ARIC Manuscript Proposal # 1240r

PC Reviewed: _6_/_5_/07	**Status: _A___**	**Priority: _2__**
SC Reviewed: _____	**Status: _____**	**Priority: ____**

1.a. Full Title: Characterization of the APOE gene region for association with LDL-C: Influence of a novel variant in the HCR2 region.

b. Abbreviated Title (Length 26 characters): The APOE HCR2 region and LDL-C

2. Writing Group:

Writing group members: Kathy Klos, Lawrence Shimmin, Eric Boerwinkle, Charlie Sing, Kiang Liu, Christie Ballantyne, Josef Coresh, Craig Hanis, James Hixson.

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

F**irst author**: **Kathy L.E. Klos**
Address: 1200 Herman Pressler
Houston, TX 77030

Phone: 713-500-9914 Fax: 713-500-0900
E-mail: kathy.klos@uth.tmc.edu

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

Phone: Fax:
E-mail:

3. Timeline: Genotyping and Analysis – February-April
Manuscript preparation – April-May
Submission - June

4. Rationale: DNA variation in the APOE gene is known to influence variation among individuals in plasma low-density lipoprotein cholesterol (LDL-C) level. We have identified 305 DNA variants in sequences from 120 hybrid cell lines (20 European-Americans, 20 African-Americans and 20 Mexican-Americans). After excluding

singleton and copy number variants, we successfully genotyped 3,999 participants (1,943 Whites and 2,046 African-Americans) of the Coronary Artery Risk Development in Young Adults (CARDIA) study for 115 SNPs. Association analysis indicated at least one SNP, a previously unreported common variant in the hepatic control region 2, was associated with LDL-C level independent of the APOE ε2/3/4 polymorphism.

5. Main Hypothesis/Study Questions: We will genotype this SNP in the ARIC cohort and in the three samples of the Genetic Epidemiology Network of Arteriopathy (GENOA) to ask if the association observed in CARDIA is also apparent in these samples.

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) Analyses to be performed in the ARIC cohort: We will exclude non-fasting individuals, individuals taking lipid-lowering medications, individuals with plasma triglyceride levels >400 mg/dL and individuals who did not consent to genetic analyses. Statistical analyses will be performed separately by race. Dependent variable : LDL-C. Covariates: gender, age, bmi, use of hypertension meds. For consistency with prior CARDIA analyses, LDL-C will be adjusted for age, age2, age3 and BMI within race and gender by fitting a linear model and adding the residual back to the race- and gender-specific grand mean. LDL-C will then be adjusted for gender within race using the same method. A general linear model analysis will be used to evaluate association between SNP genotype and LDL-C level. Two methods will be used to account for APOE ε2/3/4 genotype effect: a linear model adjustment within race (as above) and evaluation of the HCR2 region SNP within ε3/ ε3 individuals only.

2) Analyses for association with CHD case status (independent of effects on lipid metabolism): We will use a Cox proportional hazards model to estimate the hazard ratio of incident CHD, follow-up time = time from initial clinic visit to first event. Covariates as above, plus LDL-C, HDL-C, smoking, diabetes and hypertension status. The Wald ChiSquare statistic will be used to assess significance of the covariates in the model. Analyses will be performed within race.

3) We will also describe trajectories of mean lipid levels by age within apoE genotype using a random effects model with data from all 4 ARIC visits. We recognize that losses to follow-up between visits will influence the results but the ability to look at trajectories (mean levels and change from baseline) within individuals is a strength of the ARIC study.

LDL-C will the of primary focus because of the findings in CARDIA but associations with triglycerides and HDL will be explored as well.

Analysis: Kathy Klos – I have signed an internal DDA (Eric Boerwinkle PI).

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_DNA = "CVD Research" would be used? _X__ 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? _X__ Yes ____ 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"?
__X_ 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)? None related

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* 1995.07)
___ B. primarily based on ARIC data with ancillary data playing a minor role (usually control variables; list number(s)* _____ _____ _____)

Genotyping will be performed in the laboratory of Eric Boerwinkle under funding provided as part of ancillary study number 1995.07.

*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.