4. Results

Contents

Word count: 589

Results

Simulation Study

Data Simulation

Data were successfully simulated as intended. A selection of representative visualisations are presented in Figure 1. Full details of testing used to validate model outputs from parameter inputs are given in Appendix **??**. The F-statistic calculated from simulated instruments was >10, indicating that they were sufficiently strongly associated with exposure to meet the relevance assumption of instrumental variable (IV) analysis (Tables 1 and 2).

Analysis of Simulated Data

No Causal Effect

Across all cases where no causal effect was present (Table 1), the mean rate of reporting a causal effect (i.e. false positive rate) for MR-Hevo was 0.45% versus 3.6% for weighted median estimator (WME). Of the 20 combinations of scenarios and parameters, MR-Hevo exhibited a favourable false positive rate versus WME in 20 (100%). For both MR-Hevo and WME methods, causal report rate generally increased with an increasing proportion of invalid instruments, and also as assumption violations progressively presented greater data variability and bias across scenarios 1 to 3.

The mean causal effect estimate (mean reported 95% confidence interval (CI)s) across all cases was 0.041 (-0.12 to 0.21) for MR-Hevo and 0.034 (-0.12 to 0.19) for WME. The mean (range) standard error (SE) of causal effect estimates across all cases was 0.0012 (0 to 0.002) for MR-Hevo and 0.078 (0.056 to 0.099). For both MR-Hevo and WME methods, causal effect estimates, width of CIs and SE all tended to increase slightly both with an increasing proportion of invalid instruments, and as assumption violations progressively presented greater data variability and bias across scenarios 1 to 3.

Positive Causal Effect

Across all cases where positive causal effect was present (Table 2), the mean rate of reporting a causal effect (i.e. sensitivity) for MR-Hevo was 31% versus 28% for WME. Of the 24 combinations of scenarios and parameters, MR-Hevo exhibited a favourable sensitivity versus WME in 10 (42%). For both MR-Hevo and WME methods, causal report rate generally increased with an increasing proportion of invalid instruments, and also as assumption violations progressively presented greater data variability and bias across scenarios 1 to 3.

The mean causal effect estimate (mean reported 95% CIs) across all cases was 0.13 (-0.025 to 0.3) for MR-Hevo and 0.11 (-0.039 to 0.26) for WME. The mean (range) SE of causal effect estimates across all cases was 0.0013 (0.001 to 0.002) for MR-Hevo and 0.077 (0.056 to 0.1). For both MR-Hevo and WME methods, causal effect estimates, width of CIs and SE all tended to increase slightly both with an increasing proportion of invalid instruments, and as assumption violations progressively presented greater data variability and bias across scenarios 1 to 3.

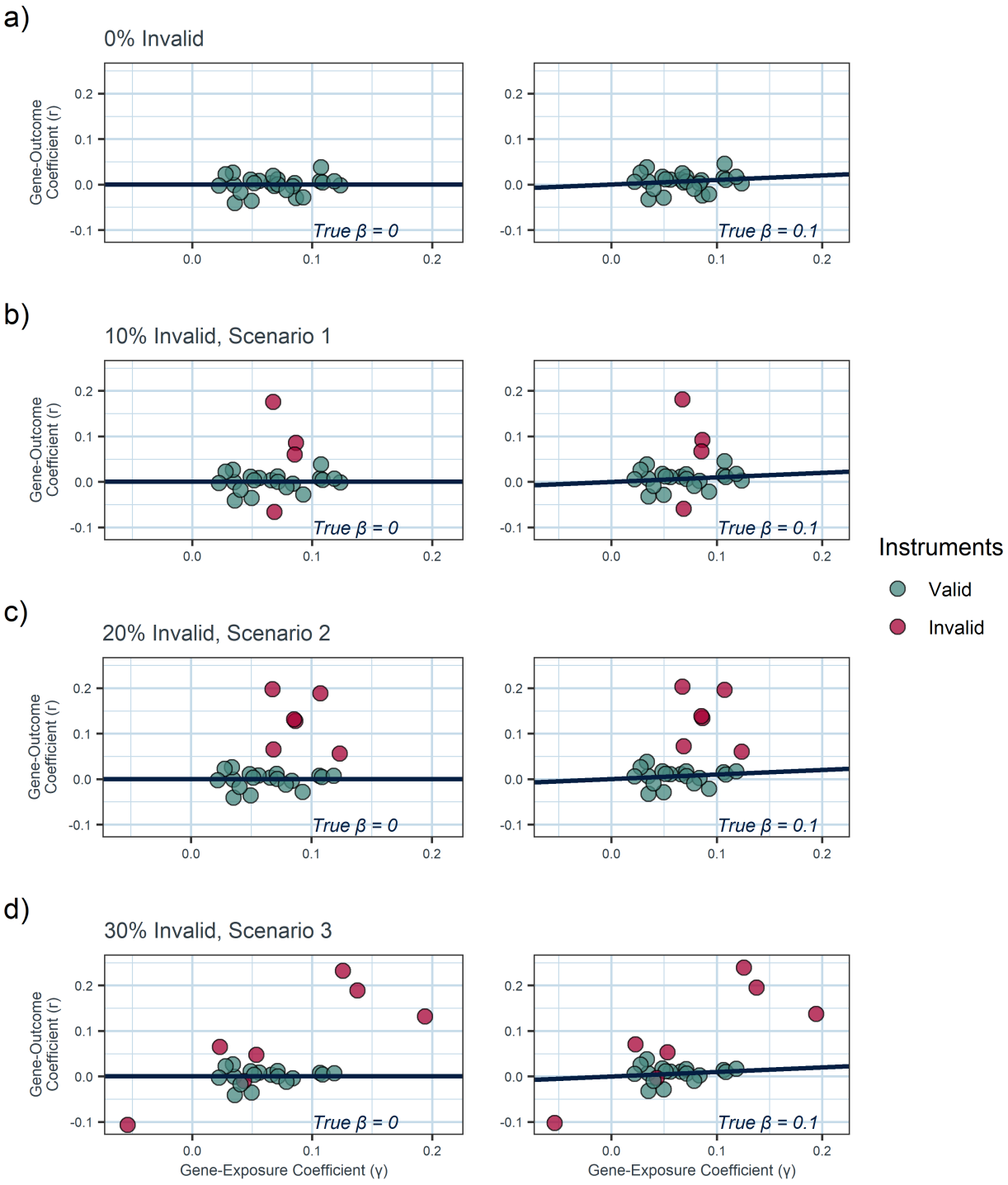

Figure 1: Plots of a representative group of simulated datasets; all simulate genetic instruments from the same index from the same random seed. Left and right columns demonstrate null and positive true causal effects, respectively. The scenario and the proportion of invalid (i.e. pleiotropic) genetic instruments changes with each row. a) 10% of instruments invalid, Scenario 1: balanced pleiotropy introduces noise around the causal effect. b) 20% of instruments invalid, Scenario 2: directional pleiotropy biases in the direction of the invalid instruments. c) 30% of instruments invalid, Scenario 3: directional pleiotropy and InSIDE assumption violation strongly biases towards a positive effect estimate.

Tables

Table 1: Summary of 1,000 simulated Mendelian randomisation studies per combination of scenario and parameters, all with null causal effect

				Weighted Median			MR Hevo		
N	Invalid IVs	*F*	R^2	Mean Estimate (Mean SE)	Mean 95% CI	Causal Report Rate	Mean Estimate (Mean SE)	Mean 95% CI	Causal Report Rate
Scenario 1: Balanced pleiotropy, InSIDE assumption satisfied									
10,000	0%	11.7	2.8%	0.001 (0.078)	-0.15 to 0.15	0.2%	0.000 (0.001)	-0.12 to 0.12	0%
10,000	10%	11.7	2.8%	0.026 (0.086)	-0.14 to 0.19	1.5%	0.032 (0.001)	-0.13 to 0.2	0%
10,000	20%	11.7	2.8%	0.022 (0.092)	-0.16 to 0.2	2%	0.037 (0.002)	-0.17 to 0.25	0%
10,000	30%	11.7	2.8%	0.014 (0.093)	-0.17 to 0.2	1.6%	0.022 (0.002)	-0.2 to 0.25	0%
20,000	0%	26.2	3.2%	0.003 (0.056)	-0.11 to 0.11	0.3%	0.001 (0)	-0.09 to 0.09	0%
20,000	10%	24.5	3%	0.022 (0.062)	-0.1 to 0.14	0.5%	0.019 (0.001)	-0.1 to 0.14	0.1%
20,000	20%	24.5	3%	0.020 (0.067)	-0.11 to 0.15	1.3%	0.022 (0.001)	-0.13 to 0.18	0%
20,000	30%	24.5	3%	0.012 (0.067)	-0.12 to 0.14	0.8%	0.014 (0.001)	-0.15 to 0.18	0%
Scenario 2: Directional pleiotropy, InSIDE assumption satisfied									
10,000	0%	11.7	2.8%	0.001 (0.078)	-0.15 to 0.15	0.3%	0.000 (0.001)	-0.12 to 0.12	0%
10,000	10%	11.7	2.8%	0.020 (0.087)	-0.15 to 0.19	0.8%	0.039 (0.001)	-0.13 to 0.22	0%
10,000	20%	11.7	2.8%	0.050 (0.093)	-0.13 to 0.23	4.1%	0.098 (0.002)	-0.11 to 0.33	1.5%
10,000	30%	11.7	2.8%	0.066 (0.094)	-0.12 to 0.25	5.8%	0.126 (0.002)	-0.09 to 0.38	3.6%
20,000	0%	24.5	3%	0.004 (0.056)	-0.11 to 0.11	0.2%	0.001 (0)	-0.08 to 0.09	0%
20,000	10%	24.5	3%	0.016 (0.062)	-0.11 to 0.14	0.7%	0.021 (0.001)	-0.1 to 0.15	0.1%
20,000	20%	24.5	3%	0.038 (0.067)	-0.09 to 0.17	2.2%	0.054 (0.001)	-0.1 to 0.22	0.5%
20,000	30%	24.5	3%	0.050 (0.068)	-0.08 to 0.18	4.9%	0.076 (0.002)	-0.08 to 0.25	1.2%
Scenario 3: Directional pleiotropy, InSIDE assumption not satisfied									
10,000	0%	11.7	2.8%	0.001 (0.078)	-0.15 to 0.15	0.2%	0.000 (0.001)	-0.12 to 0.12	0%
10,000	10%	13.7	3.3%	0.077 (0.087)	-0.09 to 0.25	8%	0.044 (0.001)	-0.12 to 0.21	0.1%
10,000	20%	14.9	3.6%	0.144 (0.099)	-0.05 to 0.34	24.7%	0.107 (0.002)	-0.1 to 0.35	1.3%
10,000	30%	12.8	3.1%	0.103 (0.097)	-0.09 to 0.29	11.9%	0.102 (0.002)	-0.11 to 0.36	0.6%

CI: Confidence Interval, IV: Instumental Variable, SE: Standard Error. Null Causal Effect (β = 0)

Table 2: Summary of 1,000 simulated Mendelian randomisation studies per combination of scenario and parameters, all with positive causal effect

				Weighted Median			MR Hevo		
N	Invalid IVs	*F*	R^2	Mean Estimate (Mean SE)	Mean 95% CI	Causal Report Rate	Mean Estimate (Mean SE)	Mean 95% CI	Causal Report Rate
Scenario 1: Balanced pleiotropy, InSIDE assumption satisfied									
10,000	0%	11.7	2.8%	0.070 (0.079)	-0.08 to 0.22	4.9%	0.085 (0.001)	-0.04 to 0.21	6.2%
10,000	10%	11.7	2.8%	0.094 (0.087)	-0.08 to 0.26	11%	0.118 (0.001)	-0.05 to 0.29	12.6%
10,000	20%	11.7	2.8%	0.089 (0.093)	-0.09 to 0.27	10.3%	0.124 (0.002)	-0.08 to 0.34	5.6%
10,000	30%	11.7	2.8%	0.081 (0.094)	-0.1 to 0.27	8.7%	0.108 (0.002)	-0.12 to 0.34	1.6%
20,000	0%	24.5	3%	0.080 (0.056)	-0.03 to 0.19	21.3%	0.089 (0.001)	0 to 0.18	62.2%
20,000	10%	24.5	3%	0.098 (0.063)	-0.03 to 0.22	27.8%	0.108 (0.001)	-0.01 to 0.23	29.9%
20,000	20%	24.5	3%	0.095 (0.067)	-0.04 to 0.23	22.6%	0.113 (0.001)	-0.04 to 0.27	15%
20,000	30%	24.5	3%	0.088 (0.068)	-0.05 to 0.22	17.7%	0.104 (0.001)	-0.06 to 0.27	5.4%
Scenario 2: Directional pleiotropy, InSIDE assumption satisfied									
10,000	0%	11.7	2.8%	0.070 (0.079)	-0.08 to 0.22	5.3%	0.085 (0.001)	-0.04 to 0.21	5.9%
10,000	10%	11.7	2.8%	0.089 (0.088)	-0.08 to 0.26	9%	0.124 (0.001)	-0.05 to 0.31	11.9%
10,000	20%	11.7	2.8%	0.119 (0.094)	-0.07 to 0.3	17.7%	0.187 (0.002)	-0.02 to 0.43	32.3%
10,000	30%	11.7	2.8%	0.133 (0.095)	-0.05 to 0.32	23.3%	0.216 (0.002)	0 to 0.47	46.1%
20,000	0%	24.5	3%	0.080 (0.057)	-0.03 to 0.19	21.1%	0.089 (0.001)	0 to 0.18	62.7%
20,000	10%	24.5	3%	0.093 (0.063)	-0.03 to 0.22	24%	0.109 (0.001)	-0.01 to 0.24	29.1%
20,000	20%	24.5	3%	0.116 (0.068)	-0.02 to 0.25	35.3%	0.146 (0.001)	-0.01 to 0.31	41.2%
20,000	30%	24.5	3%	0.127 (0.069)	-0.01 to 0.26	40.7%	0.168 (0.002)	0.01 to 0.35	56.2%
Scenario 3: Directional pleiotropy, InSIDE assumption not satisfied									
10,000	0%	11.7	2.8%	0.070 (0.079)	-0.08 to 0.22	5.2%	0.085 (0.001)	-0.04 to 0.21	5.7%
10,000	10%	13.7	3.3%	0.150 (0.089)	-0.02 to 0.32	35%	0.137 (0.001)	-0.03 to 0.31	25.1%
10,000	20%	14.9	3.6%	0.213 (0.1)	0.02 to 0.41	55.8%	0.202 (0.002)	-0.01 to 0.46	45.2%
10,000	30%	12.8	3.1%	0.169 (0.099)	-0.03 to 0.36	37.1%	0.191 (0.002)	-0.03 to 0.46	29.1%
20,000	0%	24.5	3%	0.080 (0.057)	-0.03 to 0.19	21.5%	0.089 (0.001)	0 to 0.18	62.8%
20,000	10%	30.4	3.7%	0.144 (0.064)	0.02 to 0.27	66%	0.125 (0.001)	0.01 to 0.25	63%
20,000	20%	32.4	3.9%	0.189 (0.073)	0.05 to 0.33	81.5%	0.154 (0.001)	0.01 to 0.32	58.6%
20,000	30%	31.1	3.8%	0.153 (0.071)	0.01 to 0.29	60.3%	0.146 (0.001)	-0.01 to 0.32	41%

CI: Confidence Interval, IV: Instumental Variable, SE: Standard Error. Null Causal Effect ($\beta = 0$)

Citations Search

A total of 110 abstracts and 54 full texts were screened to identify the 10 studies[1–10] listed in Table 3; the screening flow diagram is presented in Figure 2.

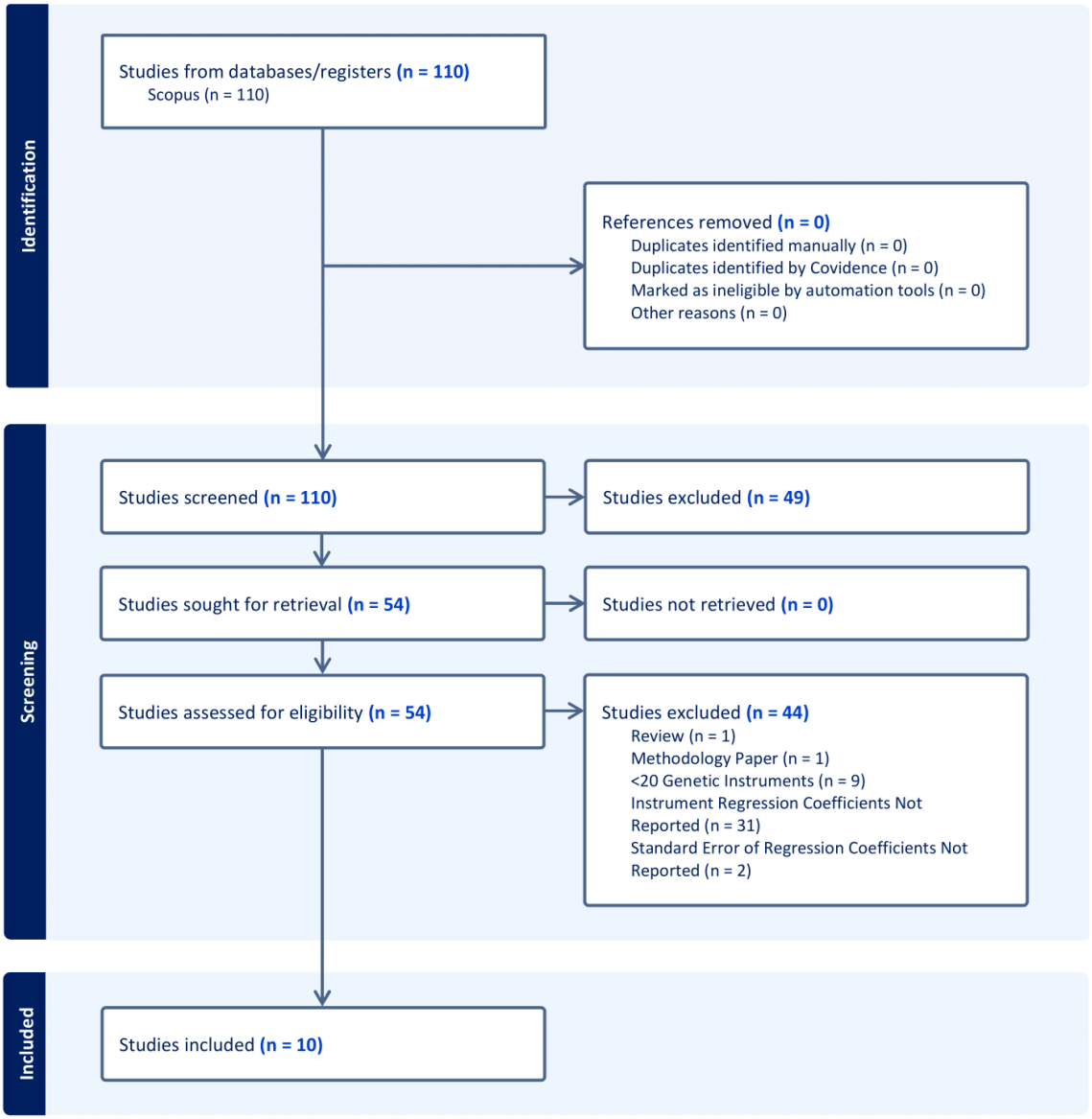

Figure 2: Flow diagram of selection for sample of ten highly-cited two-sample Mendelian randomisation articles reporting a weighted median estimate of casual effect

Table 3: Summary of ten highly-cited two-sample Mendelian randomisation articles reporting a weighted median estimate of casual effect

Author	Citations	Association	*N* Instruments	Participants			Causal Effect		Causality Reported	*p*-value
				N		Maximum Estimated Overlap				
				Exposure	Outcome		Measure	Estimate		
Budu-Aggrey et al, 2019	182	BMI vs Psoriasis	97	339,224	12,559	0%	OR	1.06 (1 to 1.12)	No	-
Carreras-Torres et al, 2017	200	Height vs Pancreatic Cancer	558	253,288	15,002	19%	OR	1.14 (1 to 1.29)	No	0.05
Carter et al, 2019	199	Education vs Coronary Disease	1,267	766,345	184,305	0%	OR	0.62 (0.57 to 0.67)	Yes	<0.001
Choi et al, 2019	492	Activity vs Depression	24	377,234	143,265	0%	OR	1.49 (0.94 to 2.36)	No	0.08
Clift et al, 2022	129	Smoking Initiation vs COVID-19 Infection	378	1,232,091	281,105	36%	OR	1.53 (1.02 to 2.28)	Yes	0.04
Ligthart et al, 2018	298	CRP vs Schizophrenia	52	204,402	82,315	0%	OR	0.89 (0.81 to 0.96)	Yes	0.004
Mokry et al, 2016	199	BMI vs Multiple Sclerosis	70	322,105	38,589	2.5%	OR	1.26 (0.98 to 1.62)	No	0.08
Pasman et al, 2018	328	Schizophrenia vs Cannabis Use	102	150,064	184,765	0%	Beta	0.163 (0.067 to 0.259)	Yes	0.001
Xie et al, 2023	138	T2DM vs NAFLD	449	441,016	218,792	0%	OR	1.61 (1.09 to 2.38)	Yes	<0.001
Xu et al, 2022	183	Coeliac vs Gut Bifidobacterium	105	15,283	24,269	63%	OR	0.998 (0.99 to 1.005)	No	0.56

A summary of the re-analysis results is presented in Table 4; estimates are presented both as β regression coefficients and \acr{OR]s to aid comparison across studies.

A number of WME estimates generated through re-analysis matched the originally reported estimates poorly: Carrera-Torres et al [carreras-torres_role_2017], Ligthart et al[3], Mokry et al[5] and Xie et al[9]. Details of instruments used

Table 4: Re-analysis of ten highly-cited two-sample Mendelian randomisation articles reporting a weighted median estimate of casual effect, comparing results of both WME and MR-Hevo causal effect estimation methods

Author	Exposure	Outcome	SNPs	Weighted Median				MR Hevo			
				β	SE	OR	Causality Reported	β	SE	OR	Causality Reported
Budu-Aggrey et al	BMI	Psoriasis	97	0 (0-0)	0.137	1 (1-1)	No	0.08 (-0.16-0.34)	0.002	1.09 (0.86-1.4)	No
Carreras-Torres et al	Height	Pancreatic Cancer	558	0 (-1.78-1.78)	0.065	1 (0.17-5.93)	No	-0.5 (-1.48-1.69)	0.908	0.61 (0.23-5.41)	No
Carter et al	Years of Education	Coronary Artery Disease	1,266	-0.46 (-0.46–0.46)	0.042	0.63 (0.63-0.63)	Yes	-0.48 (-0.54–0.42)	0.000	0.62 (0.58-0.66)	Yes
Choi et al	Self-Reported Physical Activity	Major Depressive Disorder	24	0.4 (0.4-0.41)	0.240	1.5 (1.49-1.51)	Yes	0.28 (-0.11-0.67)	0.003	1.32 (0.89-1.96)	No
Clift et al	Genetically Determined Smoking Initiation	COVID-19 Infection	378	0.43 (0.42-0.43)	0.204	1.53 (1.53-1.54)	Yes	0.37 (0.1-0.64)	0.001	1.45 (1.1-1.91)	Yes
Ligthart et al	Genetically Determined CRP	Schizophrenia	52	-0.49 (-1.26-0.28)	0.257	0.62 (0.28-1.33)	No	0.8 (0.21-1.63)	0.393	2.22 (1.23-5.12)	Yes
Mokry et al	BMI	Multiple Sclerosis	70	0.32 (0.32-0.32)	0.132	1.38 (1.37-1.38)	Yes	0.31 (0.14-0.48)	0.001	1.37 (1.16-1.61)	Yes
Pasman et al	Liability to Schizophrenia	Cannabis Use	109	0.16 (0.16-0.16)	0.048	1.18 (1.17-1.18)	Yes	0.16 (0.07-0.26)	0.001	1.18 (1.08-1.29)	Yes
Xie et al	T2DM	NAFLD	449	0.2 (0.2-0.2)	0.216	1.22 (1.22-1.23)	Yes	0.18 (-0.08-0.44)	0.002	1.2 (0.92-1.56)	No
Xu et al	Coeliac Disease	Gut Bifidobacterium	105	0 (0-0)	0.003	1 (1-1)	Yes	0 (0-0.01)	0.000	1 (1-1.01)	No

β and OR presented as: estimate (95% CI).

β: causal effect estimate, CI: Confidence Interval, OR: Odds Ratio, SE: Standard Error.

Table 4 reference[2,11]

CI confidence interval
CLT central limit theorem
IV instrumental variable
InSIDE Instrument Strength Independent of Direct Effect
MR Mendelian randomisation
RCT randomised-controlled trial
SD standard deviation
SE standard error
SNP single nucleotide polymorphism
UMREG Usher Masters Research Ethics Group
WME weighted median estimator

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