Supporting Information

Sue et al. 10.1073/pnas.1014323108

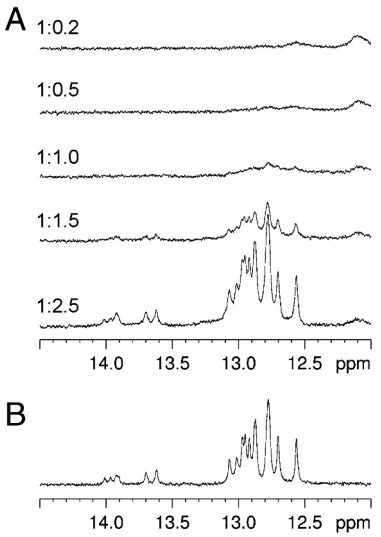

Fig. S1. Imino proton NMR spectra of the DNA duplex. (*A*) One-dimensional ^{1}H NMR spectra of solutions containing the indicated molar ratios of DNA added to a 0.12 mM solution of the complex of deuterated p50(39–350)/p65(19–321) with deuterated, ^{15}N-labeled IκBα(67–287). (*B*) One-dimensional ^{1}H NMR spectrum of the 20-nucleotide DNA duplex free in solution in the absence of protein.

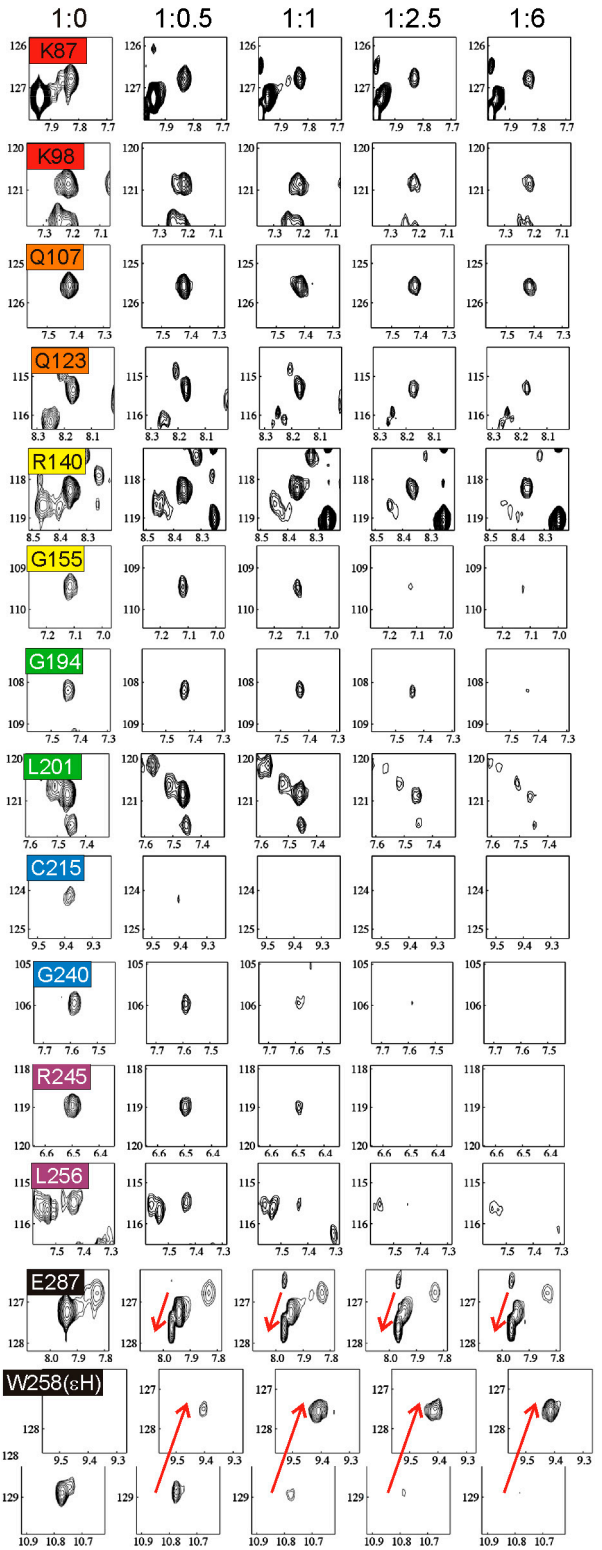

Fig. S2. Effects of DNA binding for individual ankyrin repeats. (*A*) Representative cross-peaks from the 900 MHz ^{1}H-^{15}N transverse relaxation optimized heteronuclear single-quantum coherence spectra of NF-κB-bound IκBα are color coded as in Fig. 1*B* and shown (plotted at the same contour levels in each panel) at different molar ratios of protein complex to DNA.

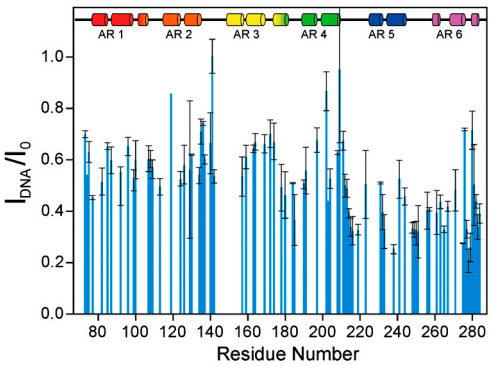

Fig. S3. Effect of DNA binding on resonance intensities of IκBα. Histogram showing experimental per-residue intensity ratios (I_{DNA}/I_0), where I_{DNA} represents the intensity of an IκBα resonance in complex with NF-κB p50(39–350)/p65(19–321) following addition of a 1:1 molar ratio of DNA and I_0 represents the intensity of the same IκBα resonance in complex with NF-κB p50(39–350)/p65(19–321) in the absence of DNA. The α-helices identified for the IκBα–NF-κB complex (1, 2) are indicated at the top for comparison. Data for overlapped resonances are omitted from the analysis.

1 Jacobs MD, Harrison SC (1998) Structure of an IκBα/NF-κB complex. *Cell* 95:749–758.
2 Huxford T, Huang DB, Malek S, Ghosh G (1998) The crystal structure of the IκBα/NF-κB complex reveals mechanisms of NF-κB inactivation. *Cell* 95:759–770.

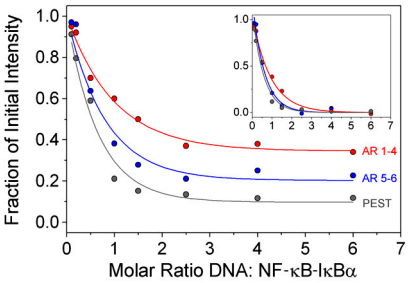

Fig. S4. Fraction of IκBα remaining in bound to NF-κB as DNA is added. Average decay data for the intensities of resonances in three portions of IκBα, ankyrin repeats (ARs) 1–4 (red), AR 5–6 (blue), and the region rich in proline, glutamate, serine, and threonine (PEST) (gray) are plotted as a function of DNA concentration. The decay curves were fitted into the first-order exponential decay function and extrapolated back to a normalized intensity of 1 in the absence of DNA where the fitted rate constant $R = 0.92$, 0.60, and 0.57 for AR-14, AR-5–6, and PEST, respectively. (*Inset*) The same curves plotted by aligning the final equilibrium state to the value of 0.