endogenous TRIM5α in HeLa cells but found no significant difference in the infectivities of the A92E and N121K mutants (data not shown). This suggests that TRIM5α does not play a role in the CypA-dependent restriction of A92E and N121K. Still, the possibility that potent, as-yet-unknown HIV-1-inhibitory factors, which associate with CypA, may be able to access this region cannot be ruled out (5).

Although CypA supports HIV-1 infection, some CA mutants showed CypA-dependent impairment of infection. Our data indicate that CypA impairs N121K infection, even in 293T and Jurkat cells that do not express CypA at very high levels. One of the proposed mechanisms for CypA-dependent restriction was that the high CypA expression level and mutation in CA modulate the stability of viral core (21). In our data, addition of CsA rescues the infection of N121K virus in 293T cells, but the infection efficiency of N121K does not exceed that of the WT virus infection even at the highest CsA concentration. It could be said that the infection of N121K might require CypA for its infection, and CypA facilitates the HIV-1 infection at different stage of replication. Indeed, CsA treatment reduced the infectivity of the WT virus in 293T cells, and restriction was observed at the early RT production step (data not shown). The stability of viral core alone cannot explain why both CypA-dependent infection and CypA-mediated inhibition are observed for the same virus. Therefore, we could assume that a potent HIV-1-inhibitory factor(s) is also involved in the CypA-dependent restriction and that each CA mutant has a different sensitivity for it. Thus, the N121K CA mutant could be a useful tool for analyzing the mechanism(s) underlying CypA-dependent restriction of HIV-1 infection. Understanding this mechanism may also help to determine the exact role played by CypA during HIV-1 replication.

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