Genetics & molecular biology

◉ Sheet ○ Slide

Number:		1
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Molecular Biology

Introduction to Molecular:

The midterm exam will be in Molecular and the questions are from six chapters.

The final exam will be in Molecular and Genetics but the larger part goes to genetics.

The book “Lehninger” is for molecular biology and biochemistry. It is the approved book for this course. The last part of it is the molecular part

What is a gene? What is DNA? Where do we find DNA? How is it a storage? How can we have so much DNA in the cell? Where in the cell can we find it? How does Replication happen? How does a cell become two? How is the DNA built? What is the metabolism of DNA? What is RNA? How do they (DNA and RNA) look like? What are the differences between them? How is the RNA built? What is the metabolism for RNA? How do we build RNA from DNA? How do we translate DNA to RNA? The next unit discusses the methods of translating RNA to proteins.

Protein is the RECEPTOR, the hormone, the enzyme that works in the body's vital processes. A vision disorder (RECEPTOR OR HORMONE OR ENZYME) can produce diseases such as insulin imbalance and a defect in certain enzymes. tThis can further cause diseases in the digestive system, in the lungs, or affects the growth in children and adults.

When we say “we have a defect in a protein”, this means that the defect is mainly from the DNA and there is a mutation in this DNA.

For example, for a person with a disease caused by a mutation in the gene, the mutation was not translated into the proper RNA, and the RNA did not translate into protein, therefore, did not give the enzyme required, or did not give the required hormone in sufficient quantity, size and function necessary, so it resulted in that disease.

Molecular Biology

1. Introduction to Genes & Chromosomes
2. DNA Metabolism (Replication) DNA تضاعف ال
3. RNA Metabolism (transcription): building DNA from RNA
4. Protien Metabolism : Modifications ممكن يصيرله

molecule: protein or any other molecule that has a biological function.

PCR:A technique responsible for polymerizing.

للكشف عن بعض الأمراض الوراثية والطفرة الموجودة بجين معّين، مثال:بيتا التلاسيميا تحدث بسبب طفرة بجين يسمى بيتا غلوبين.

Beta globin: a globin protein that produces part of the hemoglobin of the blood.

Introduction to Genes & chromosomes

DNA-Polymerase has many types in the human body and it has a high speed (approximately 100-130 ***nucleotide/seco***nd). Because of its his high speed, we get mutations.

Mutation in DNA is more dangerous than in RNA

Polymerase in Bacteria is faster than in Human

In ***human cell there is 3-3.2 billion base pairs and 6 billion nucleotides.***

1/5 of genome in the body= Repair System

In every division of a cell , we are exposed to cancer $10^{16\%}$

Every second, approximately 125 million cells die – so every year our body looses 70kg of cells.

What is the genetic material built of?

Nucleotides are made of Nucleic Acids, each neucleotide consists of:

1) Sugar

2) Nitrogen Base

3) Phosphate group

** Nucleotide is the main component of DNA & RNA, it also exists in ATP (But in ATP, it has three phosphate groups).

Example:

If the nitrogen base is Adenine, the sugar is Ribose, and there is only one phosphate group, it is Adenosine 5'-monophosphate (AMP)

*5': Five Prime

ADP: Adenosine Di Phosphate.

ATP: Adenosine Tri Phosphate.

** If we see a picture that includes a nucleotide, how can we know if it is DNA or RNA?

1) One of the differences between DNA and RNA is the type of the sugar (Ribose \ Deoxyribose):

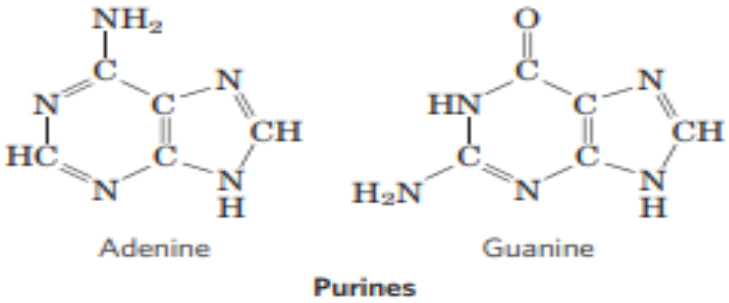
In RNA, the sugar is ribose.

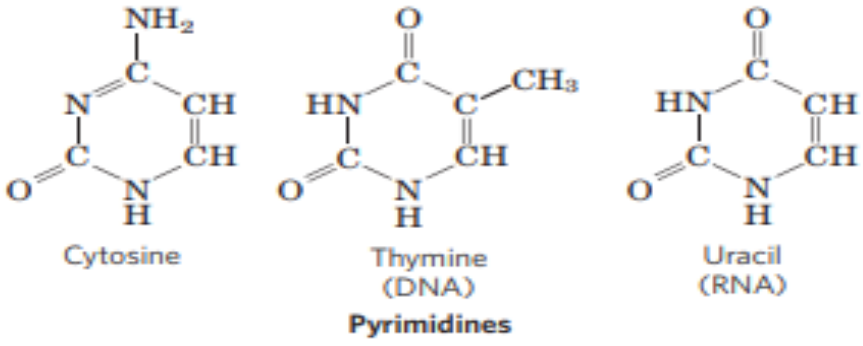
In DNA, the second carbon atom does not have oxygen. It is 2- Deoxyribose (De means without)

2) The other difference is in the nature of the existing nitrogen bases (PURINES/ PYRIMIDINES):

Purines consists of: 1) A (Adenine)

2) G (Guanine)





Pyrimidines consists of: 1) C (Cytosine)

2) T (Thymine) / U (Uracil)

PYRIMIDINES: ONE RING

PURINES: TWO RINGS

The difference between DNA and RNA, Is that RNA has Uracil (U),While in DNA, instead of Uracil (U) ,there is Thymine (T).

DNA : A , G , T ,C

RNA : A , G , U , C

3) Another difference between DNA and RNA is that the DNA is double stranded while the RNA is made of a single chain.

In summary, these are the differences between DNA and RNA:

- One or two strands.

- The nature of the nitrogen base.

- The type of sugar: DEOXYRIBOSE or RIBOSE.

**What are the links between Nucleotides?

Nucleotides bind together by a phosphate group.

The bond between a nucleotide and the other nucleotide is called a Phosphodiester bond.

**What is a phosphodiester bond?

It is a covalent bond between the Phosphate group in one nucleotide with the OH on carbon 3 of another nucleotide 3'. This called a condensation reaction because the hydrogen atom is eliminated.

The polarity is important for all DNA & RNA, so we always start from this part (5 'phosphate until the second part 3' hydroxyl) like head and tail.

5’ prime is the free part.

*The sugar and phosphate will not be different so the only difference is in the nitrogen base.

** The double stranded DNA starts from 5' to 3'

The first nitrogen base is (A)

Second (C)

Third (T)

Fourth (G)

If I want to express it: (A-C-T-G)

The second series should be: (T-G-A-C)

* The two strands are complementary to each other .

Between A and T there are two hydrogen bonds.

Between C and G there are three hydrogen bonds.

* The two chains are (Antiparallel)

If the first is from 3' to 5', the second is from 5' to 3'.

كمان شغلة في ال DNA انه ملتف على بعضه (Helix)

How does the DNA maintain its form? Why is it wrapped in a Helical shape?

1)Because there are hydrogen bonds (between A and T two hydrogen bonds, between C and G three hydrogen bonds).

2) Hydrophobic interactions: Hydro means water, Phobic of phobia means fear, fear of water.

3) Stacking: between nitrogen bases. it means they are arranged on top of each other as if they were stairs in a spiral staircase.

DNA:

- Double Helix
- Anti-parallel
- Two strains
- Complementary strands
- Hydrogen bonding
- Hydrophobic interactions

** Another characteristic of DNA:

Its room temperature is 25 ° C and its PH is 7

If we change the conditions for the DNA, such as changing the PH (adding a base) or warming it up or adding Urea (from amide), the DNA will become:

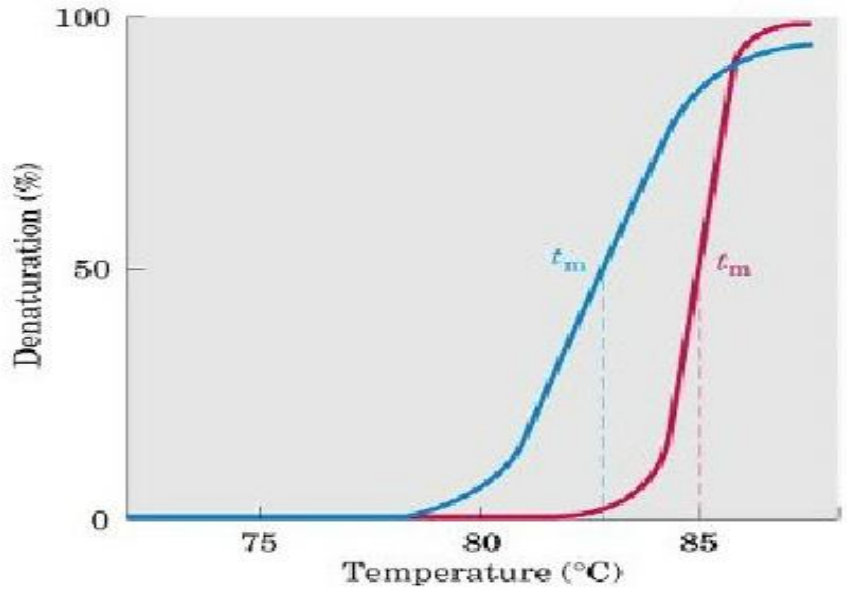
{DENATURED}: becomes (single stranded) while it was supposed to be (double stranded). This also depends on the amount of time the DNA is exposed to the conditions and the impact force (e.g. the amount of radiation)

If we reduce the heat or adjust the Ion-concentration of the salts, the DNA will undergo Renaturation, and return as it was originally(nitrogen bases bind to their complementary bases) .

To know the validity of the experiment (concentration of DNA), it is necessary to use a device called (spectrophotometer**) depending on the objects color.

- The wave at which we measure DNA is (VV 260 nm) and we also have to measure the protein level, because DNA and proteins are bound together tightly.

Analysis of DNA denaturation:



How does the double-stranded DNA behave as it is exposed to temperature?

As shown in the X axis, they started from a temperature of 75,80, 85, 90 (^{0}C)

The Y axis shows the width of the Absorption (how much light is absorbed, the rays).

- 260nm is the degree which we measure DNA.

** Double-stranded DNA has little absorption, when we raise the temperature and reach 80^{0}C, the absorption starts to rise up to 85 where it is fixed there.

This means that all the DNA was double-stranded and then it started to separate and become single-stranded.

What is melting temperature (Tm)?

It is the temperature at which which ½ the bases in a DNA sample have denatured.

(Tm) is the melting point: it depends on pH, ionic strength, and on the size and base composition of the DNA.

If its long; more heat and more time is needed to separate them.

If it’s rich in G&C, the links are more. Why?

Because G&C has three hydrogen bonds, the separating of three bonds needs more heat than the separating of 2 hydrogen bonds.

DNA denaturation and renaturation: It is the basis of many experiments in molecular biology such as (PCR).

Summary

- What is nucleotide, nucleic acid?
- DNA & RNA differences
- Hybridization
- Phosphodiester bond
- Orientation
- Properties of DNA as Complementary & antipallarar
- Nature of bonds (Hydrophobic, Hydrogen bonds {3 بين C&G, 2 بين A&T})

DNA has a double helix structure because the backbone of DNA consists of phosphoribose sugar but the ramps are nitrogenous bases.

Hydrogen bonds between nitrogenous bases

What does it mean when we say that the length of DNA is 10 pairs nitrogenous base?

It means that DNA is double stranded and it consists of 10 pairs of nitrogenous bases that are opposite to each other.

Backbone structure of DNA consists of two regions:

Major grove, where DNA strands are far away from each other

Minor grove: where DNA strands are close to each other

The attachment of these regions with other proteins helps in multiple processes; however, proteins bind with major grove regions more than minor grove regions. These regions form due to the twisting of DNA strands.

If DNA strands were parallel, we will never die.

As enzymes do not replicate their chromosome`s ends, the time of cell death is spending.

Both of the strands in the DNA double helix can grow from a 5' to 3' direction, but they grow in opposite directions due to opposite orientation of the sugar molecule in them.

RNA Types:

mRNA -5%

rRNA-80%

tRNA-15%

micro RNA- recently discovered ,it has high efficiency,and can manage translation of circular RNA

Tens of thousands are(non-coding mRNA) which do not translate to proteins, and are named (lnc RNA)=long non-coding RNA

Note that if transcription happened, this doesn't mean that there will always be translation afterwards. It is not true that every mRNA-will be translated!

DNA :

As other macromolecules, DNA consist of polymers that consist of monomers.

1. Abbreviation for Deoxyribonucleic Acid, (sugar without oxygen on carbon 2), advanced molecule (complex)

2. Every single cell has the same content of DNA (Genetic content)

3.DNA has all the information responsible for giving the shape and function to the living organisms.

4. Unit of heredity
5.Double Helix
6.Antiparallel
7.Two strains
8.Complementary to each other

9.DNA has a room temperature of 25˚ C , PH of 7 , and it is vicous

For the DNA, there is a Replication and Repair System. Replication happens with DNA Polymerase. The Repair System is for modification and decreasing mutation, and if it doesn't work then cancer will occur. Cancer means uncontrolled.

There is type of polymerase which is active through all pregnancy, once the baby is born, it becomes inactive. It can remain active in one case which is cancer.

The DNA in Mitochondria and Bacteria is almost similar.

The amount of DNA in Mitochondria is small and doesn`t make chromatin like nucleus.

DNA in Mitochondria refers to the DNA in bacteria, so we conclude that there are some types of cancer caused by a damage in the Mitochondria(uncontrolled).

DNA in mitochondria is not packed with histones, unlike that in the nucleus.

2% of our genome is for protein synthesis.

To add a new attribute to the cell, we combine the new attribute genome with a plasmid, and then we insert it in the cell.

DNA has three main structures:

1)Primary structure: sequence of nucleotides

2)Secondary structure: Double helix that consists of two twisted strands around an unreal x-axis

3)Tertiary structure: completely folded (عشان لما يدخل النواة يكون صغير حجمه)

Ribose carbons' bonding in DNA

1. First carbon is bonded to nitrogenous base

2. Second carbon may be bonded to hydrogen (DNA) or hydroxide group (RNA)

3. Fifth carbon is bonded to the phosphate group

NOTE: 5 prime end --> free phosphate end (not attached to another nucleotide)

3 prime end --> free OH end (not attached to another nucleotide)

There are different forms of DNA which are: A, B and Z مدى قرب النيوكليوتيد او بعدهم عن بعض هو الذي يحدد الشكل.

B form is the native one.

There are two processes called renaturation and denaturation of DNA.

- PCR is quite similar to DNA replication but PCR occurs inside tubes while replication inside the cells, bothare amplification.

Genes and Chromosomes

هون راح نحكي عن امثلة

(Bacteriophage T2): A virus that infects bacteria, putting it in distilled water for osmosis.(lyses ---- So the virus explodes)

Viruses are Infectious parasites, including a genetic material and coat protein that live at the expense of the Host.

Scientists have tried more than one type of virus (4, T4, X174, ...) and made the same experiment, allowing it to become two-dimensional lyses showing the dimensions of DNA (as in table 24-1).

The size of the genetic material in Each virus was varied from one virus to another (Number of base pairs in viral DNA) and each virus was the machine (length of viral DNA in nm).

The virus has a dimension of 25 with 1900 genetic material, and the virus to its dimensions 78 where DNA 1400

This is about viruses, what about other organisms such as bacteria??

E-Choli Bacteria: has 200 times more DNA than Bacteriophage lambda. When they put them under the same conditions to make the lyses, the DNA was much larger than the length of the bacteria, so this was not special in viruses.

CONCLUSION: These organisms contain more DNA in their dimensions than if they were elongated

**Other organisms:

-Yeast cell, the simplest type of eukaryote, 2.6 times more DNA than E. coli.

-The fruit fly is 35 times larger than E. coli.

-Humans have 700 times more DNA than E.coli.

The DNA of (EUKARYOTES) is arranged on chromosomes, while in bacteria there is only one chromosome

While:

The fruit fly has 8

frog 26

Hydra 30

Rabbit 44

Chicken 78

Human 46

In humans there are 46 chromosomes in the Somatic cells. In the sperm & egg, there is half of them (haploid)

* If the DNA is elongated, the 46 chromosome will be 2 meters long in the unit cell, which means each cell has two meters of DNA, although DNA takes a small space in the nucleus.

*** No introns and untranslated regions in prokaryotes

سؤال: كيف نستخرج الDNA من جسم الإنسان؟

الجواب: للكشف عن مرض وراثي، يجب أن نستخرج الDNA ونفصله عن طريق عملية تسمى PCR .أسهل طريقة لعمل ذلك هو:

١) (سحب 2 مل من الدم والذي يحتوي على RBCs) والتي لا تفيدنا في هذه الحالة لعدم احتوائها على نواة) WBCs و platelets.

2) نقوم بوضع buffer lysis أو نقوم بعملية centrifugation في جهاز الطرد المركزي لفصل الدم إلى طبقتين؛ الطبقة السفلى من الRBCs والطبقة العليا من WBCs ويكون لونها أبيض مائل للصفار

٣) نضع طبقة الWBCs في tube test عن طريق pipette ثم تبدأ عملية الextraction لل DNA

٤) بعد الحصول على الDNA يكون على شكل Double helix ،يجب فتحه حتى أعمل له تضاعف replication

سؤال: ماذا أستخدم لفتح ال strands لل DNA؟ الجواب: الحرارة، تستخدم الحرارة في عملية تسمى denaturation لفتح سلاسل ال DNA ومضاعفته لكشف المرض الوراثي.

إن درجة الحرارة العالية تكسر الروابط الهيدروجينية وهذا ما يسمى ب TM أو melting temperature . ويتم ارتباطهما مجددا بداخل tube test بخفض درجة الحرارة ولا يمكن حدوث ذلك داخل الخلية أثناء الreplication ولا يحدث synthesis لسلسة واحدة فقط، لأننا بالنهاية نحتاج لstrings two لأنه ينتجcells two daughter.

Genetic code: The DNA that is meant to be translated into mRNA. mRNA carries information to the ribosome until protein or polypeptide is built.

The genetic code means that three of the nucleotides are codon, The code is translated into an amino acid:

For example, CGU translates to Arginine (amino acid)

GGA is translated into Glycine

UAC is translated into Tyrosine

ACU is translated into threonine..... and so on

So every word in the genetic language is a triplet (triplet of nucleotide) which means an amino acid

Amino acid + Amino acid = polypeptide.