Late stage R5 HIV-1 isolates exhibit reduced baseline sensitivity to Maraviroc *in vitro*

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Background: In our previous studies CCR5-restricted (R5) HIV-1 isolates from individuals in late stage disease displayed reduced sensitivity to inhibition by natural CCR5 ligands and CCR5 antagonist TAK-779. This correlated with an altered mode of CCR5 use. As reduced sensitivity to CCR5 antagonists *in vitro* may be of clinical concern, the main objective of the present study was to investigate if late stage R5 virus also display reduced baseline sensitivity to the licensed CCR5 antagonist Maraviroc (MVC).

Methods: R5 primary isolates obtained from patients with varying CD4+ T-cell counts were evaluated for sensitivity to inhibition by MVC using an *in vitro* inhibition assay comprising stimulated peripheral blood mononuclear cells. R5 virus sensitivity to MVC inhibition was related to CD4+ T-cell count and the absence or presence of AIDS diagnosis. Furthermore, the gp120 V3 region of the R5 isolates was analysed for amino acid polymorphisms previously associated with resistance to CCR5 antagonists and/or to virologic failure in MVC clinical trials.

Results: All isolates were fully inhibited by MVC. However, late stage R5 virus, including five out of nine isolates from individuals with AIDS, displayed reduced baseline sensitivity to MVC inhibition (Figure 1). V3 amino acid polymorphisms 4L and 19S, previously associated with virologic failure in MVC clinical trials, were noted in two of the least susceptible isolates. Reduced baseline sensitivity to MVC correlated with an altered mode of CCR5 use, as displayed by an increased viral ability to utilize chimeric CXCR4/CCR5 receptors for cellular entry.

Conclusions: Late stage R5 HIV-1 infected individuals frequently harbour virus with an altered mode of CCR5 use and reduced baseline susceptibility to MVC. Virus isolates from individuals with higher CD4 T-cell counts were highly sensitive to Maraviroc. Our results provide theoretical support for *in vivo* studies that suggest a benefit of earlier initiation of CCR5 antagonist treatment rather than later. Not only because the risk of the development of CXCR4 using virus variants increases, but also due to the emergence of HIV-1 R5 viruses with reduced baseline sensitivity to MVC during late stage disease.

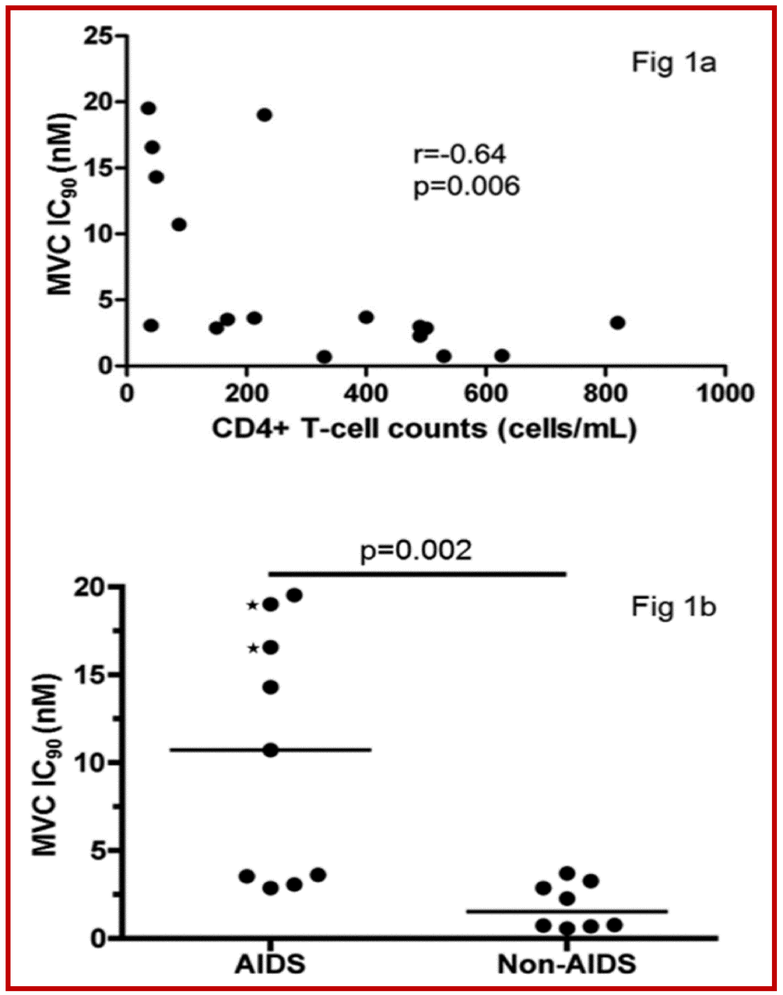

Figure 1. **R5 HIV-1 AIDS isolates display reduced baseline sensitivity to Maraviroc inhibition.** (a) CD4+ T-cell counts correlate with R5 virus baseline sensitivity to Maraviroc inhibition (r=-0.64, p=0.006). (b) All non-AIDS R5 isolates were highly sensitive to inhibition by Maraviroc. R5 HIV-1 AIDS isolates had 4-20 times higher median IC_{90} values for Maraviroc inhibition than non-AIDS R5 isolates (p=0.002). The isolates 13pl and 23pl (depicted by a star) displayed V3 polymorphisms 4L and 19S respectively that previously were related to blunted virological response in Maraviroc clinical trials. Figures display one representative out of three performed experiments.