

File S5. Notes on a multi-trait SNP association test for SNP effects estimated under the proposed joint model framework

Characterization of direct and/or indirect genetic associations for multiple traits in longitudinal studies of disease progression

Myriam Brossard,^{*,1} Andrew D. Paterson,^{†,‡} Osvaldo Espin-Garcia,^{‡,**,§§} Radu V. Craiu,^{**} Shelley B. Bull^{*,‡,1}

^{*}Lunenfeld-Tanenbaum Research Institute, Sinai Health, Toronto, M5T 3L9, Ontario, Canada;

[†]Program in Genetics and Genome Biology, Hospital for Sick Children Research Institute, M5G 1X8, Toronto, Ontario, Canada;

[‡]Division of Biostatistics, Dalla Lana School of Public Health, University of Toronto, M5T 3M7, Toronto, Ontario, Canada;

[§]Department of Biostatistics, Princess Margaret Cancer Centre, M5G 2C1, Toronto, Ontario, Canada;

^{**}Department of Statistical Sciences, University of Toronto, M5S 3G3, Toronto, Ontario, Canada;

^{§§}Department of Epidemiology and Biostatistics, Western University, N6A 5C1, London, Ontario, Canada.

¹**Corresponding authors:** Lunenfeld-Tanenbaum Research Institute, 60 Murray Street, Box #18, M5T 3L9, Toronto, Ontario, Canada. E-mails: bull@lunenfeld.ca, brossard@lunenfeld.ca. Phone number: +1 416-586-5052.

[Table of Contents](#)

1. Multi-trait SNP association test	2
2. Simulation results of the multi-trait SNP association test for SNP discovery	2
3. Application of the multi-trait SNP association test in DCCT	4
Cited literature	7

Although our primary aim is to develop inference methods to distinguish among direct and/or indirect SNP associations with each time-to-event trait, the multi-trait aspect of the joint model also lends itself to multi-trait SNP association testing for SNP discovery. In **File S5**, we present a multi-trait SNP association test derived from the parameters of the joint model and report test performance in the DCCT-based simulated data and results in application to DCCT study.

1. Multi-trait SNP association test

The proposed multi-trait SNP association test, assesses if the SNP has at a non-null association with at least one of the $L+K$ traits (*ie* at least one $\beta_{g,l}$ or $\gamma_{g,k} \neq 0$, $1 \leq l \leq L$ and $1 \leq k \leq K$) against the *global null* hypothesis that the SNP has a null effect on the traits (*ie* all $L+K$ traits $\beta_{g,l} = \gamma_{g,k} = 0$, with $1 \leq l \leq L$ and $1 \leq k \leq K$). This test is a generalized Wald statistic constructed from the SNP g effect estimates of the parameter vector $\boldsymbol{\varphi}_g = (\boldsymbol{\beta}_g, \boldsymbol{\gamma}_g)^T$, with $\boldsymbol{\beta}_g = (\beta_{g,1}, \dots, \beta_{g,l}, \dots, \beta_{g,L})$, $\boldsymbol{\gamma}_g = (\gamma_{g,1}, \dots, \gamma_{g,k}, \dots, \gamma_{g,K})$, and the bootstrap covariance matrix $\boldsymbol{\Sigma}_g$, $W_{\varphi_g} = \widehat{\boldsymbol{\varphi}}_g^T \boldsymbol{\Sigma}_g^{-1} \widehat{\boldsymbol{\varphi}}_g$ which is assumed to asymptotically follow a χ^2 distribution with $L+K$ df.

2. Simulation results of the multi-trait SNP association test for SNP discovery

We evaluate the statistical performance (power, type I error) of the multi-trait SNP association test from the joint models (JM-cmp, JM-mis), in comparison to taking the minimum p-value from the marginal models (referred as MM), fitted separately for each trait (*ie* linear mixed models fitted for each QT and Cox PH models for each time-to-event trait ignoring the QTs). Under the *global null* hypothesis, the multi-trait SNP association test does not show any marked departure from its expected distribution (χ^2 with four degrees of freedom), as shown in the following Quantile-Quantile plot (**Fig. S2-1**). Furthermore, type-1 error under the *global null* genetic scenario appears well controlled, respectively: 0.048 at $P^* = 0.05$ and 0.0097 at $P^* = 0.01$.

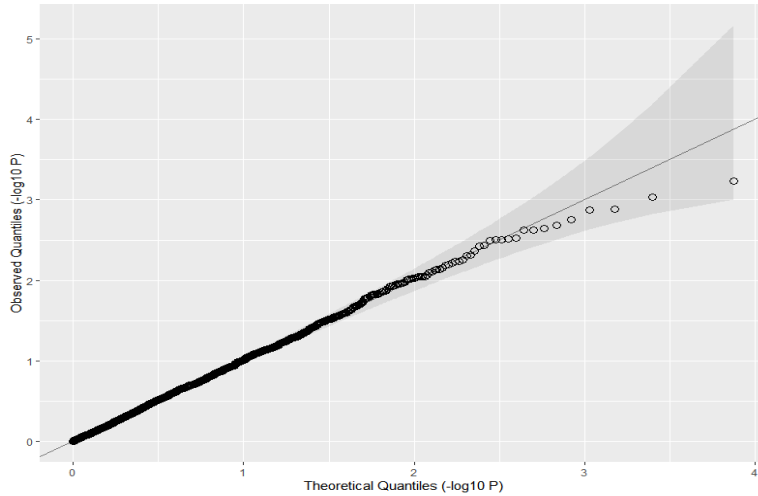
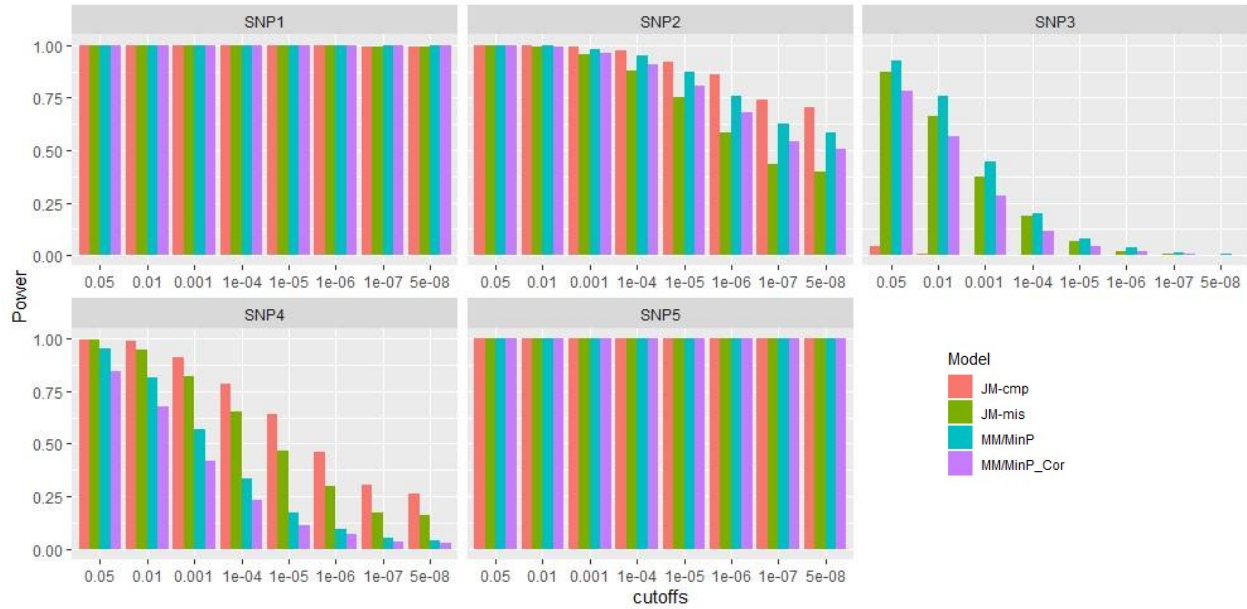


Fig. S2-1. Quantile-Quantile plot of the multi-trait SNP association test p values (JM-cmp), assessed under the global *null* genetic scenario. P-values for all 5 SNPs, were pooled, yielding to $R=5000$ replicates.

Under the *causal* genetic scenario, when the JM is fully specified (JM-cmp), as shown in **Fig. S2-2**, the power of the proposed multi-trait SNP association test appears higher (or similar) than the power based on the minimum P -value from MM in all scenarios of SNP associations (MM_MinP uncorrected and MM_MinP_cor corrected for the four traits analyzed separately assuming that the traits are independent, which is unrealistic but represents an extreme case of conservative P -value). When the joint model is mis-specified (JM-mis), the multi-trait SNP association test is still as or more powerful than the MM_MinP for all causal SNPs except for SNP2 where the power reduction is more pronounced at more stringent significant levels ($P^* \leq 10^{-5}$). Multi-trait association results for SNP3 also show attenuated power compared to the uncorrected MM_MinP; but its power remain higher than the power of the conservative MM_MinP_cor. Except for SNP5 that has both direct and indirect effects on time-to-DN, the other simulated causal SNPs have either a *single* direct or a *single* indirect effect on the time-to-event traits; the latter do not represent scenarios of SNP associations where we would expect large power improvement from the proposed multi-trait SNP association test (largest power improvement would be expected for SNPs with effects on multiple traits).

Fig. S2-2. Power of the multi-trait SNP association tests (P_{mult}) under the joint models (JM-cmp, JM-mis) to detect a SNP association, compared to the minimum SNP P -value from the marginal models (MM) fitted separately for each trait corrected and uncorrected for the number of traits tested (Min_P_Cor and MinP). Power is assessed using $R=1000$ replicates of $N=667$ DCCT subjects simulated under the *causal* genetic scenario from **Fig. 3**.



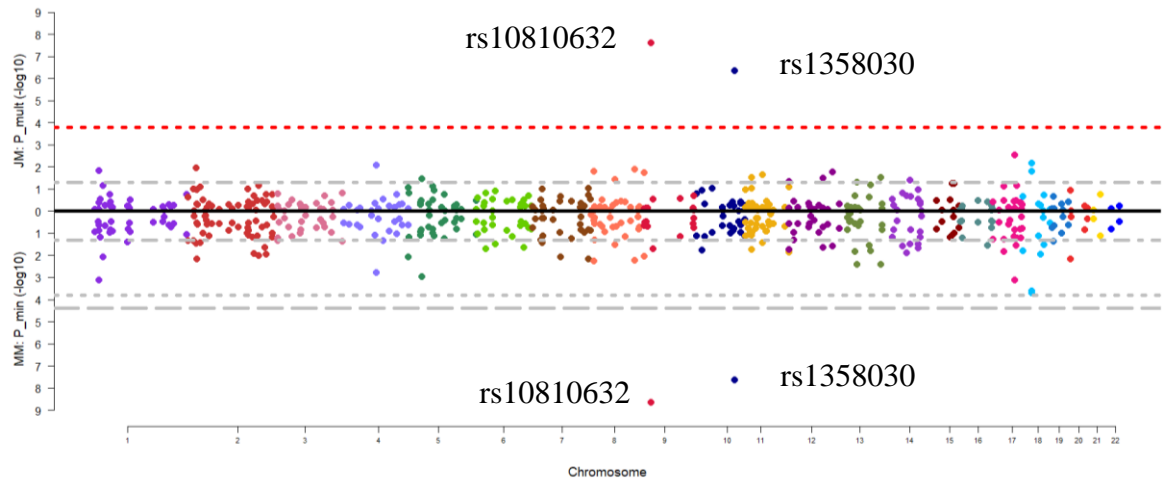
3. Application of the multi-trait SNP association test in DCCT

Out of the 307 SNPs analyzed in DCCT individuals with the joint model, we identify two SNPs reaching the Bonferroni corrected significance threshold for the effective number of SNPs tested ($P^*=1.7 \times 10^{-4}$, **Fig. S3-1**, Panel A.). These SNPs are rs10810632 (*BNC2*, $P_{\text{mult}}=9.1 \times 10^{-8}$) and rs1358030 (nearby *SORCS1*, $P_{\text{mult}}=3.4 \times 10^{-7}$) and are among the SNPs reported associated with HbA1c and time-to-T1DC traits in the previous GWAS of HbA1c in the Conventional treatment group of DCCT (Paterson et al. 2010). We obtained similar conclusions with the two other compared association structures for HbA1c on T1DC (**Fig. S3-1**, Panels B. and C.).

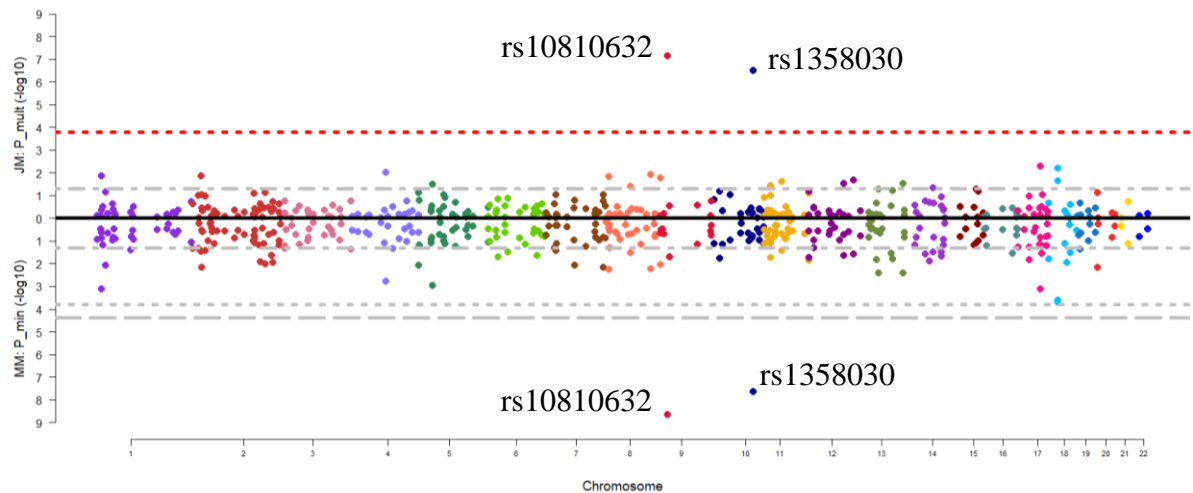
Fig. S3-1. Mirror-plots of the multi-trait SNP association test P -values ($-\log_{10}$) from the joint model using alternatively each association structure for HbA1c effects on time-to-T1DC traits. For each Manhattan plot, the upper panel represents the P -values ($-\log_{10}$) of the multi-trait SNP association test, and the lower panel represents the results of the minimum P -value from the separate analysis of each trait, included for comparison. On each upper panel, the horizontal lines represent the significance levels (in red: Bonferroni-corrected thresholds ($P^* = 1.7 \times 10^{-4}$) and in grey the nominal significance levels). On each lower panel, the grey dashed lines represent the nominal significance and Bonferroni-corrected significance levels for the effective number of SNPs tested and the Bonferroni-corrected significance level further corrected for the four traits tested (under the assumption of independent traits).

(see Figure on next page)

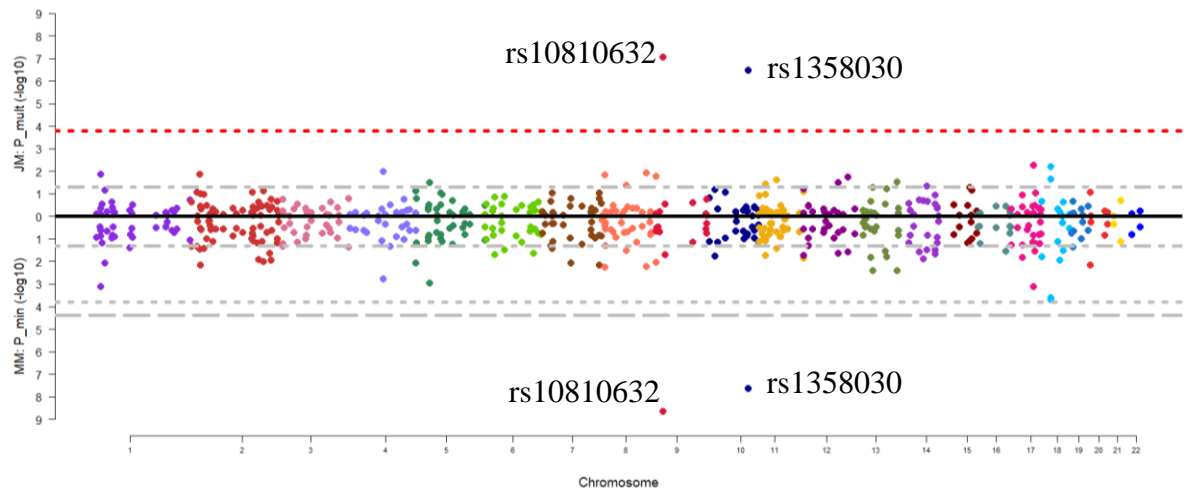
A. Contemporaneous HbA1c effects



B. Updated cumulative mean HbA1c effects



C. Time-weighted cumulative HbA1c effects.



Cited literature

Paterson AD, Waggott D, Boright AP, Hosseini SM, Shen E, Sylvestre MP, Wong I, Bharaj B, Cleary PA, Lachin JM, et al. 2010. A genome-wide association study identifies a novel major locus for glycemic control in type 1 diabetes, as measured by both A1C and glucose. *Diabetes*. 59(2):539–549. doi:10.2337/db09-0653.