ARIC Manuscript Proposal #1934

PC Reviewed: 4/17/12	**Status:** <u>A</u>	**Priority:** <u>2</u>
SC Reviewed: _____	**Status:** _____	**Priority:** ____

1.a. Full Title: Genetic contributions to statin-related rhabdomyolysis

 b. Abbreviated Title (Length 26 characters): Genetics of rhabdomyolysis

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

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3.   **Timeline**: Genotyping will be complete in April.
               Non-ARIC case samples (N=187) are in hand.
               ARIC sequencing is complete.
               Sequencing of cases will be complete by June.
               Analyses will take place in July – December
               Manuscript preparation will take place in December and January.

4.   **Rationale**:

Identifying, replicating, and understanding the mechanism of potential drug-gene interactions is a formidable challenge in a translational-research effort to use genomics to improve public health. To date, the study of severe adverse drug reactions (ADRs) has yielded some of the most impressive and useful pharmacogenetic advances to date. What used to be regarded as idiosyncratic unpredictable type B reactions--for instance, abacavir and the hypersensitivity syndrome (1,2), flucloxacillin and drug-induced liver injury (3,4), and statin-associated myopathy (5,6)--are now known to be associated with specific genetic variants. While screening appears to be effective for some immune-mediated ADRs (7), public health advances for statin-related rhabdomyolysis await additional discovery.

Recent trials and meta-analyses support not only more aggressive statin therapy in high risk individuals (8,9) but also the expansion of treatment to intermediate risk persons (10). As a result, more US prescriptions are dispensed for lipid-lowering drugs than for any other class of drug. In 2009, both simvastatin and atorvastatin were among the top 5 products dispensed. The expanded use in intermediate-risk populations and the increasing use of high doses for high-risk populations provides new opportunities for both benefits and risks. The most serious ADR associated with statins is rhabdomyolysis. The bimodal response to statins--a serious ADR in a small proportion of users and none in the vast majority of users--suggests genetic factors as a potential cause (11,12).

Indeed, an analysis of GWAS data in 185 cerivastatin users provides evidence of additional undiscovered genetic loci that influence rhabdomyolysis risk (6). Even after excluding SNPs with minor allele frequencies less than 2.5%, there was an excess of p-values below 10^{-4}, but no departure from a null p-value distribution for p-values above 10^{-2}. This excess of small p-values is consistent with the presence of undetected variants at multiple loci, and the lack of departure from the null distribution at larger p-values suggests that cryptic population structure is not the cause. There were 44 p-values below 10^{-4}, compared with an expected number of 30; this statistically significant excess (p=0.012) suggests the presence of additional loci that affect rhabdomyolysis risk.

While previous basic-science work has identified several pathways such as protein prenylation and cystolic calcium release that are important in statin-induced apoptosis, the number of such candidate genes is large but limited, on the order of 100 to 500. Exome sequencing is the most efficient method for screening a large number of genes for uncommon disabling variants that are likely to contribute to the complex genetics of this extreme adverse drug-reaction phenotype (13-15).

The purpose of the proposed project is to identify the genes and the uncommon disabling variants that determine the risk of rhabdomyolysis. We hypothesize that statin-induced rhabdomyolysis resembles a heterogeneous Mendelian disorder that may have several forms caused by a drug-gene interaction involving many loci in multiple genes. Examples of heterogeneous Mendelian disorders include Charcot-Marie-Tooth neuropathy [39 loci in 31 genes (16)], dilated cardiomyopathy [rare variants in 30 genes (17)], and long-QT syndrome [hundreds of variants in 12 genes, (18)]. Examples that result from gene-environment interactions include phenylketonuria, which is caused by dietary phenylalanine in susceptible patients [hundreds of variants in several genes (19,20)], and malignant hyperthermia, which is caused by exposure to fluorinated-inhalation anesthetics in susceptible patients [many rare variants in 6 loci (21,22)].

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5. Main Hypothesis/Study Questions:

The primary aim of the proposed manuscript is to conduct a whole-exome-sequencing case-control study of rhabdomyolysis to identify the specific mechanisms that underlie this toxic drug-response phenotype.

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).

Cases: Cases (N=187) will come from a completed case-control study of cerivastatin (Baycol).

Controls are CHS and ARIC study participants with exome sequencing completed. The ideal control group would be limited to those who are also taking a statin, cerivastatin in particular. Because rhabdomyolysis is so rare (about 5-10 per 100,000 person years), the statistical power of our analyses remains strong despite lack of documentation about the presence or absence of a history of rhabdomyolysis in controls, some of whom may not have even used statins. (The rare variants that we are looking for are likely to occur in only about 1 in 10,000 persons, so even if there are 1800 controls from CHS and ARIC, few if any are likely to have the mutations that might be a cause of rhabdomyolysis.)

Note: A companion manuscript proposal is being sent to the CHS publication committee.

- For association analyses with common variants, logistic regression will be used adjusting for sex, age, BMI and principal components to adjust for hidden substructure.
- For association analyses with rare variants, the Madson-Browning burden test will be used with the same covariates. The Madson-Browning burden test combines rare variants across a gene, and is similar to a rank sum test.
- Exploratory analyses will be carried-out assuming that rhabdomyolysis is an autosomal recessive disorder in a subset of the cases. Therefore, we will use bioinformatics analyses to identify individuals with two loss of function variants in the same gene. We will then use a permutation analysis to place a p-value on the observed distribution of loss of function variants.

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? __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 ICTDER03 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.

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)* __2009.12___)

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

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. Agree.

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. Agree.