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PRINCIPAL INVESTIGATOR: Iman A. Hakim, M.D., Ph.D., MPH

CONTRACTING ORGANIZATION: UNIVERSITY OF ARIZONA
TUCSON, AZ 85722-3308

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Iman A. Hakim, M.D., Ph.D., MPH

Email: ihakim@azcc.arizona.edu

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14. ABSTRACT

Our overall goal is to develop a safe and feasible model for the chemoprevention of a wide range of tobacco-related diseases. Our immediate goal, that was addressed over a 5-year study period, is to determine the effects of high tea consumption on biological markers of oxidative stress that mediate lung cancer risk. We completed c a 6-month randomized, controlled, double-blinded chemopreventive trial in a group of COPD subjects who are being randomized to green or black tea preparations or a control intervention (matching placebo). Levels of 8-hydroxydeoxyguanosine and 8-F2-isoprostanes are used to measure DNA and lipid damage respectively. Changes in biomarkers of oxidative damage were measured in urine and blood. The study protocol was approved by all parties in September 2003. Recruitment and screening of participants for eligibility criteria started in October 2003. Total recruitment was completed in December 2007. A total of 158 participants (86 females and 72 males) completed the study. All laboratory analyses, data entry and quality control measures were completed. Our preliminary data show that although women have a significant lower pack/year smoking history, they have a significantly higher DNA damage as measured by urinary 8-OhDG. Females smokers on green tea have a 35% significant decrease in DNA damage and a significant decrease in blood cholesterol and LDL levels. This data confirm our previous findings related to beneficial effect of green tea on DNA damage among smokers.

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INTRODUCTION

Preventive strategies require identification of cancer-susceptible individuals resulting from combinations of carcinogen exposure and lack of protective factors. Oxidative reactions have been implicated as important modulators of human health and can play a role in both disease prevention and disease development. A large number of studies have demonstrated an increased oxidant burden and consequently increased markers of oxidative stress in the airspaces, breath, blood, and urine of smokers and of patients with chronic obstructive pulmonary disease (COPD) [1,2]. Changes in dietary habits with the intake of more cancer-chemopreventive agents appear to be a practical approach for cancer prevention in subjects with increased oxidative stress as is the case of subjects with COPD and ≥ 25 pack/year of smoking history.

The present study investigated the ability of regular green and /or black tea consumption to decrease oxidative stress during the context of a randomized, controlled, double blinded, dietary intervention trial. Levels of 8-hydroxydeoxyguanosine (8-OHdG) is used to measure DNA damage and levels of 8-F2 isoprostanes **(**8-epi-PGF2) and ethanes are used to measure lipid damage**.** The ability to modulate biomarkers of oxidative stress will have a potential impact on health promotion and prevention of chronic diseases such as lung cancer and cardiovascular diseases among people at risk of increased oxidative stress, such as smokers, workers in nuclear weapons plants, Gulf War veterans, and US Marines.

BODY

Task 1. <u>Preparation, protocol development and analysis of tea extracts and placebo</u> (QC/QA) for tea polyphenols (Months 1-7)

<u>a)</u> <u>All interviewers will be trained in the specific protocols and administration of questionnaires</u>.
All the interviewers were trained in the specific protocols and administration of study questionnaires.

<u>b)</u> <u>Obtain Human subjects approval</u>
A detailed study protocol was developed, revised and approved by both USAMRAA and the University of Arizona ‘s human Subject committees. Consent and HIPPA forms were developed and approved by both USAMRAA and the University of Arizona ‘s human Subject committees. Final approval obtained on September 30, 2003. Advertisement, screening, and recruitment for the study started in October 2003.

<u>c)</u> <u>Preparation of recruitment materials (brochures, Advertisements.)</u>
Immediately after obtaining final approval study, advertisement of the study started and brochures were distributed to COPD clinics. The study cups (12 oz cups with study logo) , timers (3 minutes timer with study logo and clinic phone number), and bags (tote bags with study logo to carry forms and sealed urinary cups) were ordered and received.

<u>d)</u> <u>Obtain the study green tea , black tea and matching placebo.</u>
The study agents (green tea, black tea, and matching placebo) were ordered and received in July. All the tea bags have blank labels and were received in large barrels labeled as A, B, and C. The code is kept in a sealed envelop to be used by the medical director if needed (as in a health related emergency). Randomization is done separately under the direction of Dr. Harris the Epidemiologist and all Subjects’ tea packages are sent un-identified (only with subject ID) to the study clinic to ensure complete blindness of both staff and subjects

Task 2. **Recruitment/ eligibility, Run-In & baseline assessment of oxidative stress (Completed)**

a) Potentially eligible subjects will be recruited beginning in month 5 of the study
Recruitment of eligible subjects started in month 5 of the study and was completed in December 2007. During the last 3 years of recruitment, we found that we have to screen more than 1500 subjects to be able to find 300 eligible subjects and 40 % of the subjects are more likely to drop-out during run-in (before randomization) due to various reasons . Therefore, we expended the recruitment time to be able to have 150 subjects completing the study.

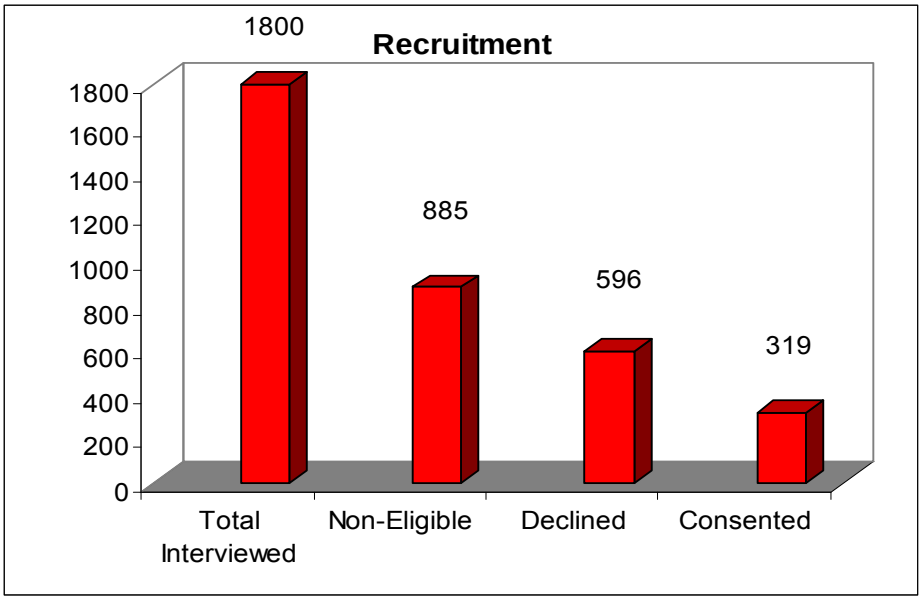
A total of 1800 subjects were interviewed by phone for eligibility criteria. Subjects were not eligible because of age, pack/year of cigarettes, medications, had cancer, or currently enrolled in another study. The main reasons for refusing to participate in the study were not willing to give up tea, cannot drink much tea, or the long duration of the study.

c) Eligible subjects will complete 1-month run-in period during which they will consume the placebo beverage and complete all baseline questionnaires.

Recruitment was successfully completed by the end of July 2007 and subjects were enrolled and followed up for the 6-month intervention period. All eligible subjects completed all baseline questionnaires and started the run-in period. Each enrolled participant, received 1-month of placebo tea bags, study teacup, a 3-minute timer, the monthly diary and health monitoring forms, and sterile urine cups. Subjects were contacted biweekly to ensure and encourage adherence and to monitor any adverse event.

d & e) Subjects who complete the run-in period will provide blood, urine and exhaled breath condensate (EBC) samples for biomarker analysis. Subjects will be asked to provide buccal cells and induced sputum samples for storage.

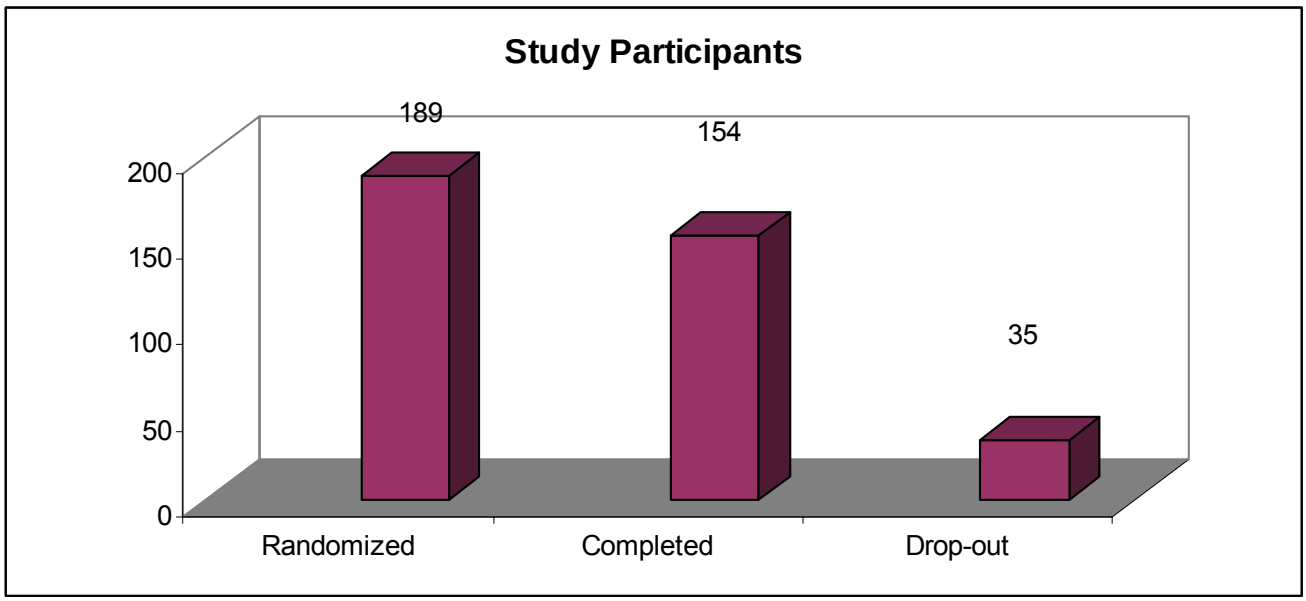
Subjects who completed the run-in period provided blood, urine and exhaled breath condensate (EBC) samples for biomarker analysis. All subjects (100%) provided buccal cells and 65% of the subjects provided induced sputum samples for storage. By the end of December (2007), 319 participants signed the consent form and were screened for confirmation of COPD eligibility criteria (spirometry for lung function tests)

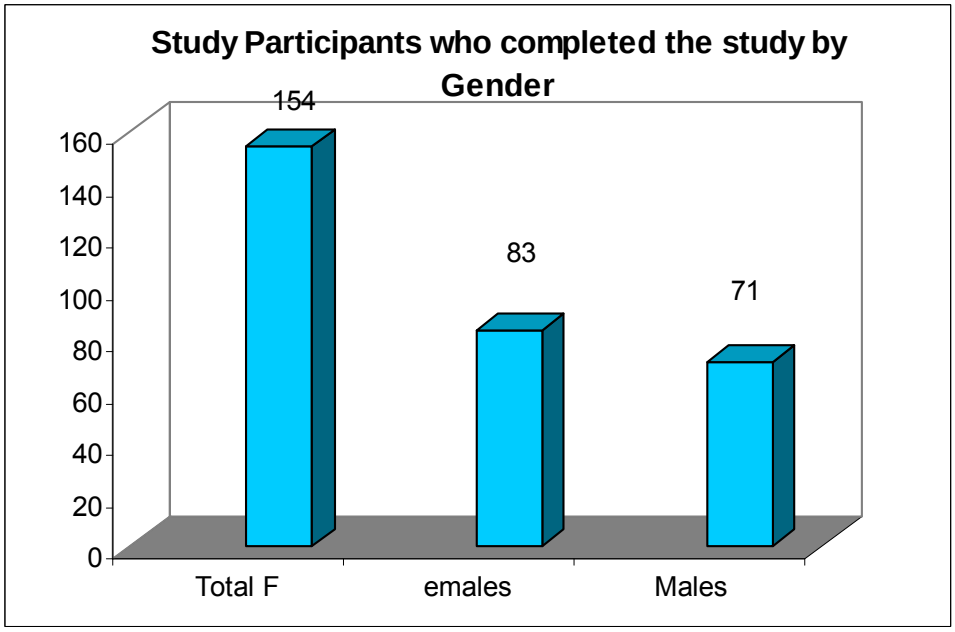


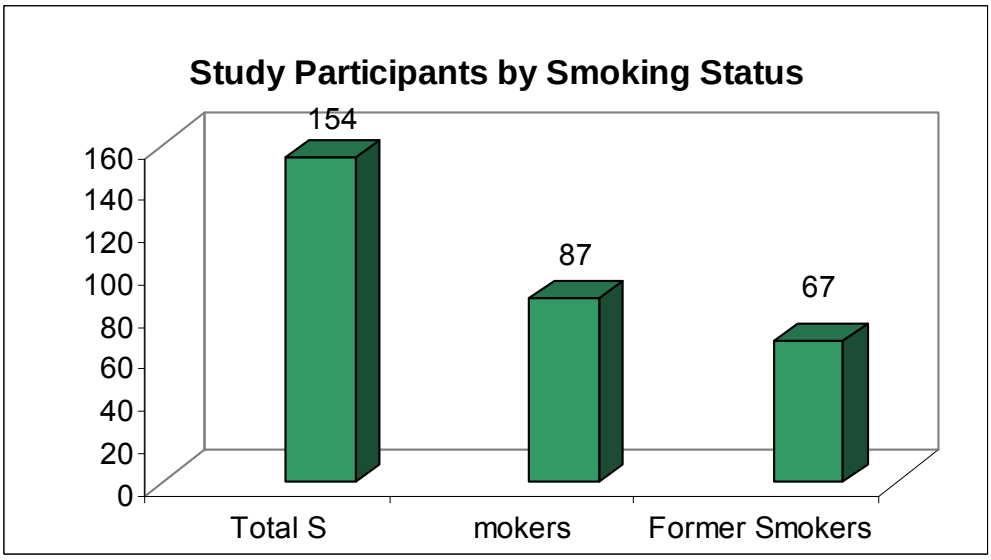
Task 3. **<u>Intervention, Follow-up & Exit focus groups</u> to study the effect of tea consumption on DNA (8-OHdG) and lipid (8-epi-PGF2) damage in blood, urine, and EBC (Completed).**

a) <u>Randomize eligible COPD chronic and former smokers into one of three interventions: black tea, green tea or placebo for 6 months</u>.

A total of 154 subjects have completed the study and they have been randomized to 1 of the 3 arms of the study. The demographics of the study population (subjects who completed the study) are shown below.







b) To maintain high adherence to the study intervention including collection of blood, urinary, and EBC samples through the 6-month intervention period and 1-month follow-up period.

Study participants were contacted biweekly by phone to ensure adherence. Subjects completed a tea and smoking diary in which they reported their daily intake of tea (amount and time) and the number of cigarettes smoked each day. They also completed a health monitoring form in which they report any change in medication use, any health-related event, or any perceived adverse event. Data is being entered for the last cohort.

c) To identify issues affecting recruitment and retention of chronic and former smokers with COPD in a lung cancer prevention trial.

d) To determine whether subjects will continue to consume tea regularly after the end of the intervention.

Exit and satisfaction questionnaire were collected from all participants that completed the study. Data has been entered into the computer database. The statistical analysis is ongoing and the final results will be available at the end of the year when all data will be analyzed. The most common causes of drop-out are too much fluid and time commitment.

Task 4. Laboratory analyses and data entry (Completed)

a) Quality control assurances of laboratory methods

We have completed all the validation and quality control measures for the biomarkers of oxidative stress. Our quality control and validation data show that the urinary biomarkers of oxidative DNA and lipid damage are stable even when left at room temperature for 3 consecutive days.

b) & c) Urine & Blood Oxidative Stress biomarkers' analyses and quality control

Laboratory analyses of urinary and blood biomarkers of oxidative damage started on time as scheduled. All laboratory analyses undergo quality control/quality assurance measures before being sent for data entry.

Measurements of 8-hydroxy-2'-deoxyguanosine (8OHdG) in human urine and lymphocyte DNA by high performance liquid chromatography-electrospray tandem mass spectrometry

A method for quantification of 8OHdG in human urine by HPLC-tandem mass spectrometry has been implemented and validated in Dr. Chow's laboratory. The analysis is performed on a ThermoFinnigan TSQ Quantum triple quadrupole mass spectrometric system in tandem with a Surveyor LC system. The urine sample (50 μl) is diluted 1:1 with water and injected onto the HPLC system. HPLC separation is achieved with a BDS Hypersil C_{18} column (150 x 2.1 mm, 5μ) and a gradient mobile phase. The gradient starts at 1% methanol and 99% 10 mM ammonium formate and is increased linearly to 80% methanol and 20% ammonium formate by 15 minutes. The system is re-equilibrated with 1% methanol and 99% ammonium formate for 5 minutes before the next injection. The flow rate is 0.2 ml/min. 8OHdG (from precursor ion *m/z* 284 to product ion *m/z* 168) and 2'-deoxyguanosine (from precursor ion *m/z* 268 to product ion *m/z* 152) are detected with multiple reaction monitoring (MRM) in the positive ion mode utilizing electrospray ionization. Linear calibration curves have been established from 0.3 to 30 ng/ml (1-100 nM). The within-day and between-day coefficient of variation of the assay is less than 10%. 8OHdG is found to be stable in urine when stored at room temperature for 72 hours.

Dr. Chow's laboratory has also tested various DNA digestion procedures for measurement of 8OHdG levels in DNA to maximize release of normal nucleosides and 8OHdG and minimize oxidation of 2'-deoxyguanosine and DNA during sample preparation and handling. Dr. Chow's lab had optimized the procedures for isolating DNA from blood lymphocytes for 8OHdG measurements. All laboratory analyses are completed.

Measurements of 8-isoprostaglandin $F_{2\alpha}$ (8-iso-$PGF_{2\alpha}$) in human urine by high performance liquid chromatography-electrospray tandem mass spectrometry

A method for quantification of 8-isoprostaglandin $F_{2\alpha}$ in human urine by HPLC-tandem mass spectrometry has been implemented and validated in Dr. Chow's laboratory. The analysis is performed on a ThermoFinnigan TSQ Quantum triple quadrupole mass spectrometric system in tandem with a Surveyor LC system. The urine sample (1 ml) is extracted with a solid phase extraction procedure before injecting onto the HPLC system. Isotope labeled 8-isoprostaglandin $F_{2\alpha}$-D4 (8-iso-$PGF_{2\alpha}$-D4) is used as the internal standard. HPLC separation is achieved with a BDS Hypersil C_{18} column (150 x 2.1 mm, 5μ) and a gradient mobile phase consisting of 2 mM ammonium acetate (A) and 5:95 methanol:acetonitrile (B). The gradient starts at 20% B and increases linearly to 35% B by 27 minutes. The system is re-equilibrated with 20% B for 10 minutes prior to the next injection. Flow rate is 0.2 ml/min. 8-iso-$PGF_{2\alpha}$ (from precursor ion *m/z* 353 to product ion *m/z* 193), 8-iso-$PGF_{2\alpha}$-D4 (from precursor ion *m/z* 357 to product ion *m/z* 197), and prostaglandin $F_{2\alpha}$ (from precursor ion *m/z* 357 to product ion *m/z* 197) are detected with multiple reaction monitoring (MRM) in the positive ion mode utilizing electrospray ionization. Linear calibration curves have been established from 20 to 5000 pg/ml. The within-day and between-day coefficient of variation of the assay is less than 10%.

We have completed the urinary analyses of biomarkers of oxidative DNA damage (8-OHdG) and lipid damage (8-F2 isoprostanes), and creatinine for all subjects who completed the 6-month study. Biomarkers were measured at baseline, month 3 (mid-intervention), and month 6 (end of intervention). The data entry and the quality control procedure for all ***8-iso-$PGF_{2\alpha}$*** data have been completed. Statistical analyses is ongoing.

d) Oxidative stress biomarkers in exhaled breath condensate

Measurements of 8-isoprostaglandin $F_{2\alpha}$ (8-iso-$PGF_{2\alpha}$) in human exhaled breath condensate

A commercially available enzyme immuno assay kit (Cayman Chemical, Catalog No. 516351) which has shown a limit of quantification of 4 pg/ml for measurements of 8-iso- $PGF_{2\alpha}$ levels in the breath condensate samples. We have established a reproducible 8-iso-$PGF_{2\alpha}$ calibration curve from 3.91 to 500 pg/ml. The between-day and within-day coefficient of variance is less than 11%. A number of baseline exhaled breath condensate samples were tested using this procedure and the 8-iso-$PGF_{2\alpha}$ levels were found to be at the low end of the calibration curve (2-4 pg/ml). With a concentrating factor of 10, we were able to observe absorbance readings comparable to those observed with concentrations of 20-30 pg/ml.

Antioxidant levels in blood

We have completed the analyses of blood antioxidants for all participants who completed the study. Data entry and statistical analyses are ongoing

Antioxidant Enzymes Data

DOD	CAT (nmol/min/g Hb)	GPx (nmol/min/g Hb)	SOD (U/g Hb)
Mean	597,248.98	26,585.17	5,366.94
Standard Error	10,804.46	549.13	108.93
Median	578,872.52	24,729.50	5,041.89
Standard Deviation	174,551.37	9,286.64	1,842.10
Sample Variance	30,468,179,876.48	86,241,725.00	3,393,341.62
Range	1,258,631.88	57,483.55	10,365.73
Minimum	209,148.44	9,711.17	1,643.06
Maximum	1,467,780.32	67,194.73	12,008.79
Sum	155,881,983.83	7,603,357.63	1,534,944.93
Count	261.00	286.00	286.00
Confidence Level(95.0%)	21,275.37	1,080.87	214.40

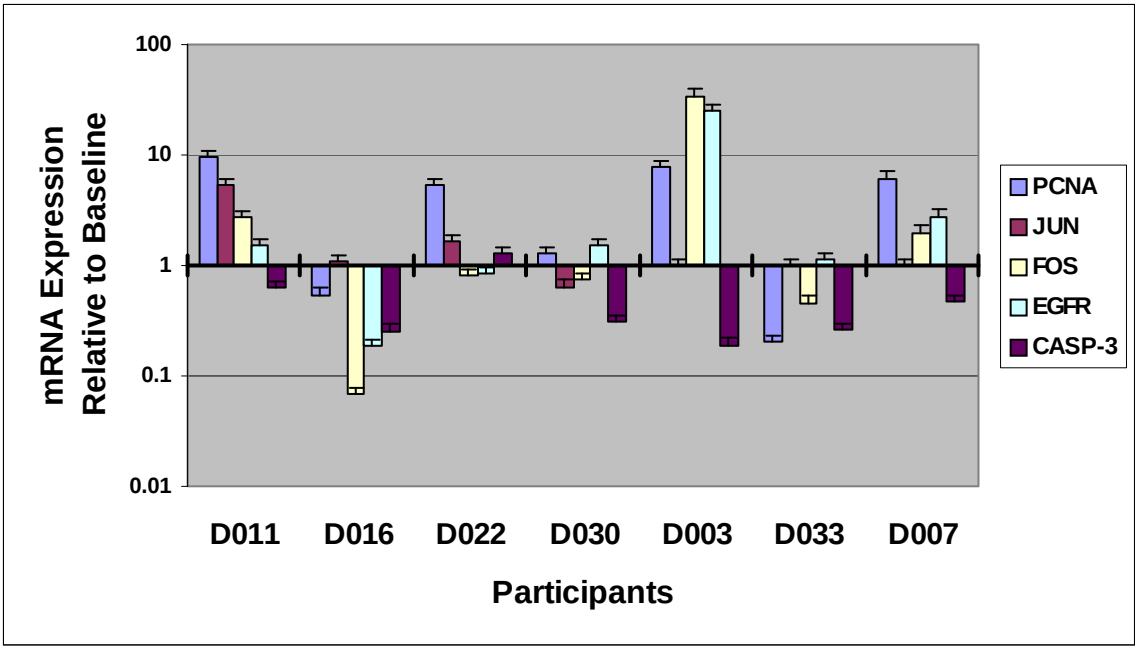
Measurements of Nitric Oxide (NO) and Ethane in Exhaled Air

Measurements of NO and ethane in exhaled air is being done at baseline and month 6 (end of intervention). All laboratory analyses undergo quality control/quality assurance measures before being sent for data entry. This preliminary summary represents the data that had been entered into our database. Summary of the overall entered data is presented in the Table below. Statistical analyses is ongoing

Visit	Nitric Oxide (ppb) Range	Carbon Monoxide (ppm) Range	Ethane (ppb) Range
Baseline	5.2 – 77.0	0 – 52.0	0.4 – 14.8
Month 6	8.1 – 85.0	0.3 – 32	0.5 – 21.4

Development of the methodology for RNA extraction from sputum

This is an innovative addition to the study. The plan was to store the sputum samples for future analyses. However**,** we have successfully developed the methodology for RNA extraction from sputum with a significant yield of RNA. Preliminary testing of gene expression of proliferation and apoptosis are successful. Preliminary data are presented below. Detailed statistical analyses will be completed later as it needs a biostatistician with expertise in genetic analyses.



KEY RESEARCH ACCOMPLISHMENTS

- Development and approval of the study protocol
- Development and approval of all study forms and questionnaires
- Successful recruitment and screening
- Successful enrollment in the study
- Successful collection of biological samples (blood, urine, EBC, buccal and sputum samples**)**
- Validation and quality control of all laboratory methods
- Complete all laboratory analyses of biological samples.
- One hundred and fifty four participants successfully completed the study.
- Successful development of methodology for RNA extraction from sputum
- Successful measurements and analyses of RNA gene expression in sputum samples
- Complete all data entry and data management
- Data analyses and manuscript preparation ongoing

REPORTABLE OUTCOMES

a) Results:

Data management was completed and data analyses started and will be ongoing for the next 6 months. Comprehensive data analyses for all other biomarkers will be completed in the following months prior to manuscripts' preparation and submission.

Preliminary results for urinary DNA damage (8-OHdG) are presented in the tables (1-3). The preliminary data show that women who smoke are at a higher risk of DNA damage compared to men smokers and thus might be at higher risk of developing lung cancer. Our data show that female smokers on green tea benefited the most from drinking green tea and showed a 35% significant decrease in DNA damage.

Table (1) Characteristics of the Study Population

	Males	Females	P
Age (years)*	64.5 ± 8.2	61.6 ± 9.2	NS
Smoking status			
Current/ former	22/38	25/36	NS
Packyear*	55.3 ± 26.0	42.6 ± 18.0	0.002
Urinary 8-OHdG*	3.19 ± 1.96	4.65 ± 2.62	0.0008

* Mean ± SD

Table (2) Characteristics of the Study Population by Group

	Green tea	Black Tea	Control	P
Females	54%	55%	52%	NS
Smokers	57%	58%	55%	NS
Age*	60.7± 8.8	60.3±9.5	60.0±9.7	NS
Packyear*	46.7± 22.5	44.6±20.4	42.3±19.5	NS

* Mean ± SD

Table (3) Mean change of 8-OHdG by Gender & Smoking Status

	Urinary 8-OHdG					
	Green Tea			Black Tea		
	Mean change	(95% CI)	P	Mean change	(95% CI)	P
Males						
Total	0.5	(-0.2, 0.4)	0.38	-0.3	(-0.4, 0.2)	0.42
Smokers	0.8	(-0.2, 0.6)	0.27	0.05	(-0.4, 0.3)	0.81
X-Smokers	0.1	(-0.4, 0.5)	0.92	-1.32 **(-37%)**	(-0.9, 0.2)	0.17
Females						
Total	-0.8 **(-19%)**	(-2.0, 0.0)	**0.05**	1.5	(-0.2, 0.5)	0.30
Smokers	1.8 **(-35%)**	(-1.1, -0.1)	**0.01**	1.5	(-0.3, 0.6)	0.60
X-Smokers	0.7	(-0.3, 0.5)	0.56	1.7	(-0.2, 0.6)	0.61

Preliminary results for lipid profiles are presented in table (4). The preliminary data show that women who were drinking green tea has a significant decrease in cholesterol and LDL levels compared to men.

Table (4) Mean changes in Lipid Profiles by Gender

	Green Tea			Black Tea		
	Mean change	(95% CI)	P	Mean change	(95% CI)	P
Males						
Cholesterol	-11.1	(-16.4, 18.8)	0.89	5.9	(-5.9, 29.6)	0.19
LDL	-1.6	(-0.22, 0.28)	0.79	-7.3	(-0.35, 0.17)	0.48
HDL	2.1	(-0.08, 0.16)	0.52	1.2	(-0.07, 0.18)	0.40
Triglycerides	12.2	(-0.32, 0.31)	0.97	-21.0	(-0.31, 0.33)	0.96
Females						
Cholesterol	-18.7	(-35.3, -2.3)	**0.03**	-6.3	(-21.4, 10.7)	0.51
LDL	- 8.9	(-0.4, 0.0)	**0.05**	-6.6	(-0.36, 0.06)	0.16
HDL	10.1	(-0.01, 0.24)	**0.07**	1.1	(-0.11, 0.14)	0.82
Triglycerides	-20.2	(-0.33, 0.09)	0.26	6.4	(-0.18, 0.23)	0.82

b) Abstracts:

Hakim IA, Chow H-H S, Harris RB, Garland L, Janine Eisnpahr, and Robbins R. A Phase II b Tea chemoprevention trial to study the effects of high tea consumption on smoking-related oxidative stress and gene expression. The 3rd Annual AACR International conference, Frontier in Cancer Prevention Research", Seattle, Washington, October 16-20, 2004.

Hakim IA, Chow H-H S, Harris R, Garland L, Rodney S, Robbins R. A Chemoprevention Trial to Study the Effects of High Tea Consumption on Smoking-Related Oxidative Stress: An Update. Peer Reviewed Medical Research Program Investigators Meeting. San Juan; Puerto Rico. May 1-5, 2006

CONCLUSIONS

During the last 5 years of the study, we were able to reach a large number of potential participants. We interviewed (initial screening) 1800 subjects and randomized 189 eligible subjects in the study. Identification of eligible participants was a challenge, however, we were successful in reaching a large pool of potential subjects. We have accomplished our goal and sample size by having 154 subjects completing the study. Laboratory analyses including all quality control procedures and data entry were completed in December 2008. The data analyses just started and our preliminary data show that women who smoke are at a higher risk of DNA damage compared to men smokers. Smokers females on green tea benefited the most from drinking green tea and showed a 35% significant decrease in DNA damage. Comprehensive data analysis is going to determine the effect of green and black tea on all the others biomarkers related to lipid damage and oxidative stress.

Because tea is one of the most popular beverages consumed worldwide, the relationship between tea consumption and human cancer incidence is an important concern. Tea can be easily consumed with one's ordinary meals making compliance and adherence to dietary intervention more likely to succeed. Thus, the role of tea drinking as a potential inhibitor of carcinogenesis merits careful evaluation. We believe that a program of nutritional intervention with realistic dietary modifications that are effective, safe, and acceptable should be the cornerstone of any cancer prevention strategy.

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APPENDICES

Abstracts:

1. ***Hakim IA,*** *Chow H-H S, Harris RB, Garland L, Janine Eisnpahr, and Robbins R. A Phase II b Tea chemoprevention trial to study the effects of high tea consumption on smoking-related oxidative stress and gene expression. The 3rd Annual AACR International conference, Frontier in Cancer Prevention Research", Seattle, Washington, October 16-20, 2004.*

BACKGROUND: Oxidative reactions have been implicated as important modulators of human health and can play a role in both disease prevention and disease development. A large number of studies have demonstrated an increased oxidant burden and consequently increased markers of oxidative stress in the airspaces, breath, blood, and urine of smokers and of patients with chronic obstructive pulmonary disease (COPD). The overall goal of this study is to develop a safe and feasible clinical research approach that will serve as a model for the chemoprevention of a wide range of tobacco-related diseases. Our immediate goal, that is addressed over a 4-year study period, is to determine the effects of high tea consumption on biological markers of oxidative stress that mediate lung cancer risk, including, 8-hydroxydeoxyguanosine **(**8-OhDG), F2-isoprostanes (8-epi-PGF2), ethanes, and nitric oxide. We will also determine if high tea consumption can modulate the genes involved in the carcinogenic process in damaged bronchoepithelial cells. **METHODS**: We are conducting a 6-month randomized, controlled, double-blinded chemopreventive trial in a group of COPD subjects (FEV1 ≤ 85%) with 25 or more pack-years of smoking history. The participants are stratified on smoking status (current or former) and gender, and are being randomized to green or black tea preparations or a control intervention (matching placebo). Levels of 8-OHdG will be used to measure DNA damage and levels of 8-epi-PGF2 and ethanes will be used to measure lipid damage. Changes in biomarkers of oxidative damage will be measured in urine, blood and exhaled breath condensate. Changes in the gene expression of biomarkers of proliferation (EGFR, PCNA, JUN, FOS, Ki-67) and apoptosis (bcl-2, caspase 3) in induced sputum will be assessed. **RESULTS:** The study protocol was approved by all parties in September 2003. Recruitment and screening of participants for eligibility criteria started in October 2003. By the end of August, 79 participants signed the consent form and were screened for eligibility criteria (spirometry for lung function tests). Eight subjects with FEV1 > 85% of the standard were excluded from the study and the remaining eligible subjects were enrolled in the study. To date, 17 subjects have completed the study and 33 have been randomized and are completing the 6-month study. **CONCLUSION**: We expect that adherence to a regular pattern of tea is feasible and quantifiable among this high risk population.

2. ***Hakim IA**, Chow H-H S, Harris R, Garland L, Rodney S, Robbins R. A Chemoprevention Trial to Study the Effects of High Tea Consumption on Smoking-Related Oxidative Stress: An Update. Peer Reviewed Medical Research Program Investigators Meeting. San Juan; Puerto Rico. May 1-5, 2006*

BACKGROUND/PURPOSE: Oxidative reactions have been implicated as important modulators of human health and can play a role in both disease prevention and disease development. A large number of studies have demonstrated an increased oxidant burden and consequently increased markers of oxidative stress in the airspaces, breath, blood, and urine of smokers and of patients with chronic obstructive pulmonary disease (COPD). The overall goal of this study is to develop a safe and feasible clinical research approach that will serve as a model for the chemoprevention of a wide range of tobacco-related diseases. Our immediate goal, that is addressed over a 4-year study period, is to determine the effects of high tea consumption on biological markers of oxidative stress that mediate lung cancer risk, including, 8-hydroxydeoxyguanosine **(**8-OhDG), F2-isoprostanes (8-epi-PGF2), ethanes, and nitric oxide. We will also determine if high tea consumption can modulate the genes involved in the carcinogenic process in damaged bronchoepithelial cells**. METHODS:** We are conducting a 6-month randomized, controlled, double-blinded chemopreventive trial in a group of COPD subjects (FEV1 ≤ 85% of the standard) with 25 or more pack-years of smoking history. The participants are stratified on smoking status (current or former) and gender, and are being randomized to green or black tea preparations or a control intervention (matching placebo). Levels of 8-OHdG are used to measure DNA damage and levels of 8-epi-PGF2 and ethanes are used to measure lipid damage. Changes in biomarkers of oxidative damage are measured in urine, blood and exhaled breath condensate. Changes in the gene expression of biomarkers of proliferation (EGFR, JUN, FOS, Ki-67) and apoptosis (caspase 3) in induced sputum are being assessed. **RESULTS:** The study protocol was approved by all parties in September 2003. Recruitment and screening of participants for eligibility criteria started in October 2003. To date, 110 subjects have been enrolled in the study and 80 have already completed the study. We have completed the urinary analyses of biomarkers of oxidative and the RNA gene expression and modulation for the first group of subjects who completed the 6-month study. Biomarkers of oxidative stress were measured at baseline, month 3 (mid-intervention), and month 6 (end of intervention) while gene expression was measured at baseline and end of the study. Summary of the overall entered data will be presented. **CONCLUSION:** We expect that adherence to a regular pattern of tea is feasible and quantifiable among this high risk population**.**

RESEARCH PERSONNEL : Include Paid and non-paid personnel involved with the study throughout the 6 years and reflects the changes in staff throughout the 6 years.

Name and Position	Research Role (PI, Co-PI, Collaborator, Sub-I, Data Manager, Study Coordinator, etc.)
Iman Hakim, MD, PhD, MPH	PI
Robin Harris; PhD; MPH	Epidemiology
H-H Sherry Chow, PhD	Director of Analytical laboratory
Linda Garland, MD	Medical director
Steve Rodney	Data management
Richard Robbins, MD	Collaborator
Michael Habib, MD	Sub investigator
Mary Lurie	Recruitment/Interviewer
Kyla Ballesteros	Laboratory Coordinator
Lisa Quale	Study Coordinator
Renee Reichard	Laboratory Technician
Tom Vincent	Coordinator
Gina Blackwell	Coordinator
Osmara Molina	Study Coordinator
Catherine Celeya/Cordova	Laboratory Technician
Carmine Martinez	Study Coordinator
Kristin Schmidt	Laboratory Coordinator
Justin Kowal	Recruitment/Interviewer
Laura Goodman	Laboratory Technician
Amber Strebing	Recruitment & monitoring
Maribel Tobar	Recruitment & monitoring
Lydia Mikail	Laboratory Technician