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Genetic analysis of the PCSK9 locus in psychological, psychiatric, metabolic and cardiovascular traits in UK Biobank

Rachel Hay¹, Breda Cullen¹, Nicholas Graham¹, Donald M. Lyall¹, Alisha Aman⁶, Jill P. Pell¹, Joey Ward¹, Daniel J. Smith¹,³ and Rona J. Strawbridge⁴,⁵

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The association between severe mental illness (SMI) and cardiovascular and metabolic disease (CMD) is poorly understood. PCSK9 is expressed in systems critical to both SMI and CMD and influences lipid homeostasis and brain function. We systematically investigated relationships between genetic variation within the PCSK9 locus and risk for both CMD and SMI. UK Biobank recruited ~500,000 volunteers and assessed a wide range of SMI and CMD phenotypes. We used genetic data from white British ancestry individuals of UK Biobank. Genetic association analyses were conducted in PLINK, with statistical significance defined by the number of independent SNPs. Conditional analyses and linkage disequilibrium assessed the independence of SNPs and the presence of multiple signals. Two genetic risk scores of lipid-lowering alleles were calculated and used as proxies for putative lipid-lowering effects of PCSK9. PCSK9 variants were associated with central adiposity, venous thrombosis embolism, systolic blood pressure, mood instability, and neuroticism (all \( p < 1.16 \times 10^{-4} \)). No secondary signals were identified. Conditional analyses and high linkage disequilibrium \( (r^2 = 0.98) \) indicated that mood instability and central obesity may share a genetic signal. Genetic risk scores suggested that the lipid-lowering effects of PCSK9 may be causal for greater mood instability and higher neuroticism. This is the first study to implicate the PCSK9 locus in mood-disorder symptoms and related traits, as well as the shared pathology of SMI and CMD. PCSK9 effects on mood may occur via lipid-lowering mechanisms. Further work is needed to understand whether repurposing PCSK9-targeting therapies might improve SMI symptoms and prevent CMD.

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INTRODUCTION

It has been long recognised that severe mental illness (SMI) is accompanied by an increased burden of cardiovascular and metabolic diseases (CMD) [1, 2]. The direct cause of this comorbidity remains ambiguous; however, it is likely influenced by factors such as genetics, socioeconomic status, poor diet, sedentary lifestyle, substance abuse and side effects of psychiatric medication [3]. Additionally, genetic studies have begun to provide evidence of shared mechanisms underlying mental and physical ill-health [4–7].

For many years CMD research has focused on PCSK9 and its encoded protein (also PCSK9), because of its key role in lipid homeostasis. Specifically, PCSK9 binds to the low-density lipoprotein (LDL) receptors (LDLR) and targets them for degradation [8]. Lipid homeostasis is important for energy storage and release and is therefore central to metabolic processes. The balance of LDL in circulation vs in tissues is important, as high circulating levels of lipids can lead to inappropriate storage of lipids, which contribute to insulin resistance and systemic inflammation observed in CMD [9]. PCSK9 has direct effects on pancreatic cells and adipose tissue, which are critical organs for maintenance of insulin sensitivity, with lipid-lowering and non-lipid-lowering mechanisms (respectively) implicated [10]. In particular, deposition of lipids in blood vessel walls is a key feature and risk marker of cardiovascular diseases including myocardial infarction and ischemic stroke. High levels of circulating LDL have a causal relationship with atherosclerotic plaques [11] and the importance of PCSK9 in these processes is highlighted by the development of PCSK9 inhibitors as a treatment of atherosclerotic disease. Clinical trials have demonstrated the benefits (and importantly no side-effects) of lipid-lowering via increased LDLR activity (reviewed in [12]). In addition to lipid-lowering, PCSK9 has direct effects on platelet activation and thrombosis risk [13], which have been prevented or reduced with PCSK9 inhibitors, experimentally [12] and clinically [14].

PCSK9’s association with SMI remains less established, however an association seems plausible due to its roles in brain functions [15, 16], which include brain cholesterol trafficking and lipoprotein trafficking [17] and insulin resistance, which is implicated in SMI [18]. Indeed, there is evidence that depressive symptoms alone can raise PCSK9 levels, with a subsequent effect on insulin resistance [19]. Under some circumstances, lipids can cross the blood brain barrier, and as well as influencing lipid regulation, PCSK9 is expressed in areas with high proliferative ability. It has a positive correlation with increased post-mitotic neurones and decreased neuroepithelial cells. Additionally, it has been shown to promote or prevent apoptosis in many neuronal pathways. This
emphasises the influence of PCSK9 on neurogenesis and neuronal apoptosis. Finally, inhibition of PCSK9 is associated with reduced neuroinflammation. PCSK9 has been investigated in neurodegenerative disorders such as Alzheimer’s disease, where altered levels of the protein levels in cerebrospinal fluid (CSF) [17] and brain samples [20] have been associated with disease. PCSK9 levels have also been reported to be upregulated in alcohol use disorders or ischemic stroke [15, 20]. Clinical studies demonstrate that, despite some initial concerns, PCSK9 inhibitors do not have adverse effects on cognition [21], although rate of adverse psychiatric reactions due to lipid-lowering (by either PCSK9 inhibition or non-PCSK9 methods such as statins) requires further examination [22].

A number of studies exploring the genetic variants in the PCSK9 locus has been conducted: One explored a single missense variant, rs11591147 (R46L), as an indicator of potential long-term effects of PCSK9 inhibition on a large number of phenotypes [23], however there is evidence that the locus contains more than one independent signal [24], thus a more extensive assessment of the locus is warranted. Another genetic study investigated a number of loss-of-function variants on cognition and Alzheimer’s disease, however this study was of limited size (total N = 878) [25]. A further study (N = 2487) identified sex-specific effects of two variants in the PCSK9 locus on CSF PCSK9 levels [20].

Given that the expression of PCSK9 is observed in the brain as well as the liver (Fig. 1), and the known functions of PCSK9, we aimed to systematically investigate whether genetic variation in the PCSK9 locus contributes to the shared genetic predisposition and underlying pathological mechanism of both CMD and SMI. Furthermore, we describe the genetic architecture in the PCSK9 locus and identify mechanisms by which genetic variants in this region may influence biological traits. Finally, we evaluate whether the effects of PCSK9 genetic variation were likely to act through LDLR-lowering pathways. If PCSK9 variation contributes to SMI by the same mechanisms as its effect on CMD, there is the potential to repurpose CMD therapies for the improvement of SMI symptoms and CMD prevention in these high CMD-risk individuals.

**METHODS**

**UK biobank**

The UK Biobank (UKB) is a large dataset of 502,613 participants, which has genetic, ethical approval granted by the National Research Ethics Service (approval letter dated dated 29th June 2021, Ref 21/NW/0157). This work was carried out using UKB application #6553 (PI Rona Strawbridge) and #17689 (PI Donald Lyall). Data is provided by participants and collected by the National Health Service as part of their care and support. A more thorough detailed description of the UKB data set can be found elsewhere in open-access reports [26, 27].

UKB volunteers were recruited through 22 assessment centres during 2006–2011. Individuals were largely aged between 40–69 at baseline. Trained clinical staff carried out physical examinations and touchscreen questionnaires to gather baseline measurements along with individual and family medical history. Blood samples allowed biomarker analysis and DNA extraction for genome-wide genotyping. The genotyping, quality control and imputation used standard protocols and took place centrally.

Average systolic and diastolic blood pressure reading was calculated, each from two measurements, and adjusted for the use of antihypertensive medications as per Ehret et al. [28], prior to analysis (SBPadj or DBPadj). Height and weight were recorded and BMI (kg/m², reflective of total obesity) calculated. Waist and hip measurements allowed for calculation of waist-hip ratio (reflective of central obesity) which was adjusted for BMI (WHRadjBMI, reflecting central obesity accounting for total obesity) prior to analysis as per Shungin et al. [29]. Type 2 diabetes status was defined as per Eastwood et al. [30] (from self-reported medical history and medication data). Reported medical histories included venous thromboembolism (VTE), stroke, and ischaemic heart disease (ISH). Self-reported smoking was categorised as current smokers vs. non or former smokers.

Psychiatric traits were analysed by different baseline questionnaires. Firstly, the 12 items of the Eysenck Personality Questionnaire-Revised Short form were used to assess neuroticism. Lack of enthusiasm in the last two weeks was recorded to assess individual’s anhedonia (data field 2060). Individuals responding “not at all” were considered controls, whilst those responding “several days”, “more than half of the days” or “nearly every day” were considered cases. Risk-taking was assessed by a self-reported question asking, “Would you describe yourself as someone who takes risks?” (data field 2040) and mood instability by the question “Does your mood often go up and down?” (data field 1920). Individuals responding “yes” were considered cases, and those responding “no” were considered as controls. Any individual responding “prefer not to answer”, or “don’t know” was excluded from the analysis. A follow-up online mental health questionnaire (collected 2016–2017) was used to clarify the lifetime history of SMI [31, 32] and related traits. This included generalised anxiety disorder (LifeGAD), major depressive disorder (LifeMDD), bipolar disorder (LifeBD), or addiction [31, 32].

**SNP selection**

The PCSK9 locus is defined as the PCSK9 gene ±250 kb (UCSC GRC37/hg19, Chromosome 1:55,505,221-55,530,525 ± flanking regions). Initially all single nucleotide polymorphisms (SNPs) within the locus were identified. Discovery analyses included only unrelated individuals of white British ancestry (N = 402,820, SNP-N = 2037). Secondary analyses were conducted in unrelated individuals of white European ancestry (N = 50,510, SNP-N = 207), south Asian ancestry (N = 7726, SNP-N = 2167), African-Caribbean ancestry (N = 7644, SNP-N = 3386) and mixed ancestry (N = 10,447, SNP-N = 2532). Genetic variants with minor allele frequencies of >1% by ancestry group were analysed.

**Statistical analyses**

Minitab 19.2 was to calculate descriptive statistics. For genetic analyses, an additive model was assumed and Plink 1.07 [33] was used with logistic or linear regression. Continuous variables were assessed for normality prior to analysis. All analyses were adjusted for age, sex, eight genetic principal components and genotyping chip, except WHRadjBMI where these covariates are included in its construction. Treatment with lipid-lowering and anti-hypertensive medications were included as covariates for analysis of ISH and stroke. The number of independent loci was determined using independent pairwise filtering using default parameters in PLINK 1.07, and Bonferroni correction for multiple testing (using number of SNPs) was applied. The threshold for significance in discovery analyses was p value <1.16 x 10^-3 and in secondary analyses, p value <1.28 x 10^-4 for South Asian, <4.69 x 10^-4 for African-Caribbean, <1.20 x 10^-5 for white European and <8.85 x 10^-5 for mixed ancestry individuals. LocusZoom [34] was used to generate regional plots.

**Conditional analysis: number of signals**

Conditional analyses were used to identify multiple independent signals. Analyses of traits with significant associations were repeated including the lead SNP as a covariate (for example, analysis of SNPs vs BMI were adjusted for age, sex, eight genetic principal components, genotyping chip and the lead BMI SNP). An additional independent signal was defined as at least one SNP meeting the threshold for significance in the conditional analysis (as well as the original analysis). Where an additional signal was identified, a further round of conditional analysis was conducted, including the lead SNPs from both signals as covariates. This process was repeated until no further significant signals remained.

**Conditional analysis: independence of signals**

Conditional analyses were also used to determine whether the signals for each significant trait were independent. Here the analyses of traits with significant associations were repeated, with the addition of the lead SNP for a different trait as a covariate (for example, analysis of SNPs vs BMI were adjusted for age, sex, eight genetic principal components, genotyping chip and the lead VTE SNP). Lead SNPs were considered independent if the effect size (Beta or OR) changed by <0.05 (arbitrary value, but consistent with the maximum magnitude of change likely with fluctuations in sample size due to inclusion of a SNP as a covariate). No distance measure was used to assess independence. Linkage disequilibrium (LD) between lead SNPs was assessed using a randomly selected 10,000 unrelated white British ancestry individuals from UKB. Haploview [35] allowed visualisation...
Fig. 1  Tissue expression patterns of genes of interest. Gene expression of A PCSK9, B DHCR24, and C USP24, by tissue (GTEx analysis release V8). Expression values are shown in transcription per million (TPM) calculated from a gene model with isoforms collapsed to a single gene. Box plots are shown as median and 25th and 75th percentiles; points are displayed as outliers if they are above or below 1.5 times the interquartile range.
of the LD between the lead SNPs, to complemented the conditional analyses.

**Effect of Lead SNPs on PCSK9 levels**
The impact of lead SNPs on circulating PCSK9 levels was investigated using publicly available genome-wide meta-analysis summary statistics [24].

**PCSK9 polygenic scores**
Three studies have highlighted genetic variants with functional effects that can be used to proxy lifelong reduction in LDL levels (Schmidt, 2019 #275; Ference, 2016 #274; Lyall, 2021 #273). Schmidt et al. used four SNPs in a polygenic score (PGS) (Schmidt, 2019 #275), whilst Ference et al. selected seven SNPs (Ference, 2016 #274). Lyall et al. used six of the seven Ference SNPs and noted that individual SNPs had little impact when removed (Lyall, 2021 #273). We constructed two PGS, using four variants (PGS4 (Schmidt, 2019 #275)) or seven variants (PGS7 (Ference, 2016 #274)), by summing the lipid-lowering alleles for each individual. Only individuals of those who completed the mental health questionnaire.

**Follow-up analyses**
The GTex portal (https://www.gtexportal.org/home/ [36], accessed 2021-08-26) was used to explore PCSK9 gene expression and genotype-specific gene expression patterns. LDEexpress was used to explore genotype-specific gene expression patterns of proxies of lead SNPs (https://ldexpress [37], 2021-08-26). The GWAS Catalog, (https://www.ebi.ac.uk/gwas/, 2021-08-17), was used to identify previous associations in the PCSK9 locus. Ensembl VEP (https://www.ensembl.org/info/docs/tools/vep/index.html, [38]) was used to explore the predicted functional effects of a SNP, with SIFT and PolyPhen being used to assess tolerance. The Human Protein Atlas (https://www.proteinatlas.org/, [39]) was used to investigate PCSK9 protein expression.

**RESULTS**
Table 1 provides the cohort characteristics. The average age of participants was 56.9 years, with 20.6% being treated with lipid-lowering medication and rates of ISH and stroke were low (6.1% and 2.1% respectively).

**PCSK9 locus and CMD**
Significant associations are summarised in Table 2 and Fig. 2. One SNP was associated with SBPadj (rs2647282-A, Beta (se) = 1.09 (1.04 – 1.11), p = 8.89 × 10⁻⁵, Fig. 2A), eight with WHRadjBMI (lead rs7543163-C, 0.001 (<0.001), p = 9.21 × 10⁻⁶, Fig. 2B) and five with VTE (rs746952505-CA, OR (95% confidence interval) 1.07 (1.04–1.11), p = 3.74 × 10⁻⁵). As the lead SNP for VTE is not available in LocusZoom, the second significant variant was used for plotting and also considered in conditional analyses (rs12071742-A, 1.09 (1.04–1.13), p = 8.89 × 10⁻⁵, Fig. 2C). One SNP was significantly associated with stroke (rs7266260-G, 0.86
Table 2. Lead SNPs in White British ancestry subset.

<table>
<thead>
<tr>
<th>Phenotype</th>
<th>SNP</th>
<th>Chr</th>
<th>BP</th>
<th>A1</th>
<th>A2</th>
<th>MAF</th>
<th>N</th>
<th>SNPs meeting the threshold for significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>SBPadj</td>
<td>rs2647282</td>
<td>55,724,437</td>
<td>A</td>
<td>C</td>
<td>0.36</td>
<td>343,109</td>
<td>1</td>
<td>p = 4.20E-05</td>
</tr>
<tr>
<td>WHRadjBMI</td>
<td>rs7543163</td>
<td>55,515,481</td>
<td>C</td>
<td>T</td>
<td>0.39</td>
<td>400,588</td>
<td>1</td>
<td>p = 9.2E-06</td>
</tr>
<tr>
<td>VTE (Lead)</td>
<td>rs746952505</td>
<td>55,750,180</td>
<td>CA</td>
<td>C</td>
<td>0.24</td>
<td>264,073</td>
<td>1</td>
<td>p = 3.74E-05</td>
</tr>
<tr>
<td>VTE (Proxy)</td>
<td>rs12071742</td>
<td>55,747,825</td>
<td>A</td>
<td>G</td>
<td>0.39</td>
<td>277,385</td>
<td>1</td>
<td>p = 8.89E-05</td>
</tr>
<tr>
<td>Stroke</td>
<td>rs72662600</td>
<td>55,732,403</td>
<td>G</td>
<td>A</td>
<td>0.08</td>
<td>339,008</td>
<td>1</td>
<td>p = 9.56E-05</td>
</tr>
<tr>
<td>Neuroticism Score</td>
<td>rs12069079</td>
<td>55,652,791</td>
<td>G</td>
<td>A</td>
<td>0.20</td>
<td>26,396</td>
<td>1</td>
<td>p = 4.53E-06</td>
</tr>
</tbody>
</table>

LD r² between these two SNPs = 0.46. A1 Effect or minor allele, A2 Non-effect allele, MAF Effect or minor allele frequency, N SNPs meeting the threshold for significance, rs746952505 is not available in locuszoom, so for plots rs12071742 is presented as the lead.

PCSK9 locus and SMI

When considering SMI-related traits (Table 2 and Fig. 2), 30 significant associations were identified for mood instability (rs11206514-C, p = 2.68 × 10⁻⁵, Fig. 2E) and for neuroticism (lead rs12069079-G, p = 4.53 × 10⁻⁶, Fig. 2F). No significant associations were observed for anhedonia, risk-taking behaviour, GAD, MDD, BD or addiction. Conditional analyses demonstrated no additional signals for mood instability (Stable 5, Sfig 5A, B) or neuroticism (Stable 6, Sfig 6A, B).

Assessment of independence of signals

Conditional analyses were used to assess whether signals for SMI and CMD traits were independent (ie. represent distinct genetic effects). If there is little or no change in the effect of a trait when adjusting for the lead of a different trait, then signals can be considered independent.

The lead SNP for SBPadj (rs2647282) demonstrated little change when conditioning on the other lead SNPs (Stable 1, Sfig 1 C–G), consistent with low LD with other lead SNPs (max r² = 0.21, Fig. 3). Effects of WHRadjBMI SNPs were unchanged when conditioning on the lead SNPs for SBP, VTE, stroke or neuroticism score (Stable 2, Sfig 2 C–E, G) but the association was rendered null by conditioning on the lead SNP for mood instability (Sfig 3F).

This is consistent with the very low LD (r² < 0.01) with other lead SNPs, but almost complete LD with the mood instability lead SNP (r² = 0.98, Fig. 3). Of note, the WHRadjBMI-increasing allele had a mood instability-increasing effect (1.02 (1.01–1.03) p = 1.50 × 10⁻⁵). The VTE signal appears to be distinct from other signals, as conditioning on other lead SNPs had little or no impact (Stable 3, Sfig 3 C–G), consistent with very low LD with other signals (max LD r² = 0.02, Fig. 3). Similarly, the stroke signal appears independent from the other signals, with conditional analyses having minimal effect (Stable 4, Sfig 4 C–G) and very low LD with any other signal (max r² = 0.05).

Visual inspection suggests that the signals for mood instability and neuroticism partially overlap (Stable 5–6, Sfigs 5–6) consistent with the results of the conditional analyses. However, mood instability is a component of the neuroticism phenotype, therefore the conditional analyses likely reflects the phenotypic relationship between these two variables, rather than a shared genetic signal, as the LD between the lead SNPs is very low (LD r² = 0.04, Fig. 3). Of note, the mood instability-increasing allele had a WHRadjBMI-increasing effect (0.001 (<0.001), p = 1.25 × 10⁻⁶).

Lead SNPs and PCSK9 levels

Of the seven lead/proxy SNPs, five were available in the meta-analysis of PCSK9 levels [24] (Table 3). Nominal p < 0.05 associations were observed for the SNPs associated with SBP (decreasing allele was associated with decreased levels of PCSK9), WHRadjBMI (increasing allele was associated with decreased levels of PCSK9 levels) and mood instability (increasing allele was associated with decreased levels of PCSK9).

PCSK9-functional SNPs

To explore whether lipid-lowering functions of PCSK9 might explain the associations observed, we tested two lipid-lowering PGS for impact on our traits of interest (PGS4 [40] and PGS7 [41]). Individual effects of each SNP in the PGS on the traits of interest are presented in Table 7. Only rs10888897 had significant effects on WHRadjBMI (T, 0.001 (<0.001), p = 3.75 × 10⁻⁵), and a non-
Fig. 2  Regional plots of main results in white British ancestry individuals. (A) SBPadj, (B) WHRadjBMI, (C) VTE, (D) stroke, (E) mood instability and (F) neuroticism score. Locuszoom regional plots of each significant SNP identified for a trait in the UKB. SNPs are aligned on the X-axis by their position on chromosome 9, and by their association with the trait on the Y-axis \((P \text{ values are on a } - \log_{10})\). Significance was set at \(p < 1.16 \times 10^{-4} \) or \(-\log_{10} P > 3.93\) (approximated by the red horizontal line). SNPs are colour coded by their estimates of pairwise LD \(r^2\) with lead SNP.
significant effect on mood instability ($T$, 1.02 (1.01–1.03) $p = 0.0003$), consistent with high LD between rs10888897 and the lead SNPs for these traits (Fig. 3). The non-significant effect on neuroticism likely reflects phenotypic similarity of these traits, as the LD with the lead neuroticism trait is low. PGS4 and PGS7 showed consistent null effects on SBP, WHRadjBMI, VTE and stroke ($p > 0.139$, STable 8), suggesting a mechanism independent of lipid-lowering. For VTE and stroke, this is consistent effects of pcsk9 on thrombosis in animal models [42].

PGS7 but not PGS4 had nominally significant ($p < 0.05$) effects on mood instability and both PGS4 and PGS7 had nominally significant ($p < 0.05$) effects on neuroticism, where the direction of effect suggests that lipid-lowering effects of the PCSK9 locus might contribute to increasing these psychological traits. Whilst rs10888897 is in high LD with the lead SNPs for mood instability ($r^2 = 0.95$, Fig. 3), it is possible that this signal is driving the PRS association with mood instability. The same is not true for the associations of the PGS with neuroticism, as PGS4 contains only SNPs with minimal LD (max $r^2 = 0.01$, Fig. 3) with the lead SNP for mood instability).

Secondary analyses

Analyses of additional ancestry groups were restricted to phenotypes significant in discovery analyses with sufficient data. No SNPs met the thresholds for significance in any analyses (neuroticism, mood instability, SBP adj or WHRadjBMI in all groups or VTE in white European ancestry). Lead SNPs for secondary analyses (reaching $p < 1 \times 10^{-5}$) are presented in STable 9 and a comparison of effect directions of the lead SNPs from the discovery analysis, across ancestry groups is provided in STable 10. The lack of significant findings in non-white British ancestry individuals is likely due to underpowered analyses. However, LD between the lead SNPs in the non-white British ancestry individuals confirmed the conclusion that the signals for

**Fig. 3** Linkage disequilibrium (LD) between lead SNPs. Haploview LD plot showing the lead SNPs in the PCSK9 locus associated with cardiometabolic or psychiatric traits, and the SNPs included in PGS4 (*) and PGS7 (#). Each box provides estimated statistics of % frequency of coinheritance ($r^2$ values). Darker shaded boxes represent stronger LD ($r^2$ colours).
### Table 4. Genotype-specific gene expression of trait-associated SNPs or proxies (I.D.R. $R^2 > 0.50$).

<table>
<thead>
<tr>
<th>Trait</th>
<th>Lead SNP</th>
<th>Proxy SNP</th>
<th>Position</th>
<th>R2</th>
<th>R2'</th>
<th>$R^2$</th>
<th>Tissue</th>
<th>mRNA expression</th>
<th>Protein expression</th>
<th>Functional effects</th>
</tr>
</thead>
<tbody>
<tr>
<td>SBP</td>
<td>rs2647282</td>
<td>rs11206513</td>
<td>55,724,437</td>
<td>0.915</td>
<td>1.00</td>
<td>1.00</td>
<td>Heart-Atrial Appendage</td>
<td>0.915</td>
<td>1.00</td>
<td>loss-of-function</td>
</tr>
<tr>
<td>NSB</td>
<td>rs11206513</td>
<td>rs11206513</td>
<td>55,724,437</td>
<td>0.915</td>
<td>1.00</td>
<td>1.00</td>
<td>Heart-Atrial Appendage</td>
<td>0.915</td>
<td>1.00</td>
<td>loss-of-function</td>
</tr>
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<td>NSB</td>
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<td>rs11206513</td>
<td>55,724,437</td>
<td>0.915</td>
<td>1.00</td>
<td>1.00</td>
<td>Heart-Atrial Appendage</td>
<td>0.915</td>
<td>1.00</td>
<td>loss-of-function</td>
</tr>
</tbody>
</table>

### DISCUSSION

This systematic analysis of the PCSK9 locus for effects on CMD and SMI-related traits in UK Biobank identified signals associated with four cardiometabolic traits (VTE, stroke, SBPadj and WHRadjBMI) and two psychological traits (mood instability and neuroticism). We demonstrated that the WHRadjBMI and mood instability signals only act through lipid-lowering mechanisms. Finally, we present evidence that these SNP regulate mRNA expression levels of PCSK9, DHCR24 and USP24.

To our knowledge this is the first study to report an effect of the PCSK9 locus, specifically on lipid-lowering effects, on psychological phenotypes (mood instability and neuroticism) of relevance to psychiatric traits. The associations with SBPadj and WHRadjBMI were unsurprising, however, it is unusual that SBPadj, but not DBPadj, was significantly associated with this locus. The
associated with the PCSK9 locus with VTE was unexpected, given that lipid accumulation and inflammation are generally considered less relevant to VTE than arterial disease. However, a non-lipid-lowering mechanism is plausible, as pck9-knockout in experimental animals reduced platelet activation [42], likely via clearance of clotting factor VIII by the LDLR family. This is consistent with our finding that the same allele of the VTE proxy (rs12071742-A) both increases VTE risk and PCSK9 levels (Table 3). A detailed summary of PCSK9 effects (and their inhibition) on platelet activation has been reported [13], with recent evidence for platelet-derived PCSK9 adding complexity [44].

The almost complete LD reported here between the lead variants for mood instability and WHRadjBMI is intriguing; does mood instability subsequently influence central fat accumulation (through diet and exercise preferences for example), or does this genetic signal have multiple effects on different genes and in different tissues (pleiotropy), or is there one mechanism common to these two very different traits? The gene expression evidence of the minor allele (associated with increased WHRadjBMI and decreased PCSK9 protein levels) both increasing PCSK9 levels in adipose tissue and decreasing PCSK9 levels in brain tissue suggests that looking at one system in isolation might not be enough.

The non-white British ancestry results were largely uninformative (due to limited power), however the consistent LD patterns across ancestry groups supports the overlap of signals for mood instability and central fat accumulation (SFig. 7).

In addition to PCSK9, expression data highlights the roles of DHCR24 and USP24 in SMI and CMD: DHCR24 has been shown to have a neuroprotective role during inflammation [45] with loss of DHCR24 expression being observed in Alzheimer’s disease [45, 46]. WHRadjBMI-increasing alleles being associated with reduced DHCR24 levels in the brain could suggest the same mechanisms underly central adiposity and lack of neuroprotection. USP24 is implicated in Parkinson’s disease and increased autophagy [47], but expression data presented here indicate a role for USP24 in heart, nerve, and muscle tissue, but not adipose or brain tissues.

Fig. 4 shows some of the potential mechanisms and pathways by which PCSK9 could contribute to SMI and CMD. Further studies are required to elucidate which of these (and/or other) pathways are most important to SMI, CMD and their comorbidity, and whether there is potential for repurposing PCSK9-inhibitors (currently used for CMD) for improving SMI symptoms and CMD prevention.

Strengths and limitations

SMI and CMD and their comorbidity are increasingly recognised as an important individual, societal and economic burden, therefore this study is very relevant. The large sample size and consistent phenotyping in the UKB is a strength, as is the conservative Bonferroni correction, both of which contribute to the reliability of these findings. The PGS provided additional depth to mechanistic insights of this study. The biases of UKB are well established (Swanson, 2012 #268), and self-report to assess psychiatric traits could provide additional biases (for example, mood instability and risk taking: Individuals with SMI may not regard their behaviour as ‘risky’ or recognize their mood to fluctuate). Therefore, UKB likely underestimates effects of SNPs on disease. Further exploration of these findings in additional ancestry groups would be of great value.

CONCLUSION

In a systematic analysis of the PCSK9 locus, we identified genetic associations with SBPsdj, WHRadjBMI, VTE, stroke, mood instability and neuroticism score, with the same signal (indicated by LD r² = 0.98) associated with WHRadjBMI and mood instability. This is the first study to implicate the PCSK9 locus in the shared pathology of CMD and SMI. The lipid-lowering PGS demonstrating a nominal association with neuroticism score and mood instability highlights important considerations for lipid-lowering drugs. Subsequent analyses should consider causal assessments and temporal patterns of PCSK9, DHCR24 and USP24 expression, to elucidate the mechanisms linking CMD and SMI. Further exploration of this locus in a more diverse cohort would improve understanding of the locus and health equality in the future.

DATA AVAILABILITY

All UK Biobank data (raw, coding and results) are available to approved researchers via application to the UK Biobank study (https://www.ukbiobank.ac.uk/). Coding and summary level results are available upon request (Dr Rona J Strawbridge, rona.strawbridge@glasgow.ac.uk).

REFERENCES


COMPETING INTERESTS
The authors have no competing interests.

ETHICAL APPROVAL
Generic ethical approval granted by the NHS National Research Ethics Service (approval letter dated 13 May 2016, Ref 16/NW/0274).

ADDITIONAL INFORMATION
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Correspondence and requests for materials should be addressed to Rona J. Strawbridge.

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