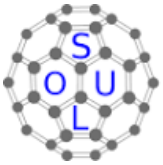
FOSSEE SUMMER FELLOWSHIP REPORT 2022



Science Open Source Software
for Teaching Learning

"3D Modeling Studies Involving Biochemistry of Nucleotides and Nucleic acids "

3D MODELING OF CHEMICAL STRUCTURES USING JMOL APPLICATION

SUBMITTED BY-

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Acknowledgement

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I am very grateful that I got to attend the weekly discussions , which were very informative and always taught me something new about the jmol application. I would also like to thank all my colleagues who shared their valuable ideas and helped me out throughout the fellowship. I've gained a lot of knowledge in the past few weeks of working with fossee , it has enhanced my skills and problem solving abilities.

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INTRODUCTION

Nucleic acids are the most important macromolecule within the organism. they're polynucleotides; that is a long chain of molecules composed of a series of nearly identical building blocks called nucleotides. There are 2 main classes of nucleic acids- 1)DNA and 2)RNA. Nucleic acids, Deoxyribonucleic acid (DNA), Ribonucleic acid (RNA) carry inheritable information which is read in cells to make the rna and proteins by which living beings work. The well known structure of the DNA; double helix,allows this information to be copied and passed on to coming generations. In this project the structures and important biochemistry related to nucleic acids, nucleotides dna and rna are summarised and shown in 3d with the help of jmol application.

Jmol is very beneficial in portraying chemical reactions related to chemistry and biochemistry. Above all it's an open source software that can be accessed by anyone free of cost which is very useful for students and teachers .It allows viewers to visualize chemical structures as 3D models. Many academicians use Jmol as an educational tool to help students visualize molecules. The 3D models are used in many websites and blogs for conceptual understanding. Jmol has features to manipulate 3D models and to display models in different styles or combinations of any or all possible styles like ball and stick,space filling, ribbon, cartoon, sticks etc.

For example; In DNA structure representation, we can show the backbone with a cartoon and the bases with ball and stick to differentiate between different bases.

This helps in gaining a better understanding of protein and nucleic acid structure. We can also get a better view of the molecule by moving it, rotating, spinning and zooming it in or out.

For my project, I have used jmol to explore nucleic acid structure, to plan experiments and to interpret experimental findings. Jmol requires typed instructions called script commands (using console) in addition to the rest of the menus. Jmol has a very simple and less complicated command line option that can be learnt by anyone easily. It also has interactive pages to simplify the use of various commands which makes it much easier to understand and learn. https://chemapps.stolaf.edu/jmol/docs/

CHAPTER 1

GENERAL DESCRIPTION OF RNA AND DNA

INTRODUCTION:

Deoxyribonucleic acid is a polymer made up of two polynucleotide chains that coil around each other to create a double helix and carry genetic instructions for all known organisms and viruses' formation, function, growth, and reproduction whereas RNA delivers genetic information that ribosomes convert into numerous proteins required for cellular functions. The three main forms of RNA involved in protein synthesis are mRNA, rRNA, and tRNA. RNA is also the genetic material that viruses use to reproduce.

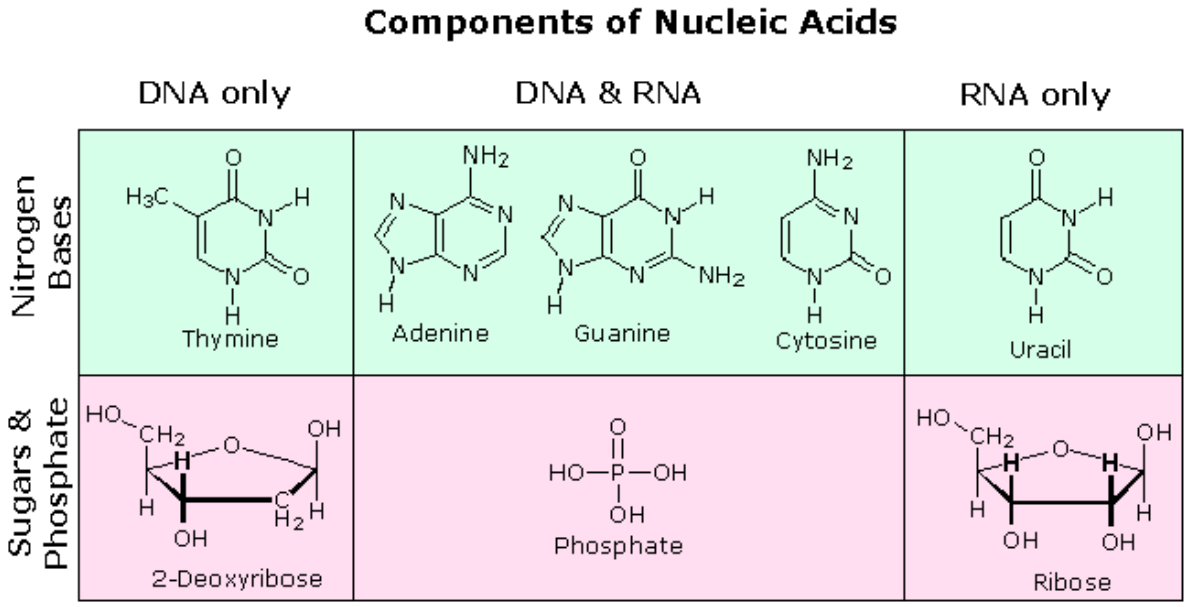


FIG 1.1-BASIC COMMONALITY & DIFFERENCE BETWEEN RNA AND DNA (figure taken from SLIDESHARE.NET)

1.1 STRUCTURAL DIFFERENCES BETWEEN RNA AND DNA

1. **DIFFERENCE IN BASES**

   DNA has four nitrogen bases adenine, thymine, cytosine, and guanine and for RNA instead of thymine, it has uracil.

   THYMINE HAS AN EXTRA METHYL GROUP ; THEREFORE DNA HAS AN EXTRA METHYL GROUP

2. **DNA is a double-stranded molecule with a long chain of nucleotides while RNA is only single-stranded. As seen in the figure above.**
3. **RNA is shorter in length due to splicing whereas DNA has an optimum length and it doesn't shorten.**
4. **DNA exists mainly in a double helix form while RNA will take on many different shapes and sizes such as the 'hairpin formation'.**
5. **There is one single type of DNA while there are many types of RNA that have different functions such as mRNA (carries DNA message to cytoplasm), tRNA (carries amino acids to mRNA and Ribosomes), rRNA (Ribosomal RNA, workbench for protein synthesis).**
6. **The pairing of bases in DNA including A-T(Adenine-Thymine) and G-C(Guanine-Cytosine)is different to that of RNA including A-U(Adenine-Uracil) and G-C(Guanine-Cytosine).**
7. **BASES AND SUGARS- DNA is a long polymer with deoxyribose sugar moiety and a phosphate backbone and RNA is a polymer with a ribose sugar moiety and phosphate backbone.**
8. **Deoxyribose sugar in DNA is less reactive because of C-H bonds on the second carbon (C_2) and DNA is stable in alkaline conditions. RNA, ribose sugar is more reactive because of the presence of hydroxyl group on C_2. RNA is not stable in alkaline conditions because bases can easily deprotonate the Hydrogen from the -OH on C_2. After deprotonation, the negatively charged oxygen may attack the Phosphate at the PO4, kicking off the Oxygen connected to the 5'C of the next nucleotide over, resulting in hydrogenation.**

1.2 SOME MORE INFO ON DNA AND RNA

1. **Attached to the 5′ carbon in both deoxyribose and ribose is a phosphate group (at "1"). The phosphate group is what makes these molecules acidic (the "A" in both DNA and RNA stands for "acid"). The negative charges on the oxygens are what's left after protons have been donated to the surrounding solution.**
2. **Attached to the 1′ carbon is one of four nitrogenous bases. They're *bases* because they have an amino functional group attached to them (at "4"), and that amino group absorbs protons from a solution, raising the pH. They're *nitrogenous* because they have a lot of nitrogen atoms. Some of these nitrogenous bases have two nitrogen rings**
3. **These polymers, at their most basic level, consist of strands of nucleotides connected in a linear sequence to one another by covalent bonds.**

4. **The 5′ carbon of one nucleotide connects to that nucleotide's phosphate group, and then that phosphate group connects to the 3′ carbon on the sugar in the *next* nucleotide. The bond connecting one nucleotide to the next is called a sugar-phosphate bond. In a DNA or RNA polymer, the connected sugars and phosphates make up a sugar-phosphate backbone.**
5. **The result of this complementary base pairing is that DNA is typically double stranded. Because the nucleotides in the two strands can form hydrogen bonds with one another only when they're oriented upside down relative to one another, the overall arrangement of the molecule is said to be antiparallel. The bond angles between the sugar and phosphates twist DNA into its famous double helix formation.**
6. **RNA is able to serve as an action molecule because it can fold into complex three-dimensional shapes. This folding emerges from hydrogen bonding between nucleotides in the same molecule of RNA.**
7. **In the Watson-Crick model, the bases are in the interior of the helix aligned at a nearly 90 degree angle relative to the axis of the helix. Purine bases form hydrogen bonds with pyrimidines, in the crucial phenomenon of base pairing**
8. **On its exterior surface, the double helix of DNA contains two deep grooves between the ribose-phosphate chains. These two grooves are of unequal size and termed the major and minor grooves. The difference in their size is due to the asymmetry of the deoxyribose rings and the structurally distinct nature of the upper surface of a base-pair relative to the bottom surface.**

Steps of model built using Jmol:

1.VISUALIZING DNA USING Jmol

PDB CODE- 1ZEW (A BDNA CONFIGURATION)
SITE FOR ACCESSING THE PDB- RCSB PDB (RCSB.org)

Steps-

> open Jmol using Jmol.jar

>go to file -> get PDB 1ZEW -> Hide water molecules and sodium atom

Using Console (click on File -> Console script) -> $Hide waters

Using model kit menu - SELECT delete atom -> Delete Na atom by clicking on it -> then EXIT the modelkit menu

>The structure appears , to show the backbone (deoxyribose-phosphate linkage)

Go to the Jmol logo at the bottom and tap on it or you could right click on your mouse

to get a panel which has various options to style , color, calculate bonds, show surfaces etc. your molecule(or 1zew)

- **Go to the Jmol logo -> choose select -> Nucleic -> SELECT BACKBONE**

Now that our backbone has been selected we can style it in different atomic models like cpk spacefill, ball & stick , wireframe etc. For that

- **Again tap on the Jmol Logo -> choose 'STYLE' -> 'SCHEME' -> Select any of those atomic models to display your DNA Backbone**

Similarly to stand out your Nucleotide Bases you could repeat the steps -

- **Go to Jmol Logo -> choose select -> Nucleic -> SELECT BASES**
- **Then -> Go to' STYLE' -> 'SCHEME' -> SELECT some other atomic model than what you chose for your Backbone to see them distinctly.**
- **IN THE FOLLOWING FIGURE BACKBONE IS IN CARTOON WHEREAS THE NUCLEOTIDE BASES ARE IN CPK SPACEFILL MODEL.**

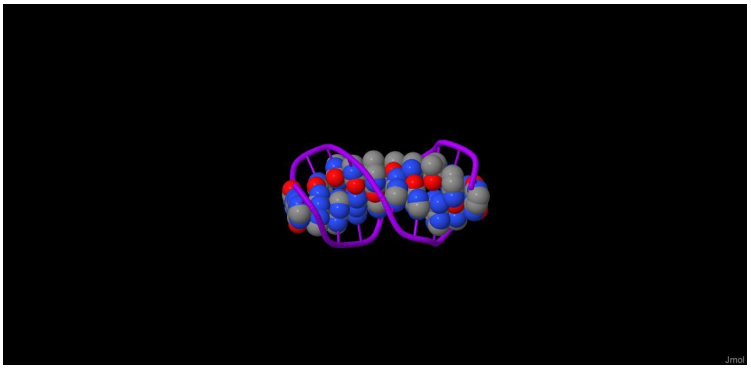


FIG 1.2 -PURPLE COLOUR DEPICTS SUGAR-PHOSPHATE BACKBONE IN CARTOON & BASES IN SPACEFILL STYLE

YOU CAN USE THE PANEL TO CHANGE THE COLOR OF THE BACKBONE AND THE NUCLEOTIDE BASES AS WELL .

- **Go to the Jmol logo -> choose select -> Nucleic -> SELECT BACKBONE OR BASES OR YOU CAN ALSO CHOOSE PARTICULARLY A-T PAIRS OR G-C PAIRS**
- **Tap on Jmol Logo again -> select 'COLOR'-> there are quite a few options you can keep it according to rasmol colors , or choose a color of your choice.**
- **We can also use the colour command on the console script to color the selected part of the molecule or the whole molecule.**

We can also show H-Bonds in a DNA Model using the Jmol panel

- **Tap on the Jmol Logo or Right click -> choose 'STYLE' ->HYDROGEN BONDS -> CALCULATE**

If you want measurements of the following H-bonds

- **Tap on the Jmol Logo or Right click -> choose 'measurements'-> click for Distance measurements**

and then according to the partial lines seen you can click on the atoms attached to the lines and get the measurements.

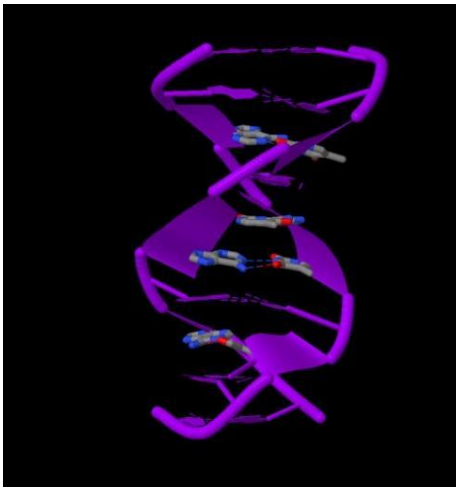


FIG1.3:In the following image G-C pairs of the DNA are in Cartoon structure whereas the A-T pairs are in Ball & stick model(G-C AND A-T CAN BE DISTINGUISHED BY LOOKING AT THE H-BONDS).

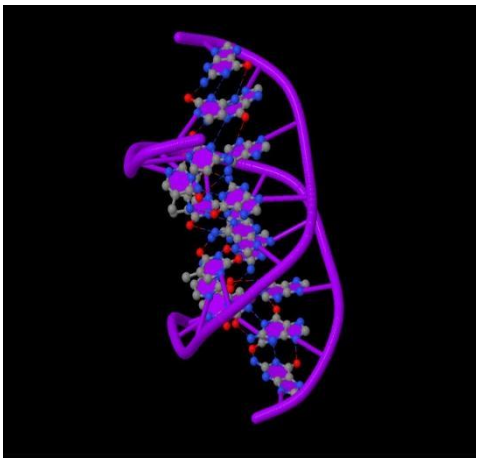


FIG1.4: H-Bonds SHOWN

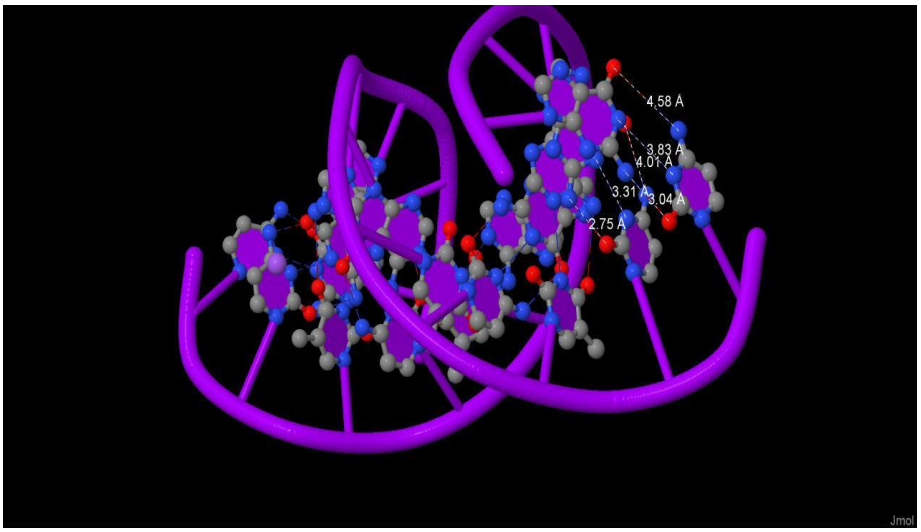


FIG1.5: Measurements of the H-Bonds SHOWN

2.VISUALIZING dsRNA USING JMOL

Double stranded RNA is similar to DNA as it has complementary base pairing and almost similar structure apart from the fact ; In dsRNA Ribose sugar is used in the backbone whereas in DNA deoxyribose sugar is present in the backbone.

Also in dsRNA instead of thymine , uracil bonds with adenine.

The similar steps can be followed with the dsRNA visualization as used in DNA visualization , FOR dsRNA we use the PDB ID- 1RNA

SITE FOR ACCESSING THE PDB- RCSB PDB(RCSB.org)

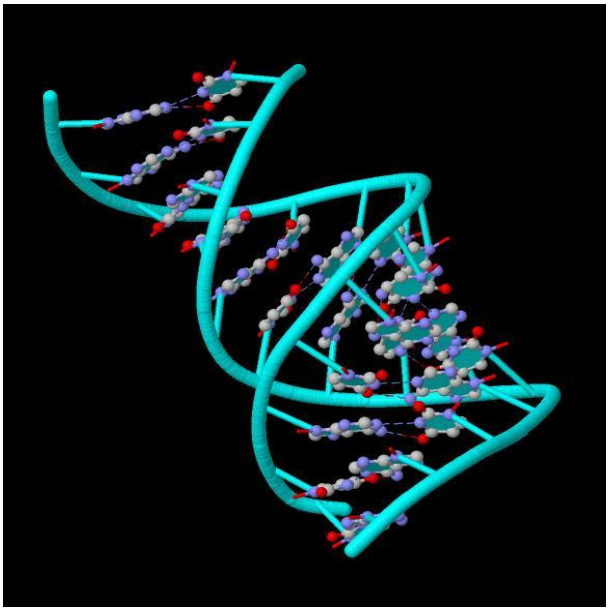


FIG1.6: dsRNA backbone represented in cartoon and bases in ball & stick atomic model

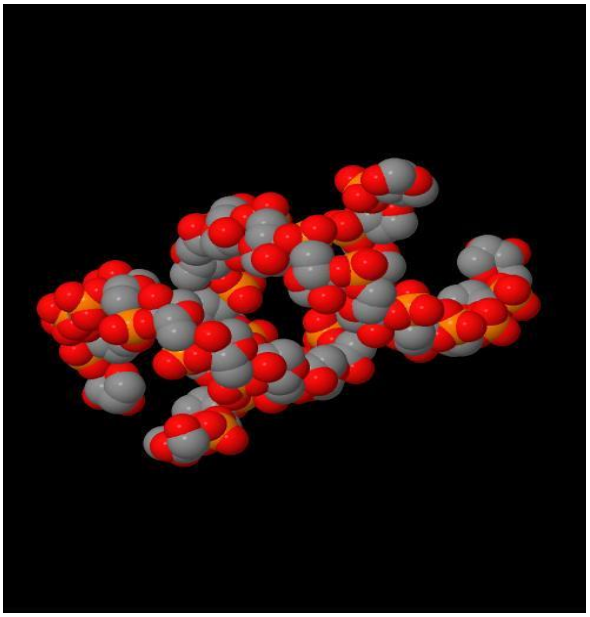


FIG1.7: CPK spacefill model of ribose sugar phosphate backbone of dsRNA

3. VISUALIZING RNA / SINGLE STRANDED RNA USING Jmol

For visualizing RNA we're using the same PDB- 1RNA

Can be accessed using- RCSB PDB

To Get ssRNA from dsRNA we need to hide its chain A or chain B(either of the 2 chains)

Open PDB through

FILE-> GET PDB -> 1RNA

FILE -> OPEN' CONSOLE SCRIPT' ->

$hide :A ; hide waters

NOW YOU HAVE YOUR RNA, YOU CAN STYLE IT, COLO IT, AND SHOW H-BONDS AND LIST THEIR MEASUREMENTS LIKE WE DID FOR DNA.

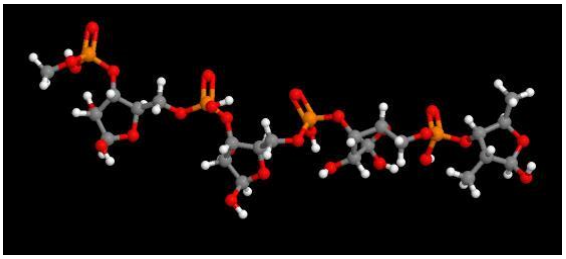


FIG1.8: BALL & STICK MODEL OF RNA BACKBONE SHOWN

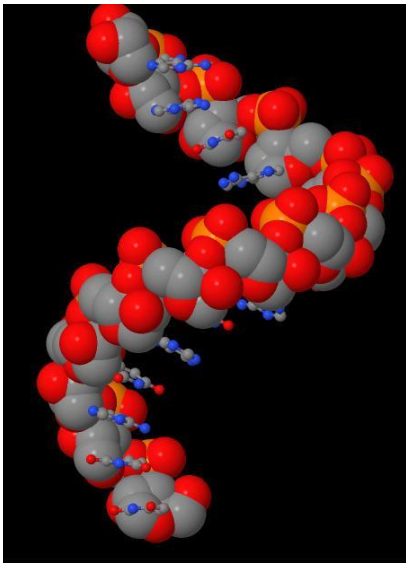


FIG1.9: CPK SPACEFILL BACKBONE AND BALL & STICK BASES OF RNA SHOWN

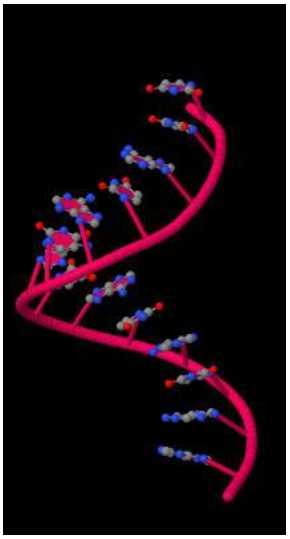


FIG1.10: CARTOON BACKBONE AND BALL & STICK BASES SHOWN

CHAPTER 2

ALL ABOUT ATP AND ADP

INTRODUCTION TO ATP & ADP

One or two extra phosphates may be covalently connected at the 5′ hydroxyl of a ribonucleotide's phosphate group connected. Nucleosides monophosphates, diphosphates, and triphosphates are the compounds that arise from this process. The chemical energy required to drive many biological reactions comes from the hydrolysis of nucleoside triphosphates. The most commonly utilised nucleoside triphosphate for this function is adenosine 5 -triphosphate, or ATP, but UTP, GTP, and CTP are also used in some operations. They serve as the activated precursors of DNA and RNA synthesis.

Abbreviations of ribonucleoside 5′-phosphates			
Base	Mono-	Di-	Tri-
Adenine	AMP	ADP	ATP
Guanine	GMP	GDP	GTP
Cytosine	CMP	CDP	CTP
Uracil	UMP	UDP	UTP

Abbreviations of deoxyribonucleoside 5′-phosphates			
Base	Mono-	Di-	Tri-
Adenine	dAMP	dADP	dATP
Guanine	dGMP	dGDP	dGTP
Cytosine	dCMP	dCDP	dCTP
Thymine	dTMP	dTDP	dTTP

FIG 2.1 : Figure taken from LEHNINGER'S BIOCHEMISTRY PG NO.-1123

The structure of the triphosphate group accounts for the energy produced by the hydrolysis of ATP and other nucleoside triphosphates. An ester linkage exists between the ribose and the α phosphate (Or the 1st phosphate linked to the ribose sugar).

The α, β and β, γ linkages are phosphoanhydride . Under typical conditions, hydrolysis of the ester linkage yields around 14 kJ/mol, whereas hydrolysis of each anhydride bond yields about 30 kJ/mol.

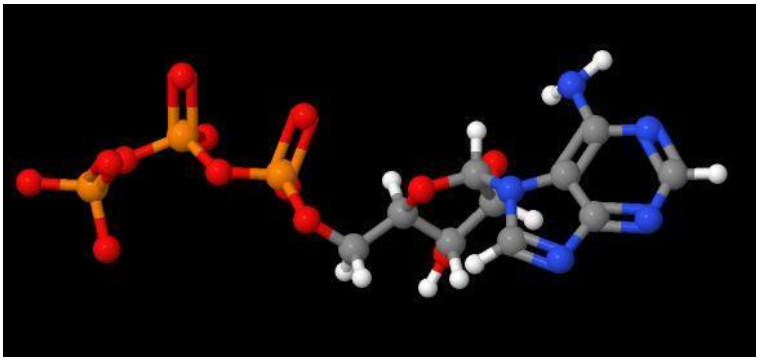


FIG2.2 : ATP MOLECULE

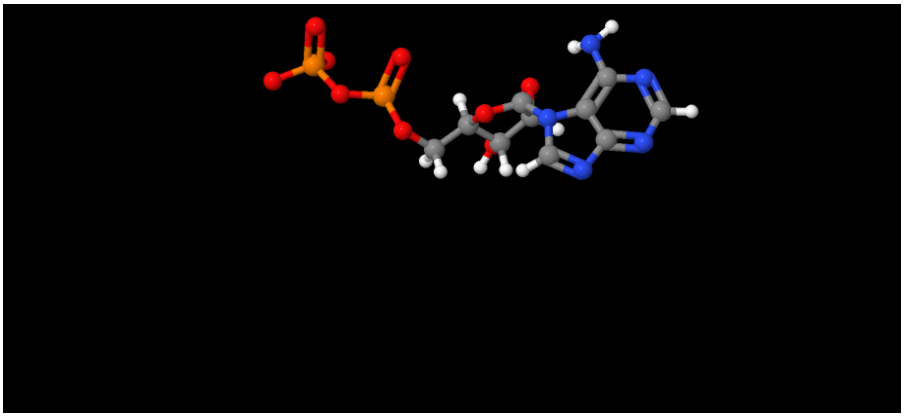
The energy gap between energy-releasing reactions (food breakdown) and energy-requiring reactions is bridged by ATP (synthesis). When a fatty acid molecule is burned, energy is released. Some of this energy is trapped in ATP molecules, while the rest is dissipated as heat. Each ATP molecule can then be transferred and utilised wherever it is needed within the cell. The triphosphate "tail" of an ATP molecule carries the energy. Covalent links connect three phosphate groups.This covalent bond is known as a pyrophosphate bond. The energy is carried by the electrons in these bonds. Energy is utilised to add one molecule of inorganic phosphate (P) to a molecule of adenosine diphosphate in the cell's power plants (mitochondria) (ADP).

ADP + P + Energy -> ATP (GIF 1)

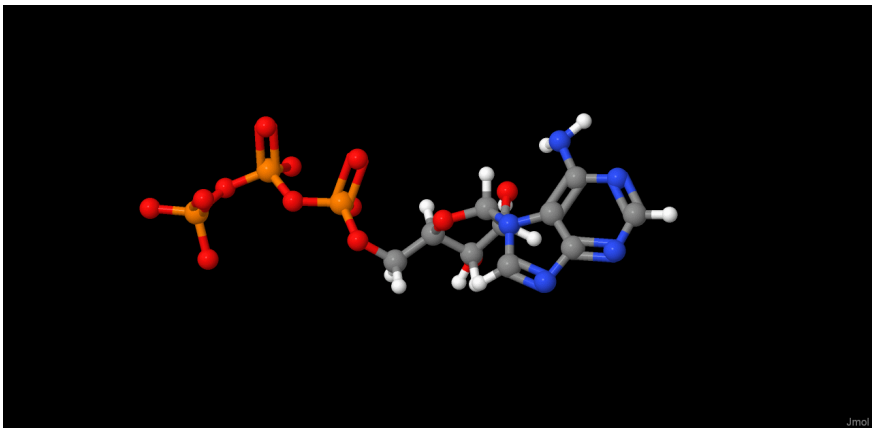
For every mole of ATP generated, around 7,300 calories of energy are stored. The last phosphate group in the tail is broken off at the energy-demanding site, releasing the energy trapped in the bond.

ATP -> ADP + P + Energy (GIF 2)

Around 7,300 calories of energy are released per mole once more. After then, the ADP and phosphate are free to return to the power plant and rejoin the system. ATP and ADP are regularly regenerated in this way.



https://drive.google.com/file/d/1dslH5bGNFo1RHeX5MbuZKr5EiwjorE9I/view?usp=sharing



https://drive.google.com/file/d/1rdzcdJJIhyAIcoLX3XuvHoBuRnqB2RQa/view?usp=sharing

The ATP-ADP Cycle is a cycle in which ATP and ADP are interconvertible. The creation of ATP requires energy, which is released when the ATP is transformed back to ADP and phosphate.

ADP-

ATP has a greater energy level than ADP, which means it obtains energy from ADP to create ATP, but it also means energy is released when ATP is converted to ADP. It is required for glycolysis. It plays an important role in the clotting of blood. It also serves as an essential component in the production of ATP

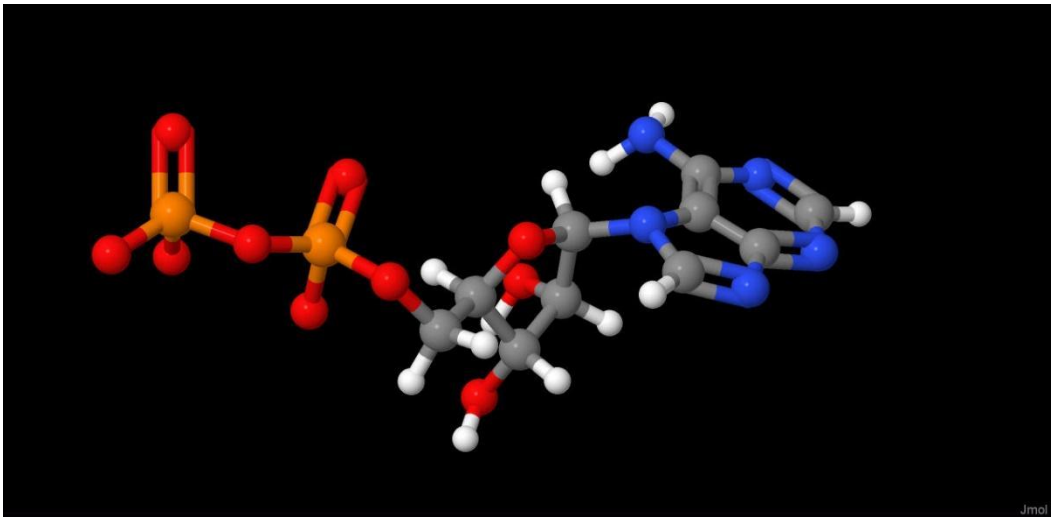


FIG2.3: ADP MOLECULE

STEPS IN JMOL-

TO MAKE ATP AND ADP I JUST LOADED ADENINE MOLECULE AND FURTHER MADE UP THE RIBOSE SUGAR AND PHOSPHATE MOLECULES BY JOINING THEM ONE BY ONE WITH THE HELP OF THE MODEL KIT MENU.

FIXED THEIR HYDROGENS AND MINIMIZED THEIR STRUCTURES.

SAVE THEIR .MOL FILES FOR FURTHER USE BY-

GO TO THE MODELKIT MENU -> SAVE FILE -> NAME THE FILE (ATP/ADP) , FIX THE FILE TYPE TO .mol-> SAVE

WE CAN STYLE AND COLOUR ADP AND ATP MOLECULES USING THE FOLLOWING STEPS :-

FILE -> OPEN -> OUR SAVED . mol FILE OF ATP/ ADP

- **TAP ON BOTTOM RIGHT JMOL LOGO -> SELECT ->SELECT ALL ->THEN GO TO 'STYLE'-> SCHEME-> ANY ATOMIC MODEL**
- **TAP ON BOTTOM RIGHT JMOL LOGO -> SELECT ->SELECT ALL->THEN GO TO 'COLOR'**

NOW TO ANIMATE ATP TO ADP WE NEED TO USE CONSOLE SCRIPT
GO TO FILE -> OPEN -> ATP.mol -> GO TO FILE -> CONSOLE SCRIPT ->
$· move 0 0 0 30 0 20 0 0 5;draw arr1 arrow diameter 0.1 (phosphorus) (atomno = 39 or atomno =37);delay 2;hide atomno >= 39 and atomno <= 43;Draw off; move 0 0 0 0 0 0 0 0 5;spin on;spin 40
CAPTURE IT BY THE CLICKING ON THE 'JMOL LOGO' -> FILE -> CAPTURE -> START CAPTURING-> SAVE IN .gif FORMAT -> ENTER THE CONSOLE COMMAND -> END CAPTURING
TO ANIMATE ADP TO ATP
IN THE SAME ATP.mol FILE
USING CONSOLE SCRIPT -> $ hide atomno >= 39 and atomno <= 43

$move 0 0 0 30 0 20 0 0 5;draw arr1 arrow diameter 0.1 (phosphorus) (atomno = 39 or atomno =37);delay 2;Display all;draw off; move 0 0 0 0 0 0 0 0 5;spin on;spin 40

CAPTURE IT BY THE CLICKING ON THE 'JMOL LOGO' -> FILE -> CAPTURE -> START CAPTURING-> SAVE IN .gif FORMAT -> ENTER THE CONSOLE COMMAND -> END CAPTURING

BY USING ATP CORR LABELLED.mol FILE(UPLOADED ON GDRIVE)

COMMANDS FOR -

atp->adp

$ Capture"ATP->ADP"SCRIPT"move 0 0 0 30 0 20 0 0 5;draw arr1 arrow diameter 0.1 (phosphorus) (atomno =38 or atomno =40);delay 2;hide atomno >= 40 and atomno <= 43;Draw off; move 0 0 0 0 0 0 0 0 5;spin on;spin 40"

$Capture end

adp->atp

$hide atomno >= 40 and atomno <= 43

$ Capture"ADP->ATP"SCRIPT"move 0 0 0 30 0 20 0 0 5;draw arr1 arrow diameter 0.1 (phosphorus) (atomno = 36 or atomno =40);delay 2;Display all;draw off; move 0 0 0 0 0 0 0 0 5;spin on;spin 40"

$Capture end

ATP<->ADP(LOOP)

Capture"ADP<->ATP"SCRIPT"delay 2;draw arr1 arrow diameter 0.1 (phosphorus) (atomno =38 or atomno =38);delay 1;hide atomno >= 40 and atomno <= 43;Draw off; move 0 0 0 0 0 0 0 0 5;spin on;delay 10;spin off;draw arr1 arrow diameter 0.1 (phosphorus) (atomno = 38 or atomno =38);delay 1;Display all;draw off; move 0 0 0 0 0 0 0 0 5;spin on;delay 12;spin off;loop 2"

$Capture end

AMP->ADP->ATP

$ hide atomno>=36 and atomno<=43

$ Capture"amp->adp->atp"SCRIPT"move 0 0 0 30 0 20 0 0 5;draw arr1 arrow diameter 0.1 (phosphorus) (atomno = 34 or atomno =34);delay 1;Display atomno>=1 and atomno<=39 ;draw off; move 0 0 0 0 0 0 0 0 5;spin on;delay 10;spin off;move 0 0 0 0 0 0 0 0 5;draw arr1

arrow diameter 0.1 (phosphorus) (atomno = 38 or atomno =38);delay 1;Display atomno>=1 and atomno<=43 ;draw off; move 0 0 0 0 0 0 0 0 5;spin on;delay 12;spin off"

$Capture end

CHAPTER 3

BIOCHEMISTRY OF NUCLEOTIDES

Introduction

Nucleic acid bases exhibit keto-enol and amino-imino prototropic tautomerism due to the presence of multiple solvent-exchangeable protons.The formation of minor tautomers can increase the overall structural and chemical diversity of nucleic acids, which enables their diverse functions in biology, The formation of minor tautomers in nucleic acids is a rare event, and their dynamics of interconversion is fast, on a millisecond to nanosecond time scale .During DNA replication, formation of minor tautomeric forms of nucleobases could lead to mispairing that generates mutations in the absence of any DNA lesion or other form of DNA damage,This phenomenon is thought to explain the appearance of spontaneous mutations during the replication of undamaged DNA. The transient generation of minor tautomers in DNA allows stabilization of mismatches, in the polymerase active site, in conformations that are indistinguishable from the structures of canonical base pairs. While assembling their double-helix model of DNA, James Watson and Francis Crick stumbled over the phenomenon of base tautomerism; in fact, their model required that the bases adopt specific tautomeric forms in order to base-pair, with the suggestion that alternate tautomers would lead to mispairing and spontaneous mutations.Since then, structural studies have shown that the formation of minor tautomers in DNA could allow wobble mismatches, such as the A-C and G-T base pairs, to adopt a geometry similar to that of canonical base pairs in Watson and Crick (W-C) conformations.

Nucleic acids display keto-enol and amino-imino types of tautomerism. This type of tautomerism, called prototropic tautomerism involves the movement of protons in the form of deprotonation at one site and protonation at another site, which is accompanied by a rearrangement of the double bonds in between the two sites. The base pairing complementarity in nucleic acids depends on the position of protons on the Watson-Crick (W-C) interface.Therefore, certain minor tautomeric forms of canonical nucleic acid bases can engage in alternative base pairings. Prototropic tautomerism is expected to influence the biology of both DNA and RNA.

adenine has the ability to adopt amino and imino tautomeric forms involving the exocyclic group at the 6-position; uracil and thymine have carbonyl functional groups that can participate in keto–enol tautomerism. Guanine and cytosine have both amino and carbonyl

groups, thus they can exhibit both amino–imino and keto–enol types of tautomerism . Of all possible tautomeric forms of the canonical nucleobases, the keto- and amino-forms predominate under physiological conditions, and are thus considered "major" tautomers. The imino- and enol forms are considered "minor" tautomers and are typically very rare.

- **DNA TAUTOMERIZATION**

  The genetic integrity of genomic DNA relies on adenine, guanine, cytosine and thymine existing predominantly in their keto and amino tautomeric forms during replication and transcription. In their work on the structure of DNA, Watson and Crick did appreciate that the formation of minor tautomeric forms would alter the base pairing properties of nucleic acid bases, potentially with mutagenic consequences . These minor tautomeric forms could arise from inter-helical transfer of protons in a DNA duplex . Transient formation of minor tautomeric forms of DNA bases, and their stabilization in the active site of DNA polymerases during replication, could lead to incorporation of mismatched base pairs.minor tautomeric forms of nucleic acid bases are rare and interconvert rapidly in aqueous solution.

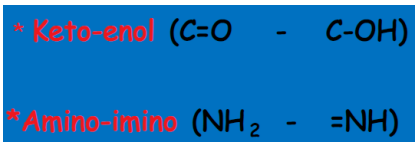


FIG 3.1: GENERAL CONVERSIONS OF KETO-ENOL &AMINO-IMINO FORMS

1. **Amino and imino forms of Adenine & Cytosine**

Standard amino and rare imino forms of C & A differ by a spontaneous proton shift (an H nucleus) between the adjacent C and N molecules. The molecular formulae do *not* change: the *amino* and *imino* forms are therefore described as tautomers of each other.

2. **Keto-enol -tautomerism in Guanine, Thymine and Uracil**

It is the most common form of tautomerism and occurs in aldehydes, ketones and related compounds (essentially carbonyl compounds with one or more hydrogens on the α-carbon). guanine, thymine, and uracil (which are cyclic amides) can exist in either lactam (keto) or lactim (enol) forms.

3.1 WHY A & C ARE IN AMINO FORM AND G & T IN KETO FORM?

It is essential that H-atoms have fixed location in DNA, if not so, A could pair with C and G could pair with T. As a consequence bases on the 2 strands will not be complementary and on replication ; the DNA cannot give 2 identical DNA strands similar to the parent molecule

The fact that the bases in DNA are not static was first pointed out by Watson and Crick. Hydrogen atoms in the bases can move from one position in a purine or pyrimidine to another position.Some of these are shown below. Such chemical fluctuations are Tautomeric shifts.The tautomeric forms of each base exist in equilibrium but the amino and lactam tautomers are more stable and therefore predominate under the conditions found inside most cells. The rings remain unsaturated and planar in each tautomer.

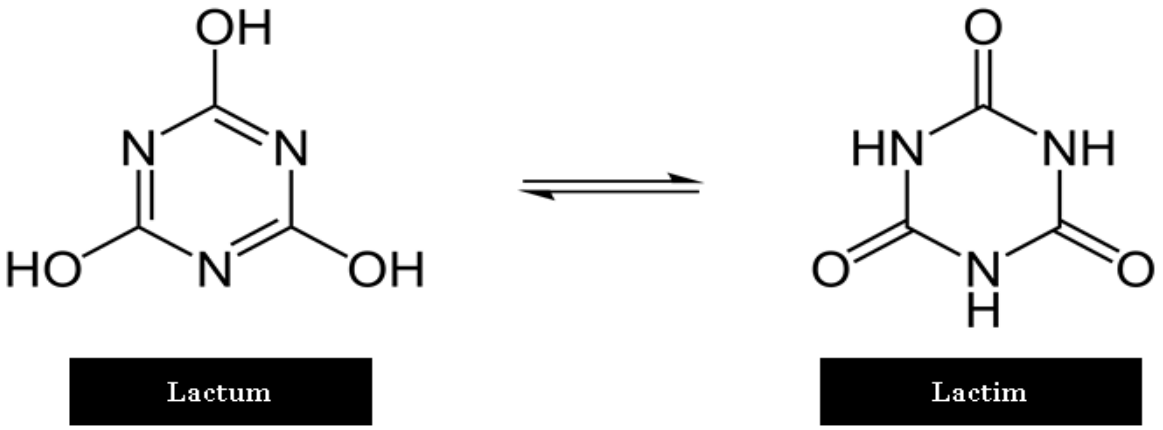


FIG 3.2: SHOWS CONVERSION OF BASES FROM LACTAM FORM TO LACTIM FORM

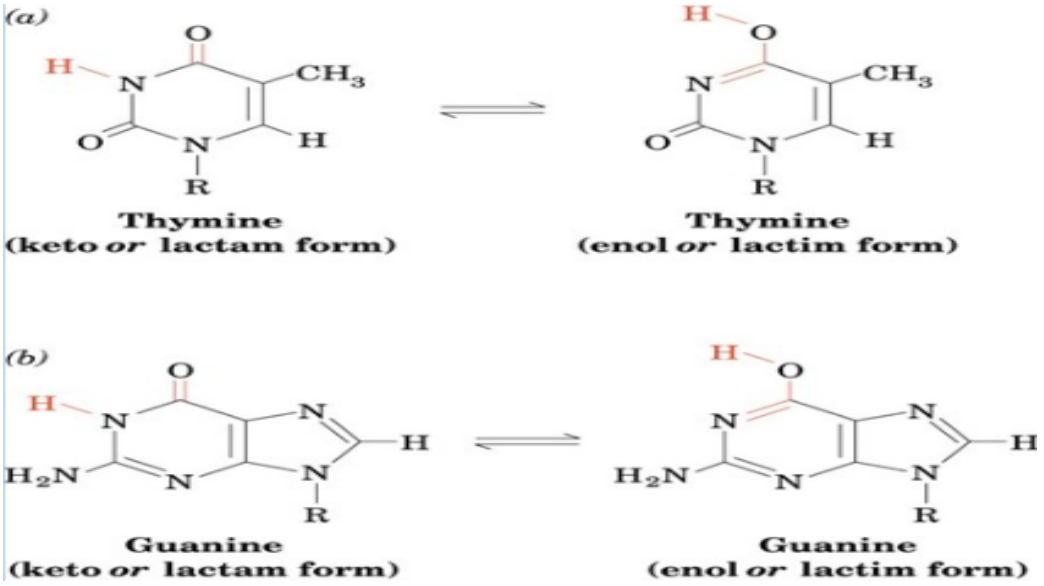


FIG 3.3: KETO -ENOL CONVERSION OF THYMINE AND GUANINE

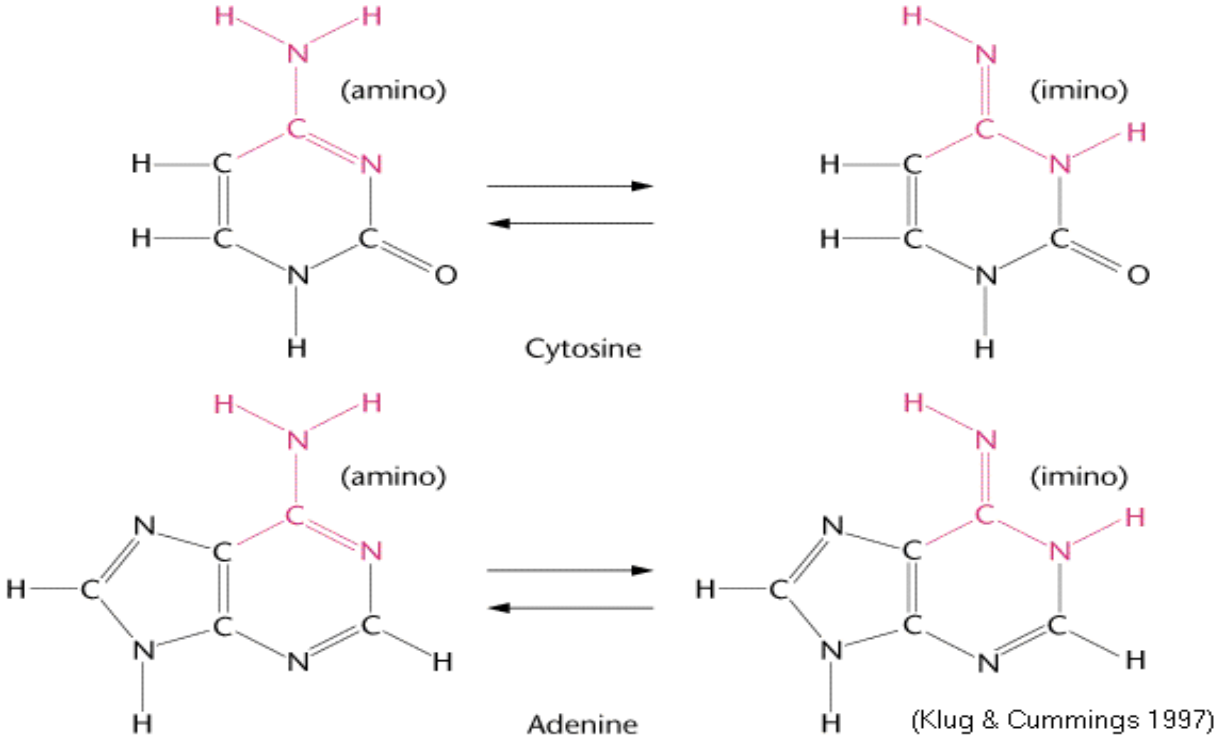


FIG 3.4: AMINO-IMINO CONVERSION OF CYTOSINE AND ADENINE

ADENINE

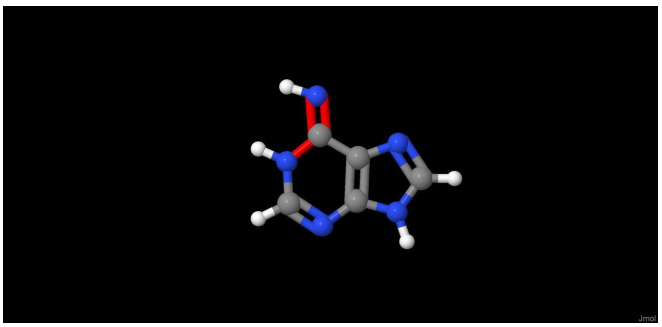


FIG 3.5: RED BONDS IN ADENINE DEPICTS THE BONDS WHICH ARE BEING INTERCHANGED FOR CONVERSION

CYTOSINE

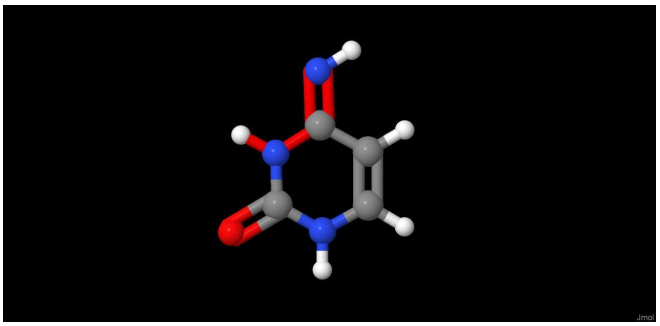


FIG 3.6: RED BONDS IN CYTOSINE DEPICTS THE BONDS WHICH ARE BEING INTERCHANGED FOR CONVERSION

GUANINE

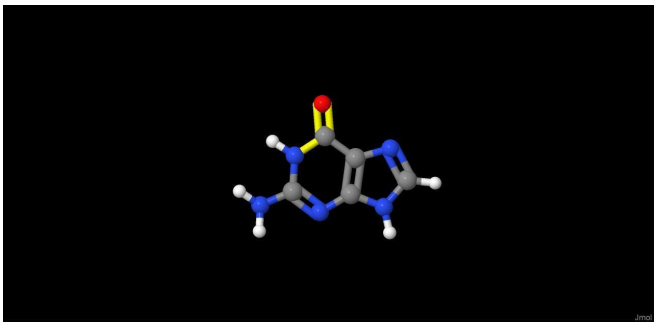


FIG 3.7: YELLOW BONDS IN GUANINE DEPICTS THE BONDS WHICH ARE BEING INTERCHANGED FOR CONVERSION

THYMINE

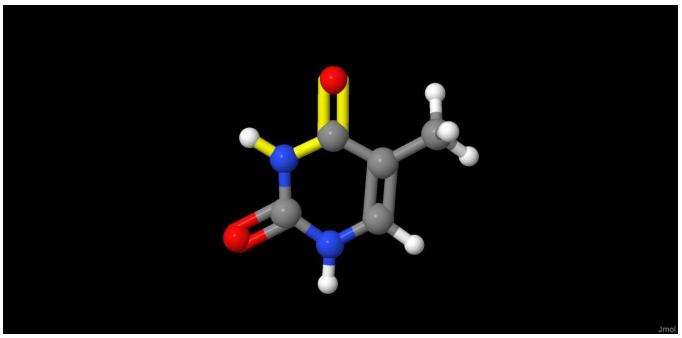


FIG 3.8: YELLOW BONDS IN THYMINE DEPICTS THE BONDS WHICH ARE BEING INTERCHANGED FOR CONVERSION

Normal Base Pairing in DNA is A-T and G-C. The tautomers forms are capable of unusual base pairings like T-G and C-A.

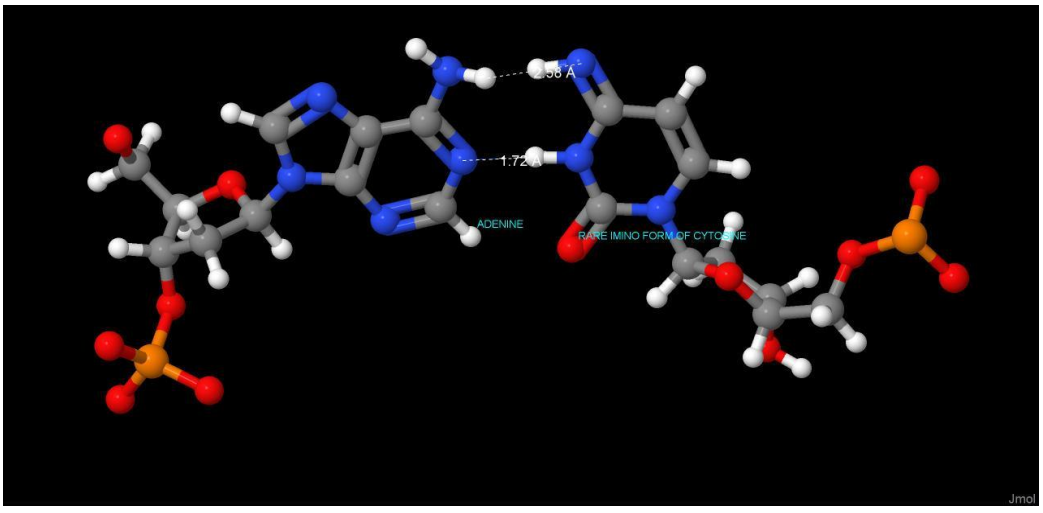


FIG 3.9: C-A MISMATCH ; used mol files(C-A MISMATCH) RARE IMINO FORM OF CYTOSINE PAIRS WITH ADENINE.mol

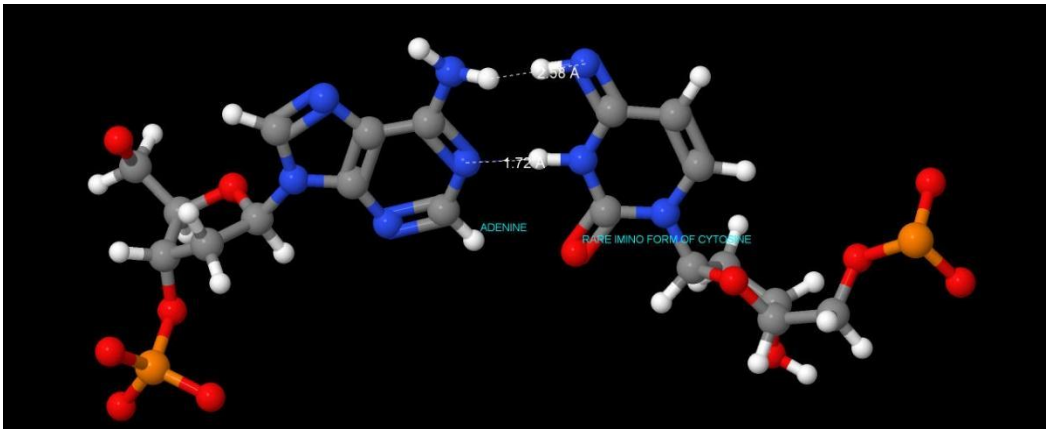


FIG 3.10: C-A MISMATCH ; used mol files(C-A MISMATCH) RARE IMINO FORM OF CYTOSINE PAIRS WITH ADENINE.mol

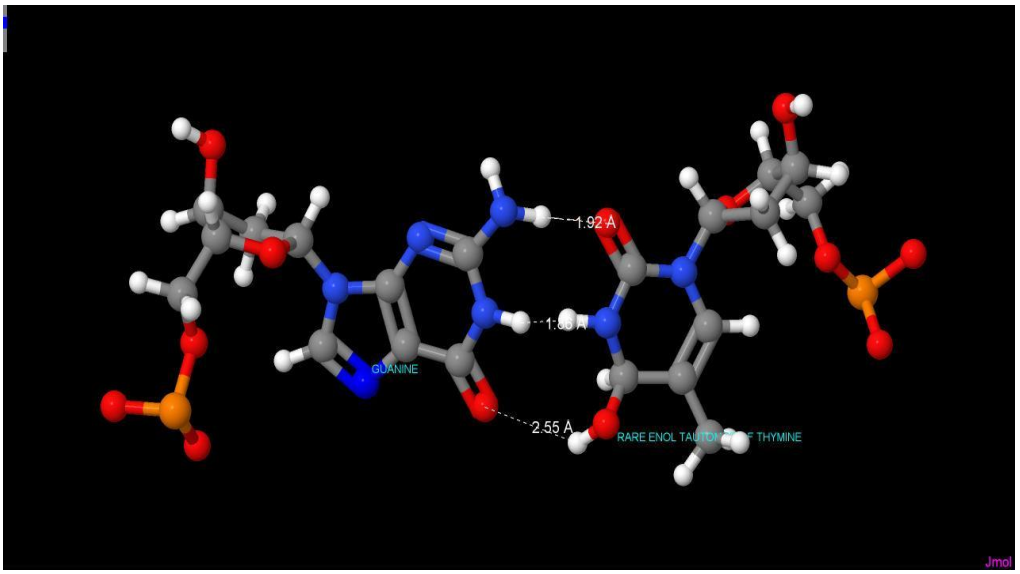


FIG 3.11: G-T MISMATCHES; used mol files; NON-CANONICAL BASE-PAIRING(RARE TAUTOMER OF THYMINE[ENOL FORM] WITH GUANINE).mol

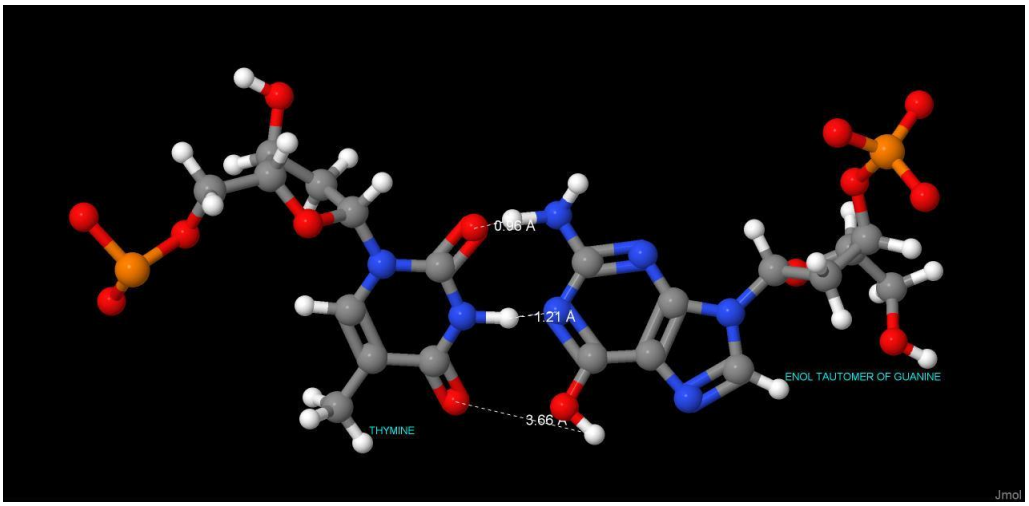


FIG 3.12: G-T MISMATCHES; used mol files; NON-CANONICAL BASE-PAIRING(RARE TAUTOMER OF THYMINE[ENOL FORM] WITH GUANINE).mol

3.2 How tautomeric shift in a base in DNA may lead to mutation?

Tautomeric shifts causing mutations. The rare, less stable tautomeric forms of base exist for only very short periods of time. However, if a base existed in the rare form at the moment that it was being replicated or being incorporated into a nascent DNA chain, a mutation might result: the rare imino or enol bases can form adenine-cytosine and Guanine-Thymine base pairs.The net effect of such an event and the subsequent replication required to segregate the "mismatched" base-pair is an A-T to G-C or G-C to A-T base pair substitution.

3.3 WHY DNA HAS SOME UNUSUAL SEQUENCES?

These sequences give different variations of DNA structure as hairpin, cruciform and H-DNA which are important in replication, transcription and recombination of DNA.

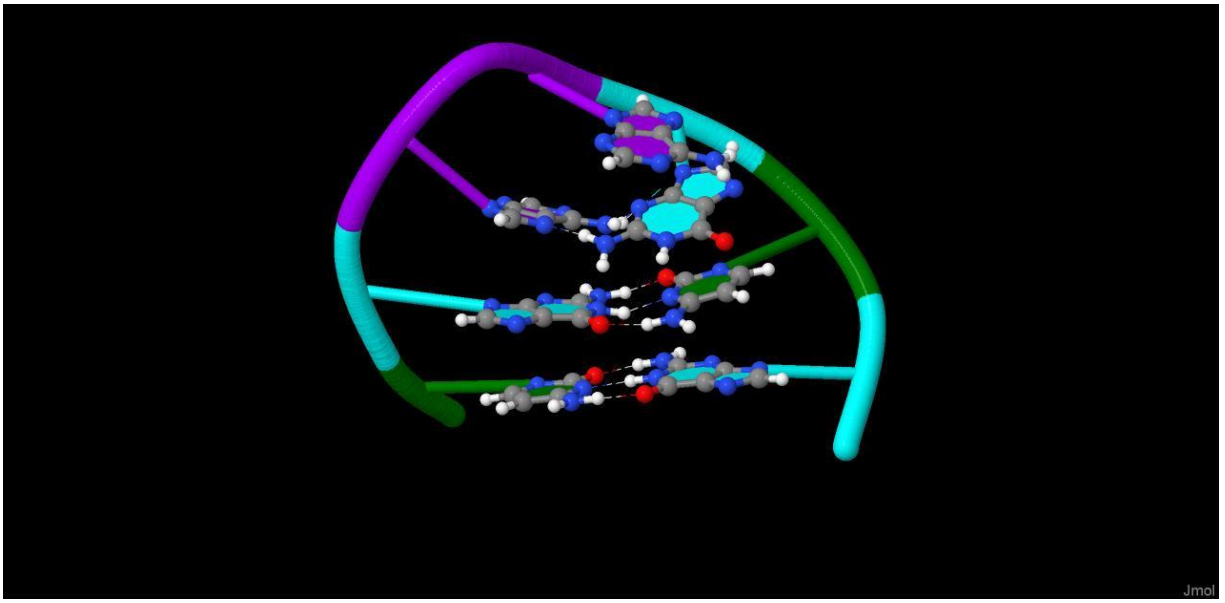


Fig 3.13: G-A mismatch leading to hairpin structure; pdb id -1KR8

(in the above figure adenine is violet, cyan is denoting guanine, cytosine is shown by green)

3.4 Methods for Studying Tautomerism in Nucleic Acids

Studying tautomerism in aqueous conditions is challenging. While tautomerism of nucleic acid bases has been studied in aprotic solvents, gas phase or excited state conditions, the conclusions derived are less relevant for RNA biochemistry. Under these conditions, tautomeric equilibria are significantly altered, and the relative proportion of major and minor tautomeric forms is changed, even for the canonical nucleobases . Challenges of studying tautomerism in aqueous conditions include the fast rates of the tautomeric equilibria, the low abundance of minor tautomers, and the high chemical and structural similarity, between the corresponding minor and major tautomeric species.

Minor tautomers of canonical nucleic acid bases and their analogs are rare, transient, and interconvert on a fast time scale. Recent developments of spectroscopic methods have allowed their direct identification in nucleic acids and in nucleoside analogs . Prototropic

tautomers differ from one another based on the position of protons, and the position of double bonds. Therefore, methods that are capable of detecting and distinguishing protons in different chemical environment (for example, NMR) are useful for the direct identification of tautomeric forms in both nucleic acids and nucleoside analogs . Similarly, methods that provide information on the bond order and strength of a specific chemical bond (such as IR and Raman spectroscopy) are useful for distinguishing between keto-enol or amino-imino tautomeric forms, because the functional groups in each tautomeric pair have very different and characteristic vibrational properties.

JMOL COMMANDS

console commands used for usual and unusual base pairings

- **calculate HBONDS**
- **minimize ADDHYDROGENS**
- **color HBONDS**

We can use the" right click jmol menu" to show HBONDS and Change the color of HBONDS . to minimize we can use modelkit menu as an alternative to script commands.

TAUTOMERISM (ADENINE); use adenine (lactam form).mol

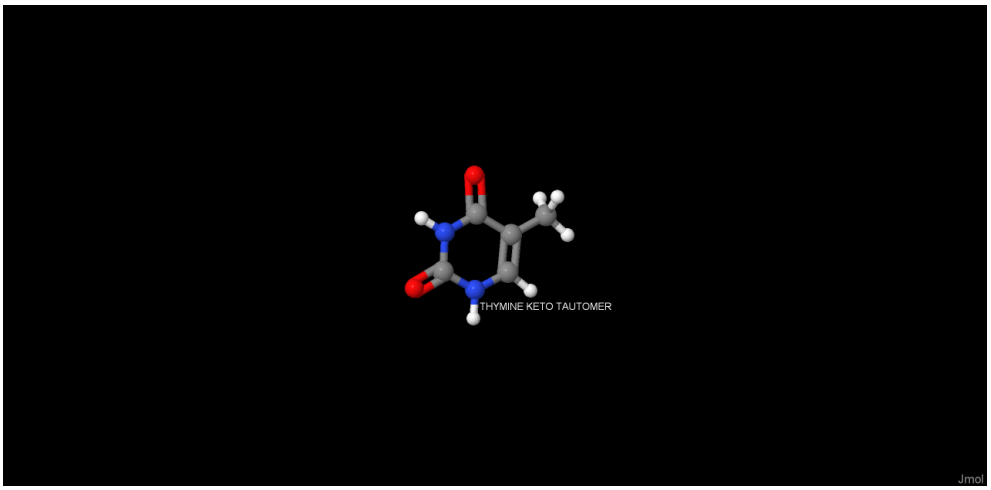
$Capture"adenine amino-imino tautomerism"SCRIPT" SELECT ({12});SPIN ON; label ADENINE(LACTAM FORM); SELECT NONE;draw arr1 arrow diameter 0.1 (NITROGEN) (atomno =9 or atomno =10);DELAY 3;draw arr2 arrow diameter 0.1 (NITROGEN) (atomno =9 or atomno =1);DELAY 3;DRAW OFF;CONNECT @1@9 SINGLE;delay 3; CONNECT @9@10 DOUBLE;delay 3; SELECT ({12}); label ADENINE(LACTIM FORM); hide ({11}); minimize ADDHYDROGENS;draw off;SELECT NONE ;draw arr1 arrow diameter 0.1 (NITROGEN) (atomno =9 or atomno =10);DELAY 3;draw arr2 arrow diameter 0.1 (NITROGEN) (atomno =9 or atomno =1);DELAY 3;DRAW OFF;CONNECT @1@9 DOUBLE; DISPLAY ALL;HIDE ({15});CONNECT @9@10 SINGLE;SELECT ({12}); label ADENINE(LACTIM FORM);SPIN OFF; LOOP" $Capture end

https://drive.google.com/file/d/116FlTWyuXiPWSVkDf-xRSlUE-bwwuC3z/view?usp=sharing

TAUTOMERISM (THYMINE); use thymine (lactam form).mol

$spin on;select @14; label THYMINE KETO TAUTOMER; draw arr1 arrow diameter 0.1 (OXYGEN)(atomno=3 or atomno=6); delay 5;connect @6@3 SINGLE;draw off; minimize ADDHYDROGENS;draw arr2 arrow diameter 0.1 (NITROGEN)(atomno=3 or atomno=8);delay 5; connect @8@3 DOUBLE;hide ({14});draw off;select @14; label THYMINE ENOL TAUTOMER;delay 5;draw arr1 arrow diameter 0.1 (OXYGEN)(atomno=3 or atomno=6); delay 5;connect @6@3 DOUBLE;hide({14 15});draw off;delay 5;draw arr2 arrow diameter 0.1 (NITROGEN)(atomno=3 or atomno=8);delay 5; connect @8@3 SINGLE;hide ({15}); draw off;minimize ADDHYDROGENS;select @14; label THYMINE KETO TAUTOMER

$Capture end



https://drive.google.com/file/d/1KZPQDrT7CkG18lM9Qg0Ww_U5vEVmEliL/view?usp=sharing

SIMILAR COMMANDS WILL BE USED FOR THE OTHER TAUTOMERIC NITROGENOUS BASES.

CHAPTER 4
METABOLISM OF NUCLEOTIDES

4.1 DEAMINATION

The most frequent and important kind of hydrolytic damage is deamination of the base cytosine. Deamination is the removal of an amine group from a molecule. Enzymes that catalyse this reaction are called deaminases. Deamination can also occur non-enzymatically. Several nucleotide bases undergo spontaneous loss of their exocyclic amino groups (deamination). For example, under typical cellular conditions, deamination of cytosine (in DNA) to uracil occurs in about one of every 10 7 cytidine residues in 24 hours. This rate of deamination corresponds to about 100 spontaneous events per day, on average, in a mammalian cell. Deamination of adenine and guanine occurs at about 1/100th this rate.

The slow cytosine deamination reaction seems innocuous enough, but it is almost certainly the reason why DNA contains thymine rather than uracil. The product of cytosine deamination (uracil) is readily recognized as foreign in DNA and is removed by a repair system . If DNA normally contained uracil, recognition of uracils resulting from cytosine deamination would be more difficult, and unrepaired uracils would lead to permanent sequence changes as they were paired with adenines during replication. Cytosine deamination would gradually lead to a decrease in G≡C base pairs and an increase in A═U base pairs in the DNA of all cells. Over the millennia, cytosine deamination could eliminate G≡C base pairs and the genetic code that depends on them. Establishing thymine as one of the four bases in DNA may well have been one of the crucial turning points in evolution, making the long-term storage of genetic information possible.

- **The deamination of cytosine to uracil happens at a significant rate in cells. Deamination can be repaired by a specific repair process which detects uracil, not normally present in DNA; otherwise the U will cause A to be inserted opposite it and cause a C:G to T:A transition when the DNA is replicated.**
- **Deamination converts adenine to hypoxanthine, which hydrogen bonds to cytosine rather than to thymine; guanine is converted into xanthine, which continues to pair with cytosine, though with only two hydrogen bonds.**
- **Deamination of 5-methylcytosine to thymine also occurs. 5- Methylcytosine occurs in the human genome at the sequence 5'CpG3', which is normally avoided in the coding regions of genes. if the meC is deaminated to T, there is no repair system which can recognise and remove it (because T is normal base in DNA). This means that whatever CpG occurs in genes it is a "hotspot" for mutation.**

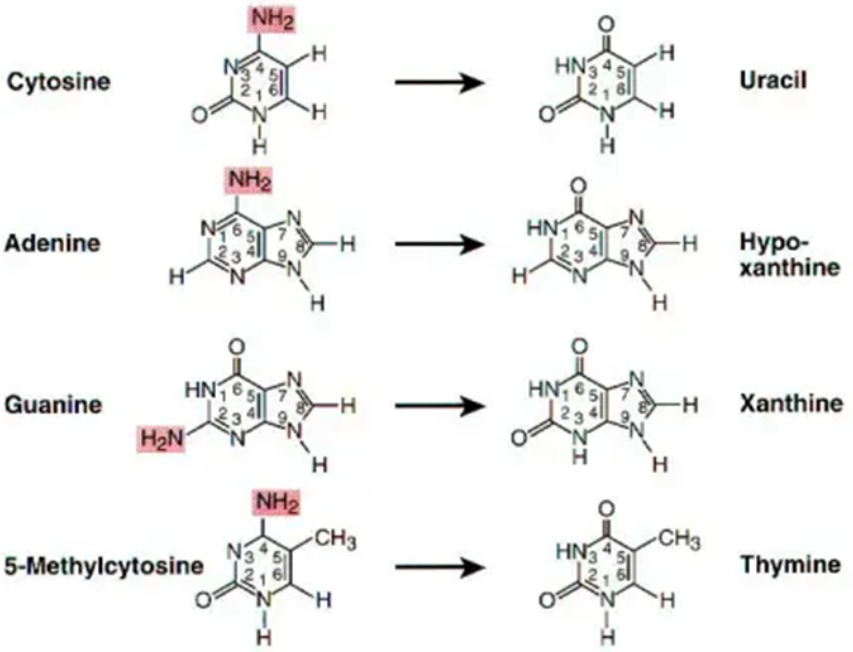


FIG 4.1: LEHNINGER'S PRINCIPLE OF BIOCHEMISTRY PG N0:-1086

4.2 DEPURINATION

DNA also undergoes depurination by spontaneous hydrolysis of the N-glycosyl linkage, and this produces an abasic site (that is deoxyribose lacking a base) in the DNA.
Depurination in DNA is a chemical reaction of purine deoxyribonucleosides, deoxyadenosine and deoxyguanosine, in which the β-N-glycosidic bond is hydrolytically cleaved releasing a nucleic base, adenine or guanine. The base is lost, creating a DNA lesion called an AP (apurinic, apyrimidinic) site or abasic site. Purines are lost at a higher rate than pyrimidines. As many as one in 10^5 purines (10,000 per mammalian cell) are lost from DNA every 24 hours under typical cellular conditions. Depurination of ribonucleotides and RNA is much slower and less physiologically significant.

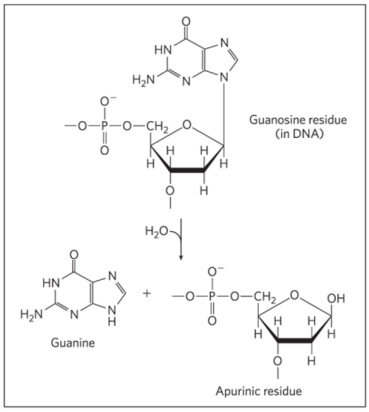


FIG 4.2:DEPURINATION(LEHNINGER'S PRINCIPLE OF BIOCHEMISTRY PG N0:-1086)

4.3 THYMINE DIMERS

Ultraviolet radiation (UV radiation) cross-links adjacent pyrimidines on the same DNA strand, forming pyrimidine dimers, usually thymine dimers. Figure shows the structure of a thymine dimer and illustrates how it interrupts base-pairing between the two DNA strands. These dimers block DNA replication because the replication machinery cannot tell which bases to insert opposite the dimer. As we will see, replication sometimes proceeds anyway, and bases are inserted at random. If these are the wrong bases, a mutation results. Non-enzymatic reactions promoted by radiation. UV light induces the condensation of two ethylene groups to form a cyclobutane ring. In the cell, the same reaction between adjacent pyrimidine bases in nucleic acids forms cyclobutane pyrimidine dimers. This happens most frequently between adjacent thymidine residues on the same DNA strand . A second type of pyrimidine dimer, called a 6-4 photoproduct, is also formed during UV irradiation. Near-UV radiation (with wavelengths of 200 to 400 nm), which makes up a significant portion of the solar spectrum, is known to cause pyrimidine dimer formation and other chemical changes in the DNA of bacteria and of human skin cells.

Ultraviolet radiation has great biological significance; it is present in sunlight, so most forms of life are exposed to it to some extent. The mutagenicity of UV radiation explains why sunlight can cause skin cancer: Its UV component damages the DNA in skin cells, which leads to mutations that sometimes cause those cells to lose control over their division.

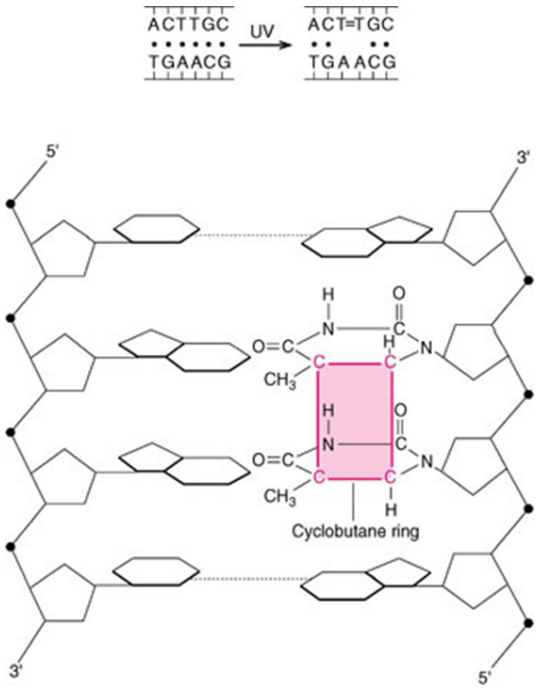


FIG 4.3: THYMINE DIMER CYCLOBUTANE RING(https://biotechkhan.files.wordpress.com/2014/10/fig3.jpg)

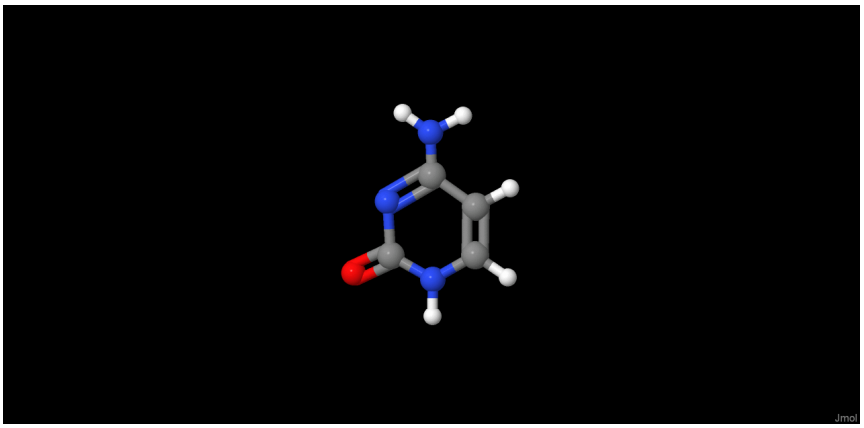
JMOL COMMANDS

DEAMINATION (CYTOSINE->URACIL) ; use cytosine deamination.mol

$hide ({12 13})

$ Capture"CYTOSINE DEAMINATION1"SCRIPT"SELECT@10;LABEL CYTOSINE;spin on;draw arr1 arrow diameter 0.1 (OXYGEN) (atomno =3 or atomno=6);DELAY 5;HIDE ({5 11 14}); draw arr1 arrow diameter 0.1 (CARBON) (atomno =3 or atomno =7);SELECT@10;LABEL URACIL;CONNECT@7@3 SINGLE;DELAY 5;DRAW OFF;delay 5;draw arr1 arrow diameter 0.1 (OXYGEN) (atomno =3 or atomno=6);DELAY 5; draw arr1 arrow diameter 0.1 (CARBON) (atomno =3 or atomno =7);DISPLAY ALL;HIDE ({12 13});SELECT@10;LABEL CYTOSINE;CONNECT@3@7 DOUBLE;draw off;delay 3;spin off; LOOP"

$Capture end

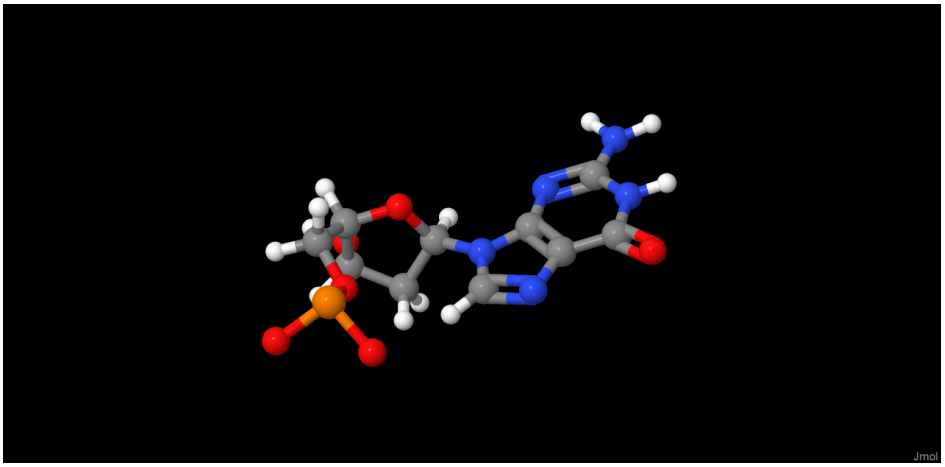


https://drive.google.com/file/d/17i7HAMReypm2wNrcfpKSgggOOL-DDOIe/view?usp=sharing

DEPURINATION (GUANINE) ; use guanosine residue.mol

$Capture" guanine depurination*"SCRIPT" select @3; label GUANOSINE RESIDUE;draw arr1 arrow diameter 0.1 (NITROGEN) (atomno =12 or atomno =11);SPIN ON;DELAY 3;DRAW OFF;SELECT ({10 11});CONNECT (SELECTED)(SELECTED)DELETE;select all;LABEL OFF;DELAY 3;SELECT @30;LABEL APURINIC RESIDUE;COLOR LABEL CYAN;SELECT @23;LABEL GUANINE;COLOR LABEL CYAN;SPIN OFF;minimize ADDHYDROGENS;Hide ({34 35});delay 5;SPIN ON; draw arr1 arrow diameter 0.1 (NITROGEN) (atomno =12 or atomno =11);DELAY 3;DRAW OFF;SELECT ALL;LABEL OFF; CONNECT @11@12 SINGLE;select@3;minimize ADDHYDROGENS;Hide ({34 35 37});Label GUANOSINE RESIDUE;DELAY 3;SPIN OFF;LOOP"

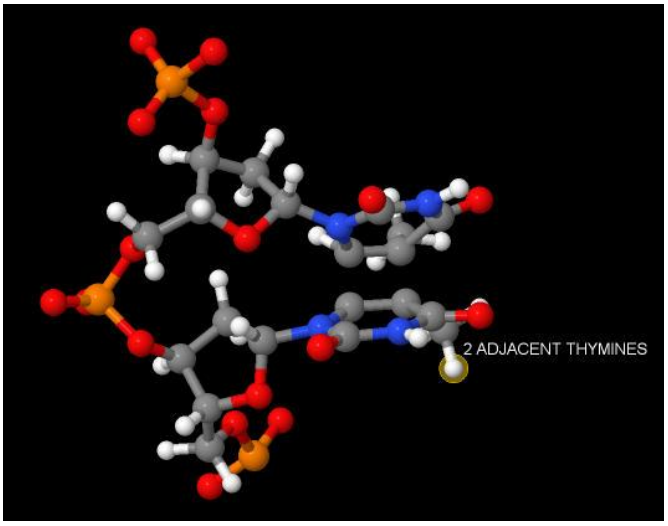
$Capture end



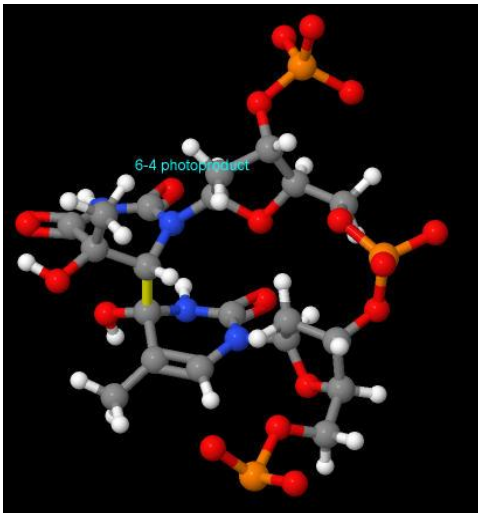
https://drive.google.com/file/d/1qeLIn11TOzonihb0ijNXO03TT--JJHZo/view?usp=sharing

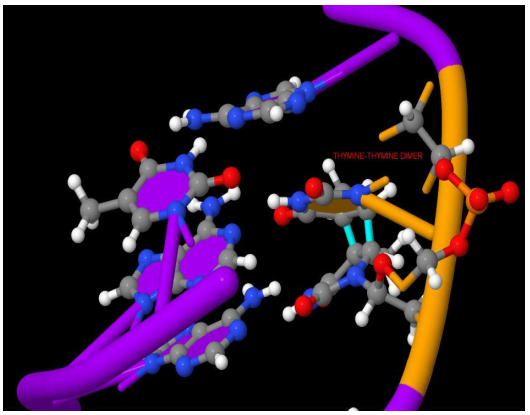
THYMINE DIMERS

PDB ID USED: 1TTD



—-UV RADIATION----->





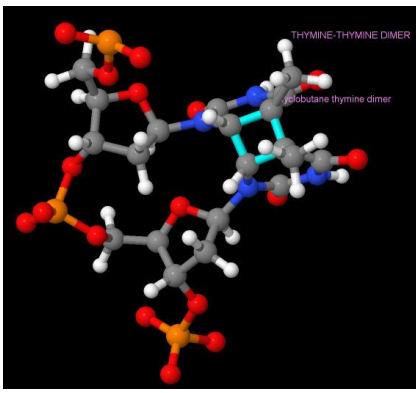


FIG 4.4: SHOWS 2 ADJACENT THYMINES
FIG 4.5: DEPICTS 6-4 PHOTOPRODUCT FORMED DUE TO REACTION OF UV
FIG 4.6: DEPICTS DIMER FORMED DUE TO REACTION OF UV HIGHLIGHTED BY CYAN.(A CYCLOBUTANE)
FIG 4.7: CYCLOBUTANE STRUCTURE OF DIMER

CONNECT COMMAND WAS USED TO CONNECT THE 2 THYMINES ON THE CONSOLE

-CONNECT@1st atomno.@2nd atomno. SINGLE

-CONNECT (SELECTED)(SELECTED)DELETE

COLOR AND LABEL COMMAND TO HIGHLIGHT THE BONDS AND THYMINES

-Select atoms of the dimer; color BONDS cyan;LABEL thymine-thymine dimer;COLOR LABEL CYAN

CHAPTER 5

NUCLEOTIDE DEGRADATION

The breakdown or cleavage of DNA and RNA or more appropriately their nucleotides through several catabolic ways. Purine and pyrimidine nucleosides can either be degraded to waste products and excreted or salvaged to form NUCLEOTIDE COMPONENTS. Nucleotide degradation or catabolism of nucleotides are basic metabolic pathways.

Nucleotides are first converted to nucleosides then these nucleosides may be directly absorbed by intestinal mucosa or undergo further degradation to free bases.

Nucleotide Degradation involves- 1) Catabolism of purines & 2) Catabolism of pyrimidines

5.1 CATABOLISM OF PURINES

The major pathways of purine nucleotide catabolism in animals and other organisms may differ but all of these pathways lead to uric acid.

DEGRADATION OF PURINES

CATABOLISM OF ADENINE NUCLEOTIDE:

An enzyme purine-5-nucleotidase hydrolyzes adenylate as a result adenosine is produced.

- **Adenosine deaminase removes ammonia from adenosine and forms inosine.**
- **Purine nucleoside phosphorylase phosphorylases inosine to ribose-1 phosphate and hypoxanthine.**
- **Xanthine oxidase then converts hypoxanthine to xanthine and then to uric acid.**

CATABOLISM OF GUANINE NUCLEOTIDE

- **GMP is hydrolyzed by purine-5-nucleotidase into guanosine**
- **Purine nucleoside phosphorylase phosphorolyses guanosine into ribose-1 phosphate and guanine.**
- **Guanine deaminase deaminates guanine to xanthine and produces ammonia.**
- **Oxidation of xanthine to uric acid is brought about by Xanthine oxidase.**

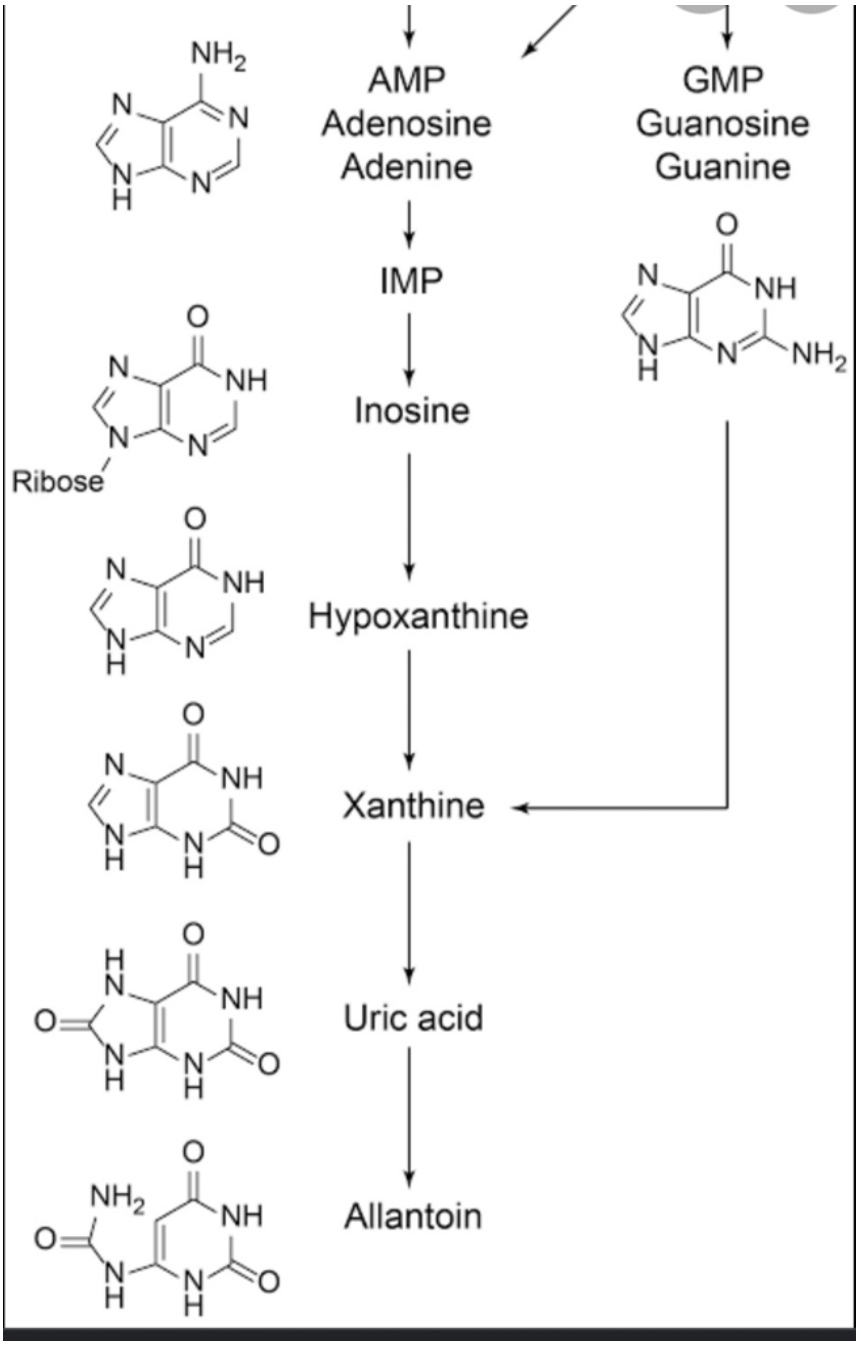


FIG 5.1: DEGRADATION OF PURINES ADENINE AND GUANINE

5.2 CATABOLISM OF PYRIMIDINES

Animal cells degrade pyrimidine nucleotides to their component bases. These reactions like those of purine nucleotides occur through DEPHOSPHORYLATION, DEAMINATION and GLYCOSIDIC BOND CLEAVAGES. The resulting uracil and thymine are then broken down in the liver through reduction rather than oxidation as in purine catabolism.

B-alanine and B-aminoisobutyrate are the end products of pyrimidine catabolism. Then these are converted to malonyl-CoA and methylmalonyl-CoA for further utilisation.

DEGRADATION OF PYRIMIDINES

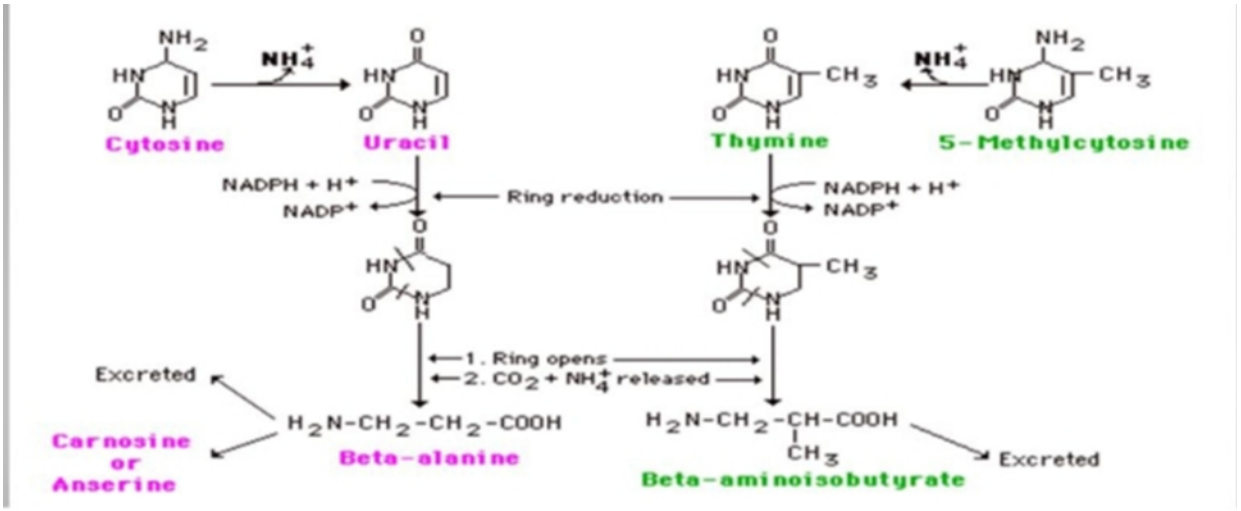


FIG 5.2: DEGRADATION OF PYRIMIDINES CYTOSINE, URACIL AND THYMINE

CATABOLISM OF CYTOSINE AND URACIL

- **Nucleotidases dephosphorylates pyrimidines to respective nucleosides Nucleosides are then phosphorolysed into free pyrimidines and ribose-1 phosphate by PI and nucleoside phosphorylasese.**
- **Dihydrouracil dehydrogenase uses NADPH to reduce uracil to 5,6-dihydrouracil.**
- **Hydropyrimidine hydrase hydrolyzes 5,6-dihydrouracil into B-ureidopropionic acid.**
- **B-ureidopropiomase hydrolyzes 5-ureidopropionic acid into carbon dioxide, ammonia and B-alanine.**
- **B-alanine can be used to produce anserine or CoA or can be oxidized to acetate, ammonia and carbon dioxide.**

CATABOLISM OF THYMINE:

- **Thymine dehydrogenase with the help of NADH reduces 5 methylcytosine to dihydrothymine.**
- **Hydrase causes the hydrolysis of dihydrothymine to give Beta-ureidaisobutyrase acid.**

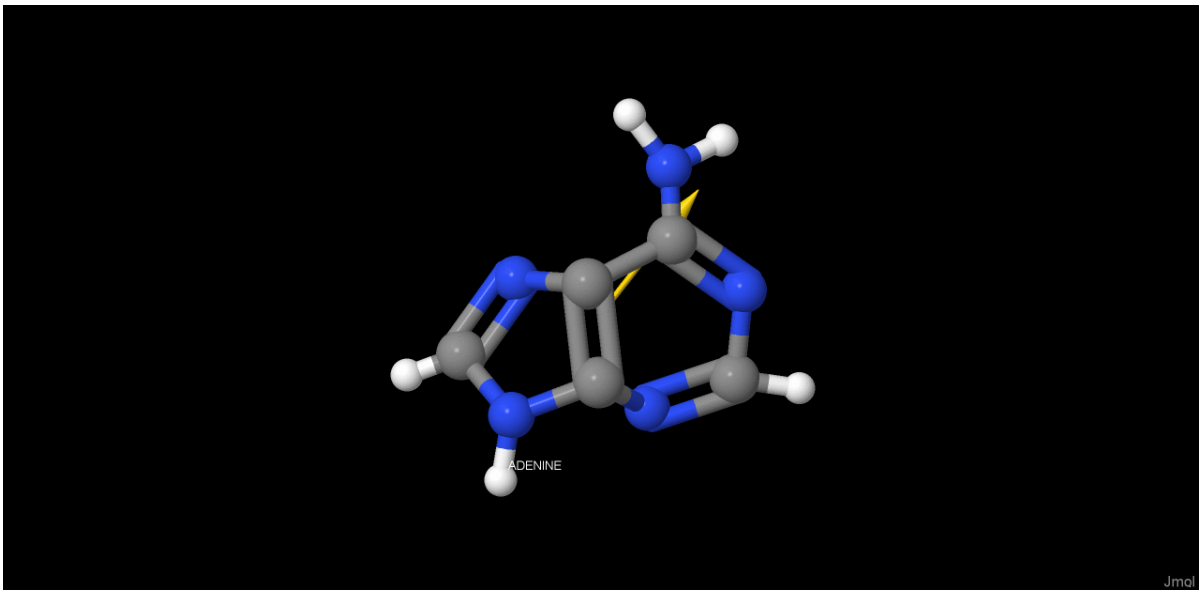
- **B-ureidaisobutyrase hydrolyzes B-ureidoisobutyric acid into carbon dioxide, ammonia and B-amino isobutyrate.**

JMOL COMMANDS

DEGRADATION OF PURINES (ADENINE/ADENOSINE -> URIC ACID -> ALLANTOIN); USE adenine degradation.mol

$HIDE ({10 12 16})

$Capture"ADENINE DEGRADATION"SCRIPT"SELECT ({15}); Label ADENINE; SELECT NONE;draw arr1 arrow diameter 0.1 (HYDROGEN) (atomno =9 or atomno=14);SPIN ON;DELAY 5;HIDE ({9 10 11 13 16 }); SELECT ({15}); Label HYPOXANTHINE; SELECT NONE ;DELAY 3; draw arr1 arrow diameter 0.1 (NITROGEN) (atomno =1 or atomno =9);DELAY 3;CONNECT@9@1 SINGLE;DELAY 5;DRAW OFF;DELAY 3;CONNECT @2@3 SINGLE; DELAY 3;draw arr1 arrow diameter 0.1 (OXYGEN) (atomno =2 or atomno=21);DELAY 3;HIDE ({9 11 13 14 16}); SELECT ({15}); Label XANTHINE; SELECT NONE ;DELAY 5;DRAW OFF; draw arr1 arrow diameter 0.1 (OXYGEN) (atomno =6 or atomno=25);delay 3; HIDE ({9 11 13 14 17 }); SELECT ({15}); Label URIC ACID; SELECT NONE ;draw off;delay 3;CONNECT @6@7 SINGLE;DELAY 5; minimize ADDHYDROGENS;DELAY 3; CONNECT @7@8 DELETE;DELAY 3; minimize ADDHYDROGENS; SELECT ({15}); Label ALLANTOIN; SELECT NONE; SPIN ON; DELAY 6; SPIN OFF"



https://drive.google.com/file/d/1y4arbNbvwlnN-4uaXCeMBUMetIIiL8-K/view?usp=sharing

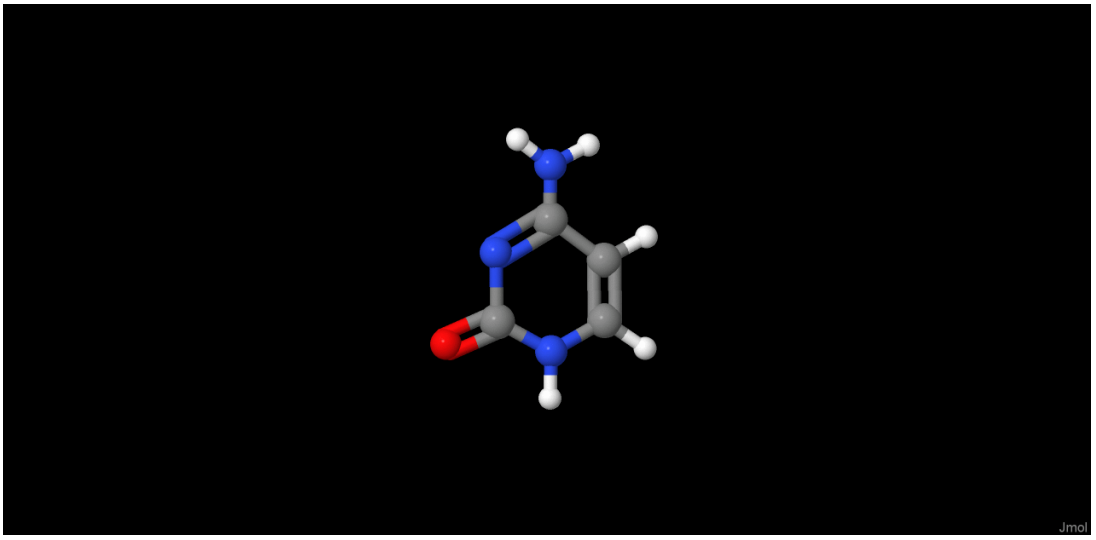
DEGRADATION OF PYRIMIDINES (CYTOSINE/URACIL -> BETA-ALANINE);

use cytosine deamination.mol

$HIDE ({12 13})

$Capture"CYTOSINE/URACIL DEGRADATION"SCRIPT" spin on;SELECT@10;LABEL CYTOSINE;draw arr1 arrow diameter 0.1 (OXYGEN) (atomno =3 or atomno=6);DELAY 5;HIDE ({5 11 14}); draw arr1 arrow diameter 0.1 (CARBON) (atomno =3 or atomno =7);SELECT@10;LABEL URACIL;CONNECT@7@3 SINGLE;DELAY 5;DRAW OFF;delay 3;connect @4@8 delete; delay 5;connect @7@3 delete;delay 5; delete ({3 6 13}); delay 3;connect @5@3 single;delay 5;connect @1@2 single;delay 3;select all; minimize ADDHYDROGENS; Select ({9}); label BETA-ALANINE; delay 5;minimize ADDHYDROGENS;delay 5; spin on"

$ Capture end; spin off



https://drive.google.com/file/d/11G8xV2ILpO9Pl8HVpqVYxISUYZjE2F7Y/view?usp=sharing

CHAPTER 6

RNA BIOSYNTHESIS

6.1 STRUCTURE OF tRNA

Transfer ribonucleic acid (tRNA) is a type of RNA molecule that helps decode a messenger RNA (mRNA) sequence into a protein. tRNAs function at specific sites in the ribosome during translation, which is a process that synthesizes a protein from an mRNA molecule. Proteins are built from smaller units called amino acids, which are specified by three-nucleotide mRNA sequences called codons. Each codon represents a particular amino acid, and each

codon is recognized by a specific tRNA. The tRNA molecule has a distinctive folded structure with three hairpin loops that form the shape of a three-leafed clover. One of these hairpin loops contains a sequence called the anticodon, which can recognize and decode an mRNA codon. Each tRNA has its corresponding amino acid attached to its end. When a tRNA recognizes and binds to its corresponding codon in the ribosome, the tRNA transfers the appropriate amino acid to the end of the growing amino acid chain. Then the tRNAs and ribosome continue to decode the mRNA molecule until the entire sequence is translated into a protein.

The secondary structure of a typical tRNA, The structure consists of hydrogen bonded stems and associated loops, which often contain nucleotides with modified bases (e.g. inosine, ribothymidine, pseudouridine, methylguanosine).

The tertiary structure of all tRNAs is similar to that of $tRNA^{ALA}$, at left, a canonical "L-shaped" molecule. As can be seen, the "cloverleaf" secondary structure results in a complex three dimensional folding of the molecule. The amino acid attachment site at the 3' end and the anticodon loop are observed at the two ends of the "L."

The hydrogen bonded stems stabilize the tertiary structure. The Acceptor and Anticodon stems are also present along with Modified bases in the Anticodon, T, and D loops . The anticodon bases are seen on the anticodon loop. The 5'wobble base that pairs with the 3' base of the mRNA .

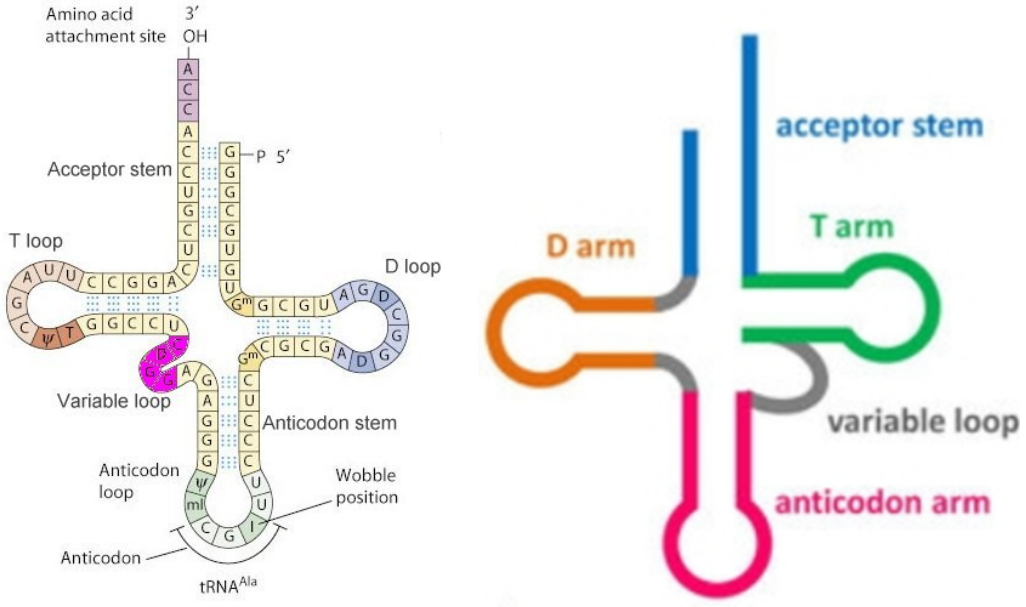


FIG 6.1:DETAILED tRNA STRUCTURE

JMOL COMMANDS

PDB ID USED : 1EHZ

- **select atoms; color “of your choice”;select “a particular atom no.”; Label “arm/loop”; color LABEL “any color of your choice”**

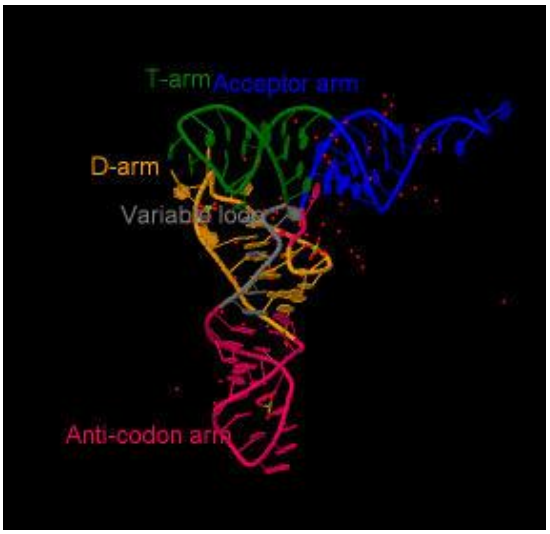


FIG 6.2: SHOWN DIFFERENT LOOPS OF tRNA WITH THE HELP OF DISTINCT COLORS AND LABELS

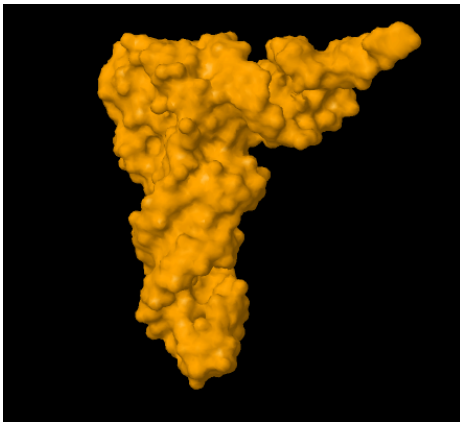


FIG 6.3: COLOURED SURFACE tRNA

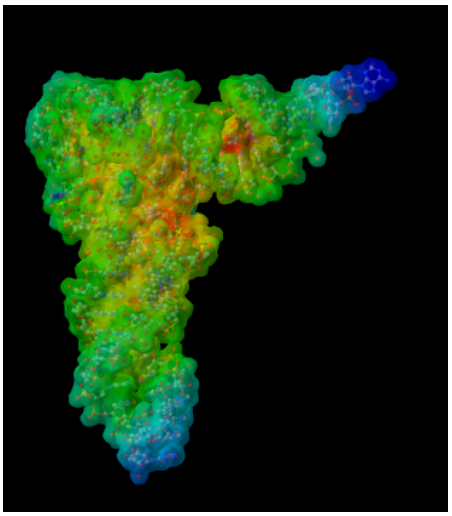


FIG 6.4: SHOWN MOLECULAR ELECTRO-POTENTIAL SURFACE TO CONFIRM THE PRESENCE OF AMINO ACIDS/ALANINE IN THIS CASE.

for surfaces we use console command

- isoSurface isosurface1 molecular

-color isosurface" -"

-isoSurface isosurface1 off

using "right click jmol taskbar"

-select Surfaces -> molecular electrostatic potential(all ranges)-> the surface is displayed on screen

The blue colour of molecular electrostatic potential surface at ends of anticodon arm and acceptor arm confirms that alanine is present there because of its highly positive PI=+6.02. Also confirms amino acids are present on the anticodon arm and the acceptor arm.

CONCLUSION

Throughout the project I used various Jmol tools to understand the biomolecules better. In this project I explored symmetry of DNA, Labelling, HBonding, Surfaces, Animations , Measurements, schematic shapes for secondary and tertiary structures and many more different uses of various tools and script commands which can be beneficial for learning basic concepts for students and can be helpful for teachers.

RESOURCES FOR PERFORMING ANIMATIONS AND ANIMATIONS CAN BE ACCESSED FROM THE FOLLOWING LINK-

https://drive.google.com/drive/folders/1ymg3pSoW4c9mkt2RiTtKXCEs2JxHv4mD?usp=sharing

References

1.Perona, J.J., Rould, M.A., Steitz, T.A.. Structural basis for transfer RNA aminoacylation by *Escheria coli* glutaminyl-tRNA synthetase. Biochemistry 32:8758. (1993).

2.Perona, J.J., Rould, M.A., Steitz, T.A.. Structural basis of anticodon loop recognition by glutaminyl-tRNA synthetase. Nature 352:213. (1991).

3.https://en.wikibooks.org/wiki/Structural_Biochemistry/Nucleic_Acid/Difference_between_DNA_and_RNA

4. The Deoxyribose molecule - rotatable in 3 dimensions

5. https://home.csulb.edu/~cohlberg/Jmolmanual.pdf

6. The Structure and Function of Nucleic Acids

7. Nucleic Acids

8.Deamination – Biotech Khan

9. https://www.mun.ca/biology/scarr/Amino_vs_Imino.html

10. https://www.pnas.org/doi/full/10.1073/pnas.1114496108

11. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4274630/

12. https://www.frontiersin.org/articles/10.3389/fmolb.2021.823253/full

13.Lehninger Principles of Biochemistry 8th edition-CHAPTER 8- NUCLEOTIDES AND NUCLEIC ACIDS

14.What is ADP in biology - 24 Hours Of Biology

15.ATP-ADP Cycle

16. ATP & ADP - Biological Energy

17. Introduction,
Editor(s): Zachary F. Burton,
Evolution Since Coding,
Academic Press,
2018,
Pages xi-xiii,
ISBN 9780128130339,
https://doi.org/10.1016/B978-0-12-813033-9.02001-1.
(https://www.sciencedirect.com/science/article/pii/B9780128130339020011)

18. Biochemistry 5: Nucleic Acids Overview – learn-biology

19. https://themedicalbiochemistrypage.org/biochemistry-of-nucleic-acids/

20.ATP AND BIOLOGICAL ENERGY

21.Fedeles BI, Li D, Singh V. Structural Insights Into Tautomeric Dynamics in Nucleic Acids and in Antiviral Nucleoside Analogs. Front Mol Biosci. 2022 Jan 25;8:823253. doi: 10.3389/fmolb.2021.823253. PMID: 35145998; PMCID: PMC8822119.

22. http://teme.alfa145.com/which-tautomeric-form-predominates

23.https://biochemden.com/tautomerization/

24. https://sandwalk.blogspot.com/2007/07/tautomers-of-adenine-cytosine-guanine.html

25. https://biotechkhan.wordpress.com/2014/10/14/dna-damage/

26. https://www.bionity.com/en/encyclopedia/Deamination.html
27. https://biotechkhan.wordpress.com/tag/depurination/
28. https://www.academia.edu/38956232/NUCLEOTIDE_DEGRADATION
29. tRNA / transfer RNA | Learn Science at Scitable - Nature
30./earth.callutheran.edu/Academic_Programs/Departments/BioDev/omm/jsmolnew/trna/trna.html