

Table S1 - Plasmids and Primers used in this study

Plasmids	Description	Reference
<i>pKOR1</i>	empty plasmid for allelic replacement	1
<i>pKOR1+pbp4</i>	<i>pKOR1</i> deletion construct for <i>pbp4</i> gene	This study
<i>pTX_Δ</i>	empty plasmid for constitutive expression	2
<i>pTX_Δ+pbp4</i> (SF8300)	constitutively expressed <i>pbp4</i> from SF8300 strain	This study
<i>pTX_Δ+pbp4</i> (SRB)	constitutively expressed <i>pbp4</i> from SRB strain	This study
<i>pTX_Δ+pbp4</i> (SRT)	constitutively expressed <i>pbp4</i> from SRT strain	This study

Primers	Sequence (5'-3')	Use
PBP4-P1	GGGGACAAGTTTGTACAAAAAAGCAGGCTAGTTTGAATTTGACATTGTGACTTGTGCGATATCTTTGCATAATACGACC	<i>pbp4</i> deletion
PBP4-P2	AAAGCGTTAATCTTCCCTTTTCCAATTCTTAAATATTCCTAAAAAGC	<i>pbp4</i> deletion
PBP4-P3	AAAAGGGAAGATTAACGCTTAACTACTAAAAACGGACAAGTTGCACATTATAAAGCTGCGAAACTTGTCCG	<i>pbp4</i> deletion
PBP4-P4	GGGGACCACTTTGTACAAGAAAGCTGGGTGAAGATTTTAAATAGATATATCACAGAAATTATGAAAAATAAGACAACG	<i>pbp4</i> deletion
PBP4- <i>Bam</i> H1 -for	TAAGGATCCGAAAAGGGAAGATTAACGCTTTATGAAAAATTTAATATC	<i>pbp4</i> cloning in <i>pTX_Δ</i>
PBP4- <i>Mlu</i> 1 -rev	TAACACGCGTTATTATAAAATAAATATAAAACGGACAAG	<i>pbp4</i> cloning in <i>pTX_Δ</i>
PBP4 pt. mut P1	GGGGACAAGTTTGTACAAAAAAGCAGGCGAGTAAGTTTGCTCTTCGTACAACATTATAACACCTTTAGCTACACAG	<i>pbp4</i> point mutation
PBP4 E183A mut 1	TACACATTTTCGTCAATCCAACGGGTGCTGCAAATTCAGATTACGTACATTTGCACC	<i>pbp4</i> point mutation
PBP4 E183A mut 2	TCTTGAATTTGCAGCACCCGTTGGATTGACGAAATGTGTATTTTTCATTC	<i>pbp4</i> point mutation
PBP4 F241R mut 1	ACGTATTACACACGCAACTTTTTCATTGGAAGGTGCAAAAATGAGTTTGC	<i>pbp4</i> point mutation
PBP4 F241R mut 2	TTCCAATGAAAAGTTGCGTGTGTAATACGTAACGTCATGCGTTTGGTG	<i>pbp4</i> point mutation
PBP4 pt. mut P2	GGGGACCACTTTGTACAAGAAAGCTGGGTAGAAGGCATTTTCGACGACTAAAATCAAACAAGAATTATATGGTAAAGATGC	<i>pbp4</i> point mutation
<i>AcrB</i> pt. mut P1	GGGGACAAGTTTGTACAAAAAAGCAGGCAGAAAATGGTATTTCTGCAAGTCAACTTGAATGCACTTGAATGAAAACCTACCAG	<i>acrB</i> point mutation
<i>AcrB</i> I960V mut 1	TAGTAACAAATGCCGTTGTGTTAATAGACCGTGTATTATAATAATG	<i>acrB</i> point mutation
<i>AcrB</i> I960V mut 2	TAACACGGTCTATTAACACAACGGCATTGTTACTACGATTCC	<i>acrB</i> point mutation
<i>AcrB</i> pt. mut P2	GGGGACCACTTTGTACAAGAAAGCTGGGTGATATAAAGTAGTCAATATCATGACCGCTTTTATGGCTTTTATATTATCTATCG	<i>acrB</i> point mutation
<i>GdpP</i> pt. mut P1	GGGGACAAGTTTGTACAAAAAAGCAGGCATCGATTTAAGTTTGAATTTGAAGCGAATCATGAAAAATAATTCTAAAAAGTG	<i>gdpP</i> point mutation
<i>GdpP</i> N182K mut 1	ACATTATTTTATAGATAAATACGATGAGATTACGCAAAATATGAATG	<i>gdpP</i> point mutation
<i>GdpP</i> N182K mut 2	TTGCGTAATCTCATCGTATTTATCTAAAAATAATGTCGCAATG	<i>gdpP</i> point mutation
<i>GdpP</i> pt. mut P2	GGGGACCACTTTGTACAAGAAAGCTGGGTATGCTTCTAAATTATTCATCATTGCAAATCTAGACACACCGATTGCTGCACC	<i>gdpP</i> point mutation

Figure S1

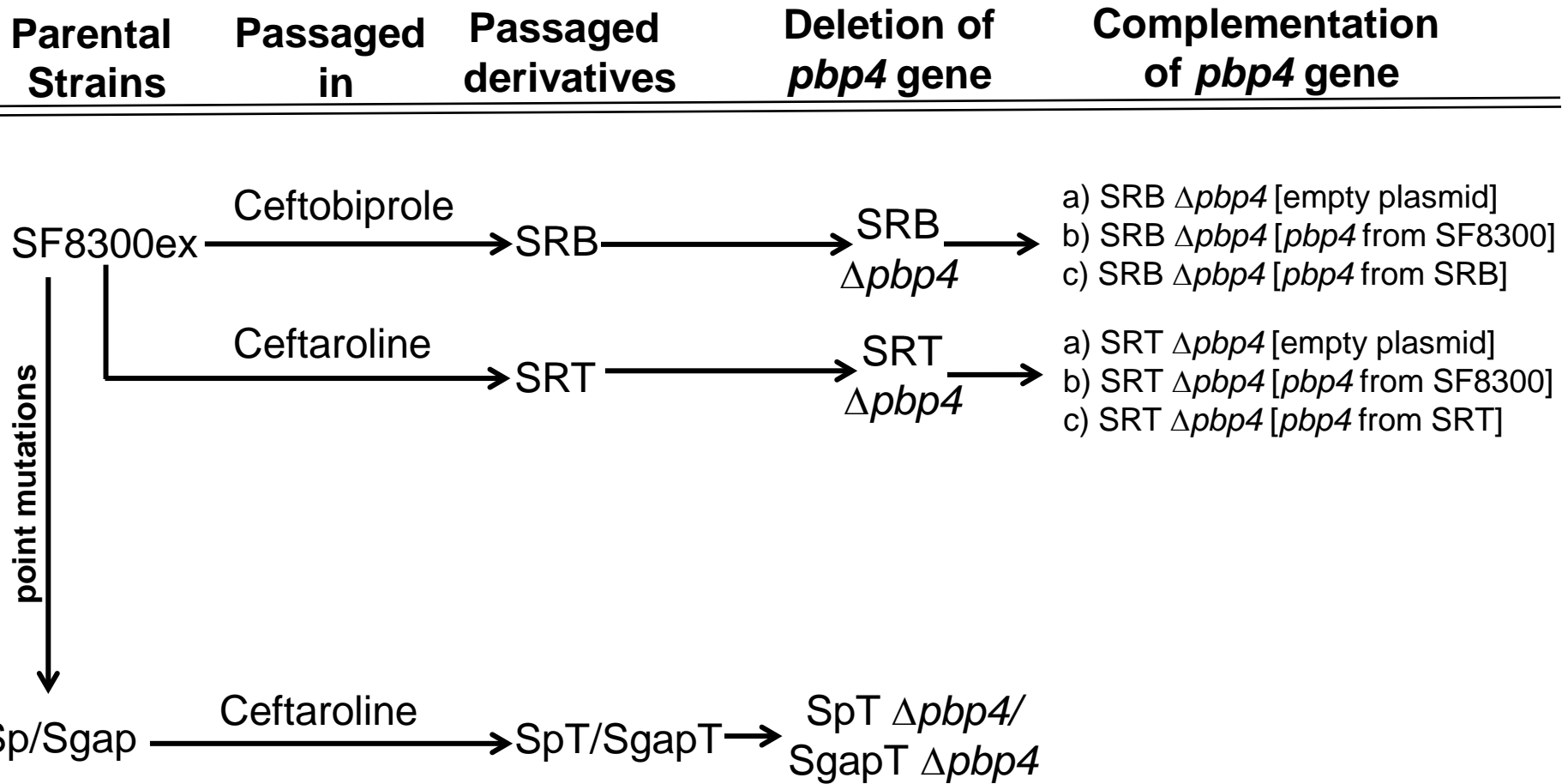


Figure S1: Scheme of passaging experiments against new generation cephalosporins.

References

1. **Bae T, Schneewind O.** 2006. Allelic replacement in *Staphylococcus aureus* with inducible counter-selection. *Plasmid* **55**:58-63.
2. **Wang R, Braughton KR, Kretschmer D, Bach TH, Queck SY, Li M, Kennedy AD, Dorward DW, Klebanoff SJ, Peschel A, DeLeo FR, Otto M.** 2007. Identification of novel cytolytic peptides as key virulence determinants for community-associated MRSA. *Nat Med* **13**:1510-1514.