Supplemental Data

STRUCTURE OF RecJ EXONUCLEASE DEFINES ITS SPECIFICITY FOR SINGLE-STRANDED DNA

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Figure S1

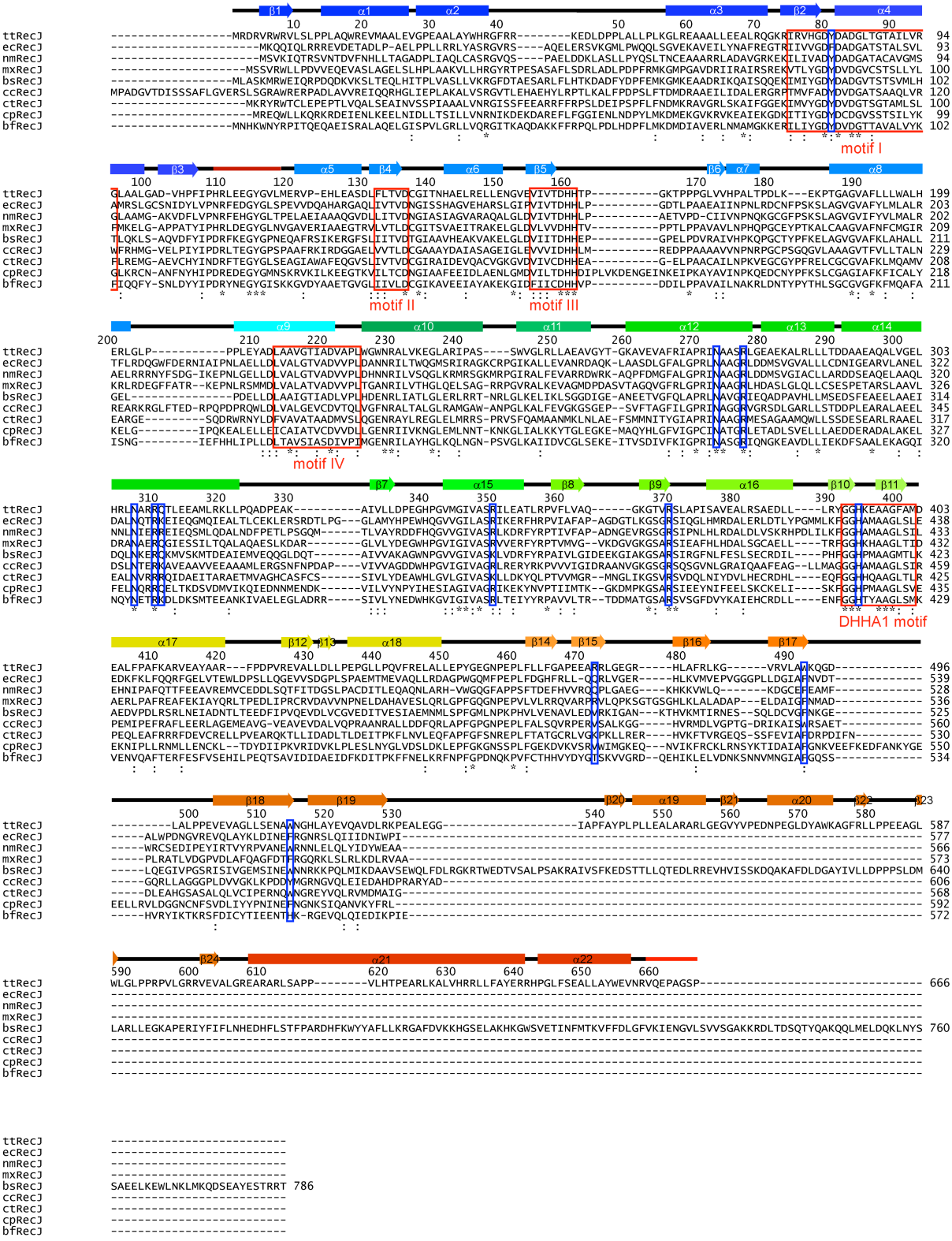

FIGURE S1. **Sequence alignments of ttRecJ and its homologs.** The secondary structures of ttRecJ are shown above the sequences; the horizontal bars indicate α-helices and arrows indicate β-strands. Red lines indicate disordered regions. The five motifs proposed by Aravind and Koonin (1) are

designated as red boxes. The possible residue for binding to ssDNA is shown by a blue box. Sequences shown are as follows: ttRecJ (*T. thermophilus* HB8), ecRecJ (*E. coli*), nmRecJ (*Neisseria meningitidis*), mxRecJ (*Myxococcus xanthus*), bsRecJ (*Bacillus subtilis*), ccRecJ (*Caulobacter crescentus*), ctRecJ (*Chlorobium tepidum*), cpRecJ (*Clostridium perfringens*), and bfRecJ (*Bacteroides fragilis*). The sequence alignment was prepared using ClustalW (2).

Figure S2

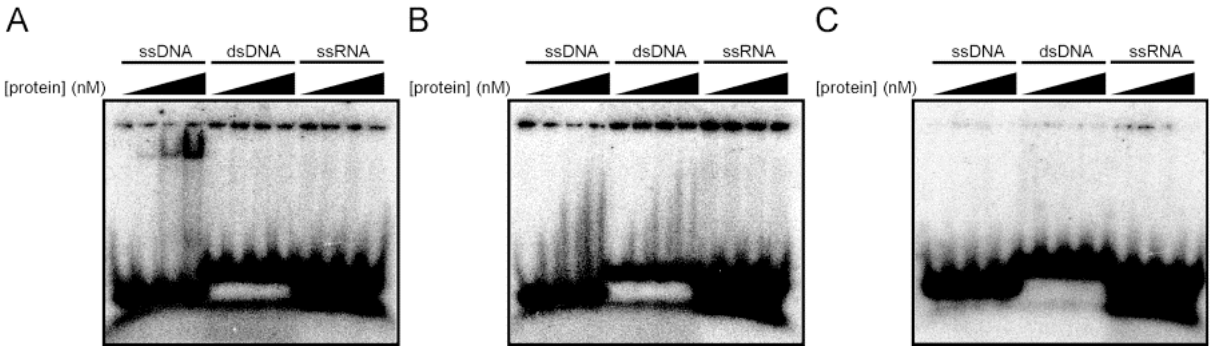

FIGURE S2. **Electrophoretic mobility shift assay with ttRecJ (*A*), ttRecJ-OB domain (*B*), and cd-ttRecJ (*C*).** The reaction mixture containing 50 mM Tris-HCl; 100 mM KCl; 20 mM EDTA; and either 10 nM 5'-^{32}P-labeled 21-mer ssDNA or 21-bp dsDNA or 21-mer ssRNA, whose sequence is the same as 21-mer ssDNA except U instead of T; along with 0, 100, 200, and 400 nM protein (pH 7.5) was incubated at 37°C for 10 min.

Figure S3

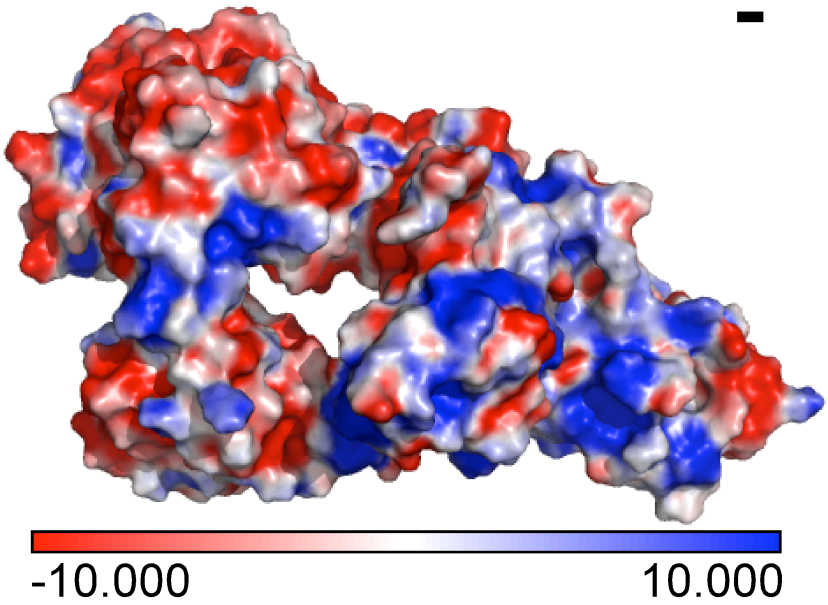

FIGURE S3. **Electrostatic potential is calculated using programs PyMOL and APBS (3).** Regions of positive electrostatic potential are shown in blue; regions of negative potential are red. The active site containing the metal center is located within the hole (at the left side wall). The scale-bar represents 10 Å.

Figure S4

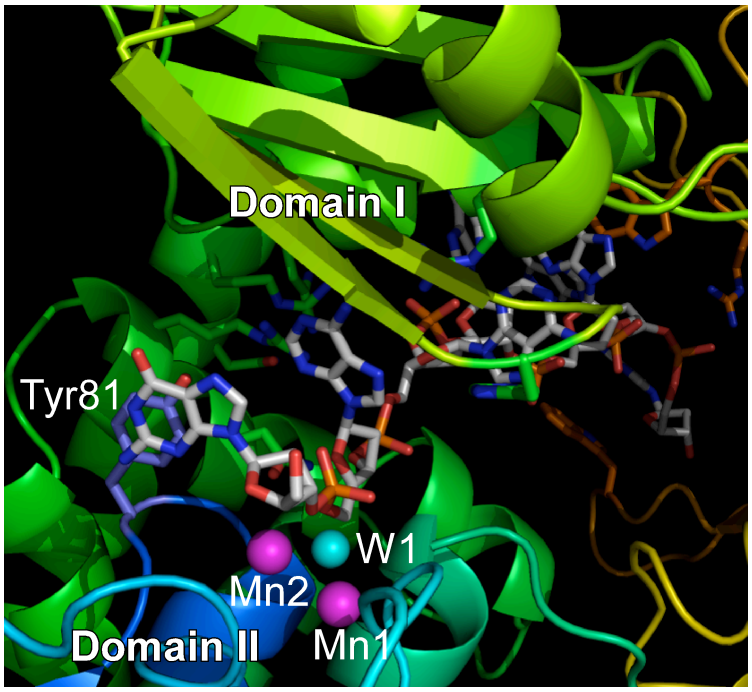

FIGURE S4. **An expanded view of the ssDNA-bound model structure.** ssDNA and the side chain of Tyr81 are shown as stick forms. Mn^{2+} ions and W1 (Fig. 3*B*) are shown as magenta spheres and a cyan sphere, respectively.

REFERENCES

1. Aravind, L., and Koonin, E. V. (1998) A novel family of predicted phosphoesterases includes *Drosophila* prune protein and bacterial RecJ exonuclease. *Trends Biochem. Sci.* **23,** 17–19
2. Thompson, J. D., Higgins, D. G., and Gibson, T. J. (1994) CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. *Nucleic Acids Res.* **22,** 4673–4680
3. Baker, N. A., Sept, D., Joseph, S., Holst, M. J., and McCammon, J. A. (2001) Electrostatics of nanosystems: Application to microtubules and the ribosome. *Proc. Natl. Acad. Sci. U.S.A.* **98,** 10037–10041