Tea Beverage in Chemoprevention of Prostate Cancer: A Mini-Review

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Abstract: Because prostate cancer has a long latency period and is typically diagnosed in elderly men, it represents an ideal candidate disease for chemoprevention. Therefore, even a modest delay achieved through intervention could have a significant impact on the outcome of this disease. Epidemiological and laboratory studies have provided convincing evidence that diet, genetic factors, and lifestyle are major causes of prostate cancer. Although surgery, radiotherapy, and hormone therapy are the most widely accepted curative options for a selected group of patients suffering from prostate cancer, the side effects of these treatments are many. In recent years, many dietary agents have been being described that show a wide range of chemopreventive effects in cell culture and selected animal model systems of prostate carcinogenesis. One such agent is the beverage tea, which, next to water, is the most popularly consumed beverage in the world. The epidemiological studies and recent data, amassed from various laboratories around the world, provide evidence that tea polyphenols such as epigallocatechin-3-gallate, epigallocatechin, and epicatechin-3-gallate may have the potential to lower the risk of prostate cancer in the human population. Recently, it has been shown that green tea polyphenols, when given to TRAMP, a transgenic mouse model that mimics progressive forms of human prostate cancer, exert remarkable preventive effects against prostate cancer development. Chemoprevention of prostate cancer by tea polyphenols appears to occur through the modulation of various molecular targets. This article attempts to address the issue of the possible use of tea, especially green tea, for the chemoprevention of prostate cancer.

Introduction

Next to skin cancer, prostate cancer is the most common cancer diagnosed in men in North America, Europe, and some parts of Africa (1). In 15 yr, prostate cancer is predicted to be the most common cancer in men and thus has become a significant public health concern (2). Currently, almost 10% of men in the United States develop clinically detectable prostate cancer in their lifetime (3). Prostate cancer is generally detected in aged men and, because of increasing life expectancy and better diagnosis of the disease, its rate of incidence is expected to increase (4). It is estimated that 220,900 new prostate cancer cases will be diagnosed in the United States alone in the year 2003, and approximately 28,900 prostate cancer-related deaths are predicted for this year alone in the United States (5). Despite the substantial morbidity and mortality, the etiology of prostate cancer is poorly understood. The only established risk factors for prostate cancer are age, race, and a family history of prostate cancer. However, it is increasingly appreciated that environment and lifestyle, particularly dietary habits, also contribute substantially to the disease outcome (6,7).

Studies of the incidence and mortality of prostate cancer in various countries suggest that changeable environmental factors are important in its etiology. The rising incidence rates of prostate cancer in several countries that previously were considered to have low incidence rates appear to be coincident with the adoption of a Western lifestyle in those populations, implicating factors such as low levels of physical activity, high relative body weight, and high dietary fat intake (8 and references therein) in the pathophysiology of prostate cancer. Nutritional factors in defined populations, especially in those with high animal fat and high intake of dairy products, have been correlated with a greater risk of prostate cancer (9). In at least one case-control study with age-adjusted analysis, a positive association of prostate cancer risk with total energy intake as well as intake of total fat was established (10). These observations, combined with an improved understanding of the molecular biology of prostate cancer, provide numerous leads to explore testable prostate cancer prevention strategies. Many types of treatments, such as hormone therapy, radiation, and surgery, are proving useful in reducing the mortality associated with this slowly progressing disease. Despite these treatments, almost all tumors (especially malignant ones) continue to progress and become refractory to treatment options, and in most cases currently available approved treatments are beneficial only to the selected group of patients. Although these treatments provided limited success in reducing the mortality in prostate cancer patients, severe side effects have been reported. These include rectal complications, urinary incontinence, impotence, loss of libido, weight gain, gynecomastia, liver inflammation, and osteoporosis (11). The reasons for these clinical phenomena are poorly understood. Because prostate cancer is a complex process and involves different
molecular events, usually occurring simultaneously, blocking or inhibiting only one event is not sufficient to prevent or delay the onset of the disease. Therefore, it is necessary to intensify our efforts for a better understanding of prostate cancer and for the development of novel approaches for its prevention and treatment.

It is increasingly appreciated that chemoprevention, which involves the use of natural or synthetic agents to suppress, block, or reverse the process of carcinogenesis, could be an effective approach to reduce the incidence of prostate cancer. Among all cancer types, prostate cancer is an ideal candidate disease for chemoprevention because of its particularly long latency period. It is typically diagnosed in elderly men and, therefore, even a modest delay in the neoplastic development achieved through pharmacological, nutritional, or therapeutic intervention could result in substantial reduction in the incidence of clinically detectable disease. In addition, the long latency period of clinical manifestation of prostate cancer provides a suitable window of opportunity for intervention during the cancer development process through chemoprevention by dietary and beverage-derived agents. Many types of natural agents that act on various molecular targets simultaneously have been reported to inhibit or delay various stages of cancer (12). Recent epidemiological studies have found a correlation between populations with higher consumption of selenium, vitamin E, fruits, and tomatoes in lowering the risk of prostate cancer (13). Consistent with this notion, several single natural agents are under study for their assessment as chemopreventive agents against prostate cancer. Tea derived from *Camellia sinensis* was shown to have antimutagenic and cancer chemopreventive effects in animal tumor models (14,15). Almost 4 yr ago, we initiated a program to assess whether tea consumption could afford chemopreventive effects against prostate carcinogenesis (16). This review summarizes the laboratory, clinical, trial, and epidemiological observations for the use of tea or its constituent polyphenols for prostate cancer chemoprevention.

**Chemoprevention and Prostate Cancer**

Chemoprevention is the administration of agents to totally prevent and partially inhibit or significantly delay progression of cancer (17). For a variety of reasons the use of naturally occurring dietary agents over synthetic agents for cancer chemoprevention is preferred (17,18). One way of considering chemoprevention is preventive maintenance of the body by use of natural agents. Important to chemoprevention is the fact that carcinogenesis is a long process of cellular growth, division, and subsequent clonal expansion of initiated cells exemplified by steps known as initiation, promotion, and progression (17). One advantage of chemoprevention is that agents can be targeted against each stage of tumorigenesis. Inhibition or slowing of any stage of carcinogenesis can potentially prevent cancers from becoming clinically significant. Chemoprevention seems to be a promising approach against prostate cancer because therapy and surgery have not been fully effective and recurrence of the metastatic form of the disease is often responsible for the low survival rate in patients (19,20).

Prostate cancer is an ideal candidate disease for chemoprevention because of its high latency period and because it is commonly diagnosed in men over the age of 50. If chemoprevention delays the clinical course of prostate cancer even by 5 yr, incidence of and deaths from this disease would substantially decrease (21). Thus, even a modest delay in the progression of this type of cancer by chemopreventive agents could result in a substantial reduction of incidence of this disease and, more importantly, improve the quality of life of patients with the disease. Most of the experimental as well as epidemiological studies conducted in last 2 decades pertaining to chemoprevention of cancer have shown that the plant-derived products are the most acceptable and promising agents that could inhibit or delay various types of cancer (17). These plant-derived agents (phytochemicals) are generally non-nutritive compounds. Currently, the chemopreventive effects of more than 30 classes of these compounds have been described, many of which may have practical implications in reducing cancer incidence, at least in high-risk individuals (22). One of the important classes of phytochemicals that has caught the attention of researchers is a group of polyphenolic compounds that have been shown to inhibit or delay many types of cancers in experimental animals and epidemiological observations (23). Many fruits, vegetables, and beverages have been reported to be a rich source of polyphenolic compounds. The chemoprevention of prostate cancer by the intervention of plant-derived polyphenolic compounds seems to be an attractive possibility.

**Tea: A Brief Overview**

Tea brews were considered healthful beverages by many ancient cultures of the East and to this day tea continues to be a popular drink throughout the world (24). The per capita worldwide consumption is approximately 120 ml brewed tea per day. Many types of tea preparations, originating from the same plant source (*C. sinensis*) but having different processing methods, are consumed today. The three most popular tea types are black tea (78%), green tea (20%), and oolong tea (2%). Green tea production involves steaming fresh leaves at elevated temperatures followed by a series of drying and rolling steps so that the chemical composition essentially remains similar to that of the fresh leaves. Black tea production involves withering of plucked leaves followed by extended fermentation. Thus, depending upon the extent of fermentation, the chemical composition of most black teas is different. Oolong tea is made by solar withering of tea leaves followed by partial fermentation.

**Polyphenols of Tea**

Tea leaves are unique as they are a rich source of catechins, caffeine, and theanine (Table 1). These constituents
Table 1. Chemical Composition of Polyphenolic Constituents of Tea

<table>
<thead>
<tr>
<th>Constituents</th>
<th>Amount (mg/l)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Green Tea</td>
</tr>
<tr>
<td>Epigallocatechin</td>
<td>129.0</td>
</tr>
<tr>
<td>Gallatecin</td>
<td>1.0</td>
</tr>
<tr>
<td>Gallicatechin</td>
<td>42.0</td>
</tr>
<tr>
<td>Epicatechin</td>
<td>8.0</td>
</tr>
<tr>
<td>Epigallocatechin</td>
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</tr>
<tr>
<td>Gallatecin</td>
<td>207.0</td>
</tr>
<tr>
<td>Catechin</td>
<td>4.0</td>
</tr>
<tr>
<td>Catechin gallate</td>
<td>84.0</td>
</tr>
<tr>
<td>Caffeine</td>
<td>233.0</td>
</tr>
<tr>
<td>Theaflavins</td>
<td>0</td>
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<tr>
<td>Gallated theaflavins</td>
<td>0</td>
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<td>Thearubigens</td>
<td>0</td>
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</table>

Impart flavor and taste to tea beverages. The catechins are characteristic water-soluble polyphenolic compounds and account for 30–42% of the dry weight of the solids in brewed tea (25). The major green tea catechins are (-)-epicatechin-3-gallate (ECG), epigallocatechin (EGC), epigallocatechin-3-gallate (EGCG), and (-)-epicatechin (EC; Fig.1). Among these, EGCG is the most abundant polyphenol and has received by far the most attention. A brewed cup of green tea contains up to 300 mg of EGCG. Black tea, in addition to catechins, also contains thearubigins and theaflavins (26). In general, the caffeine content in green tea ranges from 3 to 6% and black tea contains 2–4% of dry weight of caffeine. Tea contains phenolic acids, mainly caffic, quinic, and gallic acids (27). Theanine is an amino acid found only in tea leaves, which imparts a pleasantly sweet taste to tea. Theanine is degraded to glutamic acid and has been shown to have relaxation effects in humans. Tea contains up to one-third of dry weight of catechins and other polyphenols such as quercetin, myricitin and kaempferol (28).

Biological Activities of Tea and Its Constituents

The most widely recognized biological properties of tea polyphenols are the antioxidant properties (29). Studies have shown that oral consumption of green tea or EGCG can lower serum cholesterol levels in rats and humans (30) and increase high-density lipoprotein cholesterol (31,32) and decrease blood glucose in rats (33). Based on oral and i.p. effects of EGCG on serum hormones and nutrients, long-term consumption of green tea is considered to influence the incidence of obesity, diabetes, and cardiovascular diseases (34). A recent review on these subjects is available (35).

A number of studies in laboratory animals in various target-organ bioassay protocols, conducted in many laboratories around the world, have provided convincing evidence that the polyphenolic compounds present in tea are capable of affording protection against cancer initiation and its subsequent development (36). Tea consumption in animal model systems has been demonstrated to inhibit lung, skin, esophagus, stomach, liver, duodenum, small intestine, and pancreatic tumor formation (23). Studies in culture systems have shown that both green tea extract and EGCG are capable of

Figure 1. Structures of polyphenolic constituents found in green and black tea. R denotes gallate.
inhibiting the growth of a variety of mouse and human cancer cell types without affecting normal cells. Initial work on this topic was reported by Ahmad et al., showing that green tea may protect against cancer by causing cell cycle arrest and inducing apoptosis in various cell lines such as HaCaT and L5178Y (37). Subsequently, many laboratories, using various other cell culture systems and end points, arrived at similar conclusions (38). Tea polyphenols have been shown to inhibit the activities of transcription factors AP-1 and NFκB and synthesis of nitric oxide (39–42). Inhibition of cell transformation and cell growth by purified catechins and theaflavins has been reported (43). These activities have been attributed to the inhibition of AP-1 activity, possibly due to the inhibition of several steps of signal transduction pathways, for example, mitogen-activated protein (MAP)-kinase activities by tea polyphenols (44). It has been reported that tea and its polyphenolic constituents impart inhibitory effects on the activities of many enzymatic and metabolic pathways relevant to cancer development (36 and references therein). Based on these extensive data, clinical trials in human cancer patients with green tea are being conducted or planned (45).

**Experimental Studies on Tea and Prostate Cancer**

In a preliminary study on the effects of green tea and prostate cancer, Liao et al. showed that i.p. administration of EC1G rapidly reduced the size of human prostate tumor growth in nude mice (46). These authors further suggested that there might be a possible relationship between the higher consumption of green tea and the lower incidence of prostate cancer in some Asian countries (46). Based on this initial work and our ongoing research program on green tea and cancer chemoprevention, we started to systematically evaluate the effect of green tea consumption on prostate carcinogenesis (16). We initially showed that ornithine decarboxylase (ODC), a rate-controlling enzyme in the polyamine biosynthesis pathway, is overexpressed in prostate cancer and prostate fluid in humans (47). The induction of ODC activity is known to be mediated by high testosterone levels and exposure of prostate cancer cells to EGCG, and, in Cpb:WU rats, infusion of green tea caused a down-regulation of ODC activity in prostate cancer cells (16). We then reasoned that these preclinical studies should be conducted in a model system that mimics human disease using human achievable doses of green tea. Transgenic adenocarcinoma of the mouse prostate (TRAMP) is one such model for prostate cancer that closely mimics progressive forms of human disease (48). In this model, we provided convincing evidence that oral infusion of green tea polyphenols (equivalent to 6 cups of green tea, human consumption) inhibits prostate carcinogenesis (49). It has been reported that tea polyphenols inhibited the in vitro formation of the heterocyclic amine 2-amino-1-methyl-6-phenylimidazo[4,5-b]pyridine (PhIP), a suspect human prostate carcinogen (50). Because it is known that the development of prostate cancer depends upon a variety of factors as it progresses from small latent carcinoma to a large metastatic lesion, it is important to establish the stage of prostate carcinogenesis that is most responsive to green tea. We have developed plans to investigate this in a systematic fashion.

**Androgen Synthesis, Androgen Receptor, and Tea Polyphenols**

Descriptive epidemiological data suggest that androgens play a key role in prostate carcinogenesis (51). Because androgens are capable of both stimulating proliferation as well as inhibiting the rate of the glandular epithelial cell death within the prostate, androgen ablation therapy is commonly suggested for men with this non-organ-confined disease (52). In 1941, Huggins and Hodges first demonstrated the utilization of androgen deprivation as a treatment for advanced prostate cancer (53). Removal of androgenic stimulation has been used to treat metastatic disease for some time (54). Kao et al. reported that treatment with EGCG (85 mg/kg body weight) for 7 days reduces the circulating testosterone by 70% in male Sprague-Dawley rats, which may be associated with lowering the risk of prostate cancer (34). However, the mechanism through which tea polyphenols decrease the androgen level remained unknown at that time. Further studies have shown that tea polyphenols might block the pathway that leads to the synthesis of androgen. Populations with impaired androgen metabolism such as congenital 5α-reductase deficiency (a key enzyme that converts testosterone to 5α-dihydrotestosterone) do not develop prostate cancer, whereas those with higher circulating levels of androgen are at higher risk of prostate cancer (55). In studies of prostate cancer tumors grafted onto animals, the 5α-reductase inhibitors were found to slow the growth of previously established prostate cancer cell lines (56). The inhibition of 5α-reductase is suggested to prevent prostate cancer in rats, and various 5α-reductase inhibitors such as finasteride are approved by the U.S. Food and Drug Administration for the treatment of benign prostatic hyperplasia in humans (56–58). It has been reported that the green tea constituents EGCG and ECG (5–200 μM) inhibit the activity of type 1 rat 5α-reductase (59, 60). These polyphenols also inhibited types 1 and 2 human 5α-reductase in microsomes from rat cells that expressed the human enzyme. The IC50 values for EGCG and ECG were 15 and 12 μM, respectively (59). The relevance of these studies to in vivo prostate tumor models has not been evaluated. Several molecular mechanisms have been postulated to be responsible for the development of recurrent hormone-refractory tumors. Most of these mechanisms involve alterations in the function of the androgen receptor (AR) and its complex signaling pathways (61). Recent studies have shown that AR is expressed in all stages of prostate cancer, and at least one-third of advanced prostate cancers contain amplified AR genes (62–64). It is suggested that overexpression or mutation of the AR in prostate cancer cells may promote a growth advantage. Therefore, it has been of great interest to seek more effective means of minimizing or eliminating the function of the AR to achieve preventive and/or
therapeutic treatments for prostate neoplastic disease. Ren et al. have reported that tea polyphenols down-regulate the expression of the AR in LNCaP prostate cancer cells (65). They reported a significant reduction in AR mRNA by EGCG treatment. The basal activity of AR promoter is determined by a Sp1 binding site within the AR core promoter region (66,67). Sp1 is involved in the expression of genes related to cell proliferation (68). Because Sp1 regulates the expression of many critical genes, the decrease in this protein by tea polyphenols could somewhat decrease the growth rates of prostatic cells. EGCG has been reported to significantly decrease the Sp1 DNA binding activity (65). Prostate cancer is known to undergo a transition from an early androgen-sensitive form of cancer to a late androgen-insensitive cancer. The published results of this laboratory showed that EGCG is capable of inhibiting the prostate cancer cell growth irrespective of their androgen association, in a dose- and time-dependent manner (69). Much additional work is required to substantiate that tea constituents could affect androgen synthesis or its receptor in a manner that could lead to the protection against prostate cancer development.

Prostate Specific Antigen and Tea Polyphenols

Prostate specific antigen (PSA), a glycoprotein secreted by the prostate gland, is the most clinically used marker for prostate cancer (70). PSA is a serine protease produced by the epithelial cell lining of the acini and ducts of the prostate gland. It circulates in the serum in both free (unbound) and complexed forms. The most common cause for an elevated serum PSA is benign prostatic hyperplasia, the incidence of which increases with age, similar to prostatic cancer. It has been suggested that serum PSA levels can be decreased by the agents that lower serum testosterone levels such as leutinizing hormone-releasing hormone agonists and antagonists, anti-androgens such as flutamide and bicalutamide, and the 5a-reductase inhibitors such as finasteride (71 and references therein). Earlier published reports show that green tea polyphenols decrease the PSA levels in human prostate cancer cells (LNCaP) in a dose-dependent manner in the culture medium (65,72).

Polyamine Synthesis and Tea Polyphenols

Studies have demonstrated that prostate contains some of the highest concentrations of polyamines and polyamine-metabolizing enzymes (73,74). ODC is a key regulatory enzyme for polyamine synthesis, and the induction of its activity has been reported to be linked with various types of cancers including prostate cancer (47,75–77). Hence, ODC has been used as a biomarker for chemopreventive studies for a long time (78). An induction in the ODC activity and ODC mRNA expression mediated by testosterone has been reported in prostate cancer cells (79,80). This laboratory has reported that green tea polyphenols significantly reverse the induction of ODC activity as well ODC mRNA expression in LNCaP cells. Testosterone, when administered to C57 BL/6 mice, caused a twofold increase in ODC activity in the ventral prostate, whereas prior oral infusion of 0.2% (w/v) green tea polyphenol in drinking water resulted in 40% inhibition in this induction (16).

Gene Expression and Tea Polyphenols

Cancer is a complex process, and several genes play an important role at various stages and at different levels. These include the genes that regulate growth, cell signaling, differentiation, cell death, cell division, and cell migration. Such genes are either overexpressed or suppressed in cancer development. Tea polyphenols have been shown to modulate the function of various genes at various levels. Briefly, we have identified nine genes, including six kinases and three phosphatases, whose expression was found to be down-regulated by EGCG in human prostate LNCaP cancer cells. These genes are related to the G-protein signaling network and are responsible for cell proliferation and include adenosine kinase, protein kinase C (PKC)-ε, and type I β cGMP-dependent protein kinase (81). PKC is involved in diverse cellular functions, including cell differentiation, growth control, tumor promotion, and cell death. It is also a regulator of cell cycle events during G1 progression and G2/M transition (82 and references therein). Recent studies demonstrate that inhibition of PKC-ε gene expression could inhibit cell proliferation in the animal tumor model and in some human cancer cell lines (82). Our laboratory has shown that EGCG inhibits the expression of the PKC-ε gene, adenosine kinase, and type I β cGMP-dependent protein kinase in LNCaP cells and hence is able to block the intracellular cyclic-nucleotide signaling cascade. It was observed that the genes repressed by EGCG are broadly related to the G-protein signaling network, therefore, implicating a portion of the G-protein signaling network in the early stage of prostate cancer chemotherapy (81). The loss of tumor suppressor genes and the genes producing antigrowth factors is an important event in cancer development. We have shown that EGCG induces the expression of 16 kinases and phosphatase genes in prostate cells. These include tumor suppressor gene SHP-1 and the genes that produce pyrroline-5-carboxylate and prostatic acid phosphatase (81). Prostatic acid phosphatase and pyrroline-5-carboxylate inhibit growth by deactivation of erbB-2 and p38 MAP kinases and survival of cancer cells, respectively (83,84). The p53 tumor suppressor gene is the most frequently mutated gene found in human malignancies, including prostate cancer. Generally, no correlation between p53 mutation and early-stage prostate cancer has been noticed, but p53 mutations are shown to be associated with a small group (10–20%) of advanced prostate cancer patients with high Gleason score and distant site metastasis (85,86). Our laboratory has reported that EGCG up-regulated p53 in LNCaP cells (with wild-type p53) but not in DU145 cells (with mutant p53) (69). The stabilization of p53 is a critical step during the induction of apoptosis. Recently, we have shown that EGCG-induced apoptosis in LNCaP cells occurs
through the stabilization of p53 by phosphorylation on critical serine residues and p14ARF-mediated down-regulation of MDM2 protein (87).

**Programmed Cell Death (Apoptosis) and Tea Polyphenol**

Apoptosis is the most common form of eukaryotic cell death, acting as a physiological suicide mechanism to preserve homeostasis, and occurs naturally during tissue turnover (88). As is true in other normal cells, in normal prostate a balance between cell growth and cell death is maintained. In prostate cancer, this balance is lost in favor of cell growth. Correction of this imbalance could lead to the prevention and even ablation of prostatic cancer (89,90). Because human prostate cancer is present as a heterogeneous mixture of androgen-dependent and -independent cells, surgery and chemotherapy have failed to address this problem. Hence, one potential strategy to eradicate this mixture of cells is to modulate the apoptotic machinery by chemopreventive agents. Studies from this and other laboratories have shown that EGCG results in an induction of apoptosis in several human carcinoma cells (37,91). Previously, our laboratory has demonstrated that EGCG results in the apoptosis of both androgen-insensitive DU145 as well as androgen-sensitive LNCaP cells (69). These results were confirmed by others (92). Recently, we have shown that EGCG-induced apoptosis in human prostate carcinoma LNCaP cells is mediated via modulation of two related pathways: 1) stabilization of p53 and down-regulation of MDM2 protein and 2) negative regulation of NK-κB activity, thereby decreasing the expression of the pro-apoptotic protein Bcl-2 (87).

**Angiogenesis in Prostate Cancer and Tea Polyphenols**

The ability of cancer cells to move from their original sites and invade surrounding tissues is a phenomenon that makes cancer a deadly and life-threatening disease. Invasion of cancer cells to new sites is subsequently followed by new blood vessel formation (angiogenesis). Angiogenesis is a major event of later stages of carcinogenesis. Some hydrodases and matrix metalloproteases (MMPs) have been reported to be overexpressed during the invasion of cancer cells and angiogenesis (93,94). Once tumors become aggressive and metastasize to other organs, even systemic chemotherapy may be in vain. Jankun et al. reported that EGCG inhibits urokinase, implicated in tumor invasion (95). EGCG was found to inhibit tumor cell invasion and directly suppress the activity of MMPs MMP2 and MMP9, two of the proteases most frequently overexpressed in cancer and angiogenesis and essential in penetrating the basement membrane barriers (96–98). Oral feeding of tea polyphenol to TRAMP mice (which develop prostate cancer spontaneously) inhibited metastasis and angiogenesis by inhibiting MMPs and vascular endothelial growth factor (99). This was found to be due to inhibition of vascular endothelial growth factor, a marker of angiogenesis, and also due to suppression of the activities of MMP2 and MMP9 (99). Recently, it was shown that tea components slow progression of LNCaP human prostate tumors in SCID mice, partly by inhibiting the formation of new blood vessels (100).

**Insulin Growth Factor in Prostate Cancer and Tea Polyphenols**

Insulin-like growth factor (IGF-1) has been strongly implicated in the etiology of human prostate cancer. Recent studies have demonstrated that elevated circulating levels of IGF-1 are associated with increased risk of several common cancers, including those of the breast, prostate, lung, and colorectum (101). The level of IGF-binding protein (IGFBP-3), a major IGF-1 binding protein in serum that, in most situations, suppresses the mitogenic action of IGF-1, has been shown to be inversely associated with the risk of these cancers (102). The elevated levels of IGF-1 with concomitant lowering of IGFBP-3 levels in serum are excellent predictors of prostate cancer progression in humans. The identification of agents that inhibit the IGF-1 signaling pathway could lead to the development of highly successful prevention strategies for prostate cancer. IGF-1 has been implicated as an important factor in the initiation and progression of prostate cancer in TRAMP mice (103), and we observed that oral infusion of green tea polyphenols significantly lowered the IGF-1 levels and restored the deficient levels of IGFBP-3 in TRAMP mice (49). These results suggest that prostate regression induced by green tea polyphenols may be related to alterations in availability of IGF-1 as a result of increased production of IGFBP-3. However, the mechanism that leads to the modulation of IGF-1/IGFBP-3 by green tea polyphenol needs to be further investigated.

**Proteasome Activity in Prostate Cancer and Tea Polyphenols**

The ubiquitin-proteasome system plays a critical role in the specific degradation of cellular proteins, and two of the proteasome functions are to allow tumor cell cycle progression and to protect tumor cells against apoptosis (104–106). In addition, the chymotrypsin-like but not trypsin-like activity of proteasome is associated with tumor cell survival (107). Many cell cycle and cell death regulators such as p53, pRB, p21/CIP1, p27/KIP1, 1xB-α, and Bax have been identified as targets of the ubiquitin-proteasome-mediated degradation pathway (108). Recently, the ubiquitin-proteasome system was shown to be involved in the regulation of AR protein in prostate cancer cells (109). Ester bond-containing tea polyphenols, such as EGCG, potently and specifically inhibit the chymotrypsin-like activity of the proteasome in vitro [IC(50) = 86–194 nm] and in vivo (1–10 μm) at the concentrations found in the serum of green tea drinkers (110). This inhibition of the proteasome by EGCG in several tumor and transformed cell lines results in the accumulation of two natural proteasome substrates, p27(Kip1) and 1xB-α, an inhibi-
tor of transcription factor NF-κB, followed by growth arrest in the G1 phase of the cell cycle. This study suggests that the proteasome is a cancer-related molecular target of tea polyphenols and that inhibition of the proteasome activity by ester bond-containing polyphenols may contribute to the cancer-preventative effect of tea (110). GTP analogs (+)-EGCG and (−)-GCG potently and specifically inhibit the chymotrypsin-like activity of purified 20S proteasome and the 26S proteasome in tumor cell lysates, whereas benzyol-protected (+)-EGCG does not (111). Treatment of prostate cancer LNCaP cells with either (+)-EGCG or (−)-GCG accumulated p27 and IκB-α proteins, associated with an increased G1 population. (+)-EGCG treatment also accumulated the pro-apoptotic Bax protein and induced apoptosis in LNCaP cells expressing high basal levels of Bax but not prostate cancer DU-145 cells with low Bax expression. The synthetic GTPs significantly inhibited colony formation by LNCaP cancer cells (111).

The Cell Cycle in Prostate Cancer and Tea Polyphenols

A controlled cell cycle progression is also an important physiological event that is regarded to be essential for normal tissue homeostasis, and in recent years cell cycle-mediated apoptosis is being increasingly appreciated (88). Most of the cancer types including prostate cancer possess defects in one or more cell cycle checkpoints (112–114). Normal cell cycle progression relies on the cell’s ability to translate extracellular signals such as mitogenic stimuli and intact extracellular matrices to efficiently replicate DNA and divide. Cyclin-dependent kinases (cdk’s) respond to these signals and push cells through the cell cycle. Cyclins are cdk-binding partners that are required for kinase activity, and their protein levels are intimately linked to the cell cycle stage. Abnormal cdk activity is accomplished by cyclin amplification, cdk or substrate mutation as well as inactivation of inhibitors such as WAF1/p21, INK4a/p16, and INK4c/p18 (115). The selective growth advantage of cancer cells also stems from amplification of positive growth signals and mutation of checkpoints. The loss of cell cycle checkpoints results in the selection of cells that have a growth advantage that may result in drug resistance, invasion, and metastasis (116). Also, in recent years, inhibition of the cell cycle has been appreciated as a target for the management of cancer (117). A few reviews are available that contain information about the dysregulation of the cell cycle in prostate carcinogenesis (113,114). Tea polyphenols have been shown to arrest cell division of cancer cells and enhance the expression of cdk inhibitors (118). Earlier, our laboratory showed that ECGC exhibited a dose-dependent arrest of cells in the G0/G1 phase of the cell cycle in prostate cancer cells, thereby slowing down the growth of prostate cancer cells (69). Recently, we elucidated the molecular mechanism involved in cell cycle arrest in human prostate carcinoma cells (119). We observed that ECGG treatment of LNCaP and DU-145 cells resulted in significant dose- and time-dependent 1) up-regulation of the protein expression of WAF1/p21, INK4a/p16, and INK4c/p18, 2) down-modulation of the protein expression of cyclin D1, cyclin E, cdk2, cdk4, and cdk6 but not of cyclin D2, and 3) increase in the binding of cyclin E toward cdk2. This series of events imposes a blockade of G1 to S transition, causing a G0/G1 phase arrest of the cell cycle. However, the effect of ECGG on the interlinking among the different components of the cki-cyclin-cdk network needs further investigation.

Epidemiological Studies

As of yet no detailed case-control study has been conducted to assess the effect of consumption of green tea on human prostate cancer. All published data seeking an association between tea consumption and the risk of prostate cancer considered undefined tea preparations, mostly black tea (Table 2). At least two epidemiological studies have shown that people who regularly consume tea have a lower incidence of prostate cancer (120,121). Heilbrun et al. (120), in a prospective cohort study employing 7,833 men living in Hawaii with Japanese ancestry, observed a weak but significant negative association between black tea intake (more than one cup per day) and prostate cancer incidence (P = 0.02). In a case-control study conducted in three geographical areas of Canada, Jain et al. (121) observed a decrease in prostate cancer risk with tea intake of more than two cups per day. Other epidemiological studies conducted in Italy (122), Utah (123), and Canada (124) did not find any difference of risk for prostate cancer between tea drinkers and nondrinkers. However, most of these studies include populations that were predominantly black tea drinkers. It should be noted that most of these stud-

Table 2. Summary of Published Epidemiological Studies Showing an Association Between Tea and Prostate Cancer

<table>
<thead>
<tr>
<th>Location</th>
<th>Tea Type</th>
<th>Sample Size</th>
<th>Tea Intake</th>
<th>OR/RR</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hawaii, USA</td>
<td>Black tea</td>
<td>149 cases, 7,833 subjects</td>
<td>2–4 cups/week</td>
<td>RR = 0.4</td>
<td>120</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>&gt;1 cup/day</td>
<td>RR = 0.6 (P = 0.02)</td>
<td></td>
</tr>
<tr>
<td>Canada</td>
<td>Not defined</td>
<td>335 cases, 344 controls</td>
<td>0–500 g/day</td>
<td>OR = 0.89 (95% CI = 0.69–1.16; P = 0.05)</td>
<td>121</td>
</tr>
<tr>
<td></td>
<td></td>
<td>99 cases, 124 controls</td>
<td>&gt;500 g/day</td>
<td>OR = 0.7 (95% CI = 0.5–0.99; P = 0.05)</td>
<td></td>
</tr>
<tr>
<td>Italy</td>
<td>Black tea</td>
<td>107 cases, 6,147 controls</td>
<td>≥1 cup/day</td>
<td>RR = 0.9</td>
<td>122</td>
</tr>
<tr>
<td>Utah, USA</td>
<td>Not defined</td>
<td>179 cases, 385 controls</td>
<td>1–5 cups/week</td>
<td>OR = 0.75 (95% CI = 0.47–1.2)</td>
<td>123</td>
</tr>
<tr>
<td>Canada</td>
<td>Black tea</td>
<td>145 cases, 3,400 subjects</td>
<td>&gt;5 cups/week</td>
<td>OR = 1.06 (95% CI = 0.72–1.57)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>500 ml/day</td>
<td>RR = 1.02 (95% CI = 0.62–0.65)</td>
<td>124</td>
</tr>
</tbody>
</table>

*Abbreviations are as follows: OR, odds ratio; RR, relative risk ratio; CI, confidence interval; P, P value for trend.

Vol. 47, No. 1
ies lacked appropriate controls for comparison in categorization of tea consumption, the type of tea consumed, and the ethnicity of the subjects, which weakens the overall impact of the study. Epidemiological investigations seeking an association between green tea and prostate cancer should be undertaken to establish the validity of cell culture and animal data to human prostate cancer patients.

Clinical Trials with Green Tea

One recent phase II clinical trial explored the antineoplastic effects of green tea in patients with metastatic androgen-independent prostate cancer (125). In this study, 48 patients were instructed to take 6 g of green tea per day orally in six divided doses. Patients were monitored monthly for response and toxicity. Only 1 patient within this 42-patient cohort manifested a 50% decline in PSA values from baseline, and this decrease was not sustained beyond 2 mo. Although green tea was tolerated well for the most part, a notable percentage of patients did experience toxicity such as insomnia and fatigue, presumably from caffeine present in the tea. However, it should be noted that this study was conducted in patients with metastatic androgen-independent prostate cancer and, therefore, in principle, is not representative of the chemopreventive effects of green tea. Moreover, in this trial, the median time of study was only 1 mo, whereas previous preclinical data suggest that green tea requires prolonged exposure to exert its antitumor activity (14,97,126). Further, this trial was conducted in patients with androgen-independent prostate carcinoma only, and it is possible that green tea may exert antineoplastic effects in patients with hormoneresistant prostate carcinoma. Considering the drawbacks of this trial, the negative findings of this study do not negate the results of previous epidemiological studies that suggest that green tea may confer an antitumor effect in a relatively healthy population. For an ideal prospective study, a population with a high risk for prostate cancer development should be considered and the length of exposure of green tea to the subjects should be taken into consideration.

Future Perspectives

On the basis of information gained to date in mice or in human prostate cancer cell culture systems, tea or its constituents have been shown to cause cell cycle dysfunction, induce apoptosis, inhibit enzymes and signaling pathways associated with cellular proliferation, inhibit tumor growth, and delay onset and progression to tumorigenesis. Although studies on tea polyphenols demonstrate efficacy as a potent chemopreventive agent against prostate cancer, there are still many gaps in our existing knowledge before a recommendation could be made for tea polyphenols as preventive or therapeutic agent to humans. There are certain factors, such as bioavailability, tissue level of tea constituents, tea type, drinking habit, and race, that need to be considered before recommending tea polyphenols as therapeutic agents against prostate cancer. Knowledge of the bioavailability of tea constituents would allow a clearer interpretation of the mechanisms by which tea polyphenols exert their cancer preventive effects. The bioavailability of the active polyphenolic constituents after tea consumption by laboratory animals and humans is poorly defined. Studies conducted by Yang et al. have measured the concentration of tea polyphenols in human plasma, saliva, feces, and urine after consuming decaffeinated green tea (127–129). The bioavailability of EGCG was found to be less than that of EGC. Chronic consumption of green tea (0.6% w/v) in the drinking water of rats and mice showed that green tea polyphenols available to prostate were 245 ng/g compared with other organs such as bladder (800 ng/g), kidney (450 ng/g), etc. (130). Whether this concentration and its duration of availability of tea polyphenol in prostate are sufficient to exert the observed in vitro effects is not known and needs further investigation.

Therefore, a great deal of laboratory research and many more epidemiological studies are needed for obtaining conclusive evidence. The increase or decrease of PSA and IGF-1 and IGFBP-3 in relation to tea consumption and levels of tea polyphenols in urine samples may be used as biomarkers in prostate cancer chemoprevention. This type of study could answer the question of how much tea should be consumed by humans for prostate cancer chemoprevention and explain the mechanism involved. Prostate cancer is a disease of many etiological factors and involves several mechanisms in its progression. Therefore, the modulation of a single mechanism alone by tea polyphenols may not completely stop the progression of prostate cancer. However, tea polyphenols have been shown to modulate various molecular targets involved in progression of prostate cancer as discussed in this review. To date, there is no information available that links various molecular pathways involved in the progression of prostate cancer to each other. Presently, the combinational studies of cancer chemopreventive agents are areas of research interest (131). Various chemopreventive agents in combination may produce synergistic or additive effects (100,132–134).

Tea is one of the few agents known to inhibit tumorigenesis at the postinitiation stage and is generally safe to use. Tea shows broad chemopreventive ability from inhibition of carcinogen formation to suppression of prostate cancer progression. The uniqueness of its action lies in its specificity of killing cultured cancer cells without affecting the growth of normal cells. Integrating mechanistic studies in vitro and in vivo and paying major attention to the bioavailability of tea constituents and its metabolites, an in-depth study in animal models and humans is warranted to establish the chemopreventive potential of tea polyphenols against prostate cancer. The data obtained to date are promising and should form the basis to further examine the utility of tea in chemoprevention of human prostate cancer.
Acknowledgments and Notes

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Vol. 47, No. 1

21


22 Nutrition and Cancer 2003


