Oxidative stress has been proposed to be involved in the immunologic defect observed in effector cells of the immune system as well as in lymphocyte cell death and viral replication in human immunodeficiency virus (HIV)-infected patients. Because thiol-containing antioxidants such as N-acetylcysteine have been shown to have beneficial effects on CD4+ lymphocyte survival and to inhibit programmed cell death and HIV-1 replication, they may play a role in therapeutic strategies of this disease. In this work we have studied the cellular thiol levels and the effect of in vitro antioxidant treatment of purified CD4+ lymphocytes from HIV-infected patients, and correlated these parameters to proliferative responses and programmed cell death. We show that CD4+ lymphocytes from HIV-infected patients display impaired proliferative responses and a significant decrease in cellular thiol levels, indicating a disturbed redox status. Interestingly, antioxidant treatment succeeded to restore defective proliferative responses to CD3-mediated activation in 8 of 11 patients (high antioxidant responders). In contrast to high responders, patients failing to respond to antioxidant treatment (low antioxidant responders), were characterized by an abnormal ratio of apoptotic cells, which was not affected by N-acetylcysteine and/or 2-mercaptoethanol preincubation. These results demonstrate for the first time that antioxidant treatment is able to revert the impaired proliferative activity of CD4 cells from HIV-infected patients and could help designing therapeutic strategies with antioxidant drugs. However, this action is not observed in cells undergoing programmed cell death.

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Materials and Methods

Patients and controls. PB samples were obtained from 11 asymptomatic HIV-1-infected patients with CD4 counts >300/μL (mean = 395/μL) followed up at the Hospital of the Pasteur Institute. Seropositive status was determined by the presence of serum antibodies to HIV-1 in two separate serum samples as determined by specific enzyme-linked immunosorbent assay (Elavia I; Diagnostics Pasteur, Paris, France) and confirmed by Western blot analysis (Lavblot; Diagnostic Pasteur). Controls were 10 sex- and age-matched HIV-1 seronegative healthy volunteers.

Antibodies. Anti-CD16 (3G8, IgG1) and anti-CD56 (T199, IgG1) monoclonal antibodies (MoAbs) used for panning were purchased from Jackson ImmunoResearch (Marseille, France). Murine anti-CD19 (HD37, IgG1), anti-CD8 (DK25, IgG1), and anti-CD14 (TUK4, IgG2a) MoAbs also used for panning, and anti-CD3 MoAb (UCHT1, IgG1) used for lymphocyte stimulation were purchased from Dako (Dokkops AS, Denmark).

Isolation of CD4 T lymphocytes. Purification of CD4 T lymphocytes was proceeded as previously reported. Briefly, PB lymphocytes (PBL) isolated in a Ficoll gradient, were incubated in the presence of anti-CD19, CD8, CD16, CD56, and CD14 antibodies followed by incubation in the presence of sheep-antimouse-coated immunomagnetic beads (Dynabeads, Dynal, Norway) to achieve a 5:1 bead-to-cell ratio. After incubation for 60 minutes at 4°C, cell
fractons were separated by the application of a magnet to the outside of the tube for 3 minutes; unbound cells, enriched for CD4 T cells, were carefully aspirated and a second round of immunomagnetic separation was performed for an additional period of 60 minutes at 4°C with a bead-to-cell ratio of 10:1. Purity of subsets was evaluated at the end of the enrichment procedures. In all experiments, purity of cell preparations was greater than 90% for CD3+CD4+ cells, as determined by immunofluorescence analysis using an EPICS 752 flow cytometer (Coulter, Hialeah, FL).

Antioxidant preincubation of CD4+ lymphocytes. Addition of NAC (Sigma, St. Louis, MO) was made from freshly prepared stock solutions in RPMI-1640 titrated to pH 7.2 with NaOH. For antioxidant treatment, cells were resuspended at 5 × 10^6/mL in serum-free RPMI-1640 and incubated with NAC (20 mM/L) and 2-ME (50 μM/L; Sigma) for 2 hours at 37°C. Cells were then washed twice to remove excess of antioxidants and finally resuspended in 10% fetal calf serum (FCS) RPMI-1640.

Cell proliferation assays. For 3H-thymidine uptake, cells were cultivated in 96-well culture plates at 5 × 10^4 cells per well in a total volume of 200 μL of supplemented RPMI-1640 culture medium (10% human pooled AB serum [Jacques Boy, Paris, France], glutamine 4 mM/L, HEPES 20 mM/L, sodium pyruvate 10 mM/L, and penicillin/streptomycin). Stimulation was performed by the addition of immunomagnetic beads precoated with saturating doses of mouse-antihuman CD3 IgG MoAb at a bead-to-cell ratio of 5:1. After 3 days, cultures were pulsed with 1 μCi of [3H]-thymidine (CEA, Paris, France) during the last 18 hours of culture and then procured with a cell harvester system (Tomtec Inc, Orange, CT) and radioactivity was measured in a micro beta plate scintillation counter (LKB). All cultures were performed in quadruplicate, and for each experiment a control using only culture medium was performed.

Flow cytometry analysis. Flow cytometry analysis was performed on an EPICS 752 fluorescent activated cell sorter (Coulter Corp) as previously reported.4 To standardize the assays and to correct for day-to-day variations in instrument performance, the mean fluorescence of calibration beads (Standard-Brite; Coulter Corp) was used to adjust reference fluorescence. A total of at least 10,000 cells were analyzed for each sample and data were stored in a histogram mode.

Staining for intracellular low-molecular-weight thiols. Intracellular thiol levels were estimated by flow cytometry using the 5-chloromethyl fluorescein diacetate (CMF) probe.19 This nonfluorescent probe has been shown to form fluorescent adducts with intracellular thiol antioxidants; GSH. Levels of cellular thiols were monitored by flow cytometry and values expressed as the mean log of the relative fluorescence intensity. As depicted in Table 2, resting CD4 T cells from HIV+ individuals showed a decrease of about 15% in mean values of log CMF fluorescence with respect to CD4+ lymphocytes from healthy volunteers. As depicted in Table 2, resting CD4 T cells from HIV+ individuals showed a significant decrease in mean values of log CMF fluorescence with respect to CD4+ lymphocytes from healthy volunteers (Wilcoxon-Mann-Whitney rank test P < .05). However, no statistical correlation between LMT levels and proliferative responses in the HIV-infected group was found.

RESULTS

Defective proliferative responses of purified CD4 cells from HIV-infected patients. As previously reported, the proliferative responses of CD4 T cells to CD3-mediated from HIV-infected patients were defective and ranged from about 10% to 90% of those observed in normal CD4 T cells (Table 1, left panel).

Basal intracellular low molecular thiol levels in CD4+ lymphocytes from HIV-infected patients. In a first series of experiments we have measured the thiol levels in purified CD4 T cells from 11 HIV-infected patients and 6 healthy volunteers. For this, we used the chloromethyl-fluorescein probe, which forms fluorescent adducts with low-molecular-weight thiols, including GSH. Levels of cellular thiols were monitored by flow cytometry and values expressed as the mean log of the relative fluorescence intensity.
Table 1. Values of [3H]-Thymidine Uptake of Purified CD4+ Lymphocytes From HIV-Infected Patients and Healthy Volunteers

<table>
<thead>
<tr>
<th>Culture Media</th>
<th>Antioxidant Treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Controls</td>
<td>CD3 MoAb</td>
</tr>
<tr>
<td>HIV-infected patients (HAR)</td>
<td></td>
</tr>
<tr>
<td>HIV-01</td>
<td>0.14</td>
</tr>
<tr>
<td>HIV-02</td>
<td>0.12</td>
</tr>
<tr>
<td>HIV-03</td>
<td>0.13</td>
</tr>
<tr>
<td>HIV-04</td>
<td>0.11</td>
</tr>
<tr>
<td>HIV-05</td>
<td>0.17</td>
</tr>
<tr>
<td>HIV-06</td>
<td>0.15</td>
</tr>
<tr>
<td>HIV-07</td>
<td>0.16</td>
</tr>
<tr>
<td>HIV-08</td>
<td>0.19</td>
</tr>
<tr>
<td>HIV-09</td>
<td>0.12</td>
</tr>
<tr>
<td>HIV-10</td>
<td>0.12</td>
</tr>
<tr>
<td>HIV-11</td>
<td>0.13</td>
</tr>
</tbody>
</table>

Healthy volunteers (n = 6) mean ± SD

<p>| | | | | |</p>
<table>
<thead>
<tr>
<th></th>
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<th></th>
<th></th>
<th></th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Mean</td>
<td>SD</td>
<td>Mean</td>
<td>SD</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Controls</td>
<td>0.12 ± .05</td>
<td>50.4 ± 5.7</td>
<td>0.22 ± 0.04</td>
<td>73.8 ± 7.4</td>
</tr>
</tbody>
</table>

Values of thymidine uptake are depicted as cpm × 10^3 and were performed after 72 hours of culture.

cells when compared with untreated cells (1.5-fold increase). Preincubation with either NAC or 2-ME alone increased proliferative responses in a similar ratio for normal CD4 T cells when compared with untreated cells, although the best results were observed with the association of NAC and ME (data not shown). According to thymidine uptake increase in CD4+ lymphocytes following antioxidant treatment (Table 1, right panel), HIV-infected patients could be clearly segregated into two groups: "low antioxidant responders" (LAR, 3 of 11) and "high antioxidant responders" (HAR, 8 of 11). In the HAR group, antioxidant treatment significantly enhanced proliferative responses to approach values close to those achieved with normal CD4 T cells after antioxidant treatment. These results indicate that impaired proliferative responses observed in CD4+ lymphocytes during asymptomatic HIV-1 infection could be restored in a significant proportion of patients by antioxidants. However, as shown in Table 2, a minor fraction of HIV-infected patients studied (LAR, 3 of 11), in whom proliferative responses were strongly decreased, failed upon antioxidant treatment to significantly restore these responses.

Evaluation of culture- and activation-induced changes in cellular thiols related to proliferative responses of CD4+ lymphocytes. To evaluate whether the responses of CD4 T cells to antioxidant treatment in both groups resulted from changes in the cellular redox status, we examined thiol levels after antioxidant treatment in nonactivated and in CD3-activated cells. Results from representative individuals are depicted in Fig 2. It is shown that in vitro antioxidant treatment for 2 hours at 37°C succeeded to enhance intracellular thiol

Table 2. Low-Molecular-Weight Thiol Contents in Resting CD4+ Lymphocytes

<table>
<thead>
<tr>
<th></th>
<th>Healthy Volunteers</th>
<th>HIV-infected Patients</th>
</tr>
</thead>
<tbody>
<tr>
<td>HIV-01</td>
<td>158.2</td>
<td>158.2</td>
</tr>
<tr>
<td>HIV-02</td>
<td>157.8</td>
<td>157.8</td>
</tr>
<tr>
<td>HIV-03</td>
<td>143.9</td>
<td>143.9</td>
</tr>
<tr>
<td>HIV-04</td>
<td>153.7</td>
<td>153.7</td>
</tr>
<tr>
<td>HIV-05</td>
<td>154.5</td>
<td>154.5</td>
</tr>
<tr>
<td>HIV-06</td>
<td>147.9</td>
<td>147.9</td>
</tr>
<tr>
<td>HIV-07</td>
<td>153.1</td>
<td>153.1</td>
</tr>
<tr>
<td>HIV-08</td>
<td>145.8</td>
<td>145.8</td>
</tr>
<tr>
<td>HIV-09</td>
<td>154.8</td>
<td>154.8</td>
</tr>
<tr>
<td>HIV-10</td>
<td>150.2</td>
<td>150.2</td>
</tr>
<tr>
<td>HIV-11</td>
<td>133.5</td>
<td>133.5</td>
</tr>
</tbody>
</table>

Thiol levels were estimated by flow cytometry using the CMF probe. Values are expressed as the mean log of relative fluorescence intensity.
contents in resting lymphocytes from both healthy volunteers and HIV-infected individuals including HAR and LAR (Fig 2, Basal). However, it is important to keep in mind that the method we used is unable to determine if thiol replenishment results as a consequence of enhanced intracellular GSH, cystein, or SH containing NAC. In addition, cysteine accumulation rather than GSH after NAC replenishment, could result from impaired γ-glutamyl-cysteinesynthetase, which cannot be excluded to occur in lymphocytes from HIV-infected individuals. These results indicate that the different susceptibility of high and low responder groups of patients to recover impaired proliferative responses, is not dependent on restoration of basal lymphocyte thiol levels. However, when compared with normals and HAR, cells from LAR failed to maintain adequate intracellular thiol levels (Fig 2) after overnight cultures irrespectively of antioxidant treatment or CD3-mediated activation (more than twofold reduction). These results suggest that inability of antioxidant treatment to restore in vitro proliferative responses in CD4 T cells from LAR could be the consequence of an impairment in cellular mechanisms aimed to maintain adequate intracellular thiol levels needed for normal cellular physiology. However, the possibility of an in vivo–triggered process unrelated to redox status cannot be excluded.

Antioxidant treatment restored the proliferative responses of unresponsive CD4+ T cells but failed to affect cells undergoing PCD. PCD has been proposed to play an important role in CD4 depletion during HIV infection and there is growing evidence indicating that oxidative stress may be involved in this process. Thus, we have speculated that in vitro–enhanced proliferative responses to antioxidant preincubation in a fraction of HIV-infected individuals could be the consequence of PCD inhibition. The ratio of apoptotic cells was evaluated by fluorescence-activated cell sorting (FACS) using the merocyanine 540 fluorescent probe which binds specifically to membrane of apoptotic cells. These results were obtained for 5 of the 11 patients (HIV-06, 07, 08 from the HAR group and 09 and 10 from the LAR group). The percentage of apoptotic cells was, respectively, 5%, 7%, 8%, 25%, and 13% before stimulation and 16%, 17%, 19%, 66%, and 32% after 24 hours in culture. Because results were similar in the presence or the absence of NAC + 2-ME or in the absence or presence of anti-CD3 stimulation, these results correspond to those obtained in the presence of NAC + 2-ME and in the absence of CD3 stimulation. Figure 3 depicts the differences between extreme cases in the HAR and LAR groups. Although HIV-09 patients may represent an extreme case, because in our hands it is unusual to find 25% of cells in spontaneous apoptosis, it clearly shows that the inability to display proliferative response may result from the high number of dead cells to incorporate thymidine. On the contrary, in patients from the HAR group, cell death was not significantly different from normals (<15%), at least during the first 24 hours of culture (Fig 3). Of note, in contrast to Gougeon and Montagnier and Groux et al, who reported an increased activation-induced cell death following superantigen, calcium ionophore and soluble CD3 stimulation, in the experimental conditions used in this work, CD3-mediated stimulation of purified CD4 T cells was not associated with enhanced cell death, but with a moderate decrease in the fraction of merocyanine high (apoptotic) cells when compared with nonactivated cells. These apparently opposed results are probably accounted by differences in experimental conditions, since in a previous work we failed to substantiate increase in activation-induced cell death after stimulation with immobilized CD3 of purified CD4 T cells. Moreover, as in our experimental conditions we are unable to induce activation-mediated cell death, we cannot exclude the possibility that NAC treatment could have an effect in inhibition of this phenomenon. Taken together, these results...
showed that antioxidant treatment was able to restore proliferative responses in a high proportion of asymptomatic HIV-infected individuals, without a significant effect on the ratio of apoptotic cells. Indeed, recovery of the proliferative responses by antioxidant treatment was not mediated by inhibition of PCD, but rather by recovery of the impaired proliferative ability of nonapoptotic lymphocytes. However, taking into account that our work was performed with isolated CD4 lymphocytes, in vitro antioxidant treatment was not able to inhibit cell death and, therefore, not able to significantly influence proliferative responses.

**DISCUSSION**

Because recent reports suggested that oxidative stress and GSH depletion may play an important role in HIV-induced immunopathogenesis, we speculated that an imbalance in the cellular redox status might account, at least partially, for impaired proliferative responses of CD4 lymphocytes from HIV-infected individuals. In agreement with these reports, we have observed decreased levels of intracellular low molecular thiol in resting CD4 T cells from asymptomatic HIV-infected individuals, suggesting the possibility of an in vivo oxidative stress. In this respect, recent reports demonstrated that T lymphocytes from HIV-infected patients were characterized by a substantial decrease in reduced glutathion, in the ratio of reduced to total glutathion and a considerable increase in oxidized glutathion.

To assess the effects of replenishing cellular thiol contents on impaired proliferative responses, cells were preincubated with the thiol-containing antioxidant NAC plus 2-ME. In our experimental conditions, CD4 lymphocytes from a majority of asymptomatic HIV-infected individuals (8 of 11) restored impaired proliferative responses to a level close to that observed in normal controls under the same experimental conditions. Although several research groups have shown that antioxidants used in vitro can inhibit HIV transcription by suppressing activation of the transcription factor NFκB, there are contradictory results concerning the effect of these molecules on normal lymphocyte proliferation and function.

Several groups indicated that a series of free-radical scavengers, including NAC and 2-ME, are able to inhibit proliferation, IL-2 secretion, and induction of the IL-2 receptor initiated by alloantigens or mitogens in murine and human T cells. On the other hand, several reports have dealt with the enhancement of T-cell and PBMC proliferative activity upon antioxidant treatment, which directly or indirectly enhances the intracellular contents of thiols. In addition, recent work indicates that the capacity of PBMC to enter the cell cycle correlates with intracellular GSH contents and that the generation of reactive oxygen intermediates (ROI) at the membrane level is essential in mitogen- and antigen-induced lymphocyte activation and their associated signaling. TCR stimulation of T lymphocytes through increased production of ROI, as well as acute in vitro exposure to oxidants, induces tyrosine-kinase activation and/or tyrosine-phosphatase inhibition, resulting in an increased tyrosine-phosphorylation of these cells. In agreement with these reports, resulting in an increased tyrosine phosphorylation of these cells, Staal et al. have shown that a moderate decrease in GSH levels is able to impair Ca^2+ influx and proliferative responses in anti-CD3 stimulated T cells, which could be mediated by increased phosphorylation of inhibitory sites of Lck and fyn. Flescher et al. have recently reported that chronic low-level oxidative stress results in suppression of protein-tyrosine phosphorylation and calcium mobilization. These results could explain our previous findings of defective protein-tyrosine phosphorylation in CD4 lymphocytes from HIV patients. Therefore, chronic LMT depletion in HIV infection could determine a defective tyrosine-phosphorylation response through increased production.
of ROI, as well as acute in vitro exposure to oxidants induces increased tyrosine phosphorylation and/or tyrosine-phosphatase inhibition, resulting in an increased tyrosine phosphorylation of these cells.35,38

Indeed, GSH depletion has significantly detrimental effects in normal lymphocyte function, perhaps by causing change in the activity of vital enzymes required for cellular metabolism, DNA synthesis, and signal transduction. Additionally, GSH has an essential role in protecting cell against ROI generation. However, to define the precise mechanism accounting for the effect of antioxidant treatment, a detailed study of signal transduction pathways, assessment of the respective role of the different thiols, as well as the different metabolic pathways involved in redox regulation, is needed. Because progression into the cell cycle may depend on the levels of intracellular GSH capable of restoring the activation-induced redox imbalance,22,34 it can be assumed that the presence of antioxidants during the activation process may inhibit the generation of ROI that are postulated to play an essential role in lymphocyte signaling.35 Because in our subsequent activation proceeded in the absence of antioxidants, this resulted in significant enhancement of the proliferative activity of CD4 T cells from both normal and HIV-infected individuals. This apparent discrepancy can be explained by our results (Fig 1), indicating that when activation was performed in the presence of NAC, there was a dose-dependent inhibition of lymphocyte proliferation, whereas preincubation with NAC followed by washing of supernatants and subsequent stimulation determined opposite results. Furthermore, similar conclusions were reported by Aillet et al.23 Our results are in agreement with the idea that the capacity of PBMC to enter the cell cycle correlates with intracellular GSH contents34 and that the generation of ROI at the membrane level is essential in mitogen and antigen-induced lymphocyte activation and their associated signaling.35,36 Because progression into the cell cycle may depend on the levels of intracellular GSH capable of restoring the activation-induced redox imbalance,22,34 it can be assumed that the presence of antioxidants during the activation process may inhibit the generation of ROI, which are postulated to play an essential role in lymphocyte signaling.35

As depicted in Tables 1 and 2, there was only a rough correlation between individual thiol levels and the severity of impairment in the respective proliferative responses for HIV-infected individuals. In this respect, it is known that cellular GSH levels are submitted to a tight regulation by an enzyme-regulated precise redox equilibrium and thereby the actual low-molecular thiol levels could not reflect the state of protein sulfhydryls. Alternatively, because lymphocyte activation is dependent on adequate levels of GSH to use it in a variety of reactions, including synthesis of proteins and DNA precursors, we speculated on the possibility of a deficient recycling of cellular thiols after their activation-induced consumption. In this regard, it has been proposed that HIV-infected lymphocytes display deficient activity of enzymes acting as scavengers of reactive oxygen intermediates (ROI) generated during lymphocyte activation as superoxide dismutase (SOD), catalase, glutathione reductase, and thioredoxin.26 When thiol levels were determined after overnight in in vitro cultures (activated and nonactivated lymphocytes), we observed that CD4+ lymphocytes from the LAR responders group failed to maintain adequate levels of intracellular thiols (Fig 2), irrespective of antioxidant treatment or CD3-mediated activation. Data presented in this figure suggest an impairment in the cellular antioxidant machinery aimed to maintain an appropriate redox status in CD4 T cells from HIV-infected patients who are unresponsive to antioxidant treatment. In contrast, in the HAR group despite reduced basal cellular thiols, the replenishment of intracellular thiols resulted in restoration of functional responses, indicating that antioxidant defenses are adequate.

PCD has been proposed to play an important role in CD4+ lymphocyte depletion during HIV infection,23,25 and oxidative stress might be involved in this process.12,13,26 We speculated on the possibility that impaired proliferative responses in CD4+ lymphocytes from the LAR could result from an abnormally triggered PCD process. Data presented in Fig 3 showed that recovery of proliferative responses in CD4+ lymphocytes from HIV-infected individuals was dependent on the ratio of apoptotic cells, indicating that enhancement of proliferative responses induced by NAC + 2-ME was limited to nonapoptotic cells. Although we did not assess the percentage of apoptotic cells previous to the measurement of thymidine uptake, the number of apoptotic cells observed before culture and 24 hours after culture for five patients indicates that there is a straightforward correlation between apoptosis and results of thymidine incorporation. These results show that the NAC-mediated increase in proliferative responses was not associated with apoptosis inhibition, but with enhanced proliferative ability of "nonapoptotic cells" or anergic cells and that the absence of proliferative response enhancement and the inability to maintain adequate LMT levels in vitro may probably be accounted for by the fact that cells are dying in cultures.

An inhibitory effect of NAC on lymphocyte apoptosis was recently demonstrated in the case of apoptosis induced by TNF-α and phorbol esters in a chronically HIV-infected monomyelocytic cell line.13 This effect was proposed to be mediated via GSH replenishment by NAC treatment. Similarly, it has been reported that NAC could inhibit activation-induced DNA fragmentation in T lymphocytes as a consequence of changes in intracellular redox status.28,40 Thus, the possibility exists that the concentration of NAC or the duration of treatment in our experimental conditions may be insufficient to inhibit cell death. Because mechanisms other than oxidant-induced cell death cannot be excluded, an alternative hypothesis could be that in vitro cell death in these patients results from an irreversible process triggered in vivo, which may not be influenced by in vitro antioxidant treatment. NAC has been reported to have beneficial effects on CD4 lymphocyte survival when administrated to HIV-1-infected individuals.12 However, it is presently unclear whether the reasons for these contradictory results depend on differences in experimental conditions (cells and apoptosis inducers, in vivo v in vitro conditions). In agreement with this, a recent report shows that NAC may have variable and even opposite effects on DNA fragmentation depending on inducers and cell type tested.40 Thus, the induction of
apoptosis in T cells is a complex and multifactorial process that probably involves several different signaling pathways. Overall, these results show for the first time that antioxidant treatment is able to revert, at least in some cases, the impaired proliferative activity of CD4 cells from HIV-infected patients. However, this action was mainly occurring on unresponsive cells, but not on cells engaged in ongoing programmed cell death, although it remains unclear whether this effect could be exerted at very initial stages of this process. Taking into account the important role of intracellular low-molecular thiols in proliferative responses and in protection of T cells from oxidative damage, our data suggest that a redox imbalance may be involved in the CD4 proliferative defect observed in HIV-1 patients. Thus, the results reported here, together with others indicating that NAC is a potent inhibitor of HIV replication, may help designing therapeutic strategies with antioxidant drugs in HIV disease.

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In vitro antioxidant treatment recovers proliferative responses of anergic CD4+ lymphocytes from human immunodeficiency virus-infected individuals

A Cayota, F Vuillier, G Gonzalez and G Dighiero