Analysis of the *in vitro* susceptibility of primary isolates of HIV-2 from Portugal to dolutegravir

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Introduction

HIV-2 is endemic in West Africa and has spread throughout the World. However, the therapeutic regimens for HIV-2-infected patients are much more limited than for HIV-1. A limited number of studies have shown that HIV-2 isolates are susceptible to inhibition by currently available integrase inhibitors (raltegravir, RAL, elvitegravir, EVG, and dolutegravir, DTG). RAL is equally active against wild-type HIV-2 and HIV-1, and is therefore a promising option for the treatment of HIV-2- infected patients. DTG is the most recently approved compound of this class. It exhibits potent antiviral activity and limited cross-resistance *in vitro* to most RAL-resistant HIV-1 mutants, with the exception of G140S/Q148R mutants at the integrase gene. DTG may be a good therapeutic option for patients with HIV-2 infection, including those that previously failed other integrase inhibitors. In this study we aimed to compare the antiviral activity of DTG and RAL against a panel of 15 primary isolates from Portugal of which 12 were obtained from integrase-inhibitor patients and 3 were obtained from patients failing a RAL-based therapeutic regimen.

Methods

The antiviral activity was evaluated using a single-round viral infectivity assay. TZM-bl reporter cells were incubated with several fold dilutions of DTG or RAL, for 1h at 37°C, in growth medium supplemented with DEAE-dextran. Cells were infected with 200 TCID50 of each virus. After 48h of infection, luciferase expression was quantified with the Pierce Firefly Luc One-Step Glow Assay Kit (ThermoFisher Scientific, USA) according to manufacturer's instructions. IC50, IC90 and Hill slopes were estimated by the sigmoidal dose–response (variable slope) equation in Prism verson 5.01 for Microsoft (GrahPad Software, San Diego, California USA, www.graphpad.com).

Results

Mean Maximum Percentage of Inhibition (MPI) was similar for DGT and RAL (91.5% vs 92.7%). With the exception of isolate 00PTHDECT, DTG inhibits the replication of most primary isolates of HIV-2 as well as RAL. Mean IC_{50} was 1.3nM for DTG and 65.9nM for RAL and mean IC_{90} was 168.7nM for DTG and 1690.3nM for RAL. There was no statistically significant difference between the IC_{50} of RAL and DGT against our isolates (P= 0.2662). Importantly, DTG showed potent activity against three RAL resistant primary isolates obtained from patients failing a RAL-based therapy.

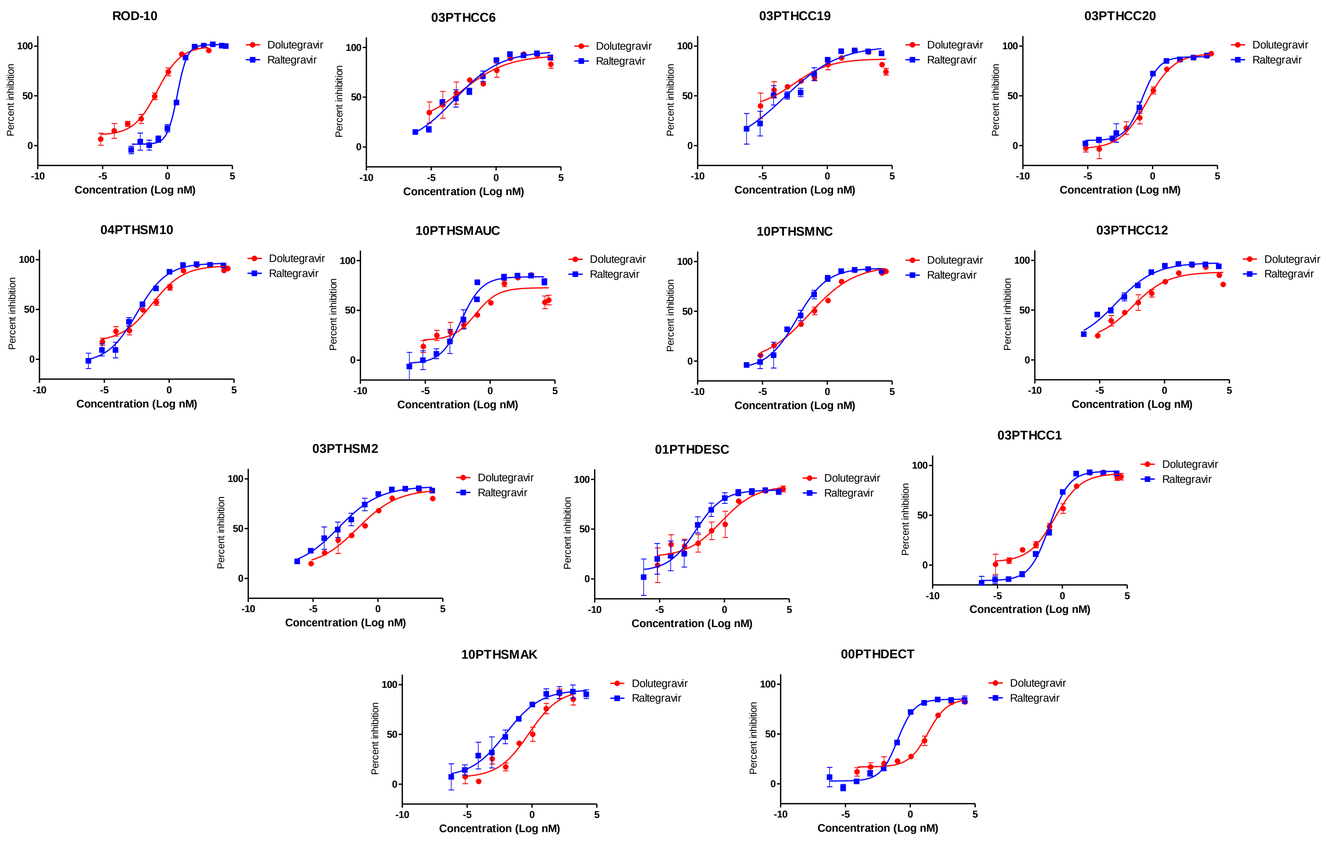

Figure 1 – Dose-response curves showing the percent inhibition of replication of HIV-2 by DTG and RAL. Only isolates that are sensitive to RAL are shown. The ROD reference strain shows some natural resistant to RAL.

Table 1 – IC50, IC90 and MPI values of DTG against HIV-2 isolates from Portugal.

Dolutegravir			
HIV-2	IC50 (nM)	IC90 (nM)	MPI
ROD10	0.1670	16.2181	98.7 (140.4nM)
15PTHCEC	0.7175	1306.1709	87.8 (33884.4nM)
10PTHJSIG	3.1780	135.2073	92.0 (1544.8nM)
15PTHJSIG	0.4140	10.1158	94.9 (140.4nM)
03PTHCC1	0.2711	44.2588	88.7 (33884.4nM)
03PTHCC6	0.0022	4.2540	93.1 (140.4nM)
03PTHCC19	0.0029	3.4324	95.6 (140.4nM)
04PTHSM10	0.0501	21.4289	94.8 (1544.8nM)
10PTHSMAUC	0.0779	5.3555	85.4 (1544.8nM)
10PTHSMNC	0.0419	290.4023	92.4 (1544.8nM)
03PTHCC12	0.0035	3.3589	95.1 (140.4nM)
03PTHSM2	0.0207	12.3311	89.7 (140.4nM)
01PTHDESC	0.6032	526.0173	90.3 (33985.8nM)
10PTHSMAK	0.1726	9.2619	91.0 (140.4nM)
03PTHCC20	0.3820	72.6106	92.4 (33884.4nM)
00PTHDECT	14.650	238.7811	82.2 (16982.4nM)
Mean	**1.2972**	**168.7003**	**91.5**

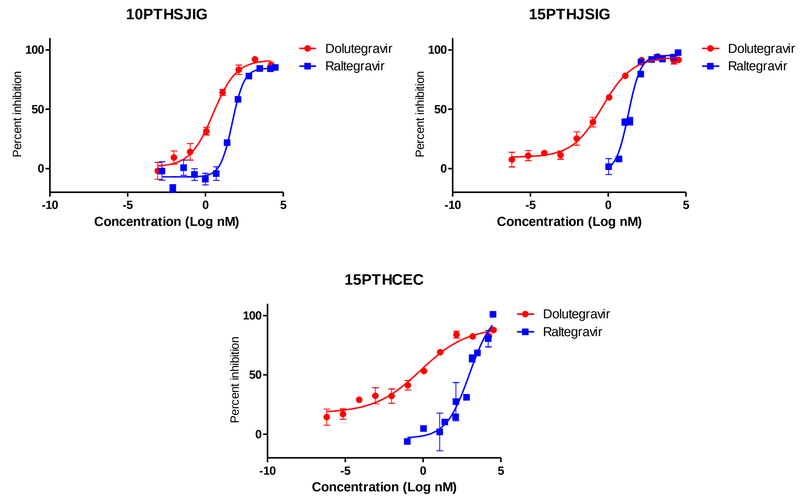

Figure 2 – Dose-response curves showing the percent inhibition of replication of HIV-2 isolates resistant to RAL by DTG. Only isolates that are resistant to RAL are shown.

Table 2 – IC50, IC90 and MPI values of RAL against HIV-2 isolates from Portugal.

Raltegravir			
HIV-2	IC50 (nM)	IC90 (nM)	MPI
ROD10	5.7860	41.1149	101.8 (3108.6nM)
15PTHCEC	974.0000	26424.0876	101.1 (30902.9nM)
10PTHJSIG	53.8600	376.7038	85.2 (31086.8nM)
15PTHJSIG	19.9800	148.5936	97.7 (31086.8nM)
03PTHCC1	0.1027	4.7044	92.2 (15488.2nM)
03PTHCC6	0.0007	5.5655	94.1 (1413.0nM)
03PTHCC19	0.0005	22.0293	95.7 (128.5nM)
04PTHSM10	0.0033	1.0993	95.6 (128.5nM)
10PTHSMAUC	0.0063	0.9470	84.9 (128.5nM)
10PTHSMNC	0.0046	1.6118	92.4 (1413.0nM)
03PTHCC12	0.0002	0.3810	94.4 (11.7nM)
03PTHSM2	0.0009	2.8054	90.5 (1413.0nM)
01PTHDESC	0.0061	1.2057	89.1 (1413.0nM)
10PTHSMAK	0.0097	8.1847	92.9 (1413.0nM)
03PTHCC20	0.1611	3.1681	90.6 (15488.2nM)
00PTHDECT	0.1051	2.8041	84.3 (15488.2nM)
Mean	**65.8767**	**1690.3129**	**92.7**

Conclusions

DGT is a potent inhibitor of the replication of primary isolates of HIV-2 obtained from patients naïve to therapy with integrase inhibitors. DGT is also a potent inhibitor of RAL resistant isolates obtained from patients failing RAL-based regimens. Overall, the results indicate that DGT can be used to treat HIV-2 infected patients even those failing therapy with RAL.

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