Mem. Inst. Butantan, v. **54**, n. 2, 1992.

THE ROLE OF COMPLEMENT IN THE PATHOGENESIS OF HIV INFECTION

M. D. Kazatchkine*

CD4 serves as the primary receptor for HIV on T lymphocytes and may be important for viral entry into CD4+ monocyte/macrophages and dendritic cells. Infection of CD4+ cells *in vitro* has been shown to be facilitated by anti-HIV antibodies through Fc receptor-mediated enhancement and by serum from HIV-infected subjects through mechanisms involving both Fc and complement receptors. Recent studies have also shown that a number of CD4- cells are susceptible to HIV infection *in vivo* and *in vitro*, indicating that alternative pathways to CD4 exist for viral penetration and that the host cell range of HIV is broader than originally described.

Complement Activation By HIV.

Intact HIV, recombinant gp 160 of HIV and HIV-infected cells have been shown to activate complement in human serum resulting in the cleavage of C3 and subsequent deposition of C3 fragments on the viral surface or on the surface of infected cells. Complement activation by HIV occurs through the classical pathway following the binding of C1q to conserved peptidic sequences of gp41 and direct activation of the C1 complex. Although activation of C1 by gp41 and by gp160 may occur in the absence of antibodies, it is significantly enhanced by anti-HIV envelope IgG. Under optimal experimental conditions, classical activation pathway by mammalian-derived recombinant gp 160 results in the deposition of one molecule of C3b/IC3b per molecule of gp160. Deposition of C3 fragments on the envelope glycoprotein complex provides the basis for antibody-dependent and independent complement-mediated enhancement of infection of C3 receptor-bearing cells.

Complement-mediated Enhancement of Infection of T Lymphocytes.

Recent studies have indentified the presence of CR1 and CR2 on subsets of human peripheral blood T lymphocytes and of CR2 on thymocytes. CR1 is expressed on approximately 15% of peripheral blood CD4+ and CD8+ T cells. CR1 + T lymphocytes also express Fc gamma receptors as assessed by their ability to bind fluoresceinated aggregated IgG. Most CR1+ T cells also express CR2 is expressed on 50% of both CD4+ and CD8+ peripheral blood T lymphocytes at an approximately 10 fold lower density than on B cells. On the human T cell line HPB-ALL, CR2 and CR1 are co-internalized when cross-linked with anti-receptor antibodies: CR2 is capable of signal transduction.

The first evidence for the role of complement is enhancing HIV infection of T cells came from the observation by Robinson that sera from more than 80% of HIV antibody-positive individuals increased infections virus release from cells of

* Unité d'Immunopathologie — Hôpital Broussais — Paris — França

the MT2 T cell line upon infection with HIV-1 *in vitro*. Enhancement of CD4+ CR2+ infection by seropositive serum required complement, anti-HIV antibodies and the CD4 molecule. We have subsequently found that complement alone (i.e. in the absence of anti-HIV antibodies) was capable of enhancing infection of MT2 cells with suboptimal amounts of HIV-1; productive viral infection of MT2 cells with low inputs of HIV that had been pre-opsonized with normal human HIV-seronegative serum occurred through the interaction of opsonized virus with CR2, since infection was blocked by cross-linked mAb against CR2 but not by a mAb against the gp120 binding site of CD4. Complement alone was also shown to enhance infection of normal peripheral blood T lymphocytes as assessed by its ability to enhance infection of PHA-stimulated seronegative peripheral blood leucocytes cocultured with leucocytes from HIV-infected individuals.

Preliminary evidence indicates that blocking of CH1 partially decreases CD4-dependent infection of the CD4+ CD8+ Cr1+ CR2+ HPB-ALL cell line with complement-opsonized HIV. CR1 on T cells could either function as a receptor mediating viral entry into the cells or as a cofactor for the cleavage of C3b into iC3b and C3dg to allow the interation of opsonized virus with CR2.

Complement-Mediated Enhancement of Infection of Monocyte/Macrophages.

Monocyte/macrophages represent a major reservoir of virus in HIV-infected individuals. The cells express CD4 at low density on the membrane. Depending on their state of maturation, cells of the monocytic lineage also express various amounts of Fc gamma RI (CD64), Fc gamma RII (CD32) and Fc gamma RIII (CD16) receptors, LFA-1 (CD11a/CD18), CR1 (CD35) and CR3 (CD11b/CD18). Complement enhances *in vitro* infection of monocyte/macrophages with HIV in the presence and in the absence of antibodies. We have recently demonstrated that complement enhances the productive infections of cultured normal peripheral blood monocytic of the promonocytic cell line THP-1, and of the cell line MonoMac 6 with HIV-1 and HIV-2. The cells and cell lines express CR1 and CR3. Cultured monocytes and THP-1 cells express low amounts of CD4 whereas MonoMac 6 cells do not express CD4 antigen nor Cd4 transcript. Thus, the enhancing effect of complement on infection of cells of the monocytic lineage may occur independently of CD4, indicating that the interaction of opsonized virus with complement receptors may be sufficient to mediate penetration of HIV into monocytes. Preincubation of target monocytic cells with F(ab)2 fragments of anti-CR1 antibodies or of monoclonal antibodies directed against the alpha chain of CR3 (but not against the alpha chain of LFA-1) suppressed infection of the cells with complement opsonized HIV.

Complement-Mediated Enhancement of Infection of B Lymphocytes.

EBV-transformed human B cell lines, EBV-cell lines and EBV-bearing normal human B lymphocytes are susceptible to HIV-infection *in vitro*. B lymphocytes are activated in HIV infected-individuals. There is no evidence, however, that normal B lymphocytes may be infected with HIV *in vitro* or *in vivo*. Lymphoblastoid B cell lines expressing CR2 and, for some of the lines also express CR1. The expression of CR2 is upregulated by EBV in EBV-transformed B cell lines. All normal mature B lymphocytes express CR1 and CR2.

Early studies have shown that EBV-transformed human B cell lines are susceptible to HIV infection *in vitro*. Depending on the cell line and on the strain of HIV, infection EBV+B cell has resulted in cell lysis or in persistent non-productive or productive infection. HIV has also been shown to infect EBV-Burkitt lymphoma

B cells lines, indicating that susceptibility to HIV infection is not strictly dependent on the presence of the EBV genome. We have recently demonstrated that CR2 maybe mediated on its own the productive infection of cells of the Raji B lymphoblastoid cell line with complement-opsonized HIV. Infection of the cells occurred independently of antibodies and of CD4 molecule since HIV-seronegative serum was used for infection and since the cells lack the expression of CD4 surface antigen and of CD4 transcript.

Enhancement of HIV-infection by complement may contribute to extend the range of target cells for the virus and to increase the rate of infection in patients with a low viral load. Complement-dependent enhancement also allows infection of cells independently of the monocytotropic or lymphocytotropic characteristics of the infective strain. The role of complement in facilitating HIV-infection should be taken into consideration in the design of vaccines and of therapeutic trials.