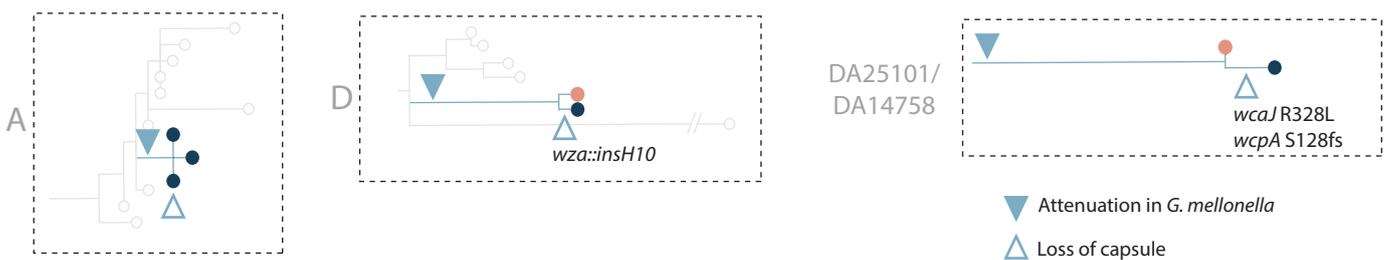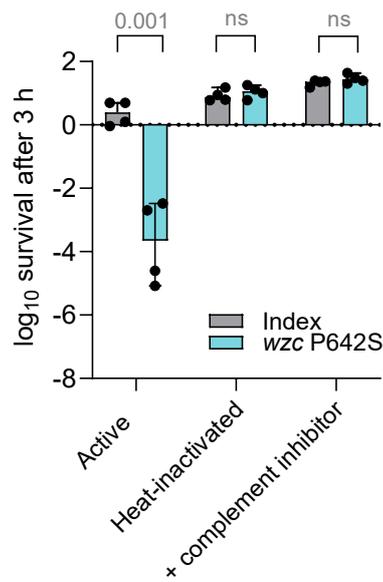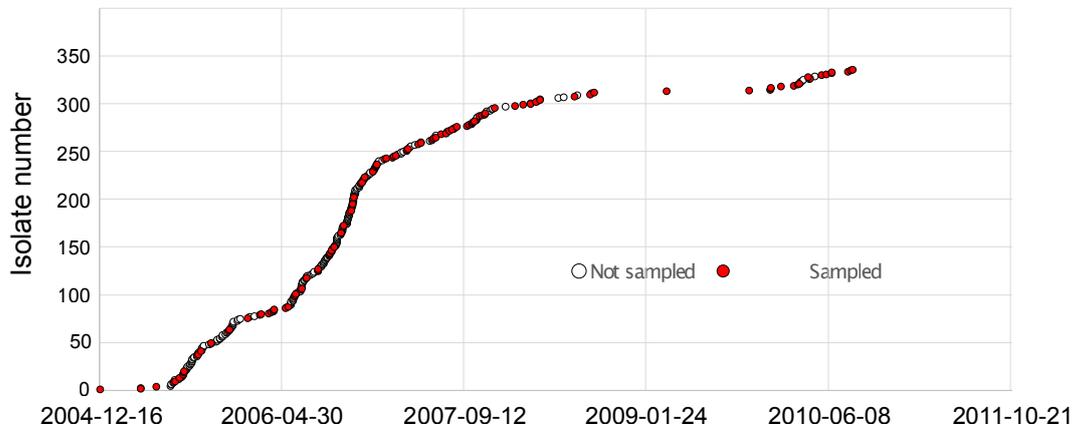# Supplementary Information

Parallel within-host evolution alters key virulence factors
in an opportunistic *K. pneumoniae* clone during a hospital outbreak

Greta Zaborskytė, Karin Hjort, Birgitta Lytsy, Linus Sandegren

Supplementary Fig. 1 | Cumulative sampling graph of isolates over time of the outbreak. Red circles denote sampled isolates in this study.

Supplementary Fig. 2 | Serum killing control with active serum, heat-inactivated serum and complement inhibitor polyanetholesulfonic acid. Results from four biological replicates with 95% CI are shown. Difference between the index isolate and *wzc* P642S mutant in each condition tested by unpaired t-test, ns - not significant.


Supplementary Fig. 3 | Phylogenetic indications of mutations reducing killing of *G. mellonella* (blue triangles) and capsule-loss mutations (white triangles) in clusters A, D, and DA25101/DA14758.
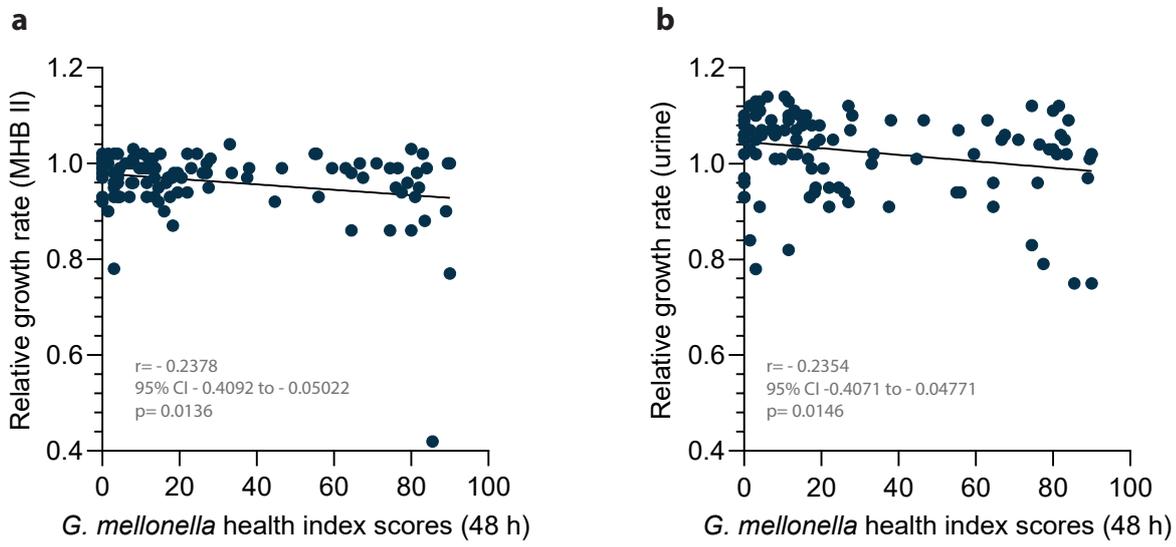
Supplementary Fig. 4 | Exponential growth rates in (**a**) MHB II medium and (**b**) urine vs health index scores in *G. mellonella* larvae at 48 h post-injection. Exponential growth rates refer to means of five biological replicates. Pearson correlation coefficients (r) with 95% CI and a two-tailed p value are shown.
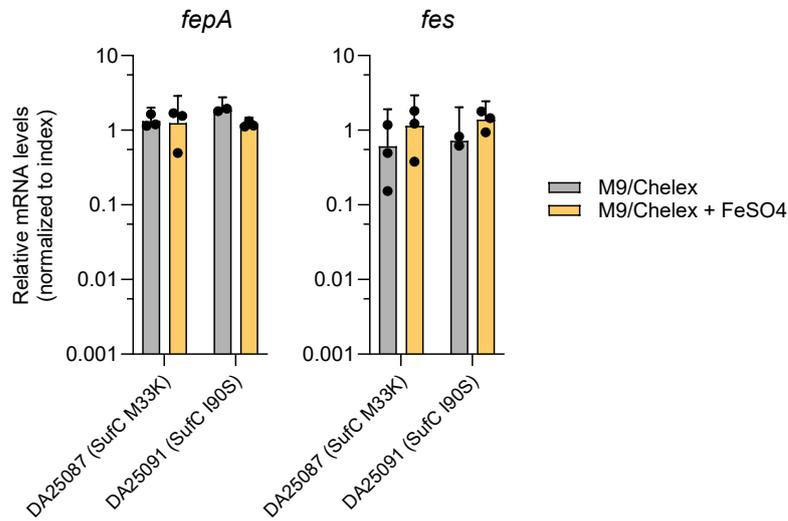
Supplementary Fig. 5 | mRNA levels of *fepA* and *fes* in control isolates with *sufC* mutations but without any *fepA/fes* changes relative to the index isolate. mRNA levels (mean of n=3 with 95% CI) are relative to the index isolate for the respective conditions. Comparison to the index isolate was done by an unpaired t-test with Bonferroni corrections for two comparisons; none of the comparisons were significant.
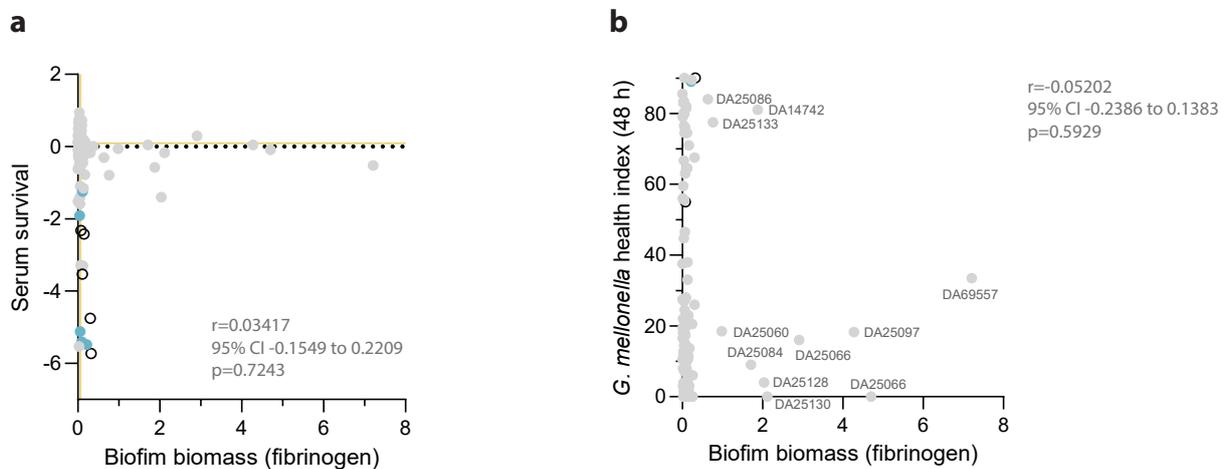
Supplementary Fig. 6 | Interconnection between biofilm biomass on silicone with fibrinogen and (**a**) survival in human serum and (**b**) health index scores of *G. mellonella* at 48 h. Blue dots indicate isolates with *wzc* missense mutations. Pearson correlation coefficients (r) with 95% CI and two-tailed p values is shown.