



Supplementary Figure S4 Characterization of the  $\Delta hslUV$  and  $C\Delta hslUV$  mutants.

(A) Identification of the  $\Delta hslUV$  and  $C\Delta hslUV$  mutants by PCR. Lane M: DNA marker (Fermentas, USA). Lane 1: amplicon (3867 bp) of the 5'arm-hsIV/U-3'arm (3627 bp) plus two extending regions (120 bp each) from wild-type *L. interrogans* strain Lai for control. Lane 2: amplicon (3189 bp) of the 5'arm-pflgB-kan-3'arm segment (2949 bp) plus two extending regions (120 bp each) from the  $\Delta hslUV$  mutant for identification. Lane 3: amplicon (6581 bp) of the 5'arm-hsIV/U-pflgB-spc-3'arm (6341 bp) plus two extending regions (120 bp each) from the  $C\Delta hslUV$  mutant for identification.

(B) Schematic diagram of sequencing result of the  $\Delta hslUV$  mutant. The positions of PCR primers used are marked below.

(C) Schematic diagram of sequencing result of the  $C\Delta hslUV$  mutant. The positions of PCR primers used are marked below.

(D) Growth curves of the  $\Delta hslUV$  and  $C\Delta hslUV$  mutants compared to wild-type *L. interrogans* strain Lai in EMJH liquid medium.

(E) Absence of HslU and HsIV proteins in the  $\Delta hslUV$  mutant, determined by Western Blot assay. Lane M: protein marker. Lane 1: HsIV or HslU protein expressed by wild-type *L. interrogans* strain Lai. Lane 2: absence of HsIV or HslU protein in the  $\Delta hslUV$  mutant. Lane 3: HsIV or HslU protein expressed by the  $C\Delta hslUV$  mutant.