ARIC Manuscript Proposal # 1402

1.a. Full Title: Retinopathy and Platelet Activation

b. Abbreviated Title (Length 26 characters): As above

2. Writing Group:
Writing group members: Anna Kucharska-Newton, Keri Monda, Richey Sharrett, Ron Klein, Nena Aleksic, Sherita Golden, Lloyd Chambless, Lynne Wagenknecht; Others are welcome

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

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3. Timeline: Data analysis to be started immediately following approval of the proposal and completed by April 2009. Manuscript preparation to be completed by July 2009.

4. Rationale:

Retinopathy is a common complication of diabetes [1, 2] and retinal microvascular changes in those without diabetes predict the onset of incident diabetes [3, 4]. Retinopathy is a marker of increased risk of macrovascular cardiovascular disease [5]. This increased risk underscores the
extent to which diabetes affects multiple metabolic pathways. One of the pathogenic elements of elevated cardiovascular disease risk in diabetes is an increased level of platelet activation and aggregation [6]. Insulin regulates platelet functions through effects on antagonists of platelet activation [7], through activation of plasminogen activator inhibitor PAI-1 [8], and possibly through anti-inflammatory effects on monocytes. Under conditions of insulin resistance, this regulatory function of insulin is diminished, resulting in increased platelet activation. Platelet activation manifests through increased density of platelet surface markers involved in the adhesion of platelets to the subendothelial matrix, platelet-platelet adhesions, and adhesion of platelets to circulating leukocytes [9]. Heterotypic platelet-monocyte aggregates are increasingly being recognized as markers of increased platelet activation [10, 11].

Association of retinopathy with platelet activation has not been studied extensively. Existing data suggest that retinopathy is associated with polymorphisms in platelet surface markers [12], increased levels of the platelet activation inhibitor-1 and factors of the coagulation pathway [13], and increased levels of platelet-leukocyte aggregates [14].

The aim of this study is to examine platelet activation and aggregation in the context of retinal microvascular changes. The flow cytometry component of the CarMRI study [15] provides a unique opportunity to examine risk factors associated with platelet activation. Retinal exams, performed as part of the CarMRI study will constitute the basis of this study. Retinopathy will be defined as the presence of specific lesions defining it (e.g., retinal microaneurysms, blot hemorrhages). Other retinal vascular changes will also be studied, e.g., presence of focal retinal arteriolar narrowing, presence of retinal arterio/venous nicking, and presence of generalized retinal arteriolar narrowing (retinal arteriolar narrowing and retinal venular widening). Density of the following platelet surface markers: CD41, CD61, CD62, as well as levels of platelet-monocyte aggregates will constitute dependent variables. Age, gender, cigarette smoking, lipid levels, blood pressure levels, and BMI will be evaluated as potential covariates. Presence of diabetes, duration of diabetes greater than 10 years (yes/no variable), and family history of diabetes will be considered as possible effect modifiers of the examined associations.

References:


5. **Main Hypothesis/Study Questions:**

Study questions:

a. Focal retinal microvascular changes and generalized arteriolar narrowing are associated with increased density of selected platelet surface markers.

b. Focal retinal microvascular changes and generalized arteriolar narrowing are associated with increased levels of platelet-monocyte aggregates.

c. Associations focal retinal microvascular changes and generalized arteriolar narrowing with the selected platelet surface markers and with levels of platelet-monocyte aggregates are stronger in persons with diabetes and in those with a positive family history of diabetes as compared to non-diabetics and those with a negative family history of diabetes.

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 population: This study will consist of two components, a cross-sectional analysis based on the CarMRI ARIC study (ARIC Visit 5) and a longitudinal analysis of change in retinal microvascular characteristics from ARIC Visit 3 to Visit 5 and its association with platelet activation.*
The CarMRI study, an ancillary ARIC study, was conducted in 2004-2005. Approximately 2000 ARIC cohort participants were selected for the study; 60% of individuals were selected based on their carotid artery wall thickness greater than 85th percentile and 40% of individuals constituted a weighted sample selected from the remaining carotid intima media thickness distribution. Retinal exams and whole blood flow cytometry analysis of platelet and monocyte surface markers were performed as part of the CarMRI study.

Data analysis: Analyses will be performed using multivariate linear regression with weighted analysis to account for sampling design.

Exclusions: missing covariates.

**Dependent variables:** Flow cytometry determined median fluorescence intensity (MFI) of the selected platelet markers (CD 41 CD 61 CD62) and of the platelet monocyte aggregates

**Independent variables:**
- Retinopathy presence
- Focal retinal microvascular changes (arteriovenous nicking, focal arteriolar narrowing, retinal hemorrhage, type of hemorrhage, microaneurysms and soft exudates),
- Generalized arteriolar narrowing (retinal arteriole-to-venule ratio (AVR), central retinal arteriolar equivalent, central retinal venular equivalent)

**Covariates:** Age, gender, cigarette smoking, lipid levels, blood pressure levels, and BMI will be evaluated as potential covariates. Presence of diabetes, duration of diabetes greater than 10 years (yes/no variable), and family history of diabetes will be considered as possible effect modifiers of the examined associations.

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

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

8.c. If yes, is the author aware that the participants with RES_DNA = ‘not for profit’ restriction must be excluded if the data are used by a for profit group? ____ 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)?
MS 1234 “10-year Incidence, Progression and Regression of Retinal Vascular Abnormalities and their Relationship with Vascular and Inflammatory Risk Markers”

11. a. Is this manuscript proposal associated with any ARIC ancillary studies or use any ancillary study data?  _____ Yes  ___X___ 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)* _________ _________ _________

*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.