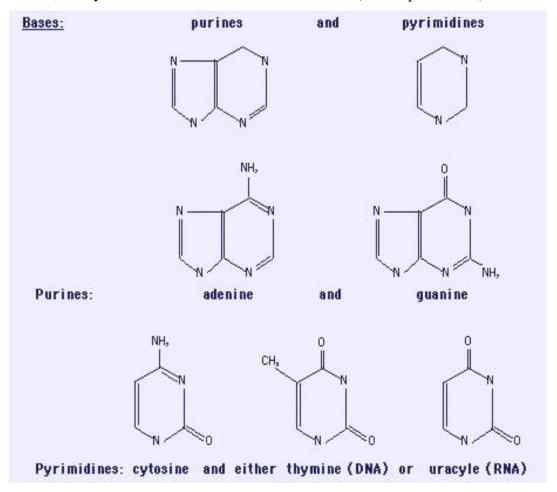


I-3 Nitrogenous bases:

Aromatic heterocycles; there are purines and pyrimidines.

- Purines: adenine (A) and guanine (G).- Pyrimidines: cytosine (C) and thymine (T) (Note: thymine is replaced by uracyle (U) in RNA).

Note: other nitrogenous bases exist, in particular methylated bases derived from the above mentioned; methylation of the bases has a functional role (see chapter ad hoc).



Glossary:

- Nucleoside names: deoxyribonucleosides in DNA: deoxyadenosine, deoxyguanosine, deoxycytidine, deoxythymidine in DNA (ribonucleosides in RNA: adenosine, guanosine, cytidine, uridine).
- Nucleotide names: deoxyribonucleotides in DNA: deoxyadenylic acid, deoxyguanylic acid, deoxycytidylic acid, deoxythymidylic acid (ribonucleotides in RNA: adenylic acid, guanylic acid, cytidylic acid, uridylic acid).

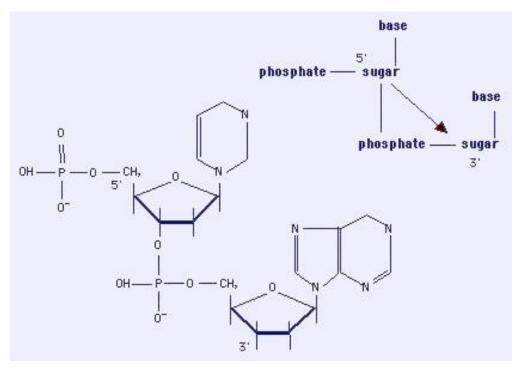
II Secondary and tertiary structures of the molecule - Three-dimentional conformation of DNA

II.1 Dinucleotides

Dinucleotides form from a phosphodiester link between 2 mononucleotides. The phosphate of a mononucleotide (in C5' of its sugar) being linked to the C3' of the sugar of the previous mononucleotide.

Then, we start with a phosphate, a 5' sugar (+base) and the 3' of this sugar, linked to a second phosphate - 5' sugar, which 3' is free for next step. The link -and the orientation of the molecule- is therefore 5' -> 3'.

Polynucleotides are made of the successive addition of monomeres in a general 5' -> 3' configuration. The backbone of the molecule is made of a succession of phosphate-sugar (nucleotide n) - phosphate-sugar (nucleotide n+1), and so on, covalently linked, the bases being aside.



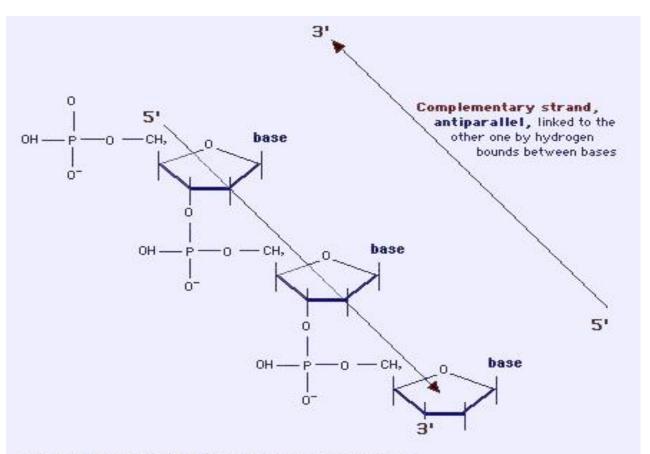
II.2 DNA molecule

DNA is made of two ("duplex DNA") dextrogyre (like a screw; right-handed) helical chains or strands ("the double helix"), coiled around an axis to form a double helix of $20A^{\circ}$ of diameter.

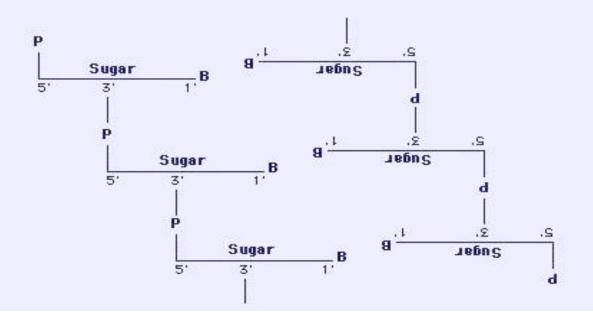
The two strands are antiparallel (id est: their 5'->3' orientations are in opposite direction). The general appearance of the polymere shows a periodicity of 3.4 A°, corresponding to the distance between 2 bases, and another one of 34 A°, corresponding to one helix turn (and also to 10 bases pairs).

II.2.1 Hydrogen bounds: bases pairing

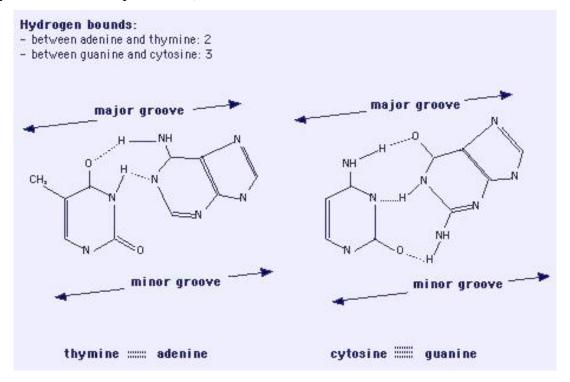
The (hydrophobic) bases are stacked on the inside, there planes are perpendicular to the axis of the double helix. The outside (phosphate and sugar) are hydrophilic. Hydrogen bounds between the bases of one strand and that of the other strand hold the two strands together.



DNA is double strand: the 2 strands are antiparallel -they run in opposite direction- and complementary. (RNA is single strand)



A purine on one strand shall link to a pyrimidine on the other strand. As a corollary, the pyrimidine purines residues equals the number of A binds T (with 2 hydrogen bounds).G binds C (with 3 hydrogen bounds: more stable link: 5.5 kcal vs 3.5 kcal). Note: the content in A in the DNA is therefore equal to the content T. in G equals the and the content content This strict correspondance (A<->T and G<->C) makes the 2 strands complementary. One is the template of the other one, and reciprocally: this property will allow exact replication (semi-conservative replication: one strand -the template- is conserved, another is newly synthesized, same with the second strand, conserved, allowing another one to be newly synthesized; see chapter ad hoc).

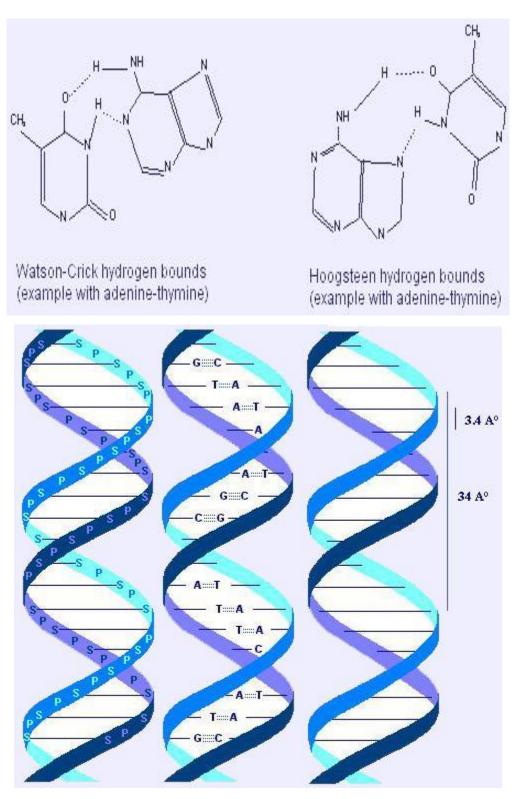


Notes:

Hydrogen bounds in base pairing are sometimes different from the model of Watson and Crick above described, using the N7 atom of the purine instead of the N1 (Hoogsteen model).

II.2.2 Major groove and minor groove

The double helix is a quite rigid and viscous molecule of an immense length and a small diameter. It presents a major groove and a minor groove.



The major groove is deep and wide, the minor groove is narrow and shallow. DNA-protein interactions are major/essential processes in the cell life (transcription activation or repression, DNA replication and repair).

Proteins bind at the floor of the DNA grooves, using specific binding: hydrogen bounds, and non specific binding: van der Waals interactions, generalized electrostatic interactions; proteins recognize H-bond donnors, H-bond acceptors, metyl groups (hydrophobic), the later being exclusively in the major groove; there are 4 possible patterns of recognition with the major groove, and only 2 with the minor groove (see iconography).- Some proteins bind DNA in its major groove, some other in the minor groove, and some need to bind to both.

Notes:

- The 2 strands are called "plus" and "minus" strands, or "direct" and "reverse" strands. At a given location where one strand (any of the two) bears coding sequences, it is unlikely (but not impossible) that the other strand also bears coding sequences.- DNA is ionized in vivo and behave like a polyanion. The double helix as described above is the "B" form of the DNA; it is the form the most commonly found in vivo, but other forms exist in vivo (see below) or in vitro. The "A" form resemble B-DNA but it is less hydrated than B-DNA, "A" form is not found in vivo.

II.3 Non-B DNA

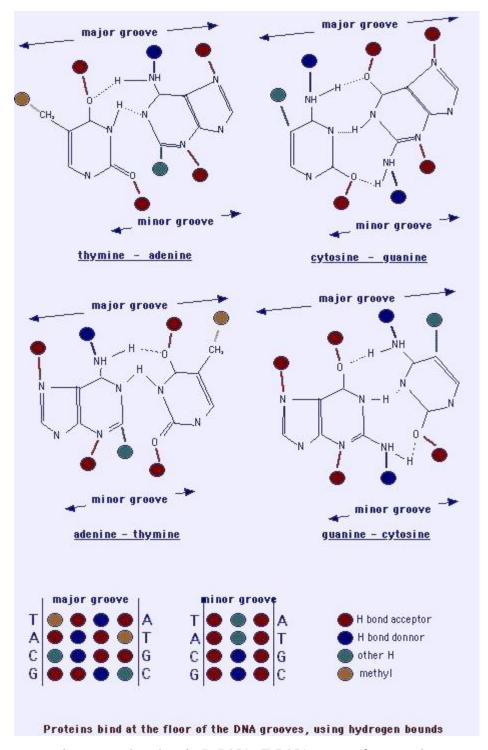
DNA is a molecule which moves, fidgets, does gymnastics, dances. The structures below cited are being proved to have funtional roles; on the other hand, they may favour DNA breaks and further deletions, amplification, recombination, and mutations.

Glossary:

Palindromes: these are names that read the same backwards and forwards (e.g. "DNA LAND"). DNA uses to play with palindromes: see below).

II.3.1 Z-DNA

- Z form is a levogyre (left handed) double helix with a zig-zag conformation of the backbone (less smooth than B-DNA). Only one groove is observed, resembling the minor groove, the base pairs being set off to the side, far from the axis. The bases (which form the major groove -close to the axis- in B-DNA) are here at the outer surface.



Phosphates are closer together than in B-DNA. Z-DNA cannot form nucleosomes.- A high G-C content favours Z conformation. Cytosine methylation, and molecules which can be present in vivo such as spermine and spermidine can stabilize Z conformation.- DNA sequences can flip from a B form to a Z form and vice versa: Z-DNA is a transient form in

vivo.- Z-DNA formation occurs during transcription of genes, at transription start sites near promoters of actively transcribed genes.

During transcription, the movement of RNA polymerase induces negative supercoiling upstream and positive supercoiling downstream the site of transcription. The negative supercoiling upstream favours Z-DNA formation; a Z-DNA function would be to absorb negative supercoiling. At the end of transcription, topoisomerase relaxes DNA back to B conformation. Certain proteins bind to Z-DNA, in particular double-stranded RNA adenosine deaminase (ADAR1), a Z-DNA binding nuclear-RNA-editing enzyme; this enzyme converts adenine to inosine in the pre-mRNA. Following, ribosomes will interpret inosine as guanine, and the protein coded with this epigenetic modification will be different (see chapter on Epigenetics).

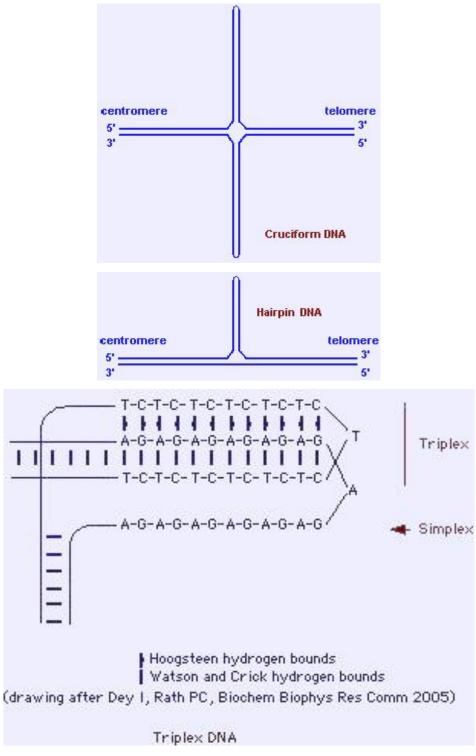
- Z-DNA antibodies are found in lupus erythematosus and other autoimmune diseases.
- Double stranded RNA (dsRNA) can adopt a Z conformation.

II.3.2 cruciform DNA and hairpin DNA

- Holliday junctions (formed during recombination) are cruciform structures. Inverted (or mirror) repeats (palindromes) of polypurine/polypyrimidine DNA stretches can also form cruciform or hairpin structures through intra-strand pairing.
- Palindromic AT-rich repeats are found at the breakpoints of the t(11;22)(q23;q11), the only known recurrent constitutional reciprocal translocation.
- Nucleases bind and cleave holliday junctions after recombination. Other well known proteins such as HMG proteins and MLL (for further reading, see: MLL) can also bind cruciform DNA.

II.3.3 H-DNA or triplex DNA

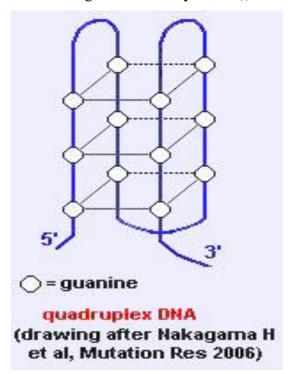
- Inverted repeats (palindromes) of polypurine/polypyrimidine DNA stretches can form triplex structures (triple helix). A triple-stranded plus a single stranded DNA are formed.-H-DNA may have a role in functional regulation of gene expression as well as on RNAs (e.g. repression of transcription).



II.3.4 G4-DNA

- G4 DNA or quadruplex DNA: folding of double stranded GC-rich sequence onto itself forming Hoogsteen base pairing between 4 guanines ("G4"), a highly stable structure. Often found near promotors of genes and at the telomeres.

- Role in meiosis and recombination; may be regulatory elements.
- RecQ family helicases are able to unwind G4 DNA (e.g. BLM, the gene mutated in Bloom syndrome (for further reading, see: Bloom syndrome)).

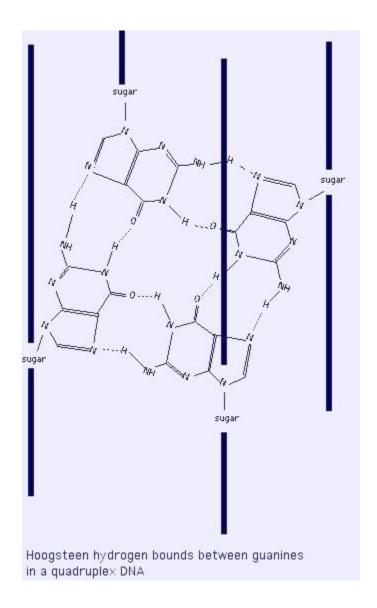


III Quaternary structure of the molecule - Chromatin

DNA is associated with proteins: histones and non histone proteins, to form the chromatin. DNA as a whole is acidic (negatively charged) and binds to basic (positively charged) proteins called histones: see chapter ChromatinThere is 3×10^9 nucleotide pairs in the human haploid genome representing about 30 000 genes dispersed over 23 chromosomes for a haploid set.

IV.1 DNA and mitochondria

- DNA is found in the nucleus of the cell, but a small amount is also present in the mitochondria.- Mitochondrias would originate from archeobacterias which became endosymbiotic to eukaryotic cells.- Their genetic code is different from the so-called



"universal" code (UGA, AUA, AGA, AGG: respectively STOP, Ile, Arg, Arg in the universal code, and Trp, Met, STOP, STOP in the mitochondria of mammals, and other meanings in mitochondria of other spieces).- The number of DNA copies in one given mitochondria is variable.- Mitochondrial DNA is circular, with a heavy and a light chains, has no introns, not any non-coding sequence.- Genes from the mitochondria code for proteins involved in electron transport, ribosomic RNAs (rRNAs), and transfer RNAs (tRNAs).

- Each DNA strand is transcribed, then cut into the mRNAs, but also into rRNAs and tRNAs.

Note: the mitochondria also use proteins imported from the cytoplasm of the cell (and coded by the nucleus); so far, proteins from the mitochondria are not exported into the cytoplasm except in case of apoptosis.

IV.2 DNA denaturation:

The double helix undergoes unwinding in vitro with heat, extremes ph, and other conditions (urea, ...). A melting point can be calculated; it is characteristic of the A/T versus G/C proportion of the specimen studied, due to the fact that there is only 2 hydrogen bounds in A/T, and 3 in G/C, a more stable binding. Upon denaturation, the physical properties of the DNA change; e.g. hyperchromic effect: light absorption at 260 nm is higher with denatured DNA than with double standed DNA. Light absorption also varies according to the A/T vs G/C proportion: it is higher in A/T rich specimens than in G/C rich ones.DNA denaturation is to be known, because: 1- it allows to measure A/T vs G/C content; 2- it is the basis of in situ hybridization techniques.

RNA: MOLECULAR STRUCTURE

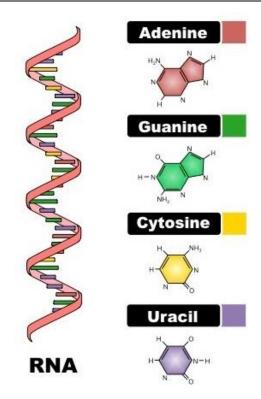
RNA, abbreviation of **ribonucleic** acid, complex compound of high molecularweight that functions in cellular protein synthesis andreplaces DNA (deoxyribonucleic acid) as a carrier of genetic codes in some viruses. RNA consists of ribose nucleotides (nitrogenous bases appended to a ribose sugar) attached by phosphodiester bonds, forming strands of varying lengths. The nitrogenous bases in RNA are adenine, guanine, cytosine, and uracil, which replaces thymine in DNA.

- RNA or ribonucleic acid is a polymer of nucleotides which is made up of a ribose sugar, a phosphate, and bases such as adenine, guanine, cytosine, and uracil.
- It is a polymeric molecule essential in various biological roles

in coding, decoding, regulation,

Figure: (a) Ribonucleotides contain the pentose sugar ribose instead of the deoxyribose found in deoxyribonucleotides. (b) RNA contains the pyrimidine uracil in place of thymine found in DNA.

RNA STRUCTURE



Like **DNA**, RNA is a long polymer consisting of nucleotides.

- RNA is a single-stranded helix.
- The strand has a 5'end (with a phosphate group) and a 3'end (with a hydroxyl group).
- It is composed of ribonucleotides.
- The ribonucleotides are linked together by $3' \rightarrow 5'$ phosphodiester bonds.

an

• The nitrogenous bases that compose the ribonucleotides include adenine, cytosine, uracil, and guanine.

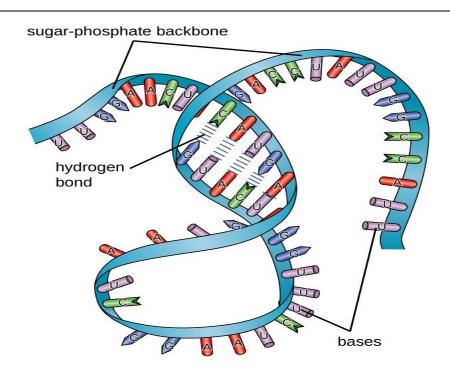
Thus, the difference in the structure of RNA from that of DNA include:

The bases in RNA are adenine (abbreviated A), guanine (G), uracil (U) and cytosine
 (C).

Thus thymine in DNA is replaced by uracil in RNA, a different pyrimidine. However, like thymine, uracil can form base pairs with adenine.

- The sugar in RNA is ribose rather than deoxyribose as in DNA.
- The corresponding ribonucleosides are adenosine, guanosine, cytidine and uridine. The corresponding ribonucleotides are adenosine 5'-triphosphate (ATP), guanosine 5'-triphosphate (GTP), cytidine 5'-triphosphate (CTP) and uridine 5'-triphosphate (UTP).

RNA Secondary Structure



- Most RNA molecules are single-stranded but an RNA molecule may contain regions
 which can form complementary base pairing where the RNA strand loops back on
 itself.
- If so, the RNA will have some double-stranded regions.

• Ribosomal RNAs (rRNAs) and transfer RNAs (tRNAs) exhibit substantial secondary structure, as do some messenger RNAs (mRNAs).

Types of RNA

In both prokaryotes and eukaryotes, there are three main types of RNA –

- rRNA (ribosomal)
- tRNA (transfer)
- mRNA (messenger)







Messenger RNA (mRNA)

Ribosomal RNA (rRNA)

Messenger RNA (mRNA)

- Accounts for about 5% of the total RNA in the cell.
- Most heterogeneous of the 3 types of RNA in terms of both base sequence and size.
- It carries the genetic code copied from the DNA during transcription in the form of triplets of nucleotides called codons.
- As part of post-transcriptional processing in eukaryotes, the 5' end of mRNA is capped with a guanosine triphosphate nucleotide, which helps in mRNA recognition during translation or protein synthesis.
- Similarly, the 3' end of an mRNA has a poly A tail or multiple adenylate residues added to it, which prevent enzymatic degradation of mRNA. Both 5' and 3' end of an mRNA imparts stability to the mRNA.

Function

mRNA transcribes the genetic code from DNA into a form that can be read and used to make proteins. mRNA carries genetic information from the nucleus to the cytoplasm of a cell.

Ribosomal RNA (rRNA)

- Found in the ribosomes and account for 80% of the total RNA present in the cell.
- Ribosomes consist of two major components: the small ribosomal subunits, which
 read the RNA, and the large subunits, which join amino acids to form a polypeptide
 chain. Each subunit comprises one or more ribosomal RNA (rRNA) molecules and a
 variety of ribosomal proteins (r-protein or rProtein).
- Different rRNAs present in the ribosomes include small rRNAs and large rRNAs, which denote their presence in the small and large subunits of the ribosome.
- rRNAs combine with proteins in the cytoplasm to form ribosomes, which act as the site of protein synthesis and has the enzymes needed for the process.
- These complex structures travel along the mRNA molecule during translation and facilitate the assembly of amino acids to form a polypeptide chain. They bind to tRNAs and other molecules that are crucial for protein synthesis.

Function

rRNA directs the translation of mRNA into proteins.

Transfer RNA (tRNA)

- tRNA is the smallest of the 3 types of RNA having about 75-95 nucleotides.
- tRNAs are an essential component of translation, where their main function is the transfer of amino acids during protein synthesis. Therefore they are called transfer RNAs.
- Each of the 20 amino acids has a specific tRNA that binds with it and transfers it to
 the growing polypeptide chain. tRNAs also act as adapters in the translation of the
 genetic sequence of mRNA into proteins. Therefore they are also called adapter
 molecules.

Structure of tRNA

tRNAs have a clover leaf structure which is stabilized by strong hydrogen bonds between the nucleotides. Apart from the usual 4 bases, they normally contain some unusual bases mostly formed by methylation of the usual bases, for example, methyl guanine and methylcytosine.

- Three structural loops are formed via hydrogen bonding.
- The 3' end serves as the amino acid attachment site.
- The center loop encompasses the anticodon.

- The anticodon is a three-base nucleotide sequence that binds to the mRNA codon.
- This interaction between codon and anticodon specifies the next amino acid to be added during protein synthesis.

DNA REPLICATION

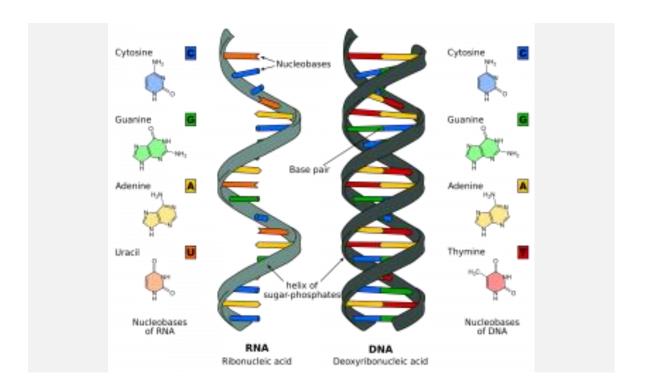
DNA replication, also known as **semi-conservative replication**, is the process by which DNA is essentially doubled. It is an important process that takes place within the dividing cell.

In this article, we shall look briefly at the **structure** of DNA, at the precise steps involved in **replicating** DNA (initiation, elongation and termination), and the **clinical consequences** that can occur when this goes wrong.

DNA is made up of millions of **nucleotides.** These are molecules composed of a deoxyribose sugar, with a phosphate and a base (or nucleobase) attached to it. These nucleotides are attached to each other in strands via phosphodiester bonds to form a 'sugar-phosphate backbone'. The bond formed is between the third carbon atom on the deoxyribose sugar of one nucleotide (henceforth known as the 3') and the fifth carbon atom of another sugar on the next nucleotide (known as the 5').

N.B: 3' is pronounced 'three prime' and 5' is pronounced 'five prime'.

There are two strands running in opposite or **antiparallel** directions to each other. These are attached to each other throughout the length of the strand through the bases on each nucleotide. There are 4 different bases associated with DNA; Cytosine, Guanine, Adenine, and Thymine. In normal DNA strands, Cytosine binds to Guanine, and Adenine binds to Thymine. The two strands together form a double helix.



Stages of DNA replication

DNA replication can be thought of in three stages; **Initiation, Elongation, Termination Initiation**

DNA synthesis is initiated at particular points within the DNA strand known as 'origins', which are specific coding regions.

These origins are targeted by initiator proteins, which go on to recruit more proteins that help aid the replication process, forming a replication complex around the DNA origin. There are multiple origin sites, and when replication of DNA begins, these sites are referred to as **replication forks**.

Within the replication complex is the enzyme **DNA Helicase**, which unwinds the double helix and exposes each of the two strands, so that they can be used as a template for replication.

It does this by hydrolysing the ATP used to form the bonds between the nucleobases, therefore breaking the bond holding the two strands together.

DNA Primase is another enzyme that is important in DNA replication. It synthesises a small **RNA primer**, which acts as a 'kick-starter' for **DNA Polymerase**. DNA

Polymerase is the enzyme that is ultimately responsible for the creation and expansion of the new strands of DNA.

Elongation

Once the DNA Polymerase has attached to the original, unzipped two strands of DNA (i.e. the **template** strands), it is able to start synthesising the new DNA to match the templates. It is essential to note that DNA polymerase is only able to extend the primer by adding free nucleotides to the **3' end.**

One of the templates is read in a 3' to 5' direction, which means that the new strand will be formed in a 5' to 3' direction. This newly formed strand is referred to as the **Leading Strand.**

Along this strand, DNA Primase only needs to synthesise an **RNA primer** once, at the beginning, to initiate DNA Polymerase. This is because DNA Polymerase is able to extend the new DNA strand by reading the template 3' to 5', synthesising in a 5' to 3' direction as noted above.

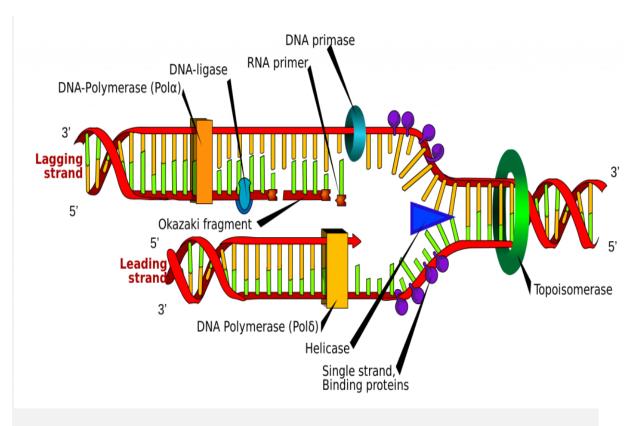
However, the other template strand (the **lagging strand**) is antiparallel, and is therefore read in a **5'** to **3'** direction. Continuous DNA synthesis, as in the **leading strand**, would need to be in the 3' to 5' direction, which is impossible as we cannot add bases to the 5' end. Instead, as the helix unwinds, RNA primers are added to the newly exposed bases on the **lagging strand** and DNA synthesis occurs **in fragments**, but still in the 5' to 3' direction as before. These fragments are known as **Okazaki fragments**.

Termination

The process of expanding the new DNA strands continues until there is either no more DNA template left to replicate (i.e. at the end of the chromosome), or two replication forks meet and subsequently **terminate.**

The meeting of two replication forks is not regulated and happens randomly along the course of the chromosome.

Once DNA synthesis has finished, it is important that the newly synthesised strands are bound and stabilized. With regards to the lagging strand, two enzymes are needed to achieve this; **RNAase H** removes the RNA primer that is at the beginning of each Okazaki fragment, and **DNA Ligase** joins fragments together to create one complete strand.



Diagrammatic representation of DNA replication

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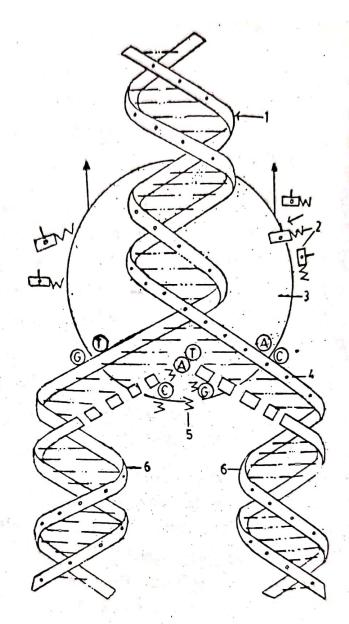
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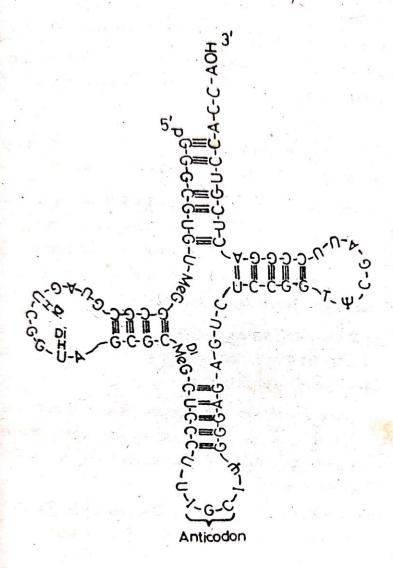
படம் 75 – DNA இரட்டித்தல்

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படம் 71 – DNA மூலக்கூறின் உருப்படிவம்

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UNIT: V

PROTEIN SYNTHESIS

Objectives

- Explain how the genetic code stored within DNA determines the protein that will form
- Describe the process of transcription
- Describe the process of translation

Most structural components of the cell are made up, at least in part, by proteins and virtually all the functions that a cell carries out are completed with the help of proteins. One of the most important classes of proteins is enzymes, which help speed up necessary biochemical reactions that take place inside the cell. Some of these critical biochemical reactions include building larger molecules from smaller components (such as occurs during DNA replication or synthesis of microtubules) and breaking down larger molecules into smaller components (such as when harvesting chemical energy from nutrient molecules). Whatever the cellular process may be, it is almost sure to involve proteins. Just as the cell's genome describes its full complement of DNA, a cell's **proteome** is its full complement of proteins.

Protein synthesis begins with genes. A **gene** is a functional segment of DNA that provides the genetic information necessary to build a protein. Each particular gene provides the code necessary to construct a particular protein. **Gene expression**, which transforms the information coded in a gene to a final gene product, ultimately dictates the structure and function of a cell by determining which proteins are made.

The interpretation of genes works in the following way. Recall that proteins are polymers, or chains, of many amino acid building blocks. The sequence of bases in a gene (that is, its sequence of A, T, C, G nucleotides) translates to an amino acid sequence. A **triplet** is a section of three DNA bases in a row that codes for a specific amino acid. Similar to the way in which the three-letter code *d-o-g* signals the image of a dog, the three-letter DNA base code signals the use of a particular amino acid. For example, the DNA triplet CAC (cytosine, adenine, and cytosine) specifies the amino acid valine. Therefore, a gene, which is composed of multiple triplets in a unique sequence, provides the code to build an entire protein, with multiple amino acids in the proper sequence. The

mechanism by which cells turn the DNA code into a protein product is a two-step process, with an RNA molecule as the intermediate.

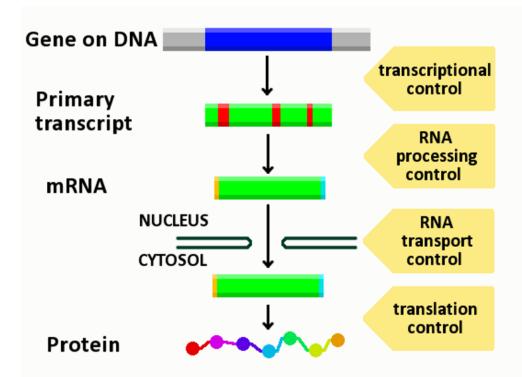
REGULATION OF GENE EXPRESSION

Gene expression is the process by which the instructions in our DNA are converted into a functional product, such as a protein.

- When the information stored in our DNA? is converted into instructions for making proteins? or other molecules, it is called gene expression?.
- Gene expression is a tightly regulated process that allows a cell to respond to its changing environment.
- It acts as both an on/off switch to control when proteins are made and also a volume control that increases or decreases the amount of proteins made.
- There are two key steps involved in making a protein, transcription and translation.

Steps I

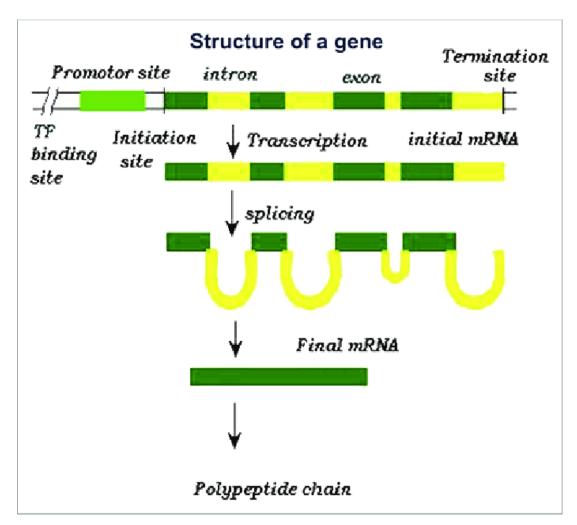
- Transcription is when the DNA in a gene? is copied to produce an RNA? transcript called messenger RNA? (mRNA).
- This is carried out by an enzyme? called RNA polymerase which uses available bases from the nucleus? of the cell to form the mRNA.
- RNA is a chemical similar in structure and properties to DNA, but it only has a single strand of bases? and instead of the base thymine ?(T), RNA has a base called uracil? (U).



Steps II

- Translation occurs after the messenger RNA (mRNA) has carried the transcribed 'message' from the DNA to protein-making factories in the cell, called ribosomes?.
- The message carried by the mRNA is read by a carrier molecule called transfer RNA ?(tRNA).
- The mRNA is read three letters (a codon) at a time.
- Each codon specifies a particular amino acid?. For example, the three bases 'GGU' code for an amino acid called glycine.
- As there are only 20 amino acids but 64 potential combinations of codon, more than one codon can code for the same amino acid. For example, the codons 'GGU' and 'GGC' both code for glycine.
- Each amino acid is attached specifically to its own tRNA molecule.
- When the mRNA sequence is read, each tRNA molecule delivers its amino acid to the ribosome and binds temporarily to the corresponding codon on the mRNA molecule.

- Once the tRNA is bound, it releases its amino acid and the adjacent amino acids all
 join together into a long chain called a polypeptide.
- This process continues until a protein is formed.
- Proteins carry out most of the active functions of a cell.



TRANSCRIPTION

DNA is housed within the nucleus, and protein synthesis takes place in the cytoplasm, thus there must be some sort of intermediate messenger that leaves the nucleus and manages protein synthesis. This intermediate messenger is **messenger RNA** (**mRNA**), a single-stranded nucleic acid that carries a copy of the genetic code for a single gene out of the nucleus and into the cytoplasm where it is used to produce proteins.

There are several different types of RNA, each having different functions in the cell. The structure of RNA is similar to DNA with a few small exceptions. For one thing, unlike DNA, most types of RNA, including mRNA, are single-stranded and contain no complementary strand. Second, the ribose sugar in RNA contains an additional oxygen atom compared with DNA. Finally, instead of the base thymine, RNA contains the base uracil. This means that adenine will always pair up with uracil during the protein synthesis process.

Gene expression begins with the process called **transcription**, which is the synthesis of a strand of mRNA that is complementary to the gene of interest. This process is called transcription because the mRNA is like a transcript, or copy, of the gene's DNA code. Transcription begins in a fashion somewhat like DNA replication, in that a region of DNA unwinds and the two strands separate, however, only that small portion of the DNA will be split apart. The triplets within the gene on this section of the DNA molecule are used as the template to transcribe the complementary strand of RNA (Figure 2). A **codon** is a three-base sequence of mRNA, so-called because they directly encode amino acids. Like DNA replication, there are three stages to transcription: initiation, elongation, and termination.

Stage 1: Initiation. A region at the beginning of the gene called a **promoter**—a particular sequence of nucleotides—triggers the start of transcription.

Stage 2: Elongation. Transcription starts when RNA polymerase unwinds the DNA segment. One strand, referred to as the coding strand, becomes the template with the genes to be coded. The polymerase then aligns the correct nucleic acid (A, C, G, or U) with its complementary base on the coding strand of DNA. **RNA polymerase** is an enzyme that adds new nucleotides to a growing strand of RNA. This process builds a strand of mRNA. Stage 3: Termination. When the polymerase has reached the end of the gene, one of three specific triplets (UAA, UAG, or UGA) codes a "stop" signal, which triggers the enzymes to terminate transcription and release the mRNA transcript.

Before the mRNA molecule leaves the nucleus and proceeds to protein synthesis, it is modified in a number of ways. For this reason, it is often called a pre-mRNA at this stage. For example, your DNA, and thus complementary mRNA, contains long regions called non-coding regions that do not code for amino acids. Their function is still a

mystery, but the process called **splicing** removes these non-coding regions from the pre-mRNA transcript (Figure 3). A **spliceosome**—a structure composed of various proteins and other molecules—attaches to the mRNA and "splices" or cuts out the non-coding regions. The removed segment of the transcript is called an **intron**. The remaining exons are pasted together. An **exon** is a segment of RNA that remains after splicing. Interestingly, some introns that are removed from mRNA are not always non-coding. When different coding regions of mRNA are spliced out, different variations of the protein will eventually result, with differences in structure and function. This process results in a much larger variety of possible proteins and protein functions. When the mRNA transcript is ready, it travels out of the nucleus and into the cytoplasm.

TRANSLATION

Like translating a book from one language into another, the codons on a strand of mRNA must be translated into the amino acid alphabet of proteins. **Translation** is the process of synthesizing a chain of amino acids called a **polypeptide**. Translation requires two major aids: first, a "translator," the molecule that will conduct the translation, and second, a substrate on which the mRNA strand is translated into a new protein, like the translator's "desk." Both of these requirements are fulfilled by other types of RNA. The substrate on which translation takes place is the ribosome.

Remember that many of a cell's ribosomes are found associated with the rough ER, and carry out the synthesis of proteins destined for the Golgi apparatus. **Ribosomal RNA (rRNA)** is a type of RNA that, together with proteins, composes the structure of the ribosome. Ribosomes exist in the cytoplasm as two distinct components, a small and a large subunit. When an mRNA molecule is ready to be translated, the two subunits come together and attach to the mRNA. The ribosome provides a substrate for translation, bringing together and aligning the mRNA molecule with the molecular "translators" that must decipher its code.

The other major requirement for protein synthesis is the translator molecules that physically "read" the mRNA codons. **Transfer RNA** (**tRNA**) is a type of RNA that ferries the appropriate corresponding amino acids to the ribosome, and attaches each new amino acid to the last, building the polypeptide chain one-by-one. Thus tRNA transfers specific amino acids from the cytoplasm to a growing polypeptide.

The tRNA molecules must be able to recognize the codons on mRNA and match them with the correct amino acid. The tRNA is modified for this function. On one end of its structure is a binding site for a specific amino acid. On the other end is a base sequence that matches the codon specifying its particular amino acid. This sequence of three bases on the tRNA molecule is called an **anticodon**. For example, a tRNA responsible for shuttling the amino acid glycine contains a binding site for glycine on one end. On the other end it contains an anticodon that complements the glycine codon (GGA is a codon for glycine, and so the tRNAs anticodon would read CCU). Equipped with its particular cargo and matching anticodon, a tRNA molecule can read its recognized mRNA codon and bring the corresponding amino acid to the growing chain.

OPERON MODEL IN PROKARYOTES

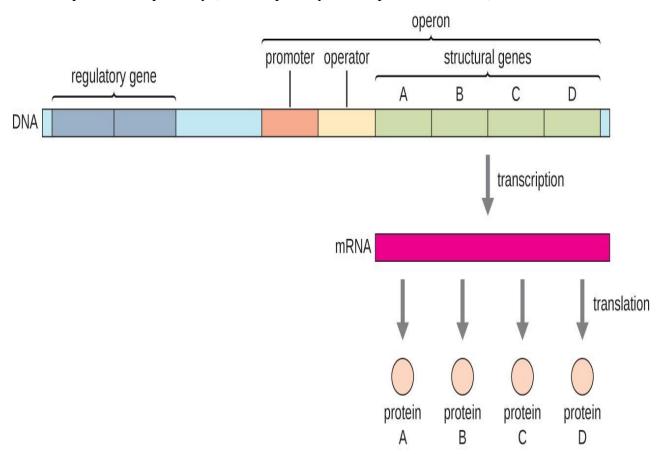
Each nucleated cell in a multicellular organism contains copies of the same DNA. Similarly, all cells in two pure bacterial cultures inoculated from the same starting colony contain the same DNA, with the exception of changes that arise from spontaneous mutations. If each cell in a multicellular organism has the same DNA, then how is it that cells in different parts of the organism's body exhibit different characteristics? Similarly, how is it that the same bacterial cells within two pure cultures exposed to different environmental conditions can exhibit different phenotypes? In both cases, each genetically identical cell does not turn on, or express, the same set of genes. Only a subset of proteins in a cell at a given time is expressed.

Genomic DNA contains both **structural genes**, which encode products that serve as cellular structures or enzymes, and **regulatory genes**, which encode products that regulate gene expression. The expression of a gene is a highly regulated process. Whereas regulating gene expression in multicellular organisms allows for cellular differentiation, in single-celled organisms like prokaryotes, it primarily ensures that a cell's resources are not wasted making proteins that the cell does not need at that time.

Elucidating the mechanisms controlling **gene expression** is important to the understanding of human health. Malfunctions in this process in humans lead to the development of cancer and other diseases. Understanding the interaction between the gene expression of a pathogen and that of its human host is important for the understanding of a particular infectious disease. Gene regulation involves a complex web of interactions within a given

cell among signals from the cell's environment, signaling molecules within the cell, and the cell's DNA. These interactions lead to the expression of some genes and the suppression of others, depending on circumstances.

Prokaryotes and eukaryotes share some similarities in their mechanisms to regulate gene expression; however, gene expression in eukaryotes is more complicated because of the temporal and spatial separation between the processes of transcription and translation. Thus, although most regulation of gene expression occurs through transcriptional control in prokaryotes, regulation of gene expression in eukaryotes occurs at the transcriptional level and post-transcriptionally (after the primary transcript has been made).



In prokaryotes, structural genes of related function are often organized together on the genome and transcribed together under the control of a single promoter. The operon's regulatory region includes both the promoter and the operator. If a repressor binds to the operator, then the structural genes will not be transcribed. Alternatively, activators may bind to the regulatory region, enhancing transcription.

Prokaryotic Gene Regulation

In bacteria and **archaea**, structural proteins with related functions are usually encoded together within the genome in a block called an **operon** and are transcribed together under the control of a single **promoter**, resulting in the formation of a polycistronic transcript (Figure 1).

In this way, regulation of the transcription of all of the structural genes encoding the enzymes that catalyze the many steps in a single biochemical pathway can be controlled simultaneously, because they will either all be needed at the same time, or none will be needed.

For example, in *E. coli*, all of the structural genes that encode enzymes needed to use lactose as an energy source lie next to each other in the lactose (or *lac*) operon under the control of a single promoter, the *lac* promoter. French scientists François **Jacob** (1920–2013) and Jacques **Monod** at the Pasteur Institute were the first to show the organization of bacterial genes into operons, through their studies on the *lac* operon of *E. coli*.

For this work, they won the Nobel Prize in Physiology or Medicine in 1965. Although eukaryotic genes are not organized into operons, prokaryotic operons are excellent models for learning about gene regulationgenerally.

There are some gene clusters in eukaryotes that function similar to operons. Many of the principles can be applied to eukaryotic systems and contribute to our understanding of changes in gene expression in eukaryotes that can result pathological changes such as cancer.

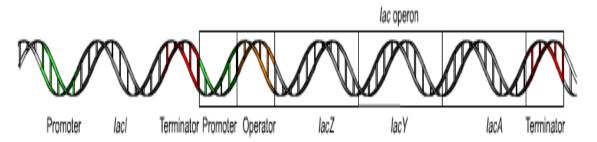
Each operon includes DNA sequences that influence its own transcription; these are located in a region called the regulatory region. The regulatory region includes the promoter and the region surrounding the promoter, to which **transcription factors**, proteins encoded by regulatory genes, can bind. Transcription factors influence the binding of **RNA polymerase** to the promoter and allow its progression to transcribe structural genes. A **repressor** is a transcription factor that suppresses transcription of a gene in response to an external stimulus by binding to a DNA sequence within the regulatory region called the **operator**, which is located between the RNA polymerase binding site of the promoter and the transcriptional start site of the first structural gene. Repressor binding physically blocks RNA polymerase from transcribing structural genes.

Conversely, an **activator** is a transcription factor that increases the transcription of a gene in response to an external stimulus by facilitating RNA polymerase binding to the promoter. An **inducer**, a third type of regulatory molecule, is a small molecule that either activates or represses transcription by interacting with a repressor or an activator.

In prokaryotes, there are examples of operons whose gene products are required rather consistently and whose expression, therefore, is unregulated. Such operons are **constitutively expressed**, meaning they are transcribed and translated continuously to provide the cell with constant intermediate levels of the protein products. Such genes encode enzymes involved in housekeeping functions required for cellular maintenance, including DNA replication, repair, and expression, as well as enzymes involved in core metabolism. In contrast, there are other prokaryotic operons that are expressed only when needed and are regulated by repressors, activators, and inducers.

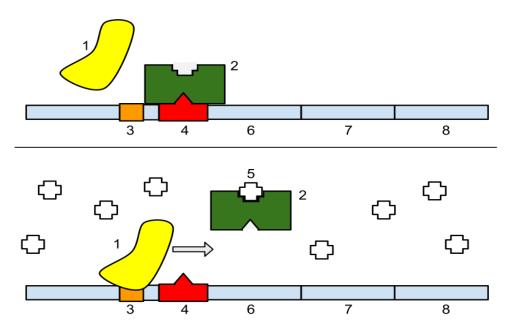
LAC OPERON MODEL

The *lactose* operon (lac operon) an operon required is for the transport and metabolism of lactose in *E.coli* and many other enteric bacteria. Although glucose is the preferred carbon source for most bacteria, the lac operon allows for the effective digestion of lactose when glucose is not available through the activity of betagalactosidase.^[1] Gene regulation of the lac operon was the first genetic regulatory mechanism to be understood clearly, so it has become a foremost example of prokaryotic gene regulation. It is often discussed in introductory molecular and cellular biology classes for this reason. This lactose metabolism system was used by François Jacob and Jacques Monod to determine how a biological cell knows which enzyme to synthesize. Their work on the lac operon won them the Nobel Prize in Physiology in 1965.



Bacterial operons are polycistronic transcripts that are able to produce multiple proteins from one mRNA transcript. In this case, when lactose is required as a sugar source for the

lac bacterium. the three genes of the operon can be expressed and and *lacA*. translated: lacZ, lacY, their subsequent proteins The of lacZ is β -galactosidase which a disaccharide, gene product cleaves lactose, into glucose and galactose. *lacY* encodes Beta-galactoside permease, a membrane protein which becomes embedded in the cytoplasmic membrane to enable the cellular transport of lactose into the cell. Finally, *lacA* encodes Galactoside acetyltransferase.



The *lac* operon. Top: Repressed, Bottom: Active. 1: RNA polymerase, 2: Repressor, 3: Promoter, 4: Operator, 5: Lactose, 6: *lacZ*, 7: *lacY*, 8: *lacA*.

It would be wasteful to produce enzymes when no lactose is available or if a preferable energy source such as glucose were available. The *lac* operon uses a two-part control mechanism to ensure that the cell expends energy producing the enzymes encoded by the *lac* operon only when necessary. ^[2] In the absence of lactose, the *lac* repressor, *lacI*, halts production of the enzymes encoded by the *lac* operon. ^[3] The lac repressor is always expressed, unless a co-inducer binds to it. In other words, it is transcribed only in the presence of small molecule co-inducer. In the presence of glucose, the catabolite activator protein (CAP), required for production of the enzymes, remains inactive, and EIIA shuts down lactose permease to prevent transport of lactose into the cell.

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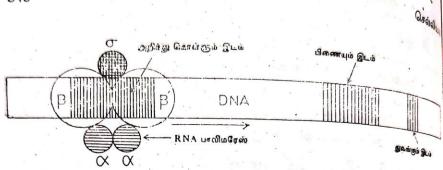
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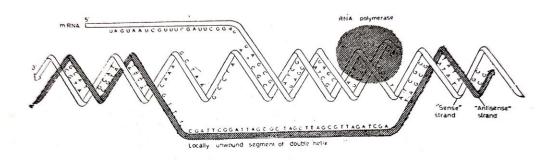
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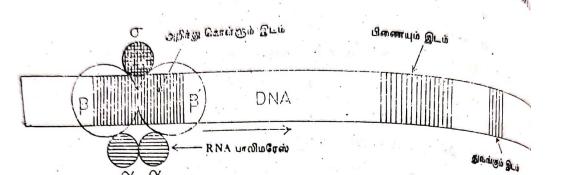
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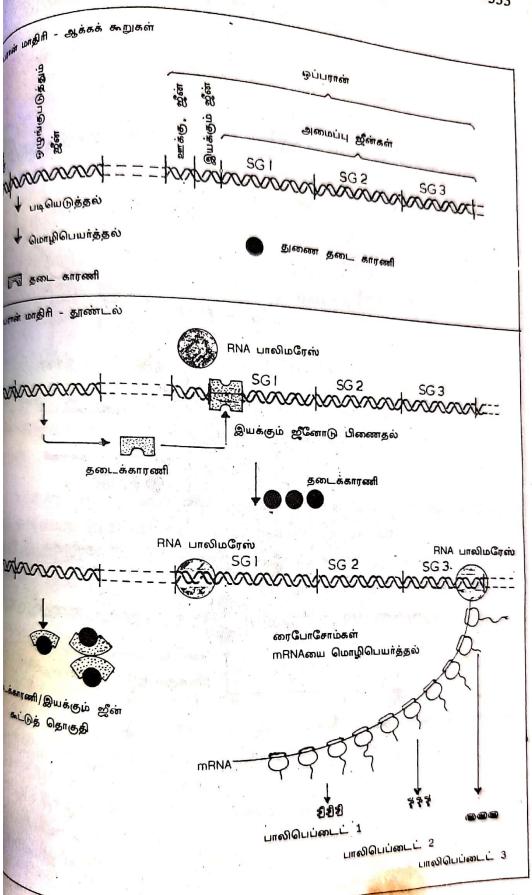
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