Human Breast Milk Inhibits Cell-free but not Cell-associated HIV-1 Infection of CD4+ Cells

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Background. Transmission of HIV-1 from mother-to-child through breastfeeding remains a global health challenge, particularly in developing countries. Recent studies suggest that infected CD4+ T lymphocytes form a significant reservoir for HIV-1 in human breast milk, and may contribute to transmission of HIV-1 during breastfeeding. Breast milk contains innate and adaptive immune factors that may limit virus infection; however the impact of innate factors in milk on HIV-1 infection in CD4+ cells is unclear. We evaluated the ability of breast milk to inhibit infection of CD4+ target cells by cell-free and cell-associated HIV-1.

Methods. Breast milk from HIV-1 negative donors was used to assess the impact of innate factors on HIV-1 infectivity. HIV-1 infection was analyzed by measuring LTR-driven luciferase expression in CD4+ TZM-bl cells, and by assessing HIV-1 reverse transcription, integration and transcription in peripheral blood mononuclear cells (PBMC) by real-time PCR. The effect of breast milk on expression of the early viral transcripts Tat and Rev was quantified by real time PCR, and release of new virions determined by quantifying viral p24 antigen. To determine the effect of breast milk on cell-associated HIV-1 infection, HIV-infected CD4+ T lymphocytes were co-cultured with uninfected TZM-bl cells.

Results. Our results show potent inhibition of cell-free HIV-1 of R5, X4 and R5/X4 tropism by breast milk in a dose dependent manner across a range of viral titers (p?0.05). We observed significant inhibition of HIV-1 reverse transcription ($p=0.004$) and integration ($p=0.02$) in PBMC. In PBMC maintained in breast milk, a significant reduction of early HIV-1 transcripts was seen 3 days post-infection ($p<0.001$) but was lost by day 6. Breast milk was ineffective at reducing HIV-1 release by PBMC throughout culture, and had no significant inhibition on transmission of HIV-1 from infected CD4+ T lymphocytes to susceptible CD4+ cells.

Conclusions. We demonstrate potent inhibition of cell-free HIV-1 by innate factors in human breast milk during early stages of infection of CD4+ cells. We found little or no inhibition of HIV-1 p24 release, and breast milk had no inhibitory effect on cell-associated HIV-1 infection of CD4+ target cells. This suggests that innate factors in breast milk may contribute to protection against transmission of cell-free HIV-1, but may be less effective at blocking cell-associated transmission of virus from infected CD4+ T lymphocytes.

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