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Oral presentation

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OA011-03. Clusterin, a natural ligand of DC-SIGN present in human semen inhibits HIV capture and transmission by dendritic cells

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Background

Although sexual transmission represents the main mode of HIV dissemination worldwide, little is known about the influence of semen on viral spread. We have shown that seminal plasma (SP) inhibits both, the attachment of HIV to dendritic cells (DC) and the ability of DC to transmit the virus to T cells. Here, we characterized the inhibitor present in SP and their functional properties.

Methods

Semen samples were collected from healthy donors. SP proteins were analyzed by 2D electrophoresis. DC-SIGN binding proteins were identified by western blot using DC-SIGN^{fc} chimera. Selected spots were cut and clusterin was identified by MS analysis as the inhibitor present in SP. SP clusterin was then purified by affinity chromatography. In all the experiments, HIV was quantified by measurement of p24 antigen by ELISA.

Results

We found that SP clusterin markedly inhibits the attachment of HIV-1 BAL (5 ng p24 antigen) to DC in a dose-dependent mode (1–40 µg/ml), being the percentage of inhibition of 54 ± 11 (n = 6, p < 0.05) when used at a concentration of 15 µg/ml. Similar levels of inhibition were observed using blocking antibodies directed to DC-SIGN.

In transmission experiments DC were cultured with HIV-1 BaL (5 ng p24Ag) in the presence of clusterin (20 µg/ml), washed and cultured with activated peripheral blood mononuclear cells (PBMCs). Clusterin markedly prevented virus transmission to DC: % inhibition = 59 ± 17, n = 5, p < 0.05). Experiments performed with THP-1-DC-SIGN⁺ cells showed that clusterin, at a concentration of 15 µg/ml, almost completely inhibited both, the attachment of HIV (% inhibition < 87%, n = 5) and the ability of THP-1-DC-SIGN⁺ cells to transmit the virus to activated PBMCs (% inhibition < 82%, n = 4).

Conclusion

Our results identified clusterin, as a novel ligand of DC-SIGN present in human semen able to inhibit the capture and transmission of HIV by DC.