Green Tea and Leukoplakia

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BACKGROUND: A feasibility chemoprevention trial of green tea and oral leukoplakia has been conducted. The purpose is to demonstrate the success and pitfalls of conducting such a population-based trial within Indian rural villages.

METHODS: Two dosages of green tea were utilized: 3.6 g per day and 5.4 g per day. Compliance was measured using tea package counts and by measuring the blood concentration of the active anticancer compound in green tea, (−)-epigallocatechin-3-gallate (EGCG).

RESULTS: Following a 3-year planning period, 13 villages were visited, and 1,203 tobacco users were interviewed in order to identify 64 participants suitable for trial inclusion. Of the subsequently 28 consenting individuals entered, 23 remained on protocol for the entire 6-month duration. Overall compliance was excellent, with participants consuming approximately 80% of prescribed packets.

CONCLUSIONS: These results provide a basis for continuing international collaborative efforts in conducting population-based chemoprevention trials against head and neck cancer. *Am J Surg.* 1997;174:552–555. © 1997 by Excerpta Medica, Inc.

Head and neck cancer represents the fifth most common cancer worldwide. In certain countries such as within central Asia, it is the most common neoplasia.[1] Unfortunately, given the continuing spread of tobacco use among developing countries, the mortality from head and neck cancer will not likely decrease in the coming years. Indeed, as the total world population increases and as the average lifespan of the world population improves, we are likely to see a further increase in the total number of individuals with the disease.[2] International efforts that merge the resources and research advances of richer nations with the expertise within medically underserved developing countries in which the disease is most common are required to effectively address this issue.

One strategy in future years that may be critical in lowering head and neck cancer mortality resides within the discipline of chemoprevention. The majority of chemopreventive trials of oral carcinogenesis have involved patients with oral leukoplakia, a precursor lesion to oral cancer.[2] However, the results of these trials, which principally involve vitamin A and vitamin A derivatives as well as carotenoids, have given mixed results.[3–5] An improved understanding of the determinants of chemopreventive agent response is required.

The present study was intended to explore the possibilities of conducting population-based chemoprevention trials in regions at high risk for head and neck cancer. The purpose was to explore the feasibility of these trials both in terms of patient compliance and trial conduct in light of limited available resources. We used as the agent in this feasibility trial, green tea.[6–8] The agent is a naturally occurring foodstuff, affordable to the population at risk, and is considered nontoxic. The potential cancer preventive effect of this agent has been the source of considerable research by both epidemiologic and basic science investigators.[6–8]

MATERIALS AND METHODS

Patient Population

This was a randomized trial involving noncancer-bearing healthy volunteers with previously untreated oral leukoplakia. All persons were more than 18 years of age. All had bidimensionally measurable disease and a Karnofsky performance status greater than 80%. Individuals with acute intercurrent illness or history of cancer were excluded from analysis. A complete history of alcohol and tobacco use was obtained from each participant.

Each leukoplakic lesion was photographed, and punch biopsies were performed in the central portion of each lesion before tea initiation. All biopsied lesions were evaluated for the presence of dysplasia by a single pathologist (AGH) using previously described criteria.[9] Informed consent was obtained from all individuals prior to trial entry. This study was conducted in compliance with Helsinki II agreement and was approved by the Institutional Review Board of the American Health Foundation.

Treatment Plan

This pilot study employed a two-arm study to evaluate the feasibility of launching in the future a randomized clinical trial with green tea (Thomas J. Lipton, Co.) in capsule or liquid form and at one of two doses. A total of 28 participants with precancerous lesions of the oral cavity was proposed to provide a moderately precise estimation of compliance and toxicity rates across a wide range of oral lesion pathology. In addition, the pilot study was intended to provide an assessment of the field logistics and procedures with a case load requiring the full attention of the on-site senior investigator. Each participant was randomized to one of two possible quantities of tea: 3.6 g or 5.4 g per day, each to be consumed in three equally divided doses (1.2 g or 1.8 g in 8 oz water). Fourteen individuals were entered into each arm. Participants were asked to consume tea for 6 months.

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Follow-up

Measurement of compliance was performed at clinic visits scheduled each month during the 6-month trial. Protocol compliance was measured by tea packet count. At each clinic visit the participant brought the tea packet container, and the number of remaining packets were counted and recorded on the data collection form. The counted packets were compared with the number given to the participants. Given restrictions secondary to the high rate of illiteracy from the participants, the initial requirements of maintaining a medication calendar of daily tea consumption was dropped from the study. In addition, compliance was assessed through spot assessments of blood EGCG ((−)-epigallocatechin-3-gallate, the active anticancer compound in green tea) levels in 4 randomly identified individuals. The methods for EGCG measurement had been established by one of us (CSY) and previously reported.[10] Results are under generation and will be presented elsewhere. During the first week of consumption, individuals who consumed less than 85% of the provided preparation were discontinued from the study and a replacement participant was identified.

Toxicity was assessed using the Division of Cancer Treatment NCI Common Toxicity Criteria table (Cancer Therapy Evaluation Program, Division of Cancer Treatment, National Cancer Institute, Bethesda, Maryland) and was assessed at each clinic visit. Likewise, counseling regarding alcohol and tobacco cessation was performed at each clinic visit. All clinic visits to assess for compliance and toxicity were performed at 1-month intervals following study entry.

Of note, clinical response was not a primary endpoint of this study. All patients remain under observation and will be assessed for outcome after adequate follow-up has been achieved. Results will be reported separately.

Surrogate Endpoint Biomarkers

Although not to be presented in this study and although not a primary goal of this feasibility study, the conduct of this trial allowed us to assess the presence of surrogate endpoint biomarkers (SEB) both within the tissues at risk as well as within the urine. The principal SEB under evaluation was 8-hydroxy-deoxyguanosine (8-OH-dGuo), a measure of oxidative damage to DNA. From each individual, a 20-mL urine sample was obtained in a plastic container without preservatives or other additives and stored at −20°C until analysis. Samples were obtained preconsumption and at 1, 2, 4, 6, and 12 months. Methods of assessment and results of 8-OH-dGuo will be reported separately.[11]

RESULTS

Population Recruitment and Characteristics

Thirteen villages in Goa, India, with populations varying from 1,000 to 6,000 were visited in order to recruit the identified participants. A total of 1,203 individuals with a history of tobacco use were interviewed. From this population, 64 individuals were identified who had leukoplakia lesions and who met criteria for trail entry. Of the 28 individuals who were entered into the study, 4 dropped out of the trial after 1 to 4 weeks of consumption for varying reasons unrelated to tea consumption, ie, changed location, automobile accident, and so on. One individual stopped after 5½ months of treatment. Thus, 23 of the original 28 participants finished the entire 6 months of treatment. Of the 4 who initially were discontinued prior to the first month, all were replaced by additional participants who then went on to complete the entire 6-month tea consumption period. One participant dropped out of the trial after 5 months.

TABLE

Participant Characteristics

Characteristics	Number of Participants
Gender	
Male	25
Female	3
Tobacco habits	
Bidi	13
Cigarettes	4
Bidi + other	8
Other	3
Alcohol	
Feni	12
Other	4
None	12
Site of leukoplakia*	
Buccal mucosa	39
Hard palate	1
Soft palate	2
Ginguia	1
Lip	4
Histology†	
Hyperplasia	6
Mild dysplasia	8
Moderate dysplasia	2
Severe dysplasia	6

** Some participants had more than one site of disease.*
† No biopsy samples were obtained on six participants.

The **Table** provides the demographic information of 28 patients who completed at least 5 months of tea consumption. The age range of the population was 40 years to 72 years. There were 25 men and 3 women. The tobacco and alcohol use patterns are also detailed in the Table. All participants consumed tobacco, with bidi smoking representing the most common form of consumption. This is consistent with the tobacco habits of the Goa population as previously identified by the coprincipal investigator of this study (SGV).[12] Of those individuals who used alcohol, the majority used a local brew called feni. All but 2 of the participants were actively employed. Twenty-three of the 28 participants had no formal education and were considered illiterate.

The majority of leukoplakia lesions were on the buccal mucosa near the oral comissure. Additionally, lesions were identified on the hard and soft palate, gingiva, and lip. Biopsies were performed on 22 (79%) of the 28 patients who remained in the study. Each individual had only one site biopsied pretreatment. Histologic assessment of the biopsy samples are, likewise, detailed in the Table. Overall, the majority of lesions were graded as mild or moderate dysplasia.

Tea packet counts were made at each monthly visit. Overall compliance in consuming and counting the required packets was excellent. Consumption by the population as a whole approximated 80% of the packets. Six of the pa-

tients failed to consume less than 80% of the packets in at least 3 of the 6 months. Monthly consumption in this latter group of 6 individuals averaged 66% during those periods. Compliance measurements also included the quantitation of EGCG within the peripheral blood. These measurements have not yet been completed and are to be reported separately. Compliance was not influenced by the amount of tea consumed, ie, 3.6 g per day or 5.4 g per day.

No significant toxicities were reported during this period that could be ascribed to the green tea. One patient developed pneumonia during the 6-month interval. There were two episodes of soft tissue infections and one episode of Herpes zoster infection.

COMMENTS

This study demonstrates that the conduct of population-based chemoprevention trials is feasible in rural underserved high-risk regions within central Asia. The compliance of the 28 patients in completing 6 months of tea consumption with cancer-preventive intent was achieved to a sufficient degree to justify further chemoprevention trials within this setting. The average number of packets consumed over the 6-month interval of testing approximated 80%. Furthermore, all but one drop-out occurred early, ie, less than 1 month from study initiation. Compliance was especially significant given the socioeconomic conditions that were prevalent. Average education levels as well as average annual income were low, both of which have been reported to adversely influence results.[13–18] We initially considered a study to evaluate overall compliance in those who consumed tea as a pill rather than as a drink. However, we noted sufficient compliance when taking the latter preparation that the pill arm was deemed no longer necessary.

Both the levels and determinants of compliance represent critical issues in chemoprevention trials.[13–18] One limitation in our understanding of these issues may stem from the limited inclusion of minorities and different ethnic populations in ongoing funded trials, a concern that is generating increasing debate. The majority of chemoprevention trials involving head and neck cancer patients have dealt with populations who may not be at greatest risk. For instance, recent SEER data have demonstrated that head and neck cancer has increased dramatically in black males. From 1973 to 1991 the incidence of squamous cell carcinoma of the oral cavity and pharynx has increased dramatically by 38%.[19] Yet, despite these very significant trends, black males are poorly represented in oral cancer prevention efforts. A recent community based trial involving vitamin E as an agent against oral leukoplakia was one of the first trials to draw attention to this fact.[20] This latter trial was noted to involve only 5% blacks, well below the proportion of head and neck cancer patients represented by this population.[20] Understanding the problems unique to high-risk populations must be considered if we are to reduce the problem of head and neck cancer in the United States, India, or other countries worldwide.

Multiple determinants govern the successful conduct of chemoprevention trials. Tangrea[13] has recently summarized these concerns, which include such issues as delays in patient accrual, beliefs of the accruing physician, available clinical trial infrastructure, informed consent process, as well as overly restrictive and complex clinical trial design. To address individually each of these considerations is beyond the scope of this study. One major strength of this study is the demonstrable success in recruitment efforts. Individual participants were identified within a 9-month interval despite the fact that recruitment was drawn from multiple localities and by a single recruitment team consisting of three individuals. In comparison, a recent community chemoprevention trial conducted by Benner et al[20] in the United States required 19 months to accrue 43 individuals. This was despite participation of 37 institutions. One obvious explanation is the increased prevalence of leukoplakia that exists within India. Additional considerations may relate to patient expectations, the manner in which the participants were recruited, ie, home-based versus either office- or institutional-based, and attitudes and competing commitments of the recruiting staff.

It should be emphasized that previous international cooperative chemoprevention trials have been conducted in rural settings. In a 1984 trial in the Phillipines involving the Ifugaos tribe, 132 participants were treated with either vitamin A, beta-carotene, or canthaxanthin.[21] Thirty-four percent of the enrollees never completed the trial. In another study involving the Inuits of Canada, drop-out rates approximated 10% of the 27 individuals entered in a beta-carotene trial designed to reverse leukoplakia.[22] Our results are comparable to these lower drop-out rates.

The choice of green tea as a chemopreventive agent was based on the preclinical information that demonstrates its chemopreventive potential. Several epidemiologic studies, although not all, have demonstrated a reduction in cancer incidence among tea drinkers.[6,7] The cancer-protective effect seems more pronounced for digestive tract cancers, including squamous cell carcinoma of the upper aerodigestive tract.[6,7] However, laboratory investigations suggest that the site of disease is not as critical as the carcinogenic mechanisms involved. The protective components within green tea consist of the plant polyphenol family of which one of the most effective compounds is the flavanol, (−)-epigallocatechin-3-gallate (EGCG). The polyphenolics comprise 30% of the dry weight of tea extract solids. These phenols can exert their protective effect in multiple ways, including the inhibition of nitrosation, the activation of host detoxifying enzymes, and the modulation of arachidonic acid metabolism, as well as functioning as free radical scavengers.[7,8] Pertinent to the problem of tobacco-induced disease, multiple animal studies have demonstrated the capacity of supplemental tea intake to inhibit carcinogenesis induced by tobacco-specific nitrosamines as well as benzo (a) pyrene.[23,24] Relevant to oral carcinogenesis, the levels of EGCG following the intake of a single cup of tea are higher in the saliva than in the blood, suggesting additional rationale for the use of tea as a preventive agent against tobacco-induced oral cancers (unpublished data from C. S. Yang). Future reports of this trial will focus on the effect of green tea on oral leukoplakia, histologically defined alterations of oral mucosa, as well as on various surrogate endpoint biomarkers.

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