

6.6 FIVE STRUCTURAL GENES OF TRP-OPERON (4)

1. Trp E : It encodes the enzyme Anthranilate synthase I
2. Trp D : It encodes the enzymes Anthranilate synthase II
3. Trp C : It encodes the enzymes N-5' phosphoribosyl anthranilate isomerase and indole -3- glyceolphosphate synthesis.
4. Trp B : It encodes the enzymes tryptophan synthase -B subunit.
5. Trp A : It encodes the enzymes tryptophan synthase-A subunit.

These structural genes are regulated to efficiently express only when tryptophan is limiting.

There are two levels to regulate the Trp-operon.

- Transcription repression by the Trp-repressor
- Attenuation: a second level of regulation of Trp-operon
- Transcription repression by the Trp-repressor

It is the first level of regulation of tryptophan operon. Tryptophan operon is **repressible operon** because the repressor binds to operator in the presence of tryptophan. In absence of tryptophan, it allows the transcription to proceed because when tryptophan concentration is low the repressor cannot bind to the operator and allows RNA polymerase to bind to the promoter to start the synthesis of proteins.

- Attenuation: A second level of Regulation of tryptophan operon.

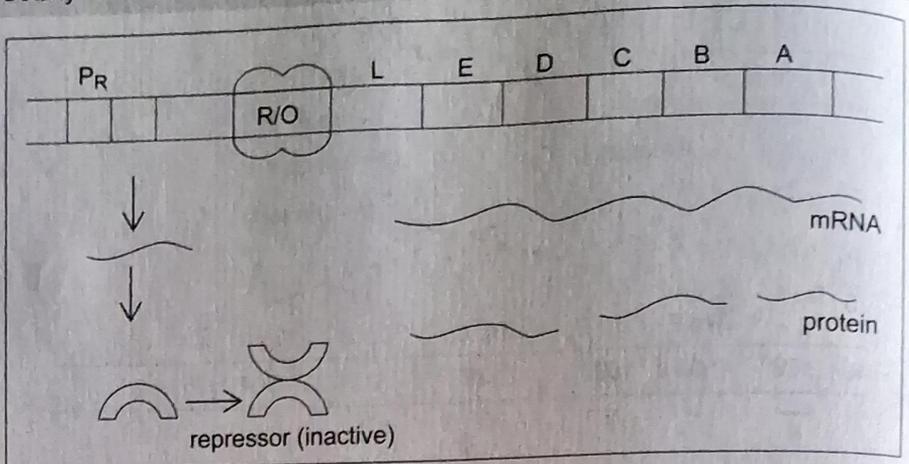
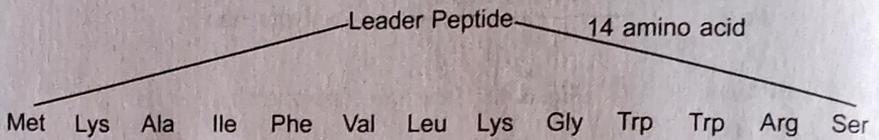


Fig. 6.5. Condition-low tryptophan.

The tryptophan operon contains a **leader sequence** upstream of the structural gene E. This leader sequence encodes a 14 amino acid **leader peptide** containing two tryptophan residues at 10th and 11th position. This plays an important role in the fine tuning of expression of genes inside the cells.



Near the end of this leader sequence, and before Trp E, is a transcription terminator which is used to compose a characteristic hairpin loop in the RNA. The leader sequence has four regions: Region 1, Region 2, Region 3 and Region 4. These regions have the capacity to form a variety of base paired stem-loop (hairpin) secondary structure. The hairpin loop is followed by uridine residues. At the end of leader, a region called attenuator is present. This is the site where transcription usually stops yielding a leader RNA of only 139 nucleotide long.

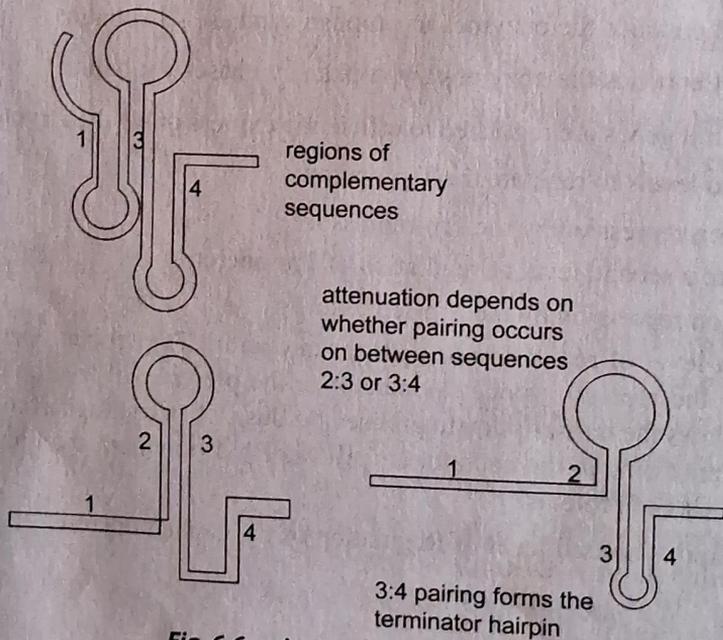


Fig. 6.6. Attenuation sequences.

Region 3 is complementary to both regions 2 and 4. If **region 3** and **region 4** base pair with each other, they form a loop like structure called **attenuator**. It forms the terminator hairpin and stops transcription. If pairing occurs between region 2 and region 3 then no attenuator is formed and transcription proceeds.

There are two conditions for regulation of Tryptophan levels:

- If low tryptophan concentration, then tRNA^{trp} is also less available to the ribosome. So, ribosome stalls at two trp codon present in a leader region 1 and in the meantime region 2 and region 3 base pairs in such a way that terminator hairpin is not formed. Now, RNA polymerase binds to the promoter region and transcription proceeds.
- If high tryptophan concentration - If there is plenty of tryptophan, the ribosome won't have to wait for a long for tRNA^{trp} because they are abundant in the environment, so ribosome moves rapidly and overlaps sequences 1 and 2, thus sequences 3 pairs with sequences 4 forming a terminator hairpin which stops transcription. The below figure represents the low tryptophan concentration tRNA less available. Ribosome stalls at the two trp codons and RNA pol continues to transcribe sequence 2 and 3 base pairs termination. Further, hairpin is not formed as transcription proceeds.

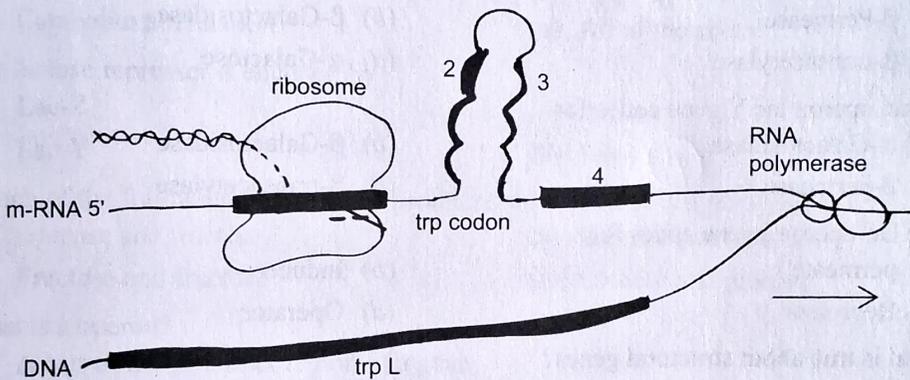


Fig. 6.7. Condition-low tryptophan concentration.

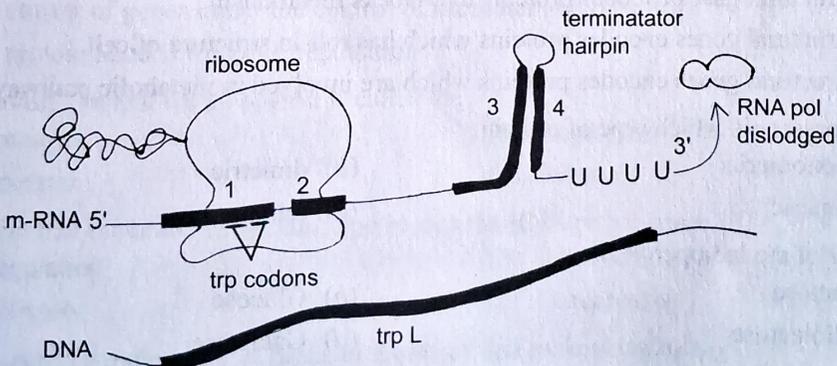


Fig. 6.8. Condition-high tryptophan concentration.

In the Figure 5.8 the high tryptophan concentration tRNA is abundant and ribosome moves rapidly and overlaps the sequences 1 and 2. RNA pol proceeds transcribing sequences 3 and 4. Sequences 3 and 4 pairs with each other and then terminator hairpin is formed and transcription stops.