Supplementary materials for *Methods for Large-scale Single Mediator Hypothesis Testing: Possible Choices and Comparisons*

Jiacong Du[1], Xiang Zhou[1], Dylan Clark-Boucher[1], Wei Hao[1], Yongmei Liu[2], Jennifer A. Smith[3] and Bhramar Mukherjee[1]
[1] Department of Biostatistics, University of Michigan, Ann Arbor, MI
[2] Department of Medicine, Divisions of Cardiology and Neurology,
Duke University Medical Center, Durham, NC
[3] Department of Epidemiology, University of Michigan, Ann Arbor, MI

S1 Comparison between traditional and causal mediation analysis

Continuous outcomes and continuous mediators

The assumption regarding the exposure-mediator interaction on the outcome is critical in the potential outcome framework (MacKinnon et al., 2020). When assessing mediation, we consider the following two regression models:

$$E(Y|M,X,C) = \beta_0 + \beta_X X + \beta M + \gamma XM + \beta_C^T C; \tag{1}$$

$$E(M|X,C) = \alpha_0 + \alpha X + \alpha_C^T C. \tag{2}$$

Following MacKinnon et al. (2020) and VanderWeele (2015), we express the natural direct effect (NDE) and the natural indirect effect (NIE):

1. $NDE = E[Y(x^*, M(x))|C] - E[Y(x, M(x))|C] = (\beta_X + \gamma\alpha_0 + \gamma\alpha x + \gamma\alpha_C^T c)(x^* - x)$;
2. $NIE = E[Y(x^*, M(x^*))|C] - E[Y(x^*, M(x))|C] = (\alpha\beta + \gamma\alpha x^*)(x^* - x)$.

The total effect (TE) = $NDE + NIE$. We see that if there is no exposure-mediator interaction affecting the outcome, i.e. $\gamma = 0$, the NIE is $\alpha\beta(x^* - x)$. In this case, the traditional product-of-coefficients approach is equivalent to the causal approach. On the other hand, if $\gamma \neq 0$, the traditional methods are invalid since they ignore the term $\gamma\alpha x^*$, and the NIE depends on the value of the intervention level of the exposure due to exposure-mediator interaction.

Continuous outcomes and binary mediators

The logistic model for the mediator M is:

$$logit(P(M = 1|X, C)) = \alpha_0 + \alpha X + \alpha_C^T C. \tag{3}$$

The two causal quantities under models (1) and (3) are (VanderWeele, 2015):

1. $NDE = E[Y(x^*, M(x))|C] - E[Y(x, M(x))|C] = \beta(x^* - x) + \gamma(x^* - x)\frac{exp(\alpha_0+\alpha x+\alpha_C^T C)}{1+exp(\alpha_0+\alpha x+\alpha_C^T C)}$;
2. $NIE = E[Y(x^*, M(x^*))|C] - E[Y(x^*, M(x))|C]$ $= (\beta + \gamma x^*)\left(\frac{exp(\alpha_0+\alpha x^*+\alpha_C^T C)}{1+exp(\alpha_0+\alpha x^*+\alpha_C^T C)} - \frac{exp(\alpha_0+\alpha x+\alpha_C^T C)}{1+exp(\alpha_0+\alpha x+\alpha_C^T C)}\right)$.

It can be seen that if the mediator is binary, regardless of the exposure-mediator interaction, the NIE depends on all the parameters in the mediator model as well as the values of confounders; thus, the product $\alpha\beta$ in traditional mediation analysis does not correspond to the NIE.

Binary outcomes and continuous mediators

The logistic regression model for the outcome Y is:

$$logit(P(Y = 1|X, M, C)) = \beta_0 + \beta_X X + \beta M + \gamma XM + \beta_C^T C. \tag{4}$$

VanderWeele and Vansteelandt (2010) extend the definition of direct and indirect effects in causal inference from the mean difference scale for the continuous outcome to the odds ratio scale for the binary outcome. To avoid the problem of non-collapsibility, they assume that the outcome is rare so that

$$\begin{aligned} P(Y=1|X,M,C) &= \frac{exp\{\beta_0+\beta_X X+\beta M+\gamma XM+\beta_C^T C\}}{1+exp\{\beta_0+\beta_X X+\beta M+\gamma XM+\beta_C^T C\}} \\ &\approx exp\{\beta_0+\beta_X X+\beta M+\gamma XM+\beta_C^T C\}. \end{aligned}$$

In addition, if the four no-unmeasured-confounding assumptions hold and the random error in model (2) is normally distributed with variance σ^2, we have (Rijnhart et al., 2021; VanderWeele, 2015; VanderWeele & Vansteelandt, 2010):

1. $OR^{NDE} = \frac{P(Y(x^*,M(x))=1|C)/P(Y(x^*,M(x))=0|C)}{P(Y(x,M(x))=1|C)/P(Y(x,M(x))=0|C)}$

   $= exp\{(\beta_X+\gamma\alpha_0+\gamma\alpha x+\gamma\alpha_C^T c+\gamma\beta\sigma^2)(x^*-x)+0.5\gamma^2\sigma^2(x^{*2}-x^2)\};$

2. $OR^{NIE} = \frac{P(Y(x^*,M(x^*))=1|C)/P(Y(x^*,M(x^*))=0|C)}{P(Y(x^*,M(x))=1|C)/P(Y(x^*,M(x))=0|C)} = exp\{(\alpha\beta+\gamma\alpha x^*)(x^*-x)\}.$

Similar to the case with continuous mediators and continuous outcomes, if there is no exposure-mediator interaction affecting the outcome, i.e. $\gamma = 0$, $\alpha\beta$ corresponds to the NIE on the log-odds-ratio scale. Thus, the traditional approach is equivalent to the causal approach. However, if $\gamma \neq 0$, it is inappropriate to use the traditional product-of-coefficients methods because they omit the part of the mediation effect modified by the exposure-mediator interaction, i.e. $\gamma\alpha x^*$.

Binary outcomes and binary mediators

When the outcome is rare, the two causal quantities under models (4) and (3) are (VanderWeele, 2015):

1. $OR^{NDE} = \frac{P(Y(x^*,M(x))=1|C)/P(Y(x^*,M(x))=0|C)}{P(Y(x,M(x))=1|C)/P(Y(x,M(x))=0|C)} = \frac{exp(\beta_X x^*)\left(1+exp(\beta+\gamma x^*+\alpha_0+\alpha x+\alpha_C^T C)\right)}{exp(\beta_X x)\left(1+exp(\beta+\gamma x+\alpha_0+\alpha x+\alpha_C^T C)\right)};$

2. $OR^{NIE} = \frac{P(Y(x^*,M(x^*))=1|C)/P(Y(x^*,M(x^*))=0|C)}{P(Y(x^*,M(x))=1|C)/P(Y(x^*,M(x))=0|C)}$

   $= \frac{\left(1+exp(\alpha_0+\alpha x+\alpha_C^T C)\right)\left(1+exp(\beta+\gamma x^*+\alpha_0+\alpha x^*+\alpha_C^T C)\right)}{\left(1+exp(\alpha_0+\alpha x^*+\alpha_C^T C)\right)\left(1+exp(\beta+\gamma x^*+\alpha_0+\alpha x+\alpha_C^T C)\right)}.$

Comparing to the NIE expression for continuous outcomes and binary mediators, we draw a similar conclusion for binary outcomes and binary mediators here: Regardless of the exposure-mediator interaction, the product $\alpha\beta$ does not correspond to the NIE and, thus, the traditional methods are inappropriate for the mediation analysis.

Hypothesis testing

In terms of hypothesis testing with continuous mediators and outcomes, the null hypothesis of having no mediation effect is:

$$H_0 : \alpha\beta + \gamma\alpha x^* = 0.$$

If the exposure-mediator interaction effect is nonzero, i.e. $\gamma \neq 0$, the traditional methods are expected to perform worse than the causal methods, since $\alpha\beta$ does not represent the NIE. If $\gamma = 0$, the null hypothesis reduces to:

$$H_0 : \alpha\beta = 0.$$

The traditional product-of-coefficients approach is equivalent to the causal approach in that rejecting the null hypothesis H_0 implies a non-zero NIE. Thus, both approaches face the issue of the composite null.

MacKinnon et al. (2020) performed simulation studies to compare the two approaches for continuous outcomes and continuous mediators with and without exposure-mediator interaction. They found that when the true value of the exposure-mediator interaction was zero, there was lower power to detect NIE from the causal approach than using the product from the traditional approach, especially with a small sample size, or with a small effect size of α or β. The reduced power was likely caused by estimating models with additional parameters. With the exposure-mediator interaction, the power of using the potential outcome framework was higher than that of using the product approach.

S2 Proof of Proposition 1

Proof. In this proof, we drop the subscript j since the statement is true for all $j \in \{1, 2, ..., J\}$. When $|Z_\beta| > |Z_\alpha|$, under H_{00},

$$T_{sobel} = \frac{|Z_\alpha|}{\sqrt{1 + Z_\alpha^2/Z_\beta^2}} \sim N(0, 1/4)$$

$$p_{max} = 2\Phi(|Z_\alpha|) \sim Beta(2, 1)$$

The p-value for Sobel-comp under H_{00} is:

$$\begin{aligned} p_{sobel}^{00} &= 2\int_{\frac{|Z\alpha|}{\sqrt{1+Z_\alpha^2/Z_\beta^2}}}^{\infty} (2\pi\frac{1}{4})^{-1/2} exp\big(-2u^2\big)du \\ &= 2\int_{\frac{2|Z_\alpha|}{\sqrt{1+Z_\alpha^2/Z_\beta^2}}}^{\infty} (2\pi)^{-1/2} exp\big(-1/2v^2\big)dv \\ &= 2\Phi\Big(\frac{2|Z_\alpha|}{\sqrt{1 + Z_\alpha^2/Z_\beta^2}}\Big) \end{aligned}$$

The p-value for HDMT under H_{00} is:

$$p_{max}^2 = 4\Phi(|Z_\alpha|)^2$$

Notice that p_{sobel}^{00} is a decreasing function of $|Z_\beta|$, and

$$\begin{aligned} & p_{sobel}^{00} < p_{max}^2 \\ \Longleftrightarrow \quad & 2\Phi\Big(\frac{2|Z_\alpha|}{\sqrt{1 + Z_\alpha^2/Z_\beta^2}}\Big) < 4\Phi(|Z_\alpha|)^2 \\ \Longleftrightarrow \quad & \frac{2|Z_\alpha|}{\sqrt{1 + Z_\alpha^2/Z_\beta^2}} > \Phi^{-1}\Big(2\Phi(|Z_\beta|)^2\Big) \\ \Longleftrightarrow \quad & |Z_\beta| > \Big\{4\Big(\Phi^{-1}\big(2\Phi(|Z_\alpha|)^2\big)\Big)^{-2} - Z_\alpha^{-2}\Big\}^{-1/2} \end{aligned}$$

Note that $\Big\{4\Big(\Phi^{-1}\big(2\Phi(|Z_\alpha|)^2\big)\Big)^{-2} - Z_\alpha^{-2}\Big\} > 0$. Therefore, a sufficient condition for $p_{sobel}^{00} < p_{max}^2$ is $|Z_\beta| > max\Big(|Z_\alpha|, \Big\{4\Big(\Phi^{-1}\big(2\Phi(|Z_\alpha|)^2\big)\Big)^{-2} - Z_\alpha^{-2}\Big\}^{-1/2}\Big)$.

□

S3 A detailed description of MESA data

MESA is a population-based longitudinal study designed to investigate the predictors and progression of subclinical cardiovascular disease in a cohort of 6,814 participants (Bild et al., 2002). Clinical, socio-demographic, lifestyle and behavior, laboratory, nutrition and medication data have been collected at multiple examinations beginning in 2000-2002. We used participants' educational attainment based on their highest degree at MESA Exam 1 as a measure of adult SES (less than a 4-year college degree as 1 vs. with a 4-year college degree or higher as 0). DNAm levels were measured using the Illumina Infinium HumanMethylation450 Beadchip on purified monocytes from a random subsample of 1,264 non-Hispanic white, African-American, and Hispanic MESA participants between April 2010 and February 2012 (corresponding to MESA Exam 5). A total of 402,339 CpG sites remained after quality control and filtering, including: "detected" methylation levels in <90% of MESA samples using a detection p-value cut-off of 0.05, overlap with a repetitive element or region, presence of SNPs within 10 base pairs according to Illumina annotation, non-reliable probes recommended by DMRcate (having SNPs with minor allele frequency > 0.05 within 2 base pairs or cross reactive probes), probes on sex chromosomes, SNPs, and other non-CpG targeting probes. Additional details about the data collection and processing procedures can be found in (Liu et al., 2013). We used HbA1c measured at Exam 5 as the outcome. Our analysis focused on the participants taking no insulin or oral hypoglycemic medication. After removing missing values, a total of 963 individuals remained for analysis.

For the j-th CpG site, where $j = 1, 2, ..., 402339$, we obtained $z_{\alpha,j}$ and $z_{\beta,j}$ from linear models for testing $\alpha_j = 0$ (effect of the exposure on the j-th mediator) and $\beta_j = 0$ (effect of the j-th mediator on the outcome). More specifically, we first remove the random effect of the methylation chip and position from the mediator and the outcome. Using the residuals, we fit two linear regression models for the continuous mediator and the continuous outcome. In both models, we adjusted for age, sex, and race as potential confounders and adjusted for the estimated proportions of residual non-monocytes (neutrophils, B cells, T cells, and natural killer cells) to account for potential contamination by non-monocyte cell types. In addition, we adjusted for the exposure in the outcome model.

Before performing mediation analysis, since correlated mediators may lead to inflated Type I error rates and spurious signals, we selected a subset of 228,088 potentially mediating CpG sites that were, at most, only weakly correlated with one another (correlation coefficient ≤ 0.3). More specifically, we first calculated the correlation matrix for all CpG sites on each chromosome. Then we found the mediator with the smallest MaxP p-value. Next, we identified and removed the group of mediators which were correlated with this mediator with correlation coefficient larger than 0.3. We repeated the previous steps until all elements in the correlation matrix were less than or equal to 0.3.

S4 Tables and figures

Table S1: The mean and standard deviation of the ratio of the false positive rates to the nominal significance level based on 2,000 replications using Sobel's test, MaxP, JT-comp, HDMT, Sobel-comp and DACT under the *Null 1(a)* scenario. n is the sample size. The total number of mediators is 100,000. With probability $\pi_{01} = 0.001$, $\alpha = 0$ and $\beta \sim N(0, \tau^2)$; with probability $\pi_{10} = 0.001, \alpha \sim N(0, 5\tau^2)$ and $\beta = 0$; with probability $\pi_{00} = 0.998, \alpha = \beta = 0$.

Cut-off	Sobel	MaxP	JT-comp	HDMT	Sobel-comp	DACT
$n = 200, \tau = 0.1$						
10^{-3}	0.00 (0.00)	0.00 (0.00)	1.11 (0.10)	1.06 (0.12)	0.89 (0.34)	1.47 (1.81)
10^{-4}	0.00 (0.00)	0.00 (0.01)	1.48 (0.37)	1.12 (0.35)	0.81 (0.66)	2.90 (4.19)
10^{-5}	0.00 (0.00)	0.00 (0.02)	3.02 (1.70)	1.29 (1.17)	1.04 (1.37)	6.54 (10.61)
10^{-6}	0.00 (0.00)	0.00 (0.00)	9.99 (10.09)	1.60 (3.95)	1.81 (4.63)	16.50 (31.56)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	15.19 (17.36)	1.84 (6.15)	2.32 (7.17)	22.65 (47.46)
$n = 500, \tau = 0.1$						
10^{-3}	0.00 (0.00)	0.00 (0.00)	1.15 (0.10)	1.05 (0.11)	0.98 (0.28)	1.20 (1.67)
10^{-4}	0.00 (0.00)	0.00 (0.01)	2.10 (0.44)	1.13 (0.34)	1.07 (0.61)	2.46 (3.88)
10^{-5}	0.00 (0.00)	0.00 (0.03)	7.53 (2.74)	1.29 (1.17)	1.64 (1.59)	6.55 (11.76)
10^{-6}	0.00 (0.00)	0.00 (0.00)	39.48 (20.10)	1.68 (4.18)	4.03 (6.68)	25.82 (71.39)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	66.28 (36.51)	1.83 (6.23)	5.51 (10.99)	42.96 (134.45)
$n = 1000, \tau = 0.1$						
10^{-3}	0.00 (0.00)	0.00 (0.00)	1.18 (0.10)	1.04 (0.10)	1.00 (0.27)	1.07 (1.62)
10^{-4}	0.00 (0.00)	0.00 (0.01)	2.90 (0.52)	1.10 (0.34)	1.17 (0.60)	2.10 (3.72)
10^{-5}	0.00 (0.00)	0.00 (0.00)	14.19 (3.73)	1.32 (1.15)	2.15 (1.75)	4.87 (10.06)
10^{-6}	0.00 (0.00)	0.00 (0.00)	88.27 (30.22)	1.74 (4.19)	6.20 (8.19)	14.31 (45.62)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	155.37 (56.59)	1.99 (6.31)	9.35 (14.05)	20.95 (82.20)
$n = 200, \tau = 0.3$						
10^{-3}	0.00 (0.00)	0.00 (0.01)	1.24 (0.10)	1.07 (0.12)	0.90 (0.36)	NA (NA)
10^{-4}	0.00 (0.01)	0.00 (0.01)	3.88 (0.59)	1.16 (0.37)	0.94 (0.81)	NA (NA)
10^{-5}	0.00 (0.00)	0.00 (0.04)	22.26 (4.57)	1.43 (1.25)	1.80 (2.19)	NA (NA)
10^{-6}	0.00 (0.00)	0.00 (0.22)	152.66 (37.96)	2.17 (4.65)	5.72 (9.60)	NA (NA)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	275.57 (72.57)	2.53 (7.09)	8.72 (16.04)	NA (NA)
$n = 500, \tau = 0.3$						
10^{-3}	0.00 (0.00)	0.00 (0.01)	1.18 (0.10)	1.04 (0.14)	1.02 (0.30)	NA (NA)
10^{-4}	0.00 (0.01)	0.00 (0.01)	5.45 (0.68)	1.11 (0.40)	1.33 (0.75)	NA (NA)
10^{-5}	0.00 (0.02)	0.00 (0.04)	38.44 (5.88)	1.35 (1.27)	3.07 (2.52)	NA (NA)
10^{-6}	0.00 (0.00)	0.00 (0.00)	293.94 (51.47)	2.09 (4.66)	11.33 (12.76)	NA (NA)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	544.06 (98.68)	2.44 (7.08)	17.35 (21.40)	NA (NA)
$n = 1000, \tau = 0.3$						
10^{-3}	0.00 (0.00)	0.00 (0.01)	1.09 (0.09)	1.02 (0.14)	1.04 (0.28)	NA (NA)
10^{-4}	0.00 (0.01)	0.00 (0.01)	6.63 (0.72)	1.07 (0.41)	1.44 (0.72)	NA (NA)
10^{-5}	0.00 (0.02)	0.00 (0.04)	51.39 (6.52)	1.34 (1.25)	3.69 (2.63)	NA (NA)
10^{-6}	0.00 (0.00)	0.00 (0.22)	412.90 (57.98)	2.01 (4.58)	14.35 (13.98)	NA (NA)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	775.58 (112.77)	2.44 (6.99)	22.73 (23.93)	NA (NA)
$n = 200, \tau = 0.7$						
10^{-3}	0.00 (0.00)	0.00 (0.01)	1.10 (0.10)	1.02 (0.16)	0.93 (0.37)	NA (NA)
10^{-4}	0.00 (0.01)	0.00 (0.01)	6.78 (0.76)	1.02 (0.46)	1.07 (0.89)	NA (NA)
10^{-5}	0.00 (0.04)	0.00 (0.04)	52.79 (6.81)	1.17 (1.30)	2.46 (2.88)	NA (NA)
10^{-6}	0.00 (0.00)	0.01 (0.32)	424.85 (61.32)	1.81 (4.46)	9.20 (13.93)	NA (NA)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	798.87 (118.48)	2.11 (6.62)	14.43 (23.41)	NA (NA)
$n = 500, \tau = 0.7$						
10^{-3}	0.00 (0.00)	0.00 (0.01)	1.04 (0.09)	1.03 (0.15)	1.04 (0.31)	NA (NA)
10^{-4}	0.00 (0.01)	0.00 (0.01)	8.12 (0.78)	1.10 (0.43)	1.48 (0.82)	NA (NA)
10^{-5}	0.00 (0.03)	0.00 (0.04)	67.30 (7.16)	1.36 (1.33)	3.91 (3.04)	NA (NA)
10^{-6}	0.00 (0.00)	0.00 (0.00)	564.02 (65.71)	2.35 (5.00)	16.41 (16.62)	NA (NA)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	1071.33 (128.36)	2.84 (7.85)	25.80 (28.32)	NA (NA)
$n = 1000, \tau = 0.7$						
10^{-3}	0.00 (0.00)	0.00 (0.01)	1.06 (0.09)	1.02 (0.14)	1.06 (0.29)	NA (NA)
10^{-4}	0.00 (0.01)	0.00 (0.02)	8.83 (0.80)	1.08 (0.41)	1.57 (0.77)	NA (NA)
10^{-5}	0.00 (0.04)	0.00 (0.04)	74.58 (7.23)	1.38 (1.27)	4.47 (3.06)	NA (NA)
10^{-6}	0.00 (0.00)	0.00 (0.22)	634.88 (66.91)	2.14 (4.69)	18.84 (16.82)	NA (NA)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	1211.53 (131.22)	2.65 (7.24)	30.22 (28.72)	NA (NA)

Table S2: The mean and standard deviation of the ratio of the false positive rates to the nominal significance level based on 2,000 replications using Sobel's test, MaxP, JT-comp, HDMT, Sobel-comp and DACT under the *Null 1(b)* scenario. n is the sample size. The total number of mediators is 100,000. With probability $\pi_{01} = 0.001$, $\alpha = 0$ and $\beta \sim N(0, \tau^2)$; with probability $\pi_{10} = 0.001, \alpha \sim N(0, 5\tau^2)$ and $\beta = 0$; with probability $\pi_{00} = 0.998, \alpha = \beta = 0$.

Cut-off	Sobel	MaxP	JT-comp	HDMT	Sobel-comp	DACT
$n = 200, \tau = 0.1$						
10^{-3}	0.01 (0.01)	0.14 (0.04)	3.48 (0.18)	1.08 (0.10)	0.04 (0.02)	7.07 (2.11)
10^{-4}	0.00 (0.01)	0.11 (0.11)	7.89 (0.89)	1.15 (0.34)	0.01 (0.03)	18.06 (7.06)
10^{-5}	0.00 (0.02)	0.10 (0.32)	18.25 (4.12)	1.26 (1.13)	0.00 (0.07)	48.93 (24.16)
10^{-6}	0.00 (0.00)	0.06 (0.80)	43.28 (20.77)	1.35 (3.77)	0.00 (0.00)	137.96 (85.53)
5×10^{-7}	0.00 (0.00)	0.07 (1.18)	56.20 (33.23)	1.55 (5.68)	0.00 (0.00)	190.14 (127.65)
$n = 500, \tau = 0.1$						
10^{-3}	0.03 (0.02)	0.24 (0.05)	4.16 (0.20)	1.04 (0.11)	0.10 (0.03)	13.74 (2.34)
10^{-4}	0.01 (0.03)	0.21 (0.14)	9.97 (0.97)	1.07 (0.33)	0.04 (0.06)	41.00 (9.13)
10^{-5}	0.01 (0.08)	0.19 (0.43)	24.31 (4.94)	1.18 (1.09)	0.02 (0.13)	126.92 (35.30)
10^{-6}	0.01 (0.32)	0.16 (1.26)	59.87 (24.24)	1.28 (3.52)	0.02 (0.39)	405.26 (141.83)
5×10^{-7}	0.00 (0.00)	0.13 (1.61)	78.44 (39.61)	1.11 (4.67)	0.02 (0.63)	576.58 (216.56)
$n = 1000, \tau = 0.1$						
10^{-3}	0.07 (0.03)	0.33 (0.06)	4.44 (0.20)	1.02 (0.11)	0.16 (0.04)	NA (NA)
10^{-4}	0.03 (0.06)	0.29 (0.17)	10.91 (1.02)	1.04 (0.32)	0.08 (0.09)	NA (NA)
10^{-5}	0.01 (0.12)	0.26 (0.50)	27.43 (5.12)	1.07 (1.05)	0.04 (0.18)	NA (NA)
10^{-6}	0.00 (0.22)	0.25 (1.55)	68.54 (26.48)	1.10 (3.35)	0.00 (0.22)	NA (NA)
5×10^{-7}	0.01 (0.45)	0.26 (2.27)	90.38 (43.37)	1.05 (4.51)	0.01 (0.45)	NA (NA)
$n = 200, \tau = 0.3$						
10^{-3}	0.19 (0.04)	0.69 (0.08)	3.05 (0.17)	0.84 (0.09)	0.25 (0.05)	NA (NA)
10^{-4}	0.13 (0.11)	0.71 (0.27)	5.79 (0.77)	0.87 (0.29)	0.16 (0.12)	NA (NA)
10^{-5}	0.09 (0.30)	0.76 (0.86)	11.15 (3.43)	0.93 (0.94)	0.11 (0.34)	NA (NA)
10^{-6}	0.10 (0.97)	0.94 (3.06)	21.78 (14.68)	1.10 (3.30)	0.11 (1.02)	NA (NA)
5×10^{-7}	0.10 (1.41)	1.07 (4.63)	27.04 (22.79)	1.20 (4.88)	0.12 (1.54)	NA (NA)
$n = 500, \tau = 0.3$						
10^{-3}	0.22 (0.05)	0.51 (0.07)	4.69 (0.21)	0.88 (0.10)	0.40 (0.06)	NA (NA)
10^{-4}	0.16 (0.13)	0.51 (0.22)	11.77 (1.07)	0.87 (0.29)	0.29 (0.17)	NA (NA)
10^{-5}	0.12 (0.36)	0.53 (0.72)	30.10 (5.59)	0.91 (0.96)	0.23 (0.48)	NA (NA)
10^{-6}	0.10 (0.97)	0.52 (2.35)	76.37 (27.35)	0.87 (3.02)	0.15 (1.20)	NA (NA)
5×10^{-7}	0.12 (1.54)	0.53 (3.27)	101.96 (45.47)	0.91 (4.31)	0.19 (1.94)	NA (NA)
$n = 1000, \tau = 0.3$						
10^{-3}	0.29 (0.05)	0.55 (0.07)	4.72 (0.20)	0.92 (0.09)	0.51 (0.07)	NA (NA)
10^{-4}	0.22 (0.15)	0.53 (0.23)	11.87 (1.07)	0.89 (0.29)	0.39 (0.20)	NA (NA)
10^{-5}	0.17 (0.42)	0.52 (0.72)	30.51 (5.44)	0.86 (0.92)	0.30 (0.54)	NA (NA)
10^{-6}	0.14 (1.15)	0.50 (2.24)	78.75 (27.61)	0.80 (2.77)	0.18 (1.33)	NA (NA)
5×10^{-7}	0.12 (1.54)	0.54 (3.24)	103.15 (46.21)	0.91 (4.26)	0.25 (2.22)	NA (NA)
$n = 200, \tau = 0.7$						
10^{-3}	0.34 (0.06)	0.63 (0.08)	4.76 (0.20)	1.02 (0.10)	0.59 (0.08)	NA (NA)
10^{-4}	0.30 (0.17)	0.69 (0.26)	12.03 (1.06)	1.12 (0.34)	0.51 (0.23)	NA (NA)
10^{-5}	0.29 (0.53)	0.84 (0.93)	31.04 (5.45)	1.30 (1.13)	0.48 (0.69)	NA (NA)
10^{-6}	0.22 (1.50)	0.96 (3.04)	81.26 (27.48)	1.59 (3.90)	0.42 (2.02)	NA (NA)
5×10^{-7}	0.20 (1.99)	1.07 (4.55)	109.32 (46.78)	1.63 (5.58)	0.40 (2.87)	NA (NA)
$n = 500, \tau = 0.7$						
10^{-3}	0.42 (0.06)	0.62 (0.08)	4.76 (0.21)	0.99 (0.10)	0.68 (0.08)	NA (NA)
10^{-4}	0.37 (0.19)	0.65 (0.25)	11.98 (1.07)	1.02 (0.32)	0.61 (0.25)	NA (NA)
10^{-5}	0.34 (0.59)	0.67 (0.82)	30.77 (5.60)	1.07 (1.03)	0.55 (0.75)	NA (NA)
10^{-6}	0.32 (1.78)	0.65 (2.64)	78.32 (27.73)	1.08 (3.37)	0.52 (2.30)	NA (NA)
5×10^{-7}	0.28 (2.35)	0.66 (3.68)	104.84 (46.12)	1.02 (4.58)	0.50 (3.12)	NA (NA)
$n = 1000, \tau = 0.7$						
10^{-3}	0.47 (0.07)	0.63 (0.08)	4.76 (0.21)	0.99 (0.10)	0.76 (0.09)	NA (NA)
10^{-4}	0.42 (0.20)	0.62 (0.25)	11.97 (1.08)	0.99 (0.31)	0.67 (0.25)	NA (NA)
10^{-5}	0.37 (0.62)	0.61 (0.78)	30.82 (5.45)	0.96 (0.97)	0.59 (0.76)	NA (NA)
10^{-6}	0.35 (1.87)	0.58 (2.37)	79.55 (27.58)	0.91 (2.97)	0.53 (2.25)	NA (NA)
5×10^{-7}	0.35 (2.62)	0.63 (3.49)	105.52 (46.71)	0.98 (4.41)	0.51 (3.15)	NA (NA)

Table S3: The mean and standard deviation of the ratio of the false positive rates to the nominal significance level based on 2,000 replications using Sobel's test, MaxP, JT-comp, HDMT, Sobel-comp and DACT under the *Null 1(c)* scenario. n is the sample size. The total number of mediators is 100,000. With probability $\pi_{01} = 0.5$, $\alpha = 0$ and $\beta \sim N(0, \tau^2)$ and with probability $\pi_{10} = 0.5, \alpha \sim N(0, 5\tau^2)$ and $\beta = 0$.

Cut-off	Sobel	MaxP	JT-comp	HDMT	Sobel-comp	DACT
$n = 200, \tau = 0.1$						
10^{-3}	0.01 (0.01)	0.21 (0.05)	2.58 (0.16)	1.09 (0.10)	0.03 (0.02)	5.70 (1.87)
10^{-4}	0.00 (0.02)	0.17 (0.13)	4.61 (0.67)	1.16 (0.34)	0.01 (0.03)	12.53 (5.30)
10^{-5}	0.00 (0.02)	0.13 (0.37)	8.37 (3.00)	1.26 (1.13)	0.00 (0.05)	29.19 (15.66)
10^{-6}	0.00 (0.00)	0.12 (1.09)	15.16 (12.08)	1.41 (3.78)	0.00 (0.00)	71.21 (49.20)
5×10^{-7}	0.00 (0.00)	0.11 (1.48)	18.39 (19.14)	1.41 (5.50)	0.00 (0.00)	93.19 (71.10)
$n = 500, \tau = 0.1$						
10^{-3}	0.05 (0.02)	0.37 (0.06)	2.86 (0.17)	1.05 (0.10)	0.08 (0.03)	13.28 (2.21)
10^{-4}	0.02 (0.04)	0.32 (0.18)	5.28 (0.76)	1.10 (0.34)	0.03 (0.06)	34.54 (7.40)
10^{-5}	0.01 (0.08)	0.28 (0.53)	9.94 (3.16)	1.19 (1.10)	0.01 (0.12)	92.82 (24.75)
10^{-6}	0.00 (0.22)	0.21 (1.45)	19.17 (13.97)	1.26 (3.48)	0.00 (0.22)	256.76 (89.89)
5×10^{-7}	0.00 (0.00)	0.15 (1.73)	23.10 (21.75)	1.17 (4.74)	0.00 (0.00)	352.29 (134.61)
$n = 1000, \tau = 0.1$						
10^{-3}	0.10 (0.03)	0.50 (0.07)	2.97 (0.17)	1.03 (0.11)	0.14 (0.04)	NA (NA)
10^{-4}	0.05 (0.07)	0.44 (0.21)	5.54 (0.72)	1.05 (0.33)	0.07 (0.09)	NA (NA)
10^{-5}	0.02 (0.15)	0.39 (0.64)	10.49 (3.30)	1.06 (1.05)	0.04 (0.19)	NA (NA)
10^{-6}	0.00 (0.22)	0.29 (1.71)	20.01 (13.77)	1.03 (3.17)	0.00 (0.22)	NA (NA)
5×10^{-7}	0.00 (0.00)	0.21 (2.04)	24.65 (21.70)	0.97 (4.39)	0.01 (0.45)	NA (NA)
$n = 200, \tau = 0.3$						
10^{-3}	0.19 (0.04)	0.69 (0.08)	3.05 (0.17)	0.84 (0.09)	0.25 (0.05)	NA (NA)
10^{-4}	0.13 (0.11)	0.71 (0.27)	5.79 (0.77)	0.87 (0.29)	0.16 (0.12)	NA (NA)
10^{-5}	0.09 (0.30)	0.76 (0.86)	11.15 (3.43)	0.93 (0.94)	0.11 (0.34)	NA (NA)
10^{-6}	0.10 (0.97)	0.94 (3.06)	21.78 (14.68)	1.10 (3.30)	0.11 (1.02)	NA (NA)
5×10^{-7}	0.10 (1.41)	1.07 (4.63)	27.04 (22.79)	1.20 (4.88)	0.12 (1.54)	NA (NA)
$n = 500, \tau = 0.3$						
10^{-3}	0.33 (0.06)	0.78 (0.09)	3.07 (0.17)	0.88 (0.09)	0.38 (0.06)	NA (NA)
10^{-4}	0.24 (0.15)	0.77 (0.28)	5.84 (0.78)	0.87 (0.30)	0.27 (0.16)	NA (NA)
10^{-5}	0.17 (0.41)	0.78 (0.87)	11.42 (3.36)	0.88 (0.93)	0.20 (0.44)	NA (NA)
10^{-6}	0.07 (0.83)	0.64 (2.59)	21.75 (15.05)	0.74 (2.76)	0.08 (0.86)	NA (NA)
5×10^{-7}	0.11 (1.48)	0.61 (3.55)	27.33 (23.93)	0.70 (3.78)	0.11 (1.48)	NA (NA)
$n = 1000, \tau = 0.3$						
10^{-3}	0.44 (0.07)	0.83 (0.09)	3.08 (0.17)	0.91 (0.10)	0.49 (0.07)	NA (NA)
10^{-4}	0.34 (0.19)	0.82 (0.29)	5.84 (0.74)	0.89 (0.31)	0.38 (0.20)	NA (NA)
10^{-5}	0.26 (0.50)	0.79 (0.90)	11.17 (3.34)	0.87 (0.94)	0.29 (0.53)	NA (NA)
10^{-6}	0.15 (1.20)	0.69 (2.62)	21.72 (14.13)	0.78 (2.79)	0.16 (1.26)	NA (NA)
5×10^{-7}	0.11 (1.48)	0.63 (3.49)	26.73 (22.69)	0.74 (3.78)	0.12 (1.54)	NA (NA)
$n = 200, \tau = 0.7$						
10^{-3}	0.52 (0.07)	0.95 (0.10)	3.11 (0.17)	1.03 (0.10)	0.57 (0.08)	NA (NA)
10^{-4}	0.46 (0.21)	1.06 (0.32)	5.94 (0.77)	1.15 (0.33)	0.50 (0.22)	NA (NA)
10^{-5}	0.43 (0.65)	1.21 (1.09)	11.51 (3.46)	1.31 (1.13)	0.46 (0.68)	NA (NA)
10^{-6}	0.47 (2.19)	1.60 (4.06)	22.53 (15.12)	1.72 (4.19)	0.53 (2.30)	NA (NA)
5×10^{-7}	0.51 (3.15)	1.77 (5.92)	28.20 (23.56)	1.84 (6.02)	0.55 (3.27)	NA (NA)
$n = 500, \tau = 0.7$						
10^{-3}	0.63 (0.08)	0.94 (0.10)	3.10 (0.18)	0.99 (0.10)	0.67 (0.08)	NA (NA)
10^{-4}	0.56 (0.24)	0.97 (0.31)	5.91 (0.78)	1.02 (0.32)	0.59 (0.24)	NA (NA)
10^{-5}	0.48 (0.69)	1.01 (0.99)	11.54 (3.42)	1.06 (1.02)	0.51 (0.71)	NA (NA)
10^{-6}	0.36 (1.94)	0.97 (3.15)	22.22 (15.28)	1.02 (3.22)	0.38 (2.00)	NA (NA)
5×10^{-7}	0.27 (2.31)	0.96 (4.46)	27.71 (24.06)	1.01 (4.60)	0.28 (2.35)	NA (NA)
$n = 1000, \tau = 0.7$						
10^{-3}	0.71 (0.08)	0.95 (0.10)	3.09 (0.17)	0.98 (0.10)	0.74 (0.09)	NA (NA)
10^{-4}	0.64 (0.25)	0.96 (0.32)	5.88 (0.74)	0.99 (0.33)	0.66 (0.26)	NA (NA)
10^{-5}	0.58 (0.76)	0.96 (0.99)	11.24 (3.33)	0.99 (1.01)	0.60 (0.77)	NA (NA)
10^{-6}	0.41 (1.98)	0.88 (2.99)	21.97 (14.16)	0.92 (3.05)	0.44 (2.06)	NA (NA)
5×10^{-7}	0.31 (2.47)	0.82 (4.02)	26.97 (22.79)	0.89 (4.17)	0.37 (2.70)	NA (NA)

Table S4: The mean and standard deviation of the ratio of the false positive rates to the nominal significance level based on 2,000 replications using Sobel's test, MaxP, JT-comp, HDMT, Sobel-comp and DACT under the *Null 2(a)* scenario. n is the sample size. The total number of mediators is 100,000. With probability $\pi_{01} = 0.001$, $\alpha = 0$ and $\beta \sim N(0, 0.3^2)$; with probability $\pi_{10} = 0.001, \alpha \sim N(0, 5 \times 0.3^2)$ and $\beta = 0$; with probability $\pi_{00} = 0.998, \alpha = \beta = 0$. R^2 is controlled in the mediator and outcome models.

Cut-off	Sobel	MaxP	JT-comp	HDMT	Sobel-comp	DACT
$n = 200, R^2 = 0.1$						
10^{-3}	0.00 (0.00)	0.00 (0.00)	1.11 (0.10)	1.06 (0.12)	0.88 (0.35)	1.51 (1.81)
10^{-4}	0.00 (0.00)	0.00 (0.01)	1.38 (0.36)	1.12 (0.36)	0.79 (0.68)	2.98 (4.23)
10^{-5}	0.00 (0.00)	0.00 (0.02)	2.23 (1.44)	1.31 (1.18)	1.00 (1.36)	6.72 (10.70)
10^{-6}	0.00 (0.00)	0.00 (0.00)	4.54 (6.66)	1.66 (4.03)	1.48 (4.08)	16.62 (29.99)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	5.79 (10.79)	1.80 (6.06)	1.87 (6.35)	22.82 (43.78)
$n = 500, R^2 = 0.1$						
10^{-3}	0.00 (0.00)	0.00 (0.00)	1.18 (0.11)	1.05 (0.11)	0.98 (0.28)	1.25 (1.67)
10^{-4}	0.00 (0.00)	0.00 (0.01)	2.13 (0.45)	1.14 (0.35)	1.10 (0.63)	2.68 (4.00)
10^{-5}	0.00 (0.00)	0.00 (0.03)	6.62 (2.57)	1.33 (1.19)	1.77 (1.68)	8.06 (15.20)
10^{-6}	0.00 (0.00)	0.00 (0.00)	26.99 (16.76)	1.75 (4.24)	4.39 (7.10)	38.21 (111.24)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	41.82 (29.40)	1.88 (6.24)	6.01 (11.63)	66.04 (210.69)
$n = 1000, R^2 = 0.1$						
10^{-3}	0.00 (0.00)	0.00 (0.00)	1.25 (0.11)	1.04 (0.11)	1.01 (0.27)	1.13 (1.64)
10^{-4}	0.00 (0.00)	0.00 (0.01)	3.22 (0.55)	1.11 (0.34)	1.22 (0.62)	2.31 (3.86)
10^{-5}	0.00 (0.00)	0.00 (0.02)	15.17 (3.91)	1.37 (1.17)	2.42 (1.89)	6.24 (13.61)
10^{-6}	0.00 (0.00)	0.00 (0.00)	84.91 (29.66)	1.85 (4.32)	7.38 (9.20)	25.98 (100.08)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	143.54 (54.73)	2.09 (6.44)	11.01 (15.57)	43.54 (192.81)
$n = 200, R^2 = 0.15$						
10^{-3}	0.00 (0.00)	0.00 (0.00)	1.13 (0.10)	1.06 (0.12)	0.88 (0.35)	1.53 (1.83)
10^{-4}	0.00 (0.00)	0.00 (0.01)	1.55 (0.38)	1.13 (0.36)	0.81 (0.69)	3.04 (4.28)
10^{-5}	0.00 (0.00)	0.00 (0.02)	2.99 (1.65)	1.32 (1.18)	1.09 (1.46)	6.90 (11.00)
10^{-6}	0.00 (0.00)	0.00 (0.00)	8.07 (8.92)	1.74 (4.14)	1.88 (4.73)	17.46 (32.68)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	10.80 (14.69)	1.95 (6.33)	2.49 (7.46)	23.91 (48.56)
$n = 500, R^2 = 0.15$						
10^{-3}	0.00 (0.00)	0.00 (0.00)	1.22 (0.11)	1.05 (0.11)	0.99 (0.29)	1.25 (1.71)
10^{-4}	0.00 (0.00)	0.00 (0.01)	2.54 (0.49)	1.15 (0.35)	1.13 (0.66)	2.60 (4.04)
10^{-5}	0.00 (0.00)	0.00 (0.03)	9.54 (3.04)	1.36 (1.21)	1.97 (1.81)	6.87 (13.35)
10^{-6}	0.00 (0.00)	0.00 (0.00)	45.83 (21.34)	1.86 (4.39)	5.42 (8.03)	27.04 (90.21)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	74.01 (38.47)	2.00 (6.45)	7.65 (13.30)	44.32 (170.63)
$n = 1000, R^2 = 0.15$						
10^{-3}	0.00 (0.00)	0.00 (0.00)	1.28 (0.11)	1.04 (0.11)	1.02 (0.27)	1.12 (1.66)
10^{-4}	0.00 (0.00)	0.00 (0.01)	3.77 (0.59)	1.12 (0.34)	1.26 (0.64)	2.22 (3.83)
10^{-5}	0.00 (0.00)	0.00 (0.03)	19.95 (4.38)	1.40 (1.19)	2.63 (2.03)	5.04 (10.00)
10^{-6}	0.00 (0.00)	0.00 (0.00)	121.86 (35.41)	1.96 (4.43)	8.54 (9.96)	13.72 (38.86)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	210.91 (66.31)	2.24 (6.62)	12.79 (16.95)	19.48 (67.64)
$n = 200, R^2 = 0.2$						
10^{-3}	0.00 (0.00)	0.00 (0.00)	1.15 (0.10)	1.06 (0.12)	0.89 (0.35)	1.55 (1.84)
10^{-4}	0.00 (0.00)	0.00 (0.01)	1.70 (0.40)	1.14 (0.36)	0.82 (0.71)	3.07 (4.31)
10^{-5}	0.00 (0.00)	0.00 (0.02)	3.80 (1.91)	1.34 (1.19)	1.16 (1.54)	6.99 (11.14)
10^{-6}	0.00 (0.00)	0.00 (0.00)	11.82 (10.73)	1.77 (4.19)	2.27 (5.21)	17.58 (33.71)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	17.19 (18.46)	1.99 (6.35)	3.01 (8.37)	24.01 (51.12)
$n = 500, R^2 = 0.2$						
10^{-3}	0.00 (0.00)	0.00 (0.00)	1.24 (0.11)	1.06 (0.11)	1.00 (0.29)	1.25 (1.73)
10^{-4}	0.00 (0.00)	0.00 (0.01)	2.86 (0.52)	1.15 (0.35)	1.17 (0.67)	2.51 (4.04)
10^{-5}	0.00 (0.00)	0.00 (0.03)	12.01 (3.40)	1.38 (1.21)	2.13 (1.90)	6.03 (11.38)
10^{-6}	0.00 (0.00)	0.00 (0.00)	62.58 (24.80)	1.95 (4.47)	6.26 (8.69)	18.98 (61.52)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	103.84 (45.14)	2.21 (6.73)	9.00 (14.57)	28.79 (113.49)
$n = 1000, R^2 = 0.2$						
10^{-3}	0.00 (0.00)	0.00 (0.00)	1.30 (0.11)	1.04 (0.11)	1.02 (0.27)	1.15 (1.68)
10^{-4}	0.00 (0.00)	0.00 (0.01)	4.18 (0.62)	1.12 (0.35)	1.29 (0.66)	2.27 (3.86)
10^{-5}	0.00 (0.00)	0.00 (0.04)	23.64 (4.72)	1.42 (1.20)	2.81 (2.14)	5.16 (9.88)
10^{-6}	0.00 (0.00)	0.00 (0.00)	151.06 (39.11)	2.02 (4.52)	9.42 (10.51)	13.62 (32.29)
5×10^{-7}	0.00 (0.00)	0.00 (0.00)	265.69 (73.67)	2.30 (6.75)	14.27 (18.08)	18.74 (50.90)

Table S5: The mean and standard deviation of the ratio of the false positive rates to the nominal significance level based on 2,000 replications using Sobel's test, MaxP, JT-comp, HDMT, Sobel-comp and DACT under the *Null 2(b)* scenario. n is the sample size. The total number of mediators is 100,000. With probability $\pi_{01} = 0.33$, $\alpha = 0$ and $\beta \sim N(0, 0.3^2)$; with probability $\pi_{10} = 0.33, \alpha \sim N(0, 5 \times 0.3^2)$ and $\beta = 0$; with probability $\pi_{00} = 0.34, \alpha = \beta = 0$. R^2 is controlled in the mediator and outcome models.

Cut-off	Sobel	MaxP	JT-comp	HDMT	Sobel-comp	DACT
			$n = 200, R^2 = 0.1$			
10^{-3}	0.00 (0.00)	0.17 (0.04)	1.57 (0.12)	1.08 (0.10)	0.01 (0.01)	2.70 (1.19)
10^{-4}	0.00 (0.00)	0.11 (0.11)	1.42 (0.37)	1.14 (0.34)	0.00 (0.01)	4.66 (2.88)
10^{-5}	0.00 (0.00)	0.07 (0.27)	1.10 (1.06)	1.20 (1.09)	0.00 (0.00)	8.32 (6.96)
10^{-6}	0.00 (0.00)	0.05 (0.71)	0.81 (2.87)	1.21 (3.57)	0.00 (0.00)	14.97 (18.81)
5×10^{-7}	0.00 (0.00)	0.06 (1.09)	0.71 (3.81)	1.36 (5.31)	0.00 (0.00)	18.27 (27.09)
			$n = 500, R^2 = 0.1$			
10^{-3}	0.02 (0.01)	0.30 (0.05)	1.61 (0.12)	1.05 (0.11)	0.07 (0.03)	9.51 (2.18)
10^{-4}	0.00 (0.02)	0.27 (0.16)	1.11 (0.34)	1.08 (0.33)	0.01 (0.04)	24.86 (7.54)
10^{-5}	0.00 (0.03)	0.25 (0.50)	0.56 (0.76)	1.20 (1.14)	0.00 (0.05)	67.52 (25.92)
10^{-6}	0.00 (0.00)	0.24 (1.55)	0.28 (1.65)	1.25 (3.53)	0.00 (0.00)	188.08 (93.66)
5×10^{-7}	0.00 (0.00)	0.21 (2.04)	0.22 (2.09)	1.32 (5.16)	0.00 (0.00)	257.10 (137.86)
			$n = 1000, R^2 = 0.1$			
10^{-3}	0.07 (0.03)	0.38 (0.06)	1.61 (0.12)	1.03 (0.10)	0.18 (0.04)	15.62 (2.39)
10^{-4}	0.02 (0.05)	0.34 (0.19)	0.95 (0.30)	1.04 (0.32)	0.06 (0.08)	46.50 (9.38)
10^{-5}	0.00 (0.07)	0.30 (0.55)	0.38 (0.62)	1.06 (1.03)	0.02 (0.13)	142.55 (36.00)
10^{-6}	0.00 (0.22)	0.29 (1.66)	0.11 (1.02)	1.05 (3.27)	0.01 (0.32)	446.62 (141.02)
5×10^{-7}	0.01 (0.45)	0.30 (2.43)	0.07 (1.18)	1.00 (4.36)	0.01 (0.45)	633.64 (217.43)
			$n = 200, R^2 = 0.15$			
10^{-3}	0.00 (0.01)	0.22 (0.05)	1.78 (0.13)	1.10 (0.10)	0.02 (0.02)	6.59 (2.01)
10^{-4}	0.00 (0.01)	0.17 (0.13)	1.65 (0.41)	1.16 (0.34)	0.00 (0.01)	15.57 (6.34)
10^{-5}	0.00 (0.00)	0.13 (0.38)	1.25 (1.15)	1.25 (1.12)	0.00 (0.00)	38.03 (20.06)
10^{-6}	0.00 (0.00)	0.08 (0.89)	0.92 (3.01)	1.45 (3.91)	0.00 (0.00)	95.13 (64.76)
5×10^{-7}	0.00 (0.00)	0.08 (1.26)	0.83 (4.09)	1.47 (5.52)	0.00 (0.00)	125.84 (93.35)
			$n = 500, R^2 = 0.15$			
10^{-3}	0.04 (0.02)	0.34 (0.06)	1.86 (0.13)	1.05 (0.11)	0.11 (0.03)	14.48 (2.42)
10^{-4}	0.01 (0.03)	0.31 (0.18)	1.39 (0.38)	1.09 (0.33)	0.03 (0.06)	42.63 (9.34)
10^{-5}	0.00 (0.04)	0.30 (0.54)	0.75 (0.88)	1.20 (1.11)	0.01 (0.08)	129.53 (35.48)
10^{-6}	0.00 (0.00)	0.29 (1.72)	0.37 (1.89)	1.21 (3.46)	0.00 (0.00)	404.50 (139.21)
5×10^{-7}	0.00 (0.00)	0.25 (2.22)	0.28 (2.35)	1.36 (5.34)	0.00 (0.00)	571.34 (211.64)
			$n = 1000, R^2 = 0.15$			
10^{-3}	0.10 (0.03)	0.42 (0.07)	1.89 (0.14)	1.03 (0.10)	0.22 (0.05)	19.83 (2.27)
10^{-4}	0.04 (0.07)	0.38 (0.20)	1.26 (0.35)	1.04 (0.32)	0.10 (0.10)	62.99 (9.48)
10^{-5}	0.02 (0.13)	0.35 (0.59)	0.56 (0.75)	1.07 (1.03)	0.04 (0.19)	204.73 (38.65)
10^{-6}	0.01 (0.32)	0.35 (1.83)	0.15 (1.22)	1.10 (3.30)	0.02 (0.39)	682.91 (162.65)
5×10^{-7}	0.01 (0.45)	0.34 (2.59)	0.12 (1.54)	1.03 (4.42)	0.02 (0.63)	981.69 (252.83)
			$n = 200, R^2 = 0.2$			
10^{-3}	0.01 (0.01)	0.26 (0.05)	1.98 (0.14)	1.11 (0.11)	0.03 (0.02)	10.35 (2.39)
10^{-4}	0.00 (0.01)	0.21 (0.15)	1.91 (0.44)	1.19 (0.35)	0.00 (0.02)	28.13 (8.56)
10^{-5}	0.00 (0.00)	0.18 (0.43)	1.47 (1.24)	1.28 (1.13)	0.00 (0.00)	79.63 (30.55)
10^{-6}	0.00 (0.00)	0.09 (0.94)	1.06 (3.25)	1.51 (3.94)	0.00 (0.00)	229.66 (111.23)
5×10^{-7}	0.00 (0.00)	0.09 (1.34)	0.98 (4.41)	1.62 (5.74)	0.00 (0.00)	317.71 (167.34)
			$n = 500, R^2 = 0.2$			
10^{-3}	0.05 (0.02)	0.37 (0.06)	2.09 (0.14)	1.05 (0.11)	0.14 (0.04)	17.98 (2.36)
10^{-4}	0.02 (0.04)	0.35 (0.19)	1.68 (0.42)	1.10 (0.33)	0.05 (0.07)	56.18 (9.56)
10^{-5}	0.01 (0.08)	0.34 (0.57)	0.99 (1.02)	1.19 (1.10)	0.01 (0.12)	180.60 (38.45)
10^{-6}	0.00 (0.00)	0.33 (1.87)	0.50 (2.20)	1.19 (3.46)	0.01 (0.32)	594.44 (159.87)
5×10^{-7}	0.00 (0.00)	0.31 (2.47)	0.46 (3.00)	1.31 (5.22)	0.00 (0.00)	853.38 (248.24)
			$n = 1000, R^2 = 0.2$			
10^{-3}	0.12 (0.04)	0.45 (0.07)	2.13 (0.15)	1.03 (0.10)	0.26 (0.05)	22.38 (2.07)
10^{-4}	0.06 (0.08)	0.41 (0.20)	1.59 (0.39)	1.05 (0.32)	0.13 (0.12)	73.34 (8.98)
10^{-5}	0.02 (0.15)	0.39 (0.62)	0.79 (0.90)	1.06 (1.01)	0.06 (0.23)	245.90 (37.71)
10^{-6}	0.01 (0.32)	0.40 (1.97)	0.23 (1.48)	1.11 (3.31)	0.02 (0.39)	840.57 (165.04)
5×10^{-7}	0.02 (0.63)	0.39 (2.77)	0.18 (1.89)	1.05 (4.51)	0.02 (0.63)	1220.59 (258.69)

Table S6: The mean and standard deviation of the ratio of the false positive rates to the nominal significance level based on 2,000 replications using Sobel's test, MaxP, JT-comp, HDMT, Sobel-comp and DACT under the *Null 2(c)* scenario. n is the sample size. The total number of mediators is 100,000. With probability $\pi_{01} = 0.5$, $\alpha = 0$ and $\beta \sim N(0, 0.3^2)$ and with probability $\pi_{10} = 0.5, \alpha \sim N(0, 5 \times 0.3^2)$ and $\beta = 0$. R^2 is controlled in the mediator and outcome models.

Cut-off	Sobel	MaxP	JT-comp	HDMT	Sobel-comp	DACT
$n = 200, R^2 = 0.1$						
10^{-3}	0.00 (0.00)	0.25 (0.05)	0.78 (0.09)	1.08 (0.10)	0.01 (0.01)	1.45 (0.73)
10^{-4}	0.00 (0.00)	0.16 (0.12)	0.41 (0.20)	1.14 (0.33)	0.00 (0.00)	1.97 (1.37)
10^{-5}	0.00 (0.00)	0.11 (0.33)	0.17 (0.41)	1.19 (1.07)	0.00 (0.00)	2.76 (2.80)
10^{-6}	0.00 (0.00)	0.06 (0.74)	0.05 (0.71)	1.35 (3.69)	0.00 (0.00)	3.91 (7.15)
5×10^{-7}	0.00 (0.00)	0.05 (1.00)	0.05 (1.00)	1.33 (5.22)	0.00 (0.00)	4.33 (10.31)
$n = 500, R^2 = 0.1$						
10^{-3}	0.03 (0.02)	0.46 (0.07)	0.59 (0.08)	1.05 (0.10)	0.05 (0.02)	8.23 (1.79)
10^{-4}	0.01 (0.02)	0.40 (0.20)	0.19 (0.14)	1.11 (0.33)	0.01 (0.03)	18.42 (5.13)
10^{-5}	0.00 (0.02)	0.36 (0.61)	0.04 (0.21)	1.20 (1.08)	0.00 (0.02)	42.73 (15.32)
10^{-6}	0.00 (0.00)	0.28 (1.77)	0.01 (0.32)	1.26 (3.50)	0.00 (0.00)	102.86 (50.33)
5×10^{-7}	0.00 (0.00)	0.22 (2.18)	0.00 (0.00)	1.21 (4.97)	0.00 (0.00)	134.24 (75.09)
$n = 1000, R^2 = 0.1$						
10^{-3}	0.11 (0.03)	0.57 (0.08)	0.50 (0.07)	1.03 (0.10)	0.15 (0.04)	15.28 (2.03)
10^{-4}	0.04 (0.06)	0.52 (0.23)	0.11 (0.10)	1.06 (0.33)	0.05 (0.07)	39.06 (6.77)
10^{-5}	0.01 (0.09)	0.48 (0.69)	0.02 (0.12)	1.08 (1.04)	0.01 (0.11)	101.97 (22.53)
10^{-6}	0.00 (0.00)	0.38 (1.98)	0.00 (0.00)	1.00 (3.14)	0.00 (0.00)	272.92 (80.91)
5×10^{-7}	0.00 (0.00)	0.27 (2.31)	0.00 (0.00)	0.94 (4.42)	0.00 (0.00)	367.48 (122.55)
$n = 200, R^2 = 0.15$						
10^{-3}	0.01 (0.01)	0.33 (0.06)	0.84 (0.09)	1.10 (0.10)	0.01 (0.01)	4.96 (1.62)
10^{-4}	0.00 (0.00)	0.25 (0.16)	0.44 (0.21)	1.18 (0.34)	0.00 (0.01)	9.77 (4.11)
10^{-5}	0.00 (0.00)	0.19 (0.44)	0.16 (0.40)	1.30 (1.13)	0.00 (0.00)	19.78 (10.89)
10^{-6}	0.00 (0.00)	0.14 (1.18)	0.05 (0.67)	1.39 (3.72)	0.00 (0.00)	41.29 (31.50)
5×10^{-7}	0.00 (0.00)	0.09 (1.34)	0.04 (0.89)	1.39 (5.36)	0.00 (0.00)	51.42 (44.47)
$n = 500, R^2 = 0.15$						
10^{-3}	0.06 (0.02)	0.52 (0.07)	0.68 (0.08)	1.05 (0.10)	0.09 (0.03)	14.02 (2.18)
10^{-4}	0.02 (0.04)	0.47 (0.22)	0.23 (0.15)	1.11 (0.33)	0.02 (0.05)	35.54 (7.09)
10^{-5}	0.00 (0.02)	0.43 (0.66)	0.05 (0.22)	1.17 (1.07)	0.00 (0.04)	92.78 (22.98)
10^{-6}	0.00 (0.00)	0.32 (1.87)	0.00 (0.22)	1.16 (3.34)	0.00 (0.00)	248.55 (82.36)
5×10^{-7}	0.00 (0.00)	0.26 (2.35)	0.00 (0.00)	1.17 (4.98)	0.00 (0.00)	337.78 (124.90)
$n = 1000, R^2 = 0.15$						
10^{-3}	0.15 (0.04)	0.63 (0.08)	0.62 (0.08)	1.03 (0.10)	0.20 (0.04)	20.22 (2.07)
10^{-4}	0.07 (0.08)	0.59 (0.24)	0.16 (0.12)	1.06 (0.33)	0.09 (0.09)	54.79 (7.41)
10^{-5}	0.02 (0.15)	0.54 (0.73)	0.02 (0.15)	1.09 (1.04)	0.03 (0.17)	151.30 (25.91)
10^{-6}	0.00 (0.22)	0.40 (2.04)	0.00 (0.00)	0.99 (3.17)	0.00 (0.22)	424.46 (99.29)
5×10^{-7}	0.00 (0.00)	0.29 (2.39)	0.00 (0.00)	0.91 (4.36)	0.00 (0.00)	580.04 (151.97)
$n = 200, R^2 = 0.2$						
10^{-3}	0.01 (0.01)	0.38 (0.06)	0.92 (0.09)	1.11 (0.10)	0.02 (0.02)	9.12 (2.19)
10^{-4}	0.00 (0.01)	0.32 (0.18)	0.49 (0.22)	1.21 (0.34)	0.00 (0.02)	21.26 (6.51)
10^{-5}	0.00 (0.00)	0.26 (0.51)	0.19 (0.43)	1.34 (1.14)	0.00 (0.00)	51.03 (19.59)
10^{-6}	0.00 (0.00)	0.21 (1.43)	0.04 (0.63)	1.45 (3.89)	0.00 (0.00)	125.84 (63.68)
5×10^{-7}	0.00 (0.00)	0.20 (1.99)	0.03 (0.77)	1.54 (5.66)	0.00 (0.00)	166.69 (92.63)
$n = 500, R^2 = 0.2$						
10^{-3}	0.08 (0.03)	0.57 (0.08)	0.78 (0.09)	1.06 (0.10)	0.11 (0.03)	18.18 (2.21)
10^{-4}	0.03 (0.05)	0.52 (0.23)	0.29 (0.18)	1.11 (0.33)	0.04 (0.06)	48.87 (7.67)
10^{-5}	0.00 (0.06)	0.49 (0.71)	0.07 (0.26)	1.17 (1.07)	0.01 (0.08)	134.32 (26.17)
10^{-6}	0.00 (0.00)	0.36 (1.97)	0.00 (0.22)	1.15 (3.34)	0.00 (0.00)	377.49 (99.53)
5×10^{-7}	0.00 (0.00)	0.30 (2.51)	0.00 (0.00)	1.14 (4.93)	0.00 (0.00)	518.23 (151.98)
$n = 1000, R^2 = 0.2$						
10^{-3}	0.19 (0.04)	0.68 (0.08)	0.73 (0.09)	1.03 (0.10)	0.23 (0.05)	23.12 (2.01)
10^{-4}	0.09 (0.10)	0.64 (0.25)	0.22 (0.14)	1.06 (0.33)	0.12 (0.11)	64.32 (7.50)
10^{-5}	0.04 (0.19)	0.59 (0.77)	0.04 (0.19)	1.08 (1.03)	0.05 (0.22)	181.69 (27.34)
10^{-6}	0.01 (0.32)	0.46 (2.16)	0.00 (0.00)	1.04 (3.20)	0.01 (0.32)	520.44 (107.54)
5×10^{-7}	0.00 (0.00)	0.33 (2.55)	0.00 (0.00)	0.89 (4.22)	0.01 (0.45)	716.24 (165.67)

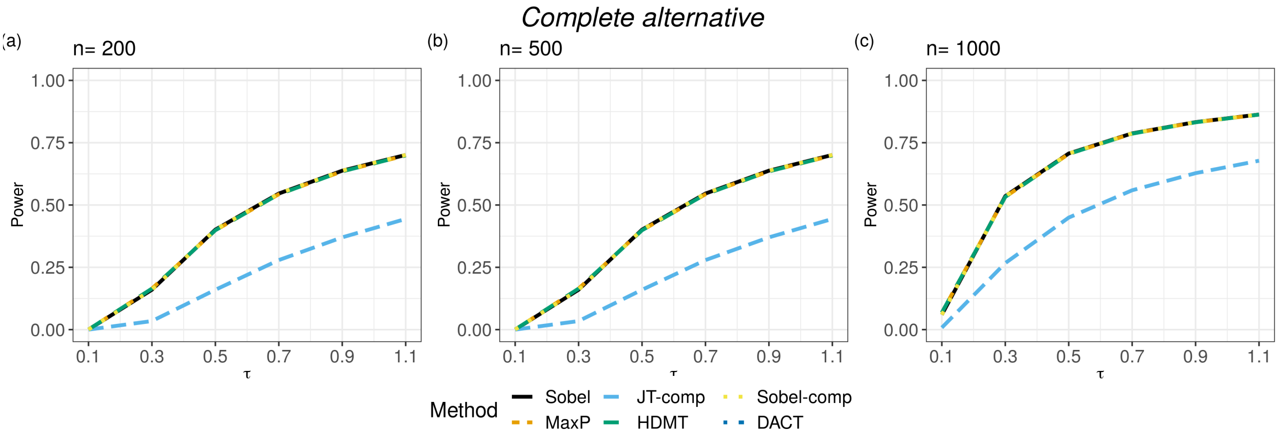

Figure S1: The average true positive rate over 200 replicates when controlling the true false discovery rate (FDR) at 0.05 for Sobel's test, MaxP, JT-comp, HDMT, Sobel-comp and DACT under the *Alternative 1(c)* scenario. The total number of mediators is 100,000. n is the sample size. For $j = 1, 2, ..., 100,000$, with probability $\pi_{11} = 0.2$, $\alpha_j \sim N(0, 5\tau^2), \beta_j \sim N(0, \tau^2)$; with probability $\pi_{01} = 0.4$, $\alpha_j = 0$ and $\beta_j \sim N(0, \tau^2)$; and with probability $\pi_{10} = 0.4, \alpha_j \sim N(0, 5\tau^2)$ and $\beta_j = 0$.

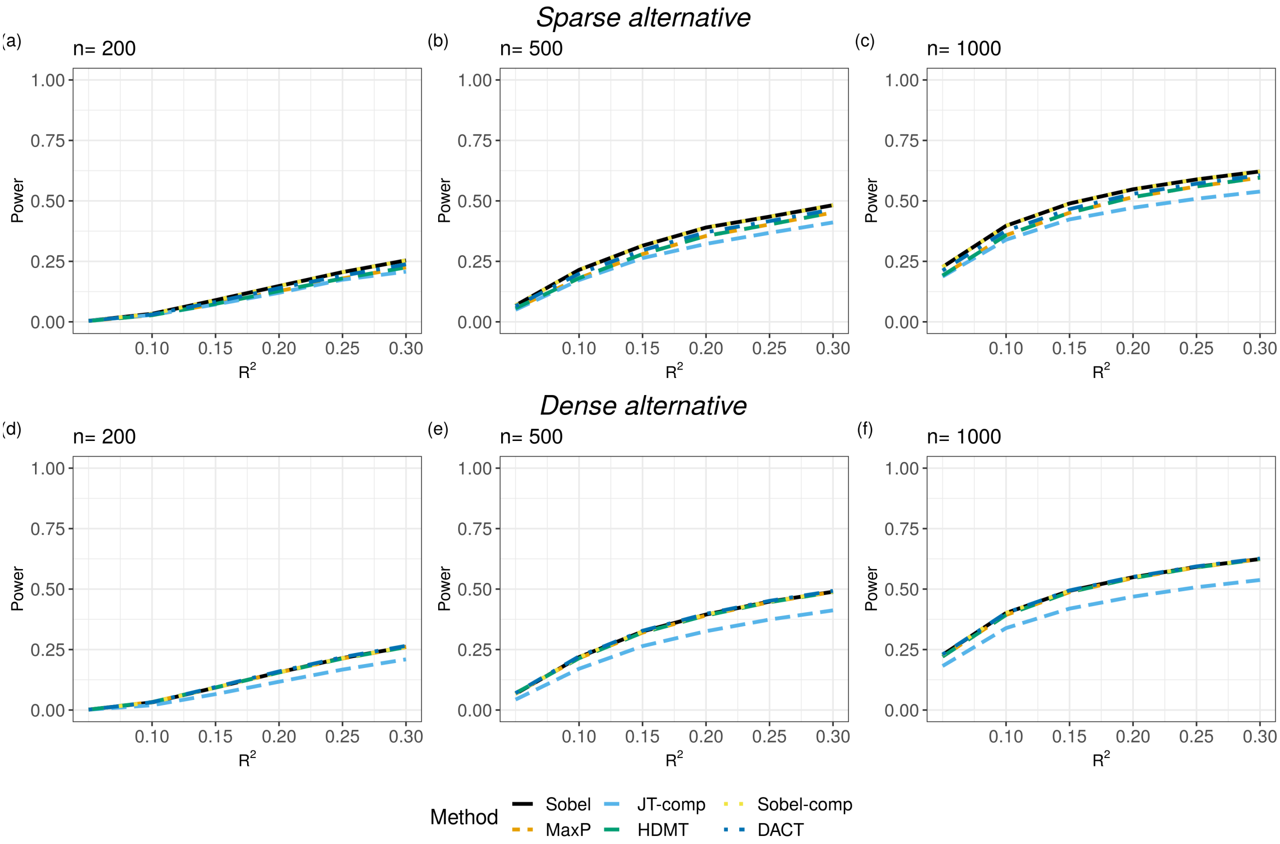

Figure S2: The average true positive rate over 200 replicates when controlling the true false discovery rate (FDR) at 0.05 for Sobel's test, MaxP, JT-comp, HDMT, Sobel-comp and DACT under the *Alternative 2(a)* and *Alternative 2(b)* scenarios. The total number of mediators is 100,000. n is the sample size. For $j = 1, 2, ..., 100,000$, with probability π_{11}, $\alpha_j \sim N(0, 5 \times 0.3^2), \beta_j \sim N(0, 0.3^2)$; with probability π_{01}, $\alpha_j = 0$ and $\beta_j \sim N(0, 0.3^2)$; with probability $\pi_{10}, \alpha_j \sim N(0, 5 \times 0.3^2)$ and $\beta_j = 0$; with probability $\pi_{00}, \alpha_j = \beta_j = 0$. R^2 is controlled in the j-th mediator and outcome models. Under the *Alternative 2(a)* scenario, $\pi_{11}, \pi_{10}, \pi_{01}, \pi_{00}$ are set as $0.001, 0.001, 0.001, 0.997$ and under the *Alternative 2(b)* scenario, $\pi_{11}, \pi_{10}, \pi_{01}, \pi_{00}$ are set as $0.2, 0.2, 0.2, 0.4$.

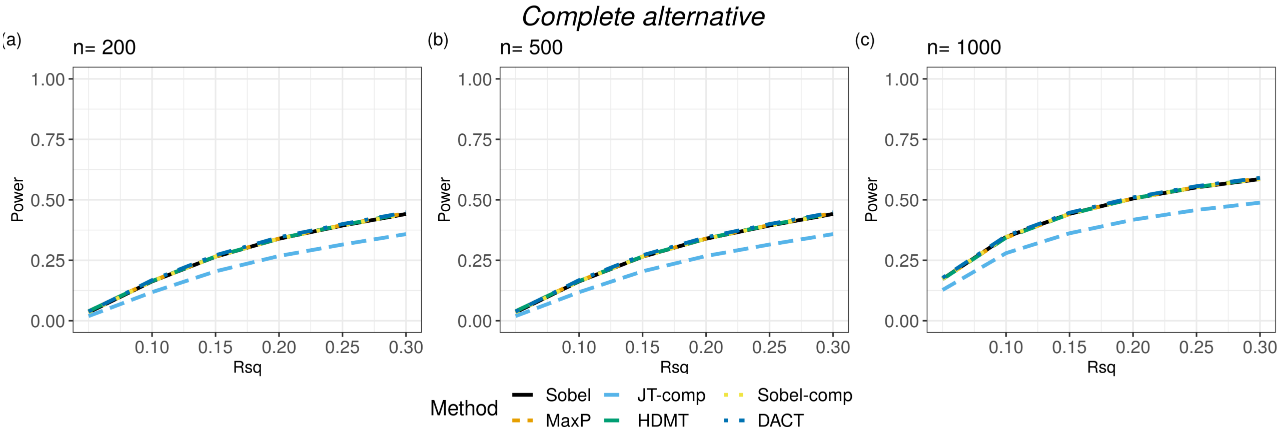

Figure S3: The average true positive rate over 200 replicates when controlling the true false discovery rate (FDR) at 0.05 for Sobel's test, MaxP, JT-comp, HDMT, Sobel-comp and DACT under the *Alternative 2(c)* scenario. The total number of mediators is 100,000. n is the sample size. For $j = 1, 2, ..., 100, 000$, with probability $\pi_{11} = 0.2$, $\alpha_j \sim N(0, 5 \times 0.3^2), \beta_j \sim N(0, 0.3^2)$; with probability $\pi_{01} = 0.4$, $\alpha_j = 0$ and $\beta_j \sim N(0, 0.3^2)$; and with probability $\pi_{10} = 0.4, \alpha_j \sim N(0, 5 \times 0.3^2)$ and $\beta_j = 0$. R^2 is controlled in the j-th mediator and outcome models.

Table S7: Three mediation pathways identified by HDMT after controlling the FWER at 0.05. The exposure is adult SES and the outcome is HbA1c. The total number of mediators is 228,088. In both models, we adjusted for age, sex, race and residual white blood cell proportions (neutrophils, B cells, T cells, and natural killer cells). In addition, we adjusted for the exposure in the outcome model. α is the effect of the exposure on the mediator (conditional on the confounders) and β is the effect of the mediator on the outcome (conditional on the exposure and the confounders).

CpG	Chr	Gene	$\hat{\alpha}$	$\hat{\beta}$	p_{HDMT}
cg01288337	14	RIN3	2.26E-01	1.51E-01	6.47E-08
cg10508317	17	SOCS3	-2.81E-01	-1.25E-01	6.49E-08
cg07571519	10	C10orf105	2.46E-01	1.26E-01	2.05E-07

Table S8: The natural indirect effect (NIE) and the proportion of mediation effect (Med. Proportion) of the exposure (adult SES) on the outcome (HbA1c) through the mediator (DNA methylation measured at a CpG site) with and without the exposure-mediator interaction in the outcome model. In the mediator and outcome models, we adjust for age, sex, race and residual white blood cell proportions (neutrophils, B cells, T cells, and natural killer cells). In addition, we adjust for the exposure in the outcome model. γ: the exposure-mediator interaction effect on the outcome. NIE: natural indirect effect. Med. Prop: proportion of the mediation effect.

	Without interaction		With interaction		
	NIE (95% CI)	Med. Proportion (95% CI)	NIE (95% CI)	Med. Proportion (95% CI)	$\hat{\gamma}$ (p-value)
cg10508317	0.035 (0.013, 0.064)	0.178 (0.063, 0.599)	0.036 (0.014, 0.063)	0.175 (0.032, 0.593)	0.002 (0.98)
cg01288337	0.034 (0.013, 0.061)	0.169 (0.055, 0.540)	0.033 (0.011, 0.061)	0.162 (0.016, 0.619)	0.012 (0.87)

Table S9: Mediation pathways identified by the multivariate mediation analysis method (HIMA (Zhang et al., 2016)) and the single-mediator hypothesis testing methods (JT-comp (Huang et al., 2019) and HDMT (Dai et al., 2022)). In both models, we adjust for age, sex, race and residual white blood cell proportions (neutrophils, B cells, T cells, and natural killer cells). In addition, we adjust for the exposure in the outcome model. The p-values under HIMA were obtained using the joint significance test. NIE: natural indirect effect. Med. Prop: proportion of the mediation effect.

CpG	$\hat{\alpha}$	$\hat{\beta}$	NIE	p-value	Med. Prop
HIMA					
cg10508317	-0.28	-0.052	0.015	4.45E-04	0.07
cg01288337	0.23	0.048	0.011	1.04E-03	0.05
cg06555281	0.25	0.038	0.009	1.99E-03	0.05
HDMT					
cg10508317	-0.28	-0.12	0.035	6.49E-08	0.18
cg01288337	0.23	0.15	0.034	6.47E-08	0.17
cg07571519	0.25	0.13	0.031	2.05E-07	0.16
JT-comp					
cg10508317	-0.28	-0.12	0.035	1.19E-07	0.18
cg01288337	0.23	0.15	0.034	1.85E-07	0.17

Table S10: Bidirectional mediation analysis for adult SES, DNA methylation and HbA1c. Mediation analysis is performed for two pathways: $SES \rightarrow DNAm \rightarrow HbA1c$ and $SES \rightarrow HbA1c \rightarrow DNAm$. We adjust for seven confounders in the model, including age, sex, race and residual white blood cell proportions (neutrophils, B cells, T cells, and natural killer cells). α is the effect of the exposure on the mediator (conditional on the confounders) and β is the effect of the mediator on the outcome (conditional on the exposure and the confounders). TE: total effect. NIE: natural indirect effect. Med. Prop: proportion of the mediation effect.

CpG	Direction	$\hat{\alpha}$	$\hat{\beta}$	TE (95% CI)	p-value	NIE (95% CI)	p-value	Med. Prop (95% CI)
cg10508317	$SES \rightarrow DNAm \rightarrow HbA1c$	-0.28	-0.12	0.20 (0.06, 0.34)	0.004	0.035 (0.013,0.063)	<0.001	0.173 (0.055, 0.591)
	$SES \rightarrow HbA1c \rightarrow DNAm$	0.20	-0.12	-0.28 (-0.42,-0.14)	<0.001	-0.023 (-0.047,-0.005)	0.008	0.076 (0.018, 0.213)
cg01288337	$SES \rightarrow DNAm \rightarrow HbA1c$	0.23	0.15	0.20 (0.07, 0.34)	0.006	0.034 (0.013,0.063)	<0.001	0.167 (0.060,0.479)
	$SES \rightarrow HbA1c \rightarrow DNAm$	0.20	0.11	0.23 (0.11, 0.35)	<0.001	0.022 (0.005, 0.044)	0.002	0.093 (0.022, 0.231)

References

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