ARIC Manuscript Proposal #2456

PC Reviewed: 10/14/14	**Status: A**	**Priority: 2**
SC Reviewed: _____	**Status: _____**	**Priority: ____**

1.a. Full Title: Associations of ϒ′ fibrinogen, factor XI and D-dimer with venous thromboembolism and atherothrombotic CVD

b. Abbreviated Title (Length 26 characters): Hemostatic factors, VTE and CVD

2. Writing Group:

Writing group members: Aaron Folsom, Weihong Tang, Alvaro Alonso, Jim Pankow, Susan Heckbert, Mary Cushman

I, the first author, confirm that all the coauthors have given their approval for this manuscript proposal. _AF_ **[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: Start in November 2014.

4. Rationale:

The LITE study of venous thromboembolism (VTE) involves ARIC and CHS. LITE recently measured ϒ′ fibrinogen and factor XI in both cohorts. The ARIC NCS study measured D-dimer. This paper will analyze these factors in relation to incident VTE and atherothrombotic CVD.

Υ′ Fibrinogen

Fibrinogen has two copies of the Aα, Bβ, and Υ chains. The Υ chain, produced under control of *FGG*, has two isoforms ΥA and Υ′. Υ′ comprises approximately 10% of total fibrinogen but varies among individuals. Clots made from Υ′ fibrinogen are more resistant to lysis than normal clots; therefore the proportion of Υ′ fibrinogen was hypothesized to be a risk factor for arterial thrombosis.[1] Epidemiologic evidence, however, is limited and contradictory. Recently, the Framingham Study showed plasma Υ′ fibrinogen was positively associated with prevalent arterial CVD (RR=2 for extreme tertiles) independent of total fibrinogen.[2] More relevant to LITE, the Leiden Thrombophilia Study found a *FGG*-H2 haplotype was strongly related to lower Υ′ fibrinogen levels.[3] The haplotype itself doubled the risk of VTE and Υ′ fibrinogen was negatively associated with VTE even though total fibrinogen was positively associated with VTE. These VTE findings, which seemingly contradict those for CHD, warrant replication. Another rationale for studying Υ′ fibrinogen is that in our VTE GWAS, known *FGG* variants did not fully explain the association of our newly discovered *FGG* locus with VTE. Measuring Υ′ fibrinogen therefore allows us to assess linkages between *FGG* variation, Υ′ fibrinogen level, and VTE risk.

Factor XI

Evidence is strong that elevated factor XI levels increase VTE risk,[4-6] but prospective data are limited. We previously did a VTE nested-case control study indicating a RR=1.8 (95% CI, 1.3-2.7) for highest vs. lowest factor XI quintiles.[7] In another nested case-control study, we showed a positive association of factor XI with stroke, but not CHD.[8,9] Another study of young women also showed a positive association with stroke but not CHD. [12] Having expanded factor XI measurement to the entire LITE sample will allow us to verify these associations with more precision, but more importantly, will allow us to more fully assess linkages between *F11* genetic variation, FXI concentration, and VTE occurrence.

D-dimer

D-dimer was associated positively with incident VTE in a LITE nested case-control study,[10] and it has been associated positively with atherothrombotic CVD in many studies.[11] We will try to replicate these findings.

5. Main Hypothesis/Study Questions:

Υ′ fibrinogen, factor XI and D-dimer are associated positively with incident VTE, CHD, and stroke.

The analysis will include ARIC for all three analytes and CHS for the first two. It is likely that we will split VTE from the other two atherothrombotic endpoints, but we request that this manuscript be approved with them lumped. In an additional analysis, we will use SNP data for a Mendelian randomization exploration.

Also, it is possible that members of the writing group will be interested in other endpoints, such as PAD, heart failure atrial fibrillation, or aortic aneurysm, but those would be proposed separately.

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).

Design: cohort (ARIC, CHS)

Endpoint: incidences of VTE, stroke, CHD

Exposure: ϒ′ fibrinogen, factor XI and D-dimer. Transformation for skewness, as needed, or use quantiles.

Exclusions: depending on the outcome under consideration, exclude VTE, stroke or CHD prior to exam visit, anticoagulant use, and missing hemostatic factor.

Covariates: VTE or CVD risk factors from the exam visit where blood was drawn (visit 3 in ARIC)

Analysis for main associations: For LITE, we typically analyze ARIC and CHS separately and pools them only if there is no obvious heterogeneity. The shape of association will be examined using cubic splines. We will use Cox proportional hazards models for each endpoint (VTE, CHD, stroke), with adjustment for relevant confounders. We will also examine Visit 1 total fibrinogen in the ϒ′ fibrinogen analysis.

Analysis for Mendelian Randomization in relation to VTE: Mendelian randomization is a type of instrumental variable analysis that seeks to estimate unconfounded causal effects between a risk factor (e.g, FXI levels) and outcome of interest (e.g., VTE) through the use of genetic instruments (genetic variants). Assuming that we find associations between intermediate traits (FXI and ϒ′ fibrinogen levels) and VTE, and that some variants in *F11* or *FGG* are significantly associated with circulating levels of their gene products, we will conduct formal analyses to compare the <u>observed</u> strength of association between *F11* or *FGG* variants and VTE to that <u>predicted</u> based on associations between the (a) gene variants and the intermediate phenotypes; and (b) intermediate phenotypes and VTE. While recognizing the important limitations of the Mendelian randomization approach, agreement between these observed and predicted associations will provide additional support for a causal relation between the intermediate phenotypes and VTE risk. This analysis will involve only whites in ARIC and CHS, with a maximum sample size of 13,000 with data on *F11* and *FGG* gene variants, FXI and ϒ′ fibrinogen levels, and VTE events. Selection of gene variants in (a) above will be based on literature and ongoing GWAS projects for these hemostatic factors in ARIC and/or CHS. GWAS projects for these hemostatic factors will be submitted in separate MS proposals.

References

1. Lovely RS, Kazmierczak SC, Massaro JM, D'Agostino RB Sr, O'Donnell CJ, Farrell DH. Gamma′ fibrinogen: Evaluation of a new assay for study of associations with cardiovascular disease. *Clin Chem* 2010;56(5):781-788.

2. Lovely RS, Yang Q, Massaro JM, Wang J, D'Agostino RB Sr, O'Donnell CJ, Shannon J, Farrell DH. Assessment of genetic determinants of the association of ϒ′ fibrinogen in relation to cardiovascular disease. *Arterioscler Thromb Vasc Biol* 2011;31(10):2345-2352.

3. Uitte de Willige S, de Visser MC, Houwing-Duistermaat JJ, Rosendaal FR, Vos HL, Bertina RM. Genetic variation in the fibrinogen gamma gene increases the risk for deep venous thrombosis by reducing plasma fibrinogen gamma' levels. *Blood* 2005;106(13):4176-83.

4. Lijfering WM, Rosendaal FR, Cannegieter SC. Risk factors for venous thrombosis – current understanding from an epidemiological point of view. *Br J Haematol* 2010;149(6):824-833.

5. Meijers JC, Tekelunburg WL, Bouma BN, Bertina RM, Rosendaal FR. High levels of coagulation factor XI as a risk factor for venous thrombosis. *N Engl J Med* 2000;342(10):696-701.

6. Doggen CJ, Rosendaal FR, Meijers JC. Levels of intrinsic coagulation factors and the risk of myocardial infarction among men: Opposite and synergistic effects of factors XI and XII. *Blood* 2006;108(13):4045-4051.

7. Cushman M, O'Meara ES, Folsom AR, Heckbert SR. Coagulation factors IX through XIII and the risk of future venous thrombosis: the Longitudinal Investigation of Thromboembolism Etiology. *Blood* 2009;114(14):2878-2883.

8. Suri MFK, Yamagishi K, Aleksic N, Hannan PJ, Folsom AR. Novel hemostatic factor levels and risk of ischemic stroke: the Atherosclerosis Risk in Communities (ARIC) Study. *Cerebrovasc Dis* 2010;29(5):497-502.

9. Yamagishi K, Aleksic N, Hannan PJ, Folsom AR. Coagulation factors II, V, IX, X, XI, XII, plasminogen, and α-2 antiplasmin and risk of coronary heart disease: the Atherosclerosis Risk in Communities (ARIC) Study. *J Atheroscler Thromb* 2010;17(4):402-409.

10. Cushman M, Folsom AR, Wang L, Aleksic N, Rosamond WD, Tracy RP, Heckbert SR. Fibrin fragment D-dimer and the risk of future venous thrombosis. *Blood* 2003;101(4):1243-1248.

11. Willeit P, Thompson A, Aspelund T, Rumley A, Eiriksdottir G, et al. Hemostatic

factors and risk of coronary heart disease in general populations: New prospective study and updated meta-analyses. *PLoS One* 2013;8(2):e55175.

12. Siegerink B, Rosendaal FR, Algra A. Antigen levels of coagulation factor XII, coagulation factor XI and prekallikrein, and the risk of myocardial infarction and ischemic stroke in young women. J Thromb Haemost. 2014;12(5):606-13.

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 ICTDER 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)?

They are our publications, cited above.

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* 2006.16 _____)

___ 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/

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.

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.