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Green Tea and its polyphenols, targeting signalling pathways for chemoprevention

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ABSTRACT: Tea polyphenols can protect against the multi-stages of cancer initiation, promotion, and progression. Research findings and epidemiologic studies suggest that the polyphenolic compounds, (–)-epigallocatechin-3-gallate (EGCG), (–)-epicatechin-3-gallate (ECG); (–)-epigallocatechin (EGC) and (–)-epicatechin (EC) found primarily in Green Tea have chemo-preventive potential. Cell signalling cascades and their interacting factors have become important targets of chemoprevention and phenolic phytochemicals and plant extracts seem to be promising in this endeavour.

KEYWORDS: *Green tea; Polyphenols; Signalling pathways; Chemoprevention; Cancer*

Introduction

Tea produced from the leaves of the plant *Camellia sinensis* is the most widely consumed beverage in the world. The consumption of green tea is second to water intake. Structural and chemical comparison of Green tea with other tea gives better SAR relationship. Green tea is consumed daily among all, the best studied for health benefits, including chemo-preventive efficacy.[1] Based on manufacturing process, Green tea is categorized as Non-fermented Green tea produced by drying and steaming the fresh leaves to inactivate the polyphenol oxidase, semi-fermented Oolong tea produced when the fresh leaves are subjected to a partial fermentation stage before drying and fermented black and red (Pu-Erh) tea which undergo a post-harvest fermentation stage before drying and steaming.[2, 3] The major components of Green tea are the polyphenols, which are flavonoids and represent 36% dry weight of Green tea. Non-fermented Green tea contains more than 80% concentration of flavonoids, while fermented Black tea has only 20–30% of this phytoconstituents.[4] Research findings and epidemiologic studies suggest that the polyphenolic compounds, (–)-epigallocatechin-3-gallate (EGCG), (–)-epicatechin-3-gallate (ECG); (–)-epigallocatechin (EGC) and (–)-epicatechin (EC) found primarily in Green tea have chemo-preventive potential.[5, 6] Green tea contains 21 mg L^{-1} catechin (C), 98 mg L^{-1} EC, 90 mg L^{-1} ECG, 411 mg L^{-1} EGC, and 444 mg L^{-1} EGCG.[7] Another component of tea that may be important in cancer prevention is caffeine. EGCG and other tea polyphenols are well known for their antioxidant activities. Besides polyphenols, Green tea comprises proteins, enzymes, amino acids, carbohydrates, lipids (linoleic and α-linolenic acids), sterols, vitamins (B, C, E), pigments (chlorophyll and carotenoids), volatile compounds (aldehyde, mineral) and trace elements.[8] Chemo-preventive effect of Green tea is against stomach, lung, prostate, breast, skin, esophagus, duodenum, pancreas, liver and colon cancers induced by various chemical carcinogens.[9–11] Many other studies showed that Green tea has preventive effects against high blood pressure, high blood cholesterol concentrations, vascular endothelial dysfunctions, atherosclerosis and coronary heart disease.[12] Further, Phase I and Phase II clinical trials have also been conducted to explore the anticancer effects of Green tea in humans.[11]

Targeting Mechanisms of Green Tea

The prevalence and incidence of cancer is increasing world-wide, about 7.6 million people suffer from this insidious disease (around 13% of all deaths) in 2008. Deaths from cancer worldwide are projected to continue rising, with an estimated 13.1 million deaths in 2030. Despite the availability of many existing treatment approaches, researchers have yet to find a curative strategy for this insidious disease. Entertainingly, many epidemiological and experimental studies have demonstrated the protective effects of dietary factors towards human cancers. The plant-derived natural products polyphenolic and flavonoids are an invaluable treasure from nature, found to block various stages of carcinogenesis including cancer initiation, promotion and progression.[13] This observation bolstered chemoprevention as a valuable approach to arrest or delay the multistage process of

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carcinogenesis prior to the development of malignancy.[14] Epidemiologic research findings indicate that tea polyphenols can protect against the multi-stages of cancer initiation, promotion, and progression, but its mechanism of action is still being intensively investigated. Number of studies suggests that the gallate structure of the flavins is important for growth inhibition of tumour cell lines.[15] Further tea-associated polyphenolic compounds exhibit potent antioxidant activity, which has been suggested as important factor in alleviating cancer-associated oxidative stress. Tea polyphenols react with reactive oxygen species (ROS) (superoxide radical, singlet oxygen, hydroxyl or peroxyl radical, peroxynitrite) and prevent auto-oxidation.[16] Due to the ability of acting as good donors for hydrogen-bonding, an accurate prediction about tea polyphenol's solubility and permeability remains, at the moment, an elusive target for anticancer activity.[17–22] It has been found that green tea polyphenols reduce levels of insulin-like growth factor-1(IGF-1) in prostate tumour cells in rat model. The antioxidant property of chitosan green tea polyphenols complex induces transglutaminase activation in wound healing.[23] Chemo-preventive effect of tea based on several signalling mechanisms, target to specific cell-signalling pathways responsible for regulating cellular proliferation or apoptosis.[6] These pathways include signal transduction pathways leading to activator protein-1 (AP-1) and/or nuclear factor kappa B (NF-κB).[6] Catechins present anti-inflammatory activity through the inhibition of transcriptional factor NF-κB-mediated production of cytokines and adhesion molecules.[24] The mechanisms of this inhibition may be due to the blockade of the mitogenic and differentiating signals through modulating EGFR function, MAPK cascades, NF-kappa B activation as well as c-myc, c-jun and c-fos expression.[25]

Bioavailability of Green Tea

Pharmacokinetic parameters of tea catechins have been studied in detail in humans and rodents.[26–27] Various processes such as gastrointestinal degradation/metabolism, poor membrane permeability, and transporter-mediated intestinal secretion/efflux may contribute to poor bioavailability of tea polyphenols. According to Zhang *et al.*,[28] non-gallate catechins are more susceptible to efflux as compared to gallate catechins. The possible mechanism of elimination of these green tea catechins is by transporter-mediated intestinal efflux.[28] The bioavailability of tea polyphenols with large molecular masses such as theaflavins (564–868 Da) is low, compared to EGCG (458 Da) , which further showed limited bioavailability as compared to smaller molecules, EGC and EC (306 and 290 Da, respectively). The plasma levels of EGCG, EGC and EC after intragastric administration of decaffeinated green tea is found to be 0.1%, 14% and 31%, respectively.[29] It also demonstrated that treatment of rats with green tea polyphenols in their drinking fluid results in increase of plasma levels of EGC and EC, which were higher than that of EGCG over a 14-day treatment period,[30] whereas the tissue levels of green tea in male Sprague-Dawley rats (300 g), which were given 0.6% green tea in their drinking water for 14 days, had highest concentration of EGCG in large intestine (487.8±121.5 ng g^{-1}), while the concentration of EGC was highest in bladder (810.4±229.4) that approximated to 1.1 μM EGCG and 2.6 μM EGC30. Tea polyphenols are rapidly methylated by S-adenosyl-methionine, catalyzed by the enzymes catechol-*O*-methyltransferase (COMT) inside the body. UDP-glucuronoryltransferase (UGT) and sulphotransferase (SULT) enzymes also catalyzed the tea polyphenols to form glucuronide and sulpahte conjugates of catechins.[31]

Moreover, fewer studies have been also conducted on the biological activities of the metabolites of green tea polyphenols. Under *in vitro* conditions, EGCG is biologically more active in cancer cells towards growth inhibition as compared to their metabolites. The polyphenolic structure of EGCG also makes them good hydrogen donors, allowing EGCG to bind tightly to proteins and nucleic acids. Recently, EGCG has been shown to bind strongly to Bcl-2 protein, laminin receptor, vimentin and proteasome, contributing to its anticancer activities.[32–33] These studies may provide important insights into the design of future strategies aiming towards the development of green tea extracts (GTE) or green tea polyphenols (GTP) as a better chemopreventive agent.

Signalling Cascade Involved in Cancer Treatment

The process by which information from an extracellular signal is transmitted from the plasma membrane into the cell and along an intracellular chain of signalling molecules to stimulate a cellular response is known as "signal transduction." A cell may respond to a stimulus by activating gene transcription through proteins known as transcription factors. A transcription factor is composed of one or more proteins that bind to specific DNA sequences in a gene. Gene transcription is the most common result of the protein binding to the DNA and is referred to as transcriptional activation.

Insight to signalling mechanisms involved in chemo-

prevention by induction of cell cycle arrest and apoptosis or inhibition of signal transduction pathways, mainly the mitogen-activated protein kinases (MAPK), protein kinases C (PKC), phosphoinositide 3-kinase (PI3K), glycogen synthase kinase (GSK), which leads to abnormal cyclooxygenase-2 (COX-2), activator protein-1 (AP-1), and nuclear factor kappa-light chain-enhancer of activated B cells (NF-κB). NF-κB and AP-1 provide mechanistic links between inflammation and cancer.

Thus, cell signalling cascades and their interacting factors have become important targets of chemoprevention and phenolic phytochemicals, and plant extracts seem to be promising in this endeavour.

Role of Green Tea Components in MAP Kinase Pathway

Green tea polyphenols and flavonoids have been shown to interact with MAP kinase pathways and mediate signalling by influencing activation and phosphorylation of these molecules. Depiction of cells to stress stimuli or mutagens, form complex that involves discrete phosphorylation cascades, resulting in activation of members of MAPK family. Three classes of MAPKs known are c-Jun N-terminal kinases/stress-activated protein kinases (JNKs/ SAPKs), p38 kinases and the extracellular signal-regulated protein kinases (ERKs).[34–37] JNKs/SAPKs and p38 kinases got activated by various forms of stress, including ultraviolet (UV) irradiation.[36] In contrast, ERKs got initiated by tumour promoters such as TPA and growth factors including EGF and platelet-derived growth factor (PDGF), which play a critical role in transmitting signals.[38,39] Number of studies reported the effect of green tea in cancer prevention, mediating their action on the signalling pathways. Dong *et al.*[40] showed that EGCG (5–20 AMol L^{-1}) in JB6 mouse epidermal cell line, inhibited the MAPK pathway. Pre-treatment of EGCG (10–40 AMol L^{-1}) before UV exposure was shown to inhibit UV-induced hydrogen peroxide production concomitant with the inhibition of phosphorylation of ERK1/2, JNK, and p38 proteins.[41] Recently, EGCG (10–20 Ag ml^{-1}) has been shown to inhibit MAPK pathway and activator protein-1 (AP-1) activity in human colon cancer cells.[42] Moreover, it has been reported that oral feeding of EGCG containing green tea polyphenols inhibits PI3K pathway in transgenic adenocarcinoma of the mouse prostate (TRAMP) model system.[43] Because the deregulation of the MAPK pathway is frequently seen in a variety of human cancers, modulation of MAPKs by EGCG may provide novel strategies for the prevention or treatment of cancer.

Role of Green Tea Components in AP-1 Transcription Factor

AP-1 transcription factor is a protein dimer, composed of homodimers or heterodimers of protein super-family specifically, *jun* and *fos* gene families,[44–47] and it regulates the transcription of various genes associated with cellular inflammation, proliferation and apoptosis.[47] High AP-1 activity is involved in the tumour promotion and progression of various types of cancers, such as lung, breast, and skin cancer.[40] In cell culture and animal models, AP-1 was shown to be involved in tumour progression and metastasis[48] and played a key role in preneoplastic-to-neoplastic transformation.[49–50] Tea acts as a key target for chemo-prevention by inhibiting AT-1 activity.[51] By comparing P^+ and P^- derivatives of the mouse epidermal JB6 cell line, the authors found that the transcriptional factor AP-1 plays a critical role in tumour promotion.[52–53] Significantly, when tumour promoter-induced AP-1 activity was blocked, neoplastic transformation was inhibited.[52] EGCG was shown to inhibit 12-*O*-tetradecanoylphorbol-13-acetate or epidermal growth factor-induced transformation of mouse epidermal cell line JB6, and the inhibitory activity was closely related to the inhibition of AP-140.

Green Tea Components Inhibit Growth Factor-Induced Signal Transduction

The epidermal growth factor receptor (EGFR) is a ligand-binded, plasma membrane glycoprotein. EGFR have two binding domain sites, among them one is extracellular ligand-binding domain that binds to single transmembrane region, and an intracellular domain that exhibits intrinsic tyrosine kinase activity. EGFR is over expressed in neoplastic or mutagenic cells, activating signal transduction pathways that promote cell proliferation and tumour progression.[54–55] Over expression of EGFR is associated with a number of cancerous diseases including breast cancer,[56] lung cancer, squamous cell carcinoma of the oral cavity,[57] bladder carcinoma and oesophageal cancer.[58] Green tea catechin exert inhibitory effect on EGFR and has been suggested by studies performed on RTK activity.[59] In this study, the authors suggested that EGCG inhibit EGFR on human epidermoid carcinoma A431 cells, which express high levels of epidermal growth factor (EGF) receptors. Moreover, green tea components exhibit more effective inhibitory activity against receptor-type protein tyrosine kinases such as EGFR, PDGF-R, and FGFR (IC50 value of 0.5–1 μg ml^{-1}) than towards non-receptor-type protein tyrosine

kinases, such as pp60v-src60. EGF-stimulated increase in phosphotyrosine levels in A431 cells also inhibited by green tea components specifically EGCG60. Additionally, EGCG and ECG blocked EGFR binding, resulting in the inhibition of EGF-R kinase activity. The output is that EGCG could block many signal transduction events initiate from the cell surface and seems that it might be a useful compound for blocking RTK signalling. However, the effects of EGCG on EGF-R remain unclear since other studies failed to reproduce earlier observations in A172 glioblastomas[61] and in vascular smooth muscle cells from rat aorta.[62]

Green Tea and Activation of NF-κB Receptor

NF-κB stress responsive transcription factor-induced signalling mechanism is well known for its importance in inflammatory pathway.[63] Activation of NF-κB by Tumour promoter cells function to intensify the transcription of a variety of genes including cytokines, growth factors and acute response proteins.[64–69] Activation of NF-κB in cell lines linked to MAP kinase signalling pathways, especially p38 kinase, and played a major role in cell cycle.[70] Recent results also point to its pivotal role in suppressing apoptosis in cancer cells.[71] The mechanism involved in activation of NF-κB is well understood. NF-κB in dormant form, found in the cytosol, is bound to an inhibitory protein called inhibitory kappa B (IκB). When enthused, IκB protein is phosphorylated, ubiquinated, released from NF-κB and subsequently degraded. Following separation from IκB, NF-κB is translocated into the nucleus, where it activates gene transcription by binding to its distinct DNA sequence found in specific genes. Activation of NF-κB is associated with initiation, propagation or acceleration of tumour genes[72] and in JB6 cells, tumour promoter-induced cell transformation was blocked by inhibition of NF-κB.[73] Although less is known about the potential effects of tea polyphenols and flavonoids on NF-κB activation, like AP-1, NF-κB may also be a potential chemo-preventive or therapeutic target for tea components. The mechanism was suggested to be related to a differential inhibition by EGCG of TNF-α- or LPS-mediated NF-κB activation in cancer compared to normal cells, with the cancer cells generally being more sensitive to EGCG.[74] Consistent with this proposal is the observation that topical application of green tea polyphenols to UVB-irradiated, SKH-1 hairless skin decreased phosphorylation and degradation of I-κ Band the subsequent activation of NF-κB.[75]

Effect of Green Tea in Cell Cycle Arrest and Apoptosis

Over-expressed and irregular growth of cells at cell cycle check-point promote cell cycles factors such as cyclin D1 and cyclin dependent kinases (CDKs), which are associated with tumorigenesis.[76–77] A protein family known as cyclins and cdks are critical components; involvement of protein takes place in cell growth at different phases of cell cycle. Several investigators have shown that EGCG can affect the expression of these cell cycle regulators and inhibit the cell cycle.[78] The authors suggested that EGCG can induce G_0/G_1 phase cell cycle arrest in several human cancerous cell lines, including breast, epidermoid, prostate, colon, skin, lung and head and neck squamous cell cancers.[74, 78, 79–83] Liang *et al.*[84] reported that EGCG treatment in MCF7 breast cancer cells give prevention in G_0/G_1 phase cell cycle arrest.[84] Expression of p21 and p27 by EGCG inhibited the activity of CDK2 and CDK4, and also caused hypophosphorylation of *Rb* gene.[85] In prostate cancer cells, EGCG increased the expression of p16, p18, p21, and p53, which are associated with negative regulation of cell cycle progression.[80, 85] EGCG also reduced the protein expression of cyclin D1, cyclin E, CDK2, CDK4, and CDK6, but not cyclin D2. Effect of green tea catechins, especially EGCG, found to be more sensitive in treatment of head and neck squamous cell carcinoma cells. The mechanism related to preventive effect of EGCG was not clear, but number of factors associated with cell cycle progression, which has direct and indirect affect in chemoprevention. Additionally, the effect of EGCG has been directly inhibited CDKs that is the primary factor.[84] The induction of various negative regulators of the cell cycle may be the consequence of this inhibition. The concentrations used in these studies were higher than those observed in blood and tissues following consumption of tea. It still remains a topic of discussion, whether the green tea catechins involved in the mechanisms of cell cycle arrest at different check-points.

Apoptosis or Programmed cell death is a biochemical event that occurs in multicellular organism, leading to a variety of morphological changes including cell shrinkage, nuclear fragmentation, chromatin condensation, and chromosomal DNA fragmentation. Several mechanisms involved in anti-apoptotic properties of cell by administering green tea catechins, as EGCG has been shown to inhibit the expression of anti-apoptotic proteins Bcl-2 and Bcl-XL, while increasing the expression of Bax and Bak pro-apoptotic proteins.[86] It has been reported that EGCG effectively inhibited cellular proliferation and

induced apoptosis in sarcoma cells. Expression levels of Bcl-2 were significantly decreased, and the levels of Bax were significantly increased.[87] Moreover, EGCG induced apoptosis of human laryngeal epidermoid carcinoma through a caspase-independent, p53-mediated pathway.[87] EGCG induced cell cycle arrest and apoptosis in tumour cells, which were correlated with altered expression of Bcl-2 family proteins, including increased expression of proapoptotic Bax and decreased expression of prosurvival Bcl2, Bcl-XL, and Mcl-1 proteins.[88]

Green Tea Exerts Antioxidant Effect in Chemoprevention

Green tea is a dietary source rich of antioxidant nutrients due to presence of several composition like polyphenols, flavonoids, carotenoids, tocopherols, ascorbic acid (vitamin C), minerals such as Cr, Mn, Se, Fe, Cu or Zn, and certain phytochemical compounds exert antioxidant effect. Polyphenols rich in green tea as catechins and gallic acid specifically exert antioxidant potential. Green tea polyphenols with antioxidant activity scavenge free radicals, reactive oxygen species, nitrogen species and chelating agents with redox active transition metals ions. Additionally, green tea exerts antioxidant activity directly by chelating metal ions such as iron and copper to inhibit fenton reaction and haber-weiss reactions.[89-91] and indirectly by inhibiting, redox-sensitive transcription factor; pro-oxidant enzymes such as nitric oxide synthase, cyclooxygenase, lipoxygenase and xanthine oxidase and by induction of antioxidant enzymes, such as glutathione-S-transferases and superoxide dismutases.

Agents that induce cancer may be chemically or radically through ionizing radiation, ultraviolet radiation, smoking, ozone depletion, oxides formation from polluted air, all induces DNA damage.[92] The damage produced in cells is due to oxidative stress through generation of free radicals and reactive oxygen species. These species play a major role in carcinogenesis by damaging DNA, altering gene expression, affecting cell growth and differentiation.[93-94] Recent studies reported that green tea polyphenols exert potential effect against the cancerous cell in animal models. Likewise, the authors suggested that skin cancer induced by solar ultraviolet radiation was prevented by topically and oral application of catechins in mice.[95-96] In rats, carcinoma cells are effectively prevented by inhibiting the 12-*O*-tetradecanoyl phorbol 13-acetate induced NO generation involved in tissue damage and inflammation.[97] Moreover, *in vitro* studies showed that catechins, specifically EGCG, ameliorate the free radical and reactive species damage sustained by DNA by reduction in both prompt DNA-single strand breakage and in residual damage to DNA base.[98-99] Interestingly, green tea catechins have an ability to perform major task at different cellular levels. Likewise, at intracellular level, these catechins inhibited 12-*O*-tetradecanoylphorbol-13-acetate induced hydrogen peroxide formation in mouse epidermis[100] and 4-(Methylnitrosamino)-1-(3-pyridyl)-1-butanone (NNK)-induced 8-hydroxydeoxyguanosine formation in mouse lung.[101, 102] Tobacco-specific-nitrosamine lung carcinoma in mice was treated by catechins.[103] GTPs are also thought to be responsible for decreased susceptibility to UVB-induced tumourigenesis through scavenging the production of H_2O_2, otherwise lead to UVB-induced phosphorylation of proteins in the Ras pathway.[104] At extracellular levels, EGCG lowers the plasma concentration of phosphatidylcholine hydroperoxide (PCOOH). PCOOH is the predominant membrane lipid hydroperoxide in human plasma. The reactivity of EGCG with ROS and its chelating activity with Cu^{2+} would be important for the reduction of PCOOH.[105-106] In the plasma membrane compartment, EGCG also inhibits anion transport caused by deoxy–oxy cycling lipid peroxidation.[107] EGCG was also shown to inhibit the production of ONOO– (peroxynitrite anion) and lipid peroxidation by suppressing the induction of iNOS.[108-109] In summary, GTPs can scavenge ROS in all cellular compartments, in a variety of cells and in different body compartments before they have time to cause damage.

Green Tea Exerts Anti-Inflammatory Property in Chemoprevention

A mediator of inflammatory pathway is prostaglandin, one among all that has been synthesized from arachidonic acid in the presence of enzyme Cyclo-oxygenases (COXs). COXs family consists of three enzymes COX-1, COX-2 and COX-3. COX-1 is ubiquitously expressed in mammalian tissues and is involved in homeostasis. COX-3 functions are unknown, but it is found in the brain and spinal cord. More recent emphasis has been laid on COX-2 as its expression is found to be up regulated in inflammation and in cancer. COX-2 plays major role in modulating cell proliferation and apoptosis in tumour growth.[110] EGCG inhibited COX-2 expression in colon cancer cells by activating AMPK pathway.[111] mRNA and protein levels of COX-2 in androgen-dependent (LNCaP cells) and independent (PC-3 cells) levels in human prostate cancer were inhibited by EGCG.[112] Green tea polyphenols have inhibited the release of free-radicals and accomplished scavenging potential in generation of ROS by inhibition of COX-2 and inactivation

Table 1: Clinical Studies Indicating Tea and Its Phytoconstituents Against Cancer

S. No.	Phyto constituents	Treatment	Ref.
1	Green tea catechins, EGCG	Anti-inflammatory, Anti-oxidant activity	114
2	Nutraceuticals, Green tea extracts, EGCG, Resveratrol	Oral cancer of Squamous cell carcinoma type	115
3	EGCG	Breast cancer, Skin cancer, GIT tract cancer	116
4	EGCG	Stem like cancerous cell, tumour initiating cells	117
5	Black tea and green tea	Smokers and non-smokers oral cancers	118
6	Green tea catechins	Colorectal adenomas in Japanese patients	119
7	Green tea polyphenols	Prostate cancer	120
8	EGCG	Hep G2 cells **deheadtion** and migration	121
9	Green tea, black tea and EGCG	Prostate cancer	122
10	Green tea, black tea	GIT tract cancer, prostate pre-malignant lesions	123
11	EGCG	Suppression of ferritin in arsenic trioxide cytotoxicity	124
12	EGCG	ER-α expression in breast cancer	125
13	EGCG	Inhibit tumour invasion and angiogenesis in tumour growth	126
14	Green tea combined with citrus fruit	Inhibit COX enzymes in cancer cell exhibit anti-inflammatory property	125
15	EGCG	Prostate cancer	127
16	Botanical supplement with green tea	Prostate cancer	128
17	EGCG	Inhibit epidermal growth factor, PI3K, AKT, ERK and breast cancer	129
18	Green tea polyphenols and catechins	Esophageal squamous cell carcinoma	130
19	EGCG	Pancreatic cancer	131
20	Green tea	Stomach cancer	132
21	Green tea	Liver cancer	133
22	EGCG	In cell cycle growth arrest and apoptosis	134
23	Green tea catechin	Prostate cancer	135

of phosphorylated NF-kappa B and Akt.[113] Thus, EGCG exerts its anti-proliferative effects *via* COX-2 inhibition that exerts potent anti-inflammatory property.

Clinical Trials of Green Tea

Effects of tea polyphenols are widely studied for cancer chemoprevention in many different parts of the world. Several excellent reviews compiling results of such studies are available. Unfortunately, the lack of a firm conclusion still exists in spite of bundle of reviews and papers that have been written on green tea. One apparent reason is that aetiological factors for different types of cancers are not the same, and the populations studied are different. From the available data of epidemiologic studies and research findings in laboratory, clinical trials are now warranted to evaluate the usefulness of green tea and polyphenols present therein. Research suggests that green tea consumption has promising effects against various types of cancers without inducing major toxicities. Studies using green tea in the treatment of cancers have focused on breast, prostate and lung cancers as well as recently on chronic lymphocytic leukemia (CLL).

Conclusions

In whole, it is concluded that intake of green tea which provides sufficient catechins has beneficial effects against cancer in animals and humans. Green tea catechins exert a variety of beneficial metabolic effects by influencing the markers such as oxidative stress, inflammation as well as altering signalling cascades. Although Green tea catechins are beneficial in improving cancer, yet the positive effects of green tea catechins on humans need further attention. Studies should be carried out on catechin metabolites, which are reported to be biologically active, but their chemopreventive effects are unknown. In addition, structural aspects as well as bioavailability criteria

of Green tea catechins should be further explored which may lead to clinically relevant strategies to prevent and treat cancer.

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References

1. Mukhtar H & Ahmad N. 2000. Tea polyphenols: Prevention of cancer and optimizing health. *Am J Clin Nutr* 71(6 Suppl): 1698S–702S.
2. Zuo Y, Chen H, & Deng Y. 2002, Simultaneous determination of catechins, caffeine and gallic acids in Green, Oolong, Black and Pu-erh teas using HPLC with a photodiode array detector. *Talanta* 57(2): 307–316.
3. McKay DL & Blumberg JB. 2002. The role of tea in human health: An update. *J Am Coll Nutr* 21(1): 1–13.
4. Balentine DA, Wiseman SA, & Bouwens LC. 1997. The chemistry of tea flavonoids. *Crit Rev Food Sci Nutr* 37(8): 693–704.
5. Siddiqui IA, Adhami VM, Saleem M, & Mukhtar H, 2006. Beneficial effects of tea and its polyphenols against prostate cancer. *Mol Nutr Food Res* 50(2): 130–143.
6. Park AM & Dong Z. 2003. Signal transduction pathways: Targets for green and black tea polyphenols. *J Biochem Mol Biol* 36(1): 66–77.
7. Kuroda Y & Hara Y. 1999. Antimutagenic and anticarcinogenic activity of tea polyphenols. *Mutat Res*. 436(1): 69–97.
8. Cabrera C, Artacho R, & Giménez R. 2006. Beneficial effects of green tea–A review. *J Am Coll Nutr* 25(2): 79–99.
9. Chen D, Milacic V, Chen MS, Wan SB, Lam WH, Huo C, Landis-Piwowar KR, Cui QC, Wali A, Chan TH, & Dou QP. 2008. Tea polyphenols, their biological effects and potential molecular targets. *Histol Histopathol* 23(4): 487–496.
10. Thangapazham RL, Singh AK, Sharma A, Warren J, Gaddipati JP, & Maheshwari RK. 2007. Green tea polyphenols and its constituent epigallocatechin gallate inhibits proliferation of human breast cancer cells *in vitro* and *in vivo*. *Cancer Lett* 245(1–2): 232–241.
11. Hou Z, Lambert JD, Chin KV, & Yang CS. 2004. Effects of tea polyphenols on signal transduction pathways related to cancer chemoprevention. *Mutat Res* 555(1–2): 3–19.
12. Stangl V, Dreger H, Stangl K, & Lorenz M. 2007. Molecular targets of tea polyphenols in the cardiovascular system. *Cardiovasc Res* 73(2): 348–358.
13. Riboli E & Norat T. 2003. Epidemiologic evidence of the protective effect of fruit and vegetables on cancer risk. *Am J Clin Nutr* 78(3 Suppl): 559S–569S.
14. Ross JA & Kasum CM. 2002. Dietary flavonoids: Bioavailability, metabolic effects, and safety. *Ann Rev Nutr* 22: 19–34.
15. Yang GY, Liao J, Li C, Chung J, Yurkow EJ, Ho CT, & Yang CS. 2000. Effect of black and green tea polyphenols on c-jun phosphorylation and H(2)O(2) production in transformed and non-transformed human bronchial cell lines: Possible mechanisms of cell growth inhibition and apoptosis induction. *Carcinogenesis* 21(11): 2035–2039.
16. Sang S, Tian S, Meng X, Stark RE, Rosen RT, Yang CS, & Ho CT. 2002. Theadibenzotropolone A, a new type pigment from enzymatic oxidation of (–)-epicatechin and (–)-epigallocatechin gallate and characterized from black tea using LC/MS/MS. *Tetrahedron Lett* 43(40): 7129–7133.
17. Katiyar SK, Afaq F, Azizuddin K, & Mukhtar H. 2001a. Inhibition of UVB-induced oxidative stress-mediated phosphorylation of mitogen-activated protein kinase signalling pathways in cultured human epidermal keratinocytes by green tea polyphenol (–)-epigallocatechin-3-gallate. *Toxicol Appl Pharmacol* 176: 110–117.
18. Katiyar SK, Bergamo BM, Vyalil PK, & Elmets CA. 2001b. Green tea polyphenols: DNA photodamage and photoimmunology. *J Photochem Photobiol B* 65(2–3): 109–114.
19. Katiyar SK & Elmets CA. 2001. Green tea polyphenolic antioxidants and skin photoprotection (Review). *Int J Oncol* 18(6): 1307–1313.
20. Owuor ED & Kong AN. 2002. Antioxidants and oxidants regulated signal transduction pathways. *Biochem Pharmacol* 64(5–6): 765–770.

21. Zhang W, Hashimoto K, Yu GY, & Sakagami H. 2002. Decline of superoxide dismutase activity during antioxidant-induced apoptosis in HL-60 cells. *Anticancer Res* 22(1A): 219–224.

22. Rozoy E, Araya-Farias M, Simard S, Kitts D, Lessard J, & Bazinet L. 2013. Redox properties of catechins and enriched green tea extracts effectively preserve L-5-methyltetrahydrofolate: Assessment using cyclic voltammetry analysis. *Food Chem* 138(2–3): 1982–1991.

23. Qin Y, Guo XW, Li L, Wang HW, & Kim W. 2013. The antioxidant property of chitosan green tea polyphenols complex induces transglutaminase activation in wound healing. *J Med Food* 16(6): 487–498.

24. Bhardwaj P & Khanna D. 2013. Green tea catechins: Defensive role in cardiovascular disorders. *Chin J Nat Med* 11(4): 345–353. doi: 10.1016/S1875-5364(13)60051-5.

25. Lin JK. 2002. Cancer chemoprevention by tea polyphenols through modulating signal transduction pathways. *Arch Pharm Res* 25(5): 561–571.

26. Chow HH, Cai Y, Hakim IA, Crowell JA, Shahi F, Brooks CA, Dorr RT, Hara Y, & Alberts DS. 2003. Pharmacokinetics and safety of green tea polyphenols after multiple-dose administration of epigallocatechin gallate and polyphenon E in healthy individuals. *Clin Cancer Res* 9(9): 3312–3319.

27. Chow HH, Hakim IA, Vining DR, Crowell JA, Ranger-Moore J, Chew WM, Celaya CA, Rodney SR, Hara Y, & Alberts DS. 2005. Effects of dosing condition on the oral bioavailability of green tea catechins after single-dose administration of Polyphenon E in healthy individuals. *Clin Cancer Res* 11(12): 4627–4633.

28. Zhang L, Chow MS, & Zuo Z. 2006. Effect of the co-occurring components from green tea on the intestinal absorption and disposition of green tea polyphenols in CaCO-2 monolayer model. *J Pharm Pharmacol* 58(1): 37–44.

29. Okushio K, Suzuki M, Matsumoto N, Nanjo F, & Hara Y. 1999. Identification of (–)-epicatechin metabolites and their metabolic fate in the rat. *Drug Metab Dispos* 27(2): 309–316.

30. Kim S, Lee MJ, Hong J, Li C, Smith TJ, Yang GY, Seril DN, & Yang CS. 2000. Plasma and tissue levels of tea catechins in rats and mice during chronic consumption of green tea polyphenols. *Nutr Cancer* 37(1): 41–48.

31. Yang CS, Sang S, Lambert JD, & Lee MJ. 2008. Bioavailability issues in studying the health effects of plant polyphenolic compounds. *Mol Nutr Food Res* 52(Suppl 1):S139–S151.

32. Chen D, Milacic V, Chen MS, Wan SB, Lam WH, Huo C, Landis-Piwowar KR, Cui QC, Wali A, Chan TH, & Dou QP. 2008. Tea polyphenols, their biological effects and potential molecular targets. *Histol Histopathol* 23(4): 487–496.

33. Yang CS, Lambert JD, Hou Z, Ju J, Lu G, & Hao X. 2006. Molecular targets for the cancer chemo preventive activity of tea polyphenols. *Mol Carcinog* 45:431–435.

34. Boulton TG, Nye SH, Robbins DJ, Ip NY, Radziejewska E, Morgenbesser SD, DePinho RA, Panayotatos N, Cobb MH, & Yancopoulos GD. 1991. ERKs: A family of protein–serine/threonine kinases that are activated and tyrosine phosphorylated in response to insulin and NGF. *Cell* 65(4): 663–675.

35. Davis RJ. 1994. MAPKs: New JNK expands the group. *Trends Biochem Sci* 19(11): 470–473.

36. Kallunki T, Su B, Tsigelny I, Sluss HK, Dérijard B, Moore G, Davis R, & Karin M. 1994. JNK2 contains a specificity-determining region responsible for efficient c-Jun binding and phosphorylation. *Genes Dev* 8(24): 2996–3007.

37. Kyriakis JM, Banerjee P, Nikolakaki E, Dai T, Rubie EA, Ahmad MF, Avruch J, & Woodgett JR. 1994. The stress-activated protein kinase subfamily of c-Jun kinases. *Nature* 369(6476): 156–160.

38. Cowley S, Paterson H, Kemp P, & Marshall CJ. 1994. Activation of MAP kinase kinase is necessary and sufficient for PC12 differentiation and for transformation of NIH 3T3 cells. *Cell* 77(6): 841–852.

39. Minden A, Lin A, McMahon M, Lange-Carter C, Dérijard B, Davis RJ, Johnson GL, & Karin M. 1994. Differential activation of ERK and JNK mitogen-activated protein kinases by Raf-1 and MEKK. *Science* 266(5191): 1719–1723.

40. Dong Z, Ma W, Huang C, & Yang CS. 1997. Inhibition of tumor promoter-induced activator protein 1 activation and cell transformation by tea polyphenols, (–)-epigallocatechin gallate, and theaflavins. *Cancer Res* 57(19): 4414–4419.

41. Afaq F, Adhami VM, Ahmad N, & Mukhtar H. 2003. Inhibition of ultraviolet B-mediated activation of nuclear factor kappaB in normal human epidermal keratinocytes by green tea Constituent (–)-epigallocatechin-3-gallate. *Oncogene* 22(7): 1035–1044.

42. Shimizu M, Deguchi A, Lim JT, Moriwaki H, Kopelovich L, & Weinstein IB. 2005. (–)-Epigallocatechin gallate and polyphenon E inhibit growth and activation of the epidermal growth factor receptor and human epidermal growth factor receptor-2 signaling pathways in human colon cancer cells. *Clin Cancer Res* 11(7): 2735–2746.

43. Adhami VM, Siddiqui IA, Ahmad N, Gupta S, & Mukhtar H. 2004. Oral consumption of green tea polyphenols inhibits insulin-like growth factor-I-induced signaling in an autochthonous mouse model of prostate cancer. *Cancer Res* 64(23): 8715–8722.

44. Cohen DR & Curran T. 1988. Fra-1: A serum-inducible, cellular immediate-early gene that encodes a fos-related antigen. *Mol Cell Biol* 8(5): 2063–2069.

45. Halazonetis TD, Georgopoulos K, Greenberg ME, & Leder P. 1988. c-Jun dimerizes with itself and with c-Fos, forming complexes of different DNA binding affinities. *Cell* 55(5): 917–924.

46. Hirai SI, Ryseck RP, Mechta F, Bravo R, & Yaniv M. 1989. Characterization of junD: A new member of the jun proto-oncogene family. *EMBO J* 8(5): 1433–1439.

47. Angel P & Karin M. 1991. The role of Jun, Fos and the AP-1 complex in cell-proliferation and transformation. *Biochim Biophys Acta* 1072(2–3): 129–157.

48. Crawford HC & Matrisian LM. 1996. Mechanisms controlling the transcription of matrix metalloproteinase genes in normal and neoplastic cells. *Enzyme Protein* 49(1–3):20–37.

49. Bernstein LR & Colburn NH. 1989. AP1/jun function is differentially induced in promotion-sensitive and resistant JB6 cells. *Science* 244(4904): 566–569.

50. Barthelman M, Bair WB III, Stickland KK, Chen W, Timmermann BN, Valcic S, Dong Z, & Bowden GT. 1998. (–)-Epigallocatechin-3-gallate inhibition of ultraviolet B-induced AP-1 activity. *Carcinogenesis* 19(12): 2201–2204.

51. McCarty MF. 1988. Polyphenol-mediated inhibition of AP-1 transactivating activity may slow cancer growth by impeding angiogenesis and tumor invasiveness. *Med Hypotheses* 50(6): 511–514.

52. Dong Z, Watts R, Sun Y, Zhan S, & Colburn N. 1995. Progressive elevation of ap-1 activity during preneoplastic-to-neoplastic progression as modeled in mouse jb6 cell variants. *Int J Oncol* 7(2): 359–364.

53. Dong Z, Birrer MJ, Watts RG, Matrisian LM, & Colburn NH. 1994. Blocking of tumor promoter-induced AP-1 activity inhibits induced transformation in JB6 mouse epidermal cells. *Proc Natl Acad Sci USA* 91(2): 609–613.

54. Salomon DS, Brandt R, Ciardiello F, & Normanno N. 1995. Epidermal growth factor-related peptides and their receptors in human malignancies. *Crit Rev Oncol Hematol* 19(3): 183–232.

55. Mendelsohn J & Baselga J. 2000. The EGF receptor family as targets for cancer therapy. *Oncogene* 19(56): 6550–6665.

56. Nicholson S, Richard J, Sainsbury C, Halcrow P, Kelly P, Angus B, Wright C, Henry J, Farndon JR, & Harris AL. 1991. Epidermal growth factor receptor (EGFr); Results of a 6 year follow-up study in operable breast cancer with emphasis on the node negative subgroup. *Br J Cancer* 63(1): 146–150.

57. Atlas I, Mendelsohn J, Baselga J, Fair WR, Masui H, & Kumar R. 1992. Growth regulation of human renal carcinoma cells: Role of transforming growth factor alpha. *Cancer Res* 52(12): 3335–3339.

58. Mukaida H, Toi M, Hirai T, Yamashita Y, & Toge T. 1991. Clinical significance of the expression of epidermal growth factor and its receptor in esophageal cancer. *Cancer* 68(1):142–148.

59. Carpenter G. 1987. Receptors for epidermal growth factor and other polypeptide mitogens. *Ann Rev Biochem* 56: 881–914.

60. Liang YC, Lin-shiau SY, Chen CF, & Lin JK. 1997. Suppression of extracellular signals and cell proliferation through EGF receptor binding by (–)-epigallocatechin gallate in human A431 epidermoid carcinoma cells. *J Cell Biochem* 67(1): 55–65.

61. Sachinidis A, Seul C, Seewald S, Ahn H, Ko Y, & Vetter H. 2000. Green tea compounds inhibit tyrosine phosphorylation of PDGF beta-receptor and transformation of A172 human glioblastoma. *FEBS Lett* 471(1): 51–55.

62. Ahn HY, Hadizadeh KR, Seul C, Yun YP, Vetter H, & Sachinidis A. 1999. Epigallocathechin-3 gallate

selectively inhibits the PDGF-BB-induced intracellular signalling transduction pathway in vascular smooth muscle cells and inhibits transformation of sis-transfected NIH 3T3 fibroblasts and human glioblastoma cells (A172). *Mol Biol Cell* 10(4): 1093–1104.

63. Silverman N & Maniatis T. 2001. NF-kappaB signalling pathways in mammalian and insect innate immunity. *Gene Dev* 15(18): 2321–2342.
64. Baeuerle PA & Henkel T. 1994. Function and activation of NF-kappa B in the immune system. *Ann Rev Immunol* 12: 141–179.
65. Baeuerle PA & Baltimore D. 1996. NF-kappa B: Ten years after. *Cell* 87(1): 13–20.
66. Baldwin AS, Jr. 1996. The NF-kappa B and I kappa B proteins: New discoveries and insights. *Ann Rev Immunol* 14: 649–683.
67. Karin M. 1998. The NF-kappa B activation pathway: Its regulation and role in inflammation and cell survival. *Cancer J Sci Am* 4(Suppl 1):S92–S99.
68. Karin M & Ben-Neriah Y. 2000. Phosphorylation meets ubiquitination: The control of NF-[kappa]B activity. *Ann Rev Immunol* 18: 621–663.
69. Karin M & Delhase M. 2000. The I kappa B kinase (IKK) and NF-kappa B: Key elements of proinflammatory signalling. *Semin Immunol* 12(1): 85–98.
70. Schulze-Osthoff K, Ferrari D, Riehemann K, & Wesselborg S. 1997. Regulation of NF-kappa B activation by MAP kinase cascades. *Immunobiology* 198(1–3): 35–49.
71. Kucharczak J, Simmons MJ, Fan Y, & Gélinas C. 2003. To be, or not to be: NF-kappaB is the answer–Role of Rel/NF-kappaB in the regulation of apoptosis. *Oncogene* 22(56): 8961–8982.
72. Gilmore TD. 1997. Clinically relevant findings. *J Clin Invest* 100(12): 2935–2936.
73. Li JJ, Westergaard C, Ghosh P, & Colburn NH. 1997. Inhibitors of both nuclear factor-kappaB and activator protein-1 activation block the neoplastic transformation response. *Cancer Res* 57(16): 3569–3576.
74. Ahmad N, Cheng P, & Mukhtar H. 2000. Cell cycle dysregulation by green tea polyphenol epigallocatechin-3-gallate. *Biochem Biophys Res Commun* 275(2): 328–334.
75. Afaq F, Ahmad N, & Mukhtar H. 2003. Suppression of UVB-induced phosphorylation of mitogen-activated protein kinases and nuclear factor kappa B by green tea polyphenol in SKH-1 hairless mice. *Oncogene* 22(58): 9254–9264.
76. Semczuk A & Jakowicki JA. 2004. Alterations of pRb1-cyclin D1-cdk4/6-p16(INK4A) pathway in endometrial carcinogenesis. *Cancer Lett* 203(1): 1–12.
77. Diehl JA. 2002. Cycling to cancer with cyclin D1. *Cancer Biol Ther* 1(3): 226–231.
78. Ahmad N, Feyes DK, Nieminen AL, Agarwal R, & Mukhtar H. 1997. Green tea constituent epigallocatechin-3-gallate and induction of apoptosis and cell cycle arrest in human carcinoma cells. *J Natl Cancer Inst* 89(24): 1881–1886.
79. Lin JK, Liang YC, & Lin-Shiau SY. 1999. Cancer chemoprevention by tea polyphenols through mitotic signal transduction blockade. *Biochem Pharmacol* 58(6): 911–915.
80. Gupta S, Hussain T, & Mukhtar H. 2003. Molecular pathway for (–)-epigallocatechin-3-gallate-induced cell cycle arrest and apoptosis of human prostate carcinoma cells. *Arch Biochem Biophys* 410(1): 177–785.
81. Liberto M & Cobrinik D. 2000. Growth factor-dependent induction of p21(CIP1) by the green tea polyphenol, epigallocatechin gallate. *Cancer Lett* 154(2): 151–161.
82. Masuda M, Suzui M, & Weinstein IB. 2001. Effects of epigallocatechin-3-gallate on growth, epidermal growth factor receptor signaling pathways, gene expression, and chemosensitivity in human head and neck squamous cell carcinoma cell lines. *Clin Cancer Res* 7(12): 4220–4229.
83. Khan N, Afaq F, Saleem M, Ahmad N, & Mukhtar H. 2006. Targeting multiple signalling pathways by green tea polyphenol (–)-epigallocatechin-3-gallate. *Cancer Res* 66(5): 2500–2555.
84. Liang YC, Lin-Shiau SY, Chen CF, & Lin JK. 1999. Inhibition of cyclin-dependent kinases 2 and 4 activities as well as induction of Cdk inhibitors p21 and p27 during growth arrest of human breast carcinoma cells by (–)-epigallocatechin-3-gallate. *J Cell Biochem* 75(1): 1–12.
85. Gupta S, Ahmad N, Nieminen AL, & Mukhtar H. 2000. Growth inhibition, cell-cycle dysregulation, and induction of apoptosis by green tea constituent (–)-epigallocatechin-3-gallate in androgen-sensitive

and androgen-insensitive human prostate carcinoma cells. *Toxicol Appl Pharmacol* 164: 82–90.

86. Qin J, Xie LP, Zheng XY, Wang YB, Bai Y, Shen HF, Li LC, & Dahiya R. 2007. A component of green tea, (−)-epigallocatechin-3-gallate, promotes apoptosis in T24 human bladder cancer cells via modulation of the PI3K/Akt pathway and Bcl-2 family proteins. *Biochem Biophys Res Commun* 354(4): 852–857.

87. Tang GQ, Yan TQ, Guo W, Ren TT, Peng CL, Zhao H, Lu XC, Zhao FL, & Han X. 2010. (–)-Epigallocatechin-3-gallate induces apoptosis and suppresses proliferation by inhibiting the human Indian Hedgehog pathway in human chondrosarcoma cells. *J Cancer Res Clin Oncol* 136(8): 1179–1185.

88. Kang HG, Jenabi JM, Liu XF, Reynolds CP, Triche TJ, & Sorensen PH. 2010. Inhibition of the insulin-like growth factor I receptor by epigallocatechin gallate blocks proliferation and induces the death of Ewing tumor cells. *Mol Cancer Ther* 9(5): 1396–1407.

89. McKay DL & Blumberg JB. 2002. The role of tea in human health: An update. *J Am Coll Nutr* 21(1): 1–13.

90. Skrzydlewsja E, Augustyniak A, Ostrowska J, Luczaj W, & Tarasiuk E. 2002. Green tea protection against aging-induced oxidative stress. *Free Radic Biol Med* 33: 555.

91. Kim JH, Kang BH, & Jeong JM. 2003. Antioxidant antimutagenic and chemopreventive activities of a phyto-extract mixture derived from various vegetables, fruits and oriental herbs. *Food Sci Biotechnol* 12: 631–638.

92. Bondy ML, Wang LE, El-Zein R, de Andrade M, Selvan MS, Bruner JM, Levin VA, Alfred Yung WK, Adatto P, & Wei Q. 2001. Gamma-radiation sensitivity and risk of glioma. *J Natl Cancer Inst* 93(20): 1553–1557.

93. Cerutti PA. 1994. Oxy-radicals and cancer. *Lancet* 344(8926): 862–863.

94. Feig DI, Sowers LC, & Loeb LA. 1994. Reverse chemical mutagenesis: Identification of the mutagenic lesions resulting from reactive oxygen species-mediated damage to DNA. *Proc Natl Acad Sci USA* 91(14): 6609–6613.

95. Wei H, Zhang X, Zhao JF, Wang ZY, Bickers D, & Lebwohl M. 1999. Scavenging of hydrogen peroxide and inhibition of ultraviolet light-induced oxidative DNA damage by aqueous extracts from green and black teas. *Free Radic Biol Med* 26(11–12): 1427–1435.

96. Zhao JF, Zhang YJ, Jin XH, Athar M, Santella RM, Bickers DR, & Wang ZY. 1999. Green tea protects against psoralen plus ultraviolet A-induced photochemical damage to skin. *J Invest Dermatol* 113(6): 1070–1075.

97. Srivastava RC, Husain MM, Hasan SK, & Athar M. 2000. Green tea polyphenols and tannic acid act as potent inhibitors of phorbol ester-induced nitric oxide generation in rat hepatocytes independent of their antioxidant properties. *Cancer Lett* 153(1–2): 1–5.

98. Anderson RF, Fisher LJ, Hara Y, Harris T, Mak WB, Melton LD, & Packer JE. 2001. Green tea catechins partially protect DNA from (.)OH radical-induced strand breaks and base damage through fast chemical repair of DNA radicals. *Carcinogenesis* 22(8): 1189–1193.

99. Elattar TM & Virji AS. 2000. Effect of tea polyphenols on growth of oral squamous carcinoma cells *in vitro*. *Anticancer Res* 20(5B): 3459–3465.

100. Wang ZY, Hong JY, Huang MT, Reuhl KR, Conney AH, & Yang CS. 1992. Inhibition of N-nitrosodiethylamine- and 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone-induced tumorigenesis in A/J mice by green tea and black tea. *Cancer Res* 52(7): 1943–1947.

101. Xu Y, Ho CT, Amin SG, Han C, & Chung FL. 1992. Inhibition of tobacco-specific nitrosamine-induced lung tumorigenesis in A/J mice by green tea and its major polyphenol as antioxidants. *Cancer Res* 52(14): 3875–3879.

102. Wang ZY, Huang MT, Ferraro T, Wong CQ, Lou YR, Reuhl K, Iatropoulos M, Yang CS, & Conney AH. 1992b. Inhibitory effect of green tea in the drinking water on tumorigenesis by ultraviolet light and 12-*O*-tetradecanoylphorbol-13-acetate in the skin of SKH-1 mice. *Cancer Res* 52(5): 1162–1170.

103. Brown BG, Richter E, Tricker AR, Ayres PH, & Doolittle DJ. 2001. The effect of a 2-h exposure to cigarette smoke on the metabolic activation of the tobacco-specific nitrosamine 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone in A/J mice. *Chem Biol Interact* 138(2): 125–135.

104. Katiyar SK, Afaq F, Azizuddin K, & Mukhtar H. 2001. Inhibition of UVB-induced oxidative stress-mediated phosphorylation of mitogen-activated protein kinase signalling pathways in cultured human

epidermal keratinocytes by green tea polyphenol (–)-epigallocatechin-3-gallate. *Toxicol Appl Pharmacol* 176(2): 110–117.

105. Miyazawa T. 2000. Absorption, metabolism and antioxidative effects of tea catechin in humans. *Biofactors* 13(1–4):55–59.

106. Nakagawa K, Ninomiya M, Okubo T, Aoi N, Juneja LR, Kim M, Yamanaka K, & Miyazawa T. 1999. Tea catechin supplementation increases antioxidant capacity and prevents phospholipid hydroperoxidation in plasma of humans. *J Agric Food Chem* 47(10): 3967–3973.

107. Ohnishi ST, Ohnishi T, Ogunmola GB. 2001. Green tea extract and aged garlic extract inhibit anion transport and sickle cell dehydration *in vitro*. *Blood Cells Mol Dis* 27(1): 148–157.

108. Paquay JB, Haenen GR, Stender G, Wiseman SA, Tijburg LB, & Bast A. 2000. Protection against nitric oxide toxicity by tea. *J Agric Food Chem* 48(11): 5768–5772.

109. Chan MM, Fong D, Ho CT, & Huang HI. 1997. Inhibition of inducible nitric oxide synthase gene expression and enzyme activity by epigallocatechin gallate, a natural product from green tea. *Biochem Pharmacol* 54(12): 1281–1286.

110. Sobolewski C, Cerella C, Dicato M, Ghibelli L, & Diederich M. 2010. The role of cyclooxygenase-2 in cell proliferation and cell death in human malignancies. *Int J Cell Biol* 21: 51–58. doi: 10.1155/2010/215158.

111. Hwang JT, Ha J, Park IJ, Lee SK, Baik HW, Kim YM, & Park OJ. 2007. Apoptotic effect of EGCG in HT-29 colon cancer cells via AMPK signal pathway. *Cancer Lett* 247(1): 115–121.

112. Hussain T, Gupta S, Adhami VM, & Mukhtar H. 2005. Green tea constituent epigallocatechin-3-gallate selectively inhibits COX-2 without affecting COX-1 expression in human prostate carcinoma cells. *Int J Cancer* 113(4): 660–669.

113. Roy P, George J, Srivastava S, Tyagi S, & Shukla Y. 2011. Inhibitory effects of tea polyphenols by targeting cyclooxygenase-2 through regulation of nuclear factor kappa B, Akt and p53 in rat mammary tumors. *Invest New Drugs* 29(2): 225–231.

114. Lecumberri E, Dupertuis YM, Miralbell R, & Pichard C. 2013. Green tea polyphenol epigallocatechin-3-gallate (EGCG) as adjuvant in cancer therapy. *Clin Nutr* pii: S0261–5614(13)00086-1. doi: 10.1016/j.clnu.2013.03.008.

115. Zlotogorski A, Dayan A, Dayan D, Chaushu G, Salo T, & Vered M. 2013. Nutraceuticals as new treatment approaches for oral cancer: II. Green tea extracts and resveratrol. *Oral Oncol* 49(6): 502–506.

116. Donejko M, Niczyporuk M, Galicka E, & Przylipiak A. 2013. Anti-cancer properties epigallocatechin-gallate contained in green tea. *Postepy Hig Med Dosw* 67: 26–34.

117. Lin CH, Shen YA, Hung PH, Yu YB, & Chen YJ. 2012. Epigallocathechin gallate, polyphenol present in green tea, inhibits stem-like characteristics and epithelial-mesenchymal transition in nasopharyngeal cancer cell lines. *BMC Complement Altern Med* 12: 201.

118. Dash C, Chung FL, Rohan JA, Greenspan E, Christopher PD, Makambi K, Hara Y, Newkirk K, Davidson B, & Adams-Campbell LL. 2012. A six-month crossover chemoprevention clinical trial of tea in smokers and non-smokers: Methodological issues in a feasibility study. *BMC Complement Altern Med* 12: 96. doi: 10.1186/1472-6882-12-96.

119. Fujiki H, Imai K, Nakachi K, Shimizu M, Moriwaki H, & Suganuma M. 2012. Challenging the effectiveness of green tea in primary and tertiary cancer prevention. *J Cancer Res Clin Oncol* 138(8): 1259–1270.

120. Davalli P, Rizzi F, Caporali A, Pellacani D, Davoli S, Bettuzzi S, Brausi M, & D'Arca D. 2012. Anticancer activity of green tea polyphenols in prostate gland. *Oxid Med Cell Longev* 2012:984219. doi: 10.1155/2012/984219.

121. Li X, Feng H, Chen B, Ng SS, Chen WN, & Chan V. 2011. Epigallocatechin-3-gallate induced modulation of cell deadhesion and migration on thermosensitive poly (N-isopropylacrylamide). *J Biomed Mater Res A* 98(3): 450–460.

122. Henning SM, Wang P, & Heber D. 2011. Chemopreventive effects of tea in prostate cancer: Green tea versus black tea. *Mol Nutr Food Res* 55(6): 905–920.

123. Yuan JM, Sun C, & Butler LM. 2011. Tea and cancer prevention: Epidemiological studies. *Pharmacol Res* 64(2): 123–135.

124. Lee TC, Cheng IC, Shue JJ, & Wang TC. 2011. Cytotoxicity of arsenic trioxide is enhanced by (–)-epigallocatechin-3-gallate via suppression of ferritin

in cancer cells. *Toxicol Appl Pharmacol* 250(1): 69–77.

125. Li Y, Yuan YY, Meeran SM, & Tollefsbol TO. 2010. Synergistic epigenetic reactivation of estrogen receptor-α (ERα) by combined green tea polyphenol and histone deacetylase inhibitor in ERα-negative breast cancer cells. *Mol Cancer* 9: 274. doi: 10.1186/1476-4598-9-274.

126. Khan N & Mukhtar H. 2010. Cancer and metastasis: Prevention and treatment by green tea. *Cancer Metastasis Rev* 29(3): 435–445.

127. Johnson JJ, Bailey HH, & Mukhtar H. 2010. Green tea polyphenols for prostate cancer chemoprevention: A translational perspective. *Phytomedicine* 17(1): 3–13.

128. Evans S, Dizeyi N, Abrahamsson PA, & Persson J. 2009. The effect of a novel botanical agent TBS-101 on invasive prostate cancer in animal models. *Anticancer Res* 29(10): 3917–3924.

129. Puig T, Relat J, Marrero PF, Haro D, Brunet J, & Colomer R. 2008. Green tea catechin inhibits fatty acid synthase without stimulating carnitine palmitoyltransferase-1 or inducing weight loss in experimental animals. *Anticancer Res* 28(6A): 3671–3676.

130. Szumiło J. 2008. [Chemoprevention of esophageal squamous cell carcinoma--clinical trials]. *Pol Merkur Lekarski* 25(147): 280–283.

131. Basu A & Haldar S. 2009. Combinatorial effect of epigallocatechin-3-gallate and TRAIL on pancreatic cancer cell death. *Int J Oncol* 34(1): 281–286.

132. Jin X, Zheng RH, & Li YM. 2008. Green tea consumption and liver disease: A systematic review. *Liver Int* 28(7): 990–906.

133. Myung SK, Bae WK, Oh SM, Kim Y, Ju W, Sung J, Lee YJ, Ko JA, Song JI, & Choi HJ. 2009. Green tea consumption and risk of stomach cancer: A meta-analysis of epidemiologic studies. *Int J Cancer* 124(3): 670–677.

134. Morrissey C, Brown M, O'Sullivan J, Weathered N, Watson RW, & Tenniswood M. 2007. Epigallocatechin-3-gallate and bicalutamide cause growth arrest and apoptosis in NRP-152 and NRP-154 prostate epithelial cells. *Int J Urol* 14(6): 545–551.

135. Bettuzzi S, Rizzi F, & Belloni L. 2007. Clinical relevance of the inhibitory effect of green tea catechins (GtCs) on prostate cancer progression in combination with molecular profiling of catechin-resistant tumors: An integrated view. *Pol J Vet Sci* 10(1): 57–60.