



**Figure S1.** Secondary pneumococcus infection after influenza virus infection with PR8 and CAL09 exacerbates disease and mortality in BALB/c mice. Mice received one intraperitoneal sham-vaccination with buffer and on day 21 were infected with 100 TCID<sub>50</sub> of influenza PR8 or 1000 TCID<sub>50</sub> CAL09 followed by secondary bacterial infection with  $1.25 \times 10^6$  CFU *S. pneumoniae* five days after viral infection. Sham-infected mice received buffer on the respective days. **(A)** Viral pulmonary titers one day before and two days after bacterial infection as well as **(B)** bacterial pulmonary loads two days post bacterial infection was assessed in 3-5 mice per group. Symbols represent the titer of an individual mouse and lines and error bars indicate group mean titers  $\pm$  SEM. The dashed line at 1.5  $\log_{10}$  EID<sub>50</sub>/mL and 10 CFU/mL indicate the LLOD of the respective assays. **(C)** Weight loss and **(D)** mortality was monitored over a period of 17 days post viral infection. Animals that lost 30% of their initial body weight (dashed line) were scored dead and humanely euthanized. Data represent the mean weight of each group  $\pm$  SEM. Differences in viral pulmonary titers on day seven were analyzed by Two-Way ANOVA with a Tukey post-hoc test and bacterial loads were analyzed by One-Way ANOVA with a Tukey correction factor. Weight loss curves were analyzed by multiple t-tests with a Holm-Sidak correction factor, survival curves were analyzed by a Log-rank test. Asterisks indicate the significance level with \*  $p_{adj} < 0.05$ , \*\*  $p_{adj} < 0.01$  and \*\*\*\*  $p_{adj} < 0.0001$ .