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Galectin-1 Acts as a Soluble Host Factor That Promotes HIV-1 Infectivity through Stabilization of Virus Attachment to Host Cells

Michel Ouellet,* Simon Mercier,* Isabelle Pelletier,† Salim Bounou,* Jocelyn Roy,* Jun Hirabayashi,‡ Sachiko Sato,‡+ and Michel J. Tremblay2‡*

The establishment of HIV type 1 (HIV-1) infection is initiated by the stable attachment of the virion to the target cell surface. Although this process relies primarily upon interaction between virus-encoded gp120 and cell surface CD4, a number of distinct interactions influence binding of HIV-1 to host cells. In this study, we report that galectin-1, a dimeric β-galactoside-binding protein, promotes infection with R5, X4, and R5X4 variants. Galectin-1 acts as a soluble adhesion molecule by facilitating attachment of HIV-1 to the cell surface. This postulate is based on experiments where galectin-1 rendered HIV-1 particles more refractory to various agents that block HIV-1 adsorption and coreceptor binding (i.e., a blocking anti-CD4, soluble CD4, human anti-HIV-1 polyclonal Abs; stromal cell-derived factor-1 refractory to various agents that block HIV-1 adsorption and coreceptor binding (i.e., a blocking anti-CD4, soluble CD4, human anti-HIV-1 polyclonal Abs; stromal cell-derived factor-1). Experiments performed with the fusion inhibitor T-20 confirmed that galectin-1 is primarily affecting HIV-1 attachment. The relevance of the present findings for the pathogenesis of HIV-1 infection is provided by the fact that galectin-1 is abundantly expressed in the thymus and lymph nodes, organs that represent major reservoirs for HIV-1. Moreover, galectin-1 is secreted by activated CD8+ T lymphocytes, which are found in high numbers in HIV-1-positive patients. Therefore, it is proposed that galectin-1, which is released in an exocrine fashion at HIV-1 replication sites, can cross-link HIV-1 and target cells and promote a firmer adhesion of the virus to the cell surface, thereby augmenting the efficiency of the infection process. Overall, our findings suggest that galectin-1 might affect the pathogenesis of HIV-1 infection. *The Journal of Immunology, 2005, 174: 4120–4126.

Human immunodeficiency virus type 1 (HIV-1) has caused over 20 million deaths by now (1). HIV-1 infection leads to a relentless decline in both numbers and function of CD4+ T lymphocytes, resulting in the development of AIDS. The initial step of the virus life cycle requires attachment of virions to target cells. This event relies primarily upon interaction of the external envelope (Env) of gp120 subunit with cell surface CD4. Conformational changes of gp120 upon CD4 binding trigger interactions of Env with appropriate coreceptors, either the CC or CXC family of chemokine receptors (2). This binding to coreceptors exposes the Env gp41 transmembrane subunit and promotes fusion of viral and cellular membranes. Because the formation of a fusion pore results in injection of the capsid into the cytoplasm, the chemokine receptors constitute essential coreceptors for HIV-1 (3). Even though the interaction between gp120 and host CD4 plays a critical role in the attachment process, the use of CD4 as a viral receptor might have evolved subsequently to that of the chemokine coreceptors (4). In fact, in a physiological setting, the binding of virion-associated gp120 to cellular CD4 is often weak (5). Furthermore, most cell types that are permissive for HIV-1 infection express low levels of CD4 (5). It has thus been proposed that a number of distinct interactions may dominate and influence the process of HIV-1 attachment to target cells. This postulate is supported by previous data showing that various host adhesion molecules that are incorporated into mature HIV-1 particles can markedly increase virus infectivity (6–11).

Recently, some members of the mammalian galectin (β-galactoside-binding protein) family have been suggested to act as adhesion molecules due to their ability to mediate both cell-to-cell and cell-to-pathogen interactions (12–16). The capacity of galectins to act as adhesion molecules is attributable to their multivalent binding and cross-linking activities. Indeed, some galectins carry two glycan-binding domains (either intrinsically or as dimers), whereas others form dimers only upon binding to their glycoconjugate ligands. Consequently, a galectin molecule can cross-link ligands expressed on different constituents. For example, galectin-1 mediates the adhesion of T lymphoblastoid cells to thymic epithelial cells (17). Another member, galectin-3, can promote the binding of L-selectin-triggered lymphocytes to dendritic cells (18) and the adhesion of neutrophils to the endothelium (19). With respect to a possible effect of galectins on microbial pathogens, galectin-3 has been shown to affect binding of bacteria to host epithelial cells (19–21), whereas galectin-9 has been demonstrated to promote interaction between Leishmania major protozoan parasites and macrophages (22).

In the last few years, it has been reported that peripheral lymphatic organs, including spleen, lymph nodes, and mucosa-associated lymphoid tissues represent major sites of HIV-1 replication (23). Among the galectin family, galectin-1 and galectin-3 are expressed in various cell types including macrophages, dendritic...
cells, epithelial cells, and lymphocytes (14, 15, 24, 25), all of which are present in lymphoid organs. We thus investigated the potential modulatory effect of these two soluble adhesion molecules on HIV-1 replication in various experimental cell systems. Our data demonstrate that galectin-1 acts as a soluble HIV-1 binding protein that can stabilize virus-cell interactions and promote virus replication.

Materials and Methods

Cell lines and tonsilar histocultures

The LuSIV cell line is derived from the CEMx174 cell line, which stably expresses a luciferase reporter gene driven by the SV40 239 long terminal repeat (LTR) region (26). LuSIV cells are highly susceptible to HIV-1, HIV-2, and SIV infection, resulting in Tat-mediated expression of luciferase, which correlates with virus infectivity (26). Three cell lines were grown in RPMI 1640-based medium as previously published (27). IG5 cell line is a Jurkat derivative stably transfected with two HIV-1 LTR-driven luciferase reporter genes (28). PM1 is a clonal derivative of the HUT-78 cell line and is sensitive to R5- and X4-tropic strains of HIV-1 (29). PBMCs were purified from blood of healthy donors by Ficoll-Hypaque centrifugation. CD4 T lymphocytes were purified from PBMCs by magnetically depleting non-Th cells with CD4 T cell isolation kits (Miltenyi Biotech) and an AutoMACS apparatus (Miltenyi Biotec) according to the manufacturer’s instructions. PBMCs and purified CD4 T lymphocytes were maintained in RPMI 1640 medium containing 10% FCS, 1 μg/ml PHA-L (Sigma-Aldrich), and 50 U/ml recombinant human IL-2 (obtained through the National Institutes of Health AIDS Repository Reagent Program, Germantown, MD) for 3 days before HIV-1 infection. Human tonsil tissues removed during routine tonsillectomy and not required for clinical purposes were processed within 4 h of excision. The tonsils were washed thoroughly with medium containing antibiotics and then sectioned into small pieces of 6–9 mm3. These tissue blocks were placed on top of collagen sponge gels in the culture medium at the air-liquid interface as we previously described (27, 30).

Galectins

Recombinant human galectin-1 and galectin-3 were purified as described previously (22, 31). Purified galectin was passed through Detoxi-Gel endotoxin-removing gels (Pierce). The cross-linking activity of galectin-1 was tested weekly by performing an hemagglutination assay with concentrations similar to what was used in HIV-1 infection and attachment studies (i.e., 0.5–2 μM).

Virus stocks

Virus particles were prepared from the culture medium of human embryonic kidney 293T cells that were transiently transfected with the infectious molecular clone pNL4-3 (X4-tropic) as previously published (32). The NL4-3-Luc E’R’ vector was constructed by inserting a frameshift mutation near the env gene and inserting the firefly luciferase reporter gene into the nef gene (33). The pcDNA-1/Amp-based expression vector coding for the HIV-1 Ada-M (R5-tropic) full-length Env protein was generously provided by N. Landau (The Salk Institute for Biological Studies, La Jolla, CA). Briefly, 293T cells were seeded at 3 × 105 cells 16 h before transient transfection, which was conducted by adding either pNL4-3 or pNL4-3-Luc E’R’ and pcDNA-1/Ada-M to the cells as a calcium phosphate precipitate. Three days posttransfection, the virus-containing supernatant was filtered and frozen at −85°C until needed. Infectious virus particles were also prepared from the culture supernatant of PBMCs, infected for 4–10 days using either a X4-tropic laboratory strain (NL4-3) or a R5X4-tropic clinical isolate of HIV-1 (93US151). Titers of virus particles were normalized by content of the capsid protein p24 as determined by a sandwich ELISA (27).

Infection studies

All cell types tested were incubated with various concentrations of galectin-1 or galectin-3 (ranging from 0 to 2 μM) in the absence or presence of 50 mM lactose and then infected with HIV-1 (5 ng of p24 per 1 × 106 cells). Reporter cells (1 × 105/well) were incubated with HIV-1 for 24 h at 37°C before lysis, and luciferase activity was measured as described previously (34). PM1 cells were infected with luciferase-encoding Ada-M-pseudotyped viruses (10 ng of p24 per 1 × 106 cells) for 48 h prior to lysis and monitoring of luciferase activity. PBMCs (5 × 105/well) were infected with HIV-1 as described above and incubated at 37°C for 30 min before being washed extensively with cold PBS to remove unbound galectin-1 and virus. LuSIV cells were then incubated for 24 h at 37°C before lysis and measurement of the luciferase activity as previously described (34). For a direct evaluation of HIV-1 attachment to a more natural cellular reservoir, PBMCs were lysed immediately after incubation with HIV-1 for 0, 5, or 30 min, and viral attachment was estimated by measuring p24 levels.

Results

Galectin-1, but not galectin-3, promotes HIV-1 infectivity in reporter cell lines

It has been well established that galectins can act as adhesion molecules by cross-linking their ligands expressed on different cell types (14, 15, 24, 25). In fact, previous works have shown that galectins can mediate not only cell–cell interaction but also cell-pathogen adhesion (19–22), raising the possibility that galectins could also influence the biology of HIV-1 by promoting the attachment of the virus to its target cell. We thus used a sensitive HIV-1 infection assay system that is based on LuSIV reporter cells to investigate the possible effect of galectins on HIV-1 replication.

This assay allows the quantitative evaluation of single-cycle infection events through activation of integrated LTR sequences driving the luciferase reporter gene following the production of the viral protein Tat by de novo viral infection (26). LuSIV cells were first incubated for 10 min at 4°C with increasing concentrations of galectin-1 or galectin-3 (0–2 μM) in the presence or absence of lactose, a galectin antagonist. Cells were then infected with a prototypic X4-tropic laboratory strain of HIV-1 (i.e., NL4-3) that was produced upon transient transfection of 293T cells. As shown in Fig. 1A, virus infectivity was increased by galectin-1 in a dose-responsive manner up to 1 μM, after which the galectin-1-promoting effect appeared to reach a plateau, possibly due to the saturation of this assay. For example, infection with NL4-3 in the absence of galectin-1 resulted in a 68-fold increase in luciferase activity compared with the mock-infected cells (1,221 ± 223 vs 18 ± 1 relative light units (RLU)), whereas addition of galectin-1 at 1 μM resulted in a 24-fold augmentation of reporter gene activity compared with HIV-1 alone (29,118 ± 3,090 RLU). This galectin-1-dependent increase was significantly inhibited by lactose, suggesting an involvement of the carbohydrate binding domain of galectin-1. Lactose did not have any effect on HIV-1 replication in the absence of galectin-1 (HIV + lactose, 0 μM galectin-1). Interestingly, HIV-1 replication was unaffected when infection studies were performed with similar concentrations of galectin-3 (Fig. 1B), therefore indicating that members of the galectin family display distinct biological functions at least with...
Galectin-1 increases HIV-1 infection in an ex vivo lymphoid tissue model

It has been well established that secondary lymphoid tissue constitute a preferred anatomical site for active HIV-1 replication. To define whether the observed galectin-1-mediated increase of HIV-1 replication in cells cultured in vitro can also take place in a cellular microenvironment such as secondary lymphoid organs, we used an established ex vivo histoculture lymphoid tissue model that supports HIV-1 infection without exogenous activation (27, 30). Human lymphoid organ sections were prepared from tonsillar tissues and inoculated with HIV-1. As shown in Fig. 2C, galectin-1 increases virus production 3- to 4-fold compared with that seen in the absence of exogenous galectin-1 or in the presence of galectin-1 and lactose. These data suggest that galectin-1 can promote HIV-1 replication in lymphoid tissues, which are considered to be major reservoirs for HIV-1 as well as sites of progressive virus proliferation.

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Galectin-1 accelerates the kinetics of HIV-1 attachment to the surface of target cells

Given the reported capacity of galectin-1 to mediate cell-to-cell adhesion and its inability to induce HIV-1 LTR-mediated transcription, it can be proposed that galectin-1 favors the initial steps of the HIV-1 life cycle by increasing interactions between the virion and the cell surface. The possible up-regulating effect of galectin-1 on HIV-1 attachment to host cells was studied by first incubating LuSIV reporter cells with galectin-1 (1 μM) for 10 min at 4°C. HIV-1 was then added to the cell/galectin-1 mixture and incubated for 0, 5, or 30 min at 37°C. Cells were washed to remove unbound HIV-1 and/or galectin-1, then resuspended in fresh culture medium and further incubated for 24 h before assessing luciferase activity. As shown in Fig. 3A, a 30-min exposure of target cells to HIV-1 alone did not result in any significant reporter gene activity, indicating, as reported before, that gp120/CD4-mediated HIV-1 attachment is a slow process. Interestingly, a very brief exposure of the reporter cells to HIV-1 and galectin-1 at 4°C, the time period required to remove unbound HIV-1 and galectin-1 from the cell surface by a centrifugation step, was sufficient to induce a 9-fold increase in HIV-1 infectivity as measured by luciferase activity (Fig. 3A, compare control vs galectin-1 at time 0).

At longer exposure times (i.e., 5 and 30 min), HIV-1 infectivity was further increased by galectin-1 because fold increases of luciferase activity over untreated controls were 52 and 74, respectively. To confirm the capacity of galectin-1 to facilitate HIV-1 adsorption to the cell surface, a virus attachment assay was performed using freshly isolated PBMCs from healthy donors. Such cells were first pretreated or not with galectin-1 (1 μM) at 4°C for 10 min before being put in contact with HIV-1. The cell-virus-galectin-1 mixture was next incubated at 37°C for 0, 5, or 30 min before being extensively washed to eliminate unbound HIV-1 and galectin-1, and the amount of cell-bound virus was estimated by assessing p24. In the absence of galectin-1, only 0.75% of the initial virus input was found associated with PBMCs after 30 min of virus exposure at 37°C (Fig. 3B). In contrast, galectin-1 significantly promoted HIV-1 adsorption because >30% of the initial virus input is attached to PBMCs after the same exposure time. The binding property of galectin-1 is clearly illustrated by the observation that only a brief exposure of galectin-1-treated PBMCs to HIV-1 at 4°C (“time 0” in the graph, i.e., <2 min, which was required for separating the cells from unbound viruses and galectin-1) was sufficient to capture >4% of the initial viral input.

Galectin-1 reduced HIV-1 sensitivity to attachment inhibitors but not to an agent blocking fusion

We have previously reported that incorporation of host-encoded ICAM-1 rendered HIV-1 particles less susceptible to neutralization by human sera from seropositive patients (37). This increased resistance was associated with an enhancement of virus attachment to target cells provided by the additional interaction between virus-anchored ICAM-1 and cell surface LFA-1. Because galectin-1 seems to augment HIV-1 infectivity in a similar mode, we have tested the effectiveness of galectin-1 to alter sensitivity of HIV-1 to neutralization by agents that block the early stages of virus infection. When exposed to a concentration of 1 μg/ml SIM.2, a monoclonal anti-CD4 Ab that inhibits gp120 binding to CD4, HIV-1 replication was reduced by 90%, whereas addition of galectin-1 resulted in a 55% inhibition of viral infection only (Fig. 4A). Sensitivity of HIV-1 to neutralization by sCD4 was also altered by galectin-1. Indeed, a 0.1 μg/ml concentration of sCD4 inhibited HIV-1 infection by only 12% in the presence of galectin-1, whereas 78% of virus infectivity was blocked in its absence (Fig. 4B). The calculated IC50 for sCD4 is 28 ng/ml in the absence of galectin-1 compared with 470 ng/ml (a 16-fold increase) in the presence of the lectin. Both neutralizing agents achieved a complete inhibition of HIV-1 replication at higher concentrations even in the presence of galectin-1, confirming that gp120/CD4 interactions remain critical to allow a successful infection. Interestingly, binding and infection studies performed with the CXCR4+ CD4-deficient T cell line A2.01 indicate that, although attachment of virions to the cell surface is increased by 6-fold, there is still no productive HIV-1 infection (data not shown). Therefore, HIV-1 infection appeared to be still mediated by CD4 despite the presence of galectin-1.

HIV-1 infection in an individual often induces the production of specific antiviral Abs that can, in some instances, neutralize HIV-1. Thus, a neutralization experiment using a pool of purified IgG Abs from three HIV-1-infected individuals was also conducted. Virus infection was reduced by >50% when using 10 μg/ml the tested human polyclonal Abs (Fig. 4C). The antiviral efficacy of pooled human Abs was significantly reduced following addition of galectin-1. Together, these data suggest that the presence of galectin-1 limits also the effectiveness of neutralizing anti-HIV-1 Abs.

Chemokines that bind to HIV-1 coreceptors have been reported to have no effect on viral attachment but were shown to inhibit the
nations, and these results are representative of three different experiments.

The amounts of PBMC-associated virus were monitored by per-

D

FIGURE 3. HIV-1 attachment to host cells is promoted by galectin-1. A. LuSIV cells (1 × 10⁶) were either left untreated or treated with galectin-1 (1 μM) for 10 min at 4°C before being exposed to NL4-3 (5 ng of p24) for 0, 5, or 30 min at 37°C. Cells were then washed twice with 10 vol of cold PBS, resuspended in complete culture medium, and incubated at 37°C for 24 h. Luciferase activity was assessed following lysis of the cells. B. PBMCs (1 × 10⁶) were first incubated in the absence or presence of 1 μM galectin-1 for 10 min at 4°C before addition of NL4-3 (5 ng of p24). Cells were incubated at 37°C for 0, 5, or 30 min and were washed twice with 10 vol of cold PBS. The amounts of PBMC-associated virus were monitored by per-

foming a p24 assay. Data shown represent the means ± SD of four determin-

ations, and these results are representative of three different experiments.

following steps required for the formation of the fusion pore (38–40). These steps include coreceptor binding followed by confor-
mational changes of gp120 and, finally, exposure of gp41 and its fusion peptide. Treatment of LuSIV reporter cells with increasing concentrations of SDF1-α resulted in a dose-dependent inhibition of HIV-1 infection (Fig. 4D). Again, sensitivity of virions to the antiviral potency of SDF-1α was severely reduced in the presence of galectin-1, shifting the IC₅₀ value >400 ng/ml. Similar results were obtained with the CC chemokine RANTES upon infection of PM1 cells with luciferase-encoding HIV-1 particles pseudotyped with the R5-tropic Ada-M Env. Indeed, PM1 cells pretreated with galectin-1 were much less sensitive to the antiviral efficacy of RANTES than untreated cells (Fig. 4E). Collectively, these data suggest that galectin-1 displays the potency to increase replication of R5- and X4-tropic strains of HIV-1 through an up-regulatory effect on the first steps in the virus life cycle.

With the recent approval of the T-20 peptide (enfuvirtide) by the U.S. Food and Drug Administration, a new class of HIV-1 inhibitors has become available for the treatment of infected individu-

als. This drug targets fusion of the viral envelope with the cellular plasma membrane and thus acts at a stage that follows viral ad-

sorption to the host cell (41). In sharp contrast with data obtained

with the previous HIV-1 blocking agents, the percentages of inhibi-
tion with T-20 remained similar for cells either untreated or treated with galectin-1 (Fig. 4F). It should be noted that, even if percentages of inhibition are comparable between cells treated or not with galectin-1, addition of galectin-1 still induced a more robust infection of cells despite the presence of a high dose of T-20 (e.g., a mean luciferase activity of 135 RLU in the absence of galectin-1 and a mean luciferase activity of 9612 RLU in the presence of galectin-1 for cells treated with T-20 at 100 ng/ml).

Discussion

One of the most limiting steps in HIV-1 life cycle is the establish-

ment of a stable and firm association between the viral entity and its target cell. The initial contact is primarily established by mul-
tivalent interactions between virus gp120 and the host cell surface CD4 glycoprotein. Although biochemical kinetics analyses of the association between purified gp120 and CD4 suggest a tight inter-

action between these two molecules (42), it has been suggested that HIV-1 adhesion to host cells often occurs under suboptimal conditions in vivo (5). This is supported by our findings that <1% of the total virus input is stably associated with the target cells after 30 min of incubation under the tested in vitro conditions. Further-

more, during the course of HIV-1 pathogenesis, neutralizing Abs against HIV-1 are produced and CC chemokines, which bind HIV-1 coreceptors, are released (43). These factors can negatively interfere with the stable attachment of HIV-1 to target cells (43). Because gp120 from primary isolates of HIV-1 is biased toward a configuration that does not allow high-affinity binding to CD4, ligation of such viruses to the cell surface is expected to be ineffi-

cient (5). In addition, most cell types that are permissive for HIV-1 infection express little CD4 on their surface (5). Neverthe-

less, despite all of these restrictive factors that potentially com-

promise HIV-1 attachment to putative target cells, a chronic state of infection is established in humans. This suggests that several interactions between the viral entity and the cell surface in addition to the normal gp120-CD4 association are also taking place. For example, recognition of virion-anchored host adhesion molecules by their counterreceptors on target cells enhances/stabilizes the initial contact between HIV-1 and cells, resulting in an enhance-

ment of virus infectivity (5, 9). Such adhesion molecules are in-

corporated into nascent virions during the budding process. Be-

cause the expression of these adhesion molecules and/or their cognate ligands is frequently up-regulated during the normal im-

mune response to invading pathogens, it is expected that these additional interactions generally improve HIV-1 infectivity.

As for other host cell molecules, recent studies have revealed that β-galactoside-binding proteins, i.e., galectins, can act as sol-

vable adhesion-modulating molecules even if they are not intrinsi-

cally membrane associated (13–16, 25). In particular, it has been recently suggested that some galectins can act as leukocyte adhe-

sion molecules during leukocyte extravasation (13, 18). Galectins, which undergo exocrine release from neighboring cells, can cross-

link their surface ligands expressed either on cells or pathogens, thus resulting in direct mediation of cell-cell (13–16, 25) or cell-

pathogen interactions (19–22, 25).

The data presented here indicate that galectin-1 increases HIV-1 infectivity by virtue of its ability to promote a more efficient bind-
ing of mature virus particles to the target cell surface. These ob-

servations were made in various in vitro infection models, includ-
ing freshly isolated CD4⁺ T cells that were infected with a clinical strain of HIV-1. Furthermore, HIV-1 infection was also promoted by galectin-1 in human lymphoid tissue cultured ex vivo. Results from this experimental cell system are physiologically relevant considering that galectin-1 is secreted in lymph nodes, especially
by activated CD8\(^+\) T cells and epithelial cells (17, 44–46). Interestingly, one of the clinical features of HIV-1 infection is the apparition of an elevated number of activated CD8\(^+\) T cells. Furthermore, CD4\(^+\) T lymphocyte, which is considered to be one of the major cell types infected by HIV-1, expresses high levels of galectin-1 ligands such as CD43 and CD45 on its surface (45). It is noteworthy that levels of galectin-1 are especially high in the thymus and lymph nodes due to constant production by epithelial cells (I. Pelletier and S. Sato, unpublished observations; Refs. 17 and 46). Thus, it is tempting to speculate that elevated galectin-1 concentrations found in the thymus might result in an increased HIV-1 infection of thymocytes, thereby disrupting mature T cell development, leading to a faster progression toward AIDS, especially in children. Similarly, high levels of galectin-1 in lymph nodes could also promote viral replication in such organs known to harbor a high percentage of activated CD4\(^+\) T cells and further facilitate HIV-1-mediated destruction of lymph node architecture, resulting in an augmentation of the viral load in the periphery.

The galectin-1-mediated promoting effect on HIV-1 replication was not shared by galectin-3, therefore suggesting that members of the galectin family display certain specificity with respect to their cross-linking abilities. Nevertheless, the fact that galectin-3 at a high dose can abolish galectin-1-mediated enhancement of HIV-1 infection suggests that these two \(\beta\)-galactoside-binding proteins are sharing some common ligand(s). This last observation might reveal some physiological significance under natural conditions because galectin-1 and galectin-3 could eventually compete for similar binding sites. It is likely that endogenous galectin-1 can modulate HIV-1 attachment to CD4\(^+\) T lymphocytes in vivo because concentrations of galectin-1 in small blocks of tonsillar tissue were found to range between 10 and 20 \(\mu\)M (I. Pelletier and S. Sato, unpublished observations). On the opposite, a much lower concentration of galectin-3 was detected in such tissue (i.e., \(\sim 0.5 \mu\)M), thus suggesting that a possible competition between galectin-1 and galectin-3 is very unlikely.

Even though galectin-1 does not permit infection of CD4-negative cells, it can still favor a more efficient binding of HIV-1 particles to such cells. It can be proposed that galectin-1 might affect the capture of mature virions by cells such as dendritic cells, which are not considered as primary targets for HIV-1 but are proposed to play a critical role in the eventual transfer in trans of HIV-1 to more susceptible targets (i.e., CD4\(^+\) T lymphocytes). Appropriate studies are currently underway to address this issue.

The advent of antiretroviral therapy for the control of HIV-1 infection has significantly lengthened the life expectancy of HIV-1-infected persons. However, this therapeutic strategy has not resulted in a complete eradication of the virus as initially expected. Existing classes of antiretroviral drugs act primarily by blocking steps in the virus life cycle that occur inside target cells. More recently, novel therapeutic strategies are aimed at disrupting the initial interactions between the HIV-1 gp120 and its ligands on the target cell, i.e., CD4 and the appropriate chemokine receptor (47). Given that our data indicate that galectin-1 confers HIV-1 resistance to neutralization by various agents interfering with virus attachment, it is possible that endogenous galectin-1 can reduce the anti-HIV-1 activity of newly developed entry inhibitors. However, our observation that galectin-1 does not reduce the efficacy of T-20 toward HIV-1 replication is comforting and confirms that the mechanism by which galectin-1 is promoting virus replication is through an effect on the initial attachment step.

Recent data showing a direct involvement of the membrane lectin DC-SIGN in HIV-1 transmission (48) further underscore the cardinal role played by glycosylation events in HIV-1 pathogenesis. Of high clinical relevance to the present study is the recent work by Lanteri et al. (49) who have reported that virus infection results in altered glycosylation patterns favoring galectin-1 binding on both latently HIV-1-infected T cell lines and peripheral CD4\(^+\) and CD8\(^+\) T cells from AIDS patients. In conclusion, the additional interaction between the virion and the cell surface that is due to cross-linking properties of galectin-1 deserves to be studied in more detail, because a therapeutic modulation of the biological functions of this lectin might represent a novel strategy for the treatment of HIV-1 infection.
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Disclosures

The authors have no financial conflict of interest.

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