Protein Synthesis

Translation of the Genetic Code

1

Review of DNA Replication

DNA is a double stranded molecule that does not leave the nucleus. DNA replication occurs exclusively in the nucleus.

With several enzymes, 1 double stranded DNA molecule will turn into 2 double stranded molecules that are identical to each other.

A bonds with T and C bonds with G.

2

Protein Synthesis

Sequences of bases in the DNA called genes can code for the production of proteins.

The process starts in the nucleus and is completed at a ribosome in the cytoplasm.

Proteins may be further modified in the Golgi Apparatus to be incorporated in the cell membrane or exported from the cell.

3

Proteins are composed of long chains of amino acids called polypeptides. Proteins can be enzymes, hormones, receptors, structural and catalysts.

- control virtually every reaction
- providing structure
- serving as signals to other cells.

The protein cannot function properly unless it folds in the proper orientation.

4

Instructions for the sequence of amino acids are encoded in DNA (genes) located in the nucleus.

The DNA contains the instructions but several steps must occur before you can build a polypeptide chain and that chain can function.

5

RNA Polymerase II

The DNA is in the nucleus and does not leave. First, the specific gene in the DNA will code for a strand of mRNA that is able to leave the nucleus. The enzyme RNA Polymerase II is needed for this process which is called TRANSCRIPTION.

6

Transcription

As you recall, in DNA, the base A will bond with T and the base G will bond with C.

In transcription to mRNA, A will bond with U, and the base T will bond with A, and the base G will bond with C and C with G.

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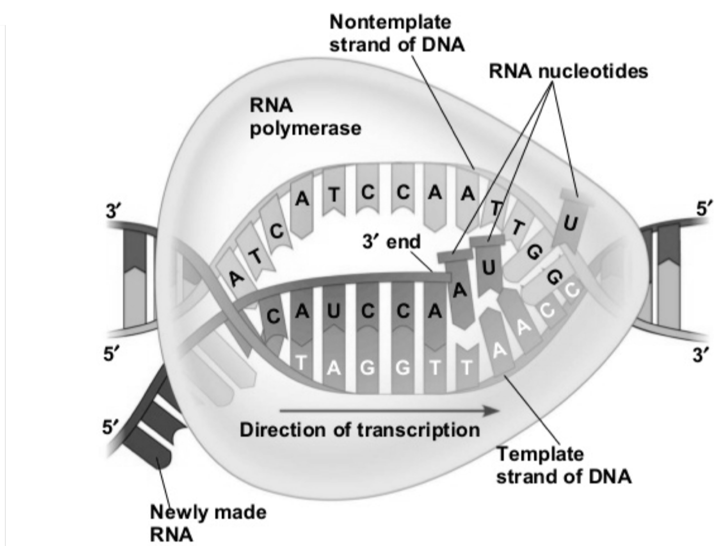

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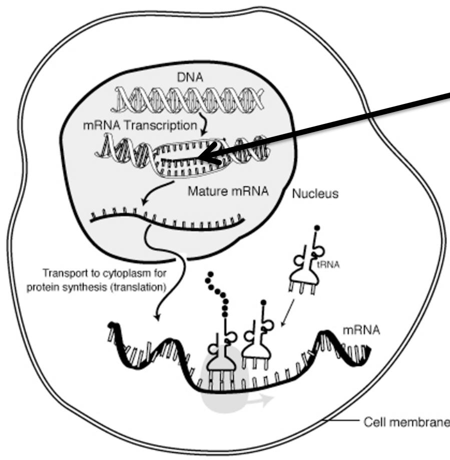

RNA Polymerase II is the enzyme necessary to form the mRNA molecule from the DNA in the nucleus.

The mRNA will move out of the nucleus into the cytoplasm, then on to a ribosome.

9

DNA Replication vs Transcription

DNA

Transcription

10

mRNA

mRNA is a single strand of nucleotides with a phosphate group, a ribose sugar and the bases A, U, G and C.

The mRNA can leave the nucleus and will join a ribosome in the cytoplasm and begin to build a polypeptide chain.

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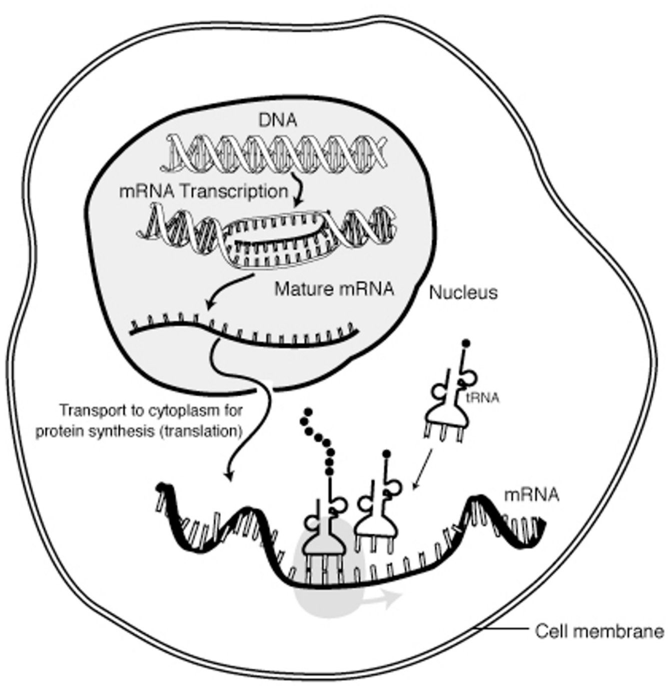

12

Let's Practice-TRANSCRIPTION

DNA → mRNA

T A C T T G C C C G G C A T T

_____ _____ _____ _____ _____

How do you know the top strand is DNA and not mRNA?

13

Codons

Each 3-base sequence of nucleotides transcribed from DNA to mRNA is called a codon. Codons are only found on mRNA. Each codon (3 nucleotides) will code for a 1 specific amino acid. You can determine which specific amino acid is coded for by using the Universal Genetic Code.

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Codon U C A

Amino Acid _______

Codon C U C

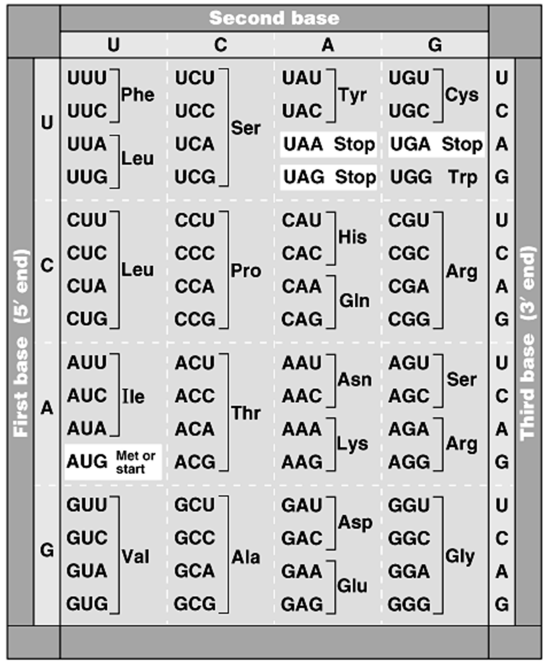
Amino Acid _______

Codon A U G

Amino Acid _______

Codon U G A

Amino Acid _______



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Beginning and Ending

Each polypeptide must begin with a START codon and end with one of three STOP codons.

What is the start codon?

What are the three stop codons?

16

Building the Polypeptide

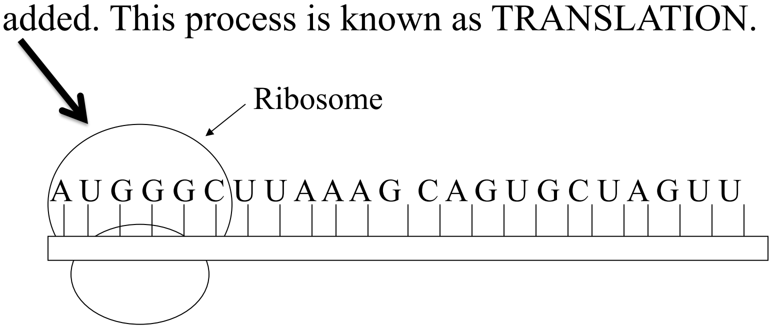
T A C T T G C C C G G C A T T

A U G A A C G G G C C G U A A

_____ _____ _____ _____ _____

17

In order to start making a protein from an mRNA strand, the strand must attach to a ribosome in the cytoplasm. This occurs at the START codon (AUG). The ribosome will move with energy provided by ATP. In reality, the ribosome covers two codons and will move down the length of the mRNA strand as soon as an amino acid is added. This process is known as TRANSLATION.



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More Practice

mRNA

A U G G G C U U A A A G C A G U G C U A G

______ ______ ______ ______ ______ ______ ______

Polypeptide
Chain

19

Where do the amino acids come from and how are they brought to the ribosome?

20

Amino acids are in the cytoplasm and come from the food we eat as well as proteins recycled inside the cells.

tRNA brings the amino acids to the ribosome.

21

Once the ribosome attaches, tRNA molecules can use the instructions on the mRNA to go and retrieve the proper amino acid. The ribosome will move along until it is instructed to drop off. That will happen at one of three STOP codons. At this point the ribosome will drop off. The resulting amino acid chain will fold and form a functioning protein.

22

Translation

The amino acids in the cytoplasm must be brought to the ribosome. This is done with a molecule of tRNA (transfer RNA). The tRNA has a specific region called the anti-codon that will form a temporary bond with the mRNA. It will then release the amino acid it is carrying and move back to the cytoplasm.

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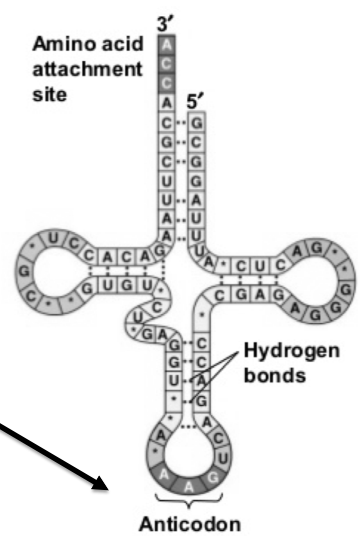
tRNA

tRNA has an area for the amino acid to bind as well as an area to bond with the mRNA (anticodon)



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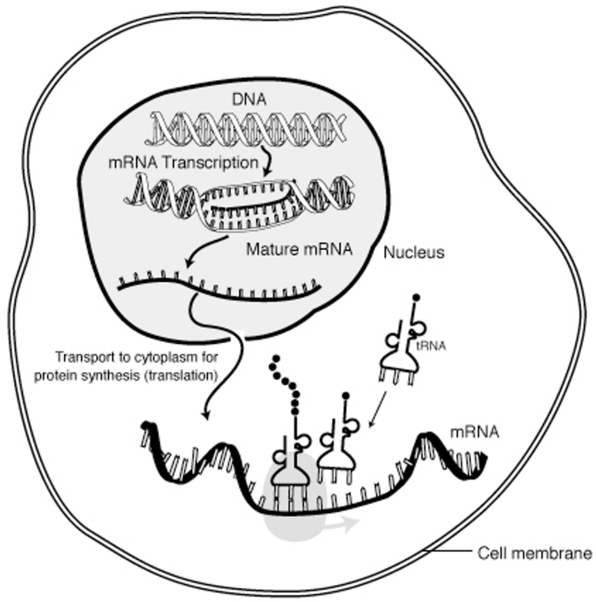
CIRCLE a codon and SQUARE an anticodon.



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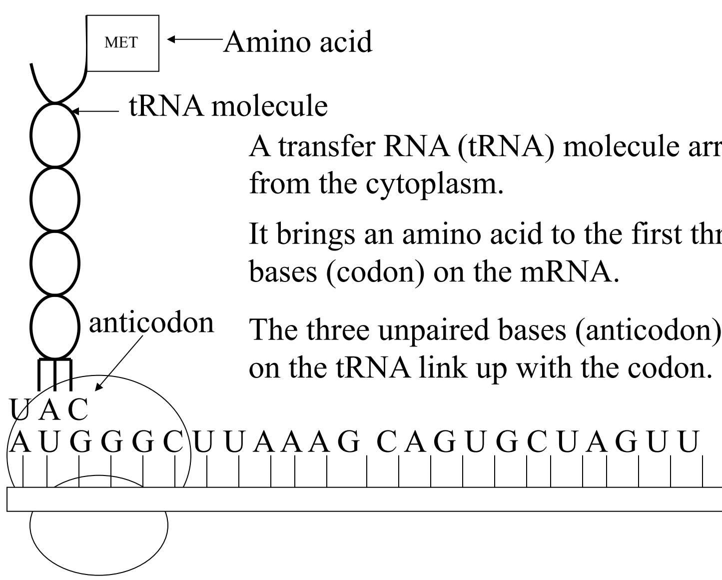
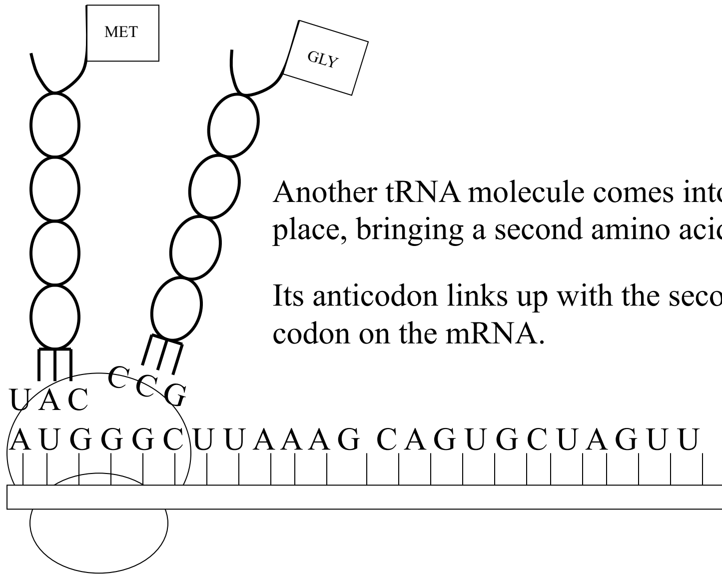


A transfer RNA (tRNA) molecule arrives from the cytoplasm.

It brings an amino acid to the first three bases (codon) on the mRNA.

The three unpaired bases (anticodon) on the tRNA link up with the codon.

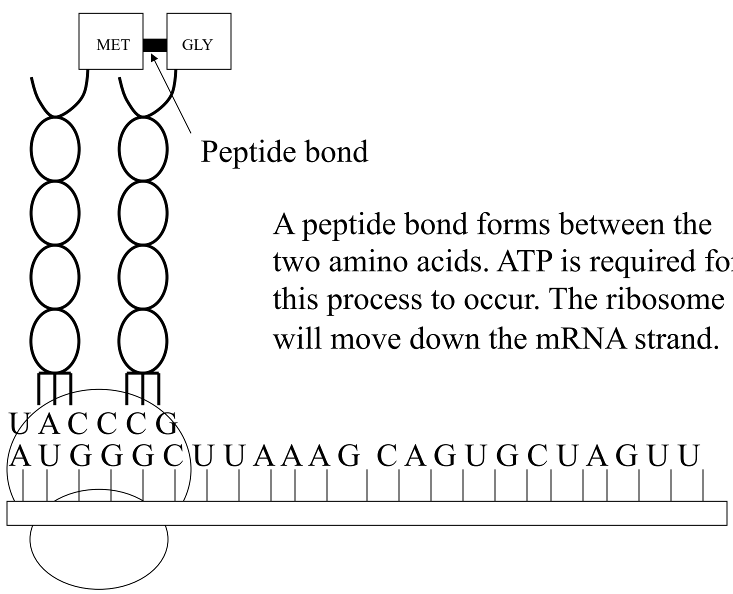
26



Another tRNA molecule comes into place, bringing a second amino acid.

Its anticodon links up with the second codon on the mRNA.

27



A peptide bond forms between the two amino acids. ATP is required for this process to occur. The ribosome will move down the mRNA strand.

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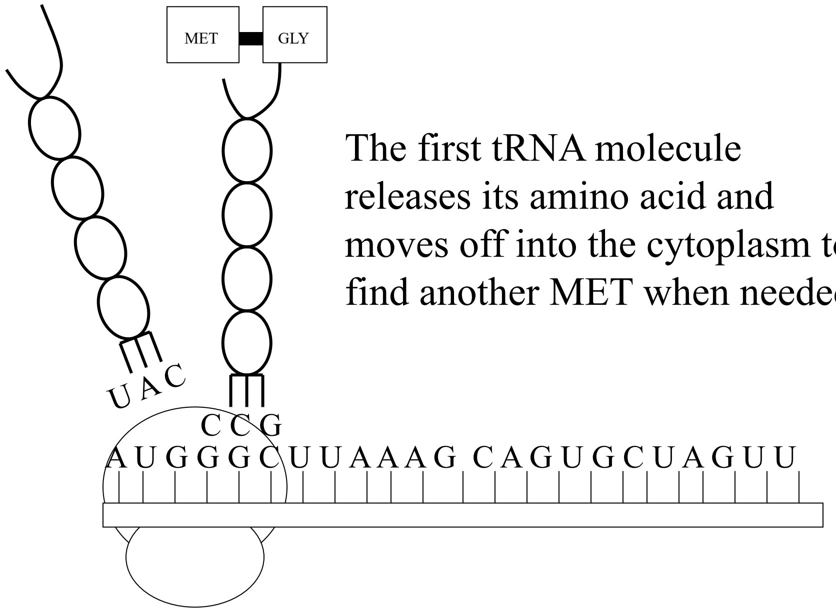

The first tRNA molecule releases its amino acid and moves off into the cytoplasm to find another MET when needed.

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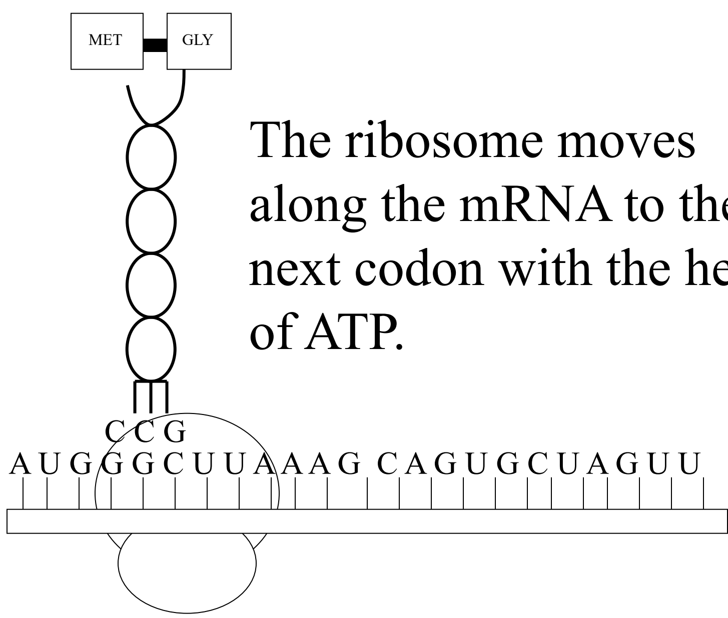
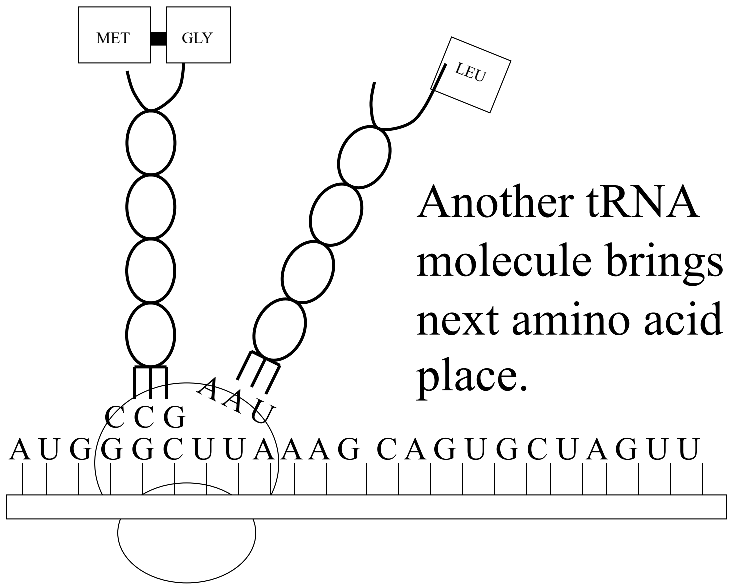


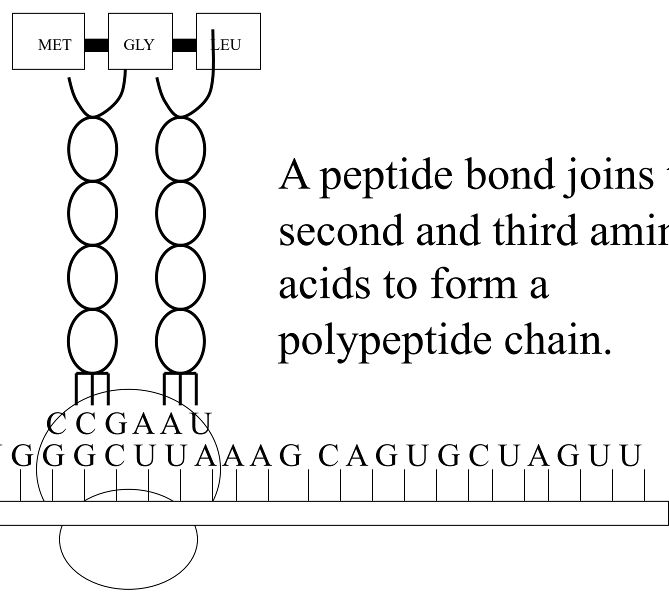
The ribosome moves along the mRNA to the next codon with the help of ATP.

30



Another tRNA molecule brings the next amino acid into place.

31



A peptide bond joins the second and third amino acids to form a polypeptide chain.

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Peptide Bonds

There will always be one less peptide bond that there will be amino acids in a polypeptide chain.

If the chain has 47 amino acids, there will be 46 peptide bonds.

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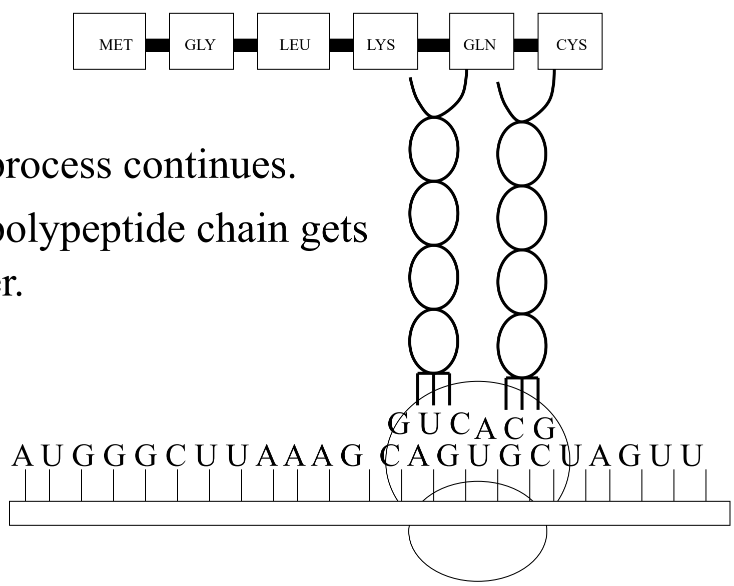
The process continues.

The polypeptide chain gets longer.



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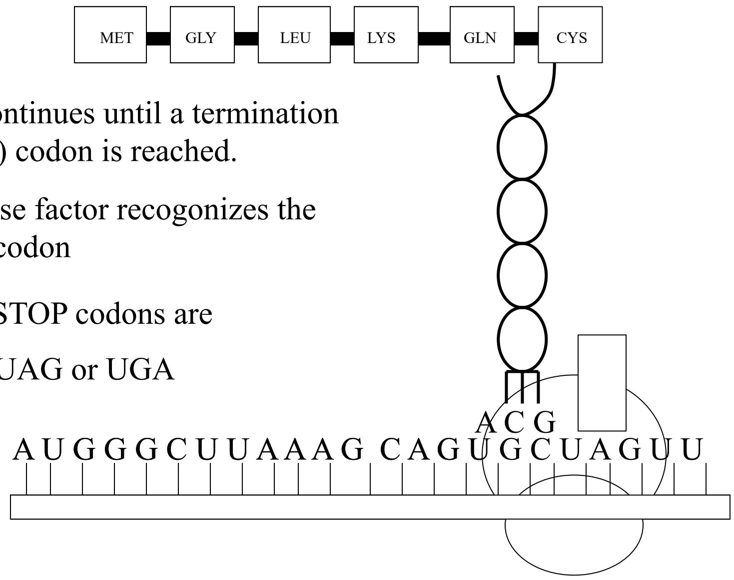
This continues until a termination (STOP) codon is reached.

A release factor recogonizes the STOP codon

Three STOP codons are

UAA, UAG or UGA



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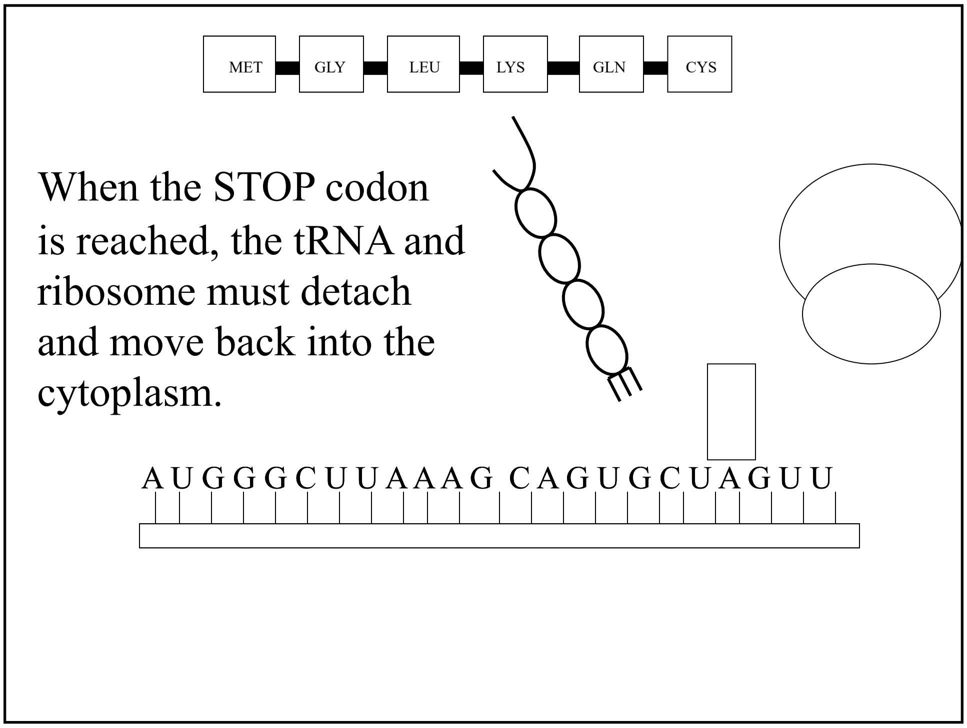
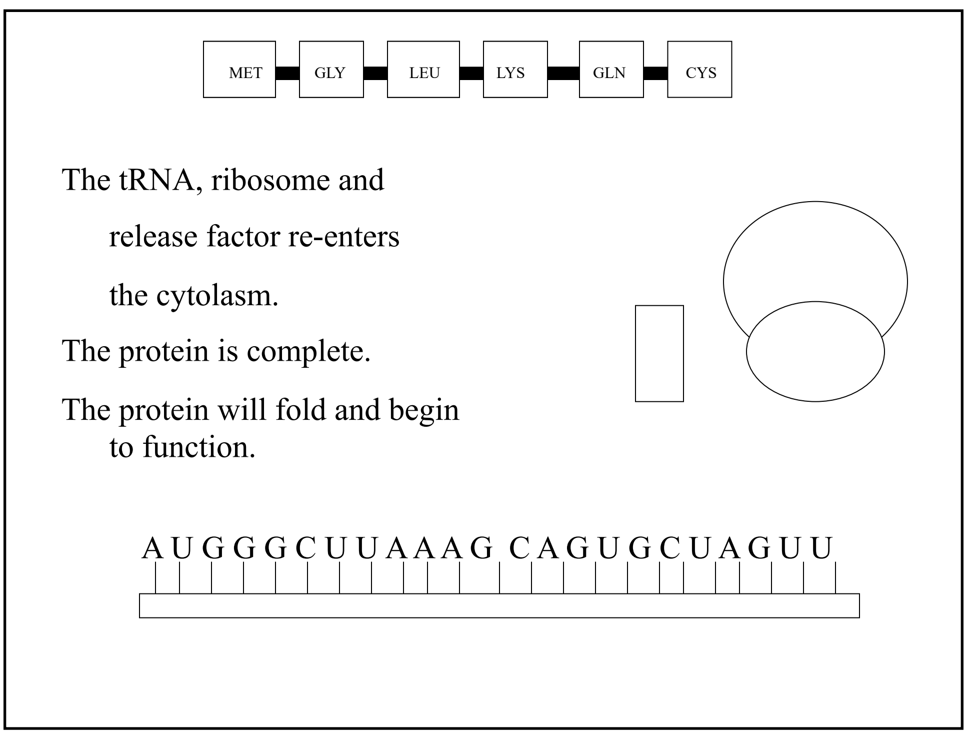
First base (5′ end)	Second base: U	Second base: C	Second base: A	Second base: G	Third base (3′ end)
U	UUU Phe	UCU Ser	UAU Tyr	UGU Cys	U
U	UUC Phe	UCC Ser	UAC Tyr	UGC Cys	C
U	UUA Leu	UCA Ser	UAA Stop	UGA Stop	A
U	UUG Leu	UCG Ser	UAG Stop	UGG Trp	G
C	CUU Leu	CCU Pro	CAU His	CGU Arg	U
C	CUC Leu	CCC Pro	CAC His	CGC Arg	C
C	CUA Leu	CCA Pro	CAA Gln	CGA Arg	A
C	CUG Leu	CCG Pro	CAG Gln	CGG Arg	G
A	AUU Ile	ACU Thr	AAU Asn	AGU Ser	U
A	AUC Ile	ACC Thr	AAC Asn	AGC Ser	C
A	AUA Ile	ACA Thr	AAA Lys	AGA Arg	A
A	AUG Met or start	ACG Thr	AAG Lys	AGG Arg	G
G	GUU Val	GCU Ala	GAU Asp	GGU Gly	U
G	GUC Val	GCC Ala	GAC Asp	GGC Gly	C
G	GUA Val	GCA Ala	GAA Glu	GGA Gly	A
G	GUG Val	GCG Ala	GAG Glu	GGG Gly	G

36


MET GLY LEU LYS GLN CYS
When the STOP codon is reached, the tRNA and ribosome must detach and move back into the cytoplasm.
A U G G G C U U A A A G C A G U G C U A G U U

37


MET GLY LEU LYS GLN CYS
The tRNA, ribosome and release factor re-enters the cytolasm.
The protein is complete.
The protein will fold and begin to function.
A U G G G C U U A A A G C A G U G C U A G U U

38

Folding Patterns/Structures

Primary-The order of amino acids as determined by the DNA

Secondary-Polypeptide chains begin to fold with hydrogen bond interactions forming alpha helices and beta pleated sheets.

Tertiary-More complex folding patterns that involve specific characteristics of the R group including polarity and affinity for water.

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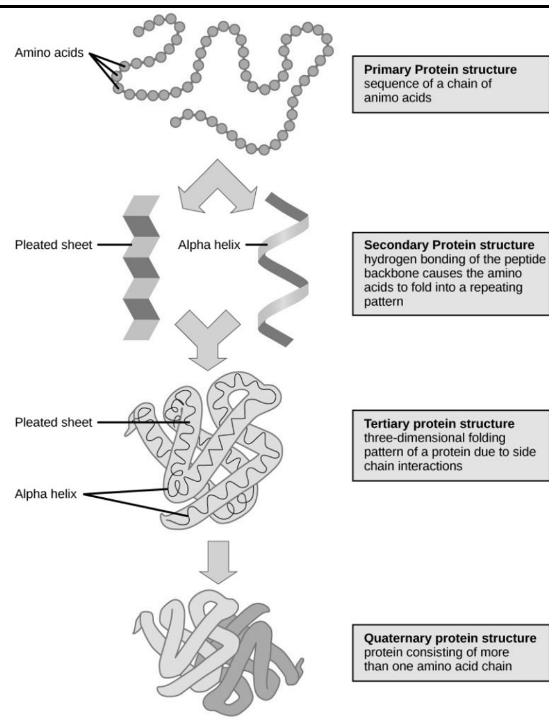

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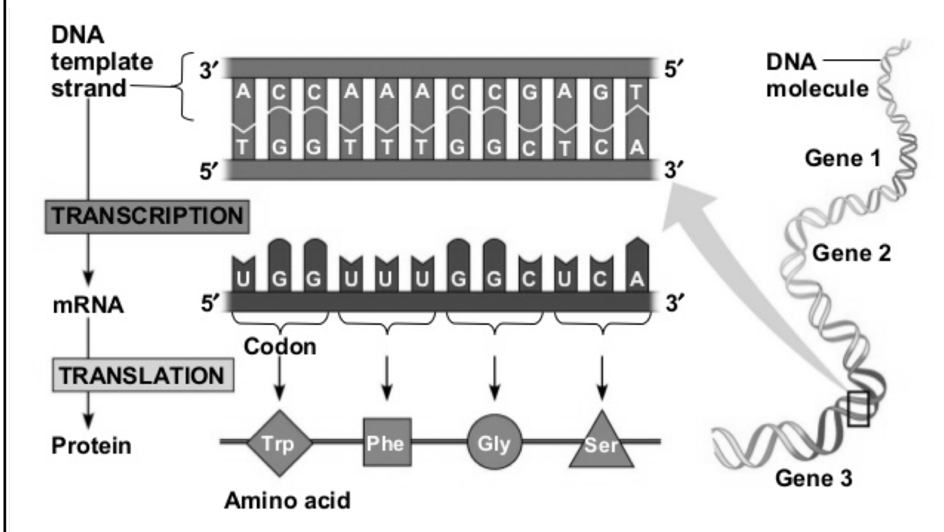

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Forming a Peptide Bond

Amino acids all have the same basic structure. The only difference is the functional R group.

The amino group of one amino acid will form a bond with the carboxyl group of a different amino acid. This will release a water molecule in a process known as dehydration synthesis.

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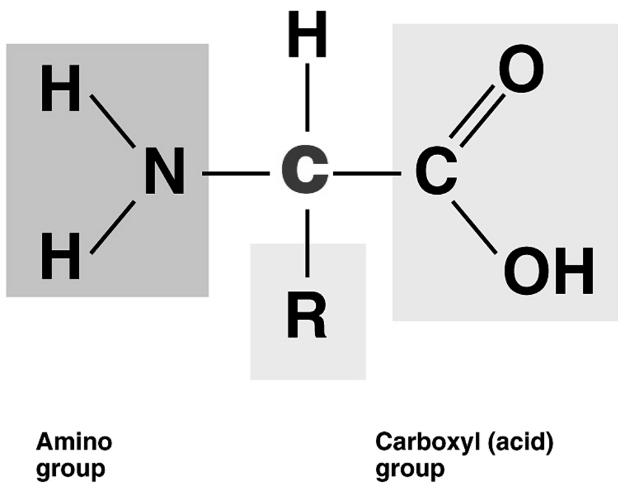
Basic Amino Acid



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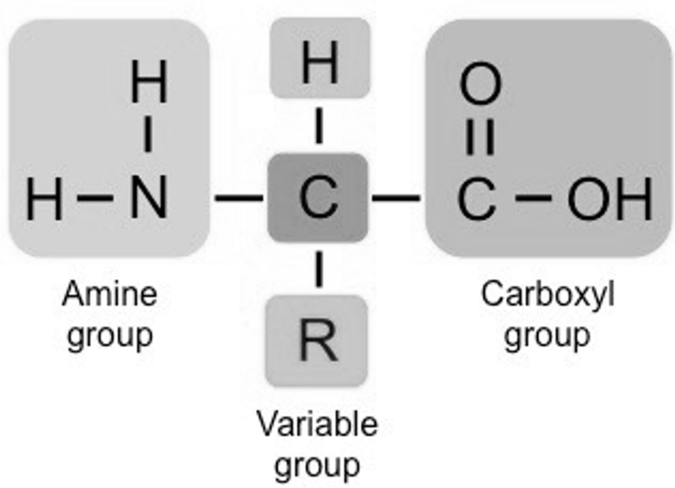

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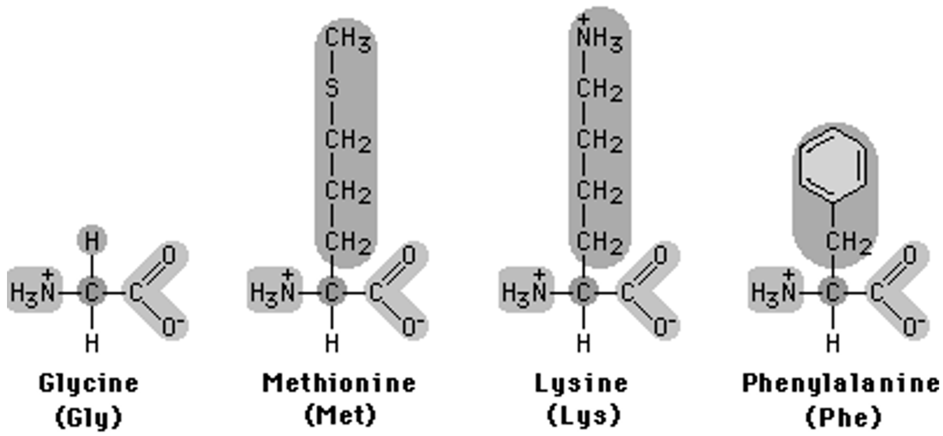
The "R" Group



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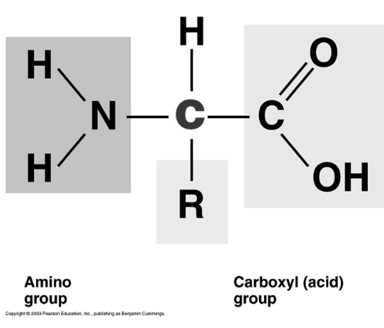
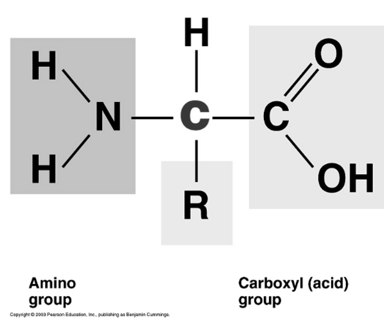
The Peptide Bond





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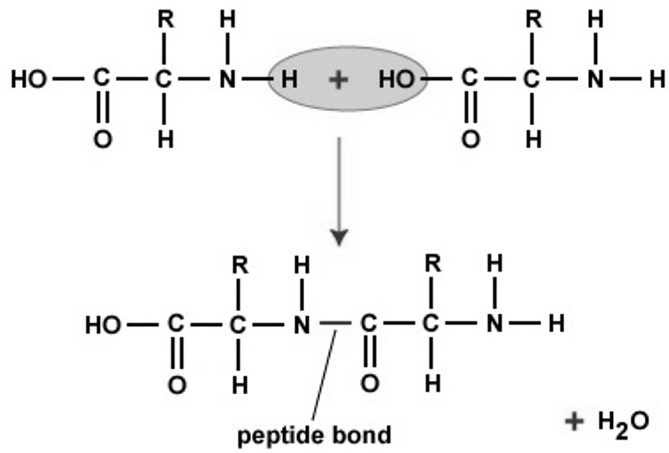
Bye-Bye Water
Dehydration Synthesis



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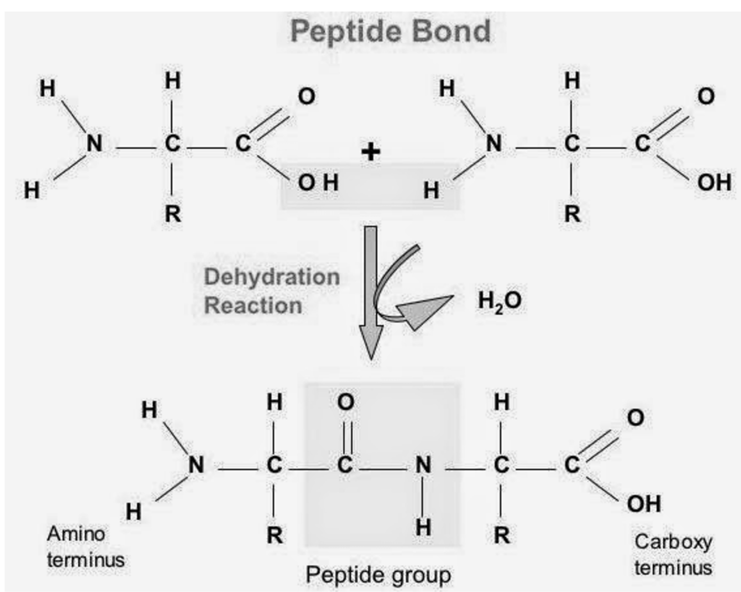

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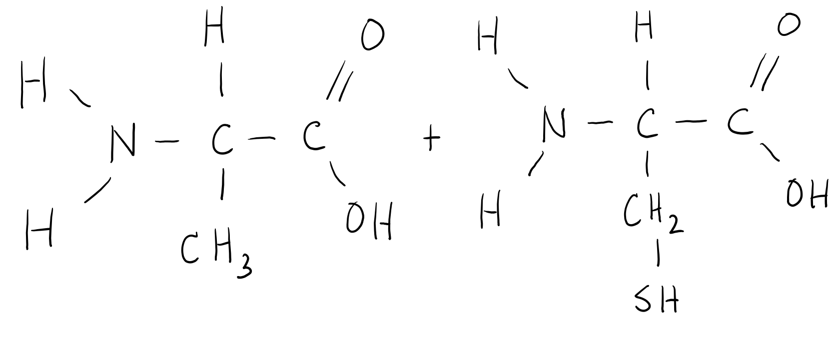
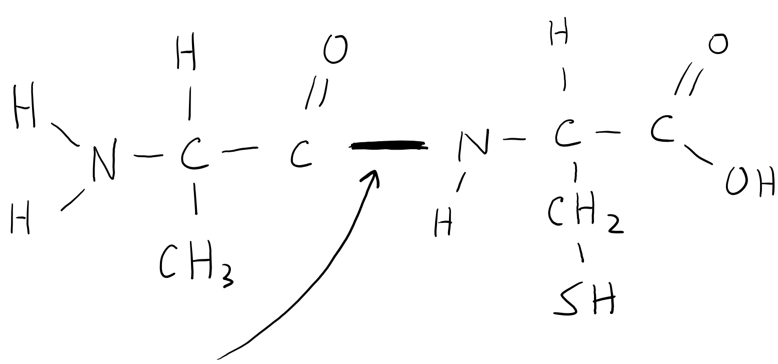
Actual Example



Alanine Cysteine

49



peptide bond

+ H_2O

50

What if there is a problem in the DNA????

51

Mutations

A mutation is a permanent change in the nuclear DNA sequence of a gene. Mutations in a gene's DNA sequence can alter the amino acid sequence of the protein encoded by the gene.

52

Causes of Mutations

Genetic-Inherited the mutation from your parents.

Caused by radiation such as ultra-violet rays from the sun, X-rays or gamma radiation from nuclear material.

Random mistake during DNA replication.

53

Cystic Fibrosis

Cystic fibrosis is an inherited mutation found on chromosome 7 which affects the lungs and digestive system. It results from mutation in a gene responsible for making a protein which is involved in the transport of ions across cell boundaries.

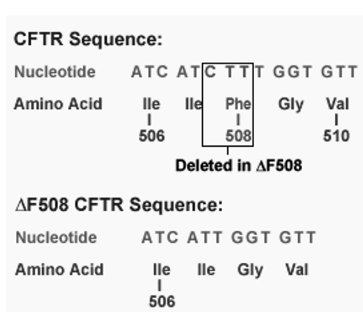
54

The effect is to produce a sticky mucus which clogs the lungs and can lead to serious infection. A similar sticky mucus also blocks the pancreas (a part of the digestive system) which provides enzymes for breaking down food. This gets in the way of the processes which convert the food into molecules which can be absorbed by the body.

55

How it Happens

The phenylalanine (Phe) in red is the amino acid which is missing from the final protein in many sufferers from cystic fibrosis. This occurs at position 508.



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Types of Mutations

Mutations can be classified as a deletion, an insertion or a substitution. In each case there can be little to no effect or the effect can be so severe that the protein does not function properly.

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Base Substitution-NEUTRAL

Here, one base is swapped with another base, but the amino acid did not change/order of amino acids does not change. In this case, the protein function will be unaffected.

Original mRNA	A C C	U A C	G A A	A
Amino Acids	___________	___________	___________	
Mutation 1 mRNA	A C C	U A U	G A A	A
Amino Acids	___________	___________	___________	

58

Base Substitution-MISSENSE

Here, one base is swapped with another base but this time, the amino acid changes to a different amino acid. The actual number of amino acids does change but the order did. In this case, the protein may not function properly or not at all.

Original mRNA	A C C	U A C	G A A	A
Amino Acids	___________	___________	___________	
Mutation 2 mRNA	A C C	A A C	G A A	A
Amino Acids	___________	___________	___________	

59

Base Substitution-NONSENSE

Here, one base is swapped with another base and resulting the amino acid turning into a STOP codon. In this case, the protein will be too short and not function at all.

Original mRNA	A C C	U A C	G A A	A
Amino Acids	___________	___________	___________	
Mutation 3 mRNA	A C C	U A A	G A A	A
Amino Acids	___________	___________	___________	

60

Deletion

Here one base is removed causing all the remaining bases to shift. This is called a frame shift and the amino acid sequence will be so badly altered that the new protein will not fold correctly and not function.

Original mRNA	A C C	U A C	G A A	A
Amino Acids	________	________	________	
		A		
Mutation 4 mRNA	A C C	U C G	A A A	
Amino Acids	________	________	________	

61

Insertion

Here one base is added causing all the remaining bases to shift. This is called a frame shift and the amino acid sequence will be so badly altered that the new protein will not fold correctly and not function.

Original mRNA	A C C	U A C	G A A	A
Amino Acids	________	________	________	
		U		
Mutation 4 mRNA	A C C	U U A	C G A	A A
Amino Acids	________	________	________	

62

How Can This Affect The Body's Ability to Produce Certain Substances?

Substance A $\xrightarrow{\text{Enzyme R}}$ Substance B $\xrightarrow{\text{Enzyme T}}$ Substance C

If the DNA is altered/mutated, Enzyme T does not function properly and the cell will be unable to create substance C.

63

Use the DNA code to make the mRNA strand. With that you can determine the amino acid chain.

DNA code	T A C	G G C	A C C	T T T	G A T	A A A	A T T
mRNA code							
Amino Acid	______	______	______	______	______	______	______

64

First Example-Use the original strand below and compare it to the example.

	T A C	G G C	A C C	T T T	G A T	A A A	A T T
DNA code	T A C	G G C	A C C	T T T	G A A	T A A	A A T
mRNA code							
Amino Acid	______	______	_______	_______	_______	_______	_______

Insertion   Deletion

Substitution   NONSENSE   MISSENSE   NEUTRAL

Affected   TOO LONG   TOO SHORT

Not Affected

65

Second Example-Use the original strand below and compare it to the example.

	T A C	G G C	A C C	T T T	G A T	A A A	A T T
DNA code	T A C	G G C	A C C	T T C	G A T	A A A	A T T
mRNA code							
Amino Acid	______	______	_______	_______	_______	_______	_______

Insertion   Deletion

Substitution   NONSENSE   MISSENSE   NEUTRAL

Affected   TOO LONG   TOO SHORT

Not Affected

66

Third Example-Use the original strand below and compare it to the example.

	T A C	G G C	A C C	T T T	G A T	A A A	A T T
DNA code	T A C	G G C	A C T	T T T	G A T	A A A	A T T
mRNA code							
Amino Acid	______	______	_______	_______	_______	_______	_______

Insertion Deletion

Substitution NONSENSE MISSENSE NEUTRAL

Affected TOO LONG TOO SHORT

Not Affected

67

Fourth Example-Use the original strand below and compare it to the example.

	T A C	G G C	A C C	T T T	G A T	A A A	A T T
DNA code	T A C	G G C	A C C	T T A	G A T	A A A	A T T
mRNA code							
Amino Acid	______	______	_______	_______	_______	_______	_______

Insertion Deletion

Substitution NONSENSE MISSENSE NEUTRAL

Affected TOO LONG TOO SHORT

Not Affected

68

Fifth Example-Use the original strand below and compare it to the example.

	T A C	G G C	A C C	T T T	G A T	A A A	A T T
DNA code	T A C	G G A	C C T	T T G	A T A	A A A	T T C
mRNA code							
Amino Acid	______	______	_______	_______	_______	_______	_______

Insertion Deletion

Substitution NONSENSE MISSENSE NEUTRAL

Affected TOO LONG TOO SHORT

Not Affected

69