A Mendelian randomization study of the effect of calcium on coronary artery disease, myocardial infarction and their risk factors

Lin Xu1,2, Shi Lin1 & C. Mary Schooling1,3

Meta-analyses of randomized controlled trials (RCTs) suggest calcium could have adverse effects on cardiovascular disease, although these findings are controversial. To clarify, we assessed whether people with genetically higher calcium had a higher risk of coronary artery disease (CAD), myocardial infarction (MI) and their risk factors. We used a two-sample Mendelian randomization study. We identified genetic variants (single nucleotide polymorphisms (SNPs)) that independently contributed to serum calcium at genome-wide significance which we applied to large extensively genotyped studies of CAD, MI, diabetes, lipids, glycaemic traits and adiposity to obtain unconfounded estimates, with body mass index (BMI) as a control outcome. Based on 4 SNPs each 1 mg/dl increase in calcium was positively associated with CAD (odds ratio (OR) 1.49, 95% confidence interval (CI) 1.02–2.17), MI (OR 1.58, 95% CI 1.06–2.35), LDL-cholesterol (0.21 standard deviations, 95% CI 0.01–0.4), total cholesterol (0.21 standard deviations, 95% CI 0.03–0.38) and possibly triglycerides (0.19 standard deviations, 95% CI −0.1–0.48), but was unlikely related to BMI although the estimate lacked precision. Sensitivity analysis using 13 SNPs showed a higher risk for CAD (OR 1.87, 95% CI 1.14–3.08). Our findings, largely consistent with the experimental evidence, suggest higher serum calcium may increase the risk of CAD.

Calcium is widely seen as part of a healthy diet that promotes bone health. Recent meta-analysis of randomized controlled trials (RCTs) has suggested that calcium has little benefit for fracture prevention1. Unexpectedly, some meta-analyses of these RCTs also suggest that calcium may increase the risk of cardiovascular events, myocardial infarction (MI) and stroke2–5, although not all meta-analyses of RCTs concur on this point6. Controversy over calcium supplements arose with the Auckland Calcium Study (ACS) reporting calcium supplements increased cardiovascular disease (CVD) events in a secondary analysis of a large RCT in 20087. However, this was a hypothesis generating study because CVD events were not one of the original study outcomes. Re-analyzing data from the US Women's Health Initiative (WHI), in a sub-group analysis of those not taking calcium supplements at randomization, the same authors of the ACS found that calcium supplementation increased the risk of MI2. Incorporating the results in a meta-analysis, the use of calcium supplementation increased the risk of MI, by 21%.2. This meta-analysis only included a small number of trials (n = 7) and compliance was low especially in the calcium arm due to adverse effects on gastrointestinal disorders, which might attenuate the potential impact on CVD events. Moreover, none of these trials were specifically designed to include primary CVD endpoints, and thus CVD events were not collected in a standard and systematic manner. In 2015, another meta-analysis of RCTs found the risk of coronary heart disease (CHD) in older women was not increased by calcium supplements8. An increase in CVD events in earlier studies was attributed to selection bias9, however, all these studies relied on adverse event reporting, whose classification is open to biases, because CVD events may not be considered as intervention-related and were not reported in some trials9. Another two recent meta-analysis of RCTs demonstrated calcium supplements (with or without vitamin D) increased risk of MI by 24–28%.10. The discrepancy between meta-analyses may be due to the inclusion of controversial RCTs, such as trials with participants who

1School of Public Health, Li Ka Shing Faculty of Medicine, The University of Hong Kong, Hong Kong SAR, China. 2School of Public Health, Sun Yat-sen University, Guangzhou, Guangdong Province, China. 3School of Urban Public Health, Hunter College and CUNY School of Public Health, New York, New York, USA. Correspondence and requests for materials should be addressed to L.X. (email: linxu@hku.hk)
Serum calcium. From a large meta-analysis of genome-wide association studies (GWAS) including 20,611 individuals of European ancestry, we obtained single nucleotide polymorphisms (SNPs) independently contributing to serum calcium at genome-wide significance ($p < 5 \times 10^{-8}$). We assessed correlation (linkage disequilibrium) between SNPs using SNP Annotation and Proxy (SNAP) search system (http://www.broadinstitute.org/mpg/snap ldsearchpw.php) for the same reference catalog and population. When the correlation coefficient between SNPs was high ($R^2 \geq 0.8$) we discarded the SNP with the larger $P$ value, when the correlation was lower we kept all SNPs but took into account their correlation matrix. We identified pleiotropic effects of these SNPs from Ensembl (Homo sapiens – phenotype) (http://grch37.ensembl.org/Homo_sapiens/Info/Index), a comprehensive genotype to phenotype cross-reference. We used SNPs that are approximately independent as determinants of calcium ($i.e., R^2 < 0.01$) as the main analysis. As a sensitivity analysis we further included SNPs with low correlation (0.01 $\leq R^2 < 0.8$), identified from the linkage disequilibrium correlation matrix (Supplementary Table 1).

Control outcome. We used BMI as a control outcome that, based on RCTs, should be unrelated to serum calcium. Genetic associations with BMI (kg/m$^2$) have been contributed by The Genetic Investigation of Anthropometric Traits (GIANT) investigators and have been downloaded from https://www.broadinstitute.org/collaboration/giant/index.php/GIANT_consortium_data_files which has BMI for 152,893 men and 171,977 women of European ancestry without diabetes.

Statistical analysis. SNP-specific Wald estimates (ratio of SNP on outcome to SNP on calcium) of the effect of calcium on each outcome were combined using weighted generalized linear regression to account for correlation between the SNPs, giving an odds ratio (OR) for CAD, MI, T2DM, and regression coefficients ($\beta$) for the other outcomes with 95% confidence interval (CI). As a sensitivity analysis we used inverse variance weighted (IVW) estimator with fixed effects. Provided that the genetic variants are uncorrelated, the IVW estimate is asymptotically equal to the two-stage least squares estimate commonly used with individual-level data. In IVW, the ratio estimates from each IV (or SNP) are combined in an inverse-variance weighted estimator. We also used a weighted median method to combine the SNP specific estimates for the uncorrelated SNPs. Even after
shown in Table 1. For the 13 selected SNPs, 9 were correlated (R² ≥ 0.8), and 2 SNPs with pleiotropic effects (on bilirubin and parathyroid hormone) giving 13 genome-wide significant SNPs (rs4306808 in CSTA, rs17711722 in VKORC1LI, rs1067 in WRDSB) as associated with serum calcium concentrations. The first-stage F-statistics for the IV including these 4 SNPs was 52.

Association of genetically determined serum calcium concentrations with CAD risk. Table 2 shows that the estimates for the causal effect of 1 mg/dl higher serum calcium were consistently in the direction of higher risk of CAD and MI based on 4 SNPs, although sometimes the lower limit of the confidence interval included the null value in both CARDioGRAM (OR 1.62, 95% CI 0.86 to 3.07) and CARDioGRAMplusC4D 1000 Genomes-based GWAS (OR 1.49, 95% CI 1.02 to 2.17 for CAD, and 1.58, 95% CI 1.06 to 2.35 for MI) using IVW or using a weighted median (1.66, 95% CI 1.12 to 1.81 for CAD, and 1.65, 95% CI 1.06 to 2.56 for MI). The results were consistent based on 13 SNPs in CARDioGRAM (OR 1.87, 95% CI 1.14 to 3.08) and CARDioGRAMplusC4D 1000 Genomes-based GWAS using IVW (1.25, 95% CI 0.92 to 1.70 for CAD, and 1.32, 95% CI 0.94 to 1.85 for MI).

Association of genetically determined calcium with CAD risk factors. In all analyses the estimated effect of calcium on BMI was null but lacked precision. Similar observations from results using 4 SNPs were found for T2DM (OR 1.34, 95% CI 0.65 to 2.74) and glycemic parameters including fasting glucose, insulin and log HOMA-IR. In contrast, based on 4 SNPs, the estimates for the causal effect of 1 mg/dl higher serum calcium on LDL-cholesterol (0.21 standard deviation, 95% CI 0.01 to 0.40) and total cholesterol (0.29 standard deviation, 95% CI 0.09 to 0.48) were positive but in sensitivity analysis the confidence interval included the null for triglycerides (0.19 standard deviation, 95% CI -0.1 to 0.48). Using the MR-Egger method with the 4 SNPs, we could not estimate the MR-Egger slope estimate is the same as the regression coefficient from IVW. If the intercept is zero it suggests that there is no violation of the exclusion restriction criteria (i.e., no horizontal pleiotropy); it provides an estimate of the average pleiotropic effect across all of the genetic variants, which reflects the effect of the joint instruments on outcome (e.g., CAD/MI when there is zero effect of the genetic variants on the risk factor (e.g. calcium). An intercept term that differs from zero suggests horizontal pleiotropy and that the IV estimate may be biased. The weakness of the instruments was evaluated using the first-stage F-statistics calculated by F = R²/K 

Table 1. Characteristics of the single nucleotide polymorphisms (SNP) used for genetically determined serum calcium. MAF: minor allele frequency. *Increase in calcium (mg/dl) per effect allele.

| SNP          | Nearest gene | Effect allele | Other allele | Effect | Standard error | MAF  | R²   | Main analysis |
|--------------|--------------|---------------|--------------|--------|----------------|------|------|--------------|
| rs4306808    | FAM162A      | G             | C            | 0.054  | 0.0084         | 0.1675 | 0.08 |
| rs7336933    | DGKH/KIAA0564| G             | A            | 0.022  | 0.0040         | 0.1248 | 0.01 ✓ |
| rs17711722   | VKORC1LI     | T             | C            | 0.021  | 0.0030         | 0.4355 | 0.02 ✓ |
| rs17267388   | PARP9        | A             | G            | 0.036  | 0.0059         | 0.1300 | 0.03 |
| rs1067       | WRDSB        | A             | G            | 0.033  | 0.0059         | 0.1444 | 0.03 |
| rs13095172   | CSTA;CASR    | T             | C            | 0.028  | 0.0046         | 0.3524 | 0.04 |
| rs11929034   | PARP9        | A             | G            | 0.038  | 0.0063         | 0.1312 | 0.03 |
| rs4491840    | CCDC58       | A             | G            | 0.042  | 0.0060         | 0.1673 | 0.05 |
| rs16832956   | CSTA;CASR    | G             | C            | 0.044  | 0.0053         | 0.1641 | 0.05 |
| rs17251221   | CASR         | G             | A            | 0.061  | 0.0063         | 0.0942 | 0.06 ✓ |
| rs10222633   | CASR         | G             | A            | 0.030  | 0.0042         | 0.3852 | 0.04 |
| rs10491003   | GATA3        | T             | C            | 0.027  | 0.0050         | 0.1040 | 0.01 ✓ |
| rs9864290    | CSTA         | C             | T            | 0.028  | 0.0047         | 0.4453 | 0.04 |

Genetic determinants of serum calcium. GWAS gave 128 SNPs related to serum calcium with p < 1 × 10⁻⁵, from which we excluded 84 SNPs that do not reach genome wide significance (p < 5 × 10⁻⁸), 29 highly correlated SNPs (R² ≥ 0.8), and 2 SNPs with pleiotropic effects (on bilirubin and parathyroid hormone) giving 13 genome-wide significant SNPs (rs4306808 in FAM162A, rs7336933 near DGKH/KIAA0564, rs17711722 in VKORC1LI, rs1067 in WRDSB, rs17267388 and rs11929034 in PARP9, rs4491840 in CCDC58, rs9864290 in CSTA, rs10491003 (closest gene GATA3), and rs16832956, rs17251221, rs13095172 and rs10222633 in CASR) as shown in Table 1. For the 13 selected SNPs, 9 were correlated (R² > 0.1) among each other according to SNAP with HapMap release 22, as shown in the correlation matrix in Supplementary Table 1. Of these 13 SNPs, 4 from 4 different genes (rs7336933 (DGKH/KIAA0564), rs17711722 (VKORC1LI), rs17251221 (CASR) and rs10491003 (GATA3)) were uncorrelated R² < 0.05 in HapMap CEU population. The first-stage F-statistics for the IV including these 4 SNPs was 52.

| SNP          | Nearest gene | Effect allele | Other allele | Effect | Standard error | MAF  | R²   | Main analysis |
|--------------|--------------|---------------|--------------|--------|----------------|------|------|--------------|
| rs4306808    | FAM162A      | G             | C            | 0.054  | 0.0084         | 0.1675 | 0.08 |
| rs7336933    | DGKH/KIAA0564| G             | A            | 0.022  | 0.0040         | 0.1248 | 0.01 ✓ |
| rs17711722   | VKORC1LI     | T             | C            | 0.021  | 0.0030         | 0.4355 | 0.02 ✓ |
| rs17267388   | PARP9        | A             | G            | 0.036  | 0.0059         | 0.1300 | 0.03 |
| rs1067       | WRDSB        | A             | G            | 0.033  | 0.0059         | 0.1444 | 0.03 |
| rs13095172   | CSTA;CASR    | T             | C            | 0.028  | 0.0046         | 0.3524 | 0.04 |
| rs11929034   | PARP9        | A             | G            | 0.038  | 0.0063         | 0.1312 | 0.03 |
| rs4491840    | CCDC58       | A             | G            | 0.042  | 0.0060         | 0.1673 | 0.05 |
| rs16832956   | CSTA;CASR    | G             | C            | 0.044  | 0.0053         | 0.1641 | 0.05 |
| rs17251221   | CASR         | G             | A            | 0.061  | 0.0063         | 0.0942 | 0.06 ✓ |
| rs10222633   | CASR         | G             | A            | 0.030  | 0.0042         | 0.3852 | 0.04 |
| rs10491003   | GATA3        | T             | C            | 0.027  | 0.0050         | 0.1040 | 0.01 ✓ |
| rs9864290    | CSTA         | C             | T            | 0.028  | 0.0047         | 0.4453 | 0.04 |
not reject the hypothesis that an association of these 4 SNPs with CAD, MI, T2DM or their risk factors was not independent of the effects on calcium (Supplementary Table 2).

Discussion

Consistent with most previous meta-analyses of RCTs, we found that higher serum calcium was associated with a higher risk of CAD and probably MI, although we cannot definitively rule out the possibility of no effect. Findings for CAD risk factors, LDL- and total cholesterol were also positive, consistent with previous RCTs. In addition, as expected, calcium appeared not to be associated with BMI. As such, our study replicates findings from RCTs and extends them by showing the same pattern of associations for endogenous calcium in very large studies including substantial numbers of men as well as of women.

Our findings for CAD are consistent with most previous meta-analyses of RCTs pertaining to women, although the estimate is higher than that suggested by these RCTs, which could be because calcium has a stronger association with MI in men. However, MR is more suitable for establishing direction than exact effect sizes, because genetically determined calcium represents lifetime exposure, whereas the RCTs were relatively short in duration and potentially biased towards the null by non-compliance, if the participants were successfully blinded.

More generally, our findings are consistent with the observation that countries with higher calcium intake, such as Northern European countries, also have higher CAD mortality rates, while countries with low calcium intake tend to have low CAD mortality, such as China, Korea and Japan. Of course, ecological evidence never proves causality, but this observation does require some explanation.

A previous Mendelian randomization study, using 17 SNPs for calcium from in and around the CASR gene, gave no association of calcium with fasting glucose (beta = 0.001, 95% CI 0.14 to 0.14). We also cannot completely rule out the possibility that the result on glucose was due to an insufficient sample size. A post-hoc power calculation for the association with fasting glucose (beta = 0.001, 95% CI 0.14 to 0.14) showed power of less than 20%, suggesting larger MR studies are necessary to further clarify the effect on fasting glucose. Our results are consistent with previous RCTs showing calcium supplementation has no effects on fasting glucose, although calcium supplements intake tends to reduce fasting insulin and improve insulin resistance.

The heritability for total calcium is between 33% and 78% in twin studies, suggesting that serum calcium levels are tightly regulated. Three major hormones are involved in the regulation of calcium homeostasis, parathyroid hormone (PTH), calcitonin and 1,25-dihydroxyvitamin D, which act on their corresponding receptors in bone, gut and kidney to maintain serum calcium concentrations. A key regulator of the PTH release is the calcium-sensing receptor (CASR), which is mainly in the plasma membrane of chief cells of the parathyroid gland and in cells of the renal tubule. The CASR gene encodes a protein which binds to calcium and thus.

|                | Main analysis (4 SNPs) | Weighted-median | Sensitivity analysis (13 SNPs) |
|----------------|------------------------|-----------------|-------------------------------|
|                | IVW                    | WGL regression  |                                |
|                | OR (95% CI)            | OR (95% CI)     | OR (95% CI)                   |
| CAD (CARDIoGRAM) |                        |                 |                               |
| CARDIoGRAM GWAS | 1.62 (0.86 to 3.07)    | 1.48 (0.80 to 2.71) | 1.87 (1.14 to 3.08)           |
| 1000 Genomes   | 1.49 (1.02 to 2.17)    | 1.66 (1.12 to 1.81) | 1.25 (0.92 to 1.70)          |
| 1000 Genomes –MI | 1.58 (1.06 to 2.35)   | 1.65 (1.06 to 2.56) | 1.32 (0.94 to 1.85)          |
| T2DM           |                        |                 |                               |
| DIAGRAM GWAS   | 1.34 (0.65 to 2.74)    | 1.31 (0.62 to 2.8) | 1.64 (0.87 to 3.10)          |
| Trans-ethnic GWAS meta-analysis | 1.21 (0.71 to 2.07) | 1.31 (0.71 to 2.5) | 1.06 (0.65 to 1.75)          |

Table 2. Mendelian randomization estimates of the causal association of serum calcium (mg/dL) with coronary artery disease (CAD), myocardial infarction (MI) and their risk factors. WGL: weighted generalized linear; IVW: inverse variance weighted; SD: standard deviation; OR: odds ratio; CI: confidence interval; T2DM: type-2 diabetes mellitus; BMI: body mass index; LDL-C: low-density lipoprotein cholesterol; HDL-C: high-density lipoprotein cholesterol; TG: triglycerides; TC: total cholesterol; HOMA-IR: homeostatic model assessment of insulin resistance. *P < 0.05; **P < 0.01.
plays an essential role in calcium homeostasis. Apart from CASR, another gene, GATA3 encodes a GATA transcription factor involved in T cell lymphopoiesis, renal and vestibular morphogenesis, and parathyroid gland development. Functional studies have shown that GATA3 haploinsufficiency causes hypoparathyroidism in populations of different ethnicities. Other mechanisms exist by which calcium might contribute to CAD, for example by promoting carotid intima-media thickness, coronary artery calcification, as occurs with calcium based phosphate binders versus non-calcium based phosphate binders, and coagulation. Acute induction of severe hypercalcemia in animal models reduces blood clotting time by 50%. In vitro, increasing calcium concentrations across the physiological range reduces the clotting time of human blood. These mechanisms could underlie effects of calcium on CAD.

Several methodologic considerations and limitations bear discussion. First, the genetic variants used for genetically determined calcium were all strongly related to calcium at GWAS level significance. No obvious reason exists for the existence of confounders of the association of these genetic variants with the outcomes considered here, for example by population stratification, because the underlying studies relate to relatively ethnically homogeneous populations of mainly European ancestry. Both calcium levels and CVD rates vary across Europe, which could be due to other factors determining calcium and CVD, of which the calcium related genetic variants are only a marker. However, the estimate was dominated by genetic variants from the calcium-sensing receptor (CASR) gene functionally relevant to calcium, making such confounding unlikely. Second, the genetic variants used are not known to be associated with other phenotypes that might influence CAD and its risk factors, thus making biases from direct associations of SNPs with the outcomes, i.e., “pleiotropy” or violation of the “exclusion-restriction” assumption, unlikely. Moreover, we found no evidence of directional pleiotropy, i.e. that the genetic variants used to predict calcium had effects on CAD or its risk factors independent of effects via calcium. Third, we replicated established experimental evidence from meta-analysis of RCTs by showing that calcium had no effect on BMI, which gives more credence to the other estimates using the same genetic determinants of calcium. Fourth, the use of summarized data in two samples, serum calcium levels were not measured in the sample with the outcome. However, two-sample instrumental variable analysis is more robust to chance associations than analysis of a single sample. Fifth, it is not possible to perform sub-group analysis or multivariable analysis as rigorously in two-sample MR as in one-sample MR using individual-level data. For example, whether the SNPs for serum calcium have different effects on CAD, T2DM or other CVD risk factors at different levels of serum calcium, by sex or at different ages could not be tested, and whether there was significant heterogeneity among sub-populations for individual instruments could not be assessed. In addition, there might be participant overlap in this two-sample MR (i.e., the same data used both for SNP selection and to calculate the IV effect). However, given the very large sample size of the CARDIoGRAMplusC4D consortium, assuming a 50% overlapping the sample overlap in this study is only 12%, because the dataset for deriving calcium related SNP was much smaller (n = 20,611). Thus bias from sample overlapping, if any, should not be a major concern. Finally, both IVW and MR-Egger methods use weights that under the “NO Measurement Error (NOME) assumption”, that is assuming the SNP-exposure associations to be known, rather than estimated. This assumption cannot be tested directly. However, we used P statistics to quantify the strength of NOME violation for MR-Egger and did not find significant evidence of the violation.

Our study indicates that genetically higher serum calcium concentrations could have a harmful effect on MI and CAD. On the precautionary principle, given calcium does not seem as important in bone health as thought, our findings suggest reconsideration of the use of calcium supplementation and particularly fortification in the general population, especially in products used by older people who have higher risk of CAD.

References
1. Tai, V., Leung, W., Grey, A., Reid, I. R. & Bolland, M. J. Calcium intake and bone mineral density: systematic review and meta-analysis. BMJ 351, h183, doi: 10.1136/bmj.h183 (2015).
2. Bolland, M. J., Grey, A., Avenell, A., Gamble, G. D. & Reid, I. R. Calcium supplements with or without vitamin D and risk of cardiovascular events: reanalysis of the Women’s Health Initiative limited access dataset and meta-analysis. BMJ 342, d2040, doi: 10.1136/bmj.d2040 (2011).
3. Bolland, M. J. et al. Effect of calcium supplements on risk of myocardial infarction and cardiovascular events: meta-analysis. BMJ 341, c3691, doi: 10.1136/bmj.c3691 (2010).
4. Mao, P. J. et al. Effect of calcium or vitamin D supplementation on vascular outcomes: a meta-analysis of randomised controlled trials. International journal of cardiology 169, 106–111, doi: 10.1016/j.ijcard.2013.08.055 (2013).
5. Wang, L., Manson, J. E. & Sesso, H. D. Calcium intake and risk of cardiovascular disease: a review of prospective studies and randomized clinical trials. American journal of cardiovascular drugs: drugs, devices, and other interventions 12, 105–116, doi: 10.2165/11595400-000000000-00000 (2012).
6. Lewis, J. R. et al. The Effects of Calcium Supplementation on Verified Coronary Heart Disease Hospitalization and Death in Postmenopausal Women: A Collaborative Meta-Analysis of Randomized Controlled Trials. Journal of Bone and Mineral Research 30, 165–175, doi: 10.1002/jbmr.2311 (2015).
7. Bolland, M. J. et al. Vascular events in healthy older women receiving calcium supplementation: randomised controlled trial. BMJ 336, 262–266, doi: 10.1136/bmj.39440.525752.BE (2008).
8. Ioannidis, J. A. Adverse events in randomized controlled trials. International journal of epidemiology 38, 737–739, doi: 10.1093/ije/dyr253 (2009).
9. Reid, I. R., Bristow, S. M. & Bolland, M. J. Cardiovascular complications of calcium supplements. Journal of cellular biochemistry 116, 494–501, doi: 10.1002/jcb.25028 (2015).
10. Larsen, E. R., Mosedalde, L. & Foldspang, A. Vitamin D and Calcium Supplementation Prevents Osteoporotic Fractures in Elderly Community Dwelling Residents: A Pragmatic Population-Based 3-Year Intervention Study. Journal of Bone and Mineral Research 19, 370–378, doi: 10.1359/jbmr.0901240 (2004).
11. Tabesh, M., Azadbakht, L., Faghihimani, E., Tabesh, M. & Esmaillzadeh, A. Effects of calcium-vitamin D co-supplementation on metabolic profiles in vitamin D insufficient people with type 2 diabetes: a randomised controlled clinical trial. Diabetologia 57, 2038–2047, doi: 10.1007/s00125-014-3313-x (2014).
12. Sanchez, M. et al. Oral calcium supplementation reduces intraplatelet free calcium concentration and insulin resistance in essential hypertensive patients. Hypertension 29, 531–536 (1997).
Author Contributions
L.X. and S.L.L. did data analysis, L.X. and C.M.S. wrote the main manuscript text. All authors reviewed the manuscript.

Additional Information
Supplementary information accompanies this paper at http://www.nature.com/srep

Competing financial interests: The authors declare no competing financial interests.

How to cite this article: Xu, L. et al. A Mendelian randomization study of the effect of calcium on coronary artery disease, myocardial infarction and their risk factors. Sci. Rep. 7, 42691; doi: 10.1038/srep42691 (2017).

Publisher’s note: Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

This work is licensed under a Creative Commons Attribution 4.0 International License. The images or other third party material in this article are included in the article’s Creative Commons license, unless indicated otherwise in the credit line; if the material is not included under the Creative Commons license, users will need to obtain permission from the license holder to reproduce the material. To view a copy of this license, visit http://creativecommons.org/licenses/by/4.0/

© The Author(s) 2017