# tRNA structure and function

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# tRNA

- t-RNA (transfer RNA) is also named as S-RNA (soluble or supernatant RNA) and adaptor RNA.
- t-RNA is a family of nearly 60 small sized ribonucleic acids.
- 10 – 15% of total cellular RNA is t-RNA.
- t-RNAs are small molecules with about 74 – 95 ribonucleotides.
- Sedimentation constant – 3.8S
- Molecular weight – nearly 25,000 – 30,000 Dalton
- t-RNAs are made up of a single stranded polynucleotide chain

# Unique feature of tRNA

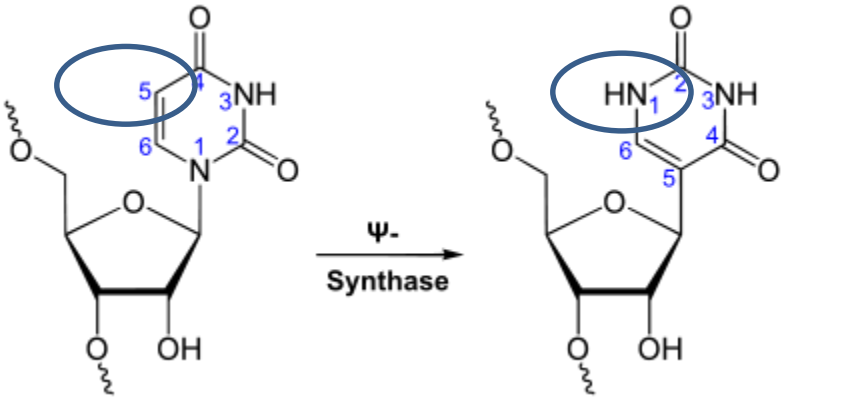
- **Presence of un usual base pairs**

- In addition to usual N-bases (A,U,G,C) tRNA contains number of unusual bases.
- These unusual bases are important as they **protect t-RNA molecules from dehydration by Rnase**, when tRNAs are floating freely in cytoplasm.
- **Protects mainly by methylation**

❖Inosine (I) – Adenine

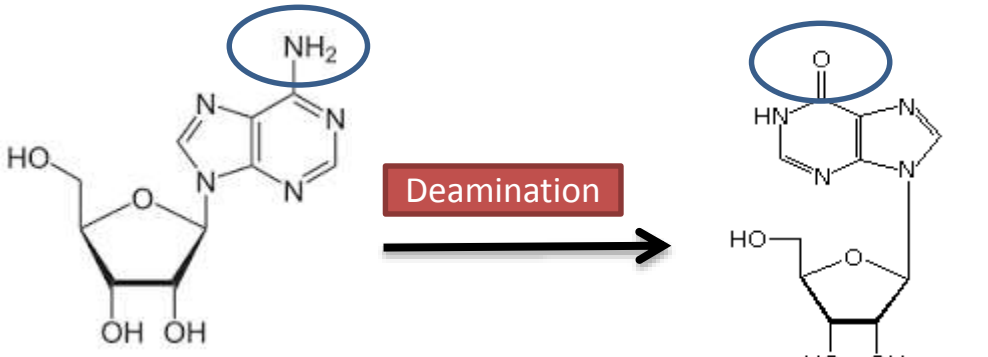
❖Pseudouracil ($\psi$) - Uracil

❖Dihydroxyuridine- urdine



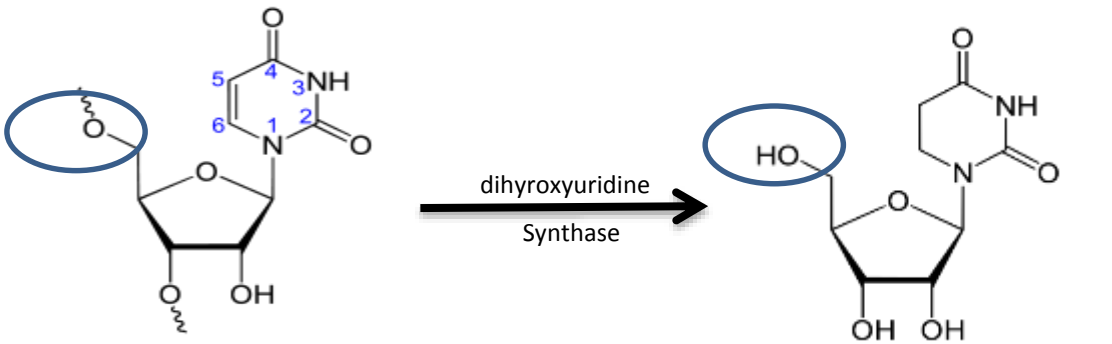

uridine

Psuedouridine




Adenosine

Inosine




uridine

dihyroxyuridine

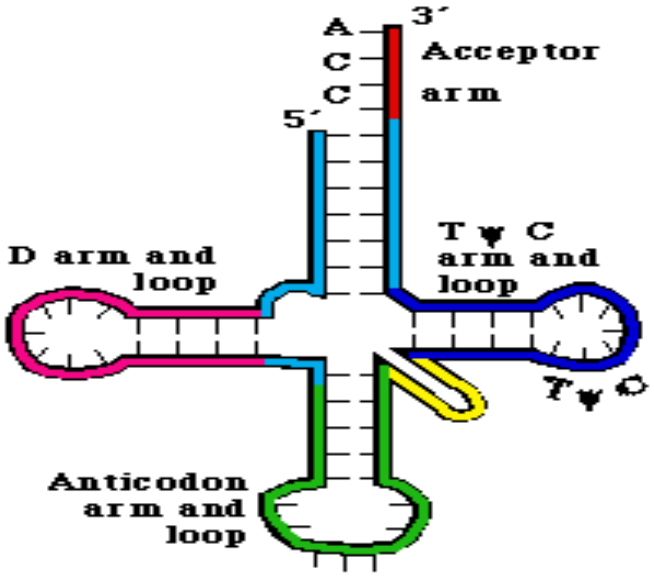
# Structure of tRNA

- **Primary structure-** linear sequence of nucleotides
- **Secondary structure**-Clover leaf model
- **Tertiary structure-** 3-D structure of tRNA , L shape, Helix stacking

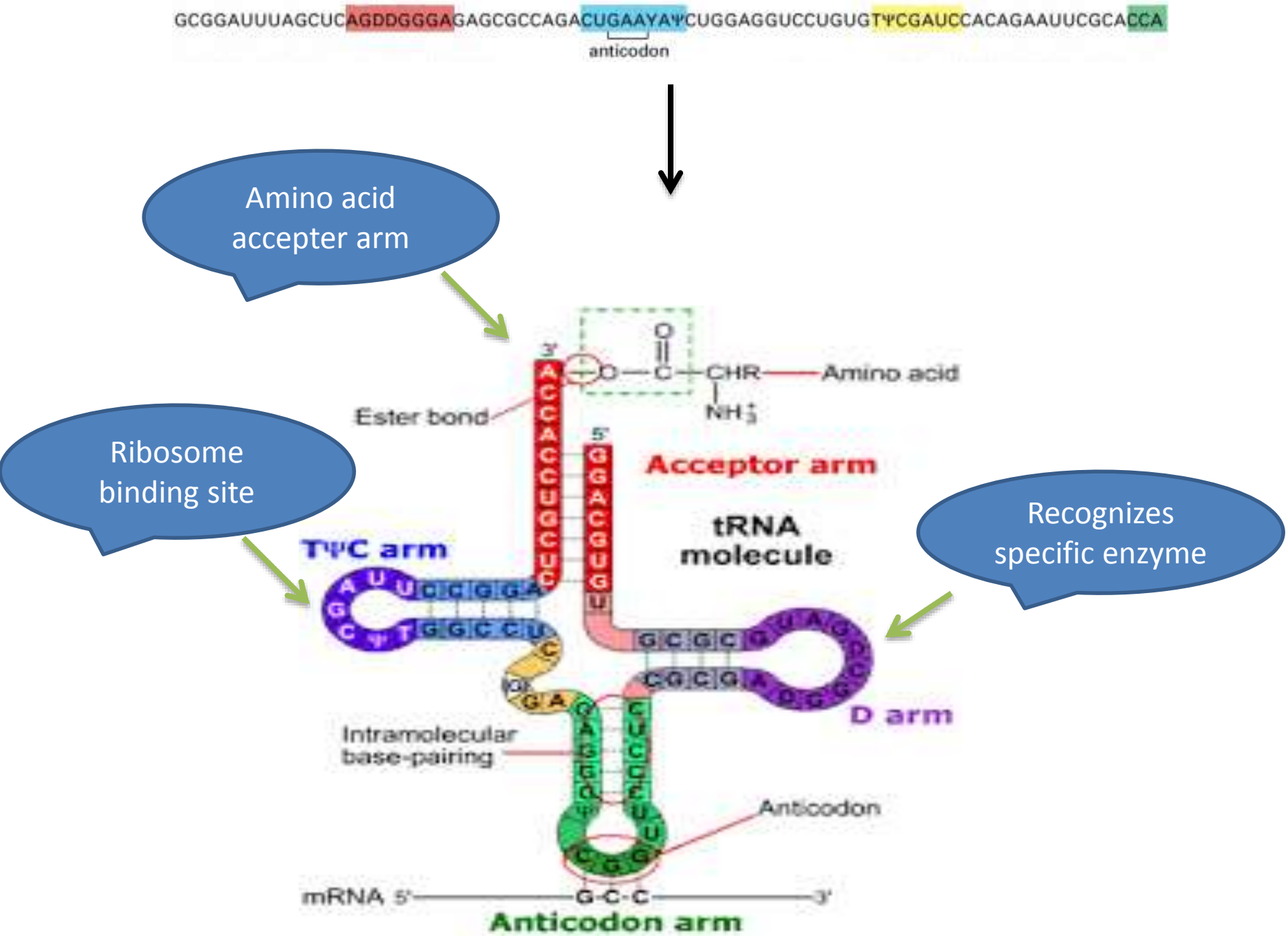
# Primary Structure

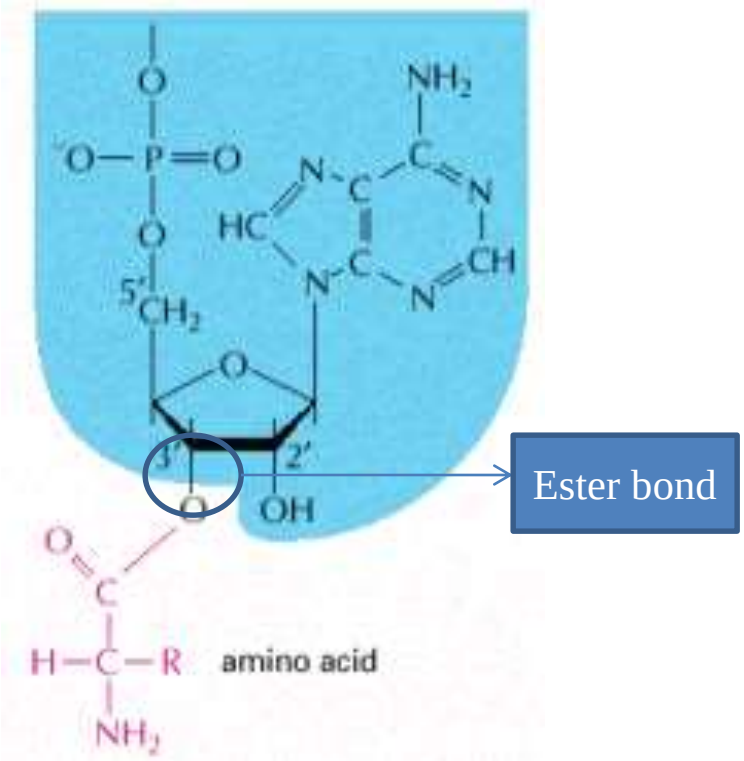

- Linear sequence of nucleotides is 60-90 in nt long but most commonly 76
- Many **modified bases,** sometimes accounting for **20%** of the total bases in any one tRNA molecules
- All of them are created post transcriptionally.

# Secondary structure/ clover leaf model



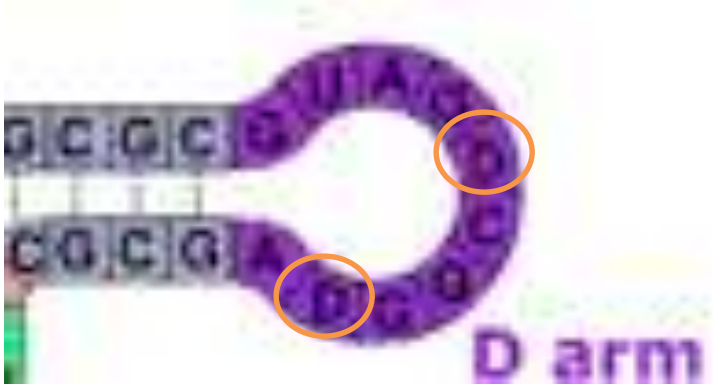

- Robert Holley proposed clover leaf model for the first time in 1968.
- It is a two dimensional description of the t-RNA.


GCGGAUUUAGCUCAGDDGGGAGAGCGCCAGACUGAAYAΨCUGGAGGUCCUGUGTΨCGAUCCACAGAATTCGCACCA
anticodon
Amino acid accepter arm
Ribosome binding site
Recognizes specific enzyme
Amino acid
Ester bond
Acceptor arm
tRNA molecule
TΨC arm
D arm
Intramolecular base-pairing
Anticodon
mRNA 5'
G-C-C
3'
Anticodon arm




**Amino acid acceptor arm**

- Double helical (both 5’ and 3’ ends of tRNA)
- 7 base pairs unpaired
- At 3’ end**, 5’CCA3**’ protrudes with –OH at the tip
- Site for **attachment of amino acid**
- -COOH of specific amino acid joins with –OH of A in CCA to form amino acyl tRNA

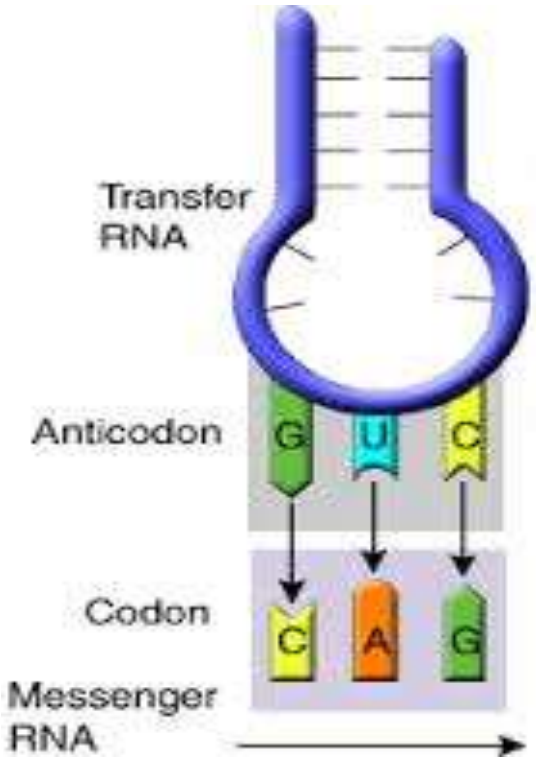



D arm

- **DHU or D arm** – This arm consists of stem and loop with unusual pyrimidine nucleotide **dihydrouracil**.
- 4 bp stem with a loop contain dihydrouridine
- Recognition site for the **specific enzyme aminoacyl-tRNA synthetase** that activate the amino acid
- Play a important role in the stabilization of the tRNA's tertiary structure.


T ψ C arm

- **TψC arm is named for the presence of sequence TψC (thymine – pseudouridine (ψ) – cytosine), where pseudouridine is unusual base.**
- **This arm also consists of stem and loop.**
- **Stem contains 5 base pairs; outermost of these pairs is C-G. Loop contains 7 unpaired nucleotides**
- **This loop contains a ribosome recognition site.**




**Anticodon loop**

- **This arm also contains stem and loop.**
- **Stem consists of 5 base pairs and loop (called as anticodon loop or loop II) contains 7 unpaired nucleotides.**
- **Out of these 7 unpaired nucleotides the middle three form anticodon.**
- **Anticodon recognizes and codon of mRNA and binds to it.**

- The original wobble pairing rules, as proposed by Crick. Watson-Crick base pairs are shown in **bold**, wobble base pairs in *italic*:

- The thermodynamic stability of a wobble base pair is comparable to that of a Watson-Crick base pair

| tRNA 5' anticodon base | mRNA 3' codon base |
|---|---|
| A | **U** |
| C | **G** |
| G | **C** or *U* |
| U | **A** or *G* |
| I | *A* or *C* or *U* |

# Variable arm

- The variable arm has between 3 and 21 nucleotides, depending on which amino acid the tRNA encodes.
- Between anticodon loop and TΨU loop
- This tRNA's variable arm is very short so it looks quite different from the other arms of the molecule.
- May present or absent, it depends on species.
- The length of the variable arm is important in the recognition of the aminoacyl tRNA synthetase for the tRNA.
- Variable arm helps is stability of tRNA
- tRNAs are called class 1 if they lack it, and class 2 if they have it.

# Tertiary structure : 3D structure

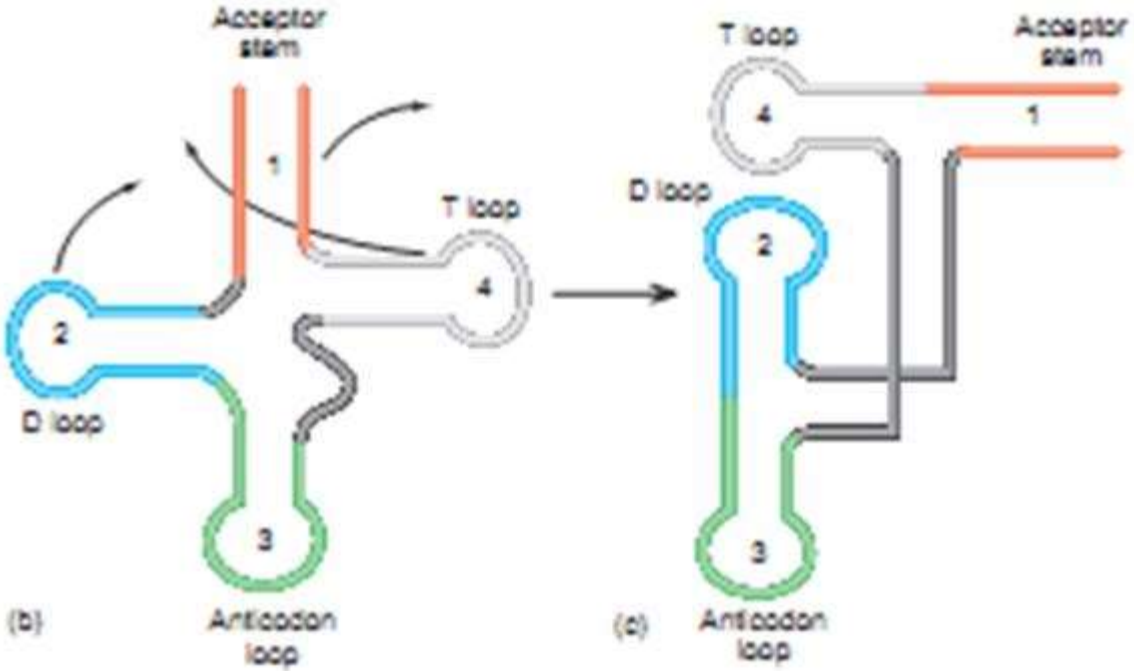
## L-Shaped structure

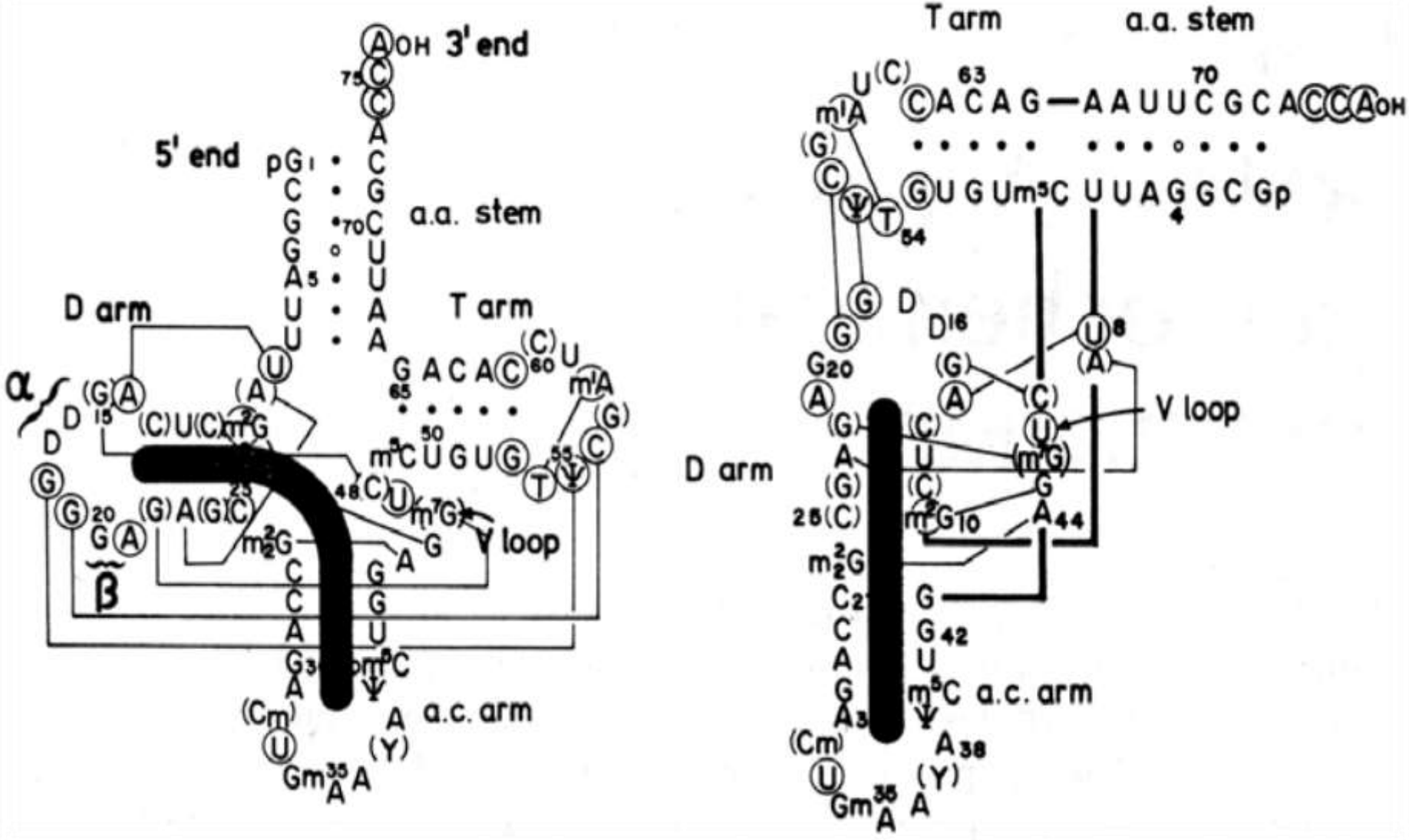
(i) Acceptor stem and ΨU stem form extended helix

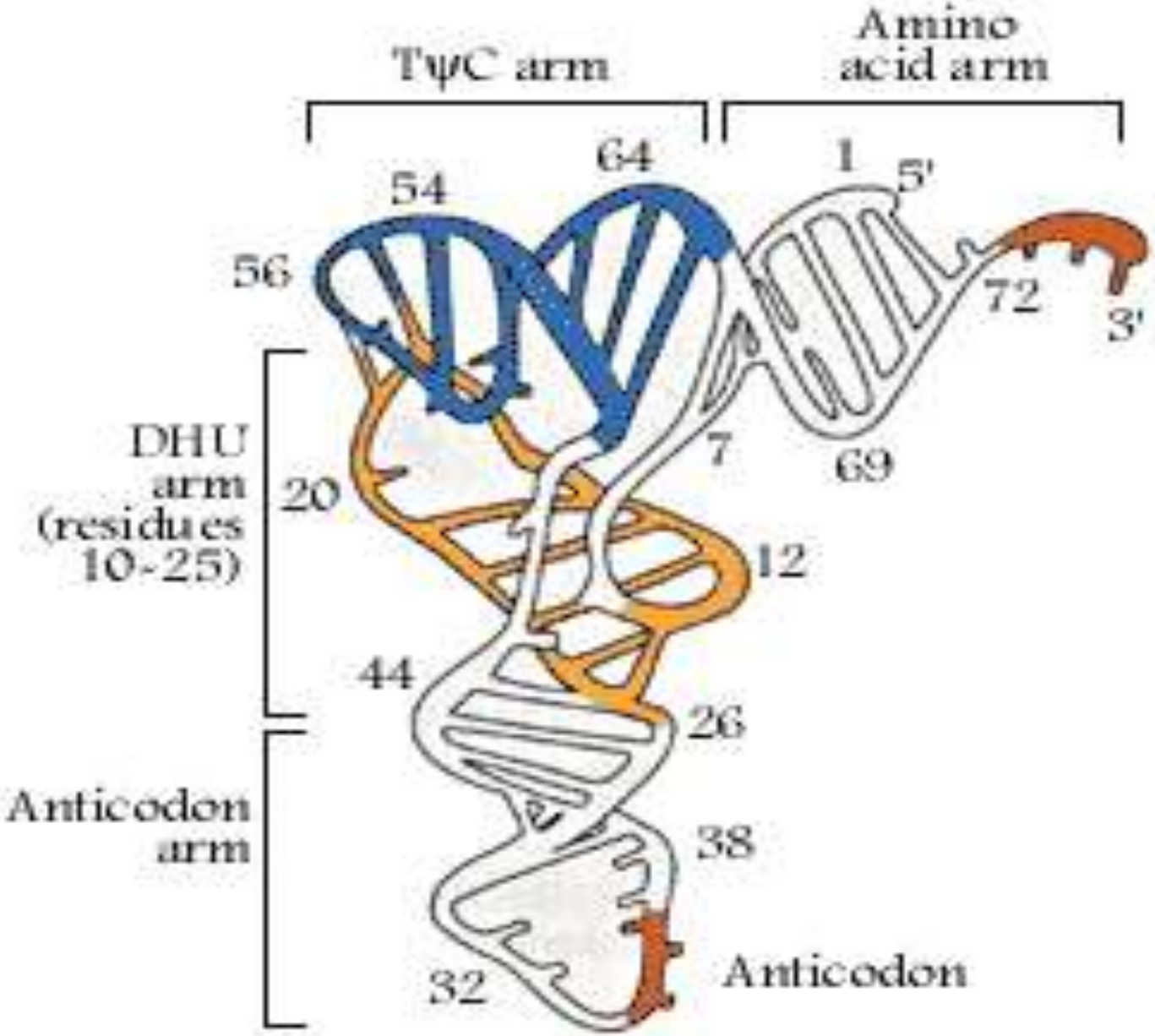
(ii) Anticodon stem and D-loop stem form extended helix

(iii) D-loop and T loop align together

(iv) Both extended helices align at right angle




5' end
3' end
a.a. stem
D arm
T arm
V loop
a.c. arm
α
β
D arm
T arm
a.a. stem
V loop
a.c. arm


TψC arm
Amino acid arm
64
54
1
5'
56
72
3'
DHU arm (residues 10-25)
7
69
20
12
44
26
Anticodon arm
38
32
Anticodon

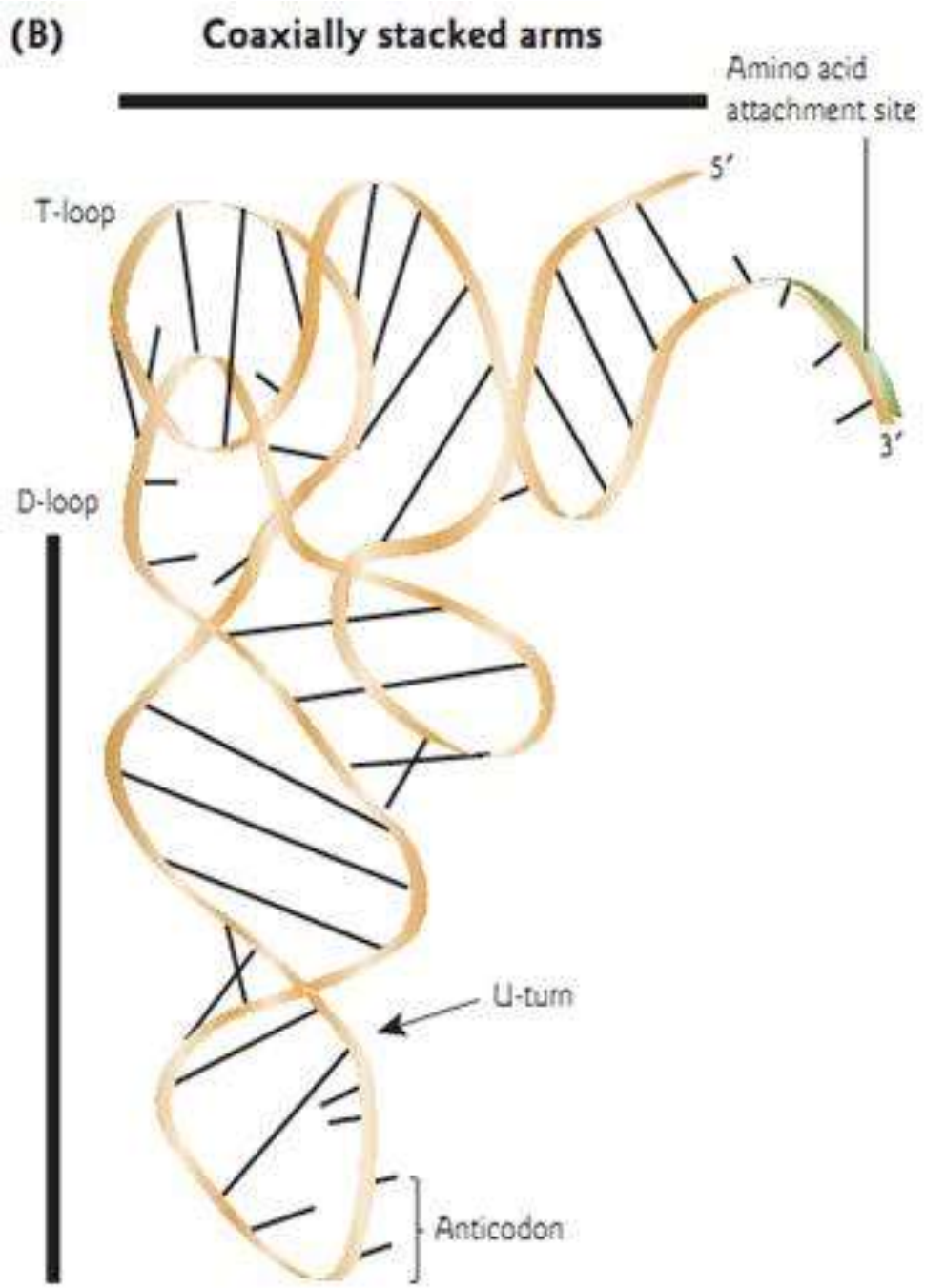
## Stability of L-structure

- Tertiary structure of t-RNA is produced by hydrogen bonding –

✓ Between N-bases

✓ Between N-bases and ribose-phosphate backbone

✓ Between ribose-phosphate backbone

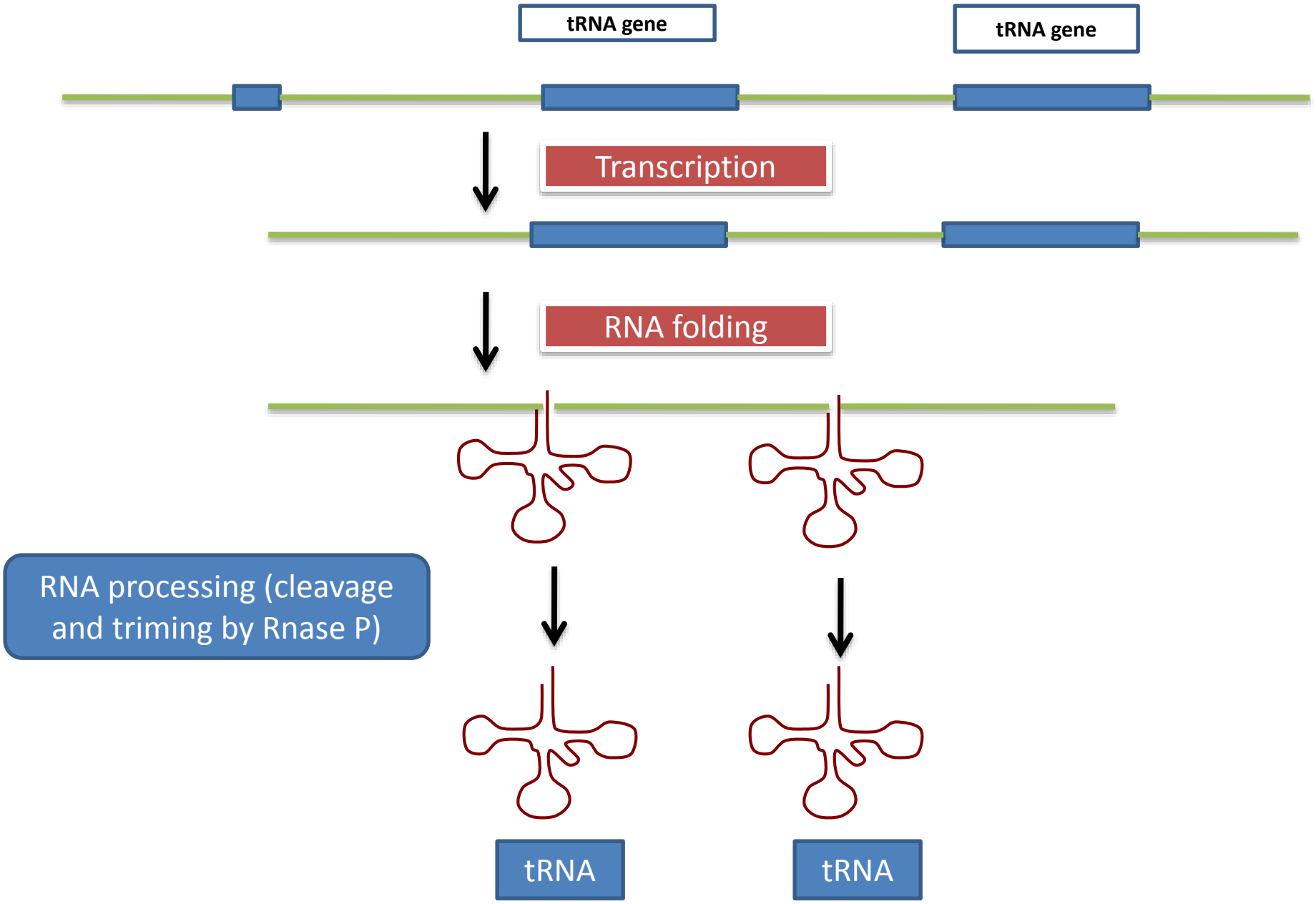


# tRNA processing

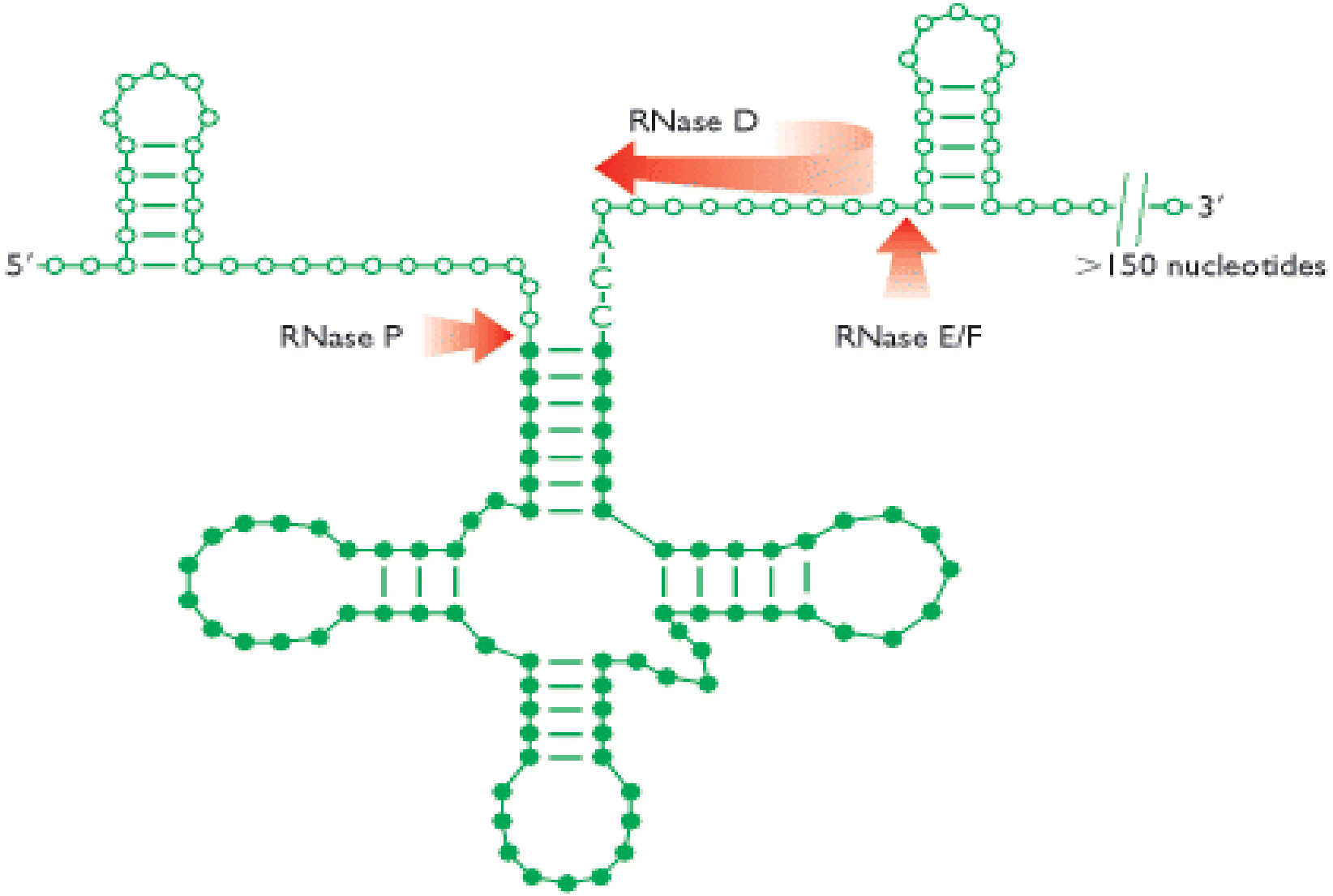
- The rRNA operons of *E coli* contain coding sequences for tRNAs.

- In addition, there are other operons that contain up to seven tRNA genes separated by spacer sequences.

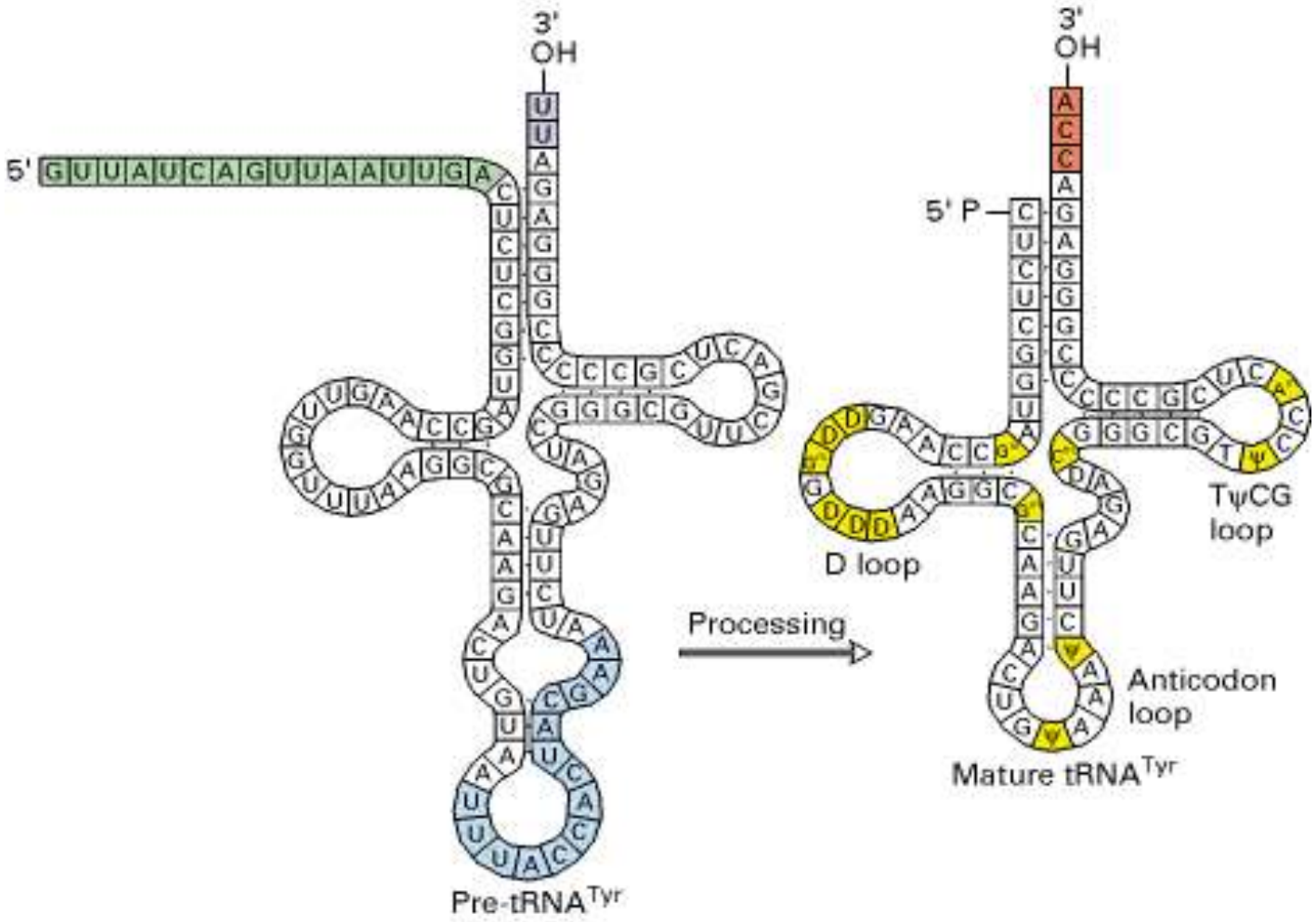
- tRNAs are transcribed by **RNA polymerase III** as pre-tRNAs in the nucleus




Transcription and processing of tRNA in prokaryotes




**Processing of prokaryotic tRNA**

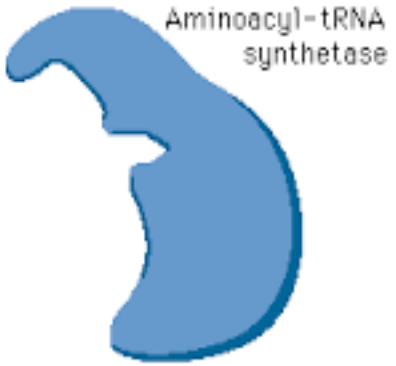



**Processing of yeast pre-tRNA$_{tyr}$ involves four types of changes.** A 14-nucleotide intron (blue) in the anticodon loop is removed by splicing. A 16-nucleotide sequence (green) at the 5′ end is cleaved by RNase P. U residues at the 3′ end are replaced by the CCA sequence (red) found in all mature tRNAs. Numerous bases in the stem-loops are converted to characteristic modified bases (yellow). Not all pre-tRNAs contain introns that are spliced out during processing, but they all undergo the other types of changes shown here. D = dihydrouridine; Y = pseudouridine.

**Structure of a tRNA-splicing endonuclease docked to a precursor tRNA.** The endonuclease (a four-subunit enzyme) removes the tRNA intron *(blue).* A second enzyme, a multifunctional tRNA ligase (not shown), then joins the two tRNA halves together

# Aminoacyl-trna synthetases

- catalyzes the esterification of a specific amino acid
- The accuracy of protein translation depends on the fidelity with which the correct amino acids are esterified to their cognate tRNA molecules by aminoacyl tRNA synthetases.
- This enzyme catalyzes the union of amino acid and tRNA

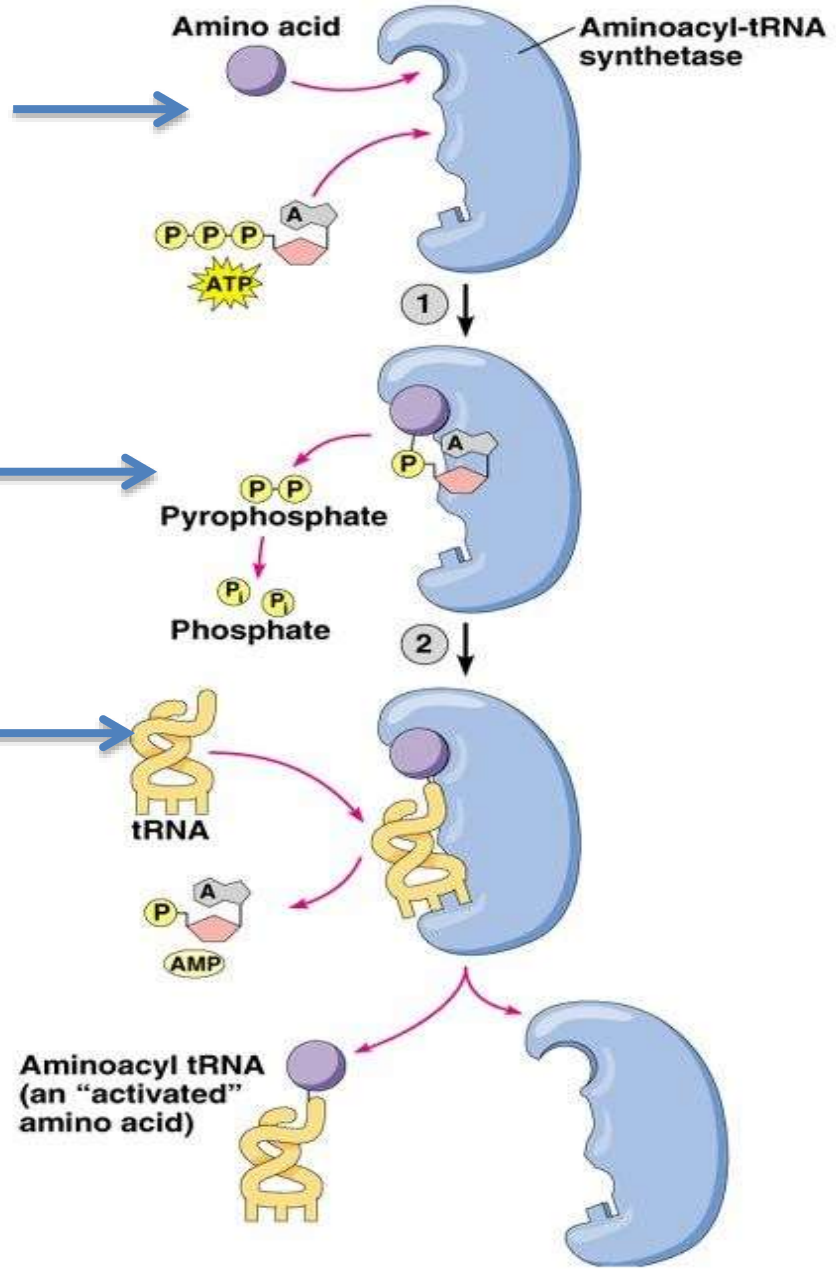


Enzyme binds the amino acid and joins it to a molecular of ATP

Cleaving two phosphate groups from a molecule of ATP

Enzyme binds the aminoacyl portion of this complex to an appropriate tRNA molecule while releasing the AMP molecule.

Union of tRNA and amino acid

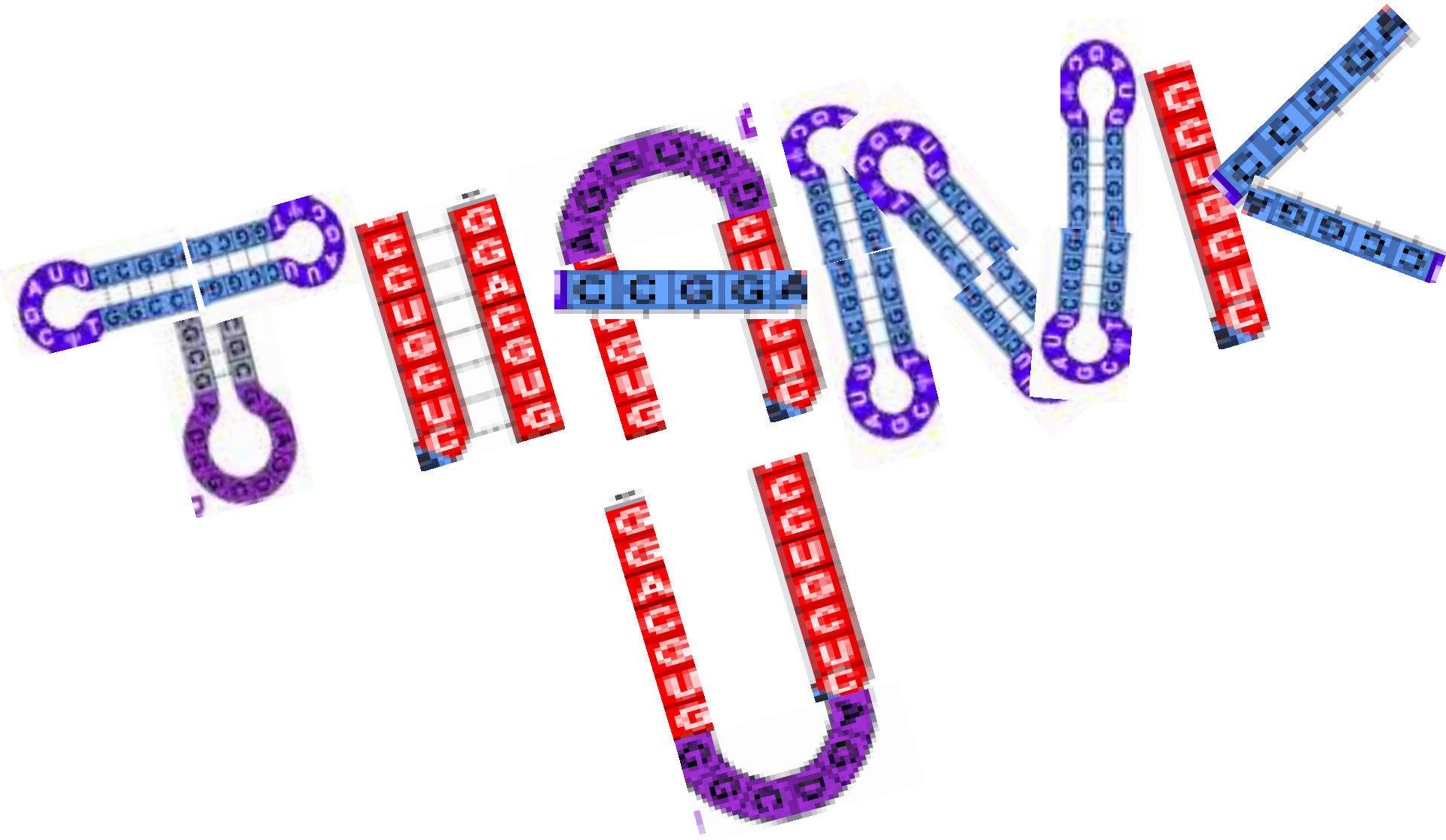


# Functions of tRNA

- Help in the recognition of *Aminoacyl tRNA synthetase* enzyme
- Picks up specific amino acid from cytoplasm and carries to site of protein synthesis
- Attaches itself to ribosome in accordance with sequence specified by mRNA
- Transmits amino acid to polypeptide chain
- **Participate in non protein synthetic processess such as a primer during reverse transcription in retrovirus life cycles**

# References


- *Instant notes* Molecular biology- by P.C Turner, A.G McLenman, A.D.Bates and M.R.H. White
- Life sciences fundamentals and practice part I- by Pranav kumar and Usha Meena
- *Theagricos* knowledge database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