Antioxidants: The good, the bad and the ugly

Jean-Claude Tardif MD FRCPC FACC

J-C Tardif. Antioxidants: The good, the bad and the ugly. Can J Cardiol 2006;22(Suppl B):61B-65B.

Inflammation has a fundamental role in mediating all stages of atherosclerotic disease. The key role of oxidation in linking lipids and inflammation to atherosclerosis is compelling and is supported by experimental evidence. However, the relevance of the antioxidant hypothesis for the treatment of patients with atherosclerosis has not been definitively proven. Results of randomized trials with 'antioxidant' vitamins have been disappointing, and there are potentially important problems associated with their use, including their potential pro-oxidant effects. Probucol has reduced postpercutaneous coronary intervention (PCI)-restenosis and progression of carotid atherosclerosis in clinical trials. The antioxidant vascular protectant AGI-1067 has also been effective at preventing atherosclerosis in all tested animal models. The nonintervened reference coronary segments of the PCI vessel demonstrated improvements with AGI-1067 in the Canadian Antioxidant Restenosis Trial-1 (CART-1), evidence supportive of a clinical effect on slowing atherosclerosis progression. Two trials test the antioxidant/anti-inflammatory hypothesis with AGI-1067; CART-2 assesses its value for the reduction of both atherosclerosis progression and post-PCI restenosis, and Aggressive Reduction of Inflammation Stops Events (ARISE), which is evaluating its effects on hard cardiovascular outcomes.

Key Words: *Antioxidants; Atherosclerosis; Inflammation; Oxidative stress*

Antioxydants : Le bon, la brute et le truand

L'inflammation joue un rôle fondamental de médiation à tous les stades de l'athérosclérose. Le rôle central de l'oxydation dans les phénomènes qui relient les lipides et l'inflammation à l'athérosclérose n'est plus à démontrer et s'appuie sur des preuves expérimentales. Par contre, la pertinence de l'hypothèse antioxydante pour le traitement des patients qui souffrent d'athérosclérose n'a pas encore été prouvée hors de tout doute. Les résultats d'essais randomisés menés sur des vitamines « antioxydantes » ont donné des résultats décevants et leur utilisation est associée à des problèmes potentiellement importants, notamment à un risque d'effets pro-oxydants. Le probucol a réduit la resténose consécutive aux angioplasties coronariennes percutanées (ACP) et a ralenti la progression de l'athérosclérose carotidienne lors d'essais cliniques. L'AGI 1067, un antioxydant vasoprotecteur, s'est également révélé efficace à prévenir l'athérosclérose dans tous les modèles animaux testés. Les segments coronariens de référence sur lesquels les chercheurs ne sont pas intervenus lors d'ACP ont montré des signes d'amélioration avec l'AGI-1067 dans le cadre de l'étude CART-1 (pour *Canadian Antioxidant Restenosis Trial-1*); ces signes en confirment l'effet clinique sur le ralentissement de l'athérosclérose. Deux essais vérifient par ailleurs l'hypothèse de l'effet antioxydant/anti-inflammatoire de l'AGI 1067 : l'essai CART-2 évalue son utilité dans le ralentissement de la progression de l'athérosclérose et la réduction de la resténose post-ACP et l'essai ARISE (pour *Aggressive Reduction of Inflammation Stops Events*) mesure ses effets sur les paramètres cardiovasculaires définitifs.

Reactive oxygen species (ROS) include both free radicals, which typically have an oxygen- or nitrogen-based unpaired electron in their outer orbits, and other species, such as hydrogen peroxide, that act as oxidants. The mitochondria, the cellular membrane oxidases, such as NADPH oxidase, nitric oxide synthase and myeloperoxidase are major sources of ROS. The byproducts associated with the metabolism of arachidonic acid by cyclooxygenase, lipoxygenase and cytochrome P450 monooxygenase also result in the production of ROS. The targets for damage from ROS are not only lipids, such as polyunsaturated fatty acids in cellular membranes and low density lipoproteins (LDL), but also proteins and nucleic acids.

Lipid peroxidation products react strongly with various biological substrates leading to cellular injury. Oxidative damage to polyunsaturated fatty acids of membrane phospholipids leads to a destabilization in their intermolecular packing characteristics and subsequent loss in membrane integrity. Lipid peroxides break down into smaller molecules that either remain covalently linked to the phospholipid glycerol backbone or are released into the cytosol. In addition, the oxidized LDL particle does not effectively bind to the high-affinity LDL receptor, thus extending its circulation and enhancing its uptake into macrophages. Besides lipids, other constituents of the cell are vulnerable to ROS-induced damage, especially nucleic acids associated with nuclear and mitochondrial DNA, and oxidizable proteins. Damage to nucleic acids can be broadly categorized as either strand breaks or base modifications. Oxidizable amino acids associated with cellular proteins are also vulnerable to ROS-induced damage, leading to a loss in normal function. The effects of protein oxidation products such as residue-specific changes, fragmentation products and cross-linked reaction products with other intracellular molecules can be highly deleterious to the cell (for example, disrupting proteins involved in transport or cell regulation).

OXIDATIVE STRESS AND ATHEROSCLEROSIS

Atherosclerosis is now understood to be a chronic inflammatory disease characterized by the excess accumulation of monocyte-derived macrophages within the arterial wall (1). Compelling

Montreal Heart Institute, Montreal, Québec
Correspondence: Dr Jean-Claude Tardif, Director, Montreal Health Institute Research Centre, 5000 Belanger Street, Montreal, Québec H1T 1C8. Telephone 514-376-3330 ext 3612, fax 514-593-2500, e-mail jean-claude.tardif@icm-mhi.org

evidence points to oxidative stress as an important trigger in the complex chain of events leading to atherosclerosis (2). The expression of chemotactic factors, such as monocyte chemotactic protein-1, and of adhesion molecules, such as vascular cell adhesion molecule-1 (VCAM-1), are enhanced by oxidative stress and oxidized LDL (3,4). The release of macrophage colony-stimulating factor is also stimulated by modified LDL (5). Expression of these factors results in the attraction and adhesion of monocytes to the arterial wall, and the promotion of their differentiation into tissue macrophages. Exposure to the superoxide ion activates the nuclear factor-kappa B regulatory complex and triggers the transcription of several atherosclerosis-related genes (6). This series of events leads to the accumulation of macrophages in the arterial wall, which incorporate oxidized LDL to form foam cells. Oxidized LDL, in turn, stimulates the release of interleukin-1 from macrophages (7). The activity of matrix metalloproteinases, also regulated by oxidative stress, appears to be closely coupled with smooth muscle cell activation and migration, and has also been implicated in the pathophysiology of plaque rupture (8). Furthermore, reactive oxygen species can lead to platelet activation and thrombus formation (9). Therefore, oxidative stress appears to be important in both the early and the later stages of the atherosclerotic process.

There is also evidence that oxidative stress occurs early after angioplasty (10,11). Reactive intermediates can be generated by damaged endothelium, activated platelets and neutrophils at the angioplasty site. These oxidizing metabolites can induce chain reactions that result in endothelial dysfunction, macrophage activation, smooth muscle cell migration and proliferation, and matrix remodelling (12,13).

CONCERNS ASSOCIATED WITH THE USE OF 'ANTIOXIDANT' VITAMINS

The results of prospective epidemiological studies have supported a protective role for antioxidant vitamins in cardiovascular diseases (14). Such observational studies are limited by their inability to control for the effects of unknown or unmeasured confounders. In contrast, the results of randomized clinical trials with antioxidant vitamins have been disappointing. An excess risk for cancer and cardiovascular mortality was observed with beta-carotene in the large Alpha-Tocopherol/Beta-Carotene (ATBC) (15) and Carotene And Retinol Efficacy Trial (CARET) (16) studies. In terms of the results with vitamin E, except for the Cambridge Heart Antioxidant Study (CHAOS) (17), most secondary prevention trials have not shown protective effects (18-20). The Gruppo Italiano per lo Studio della Sopravivenza nell'Infarcto miocardico (GISSI) prevention trial was an open-label, randomized trial assessing dietary supplements of vitamin E (300 mg daily) and n-3 polyunsaturated fatty acids in 11,324 patients who had had a recent myocardial infarction (18). The Heart Outcomes Prevention Evaluation (HOPE) study tested an angiotensin-converting enzyme inhibitor and vitamin E (400 U daily) in a factorial design for the prevention of cardiovascular morbidity and mortality (19). The study population included more than 9000 patients who were at high risk of developing a major cardiovascular event. The Heart Protection Study (HPS) recruited more than 20,000 patients at high risk of atherosclerosis-related events because of a past history of myocardial infarction, or other evidence of atherosclerosis, diabetes mellitus or hypertension (20). The patients were randomly assigned in a two by two factorial design to receive simvastatin alone, antioxidant vitamins (600 mg vitamin E, 250 mg vitamin C and 20 mg beta-carotene daily), a combination of simvastatin and vitamins, or placebo. Vitamin E did not offer cardiovascular protection in the GISSI, HOPE and HPS trials (18-20).

There are potentially important problems associated with the use of vitamins, which include their potential pro-oxidant effects (21,22). Alpha-tocopherol itself becomes a radical when it scavenges a free electron. Because the tocopherol radical is relatively unstable, it can become a donor of free radicals or act as a pro-oxidant. Bowry et al (21) showed that, under high concentrations of vitamin E, lipid peroxidation is actually faster in the presence of alpha-tocopherol, and lipid peroxidation is propagated within LDL particles by reaction with the tocopherol radical. This may explain why a high dose of alpha-tocopherol worsens endothelial-dependent vasodilation, whereas a low dose improved it in cholesterol-fed rabbits (22). These problems associated with vitamin E supplementation may explain the negative results of the vitamin arms of the GISSI, HOPE, HPS, Women's Angiographic Vitamin and Estrogen (WAVE) and MultiVitamins and Probucol (MVP) trials (18-20,23,24). Moreover, antioxidant vitamins negated in part the beneficial effects of lipid-lowering agents in patients with coronary artery disease in the recently published High density lipoprotein Atherosclerosis Treatment Study (HATS) (25). This phenomenon had also been previously observed in the MVP trial (24,26,27).

RESULTS WITH POWERFUL SYNTHETIC ANTIOXIDANTS

Animal studies have shown a beneficial effect of antioxidants on both neointimal formation and arterial remodelling after balloon angioplasty (28-32). More recently, the antioxidant probucol was shown to promote the regeneration of functional endothelium in balloon-injured rabbit aortas (33). Small clinical studies, along with the MVP and the Probucol Angioplasty Restenosis Trial (PART), have shown that probucol started before balloon angioplasty reduces coronary restenosis (13,24,26,27,34-38). However, prolongation of the QT interval and lowering of high density lipoprotein cholesterol with probucol remain potential long-term safety concerns. An agent that would offer antioxidant properties similar to probucol but with more favourable safety and pharmacokinetic profiles would have significant potential for the prevention of both restenosis and atherosclerosis progression. The recently developed antioxidant and vascular protectant AGI-1067 appears to fulfill these criteria and was therefore assessed in the Canadian Antioxidant Restenosis Trial-1 (CART-1) (39).

AGI-1067, the monosuccinic acid ester of probucol, is a phenolic antioxidant member of a novel class of agents termed vascular protectants (40). It has strong antioxidant properties, equipotent to those of probucol, and anti-inflammatory properties (4,41). AGI-1067 also exhibits greater water solubility and cell permeability than probucol. AGI-1067 has the ability to selectively block the expression of oxidation-sensitive inflammatory genes that code for VCAM-1 and monocyte chemotactic protein-1 (42). Although AGI-1067 and probucol are equipotent antioxidants, they produce different effects on VCAM-1 expression in human aortic endothelial cells. AGI-1067 produces a concentration-related decrease in tumour necrosis factor-alpha-stimulated VCAM-1 expression in the

concentration range of 2.5 µmol/L to 10 µmol/L, while probucol was shown to have no effect at concentrations as high as 100 µmol/L (41,43). AGI-1067 has also been a powerful inhibitor of smooth muscle cell proliferation in experimental studies.

CART-1

CART-1 (39) was a double-blind, double-dummy, multicentre trial in which 305 patients scheduled to undergo elective percutaneous coronary intervention (PCI) with or without stent placement (85% were treated with stents) were randomly assigned to placebo, probucol 500 mg twice daily, or AGI-1067 70 mg, 140 mg or 280 mg once daily. Patients were treated for two weeks before and four weeks after PCI in this phase 2 trial. The minimal lumen area at the site of PCI on follow-up intravascular ultrasound (IVUS) was on average 2.66 mm^2 in the placebo group, 3.69 mm^2 in the probucol group, 2.75 mm^2 in the AGI-1067 70 mg group, 3.17 mm^2 in the AGI-1067 140 mg group and 3.36 mm^2 in the AGI-1067 280 mg group (P<0.05 for AGI-1067 280 mg and probucol versus placebo) (39). There was also a significant dose-response relationship of AGI-1067 for the primary end point (P=0.02, Figure 1). The benefit of AGI-1067 and probucol was present immediately after PCI, raising the possibilities that countering oxidative stress may rapidly improve endothelial function (44) or that changes in plaque content (45) may have contributed to an optimal response to PCI.

Volumetric (three-dimensional) changes of nonintervened coronary reference segments, away from the PCI site, were also evaluated with IVUS in a post hoc analysis in CART-1 (39). The mean change in lumen volume (follow-up minus baseline) in the reference segments was –5.3 mm^3 in the placebo group, –0.2 mm^3 in the probucol group and –2.4 mm^3 in the AGI-1067 70 mg group, but was +3.5 mm^3 in the AGI-1067 140 mg group and +1.8 mm^3 in the AGI-1067 280 mg group (P=0.05 for AGI-1067 140 mg versus placebo; P=0.077 for dose-response relationship). These findings may represent the first clinical evidence of vascular protection with AGI-1067 (46). This effect was due to trends for both the inhibition of negative remodelling and reduction of plaque burden. These clinical results are supported by demonstration of atherosclerosis prevention by AGI-1067 in all tested animal models (41), including in apolipoprotein E knockout and LDL receptor-deficient mice, and in hyperlipidemic primates.

The preclinical and clinical results obtained with AGI-1067 are also concordant with those in the Fukuoka AtheroSclerosis Trial (FAST) in which probucol induced regression of carotid atherosclerosis in 246 asymptomatic hypercholesterolemic patients (47). The incidence of major cardiac events was also significantly lower with probucol than in the control group in FAST (2.4% versus 13.6%; P<0.05). The results of the Probucol Quantitative Regression Swedish Trial (PQRST) appear to be discordant with those described here, but the design of that study raised several important issues (48). The primary end point of that study was lumen volume of femoral arteries using three-dimensional angiography, an approach rarely used in other clinical trials. The choice of the femoral location for assessment is also questionable in light of the preferential effect of probucol on younger lesions in the proximal thoracic aorta compared with the more advanced iliac lesions in experimental atherosclerosis in nonhuman primates (49). In addition, probucol was given to all patients (including those in the placebo group) for two months during the prerandomization phase, which represents another problematic design feature considering that probucol accumulates in tissues for prolonged periods.

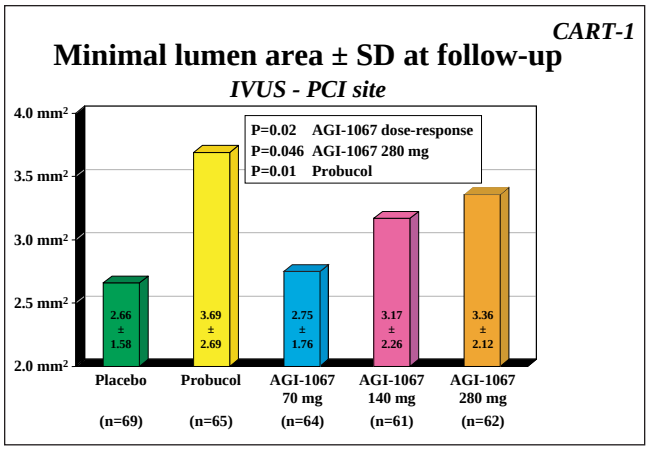

Figure 1) *Primary end point results in the Canadian Antioxidant Restenosis Trial-1 (CART-1). IVUS Intravascular ultrasound; PCI Percutaneous coronary intervention*

AGI-1067 was not different from placebo with respect to its effects on the QT interval after six weeks of therapy in CART-1, in contrast to probucol (46). This allows testing of the antioxidant/anti-inflammatory hypothesis with AGI-1067 in two other clinical trials.

RATIONALE AND DESIGN OF THE CART-2

AGI-1067 has been effective at preventing atherosclerosis in all tested animal models. The positive results observed with AGI-1067 in reference segments in CART-1 have also been described. Therefore, CART-2 assesses the value of AGI-1067 for the reduction of both post-PCI restenosis and atherosclerosis progression in non-PCI vessels after 12 months of treatment (measured with IVUS [50]). Based on preclinical and clinical data and also on its beneficial effects on prevention of LDL-lipid oxidation, blockage of monocyte adhesion and recruitment to inflamed endothelium through inhibition of endothelial adhesion molecule expression, it was hypothesized that AGI-1067 may significantly slow atherosclerosis progression or even induce regression in CART-2. Considering that oxidative stress and inflammation may persist for a prolonged period after stenting (51), treatment with AGI-1067 for the entire period of risk after PCI may result in enhanced protection against luminal renarrowing. This hypothesis is also tested in CART-2 (52).

RATIONALE AND DESIGN OF THE AGGRESSIVE REDUCTION OF INFLAMMATION STOPS EVENTS (ARISE) TRIAL

Despite improvements in imaging modalities, visualization of morphological details indicative of plaque stabilization is not yet possible with IVUS (50), and its predictive value for future clinical events is not yet definitely known. The greatest impact AGI-1067 may have clinically is the prevention of plaque rupture and subsequent cardiovascular morbidity and mortality. Modifications resulting from the administration of AGI-1067, which stabilize the plaque but do not result in overall plaque volume changes, are only evaluable through the conduct of a

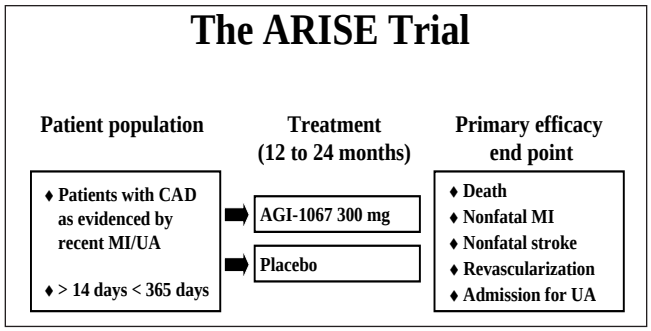

Figure 2) *Study design of the Aggressive Reduction of Inflammation Stops Events (ARISE) multinational, multicentre trial. CAD Coronary artery disease; MI Myocardial infarction; UA Unstable angina*

clinical events trial. Therefore, the ARISE trial is a necessary addition to the full evaluation of the potential utility of AGI-1067. Furthermore, ARISE represents a unique opportunity to test in a definitive fashion the antioxidant/anti-inflammatory hypothesis (53). The primary objective of ARISE is to determine whether long-term treatment with AGI-1067 will prevent major cardiovascular events in patients with coronary artery disease. Because the beneficial effects of AGI-1067 would likely be related to its potent antioxidant and anti-inflammatory properties, another key objective is to determine the effects of AGI-1067 on markers of inflammation and oxidation.

ARISE is a multicentre, double-blind, randomized, placebo-controlled trial involving more than 200 study sites (Figure 2). Patients with a recent diagnosis of coronary artery disease (unstable angina or myocardial infarction) and one additional risk factor are enrolled in the United States, Canada, Europe and South Africa. Patients are randomly assigned to AGI-1067 or placebo in a 1:1 ratio and treated for at least 12 months. The study will be complete when at least 990 patients have experienced a primary event. The primary study end point is the combined incidence of cardiovascular mortality, resuscitated cardiac arrest, nonfatal myocardial infarction, nonfatal stroke, need for coronary revascularization and urgent hospitalization for angina pectoris with objective evidence of ischemia.

CONCLUSION

Despite medical advancements made over the past decade, cardiovascular disease originating from atherosclerosis inflicts a large burden in terms of life expectancy, life quality and societal costs. Based on the aging of the population and trends toward environmental and lifestyle factors that increase the risk for atherosclerosis, it is anticipated that this burden will not dissipate in the near future. There is, therefore, a need for a pharmacological intervention that would provide further cardiovascular protection in patients with atherosclerosis over and above the protection offered by other medications available, including statins. In light of its favourable effects, ARISE is evaluating the value of AGI-1067 on clinical end points.

ACKNOWLEDGEMENT: Dr Tardif holds the Canadian Institutes of Health Research (CIHR) and Pfizer Chair in Atherosclerosis.

REFERENCES

1. Ross R. Atherosclerosis – An inflammatory disease. N Engl J Med 1999;340:115-26.
2. Kunsch C, Medford RM. Oxidative stress as a regulator of gene expression in the vasculature. Circ Res 1999;85:753-66.
3. Cushing SD, Berliner JA, Valente AJ, et al. Minimally modified low density lipoprotein induces monocyte chemotactic protein 1 in human endothelial cells and smooth muscle cells. Proc Natl Acad Sci USA 1990;87:5134-8.
4. Marui N, Offermann MK, Swerlick R, et al. Vascular cell adhesion molecule-1 (VCAM-1) gene transcription and expression are regulated through an antioxidant-sensitive mechanism in human vascular endothelial cells. J Clin Invest 1993;92:1866-74.
5. Rajavashisth TB, Andalibi A, Territo MC, et al. Induction of endothelial cell expression of granulocyte and macrophage colony-stimulating factors by modified low-density lipoproteins. Nature 1990;344:254-7.
6. Bourcier T, Sukhova G, Libby P. The nuclear factor kappa-B signaling pathway participates in dysregulation of vascular smooth muscle cells in vitro and in human atherosclerosis. J Biol Chem 1997;272:15817-24.
7. Thomas CE, Jackson RL, Ohlweiler DF, Ku G. Multiple lipid oxidation products in low density lipoproteins induce interleukin-1 beta release from human blood mononuclear cells. J Lipid Res 1994;35:417-27.
8. Rajagopalan S, Meng XP, Ramasamy S, Harrison DG, Galis ZS. Reactive oxygen species produced by macrophage-derived foam cells regulate the activity of vascular matrix metalloproteinases in vitro. Implications for atherosclerotic plaque stability. J Clin Invest 1996;98:2572-9.
9. Ikeda H, Koga Y, Oda T, et al. Free oxygen radicals contribute to platelet aggregation and cyclic flow variations in stenosed and endothelium-injured canine coronary arteries. J Am Coll Cardiol 1994;24:1749-56.
10. Roberts MJ, Young IS, Trouton TG, et al. Transient release of lipid peroxides after coronary artery balloon angioplasty. Lancet 1990;336:143-5.
11. Iuliano L, Pratico D, Greco C, et al. Angioplasty increases coronary sinus F2-isoprostane formation: Evidence for in vivo oxidative stress during PTCA. J Am Coll Cardiol 2001;37:76-80.
12. Kugiyama K, Kerns SA, Morrisett JD, Roberts R, Henry PD. Impairment of endothelium-dependent arterial relaxation by lysolecithin in modified low-density lipoproteins. Nature 1990;344:160-2.
13. Tardif J-C, Gregoire J, Bourassa MG. Antioxidants and restenosis – human studies. Tardif J-C, Bourassa MG, eds. In: Antioxidants and Cardiovascular Disease. Netherlands: Kluwer Academic Press, 2000:175-94.
14. Christen WG, Hennekens CH. Antioxidant vitamins and cardiovascular disease: Evidence from observational epidemiologic studies and randomized trials. Tardif J-C, Bourassa MG, eds. In: Antioxidants and Cardiovascular Disease. Netherlands: Kluwer Academic Press, 2000:135-44.
15. The Alpha-Tocopherol, Beta Carotene Cancer Prevention Study Group. The effect of vitamin E and beta carotene on the incidence of lung cancer and other cancers in male smokers. N Engl J Med 1994;330:1029-35.
16. Omenn GS, Goodman GE, Thornquist MD, et al. Effects of a combination of beta carotene and vitamin A on lung cancer and cardiovascular disease. N Engl J Med 1996;334:1150-5.
17. Stephens NG, Parsons A, Schofield PM, Kelly F, Cheeseman K, Mitchinson MJ. Randomized controlled trial of vitamin E in patients with coronary disease: Cambridge Heart Antioxidant Study (CHAOS). Lancet 1996;347:781-6.
18. Gruppo Italiano per lo Studio della Sopravivenza nell'Infarcto miocardico. Dietary supplementation with n-3 polyunsaturated fatty acids and vitamin E after myocardial infarction: Results of the GISSI-Prevenzione trial. Lancet 1999;354:447-55. (Erratum in 2001;357:642).
19. Yusuf S, Dagenais G, Pogue J, Bosch J, Sleight P. Vitamin E supplementation and cardiovascular events in high-risk patients. The Heart Outcomes Prevention Evaluation Study Investigators. N Engl J Med 2000;342:154-60.

20. MRC/BHF Heart Protection Study of cholesterol-lowering therapy and of antioxidant vitamin supplementation in a wide range of patients at increased risk of coronary heart disease death: Early safety and efficacy experience. Eur Heart J 1999;20:725-41.
21. Bowry VW, Ingold KU, Stocker R. Vitamin E in human low-density lipoprotein. When and how this antioxidant becomes a pro-oxidant. Biochem J 1992;288:341-4.
22. Keaney JF Jr, Gaziano JM, Xu A, et al. Low-dose alpha-tocopherol improves and high-dose alpha-tocopherol worsens endothelial vasodilator function in cholesterol-fed rabbits. J Clin Invest 1994;93:844-51.
23. Waters DD, Alderman EL, Hsia J, et al. Effects of hormone replacement therapy and antioxidant vitamin supplements on coronary atherosclerosis in postmenopausal women: A randomized controlled trial. JAMA 2002;288:2432-40.
24. Tardif J-C, Cote G, Lesperance J, et al. Probucol and multivitamins in the prevention of restenosis after coronary angioplasty. Multivitamins and Probucol Study Group. N Engl J Med 1997;337:365-72.
25. Brown BG, Zhao X-Q, Chait A, et al. Simvastatin and niacin, antioxidant vitamins, or the combination for the prevention of coronary disease. N Engl J Med 2001;345:1583-92.
26. Rodes J, Cote G, Lesperance J, et al. Prevention of restenosis after angioplasty in small coronary arteries with probucol. Circulation 1998;97:429-36.
27. Cote G, Tardif J-C, Lesperance J, et al. Effects of probucol on vascular remodeling after coronary angioplasty. Circulation 1999;99:30-5.
28. Ferns GA, Forster L, Stewart-Lee A, Konneh M, Nourooz-Zadeh J, Anggard EE. Probucol inhibits neointimal thickening and macrophage accumulation after balloon injury in the cholesterol-fed rabbit. Proc Natl Acad Sci USA 1992;89:11312-6.
29. Schneider JE, Berk BC, Gravanis MB, et al. Probucol decreases neointimal formation in a swine model of coronary artery balloon injury. A possible role for antioxidants in restenosis. Circulation 1993;88:628-37.
30. Freyschuss A, Stiko-Rahm A, Swedenborg J, et al. Antioxidant treatment inhibits the development of intimal thickening after balloon injury of the aorta in hypercholesterolemic rabbits. J Clin Invest 1993;91:1282-8.
31. Nunes GL, Sgoutas DS, Redden RA, et al. Combination of vitamins C and E alters the response to coronary balloon injury in the pig. Arterioscler Thromb Vasc Biol 1995;15:156-65.
32. Lafont A, Vinchon F, Addad F, et al. Antioxidants and restenosis: Animal studies. In: Tardif J-C, Bourassa MG, eds. Antioxidants and Cardiovascular Disease. Netherlands: Kluwer Academic Press, 2000:161-74.
33. Lau AK, Leichtweis SB, Hume P, et al. Probucol promotes functional reendothelialization in balloon-injured rabbit aortas. Circulation 2003;107:2031-6.
34. Setsuda M, Inden M, Hiraoka N, et al. Probucol therapy in the prevention of restenosis after successful percutaneous transluminal coronary angioplasty. Clin Ther 1993;15:374-82.
35. Watanabe K, Sekyia M, Ikeda S, Miyagawa M, Hashida K. Preventive effects of probucol on restenosis after percutaneous transluminal coronary angioplasty. Am Heart J 1996;132:23-9.
36. Lee YJ, Daida H, Yokoi H, et al. Effectiveness of probucol in preventing restenosis after percutaneous transluminal coronary angioplasty. Jpn Heart J 1996;37:327-32.
37. Tardif J-C, Cote G, Lesperance J, et al. Impact of residual plaque burden after balloon angioplasty in the MultiVitamins and Probucol (MVP) trial. Can J Cardiol 2001;17:49-55.
38. Yokoi H, Daida H, Kuwabara Y, et al. Effectiveness of an antioxidant in preventing restenosis after percutaneous transluminal coronary angioplasty: The Probucol Angioplasty Restenosis Trial. J Am Coll Cardiol 1997;30:855-62.
39. Tardif J-C, Gregoire J, Schwartz L, et al. Effects of AGI-1067 and probucol after percutaneous coronary interventions. Circulation 2003;107:552-8.
40. Meng CQ, Somers PK, Rachita CL, et al. Novel phenolic antioxidants as multifunctional inhibitors of inducible VCAM-1 expression for use in atherosclerosis. Bioorg Med Chem Lett 2002;12:2545-8.
41. Wasserman MA, Sundell CL, Kunsch C, Edwards D, Meng CQ, Medford RM. Chemistry and pharmacology of vascular protectants: A novel approach to the treatment of atherosclerosis and coronary artery disease. Am J Cardiol 2003;91(Suppl):34A-40A.
42. Kunsch C, Luchoomun J, Grey JY, et al. Selective inhibition of endothelial and monocyte redox-sensitive genes by AGI-1067: A novel antioxidant and anti-inflammatory agent. J Pharmacol Exp Ther 2004;308:820-9.
43. Sundell CL, Somers PK, Meng CQ, et al. AGI-1067: A multifunctional phenolic antioxidant, lipid modulator, anti-inflammatory and antiatherosclerotic agent. J Pharmacol Exp Ther 2003;305:1116-23.
44. Anderson TJ, Meredith IT, Yeung AC, Frei B, Selwyn AP, Ganz P. The effect of cholesterol-lowering and antioxidant therapy on endothelium-dependent coronary vasomotion. N Engl J Med 1995;332:488-93.
45. McLean LR, Thomas CE, Weintraub B, Hagaman. Modulation of the physical state of cellular cholesteryl esters by 4,4'-(isopropylidenedithio)bis(2,6-di-t-butylphenol) (probucol). J Biol Chem 1992;267:12291-8.
46. Tardif J-C. Clinical results with AGI-1067: A novel antioxidant vascular protectant. Am J Cardiol 2003;91(Suppl):41A-49A.
47. Sawayama Y, Shimizu C, Maeda N, et al. Effects of probucol and pravastatin on common carotid atherosclerosis in patients with asymptomatic hypercholesterolemia. Fukuoka Atherosclerosis Trial (FAST). J Am Coll Cardiol 2002;39:610-6.
48. Walldius G, Erikson U, Olsson AG, et al. The effect of probucol on femoral atherosclerosis: The Probucol Quantitative Regression Swedish Trial (PQRST). Am J Cardiol 1994;74:875-83.
49. Sasahara M, Raines EW, Chait A, et al. Inhibition of hypercholesterolemia-induced atherosclerosis in the non-human primate by probucol. I. Is the extent of atherosclerosis related to resistance of LDL to oxidation? J Clin Invest 1994;94:155-64.
50. Guedes A, Tardif J-C. Intravascular ultrasound assessment of atherosclerosis. Curr Atheroscler Rep 2004;6:219-24.
51. Nunes GL, Robinson K, Kalynych A, King SB III, Sgoutas DS, Berk BC. Vitamins C and E inhibit O^{2-} production in the pig coronary artery. Circulation 1997;96:3593-601.
52. Tardif J-C, Gregoire J, L'Allier PL. Prevention of restenosis with antioxidants: Mechanisms and implications. Am J Cardiovasc Drugs 2002;2:323-34.
53. Tardif J-C. Insights into oxidative stress and atherosclerosis. Can J Cardiol 2000;16(Suppl D):2D-4D.