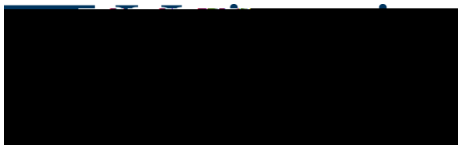

Methods and Results: We estimated the association of CRP genetic variant +1444C T (rs1130864) with CRP levels and with CHD in five studies and then pooled these analyses (N = 18,637 participants amongst whom there were 4,610 cases). CRP was associated with potential confounding factors (socioeconomic position, physical activity, smoking and body mass) whereas genotype (rs1130864) was not associated with these confounders. The pooled odds ratio of CHD per doubling of circulating CRP level after adjustment for age and sex was 1.13 (95%CI: 1.06, 1.21), and after further adjustment for confounding factors it was 1.07 (95%CI: 1.02, 1.13). Genotype (rs1130864) was associated with circulating CRP; the pooled ratio of geometric means of CRP level among individuals with the TT genotype compared to those with the CT/CC genotype was 1.21 (95%CI: 1.15, 1.28) and the pooled ratio of geometric means of CRP level per additional T allele was 1.14 (95%CI: 1.11, 1.18), with no strong evidence in either analyses of between study heterogeneity (I^2 = 0%, p 0.9 for both analyses). There was no association of genotype (rs1130864) with CHD: pooled odds ratio 1.01 (95%CI: 0.88, 1.16) comparing individuals with TT genotype to those with CT/CC genotype and 0.96 (95%CI: 0.90, 1.03) per additional T allele (I^2 = 7%, p 0.6 for both meta-analyses). An instrumental variables
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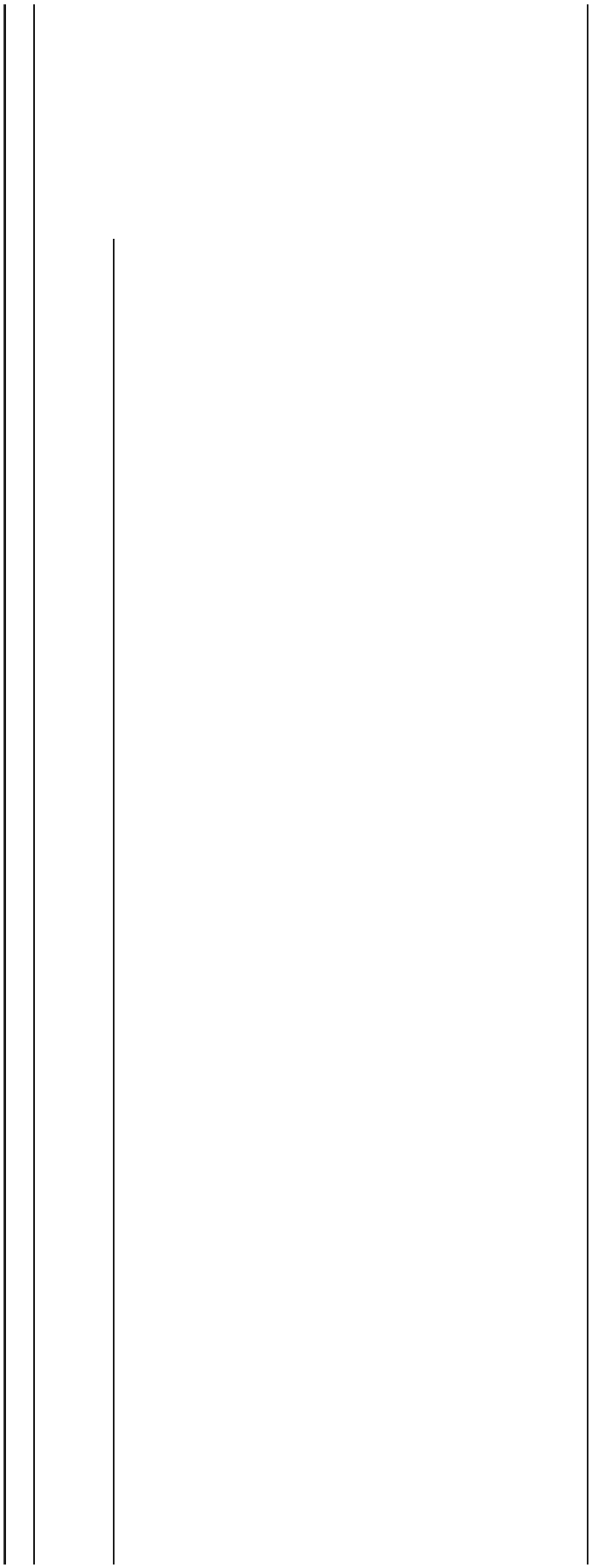

heart attack.[26] For any participant who indicated that they had a physician diagnosis of myocardial infarction their medical records were reviewed and the myocardial infarction only

altered CRP levels but did not affect any other cardiovascular risk factors. To our knowledge no such trials have been conducted. The use of genetic variants as instrumental variables to determine the causal association between circulating CRP and CHD provides an alternative to such a randomized controlled trial, with the advantage that this approach can be performed in

The rs1130864 SNP has been consistently shown to be associated

stratification is unlikely to have importantly confounded our genetic association results. Developmental canalisation (the process by which target receptors or organs develop differently in response to varying levels of the exposure of interest during key developmental periods) might limit the Mendelian randomization process. The extent to which this occurs with modest effects such

The fact that rs1130864 explains less than 1% of the variation in CRP within each of our studies, whilst affecting statistical precision is unlikely to result in bias. As noted previously,[8,9,10] many medications that are used in randomised controlled trials to determine causality explain a similarly small proportion of variation in the potentially causal risk factor, but with adequate sample sizes (sometimes, as in our Mendelian randomization study presented here, obtained only through meta-analysis of data from a number of trials) provide precise and valid estimates of the causal effect on clinical endpoints.

For example, blood pressure lowering therapies explain ~2% of the variation in blood pressure, and in participants who are randomised to either active blood pressure lowering therapy or control in randomised controlled trials there will be many other environmental and genetic factors that influence variation in blood pressure. Nonetheless, an adequately powered randomised trial of the effect of blood pressure medication on stroke (or other cardiovascular outcomes) is, rightly, accepted as unbiased evidence of the causal effect of blood pressure on stroke risk.[9,10]

With respect to other Mendelian randomization studies, single SNPs that have been shown to be robustly associated with low density lipoprotein cholesterol (LDLc), and that explain less than 1% of the variation in circulating LDLc, are robustly associated with CHD, with the magnitude of this association being somewhat larger than that predicted from the causal randomised controlled trial evidence relating statins (which reduce LDLc) to CHD.[49,50] It has been suggested that the somewhat stronger effects with these genetic variants relates to the fact that the randomised difference in LDLc occurring as a result of genetic variants is life-long, whereas that occurring as a result of statins is from mid-adult life only.[51,52] Similarly, we have recently shown that a single SNP in *FTO* (which again explains less than 1% of variation in body mass index or total fat mass) is associated with a wide range of vascular and metabolic outcomes, including fasting glucose, insulin and lipids, with magnitudes of association that hat an 8ntlyobserv

ed between 210 to 1062 CHD cases, have also found genetic variants within the *CRP* gene to be unrelated to prevalent and incident CHD events, despite these variants being associated with CRP levels,[13–17] or have found associations in the opposite direction to what one would anticipate if higher CRP levels were causally associated with increased CHD risk,[13] or associations only in very specific subgroups.[18,19] Whilst such subgroup analyses might represent true differences in the effect of *CRP* or CHD, in general subgroup analyses fail to replicate and should be treated with caution until replicated in other independent studies. Furthermore, none of the large genome wide association studies of CHD have found variation in *CRP* to be robustly associated with CHD.[46–48] By contrast variants associated with established risk factors for CHD (e.g. LDLc) are identified in these genome wide association studies CHD. Taking these findings together with our own results–the largest study to date (N = 4160 cases; four times the largest previous published study) to relate variation in *CRP* to CHD–there does not appear to be any strong evidence for an association of *CRP* with CHD events, despite its association with CRP levels. These findings together would suggest that variation in circulating CRP level is not importantly causally related to CHD risk and that genetic profiling of *CRP* is unlikely to be useful in the prediction of CHD. However, we acknowledge that a very large genetic (Mendelian randomization) study is required to definitively demonstrate whether there is no causal association of CRP with CHD. A newly established collaboration that will include at least 30,000 CHD events will over the coming years be able to provide this definitive answer.[55]

In a companion paper we examined the association of 3 tag SNPs [+1444T. C (rs1130864); +2303G. A (rs1205) and +4899T. G (rs 3093077)] in the *CRP* gene with serum CRP and carotid intima-mediaTD02.5(4);)]1.1055(ackno0010TD1emTD1emTD1emTD1.

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