Vitamin D supplementation and major cardiovascular events: D-Health randomised controlled trial

Bridie Thompson,[1] Mary Waterhouse,[1] Dallas R English,[2] Donald S McLeod,[1]
Bruce K Armstrong,[3] Catherine Baxter,[1] Briony Duarte Romero,[1] Peter R Ebeling,[4]
Gunter Hartel,[5] Michael G Kimlin,[6] Sabbir T Rahman,[1] Jolieke C van der Pols,[7] Alison J Venn,[8]
Penelope M Webb,[1] David C Whiteman,[1] Rachel E Neale[1]

ABSTRACT

OBJECTIVE
To investigate whether supplementing older adults with monthly doses of vitamin D alters the incidence of major cardiovascular events.

DESIGN
Randomised, double blind, placebo controlled trial of monthly vitamin D (the D-Health Trial). Computer generated permuted block randomisation was used to allocate treatments.

SETTING
Australia from 2014 to 2020.

PARTICIPANTS
21 315 participants aged 60-84 years at enrolment. Exclusion criteria were self-reported hypercalcaemia, hyperparathyroidism, kidney stones, osteomalacia, sarcoidosis, taking >500 IU/day supplemental vitamin D, or unable to give consent because of language or cognitive impairment.

INTERVENTION
60 000 IU/month vitamin D_3 (n=10 662) or placebo (n=10 653) taken orally for up to five years. 16 882 participants completed the intervention period: placebo 8270 (77.6%); vitamin D 8552 (80.2%).

MAIN OUTCOME MEASURES
The main outcome for this analysis was the occurrence of a major cardiovascular event, including myocardial infarction, stroke, and coronary revascularisation, determined through linkage with administrative datasets. Each event was analysed separately as secondary outcomes. Flexible parametric survival models were used to estimate hazard ratios and 95% confidence intervals.

RESULTS
21 302 people were included in the analysis. The median intervention period was five years. 1336 participants experienced a major cardiovascular event (placebo 699 (6.6%); vitamin D 637 (6.0%)). The rate of major cardiovascular events was lower in the vitamin D group than in the placebo group (hazard ratio 0.91, 95% confidence interval 0.81 to 1.01), especially among those who were taking cardiovascular drugs at baseline (0.84, 0.74 to 0.97; P for interaction=0.12), although the P value for interaction was not significant (<0.05). Overall, the difference in standardised cause specific cumulative incidence at five years was −5.8 events per 1000 participants (95% confidence interval −12.2 to 0.5 per 1000 participants), resulting in a number needed to treat to avoid one major cardiovascular event of 172. The rate of myocardial infarction (hazard ratio 0.81, 95% confidence interval 0.67 to 0.98) and coronary revascularisation (0.89, 0.78 to 1.01) was lower in the vitamin D group, but there was no difference in the rate of stroke (0.99, 0.80 to 1.23).

CONCLUSIONS
Vitamin D supplementation might reduce the incidence of major cardiovascular events, although the absolute risk difference was small and the confidence interval was consistent with a null finding. These findings could prompt further evaluation of the role of vitamin D supplementation, particularly in people taking drugs for prevention or treatment of cardiovascular disease.

TRIAL REGISTRATION
ACTRN12613000743763

WHAT IS ALREADY KNOWN ON THIS TOPIC

Observational studies have consistently shown inverse associations between 25-hydroxyvitamin D concentration and cardiovascular disease

Randomised controlled trials have not shown that vitamin D supplementation reduces the incidence of major cardiovascular events, although most trials were not adequately powered to investigate this issue

WHAT THIS STUDY ADDS

Vitamin D supplementation might reduce the risk of major cardiovascular events, although the absolute risk difference was small and the confidence interval was consistent with a null finding

Further evaluation is warranted, particularly in people taking statins or other cardiovascular disease drugs

Introduction

Coronary heart disease and stroke are the leading causes of death globally.[1] The risk of these events increases with age, and they are more prevalent in men than women.[2] The number of cardiovascular disease events will probably continue to increase in developed countries as populations age, and in low to middle income countries as non-communicable diseases become dominant.[3] Vitamin D has biological effects which suggest it could influence cardiovascular disease. The vitamin D receptor is expressed in cells throughout the vascular system; many of these also express 1α-hydroxylase, and are therefore able to convert 25-hydroxyvitamin D (25(OH)D) to calcitriol, the active form of vitamin D. Calcitriol reduces inflammation, regulates the renin-angiotensin-aldosterone system, and inhibits proliferation of vascular smooth muscle.[4]

Meta-analyses of observational studies have found inverse associations between serum 25(OH)D concentration and risk of cardiovascular disease.[5-9] However, these findings might be due to reverse causality or uncontrolled confounding. Of three Mendelian randomisation studies, which largely overcome these biases, one reported an inverse association between genetically predicted 25(OH)D concentration up to 50 nmol/L and cardiovascular disease.[10] The other studies found no association, but did not allow for nonlinear effects.[11 12] A meta-analysis of randomised controlled trials concluded that vitamin D supplementation does not prevent cardiovascular events.[13] However, 45% of the 83 291 participants included in the meta-analysis were from the Women's Health Initiative Trial, which was restricted to women, used a low dose of vitamin D, and had relatively low compliance.[14] Cardiovascular disease was the primary outcome of the Vitamin D Assessment (ViDA) study[15] and the Vitamin D and Omega 3 trial (VITAL).[16] Despite different outcome definitions, both randomised controlled trials found that vitamin D supplementation had no effect on cardiovascular disease,[15 16] but VITAL excluded people with a history of cardiovascular disease and the ViDA study had relatively few events.

We launched the D-Health Trial to determine if monthly vitamin D supplementation can improve health outcomes in the older general population. It was a large intermittent dosing trial of vitamin D supplementation (n=21 315). Previous analysis of the D-Health cohort found that vitamin D supplementation did not reduce all cause mortality (the primary outcome of the overall trial) or mortality due to cardiovascular disease,[17] but the effect on the incidence of major cardiovascular events has not been analysed.

For the current study we analysed data from the D-Health Trial to examine whether supplementing Australians aged ≥60 years with monthly doses of 60 000 IU of vitamin D altered the incidence of major cardiovascular events.

Methods

Study design, recruitment, and participants

The D-Health Trial was a randomised, double blind, placebo controlled trial with two parallel arms.[18] Between January 2014 and May 2015, randomly selected adults, aged 60-79 years, were invited from all Australian states and territories (except the Northern Territory) using a population register, the Commonwealth Electoral Roll, as the sampling frame (in Australia it is compulsory to register to vote). Volunteers aged 60-84 years were also recruited via media stories and contacts of participants. Exclusion criteria included a history of hypercalcaemia, hyperparathyroidism, kidney stones, osteomalacia, sarcoidosis, or daily intake of >500 IU of supplemental vitamin D. The full trial protocol is available online (https://dhealth. qimrberghofer.edu.au/page/Publications/).

Randomisation and blinding

We used computer generated permuted block randomisation, stratified by age, sex, and state of residence, to randomly allocate participants in a 1:1 ratio to 60 000 IU of vitamin D_3 (cholecalciferol) or placebo tablets, taken as monthly oral doses. Vitamin D_3 and placebo tablets were identical in appearance. Participants, staff, and investigators were blinded to study group allocation during the intervention. Participants were notified of their allocation in March 2020. Staff and investigators remained blinded until the analyses of all cause mortality[17] were finalised. We wrote the statistical code for the current analysis blind to study group using a dataset from which the allocation variable had been removed and participants were randomly assigned to two groups of equal size. After the statistical code for prespecified analyses was finalised, we implemented it on the original dataset.

Intervention

Each year, participants were sent 12 study tablets. We reminded participants to take one tablet at the beginning of each month through text message, email, or automated landline message. The intervention period ended at five years after randomisation, or on 1 February 2020 for the 507 participants randomised after February 2015.

Baseline information

Participants completed a baseline questionnaire in which they reported sociodemographic and lifestyle factors, pre-existing health conditions, and intake of food and supplements containing vitamin D. We calculated body mass index by dividing self-reported weight (kg) by height squared (m^2). Serum 25(OH)D concentration was not measured at baseline; rather we developed and internally validated a model to predict deseasonalised baseline serum 25(OH)D concentration using data and serum 25(OH)D measures collected from a random subset of participants in the placebo group during the trial.[19]

Adherence and adverse event reporting

Annually, participants were asked to report the number of study tablets taken and their use of any other supplements containing vitamin D not related to the study. We calculated adherence by dividing the number of tablets taken by the number they would have taken had they been fully adherent (60, except for those who died during the trial). We encouraged participants to minimise the use of off-trial vitamin D supplements, but allowed them to remain in the trial provided they took no more than 2000 IU/day. This strategy ensured participants remained below the tolerable upper intake level of 4000 IU, enabled us to capture information about off-trial vitamin D intake, and minimised missing participant reported outcome information.

Each year, we randomly selected approximately 800 participants (stratified by study group, age, sex, state, and month of recruitment) and asked them to provide

blood samples for measurement of serum 25(OH)D concentration.

Participants were asked to contact the trial helpline if they experienced any health events; these were coded using the Medical Dictionary for Regulatory Activities. Diagnoses of kidney stones, hypercalcaemia, and hyperthyroidism were also captured in annual surveys.

Determination of major cardiovascular events

Cardiovascular events were a prespecified tertiary outcome of the D-Health Trial. Our published statistical analysis plan included 45 tertiary outcomes.[20] In the statistical analysis plan for the current study, we prespecified that the main outcome for this analysis was first major cardiovascular event, defined as any of myocardial infarction, stroke, or coronary revascularisation. We prespecified the first

of myocardial infarction, stroke (total, ischaemic, and haemorrhagic), and coronary revascularisation separately as secondary outcomes.

We used linked hospital admissions data, Medicare Benefits Schedule records, and mortality data to determine major cardiovascular events. Medicare is Australia's universal health insurance system, and procedures that take place outside public hospitals are recorded in the Medicare Benefits Schedule dataset. Hospital admissions data were available from each state, but not from the Northern Territory or Australian Capital Territory. Admissions to private hospitals were not available from Tasmania or South Australia. Supplementary table 1 shows the principal diagnosis codes (international classification of diseases 10th revision), procedure codes, and Medicare Benefits Schedule item numbers used to determine events. If a death from myocardial infarction or stroke occurred, with no previous hospital admission for these conditions or for coronary revascularisation, the date of major cardiovascular event was considered to be the date of death. Determination of cause of death is described elsewhere.[17]

Use of cardiovascular drugs at baseline

The Pharmaceutical Benefits Scheme captures information about prescription drugs dispensed to Australian citizens and permanent residents. We used linked Pharmaceutical Benefits Scheme data to determine use of statins (Anatomical Therapeutic Chemical code C10) and other cardiovascular drugs (codes C01-C09). Use within three months after randomisation indicated baseline use. For people who did not consent to linkage with the Pharmaceutical Benefits Scheme (n=1812), we used self-reported treatment at baseline for hypercholesterolaemia and hypertension; agreement between self-reported drug use and Pharmaceutical Benefits Scheme data was high (supplementary tables 2a and 2b).

Sample size and power

The sample size for the D-Health Trial was chosen to enable 80% power to detect a difference of 9% in the mortality rate with a type 1 error rate of 0.05.[18] We estimated that given the sample size available for this analysis (n=21 302) we would have 80% power to detect a difference of 16% in the incidence of first major cardiovascular event (based on VITAL data[16] from which we estimated that 508 events would be expected in the placebo group). This effect size calculation was performed before starting analyses (before knowing the actual number of events).

Statistical analysis

Analyses followed the intention-to-treat principle and were conducted in SAS version 9.4 (SAS Institute, Cary, North Carolina, USA), R version 4.1.1 (R Foundation for Statistical Computing, Vienna, Austria), and Stata version 17 (Stata Corp, Texas, USA). The D-Health Trial statistical analysis plan has been published previously.[20]
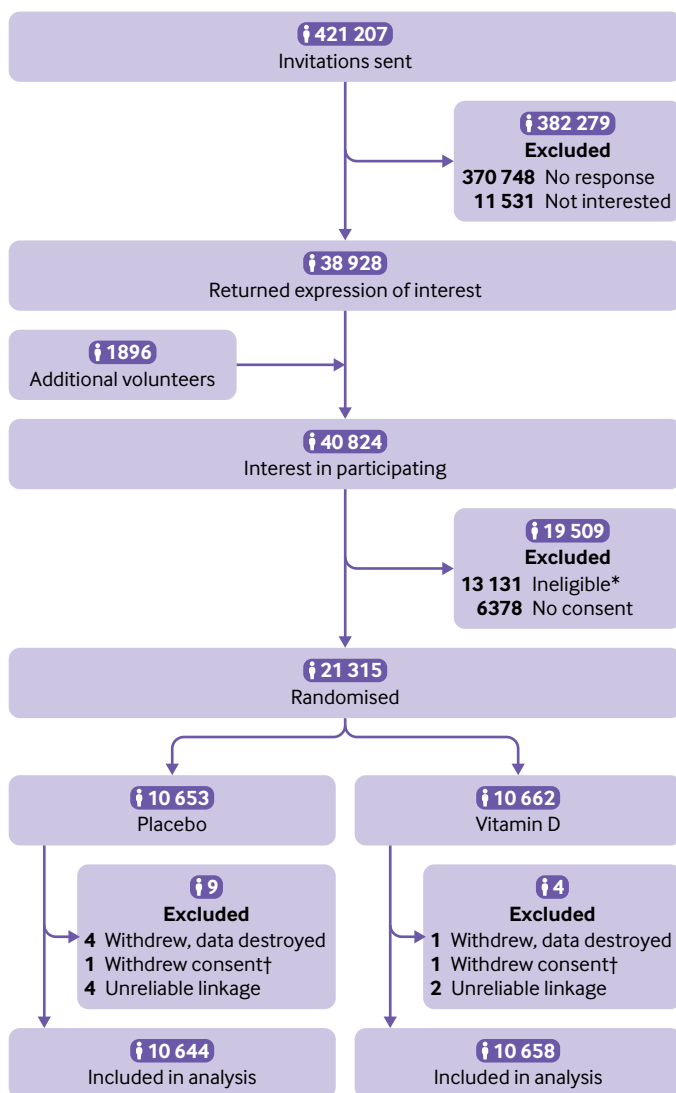

Fig 1 | Participant flow for analyses of major cardiovascular events (Consolidated Standards of Reporting Trials—CONSORT flow diagram). *People with self-reported history of hypercalcaemia, kidney stones, hyperarathyroidism, osteomalacia, or sarcoidosis, or those taking ›500 IU/day of supplemental vitamin D were ineligible. †Withdrew consent to link to health registers

Table 1 | Baseline characteristics according to randomisation group

Characteristic	Vitamin D (n=10 658)	Placebo (n=10 644)
Age (years)		
60-64	2627 (24.6)	2624 (24.7)
65-69	2920 (27.4)	2914 (27.4)
70-74	2901 (27.2)	2891 (27.2)
≥75	2210 (20.7)	2215 (20.8)
Sex		
Men	5765 (54.1)	5760 (54.1)
Women	4893 (45.9)	4884 (45.9)
Body mass index		
<25	3257 (30.7)	3157 (29.8)
≥25	7344 (69.3)	7425 (70.2)
Missing	57	62
Predicted 25(OH)D concentration (nmol/L)		
<50	2562 (24.0)	2637 (24.8)
≥50	8096 (76.0)	8007 (75.2)
Statin use at baseline*		
No	6880 (64.6)	6956 (65.4)
Yes	3769 (35.4)	3681 (34.6)
Missing	9	7
Cardiovascular drug use at baseline†		
No	5655 (53.1)	5755 (54.1)
Yes	4994 (46.9)	4882 (45.9)
Missing	9	7
Self-reported history of hypertension		
No	6129 (57.8)	6240 (58.8)
Yes	4483 (42.2)	4368 (41.2)
Missing	46	36
Self-reported history of hypercholesterolaemia		
No	7164 (67.5)	7192 (67.8)
Yes	3445 (32.5)	3412 (32.2)
Missing	49	40
Self-reported history of other cardiovascular disease‡		
No	8233 (77.7)	8220 (77.6)
Yes	2357 (22.3)	2366 (22.4)
Missing	68	58

Data are numbers (%).

*Anatomical Therapeutic Chemical code C10 in Pharmaceutical Benefits Scheme data within three months of randomisation, or self-reported treatment for hypercholesterolaemia before randomisation.

†Anatomical Therapeutic Chemical codes C01-C09 in Pharmaceutical Benefits Scheme data within three months of randomisation, or self-reported treatment for hypertension before randomisation.

‡A history of cardiovascular disease was assumed if a participant reported having experienced or received a diagnosis of the following before randomisation: heart attack, stroke, transient ischaemic attack, angioplasty, pacemaker insertion, insertion of a coronary stent, coronary artery bypass graft, thrombosis, angina, or arrhythmia.

The detailed plan for this analysis is available at https://dhealth.qimrberghofer.edu.au/page/Publications/. Although this is one of several outcomes analysed, we have not adjusted for multiple testing.

For each outcome, follow-up began at randomisation and ended at the earliest of first major cardiovascular event of interest; last known date alive; five years and one month after randomisation; or 31 December 2019 (the date to which hospital data were provided for all states). We used Aalen-Johansen methods to plot the cause specific cumulative incidence of an outcome according to randomisation group. We used flexible parametric survival models to estimate the effect of vitamin D supplementation on outcomes. To estimate an overall hazard ratio, we used a flexible parametric survival model without any time varying coefficients. To allow the hazard ratio to vary with time, we fitted a second flexible parametric survival model that included an interaction between randomisation group and time since randomisation. We used flexible parametric survival models to estimate the difference in cause

specific standardised cumulative incidence, treating death without previous major cardiovascular event as a competing risk. All flexible parametric survival models included the randomisation stratification variables of age, sex, and state of residence at baseline. Additional details of the flexible parametric survival models are included in the supplementary methods.

We assessed whether the effect of vitamin D supplementation on major cardiovascular events was modified by the following prespecified baseline characteristics: age (<70, ≥70 years); sex (men, women); body mass index (<25, ≥25); predicted deseasonalised serum 25(OH)D concentration (<50 nmol/L, ≥50 nmol/L)[19]; statin use; and use of (non-statin) cardiovascular drugs. Participants missing data for a stratifying variable were excluded from the relevant analysis.

In prespecified sensitivity analyses, we excluded people who did not have Pharmaceutical Benefits Scheme data from the subgroup analyses by statin use and cardiovascular drug use (rather than using self-reported use of these drugs). We have not shown the data because this exclusion made no meaningful difference to the results.

In exploratory analyses (performed after the prespecified results were available) we estimated the overall effect of vitamin D supplementation on total major cardiovascular events within subgroups defined according to use of statins and non-statin cardiovascular drugs versus no use of these drugs (as opposed to use or non-use of each of these drug classes separately); and myocardial infarction and coronary revascularisation within stratums defined according to use of statins and cardiovascular drugs at baseline.

Patient and public involvement

The public were not involved in the design or reporting of this study. A participant committee contributed to the design of the participant information sheet, surveys, and newsletters.

Results

Between January 2014 and May 2015 we invited 421 207 people to participate in the D-Health Trial. From 38 928 people who expressed interest and an additional 1896 volunteers, we recruited 21 315 eligible people (fig 1). Five participants subsequently requested that their data be destroyed and eight had incomplete hospital data, leaving 21 302 in this analysis (vitamin D, n=10 658; placebo, n=10 644).

For the trial overall, 16 822 (79%) participants (vitamin D, n=8552 (80%); placebo, n=8270 (78%)) were still taking tablets at the end of five years; 866 people died before they completed the intervention period. The median treatment duration was five years and more than 80% of participants reported taking at least 80% of the study tablets (vitamin D, n=9006 (84%); placebo, n=8783 (82%)). During the intervention, the mean serum 25(OH)D concentration was 77 nmol/L (standard deviation 25) in the placebo group and 115 nmol/L (standard deviation 30) in the
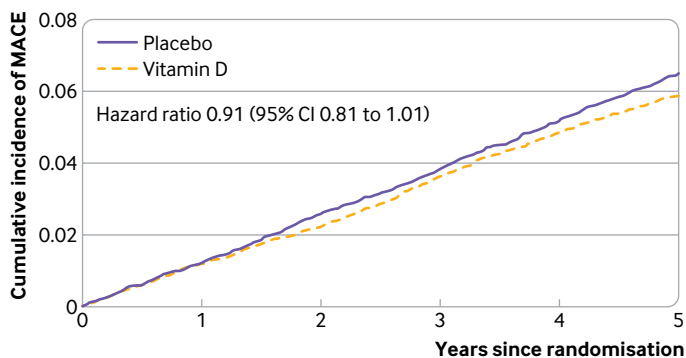
Fig 2 | Cause specific cumulative incidence of major cardiovascular events according to randomisation group and time since randomisation. Curves estimated using Aalen-Johansen methods, treating death without previous major cardiovascular event as a competing risk. Hazard ratio (vitamin D v placebo) was estimated using a flexible parametric survival model that included randomisation group, age, sex, and state of residence at baseline. 95% CI=95% confidence interval; MACE=major cardiovascular event

vitamin D group. The incidence of adverse events was similar in the two groups.[17]

Baseline characteristics of participants included in the current analysis, including use of statins and cardiovascular drugs, were well balanced between groups (table 1, supplementary table 3). Fifty four per cent of participants were men and the mean age was 69 years (standard deviation 5). The median follow-up was five years.

Major cardiovascular events
There were 1336 major cardiovascular events during follow-up (vitamin D, n=637 (6.0%); placebo, n=699 (6.6%)). Compared with the placebo group, the rate of major cardiovascular events was lower in the vitamin D group (hazard ratio 0.91, 95% confidence interval 0.81 to 1.01), although the upper bound of the confidence interval is consistent with there being no effect (fig 2, table 2). The hazard ratio did not change with time (supplementary fig 1, supplementary table 4). The difference in the standardised cause specific cumulative incidence at five years was −5.8 events per 1000 participants (95% confidence interval −12.2 to 0.5 per

1000 participants), resulting in a number needed to treat to avoid one major cardiovascular event of 172.

No effect modification was found according to baseline age, sex, or body mass index (fig 3, supplementary figs 2-7). The hazard ratio was lower in people with predicted baseline 25(OH)D concentration ≥50 nmol/L than in those with predicted baseline 25(OH)D <50 nmol/L (hazard ratio 0.87, 95% confidence interval 0.76 to 0.98 v 1.04, 0.84 to 1.27; P for interaction=0.14; fig 3, supplementary figs 8 and 9). The hazard ratio was also lower in people using statins at baseline versus those who were not (0.83, 0.71 to 0.97 v 0.98, 0.84 to 1.13; P for interaction=0.14), and in those who were using cardiovascular drugs at baseline versus those who were not (0.84, 0.74 to 0.97 v 1.01, 0.84 to 1.20; P for interaction=0.12; fig 3, supplementary figs 10-13). In exploratory analyses within subgroups defined according to use of statins or cardiovascular drugs at baseline versus no use, similar patterns were observed (supplementary table 5). In an exploratory analysis requested by reviewers, we performed analyses within subgroups defined by self-report of a major cardiovascular event before baseline. In contrast to the above findings, the effect was stronger in people who did not report a history of major cardiovascular event (0.89, 0.78 to 1.01) versus those who did report an event (0.95, 0.79 to 1.15; supplementary table 5). However, the confidence interval for those reporting an event was wide and the P value for interaction high (0.53).

Specific cardiovascular events
The cumulative incidence and hazard of myocardial infarction were lower in the vitamin D group (hazard ratio 0.81; 95% confidence interval 0.67 to 0.98; table 2, supplementary figs 14 and 15). The same was true of coronary revascularisation, although the confidence interval for the hazard ratio included the null (0.89, 0.78 to 1.01; table 2, supplementary figs 16 and 17). There was no interaction with elapsed time for these outcomes (supplementary figs 15 and 17). The intervention had no apparent effect on stroke (0.99, 0.80 to 1.23; table 2, supplementary fig 18-23). In exploratory analyses of myocardial infarction and coronary revascularisation, we did not find evidence of interactions with baseline statin and other cardiovascular drug use (supplementary table 6).

Discussion
Principal findings
In this analysis of data from the D-Health Trial we found some evidence that supplementation with 60 000 IU of vitamin D_3 per month for up to five years reduced the incidence of major cardiovascular events, particularly myocardial infarction and coronary revascularisation. The absolute differences were small, and the confidence intervals for total major cardiovascular events and coronary revascularisation were consistent with null findings. For total major cardiovascular events, there was some indication of a stronger effect in those who were using statins or other cardiovascular drugs at baseline, or who had higher

Table 2 | Hazard ratios for vitamin D in relation to major cardiovascular events			
Outcome*	Vitamin D (n=10 658)	Placebo (n=10 644)	HR (95% CI)¶
Major event†	637 (6.0)	699 (6.6)	0.91 (0.81 to 1.01)
Myocardial infarction	194 (1.8)	238 (2.2)	0.81 (0.67 to 0.98)
Coronary revascularisation‡	413 (3.9)	462 (4.3)	0.89 (0.78 to 1.01)
Stroke§	172 (1.6)	173 (1.6)	0.99 (0.80 to 1.23)
Haemorrhagic stroke	40 (0.4)	41 (0.4)	0.97 (0.63 to 1.50)
Ischaemic stroke	116 (1.1)	113 (1.1)	1.03 (0.79 to 1.33)

Data are numbers (%).
95% CI=95% confidence interval.
*The number (%) of people with at least one event.
†Composite endpoint including myocardial infarction, stroke, and coronary revascularisation. The number of people who experienced at least one major event is less than the total of the numbers presented for myocardial infarction, stroke, and coronary revascularisation because participants could experience more than one type of event.
‡Composite endpoint including percutaneous coronary intervention (insertion of stent or balloon, artherectomy, thromboectomy, and endarterectomy) or coronary artery bypass grafting.
§Stroke includes diagnosis code I64 (unspecified) and therefore exceeds the total number of ischaemic and haemorrhagic strokes.
¶Estimated using flexible parametric survival models that included age, sex, and state of residence at baseline. The baseline log cumulative hazard function was modelled using restricted cubic spline with two internal knots (placed at the 33rd and 67th percentiles of the uncensored log survival times).
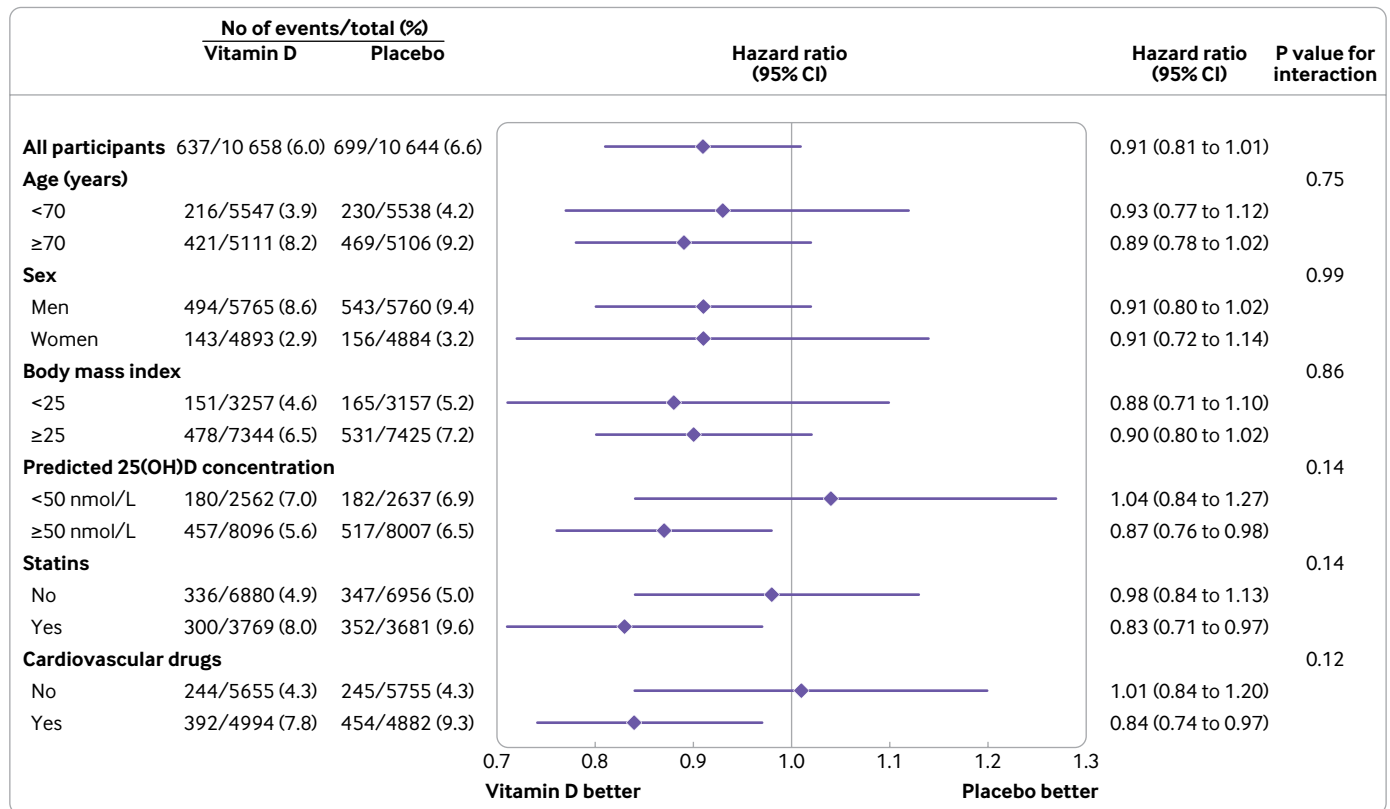
	No of events/total (%)		Hazard ratio (95% CI)	Hazard ratio (95% CI)	P value for interaction
	Vitamin D	Placebo			
All participants	637/10 658 (6.0)	699/10 644 (6.6)		0.91 (0.81 to 1.01)	
Age (years)					0.75
<70	216/5547 (3.9)	230/5538 (4.2)		0.93 (0.77 to 1.12)	
≥70	421/5111 (8.2)	469/5106 (9.2)		0.89 (0.78 to 1.02)	
Sex					0.99
Men	494/5765 (8.6)	543/5760 (9.4)		0.91 (0.80 to 1.02)	
Women	143/4893 (2.9)	156/4884 (3.2)		0.91 (0.72 to 1.14)	
Body mass index					0.86
<25	151/3257 (4.6)	165/3157 (5.2)		0.88 (0.71 to 1.10)	
≥25	478/7344 (6.5)	531/7425 (7.2)		0.90 (0.80 to 1.02)	
Predicted 25(OH)D concentration					0.14
<50 nmol/L	180/2562 (7.0)	182/2637 (6.9)		1.04 (0.84 to 1.27)	
≥50 nmol/L	457/8096 (5.6)	517/8007 (6.5)		0.87 (0.76 to 0.98)	
Statins					0.14
No	336/6880 (4.9)	347/6956 (5.0)		0.98 (0.84 to 1.13)	
Yes	300/3769 (8.0)	352/3681 (9.6)		0.83 (0.71 to 0.97)	
Cardiovascular drugs					0.12
No	244/5655 (4.3)	245/5755 (4.3)		1.01 (0.84 to 1.20)	
Yes	392/4994 (7.8)	454/4882 (9.3)		0.84 (0.74 to 0.97)	

0.7 0.8 0.9 1.0 1.1 1.2 1.3
Vitamin D better   Placebo better

Fig 3 | Effect of vitamin D supplementation on incidence of major cardiovascular events for all participants and by selected baseline characteristics. Hazard ratios (vitamin D *v* placebo) were estimated using flexible parametric survival models. All models included randomisation group, age, sex, and state of residence at baseline. Models producing estimates by levels of age, sex, body mass index, predicted 25-hydroxyvitamin D (25(OH)D) concentration, use of statins, and use of (non-statin) cardiovascular drugs include the characteristic of interest and an interaction between randomisation group and the characteristic of interest. P value for interaction is from a likelihood ratio test comparing models with and without the interaction term. 95% CI=95% confidence interval

predicted vitamin D status, although the interaction terms were not statistically significant. We found no evidence of interaction with age, sex, or body mass index.

Strengths and limitations
The D-Health Trial has several strengths. Over 21 000 people were recruited from the general population and supplemented for five years, with extremely high retention and adherence.[17] Determination of cardiovascular events and mortality outcomes was achieved through comprehensive data linkage to population based administrative data sources. The lack of private hospital data for South Australia and Tasmania would have resulted in a small underestimate of events. However, the underestimation would have been low because only a quarter of participants came from these states, we captured public hospital data, and procedures were able to be identified through Medicare Benefits Schedule data. Importantly, any underestimate would probably not have differed between the study groups.

Comparison with other studies
A meta-analysis of randomised controlled trials, including the VITAL and ViDA studies that had major cardiovascular events or cardiovascular

disease as the primary outcome, concluded that vitamin D supplementation does not prevent cardiovascular events.[13] VITAL did not observe a protective effect for overall major cardiovascular events (including myocardial infarction, stroke, death from cardiovascular causes, and coronary revascularisation; hazard ratios ranged from 0.95 to 0.96).[16] Similarly, the ViDA study concluded that vitamin D supplementation was not protective against total cardiovascular disease (hazard ratio 1.02, 95% confidence interval 0.87 to 1.20) or stroke (0.95, 0.55 to 1.62).[15] The hazard ratio for myocardial infarction was similar to the D-Health Trial findings, although the confidence interval was wide (0.90, 0.54 to 1.50). The D-Health Trial has multiple outcomes, increasing the likelihood of chance findings. However, if the effect on myocardial infarction observed in the D-Health Trial is a true effect, and not due to chance, the reasons for the lack of consistency across studies are unclear. The discrepancy with VITAL might partly be caused by differences in study design and adherence. For example, VITAL excluded people with a history of cardiovascular disease (other than hypertension), and the cohort was more racially diverse. Whereas we used linked data to capture major cardiovascular events, VITAL captured events through participant report in annual surveys, followed by verification of

doi: 10.1136/bmj-2023-075230 | *BMJ* 2023;381:e075230 | the**bmj**

reported events. Differential reporting between study groups might have masked any protective effect of vitamin D. Further, unlike D-Health and ViDA, VITAL used a daily dosing regimen of 2000 IU/day. While evidence is emerging to suggest that daily dosing is of greater benefit for health outcomes such as cancer mortality and infection, the monthly dosing regimen might have led to higher adherence in D-Health than in VITAL; in D-Health 80% of participants reported taking approximately 80% of study tablets, whereas in VITAL around 80% reported taking two thirds of study tablets.[16]

We did not observe a protective effect of vitamin D on stroke. However, the number of stroke events was relatively low, particularly when haemorrhagic stroke, which has different pathophysiology, was excluded; therefore, the confidence intervals were wide and consistent with benefit or harm. Moreover, there are several examples where associations with myocardial infarction and stroke differ,[21-23] so this finding is not entirely unexpected.

In prespecified subgroup analyses, we observed an effect of vitamin D on major cardiovascular events in people who were taking statins or cardiovascular drugs at baseline, but not in those who were not taking these drugs. The interactions were not significant at P<0.05, and it is plausible that these are chance findings. Nevertheless, given the lower power to detect interactions compared with main effects, and the observed strong protective effect in those taking these drugs, these interactions are of interest. There was high concurrent use of statins and other cardiovascular drugs (supplementary table 7), and the interaction could reflect an effect in people who are already at high risk of experiencing a cardiovascular event, rather than a synergistic effect between vitamin D and a particular drug. However, the exploratory analysis by self-reported history of major cardiovascular events was inconsistent with this hypothesis, and it is plausible that there is an interaction between vitamin D and the drugs examined. For example, a number of commonly used statins depend on the enzyme CYP3A4 for activation, and the CYP3A4 gene is responsive to calcitriol, suggesting that vitamin D might alter the effect of statin use.[24] Further investigation of these potential interactions is warranted.

Although we observed a protective effect for vitamin D on major cardiovascular events among people predicted to be vitamin D sufficient at baseline, but not on those predicted to be insufficient, this finding needs to be interpreted with caution because we used predicted rather than measured vitamin D status. Because of the relatively low positive predictive value of the model (0.23), a considerable proportion of those predicted to be in the low group will have been vitamin D replete. While it is plausible that vitamin D supplementation becomes protective at higher serum 25(OH)D concentrations, we found that the 25(OH)D concentration attained in the vitamin D group was only slightly higher in those with predicted deseasonalised baseline serum 25(OH)D concentration ≥50 nmol/L than in those predicted to be deficient (supplementary fig 24).

Generalisability of findings

We need to consider whether the D-Health findings are generalisable to the broader population. A direct comparison with Australian rates of myocardial infarction or major cardiovascular events is not possible because national statistics report myocardial infarction and angina as a single entity. The incidence rates for stroke were a little lower in the D-Health Trial cohort (Australia: 360/100 000 v D-Health: 302/100 000 person years),[2] probably reflecting the better overall health of D-Health participants, who were less likely to report having poor overall health, and less likely to be current smokers.[25] D-Health participants were also less likely to be statin users (35% v 44%),[26] suggesting that if our findings of a more marked effect in statin users at baseline is real, a greater effect might be expected in the Australian population.

The mean 25(OH)D concentration of the D-Health placebo participants throughout the trial, probably indicative of baseline concentration in the cohort, was 77 nmol/L, and the proportion with 25(OH)D <50 nmol/L was 13%.[17] These figures are reasonably representative of the Australian population; in 2011-12 the mean serum 25(OH)D concentration was 69 nmol/L and 16% of people aged ≥65 years had 25(OH)D concentration <50 nmol/L.[27] However, the findings cannot be generalised to populations with a greater prevalence of vitamin D deficiency.

Conclusions

In conclusion, these findings indicate that vitamin D supplementation might reduce the incidence of major cardiovascular events, particularly myocardial infarction and coronary revascularisation. This protective effect could be more marked in those taking statins or other cardiovascular drugs at baseline. Subgroup analyses in other large trials might help to clarify this issue. In the meantime, these findings suggest that conclusions that vitamin D supplementation does not alter risk of cardiovascular disease are premature.

AUTHOR AFFILIATIONS
[1]Population Health Program, QIMR Berghofer Medical Research Institute, Herston, Queensland, Australia

[2]Melbourne School of Population Health, University of Melbourne, Carlton, Victoria, Australia

[3]School of Public Health, The University of Sydney, Sydney, New South Wales, Australia

[4]Department of Medicine, School of Clinical Sciences, Monash University, Clayton, Victoria, Australia

[5]Statistics Unit, QIMR Berghofer Medical Research Institute, Herston, Queensland, Australia

[6]School of Biomedical Science, Faculty of Health, Queensland University of Technology, Kelvin Grove, Queensland, Australia

[7]School of Exercise and Nutrition Sciences, Faculty of Health, Queensland University of Technology, Kelvin Grove, Queensland, Australia

[8]Menzies Institute for Medical Research, University of Tasmania, Hobart, Tasmania, Australia

1 World Health Organization. The top 10 causes of death. Geneva: World Health Organization, 2020. https://www.who.int/news-room/fact-sheets/detail/the-top-10-causes-of-death.

2 Australian Institute of Health and Welfare. Heart, stroke and vascular disease: Australian facts. Canberra: AIHW, 2023. https://www.aihw.gov.au/reports/heart-stroke-vascular-diseases/hsvd-facts/contents/all-heart-stroke-and-vascular-disease.

3 World Health Organization. Noncommunicable diseases country profiles 2018. Geneva: WHO, 2018. https://www.who.int/publications/i/item/9789241514620.

4 von Essen MR, Geisler C. VDR, the vitamin D receptor. In: Choi S, ed. Encyclopedia of Signaling Molecules. Springer New York, 2016: 1-8. doi:10.1007/978-1-4614-6438-9_287-1.

5 Zhang R, Li B, Gao X, et al. Serum 25-hydroxyvitamin D and the risk of cardiovascular disease: dose-response meta-analysis of prospective studies. Am J Clin Nutr 2017;105:810-9. doi:10.3945/ajcn.116.140392

6 Gholami F, Moradi G, Zareei B, et al. The association between circulating 25-hydroxyvitamin D and cardiovascular diseases: a meta-analysis of prospective cohort studies. BMC Cardiovasc Disord 2019;19:248. doi:10.1186/s12872-019-1236-7

7 Wang L, Song Y, Manson JE, et al. Circulating 25-hydroxy-vitamin D and risk of cardiovascular disease: a meta-analysis of prospective studies. Circ Cardiovasc Qual Outcomes 2012;5:819-29. doi:10.1161/CIRCOUTCOMES.112.967604

8 Elamin MB, Abu Elnour NO, Elamin KB, et al. Vitamin D and cardiovascular outcomes: a systematic review and meta-analysis. J Clin Endocrinol Metab 2011;96:1931-42. doi:10.1210/jc.2011-0398

9 Ke L, Mason RS, Kariuki M, Mpofu E, Brock KE. Vitamin D status and hypertension: a review. Integr Blood Press Control 2015;8:13-35. doi:10.2147/IBPC.S49958

10 Zhou A, Selvanayagam JB, Hyppönen E. Non-linear Mendelian randomization analyses support a role for vitamin D deficiency in cardiovascular disease risk. Eur Heart J 2022;43:1731-9. doi:10.1093/eurheartj/ehab809

11 Meng X, Li X, Timofeeva MN, et al. Phenome-wide Mendelian-randomization study of genetically determined vitamin D on multiple health outcomes using the UK Biobank study. Int J Epidemiol 2019;48:1425-34. doi:10.1093/ije/dyz182

12 Huang T, Afzal S, Yu C, et al, China Kadoorie Biobank Collaborative Group. Vitamin D and cause-specific vascular disease and mortality: a Mendelian randomisation study involving 99,012 Chinese and 106,911 European adults. BMC Med 2019;17:160. doi:10.1186/s12916-019-1401-y

13 Barbarawi M, Kheiri B, Zayed Y, et al. Vitamin D supplementation and cardiovascular disease risks in more than 83000 individuals in 21 randomized clinical trials: a meta-analysis. JAMA Cardiol 2019;4:765-76. doi:10.1001/jamacardio.2019.1870

14 Jackson RD, LaCroix AZ, Gass M, et al, Women's Health Initiative Investigators. Calcium plus vitamin D supplementation and the risk of fractures. N Engl J Med 2006;354:669-83. doi:10.1056/NEJMoa055218

15 Scragg R, Stewart AW, Waayer D, et al. Effect of monthly high-dose vitamin D supplementation on cardiovascular disease in the vitamin D assessment study: a randomized clinical trial. JAMA Cardiol 2017;2:608-16. doi:10.1001/jamacardio.2017.0175

16 Manson JE, Cook NR, Lee IM, et al, VITAL Research Group. Vitamin D supplements and prevention of cancer and cardiovascular disease. N Engl J Med 2019;380:33-44. doi:10.1056/NEJMoa1809944

17 Neale RE, Baxter C, Duarte Romero B, et al. The D-Health Trial: a randomised controlled trial of the effect of vitamin D on mortality. Lancet Diabetes Endocrinol 2022;10:120-8. doi:10.1016/S2213-8587(21)00345-4

18 Neale RE, Armstrong BK, Baxter C, et al. The D-Health Trial: a randomized trial of vitamin D for prevention of mortality and cancer. Contemp Clin Trials 2016;48:83-90. doi:10.1016/j.cct.2016.04.005

19 Waterhouse M, Baxter C, Duarte Romero B, et al. Predicting deseasonalised serum 25 hydroxy vitamin D concentrations in the D-Health Trial: an analysis using boosted regression trees. Contemp Clin Trials 2021;104:106347. doi:10.1016/j.cct.2021.106347

20 Waterhouse M, English DR, Armstrong BK, et al. A randomized placebo-controlled trial of vitamin D supplementation for reduction of mortality and cancer: Statistical analysis plan for the D-Health Trial. Contemp Clin Trials Commun 2019;14:100333. doi:10.1016/j.conctc.2019.100333

21 Borges MC, Schmidt AF, Jefferis B, et al, UCLEB Consortium**.
    Circulating fatty acids and risk of coronary heart disease and stroke:
    individual participant data meta-analysis in up to 16 126 participants.
    J Am Heart Assoc 2020;9:e013131. doi:10.1161/JAHA.119.013131

22 Colantonio LD, Bittner V, Safford MM, et al. Lipoprotein(a) and the
    risk for coronary heart disease and ischemic stroke events among
    black and white adults with cardiovascular disease. *J Am Heart
    Assoc* 2022;11:e025397. doi:10.1161/JAHA.121.025397

23 Ricci C, Wood A, Muller D, et al. Alcohol intake in relation to non-fatal
    and fatal coronary heart disease and stroke: EPIC-CVD case-cohort
    study. *BMJ* 2018;361:k934. doi:10.1136/bmj.k934

24 Wakeman M. A literature review of the potential impact of medication
    on vitamin D status. *Risk Manag Healthc Policy* 2021;14:3357-81.
    doi:10.2147/RMHP.S316897

25 Australian Institue of Health and Welfare. Older Australians. Canberra:
    AIHW, 2021. https://www.aihw.gov.au/reports/older-people/older-
    australians.

26 Ofori-Asenso R, Ilomäki J, Zomer E, Curtis AJ, Zoungas S, Liew D.
    A 10-year trend in statin use among older adults in Australia: an
    analysis using national pharmacy claims data. *Cardiovasc Drugs
    Ther* 2018;32:265-72. doi:10.1007/s10557-018-6794-x

27 Malacova E, Cheang PR, Dunlop E, et al. Prevalence and predictors
    of vitamin D deficiency in a nationally representative sample of
    adults participating in the 2011-2013 Australian Health Survey. *Br J
    Nutr* 2019;121:894-904. doi:10.1017/S0007114519000151

Web appendix: Supplementary material