

Figure S1. Heat-killed *B. burgdorferi* bind less efficiently than live *B. burgdorferi* to the vasculature of mice. C3H/HeN mice were retro-orbitally inoculated with 1×10^8 total live or heat-killed (dead) infectious *B. burgdorferi* strain B31-A3. Bacteria burdens were determined for each perfused tissue 1 hr post inoculation. Significantly fewer heat-killed bacteria were bound to the vasculature of the lung, liver, heart, spleen, bladder, and joint after perfusion than infectious WT. No significant difference was observed in the ear of mice inoculated with live compared to heat-killed bacteria. The heat-killed bacteria were also cleared from the bloodstream more efficiently than the living bacteria. *=Statistical significance determined by Mann-Whitney unpaired t-test, p-value < 0.0079, n=5.

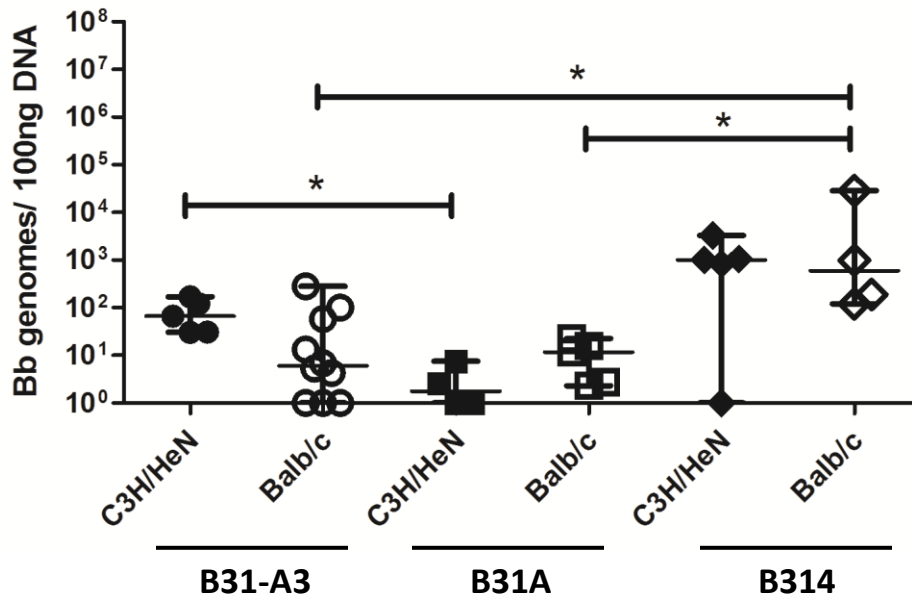


Figure S2. Blood burdens of *B. burgdorferi* strain B31-A3 and B31A are similar in C3H/HeN and Balb/c mice, while B314 shows increased blood burdens in Balb/c mice. Balb/c and C3H/HeN mice were retro-orbitally inoculated with 1×10^8 total infectious B31-A3, or non-infectious strains B31A or B314. After 1 hour, blood was collected and bacterial genomes were enumerated in each sample. Blood burdens of B314 were significantly higher than infectious strain B31-A3 (p-value=0.0129) and non-infectious strain B31A (p-value=0.0159) in Balb/c mice. There was not a significant difference in B31A and B31-A3 burdens in the bloodstream between C3H/HeN and Balb/c mice 1 hour post inoculation. *=Statistical significance as determined by non-parametric Mann-Whitney t-test, p-value<0.05). Balb/c data are replicated from Figure 1 and supplementary Figure S1.

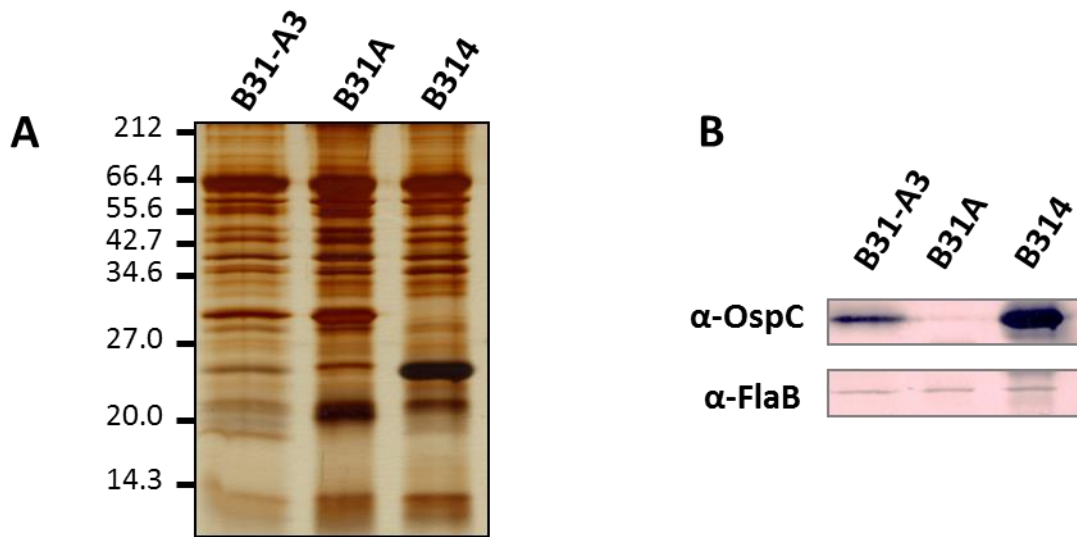


Figure S3. Variation in protein profile of infectious and non-infectious *B.*

***burgdorferi* strains.** Infectious *B. burgdorferi* strain B31-A3 and non-infectious strains B31A and B314 grown in OspC-inducing culture conditions were lysed and separated by 15% SDS-PAGE. (A) Gel was silver-stained to detect total protein. (B) Immunoblotting was performed on the separated proteins and incubated with antibody to detect OspC (α -OspC) and Flagellin (α -FlaB).

Table S1. Oligonucleotides for qPCR

Primer Name	Target	Strain of <i>Bb</i> Detected	Annealing Temp. (°C)	Sequence	Reference
RecA nTM16.Fwd	<i>RecA</i>	B31-A3	63.3	5'-gtggatctattgtattagatgaggctctcg-3'	(1)
RecA nTM16.Rev	<i>RecA</i>	B31-A3	63.3	5'-gccaaagtctgcaacattaacacctaag-3'	(1)
GCB908 B31A RecA.Fwd	<i>RecA</i>	B31A	58.3	5'- gcagctatcccaccttctt-3'	<i>This study</i>
GCB908 B31A RecA.Rev	<i>RecA</i>	B31A	58.3	5'- atgaggctctcggcattg-3'	<i>This study</i>
β -actin.Fwd	β -actin	B31-A3	65.0	5'- tcaccacactgtgccatctacga-3'	(1)
β -actin.Rev	β -actin	B31-A3	65.0	5'-ggatgccacaggattccatacca-3'	(1)

1. Ristow LC, Miller HE, Padmore LJ, Chettri R, Salzman N, Caimano MJ, Rosa PA, Coburn J. 2012. The β 3-integrin ligand of *Borrelia burgdorferi* is critical for infection of mice but not ticks. *Mol Microbiol* **85**:1105-1118.