# CARDIA Y20 EXAM VII
## PROTOCOL
### TABLE OF CONTENTS

<table>
<thead>
<tr>
<th>Section</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>A. INTRODUCTION</td>
<td>3</td>
</tr>
<tr>
<td>B. STUDY OBJECTIVES</td>
<td>4</td>
</tr>
<tr>
<td>C. EXAM COMPONENTS</td>
<td>6</td>
</tr>
<tr>
<td>D. STUDY DESIGN</td>
<td>6</td>
</tr>
<tr>
<td>1. Population</td>
<td>7</td>
</tr>
<tr>
<td>2. Informed Consent</td>
<td>7</td>
</tr>
<tr>
<td>3. Data Collected</td>
<td>8</td>
</tr>
<tr>
<td>4. Comparability Studies</td>
<td>9</td>
</tr>
<tr>
<td>E. RATIONALE AND METHODS</td>
<td>12</td>
</tr>
<tr>
<td>1. Racial Differences</td>
<td>12</td>
</tr>
<tr>
<td>2. CAC Measurement</td>
<td>14</td>
</tr>
<tr>
<td>3. Carotid IMT Measurement</td>
<td>16</td>
</tr>
<tr>
<td>4. Blood Pressure</td>
<td>17</td>
</tr>
<tr>
<td>5. Obesity and Anthropometry</td>
<td>18</td>
</tr>
<tr>
<td>6. Diet</td>
<td>21</td>
</tr>
<tr>
<td>7. Laboratory Measures</td>
<td>22</td>
</tr>
<tr>
<td>a. Glycemia</td>
<td>22</td>
</tr>
<tr>
<td>b. Lipids</td>
<td>24</td>
</tr>
<tr>
<td>c. Serum Creatinine</td>
<td>24</td>
</tr>
<tr>
<td>d. Inflammatory Markers</td>
<td>25</td>
</tr>
<tr>
<td>e. Urinary Albumin Excretion</td>
<td>27</td>
</tr>
<tr>
<td>8. Pulmonary Function</td>
<td>30</td>
</tr>
<tr>
<td>9. Psychosocial Measures</td>
<td>31</td>
</tr>
<tr>
<td>10. Sleep</td>
<td>32</td>
</tr>
<tr>
<td>11. Medical History, Hospitalizations and Medications</td>
<td>33</td>
</tr>
<tr>
<td>12. Physical Activity</td>
<td>36</td>
</tr>
<tr>
<td>13. Genetic Analyses</td>
<td>37</td>
</tr>
</tbody>
</table>
F. EXAM IMPLEMENTATION ........................................................................................................... 43
   1. Recruitment ........................................................................................................................ 43
   2. Exam Flow ......................................................................................................................... 44
   3. Prioritization Schedule ...................................................................................................... 45
   4. Quality Control .................................................................................................................. 46
   5. Referrals and Results Reporting ......................................................................................... 48
G. DATA MANAGEMENT ................................................................................................................. 49
   1. Flow and management of Data Forms ............................................................................... 50
   2. Electronic Storage and Management of Data ................................................................. 52
   3. Field Center System .......................................................................................................... 52
   4. Data Entry at the Coordinating Center ........................................................................... 55
   5. Data Transfer System ........................................................................................................ 57
H. STUDY ORGANIZATION ............................................................................................................ 58
   1. NHLBI ............................................................................................................................... 58
   2. OSMB ................................................................................................................................ 58
   3. CARDIA Coordinating Center ......................................................................................... 59
   4. CARDIA Field Centers ...................................................................................................... 60
   5. Reading Centers .............................................................................................................. 61
   6. Laboratories ..................................................................................................................... 62
   7. Role and Composition of Steering Committee ................................................................. 62
   8. Role and Composition of Executive Committee .............................................................. 64
   9. Role and Composition of Emerging Science Committee ............................................... 64
   10. Role and Composition of Subcommittees ...................................................................... 65
I. TIME TABLE .............................................................................................................................. 76
J. REFERENCES ............................................................................................................................. 77
A. INTRODUCTION

The Coronary Artery Risk Development in (Young) Adults (CARDIA) Study was initiated in 1984 by the National Heart, Lung, and Blood Institute to assist in providing a better understanding of the trends and determinants of coronary heart disease in the U.S. The study began by focusing on young adults, that is, individuals 18 to 30 years of age at the time of the initial Y0 screening, undertaken between March 1985 and June 1986. A random selection of 5,115 black and white men and women identified by each of the four CARDIA Field Centers constituted the cohort. The selection procedures, study populations and methods used are described elsewhere [1, 2] and the consequent baseline results have been reported [3, 4].

Subsequent exams were held at Y2 (June 1987-June 1988), Y5 (June 1990-June 1991), Y7 (June 1992-June 1993), Y10 (June 1995-June 1996) and Y15 (June 2000-June 2001). The contents and methods used in these prior examinations are described in the previous CARDIA Manuals of Operations and Protocols [5-14]. Follow-up examinations at Y2, Y5, Y7, Y10, and Y15 achieved high retention rates, collected a rich set of high quality data and stored specimens bearing on the risk factors and possible causes of cardiovascular disease (CVD), and led to 213 peer-reviewed publications (as of October 2005).

This document describes the Protocol for the Y20 Exam (also referred to as Exam VII) undertaken from June 2005, with initial target end-date of May 30, 2006 on the CARDIA cohort, who are now 38-50 years old. We propose to re-examine at least 73% of those surviving (3,650 participants study-wide). This document provides the rationale, objectives and methods for the Exam VII cycle for the members of the CARDIA cohort. Coronary calcification and carotid artery intima-medial thickness (IMT) of the carotid artery and their relationships to the cardiovascular risk factors are major focuses for this exam cycle.

The core Y20 data, as well as findings in separately funded ancillary studies, will allow examination of the antecedents and prevalence of subclinical atherosclerosis in diverse populations, analyses of the role of predisposing genetic traits in the presence of behavioral and physiologic risk factors in order to detect genotype-by-environment interactions, and determination of how these differ in men and women, and in blacks and whites. These findings will increase understanding of the 20-year antecedents of middle-aged risk factors and subclinical disease. This knowledge will be important in designing preventive medicine policies and interventional studies that address the growing epidemic of obesity and reduce the public
health burden of CVD, and that are tailored to specific population subgroups and settings where they will be most effective.

B. STUDY OBJECTIVES

The CARDIA Y20 Exam offers a unique opportunity to examine the evolution of lifestyle and risk factor profiles from young adulthood into middle age, and their effect on both incidence and progression of subclinical disease. Of particular importance in this context is the emerging epidemic of obesity and its role in pathogenesis. More generally, we will assess how phenotypic expression of genetic traits, environmental exposures, and ethnicity operate in conjunction with inflammatory markers and other intermediaries to influence atherosclerosis and its early manifestations. Specifically, the primary objectives are:

1. **To identify predictors of earlier development and more rapid progression of subclinical atherosclerosis:** Test the relationship between 20-year patterns in risk factors (established, novel, lifestyle, psychosocial, and socioeconomic) and the development and progression of subclinical atherosclerosis (coronary artery calcification [CAC] and carotid intima-media thickness [IMT]); Examine the antecedents of these risk factors, with special attention to the growing epidemic of obesity; Examine whether risk factors differ for CAC vs. IMT, explore the impact of short-term vs. long-term exposure to risk factors, and assess the contribution of conditions such as obesity, diabetes, renal impairment, low socio-economic status, and poor health care access/utilization to subclinical atherosclerosis.

2. **To assess racial differences in severity and progression of early subclinical disease to elucidate possible differences in CVD pathogenesis:** Examine whether the higher prevalence of CAC in whites than blacks observed at Y15 continues; whether IMT is greater in blacks than whites at these relatively young ages as it is in older cohorts; and whether the rate of CAC progression differs by race; (If racial differences in these subclinical measures exist) Examine factors that are associated with CAC and IMT within each race-gender group, and factors that may explain these differences.

3. **To test whether inflammation precedes subclinical disease:** Explore whether inflammation is a predictor or a consequence of atherosclerosis, examining the time course of the association between inflammatory markers such as C-reactive protein (CRP) and the occurrence of CAC and IMT; Identify predictors of inflammation (including infection), the
impact of obesity and of visceral adiposity, and the time course of inflammation-associated
dysregulation of normal physiology.

4. **To assess the roles of genetic variation and gene by environment interactions in early
development and progression of subclinical disease:** Explore the role of genetic variation and
gene by environment interactions in the etiology of risk factors; Test if allelic variation in
genes such as those regulating bone mineralization, obesity, dyslipidemia, and blood pressure
are associated with CAC and IMT.

In addition, objectives from previous examinations that will continue to be addressed are:

1. To measure physiologic correlates of blood pressure and hypertension:
   a. Compare relationships between physiologic measures and blood pressure in blacks and
      whites;
   b. Determine relationships between physiologic correlates of blood pressure and other
      cohort characteristics; and
   c. Identify and quantify subclinical conditions associated with elevated blood pressure,
      such as early renal dysfunction and structural vascular changes.
2. To assess the levels and determinants of obesity, body fat distribution and weight change.
3. To continue the follow up to:
   a. Identify correlates of smoking cessation and other changes in smoking behavior.
   b. Identify correlates of changes in blood lipid and lipoprotein levels.
   c. Study the relationship of levels and changes in blood lipids and lipoproteins to the early
      stages of both cardiovascular and non-cardiovascular diseases.
4. To continue monitoring cardiovascular and other disease outcomes and relate these
   outcomes to both baseline characteristics and to changes in risk factor levels.

Secondary Objectives for this exam are:

1. To develop and apply effective methods of follow-up during these young adult years to re-
   examine at Y20 Exam at least 73% of the surviving participants.
2. To assess risk factors for CVD and disease endpoints in these participants with high
   accuracy and precision.
3. To continue developing and using the most appropriate analytic methods, especially
   longitudinal data analysis methods, for example utilizing both cross-sectional and cohort
   data to assess age-related trends in risk factors during young adulthood.
## C. Exam Components

Exam Components for all CARDIA core exams, including Y20, are listed in the following:*  

### Table 1. Schedule of CARDIA components by examination

<table>
<thead>
<tr>
<th>Components</th>
<th>Year/Exam</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>85 87 90 92 95 00 05 05</td>
</tr>
<tr>
<td></td>
<td>0 2 5 7 10 15 20</td>
</tr>
<tr>
<td><strong>Standard Risk Factors</strong></td>
<td></td>
</tr>
<tr>
<td>Blood Pressure</td>
<td>X X X X X X X X</td>
</tr>
<tr>
<td>Lipids</td>
<td>X X X X X X X</td>
</tr>
<tr>
<td>Anthropometry</td>
<td>X X X X X X X</td>
</tr>
<tr>
<td>Psychosocial Measures</td>
<td>X X X X X X X</td>
</tr>
<tr>
<td><strong>Chemistries</strong></td>
<td></td>
</tr>
<tr>
<td>Fasting Insulin/Glucose</td>
<td>X - - X X X</td>
</tr>
<tr>
<td>Oral Glucose Tolerance Test</td>
<td>- - - - X - X</td>
</tr>
<tr>
<td>Stored Plasma/Serum</td>
<td>X X X X X X X</td>
</tr>
<tr>
<td>Creatinine</td>
<td>X - - - X X X</td>
</tr>
<tr>
<td>Uric Acid</td>
<td>X - - - X X -</td>
</tr>
<tr>
<td>C-reactive Protein</td>
<td>- - - X - X X</td>
</tr>
<tr>
<td>Microalbuminuria</td>
<td>- - - - X X X</td>
</tr>
<tr>
<td>Interleukin-6</td>
<td>- - - - - X X</td>
</tr>
<tr>
<td>Fibrinogen</td>
<td>- - X - - X X</td>
</tr>
<tr>
<td>Infectious Disease Markers</td>
<td>- - - - - X -</td>
</tr>
<tr>
<td>Family History</td>
<td>X - X - X - -</td>
</tr>
<tr>
<td><strong>Physical Activity/Fitness</strong></td>
<td></td>
</tr>
<tr>
<td>Physical Activity History</td>
<td>X X X X X X X X</td>
</tr>
<tr>
<td>Graded Exercise Test</td>
<td>X - - - X - -</td>
</tr>
<tr>
<td><strong>Nutrient Intake</strong></td>
<td></td>
</tr>
<tr>
<td>CARDIA Diet History</td>
<td>X - - X - - X</td>
</tr>
<tr>
<td>Food Frequency</td>
<td>- X - - - - -</td>
</tr>
<tr>
<td>Obesity Questionnaires</td>
<td>- - X X X X X</td>
</tr>
<tr>
<td><strong>Pulmonary Function</strong></td>
<td></td>
</tr>
<tr>
<td>Testing</td>
<td>X X X - X - X</td>
</tr>
<tr>
<td>Questionnaire</td>
<td>X X - - X X X</td>
</tr>
<tr>
<td><strong>Electrocardiogram</strong></td>
<td></td>
</tr>
<tr>
<td>Exercise</td>
<td>X - - X - - -</td>
</tr>
<tr>
<td>Resting</td>
<td>- - - - - - -</td>
</tr>
<tr>
<td>Echocardiography</td>
<td>- - X - X - -</td>
</tr>
<tr>
<td>Coronary Artery Calcification</td>
<td>- - - - - X X</td>
</tr>
<tr>
<td>Carotid IMT</td>
<td>- - - - - - - X</td>
</tr>
<tr>
<td>Genetic Material</td>
<td>- - X - X X X</td>
</tr>
</tbody>
</table>

*Ancillary studies and substudies are not included in this Table.

**Also included: smoking, medical history, interim hospitalizations, events and substance use."
D. STUDY DESIGN

1. Population

Of the 5,115 participants, between the ages of 18-30, enrolled by CARDIA Field Centers in 1985-86, 45.5% were men, 51.5% were Black and 48.5% White. It is anticipated that approximately 3,639 (~73% of those surviving) will be examined during the Y20 Exam at the four Field Centers: 834 in Birmingham, 816 in Chicago, 989 in Minneapolis and 1000 in Oakland. Participants will be invited to participate in the Y20 Exam during its entire period which extends approximately from June, 2005 through May, 2006.

2. Informed Consent

All participants will go through an informed consent process prior to initiating the Y20 Exam. Each Field Center has a consent form approved by their institutional review board, which while sharing common consent sections, vary slightly. Each form explains the procedures of the Y20 Exam with accompanying risks and benefits. After the participant has read and had any questions answered about the consent form, they are asked to individually check or initial each consent activity as follows: (wording and format varying slightly by Field Center)

a. Participate in the CARDIA Y20 Exam to measure weight, cholesterol, blood pressure and other physical factors related to CVD including an ultrasound image of the arteries of the neck.

b. Donate a blood and urine sample to be frozen, stored, and used for future research.

c. Allow CARDIA investigators to extract DNA from blood samples to be analyzed for genetic information related to heart disease, obesity, blood pressure, diabetes and other conditions affecting people in midlife, understanding that the information will be kept confidential at all times.

d. Allow genetic/DNA samples and data to be released, for research purposes, to other qualified non-CARDIA investigators who have met CARDIA’s standards for confidentiality.

e. Allow researchers from private or non-profit organizations to have access, in a way that cannot identify you, to specimens, DNA or other CARDIA information in order to develop diagnostic laboratory tests, medications, or other therapies that could benefit many people.
f. Wish to know your results if a gene is found that is linked to a medically treatable genetic disease.

g. Agree to have a CT exam to look for calcium in the arteries of your heart and other findings that may be related to heart disease.

Consent is sought for an ancillary study, YALTA, at the same time as the core consent. Each Field Center has a different consent form configuration as follows:

- BHAM – Core consent form with CT addendum; second consent form for YALTA
- CHIC – One consent form which includes Core, CT and YALTA
- MINN – Core consent form which includes CT; second consent form for YALTA
- OAKL - Core consent form which includes CT; second consent form for YALTA

3. Data collected

The Y20 Exam measures numerous risk factors, including plasma lipids; blood pressure; anthropometry; smoking behavior; blood levels of lipids, insulin and glucose; physical activity; psychosocial factors; pulmonary function (PF); creatinine; and microalbuminuria in order assess cohort changes and trends. In order to enhance comparability, these measurements will use the same data collection and analysis techniques that were previously used, where possible, as described for key measures in Sections E7-8. Data on socio-economic status (SES), alcohol intake, medication use, recent hospitalizations, and medical diagnoses is collected using the same methods and questionnaires as described in previous years’ exam manuals. Y20 will enrich data on environmental exposures by measuring diet with the CARDIA diet history, which was previously assessed at Y0 and Y7. These measures will allow the study of interactions between ethnicity, gender, SES, and modifiable environmental characteristics such as diet and smoking, in relation to subclinical disease. Primary outcomes collected at Y20 to detect subclinical disease are coronary calcification measured by Computed Tomography (CT) and IMT of the carotid arteries measured by ultrasound.

In addition to the measurements mentioned above, numerous aliquots of serum and plasma, buffy coat for DNA, and urine are set aside. These will be inventoried with the previous exams’ specimens for future case-control studies of associations and longitudinal trends in new analytes and potential risk factors as they are discovered.
4. Comparability Studies

A series of comparability studies were done to ensure that Y20 measures are consistent with measures from previous years. An explanation of these studies follows.

a. **Blood Pressure:** In August of 2004, the Design and Analysis (D&A) Subcommittee designed a blood pressure comparability study that was conducted at the Minnesota Field Center under the leadership of Dr. Pamela Schreiner. After a careful review of the literature, the Subcommittee recommended the use of the Omron® HEM 907XL ([http://www.omronhealthcare.com/enTouchCMS/FileUplFolder/HEM-907XL.pdf](http://www.omronhealthcare.com/enTouchCMS/FileUplFolder/HEM-907XL.pdf)) for that comparability study. The study, designed to compare the Omron (OM) and the Y15 Random Zero (RZ) devices, consisted of 100 volunteer participants, aged 18 to 77. For each participant, two sets of blood pressure levels were measured. Not only was there simultaneous measurement (one by OM and one by RZ) but by allowing two replications, each observer had the opportunity to use both machines on the same participant. For both observers, the RZ readings of systolic and diastolic blood pressure were, on average, lower than the corresponding OM readings; however the differences may have been influenced by the observers. The correlation between the RZ and OM devices was 0.954 for Observer 1 and 0.92 for Observer 2. As a result of this study, it was determined that the assumption of linearity between the RZ and OM blood pressure (SBP and DBP) is reasonable. It was also determined that observer difference may impact the precision and accuracy of the equations and that the sample is too small to provide a definitive predictive equation. As a result, the D & A Subcommittee recommended that the comparability study be enlarged to include a calibration study whose purpose would be to establish a calibration equation for transforming OM levels to RZ levels in order to monitor blood pressure trend over time. This expanded study would include 200 participants (about 50 from each race and gender group) from each of the four Field Centers and is conducted during the beginning of the Y20 Exam period. The blood pressure distributions would be monitored to ensure that people with blood pressure greater than 135/85 mmHg will be included. Measurement protocol, developed by Dr. Ronald Prineas, is detailed in Section 3 E of the CARDIA Y20 Manual of Operations (MOO).

b. **Computed Tomography (CT)** - The CT comparability study design includes retrospective scoring of Y15 Exams with the new TerraRecon software. A 5% sample
(n=150) of the total number of participant scans (≈3,000) was randomly selected to be re-read for this study. The sample was configured so that the prevalence of exams positive for calcified plaque was ~2/3 (100 participants with non-zero scores and 50 participants with zero scores on the Y15 Exam). The results showed that there was a high level of agreement between the two methods. Fifty-five of 58 (94.8%) of scans that were originally scored as negative for CAC by the Y15 method were also scored as negative for CAC by the TerraRecon® software, and 86 of 88 (97.7%) of the scans scored as positive for CAC by the Y15 method were also scored as positive for CAC by the TerraRecon software. For the 86 cases in which both the Y15 and TerraRecon scores were nonzero, the mean score was 78.2 for the Y15 reads and 82.3 for the TerraRecon rereads (mean difference 4.2).

c. **Laboratory Assays** - To ensure the reproducibility of results, a sample of 100 of each of the measures used previously (Y7-fibrinogen, Y15-urinary albuminuria, urinary creatinine, fasting insulin, glucose, serum creatinine, C-reactive protein, total cholesterol, HDL cholesterol, and triglycerides) will be rerun. Agreement between original Y15 and rerun Y15 measures will be determined by examining mean differences, standard deviations, and correlations. Each of the laboratories and assay methods used for Y20 are the same as those used in Y15 with a few exceptions (albuminuria was originally run by the Lipid laboratory in Y15 but was rerun by the new Albuminuria Laboratory; C-reactive protein was originally run using an ultrasensitive ELISA assay in Y15 but was later rerun using the BNII method which has been adopted for Y20). The sampling scheme, which was based on the distribution of values, has been approved by the Steering Committee and specimen retrieval lists are produced accordingly.

**CARDIA Laboratory Assay Sampling Scheme for Lab Comparability Study**

<table>
<thead>
<tr>
<th>Laboratory</th>
<th>Assay</th>
<th>Distributional Cut Points</th>
<th>Value Range</th>
<th>Number to be Sampled</th>
</tr>
</thead>
<tbody>
<tr>
<td>Albuminuria</td>
<td>Urinary Albumin (G/DL)</td>
<td>Q1 0 – 0.34</td>
<td>25</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Q2 0.34 – 0.62</td>
<td>25</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Q3 0.62 – 1.09</td>
<td>25</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Q4 1.09 – 25.1</td>
<td>25</td>
<td></td>
</tr>
<tr>
<td>Urinary Creatinine (MG/DL)</td>
<td>Q1 0 – 92</td>
<td>25</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Q2 92 – 152.4</td>
<td>25</td>
<td></td>
</tr>
<tr>
<td>Chemistry</td>
<td></td>
<td>Q3</td>
<td>152.4 – 213.5</td>
<td>25</td>
</tr>
<tr>
<td>---------------------------------------</td>
<td>----------------</td>
<td>-----</td>
<td>---------------</td>
<td>----</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Q4a</td>
<td>213.5 – 417.4</td>
<td>25</td>
</tr>
<tr>
<td>Fasting Insulin (MicroU / ML)</td>
<td>Q1</td>
<td>0 – 8</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Q2</td>
<td>8 – 12</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Q3</td>
<td>12 – 17</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Q4a</td>
<td>17 – 53</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fasting Glucose (MG / DL)</td>
<td>Q1</td>
<td>0 – 78</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q2</td>
<td>78 – 83</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q3</td>
<td>83 – 90</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q4a</td>
<td>90 - 189</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td>Serum Creatinine (MG / DL)b</td>
<td>Group 1</td>
<td>0.3 – 0.7</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Group 2</td>
<td>0.8</td>
<td></td>
<td>15</td>
</tr>
<tr>
<td></td>
<td>Group 3</td>
<td>0.9</td>
<td></td>
<td>15</td>
</tr>
<tr>
<td></td>
<td>Group 4</td>
<td>1.0</td>
<td></td>
<td>15</td>
</tr>
<tr>
<td></td>
<td>Group 5</td>
<td>1.1</td>
<td></td>
<td>15</td>
</tr>
<tr>
<td></td>
<td>Group 6</td>
<td>1.2</td>
<td></td>
<td>15</td>
</tr>
<tr>
<td>Inflammatory Markers</td>
<td>C-reactive Protein (MicroG/ML)—BNII method</td>
<td>Q1c</td>
<td>0.17 – 0.55</td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q2</td>
<td>0.55 – 1.39</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q3</td>
<td>1.39 – 3.81</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q4a</td>
<td>3.81 – 22.6</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td>Fibrinogen (MG/DL)d</td>
<td>Q1</td>
<td>0 – 221</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q2</td>
<td>221 – 255</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q3</td>
<td>255 – 301</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q4a</td>
<td>301 - 448</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td>Lipids</td>
<td>Total Cholesterol (MG/DL)</td>
<td>Q1</td>
<td>0 – 160</td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q2</td>
<td>160 – 182</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q3</td>
<td>182 – 206</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q4a</td>
<td>206 – 284</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td>HDL Cholesterol (MG/DL)</td>
<td>Q1</td>
<td>0 – 40</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q2</td>
<td>40 – 49</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q3</td>
<td>49 – 59</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q4a</td>
<td>59 – 92</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td>Triglycerides (MG/DL)</td>
<td>Q1</td>
<td>0 – 59</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q2</td>
<td>59 – 83</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q3</td>
<td>83 – 122</td>
<td></td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Q4a</td>
<td>122 - 420</td>
<td></td>
<td>25</td>
</tr>
</tbody>
</table>

*a Top 1% to be removed from sampling scheme.

*b This sampling framework was used previously during the Y15 Exam because of the concern of elevated creatinine values associated with a recalled reagent. This quartile starts at 0.17 (5th percentile) rather than 0 since values less than 0.15 are undetectable.

c Fibrinogen assays to be conducted on Y7 samples.

d. **Pulmonary Function (PF)**

The comparability studies for PF are conducted in two phases: i) machine and ii) human testing.

i. The OMI dry rolling-seal spirometer ([http://www.occupational.com/spiro1.htm](http://www.occupational.com/spiro1.htm)) was selected for the Y20 Exam and is very compatible with the Y10 Collins II Survey Spirometers that have been utilized in the previous measurements of PF. Concordance with earlier data will be obtained by carefully measuring the exact performance of the
Collins II survey spirometers at the testing facility at Latter Day Saints (LDS) Hospital. Specifically, each Field Center sent their Y10 spirometers to LDS for testing. Each of the spirometers, both Y10 and Y20 devices, are compared against a “gold standard,” the pulmonary wave-form (PWF) generator. The generator moves a precise amount of air through a high precision robotic motor allowing very specific measurements. The accuracy of both spirometer devices were assessed and compared to determine any bias between the two.

ii. To further assess comparability, testing is extended to human subjects. This phase, carried out at LDS with twenty-five participant-like volunteers, requires testing on both the Y10 and Y20 instruments, providing a side-by-side comparison of a functioning older machine with a new machine.

E. RATIONALE AND METHODS

The development of atherosclerosis is a complex process involving atherogenesis, the deposition of lipids, smooth muscle and macrophage infiltration and collagen overgrowth, and sclerosis, which is linked in part to calcification of an injured arterial wall. Individual differences in the development and presence of these components characterize early disease, and conventional CVD risk factors may be more strongly linked to one part of the process than another. Further, while several studies suggest that IMT and CAC are correlated [15, 16], there is evidence that considerable variability exists in the association between CAC and atherosclerosis, both within arteries and between arterial beds [17]. Thus, calcification may be part of a common process while the progression from atherosclerosis to calcified plaque may not occur consistently across all arterial sites.

Subclinical atherosclerosis is measured via CAC and IMT to identify predictors of subclinical disease development and progression, and to determine how these measures are related to interactions of genetic variation with lifestyle, behavioral and physiologic risk factors, and the evolution of these risk factors.

1. Racial Differences

Previous studies in older cohorts have found ethnic differences in subclinical disease prevalence, suggesting that there may be ethnic differences in the pathologic significance of
these measures, and in correlations with risk factors [18]. CHS and IRAS found that African Americans have significantly thicker common carotid artery walls than whites, independent of CVD risk factors;[19, 20] conversely, African Americans had lower levels of calcification in a subset of the CHS [21]. Another study found that while CAC prevalence was significantly lower in African Americans than in whites of similar risk levels, African Americans suffered more MI and angina, and similar CHD death and revascularization over follow-up [22]. Other studies have found ethnic differences in plaque characteristics (e.g., calcification, fibrosis and density), plaque volume, arterial distensibility and endothelial function [23-26]. Thus, there is emerging evidence of ethnic differences in the pathology of atherosclerosis which could influence the design of preventive medicine programs.

A continuing strength of CARDIA is our ability to disentangle the effects of ethnicity and SES on CVD rates. African-Americans are at higher risk for early CVD than whites, but are also disproportionately represented in SES strata related to CVD risk factors and outcomes.[27] The depth of psychosocial and biologic data in CARDIA constitutes a great resource for elucidating the independent and combined effects of SES and African-American ethnicity on CVD pathogenesis and subclinical disease.

We have observed striking increases in obesity prevalence across exams, particularly among Black women (51% at Y15 with BMI ≥ 30.0 vs 22% at Y0), and the attendant rise in clinical disorders such as hypertension, hyperlipidemia, and diabetes is alarming. Thus, a particular focus is on hypotheses concerning the health consequences of obesity, including the importance of central fat patterning, duration of obesity, and the interplay of environmental and genetic factors in contributing to the metabolic syndrome and subclinical CVD. For example, we observed that the favorable downward secular trend in both LDL and total cholesterol between Y0 and Y7 [28] had leveled off in Y15 (white women, black men and black women) or reversed.[29] Body mass index (BMI) and waist circumference were the factors that most affected these adverse trends. In the context of a national obesity epidemic, further follow-up of the CARDIA cohort will determine if an upward secular trend in LDL is taking place in the other three race-gender groups; and will define trends in HDL and triglycerides.

The impact of genetic variation on subclinical CVD risk likely differs in the presence of other risk factors such as obesity or hypertension. Thus, the genetic variants and risk factors related to CAC and IMT may drive racial disparities through differential progression of early subclinical disease. We will take several steps to assess racial differences in prevalence and
progression of CAC and IMT, and to explore factors that may be responsible. We will first examine whether Blacks have increased IMT, but lower CAC scores and/or slower progression of CAC. If there are differences, we will systematically examine, in univariate and multivariate analyses, relationships of new and established psychosocial, lifestyle, genetic and biomedical risk factors with IMT, CAC and changes in CAC in Blacks and whites. We will also compare the characteristics of individuals with varying degrees of IMT and CAC to identify factors that are more relevant to carotid IMT or CAC. Based on these findings, we will explore whether racial differences in different subclinical disease measures, and in the progression of CAC, can be explained by differences in lifestyle, psychosocial, genetic and biomedical risk factors.

The NHLBI–NIDDK Pathophysiology of Obesity-Associated Cardiovascular Disease Working Group recommended research with emphasis on the cardiovascular consequences of obesity in the young, and the role of adipose tissue as a proinflammatory secretory organ affecting multiple components of the cardiovascular system (eg., blood pressure, lipid metabolism, clotting and inflammatory pathways, cardiac conduction systems); and on the cardiovascular consequences of obesity among the young. CARDIA is well positioned to address these issues due to the age and composition of the cohort, and the wealth and quality of the longitudinal phenotypic data and genetic material available. We can search for predictors and correlates in the early stages of disease, perhaps leading to new and important interventions.

2. CAC Measurement

CAC corresponds to coronary plaque quantity [30] and the probability of significant angiographic disease [31-34]. The Muscatine Study documented CAC in 31% of men and 10% of women aged 29-37 years and found CAC associated with BMI, systolic blood pressure, LDL-C, and HDL-C [35, 36]. While prevalence of any calcification (Agatston score > 0) was relatively low at CARDIA Y15, CAC varied by race-gender group (11.3%, 4.9%, 17.6% and 5.3% in black men, black women, white men and white women, respectively). Odds of CAC were higher with increasing age and cigarettes smoked in all race-gender groups, and with increased LDL-C, SBP, and BMI in some groups [37].

The CT Reading Center (CTRC) developed the Y20 protocol for CAC measurement. We measured CAC at Y15 with a scanning protocol developed by a committee of cardiologists, radiologists, and a physicist in order to provide almost uniformly acquired scans with different technologies, electronic beam computed tomography (EBT) and multi-detector computed
tomography (MDCT). Participants were all scanned over a hydroxyapatite phantom in order to allow monitoring of image brightness and noise and adjust for scanner differences in brightness levels during reading. Two scans were obtained on each participant 1-2 minutes apart. Using a 35 cm field of view, we obtained sequential axial scans with 10.5 cm of data in each scan (3 mm slice thickness for EBT and 2.5 mm for MDCT). One cardiovascular radiologist scored all the scans using specially developed software, and each scan set with at least one non-zero score and a random sample of scan sets that were negative for CAC was reviewed by an expert investigator from the Reading Center (Dr. Robert Detrano, UCLA-Harbor) without knowledge of the scan scores to verify the presence of coronary calcium. A second expert reader (Dr. Jeffrey Carr, Wake Forest) who was not from the Reading Center was trained in software use and reviewed 100 scans using the same adjudication process. He and the Reading Center scorer agreed in 91% of cases, including 95% of concordant scan sets (74 of 78 scan sets, both with zero scores or both with non-zero scores) and 82% of discordant scan sets (18 of 22 scan, one scan with a non-zero score and one a zero score). Since most nonzero scores were low, the procedures developed by CARDIA represent a method of interpreting scans detecting small amounts of calcium that will enable us to reliably determine the progression of coronary calcium in a dichotomous manner (ie., zero to nonzero scores) from Y15 to Y20. Prior to the start of the Y20 Exam, the CARDIA Imaging Subcommittee and the CTRC also worked together to design and implement a CT comparability study that included 150 participants to ensure comparability of Y15 and Y20 coronary arteray calcium measures (Refer to Section D.4.b of this protocol for details concerning this comparability study).

The Y20’s CT scanning of the chest for the measurement of coronary calcium are scheduled by the Field Center staff; generally within three weekdays of a participant’s Y20 CARDIA examination. Coronary calcium is measured using the following scanners at the Field Centers:

- Birmingham GE Lightspeed QX/I (tentative)
- Chicago Imatron C-150
- Minneapolis Siemens S4+ Volume Zoom
- Oakland Imatron C-150

The following individuals will be excluded from the CT component:

1. Weight greater than 160 kg (352 lbs), but may vary by Field Center site
2. Pregnant, pregnancy status unknown, or any woman who has not undergone a hysterectomy or a tubal ligation but who has had unprotected sex at any time within the 7 days prior to the scheduled CT scan (this is because an urine pregnancy test may stay negative for several days after a woman becomes pregnant).

Each participant undergoes two sequential scans during one session. Data from both scans is transmitted electronically to the CTRC for analysis. Measurements made include calcium volume, calcium score, and calcium volume score, and calcium mass. The score is the sum of the areas of the calcium foci identified, weighted by the density within each focus. The volume takes into account slice thickness. The volume score is calculated similarly to calcium volume except that it is calculated from interpolated images with a slice thickness equal to the pixel width. The mass calibrates the measure to concentrations of hydroxyapatite from the calibration phantom. These measures are calculated for each of the main coronary arteries (left main, left circumflex, left anterior descending and right) and as a total value.

3. Carotid IMT Measurement

The association of CVD risk factors with IMT, the relationships between baseline IMT and incident CHD in middle-aged and elderly adults [37-44], and risk factors such as LDL-C and smoking are well known [35, 45]. These data suggest that IMT can be considered a marker of early atherosclerosis. IMT can measure wall thickening of any etiology, including medial hyperplasia from chronic hypertension. While thickening of the internal carotid artery and the carotid bifurcation are more likely due to fatty and fibrous accumulation, the composition of the common carotid makes it prone to muscular changes. Given the increase in elevated blood pressure observed at Y15, data from the common carotid artery will add to our understanding of the independent effects of blood pressure changes across young adulthood.

The CARDIA Study will use a variant of the MESA/CHS protocol. The scanning protocol is presented in two parts. Part 1 – CCA Video is the capture of a video stream of the right common carotid artery. Part 2 – Carotid IMT is performed first on the right and then on the left side. On each side Part 2 – Carotid IMT scanning begins with a transverse sweep from the base of the common carotid up through the internal carotid. The transverse sweep is followed by a pulse-wave Doppler measurement in the ICA and then grayscale images of the common carotid, the carotid bulb, and the internal carotid arteries. Five standardized B-mode images will be acquired from each of the right and left carotid arteries. One measurement will be made of the
common carotid, two of the carotid bulb, and two of the internal carotid artery. The imaging priorities are primarily arterial wall thickness.

4. Blood Pressure

High blood pressure, or hypertension, is a serious public health problem that afflicts approximately 25% of American adults [46]. Hypertension is a major risk factor for heart attacks, congestive heart failure, stroke, and kidney disease[47]. These risks are also increased in adults whose blood pressure level is not hypertensive but is above optimal [48]. More than one-half of all heart attacks and greater than three-fourths of all strokes occur in patients with hypertension. In addition, most patients who develop kidney failure and require dialysis are also hypertensive. Although long-term management can reduce the risks associated with high blood pressure, many of the risks have not been eliminated completely [49].

Hypertension is multifactorial; genetic, neural, humoral, vascular, cardiac, renal, nutritional, and psychosocial factors all play interdependent roles [47]. The hypertensive population is also heterogenous with respect to the degree of response to manipulation of environmental factors and medications, and the tendency to develop blood pressure-related complications. Major progress has been made in improving understanding of the causes of hypertension and in developing effective therapeutic programs to improve its control [50]. A substantial contribution to this body of information has been made from results of work with CARDIA data. Some examples from the CARDIA Study include: higher blood pressures in blacks that increased over time was due to physical activity and alcohol intake levels [51]; benefits of plant food and adverse effects of meat intakes on blood pressure [52]; psychosocial factors (e.g., job strain) assessed early in CARDIA are associated with increased risk of hypertension [53]; blood pressure reactivity to psychological stress predicts hypertension [54]; and, increased risk of hypertension development in young blacks with depression [55].

During the previous examinations, blood pressure was measured using the Hawksley random zero (RZ) sphygmomanometer. Due to the risks of mercury toxicity, Hawksley no longer manufactures or services this device. As a result, a new instrument is used for the Y20 exam: the OmRON HEM907XL Each participant sits in a quiet room for five minutes prior to having three blood pressure measurements taken from the right arm. The second and third blood pressure readings are averaged for analyses. Pulse is taken by standard count methods using the radial pulse.
5. Obesity and Anthropometry

In recent decades, the prevalence of obesity (BMI $\geq 30.0$) increased markedly in the United States [56] and in 1999, almost 108 million adults in the U. S. were overweight or obese [57]. Multiple investigations have shown that obesity and overweight increase the risk of morbidity from hypertension; dyslipidemia; type 2 diabetes; coronary heart disease; stroke; gallbladder disease; osteoarthritis; sleep apnea and respiratory problems; and endometrial, breast, prostate, and colon cancers, and all-cause [57]. Therefore, any effort to prevent and control it will have an important impact on the health care system.

There are three primary reasons for including body size measurements in CARDIA:

- They provide a standardized basis for examining the association between physical size, stature, or ponderosity, and risk for developing coronary heart disease, hypertension and other diseases of interest (e.g., chronic obstructive lung disease, diabetes).

- Repeated measures of body habitus over time permit investigation of whether changes in body size directly contribute favorably or unfavorably to an individual's risk profile (i.e., tracking of secular and aging trends of known cardiovascular disease risk factors).

- Because the data will be collected using standardized methods employed in other observational studies besides CARDIA, the representative nature of the CARDIA data set can be determined using existing data and ethnic groups. Given the patterns of weight gain in the American population and the increasing prevalence of overweight and obesity, including central fat patterning, it is important to continue obtaining body size measurements on this cohort.

Previous body size measurements have been reported, either independently or collectively, and they have been correlated with a specific risk factor or clinical manifestations of disease. These measures also represent important indicators of growth and nutritional status. Although numerous additional anthropometric and/or body size measurements have been identified, this study focuses on indices particularly relevant to the risk of developing coronary heart disease or systemic arterial hypertension. Measurements were limited to those that are the most relevant and that can be performed using standardized methodology within and between survey centers because of time limitations and other practical constraints of the study.
Specifically, the anthropometric and body measurements included in studies such as the National Health and Nutrition Examination Survey (NHANES), Framingham, and the Lipids Research Clinics Prevalence Survey served as a model for the proposed measurements [58-60]. In addition, the age group involved in CARDIA suggested the need for monitoring growth and development inherent in the aging process which may have been unrelated to environmental influences such as dietary or exercise patterns.

a. Height and Weight - Among all the possible measurements, height and weight are universally accepted as primary indices of body size. Weight for height standards are available for both men and women from studies such as the Build and Blood Pressure Study and the updated Metropolitan Life Insurance tables. Based on standards such as these, relative weight can be determined. In addition, several ratios of weight/height, such as the body mass index (BMI) (Quetelet's Index\(^1\)) or the Ponderal Index\(^2\), have been positively correlated with risk factors for CVD, including hypertension, hyperlipidemia and diabetes.

BMI is a more reproducible measure than some other measures of body fat, such as skinfold thicknesses [61], and is more strongly associated with blood pressure and serum cholesterol in some populations[61]. Epidemiologic data have shown that body mass index above 25 kg/m\(^2\) is associated with increased mortality, which tends to be modest until a body mass index of 30 kg/m\(^2\) is reached [62-64]. As consequence, the National Institutes of Health recommended to classify patients as overweight when the body mass index is between 25.0 and 29.9 kg/m\(^2\) and obesity with a body mass index ≥ 30 kg/m\(^2\) [57], classification that would be used to analyze the data of the CARDIA study.

Not only the existence of obesity, but also the age of onset of obesity are reportedly related to susceptibility to hypertension [65]. Weight gained early in adult life may be a sensitive indicator of increased risk for CVD. Hypertrophic obesity experienced during adulthood may impose different risks than hyperplastic obesity developed during youth, and these risks may vary among different age, sex and ethnic groups [66]. On the other hand, changes in weight or BMI over time, particularly decreases in BMI [67] and

\(^1\) Quetelet index is calculated as: weight divided by height squared (W/H\(^2\) [kg/m\(^2\)]). Conversion [w(pounds)/h (inches\(^2\)] x 703 (1 lb=0.45 kg) (1 in.=2.54 cm=0.0254 m).

\(^2\) Ponderal index is computed as weight (in kg)/ht\(^3\) (in meters).
weight cycling [68], have been associated with increased risk of all-cause, cardiovascular and CHD mortality, independently of baseline BMI and other risk factors. For these reasons it is important that CARDIA include measures of height and weight during each examination.

Nevertheless, it is well known that there are numerous limitations to indices of relative weight [69]. For example, height and weight tables do not reflect the variations across different ethnic, socioeconomic and/or occupational groups nor do they indicate differences in body composition or fat distribution. Thus, in order to determine body composition and fat distribution more accurately, waist measurement is also performed in CARDIA.

b. Waist Girth - Central or upper body obesity is associated with insulin resistance, hypertriglyceridemia and reduced HDL-cholesterol and with the future development of diabetes, myocardial infarction, angina pectoris, stroke, and all-cause mortality [70-72]. Visceral (intra-abdominal) adipose tissue shows the strongest link to risk factors for CVD and type 2 diabetes mellitus [70-74]. Waist circumference is a measure of central adiposity that correlates well with visceral fat [75-78]. Among nearly 400 CARDIA participants who participated in an ancillary study in which visceral fat was measured, the correlation of waist circumference with visceral fat is 0.44 for black men, 0.64 for white men, 0.62 for black women, and 0.66 for white women.

c. Rational for Changes to Anthropometry Form

This section is included in the Y20 protocol in order to document changes to the anthropometry form for future CARDIA studies. As part of the process of reviewing CARDIA forms for Y20, the anthropometry form was checked for wording, and compared with instructions provided in the manual of operations, and training and certification document.

In Y15, technicians were instructed to record height to the nearest 0.50 cm and record any modification to the protocol if a participant was taller than 200 cm (upper limit of ruler) or if there were problems with hairstyle or other issue that might impact the precise estimate of height. In addition, technicians were instructed to “NOT measure pregnant women.”
The analysis of the questionnaire’s responses showed inconsistency in the recording of responses to the question: Was there a modification in protocol? The Coordinating Center found examples that were inconsistent:

- a participant with a height of 203.5 cm and the technician recorded a modification in the protocol. Another participant had a height of 201.5 and it was considered as NO modification in protocol.

In order to avoid these inconsistencies, there is a change to the second part of the question to include a clear reason for protocol modification. To determine these reasons, the Coordinating Center analyzed the responses to question 4 from Y15. These responses were grouped into categories and then included in the new version as options.

For weight and waist girth, a similar process was followed to modify these sections of the form.

In addition, the Y15 MOO indicated that two measurements of waist girth were to be taken, but the anthropometry form has room only to record one measurement of waist girth. It is not clear if technicians averaged the two measurements or if they only measured once. To avoid this problem, we included an additional response space to record the second waist girth measurement. All changes were approved by the Steering Committee.

Anthropometry comprises a simple, but essential, exam component since the dominant experience of the CARDIA cohort has been weight gain, regardless of ethnicity, sex, or SES [79]. Height and weight are measured to compute BMI. Waist circumference is also measured as it correlates well with visceral fat [80]. Waist circumference is measured twice with a Gulick II Plus measurement tape, using the average for analyses. Measures will use the CARDIA methods implemented in previous exams.

6. Diet

For Y20, we again used the CARDIA Diet History (DH) [81], which assesses usual eating patterns—food groups as well as nutrients—over a relatively long period (30 days) rather than actual foods eaten in a short period (eg, the 24 hour recall). CARDIA analyses have shown that fiber and whole grain intake are associated with reduced obesity, lower insulin and blood pressure levels, and better lung function [82, 83]. Increased consumption of dairy products, regardless of their fat content, was related to reduced obesity and insulin resistance among those
overweight at baseline [84]. Albuminuria was positively associated with animal protein or red meat consumption, and negatively with vegetable protein or whole grain intake [85]. Frequent fast food intake was associated with weight gain and worsened insulin status [86]. Intake of whole grain and fruit was associated with reduced risk of future hypertension, while intake of meat was associated with increased risk [87]. Diet was predictive of gamma-glutamyl transferase [88], an enzyme shown to influence risk of diabetes [89] and other clinical conditions. Dietary patterns by race, gender and SES have undoubtedly changed since Y7 when diet was last assessed, and a major goal of the Y20 assessment will be to explore the relevance of these changes to risk factor trends and health outcomes.

At Y0, the DH was highly correlated with both macronutrient and micronutrient assessment in seven 24-hour recalls randomly collected over one month [90], with coefficients somewhat stronger in whites than blacks. The DH, administered in 60-90 minutes, includes questions about typical eating behaviors (e.g., water, vitamin and mineral consumption, number of meals per day, snacking, eating out, and adherence to special diets), a quick review of each dietary item followed by a series of questions about typical intake over the preceding 30 days, using food models and form shapes for quantification.

For Y20, a Diet Reading Center (DRC) was established at the University of Minnesota, headed by Lyn Steffen, PhD. The DRC utilizes an updated nutrient database from the University of Minnesota Nutrition Coordinating Center (NCC), which is based on the USDA Nutrient Database (Surveynet) and other food and nutrient data obtained from food manufacturers. DRC trained and certified Field Center interviewers administer the Diet History according to standardized procedures described in the Y20 Diet MOO. Quality control procedures, conducted by the DRC jointly with the Coordinating Center, will include review of dietary data for outlying nutrient values and unrealistic food consumption, tape-recorded interviews, and site visits.

7. Laboratory Measures

   a. Glycemia: We will assess fasting glucose and insulin and perform a 2-hour oral glucose tolerance test (OGTT). Each Field Center will recruit all willing Y20 participants into the OGTT study, excluding the following individuals:

      i. Those who arrive at the clinic after 10 am (although may participate on a future date),

      ii. Those who did not fast for at least eight hours (although may participate on a
future date),

iii. Those who are diabetic and take insulin and/or oral diabetes medications,

iv. Those using steroids (not including oral inhalers), or

v. Those who are, or may be, pregnant.

Based on Y10 experience, 65% of participants are projected to consent.

Diabetes has long been recognized as a major risk factor for coronary heart disease, and prospective studies have demonstrated that type 2 diabetes is strongly associated with the incidence of myocardial infarction and stroke as well as mortality from CHD and CVD[91-98]. Diabetes is associated with lower HDL-c and higher triglyceride levels than normoglycemia and altered hemostatic factors [99-105]. Higher mean blood pressure and greater prevalence of hypertension have been observed in patients with diabetes, with obesity accounting for only a portion of these differences [106-109]. Further, elevated insulin levels have been associated with high triglycerides, blood pressure, and uric acid and low HDL-c in nondiabetic populations [108, 110-112]. Fasting insulin levels or insulin response to a glucose challenge is an independent risk factor for incident CVD in persons with or without diabetes. Type 2 diabetes may be preceded by long periods of hyperinsulinemia [113-116]. CARDIA has measures of fasting glucose and insulin at most of the previous exams, but as the cohort enters middle age with concomitant weight gain and lifestyle changes, the assumption that fasting glucose and insulin rise linearly may not be correct. CARDIA can examine the effects of aging as well as having unique data to assess the impact of glucose and insulin in young adulthood on incident subclinical disease (coronary artery calcification and IMT far wall thickness) as well as progression of subclinical disease by gender and by ethnicity.

Further, because individuals in Western societies are rarely fasting for prolonged periods of time, we will re-administer the OGTT. At Y10, the prevalence of diabetes was relatively low, but we anticipate that both the prevalence and incidence of diabetes and impaired glucose tolerance will be considerably higher at Y20, and that these endpoints will be strongly related to blood pressure, body weight, dyslipidemia, elevated hemostatic factors, and subclinical CVD including albuminuria. The addition
of OGTT will help to detect those individuals who are pre-diabetic and who are at increased risk of both future diabetes and CVD.

At Y20, glycemia chemistries are measured as follows: insulin by radioimmunoassay and glucose by hexokinase ultraviolet method.

b. **Lipids:** As in all previous exams, we will measure lipids, including total cholesterol, LDL, HDL, and triglycerides [117-120]. Elevated blood lipids as well as low HDL-cholesterol are well established risk factors for CVD morbidity and mortality[121]. Cholesterol levels tend to increase with age in Western populations [122-125]; however, national surveys suggest that total cholesterol has been declining over the past decade in some subgroups of the population [126]. Given the increase in HMG co-A reductase inhibitors (statins) since the Y15 Exam, continued tracking of lipids as part of the Y20 Exam is a critical component of CVD risk assessment. In addition, the women attending the Y20 Exam will be 38-50 years of age, and a moderate proportion of them will be entering the perimenopausal transition when lipids start to rise as a consequence of endogenous estrogen deficiency. In addition to the traditional roles of lipids in CVD risk, with the publication of the 3rd Adult Treatment Panel recommendations from the National Cholesterol Education Program [127], the importance of low HDL as a component of the metabolic syndrome has become more evident. Linked with the increase in the average BMI of both CARDIA participants and the U. S. population over the past 20 years, lipids also are key predictors of impaired glucose tolerance and type 2 diabetes.

At Y20, lipid assays are using the same as used in previous CARDIA exams, as follows: total cholesterol by trinder-type method and determined enzymatically on the Abbot Spectrum (using Hitachi 917 – R1 cholesterol reagent); HDL cholesterol by trinder-type method and determined enzymatically after dextran sulfate –magnesium precipitation on the Abbot Spectrum; and, triglycerides by ultraviolet method and determined enzymatically on the Abbot Spectrum (using Hitachi 917 – R1Buffer/4-Chloropheno/Enzymes). LDL cholesterol will be calculated using the Friedewald equation.

c. **Serum Creatinine:** We will measure creatinine at Y20 by a modified-rate Jaffé method, essentially identical to the method used in previous CARDIA examinations,
thereby insuring comparability. Creatinine reflects kidney function and is both a predictor and a consequence of hypertension [128, 129]. Previous reports have suggested [130] that impaired renal function is both a predictor of CVD and an intermediate outcome of the risk factors that CARDIA has been tracking. Longitudinal trends in serum creatinine will also be examined for their role in CAC prevalence and progression as well as carotid IMT prevalence.

At Y20, serum creatinine is assayed by kinetic in vitro tests using rate-blanking and compensation for the quantitative determination of creatinine in human serum and plasma; kinetic, substrate triggered, rate-blanked method.

d. Inflammatory Markers (CRP, IL-6, Fibrinogen): A unifying hypothesis concerning inflammation, infection, oxidation and hemostasis in relation to endothelial dysfunction and early atherosclerosis is that any condition which induces an acute phase response will alter metabolism in ways that can be proatherogenic and that atherosclerosis is an inflammatory process at all stages of development [131-135]. Plaque destabilization and thrombosis become especially important in progression, while inflammation may be both a cause and a consequence of the atherosclerotic process in both initiation and progression.

Markers of inflammation have been studied in older populations, but there are limited data in young adults [136]. In addition to CVD, inflammatory markers have been related to risk of type 2 diabetes mellitus [137], and may contribute to the relationship of the insulin resistance syndrome to heightened CVD risk. Of particular interest due to the obesity epidemic is the emerging role of adipose tissue, particularly visceral adipose tissue, as a key regulator of inflammation, coagulation, and fibrinolysis [138].

CARDIA measured fibrinogen at Y5 and an ancillary study remeasured it at Y7; CARDIA has now re-measured C-reactive protein (CRP) for Y7 and Y15, by a new, greatly improved assay (see below). Cross-sectional relationships of CRP with CAC in Y15 of CARDIA are not as strong as they are with other CHD markers in other studies, perhaps due to the age of the cohort and/or the markers of atherosclerosis used. Inflammation may increase with a number of factors (e.g., smoking, obesity, oxidative stress), and CARDIA is an ideal setting to examine the natural history of inflammation
and subclinical atherosclerosis and the role of CRP as cause or consequence of the atherosclerotic process.

For Y20, CRP [139-143], interleukin-6 (IL-6), and fibrinogen are being measured. CRP has been associated with subclinical disease measures, including CAC and IMT, but not all studies have consistently shown this relationship [21, 144-146]. Because some studies have shown greater correlations of CRP with risk of clinical events than with measures of subclinical disease, some speculate that CRP may reflect the presence of vulnerable plaque, rather than atherosclerotic burden [147]. Indeed, some data suggest that in nondiabetics, CRP and CAC may provide complementary risk information, perhaps indicating presence, amount, and stability of coronary atherosclerosis [148]. IL-6 is a key cytokine that mediates the acute-phase inflammatory response, is a primary determinant of CRP production by the liver, and has been associated with risk of MI [140, 149]. While some studies comparing the two have found CRP to be a superior predictor [140], we may derive valuable and complementary information by measuring both, as a strategy to decrease measurement and biologic variation, and to assess different aspects of inflammation [136, 150].

Recently, inflammation and coagulation/thrombolysis have been recognized as related processes, such that inflammation may promote local thrombosis which then amplifies inflammation[151]. In addition to being a clotting factor, fibrinogen is an acute phase reactant and is correlated with inflammatory markers and several CVD risk factors [152, 153]. Moreover, fibrinogen levels are related to subclinical CVD, have predicted future CVD events in several studies in both sexes, and in a range of ages, and predict all-cause death [154-156]. Fibrinogen levels are adversely related to level and change in lung function in CARDIA [157]. Fibrinogen levels are determined by identified genetic polymorphisms, as well as environmental [158, 159], and psychosocial factors [160]. There are few data comparing directly the predictive information derived from multiple inflammatory markers examined over considerable periods of time or involving younger study populations.

The multiple measurements of CRP over time will help to clarify whether inflammation precedes the development of coronary calcium. For example, we will examine the relationship between Y7 CRP and presence of CAC at Y20, and the Y20 cross-sectional relationship between CRP and presence of CAC. In particular, we will
assess whether Y7 CRP is significantly associated with Y20 CAC independent of Y20 CRP. In these analyses, we will be sensitive to potential colinearity problems that may be caused by having both Y7 and Y20 CRP levels in the model. In addition, we will also study the relationship between Y15 CRP and incident cases of Y20 CAC, and compare the 5-year (Y15 to Y20) changes in CRP between those with and those without CAC at Y15. The findings from these analyses could help to clarify the temporal sequence of CRP and CAC. Similar analyses will be performed for the relationship between CRP and prevalent carotid IMT at Y20.

The relationship between Y5 fibrinogen level and the presence of Y20 CAC (or Y20 IMT as a continuous variable), and the Y20 cross-sectional relationship between fibrinogen and CAC (or IMT) will be examined. We will assess whether Y5 fibrinogen is significantly associated with presence of Y20 CAC independent of Y20 fibrinogen or vice versa. Again, these analyses will help to clarify the temporal relationship between presence of CAC and fibrinogen. In addition, we will include both fibrinogen and CRP in the analyses to examine the relative importance of these factors in relationship to early subclinical disease as measured by CAC and carotid IMT. The study originally measured CRP with the ultrasensitive ELISA assay [161, 162] based on purified protein and polyclonal anti-CRP antibodies (Calbiochem) at the University of Vermont. Subsequently, Dr. Russ Tracy’s lab has switched to a nephelometry-based high throughput assay that offers even greater sensitivity and much greater reproducibility, improving upon previous procedures for CRP from this laboratory. This method is used for Y20, which is the method used for the re-assays for the Y15 and the initial assay for the Y7 samples. This laboratory is measuring IL-6 with an ELISA assay from R&D Systems, methodology well established in the laboratory. Fibrinogen is assayed at Y20 with an immunologic method, selected for consistency with the previously utilized Claus method. Furthermore, additional measurements are completed with stored samples from earlier examinations to assure comparability of results with those produced earlier in CARDIA.

e. **Urinary albumin excretion** - The kidneys in both blacks and whites are susceptible to blood pressure-related target-organ damage even at blood pressure levels within the “normal” or “pre-hypertension” (<140/90 mmHg) range [163]. Serum creatinine has been used as a crude measure of overall renal function in many studies. Urinary
albumin excretion has been proposed as a sensitive and reasonably specific test to identify impaired renal function prior to gross creatinine elevations. This test has the potential to improve risk stratification for persons with high normal to elevated blood pressures.

A small amount (<20 µg/min) of albumin is normally excreted by the kidney. When urine albumin excretion exceeds 200 µg/min, it is usually detectable by dipstick methods. Mildly increased urine albumin excretion, “albuminuria” in the range of 20 to 200 µg/min often is undetectable by such methods. Quantification of urine albumin excretion has been performed in several ways including total 24 hour excretion, amount excreted per mg of creatinine, and the amount of albumin excreted per unit of time. The albumin excreted in the urine over a 24 hour period is considered the “gold standard” for assessing albumin level and defining albuminuria [164]. “Spot” urine collections indexed to creatinine excretion have also been used in other studies [65]. A study reported high correlation between the albumin:creatinine ratio and the 24-h urinary albumin excretion for men (0.949) and women (0.942). The correlation was lower between the urinary albumin concentration with the 24-hour urinary albumin excretion (0.881 for men and 0.816 for women [164].

Albuminuria is a risk factor for kidney failure [165], stroke [166] and cardiovascular and all-cause mortality [167], particularly in diabetic and hypertensive patients. Urinary excretion of albumin above 20 µg/min correlates with a higher prevalence of blood pressure-related target-organ damage (i.e., greater left ventricular mass (LVM) and glomerular filtration rates) in cross-sectional studies and portends a higher incidence of cardiovascular morbidity and mortality, even in non-diabetics [65, 168, 169]. Increased levels of urine albumin excretion probably reflect glomerular damage (i.e., increased permeability) which may be part of a generalized membrane defect leading to a "vascular leak" and possibly to hypertension. In TOMHS, hypertension treatment with contemporary antihypertensive drug classes reduced "spot" but not overnight (8 hours) urine albumin excretion [65]. “Spot” urine albumin excretion also was correlated positively to age, left ventricular function, and systolic blood pressure. Other studies have documented positive associations of urine albumin excretion with black race, postprandial glucose and insulin concentrations [169, 170] and triglycerides [169]. CARDIA reported that men and blacks had higher “spot” urine
album excretion than women and whites. The strongest correlates of microalbuminuria were diabetes and blood pressure, although only 37% of cases of microalbuminuria occurred in people with either of these conditions [171, 172].

Because of the relatively young age of the CARDIA cohort, blood pressures are lower on average and ranges wider than clinic series, thus providing a unique opportunity to describe the epidemiology of urine albumin excretion and the associations with CVD risk factors and longitudinal blood pressure change. The overall prevalence of albuminuria in the CARDIA cohort is likely to be 10% or less, and elevated excretion may cluster among persons with diabetes mellitus, hypertension, left ventricular hypertrophy, and abnormal lipoprotein levels. Nevertheless, it is of interest to measure urinary albumin excretion - putatively a marker for a generalized disruption of the endothelial barrier - in the entire CARDIA cohort, particularly given our keen interest in blood pressure and atherosclerosis precursors and the central role of the kidney in blood pressure regulation. Also, there are no biochemical risk markers for renal disease/dysfunction in the CARDIA data set with the exception of creatinine. Albuminuria in small selected hypertensive study samples of older ages than the CARDIA participants is associated with target-organ damage such as left ventricular hypertrophy as well as salt-sensitivity [173], insulin resistance, and an atherogenic lipoprotein profile [173]. Yet, there is no reason to believe that such associations are specific to those with blood pressure above arbitrary blood pressure cut-points as used to define hypertension. Furthermore, there are limited opportunities for causal and mechanistic inferences when making cross-sectional associations.

Albuminuria measured in the entire CARDIA cohort at the Y10 Exam allowed for establishment of a baseline of albumin excretion in the cohort and occurrence of clinical events with a clear understanding of the temporal relationships. A repeat measurement at Y15 permitted longitudinal trend data to be examined.

Urinary albumin excretion (as calculated from spot measurement of urinary albumin and urinary creatinine) will again be measured at the Y20, using methods comparable to those used at the Y10 and Y15 Exam. The Y10 value will be used as a baseline to establish change, or to focus on albuminuric or normoalbuminuric people. Albuminuria at Y10 was observed to be closely related to race (blacks higher), blood pressure (even at levels as low as 120 mmHg systolic), and diabetes. All these will be
followed closely in the Y20 Exam. In addition, having a third time point (Y10, Y15, and Y20) will permit assessment of linear versus nonlinear trends in urine albumin both overall and in high risk subgroups.

Besides its role as a direct measure of kidney function, albuminuria is viewed as a marker of endothelial dysfunction. It is expected to correlate with coronary calcification prevalence and progression, as well as with blood markers of inflammation, hemostatic activity, and oxidation, and with factors such as diet and smoking.

8. Pulmonary Function

Low levels of lung function predict increased all-cause mortality [174] and have been associated with fatal MI [175, 176], hypertension[130], and fatal stroke [177]. Reduced lung function is a powerful health status indicator. Ethnicity, gender, smoking, physical activity level, and BMI may be determinants of asthma [178]. CARDIA has collected respiratory history, risk factor, and pulmonary function data at four examinations, the most recent being the Y10 Exam. Using these data, we observed increases in FEV1 between age 18 and the early 20s, followed by a plateau, then decline, with these age-patterns modified by race, sex, smoking, and asthma [179]. Less favorable patterns were associated with childhood exposure to familial smoking, early onset of smoking, higher baseline BMI, and increasing BMI. Lower childhood socioeconomic status is related to reduced lung function [180]. Higher fibrinogen [157], indicating chronic mild inflammation, and lower total blood carotenoids, indicating susceptibility to oxidative stress, are also associated with less favorable age patterns in lung function. Lower levels of lung function at Y10 are associated with increased occurrence of microalbuminuria at Y10 and Y15, and CAC at Y15. We hypothesize that deterioration of lung tissue and glomerular endothelial tissue (reflected in albuminuria), and deposition of calcium in coronary atherosclerotic plaque are occurring simultaneously and are related to many of the same risk factors, including inflammation. Lung function data is collected again at Y20, providing unique opportunities to compare asthma risk and deteriorating lung function in young to middle-aged Black and white men and women, to assess the role of genetics and the environment, and to explore the relation to CVD.

Since the Y10 pulmonary function equipment (Collins Survey Spirometer with Eagle II Microprocessor) is outmoded, new equipment was selected and calibrated against that used
previously. For Y20, pulmonary function is measured by the OMI (Occupational Med, Inc., Texas) rolling seal spirometer, as used in NHANES. The protocol for participant testing will remain the same as in previous examinations. To assess the impact of upgrading our equipment, each site’s Y10 equipment will be tested against the new equipment using a series of standard waveforms and lung volumes [181, 182]. Comparability study results will provide calibration equations, if needed, for longitudinal analyses.

A comparability study of the present equipment against the new equipment is also to be performed on 25 volunteers. This is to be performed at the Pulmonary Function Reading Center in Utah, as early in the Y20 Exam as possible.

9. Psychosocial Measures

Accumulating evidence suggests that psychosocial factors play a role in the etiology of hypertension and CVD [183, 184]. Individuals who are less educated, have lower income, or occupy jobs of lower prestige are at elevated risk for all cause and CVD mortality relative to their higher socioeconomic status (SES) counterparts. Individuals employed in more demanding or unrewarding jobs are at elevated risk for hypertension, CVD and all-cause mortality. Persons who are hostile, anxious, and depressed are at elevated risk for clinical coronary events.

Evidence for the prospective role of psychosocial factors comes largely from studies of middle-aged white men with clinical CVD. We have reported from our young adult cohort that hostile attitudes predict CAC; [53, 185] depressive symptoms predict incident hypertension in blacks: [186] greater job strain predicts incident hypertension in whites; [53] a sense of time urgency and impatience predicts hypertension in both ethnic groups; [187] and health care access predicts smoking cessation [188]. With IMT and the re-assessment of CAC, we will obtain important new information about psychosocial risk involved in early disease in a more diverse population. Since total exposure over time to a psychosocial risk factor such as hostility or depression is likely to be a better estimate of risk factor burden than a single measure, psychosocial measures are collected at Y20 and will be summed along with the same measures obtained at earlier examinations. In other words, like person years of exposure to smoke, we can develop indices of person years of exposure to stress.

We can also use the available genetic data to conduct gene-environment analyses, exploring interactions with psychosocial measures such as SES, depressive symptoms, job strain, and other lifestyle variables including BMI and physical inactivity. CARDIA’s detailed psychosocial and
biological longitudinal data will help answer questions on the independent and combined effects of SES and Black ethnicity on subclinical CVD.

In addition to individual-level psychosocial risk factors, another important area will be contextual and multilevel analyses. There is increasing evidence that neighborhood quality is significantly associated with health, independent of individual risk factors and SES.[189] Given the utility of individual perception in characterizing neighborhood quality, we will also explore participants’ perceptions of neighborhood, using validated instruments of perceived neighborhood crime, lack of amenities, environmental problems, and cohesion.

Twenty or thirty minutes of time is dedicated to psychosocial assessment, with all being assessed via self-administered questionnaire, except for Sociodemographic characteristics which are collected via interview. The assessments being collected are noted below with exam years where previously used:

- Sociodemographic characteristics (Form 3) – all years
- CES-Depression (Form 36) – Y5, 10, and 15
- Quality of Life SF-12 (Form 65) – Y15
- Subjective Standing (Form 66) – Y15
- Framingham Type A (Form 16) – Y0 and Y2
- Anger in Expression (Form 38) – Y5
- Chronic Burden (Form 64) – Y15
- Caregiving Stress (Form 75) - new
- Social Network (Form 63) – Y15
- Social Support and Conflict (Form 62) – Y15
- Loneliness (Form 14B) – Y0 and Y2
- Goal-Striving Stress (Form 74) - new
- Neighborhood Cohesion (Form 56) - new

10. Sleep

Millions of Americans obtain inadequate sleep because of life style factors, while millions of others suffer from chronic sleep disorders such as sleep-disordered breathing (SDB) [190].
Growing evidence suggests that lack of sleep and SDB may be important etiologic factors for the development of CVD risk factors and clinical disease [21, 191-194]. CARDIA participants who reported six or fewer hours of sleep at Y15 had a higher prevalence of hypertension and obesity than those with longer sleep duration[195]. Given the high prevalence of inadequate sleep and sleep-disorder breathing, and the recently discovered associations with CVD risk factors and disease, it is important to assess chronic sleep disorders and determine their associations with known risk factors and subclinical endpoints such as CAC and IMT.

Selected questions from the baseline Sleep Heart Health Study questionnaire, specifically questions #4-5 (usual hours of sleep on weekdays and weekends), #8-17 (snoring and sleep apnea), and #20 (Epworth scale of daytime sleepiness) are used. This questionnaire is designed for self-administration with the subset of questions proposed here requiring about 5-8 minutes.

11. Medical History, Hospitalizations and Medications

Medical history information is extremely important to assess in CARDIA. First, CARDIA began as a study of cardiovascular risk development. Thus, assessment of such conditions as self-report of physician diagnosed hypertension, and use of medications for its treatment, are important for achieving this objective, and serving in analyses as endpoints[196]. In addition, use of certain medications or presence of specific medical conditions could affect lifestyle, cardiovascular risk factor levels, or increase risk for clinical outcomes. For example, diabetes, assessed in CARDIA by both self-report and measurement of serum glucose, and self-reported liver and kidney disease were among the independent predictors of mortality in CARDIA [197].

CARDIA participants are asked to complete questionnaires and interviews in order for the study to thoroughly review self-reported health conditions and medication use (including adherence).

a. Tobacco use - Smoking, particularly cigarette smoking, has long been known to be a major risk factor for cardiovascular events as well as for subclinical disease [16, 35]. Smoking is also known to increase inflammation, a process known to be intimately involved in all stages of atherosclerosis[198]. In addition, the relationship between genetic variants and cardiovascular risk differs in the context of age, gender and environmental exposures such as smoking.

Not only is smoking important to CVD endpoints, it also is a significant determinant of other endpoints and disease measures important to CARDIA, including
asthma and pulmonary function. For example, using respiratory history, risk factors, and pulmonary function data at four examinations, CARDIA observed increases in FEV\textsubscript{1} between age 18 and the early 20s, followed by a plateau, then decline, with these age-patterns modified by race, sex, smoking, and asthma [179]. Further, cigarette smoking was related to all cause mortality in analyses of early mortality in CARDIA [197]. Even though CARDIA smokers are still young with relatively few pack years of exposure, we observe a relative risk of 1.8 over 15 years of follow-up for a composite of smoking-related clinical events for baseline smokers compared to baseline never smokers who were not exposed to passive smoke [199].

A particular strength of CARDIA is its ability to examine potential race/ethnicity, gender, and SES because of its diverse cohort. These issues are extremely important in analyses of health behaviors and lifestyle factors as they pertain to outcomes of interest to CARDIA and are exemplified by the findings of differences in smoking patterns and cessation rates by ethnicity, health care access and other factors [200] and by greater passive smoke exposure in those with lower education, even in the face of widespread restrictions on smoking in public places [201]. Thus, detailed data on smoking exposure is collected in CARDIA.

b. Alcohol use - Alcohol intake is known to affect several cardiovascular risk factors and has complex relationships with CVD. Among drinkers who do not binge drink, the lowest mortality rates are among light to moderate drinkers, with higher rates among abstainers and heavy drinkers [202]. There is evidence that “binge” drinking (consumption of five or more drinks on any given occasion) negates the benefits of moderate alcohol consumption, leading to higher rates of CHD in most studies [203-207]. In addition, ethnicity [208], gender [202, 209, 210], and type of alcohol beverage [211] also influence the relationship between alcohol consumption and CHD in some studies.

The mechanism(s) for potential benefits of alcohol on CHD risk include effects on risk factors –(HDL cholesterol [212], clotting and platelet aggregation [212], systemic inflammation [213], endothelial function [214, 215], and resistance of myocytes to ischemic injury [216]). On the other hand, it has mixed effects on glucose tolerance [217-219], and detrimental effects on blood pressure [220, 221]. In CARDIA data from Y15, moderate alcohol intake was not associated with lower risk of CAC, while binge
drinking was related to presence of CAC, with the association being strongest in Black men [222].

Alcohol intake is assessed for different types of beverages (wine, beer, liquor) and then summed for total consumption. Questions on binge drinking are also asked.

c. Non-medical drug use - “Non-medical drug” use is prevalent in the U.S., particularly in young people. Use patterns differ for different substances and demographic groups. In CARDIA, cocaine use is more common in men, smokers, drinkers, those with lower income, and less education. Use of these substances can have profound effects on health. Cocaine has acute effects on the heart and circulatory system, including sympathomimetic increases in BP, pulse, contractility and vasoconstriction, and acutely increases the risk of MI. Non-medical drug use also has other profound effects on health. In CARDIA, non-medical drug use was associated with health related quality of life measured at Y15. Reduced mental health-related quality of life (HRQOL) was associated with cocaine use, physical HRQOL with opiate use, within-person general health decline with methamphetamine use[223].

Non-medical drug use is collected for recent and lifetime use for several substances.

d. Interim hospitalizations and serious illnesses and injuries - CVD outcome ascertainment is important in a study of longitudinal change in cardiovascular risk. While CVD morbidity and mortality has been relatively low due to the young age of the cohort initially, there has been a steady increase in the number of possible endpoints requiring adjudication in CARDIA. Outcomes that occur in individuals of young age at onset are frequently suspected to have some heritable trait causing their increased risk. Thus, they are important to study such as CARDIA that is assessing gene-environment interactions on CVD risk. In addition, standardized assessment of other outcomes of interest, such as hospitalized asthma, gives specific information on diagnosis and therapy.

Standardized self-report, or informant report in case of death, of potential outcomes of interest is collected at each exam and each annual contact of the cohort. At Y20, these outcomes include hospitalization for acute MI, outpatient vascular procedures for peripheral arterial disease, and potential cardiovascular death. Once interview
information is collected, it is coded to determine if medical records are needed and if so, they are adjudicated by physicians using standardized criteria.

e. Women’s reproductive health - The perimenopausal transition is relatively understudied, in part because it is difficult to define. It is known that gradual declines in estrogen occur, as well as increases in gonadotrophins. Symptoms include changes in cycle length and menstrual flow, short term amenorrhea and irregularity, as well as hot flushes and other menopausal symptoms. Concomitantly, a myriad of cardiovascular risk factor changes occur that may potentiate future cardiovascular disease. The transition may occur earlier in women who smoke compared to those who don’t, due to the anti-estrogenic effects of cigarette smoking [224].

The women in CARDIA are 38-50 years old during Y20. A number of women will be postmenopausal or experiencing the perimenopausal transition. This is an optimal cohort to examine the effects of menopause on cardiovascular disease risk, including the opportunity to examine longitudinal associations with subclinical disease, using Y15 and Y20 CAC.

Women will again complete a detailed questionnaire on menstrual history, including intervals between periods, menstrual flow, instances of amenorrhea, symptoms (e.g., hot flashes, vaginal dryness), and use of oral contraceptives and hormone therapy.

12. Physical Activity

Physiologic studies have shown that physical inactivity is associated with the risk of coronary heart disease (CHD) [225, 226] while total physical activity, running, weight training and rowing are inversely associated with the risk of CHD [227].

Physical activity is a behavior consisting of many different components including occupational activities, discretionary activities, optional household tasks, socially desirable activities and activity for physical fitness and the promotion of health [228]. In the CARDIA study it is important to measure which activities are prevalent, and how this activity spectrum changes as the CARDIA population ages. Further it is important to reliably rank people for overall level of all and of intense physical activity, and to determine the impact of types and levels of physical activity on the prevention of cardiovascular events.
Furthermore, it is important to determine the role of ethnicity on the association physical activity-cardiovascular disease particularly in the hypertension area. Some studies have shown that blacks who performed moderate to vigorous activity have less hypertension than their inactive peers [229, 230].

The existing questionnaires which appeared to be best suited for use in CARDIA were the Minnesota Leisure Time Physical Activity Questionnaire [231] and the Seven Day Physical Activity Recall[232]. The former, which spans 12 months, has been validated against physiologic endpoints, but has not been shown to be adequately reliable on reapplication, and takes too long to administer. The latter spans only seven days, has considerable accuracy, has passed some validity studies and is suitably brief [233]. However, it too suffers from low repeatability since one week's physical activity is often not representative of general patterns.

In devising a briefer new questionnaire, it was theorized that people could accurately recall or summarize for themselves what and how much activity they do within broad categories. Therefore details were avoided as being both poorly repeatable and irrelevant.

Using an extensive database from the Minnesota Heart Health Program, tabulations were made of activity participation for men and women aged 25-29. Based on these tabulations questions were worded for sets of activities which might be logically classed together.

At the Y20 Exam, the original CARDIA physical activity history is asked, as in prior exams, including comparison to others of the same age and sex, and 13 sets of questions about various moderate and vigorous intensity activities. As before, TV watching is queried. In addition, because it has been observed that activity performed mostly by women (e.g., household chores and child care) [234] is related to mortality outcome [235], household chores and childcare activities are queried.

13. Genetic analyses

While familial aggregation of CHD and the genes influencing several risk factors, including lipids, blood pressure, and obesity have been well studied [236-239] the genes underlying CHD susceptibility remain largely unidentified. The large proportion of asymptomatic individuals with CHD [240-242] and the potential for early risk factor modification make the study of subclinical disease appealing. Multiple studies of familial aggregation of IMT and CAC provide heritability estimates of .40 for CAC [240, 241], ranging from .30 to .90 for IMT [243-246], with concomitant identification of candidate genes and candidate gene regions contributing to
development of subclinical atherosclerosis [245, 247, 248]. Although the molecular mechanisms contributing to the development and progression of atherosclerotic lesions are being articulated [249], studies are needed beyond family studies and molecular mechanisms to determine the impact of the identified candidate genes on the risk for subclinical atherosclerosis in the population. CARDIA has several publications reporting associations with genetic markers [250-254].

CARDIA collected buffy coat for DNA extraction at Y5, Y10, and Y15. At Y15, cryopreserved cells were collected for future transformation and immortalization. Buffy coat is collected at Y20 and stored for later DNA isolation. We have extracted DNA on all Y10 participants, which constitutes the primary source of genetic material for the current CARDIA studies. We will supplement this resource with material from Y20 in order to obtain as complete a genetic dataset as possible.

CARDIA will genotype approximately 50 polymorphisms on identified genes, as agreed upon by the Genetics Subcommittee and approved by the CARDIA Steering Committee. These genes, which may be implicated in the biology of CAC and IMT or may influence their risk factors, belong to 4 broad categories: bone mineralization regulation; obesity; dyslipidemia; and “others,” including blood pressure regulation. These genotyping studies will build upon and expand the genetic data already collected in CARDIA. Using association study methods, we will test the hypothesis that common polymorphisms and patterns of variation in these genes are risk factors for subclinical atherosclerosis in young adults. These methods are increasingly being used to investigate disease etiology and are well suited to studies of complex multi-factorial traits with weak-to-moderate effects [255, 256].

Two complementary approaches have been advocated for such studies: [257] the first directly tests all of the common, known functional variants in a candidate gene for disease association. Collins et al. [257] have argued that only two to three variants in any coding sequence will be sufficiently frequent for such studies. The second examines a dense collection of single nucleotide polymorphisms (SNPs) located throughout or very close to the gene of interest, relying on linkage disequilibrium between the set of SNPs and the disease-causing variant while examining linear combinations of polymorphisms (haplotypes) for disease association. We propose to use features of both methods to test hypotheses about the influence on subclinical atherosclerosis of sequence variation in the genes involved in obesity, dyslipidemia, blood pressure and bone mineralization regulation.
The relationship between genetic variants and CHD risk has been noted to differ in the context of age, gender, environmental exposures such as smoking, diet, etc., or other genes. Investigations of such context-dependent genetic effects or “gene-environment interaction” and “gene-gene interaction or epistasis” contribute to a more complete characterization of disease etiology, improving accuracy and precision in the assessment of genetic and non-genetic influences on CHD risk. Our heightened understanding of these interactions has important public health implications, permitting more accurate CVD risk assessment, and providing a rational basis for recommending behavior modification and disease prevention strategies.

Given its extensive longitudinal data on lifestyle, behavior, and biochemical risk factors, CARDIA provides a unique opportunity to examine the context-dependent effects of candidate gene variation on subclinical measures of atherosclerosis. Figure 1 illustrates examples of context variables which may interact with variation in selected candidate genes to influence subclinical disease and its risk factors. For example, one such question is whether the Trp64Arg missense mutation of the β3-adrenergic receptor interacts with longitudinal weight gain to drive greater central fat deposition, in turn leading to increased risk of type 2 DM. Another is whether the IL-6-174CC genotype affects LDL-C response to statin therapy and the level of risk reduction in CHD accomplished with treatment [258]. The larger number of outcomes such as central obesity and hypertension anticipated at the Y20 Exam will provide additional power to examine these types of hypotheses within race-gender subgroups, and other demographic or environmental strata.

**Figure 1- Examples of genes and environmental strata which may interact to influence subclinical coronary artery disease**

**Candidate Genes**
- Osteopontin
- Matrix GlA
- Leptin
- Angiotensinogen
- β3 Adrenergic Receptor Lipoprotein
- Lipase Hepatic Lipase CETP

**Environmental Strata**
- Race
- Gender
- Age
- Body size
- Diet
- Physical activity
- Smoking
- Alcohol use
- Socioeconomic status
- Urinary Na & K
- Medication use

**Intermediate Risk Factor**
- Obesity
- Insulin Resistance
- Hypertension
- BP reactivity
- Increased LV mass
- Central fat patterning
- Dyslipidemia
- Diabetes
- Microalbuminuria
- Serum Creatinine
- Inflammation markers

**Subclinical disease**
- CAC
- IMT
a. **Candidate gene selection in relation to CAC and IMT**-- The table below lists examples of candidate genes which may be proposed for study. The list is not static and will be continually reviewed by the CARDIA Genetics Subcommittee during the course of the study to take into account new information.

<table>
<thead>
<tr>
<th>Bone Mineralization</th>
<th>Obesity</th>
<th>Dyslipidemia</th>
<th>Hypertension; Others</th>
</tr>
</thead>
<tbody>
<tr>
<td>Matrix Gla</td>
<td>Leptin</td>
<td>Hepatic Lipase</td>
<td>Angiotensinogen</td>
</tr>
<tr>
<td>Osteoprotegerin</td>
<td>Leptin Receptor</td>
<td>Lipoprotein Lipase</td>
<td>Angiotensin-Conv Enzyme</td>
</tr>
<tr>
<td>Osteoprotegerin Ligand (RANKL)</td>
<td>Neuropeptide Y</td>
<td>Endothelial Lipase</td>
<td>Endothelial Nitric Oxide Synthase</td>
</tr>
<tr>
<td>Osteoprotegerin Ligand Receptor [131]</td>
<td>NPY Receptor 1 and 5</td>
<td>Cholesteryl Ester Transfer Protein</td>
<td></td>
</tr>
<tr>
<td>Osteopontin</td>
<td>Melacortin 4 Receptor</td>
<td>Paraoxonase 1</td>
<td></td>
</tr>
<tr>
<td>Osteocalcin</td>
<td>Pro-opiomelanocortin</td>
<td>B3-Adrenergic Receptor</td>
<td></td>
</tr>
<tr>
<td>Osteonectin</td>
<td>Adiponectin</td>
<td>Apolipoprotein AV</td>
<td></td>
</tr>
<tr>
<td>Matrix Metalloproteinase 3</td>
<td>Ghrelin</td>
<td>Apolipoprotein AI</td>
<td></td>
</tr>
<tr>
<td>Matrix Metalloproteinase 9</td>
<td>PPAR alpha</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bone Morphogenic Protein 2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bone Morphogenic Protein 4</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

b. **Polymorphisms selection within candidate genes**— Initially, we propose to study genes in which multiple polymorphisms have already been identified. From previous research, we know that any single DNA polymorphism within a gene sequence is unlikely to capture adequately the overall variation in that gene [259, 260]. Success in identifying associations thus will likely rest on examining a dense collection of SNPs since the distance over which significant linkage disequilibrium can be detected varies greatly within a gene region; however, it is difficult to predict the ideal density of SNPs required in these studies. In addition, we must consider and make appropriate correction for multiple comparisons when performing analyses with a large number of SNPs. Statistical analysis based on haplotypes addresses these problems very well [261]. Since the extent of linkage disequilibrium between marker locus and trait locus is not known *a priori*, we propose to genotype a minimum of 3 to 5 polymorphisms in each sequence of the selected genes involved in obesity, dyslipidemia, hypertension, and bone mineralization. We will give priority to those polymorphisms of recognized or inferred functional significance (e.g., affecting mRNA regulation, or protein structure or function). As the ongoing efforts for large-scale discovery of DNA sequence variation in candidate genes intensify, and data on the linkage disequilibrium structure of this variation in black and white populations becomes available (see Program for Genomic Applications, for example), we will refine our list of polymorphisms based on...
the most current data. In both single-locus and haplotype analyses, we will test these polymorphisms for associations with subclinical disease.

c. Genotyping Methods - The protocols and quality control procedures in practice in the CARDIA DNA laboratory have evolved from experience and continued participation in large collaborative and inter-disciplinary studies. For each polymorphism, the choice of method is based on feasibility (i.e., nature of the polymorphic site and the sequence surrounding it), and quality of the data generated during the optimization procedure. Two methods will be utilized: preferentially the TaqMan assay (Perkin Elmer Applied Biosystems), and Matrix-Assisted Laser Desorption Ionization Time of Flight (MALDI-TOF) mass spectrometry genotyping, using the MassArray system (Sequenom). These procedures provide accurate, high throughput and cost-efficient genotyping, and are well suited to the scope of this study.

i. Mass-spectrometry genotyping - Matrix-Assisted Laser Desorption Ionization Time of Flight (MALDI-TOF) mass spectrometry (MS) will be preferentially used for high throughput genotyping because it provides a fully-automated procedure with high level multiplex analysis (up to 10 SNPs can be routinely genotyped from a single assay). The method is based on oligonucleotide extension reactions (mini-sequencing). It first requires amplification by PCR of a short sequence (40 to 100 base pairs) containing the variant site(s) in a minimal reaction volume (5µl).
Following PCR, the reaction mix is treated with shrimp alkaline phosphatase in order to digest unincorporated dNTPs and PCR primers. This is followed by a mini-sequencing reaction in which an additional oligonucleotide primer is annealed immediately upstream of the polymorphic site, extended over it, and rapidly terminated. Use of the appropriate dideoxynucleotide triphosphates (ddNTPs) and DNA polymerases, such as Thermosequenase, that incorporate ddNTPs with high efficiency, results in rapid termination of extension. This reaction generates allele-specific primer extension products that are generally 1-4 bases longer than the original oligonucleotide. Multiplexing can be achieved at the level of oligonucleotide extension. Extension primers and products utilized in multiplexed mini-sequencing reactions are designed so that the masses of un-extended and extended primers can be distinguished from one another. Masses of the mini-sequencing reaction products are assessed using delayed extraction MS.
The laboratory currently uses the MassArray system from Sequenom. This turn-key commercial MS genotyping system incorporates the elements described above and is extended to facilitate high-throughput by including software for design of genotyping assays and for allele-calling to automate extraction of genotype information from mass spectra, and methods to simplify preparation of samples for MS genotyping.

ii. *TaqMan genotyping assay* - TaqMan genotyping assays will be used in complement to the MS method when assays could not be designed in an efficient manner by the MS method. This method has been used for genotyping of the limited collection of SNPs in the CARDIA study. Single base pair substitutions as well as short insertions/deletions can be genotyped by this assay. The TaqMan assay is a 3-step process. First, allele-specific fluorogenic probes, approximately 20 bp in length and labeled at the 5’ and 3’ ends with fluorescent reporter and quencher dyes, respectively, hybridize to the PCR amplified-target DNA in a sequence-specific manner. When the probes are intact, the proximity of the reporter dye to the quencher dye suppresses the fluorescent activity of the reporter dye. Second, during PCR amplification, flanking PCR primers are extended in the 3’ direction by Taq DNA polymerase activity. The 3’ end of the fluorogenic probes is blocked to prevent extension. Third, as the DNA strand extends from the primer to the bound probe, the 5’ nuclease activity of Taq DNA polymerase cleaves the bound probe at the 5’ end and releases the reporter dye, causing an increase in fluorescent intensity of the reporter dye. Each allele-specific probe is labeled with a different reporter dye, usually TET and VIC. Primers for PCR amplification and custom fluorescent probes are designed using the Primer Express™ software (ABI, Foster City, CA). PCR reagents available as a kit and dye-labeled custom probes are purchased from ABI. After PCR amplification, analysis of fluorescent signals of the distinct reporter dyes leads to automated genotype determination with a commercial software. An increase in only one of the fluorescent signal indicates that the sample is homozygous for either the VIC- or TET-specific allele, while an increase in both signals indicates that the sample is heterozygous.

d. *Genotyping QC procedures* - The laboratory rigorously follows established QC protocols and procedures. Those include the use of robotic instrumentation, a
standardized barcode system for sample identification and tracking, automated allele calling procedures with proprietary software (see above), verification of the genotype data for Hardy-Weinberg equilibrium (HWE), and monitoring of genotyping error using replicate pairs on a 5% random sample. The laboratory has an excellent track record of high quality genotyping in CARDIA and other studies with a low error rate (average $\kappa$ statistic – representing a measure of agreement corrected for chance agreement: 0.87).

F. EXAM IMPLEMENTATION

1. Recruitment

Retention of CARDIA participants across the four Field Centers is a primary focus of the Y20 Exam period. Participants are seen approximately 20 years from the date of their original exam with a window of eligibility of $\pm$ two months. Scheduling participants will be sequential from the first person seen until the last participant over a twelve month period.

All participants in the Y0 Exam are eligible for the Y20 Exam. Pregnancy or lactation will not eliminate a participant from the entire exam, but may preclude participation in specific exam components. Pregnancy and/or lactation may extend the window of eligibility for a participant. Similarly, participants who have moved more than 50 miles from the clinic will be permitted an extended window of eligibility in order to maximize retention of the cohort and minimize the costs of retention. Reimbursements will be allowed within limits, at the discretion of the Field Center Principal Investigator, to bring back for examination participants who have moved more than 50 miles.

The Y20 recruitment plan is designed to re-examine at least 73% of the surviving CARDIA cohort examined at the Y0 Exam. The plan also provides for extensive monitoring of the process. Each Field Center is provided scheduling lists designed to achieve the completion of the re-examination within 12 months. Participants who have moved will be monitored so that special recruitment and cost planning can be made in advance. Clinic staff will make every effort to contain the costs associated with bringing moved participants to the clinic during the exam. A description of the initial contact and recruitment process can be found in the MOO, sections 2 and 14.
Every effort is made to maximize the return rate of the CARDIA cohort. The efforts include performing a partial exam on participants who refuse or are unable to participate in a full exam; examination at another Field Center clinic that is more convenient to the participant; and, a telephone interview if a participant is unable to participate in any other way. Two of the Field Centers may institute at-home visits for those unable to attend the exam, if considered necessary in the final few months of the Exam. The CARDIA Study is aiming at as high a rate as possible in order to maximize the long term potential of the study to chart the natural history and/or evolution of CVD risk factors.

2. Exam Flow

A sample form for tracking clinic flow is provided to each Field Center via the Scheduling System. Each clinic can determine their own flow based on their set-up, staffing and needs. The following general flow should typically be followed when planning the exam flow for each participant – detailed information can be found in the Y20 MOO – Section 2.

   a. Greeting, including informed consent and pregnancy screening (urine)
   b. Blood pressure (BP) measurements before blood draw and/or any potentially-stressful interview.
   c. Phlebotomy - Initial blood draw must occur before 10:30 am.
   d. Anthropometry
   e. Self-administered questionnaire
   f. Interview
      i. Diet Interview
   j. Carotid Ultrasound
   k. Pulmonary Function
   l. CT or CT scheduling
   m. BP comparability study, if participating
   n. Exit interview

3. Prioritization schedule

CARDIA Y20 Protocol 23Jan06
If a participant refuses to stay for the entire exam (i.e. *I only have one and a half hours available*) then the performance of exam components are prioritized as follows:

a. Highest Priority Level

**NOTE:** CT is out of clinic for 3 of the 4 Field Centers and is in a separate category than core components, but is of very high priority (Forms 73 and 76).

i. Consent

ii. Exit Interview (Follow-up contact)

iii. Blood Pressure (Form 2)

iv. Fasting Phlebotomy (Form 5) **and** Urine (Form 51)

(priority based on groupings below)

<table>
<thead>
<tr>
<th>Group 1</th>
<th>Group 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>• lipid profile</td>
<td>• hsCRP</td>
</tr>
<tr>
<td>• glucose</td>
<td>• IL6</td>
</tr>
<tr>
<td>• insulin</td>
<td>• fibrinogen</td>
</tr>
<tr>
<td>• creatinine</td>
<td></td>
</tr>
<tr>
<td>• urinary albumin/creatinine</td>
<td></td>
</tr>
</tbody>
</table>

v. Carotid IMT Ultrasound (Form 77)

vi. Height, Weight, Waist Girth (Form 20)

vii. Medical History (Form 8)

viii. Interim Hospitalizations (Form 31)

ix. Sociodemographics (Form 3)

x. Tobacco Use (Form 10 and Form 9-TOB)

xi. Pulmonary Function Test

b. Second Priority Level

i. CES-D (Form 36)

ii. Diet (Form 6 and Form 48)

iii. Physical Activity (Form 18)

iv. OGTT (Form 51)

v. History of Lung Problems (Form 12)

vi. Sleep (Form 67)

c. Third Priority Level

Other **Interviewer-Administered Questionnaires**
i. Alcohol Use (Form 7)
ii. Follow-up forms to Form 8 (Form 9 MED-ADH, Form 9 MED, Form 9 MED-ASP, Form 9 MED-BCP, Form 9 TB, Form 9 OVA, Form 9 PREG)

Other Self-Administered Questionnaires
i. Women’s Reproductive Health (Form 68)
ii. Non-Medical Drug Use (Form 17)
iii. Quality of Life (Form 65)
iv. Subjective Standing (Form 66)
v. Framingham Type A (Form 16)
vi. Anger in Expression (Form 38)
vii. Chronic Burden (Form 64)
viii. Caregiving Stress (Form 75)
ix. Social Network (Form 63)
x. Social Support and Conflict (Form 62)
xii. Loneliness (Form 14B)
xii. Goal-Striving Stress (Form 74)
xiii. Neighborhood Cohesion (Form 56)
xiv. Dietary Habits (Form 79)
xv. CSQ Medical History (Form 78)

4. Quality Control

To ensure high quality data, quality control procedures developed during prior CARDIA exams are used. These include:

a. Initial Field Center personnel were centrally trained and certified by CC staff, and locally recertified during the exam.

b. Anthropometric technicians are evaluated monthly using duplicate measures from a 7% random sample of participants to assess inter- and intra technician variability. Terminal digit distribution is used to estimate technician’s performance monthly as well.

c. Overall Field Center performance is evaluated by a site visit early in the exam cycle and monthly assessment of forms and logs.

d. Laboratories are evaluated using blinded pairs results from a 7% random sample of participants.
e. The Diet Reading Center reviews 10% of audiotapes of diet interviews monthly and assesses these for protocol adherence and the degree of misrecorded information. As well, the Diet Reading Center will review one interview per nutritionist each month to provide individual staff feedback.

f. The Field Center Clinic Coordinator reviews one regular interview per interviewer and completes a rating form monthly. A Coordinating Center staff member reviews the tape and completes an evaluation form to provide Field Center Principal Investigator feedback.

g. Pulmonary function acquisitions and readings are reviewed by the Reading Center and compared to the data recorded on Form 80. All images are assigned the quality scores that are monitored at the Coordinating Center monthly to evaluate PFT technician’s performance. Re-readings of the images will be done in 5% to assess intra- and inter reader variability.

h. Quality control of CT data will include monthly regular scanning of standard phantoms (CAC) and duplicate readings of 5% CTs - with positive and negative CAC ratio as 2:1. All images are assigned quality scores which are monitored at the Coordinating Center on the monthly basis. The CTRC will analyze Y15 vs. Y20 CT decreases and large increases in the calcium score as well as reread any large differences (≥3 SD) between two duplicate scans collected at Y20.

i. Carotid Ultrasound tapes will be reviewed by the Ultrasound Reading Center (URC) and given quality ratings. Technician performance will be evaluated doing 3% of re-measurements at the Field Centers. Technician performance reports will be given to Field Center PIs on a regular basis. To estimate inter- and intra-readers variability re-readings will be done at the URC in 5% of tapes.

j. Data entry quality will be assessed by utilizing duplicate entry of 10% of data forms (dual monitor and TeleForms) and monthly review of error rates.

k. All data will be subject to range and logic checks; calculation of the proportion of missing data; calculation of mean and SD for continuous variables with monthly trends; calculation of crude percentages for categorical variables with monthly trends.
1. Equipment maintenance and calibration is evaluated using quality control logs reported to the Coordinating Center on a monthly basis.

5. Referrals and Results Reporting

The study will utilize criteria for medical referral of abnormal blood pressure, blood lipids and glucose levels, GFR and urine albumin levels based on commonly accepted guidelines where available, e.g., the JNC7 for blood pressure and ATPIII for lipids. The table below summarizes the referral criteria established for this exam.

Summary of Y20 referral criteria, CARDIA Study

<table>
<thead>
<tr>
<th>Measure</th>
<th>Referral Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Systolic blood pressure</td>
<td>$\geq 140$ mmHg</td>
</tr>
<tr>
<td>Diastolic blood pressure</td>
<td>$\geq 90$ mmHg</td>
</tr>
<tr>
<td>Triglycerides</td>
<td>$&gt;500$ mg/dl</td>
</tr>
<tr>
<td>LDL cholesterol</td>
<td>$\geq 160$ mg/dl</td>
</tr>
<tr>
<td>Fasting blood glucose</td>
<td>$&lt;60$ mg/dl or $\geq 126$ mg/dl</td>
</tr>
<tr>
<td>2-hour post load glucose</td>
<td>$\geq 200$ mg/dl</td>
</tr>
<tr>
<td>GFR</td>
<td>$&lt;60$ ml/min/1.73 m²</td>
</tr>
<tr>
<td>Urinary albumin excretion</td>
<td>$\geq 300$ mg/g $[\text{estimated from } (\text{albumin mg/dl})/(\text{creatinine mg/dl})]$</td>
</tr>
<tr>
<td>IMT</td>
<td>a Doppler flow velocity value $\geq 250$ cm/sec on either side for immediate alert referral; Doppler flow velocity value $\geq 150$ and $&lt;250$ cm/sec on either side for regular referral.</td>
</tr>
<tr>
<td>Pulmonary Function</td>
<td>$&lt;80%$ of predicted (based on height, age, gender and ethnicity)</td>
</tr>
</tbody>
</table>

Participants will receive printed report results of clinically useful tests as noted in the table below. Copies of the results reports/letters can be found in the Y20 MOO, section 12.
<table>
<thead>
<tr>
<th>Measure</th>
<th>Given</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blood pressure</td>
<td>In clinic</td>
<td>Systolic and Diastolic blood pressure</td>
</tr>
<tr>
<td>Anthropometry</td>
<td>In clinic</td>
<td>Height, Weight, BMI, Waist circumference</td>
</tr>
<tr>
<td>Lipids</td>
<td>Form sent</td>
<td>Total Cholesterol, HDL, LDL, Triglycerides</td>
</tr>
<tr>
<td>Chemistries, OGTT</td>
<td>Form sent</td>
<td>Fasting glucose, 2-hour glucose</td>
</tr>
<tr>
<td>Urine</td>
<td>Form sent</td>
<td>Urinary creatinine, albumin, albumin/creatinine ratio and GFR</td>
</tr>
<tr>
<td>CT</td>
<td>Form sent</td>
<td>Coronary findings (other incidental findings only for those with positive findings at Y15 and Birmingham participants)</td>
</tr>
<tr>
<td>Diet</td>
<td>Form sent</td>
<td>% calories from fat; % calories from saturated fat; % calories from carbohydrate; % calories from protein; Total dietary cholesterol/1000 calories; grams of fiber/1000 calories; calcium</td>
</tr>
<tr>
<td>Ultrasound</td>
<td>Form sent</td>
<td>Doppler value - cm/sec</td>
</tr>
<tr>
<td>Pulmonary Function</td>
<td>Form sent</td>
<td>% of predicted (based on height, age, gender and ethnicity); FEV₁/FVC ratio</td>
</tr>
</tbody>
</table>

G. DATA MANAGEMENT

The data management system for the CARDIA Y20 Exam includes three main components:

1. A Field Center system for participant scheduling, form generation, scanning of forms to produce form image files and accumulation of the image files into sets for transmission to the Coordinating Center.

2. A data transmission system which automates the shipping of form sets from the Field Centers to the Coordinating Center and which returns tracking information and retention information to the Field Centers.

3. The data entry system for use at the Coordinating Center.

All data entry will be done at the Coordinating Center by a paid data entry staff. The data entry system automates much of the range checking and level 1 data checking formerly done.
with SAS programs. Based on range and logic checks, queries are automatically generated to the appropriate Field Center’s data tracking page for correction or verification.

1. Flow and Management of Data Forms

For exams at Y0, Y2, Y5, Y7, and Y10, the CARDIA study used distributed data entry systems with data entry performed locally at each of the Field Centers. For the Y15 Exam, we utilized a different approach with paper questionnaires and forms generated at each Field Center from software provided by the Coordinating Center. A similar system is being used for the Y20 Exam. On each exam form page, a printed bar code provides a unique participant ID (PID) and data form page. Once data are entered on the forms, the forms will be scanned electronically to image files at the Field Centers for transfer to the Coordinating Center and automatically merged into the image portion of the centralized database. Data entry is done at the Coordinating Center from the scanned image by one of two methods: a Verity TeleForm application or a dual-monitor application (the scanned form will be visible on one screen while data entry will occur on the other). The data is then directly imported into the centralized database. Redundancy and monitoring are incorporated into this system. The original paper forms will be archived at the Field Centers to ensure against loss of the scanned images.

Each Field Center will have access to the CARDIA Recruitment System. The Recruitment System allows on-screen access to participant data and scheduling, as well as a variety of reports used in the recruitment process. A Web-based tracking system which includes scheduling reports along with tracking logs for forms and participant samples is available on the CARDIA Internal Website.

The CARDIA Recruitment System includes a configurable reporting feature. The reporting feature will allow the user to filter the master list of participants by Field Center, Status, Category, Exam Type, and Name. Each participant is assigned a status based on the last action performed on that participant. Categories are manually assigned to each participant. Each Field Center will be provided with a set of predetermined category codes for the master participant list; however, Field Centers may create additional categories and assign them to the participants as well. Utilizing the filters, Field Centers will be able to produce several reports, such as the \( \geq 50 \) Mile List which includes participants who have moved 50 miles or more from their “home” Field Center (the site at which they participated in the Y0 Exam).
To ensure the integrity of data collected, the process must be monitored and data tracked carefully. The information collected in this exam will be analyzed and published to advance understanding about the development of CVD. The value of this information is a reflection of the care and attention paid to the completeness and accuracy of the data during its collection.

Both push and pull strategies of moving data from one point to another are utilized in creating a system to track and verify the status of data at each site, at any point in time. These sites include:

- Field Centers: Birmingham, Chicago, Minneapolis, Oakland
- Coordinating Center
- Assay laboratories: Chemistry, Lipids, Albuminuria/YALTA, Inflammatory Markers
- Storage laboratories: Solomon Park Research Institute Laboratory, DNA Storage and Genotyping
- Reading Centers: Diet Center, Ultrasound Reading Center, CT Reading Center, Pulmonary Function Reading Center

These sites are collectively referred to as “centers.”

The purpose of the Tracking System is to employ effective methods of data tracking for Y20 by utilizing technologically-advanced systems for maintaining data availability and sustaining accurate data status throughout the exam period. The aims of the Tracking System are to:

- assure the availability of complete and accurate data
- assure timely transfer of complete and accurate data
- store and archive data in a readily-retrievable manner

Computer based information systems for all aspects of data collection, transmission and entry have been developed by the Coordinating Center Data Information and Statistical Computing staff. All systems have been developed in the C++ programming language and make use of the standard Windows application programming interface. The web-based Tracking system is written in ASP. Data forms for the Y20 Exam are printed at the Field Center prior to the examination date of each participant. The data forms are personalized by having the PID printed at the top of each page, both numerically and as a bar code. In addition, each page of the
form is clearly identified with the PID, form number, page number, and version number for ease of use. Each participant's forms and ancillary data (e.g., pulmonary function test print-outs, etc.) should be filed in the participant's Y20 Exam notebook/file. Forms for a participant are to be checked for completeness, accuracy and legibility at the clinic before the participant leaves. After the forms are checked, they are to be scanned to produce electronic images that are automatically collected into sets for daily automatic transmission to the Coordinating Center. The original forms are to be filed immediately in the participant’s Y20 file to avoid damage or loss.

In addition, a set of routines to identify bar codes that were not read by the Verity TeleForm application has been written to assist in identifying and entering the form images within the Coordinating Center in order to minimize the forms that must be hand-entered. A Query system that allows for visible inspection of problems identified has also been implemented so that corrections can be entered into the system by a reviewer electronically.

All files are kept in a logical and consistent manner to provide accessibility for the duration of the Y20 Exam period and thereafter until deemed unnecessary by the study. Participant notebooks are filed in numerical order by PID and stored in a secure but accessible place and manner.

2. Electronic Storage and Management of Data

The electronic storage and management of the data begins at the Field Centers when the forms are scanned and prepared for transmission to the Coordinating Center. The data entry takes place at the Coordinating Center and the resulting data files are stored on a secure network server. The files are organized into a SAS database by the Statistical Computing Unit of the Coordinating Center. Backup of the data files is done daily and weekly backups are maintained in a secure off-site location to guard against loss in the event of a catastrophic failure of the Coordinating Center network.

3. Field Center System

The Field Center system is a native mode Windows NT application which utilizes the standard Windows graphical user interface and security. The systems provided to the Field Centers include a designated Scheduling System computer, interface so that additional Field Center computers can link to this system, color printer, laser printer and scanner and hand held bar code scanner. The system provides support for the following activities:
a. Participant Scheduling - The *Scheduling System* provides a one week window for the expected date based on each participant’s baseline examination date. The system provides a link between the PID and actual participant demographic information such as name, address and contact information. It has a calendar format so that participants scheduled for each day is clear for the clinic personnel. The *System* allows certain days to be blocked out so that no one is scheduled at a time or date during which the clinic is not open. It also graphically shows the week’s schedule against the “ideal schedule” to assist staff in tracking daily progress for retention goals.

b. Participant Event Tracking - The *System* keeps track of each participant’s scheduled clinic visits and the outcome of each visit. Thus, if a participant does not keep his/her appointment, the system can record this and return the participant to the pool of individuals who need to be scheduled. The *System* maintains a database of this information and can produce tracking and retention reports throughout the study period.

c. Data Sharing Between Field Centers - The *Recruitment System* maintains study-wide demographic databases of non-sensitive site-specific information. The *Scheduling System* works in conjunction with the data transmission system to provide synchronized information for the clinics. The data in the files associated with this synchronization are generated at the Coordinating Center and automatically uploaded to the Field Centers in the form of read-only files. Thus, if a participant makes an appointment to be seen at a clinic other than their home clinic (the one at which they were examined at Y0), all the demographic information needed for scheduling will be available in this database. In addition, a Field Center can query the systems at the other Field Centers to obtain a list of any of their participants who have been scheduled or seen at another clinic during the exam cycle.

d. Reports of Recruitment/Scheduling Progress – An extensive report generation capability is offered by the *Scheduling System*. Among the standard reports which are produced are the tracking and retention reports. With the synchronization of the recruitment information across clinic sites, all Field Centers can produce their own as well as study-wide reports for real-time comparison. The same reports will be posted on the CARDIA internal website for Committee members to review.
e. Production of Personalized Blank Forms – All Y20 Exam forms are generated at the clinic site on an as-needed basis. The forms are tailored for each participant with each page of the form having the PID number represented numerically and by bar code at the top of each page, and only those pages required for each participant are produced (e.g., male participants do not have forms pertaining to female only issues printed). The Field Centers each have a laser printer to print the forms. The system is flexible so that forms can be printed on a daily, weekly or monthly basis and can be generated in the order best suited to an individual clinic’s flow. Each page of the form is clearly labeled with the form number and page within the form information. Labels (with bar coded PID) for laboratory use, diet and Pulmonary Function CDs, ultrasound tapes and extra forms are printed on the color printer.

f. Scanning of Completed Forms for Transmission to the Coordinating Center and Archiving of Form Images at the Field Center - When any form for a participant's Y20 Exam is completed, it is scanned into data files for transmission of form images to the Coordinating Center for data entry. The Field Center system provides the user interface for this process. In addition, it collects the scanned images into a set of files on the scheduling computer which are later collected by the Coordinating Center automatically. Once scanned, the original forms are stored at the clinic in whatever manner their system utilizes.

g. Quality Control ID Generation - The master list of Quality Control IDs is generated by the Coordinating Center and the weekly assignments are done at Field Center level. The Field Center system generates the randomly selected list of participants who will be used for quality control (QC). During the early part of the examination period (first three months) over sampling is used to be sure that all quality control measures are working. After this initial phase, the sampling plan is implemented in such as way as to produce about a 10% quality control sample for the entire examination period. The Scheduling System allows for the Field Center to record which participant is involved in each QC sample. All data relating to QC is transmitted along with the normal recruitment activities to the Coordinating Center for review and monitoring.
4. Data Entry at the Coordinating Center

Like the Field Center system, the data entry system at the Coordinating Center is a server-based application. In structure, the system is flexible so that forms can be added and removed for the system without making extensive changes to the basic program. Details of this flexibility are included where appropriate in the description which follows.

The data entry program performs an extensive set of tasks which include:

a. Form Scripting Program - A set of form scripting programs, which are very much like a standard word processing program, are used to prepare forms for use by the system. Because of the flexibility of the system, changes to forms after the exam cycle has started can easily be implemented and internal revision numbers are recorded for all data collected in order to track the form actually used. The form printing utility will also reflect these changes not only on the form which is printed but also in the unique QC bar coding.

b. Data Entry of Scanned Images from the Clinic - The data entry system utilizes either the Verity TeleForm application or the dual monitor scheme. The Verity TeleForm application reads and verifies data that is recognizable, and within the specified range checks, while data that is not recognizable or outside of the range check is sent to a queue for manual verification. The dual monitor system also performs a series of edits including range checks, checks for completeness of answers and checks of skip patterns. The system maintains a status file for all entries which flags questionable answers and provides the basis for reporting data which require clarification from the clinic staff, after first being reviewed by the Coordinating Center staff.

c. Data Export - The system has the ability to create data files from the input forms in a number of formats. For most purposes these files will be either ASCII delimited text files or standard DBF files such as those produced by a number of commercial database engines. All of the formats can be read directly by the SAS analysis platform. All variable names and data types exported have been chosen by the Data Analysis and Verification group so that no translation of name or type is required after input to SAS for analysis purposes. The standard database format for all CARDIA Y20 Exam files will be SAS v604 (SSD) files. This random selection of forms will be reviewed with the data setup for quality control purposes.
d. Problem Data Resolution - The data entry systems do data checking at the time of entry. This includes range checks for continuous variables and checking for validity against a list for discrete variables. The system views data as occurring in pages (corresponding to pages of a form) and will not allow a page to be saved into the database until the page is complete. If for some reason a page cannot be completed and the issue which causes the problem cannot be immediately resolved, the data entry person has the option to save the page with a status result indicating the specific problem. The page is saved to a temporary file from which it can be edited. The system maintains variable level status information for each page and all pages in a form. When a page cannot be accepted, an entry is made in an error log which is later used to determine a satisfactory resolution to the problem. No page is added to the preliminary data set until all such errors are resolved. The data entry system will generate a random sample of forms to be either re-entered blindly by the original data entry technician and by a second independent staff person and the results compared to determine both intra and inter technician reliability. As with past CARDIA exams, all data entry must meet the accuracy requirements of the study. Regular reports of the accuracy statistics will be reported regularly.

e. Quality Control Administration - This section of the program provides the tracking for the QC forms. It maintains three lists:

i. A list of forms which have been entered a second time as part of the duplicate data entry QC effort.

ii. A list of forms as mentioned in the previous section. These are forms for which data entry found a problem which could not be resolved by the data entry person. Generally these forms will require interaction with field center staff to resolve the problem.

iii. The list of forms which have been accepted into the database as having passed the level I QC protocol. At any given time a form which has been transmitted to the Coordinating Center and been entered will be in one of these lists.

5. Data Transfer System (Between Field Centers and Coordinating Center)

The file transfer system is responsible for transferring data to and from the Field Centers and the Coordinating Center. It is also responsible for maintaining archives of the Field Center data
and maintaining logs of all data transfers. The type of data transferred falls into two categories: (1) recruitment information; and (2) scanned forms.

a. Recruitment Transfer - Only a subset of the recruitment system is transferred from the Field Center to the Coordinating Center. This subset contains the current schedule of each participant entered at the Field Center. This information is then combined and sent to all Field Centers. This information, when combined with the Field Center’s local data, allows each Field Center to view the schedule for any participant in the system. Please note that although these data are archived nightly, the archives will only be maintained for five days. Below is a flow chart representing the flow of the recruitment data:

Step 1: Each night the system will activate a routine that will make an archival copy of key files in the Recruitment System.

Step 2: A copy of the archived data is then transferred to a subdirectory at the Coordinating Center. Each Field Center will have its own subdirectory.

Step 3: All Field Center archives are decompressed and combined into one set of files. These master files are then used by the Coordinating Center to monitor recruitment efforts.

Step 4: The master set of files is sent to each Field Center replacing the previous copy. The recruitment system uses this master list to provide a complete picture of participant recruitment.

b. Scanned Forms - During the process of scanning the forms, the Field personnel will place the Scanned TIFF images into a preset subdirectory. The File transfer system takes these images, archives them, and transfers them to the Coordinating Center. Below is a flow chart detailing this process:

Step 1: Each night the system will activate a procedure which will compress the scanned images and supporting documentation into a time-stamped file. After the creation of the compressed file, the system will copy the source files into an archival subdirectory.

Step 2: The copy of the compressed files is then transferred to the Field Center’s subdirectory at the Coordinating Center.
**Step 3:** The compressed file is decompressed and the supporting documentation is processed to log receipt of the scanned images and prepare them for access by the data-entry system.

During each of the steps listed above, the system maintains detailed logs that provide clear audit trail of the disposition of all data elements processed through the system.

**H. STUDY ORGANIZATION**

The organization of the CARDIA Study and the CARDIA Study Centers is illustrated in Figures 2 and 3, respectively, at the end of this section. Below is detailed the role of the CARDIA Study entities.

1. **NHLBI**

   The CARDIA Study operates under contracts between the institutions involved and the National Heart, Lung, and Blood Institute (NHLBI). The NHLBI has assigned a Project Officer to serve as the main scientific link between NHLBI and the Program.

   **Functions and Responsibilities**

   i. Provide necessary scientific and administrative communication, liaison and direction to the Principal Investigators of the Field Centers and the Coordinating Center to assure fulfillment of the scientific objectives and administrative aspects of this program for NHLBI and NIH review --including protocol, program progress, forms and publications.

   ii. Serve as a direct link from the organization of CARDIA to the Director of the NHLBI, to channel inquiries, recommendations, and policy directives. Since this is a contract, NHLBI will be the final authority for determining program policy and how problems should be handled. A CARDIA Observational Study Monitoring Board (OSMB) composed of outside scientists will provide advice to NHLBI on the conduct of the study.

2. **OSMB**

   An external Observational Study Monitoring Board (OSMB) (referred to in CARDIA as the “Review Board”) whose membership is assigned by NHLBI meets annually to monitor and
evaluate the study's progress in all areas. The Review Board advises and makes recommendations to the Steering Committee and NHLBI concerning any scientific or administrative issues which may be of concern.

**CARDIA OSMB MEMBERS**

<table>
<thead>
<tr>
<th>Investigator</th>
<th>Affiliation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ingrid Borecki, PhD</td>
<td>Washington University, School of Medicine, Division of Biostatistics</td>
</tr>
<tr>
<td>Luther T. Clark, MD</td>
<td>SUNY Health Science Center at Brooklyn, Cardiovascular Medicine, Department of Medicine</td>
</tr>
<tr>
<td>Patricia Elmer, PhD, RD</td>
<td>Kaiser Permanente Center for Health Research</td>
</tr>
<tr>
<td>Gerardo Heiss, MD</td>
<td>University of North Carolina at Chapel Hill, Department of Epidemiology</td>
</tr>
<tr>
<td>Barbara Phillips, MD, MPH</td>
<td>University of Kentucky, Department of Medicine, Pulmonary and Critical Care</td>
</tr>
<tr>
<td>Paul Sorlie, PhD, Executive Secretary</td>
<td>National Heart, Lung and Blood Institute</td>
</tr>
<tr>
<td>David Strogatz, PhD</td>
<td>State University of New York, Albany, School of Public Health, Department of Epidemiology</td>
</tr>
<tr>
<td>Kim Suton-Tyrrell, PhD</td>
<td>University of Pittsburgh, Graduate School of Public Health, Department of Epidemiology</td>
</tr>
<tr>
<td>Alan B. Weder, MD, Chair</td>
<td>University of Michigan Medical Center, Department of Internal Medicine</td>
</tr>
</tbody>
</table>

3. **CARDIA Coordinating Center**

The CARDIA Coordinating Center is a facilitator of the study's design, monitoring, analysis, and manuscript production. The Coordinating Center takes the lead role in the generation of the study Protocol and the corresponding Manual of Operations. It also takes the organizing role for the central training and certifications. It has the responsibility for developing and implementing systems necessary for data collection, editing, management and analysis.

Functions and Responsibilities

i. Prepare the Protocol and Manual of Operations with the cooperation of the Steering Committee.

ii. Prepare and implement a system for data collection, transmittal, logging, editing, management, extraction and analysis.

iii. Work with the investigators in the development, pretesting, construction of forms and associated procedures.

   - Obtain inter- and intra-observer reliability measurements.
   - Obtain reliability for all questionnaires and procedures used in the study so that such information might be incorporated into the analyses.
iv. Manage subcontracts for central laboratories as requested by NHLBI.

v. Monitor quality control of external laboratories.

vi. Interact with the Clinic Coordinators and Principal Investigators to orchestrate the training sessions and certification for the Y20 Exam.

vii. Produce minutes of the Steering Committee meetings and conference calls.

viii. Assist in the organization and conduct of site visits to each center to ensure compliance with the provisions of the Manual of Operations.

ix. Monitor the completeness and accuracy of submitted data forms. Notify Field Centers and the relevant study units of error rates and/or deficiencies in forms submitted.

x. Design and implement a distributed data analysis system.

xi. Notify the Principal Investigator if any specific local problems arise in the Field Center's performance. Notify Project Officer if timely resolution is not possible.

xii. Issue periodic reports to NHLBI, the Steering Committee and the OSMB. Specific reports will include predefined quality control monitoring reports.

xiii. Develop new or modify existing statistical methods for data analyses.

xiv. Facilitate the preparation of study publications in cooperation with other investigators and according to the publications policies.

xv. Take the lead on specific manuscripts depending on investigator interest and in accordance with the publications policies.

xvi. Lead efforts to periodically review and revise the publications policies.

4. **CARDIA Field Centers**

The Field Centers are clinical research units supervised by the Principal Investigators and supported by individual agreement research contracts from the NHLBI. Each Field Center is responsible for following the provisions of the Protocol, Manual of Operations and Quality Control Document. The Principal Investigator of each Field Center is a voting member of the Steering Committee.

Functions and Responsibilities
i. Recruit and train staff to perform the procedures of the study effectively.

ii. Retain participants according to the CARDIA Manual of Operations (MOO) and examine/evaluate them.

iii. Establish and maintain good relations with the participants, their families, other attending physicians (if any), and the public.

iv. Collect and transmit all required data, specimens, etc. in accordance with established procedures and schedules.

v. Evaluate the progress of the study and alert the Coordinating Center and Steering Committee to major problems.

vi. Work with the Coordinating Center to maintain the quality of data collected.

5. Reading Centers

For the Y20 Exam, there are four reading centers, each functioning to review data from the Field Centers and report results to the Coordinating Center. As well, each reading center is responsible for training CARDIA Field Center staff in their area and participating on the Subcommittee which oversees their exam component.

a. The Computed Tomography Reading Center (CTRC), Jeff Carr, MD, Principal Investigator, is located at Wake Forest University. The CTRC is responsible for the review and reading of all CT scans for this exam. As well, the CTRC will provide quality scores for scans and reread scans for quality control purposes, as directed by the Imaging Committee and Coordinating Center.

b. The Diet Reading Center (DRC), Lyn Steffen, PhD, Principal Investigator, is located at the University of Minnesota. The DRC is responsible for reviewing Diet interviews for quality control purposes and converting the raw diet data into nutrient data.

c. The Ultrasound Reading Center (URC), Dan O’Leary, MD, Principal Investigator, is located at Tufts University. The URC is responsible for reviewing all ultrasound tapes and providing quality ratings for each scan and feedback to Field Center sonographers.

d. The Pulmonary Function Reading Center (PFRC), Robert Jensen, PhD, Principal Investigator, is located at the Latter Day Saints Hospital in Salt Lake City. The PFRC is
responsible for reviewing the pulmonary function data and providing quality scores for the tests.

6. Laboratories

There are four assay labs and two storage labs for the Y20 Exam. They are as follows:

a. Lipids – Northwest Lipids Research Laboratories – Santica Marcovina, PhD, ScD
b. Chemistries – Linco Research, Inc. – Ronald Gingerich, PhD
c. Central Repository – Solomon Park Laboratory – Patric Clapshaw, PhD
d. DNA Genotyping and Storage – University of Texas-Houston – Myriam Fornage, PhD
e. Albuminuria – Metabolic, environmental and behavioral research laboratory – University of Minnesota – Myron Gross, PhD
f. Inflammatory Markers – Laboratory of Clinical Biochemistry Research – University of Vermont – Russell Tracy, PhD

7. Role and Composition of Steering Committee

The Steering Committee is the governing body of the CARDIA Study. It is composed of the Steering Committee chair; the NHLBI Project Officers and Contracting Officers; Principal Investigators from the four Field Centers and the Coordinating Center; Principal Investigators of the DNA Genotyping and Storage Laboratory, the CT Reading Center, and the Diet Reading Center; the Co-Principal Investigators of each of the Field Centers and the Coordinating Center; Subcommittee Chairs; and the members of the Emerging Science Committee. The Steering Committee provides overall scientific direction for the study and serves as the focal point of the organizational structure.

The Steering Committee meets regularly to review study progress, currently twice each month on the second and fourth Tuesdays for one hour at 10:30 am Central Time. Each center is required to be represented by an investigator or proxy at each meeting of this committee. If a vote is needed, it will require a simple majority of the members. Minutes are taken by a member of the Coordinating Center.

The Steering Committee performs the following functions:

b. Review and approve major changes in the Manual of Operations and Data Collection Forms.

c. Provide advice and assistance to the Coordinating Center, Field Centers or NHLBI on operational matters.

d. Resolve problems brought to the Steering Committee by investigators, the Coordinating Center, and any other of the central units (central labs, NHLBI, etc.).

e. Monitor the performance of all participating Field Centers through site visits according to information provided by the Coordinating Center in order to insure that an adequate number of participants are retained; that the Field Centers adhere to the protocol; and that high quality data are collected.

### CARDIA STEERING COMMITTEE MEMBERS

<table>
<thead>
<tr>
<th>Investigator</th>
<th>Affiliation</th>
</tr>
</thead>
<tbody>
<tr>
<td>David Allison, PhD</td>
<td>University of Alabama at Birmingham, School of Public Health, Department of Epidemiology</td>
</tr>
<tr>
<td>Eric Boerwinkle, PhD</td>
<td>University of Texas-Houston, Health Science Center, Human Genetics Center</td>
</tr>
<tr>
<td>Jeff Carr, MD, MS</td>
<td>Wake Forest University School of Medicine</td>
</tr>
<tr>
<td>Martha Daviglus, MD</td>
<td>Northwestern University, Feinberg School of Medicine, Department of Preventive Medicine</td>
</tr>
<tr>
<td>Myriam Fornage, PhD</td>
<td>University of Texas-Houston, Health Science Center, Institute of Molecular Medicine</td>
</tr>
<tr>
<td>Myron Gross, PhD, MS</td>
<td>University of Minnesota, Molecular Epidemiology and Biomarker Research Laboratory</td>
</tr>
<tr>
<td>Stephen Hulley, MD, MPH</td>
<td>University of California, San Francisco</td>
</tr>
<tr>
<td>Carlos Iribarren, MD, MPH, PhD</td>
<td>Kaiser Permanente, Division of Research</td>
</tr>
<tr>
<td>David R. Jacobs, Jr., PhD</td>
<td>University of Minnesota, School of Public Health, Division of Epidemiology &amp; Community Health</td>
</tr>
<tr>
<td>Cheryl Jennings</td>
<td>National Heart, Lung and Blood Institute, Contracts Office</td>
</tr>
<tr>
<td>Catarina Kiefe, MD, PhD</td>
<td>University of Alabama at Birmingham, School of Medicine, Department of Medicine, Division of Preventive Medicine</td>
</tr>
<tr>
<td>Shriki Kumanyika, PhD, MPH</td>
<td>University of Pennsylvania, School of Medicine</td>
</tr>
<tr>
<td>Cora E. Lewis, MD, MSPH</td>
<td>University of Alabama at Birmingham, School of Medicine, Department of Medicine, Division of Preventive Medicine</td>
</tr>
<tr>
<td>Kiang Liu, PhD</td>
<td>Northwestern University, Feinberg School of Medicine, Department of Preventive Medicine</td>
</tr>
<tr>
<td>Vivian Lucas</td>
<td>National Heart, Lung and Blood Institute, Contracts Office</td>
</tr>
<tr>
<td>Karen Matthews, PhD</td>
<td>University of Pittsburgh, Department of Psychiatry</td>
</tr>
<tr>
<td>Cheryl Nelson, MS</td>
<td>National Heart, Lung and Blood Institute; Project Office</td>
</tr>
<tr>
<td>Sharina Person, PhD</td>
<td>University of Alabama at Birmingham, School of Medicine, Department of Medicine, Division of Preventive Medicine</td>
</tr>
<tr>
<td>Mark Pletcher, MD</td>
<td>University of California San Francisco, Department of Epidemiology and Biostatistics</td>
</tr>
<tr>
<td>Pamela Schreiner, MS, MS, PhD</td>
<td>University of Minnesota, School of Public Health, Division of Epidemiology &amp; Community Health</td>
</tr>
</tbody>
</table>
8. Role and Composition of Executive Committee

The Executive Committee is responsible for making major decisions regarding operational and policy matters of the study. In addition, they are responsible for the review and approval of Ancillary Study proposals submitted to the study. The Executive Committee meets on a monthly basis, currently the first Tuesday of each month from 11:30-12:30 Central Time. Its membership consists of the Principal Investigators from the Field and Coordinating Centers, the Chair of the Steering Committee, and the Project Officers from NHLBI.

### Executive Committee Members

<table>
<thead>
<tr>
<th>Member</th>
<th>Affiliation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Beth Lewis</td>
<td>BHAM Field Center</td>
</tr>
<tr>
<td>Kiang Liu</td>
<td>CHIC Field Center</td>
</tr>
<tr>
<td>Steve Sidney</td>
<td>OAKL Field Center</td>
</tr>
<tr>
<td>Pam Schreiner</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td>Dale Williams, Chair</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td>Gina Wei</td>
<td>NHLBI</td>
</tr>
<tr>
<td>Cheryl Nelson</td>
<td>NHLBI</td>
</tr>
<tr>
<td>Steve Hulley</td>
<td>UCSF</td>
</tr>
</tbody>
</table>

9. Role and Composition of Emerging Science Committee

In 1998, it was decided that the study could benefit from the addition of new colleagues with diverse scientific expertise in emerging areas of cardiovascular research. The strategy for the Emerging Science Committee (ESC) is that members are to be substantially and directly involved in all phases of the design and implementation of the CARDIA Study. Members participate in Steering Committee meetings and conference calls, in the study's publications generation and review process, and in subcommittee and subgroup activities.

The ESC members perform the following functions:
a. Attend at least one Steering Committee meeting per year. The strategy for meetings may be revised to better accommodate ESC input.

b. Participate in discussions of scientific issues on scheduled conference calls.

c. Submit one manuscript proposal per year and lead the preparation and submission to a peer-reviewed journal of at least one manuscript per year.

d. Serve as chief reviewer for at least two CARDIA manuscripts per year.

e. Present work-in-progress reports based on CARDIA data at CARDIA Steering Committee meetings, as appropriate.

f. Serve on CARDIA Working Groups or Subcommittees as appropriate.

**CARDIA EMERGING SCIENCE COMMITTEE MEMBERS**

<table>
<thead>
<tr>
<th>Member</th>
<th>Affiliation</th>
</tr>
</thead>
<tbody>
<tr>
<td>David Allison, PhD</td>
<td>University of Alabama at Birmingham, School of Public Health, Department of Epidemiology</td>
</tr>
<tr>
<td>Eric Boerwinkle, PhD</td>
<td>University of Texas-Houston, Health Science Center, Human Genetics Center</td>
</tr>
<tr>
<td>Shriki Kumanyika, PhD, MPH</td>
<td>University of Pennsylvania, School of Medicine</td>
</tr>
<tr>
<td>Karen Matthews, PhD</td>
<td>University of Pittsburgh, Department of Psychiatry</td>
</tr>
<tr>
<td>David Siscovick, MD, MPH</td>
<td>University of Washington</td>
</tr>
<tr>
<td>Russ Tracy, PhD</td>
<td>University of Vermont, Inflammatory Markers Laboratory</td>
</tr>
<tr>
<td>Mike Steffes, MD, PhD</td>
<td>University of Minnesota</td>
</tr>
</tbody>
</table>

10. Role and Composition of Subcommittees

For the Y20 Exam there are

a. Clinic Coordination and Retention Subcommittee

The Clinic Coordination and Retention Subcommittee’s purpose is to:

- Develop a communications support network for the Clinic Coordinators, the Coordinating Center and the Project office.

- Establish a mechanism for providing input and feedback to the Steering Committee.

- Convene annual meetings to discuss clinic operational issues, and summarize these for the Steering Committee.
• Provide encouragement to Clinic Coordinators to participate in other study activities.

• Plan and coordinate a retention program which will maximize the number of baseline participants to be re-examined during the Y20 Exam.

• Collaborate with the Coordinating Center in designing and updating a monitoring system to evaluate and track the retention efforts.

The Clinic Coordination and Retention Subcommittee will meet periodically to review the effectiveness of the recruitment/retention plan and create new strategies as needed.

b. Design and Analysis Subcommittee

The Design and Analysis Subcommittee’s purpose is to:

• Develop analytic strategies to deal with longitudinal data.

• Identify appropriate statistical models from specific epidemiologic problems.

• Plan analyses and publications which encompass the hypothesized epidemiologic models.

c. Endpoints and Adjudication Subcommittee

The Endpoints and Adjudication Subcommittee’s purpose is to:

• Develop strategies for obtaining clinical information on all deaths, and on hospitalizations deemed relevant to CARDIA.

• Enhance the completeness with which this information is obtained.

• Adjudicate the causes of these disease events.

d. Publications and Presentations Committee

The Publications and Presentations Committee is responsible for overseeing all CARDIA publications and presentations activities, with final adjudication of decisions by the Steering Committee, and stimulating and enhancing the timely production of an optimal set of publications and presentations. Further, they are to plan and implement a policy to oversee all study publications and presentations.

The Publications and Presentations Committee’s purpose is to:
• Provide a system for tacking progress on each proposed manuscript through its completion.

• Provide scientific leadership and long-range planning for CARDIA publications activities.

• Determine areas and topics of high scientific priority for CARDIA manuscripts and periodically re-examine these priorities.

• Periodically examine CARDIA publications (completed and in progress) and determine whether scientific priorities are being met. Propose strategies for progress.

• Recruit new investigators, including junior investigators, to produce manuscripts.

• Identify areas of potential collaboration with other studies.

e. Quality Control Subcommittee

The Quality Control Subcommittee is responsible for designing and overseeing quality control approaches for all aspects of CARDIA not specifically addressed by another subcommittee, and will recommend strategies to improve the quality of CARDIA data.

The purpose of the Quality Control Committee is to:

• Design appropriate measures and techniques that assess data quality.

• Design appropriate measures and techniques that assess quality of the laboratories and reading centers.

• Review the quality control reports on the data.

• Make recommendations to improve the quality of the data, as needed.

• Recommend remedial protocols to improve the performance of a laboratory or reading center.

• Prepare a document summarizing and assessing the quality of the collected data.

f. Imaging Subcommittee
The Imaging Subcommittee is responsible for helping to plan and address issues concerning the measurement of coronary artery calcium and carotid intimal media thickness, quality control of these data, and analysis and interpretation of results.

The Imaging Subcommittee’s purpose is to:

- Develop and oversee the implementation of the protocol, quality control procedures, and analysis of carotid ultrasound, coronary calcium, and other relevant imaging data.

- Advise the Steering Committee on issues related to CAC and Carotid imaging.

- Communicate with the two CARDIA imaging reading centers.

g. Laboratory Subcommittee

The Laboratory Subcommittee is responsible for planning and overseeing laboratory measurements on blood and urine specimens from Y20 Exam, oversee use of stored specimens from prior exams, and guide analyses and interpretation of results.

The Laboratory Subcommittee’s purpose is to:

- Plan and oversee the implementation of the laboratory procedures for the Y20 Exam.

- Review and recommend laboratory measures (for blood and urine) to be included in the Y20 Exam.

- Review results of internal and external quality results during the Y20 Exam.

- Meet periodically to discuss measures to be conducted in case-control design following the Y20 Exam.

h. Psychosocial Subcommittee

The Psychosocial Subcommittee is responsible for designing and overseeing the psychosocial measures in CARDIA, and guide analysis and interpretation of results.

The Psychosocial Subcommittee’s purpose is to:

- Advise the Steering Committee on selection of psychosocial aspects of CARDIA, including selection of constructs for inclusion in the protocol; measurement; interpretation; implementation in the protocol.
• Provide suggestions for psychosocial paper proposals.

• Work as a liaison with ancillary projects that involve psychosocial hypotheses.

i. Genetics Subcommittee

The Genetics Subcommittee is responsible for designing and coordinating genetic activities in CARDIA, including research questions, participant consent, analyses, substudies, publications and presentations.

The Genetics Subcommittee’s purpose is to:

• Track ongoing genetic studies using CARDIA DNA.

• Recommend to the Steering Committee approximately 50 polymorphisms on identified genes for genotyping.

• Review proposed substudies and ancillary studies, from investigators both within and external to the CARDIA Study.

• Establish an ancillary study agreement for investigators using CARDIA DNA that will address a formal policy on data distribution and submitting progress reports, what to do with a non-cooperative ancillary study investigators, and a formal policy on either destroying or returning excess DNA to its source.

i. Diet Subcommittee

The Diet Subcommittee is responsible for designing and overseeing the approaches for collecting dietary data in CARDIA and to analyze and publish the findings.

The Diet Subcommittee’s purpose is to:

• Advise the Steering Committee on selection of instruments to collect information related to diet for inclusion in the protocol as well as aspects of measurement, interpretation and implementation in the protocol.

• Provide suggestions for diet paper proposals.

• Work as a liaison with ancillary projects that involve diet hypotheses.

j. Pulmonary Subcommittee

The Pulmonary Subcommittee is responsible for monitoring the collection and quality
of the pulmonary function data, including review of data and protocols.

The Pulmonary Subcommittee’s purpose is to:

- Review the Manual of Operations written by the Pulmonary Function Reading Center to ensure it adheres to the CARDIA plan.
- Provide suggestions for pulmonary related paper proposals.
- Review Pulmonary data during the exam.
- Advise the Steering Committee on issues related to pulmonary testing or data.
## Subcommittee Memberships

<table>
<thead>
<tr>
<th>Committee</th>
<th>Member</th>
<th>Affiliation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clinic Coordination and Retention</td>
<td>James Shikany</td>
<td>BHAM Field Center</td>
</tr>
<tr>
<td></td>
<td>Phil Johnson</td>
<td>BHAM Field Center</td>
</tr>
<tr>
<td></td>
<td>Christie Oden</td>
<td>BHAM Field Center</td>
</tr>
<tr>
<td></td>
<td>Julia Wilkoff</td>
<td>CHIC Field Center</td>
</tr>
<tr>
<td></td>
<td>Valerie Bruce</td>
<td>CHIC Field Center</td>
</tr>
<tr>
<td></td>
<td>Claudia Chambers</td>
<td>OAKL Field Center</td>
</tr>
<tr>
<td></td>
<td>Bev Peters</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>Valerie Green</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>Pam Schreiner, Chair</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>Mary Zubrzycki</td>
<td>NHLBI</td>
</tr>
<tr>
<td></td>
<td>Joanie Tool</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Kathy Harrington</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Cheryl Nelson</td>
<td></td>
</tr>
<tr>
<td>Design and Analysis</td>
<td>Kiang Liu, Chair</td>
<td>CHIC Field Center</td>
</tr>
<tr>
<td></td>
<td>David Jacobs</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>Dale Williams</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Sharina Person</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Cheryl Nelson</td>
<td>NHLBI</td>
</tr>
<tr>
<td></td>
<td>David Siscovick</td>
<td>UWash</td>
</tr>
<tr>
<td></td>
<td>Myriam Fornage</td>
<td>Genotyping Lab - UTH</td>
</tr>
<tr>
<td>Diet</td>
<td>Linda Van Horn</td>
<td>CHIC Field Center</td>
</tr>
<tr>
<td></td>
<td>Martha Daviglus</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>David Jacobs, Chair</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>Lyn Steffen</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Dale Williams</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Kathy Harrington</td>
<td>NHLBI</td>
</tr>
<tr>
<td></td>
<td>Cay Loria</td>
<td>UPenn</td>
</tr>
<tr>
<td></td>
<td>Shriki Kumanyika</td>
<td></td>
</tr>
<tr>
<td>Endpoints &amp; Surveillance &amp; Adjudication</td>
<td>Beth Lewis, Chair</td>
<td>BHAM Field Center</td>
</tr>
<tr>
<td>------------------------------------------</td>
<td>-------------------</td>
<td>--------------------</td>
</tr>
<tr>
<td></td>
<td>James Shikany</td>
<td>BHAM Field Center</td>
</tr>
<tr>
<td></td>
<td>Phil Johnson</td>
<td>BHAM Field Center</td>
</tr>
<tr>
<td></td>
<td>Phillip Greenland</td>
<td>CHIC Field Center</td>
</tr>
<tr>
<td></td>
<td>Steve Sidney</td>
<td>OAKL Field Center</td>
</tr>
<tr>
<td></td>
<td>Carlos Iribarren</td>
<td>OAKL Field Center</td>
</tr>
<tr>
<td></td>
<td>Richard Crow</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>Catarina Kiefe</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Jo Crawford</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Alex Arynchyn</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Gina Wei</td>
<td>NHLBI</td>
</tr>
<tr>
<td></td>
<td>Mark Pletcher</td>
<td>UCSF</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Genetics</td>
<td>Martha Daviglus</td>
<td>CHIC Field Center</td>
</tr>
<tr>
<td></td>
<td>Pam Schreiner</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>Myron Gross</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>Dale Williams</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Kathy Harrington</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Ebony Bookman</td>
<td>NHLBI</td>
</tr>
<tr>
<td></td>
<td>Myriam Fornage, Chair</td>
<td>Genotyping Lab - UTH</td>
</tr>
<tr>
<td></td>
<td>David Siscovick</td>
<td>UWash</td>
</tr>
<tr>
<td></td>
<td>David Allison</td>
<td>UAB</td>
</tr>
<tr>
<td></td>
<td>Eric Boerwinkle</td>
<td>Genotyping Lab - UTH</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Laboratory</td>
<td>Donald Lloyd-Jones</td>
<td>CHIC Field Center</td>
</tr>
<tr>
<td></td>
<td>Pam Schreiner, Chair</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>Myron Gross</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>Dale Williams</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Jennifer Wammack</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Gina Wei</td>
<td>NHLBI</td>
</tr>
<tr>
<td></td>
<td>David Siscovick</td>
<td>UWash</td>
</tr>
<tr>
<td></td>
<td>Mike Steffes</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>Russ Tracy</td>
<td>IM Lab - UVT</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pulmonary</td>
<td>Lewis Smith</td>
<td>CHIC Field Center</td>
</tr>
<tr>
<td></td>
<td>Carlos Iribarren</td>
<td>OAKL Field Center</td>
</tr>
<tr>
<td></td>
<td>David Jacobs</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>Dale Williams</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Jo Crawford</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Alex Arynchyn</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Gina Wei</td>
<td>NHLBI</td>
</tr>
<tr>
<td></td>
<td>Robert Jensen</td>
<td>US Reading Center – Utah</td>
</tr>
<tr>
<td></td>
<td>Robert Crapo</td>
<td>US Reading Center – Utah</td>
</tr>
<tr>
<td>Psychosocial</td>
<td>Diane Tucker</td>
<td>BHAM Field Center</td>
</tr>
<tr>
<td>--------------------------------</td>
<td>-------------------</td>
<td>-------------------------</td>
</tr>
<tr>
<td></td>
<td>Pam Schreiner</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>Catarina Kiefe</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Kathy Harrington</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Cheryl Nelson</td>
<td>NIH</td>
</tr>
<tr>
<td></td>
<td>Karen Matthews, Chair</td>
<td>UPitt</td>
</tr>
<tr>
<td></td>
<td>Anna Diez-Roux</td>
<td>UPitt</td>
</tr>
<tr>
<td></td>
<td>David Williams</td>
<td>UMich</td>
</tr>
<tr>
<td></td>
<td>Mary Whooley</td>
<td>UCSF</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Publications &amp; Presentations</th>
<th>Beth Lewis</th>
<th>BHAM Field Center</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Martha Daviglus</td>
<td>CHIC Field Center</td>
</tr>
<tr>
<td></td>
<td>Steve Sidney</td>
<td>OAKL Field Center</td>
</tr>
<tr>
<td></td>
<td>David Jacobs</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>Catarina Kiefe, Chair</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Cay Loria</td>
<td>NHLBI</td>
</tr>
<tr>
<td></td>
<td>Gina Wei</td>
<td>NHLBI</td>
</tr>
<tr>
<td></td>
<td>Steve Hulley</td>
<td>UCSF</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Quality Control</th>
<th>Phil Johnson</th>
<th>BHAM Field Center</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Kiang Liu, Chair</td>
<td>CHIC Field Center</td>
</tr>
<tr>
<td></td>
<td>David Jacobs</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>Dale Williams, Co-Chair</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Sharina Person</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Alex Arynchyn</td>
<td>COORDINATING CENTER</td>
</tr>
<tr>
<td></td>
<td>Gina Wei</td>
<td>NHLBI</td>
</tr>
<tr>
<td></td>
<td>David McPherson</td>
<td>CHIC Field Center</td>
</tr>
<tr>
<td></td>
<td>Mike Steffes</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>Lyn Steffen</td>
<td>MINN Field Center</td>
</tr>
<tr>
<td></td>
<td>Myriam Fornage</td>
<td>Genotyping Lab - UTH</td>
</tr>
<tr>
<td></td>
<td>Jeff Carr</td>
<td>CT Reading Center</td>
</tr>
</tbody>
</table>
FIGURE 2. CARDIA ORGANIZATION CHART

NHLBI Director: Nabel

CARDIA Review Board: Weder, Chair

NHLBI Contract Office: Jennings; Lucas

NHLBI Project Office: Wei; Nelson

Executive Committee: Williams, Chair

Steering Committee: Hulley, Chair

Emerging Science Committee

Standing Subcommittees

- Quality Control (Liu, Chair)
- Publications and Presentations (Kiefe, Chair)
- Design & Analysis (Liu, Chair)
- Clinic Coordination and Retention (Schreiner, Chair)
- Endpoints and Adjudication (Lewis, Chair)
- Pulmonary Function (Smith, Chair)
- Imaging (Sidney, Chair)
- Laboratory (Schreiner, Chair)
- Psychosocial (Matthews, Chair)
- Genetics (Fornage, Chair)
I. **Timetable**

The Study is grouped into three phases:

**Phase I**

Planning, pilot testing, cohort follow-up: April 1, 2004 - May 31, 2005 (14 months)

1. Continued periodic contacts of cohort members
2. Planning for the Y20 Exam
3. Pilot testing for the Y20 Exam
4. Ongoing analysis and publication of data
5. Morbidity and mortality follow-up activities
6. Preparation of public use data set for data prior to Y20 Exam
7. Monitoring participant contact rates

**Phase II**

Cohort Examination: June 1, 2005 – May 30, 2006 (12 months)

8. Monitoring participant recruitment
9. Conduct of the Y20 Exam
10. Monitoring periodic contacts of cohort members
11. Morbidity and mortality follow-up activities
12. Ongoing analysis and publication of data
13. Preparation of public use data set for Y20 Exam

**Phase III**

Close-out and Final Data Analysis: June 1, 2006 - September 30, 2008 (28 months)

14. Close-out of clinic operations
15. Final data analysis and publication of data
16. Morbidity and mortality follow-up activities
17. Preparation of Y20 Exam public use data set
J. REFERENCES


2. The CARDIA Study Steering Committee, *Coronary Artery Risk Development in (Young) Adults (CARDIA): Year 0 Exam Protocol*. February 1985: The CARDIA Study Coordinating Center, University of Alabama at Birmingham.


8. The CARDIA Coordinating Center, *Coronary Artery Risk Development in (Young) Adults (CARDIA): Year 15 Data Systems Documentation*. April 2000: The CARDIA Study Coordinating Center, University of Alabama at Birmingham.


37. Loria, C.M., et al., *Sex and race differences in prevalence and predictors of early coronary calcification: The CARDIA Study (abstract) Presented at the American Heart Association 42nd Annual Conference on Cardiovascular Disease Epidemiology and Prevention, Honolulu, HI. 2002.*


45. Folsom, A., *personal communication*.


58. National Center for Health Statistics, **Skinfolds, body girths, biacromial diameter and selected anthropometric indices of adults.** Data from the National Health Survey, Series 11, Number 35. DHEW Publication No. (HRA). p. 74-1281, 1970.


60. Khoury, P., et al., **Weight change since age 18 years in 30- to 55-year-old whites and blacks. Associations with lipid values, lipoprotein levels, and blood pressure.** Jama, 1983. 250(23): p. 3179-87.


186. Davidson, et al., *Do depression symptoms predict early hypertension incidence in Young Adults in the CARDIA Study?* Archives of Internal Medicine, 2000. 160: p. 1495-1500.


