# HIV co-receptor tropism prediction remains stable over time in treatment naive patients

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## INTRODUCTION

HIV co-receptor tropism must be established before the initiation of treatment with CCR5 receptor antagonists such as Maraviroc. Tropism largely results from to the hypervariable V3 loop of the gp120 HIV molecule, as this is the specific section that binds the CD4 cells' co-receptor. Consequently, determining the amino acid sequence of this region can be used to predict co-receptor usage (Beerenwinkel 2003, Jensen 2003, Low 2008), and this forms the basis of genotypic tropism assessment of HIV-1. HIV-1 co-receptor tropism can be grouped as CCR5 (R5), CXCR4 (X4), dual-tropic or dual mixed (D/M, containing both X4 and R5-tropic viruses).

Current UK guidelines suggest genotypic tropism predictions can be considered valid for up to 90 days. However, data to support this recommendation are limited. We assessed the evolution of the HIV third hyper-variable (V3) loop sequence, and thus HIV-1 co-receptor tropism, in ART-naive patients over time. The aim was to establish how long baseline tropism testing remains valid.

## METHODS

Co-receptor tropism was assessed in 19 ART-naive patients with ongoing viral replication. We retrospectively genotyped samples taken between the baseline, first clinic visit, and final sample before the start of ART, median 53 months (range 33-80 months). Each patient had samples at approximately yearly intervals.

We selectively amplified the V3 loop region of the HIV-1 envelope and performed population sequencing of amplicons. Triplicate testing was performed to increase sensitivity of detection of minority X4 variant quasispecies and the Geno2Pheno system used to predict co-receptor tropism. Samples with clonal false positive rates <6% were deemed X4 tropic.

Both clonal and clinical predictions were made. Clonal predictions made upon the genotypic V3 sequence alone. Clinical predictions combined the genotypic sequence with clinical data – viral load (VL), Nadir CD4 and Nadir % CD4.

Any samples with false positive rates <6%, in any of the triplicate amplifications, were deemed X4 tropic. This is in line with current clinical practice.

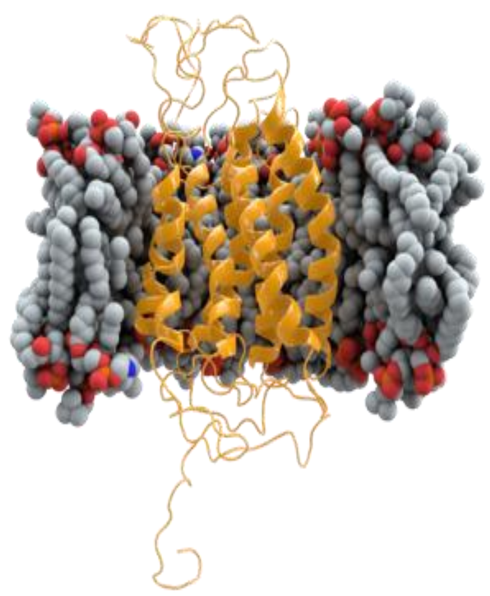

**Fig1.** CCR5 in a cell membrane

## RESULTS

**Table.1.** Tropism predictions in samples over time

| Sample number: | 1 (base line) | 2 | 3 | 4 | 5 | 6 | 7 | 8 |
|---|---|---|---|---|---|---|---|---|
| Patient Number | | | | | | | | |
| 1 | R5 | R5 | R5 | | | | | |
| 2 | R5 | R5 | R5 | R5 | R5 | | | |
| 3 | X4 | X4 | X4 | X4 | X4 | | | |
| 4 | R5 | R5 | R5 | R5 | R5 | R5 | R5 | X4 |
| 5 | R5 | R5 | R5 | R5 | R5 | R5 | | |
| 6 | R5 | R5 | R5 | R5 | R5 | R5 | | |
| 7 | R5 | R5 | R5 | R5 | R5 | R5 | R5 | |
| 8 | R5 | R5 | R5 | R5 | | | | |
| 9 | D/M | D/M | R5 | R5 | R5 | | | |
| 10 | R5 | R5 | R5 | R5 | R5 | R5 | D/M (42m post BL) | |
| 11 | R5 | R5 | R5 | R5 | R5 | R5 | | |
| 12 | R5 | R5 | R5 | R5 | R5 | R5 | | |
| 13 | R5 | R5 | R5 | R5 | R5 | R5 | | |
| 14 | R5 | R5 | R5 | R5 | R5 | R5 | R5 | R5 |
| 15 | X4 | X4 | X4 | X4 | D/M | D/M | | |
| 16 | R5 | R5 | R5 | R5 | R5 | X4 (55m post BL) | R5 | |
| 17 | R5 | R5 | R5 | R5 | R5 | R5 | | |
| 18 | R5 | R5 | R5 | R5 | X4 (54m post BL) | | | |
| 19 | R5 | R5 | R5 | R5 | R5 | R5 | | |

BLUE: non-R5 virus at baseline
BLACK: R5 throughout all samples
RED: R5 at baseline with subsequent X4. Number of months from baseline sample to first X4 detected shown in brackets

Median plasma HIV VL was 20,049 copies/ml (range 965 - 882,256 copies/ml).

Key Results:

- Three patients were identified as having non-R5 tropic virus present at baseline
- Thirteen patients were R5-tropic at baseline and remained R5-tropic throughout a median of 54 months of follow up (range 22-81 months).
- All 16 patients identified as R5-tropic virus at baseline remained R5-tropic for a median of 42 months (range 22-81 months). Hence, these 16 patients eligible for MVC at baseline, remained suitable for MVC treatment for the same period.
- In 3 patients identified as R5-tropic at baseline, X4-tropic virus evolved after a median of 54 months (range 42-55 months).

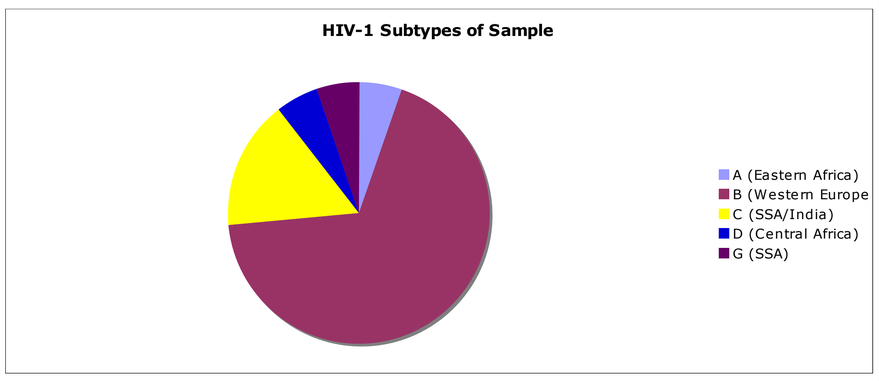


**Fig.2.** The variety and numbers of different HIV-1 subtypes represented in the study

## CONCLUSIONS

Co-receptor tropism in treatment naive patients with ongoing viral replication appears to show a high level of stability over time. Including the specific components that determine co-receptor tropism.

100% of CCR5 antagonist eligible patients remained so at 22 months after initial genotyping. our data suggest that baseline genotypic tropism prediction may be valid for a significant duration in patients delaying the start of anti-retroviral therapy.

This has significant clinical and financial implications, particularly following withdrawal of commercial funding to cover the cost of a tropism test.

**Fig3.** HIV-1 binding to CD4+ cell

## LITERATURE CITED

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