4241

Teaching Guide

Gene Machine

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JHRM software
A division of Human Relations Media

Apple II, II+, and IIe

<u>About the Author</u>

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GENE MACHINE

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INTRODUCTION

The molecular model of the genetic material, DNA (deoxyribonucleic acid) was first described by Watson and Crick in 1953. Since that time, there have been rapid and exciting advances in the study of the structure and function of the genetic material from a molecular perspective.

Gene Machine has been designed to allow students to explore some of the basic ideas concerning the structure and function of the nucleic acids, DNA and RNA (ribonucleic acid).

The program consists of:

- an introductory tutorial which allows students to review some of the basic principles

and sections on the following:

- DNA replication

- transcription of mRNA from a DNA template

- the genetic code

- translation of a polypeptide chain from an mRNA molecule.

This version of Gene Machine runs on Apple II, II+, or IIe computers with 48K RAM and DOS 3.3. A color monitor is preferable. A version for the TRS-80 Model III and Model IV computers is also available.

TARGET AUDIENCE

The program is designed for use by biology students at the high school level and is intended to complement their study of gene action. It is also appropriate for introductory college courses in biology and for high school and college chemistry courses that include aspects of the chemical basis of heredity. We recommend that the program be used with students who have some prior knowledge of the topic of molecular genetics.

LEARNING OBJECTIVES

The program is directed towards enhancing students' understanding of aspects of the structure and function of the nucleic acids, DNA and RNA.

In particular, the program aims to provide a review of—

- the base-pairing rules governing the synthesis of a DNA sequence complementary to a DNA primer strand.

- the base-pairing rules for synthesis of an RNA sequence from a DNA template strand.

- aspects of the process of translation of a polypeptide chain from an mRNA template, including the specification of the amino acid sequence in a polypeptide chain by the sequence of codons in mRNA.

The program is not designed to replace current teaching activities; rather it aims to complement and extend the usual teaching/learning activities in this area of molecular genetics.

The program is designed to give the user a high degree of control over the rate of progress through, and the sequence of execution of, the material.

RUNNING THE PROGRAM

When the disk is booted, Gene Machine will automatically load and commence running.

Users are first given the choice of reviewing aspects of the structure of the nucleic acids, DNA and RNA. Worksheet I in the Appendix is provided for students who wish to record the review information.

The program is menu-driven and the main menu is shown below:*

```
        YOU MAY . . .

  1.  MAKE DOUBLE-STRANDED DNA

  2.  TRANSCRIBE M-RNA FROM DNA

  3.  EXPLORE THE GENETIC CODE

  4.  LOOK AT REVIEW

  5.  END THE PROGRAM


  TYPE 1, 2, 3, 4 OR 5
```

A. If the first option is selected from the main menu--

"MAKE DOUBLE-STRANDED DNA"

the following choices are presented:

```
  YOU WILL BE SHOWN PORTION OF
    A SINGLE STRAND OF DNA . .

         YOU MAY

  1.  WATCH SYNTHESIS OF
      COMPLEMENTARY STRAND

  2.  BE IN CHARGE

  3.  RETURN TO MAIN MENU


  TYPE 1, 2 OR 3
```

* Because most Apple computers do not have upper and lower case, mRNA will be shown on the screen as M-RNA.

1. WATCH SYNTHESIS OF COMPLEMENTARY STRAND

A random sequence of five nucleotides in DNA is generated. The user can watch as the complementary strand is formed.

2. BE IN CHARGE

A random sequence of five nucleotides in DNA is generated. The user is required to pair each base in turn and form the complementary sequence by pressing the key corresponding to the matching base. A second sequence of five bases is then given for pairing. Following this, the user can choose to exit from this section or can continue with pairing more sequences.

B. If the second option is selected from the main menu--

"TRANSCRIBE M-RNA FROM DNA"

the following choices are presented:

```
FROM A SINGLE STRAND OF DNA . .

          YOU MAY

1.  WATCH M-RNA BEING TRANSCRIBED

2.  BE IN CHARGE

3.  RETURN TO MAIN MENU


TYPE 1, 2 OR 3
```

1. WATCH M-RNA BEING TRANSCRIBED

A random sequence of five nucleotides in DNA is presented. A user can simply watch as this sequence is transcribed into a sequence of five nucleotides in an mRNA molecule. This process is repeated.

For those users with a color monitor, the nucleotides are rendered in color. The color code is as follows:

adenine: magenta (reddish blue)   uracil: purple

thymine: blue   ribose: dark green

guanine: orange   phosphate: white

cytosine: yellow   deoxyribose: aqua
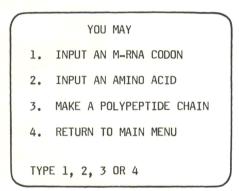
2. BE IN CHARGE

A random sequence of DNA nucleotides is generated. The user is required to identify the mRNA transcript that would be determined by this DNA template. The user identifies each mRNA nucleotide in turn by pressing the key corresponding to the matching base. This process is repeated.

Following this, the user can choose to exit from this section or can continue with transcribing more sequences.

C. If the third option is selected from the main menu--

"EXPLORE THE GENETIC CODE"

the following choices are presented:

```
           YOU MAY

1.   INPUT AN M-RNA CODON

2.   INPUT AN AMINO ACID

3.   MAKE A POLYPEPTIDE CHAIN

4.   RETURN TO MAIN MENU


TYPE 1, 2, 3 OR 4
```

1. INPUT AN M-RNA CODON

Users are required to input an mRNA codon. This is done by typing a three-letter sequence of bases from an mRNA molecule (e.g., AAG, UGG, etc.). As each letter is pressed, its symbol appears on the screen. When the codon is complete, the amino acid which is determined by that codon is shown on the screen. The user can continue with this process until he/she chooses to exit from this section.

2. INPUT AN AMINO ACID

The user is required to enter one of the standard three-letter abbreviations for an amino acid (e.g., ALA, CYS, etc.). A list of the twenty amino acids commonly found in proteins and their standard abbreviations is given in the Appendix (Table 1) and can also be called up from within the program.

After the user has entered an amino acid, the mRNA codon(s) which determine(s) this specific amino acid is (are) displayed on the screen.

This procedure can be continued as long as the user wishes.
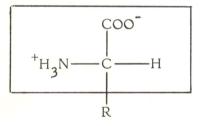
3. MAKE A POLYPEPTIDE CHAIN

The user selects the number of amino acids in the polypeptide chain:

```
┌─────────────────────────────────────┐
│                                      │
│   YOUR POLYPEPTIDE CHAIN WILL BE     │
│  ┌──────────┐                        │
│  │TRANSLATED│ FROM AN M-RNA MOLECULE │
│  └──────────┘                        │
│                                      │
│   CHOOSE THE NUMBER OF AMINO         │
│   ACIDS IN THE CHAIN (5-25)?         │
│                                      │
│                                      │
│                                      │
│   PRESS RETURN TO CONTINUE           │
│                                      │
└─────────────────────────────────────┘
```

Having defined the length of the polypeptide chain to be synthesized, the student is given information about the number of codons required, including a "start" and a "stop" codon.

The student then enters each codon in turn by typing a three-letter sequence, e.g., AAG. As each codon is completed, the corresponding amino acid is displayed on the screen. Each amino acid is brought into place by a tRNA molecule. The anticodon in the tRNA pairs with the codon of the mRNA. Adjacent amino acids are then joined by a peptide bond. In the program, this linking is symbolized by the appearance of a small block joining the amino acids.

The general formula of an amino acid may be shown as:

$$^+H_3N \longrightarrow \overset{\displaystyle COO^-}{\underset{\displaystyle R}{\overset{|}{\underset{|}{C}}}} \longrightarrow H$$

The portion shown in the block is common to all amino acids.
The side chain or R group distinguishes the different amino acids.
The symbols used for the amino acids in Gene Machine are color coded in terms of the nature of the side chain:
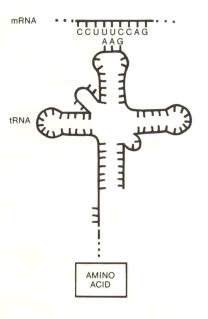
pink is used to denote amino acids with non-polar hydrophobic R groups

blue is used to denote amino acids with polar, but uncharged, R groups

green is used to denote amino acids with negatively charged R groups

white is used to denote amino acids with positively charged R groups

yellow is used to denote cysteine, the amino acid which can form a disulfide cross link with a second cysteine.

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When the polypeptide chain is completed, the entire sequence is shown on the screen.

```
YOUR POLYPEPTIDE HAS THE SEQUENCE
1    2    3    4    5    6
FMET-PHE-GLY-PRO-SER-GLY


THE INITIAL FMET IS REMOVED LATER

MAKE ANOTHER POLYPEPTIDE?  (Y/N)
```

THE UNDERLYING MODEL

1. The bases that comprise part of the nucleotides of DNA and RNA are restricted to the major bases: adenine, guanine, cytosine, and thymine or uracil, as appropriate. The so-called "minor" bases, which are chemical modifications of the major bases and which occur in small amounts in many organisms, are excluded from the program. (Minor bases include compounds, such as 5-methylcytosine, inosine, 5-hydroxy-methylcytosine.)

2. The processes of DNA replication, gene transcription, and gene translation are reduced to their simplest form. The program does not consider the number and complexity of enzymes involved in these processes.

3. The DNA presented in the program consists of very short nucleotide sequences. In fact, DNA typically consists of several hundred nucleotides organized into a double helix.

4. The process of transcription is reduced to a simplified form. The product of gene transcription is identified as in mRNA. The relationship of heterogeneous nuclear RNA (HnRNA) to mRNA is not introduced, nor are the concepts of exons and introns considered.

5. Each tRNA molecule consists of a chain of 80 nucleotides, folded into a complex three-dimensional shape. A specific tRNA molecule can transfer or carry a specific amino acid to the side of polypeptide synthesis. In the program, however, each tRNA molecule is simply represented by an anticodon, that is a sequence of three bases that is complementary to a specific codon in an mRNA molecule.

6. The base pairing between mRNA codons and tRNA anticodons is given in terms of A-T and C-G base pairing only. The "wobble hypothesis" is not included in the program.

7. The term "polypeptide" has been used in the program. Since many proteins consist of two or more polypeptide chains encoded by two or more separate genes, the term "polypeptide" has been used for the product of gene translation, rather than the term "protein."

SOME SUGGESTED ACTIVITIES FOR USE WITH THE PROGRAM

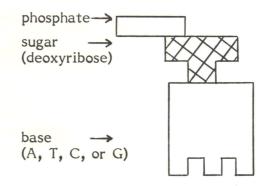Several activities which might be undertaken with Gene Machine are described below.

These activities are offered as a guide to possible uses of the program. It is suggested that teachers modify these activities as they wish or write alternative activities appropriate to the needs of their own students. For example, an instructor using the program as part of a study of DNA replication may choose for one group of students to introduce the antiparallel orientation of the two strands of the DNA molecule, and the polarity of each DNA strand with a 5' and a 3' terminus. For another group of students, discussion of these aspects may not be appropriate, and the sole focus could be the identification of the complementary base pairs in a DNA double-stranded molecule.

Activity A1: EXPLORING THE STRUCTURE OF DNA

Choose the first option from the main menu: "MAKE DOUBLE-STRANDED DNA" and then select the option: "BE IN CHARGE."

A short single strand of DNA will appear building from left to right. The basic unit of which DNA is built is called a nucleotide.

Note that each basic unit consists of--

phosphate →

sugar →
(deoxyribose)

base →
(A, T, C, or G)
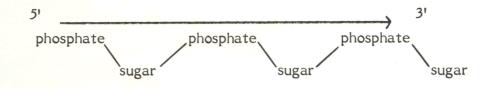
• How many nucleotides are present in the strand?

• How are adjacent nucleotides joined?

• To what is each base linked?

Note that--

• the two ends of the strand are different. The end of the strand at the left of the screen terminates with a phosphate grouping; this is known as the 5' end. The other end of the strand terminates with a sugar; this is known as the 3' end.

• the backbone of the strand consists of repeating phosphate and sugar units:

5'   3'

phosphate   phosphate   phosphate

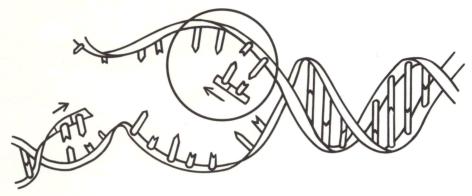sugar   sugar   sugar

• each base is linked to a sugar.

In the nucleus of the cell, DNA replicates when the original molecule partly unwinds, so that the two strands can separate. A new strand is formed by complementary base pairing on each of the original strands. The circled area in the diagram below shows the screen display. You should be aware that you see replication of one DNA strand only on the screen.

DNA REPLICATION

Arrows show direction of replication for each strand

Examine the template DNA strand, and type the letter corresponding to the complementary base. In this way, you will build up a new double-stranded DNA molecule. The new strand is oriented in the opposite direction from the first strand.

The two strands of a DNA molecule are said to be antiparallel.

Record the base sequence in the double-stranded DNA for at least 20 base pairs:

STRAND 1
5' end

STRAND 2
3' end

In the double-stranded DNA molecule you have built --

Number of A's =

Number of T's =

Number of C's =

Number of G's =

Because of the complementary base pairing in DNA, you should find that number of A's is equal to the number of T's, and similarly, the number of C's is equal to the number of G's.

Calculate the $\frac{A + T}{C + G}$ ratio for your double-stranded segment of DNA.

The A + T/C + G ratio differs in different organisms.

For example, DNA has been isolated from tissues of various species and has been analyzed.

Some typical results are shown in the following table:

SPECIES	A + T/C + G
human	1.67
cattle	1.26
Euglena	0.88
Drosophila	1.22
cow	1.04
cat	1.34
salmon	1.43

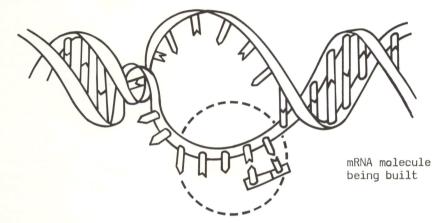From what organism might your sample of DNA have been isolated?

Activity A2: MAKING M-RNA

DNA is a double-stranded molecule, but only one of the two strands acts as the template for mRNA synthesis. The two DNA strands separate, and the mRNA is formed (or transcribed) on one of the separated DNA strands.

Select the option: "TRANSCRIBE M-RNA FROM DNA " and then select the option: "BE IN CHARGE."

A short single strand of DNA appears on the screen.

Build up an mRNA molecule by typing the key corresponding to the matching base, starting at the right-hand end.

TRANSCRIPTION
of mRNA from
DNA template

mRNA molecule
being built
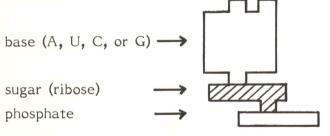
The circled area shows the screen display. Note that the screen display shows only one strand of the DNA molecule.

The mRNA is built up from building blocks also called nucleotides.

Each nucleotide in mRNA consists of

base (A, U, C, or G) ⟶

sugar (ribose) ⟶

phosphate ⟶

How does this nucleotide differ from that found in DNA?

Note that--

- the mRNA molecule is built up in an antiparallel direction to the DNA template strand

- the "backbone" of the mRNA strand consists of repeating units of

```
              sugar            sugar            sugar
            /      \         /      \         /
 phosphate           phosphate         phosphate
```

- each base is joined to a sugar molecule.

Complete the following table:

BASE IN DNA	MATCHING BASE IN mRNA
A	
T	
C	
G	

Record a sequence of at least 20 nucleotides in the single-stranded mRNA molecule you have built:

Number of A's =

Number of U's =

Number of C's =

Number of G's =

In double-stranded DNA, the number of C's is equal to the number of G's. Does a similar relationship hold for mRNA? Why/Why not?

Activity A3: MAKING A POLYPEPTIDE

A segment of one strand of the DNA double helix contains in its base sequence the information required for the synthesis of a specific polypeptide.

The information in DNA is first copied into an mRNA molecule. This process occurs in the nucleus and is called gene transcription.
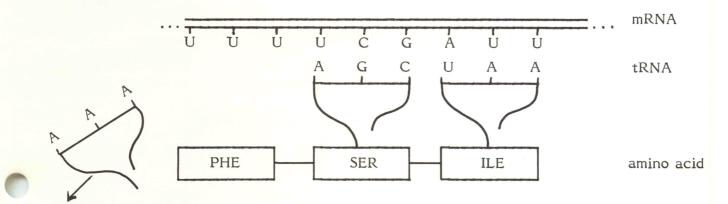
The mRNA molecule moves to the cytoplasm, where it attaches to the surface of ribosomes.

Amino acids are brought into place by specific tRNA molecules. Adjacent amino acids are joined by a peptide bond to form a polypeptide chain.

A sequence of 15 bases in an mRNA strand is shown in the table below. Write the sequence of bases in the DNA template strand from which this mRNA was transcribed.

base sequence in DNA							
base sequence in mRNA	"START"	G U G	C G C	A U A	A A A	C U G	"STOP"
amino acid sequence in polypeptide							

How many codons are represented by these 15 bases in mRNA? _____

How many amino acids will be encoded by these 15 bases? _____

Select the option: "EXPLORE THE GENETIC CODE" and then choose: "MAKE A POLYPEPTIDE CHAIN."

You will be asked to indicate the number of amino acids in the polypeptide chain. A reminder will be given that to make this polypeptide, you will also require a "start" and a "stop" codon.

Type in the mRNA codons one by one, commencing with a "start" codon. As each codon is typed, you will see an amino acid brought into place by a tRNA molecule.

Note that:

  • adjacent amino acids are joined by peptide bond.

  • each tRNA molecule releases its amino acid and moves away.

Finish the synthesis of your polypeptide chain by typing a "stop" codon. Write the amino acid sequence of your polypeptide chain in the table above.

  • What is the codon to "start" synthesis of a polypeptide chain?
  • Does the "start" codon put an amino acid into the chain?
  • What is the signal to terminate synthesis?
  • Does the "stop" signal put an amino acid into place in the chain?

Additional activity:

Make a polypeptide, using the mRNA sequence in the table below. The number of amino acids coded by this sequence is 6.

base sequence in mRNA	"START"	C U U	C U C	C U A	C U G	U U A	U U G	"STOP"
amino acid sequence in polypeptide								

What do your results illustrate about the nature of the genetic code?

Activity A4: EFFECTS OF MUTATION

Agents, such as chemicals or radiation, can cause a change in the base sequence of a DNA molecule.

If one of the bases in a DNA molecule is altered, the DNA is said to have undergone a mutation.

A DNA base sequence and the corresponding mRNA base sequence are shown below:

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18
DNA	T	A	C	T	A	T	G	C	C	A	T	G	A	C	C	A	T	T
mRNA	A	U	G	A	U	A	C	G	G	U	A	C	U	G	G	U	A	A
amino acid sequence	"START" F-MET			ILE			ARG			TYR			TRP			"STOP"		

Choose option 3: "EXPLORE THE GENETIC CODE" and then select: "INPUT AN M-RNA CODON."

Type in the mRNA codons in turn and record the sequence of amino acids. It should be as shown in the table above.

The DNA sequence could be changed by a mutagenic agent as follows: each of these changes affects the fourth codon in the mRNA.

Change base #10 in original DNA strand from A to G

ORIGINAL DNA	T A C	T A T	G C C	A T G	A C C	A T T
MUTATED DNA	T A C	T A T	G C C	G*T G	A C C	A T T
mRNA						
amino acid sequence						

Change base #11 in original DNA strand from T to A

ORIGINAL DNA	T A C	T A T	G C C	A T G	A C C	A T T
MUTATED DNA	T A C	T A T	G C C	A A*G	A C C	A T T
mRNA						
amino acid sequence						

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Change base #12 in original DNA strand from G to T

ORIGINAL DNA	T A C	T A T	G C C	A T G	A C C	A T T
MUTATED DNA	T A C	T A T	G C C	A T T*	A C C	A T T
mRNA						
amino acid sequence						

Change base #12 in original DNA strand from G to A

ORIGINAL DNA	T A C	T A T	G C C	A T G	A C C	A T T
MUTATED DNA	T A C	T A T	G C C	A T A*	A C C	A T T
mRNA						
amino acid sequence						

For each of the mutations above, record the new mRNA sequence and type in the mRNA codons one by one. Record the sequence in the polypeptide chain.

- What, if any, has been the effect of each DNA mutation?
- Is every change in DNA expressed as a change in the amino acid sequence of a polypeptide?

Additional activity

Each of the mutations above is an example of a base substitution in DNA.

Mutations can also occur when DNA is changed in other ways:

e.g., by base addition when an extra base is inserted into a DNA base sequence.

e.g., by base deletion when a base is removed from a DNA sequence.

These mutations are known as frame shifts.

Use Gene Machine to explore the effects of a base addition to DNA and a base deletion from DNA in terms of the change in the polypeptide chain produced by the DNA.

Activity A5: <u>IDENTIFYING A MESSENGER</u>

The hormone vasopressin is a polypeptide, consisting of a chain of nine amino acids. Vasopressin has the amino acid sequence: CYS-TYR-PHE-GLN-ASN-CYS-PRO-ARG-GLY.

Select option 3: "EXPLORE THE GENETIC CODE" and then select the option: "INPUT AN AMINO ACID."

Enter each amino acid in turn. Select one of the mRNA codons which governs the addition of that amino acid to a polypeptide chain. Record your results in the table below:

amino acid	CYS	TYR	PHE	GLN	ASN	CYS	PRO	ARG	GLY
mRNA CODON									

Is your final mRNA base sequence identical with that of other students?

If not, why are the sequences different? What does this show about the nature of the genetic code?

What is meant by the statement: "The genetic code is non-overlapping"?

TABLE 1

The Twenty Amino Acids Common in Proteins

Nonpolar hydrophobic R groups	
Alanine	ALA
Isoleucine	ILE
Leucine	LEU
Methionine	MET*
Phenylalanine	PHE
Proline	PRO
Tryptophan	TRP
Valine	VAL
Polar but uncharged R groups	
Asparagine	ASN
Cysteine	CYS
Glutamine	GLN
Glycine	GLY
Serine	SER
Threonine	THR
Tyrosine	TYR
Negatively charged R groups	
Aspartic acid	ASP
Glutamic acid	GLU
Positively charged R groups	
Arginine	ARG
Histidine	HIS
Lysine	LYS

* The amino acid, MET, also occurs in a modified form known as formylmethionine (F-MET).

APPENDIX

TUTORIAL WORKSHEET

1. DNA and RNA are made up of building blocks called _____.

2. Each nucleotide consises of _____

   + _____

   + _____

3. a) The sugar in RNA is _____.

   b) The sugar in DNA is _____.

4. True or false:

   a) DNA and RNA differ in their sugar component (   )

   b) DNA and RNA differ in the number of strands in their normal structure (   )

5. a) DNA is a _____ -stranded molecule.

   b) RNA is a _____ -stranded molecule.

6. The base pairs found in DNA are:

| G | ≡ | ? |

| A | — | ? |

| C | ≡ | ? |

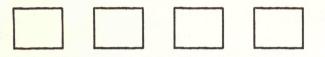| T | — | ? |

7.   The bases found in RNA are

    □   □   □   □

8.   The bases found in DNA and RNA are commonly denoted by a single letter (e.g., G, A).

    Complete the table below showing the letter abbreviation for each base, the name in full, and where it is found.

	ABBREVIATION	BASE	FOUND IN
e.g.	A	adenine	DNA & RNA

Additional activities

*   The bases guanine (G) and adenine (A) are purines, and the bases cytosine (C), thymine (T), and uracil (U) are smaller pyrimidine molecules. Refer to a textbook and draw the molecular structure of one the larger purine bases and one the smaller pyrimidine bases.

*   The G-C base pairs are shown with three links between the bases while the A-T pairs are shown with two links. What is the significance of this?

7. The bases found in RNA are

8. The bases found in DNA and RNA are commonly denoted by a single letter (e.g. G, A).

Complete the table below showing the letter abbreviation for each base, the name in full, and where it is found.

ABBREVIATION	BASE	FOUND IN
A	adenine	DNA & RNA

Additional activities

The bases guanine (G) and adenine (A) are purines, and the bases cytosine (C), thymine (T), and uracil (U) are smaller pyrimidine molecules. Refer to a text-book and draw the molecular structure of one the larger purine bases and one the smaller pyrimidine bases.

The G-C base pairs are shown with three links between the bases while the A-T pairs are shown with two links. What is the significance of this?