Mathematical analysis of clinical data suggests that HCV genotype 1 has a high cellular infection rate that is reduced by pegylated-interferon alfa-2a/ribavirin

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Introduction

- Mathematical modelling of HCV RNA response during treatment with pegylated interferon-alfa (P) and ribavirin (R) has shown that the viral production rate is inhibited;[1] however, some important system parameters, such as the cellular infection rate, are difficult to estimate. Indeed, the rapid breakthrough of resistant variants observed during monotherapy with many specifically targeted antiviral therapies (STAT-Cs) suggests that the initial infection rate may have been underestimated.

- Here we describe the HCV RNA profiles from patients from the Optimal Protease inhibitor Enhancement of Response to TherApy (OPERA)-1 study who received the HCV NS3/4A protease inhibitor, TMC435, as monotherapy followed by triple therapy (TMC435 in combination with P/R), in order to investigate the mode of action of P/R and to obtain a better estimate of the infection rate.

Patient data

- Data from two cohorts of the Phase IIa OPERA-1 study (TMC435-C201) were used, in which treatment-naive HCV-genotype-1-infected patients received one week of TMC435 at three different doses followed by three weeks of triple therapy (TMC435 in combination with P/R), and subsequent therapy with P/R alone.[2]

- This analysis focused on patients who had an increase of >0.5 log$_{10}$ IU/mL in plasma HCV RNA from the lowest level reached during TMC435 monotherapy and an immediate (patients A and B) or delayed (patients C and D) HCV RNA decline upon addition of P/R therapy (Figure 1).
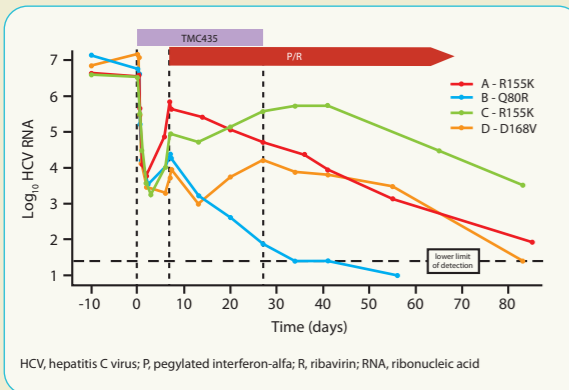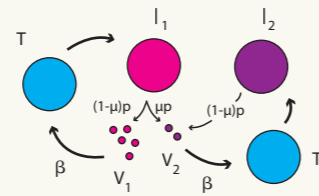

Figure 1. Individual HCV RNA profiles following up to 12 weeks of therapy in four patients who experienced an increase of >0.5 log$_{10}$ IU/mL in plasma HCV RNA from nadir during TMC435 monotherapy, and an immediate (patients A and B) or delayed (patients C and D) HCV RNA decline upon addition of P/R therapy. For each patient, an NS3 protease mutation was assigned based on emerging mutations determined using population sequencing.

HCV, hepatitis C virus; P, pegylated interferon-alfa; R, ribavirin; RNA, ribonucleic acid

- The following mutations emerged and were considered for this analysis: Q80R (patient B), R155K (patients A and C) and D168V (patient D).

Mathematical modelling and simulation

- Simulations of HCV RNA profiles during therapy with P/R were performed using the one-strain Neumann mathematical model.[1]

- This model was then extended towards a two-strain framework to account for the emergence of resistant viral variants (Figure 2).
  - This model fitted the data better than alternatives using different assumptions regarding the T-compartment of uninfected hepatocytes (data not shown).[3-5]

$$dT/dt = \lambda - dT - (1-\eta)\beta(V_1 + V_2)T$$
$$dI_1/dt = (1-\eta)\beta V_1 T - \delta I_1$$
$$dI_2/dt = (1-\eta)\beta V_2 T - \delta I_2$$
$$dV_1/dt = (1-\epsilon_1)(1-\mu)pI_1 - cV_1$$
$$dV_2/dt = (1-\epsilon_1)\mu pI_1 + (1-\epsilon_2)(1-\mu)f_2 pI_2 - cV_2$$

T, uninfected hepatocytes; I, hepatocytes infected with strain i; V, virions of strain i; d, loss rate of T; δ, loss rate of I; c, viral clearance rate; λ, supply rate of uninfected hepatocytes; p, viral production rate; β, viral infection rate; μ, mutation rate; f$_2$, fitness of strain 2; ε$_i$, drug efficacy at inhibiting viral production for strain i; η$_i$, drug efficacy at inhibiting viral infection

Figure 2. The Neumann mathematical model extended towards two viral strains.

Simulations focusing on HCV infection rate in a one-strain model

- The two most common HCV RNA profiles during P/R therapy are a biphasic decline and a decline to a new steady state.[1] Simulated examples are shown in Figure 3 (red and green dashed lines, respectively).
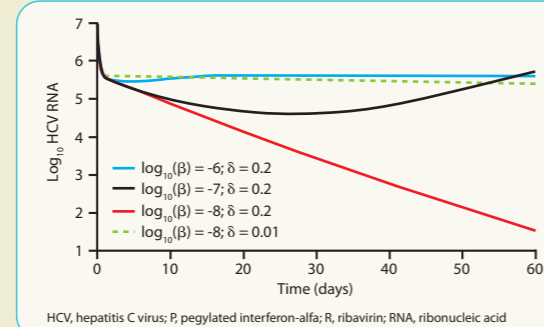

Figure 3. Simulations of HCV RNA profiles using the one-strain Neumann mathematical model[1] for P/R therapy using a typical 90% inhibition of viral production (ε$_{P/R}$) and a baseline HCV RNA level (V$_0$) of 6.6 log$_{10}$ IU/mL. Changing parameters are the infection rate (β) and the loss rate (δ) of infected cells.

HCV, hepatitis C virus; P, pegylated interferon-alfa; R, ribavirin; RNA, ribonucleic acid

- Biphasic decline can only be modelled with a low infection rate (β): log$_{10}$ of the order -8 (note that this is an estimate for β under P/R therapy).

- Decline to a new steady state can be modelled either conventionally by a low β and loss rate (δ) of infected cells (green dashed line), or with a high β and δ (blue line).

A high infection rate can describe the rapid rise in HCV RNA after initial decline

- Given the steady-state baseline HCV RNA levels, only two parameters have an influence on the slope of the rising mutant virions: β and δ.

- δ was estimated from the decline during P/R therapy.

- β and fitness of viral variants were estimated based on the first week of monotherapy to remove the influence of P/R therapy.

- The inhibition of viral production for the selected mutations was deduced from an E-max formula (considering PK and in vitro data) and was fixed as: ε$_{Q80R}$ = 0.75, ε$_{R155K}$ = 0.5, ε$_{D168V}$ = 0.01.

- The infection rate best fitting the HCV RNA profiles was two orders of magnitude higher than conventionally assumed (Figure 4).

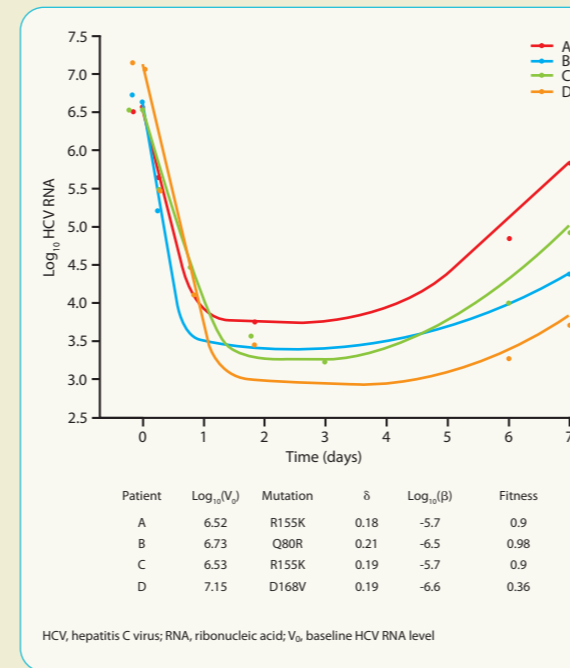| Patient	Log$_{10}$(V$_0$)	Mutation	δ	Log$_{10}$(β)	Fitness
A	6.52	R155K	0.18	-5.7	0.9
B	6.73	Q80R	0.21	-6.5	0.98
C	6.53	R155K	0.19	-5.7	0.9
D	7.15	D168V	0.19	-6.6	0.36

Figure 4. Best-fit models of changes in HCV RNA levels during the first week of TMC435 monotherapy. Dots represent observed HCV RNA levels, with curves representing best-fit models. Baseline HCV RNA levels and associated NS3 protease mutation, fits for loss rate (δ) of infected cells obtained from the decline during P/R therapy (see Figure 1), infection rate (β) and fitness of the mutant are shown for each patient.

HCV, hepatitis C virus; RNA, ribonucleic acid; V$_0$, baseline HCV RNA level

Inhibition of infection rate by P/R therapy

- A second perturbation occurred when P/R was added to the therapy.

- Several alternative models for the mode of action of P/R were examined to describe the observed profiles:
  - immediate or gradual increase in the rate of infected cell loss[3]
  - gradual reduction of the viral production rate
  - gradual inhibition of the infection rate.

- The observed profiles were best described using the third model.

- The inhibition (η) of the infection rate by P/R was estimated at ~99% and the individual time (t$_η$) to reach 50% of the maximal inhibition ranged from 3–20 days (Figure 5).

Patient	η$_{max}$	t$_η$ (days)
A	0.996	5.7
B	0.99	3
C	0.997	20
D	0.99	17

HCV, hepatitis C virus; P, pegylated interferon-alfa; R, ribavirin; η$_{max}$, maximum inhibition of the infection rate; RNA, ribonucleic acid; t$_η$, time to reach 50% of η$_{max}$
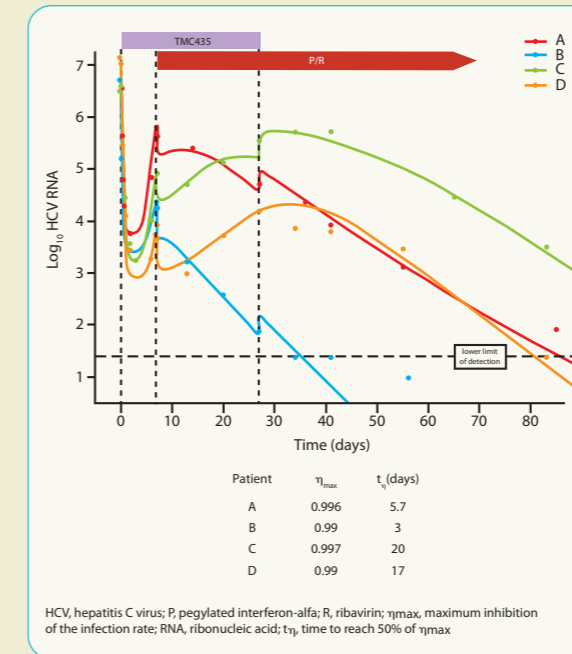
Figure 5. Best-fit models of changes in HCV RNA levels during one week of TMC435 monotherapy followed by triple therapy (TMC435 in combination with P/R). Fitting was based on the hypothesis that P/R therapy augments the inhibition (η) of the infection rate (β) from 0 to η$_{max}$ and reaches 50% of its maximal inhibition in t$_η$ days. Dots represent observed HCV RNA levels, with curves representing best-fit models. Fitted values for η$_{max}$ and t$_η$ are shown for each profile.

The proposed model successfully describes triphasic HCV RNA profiles during P/R therapy

- Besides the most common biphasic HCV RNA profiles, triphasic profiles have also been reported during P/R therapy.[3]

- The proposed inhibition of the infection rate by P/R can also describe these triphasic HCV RNA profiles (Figure 6).

Profile	Log$_{10}$(V$_0$)	ε$_{P/R}$	δ	Log$_{10}$(β)	η$_{max}$	t$_η$ (days)
1	6.35	0.53	0.45	-6.3	0.98	20
2	6.36	0.77	0.17	-6.3	0.98	14
3	5.92	0.37	0.26	-6.0	0.99	10

HCV, hepatitis C virus; P, pegylated interferon-alfa; R, ribavirin; RNA, ribonucleic acid
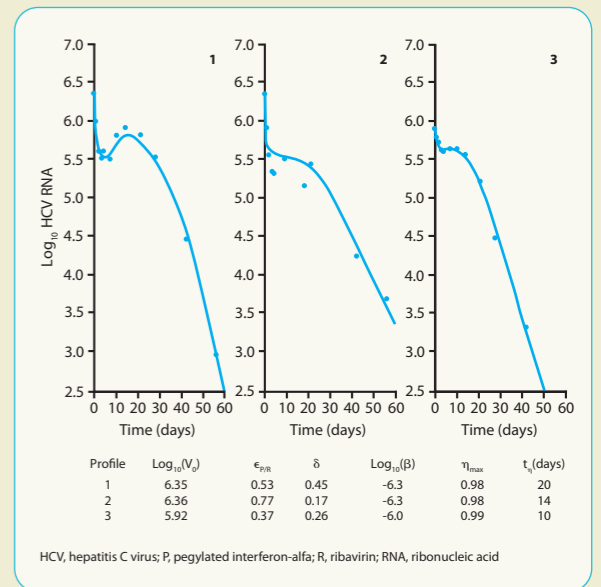
Figure 6. Triphasic changes in HCV RNA levels during P/R therapy. Dots represent digitised HCV RNA levels from triphasic profiles during P/R therapy, as reported by Herrmann et al.[3] Full lines represent the best fit based on the one-strain Neumann model with gradual inhibition of infection rate. Baseline HCV RNA levels (V$_0$), fitted percentage inhibition (ε$_{P/R}$), loss rate (δ) of infected cells, infection rate (β), maximum inhibition (η$_{max}$) of the infection rate and time (t$_η$) to reach 50% of η$_{max}$ are shown for each profile.

Conclusions

- The cellular infection rate in genotype-1 HCV may have been underestimated.

- A higher cellular infection rate may help explain the rapid breakthrough of mutant variants observed during STAT-C monotherapy.

- In addition to inhibiting viral production, P/R may also inhibit cellular infection and thus interfere with the HCV replication cycle on two levels.

- The underlying mechanism of inhibition of the cellular infection rate by P/R remains speculative.[6,7]

References
1. Neumann U et al. Science 1998; 282: 103–107.
2. Manns M et al. Presented at the 44th Annual Meeting of European Association for the Study of the Liver (EASL), Copenhagen, Denmark, 22–26 April, 2009.
3. Herrmann E et al. Hepatology 2003; 37: 1351–1358.
4. Dahari H et al. Hepatology 2007; 46: 16–21.
5. Colombatto P et al. Clin Pharmacol Ther 2008; 84: 212–215.
6. Dixit NM et al. Nature 2004; 432: 922–924.
7. Murao K et al. Gut 2008; 57: 664–671.