Supporting information for Leng and McMacken (2002) *Proc. Natl. Acad. Sci. USA*, 10.1073/pnas.142002099.

Table 1. Effect of specific DNA-binding proteins on transcription-induced DNA supercoiling by T7 RNA polymerase*

Protein(s) added	DNA template	Binding sites†	Mass, kDa‡	K_d, nM§	DNA-bending angle‖	Stimulation of supercoiling**	Ref.††
λ O	pRLM375	4	270	3	85°	++++	1
λ O	pRLM411	1	67	3	85°	+	1
λ O	pRLM412	2	135	3	85°	++	1
λ O_N	pRLM375	4	111	5	74°	++++	1
*Eco*RI	pRLM375	1	62	0.002	20°	+	2, 3
E.coli RNAP + rif	pRLM375	≥3	≥1347	0.01	60–70°	+++	4, 5
λ *cI* repressor	pRLM389	3	156	3	40°	+	6, 7
λ *cI* repressor	pRLM409	6	313	3	40°	+++	6, 7
GalR	pRLM419	2	148	4	105°	++++	8, 9
GalR + galactose	pRLM419	2	148	3	ND	–	8
LacI	pRLM420	2	154	3×10^{-5}	90°	++++	10, 11
LacI + IPTG	pRLM420	2	154	0.03	ND	–	10
λ O + *Eco*RI	pRLM411	2	130	¶	¶	++++	1, 2
λ O + *Eco*RI	pRLM412	3	197	¶	¶	++++	1, 2

*Standard transcription-supercoiling reactions were performed with T7 RNA polymerase and DNA gyrase as described under *Materials and Methods*, with the exception that the plasmid DNA templates were varied as indicated and the reaction mixtures were supplemented with the specified site-specific DNA-binding proteins. After completion of each reaction, the plasmid DNA template was electrophoresed through 1% agarose gels containing 5 μg/ml of chloroquine. The amount of hypernegatively supercoiled DNA produced during the reaction was estimated. Abbreviations: O_N, N-terminal domain of λ O protein; RNAP, RNA polymerase; rif, rifampicin; GalR, *E. coli* galactose operon repressor; LacI, *E. coli* lactose operon repressor; *Eco*RI, *Eco*RI-Gln111 active site mutant of *Eco*RI restriction endonuclease.

†The number of specific recognition sites present on the DNA template for the indicated site-specific DNA-binding protein(s) is listed.

‡The total mass in kDa of specific nucleoprotein structures that are assembled on the template DNA is listed. These masses are exclusive of the masses of transcribing RNAP molecules and DNA gyrase.

§The equilibrium dissociation constant of the added specific DNA-binding protein for a single recognition site.

¶See individual listings for λ O and *Eco*RI in this table.

‖The estimated angle of bending of the DNA helical axis upon binding of the added site-specific binding protein to a single recognition sequence. ND, not determined.

**Estimated stimulation of the formation of hypernegatively supercoiled DNA by the added site-specific DNA-binding protein(s). ++++, maximal stimulation; +++, strong stimulation; ++, moderate stimulation; +, weak stimulation; – , no stimulation. See individual Figs. 2 and 5 for representative examples across a wide range of stimulation levels.

††References for the equilibrium dissociation constants and DNA-bending angles for the protein-DNA interactions of the specified DNA-binding proteins.

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