<Barresi13e, Further Development_c03_07>

Poised Chromatin

Promoters can exist in three major states: an active state, a repressed state, and an intermediate, or "poised" state (see Figure 3.14 in the text). This poised chromatin state allows for a rapid response to developmental signals, and it characterizes the high CpG-content promoters (HCPs) that regulate the transcription of developmental control genes. The DNA of HCPs is relatively unmethylated, and nucleosomes tend to be enriched with "activating" H3K4me3. As a result, RNA poly- merase II is usually already present on HCPs (Hon et al. 2009; Ernst and Kellis 2010). Indeed, there is often a small, truncat- ed transcript of nRNA already initiated (but not completed) at these promoters (see Figure 2.15). DNA methylation does not appear to play a major role in HCP regulation. Rather, HCPs can be repressed by modifying the histone 3 to H3K27me3, which recruits Polycomb repressive complex 2 (Peng et al. 2009; Li et al. 2010), a complex that appears to inhibit further RNA polymerase II binding as well as preventing elongation of the existing nRNA transcripts.

HCPs become poised for activation by having nucleo- somes containing both H3K4me3 (activating) and H3K27me3 (repressive) histones (this is sometimes called a bivalent state). Thus, the rate-limiting step of RNA transcription from HCPs is not the *initiation* of transcription (as it is in the LCPs), but RNA *elongation*. This "poised for activation" state may be predominant during early development (Muse et al. 2007; Zeitlinger 2007). The genes may be put into an active state by specific transcription factors that activate the elongation of RNA transcripts (Peterlin and Price 2006), and they may be repressed later in development (Hargreaves et al. 2009; Ramirez-Carrozzi et al. 2009; Rahl et al. 2010).

These transcription factors may act on several levels to promote RNA elongation. In mammalian cells, where about 30% of the genes have promoters that already contain RNA polymerase II and nascent RNA chains (Core and Lis 2008), transcription factors appear to act through the Mediator complex. Here transcription is paused because the RNA polymerase II remains tethered to TFIID, which remains bound to the promoter sequence of the gene. This tethering is accomplished by the Mediator complex, especially by the Mediator protein Med26, which binds the Mediator to TFIID (Figure 1). In order to elongate the RNA, a signal must enable the multiprotein transcription elongation com- plex (TEC) to compete with TFIID for the favors of Med26. Once the transcription elongation complex frees the RNA polymerase II from TFIID, RNA polymerase II can become phosphorylated and travel along the DNA to transcribe the gene (Takahashi et al. 2011).

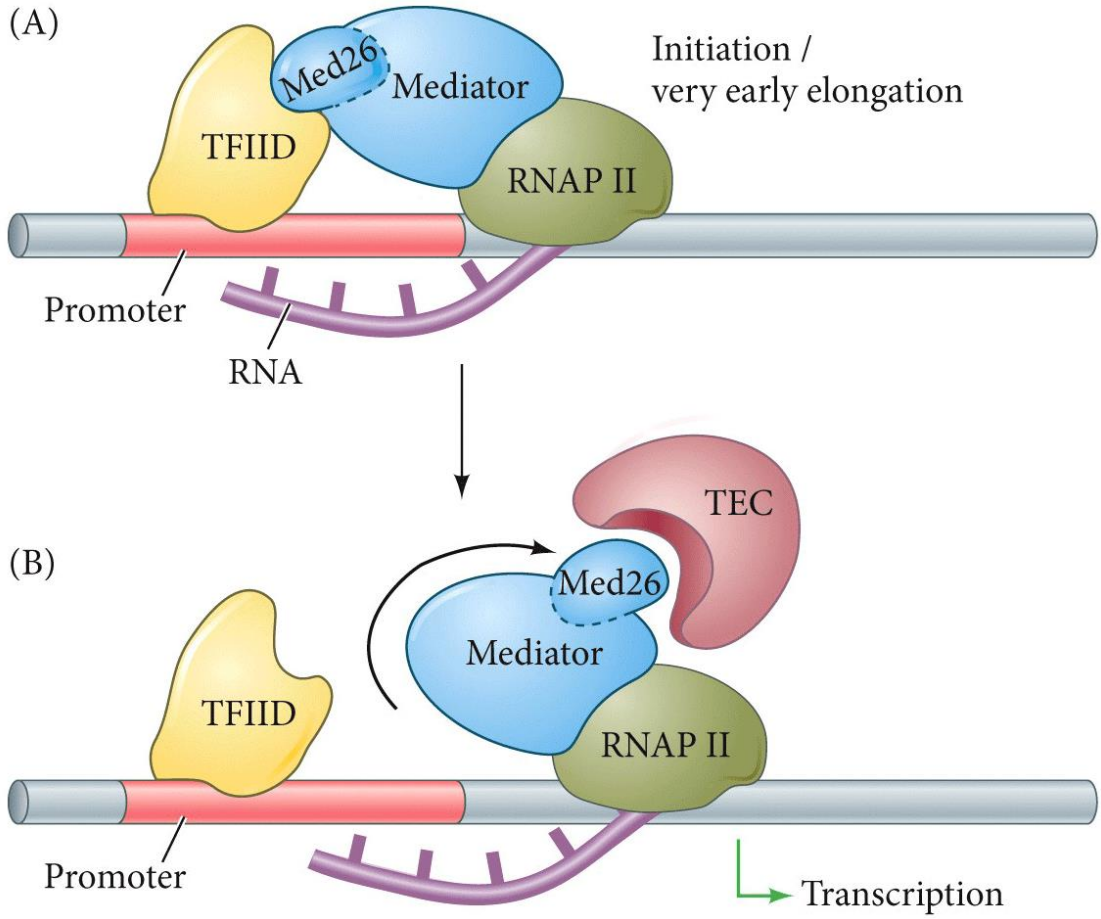

Figure 1 Model for the regulation of RNA elongation by the Mediator protein Med26. In the initiation and early elongation phase of transcription, the Mediator tethers RNA polymerase II to TFIID at the promoter through its Med26 protein. The Med26 protein can also bind to the transcription elongation complex (TEC). Transcription elongation can be reactivated by transcription factors promoting the binding of Med26 to the TEC rather than to TFIID. (After Takahashi et al. 2011.)

Drosophila may use a slightly different mechanism to pause the transcription from HCPs. In many of these genes, there appears to be a DNA sequence in the proximal promoter (i.e., the sequences of the promoter closest to the exons) that acts as a “pause button” (Hendrix et al. 2008). About 1500 genes in Drosophila embryos have RNA polymerase II already on their promoters, and these genes are primarily those active in regulating early development (Muse et al. 2007; Zeitlinger et al. 2007). It is possible that these “pause button” sequences may be more difficult to unwind, and the presence of the poly- merase may enable elongation inhibitory factors to assemble there (Levine 2011).

But how does the release of a single transcript influence the synthesis of that protein? It certainly takes more than one transcript to produce significant amounts of gene product. In some cases, it appears that the transcript of the paused poly- merase can recruit histone-activating proteins, enabling fur- ther transcription to occur as soon as elongation commences (Petesch and Lis 2008). It is also possible that paused RNA polymerase II prevents the assembly of new nucleosomes on the promoter, keeping the gene in an open configuration (Gilchrist et al. 2010; Nechaev et al. 2010).

Thus, both high and low CpG-content promoters regulate RNA synthesis, but they do so in different manners. LCPs are usually turned off, requiring transcription factors to enable gene expression by promoting access of RNA polymerase II to the DNA. HCPs already have initiated transcription, but transcription is not completed. Here, developmental signals allow the elongation of the nascent

nRNA. Both LCPs and HCPs have repressed states that prevent transcription, as well as poised states that enable the genes to be transcribed immediately when the appropriate signal is received.

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