Agents and Actions, vol. 22, 3/4 (1987)

0065-4299/87/040340-03 $ 1.50+0.20/0

Lipid peroxidation and protective enzymes during the course of myocardial infarction

J. Loeper, J. Goy, O. Bedu and L. Rozensztajn

CAU Saint Antoine, 27, rue Chaligny, F-75012 Paris

Formation of atheromatous plaques from lipids deposited in the intima of arteries is a process which has not yet been completely understood despite several hypotheses which have been proposed, and it thus was of interest to investigate whether the processes involved in lipid peroxidation might affect the mechanism of atherosclerosis.

Peroxidation acts upon fatty acids which have many double bonds and gives rise to free radicals and endogenous peroxides that are very active, possessing chemotactic and cytotoxic properties; one free radical generates another free radical, and these chain reactions may be very harmful for cells if they are not stopped by enzymes of defense [1]: peroxides predispose to platelet aggregation; exposure of cell membranes to peroxides may induce changes in cell permeability and lead to necrosis; it is also probable that an excess of peroxides prevents prostacyclin synthesis, which opposes platelet aggregation.

It is logical to believe that lipid oxidation and peroxidase activity play a part in the mechanism of atherosclerosis to facilitate its onset and to enter into the complications which develop as a consequence.

The main parameters which were assessed in this research were plasma malondialdehyde which is an end-product of the catabolism of polyunsaturated acids by free radicals and is an indicator of peroxidation; and two enzymes of defense present in erythrocytes were studied: superoxide dismutase which catalyzes the reaction of O_2^-. into H_2O_2 and glutathione peroxidase which eliminates the H_2O_2 by transforming it into H_2O.

Material and methods

Malondialdehyde (MDA): in the plasma, proteins are precipitated by phosphotungstic acid and MDA is assayed by fluorimetry with thiobarbituric acid.

Superoxide dismutase (SOD): SOD catalyses the transformation of O_2^-. into H_2O_2: the oxidation of xanthine by xanthine oxidase releases H^+ ions which reduce cytochrome, an H^+ receiver; this reduction causes an increase in the O.D. which is inhibited by SOD the results are given in SOD units able to inhibit cytochrome reduction by 50%.

Glutathione peroxidase (GP): GP reduces H_2O_2 into H_2O by the oxidation of reduced glutathione; the method employed consisted in assaying reduced glutathione by the intermediary NADPH; the results are expressed in units per g of hemoglobine.

The study was conducted with patients with hyperlipidemia, with arterial disease or not.

Results

In an initial clinical study, we sought to detect changes in plasma levels of MDA in a population with normal levels of serum lipids without arterial disease compared to a population with hyperlipi-

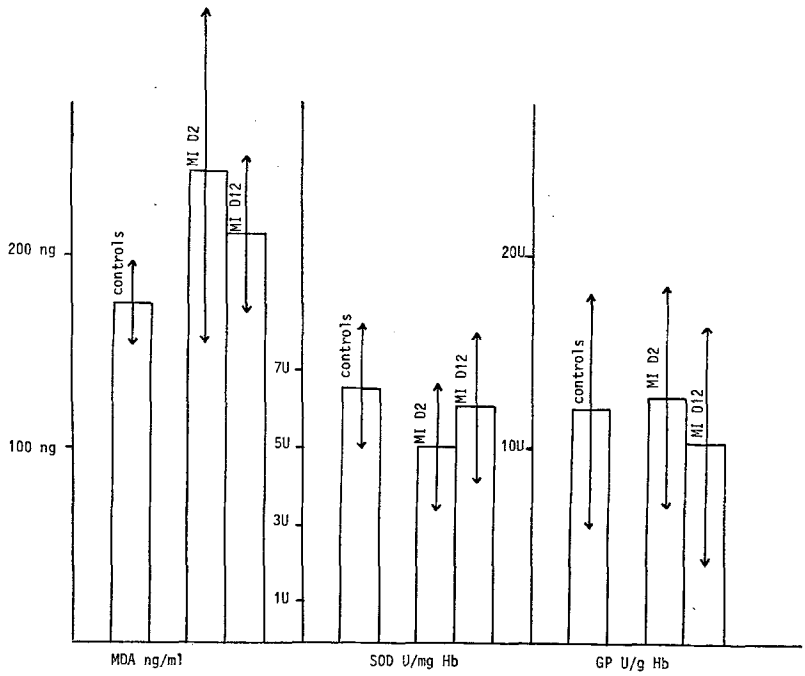

Figure 1
Malonedialdehyde, superoxidismutase and glutathion peroxidase in patients with myocardial infarction compared to controls, at day 2 and day 12. Results are given as mean ± standard deviation.

demia with or without arterial disease; we compared 40 control subjects to 41 patients with hyperlipidemia without arterial disease (Hlp0) and 32 patients suffering from arteriosclerosis obliterans of the lower limbs, or cerebral atherosclerosis or myocardial infarction (Hlp+): the level of MDA was 157 ng in control subjects, 220 ng in the Hlp0 subjects and 249 ng in Hlp+ patients. Differences between these three groups are significant.

These results prove the involvement of peroxidase oxidation during the course of atherosclerotic diseases and are confirmed by the increase of unsaturated fatty acids observed in patients with atherosclerosis [2].

Our present study involved more specifically patients who presented with a clinically-defined myocardial infarction and we assayed MDA in plasma, and SOD and GP in the erythrocytes of:

- 25 patients with myocardial infarction (MI)
- 8 patients with unstable angina
- 20 control subjects

Assays were made at the time the patients were admitted to the hospital in an interval no greater than 48 hours after the onset of the attack and then 12 days later, to determine changes in the different parameters.

Results of changes in levels of MDA, SOD and GP on the average between days 2 and 12 are listed in Figure 1: levels of MDA are significantly increased compared to control subjects (243 ng vs. 175 ng) during the 48 hours period following the MI and there was a decrease between day 12 (212 ng) and day 2 (243 ng). SOD was decreased in the cardiac patients (5.08 U.) compared to control subjects (6.6 U.) during the 48 hours period, and increased between day 12 (6.1 U.) and day 2 (5.08 U.). Levels of GP in cardiac patients and control subjects were the same at the onset of the MI (12.11 U.) and decreased on day 12 (10.29 U.) compared to day 2 (12.68 U.). Variations in MDA and enzyme levels in patients with unstable angina were identical.

The course of the three parameters expressed in percent between the onset of the MI and 12 days later were a decrease of 13% for MDA, 19% for GP and a 20% increase for SOD.

Discussion

Many investigators have studied the mechanism involved in lipid oxidation but only a few have looked at its role in the genesis of atherosclerosis. The increase in levels of peroxides was observed in serum lipoproteins of atherosclerotic rabbits in 1973; in 1982, Dousset [3] observed an increase in MDA in patients with MI within the first three days following the attack. More recently, Ledwozyw [4] investigated the relationship between serum lipids and peroxidation in patients with atherosclerotic disease. In an experimental study [5] we ourselves observed an increase in levels of plasma and aortic MDA in rabbits fed a hypercholesterol diet compared to control rabbits.

In the present study, we found an increase in levels of MDA after the onset of a myocardial infarction, at the same time as a decrease in superoxidismutase. It is probable that hypoxia, ischemia and reoxygenation which occur with a myocardial infarction induce a rush of free radicals which slow momentarily SOD activity and increase MDA. There are opposite changes in SOD and GP and it is clear that an increase in MDA and a decrease in SOD without a large addition of GP has a dangerous effect.

SOD treatment is currently being assessed in trials with patients suffering from vascular disorders.

References

[1] J. McCord and I. Fridovich, *The Biology and Pathology of Oxygen Radicals,* Annals of International Medicine *89,* 122–127 (1978).

[2] J. Loeper, J. Goy, J. Emerit, L. Rozensztajn, C. Jeny and O. Bedu, *Etude des acides gras et de la peroxydation lipidique au cours de l'athérosclérose humaine,* Sem. Hôp. Paris *59,* N° 22, 1657–1660 (1983).

[3] J. C. Dousset, M. Trouilh and M. J. Foglietti, *Plasma malonaldehyde levels during myocardial infarction,* Clin. Chim. Acta *129,* 319–322 (1983).

[4] A. Ledwozyw, J. Michalak, A. Stepien and A. Kadziolka, *The relationship between plasma triglycerides, cholesterol, total lipids and lipid peroxidation products during human atherosclerosis,* Clin. Chim. Acta *155,* 275–284 (1986).

[5] J. Loeper, J. Emerit, J. Goy, L. Rozensztajn and M. Fragny, *Etudes des acides gras et de la peroxydation lipidique dans l'athérome expérimental du lapin,* Path. Biol. *32,* N° 6, 693–697 (1984).