Mendelian Randomization Theory

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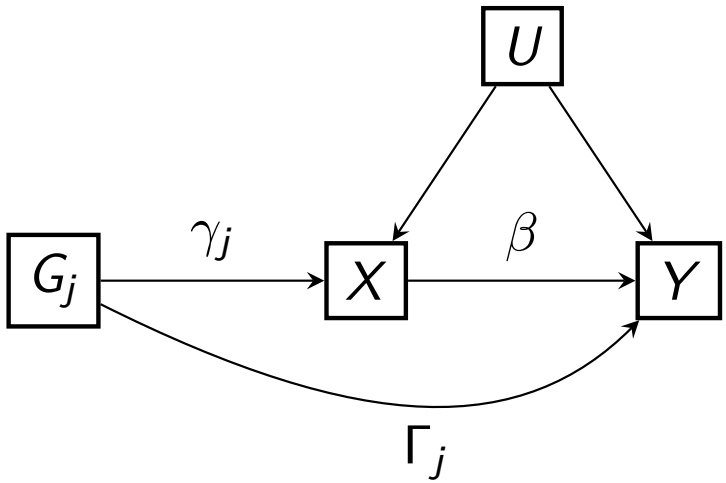
Table of Contents

General modeling framework



- G_j: Genetic variant (SNP) j
- X: Exposure
- Y: Outcome
- U: Confounder

- γ_j: SNP-Exposure association

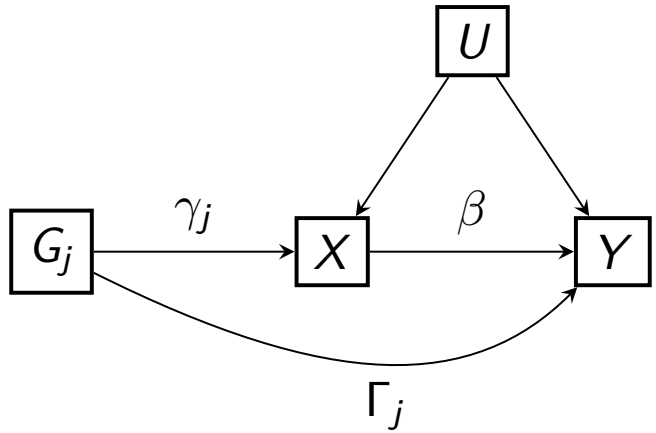
$$\hat{\gamma}_j = \mathrm{lm}(X \sim G_j) \tag{1}$$

- Γ_j: SNP-Outcome association

$$\hat{\Gamma}_j = \mathrm{lm}(Y \sim G_j) \tag{2}$$

- β: Exposure-Outcome association, aka the **causal estimate**, what MR tries to find.

Structural equations



- **Structural equations for variant** G_j

$$X = \gamma_j G_j + U^X + \varepsilon_j^X, \tag{3}$$

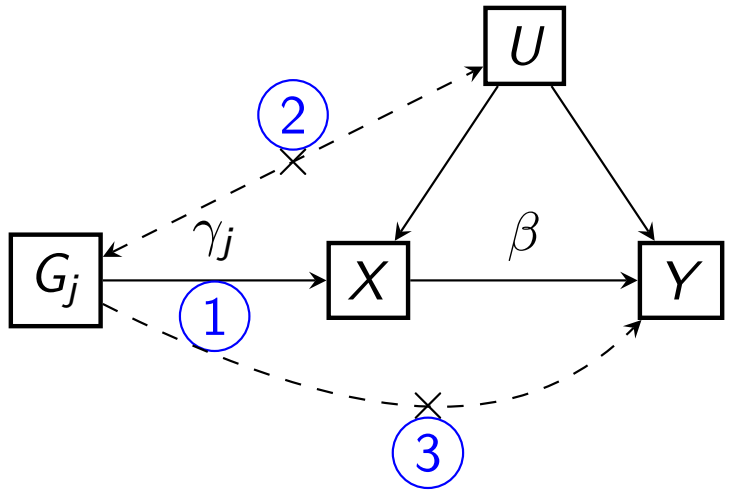
$$Y = \beta X + \alpha_j G_j + U^Y + \varepsilon_j^Y \tag{4}$$

- The association between G_j and Y can be decomposed as

$$\Gamma_j = \gamma_j \beta \; + \; \alpha_j, \tag{5}$$

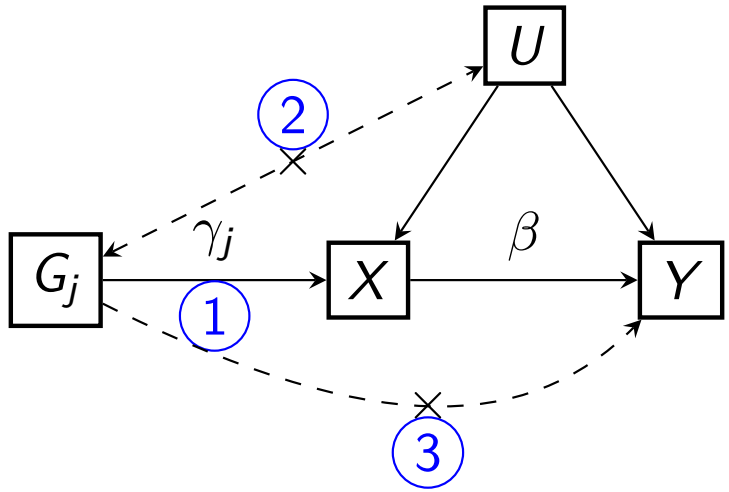
where α_j captures any direct (**pleiotropic**) effect of G_j on Y not mediated through X.

Instrumental variables (IV) assumptions



- IV1 (Relevance): The genetic variant is robustly associated with the exposure. Implies $\gamma_j \neq 0$.
- IV2 (Independence): The genetic variant is independent of confounders U;
- IV3 (Exclusion restriction): The genetic variant is independent of the outcome Y conditional on the exposure X and confounders U. Implies $\alpha_j = 0$

IV assumptions examined



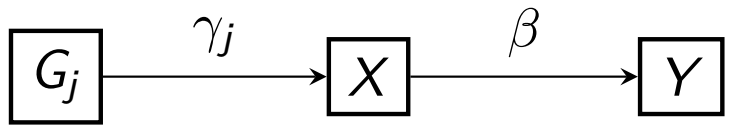
- IV1 (Relevance): Supported by GWAS.
- IV2 (Independence): Guaranteed by Mendel's law of independent assortment (under LD clamping). Only concern is population stratification.
- IV3 (Exclusion restriction): Often problematic due to **pleiotropy**.

Pleiotropy: vertical vs horizontal

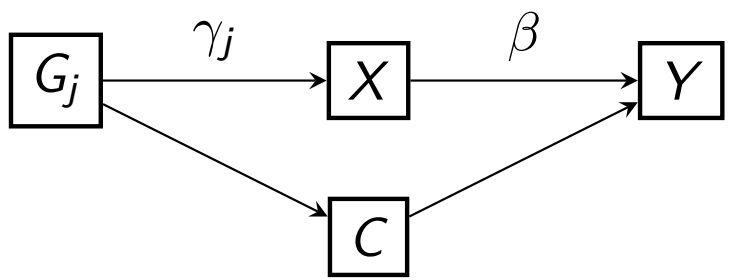
A genetic variant G_j is *pleiotropic* if it influences more than one phenotype. In the linear MR model:

$$\Gamma_j \;=\; \beta\,\gamma_j \;+\; \alpha_j, \tag{6}$$

- **Vertical pleiotropy**: variant affects multiple traits via the same causal chain, i.e. $\alpha_j = 0$ (valid IV)



- **Horizontal pleiotropy**: variant also affects outcome through a pathway that does not run through the exposure, $\alpha_j \neq 0$ (invalid IV)



Horizontal pleiotropy

Balanced vs. directional pleiotropy

- **Balanced pleiotropy**: Average pleiotropic effect is zero, $\mathbb{E}[\alpha_j] = 0$.
- **Directional pleiotropy**: Non-zero average pleiotropic effect, $\mathbb{E}[\alpha_j] \neq 0$.

Uncorrelated vs. correlated pleiotropy

- **Uncorrelated pleiotropy**: Instrument Strength Independent of Direct Effect (aka InSIDE), $\text{Cov}(\gamma_j, \alpha_j) = 0$.
- **Correlated pleiotropy**: $\text{Cov}(\gamma_j, \alpha_j) \neq 0$. This also violates IV2 (Independence).

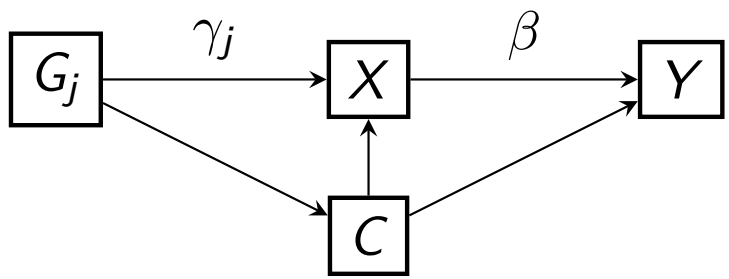


Table of Contents

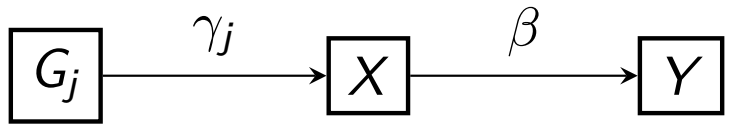
Individual-Level Data

- **What does it look like?** It's typically a large table or spreadsheet where you have one record per participant:

Participant ID	SNP (rs123)	SNP (rs456)	... (SNPs)	Exposure (X)	Outcome (Y)	Covariates
001	1	0	...	120.5	1 (Case)	...
002	0	2	...	115.2	0 (Control)	...
003	2	1	...	131.0	1 (Case)	...
...	...	...	...	...	...	...

- Matrix G: for person i and SNP j G with $G_{ij} \in \{0, 1, 2\}$ = count of the *effect* allele.
- This format enables flexible and powerful analyses, like the two-stage least squares (TSLS) method, but accessing such data can be challenging due to privacy and logistical reasons.

Two-stage least-squares (TSLS)



- **First stage regression**

$$X = \gamma_0 + G\boldsymbol{\gamma} + \varepsilon^X \tag{7}$$

Estimate predicted exposure $\widehat{X}$ via OLS.

- **Second stage regression**

$$Y = \beta_0 + \beta\widehat{X} + \varepsilon^Y \tag{8}$$

Regress outcome Y on predicted exposure $\widehat{X}$, yielding the estimated exposure-outcome association $\hat{\beta}$.

- Under IV assumptions, $\hat{\beta}$ is a **consistent estimator** of the causal effect.
- **Potential problem:** TSLS may suffer from low power and for γ_j small from the "many weak instruments bias", see Davies et al. [2015].

Allele Scores (Genetic Risk Scores)

- Allele scores can then be used to increase power and reduce the risk from many weak instruments.
- Combines J independent SNPs into one genetic instrument.

$$S_i^{\text{unweighted}} = \sum_{j=1}^{J} G_{ij} \qquad S_i^{\text{weighted}} = \sum_{j=1}^{J} w_j \, G_{ij} \tag{9}$$

- Replace first stage regression with

$$X = \gamma_0 + \gamma \, S + \varepsilon^X \tag{10}$$

"Risk-increasing" (effect) allele

- Before scoring, reorient each SNP so all effect alleles push the phenotype in the *same* direction; otherwise opposite signs cancel and attenuate power[1].

[1] SNP reorientation can be problematic under directional pleiotropy, see MR-GRIP.

Table of Contents

Motivation

GWAS summary statistics

SNP	b.exposure	b.outcome	se.outcome	se.exposure
rs1000940	0.018	-0.0091	0.0162	0.0033
rs10132280	-0.022	-0.0169	0.0164	0.0033
rs1016287	-0.022	-0.0254	0.0155	0.0033
rs10182181	0.030	0.0009	0.0143	0.0029
rs10733682	-0.018	-0.0212	0.0159	0.0030
rs10840100	0.020	-0.0133	0.0143	0.0030

- All methods discussed herein are founded upon Wald ratios.
- The essence involves constructing statistical tests from the properties of the sampling distribution of Wald ratio statistics.
- Typically, this is accomplished within a "precision-weighted space".

Wald Ratio (single-variant MR estimate)

- Definition

$$\hat{\beta}_{\text{Wald}} = \frac{\hat{\Gamma}_j}{\hat{\gamma}_j} \tag{11}$$

- Under IV assumptions, the **Wald ratio** provides a *consistent* estimate of the causal effect β.
- Using the **Delta method** and assuming $\Gamma_j \equiv \beta_j \gamma_j$ with precise instruments ($\sigma^2_{\hat{\gamma}_j}$ small)[2], we can obtain

$$\hat{\beta}_j = \frac{\hat{\Gamma}_j}{\hat{\gamma}_j} \sim \mathcal{N}(\beta, \sigma^2_{\hat{\beta}_j}) \tag{12}$$

with

$$\sigma^2_{\hat{\beta}_j} \approx \frac{\sigma^2_{\hat{\Gamma}_j}}{\hat{\gamma}_j^2} \tag{13}$$

[2] This is sometimes called the NOME (NO Measurement Error) assumption

MR methods

- Each SNP produces an independent Wald estimator: $\hat{\beta}_j = \hat{\Gamma}_j / \hat{\gamma}_j$.

$$\hat{\beta}_j = \frac{\hat{\Gamma}_j}{\hat{\gamma}_j} \sim \mathcal{N}(\beta, \sigma^2_{\hat{\beta}_j}) \tag{14}$$

- Combine individual Wald ratio estimates in different ways:

1. Inverse-variance weighting

$$\hat{\beta}_{\text{IVW}} = \text{Mean}(\hat{\beta}_j, \text{weights} = 1/\sigma^2_{\hat{\beta}_j}) \tag{15}$$

2. MR-Egger regression

$$\hat{\beta}_{\text{Egger}} = \text{lm}(\hat{\Gamma}_j \sim \hat{\gamma}_j, \text{weights} = 1/\sigma^2_{\hat{\Gamma}_j}) \tag{16}$$

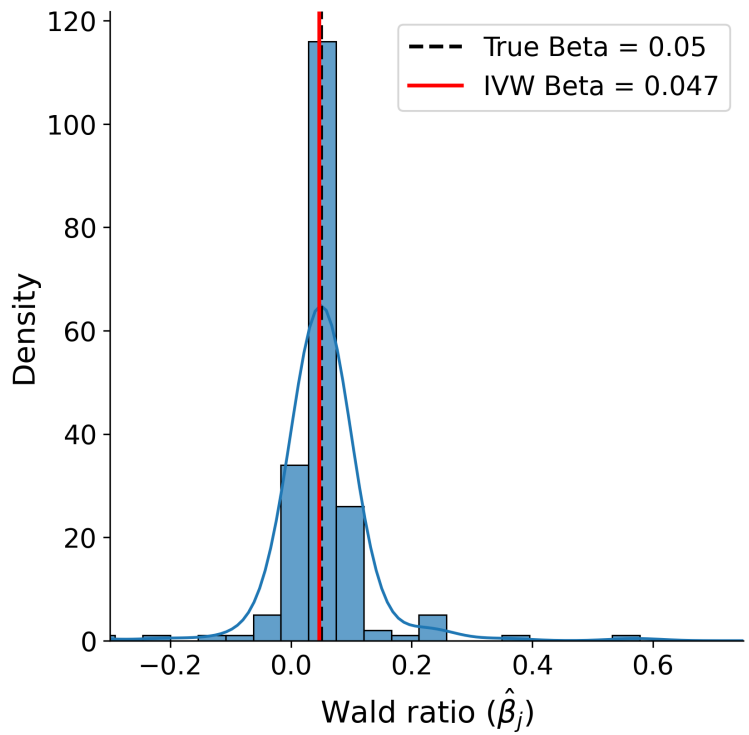
3. Weighted median

$$\hat{\beta}_{\text{WM}} = \text{Median}(\hat{\beta}_j, \text{weights} = 1/\sigma^2_{\hat{\beta}_j}) \tag{17}$$

Inverse-variance weighted (IVW) estimate

- IVW Idea: Weigh SNP Wald ratios by the inverse variance of the Wald ratios $w_j = \sigma^{-2}_{\hat{\beta}_j} \approx \widehat{\gamma}_j^2 / \sigma^2_{\hat{\Gamma}_j}$.
- The IVW causal estimate is then the weighted arithmetic mean:

$$\hat{\beta}_{\mathrm{IVW}} = \frac{\sum_j w_j \hat{\beta}_j}{\sum_j w_j} = \frac{\sum_j \sigma^{-2}_{\hat{\Gamma}_j} \hat{\Gamma}_j \hat{\gamma}_j}{\sum_j \sigma^{-2}_{\hat{\Gamma}_j} \hat{\gamma}_j^2}. \tag{18}$$

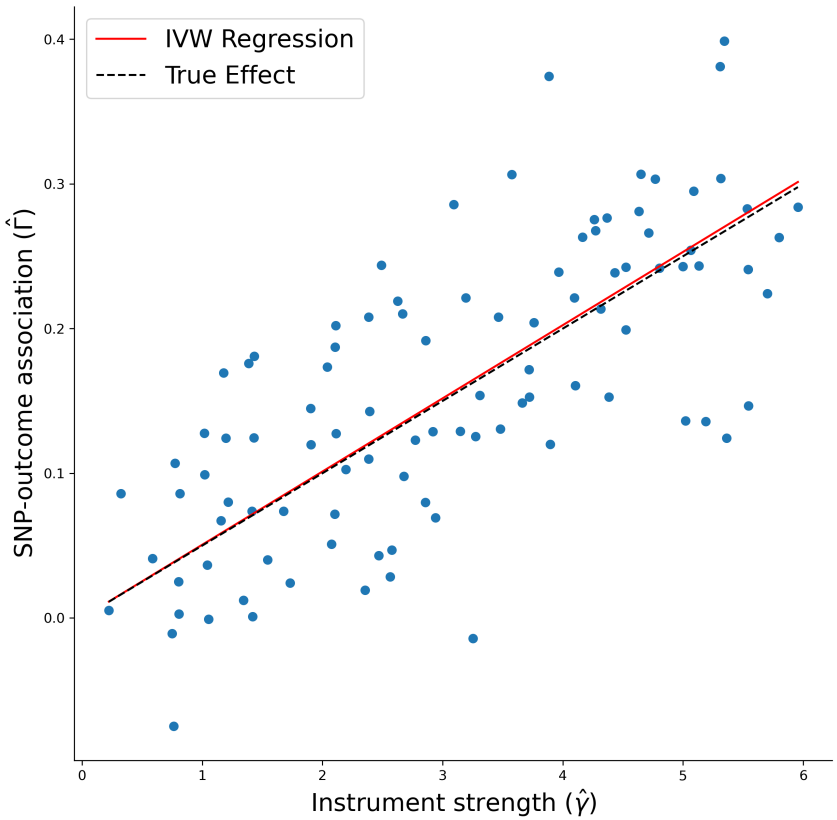


IVW - Weighted Least Squares (WLS) perspective

- Consider linear model

$$\widehat{\Gamma}_j = \beta\,\widehat{\gamma}_j + \nu_j, \qquad \nu_j \sim \mathcal{N}\big(0, \sigma^2_{\hat{\Gamma}_j}\big), \tag{19}$$

with weights $\sigma^{-2}_{\hat{\Gamma}_j}$.



IVW - WLS loss function

- Weighted least squares (WLS) minimizes the loss function

$$Q(\beta) = \sum_j \sigma_{\hat{\Gamma}_j}^{-2} \left(\widehat{\Gamma}_j - \beta\,\widehat{\gamma}_j\right)^2. \tag{20}$$

- Recall that we let $w_j = \widehat{\gamma}_j^2/\sigma_{\hat{\Gamma}_j}^2$, and after rearranging we get

$$Q(\beta) = \sum_j w_j \left(\hat{\beta}_j - \beta\right)^2 \tag{21}$$

which yields

$$\arg\min_{\beta}\ Q(\beta) = \frac{\sum_j w_j\,\hat{\beta}_j}{\sum_j w_j} \tag{22}$$

$$\equiv \hat{\beta}_{\mathrm{IVW}}. \tag{23}$$

IVW – WLS loss & Cochran's Q-test

- **Weighted-least-squares loss**

$$Q(\beta) = \sum_j w_j \left(\hat{\beta}_j - \beta\right)^2 \tag{24}$$

- **Cochran's Q-test (heterogeneity)** At $\beta = \hat{\beta}_{\mathrm{IVW}}$,

$$w_j^{1/2}(\hat{\beta}_j - \hat{\beta}_{\mathrm{IVW}}) \sim \mathcal{N}(0,1) \tag{25}$$

And therefore asymptotically[3] (large sample)

$$Q(\hat{\beta}_{\mathrm{IVW}}) \sim \chi^2_{J-1} \tag{26}$$

A large Q (small p-value) indicates residual variance beyond sampling error, i.e. heterogeneity / pleiotropy.

[3] Special case of Cochran's Theorem

IVW is unbiased under balanced pleiotropy and InSIDE

$$\hat{\beta}_{\text{IVW}} = \frac{\sum_j v_j \hat{\gamma}_j \hat{\Gamma}_j}{\sum_j v_j \hat{\gamma}_j^2}, \qquad v_j := \frac{1}{\hat{\sigma}^2_{\hat{\Gamma}_j}}. \tag{27}$$

- Plug in pleiotropy model and rewrite in terms of bias:

$$\hat{\beta}_{\text{IVW}} = \frac{\sum_j v_j \hat{\gamma}_j (\beta \hat{\gamma}_j + \alpha_j)}{\sum_j v_j \hat{\gamma}_j^2} = \beta + \frac{\sum_j v_j \hat{\gamma}_j \alpha_j}{\sum_j v_j \hat{\gamma}_j^2}. \tag{28}$$

- When $\mathbb{E}[\alpha_j] = 0$ and $\text{Cov}(\hat{\gamma}_j, \alpha_j) = 0$ we can apply Slutsky's Theorem to show that

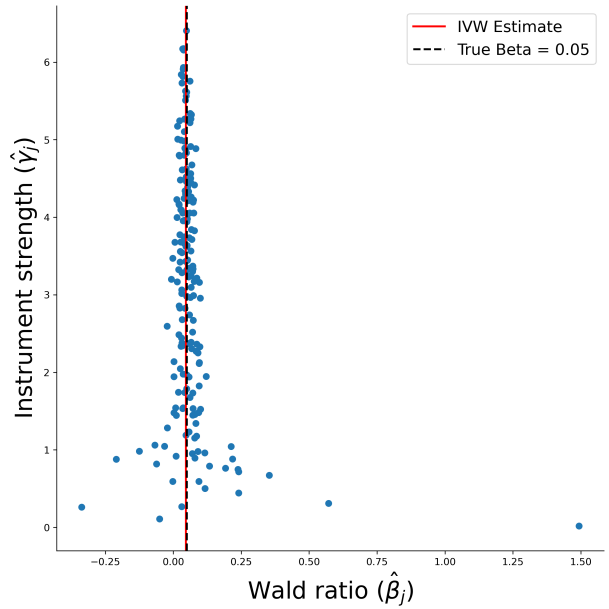
$$\mathbb{E}[\hat{\beta}_{\text{IVW}}] = \beta \tag{29}$$

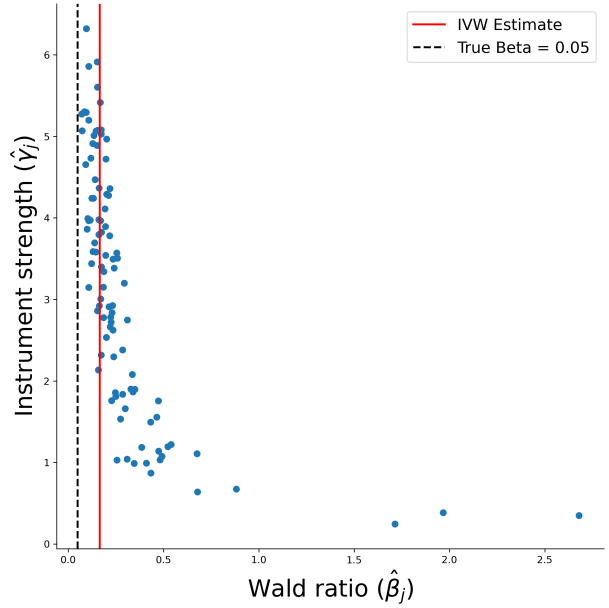
Funnel plots - visualizing pleiotropy

- Funnel plot shows precision of causal estimates vs. the causal estimates themselves.
- Under *balanced* pleiotropy the cloud is symmetric around the true causal effect line.
- **Directional pleiotropy** tilts or shifts the funnel $\Rightarrow$ IVW may be biased.
- Potential solution: estimate intercept

$$\hat{\Gamma}_j = \beta\,\hat{\gamma}_j + \text{intercept} + \nu_j \quad (30)$$

A non-zero intercept $\Rightarrow$ evidence of directional pleiotropy.

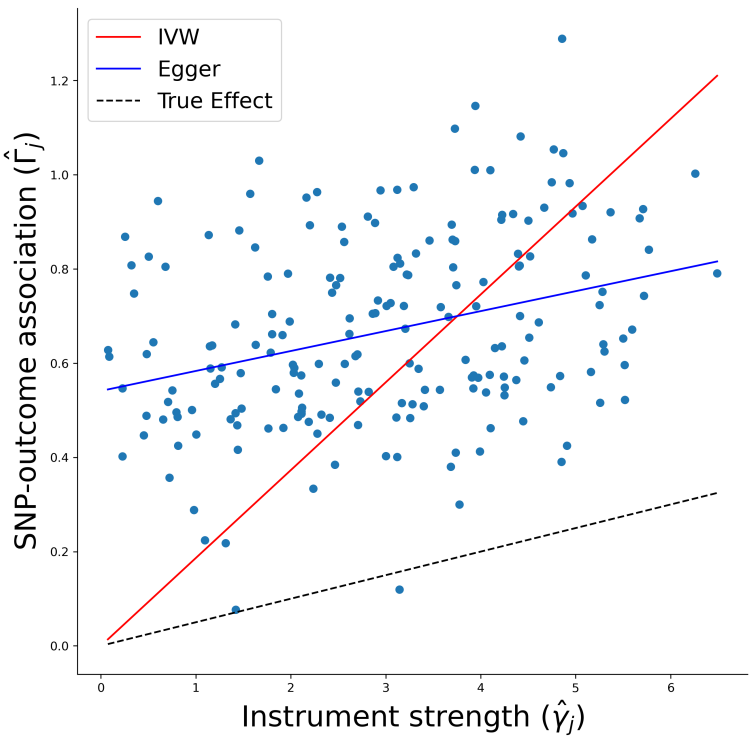




Detecting directional pleiotropy with MR-Egger

$$\hat{\Gamma}_j \;=\; \beta_{0E} \;+\; \beta_E\,\hat{\gamma}_j + \nu_j \tag{31}$$

- Estimate coefficient $\hat{\beta}_E$ and intercept $\hat{\beta}_{0E}$ via WLS with weights $v_j = \sigma^{-2}_{\hat{\Gamma}_j}$.
- Intercept estimate $\hat{\beta}_{0E}$ represents the *average pleiotropic effect*.
- A non-zero $\hat{\beta}_{0E} \implies$ evidence of overall **directional pleiotropy**.



Egger estimate is unbiased under the InSIDE assumption

- Consider OLS Egger regression, the slope estimate is given by

$$\hat{\beta}_E = \frac{\text{cov}(\hat{\Gamma}, \hat{\gamma})}{\text{var}(\hat{\gamma})} = \beta + \frac{\text{cov}(\hat{\alpha}, \hat{\gamma})}{\text{var}(\hat{\gamma})}. \tag{32}$$

- Under InSIDE ($\text{Cov}(\alpha, \gamma) = 0$) the bias term vanishes as

$$\text{Cov}(\hat{\alpha}, \hat{\gamma}) \overset{N\to\infty}{\longrightarrow} \text{Cov}(\alpha, \gamma) \overset{J\to\infty}{\longrightarrow} 0, \tag{33}$$

  and $\hat{\beta}_E$ is a *consistent estimator* of the true causal effect β.

- Violating InSIDE couples pleiotropy with instrument strength, biasing the slope.

- A similar bias decomposition can be performed for Egger estimates via WLS with weights $v_j = \sigma_{\hat{\Gamma}_j}^{-2}$.

Weighted-median MR

Robust alternative to IVW

- IVW is biased under directional pleiotropy.
- In weighted-median MR invalid variants appear as **outliers**; the median resists influence of those extremes, ensuring robustness.
- *Key identifying assumption*: at least $\geq 50\%$ of the *total inverse-variance weight* comes from valid (non-pleiotropic) instruments.
- Procedure
  1. Construct a weighted emperical distribution of $\hat{\beta}_j$'s using inverse-variance weights.
  2. Return the *weighted median* (i.e. 50th percentile).

Share your Feedback!

Scan the QR code to let us know your thoughts on this tutorial. Thank you!

Extra slides

Delta-method - Theorem (univariate)

- Given a (weak) convergence of a sequence of random variables X_n to a standard normal

$$\sqrt{n}(X_n - \theta) \xrightarrow{D} \mathcal{N}(0, \sigma^2) \tag{34}$$

- we can compute the convergence of a smooth transformation $f(X_n)$

$$\sqrt{n}(f(X_n) - f(\theta)) \xrightarrow{D} \mathcal{N}(0, \sigma^2 \cdot f'(\theta)^2) \tag{35}$$

- Thus, for large n we can estimate

$$\mathrm{SE}(\hat{\beta}_j) = \mathrm{SE}(f(\hat{\Gamma}_j, \hat{\gamma}_j) \equiv \mathrm{SE}(\frac{\hat{\Gamma}_j}{\hat{\gamma}_j}). \tag{36}$$

Delta-method setup

Wald ratio for a single SNP j

$$\hat{\beta}_j = \frac{\hat{\Gamma}_j}{\hat{\gamma}_j} \tag{37}$$

Let $f(\hat{\Gamma}_j, \hat{\gamma}_j) = \hat{\Gamma}_j / \hat{\gamma}_j$ be a new random variable with an associated sampling distribution:

$$\begin{pmatrix} \hat{\Gamma}_j \\ \hat{\gamma}_j \end{pmatrix} \sim \mathcal{N}\left(\begin{pmatrix} \Gamma_j \\ \gamma_j \end{pmatrix}, \Sigma_j \right) \tag{38}$$

- *Next:* Study its theoretical properties.
- What's Σ_j?
- Main stats tool: **Delta method** - use first order Taylor expansion of $\hat{\beta}_\text{j}$ around its mean $E\hat{\beta}_j = \beta_j$.

Derivation of SE

- Joint sampling covariance of $(\hat{\Gamma}_j, \hat{\gamma}_j)^\top$:

$$\Sigma_j = \begin{pmatrix} \sigma^2_{\Gamma j} & \rho_j \, \sigma_{\Gamma j} \sigma_{\gamma j} \\ \rho_j \, \sigma_{\Gamma j} \sigma_{\gamma j} & \sigma^2_{\gamma j} \end{pmatrix} \tag{39}$$

- Gradient at (Γ_j, γ_j):

$$\nabla f(\Gamma_j, \gamma_j) = \left(\frac{1}{\gamma_j}, \; -\frac{\Gamma_j}{\gamma_j^2} \right)^\top \tag{40}$$

- Delta-method variance formula:

$$(\mathrm{SE}(\hat{\beta}_j))^2 = \mathrm{Var}(\hat{\beta}_j) \approx [\nabla f(\hat{\Gamma}_j, \hat{\gamma}_j)]^\top \; \Sigma_j \; [\nabla f(\hat{\Gamma}_j, \hat{\gamma}_j)] \tag{41}$$

$$= \frac{\sigma^2_{\Gamma j}}{\hat{\gamma}_j^2} + \frac{\hat{\Gamma}_j^2 \, \sigma^2_{\gamma j}}{\hat{\gamma}_j^4} - \frac{2 \, \hat{\Gamma}_j \, \rho_j \, \sigma_{\Gamma j} \sigma_{\gamma j}}{\hat{\gamma}_j^3}. \tag{42}$$

Two-sample vs. one-sample MR

$$\mathrm{Var}(\hat{\beta}_j) \approx \frac{\sigma^2_{\Gamma j}}{\hat{\gamma}_j^2} + \frac{\hat{\Gamma}_j^2\,\sigma^2_{\gamma j}}{\hat{\gamma}_j^4} - \frac{2\,\hat{\Gamma}_j\,\rho_j\,\sigma_{\Gamma j}\sigma_{\gamma j}}{\hat{\gamma}_j^3}. \tag{43}$$

One-sample MR (same cohort)

$$\rho_j \neq 0 \;\Rightarrow\; \mathrm{SE}(\hat{\beta}_j) = \sqrt{\frac{\sigma^2_{\Gamma j}}{\hat{\gamma}_j^2} + \frac{\hat{\Gamma}_j^2\,\sigma^2_{\gamma j}}{\hat{\gamma}_j^4} - \frac{2\,\hat{\Gamma}_j\,\rho_j\,\sigma_{\Gamma j}\sigma_{\gamma j}}{\hat{\gamma}_j^3}}. \tag{44}$$

Two-sample MR (independent cohorts)

$$\rho_j = 0 \;\Rightarrow\; \mathrm{SE}(\hat{\beta}_j) = \sqrt{\frac{\sigma^2_{\Gamma j}}{\hat{\gamma}_j^2} + \frac{\hat{\Gamma}_j^2\,\sigma^2_{\gamma j}}{\hat{\gamma}_j^4}}. \tag{45}$$

If $\sigma^2_{\gamma_j}$ is negligible (precise $\hat{\gamma}_j$):

$$\mathrm{SE}(\hat{\beta}_j) \approx \frac{\sigma_{\Gamma j}}{|\hat{\gamma}_j|}. \tag{46}$$

Take-away: covariance term present in one-sample MR, zero in two-sample MR.

References I

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