

ARIC Manuscript Proposal # 1371

PC Reviewed: 05/13/08
SC Reviewed: _____

Status: A
Status: _____

Priority: 2
Priority: _____

1.a. Full Title: Association of the NOS3 Glu298Asp SNP with hypertension and possible effect modification of dietary fat intake.

b. Abbreviated Title (Length 26 characters): NOS3, hypertension and dietary fat

2. Writing Group: Pascal L. Kingah, MD; Hung Luu, MD, MPH; Kelly Volcik, PhD; Alanna Morrison, PhD; Eric Boerwinkle, PhD; Jennifer Nettleton, PhD.

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

First author: Pascal L Kingah, MD

Address: UT School of Public Health
University of Texas Health Science Center at Houston
1200 Herman Pressler, Box 134
Houston, TX 77030
Phone: 713-432-7666 Fax: 713-432-7666
E-mail: Pascal.L.Kingah@uth.tmc.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: Kelly Volcik
Address: Human Genetics Center
UTHSCH School of Public Health
1200 Herman Pressler
Houston, TX 77030
Phone: 713-500-9891 Fax: 713-500-0900
E-mail: Kelly.A.Volcik@uth.tmc.edu

3. Timeline: Genotyping for the NOS3 Glu298Asp SNP (rs1799983) is complete for the entire ARIC cohort. Statistical analysis will begin immediately, with a first draft manuscript prepared by August 2008.

4. Rationale:

In the United States, 1 out of every 3 persons aged 20 and older has hypertension, with more than 100 million people having either pre-hypertension or definite hypertension (1-2). Both human and animal studies have revealed evidence of hypertension due to the loss of the effect of

nitric oxide, which acts as the endothelium derived relaxing factor (3). Nitric oxide is catalyzed by endothelial nitric oxide synthase (eNOS), an enzyme with multiple genetic variants that might confer risk for hypertension (4). Three SNPs in the NOS3 gene have been shown to be associated with hypertension, including Glu298Asp, T786C, and a base pair VNTR in intron 4 (5). However, inconsistent findings on the relationship between NOS3 polymorphisms and hypertension have been observed in previous studies, with significant associations reported from Japan (6) and Singapore(7) , but no associations observed in China(8) and Australia(9). In a recent meta-analysis by Pereira et al, the association between the NOS3 Glu298Asp SNP and hypertension was shown to be modified by specific dietary patterns (e.g. diets rich in saturated fat) and conditions (e.g. hypercholesterolemia) (10). This meta-analysis included studies comprised of subjects from Europe, China and Japan, but did not include subjects from the United States. Apart from publication bias, one of the most common limitations among these previously reported studies is the low power to detect the strength of association due to small sample sizes.

Being a cohort with a large sample size, the ARIC study offers an opportunity to determine stronger effect measures between NOS3 and hypertension in both whites and blacks, with additional analyses of possible gene-diet interactions. Studies have shown that the impact of n-3 fatty acids on endothelial function depends on eNOS genotype with the influence of n-3 fatty acid level being greater in Asp298 carriers.(11) Interactions between cholesterol and regulatory mechanisms of blood pressure are poorly understood and the role of individual fatty acids in blood pressure regulation remains unclear.(12,13,14). We propose to evaluate the association between the NOS3 Glu298Asp SNP (rs1799983) and hypertension, as well as the possible effect modification of dietary fat intake (saturated fat, monounsaturated fat, polyunsaturated fat).

5. Main Hypothesis/Study Questions:

- 1- Determine the frequency distribution of the NOS3 Glu298Asp SNP in the ARIC study. If allele frequencies are markedly different with respect to race, all subsequent analyses will be stratified by race.

- 2- Using logistic regression, evaluate the independent effect of the NOS3 Glu298Asp SNP on hypertension status. Models will be adjusted for age, gender, race, BMI, smoking status, diabetes and physical activity.

- 3- Evaluate whether dietary intake modulates the independent effect of the NOS3 Glu298Asp SNP on hypertension status. Models will be adjusted for age, gender, race, BMI, smoking status, diabetes and physical activity.

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).

Hypertension status at visit 1 will be the primary dependent variable (HYPERT05). The usual DNA restriction, ethnic group and missing data exclusion criteria will be used. Exclusions will include the following: 1) prohibited use of DNA, 2) ethnic background other than white or

African American, as well as African Americans not from Jackson or Forsyth. Independent variables include but are not limited to NOS3 genotype status and traditional risk factors such as age, gender, smoking, BMI and diabetes.

For the analyses of gene-diet interaction, we will focus on dietary intakes of saturated fat (SFAT), monounsaturated fat (MFAT) and polyunsaturated fat (PFAT). We acknowledge that diet is notoriously measured with error, and we will explore the effects of bias due to this error. If we feel the degree of bias is large, we will explore methods to correct for some degree of the error in reported dietary intake.

7.a. Will the data be used for non-CVD analysis in this manuscript? ___ Yes 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

8.a. Will the DNA data be used in this manuscript? 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"? 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.
 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)?

MS#950, North – Association of the NOS3 Glu298Asp SNP with non-invasively measured atherosclerotic burden and/or risk of adverse CVD events

MS#1131, Bielinski – Association of the NOS3 Glu298Asp SNP with serum levels of inflammation biomarkers and possible effect modification of dietary antioxidants

MS#1228, Bressler – Association of the NOS3 Glu298Asp SNP with diabetes and possible effect modification of obesity

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

11.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)* _____)**

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. Agree

References

1. Kearney PM, Whelton M, Reynolds K, Muntner P, Whelton PK, He J. Global burden of hypertension: analysis of worldwide data. *Lancet* 2005; **365**:217–223.
2. Qureshi AI, Suri MF, Kirmani JF, Divani AA. Prevalence and trends of prehypertension and hypertension in United States: National Health and Nutrition Examination Surveys 1976 to 2000. *Med Sci Monit* 2005;**11**:CR403–CR409.
3. Lockette W, Otsuka Y, Carretero O. The loss of endothelium-dependent vascular relaxation in hypertension. *Hypertens*. 1986;**8(Supp. II)**:61-II-66.
4. Nadaul S, Bonnadaux A, Lathrop M, Soubrier F. Gene structure polymorphism and mapping of the human endothelial nitric oxide synthase gene. *Biochem Biophys Res Commun*. 1994;**198**:1027-1033.
5. Wattanapitayakul SK, Mihm MJ, Young AP, Bauer JA. Therapeutic implications of human endothelial nitric oxide gene polymorphism. *Trends Pharmacol Sci* 2001;**22**:361-368.
6. Miyamoto Y, Saito Y, Kajiyama N, Yoshimura M et al. Endothelial nitric oxide synthase gene is positively associated with essential hypertension. *Hypertension* 1998; **32**:3-8.
7. Moe KT, Lim ST, Wong P et al. Association analysis of endothelial nitric oxide synthase gene polymorphism with primary hypertension in a Singapore population. *J Hum Hypertens*. 2006;**20**:956-963.
8. Zhao Q, Su SY, Chen SF et al. Association study of the endothelial nitric oxide synthase gene polymorphisms with essential hypertension in northern Han Chinese. *Chin Med J(Engl)* 2006;**119**:1065-1071.
9. Granath B, Taylor RR, van Bockxmeer FM, Mamotte CD. Lack of evidence for association between endothelial nitric oxide synthase gene polymorphisms and coronary artery disease in the Australian Caucasian population. *J Cardiovasc Risk* 2001; **8**:235-241.
10. Pereira VT, Rudnicki M, Cheung MYB et al. Three endothelial nitric oxide (NOS3) gene polymorphisms in hypertensive and normotensive individuals: meta-analysis of 53 studies reveals evidence of publication bias. *Journal of Hypertension*. 2007;**25**: 1763 -1774.
11. Leeson CPM, Hingorani AD, Mullen MJ et al. Glu298Asp endothelial Nitric oxide synthase gene polymorphism interacts with environmental and dietary factors to influence endothelial function. *Circulation Research* 2002; 90: 1153-1158.
12. Rubattu S, Volpe M, Enea I et al. Influence of hypercholesterolemia on adrenal steroid metabolism and electrolyte balance in spontaneously hypertensive rats *Endocrinology*. 1993 **133**: 2015-2021
13. Pomposiello IS, Alva M, Wilde WD et al. Linoleic Acid Induces Relaxation and Hyperpolarization of the Pig Coronary Artery. *Hypertension*. 1998;**31**(2):615-620
14. Grimsgaard S, Bønaa HK, Jacobsen KB et al. Plasma Saturated and Linoleic Fatty Acids Are Independently Associated With Blood Pressure. *Hypertension*. 1999;**34**: 478-483.