

Figure S1. Structural similarity between Spn1 core and proteins that bind RNAPII CTD and conservation of amino acid sequences. Related to Figure 2.

(A) An alignment of Spn1 (gray) and Pcf11 (yellow/green). Structures of the Spn1 core and Pcf11 (Meinhart and Cramer, 2004) and SCAF8 (Becker et al., 2008) were aligned with DALI Lite (Holm and Park, 2000). A population of overlaps of both proteins with Spn1 were obtained that typically displayed Z scores ~ 6 and RMSD values of 3-4 Å for overlap on ~ 100 residues. Sequence identity after structural alignment is just 5-10%. Alignments on SCAF8 are essentially identical.

(B) Spn1 surface with the Pcf11-bound RNAPII CTD peptide shown after overlap on the proteins. The Spn1 K192 pocket that has been implicated in interactions with RNAPII (Zhang et al., 2008) is indicated. As indicated in the main text, despite the structural similarity, we did not detect measurable binding of CTD peptides with Spn1.

(C-D) Alignments of homologs of (C) the Spn1 core and (D) the fragment of Spt6 that binds Spn1. S.c. *Saccharomyces cerevisiae*; S.p. *Schizosaccharomyces pombe*; C.e. *Caenorhabditis elegans*; D.m. *Drosophila melanogaster*; D.r. *Danio rerio*; H.s. *Homo sapiens*. Secondary structure is indicated above. Residues that make contact across the interface are indicated with a black square or with an asterisk if they were mutated in this study. Numbering refers to the *S. cerevisiae* proteins. Coloring represents degrees of conservation: dark green and red (high), light green and yellow (medium). Amino acid sequences were aligned using the T-coffee multiple sequence alignment method (Notredame et al., 2000) and slightly adjusted manually in light of the structure. Secondary structure designation was by ESPript (Gouet et al., 2003).