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ORIGINAL RESEARCH

Combining Biomarkers and Imaging for Short-Term Assessment of Cardiovascular Disease Risk in Apparently Healthy Adults

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BACKGROUND: Current strategies for cardiovascular disease (CVD) risk assessment focus on 10-year or longer timeframes. Shorter-term CVD risk is also clinically relevant, particularly for high-risk occupations, but is under-investigated.

METHODS AND RESULTS: We pooled data from participants in the ARIC (Atherosclerosis Risk in Communities study), MESA (Multi-Ethnic Study of Atherosclerosis), and DHS (Dallas Heart Study), free from CVD at baseline (N=16 581). Measurements included N-terminal pro-B-type natriuretic peptide (>100 pg/mL prospectively defined as abnormal); high-sensitivity cardiac troponin T (abnormal >5 ng/L); high-sensitivity C-reactive protein (abnormal >3 mg/L); left ventricular hypertrophy by ECG (abnormal if present); carotid intima-media thickness, and plaque (abnormal >75th percentile for age and sex or presence of plaque); and coronary artery calcium (abnormal >10 Agatston U). Each abnormal test result except left ventricular hypertrophy by ECG was independently associated with increased 3-year risk of global CVD (myocardial infarction, stroke, coronary revascularization, incident heart failure, or atrial fibrillation), even after adjustment for traditional CVD risk factors and the other test results. When a simple integer score counting the number of abnormal tests was used, 3-year multivariable-adjusted global CVD risk was increased among participants with integer scores of 1, 2, 3, and 4, by ≈2-, 3-, 4.5- and 8-fold, respectively, when compared with those with a score of 0. Qualitatively similar results were obtained for atherosclerotic CVD (fatal or non-fatal myocardial infarction or stroke).

CONCLUSIONS: A strategy incorporating multiple biomarkers and atherosclerosis imaging improved assessment of 3-year global and atherosclerotic CVD risk compared with a standard approach using traditional risk factors.

Key Words: carotid intima-media thickness ■ coronary artery calcium ■ high-sensitivity cardiac troponin T ■ high-sensitivity C-reactive protein ■ N-terminal pro B-type natriuretic peptide ■ plaque

See Editorial by Osei and Blaha

Current clinical strategies for the primary prevention of cardiovascular disease (CVD) rely on estimation of the absolute risk of incident atherosclerotic CVD (ASCVD) events in individual patients. The American Heart Association and American College of Cardiology CVD risk assessment guidelines focus on 10-year and longer-term risk of hard ASCVD events, defined as non-fatal myocardial infarction, non-fatal stroke, or death from coronary heart disease or stroke.[1] Use of pooled cohort equations (PCE) to estimate 10-year ASCVD risk is recommended to guide therapeutic decisions, including initiation of pharmacotherapy for hypercholesterolemia and hypertension,[2,3] while longer-term risk assessment is suggested as

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CLINICAL PERSPECTIVE

What Is New?

- A novel strategy combining plasma biomarkers (N-terminal pro-B-type natriuretic peptide; high-sensitivity cardiac troponin T; high-sensitivity C-reactive protein), and imaging (carotid intima-media thickness; plaque; coronary artery calcium) improves assessment of short-term (3-year) global and atherosclerotic cardiovascular disease risk among adults without known cardiovascular disease.

What Are the Clinical Implications?

- This strategy can be used to better assess cardiovascular risk in apparently healthy individuals before critical missions such as prolonged space flight and may also have broader implications for future precision prevention efforts.

Nonstandard Abbreviations and Acronyms

ARIC	Atherosclerosis Risk in Communities study
ASCVD	atherosclerotic cardiovascular disease
CAC	coronary artery calcium
DHS	Dallas Heart Study
ECG-LVH	left ventricular hypertrophy by ECG
hs-CRP	high-sensitivity C-reactive protein
hs-cTnT	high-sensitivity cardiac troponin T
IMT	intima-media thickness
MESA	Multi-Ethnic Study of Atherosclerosis
NT-proBNP	N-terminal pro B-type natriuretic peptide
PCE	pooled cohort equations

reasonable to motivate lifestyle modifications, particularly in younger individuals and in those with low 10-year risk.[1]

The focus on ASCVD risk does not consider risk for incident heart failure or atrial fibrillation, which represent a significant proportion of overall CVD events, particularly in older populations.[4,5] Moreover, the 10-year risk window may not be appropriate for situations that require more comprehensive estimation of CVD risk over a shorter timeframe. For instance, the National Aeronautics and Space Administration Authorization Act of 2017 set the goal for a crewed mission to Mars in the 2030s,[6] estimated to involve ≈1 year of Earth-based intensive astronaut training, followed by ≈2 years of actual space mission. Among the many challenges involved in evaluating and preparing astronauts for such missions, comprehensive assessment of 3-year CVD risk will be essential for crew selection. Accurate global CVD risk assessment is necessary both to minimize the risk of events that can have life- and mission-threatening consequences, and also to avoid exclusion of otherwise qualified individuals with borderline elevations in longer-term ASCVD risk. The present study was supported by National Aeronautics and Space Administration to develop a short-term (3-year) global CVD risk assessment strategy for this purpose.

We hypothesized that a composite model, incorporating multiple CVD biomarkers associated with distinct pathophysiological processes, would predict 3-year global CVD and ASCVD risk among adults without known CVD at baseline, even after adjustment for traditional risk factors. The biomarkers prospectively selected for this study were NT-proBNP (N-terminal pro B-type natriuretic peptide),[7] hs-cTnT (high-sensitivity cardiac troponin T),[8,9] hs-CRP (high-sensitivity C-reactive protein),[10] a composite of carotid intima-media thickness and plaque measured by ultrasound[11] or coronary artery calcium (CAC) measured by computed tomography,[12] and 12-lead ECG assessment of left ventricular hypertrophy by ECG (ECG-LVH).[13] A similar approach was previously shown to improve 10-year global CVD and ASCVD risk prediction in the Dallas Heart Study and MESA cohorts,[14] but its utility for shorter-term risk prediction is unknown.

METHODS

The anonymized data that support the findings of this study are available from the corresponding author upon reasonable request.

Study Population

The study included participants enrolled in 3 independent, multi-ethnic population-based cohorts, the ARIC (Atherosclerosis Risk in Communities study), DHS (Dallas Heart Study), and MESA (Multi-Ethnic Study of Atherosclerosis). Detailed methods for these studies have been published elsewhere.[15–17] ARIC enrolled 15 792 participants aged 45 to 64 years, between 1987 and 1989, with data from the second examination (collected between 1990 and 1992 from 14 348 participants) used as the baseline assessment in the present study. DHS enrolled 6101 participants aged 30 to 65 years between 2000 and 2002, of whom 3072 completed all 3 phase 1 visits (in-home survey, laboratory testing, imaging tests, and ECG), with resulting data used in the present study.

MESA enrolled 6814 participants 45 to 84 years of age, with no history of CVD, between 2000 and 2002, with data from the first examination used in the present study. From each cohort, we excluded participants with CVD at baseline, missing data on CVD biomarkers or covariates (age, sex, race, total cholesterol, high-density lipoprotein cholesterol, systolic blood pressure, antihypertensive medication use, current smoking, diabetes mellitus status, body mass index, and estimated glomerular filtration rate [eGFR]), and incomplete follow-up (<3 years). The final study population consisted of 16 581 participants from the 3 combined cohorts (7723 from ARIC; 2237 from DHS; 6621 from MESA). Each study had Institutional Review Board approval at participating institutions, and all participants provided written informed consent.

Data Collection and Definitions

Detailed descriptions of data collection for each cohort have been previously published,[15–18] and variable definitions are in accordance with current clinical guidelines. We defined hypertension as systolic blood pressure ≥130 mm Hg or diastolic blood pressure ≥80 mm Hg, and stage 2 hypertension as systolic blood pressure ≥140 mm Hg or diastolic blood pressure ≥80 mm Hg, based on guidelines recently updated by the American Heart Association and the American College of Cardiology.[3]

Measurement of Biomarkers

ECG determination of cardiac hypertrophy (ECG-LVH) was based on 12-lead ECGs using the Sokolow-Lyon voltage criteria, and categorized as present or absent.[19] Plasma biomarkers were measured from aliquots previously stored at −70°C or −80°C. hs-CRP was measured using the Roche Modular P chemistry analyzer (Roche Diagnostics, Indianapolis, IN) in ARIC,[20] the Roche/Hitachi 912 System in DHS,[21] and the Dade Behring BNII system (Siemens Healthcare GmbH, Erlangen, Germany) in MESA.[22] NT-proBNP and hs-cTnT were measured using the Roche Elecsys 2010 analyzer in ARIC and DHS,[23–25] and the Roche Cobas e601 in MESA.[14,26] Thresholds defining abnormal plasma biomarkers were prospectively selected at hs-CRP ≥3 mg/L, NT-proBNP ≥100 ng/L and hs-cTnT ≥5 ng/L.[14] In ARIC, trained readers used ultrasound to measure carotid intima-media thickness (IMT) at 3 locations bilaterally, with up to 11 measurements per location, and to assess plaque presence or absence, as previously described.[27] Abnormal carotid ultrasound was defined as either the presence of carotid plaque or IMT ≥75th percentile for age and sex. CAC measurements were performed in duplicate using either electron beam or multidetector CT in MESA,[28] and electron beam computed tomography in DHS,[29] with the mean of the 2 scans expressed in Agatston units used for analyses. The threshold for positive CAC status was defined as a mean Agatston score >10 U.[29]

Cohort Follow-Up and Study End Points

Study outcomes included global CVD (composite of cardiovascular death, myocardial infarction, stroke, coronary revascularization, incident heart failure, or atrial fibrillation) and ASCVD (fatal or non-fatal myocardial infarction or stroke) during the first 3 years of follow-up. The methodology for assessing incident CVD events during follow-up has been previously described for all 3 cohorts.[15–17] Briefly, continuous retrospective surveillance of deaths and hospitalizations was performed for ARIC participants, with relevant medical records from all hospitals admitting residents from the 4 ARIC communities abstracted for CVD outcomes. Participants in DHS completed an annual health survey including questions about CVD diagnoses and hospitalizations. For those DHS participants who consented to hospitalization tracking (>90%), admissions at hospitals participating in the Dallas–Fort Worth Hospital Council Data Initiative Database (70 out of 72 total hospitals in the region) were also recorded and matched with survey data. Fatal events were assessed using the National Death Index and classified as CVD deaths if they included *International Classification of Diseases*, *Tenth Revision* (*ICD-10)* codes I00–I99. Participants in MESA were contacted by a trained telephone interviewer at 9- to 12-month intervals, with questions about CVD diagnoses, hospitalizations, and deaths. Medical records and death certificates were subsequently obtained for a vast majority (98%) of suspected CVD events. For all 3 cohorts, source documents were collected and reviewed for all suspected CVD events and were independently adjudicated by blinded end-point committees.

Statistical Analysis

Participant characteristics were analyzed in ARIC, DHS, and MESA separately, as well as in the pooled cohort. Because of the relatively low number of events over 3 years of follow-up, analyses of associations of test results with outcomes were performed only in the pooled cohort. Test results were analyzed as dichotomous categorical variables, with either present or absent ECG-LVH and plaque, and pre-specified thresholds for abnormal hs-CRP, NT-proBNP, hs-cTnT, IMT, and CAC as defined previously. We also modeled hs-CRP, NT-proBNP, and hs-cTnT as log-transformed continuous variables in separate analyses. The associations of biomarkers with global CVD and atherosclerotic CVD were assessed using Cox

proportional hazards models, both unadjusted and adjusted for pre-specified covariates. Cohort-specific baseline hazard functions were used in all Cox models with robust standard errors to account for the clustering of patients by study. Covariates included traditional cardiovascular risk factors (age, sex, black race, total cholesterol, high-density lipoprotein cholesterol, systolic blood pressure, lipid-lowering and antihypertensive medication use, current smoking, diabetes mellitus status, and body mass index), eGFR, and the other test results. Schoenfeld residuals were used to test Cox proportional hazards model assumptions. Improvement in discrimination after addition of biomarkers to the base model of traditional cardiovascular risk factors was determined from improvement in Harrell c-statistic using bootstrap resampling. Imaging and plasma biomarkers that were independently associated with global CVD or atherosclerotic CVD after full adjustment (for the base model, eGFR, and the other biomarkers) were used to create an integer score, counting the number of abnormal test results. Associations of the integer scores with global CVD and ASCVD were assessed using Kaplan–Meier methods and multivariable Cox proportional hazards models. Subgroup analyses were performed based on age, sex, race/ethnicity, and estimated ASCVD risk using the PCE. All statistical analyses were performed using SAS version 9.4 (SAS Institute Inc.).

RESULTS

Baseline characteristics for each cohort and for the 3 cohorts combined are presented in Table 1. The mean age of the pooled cohort was 57.3±10 years, 55.7% were women, and 25.7% were black. Approximately 30% were on antihypertensive medications, and 9% on statins. Table 1 also shows baseline levels and prevalence of abnormal test results for each biomarker included in the study. During the 3-year follow-up period, incident global CVD occurred in 3.3% of the study population (553 events), and incident ASCVD occurred in 1.6% (260 events).

Association of Biomarkers With Clinical Outcomes

Abnormal hs-CRP, NT-proBNP, hs-cTnT, and imaging markers of subclinical atherosclerosis were each associated with the primary outcome of global CVD events after adjustment for traditional CVD risk factors and body mass index (Table 2). These associations were only minimally attenuated and remained statistically significant after further adjustment for eGFR and after adjusting for the other biomarkers (Table 2). Significant associations with global CVD were also observed when hs-CRP, NT-proBNP, and hs-cTnT were analyzed as log-transformed continuous variables (Table 2). Each test was associated with ASCVD in unadjusted analyses, but only NT-proBNP and the composite subclinical atherosclerosis variable were independently associated with ASCVD events (Table 3). We also considered ECG-LVH, but it was not associated with global CVD (hazard ratio [HR], 1.11; 95% CI, 0.87–1.43) or ASCVD (HR, 0.98; 95% CI, 0.68, 1.42) after adjustment for traditional CVD risk factors. For this reason, ECG-LVH was not included as a variable in further analyses.

Three-Year CVD Risk Prediction Improvement With Biomarkers

For the primary outcome of global CVD events, addition of all 4 biomarkers as categorical variables to the base model of traditional CVD risk factors resulted in a significant increase in the c-statistic (Table 4). Improvements in c-statistic were comparatively smaller for atherosclerotic CVD events, but remained statistically significant (Table 4). When the biomarkers were analyzed as continuous variables, improvements in c-statistic were slightly greater than when entered as categorical variables.

We created a simple integer score to describe how many of the 4 biomarkers (hs-CRP, NT-proBNP, hs-cTnT, and the composite imaging marker) were abnormal in each study participant, with values ranging from 0 to 4. Figure 1 shows Kaplan–Meier estimates of the rates of global CVD and ASCVD outcomes in the combined cohorts, stratified by the number of abnormal biomarkers. When compared with those with an integer score of 0, there was a significant increase in 3-year global CVD risk for participants with integer scores of 1, 2, 3, and 4, by ≈2-, 3-, 4.5- and 8-fold, respectively, after adjustment for traditional CVD risk factors and eGFR (Table 5). All integer scores ≥1 were also associated with significantly increased risk of ASCVD, albeit of lesser magnitude than for global CVD (Table 5). As shown in Figure 2, qualitatively similar results were obtained in analyses stratified by type of subclinical atherosclerosis imaging test used (IMT or plaque in ARIC; CAC in DHS and MESA), sex, race, age group, and estimated CVD risk category using the PCE equations. We also conducted sensitivity analyses in the subset of participants that had either CAC=0 or normal carotid IMT at baseline (N=9945), and found that integer scores of 2 or 3 (now based solely on plasma biomarkers, with a score scale of 0 to 3) were still associated with a significantly higher 3-year global CVD risk after adjustment for traditional CVD risk factors (HR, 1.36; 95% CI, 0.84–2.18 for integer score 1; HR, 2.63; 95% CI, 1.58–4.36 for integer score 2; HR, 3.67; 95% CI, 1.84–7.33 for integer score

Table 1. Baseline Characteristics

	ARIC (n=7723)	DHS (n=2237)	MESA (n=6621)	Combined Cohorts (n=16 581)	
	Mean±SD or Median (IQR) or Percent	Mean±SD or Median (IQR) or Percent	Mean±SD or Median (IQR) or Percent	Mean±SD or Median (IQR) or Percent	N
Age, y	56.8±5.7	44.7±9.2	62.2±10.2	57.3±10	16 581
Women, %	58.2	55.8	52.7	55.7	16 581
Race/ethnicity					
Black, %	8	47.3	27.4	25.7	16 581
Hispanic, %	···	16.5	22.2	11.1	16 581
White, %	82.0	34.1	38.4	58.1	16 581
Other/unknown %	···	2.2	12.0	5.1	16 581
Body mass index, kg/m^2	27.5±4.9	29.0±6.1	28.3±5.5	28±5.3	16 566
Diabetes mellitus, %	7.9	9.3	12.6	9.95	16 581
Current smoker, %	19.5	27	13	17.9	16 581
Systolic blood pressure, mm Hg	120.1±17.8	123.6±17.9	127.1±21.4	123.3±19.6	16 580
Diastolic blood pressure, mm Hg	71.7±9.9	77.8±9.8	72.2±10.3	72.7±10.3	16 580
Hypertension, %	32.4	42.2	45.6	39.0	16 581
Stage 2 hypertension, %	14.3	17.5	25.9	19.4	16 581
Antihypertensive medications, %	27.4	18.8	37.2	30.2	16 581
Total cholesterol, mg/dL	207 (184–232)	180 (157–205)	192 (170–215)	197 (174–223)	16 581
LDL cholesterol, mg/dL	130 (109–155)	106 (84–129)	116 (96–136)	121 (100–145)	16 408
HDL cholesterol, mg/dL	47 (39–60)	48 (40–58)	48 (40–59)	48 (40–59)	16 534
Triglycerides, mg/dL	112 (81–158)	96 (68–146)	111 (78–161)	110 (78–158)	16 580
Hypercholesterolemia, %	19.0	7.0	9.2	13.5	16 581
Statin medications, %	5.54	5.59	14.88	9.3	16 581
Creatinine, mg/dL	0.74±0.38	0.88±0.34	0.96±0.29	0.85±0.36	16 534
10-y ASCVD risk (pooled cohort equations), %	7.6 (14.6–26.5)	2.1 (0.7–5.5)	9.4 (3.8–19.8)	10.3 (4.2–21.5)	16 534
ECG-LVH, %	8.24	8.67	9.23	8.69	16 581
hs-CRP, mg/L	2.03 (0.98–4.2)	2.7 (1.1–6.2)	1.91 (0.84–4.24)	2.05 (0.94–4.43)	16 581
hs-CRP ≥3 mg/L, %	35.4	46	36.1	37.1	16 581
NT-proBNP, ng/L	51.3 (28.7–89.7)	27.7 (12.7–56.9)	53 (24–107.7)	48.1 (24–91.5)	16 581
NT-proBNP ≥100 ng/L, %	20.7	11	27.4	22.1	16 581
hs-cTnT, ng/L	1.5 (1.5–6)	1.5 (1.5–1.5)	4.4 (3–7.5)	3.1 (1.5–6)	16 581
hs-cTnT ≥5 ng/L, %	33.5	14	43.7	34.9	16 581
CAC, Agatston U	···	0.5 (0–4.5)	0 (0–86.5)	0.5 (0–52)	8858
CAC >10, %	···	19.2	42.4	36.6	8858
Plaque, %	33.1	···	···	33.1	7469
IMT	0.67 (0.6–0.77)	···	···	0.67 (0.6–0.77)	7603
IMT >75th percentile, %	23.6	···	···	23.6	7603
Plaque and/or IMT >75th percentile and/or CAC >10, %	43.8	19.2	42.4	39.95	16 581

ARIC indicates Atherosclerosis Risk in Communities study; ASCVD, atherosclerotic CVD; CAC, coronary artery calcium score; ECG-LVH, left ventricular hypertrophy by ECG; HDL, high-density lipoprotein; hs-CRP, high sensitivity C-reactive protein; hs-cTnT, high-sensitivity cardiac troponin T; IMT, intima-media thickness; IQR, interquartile range; LDL, low-density lipoprotein; and NT-proBNP, N-terminal prohormone of B-type natriuretic peptide.

3). Figure 3 shows absolute 3-year global CVD event rates in the combined cohorts stratified by PCE-estimated cardiovascular risk category and the number of abnormal test results. Even among individuals at low (<5%) PCE-estimated 10-year ASCVD risk, the absolute 3-year global CVD risk among individuals with scores of ≥3 was >5%. Conversely, individuals with borderline PCE-estimated 10-year risk (5%–7.5%)

Table 2. Hazard Ratios (95% CIs) for the Associations of Biomarkers With Risk of Global CVD Events in the Combined Cohorts

	Unadjusted	Adjusted for Base Model*	Adjusted for Base Model and eGFR	Adjusted for Base Model, eGFR, and the Other Biomarkers
Total, n	16 581	16 551	16 506	16 506
Number of events	553	553	551	551
Biomarkers as categorical variables				
ECG-LVH	1.11 (0.87–1.43)	1.10 (0.85–1.41)	…	…
hs-CRP ≥3 mg/L	1.4 (1.18–1.65)	1.30 (1.08–1.56)	1.28 (1.07–1.54)	1.23 (1.03–1.48)
NT-proBNP ≥100 pg/mL	2.06 (1.73–2.44)	2.20 (1.82–2.67)	2.16 (1.78–2.62)	2.00 (1.65–2.43)
hs-cTnT ≥5 ng/L	2.16 (1.80–2.60)	1.47 (1.21–1.79)	1.45 (1.19–1.77)	1.32 (1.08–1.62)
Plaque and/or IMT >75th percentile (ARIC only)	3.31 (2.69–4.08)	1.96 (1.57–2.45)	1.95 (1.56–2.44)	1.8 (1.43–2.26)
CAC >10 (DHS and MESA only)	5.07 (4.24–6.05)	2.38 (1.96–2.90)	2.38 (1.96–2.89)	2.2 (1.81–2.68)
Plaque and/or IMT >75th percentile and/or CAC >10 (combined cohorts)	3.28 (2.69–3.99)	2.16 (1.76–2.67)	2.15 (1.75–2.65)	1.99 (1.61–2.46)
Biomarkers as log-transformed continuous variables				
hs-CRP (log)	1.26 (1.16–1.36)	1.18 (1.07–1.29)	1.17 (1.06–1.28)	1.13 (1.03–1.24)
NT-proBNP (log)	1.87 (1.72–2.03)	1.65 (1.52–1.80)	1.70 (1.55–1.87)	1.59 (1.44–1.75)
Hs-cTnT (log)	1.90 (1.77–2.04)	1.36 (1.24–1.49)	1.35 (1.23–1.49)	1.19 (1.08–1.31)

ARIC indicates Atherosclerosis Risk in Communities study; CAC, coronary artery calcium score; ECG-LVH, left ventricular hypertrophy by ECG; HDL, high-density lipoprotein; hs-CRP, high sensitivity C-reactive protein; hs-cTnT, high-sensitivity cardiac troponin T; IMT, intima-media thickness; LDL, low-density lipoprotein; and NT-proBNP, N-terminal prohormone of B-type natriuretic peptide.

*Base model adjustment included pooled cohort equation variables (age, sex, black race, total cholesterol, high-density lipoprotein [HDL] cholesterol, systolic blood pressure, antihypertensive medication use, current smoking, and diabetes mellitus status) and body mass index.

but an integer score of 0 or 1 had an absolute 3-year global CVD risk <1.5%.

Consistent with previous findings from DHS and MESA,[14] the predictive value of integer scores in this study was preserved in separate analyses of longer term, 10-year global CVD and ASCVD risk (data not shown).

DISCUSSION

Plasma and Imaging Biomarkers Improve 3-Year CVD Risk Prediction

In the present study, we tested whether a combination of biomarkers including both plasma- and imaging-based tests reflecting distinct pathophysiological processes is useful for 3-year global CVD and ASCVD risk stratification in adults free from CVD at baseline. Participants enrolled in 3 independent population-based cohorts were pooled in the study, a strategy planned prospectively because of small numbers of CVD events expected over the 3-year follow-up period. We included in our analyses circulating biomarkers reflecting neurohormonal activation and cardiac wall stretch (NT-proBNP), cardiomyocyte injury (hs-cTnT), and inflammation (hs-CRP), imaging test results reflecting subclinical atherosclerosis (a composite of IMT or plaque in one cohort, and CAC in the other 2 cohorts), as well as ECG measures of cardiac hypertrophy (ECG-LVH). Each test result except for ECG-LVH (which was therefore omitted from subsequent analyses) was independently associated with the primary outcome of global CVD after full adjustment for traditional CVD risk factors, body mass index, eGFR, and the other test results. These findings suggest that NT-proBNP, hs-cTnT, hs-CRP, and imaging markers of subclinical atherosclerosis each contributes non-redundant information for short-term (3-year) global CVD risk prediction.

Multimodality Strategy for Short-Term CVD Risk Assessment

To estimate the combined predictive value of these tests taken together, we calculated a simple integer score reflecting the number of abnormal tests results in each study participant, with possible values from 0 to 4. This multimodality strategy for CVD risk assessment has been proposed previously for longer-term risk,[14] is relatively easy to implement in both clinical and research settings, and can potentially be used even in patients with missing or incomplete data. Moreover, our findings suggest that this approach could be used

Table 3. Hazard Ratios (95% CIs) for the Associations of Biomarkers With Risk of Atherosclerotic CVD Events in the Combined Cohorts

	Unadjusted	Adjusted for Base Model*	Adjusted for Base Model and eGFR	Adjusted for Base Model, eGFR, and the Other Biomarkers
Total, n	16 581	16 565	16 519	16 519
Number of events	260	260	259	259
Biomarkers as categorical variables				
ECG-LVH	1.15 (0.89–1.50)	0.98 (0.68–1.42)	···	···
hs-CRP ≥3 mg/L	1.31 (1.02–1.67)	1.12 (0.92–1.56)	1.19 (0.91–1.55)	1.14 (0.88–1.49)
NT-proBNP ≥100 pg/mL	1.85 (1.43–2.38)	1.76 (1.33–2.33)	1.73 (1.3–2.31)	1.66 (1.25–2.21)
hs-cTnT ≥5 ng/L	1.8 (1.38–2.34)	1.07 (0.81–1.42)	1.06 (0.8–1.41)	0.98 (0.74–1.31)
Plaque and/or IMT >75th percentile (ARIC only)	3.43 (2.49–4.72)	2.03 (1.45–2.85)	2.01 (1.43–2.83)	1.91 (1.36–2.67)
CAC >10 (DHS and MESA only)	4.14 (3.25–5.28)	1.66 (1.27–2.16)	1.66 (1.27–2.16)	1.56 (1.20–2.03)
Plaque and/or IMT >75th percentile and/or CAC >10 (combined cohorts)	3.12 (2.35–4.13)	1.80 (1.34–2.41)	1.78 (1.33–2.4)	1.69 (1.26–2.28)
Biomarkers as log-transformed continuous variables				
hs-CRP (log)	1.20 (1.07–1.35)	1.11 (0.97–1.27)	1.10 (0.96–1.27)	1.08 (0.94–1.24)
NT-proBNP (log)	1.63 (1.45–1.84)	1.37 (1.21–1.55)	1.39 (1.21–1.59)	1.33 (1.16–1.52)
Hs-cTnT (log)	1.81 (1.62–2.01)	1.21 (1.05–1.38)	1.20 (1.04–1.38)	1.10 (0.95–1.27)

ARIC indicates Atherosclerosis Risk in Communities study; CAC, coronary artery calcium score; ECG-LVH, left ventricular hypertrophy by ECG; HDL, high-density lipoprotein; hs-CRP, high sensitivity C-reactive protein; hs-cTnT, high-sensitivity cardiac troponin T; IMT, intima-media thickness; LDL, low density lipoprotein; and NT-proBNP, N-terminal prohormone of B-type natriuretic peptide.

*Base model adjustment included pooled cohort equations variables (age, sex, black race, total cholesterol, high-density lipoprotein cholesterol, systolic blood pressure, antihypertensive medication use, current smoking, and diabetes mellitus status) and body mass index.

with either carotid or coronary imaging approaches. All integer scores ≥1 in our study were associated with significantly and incrementally higher risk of both global CVD and ASCVD compared with those with a score of 0. Almost 1 in 10 study participants with an integer score ≥3, and 1 in 25 of those with an integer score of 2, had a global CVD event within 3 years, compared with 1 in 145 of those with an integer score of 0. Findings were consistent in multiple subgroups, and absolute risk for those with higher scores was notable even among individuals at low predicted risk based on the PCE. Conversely, those with an integer score of 0 or 1 had a low absolute short-term risk across PCE risk categories.

In addition, sensitivity analyses conducted in the subset of participants that had either CAC=0 or normal carotid IMT at baseline found that integer scores of 2 or 3 (now based only on plasma biomarkers, thus having possible values from 0 to 3) were still associated with a significantly higher 3-year risk of global CVD, suggesting that an initial finding of normal CAC or IMT does not obviate the utility of the other tests.

Table 4. Changes in c-Statistic With All Four Biomarkers Added to the Base Model*, Both as Categorical and as Continuous Variables, in the Combined Cohorts

Global CVD			Atherosclerotic CVD		
c-Statistic Base Model (95% CI)	c-Statistic Base Model+Biomarkers (95% CI)	*P* Value for c-Statistic w/ vs w/o Biomarkers	c-Statistic Base Model (95% CI)	c-Statistic Base Model+Biomarkers (95% CI)	*P* Value for c-Statistic w/ vs w/o Biomarkers
Biomarkers as categorical variables†					
0.780 (0.762, 0.798)	0.801 (0.784, 0.818)	<0.0001	0.799 (0.774, 0.823)	0.812 (0.789, 0.835)	0.006
Biomarkers as log-transformed continuous variables					
0.780 (0.762, 0.798)	0.805 (0.788, 0.822)	<0.0001	0.799 (0.774, 0.823)	0.811 (0.788, 0.834)	0.006

CAC indicates coronary artery calcium; CVD, cardiovascular disease; hs-CRP, high-sensitivity C-reactive protein; IMT, intima-media thickness; NT-proBNP, N-terminal pro B-type natriuretic peptide.

*Base model includes traditional CVD risk factors, including pooled cohort equation variables (age, sex, black race, total cholesterol, high-density lipoprotein [HDL] cholesterol, systolic blood pressure, antihypertensive medication use, current smoking, and diabetes mellitus status), and bone mass index.

†Thresholds were hs-CRP ≥3 mg/L; NT-proBNP ≥100 pg/mL; hs-cTnT ≥5 ng/L; plaque and/or IMT >75th percentile and/or CAC >10.

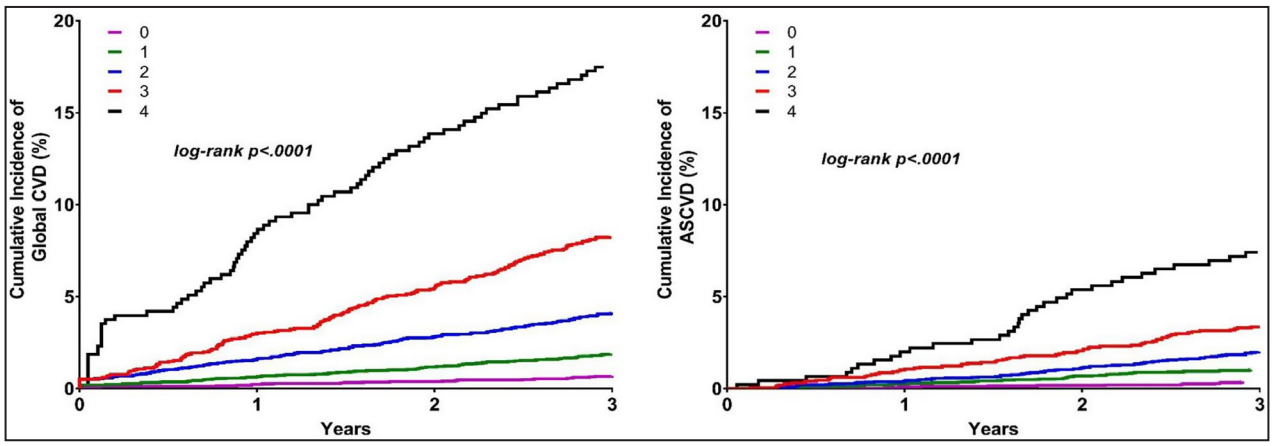

Figure 1. Cumulative incidence rates of global cardiovascular disease (CVD) and atherosclerotic cardiovascular disease composite outcomes stratified by the number of abnormal test results.
We pooled data from participants in 3 large population-based cohorts, free from CVD at baseline (N=16 581), with measurements including N-terminal pro-B-type natriuretic peptide; high-sensitivity cardiac troponin T; high-sensitivity C-reactive protein; and a composite imaging measure of subclinical atherosclerosis (coronary artery calcium or carotid intima-media thickness or plaque). Using an integer score to count the number of abnormal tests in each study participant, higher integer scores were associated with higher incidence of global CVD events (myocardial infarction, stroke, coronary revascularization, incident heart failure, or atrial fibrillation) and atherosclerotic CVD events (fatal or non-fatal myocardial infarction or stroke) during 3 years of follow-up. ASCVD indicates atherosclerotic cardiovascular disease; and CVD, cardiovascular disease.

Clinical Applications

The multi-modality approach for 3-year global CVD risk assessment tested in the present study may have several clinical applications. First, accurate estimation of short-term global CVD risk is particularly important for the medical evaluation of apparently healthy individuals before critical missions, such as prolonged space flight. The estimated duration of a crewed mission to Mars is up to 3 years, and any CVD event affecting an astronaut during this time may become both life- and mission-threatening. Other high-risk or high-stakes occupations may also benefit from accurate short-term global CVD risk assessment, either to assess job readiness or to guide the type and intensity of preventive measures. Examples include airline

Table 5. Association Between the Number of Abnormal Biomarkers and Composite CVD End Points in the Combined Cohorts

	n (% of Overall Cohort)	Number of Events	Absolute 3-y Event Rate	Unadjusted Hazard Ratio (95% CI)	Adjusted * Hazard Ratio (95% CI)
Global CVD					
Overall cohort	16 567	553	3.38%	···	···
Integer score 0	3917 (23.6%)	27	0.69%	Ref	Ref
Integer score 1	5970 (36%)	112	1.88%	2.74 (1.8, 4.18)	2.01 (1.32, 3.08)
Integer score 2	4271 (25.8%)	176	4.12%	5.93 (3.95, 8.9)	3.03 (1.99, 4.61)
Integer score 3	1956 (11.8%)	160	8.18%	11.67 (7.74, 17.59)	4.56 (2.95, 7.06)
Integer score 4	453 (2.8%)	78	17.22%	28.89 (16.66, 40.23)	7.85 (4.84, 12.72)
Atherosclerotic CVD					
Overall cohort	16 581	260	1.57%	···	···
Integer score 0	3917 (23.6%)	15	0.38%	Ref	Ref
Integer score 1	5972 (36%)	61	1.02%	2.73 (1.55, 4.8)	1.87 (1.05, 3.31)
Integer score 2	4276 (25.8%)	85	1.99%	5.28 (3.04, 9.17)	2.33 (1.32, 4.13)
Integer score 3	1961 (11.8%)	66	3.37%	8.78 (4.99, 15.45)	2.73 (1.49, 5.00)
Integer score 4	455 (2.7%)	33	7.25%	19.12 (10.33, 35.41)	4.34 (2.20, 8.56)

Hazard ratios are vs participants with no abnormal biomarkers (ie, integer score of 0).
*Adjusted for pooled cohort equation variables (age, sex, black race, total cholesterol, high-density lipoprotein [HDL] cholesterol, systolic blood pressure, antihypertensive medication use, current smoking, and diabetes mellitus status), body mass index, and eGFR. CVD indicates cardiovascular disease; eGFR, estimated glomerular filtration rate.

pilots, commercial truck drivers especially when transporting hazardous materials, and certain active-duty military personnel. For instance, US Navy submarine officers are typically attached to a submarine for 3 years,[30] with many months of continuous deployment during this time, including in remote regions of the globe where timely medical assistance or evacuation may be difficult or impossible.

These findings also may have broader implications for cardiovascular prevention beyond high-risk occupations. Short-term risk assessment may also prove beneficial to communicate risk and promote healthy behavioral changes for CVD prevention, including among apparently healthy individuals. Individual awareness of both short-term and longer-term risk estimates may be more effective in motivating healthy behavioral changes than longer-term risk alone, because risk perception is profoundly affected by the imminence of risk, and health risk perception is a key motivator for healthy behavioral changes.[31–33] Multiple abnormal test results identified individuals at substantial 3-year global CVD risk, even when estimated 10-year ASCVD risk was low. Such patients may warrant more aggressive lipid and blood pressure control targets than would be recommended based on the PCE, although the utility and cost-effectiveness of extensive cardiovascular screening in young asymptomatic individuals, outside of high-risk occupations, are presently unknown. Finally, while not addressed in this study, another potential application of short-term CVD

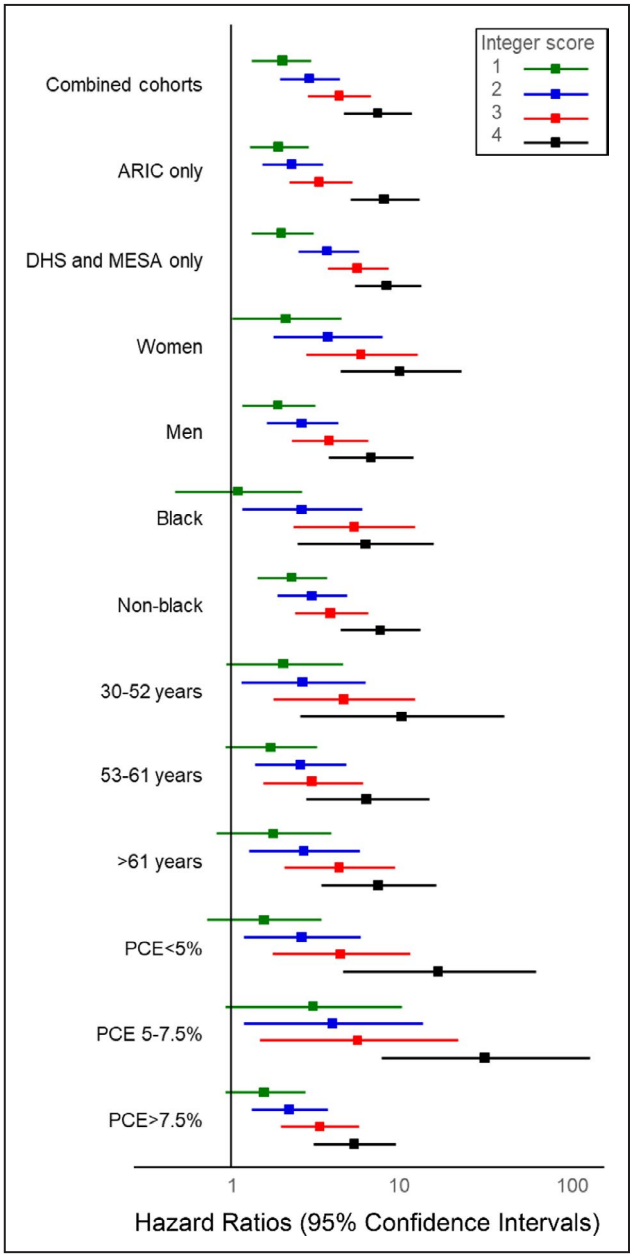

Figure 2. Association between the number of abnormal test results and global cardiovascular disease outcomes in study subgroups.
Higher integer scores (representing the number of abnormal test results) were associated with global cardiovascular disease after multivariable adjustment regardless of the type of subclinical atherosclerosis imaging test used (carotid intima-media thickness or plaque in ARIC; coronary artery calcium in DHS and MESA), and across categories of sex, race, age, and 10-year cardiovascular risk estimated using pooled cohort equations. There were no significant interactions across subgroups (*P* interaction >0.05 across each subgroup). ARIC indicates Atherosclerosis Risk in Communities study; DHS, Dallas Heart Study, MESA, Multi-Ethnic Study of Atherosclerosis; and PCE, pooled cohort equations.

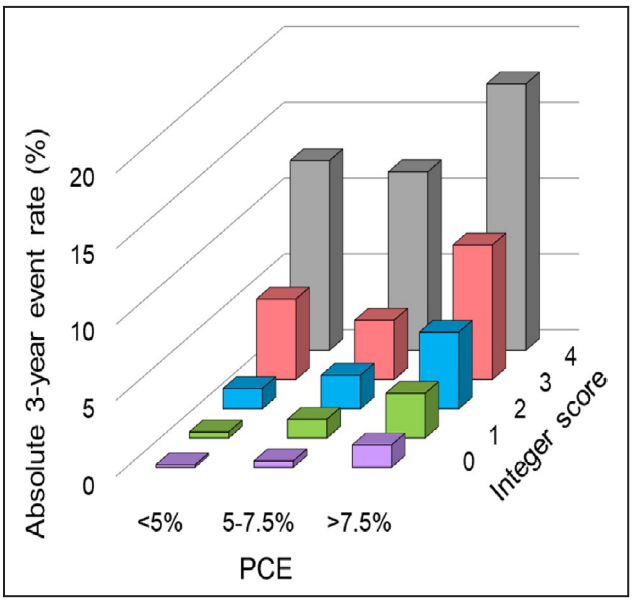

Figure 3. Absolute 3-year global cardiovascular disease event rates in the combined cohorts stratified by pooled cohort equation-estimated risk and the number of abnormal test results.
Higher integer scores (representing the number of abnormal test results) were associated with higher 3-year global cardiovascular disease risk across pooled cohort equation categories, and, conversely, a low integer score (0 or 1) was associated with lower global cardiovascular disease risk even among individuals with borderline or elevated 10-year risk by pooled cohort equations. PCE indicates pooled cohort equations.

risk assessment may be serial evaluation in the same individual over time, providing a time-updated risk profile that may be used as a tool for precision CVD prevention. Further studies are required to explore this possibility.

Study Findings in Context

Few prior data are available evaluating short-term risk assessment approaches. A recent study evaluating older adults (mean age, 75 years) free from CVD at the time of ARIC visit 5 demonstrated improved prediction of global CVD events over a median of ≈4 years, when the PCE was combined with NT-pro-BNP, hs-cTnT, and hs-CRP.[34] To our knowledge, our study is the first to combine biomarkers with imaging, and focused on a much younger population that would be more relevant for cardiac evaluation before selection for high-risk occupations.

Strengths and Limitations

Strengths of the present study include the large sample size of >16 500 individuals free from CVD at baseline, from 3 diverse and well-characterized population-based cohorts, with >550 global CVD events and 250 ASCVD events over the 3-year follow-up period. Methods of biomarker assessment were harmonized across the cohorts, comprehensive data capture was accomplished in nearly all participants, and rigorous and blinded assessment of outcomes was performed. Study findings were robust across subgroups defined by age, sex, race, type of imaging test used, and PCE-estimated cardiovascular risk category, and were qualitatively similar when plasma biomarkers were analyzed as categorical and as continuous variables.

This study has several limitations. First, the measurement of subclinical atherosclerosis differed in ARIC versus DHS and MESA. Importantly, however, our stratified analyses suggest that the carotid measurement in ARIC provided comparable prognostic information as CAC in DHS and MESA. Second, we used a simple integer score counting the number of abnormal test results as a simple way to facilitate real-life application of this novel risk assessment tool, but it is possible that other approaches that do not give the same weight to various biomarkers may perform better. Finally, measurements were performed at a single time point. Future studies are needed to validate these findings, and to determine whether periodic reassessment and incorporation of time-updated test results enhance risk prediction.

CONCLUSIONS

A novel strategy incorporating 3 different plasma biomarkers and a composite imaging measure of subclinical atherosclerosis improves assessment of 3-year global and atherosclerotic CVD risk among adults without known CVD. Even among individuals at low PCE-estimated ASCVD risk (<5% estimated 10-year risk), the absolute 3-year global CVD risk among individuals with scores of ≥3 was >5%. Further studies are required to explore the utility of this novel short-term risk assessment tool for CVD prevention, including for the development of precision CVD prevention strategies.

ARTICLE INFORMATION

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