

Protein Biosynthesis: Basic details

Introduction:

Def: Translation is the process in which genetic message carried out by mRNA from DNA is converted into polypeptide chain having a specific sequence of amino acids.

- These polypeptides (more correctly proteins) determine the phenotype.
- structural ~~and~~ protein and functional protein i.e. enzymatic protein both are produced as a result of translation.
- In going from RNA to protein the language is changed from a nucleotide sequence to an amino acid sequence.

Minimum necessary materials:

1. Amino acid (20 types)
2. Ribosomes
3. mRNA
4. tRNA
5. Enzymes
 - (a) Amino acyl-tRNA synthetase.
6. ATP energy source
7. GTP for synthesis of peptide bond.
8. Various inorganic ~~for~~ cations (K^+ , NH^+ , Mg^{++})

Rule of translation:

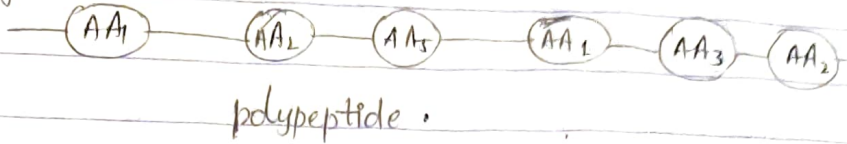
• language of mRNA is written in four letter alphabet i.e. A, U, G, C

eg. 5' - AUCA CCG AAG CUC UGA UUA 3'

codon - add of mol.

- language of protein is written by 20 letter alphabet.

e.g.



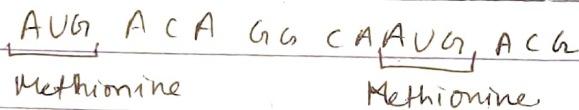
Genetic codes - Term proposed by Crick.

- Combination of 3-bases → Genetic code (codon)
- Specific base sequence of 3-nitrogenous bases form one codon for one amino acid.

Therefore, no. of possible codons = $(4)^3 = 64$

Initiation codon (start signal)

AUG - It act as capital letter in beginning of sentence i.e. translation.



- ✓ In bacteria, in place of AUG, GUG also function as initiation codon.

Normally GUG codes for valine but when it present in start point, it codes for methionine.

Termination codon (stop codon)

UAA

UGA

UAG

- These function as full stop signal. It means that it can stop protein synthesis but will not code for any protein.
- Normally tRNA are responsible for ^{sensing} codons but no any tRNA is capable of sensing them.

so these are c/a non-sense codon.

- Thus total codons, coding for amino acid are $(64-3) = 61$.
- More than one codon can code a single amino acid but not the vice-versa i.e. a single codon can never code more than one amino acid.
- If one amino acid is coded by more than one codon then ^{these} codon only differ at their 3rd base posⁿ. This 3rd base posⁿ of codon is c/a Wobble posⁿ. Wobble posⁿ is less specific for amino acid.
- * Specificity for amino acid lies in 1st two bases of codon.

Degeneracy of codon - The codon which differ only at 3rd base posⁿ i.e. wobble posⁿ.

Non-overlapping type codon -

AUG, ACA, GAC, AUG, AAA, AC.....

- A single base can never be part of two adjacent codon.
- Codons are universal.

Mechanism of translation:

Translation is combination of two major events

- (1) Aminoacylation of tRNA
- (2) stages of polypeptide synthesis.

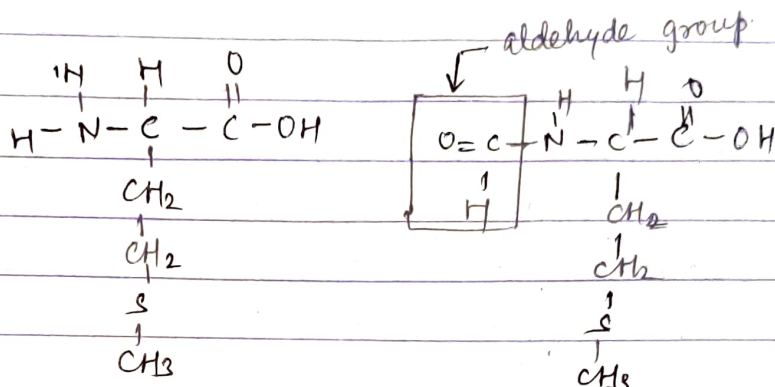
(1) Aminoacylation of tRNA -

- binding of activated amino acid with tRNA.
- Transfer RNA is different for different amino acid as for

~~met~~ methionine - tRNA^{met}
valine - tRNA^{val}
alanine - tRNA^{ala}

- For amino acid methionine, two tRNA found:
 - (a) tRNA^{met}
 - (b) tRNA^{fmet}.

- Methionine (a sulphur containing amino acid) which comes in first of chain, ^{at P-site,} is of a formylated methionine (fmet) and methionine which comes in b/w the chain (base region) is simply called methionine (met). (at A-site)



* fmet starts protein synthesis.

* In prokaryotes fmet is present but not in eukaryotes.

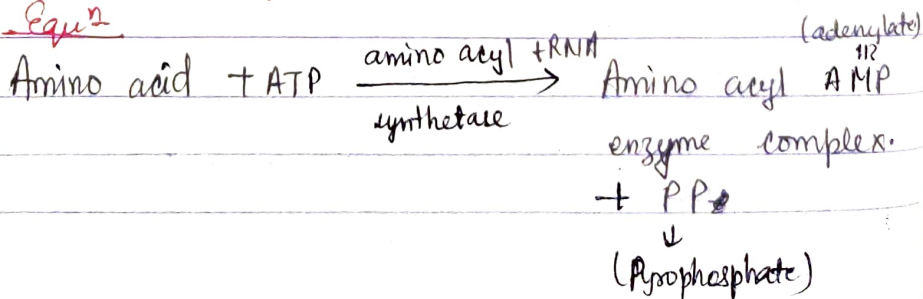
* Formylation of methionine protects its amino group from taking part in peptide bond formation.

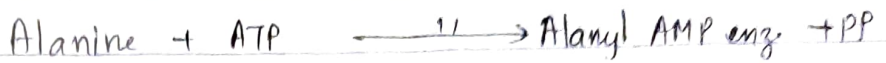
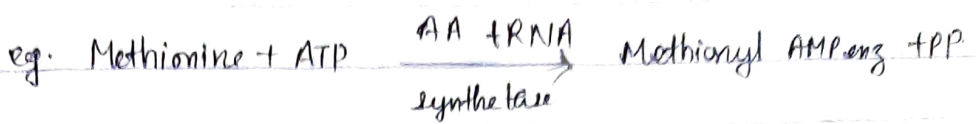
* Possible no. of tRNA = 34.

• Aminoacylation of tRNA is completed in following two steps.

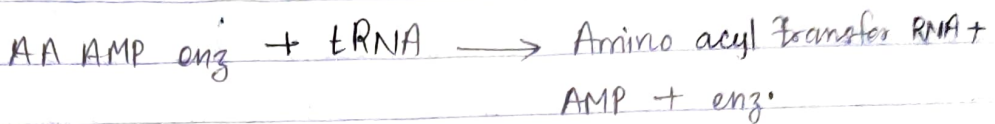
(i) Activation of amino acid -

Equⁿ





(i) Attachment of activated a. acid to tRNA -



→ It is the only step in protein synthesis in which identity of a. acid (i.e. R group) plays role.

- After attachment of with a. acid, tRNA becomes charged and that tRNA which is not attached to a. acid is uncharged.

(2) stages of polypeptide synthesis -

There are 3 stages

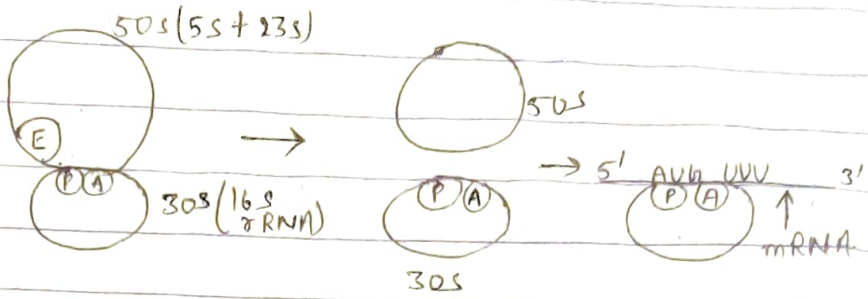
- (i) Initiation
- (ii) Elongation
- (iii) Termination.

(i) Initiation -

- Both subunits of ribosome dissociate.
- There are 3 hypothetical sites in ribosomes.
 - P site - peptidyl site.
 - A site - amino acyl site.
 - E. site - exit site.

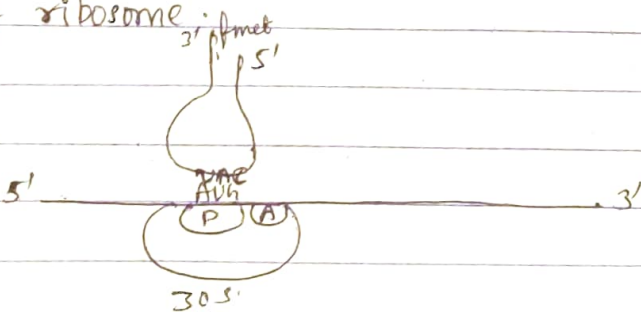
- Properties of P site and A site contributed by both subunits of ribosome but property of E-site

is only due to larger subunit i.e. (50s)



- During binding of tRNA, initiation codon is attached at P-site i.e. AUG.
- mRNA is attached to smaller subunit (30s in case of prokaryote and 40s in case of eukaryotes)
- Correct positioning of mRNA is decided by **purine rich "Shine - Dalgarno repetitive sequence of bases"**. Shine - Dalgarno sequence of bases present towards mRNA before initiation codon.
- Purine rich Shine - Dalgarno sequence of bases are complementary to pyrimidine rich bases towards smaller 3'-end of 16s rRNA present in smaller subunit of ribosome.

- fmet tRNA^{fmet} only, can recognize the P-site of ribosome.



- larger subunit of ribosome join with this

to produce initiation complex.

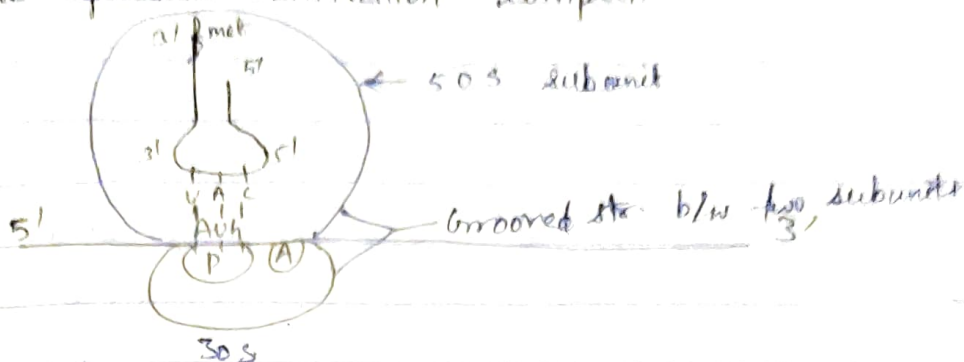


Fig: INITIATION COMPLEX,

• Mainly 3 initiation factors are

IF 1

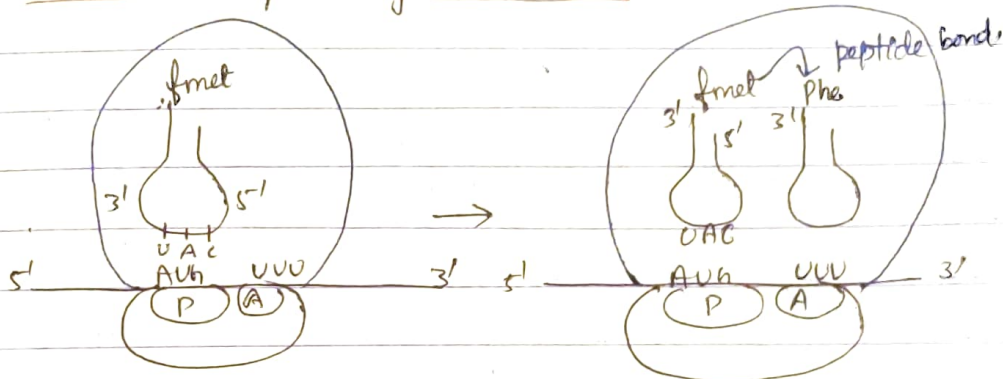
IF 2

IF 3

Here energy molecule needed is GTP.

(ii) Elongation -

(1) Attachment of charged tRNA at A-site.



• tRNA charged with a. acid as per codon of A-site comes and attaches to this site.

(2) Peptide bond formation

• For peptide bond formation, amino acid from tRNA of P-site dissociate and moves on A-site. Thus, peptide bond formation occurred A-site.