1.a. Full Title: Estimated glomerular filtration rate upper reference limits and outcomes associated with NT-proBNP and high sensitivity troponin T

b. Abbreviated Title (Length 26 characters): Cardiac biomarkers and kidney disease

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3. **Timeline:**

We plan to complete data analyses by the end of 2016/early 2017. The manuscript will be written and submitted in the Spring of 2017.

4. **Rationale:**

N-terminal pro-B-type natriuretic peptide (NT-proBNP) is secreted from cardiac myocytes in response to myocardial stretch from pressure, volume overload\(^1\) or increasing left ventricular mass.\(^2\)\(^-\)\(^4\) Cardiac troponin T levels rise in response to myocardial injury, remodeling or left ventricular hypertrophy.\(^5\)\(^-\)\(^6\) While NT-proBNP and cTnT levels are widely used for the diagnoses of acute decompensated heart failure and myocardial infarction, respectively, levels of these biomarkers are also strongly predictive of incident cardiovascular events and death in the asymptomatic general population.\(^7\)\(^-\)\(^25\) The utility of troponin T and NT-proBNP for both diagnosis and prognosis among patients with CKD has been limited due to several concerns: elevated concentrations may be due to reduced renal excretion\(^2\)\(^-\)\(^6\)\(^,\)\(^26\)\(^-\)\(^29\) and that other novel CKD-specific pathways to cardiovascular disease\(^30\)\(^,\)\(^31\) may render those reflected by these biomarkers less important. Recent studies in CKD populations have shown strong associations of troponin T and NT-proBNP with adverse clinical outcomes.\(^32\)\(^-\)\(^36\) Despite observational data from a number of epidemiological studies demonstrating a strong link between elevated NT-proBNP and troponin T with adverse outcomes, there is a lack of data on a cut-point to define elevated risk across the spectrum of kidney function.

Our group has previously shown that cut-points defining abnormal troponin T when measured by a highly sensitive assay likely differ by age and sex.\(^37\) A better understanding of the distribution of NT-proBNP and troponin T among those with CKD and across CKD stages, and their associations with adverse outcomes may have important clinical applications for use of these biomarkers in the setting of decreased kidney function. Thus, we propose to estimate the reference range of NT-proBNP and hs cTnT, and the associations of these biomarkers with clinical outcomes among participants with a broad range of kidney function from 5 community based cohorts.

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

1. **To determine the median, IQR, 95\%, and 99\%** percentile upper reference limits for high sensitivity troponin T and NT-proBNP across eGFR categories among patients with CKD using individual patient-level data across 5 large independent cohorts.

2. **To compare the association and discrimination of NT-proBNP and hs-TnT with incident heart failure, coronary heart disease and death among participants with and without chronic kidney disease across eGFR categories.**
We hypothesize that - although the reference range will be higher for those with lower kidney function - associations of each biomarker with adverse outcomes will be similar across the range of kidney function.

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 populations:
Patients with and without CKD without prior CVD (CHD, HF, atrial fibrillation, stroke) in the following cohorts:
1. ARIC
2. DHS
3. CHS
4. MESA
5. CRIC

(Main analysis) All subjects with previously measured NT-proBNP and hs cTnT, with and without CKD, and without prior clinically overt CVD (CHD, HF, atrial fibrillation, stroke) in the following cohorts:
1. ARIC
2. DHS
3. CHS
4. MESA
5. CRIC

(Secondary analysis): All subjects as defined in the main analysis without subclinical CVD (defined as presence of Left Ventricular Hypertrophy and/or depressed LV Ejection Fraction (LVEF<55%).

Data:
Exposures
1. Estimated glomerular filtration rate at baseline will be calculated using baseline serum creatinine measures and the CKD-EPI equation. We will examine categories of eGFR: >90, 60-89, 45-59, 30-44, <30 ml/min/1.73 m2. Chronic Kidney Disease stage 3 or greater will be defined by eGFR_{CKD-EPI}<60 ml/min/1.73m^2
2. NT-proBNP: baseline for MESA, DHS and CRIC, year 5 for CHS, ARIC at year 4
3. High-sensitivity troponin T: baseline for MESA, DHS and CRIC, year 5 for CHS, ARIC at year 4

Outcomes
Study Question #2: Incident heart failure, coronary heart disease, stroke, atrial fibrillation and all-cause mortality (all individual outcomes).

4. Covariates

Demographics: age, sex, race/ethnicity, education, study cohort, smoking, diabetes, hypertension, systolic blood pressure, height, weight, LDL cholesterol, HDL cholesterol, anti-hypertensive medications, statins

ECG (CHS and ARIC)/Echocardiogram (DHS, CRIC)/cardiac MRI (MESA) covariates: left ventricular mass, left ventricular ejection fraction.

(For CHS, we will do a secondary analysis using echocardiogram instead of ECG).

Analytic Plan:

We will begin by comparing characteristics of patients with and without CKD (defined as eGFR<60 ml/min/1.73 m2) across study cohorts.

Aim 1: We will estimate and statistically compare the distribution and 99th centile values (with 95% CI) for hs-TnT and NT-proBNP for each eGFR category (>90, 60-89, 45-59, 30-44, <30 ml/min/1.73 m2 in the pooled 5-cohort study sample as defined above). We will also generate LOWESS curves to explore the association of each cardiac biomarker and eGFR (continuous). In secondary analyses, we will further stratify by age (by decade of age), sex and race/ethnicity (African-American vs. other).37

Aim 2: We will first calculate incidence rates of each CVD event type in each eGFR stratum across quartiles of each biomarker and examine absolute risk differences. We will explore the functional form of the association of each cardiac biomarker with each outcome using restricted cubic splines in each stratum of eGFR. For these analyses, Troponin T levels<LLD will be modeled separately. We will then perform stratified Cox models to test the associations of each biomarker (continuous and in categories [based on distributions in analysis 1 and splines]) with associations of incident heart failure, coronary heart disease, stroke, and death in each strata of eGFR. We will perform a series of nested Cox regression models: (1) unadjusted; (2) adjusting for demographics and study cohort and (3) diabetes, hypertension, systolic blood pressure, body mass index, LDL cholesterol, HDL cholesterol, anti-hypertensive medications, statins.

Next, we will examine the C-statistic and the net reclassification improvement (NRI) for a “base” model in each eGFR category. This base model will include traditional CVD risk factors such as (but not limited to): age, sex, race/ethnicity, diabetes, hypertension, systolic blood pressure, BMI, LDL cholesterol and HDL cholesterol. We will examine the change in C-statistic with the addition of NT-proBNP and hs-TnT for each eGFR category. These ΔC-statistics and NRIs will be compared across eGFR categories to quantitatively
compare the population-level risk reclassification and discrimination of each biomarker for predicting incident CV events and mortality.

In sensitivity analyses, we will further exclude participants with evidence of subclinical CVD – by echocardiogram, ECG, or cardiac MRI- and repeat our main analyses.

In exploratory analyses, we will try to define risk “thresholds” of each biomarker for each outcome of interest by eGFR strata using restricted cubic splines and AUC models.

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

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? __x__ Yes _____ No

11.b. If yes, is the proposal

        __x__ 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.