ARIC Manuscript Proposal #2817

PC Reviewed: 8/9/16 **Status: _____** **Priority: 2**
SC Reviewed: _________ **Status: _____** **Priority: ____**

1.a. Full Title: Lifestyle-related health behaviors and six-year change in high-sensitivity cardiac troponin T

b. Abbreviated Title (Length 26 characters): **Health Behaviors and Troponin T**

2. Writing Group:
Writing group members:

Anna Fretz; John McEvoy; Casey Rebholz; Chiadi Ndumele; Christie Ballantyne; Liz Selvin.. others welcome

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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3. Timeline: Data is currently available. Analysis is planned to start as soon as approval is obtained and will take between 3 and 6 months. Manuscript will be prepared during the 3 months following the completion of the analysis.

4. Rationale:

Cardiac troponin T is a widely-used biomarker in clinical practice for diagnosing myocardial infarction in persons with chest pain. A novel highly sensitive assay for cardiac troponin T can detect concentrations of troponin 10-times lower than assays currently used in clinical practice, extending the potential utility of this biomarker to asymptomatic populations. Previous studies have demonstrated that roughly 70% of a healthy middle-aged population have detectable troponin levels using this assay (1-3). Having detectable levels of high-sensitivity cardiac troponin T (hs-cTnT) has been shown to strongly and independently predict cardiovascular morbidity and mortality (2-6). Moreover, in studies using serial measures of hs-cTnT, individuals with increases in hs-cTnT over time demonstrate greater risk of cardiovascular events relative to individuals with no significant change (5-7). Additionally, individuals with decreases in hs-cTnT over time demonstrate lower risk of cardiovascular events than those with no significant change (6,7). Given the prognostic utility of serial measures of hs-cTnT, it is important to understand determinants that may be associated with increased or decreased subclinical myocardial damage over time.

Only one previous study has explored clinical factors associated with temporal changes in hs-cTnT (8). This study demonstrated that hypertension, obesity and diabetes were most strongly associated with progression of subclinical myocardial damage. It also demonstrated that among modifiable clinical risk factors, individuals with obesity and diabetes were less likely to have temporal decreases in hs-cTnT. Because hypertension, obesity and diabetes are strongly influenced by lifestyle and environmental factors, the impact of health behaviors on temporal changes in hs-cTnT is of substantial clinical interest. To our knowledge, only two previous studies have addressed a component of this question, by examining the association between physical activity and temporal increases in hs-cTnT (9,10). In these studies, low levels of physical activity were indeed associated with progression of subclinical myocardial damage.

However, it is important to better understand other modifiable health behaviors that are associated with temporal reductions in hs-cTnT, as this has potential utility for monitoring the beneficial impacts of health behaviors on CVD risk through regression of myocardial damage during the subclinical period.

There are five key health behaviors, highlighted by the American Heart Association, recommended for the prevention of cardiovascular disease: physical activity, diet, weight management, smoking cessation and emotional wellbeing (11). There is exhaustive literature on the cardiovascular benefits of these health behaviors. With the exception of physical activity that has already been studied (9,10), our study will focus on diet, adiposity, smoking, alcohol consumption and emotional well-being as potential lifestyle risk factors for subclinical myocardial damage. We will also assess health behaviors modeled in aggregate, using the American Heart Association Life's Simple 7 score, which has shown that a higher health score is associated with reduced risk of cardiovascular disease (12).

Diet is a well-studied risk factor for cardiovascular disease. Previous studies show that having a healthy diet or improving diet over time is associated with decreased risk of cardiovascular events (13-18) perhaps through improved endothelial function and reduced levels of inflammatory markers and oxidized lipids, which contribute to vascular disease (19-21).

Adiposity is strongly influenced by diet, but has been independently studied as a risk factor for cardiovascular disease and mortality, both in terms of excess weight (22-24) and underweight (25). Previous studies also demonstrate that weight gain is associated with increased CVD risk, compared to stable weight (26-30). However, data are conflicted on the association of weight loss and cardiovascular risk, with some studies showing increased CVD risk (29,30), some showing no association (26-28) and some showing decreased mortality, only when weight loss is intentional (31,32). It is hypothesized that the data are conflicted due to confounding factors such as comorbidities and illness that could cause weight loss, but also lead to increased risk of cardiovascular events and mortality. However from a pathophysiologic standpoint, weight loss is associated with improved cardiometabolic markers (33-36), reduced blood pressure (37,38) and reduced cardiac wall stress (39,40).

Smoking cessation is associated with rapid reduction in risk of cardiac events (41-43), but its mechanism is less well-studied than the risks associated with active smoking. The literature suggests that there are improvements in endothelial function after just one year (44). It is also hypothesized that many of the negative impacts of cigarette smoking, such as increased sympathetic activity of the heart (45), increased inflammatory markers (46,47) and inhibition of thrombolytic processes (47), are simply reversed with cessation (48).

Poor **emotional wellbeing** and psychological stress have been assessed in population-based studies as vital exhaustion using the well-established Maastricht Vital Exhaustion Questionnaire (49,50). High levels of baseline vital exhaustion are independently associated with increased coronary events (51-54) as well as microvascular disease

(55). The literature suggests that this risk could be mediated by reduced cardiac parasympathetic activity (56) as well as impaired thrombolysis (57,58).

Lastly, moderate **alcohol consumption**, although not part of the core AHA prevention guidelines, is a known health behavior that demonstrates cardiovascular protective effects (59-64). The mechanisms by which there is decreased risk are hypothesized to be: improved endothelial function (65,66), decreased inflammation of the endothelium and myocardium (67,68), reduced fibrinogen levels and platelet aggregation leading to improved hemodynamics (69-73), as well as inhibition of oxidation of lipids (74,75).

Given the associations between these health behaviors and cardiovascular pathology, as well as clinical cardiovascular outcomes, it could be of clinical utility to understand their association with changes in hs-cTnT, specifically temporal reductions, to assess for regression in myocardial damage during the subclinical stage.

5. Main Hypothesis/Study Questions:

Aim 1. To evaluate the association between baseline health behaviors (healthy diet score, moderate alcohol consumption, smoking status, body mass index (BMI) and vital exhaustion) and six-year categorical changes in hs-cTnT. These associations will be evaluated before and after adjustment for demographic and traditional cardiovascular risk factors.

> Hypothesis: Healthier behaviors will be positively associated with incident undetectable hs-cTnT (<5 ng/L) among persons with detectable hs-cTnT (≥5ng/L) at baseline, compared to those who had sustained detectable hs-cTnT levels. We will also conduct analyses assessing risk of incident detectable hs-cTnT (≥5ng/L) among those with undetectable levels (<5ng/L) at visit 2, which we hypothesize to have a negative association.

Aim 2. To evaluate the association between health behaviors (healthy diet score, alcohol consumption, smoking status, body mass index (BMI) and vital exhaustion), treated as time-varying exposure variables, and six-year categorical change in hs-cTnT. These associations will be evaluated before and after adjustment for demographic and traditional cardiovascular risk factors.

> Hypothesis: Increased duration of exposure to better health behaviors will be positively associated with incident undetectable hs-cTnT among those with detectable hs-cTnT at baseline compared to those who had sustained detectable hs-cTnT levels. We will also conduct analyses assessing risk of incident detectable hs-cTnT among those with undetectable hs-cTnT at baseline, which we hypothesize to be a negative association.

Aim 3. To evaluate the association between Life's Simple 7 score and six-year categorical change in hs-cTnT.

Hypothesis: A higher score (indicative of healthier behaviors) will be positively associated with incident undetectable hs-cTnT (<5ng/L) among persons with detectable hs-cTnT (≥5ng/L) at baseline, compared to individuals with sustained detectable hs-cTnT levels.

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 Design: Prospective analysis to measure the association of health behaviors and Life's Simple 7 score with changes in hs-cTnT, measured at two time points, six years apart (visit 2: 1990-1992 and visit 4: 1996-1998). Visit 2 will serve as baseline for the study. We will also assess health behaviors as time-varying exposures, using information from visit 3 and visit 4.

Study Population (Inclusion/Exclusion Criteria):
All ARIC participants who attended visits 2 and 4 and who did not meet any of the following exclusion criteria:
-MI, angina, stroke, or revascularization before visit 2
-Missing hs-cTnT value at visits 2 or 4
-Missing drinking status, smoking status, diet information, BMI, or vital exhaustion score at visit 2
-Missing any component of the Life's Simple 7 score (BMI, physical activity score, diet information, smoking status, total cholesterol, fasting blood glucose, blood pressure)
-Identify as non-white race in Minnesota or Maryland
-Identify as non-black race in Mississippi

Exposure:
There are five health behaviors that will be assessed: physical activity, diet, smoking status, alcohol consumption, weight loss, and emotional well-being. We will also assess Life's Simple 7 score, which is composed of: total cholesterol, fasting blood glucose, blood pressure, smoking, body mass index, physical activity, and diet.

Diet will be measured using a healthy food score, adapted from a study by Rebholz et al (76). The score will be a sum of 5 individual food groups obtained from the Food Frequency Questionnaire (FFQ): fruits and vegetables, fish, fiber-rich whole gains, sodium, and sugar-sweetened beverages. A point will be given for each group where recommended intake levels were met: (1) ≥4.5 servings of fruits and vegetables per day; (2) ≥7 ounces of fish per week; (3) ≥3 ounces of fiber-rich whole grains per day

(≥1.1 g of dietary fiber/10 g of carbohydrate per day); (4) <1500 mg of sodium per day; and (5) ≤36 ounces of sugar-sweetened beverages per week. Thus, a higher score indicates a healthier diet. This score demonstrates consistency with AHA scientific statements (77, 78) as well as the US Dietary Guidelines (79).

Smoking Status will be measured by self-reported current, former or never user.

Alcohol Consumption will be measured by self-reported drinking status and number of drinks per week. Alcohol consumption will be divided as never drinker, former drinker, current drinker up to 7 drinks per week, current drinker 7-14 drinks per week, current drinker 14-21 drinks per week, and current drinker greater than 21 drinks per week.

Body Mass Index will be measured using height and weight, assessed by standard protocol.

Emotional Well-being will be assessed using Vital Exhaustion, measured by the Maastricht Questionnaire. This questionnaire consists of 21 questions with a higher score corresponding to greater level of vital exhaustion and poorer emotional well-being. A score of ≥14 is considered the threshold for clinically evident vital exhaustion and thus is the threshold for our analyses (80).

Life's Simple 7 score will be determined by summing the number of the 7 individual health components that are achieved at the ideal level. Ideal levels of the health components were: healthy diet score ≥4, body mass index <25 kg/m^2, ≥ 150 minutes/week of physical activity, never smoker or quit >12 months ago, blood pressure <120/80 mmHg, fasting blood glucose <100mg/dL, and total cholesterol <200 mg/dL.

Outcome:

Cardiac troponin T was measured using a highly-sensitive novel (pre-commercial) assay, Elecsys Troponin T (Roche Diagnostics, Indianapolis, Indiana). For the purposes of these analyses, we will define 5.0 ng/L as the lower limit of detection (81,82) although we will conduct sensitivity analyses using the lowest level of measurability (3.0 ng/L). The assay demonstrated between-assay coefficients of 2.6% for control materials with mean cTnT concentration of 2378 ng/L and 6.9% for control materials with mean cTnT concentration of 29 ng/L. Laboratory reliability was 0.99 in participants with HF and 0.94 in participants without HF (83); reliability coefficient was assessed to be r=0.98 with a coefficient of variation of 15% after excluding outliers of greater than SD from the mean. Serum samples were drawn at visit 2 and plasma samples were drawn at visit 4. These samples were stored at -80° until time of assay, which was 2011 for visit 4 plasma samples and 2013 for visit 2 serum samples. A formal calibration study demonstrated comparability between visit 2 serum and visit 4 plasma measurements (84). Detectable hs-cTnT will be defined as concentrations ≥5ng/L; undetectable hs-cTnT will be defined as concentrations<5ng/L.

Covariates:

Age (years, continuous), sex (male/female), race/field center (Maryland whites; Minnesota whites, North Carolina whites; North Carolina blacks; Mississippi blacks), education (years of education), diabetes (yes/no), total cholesterol (continuous), HDL cholesterol (continuous), systolic and diastolic blood pressure (continuous), blood pressure medication use (yes/no), eGFR (mL/min/1.73 m2), LV hypertrophy (yes/no). All measured at baseline visit 2, unless otherwise noted.

Statistical Analysis:
For the primary analysis, we will look at hs-cTnT as a binary outcome. The primary outcome will be incident undetectable hs-cTnT (<5ng/L) at visit 4 among persons with detectable hs-cTnT (≥5ng/L) at visit 2, using those who had sustained detectable hs-cTnT at visit 4 as the reference group. For these binary outcomes we will use Poisson regression to generate adjusted risk ratios.

We will perform two statistical models:

- Model 1: adjust for demographic factors (age, gender, race/field center [only if not a modifier], education level)
- Model 2: Model 1 + additional cardiovascular risk factors (total cholesterol, HDL cholesterol, systolic and diastolic blood pressure, blood pressure medication use, diabetes, eGFR, LVH)

Sensitivity Analyses:
We will perform 4 sensitivity analyses:

1) Sensitivity analysis excluding individuals with a cardiovascular event between visits 2 and 4 (MI, HF).
2) Sensitivity analysis for the prospective association of incident non-elevated hs-cTnT (<14 ng/L) among those with elevated hs-cTnT (≥14ng/L) at visit 2, using those with sustained elevation of hs-cTnT as the reference group.
3) Sensitivity analysis imputing visit 4 hs-cTnT as 60ng/L for those who died between visit 2 and 4.
4) Sensitivity analysis looking at continuous change in hs-cTnT among those with detectable hs-cTnT at visit 2.
5) Sensitivity analysis using data only from North Carolina to assess race-field center aliasing.

Potential effect modifiers:
We will test for interaction by age, race, and sex. A stratified analysis will be performed if statistically significant effect modification for any of these variables is observed.

Limitations:

We are limited by the two measurements of hs-cTnT at visit 2 and then visit 4, which are six years apart, as a means of characterizing the progression or regression of subclinical myocardial injury. There may be more detailed trends in changes of hs-cTnT that we are unable to assess. The use of Vital Exhaustion as a variable to measure emotional well-being is not ideal, but has been shown to be of use in predicting cardiovascular outcomes (51). Lastly, because this is an observational study only, we cannot rule out the potential of residual confounding.

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)?

MS #2128: Six-year change in high sensitivity cardiac troponin T and risk of cardiovascular events (Selvin)

MS #2441: Obesity, Physical Activity and Myocardial Injury: The Atherosclerosis Risk in Communities (ARIC) Study

MS #2269: Risk factors for Progression of Subclinical Myocardial Injury: Six-year change in highly-sensitive troponin T in a community-based population study (McEvoy)

MS #2025: Obesity and Subclinical Myocardial Injury: The Atherosclerosis Risk in Communities (ARIC) Study (Ndumele)

MS #2442: Alcohol consumption and myocardial biomarkers

11.a. Is this manuscript proposal associated with any ARIC ancillary studies or use any ancillary study data? _X_ Yes ____ No

ARIC Ancillary Study #2008.10: Measurement of NT-pro-BNP and troponin T at visit 4 for the full ARIC cohort (Ballantyne)

ARIC Ancillary Study #2009.16: Short-term Markers of Glycemia and Long-term Outcomes (Selvin)

11.b. If yes, is the proposal
_X_ A. primarily the result of an ancillary study (list number* #2008.10, #2009.16)
___ 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.

References:

1. Sherwood MW, Newby LK. High-sensitivity troponin assays: evidence, indications, and reasonable use. J. Am. Heart Assoc 2014;3(1): e000403.
2. Saunders JT, Nambi V, de Lemos JA et al. Cardiac Troponin T Measured by a Highly Sensitive Assay Predicts Coronary Heart Disease, Heart Failure, and Mortality in the Atherosclerosis Risk in Communities Study. Circulation 2011; 123:1367-76.
3. Eggers KM, Al-Shakarchi J, Berglund L et al. High-sensitive cardiac troponin T and its relations to cardiovascular risk factors, morbidity, and mortality in elderly men. Am Heart J. 2013; 166(3): 541-8.
4. de Lemos JA, Drazner MH, Omland T et al. Association of troponin T detected with a highly sensitive assay and cardiac structure and mortality risk in the general population. JAMA 2010; 304(22): 2503-12.
5. Hussein AA, Gottdiener JS, Bartz TM et al. Cardiomyocyte injury as assessed by a highly sensitive troponin assay and sudden cardiac death in the community: The Cardiovascular Health Study. J. Am. Coll. Cardiol. 2013; 62(22): 2112-20.
6. deFilippi CR, de Lemos JA, Christenson RH et al. Association of serial measures of cardiac troponin T using a sensitive assay with incident heart failure and cardiovascular mortality in older adults. JAMA 2010;304:2494 –502.
7. McEvoy JW, Chen Y, Ndumbele CE et al. Six-Year Change in High-Sensitivity Cardiac Troponin T and Risk of Subsequent Coronary Disease, Heart Failure, and Death. JAMA Cardiol. Published online June 08, 2016. doi:10.1001/jamacardio.2016.0765.
8. McEvoy JW, Lazo M, Chen Y et al. Patterns and determinants of temporal change in high-sensitivity cardiac troponin-T: The Atherosclerosis Risk in Communities Cohort Study. Int J Cardiol 2015;187;651-7.
9. deFilippi CR, de Lemos JA, Tkaczuk AT et al. Physical Activity, Change in Biomarkers of Myocardial Stress and Injury, and Subsequent Heart Failure Risk in Older Adults. JACC 2012;60(24): 2539-47.
10. deFilippi JR, de Lemos JA, Newman AB et al. Impact of moderate physical activity on the longitudinal trajectory of a cardiac specific biomarker of injury: Results from a randomized pilot study of exercise intervention. American Heart Journal 2016; 179: 151-156.
11. Healthy Living. American Heart Association. 2016.
12. Folsom AR, Yatsuya H, Nettleton JA et al. Community prevalence of ideal cardiovascular health, by the American Heart Association definition, and

relationship with cardiovascular disease incidence. J Am Coll Cardiol 2011; 57(16);1690-6.

13. Estruch R, Ros E, Salas-Salvadó J, et al. Primary prevention of cardiovascular disease with a Mediterranean diet. N Engl J Med 2013; 368:1279.
14. Sotos-Prieto M, Bhupathiraju SN, Mattei J, et al. Changes in Diet Quality Scores and Risk of Cardiovascular Disease Among US Men and Women. Circulation 2015; 132:2212.
15. Reedy J, Krebs-Smith SM, Miller PE et al. Higher diet quality is associated with decreased risk of all-cause cardiovascular disease, and cancer mortality among older adults. J Nutr 2014;144(6);881-9.
16. Fung TT, Rexrode KM, Mantzoros CS et al. Mediterranean Diet and Incidence of and Mortality From Coronary Heart Disease and Stroke in Women. Circulation 2009; 119; 1093-1100
17. Trichopoulou A, Costacou T, Bamia C, Trichopoulos D. Adherence to a Mediterranean diet and survival in a Greek population. *N Engl J Med.* 2003; 348: 2599–2608.
18. McCullough ML, Feskanich D, Stampfer MJ, Giovannucci EL, Rimm EB, Hu FB, Spiegelman D, Colditz GA, Hunter DJ, Willett WC. Diet quality and major chronic disease risk in men and women: moving toward improved dietary guidance. *Am J Clin Nutr.* 2002;76(6): 1261–1271.
19. Fung TT, McCullough ML, Newby PK, Manson JE, Meigs JB, Rifai N, Willett WC, Hu FB. Diet-quality scores and plasma concentrations of markers of inflammation and endothelial dysfunction. *Am J Clin Nutr.* 2005; 82: 163–173
20. Dai J, Miller AH, Bremner JD, Goldberg J, Jones L, Shallenberger L, Buckham R, Murrah NV, Veledar E, Wilson PW, Vaccarino V. Adherence to the Mediterranean diet is inversely associated with circulating interleukin-6 among middle-aged men: a twin study. *Circulation.* 2008; 117: 169–175
21. Esposito K, Marfella R, Ciotola M, Di Palo C, Giugliano F, Giugliano G, D'Armiento M, D'Andrea F, Giugliano D. Effect of a Mediterranean-style diet on endothelial dysfunction and markers of vascular inflammation in the metabolic syndrome: a randomized trial. *JAMA.*2004; 292: 1440–1446.
22. Global Burden of Metabolic Risk Factors for Chronic Diseases Collaboration (BMI Mediated Effects), Lu Y, Hajifathalian K, et al. Metabolic mediators of the effects of body-mass index, overweight, and obesity on coronary heart disease and stroke: a pooled analysis of 97 prospective cohorts with 1·8 million participants. Lancet 2014; 383:970.
23. Kenchaiah S, Evans JC, Levy D, et al. Obesity and the risk of heart failure. N Engl J Med 2002; 347:305.
24. Stevens J, Erber E, Truesdale KP et al. Long- and Short-term Weight Change and Incident Coronary Heart Disease and Ischemic Stroke: The Atherosclerosis Risk in Communities Study. Am J Epidemiol 2013; 178(2);239-248.
25. Lavie CJ, McAuley PA, Church TS, et al. Obesity and cardiovascular diseases: implications regarding fitness, fatness, and severity in the obesity paradox. J Am Coll Cardiol. 2014; 63(14): 1345-54.

26. Willett WC, Manson JE, Stampfer MJ, et al. Weight, weight change, and coronary heart disease in women. Risk within the 'normal' range.JAMA. 1995;273(6):461–465.
27. Rimm EB. Body size and fat distribution as predictors of coronary heart disease among middle-aged and older US men. Am J Epidemiol.1995;141(12):1117–1127.
28. Galanis DJ, Harris T, Sharp DS, et al. Relative weight, weight change, and risk of coronary heart disease in the Honolulu Heart Program. Am J Epidemiol. 1998;147(4):379–386.
29. Rosengren A, Wedel H, Wilhelmsen L. Body weight and weight gain during adult life in men in relation to coronary heart disease and mortality: a prospective population study. Eur Heart J. 1999;20(4):269–277.
30. Chei CL, Iso H, Yamagishi K, et al. Body mass index and weight change since 20 years of age and risk of coronary heart disease among Japanese: the Japan Public Health Center-Based Study. Int J Obes (Lond) 2008;32(1):144–151.
31. Gregg EW, Gerzoff RB, Thompson TJ, et al. Intentional weight loss and death in overweight and obese U.S. adults 35 years of age and older. Ann Intern Med. 2003;138(5):383–389.
32. Williamson DF, Thompson TJ, Thun M, et al. Intentional weight loss and mortality among overweight individuals with diabetes.Diabetes Care. 2000;23(10):1499–1504.
33. Janiszewski PM, Ross R. Effects of weight loss among metabolically healthy obese men and women. *Diabetes Care* 2010; 33: 1957–1959.
34. Shin MJ, Hyun YJ, Kim OY, Kim JY, Jang Y, Lee JH. Weight loss effect on inflammation and LDL oxidation in metabolically healthy but obese (MHO) individuals: low inflammation and LDL oxidation in MHO women. *Int J Obes (Lond)* 2006; 30: 1529–1534.
35. Rosito GA, D'Agostino RB, Massaro J, et al. Association between obesity and a prothrombotic state: the Framingham Offspring Study. Thromb Haemost 2004; 91:683.
36. Davì G, Guagnano MT, Ciabattoni G, et al. Platelet activation in obese women: role of inflammation and oxidant stress. JAMA 2002; 288:2008.
37. Horvath K, Jeitler K, Siering U, et al. Long-term effects of weight-reducing interventions in hypertensive patients: systematic review and meta-analysis. Arch Intern Med 2008; 168:571.
38. Douketis JD, Macie C, Thabane L, Williamson DF. Systematic review of long-term weight loss studies in obese adults: clinical significance and applicability to clinical practice. Int J Obes (Lond) 2005; 29:1153.
39. Klein S, Burke LE, Bray GA, et al. Clinical implications of obesity with specific focus on cardiovascular disease: a statement for professionals from the American Heart Association Council on Nutrition, Physical Activity, and Metabolism: endorsed by the American College of Cardiology Foundation. Circulation 2004; 110:2952.
40. Poirier P, Giles TD, Bray GA, et al. Obesity and cardiovascular disease: pathophysiology, evaluation, and effect of weight loss. Arterioscler Thromb Vasc Biol 2006; 26:968.

41. Rose G, Hamilton PJ, Colwell L, Shipley MJ. A randomised controlled trial of anti-smoking advice: 10-year results. J Epidemiol Community Health 1982; 36:102.
42. Multiple risk factor intervention trial. Risk factor changes and mortality results. Multiple Risk Factor Intervention Trial Research Group. JAMA 1982; 248:1465.
43. Hjermann I, Velve Byre K, Holme I, Leren P. Effect of diet and smoking intervention on the incidence of coronary heart disease. Report from the Oslo Study Group of a randomised trial in healthy men. Lancet 1981; 2:1303.
44. Johnson HM, Gossett LK, Piper ME, et al. Effects of smoking and smoking cessation on endothelial function: 1-year outcomes from a randomized clinical trial. J Am Coll Cardiol 2010; 55:1988.
45. Grassi G, Seravalle G, Calhoun DA et al. Mechanisms responsible for sympathetic activation by cigarette smoking in humans. Circulation 1994; 90(1); 248-53.
46. Celermajer DS, Sorensen KE, Georgakopoulos D, et al. Cigarette smoking is associated with dose-related and potentially reversible impairment of endothelium-dependent dilation in healthy young adults. Circulation 1993; 88:2149.
47. Ambrose JA, Barua RS. The pathophysiology of cigarette smoking and cardiovascular disease: an update. J Am Coll Cardiol 2004;43(10);1731-7.
48. McEvoy JW, Nasir K, DeFillippis AP et al. Relationship of cigarette smoking with inflammation and subclinical vascular disease: the Multi-Ethnic Study of Atherosclerosis. Arterioscler Thromb Vasc Biol 2015; 35(4);1002-10.
49. Appels A, Höppener P, Mulder P. A questionnaire to assess premonitory symptoms of myocardial infarction. Int J Cardiol. 1987;17:15–24.
50. Appels A, Mulder P. Excess fatigue as a precursor of myocardial infarction. Eur Heart J. 1988;9:758–764.
51. Williams JE, Mosley TH, Jr, Kop WJ, Couper DJ, Welch VL, Rosamond WD. Vital exhaustion as a risk factor for adverse cardiac events (from the Atherosclerosis Risk In Communities [ARIC] study). Am J Cardiol.2010;105:1661–1665.
52. Cole SR, Kawachi I, Sesso HD, Paffenbarger RS, Lee IM. Sense of exhaustion and coronary heart disease among college alumni. Am J Cardiol.1999;84:1401–1405.
53. Appels A, Falger PR, Schouten EG. Vital exhaustion as risk indicator for myocardial infarction in women. J Psychosom Res 1993;37:881–890.
54. Schuitemaker GE, Dinant GJ, van der Pol GA, Appels A. Assessment of vital exhaustion and identification of subjects at increased risk of myocardial infarction in general practice. Psychosomatics 2004;45:414–418.
55. Cheung N, Rogers S, Mosley TH, Klein R, Couper D, Wong TY. Vital exhaustion and retinal microvascular changes in cardiovascular disease: atherosclerosis risk in communities study.. Psychosom Med 2009; 71(3):308-12
56. Watanabe T, Sugiyama Y, Sumi Y, Watanabe M, Takeuchi K, Kobayashi F, Kono K. Effects of vital exhaustion on cardiac autonomic nervous functions assessed by heart rate variability at rest in middle-aged male workers. Int J Behav Med. 2002;9:68–75.

57. Kop WJ, Hamulyak K, Pernot C, Appels A. Relationship of blood coagulation and fibrinolysis to vital exhaustion. Psychosom Med. 1998;60:352–8.
58. von Känel R, Frey K, Fischer JE. Independent relation of vital exhaustion and inflammation to fibrinolysis in apparently healthy subjects. Scand Cardiovasc J. 2004;38:28–32.
59. O'Keefe JH, Bybee KA, Lavie CJ. Alcohol and cardiovascular health: the razor-sharp double-edged sword. J Am Coll Cardiol 2007; 50:1009.
60. Ronksley PE, Brien SE, Turner BJ, et al. Association of alcohol consumption with selected cardiovascular disease outcomes: a systematic review and meta-analysis. BMJ 2011; 342:d671.
61. Rimm EB, Giovannucci EL, Willett WC, et al. Prospective study of alcohol consumption and risk of coronary disease in men. Lancet 1991; 338:464.
62. Stampfer MJ, Colditz GA, Willett WC, et al. A prospective study of moderate alcohol consumption and the risk of coronary disease and stroke in women. N Engl J Med 1988; 319:267.
63. Bryson CL, Mukamal KJ, Mittleman MA, et al. The association of alcohol consumption and incident heart failure: the Cardiovascular Health Study. J Am Coll Cardiol 2006; 48:305.
64. Walsh CR, Larson MG, Evans JC, et al. Alcohol consumption and risk for congestive heart failure in the Framingham Heart Study. Ann Intern Med 2002; 136:181.
65. Xia L, Wang XX, Hu XS, et al. Resveratrol reduces endothelial progenitor cells senescence through augmentation of telomerase activity by Akt-dependent mechanisms. Br J Pharmacol 2008; 155:387.
66. Hamed S, Alshiek J, Aharon A, et al. Red wine consumption improves in vitro migration of endothelial progenitor cells in young, healthy individuals. Am J Clin Nutr 2010; 92:161.
67. Volpato S, Pahor M, Ferrucci L, et al. Relationship of alcohol intake with inflammatory markers and plasminogen activator inhibitor-1 in well-functioning older adults: the Health, Aging, and Body Composition study. Circulation 2004; 109:607.
68. Albert MA, Glynn RJ, Ridker PM. Alcohol consumption and plasma concentration of C-reactive protein. Circulation 2003; 107:443.
69. Renaud SC, Beswick AD, Fehily AM, et al. Alcohol and platelet aggregation: the Caerphilly Prospective Heart Disease Study. Am J Clin Nutr 1992; 55:1012.
70. Lacoste L, Hung J, Lam JY. Acute and delayed antithrombotic effects of alcohol in humans. Am J Cardiol 2001; 87:82.
71. Landolfi R, Steiner M. Ethanol raises prostacyclin in vivo and in vitro. Blood 1984; 64:679.
72. Meade TW, Chakrabarti R, Haines AP, et al. Characteristics affecting fibrinolytic activity and plasma fibrinogen concentrations. Br Med J 1979; 1:153.
73. Folsom AR, Wu KK, Davis CE, et al. Population correlates of plasma fibrinogen and factor VII, putative cardiovascular risk factors. Atherosclerosis 1991; 91:191.
74. Frankel EN, Kanner J, German JB, et al. Inhibition of oxidation of human low-density lipoprotein by phenolic substances in red wine. Lancet 1993; 341:454.

75. Miyagi Y, Miwa K, Inoue H. Inhibition of human low-density lipoprotein oxidation by flavonoids in red wine and grape juice. Am J Cardiol 1997; 80:1627.
76. Rebholz CM, Anderson CAM, Grams ME et al. Relationship of the American Heart Association's impact Goals (Life's Simple 7) With Risk of Chronic Kidney Disease: Results from the Atherosclerosis Risk in Communities (ARIC) Cohort Study. J Am Heart Assoc 2016;5; e003192.
77. American Heart Association Nutrition Committee, Lichtenstein AH, Appel LJ, Brands M et al. Diet and lifestyle recommendations revision 2006: a scientific statement from the American Heart Association Nutrition Committee. Circulation 2006; 114; 82-96.
78. Johnson RK, Appel LJ, Brands M et al. Dietary Sugars Intake and Cardiovascular Health: A Scientific Statement from the American Heart Association. Circulation 2009; 120; 1011-20.
79. US Department of Health and Human Services and US Department of Agriculture, 2005. 6th Edition. Washington, DC. U.S. Government Printing Office, 2005.
80. Appels A, Höppener P, Mulder P. A questionnaire to assess premonitory symptoms of myocardial infarction. Int J Cardiol. 1987;17:15–24
81. Armbruster, D.A. and T. Pry, *Limit of Blank, Limit of Detection and Limit of Quantitation.* Clinical Biochemistry Review, 2008. 29: p. S49-52.
82. Diagnostics, Roche., *Troponin T hs: Elecsys and cobas e analyzers* 2011, Roche: Mannheim, Germany.
83. Agarwal, S.K., et al., *Sources of variability in measurements of cardiac troponin T in a community-based sample: the atherosclerosis risk in communities study.* Clin Chem, 2011. 57(6): p. 891-7.
84. Parrinello CM Grams ME, Couper D et al. Recalibration of blood analytes over 25 years in the Atherosclerosis Risk in Communities Study: impact of recalibration on chronic kidney disease prevalence and incidence*.* Clin Chem 2015; 61(7); 938-47.