

SUPPLEMENTAL FIGURES

The *Borrelia burgdorferi* infection critical BBK13 protein forms large oligomers in the spirochete membrane.

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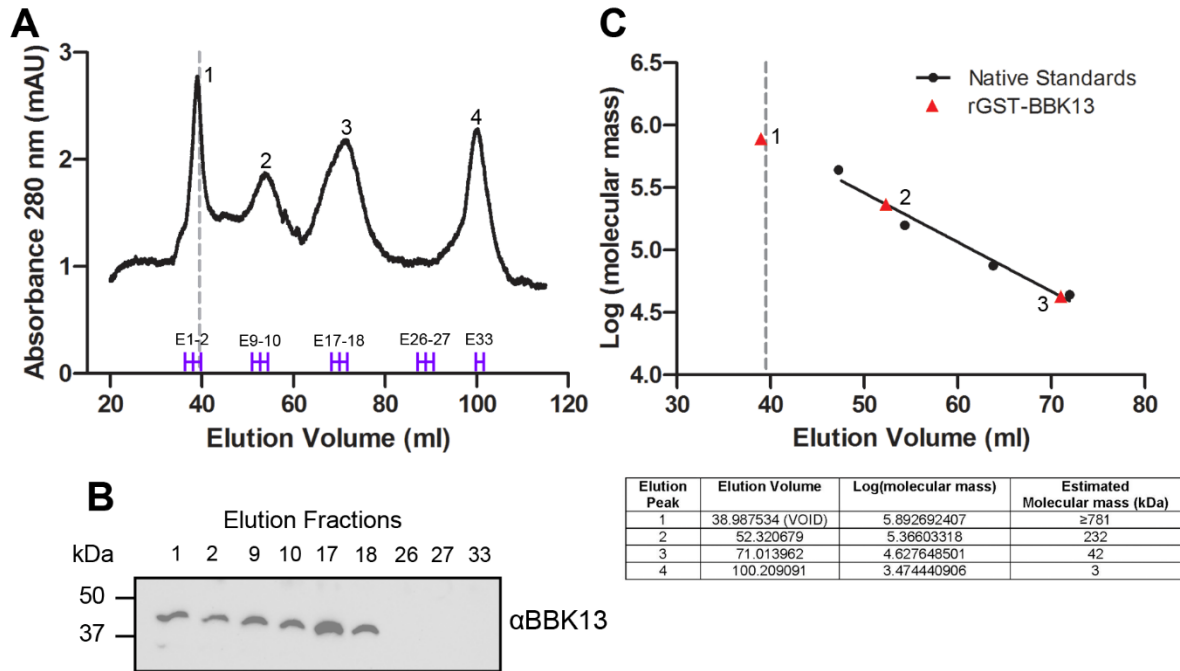


Figure S1. Recombinant BBK13 complex size estimation by SEC.

(A) Analytical size exclusion chromatography (SEC) of 500 μ g purified rGST-BBK13 via FPLC using High Prep 16/60 Sephacryl 300 resin. The void volume is indicated by the gray dashed line and the elution fractions used for BBK13 immunoblot analysis are indicated with purple brackets. Absorbance peaks are labeled 1-4. (B) Immunoblot analysis of select SEC elution fractions using anti-BBK13 antibodies (α BBK13). Molecular mass is shown in kilodaltons (kDa). (C) Standard curve plot of the elution volumes of the molecular mass standards (Native standards) were plotted versus the log of the molecular masses. The estimated molecular masses of the SEC absorbance peaks are shown the table and the rGST-BBK13 complexes are mapped on the standard curve plot (red triangles).

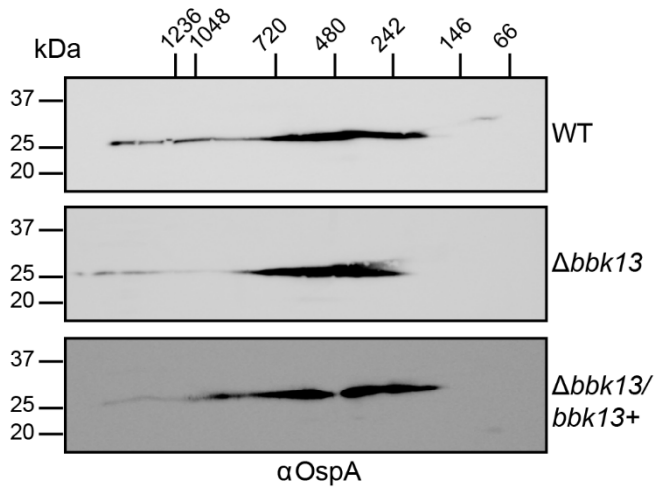


Figure S2: Detection of OspA membrane protein complexes by 2D-Blue native-PAGE.

Total membrane protein complexes isolated from wild type, $\Delta bbk13$ and $\Delta bbk13/bbk13+$ *B. burgdorferi* were resolved by two dimensional blue native-PAGE followed by immunoblot for OspA (α OspA). Molecular mass standards for the second dimension SDS-PAGE (left) and the first dimension BN-PAGE (top) are shown in kilodaltons (kDa).

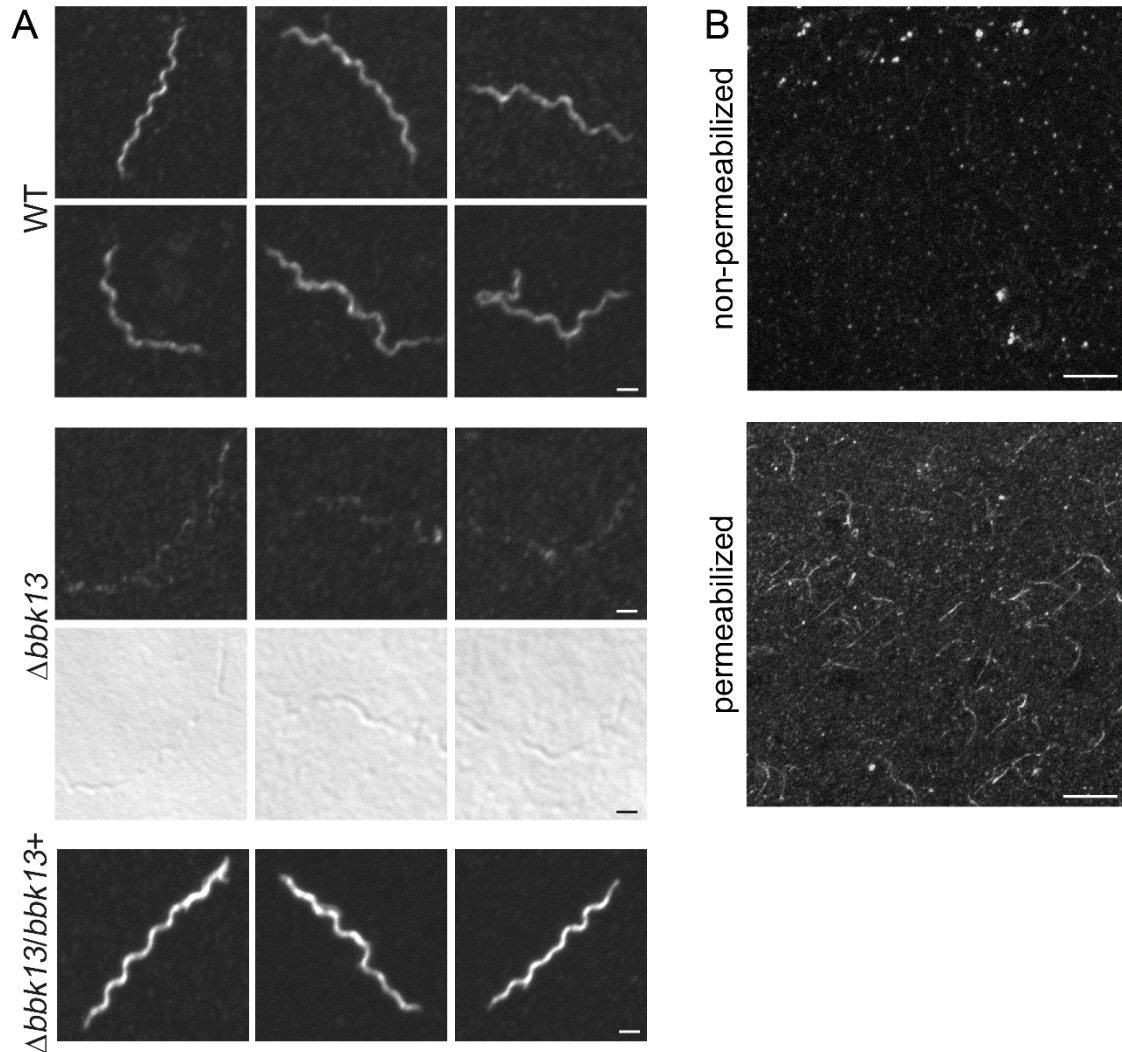


Figure S3. Immunofluorescence reveals uniform localization of BBK13 across the spirochete. Immunofluorescence assay of log phase grown *B. burgdorferi* probed with biotinylated anti-BBK13 antibodies, detected with Alexa 488-conjugated streptavidin and imaged by confocal fluorescent microscopy. (A) Formaldehyde-fixed wild type (WT), $\Delta bbk13$ and $\Delta bbk13/bbk13+$ *B. burgdorferi*. Representative fluorescent and phase contrast ($\Delta bbk13$ only) images are shown. A scale bar of 1 μ m is represented by a white or black line. (B) Heat fixed wild type *B. burgdorferi* treated with (permeabilized) or without (nonpermeabilized) 0.1% triton-X-100. A scale bar of 20 μ m is represented by a white line.