

## DNA transcription

### prokaryotic cell

The DNA-dependent RNA polymerase of E. Coli is a large complex enzyme with five core subunits and a sixth subunit. The sigma subunit of RNA polymerase binds transiently to the core and directs the enzyme to specific binding sites on the DNA. RNA synthesis begins at promoters which is between positions -70 and +30. The RNA polymerase bonds with the promoter by helping the sigma factor bind with the consensus sequence between -35 and -10. The sigma 2 binds to the -10 region whereas sigma 4 binds to the -35 region. The sigma factor one prevents the dissociation of RNA polymerase. The sigma factor two separates dsDNA and stabilises ssDNA. Sigma factors three and four will help position incoming rNTP on RNA polymerase's active site. The alpha unit of RNA polymerase bonds to UP elements, an A-T-rich recognition element. Then transcription is initiated within the complex.

### eukaryotic cell

#### initiation

The transcription of Eukaryotic cells is more complicated. There are several transcription factors which are necessary for transcription initiation. The step-by-step process starts with the TBP(TATA-binding protein), the main unit of TFIID binding with the TATA box in the promoter, causing distortion. Then TFIID-TBP associate factors bind to several elements on the template strand. TFIIA bind to the promoter and stabilises the TBP complex. TFIIB binds to the promoter complex which is the TFIIB recognition element, providing an important link to RNA polymerase II. Then the TFIIF interact TBP-TFIIB complex and facilitated RNA polymerase II binds with the TBP-TFII complex. After RNA polymerase II bind with the promoter complex, the transcription bubble starts forming. Then TFIIE binds with these initiation complexes, leading other factor TFIIH to bind with them. Then a closed initiation complex formed. TFIIH has nine subunits and includes a DNA helicase activity that could unwind the DNA. Two of the subunits in TFIIH have ATPase activity which provides energy to unwinding DNA. In the end, the open complex formed so the transcription starts.

#### elongation

When transcription is formed, 8pb of DNA-RNA hybrid is formed. There is an abortive synthesis before elongation, releasing the failure of RNA short 2-9bp RNAs. Then the carboxyl-terminal domain(CTD) of RNA pol II is phosphorylated by several kinases. Then rNTP enters the active side of RNA pol II and pairs in a processive manner. Transcription bubble size remains constant. Supercoiling caused by chromatin flanks the transcription bubble Torsion is released through topoisomerases

#### Termination

##### Prokaryotic

There are two strategies for prokaryotic cell termination. One is intrinsic termination, by using lots of 8-10 U bases from an RNA hairpin loop to pull RNA out of the polymerase active site. At other sites, however, termination requires the participation of an additional factor. Which is rho factor-dependent termination. There is a ring structure which contains ATPase activity in the rho factor. The C-rich domains in the rho factor activate it to jump into the transcription complex and chase them. Then the rho factor terminates the transcription by changing the RNA pol II and pulling the RNA out of the pol II with the hydrolysis of ATP.

## **Eukaryotic cell**

There are two strategies which are allosteric modules and torpedo for termination. The allosteric termination needs two factors, CPSF and CSTF. First, The CTD(as discussed before) of pol II phosphorylation changes then CPSF and CSTF bind to the CTD of pol II. Then the polyA signal sequence is added to pol II, leading to CPSF and CSTF transfer to RNA and cleaving it. Then pol II change the conformation which helps the dissociation of RNA from DNA.

As for torpedo termination, the RNase bind to pol II, and when the polyA sequence is added to pol II, it activates the RNase to degrade nascent RNA, and also the uncapped RNA. Then transcription ends and pol II dissociates from DNA. A combination of torpedo and allosteric models may be responsible for termination.