Virological response to short-course maraviroc monotherapy does not predict viral tropism in HIV-1-infected treatment-naive patients

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Objectives: We aimed to evaluate whether virological response to a short course of maraviroc monotherapy could predict HIV-1 tropism.

Methods: A clinical trial was performed in HIV-1 treatment-naive patients infected with R5- or non-R5-tropic virus determined using the Trofile® assay, with >1000 HIV-1 RNA copies/mL. Maraviroc was administered for 10 days. Viral load was measured at baseline and days 4, 7, 10 and 28. The main outcome measurement was the decline in HIV-1 RNA at day 10. The trial was registered in the ClinicalTrials.gov database (NCT01060618; TROPISMVC).

Results: Forty patients [30 R5 and 10 dual/mixed (D/M)] were recruited. There was a significant decrease in HIV-1 RNA after 10 days of maraviroc treatment in patients with R5-tropic virus (median 1.52 log10 RNA copies/mL; 95% CI 1.23–1.63; P = 0.0001), but also in patients with D/M-tropic virus (median 1.62 log10 RNA copies/mL; 95% CI 0.33–1.88; P = 0.00024). The difference in the HIV-1 RNA decrease (−0.16 log10 RNA copies/mL; 95% CI −0.53 to 0.22) was not significant (P = 0.410). A decrease >0.5 log10 RNA copies/mL was found in 96.3% of patients with R5-tropic virus and in 70% of patients with D/M-tropic virus (P = 0.052). The differences were not significant when a decline of 1 log10 RNA copies/mL was considered (92.6% versus 70%; P = 0.11).

Conclusions: Treatment-naive patients infected with R5- or D/M-tropic virus have similar virological responses to a short course of maraviroc monotherapy. This clinical test thus cannot be used as a surrogate marker of viral tropism in this population.

Keywords: clinical trials, R5-tropic, dual/mixed-tropic

Introduction

Maraviroc, a CCR5 coreceptor antagonist, is indicated as part of combination antiretroviral therapy (cART) in R5-tropic HIV-1-infected patients; an HIV-1 tropism assessment must therefore be done prior to its administration. There is no global agreement on which test should be used, and both genotypic and phenotypic tests have been clinically validated. The Department of Health and Human Services Panel on Antiretroviral Guidelines for Adults and Adolescents recommends a phenotypic test as the preferred option, while the European Consensus Group on Clinical Management of Tropism Testing recommends both types with the same level of evidence, leaving the choice to the local assessment of logistics and costs. The prerequisite for tropism testing potentially limits the access of patients to maraviroc and hence it is desirable to find an alternative diagnostic test feasible for use by low-resource countries.

Maraviroc monotherapy administered for 10 days in dosagefinding studies resulted in important viral load (VL) reductions (1.7 log10), and when added to optimized backbone treatments led to statistically significant VL reductions compared with placebo.

Potential worries have arisen regarding the de novo emergence of X4-tropic virus or pre-existing X4-tropic variants and the selection of maraviroc resistance. Available in vitro data show that repeated HIV-1 passes in the presence of maraviroc do not result in the selection of X4-tropic HIV-1 and maraviroc-resistant strains are difficult to select. These data have been confirmed in clinical trials, including Phase II studies using maraviroc monotherapy. Considering all these facts, we aimed to evaluate whether the reduction of VL after a short course of maraviroc monotherapy in HIV-1-infected patients who had never received cART correlated with HIV-1 tropism determined using the Trofile® assay (Monogram Biosciences®, South San Francisco, CA, USA).
Virological response to maraviroc does not predict tropism

 Patients and methods

Study design and setting

A pilot, open-label Phase II clinical trial was conducted at Hospital Universitario Ramón y Cajal–IRYCIS and Hospital Universitario La Paz–IdiPaz (Madrid, Spain) between 2008 and 2012. Maraviroc monotherapy was administered for 10 days. Maraviroc has been developed and was provided by Pfizer Inc. (New York City, NY, USA).

Patients

Eligible patients were aged ≥18 years, had chronic HIV-1 infection, had not received previous ART or did not meet guideline criteria for ART initiation and had HIV-1 RNA >1000 copies/mL. No patient was recruited if current or planned pregnancy during the study was anticipated.

Sample size

Based on previous studies on HIV-1 treatment-naive patients, it was expected that ~85% of the patients would have R5-tropic virus and 15% would have non-R5-tropic virus. The inclusion of 30 patients per group was calculated to be enough to find an association between VL reduction and the Tropfile assay result. Recruitment would have been stopped when 30 subjects per group were reached. To include 30 subjects with non-R5-tropic virus, it would have been necessary to perform the Tropfile assay in ~200 patients.8

Development of the study

The research was conducted in accordance with the Declaration of Helsinki and the national and institutional standards. The study protocol (EudraCT registration number 2008-007208-28) was approved by the Spanish Regulatory Agency (Agencia Española del Medicamento y Productos Sanitarios–AEMPS; ref.: MUH/Clin N° 8585; 6 May 2009) and the Ethics Committee (acta n° 208; 24 November 2008) of Hospital Universitario Ramón y Cajal–IRYCIS. Written informed consent was obtained from all participants. The trial was registered in the ClinicalTrials.gov database (NCT01060618; TROPISMVC).

Once informed consent was obtained, plasma samples were sent to Monogram Biosciences®. Since June 2008, the Tropfile assay has been able to detect X4-tropic variants at a threshold of 0.3% of the viral population.8 Plasma VL testing was done in the infectious diseases research laboratory of Hospital Universitario Ramón y Cajal–IRYCIS using bDNA technology (VERSANT HIV-1 RNA 3.0 Assay, Siemens Healthcare Diagnostics Inc., Tarrytown, NY, USA).

After the initiation of the study, patients received open-label maraviroc monotherapy (300 mg/12 h) for 10 days. During this period, HIV-1 VL was quantified with a lower threshold of 50 HIV-1 RNA copies/mL (VERSANT HIV-1 RNA 3.0 Assay) at the baseline visit, at days 4, 7 and 10 while on maraviroc and 18 days after maraviroc withdrawal at the final safety visit.

Statistical analysis

The main outcome measurement was the decay of the log10 HIV-1 RNA VL between baseline and the day 10 maraviroc visit. Decay >0.5 log10 was considered clinically significant and was used as the cut-off. The operational characteristics (sensitivity and specificity) of this cut-off to distinguish among R5 and non-R5 tropism were calculated, together with the exact 95% CI. A cut-off of 1 log10 decay was also explored. Comparison of log10 VL decay between the R5 and non-R5 groups was done by means of the Mann–Whitney U-test, with a 5% significance level. The same analyses were done using the VL decay at days 4 and 7 of maraviroc treatment.

Results

The clinical trial was initially planned to enrol 60 patients: 30 with R5-tropic virus and 30 with non-R5-tropic virus, after screening 200 patients. As the recruitment of patients with non-R5-tropic virus was harder than expected, an interim analysis was conducted. After screening 58 patients, 40 subjects were recruited: 30 with R5-tropic virus and 10 with dual/mixed (D/M)-tropic virus. Three patients with R5-tropic virus were excluded from the analysis due to poor adherence. Table 1 summarizes patients’ baseline characteristics. Maraviroc was well tolerated by all the patients and no serious adverse events were reported.

Decrease in HIV RNA according to Tropfile–determined tropism

Considering the asymmetry of both distributions, mainly that of patients with D/M-tropic virus, the median of the log10 VL decay after 10 days of maraviroc monotherapy was estimated together with the 95% CI. Figure 1 shows the frequency distribution of log10 HIV-1 RNA decay in patients with R5-tropic (left panel) and D/M-tropic (right panel) viruses.

There was a significant decrease in HIV-1 RNA VL after 10 days of maraviroc treatment in patients infected with R5-tropic virus (median 1.52 log10 HIV-1 RNA copies/mL; 95% CI 1.23–1.63; P<0.0001) or D/M-tropic virus (median 1.62 log10 HIV-1 RNA copies/mL; 95% CI 0.33–1.88; P=0.00024). The difference in HIV-1 RNA decay (−0.16 log10 RNA copies/mL; 95% CI −0.53 to 0.22) was not significant (P=0.410).

A decay >0.5 log10 HIV-1 RNA copies/mL was found in 96.3% of patients with R5-tropic virus and in 70% of patients with D/M-tropic virus (P=0.052). The differences were not significant even when a 1 log10 HIV-1 RNA copies/mL decline was considered

Table 1. Baseline characteristics, N=37

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male, n (%)</td>
<td>32 (86.5%)</td>
</tr>
<tr>
<td>Age (years), mean (range)</td>
<td>37 (23–53)</td>
</tr>
<tr>
<td>Risk practice for HIV-1 infection, n (%)</td>
<td></td>
</tr>
<tr>
<td>intravenous drug use</td>
<td>2 (5.4)</td>
</tr>
<tr>
<td>homo/heterosexual</td>
<td>34 (91.9)</td>
</tr>
<tr>
<td>other</td>
<td>1 (2.7)</td>
</tr>
<tr>
<td>Years of HIV-1 infection, mean (range)</td>
<td>3 (0–24)</td>
</tr>
<tr>
<td>Nadir CD4 cell count (cells/mm³), mean (range)</td>
<td>503 (264–934)</td>
</tr>
<tr>
<td>Maximum log10 HIV-1 VL, mean (range)</td>
<td>4.4 (2.7–5.8)</td>
</tr>
<tr>
<td>Baseline CD4 cell count (cells/mm³), mean (range)</td>
<td>617 (332–1253)</td>
</tr>
<tr>
<td>Baseline log10 HIV-1 VL, mean (range)</td>
<td>4.1 (2.0–5.5)</td>
</tr>
<tr>
<td>HIV-1 subtype, n (%)</td>
<td></td>
</tr>
<tr>
<td>B</td>
<td>26 (70.3)</td>
</tr>
<tr>
<td>other</td>
<td>2 (5.4)</td>
</tr>
<tr>
<td>not determined</td>
<td>9 (24.3)</td>
</tr>
</tbody>
</table>
(92.6% versus 70%; \( P=0.11 \)). The operational characteristics of the VL decay were a sensitivity of 96.30% (95% CI 89.17–100.00) and a specificity of 30.00% (95% CI 1.60–58.40).

VL decay at days 4 and 7 was also explored, but no significant differences among R5- or D/M-tropic viruses were documented (data not shown). After analysing the results of this interim analysis, the clinical trial was stopped ahead of time owing to futility, as the main hypothesis was not fulfilled. No more patients were recruited.

**Discussion**

Our results show that a 10 day course of maraviroc monotherapy in HIV-1-infected patients treatment-naive to cART leads to no differences in VL decay, regardless of the Trofile\textsuperscript{w}-determined tropism. In our opinion, the lack of difference in the VL decay kinetics might be due to the fact that none of the non-R5 tropic viruses was pure X4-tropic virus, as none was detected after screening 58 HIV-1-infected patients. The low abundance of pure X4-tropic virus in HIV-1-infected treatment-naive patients has been previously described.\textsuperscript{7} Indeed, among the 10 D/M-tropic recruited patients, the Trofile\textsuperscript{w} report highlighted the fact that in six cases X4-tropic variants were detected at a low level, close to the test sensitivity threshold, so the great majority of the strains were R5-tropic.

By contrast, Genebat et al.\textsuperscript{9} in a study comprising 34 consecutively included HIV-infected treatment-experienced patients showed that VL reduction after short-term maraviroc exposure correlated with Trofile\textsuperscript{w}-determined tropism. The disagreement between the studies might be explained by the fact that X4-tropic variants are more frequent in pretreated patients and at advanced stages of HIV-1 infection.\textsuperscript{7,10,11} This was the case in the study by Genebat et al.,\textsuperscript{9} which included 19 patients undergoing supervised treatment interruption with maraviroc as monotherapy, while 15 were failing cART, with detectable VL, to which maraviroc was added.

In our study, a significant median decrease of 1.62 log\textsubscript{10} HIV-1 RNA copies/mL after 10 days of maraviroc treatment was observed in treatment-naive patients infected with D/M-tropic virus. Moreover, when considering only those with X4-tropic variants detected at a level close to the 0.3% threshold, the median decay was higher (1.77 log\textsubscript{10} HIV-1 RNA copies/mL; data not shown). Acknowledging the low sample size, if a clinically significant cut-off level could be set for minority X4-tropic variants, an issue under ongoing research,\textsuperscript{12–15} patients with D/M-tropic viruses could benefit to some extent from maraviroc in cART, regardless of tropism, provided the rest of drugs in the regimen were effective.

**Conclusions**

Treatment-naive patients infected with R5- or D/M-tropic HIV-1 have similar virological responses to a short course of maraviroc monotherapy. This clinical test cannot be used as a surrogate marker for viral tropism in this population.

**Acknowledgements**

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Transparency declarations
None to declare.

Author contributions
B. H.-N. and S. M. contributed to the clinical trial design and coordinated the study. F. D., M. J. P.-E., J. L. C., J. A. P.-M., A. M., M. E., J. G. and S. M. recruited and followed the patients. N. M.-E. processed the samples for HIV-1 VL quantification. J. Z. designed and carried out the statistical analysis. All authors contributed to the writing of the manuscript.

References