1.a. **Full Title**: Association of a common variant in Endothelial Nitric Oxide Syntase (Glu298Asp) with non-invasively measured atherosclerotic burden and/or risk of adverse cardiovascular events: The Atherosclerosis Risk in Communities Study

b. **Abbreviated Title (Length 26 characters)**: eNOS and atherosclerosis

2. **Writing Group (list individual with lead responsibility first)**:

   **Lead**: Kari North  
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   Writing group members: Molly Bray, Eric Boerwinkle, Joe Coresh, Darryl Zeldin, Craig Lee, David Couper, Aaron Folsom, Gerardo Heiss (others are welcome)

3. **Timeline**:

   Anticipate data will be available: Immediately  
   Analysis to be completed: 10/2003  
   First draft anticipated: 12/2003

4. **Rationale**:

   Endothelial dysfunction (ED) may play an important role in the pathogenesis of atherosclerotic cardiovascular disease. ED can be manifested by reductions in endothelial-dependent vasodilatation, primarily due to functional impairments in endothelial nitric oxide (NO) synthase (eNOS) and NO availability.

   Although the impact of the Glu298Asp polymorphism on endothelial NO function remains to be clarified, recent studies have shown an association between this mutation and reduced basal NO production (1). Moreover, Philip and colleagues (2) demonstrated enhanced vasoconstriction in response to phenyleprine in Asp298 homozygotes that is suggestive of impaired endothelial NO modulation of adrenergic vasoconstriction. Tanus-Santos et al.(3) found marked interethnic
differences in the distribution of the Asp298 eNOS variants, being more common in Caucasians (34.5%) than in African-Americans (15.5%) or Asians (8.6%) (P < 0.0001).

Several studies have shown that the Glu298Asp polymorphism is associated with increased risk of myocardial infarction (4-6), stroke (7), measures of carotid atherosclerosis (8), and coronary artery disease (4). In contrast, no relationship between the Asp polymorphism and stroke (9), transient ischemic attacks (9), coronary artery disease (10), or carotid artery disease (11) were detected. Gene-Environment interaction may be important to consider as recent studies have identified the importance of gene (Asp variant) by smoking(12) interaction for endothelial functioning.

5. Main Hypothesis/Study Questions:

To determine if the Glu298Asp polymorphism in the gene encoding eNOS, an enzyme important in the biosynthesis of NO, is significantly associated with non-invasively measured atherosclerotic burden and/or risk of adverse cardiovascular events in individuals enrolled in the ARIC Study.

Hypothesis 1: The eNOS Glu298Asp polymorphism is associated with incident coronary heart disease (CHD).
Hypothesis 2: The eNOS Glu298Asp polymorphism is associated with increased carotid artery intima-media thickness (IMT).
Hypothesis 3: The eNOS Glu298Asp polymorphism is associated with peripheral arterial disease (PAD).
Hypothesis 4: The eNOS Glu298Asp polymorphism is associated incident stroke.
Hypothesis 5: The eNOS Glu298Asp polymorphism is associated with MRI-detected cerebral infarct-like brain lesions (CI).

Case-cohort case-control analyses will be used, per the design selected for each of the end points. The analyses will be run by K. North in collaboration with D. Couper, at the ARIC Coordinating center. Because of the commonalities between the study end points hypotheses 1-5 will be tested in a first set of parsimonious analyses. The results will be reviewed by the writing group, to inform additional analyses and to determine whether several manuscripts are warranted.

6. Data (variables, time window, source, inclusions/exclusions):

A. eNOS genotype data
B. Case status on five endpoints quantifying subclinical atherosclerosis and clinical atherosclerotic events: incident coronary heart disease (CHD), increased carotid artery intima-media thickness (IMT), peripheral arterial disease (PAD), incident stroke, and MRI-detected cerebral infarct-like brain lesions (CI).
C. The corresponding CRS or control group(s)
D. Information on demographic covariates (e.g., age, sex, race)
E. Information on other cardiovascular disease risk factors, to use as covariates (e.g., smoking, LDL cholesterol levels, BMI, etc.)

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://bios.unc.edu/units/cscc/ARIC/study/studymem.html

___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. 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.

