1.a. Full Title: The association between morning salivary cortisol, cardiovascular risk factors, and carotid atherosclerosis: The ARIC Carotid MRI Study

b. Abbreviated Title (Length 26 characters): Salivary cortisol and atherosclerosis

2. Writing Group: Sherita Hill Golden, MD, MHS; Gary Wand, MD; Josef Coresh, MD, PhD; Saurabh Malhotra, MD, MPH; Bruce Wasserman, MD

I, the first author, confirm that all the coauthors have given their approval for this manuscript proposal. [please confirm with your initials electronically or in writing]

First author: Sherita Hill Golden

Corresponding/senior author (if different from first author correspondence will be sent to both the first author & the corresponding author): Same

3. Timeline: To begin immediately once salivary cortisol data have been entered at the Coordinating Center and forwarded to Dr. Golden

4. Rationale:

   Obesity, type 2 diabetes, and cardiovascular disease continue to be major public health burdens and type 2 diabetes is rising in epidemic proportions. Thus, identification of novel risk factors for these diseases is important in guiding the development of preventive interventions. Psychological stress, particularly depression, has been shown to be a risk factor for the development of both type 2 diabetes (1-5) and cardiovascular disease (6-9) but the mechanism remains unclear. Neuroendocrine changes induced by these psychological factors, specifically activation of the hypothalamic-pituitary-adrenal (HPA) axis, might provide a unifying explanation. Dysregulation of the HPA axis has been documented in individuals with various forms of psychological stress, including depression (10-13), anxiety(14), history of childhood abuse (15) (16,17)and/or adult trauma (17)and post-traumatic stress disorder(18). Neuroendocrine dysfunction has also been documented in obesity (19-22)and diabetes(23). Cross-sectional studies assessing the relation of HPA axis activity, by measurement of salivary cortisol, and cardiovascular risk factors have demonstrated a positive association between morning cortisol and cardiovascular risk factors, such as blood pressure, lipids, glucose, and intimal-medial thickness reactivity (24-26), although one study found no association between salivary cortisol response to awakening and the metabolic syndrome(27). With the exception of one study of 509 South Asian individuals, these studies were limited by small sample sizes and by being performed primarily in Caucasians in Europe.
Our Salivary Cortisol Ancillary Study to the ARIC Carotid MRI Study, provides a unique opportunity to assess the association of salivary cortisol, as a proxy of hypothalamic-pituitary-adrenal axis activity, with multiple cardiovascular risk factors and carotid atherosclerosis that have been assessed thoroughly, uniformly and in a large, bi-racial cohort.

5. Main Hypothesis/Study Questions:

1. Is morning salivary cortisol associated with prevalent coronary heart disease (CHD) and prevalent type 2 diabetes mellitus?

2. Among individuals without prevalent type 2 diabetes, is morning salivary cortisol associated with cardiovascular risk factors, including adiposity (waist circumference, body-mass index), fasting plasma glucose, fasting plasma insulin, dyslipidemia (triglycerides, HDL-cholesterol), and systolic and diastolic blood pressure?

3. Is morning salivary cortisol associated with carotid wall thickness and the presence and characteristics of carotid plaque, assessed by carotid MRI?

6. Design and analysis (study design, inclusion/exclusion, outcome and other variables of interest with specific reference to the time of their collection, summary of data analysis, and any anticipated methodologic limitations or challenges if present).

Study design: Cross-sectional analysis

Data Variables: Age, race/ethnicity, gender, ARIC site, prevalent CHD, prevalent type 2 diabetes mellitus, prevalent hypertension, fasting plasma glucose, fasting plasma insulin, lipids (total cholesterol, HDL-cholesterol, LDL-cholesterol, triglycerides), systolic blood pressure, diastolic blood pressure, smoking status, dietary intake, physical activity, medications (cholesterol lowering, anti-hypertensive therapy), adiposity (waist circumference, body-mass index), carotid wall thickness (mean and maximum), presence of carotid plaque, lipid core size (as proxy of plaque vulnerability), plaque contrast enhancement (as proxy of inflammation and neovascularization), and morning salivary cortisol.

Brief analysis plan and methods

Table 1. Outline of analysis plan.

<table>
<thead>
<tr>
<th>Markers of HPA Axis Activation</th>
<th>Consequences of Neuroendocrine Activation</th>
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<tbody>
<tr>
<td>8 am salivary cortisol</td>
<td>Visceral obesity</td>
</tr>
<tr>
<td></td>
<td>Hypertension</td>
</tr>
<tr>
<td></td>
<td>Glucose intolerance/insulin resistance</td>
</tr>
<tr>
<td></td>
<td>Dyslipidemia</td>
</tr>
<tr>
<td></td>
<td>Atherosclerosis</td>
</tr>
</tbody>
</table>

Confounders/Mechanisms: Diet, physical activity, smoking status, obesity
Correlation analyses and linear regression models will be used to determine the correlation between morning salivary cortisol and the following measures: (a) adiposity, assessed by waist circumference and body-mass index, (b) fasting glucose, (c) fasting insulin, (d) lipid parameters (total cholesterol, triglycerides, HDL-cholesterol, LDL-cholesterol), and (e) blood pressure.

Correlation analyses and linear regression models will be used to determine the correlation between morning salivary cortisol and the following measures of carotid atherosclerosis: (a) mean and maximum carotid wall thickness, (b) presence of carotid plaque, (c) lipid core size (as a proxy of plaque vulnerability), and (d) plaque contrast enhancement (as a proxy of inflammation and neovascularization).

Individuals with diabetes will be excluded since treatment of glycemia may confound the association between salivary cortisol and fasting glucose. Subsidiary analyses will be conducted in individuals with and without prevalent cardiovascular disease to determine if the associations differ in these two populations. In multivariate analyses, we will be able to adjust for potential confounders of the association between morning cortisol and metabolic risk factors using data already collected in the study on demographics, diet, physical activity, smoking status, and medication use.

Conclusion:

ARIC provides a unique opportunity to examine the association of salivary cortisol, as a proxy of hypothalamic-pituitary-adrenal axis activity, with multiple cardiovascular risk factors and carotid atherosclerosis that have been assessed thoroughly, uniformly and in a bi-racial cohort. To our knowledge, there are no large epidemiological studies that have examined these hypotheses previously. The addition of this hormonal assessment has placed ARIC in a position to expand the field of cardiovascular epidemiology to include less traditional hormonal measurements.

7.a. Will the data be used for non-CVD analysis in this manuscript? __ Yes __x__ No

b. If Yes, is the author aware that the file ICTDER02 must be used to exclude persons with a value RES_OTH = “CVD Research” for non-DNA analysis, and for DNA analysis RES_DÑA = “CVD Research” would be used? ____ Yes ____ No

(This file ICTDER02 has been distributed to ARIC PIs, and contains the responses to consent updates related to stored sample use for research.)

8.a. Will the DNA data be used in this manuscript? ____ Yes __x__ No
8.b. If yes, is the author aware that either DNA data distributed by the Coordinating Center must be used, or the file ICTDER02 must be used to exclude those with value RES_DNA = “No use/storage DNA”? ____ Yes ____ No  N/A

9. The lead author of this manuscript proposal has reviewed the list of existing ARIC Study manuscript proposals and has found no overlap between this proposal and previously approved manuscript proposals either published or still in active status. ARIC Investigators have access to the publications lists under the Study Members Area of the web site at: http://www.cscc.unc.edu/ARIC/search.php
__x__ Yes _______ No

10. What are the most related manuscript proposals in ARIC (authors are encouraged to contact lead authors of these proposals for comments on the new proposal or collaboration)?

There are none.

11. a. Is this manuscript proposal associated with any ARIC ancillary studies or use any ancillary study data? __x__ Yes ____ No

11.b. If yes, is the proposal
__X__ A. primarily the result of an ancillary study (list number* _________)
___ B. primarily based on ARIC data with ancillary data playing a minor role (usually control variables; list number(s)* __________ __________ __________)

*ancillary studies are listed by number at http://www.cscc.unc.edu/aric/forms/

12. Manuscript preparation is expected to be completed in one to three years. If a manuscript is not submitted for ARIC review at the end of the 3-years from the date of the approval, the manuscript proposal will expire.


22. Bjorntorp P, Rosmond R: Obesity and cortisol [In Process Citation]. *Nutrition* 16:924-936, 2000


