Retrovirology

Poster presentation

P04-52 LB. The importance of well-defined HIV-1 neutralization assays

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Background

Plasma from HIV-1 seropositive individuals show a range of specificities from variant specific to broad cross-neutralizing. The plasma also show different kinetics of neutralization which may be linked to their specificity. The properties may be a consequence of being a polyclonal mixture of antibodies. Isolating monoclonal antibodies from these people would help to resolve these issues.

Methods

Panels of primary isolates of HIV-1 subtypes A, B, C and circulating recombinant form CRF02_AG were prepared in mitogen stimulated PBMCs. Neutralization assays were performed with different lengths of incubation (1 h vs. 24 h), absorption (1 h vs. 24 h) and culture phases (7 days or 14 days for PBMCs and 2 days for cell lines). Plasma from patients infected with either HIV-1 subtype C or CRF02_AG and attending the HIV clinic in Antwerp were screened in PBMC neutralization assays. Monoclonal antibodies were then isolated from memory B cells of individuals whose plasma cross-neutralized strains from multiple subtypes in a high throughput HOS cell based assay using pseudoviruses.

Results

The original donors' plasma were active against relatively resistant HIV-1 isolates in assays with extended incubation phases. In contrast, the monoclonal antibodies were active against relatively sensitive isolates in assays with extended absorption phases. Pseudoviruses had a tendency to be easier neutralized compared to their primary viruses.

Conclusion

The neutralizing properties of the monoclonal antibodies do not reflect those of the plasma from the original donors. Either the method of selecting the monoclonals or the proportion of monoclonals isolated may be a problem. Mixtures of monoclonals may be required for activity against the relatively resistant isolates and the amount of specific antibodies in the plasma is unknown. Alternatively, there may be a discrepancy between specificities of the bone marrow plasma cells and the circulating memory B cells.