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L.C. Sieker[1], M. Holmes[2], I. Le Trong[1], S. Turley[1], B.D. Santarsiero[3], M.-Y. Liu[4], J. LeGall[4,5] and R.E. Stenkamp[1]

[1]Department of Biological Structure and Biomolecular Structure Center, Box 357420, University of Washington, Seattle, Washington 98195-7420. [2]C3-168 Molecular Medicine, Fred Hutchinson Cancer Research Center, 1100 Fairview Ave. N., Seattle, Washington 98109. [3]504 Stanley Hall, Physical BioSciences Division, E.O. Lawrence Berkeley National Laboratory, Berkeley, California 94720. [4]Department of Biochemistry and Molecular Biology, The University of Georgia, Athens, Georgia 30602-7229. [5]Instituto de Tecnologia Quimica e Biologica, Universidade Nova de Lisboa, Apartado 127, 2780 Oeiras, Portugal.

Correspondence should be addressed to R.E.S. email: stenkamp@u.washington.edu

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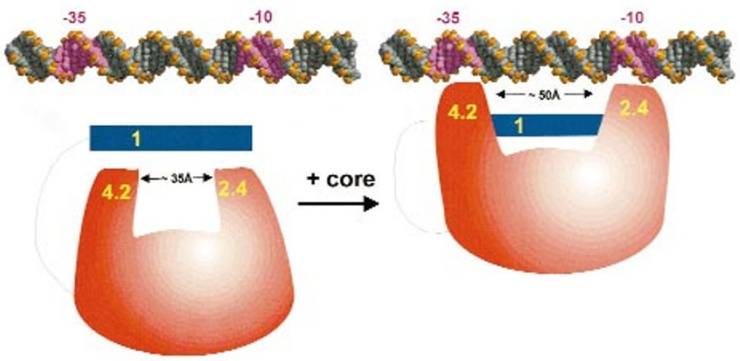
Move over and bind

In bacteria, all RNA synthesis is carried out by a multisubunit enzyme composed of two small α subunits, large β and β' subunits, and the σ subunit, of which there are several varieties. It exists in two forms — the core enzyme ($\alpha_2\beta\beta'$) and the holoenzyme ($\alpha_2\beta\beta'\sigma$). While the core polymerase can elongate RNA, only the holoenzyme can initiate transcription at specific promoter sites. The promoter consists of two highly conserved sequences (-10 and -35 with respect to the transcription start site +1), each six nucleotides long and separated from each other by ~17 nucleotides. Thus, the σ subunit recognizes the promoter but only when it is bound to the core enzyme. The σ^{70} subunit is composed of a conserved autoinhibitory N-terminal domain (region 1) and two DNA binding domains (regions 2.4 and 4.2) that are involved in recognition of the -10 and -35 promoter elements. These and other findings have led to a model in which region 1 sterically blocks access of the DNA binding domains of σ^{70} to promoter DNA in the absence of core polymerase. Binding of the core enzyme to σ^{70} was proposed to induce movement of region 1 to unmask the DNA binding domains of σ^{70}. A schematic diagram of this conformational change is depicted, but no structural details are known.



To test whether core enzyme does induce conformational changes in σ^{70}, Heyduk and coworkers (Callaci, S., Heyduk, E., & Heyduk, T. *Mol. Cell* **3**, 229–238; 1999) used a combination of site-directed mutagenesis and luminescence resonance energy transfer measurements to introduce luminescence donors and acceptors in different conserved regions of the protein and monitor the distances between them in the free σ^{70} and in the holoenzyme. Their studies have provided direct evidence for a major movement of region 1 induced by core binding. Region 1 moves by ~20 Å away from the DNA binding domains. In addition, their experiments have suggested another way in which core binding could regulate promoter recognition by σ^{70}.

In the absence of core, regions 2.4 and 4.2 of σ^{70} are too close together to interact simultaneously with the -10 and -35 regions on the DNA. The distance between regions 2.4 and 4.2 is increased by ~15 Å upon core binding which is more compatible with binding to the promoter. Thus core binding to σ^{70} has at least two effects with regard to promoter binding: it displaces the autoinhibitory region 1 and it increases the spacing between the two DNA binding domains. Both of these changes increase the affinity of the holoenzyme for the promoter DNA.

One interesting question is whether the movements observed within σ^{70} upon core binding are interdependent. For example, does the modulation of spacing between DNA binding regions occur in the absence of region 1? If not, can this movement be restored if domain 1 is added back in trans? Answers to these questions would address the role of region 1 in promoter recognition by regions 2.4 and 4.2. In one scenario, region 1 holds the DNA binding domains together in the absence of core and when core binds they spring apart. Alternatively, region 1 could push the DNA binding regions apart upon core binding. Clearly, even in this 'simple' system, many important dynamic properties have yet to be revealed.

Boyana Konforti

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