

## MINIREVIEW

# Role of the Fas/Fas Ligand Apoptotic Pathway in Human Immunodeficiency Virus Type 1 Disease

DAVID KAPLAN\* AND SCOTT SIEG

*The Institute of Pathology, Case Western Reserve University, Cleveland, Ohio 44106-4943*

### APOPTOSIS AND THE DEPLETION OF CD4<sup>+</sup> T CELLS

Infection with human immunodeficiency virus type 1 (HIV-1) is characterized by the gradual and inexorable depletion of CD4<sup>+</sup> T lymphocytes. The pathogenetic importance of the loss of these cells is unquestioned since it correlates with the loss of immune capabilities and the consequent occurrence and severity of opportunistic infections and neoplasias. Thus, it is axiomatic that understanding the pathogenesis of HIV-1 disease requires an appreciation for the mechanisms involved in the loss of CD4<sup>+</sup> T cells.

In 1991, several groups of scientists proposed that apoptosis is important in the pathogenesis of infection with HIV-1 (2, 32, 60). Most cogently, Amieson and Capron hypothesized that the depletion of CD4<sup>+</sup> T lymphocytes in HIV-1 disease occurs by apoptosis secondary to inappropriate induction of activation-induced cell death (2). The incorporation of the concept of apoptosis into a reasonable conceptualization of HIV-1 infection was met with enthusiasm. The attractiveness of this model was due to its potential explanation of an organismal variable (the depletion of CD4<sup>+</sup> T cells in infected persons) in terms of a cellular mechanism (apoptosis). These scientists went on to propose the pharmacologic prevention of activation-induced apoptosis as a therapeutic modality for HIV-1-infected persons (2).

Soon after its publication, the hypothesis of inappropriate apoptosis in HIV-1 disease was supported by the findings of two independent groups (23, 39). They found that a greater proportion of T cells from HIV-1-infected persons died in culture than did T cells from healthy individuals. This greater propensity for apoptosis was observed for cells either left without any overt activation (spontaneous cell death) or for cells specifically activated (activation-induced cell death). Nevertheless, it should be noted that no correlation between these *in vitro* analyses and disease progression or virus levels has been subsequently found (40, 48).

### THE FAS/FAS LIGAND APOPTOTIC PATHWAY IN HIV-1 DISEASE

In 1995, the molecular mechanism of activation-induced T-cell death was determined to involve a pair of complementary molecules called Fas (also known as CD95 and APO-1) and Fas ligand (1, 12, 18, 24). Moreover, the ligation of Fas with Fas ligand was recognized as an important homeostatic control mechanism for maintaining appropriate numbers of T cells

(57). Thus, many investigators sought to test the possibility that the Fas/Fas ligand apoptotic pathway was accentuated in HIV-1-infected patients and thereby operative in depleting their CD4<sup>+</sup> T lymphocytes.

This suggestion appeared to be confirmed when several groups of scientists reported that the proportion of Fas-expressing T cells in HIV-1-infected persons increased with disease progression (5, 16, 22, 25, 52). Additionally, ligation of Fas with antibodies or with an active, soluble form of Fas ligand induced these cells to die by apoptosis (22, 25, 52). However, the implications of these findings in terms of CD4<sup>+</sup> T-cell depletion were not clear, since both CD4<sup>+</sup> and CD8<sup>+</sup> T cells demonstrated increased proportions of Fas expression and apoptosis upon ligation.

Further investigation concerning the possible involvement of the Fas/Fas ligand pathway in activation-induced cell death of T lymphocytes from HIV-1-infected persons has not produced a clear consensus. Some have found that activation-induced T-cell death is Fas dependent (10, 20), but others have not found any involvement of Fas and Fas ligand (26, 27). Recent data suggest that tumor necrosis factor (TNF)-related apoptosis-inducing ligand (TRAIL) but not Fas ligand mediates the apoptosis of activated T cells from HIV-1-infected persons (26).

The more important issue involves the possible role of the Fas/Fas ligand pathway in disease pathogenesis. However, the experiments reported thus far have not directly addressed this concern since the *in vivo* relevance of activating T cells *in vitro* has never been established. Moreover, there is no involvement of Fas and Fas ligand in the spontaneous apoptosis of peripheral blood cells from HIV-1-infected persons, which might reasonably be considered more reflective of *in vivo* events than activation-induced apoptosis (10, 27).

More relevant to the potential of Fas-dependent apoptosis acting *in vivo* was the finding that freshly isolated T lymphocytes from HIV-1-infected persons possessed Fas ligand message (10, 41, 50). Although this finding is important in considering the involvement of the Fas/Fas ligand pathway *in vivo*, these scientists did not observe spontaneous Fas ligand activity by blood cells in spite of the presence of the message. Consequently, the significance of the presence of the message in terms of HIV-1 disease pathogenesis is unclear.

While scientists were investigating Fas expression and activation-induced cell death, others discovered that *in vitro* exposure of monocytes or peripheral blood cells from healthy volunteers to HIV-1 enhanced Fas ligand mRNA and Fas ligand activity in these cells (7, 65). These data were used to support the contention that Fas ligand is involved in the depletion of T cells in HIV-1-infected persons. Additionally, simian immunodeficiency virus (SIV) infection of concanavalin A-stimulated peripheral blood mononuclear cells from healthy

\* Corresponding author. Mailing address: The Institute of Pathology, Case Western Reserve University, Biomedical Research Building, Room 926, 2109 Adelbert Rd., Cleveland, OH 44106-4943. Phone: (216) 368-1279. Fax: (216) 368-1300. E-mail: drk5@po.cwru.edu.

macaques enhanced Fas ligand expression, and this enhancement was related to expression of the Nef protein (68). However, the more direct demonstration that freshly isolated monocytes or other blood cells from the HIV-1-infected patients or from the SIV-infected macaques actually express cell surface Fas ligand protein and possess Fas ligand activity was not provided.

### VIRAL REGULATION OF APOPTOSIS

While the concept of apoptosis in HIV-1 disease was being promulgated, a different perspective on the role of programmed cell death in viral infections was developed. Virologists were finding that viruses actively regulate apoptosis in a complex way. Viruses require live, functional cells for their own replication. An indication of the importance of cellular viability for viruses is the prevalence of viruses that have been shown to inhibit physiological cell death upon infection (47, 59, 63, 64).

Although viruses require live cells to replicate, they also have the potential to kill cells through a variety of different mechanisms (47, 59). Viruses and viral replication are often deleterious to cells and consequently can enhance apoptosis. Thus, massive viral replication, as in syncytium formation induced by HIV-1, can result in cell death (36, 56). Also, viruses produce various proteins that are noxious such as (i) the HIV-1 Tat protein, which by itself has been shown to induce apoptosis in uninfected T lymphocytes (34, 46); (ii) HIV-1 gp120, which cross-links CD4 and thereby primes cells for apoptosis (9, 35); and (iii) HIV-1 Vpr, which has a cytostatic or cytotoxic effect on cells (6, 44, 54).

It may seem paradoxical and self-contradictory to suggest that the same virus can both prevent apoptosis and induce apoptosis. Perhaps it is most clearly understood by suggesting that viruses regulate apoptosis for their own purposes which relate to their own life cycles. Thus, viruses may prevent apoptosis early in infection before viral replication has proceeded and induce apoptosis late in infection as a means of releasing viral particles. Similarly, some viruses may induce death in cells responsible for immunity but prevent death in cells that are actively infected.

Many viruses have been found to both inhibit and enhance apoptosis, including adenovirus (3, 17, 37, 59, 61, 67), baculovirus (14, 15, 45), vaccinia virus (19, 28, 33), and poliovirus (62). It is clear that HIV-1 can also regulate physiological cell death in a complex way. Some of the molecular and cellular mechanisms that mediate the various effects on apoptosis have been investigated (Table 1), and pleomorphic effects of specific HIV-1 proteins on cell death have been found. For instance, although HIV-1 Tat can induce apoptosis (34, 38, 46), the protein also has the capacity to protect cells from dying (38, 69). Recently, HIV-1 Vpr has been shown to induce the physiological cell death of T lymphocytes in the absence of cellular activation, but the apoptosis of activated cells, which may be considered targets of viral infection, is inhibited by Vpr via I $\kappa$ B-mediated suppression of NF- $\kappa$ B (6). Understanding the complex regulation of apoptosis by viruses is key to understanding the pathogenesis of the diseases they cause.

### REGULATION OF APOPTOSIS IN HIV-1 DISEASE

Several findings in the last few years have suggested that the role of apoptosis in HIV-1 disease pathogenesis is more complex than is accounted for by simply proposing that the Fas/Fas ligand apoptotic pathway inappropriately induces the death of CD4<sup>+</sup> T cells. As with other viruses, HIV-1 most likely regulates apoptosis in an intricate way for the benefit of its own

TABLE 1. HIV-1 modulation of apoptosis

Viral protein involved	Mechanism	Reference(s)
<b>Inhibition</b>		
Tat	Increased Bcl-2 expression	38, 69
Vpr	I $\kappa$ B-mediated inhibition of NF- $\kappa$ B	6
p24/p17	Production of antagonistic epitopes that inhibit cytotoxic T lymphocytes	29
Not known	Inhibition of Fas ligand expression	50
<b>Enhancement</b>		
Tat	Increased activation of cyclin-dependent kinases	69
	Inhibition of manganese-dependent superoxide dismutase	66
	Enhanced Fas-dependent signal transduction	31
gp120/gp41	Syncytium formation	36, 56
	CD4 cross-linking	9, 35
	Cell death enhancement without hallmarks of apoptosis	31
Vpr	Not known	54

replication and transmission. The currently prevalent hypothesis that virus-induced, Fas/Fas ligand-dependent apoptosis is responsible for the depletion of CD4<sup>+</sup> T cells in HIV-1 disease does not take into account the varied and complex uses of apoptosis that other viruses have enlisted for their life cycles. It also does not adequately explain several relevant experimental results.

We have demonstrated that freshly isolated blood cells from HIV-1-infected patients have a deficiency in Fas ligand cell surface expression and activity compared to blood cells from uninfected, healthy volunteers (50). We tested these cells directly *ex vivo* for the presence of Fas ligand activity without any stimulation *in vitro*. Although this finding may seem paradoxical considering the presence of Fas ligand mRNA (10, 41, 50), we propose that there may be an active suppression of Fas ligand expression in HIV-1 disease as we have seen in cells infected with herpes simplex virus type 2 (51).

This suggestion is interesting in light of a recent finding indicating that cross-linking CD4 inhibits anti-CD3-induced apoptosis by inhibiting Fas ligand expression (43). Thus, virus-associated HIV-1 gp120 may be responsible for affecting Fas ligand surface levels. Another mechanism that could explain the decrease in Fas ligand expression is excessive enzymatic removal of the molecule from the cell surface. Soluble Fas ligand has been demonstrated to be elevated in the plasma of HIV-1-infected persons (8, 53). Moreover, recent findings have indicated that soluble Fas ligand inhibits the activity of the membrane-bound form of the molecule (55, 58).

Regardless of the mechanism for decreased membrane Fas ligand expression in HIV-1 disease, the suggestion that an inappropriate accentuation of the Fas/Fas ligand apoptotic pathway accounts for the depletion of CD4<sup>+</sup> T lymphocytes in HIV-1-infected persons is not likely. Our findings suggest that the Fas/Fas ligand pathway is suppressed by the viral infection in these patients (50).

Other observations also suggest that Fas-dependent apoptosis is not accentuated in HIV-1 disease. Patients preferentially accumulate T cells that express Fas and that are susceptible to killing by Fas ligand agonists (5, 16, 22, 25, 52). The accumulation of these cells suggests that they persist in infected persons instead of being depleted by apoptosis. Moreover, one group of investigators has shown that blood cells from HIV-1-

infected persons expressed high levels of Fas and Fas ligand mRNA, but they did not observe spontaneous cell death in these patient samples (41). Other groups observed spontaneous T-cell death from HIV-1-infected patients but failed to inhibit it by suppressing the Fas/Fas ligand pathway (10, 27). Consequently, there is no definitive indication that the Fas/Fas ligand apoptotic pathway is operative in HIV-1-infected patients.

Additional findings have suggested that HIV-1-mediated regulation of apoptosis is likely to have complex effects. Inhibition of apoptosis of cells infected in culture has been found to markedly enhance HIV-1 production and to facilitate persistent infection (4, 13, 49). In these experiments, inhibition of apoptosis was achieved by the constitutive expression of Bcl-2 (49) or adenovirus E1B 19K (4) via transfection and by the addition of inhibitors of the proapoptotic interleukin-1 $\beta$ -converting enzyme-like proteases (13). Thus, the inhibition of apoptosis in cells infected with HIV-1 has an enhancing effect on viral replication in culture. Moreover, we have shown that the reconstitution of Fas ligand activity in culture results in a marked decrease in virus production (50). These results suggest that the regulation of apoptosis in HIV-1-infected persons could have profound implications in terms of virus levels. Since virus levels are important for the HIV-1 life cycle, especially in terms of the likelihood of transmission, it seems reasonable to consider the possibility that the virus inhibits apoptosis of infected cells.

We hypothesize that HIV-1 regulates apoptosis in a way that is beneficial for itself in terms of the requirements of its own life cycle, especially those involving virus production and transmission. This regulation is likely to be complex since it involves both the inhibition and the induction of apoptosis. We propose that the complexity of apoptosis regulation in HIV-1 disease may be reflected in part by differential effects with respect to time so that newly infected cells are prevented from dying and with respect to different types of cells so that some uninfected cells—perhaps cells that have the potential to mediate an effective immune response against the virus—are induced to die.

The complex interaction of apoptosis and HIV-1 pathogenesis is also likely to be reflected by the various apoptosis-inducing molecules such as Fas ligand, TRAIL, and TNF, which have differential activities and implications with regard to HIV-1 production. For instance, it is known that TNF induces apoptosis as well as stimulates HIV-1 production, whereas Fas ligand induces apoptosis without the stimulation of concomitant viral replication (11, 30). The activity of TRAIL in terms of HIV-1 production is not yet known.

It is also important to consider apoptotic events in different anatomical locations. It is known that lymph nodes from HIV-1-infected persons demonstrate enhanced levels of apoptosis compared to lymph nodes from seronegative individuals and that the level of apoptosis is correlated with the activation status of the cells (21, 42). Moreover, in one study no correlation was found between apoptosis and either disease progression or viral burden (42). The molecular and cellular mechanisms of physiological cell death in lymph nodes from infected persons have not been investigated.

At the same time that HIV-1 manipulates apoptosis in its favor, the host uses various mechanisms of programmed cell death to counteract the virus. For instance, the cytotoxic immune response can be conceived of as a means to provide an exogenous trigger for the apoptosis of infected cells. In order to relate the organismal phenomena of CD4<sup>+</sup> T-cell depletion to the cellular mechanism of apoptosis, scientists must account for the many, different, independently regulated pathways of cell death. The capacity of infected cells to kill themselves in order to protect the organism from viral infection, the propen-

sity of immune cells to mediate immunity by cytotoxic mechanisms, the requirement of viruses to inhibit or at least delay apoptosis so that virus replication can occur, and the production of viral proteins that induce cell death combine to form a complex set of events that may together help to explain variations in organismal parameters such as fluctuations in CD4<sup>+</sup> T-lymphocyte levels as well as virus levels. Integration of these various components of cell death is most likely to produce a greater understanding of HIV-1 infection at an organismal level.

The regulation of apoptosis has profound implications for both the virus and the human host. It has the potential of significantly influencing virus levels and transmission as well as the host immune response and consequently health. A greater appreciation of the intricately choreographed regulation of apoptosis in HIV-1 disease pathogenesis will likely provide new opportunities in developing diagnostic, prognostic, and therapeutic modalities and thus may eventually help us advance the care of infected persons.

#### ACKNOWLEDGMENTS

We thank Neil Greenspan and Scott Vande Pol for helpful comments and Dawn Smith for participation in our experimental investigations.

#### REFERENCES

1. Alderson, M., T. Tough, T. Davis-Smith, S. Braddy, B. Falk, K. Schooley, R. Goodwin, C. Smith, F. Ramsdell, and D. Lynch. 1995. Fas ligand mediates activation-induced cell death in human T lymphocytes. *J. Exp. Med.* **181**:71–77.
2. Ameisen, J. C., and A. Capron. 1991. Cell dysfunction and depletion in AIDS: the programmed cell death hypothesis. *Immunol. Today* **12**:102–105.
3. Andersson, M., A. McMichael, and P. A. Peterson. 1987. Reduced allorecognition of adenovirus-2 infected cells. *J. Immunol.* **138**:3960–3966.
4. Antoni, B., P. Sabbatini, A. Rabson, and E. White. 1995. Inhibition of apoptosis in HIV-infected cells enhances virus production and facilitates persistent infection. *J. Virol.* **69**:2384–2392.
5. Aries, S. P., B. Schaaf, C. Muller, R. H. Dennin, and K. Dalhoff. 1995. Fas (CD95) expression on CD4<sup>+</sup> T cells from HIV-infected patients increases with disease progression. *J. Mol. Med.* **73**:591–593.
6. Ayyavoo, V., A. Mahboubi, S. Mahalingam, R. Ramalingam, S. Kudchodkar, W. V. Williams, D. R. Green, and D. B. Weiner. 1997. HIV-1 Vpr suppresses immune activation and apoptosis through regulation of nuclear factor kappa B. *Nat. Med.* **3**:1117–1123.
7. Badley, A., J. McElhinny, P. Leibson, D. Lynch, M. Alderson, and C. Paya. 1996. Upregulation of Fas ligand expression by HIV in human macrophages mediates apoptosis of uninfected T lymphocytes. *J. Virol.* **70**:199–206.
8. Bahr, G. M., A. Capron, J. Dewulf, S. Nagata, M. Tanaka, J. Bourez, and Y. Mouton. 1997. Elevated serum level of Fas ligand correlates with the asymptomatic stage of human immunodeficiency virus infection. *Blood* **90**:896–898.
9. Banda, N., J. Bernier, D. Kurahara, R. Kurrel, N. Haigwood, R. Sekaly, and T. Finkel. 1992. Crosslinking CD4 by HIV gp120 primes T cells for activation induced apoptosis. *J. Exp. Med.* **176**:1099–1106.
10. Baumler, C., T. Bohler, I. Herr, A. Benner, P. Krammer, and K. Debatin. 1996. Activation of the CD95 (APO-1/Fas) system in T cells from HIV-1-infected children. *Blood* **88**:1741–1746.
11. Biswas, P., G. Poll, J. Orenstein, and A. S. Fauci. 1994. Cytokine-mediated induction of human immunodeficiency virus type 1 (HIV-1) expression and cell death in chronically infected U1 cells: do TNF alpha and gamma interferon selectively kill HIV-1-infected cells? *J. Virol.* **68**:2598–2604.
12. Brunner, T., R. Mogil, D. LaFace, N. Yoo, A. Mahboubi, F. Echeverri, S. Martin, W. Force, D. Lynch, C. Ware, and D. Green. 1995. Cell-autonomous Fas/Fas-ligand interaction mediates activation-induced apoptosis in T-cell hybridomas. *Nature* **373**:441–444.
13. Chinnaiyan, A., C. Woffendin, V. Dixit, and G. Nabel. 1997. The inhibition of pro-apoptotic ICE-like proteases enhances HIV-1 replication. *Nat. Med.* **3**:333–337.
14. Clem, R. J., M. Fechheimer, and L. K. Miller. 1991. Prevention of apoptosis by a baculovirus gene during infection of insect cells. *Science* **254**:1388–1390.
15. Crook, N. E., R. J. Clem, and L. K. Miller. 1993. An apoptosis-inhibiting baculovirus gene with a zinc finger-like motif. *J. Virol.* **67**:2168–2174.
16. Debatin, K., A. Faissner, S. Stodt, W. Drez, A. Benner, and P. Krammer. 1994. High expression of APO-1 (CD95) on T lymphocytes from HIV-1-infected children. *Blood* **83**:3101–3103.
17. Debbas, M., and E. White. 1993. Wild-type p53 mediates apoptosis by E1A which is inhibited by E1B. *Genes Dev.* **7**:546–554.
18. Dhein, J., H. Walczak, C. Baumler, K. Debatin, and P. Krammer. 1995.

- Autocrine T-cell suicide mediated by APO-1(Fas/CD95). *Nature* **373**:438–441.
19. Dobbelsstein, M., and T. Shenk. 1996. Protection against apoptosis by the vaccinia virus SPI-2 (B13R) gene product. *J. Virol.* **70**:6479–6485.
  20. Estaquier, J., M. Tanaka, T. Suda, S. Nagata, P. Golstein, and J. Amies. 1996. Fas-mediated apoptosis of CD4+ and CD8+ T cells from HIV-1-infected persons: differential in vitro preventive effect of cytokines and protease antagonists. *Blood* **87**:4959–4966.
  21. Finkel, T., G. Tudor-Williams, N. Banda, M. Cotton, T. Curriel, C. Monks, T. Baba, R. Rubrecht, and A. Kupffer. 1995. Apoptosis occurs predominantly in bystander cells and not in productively infected cells of HIV- and SIV-infected lymph nodes. *Nat. Med.* **1**:129–134.
  22. Gehri, R., S. Hahn, M. Rothen, M. Steuerwald, R. Nuesch, and P. Erb. 1996. The Fas receptor in HIV infection: expression on peripheral blood lymphocytes and role in the depletion of T cells. *AIDS* **10**:9–16.
  23. Groux, H., G. Torpier, D. Monte, Y. Mouton, A. Capron, and J. C. Ameisen. 1992. Activation-induced death by apoptosis in CD4+ T cells from HIV-1-infected asymptomatic individuals. *J. Exp. Med.* **175**:331–340.
  24. Ju, S.-T., D. Panka, H. Cui, R. Ettinger, M. El-Khatib, D. Sherr, B. Stanger, and A. Marshak-Rothstein. 1995. Fas/FasL interactions required for programmed cell death after T-cell activation. *Nature* **373**:444–448.
  25. Katsikis, P., E. Wunderlich, C. Smith, L. A. Herzenberg, and L. A. Herzenberg. 1995. Fas antigen stimulation induces marked apoptosis of T lymphocytes in HIV-1 infected individuals. *J. Exp. Med.* **181**:2029–2036.
  26. Katsikis, P. D., M. E. Garcia-Ojeda, J. F. Terres-Roca, I. M. Tijoe, C. A. Smith, L. A. Herzenberg, and L. A. Herzenberg. 1997. Interleukin-1 $\beta$  converting enzyme-like protease involvement in Fas-induced and activation-induced peripheral blood T cell apoptosis in HIV-1 infection. TNF-related apoptosis-inducing ligand can mediate activation-induced T cell death in HIV-1 infection. *J. Exp. Med.* **186**:1365–1372.
  27. Katsikis, P. D., M. Garcia-Ojeda, E. S. Wunderlich, C. Smith, H. Yagita, K. Okumura, N. Kayagaki, M. Alderson, L. A. Herzenberg, and L. A. Herzenberg. 1996. Activation-induced peripheral blood T cell apoptosis is Fas independent in HIV-1-infected individuals. *Int. Immunol.* **8**:1311–1317.
  28. Kibler, K. V., T. Shors, K. B. Perkins, C. Zeman, M. Banaszak, J. Biesterfeldt, J. Langland, and B. Jacobs. 1997. Double-stranded RNA is a trigger for apoptosis in vaccinia virus-infected cells. *J. Virol.* **71**:1992–2003.
  29. Klenerman, P., S. Rowland-Jones, S. McAdam, J. Edwards, S. Daenke, D. Laloo, B. Koppe, W. Rosenberg, D. Boyd, A. Edwards, P. Giangrande, R. E. Phillips, and A. J. McMichael. 1994. Cytotoxic T-cell activity antagonized by naturally occurring HIV-1 Gag variants. *Nature* **369**:403–407.
  30. Kobayashi, N., Y. Hamamoto, N. Yamamoto, A. Ishii, M. Yonehara, and S. Yonehara. 1990. Anti-Fas monoclonal antibody is cytotoxic to human immunodeficiency virus-infected cells without augmenting viral replication. *Proc. Natl. Acad. Sci. USA* **87**:9620–9624.
  31. Kolesnitchenko, V., L. King, A. Riva, Y. Tani, S. Korsmeyer, and D. Cohen. 1997. A major human immunodeficiency virus type 1-initiated killing pathway distinct from apoptosis. *J. Virol.* **71**:9753–9763.
  32. Laurent-Crawford, A., B. Krust, S. Muller, Y. Riviere, M. Rey-Cuille, J. Bechet, L. Montagnier, and A. Hovanessian. 1991. The cytopathic effect of HIV is associated with apoptosis. *Virology* **185**:829–839.
  33. Lee, S. B., and M. Esteban. 1994. The interferon-induced double-stranded RNA-activated protein kinase induces apoptosis. *Virology* **199**:491–496.
  34. Li, C., D. Friedman, C. Wang, V. Metelev, and A. Pardee. 1995. Induction of apoptosis in uninfected lymphocytes by HIV-1 Tat protein. *Science* **268**:429–431.
  35. Lu, Y., Y. Koga, K. Tanaka, M. Sasaki, G. Kimura, and K. Nomoto. 1994. Apoptosis induced in CD4+ cells expressing gp160 of human immunodeficiency virus type 1. *J. Virol.* **68**:390–399.
  36. Maldarelli, F., H. Sato, E. Berthold, J. Orenstein, and M. Martin. 1995. Rapid induction of apoptosis by cell-to-cell transmission of human immunodeficiency virus type 1. *J. Virol.* **69**:6457–6465.
  37. Marcellus, R. C., J. G. Teodoro, T. Wu, D. Brough, G. Ketner, G. C. Shore, and P. E. Branton. 1996. Adenovirus type 5 early region 4 is responsible for E1A-induced p53-independent apoptosis. *J. Virol.* **70**:6207–6215.
  38. McCloskey, T. W., M. Ott, E. Tribble, S. Khan, S. Teichberg, M. O. Paul, S. Pahwa, E. Verdin, and N. Chirmule. 1997. Dual role of HIV Tat in regulation of apoptosis in T cells. *J. Immunol.* **158**:1014–1019.
  39. Meyaard, L., S. A. Otto, R. R. Jonker, M. J. Mijster, R. P. M. Keet, and F. Miedema. 1992. Programmed death of T cells in HIV-1 infection. *Science* **257**:217–219.
  40. Meyaard, L., S. A. Otto, I. P. M. Keet, M. T. L. Roos, and F. Miedema. 1994. Programmed death of T cells in HIV infection. No correlation with progression to disease. *J. Clin. Invest.* **93**:982–988.
  41. Mitra, D., M. Steiner, D. H. Lynch, L. Staiano-Coico, and J. Laurence. 1996. HIV-1 upregulates Fas ligand expression in CD4+ T cells in vitro and in vivo: association with Fas-mediated apoptosis and modulation by aurointricarboxylic acid. *Immunology* **87**:581–585.
  42. Muro-Cacho, C., G. Pantaleo, and A. Fauci. 1995. Analysis of apoptosis in lymph nodes of HIV-infected persons. Intensity of apoptosis correlates with the general state of activation of the lymphoid tissue and not with stage of disease of viral burden. *J. Immunol.* **154**:5555–5566.
  43. Oberg, H., R. Sanzenbacher, B. Lengel-Janssen, T. Dobmeyer, S. Flindt, O. Janssen, and D. Kabelitz. 1997. Ligation of cell surface CD4 inhibits activation-induced death of human T lymphocytes at the level of Fas ligand expression. *J. Immunol.* **159**:5742–5749.
  44. Planelles, V., F. Bachelier, J. Jowett, A. Haislip, Y. Xie, P. Banooni, T. Masuda, and I. S. Y. Chen. 1995. Fate of the human immunodeficiency virus type 1 provirus in infected cells: a role for Vpr. *J. Virol.* **69**:5883–5889.
  45. Prikhod'ko, E. A., and L. K. Miller. 1996. Induction of apoptosis by baculovirus transactivator IE1. *J. Virol.* **70**:7116–7124.
  46. Purvis, S., J. Jacobberger, M. Sramkoski, A. Patki, and M. Lederman. 1995. HIV type 1 Tat protein induces apoptosis and death in Jurkat cells. *AIDS Res. Hum. Retroviruses* **4**:443–450.
  47. Razvi, E. S., and R. M. Welsh. 1995. Apoptosis in viral infections. *Adv. Viral Res.* **45**:1–60.
  48. Rothen, M., S. Gratzl, H. H. Hirsch, and C. Moroni. 1997. Apoptosis in HIV-1-infected individuals is an early marker occurring independently of high viremia. *AIDS Res. Hum. Retroviruses* **13**:771–779.
  49. Sandstrom, P., D. Pardi, C. Goldsmith, D. Chengying, A. Diamond, and T. Folks. 1996. Bcl-2 expression facilitates human immunodeficiency virus type 1-mediated cytopathic effects during acute spreading infections. *J. Virol.* **70**:4617–4622.
  50. Sieg, S., D. Smith, Z. Yildirim, and D. Kaplan. 1997. Fas ligand deficiency in HIV disease. *Proc. Natl. Acad. Sci. USA* **94**:5860–5865.
  51. Sieg, S., Z. Yildirim, D. Smith, N. Kayagaki, H. Yagita, Y. Huang, and D. Kaplan. 1996. Herpes simplex virus type 2 inhibition of Fas ligand expression. *J. Virol.* **70**:8747–8751.
  52. Silvestris, F., P. Cafforio, M. Frassanito, M. Tucci, A. Romito, S. Nagata, and F. Dammacco. 1996. Overexpression of Fas antigen on T cells in advanced HIV-1 infection: differential ligation constantly induces apoptosis. *AIDS* **10**:131–141.
  53. Sloan, E., N. Young, P. Kumar, F. Weichold, T. Sato, and J. Maciejewski. 1997. Role of Fas ligand and receptor in the mechanism of T-cell depletion in acquired immunodeficiency syndrome: effect on CD4+ lymphocyte depletion and human immunodeficiency virus replication. *Blood* **89**:1357–1363.
  54. Stewart, S., B. Poon, J. Jowett, and I. Chen. 1997. Human immunodeficiency virus type 1 Vpr induces apoptosis following cell cycle arrest. *J. Virol.* **71**:5579–5592.
  55. Suda, T., H. Hashimoto, M. Tanaka, T. Ochi, and S. Nagata. 1997. Membrane Fas ligand kills human peripheral blood T lymphocytes, and soluble Fas ligand blocks the killing. *J. Exp. Med.* **186**:2045–2050.
  56. Sylwester, A., S. Murphy, D. Shutt, and D. Soll. 1997. HIV-1-induced T cell syncytia are self-perpetuating and the primary cause of T cell death in culture. *J. Immunol.* **158**:3996–4007.
  57. Takahashi, T., M. Tanaka, C. Brannan, N. Jenkins, N. Copeland, T. Suda, and S. Nagata. 1994. Generalized lymphoproliferative disease in mice, caused by a point mutation in the Fas ligand. *Cell* **76**:969–976.
  58. Tanaka, M., T. Itai, M. Adachi, and S. Nagata. 1998. Downregulation of Fas ligand by shedding. *Nat. Med.* **4**:31–36.
  59. Teodoro, J. G., and P. E. Branton. 1997. Regulation of apoptosis by viral gene products. *J. Virol.* **71**:1739–1746.
  60. Terai, C., R. S. Kornbluth, C. D. Pauza, D. D. Richman, and D. A. Carson. 1991. Apoptosis as a mechanism of cell death in cultured T lymphoblasts acutely infected with HIV-1. *J. Clin. Invest.* **87**:1710–1715.
  61. Tollefson, A., A. Scaria, T. Hermiston, J. Ryerse, L. Wold, and W. Wold. 1996. The adenovirus death protein (E3-11.6K) is required at very late stages of infection for efficient cell lysis and release of adenovirus from infected cells. *J. Virol.* **70**:2296–2306.
  62. Tolskaya, E., L. Romanova, M. Kolesnikova, T. Ivannikova, E. Smirnova, N. Raikhlin, and V. Agol. 1995. Apoptosis-inducing and apoptosis-preventing functions of poliovirus. *J. Virol.* **69**:1181–1189.
  63. Vaux, D. L., G. Haeccker, and A. Strasser. 1994. An evolutionary perspective on apoptosis. *Cell* **76**:777–779.
  64. Vaux, D. L., and A. Strasser. 1996. The molecular biology of apoptosis. *Proc. Natl. Acad. Sci. USA* **93**:2239–2344.
  65. Westendorp, M., R. Frank, C. Ochsenbauer, K. Stricker, J. Dhein, H. Walczak, G. Debatin, and P. Krammer. 1995. Sensitization of T cells to CD95-mediated apoptosis by HIV-1 Tat and gp120. *Nature* **375**:497–500.
  66. Westendorp, M., V. Shatrov, K. Schulze-Osthoff, R. Frank, M. Kraft, M. Los, P. Krammer, W. Droge, and V. Lehmann. 1995. HIV-1 Tat potentiates TNF-induced NF- $\kappa$ B activation and cytotoxicity by altering the cellular redox state. *EMBO J.* **14**:546–554.
  67. Wold, W. S. M., and L. R. Gooding. 1991. Region E3 of adenovirus: a cassette of genes involved in host immunosurveillance and virus-cell interactions. *Virology* **184**:1–8.
  68. Xu, X., G. Sreaton, F. Gotch, T. Dong, R. Tan, N. Almond, B. Walker, R. Stebbings, K. Kent, S. Nagata, J. Stott, and A. McMichael. 1997. Evasion of cytotoxic T lymphocyte (CTL) responses by Nef-dependent induction of Fas ligand (CD95L) expression on simian immunodeficiency virus-infected cells. *J. Exp. Med.* **186**:7–16.
  69. Zauli, G., D. Gibellini, D. Milani, M. Mazzoni, P. Borgatti, M. La Placa, and S. Capitani. 1993. HIV-1 Tat protein protects lymphoid, epithelial, and neuronal cell lines from death by apoptosis. *Cancer Res.* **53**:4481–4485.