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Symposium: Molecular Mechanisms of Protective Effects of Vitamin E in Atherosclerosis

Nonantioxidant Functions of α-Tocopherol in Smooth Muscle Cells[1,2]

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ABSTRACT Most tocopherols and tocotrienols, with the exception of α-tocopherol, are not retained by humans. This suggests that α-tocopherol is recognized uniquely; therefore, it may exert an exclusive function. α-Tocopherol possesses distinct properties that are independent of its prooxidant, antioxidant or radical-scavenging ability. α-Tocopherol specifically inhibits protein kinase C, the growth of certain cells and the transcription of the CD36 and collagenase genes. Activation events have also been seen on the protein phosphatase 2A (PP_2A) and on the expression of other genes (α-tropomyosin and connective tissue growth factor). Neither β-tocopherol nor probucol possessed the same specialty functions as α-tocopherol. Recently, we isolated a new ubiquitous cytosolic α-tocopherol binding protein (TAP). Its motifs suggest that it is a member of the hydrophobic ligand-binding protein family (CRAL-TRIO). TAP may also be involved in the regulation of cellular α-tocopherol concentration and α-tocopherol–mediated signaling. J. Nutr. 131: 378S–381S, 2001.

KEY WORDS: • *tocotrienols* • *tocopherols* • *cell signaling* • *α-tocopherol binding protein*

In 1922, Evans and Bishops named the animal nutritional factor essential of reproduction "Vitamin E" (1). Later, in the 1960s, it was associated with antioxidant function (2); nonoxidant properties were discovered 25 y after that (3,4). α-Tocopherol is the member of the vitamin E group (α-, β-, γ- and δ-tocopherols and tocotrienols) with the most biologically significant properties (5–9). Unlike others in the vitamin E group, α-tocopherol is found predominantly in mammalian tissue, rather than in plants (10–13).

When α-tocopherol is attacked by fatty acid peroxy radicals, it becomes, via one-electron oxidation, the α-tocopheryl radical; as a consequence of two-electron oxidation, it becomes α-tocopherylquinone. Under physiologic conditions, the reducing agents, ascorbic acid and lypoic acid, continuously repair oxidized α-tocopherol, thus preventing a loss of α-tocopherol–dependent cell-signaling events. If the rate of oxidation is greater than the rate of repair, α-tocopherol concentrations in the body will decrease.

Low levels of α-tocopherol have been associated with increased incidence of coronary artery disease. Conversely, increased intake of α-tocopherol has been shown to have protective effects against heart disease. Advances have been made in understanding the molecular basis of atherogenesis, elucidating functions of α-tocopherol beyond that of preventing LDL oxidation. We are on the verge of understanding the regulatory, nonoxidative response to α-tocopherol by crucial cells. Such responses include inhibition of smooth muscle cell proliferation, preservation of endothelial function, inhibition of monocyte-endothelial adhesion, inhibition of monocyte reactive oxygen species and cytokine release, and inhibition of platelet adhesion and aggregation.

These cellular responses to α-tocopherol are associated with transcriptional and post-transcriptional events. Activation of diacylglycerol kinase and protein phosphatase 2A (PP_2A),[4] and the inhibition of protein kinase C (PKC), cyclooxygenase, lipoxygenase and cytokine release by α-tocopherol are all examples of post-transcriptional regulation. α-Tocopherol also modulates the transcriptional regulation of a number of genes, including the liver collagen αI gene, the α-tocopherol transfer protein gene, the α-tropomyosin gene and the collagenase (metallo-proteinase 1) gene. In recent years, several reviews have reported on the action of α-tocopherol at the cellular level (14–20). This brief report will emphasize the nonantioxidant role of α-tocopherol in cellular modulation.

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[4] Abbreviations: IL, interleukin; PKC, protein kinase C; PP_2A, protein phosphatase 2A; TAP, tocopherol-associated protein.

Inhibition of PKC and associated cellular functions

Inhibition of PKC activity by α-tocopherol was discovered in 1991 to be the cause of the inhibition of vascular smooth muscle cell proliferation by α-tocopherol (3,4,21–25). Subsequent reports have confirmed that the inhibition of PKC by α-tocopherol occurs in different cell types such as monocytes, macrophages, neutrophils, fibroblasts and also mesangial cells (14,26–38). α-Tocopherol was also found to inhibit thrombin-induced PKC activation and endothelin secretion in edothelial cells; β-tocopherol did not have a similar ability (39). α-Tocopherol inhibits phorbol ester–induced shape changes in erythroleukemia cells (40), and also inhibits PKC-mediated neutrophil-superoxide generation (31). In animal models of atherosclerosis, PKC inhibition by α-tocopherol has also been demonstrated (41,42). α-Tocopherol inhibits PKC activity in a specific manner because β-tocopherol or Trolox (43) does not exert such an effect. α-Tocopherol also produces a significant decrease in monocyte superoxide anion release, lipid oxidation, and interleukin-1 (IL-1 β) release and adhesion to the endothelium. A similar antioxidant, β-tocopherol, had no effect on IL-1 β release (44). α-Tocopherol inhibits production of chemokines and inflammatory cytokines in addition to inhibition of adhesion of monocytes to human aortic endothelial cells by reducing the expression of adhesion molecules when cells are activated by inflammatory cytokines (45).

The proliferation and inhibition of PKC by a physiologic concentration of α-tocopherol are parallel events in vascular smooth muscle cells (46–48). β-Tocopherol is ineffective in either process and prevents the inhibitory effect of α-tocopherol. Because α-tocopherol and β-tocopherol have very similar radical-scavenging abilities, it is clear that the mechanism by which α-tocopherol acts on PKC is not related to these scavenging properties (49). Inhibition by α-tocopherol may be seen only at the cellular level and is not evident with recombinant PKC. The inhibitory effect of α-tocopherol on PKC can be correlated to a dephosphorylation of PKCα. PP_2A can be activated in vitro by treatment with α-tocopherol (50,51). This event may be crucial to the dephosphorylation of PKC and its subsequent decrease in activity.

α-Tocopherol has a PKC-mediated protective effect on human mesangial cells when exposed to high glucose concentrations (37). Our group observed a similar protective effect (50, 51). In the studies of King's group (37), PKC β-isoform expression was induced by high glucose. Interestingly, high glucose is concurrently responsible for an increase in diacylglycerol synthesis. It can be concluded that, although the mechanism of PKC inhibition by α-tocopherol has been interpreted differently in different laboratories and cellular systems, considerable agreement exists concerning the inhibition of PKC by α-tocopherol.

Transcriptional regulation of cellular reactions

α-Tocopherol concentration in organisms is dependent upon its uptake and destruction in radical reactions. Because of this, modulation of gene expression takes place as α-tocopherol concentrations increase or decrease (52). α-Tropomyosin expression is upregulated by α-tocopherol, but not by β-tocopherol, in rat vascular smooth muscle cells (53) in a reaction not mediated by PKC. Age-dependent increase of collagenase (MPP1) expression can be reduced by α-tocopherol (54) in human skin fibroblasts.

The liver α-tocopherol transfer protein and its mRNA are modulated by dietary vitamin E deficiencies in rats (55), and α- and β-tocopherol induce expression of hepatic α-tocopherol transfer protein mRNA (56,57). Scavenger receptors are also under α-tocopherol control. α-Tocopherol downregulates the activity of class A scavenger receptors in macrophages (58). Another scavenger receptor gene, CD36, is downregulated at the transcriptional level by α-tocopherol in macrophages and smooth muscles cells. β-Tocopherol, however, does not have this regulating ability (59). In conclusion, it appears that α-tocopherol is able to regulate the expression of a number of genes that are correlated with α-tocopherol–associated pathologies. To what extent these regulatory events are the direct consequence of the interaction of α-tocopherol with a receptor, a transcription factor or an element of the signal transduction pathways (e.g., PKC or phosphatase) remains a matter of investigation.

TABLE 1

Effect of α-tocopherol and β-tocopherol on protein kinase C-α phosphorylation state, autophosphorylating activity and activity towards Histone III-S[1]

	^{32}P-Protein kinase Cα	Autophosphorylating activity of protein kinase C-α	Histone activity	Cell proliferation
		%		
PMA[2]	100	100	100	100
α-Tocopherol	18.5	36.4	56.0	30
β-Tocopherol	74.1	84.9	79.0	90

[1] Source: Ref. 25.
[2] PMA, phorbol myristate acetate.

α-Tocopherol–associated protein (TAP)

Showing that α-tocopherol is involved in the regulation of several genes offers a very challenging opportunity for future studies. Here, only the existence of a common denominator (an α-tocopherol receptor protein, an α-tocopherol sensitive promoter element or an α-tocopherol sensitive transcription factor) has been postulated. Using molecular cloning into *Escherichia coli* and in vitro expression, we recently identified a human (hTAP) and bovine TAP (60). This protein appears to belong to a family of hydrophobic ligand-binding proteins, which all have the CRAL (*cis*-retinal binding motif) sequence in common. By using a biotinylated α-tocopherol derivative and the IASys resonant mirror biosensor, the purified recombinant protein was shown to bind tocopherol at a specific binding site with a K_d of 4.6×10^{-7} mol/L. Northern analysis shows that hTAP mRNA has a size of ~2.8k bp and is expressed ubiquitously. The highest amounts of hTAP message are found in the liver, brain and prostate. In conclusion, hTAP has significant sequence homology with proteins containing the CRAL-TRIO structural motif (RALBP, CRALBP, α-TTP, SEC 14, PTN 9, RSEC 45). TAP binds specifically to α-tocopherol and biotinylated tocopherol, suggesting the existence of a hydrophobic pocket possibly analogous to that of SEC14.

The newly discovered TAP is coded for in the human genome by three genes having slightly different 3′-sequences. The real function of these three genes products cannot be predicted precisely, but the very existence of three copies and their ubiquitous distribution point towards an important cellular role. Unbiased hypotheses may consider TAP a cellular binding or interorganelle transport protein, although the possibility of the identification of TAP with a cell receptor, a coreceptor or a transcription factor modulator cannot be un-

TABLE 2

The growth inhibitory effect of α-tocopherol on different cell lines

Cell lines	Sensitivity	Origin
A10	Yes	Rat aorta smooth muscle
A7r5	Yes	Rat aorta smooth muscle
T/G	Yes	Human aorta smooth muscle
NB2A	Yes	Mouse neuroblastoma
Balb/3T3	Yes	Mouse fibroblast
Human fibroblast	Yes	Primary cell lines
DU-145, PC-3	Yes	Human prostate cancer
LNCaP	Yes	Human prostate cancer (androgen sensitive)
Human pigmented retinal epithelial cells	Yes	hPRE
Human leukaemia	Yes	U937
Mouse fibroblast	Yes	Balb/c-3T3
Glioma	Yes	C6
P388 D1	No	Mouse monocyte macrophage
LR73	No	Chinese hamster ovary
Saos-2	No	Human osteosarcoma
Human hepatocarcinoma	No	HepG2
Human colon adenocarcinoma	No	CaCo2

derestimated. Coprecipitation experiments and two hybrid studies in progress in our laboratory may give indications, by nearest-neighbor protein interactions, concerning the function of these new cellular tocopherol binding proteins.

LITERATURE CITED

1. Evans H. M. & Bishop, B.K.S. (1922) Fetal resorption. Science (Washington, DC) 55: 650.
2. Epstein, S. S., Forsyth, J., Saporoschetz, I. B. & Mantel, N. (1966) An exploratory investigation on the inhibition of selected photosensitizers by agents of varying antioxidant activity. Radiat. Res. 28: 322–335.
3. Boscoboinik, D., Szewczyk, A. & Azzi, A. (1991) α-Tocopherol (vitamin E) regulates vascular smooth muscle cell proliferation and protein kinase C activity. Arch. Biochem. Biophys. 286: 264–269.
4. Boscoboinik, D., Szewczyk, A., Hensey, C. & Azzi, A. (1991) Inhibition of cell proliferation by α-tocopherol. Role of protein kinase C. J. Biol. Chem. 266: 6188–6194.
5. Ingold, K. U., Burton, G. W., Foster, D. O., Hughes, L., Lindsay, D. A. & Webb, A. (1987) Biokinetics of and discrimination between dietary *RRR*- and *SRR*-α-tocopherols in the male rat. Lipids 22: 163–172.
6. Kiyose, C., Muramatsu, R., Kameyama, Y., Ueda, T. & Igarashi, O. (1997) Biodiscrimination of α-tocopherol stereoisomers in humans after oral administration [see comments]. Am. J. Clin. Nutr. 65: 785–789.
7. Weiser, H. & Vecchi, M. (1981) Stereoisomers of α-tocopheryl acetate—characterization of the samples by physico-chemical methods and determination of biological activities in the rat resorption-gestation test. Int. J. Vitam. Nutr. Res. 51: 100–113.
8. Weiser, H., Vecchi, M. & Schlachter, M. (1986) Stereoisomers of α-tocopheryl acetate. IV. USP units and α-tocopherol equivalents of all-*rac*-, 2-*ambo*- and *RRR*-α-tocopherol evaluated by simultaneous determination of resorption-gestation, myopathy and liver storage capacity in rats. Int. J. Vitam. Nutr. Res. 56: 45–56.
9. Weber, P., Bendich, A. & Machlin, L. J. (1997) Vitamin E and human health: rationale for determining recommended intake levels. Nutrition 13: 450–460.
10. Netscher, T. (1996) Stereoisomers of tocopherols—synthesis and analytics. Chimia 50: 563–567.
11. Bauernfeind, J. C. (1977) The tocopherol content of food and influencing factors. CRC Crit. Rev. Food Sci. Nutr. 8: 337–382.
12. Threlfall, D. R. (1971) The biosynthesis of vitamins E and K and related compounds. Vitam. Horm. 29: 153–200.
13. Bauernfeind, J. B. (1980) In: Vitamin E: A Comprehensive Treatise, pp. 99–167, Marcel Dekker, Inc., New York, NY.
14. Devaraj, S. & Jialal, I. (1998) The effects of α-tocopherol on critical cells in atherogenesis. Curr. Opin. Lipidol. 9: 11–15.
15. Gokce, N. & Frei, B. (1996) Basic research in antioxidant inhibition of steps in atherogenesis. J. Cardiovasc. Risk 3: 352–357.
16. Frei, B. (1999) On the role of vitamin C and other antioxidants in atherogenesis and vascular dysfunction. Proc. Soc. Exp. Biol. Med. 222: 196–204.
17. Steiner, M. (1993) Vitamin E: more than an antioxidant. Clin. Cardiol. 16: I16–118.
18. Wu, H. P., Tai, T. Y., Chuang, L. M., Lin, B. J., Wang, J. D. & Teng, C. M. (1992) Effect of tocopherol on platelet aggregation in non-insulin-dependent diabetes mellitus: ex vivo and in vitro studies. J. Formos. Med. Assoc. 91: 270–275.
19. Steiner, M. (1991) Influence of vitamin E on platelet function in humans. J. Am. Coll. Nutr. 10: 466–473.
20. Traber, M. & Packer, L. (1995) Vitamin E: beyond antioxidant function. Am. J. Clin. Nutr. 62: 1501S–1509S.
21. Azzi, A., Boscoboinik, D. & Hensey, C. (1992) The protein kinase C family. Eur. J. Biochem. 208: 547–557.
22. Azzi, A., Boscoboinik, D., Chatelain, E., Özer, N. K. & Stäuble, B. (1993) *d*-α-Tocopherol control of cell proliferation. Mol. Asp. Med. 14: 265–271.
23. Chatelain, E., Boscoboinik, D. O., Bartoli, G. M., Kagan, V. E., Gey, F. K., Packer, L. & Azzi, A. (1993) Inhibition of smooth muscle cell proliferation and protein kinase C activity by tocopherols and tocotrienols. Biochim. Biophys. Acta 1176: 83–89.
24. Boscoboinik, D., Chatelain, E., Bartoli, G. M. & Azzi, A. (1992) In: Free Radicals and Aging (Emerit, I. & Chance, B., eds.), pp. 164–177. Birkhäuser Verlag, Basel, Switzerland.
25. Tasinato, A., Boscoboinik, D., Bartoli, G. M., Maroni, P. & Azzi, A. (1995) *d*-α-Tocopherol inhibition of vascular smooth muscle cell proliferation occurs at physiological concentrations, correlates with protein kinase C inhibition, and is independent of its antioxidant properties. Proc. Natl. Acad. Sci. U.S.A. 92: 12190–12194.
26. Devaraj, S., Li, D. & Jialal, I. (1996) The effects of alpha tocopherol supplementation on monocyte function. Decreased lipid oxidation, interleukin 1 beta secretion, and monocyte adhesion to endothelium. J. Clin. Investig. 98: 756–763.
27. Devaraj, S., Adams-Huet, B., Fuller, C. J. & Jialal, I. (1997) Dose-response comparison of *RRR*-α-tocopherol and all-racemic α-tocopherol on LDL oxidation. Arterioscler. Thromb. Vasc. Biol. 17: 2273–2279.
28. Freedman, J. E., Farhat, J. H., Loscalzo, J. & Keaney, J. F., Jr. (1996) α-Tocopherol inhibits aggregation of human platelets by a protein kinase C-dependent mechanism. Circulation 94: 2434–2440.
29. Hehenberger, K. & Hansson, A. (1997) High glucose-induced growth factor resistance in human fibroblasts can be reversed by antioxidants and protein kinase C-inhibitors. Cell Biochem. Funct. 15: 197–201.
30. Kanno, T., Utsumi, T., Kobuchi, H., Takehara, Y., Akiyama, J., Yoshioka, T., Horton, A. A. & Utsumi, K. (1995) Inhibition of stimulus-specific neutrophil superoxide generation by α-tocopherol. Free Radic. Res. 22: 431–440.
31. Kanno, T., Utsumi, T., Takehara, Y., Ide, A., Akiyama, J., Yoshioka, T., Horton, A. A. & Utsumi, K. (1996) Inhibition of neutrophil-superoxide generation by α-tocopherol and coenzyme Q. Free Radic. Res. 24: 281–289.
32. Okada, S., Takehara, Y., Yabuki, M., Yoshioka, T., Yasuda, T., Inoue, M. & Utsumi, K. (1996) Rapid reduction of nitric oxide by mitochondria, and reversible inhibition of mitochondrial respiration by nitric oxide. Biochem. J. 315: 295–299.
33. Koya, D., Lee, I. K., Ishii, H., Kanoh, H. & King, G. L. (1997) Prevention of glomerular dysfunction in diabetic rats by treatment with *d*-α-tocopherol. J. Am. Soc. Nephrol. 8: 426–435.
34. Rattan, V., Sultana, C., Shen, Y. M. & Kalra, V. K. (1997) Oxidant stress-induced transendothelial migration of monocytes is linked to phosphorylation of PECAM-1. Am. J. Physiol. 273: E453–E461.
35. Skolnick, A. A. (1997) Novel therapies to prevent diabetic retinopathy [news]. J. Am. Med. Assoc. 278: 1480–1481.
36. Studer, R. K., Craven, P. A. & DeRubertis, F. R. (1997) Antioxidant inhibition of protein kinase C-signaled increases in transforming growth factor-β in mesangial cells. Metabolism 46: 918–925.
37. Tada, H., Ishii, H. & Isogai, S. (1997) Protective effect of *d*-α-tocopherol on the function of human mesangial cells exposed to high glucose concentrations. Metabolism 46: 779–784.
38. Takehara, Y., Kanno, T., Yoshioka, T., Inoue, M. & Utsumi, K. (1995) Oxygen-dependent regulation of mitochondrial energy metabolism by nitric oxide. Arch. Biochem. Biophys. 323: 27–32.
39. Martin-Nizard, F., Boullier, A., Fruchart, J. C. & Duriez, P. (1998) α-Tocopherol but not β-tocopherol inhibits thrombin-induced PKC activation and endothelin secretion in endothelial cells. J. Cardiovasc. Risk 5: 339–345.
40. Steiner, M., Li, W., Ciaramella, J. M., Anagnostou, A. & Sigounas, G. (1997) *dl*-α-Tocopherol, a potent inhibitor of phorbol ester induced shape change of erythro- and megakaryoblastic leukemia cells. J. Cell. Physiol. 172: 351–360.
41. Özer, N. K., Sirikci, Ö., Taha, S., San, T., Moser, U. & Azzi, A. (1998) Effect of vitamin E and probucol on dietary cholesterol-induced atherosclerosis in rabbits. Free Radic. Biol. Med. 24: 226–233.
42. Sirikci, Ö., Özer, N. K. & Azzi, A. (1996) Dietary cholesterol-induced changes of protein kinase C and the effect of vitamin E in rabbit aortic smooth muscle cells. Atherosclerosis 126: 253–263.

43. Cachia, O., El Benna, J., Pedruzzi, E., Descomps, B., Gougerot-Pocidalo, M. A. & Leger, C. L. (1998) α-Tocopherol inhibits the respiratory burst in human monocytes—attenuation of p47phox membrane translocation and phosphorylation. J. Biol. Chem. 273: 32801–32805.

44. Devaraj, S. & Jialal, I. (1999) α-Tocopherol decreases interleukin-1β release from activated human monocytes by inhibition of 5-lipoxygenase. Arterioscler. Thromb. Vasc. Biol. 19: 1125–1133.

45. Wu, D., Koga, T., Martin, K. R. & Meydani, M. (1999) Effect of vitamin E on human aortic endothelial cell production of chemokines and adhesion to monocytes. Atherosclerosis 147: 297–307.

46. Gey, K. F. (1990) The antioxidant hypothesis of cardiovascular disease: epidemiology and mechanisms. Biochem. Soc. Trans. 18: 1041–1045.

47. Schultz, M., Leist, M., Petrzika, M., Gassmann, B. & Brigelius-Flohé, R. (1995) Novel urinary metabolite of α-tocopherol, 2,5,7,8-tetramethyl- 2(2′-carboxyethyl)-6-hydroxychroman, as an indicator of an adequate vitamin E supply. Am. J. Clin. Nutr. 62 (suppl.): 1527S–1534S.

48. Traber, M. G., Ramakrishnan, R. & Kayden, H. J. (1994) Human plasma vitamin E kinetics demonstrate rapid recycling of plasma *RRR*-α-tocopherol. Proc. Natl. Acad. Sci. U.S.A. 91: 10005–10008.

49. Pryor, A. W., Cornicelli, J. A., Devall, L. J., Tait, B., Trivedi, B. K., Witiak, D. T. & Wu, M. (1993) A rapid screening test to determine the antioxidant potencies of natural and synthetic antioxidants. J. Org. Chem. 58: 3521–3532.

50. Ricciarelli, R., Tasinato, A., Clément, S., Özer, N. K., Boscoboinik, D. & Azzi, A. (1998) α-Tocopherol specifically inactivates cellular protein kinase C alpha by changing its phosphorylation state. Biochem. J. 334: 243–249.

51. Clement, S., Tasinato, A., Boscoboinik, D. & Azzi, A. (1997) The effect of α-tocopherol on the synthesis, phosphorylation and activity of protein kinase C in smooth muscle cells after phorbol 12-myristate 13-acetate down-regulation. Eur. J. Biochem. 246: 745–749.

52. Azzi, A., Boscoboinik, D., Fazzio, A., Marilley, D., Maroni, P., Ozer, N. K., Spycher, S. & Tasinato, A. (1998) *RRR*-α-Tocopherol regulation of gene transcription in response to the cell oxidant status. Z. Ernaehrwiss. 37: 21–28.

53. Aratri, E., Spycher, S. E., Breyer, I. & Azzi, A. (1999) Modulation of α-tropomyosin expression by α-tocopherol in rat vascular smooth muscle cells. FEBS Lett. 447: 91–94.

54. Ricciarelli, R., Maroni, P., Ozer, N., Zingg, J. & Azzi, A. (1999) Age-dependent increase of collagenase expression can be reduced by α-tocopherol via protein kinase C inhibition. Free Radic. Biol. Med. 27: 729–737.

55. Shaw, H. M. & Huang, C. (1998) Liver α-tocopherol transfer protein and its mRNA are differentially altered by dietary vitamin E deficiency and protein insufficiency in rats. J. Nutr. 128: 2348–2354.

56. Fechner, H., Schlame, M., Guthmann, F., Stevens, P. A. & Rustow, B. (1998) α- and δ-Tocopherol induce expression of hepatic α- tocopherol-transfer-protein mRNA. Biochem. J. 331: 577–581.

57. Kim, H. S., Arai, H., Arita, M., Sato, Y., Ogihara, T., Inoue, K., Mino, M. & Tamai, H. (1998) Effect of α-tocopherol status on α-tocopherol transfer protein expression and its messenger RNA level in rat liver. Free Radic. Res. 28: 87–92.

58. Teupser, D., Thiery, J. & Seidel, D. (1999) α-Tocopherol down-regulates scavenger receptor activity in macrophages. Atherosclerosis 144: 109–115.

59. Ricciarelli, R., Zingg, J.-M. & Azzi, A. (2000) Downregulation of the CD36 scavenger receptor by α-tocopherol. Circulation 102: 82–87.

60. Stocker, A., Zimmer, S., Spycher, S. E. & Azzi, A. (1999) Identification of a novel cytosolic tocopherol-binding protein: structure, specificity, and tissue distribution. JUBMB 48: 49–55.