STRUCTURAL BIOLOGY

Volume 74 (2018)

Supporting information for article:

Biochemical characterization of *Mycobacterium tuberculosis* LexA and structural studies on its C-terminal segment

A. V. Chandran, R. Srikalaivani, A. Paul and M. Vijayan

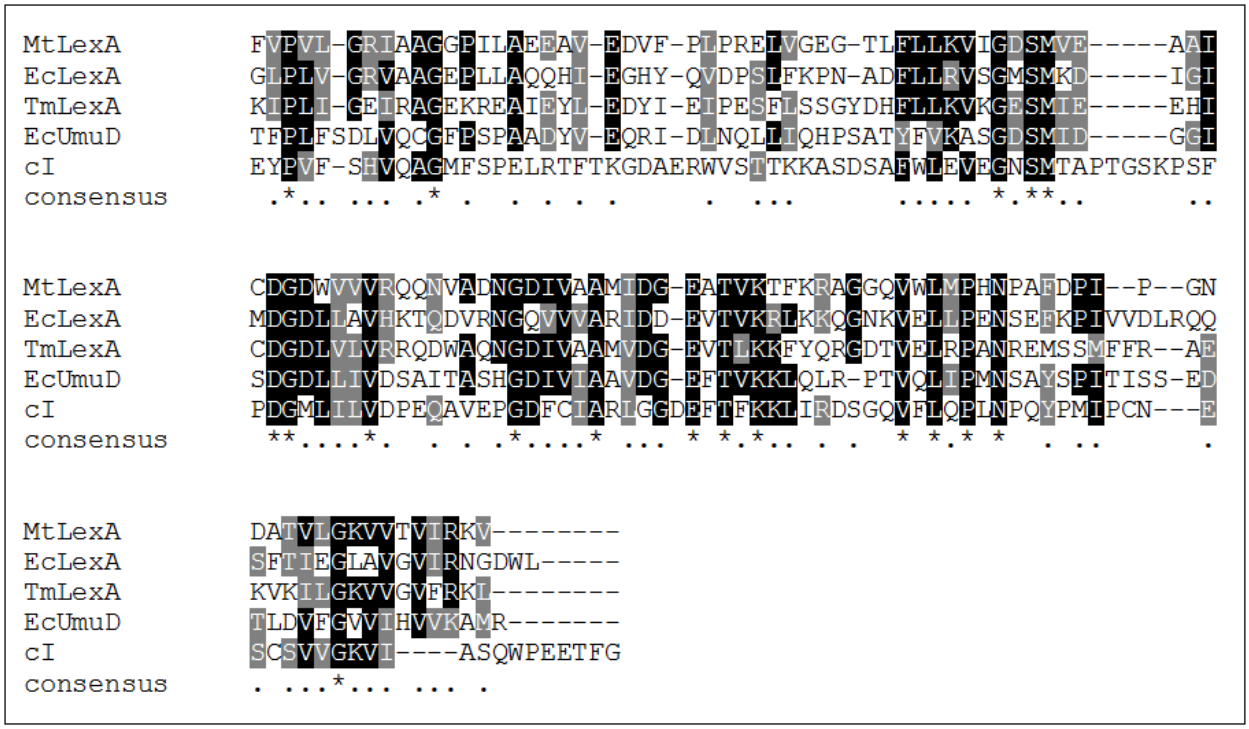

Figure S1 Multiple sequence alignment of the catalytic core of *Mt*LexA and related proteins. Boxshade was used for generating this figure from multiple sequence alignment. The amino acids are shaded from white (not conserved) to black (highly conserved).

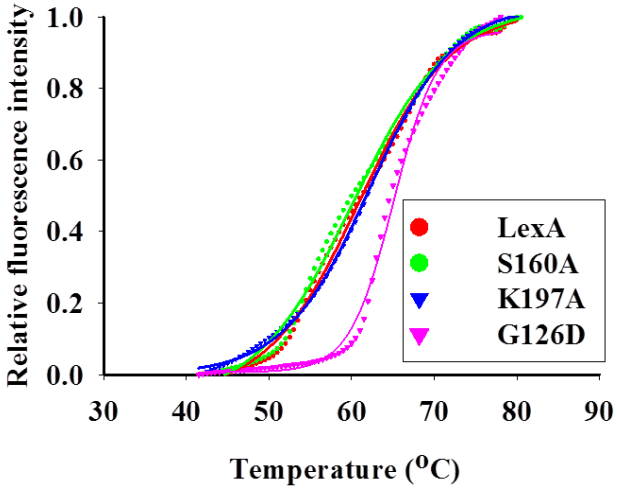

Figure S2 Thermal melting curve of *Mt*LexA and mutants.

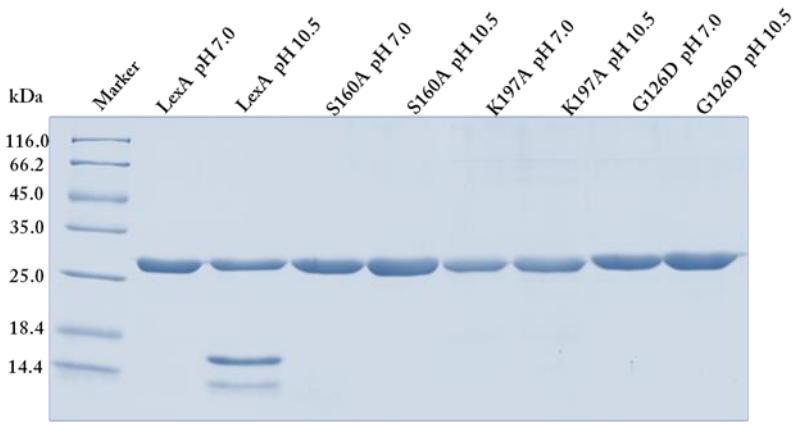

Figure S3 Cleavage of mutants compared to native *Mt*LexA.

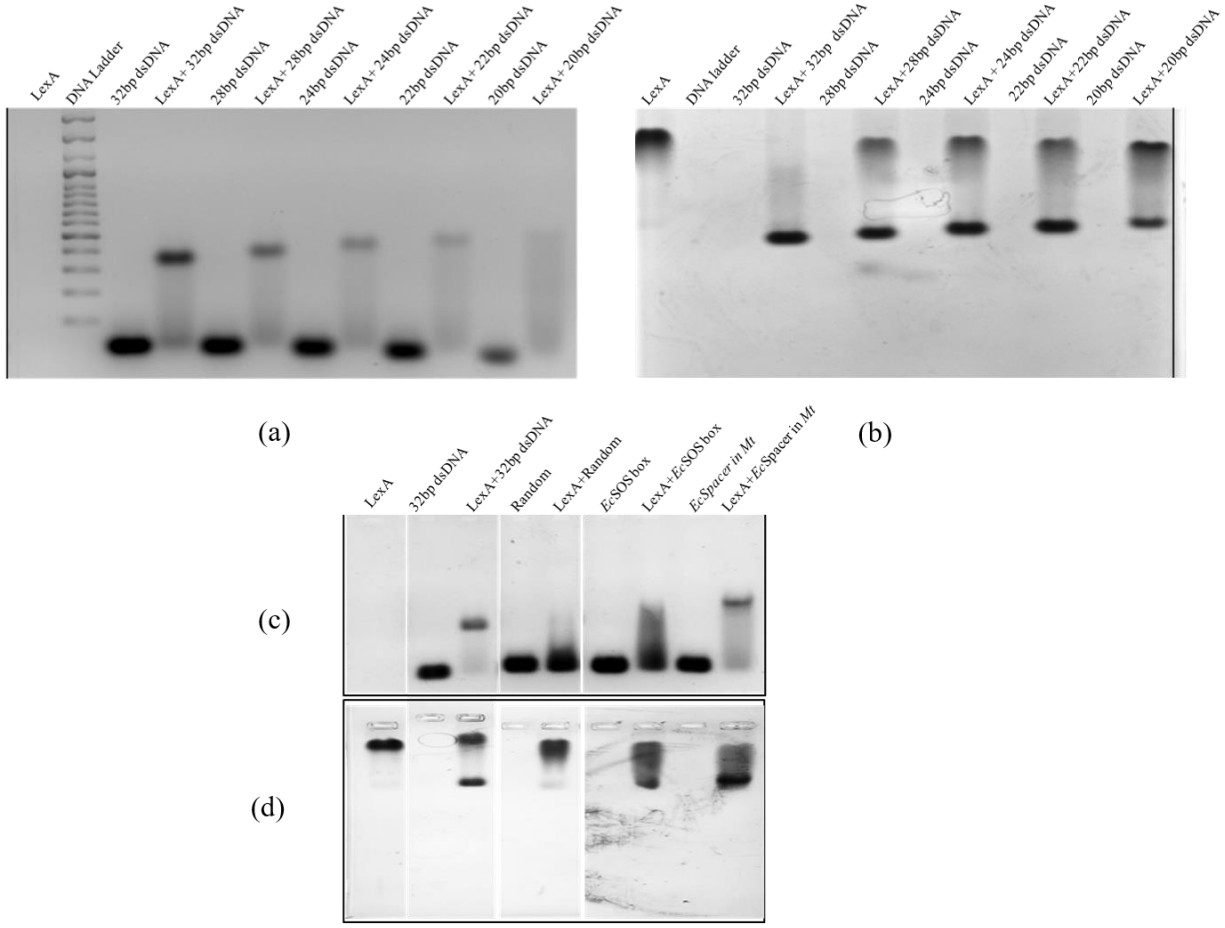

Figure S4 Gel shift assay on binding of *Mt*LexA with different lengths of flanking regions and different types of spacer DNA. Different lengths of DNA: (a) Gel stained with ethidium bromide (EtBr) (b) Gel stained with Coomassie Brilliant Blue (CBB). Different types of spacer DNA: (c) Gel stained with EtBr (d) Gel stained with CBB.

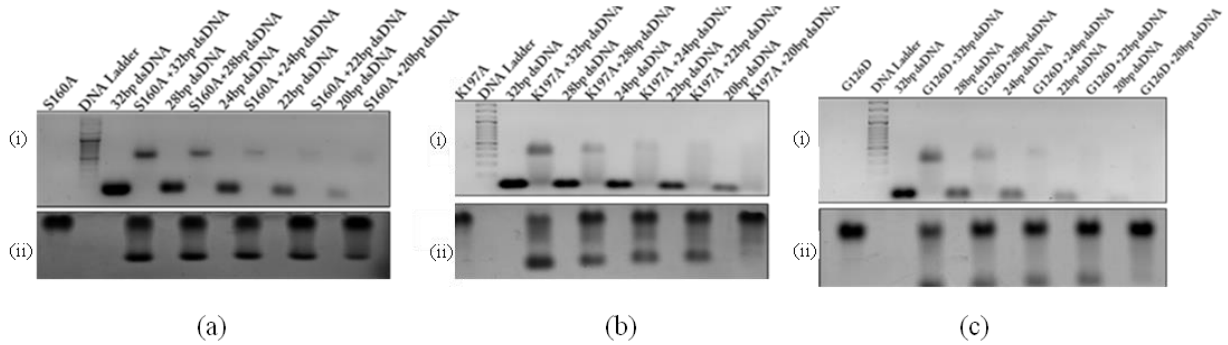

Figure S5 Gel shift assay on DNA binding in *Mt*LexA mutants with different length of flanking sequence: (a) S160A (b) K197A (c) G126D. (i) Gels stained with EtBr. (ii) Gels stained with CBB.

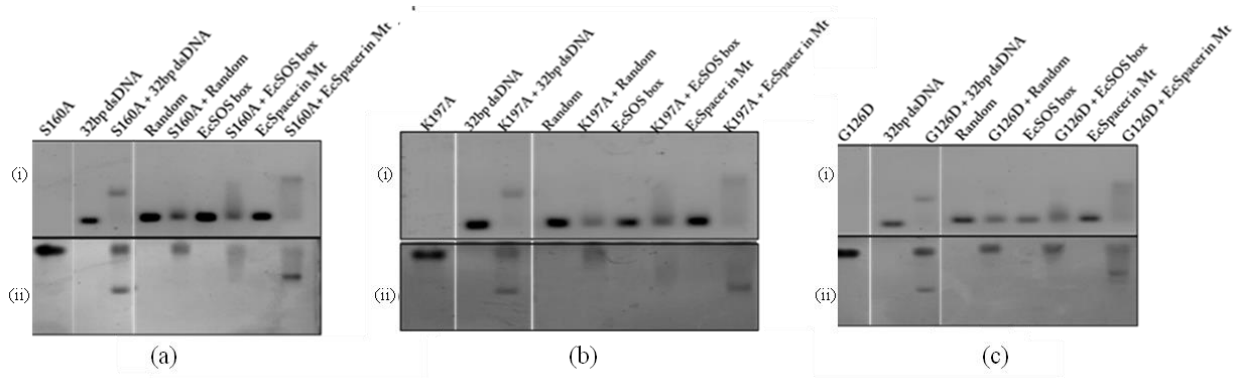

Figure S6 Gel shift assay on binding of *Mt*LexA mutants with different types of spacer DNA: (a) S160A (b) K197A (c) G126D. (i) Gels stained with EtBr. (ii) Gels stained with CBB.

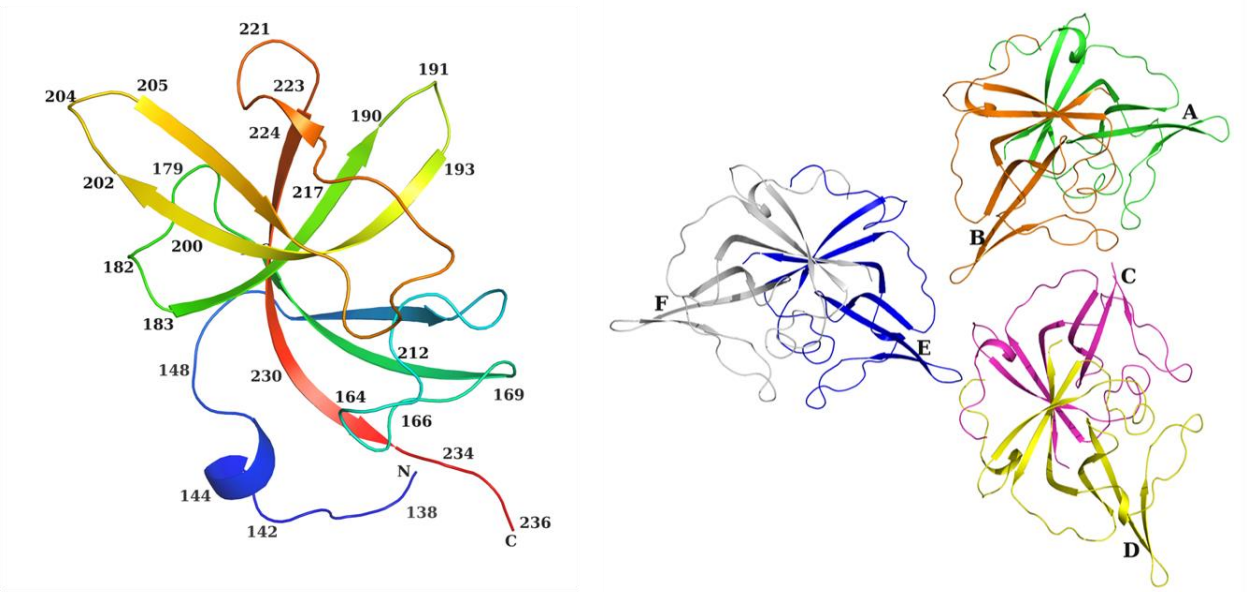

Figure S7 Contents of asymmetric unit in (a) Form I′ and (b) Form II crystals.

Table S1 List of primers used in the cloning of constructs for the present study. Restriction sites for cloning and the base changes for mutation in the primers are shown in bold.

Construct/Mutant	Primers
S160A	5' GTGATCGGTGA**CGC**GATGGTCGAAGCC 3'
	5' GGCTTCGACCATC**GCG**TCACCGATCAC 3'
K197A	5' GAGGCCACCGTC**GCG**ACGTTCAAA3'
	5' TTTGAACGT**CGC**GACGGTGGCCTC3'
G126D	5' CGTATCGCGGCC**GAC**GGCCCGATCCTT 3'
	5' AAGGATCGGGCC**GTC**GGCCGCGATACG3'
C-domain	5' AGC**CATATG**GGCGGCCCGATCCTTGCCGA3'
	5' CTGCC**GGATCC**TCGTCATCCTCGGCACCGTC3'
N-domain	5' CTG**CATATG**GTGCTGTCCGCAGATTCGG3'
	5' CTGCC**GGATCC**GGCCGCGATACGTCCCAC3'

Table S2 List of oligonucleotides used for binding studies with DNA.

Duplex name	DNA sequence
32bp	5'CACGCCTGTCGAACACATGTTTGATTCTTGGT3'
	5'ACCAAGAATCAAACATGTGTTCGACAGGCGTG3'
28bp	5'CGCCTGTCGAACACATGTTTGATTCTTG3'
	5'CAAGAATCAAACATGTGTTCGACAGGCG3'
24bp	5'CCTGTCGAACACATGTTTGATTCT3'
	5'AGAATCAAACATGTGTTCGACAGG3'
22bp	5'CTGTCGAACACATGTTTGATTC3'
	5'GAATCAAACATGTGTTCGACAG3'
20bp	5'TGTCGAACACATGTTTGATT3'
	5'AATCAAACATGTGTTCGACA3'
Random	5′GCTGTCCGCACCAGCGGCTCCGCCGGCAACCG3′
	5′CGGTTGCCGGCGGAGCCGCTGGTGCGGACAGC3′
*Ec*SOS box	5'TGCGGATACTGTATGATCATACAGTATCAATT3'
	5'AATTGATACTGTATGATCATACAGTATCCGCA3'
*Ec*spacerIn*Mt*	5'CACGCCTGTCGAACATGATCATGTTCGATTCTTGGT3'
	5'ACCAAGAATCGAACATGATCATGTTCGACAGGCGTG3'