# Sequential infection experiments for quantifying innate and adaptive immunity during influenza infection

## Text S2:
## Notes on biologically plausible ranges for the parameters $p_{Vratio}$, $\alpha$ and $\gamma$

First, we examine the parameter $\alpha$. The concentration of virions in nasal wash is lower than the concentration of virions in the respiratory tract by a factor of $a = 1 - 100$ [1]. The volume of the ferret upper respiratory tract, which was assumed to be the site of infection, is approximately $v = 1$mL [2], while the concentration of nasal wash was reported per $b = 0.1$mL [3]. We assumed that 1 RNA copy number corresponded to one (infectious or non-infectious) virion. Hence, the number of RNA copies/100$\mu$L of nasal wash corresponding to one virion in the respiratory tract ($\alpha$) was $(b/v)/a$. Based on the bounds for $a$ and $b$, the bounds for $\alpha$ were then $[10^{-3}, 10^{-1}]$.

Next, we examine the parameter $\gamma$. Petrie *et al.* [4] used the bounds $[10^0, 10^5]$ to fit the initial ratio of total to infectious virions. These bounds were in units of RNA copy number/$\text{TCID}_{50}$ ($f$). These bounds were based on the variability across different inocula given to donor ferrets in the experiments by Butler *et al.* [5]; the study by Petrie *et al.* [4] analysed data from these experiments. However, in our study, $\gamma$ was required to be in units of RNA copy number/virion, as we accounted for the loss of a single virion due to its infecting a target cell. Assays to measure the amount of infectious virus cannot measure the number of virions directly; hence, we needed to estimate the conversion factor between $\text{TCID}_{50}$ and virions ($h$). At the minimum, one virion was required to establish an infection; hence, the lower bound for the number of virions corresponding to 1 $\text{TCID}_{50}$ was 1 virion. Handel *et al.* [1] estimated that 1 $\text{TCID}_{50}$ corresponds to 1–100 virions, so the bounds for $h$ were $[1, 100]$. Hence, the initial ratio of total to infectious virions in the respiratory tract in units of RNA copy number/virion ($\gamma$), which by definition had a lower bound of 1, was $\gamma = f/h$. Based on the bounds for $f$ and $h$, the bounds for $\gamma$ were then $[10^0, 10^5]$.

Lastly, we tackle the parameter $p_{Vratio}$. Petrie *et al.* [4] used the bounds $[10^0, 10^6]$ to fit the ratio of production of total to infectious virions, in units of RNA copy number/$\text{TCID}_{50}$ ($q$). These bounds were based on the variability of this ratio in the data provided by Butler *et al.* [5]. The ratio of production of total to infectious virions in units of RNA copy number/virion ($p_{Vratio}$), which also by definition had a lower bound of 1, was then $p_{Vratio} = q/h$. Based on

the bounds for $h$ and $q$, the bounds for $p_{Vratio}$ were then $[10^0, 10^6]$.

# References


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