

**HOST FACTORS MODULATING HIV-1 INFECTIVITY
AND RESTRICTION**

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ABSTRACT

Host Factors Modulating HIV-1 Infectivity and Restriction

Retroviruses are obligate cellular parasites that depend on host cell factors for their replication. In particular, one such cellular factor, Cyclophilin A (CypA), promotes replication of Human Immunodeficiency Virus Type 1 (HIV-1) in human cells. CypA belongs to the family of peptidyl-prolyl isomerases and binds to an exposed proline-rich loop in the HIV-1 capsid protein (CA), as demonstrated by *in vitro* binding experiments. Although CypA has been conclusively demonstrated to facilitate HIV-1 spread in tissue culture, its role in HIV-1 replication remains unknown. Though CypA binds to mature HIV-1 CA, it is also incorporated into nascent HIV-1 virions via interactions with the CA domain of the Gag polyprotein. These findings raised the possibility that CypA might act at multiple steps of the retroviral life cycle. Disruption of the CypA incorporation into HIV-1 virions suggested that producer cell CypA was required for full virion infectivity. However, recent studies indicate that CypA within the target cell regulates HIV-1 infectivity by modulating retroviral restriction. In this study, we exploit multiple tools that disrupt the HIV-1 CA-CypA interaction to examine the relative contribution of producer cell CypA and target cell CypA to HIV-1 replication. Our results clearly demonstrate that target cell CypA, and not producer cell CypA, is important for HIV-1 CA-mediated function. Inhibition of HIV-1 infectivity resulting from virion production in the presence of CsA occurs independently of the CA-CypA interaction or even of CypA.

A recent discovery identified TRIM5 α as the factor mediating retroviral

restriction in cells from mammalian species. While in non-human primates, TRIM5 α potently blocks HIV-1, the human TRIM5 α orthologue specifically targets N-tropic Murine Leukemia Virus (N-MLV). Paradoxically, in cells from African green monkeys and rhesus macaques, CypA is required for TRIM5 α -mediated restriction of HIV-1. These observations suggest that human TRIM5 α also restricts HIV-1, but only when the association of CypA and HIV-1 CA is disrupted - in other words, HIV-1 recognition by the human TRIM5 α is precluded by CypA. In this study, effects of CypA and TRIM5 α on HIV-1 restriction were examined directly. RNA interference (RNAi) was used to show that endogenous human TRIM5 α does indeed restrict HIV-1, but the magnitude of this antiviral activity is not altered by disruption of the CA-CypA interaction or by elimination of CypA protein. Conversely, the stimulatory effect of CypA on HIV-1 infectivity was completely independent of human TRIM5 α . These data suggest that TRIM5 α and CypA independently regulate HIV-1 infectivity, and that in human cells CypA protects HIV-1 from an antiviral activity other than TRIM5 α . In addition, cross-saturation experiments suggest that both restrictions utilize a common saturable target that is epistatic to both TRIM5 α and the putative CypA-regulated restriction factor.

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