Efficient infection of healthy donors´ monocyte derived macrophages by erythrocyte-associated HIV

L. Fazzi, M.N. García, C. Cevallos, R.D. Rabinovich, M.M. Ávila
Buenos Aires University. CONICET, Instituto de Investigaciones Biomédicas en Retrovirus y Sida, Buenos Aires, Argentina

Background: Erythrocytes of HIV+ individuals present p24 Antigen, RNA-HIV and bounded-specific antibodies. This HIV- erythrocyte association was observed to be mediated by immune complexes or complement factors with CR1 receptors in the erythrocyte membrane, and/or the virus associated to the Duffy antigen receptor for chemokines (DARC). Taking into account the importance of macrophagotropic strains in HIV infection, and the close contact of erythrocytes and macrophages during the erythrocyte-clearance process in the spleen and liver, this work focuses on the capacity of the erythrocyte-associated HIV to infect macrophages.

Methods: Erythrocytes and monocyte derived macrophages (MDM) were obtained from 12 healthy donors' buffy coats. Experiments were carried out with erythrocytes and MDM from the same donor. Erythrocytes were incubated with virus in 3 different conditions: **A-** macrophagotropic strain BaL virus; **B-** BaL virus + complement (normal human serum); **C-** BaL virus + antibodies (pool of inactivated serum of HIV+ patients with VL< 50 copies/ml) + complement. After quantification of p24 Ag on erythrocytes, MDM were incubated with erythrocytes obtained in conditions **A**, **B** and **C** for 2 hours at 37ºC. At 2 to 14 days post inoculation (pi), p24Ag was determined on the culture supernatant and immunofluorescence was performed on MDM.

Results: The amount of virus captured by erythrocytes in the 3 *in-vitro* conditions was similar to that present in erythrocytes of patients.
Under condition **B** (HIV + complement) p24Ag was positive in MDM culture supernatant in all samples (80 pg/ml at day 13 pi). Besides, in 4 samples p24Ag was positive in the culture supernatant in conditions **A** and **C**, indicating a productive infection of the MDM. Infectivity was proved on Ghost cells and in other MDM. The MDM immunofluorescence for p24Ag was positive for all samples as from day 6 pi, even in those where no supernatant p24Ag was found (A and C), suggesting that HIV-1 can enter MDM, although no viral replication was observed.

Conclusion: Given that erythrocyte-associated HIV through complement maintains its infectivity on MDM, this viral fraction could be important in the early stages of infection and for reservoir generation.