HIV-1 Protein Vpr Suppresses IL-12 Production from Human Monocytes by Enhancing Glucocorticoid Action: Potential Implications of Vpr Coactivator Activity for the Innate and Cellular Immunity Deficits Observed in HIV-1 Infection

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HIV-1 Protein Vpr Suppresses IL-12 Production from Human Monocytes by Enhancing Glucocorticoid Action: Potential Implications of Vpr Coactivator Activity for the Innate and Cellular Immunity Deficits Observed in HIV-1 Infection

Marco Mirani,* Ilia Elenkov,‡ Simona Volpi,† Naoki Hiroi,* George P. Chrousos,* and Tomoshige Kino1*

The HIV-1 protein Vpr has glucocorticoid receptor coactivator activity, potently increasing the sensitivity of glucocorticoid target tissues to cortisol. Patients with AIDS and normal cortisol secretion have manifestations compatible with glucocorticoid hypersensitivity of the immune system, such as suppression of innate and cellular immunities. The latter can be explained by glucocorticoid-induced inhibition of cytokine networks regulating innate and Th1-driven cellular immunity. We demonstrated that extracellularly administered Vpr protein dose-dependently potentiated glucocorticoid-induced suppression of both mRNA expression and secretion of IL-12 subunit p35 and IL-12 holo-protein, but not IL-12 subunit p40 or IL-10, by human monocytes/macrophages stimulated with LPS or heat-killed, formalin-fixed Staphylococcus aureus (Cowan strain 1). This effect was inhibited by the glucocorticoid receptor antagonist RU 486. Also, Vpr changed the expression of an additional five glucocorticoid-responsive genes in the same direction as dexamethasone and was active in potentiating the trans-activation, but not the trans-repression, properties of the glucocorticoid receptor on nuclear factor κB- or activating protein 1-regulated simple promoters. Thus, extracellular Vpr enhances the suppressive actions of the ligand-activated glucocorticoid receptor on IL-12 secretion by human monocytes/macrophages. Through this effect, Vpr may contribute to the suppression of innate and cellular immunities of HIV-1-infected individuals and AIDS patients. The Journal of Immunology, 2002, 169: 6361–6368.

Infection with HIV-1 leads to a decline in CD4 T cell numbers and an impaired cellular immune response, permitting developmental AIDS, through elimination of these cells by the viral infection (1–3). In parallel, the defect in innate and cellular immunities seen in HIV-1-infected patients is also characterized by suppression of the cytokytic activities of NK cells and cytotoxic T lymphocytes and by inhibition of T cell proliferation and of delayed-type hypersensitivity reactions (3). One important mechanism of this immune dysregulation is a selective impairment of monocytes/macrophages, dendritic cells, and Th1 lymphocytes and their cytokine networks (4, 5). Thus, the production of IL-12 and IFN-γ by NK and T cells, two cytokines that regulate and promote innate, cellular, and antiviral immunities, is decreased during HIV-1 infection (6).

Cytokines that are responsible for polarization of Th-type responses are of particular interest in AIDS (7). IL-12 is a central inducer of the Th1 response and cell-mediated immunity by stimulating the production of IFN-γ from NK and T cells and by inducing differentiation and proliferation of Th1-type cells (8). Thus, this cytokine, which is produced by APCs of the innate immune system, such as monocytes/macrophages and dendritic cells, appears to be critical for the promotion of cellular and, hence, antiviral immunities (9–11). IL-12 is a 75-kDa heterodimeric molecule composed of p35 and p40 subunits encoded by two separate genes (12–14). Both p35 and p40 are up-regulated in response to various stimuli, including intracellular bacteria or viruses, several endotoxins such as LPS, and intracellular proteins released by necrotic cells (14–19). IFN-γ, on the other hand, does not stimulate IL-12 production, but functions as a costimulatory signal, priming the cells to produce higher amounts of IL-12 after appropriate stimuli (20).

PBMC or whole blood cultures from HIV-infected individuals express marked deficiency in the production of both IL-12 p40 and p70 compared with cells from healthy individuals (6). This deficiency is observed across a broad panel of stimuli derived from infectious pathogens (21). The impairment of IL-12 production in HIV-1-infected individuals appears to be selective, since no variation in the secretion of TNF-α, IL-1β, or IL-10 has been found (22). The low production of IL-12 may be one of the critical factors for the impaired innate and cellular immunity in HIV-1-infected patients, since exogenous IL-12 is able to partially restore the depressed T cell proliferation, T and NK cell production of IL-2 and IFN-γ, and NK lytic activities in cells from HIV-1-infected individuals (23). The mechanism(s), however, underlying the reduced IL-12 production in HIV-1-infected patients remains poorly understood.

HIV-1 encodes a 96-aa, virion-associated accessory protein, Vpr, which has multiple, diverse functions (2, 24, 25). Vpr enhances the viral replication in monocyte- and lymphocyte-derived cell lines (26–28) and functions as a transcriptional activator of several viral promoters, including the HIV-1 long terminal repeat.

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promoter (29–32), and causes cell cycle arrest at the G2/M phase (33–35). Vpr may also contribute to nuclear translocation of the HIV-1 preintegration complex, possibly helping HIV-1 to efficiently infect nondividing cells, such as monocytes/macrophages and resting lymphocytes (36–38). Because Vpr is virion associated, it is delivered to the infected cells with the viral particle at an early phase of HIV-1 infection, affecting local infected lymphocytes, monocytes/macrophages, and dendritic cells (39). However, the same viral polypeptide is also expressed and secreted by infected cells of the host after successful integration of the HIV-1 provirus into the host genome (40). Indeed, this protein is detected in the extracellular fluids of HIV-1-infected patients, such as plasma and cerebrospinal fluid. Since extracellularly administered synthetic Vpr is bioactive, causing cell cycle arrest by penetrating the cell membrane, Vpr secreted into the extracellular space may exert its effects on proximal and distal cells and tissues, which are not infected by the virus (41).

We previously reported that Vpr dramatically enhances glucocorticoid receptor (GR)2 activity in several different cell lines, functioning as a potent coactivator of the GR, through a classic LXXLL coactivator signature motif (30). We further demonstrated that Vpr behaved as an adaptor molecule between promoter-bound transcription factors and p300/CBP coactivators in a glucocorticoid-responsive promoter (42). We also reported that nuclear receptor-responsive promoters employed the same set of coactivators and transcription-elongation complexes as the HIV-1 long terminal repeat, and that Vpr potentiated both types of promoter activities through the same mechanism, cooperating with host coactivators (43).

Glucocorticoids strongly inhibit IL-12 production from peripheral monocytes/macrophages, and this was suggested to be a major mechanism responsible for their selective suppression of Th1 functions and cellular immunity (44–46). Since Vpr functions as an enhancer of glucocorticoid activity, we hypothesized that this viral protein might contribute to the impaired IL-12 production seen in AIDS patients. We found that extracellularly administered Vpr suppressed both mRNA and protein production of IL-12, but not IL-10, from human peripheral monocytes. This viral peptide potentiated the effect of dexamethasone on IL-12 production, and this effect was inhibited by the glucocorticoid antagonist RU 486. The actions of Vpr and dexamethasone on the expression of other glucocorticoid-regulated mRNAs were similar. These results indicate that Vpr may inhibit IL-12 production by enhancing endogenous glucocorticoid action, potentially contributing to the innate and cellular immunity deficits of HIV-1-positive individuals and AIDS patients.

Materials and Methods

**Purification of recombinant Vpr**

Recombinant Vpr protein was produced using a baculovirus expression system. Vpr cDNA was amplified by PCR using primers 5′-ATGATAG GATCCATGGACACAGCCCGAAGACC-3′ and 5′-ACATGTAAGCT TCTAGGATCTACTGGCTC-3′ from the pCDNA3-Vpr that contains the wild-type Vpr sequence of the HIV-1 NL43 strain (30). The pFastBacHtB-Vpr was constructed by subcloning Vpr cDNA into BamHI and HindIII sites of pFastBacHtB (Life Technologies, Gaithersburg, MD) and expressed six-histidine-tagged Vpr protein in insect cells. Viruses that contain the His-Vpr sequence were isolated from DH10bac cells (Life Technologies) transformed by pFastBacHtB-Vpr. S9 cells were then infected with such viruses, and the cell lysates and culture media were collected to test for the expression of Vpr. A significant quantity of His-Vpr was found in the cell lysates, and subsequent purification was conducted using the cell pellets.

The method for the purification of recombinant His-Vpr is as follows. Frozen cell pellets were resuspended in lysis buffer (20 mM potassium phosphate (pH 7.4), 0.5 M NaCl, 5 mM imidazole, 1% Tween 20, 1 μg/ml aprotinin, 1 μg/ml leupeptin, and 1 mM PMSF) and sonicated to disrupt the cell membranes. The homogenates were centrifuged at 14,000 rpm for 15 min, and the supernatants were collected and loaded on a His-tag affinity column (Amersham Pharmacia Biotech, Piscataway, NJ). His-Vpr was eluted using elution buffer (20 mM potassium phosphate (pH 7.4), 0.5 M NaCl, 1 μg/ml aprotinin, 1 μg/ml leupeptin, 1 mM PMSF, and 200 mM or 400 mM imidazole) and was further concentrated using Centriprep YM-10 (nominal m.w. limit, 10,000) and YM-30 (nominal m.w. limit, 30,000; Millipore, Bedford, MA). The fraction filtered with YM-10 was used to dissolve the HIV-1 reverse transcriptase (Pol; AIDS Research and Reagent Program, National Institutes of Health, Bethesda, MD), which was used as a control for purified His-Vpr. In all procedures described above we used no steroids including glucocorticoids. A SilverXpress silver staining kit (Invitrogen, Carlsbad, CA) was used to visualize proteins run on the SDS-PAGE gel following the manufacturer's instructions, and gels were dried on paper filters. His-Vpr, separated on an SDS-PAGE gel, was transferred to the nitrocellulose membrane and blotted with anti-Vpr Ab (a gift from Dr. J. B. Kopp, National Institutes of Health) or anti-His Ab (Santa Cruz Biotechnology, Santa Cruz, CA).

**ELISA for cytokine measurements**

IL-12 p70, IL-12 p40, and IL-10 were measured by specific ELISA kits purchased from R&D Systems (Minneapolis, MN). The absorbance of each sample was measured with the microplate reader (model 550; Bio-Rad, Richmond, CA), and results were transformed to cytokine concentration (in picograms per milliliter) using a standard curve computed by Microplate Manager III Macintosh data analysis software (Bio-Rad).

**Elutriation and culture of monocytes from human donors**

Human monocytes were obtained from HIV-1-negative healthy donors. Mononuclear cells were isolated in lymphocyte separation medium, and the monocytes were purified by the counterflow centrifugal elutriation as reported previously (47). The monocytes were cultured at 5 × 10^5 cells/ml in RPMI 1640 medium supplemented with 10% FBS, 1% glutamine, and 50 μg/ml gentamicin to yield a final volume of 1 ml in 24-well plates. In a separate experiment dextran/charcoal-treated FBS was used instead of regular FBS. Cells were primed with 100 ng/ml of IFN-γ (R&D Systems) and cultured in the presence of increasing concentrations of Vpr in the absence or the presence of several concentrations of dexamethasone and/or RU 486, followed by stimulation with 1 μg/ml of LPS or 0.1% heat-killed, formalin-fixed *Staphylococcus aureus* (Cowan strain 1; SAC) in 5% CO2 at 37°C for 18 h. After incubation, the plates were centrifuged, and cell-free medium was collected and stored at −70°C for determination of p70 and p40.

![Figure 1](http://www.jimmunol.org/Downloaded.from/http://www.jimmunol.org)
p40 components of IL-12 and IL-10. To minimize variation of results from different donors, the experiments were repeated at least four times using peripheral monocytes from different individuals. The mean ± SE of LPS-induced levels of p70, p40, and IL-10 were 403 ± 16.9, 8956 ± 255, and 125.2 ± 5 pg/ml, respectively.

**Quantitation of glucocorticoid-regulated mRNAs by real-time PCR**

Human monocytes were cultured at 1.5 × 10^7 cells in the presence of 10 ng/ml of Vpr and/or 10^{-8} M dexamethasone, followed by stimulation with 1 μg/ml of LPS. Total RNA was isolated by using TRizol (Life Technologies, Gaithersburg, MD) according to the manufacturer’s instruction. The cDNAs were reverse transcribed with TagMan reverse transcription reagents (PE Applied Biosystems, Foster City, CA). mRNA levels of p35 and p40, and IL-10 were determined using TagMan cytokine gene expression plate 1 (PE Applied Biosystems) on the ABI PRISM 7700 sequence detection system, as previously reported. mRNA levels of human CD44, human Toll-like receptor 4, and indolamine 2,3-dioxigenase; the human macrophage receptor with collagenase structure and thrombospondin 1, which are negatively or positively regulated by glucocorticoids, respectively, were determined using primer sets as described previously (48).

**Transient transfection and reporter assay**

Human cervical carcinoma HeLa cells were kept in DMEM containing 10% FBS, 50 μg/ml of streptomycin, and 50 U/ml of penicillin. They were transfected with 1.0 μg/well of pCDNA3-Vpr together with 1.5 μg/ml of p(CAT)3 luciferase (Luc) or AP-1-dk81-Luc and 0.5 μg/ml of pSV40-β-Gal by the lipofectin (Life Technologies) method as previously described (30). pRSV-RelA (0.5 μg/well) or tetradecanoyl phorbol acetate (10^{-9} M) was cotransfected or added to examine NF-κB or AP-1 activities, respectively. To keep the same amount of DNA, Bluescript SK² and/or pCDNA3 were added. After transfection, the media were replaced with regular medium, and cells were cultured for an additional 24 h. Then, cells were stimulated with 1 × 10^{-8} M dexamethasone or vehicle, and cell lysates were collected for the luciferase and β-galactosidase assays after an additional 24 h. These enzyme activities were determined as previously described (49). pRSV-RelA, which expresses the p65 component of NF-κB, was obtained from the National Institutes of Health AIDS Research and Reference Reagent Program. pCDNA3-Vpr, which expresses wild-type Vpr, was described previously. (1x8B)-Luc, which contains three κB-responsive elements from the 1x8B promoter, was a gift from Dr. T. Fujita (Tokyo Metropolitan Institute of Medical Science, Tokyo, Japan). AP-1-dk81-Luc, which has four AP-1-responsive elements upstream of a proximal portion of the herpes simplex thymidine kinase promoter, was a gift from Dr. C. Bamberger (IHF, Institute for Hormone and Fertility Research, Hamburg, Germany). Mouse mammary tumor virus (MMTV) promoter-luciferase (pMMTV-Luc), which has the full-length MMTV promoter that contains four glucocorticoid-responsive elements (GREs), was provided by Dr. G. L. Hager (National Cancer Institute, Bethesda, MD). Bluescript SK², pCDNA3, and pSV40-β-galactosidase were purchased from Stratagene (La Jolla, CA), Invitrogen (Carlsbad, CA), and Promega (Madison, WI), respectively.

**Statistical analyses**

All data are presented as the mean ± SE. Statistical analyses were performed by one-way ANOVA, followed by Student’s t test after appropriate Bonferroni correction.

**Results**

Vpr suppresses the production of LPS-induced IL-12 p70, but not IL-12 p40 or IL-10, in human monocytes

Purified, insect cell-derived Vpr protein showed a single band with a Mr of 17 kDa detected with the silver-staining method on an (compared with baseline). B, Vpr suppressed the secretion of IL-12 p70 from the human peripheral monocytes stimulated with SAC in addition to LPS. Human peripheral monocytes were incubated with 10 ng/ml of Vpr with LPS or SAC. Bars represent the mean ± SE percentage of each result compared with the values obtained with LPS stimulation. ***, p < 0.01 (compared with baseline). C and D, Vpr did not change the secretion of IL-12 p40 (C) and IL-10 (D) by LPS-stimulated human peripheral monocytes. Human peripheral monocytes were incubated with increasing concentrations of Vpr or 10 ng/ml of Pol. Bars represent the mean ± SE percentage of each result compared with the values obtained with LPS stimulation.
SDS-PAGE gel (Fig. 1A). The protein band with the same Mr was detected with anti-Vpr (i) or anti-His (ii) Ab in a Western blot (Fig. 1B). The addition of increasing concentrations of His-Vpr resulted in a dose-dependent inhibition of IL-12 p70 production from LPS-stimulated human monocytes (Fig. 2A). Very low concentrations of Vpr, such as 0.1 ng/ml, already suppressed IL-12 production to 84% of baseline. The Vpr effect became more significant at concentrations of 1 and 10 ng/ml with a cytokine production of 73, and 65% of baseline, respectively. The same concentration of the control protein, HIV-1 Pol, dissolved in the void fluid from YM-10 concentration during Vpr purification, did not affect p70 production. To determine whether the effects of Vpr on IL-12 production were limited to LPS-stimulated IL-12, monocytes were treated with SAC instead of LPS and showed the same effect (Fig. 2B).

We also tested the effect of Vpr on the production of IL-12 p40 and IL-10 in the same assay system. Neither Vpr nor HIV-1 Pol affected the production of these molecules in LPS-stimulated monocytes (Fig. 2, C and D). Since we reported that Vpr enhances the glucocorticoid effect, functioning as a coactivator of the GR (30), we examined the effects of dexamethasone on the production of IL-12 p70 and p40, and IL-10 to compare with Vpr activity (Fig. 3, A–C). Dexamethasone at $10^{-8}$ M suppressed IL-12 p70 and p40, but not IL-10 production in LPS-treated monocytes, as we reported previously. It was equivalent to $10$ ng/ml of Vpr for the suppression of production of these cytokines. RU 486, a competitive antagonist for the GR, at a concentration of $10^{-6}$ M did not affect the production of any of the cytokines examined by itself, while it completely abolished the suppressive effect of dexamethasone on p70.

Vpr enhances the action of endogenous glucocorticoids and potentiates the dexamethasone effect on IL-12 production

To further investigate the hypothesis that Vpr acts by modulating GR activity, we tested RU 486 on the action of Vpr in the same experiment procedure. RU 486 at $10^{-6}$ M completely abolished the suppression of Vpr-induced p70 production, indicating that Vpr enhanced the pathway(s) regulated by the GR (Fig. 3D). Since it is likely that Vpr potentiated the glucocorticoid effect present in regular FBS, we tested the effects of Vpr on IL-12 p70 production in medium supplemented with dextran/charcoal-treated FBS. As expected, Vpr did not suppress p70, while it was fully active in the medium containing FBS (Fig. 3E).

We also tested the effects of Vpr in the presence of increasing concentrations of dexamethasone. Incubation of these cells with 1

![Graph](http://www.jimmunol.org/)
ng/ml of Vpr shifted the dexamethasone dose-response curve of p70 production to the left and downward, suggesting that it potentiated the effect of administered dexamethasone on p70 production. The Vpr effect was particularly significant at concentrations from $10^{-10}$ to $10^{-9}$ M dexamethasone, which are equivalent to physiologic concentrations of cortisol in humans (Fig. 3F).

Vpr regulates mRNA expression of glucocorticoid-responsive genes, including IL-12 p35, but not p40

We next examined the effects of Vpr on the mRNA expression of IL-12 p40 and p35, and IL-10 in human peripheral monocytes using TaqMan cytokine gene plate I. Expression of IL-12 p35 mRNA was suppressed by Vpr, while that of IL-12 p40 and IL-10 was not changed. In a parallel experiment, $10^{-8}$ M dexamethasone suppressed both IL-12 p35 and p40 mRNA expression, but did not change the abundance of IL-10 mRNA. Vpr and $10^{-8}$ M dexamethasone cooperatively suppressed IL-12 p35 mRNA levels (Fig. 4). We also measured the mRNA concentrations of CD44, Toll-like receptor 4, and indolamine 2,3-dioxygenase, all of which are strongly suppressed by glucocorticoids, and macrophage receptor with collagenous structure and thrombospondin 1, which are potently stimulated by these hormones. A concentration of 10 ng/ml of Vpr or $10^{-8}$ M dexamethasone changed the expression of all tested mRNAs in the same direction, suggesting that Vpr also affects the gene expression of other glucocorticoid-regulated genes (Fig. 5).

Vpr does not affect trans-repression of GR that is mediated by protein-protein interactions with NF-κB and AP-1

Vpr suppressed IL-12 p35 mRNA production, but not that of IL-12 p40, and it was active on both positively and negatively regulated genes by dexamethasone. Since GR has two major activities, GRE-mediated trans-activation of the glucocorticoid-responsive promoters and trans-repression of promoters regulated by other transcription factors through the protein-protein interactions (50), the above discrepancy might result from selective action of Vpr on these GR activities. We previously showed that Vpr enhanced trans-activation by the GR through its coactivator activity (30). Here we tested Vpr on GR-induced trans-repression of NF-κB- and AP-1-responsive promoters in transient transfection-based reporter assay systems. The IL-12 p40 promoter contains functional

**FIGURE 4.** Vpr suppressed the production of IL-12 p35 mRNA, but not IL-12 p40 and IL-10, in LPS-stimulated human peripheral monocytes. Peripheral human monocytes were incubated with vehicle (−), $10^{-8}$ M dexamethasone (Dex), or 10 ng/ml Vpr and stimulated with LPS. Total RNA was collected, and the mRNA abundance of indicated polypeptides was determined using the TaqMan PCR procedure. Bars represent the mean ± SE percentage of each result compared with the values obtained with LPS stimulation. **, p < 0.01 (compared with baseline).

**FIGURE 5.** Vpr and dexamethasone induced the same direction changes in the expression of glucocorticoid-regulated genes. LPS-stimulated human peripheral monocytes were incubated with vehicle (−), 10 ng/ml of Vpr, or $10^{-8}$ M dexamethasone (Dex). Total RNA was collected, and the mRNA abundance of the indicated polypeptides was determined using the TaqMan PCR procedure. Bars represent the mean ± SE percentage of each result compared with the values obtained with LPS stimulation. **, p < 0.01 (compared with baseline).
NF-κB- and AP-1-responsive elements, and glucocorticoid interference with the transcriptional activity of these factors may explain their suppressive effect of dexamethasone on p40 mRNA expression (51, 52). Interestingly, Vpr did not affect dexamethasone-suppressed NF-κB- or AP-1-responsive promoter activities, while it enhanced GR activity on the MMTV promoter, as previously reported (30) (Fig. 6). These results indicate that Vpr is effective primarily in GRE-mediated trans-activation activity, but it appears to have no effect on its trans-repressive actions on other transcription factors, such as NF-κB and AP-1.

**Discussion**

We demonstrated that extracellularly administered Vpr moderately suppressed IL-12 production, measured as the p70 dimeric molecule from IFN-γ-primed, LPS-stimulated human peripheral monocytes, suggesting downstream suppression of innate and Th1-type cellular immunity by this viral protein. Vpr, on the other hand, did not change the production of IL-10, which plays a critical role in cellular immunity by this viral protein. Vpr, on the other hand, did not change the production of IL-10, which plays a critical role in cellular immunity.

Steroid hormones from serum, completely abolished Vpr’s effect on IL-12 and IL-10 production and mRNA expression was similar, but not identical, with to of dexamethasone (44, 46). The GR antagonist, RU 486, or the use of dextran/charcoal-treated FBS, which strips glucocorticoid-responsive genes. These results strongly support the hypothesis that extracellular Vpr enters human cells and suppresses IL-12 production by enhancing ligand-bound GR activity.

Vpr enhanced the activity of glucocorticoids by suppressing IL-12 p35, but not p40, expression. Since the IL-12 p35 subunit is produced in far smaller amounts than the p40 subunit, HIV-1 may efficiently suppress IL-12 p70 production by down-regulating this rate-limiting component of IL-12 (18). While Vpr did not change mRNA or protein levels of IL-12 p40, however, dexamethasone did. The promoter of the IL-12 p40 subunit gene contains functional cis-acting sequences, including responsive elements for NF-κB and AP-1, which are key transcription factors in inflammation (20, 51–53). Since glucocorticoids induce strong inhibition of NF-κB and AP-1 through protein-protein interactions between ligand-activated GR and each of these transcription factors, it is likely that glucocorticoids suppress p40 production by inhibiting the activity of these factors (54).

The mechanism of p35 promoter suppression by glucocorticoids is not well understood. In humans there are two types of p35 transcripts, which are regulated by an upstream promoter characterized by a CpG island and a downstream promoter containing a TATA box (55). The former is active in B cell lines transformed with EBV, while the latter plays a major role in monocytes/macrophages. The latter promoter contains IFN-responsive elements that strongly influence p35 expression (56). The same promoter also contains several half-GREs and responsive elements for NF-κB, AP-1, Sp 1, and C/EBP; however, the functional significance of these sequences has not been determined as yet (56). Because our results indicate that Vpr does not affect the suppressive effect of GR on simple NF-κB- or AP-1-responsive promoters (Fig. 6), we concluded that Vpr might not generically disrupt the proper function of the transcriptional of IL-12 from monocytes/macrophages or dendritic cells at local sites of infection. Thus, Vpr may contribute to the inefficient activation of NK cells, blockade of Ag recognition, and deficient production of sensitized cytotoxic T lymphocytes with anti-viral specificity that characterize HIV-1-infected AIDS patients. Through these effects it could play an important role in the successful infection and proliferation of the HIV-1 virus. We also demonstrated that extracellular Vpr changed the expression of an additional five glucocorticoid-regulated genes in the same direction as dexamethasone. This result indicates that Vpr may regulate numerous glucocorticoid-responsive genes, regardless of actual infection of cells, through enhancing the trans-activation activity of the GR even at normal or low concentrations of circulating glucocorticoids. Indeed, AIDS patients have several manifestations seen in typical glucocorticoid excess states, such as...
myopathy, muscle wasting, dyslipidemia, and visceral obesity-related insulin resistance, in addition to immunosuppression similar to that induced by pharmacologic levels of glucocorticoids (57–64). It is possible that extracellular Vpr may contribute to the development of these pathologic states; thus, inhibition of Vpr co-activator activity by neutralizing Ab to Vpr or administration of RU 486 might help improve the clinical manifestations and disease course of AIDS patients.

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