Ebola viral dynamics in nonhuman primates: insights into virus immuno-pathogenesis and antiviral strategies

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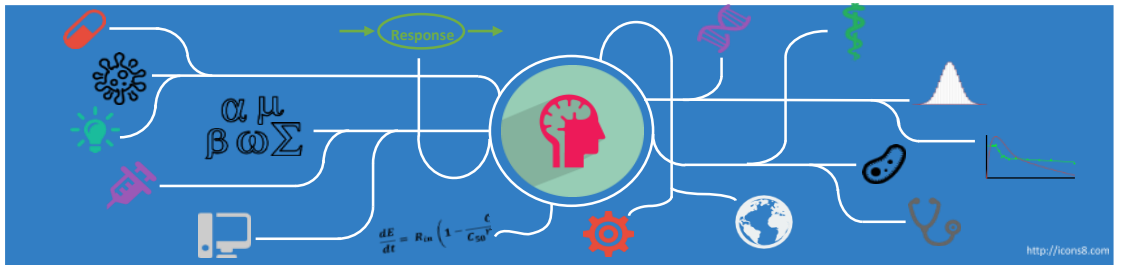
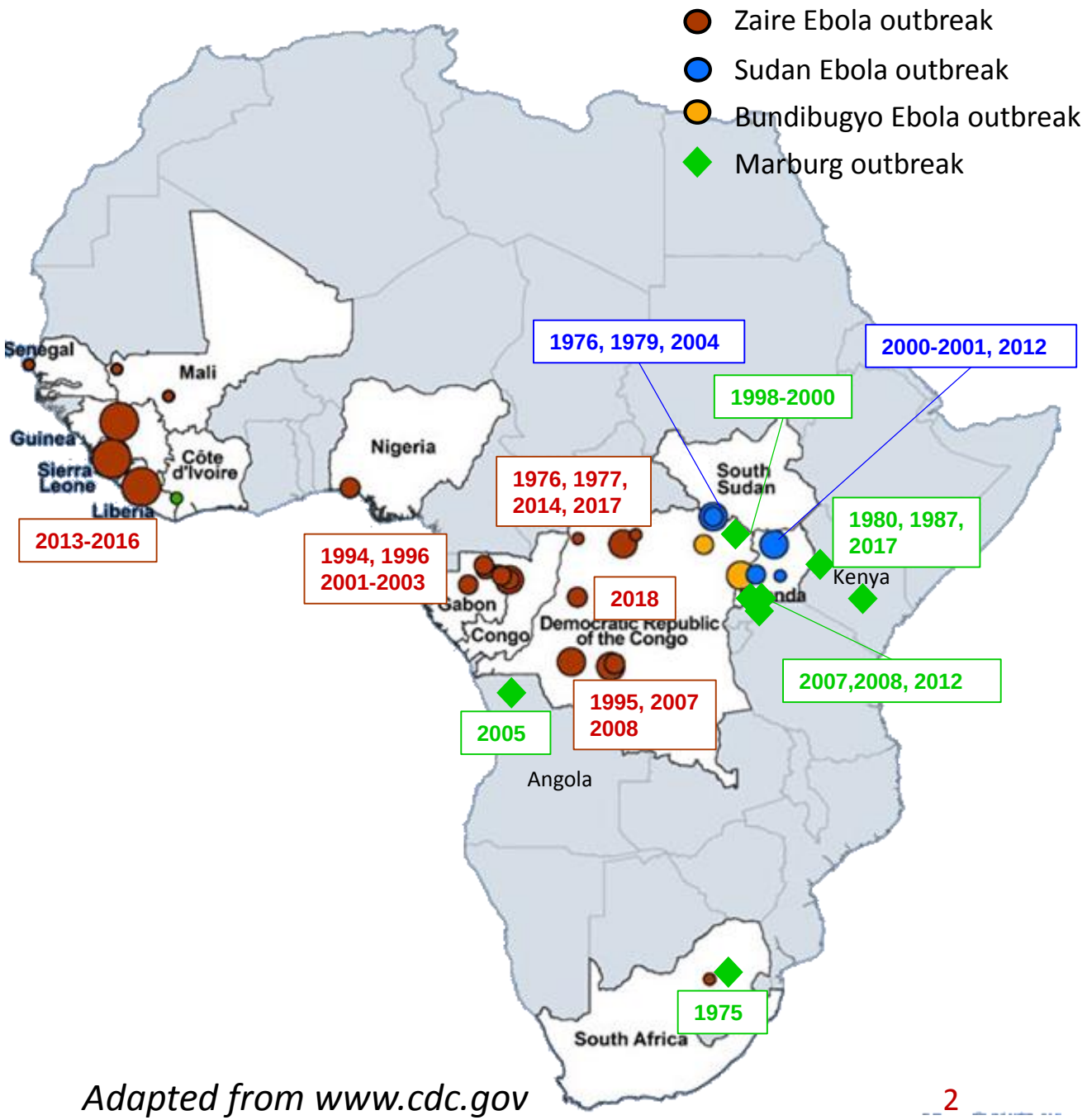
Page Meeting, June 1st 2018



This project has received funding from the *European Union's Horizon 2020 research and innovation programme* under grant agreement No° 666092

Background

- Hemorrhagic fevers represent a constant threat to public health in Africa and beyond
- The 2013-2016 Ebola outbreak in West Africa confirmed its potential to develop into regional epidemics
- None of the antiviral drug tested could demonstrate a significant effect on survival rate



Adapted from www.cdc.gov

Background

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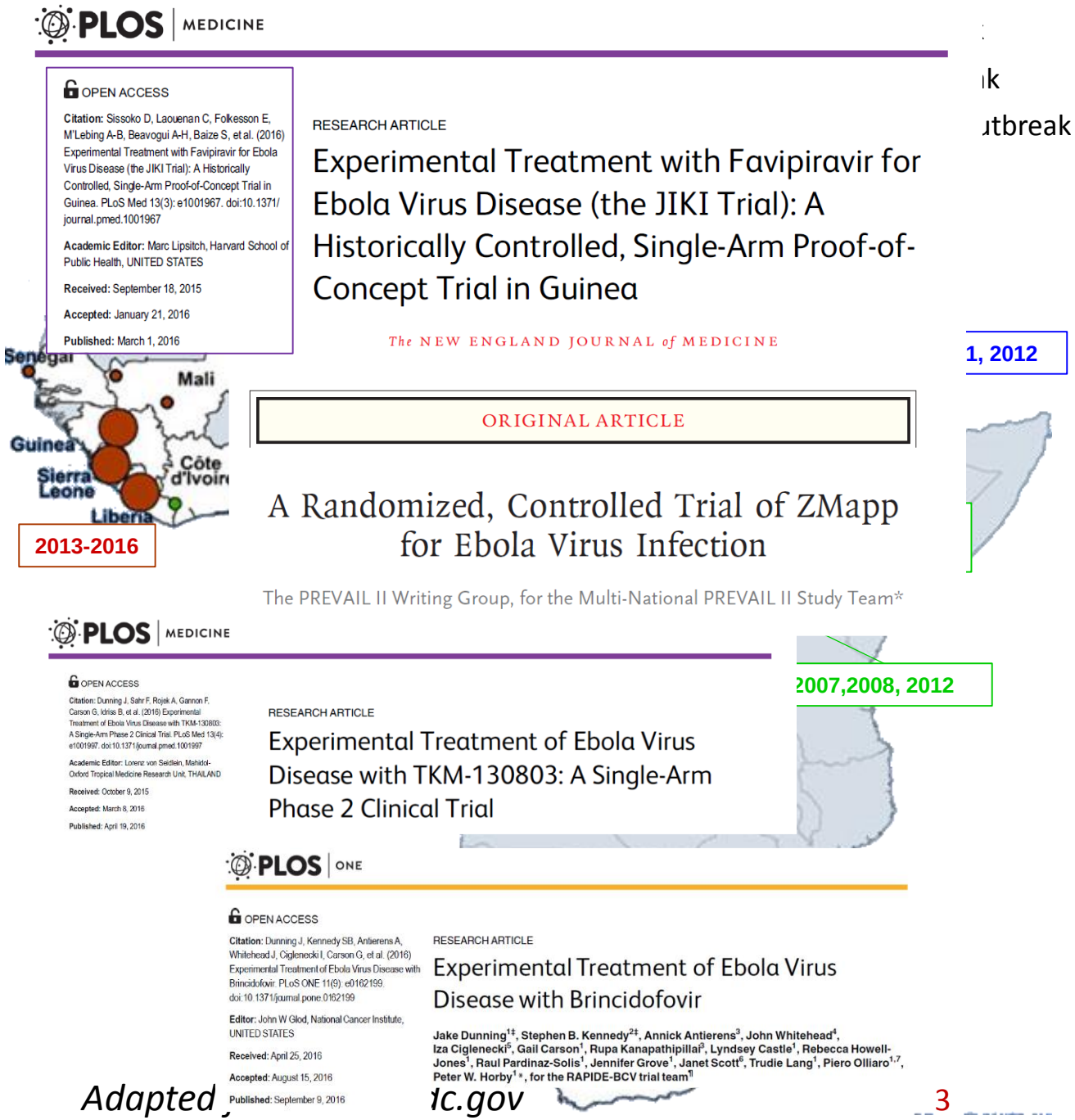
Sissoko et al, Plos Med, 2016
Prevail study group, NEJM, 2016
Dunning et al, Plos Med, 2016
Dunning et al, Plos One, 2016



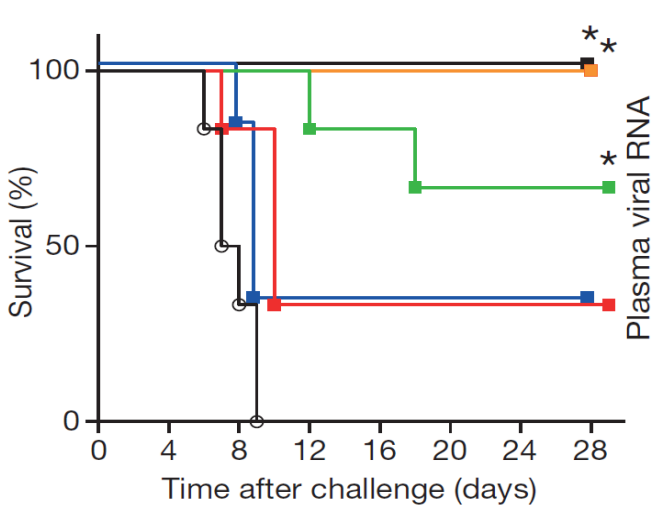
Background

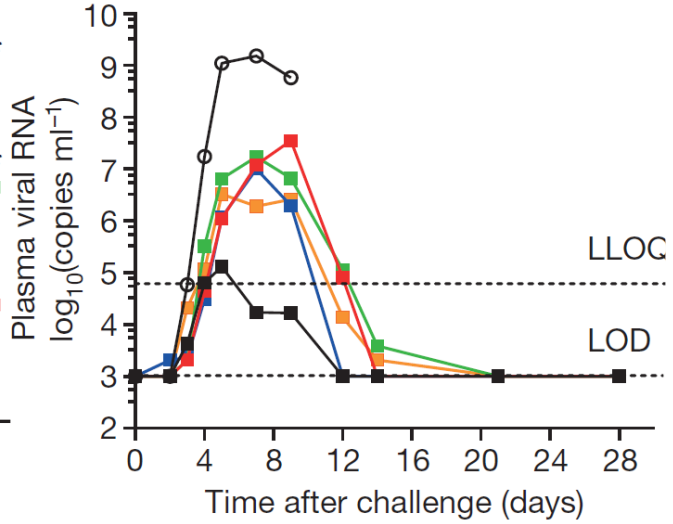
- Most of these candidate treatments, and others, demonstrated *in vivo* antiviral effect, and were found to significantly improve survival in NHP experiments

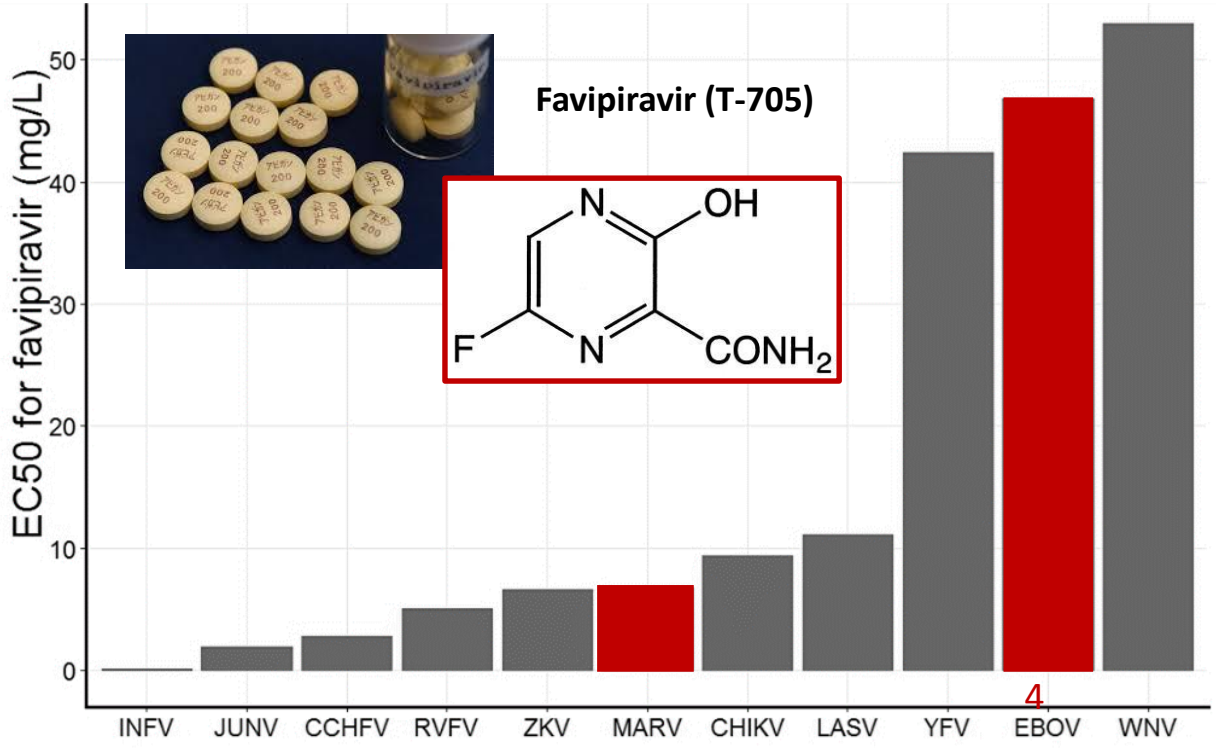
Warren et al, Nature, 2014
Qiu et al, Nature, 2014
Thi et al, Nature, 2015
Warren et al, Nature, 2016

- Our group focused on favipiravir, a RNA polymerase inhibitor approved for influenza, with activity against various etiological agents of hemorrhagic fevers

Oestereich et al, Antiviral res, 2014
Guedj et al, Plos med, 2018



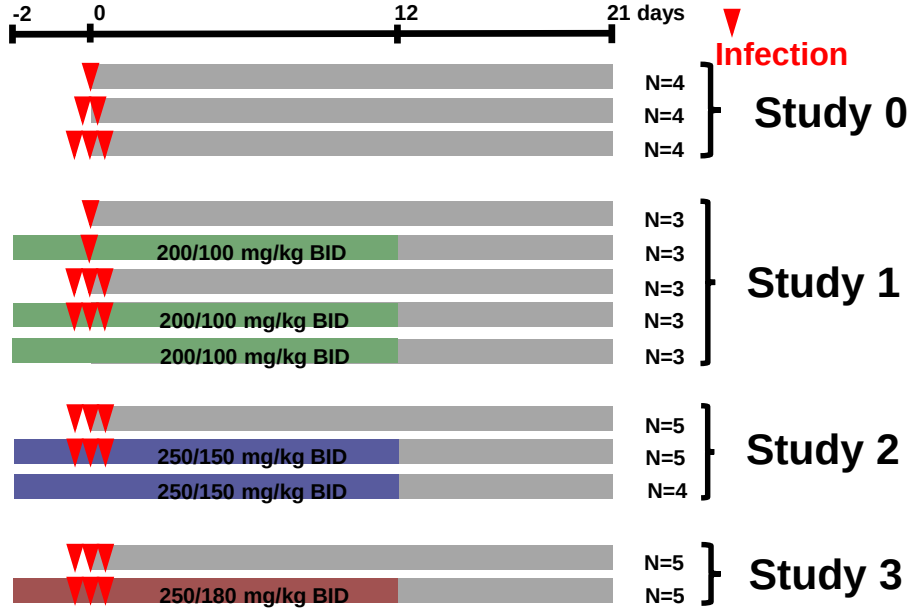




Objectives

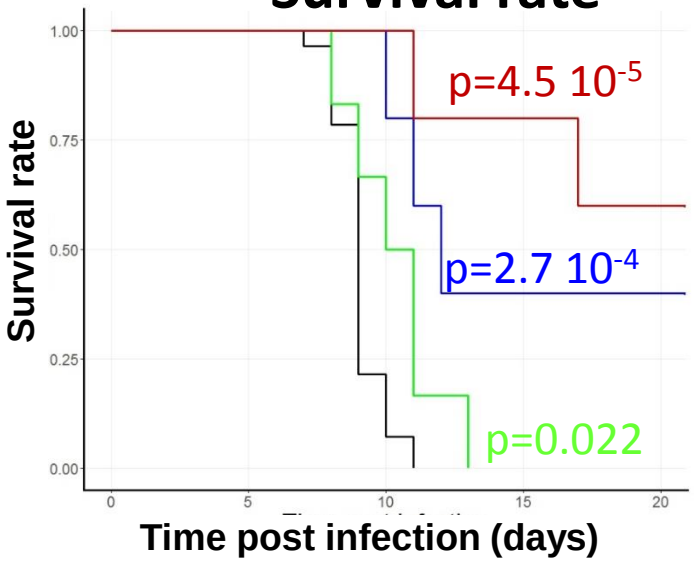
- To develop a mathematical mechanistic model to characterize the pathogenesis and the determinants of the death of Ebola virus disease (EVD) in NHPs
- To assess the activity of favipiravir in EVD
- To predict the impact of treatment potency and timing of initiation on survival rate

Study design and data

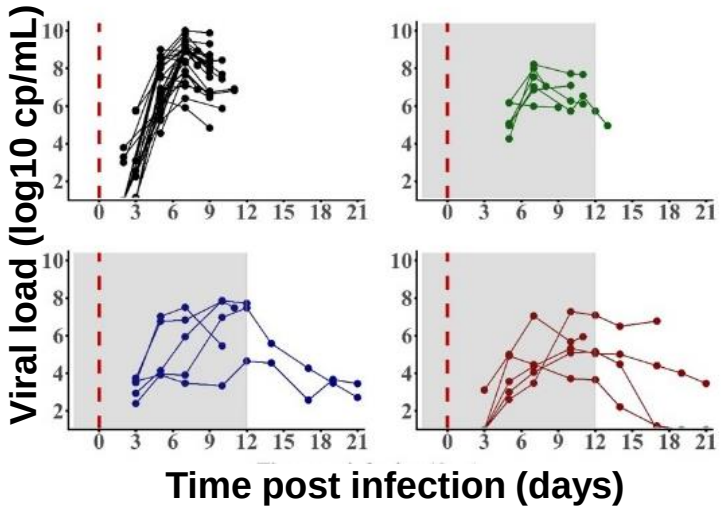


Guedj et al, Plos med, 2018

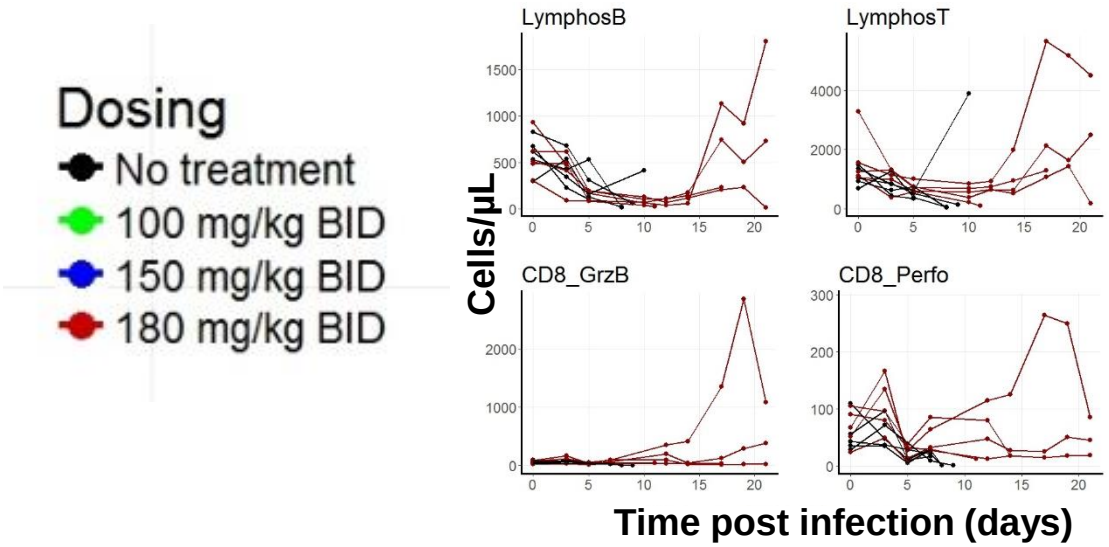
Survival rate



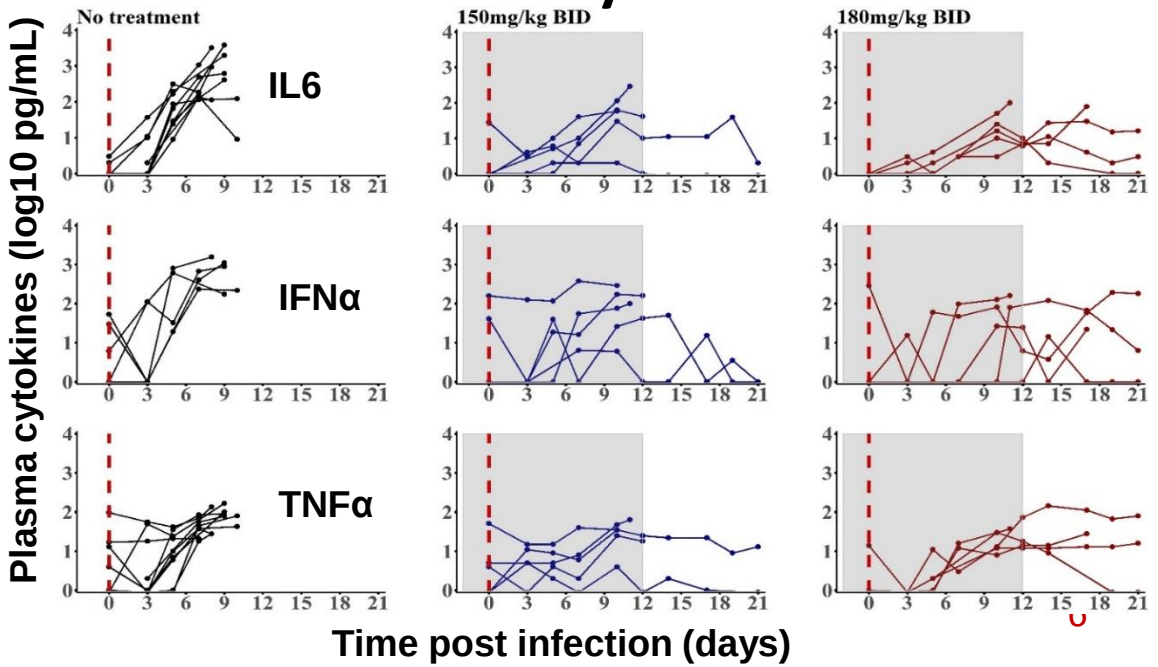
Viremia



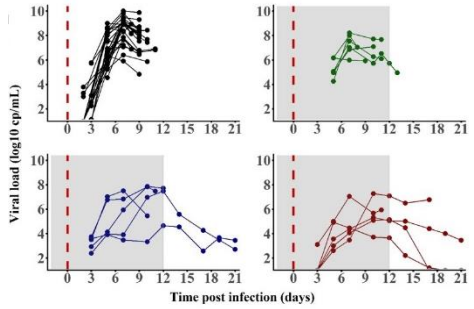
Cytometry



Cytokines

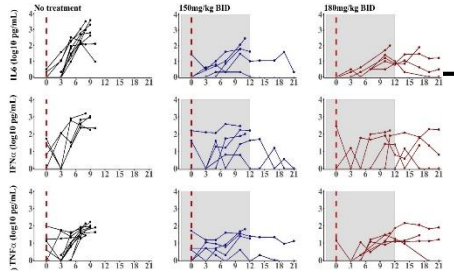


Model building strategy



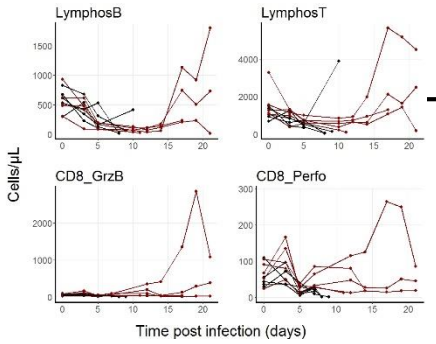
Viremia data

Target cell limited model



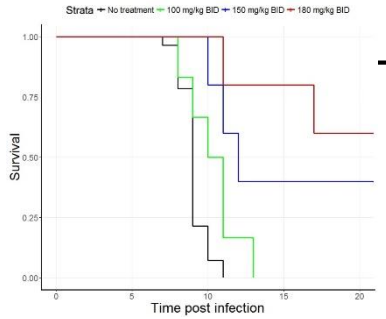
Cytokine data

Innate response model to describe the early stage of the infection



Cytometry data

Adaptive reponse model to describe the late stage of the infection



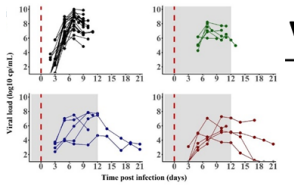
Time to death

Joint model to assess the impact of viral load and cytokine dynamics on survival

Improvement of viremia OFV

Parameter estimation was performed using the SAEM algorithm implemented in Monolix software

Target cell limited model



Viremia data

Target cell limited model

- Assumes that viral control results from target cell depletion
- Favipiravir concentrations are described by a previously developed PK model and inhibits viral production

$$\frac{dT}{dt} = -\beta TV$$

$$\frac{dI_1}{dt} = \beta TV - kI_1$$

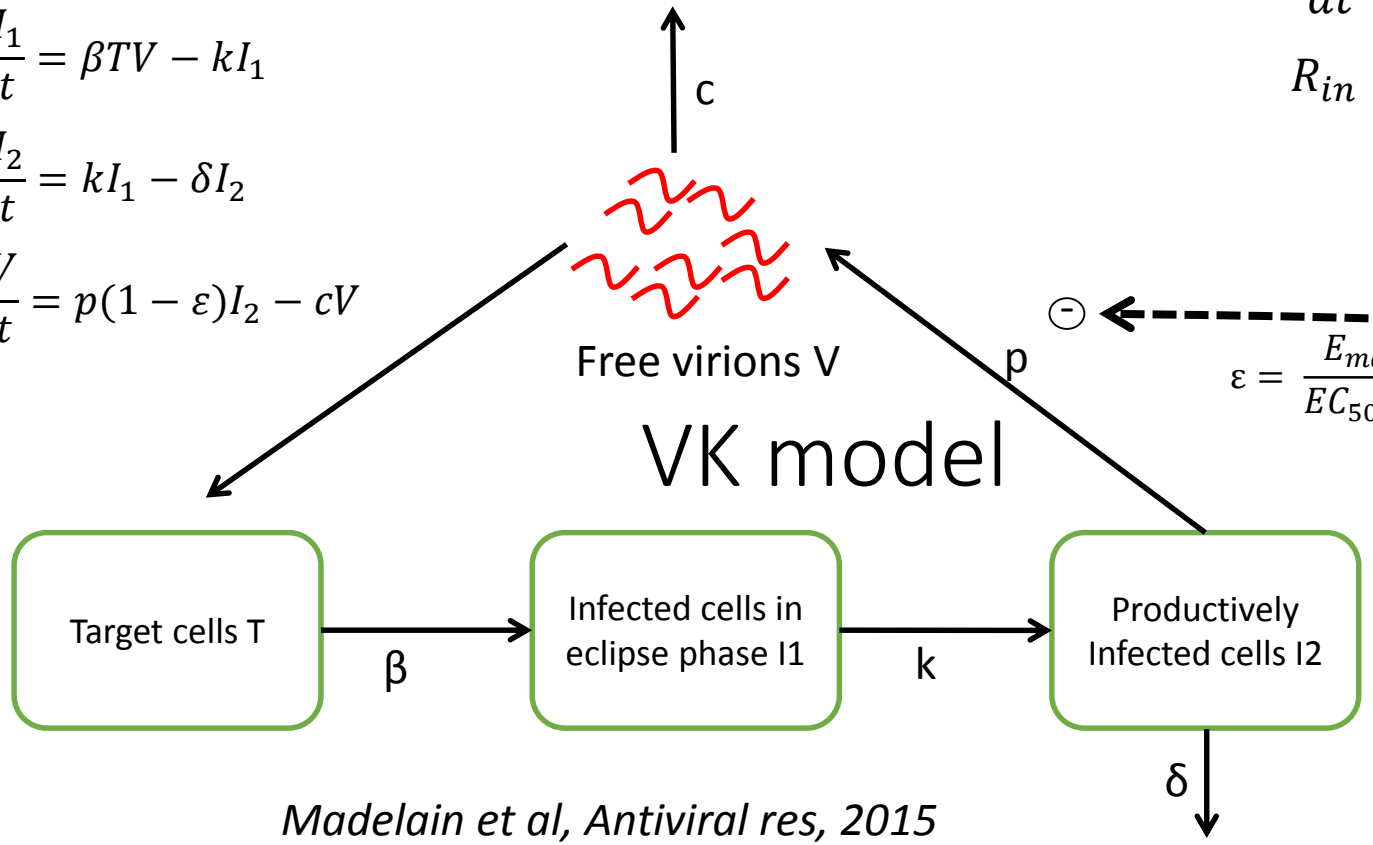
$$\frac{dI_2}{dt} = kI_1 - \delta I_2$$

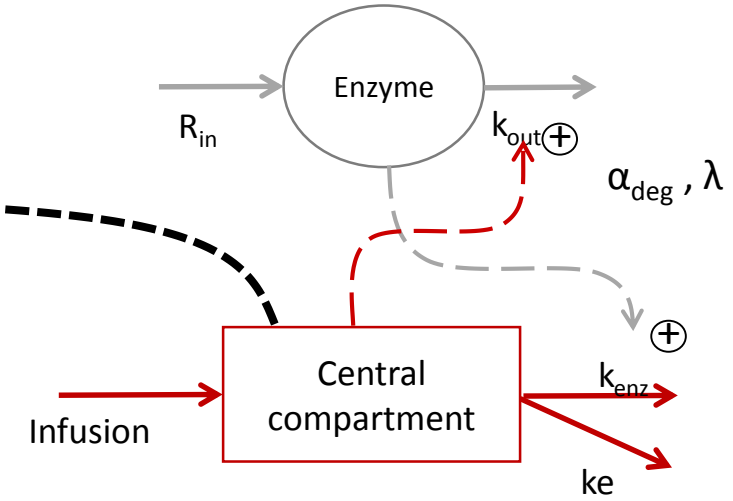
$$\frac{dV}{dt} = p(1-\varepsilon)I_2 - cV$$

$$\frac{dA_c}{dt} = -k \times A_c - V_{max} \times A_e \times A_c$$

$$\frac{dA_e}{dt} = R_{in} - k_{out} \times (1 + C_c \times k_{el} \times e^{-\lambda \times t}) \times A_e$$

$$R_{in} = k_{out} \times A_{e0} \quad ; C_c = \frac{A_c}{V}$$

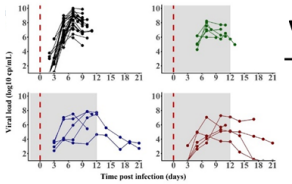




Madelain et al, Antiviral res, 2015

Madelain et al, AAC, 2017

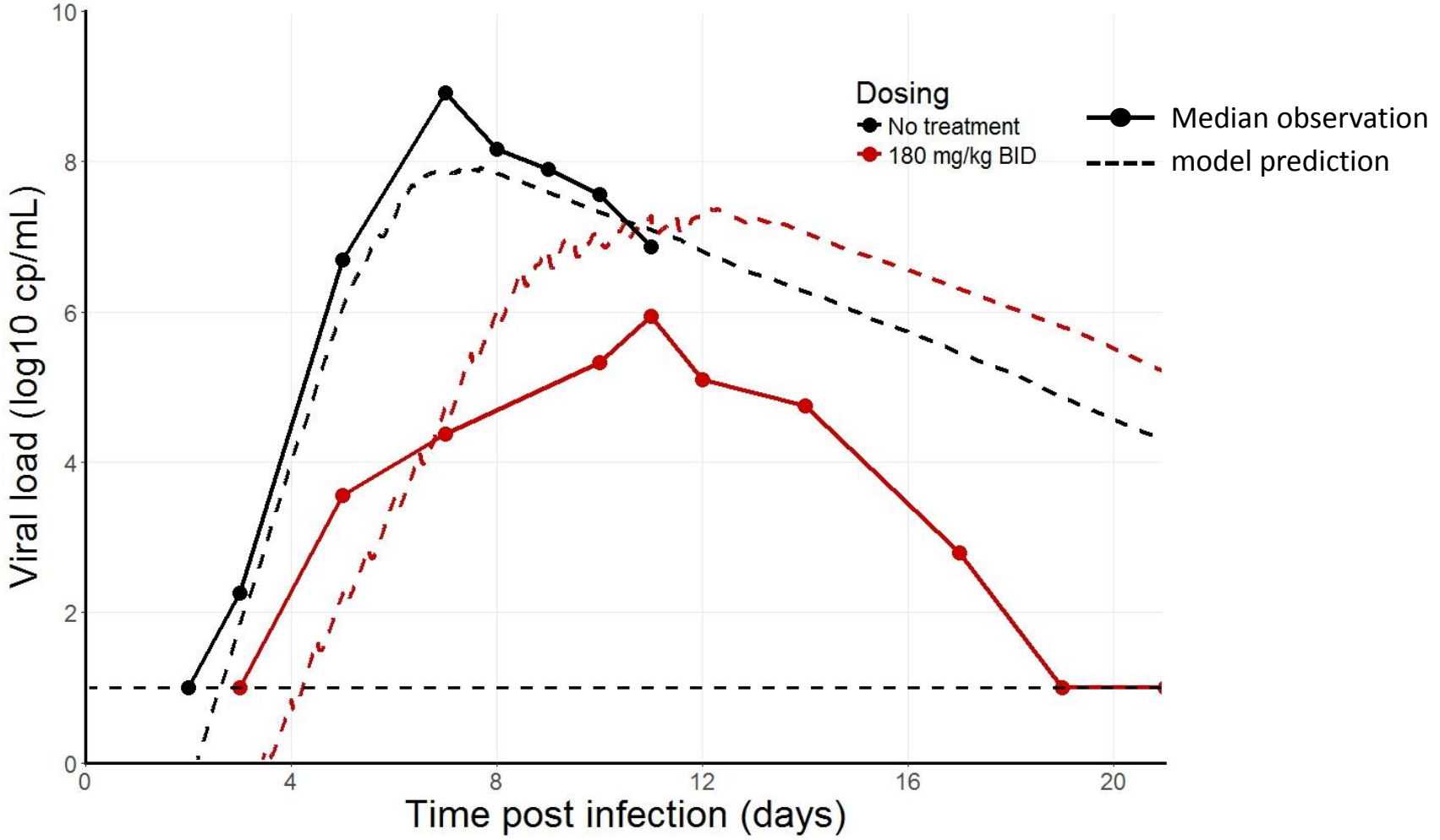
Target cell limited model



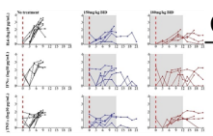
Viremia data

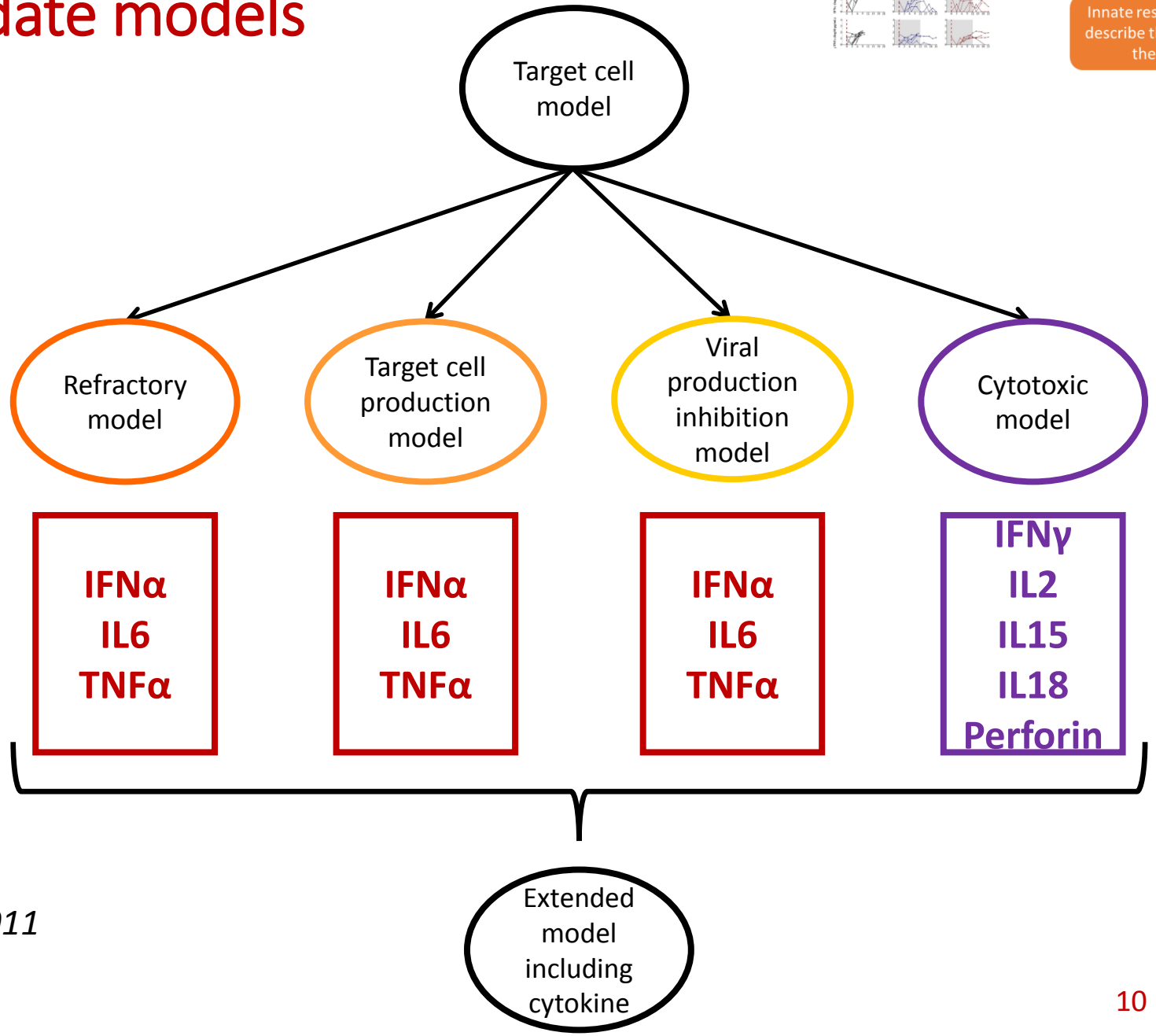
Target cell limited model

- Simulations showed that the model underestimates the effect of treatment on peak viremia



Evaluation of the candidate models



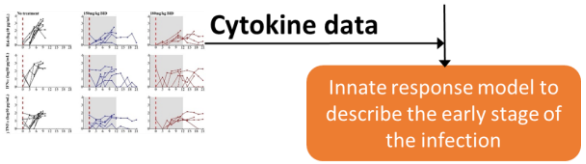


Pawelek et al, Plos Comp Bio, 2012

Smith and Perelson, WIREs SBM, 2011

Li and Handel, JTB, 2014

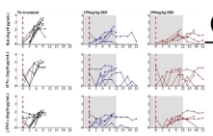
Selection of the candidate models



cytokine	model	effect of cytokine	-2logL viremia	Additional parameters
None (step 1)	target cell limited	none	645.7	_
IL6	**refractory**	**increase refractory cell production**	**618.0**	**4**
IL6	target cell increase	increase of target cells	644.8	4
IL6	prod inhibition	non linear inhibition of viral production	645.7	4
IFNα	**refractory**	**increase refractory cell production**	**622.4**	**4**
IFNα	target cell increase	increase of target cells	641.8	4
IFNα	prod inhibition	non linear inhibition of viral production	692.8	4
TNFα	**refractory**	**increase refractory cell production**	**621.8**	**4**
TNFα	target cell increase	increase of target cells	633.1	4
TNFα	prod inhibition	non linear inhibition of viral production	633.3	4
IFNγ	cytotox	increase infected cell elimination	634.5	4
IL2	cytotox	increase infected cell elimination	636.3	5
perforin	cytotox	increase infected cell elimination	634.6	4
IL15	cytotox	increase infected cell elimination	634.6	5
IL18	cytotox	increase infected cell elimination	634.8	4

→ Refractory models relying on IFNα, IL6 and TNFα and assuming a conversion of target cells into refractory cells provided the best description of viremia

Model including IFNα dynamics



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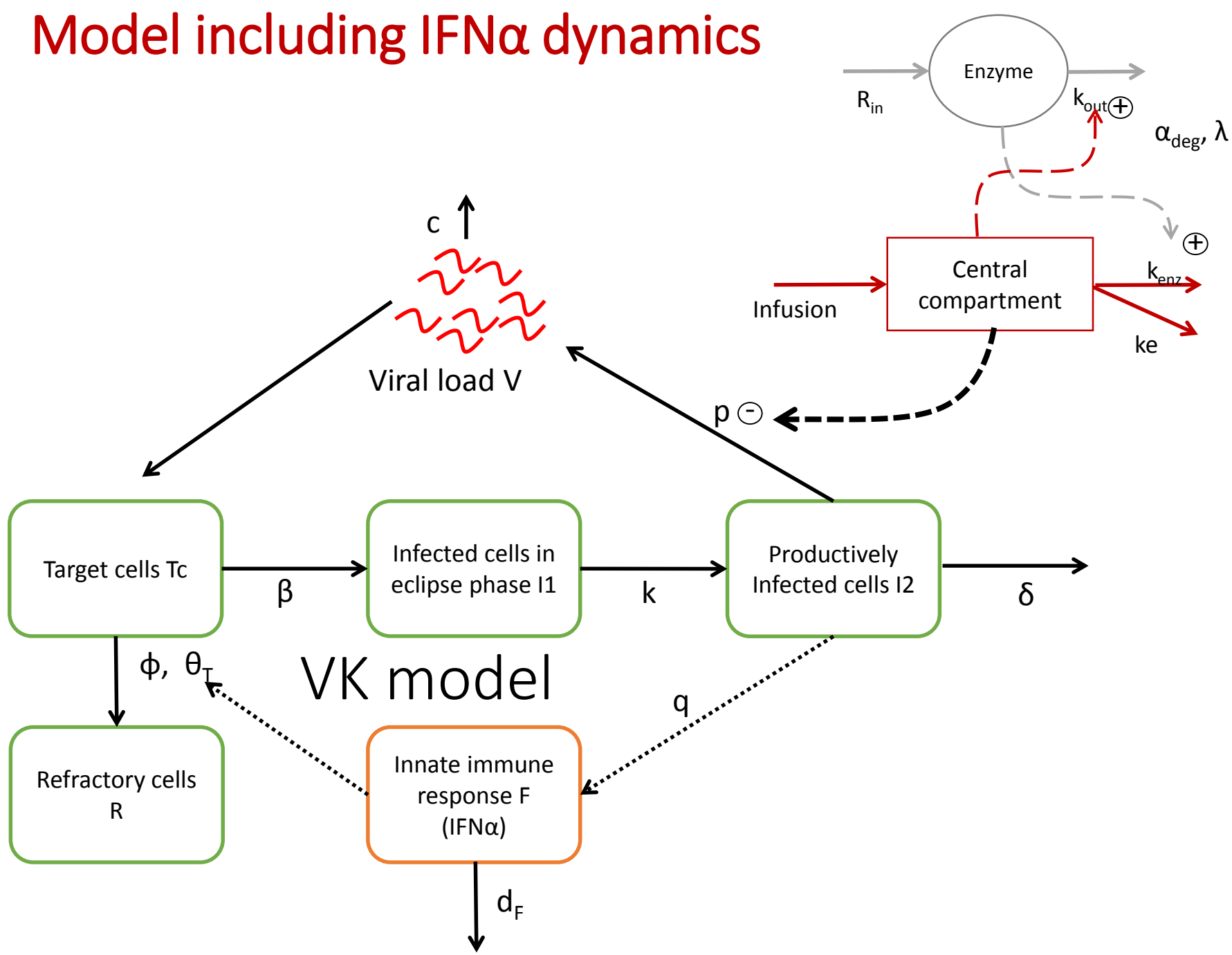


$$\frac{dT}{dt} = -\beta TV - \frac{\phi TF}{F + \theta_T}$$

$$\frac{dI_1}{dt} = \beta TV - kI_1$$

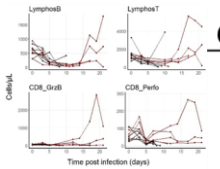
$$\frac{dI_2}{dt} = kI_1 - \delta I_2$$

$$\frac{dR}{dt} = \frac{\phi TF}{F + \theta_T}$$

$$\frac{dV}{dt} = p(1-\varepsilon)I_2 - cV$$

$$\frac{dF}{dt} = qI_2 - d_F F$$

Selection of the candidate models



Cytometry data

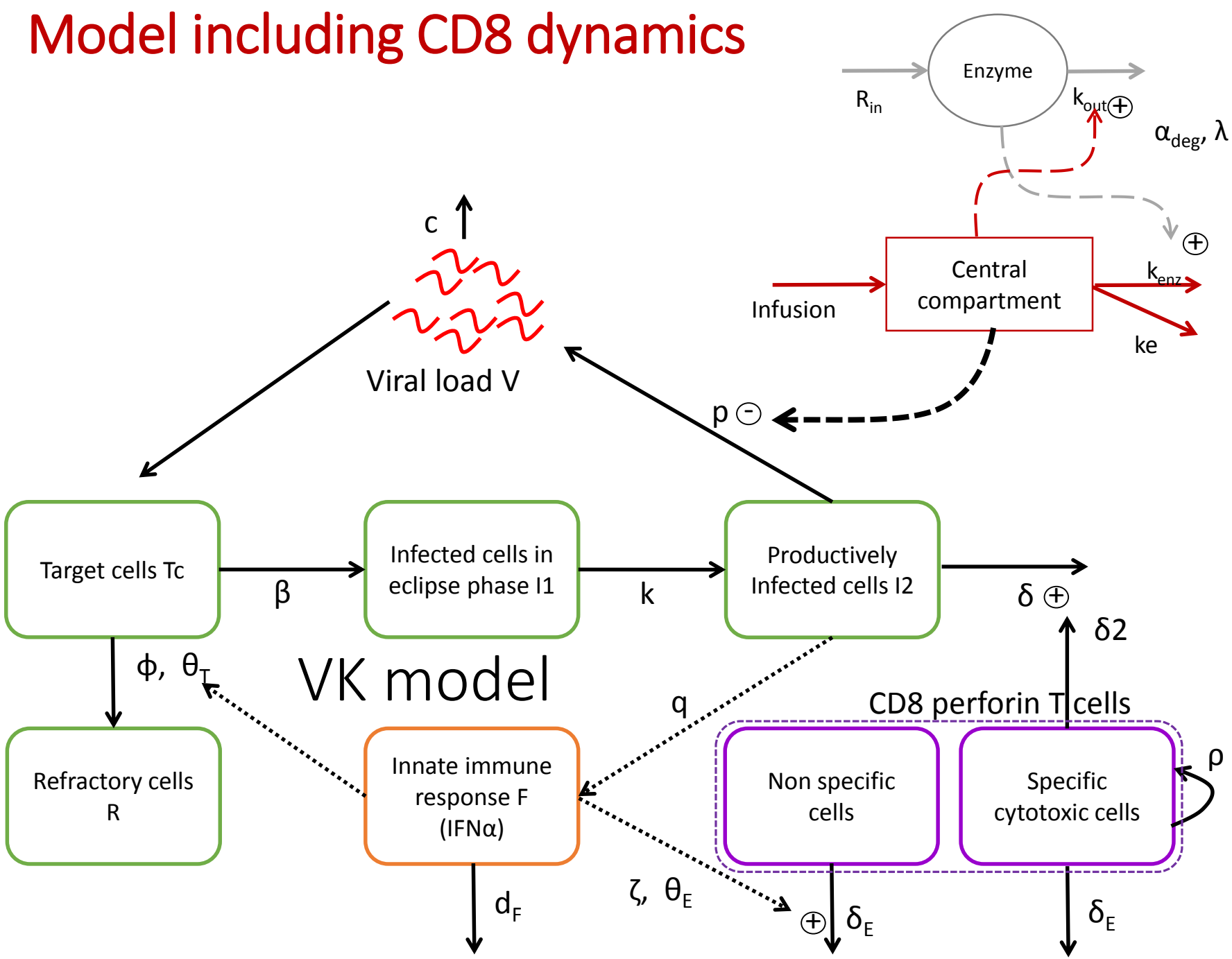
Adaptive reponse model to describe the late stage of the infection

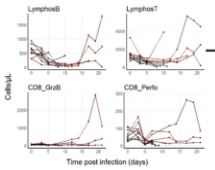
Adaptive response models including CD8 T cells expressing cytotoxic activity NKp80, perforin and granzymeB were compared

model	CD8 T cell data	-2logL viremia	Additional parameters
target cell limited (step 1)	_	645.7	_
Refractory (step 2)	_	618	4
refractory	**CD8 perforin+**	**605.5**	**12**
refractory	CD8 granzymeB+	611.6	12
refractory	CD8 NKp80+	608.4	12

→ CD8 T cell population expressing perforin was selected and included

Model including CD8 dynamics





Cytometry data

Adaptive reponse model to describe the late stage of the infection

$$\frac{dT}{dt} = -\beta TV - \frac{\phi TF}{F+\theta_T}$$

$$\frac{dI_1}{dt} = \beta TV - kI_1$$

$$\frac{dI_2}{dt} = kI_1 - \delta I_2 - \kappa I_2 E_2$$

$$\frac{dR}{dt} = \frac{\phi TF}{F+\theta_T}$$

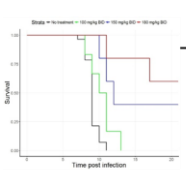
$$\frac{dV}{dt} = p(1-\varepsilon)I_2 - cV$$

$$\frac{dF}{dt} = qI_2 - d_F F$$

$$\frac{dE_1}{dt} = \sigma - \frac{\zeta F E_1}{F+\theta_E} - \delta_E E_1$$

$$\frac{dE_2}{dt} = \rho E_2\left(1-\frac{E_2}{E_0}\right) - \delta_E E_2$$

Impact of viral and cytokine dynamics on survival



Time to death

Joint model to assess the impact of viral load and cytokine dynamics on survival

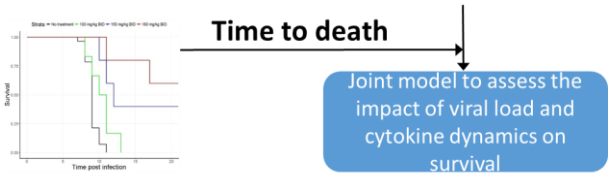
Extension to a joint model to assess the impact of viremia and cytokines on the hazard rate, where:

- h(t) is the hazard rate : $h(t) = \lambda_0 \times e^{\sum_k \beta_k \times \log(X_k(t))}$

  where $X_k(t)$ is current or lag-value of viral load, IL6, IFNα or TNFα

- S(t) is the probability to be alive up to time t: $S(t) = e^{-\int_0^t h(u)du}$

Final joint model



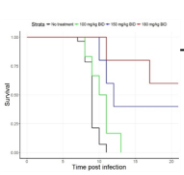
Model compartment driving the hazard of death	BIC
viremia	1545.0
IL6	**1506.5**
IFNα	**1506.5**
TNFα	1526.8

Best fit to the survival data was obtained with a model including IFNα or IL6

Enzyme
R_{in}
k_{out} ⊕
α_{deg} , λ
k_{enz} ⊕
Central compartment
Infusion
ke
⊖
c
Viral load VI
p
Target cells Tc
β
Infected cells in eclipse phase I1
k
Productively Infected cells I2
δ ⊕
δ2
ϕ, θ_T
q
CD8+ perforin+ T cells
Refractory cells R
Innate immune response F (IFNα)
Non specific cells
Specific cytotoxic cells
ρ
ζ, θ_E
d_F
⊕ δ_E
δ_E
Hazard of death

$$h(t) = \lambda_s \times e^{\beta_{il} \times \log(IFN\alpha_{eff})}$$

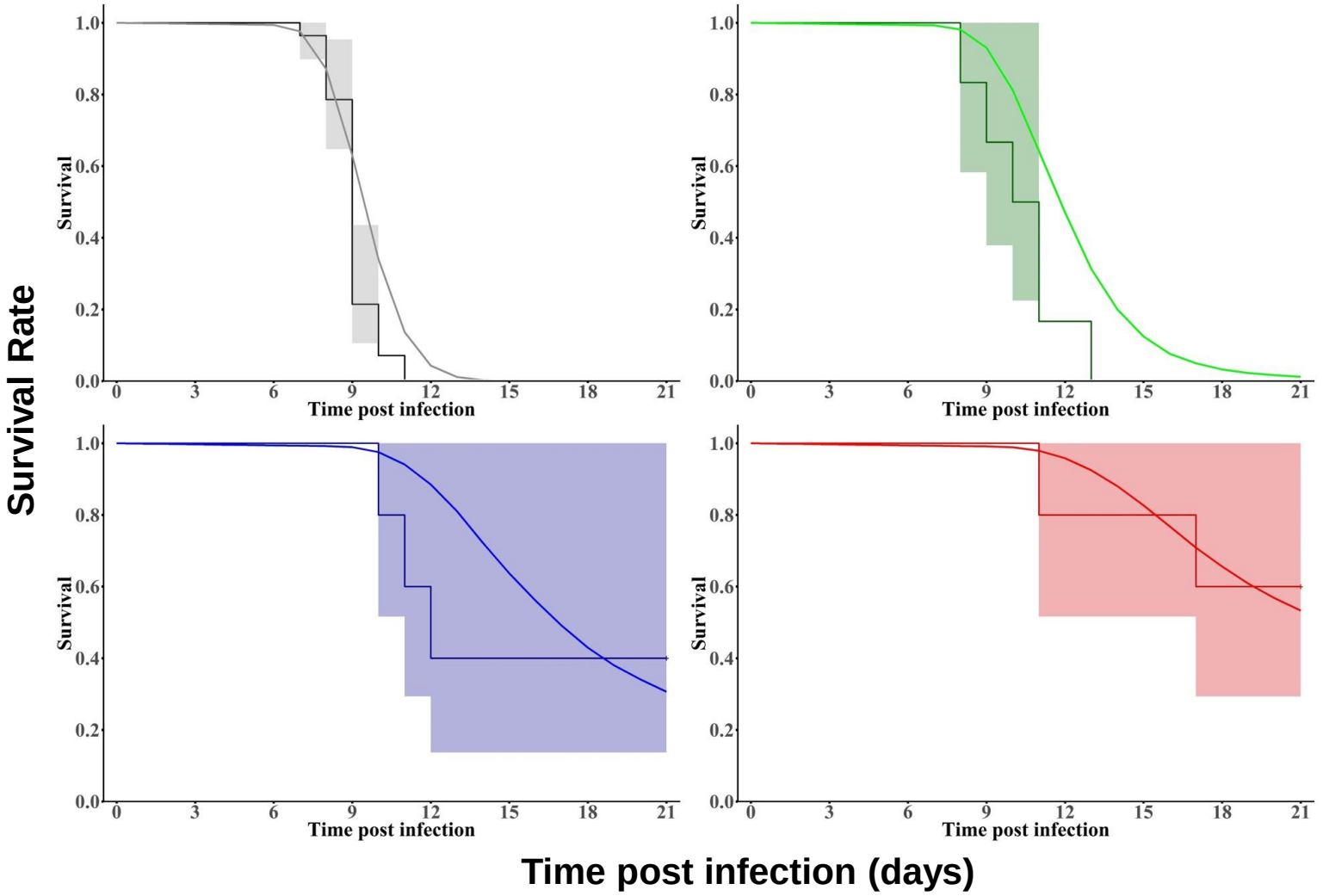
Model predictions of survival rate



Time to death

Joint model to assess the impact of viral load and cytokine dynamics on survival

- The joint model recapitulates the survival rate at day 21 in each treatment group



Validation of model predictions using remdesivir data

Warren et al, Nature, 2016

LETTER

doi:10.1038/nature17180

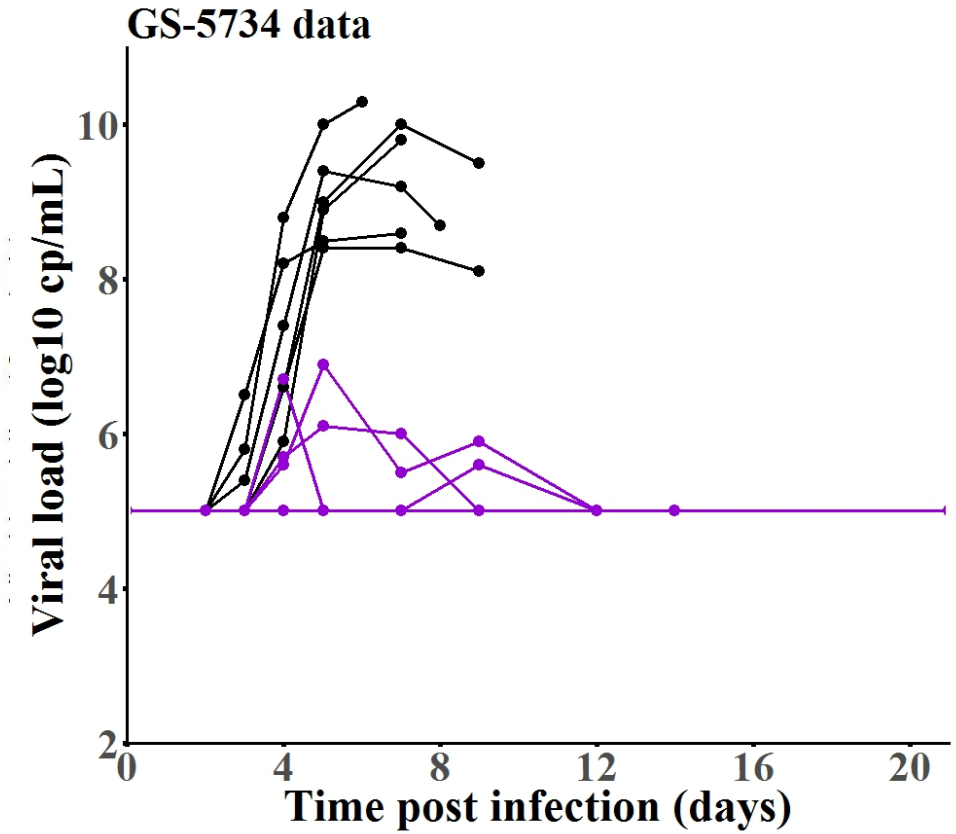
Therapeutic efficacy of the small molecule GS-5734 against Ebola virus in rhesus monkeys

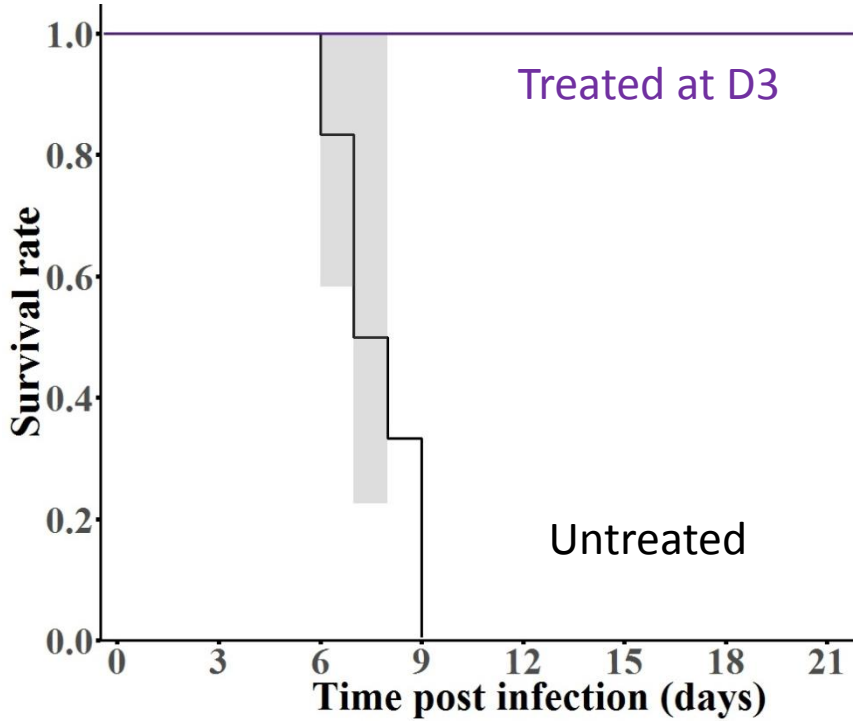
including testes, eyes, and brain. In a rhesus monkey model of EVD, once-daily intravenous administration of 10 mg kg^{-1} GS-5734 for 12 days resulted in profound suppression of EBOV replication and protected 100% of EBOV-infected animals against lethal disease, ameliorating clinical disease signs and pathophysiological markers, even when treatments were initiated three days after virus exposure

We used the model (with variation of R_0 parameter) to fit viral load data only:

→Found ε = 0.9

→With this level of effectiveness 100% survival predicted by the model





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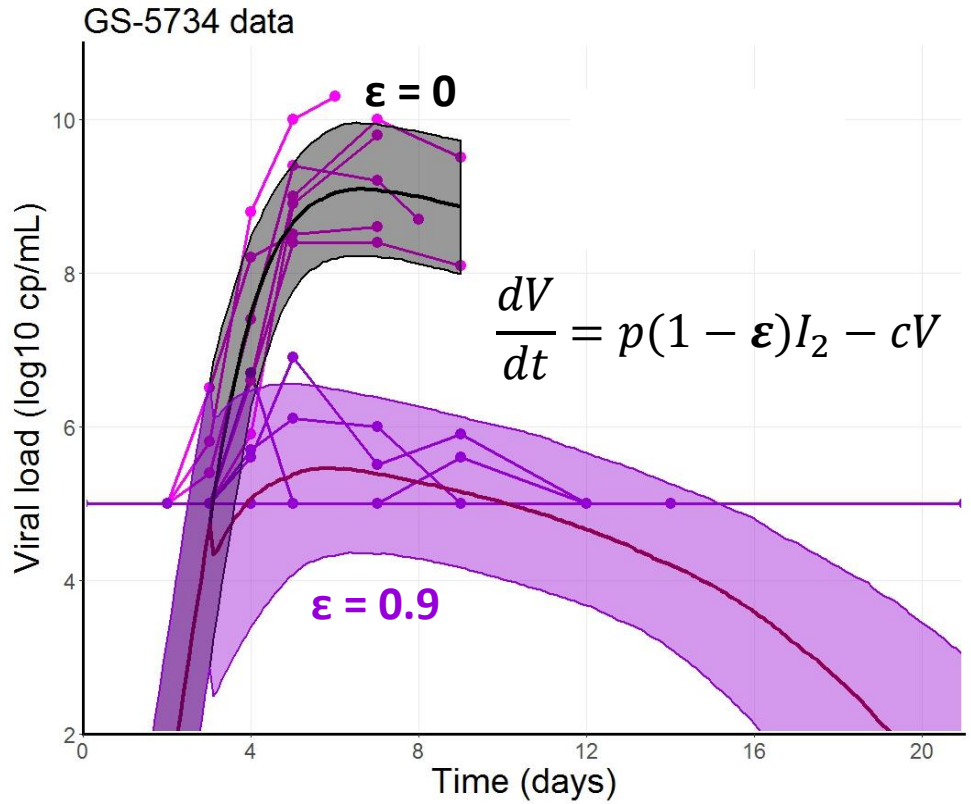
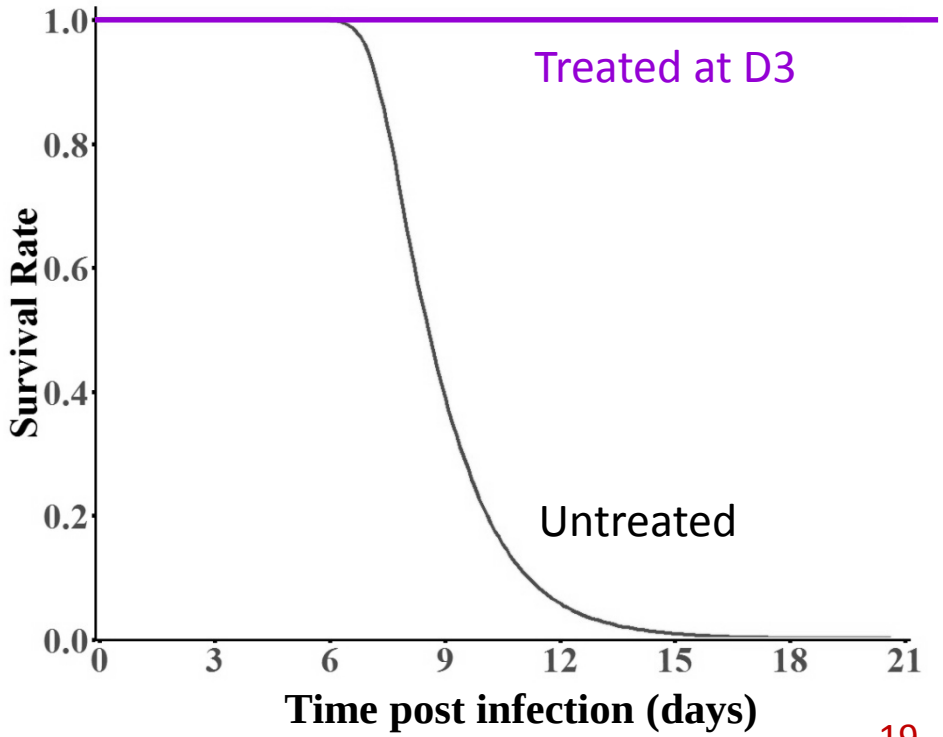
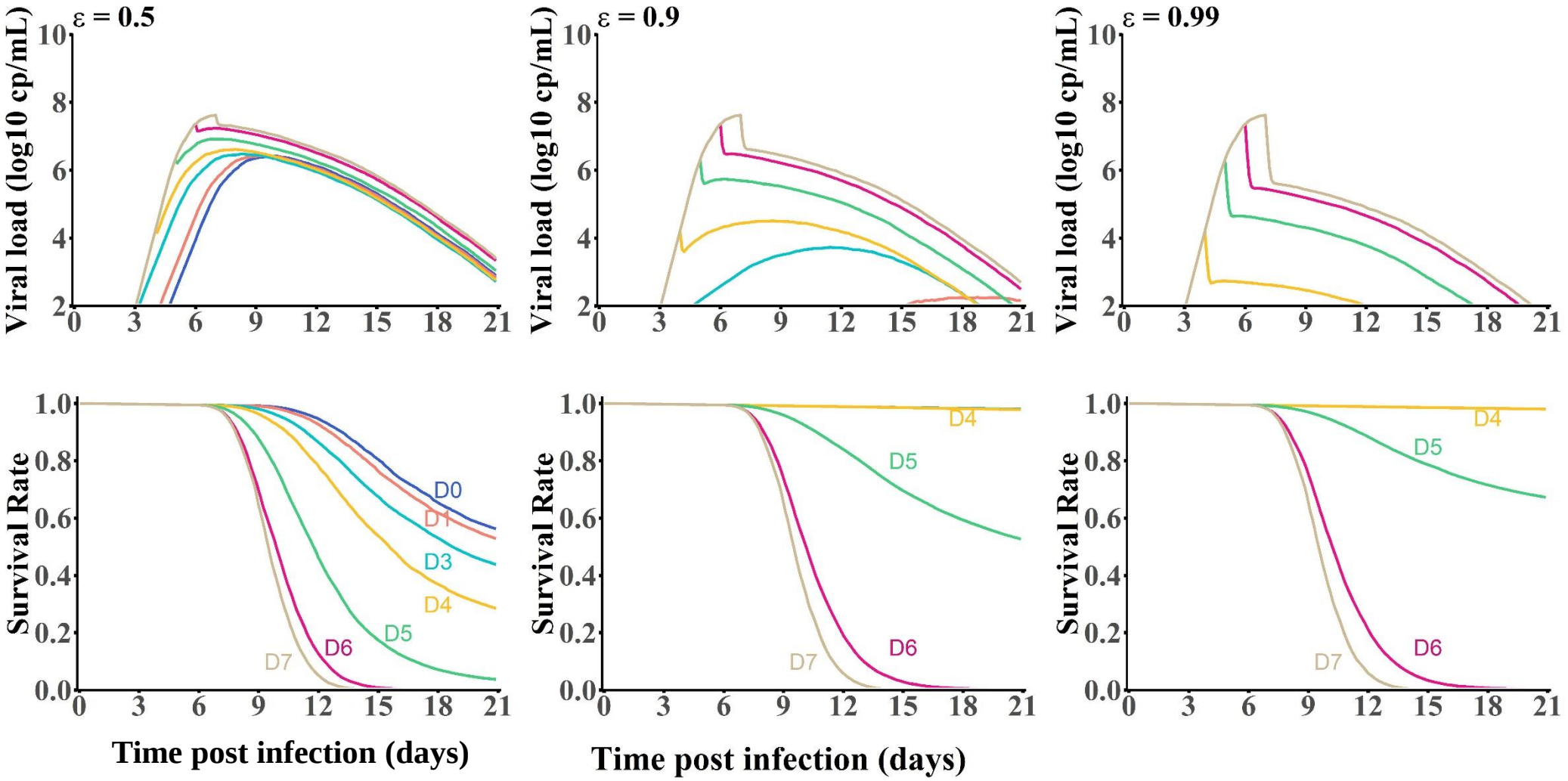




Impact of the timing of treatment initiation on survival



Treatment initiation: Day 0, Day 1, Day 3, Day 4, Day 5, Day 6, Day 7

Summary on EVD modeling

- Best fit to the data was obtained with models assuming that pro-inflammatory cytokines (IFNα, IL6) were associated with
  → control of viremia via the reduction of target cell population during acute infection
  → time-to-death with a stronger impact than viral load

- Favipiravir initiated two days prior the infection had a moderate impact on viral replication with average ε = 50% at 180 mg/kg BID

- Model predicts that antiviral drugs may improve survival rate in NHPs only if initiated before the cytokine storm

Patient admission occurred most often close to the viremia peak

PLOS | MEDICINE

RESEARCH ARTICLE

Experimental Treatment with Favipiravir for Ebola Virus Disease (the JIKI Trial): A Historically Controlled, Single-Arm Proof-of-Concept Trial in Guinea

The NEW ENGLAND JOURNAL of MEDICINE

ORIGINAL ARTICLE

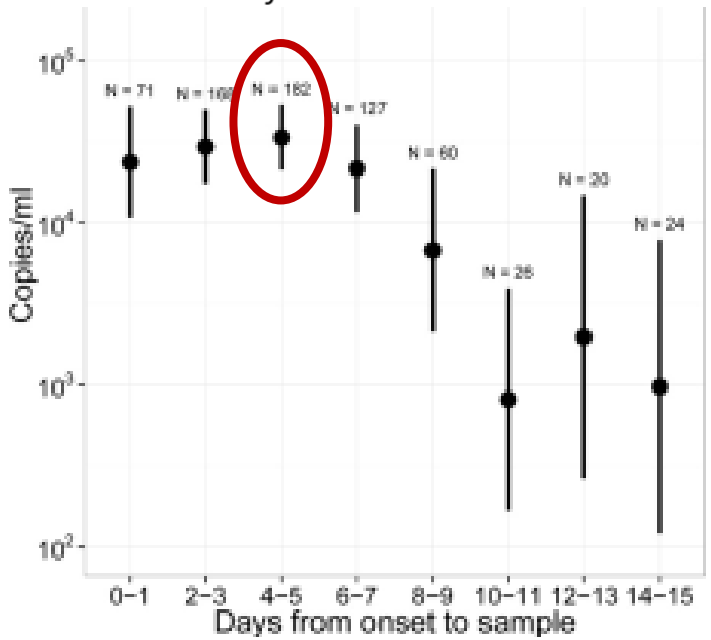
A Randomized, Controlled Trial of ZMapp for Ebola Virus Infection

The PREVAIL II Writing Group, for the Multi-National PREVAIL II Study Team*

PLOS | MEDICINE

RESEARCH ARTICLE

Use of Viremia to Evaluate the Baseline Case Fatality Ratio of Ebola Virus Disease and Inform Treatment Studies: A Retrospective Cohort Study



- Time from symptom onset to inclusion were 4.2 and 3.5 days in the two main clinical trials during the last EVD outbreak
- Retrospective analysis indicated that maximal level of viremia was observed 4 to 5 days after symptoms onset
- These results suggest that majority of patients initiated treatment close to their time to viremia peak
- This supports the design of prophylaxis or post exposure trials for the evaluation of direct antiviral in future outbreaks

Sissoko et al, Plos med, 2016
Prevail study group, NEJM, 2016
Faye et al, Plos med, 2015

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