ARIC Manuscript Proposal #2615

PC Reviewed: 9/8/15	**Status: A**	**Priority: 2**
SC Reviewed: _____	**Status: _____**	**Priority: ____**

1. a. Full Title: Serum magnesium and the incidence of coronary heart disease over 20 years of follow-up: the ARIC study.

b. Abbreviated Title (Length 26 characters): Mg and CHD

2. Writing Group:
Writing group members: Jeffrey R. Misialek, Alvaro Alonso, Aaron R. Folsom, Erin D. Michos, Casey M. Rebholz, Pamela L. Lutsey

I, the first author, confirm that all the coauthors have given their approval for this manuscript proposal. JM **[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:
Data analysis will begin immediately. We anticipate completion of the manuscript within 1 year.

4. Rationale:

Magnesium (Mg) is an abundant cation and micronutrient that plays many crucial roles in the body by activating enzymes, contributing to energy production, and regulating calcium levels and related biomarkers.[1,2] Serum Mg measured from blood has been shown to be usually predictive of the total Mg levels in the body and has been used to examine the relationship between Mg levels in the body and many outcomes such as cardiovascular disease (CVD). Low serum Mg levels may be associated with CVD through physiological pathways and roles that lead to elevated blood pressure, chronic inflammation, impaired vasomotor tone and peripheral blood flow.[1,3-5]

A 2013 meta-analysis summarized findings from prospective studies evaluating the associations of serum Mg with CVD risk and concluded that there was an inverse association, with low serum Mg levels or intake leading to a higher CVD risk. Looking specifically at coronary heart disease (CHD) events and fatal CHD, an inverse association was also identified for both outcomes, but the CHD results were non-significant while the fatal CHD results were borderline significant.[6] Included in this meta-analysis was a 1998 ARIC manuscript that examined Mg and incident CHD. Over the four to seven years of follow-up, there were only 319 incident CHD events, and a sex interaction was reported whereby an association was present and stronger among women than men.[7] Since the publication of this manuscript, though, the number of incident CHD cases has risen to 2,279 through the end of 2012.

Using data from the ARIC study, we propose to reexamine the prospective association between serum Mg and CHD in both whites and African Americans at visit 1 through the end of 2012. Given how little is known about the association between serum Mg and incident CHD, we believe that it is important to reexamine this association with ARIC's present capacity to attain more precise estimates and evaluate subgroup associations.

5. Main Hypothesis/Study Questions:

Aim #1: To determine if serum Mg level is associated with the incidence of CHD through the end of 2012.

Hypothesis: Individuals with lower serum Mg levels will have an increased risk for CHD.

Aim #2: To determine if age, gender, and race modify the association between serum Mg level and CHD.

Hypothesis: The Mg-CHD association will be present and of a similar magnitude regardless of stratifying by age, gender, or race.

Aim #3: To evaluate whether prevalent diabetes or hypertension explain the association between serum Mg and incident CHD.

Hypothesis: Adjustment for diabetes and hypertension will partially explain the association between Mg and incident CHD.

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:
Prospective cohort from Visit 1 through the end of follow-up.

Inclusion/exclusion criteria:
We will exclude those individuals based on the following criteria:

(1) A race other than white or African-American.
(2) African Americans from the Minnesota and Washington County sites.
(3) Prevalent CHD or missing information on prevalent CHD at baseline.
(4) Missing a baseline serum Mg measurement at visit 1.
(5) Not fasting at least eight hours before the baseline examination.
(6) Missing other covariate information.

Variables of interest:

Main outcome of interest: CHD incidence
The time to incident CHD cases from baseline through December 31, 2012, will be the main outcome variable.

Main independent variables of interest: Serum Mg
In the ARIC study, serum Mg was assessed through laboratory tests at visits 1 and 2 from fasting blood samples. To increase precision, we will average these measures for the primary analyses. We will also look at visit 1 Mg as a secondary analysis.

Covariates
From visit 1, the categorical covariates to be included in this analysis are gender, race, study site, diabetes (yes, no), drinking status (current, former, never), educational level (<high school graduate, high school graduate, college/graduate school), eGFR category (15-59, 60-89, $\geq$90 ml/min/1.73 m^2), smoking status (current, former, never), use of antihypertensive medications (yes, no), use of lipid-lowering medications (yes, no), and hormone replacement therapy (men, women on HRT, women not on HRT). The following continuous variables will be included as well: age, body mass index (BMI), cigarette years, ethanol intake, high-density lipoprotein cholesterol (HDL-c), low-density lipoprotein cholesterol (LDL-c), sports index, systolic blood pressure, fruit intake, and vegetable intake.

Statistical analysis:

Cox proportional hazards models will be used to determine the association between serum Mg and incident CHD through 2012. Initially, we will explore the shape of the association between Mg and CHD risk using restricted cubic splines. If appropriate, serum Mg will be divided into ranked quintiles based on the average of the two visit measurements. For individuals who did not attend visit 2 or were censored before the visit, their visit 1 serum Mg measurement will be used for ranking. For those individuals who attended both visits and were censored after visit 2, the mean value of their two serum Mg measurements will be used for ranking. The following models will be used to analyze the serum Mg-CHD association:

- Model 1: adjustment for age, gender, race, and ARIC study site
- Model 2: Model 1 + adjustment for BMI, cigarette years, drinking status, ethanol intake, education level, smoking status, sports index, fruit intake and vegetable intake.
- Model 3: Model 2 + adjustment for diabetes, HDL-c, LDL-c, lipid-lowering medications, systolic blood pressure, antihypertensive medications, eGFR, and hormone replacement therapy.

Effect modification will also be evaluated by age, gender, race, and diabetes by including multiplicative terms between the potential effect modifier and Mg measures in the models. Stratified results will be reported as appropriate. Regardless of whether a significant sex interaction is present, we plan to present results overall and stratified by sex based on the prior ARIC study results.

We will examine the serum Mg-CHD association further in the following sensitivity analyses:

(1) Excluding those individuals on diuretics, which tend to decrease serum Mg concentrations in the body.
(2) Repeat the analysis through ten years of follow-up to determine if the association was a similar magnitude to the complete follow-up results.
(3) Examining the association using the visit 1 serum Mg measurement only to see if the results are similar to the results using both visit 1 and visit 2 serum Mg measurements.
(4) Looking at the association of non-fatal CHD and fatal CHD as separate outcomes with serum Mg.

<u>Strengths and limitations</u>:
Strengths of the study include the large sample size and power to measure associations between serum Mg and CHD, especially in a biracial population. Limitations include some misclassification of the Mg exposure since there is no follow-up information on serum Mg beyond visit 2. In our interpretation, we will note that Mg levels may not be reflective of the serum Mg level immediately preceding a CHD event, which may be more relevant as a predictor of CHD.

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**

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, but overlap was found. _____ 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)?

The previous manuscript proposal#438 in ARIC specifically examined the association between serum Mg and CHD. However, as noted in the Rationale, we believe it is worthwhile to reevaluate this association with more CHD cases. Other ARIC manuscripts have explored the association between individual serum Mg/dietary Mg and CVD.

#1196 Mg and sudden death
#1268 Mg and stroke
#1819 Mg and atrial fibrillation
#1893 Mg and heart failure

11. a. Is this manuscript proposal associated with any ARIC ancillary studies or use any ancillary study data? ___ Yes X **No**

b. If yes, is the proposal
__ A. primarily the result of an ancillary study (list number*)
__ 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.

References:

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4. Rude RK. Magnesium. In: Coates PM, Betz JM, Blackman MR, Cragg GM, eds. *Encyclopedia of Dietary Supplements*. 2nd ed. New York: Informa Healthcare; 2010:527-537.
5. Jee SHa, Miller ER, Guallar E, Singh VK, Appel LJ, Klag MJ. The effect of magnesium supplementation on blood pressure: a meta-analysis of randomized clinical trials. *American Journal of Hypertension.* 2002;15(8):691-696.
6. Del Gobbo LC, Imamura F, Wu JH, de Oliveira Otto MC, Chiuve SE, Mozaffarian D. Circulating and dietary magnesium and risk of cardiovascular disease: a systematic review and meta-analysis of prospective studies. *Am J Clin Nutr.* 2013;98(1):160-173.
7. Liao F, Folsom AR, Brancati FL. Is low magnesium concentration a risk factor for coronary heart disease? The Atherosclerosis Risk in Communities (ARIC) Study. *Am Heart J.* 1998;136(3):480-490.