## Supplemental Figure Legends.

**Supp. Fig. 1. Screen shots from CLC Genomics Workbench.** In the local alignment (top), the DAP-B IS*256* insertion is not detected by polymorphism analysis. In the global alignment (bottom), the DAP-B IS*256* insertion is detected as multiple, consecutive polymorphisms surrounding the insertion site.

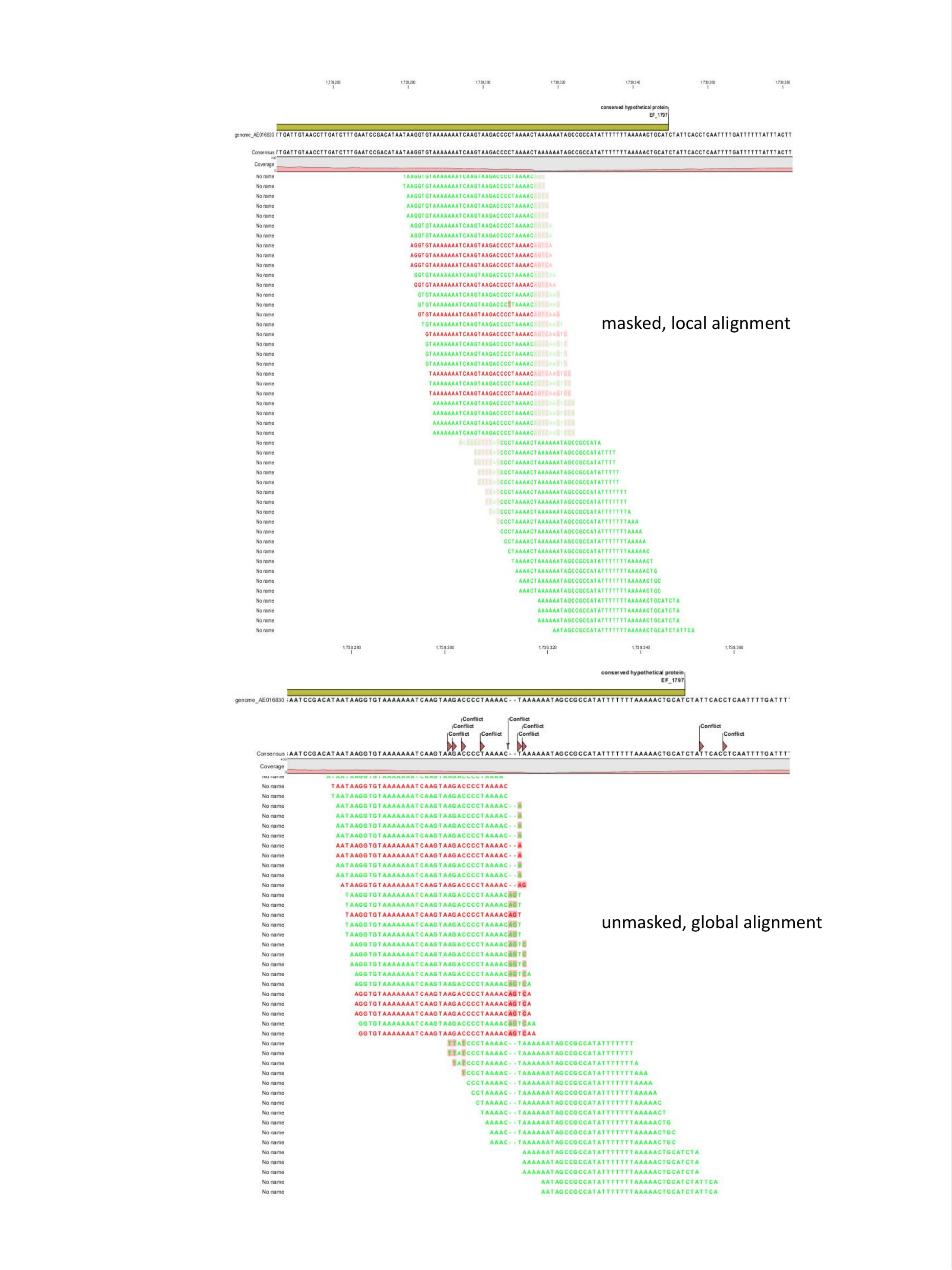
**Supp. Fig. 2. Alignment of all *E. faecalis* EF0631 protein sequences from this study.** Identical amino acids are shaded in grey; amino acid changes are shown in white. The location of the DAP-A N77_Q79del is boxed in green. Predicted PLD motifs are shown. The location of the DAP-B and DAP-C R218Q substitution is indicated by an asterisk. Amino acids encoded after the frameshift mutation in *E. faecalis* T2 are not shown.

**Supp. Fig. 3. Alignment of EF0631 and EF1608 predicted cardiolipin synthases.** Identical and similar amino acids are shaded in grey; non-similar amino acids are shown in white. The location of the DAP-A N77_Q79del is boxed in green. Predicted PLD motifs are shown. The location of the DAP-B and DAP-C R218Q substitution is indicated by an asterisk.

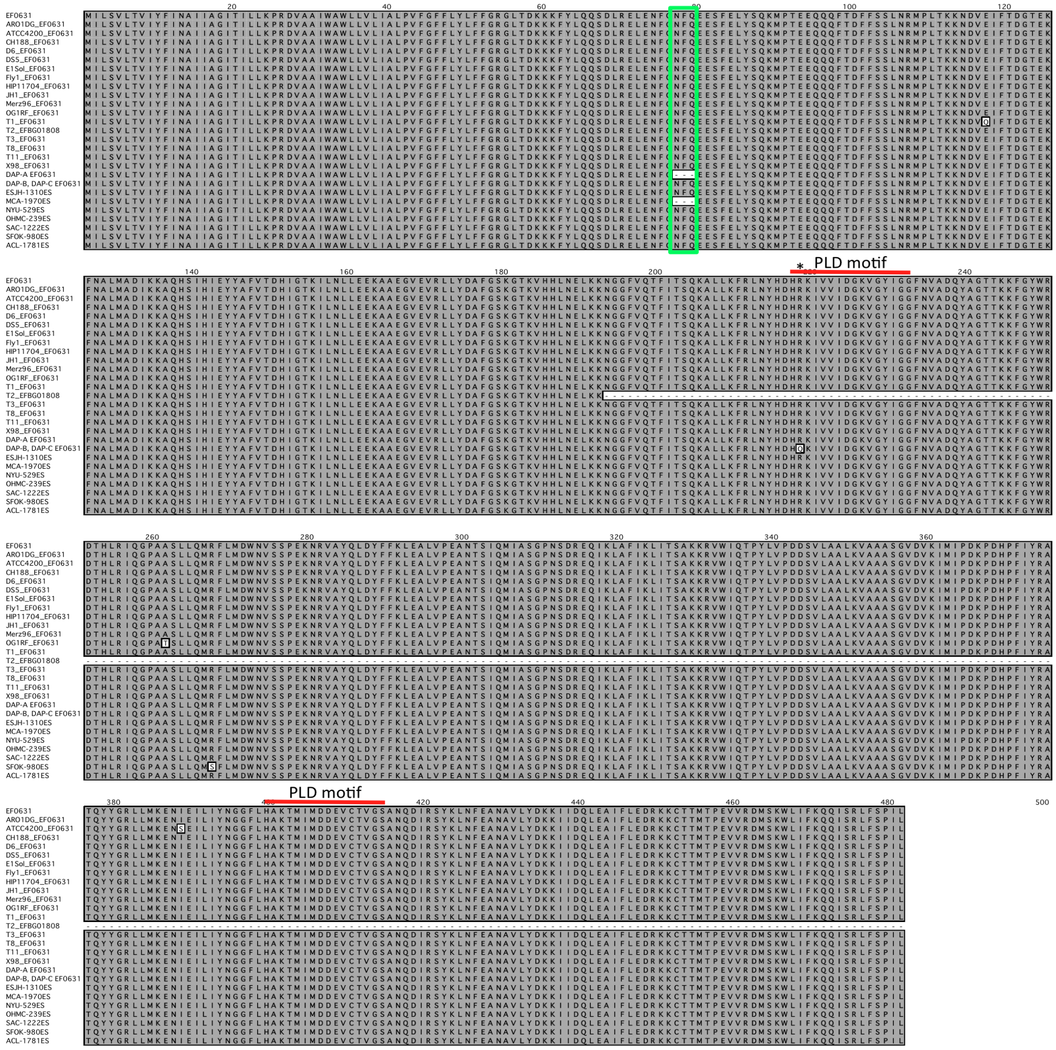
**Supp. Fig. 4. Alignment of EF1797 sequences from *E. faecalis*, *E. faecium* and *Listeria* sp.** Identical and similar amino acids are shaded in grey; non-similar amino acids are shown in white. The green bar indicates the location of the DAP-C G130_F154del.

**Supp. Fig. 5. Alignment of EF0631 sequences from *E. faecalis* and *E. faecium*.** Identical amino acids are shown in dark grey, similar amino acids are shaded in light grey, and non-similar amino acids are shown in white. The location of the DAP-A N77_Q79del is boxed in green. Predicted PLD motifs are shown. The location of the DAP-B and DAP-C R218Q substitution is indicated by an asterisk.

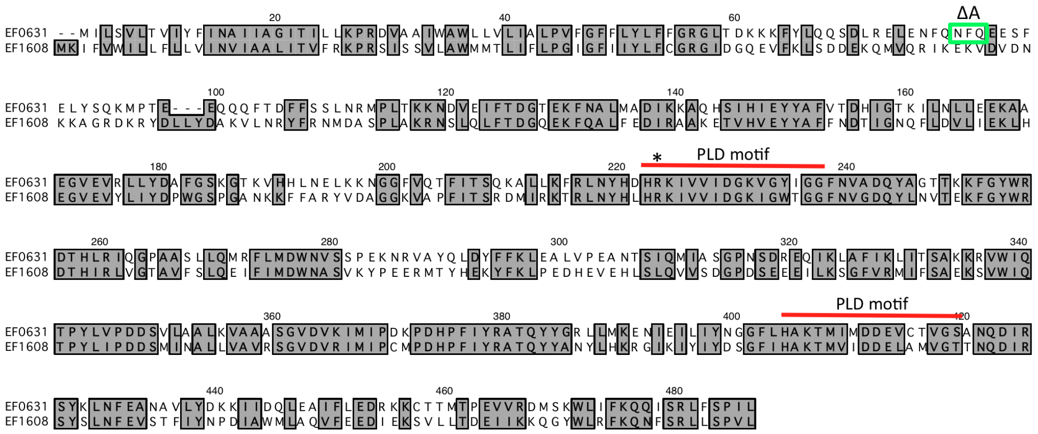
## Supplemental Fig. 1



## Supplemental Fig. 2.



Supplemental Fig. 3.



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EF0631 --MILSVLTVIYFINAIIAGITILLKPRDVAAIWAWLLVLIALPVFGFFLYLFFGRGLTDKKKFYLQQSDLRELENFQNFQEESF
EF1608 MKIFVWILLFLLVINVIAALITVFRKPRSISSVLAWMMTLIFLPGIGFIIYLFCGRGIDGQEVFKLSDDEKQMVQRIKEKVDVDN

EF0631 ELYSQKMPTE---EQQQFTDFFSSLNRMPLTKKNDVEIFTDGTEKFNALMADIKKAQHSIHIEYYAFVTDHIGTKILNLLEEKAA
EF1608 KKAGRDKRYDLLYDAKVLNRYFRNMDASPLAKRNSLQLFTDGQEKFQALFEDIRAAKETVHVEYYAFFNDTIGNQFLDVLIEKLH

EF0631 EGVEVRLLYDAFGSKGTKVHHLNELKKNGGFVQTFITSQKALLKFRLNYHDHRKIVVIDGKVGYIGGFNVADQYAGTTKKFGYWR
EF1608 EGVEVYLIYDPWGSPGANKKFFARYVDAGGKVAPFITSRDMIRKTRLNYHLHRKIVVIDGKIGWTGGFNVGDQYLNVTEKFGYWR

EF0631 DTHLRIQGPAASLLQMRFLMDWNVSSPEKNRVAYQLDYFFKLEALVPEANTSIQMIASGPNSDREQIKLAFIKLITSAKKRVWIQ
EF1608 DTHIRLVGTAVFSLQEIFIMDWNASVKYPEERMTYHEKYFKLPEDHEVEHLSLQVVSDGPDSEEEILKSGFVRMIFSAEKSVWIQ

EF0631 TPYLVPDDSVLAALKVAAASGVDVKIMIPDKPDHPFIYRATQYYGRLLMKENIEILIYNGGFLHAKTMIMDDEVCTVGSANQDIR
EF1608 TPYLIPDDSMINALLVAVRSGVDVRIMIPCMPDHPFIYRATQYYANYLHKRGIKIYIYDSGFIHAKTMVIDDELAMVGTTNQDIR

EF0631 SYKLNFEANAVLYDKKIIDQLEAIFLEDRKKCTTMTPEVVRDMSKWLIFKQQISRLFSPIL
EF1608 SYSLNFEVSTFIYNPDIAWMLAQVFEEDIEKSVLLTDEIIKKQGYWLRFKQNFSRLLSPVL
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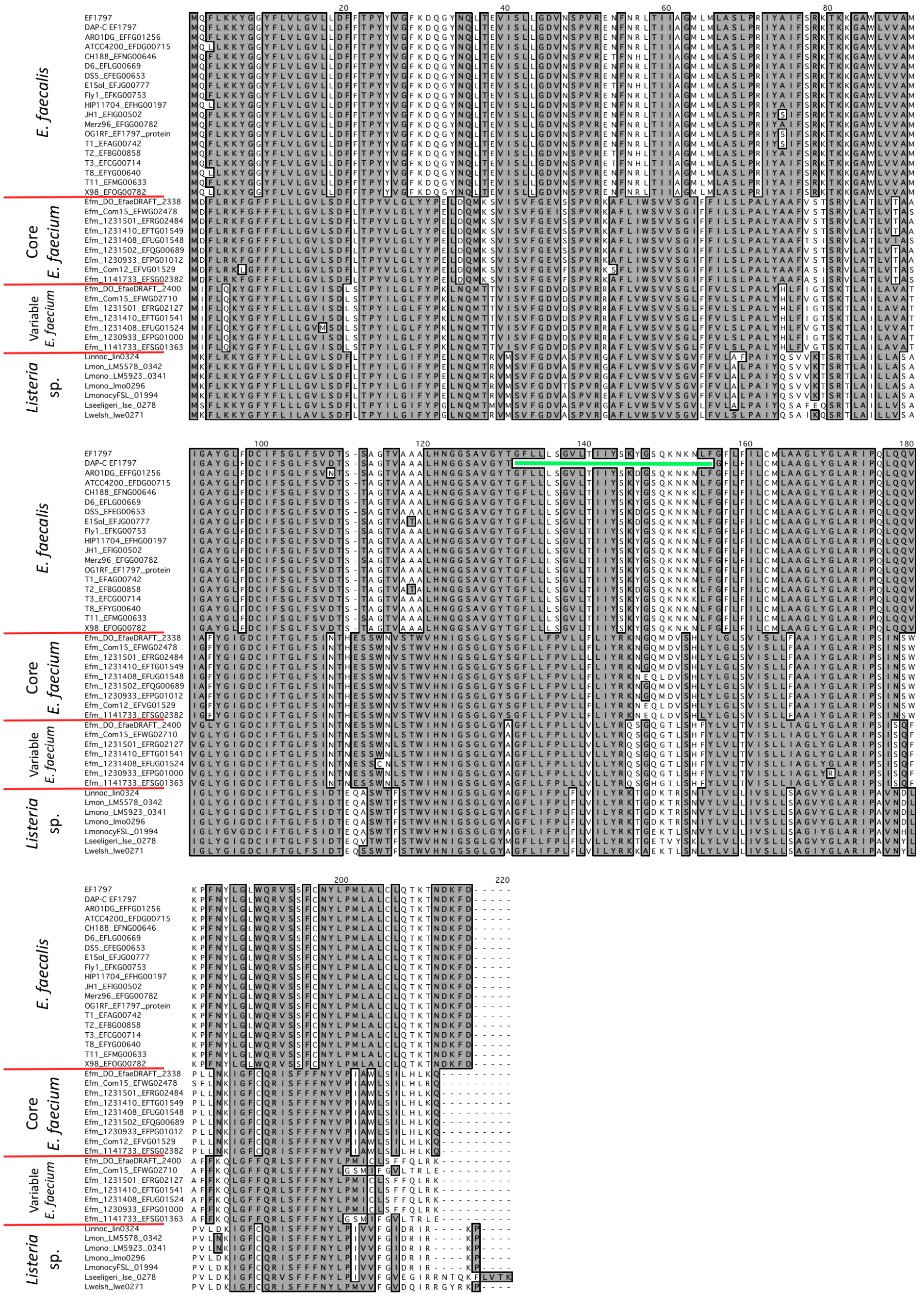
Supp. Figure 4.



Supp. Figure 5.