Identifying Young Adults At High Risk Of Cardiometabolic Disease Using Cluster Analysis and the Framingham 30-yr Risk Score

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Abstract

Background and Aims: Current strategies to reduce cardiovascular disease (CVD) risk in young adults are largely limited to those at extremes of risk. In cohort studies we have shown cluster analysis identified a large sub-group of adolescents with multiple risk factors. This study examined if individuals classified at 'high-risk' by cluster analysis could also be identified by their Framingham risk scores.

Methods and Results: Raine Study data at 17- (n=1048) and 20-years (n=1120) identified high- and low-risk groups by cluster analysis using continuous measures of systolic BP, BMI, triglycerides and insulin resistance. We assessed:- CVD risk at 20-years using the Framingham 30yr-risk-score in the high- and low-risk clusters, and cluster stability from adolescence to adulthood.

Cluster analysis at 17- and 20-years identified a high-risk group comprising, 17.9% and 21.3%, respectively of the cohort. In contrast, only 1.2% and 3.4%, respectively, met the metabolic syndrome criteria, all of whom were within the high-risk cluster. Compared with the low-risk cluster, Framingham scores of the high-risk cluster were elevated in males (9.4%; 99%CI 8.3, 10.6 vs 6.0%; 99%CI 5.7, 6.2) and females (4.9%; 99%CI 4.4, 5.4 vs 3.2%; 99%CI 3.0, 3.3) (both $P<0.0001$). A score >8 for males and >4 for females identified those at high CVD risk with 99% confidence.

Conclusion: Cluster analysis using multiple risk factors identified ~20% of young adults at high CVD risk. Application of our Framingham 30yr-risk cut-offs to individuals allows identification of more young people with multiple risk factors for CVD than conventional metabolic syndrome criteria.

Key words: cardiovascular risk, cluster analysis, young adults, adolescents, Framingham scores

Introduction

The World Health Organization 2018 report indicates that 18.4% of individuals aged 5-19 years are overweight or obese [1]. In Australia, the 2014-15 Australian Institute of Health and Welfare reported that 16.5% of those aged 18-21 years were obese, more than double that of individuals of the same age born 20 years earlier (7.2%) [2]. Adiposity in childhood and adolescence associates with higher blood pressure, dyslipidaemia and relative insulin resistance. Clustering of cardiometabolic risk factors has traditionally been defined as the metabolic syndrome (MetS) that associates with pro-inflammatory and pro-thrombotic states [3], and is a substantial contributor to future disease burden associated with CVD [4]. Recognition of individuals at increased cardiometabolic risk at a young age offers an opportunity to intervene early to prevent future cardiovascular disease (CVD). However, the use of MetS cut-points [5, 6] for several quantitative variables that are each linearly related to CVD is likely to substantially underestimate the number of young adults at high risk of later cardiovascular disease, diabetes and related metabolic abnormalities.

We have previously used cluster analysis to assess CVD risk in children and adolescents [7-9] in the Raine Study a pregnancy cohort study from Western Australia. An advantage of this approach is that it considers multiple co-existing CVD risk factors as continuous variables to assign individuals to a high- or low-risk cardiometabolic cluster. Cluster analysis arranges the groupings such that within a single cluster, the participants are relatively homogeneous, sharing similar traits and being dissimilar to those in the other cluster. At the age 14 follow-up, 29.1% of adolescents were identified as high-risk using cluster analysis whereas only 2.2% had the MetS using International Diabetes Federation (IDF) paediatric criteria [9].

Given that cluster analysis is not a diagnostic tool that can used for individuals in a clinical setting, this study used data from the Raine Study at 20-years of age to examine if individuals classified at 'high risk' by cluster analysis could also be identified by their Framingham 30yr-risk-score[10]. The high- and low-risk clusters were generated for each sex using triglycerides, BMI, the Homeostatic Model Assessment for Insulin Resistance (HOMA-IR) and systolic BP (SBP). We also report the stability of the high-risk cluster between 17- and 20-years of age, and the association between clusters and a range of inflammatory markers known to be associated with increased risk of CVD.

Methods

Study populations

The Raine Study recruited 2900 pregnant women from the general population at King Edward Memorial Hospital (Subiaco, Western Australia) and nearby private practices from 1989-1991 [11]. The women gave birth to 2868 babies (Generation 2). The 17- and 20-year follow-ups of Generation 2 involved 1726 and 1462 participants, respectively. Human ethics committees of King Edward Memorial Hospital, Princess Margaret Children's Hospital and the University of Western Australia approved the study. Participation was voluntary and informed written consent was obtained. At 17- and 20-years of age, data from 1048 and 1120 participants, respectively, were used in cluster analysis and to define the MetS. At 17-years, plasma cytokines and adipokines were measured in 1027 participants. The stability of cluster membership was assessed in 806 participants who had measurements at both survey times. Those included in the analysis on both occasions were not different to those excluded from analysis in terms of birthweight, sex and BMI at 17- and 20-years (all $P>0.2$).

Clinical and laboratory measurements

Body weight and height were measured using a Wedderburn Chair Scale and a Holtain Stadiometer, respectively. Body mass index (BMI) was calculated as weight (kg)/height^2 (m). Waist circumference was measured at the umbilicus level with a tape measure (to the nearest 0.1 cm). Information on smoking was obtained from questionnaires at 17-and 20-years. Resting systolic and diastolic BP were measured after 5 mins supine rest using an oscillometric sphygmomanometer (DINAMAP 8100, DINAMAP XL or DINAMAP ProCare 100 vital signs monitors; GE Healthcare, USA). Six readings were recorded, each two minutes apart, with the last five readings averaged.

Fasting bloods were analysed for serum insulin, glucose, triglycerides, total cholesterol, HDL-cholesterol, and high-sensitivity C-reactive protein (hsCRP) (PathWest Laboratory, Royal Perth Hospital). LDL-cholesterol was calculated using the Friedewald equation. Insulin resistance was estimated using HOMA-IR calculated as fasting insulin [µU/ml] x fasting glucose [mmol/L]/22.5). Serum leptin was measured by the ACTIVE® Human Leptin ELISA kit (DSL-10-23100, Diagnostic Systems Laboratories, Webster, TX, USA) and adiponectin by the Quantikine® Human Total Adiponectin/Acrp30 Immunoassay (R&D Systems, Minneapolis, MN, USA).

Plasma interleukin-18 (IL-18) was measured by ELISA (Medical Biological Laboratories, Nagoya, Japan) and plasma IL-18 binding protein (IL-18BP primary isoform a) using a DuoSet ELISA development system (R&D Systems, Minneapolis, MN, USA). Plasma IP-10, sTNFR1 and sTNFR2 were quantified using Cytometric Bead Array (CBA) Flex sets (BD PharMingen, San Diego, CA) on a BD FACSArray^{TM} bioanalyser (BD Biosciences, San Jose, California, USA). Individual cytokine concentrations were determined using FCAP Array software (BD Biosciences).

Cluster Analysis, the MetS and the Framingham 30-year CVD risk score

Two-step cluster analysis was used to identify individuals at high risk. This technique is suitable for data with natural groupings, and classifies data into subsets, known as clusters, such that between-subject variation is minimized within a cluster and maximised between clusters[12]. Using triglycerides, BMI, HOMA-IR, and SBP as continuous variables in each sex at 17- and 20-years, two cluster groups were identified as previously described [7, 9]. The high-risk cluster characteristically had significantly elevated waist circumference, SBP, triglycerides and glucose and reduced HDL-cholesterol compared with the low-risk cluster.

The MetS was defined using the IDF clinical criteria at each time-point [6].

To assess the potential long-term significance of the high-risk cluster, the Framingham 30-year CVD risk score[10] (https://framinghamheartstudy.org/fhs-risk-functions/cardiovascular-disease-30-year-risk/) was calculated using data collected at 20 years. A Framingham 30yr-risk-score was calculated for each individual using age, sex, SBP, use of antihypertensive treatment (yes/ no), smoking, diabetes mellitus, total cholesterol, and HDL-cholesterol as input variables. Increasing age, diabetes, smoking, use of antihypertensive treatment and male sex have the highest hazard ratios (between 1.4 and 2) and are the largest contributors to a high Framingham risk score. Elevated SBP and total cholesterol also contribute to a high score (hazard ratios >1.2), whereas increasing HDL-cholesterol is protective (hazard ratio < 1). The equation is validated for ages 20-60 years and provides estimates of 30-year risk of 'Hard CVD' (coronary death, myocardial infarction, and fatal and nonfatal stroke) and 'Full CVD' (hard CVD or coronary insufficiency, angina pectoris, transient ischaemic attack, intermittent

claudication, and congestive heart failure), adjusted for the competing risk of non-CVD death.

Statistical analyses

Analyses were performed using SPSS (version 23.0). Skewed data were log-transformed where appropriate. Subject characteristics were described by arithmetic or geometric means and 95% confidence intervals.

Cluster analysis

Crosstabs in SPSS were used to assess the prevalence of the high- and low-risk cluster groups and the MetS at each time-point [6]. One-way ANOVA with Bonferroni correction was used to assess differences in cardiometabolic risk factors according to cluster at 17- and 20-years.

Framingham 30yr-risk-score

Between cluster differences in the Framingham 30yr-risk-score at 20-years was assessed using one-way ANOVA with Bonferroni correction. The lower 99% confidence interval that associated with the high-risk cluster for each sex was used as a guide for identification of individuals at high-risk of future CVD.

Cluster stability

Crosstabs in SPSS were used to assess changes in high- and low-risk cluster membership between 17-20 years as a measure of cluster stability. CVD risk factors at 17-and 20-years and the Framingham 30yr-risk-score at 20-years were compared in individuals that remained in, or changed clusters using one-way ANOVA with Bonferroni correction.

The effect of cluster and sex on adipokines and cytokines at 17-years was assessed using univariate analysis with sex as a covariate.

Results

Cardiometabolic risk clusters at 17- and 20-years

At 17-years, 17.9% of participants (16.4% of males, 19.5% of females) were in a high-risk cluster and 82.1% (83.6% of males, 80.5% of females) were in the low-risk cluster (Table 1). Males and females in the high-risk cluster had substantially higher waist circumference, BMI, HOMA-IR, SBP, DBP, cholesterol, triglycerides, and hs-CRP and lower HDL-cholesterol. Plasma cytokines sTNFR1, sTNFR2 were also significantly elevated in the high-risk cluster in both sexes. Serum leptin was significantly elevated and adiponectin significantly reduced in the high-risk cluster males and females (Table 1). Lower levels of IP-10, IL-18BP, sTNFR1 and sTNFR2 were observed in females compared with males when the clusters were combined ($p<0.001$).

At 20-years a similar pattern was observed: the high-risk cluster comprised 21.3% of participants (16.1% of males, 24.1% of females) (Table 2). Individual risk factors were all significantly elevated in the high-risk cluster regardless of sex.

The MetS was identified in only 1.2% of the cohort at 17-years (2% of males, 0.4% of females) and 3.4% at 20-years (5.3% of males, 1.2% of females). On both occasions, those with the MetS were identified within the high-risk cluster (Tables 1 and 2).

Framingham 30yr-Risk-Score at 20-years

The Framingham 30yr-risk-score clearly delineated the clusters at 20-years (Table 2, Figure 1). The risk score for individuals in the high-risk cluster was elevated by 58% in males and 53% in females (both $P<0.0001$) compared with those in the low-risk cluster. The mean and 99% confidence intervals for the Framingham risk scores according to cluster are shown in Table 2. These values indicate that a Framingham score >8 for males and >4 for females will identify young adults at high-risk of CVD with 99%

confidence. For CVD based on coronary death, myocardial infarction, and fatal and nonfatal stroke, the risk scores were >6 for males and >3 for females. Figure 1 demonstrates that the high-risk cluster is clearly separated from the low-risk cluster in terms of Framingham risk score and individual risk factors for CVD. Within the high-risk cluster, individuals with the MetS had the highest Framingham risk scores: males 10.9% (95% CI 9.5,12.2) and females 7.4% (95% CI 4.3,10.4), compared with the remainder of the high-risk cluster: 8.6% (95% CI 7.4, 9.8) and females 4.7% (95% CI 4.4, 5.0)

Stability of cluster membership between 17- and 20-years and Framingham risk score

There was a high degree of stability of cluster membership from 17- to 20-years with 90% (93% of males and 84.7% of females) of the low-risk cluster and 72% (60.2% of males and 85.5% of females) of the high-risk cluster remaining in the same cluster. Compared with the low-risk cluster, the Framingham risk scores were elevated ($P<0.001$) in males (Figure 2A) and females (Figure 2B) that remained or moved to high-risk cluster ($P<0.01$). Conversely, those who remained or were reclassified to a low-risk cluster had lower Framingham risk scores (in males both $P<0.01$; females $P<0.01$ and $P=0.06$, respectively) compared with those who remained in the high-risk cluster.

Discussion

We have shown for the first time that the Framingham 30yr-risk-score clearly discriminates young adults at high risk for CVD with 99% confidence as identified using cluster analysis. Individuals in a high-risk cluster, who represented 18-21% of the cohort at 17- and 20-years of age, had a Framingham score for total CVD of > 8 for

males and > 4 for females at 20-years. These values are fourfold higher than values considered to be optimal for males (2%) and females (1%) aged 20-years, according to the Framingham 30yr-risk calculator (https://framinghamheartstudy.org/fhs-risk-functions/cardiovascular-disease-30-year-risk/). For Framingham predicted events of coronary death, myocardial infarction, and fatal and nonfatal stroke, the risk scores were >6 for males and >3 for females. Individuals identified at high-risk for CVD had substantially increased BMI (+7kg/m^2), waist circumference (+13cm), SBP (+10mmHg), triglycerides (~60%) and HOMA-IR (~2-3 fold) and lower HDL-cholesterol.

Cluster movement between adolescence and young adulthood associated with a high degree of stability with 72% of high-risk cluster and 90% of the low-risk cluster participants remaining in the same cluster. The Framingham risk score was responsive to, and reflected changes in cluster between adolescence and young adulthood, suggesting that this risk score could be employed to assess the success of individual or group lifestyle management to reduce CVD risk.

The prevalence of the high-risk cluster is consistent with our analyses of the Raine Study at 8 and 14-years of age [8, 9]. In contrast, a very small proportion, less than 4% of the cohort met the criteria for the MetS. The very low prevalence of the MetS is similar to that of other cohorts in adolescence [13, 14]. Importantly all individuals with the MetS were encompassed within the high-risk cluster.

CVD risk factors (smoking, BMI, SBP, DBP, LDL- and HDL-cholesterol) measured at younger ages have been shown to be better predictors of CVD than those measured concurrently with atherosclerosis [15, 16]. However, current approaches to risk

management in adolescents and young adults still tend to focus on thresholds for individual risk factors with labelling of adiposity/obesity, pre/hypertension, pre/diabetes and hypercholesterolaemia, and with less attention to multiple co-existent risk factors at levels below the arbitrary thresholds for disease labelling. Given that atherosclerosis begins in childhood and progresses with aging, the results from these studies provide strong support for early intervention targeting all major risk factors. In our study, the consistent individual risk factors that accompanied change in cluster in either direction were BMI, waist circumference and SBP in males and females, as well as HOMA-IR in females. These findings suggest that high-risk cluster individuals might benefit from early lifestyle intervention in terms of reducing the risk of cardiometabolic complications such as diabetes, hypertension and non-alcoholic fatty liver disease as well as later life onset CVD.

The Framingham 30yr-risk-score clearly delineates the low- and high- risk- clusters in both males and females and has been validated for 20-year-olds. It is relatively easy to use requiring input of commonly measured risk factors https://framinghamheartstudy.org/fhs-risk-functions/cardiovascular-disease-30-year-risk/. Our data suggest that high-risk individuals could be identified with 99% confidence if their risk score for total CVD is >8 for males and >4 for females. This information could be used to assist individuals and health carers to implement lifestyle changes to improve CVD risk at an early age. Individuals with very high levels of individual risk factors that may need more urgent clinical attention would likely also be captured using this approach.

A strength of this study is that we examined the prevalence and stability of a high-risk cluster sequentially in adolescence and young adulthood in a large phenotypically well-

defined population cohort; cluster membership could be defined by an individual's Framingham risk score with a high degree of confidence. Regardless of the fact that different CVD risk factors were used to derive the clusters and the Framingham 30y-risk-score, the high-risk cluster was defined by a significantly higher Framingham score. Further evidence that the high-risk cluster identified individuals at increased risk of future CVD is supported by their elevated levels of a range of inflammatory markers known to be associated with increased risk of cardiometabolic disorders [17-21, 22 , 23] We acknowledge that the cohort was predominantly Caucasian; there was attrition in the cohort from birth to the years of our surveys; and there were reduced numbers of participants studied at both ages. These factors may limit the generalizability of our findings, however, the overall demographic characteristics of individuals studied were not significantly different from the general population in Western Australia [24]. We acknowledge that adolescent and young adult CVD risk assessment using the Framingham risk score is limited and that this risk score was not derived from an Australian population. This however does not detract from the ability of the Framingham scores proposed to identify high-risk individuals with a cluster of cardiometabolic risk phenotypes. Similar studies should be carried out in comparative age population cohorts from other countries and other ethnic groups where the relevant phenotypic data are available. Of value would be further long-term follow up of young adults to assess the validity of this approach for prediction of CVD morbidity and mortality in later life.

In conclusion, cluster analysis using cardiometabolic risk factors has identified a high-risk group defined by the Framingham 30yr-risk-score. A score > 8 for males and > 4 for females identified approximately 20% of 20-year-olds at high-risk of CVD with 99% confidence. These scores if confirmed in other studies could be used to assess

CVD risk in young adults and form a basis for introducing and assessing interventions to reduce CVD risk in young adults.

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Figure legends.

Figure 1. Comparison of Framingham 30-yr Risk of Full CVD and CVD risk factors in the high- and low-risk clusters and for the cohort (All), in males and females at age 20 years.

Values are means and 99% CI for Framingham risk scores and means and 95% CI for BMI, SBP, Triglycerides and HOMA-IR. †P<0.001, compared with the low risk cluster

Figure 2. Stability of cluster membership between 17- and 20-years and Framingham 30-yr Risk of Full CVD.

Values are means and SEM. Bonferroni adjusted group differences [a] P<0.001, compared with the those that stayed in the low-risk cluster and [b] P<0.01, compared with those that stayed in the the high-risk cluster. Stayed in low-risk cluster = (Stayed LRC); moved from the high-risk cluster to the low-risk cluster = (HRC→LRC); moved from the low-risk cluster to the high-risk cluster = (LRC→HRC); stayed in the high-risk cluster = (Stayed HRC).

Table 1 Description of the "Raine cohort" at age of 17 years.

	LR-Cluster		HR-Cluster	
Characteristics	Males (453)	Females (407)	Males (89)	Females (99)
Metabolic syndrome (n)	0	0	11	2
BMI (kg/m^2)	21.4 (21.2, 21.6)	21.5 (21.2, 21.7)	28.7 (27.7,29.8) [a]	29.1 (28.3, 30.2) [a]
Waist circumference (cm)	77.5 (76.8, 78.1)	74.0 (73.2, 74.8)	95.6 (92.6,98.7) [a]	92.4 (89.8, 95.1) [a]
Systolic blood pressure (mmHg)	118 (118, 119)	109 (107, 109)	128 (126, 130) [a]	115 (113, 118) [a]
Diastolic blood pressure (mmHg)	59 (58, 60)	59 (59, 60)	61 (59, 63) [b]	61 (60, 63) [b]
Fasting glucose (mmo/L)	4.8 (4.8, 4.9)	4.6 (4.6, 4.6)	5.0 (4.9, 5.1 [b]	4.8 (4.7, 4.8) [a]
HOMA-IR*^	1.5 (1.4,1.6)	1.6(1.5, 1.6)3	3.7 (3.2, 4.2) [a]	4.6 (3.3,6.0) [a]
Triglycerides (mmol/L)*	0.9 (0.8, 0.9)	0.9 (0.9, 1.0)	1.6 (1.5, 1.8) [a]	1.5 (1.3, 1.6) [a]
Total cholesterol (mmol/L)	3.8 (3.8, 3.9)	4.3 (4.2, 4.3)	4.4 (4.2, 4.5) [a]	4.4 (4.3,4.6) [b]
HDL cholesterol (mmo/L)	1.2 (1.2, 1.3)	1.4 (1.4, 1.5)	1.1 (1.0, 1.1) [a]	1.2 (1.2, 1.3) [a]
Calculated LDL cholesterol (mmol/L)	2.2 (2.1,2.2)	2.4 (2.3, 2.5)	2.5 (2.3,2.6) [b]	2.5 (2.4, 2.7
C-reactive protein (mg/L)*	0.5 (0.4, 0.5)	0.7 (0.6, 0.8)	1.08 (0.8, 1.4) [a]	2.2 (1.7, 2.7) [a]
Adiponectin (mg/L)*	7.4 (7.0, 7.7)	10.5 (9.8, 11.0)	6.0 (5.4, 6.6) [a]	7.7 (7.0, 8.5) [a]
Leptin (mg/L)*	2.8 (2.6, 3.0)	20.0 (81.1, 21.5)	13.6 (11.3, 16.4) [a]	52.1(47.1, 57.8) [a]
IP-10 (pg/mL)*	106 (99, 114)	100 (92, 110)	137 (114, 165) [a]	99 (84, 117)
Total IL-18 (pg/mL)*	288 (276, 299)	280 (269, 291)	314 (287, 342)	328 (305, 353) [a]
IL-18 binding protein (ng/mL)*	13.9 (13.6,14.3)	12.3 (12.1, 12.7)	14.7 (13.7, 15.7)	12.8 (12.2, 13.4)
Soluble TNFα receptor 1 (pg/mL)*	354 (343, 368)	291 (276, 305)	397 (363, 435) [b]	354 (325, 386) [a]
Soluble TNFα receptor 2 (pg/mL)*	3192 (3104, 3281)	3056 (2959, 3157)	3554 (3322, 3803) [b]	3389 (3162, 3632) [a]

Values are means or*Geometric means and 95% CI; [a]P<0.01 [b]P<0.05 comparing HR-Cluster and LR-Clusters within sex

Table 2. Description of the "Raine cohort" at age of at 20 years with Framingham 30-yr risk scores

	LR-Cluster		HR-Cluster	
Characteristics	Males (504)	Females (377)	Males (97)	Females (142)
Metabolic Syndrome (n)	0	0	32	6
Smokers (%)	14.5	11.0	23.2	17.7
BMI (kg/m^2)	23.1 (22.8, 23.3)	21.9 (21.5, 22.2)	30.6 (29.6, 31.7) [a]	29.9 (29.0, 30.9) [a]
Waist circumference (cm)	77.6 (77.0, 78.4)	73.7 (72.8, 74.5)	93.3 (90.2, 96.6) [a]	85.3 (83.0, 87.7) [a]
Systolic blood pressure (mmHg)	121.7 (120.7, 122.8)	108.5 (107.6, 109.5)	131.5 (129.0, 134.0) [a]	119.1 (117.3, 121.0) [a]
Diastolic blood pressure (mmHg)	64.9 (64.2, 65.6)	64.4 (63.4, 65.2)	70.2 (68.3, 71.8) [a]	68.2 (66.9, 69.4) [a]
Triglycerides (mmol/L)*	0.9 (0.9, 0.9)	0.9 (0.8, 0.9)	1.6 (1.4, 1.8) [a]	1.2 (1.2, 1.3) [a]
Total cholesterol (mmol/L)	4.1 (4.0, 4.2)	4.4 (4.4, 4.5)	4.5 (4.4, 4.7) [b]	4.7 (4.5, 4.8) [b]
Calculated LDL cholesterol (mmol/L)	2.4 (2.3, 2.5)	2.5 (2.5, 2.6)	2.6 (2.5, 2.8) [a]	2.8 (2.7, 2.9) [a]
HDL cholesterol (mmo/L)	1.3 (1.2, 1.3)	1.5 (1.4, 1.5)	1.1 (1.0, 1.1) [a]	1.3 (1.2, 1.3) [a]
Fasting glucose (mmo/L)	5.0 (4.9, 5.0)	4.8 (4.7, 4.8)	5.3 (5.2, 5.4) [a]	5.0 (4.9, 5.1)[a]
HOMA-IR*^	0.6 (0.5, 0.6)	0.6 (0.6, 0.6)	2.1 (1.8, 2.5) [a]	1.7 (1.5, 1.9) [a]
C-reactive protein (mg/L)*	0.8 (0.7, 0.9)	1.2 (1.1, 1.4)	1.6 (1.3, 2.0) [a]	2.9 (2.4, 3.4) [a]
Framingham 30-yr risk score				
Full CVD %[††]**	**6.0 (5.7, 6.2)**	**3.2 (3.0, 3.3)**	**9.4 (8.3, 10.6)**	**4.9 (4.4, 5.4)**
Hard CVD %[††]**	**4.2 (4.0, 4.5)**	**1.9 (1.8, 2.0)**	**7.1 (6.0, 8.2)**	**3.0 (2.6, 3.3)**

Values are means or*Geometric means and 95% CI; ** or for Framingham risk scores mean and 99% CI; [††] P<0.0001; [a]P<0.01 [b]P<0.05 comparing HR-Cluster and LR-Clusters within sex

Figure 1

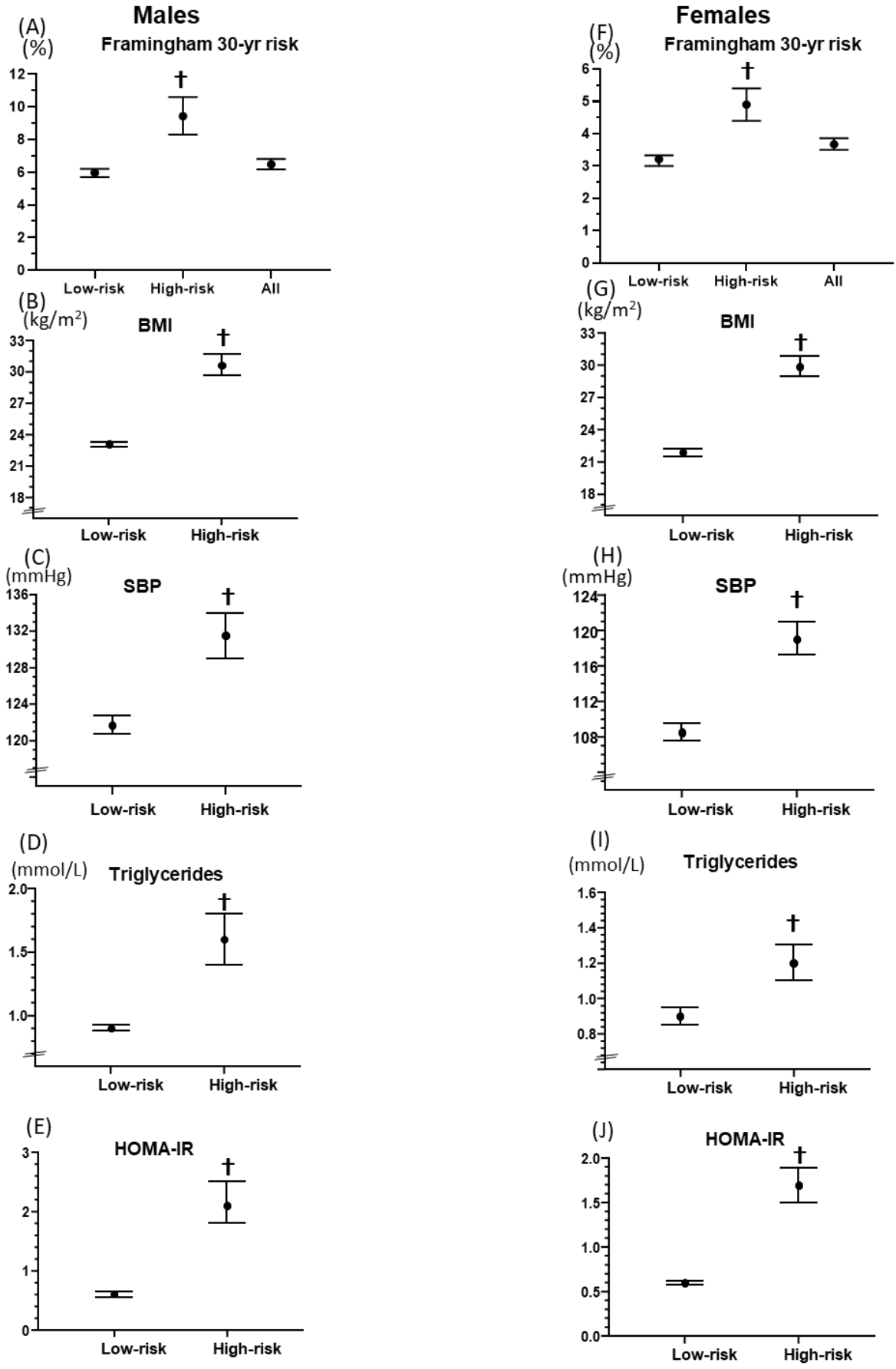
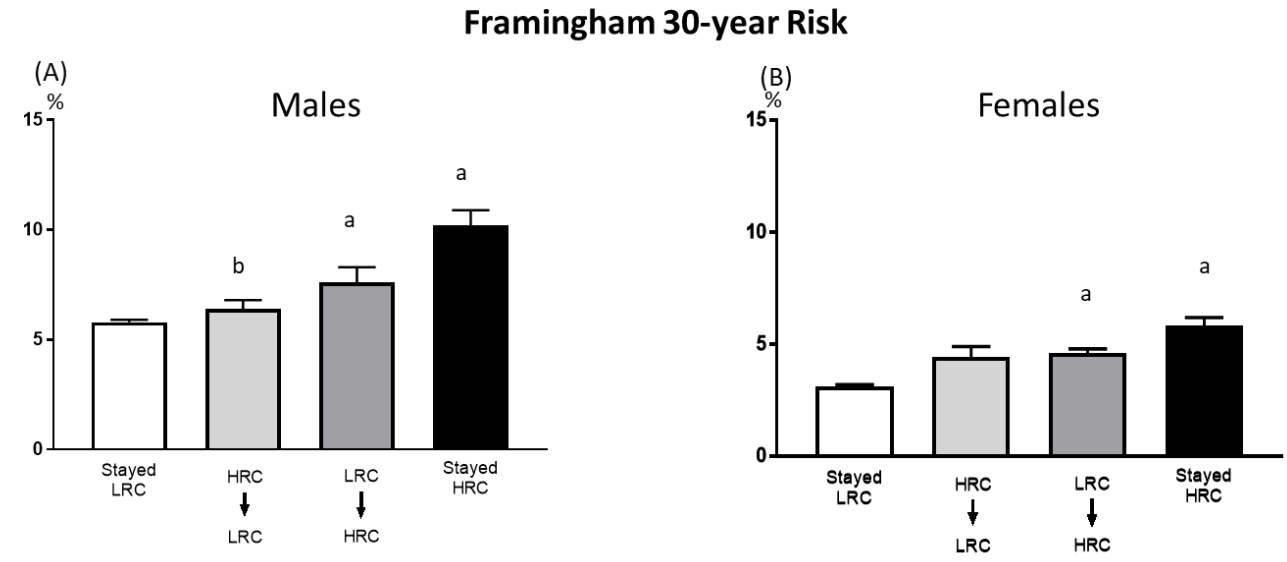

Males
(A)
(%)
Framingham 30-yr risk
12
10
8
6
4
2
0
Low-risk
High-risk
All
(B)
(kg/m²)
BMI
33
30
27
24
21
18
Low-risk
High-risk
(C)
(mmHg)
SBP
136
132
128
124
120
Low-risk
High-risk
(D)
(mmol/L)
Triglycerides
2.0
1.5
1.0
Low-risk
High-risk
(E)
HOMA-IR
3
2
1
0
Low-risk
High-risk
Females
(F)
(%)
Framingham 30-yr risk
6
5
4
3
2
1
0
Low-risk
High-risk
All
(G)
(kg/m²)
BMI
33
30
27
24
21
18
Low-risk
High-risk
(H)
(mmHg)
SBP
124
120
116
112
108
Low-risk
High-risk
(I)
(mmol/L)
Triglycerides
1.6
1.4
1.2
1.0
0.8
Low-risk
High-risk
(J)
HOMA-IR
2.0
1.5
1.0
0.5
0.0
Low-risk
High-risk

Figure 2


Framingham 30-year Risk
(A)
%
Males
15
10
5
0
b
a
a
Stayed LRC
HRC ↓ LRC
LRC ↓ HRC
Stayed HRC
(B)
%
Females
15
10
5
0
a
a
Stayed LRC
HRC ↓ LRC
LRC ↓ HRC
Stayed HRC