Module 11 / RNA Synthesis

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Describe the difference between a eukaryotic and prokaryotic Operon	Describe the similarities between the functioning of DNA plymerase and RNA polymerase	Describe the differences between the substrate requirements for DNA polymerase and RNA polymerase
Explain the difference between a weak and a strong promoter including the basis for the difference in "strength" of the promoter	Describe the steps in the process of transcription (RNA synthesis) starting with RNA polymerase binding to the promoter and ending with the release of polymerase from the terminator	Name the products of transcription and differentiate each from the others

Transcription

The process of DNA transcription is described as **DNA-directed RNA synthesis**. This process, which takes place in the nucleus of eukaryotic cells, has many similarities with DNA replication. The enzyme to catalyze the process is **RNA Polymerase** that likewise has some characteristics similar to those of DNA polymerase used in replication but it also has significant differences.

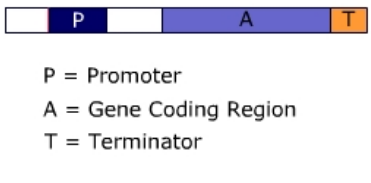
- The DNA must be unwound starting at a specific DNA sequence
  - **Replication** – Origin of Replication
  - **Transcription** – The Promoter
- The process requires a template that is DNA
  - **Replication** – Complementary copies of both strands are created simultaneously
  - **Transcription** – A complementary copy is made of one strand
- The process does not require a primer
  - **Replication** – A primer with a 3' hydroxyl is required for DNA polymerase
  - **Transcription** – No primer is required for RNA polymerase
- Synthesis is unidirectional in the 5' to 3' direction
- Synthesis requires four nucleotide triphosphates
  - **Replication** – deoxyribonucleotide triphosphates include dATP, dGTP, dCTP, dTTP
  - **Transcription** – ribonucleotide triphosphates include ATP, GTP, CTP, UTP

The Operon

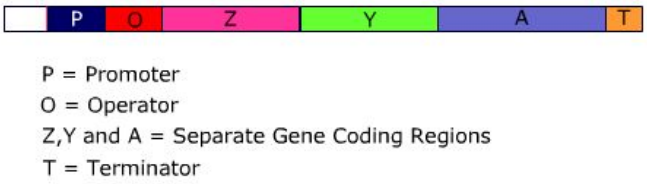
The organization of the genetic information on the chromosomes of prokaryotes and eukaryotes is different. In each case, as indicated below, the sequence for initiation of transcription is the Promoter and the

sequence signaling the end of transcription is the Terminator. However, in eukaryotes there is generally one

sequence signaling the end of transcription is the Terminator. However, in eukaryotes there is generally one gene that codes for one product under the control of a single promoter. In contrast, prokaryotes generally have multiple genes, each coding for a separate product, under the control of a single promoter. This unit of a promoter, a terminator and the intervening gene or genes is called an **Operon**. The operon also contains the controlling elements for the operon. The control of expression of an operon is the topic for a separate unit.



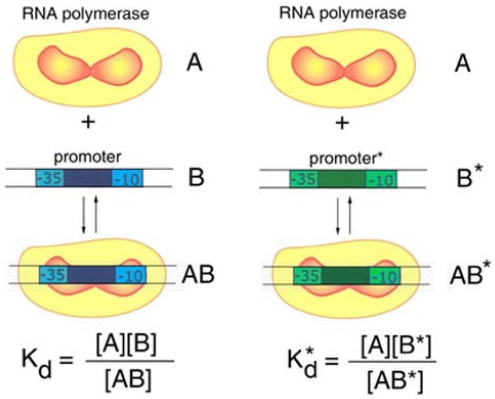
Eukaryotic Operon



Prokaryotic Operon

The Promoter

RNA polymerase is an example of a quaternary structure composed of a core protein for the synthesis the complementary RNA strand and a subunit, the Sigma subunit, that first binds to the promoter region of the Operon and creates an open complex by unwinding the double stranded DNA. The promoter region of the operon is critical in defining how much or the frequency with which transcription of an operon takes place. Promoters have different sequences. The sigma subunit binds to the different promoter sequences with different affinities. In the illustration below, the sigma subunit binds to both of the promoters and a dissociation constant, Kd, can be written for each binding.



Different promoters result in different K_d values.

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If Kd > Kd*, which promoter is bound more tightly to RNA polymerase? Which has the higher probability of initiating transcription?

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Promoters can be characterized as being strong or weak promoters depending on their affinity for RNA

Promoters can be characterized as being strong or weak promoters depending on their affinity for RNA polymerase. The result of a strong promoter with a high affinity (tight binding) to RNA polymerase is a greater frequency of starting transcription from that promoter. Therefore, more product from transcription initiated at a strong promoter is possible compared to product formation initiated from a weak promoter in the same amount of time.

RNA Synthesis

Once RNA polymerase binds to the promoter, initiation of RNA synthesis or Transcription proceeds. The following animation depicts the steps of transcription.

The Products of Transcription

The product of the process of transcription is RNA. There are three distinctly different RNA products that result from transcription based on their function within the cell.

- **Ribosomal-RNA (rRNA**) is a large defined length of RNA that when processed is folded into a tertiary structure that is the scaffolding for the structure of the ribosome. The ribosome is a protein-RNA complex quaternary structure where protein synthesis (translation) takes place.
- **Messenger-RNA (mRNA)** is a variable length of RNA whose length is dependent on the length of the coding needed to produce a protein by translation. This RNA is the message that carries the information that codes for the synthesis of a protein. It has a relatively short lifetime in the cell.
- **Transfer-RNA (tRNA)** is a set of short RNA molecules generally 75-80 nucleotides in length. Each tRNA molecule is specific for transporting a specific amino acid to the ribosome during protein synthesis (translation). All of the tRNA molecules have a common, compact tertiary structure.

Each of the RNA classes (rRNA, mRNA, and tRNA) is produced during transcription using a different RNA polymerase.

- rRNA – RNA polymerase I
- mRNA – RNA polymerase II
- tRNA – RNA polymerase III

Bioselectivity describes the discrimination of a specific RNA polymerase for a specific promoter yielding different RNA products. Equilibrium binding of the polymerase with the promoter explains the differential production of products using the same polymerase and different promoter sequences.

did I get this

Which of the following special base sequences signals the start of transcription?

- promotor
- stop codon
- operator
- transcription termination signal
- CAP site

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