

# Supplement for: Correlation Between Binding Rate Constants and Individual Information of *E. coli* Fis Binding Sites

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This document contains:

1. Fis binding sequences used in EMSA experiments - Fig. 1
2. Graphs of Fis binding rates vs. information - Fig. 2

## Computation of $K_D$ values

See reference [1] for definitions of symbols, methods and the equations used. The competitor EMSA experiments were repeated three times with different competitor concentrations ( $C_0$ ).  $K_{Dw}$  and  $R_0$  were simultaneously adjusted to fit two criteria. First, the  $K_D$  of the Hin-180 oligo was made to match the published value of Hin-D,  $2 \times 10^{-9}$  M [2]. Second, the  $K_{Dw}$  constant was made to match the computed  $K_D$  of the **con** oligo. The top and bottom bands (Figure 5) were quantitated using the ImageQuant program (Molecular Dynamics, Sunnyvale, CA). The  $K_{Dc}$ s were computed using the method given in [1] and the average  $K_{Dc}$ s and standard deviations were computed from the three values.

Additional supplementary files are:

- Fis-koff-raw-data.xls - The raw SPR data.
- Fis-kd-raw-data.xls - The raw EMSA data.
- Fis-kon-koff-kd-ri.xls - Combination of SPR and EMSA data, graphed in Fig. 2.

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## References

- [1] I. G. Lyakhov, P. N. Hengen, D. Rubens, and T. D. Schneider. The P1 Phage Replication Protein RepA Contacts an Otherwise Inaccessible Thymine N3 Proton by DNA Distortion or Base Flipping. *Nucleic Acids Res.*, 29(23):4892–4900, 2001.  
<http://www.ccrnp.ncifcrf.gov/~toms/paper/repan3/>.
- [2] C. Q. Pan, S. E. Finkel, S. E. Cramton, J. A. Feng, D. S. Sigman, and R. C. Johnson. Variable structures of Fis-DNA complexes determined by flanking DNA-protein contacts. *J. Mol. Biol.*, 264:675–695, 1996.

Sequences used for EMSA Experiments

#	Ri (bits)	Name	Sequence
1	-30.6	anti-con	CTGACCCCGGGTCAGGCGAAGCCTGACCCGGGGTCAG
3	4.9	cin-336	GTCTAACTTCCATACGCGAAGCGTATGGAAGTTAGAC
2	5.4	hin-1096	GAACAAATCCCAGTCGCGAAGCGACTGGGATTTGTTC
4	6.6	lacP-560	GCTCACTCATTAGGCGCGAAGCGCCTAATGAGTGAGC
6	8.2	ndhII-188	GCCTATCTTTTCAGCGCGAAGCGCTGAAAAGATAGGC
7	8.2	comp-ndhII-188	GCTGAAAAGATAGGCGCGAAGCGCCTATCTTTTCAGC
9	10.1	fis-333	GGTCAAAGTTTGGCCGCGAAGCGCCAAACTTTGACC
8	10.2	tgt-1824	GCTAAAAAATTCATCGCGAAGCGATGAATTTTTTAGC
5	10.4	hin-180	GGTCACAATTTGCACGCGAAGCGTGCAAATTTGTGACC
10	10.9	thrU-87	GGTCACATTTTATGCGCGAAGCGCATAAAATGTGACC
11	12.7	ndhI-137	GCTCAAATAATAAACGCGAAGCGTTTATTATTTGAGC
12	12.8	mut-con	GGTTAAATGTTAACCGCGAAGCGGTTAACATTTAACC
13	14.9	con	GGTTAAATTTTAACCGCGAAGCGGTTAAAATTTAACC

Figure 1: Fis binding sequences used in EMSA experiments.

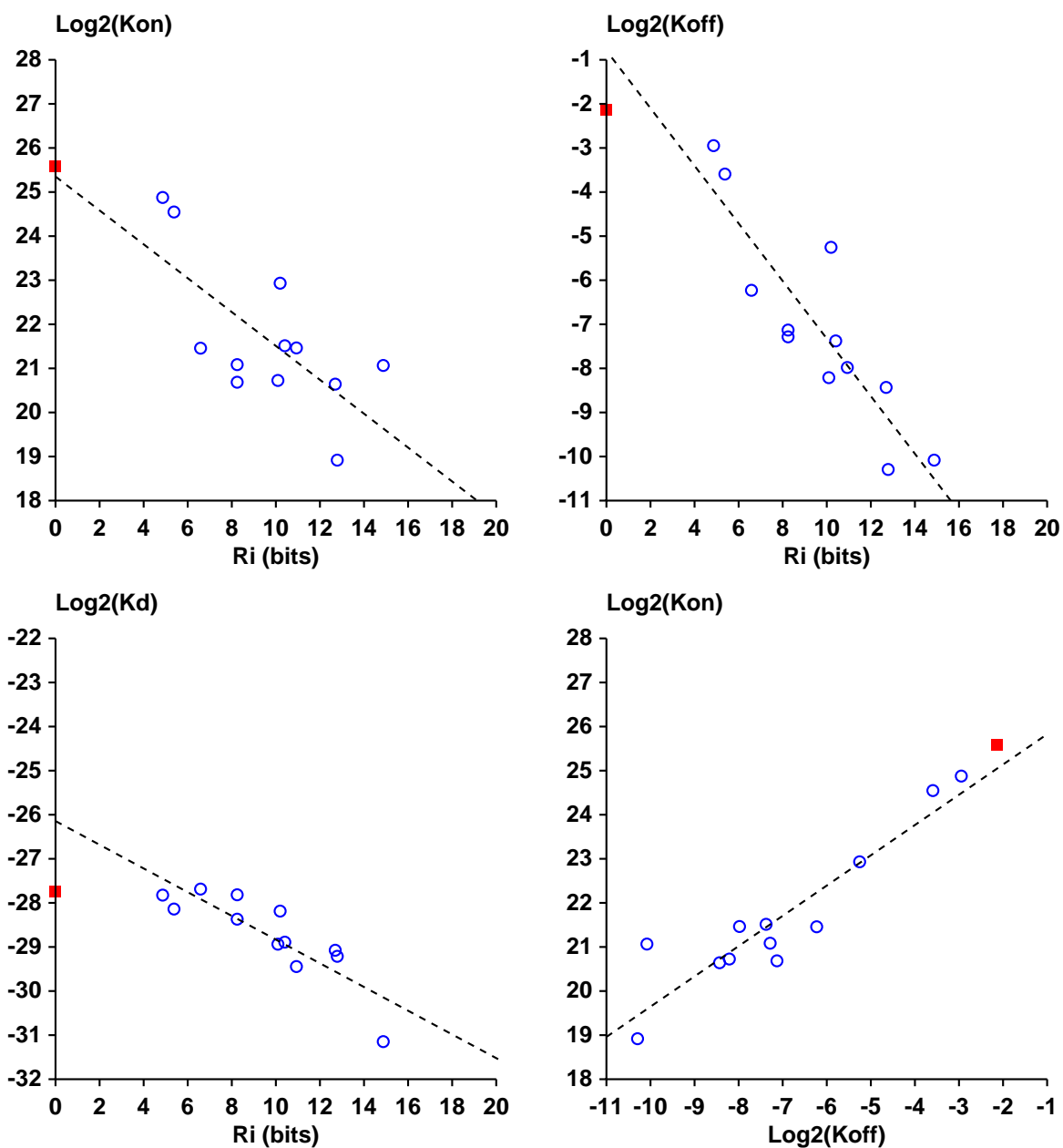


Figure 2: Fis binding rates versus information.

The axes of each rate graph always covers 10 powers of 2 so that the slopes of the regression lines can be compared.  $k_{off}$  values were averaged for each oligo. The anti-con oligo is plotted at 0 bits as a square but was not included in the regressions.