

Table S7. PCR primers used in this study			
Target	Gene ID ^a	Use	Sequence ^b (5' to 3')
<i>km</i> cassette of plasmid pKD4	<i>kan</i>	PCR of pKD4 <i>km</i> cassette and flanking FRTs	GCGATTGTGTAGGCTGGAGCTGCTTC; ATGGGAATTAGCCATGGTCC
T6SS-4 upstream region	YPTB0639; y3677	Deletion of T6SS-4 locus	GCCATTCCAGATTATAGGC; gaagcagctccagcctacacCTAGTGTCTATCTGACATGCAAAGGATCTA AAATCTCTGGAGATTAAG
T6SS-4 downstream region	YPTB0657; y3657	Deletion of T6SS-4 locus	CCATGTTGCAACAAGCCGAAGC ; ggaccatggctaattcccatGTAGCGGCAGTCGTCAGACAAAAAATATC CGGGGGAATGCCCGGATATC
T6SS-4 locus	YPTB0639- YPTB0657, y3657-y3677	Verification of the T6SS-4 deletion	CGGCAAATAATACGTTTCGAGC; CCGAAACTTGTCAACTGACCC
<i>hcp</i> upstream region	YPTB643; YPO0502	Deletion of <i>hcp</i>	CGGCGAACGCCAGATTATCGGC; gaagcagctccagcctacacCATTTTTTCACTCCTGAATTAATGTCATTA AGTTGGG
<i>hcp</i> downstream region	YPTB643; YPO0502	Deletion of <i>hcp</i>	CGCAACTCTTCGTTGTATAGAGC; ggaccatggctaattcccattaagtcagaaatcattaagactgctctcgcgagtagg
<i>hcp</i>	YPTB643; YPO0502	PCR of <i>hcp</i>	CCATATCGAAGGTGTAGATGGTGAG; GCCATCAGGTTGCTGCTCTTTG
T6SS-4 locus gene	YPTB0639, y3677	Deletion	gaagcagctccagcctacacCTAGTGTCTATCTGACATGCAAAGGATCTA AAATCTCTGGAGATTAAG; ggaccatggctaattcccatCATATCTATGTCTCTTATTTTGGCTATTCAT CCACGTCATCGTGCTAATGG
<i>psaA</i> upstream region	YPTB1334; y2882	Deletion of <i>psaA</i>	CCATACGTCCTATCATGACG; gaagcagctccagcctacacCATGAGAACAGTCTCCATTAATGTAATAA TTGC
<i>psaA</i> downstream region	YPTB1334; y2882	Deletion of <i>psaA</i>	GGGAATTTTCCACCGGTCCG; ggaccatggctaattcccataAATAACGAGTAATCATAGAACATGATGGC TTGCTG
<i>psaA</i>	YPTB1334; y2882	PCR of <i>psaA</i>	CGCTTCTACTGTCATTAACTCC; GCCTAACGTACAGATGATC
<i>ddhB-gmd</i> O-antigen locus	YPTB1000- 1009; y1069- 1079	Deletion of <i>ddhB-gmd</i> locus	TGAGTTTATGGTTGCAAACCATGGGGGCAACGGTAAAAGGTTA CTCTATTCCGGGATCCGTCGACC; CTAAGAGCTAGACCGTAGTAAAACCAAAACC
<i>ddhB-gmd</i> O-antigen locus	YPTB1000- 1009; y1069- 1079	Deletion of <i>ddhB-gmd</i> locus	CATGCCACGATGCCCCGCAATAAATACGCGTTTCTTATCCATTT GTA GTGTAGGCTGGAGCTGCTTCG; CTAAGAGCTCTGATAACACCACCCGAG
<i>ddhB-gmd</i> O-antigen locus	<i>ddhB</i> , <i>ddhC</i> ; <i>wbyK</i> , <i>gmd</i>	Addition of <i>Y. pseudtb</i> O-Ag genes to <i>Y. pestis</i> KIM6+ Δ <i>ddhB-gmd</i> (1)	TGACGCATGCACATGTCGGATATTACCTT; CGGGATCCAATTGGCTCCCCAAATAACC; CGGGATCCTGCAAATATGGGAGGGGTA; CTAAGAGCTCGGCTTACGTCCATCAATTT
<i>ddhB-gmd</i> O-antigen locus	<i>prt</i> , <i>wbyH</i>	Addition of <i>Y. pseudtb</i> O-Ag genes to <i>Y. pestis</i> KIM6+ Δ <i>ddhB-gmd</i> (2)	TGACGCATGCATCGCGGAACTGAAGAGTA; CGGGATCCAGCCGTTTCTAAGGGGAGAT; CGGGATCCTCGCAAAGTTGCAAATGAA; CTAAGAGCTCATCCTGCACTCATCGCTTCT
<i>ddhB-gmd</i> O-antigen locus	<i>wzx</i> , <i>wzy</i> , <i>wbyK</i>	Addition of <i>Y. pseudtb</i> O-Ag genes to <i>Y. pestis</i> KIM6+ Δ <i>ddhB-gmd</i> (3)	TGACGCATGCAAAGAAATGCGTTATCC; CGGGATCCTCGGATTTTACCCTCAACTC; CGGGATCCTCGGATTTTACCCTCAACTC CTAAGAGCTCGCAAATATAACTCACTCACTT
<i>ddhB-gmd</i> O-antigen locus	<i>wbyI</i> , <i>wbyJ</i>	Addition of <i>Y. pseudtb</i> O-Ag genes to <i>Y.</i>	TGACGCATGCGTGGTCCGATTAATTTTG; CTAAGAGCTCACCAATCCATAATGCGATGC

		<i>pestis</i> KIM6+ <i>ΔddhB-gmd</i> (4)	
<i>chb</i> (<i>nghA</i>)	YPTB1123	Replacement with <i>Y. pestis</i> pseudogene	TGACGCATGCATGACAACCACCAATATCC; CTAAGAGCTCGGCTTCTTTCTCTTTACCCTG
<i>flhD</i>	YPTB2409	Replacement with <i>Y. pestis</i> pseudogene	TGACGCATGC AGCGGGTACACTGATTGCT; CTAAGAGCTC GAAAACCGCACCTAAGACCA
<i>chiC</i>	YPTB3365-3366	Replacement with <i>Y. pestis</i> pseudogene	TATCAGAAATAACATTAATC GTCGCTAAGGGTAAG; GATCTCTAGATTCTACGATGACATCTCATAATA CGCGCATGCCTTGATTGTGGCATAAGCGGTC
<i>barA</i>	YPTB0750	Replacement with <i>Y. pestis</i> homolog	TGACGCATGC TATCCAACGTACGCACCTGA; CTAAGAGCTC AAATGACCTTGGGTCTGCTG
YPTB2490 upstream region	YPTB2490	Deletion of YPTB2490-2497	TGACGCATGCCATCGGGACAAATTTATGG; CGGGATCCGCTACGGATCATCCCTTCA
YPTB2497 downstream region	YPTB2497	Deletion of YPTB2490-2497	CGGGATCCGATTGGCATGTTCTGGTCT; CTAAGAGCTCGTCAGACCGTCCCACTCAAT
YPTB0556 upstream region	YPTB0562	Deletion of YPTB0556-0562	TGACGCATGCCAAAGGGAATTAGTGG; CGGGATCCAGCTTCAACCACAAAAGCA
YPTB0562 downstream region	YPTB0556	Deletion of YPTB0556-0562	CGGGATCCAATCGCTCTATCGCATTCA; CTAAGAGCTCTGCATCACATTGCGACACTA
YPTB0871 upstream region	YPTB0871	Deletion of YPTB0871-0880	TGACGCATGCTCTGCTGGATTGCAAAGTG; CGGGATCCTGCCAAACCTGCATAC
YPTB0880 downstream region	YPTB0880	Deletion of YPTB0871-0880	CGGGATCCGACGAATGGTATTGGTGTG; CTAAGAGCTCACCGTGGCTATTGTCTGAA
<i>nhaC</i> locus	y3550-3555	Deletion of y3550-3555	AGCAACAGTGAATTATGACAATGTCCTATGGAAATCCGCTGT TAGAGAGttgtcgggaagatgcgtg; TCCGCTCAGATCATGCAGTCACCCGATTAGCCATACCCATAAA TACAACGCcggtcgtatgtgtgtg
<i>lsrR</i>	YPTB0553	Cloning of <i>Y. pseudotub</i> <i>lsrR</i>	TGGATTTACCCGCGCCATTTTC; ATCGAGTTGACTCATTGCGCG
PDE4	YPTB3828	Cloning of <i>Y. pseudotub</i> PDE4 gene	CGCCACCAATGACAACAGGAT; CCCAGTTATCGGTCTCCACTTC
PDE4	YPTB3828	Deletion of <i>Y. pseudotub</i> PDE4 gene (inverse PCR)	GGCACTCCCAGTGTGTTTTGCGGCTAACGCTCCCTAAG; CTGTTGGTCATCATCAGTTTAGTCGCCATGTTTCAGGGTA
^a gene annotation number for <i>Y. pestis</i> KIM and <i>Y. pseudotuberculosis</i> IP32953 ^b sequences complementary to the kanamycin resistance cassette are in lowercase letters; 6-nt sequences in bold type are restriction enzyme sites added to facilitate cloning			