Supplementary Material

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Overview

The MDD2000+ project comprises a total of 2431 cases with MDD and 3673 screened controls from different sources and genotyped on different platforms (main paper ***Tables 1*** and ***2***). Samples were provided by the Queensland Institute of Medical Research (QIMR, Australia), the Netherlands Study of Anxiety and Depression (NESDA), the Netherlands Twin Registry (NTR), the University of Edinburgh (UoE, United Kingdom), and the Molecular Genetics of Schizophrenia (MGS) study (controls only, United States). Genotyping was conducted on different Illumina and Affymetrix platforms and because the overlap in genotyped single nucleotide polymorphisms (SNPs) is limited, association analysis is based on a set of >1M imputed SNPs. The numbers of SNPs for each analysis set (main table ***Table 1***), represents genotyped SNPs surviving all quality control (QC) criteria that were used for imputation. Differing QC measures were applied to each sample (described below), before imposing uniform QC across all sample sets as described in the main paper.

Genotyping and initial quality control for the QIMR I317, I370 and I610 samples

The QIMR Genetic Epidemiology Laboratory collects a wide range of phenotypic variables on twin individuals and their family members. DNA samples from some of these subjects have been submitted for genotyping under projects funded with focus on a number of primary phenotypes. The QIMR samples genotyped on Illumina platforms are described in detail in Medland et al (1) (see their Table 2 projects 1-4,6). The Illumina platforms corresponding to the I317, I370 and I610 sample sets are the Illumina 317k, Illumina HumanCNV370-Quadv3 and the Illumina Human610-Quad respectively. QC steps for each project (1) included rejection of SNPs with Bead Studio Gen Call Scores < 0.7, SNP call rate < 0.95, Hardy Weinberg Equlibrium (HWE) test $p < 10^{-6}$, and SNPs with minor allele frequency (MAF) < 0.01. The different genotyping projects included overlapping individuals and families. After merging of the QIMR Illumina projects, the data were screened for missingness within individuals (removal of individuals where > 0.05 of genotyped SNPs failed), pedigree, sex errors, and Mendelian errors (genotypes for all family members for a given SNP were removed on detection of errors). Non-European ancestry outliers were removed. After QC, where one individual from a monozygotic (MZ) twin pair had been genotyped, duplicate genotypes were assigned to the ungenotyped co-twin. All but one of the genotyping projects represented the community samples of twins interviewed. One sample (Project 3 in Medland Table 2), contributing to analysis set I610, was a sample of migraine cases and non-migraine controls. Individuals interviewed in the NAG/IRPG studies (see METHODS) featured in all analysis sets (Table 1) but they were preferentially genotyped in the I370 analysis ste where they comprise 92% of the QIMR samples. Individuals interviewed Individuals surviving QC and satisfying the case and control definitions of section 2i) were selected as QIMR cases and controls and uploaded for joint analysis of all data.

Genotyping and initial quality control for the I370 NTR controls

In total 1405 individuals were submitted for genotyping to the Institute of Human Genetics, LIFE & BRAIN Center, University of Bonn, Germany on the Human610-Quad platform. Of these 577 satisfied the criteria for controls. No QC was applied to the genotypes prior to uploading of 657,366 SNPs for joint MDD2000+ analysis. Given that the ratio of controls:cases was ~2.5:1 for QIMR samples in the I610 set, it was decided to impute the NTR controls together with the QIMR samples in the I370 set where the ratio of controls:cases was much lower, hence only a subset of the genotyped SNPs was used.

Genotyping and initial quality control for A6.0 case sample

DNA from 1874 samples with MDD (1248 QIMR, 160 NESDA/NTR and 466 UoE) were submitted for genotyping to the Translational Genomics Research Institute (Tgen), Phoenix, Arizona. Since these samples have not been described elsewhere, we provide full details of the QC process. DNA samples were shipped on 96 well plates, 20uL per sample at 50ng of DNA per uL. At Tgen samples were re-plated ensuring that each centre was proportionally represented on each plate. A single QC sample was allocated to a standard position on each of the 21 plates.

Genotypes were initially called at Tgen using Affymetrix's Genotyping Console Software which runs the Birdseed v2 algorithm plate by plate. Only samples with a contrast statistic > 0.4 were included. Samples with a Birdseed call rate < 97.0 were excluded iteratively, each time excluding the sample with the lowest call rate. Samples with poor genotyping results were replated and the process repeated (using two additional plates). Genotypes were provided by Tgen for 1867 samples (1827 after subtracting deliberate duplicates and QC controls).

We calculated identity-by-state (IBS) matrix from the genotypes of autosomal markers and calculated the mean identity between all sample pairs. This provided evidence for contamination of a small number of samples (high mean IBS between

adjacent plated samples). These samples were excluded and genotypes were recalled plate by plate using the BirdSeed v2 algorithm as implemented in the BirdSuite software (2, 3). Individual genotypes were filtered after calling, changing the confidence score threshold from the default of 0.1 the more stringent value of 0.02. This threshold was established by investigating the effect of a range of thresholds on a number of QC measures. The QC checks listed in ***Table S1*** Stages 1-3 were performed at each threshold level. We compared the number of SNPs dropped at each stage and compared the quantile-quantile (Q-Q) plot from HWE tests of the SNPs surviving all QC steps. A genomic inflation of HWE test statistics is commonly observed in other data sets using the Affymetrix chips reflecting residual batch effects. We found that more stringent thresholds preferentially excluded poor performing SNPs i.e. the proportion of SNPs failing QC Stages 2 and 3 decreased as the confidence score threshold became more stringent.

Importantly, a total of 327 individuals had been previously genotyped on an Illumina platform and we used the 268 of these that had excellent dm.all_qc statistics (from program apt-geno-qc in the Affymetrix Power Tools) of 0.95 or higher to investigate genotype discordancy on SNPs genotyped in common across the platforms (Stage 4). We chose a confidence score threshold of 0.02 as the best balance between SNP numbers and genotype confidence. In total 646,601 SNPs remained. This represents about 70,000 SNPs fewer than often used in other studies using the Affymetrix 6.0 chip (e.g.(4)). However, we believe the extra stringency is justified because only cases were genotyped on this platform. The default confidence score threshold was used for calling individuals' genotypes for the SNPs that survived this QC step. As an additional check, sex for all samples was derived using the program apt-geno-qc (directly from the CEL files) and also by examining chrX and chrY.

Samples were required to have ≥95% call rate across all SNPs before entering the QC process. This threshold was also imposed post SNP QC resulting with the loss of 254 samples. Many studies would not impose such stringent QC, but using the 327 of the QIMR samples already been genotyped on an Illumina platform we were able to undertake additional checks not usually possible (***Table S1***, Stage 4). Discordance rates relative to the Illumina genotyping varied widely between samples (with a maximum of ~4.7% but a median of only ~0.13%), and imposing ≥95% call rate post QC eliminated 39 of the 45 most discordant samples.

Next, we merged the QIMR A6.0 genotypes with the full merged QIMR Illumina genotype data (1) (of which the I317, I370 and I610 QIMR sets used here are a subset) for the subset of ~138k SNPs which were overlapping and strand-unambiguous. Since family members of the unrelated individuals in the A6.0 sample had been genotyped on the Illumina platforms we used pairwise IBS analysis to verify that the genotyped individuals bore the expected relationship to their families. This identified two otherwise unknown sample mixups (a swap plus a mislabeled sample) and tested the quality of deliberate repeats. These checks also identified 20 pairs of related samples amongst the QIMR set: repeat genotypings (4 pairs representing multiple recruitments of the same individual); MZ (1 pair); siblings (7 pairs); parent/child (1 pair); avuncular (3 pairs) ; 1st cousins (4 pairs). All of the first degree relationships and multiple recruitments were already recorded in our database, and one member of each pair was dropped.

Additional samples were excluded if they were identified as gross ancestry outliers using Principal Component Analysis (PCA) when analysed together with genotype data from 16 global populations sourced from HapMap Phase 3 (HM3) (11 populations) and a previously published study of Northern European genetic diversity (GEUT- 5 populations) (5) using the same methods described in Medland et al (1). PCA was conducted using the autosomal SNPs that were genotyped in common between the A6.0 cases, HM3 and GEUT populations with the further proviso that SNP missing rates were < 2.5% in all individual cohorts and populations. A total of ~73K SNPs fulfilled these requirements. The EIGENSOFT package was used to conduct the PCA (6). Only those individuals in the 16 reference populations (n = 2317) were included in the initial PCA used to generate the top 10 Eigenvectors or Principal Components (PCs). The A6.0 cases were then projected onto this 'genetic space' background. The A6.0 case individuals cluster with Europeans as expected and the QIMR, NESDA/NTR and UoE samples cluster together. However, a few individuals show evidence of African or Asian ancestry. We calculated the mean and standard deviations for PC1 and PC2 of the non-Australian European reference and excluded any MDD2000 individuals more than 6 standard deviations from these means as ancestry outliers. A total of 15 QIMR, 7 NESDA/NTR and 5 UoE individuals that had survived QC were excluded as outliers.

Table S1 SNP and sample QC criteria for A6.0 case sample.

	SNPs removed	SNPs remaining
Stage 1		
No. SNPs called/genotyped		909,622
Monomorphic or MAF < 1%	145,911	
Not mapped reliably (includes Affymetrix proprietary probes)	4,049	
>5% missing	103,645	656,017
Stage 2		
Allele frequency difference on one plate vs rest ($P<10^{-6}$) removed only for the failing plate.	≤ 64 on each plate	
Stage 3		
$p< 10^{-6}$ for Hardy-Weinberg exact test, either across all samples and/or by plate.	519	
Minor allele frequency < 1%	71	
Positive controls on two or more plates disagreed with the consensus genotype for controls on the remaining plates	102	
Non-random genotypic failures as inferred by the flanking haplotypic background PLINK --mishap test ($P<10^{-10}$)	8090	647,235
Stage 4		
High discordance (>1% of individuals) for 268 individuals with Affymetrix contrast statistics ≥0.95 with an Illumina platform (see ii below) (strand-unambiguous SNPs genotyped on both platforms)	579	
Allele frequency differences ($P< 10^{-8}$) between the QIMR cases genotyped on Affymetrix vs 1205 unrelated cases genotyped on Ilumina platform (for same 137761 SNPs tested previously)	0	
Strand ambiguity, AT or CG SNPs with allele frequency close to 0.5	55	**646,601**

a: 646,601 SNPs comprised 621,111 autosomal, 25347 X and 143 Y chromosome SNPs
Following all QC procedures 646,601 SNPs for 1001 QIMR, 139 NESDA/NTR and 375 UoE MDD cases were included in the next stage of analysis.

Genotyping and initial quality control for the A6.0 control (MGS) sample

A6.0 controls comprised samples screened negative for major psychiatric disorders as part of the Molecular Genetics of Schizophrenia (MGS) study genotyped on the Affymetrix 6.0 at the Broad Institute Center for Genotyping and Analysis in two batches. Permission to access the genotypes was granted from the data access committees of the NIMH (GENRED controls) and dbGAP (GAIN schizophrenia controls) repositories. Only the samples (1636 controls) surviving the QC steps described in Shi et al (7) were used.

Statistical power

For a complex disease with lifetime population prevalence of *K*, assume a causal variant having two alleles (A and a) with frequencies of *p* and (1-*p*) . Let $(1-p)^2$, $2p(1-p)$ and p^2 be the frequencies of genotypes aa, Aa and AA (in Hardy-Weinberg equilibrium), with risks of f_0, f_1 and f_2. If we assume a multiplicative model then $f_1 = f_0 \lambda$ and $f_2 = f_0 \lambda^2$ where λ is the relative risk with respect to the causal variant. Let p_{case} and $p_{control}$ be the frequency of allele A in cases and controls, respectively, defined so that $Kp_{case} + (1-K)\, p_{control} = p$,

$p_{case} = \frac{p\lambda}{1+p(\lambda-1)}$ and $p_{control} = \frac{p}{1-K}\left(1 - \frac{K\lambda}{1+p(\lambda-1)}\right)$.

If the number of cases and controls in the sample are N_{case} and $N_{control}$, respectively then $N = N_{case} + N_{control}$ and $v = N_{case}/N$. The non-centrality parameter (NCP) of the X^2 test of association is

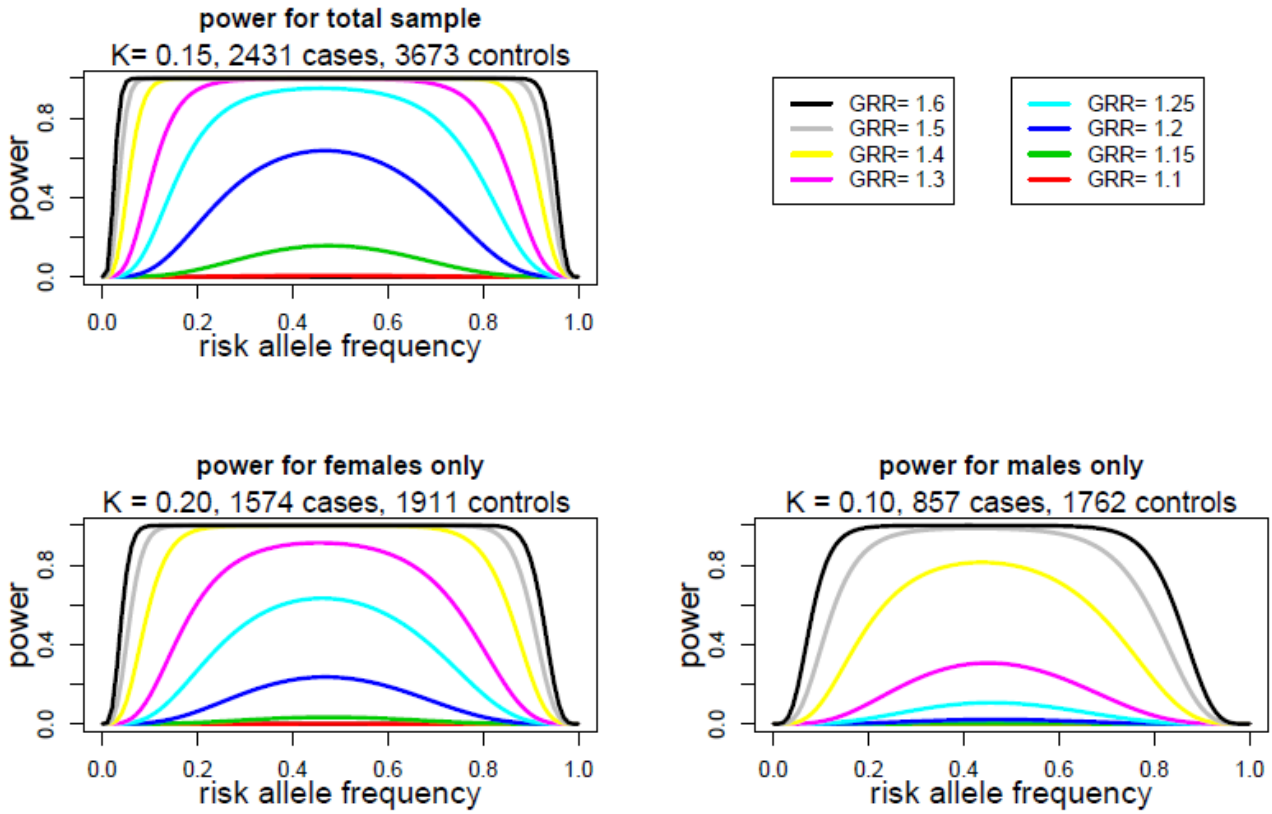
$$\text{NCP} = \frac{N^2(p_{case}-p_{control})^2}{Var(\hat{p}_{case}-\hat{p}_{control})} = \frac{2Nv(1-v)(p_{case}-p_{control})^2}{\bar{p}(1-\bar{p})} = \frac{2Nv(1-v)p^2(1-p)^2(\lambda-1)^2}{(1+p(\lambda-1))^2(1-K)^2\bar{p}(1-\bar{p})}, \quad [1]$$

[1]

where $\bar{p} = v\, p_{case} + (1-v)\, p_{control}$

We calculate power as the normal probability p(Z > T+√NCP), where Z ~N(0,1) and T is the normal deviate corresponding to the type I probability level, ie 5e-8 for genome-wide association. Power curves are presented in FigureS1. The estimated power is from equation [1], the same as from the Genetic Power Calculator(8).

Figure S1. Power curves for the total, male and female only study samples for different genotype relative risks (GRR= λ).



Comparing the power of case control studies of MDD and schizophrenia (SCZ)

From equation [1] we can write a NCP for the association test of a case-control study of MDD or SCZ. We assume both have equal proportions of cases in their samples (*v*), and the associated variant for each disorder has frequency, *p*.

$$NCP_{SCZ} = \frac{2N_{SCZ}v(1-v)p^2(1-p)^2(\lambda_{SCZ}-1)^2}{(1+p(\lambda_{SCZ}-1))^2(1-K_{SCZ})^2\bar{p}_{SCZ}(1-\bar{p}_{SCZ})}$$

and

$$NCP_{MDD} = \frac{2N_{MDD}v(1-v)p^2(1-p)^2(\lambda_{MDD}-1)^2}{(1+p(\lambda_{MDD}-1))^2(1-K_{MDD})^2\bar{p}_{MDD}(1-\bar{p}_{MDD})}$$

Following Yang et al (9), under the liability threshold model, we consider variants which explain the same proportion (q^2) of phenotypic variance on the liability scale so that

$$q^2 = 2p(1-p)\frac{(\lambda_{MDD}-1)^2}{i_{MDD}^2} = 2p(1-p)\frac{(\lambda_{SCZ}-1)^2}{i_{SCZ}^2}$$

where i_{SCZ} *and* i_{MDD} are the mean phenotypic liability of individuals with SCZ and MDD respectively, calculated as $i = z/K$ where z is the height of the standard normal curve at the truncation of proportion *K*. Therefore,

$$\lambda_{MDD} = 1 + \frac{i_{MDD}}{i_{SCZ}}(\lambda_{SCZ}-1)$$

i.e. a risk variant for schizophrenia $\lambda_{SCZ} = 1.20$ explains the same proportion of variance in liability as a risk variant of equal frequency in the population for MDD of $\lambda_{MDD} = 1.11$, assuming disease prevalence, K_{SCZ} = 0.007 and K_{MDD} = 0.15 so that i_{SCZ} = 2.78 and i_{MDD} = 1.55. The increase in sample size for a case control study for MDD to detect a risk variant which explains the same proportion of the variance in liability

$$\frac{N_{MDD}}{N_{SCZ}} = \frac{(\lambda_{SCZ}-1)^2}{(\lambda_{MDD}-1)^2}\frac{(1-K_{MDD})^2}{(1-K_{SCZ})^2}\frac{(1+p(\lambda_{MDD}-1))^2}{(1+p(\lambda_{SCZ}-1))^2}\frac{\overline{p}_{MDD}(1-\overline{p}_{MDD})}{\overline{p}_{SCZ}(1-\overline{p}_{SCZ})}$$

$$= \frac{i_{SCZ}^2}{i_{MDD}^2}\frac{(1-K_{MDD})^2}{(1-K_{SCZ})^2}\frac{(1+p\,\frac{i_{MDD}}{i_{SCZ}}(\lambda_{SCZ}-1))^2}{(1+p(\lambda_{SCZ}-1))^2}\frac{\overline{p}_{MDD}(1-\overline{p}_{MDD})}{\overline{p}_{SCZ}(1-\overline{p}_{SCZ})} \qquad [2]$$

$$\approx \frac{i_{SCZ}^2}{i_{MDD}^2}\frac{(1-K_{MDD})^2}{(1-K_{SCZ})^2} \qquad [3]$$

The increased sample size required for MDD to detect a variant that explains the same proportion of phenotypic liability as a variant for SCZ is demonstrated using equation [2] in Figure S2 (assuming both studies have equal proportion of cases). Using the approximation of equation [3] $\frac{N_{MDD}}{N_{SCZ}} \approx 2.4$.

Hospital-based MDD cohorts may represent a more extreme phenotype, with both lower prevalence and higher heritability (10). Using a prevalence for such clinical samples to be K_{MDD} = 0.06 (the average across sexes (11)) still requires a sample size ~1.8 times (equation [3])greater for a case control study of MDD compared to one for schizophrenia.

This comparison only accounts for the difference in prevalence rates and hence the lower difference in mean liability between cases (*i*) and controls (-iK/(1-K)) i.e. , K_{SCZ} = 0.007 and K_{MDD} = 0.15 the difference in mean liability between cases and controls is 2.78 s.d units for SCZ and 1.57 s.d. units for MDD. If we assume that the number and frequency of risk variants underlying SCZ and MDD is the same then the difference in heritability must reflect lower effect sizes in MDD. In

this case the ratio of NMDD/NSCZ can be derived by equating the NCP on equal proportions of genetic variance explained (i.e. scaling q2 by the heritability) so that equation [3] is inflated by the ratio of heritabilities $\frac{h^2_{SCZ}}{h^2_{MDD}}$, which may be as great as a factor of 2, ie sample sizes 4-5 times those for schizophrenia may be needed for MDD.

Figure S2 Ratio of sample size for a case-control study for MDD compared to one for SCZ assuming variants of equal population frequency that explain an equal proportion of the phenotypic variance.

A sample size of ~2.4 times greater (equation [3]) is needed for MDD compared to to detect variants that explain the same proportion of variance in *phenotypic* liability and that have the same frequency in the total population, expressed in terms of genotypic relative risk in SCZ (GRR = λ_{SCZ}).

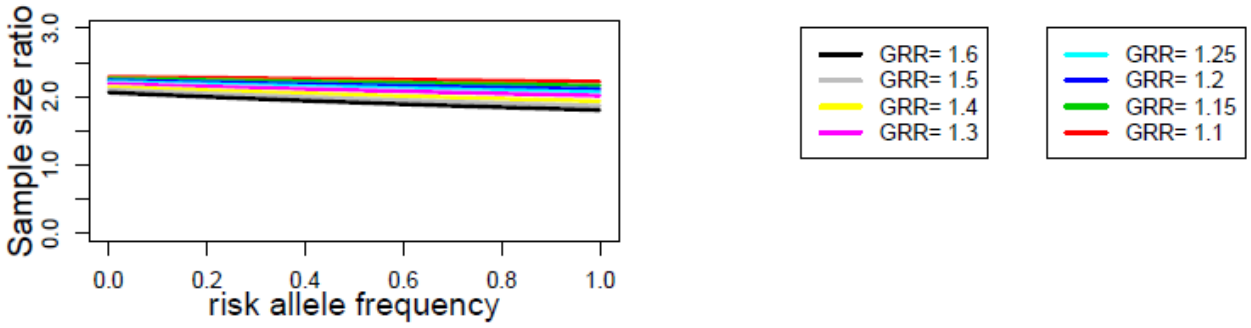


Figure S3 Manhattan Plot for full analysis of 2431 major depression cases and 3673 controls.

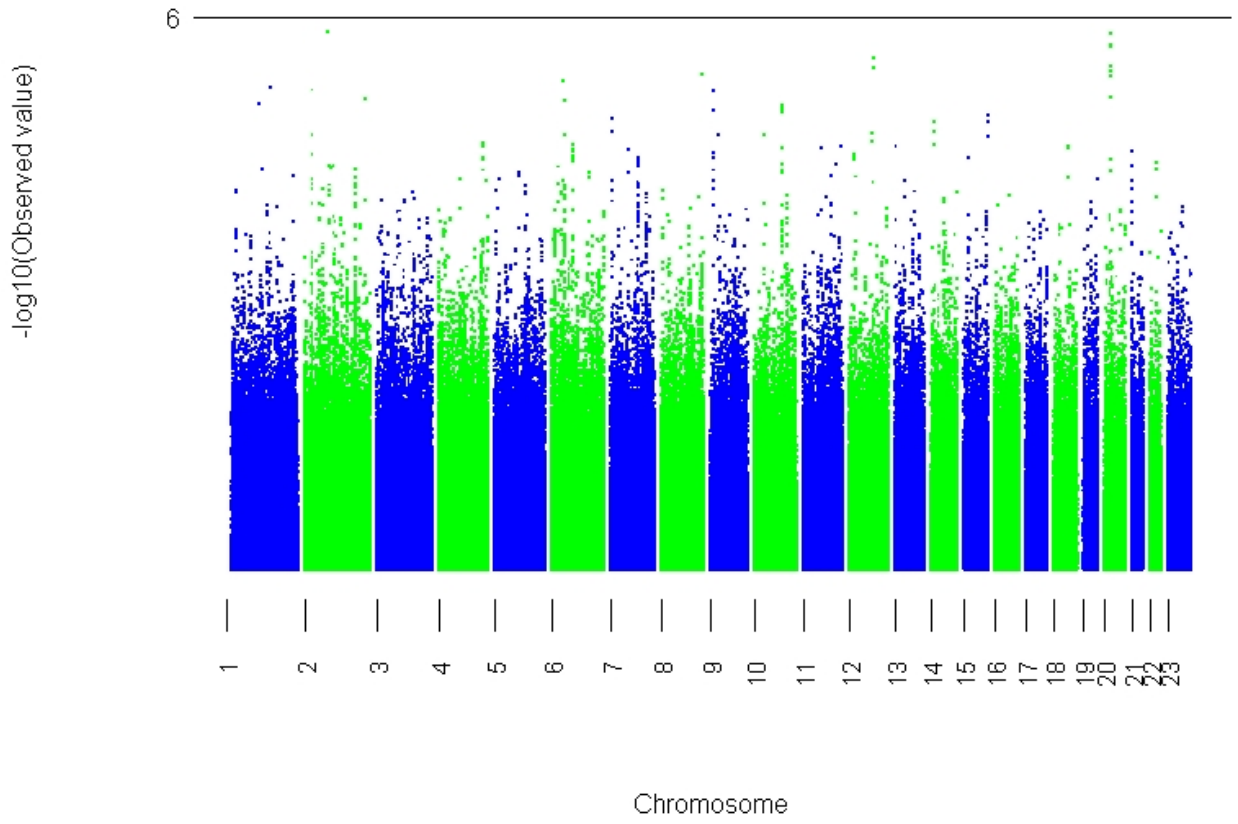


Figure S4 QQ plot from association of i) male only and ii) female only analyses

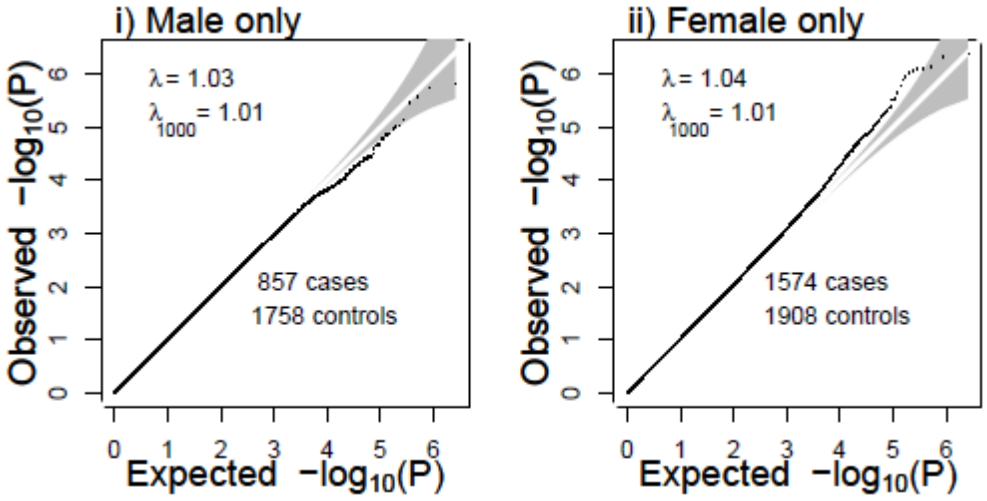


Table S2 SNPs in the PCLO region investigated by analysis set and sample source.

The gene *PCLO* gene, a novel but plausible candidate, identified in the GAIN MDD study (1) located on chromosome 7q11.23-q21.3 and bounded by two other plausible candidate genes *CACNA2D1* and *SEMA3E* . The most associated *PCLO* SNPs were rs2715148 and the non-synonymous rs2522833.
The most associated *PCLO* SNPs were rs2715148 and the non-synonymous rs2522833. The GAIN MDD sample comprises cases and controls from the Dutch NESDA and NTR studies, the same studies which contribute 127 cases and 577 controls to MDD2000+.
In addition, two of the replication samples comprised cases and controls from UoE and QIMR, the latter showing support for association across the region with p = 0.076 and 0.028 for rs2715148 and rs2522833, respectively.
Here we explore the 16 SNPs in our genotype set that were among the 30 SNPs considered in the GAIN MDD replication study, investigating allele frequency and association by analysis set and sample source.
This investigation shows that the genotyping results here are consistent with those presented in Sullivan et al (2009) for QIMR samples, but that the choice of different QIMR study participants
(overlap with MDD2000+ of 562 cases and 264 controls), the ascertainment of QIMR samples reflected in the summary statistics of Table 2 in the main text, and the combining of samples from different sources results in the lack of association in the total MDD2000+ sample.

		QIMR sample (Sullivan et al)			MDD2000+			A6.0									I317 (QIMR)			I370				I610 (QIMR)				ALL
SNP	Associated allele FWD strand	Freq controls [N = 1039]	Freq cases [N = 969]	P	Freq Cont [N=3673]	Freq Case [N=2431]	P	Freq Cont (MGS) [N=1636]	Freq All Case [N=1441]	Freq QIMR case [N=941]	Freq UoE Case [N=373]	Freq NESDA/ NTR Case [N=127]	P All case	P QIMR case	P UoE Case	P NESDA/ NTR case	Freq Cont [N=237]	Freq Case [N=-84]	P	Freq All Cont [N=1372]	Freq QIMR Cont [N=795]	Freq Case (QIMR) [N=737]	P	Freq Contr [N=428]	Freq Case [N=169]	p	P QIMR
rs7780196	G	0.41	0.46	**3.2E-03**	0.44	0.43	2.7E-01	0.43	0.44	0.44	0.43	0.44	1.2E-01	1.0E-01	2.9E-01	6.3E-01	0.42	0.37	1.9E-01	0.45	0.46	0.41	**1.9E-03**	0.45	0.44	6.9E-01	**7.0E-04**
rs10954689	T	0.44	0.49	**3.4E-03**	0.47	0.47	5.5E-01	0.45	0.48	0.49	0.46	0.46	8.2E-02	**2.6E-02**	6.8E-01	7.0E-01	0.48	0.47	9.3E-01	0.48	0.48	0.46	2.9E-01	0.48	0.46	4.5E-01	2.9E-01
rs12672552	T	0.89	0.90	7.4E-01	0.89	0.89	4.4E-01	0.89	0.89	0.89	0.91	0.88	9.7E-01	6.2E-01	1.6E-01	5.9E-01	0.87	0.86	6.1E-01	0.90	0.91	0.89	4.1E-01	0.91	0.90	5.8E-01	2.4E-01
rs6948464	C	0.64	0.65	2.9E-01	0.65	0.65	8.8E-01	0.65	0.65	0.66	0.64	0.64	8.1E-01	7.4E-01	3.3E-01	8.2E-01	0.66	0.65	7.4E-01	0.64	0.65	0.64	9.7E-01	0.66	0.65	7.5E-01	5.5E-01
rs13227462	G	0.95	0.95	7.9E-01	0.94	0.94	4.0E-01	0.94	0.95	0.94	0.95	0.96	**1.3E-02**	1.3E-01	**3.9E-02**	**3.6E-02**	0.94	0.93	7.5E-01	0.95	0.94	0.94	9.2E-02	0.94	0.94	9.1E-01	3.6E-01
rs6979066	A	0.33	0.37	**6.7E-03**	0.35	0.35	9.9E-01	0.34	0.36	0.37	0.36	0.34	7.9E-02	5.4E-02	2.4E-01	6.2E-01	0.33	0.32	8.2E-01	0.37	0.37	0.34	7.3E-02	0.37	0.34	2.7E-01	**3.8E-02**
rs6965452	G	0.84	0.86	**4.4E-02**	0.85	0.85	3.2E-01	0.84	0.86	0.86	0.85	0.87	**3.2E-03**	**8.2E-03**	8.7E-02	3.0E-01	0.84	0.83	7.2E-01	0.86	0.85	0.84	5.9E-02	0.84	0.84	9.6E-01	2.5E-01
rs12668093	C	0.85	0.86	6.8E-01	0.85	0.85	3.2E-01	0.85	0.85	0.85	0.87	0.82	6.7E-01	5.1E-01	3.4E-01	1.7E-01	0.82	0.84	7.6E-01	0.86	0.87	0.86	5.5E-01	0.87	0.84	2.0E-01	4.3E-01
rs2522833	C	0.41	0.45	**2.5E-02**	0.43	0.44	5.1E-01	0.42	0.44	0.45	0.42	0.43	**4.9E-02**	**1.2E-02**	8.4E-01	7.6E-01	0.43	0.42	9.4E-01	0.44	0.45	0.42	2.0E-01	0.45	0.43	5.2E-01	1.7E-01
rs13233504	T	0.20	0.21	2.7E-01	0.21	0.21	8.9E-01	0.21	0.21	0.22	0.20	0.19	9.8E-01	5.2E-01	8.6E-01	1.7E-01	0.19	0.21	4.3E-01	0.21	0.21	0.21	7.3E-01	0.21	0.20	9.5E-01	6.5E-01
rs2888018	G	0.32	0.33	5.7E-01	0.33	0.33	4.9E-01	0.33	0.34	0.34	0.31	0.34	5.9E-01	2.1E-01	3.7E-01	7.4E-01	0.33	0.33	9.4E-01	0.33	0.32	0.32	9.9E-01	0.31	0.34	2.7E-01	9.8E-01
rs10954694	C	0.35	0.37	1.9E-01	0.37	0.37	9.7E-01	0.35	0.38	0.38	0.38	0.34	8.1E-02	5.6E-02	1.6E-01	4.7E-01	0.35	0.32	4.2E-01	0.38	0.39	0.36	2.2E-01	0.40	0.35	1.0E-01	1.5E-01
rs9690648	T	0.93	0.93	9.7E-01	0.93	0.94	4.4E-01	0.93	0.93	0.93	0.94	0.91	6.9E-01	8.0E-01	4.4E-01	2.7E-01	0.93	0.96	1.6E-01	0.94	0.94	0.94	4.9E-01	0.93	0.92	3.6E-01	3.5E-01
rs6959723	A	0.81	0.82	4.8E-01	0.81	0.81	7.3E-01	0.80	0.82	0.82	0.80	0.82	6.6E-02	**4.0E-02**	6.0E-01	6.4E-01	0.82	0.77	2.4E-01	0.83	0.82	0.80	**3.3E-02**	0.81	0.80	8.7E-01	8.4E-02
rs7799260	G	0.52	0.53	4.2E-01	0.52	0.52	9.3E-01	0.51	0.53	0.54	0.50	0.52	3.1E-01	7.5E-02	6.2E-01	8.6E-01	0.52	0.46	1.8E-01	0.53	0.53	0.51	5.4E-01	0.54	0.52	4.2E-01	3.9E-01
rs12669254	T	0.91	0.92	3.3E-01	0.91	0.91	3.4E-01	0.91	0.91	0.91	0.93	0.90	6.5E-01	7.2E-01	**2.1E-02**	4.7E-01	0.91	0.90	6.8E-01	0.92	0.92	0.91	8.3E-02	0.92	0.91	5.3E-01	6.3E-02

16 out of the 30 SNPs used in the replication study undertaken Sullivan et al (2009) were included in our final imputed SNP set.
562 MDD cases and 264 controls were in common between the QIMR replication sample used in Sullivan et al (2009) and MDD2000+, of which 404 MDD cases were included in the A6.0 analysis set.
For SNPs directly genotyped in MDD2000+ the maximum discordance in genotypes was 2.
In Sullivan et al (2009) UoE cases vs UoE controls showed association of p = 4.0E-03 and p =7E-02 for SNPs rs13227462 and rs 12669254, respectively; these were the only 2 SNPs of the 16 to have p < 0.05 in the Sullivan et al (2009) paper for UoE samples
The lack of support for PCLO by the total MDD2000+ does not imply any inconsistency with the results reported in Sullivan et al (2009).

Table S3 Regions containing at least on SNP with a p-value of $p<10^{-5}$ in the meta-analysis of associations from the MDD2000+, GAIN and UK samples.

					Frequency	OR				P				
CHR	SNP	BP	A1	A2	MDD2000+	MDD2000+	GAIN	UK	Meta	MDD2000+	GAIN	UK	Meta	Gene +/- 50kb
1	rs7513908	8225206	A	G	0.17	1.18	1.11	1.18	1.16	1.2E-03	1.2E-01	1.4E-02	1.9E-05	
1	rs12407717	30198601	T	C	0.12	1.18	1.11	1.25	1.17	5.7E-03	1.4E-01	3.6E-03	4.0E-05	
1	rs11579964	222605563	T	C	0.16	0.84	0.84	0.84	0.84	3.5E-03	1.4E-02	1.0E-02	4.4E-06	*CNIH4 NVL WDR26*
3	rs581190	62821149	A	G	0.44	1.16	1.09	1.08	1.11	2.5E-04	9.0E-02	1.5E-01	4.3E-05	*CADPS*
3	rs12714788	72739803	C	A	0.27	0.87	0.84	0.96	0.88	1.1E-03	1.8E-03	4.9E-01	2.6E-05	
3	rs7647854	186359477	G	A	0.16	1.22	1.20	1.12	1.19	5.0E-04	7.4E-03	1.1E-01	4.6E-06	*C3orf70, EHHADH*
4	rs1826690	70386855	G	A	0.25	0.96	0.82	0.80	0.88	4.3E-01	8.1E-04	1.4E-04	1.9E-05	
5	rs1990950	156853334	T	G	0.40	1.09	1.17	1.10	1.12	2.5E-02	2.4E-03	6.5E-02	4.9E-05	*UGT2B4*
6	rs6568842	114907070	A	G	0.15	1.19	1.13	1.17	1.17	1.0E-03	8.4E-02	3.6E-02	2.6E-05	*ADAM19,ICHTHYIN*
10	rs7100942	25863759	A	C	0.43	1.12	1.09	1.13	1.11	4.2E-03	8.3E-02	1.8E-02	4.8E-05	*GPR158*
16	rs11075236	14372157	C	A	0.34	1.09	1.14	1.16	1.12	3.4E-02	1.4E-02	5.6E-03	3.3E-05	
16	rs3852700	65829359	C	T	0.07	1.28	1.19	1.22	1.24	1.5E-03	6.9E-02	4.4E-02	3.7E-05	*E2F4, ELMO3, EXOC3L, FHOD1, LRRC29, SLC9A5,TMEM208*
16	rs12446956	72059037	C	T	0.13	1.22	1.15	1.28	1.22	6.4E-04	6.1E-02	2.0E-03	1.1E-06	*SYT4*
18	rs12457996	39126271	C	T	0.23	0.88	0.88	0.84	0.87	5.3E-03	2.3E-02	4.5E-03	5.7E-06	
18	rs9951150	50972122	G	A	0.45	1.12	1.08	1.15	1.12	5.0E-03	1.0E-01	5.0E-03	2.6E-05	
19	rs2116877	11219440	C	T	0.11	0.86	0.85	0.79	0.84	1.6E-02	4.3E-02	4.4E-03	2.9E-05	*DOCK6,LOC55908, TSPAN16*
20	rs1539470	59771861	C	A	0.21	0.91	0.83	0.89	0.88	4.2E-02	1.5E-03	4.8E-02	4.3E-05	
22	rs5755867	34403989	C	T	0.04	1.51	1.32	1.17	1.36	9.2E-05	5.5E-02	2.5E-01	2.3E-05	*APOL5,APOL6*
23	rs17282946	120144590	A	G	0.05	0.78	0.83	0.78	0.79	9.4E-04	1.4E-01	4.7E-02	4.0E-05	

a: INFO =0.7; all other INFO statistics for MDD2000 and GAIN > 0.8

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