Incorporating a polygenic risk score-triaged coronary calcium score into cardiovascular disease examinations to identify subclinical coronary artery disease (ESCALATE): Protocol for a prospective, nonrandomized implementation trial

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Background Identifying and targeting established modifiable risk factors has been a successful strategy for reducing the burden of coronary artery disease (CAD) at the population-level. However, up to 1-in-4 patients who present with ST elevation myocardial infarction do so in the absence of such risk factors. Polygenic risk scores (PRS) have demonstrated an ability to improve risk prediction models independent of traditional risk factors and self-reported family history, but a pathway for implementation has yet to be clearly identified. The aim of this study is to examine the utility of a CAD PRS to identify individuals with subclinical CAD via a novel clinical pathway, triaging low or intermediate absolute risk individuals for noninvasive coronary imaging, and examining the impact on shared treatment decisions and participant experience.

Trial Design The ESCALATE study is a 12-month, prospective, multicenter implementation study incorporating PRS into otherwise standard primary care CVD risk assessments, to identify patients at increased lifetime CAD risk for noninvasive coronary imaging. One-thousand eligible participants aged 45 to 65 years old will enter the study, which applies PRS to those considered low or moderate 5-year absolute CVD risk and triages those with CAD PRS $\geq$80% for a coronary calcium scan. The primary outcome will be the identification of subclinical CAD, defined as a coronary artery calcium score (CACS) >0 Agatston units (AU). Multiple secondary outcomes will be assessed, including baseline CACS $\geq$100 AU or $\geq$75th age-/sex-matched percentile, the use and intensity of lipid- and blood pressure-lowering therapeutics, cholesterol and blood pressure levels, and health-related quality of life (HRQOL).

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Abbreviations: ACS, acute coronary syndrome; ANZCTR, Australian New Zealand Clinical Trials Registry; AU, Agatston units; AUC, area under the receiver-operating characteristic curve; CACS, coronary artery calcium score; CAD, coronary artery disease; CCS, coronary calcium scan; CRF, case report form; CT, computed tomography; CVD, cardiovascular disease; ECG, electrocardiogram; GPAQ, Global Physical Activity Questionnaire; GWAS, genome-wide association studies; HRQOL, health-related quality of life; MACE, major adverse cardiovascular events; MBS, Medicare Benefits Schedule; MESA, Multi-Ethnic Study of Atherosclerosis; MI, myocardial infarction; NHMRC, National Health and Medical Research Council; PBS, Pharmaceutical Benefits Scheme; PPP-CAD, Partnership for Precision Prevention in Coronary Artery Disease; PRO, patient-reported outcome; PRS, polygenic risk score; SC, Steering Committee; SCT, stacked clumping and threshold; SNP, single-nucleotide polymorphism; TMC, Trial Management Committee.

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Conclusion This novel trial will generate evidence on the ability of a PRS-triaged CACS to identify subclinical CAD, as well as subsequent differences in traditional risk factor medical management, pharmacotherapy utilization, and participant experience.

Trial Registration Australian New Zealand Clinical Trials Registry, ACTRN12622000436774. Trial was prospectively registered on March 18, 2022. https://www.anzctr.org.au/Trial/Registration/TrialReview.aspx?id=383134 (Am Heart J 2023;264:163–173.)

Introduction

Clinical outcomes for patients with coronary artery disease (CAD) have improved substantially over the past 6 decades, broadly due to advances in acute management of myocardial infarction (MI) and primary prevention strategies targeting population-level risk factors. However, the substantial, enduring morbidity, mortality, and overall healthcare utilization associated with CAD highlight the unmet need for early plaque detection and equitable application of available pharmacotherapy, such as high-dose statins, which can stabilise plaque and prevent progression and associated events.[1] Whilst dyslipidaemia, blood pressure, smoking, and diabetes as factors can be combined and used to report individual risk using community-level statistics, the many algorithms that rely on these are recognized for their weaknesses in reflecting individual factors mediating the host response. Indeed, it is not uncommon for a patient to present with extensive atherosclerosis and life-threatening heart attack in the absence of any such modifiable risk factors reaching threshold, believed to occur in up to 27% of patients presenting with a first MI attributable to plaque rupture.[2]

Traditional risk prediction & CAD diagnosis

Multivariable CAD risk prediction models have been developed and account for the contribution of multiple fixed and modifiable risk factors and to guide more comprehensive CVD risk management. The use of five- or 10-year absolute CVD risk calculators to appropriately target primary prevention has been incorporated into major CVD guidelines. However, applying risk algorithms to the management of an individual patient requires acceptance of the tools' limitations. These include suboptimal ancestry, geographic, and socioeconomic diversity in the derivation cohort; epidemiologic and practice pattern changes since equation development; and inconsistent event definitions used as outcomes to derive the equation, frequently the catastrophic clinical consequences of the disease rather than direct detection of CAD in the subclinical phase.

In 2000, the Multi-Ethnic Study of Atherosclerosis (MESA) offered a novel opportunity to understand the incidence, prevalence, and progression of coronary calcification; the influence of previously identified risk factors on this disease activity; and the impact of subclinical calcification on clinical outcomes in an asymptomatic cohort. In 2007, Kronmal, et al. demonstrated the association of eight risk factors (age, male sex, European ancestry, hypertension, body mass index, diabetes, fasting glucose, and family history of MI) with increased risk of incident coronary calcium and progression of prevalent calcium on follow-up coronary calcium scan (CCS).[3] From the same cohort, Detrano, et al. reported the clinical significance of a nonzero coronary artery calcium score (CACS), identifying an increased risk of myocardial infarction (MI) or CAD-related mortality by a factor of 3.89 (95% CI 1.72-8.79, $P < .001$) in participants with a CACS of 1 to 100 Agatston units (AU) compared to participants with a 0 AU CACS.[4] The investigators finally demonstrated value in including CACS in addition to traditional risk factors, with statistically significant area under the receiver-operating characteristic curve (AUC) increases for ability to predict MI or CAD-related mortality in Chinese and Black ethnicities and ability to predict any coronary event in White, Chinese, and Black participants. The ability of CACS to improve risk stratification and prediction of future clinical outcomes independent of traditional risk factors has been further reported in separate cohorts of both symptomatic and asymptomatic participants.[5,6] Though CACS is not itself a direct measure of higher-risk atherosclerotic plaque phenotypes, CACS has been correlated with coronary plaque volume.[7]

Despite the demonstrated ability of CACS to improve CAD risk stratification, population-level CAD screening with noncontrast computed tomography (CT) is neither feasible nor recommended. Modern CVD guidelines currently recommend CACS as a risk modifier with reclassification potential for cases where an individual's absolute risk is at a decisional threshold. CACS is currently not recommended for low-risk individuals, despite studies demonstrating the presence and strong prognostic value of coronary calcium in these populations,[8] in part due to a lack of prospective evidence on subsequent changes in management and outcomes.

Polygenic risk scoring

A heritable risk factor for CAD has long been suspected following early reports of familial clustering of premature

cases. However, CAD has not generally demonstrated monogenic inheritance patterns, apart from rare lipid metabolism disorders such as familial hypercholesterolaemia. Contemporary CAD genomic studies focus on common variants throughout the genome, each demonstrating small, continuous effects through the *common disease, common variants* hypothesis. High-throughput genome-wide association studies (GWAS) and improvements in linkage disequilibrium characterization have allowed millions of DNA markers to be interrogated. Allele frequencies for single-nucleotide polymorphisms (SNP) can be compared in case-control studies to identify variants associated with disease and their effect sizes, without surveillance bias to specific biological pathways or genes of interest.

The polygenic risk score (PRS) is a method quantifying an individual's genetic disease predisposition based on SNP:disease association and the SNP's effect size. Mathematically, the weighted PRS is a sum representing an individual's total genetic contributions proportional to the risk of disease. SNPs selected for the weighted PRS are traditionally independent and additive. The equation can be represented as follows, where β represents the effect size (obtained from GWAS summary statistics and expressed as log-odds), X_{ij} represents genotype for the i individual and j SNP.

$$PRS_i = \sum_{j=1}^{m} X_{ij}\beta_j$$

For nearly two decades, these evolving PRS algorithms have been studied with expanding evidence regarding their discriminatory capability for prevalent CAD cases, risk of developing incident CAD, and risk of CVD-events, including MACE. PRS are traditionally analyzed in a categorical framework, considering differences in relative risk between the highest decile or quintile and the lowest. Early studies demonstrated mixed performance, in part due to limitations in study design, sample sizes, numbers of events, and inclusion of non-CAD events (eg, stroke) in a composite endpoint. However, many recent PRS studies have now demonstrated strong independent association with hard CAD endpoints, along with some ability to improve risk prediction models independent of traditional risk factors.[9] Meta-analytic approaches have been applied to develop a "meta"-PRS consisting of 1.7 million genetic variants, showing those in the top 20% of PRS-predicted risk to have a 4.17-fold increased risk of MI events compared with those in the bottom 20%[10]. However, despite strong association data, there is a stark paucity of implementation studies testing this potentially powerful tool in prospective studies evaluating the feasibility, patient experience, impact on risk assessment and management, and the health economic potential.[11] The use in primary prevention is particularly attractive given the distinct lack of association ($r^2 < 0.004$) of the CAD PRS with the traditional Framingham Risk Score,[10] highlighting a potential synergy and opportunity for benefit for those in whom traditional scores perform poorly.

The *Implementation Study of Incorporating a Polygenic Risk Score into Cardiovascular Disease Examinations to Identify **SubClinicAL** coron**A**ry ar**TE**ry Disease* (ESCALATE) trial was designed and initiated to test the impact of incorporating a PRS-triaged CCS into the primary care setting on identification of subclinical CAD and resulting differences in medical management and participant experience.

Study objectives

The primary objective of the ESCALATE trial is to assess whether incorporating a PRS-triaged CCS into primary care CVD examinations identifies subclinical CAD in participants considered low or moderate risk by traditional risk algorithms. Secondary objectives are to measure the impact of this diagnostic strategy on: (1) traditional CVD risk factor management; (2) primary prevention pharmacotherapy utilization; (3) fasting cholesterol levels; (4) blood pressure measures; (5) patient-reported outcomes (PRO); and (6) participant reported experience. The health economic impact of the intervention will be evaluated, with comparisons to traditional Australian primary care CVD risk assessment.

Methods

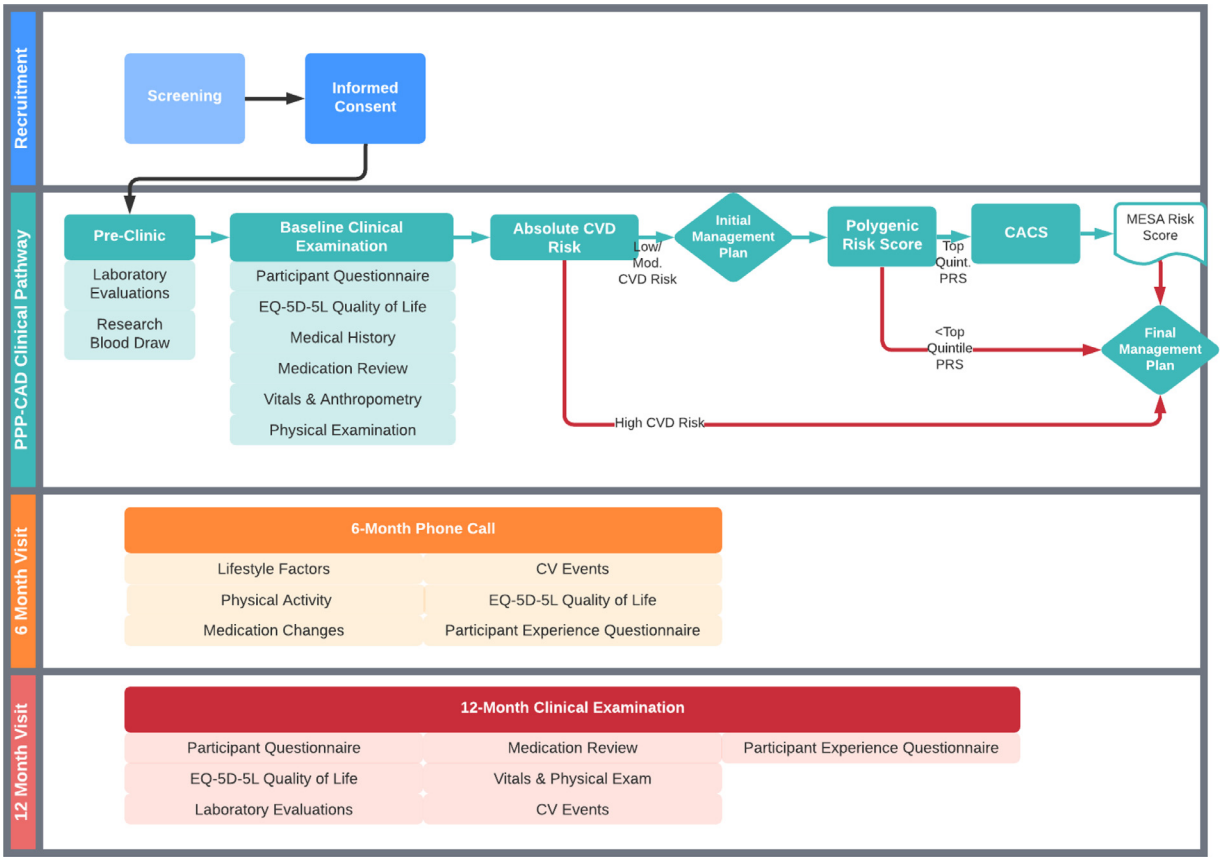
Study design

The ESCALATE study is a 12-month, prospective, multicentre, nonrandomized implementation study designed to demonstrate the impact of a PRS-triaged CCS in Australian primary care CVD risk assessments. This intervention seeks to identify patients with: a) low or moderate risk of CVD at 5-year by the Australian Absolute CVD Risk Calculator; and b) PRS ≥80% ("Top Quintile PRS") for noncontrast cardiac CT and CACS calculation. One-thousand participants are expected to undergo the intervention, with an estimated 20% being referred for CCS. The study diagram is presented in Figure 1 and full schematic in the Supplementary Materials. Participants will undergo a follow-up CVD risk assessment at 12-months. Participants will be invited to provide consent for further research use of the SNP array data used to calculate their PRS. Participants will finally be invited to provide consent for Medicare Benefits Schedule (MBS) and/or Pharmaceutical Benefits Scheme (PBS) administrative data linkage, facilitating pragmatic 5-year follow-up data during the study extension phase.

CAD polygenic risk score

A SNP array will be generated for each participant utilizing the Infinium GSA-24 v30 BeadChip. The ESCALATE trial uses several genetic ancestry-specific CAD PRS developed by Allelica, which are available from the company for research purposes. The CAD PRS that will be

Figure 1



ESCALATE study diagram.

applied to individuals of European genetic ancestry has been previously described in the literature.[12] Briefly, a stacked clumping and threshold (SCT) algorithm was applied to a UK Biobank training dataset, utilizing Nikpay, et al. GWAS summary statistics as input.[13] The SCT algorithm output was combined with SNPs identified by Inouye, et al.,[14] for a final metaPRS containing 1,926,521 SNPs (SCT-I PRS Panel).

Non-European PRS panels were developed to improve prediction accuracy over SCT-I PRS Panel using ancestry-specific GWAS datasets where possible[15,16] Ancestry-specific PRS panels for African, South and East Asian, and Admixed American ancestry individuals have been validated and tested utilizing the UK Biobank and MESA datasets (see Supplementary Materials).

Participants

The eligibility criteria have been pragmatically designed to enrol a population targeted for CVD primary prevention and for whom CACS would be clinically relevant.

1. Males aged 45 to 60 years old;
2. Females aged 50 to 65 years old;
3. Eligible for a Medicare (Australian government) rebated CVD consultation with a GP; and
4. Willingness to undergo the PRS and/or CCS interventions as indicated by the diagnostic clinical pathway.

Participants with conditions placing them at clinically determined high CVD risk will be ineligible for the study. In addition, participants with prior CVD events will also be ineligible for enrolment.

1. Conditions placing the individual at clinically determined high risk of CVD, where application of the Absolute CVD risk calculator is not appropriate:
   a. Diabetes and age >60 years
   b. Diabetes with microalbuminuria (>20 mcg/min or UACR >2.5 mg/mmol for males, >3.5 mg/mmol for females)

   c. Moderate or severe chronic kidney disease (CKD) (persistent proteinuria or eGFR <45 mL/min/1.73m^2)
   d. Previous diagnosis of familial hypercholesterolaemia
   e. Systolic blood pressure (SBP) ≥180 mm Hg or DBP ≥110 mm Hg
   f. Serum total cholesterol >7.5 mmol/L;
2. Symptomatic or previously documented CAD;
3. Previous CVD event, including angina, myocardial infarction (MI), percutaneous coronary intervention (PCI), coronary artery bypass grafting (CABG), ischaemic stroke, transient ischaemic attack, heart failure, or peripheral arterial disease;
4. Prior mediastinal radiation exposure or therapy; and
5. Prior medical diagnosis of rheumatoid arthritis (RA) or systemic lupus erythematosus (SLE).

Procedures

Screening and enrolment

Primary health networks will be informed of the trial and invited to inform member GP practices of the opportunity to participate as a clinical site. Patients at participating clinical sites who are eligible for a clinically indicated CVD clinical examination are introduced to the study by a member of the clinical team. Australians are eligible for an annual CVD risk assessment through the Heart Health Check MBS Item Number, or at the discretion of the GP. The study seeks to enrol equivalent numbers of males and females, across the entire age range, and an ethnically and socioeconomically diverse cohort. Following a comprehensive discussion of the study background, design, risks, benefits, and alternatives to participating, the participant is invited to provide written informed consent. The participant is then invited to provide independent consent for MBS and PBS five-year administrative data linkage; participants are not required to provide consent for the administrative data linkage extension phase to participate in the PRS intervention.

After providing informed consent, the participant is provided a referral for a research blood collection. Blood samples are processed utilizing standard fractionation and DNA isolation protocols, with plasma being stored to support future study-related biomarker analyses.

Following enrolment, participants complete three baseline questionnaires. The first questionnaire has been designed by the study investigators and collects demographics, lifestyle (eg, active/passive smoking and alcohol use), and nutrition data. Participants then complete the Global Physical Activity Questionnaire (GPAQ). Developed by the World Health Organization in 2002, the GPAQ is a validated physical activity tool assessing intensity, frequency, and duration of physical activity within work, day-to-day transportation, and recreational domains.[17] Finally, participants complete the 5-level EQ-5D (EQ-5D-5L) HRQOL tool.[18]

Intervention: PRS-triaged CCS

ESCALATE participants will complete a baseline clinical examination with their GP within thirty days of study enrolment. Clinical pathology will be collected prior to the baseline visit and is expected to include fasting cholesterol panel, glucose/HbA1c, high-sensitivity C-reactive protein, eGFR, and urine albumin/creatinine ratio. The baseline clinical examination includes a comprehensive review of past medical history, family history, anthropometrics, medications, clinical pathology results, 12-lead electrocardiogram (ECG), and in-office vitals. The GP utilizes information from the initial clinical visit to formally assess whether there are any modifiable risk factors to address and, if so, whether he or she intends to initiate pharmacotherapy; in addition, the GP will report LDL-C or BP targets if lipid- or BP-lowering therapies will be used. A study-specific form has been designed and will be used to record any management escalation during the study. Information from this baseline visit is used to calculate a five-year absolute CVD risk score using the Australian Absolute CVD Risk Calculator (Low-Risk = <10%; Moderate-Risk = 10%-15%; High-Risk = >15%). High-Risk participants do not have a PRS calculated and will be managed aggressively by the clinical team based on their elevated absolute risk. A CAD PRS is calculated for Low-Risk and Moderate-Risk participants. Participants with a PRS <80% ("Nontop Quintile PRS") do not continue in the diagnostic pathway; risk factors and any subsequent diagnostics are completed by the GP based on current expert consensus guidelines. Participants with Top Quintile PRS (≥80%) continue to the final step and receive a CCS referral. At the end of the diagnostic pathway, with all additional information obtained after the baseline visit, the GP confirms or modifies the medical management strategy to be used based on expert consensus treatment guidelines.

All participants complete two follow-up study visits. Six months after the baseline clinical visit, participants complete a questionnaire assessing lifestyle factors, physical activity, HRQOL, medication changes, and CVD events. Twelve months after the baseline clinical visit, participants complete a second clinical examination with the GP, including a follow-up assessment of lifestyle, physical activity, nutrition, HRQOL, laboratory evaluation, review of medication and diagnosis changes, clinical examination, and assessment of CVD events since enrolment. CVD event data will be used to informed healthcare utilization analyses.

Administrative data linkage

During the initial study enrolment, participants are invited to provide consent for five-year administrative data

linkage for Australian Government MBS and PBS payments, facilitating pragmatic five-year analyses of healthcare utilization and medication claim changes.

Outcomes

The primary and secondary outcomes will be analyzed from data collected during three timepoints: (1) the baseline visit (including all visits required to complete the diagnostic pathway); (2) six-month survey; (3) 12-month clinic examination.

Primary outcome

The primary outcome is the proportion of participants considered at low or moderate five-year absolute CVD risk, and a Top Quintile PRS, with subclinical CAD defined by CACS >0 AU. Non-zero CACS has been associated with events, including ACS and sudden cardiac death,[19] and additionally been demonstrated to be an independent predictor of mortality in observational studies.[20]

Secondary outcomes

The secondary and exploratory outcomes are included in Table I.

- *Clinically significant CAD:* The proportion of participants at low or moderate five-year absolute CVD risk, and Top Quintile PRS, with a CACS ≥100 AU or ≥75th age/sex percentile CACS.
- *MESA Risk Calculator:* The proportion of participants completing the PPP-CAD Diagnostic Pathway who experience an upward reclassification of MESA Risk Score after incorporation of CACS into the equation. This risk equation is used to predict 10-year CAD-specific events utilizing traditional risk factors ± CAC measurement. CAC inclusion in the equation resulted in significant risk prediction improvements (C-statistic 0.80 v. 0.75, $P < .001$).[21]
- *Lipid- and Blood Pressure-Lowering Therapy:* The proportion of participants on lipid- and blood pressure-lowering medications at 12 months. Proportions of participants on these medications at five years will be analyzed and reported from administrative data linkage.
- *Fasting Cholesterol Changes:* The change in fasting total cholesterol, triglycerides, HDL-C, and LDL-C levels between baseline and 12 months.
- *Blood Pressure Changes:* The change in systolic and diastolic blood pressure between baseline and 12 months.
- *HRQOL:* The change in EQ-5D-5L score between baseline and 12-months.
- *Participant Experience*: Participants are surveyed regarding their experience with the intervention and utilizing a genetic test for CAD diagnosis at two timepoints: (1) at the end of the baseline visit(s); and (2) after the 12-month clinic evaluation. A self-reporting tool has been designed by the investigators, based on the Australian Hospital Patient Experience Question Set,[22] asking participants to rate their agreement with eight statements on a Likert scale (5 = Strongly agree; 1 = Strongly disagree). Statements include the participants' perceptions that their views and concerns about screening were listened to, they are confident in their screening, they understood the screening process, they felt they were involved in their treatment and care, they were confident in the treatment and care, whether they experienced any worry as a result of their CAD screening, and whether those worries were appropriately addressed.

Healthcare utilization

Participants will be asked to report on their recent healthcare utilization during the six- and 12-month follow-up visits. Expenditures related to their consultations will be recorded, including direct costs of obtaining care, indirect costs related to travel and incidentals, and opportunity costs related to time off work will be recorded. Service utilization will be classified as related or unrelated to CAD and the study intervention, to capture the direct system impact of the pathway and the indirect physical or psychological consequences. This will complement data obtained from the MBS/PBS administrative data linkage.

Statistical methods

Baseline clinical examination will be summarized using descriptive statistics. The primary and secondary outcomes will be reported using descriptive statistics. To assess changes in fasting cholesterol and blood pressure between baseline and 12-months, linear mixed effects models will be used. Baseline participant characteristics—including age, sex, race, BMI, smoking status, and the outcome values at baseline—will be adjusted for in the models. Changes to risk factor management strategies and therapeutic targets during the intervention will be reported using descriptive statistics. Participant experience will be analyzed as proportion of participants who agree with the statements in each of the seven domains.

Sample size and power

The sample size has been determined based on the primary objective to examine whether incorporation of PRS into a cardiovascular disease examination is feasible and identifies individuals with unexpected atherosclerosis, providing an opportunity for more aggressive risk factor management.

One thousand individuals with traditional risk scores in the low and intermediate category will be recruited

Table I. ESCALATE study outcome measures.

PRIMARY OUTCOME
Proportion of participants considered to be low or moderate Absolute CVD Risk and in the Top Quintile PRS Risk with subclinical CAD—defined as CACS >0 Agatston units—identified using the CAD diagnostic pathway
SECONDARY OUTCOMES
Proportion of participants considered to be low or moderate Absolute CVD Risk, and in Top Quintile PRS Risk, with a CACS ≥100 AU or ≥75th age/sex/race percentile CACS identified utilizing the CAD diagnostic pathway. Proportion of participants considered to be low or moderate Absolute CVD Risk, and in the Top Quintile PRS Risk with an upward reclassification of 10-y MESA Risk Score after incorporation of CACS Between group difference in proportion of participants prescribed lipid-lowering therapy at 12-mo, comparing participants managed based on risk factor + CAC to participants managed by risk factors alone Change in cholesterol levels between baseline and 12-mo, comparing participants managed based on risk factor + CAC to participants managed by risk factors alone • Total >cholesterol • Triglycerides • HDL cholesterol • LDL cholesterol Between group difference in the proportion of participants prescribed blood pressure-lowering therapy at 12-mo, comparing participants managed based on risk factor + CAC to participants managed by risk factors alone Change in blood pressure between baseline and 12-mo, comparing participants managed based on risk factor + CAC to participants managed by risk factors alone • SBP • DBP Between group difference in change in 5-level EQ-5D (EQ-5D-5L) score from baseline to 12-mo, comparing participants managed based on risk factor + CAC to participants managed by risk factors alone Between group difference in proportion of participants prescribed lipid-lowering therapy at 5-y, comparing participants managed based on risk factor + CAC to participants managed by risk factors alone Between group difference in proportion of participants prescribed blood-pressure lowering therapy at 5-y, comparing participants managed based on risk factor + CAC to participants managed by risk factors alone
EXPLORATORY OUTCOMES
Sex differences in proportion of participants considered to be low or moderate Absolute CVD Risk, and in the Top Quintile PRS Risk, with subclinical CAD—defined as: (1) CACS >0 AU; or (2) CACS ≥100 AU or ≥75th age/sex/race percentile CACS—identified using the CAD diagnostic pathway

which, by definition, is expected to result in 200 individuals in Top Quintile PRS Risk. Conservatively, it is estimated that 50% of these individuals will have a positive CAC.[21,23] This is expected to result in an estimated 100 individuals who would have been "missed" and who will be prescribed precision primary prevention therapy. The ESCALATE trial has 81.13% power to detect a 10 percent difference in proportion of Top Quintile PRS participants with positive coronary calcium, assuming a general population prevalence of 45% in this age group.[21]

Study and data management

The study is coordinated at the Kolling Institute of Medical Research at the University of Sydney. Study data are either completed by the participant through approved study surveys or collected by members of the research team on case report forms (CRF). Data will be managed electronically during the collection period by the coordinating center utilizing a REDCap database, hosted at the University of Sydney. ECGs, noncontrast CT scans, and SNP arrays will be collected per standard processes.

Long-term data storage of de-identified research datasets—including clinical, imaging, genomic, and molecular data—will occur on research servers, hosted by the University of Sydney. All informed consent documents will be securely transferred from the REDCap data collection tool to research servers managed by the University of Sydney Archives and Records Management Services. All data and source documents will be stored in accordance with research guidelines for a minimum of 15 years after data analysis.

As the ESCALATE study is a nonrandomized implementation study with a low-risk intervention, no interim analysis or data monitoring committee are planned.

The coordinating center will provide data management services and ensure that personally identifiable information is removed from study data exports, including all analysis and archive datasets. Identifiable participant data will be restricted and provided on a need-to-know basis only to appropriate study members, including principal

investigators, senior coordinating center staff, and ethics committee representatives.

A 20-member Steering Committee (SC) has been constituted, representing academic/clinical cardiology, primary care, and radiology; clinical genomics; clinical trial design; public health; health policy; health economics; and relevant members of industry. The SC is broadly responsible for determining overall strategic direction, identifying and progressing novel funding opportunities, and disseminating results for use in further interventional studies or health policy discussions. A seven-member Trial Management Committee (TMC), not including members from industry, is solely responsible for the design and oversight of the ESCALATE trial, approval of resulting abstracts and publications, and approval of requests for data. The TMC is responsible for the monitoring of participant safety in the absence of a data monitoring committee.

The study is an investigator-initiated study supported by an NHMRC Partnerships award. No additional, extramural funding will be used to conduct the trial. Per funding guidelines, partner organizations must provide at least half the research budget for funded trials through cash and/or in-kind support. No cash support for the ESCALATE trial design and execution has been received from for-profit, commercial interests. 23Strands has committed to providing financial support for a nonposttrial investigators' meeting. Allelica SRL (Italy), Castlereagh Imaging, North Shore Radiology & Nuclear Medicine, and Sonic Healthcare have provided in-kind support through reduced rates for trial services.

Results dissemination

The ESCALATE study anticipates disseminating trial results through presentations at cardiovascular and clinical genomic society congresses. In addition, study results are planned to be published—whether positive, negative, or inconclusive—in peer-reviewed journals and reports. Study data may be used in research theses. Importantly, all participants are asked during the informed consent process if they would like to receive updates prepared by the study team on presentations and publications arising from the trial.

Study ethics and governance

The study will be conducted according to the Declaration of Helsinki. The study has received ethical approval from the University of Sydney Human Research Ethics Committee (2021/913). The Australian Institute of Health and Welfare External Request Evaluation Committee approved the prospective consent collection mechanism for MBS/PBS data linkage. The University of Sydney serves as the study sponsor. The trial was prospectively registered on the Australian New Zealand Clinical Trials Registry (ANZCTR) on March 18, 2022 (Trial Number: ACTRN12622000436774).

Discussion

This prospective, multicentre study tests the use of PRS to identify patients for noninvasive coronary imaging who are at increased lifetime CAD risk, but considered to be low or moderate risk by traditional risk algorithms, to detect subclinical CAD and inform shared decision-making. Whilst traditional risk scores perform well at a population level, many individuals develop CAD and MI despite no standard, modifiable cardiovascular risk factors.[2] There is a significant unmet need for new diagnostic strategies independent of these factors to detect CAD in its subclinical phase. Initial results from on-going, population-level clinical trials—including Danish Cardiovascular Screening (DANCAVAS)—have demonstrated difficulty in applying untargeted CVD screening in a positive impact on mortality.[24] However, studies such as MESA and the Swedish Cardiopulmonary Bioimage Study (SCAPIS) continue to show significant silent coronary disease at the population-level. Together, these exhibit the continued unmet need to identify and prospectively trial implementation of novel risk markers for more aggressive diagnostic workup in targeted populations. Given the relative independence of the risk represented by the PRS and CAD traditional risk factors, and the prognostic significance of CACS across traditional risk score strata, our approach has the potential to test further CAD screening in individuals at higher risk of atherosclerotic CAD based on observational cohort studies.

It is important to note that whilst a CT CCS and the resulting CACS is an excellent noninvasive method for detecting subclinical atherosclerosis, it is unable to detect noncalcified plaque. Furthermore, it is not intended to be diagnostic for functionally significant stenosis. Given the population recruited for ESCALATE is asymptomatic, this is unlikely to be of relevance. However, the presence of calcified plaque and elevated CACS may prompt more thorough history and either CT coronary angiography or functional study (eg, stress echocardiography) where indicated.

To the authors' knowledge, this is the first prospectively registered clinical trial directly testing both: (1) the ability of a CAD PRS to identify individuals with subclinical CAD detected by triaged CCS referral; and (2) the impact of the knowledge gained during this diagnostic pathway on shared decision-making and participant experience. Interventions incorporating PRS into CAD diagnostic strategies have previously been tested in several related clinical trials, with distinct differences to the ESCALATE study (see Supplementary Materials). For example, the EstPerMedCV (Trial #NCT04291157), GenoVA (Trial #NCT04331535), HEART (Trial #NCT05294419), PEPRS1 (Trial #NCT05072275), and PEPRS2 (Trial

#NCT05175651) studies seek to characterize the impact of PRS on CAD diagnosis in the primary care setting on endpoints including time to new diagnosis, impact on clinical decision making, participant/provider experience, and MACE. However, none use the innovative triage approach we have adopted in the ESCALATE trial to select an enriched population at higher risk of disease, which when visualized by CACS can drive shared decision making on treatment. Recently, in a pilot implementation study utilizing PRS to recall individuals at differential CAD genetic risk for CACS screening, investigators at the Icahn School of Medicine at Mount Sinai demonstrated a 2.8-fold increase in CACS in participants with high PRS compared to low (95% CI 1.2-6.9, $P = .027$).[25] However, this study was limited in study setting, sample size, cohort diversity, and exclusion of individuals with nonextreme CAD PRS.

Whilst the ESCALATE study will contribute to identified literature gaps, the authors note the following limitations due to study design. Firstly, ESCALATE utilizes a nonrandomized design to test the PRS-triaged CCS. The investigators will report the proportion of individuals at low or moderate five-year absolute CVD risk and Top Quintile PRS with subclinical CAD. Insights will be described on risk factor management strategies utilized during the diagnostic pathway and during follow-up. However, it will not be possible to compare this strategy to standard-of-care. Importantly, however, participant experience will be reported, allowing the study design and patient communication tactics to be refined prior to testing under randomized conditions, minimising further structural opportunities for bias including selection bias. Secondly, as participants will be providing informed consent for this novel diagnostic strategy, study generalizability could be impacted by engaging a cohort of individuals with healthy seeking behaviours; this will be explored and described through analysis of self-reported participant data (eg, physical activity, nutrition). Finally, CACS will not systematically be collected in non-Top Quintile PRS participants. Therefore, the investigators will be unable to calculate differences in subclinical CAD between PRS strata, allowing PRS referral thresholds to be refined for future research studies.

Several limitations exist within the use of PRS in any research setting, which warrant consideration during study planning and interpretation processes. A fundamental limitation of PRS is its development from cohort studies that have historically included mostly European participants. The predictive ability of PRS has generally attenuated in validation studies of non-European populations. GWAS and PRS development in ancestrally diverse and admixed cohorts will be critical to future clinical use of PRS and improve generalizability of the strategy. In addition, though PRS has demonstrated predictive ability for CAD, the biological mechanism and interaction with standard risk factors remain largely unknown. Variants may have pleiotropic or differential effects depending on the specific environmental, biochemical, and genetic milieu of an individual, which are not accurately reflected in the GWAS summary statistics used in PRS development.

Further perceived limitations in the medical community discouraging implementation of this potentially powerful tool stem from a lack of understanding of how the PRS operates, as well as the rapidly evolving algorithms. Our pragmatic approach, incorporating a binary PRS cut-off into a diagnostic decision tree, provides a pathway utilizing the potentially powerful prognostic ability of the enigmatic PRS, but allowing clinical decision-making to be based on plaque burden itself. This approach additionally establishes a framework for easier application of evolving future PRS, including a model to support patient understanding to reduce the need for genetic counselling.

Despite these potential limitations, the ESCALATE trial represents a unique opportunity to begin systematically testing a novel diagnostic strategy to identify subclinical CAD. Initial data will inform future study design, including intervention testing under randomized conditions and against hard CAD endpoints, including MACE. Patients with subclinical CAD identified through this pathway will have the opportunity for earlier shared decision-making with their primary care physician on the initiation or escalation of pharmacotherapy associated with survival benefits.

Conclusion

The ESCALATE study is a prospective, multicentre, nonrandomized implementation study of a PRS to identify additional patients in the primary care setting for noninvasive coronary imaging, to identify subclinical CAD. The ESCALATE study aims to generate evidence on the impact of the PPP-CAD Diagnostic Pathway on identification of subclinical CAD and whether this diagnostic pathway results in differences in CAD risk factor management, HRQOL, and participant reported experience. The initial study results are expected in mid-2025.

Authors' contribution

GAF conceived of the study. *GAF, MPG, AH, GJ, GK, PM, JR, TU,* and *SJN* initiated the study design and *YB, JH,* and *JFL* helped with implementation. *KH* provided statistical expertise in the study design. *MPG and GAF* wrote the original draft of the manuscript. *YB, SMG, AH, JH, KH, GJ, JFL, JR, TU, and SJN* reviewed and edited the manuscript. All authors reviewed and approved the manuscript prior to submission.

Trial status

The ESCALATE study initiated recruitment on August 8, 2022 and, as of July 4, 2023, 366 participants have been enrolled in the trial. The study is in the recruitment phase, which is expected to conclude by May 2024.

Public availability of data

A decision on the public availability of trial data has not yet been made by the Trial Management Committee (TMC).

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Disclosure

G. Bottà is an employee and holds equity in Allelica Srl.

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Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.ahj.2023.06.009.

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