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COMMENTARY

Oxidative stress in immunodeficiency

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Oxidative modifications of biological molecules are essential for aerobic cells, and these cells are therefore permanently exposed to a variety of oxidants. Free radicals emerge in cases of excessive or uncontrolled oxidation, and as a consequence the pro-oxidant–antioxidant balance is disturbed in favour of the pro-oxidant, a condition known as oxidative stress [1]. In other words, there is a measurable shift in one or more redox couples to a more electron-deficient (oxidized) steady state [2]. Free radical damage is believed to be important in various areas of clinical medicine, including respiratory diseases, toxicology, mutations and inductions of neoplasms, vascular injury and cardiac disease, as well as dysfunction of the immune system [3]. Concerning the last-mentioned, the activation and proliferation of T lymphocytes, in particular, requires a delicately orchestrated action of oxidizing and reducing substances [4], and enhanced oxidative stress could therefore be important in the pathogeneses of immunodeficiency.

The pro-oxidant–antioxidant balance and/or redox status were recently determined in patients immunodeficient as a result of infection with human immunodeficiency virus (HIV) or because they suffer from common variable immunodeficiency (CVI) syndrome. Various investigators have monitored different parameters of oxidative stress in HIV-infected patients, often with conflicting results. Products of lipid peroxidation, as measured by the concentration of thiobarbituric acid-reactive substances, were found to be significantly higher in HIV-seropositive subjects, although we could not confirm this finding [5,6]. Reduced concentrations of polyunsaturated fatty acids, vitamin A, vitamin E and selenium may also be associated with enhanced oxidative stress, and indeed several investigators have found reduced concentrations of these parameters in HIV infection [7–9]; others have found normal or even increased levels [10,11]. HIV infection is also reported to be associated with systemic deficiency of glutathione (GSH), and the importance of this finding for the pathogenesis of this infection has received wide attention [12,13]. The measurement of blood GSH levels has pitfalls, however, that were not always accounted for. Decreased GSH levels were measured intracellularly in $CD4^+$ and $CD8^+$ T lymphocytes, but monochlorobimane was used as fluorescent label for the determination [14]. We have demonstrated that this probe lacks specificity and that the results of such studies should be interpretated with great caution [15]. Analysis of GSH levels using validated high-performance liquid chromatography (HPLC) methods did not confirm the presence of decreased intracellular GSH levels [6,16,17]. On the contrary, Aukrust *et al.* [17] as well as our own group [6] showed significantly enhanced concentrations of both oxidized and total GSH in $CD4^+$ lymphocytes but not in other cell subsets. This indicates that the GSH redox potential is disturbed in $CD4^+$ T lymphocytes from HIV-infected patients, a finding consistent with enhanced oxidative stress and possibly important for the pathogenesis of HIV infection. Aukrust *et al.* [17] also measured GSH and cysteine in plasma, but in this compartment the total and oxidized concentrations of these thiols were both reduced. Also, protein-bound cysteine was decreased in plasma from HIV-infected patients, and it seems therefore that HIV infection is associated with low disulphide formation in plasma. Thus, the pro-oxidant status is limited to $CD4^+$ T lymphocytes and is not present in plasma. The same authors also report elevated plasma concentrations of reduced homocysteine in HIV infection [18], and they argue that reduced homocysteine enhances free radical formation and stimulates HIV replication through NF-kappa B activation.

In a paper in this issue [19], as well as in an earlier report [20], plasma thiol levels in CVI patients are presented. Common variable immunodeficiency is a heterogeneous group of B-cell deficiency syndromes characterized by defective antibody production, and many patients also show some abnormality in T-cell function. Patients with this disorder are often afflicted by recurrent bacterial infections, gastrointestinal diseases including malabsorption and lymphoid hyperplasia, autoimmune diseases and increased incidences of lymphoid and gastrointestinal malignancies; however, they do not have an excess risk for cardiovascular diseases [21]. A subgroup of CVI patients is characterized by persistent immune activation *in vivo*. In this issue it is reported that, especially in this group, high plasma levels of reduced homocysteine are significantly correlated with increased concentrations of a product of lipid peroxidation (high plasma malonaldehyde) and low plasma levels of vitamin E [19]. It is concluded that elevated plasma levels of reduced homocysteine are a marker of enhanced oxidative stress.

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It should first be questioned whether enhanced oxidative stress was demonstrated in these patients. Antioxidants appear to be indicators of the susceptibility of a subject to reactive oxygen species, reflecting the risk rather than the actual level of free radical damage. By assaying antioxidants such as vitamin E and carotene, it is difficult to discern cause and consequence of an observed effect [22]. Most pronounced effects have been observed in CVI patients with persistent immune activation, and these patients are characterized by low CD4 cell counts and the occurrence of splenomegaly and nodular intestinal lymphoid hyperplasia [23]. However, especially in these patients, gastrointestinal problems may arise and the absence of diarrhoea or hypoalbuminaemia does not exclude vitamin deficiency due to malabsorption. In animal experiments, food elements were found to be an important factor for the excretion of malonaldehyde; various fatty acids induced marked changes and vitamin E depletion or iron supplementation greatly increased malonaldehyde levels [24].

Free radical damage can indeed be analysed by monitoring products of lipid peroxidation and shifts in redox status. Lipid peroxidation was assessed by measurement of plasma malonaldehyde concentration; however, this has important limitations, and many alternative parameters are available for this purpose [22]. Thiols other than homocysteine were measured in these patients, but the results, which are not in agreement with previous theories regarding enhanced oxidative stress, are not discussed. In previous reports it was found that elevated levels of reduced homocysteine were associated with a decrease in cysteine levels [25], and the role of scavenger in a pro-oxidant state was therefore attributed to cysteine. However, in the present study no abnormality of cysteine was found. Enhanced levels of reduced plasma homocysteine have been found in HIV-infected as well as CVI patients, but without a significant reduction in plasma levels of oxidized thiols such as GSH [20] or cysteine [19], as would be expected in the case of a shift in redox status. In addition, homocysteine is supposed to cause endothelial damage through free radical damage; is it therefore not peculiar that no cardiovascular diseases are reported in these patients?

Overall, it can be concluded that some immunodeficiency states are accompanied by metabolic changes, but the question remains whether these metabolic changes are simply manifestations of the damage that is caused by long-term immunodeficiency or whether these changes also compromise immune function.

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