

Poster presentation

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P04-I9. Analysis of the binding of multiple clades of HIV-1 by a modified virus capture assay

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from AIDS Vaccine 2009
Paris, France. 19–22 October 2009

Published: 22 October 2009

Retrovirology 2009, **6**(Suppl 3):P47 doi:10.1186/1742-4690-6-S3-P47

This abstract is available from: <http://www.retrovirology.com/content/6/S3/P47>

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Background

We have developed a modified virus capture assay to demonstrate the binding of multiple clades of HIV-1 to polyclonal and monoclonal anti-HIV-1 antibodies on protein G coated plates. The results from the virus capture assay were compared with the TZM-bl virus neutralization assay.

Methods

Protein G plates were coated with 2F5, 4E10, 2G12, 21.2 (pooled positive HIV-1 human sera), or with anti-IgM antibodies. Primary HIV-1 or pseudoviruses (pv) from clades A [92UG(pv), 93RW(pv)], B [MN, US-1, MN(pv), US-1(pv)], C [GS016], and D [57128(pv)], A07412(pv)] were then added to the plates in the presence or absence of sCD4 to determine the ability of the antibodies to capture virus. The amount of bound virus was determined by a p24 ELISA. The appropriate viruses were also tested in a TZM-bl neutralization assay using mAbs 2F5 and 4E10.

Results

Several viruses were captured by both 2F5 and 4E10 mAbs. However, the binding to 4E10 was much greater than the binding to 2F5. US-1(pv) and GS016 were not captured by either 4E10 or 2F5 antibodies and also were not neutralized by the mAbs in the TZM-bl assay. In contrast, 4E10 did not neutralize 93RW(pv) even though it captured the virus. Addition of sCD4 resulted in a significant enhancement of binding of 93RW(pv), MN and A3R5-US1 viruses suggesting that additional binding sites on the viral surface were exposed. A decrease in virus cap-

ture was observed with the mAb 2F5 IgM isotype compared to the IgG isotype.

Conclusion

The modified virus capture assay is highly versatile and is currently being used to screen polyclonal and monoclonal antibodies as well as MPER-specific multispecific IgM mAbs that exhibit both protein and lipid binding.