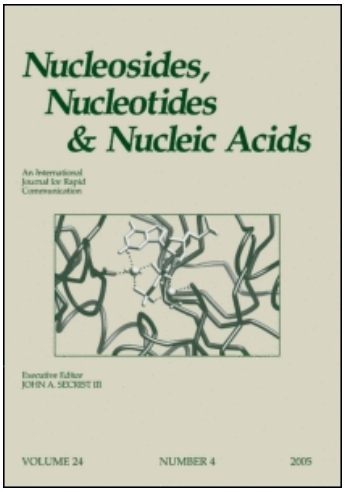

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Antioxidant Status and Purine Bases in Carotid Artery Plaque

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ANTIOXIDANT STATUS AND PURINE BASES IN CAROTID ARTERY PLAQUE

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□ *Free radical excess and oxidative stress are implicated in the formation and progression of atherosclerotic plaque through actions on susceptible vascular cells, such as by activating xanthine oxidase. Purine bases and other antioxidant compounds could play important protective roles in atherogenesis, as could nonenzymatic low molecular weight thiol defenses, not previously evaluated in carotid artery plaque. Therefore, we measured purine catabolites (hypoxanthine, xanthine, uric acid, allantoin) and antioxidant compounds (total sulphydryl groups, homocysteine, cysteine, and glutathione) in advanced carotid artery plaque and found a high ratio of allantoin to uric acid, suggesting a ongoing local oxidative stress.*

Keywords Antioxidant; SH group; purine catabolites

INTRODUCTION

Although atherosclerosis is a multifactorial disease, an important role in its development is attributed to oxidative stress.[1–3] Arterial wall endothelial cells are continuously exposed to changing oxygen pressure and high rates of free radical formation that can modify LDL and initiate events that lead to progression of plaque.[4–6] Such modifications could also potentially result from reduced antioxidant defenses normally provided by low molecular weight thiols, such as homocysteine (Hcys), cysteine (cys), glutathione (GSH), and uric acid (UA). Although UA is the most abundant scavenger of free radicals in humans by virtue of the absence of activity of uricase, it is important to note that purine catabolism increases in states of oxidative stress,[7–8] and that, conversely, activation of xanthine oxidase promotes production of free radicals.

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MATERIALS AND METHODS

Plaque material was obtained from 20 subjects (10 males and 10 females with an age range of 57–83 years), hospitalized at the Department of Surgery, University of Siena, Italy for carotid endarterectomy. Plaques were rinsed twice in cold phosphate buffered saline solution to minimize blood residue, frozen in liquid nitrogen, homogenized by three 1-minute cycles on a Dismembrator (Braun AG, Melsungen, Germany). Twenty mg samples of plaque homogenate were vigorously resuspended in 1 ml of phosphate buffered saline, and, after centrifugation at 15000 × g (Mikro 12-24, Hettich D-78532 Tuttlingen) for 15 minutes, the supernatant layer was used for assay of total thiol groups (SH) by spectrophotometric assay (Ellman's reagent),[9] and for UA, allantoin (ALL), Hx, X,[10] and Hcys, Cys, GSH[11] by HPLC.

RESULTS

We analyzed purine catabolite (UA, ALL, Hx, X) and thiol compound (SH, Hcys, cys, GSH) levels in carotid artery plaque homogenates. All concentrations (μmol/g tissue) are expressed as mean ± standard deviation. The results are: UA = 4.9 ± 1.28; ALL = 21.49 ± 15.10; Hx = 27.26 ± 8.72; X = 8.79 ± 2.71; SH = 112.21 ± 62.4; Hcys = 0.40 ± 0.33; cys = 5.46 ± 2.28; GSH = 1.35 ± 0.8.

A typical HPLC chromatogram of plaque homogenate is shown in Figure 1.

DISCUSSION

We acknowledge that, in the absence of control tissue analyses, the significance of the absolute purine base and thiol compound levels measured in advanced atherosclerotic plaque remains uncertain. Nevertheless, the most striking finding of this study was the relatively high levels of ALL observed. ALL can be formed only from the non-catalytic oxidative action of free radicals on uric acid, because of the absence of uricase in human tissues. ALL was found in higher concentrations than UA (mean ALL/UA >4/1), leading us to propose that this finding may reflect continued oxidative stress, even in the advanced atherosclerotic lesions. Further studies to determine the contents of these compounds in other tissues of interest to the progression of the atherosclerotic plaque and to measure the effects of interventions to improve purine base-mediated antioxidant levels are warranted.

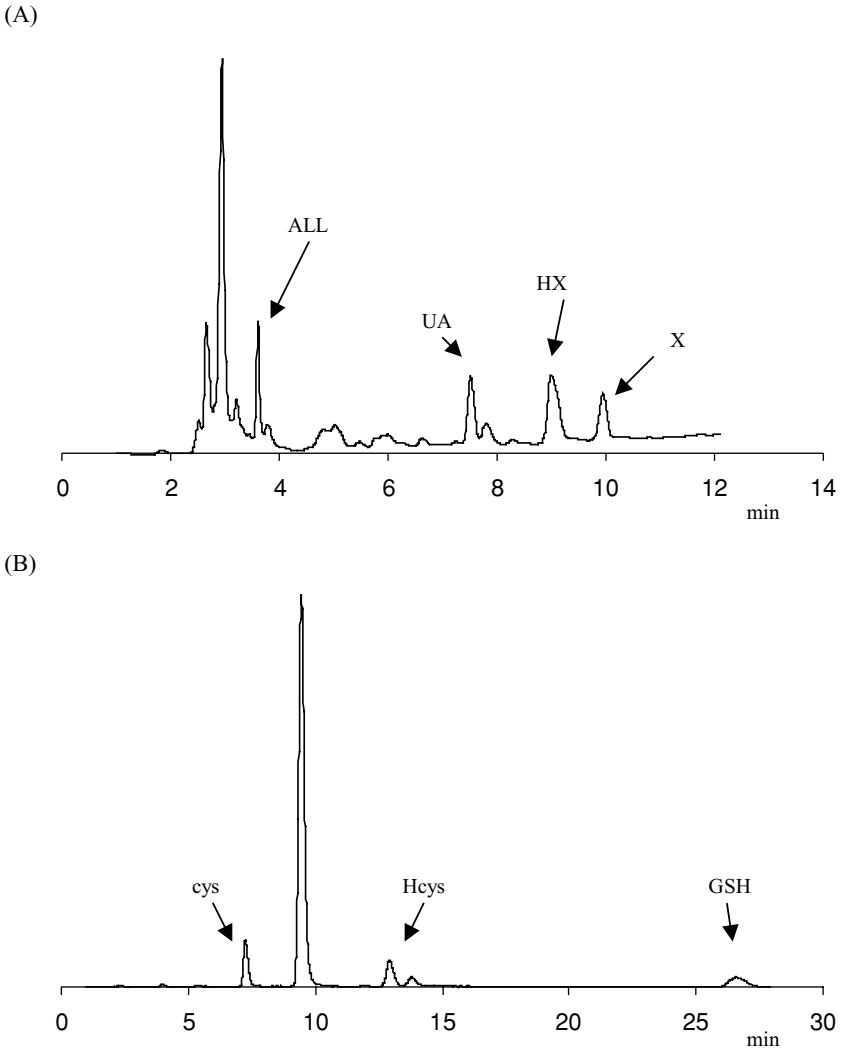

FIGURE 1 Typical HPLC chromatogram of atherosclerotic plaque. A): uric acid (UA), allantoin (ALL), hypoxanthine (Hx), xanthine (X); B): cysteine (cys), homocysteine (Hcys), glutathione (GSH).

REFERENCES

1. Khatri, J.J.; Johnson, C.; Magid, R.; Lessner, S.M.; Laude, K.M.; et al. Vascular oxidant stress enhances progression and angiogenesis of experimental atheroma. *Circulation* **2004**, 109, 520–525.
2. Martinet, W.; de Meyer, G.R.; Herman, A.G.; Kockx, M.M. Reactive oxygen species induce RNA damage in human atherosclerosis. *Eur. J. Clin. Invest.* **2004**, 34, 323–327.
3. Martinet, W.; Knaapen, M.W.; De Meyer, G.R.; Herman, A.G.; Kockx, M.M. Elevated levels of oxidative DNA damage and DNA repair enzymes in human atherosclerotic plaques. *Circulation* **2002**, 106, 927–932.

4. Navab, M.; Imes, S.S.; Hama, S.Y.; Hough, G.P.; Ross, L.A.; et al. Monocyte transmigration induced by modification of low density lipoprotein in cocultures of human aortic wall cells is due to induction of monocyte chemotactic protein 1 synthesis and is abolished by high density lipoprotein. *J. Clin. Invest.* **1991**, 88, 2039–2046.
5. Cushing, S.D.; Berliner, J.A.; Valente, A.J.; Territo, M.C.; Navab, M.; et al. Minimally modified low density lipoprotein induces monocyte chemotactic protein 1 in human endothelial cells and smooth muscle cells. *Proc. Natl. Acad. Sci.* **1990**, 87, 5134–5138.
6. Liu, K.Z.; Cuddy, T.E.; Pierce, G.N. Oxidative status of lipoproteins in coronary disease patients.*Am. Heart. J.* **1992**, 123, 285–290.
7. Skibba, J.L.; Powers, R.H.; Stadnicka, A.; Cullinana, D.W.; Almagro, U.A.; et al. Oxidative stress as a precursor to the irreversible hepatocellular injury caused by hyperthermia. *Int. J. Hypotermia* **1991**, 7, 749–761.
8. Chen, Y.F.; Li, P.L.; Zou, A.P. Oxidative stress enhances the production and actions of adenosine in the kidney. *Am. J. Physiol. Regul. Integr. Comp. Physiol.* **2001**, 281, R1808–1816.
9. Hu, M.L.; Dillard, C.J.; Tappel, A.L. In vivo effects of aurothioglucose and sodium thioglucose on rat tissue sulfhydryl levels and plasma sulfhydryl reactivity. *Agents Actions* **1988**, 25, 132–138.
10. Terzuoli, L.; Porcelli, B.; Setacci, C.; Giubbolini, M.; Cinci, G.; et al. Comparative determination of purine compounds in carotid plaque by capillary zone electrophoresis and high performance liquid chromatography. *J. Chrom.* **1991**, 728, 185–192.
11. Kuo, K.; Still, R.; Cale, S.; McDowell, I. Standardization (external and internal) of HPLC assay for plasma homocysteine. *Clin. Chem.* **1997**, 43, 1653–1655.