PROTOCOL TITLE:

Biological Pathways in Stress Reactivity and Nicotine Addiction among African American and White Smokers

PRINCIPAL INVESTIGATORS:

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1.0 Objectives / Specific Aims

This study will examine racial differences in stress responses between African American and white male smokers as part of the Translational Research Center in Lung Cancer Disparities (TRACER).

- **Aim 1: Examine racial differences in acute physiological stress responses between African American and white male smokers using a laboratory model. The association between acute physiological stress response and tobacco-related behaviors will also be evaluated.**

  Hypotheses: African American smokers will be more likely than white smokers to have a blunted cortisol response to an acute laboratory stressor. African American smokers will also have significantly greater cigarette craving and be more likely to smoke following exposure to an acute physiological stressor compared to white smokers following an acute laboratory stressor.

- **Aim 2: Evaluate racial differences in daily cortisol patterns between African American and white male smokers**.

  Hypotheses: African American smokers will be more likely than white smokers to have blunted daily diurnal cortisol slopes. Compared to white smokers, African American smokers will have lower absolute cortisol values during the waking day.

- **Aim 3: Identify factors that are important to acute and daily stress responses among African American and white male smokers by examining acute and daily cortisol patterns based on exposure to chronic stressors.**

  Hypotheses: Acute and daily cortisol responses will be associated with chronic stressors such that men who have greater exposure to SES, structural, and psychosocial stressors will have blunted acute and daily cortisol patterns. The relationship between acute and daily cortisol patterns, nicotine metabolism, and nicotine dependence will also be explored among African American and white male smokers as part of this aim.

- **Aim 4: Examine social and clinical factors associated with study enrollment and retention**.

  Hypotheses: Those with greater comorbidities and social issues (i.e. lower SES, education, social support) will have lower rates of study enrollment and retention compared to those with fewer comorbidities and social issues.

2.0 Background

Cigarette smoking is a leading cause of morbidity and mortality from lung cancer among adults in the US; smoking cessation is the best preventive strategy for reducing lung cancer risk and addressing racial disparities in outcomes. Despite this, many smokers are unwilling or unable to make a quit attempt, and among those who try to quit smoking, utilization of evidence-based treatment is poor, especially in racial minorities. Stress relief, and the avoidance of adverse psychological and physiological reactions, are among the primary reasons for smoking initiation, maintenance, and relapse in all populations. Stress responses have both psychological and physiological consequences that are important to smoking behaviors and cessation outcomes, and racial differences in physiological stress responses have not been examined between African American and white smokers.

Physiological stress responses are among the multilevel determinants of cancer health disparities that need to be examined within the context of other SES, psychosocial, and structural factors. This study will be the first to examine racial differences in stress responses between African American and white male smokers at increased risk for lung cancer morbidity and mortality by measuring acute cortisol responses to a laboratory stressor and evaluating racial disparities in daily cortisol patterns among these men in their

neighborhoods and communities. Together with contextualized data on acute and chronic stress responses based on SES, psychosocial, and structural factors, this study will generate novel empirical data that can be leveraged into precision strategies for lung cancer prevention through smoking cessation and other intervention approaches.

3.0 Study Setting

This study will be conducted at MUSC Health. Participants will be identified using electronic health records (EHR) and will participate in laboratory visits at the Research Nexus Laboratory in the Clinical Sciences Building at MUSC to measure acute stress responses. All baseline and follow-up assessments will be conducted by telephone or online using a redcap weblink. *This study is being conducted as part of the Translational Research Center in Lung Cancer Disparities (TRACER) that includes: MUSC, the Massey Cancer Center at the Virginia Commonwealth University (VCU), and the City of Hope Comprehensive Cancer Center (COH).*

4.0 Inclusion and Exclusion Criteria/ Study Population

Primary care patients at MUSC Health will be identified using EHR and those who meet inclusion criteria will be contacted by telephone to complete a screening interview and baseline assessment that measures socioeconomic stressors and other variables. To be eligible to participate in the study, the following criteria will be applied.

Inclusion Criteria

- African American and white males
- Between the ages of 18-75 years old
- Smoke at least 5 to 10 cigarettes per day

Exclusion Criteria

- Smokers who have a serious cognitive disorder or psychiatric illness
- Have a personal history of lung cancer
- Personal history of usage of illicit drug and alcohol abuse
- Enrollment in a smoking cessation treatment program during the past 6-months
- Current use of a nicotine replacement therapy
- Have any positive response on the Mini International Neuropsychiatric Interview (MINI) screener

5.0 Number of Subjects

The study will enroll 100 African American (n=50) and white (n=50) male smokers.

6.0 Recruitment Methods

Two recruitment strategies will be used to identify smokers to participate in the study. First, the initial recruitment strategy will be identifying patients by searching EHRs using smoking status and ICD-10 diagnosis codes (e.g., smoking status, psychiatric illness, cognitive disorders) to identify smokers who are potentially eligible to participate in the study. Additionally, text algorithms will be applied to chart notes to help supplement the EHR search because smoking behaviors may be under-reported. Following identification and application of exclusion criteria to the EHR, a patient list of individuals who are identified as current smokers and meeting other eligibility criteria will be created and provided to the investigative team. These patients will be mailed an invitation letter that describes the purpose of the study and the procedures involved in participation. The letter will also include a local number and email address to respond if they are not interested in being contacted for study participation. Those who do not opt out of

participation will be contacted for the baseline and screening telephone interview. The South Carolina Translational Research Center (SCTR) will conduct the EHR search and generate patient lists. Second, a community-based approach in which smokers self-refer for participation from newspaper, radio advertisements, and social media will be used to recruit study participants. For individuals who self-refer for study participation, the screening interview and baseline survey will be completed to determine eligibility and complete enrollment procedures. Marketing materials will be developed using a evidence-based community engagement strategies. Project investigators will consult with members of the community engagement core to develop materials that are tailored and appropriate. Once these materials are developed they will be submitted to the IRB for review and approval.

7.0 Consent Process

This study will use the procedures described below to obtain informed consent for study participation.

Electronic Health Record Data Abstraction. A full waiver of HIPAA Authorization is requested to generate the EHR query that will be used to establish the list of patients who are potentially eligible to participate in the study and to obtain other personal health identifiers (e.g., MRN, names, address, telephone numbers) for sending invitation letters and conducting recruitment calls. The waiver of HIPAA Authorization will also include permission to abstract clinical (e.g., co-morbidities, medications, vital signs) and socioeconomic data (e.g. insurance type, race/ethnicity), and to utilize text algorithms developed by BMIC to abstract information from clinical notes relating to smoking status. Clinical and sociodemographic data abstracted from the EHR will be included in de-identified datasets for analysis of study outcomes relating to factors (i.e. zipcode, co-morbidities) associated with smoking behaviors, stress responses, and enrollment and retention . The outcomes rendered from the EHR data that is obtained will help to inform future studies that further address disparities relating to lung cancer risk.

All PHI data will be collected at the time of screening. No further data collection occurs on these participants following screening and EHR abstraction under the full waiver. Further data collection for participants who choose to enroll will be covered under the partial waiver for the baseline interview, and the signed informed consent form and HIPAA if they choose to participate further in the study.

Screening Interview and Baseline Survey. A waiver of written informed consent/partial waiver of signature element is requested to complete the screening interview and baseline survey. Patients will be read a statement of study information before obtaining verbal consent. Once verbal consent is achieved, the screening interview will be completed by telephone. Baseline surveys will be completed according to the participant's preference either by telephone or through self-administration via a redcap survey weblink.

Laboratory Visits and Biorepository Biospecimen Donation. This study will involve laboratory visits for biospecimen collection and stress assessments, followed by at-home saliva collections at two follow-up timepoints (see section 8.0 for detailed methods). Written informed consent will be obtained at the Pre-Challenge laboratory visit (1st laboratory visit) prior to the initiation of any study activities. The written informed consent will include permission to collect biospecimens (e.g. urine, saliva), completion of a social stress test, perform at-home saliva collection and smoking assessment following the laboratory visits, and to use data on exposure to chronic, psychosocial, and structural stressors obtained during the study. The informed consent will also include an option for participants to donate biospecimens to the TRACER Biorepository located at VCU. If consent for biospecimens and data is provided, this information will be processed and stored using the procedures described in study design and methods. Provision of consent for biospecimens and data among individuals who do not meet final eligibility determination will be recorded in the data management system.

To obtain written informed consent, the research assistant (RA) will meet participants in a private room to review the informed consent form (ICF) and answer questions about the procedures involved in participating in the study, describe the voluntary nature of study participation, and discuss procedures for withdrawing. Following the informed consent discussion, participants will immediately be asked to sign the ICF, the RA will also sign and date the ICF, and a copy of the signed ICF will be provided to participants. The original ICF will be filed in the participants study chart and stored in a locked file cabinet. The RA will also complete an informed consenting process documentation form that contains a checklist for the RA to adhere to while conducting the informed consenting process. This form will also be filed in the participant's chart.

8.0 Study Design / Methods

This research is a prospective observational study that will measure acute and chronic stress responses among African American and white smokers. Acute stress responses will be measured following exposure to a validated laboratory social stress test and chronic stress responses will be measured at 4- and 8-weeks following the laboratory stress test using at home collection. The procedures described below will be completed.

Screening Interview and Baseline Assessment. A 30-minute telephone interview will be conducted by research assistants from MUSC to screen participants for initial eligibility and those who are eligible will be invited to enroll into the study and complete a baseline survey. Participants that agree to enroll into the study, will be given the option to participate in the baseline survey over the telephone or through self-administration via a redcap survey link. The baseline survey will obtain information relating to sociodemographic characteristics and measure exposure to chronic economic stressors (e.g., low income, financial strain), psychosocial stressors (e.g., perceived stress, social isolation), exposure to structural stressors (e.g., violence, crime), and community perceptions and resources (e.g., collective efficacy). Additionally, other factors relating to smoking history, nicotine dependence, and reasons for smoking will also be measured, along with perceived risk and control over developing lung cancer, utilization of lung cancer screening, and interest in quitting smoking will also be measured during the baseline assessment. Zip code and street address will be confirmed as part of the telephone interview to mail financial incentives and send study reminders. Once participants completed the baseline survey, they will be invited to participate in the laboratory assessments described below.

Laboratory Visits. This study will involve four laboratory visits that will include a pre-challenge visit (lab visit 1) to collect biospecimens, a stress exposure visit (lab visit 2) that will involve conducting the Trier Social Stress Test (TSST), and two visits (lab visits 3 and 4) at 4- and 8-weeks after the TSST visit to collect final saliva samples. Each visit is explained in more detail below.

Pre-Challenge Visit. Following completion of the screening interview and baseline survey, participants will complete a pre-challenge visit to obtain urine samples that will be used to determine final eligibility. The pre-challenge visit will be completed at the Research Nexus Lab at MUSC. Participants will be asked to smoke as usual prior to the pre-challenge visit but to not eat or drink one hour before the session. During the Pre-Challenge Visit, screening tests will determine final eligibility, these include urine toxicology; individuals whose results indicate current substance abuse (e.g., cocaine) will be excluded (not eligible for further participation). Saliva samples will also be obtained during the pre-challenge visit to determine cortisol and cotinine levels using established salivary swab collection methods and validated thresholds. To further evaluate and confirm active smoking behaviors, a breath monitor will be used to measure levels of carbon monoxide in the blood. The MINI International Neuropsychiatric Interview (MINI) will be administered by a trained research technician to evaluate mental status and psychiatry exclusion criteria. Participants will be excluded from participation in the laboratory stress visit if they meet the exclusion criteria (e.g., positive responses on the MINI, or positive toxicology result). Participants eligible following pre-challenge visit will be scheduled for the stress exposure visit described below.

TSST Stress Exposure Visit. The stress exposure visit will be completed within 72-hours of completing the pre-challenge visit. As with the pre-challenge visit, participants will be instructed to smoke as usual, but not to eat or drink during 1-hour prior to stress exposure visit. The number and brand of cigarettes smoked prior to the TSST will be recorded. Participants will be asked to smoke a single cigarette of their usual brand prior to initiating the stress test to standardize the time since last smoking. Fifteen minutes later, baseline clinical and subjective measures will be obtained. Clinical measurements will include heart rate and blood pressure using a standardized monitor, and saliva collection using a Salivette. Following collection of these baseline measures, participants will be escorted back to their room to start the TSST. The TSST is a standardized psychological stress challenge that has been validated and used extensively in research studies; a meta-analysis supports that it is the gold standard for evoking an HPA-axis stress response in a laboratory setting. The TSST (see **TSST Procedures and Timeline**) will be administered in

the Research Nexus Lab. Outcome measures (e.g., cortisol responses) will be collected prospectively during the stress exposure visit. Heart rate variability (HRV) and blood pressure will also be measured using standard medical equipment and procedures during the TSST. Cigarette craving and smoking behavior will be measured after the TSST has been completed. Specifically, smokers will rate their urge to smoke immediately after the last saliva sample has been taken as part of the TSST. After completing this self-report measure, they will be given the opportunity to smoke a cigarette using a pocket CReSS system that measures and records smoking topography (Puff #, Volume, IPI). Smoking behavior (smoke versus not smoke) will be recorded in the participant's study chart and stored in study's data management Redcap system.

TSST Procedures and Timeline		
TIME	**EXPERIMENTAL PROCEDURE**	**STRESS ASSESSMENT**
3:00pm	Acclimation period (15 minute duration)	Patient arrives and given relaxation tools (i.e. spa video, magazine, nature scenes)
Baseline (immediately following acclimation period)	Stress measurements	Take cortisol, BP, HR, and assess SUD
Immediately before TSST (12 minutes following baseline)	Stress measurements	Take cortisol, BP, HR, assess SUD, and explain the task and give participant paper to write notes
3:33- 3:44	TSST	
5	Interview task	
5	Arithmetic task	
Post TSST stress assessments (12 minute increments)		
Post TSST (2 minutes following TSST)	Post-TSST Assessment	Cortisol, BP, HR, SUD
12 mins post TSST	Post-TSST Assessment	Cortisol, BP, HR, SUD
24 mins post TSST	Final Post-TSST Assessment	Cortisol, BP, HR, SUD

Post-Stress Test Saliva Collection. At the end of the TSST, participants will be given saliva kits (Salimetrics oral swab) to collect samples at home 4- and 8-weeks after the stress exposure visit**.** Self-reported data on perceived stress and life experiences during the past month will also be obtained using validated instruments. Standard collection procedures (e.g., avoid eating/drinking/brushing teeth within 1-hour of sample collection; do not engage in vigorous physical activity; no alcohol within 12-hours of sample collection) will be provided to participants as an instructional sheet. Saliva will be collected at fixed intervals (upon wakening, 30-minutes after wakening, 4:00 pm, and 6:30 pm). The final 6:30pm saliva collection will occur at the Nexus Laboratory and participants will be instructed to return their other collected samples from home at this time. Collection dates will be based on the date the stress exposure visit was completed and sample collection procedures will be explained verbally and in writing at the end of the stress exposure visit. Participants will be given an instructional sheet for how to perform the sample collections. Participants will also be given a collection log to record the date and time of each collection, the number and brand of cigarettes smoked on the collection day, quit attempts, the amount of stress experienced during the past month, and positive and negative life experiences during the past month. Participants will be contacted the night before the collection date by telephone, email, and text to remind them about the sample collection date, collection and storage procedures, and the information that should be recorded in the sample collection log. Saliva samples will be collected at 4- and 8-weeks after the TSST to be consistent with the collection timepoints used in previous research with African American and white treatment-seeking smokers.

9.0 Data Collection and Storage.

9.1 Specimen Collection and Banking for Future Use

As described previously, biospecimens (urine, saliva) will be collected after obtaining written informed consent at the Pre-Challenge Visit and Stress Exposure Visit and these biospecimens will be stored at the Research Nexus Laboratory at MUSC to complete procedures related to determining eligibility (Pre-Challenge Visit) and evaluating stress responses (Stress Exposure Visit). Saliva samples will also be obtained at home following the Stress Exposure Visit; these biospecimens will be processed and stored at the Research Nexus Laboratory until statistical analyses have been completed. Once statistical analyses have been completed, biospecimens will be stored in the Nexus Laboratory until the end of the project period. All biospecimens will be stored in the Nexus Laboratory using a unique numeric identification

number to link samples with participants. Further, all biospecimens will be coded with a numeric identifier to link test results with (e.g., changes in cortisol, cotinine levels) with subject data obtained during the screening and baseline telephone interview and clinical data (e.g., co-morbidities) abstracted from electronic health records. All linked subject data will be de-identified and stored in a secured, password-protected database at MUSC using their identification number.

Participants also have the option to provide written informed consent for biospecimens to be included in the TRACER Biorepository. The TRACER biorepository is located at VCU; biospecimens from this study will be shipped to the TRACER biorepository using standard shipping procedures. The Project Manager for this study will work with Nexus staff to ensure that samples are sent to the TRACER Biorepository and to document receipt of biospecimens. Additionally, de-identified participant-level data will be uploaded into a centralized database at the TRACER Biorepository. Access to biospecimens, test results, and linked data will be limited to study investigators, research staff within the scope of their roles (e.g., Project Manager), and investigators in the TRACER Biorepository.

9.2 Specimen/Banking for Future Use

The TRACER Biorepository will bank biospecimens in the facility located at VCU and de-identified participant data (e.g., race, age, zip code, self-reported data on income, education, etc., and census level information on social determinants of health) that are linked with these biospecimens will be stored on a centralized, secure database in the TRACER Biorepository. This information will be stored in the TRACER Biorepository until the end of the project period. At the end of TRACER, samples and de-identified participant data will be destroyed. Participants can request that their biospecimen/data be withdrawn from the TRACER Biorepository by submitting a Withdrawal of Informed Consent for Use of Specimens to the study project manager. Once this request is received, biospecimens and data will be destroyed or removed from the TRACER Biorepository.

Biospecimens and participant data collected as part of this study will be used in future research that is conducted by TRACER investigators and may include genomic/genetic analyses. A standard process will be used to release biospecimens and data to TRACER investigators. Specifically, TRACER investigators will submit a written Request for Samples. The Request for Samples will describe the type of samples needed, the purpose of the study, and documentation of IRB approval or exempt from human subjects designation. The Request for Samples will be reviewed by investigators in the TRACER Biorepository with a recommendation to approve or deny the request for samples. The TRACER Executive Council will make final decisions about approving or denying the request for samples. Only de-identified biospecimens and/or data will be released to TRACER investigators. A Material Transfer Agreement/Data Transfer Agreement (MTA/DAT) will be established by TRACER institutions prior to inter-institutional data sharing and transfer of biospecimens and data.

10. Data Management

10.1 Statistical Considerations and Sample Size Justification by Specific Aim. This section describes the sample size and power estimates for the study and also describes the statistical analyses that will be completed to address the specific aims for the study.

Aim 1: Examine racial differences in acute physiological stress responses between AA/Black and white male smokers using a laboratory model. The association between acute physiological stress response and tobacco-related behaviors will also be evaluated. *Hypotheses:* AA/Black smokers will be more likely than white smokers to have a blunted cortisol response to an acute laboratory stressor. AA/Black smokers will have lower absolute cortisol values during the acute stressor compared to white smokers. AA/Black smokers will have significantly greater cigarette craving and be more likely to smoke following exposure to an acute physiological stressor compared to white smokers following an acute laboratory stressor.

Sample Size and Power Calculations: This aim will examine racial disparities in cortisol responses to an acute stressor among AA/Black and white male smokers in a laboratory setting. Changes in cortisol levels will be the primary outcome variable and will be measured multiple times during the course of the TSST. A sample size of 50 AA/Black (Mean DCS=0.20, SD=0.20) and 50 white (Mean DCS=0.35, SD=0.30) males achieves 84% power to detect a difference of 0.15µg/dL between the 2 groups with a significance level of 0.05 using a two-sided two-sample t-test. This sample size would also provide 80% power to detect a difference of 0.14µg/dL in absolute cortisol value between AA/Black (mean=0.17, SD=0.19) and white (mean=0.31, SD=0.30) males with a significance level of 0.05 using a two-sided two-sample *t*-test.

Statistical Analysis: First, descriptive statistics will be generated to characterize enrollment outcomes using clinical data (e.g., co-morbidities, age, etc) obtained from the EHR during the recruitment process. Statistical analyses will compare enrollees vs. decliners in terms of race, clinical factors, and smoking history to identify variables that should be controlled for in the analysis of stress reactivity and smoking behaviors.

To examine the effects of race on acute stress responses to the TSST laboratory stressor the response area under the curve (rAUC) for cortisol will be calculated using trapezoidal approximation with the samples obtained at time zero (T0) to the last measurement (T24m). Analysis will adjust for baseline cortisol levels by subtracting the rectangle defined the baseline sample and total observation time (T24min) from the total area under the curve, thus, it is possible to have negative rAUC values if cortisol levels decline below baseline.[91] A generalized linear regression model will be conducted with rAUC as the dependent variable and race (AA/Black, white) as the between-subjects factor. The effects of nicotine metabolism rates, nicotine dependence, and cigarettes smoked per day will be examined in analysis of covariance that include rAUC as the dependent variable and race as the between-subjects variable. The extent to which cortisol changes are blunted will be determined by calculating the difference between the first and last cortisol assessment during the TSST.[90] Cortisol values that do not change over time or those that are closer to zero will be considered blunted.[30] Similar regression models will be used to test for racial differences in cortisol levels at each timepoint following exposure to an acute laboratory stressor, the proportion who smoke after the TSST, and scores for cigarette craving. A random effect will be added to the statistical models to allow for differences in participants recruited from different cites (MUSC and VCU). Models will be adjusted for known and potential confounders (e.g., nicotine metabolism rates).

Aim 2. Evaluate racial differences in *daily* cortisol patterns between AA/Black and white male smokers. *Hypotheses:* AA/Black smokers will be more likely than white smokers to have blunted daily DCS. Compared to white smokers, AA/Black smokers will have lower absolute cortisol values during the waking day. This aim will examine racial disparities in cortisol responses between AA/Black and white male smokers in their neighborhood and community settings at 4- and 8-weeks after the TSST has been completed. Changes in cortisol levels will be the primary outcome variable and will be measured at multiple critical timepoints during the waking day.

Sample Size and Power Calculations: A sample size of 50 AA/Black (Mean DCS=0.12, SD=0.15) and 50 white (Mean DCS=0.26, SD=0.30) males achieves 84% power to detect a difference of 0.14µg/dL between the 2 groups with a significance level of 0.05 using a two-sided two-sample *t*-test, assuming similar differences between the groups at 4 and at 8 weeks of follow-up. This sample size would also provide 83% power to detect a difference of 0.13 µg/dL in absolute cortisol value between AA/Black (mean=0.35, SD=0.19) and white (mean=0.48, SD=0.35) males with a significance level of 0.05 using a two-sided two-sample *t*-test.

Statistical Analysis: A similar approach to Specific Aim 1will be used to examine racial differences in daily cortisol levels. Racial differences in mean cortisol levels at 4 weeks will be modeled by a generalized linear model similar to Specific Aim 1 with the addition of a repeated statement to account for the multiple measures takes for each participant throughout the day and a random intercept to allow for individual differences. The same will be done at 8 weeks post TSST. Models will include race and time-of-day as fixed effects and the interaction between them. Model-based contrasts will be used to conduct group

comparisons based on the fitted model. Blunted cortisol DCS will be defined similarly as in Specific Aim 1. Generalized linear models will be used to examine racial differences in blunted DCS at each post-TSST assessment. Similar models will examine the relationship between blunted DCS (at 4 and 8 weeks post-TSST) and baseline measures of smoking behavior (e.g., smoking history, nicotine dependence, nicotine metabolism rates), and stressors (chronic SES stressors, psychosocial stressors, and structural stressors).

Aim 3: Identify factors that are important to acute and daily stress responses among AA/Black and white male smokers by examining acute and daily cortisol patterns based on exposure to chronic stressors. *Hypotheses:* Acute and daily cortisol responses will be associated with chronic stressors such that men who have greater exposure to SES, structural, and psychosocial stressors will have blunted acute and daily cortisol patterns. The relationship between acute and daily cortisol patterns, nicotine metabolism, and nicotine dependence will also be explored.

Sample Size and Power Calculations: It is hypothesized that men who have a higher nicotine addiction (daily versus non-daily or higher FTND score versus lower) will have higher cortisol levels. A sample size of 100 males achieves 82% power to detect a ratio of 1.25 (25% higher cortisol levels among the highly addicted) when the ratio under the null hypothesis is 1 and the coefficient of variation on the original scale is 0.4 with a significance level of 0.05 using a two-sided two-sample *t*-test.

Statistical Analysis: Similar to Specific Aims 1 and 2, Specific Aim 3 multivariable generalized linear regression models will be used to identify factors that are important to acute (baseline) and daily (4 and 8 weeks post-TSST) stress responses (cortisol patterns). Independent associations between blunted cortisol responses and race, smoking behaviors, nicotine dependence at baseline, SES, and psychosocial and structural stressors will be evaluated.

10.2 Data Storage and Privacy Protection. The data management system for the study will be designed to achieve the major elements of the study including: (1) determination of study eligibility, (2) monitoring recruitment and accrual, (3) generation of study materials, and (4) storage of data from telephone interviews and laboratory assessments. All data collected as part of this study will be stored on a MUSC-based secure password-protected network server. Moreover, access to the server will be monitored by the PI and limited to pertinent study personnel.

We will take extensive precautions to protect the privacy of the participants. Personal health identifiers (e.g., name, address) will not be used to identify participants in study databases or on laboratory materials. We will use a confidential subject identification number to identify all participant data in research databases and on study documents (e.g., baseline telephone assessments), and laboratory materials (i.e. clinic flow sheets, biospecimen tubes). Cortisol saliva kits and other biospecimen collection tubes will be labeled with the study identification number. A key containing each participant's name, study identification number, and telephone number will be kept on a secure, password-protected database on a MUSC network server until study procedures have been completed and the data have been checked for completeness and accuracy. At that time, this identifying information will be destroyed. Thus, we will retain no identifying information in the study data files. Moreover, all computerized study databases for clinical and questionnaire data will be housed on a secure, password-protected network server. All personal contact information will be kept in a database that will be housed separately from the database containing questionnaire data. There will be limited access to study files and study databases throughout the duration of the study; only pertinent study staff will have access to study information.

All MUSC data including biospecimens, social determinants of health, clinical data, and sociodemographic data will be de-identified before being sent to the TRACER VCU centralized database that will be the repository for all TRACER-affiliated projects. The centralized database will ensure data harmonization, uniformity, quality control and privacy. VCU TRACER investigators will be responsible for abiding to HIPPA

compliance, and compliance with federal and local regulations. Data will be collected in REDCap instruments to facilitate data integration into the centralized database.

Handling Incidental Findings and Adverse Effects. In the event of any unexpected events related to incidental findings, safety concerns, and adverse effects, the PIs and/or study staff will contact the MUSC and VCU IRBs to discuss appropriate action.

11.0 Risks to Subjects

Potential Risks. There is a slight risk that participants may experience adverse psychological reactions such as anxiety or stress as a result of discussing smoking behaviors, personal health behaviors, and social and psychological stressors during the **screening and baseline telephone interview**. We will minimize this risk by ensuring that telephone interviewers are trained to detect increased anxiety and how to intervene accordingly. Also, participants will be given rest breaks as needed throughout the telephone interviews, and if warranted, any referrals for follow-up psychological care will be made as needed.

Participants may also potentially experience a loss of privacy as a result of providing information about their past medical history and personal health behaviors. However, we are using items that have been tested and validated and are commonly used in clinical settings and it is likely that participants would have been exposed to similar types of questions through their routine medical care. Therefore, we do not anticipate that participants will feel that their privacy has been violated. Also, we will obtain permission from the IRB to obtain informed consent over the telephone prior to initiating any study activity. Eligible participants will be required to verbally agree to participate in the study during the initial scripted telephone call after the invitations letters have been mailed out. Only participants that express interest in participating will be administered the baseline telephone interview. Written informed consent will be obtained prior to conducting the laboratory assessments. We will also give participants the option to opt out of being contacted if they do not want to be contacted about participating in the study. Participants will be informed that their participation is voluntary and that they can withdraw their participation at any time without any adverse consequences.

There are also minimum risks associated with the **laboratory assessments.** Participants may experience some mild physical discomfort from the biospecimen collection (e.g., saliva swab) at the laboratory visits. Participants will also be exposed to a psychological stressor (giving a speech and arithmetic task) to induce a stress response. It is likely that the stress test will produce a certain amount of stress and may cause an increase in blood pressure and increased heart rate. We do not anticipate that the laboratory stressor would provoke any significant adverse reactions that are not usually encountered on a day-to-day basis. The Research Assistant conducting the in-person laboratory assessment will be highly trained in administering the laboratory stress test. As part of this training, she/he will be trained to determine if participants are experiencing any adverse reactions and will terminate the assessment immediately. If warranted, referrals for follow-up psychological care will be made as needed. Our study team includes a Clinical Psychological (Carla Kmett Danielson, PhD) who has extensive experience administering the Trier Social Stress Test (TSST), monitoring responses, and making referrals to psychological services as needed.

Protection Against Risks. We will take every precaution to protect the privacy and autonomy of participants and to assure that the consent is truly informed. As we described above, verbal informed consent will be obtained for screening, study enrollment and completion of the baseline and follow-up assessments. As part of the verbal informed consent process, the purpose of the study will be explained orally and the right to refuse to answer all or some of the questions will be emphasized. It will also be stressed that they can start or stop their participation at any time throughout the duration of the study. Participants who refuse to complete the laboratory assessment or those who withdraw from the study will be thanked and all further contacts will be terminated.

We will obtain written informed consent prior to conducting the laboratory assessments. As part of obtaining written informed consent, we will review the purpose, procedures, duration of participation, and their rights for being a research participant. Participants will be given the opportunity to ask questions before signing the informed consent form and proceeding with the laboratory assessments.

To protect the privacy of participants that provide permission to donate biospecimens for future use, biospecimens will be given a coded number and stored based on that code. This coded number can be linked back to limited personal health information and other clinical and social data pertaining to them. Any information learned from this study in which you might be identified will be confidential and disclosed only with their permission. Every effort will be made to keep any information collected about the study participants confidential. Study investigators will adhere to compliance and ethical conduct of research procedures to protect the privacy and welfare of participants.

We will also take extensive precautions to protect the privacy of the participants. Personal health identifiers (e.g., name, address) will not be used to identify participants in study databases or on laboratory materials. We will use a confidential subject identification number to identify all participant data in research databases and on study documents (e.g., baseline telephone interview), and laboratory materials (i.e. biospecimen tubes). Cortisol saliva kits and plasma tubes will be labeled with the study identification number. A key containing each participant's name, study identification number, and telephone number will be kept in a secured location until study procedures have been completed and the data have been checked for completeness and accuracy. At that time, this identifying information will be destroyed. Thus, we will retain no identifying information in the study data files. Moreover, all computerized study databases for questionnaire data will be housed on a secure, password-protected network server. All personal contact information will be kept in a database that will be housed separately from the database containing questionnaire data. There will be limited access to study files and study databases throughout the duration of the study; only pertinent study staff will have access to study information.

12.0 Potential Benefits to Subjects or Others

There are no direct clinical benefits to participating in this study. The potential risks associated with study participation are minimal and the information that is learned through this research is likely to have significant benefits. Racial disparities exist as it relates to smoking cessation outcomes among AA/Black smokers. Observational studies have shown that psychosocial stress is a potential intervention target for smoking cessation treatment programs. Therefore, evaluating racial differences in biological pathways involved in stress reactivity and nicotine addiction would provide empirical evidence necessary for designing precision medicine strategies for tobacco control interventions that address the smoking patterns and cessation needs among racial minorities and individuals from other medically underserved groups.

Importance of Knowledge to be Gained. Cigarette smoking continues to be a leading cause of morbidity and mortality from lung cancer and other chronic diseases among adults in the US. Data from national studies continue to expose the disparities that exist among AA/Black smokers as it relates to poorer smoking cessation rates, unfavorable socioeconomic factors, and greater levels of perceived stress that impact tobacco use. This study will be the first to examine the complex ways in which psychological, behavioral, and biological pathways contribute to NMR and smoking behaviors among AA/Black and white smokers. Our research will move beyond black-white comparisons in smoking behaviors and cessation rates by taking a biobehavioral results of this research will provide greater insights into behavioral, psychological, and biological mechanisms that contribute to racial disparities in smokers and will generate novel empirical data that can be translated into targeted interventions to reduce racial disparities in smoking behaviors and improve tobacco control outcomes.

Additionally, this study will support the collection, analyses, and storage of biospecimens (saliva, urine) from a racially and ethnically diverse sample of smokers. Biospecimens from a racially diverse sample in addition to collected data on social determinants of health and other biological and clinical data will help

with enhancing our understanding of factors related to smoking disparities and enable investigators to conduct studies that address multilevel determinants of cancer risk and outcomes.

13.0 Sharing of Results with Subjects

The results of this study will not be shared with participants or their physicians. Research results will not be placed in participant's medical charts.