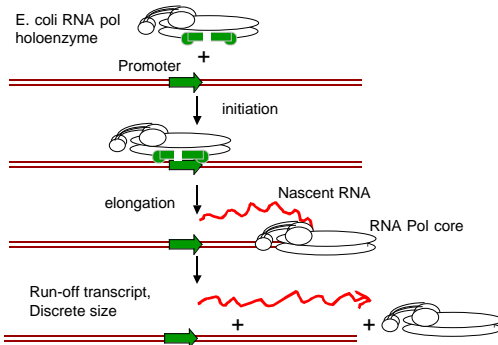
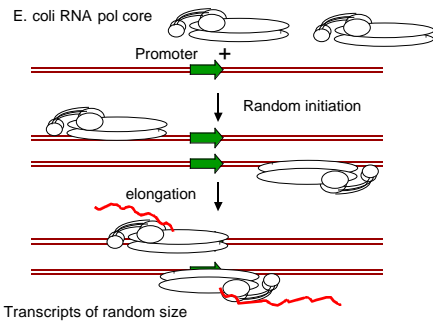


General Transcription Initiation Factors

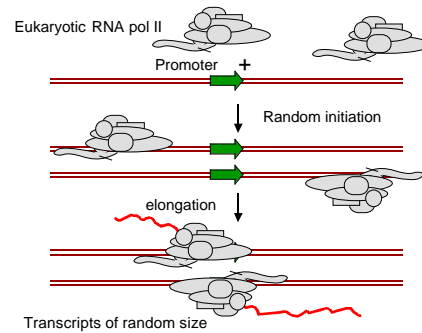
Accurate initiation by bacterial RNA polymerase



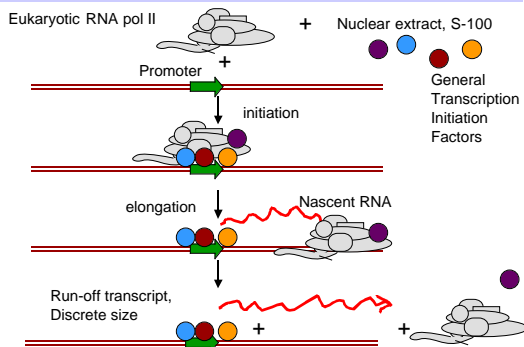
Random transcription by bacterial RNA polymerase core



Random transcription by eukaryotic RNA polymerase

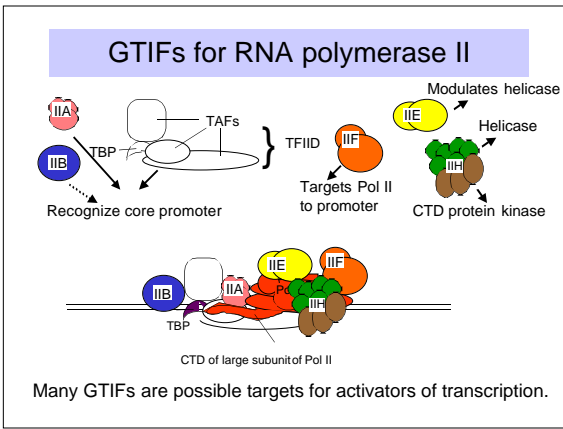
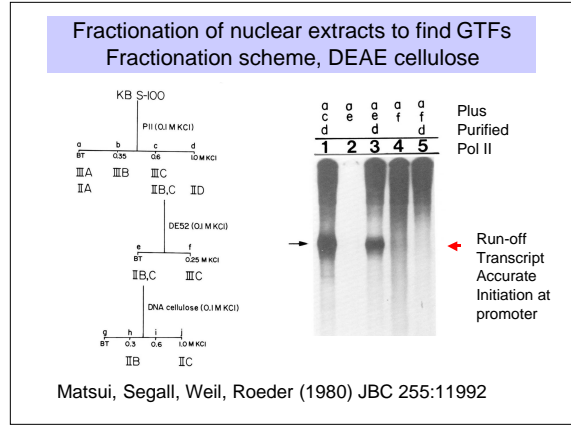
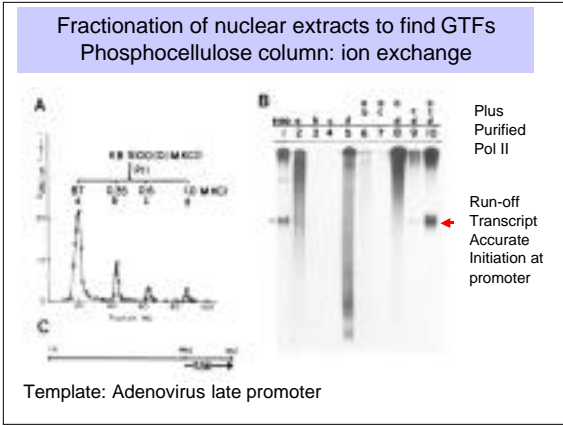


Accurate initiation by Euk RNA polymerase II plus factors in the nucleus

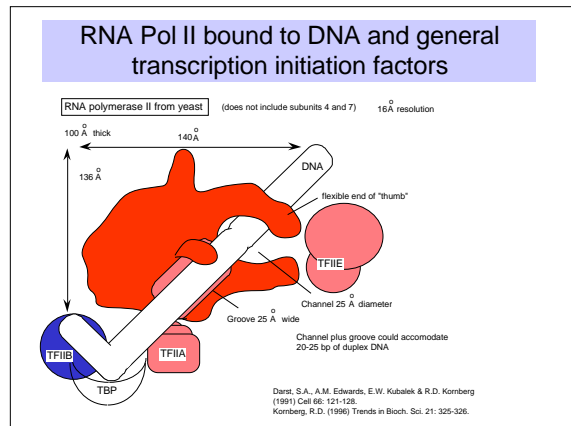
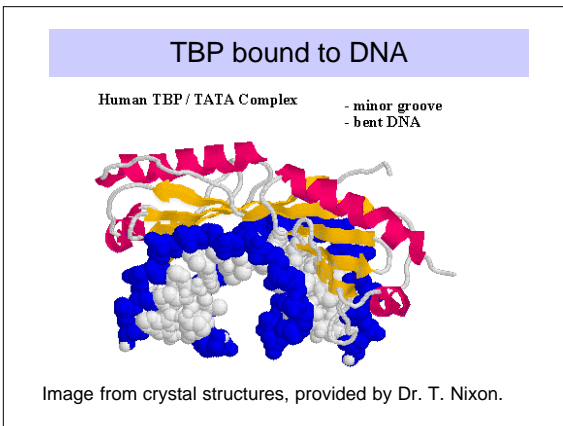


General transcription factors = GTFs

- Proteins other than RNA polymerase involved in transcription
 - Initiation, Elongation, Termination
- General transcription **initiation** factors (GTIFs)
 - Proteins **required** for specific transcription from a minimal promoter (core)
 - **Not** subunits of purified RNA polymerase.
 - Required for RNA polymerase to bind avidly and specifically to promoters.
 - GTIFs for RNA polymerase II are called TFI_x, where x = A, B, D, ...
 - Can have multiple subunits



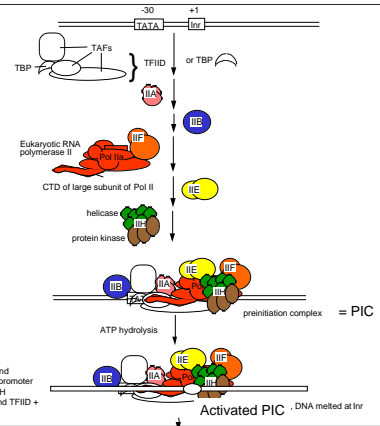
- ### TATA Binding Protein = TBP
- TBP binds in the narrow groove of DNA at the TATA box found about 20-25 bp 5' to the start site for transcription of *many* (but not all) genes transcribed by RNA polymerase II.
 - TBP bends the DNA about 90 degrees.
 - TBP alone or with TBP-associated proteins (TAFs) plays an important role in recognizing the core promoter and recruiting RNA polymerase II to the promoter.



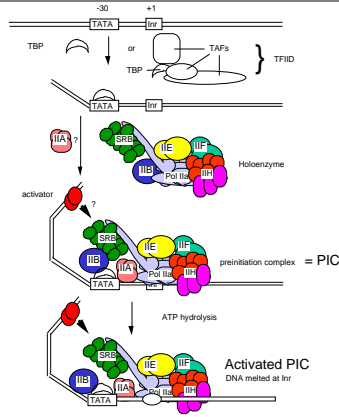
Movie of TBP, then binding to TATA DNA (and bending it), and then TFIIA binding to TBP-TATA

Thanks to Drs. Song Tan and Tracy Nixon for movies

Sequential Binding Model for assembly of preinitiation complex



Direct Binding of Holoenzyme model for assembly of the preinitiation complex



Other proteins involved in transcription and regulation

- In addition to RNA polymerase II and GTFs:
- Proteins required for regulation**, e.g.
 - Gal11: regulation of the GAL operon
 - Rgr1: resistance to glucose repression
- Srb proteins**
 - Yeast strains with truncations in the CTD of the large subunit of RNA polymerase B are cold-sensitive
 - SRB genes: when mutated, suppress the phenotype of CTD deletions
 - Extragenic suppressors: implicated in RNA polymerase function

RNA polymerase II Holoenzyme and Mediator

- Holoenzyme**
 - RNA polymerase II + (TFIIB, E, F, H) + (Srb2, 4, 5, 6) + (Rgr1, Gal11, others)
 - Correct initiation in presence of TBP (TFIID)
 - Responds to transcriptional activators
- Mediator**
 - Complex needed for a response to transcriptional activators by purified RNA Pol II plus GTFs
 - Yeast Mediator has 20 subunits, including Srb2, 4, 5, 6; Srb7, Rgr1, Gal11, Med 1, 2, 6, 7, Pgd1, Nut 1, 2, and others
- RNA Pol II + Mediator (+ some GTFs?) = Holoenzyme**

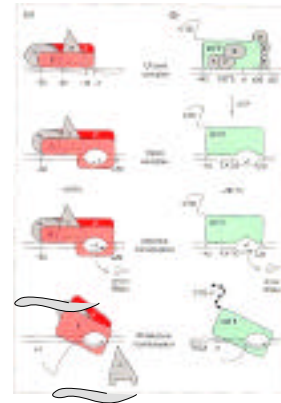
Expanding the functions of RNA polymerase

Polymerase	Transcribe	Start at Promoter	Respond to Activator
RNA Pol II	Yes	No	No
RNA Pol II + GTFs	Yes	Yes	No
RNA Pol II holoenzyme + GTFs	Yes	Yes	Yes

Stages in Initiation of Transcription

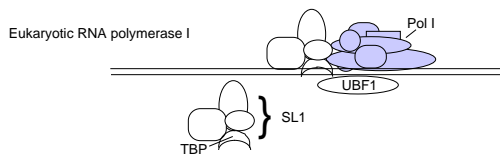
- | | |
|--|--|
| <ul style="list-style-type: none"> • Bacterial transcription • Closed complex: holoenzyme+promoter • Open complex (DNA melting, not need ATP) • Abortive transcription • Productive initiation <ul style="list-style-type: none"> – Transcribe past +9 – Sigma dissociates • Elongation | <ul style="list-style-type: none"> • Eukaryotic transcription • Preinitiation complex (PIC) assembly • PIC activation (DNA melting, needs ATP) • Abortive transcription • Productive initiation <ul style="list-style-type: none"> – CTD phosphorylated – Promoter clearance • Elongation |
|--|--|

Parallels between initiation pathway in prokaryotes and eukaryotes

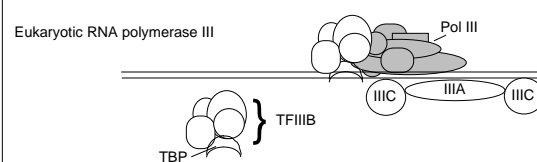


From Eick et al. (1994)
Trends in Genetics 10: 292-296

GTFs for RNA polymerase I



GTFs for RNA polymerase III



TBP is used by all 3 RNA polymerases

- TBP is a subunit of an important GTF for each of the 3 RNA polymerases:
 - TBP or TFIID for Pol II
 - SL1 for Pol I
 - TFIIIB for Pol III
- It does NOT always bind to TATA boxes; promoters for RNA Pol I and Pol III (and even some for Pol II) do not have TATA boxes, but TBP is still used.
- The GTFs that contain TBP may serve as **positioning factors** for their respective polymerases.