

Supporting Online Material

***Borrelia burgdorferi* Oxidative Stress Regulator BosR Directly Represses Lipoproteins
Primarily Expressed in the Tick during Mammalian Infection**

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Table S1. Oligos used in this study.

Name ^a	Sequence (5' – 3')	Purpose
<i>flaB</i> _+441_For	AGCTGAAGAGCTTGGAATGC	q PCR
<i>flaB</i> _+543_Rev	TTGGTTTGCTCCAACATGAA	q PCR
<i>bosR</i> _+41_For	AAGTCGGCATTACAAACGAT	q PCR
<i>bosR</i> _+153_Rev	TTTTGGGTTTGATGCTATGTAT	q PCR
<i>ospA</i> _+166_For	GCAACAGTAGACAAGCTTGAGC	q PCR
<i>ospA</i> _+295_Rev	GTGTGGTTTGACCTAGATCGTCA	q PCR
<i>ospC</i> _+12_For	TACATTAAGTGCAATATTAATGACT	q PCR
<i>ospC</i> _+125_Rev	AGATTAGGCCCTTTAACAGA	q PCR
<i>rpoS</i> _+108_For	AGGCAATGCAAAAGCAAAAA	q PCR
<i>rpoS</i> _+232_Rev	ATCCCAAGTTGCCTTCTTGA	q PCR
<i>bosR</i> _+4_For	GCGGATCCAACGACAACATAATAGACGTA	Cloning
<i>bosR</i> _+531_Rev	CGCTCGAGTCATAA AGTGATTCCTTGTT	Cloning
1 st Lys_For	AAGAGGAGAAATTA ACTATGAAACATCACCATCACCATCA	Mutagenesis
1 st Lys_Rev	TGATGGTGATGGTGATGTTTCATAGTTAATTTCTCCTCTT	Mutagenesis
2 nd Lys_For	ATCACCATCACCATCACAAAAACGACAACATAATAGA	Mutagenesis
2 nd Lys_Rev	TCTATTATGTTGTCGTTTTTGTGATGGTGATGGTGAT	Mutagenesis

<i>ospA</i> _-330_For_B	ATCAAGACAAACATTGCTGCTTTAA	EMSA, q PCR
<i>ospA</i> _+20_Rev_B	CCCAATAAATATTTTTTCATAATATATTCTCCTT	EMSA, q PCR
<i>ospA</i> _-330_For_F	ATCAAGACAAACATTGCTGCTTTAA	Footprint
<i>ospA</i> _+20_Rev_V	TTCCAATAAATATTTTTTCATAATATATTCTCCTT	Footprint
<i>rpoS</i> _-277_For_B	CTTGTGTTCTCTTACTGATTTTAAATATATGTTT	EMSA, q PCR
<i>rpoS</i> _+60_Rev_B	TGCTAAACGGAGGCCAAGTA	EMSA, q PCR
<i>bosR</i> _-183_For_B	TGTCGTTTATGATTATACCTTTTTTGT	EMSA, q PCR
<i>bosR</i> _+10_Rev_B	CTGAATTCAAAAATAAAAAATTTAATTTTTTATACT	EMSA, q PCR
<i>bicA</i> _-170_For_B	TCTTCTTTTGTATCTATTTTATGCATTGT	EMSA, q PCR
<i>bicA</i> _+159_Rev_B	AGTTTTTTTGTGAATAACAAAGAAATTGGT	EMSA, q PCR
<i>bicA</i> _-1_Rev_B	AACTATCTCCTTTATATAATTATTATAATAC	q PCR
53%G+C_For_B	CTATTCTGGACTACCTGCTGA	EMSA
53%G+C_Rev	CAATGTCTTTGCGTTTTGCTT	EMSA
<i>ospD</i> _-231_For_B	AGCATCATTAAACATCCTTTCAACTCA	EMSA
<i>ospD</i> _+68_Rev_B	ATCATGAACACAAGATATTGAGAGCAAT	EMSA
Rnd_For	GGGTATGAGTCAATGAAGATGACTGGG	EMSA, FA
Rnd_Rev	CCCAGTCATCTTCATTGACTCATACCC	EMSA
Rnd_Rev_F	CCCAGTCATCTTCATTGACTCATACCC	FA

Fur_For	GGGAAATGATAATAATTATCATTTGGG	EMSA
Fur_Rev	CCCAAATGATAATTATTATCATTTCCC	EMSA
Per_For	GGGAATTTATAATTATTATAAATTGGG	EMSA
Per_Rev	CCCAATTTATAATAATTATAAATTCCC	EMSA
DR_For	GGGTAAATTAATTAATTAATTAATGGG	EMSA
DR-Rev	CCCATTTAATTTAATTTAATTTACCC	EMSA
RndAT_For	GGGTATAATATATTTAAAATAAAGGG	EMSA, FA
RndAT_Rev	CCCTTTATTTTAAATATATTATACCC	EMSA
RndAT_Rev_F	CCCTTTATTTTAAATATATTATACCC	FA
Per/7-0-7_For	GGGAATTTATAATATTATAAATTGGG	EMSA
Per/7-1-7_Rev	CCCAATTTATAATATTATAAATTCCC	EMSA
Per/7-2-7_For	GGGAATTTATAATTTATTATAAATTGGG	EMSA
Per/7-2-7_Rev	CCCAATTTATAATAAATTATAAATTCCC	EMSA
<i>ospA</i> _-60_For_F	TAATCTTATAATATAATTATACTT	FA
<i>ospA</i> _-37_Rev	AAGTATAATTATATTATAAGATTA	FA
<i>ospD</i> _-85_For_F	ATAATTGATATTTAAATATAATTGAT	FA
<i>ospD</i> _-60_Rev	ATCAATTATATTTTAAATATCAATTAT	FA

^a Indicates the target gene (when applicable), direction (For, forward; Rev, reverse), and 5'-modification (B, biotin; F, 6-FAM; V, VIC) of the oligo.

Table S2. Putative BosR-binding sites.

Gene	Location ^a	Spacer ^b	Mismatch ^c	Sequence
BB_0001	-36 ~ -23	0	2	ATATAATATTATTA
BB_0026	-115 ~ -101	1	2	TTAAAATTATTATCC
BB_0027	-29 ~ -15	1	2	GGATAATAATTTTAA
BB_0045	-69 ~ -55	1	2	TTTTATTTATTATAA
BB_0046	-37 ~ -23	1	2	TTATAATAAATAAAA
BB_0050	-157 ~ -144	0	2	TTACAATATTAACA
BB_0061	-70 ~ -55	2	2	TTATAATTCCTTATTA
BB_0062	-19 ~ -4	2	2	TAATAAGGAATTATAA
BB_0083	-106 ~ -92	1	2	TTAAAATAATTATTA
	-148 ~ -134	1	2	TTTTAATTATTATAT
BB_0084	-91 ~ -77	1	2	TAATAATTATTTTAA
	-49 ~ -35	1	2	ATATAATAATTAATAA
BB_0096	-112 ~ -99	0	2	TTTTAATATTGTAA
BB_0106	-99 ~ -85	1	2	TTATTCTTATTATCA
BB_0123	-147 ~ -134	0	2	TGATAATTTTATTA
	-80 ~ -67	0	2	TTATATTCTTATAA

BB_0133	-154	~	-141	0	1	TTTTAATATTATAA
BB_0134	-43	~	-30	0	1	TTATAATATTTAAA
BB_0142	-58	~	-43	2	2	TTATAATAATTAATCA
BB_0143	-34	~	-19	2	2	TGATTAATTATTATAA
BB_0142	-63	~	-50	0	2	TTATAAATTATAATAA
BB_0143	-27	~	-14	0	2	TTATTATAATATAA
BB_0142	-68	~	-53	2	2	TTGTATTATATTATAA
BB_0143	-24	~	-9	2	2	TTATAATATAATACAA
BB_0166	-60	~	-45	2	2	TTATTATAATTTATAA
BB_0167	-119	~	-104	2	2	TTATAAATTATAATAA
BB_0166	-66	~	-52	1	1	TGATATTTATTATAA
BB_0167	-112	~	-98	1	1	TTATAATAAATATCA
BB_0179	-17	~	-2	2	2	TTATAATACATTAAG
	-69	~	-54	2	1	TTATAAGCTATTATAA
	-78	~	-64	1	2	TGAAATTAATTATAA
BB_0180	-82	~	-67	2	2	CTTTAATGTATTATAA
	-30	~	-15	2	1	TTATAATAGCTTATAA

	-20	~	-6	1	2	TTATAATTAATTTCA
BB_0204	-81	~	-66	2	2	TTATTATCTACTATAA
	-71	~	-57	1	2	CTATAAAGATTATAA
BB_0211	-37	~	-24	0	2	TGAAAATATTAATAA
BB_0217	-62	~	-47	2	2	TTATAATCAATTCTAT
BB_0226	-92	~	-77	2	2	TTATAAGATATTATAT
BB_0227	-53	~	-38	2	2	ATATAATATCTTATAA
BB_0233	-25	~	-12	0	2	TAATAATATTATTA
BB_0253	-90	~	-77	0	2	TTATTATTTTATAA
BB_0254	-199	~	-186	0	2	TTATAAAATAATAA
BB_0260	-84	~	-70	1	2	TTATAGTTATAATAA
	-94	~	-79	2	2	TTATATTATATTATAG
BB_0261	-149	~	-135	1	2	TTATCATTATTATTA
	-131	~	-116	2	2	CTATAATATAATATAA
BB_0309	-159	~	-145	1	2	TTATAAGGATTTTAA
	-103	~	-90	0	2	TTATAAATTTATAA
BB_0328	-73	~	-58	2	2	TTAAAATTTATTAATAA

BB_0330	-34 ~ -19	2	2	TTATAAATAATTTTAA
	-131 ~ -118	0	2	TTATAATATTTAAAT
BB_0358	-109 ~ -95	1	2	TTTTAATTACTATCA
BB_0360	-175 ~ -160	2	2	TGATAACTTATTATTA
	-18 ~ -5	0	2	ATATAATATTTTAA
BB_0381	-46 ~ -32	1	2	TTATAATAAATAGAA
BB_0399	-97 ~ -82	2	2	TTATAATGACCTATAA
BB_0403	-127 ~ -114	0	2	TGATATTATTTTAA
BB_0404	-97 ~ -84	0	2	TTAAAATAATATCA
BB_0421	-96 ~ -83	0	2	ATATAATATTATTA
BB_0431	-64 ~ -50	1	2	TTATTATTTTTATCA
	-72 ~ -59	0	2	TTAAAATATTATTA
BB_0445	-59 ~ -44	2	2	GTATAATTTATTATAG
BB_0473	-245 ~ -230	2	2	TTATTATTTATTATAT
BB_0512	-68 ~ -54	1	2	TTTTCATGATTATAA
BB_0524	-36 ~ -21	2	2	TTAGAATTTATTATTA
BB_0543	-77 ~ -63	1	2	TGGTAATTATTATAT

	-71 ~ -58	0	2	TTATTATATAATAA
BB_0551	-85 ~ -72	0	2	AGACAATATTATAA
	-219 ~ -206	0	2	TTATTATTTTATCA
BB_0552	-170 ~ -157	0	2	TTATAATATTGTCT
	-36 ~ -23	0	2	TGATAAAATAATAA
BB_0554	-238 ~ -225	0	2	TTATAATGTTAGCA
BB_0557	-69 ~ -54	2	2	TTTTTATGTATTATAA
BB_0560	-51 ~ -36	2	2	TTATGTTATATTATAA
BB_0561	-77 ~ -62	2	2	TTATAATATAACATAA
BB_0574	-178 ~ -164	1	2	TAATAAAAATTATAA
	-169 ~ -156	0	2	TTATAAGATTAAAA
BB_0576	-47 ~ -33	1	2	ATATACTAATTATAA
BB_0577	-107 ~ -93	1	2	TTATAATTAGTATAT
BB_0582	-25 ~ -10	2	2	CTAAAATATATTATAA
BB_0583	-109 ~ -94	2	2	TTATAATATATTTTAG
BB_0591	-14 ~ -1	0	2	TAAGAATATTATAA
	-37 ~ -22	2	2	ATATAATTATTTATAA

BB_0592	-115 ~ -102	0	2	TTATAATATTCTTA
	-94 ~ -79	2	2	TTATAAATAATTATAT
BB_0603	-106 ~ -91	2	2	TGTTAATATATTATAC
	-134 ~ -121	0	2	TTTTAATATTATAT
BB_0604	-69 ~ -56	0	2	TTGTAATATTAAAA
BB_0605	-42 ~ -29	0	2	TTTTAATATTACAA
BB_0608	-66 ~ -51	2	2	ATATTATTAATTATAA
BB_0610	-40 ~ -26	1	1	TTTTAATAATTATCA
BB_0620	-122 ~ -108	1	2	TGCTAATAAATATAA
	-150 ~ -135	2	2	TTATTATAAAGTATAA
BB_0621	-191 ~ -177	1	2	TTATATTTATTAGCA
	-164 ~ -149	2	2	TTATACTTTATAATAA
BB_0628	-46 ~ -31	2	2	ATATATTCTATTATAA
BB_0647	-133 ~ -120	0	2	TTATACTAATATAA
BB_0649	-237 ~ -224	0	2	TAATAATAATATAA
BB_0657	-116 ~ -102	1	2	TACTAATAATTATAA
BB_0658	-47 ~ -33	1	2	TTATAATTATTAGTA

Bb_0660	-58 ~ -43	2	1	ATATAATTAATTATAA
BB_0661	-62 ~ -47	2	1	TTATAATTAATTATAT
BB_0664	-28 ~ -14	1	2	TTATAATAATCATAT
BB_0665	-44 ~ -30	1	2	ATATGATTATTATAA
BB_0664	-48 ~ -34	1	2	TTAAAATCCTTATAA
BB_0665	-24 ~ -10	1	2	TTATAAGGATTTTAA
BB_0676	-190 ~ -175	2	2	TTCTAATGAAATATAA
BB_0677	-68 ~ -53	2	2	TTATATTTTATTAGAA
BB_0680	-150 ~ -135	2	2	TAATAATAGTTTATAA
BB_0682	-65 ~ -52	0	2	GTATTATATTATAA
BB_0683	-42 ~ -29	0	2	TTATAATATAATAC
BB_0689	-62 ~ -47	2	2	TTATAATACATAATGA
	-71 ~ -57	1	1	ATATAATTATTATAA
BB_0690	-38 ~ -23	2	2	TCATTATGTATTATAA
	-28 ~ -14	1	1	TTATAATAATTATAT
BB_0709	-101 ~ -86	2	2	TTATTATTGGTTATAA
BB_0727	-25 ~ -12	0	2	TTATAATTCTATAA

BB_0734	-97 ~ -84	0	2	TTATAATATAAAAA
	-100 ~ -87	0	2	TTATTATAATATAA
BB_0735	-22 ~ -9	0	2	TTTTTATATTATAA
	-19 ~ -6	0	2	TTATATTATAATAA
BB_0756	-167 ~ -152	2	2	TTATAATTGAAAATAA
	-177 ~ -162	2	2	TAATGATATATTATAA
BB_0757	-49 ~ -34	2	2	TTATTTTCAATTATAA
	-39 ~ -24	2	2	TTATAATATATCATTAA
BB_0785	-102 ~ -87	2	2	TTAAAATTAATTAATAA
BB_0793	-73 ~ -59	1	2	CGATAATTATTATTA
	-64 ~ -51	0	2	TTATTATATAATAA
BB_0804	-147 ~ -132	2	2	TAATAATTTATTATTA
BB_0828	-31 ~ -16	2	2	TTATAATACAATATAT
BB_0829	-59 ~ -44	2	2	ATATATTGTATTATAA
BB_0835	-66 ~ -51	2	1	ATATAATTCATTATAA
	-89 ~ -74	2	2	TGATAATAAATTCTAT
BB_0840	-106 ~ -93	0	2	TAATAATATTAATAA

BB_0841	-86 ~ -73	0	2	TTTTAATATTATTA
BB_A03	-106 ~ -91	2	2	TAATAATTAATAATAA
	-35 ~ -21	1	2	TAATAATAATTAAAA
BB_A15	-63 ~ -50	0	2	TGTTAATCTTATAA
	-28 ~ -15	0	2	TTATATTAATATAA
BB_A38	-138 ~ -125	0	2	TTATAAAATTTTAA
	-78 ~ -64	1	2	TAATAATTAATATCA
BB_A61	-53 ~ -40	0	2	TTTTAATATTTTAA
	-219 ~ -204	2	2	TTATATTAAAATATAA
	-224 ~ -209	2	2	TGATATTATATTAATAA
BB_A62	-208 ~ -195	0	2	TTAAAATATTAATAA
	-44 ~ -29	2	2	TTATATTTTAATATAA
	-39 ~ -24	2	2	TTTTAATATAATATCA
BB_A66	-36 ~ -21	2	2	TTAAAATTCATTAAAA
	-72 ~ -57	2	2	TAATAATTAATTTTAA
BB_A68	-81 ~ -67	1	2	TGACAATTATTGTAA
BB_A69	-62 ~ -49	0	2	TTATAATATTATTT

	-71	~	-57	1	2	TTAATATAATTATAA
	-101	~	-88	0	2	TAATAATATTATTA
BB_A0078	-152	~	-139	0	2	TTAAAATATTTAAAA
BB_B01	-85	~	-70	2	2	TTAAAATATAATATAA
BB_B04	-70	~	-57	0	2	TTATATTATAATAA
BB_B04	-128	~	-114	1	2	TTATAAATATTGTAA
BB_B04	-223	~	-210	0	2	ATATTATATTATAA
BB_B05	-229	~	-216	0	2	TTATTATAATATAA
BB_B05	-172	~	-158	1	2	TTACAATATTTATAA
BB_B05	-76	~	-63	0	2	TTATAATATAATAT
BB_B16	-169	~	-156	0	2	TTATAATCTTTTCA
BB_D001	-50	~	-37	0	1	CTATAATATTATAA
BB_D04	-222	~	-207	2	2	ATATGATTTATTATAA
BB_D10	-36	~	-22	1	2	TTAAAATAAATATAA
BB_D15	-158	~	-143	2	2	TTATATTTCTTTATAA
	-220	~	-207	0	2	TGATAATAATTTAA
BB_E17	-40	~	-27	0	2	TTAGAATAGTATAA

BB_F06	-225	~	-210	2	1	ATATAATTTATTATAA
BB_F20	-129	~	-115	1	2	TTTTAATTATTAATAA
BB_G01	-101	~	-86	2	2	TTATATTTTATTGTAA
BB_G07	-105	~	-92	0	2	TTAGAATATAATAA
	-85	~	-71	1	2	TAATAATAATTATTA
	-73	~	-58	2	2	TTATTATGAATTATTA
BB_H02	-168	~	-155	0	1	CTATAATATTATAA
BB_H04	-129	~	-116	0	2	CTATAATATTATGA
BB_H40	-18	~	-4	1	2	TTATAATTATAAAAAA
BB_I02	-126	~	-111	2	2	TGTAAATACATTATAA
BB_I12	-34	~	-20	1	2	CTATAATAATTTTAA
BB_I16	-237	~	-223	1	2	TTTAAATAATTATAA
	-55	~	-41	1	1	TTATAATAAATATAA
BB_I36	-64	~	-51	0	1	TTGTAATATTATAA
	-80	~	-67	0	1	TGATAATAATATCA
BB_I38	-64	~	-51	0	1	TTGTAATATTATAA
	-80	~	-67	0	1	TGATATTATTATCA

BB_I39	-64 ~ -51	0	2	TTGTAATATTATAT
	-74 ~ -59	2	2	TGATATTA AATTGTAA
BB_I41	-21 ~ -7	1	2	ATATTATAATTATAA
	-26 ~ -13	0	2	ATATGATATTATAA
BB_J08	-116 ~ -101	2	2	TTATATTTTATTGTAA
	-66 ~ -51	2	2	TTATTATTATTTATAA
BB_J09	-148 ~ -133	2	2	TGATATTA A AATATAA
	-131 ~ -116	2	2	TGATATTA A AATATAA
	-114 ~ -99	2	2	TGATATTA A AATATAA
	-97 ~ -82	2	2	TGATATTA A AATATAA
	-80 ~ -65	2	2	TGATATTA A AATATAA
	-63 ~ -48	2	2	TGATATTGAAATATAA
	-46 ~ -31	2	2	TGATATTA A AATATAA
BB_J19	-37 ~ -23	1	2	TAATAATAATTATTA
BB_J0056	-114 ~ -100	1	2	TAATAATTATTATTA
BB_J36	-54 ~ -41	0	2	TTATAATA A AATAA
BB_J41	-64 ~ -51	0	2	TTGTAATATTATTA

BB_J46	-41 ~ -28	0	1	TTATAATATTATAT
BB_K01	-41 ~ -27	1	2	TTATAATAATTATTC
	-48 ~ -33	2	2	TTATTATTTATAATAA
	-101 ~ -86	2	2	TTACAATTTATTGTAA
BB_K15	-208 ~ -194	1	2	TTATAATGACTACTA
	-191 ~ -177	1	2	TAATATTTATTATAA
BB_K47	-79 ~ -65	1	1	TTATAATTATTATTA
	-87 ~ -74	0	2	TTTTTATATTATAA
BB_K49	-79 ~ -65	1	1	TTATAATTATTATTA
	-87 ~ -74	0	2	TTTTTATATTATAA
BB_K50	-46 ~ -31	2	2	TGATAAAATATTCTAA
BB_L35	-130 ~ -116	1	2	TTATGATTTTTATAA
	-53 ~ -39	1	2	TTAAAATAATTATGA
BB_M35	-244 ~ -230	1	2	TTTTATTTATTATAA
BB_N19	-57 ~ -44	0	2	TTATGATATTAACA
BB_N35	-244 ~ -230	1	2	TTTTATTTATTATAA
BB_P30	-36 ~ -21	2	2	TAATAATTAATTATTA

BB_P35	-244	~	-230	1	2	TTTTATTTATTATAA
BB_Q05	-213	~	-200	0	2	TTATATTATTATTA
	-149	~	-136	0	1	TTAAAATATTATAA
BB_Q42	-130	~	-116	1	2	TTATGATTTTTATAA
BB_Q85	-222	~	-207	2	2	ATATGATTTATTATAA
BB_Q89	-50	~	-37	0	1	CTATAATATTATAA
BB_R36	-244	~	-230	1	2	TTTTATTTATTATAA
BB_R41	-94	~	-79	2	2	TGATAATTTCTTGTA
BB_S41	-180	~	-165	2	2	TTATAATCTTTTGTA
BB_T02	-104	~	-89	2	2	TGTAAATACATTATAA
BB_U02	-126	~	-111	2	2	TGTAAATACATTATAA

^a Location of each site is shown as relative to the start of each gene. Only sites within 250 bps upstream of a gene are shown.

^b Indicates the length of the spacer in base pairs.

^c Indicates the number of mismatches from the consensus sequence T(T/G)ATAAT-N{0,2}-ATTAT(A/C)A.

References

- Burtnick, M.N., J.S. Downey, P.J. Brett, J.A. Boylan, J.G. Frye, T.R. Hoover & F.C. Gherardini, (2007) Insights into the complex regulation of *rpoS* in *Borrelia burgdorferi*. *Mol Microbiol* **65**: 277-293.
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- Margolis, N., D. Hogan, K. Tilly & P.A. Rosa, (1994) Plasmid location of *Borrelia* purine biosynthesis gene homologs. *J Bacteriol* **176**: 6427-6432.
- Norris, S.J., C.J. Carter, J.K. Howell & A.G. Barbour, (1992) Low-passage-associated proteins of *Borrelia burgdorferi* B31: characterization and molecular cloning of OspD, a surface-exposed, plasmid-encoded lipoprotein. *Infect Immun* **60**: 4662-4672.

PospAB, -330 ~ +20, 20% G+C

ATCAAGACAAACATTGCTGCTTTAAAAAATCACATTGACAAAATAAAACCAATTGCAATGCAGATTTACAAAAATACTCAAAAAATATACCTTAATACT
TTAAAAGACATTTAACTTTTCTTTTCTGAAAGTCCCAAACTGGGACTTTTTTTAAATAAAAAATCTACATTAATCTAAGCTTAATTAGAACCAAAC
TTAATTAACCAAACCTAATTGAAGTTATTATCATTTTATTTTTTTTCAATTTTCTATTTGTTAATTGTTAATCTATAATATAATATACCTGTATTA
AGTTATATTAATATAAAAGGAGAAATATATATGAAAAAATATTTTATTGGG

PospD, -231 ~ +68, 19% G+C

AGCATCATTAAACATCCTTTCAACTACTACTATTGTTTTCTTAGCCTTAAGCTAGCCAAGCTAAATAGAAATTAGTAGGCAATTGATATTAATA
TAATTGATATTAATAATAATTGATATTAATAATAATTGATATTAATAATAATTGATATTAATAATAATTGATATTAATAATAATTGATATTAATA
TATAATTTAAGACATTATATTTAAGGAGTATAAATATGAAAAAATATAAAAAATACTACTGTTAAGTTTATTTTTATTGCTCTCAATATCTTGTGTTTAT
GAT

PrpoS, -277 ~ +60, 28% G+C

TGCTAAACGGAGGCCAAGTAGAAGAAATTTTTGAAGATATGCTTTGCGAACAAAGAGCAAAACAAATGGCACAAGCTCAAAGCTTTGGCCTTGCCGATT
TAATTTACAATGAATTACAAAAAGTAAATAATCAAAAAATACTCCCCCTAAACTCAAAATATATCCTATTTAGTTTAAACCATTTTTAAATTAAT
TGGCACAGTTTTTGCATGGAAATTAAGTAGTAAAACTTAATCACAATATTCAGAAAGGGGAGAAAATATAATAACTATGAACATATTTAGTAATGAGG
ATTTAAACATATATTTAAATCAGTAAGAGAACACAAG

PbicA, -170 ~ +159, 21% G+C

TCTTCTTTGTATCTATTTATGCATTGTACTTAAATTGCATGCTTGAGATAAAAACAGTGTAAAAATTATAATCAATTTTTTCATTAAATATCCTTTTT
ATTTATATAGGTATTAACCATAATTATATCTCATTATGTATTATAATAATTATATAAAGGAGATAGTTATGGAAAAGTATTTAAGCTATATAAAAAAG
GATGATTTAGACGCAATACAATTAATAAATACAAGAATTGTAGCAAGTTTGCATATTTTTTATTCTAATTTAAGAGGTATTCATTGGAATATAAAAAAGATA
CCAATTTCTTTGTATTACAAAAAACT

PbosR, -183 ~ +10, 15% G+C

AATTTTGATAACAAAATAAACTGAATTCAAAAATAAAAAATTTAATTTTTTATACTAATATAAAAAATATGAAAAATAAATAAATAAAGTAGTAAAA
TATTAATAACTGGGTATAAAATATCCTAAGAAGAACATAAAAAAGTATTTAATCTTAATTTAAACAAAAAGGTATAATCATATGAACGACA

PospC, -330 ~ +23, 22% G+C

ACTTTTGTATAAACGCCAATTTCTCTAATTTCTTCTGCAATTAGTTGGCTATATTGGGATCCAAAATCTAATACAAGTATTGCCTGAGTATTCATTA
TATAAGTCCTTAAATAAATTAACCTTTTTTTTATTAAGTATACTTCATTTAATTTTAGCATATTTGGCTTTGCTTATGTGCGATTTTAAATCAAATTA
GACAATATTTTCAAATCTTCAATATTTATTCAAGATATTGAAGAATTTGAAAAAATATTTTTTCAAATAAAAAATGAAACAAAATTTGTTGGACT
AATAATTCATAAATAAAAGGAGGCACAAATTAATGAAAAAGAATACATTAAGTGC

Pech193, -246 ~ +3, 23% G+C

GAGAAATATAACCTTTAAATATGTTGAATACTTTCAAATCATAACATAATAGTATGGTGAAGTCAAATGAAAAATGCATTTTAAATATTTAAATCTT
ATCTTTAAGTTCTTTGAGGAAAATAAGAAATTTGCAATATGTTGTTAAAGCAATATGATTAATTAGTCAAGCTTGTCAAATATGAAAAAGTAAAAAG
ATGTCCTTTTTGAAAAGGATATGTAATAGGAGGATATTTAAGTTATATG

Pech818, -542 ~ +3, 25% G+C

AAGCATATCTTTTAAATGTTTTAAAAATTTTTCTAGTTAGGTCTATGTATAGCATTATAAAAAATACTTGAAGCTTTAAACATTTTTAAGCTGTAATTAT
GTGTGCTTATTTGCAACAGATATAATTTATGCGCAAATGTGTAGTACATATGAATAATACAACTTAAAAACAAGTAAAAAGTGCCTTTATGAAGGATA
ATTTTTAACTGAAGTACCAATAAGCAAAGTCAATATAAGTAAAAATGTACATTTGTATGTGTACAGCTTAAATGGTTATAATTGGTAGGGAATTTGT
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TTTGTCTTAAACATCAAATCAGAATAGTATTGTTGAGCATGTAGTAAAGGATATATCCTAAAATCAAGTATTGTTAATAGTATTGACACAGAGAATAAAT
GAGGTTATATCTTTTTATTCTGTTATAATTAGTGTCTAAAATG

53% G+C

CTATTTGACTACTCTGCTGACGCTGCACCCGGTTCGTAAGTGGACAACAAAATTTAGGACGCAATGCTGGCCCGCGGATTGAACGTGCCGAAGAACT
GGGCCCGGATCTGGACAACTGACCTTTATCGATGAAGACCTGACGAAACATGGCAAAAAAGATTTTCGGTGCCTGGCCAGCAATCCGGATATCCAGACC
CTGGTGGGCGCCCGCTGCGTGCAGGTAATGTTGCACTGGCTAAATCTATCGAACAACTGATCCCGGAAGCGAAACGCAAAGACATTG

Fig S1. Sequences of EMSA probes. Whenever applicable, the position of a probe is indicated as relative to the start of a gene, and the ATG start codon and the Shine-Dalgarno sequence of each gene are indicated with double underlines. The transcription start sites for *ospAB*, *ospD*, *rpoS*, and *ospC* had been previously determined (Jonsson *et al.*, 1992; Norris *et al.*, 1992; Burtnick *et al.*, 2007; Margolis *et al.*, 1994) and are indicated with asterisks. Putative -35, -24/-12, and -10 promoter regions that precede these experimentally determined transcription start sites are indicated with brackets.

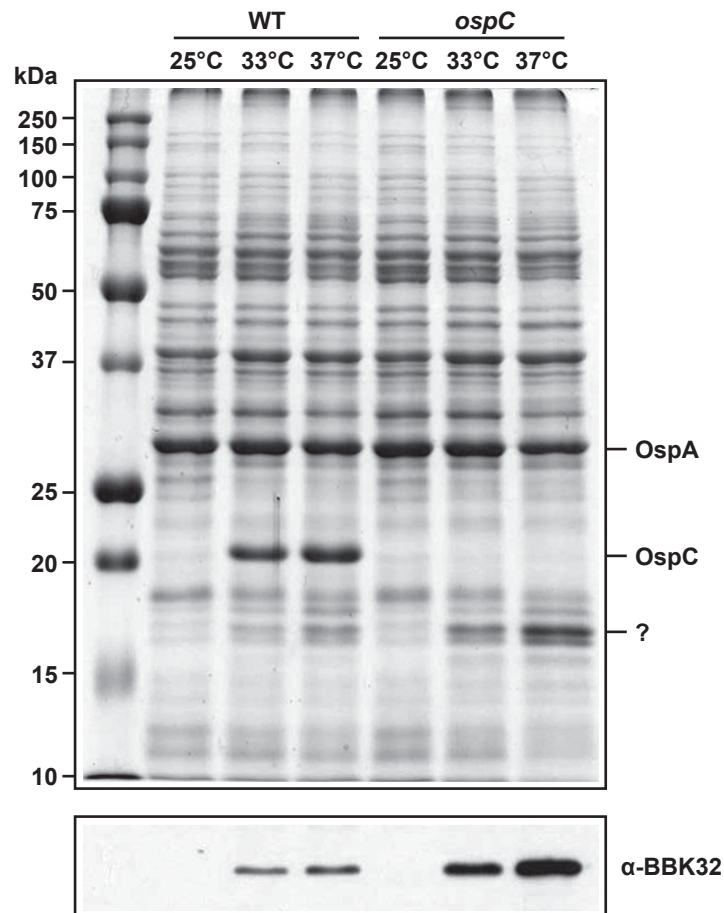


Fig. S2. Up-regulation of BBK32 in an *ospC* mutant. The *ospC* mutant was constructed from an infectious clone of *B. burgdorferi* strain 297. Up-regulation of BBK32 in the *ospC* mutant was evident in an immunoblot analysis using α -BBK32 antibodies. The identity of another protein that was also significantly up-regulated in the *ospC* mutant, indicated by a question mark, remains unknown. However, given that this protein was significantly up-regulated at 37°C and also reacted strongly with sera from mice infected with *B. burgdorferi* through tick inoculation (data not shown), this protein could be a lipoprotein belonging to the RpoS regulon.

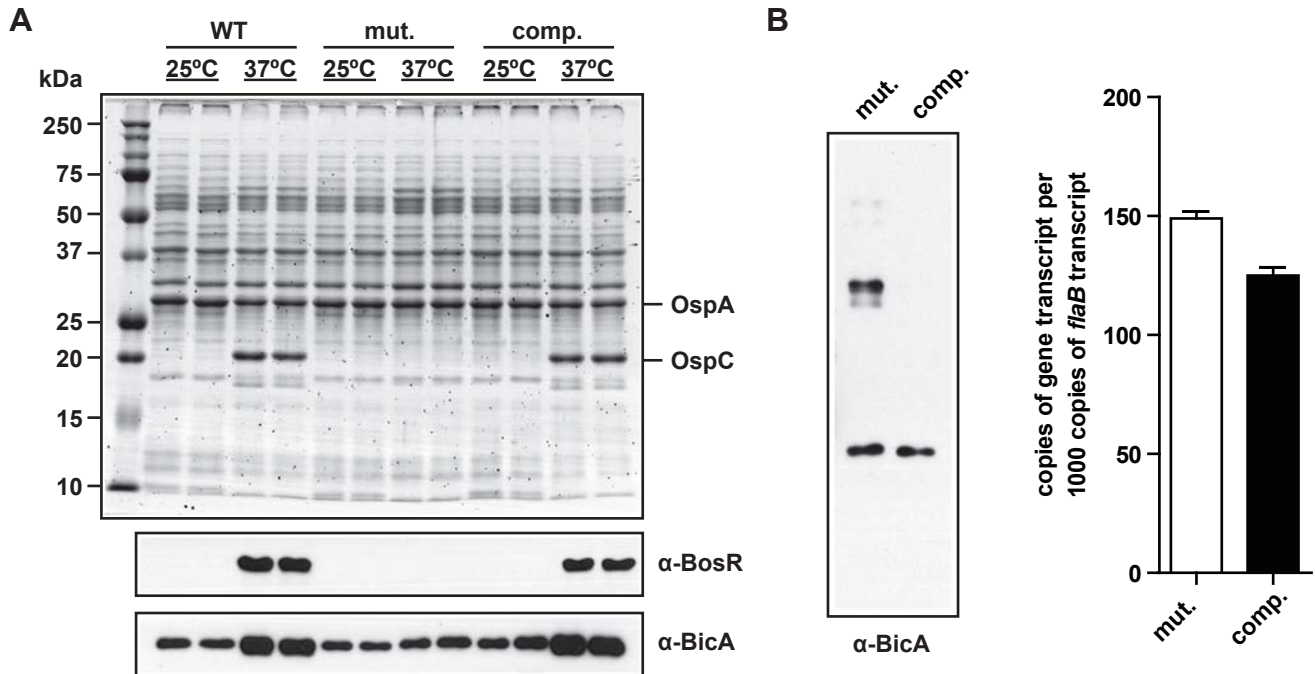


Fig. S3. BosR-dependent activation and repression of BicA. (A) When cultured *in vitro*, BicA expression in the wild type (WT) was significantly elevated at 37°C as compared to 25°C. This activation was clearly dependent on BosR based on analyses of the *bosR* mutant (mut.) and the complemented *bosR* mutant (comp.). (B) In host-adapted spirochetes harvested from DMCs, the presence of BosR (comparing the complemented strain with the *bosR* mutant) appeared to be associated with a modest reduction in the BicA protein level (left) as well as in the *bicA* mRNA level (right).

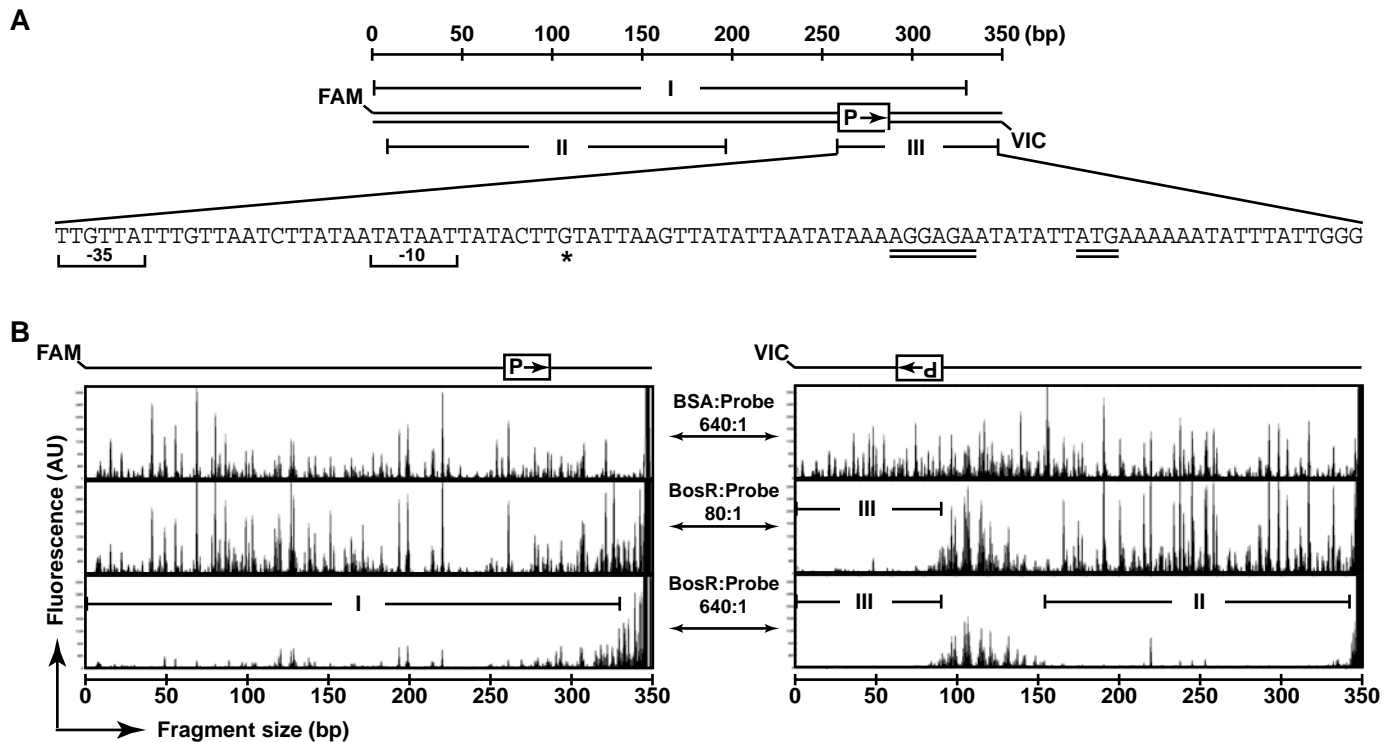


Fig. S4. Footprint analysis of BosR binding to P_{ospAB} . (A) A 350-bp DNA fragment containing the *ospAB* promoter was PCR amplified using a pair of primers, one labeled at the 5' end with FAM and the other labeled at the 5' end with VIC. (B) Electropherograms of DNase I-digested probe following incubation with 640-fold molar excess of BSA or 80- or 640-fold molar excess of BosR. The y-axis scale is the same for all electropherograms, ranging from 0 to 3,000 arbitrary units (AU). The direction of the promoter (P) is indicated with an arrow. Three regions of the probe that were protected from DNase I digestion by BosR are designated I, II, and III. The nucleotide sequence of the opposite strand of footprint III is shown. The -35 and -10 promoter regions are indicated with brackets, and the Shine-Dalgarno sequence and the ATG start codon are indicated with double underlines. The transcription start site, as previously determined (Jonsson et al., 1992), is indicated with an asterisk.