Multiple Functions of the Nonconserved N-Terminal Domain of Yeast TATA-Binding Protein

Mark Lee1 and Kevin Struhl

Department of Biological Chemistry and Molecular Pharmacology, Harvard Medical School, Boston, Massachusetts 02115

Manuscript received December 15, 2000
Accepted for publication January 26, 2001

ABSTRACT

The TATA-binding protein (TBP) is composed of a highly conserved core domain sufficient for TATA-element binding and preinitiation complex formation as well as a highly divergent N-terminal region that is dispensable for yeast cell viability. In vitro, removal of the N-terminal region domain enhances TBP-TATA association and TBP dimerization. Here, we examine the effects of truncation of the N-terminal region in the context of yeast TBP mutants with specific defects in DNA binding and in interactions with various proteins. For a subset of mutations that disrupt DNA binding and the response to transcriptional activators, removal of the N-terminal domain rescues their transcriptional defects. By contrast, deletion of the N-terminal region is lethal in combination with mutations on a limited surface of TBP. Although this surface is important for interactions with TFIIA and Brf1, TBP interactions with these two factors do not appear to be responsible for this dependence on the N-terminal region. Our results suggest that the N-terminal region of TBP has at least two distinct functions in vivo. It inhibits the interaction of TBP with TATA elements, and it acts positively in combination with a specific region of the TBP core domain that presumably interacts with another protein(s).

THE TATA-binding protein (TBP) plays a central role in eukaryotic transcription, being required for accurate transcriptional initiation by all three nuclear RNA polymerases (Hernandez 1993; Struhl 1994; Burley and Roeder 1996). TBPs from a wide variety of eukaryotes and arabia have a highly conserved 180-residue C-terminal core domain (80% identical between yeast and human) that is sufficient for TATA-element binding and preinitiation complex formation. TBPs also contain an N-terminal region that is very divergent across species with respect to both length and sequence and whose function is poorly understood. Human TBP does not support yeast cell growth, but species-specific difference maps to the C-terminal core domain, not the N-terminal region (Cormack et al. 1991; Gill and Tjian 1991). Indeed, a single amino acid change within the core domain enables human TBP to substitute for all of the essential activities of yeast TBP in vivo (Cormack et al. 1994).

The TBP core domain and full-length TBP behave similarly in many biochemical assays, but there are several differences that provide insight into the function of the N-terminal domain. First, deletion of the N-terminal region results in enhanced binding and bending of DNA relative to full-length TBP (Horikoshi et al. 1990; Lieberman et al. 1991), primarily by increasing the rate of TBP-TATA association (Kiddus and Schmidt 1993). As a consequence, DNA binding by TBP mutants defective for the TBP-TATA interaction can be restored by removal of the N-terminal domain (Lee et al. 1992). Second, the yeast TBP core domain readily forms transcriptionally inert dimers and higher order oligomers in vitro (Kato et al. 1994; Coleman et al. 1995). Although the physiological relevance of TBP dimerization is controversial (Jackson-Fisher et al. 1999; Geisberg and Struhl 2000), the N-terminal domain inhibits oligomerization of full-length TBP, potentially enhancing the probability that TBP is monomeric under physiologic conditions (Campbell et al. 2000). Third, a peptide or a monoclonal antibody corresponding to a portion of the N-terminal region selectively blocks TATA-dependent transcription by RNA polymerases II and III while sparing RNA polymerase I and TATA-less promoter transcription (Lescure et al. 1994). Fourth, yeast and human NC2 are most effective in repressing transcription in reactions reconstituted with TBP from the same species, and maximal repression requires the N-terminal region (Goppelt and Meisterernst 1996). Fifth, removal of the N-terminal region of yeast TBP increases interactions with human TBP-associated factors (TAFs) and the formation of a hybrid TFIIID complex (Zhou and Berk 1995). Sixth, the N-terminal region of human TBP mediates cooperative binding with the small nuclear RNA activating protein (SNAP) complex to the U6 promoter and enhances U6 transcription (Mittal et al. 2001).
and Hernandez 1997). Taken together, these results suggest a regulatory role for the N-terminal domain in governing the interactions of TBP with the TATA element, with other transcription factors, and with TBP itself. However, the physiological relevance of these biochemical properties of the TBP N-terminal region remains to be established.

In yeast cells, expression of the TBP core domain at physiological levels has minimal phenotypic consequence, indicating that the 63-residue N-terminal region of TBP is dispensable for viability and the response to transcriptional activators (Cormack et al. 1991; Reddy and Hahn 1991). However, overexpression of the TBP core domain inhibits cell growth, particularly on nonfermentable carbon sources (Gill and Tjian 1991; Zhou et al. 1991), and this toxicity is suppressed by concurrent overexpression of Std1 (Msn3) (Ganster et al. 1993). Std1 directly associates with TBP, and deletion of the N-terminal region qualitatively alters this interaction (Tillman et al. 1995). Apart from these studies, however, relatively little is known about the function of the TBP N-terminal region in vivo.

To examine the physiological role of the N-terminal region of yeast TBP, we deleted this region in the context of a large number of yeast TBP mutants. Our results suggest that the N-terminal region of TBP has at least two distinct functions in vivo. It inhibits the interaction of TBP with TATA elements, and it functions in concert with a specific surface of TBP that presumably interacts with another transcriptional regulatory protein.

MATERIALS AND METHODS

DNA molecules: TBP mutants used in this study have been described previously (Cormack and Struhl 1993; Lee and Struhl 1995, 1997). The N-terminally deleted derivatives in Table 1 derive from pBC83, a TRP1-marked centromeric plasmid with the 2.4-kb EcoRI-BamHI fragment containing the yeast TBP gene with a deletion of residues 4–63 (Cormack et al. 1991). The N-terminally deleted derivatives in Figure 2 derive from pML3095, a derivative of pBC83 in which a FLAG epitope was engineered at the N-terminus of the TBP core domain by subcloning of annealed oligonucleotides. Serial deletions of the N-terminal region were generated by PCR using the wild-type TBP gene as template.

Phenotypic analyses: TBP derivatives were assayed for their ability to support cell growth by spotting 10^4 and 10^5 cells on glucose medium supplemented with casamino acids and 5-fluoroorotic acid, using the plasmid shuffle assay and strain BYΔ2 (Cormack et al. 1991); growth was monitored after 2–3 days incubation at 30°. SI nuclease protection assays for measuring RNA levels and β-galactosidase assays for determining Gal4-dependent activation were performed as described elsewhere (Cormack and Struhl 1992; Lee and Struhl 1995; Iyer and Struhl 1996). Intracellular levels of N-terminally deleted TBPα were determined by Western blotting using monoclonal antibody mAb58C9 (Santa Cruz Biotechnologies) against the FLAG epitope. To avoid the confounding effects of homeostatic mechanisms on TBP levels, mutant TBP levels were measured in a strain containing hemagglutinin-tagged TBP (Lee and Struhl 1997).

FIGURE 1.—N-terminal truncation can rescue the transcriptional phenotypes of a subset of activation-deficient DNA-binding mutants of yeast TBP. β-Galactosidase activity of a Gal4-dependent LacZ reporter in wild-type and mutant TBP strains grown with galactose as the sole carbon source (inducing conditions) is shown. Standard errors are given as error bars. Solid bars, full-length TBP derivatives; stippled bars, N-terminally truncated derivatives (Δ4-63).

RESULTS

Deletion of the N-terminal region rescues the activation-deficient phenotype of a subset of DNA-binding mutants: Certain TBP mutants deficient in the response to transcriptional activators contain mutations on the DNA-binding surface that dramatically reduce TATA-element binding in vitro (Kim et al. 1994; Arndt et al. 1995; Lee and Struhl 1995). As removal of the N-terminal region can restore TBP-TATA association in vitro for some TBP mutants with DNA-binding defects (Lee et al. 1992), we examined the genetic properties of N-terminally deleted versions of three TBP mutants deficient for DNA binding and transcriptional activation (Lee and Struhl 1995). All three N-terminally deleted derivatives support yeast cell growth at levels comparable to their full-length counterparts (Table 1). As previously reported (Lee and Struhl 1995), the F148L, N159L, and V161A derivatives are compromised for Gal4-dependent activation when compared to wild-type TBP (39, 42, and 8% of wild-type activity, respectively; Figure 1). Removal of the N-terminal region from wild-type TBP results in a 40% decrease in Gal4-dependent activation, in accord with previous results (Cormack et al. 1991; Reddy and Hahn 1991). By contrast, N-terminal deletion confers increased transcriptional activation by the N159L, V161A, and perhaps the F148L derivatives.

Deletion of the N-terminal region of TBP does not affect HIS3 TATA-element utilization: The results above and previous biochemical experiments indicate that the N-terminal region can act as an inhibitor of TBP function. For this reason, we examined whether the N-terminal region of TBP affects TATA-element utilization at the HIS3 promoter in a manner similar to that of other general inhibitors of TBP function such as Mot1 (Gollart 1996), NC2 (Lemaire et al. 2000), and TAF130.
Functions of the TBP N-Terminal Region

### TABLE 1

<table>
<thead>
<tr>
<th>TBP derivative</th>
<th>Biochemical defect</th>
<th>Viability as</th>
<th>Full length TBP</th>
<th>ΔN terminus</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wild type (TBP⁺)</td>
<td>None</td>
<td>++</td>
<td>++</td>
<td></td>
</tr>
<tr>
<td>L114K</td>
<td>DNA binding</td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>S118L</td>
<td>DNA binding</td>
<td>+++</td>
<td>++</td>
<td></td>
</tr>
<tr>
<td>N159L</td>
<td>DNA binding</td>
<td>++</td>
<td>++</td>
<td></td>
</tr>
<tr>
<td>V161A</td>
<td>DNA binding</td>
<td>+++</td>
<td>+++</td>
<td></td>
</tr>
<tr>
<td>F148L</td>
<td>DNA binding</td>
<td>+++</td>
<td>+++</td>
<td></td>
</tr>
<tr>
<td>D130A</td>
<td>TFIIA interaction</td>
<td>+++</td>
<td>++</td>
<td></td>
</tr>
<tr>
<td>R137A</td>
<td>TFIIA interaction</td>
<td>+++</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>D130A, R137A</td>
<td>TFIIA interaction</td>
<td>++</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>Y139A</td>
<td>TFIIA interaction</td>
<td>+++</td>
<td>+++</td>
<td></td>
</tr>
<tr>
<td>E186A</td>
<td>TFIIB interaction</td>
<td>–</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>E188A</td>
<td>TFIIB interaction</td>
<td>+++</td>
<td>+++</td>
<td></td>
</tr>
<tr>
<td>L189K</td>
<td>TFIIB interaction</td>
<td>–</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>L134A, E222A</td>
<td>TFIIF, Pol II interaction</td>
<td>+++</td>
<td>++</td>
<td></td>
</tr>
<tr>
<td>R137W</td>
<td>TDS4 interaction</td>
<td>++</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>A140R</td>
<td>TDS4 interaction</td>
<td>++</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>F152E</td>
<td>TDS4 interaction</td>
<td>+++</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>F155S</td>
<td>TDS4 interaction</td>
<td>+</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>I160H</td>
<td>TDS4 interaction</td>
<td>+++</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>G162Y</td>
<td>TDS4 interaction</td>
<td>++</td>
<td>++</td>
<td></td>
</tr>
<tr>
<td>R220A</td>
<td>TDS4 interaction</td>
<td>++</td>
<td>++</td>
<td></td>
</tr>
<tr>
<td>Y224K</td>
<td>TDS4 interaction</td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>Y231V</td>
<td>TDS4 interaction</td>
<td>++</td>
<td>++</td>
<td></td>
</tr>
<tr>
<td>R238D</td>
<td>TDS4 interaction</td>
<td>++</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>A135T</td>
<td>Decreased Pol I, Pol II</td>
<td>++</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>K138W</td>
<td>TFIIA±</td>
<td>++</td>
<td>–</td>
<td></td>
</tr>
</tbody>
</table>

(Moqtaderi et al. 1996). The HIS₃ promoter contains a noncanonical TATA-like element (Tₐ) that is responsible for initiation from the +1 site and a consensus TATA element (Tᵢ) that is responsible for initiation from the +13 site (Iyer and Struhl 1995). Loss of Mot1, NC2, or TAF130 results in a dramatic decrease in Tᵢ-dependent transcription from the +1 site, but does not affect Tₐ-dependent transcription from the +13 site. In contrast, deletion of the N-terminal region of TBP does not affect the pattern of transcriptional initiation, and hence TATA-element utilization, at the HIS₃ promoter region (Figure 2). In addition, the N-terminally deleted version of TBP has minimal effects on transcription by RNA polymerases I and III. Thus, by the criterion of TATA-element utilization at the HIS₃ promoter, the N-terminal region of TBP behaves differently from other proteins that can function as inhibitors of TBP function.

Mutations within the core domain of TBP render the N-terminal domain essential for viability: In addition to its negative effect on DNA binding, the N-terminal region of TBP has been postulated to participate in protein-protein interactions with other components of the transcription apparatus (LesCure et al. 1994; Zhou and Berk 1995; Goppelt and Meisterernst 1996). Deletion of the N-terminal region has minimal effects on growth and on transcription by all three nuclear RNA polymerases. Thus, if the N-terminal region mediates physiologically important functions, such functions must be also performed by another region of TBP or by another protein(s).

To search for such potential redundant functions, we constructed N-terminally deleted versions of a panel of TBP mutants with specific defects for interactions with TFIIA (Stargell and Struhl 1995; Lee and Struhl 1997), TFIIB (Kim et al. 1994; Lee and Struhl 1997), TFIIF, RNA polymerase II (Lee and Struhl 1997), TFIIF, RNA polymerase III factor Brf1 (Cormack and Struhl 1993). Deletion of the N-terminal region from TBP mutants deficient for interaction with DNA, TFIIA, TFIIF, and RNA polymerase II produces only modest changes in growth (Table 1). In contrast, N-terminal deletion of a subset of mutations involving the TFIIA-

[548x755]
TABLE 2
Viability of N-terminally truncated TBP derivatives with and without an N-terminal FLAG epitope

<table>
<thead>
<tr>
<th>TBP derivative</th>
<th>Full-length TBP</th>
<th>ΔN terminus</th>
<th>FLAG-ΔN terminus</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wild type (TBP)</td>
<td>+++</td>
<td>+++</td>
<td>+++</td>
</tr>
<tr>
<td>A135T</td>
<td>++</td>
<td>–</td>
<td>+</td>
</tr>
<tr>
<td>K138W</td>
<td>++</td>
<td>–</td>
<td>++</td>
</tr>
<tr>
<td>R238D</td>
<td>++</td>
<td>–</td>
<td>+</td>
</tr>
<tr>
<td>R137A</td>
<td>+++</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>A140R</td>
<td>++</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>F152E</td>
<td>+++</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>F155S</td>
<td>++</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>I160H</td>
<td>+</td>
<td>–</td>
<td>–</td>
</tr>
</tbody>
</table>

level achieved by N-terminally deleted wild-type TBP (Figure 3).

N-terminal-dependent mutations map to a limited surface of TBP and define a new TBP function: The mutations that confer dependence on the N-terminal domain lie in close proximity within the three-dimensional structure of the TBP core domain (Figure 4). Specifically, the five positions define a limited surface on the convex face of TBP that lines the inner portion of the groove formed by helix H2, helix H2', and the segments preceding strand S1 and connecting helix H2 and strand S1'. Amino acid substitutions at neighboring positions where the side chains project away from this putative nuclease protection assays for determination of H153, DED1, rRNA, and tRNA transcript levels in yeast strains supported by either full-length or N-terminally deleted (∆4-63) TBP are shown. All bands in the right lane are ~50% higher in intensity than in the left lane and likely represent a difference in total RNA used for each assay.

and Brf1-interaction surfaces results in lethality. Although removal of the N-terminal domain does not destabilize wild-type TBP (Cormack et al. 1991; Reddy and Hahn 1991; Ganster et al. 1993), it might cause protein instability of TBP derivatives with mutations within the core domain. To address this possibility, FLAG-tagged versions of these inviable N-terminally deleted TBP mutants were generated to facilitate detection of their protein levels in vivo. Unexpectedly, three of these mutants (A135T, K138W, and R238D) are viable in the context of this N-terminal FLAG epitope tag (Table 2). The ability of an unrelated peptide (i.e., FLAG) to functionally substitute for the N terminus in these three mutants suggests that these mutants, in the absence of the N terminus, are unstable or misfolded. The remainder of the inviable N-terminally truncated TBP mutants, however, are inviable in the context of the FLAG epitope, and their in vivo protein levels (with the possible exception of F152E) are comparable to the

Figure 2.—Deletion of the N terminus of TBP does not significantly alter Pol I, II, or III transcription in vivo. S1 nuclease protection assays for determination of H153, DED1, rRNA, and tRNA transcript levels in yeast strains supported by either full-length or N-terminally deleted (∆4-63) TBP are shown. All bands in the right lane are ~50% higher in intensity than in the left lane and likely represent a difference in total RNA used for each assay.

Figure 3.—Western blot analysis of inviable N-terminally truncated TBP mutants. Whole-cell extracts from yeast strains supported by wild-type TBP and coexpressing the indicated TBP derivatives were analyzed with monoclonal antibody mAb58C9. *, cross-reacting band.
Functions of the TBP N-Terminal Region

Figure 5.—Sequential truncation of the N terminus of TBP. Viability of TBP derivatives with successive deletions of the N-terminal domain was assessed by growth on medium containing 5-fluoroorotic acid (FOA) in a plasmid shuffle assay. R137A and A140R respectively affect the TFIIA- and Brf1-interaction surfaces.

**DISCUSSION**

A role for the N-terminal domain in DNA binding by TBP in vivo: The association of TBP with the TATA element is a crucial regulatory step in the formation of productive transcription complexes in vivo. Direct recruitment and stabilization of TBP at the promoter through fusions with heterologous DNA-binding domains activates transcription (Chatterjee and Struhl 1995; Klages and Strubin 1995; Xiao et al. 1995), and natural transcriptional activators enhance TATA occupancy by TBP at chromosomal promoters (Klein and Struhl 1994; Kuras and Struhl 1999; Li et al. 1999). A subset of TBP mutations that selectively impair activated transcription maps to the TBP-TATA interface (Kim et al. 1994; Arndt et al. 1995; Lee and Struhl 1995). In this report, we demonstrate that removal of the absence of a particular interaction(s) normally mediated by the core domain of TBP, at least one function of the N-terminal region becomes essential for yeast cell growth. To localize the region within the N terminus that performs this function, we analyzed a series of successive deletions within the N-terminal region in the context of two TBP mutants that require the N-terminal domain: R137A (TFIIA-interaction mutant) and A140R (Brf1-interaction mutant). Remarkably, all but the final 10 amino acids of the N-terminal domain could be removed without affecting the viability of these two TBP mutants (Figure 5). This 10-amino-acid segment likely encodes a specific function, as an unrelated sequence (FLAG) could not be functionally substituted (Table 2).

A 10-amino-acid segment of the N-terminal region is sufficient to restore viability to TBP mutants dependent on the N terminus: The above results suggest that, in

Type (Table 1). Second, Y139A, which is 20–30 times more deficient for TFIIA interaction than D130A, R137A (Lee and Struhl 1997), is unaffected by removal of the N terminus. Third, overexpression of Brf1 fails to suppress the requirement for the N-terminal domain in these mutants (data not shown), even though it does suppress the Pol III defect (Cormack and Struhl, 1993). Std1 (Msn3) interacts directly with yeast TBP in vitro and in vivo, and this interaction is modulated by the N-terminal domain of TBP (Tillman et al. 1995). However, neither overexpression nor deletion of STD1 relieves the N-terminal dependence of these TBP mutants (data not shown).

A 10-amino-acid segment of the N-terminal region is sufficient to restore viability to TBP mutants dependent on the N terminus: The above results suggest that, in...
the N-terminal region from these DNA-binding mutants of TBP can partially restore their activation competency. This result provides evidence that the inhibitory effect of the TBP N-terminal domain on DNA binding, long recognized in vitro (Horikoshi et al. 1990; Lieberman et al. 1991; Lee et al. 1992; Kuddus and Schmidt 1993), has physiological relevance.

More generally, our results indicate that the N-terminal region of TBP has an autoinhibitory role in vitro. In otherwise wild-type cells, loss of this autoinhibitory function does not significantly affect cell growth or general transcriptional functions of TBP. In part, the minimal phenotype caused by loss of the TBP N-terminal region might be due to functional redundancy with other negative regulators of TBP function such as NC2 (Gadbois et al. 1997; Kim et al. 1997, 2000; Prelieva 1997; Lemaire et al. 2000; Xie et al. 2000), Mot1 (Auble et al. 1994, 1997; Collart 1996; Madison and Winston 1997), the N terminus of TAF145 (Collart et al. 1998), and the NOT complex (Collart and Struhl 1994; Collart 1996). Nevertheless, we suspect that loss of this autoregulatory function might result in the generation of a hyperfunctional TBP, thereby explaining why overexpression of the TBP core domain strongly inhibits cell growth even in the presence of wild-type TBP (Gill and Tjian 1991; Zhou et al. 1991).

Functional redundancy between the N terminus and a surface of the TBP core domain: We examined the effect of N-terminal deletion in the context of TBP mutations representing 25 unique positions with the core domain. Mutations at only 5 of those positions yield a lethal phenotype when combined with the N-terminal deletion (excluding the mutants that are rescued by the presence of an N-terminal FLAG epitope). Strikingly, these 5 positions define a limited surface of TBP that lines the inner portion of the groove formed by helix H2, helix H2', and the segments preceding strand S1 and connecting helix H2 and strand S1'. This synthetic lethality between TBP mutations lacking the N-terminal region and those affecting a specific surface in the core domain provides genetic evidence for functional redundancy between these two regions of TBP.

These observations provide evidence both for a positive function for the N-terminal region of TBP and for a previously unidentified surface of the TBP core domain that interacts with a transcriptional regulatory protein(s). The factor(s) that interacts with this surface of the TBP core domain is unknown, although TFIIA, TFIIIB, and Brf1 do not appear to be strong candidates. A role for the TBP N terminus in interactions with other transcription factors is consistent with observations in vitro in which the presence of the N-terminal domain affects the association of TBP with NC2 (Goppelt and Meisterernst 1996), TAFs (Zhou and Berk 1995), Std1 (Tillman et al. 1995), and the SNAP complex (Mittal and Hernandez 1997).

We consider two models by which the N-terminal region of TBP performs a positive function. In one model, the N-terminal domain stabilizes the interaction between TBP and the factor that interacts with the surface of the core domain identified by virtue of the synthetic lethal effects described above. The role of the N-terminal region of human TBP in mediating cooperative binding with the SNAP complex at the U6 promoter (Mittal and Hernandez 1997) is a precedent for such a model, although the SNAP complex and its target site (PSE) do not exist in yeast cells. In a second model, the apparent positive role of the N-terminal region might actually be a negative function that is redundant with a distinct negative regulatory function that is mediated by the limited surface of the TBP core domain. In this view, synthetic lethality results from the simultaneous elimination of multiple negative regulators of TBP function. In accord with this model, the region immediately adjacent to yeast TBP core domain is required both for the synthetic lethal interactions observed here (Figure 5) and for the toxicity observed upon overexpression of the core domain (Zhou et al. 1991). By either model, our results and previous observations (Zhou and Berk 1995; Goppelt and Meisterernst 1996; Mittal and Hernandez 1997) suggest that the evolutionarily divergent N-terminal region coevolved with other transcriptional regulators to mediate certain species-specific functions of TBP.

We thank Martin Schmidt for providing STD constructs, Laurie Stargell for helpful discussions at the initial stages of this work, and Lisete Fernandes and Ada Garcia for assistance in the preparation of this manuscript. We also thank Susanna Chou, Sukalyan Chatterjee, and Irene Wu for their discussions and critical reading of the manuscript. This work was supported by a predoctoral fellowship to M.L. from the Howard Hughes Medical Institute and by research grants to K.S. from the National Institutes of Health (GM 30186 and GM 53720).

LITERATURE CITED


Functions of the TBP N-Terminal Region


Communicating editor: M. Hampsy