

Out of Its Element: TBP Function at TATA-Less Promoters

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Transcription of genes is initiated at specialized regions of DNA called promoters, which contain binding sites for numerous factors. One such protein is TATA-binding protein (TBP), so named for its ability to bind TATA-containing DNA with high affinity. However, genome-wide analysis of promoter sequences in organisms from yeast to human has shown that the majority of promoters lack TATA boxes. This leaves open the question of whether the DNA binding activity of TBP is required for transcription from TATA-less promoters. To dissect the functions of TBP at TATA-containing and TATA-less promoters, graduate student Ivanka Kamenova and colleagues in the laboratory of Dr. Steven Hahn (Basic Sciences Division) tested the effects of mutations in the DNA-binding surface of TBP on transcription from each type of promoter.

The researchers first created a series of yeast strains harboring mutations in the DNA-binding domain of TBP. Notably, with the exception of a single mutant, each TBP derivative supported the growth of yeast, though three additional mutants grew more slowly than wild-type. As stress-induced genes are generally transcribed from TATA-containing promoters, the researchers also tested the growth of their TBP mutant strains on medium containing a drug that mimics amino acid starvation. Most of the TBP mutants are sensitive to this drug, consistent with the idea that sequence-specific TBP DNA binding is primarily required for transcription from TATA-containing promoters.

To test the effect of these TBP DNA-binding mutants on *in vivo* transcription, Kamenova et al examined the expression of two TATA-containing genes, *HIS4* and *SNZ1*, and two TATA-less genes, *RPS5* and *RPL5* in strains containing the TBP mutants. All but two TBP derivatives were unable to rescue the expression of *HIS4* and *SNZ1*, while all but two TBP mutants were able to complement transcription of *RPS5* and *RPL5*. These data support the notion that the specific DNA binding activity of TBP is not important for transcription from TATA-less promoters. Further analysis using an *in vitro* transcription system revealed that TBP, but not its intact DNA-binding surface, was required for transcription of a TATA-less gene.

The lack of a requirement for an intact DNA binding surface of TBP for TATA-less transcription led Kamenova et al. to assess the ability of TBP to associate with TATA-less promoters. *In vitro*, they

found that TBP was able to associate with the RPS5 promoter. Using deoxyribonuclease (DNase) I digestion of the TBP-RPS5 promoter complex to determine the regions of DNA bound by TBP, the authors identified three “footprints” of TBP occupancy. Interestingly, mutation of any of these regions did not substantially alter RPS5 transcription, further supporting the idea that specific DNA binding is not important for TBP-mediated transcription from TATA-less promoters.

These results show important differences in the requirement for TBP DNA binding at TATA-containing versus TATA-less promoters, providing insight into the complexity of eukaryotic transcriptional regulation. "Our results are consistent with the model that sequence-specific TBP-DNA contacts are not important at yeast TATA-less genes and suggest that other general transcription factors or coactivator subunits are responsible for recognition of TATA-less promoters," said Dr. Hahn. "These findings are contrary to the common assumption that TATA DNA binding activity is an essential function of TBP. Our future studies aim to understand the role of TBP and its DNA binding function at genes that lack TATA elements."

[Kamenova I, Warfield L, Hahn S.](#) 2014. Mutations on the DNA binding surface of TBP discriminate between yeast TATA and TATA-less gene transcription. *Mol Cell Biol* 34(15):2929-2943.

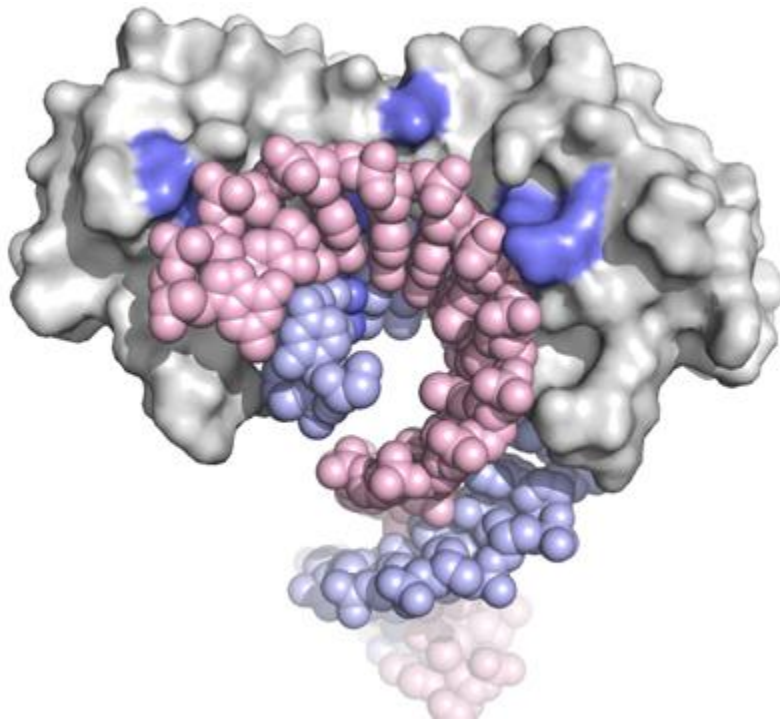


Image provided by Dr. Steven Hahn.

Structure of yeast TBP (grey) in complex with DNA (pink/purple). Residues targeted for mutation in this study are indicated in blue.