ARIC Manuscript Proposal #2056

PC Reviewed: 2/12/13	**Status: A**	**Priority: 2**
SC Reviewed: _____	**Status: _____**	**Priority: ____**

1.a. Full Title:
A Medium-Term Reliability Study of the Human Serum Metabolome: the Atherosclerosis Risk in Communities (ARIC) Study

b. Abbreviated Title (Length 26 characters): Metabolomics, reliability

2. Writing Group:
Writing group members: Yan Zheng; Bing Yu; Danny Alexander; David Couper; Jennifer A. Nettleton; and Eric Boerwinkle

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

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ARIC author to be contacted if there are questions about the manuscript and the first author does not respond or cannot be located (this must be an ARIC investigator).
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3. Timeline:
We expect that the manuscript will be prepared within six months from approval of the analysis plan.

4. Rationale:
Metabolomics is the systematic study of small molecule metabolites resulting from a host of cellular and physiologic processes(1). The metabolome is dynamic and sensitive to

external stimuli and thus there may be considerable day-to-day variations that are separate from technical laboratory phenomenon or measurement error. However, the within-subject variance over time should be small compared with the between-subject variance in order to gain a reliable effect estimate from a single blood measurement, as most metabolomic studies have reported(2). It is surprising that there has been little documentation concerning the biological variance reliability associated with metabolomic biomarkers (2, 3). The proposed study is designed to investigate biological reliability of the human metabolome in fasting serum samples over a period of 4-6 week apart. Using a convenience subsample of 60 participants from the Atherosclerosis Risk in Communities (ARIC) Carotid MRI Study(4, 5), intraclass correlation coefficient (ICC)(6) for each of the detected metabolites is going to be calculated to estimate its biological reliability. This approach of reliability measurement has been successfully applied in several similar settings (7, 8).

5. Main Hypothesis:
Some parts of the human fasting serum metabolome (such as Amino Acids and Fatty Acids) are more reliable across the medium-term period than the other parts (such as Xenobiotics).

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).

1. The proposed study is designed to investigate biological reliability of the metabolome in fasting serum samples over a period of 4-6 weeks apart by using a random subsample of 60 participants from the Atherosclerosis Risk in Communities (ARIC) Carotid MRI Study. These participants will be assigned alternate IDs to ensure blinding of laboratory technicians.
2. All analyses will be done separately for each metabolite across the 60 paired samples. For pairs having missing or below the detection limit values (missing/BDL values) in either sample <50% (i.e., <30 pairs), the value of this metabolite will be analyzed as continuous variable, where the missing/BDL values assigned the lowest detected value for that metabolite in all samples. For pairs having missing/BDL values in either sample between 50-80% (i.e., between 30-48 pairs), the value of this metabolite will be analyzed as ordinal variable with three levels: 1) missing/DBL values, 2) detected values < the median; and 3) detected values $\geqslant$ the median. For pairs with missing/BDL values in either sample >80% (i.e., >48 pairs), the value of this metabolite will not be analyzed in this study.
3. Repeated-measures analysis of variance (ANOVA) will be used to calculate the mean, total variance and between-person variance for each metabolite detected. ICCs (6), which is to estimate the biological reliability of metabolite, will be

calculated to by dividing the between-person variance by the total variance (sum of between- and within-individual variances) (9).

4. ICCs will be interpreted as >0.8, almost perfect; 0.61 to 0.8, substantial; 0.41-0.6, fair; <0.4, poor to slight (10).
5. All statistical analyses will be performed in SAS version 9.2 (SAS Institute, Cary, NC). The level of statistical significance will be set at P<0.05 for two-sided testing.

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

8.c. If yes, is the author aware that the participants with RES_DNA = 'not for profit' restriction must be excluded if the data are used by a for profit group?
___ 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

There is no overlap between this proposal and any current proposals.

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)?
The present proposal complements the metabolomic HF proposal submitted by Zheng and Nettleton (MS #1847), the metabolomic CKD proposal submitted by Yu and Boerwinkle (MS #1882), the metabolomic BP proposal submitted by Zheng and Nettleton (MS #1918). All of them are authors in the proposed study.

11. a. Is this manuscript proposal associated with any ARIC ancillary studies or use any ancillary study data? Yes

11.b. If yes, is the proposal

___ A. primarily the result of an ancillary study (list number* 2008.16 **)**

___ B. primarily based on ARIC data with ancillary data playing a minor role (usually control variables; list number(s)* 2008.16 "Metabolomics & Heart Failure: A Novel Approach to Biomarker Discovery")

*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.

Yes, the lead author is aware that manuscript preparation is expected to be completed in 1-3 years, and if this expectation is not met, the manuscript proposal will expire.

References:

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4. Wasserman BA, Astor BC, Sharrett AR, Swingen C, Catellier D. MRI measurements of carotid plaque in the atherosclerosis risk in communities (ARIC) study: methods, reliability and descriptive statistics. Journal of magnetic resonance imaging : JMRI 2010;31(2):406-15.
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6. Fleiss J. The Design and Analysis of Clinical Experiments. New York, NY: John Wiley & Sons; 1986.
7. Bijari PB, Antiga L, Wasserman BA, Steinman DA. Scan-Rescan reproducibility of carotid bifurcation geometry from routine contrast-enhanced MR angiography. Journal of magnetic resonance imaging : JMRI 2011;33(2):482-9.
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