ARIC Manuscript Proposal #2704

PC Reviewed: 2/9/16	**Status: A**	**Priority: 2**
SC Reviewed: _____	**Status: _____**	**Priority: ____**

1.a. Full Title: Repeatability of measures of insulin resistance: The Atherosclerosis Risk in Communities (ARIC) Study

b. Abbreviated Title (Length 26 characters): Repeatability of IR measures

2. Writing Group:
Writing group members: Anna K Poon, Michelle L Meyer, Gerald Reaven [invited], Joshua W Knowles, Elizabeth Selvin, James S Pankow, David Couper, Christie M Ballantyne, Laura Loehr, Gerardo Heiss, others welcome

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

First author: Anna K Poon
Address:
137 E. Franklin Street, Suite 303-A
Chapel Hill, NC 27514

Phone: 312-804-1672 Fax: 919-966-9800
E-mail: apoon@email.unc.edu

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).
Name: Gerardo Heiss
Address:
137 E. Franklin Street, 4th Floor
Chapel Hill, NC 27514
Phone: 919-962-3253 Fax: 919-966-9800
E-mail: gerardo_heiss@unc.edu

3. Timeline: We will conduct the analysis and draft the manuscript within 6 months of approval

4. Rationale:

Insulin resistance (IR) is a condition of reduced tissue sensitivity to insulin-mediated glucose uptake (Reaven, 1988). Chronic IR often precedes the development of type 2 diabetes mellitus and is a risk factor for coronary artery disease (Reaven, 1991, Reaven and Laws, 1994, Abbasi et al., 2002). The prospective study of its trajectory over time may therefore be useful for risk stratification and early identification of those at increased cardiovascular risk. Measurement protocols for standard reference measures of IR and insulin sensitivity, such as the insulin suppression test and hyperinsulinemic-euglycemic clamp, tend to be time-consuming, invasive, and thus not practical in large population-based studies (DeFronzo et al., 1979, Greenfield et al., 1981). Furthermore, while surrogate measures of IR are routinely used in research, indexed for example, by the homeostatic model assessment of insulin resistance (HOMA-IR), the need for insulin determinations and the lack of standardization across insulin assays hamper its clinical utility in assessing IR (Bonora et al., 2000, Staten et al., 2010). A practical alternative measure of IR is therefore needed.

Lipoprotein metabolism is dysregulated in IR (Reaven, 1988). This dysregulation is marked by higher levels of triglycerides (TG) and lower levels of high-density lipoprotein cholesterol (HDL-C) (Laws and Reaven, 1992). As a result, both single lipid measures and lipid ratio measures have been used to estimate IR. The triglyceride-to-high-density lipoprotein cholesterol (TG/HDL-C) ratio is a simple marker of IR. A high distribution-based TG/HDL-C ratio identifies IR with a performance similar to that of fasting plasma insulin concentrations, and is better than either lipid measure alone for detecting IR and predicting future cardiovascular disease events (Salazar et al., 2013b, Salazar et al., 2013a, McLaughlin et al., 2005, McLaughlin et al., 2003, Lorenzo et al., 2015). The TG/HDL-C ratio has the advantage of ready availability of TG and HDL-C in clinical settings and epidemiologic studies. The TG and HDL-C laboratory assay methods employed in clinical and research settings are also well standardized. These properties make the TG/HDL-C ratio a promising and practical marker of IR.

The measurement properties of the TG/HDL-C ratio have implications for the assessment of its change in prospective studies and the estimation of associations. Measurement error or variability is typically related to two broad sources of error: 1) biologic (intra-individual) variability, and 2) process- or method-related variability, such as variability due to blood drawing, local processing, shipping, central laboratory handling, and assay variability. The repeatability of both fasting TG and fasting HDL-C has been previously assessed in the Atherosclerosis Risk in Communities (ARIC) study. Between-person, within-person, and method variability were estimated; reliability coefficients indicated high reliability for both markers (Chambless et al., 1992a) over a 1-2 week period. This study captured repeatability in adults aged 45-64 years. The intra-individual variability of a measure, however, may change with time due to age-related changes in metabolic regulation or comorbidities. Whether the repeatability of fasting TG and fasting HDL-C, and the TG/HDL-C ratio, is constant over time from mid-life to late-life is unknown.

Our goal is to evaluate and compare the sources of variability in markers of IR as follows: 1) evaluate and compare the 3-month, 6-month, and 12-month repeatability of the TG/HDL-C ratio with HOMA-IR at repeated examination visits (conducted by the ARIC study

and the NHLBI Family Heart Study (FHS)); and 2) evaluate and compare the repeatability of the TG/HDL-C ratio and HOMA-IR at different life epochs, i.e., mid-life and older adulthood.

5. Main Hypothesis/Study Questions:

- Aim: Quantify and compare the repeatability of the TG/HDL-C ratio and HOMA-IR, defined continuously and by distribution-based cut points, at different life epochs from mid-life to older adulthood

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: Our analysis will be conducted in the ARIC study and the NHLBI FHS. Briefly, the NHLBI FHS is a population-based study of genetic and non-genetic determinants of coronary heart disease, atherosclerosis, and cardiovascular risk factors conducted in 1994-1995. FHS participants were selected from three ongoing epidemiologic studies: the Forsyth County, North Carolina and Minneapolis, Minnesota cohorts of the ARIC study; the Framingham Heart Study in Framingham, Massachusetts; and the Health Family Tree Program in Salt Lake City, Utah (Higgins et al., 1996).

Repeatability will be evaluated in the ARIC study at repeated examination visits in 1988, 2005-2006, and 2011-2013. Repeatability will also be evaluated at 3-, 6-, and 12-months for participants in both the ARIC study (Visit 3, 1993-1995; Visit 4, 1996-1998) and the NHLBI FHS (1994-1995) at exams conducted one-year apart, as previously done by Weatherley and colleagues (Weatherley et al., 2006). A sample of repeatability studies at these visits is included at the end of this document.

Inclusion/Exclusion: Participants will be excluded if they have not fasted for 8 hours, if they are taking cholesterol-lowering medications, if they have type 2 diabetes mellitus (defined as fasting glucose ≥126 mg/dL, non-fasting glucose ≥200 mg/dL, or use of diabetes medication), or if they are missing analytes of interest (i.e., fasting TG, fasting HDL-C, fasting glucose, fasting insulin).

Outcome: The analytes of interest include fasting TG, fasting HDL-C, fasting glucose, and fasting insulin. The surrogate IR measures include: 1) the TG/HDL-C ratio, which will be calculated by dividing fasting TG concentrations (mg/dL) by fasting HDL-C concentrations (mg/dL) ($TG \div HDL$); and 2) HOMA-IR, which will be calculated by multiplying fasting glucose concentrations (mg/dL) by fasting insulin concentrations (μU/mL) and dividing by 405 $[(glucose \times insulin) \div 405]$. The TG/HDL-C ratio and HOMA-IR will be evaluated continuously and categorically. Race and gender specific receiver operator characteristic curves will be used to determine cut points corresponding to the 75^{th} percentile of fasting insulin, optimized using the Youden index. Cut points proposed in the literature will also be used (Salazar et al., 2013b, Sumner et al., 2010).

Other variables of interest: Body mass index, abdominal obesity (e.g., waist circumference, waist-to-hip ratio), race/ethnicity, gender, age, and physical activity.

Data Analysis: Summary measures will be estimated as means and standard deviations for normally distributed analytes, and as medians and interquartile ranges for skewed distributions. We will use a nested random-effects analysis of variance model to separate the variance of the measures into between-person variance (σ^2_{BP}), between-visit variance (σ^2_{BV}), and method variance (σ^2_e). The model will be as follows: $Y_{ijk} = \mu + Person_i + Visit(Person)_{j(i)} + Error_{k(ij)}$, where Y = the outcome of interest, μ = the intercept, i = 1,2,3 to the X^{th} participant, j =visit one or visit two, and k = the first or second measurement.

Repeatability will be evaluated in the ARIC study at repeat exams in 1988, 2005-2006, and 2011-2013. We will compute the intraclass correlation coefficient (ICC) by dividing the between-person variance by the total variance [$ICC = \sigma^2_{BP}/\sigma^2_{TOT} = \sigma^2_{BP}/(\sigma^2_{BP} + \sigma^2_{BV} + \sigma^2_e)$]. Skewed distributions of the outcome will be log transformed to calculate the ICC. SAS Proc GLIMMIX will be used to estimate variances if transformation of the variables does not result in a normal distribution. Repeatability will be interpreted based on the following benchmarks: slight, 0 to 0.2; fair, 0.21 to 0.4; moderate, 0.41 to 0.6; substantial, 0.61 to 0.8; and almost perfect, 0.81 to 1.0 (Fleiss, 1981).

Repeatability at 3, 6, and 12 months will also be evaluated in the ARIC study (Visit 3, 1993-1995; Visit 4, 1996-1998) and FHS (1994-1995) at repeated examination visits conducted one-year apart, as previously done by Weatherly and colleagues (Weatherley et al., 2006). In addition to the ICC, we will estimate the chance-corrected agreement of categorical IR at 3, 6, and 12 months evaluated separately for IR indexed by TG/HDL-C ratio cut points and HOMA-IR cut points. This analysis will inform classification of categorically defined IR.

Methodologic limitations or challenges:

Our study will evaluate the repeatability of surrogate IR measures at different repeat examinations in the ARIC study and in FHS. The length of time elapsed between repeat visits differs by participant and may require adjustment. Our study will also compare the repeatability of surrogate IR measures at different life epochs from mid-life to older adulthood. Laboratory assay methods are subject to drift due to a change in methods or change in control materials, which have implications for the accurate measurement of temporal changes and the estimation of associations. Systematic differences in fasting TG and HDL-C over time, however, were assessed in the ARIC study (Parrinello et al., 2015a). Blood samples were re-assayed for analytes at multiple visits in 2011-2013 for 200 men and women attending all 5 study visits (ARIC, 2012). The original values were then recalibrated to remeasured values using Deming regression. For fasting TG and fasting HDL-C, the percent bias between original values and remeasured values was less than 10% at each visit. Based on these results and other considerations, recalibration equations were not recommended for either fasting TG or HDL-C (Parrinello et al., 2015a). This work will be taken into consideration in our analysis.

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 ICTDER 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)?

- **Please see the table attached at the end of this document for related studies on repeatability.**

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
___ A. primarily the result of an ancillary study (list number* _____)
__X__ 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/

This proposal will use data from the following ancillary studies:

- **NHLBI Family Heart Study (FHS)**

- **ARIC Carotid MRI Flow Cytometry Study**

12a. 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.

12b. The NIH instituted a Public Access Policy in April, 2008 which ensures that the public has access to the published results of NIH funded research. It is **your responsibility to upload manuscripts to PUBMED Central** whenever the journal does not and be in compliance with this policy. Four files about the public access policy from http://publicaccess.nih.gov/ are posted in http://www.cscc.unc.edu/aric/index.php, under Publications, Policies & Forms. http://publicaccess.nih.gov/submit_process_journals.htm shows you which journals automatically upload articles to PubMed central.

13. Per Data Use Agreement Addendum for the Use of Linked ARIC CMS Data, approved manuscripts using linked ARIC CMS data shall be submitted by the Coordinating Center to CMS for informational purposes prior to publication. Approved manuscripts should be sent to Pingping Wu at CC, at pingping_wu@unc.edu. I will be using CMS data in my manuscript.
____ Yes __X__ No

Repeatability Studies Published in the ARIC study

	Title	N	Year	Time Between Visits
1[a]	Short-term intraindividual variability in lipoprotein measurements: the Atherosclerosis Risk in Communities (ARIC) Study (Chambless et al., 1992a)	40	1988	Visits were 1 to 2 weeks apart (mean: 8 days; range: 6 to 14 days); 45-64 years old
2[a]	Short-term intraindividual variability in hemostasis factors. The ARIC Study. Atherosclerosis Risk in Communities Intraindividual Variability Study (Chambless et al., 1992b)	39	1988	Visits were 1 to 2 weeks apart (mean: 8 days; range: 6 to 14 days); 45-64 years old
3[a]	ARIC hemostasis study -- IV. Intraindividual variability and reliability of hemostatic factors. The Atherosclerosis Risk in Communities (ARIC) (Nguyen et al., 1995)	39	1988	Visits were 1 to 2 weeks apart (mean: 8 days; range: 6 to 14 days); 45-64 years old
4	Atherosclerosis Risk in Communities (ARIC) Carotid MRI flow cytometry study of monocyte and platelet markers: intaindividual variability and reliability (Catellier et al., 2008)	55	2005-2006	Visits were 4 to 8 weeks apart.
5	Total Short-Term Variability in Biomarkers of Hyperglycemia in Older Adults (Parrinello et al., 2015b)	153	2011-2013	Visits were 3 to 15 weeks apart (mean: 45 days; range: 23 to 102 days); mean age 76
6	The reliability of the ankle-brachial index in the Atherosclerosis Risk in Communities (ARIC) study and the NHLBI Family Heart Study (FHS) (Weatherley et al., 2006)	119	FHS (1994-1995); ARIC (1993-1995; 1996-1998)	Repeat visits within 365 days (mean: 228 days)

[a] Same Study

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