ARIC Manuscript Proposal # 1767

PC Reviewed: 3/8/11	**Status: A**	**Priority: 2**
SC Reviewed: _____	**Status: _____**	**Priority: ____**

1. **a. Full Title**: Association of blood lactate with carotid atherosclerosis: The Atherosclerosis Risk in Communities Carotid MRI Study
   b. Abbreviated Title (Length 26 characters): Lactate & atherosclerosis

2. **Writing Group**:
   Ghanshyam Palamaner Subash Shantha
   Brad Astor
   Bruce Wasserman
   Josef Coresh
   Fred Brancati
   Ron Hoogeveen
   Christie Ballantyne
   J. Hunter Young (senior)

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

First author: **Ghanshyam Palamaner Subash Shantha**
Address: Department of Epidemiology
Johns Hopkins Bloomberg School of Public Health
Welch Center for Prevention, Epidemiology, & Clinical Research
2024 E. Monument Street, Suite 2-600
Baltimore, MD 21205-2223

Phone: 443-825-2546 E-mail: gpalaman@jhsph.edu

Corresponding/senior author (must be an ARIC investigator for the proposal but can be different in the published paper; correspondence will be sent to both the first author & the corresponding author**)**:

Senior author: J. Hunter Young, MD MHS
Address: Assistant Professor of Medicine
Welch Center for Prevention, Epidemiology, & Clinical Research
Johns Hopkins School of Medicine
2024 E. Monument Street, Suite 2-600
Baltimore, MD 21205-2223

Phone: 410-502-5808 Fax: 410-955-0476
E-mail: jhyoung@jhmi.edu

3. Timeline: Manuscript to be completed by May 27th 2011.

4. Rationale:

Atherosclerosis is a complex inflammatory process that is characterized by the formation of raised plaques resulting in the narrowing of the lumen in a number of arterial

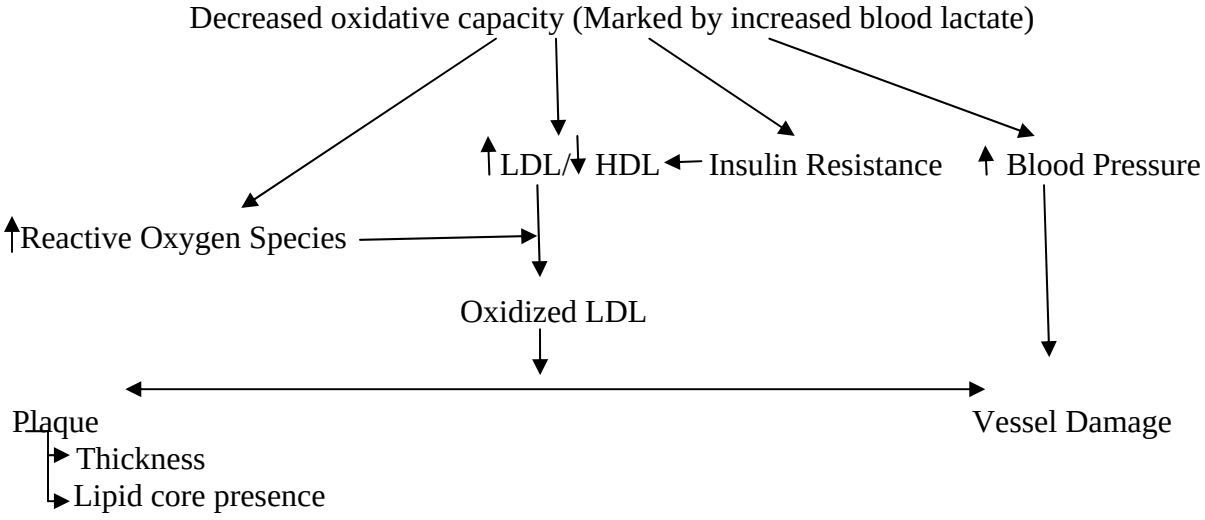
beds.(1) Oxidized LDL plays a central role in plague formation. Macrophages preferentially take up oxidized LDL becoming foam cells, which contribute to the formation of fatty streaks and the plaque's lipid core. So-called vulnerable or unstable plaques (UP) characteristically have a thin fibrous cap and a prominent lipid core.(2) Oxidized LDL enhances the release of cytokines, inhibits nitric oxide production in the arterial wall, increases the expression of adherence molecules, and impairs endothelial function.(3) Therefore, oxidized LDL contributes to both plague development and plague instability through multiple pathways.

Oxidative stress expressed in the form of increased production of reactive oxygen species is responsible for the increased occurrence of oxidized LDL, at least in part. ROS may contribute to atherosclerosis through other pathways as well (4). For example, reactive oxygen species are produced in the arterial endothelium, smooth muscle cells, and adventitia. In addition to promoting lipid oxidation, ROS has direct, pro-atherosclerotic effects on expression of adhesion molecules, apoptosis, and activation of metalloproteinases.(4)

Despite its importance in the pathogenesis of atherosclerosis, the source of increased oxidized LDL is poorly understood. Mitochondrial dysfunction is a potential source of ROS (5). Several lines of evidence show that increased mitochondrial uncoupling leads to increased production of ROS (5). Furthermore, those with decreased aerobic capacity demonstrate elevated levels of ROS (6). However, no population-based study has investigated the potential relationship between mitochondrial dysfunction and ROS.

Blood lactate is an indirect indicator of insufficient oxidative capacity: when oxidative capacity decreases, flux through glycolytic pathways increases and blood lactate rises.(7-10) Prior work suggests that lactate is elevated among obese, insulin resistant subjects.(11-12) Moreover, cross-sectional studies have shown that lactate is associated with blood pressure, type 2 DM.(13, 14) and with coronary atherosclerosis (15). These studies suggests that decreased oxidative capacity marked by increased blood lactate is associated with cardio-metabolic risk factors and atherosclerosis. The goal of this study is to assess the relationship between blood lactate levels and plaque thickness and plaque characteristics such as size of the lipid core in the carotid artery and carotid stenosis among ARIC participants.

Flowchart:



5. Main Hypothesis/Study Questions:

H1. It is well established that inflammation is a primary component of plaque formation. Decreased oxidative capacity may contribute to LDL oxidation via reactive oxidative species (ROS) production and subsequent LDL oxidation. In order to assess the relative role of decreased oxidative capacity, we intend to examine the association of lactate, a marker of decreased oxidative capacity and markers of inflammation with plaque thickness in the Atherosclerosis Risk in Communities Study (ARIC). The outcome will be carotid wall plaque as measured via 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 population

The study population consists of all persons participating in the ARIC Carotid MRI study (n=2066). Subjects were recruited to the Carotid MRI study based upon their intima-media thickness (IMT), as measured by B-Mode ultrasound at the most recent ARIC visit. The study consists of 1,250 participants who had an IMT value greater than the 85th percentile, and 816 individuals randomly sampled from the remainder of the IMT distribution (<85th percentile).

Lactate

We measured lactate among all Carotid MRI study participants as part of ARIC Ancillary Study # 2006.04**C**, "Assessing the association between mitochondrial dysfunction and insulin resistance via the measurement of cellular energy intermediates: The Atherosclerosis Risk in Communities Carotid MRI Study." Lactate measurements were completed in December, 2007 and available for 1964 ARIC-MRI participants.

Data Analysis

Aim 1: Association of lactate with plaque characteristics

- Outcome variable: Increased atherosclerosis is measured by
  1) Maximum lipid core area (MLA) (cm2)
  2) Total lipid core volume (TLV) (mm3)
- Independent variables: lactate log-transformed, categorized into quartiles
- Covariates
  1. Age
  2. Gender
  3. Ethnicity
  4. Field center
  5. BMI (kg/m2)
  6. Systolic Blood pressure (mmhg)
  7. Diastolic blood pressure (mmhg)
  8. Glucose (mg/dl)
  9. Smoking
  10. Triglycerides
  11. HDL
  12. LDL

13. HsCRP
14. Statins

Aim 1: Association of lactate with carotid stenosis

A potential mechanism linking atheroscelosis with lactate is low blood flow due to peripheral vascular disease as indicated by increased carotid stenosis. In order to examine this possibliliy we will perform an analysis examing the relationship of carotid stenosis with lactate.

- Outcome variable: Carotid stenosis (lumen area, rcca)
- Independent variables: lactate log-transformed, categorized into quartiles
- Covariates
    15. Age
    16. Gender
    17. Ethnicity
    18. Field center
    19. BMI (kg/m2)
    20. Systolic Blood pressure (mmhg)
    21. Diastolic blood pressure (mmhg)
    22. Glucose (mg/dl)
    23. Smoking
    24. Triglycerides
    25. HDL
    26. LDL
    27. HsCRP
    28. Statins

Statistical Analysis:

Data will be expressed as means and 95% confidence intervals. All statistical analyses will incorporate the stratified random sampling design, for estimation, testing and confidence intervals. Sampling weights will be based on the probability of being selected from each field center based upon the high IMT status of each participant.

Characteristics of subjects participating in the ARIC-MRI study will be compared across lactate quartiles.

Step 1: coding MLA and TLV as continuous variables multiple linear regression will be performed including all the covariates in the model.

Step 2: Categorize MLA and TLV. For example, the following analyses will be performed.

- Unadjusted log- binomial regression with dichotomized MLA and TLV as dependent variables and the covariates as independent variables and lactate categorized into quartiles.
- Multiple log binomial regression will be performed with MLA and TLV as dependent variables, lactate categorized according to quartiles after adjusting for covariates.

Step 3:

- Internal validation: Dividing the data into training set (70%) and validation set (30%). Develop a prediction equation with the training set and use it in the validation set and analyze discrimination and calibration in the validation set.
- Use Framingham risk score in our data and see if that predicts carotid atherosclerosis in our data.

7.a. Will the data be used for non-CVD analysis in this manuscript?

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 ICTDER03 must be used to exclude persons with a value RES_OTH = "CVD Research" for non-DNA analysis, and for DNA analysis RES_DNA = "CVD Research" would be used? ____ Yes ____ No
(This file ICTDER03 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 ICTDER03 must be used to exclude those with value RES_DNA = "No use/storage DNA"? ____ Yes ____ No

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)?

This is the first study of lactate and insulin resistance to my knowledge in the ARIC cohort.

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* 2006.04**C)**

___ 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.

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5) Rego AC, Oliveira CR. Mitochondrial dysfunction and reactive oxygen species in excitotoxicity and apoptosis: implications for the pathogenesis of neurodegenerative diseases. Neurochem Res. 2003;28(10):1563-74.

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(11) Doar JW, Wynn V, Cramp DG. Blood pyruvate and plasma glucose levels during oral and intravenous glucose tolerance tests in obese and non-obese women. Metabolism 1968 Aug;17(8):690-701.

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