1.a. Full Title: Preclinical risk stratification for incident stroke using retinal vasculopathy severity score: The ARIC study

b. Abbreviated Title (Length 26 characters): Retinal vasculopathy predicts stroke

2. Writing Group:
   Writing group members: Michelle P. Lin (first author), Rebecca Gottesman (senior author), A. Richey Sharrett, Ron Klein, Barbara Klein, Jennifer Deal, Others welcome.

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

First author: Michelle P. Lin, MD, MPH
Address: Johns Hopkins University School of Medicine
Department of Neurology
600 N Wolfe Street
Phipps Building, Suite 486
Baltimore, Maryland 21287

Phone: 410-955-2228   Fax: 410-955-0793
E-mail: michelle.py.lin@gmail.com

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: Rebecca Gottesman, MD, PhD
Address: Johns Hopkins University School of Medicine
Department of Neurology
600 N Wolfe Street
Phipps Building, Suite 486
Baltimore, Maryland 21287

Phone: 410-614-2381   Fax: 410-955-0793
E-mail: rgottesm@jhmi.edu

3. Timeline: analysis will be performed following approval of current proposal. We aim to complete manuscript within 3 months after completion of analysis.

4. Rationale:
The retinal blood vessels are accessible to direct non-invasive visualization and have similar anatomical, physiological, and embryological characteristics to small cerebral vessels. Prior studies have shown that pathological changes in the retinal vasculatures (e.g., microaneurysms, retinal hemorrhage, arteriovenous nicking, focal arteriolar narrowing) are associated with clinical and radiographic cerebral microangiopathy (Wong 2002, Ogagarue 2013, ...
Recent *in vivo* data also suggest that retinal vascular patterns may predict pial collateral extent, which is a pivotal modifier of stroke severity and outcomes (Prabhakar 2015). It is therefore conceivable that surveillance retinal photos may enable preclinical risk stratification of cerebrovascular diseases such as stroke and silent cerebrovascular disease, and help identify people who would benefit from early lifestyle changes and preventative therapies, as well as assessing the efficacy of treatment intensity.

While studies have shown that retinal vascular abnormalities including microaneurysms, retinal hemorrhage, arteriovenous nicking, focal arterionarrowing each predict cerebrovascular events, less is known about the utility of these retinal vasculopathy biomarkers in combination. We hypothesize that simultaneous assessment of all 4 retinal vasculopathy biomarkers could provide a simple, preclinical risk stratification schema for incident stroke. We also hypothesize that retinal vasculopathy differs between stroke types, with strongest associations with lacunar strokes, since these reflect microvascular disease. We will test these hypotheses using ARIC visit 3 (1993-1995) retinal imaging data, with cohort followup through 2013.

5. Main Hypothesis/Study Questions:

   Hypothesis 1. We hypothesize that increased severity of retinal vasculopathy is associated with increased risk of stroke, particularly lacunar stroke.

   Hypothesis 2. The addition of retinal vasculopathy improves prediction of incident stroke beyond traditional risk factors (specifically, beyond the ARIC stroke risk score).

   Study questions include: To evaluate the association between severity of retinal vasculopathy (0-4) and incident stroke. To describe retinal vasculopathy pattern across stroke types (hemorrhagic, ischemic, cardioembolic, lacunar, non-lacunar). To evaluate how well additional retinal vasculopathy findings improve prediction of incident stroke beyond traditional risk factors.

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).

   **Participants/ Study population:** ARIC participants who had retinal photography at ARIC visit 3

   **Inclusion:** participants of ARIC visit 3 1993-1995 with retinal photos

   **Exclusion:** prevalent stroke before 1993-1995

   **Outcome Measure:** Incident stroke; in addition to overall stroke (definite/ probable), stroke subtypes will be considered (hemorrhagic, ischemic, cardioembolic, lacunar, non-lacunar).

   **Retinal vasculopathy score** (0-4, 1 point is allotted to each of the below findings)

   Microaneurysm (yes/no)
   Retinal hemorrhage (yes/no)
   Arteriovenous (AV) nicking (yes/no)
   Focal arteriolar narrowing (yes/no)

   **Covariates, to be evaluated from ARIC visit 3:**

   - demographics: age, sex, race-center, level of educational attainment
   - comorbidities: hypertension, diabetes, dyslipidemia, atrial fibrillation, coronary heart disease, left ventricular hypertrophy, chronic kidney disease, peripheral artery disease, smoking, antihypertensive use, antithrombotic use
   - Blood pressure (SBP, DBP)
   - Use of antihypertensive medication (yes/no)
   - Use of anti-thrombotic medication (yes/no)
Analysis Plan
The primary independent variable is retinal vasculopathy severity (0-4), while the outcome variable is incident stroke. The relationship between retinal vasculopathy score (0-4) and outcomes (incident stroke by types) will be explored using 2 models, as will the separate retinal markers. Cox proportional hazards models will be performed to estimate incidence rate, hazard ratios (HRs) and 95% confidence intervals of incident stroke before and after adjusting for covariates. Model 1 is to adjust for sociodemographic (age, sex, race-center, education). Model 2 will adjust for sociodemographics and comorbidities (hypertension, diabetes mellitus, dyslipidemia, coronary heart disease, left ventricular hypertrophy, chronic kidney disease, smoking). We’ll evaluate for a dose-dependent relationship between the number of abnormal retinal vasculopathy markers (between 0 and 4) and outcomes. Separate analyses will also be performed on persons with and without diabetes at visit 3. Models will be repeated evaluating stroke subtypes.

To assess the performance of retinal vasculopathy score in predicting incident stroke (all stroke and by types), and specifically in improving predicting beyond the ARIC model, we will estimate the area under the receiver operating characteristic curve (AUC), and compare AUC across models that contain traditional vascular risk factors and models with addition of retinal vasculopathy scores (0-4). The “ARIC-model” (Chambless 2004) includes age, black race, smoking status, diabetes mellitus, left ventricular hypertrophy, previous coronary heart disease, use of antihypertensive medication, and systolic blood pressure (20mmHg).

Finally, we will test for interaction by antihypertensive and antithrombotic use on the retinal vasculopathy and stroke relationship.

Limitations: Potential limitation to this approach is the loss of quantitative information of each of the retinal vasculopathy measures. Severity of each biomarker may confer different relative risks for individual components of the composite endpoint. The current binary scoring assumes that the risk associated with each positive retinal vasculopathy finding is equivalent, which may not be the case. Nevertheless, using binary cutpoints, however, enables clinicians to integrate data rapidly from all 4 biomarkers into a simple scoring system for fast risk-stratification.

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? _____ 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

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)?
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/

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. Understood.

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.

13. Per Data Use Agreement Addendum, approved manuscripts using CMS data shall be submitted by the Coordinating Center to CMS for informational purposes prior to publication. Approved manuscripts should be sent to Pingping Wu at CC, at pingping_wu@unc.edu. I will be using CMS data in my manuscript ____ Yes _x_ No.

References:


