
It is generally recognized that high levels of low density lipoprotein are associated with the development of atherosclerosis. A considerable body of evidence has accumulated to indicate that a critical step in atherogenesis, is oxidation of LDL, and that circumstances that promote oxidation of LDL may constitute a major risk factor for atherosclerosis (1). A major catalyst of lipoprotein oxidation in vivo is the enzyme 15-lipoxygenase. This enzyme can oxidize polyunsaturated fatty acids present in intact lipids and lipoproteins in situ. Steinberg has estimated that up to 75% of the oxidative damage to LDL by macrophages is caused by 15-lipoxygenase. Atherosclerotic lesions in rabbit aorta contain increased 15-lipoxygenase activity (2) and oxidized LDL epitopes colocalize with immunoreactive 15-lipoxygenase protein in the lesions (1). Hypercholesterolemia is associated with increased 15-lipoxygenase in a number of rabbit tissues - particularly lung - where elevations of several hundred fold were observed in some animals (3). Systemic induction of the 15-lipoxygenase was duplicated by administration of the cytoplytic agent phenylhydrazine, and was accompanied by systemic lipoprotein oxidation detected by accumulation of the characteristic oxidation products 15-HETE and 13-HODE in the serum lipids.

The phenomenon of a hyperresponsive 15-lipoxygenase phenotype was encountered during analysis of vascular tissues in hypercholesterolemic rabbits. Of a total of 32 rabbits, 7 animals (21%) were hyperoxidizers with elevations of 15-lipoxygenase in the range of 150-250 units/g, compared to a normal level of 0.5-1 units/g. 10 Rabbits were in the medium range (5-25 units/g) and 15 were in the low range (1-5 units/g).

In studies with human blood donors, similar elevations in 15-lipoxygenase activity in polymorphonuclear leukocytes, have been observed in response to cytolytic concentrations of drugs such as ibuprofen (4,5). The 15-lipoxygenase responses of PMN's from a group of 20 subjects were therefore analyzed for evidence of a hyperoxidizer phenotype (Table 1).

Table 1: Activation of 15-Lipoxygenase in Human PBL's

<table>
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<tr>
<th>No. of Subjects</th>
<th>15-Lipoxygenase Activity/10^7 cells</th>
<th>Increases in 15-Lipoxygenase activity/10^7 cells</th>
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<tr>
<td>20 Subjects</td>
<td>0.64 ± 0.12 (Basal Activity)</td>
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Legend: Blood was obtained from 20 human donors who had not taken any aspirin-like drugs during the preceding 2 weeks. PMN leukocytes were isolated using a modified hypaque-ficol technique, and 15-lipoxygenase activity was measured both before and immediately following exposure to ibuprofen (5 mM).

Basal levels of the enzyme were low in all subjects, averaging 0.64 ± 0.12 units/10^7 cells. A hyperoxidizer profile was observed in almost half (45%) of the tested subjects, with 15-lipoxygenase elevations in the 11-70 fold range. The remaining individuals displayed responses in the low to medium (2-10 fold) range. The hyperoxidizer profile was reproducible when subjects were tested at intervals up to 3 months, suggesting that it was probably an inherent characteristic of the individual rather than a temporary phenomenon due to some environmental or other external circumstance.

The results were further analyzed to determine if the basal levels of 15-lipoxygenase in PMN's are predictive of the hyperoxidizer profile. The basal and activated levels of the enzyme for 4 individuals who were hyperoxidizers are compared in Figure 1 with those of 4 individuals whose 15-lipoxygenase displayed only low or moderate elevation following exposure to 5 mM ibuprofen.

Figure 1: 15-Lipoxygenase activation in human subjects

In general, the basal level of the enzyme was not predictive of the hyperoxidizer profile, since individuals such as subjects #6 and #7 with basal levels in the low range, displayed 10-20 fold activation, whereas subject # 3 with an above average basal level gave less than a 30% elevation following drug treatment. Of interest also is the retest of subject # 7 after 4 week interval, indicating the stability of the hyperoxidizer phenotype.

It has been shown that systemic activation of the 15-lipoxygenase in rabbits is associated with increased lipoprotein oxidation (3). The results reported here in humans raise the question whether a hyperoxidizer phenotype is associated with an increased risk of lipoprotein oxidation and atherosclerosis, and whether antioxidant vitamin therapy is protective. Experiments to determine if the hyperoxidizer phenotype is associated with a family history of atherosclerosis are now being planned.

References


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