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What controls the acute viral infection following Yellow Fever vaccination?

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Abstract

Does target cell depletion, innate immunity, or adaptive immunity play the dominant role in controlling primary acute viral infections? Why do some individuals have higher peak virus titers than others? Answering these questions is a basic problem in immunology and can be particularly difficult in humans due to limited data, heterogeneity in responses in different individuals, and limited ability for experimental manipulation. We address these questions for infections following vaccination with the live attenuated Yellow Fever Virus (YFV-17D) by analyzing viral load data from 80 volunteers. Using a mixed effects modeling approach, we find that target cell depletion models do not fit the data as well as innate or adaptive immunity models. Examination of the fits of the innate and adaptive immunity models to the data allows us to select a minimal model that gives improved fits by widely used model selection criteria (AICc and BIC), and explains why it is hard to distinguish between the innate and adaptive immunity models. We then ask why some individuals have over 1000 fold higher virus titers than others, and find that most of the variation arises from differences in the initial/maximum growth rate of the virus in different individuals.

1 Introduction

Primary acute viral infections are characterized by an initial period of rapid growth followed by a decline and clearance of the virus [35,7,26]. What accounts for these basic features of infection and in particular what is responsible for the initial decline in the density of virus? Three hypotheses are widely considered – target cell depletion [5,7,44,16,9], innate immunity [42,34,11,29,18, 16] and adaptive immunity [40]. In the target cell depletion (sometimes called resource depletion) hypothesis, the virus infects and kills the cells that are

the target for viral replication faster than they can be replenished. The depletion of these target cells results in a decline in the density of virus. In the innate immunity hypotheses the virus induces the innate immune response which includes the production of cytokines such as type I interferons and/or the activation of phagocytes such as macrophages and neutrophils. These factors cause a decline in viral density by a combination of reduction in virus growth and increased virus clearance. In the adaptive immunity hypothesis, the virus stimulates the clonal expansion of antigen-specific B and T cells, which then control the infection via virus-specific antibodies and the killing of infected cells by CD8 T cells. Determining the role of target cell depletion, innate and adaptive immunity in the control of infections has practical implications for the use of drugs to control infection and the generation of resistance [35,19].

Discriminating between these hypotheses is challenging as mathematical models incorporating any of these three hypotheses can generate dynamics consistent with the basic features of an acute infection (i.e. rapid virus growth followed by decline). Model discrimination is particularly difficult for data from human infections for a number of reasons. It is difficult to obtain detailed time courses of infection and immunity. Ethical constraints preclude most experimental manipulations. Finally host genetic diversity and differences in environment introduce much larger heterogeneity in the dynamics of infection and immunity than is seen in inbred laboratory mice kept in controlled conditions [39,6,32,41,33,12,38,14].

In the case of influenza A infection, an early modeling study [5] in humans proposed that the loss of target cells played a dominant role in the control of virus. Later studies using more extensive data from horses resulted in reevaluation of this conclusion as models incorporating an interferon response were better fits to the data [42,34]. In the case of SIV and HIV infections, early studies proposed that target cell depletion plays a key role in the decline in virus during the acute phase of infection [36,46]. A subsequent study analyzed data on SIV infection of rhesus macaques following depletion of CD8 T cells, and suggested that these cells play a crucial role in the decline of virus during the acute phase of infection [40]. While it is an intracellular protozoa and not a virus, a similar debate exists for malaria infection. Although it is clear that adaptive immunity is required for the long-term control and clearance of infection, the roles of target cell depletion and innate immunity during the initial decline in parasitemia are less clear [28,21,4,23,30]. Finally, determining the role of target cell depletion and innate immunity in the control of dengue and zika is of current interest and one modeling study has suggested that an innate immunity plays an important role [8]. When the data comes from experimental infections of animals, manipulations that alter the numbers of target cells or deplete components of the innate and adaptive immune responses can facilitate discriminating between the different hypotheses [47,40, 20].

In this paper we study what controls viral infections of humans that arise following immunization with the yellow fever virus strain YFV-17D. YFV-17D is widely used as a safe and effective live attenuated virus vaccine. Following immunization the virus generates a relatively mild acute infection that lasts for about 1 to 2 weeks and ends with the clearance of the virus and the generation of long-lasting immunity [31]. We use data from 80 naive individuals who were seronegative for YFV prior to vaccination. Following immunization

with YFV-17D these individuals were intensively sampled and virus and CD8 T cell responses quantified. This gave us a fine-grained look at the dynamics of viral load following immunization [15,1].

An outline of the paper is as follows. We begin in Section 2 by describing the data on the dynamics of virus following immunization with YFV-17D. In Section 3 we construct mathematical models incorporating the three different hypotheses (target cell depletion, innate immunity, or adaptive immunity), and in Section 4 we consider which model best reproduces the virus dynamics measured in the data from the 80 vaccinated individuals.

2 Data

The data is described in detail in earlier papers [15,1]. Briefly, 80 healthy volunteers were injected with Yellow Fever vaccine virus (YFV-17D), a live attenuated virus. Viral titers were recorded on day 0, 1, 2, 3, 5, 7, 9, 11 and 14. The data showed that the peak virus load typically occurred at day 5 or 7 following immunization, and the virus was rapidly cleared in the subsequent 2 to 4 days. The CD8 T cell response to YFV was measured by following the number of activated Ki67⁺, Bcl2⁺ CD8 T cells in the blood at days 0, 3, 7, 11, 14, 28 and 90 following infection and this was supplemented by measurement of responses to the HLA-A2 restricted epitope in the NS4B protein of YFV in a subset of individuals possessing HLA-A2 MHC. The number of activated T cells were not significantly above background (day 0) at day 3 following infection and only just above background at the peak observed viremia. Consequently the CD8 T cell data was not of sufficient resolution to be useful for the timeframe of this analysis and we focused on the virus data alone.

3 Models of viral dynamics

We first present three within host models of viral dynamics that correspond to the three hypotheses for the control of infection. These are the target cell model, the innate immunity model and the adaptive immunity model. The basic outline of the models is shown in Fig. 2. We consider alternative versions of these models in S1 in Online Resource 1.

The **target cell depletion model**, or target cell model, assumes that the viral infection will continue to grow provided that there are sufficient uninfected target cells. If S and I are the number of susceptible and infected target cells, and V equals the number of virions, then

$$\begin{array}{l} \text{target cell model} \quad \begin{array}{l} \frac{dS}{dt} = -\beta SV \\ \frac{dI}{dt} = \beta SV - \delta I \\ \frac{dV}{dt} = pI - cV \\ S(0) = S_0 \quad I(0) = 0 \quad V(0) = V_0 \end{array} \quad (1) \end{array}$$

where β is the parameter describing infectivity, p is rate of production of virions by infected cells, δ is the death rate of infected cells, and c is the decay rate of virions. As the number of target cells and infected cells is unobserved, we can set $p \equiv 1$.

The **innate immunity model** assumes that the virus induces an innate immune response R that is responsible for controlling the infection. We let the virus grow exponentially at rate r in the absence of the immune response. We assume that there is a fixed population of innate immune cells. These start in a resting state but can become active in the presence of virus. Therefore the innate immune response has a maximum response level which we scale to unity [45,2]. R represents the proportion of the maximum innate response – and can be thought of as the fraction of innate immune cells such as macrophages or dendritic cells that are in an activated state and producing cytokines that control the infection. The activation of immune cells is incorporated as a saturating function of viral load. Similarly the control of viral infection is incorporated as a saturating function of cytokines [25]. However, we assume that the lifespan of cytokines is short, so their concentration is proportional to R . The innate immunity model is described by

$$\begin{aligned} \text{innate immunity model} \quad & \frac{dV}{dt} = rV - kV \frac{R^n}{R^n + \psi^n} \\ & \frac{dR}{dt} = s \frac{V^m}{V^m + \phi^m} (1 - R) - \delta R \\ & V(0) = V_0 \quad R(0) = 0 \end{aligned} \quad (2)$$

where s , ϕ , and m are the parameters for the activation of innate immunity by virus, δ is the rate of inactivation of innate immune cells in the absence of virus, and k , ψ , and n are the parameters for control of the virus by innate immunity. We note that R is the inducible component of the innate response and any constitutive (non-varying component) would be incorporated by a change in the initial/maximum growth rate of the virus r .

The **adaptive immunity model** is similar to the innate immune model except for the term for the growth of immunity. In contrast to innate immunity which involves the activation of a fixed population of cells, adaptive immune responses are generated by clonal expansion of virus-specific cells in a manner dependent on the density of the virus. Consequently, the adaptive immune response can potentially reach a higher magnitude than innate immunity [40, 25,26]. The adaptive immunity model is described by

$$\begin{aligned} \text{adaptive immunity model} \quad & \frac{dV}{dt} = rV - kV \frac{X^n}{X^n + \psi^n} \\ & \frac{dX}{dt} = s \frac{V^m}{V^m + \phi^m} X - \delta(X - X_0) \\ & V(0) = V_0 \quad X(0) = X_0 = 1 \end{aligned} \quad (3)$$

where s , ϕ , and m are the parameters for the activation of adaptive immunity by virus, δ is the rate of loss of adaptive immunity in the absence of virus, and k , ψ and n are the parameters for control of the virus by adaptive immunity. We have chosen $X(0) = 1$ which amounts to rescaling of the response to its initial value. Note that the term $\delta(X - X_0)$ represents the maintenance of X at a stable initial value in the absence of antigen stimulation. Differences in the initial value of the response in different individuals can be accounted for by changes in the value of ψ .

We note that the parameters s , ϕ , m , etc take different values for innate and adaptive responses. Further, as the rate of virus clearance is relatively rapid compared with the timescale for loss of both innate and adaptive immunity we set the parameter δ for the rate of decay of these responses to zero.

4 Model comparison

There are a number of approaches to fitting models to longitudinal data for the dynamics of virus infections. The simplest is to fit the model to the average data for the dynamics of infection. A more widely used approach is to fit the model to each individual independently and examine the parameters for the different individuals [5,42,34]. Ideally, the number of observations for each individual is large compared to the number of model parameters to prevent overfitting. A more sophisticated approach is mixed effects modeling [11,25, 9], in which the mean and standard deviation for each parameter within a population is estimated. As the data for each individual is relatively limited we focus on the mixed effects modeling framework and where relevant mention its advantages compared with the previous approaches.

To compare the fit of each of our models to data, we calculate the likelihood using a mixed effects framework, assuming independent log-normal priors for each model parameter. The mean and standard deviation of these priors is calculated by minimizing the likelihood function

$$\begin{aligned}
 L &= \text{Likelihood} \\
 Y_i &= \text{Data from volunteer } i \\
 \theta, \omega &= \text{Prior mean and standard deviation} \\
 \lambda &= \text{Parameter value} \\
 f &= \text{Dynamic Model} \\
 L &= \sum_i \int_{\lambda} p(Y_i | f, \lambda) q(\lambda | \theta, \omega) d\lambda
 \end{aligned} \tag{4}$$

where $p(Y_i | f, \lambda)$ is the probability that model f produces the data with parameter choice λ , and $q(\lambda | \theta, \omega)$ is the probability of λ according to the prior. As many of our measurements fall below the limit of detection, we use a left censoring approach [17] to calculate $p(Y_i | f, \lambda)$. If the measurement is below the limit of detection, $y_c = 10$ genomes per ml, then we evaluate the probability that the model would produce any subthreshold value.

$$\begin{aligned}
\sigma &= \text{Estimate of noise in model} \\
Y_{ij} &= \text{Data from volunteer } i \text{ at time } j \\
\hat{y} &= f_j(\lambda) = \text{Dynamic Model evaluated at time } j \\
p(Y_i | f, \lambda) &= \prod_j g(Y_{ij}, f_j(\lambda)) \\
g(Y, \hat{y}) &= \begin{cases} \sqrt{2\pi\sigma^2} \exp\left(-(Y-\hat{y})^2/(2\sigma^2)\right) & Y > y_c \\ \sqrt{2\pi\sigma^2} \int_{-\infty}^{y_c} \exp\left(-(Y-\hat{y})^2/(2\sigma^2)\right) dy & Y < Y_c \end{cases} \quad (5)
\end{aligned}$$

where the value of σ must also be estimated for each model by maximizing (4). For further details about the calculation of the likelihood see section S2 in Online Resource 1 as well as [27,37,17]. We use the software package Monolix to maximize this likelihood function [24,43].

We examine the fit of a model to the data by minimizing $-2 \log L$ and smaller numbers indicate better model fits. In order to statistically compare the fit of models with different numbers of parameters we used two commonly used measures the corrected Akaike's Information Criterion (AICc) and the Bayesian Information Criterion (BIC) which are information-based criteria that assess model fit. Both AICc and BIC are based on $-2 \log L$. Both add a penalty based on number of parameters and have corrections for finite sample sizes. $AICc = -2 \log L + 2NK/(N - K - 1)$ and $BIC = -2 \log L + K \log N$ where N is the number of data points and K is the number of parameters. A difference in BIC between two models of over 10 is considered to be evidence of strong support in favor of the model with the lower BIC.

5 Results

5.1 All models can fit the average dynamics of infection and data from individual volunteers

We begin by examining if the target cell model, innate immunity model and adaptive immunity model could fit the average dynamics of virus during the infection. In Figure 3 we see that all three models can fit the data on the average dynamics of infection, and indeed they produce almost indistinguishable fits. The models give virtually identical log likelihoods, and thus AICc and BIC both favor the target cell model as it has the fewest number of parameters (see Table S1 in Online Resource 1). We also find that all three models are able to fit to the data for each individual separately. Figure 4 shows the results for a few representative individuals. In some individuals such as Panel A of Figure 4 the target cell model can give a very different trajectory for the dynamics of infection compared with the trajectories generated by the innate and adaptive immunity models. The problem is that there are only a few datapoints for the dynamics of infection in a given individual and the models compared with the number of parameters for each of the models, and consequently we run into the problem of overfitting. These results are not unexpected, but we feel they illustrate the limitations of these approaches and motivate the mixed effects modeling framework.

5.2 A mixed effects modeling approach rejects the target cell model

We use a mixed effects modeling approach to help discriminate between models that we were not able to distinguish between by fitting to the mean data. The mixed effects framework must reproduce both the dynamics of infection and the pattern of variation between individuals.

The results of fitting the three different models to the viral load data using a mixed effects approach is shown in Table 1. The log-likelihood indicates that the innate and adaptive immunity models fit significantly better than the target cell model. This is supported by the AICc and BIC values which take the different numbers of parameters into account – the difference in BIC between the target cell and the other two models is about 40 which indicates very strong statistical support for rejecting the target cell model. The difference in BIC between the innate and adaptive models is small indicating that it is not possible to discriminate between these two models.

Representative fits are shown in Figure 5, and the fits to all 80 individuals are shown in SI Figure 1. Visual inspection reveals that while all three models are able to capture the basic features of the expansion of virus followed by its control, the innate and adaptive immunity models appear able to capture the dynamics better than the target cell model in a few individuals. In particular comparison of the fits to each individual separately with the fits using the mixed effects framework (i.e. comparing Figures 4 & 5) indicate that the fits of the target cell model become significantly worse when the mixed effects framework constrains the variation in parameters between different individuals.

The mixed effects approach rejects the target cell model because it cannot reproduce the variance in peak viral load. Changing the number of target cells in this model can change the peak viral load. However, such a change would also lead to much more rapid transmission of the pathogen, which is not observed. Therefore to match the data for each individual, the target cell model must simultaneously change S_0 and β in almost exact proportion with each other. This tight correlation of parameters is heavily penalized in a mixed effects framework where β and S_0 are assumed to vary independently. There is no biological reason for S_0 and β to be correlated.

5.3 Parameters and a minimal model

In Table 2 we show the parameters for the innate and adaptive immunity models that give us the best overall fit and the standard deviations of these parameters between different individuals. Both models give comparable rates of viral growth (r) and the level of virus required for triggering of the responses (ϕ). The growth rate r corresponds to a doubling times for the virus of around $\ln(2)/r = 9$ hours, and innate and adaptive immunity are triggered at half maximal rates at relatively low viral loads ϕ of around 25 genomes/ml. The hill coefficients for the terms for stimulation of immunity and killing of virus are significantly greater than one.

When fitting the data, we find that n and m are generally large enough to warrant replacing the hill functions in (2) and (3) with step functions, with the possible exception of m in the

adaptive model. We therefore hypothesized that we might further improve the model by replacing the hill functions with step function. This results in two discontinuous ODEs

$$\begin{aligned}\frac{dV}{dt} &= rV - kH(R - \psi)V \\ \frac{dR}{dt} &= SH(V - \phi)(1 - R) - \delta R \\ V(0) &= V_0 \quad R(0) = 0\end{aligned}\quad (6)$$

and

$$\begin{aligned}\frac{dV}{dt} &= rV - kH(X - \psi)V \\ \frac{dX}{dt} &= SH(V - \phi)X - \delta(X - X_0) \\ V(0) &= V_0 \quad X(0) = X_0 = 1\end{aligned}\quad (7)$$

where H is the heaviside step function. Both (6) and (7) reduce to the following form

$$\begin{aligned}\text{minimal immunity} \quad V &= \begin{cases} V_0 e^{rt} & t \leq t_{\max} \\ V_{\max} e^{(r-k)(t-t_{\max})} & t > t_{\max} \end{cases} \\ V_{\max} &= \phi e^{rt_d} \\ t_{\max} &= (1/r) \ln(\phi/V_0) + t_d\end{aligned}\quad (8)$$

In the above, t_d is the time it takes for the immune response to reach the threshold ψ . Its value is given by

$$t_d = \begin{cases} -\frac{1}{s+\delta} \log(1 - \psi(s+\delta)/s) & \text{Innate Model} \\ \frac{1}{s-\delta} \log(\delta/s + \psi(s-\delta)/s) & \text{Adaptive Model} \end{cases} \quad (9)$$

The virus starts at titer V_0 (at time 0) and grows exponentially at rate r followed by exponential decay at rate $(r - k)$ when the induced innate or adaptive response reaches a critical threshold. Immunity would be triggered when the virus titer reaches ϕ and reach the threshold for virus control after a delay t_d . This model has three fewer parameters than the prior innate and adaptive immunity models described by (2) & (3): t_d is a composite of the parameters that includes s and ψ in the innate and adaptive immunity models. Therefore, the values in Table 2 for the innate and adaptive models should only be viewed as a parameter set consistent with the data. Thus we cannot say how rapidly the innate response saturates or how many divisions the cells of the adaptive response undergo.

A comparison of fitting the minimal inducible immunity model with the previous three different models to the viral load data using a mixed effects approach is shown in Table 1. The log-likelihood indicates that the minimal inducible immunity model fits the data almost as well as the innate and adaptive immunity models and significantly better than the target cell model. When we take the number of parameters into account, the AICc and BIC values tell us that the minimal inducible immunity model is statistically the best model having an AICc roughly 5 less and a BIC roughly 19 less than the more complex innate and adaptive immunity models.

To isolate which parameters are important for generating this variability, we vary one parameter at a time and then calculate the peak viral load, integral viral load, time of peak, and time of clearance for each individual. We set the test parameter equal to the posterior mode for each volunteer, and hold the others at the population means given in Table 2. Both the innate model and the adaptive models primarily generate the variance in viral load via the parameter r which represents the initial viral growth rate (Table 3), although there is a small contribution of the immune killing rate k to the clearance time. The variation in r could reflect between host differences such as the infectivity, density, and life span of susceptible cells and the rate of production and clearance of virus. All of these factors could be due the standing level of innate immunity; our model only predicts that the inducible component does not contribute to the variation in load observed between different individuals. This is primarily because we find very little variation between individuals in inducible immune response.

6 Robustness

We have demonstrated that three immune response models are a better fit to the data than a target cell depletion model. It is important to examine the extent to which this result is robust to the details of the models, such as whether there is a delay between infection of a target cell and the production of virus as in [5] or the nature of the function that describes activation of innate or adaptive immunity. We explored the following different models for each of the hypotheses (equations in Online Resource 1).

For the target cell models we explored the effect of: adding an eclipse phase [5]; simplifying the model by making a quasi-steady-state assumption that the decay rate of virus is fast [35]; and adding these together. For the innate immunity model we explored the effect of: using a simplified model for innate immunity with $n = m = 1$ [2]; using the model for cytokine production suggested by Pawelek et al [34]; using a simplified version of the Pawelek model [34]; and using a model suggested by Saenz et al [42]. For adaptive immunity we use: a simplified model for the dynamics of adaptive immunity with $n = m = 1$ [3]; and a simplified model for adaptive immunity with saturation in killing [25]. The quality of fits for all models is listed in Table 4.

None of the other models considered could fit the data as well as the three immune models (2), (3), or (8). Models incorporating an immune response are better fits by AICc and BIC than those that only include depletion of target cells. The only exception to this is the simplified adaptive immunity model with mass action killing, however this is fixed by

incorporating a saturation term to the killing rate. Therefore we conclude that the primary mechanism controlling infection in these individuals is an immune response and not depletion of target cells.

7 Discussion

We have used a combination of mathematical modeling and statistical analysis of viral dynamics data to explore which factor is primarily responsible for controlling the yellow fever virus YFV-17D following immunization of humans. Our results reject target cell depletion, and suggest that some form of inducible immunity is likely to be the principal factor in the control of YFV-17D. Our results suggest that the current data is consistent with either innate or adaptive immune responses being the principle factor for the control of the virus, and we were not able to discriminate between these hypotheses. A previous modeling study by Le et al. [25] suggested that the adaptive immune response (YFV-specific CD8 cells) controlled YFV-17D infections. The Le et al study showed that the dynamics of virus was consistent with an adaptive CD8 T cell response but did not consider alternative hypotheses. Our results suggest that it is not possible to discriminate between control by adaptive or innate immunity and highlight the importance of considering multiple hypotheses [22,13].

As described in the introduction, the identification of the principal factor responsible for control of acute infections has been the subject of considerable interest and many modeling studies. A number of factors make this problem potentially difficult, as we briefly discuss below. First, using models to discriminate between different hypotheses requires sufficient data, which can be problematic for human infections. We were fortunate to use a study with extensive sampling of virus at days 0, 1, 2, 3, 5, 7, 9, 11, 14 in the blood. Second, even with this unprecedented level of sampling there were usually only between 1 and 4 timepoints when the virus was above the threshold of detection. Fitting separately to each individual as in [34,42,5] was impractical for this data set. In this situation a mixed effects modeling approach can help as fewer parameters need to be estimated from the data. Third, each of the hypotheses can be described by a number of alternative models. Our approach to this problem was to examine multiple models that would describe each hypothesis. We examined 4 different target cell models, 5 innate immunity models and 3 different adaptive immunity models. We now describe some of the key assumptions and corresponding caveats of our study. The data that we model comprises of measurements of the virus titer in the blood of individuals, rather than from tissue samples. This is a common limitation of human studies. Second, we focus on the virus data. We lack data on cytokines and other measures of activation of the innate immune response, and the data on CD8 T cell and antibody responses are barely above background at the time of peak viremia (see section 2 on Data). Third we use simple deterministic models which ignore stochastic effects.

In our data, peak viral load could vary over 10^4 fold between individuals. Therefore, we also explored which factors might be responsible for this heterogeneity. We found that in our model a single parameter, the initial viral growth rate of the virus (r in (2) & (3)), explains most of this variation, rather than the parameters that describe how immunity is elicited or how fast it grows. This is supported by the observation that the peak virus density happens in

a relatively narrow time-range (typically at day 5 or 7 of the measurements). A number of biological factors could result in changes in the initial growth rate of the virus including differences in the susceptibility of cells to infection, the burst size of the virus, as well as constitutive levels of innate immune responses in different individuals. Further studies will need to be done to answer this question.

Interestingly, it is the heterogeneity in viral dynamics between individuals that allows us to distinguish between models. A mixed effects approach requires the models to reproduce both the dynamics of infection in individuals and the heterogeneity within the population. If this heterogeneity had been smaller in our data set, model discrimination would have been much harder as all models could reproduce the mean data. For example, a recent study on Zika virus infection in Rhesus Macaques [9] found that a target cell limited model could reproduce their data. However, their viral load data showed considerably lower between subject variability than we observed. For many years, immunological studies have focused on inbred animal models, in relatively sterile environments as these conditions minimize variability in the outcome of experiments [39,10]. The present study suggests that, at least in some situations, data obtained from ‘dirty’ mice, which may be exposed to a variety of pathogens, and outbred mice with variable genetics, may prove useful to quantitative modelers.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Fig. 1. Summary of Yellow Fever Vaccine data

80 volunteers were vaccinated with yellow fever vaccine and their plasma viral load was tracked for two weeks [15,1]. Each solid line represents a different volunteer, dashed line is the limit of detection. Time points with undetectable virus are imputed at 5 genome per ml (half the limit of detection) to aid visualization.

Fig. 2. Models for the three hypotheses

In Panel A we show the target cell model. In this model virus infection converts uninfected target cells to an infected state. Infected cells produce virus and have an increased death rate. In Panel B we show the innate immunity model. The virus grows exponentially and induces the activation of resting immune cells, which become activated. Activated innate immune cells are responsible for the control of virus. In Panel C we show the adaptive immunity model. In this model the virus induces the clonal expansion (replication) of virus-specific cells. These cells are responsible for clearance of the virus.

Fig. 3. Fits to average virus data

We plot the raw data viral titer in grey, the mean at each timepoint as black dots, and the median as black crosses. All three models produce virtually identical fits to the average dynamics of infection.

Fig. 4. Individual fits to dynamics of virus in four subjects

We fit the three models (i.e. the target cell, innate and adaptive immunity models) to the data on the dynamics of infection in four different individuals. We see that all three models can fit the data well. However in some individuals (e.g. Panel A) the target cell model can give a very different trajectories for the dynamics of infection from the innate and adaptive immunity models.

Fig. 5. Fits from the mixed effects modeling approach

We show the fits obtained using the mixed effects modeling framework to the same four individuals shown in Figure 4. The circles are the data collected for each individual and the dashed line represents the limit of detection. We see that the innate and adaptive immunity models produce similar dynamics of infection and fit the data better than the target cell model. Fits to all 80 individuals are shown in the Online Resource 1.

Fig. 6. Minimal inducible-immunity model

A minimal model to explain the data for virus and inducible immunity. The virus exhibits exponential growth and decay and the immune response is induced at virus density ϕ and controls the virus with a time delay t_d . We show.

Table 1**Comparison of virus models**

Log likelihood, AICc, and BIC for mixed effects fits. N_P is the number of parameters.

	-2LogL	AICc	BIC	N_P
Target cell model	1301	1323	1349	15
Innate immunity model	1235	1270	1309	17
Adaptive immunity model	1234	1269	1308	17
Minimal immunity model	1241	1264	1289	11

Table 2
Parameter fits for innate/adaptive/minimal immune mixed effects models

These values represent the mean and standard deviation of the priors that maximize the likelihood as calculated by monolix. All parameters are assumed to be log-normally distributed, constraining all parameters to be positive. In some cases the standard deviation is greater than the mean, because the lognormal distribution can be highly skewed. (gen/ml) refers to viral genomes per ml of blood. Parameter distributions for all models are show in Online Resource 1 Figures 2–5

Parameter	Description	Innate		Adaptive		Minimal	
		Mean	S.D.	Mean	S.D.	Mean	S.D.
r (1/day)	Viral growth rate	1.69	0.48	1.83	0.46	1.60	0.47
k (1/day)	Max killing rate	4.18	0.46	4.11	0.56	3.68	0.66
s (1/day)	Immune growth/activation rate	0.17	0.01	0.52	0.02		
ϕ (gen/ml)	V req'd for 50% max activation	25.2	9.67	23.6	7.67	11.2	3.90
ψ	R/X req'd for 50% max killing	0.37	0.01	3.78	0.22		
V_0 (gen/ml)	Viral load at $t = 0$	0.10	0.08	0.10	0.08	0.10	0.04
n	Hill exponent in killing term	27.9	48.4	6.97	10.38		
m	Hill exponent in activation term	3.67	9.22	1.62	0.16		
t_d (days)	Time before immune response					2.90	0.26

Contribution of parameters in the minimal immunity model to variance in viral dynamics

Table 3

We first calculate the best fit parameter set for each individual. Then we set all but one parameter to the mean value and calculate the variance in peak viral load (log scale), integral viral load (log scale), time of peak and time of clearance. The first column shows the variance in these measures when all parameters are allowed to vary. We see that the parameter r is by far the most significant in producing the observed heterogeneity in viral load. The variance in the peak viral load in the data is roughly 4.5 logs, so the best model explain roughly 50% of the variance.

	Full	r	k	ϕ	V_0	t_d	s	ψ	n	m
Peak	Minimal	2.30	2.20	0.00	0.00	0.02	0.01			
Virus	Innate	2.51	1.55	0.00	0.02	0.06	0.01	0.00	0.04	0.00
	Adaptive	2.75	1.79	0.00	0.00	0.12	0.00	0.00	0.27	0.00
Area	Minimal	0.81	0.68	0.00	0.01	0.01	0.01			
Under	Innate	0.74	0.58	0.00	0.01	0.05	0.00	0.00	0.01	0.01
Curve	Adaptive	0.52	0.22	0.00	0.00	0.09	0.00	0.00	0.05	0.00
Peak	Minimal	1.20	0.47	0.18	0.02	0.01	0.04			
Time	Innate	1.43	0.18	0.02	0.02	0.05	0.01	0.00	0.05	0.02
	Adaptive	1.06	0.24	0.08	0.01	0.08	0.00	0.00	0.07	0.00
Clearance	Minimal	1.99	1.85	0.02	0.01	0.00	0.02			
Time	Innate	2.16	1.31	0.00	0.02	0.01	0.01	0.00	0.05	0.01
	Adaptive	2.29	1.44	0.01	0.00	0.03	0.00	0.00	0.20	0.00

Table 4
Comparison of goodness of fit to viral load data

We compare the loglikelihood, AICc and BIC of each model using both fits to the mean data and mixed effects fits. Each row represents a different model and the models are clustered by type.

Model	-2logl	AICc	BIC
Target cell models			
Target cell model (1)	1298	1320	1346
Target cell limited w/eclipse phase	1294	1321	1351
...with virus in steady state (QSS)	1312	1330	1352
...with QSS and eclipse phase	1314	1336	1362
Innate immunity models			
Innate immunity model (2)	1235	1270	1309
Simplified induction of immunity	1282	1304	1330
Pawelek model	1278	1312	1352
Simplified Pawelek	1291	1309	1330
Saenz model	1258	1315	1377
Adaptive immunity models			
Adaptive immunity model (3)	1234	1269	1308
Simplified adaptive immunity	1312	1335	1360
Simplified adaptive w/saturated killing	1269	1300	1335
Minimal immunity model (8)	1241	1264	1289