

Multi-omic strategies for transcriptome-wide prediction and association studies

Supplementary Materials

1 Supplementary methods

1.1 Asymptotic test of total mediation effect

In DePMA, a distal-eQTL s is tested for its total mediation effect on gene G through m mediators that are local to s . Consider the following mediation model for $1 \leq j \leq m$:

$$\begin{aligned} Y_G &= X_s \beta_s + \mathbf{M} \beta_{\mathbf{M}} + \mathbf{X}_C \beta_C + \varepsilon_{Y_G} \\ M_j &= X_s \alpha_{M_j} + \mathbf{X}_C \alpha_{C,j} + \varepsilon_{M_j} \end{aligned} \tag{1}$$

Here, we construct the total mediation effect

$$\text{TME} = \alpha_{\mathbf{M}}^T \beta_{\mathbf{M}} = \sum_{i=1}^m \alpha_{M_i} \beta_{M_i}.$$

Note that TME is distributed as the product of two multivariate Normal distributions. By the multivariate Delta method¹, we can obtain the standard error for the estimated TME. Let $\boldsymbol{\theta} = (\alpha_{\mathbf{M}}, \beta_{\mathbf{M}})$ and define $f(\boldsymbol{\theta}) = \text{TME} = \sum_{i=1}^m \alpha_{M_i} \beta_{M_i}$.

The first order partial derivative of $f(\hat{\boldsymbol{\theta}})$ is

$$d_{\hat{\boldsymbol{\theta}}} = \frac{\partial(\sum_{i=1}^m \alpha_{M_i} \beta_{M_i})}{\partial \hat{\boldsymbol{\theta}}} = [\beta_{\mathbf{M}} \ \alpha_{\mathbf{M}}]^T.$$

We also obtain the estimated variance-covariance matrix $\hat{\boldsymbol{\Sigma}}$ of $\hat{\boldsymbol{\theta}}$:

$$\hat{\boldsymbol{\Sigma}} = \begin{bmatrix} \hat{\boldsymbol{\Sigma}}_{\alpha_{\mathbf{M}}} & \hat{\boldsymbol{\Sigma}}_{\alpha_{\mathbf{M}}\beta_{\mathbf{M}}} \\ \hat{\boldsymbol{\Sigma}}_{\alpha_{\mathbf{M}}\beta_{\mathbf{M}}} & \hat{\boldsymbol{\Sigma}}_{\beta_{\mathbf{M}}} \end{bmatrix},$$

where $\hat{\Sigma}_{\alpha_{\mathbf{M}}}$, $\hat{\Sigma}_{\beta_{\mathbf{M}}}$, and $\hat{\Sigma}_{\alpha_{\mathbf{M}}\beta_{\mathbf{M}}}$ are the variances and covariance of $\hat{\alpha}_{\mathbf{M}}$, $\hat{\beta}_{\mathbf{M}}$, and between $\hat{\alpha}_{\mathbf{M}}$ and $\hat{\beta}_{\mathbf{M}}$, respectively. Sobel previously has shown, that with sufficient sample size, $\hat{\Sigma}_{\alpha_{\mathbf{M}}\beta_{\mathbf{M}}} \approx 0^{2,3}$. Thus, the standard error of $\hat{\theta}$ is given by

$$\hat{\sigma}_{\hat{\theta}}^2 = d_{\hat{\theta}}^T \hat{\Sigma} d_{\hat{\theta}}.$$

We can then test $H_0 : \text{TME} = 0$ against $H_1 : \text{TME} \neq 0$ with the two-sided Wald-type test with the test statistic $Z = \frac{\alpha_{\mathbf{M}}^T \beta_{\mathbf{M}}}{\sqrt{\hat{\sigma}_{\hat{\theta}}^2}}$ and comparing to the null standard Normal distribution.

We illustrate the trade-off between power and computational speed using the asymptotic Sobel test and the permutation speed. Consider the following simulation framework with $m = 5$ mediators, 3 covariates and a sample size of $n \in \{200, 500, 700, 1000\}$ for the model in Equations 1:

- an n -length genotype vector for SNP s is drawn from $\text{Binomial}(2, MAF)$, where the minor allele frequency MAF is set at 0.1 in **Supplemental Figure 1** below;
- Under the alternative, we simulated $\beta_X \sim N(0, 1)$, $\beta_{\mathbf{M}} \sim \mathbf{N}_5(\mathbf{0}, \mathbf{I}_5)$, $\beta_C \sim \mathbf{N}_3(\mathbf{0}, \mathbf{I}_3)$, $\alpha_{M_j}|_{j=1}^{m=5} \sim N(0, 1)$, $\alpha_C \sim \mathbf{N}_5(\mathbf{0}, \mathbf{I}_5)$.
- Under the null, all regression parameters were simulated as in the alternative case. However, we set $\alpha_{M_j} = 0|_{j=1}^m$ and $\beta_{\mathbf{M}} = \mathbf{0}$.
- Lastly, $\varepsilon_{Y_G} \sim N(0, 1 - h^2)$ and $\varepsilon_{M_j} \sim N(0, 1 - h_M^2)$, where $h^2 = h_M^2 = 0.1$ in **Supplemental Figure 1** below.
- We then constructed Y_G and \mathbf{M} using Equations 1.

We found, that over 10,000 simulations, the permutation test was considerably more powerful, albeit considerably slower. However, in most cases of implementing DePMA, the number of tests of mediation are usually on the order of 10^1 to 10^2 . We recommend the permutation test in most cases, unless gene G has thousands of identified distal-eQTLs. Parallel implementations have been offered as options in the MOSTWAS package.

1.2 Added-last test of distal-SNPs

Here, we propose a test to assess the information added from distal-eSNPs in the weighted burden test beyond what we find from local SNPs. Let \mathbf{Z}_l (an n_l -vector) and \mathbf{Z}_d (an n_d -vector) be the Z -scores local and distal SNPs identified by a MOSTWAS model, with $\mathbf{Z} = [\mathbf{Z}_l \ \mathbf{Z}_d]^T$ (an n vector). The local and distal SNP effects from the MOSTWAS model are represented in \mathbf{w}_l (an n_l -vector) and \mathbf{w}_d (an n_d -vector), with $\mathbf{w} = [\mathbf{w}_l \ \mathbf{w}_d]^T$ (an n vector). Here, we are interested in testing

$$H_0 : \mathbf{w}_d^T \mathbf{Z}_d | \mathbf{w}_l^T \mathbf{Z}_l = \tilde{Z}_{l,\text{obs}} = 0,$$

where $\tilde{Z}_{l,\text{obs}}$ is the observed weighted Z -score from local SNPs.

Under the null distribution, as proposed by Pasaniuc et al and Gusev et al in the Imp-G framework^{4,5}, we assume that $\mathbf{Z} \sim N_n(\mathbf{0}, \mathbf{\Sigma})$, where

$$\mathbf{\Sigma} = \begin{bmatrix} \mathbf{\Sigma}_l & \mathbf{\Sigma}_{l,d} \\ \mathbf{\Sigma}_{l,d}^T & \mathbf{\Sigma}_d \end{bmatrix}$$

is the LD matrix for the SNPs, as estimated from the reference panel. $\mathbf{\Sigma}_l$ and $\mathbf{\Sigma}_d$ represent the LD matrices for local and distal SNPs, respectively. The LD matrix between local and distal SNPs $\mathbf{\Sigma}_{l,d}$ can be assumed to be zero, though recent studies have showed long-range LD in the human genome^{6,7}. We allow the user to set cross-chromosomal LD to 0, though by default, we estimate LD from the reference panel.

Now, we see that, under this null hypothesis, the joint distribution of $(\tilde{Z}_l, \tilde{Z}_d) = (w_l^T \mathbf{Z}_l, w_d^T \mathbf{Z}_d)$ is given by:

$$\begin{pmatrix} \tilde{Z}_l \\ \tilde{Z}_d \end{pmatrix} \sim N_2 \left(\mathbf{0}, \begin{bmatrix} w_l^T \mathbf{\Sigma}_l w_l & w_l^T \mathbf{\Sigma}_{l,d} w_d \\ w_d^T \mathbf{\Sigma}_{l,d}^T w_l & w_d^T \mathbf{\Sigma}_d w_d \end{bmatrix} \right).$$

It follows that, given $\tilde{Z}_l = \tilde{Z}_{l,\text{obs}}$,

$$\tilde{Z}_d | \tilde{Z}_l = \tilde{Z}_{l,\text{obs}} \sim N \left(\frac{w_l^T \mathbf{\Sigma}_{l,d} w_d}{w_l^T \mathbf{\Sigma}_l w_l} \tilde{Z}_{l,\text{obs}}, w_d^T \mathbf{\Sigma}_d w_d - \frac{[w_l^T \mathbf{\Sigma}_{l,d} w_d]^2}{w_l^T \mathbf{\Sigma}_l w_l} \right).$$

We can use this null distribution for the one-sided test of $H_0 : \mathbf{w}_d^T \mathbf{Z}_d | \mathbf{w}_l^T \mathbf{Z}_l = \tilde{Z}_{l,\text{obs}} = 0$ against $H_1 : \mathbf{w}_d^T \mathbf{Z}_d | \mathbf{w}_l^T \mathbf{Z}_l = \tilde{Z}_{l,\text{obs}} > 0$. These test is implemented in MOSTWAS as a follow-up to the weighted-burden test.

2 Supplemental Tables and Figures

	TCGA-BRCA	ROS/MAP
Local-only	0.037 (0.053)	0.079 (0.119)
MeTWAS	0.040 (0.066)	0.135 (0.099)
DePMA	0.383 (0.194)	0.405 (0.118)

Table 1: *Comparison of h^2 across local-only, MeTWAS, and DePMA predictive models.* The mean and standard deviation of h^2 across all genes that are significantly heritable with the genetic loci considered in the design matrix of each predictive model.

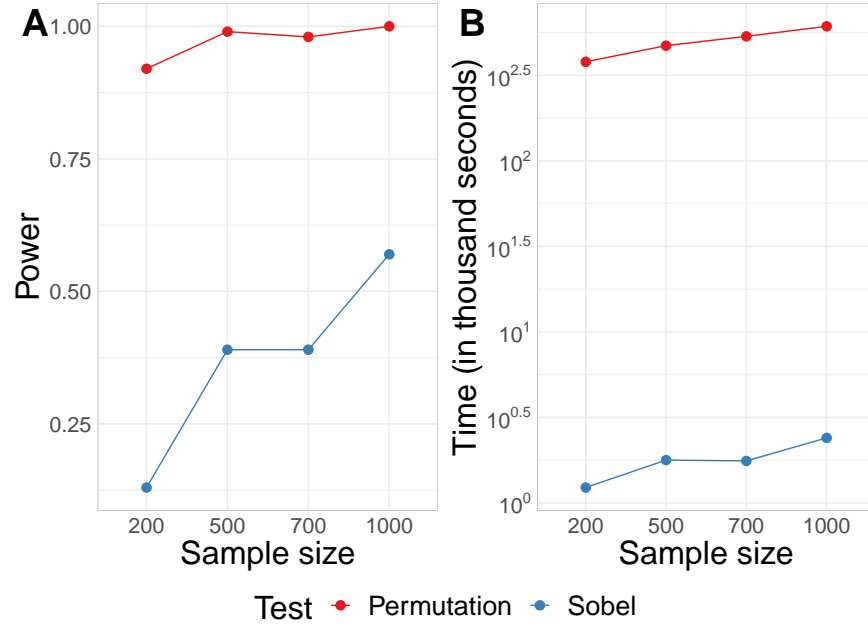


Figure 1: *Comparison of power and computational speed comparison of permutation and Sobel test.* Power (A) and computational speed (B) of permutation test (red) and asymptotic Sobel test (blue) in simulation framework

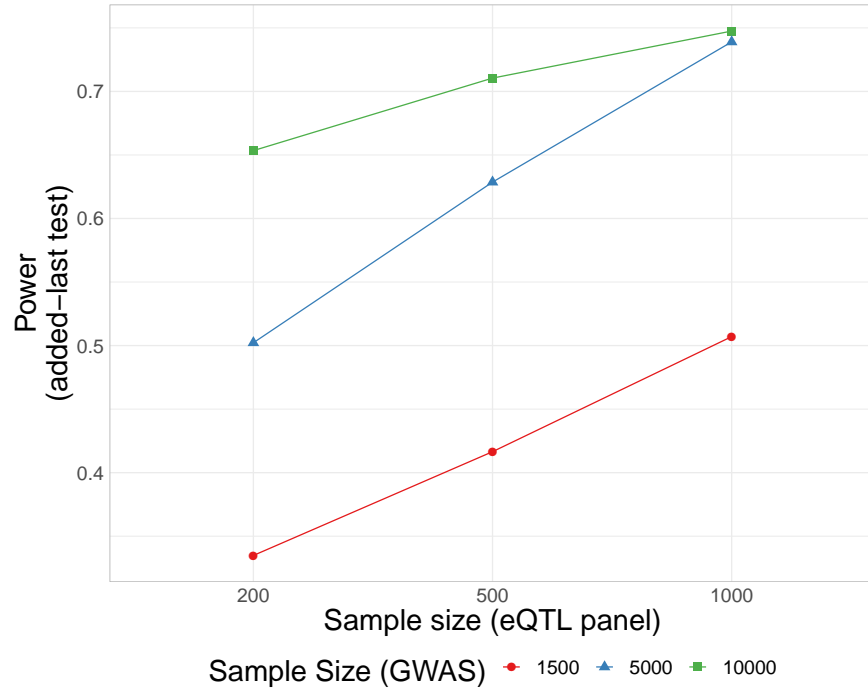


Figure 2: *Simulation analysis for the power of the distal variants added-last test.* Across various sample sizes for the eQTL reference (X -axis) panel and GWAS imputation panel (color), the power of the distal added-last test to detect a significant association with distal variants conditional on a significant local association at FDR-adjusted $P < 0.05$.

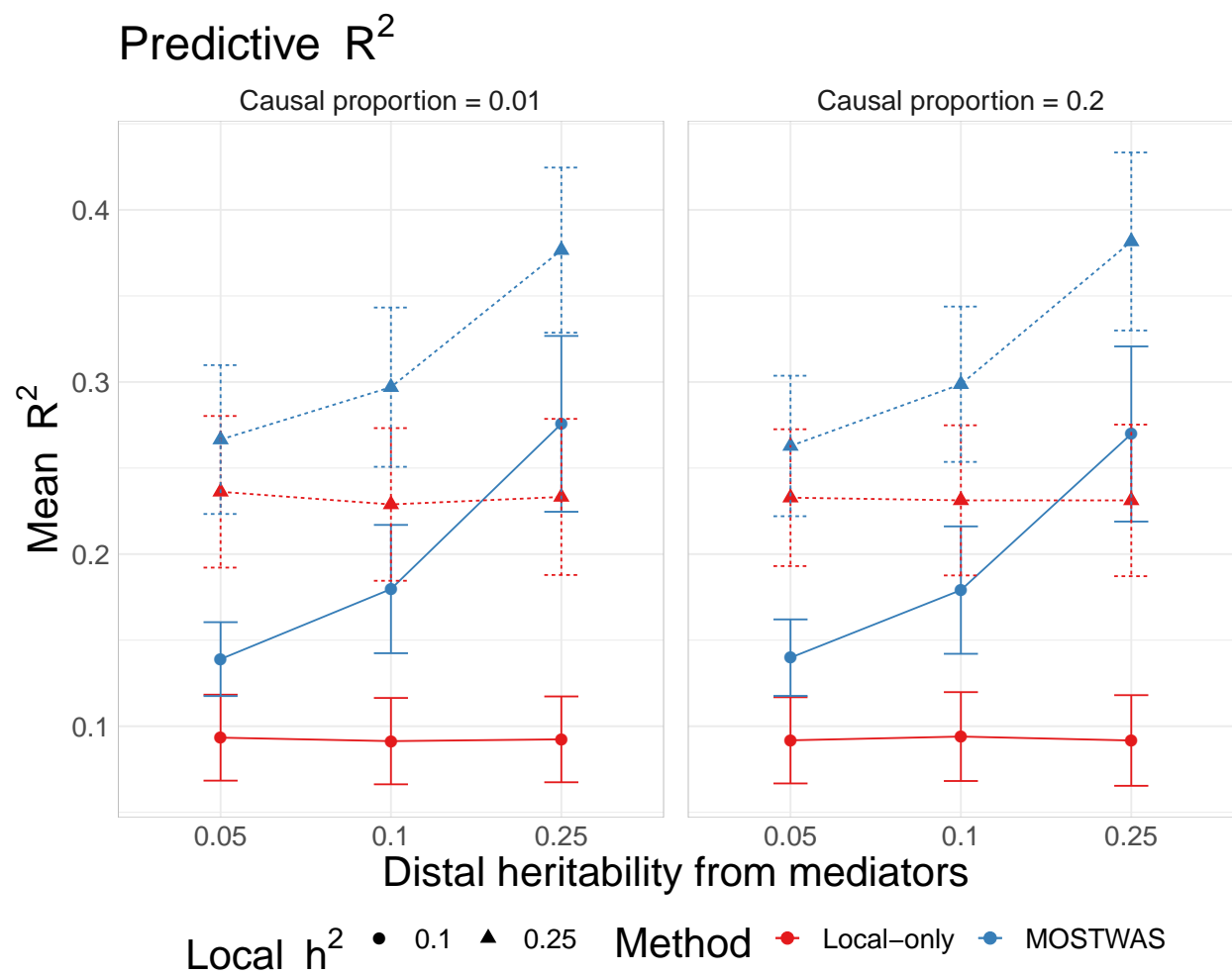


Figure 3: *Comparison of predictive R^2 in simulations.* Mean adjusted R^2 across various local and distal expression heritabilities, trait heritabilities, and causal proportions using local-only (red) and the best MOSTWAS (blue) models. The error bars reflect a width of 1 standard deviation of the 1,000 simulated adjusted R^2 values.

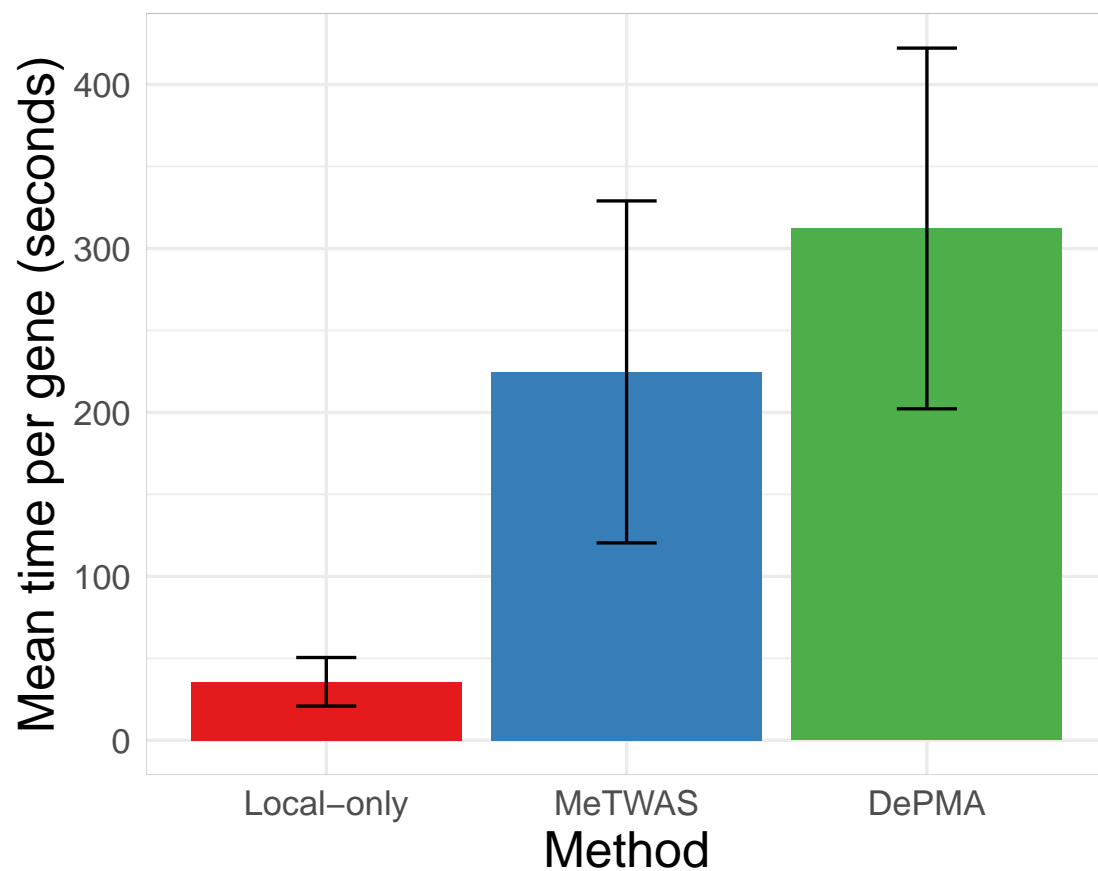


Figure 4: *Comparison of computation times between local-only and MOSTWAS modelling.* Mean and standard deviation of per-gene computation time across 50 randomly selected genes in TCGA-BRCA. Computations here were done with a 24-core, 3.0 GHz processor.

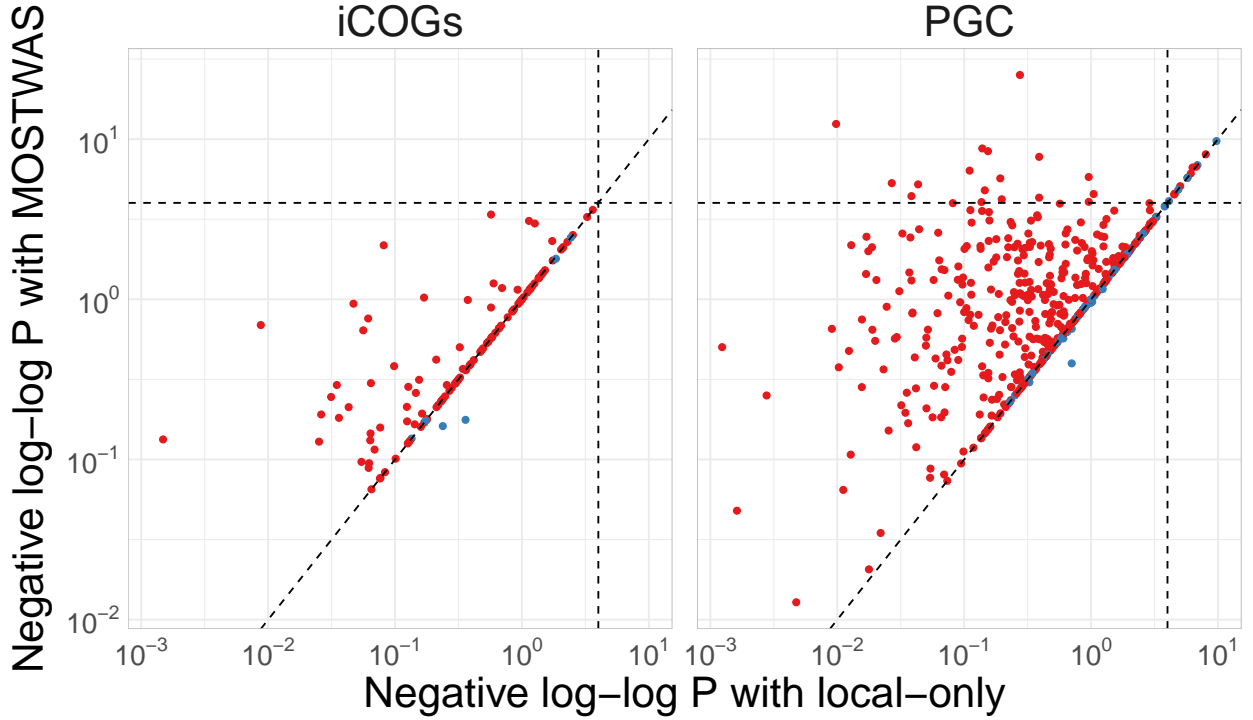


Figure 5: *Gene-trait associations in iCOGs and PGC using local-only and MOSTWAS models.* $-\log_{10}P$ -values of weighted burden gene-trait associations using iCOGs survival GWAS in European-ancestry women (left) and PGC MDD risk GWAS in predominantly European-ancestry patients (right) among genes that were predicted at cross-validation $R^2 \geq 0.01$ using both local-only and MOSTWAS models. The X - and Y -axes display the $-\log_{10}P$ -values for local-only and the best MOSTWAS model, respectively. Note that the scales of both axes are on a doubly logarithmic scale. Points are colored red if P -value of association is less than or equal using the MOSTWAS model. The horizontal and vertical reference lines indicate overall Bonferroni-corrected significance thresholds.

Gene	Cross-validation R^2	iCOGs Z-statistic (Added-last Z)	Top GWAS SNP location (P)	Permutation FDR-adjusted P
C16orf13	0.019	4.51 (5.18)	Chr3:10720351 (0.13)	0.03
C9orf169	0.011	4.18 (3.97)	Chr19:44949849 (0.04)	0.03
CTRL	0.051	4.51 (3.88)	Chr10:2798136 (0.06)	0.03
DNAL4	0.013	-3.94 (-4.57)	Chr22:38681840 (0.01)	0.04
LOC221710	0.014	5.34 (5.08)	Chr1:152983865 (8.5×10^{-4})	0.03
MAP3K6	0.021	-4.10 (-4.00)	Chr1:27686314 (0.01)	0.05
MAP4K5	0.020	3.76 (1.26)	Chr14:50502944 (1.3×10^{-4})	0.03
NPAT	0.115	-3.92 (-3.72)	Chr20:4217738 (0.02)	0.04
RPLP1	0.040	-3.82 (-3.83)	Chr18:1592917 (0.18)	1.4×10^{-4}
SPATA5L1	0.042	3.76 (No distal SNPs in model)	Chr15:45593323 (0.01)	1.4×10^{-4}
TXNRD2	0.047	3.91 (4.64)	Chr22:19735425 (3.5×10^{-3})	0.05

Table 2: *Summary statistics for 11 breast cancer-specific survival-associated loci identified by MOSTWAS models. TWAS associations with breast cancer survival from GWAS statistics from iCOGs with permutation test results and added-last Z-statistics. The top iCOGs GWAS SNP in the identified loci with its location and P-value are provided.*

Gene	Z-statistic (FDR-adjusted P)	Cross-validation R^2	TOP GWAS SNP location (P)	Permutation FDR-adjusted P	Added last FDR-adjusted P
ABCA7	-1.82 (0.09)	0.011	Chr19:553,066 (0.135)	NA	NA
ADAM10	-1.25 (0.23)	0.014	Chr15:59,052,072 (1.68×10^{-4})	NA	NA
APOE	2.82 (0.02)	0.119	Chr19:45,545,562 (3.0×10^{-5})	5.0×10^{-3}	0.03
BIN1	1.91 (0.08)	0.010	Chr22:24,199,787 (8.53×10^{-4})	NA	NA
CD2AP	1.52 (0.15)	0.011	Chr6:47,432,637 (1.23×10^{-4})	NA	NA
CLU	-2.41 (0.04)	0.012	Chr8:27,465,312 (1.33×10^{-4})	0.83	0.44
FERMT2	2.13 (0.06)	0.017	Chr14:53,305,626 (1.38×10^{-4})	NA	NA
MEF2C	2.20 (0.06)	0.016	Chr5:88,359,039 (0.020)	NA	NA
PLCG2	-2.48 (0.04)	0.010	Chr16:81,879,218 (0.037)	0.66	0.07
SORL1	2.91 (0.02)	0.043	Chr11:121,446,813 (0.032)	0.04	4.5×10^{-3}
ZCWPW1	-4.56 (6.1×10^{-5})	0.018	Chr7:100,435,157 (0.074)	0.03	1.3×10^{-5}

Table 3: *Summary statistics for known Alzheimer’s risk-associated loci identified by MOSTWAS models. TWAS associations (weighted Z-score and FDR-adjusted⁸ P -value) with late-onset Alzheimer’s risk from GWAS statistics from IGAP⁹. The top IGAP GWAS SNP in the identified loci with its location and P -value are provided. For the 6 loci with significant TWAS associations, the FDR-adjusted P -value for the follow-up distal SNP added last test is provided.*

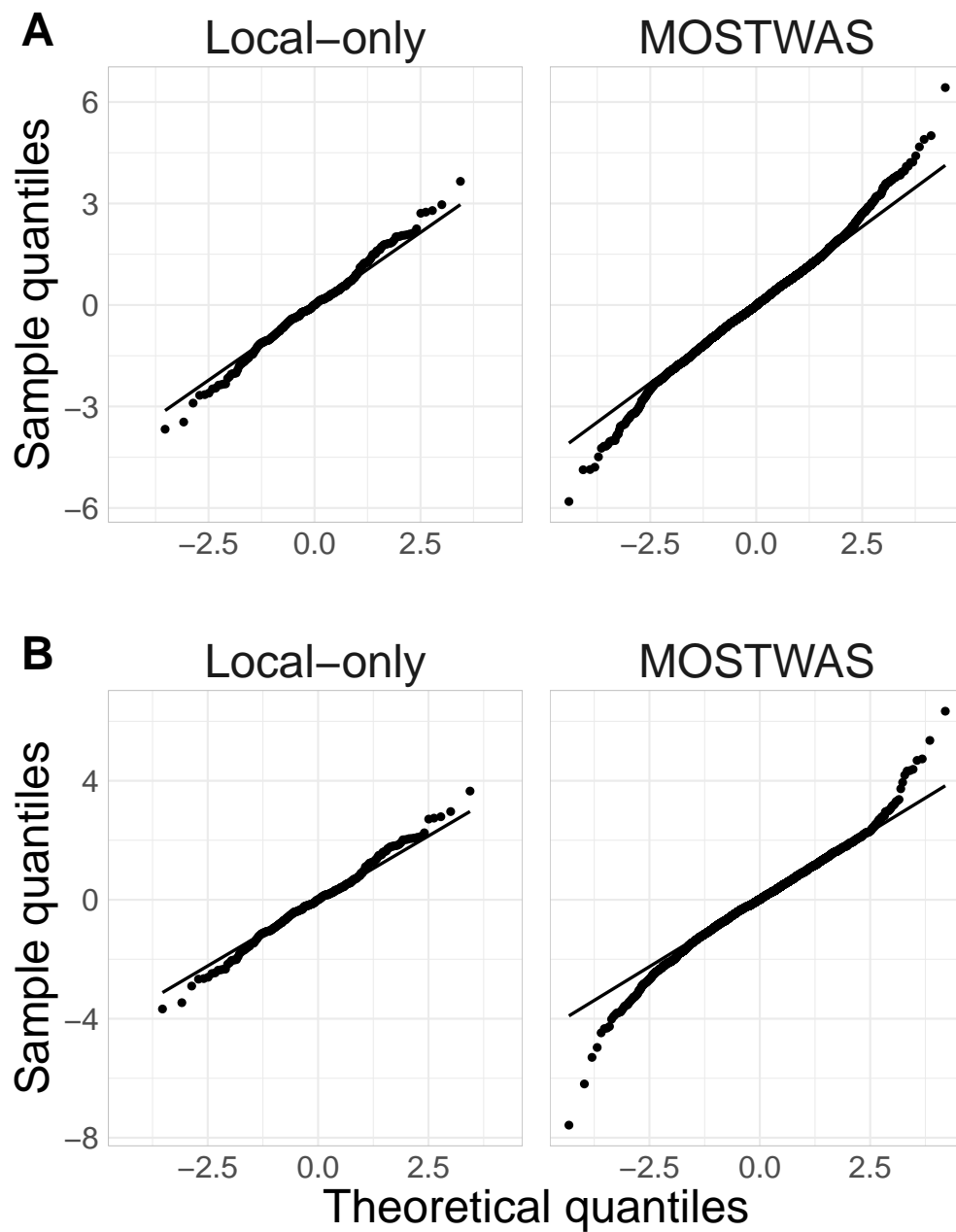


Figure 6: *Comparison of QQ-plots from TWAS associations.* QQ-plots from TWAS for breast cancer-specific survival in iCOGs (A) and MDD in PGC (B) with local-only models (left) and MOSTWAS (right)

Gene	Cross-validation R^2	PGC Z -statistic (UKBB Z)	Top GWAS SNP location (P)	Permutation FDR-adjusted P
ADAD2	0.050	5.89 (4.16)	Chr5:35,639,107 (4.05×10^{-3})	3.5×10^{-5}
CACNA2D3	0.033	3.41 (2.88)	Chr7:12,268,243 (1.27×10^{-2})	0.046
FAM43B	0.035	-4.03 (-2.85)	Chr2:73,148,399 (2.09×10^{-2})	0.028
MGC29506	0.022	3.51 (5.54)	Chr5:139,536,922 (1.48×10^{-3})	3.5×10^{-5}
OR8U1	0.022	-3.19 (-4.21)	Chr11:56,676,947 (4.90×10^{-5})	0.049
SYT1	0.015	-5.58 (-3.16)	Chr7:12,269,417 (1.29×10^{-2})	0.040
YJEFN3	0.010	5.82 (7.22)	Chr7:12,276,011 (1.35×10^{-2})	0.038

Table 4: *Summary statistics for 7 MDD risk-associated loci identified by MOSTWAS models.* TWAS associations with major depressive disorder from GWAS statistics from Psychiatric Genomics Consortium that were replicated with GWAX summary statistics in UK Biobank. The top PGC GWAS SNP in the identified loci with its location and P -value are provided.

References

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