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Mechanisms of combined action of different chemopreventive dietary compounds: a review

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Abstract

Consumption of fruits and vegetables has generally been associated with a decrease in cancer incidence and cardiovascular disease. Over the years, numerous bioactive compounds have been identified that contribute to these beneficial health effects. More recently, evidence is emerging that specific combinations of phytochemicals may be far more effective in protecting against cancer than isolated compounds. Combinatorial effects have been observed where any one of the single agents is inactive. Apart from interactions among dietary micronutrients, drug-phytochemical interactions have also been observed, indicating possibilities for improved cancer therapeutic strategies. Our understanding of the molecular mechanisms underlying such synergistic effects is still limited, but it appears that different combinations of complementary modes of actions are involved. In this review we discuss the molecular mechanisms that are likely to be involved in cancer chemoprevention and summarize the most important findings of those studies that report synergistic chemopreventive effects of dietary compounds.

Keywords: Chemoprevention, Phytochemicals, Synergistic effects, Polyphenols, Vegetables

Introduction

The relatively consistent epidemiological finding, that consumption of specific whole foods, such as fruits, vegetables and whole grains is strongly associated with reduced risk of cancer and other chronic diseases, has led to the hypothesis that specific phytochemicals may be responsible for the observed preventive action [1-5]. As a result, numerous bioactive compounds have been isolated and identified, and their potential health promoting effects have been evaluated extensively, both *in vitro* and *in vivo* [6-10]. One of the key issues in this field of research is, however, that purified phytochemicals do not necessarily exert the same beneficial health effect as when the compound source is in a food or even a complete specific diet. There is a growing body of evidence that the actions of phytochemicals administered as dietary supplements alone, do not explain the observed health benefits of diets rich in for instance fruits, vegetables and whole grains [11-15]. Although relatively high doses of single bioactive agents may show potent anticarcinogenic effects, the chemopreventive properties of interactions among various dietary ingredients that potentiate the activities of any single constituent may better explain the observed preventive effect of whole foods and diets in many epidemiological studies. In this paper, the evidence that bioactive compounds act synergistically is reviewed.

Mechanisms of anticarcinogenic effects

Carcinogenesis is an extremely complex multistep process, in which numerous molecular mechanisms play different crucial roles. As a result, cancer preventive dietary compounds may interfere with these processes at various levels. Table 1 summarizes mechanisms of action by

which phytochemicals can modulate cancer risk, both by blocking initiation and by suppressing the later stages involving promotion, progression, angiogenesis, invasion and metastasis [14,16].

Blocking mechanisms

Possible ways in which initiation of carcinogenesis can be blocked by phytochemicals include prevention of reactive oxygen species attack on DNA, altered metabolism of procarcinogens in favour of conjugation and excretion of reactive metabolites, inhibition of carcinogen uptake into cells and enhanced DNA repair. Many chemopreventive compounds possess antioxidant or free radical scavenging potential, which varies depending on the hydroxylation status of the benzene rings. Examples include quercetin (a flavonol in vegetables, apples and onions), xanthohumol (a chalone in hops and beer) and genistein (an isoflavone in soy). An early study by Duthie et al. reported that quercetin protected human lymphocytes from hydrogen peroxide-induced DNA damage [17]. Similar findings were reported by Wilms et al. [18,19], who also found that quercetin protected human lymphocyte DNA from *ex vivo* induced oxidative damage, an effect that was influenced by different genetic polymorphisms. In this study, volunteers consumed quercetin-rich blueberry/ apple juice for 4 weeks, which led to a significant increase in antioxidant capacity of plasma. Bulky adduct formation following treatment of lymphocytes with benzo[a]pyrene was however increased after the intervention.

Several types of bioactive compounds, including flavonoids, indoles, and bergamottin in citrus fruits, can interact with the aryl hydrocarbon receptor (AhR) as agonists or antagonists, depending on structure and cell context [20]. Such interactions influence the expression of drug metabolising enzymes such as cytochromes P450 [21]. They have also been shown to influence the multi-drug resistance phenotype acquired by many tumour cells.

Xanthohumol, present in small amounts in St. John's wort and hop extracts, possesses several useful properties to block carcinogenesis including modulation of enzymes involved in carcinogen metabolism and detoxification (inhibition of CYP1A, induction of quinone reductase activity), scavenging of ROS, including hydroxyl and peroxyl radicals, along with inhibition of superoxide anion radical formation and nitric oxide production [22].

Suppressing mechanisms

Mechanisms which result in suppression, or even better, elimination of tumour cells, include growth inhibition by induction of cell cycle arrest or apoptosis. A significant number of flavonoids, alone and in combination, have been shown to induce G_2/M arrest in SW480 and CaCo2 human colon carcinoma cells [23]. Tricin, a novel flavonol in rice bran, was shown to inhibit the growth of breast tumour cells, causing G_2/M arrest, but not apoptosis [24] . In a subsequent study by the same group [25], tricin decreased the number of intestinal adenomas in APCMin/+ mice by 33%, with inhibition of COX-1 and COX-2 activity. The latter led to a 34% reduction of PGE2 levels in

small intestinal mucosa and blood. Xanthohumol was also found to inhibit COX-1 and COX-2 activities, and to be anti-estrogenic [22]. The inhibitory effect of other flavonoids on COX-2 expression and activity has been reviewed by O'Leary et al. [26]. During later stages of carcinogenesis additional useful mechanisms include inhibition of angiogenesis, invasion and metastasis.

A range of tumour suppressing activities is shown for quercetin, a compound that has been intensively studies as a model flavonoid (Table 2). Resveratrol, genistein and epigallocatechin gallate (EGCG) (reviewed in [27]) have a number of effects in common with those detailed for quercetin, including inhibition of signalling through the EGFR family, NF-κB, and pAkt, induction of cell cycle arrest involving a decrease in cyclin D1 and phosphorylation of Rb, accompanied by upregulation of p21 and p27, and induction of apoptosis involving release of cytochrome c from mitochondria, activation of caspases 3 and 9 and downregulation of Bcl family members. However, depending on cell type and experimental conditions, flavonoids can both up- and down-regulate key molecules, including JNK, AP-1, p21, p27, cdc2, cyclin D1, p53, and PI3K.

One recent report by Fenton and Hord [39] has suggested a novel chemopreventive mechanism for flavonoids. In normal colon, epithelial cells migrate to the apex of the crypt, a process involving the APC gene, which is often mutated in colon cancer. These authors reported that apigenin, epicatechin, naringin and hesperidin induced a greater migratory response in APCMin/+ cells compared to those expressing wild type APC. Such flavonoid-induced migration was dependent on matrix metalloproteinase activity.

During the carcinogenic process, both hypermethylation of the promoter regions of tumour suppressor genes and hypomethylation of oncogenes can occur, resulting in under- or over-expression. Both EGCG [40] and genistein [41] have been shown to reactivate a number of key genes, such as the cell cycle inhibitor p16 and the retinoic acid receptor (RARß), in several different cancer cell types. The mechanism proposed was through inhibition of DNA methyltransferase, which, in the case of EGCG, involved direct interaction with the enzyme.

Evaluation of synergistically acting phytochemicals

In natural foods, combinations of phytochemicals are likely to influence cancer risk by affecting overlapping and complementary mechanisms. On the other hand, isolated pure compounds may lose their biological activity or may not behave in the same way as in the complex matrix of the original food item. This is illustrated, for instance, by the effects of increased intake of carotenoids and vitamin C in diets high in fruits, green and yellow vegetables, which are generally associated with cancer preventive effects. The effect of increased intake of ß-carotene or vitamin C as supplements is, however, questionable. Some studies show no reduced cancer incidence as a result of vitamin C [12] or ß-carotene supplementation [13], whereas even an increased lung

cancer incidence has been reported in smokers receiving supplemental ß-carotene [11,42]. Therefore, in addition to the characterisation of chemopreventive effects of individual compounds, evaluation of synergistically acting phytochemicals is of particular interest.

Synergistic effects of combinations of various polyphenols

A number of studies report enhanced chemopreventive effects of mixtures of polyphenols from green tea or other dietary sources. In Table 3, a selection of relevant studies is presented that describe such synergistic effects. Suganuma et al. [43] reported that the incorporation of tritium labelled EGCG into human lung cancer cells was enhanced by epicatechin (EC), another green tea polyphenol, but one which lacks a galloyl moiety. Epicatechin enhanced EGCG-induced apoptosis, growth inhibition of PC-9 lung tumour cells, and the inhibition of tumour necrosis factor-α release. These effects when induced by other tea polyphenols with a galloyl moiety, were also enhanced in a dose-dependent way by EC. The results of this study indicate that as a result of synergistic effects between green tea polyphenols, whole tea is a more efficient mixture for cancer prevention than supplementation with EGCG alone.

Synergistic effects of green tea catechins on cell growth and induction of apoptosis were also found in gastric carcinoma cells [44]. The results indicated that various gastric cell lines differed in their susceptibility to EGCG treatment. EC had almost no effect on cell growth or induction of apoptosis, but a significant synergistic effect on the induction of apoptosis was observed when EC was combined with other catechins. After treatment, the activity levels of caspases-3, -8 and -9 were elevated, indicating that these caspases are involved in catechin-induced apoptosis. Interestingly, catalase blocked the synergistic effect of EC and EGCG, suggesting that the production of hydrogen peroxide and reactive oxidative species are involved in the mechanism of synergy [44].

As cytochrome P450 (CYP) enzymes are responsible for the metabolism of many environmental carcinogens, modulation of their expression and activity by phytochemicals is a potential mechanism by which cancer risk may be influenced. Some of the cytochrome P450 genes are expressed constitutively, whereas others are inducible by xenobiotic compounds or phytochemicals. Enzyme induction usually enhances detoxification, but under some circumstances substrates may be activated to mutagens, carcinogens or cytotoxic substances [45]. Induction of the CYP1A enzymes by PAH and dioxins like TCDD, occurs at the level of transcription and is mediated by the cytosolic aryl hydrocarbon receptor (AhR) [45]. Williams et al. [46] demonstrated that complex green tea extracts exert mixed agonist/antagonist activity on the Ah-receptor, whereas EGCG acts as a strict AhR antagonist. Therefore, the authors conclude that modulation of human CYP1A1 expression by green tea extracts cannot be attributed to the

action of a single tea catechin, but rather is due to the effects of the complex mixture. Co-treatment of human hepatocytes with TCDD and different mixtures of tea catechins synergistically inhibited TCDD-induced CYP1A promoter-driven luciferase reporter activity (in HepG2 cells) and CYP1A1 expression (in HepG2 and primary human hepatocytes). The optimal synergy was found for a combination of the four major tea catechins, EC, EGCG, epigallocatechin (EGC), and epicatechin gallate (ECG), and was not improved by further addition of other compounds [47].

Chemopreventive synergism was also observed between EGCG and curcumin, a major phenolic antioxidant and anti-inflammatory agent in the spice *Curcuma longa* [48]. In malignant and premalignant human oral epithelial cells, the combination of both agents showed synergistic interactions in growth inhibition and increased sigmoidicity (steepness) of the dose-response curves. Calculated dose reduction indices (DRI) indicated that the combination allowed approximately a 4 – 8 fold dose reduction for ECGC and 2 – 3 fold for curcumin. On the other hand, antagonistic effects of this combination have been observed at the level of involucrin gene expression, involved in normal keratinocyte differentiation [49]. The same authors argue, however, that combined treatment with EGCG and curcumin may result in more efficient cancer chemoprevention than treatment with each agent alone; despite the fact that these compounds have opposing action on cell differentiation, they may still be effective when used together because of the shared property of growth suppression [50].

Synergistic effects of polyphenols with other phytochemicals

In two different studies, Zhou et al. investigated potential synergistic effects of the combination tea bioactive components and soy phytochemicals on androgen-sensitive human prostate tumours and estrogen-dependent human breast carcinoma in mice models [51,52]. In these studies, a soy phytochemical concentrate (SPC) and green and black tea infusions were used (Table 2). Multiple studies demonstrated that bioactive compounds in tea (particularly EGCG [53]) and soy (the soy isoflavone genistein as well as SPC) inhibit prostate cancer progression and tumour metastasis *in vivo* [54]. The combination of SPC and tea synergistically inhibited tumourigenicity, final tumour weight and metastasis to lymph nodes *in vivo*. The synergistic inhibition by the green tea and SPC combination on prostate tumour progression and metastasis was associated with effective reduction of serum levels of both testosterone and dihydrotestosterone, a biologically more active metabolite of testosterone and prerequisite for the development of benign prostatic hyperplasia and prostate cancer [51]. In an immune deficient mouse model, with implanted MCF-7 human breast cancer cells, SPC combined with green tea showed synergistic inhibition of tumour cell growth. This inhibition was associated with inhibited tumour angiogenesis and reduced estrogen receptor (ER)-alpha expression and serum levels of insulin-like growth factor (IGF)-I, both crucial factors in breast cancer development. Modulation of

these two different mechanisms of action may explain the synergistic effects of the combined phytochemicals [52].

A tumour specific growth protein with NADH oxidase activity (tNOX) has emerged as a potential target of the anticancer action of plant polyphenols and flavonoids [55]. NOX proteins are located at the cell surface and responsible for the increases in cell size following cell division [56]. Cells in which NOX activity is blocked, for instance by phytochemicals, are unable to enlarge, cease to divide and eventually undergo apoptosis. An exceptionally strong (10-fold) synergy was shown between grape polyphenols and tea catechins in the inhibition of tNOX [55]. The strongest synergistic activity was found with ethanol extracts of grape skins, whereas no activity was found for extracts of grape seeds, indicating that the effects were probably not caused by resveratrol, which is found in relatively high amounts in the seeds. These results suggest more efficient cancer prevention and therapy by using combinations of different phytochemicals.

Polyphenols and dietary antioxidant vitamins may also have synergistic inhibitory effects on lipid peroxidation and co-oxidation of dietary antioxidants. In simulated stomach fluid, it was demonstrated that phytochemicals can prevent the build-up of oxidized lipid products (lipid hydroperoxides and malondialdehyde) and destruction of vitamin E and ß-carotene (and vitamin C to a lesser extent) [57]. In the gastric fluid, vitamin C could enhance the activity of polyphenols through a synergistic antioxidant effect. The authors suggest that the antioxidant network in the stomach could thereby decrease the levels of hydroperoxides and other cytotoxic compounds and, in parallel, increase the vitamin antioxidants that reach the blood system. This would result in a synergistic increased systemic antioxidant effect that may also explain the French paradox (the fact that people in France suffer from relatively low incidence of coronary heart disease, despite their unhealthy dietary habits and high consumption of alcohol in the form of red wine) and the beneficial effect of Mediterranean and Japanese diets in which complex combinations of polyphenols and other antioxidants are found [57]. By studying the kinetics of the reaction of α-tocopherolyl radicals with green tea polyphenols using stopped-flow electron paramagnetic resonance, Zhou et al. [58] demonstrated unambiguously that several green tea polyphenols (EC, EGCG, EGC, ECG and gallic acid) can effectively reduce α-tocopheroxy radicals to regenerate α-tocopherol. Furthermore, these green tea polyphenols were able to trap the initiating radical ($ROO^{\bullet}$) as well as the propagating lipid peroxyl radicals ($LOO^{\bullet}$). It is particularly the elimination of the pro-oxidant effect of vitamin E (or the so-called tocopherol-mediated peroxidation), which may occur in absence of other oxidants [59], combined with the α-tocopherol regenerating reaction by coexisting antioxidants, that plays a crucial role in the enhancement of the antioxidant efficiency of vitamin E. These combined effects may also explain the synergistic antioxidant effects of tea

polyphenols and α-tocopherol in micelles and human low-density lipoprotein reported by the same group of researchers [60,61].

Studies on combination effects of isothiocynates and indoles, derived from cruciferous vegetables, have demonstrated that synergistic effects may depend on experimental conditions and concentrations. In human colon cancer cells, combinations of sulforaphane and 3,3'-diindolylmethane showed antagonistic effects on cell proliferation, cell cycle progression and apoptosis at physiologically low concentrations (2.5 μM), an effect that gradually turned into a synergistic interaction at the highest combined cytotoxic concentration of 40 μM [62]. These findings underline the need to elucidate mechanistic interactions in order to better predict beneficial health effects of bioactive food ingredients.
The combined effect of two other bioactive compounds from crucifers, indolo-3-carbinol and crambene, was studied in a rat model. The high dose experimental groups were protected against aflatoxin B1 induced toxicity, showing synergistic effects, whereas no effect was observed in the low dose groups [63].

Synergistic effects of whole foods / complex mixtures

Besides the synergistic effects of several individual compounds on biomarkers of cancer prevention, the synergistic effects of whole foods and other complex mixtures has been reported. In recent animal studies examining the effect of vegetable consumption on the modulation of gene expression, it was found that most of the genes that were differentially expressed before and after feeding with vegetables, represented changes in expression that could be interpreted as a cancer preventive effect [64,65]. Moreover, the results of these studies indicated that the effect of four individual vegetables on gene expression changes in the colon and lung in female C57Bl6 mice, differed from the effect of the mixture of the four vegetables. Furthermore, the mixture was able to modulate genes which were not significantly modulated by one of the specific vegetables present in the mixture. On the other hand, the individual vegetables were able to modulate genes which were not significantly modulated by the mixture, indicating that combinations of different foods containing different complex mixtures of phytochemicals can also have an antagonistic effect on gene expression.
Another example of the assessment of synergistic effects in complex mixtures is found in studies aiming to unravel the antioxidant capacity of red wines. In a large number of pinotage wines, the Trolox equivalent antioxidant capacity (TEAC) values and phenolic composition was determined [66]. The contribution of individually quantified phenolic compounds was calculated, and it was found that only between 11 and 24 % of the TEAC could be explained by the sum of the individual compounds. Different mixtures of 12 phenolic compounds in typical concentrations found in red wines, revealed 16 to 23 % of synergistic antioxidant activity. This implies that apart

from synergistic effects among phenolics, synergy between phenolic compounds and other wine constituents may also play a role. The authors exclude a potential role of sulphur dioxide in the regeneration of phenolic compounds from their phenoxyl radicals as it does not contribute to the total antioxidant potential at the concentrations normally present in red wines [67]. However, Jørgensen et al. [68] demonstrated for instance the regeneration of quercetin from its phenoxyl radical by (+)-catechin, thereby indicating the regeneration of phenoxy radicals by phenolic compounds as a possible mechanism for the synergistic effects observed for mixtures.

Other synergistic effects

In addition to the synergistic effects between several phytochemicals as discussed above, studying the combined effects of dietary factors and therapeutic compounds may be a promising approach to optimise pharmacological strategies for cancer prevention and therapy [16,69]. Administration of multiple agents may increase efficacy and potency of the chemopreventive action and, at the same time, reduce toxic side effects. Based on their synergistic activity *in vitro* or in animal studies, several drug combinations have been proposed for clinical development, such as retinoids in combination with SERMs (selective estrogen receptor modulators) like tamoxifen or raloxifene [70,71]. Also, the effects of EGCG on the induction of apoptosis in human lung cells *in vitro* reported by Suganuma et al. [43] were synergistically enhanced by cancer preventive agents, such as sulindac and tamoxifen. The same conclusion was drawn from animal studies, where co-treatment of rats with EGCG and sulindac resulted in significantly reduced aberrant crypt formation after treatment with azoxymethane [72]. The results of this study also revealed that EGCG and sulindac synergistically enhanced apoptosis. These data indeed confirm that combinations of phytochemicals and therapeutic agents result in even more effective cancer preventive therapies, and side effects, particularly of sulindac, may be reduced without loss of activity.

Quercetin and silymarin were found to inhibit MRP1/4/5-mediated drug transport from intact erythrocytes with high affinity, in a manner which suggested that they interact at the substrate-binding sites. Such interactions might influence bioavailability of anti-cancer drugs *in vivo* and could be considered for combination therapies [73].

In another recent study, the flavonols, quercetin and kaempferol, reduced P-glycoprotein expression and function in multi-drug resistant human cervical carcinoma KB-IV cells, while the isoflavones, genistein and daidzein, modulated intracellular drug levels by inhibiting function, without affecting expression [74].

Several other drug-phytochemical interactions have been studied, and almost all interactions between pharmaceutical drugs and dietary quercetin, genistein, curcumin and catechins showed increased therapeutic effects by blocking one or more targets of the signal transduction

pathways, by increasing the bioavailability of the other drug or, by stabilizing the other drug in the system [69,75-77].

Conclusions

A growing number of *in vitro* and *in vivo* studies indicate that combinations of dietary chemopreventive agents can sometimes result in significant activities at concentrations where any single agent is inactive. Many of these phytochemicals are reported to act synergistically, which may explain why some food items or diets may show cancer preventive effects which cannot be explained based on individual bioactive ingredients. Although our understanding of the molecular mechanism behind the observed combinatorial effects is still limited, it appears that many different combinations of complementary modes of action may be involved. The synergistic effects of dietary phytochemicals should be further explored for additional beneficial and reliable outcomes in the field of cancer prevention. Especially the development of new supplement regimens, cancer therapies and nutraceuticals may benefit from improved insight in the mechanisms behind synergistic effects of both natural and synthetic chemopreventive compounds.

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Table 1: Proposed mechanism by which dietary phytochemicals in general may prevent cancer

Antioxidant activity
Scavenging of free radicals and reduction of oxidative stress
Inhibition of cell proliferation
Induction of cell differentiation
Inhibition of oncogene expression
Induction of tumour suppressor gene expression
Induction of cell-cycle arrest
Induction of apoptosis
Inhibition of signal transduction pathways
Enzyme induction and enhancement of detoxification
Phase II enzymes
Glutathione peroxidase
Catalase
Superoxide dismutase
Enzyme inhibition
Phase I enzyme (blocking activation of carcinogens)
Cyclooxygenase-2
Inducible nitric oxide synthase
Xanthine oxidase
Enhancement of immune functions and surveillance
Inhibition of inflammation
Antiangiogenesis
Inhibition of cell adhesion and invasion
Inhibition of nitrosation and nitration
Prevention of DNA adduct formation or DNA intercalation
Regulation of steroid hormone metabolism
Regulation of estrogen metabolism
Antibacterial and antiviral effects

Modified from Liu et al., 2004 [14]

Table 2: Chemopreventive suppressing effects of quercetin

Tissue/cell type	Mechanism	Effect	Reference
HL60 human myeloid leukeamia cells	↑Bax; ↑phosphoBcl2; ↓Pgp	Apoptosis	28
Jurkat T cells	Inhibiting chymotrypsin-like activity of 20S and 26S proteosomes; ↑Bax; ↑IκBα	Apoptosis	29
Breast and prostate cancer cells	↓fatty acid synthase activity	Growth inhibition and apoptosis	30
Colonic aberrant crypt foci	↑Bax; ↓Bcl2; ↑cleavage of caspase 9	Suppression by 4-fold; apoptosis ↑3-fold	31
MiaPaCa pancreatic tumour cells	↓phosphoFAK	Decreased invasion	32
MCF7 breast tumour cells	↑PTEN; ↑p27; ↓Akt	Growth inhibition and apoptosis	33
LNCaP, PC3 prostate tumour cells	↓Sp1 interaction with AR; ↑c-jun	Inhibition of androgen receptor activity	34
HT29, SW480 colon cancer cells	↓ErbB2/3; ↓Bcl2; ↓phosphoAkt	Growth inhibition and apoptosis	35
SW480 colon cancer cells	↓β-catenin/Tcf transcriptional activity	↓c-myc	36
A549, H1299 human lung carcinoma cells	↑cyclin B1; ↑phospho cdc2; ↑survivin; ↑p53; ↑p21	Growth inhibition G_2/M arrest	37
PC3 prostate cancer cells	↓HSP70	Apoptosis	38

Table 3: Selection of studies on synergistic effects of mixtures of polyphenols and combinations with other types of phytochemicals

Combination of compounds	**Synergistic effect**	**Mechanisms involved**	**Reference**
Polyphenol mixtures			
EGCG and EC, sulindac or tamoxifen	Inhibition of lung cancer cell growth of human lung cancer cells	Enhanced cellular uptake of EGCG, enhanced apoptosis, reduced release of TNF-α	43
EGCG and EC	Inhibition of cell growth and induction of apoptosis in gastric carcinoma cells	Increased production of caspases-3,-8 and –9; Extracellular production of oxygen species	44
EGCG, EC, EGC and ECG	Modulation of CYP1A1 expression in human hepatocytes	Antagonism of TCDD-induced transcription of human CYP1A1, via interaction with the Ah-receptor	46,47
EGCG and Curcumin	Growth inhibition in (pre-) malignant human oral epithelial cells	Combined blocking of cell cycle at the G_1 and S/G_2M phase	48
EGCG and Curcumin	*Antagonistic* effects at the level of keratinocyte differentiation	Modulation of involucrin gene expression	49,50
Polyphenols and other phytochemicals			
Green/black tea and soy (SPC)*	Inhibition of prostate tumours, tumour weight and metastasis	Reduction of serum levels of testosterone and DHT	51
Green/black tea and soy (SPC)	Inhibition of breast tumour cell growth	Inhibition of tumour angiogenesis, reduced estrogen receptor-α protein levels and reduction of serum levels of IGF-I.	52
Green tea infusions and grape or grape skin extracts	Reduced tumour cell growth	Inhibition of tNOX, induction of apoptosis,	55
Polyphenols, vitamin E, A and ß-carotene	Reduced oxidative stress	Reduced formation of lipid hydroperoxides and malondialdehyde; reduced co-oxidation of vitamin E,C and ß-carotene	57
EGCG, EC, EGC and ECG or gallic acid and α-tocopherol	Reduced oxidative stress in micelles and human LDL	Reduction of α-tocopheryl radical, trapping of lipid peroxyl radicals and regeneration of vitamin E	58,60,61

* SPC: Soy Phytochemical Concentrate; DHT: dihydrotestosterone