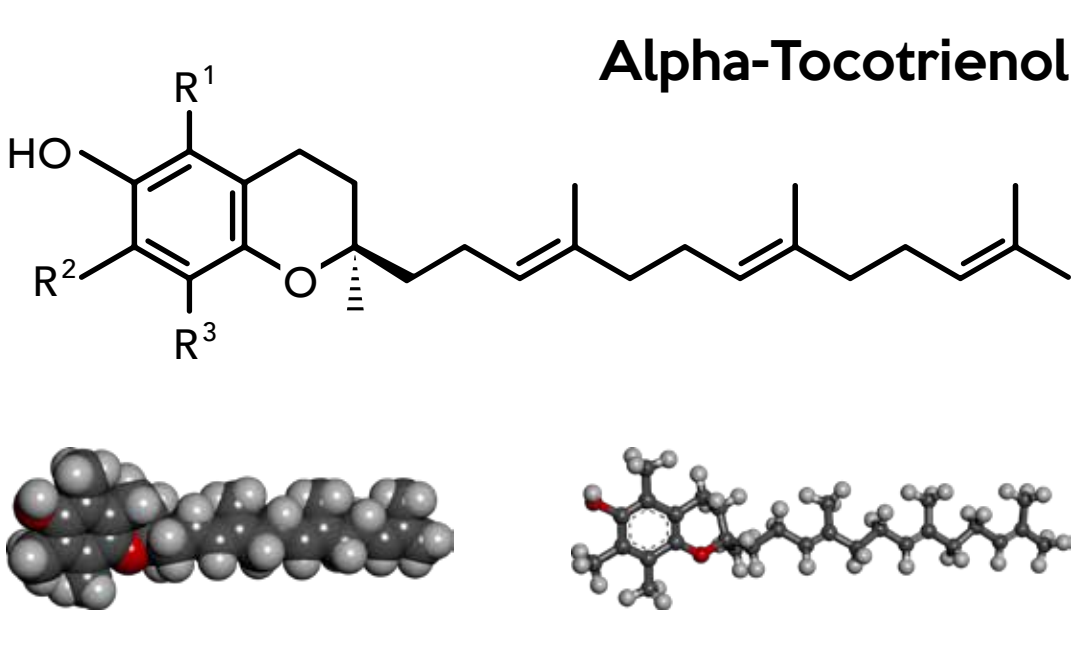
Tocotrienols, the Vitamin E of the 21st Century:

It's Potential Against Abnormal cells and Other Chronic Diseases

David Terrero, Phd
Scientific Advisor
Bixahuman Lab

Alpha-Tocotrienol



ABSTRACT

Initially discovered in 1938 as a "fertility factor," vitamin E now refers to eight different isoforms that belong to two categories, four saturated analogues (α, β, γ, and δ) called tocopherols and four unsaturated analogues referred to as tocotrienols. While the tocopherols have been investigated extensively, little is known about the tocotrienols. Very limited studies suggest that both the molecular and therapeutic targets of the tocotrienols are distinct from those of the tocopherols. For instance, suppression of inflammatory transcription factor NF-κB, which is closely linked to tumorigenesis and inhibition of HMG-CoA reductase, mammalian DNA polymerases and certain protein tyrosine kinases, is unique to the tocotrienols. This review examines in detail the molecular targets of the tocotrienols and their roles in cancer, bone resorption, diabetes, and cardiovascular and neurological diseases at both preclinical and clinical levels. As disappointment with the therapeutic value of the tocopherols grows, the potential of these novel vitamin E analogues awaits further investigation.

Keywords tocotrienols; anticancer; cholesterol; atherosclerosis; diabetes; bone resorption; neuroprotective

1. Introduction

Preventing beriberi by eating unpolished rice, curing scurvy by eating citrus fruits, and supporting fertility by eating leafy vegetables—all of these life-sustaining properties of foods are related to factors that in 1912 came to be called vitamins (vita means life). In 1922, Herbert Evans and Katherine Bishop, two prominent researchers from Berkeley, first isolated fatsoluble vitamin E from green leafy vegetables and described it as a fertility factor. Vitamin E was named tocopherol in 1924 and synthesized in 1938 [for references, see [1]]. Deficiency of this vitamin is now known to cause severe degenerative diseases such as ataxia, Duchenne muscular dystrophy-like muscle degeneration, and infertility. Vitamin E is present in most edible oils to various extents, including those extracted from wheat germ oil, wheat, rice bran (0.035%), barley (0.012% or 44 mg/g oil), oats (0.03%), coconut (0.019%) and palm (0.044%; 0.78–1.08 mg/g oil) (http://www.tocotrienol.org/sources.htm).

While alpha-tocopherol was the first vitamin E analogue to be recognized, eight chemically distinct analogues are now known, consisting of alpha (α), beta (β), gamma (γ) and delta (δ)tocopherols (TP) and alpha, beta, gamma and delta-tocotrienols (T3); all of them are referred to as vitamin E (Fig. 1). The tocopherols are saturated forms of vitamin E, whereas the tocotrienols are unsaturated and possess an isoprenoid side chain. Some evidence suggests that human tissues can convert tocotrienols to tocopherols [2,3]. Tocopherols consist of a chromanol ring and a 15-carbon tail. The presence of three trans double bonds in the tail distinguishes tocopherols from tocotrienols. The isomeric forms of tocotrienol are distinguished by the number and location of methyl groups on the chromanol rings: αtocotrienol is 5, 7, 8, trimethyl; β-tocotrienol is 5, 8, dimethyl; γ-tocotrienol is 7, 8, dimethyl and δ-tocotrienol is 8, monomethyl. While leaves and seeds of most plants contain tocopherols, tocotrienols are present in only a very small fraction of plants (Fig. 2a, b). Although some activities of tocopherols and tocotrienols are compared in this review, tocotrienols are the primary focus.

The name tocotrienol to denote a tocopherol with a true isoprenoid side chain was first suggested by Bunyan et al [4], and the tocotrienols were described in Nature when isolated from the latex of the rubber plant, Havea brasiliensis, in 1964 [5,6]. The tocotrienols attracted no real attention until the 1980s and 1990s when their cholesterol-lowering potential [7] and anticancer effects were described [8,9]. Subsequently, rice bran, palm, and annatto (90% delta and 10% gamma) oils were described as some of the richest sources of tocotrienols by Tan and his coworkers. The tocopherols: tocotrienols ratios in rice bran, palm and annatto oils are 50:50; 25:75 and 0.1:99.9, respectively [10]. Besides tocopherols, various isomers of tocotrienols have also been detected in human milk [11].

2. Molecular Targets

Like tocopherols, tocotrienols exhibit antioxidant activities, and most of its effects can be linked to its antioxidant function. Molecular targets of tocotrienols can be classified as those that are modulated by binding directly[12–17] and those that are modulated indirectly. Modulation of various targets by tocotrienols may occur at the transcriptional, translational, or post-translational levels, or by direct interactions with cellular targets (Table 1). For instance, src and 3-hydroxy-3-methyl-glutaryl coenzyme A (HMG-CoA) reductase are modulated through direct binding, whereas inflammatory transcription factors and the genes regulated by them and death receptors are modulated indirectly (Fig. 3a, b). Various studies indicate that tocotrienols exhibit antioxidant, antiproliferative, antisurvival, proapoptotic, antiangiogenic, and anti-inflammatory activities.

The antioxidant activities of this vitamin E (tocotrienols) are mediated through induction of antioxidant enzymes such as superoxide dismutase [18,19], NADPH:quinone oxidoreductase [20], and glutathione peroxidase [21], which quench free radicals such as superoxide radicals [22] (Table 1). The antiproliferative activity of tocotrienols are mediated through modulation of growth factors such as vascular endothelial growth factor (VEGF)[23], basic fibroblast growth factor (bFGF) [24] and transforming growth factor beta (TGF-β) [25], HER2/neu [26], and interleukin-6 (IL-6) [27]. Cyclin-dependent kinases (CDK2, CDK4, CDK6) and their inhibitors, such as p21, p27 and p53 [28,29] and downregulation of Rb phosphorylation [29– 31] also mediate the growth-suppressive effects of this agent. Moreover, inhibition of mitogenactivated protein kinases (MAPK) such as ERK [32], p38 MAPK and JNK [33] is critical to the antiproliferative effects of tocotrienols. The suppression of cyclin D1 expression induced by tocotrienols also plays an important role in the growth-inhibitory activities of this vitamin [20,29–31,34,35]. Tocotrienols impede the survival of various tumor cells by inhibiting expression of cell survival proteins such as XIAP, IAP-1, IAP-2, bcl-2, bcl-xl, cFLIP, TRAF-1, survivin and Bfl-1/A1 [34]. Suppression of the phosphotidyl-inositol-3-kinase (PI3K)/AKT pathway by tocotrienols could account for its antisurvival activities [36]. Downregulation of the telomerase, c-myc, and raf-ERK signaling pathways has been linked to tocotrienol's ability to inhibit cell survival [32,37].

Various studies have revealed that tocotrienols can induce apoptosis in a wide variety of tumor cells. These effects are mediated through activation of both extrinsic and intrinsic pathways by the vitamin. The extrinsic pathways involve induction of death receptors [33] and activation of caspase-8, which leads to caspase-3 activation [38]. The activation of intrinsic pathways by tocotrienols involves mitochondrial depolarization [39] and is mediated through the upregulation of Bax [32,40,41], cleavage of Bid [40], release of cytochrome C [22,28,39,42, 43], and activation of caspase-9, which in turn leads to activation of caspase-3 [28,40,44,45]. This unsaturated form of vitamin E also mediates apoptosis through DNA fragmentation [39, 46] and upregulation of p53 [28] in certain cells.

The suppression of angiogenesis by tocotrienols is mediated through inhibition of VEGF expression [23,47] and VEGF receptor signaling [48–50]. Suppression of the matrix metalloproteinase (MMP)-9 gene could also contribute to the angiogenesis-suppressive activity [34,51]. Although TWIST, CXCR4, TNF, FGF, TGF-β, PDGF and IL-8 all have been linked with angiogenesis, whether any of these pathways is modulated by tocotrienols [47] is poorly understood.

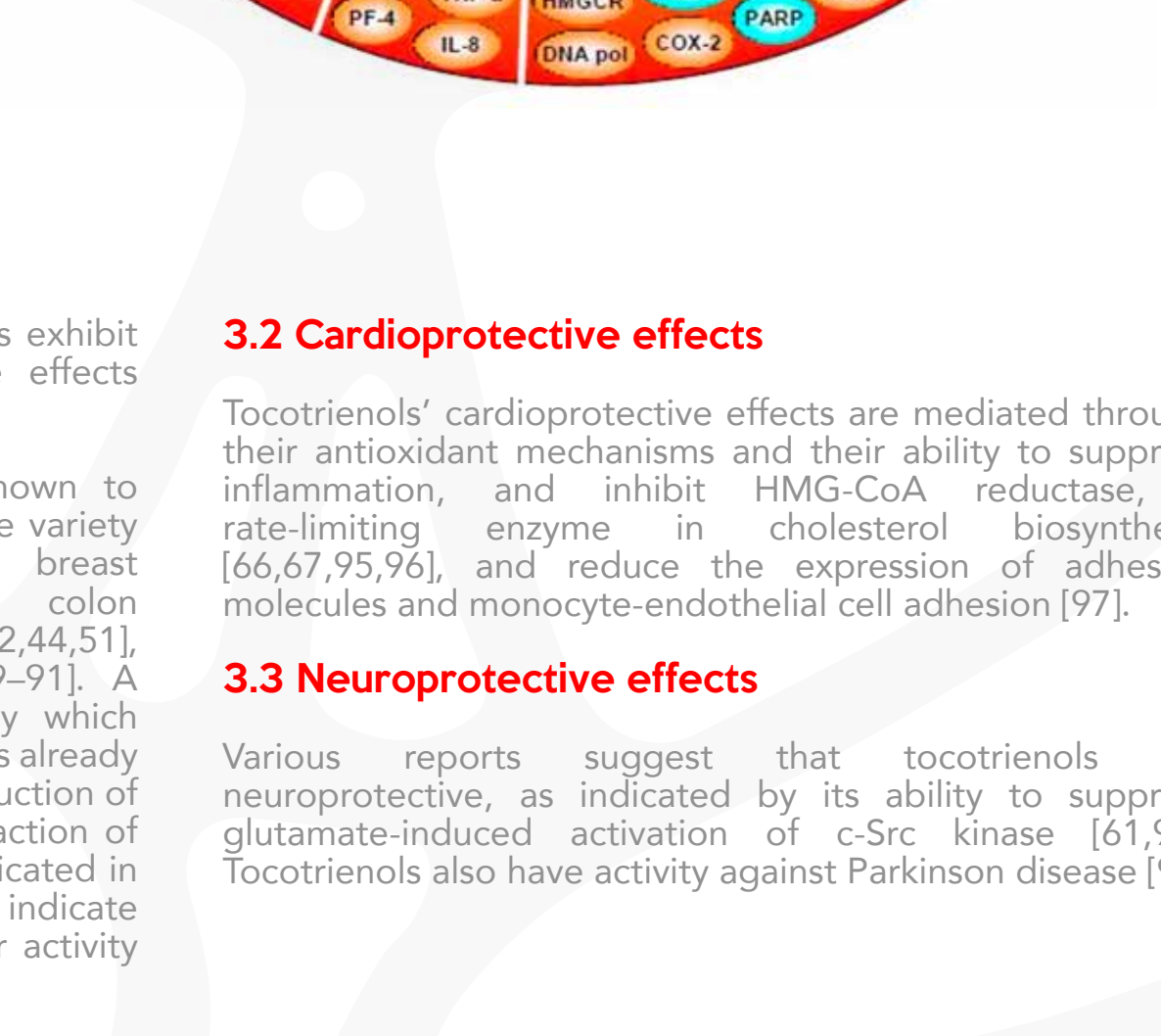
Numerous lines of evidence suggest that tocotrienols exhibit potent anti-inflammatory activity. First, activation of the transcription factor NF-κB has been closely linked with inflammation [34,52]. Second, tocotrienols has been shown to suppress the expression of TNF [34,53], IL-1 [54], IL-6 [55], IL-8 [47], inducible nitric oxide synthase [56], and cyclo-oxygenase 2 [34, 52,56,57], all of which mediate inflammation. Third, tocotrienols has been shown to suppress STAT3 cell signaling pathway, also involved in inflammation [58,59]. Hypoxia-induced factor-1 is another pathway that has been linked with inflammation and is modulated by tocotrienols [47].

Tocotrienols inhibit various protein kinases, including protein kinase C [60,61], p60 Src [61], IκBα kinase [62], and GSK-3β [38]. Inhibition of HMG-CoA reductase, an enzyme that is rate limiting in the pathway to cholesterol biosynthesis [63], also plays an essential role in the various activities attributed to this vitamin. There are, for instance, reports that the antitumor effects of tocotrienols are linked to its ability to inhibit HMG-CoA reductase [64,65]. Different isomeric forms of tocotrienols vary in their ability to lower cholesterol, as follows: δ > γ > α > β [66]. The reduction of HMG-CoA occurs through two separate mechanisms, first the enhancement of degradation of the reductase protein and second the decrease in efficiency of translation of the reductase mRNA [66,67].

The modification by tocotrienols of various cell-signaling pathways described here has been linked to its effects against cancer, diabetes, and cardiovascular and neurological diseases.

≋ BIXA

Molecular targets of tocotrienols



3. In Vitro Studies

Numerous in vitro studies indicate that tocotrienols exhibit anticancer, cardioprotective, and neuroprotective effects (Table 2).

3.1 Anticancer effects Tocotrienols has been shown to suppress proliferation and induce apoptosis in wide variety of tumor cells including those of the breast [13,25,26,29,30,33,36,42,43,45,52,59,62,68–78], colon [28,41,79], liver [40,80–82], lung[83–85], stomach [32,44,51], skin [86,87], pancreas [88], and prostate [46,89–91]. A number of mechanisms have been proposed by which tocotrienols induce apoptosis in these cancer cells, as already described. Some additional mechanisms involve induction of death receptor-5, as described recently [33]. Interaction of tocotrienols with estrogen receptors has been implicated in studies of breast cancer cells [73,92]. Various results indicate that γ- and δ-tocotrienol exhibit greater anticancer activity than α- or βtocotrienol [27,75,82,89,93,94].

3.2 Cardioprotective effects

Tocotrienols' cardioprotective effects are mediated through their antioxidant mechanisms and their ability to suppress inflammation, and inhibit HMG-CoA reductase, a rate-limiting enzyme in cholesterol biosynthesis [66,67,95,96], and reduce the expression of adhesion molecules and monocyte-endothelial cell adhesion [97].

3.3 Neuroprotective effects

Various reports suggest that tocotrienols are neuroprotective, as indicated by its ability to suppress glutamate-induced activation of c-Src kinase [61,98]. Tocotrienols also have activity against Parkinson disease [99]

4. Animal Studies with tocotrienols

4.1 Anticancer effects

Tocotrienols exhibit activity in different models of both prevention and treatment of cancer (Table 3). Perhaps the first report about the therapeutic potential of tocotrienols for cancer in animal models was by Kato et al., who in 1985 showed that tumor-bearing rats administered with tocotrienols had an extended life span [8]. Komiyama et al. observed antitumor activity when tocotrienols were administered intraperitoneally to mice with established murine Meth A fibrosarcoma. They showed that tocotrienols were more effective than α-tocopherol, and among the tocotrienols, γ-tocotrienol was more effective than α-tocotrienol as an antitumor agent [100]. They also showed that tocotrienols are better antioxidants than tocopherols. The growth of highly metastatic B16 melanoma in female mice was inhibited by tocotrienols, and δ-tocotrienol was more active than γ-tocotrienol in this setting [101]. In mice implanted with hepatoma, both γ-tocotrienol and δ-tocotrienol delayed tumor growth, and when examined for levels of tocotrienols, the tumors contained a specific accumulation of these analogues [102].

The antitumor effects of tocotrienols appear to be mediated in part through their ability to suppress angiogenesis [23,103]. Suppression of angiogenesis is mediated through reduction in serum levels of VEGF and inhibition of the PI3K-AKT pathway. The inhibition of HMG-CoA reductase and the consequent decrease in serum cholesterol level has been linked with the tumor-suppressive action of tocotrienols [64]. Tocotrienols have also been shown to enhance the antitumor effects of other agents. In one study, δ-tocotrienol was reported to enhance the growth-suppressive effects of lovastatin in the B16 melanoma model in mice [87]. γTocotrienol preferentially sensitized human prostate cancer in nude mice to radiation [104].

Besides antitumor effects against established tumors, tocotrienols have also been shown to be effective in cancer prevention models. Sundram et al. showed that palm oil, one of the richest dietary sources of tocotrienols, is effective in preventing 7,12-dimethylbenz[α]anthracene (DMBA)-induced mammary carcinogenesis in rats, but corn oil and soybean oil, which contain tocopherols but not tocotrienols, lack this activity [9]. Gould et al. reported a statistically significant increase in tumor latency in the DMBA-induced rat mammary tumor model with tocotrienols but not with tocopherols [105]. Inhibition of tumor promotion by various palmoil tocotrienols was also reported by Goh and his colleagues [106], in an in vitro assay utilizing the activation of Epstein-Barr virus (EBV) early antigen expression in EBV-genome–carrying human lymphoblastoid cells. They showed that γ- and δ-tocotrienol derived from palm oil exhibit strong activity against tumor promotion by inhibiting EBV early antigen expression in Raji cells induced by phorbol ester. However, α- and γ-tocopherol and dimers of γ-tocotrienol or γ-tocopherol lack this activity [98]. Iqbal et al. showed that feeding tocotrienol-rich fraction (TRF; 10 mg/kg) to DMBA-administered rats suppressed mammary carcinogenesis, and this correlated with declines in serum cholesterol, low-density lipoprotein (LDL)-cholesterol, and HMG CoA reductase protein [65]. Wada et al. examined the effect of 0.05% oral tocotrienols on spontaneous liver carcinogenesis in male mice and on glycerol-induced lung tumor promotion in male mice initiated with 4-nitroquinolone 1-oxide [82]. Incidence of liver and lung tumors was almost 80% lower in treated animals than in untreated animals.

Tocotrienols has been shown to prevent chemical-induced carcinogenesis of the liver [107] and found to suppress 2-acetylaminofluorene (AAF)–induced hepatocarcinogenesis [108]. In another study, Rahmat et al. [109] examined the effect of long-term administration of tocotrienols on hepatocarcinogenesis in rats. Liver carcinogenesis was induced by diethylnitrosamine and AAF in rats fed a diet containing 30 mg/kg tocotrienols for 9 months. Expression of biomarkers of liver carcinogenesis such as glutathione, alkaline phosphatase, and gamma-glutamyl transpeptidase was enhanced by the carcinogens but attenuated by tocotrienols, decreasing the impact of the carcinogens. A similar study by others confirmed these findings [110]. All these studies suggest that tocotrienols have potential to both prevent and treat cancer.

4.2 Cardioprotective effects

Persistent hypertension is one of the risk factors for strokes, heart attacks, and heart failure and is a leading cause of chronic renal failure. Most of the cardioprotective effects of tocotrienols are mediated through their ability to inhibit a rate-limiting enzyme in cholesterol biosynthesis and their antioxidant and anti-inflammatory activities. In one study, tocotrienols significantly depressed age-related increases in systolic blood pressure of spontaneously hypertensive rats, and the investigators concluded that the tocotrienols' effects were more pronounced than those of α-tocopherol [111]. TRF from palm oil can reduce total cholesterol and LDL-cholesterol levels through downmodulation of hepatic HMG-CoA reductase activity [65]. Whether rice bran oil with its high content of γ-oryzanol and γ-tocotrienol has the same effect has been investigated in rats [112]. A rice bran oil diet lowered plasma triglyceride, LDL-cholesterol and hepatic triglyceride concentrations and increased hepatic cholesterol 7-alpha-hydroxylase, hepatic LDL receptor, and HMG-CoA reductase mRNA in rats. The γ-oryzanol and γtocotrienol in rice bran oil can lead to increased neutral sterol and bile acid excretion in feces via upregulation of cholesterol synthesis and catabolism. Chou et al. observed that rice bran oil improved lipid abnormalities, reduced the atherogenic index and suppressed the hyperinsulinemic response in rats with streptozotocin/nicotinamide–induced type 2 diabetes mellitus [113].

In atherosclerosis, build-up of fatty materials such as cholesterol leads to artery wall thickening. Nafeeza et al. investigated the effect of TRF on the microscopic development of atherosclerosis and lipid peroxidation in the aortas of rabbits. After 10 weeks of treatment with TRF, cholesterol-fed rabbits had lower aortic contents of malondialdehyde, less intimal thickening and greater preservation of the internal elastic lamina than untreated rabbits [114]. Because TRF lowered lipid peroxidation, which in turn reduces intimal thickening and preserves the internal elastic lamina, they concluded that the antioxidant activities of TRF could reduce experimental atherosclerosis.

TRF and isomers of tocotrienols can improve postischemic ventricular function and reduce myocardial infarct size. They exert this cardioprotective effect through downmodulation of cSrc and upregulation of phosphorylation of Akt, thus generating a survival signal [115]. A 6week treatment of diet supplemented with either d-P(21)-T3, d-P(25)-T3, γ-T3, or TRF showed significant effects on lipid metabolism in swine expressing hereditary hypercholesterolemia [116]. Levels of serum total cholesterol, LDL-cholesterol, apolipoprotein B, platelet factor 4, thromboxane B(2), glucose, triglycerides, and glucagon were reduced in all of the treatment groups relative to the control. The hepatic HMG-CoA reductase activity was lower, and cholesterol and fatty acid levels in various tissues were lower in all of the treatment groups.

Activation of the nitric oxide-cGMP pathway is associated with myocardial protection against ischemia; in ischemia, the function of this pathway is disturbed. Esterhuyse et al. investigated the effects of red palm oil on the myocardial nitric oxide-cGMP signaling pathway [117]. Treatment with red palm oil increased aortic output and increased levels of cGMP and polyunsaturated fatty acid in rat hearts. Their findings suggest that dietary red palm oil protects via the nitric oxide-cGMP pathway and/or changes in polyunsaturated fatty acid composition during ischemia/reperfusion. As red palm oil contains both tocopherols and tocotrienols, it is unclear which of these constituents exerted the cardioprotective effect. Newaz et al. determined the effects of γ-tocotrienol on lipid peroxidation and total antioxidant status of spontaneously hypertensive rats. Their study showed that a 3-month antioxidant trial with γ-tocotrienol reduced blood and plasma concentrations of lipid peroxides and improved total antioxidant status and superoxide dismutase activity [19]. The investigators concluded that antioxidant supplementation with γ-tocotrienol may prevent development of increased blood pressure, reduce lipid peroxides in plasma and blood vessels and enhance total antioxidant status, including superoxide dismutase activity.

Myocardial ischemic injury results from severe impairment of coronary blood supply and produces a spectrum of clinical syndromes. Although all of the tocotrienol isomers have cardioprotective properties against myocardial ischemic injury, γ-tocotrienol was the most protective. The differential interaction of MAPK with caveolin 1/3 in conjunction with proteasome stabilization plays a unique role in tocotrienols-mediated cardioprotection, possibly by altering the availability of prosurvival and antisurvival proteins [118].

In a study of the cardioprotective properties of γ-tocotrienol in combination with resveratrol, the two agents acted synergistically, providing a greater degree of cardioprotection than either alone [119]. The basis of this effect is their ability to induce autophagy accompanied by activation of Beclin and LC3-II as well as mTOR signaling while simultaneously generating a greater amount of survival signal through activation of the Akt-Bcl-2 survival pathway.

4.3 Effects against diabetes mellitus

In diabetes the blood glucose level is persistently high because of insufficient insulin production or insulin resistance. TRF prevented increases in serum levels of advanced glycosylation end-products (AGE) in normal rats and decreased blood glucose and glycated hemoglobin levels in diabetic rats [120]. In a similar study, TRF treatment not only reduced serum glucose and glycated hemoglobin concentrations, it also reduced plasma total cholesterol, LDL-cholesterol and triglyceride levels and increased levels of high-density lipoprotein (HDL)-cholesterol, as compared to the untreated group [121]. Tocotrienols exert these effects through increasing superoxide dismutase activity and levels of vitamin C in plasma and decreasing levels of plasma and aorta malondialdehyde and 4-hydroxynonenal and oxidative DNA damage. Thus TRF lowers blood glucose level and oxidative stress markers, improves dyslipidemia, and maintains vessel wall integrity. A combination of insulin and tocotrienol treatment attenuated the diabetic condition and reversed neuropathic pain through modulation of oxidative-nitrosative stress and release of inflammatory cytokines and caspase-3 in diabetic rats [122,123]. In another study, suppression of the NF-κB signaling pathway by tocotrienols prevented diabetes-associated cognitive deficits. Rats with streptozotocin-induced diabetes were treated with oral tocotrienols (25, 50 or 100 mg/kg body weight) for 10 weeks, which significantly prevented behavioral, biochemical and molecular changes associated with diabetes, in part through suppression of activation of the NF-κB signaling pathway [53].

Oxidative stress is considered to be a key factor in the development of diabetes and its complications. Kanaya et al. examined the antioxidative effects of a crude lipophilic rice bran extract, Ricetrienol, which contains α-tocopherol, tocotrienols, and phytosterol, in obese diabetic KKAy mice [124]. While Ricetrienol did not affect hyperglycemia, body weight, or hyperlipidemia, it did significantly suppress elevation of plasma malondialdehyde and significantly increase glutathione peroxidase (GPx) mRNA expression at the 0.1% concentration; the authors suggested that Ricetrienol exerts a protective effect against oxidative damage in diabetes mellitus. Yoshida et al evaluated the antioxidant properties of natural and synthetic dietary antioxidants by using the biomarker, total hydroxyoctadecadienoic acid (tHODE) [125]. Remarkable increases in tHODE and total 8-iso-prostaglandin F (2alpha) (t8iso-PGF (2alpha)) levels were observed in the plasma, erythrocytes, liver and brain of mice that were fed an α-tocopherol–stripped (E-free) diet, whereas levels of these markers were reduced in mice treated with the E-free diet supplemented with a lipophilic antioxidant (0.04 % by wt) containing α-tocopherol, α-tocotrienol, and γ-tocopherol.

Fang et al. investigated the mechanism through which tocotrienols reduce blood glucose levels in patients and in preclinical animal models [126]. They proposed that tocotrienols function as peroxisome proliferator-activated receptor (PPAR) modulators. PPARs are ligand-regulated transcription factors that play essential roles in energy metabolism. Synthetic PPARα and PPARγ ligands have been used recently in the treatment of hyperlipidemia and diabetes. Both α- and γ-tocotrienol activated PPARα, while δ-T3 alone activated PPARα, PPARγ, and PPARδ in reporter-based assays. Tocotrienols enhanced the interaction between the purified ligand-binding domain of PPARα and the receptor-interacting motif of coactivator PPARγ coactivator-1alpha. They also found that TRF improved whole-body glucose utilization and insulin sensitivity of diabetic Db/Db mice by selectively regulating PPAR target genes [118]. All of these results indicate that tocotrienols have antidiabetic potential.

4.4 Neuroprotective effects

Numerous reports indicate that tocotrienols exhibit neuroprotective effects under a wide variety of conditions [53,122,123,127–129]. Chopra and her group noted neuroprotection by tocotrienols in an experimental model of diabetic neuropathy [123], in the rat model of alcoholic neuropathy [128], in chronic alcohol-induced cognitive dysfunction in rats [129], in intracerebroventricular streptozotocin-induced cognitive impairment and oxidative-nitrosative stress in rats [127], in diabetic nephropathy [53], and in diabetes-associated cognitive deficits [122], all through suppression of proinflammatory pathways. Sen and his group have examined extensively the prevention of glutamate-induced neurodegeneration by tocotrienols [16,17, 61,98,130–132]. They found that modulation of c-Src, 12-lipoxygenase and PLA2 is involved in the neuroprotective effects of tocotrienols. Khanna et al. showed that a subattomole quantity of α-tocotrienol, but not γ-tocopherol, protected neurons from glutamate challenge. Rats given a α-tocotrienol supplement showed more protection against stroke-induced injury through downregulation of c-Src activation and 12-lipoxygenase phosphorylation at the stroke site [131]. Roy et al. reported that dietary tocotrienols are bioavailable to both mother and fetal brains and that the enrichment is greater in fetal brain tissue. They also identified a specific set of vitamin E–sensitive genes in the developing rat fetal brain using GeneChip microarray expression profiling. HO-3, LINE-1, and ApoB are some of the vitamin E–sensitive genes affected by vitamin E treatment [132].

A cerebral infarction is an ischemic kind of stroke caused by a disturbance in the blood vessels supplying blood to the brain. α-Tocopherol, α-tocotrienol and γ-tocopherol significantly decreased the size of cerebral infarcts in the mice middle cerebral artery occlusion model, while γ-tocotrienol, δ-tocopherol and δ-tocotrienol showed no effect [133]. Tiwari et al. demonstrated the effectiveness of tocotrienols in attenuation of alcoholic neuropathy [128]. Treatment with α-tocopherol and tocotrienols (mixture of α-, β-, γ-tocotrienol) for 10 weeks significantly improved nociceptive threshold, paw-withdrawal threshold and superoxide dismutase levels and decreased tumor necrosis factor alpha (TNF-α) and IL-1β levels in male Wistar rats. In another study, they investigated the effect of α-tocopherol and α-tocotrienol against intracerebroventricular streptozotocin–induced cognitive impairment and oxidativenitrosative stress in rats. Both isoforms effectively attenuated the reductions in glutathione and catalase and reduced the malondialdehyde, nitrite and cholinesterase activity in the brains of these rats, but the effect was more potent with tocotrienols [127].

4.5 Effects on bone metabolism

Tobacco smoking has been identified as a risk factor in the development of osteoporosis, vitamin E supplements reverse nicotine-induced bone loss and stimulate bone formation [134]. Another group has shown that tocotrienols can reverse nicotine-induced bone loss in rats (Table 3) [54]. Bone histomorphometric parameters of adult male rats treated with TRF or γ-tocotrienol but not with γ-tocopherol (60 mg/kg) following nicotine treatment showed significantly higher trabecular thickness and less eroded surface than the control group. Tocotrienols are slightly superior to tocopherols in attenuating the effects of tobacco; γtocotrienol especially may have therapeutic potential to repair bone damage caused by chronic smoking. This vitamin is an anabolic agent for bone in normal male rats [134–136].

Other studies have shown that tocotrienols can reverse glucocorticoid-induced or free radical– induced bone loss in adrenalectomized rats [55,135,137] and improve normal bone structure [134,135,138], possibly through its antioxidant activity in bone [137]. Maniam et al. investigated the effects of vitamin E on lipid peroxidation and antioxidant enzyme levels in rat bones [137]. They found that palm oil tocotrienols at the dose of 100 mg/kg body weight significantly reduced the level of thiobarbituric acid–reactive substance in the femur while significantly increasing glutathione peroxidase activity compared to the control group; these effects were not observed in rats treated with γ-tocopherol. Tocotrienols also showed a protective effect against free radical damage in the rat femur bones. Long-term glucocorticoid treatment is associated with severe side effects, such as obesity and osteoporosis. Ima-Nirwana et al. showed that treatment with γ-tocotrienol (60 mg/kg body weight/day) reduced body fat mass and increased fourth lumbar vertebra bone calcium content in rats, while α-tocopherol was ineffective [139]. Therefore, palm oil-derived γ-tocotrienol has the potential to be utilized as a prophylactic agent in prevention of the skeletal side effects of long-term glucocorticoid and tobacco use.

4.6 Immunomodulatory effects

Gu et al. demonstrated the immunoregulatory effects of dietary α-tocopherol and mixture of tocotrienols on humoral and cell-mediated immunity [140]. Their results showed that tocopherols or tocotrienols feeding enhanced expression of interferon-gamma, IgA, and IgG, but not IgE, and decreased the proportion of CD4+ T cells. Interestingly, tocotrienols decreased the expression of TNF-α. These investigators concluded that oral administration of tocopherols and tocotrienols affects the proliferation and function of spleen and mesenteric lymph node lymphocytes.

4.7 Gastroprotective effects

Azlina et al. compared the impacts of tocopherols and tocotrienols on gastric acidity, gastric tissue content of parameters such as malondialdehyde and prostaglandin E2, and serum levels of gastrin and glucagon-like peptide-1 in rats exposed to restraint stress. They found that tocotrienol-treated animals, both stressed and non-stressed, had comparable gastric acidity and gastrin levels [141]. Both tocopherols and tocotrienols had gastroprotective effects against damage by free radicals generated in stress conditions, but only tocotrienols had the ability to block stress-induced changes in gastric acidity and gastrin level. Another group showed that tocotrienols can prevent aspirin-induced gastric lesions through their ability to limit lipid peroxidation [142].

5. Pharmacokinetics of tocotrienol

Numerous studies on the pharmacokinetics, organ and tissue distribution and toxicity of tocopherols and tocotrienols have been carried out [143–155]. Yap et al. determined the pharmacokinetics and bioavailability of α-, γ-, and δ-tocotrienol given via oral, intravenous, intramuscular and intraperitoneal routes in rats. They found that oral absorption of all forms of tocotrienols were incomplete and that absorption of tocotrienols given via the intramuscular or intraperitoneal routes was negligible; they concluded that these routes of administration should be avoided [155]. They also found that α-tocotrienol had greater bioavailabity than γ tocotrienol and δ-tocotrienol. The absolute bioavailability of α-tocotrienol was approximately 28%, while the bioavailability of γ- and δ-tocotrienol were around 9% [155]. Phase I cytochrome p450 3A4 enzyme and P-glycoprotein at the gastrointestinal epithelium are implicated in the oral absorption of tocotrienols. Preferential absorption of α-tocotrienol over γ-tocotrienol and δ-tocotrienol in rats is in agreement with other reports in pigs [156] and lymphatic cannulated rats [146]. These differences may be linked to the number of methyl groups in the chromanol ring, as α-tocotrienol has three, γ-tocotrienol has two, and δ-tocotrienol has one methyl group, and thus they have different lipophilicities.

In another study, following a single oral administration of δ-tocotrienol (100 mg/kg), the peak plasma concentration was 57 +/− 5 micromol/l, the time required to reach peak plasma concentration was 2 hours, and the plasma half-life was 3.5 hours. The tocotrienols were cleared from plasma and liver within 24 hours, but clearance from the pancreas was delayed [145]. δ-tocotrienol was 10-fold more concentrated in the pancreas than in the tumor and no toxicity was shown by δ-tocotrienol (100mg/ kg) in mice. Intestinal epithelial cells absorb γ-tocotrienol faster than α-tocopherol. Tocotrienol isomers accumulated rapidly in Caco2 cells treated with micelles of vitamin E isomers consisting of bile salts, lysophospholipids, free fatty acid, and 2-monoacylglycerols and was greater than the accumulations of corresponding tocopherol isomers [154]. This finding shows that the difference in saturation of the side chains of tocopherols and tocotrienols, rather than the difference in their rings, was responsible for the rapid epithelial transport into the Caco2 cell membranes. α-Tocopherol, α-tocotrienol and γtocotrienol can all be retained abundantly by the skin of rats and mice, but only α-tocopherol is retained by the liver, kidney, and plasma of these animals [147]. Dietary sesame seeds can elevate absorption and concentrations of α- and γ-tocotrienol in skin and adipose tissue [145]. Kawakami et al. investigated the distribution of tocotrienols in rats and reported that γtocotrienol was significantly distributed to the adipose tissue and that the adipose tissue concentration increased from 1.1 to 10.2 nmol/g according to rice bran tocotrienols intake [149].

Nakamura examined the 13-week oral toxicity of a tocotrienol preparation in rats and found that the no-observed-adverse-effect level of tocotrienols was 0.019% in the diet (i.e., 120 mg/ kg body weight/day for male and 130 mg/kg body weight for female rats). A decrease in total cholesterol was observed in males in line with the hypocholestrolemic activity of this vitamin [157].

6. Clinical Studies with Tocotrienols

Numerous clinical studies have been performed to examine bioavailability and various therapeutic effects of tocotrienols in humans (Table 4; Figure 4).

6.1 Pharmacodynamics and pharmacokinetics

In a double-blind placebo-controlled study, the bioavailability of purified α-, γ- or δ-tocotrienol (250 mg/day for 8 weeks) in hypercholesterolemic humans was examined. At the end of the study period, plasma levels of α-tocotrienol, γ-tocotrienol and δ-tocotrienol were 0.8 μM, 0.54 μM and 0.09 tocotrienol were 0.8 μM, 0.54 μM and 0.09 M, respectively [158]. The preferential absorption of α-tocotrienol in humans noted here is in agreement with that noted in rats [146]. Hayes et al. reported that tocotrienols were transported by chylomicrons and disappeared from the plasma during chylomicron clearance [159]. Another study investigated the pharmacokinetics and bioavailability of a single oral dose (300 mg) of α-tocotrienol, γtocotrienol and δ-tocotrienol in healthy volunteers (N=8) under fed and fasting conditions. Oral bioavailability of all tocotrienol analogues was markedly increased when taken with food, with peak plasma concentrations (1.52–ss5.87 tocotrienol were 0.8 μM, 0.54 μM and 0.09 M) occurring between 3 and 5 hours after ingestion. The biological half-lives of α-tocotrienol, γtocotrienol and δ-tocotrienol were 2.3, 4.4, and 4.3 hours, respectively. The half-life of αtocopherol is about 20 hours; thus the half-lives of the tocotrienols are 4.5- to 8.7-fold shorter [160]. When the tocotrienol analogues were given at the same dose, plasma levels of α tocotrienol were twice those of γ-tocotrienol and ten times higher than those of δ-tocotrienol. Another study showed that α- and γ-tocotrienol are metabolized to carboxyethylhydroxychroman derivatives and excreted in human urine [161]. When human subjects (n=6) consumed 125 mg of tocotrienyl acetate daily for the first week, 500 mg daily for the second week, 125 mg daily for the third week and 500 mg daily for the fourth week, only 1–2% of αtocotrienol and 4–6% of γ-tocotrienol metabolites was recovered in the urine. To overcome the limited oral bioavailability of tocotrienols, self-emulsifying formulations have been tested in healthy human volunteers with favorable results [162].carcinogenesis such as glutathione, alkaline phosphatase, and gamma-glutamyl transpeptidase was enhanced by the carcinogens but attenuated by tocotrienols, decreasing the impact of the carcinogens. A similar study by others confirmed these findings [110]. All these studies suggest that tocotrienols have potential to both prevent and treat cancer.

6.2 Effects on cardiovascular system

About 50% of persons consuming the typical Western diet will die of coronary heart disease or stroke. Hypercholesterolemia and inflammation of the coronary artery are the major risk factors for development of coronary heart disease. While dietary fat has been associated with coronary heart disease, a diet of predominantly plant foods, such as rice, oats and barley (all of which contain tocotrienols), can retard this disease. While some studies indicate that tocotrienols have cardioprotective properties in humans [2,3,158,163–171], others have failed to show the benefit [172–174].

In the first study ever performed on the effects of tocotrienols in human subjects, 22 healthy volunteers took one capsule daily containing a palm oil-vitamin E concentrate (palmvitee) that comprised approximately 18 mg tocopherols, 42 mg tocotrienols and 240 mg palm olein for 30 days [170]. The investigators observed decreases in total cholesterol ranging from 5% to 35.9% and in LDL-cholesterol from 0.9% to 37% [170]. These cholesterol-lowering effects were attributed to tocotrienols, as tocopherols has been shown in human subjects to lack these effects [175]. Qureshi et al. performed a double-blind, crossover 8-week study comparing the effect of 200 mg palmvitee/day with that of 300 mg corn oil (which lacks tocotrienols) on the serum lipid levels of 25 hypercholesterolemic human subjects. The serum cholesterol levels of seven subjects decreased by 31% during the 4-week period of treatment that included tocotrienols, and this effect persisted even 2 weeks after the capsules were discontinued [167]. Later, in another clinical trial, Qureshi et al. administered TRF from rice bran oil in a 12-week double-blind study in 21 hypercholesterolemic subjects. A 12% decrease in total cholesterol and 16% reduction in LDL-cholesterol were noted in subjects given TRF during the 4-week period in which the dose was 200 mg but not in the placebo group given 1.2 g corn oil. Furthermore, a 17% decrease in lipoprotein-a was noted in the treated group, which is remarkable as most cholesterol-lowering drugs do not affect lipoprotein-a [166].

The antioxidant benefit of tocotrienols has been reported in a group of patients with cerebrovascular diseases. Both tocopherols and 240 mg mixed tocotrienols were used in this trial [171]. In another clinical trial, low doses of TRF for 25 weeks were found to exhibit synergistic effects with lovastatin on various lipid parameters in hypercholesterolemic humans [2]. Further studies revealed that the effect of TRF on serum cholesterol levels was dose dependent when administered at 25, 50, 100, or 200 mg/day. The maximum decrease (25%) was seen at the 100 mg/day dose [3].

In contrast to these studies, Mensink et al. showed, in a randomized, double-blind, placebocontrolled trial in 20 mildly hypercholesterolemic men who received 140 mg tocotrienols and 20 mg α-tocopherol for 4 weeks, that this regimen had no effect on serum lipid levels [172]. Whether the negative results were due to the presence of the tocopherols are not clear, but αtocopherol has been shown to neutralize the HMG-CoA reductase–inhibitory activity of tocotrienols [63]. These results do agree, however, with those of Wahlqvist et al. [176]. In another double-blind, placebo-controlled study, the serum cholesterol-lowering efficacy of purified α-, γ- or δ-tocotrienol (250 mg/d) for 8 weeks in hypercholesterolemic humans was examined. Although at the end of the study period, plasma levels of α-tocotrienol, γ-tocotrienol and δ-tocotrienol were 0.98 tocotrienol were 0.8 μM, 0.54 μM and 0.09 M, 0.54 tocotrienol were 0.8 μM, 0.54 μM and 0.09 M and 0.09 tocotrienol were 0.8 μM, 0.54 μM and 0.09 M, respectively, no change in serum or LDL-cholesterol levels were observed [158]. Alphatocotrienol did decrease the oxidizing potential of LDL. Mustad et al. also showed a lack of effect of supplementation of tocotrienols (200 mg/day) on hypercholesterolemia [173].

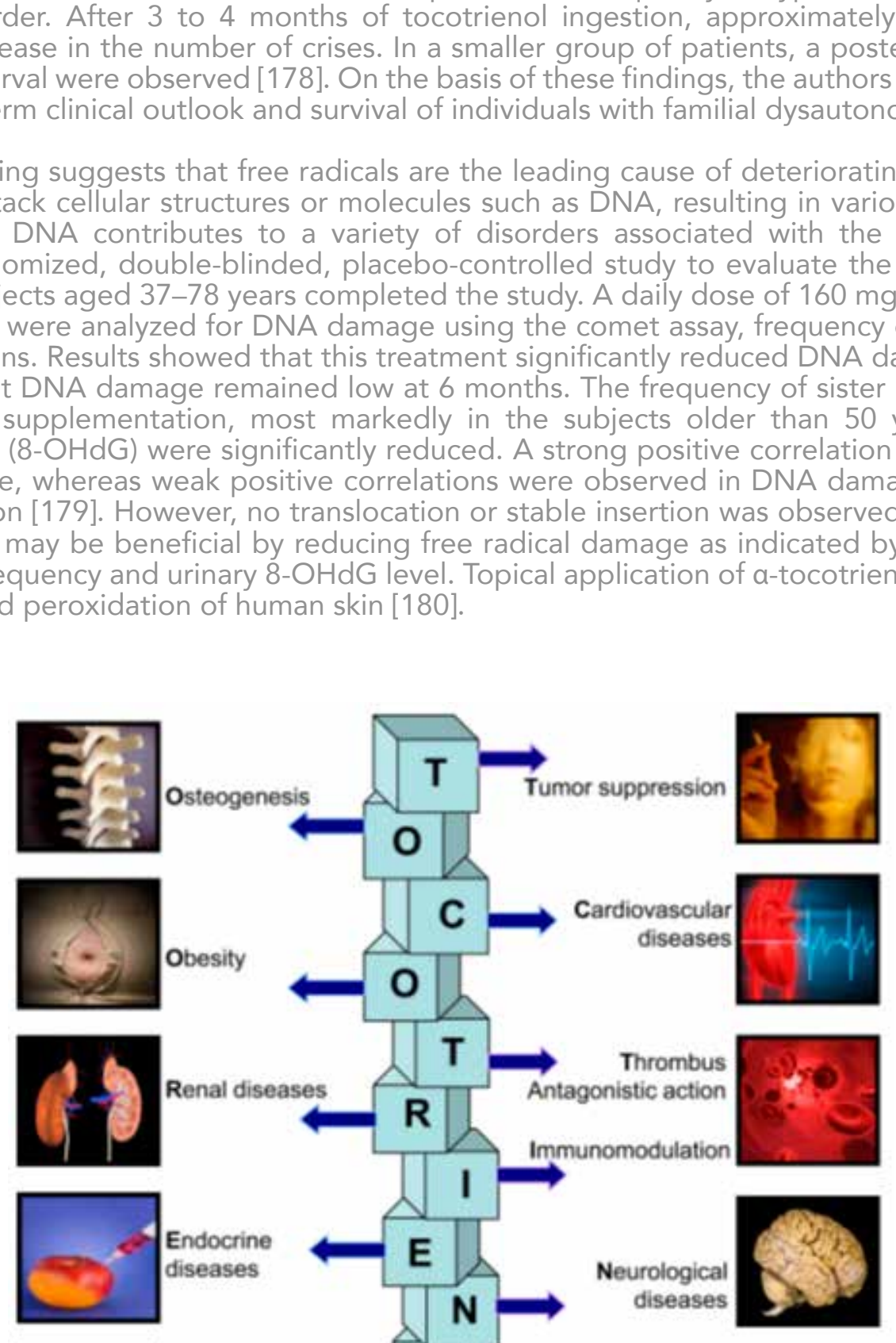
Another study examined the effects of three doses of tocotrienol-rich vitamin E (TRE) on plasma tocotrienol isomer concentrations, arterial compliance, plasma total antioxidant status, aortic systolic blood pressure, and serum total cholesterol and LDL-cholesterol levels in healthy men. This randomized, blinded endpoint, placebo-controlled clinical trial with a parallel design involved 36 male subjects who took either an oral placebo or TRE at doses of 80, 160, or 320 mg daily for 2 months. Baseline tocotrienol isomer concentrations were low and in some subjects not detectable. At the end of the study period, all TRE-treated groups showed significant increases in α-, δ- and γ-tocotrienol concentrations from baseline relative to the placebo group. There was a linear dose and blood level relationship for all the isomers. There was no significant difference between groups, however, in pulse wave velocity, arterial compliance, plasma total antioxidant status, aortic systolic blood pressure, or serum levels of total cholesterol or LDL-cholesterol from baseline to end of treatment. Groups receiving 160 mg or 320 mg of TRE showed significant reductions in their aortic systolic blood pressure, and the group receiving 320 mg showed a significant 9.2% improvement in total antioxidant status [174].

The progression of atherosclerosis is more rapid in individuals with type 2 diabetes than in the general population, and 80% of those with type 2 diabetes will die of an atherosclerotic event. Since in these patients hyperglycemia per se confers increased risk for cardiovascular disease, the presence of even a borderline-high-risk LDL-cholesterol level signals the need for aggressive LDL-lowering therapy. Thus Baliarsingh et al. investigated the therapeutic impacts of tocotrienols on serum and lipoprotein lipid levels in patients with type 2 diabetes in a randomized, double blind, placebo-controlled design involving 19 subjects with type 2 diabetes and hyperlipidemia. After 60 days of TRF treatment, subjects showed average declines of 23%, 30%, and 42% in serum total lipids, total cholesterol, and LDL-cholesterol, respectively. Tocotrienols mediated a reduction of LDL-cholesterol level from an average of 179 mg/ml to 104 mg/ml. No hypoglycemic effect was observed in these patients because their glucose and glycated hemoglobin levels at baseline were close to normal values [164]. These findings suggest that daily intake of dietary TRF by individuals with type 2 diabetes will be useful in the prevention and treatment of hyperlipidemia and atherogenesis.

6.3 Other uses

Familial dysautonomia, a genetic neurodegenerative disorder affecting primarily individuals of Ashkenazi Jewish descent, is caused by mutations in the IKBKAP gene, which encodes the IkappaB kinase complex-associated protein (IKAP). The more common or major mutation causes aberrant splicing, resulting in a truncated form of IKAP. Tissues from individuals homozygous for the major mutation contain both mutant and wild-type IKAP [177] transcripts. The apparent leaky nature of this mutation prompted a search for agents capable of elevating the level of expression of the wild-type IKAP transcript. It has been shown that tocotrienols can increase the transcription of IKAP mRNA in familial dysautonomia–derived cells, with corresponding increases in the correctly spliced transcript and normal protein. Because ingestion of tocotrienols elevates IKAP and MAO-A in familial dysautonomia patients, Rubin et al. examined their impact on the frequency of hypertensive crises and cardiac function in individuals with this disorder. After 3 to 4 months of tocotrienol ingestion, approximately 80% of patients reported a significant (> or = 50%) decrease in the number of crises. In a smaller group of patients, a postexercise increase in heart rate and a decrease in the QT interval were observed [178]. On the basis of these findings, the authors hypothesized that tocotrienol therapy improves the long-term clinical outlook and survival of individuals with familial dysautonomia.

The free radical theory of aging suggests that free radicals are the leading cause of deteriorating physiologic function during senescence. Free radicals attack cellular structures or molecules such as DNA, resulting in various modifications to the DNA. Accumulation of unrepaired DNA contributes to a variety of disorders associated with the aging process. Chin and his coworkers performed a randomized, double-blinded, placebo-controlled study to evaluate the effect of Tri E tocotrienol on DNA damage. Sixty-four subjects aged 37–78 years completed the study. A daily dose of 160 mg of Tri E tocotrienol was given for 6 months. Blood samples were analyzed for DNA damage using the comet assay, frequency of sister chromatid exchange, and chromosome 4 aberrations. Results showed that this treatment significantly reduced DNA damage as measured by comet assay after 3 months and that DNA damage remained low at 6 months. The frequency of sister chromatid exchange was also reduced after 6 months of supplementation, most markedly in the subjects older than 50 years, while urinary levels of 8hydroxy-2'-deoxyguanosine (8-OHdG) were significantly reduced. A strong positive correlation was observed between sister chromatid exchange with age, whereas weak positive correlations were observed in DNA damage and 8-OHdG, which were reduced with supplementation [179]. However, no translocation or stable insertion was observed in chromosome 4. Thus Tri E tocotrienol supplementation may be beneficial by reducing free radical damage as indicated by reductions in DNA damage, sister chromatid exchange frequency and urinary 8-OHdG level. Topical application of α-tocotrienol has been shown to prevent benzyl peroxide–induced lipid peroxidation of human skin [180].



7. Tocotrienols vs. Tocopherols

Tocotrienols differ from tocopherols in that the former contain three double bonds in their isoprenoid side chain while the latter do not; this may account for the differences in their efficacy and potency in vitro and in vivo (Table 5) [61,133,181–183]. While over 30,000 papers have been published on tocopherols, fewer than 600 exist on tocotrienols, most published within the last 5 years. Tocopherols are present mainly in corn, wheat and soybeans, whereas tocotrienols occur mainly in barley, oats, palm, and rice bran. Although tocopherols and tocotrienols are structurally very similar and both are metabolized through similar mechanisms involving initial w-hydroxylation followed by five cycles of β-oxidation [184], tocotrienols have been found to exhibit superior antioxidant activity [96,181,185]. McIntyre et al. showed that tocotrienols were several-fold more effective than tocopherols in inhibiting the proliferation of mouse mammary tumor epithelial cells and in inducing apoptosis [70].

Almost millimolar doses of tocopherols were required for antiproliferative effects [186]. The authors showed that these differences could be linked to preferential accumulation of tocotrienols as compared to tocopherols. These differences may also be due to α-tocopherol transfer protein, which binds to α-tocopherol with higher affinity than to tocotrienols [187]. When their respective effects on proliferation of bovine endothelial cells, a marker of angiogenesis, were measured, only tocotrienols (not tocopherols) inhibited this proliferation [188]. Another study showed that oral administration of tocotrienols but not tocopherols blocked tumor-induced angiogenesis. These investigators showed that tocotrienols downregulated VEGF receptor expression in HUVEC cells and blocked VEGF signaling [189].

Suarna et al. reported that when rats or humans were treated with tocotrienols and tocopherols, tocotrienols provided oxidative protection but tocopherols did not [190]. α-tocopherol has been reported to attenuate the inhibitory effects of tocotrienols on HMG-COA activity [63]. Tocotrienols has been shown to be converted to tocopherols in vivo [2]. High concentrations of γ-tocotrienol but not α-tocopherol were cytotoxic to astrocytes. This difference was attributed to the greater prooxidant activity of tocotrienols at high concentrations. At low concentrations, however, tocotrienols were found to be antioxidant and to protect cells from hydrogen peroxide–induced killing [191]. This is consistent with studies showing that tocotrienols are more effective than tocopherols in protecting against glutamate-induced cell death in HT4 neuron cell culture [61]. Whether these differences were due to the differential rates of uptake of tocotrienols and tocopherols by the neuronal cells is controversial [191]. Tocotrienols were many times as potent as tocopherols in protecting cerebellar granule cells against methyl mercury toxicity, an effect that was linked to the difference in the antioxidant potency of the two forms of the vitamin [192]. In vitro studies showed that tocotrienols has greater anti-inflammatory activity than tocopherols as measured by lipopolysaccharideinduced production of IL-6 and prostaglandin E2 [193]. Mishima et al. showed that αtocotrienol and γ-tocotrienol were more effective than α-tocopherol in preventing cerebral infarction in vivo in mice [133].

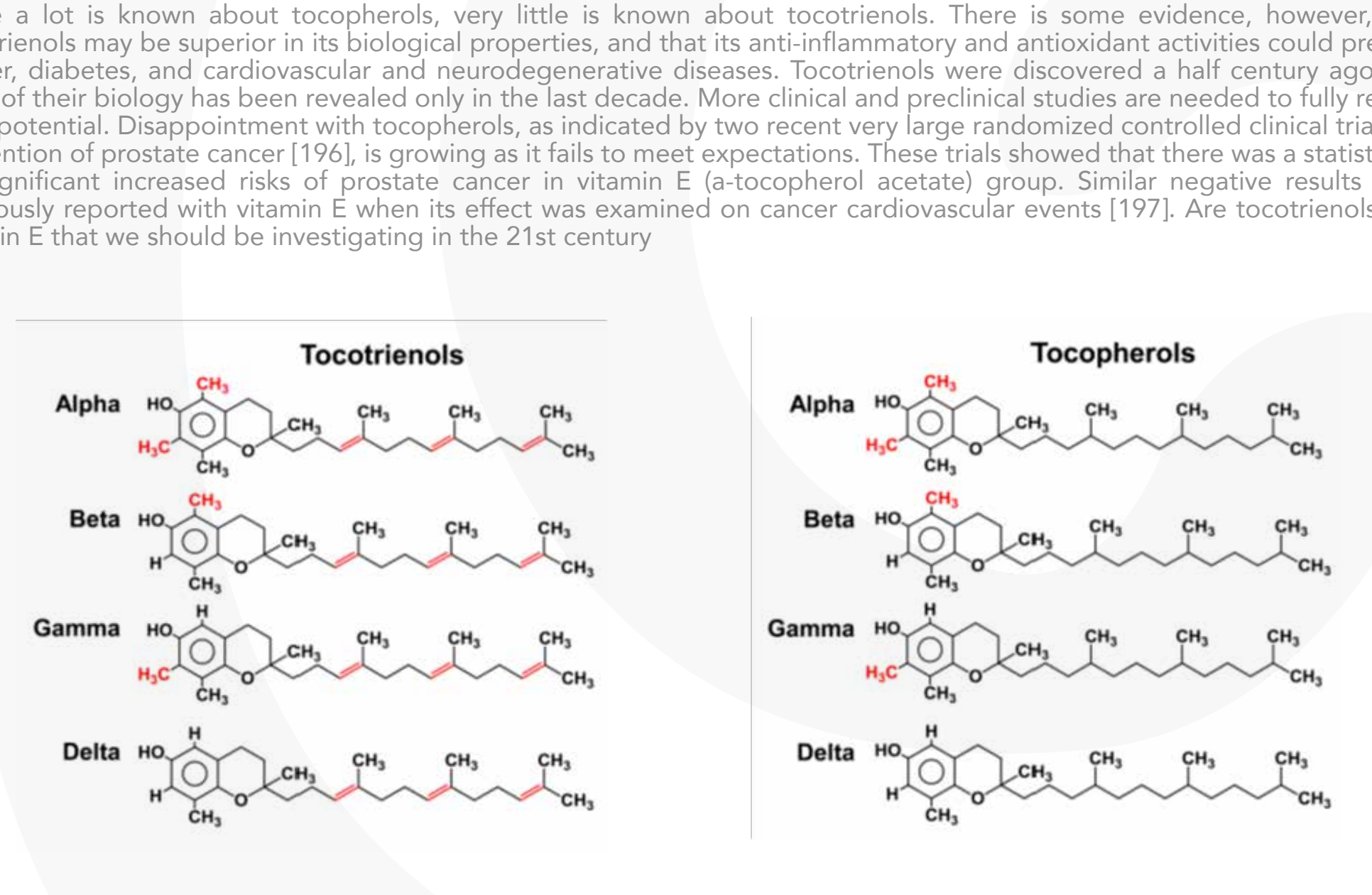
Any number of mechanisms could account for the difference in potency of tocotrienols and tocopherols. First, because of structural differences, tocotrienols may be more uniformly distributed in the lipid bilayer. Second, the chromanol ring of tocotrienols may interact more efficiently with the lipid bilayer than that of tocopherols. Third, tocotrienols may have a higher recycling efficiency [194]. Fourth, cellular uptake of tocotrienols is 70 times higher than that of tocopherols [151]. All of these factors may contribute to tocotrienol's greater efficacy. It has also been shown that tocotrienol isomers are accumulated and secreted at greater rates in Caco2 cells than tocopherol isomers. When administered orally to mice, tocotrienols appeared faster in the plasma but at lower levels than tocopherols [154].

Alpha-tocotrienol mediates some of its effect by inhibiting HMG-CoA reductase activity [66], while α-tocopherol induces HMG-CoA reductase activity [63].

7. Tocotrienol isoforms The isoforms of tocotrienols, which differ in their number of methyl groups, also differ in their biological activities (Table 6). While various studies have indicated that α-tocotrienol is highly neuroprotective [61,131], δ- and γ-tocotrienol have been shown to exhibit the greatest anticancer effects. In vitro studies suggest that there may be as much as a 30-fold difference in the ability of α, γ, and δ isomers of tocotrienol to inhibit cholesterol biosynthesis [67]. The antioxidant capacity of these three isomers is α-tocotrienol > γ-tocotrienol > δ-tocotrienol [185]. The antioxidant activity of α-tocotrienol is similar to that of α-tocopherol [190]. McIntyre et al. showed that various tocotrienols differ in their potency in inhibiting the proliferation of mouse mammary tumor epithelial cells and in inducing apoptosis [70]. They Aggarwal et al. Page 13 identified the relative potencies of these three isomers as δ-tocotrienol ≥ γ-tocotrienol> αtocotrienol. These observations agree with that of Inkouchi et al., who showed that the relative potencies for the suppression of proliferation of bovine and human endothelial cells and tube formation were δ-tocotrienol > β-tocotrienol > γ-tocotrienol = α-tocotrienol [188,189,195]. All these reports point to differences in the mechanisms of action of the tocotrienol isomers.

8. Conclusion

While a lot is known about tocopherols, very little is known about tocotrienols. There is some evidence, however, that tocotrienols may be superior in its biological properties, and that its anti-inflammatory and antioxidant activities could prevent cancer, diabetes, and cardiovascular and neurodegenerative diseases. Tocotrienols were discovered a half century ago, but most of their biology has been revealed only in the last decade. More clinical and preclinical studies are needed to fully realize their potential. Disappointment with tocopherols, as indicated by two recent very large randomized controlled clinical trials for prevention of prostate cancer [196], is growing as it fails to meet expectations. These trials showed that there was a statistically nonsignificant increased risks of prostate cancer in vitamin E (a-tocopherol acetate) group. Similar negative results were previously reported with vitamin E when its effect was examined on cancer cardiovascular events [197]. Are tocotrienols, the vitamin E that we should be investigating in the 21st century



References

1. Sen CK, Khanna S, Rink C, Roy S. Tocotrienols: the emerging face of natural vitamin E. Vitam Horm 2007;76:203–261. [PubMed: 17628176]

2. Qureshi AA, Sami SA, Salser WA, Khan FA. Synergistic effect of tocotrienol-rich fraction (TRF(25)) of rice bran and lovastatin on lipid parameters in hypercholesterolemic humans. J Nutr Biochem 2001;12:318–329. [PubMed: 11516635]

3. Qureshi AA, Sami SA, Salser WA, Khan FA. Dose-dependent suppression of serum cholesterol by tocotrienol-rich fraction (TRF25) of rice bran in hypercholesterolemic humans. Atherosclerosis 2002;161:199–207. [PubMed: 11882333]

4. Bunyan J, Mc HD, Green J, Marcinkiewicz S. Biological potencies of epsilon- and zeta-1-tocopherol and 5-methyltocol. Br J Nutr 1961;15:253–257. [PubMed: 13688870]

5. Dunphy PJ, Whittle KJ, Pennock JF, Morton RA. Identification and estimation of tocotrienols in Hevea latex. Nature 1965;201:521–522.

6. Whittle KJ, Dunphy PJ, Pennock JF. The isolation and properties of delta-tocotrienol from Hevea latex. Biochem J 1966;100:138–145. [PubMed: 5965249]

7. Qureshi AA, Burger WC, Peterson DM, Elson CE. The structure of an inhibitor of cholesterol biosynthesis isolated from barley. J Biol Chem 1986;261:10544–10550. [PubMed: 3733719]

8. Kato A, Yamaoka M, Tanaka A, Komiyama Ka, Umezawa I. Physiological effect of tocotrienol. J Japan Oil Chem Soc (Yukugaku) 1985;34:375–376.

9. Sundram K, Khor HT, Ong AS, Pathmanathan R. Effect of dietary palm oils on mammary carcinogenesis in female rats induced by 7,12-dimethylbenz(a)anthracene. Cancer Res 1989;49:1447– 1451. [PubMed: 2493981]

10. Tan B. Tocotrienols: The New Vitamin E. Spacedocnet.

11. Kobayashi H, Kanno C, Yamauchi K, Tsugo T. Identification of alpha-, beta-, gamma-, and deltatocopherols and their contents in human milk. Biochim Biophys Acta 1975;380:282–290. [PubMed: 1120147]

12. van Haaften RI, Haenen GR, Evelo CT, Bast A. Tocotrienols inhibit human glutathione S-transferase P1-1. IUBMB Life 2002;54:81–84. [PubMed: 12440523]

13. Comitato R, Nesaretnam K, Leoni G, Ambra R, Canali R, Bolli A, et al. A novel mechanism of natural vitamin E tocotrienol activity: involvement of ERbeta signal transduction. Am J Physiol Endocrinol Metab 2009;297:E427–E437. [PubMed: 19491296]

14. Upadhyay J, Misra K. Towards the interaction mechanism of tocopherols and tocotrienols (vitamin E) with selected metabolizing enzymes. Bioinformation 2009;3:326–331. [PubMed: 19707294]

15. Zhou C, Tabb MM, Sadatrafiei A, Grun F, Blumberg B. Tocotrienols activate the steroid and xenobiotic receptor, SXR, and selectively regulate expression of its target genes. Drug Metab Dispos 2004;32:1075–1082. [PubMed: 15269186]

16. Khanna S, Roy S, Park HA, Sen CK. Regulation of c-Src activity in glutamate-induced neurodegeneration. J Biol Chem 2007;282:23482–23490. [PubMed: 17569670]

17. Khanna S, Roy S, Ryu H, Bahadduri P, Swaan PW, Ratan RR, et al. Molecular basis of vitamin E action: tocotrienol modulates 12-lipoxygenase, a key mediator of glutamate-induced neurodegeneration. J Biol Chem 2003;278:43508–43515. [PubMed: 12917400]

18. Lee SP, Mar GY, Ng LT. Effects of tocotrienol-rich fraction on exercise endurance capacity and oxidative stress in forced swimming rats. Eur J Appl Physiol 2009;107:587–595. [PubMed: 19705143]

19. Newaz MA, Nawal NN. Effect of gamma-tocotrienol on blood pressure, lipid peroxidation and total antioxidant status in spontaneously hypertensive rats (SHR). Clin Exp Hypertens 1999;21:1297– 1313. [PubMed: 10574414]

20. Hsieh TC, Wu JM. Suppression of cell proliferation and gene expression by combinatorial synergy of EGCG, resveratrol and gamma-tocotrienol in estrogen receptor-positive MCF-7 breast cancer cells. Int J Oncol 2008;33:851–859. [PubMed: 18813800]

21. Adam A, Marzuki A, Ngah WZ, Top GM. Nitrofurantoin-induced hepatic and pulmonary biochemical changes in mice fed different vitamin E doses. Pharmacol Toxicol 1996;79:334–339. [PubMed: 9000262]

22. Renuka Devi R, Arumughan C. Antiradical efficacy of phytochemical extracts from defatted rice bran. Food Chem Toxicol 2007;45:2014–2021. [PubMed: 17574716]

23. Weng-Yew W, Selvaduray KR, Ming CH, Nesaretnam K. Suppression of tumor growth by palm tocotrienols via the attenuation of angiogenesis. Nutr Cancer 2009;61:367–373. [PubMed: 19373610]

24. Rashid SA, Halim AS, Muhammad NA. The effect of vitamin E on basic fibroblast growth factor level in human fibroblast cell culture. Med J Malaysia 2008;63 Suppl A:69–70. [PubMed: 19024988]

25. Shun MC, Yu W, Gapor A, Parsons R, Atkinson J, Sanders BG, et al. Pro-apoptotic mechanisms of action of a novel vitamin E analog (alpha-TEA) and a naturally occurring form of vitamin E (deltatocotrienol) in MDA-MB-435 human breast cancer cells. Nutr Cancer 2004;48:95–105. [PubMed: 15203383]

26. Pierpaoli E, Viola V, Pilolli F, Piroddi M, Galli F, Provinciali M. gamma- and delta-tocotrienols exert a more potent anticancer effect than alpha-tocopheryl succinate on breast cancer cell lines irrespective of HER-2/neu expression. Life Sci 2010;86:668–675. [PubMed: 20188744]

27. Wu SJ, Ng LT. Tocotrienols inhibited growth and induced apoptosis in human HeLa cells through the cell cycle signaling pathway. Integr Cancer Ther 2010;9:66–72. [PubMed: 20150225]

28. Agarwal MK, Agarwal ML, Athar M, Gupta S. Tocotrienol-rich fraction of palm oil activates p53, modulates Bax/Bcl2 ratio and induces apoptosis independent of cell cycle association. Cell Cycle 2004;3:205–211. [PubMed: 14712090]

29. Samant GV, Wali VB, Sylvester PW. Anti-proliferative effects of gamma-tocotrienol on mammary tumour cells are associated with suppression of cell cycle progression. Cell Prolif 2010;43:77–83. [PubMed: 19922488]

30. Elangovan S, Hsieh TC, Wu JM. Growth inhibition of human MDA-mB-231 breast cancer cells by delta-tocotrienol is associated with loss of cyclin D1/CDK4 expression and accompanying changes in the state of phosphorylation of the retinoblastoma tumor suppressor gene product. Anticancer Res 2008;28:2641–2647. [PubMed: 19035289]

31. Wali VB, Bachawal SV, Sylvester PW. Combined treatment of gamma-tocotrienol with statins induce mammary tumor cell cycle arrest in G1. Exp Biol Med (Maywood) 2009;234:639–650. [PubMed: 19359655]

32. Sun W, Wang Q, Chen B, Liu J, Liu H, Xu W. Gamma-tocotrienol-induced apoptosis in human gastric cancer SGC-7901 cells is associated with a suppression in mitogen-activated protein kinase signalling. Br J Nutr 2008;99:1247–1254. [PubMed: 18081943]

33. Park SK, Sanders BG, Kline K. Tocotrienols induce apoptosis in breast cancer cell lines via an endoplasmic reticulum stress-dependent increase in extrinsic death receptor signaling. Breast Cancer Res Treat. 2010

34. Ahn KS, Sethi G, Krishnan K, Aggarwal BB. Gamma-tocotrienol inhibits nuclear factor-kappaB signaling pathway through inhibition of receptor-interacting protein and TAK1 leading to suppression of antiapoptotic gene products and potentiation of apoptosis. J Biol Chem 2007;282:809–820. [PubMed: 17114179]

35. Gysin R, Azzi A, Visarius T. Gamma-tocopherol inhibits human cancer cell cycle progression and cell proliferation by down-regulation of cyclins. FASEB J 2002;16:1952–1954. [PubMed: 12368234]

36. Samant GV, Sylvester PW. gamma-Tocotrienol inhibits ErbB3-dependent PI3K/Akt mitogenic signalling in neoplastic mammary epithelial cells. Cell Prolif 2006;39:563–574. [PubMed: 17109639]

37. Eitsuka T, Nakagawa K, Miyazawa T. Down-regulation of telomerase activity in DLD-1 human colorectal adenocarcinoma cells by tocotrienol. Biochem Biophys Res Commun 2006;348:170–175. [PubMed: 16875674]

38. Shah S, Sylvester PW. Tocotrienol-induced caspase-8 activation is unrelated to death receptor apoptotic signaling in neoplastic mammary epithelial cells. Exp Biol Med (Maywood) 2004;229:745–755. [PubMed: 15337828]

39. Rickmann M, Vaquero EC, Malagelada JR, Molero X. Tocotrienols induce apoptosis and autophagy in rat pancreatic stellate cells through the mitochondrial death pathway. Gastroenterology 2007;132:2518–2532. [PubMed: 17570223]

40. Sakai M, Okabe M, Tachibana H, Yamada K. Apoptosis induction by gamma-tocotrienol in human hepatoma Hep3B cells. J Nutr Biochem 2006;17:672–676. [PubMed: 16517139]

41. Xu WL, Liu JR, Liu HK, Qi GY, Sun XR, Sun WG, et al. Inhibition of proliferation and induction of apoptosis by gamma-tocotrienol in human colon carcinoma HT-29 cells. Nutrition 2009;25:555–566. [PubMed: 19121919]

42. Shah SJ, Sylvester PW. Tocotrienol-induced cytotoxicity is unrelated to mitochondrial stress apoptotic signaling in neoplastic mammary epithelial cells. Biochem Cell Biol 2005;83:86–95. [PubMed: 15746970]

43. Takahashi K, Loo G. Disruption of mitochondria during tocotrienol-induced apoptosis in MDAMB- 231 human breast cancer cells. Biochem Pharmacol 2004;67:315–324. [PubMed: 14698044]

44. Sun W, Xu W, Liu H, Liu J, Wang Q, Zhou J, et al. gamma-Tocotrienol induces mitochondriamediated apoptosis in human gastric adenocarcinoma SGC-7901 cells. J Nutr Biochem 2009;20:276–284. [PubMed: 18602811]

45. Sylvester PW, Shah S. Intracellular mechanisms mediating tocotrienol-induced apoptosis in neoplastic mammary epithelial cells. Asia Pac J Clin Nutr 2005;14:366–373. [PubMed: 16326643]

46. Srivastava JK, Gupta S. Tocotrienol-rich fraction of palm oil induces cell cycle arrest and apoptosis selectively in human prostate cancer cells. Biochem Biophys Res Commun 2006;346:447–453. [PubMed: 16762318]

47. Shibata A, Nakagawa K, Sookwong P, Tsuduki T, Tomita S, Shirakawa H, et al. Tocotrienol inhibits secretion of angiogenic factors from human colorectal adenocarcinoma cells by suppressing hypoxiainducible factor-1alpha. J Nutr 2008;138:2136–2142. [PubMed: 18936210]

48. Miyazawa T, Shibata A, Nakagawa K, Tsuzuki T. Anti-angiogenic function of tocotrienol. Asia Pac J Clin Nutr 2008;17 Suppl 1:253–256. [PubMed: 18296349] Aggarwal et al. Page 16 Biochem Pharmacol. Author manuscript; available in PMC 2011 December 1.

49. Nakagawa K, Eitsuka T, Inokuchi H, Miyazawa T. DNA chip analysis of comprehensive food function: inhibition of angiogenesis and telomerase activity with unsaturated vitamin E, tocotrienol. Biofactors 2004;21:5–10. [PubMed: 15630161]

50. Shibata A, Nakagawa K, Sookwong P, Tsuduki T, Oikawa S, Miyazawa T. delta-Tocotrienol suppresses VEGF induced angiogenesis whereas alpha-tocopherol does not. J Agric Food Chem 2009;57:8696–8704. [PubMed: 19702331]

51. Liu HK, Wang Q, Li Y, Sun WG, Liu JR, Yang YM, et al. Inhibitory effects of gamma-tocotrienol on invasion and metastasis of human gastric adenocarcinoma SGC-7901 cells. J Nutr Biochem 2010;21:206–213. [PubMed: 19195866]

52. Shirode AB, Sylvester PW. Synergistic anticancer effects of combined gamma-tocotrienol and celecoxib treatment are associated with suppression in Akt and NFkappaB signaling. Biomed Pharmacother 2010;64:327–332. [PubMed: 19954924]

53. Kuhad A, Chopra K. Attenuation of diabetic nephropathy by tocotrienol: involvement of NFkB signaling pathway. Life Sci 2009;84:296–301. [PubMed: 19162042]

54. Norazlina M, Lee PL, Lukman HI, Nazrun AS, Ima-Nirwana S. Effects of vitamin E supplementation on bone metabolism in nicotine-treated rats. Singapore Med J 2007;48:195–199. [PubMed: 17342286]

55. Ahmad NS, Khalid BA, Luke DA, Ima Nirwana S. Tocotrienol offers better protection than tocopherol from free radical-induced damage of rat bone. Clin Exp Pharmacol Physiol 2005;32:761–770. [PubMed: 16173934]

56. Wu SJ, Liu PL, Ng LT. Tocotrienol-rich fraction of palm oil exhibits anti-inflammatory property by suppressing the expression of inflammatory mediators in human monocytic cells. Mol Nutr Food Res 2008;52:921–929. [PubMed: 18481320]

57. Jiang Q, Yin X, Lill MA, Danielson ML, Freiser H, Huang J. Long-chain carboxychromanols, metabolites of vitamin E, are potent inhibitors of cyclooxygenases. Proc Natl Acad Sci U S A 2008;105:20464–20469. [PubMed: 19074288]

58. Kashiwagi K, Virgona N, Harada K, Kido W, Yano Y, Ando A, et al. A redox-silent analogue of tocotrienol acts as a potential cytotoxic agent against human mesothelioma cells. Life Sci 2009;84:650–656. [PubMed: 19232361]

59. Bachawal SV, Wali VB, Sylvester PW. Combined gamma-tocotrienol and erlotinib/gefitinib treatment suppresses Stat and Akt signaling in murine mammary tumor cells. Anticancer Res 2010;30:429–437. [PubMed: 20332450]

60. Ozer NK, Boscoboinik D, Azzi A. New roles of low density lipoproteins and vitamin E in the pathogenesis of atherosclerosis. Biochem Mol Biol Int 1995;35:117–124. [PubMed: 7735126]

61. Sen CK, Khanna S, Roy S, Packer L. Molecular basis of vitamin E action. Tocotrienol potently inhibits glutamate-induced pp60(c-Src) kinase activation and death of HT4 neuronal cells. J Biol Chem 2000;275:13049–13055. [PubMed: 10777609]

62. Sylvester PW, Shah SJ, Samant GV. Intracellular signaling mechanisms mediating the antiproliferative and apoptotic effects of gamma-tocotrienol in neoplastic mammary epithelial cells. J Plant Physiol 2005;162:803–810. [PubMed: 16008108]

63. Qureshi AA, Pearce BC, Nor RM, Gapor A, Peterson DM, Elson CE. Dietary alpha-tocopherol attenuates the impact of gamma-tocotrienol on hepatic 3-hydroxy-3-methylglutaryl coenzyme A reductase activity in chickens. J Nutr 1996;126:389–394. [PubMed: 8632210]

64. Elson CE, Qureshi AA. Coupling the cholesterol- and tumor-suppressive actions of palm oil to the impact of its minor constituents on 3-hydroxy-3-methylglutaryl coenzyme A reductase activity. Prostaglandins Leukot Essent Fatty Acids 1995;52:205–207. [PubMed: 7784459]

65. Iqbal J, Minhajuddin M, Beg ZH. Suppression of 7,12-dimethylbenz[alpha]anthracene-induced carcinogenesis and hypercholesterolaemia in rats by tocotrienol-rich fraction isolated from rice bran oil. Eur J Cancer Prev 2003;12:447–453. [PubMed: 14639121]

66. Parker RA, Pearce BC, Clark RW, Gordon DA, Wright JJ. Tocotrienols regulate cholesterol production in mammalian cells by post-transcriptional suppression of 3-hydroxy-3-methylglutarylcoenzyme A reductase. J Biol Chem 1993;268:11230–11238. [PubMed: 8388388]

67. Pearce BC, Parker RA, Deason ME, Qureshi AA, Wright JJ. Hypocholesterolemic activity of synthetic and natural tocotrienols. J Med Chem 1992;35:3595–3606. [PubMed: 1433170] Aggarwal et al. Page 17 Biochem Pharmacol. Author manuscript; available in PMC 2011 December 1.

68. Ali H, Shirode AB, Sylvester PW, Nazzal S. Preparation, characterization, and anticancer effects of simvastatin-tocotrienol lipid nanoparticles. Int J Pharm 2010;389:223–231. [PubMed: 20123009]

69. Bachawal SV, Wali VB, Sylvester PW. Enhanced antiproliferative and apoptotic response to combined treatment of gamma-tocotrienol with erlotinib or gefitinib in mammary tumor cells. BMC Cancer 2010;10:84. [PubMed: 20211018]

70. McIntyre BS, Briski KP, Gapor A, Sylvester PW. Antiproliferative and apoptotic effects of tocopherols and tocotrienols on preneoplastic and neoplastic mouse mammary epithelial cells. Proc Soc Exp Biol Med 2000;224:292–301. [PubMed: 10964265]

71. Wali VB, Bachawal SV, Sylvester PW. Suppression in mevalonate synthesis mediates antitumor effects of combined statin and gamma-tocotrienol treatment. Lipids 2009;44:925–934. [PubMed: 19777282]

72. Wali VB, Bachawal SV, Sylvester PW. Endoplasmic reticulum stress mediates gamma-tocotrienolinduced apoptosis in mammary tumor cells. Apoptosis 2009;14:1366–1377. [PubMed: 19771520]

73. Guthrie N, Gapor A, Chambers AF, Carroll KK. Inhibition of proliferation of estrogen receptornegative MDA-MB-435 and -positive MCF-7 human breast cancer cells by palm oil tocotrienols and tamoxifen, alone and in combination. J Nutr 1997;127:544S–548S. [PubMed: 9082043]

74. Nesaretnam K, Guthrie N, Chambers AF, Carroll KK. Effect of tocotrienols on the growth of a human breast cancer cell line in culture. Lipids 1995;30:1139–1143. [PubMed: 8614304]

75. Nesaretnam K, Stephen R, Dils R, Darbre P. Tocotrienols inhibit the growth of human breast cancer cells irrespective of estrogen receptor status. Lipids 1998;33:461–469. [PubMed: 9625593]

76. Shah S, Gapor A, Sylvester PW. Role of caspase-8 activation in mediating vitamin E-induced apoptosis in murine mammary cancer cells. Nutr Cancer 2003;45:236–246. [PubMed: 12881019]

77. Sylvester PW, Nachnani A, Shah S, Briski KP. Role of GTP-binding proteins in reversing the antiproliferative effects of tocotrienols in preneoplastic mammary epithelial cells. Asia Pac J Clin Nutr 2002;11 Suppl 7:S452–S459. [PubMed: 12492634]

78. Sylvester PW, Shah SJ. Mechanisms mediating the antiproliferative and apoptotic effects of vitamin E in mammary cancer cells. Front Biosci 2005;10:699–709. [PubMed: 15569611]

79. Yang Z, Xiao H, Jin H, Koo PT, Tsang DJ, Yang CS. Synergistic actions of atorvastatin with gammatocotrienol and celecoxib against human colon cancer HT29 and HCT116 cells. Int J Cancer 2010;126:852–863. [PubMed: 19626588]

80. Har CH, Keong CK. Effects of tocotrienols on cell viability and apoptosis in normal murine liver cells (BNL CL.2) and liver cancer cells (BNL 1ME A.7R.1), in vitro. Asia Pac J Clin Nutr 2005;14:374–380. [PubMed: 16326644]

81. Sakai M, Okabe M, Yamasaki M, Tachibana H, Yamada K. Induction of apoptosis by tocotrienol in rat hepatoma dRLh-84 cells. Anticancer Res 2004;24:1683–1688. [PubMed: 15274341]

82. Wada S, Satomi Y, Murakoshi M, Noguchi N, Yoshikawa T, Nishino H. Tumor suppressive effects of tocotrienol in vivo and in vitro. Cancer Lett 2005;229:181–191. [PubMed: 16098658]

83. Kashiwagi K, Harada K, Yano Y, Kumadaki I, Hagiwara K, Takebayashi J, et al. A redox-silent analogue of tocotrienol inhibits hypoxic adaptation of lung cancer cells. Biochem Biophys Res Commun 2008;365:875–881. [PubMed: 18042466]

84. Nakashima K, Virgona N, Miyazawa M, Watanabe T, Yano T. The tocotrienol-rich fraction from rice bran enhances cisplatin-induced cytotoxicity in human mesothelioma H28 cells. Phytother Res. 2010

85. Yano Y, Satoh H, Fukumoto K, Kumadaki I, Ichikawa T, Yamada K, et al. Induction of cytotoxicity in human lung adenocarcinoma cells by 6-O-carboxypropyl-alpha-tocotrienol, a redox-silent derivative of alpha-tocotrienol. Int J Cancer 2005;115:839–846. [PubMed: 15723336]

86. Chang PN, Yap WN, Lee DT, Ling MT, Wong YC, Yap YL. Evidence of gamma-tocotrienol as an apoptosis-inducing, invasion-suppressing, and chemotherapy drug-sensitizing agent in human melanoma cells. Nutr Cancer 2009;61:357–366. [PubMed: 19373609]

87. McAnally JA, Gupta J, Sodhani S, Bravo L, Mo H. Tocotrienols potentiate lovastatin-mediated growth suppression in vitro and in vivo. Exp Biol Med (Maywood) 2007;232:523–531. [PubMed: 17392488]

88. Hussein D, Mo H. d-Dlta-tocotrienol-mediated suppression of the proliferation of human PANC-1, MIA PaCa-2, and BxPC-3 pancreatic carcinoma cells. Pancreas 2009;38:e124–e136. [PubMed: 19346993]

89. Constantinou C, Hyatt JA, Vraka PS, Papas A, Papas KA, Neophytou C, et al. Induction of caspaseindependent programmed cell death by vitamin E natural homologs and synthetic derivatives. Nutr Cancer 2009;61:864–874. [PubMed: 20155628]

90. Conte C, Floridi A, Aisa C, Piroddi M, Galli F. Gamma-tocotrienol metabolism and antiproliferative effect in prostate cancer cells. Ann N Y Acad Sci 2004;1031:391–394. [PubMed: 15753178]

91. Yap WN, Chang PN, Han HY, Lee DT, Ling MT, Wong YC, et al. Gamma-tocotrienol suppresses prostate cancer cell proliferation and invasion through multiple-signalling pathways. Br J Cancer 2008;99:1832–1841. [PubMed: 19002171]

92. Comitato R, Leoni G, Canali R, Ambra R, Nesaretnam K, Virgili F. Tocotrienols activity in MCF-7 breast cancer cells: Involvement of ERbeta signal transduction. Mol Nutr Food Res 2010;64:327–332.

93. Kamat JP, Sarma HD, Devasagayam TP, Nesaretnam K, Basiron Y. Tocotrienols from palm oil as effective inhibitors of protein oxidation and lipid peroxidation in rat liver microsomes. Mol Cell Biochem 1997;170:131–137. [PubMed: 9144327]

94. Yu W, Simmons-Menchaca M, Gapor A, Sanders BG, Kline K. Induction of apoptosis in human breast cancer cells by tocopherols and tocotrienols. Nutr Cancer 1999;33:26–32. [PubMed: 10227040]

95. Chao JT, Gapor A, Theriault A. Inhibitory effect of delta-tocotrienol, a HMG CoA reductase inhibitor, on monocyte-endothelial cell adhesion. J Nutr Sci Vitaminol (Tokyo) 2002;48:332–337. [PubMed: 12656204]

96. Pearce BC, Parker RA, Deason ME, Dischino DD, Gillespie E, Qureshi AA, et al. Inhibitors of cholesterol biosynthesis. 2. Hypocholesterolemic and antioxidant activities of benzopyran and tetrahydronaphthalene analogues of the tocotrienols. J Med Chem 1994;37:526–541. [PubMed: 8120870]

97. Theriault A, Chao JT, Gapor A. Tocotrienol is the most effective vitamin E for reducing endothelial expression of adhesion molecules and adhesion to monocytes. Atherosclerosis 2002;160:21–30. [PubMed: 11755919]

98. Khanna S, Roy S, Parinandi NL, Maurer M, Sen CK. Characterization of the potent neuroprotective properties of the natural vitamin E alpha-tocotrienol. J Neurochem 2006;98:1474–1486. [PubMed: 16923160]

99. Liu X, Yamada N, Osawa T. Assessing the neuroprotective effect of antioxidative food factors by application of lipid-derived dopamine modification adducts. Methods Mol Biol 2009;580:143–152. [PubMed: 19784598]

100. Komiyama K, Iizuka K, Yamaoka M, Watanabe H, Tsuchiya N, Umezawa I. Studies on the biological activity of tocotrienols. Chem Pharm Bull (Tokyo) 1989;37:1369–1371. [PubMed: 2630104]

101. He L, Mo H, Hadisusilo S, Qureshi AA, Elson CE. Isoprenoids suppress the growth of murine B16 melanomas in vitro and in vivo. J Nutr 1997;127:668–674. [PubMed: 9164984]

102. Hiura Y, Tachibana H, Arakawa R, Aoyama N, Okabe M, Sakai M, et al. Specific accumulation of gamma- and delta-tocotrienols in tumor and their antitumor effect in vivo. J Nutr Biochem 2009;20:607–613. [PubMed: 18824342]

103. Nakagawa K, Shibata A, Yamashita S, Tsuzuki T, Kariya J, Oikawa S, et al. In vivo angiogenesis is suppressed by unsaturated vitamin E, tocotrienol. J Nutr 2007;137:1938–1943. [PubMed: 17634267]

104. Kumar KS, Raghavan M, Hieber K, Ege C, Mog S, Parra N, et al. Preferential radiation sensitizationof prostate cancer in nude mice by nutraceutical antioxidant gamma-tocotrienol. Life Sci 2006;78:2099–2104. [PubMed: 16413038]

105. Gould MN, Haag JD, Kennan WS, Tanner MA, Elson CE. A comparison of tocopherol and tocotrienol for the chemoprevention of chemically induced rat mammary tumors. Am J Clin Nutr 1991;53:1068S, 1070S. [PubMed: 1845366]

106. Goh SH, Hew NF, Norhanom AW, Yadav M. Inhibition of tumour promotion by various palm-oil tocotrienols. Int J Cancer 1994;57:529–531. [PubMed: 8181855]

107. Iqbal J, Minhajuddin M, Beg ZH. Suppression of diethylnitrosamine and 2-acetylaminofluoreneinduced hepatocarcinogenesis in rats by tocotrienol-rich fraction isolated from rice bran oil. Eur J Cancer Prev 2004;13:515–520. [PubMed: 15548946]

108. Ngah WZ, Jarien Z, San MM, Marzuki A, Top GM, Shamaan NA, et al. Effect of tocotrienols on hepatocarcinogenesis induced by 2-acetylaminofluorene in rats. Am J Clin Nutr 1991;53:1076S–1081S. [PubMed: 1672785]

109. Rahmat A, Ngah WZ, Shamaan NA, Gapor A, Abdul Kadir K. Long-term administration of tocotrienols and tumor-marker enzyme activities during hepatocarcinogenesis in rats. Nutrition 1993;9:229–232. [PubMed: 8102564]

110. Shamaan NA, Wan Ngah WZ, Ibrahim R, Jarien Z, Top AG, Abdul Kadir K. Effect of tocotrienol on the activities of cytosolic glutathione-dependent enzymes in rats treated with 2-acetylaminofluorene. Biochem Pharmacol 1993;45:1517–1519. [PubMed: 8471073]

111. Koba K, Abe K, Ikeda I, Sugano M. Effects of alpha-tocopherol and tocotrienols on blood pressure and linoleic acid metabolism in the spontaneously hypertensive rat (SHR). Biosci Biotechnol Biochem 1992;56:1420–1423. [PubMed: 1368948]

112. Chen CW, Cheng HH. A rice bran oil diet increases LDL-receptor and HMG-CoA reductase mRNA expressions and insulin sensitivity in rats with streptozotocin/nicotinamide-induced type 2 diabetes. J Nutr 2006;136:1472–1476. [PubMed: 16702306]

113. Chou TW, Ma CY, Cheng HH, Chen YY, Lai MH. A rice bran oil diet improves lipid abnormalities and suppress hyperinsulinemic responses in rats with streptozotocin/nicotinamide-induced type 2 diabetes. J Clin Biochem Nutr 2009;45:29–36. [PubMed: 19590704]

114. Nafeeza MI, Norzana AG, Jalaluddin HL, Gapor MT. The effects of a tocotrienol-rich fraction on experimentally induced atherosclerosis in the aorta of rabbits. Malays J Pathol 2001;23:17–25. [PubMed: 16329543]

115. Das S, Lekli I, Das M, Szabo G, Varadi J, Juhasz B, et al. Cardioprotection with palm oil tocotrienols: comparision of different isomers. Am J Physiol Heart Circ Physiol 2008;294:H970–H978. [PubMed: 18083895]

116. Qureshi AA, Peterson DM, Hasler-Rapacz JO, Rapacz J. Novel tocotrienols of rice bran suppress cholesterogenesis in hereditary hypercholesterolemic swine. J Nutr 2001;131:223–230. [PubMed: 11160537]

117. Esterhuyse AJ, Toit ED, Rooyen JV. Dietary red palm oil supplementation protects against the consequences of global ischemia in the isolated perfused rat heart. Asia Pac J Clin Nutr 2005;14:340–347. [PubMed: 16326640]

118. Das M, Das S, Wang P, Powell SR, Das DK. Caveolin and proteasome in tocotrienol mediated myocardial protection. Cell Physiol Biochem 2008;22:287–294. [PubMed: 18769056]

119. Lekli I, Ray D, Mukherjee S, Gurusamy N, Ahsan MK, Juhasz B, et al. Co-ordinated autophagy with resveratrol and gamma-tocotrienol confers synergetic cardioprotection. J Cell Mol Med. 2009

120. Wan Nazaimoon WM, Khalid BA. Tocotrienols-rich diet decreases advanced glycosylation endproducts in non-diabetic rats and improves glycemic control in streptozotocin-induced diabetic rats. Malays J Pathol 2002;24:77–82. [PubMed: 12887164]

121. Budin SB, Othman F, Louis SR, Bakar MA, Das S, Mohamed J. The effects of palm oil tocotrienolrich fraction supplementation on biochemical parameters, oxidative stress and the vascular wall of streptozotocin-induced diabetic rats. Clinics (Sao Paulo) 2009;64:235–244. [PubMed: 19330251]

122. Kuhad A, Bishnoi M, Tiwari V, Chopra K. Suppression of NF-kappabeta signaling pathway by tocotrienol can prevent diabetes associated cognitive deficits. Pharmacol Biochem Behav 2009;92:251–259. [PubMed: 19138703]

123. Kuhad A, Chopra K. Tocotrienol attenuates oxidative-nitrosative stress and inflammatory cascade in experimental model of diabetic neuropathy. Neuropharmacology 2009;57:456–462. [PubMed: 19555701]

124. Kanaya Y, Doi T, Sasaki H, Fujita A, Matsuno S, Okamoto K, et al. Rice bran extract prevents the elevation of plasma peroxylipid in KKAy diabetic mice. Diabetes Res Clin Pract 2004;66 Suppl 1:S157–S160. [PubMed: 15563968]

125. Yoshida Y, Hayakawa M, Habuchi Y, Itoh N, Niki E. Evaluation of lipophilic antioxidant efficacy in vivo by the biomarkers hydroxyoctadecadienoic acid and isoprostane. Lipids 2007;42:463–472. [PubMed: 17476550]

126. Fang F, Kang Z, Wong C. Vitamin E tocotrienols improve insulin sensitivity through activating peroxisome proliferator-activated receptors. Mol Nutr Food Res 2010;54:345–352. [PubMed: 19866471]

127. Tiwari V, Kuhad A, Bishnoi M, Chopra K. Chronic treatment with tocotrienol, an isoform of vitamin E, prevents intracerebroventricular streptozotocin-induced cognitive impairment and oxidativenitrosative stress in rats. Pharmacol Biochem Behav 2009;93:183–189. [PubMed: 19464315]

128. Tiwari V, Kuhad A, Chopra K. Tocotrienol ameliorates behavioral and biochemical alterations in the rat model of alcoholic neuropathy. Pain 2009;145:129–135. [PubMed: 19541419]

129. Tiwari V, Kuhad A, Chopra K. Suppression of neuro-inflammatory signaling cascade by tocotrienol can prevent chronic alcohol-induced cognitive dysfunction in rats. Behav Brain Res 2009;203:296–303. [PubMed: 19464322]

130. Khanna S, Parinandi NL, Kotha SR, Roy S, Rink C, Bibus D, et al. Nanomolar vitamin E alphatocotrienol inhibits glutamate-induced activation of phospholipase A2 and causes neuroprotection. J Neurochem 2010;112:1249–1260. [PubMed: 20028458]

131. Khanna S, Roy S, Slivka A, Craft TK, Chaki S, Rink C, et al. Neuroprotective properties of the natural vitamin E alpha-tocotrienol. Stroke 2005;36:2258–2264. [PubMed: 16166580]

132. Roy S, Lado BH, Khanna S, Sen CK. Vitamin E sensitive genes in the developing rat fetal brain: a high-density oligonucleotide microarray analysis. FEBS Lett 2002;530:17–23. [PubMed: 12387859]

133. Mishima K, Tanaka T, Pu F, Egashira N, Iwasaki K, Hidaka R, et al. Vitamin E isoforms alphatocotrienol and gamma-tocopherol prevent cerebral infarction in mice. Neurosci Lett 2003;337:56– 60. [PubMed: 12524170]

134. Hermizi H, Faizah O, Ima-Nirwana S, Ahmad Nazrun S, Norazlina M. Beneficial effects of tocotrienol and tocopherol on bone histomorphometric parameters in sprague-dawley male rats after nicotine cessation. Calcif Tissue Int 2009;84:65–74. [PubMed: 19020790]

135. Mehat MZ, Shuid AN, Mohamed N, Muhammad N, Soelaiman IN. Beneficial effects of vitamin E isomer supplementation on static and dynamic bone histomorphometry parameters in normal male rats. J Bone Miner Metab. 2010

136. Shuid AN, Mehat Z, Mohamed N, Muhammad N, Soelaiman IN. Vitamin E exhibits bone anabolic actions in normal male rats. J Bone Miner Metab 2010;28:149–156. [PubMed: 19779668]

137. Maniam S, Mohamed N, Shuid AN, Soelaiman IN. Palm tocotrienol exerted better antioxidant activities in bone than alpha-tocopherol. Basic Clin Pharmacol Toxicol 2008;103:55–60. [PubMed: 18598299]

138. Norazlina M, Ima-Nirwana S, Abul Gapor MT, Abdul Kadir Khalid B. Tocotrienols are needed for normal bone calcification in growing female rats. Asia Pac J Clin Nutr 2002;11:194–199. [PubMed: 12230232]

139. Ima-Nirwana S, Suhaniza S. Effects of tocopherols and tocotrienols on body composition and bone calcium content in adrenalectomized rats replaced with dexamethasone. J Med Food 2004;7:45–51. [PubMed: 15117552]

140. Gu JY, Wakizono Y, Sunada Y, Hung P, Nonaka M, Sugano M, et al. Dietary effect of tocopherolsand tocotrienols on the immune function of spleen and mesenteric lymph node lymphocytes in Brown Norway rats. Biosci Biotechnol Biochem 1999;63:1697–1702. [PubMed: 10586497]

141. Azlina MF, Nafeeza MI, Khalid BA. A comparison between tocopherol and tocotrienol effects ongastric parameters in rats exposed to stress. Asia Pac J Clin Nutr 2005;14:358–365. [PubMed: 16326642]

142. Nafeeza MI, Fauzee AM, Kamsiah J, Gapor MT. Comparative effects of a tocotrienol-rich fraction and tocopherol in aspirin-induced gastric lesions in rats. Asia Pac J Clin Nutr 2002;11:309–313. [PubMed: 12495264]

143. Freiser H, Jiang Q. Gamma-tocotrienol and gamma-tocopherol are primarily metabolized to conjugated 2-(beta-carboxyethyl)-6-hydroxy-2,7,8-trimethylchroman and sulfated long-chain carboxychromanols in rats. J Nutr 2009;139:884–889. [PubMed: 19297424]

144. Hattori A, Fukushima T, Yoshimura H, Abe K, Imai K. Production of LLU-alpha following an oral administration of gamma-tocotrienol or gamma-tocopherol to rats. Biol Pharm Bull 2000;23:1395– 1397. [PubMed: 11085376]

145. Husain K, Francois RA, Hutchinson SZ, Neuger AM, Lush R, Coppola D, et al. Vitamin E deltatocotrienol levels in tumor and pancreatic tissue of mice after oral administration. Pharmacology 2009;83:157–163. [PubMed: 19142032]

146. Ikeda I, Imasato Y, Sasaki E, Sugano M. Lymphatic transport of alpha-, gamma- and deltatocotrienols and alpha-tocopherol in rats. Int J Vitam Nutr Res 1996;66:217–221. [PubMed: 8899454]

147. Ikeda S, Niwa T, Yamashita K. Selective uptake of dietary tocotrienols into rat skin. J Nutr Sci Vitaminol (Tokyo) 2000;46:141–143. [PubMed: 10955281]

148. Ikeda S, Toyoshima K, Yamashita K. Dietary sesame seeds elevate alpha- and gamma-tocotrienol concentrations in skin and adipose tissue of rats fed the tocotrienol-rich fraction extracted from palm oil. J Nutr 2001;131:2892–2897. [PubMed: 11694614]

149. Kawakami Y, Tsuzuki T, Nakagawa K, Miyazawa T. Distribution of tocotrienols in rats fed a rice bran tocotrienol concentrate. Biosci Biotechnol Biochem 2007;71:464–471. [PubMed: 17284857]

150. Khosla P, Patel V, Whinter JM, Khanna S, Rakhkovskaya M, Roy S, et al. Postprandial levels of the natural vitamin E tocotrienol in human circulation. Antioxid Redox Signal 2006;8:1059–1068. [PubMed: 16771695]

151. Saito Y, Yoshida Y, Nishio K, Hayakawa M, Niki E. Characterization of cellular uptake and distribution of vitamin E. Ann N Y Acad Sci 2004;1031:368–375. [PubMed: 15753172]

152. Tanito M, Itoh N, Yoshida Y, Hayakawa M, Ohira A, Niki E. Distribution of tocopherols and tocotrienols to rat ocular tissues after topical ophthalmic administration. Lipids 2004;39:469–474. [PubMed: 15506242]

153. Tasaki M, Umemura T, Inoue T, Okamura T, Kuroiwa Y, Ishii Y, et al. Induction of characteristic hepatocyte proliferative lesion with dietary exposure of Wistar Hannover rats to tocotrienol for 1 year. Toxicology 2008;250:143–150. [PubMed: 18675878]

154. Tsuzuki W, Yunoki R, Yoshimura H. Intestinal epithelial cells absorb gamma-tocotrienol faster than alpha-tocopherol. Lipids 2007;42:163–170. [PubMed: 17393222]

155. Yap SP, Yuen KH, Lim AB. Influence of route of administration on the absorption and disposition of alpha-, gamma- and delta-tocotrienols in rats. J Pharm Pharmacol 2003;55:53–58. [PubMed: 12625867]

156. Qureshi AA, Qureshi N, Hasler-Rapacz JO, Weber FE, Chaudhary V, Crenshaw TD, et al. Dietary tocotrienols reduce concentrations of plasma cholesterol, apolipoprotein B, thromboxane B2, and platelet factor 4 in pigs with inherited hyperlipidemias. Am J Clin Nutr 1991;53:1042S–1046S. [PubMed: 2012015]

157. Nakamura H, Furukawa F, Nishikawa A, Miyauchi M, Son HY, Imazawa T, et al. Oral toxicity of a tocotrienol preparation in rats. Food Chem Toxicol 2001;39:799–805. [PubMed: 11434987]

158. O'Byrne D, Grundy S, Packer L, Devaraj S, Baldenius K, Hoppe PP, et al. Studies of LDL oxidation following alpha-, gamma-, or delta-tocotrienyl acetate supplementation of hypercholesterolemic humans. Free Radic Biol Med 2000;29:834–845. [PubMed: 11063909]

159. Hayes KC, Pronczuk A, Liang JS. Differences in the plasma transport and tissue concentrations of tocopherols and tocotrienols: observations in humans and hamsters. Proc Soc Exp Biol Med 1993;202:353–539. [PubMed: 8437992]

160. Yap SP, Yuen KH, Wong JW. Pharmacokinetics and bioavailability of alpha-, gamma- and deltatocotrienols under different food status. J Pharm Pharmacol 2001;53:67–71. [PubMed: 11206194]

161. Lodge JK, Ridlington J, Leonard S, Vaule H, Traber MG. Alpha- and gamma-tocotrienols are metabolized to carboxyethyl-hydroxychroman derivatives and excreted in human urine. Lipids 2001;36:43–48. [PubMed: 11214728]

162. Yap SP, Yuen KH. Influence of lipolysis and droplet size on tocotrienol absorption from selfemulsifying formulations. Int J Pharm 2004;281:67–78. [PubMed: 15288344]

163. Ajuluchukwu JN, Okubadejo NU, Mabayoje M, Ojini FI, Okwudiafor RN, Mbakwem AC, et al. Comparative study of the effect of tocotrienols and -tocopherol on fasting serum lipid profiles in patients with mild hypercholesterolaemia: a preliminary report. Niger Postgrad Med J 2007;14:30– 33. [PubMed: 17356586]

164. Baliarsingh S, Beg ZH, Ahmad J. The therapeutic impacts of tocotrienols in type 2 diabetic patients with hyperlipidemia. Atherosclerosis 2005;182:367–374. [PubMed: 16159610]

165. Qureshi AA, Bradlow BA, Brace L, Manganello J, Peterson DM, Pearce BC, et al. Response of hypercholesterolemic subjects to administration of tocotrienols. Lipids 1995;30:1171–1177. [PubMed: 8614309]

166. Qureshi AA, Bradlow BA, Salser WA, Brace LD. Novel tocotrienols of rice bran modulate cardiovascular disease risk parameters of hypercholesterolemic humans. Nutritional Biochemistry 1997;8:290–298.

167. Qureshi AA, Qureshi N, Wright JJ, Shen Z, Kramer G, Gapor A, et al. Lowering of serum cholesterol in hypercholesterolemic humans by tocotrienols (palmvitee). Am J Clin Nutr 1991;53:1021S– 1026S. [PubMed: 2012010]

168. Rasool AH, Rahman AR, Yuen KH, Wong AR. Arterial compliance and vitamin E blood levels with a self emulsifying preparation of tocotrienol rich vitamin E. Arch Pharm Res 2008;31:1212– 1217. [PubMed: 18806966]

169. Roza JM, Xian-Liu Z, Guthrie N. Effect of citrus flavonoids and tocotrienols on serum cholesterol levels in hypercholesterolemic subjects. Altern Ther Health Med 2007;13:44–48. [PubMed: 17985810]

170. Tan DT, Khor HT, Low WH, Ali A, Gapor A. Effect of a palm-oil-vitamin E concentrate on the serum and lipoprotein lipids in humans. Am J Clin Nutr 1991;53:1027S–1030S. [PubMed: 2012011]

171. Tomeo AC, Geller M, Watkins TR, Gapor A, Bierenbaum ML. Antioxidant effects of tocotrienols in patients with hyperlipidemia and carotid stenosis. Lipids 1995;30:1179–1183. [PubMed: 8614310]

172. Mensink RP, van Houwelingen AC, Kromhout D, Hornstra G. A vitamin E concentrate rich in tocotrienols had no effect on serum lipids, lipoproteins, or platelet function in men with mildly elevated serum lipid concentrations. Am J Clin Nutr 1999;69:213–219. [PubMed: 9989682]

173. Mustad VA, Smith CA, Ruey PP, Edens NK, DeMichele SJ. Supplementation with 3 compositionally different tocotrienol supplements does not improve cardiovascular disease risk factors in men and women with hypercholesterolemia. Am J Clin Nutr 2002;76:1237–1243. [PubMed: 12450888]

174. Rasool AH, Yuen KH, Yusoff K, Wong AR, Rahman AR. Dose dependent elevation of plasma tocotrienol levels and its effect on arterial compliance, plasma total antioxidant status, and lipid profile in healthy humans supplemented with tocotrienol rich vitamin E. J Nutr Sci Vitaminol (Tokyo) 2006;52:473–478. [PubMed: 17330512]

175. Stampfer MJ, Willet W, Castelli WP, Taylor JO, Fine J, Hennekens CH. Effect of vitamin E on lipids. Am J Clin Pathol 1983;79:714–716. [PubMed: 6342362]

176. Wahlqvist ML, Krivokuca-Bogetic Z, Lo CS, Hage B, Smith R, Lukito W. Differential serum responses of tocopherols and tocotrienols during vitamin supplementation in hypercholesterolemic individuals without change in coronary risk factors. Nutr Res 1992;12 supple

177. Anderson SL, Rubin BY. Tocotrienols reverse IKAP and monoamine oxidase deficiencies in familial dysautonomia. Biochem Biophys Res Commun 2005;336:150–156. [PubMed: 16125677]

178. Rubin BY, Anderson SL, Kapas L. Can the therapeutic efficacy of tocotrienols in neurodegenerative familial dysautonomia patients be measured clinically? Antioxid Redox Signal 2008;10:837–841. [PubMed: 18177231]

179. Chin SF, Hamid NA, Latiff AA, Zakaria Z, Mazlan M, Yusof YA, et al. Reduction of DNA damage in older healthy adults by Tri E Tocotrienol supplementation. Nutrition 2008;24:1–10. [PubMed: 17884341]

180. Weber SU, Thiele JJ, Han N, Luu C, Valacchi G, Weber S, et al. Topical alpha-tocotrienol supplementation inhibits lipid peroxidation but fails to mitigate increased transepidermal water loss after benzoyl peroxide treatment of human skin. Free Radic Biol Med 2003;34:170–176. [PubMed: 12521598]

181. Serbinova E, Kagan V, Han D, Packer L. Free radical recycling and intramembrane mobility in the antioxidant properties of alpha-tocopherol and alpha-tocotrienol. Free Radic Biol Med 1991;10:263–275. [PubMed: 1649783]

182. Suzuki YJ, Tsuchiya M, Wassall SR, Choo YM, Govil G, Kagan VE, et al. Structural and dynamic membrane properties of alpha-tocopherol and alpha-tocotrienol: implication to the molecular mechanism of their antioxidant potency. Biochemistry 1993;32:10692–10699. [PubMed: 8399214]

183. Yoshida Y, Niki E, Noguchi N. Comparative study on the action of tocopherols and tocotrienols as antioxidant: chemical and physical effects. Chem Phys Lipids 2003;123:63–75. [PubMed: 12637165]

184. Birringer M, Pfluger P, Kluth D, Landes N, Brigelius-Flohe R. Identities and differences in the metabolism of tocotrienols and tocopherols in HepG2 cells. J Nutr 2002;132:3113–3118. [PubMed: 12368403]

185. Kamal-Eldin A, Appelqvist LA. The chemistry and antioxidant properties of tocopherols and tocotrienols. Lipids 1996;31:671–701. [PubMed: 8827691]

186. Sigounas G, Anagnostou A, Steiner M. dl-alpha-tocopherol induces apoptosis in erythroleukemia, prostate, and breast cancer cells. Nutr Cancer 1997;28:30–35. [PubMed: 9200147]

187. Hosomi A, Arita M, Sato Y, Kiyose C, Ueda T, Igarashi O, et al. Affinity for alpha-tocopherol transfer protein as a determinant of the biological activities of vitamin E analogs. FEBS Lett 1997;409:105–108. [PubMed: 9199513]

188. Inokuchi H, Hirokane H, Tsuzuki T, Nakagawa K, Igarashi M, Miyazawa T. Anti-angiogenic activity of tocotrienol. Biosci Biotechnol Biochem 2003;67:1623–1627. [PubMed: 12913317]

189. Miyazawa T, Tsuzuki T, Nakagawa K, Igarashi M. Antiangiogenic potency of vitamin E. Ann N Y Acad Sci 2004;1031:401–404. [PubMed: 15753181]

190. Suarna C, Hood RL, Dean RT, Stocker R. Comparative antioxidant activity of tocotrienols and other natural lipid-soluble antioxidants in a homogeneous system, and in rat and human lipoproteins. Biochim Biophys Acta 1993;1166:163–170. [PubMed: 8443232]

191. Mazlan M, Sue Mian T, Mat Top G, Zurinah Wan Ngah W. Comparative effects of alpha-tocopherol and gamma-tocotrienol against hydrogen peroxide induced apoptosis on primary-cultured astrocytes. J Neurol Sci 2006;243:5–12. [PubMed: 16442562]

192. Shichiri M, Takanezawa Y, Uchida K, Tamai H, Arai H. Protection of cerebellar granule cells by tocopherols and tocotrienols against methylmercury toxicity. Brain Res 2007;1182:106–115. [PubMed: 17949699]

193. Yam ML, Abdul Hafid SR, Cheng HM, Nesaretnam K. Tocotrienols suppress proinflammatory markers and cyclooxygenase-2 expression in RAW264.7 macrophages. Lipids 2009;44:787–797. [PubMed: 19655189]

194. Serbinova EA, Packer L. Antioxidant properties of alpha-tocopherol and alpha-tocotrienol. Methods Enzymol 1994;234:354–366. [PubMed: 7808307]

195. Miyazawa T, Inokuchi H, Hirokane H, Tsuzuki T, Nakagawa K, Igarashi M. Anti-angiogenic potential of tocotrienol in vitro. Biochemistry (Mosc) 2004;69:67–69. [PubMed: 14972020]

196. Lippman SM, Klein EA, Goodman PJ, Lucia MS, Thompson IM, Ford LG, et al. Effect of selenium and vitamin E on risk of prostate cancer and other cancers: the Selenium and Vitamin E Cancer Prevention Trial (SELECT). JAMA 2009;301:39–51. [PubMed: 19066370]

197. Lonn E, Bosch J, Yusuf S, Sheridan P, Pogue J, Arnold JM, et al. Effects of long-term vitamin E supplementation on cardiovascular events and cancer: a randomized controlled trial. JAMA 2005;293:1338–1347. [PubMed: 15769967]

198. Schroeder MT, Becker EM, Skibsted LH. Molecular mechanism of antioxidant synergism of tocotrienols and carotenoids in palm oil. J Agric Food Chem 2006;54:3445–3453. [PubMed: 16637706]

199. Sookwong P, Nakagawa K, Yamaguchi Y, Miyazawa T, Kato S, Kimura F. Tocotrienol distribution in foods: estimation of daily tocotrienol intake of Japanese population. J Agric Food Chem 2010;58:3350–3355. [PubMed: 20158257]

200. Hassanein MM, Abedel-Razek AG. Chromatographic quantitation of some bioactive minor components in oils of wheat germ and grape seeds produced as by-products. J Oleo Sci 2009;58:227–233. [PubMed: 19367078]

201. Amaral JS, Casal S, Alves MR, Seabra RM, Oliveira BP. Tocopherol and tocotrienol content of hazelnut cultivars grown in portugal. J Agric Food Chem 2006;54:1329–1336. [PubMed: 16478256]

202. Cunha SC, Amaral JS, Fernandes JO, Oliveira MB. Quantification of tocopherols and tocotrienols in portuguese olive oils using HPLC with three different detection systems. J Agric Food Chem 2006;54:3351–3356. [PubMed: 16637695]

203. Kallio H, Yang B, Peippo P, Tahvonen R, Pan R. Triacylglycerols, glycerophospholipids, tocopherols, and tocotrienols in berries and seeds of two subspecies (ssp. sinensis and mongolica) of Sea Buckthorn (Hippophae rhamnoides). J Agric Food Chem 2002;50:3004–3009. [PubMed: 11982433]

204. Milagros Delgado-Zamarreno M, Bustamante-Rangel M, Sierra-Manzano S, Verdugo-Jara M, Carabias-Martinez R. Simultaneous extraction of tocotrienols and tocopherols from cereals using pressurized liquid extraction prior to LC determination. J Sep Sci 2009;32:1430–1436. [PubMed: 19330788]

205. Panfili G, Fratianni A, Irano M. Normal phase high-performance liquid chromatography method for the determination of tocopherols and tocotrienols in cereals. J Agric Food Chem 2003;51:3940–3944. [PubMed: 12822927]

206. Bozan B, Temelli F. Chemical composition and oxidative stability of flax, safflower and poppy seed and seed oils. Bioresour Technol 2008;99:6354–6359. [PubMed: 18198133]

207. Hafid SR, Radhakrishnan AK, Nesaretnam K. Tocotrienols are good adjuvants for developing cancer vaccines. BMC Cancer 2010;10:5. [PubMed: 20051142]

208. Naito Y, Shimozawa M, Kuroda M, Nakabe N, Manabe H, Katada K, et al. Tocotrienols reduce 25-hydroxycholesterol-induced monocyte-endothelial cell interaction by inhibiting the surface expression of adhesion molecules. Atherosclerosis 2005;180:19–25. [PubMed: 15823271]

209. Noguchi N, Hanyu R, Nonaka A, Okimoto Y, Kodama T. Inhibition of THP-1 cell adhesion to endothelial cells by alpha-tocopherol and alpha-tocotrienol is dependent on intracellular concentration of the antioxidants. Free Radic Biol Med 2003;34:1614–1620. [PubMed: 12788481]

210. Mizushina Y, Nakagawa K, Shibata A, Awata Y, Kuriyama I, Shimazaki N, et al. Inhibitory effect of tocotrienol on eukaryotic DNA polymerase lambda and angiogenesis. Biochem Biophys Res Commun 2006;339:949–955. [PubMed: 16325764]

211. Makpol S, Shamaan NA, Jarien Z, Top AG, Khalid BA, Wan Ngah WZ. Different starting times of alpha-tocopherol and gamma-tocotrienol supplementation and tumor marker enzyme activities in the rat chemically induced with cancer. Gen Pharmacol 1997;28:589–592. [PubMed: 9147029]

212. Ong FB, Wan Ngah WZ, Shamaan NA, Md Top AG, Marzuki A, Khalid AK. Glutathione Stransferase and gamma-glutamyl transpeptidase activities in cultured rat hepatocytes treated with tocotrienol and tocopherol. Comp Biochem Physiol C 1993;106:237–240. [PubMed: 7903615]

213. Ong FB, Wan Ngah WZ, Top AG, Khalid BA, Shamaan NA. Vitamin E, glutathione S-transferase and gamma-glutamyl transpeptidase activities in cultured hepatocytes of rats treated with carcinogens. Int J Biochem 1994;26:397–402. [PubMed: 7910569]

214. Shibata A, Nakagawa K, Sookwong P, Tsuzuki T, Oikawa S, Miyazawa T. Tumor anti-angiogenic effect and mechanism of action of delta-tocotrienol. Biochem Pharmacol 2008;76:330–339. [PubMed: 18599020]

215. Landes N, Pfluger P, Kluth D, Birringer M, Ruhl R, Bol GF, et al. Vitamin E activates gene expression via the pregnane × receptor. Biochem Pharmacol 2003;65:269–273. [PubMed: 12504802]

216. Uto-Kondo H, Ohmori R, Kiyose C, Kishimoto Y, Saito H, Igarashi O, et al. Tocotrienol suppresses adipocyte differentiation and Akt phosphorylation in 3T3-L1 preadipocytes. J Nutr 2009;139:51–57. [PubMed: 19056650]

217. Bi S, Liu JR, Li Y, Wang Q, Liu H, Yan YG, et al. gamma-Tocotrienol modulates the paracrine secretion of VEGF induced by cobalt (II) chloride via ERK signaling pathway in gastric adenocarcinoma SGC-7901 cell line. Toxicology. 2010

218. Li F, Tan W, Kang Z, Wong CW. Tocotrienol enriched palm oil prevents atherosclerosis through modulating the activities of peroxisome proliferators-activated receptors. Atherosclerosis. 2010

219. Wang Q, Theriault A, Gapor A, Adeli K. Effects of tocotrienol on the intracellular translocation and degradation of apolipoprotein B: possible involvement of a proteasome independent pathway. Biochem Biophys Res Commun 1998;246:640–643. [PubMed: 9618265]

220. Theriault A, Wang Q, Gapor A, Adeli K. Effects of gamma-tocotrienol on ApoB synthesis, degradation, and secretion in HepG2 cells. Arterioscler Thromb Vasc Biol 1999;19:704–712. [PubMed: 10073977]

221. Galli F, Stabile AM, Betti M, Conte C, Pistilli A, Rende M, et al. The effect of alpha- and gammatocopherol and their carboxyethyl hydroxychroman metabolites on prostate cancer cell proliferation. Arch Biochem Biophys 2004;423:97–102. [PubMed: 14871472]

222. Brigelius-Flohe R. Induction of drug metabolizing enzymes by vitamin E. J Plant Physiol 2005;162:797–802. [PubMed: 16008107]

223. Clarke MW, Burnett JR, Croft KD. Vitamin E in human health and disease. Crit Rev Clin Lab Sci 2008;45:417–450. [PubMed: 18712629]

224. Yap WN, Zaiden N, Tan YL, Ngoh CP, Zhang XW, Wong YC, et al. Id1, inhibitor of differentiation, is a key protein mediating anti-tumor responses of gamma-tocotrienol in breast cancer cells. Cancer Lett 2010;291:187–199. [PubMed: 19926394]

225. McIntyre BS, Briski KP, Tirmenstein MA, Fariss MW, Gapor A, Sylvester PW. Antiproliferative and apoptotic effects of tocopherols and tocotrienols on normal mouse mammary epithelial cells. Lipids 2000;35:171–180. [PubMed: 10757548]

226. Wali VB, Sylvester PW. Synergistic antiproliferative effects of gamma-tocotrienol and statin treatment on mammary tumor cells. Lipids 2007;42:1113–1123. [PubMed: 17701065]

227. Saito H, Kiyose C, Yoshimura H, Ueda T, Kondo K, Igarashi O. Gamma-tocotrienol, a vitamin E homolog, is a natriuretic hormone precursor. J Lipid Res 2003;44:1530–1535. [PubMed: 12730299]

228. Kausar H, Bhasin G, Zargar MA, Athar M. Palm oil alleviates 12-O-tetradecanoyl-phorbol-13- acetate-induced tumor promotion response in murine skin. Cancer Lett 2003;192:151–160. [PubMed: 12668279]

229. Nesaretnam K, Ambra R, Selvaduray KR, Radhakrishnan A, Reimann K, Razak G, et al. Tocotrienolrich fraction from palm oil affects gene expression in tumors resulting from MCF-7 cell inoculation in athymic mice. Lipids 2004;39:459–467. [PubMed: 15506241]

230. Yamada Y, Obayashi M, Ishikawa T, Kiso Y, Ono Y, Yamashita K. Dietary tocotrienol reduces UVB-induced skin damage and sesamin enhances tocotrienol effects in hairless mice. J Nutr Sci Vitaminol (Tokyo) 2008;54:117–123. [PubMed: 18490840]

231. Yap WN, Zaiden N, Luk SY, Lee DT, Ling MT, Wong YC, et al. In vivo Evidence of gamma- Tocotrienol as a Chemosensitizer in the Treatment of Hormone-Refractory Prostate Cancer. Pharmacology 2010;85:248–258. [PubMed: 20375535]

232. Qureshi AA, Salser WA, Parmar R, Emeson EE. Novel tocotrienols of rice bran inhibit atherosclerotic lesions in C57BL/6 ApoE-deficient mice. J Nutr 2001;131:2606–2618. [PubMed: 11584079]

233. Newaz MA, Yousefipour Z, Nawal N, Adeeb N. Nitric oxide synthase activity in blood vessels of spontaneously hypertensive rats: antioxidant protection by gamma-tocotrienol. J Physiol Pharmacol 2003;54:319–327. [PubMed: 14566071]

234. Nakano M, Onodera A, Saito E, Tanabe M, Yajima K, Takahashi J, et al. Effect of astaxanthin in combination with alpha-tocopherol or ascorbic acid against oxidative damage in diabetic ODS rats. J Nutr Sci Vitaminol (Tokyo) 2008;54:329–334. [PubMed: 18797156]

235. Ima-Nirwana S, Kiftiah A, Sariza T, Gapor MT, Khalid BAK. Palm vitamin E improves bone metabolism and survival rate in thyrotoxic rats. Gen Pharmacol 1999;32:621–662. Abd. [PubMed: 10382867]

236. Rukmini C, Raghuram TC. Nutritional and biochemical aspects of the hypolipidemic action of rice bran oil: a review. J Am Coll Nutr 1991;10:593–601. [PubMed: 1770191] 237. Khor HT, Ng TT. Effects of administration of alpha-tocopherol and tocotrienols on serum lipids and liver HMG CoA reductase activity. Int J Food Sci Nutr 2000;51 Suppl:S3–S11. [PubMed: 11271854]

238. Poveda E, Ayala P, Milena R, Ordonez E, Baracaldo C, Delgado W, et al. [Effects of vegetal oil supplementation on the lipid profile of Wistar rats]. Biomedica 2005;25:101–109. [PubMed: 15962906]

239. Gupta A, Chopra K. Effect of tocotrienols on iron-induced renal dysfunction and oxidative stress in rats. Drug Chem Toxicol 2009;32:319–325. [PubMed: 19793023]

240. Fairus S, Nor RM, Cheng HM, Sundram K. Postprandial metabolic fate of tocotrienol-rich vitamin E differs significantly from that of alpha-tocopherol. Am J Clin Nutr 2006;84:835–842. [PubMed: 17023711]

241. Radhakrishnan AK, Lee AL, Wong PF, Kaur J, Aung H, Nesaretnam K. Daily supplementation of tocotrienol-rich fraction or alpha-tocopherol did not induce immunomodulatory changes in healthy human volunteers. Br J Nutr 2009;101:810–815. [PubMed: 18702848]

242. Rautalahti M, Albanes D, Hyvonen L, Piironen V, Heinonen M. Effect of sampling site on retinol, carotenoid, tocopherol, and tocotrienol concentration of adipose tissue of human breast with cancer. Ann Nutr Metab 1990;34:37–41. [PubMed: 2331139]

243. Nesaretnam K, Gomez PA, Selvaduray KR, Razak GA. Tocotrienol levels in adipose tissue of benign and malignant breast lumps in patients in Malaysia. Asia Pac J Clin Nutr 2007;16:498–504. [PubMed: 17704032]

244. Weinstein SJ, Wright ME, Lawson KA, Snyder K, Mannisto S, Taylor PR, et al. Serum and dietary vitamin E in relation to prostate cancer risk. Cancer Epidemiol Biomarkers Prev 2007;16:1253–1259. [PubMed: 17548693]

245. Nur Azlina MF, Nafeeza MI. Tocotrienol and alpha-tocopherol reduce corticosterone and noradrenalin levels in rats exposed to restraint stress. Pharmazie 2008;63:890–892. [PubMed: 19177905]

246. Kamat JP, Devasagayam TP. Tocotrienols from palm oil as potent inhibitors of lipid peroxidation and protein oxidation in rat brain mitochondria. Neurosci Lett 1995;195:179–182. [PubMed: 8584204]

247. Raederstorff D, Elste V, Aebischer C, Weber P. Effect of either gamma-tocotrienol or a tocotrienol mixture on the plasma lipid profile in hamsters. Ann Nutr Metab 2002;46:17–23. [PubMed: 11914511]

248. Osakada F, Hashino A, Kume T, Katsuki H, Kaneko S, Akaike A. Alpha-tocotrienol provides the most potent neuroprotection among vitamin E analogs on cultured striatal neurons. Neuropharmacology 2004;47:904–915. [PubMed: 15527824]

249. Palozza P, Verdecchia S, Avanzi L, Vertuani S, Serini S, Iannone A, et al. Comparative antioxidant activity of tocotrienols and the novel chromanyl-polyisoprenyl molecule FeAox-6 in isolated membranes and intact cells. Mol Cell Biochem 2006;287:21–32. [PubMed: 16645729]

250. Song BL, DeBose-Boyd RA. Insig-dependent ubiquitination and degradation of 3-hydroxy-3- methylglutaryl coenzyme a reductase stimulated by delta- and gamma-tocotrienols. J Biol Chem 2006;281:25054–25061. [PubMed: 16831864]

Combination Efect in Delta Tocotrienol y Gamma Tocoferol on protaste cancer cell growth

(Chiaki Sato et al, 2017, J of Nut Sci Vitaminol, 63, 349-354, 2017)

Summary Tocotrienols (T3s) and tocopherols (Tocs) are both members of the vitamin E family. It is known that δ-tocotrienol (δ-T3) has displayed the most potent anti-cancer activity amongst the tocotrienols. On the other hand, γ-tocopherol (γ-Toc) is reported to have a protective effect against prostate cancer. Therefore, we investigated whether the combination of γ-Toc and δ-T3 could strengthen the inhibitory effect of δ-T3 on prostate cancer cell growth. In this study the effect of combined δ-T3 (annatto T3 oil) and γ-Toc (Tmix, γ-Toc-rich oil) therapy was assessed against human androgen-dependent prostate cancer cells (LNCaP). We found that combined treatment of δ-T3 (10 μM) and γ-Toc (5 μM) resulted in reinforced anti-prostate cancer activity. Specifically, cell cycle phase distribution analysis revealed that in addition to G1 arrest caused by the treatment with δ-T3, the combination of δ-T3 with γ-Toc induced G2/M arrest. Enhanced induction of apoptosis by the combined treatment was also observed. These findings indicate that combination of δ-T3 and γ-Toc significantly inhibits prostate cancer cell growth due to the simultaneous cell cycle arrest in the G1 phase and G2/M phase.