

Genetics of Smoking and Risk of Spinal Degenerative Diseases: A Two-Sample Mendelian Randomization Study

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Table S1. Detailed information for instrumental variables of smoking status

Chr	Position	Beta	SE	<i>P</i>	Sample size	SNP	Effect_al lele	Other_al lele	EAF	Exposur e	F- statistic
1	4403768 5	0.033512	0.003898	8.12E-18	632802	rs300172 3	A	G	0.321	smoking initiation id:ieu- b-4877	74
1	7376603 7	-0.02414	0.003556	1.14E-11	632802	rs755550 7	T	C	0.496	smoking initiation id:ieu- b-4877	46
1	5062597 9	0.026004	0.004396	3.36E-09	632802	rs666983 9	T	C	0.204	smoking initiation id:ieu- b-4877	35
1	9119617 6	-0.02228	0.003568	4.22E-10	632802	rs120421 07	C	T	0.527	smoking initiation id:ieu- b-4877	39
1	6647020 6	0.026057	0.003586	3.61E-13	632802	rs218612 2	T	A	0.561	smoking initiation id:ieu- b-4877	53

1	8481016	0.021468	0.003613	2.8E-09	632802	rs301805	G	T	0.559	smoking initiation id:ieu- b-4877	35
1	1.54E+08	-0.033	0.005339	6.52E-10	632802	rs120252 37	C	A	0.124	smoking initiation id:ieu- b-4877	38
1	8790582 8	-0.02055	0.003708	3E-08	632802	rs205058 6	C	G	0.355	smoking initiation id:ieu- b-4877	31
1	2.1E+08	-0.02481	0.004478	3.03E-08	632802	rs204685 0	T	C	0.187	smoking initiation id:ieu- b-4877	31
2	623976	0.035449	0.004733	6.73E-14	632802	rs672872 6	C	T	0.829	smoking initiation id:ieu- b-4877	56
2	5817122 0	0.020536	0.003659	2.03E-08	632802	rs784111 60	C	A	0.631	smoking initiation id:ieu- b-4877	31
2	1.82E+08	0.022448	0.004058	3.16E-08	632802	rs643389 7	C	T	0.754	smoking initiation	31

2	1.04E+08	-0.03051	0.003739	3.36E-16	632802	rs266047	A	G	0.529	id:ieu- b-4877 smoking initiation	67
2	2.26E+08	-0.02521	0.004436	1.32E-08	632802	rs467499 3	G	A	0.207	id:ieu- b-4877 smoking initiation	32
2	4514317 5	0.02868	0.003596	1.5E-15	632802	rs578584	T	A	0.605	id:ieu- b-4877 smoking initiation	64
2	1.38E+08	0.025244	0.004231	2.43E-09	632802	rs357025 15	T	G	0.162	id:ieu- b-4877 smoking initiation	36
2	1.46E+08	0.036093	0.003556	3.56E-24	632802	rs130309 94	A	G	0.485	id:ieu- b-4877 smoking initiation	103
2	1.63E+08	0.027633	0.003582	1.25E-14	632802	rs124745 87	T	G	0.404	id:ieu- b-4877 smoking initiation	60
										id:ieu- b-4877	

2	2.01E+08	-0.0272	0.004925	3.27E-08	632802	rs210730 0	G	C	0.845	smoking initiation id:ieu- b-4877	30
2	6002485 7	0.0224	0.003728	1.88E-09	632802	rs758557 9	G	C	0.505	smoking initiation id:ieu- b-4877	36
2	1.56E+08	0.023993	0.003565	1.68E-11	632802	rs144564 9	C	T	0.525	smoking initiation id:ieu- b-4877	45
3	8562413 1	-0.03135	0.003689	1.91E-17	632802	rs678809 8	T	A	0.623	smoking initiation id:ieu- b-4877	72
3	5022422 5	-0.02338	0.003753	4.78E-10	632802	rs126321 10	G	A	0.647	smoking initiation id:ieu- b-4877	39
3	7500901 9	-0.02705	0.004578	3.51E-09	632802	rs117126 80	C	A	0.174	smoking initiation id:ieu- b-4877	35
3	1.18E+08	0.032622	0.004912	3.12E-11	632802	rs115469 3	G	A	0.856	smoking initiation	44

3	8598532 4	-0.02027	0.003653	2.83E-08	632802	rs666808 00	T	G	0.397	id:ieu- b-4877 smoking initiation	31
3	5724536	0.019741	0.003563	2.97E-08	632802	rs186924 3	C	T	0.481	id:ieu- b-4877 smoking initiation	31
3	8576602 5	0.024047	0.004142	6.32E-09	632802	rs983577 2	T	A	0.235	id:ieu- b-4877 smoking initiation	34
4	2847352 4	0.023718	0.004038	4.37E-09	632802	rs962625	G	A	0.24	id:ieu- b-4877 smoking initiation	35
4	6782589 4	-0.02593	0.004292	1.53E-09	632802	rs993700	C	T	0.766	id:ieu- b-4877 smoking initiation	37
4	1.41E+08	-0.02325	0.003663	2.14E-10	632802	rs131457 28	C	G	0.358	id:ieu- b-4877 smoking initiation	40

4	1.48E+08	-0.02499	0.003642	6.65E-12	632802	rs100013 65	A	G	0.405	smoking initiation id:ieu- b-4877	47
4	9405285 4	0.020772	0.003589	7.2E-09	632802	rs116068 5	G	C	0.478	smoking initiation id:ieu- b-4877	33
5	6037491 2	-0.0241	0.004074	3.25E-09	632802	rs689375 2	G	A	0.766	smoking initiation id:ieu- b-4877	35
5	1.04E+08	-0.03326	0.005021	3.42E-11	632802	rs121867 38	T	G	0.154	smoking initiation id:ieu- b-4877	44
5	1.55E+08	0.024662	0.004157	3E-09	632802	rs138510 8	T	C	0.239	smoking initiation id:ieu- b-4877	35
5	1.67E+08	-0.02784	0.003711	6.08E-14	632802	rs404432 1	G	A	0.642	smoking initiation id:ieu- b-4877	56
5	8775682 1	-0.02753	0.003569	1.22E-14	632802	rs435262 9	T	C	0.492	smoking initiation	59

5	1.07E+08	-0.03289	0.005286	5.02E-10	632802	rs727896 32	T	C	0.12	id:ieu- b-4877 smoking initiation	39
6	9874800 8	0.027731	0.003986	3.47E-12	632802	rs940177 0	A	G	0.273	id:ieu- b-4877 smoking initiation	48
6	5291606 2	-0.02532	0.004428	1.08E-08	632802	rs222449	T	A	0.793	id:ieu- b-4877 smoking initiation	33
6	1.09E+08	0.022812	0.004058	1.93E-08	632802	rs380022 7	G	A	0.701	id:ieu- b-4877 smoking initiation	32
6	6740533 7	0.02061	0.003556	6.62E-09	632802	rs104988 46	T	C	0.473	id:ieu- b-4877 smoking initiation	34
6	1.12E+08	-0.04104	0.004837	2.16E-17	632802	rs240963	C	T	0.836	id:ieu- b-4877 smoking initiation	72

7	9918540 6	-0.02905	0.004801	1.44E-09	632802	rs123337 60	C	T	0.204	smoking initiation id:ieu- b-4877	37
7	1.18E+08	0.027069	0.003557	2.75E-14	632802	rs102330 18	G	A	0.503	smoking initiation id:ieu- b-4877	58
7	1.34E+08	-0.02142	0.003663	5E-09	632802	rs102792 61	A	G	0.619	smoking initiation id:ieu- b-4877	34
7	1889773	-0.02032	0.003609	1.75E-08	632802	rs102609 68	A	G	0.597	smoking initiation id:ieu- b-4877	32
7	6973525 1	-0.02453	0.004043	1.34E-09	632802	rs121126 38	G	A	0.275	smoking initiation id:ieu- b-4877	37
7	1708080	-0.02477	0.003557	3.35E-12	632802	rs423625 9	G	T	0.499	smoking initiation id:ieu- b-4877	48
7	3407568	-0.02326	0.003734	4.7E-10	632802	rs214011 4	T	C	0.518	smoking initiation	39

7	9663826 7	-0.02206	0.00374	3.74E-09	632802	rs380128 9	C	A	0.351	id:ieu- b-4877 smoking initiation id:ieu- b-4877	35
8	2742607 7	-0.03762	0.004461	3.42E-17	632802	rs156573 5	A	T	0.212	smoking initiation id:ieu- b-4877	71
8	9320103 6	0.026448	0.003887	1.04E-11	632802	rs189989 6	T	C	0.286	smoking initiation id:ieu- b-4877	46
8	5981466 6	-0.02689	0.003556	3.9E-14	632802	rs132616 66	T	G	0.522	smoking initiation id:ieu- b-4877	57
8	6507360 5	0.020281	0.003637	2.43E-08	632802	rs125450 53	G	A	0.397	smoking initiation id:ieu- b-4877	31
8	9199557 7	-0.02296	0.004028	1.18E-08	632802	rs263102 4	G	A	0.737	smoking initiation id:ieu- b-4877	33

9	3014254	0.021931	0.003562	7.46E-10	632802	rs454359 2	C	T	0.468	smoking initiation id:ieu- b-4877	38
9	8670728 9	0.020948	0.003566	4.16E-09	632802	rs237866 2	A	G	0.556	smoking initiation id:ieu- b-4877	35
9	1107016 5	-0.02551	0.004532	1.81E-08	632802	rs101144 90	A	G	0.198	smoking initiation id:ieu- b-4877	32
10	8803551	-0.02396	0.004145	7.35E-09	632802	rs109054 61	C	T	0.718	smoking initiation id:ieu- b-4877	33
10	6367488 5	-0.02546	0.003558	8.26E-13	632802	rs792137 8	C	G	0.463	smoking initiation id:ieu- b-4877	51
10	1.05E+08	0.03937	0.005049	6.27E-15	632802	rs123568 21	C	G	0.14	smoking initiation id:ieu- b-4877	61
10	2176696 9	0.02625	0.003727	1.84E-12	632802	rs101595 45	G	C	0.375	smoking initiation	50

10	1.26E+08	-0.02051	0.003708	3.21E-08	632802	rs942327 9	G	C	0.641	id:ieu- b-4877 smoking initiation	31
11	1.13E+08	0.043791	0.003637	2.71E-33	632802	rs793881 2	G	T	0.424	id:ieu- b-4877 smoking initiation	145
11	2767991 6	-0.03179	0.004578	3.77E-12	632802	rs6265	T	C	0.203	id:ieu- b-4877 smoking initiation	48
11	8598095 8	0.024238	0.004285	1.56E-08	632802	rs792951 8	G	A	0.765	id:ieu- b-4877 smoking initiation	32
11	7950797	-0.02061	0.003643	1.55E-08	632802	rs452368 9	G	A	0.408	id:ieu- b-4877 smoking initiation	32
12	1674872 1	-0.02093	0.003579	4.85E-09	632802	rs110570 05	G	A	0.43	id:ieu- b-4877 smoking initiation	34
										id:ieu- b-4877	

12	5650840 9	-0.02169	0.003934	3.58E-08	632802	rs475922 8	C	G	0.27	smoking initiation id:ieu- b-4877	30
12	6965516 7	-0.02438	0.003959	7.31E-10	632802	rs796955 9	G	A	0.688	smoking initiation id:ieu- b-4877	38
12	1.21E+08	0.028507	0.004925	7.06E-09	632802	rs197131 8	T	C	0.141	smoking initiation id:ieu- b-4877	33
13	1.01E+08	-0.02557	0.004335	3.58E-09	632802	rs732287 2	T	C	0.782	smoking initiation id:ieu- b-4877	35
13	3835747 1	-0.02116	0.003577	3.23E-09	632802	rs390451 2	A	G	0.429	smoking initiation id:ieu- b-4877	35
13	6694712 4	-0.01955	0.003558	3.82E-08	632802	rs954072 9	T	A	0.501	smoking initiation id:ieu- b-4877	30
14	2950013 0	-0.02499	0.004547	3.99E-08	632802	rs762148 62	C	A	0.202	smoking initiation	30

15	8392238 7	-0.02921	0.004523	1.06E-10	632802	rs124419 07	A	C	0.186	id:ieu- b-4877 smoking initiation	42
15	4793584 3	0.029415	0.003591	2.64E-16	632802	rs143574 1	A	G	0.425	id:ieu- b-4877 smoking initiation	67
16	6560465 2	-0.02047	0.003659	2.26E-08	632802	rs478583 6	C	T	0.398	id:ieu- b-4877 smoking initiation	31
16	717085	-0.02477	0.004169	2.77E-09	632802	rs719707 2	T	C	0.238	id:ieu- b-4877 smoking initiation	35
16	8744373 4	-0.02162	0.003589	1.67E-09	632802	rs105084 7	T	C	0.505	id:ieu- b-4877 smoking initiation	36
16	1757267 4	-0.02387	0.004365	4.54E-08	632802	rs478197 7	C	T	0.205	id:ieu- b-4877 smoking initiation	30
										id:ieu- b-4877	

17	7795972	-0.02017	0.003606	2.23E-08	632802	rs110787 13	G	A	0.454	smoking initiation id:ieu- b-4877	31
17	3065705 8	-0.02071	0.003655	1.43E-08	632802	rs722474 2	T	C	0.595	smoking initiation id:ieu- b-4877	32
17	2072949	0.020136	0.003611	2.43E-08	632802	rs116588 81	G	A	0.418	smoking initiation id:ieu- b-4877	31
18	5002614 2	-0.02069	0.003586	7.97E-09	632802	rs650814 4	G	C	0.563	smoking initiation id:ieu- b-4877	33
18	7253528 2	-0.02477	0.004095	1.43E-09	632802	rs118723 97	A	G	0.252	smoking initiation id:ieu- b-4877	37
18	4263265 2	-0.02689	0.004837	2.75E-08	632802	rs728968 86	C	G	0.144	smoking initiation id:ieu- b-4877	31
19	4474725	-0.04956	0.00826	1.94E-09	632802	rs766085 82	A	C	0.0389	smoking initiation	36

20	31175258	0.022555	0.003823	3.65E-09	632802	rs1555445	T	A	0.337	id:ieu-b-4877 smoking initiation	35
21	40555561	0.02929	0.005269	2.76E-08	632802	rs117143374	C	T	0.12	id:ieu-b-4877 smoking initiation	31
22	28781758	-0.01998	0.003661	4.85E-08	632802	rs134529	C	T	0.349	id:ieu-b-4877 smoking initiation	30
										id:ieu-b-4877	

Chr, chromosome; EAF, allele frequency of effect allele; SE, standard error of beta; Sample size, available sample size for each variant in according studies; F-statistic calculated as: $F\text{-statistic} = R^2 * (\text{SampleSize} - 2) / (1 - R^2)$.

Table S2. MR analyses of the effect of smoking initiation on cigarettes per day and current tobacco smoking

Outcome	Method	N.SNPs	OR (95% CI)	P value
Cigarettes per Day	MR Egger	85	1.04 (0.66, 1.65)	0.871825
	Weighted median	85	1.21 (1.11, 1.31)	8.26E-06
	Inverse variance weighted	85	1.20 (1.09, 1.31)	0.000101
	Simple mode	85	1.33 (1.06, 1.67)	0.015903
	Weighted mode	85	1.29 (1.03, 1.62)	0.031194

Current tobacco smoking	MR Egger	84	1.19 (1.11, 1.28)	6.94E-06
	Weighted median	84	1.18 (1.16, 1.21)	6.17E-76
	Inverse variance weighted	84	1.19 (1.17, 1.20)	9.69E-125
	Simple mode	84	1.23 (1.17, 1.29)	1.41E-11
	Weighted mode	84	1.22 (1.16, 1.28)	9.22E-12

N.SNPs: number of SNPs used in MR; OR, odds ratio.

Table S3. MR analyses of the effect of smoking initiation on body mass index, type 2 diabetes, overweight and waist circumference

Outcome	Method	N.SNPs	OR (95% CI)	P value
Body mass index	MR Egger	84	1.39 (0.96, 2.03)	0.088438
	Weighted median	84	1.12 (1.07, 1.17)	5.61E-07
	Inverse variance weighted	84	1.20 (1.12, 1.29)	1.24E-06
	Simple mode	84	1.25 (1.13, 1.38)	4.88E-05
	Weighted mode	84	1.24 (1.05, 1.46)	0.013712
Type 2 diabetes	MR Egger	76	1.18 (0.64, 2.17)	0.600831
	Weighted median	76	1.27 (1.12, 1.44)	0.000149
	Inverse variance weighted	76	1.29 (1.15, 1.46)	2.74E-05
	Simple mode	76	1.37 (0.98, 1.91)	0.068801
	Weighted mode	76	1.30 (0.99, 1.71)	0.0637
Overweight	MR Egger	74	1.27 (0.55, 2.89)	0.578315
	Weighted median	74	1.08 (0.94, 1.25)	0.294699
	Inverse variance weighted	74	1.19 (1.01, 1.40)	0.038241
	Simple mode	74	1.09 (0.77, 1.54)	0.635137
	Weighted mode	74	1.09 (0.79, 1.50)	0.606926
Waist circumference	MR Egger	84	1.38 (1.04, 1.84)	0.028129

Weighted median	84	1.10 (1.06, 1.14)	1.16E-06
Inverse variance weighted	84	1.17 (1.10, 1.24)	5.41E-08
Simple mode	84	1.19 (1.04, 1.37)	0.014691
Weighted mode	84	1.01 (0.92, 1.10)	0.894964

N.SNPs: number of SNPs used in MR; OR, odds ratio.

Table S4. Cochran's Q test for heterogeneity among MR analyses*

Exposure	Outcome	Method	Q	Q_df	Q_pval
Smoking initiation	Intervertebral disc degeneration	MR Egger	125.6844	82	0.001377
		Inverse variance weighted	126.1118	83	0.001607
	Sciatica	MR Egger	112.2543	82	0.014908
		Inverse variance weighted	114.3621	83	0.012819
	Low back pain	MR Egger	124.7383	82	0.001652
		Inverse variance weighted	124.9137	83	0.002016

*The Q statistic has a chi-squared distribution on N-1 degrees of freedom (df) under the null hypothesis that all genetic variants are valid IVs and the same causal effect is identified by all variants. N is the number of genetic variants.

Table S5. MR Egger test of directional pleiotropy

Exposure	Outcome	Egger_intercept	SE	P value
Smoking initiation	Intervertebral disc degeneration	-0.00469	0.00889	0.598873
	Sciatica	0.014468	0.01166	0.218193
	Low back pain	-0.0035	0.010307	0.735095

Table S6. Causal relationships of smoking on outcome estimated by multivariable MR*

Exposure	Outcome	N.SNP	Beta	SE	P value
Smoking initiation	Low back pain	25	0.283808	0.0953	0.002901
Type 2 diabetes		55	0.021676	0.02698	0.421742
Overweight		8	-0.04123	0.07969	0.604919
Body mass index		247	-0.08155	0.249103	0.743398
Waist circumference		198	0.465507	0.287662	0.10561
Smoking initiation	Intervertebral disc degeneration	25	0.211893	0.086747	0.01458
Type 2 diabetes		55	0.052602	0.024579	0.032346
Overweight		8	-0.01419	0.072747	0.845332
Body mass index		247	0.159043	0.226555	0.482675
Waist circumference		198	0.068603	0.261764	0.79326

Smoking initiation	Sciatica	25	0.055412	0.105866	0.600685
Type 2 diabetes		55	0.049323	0.029958	0.099678
Overweight		8	-0.08117	0.088616	0.359657
Body mass index		247	0.269499	0.276649	0.329979
Waist circumference		198	0.102085	0.319355	0.749226

*Multivariable Mendelian randomization using inverse variance weighted approach; N.SNPs, number of SNPs used in MR.

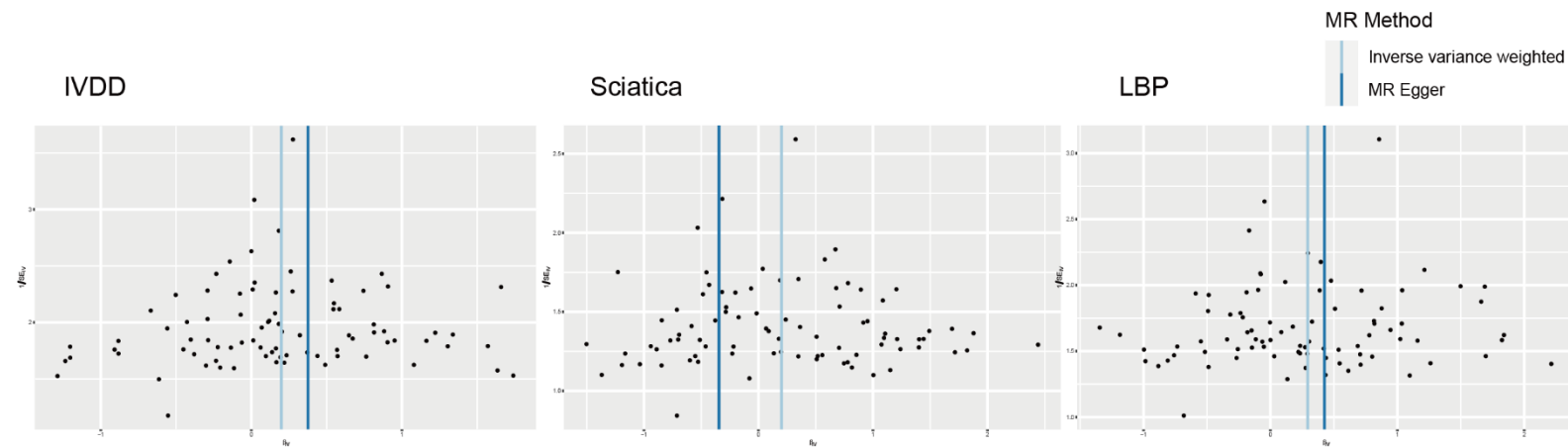


Figure S1. Funnel plots to show symmetrical distribution of individual variant estimates around the point estimate. The x-axis represents the MR estimate of individual variants. The y-axis represents the inverse of their standard error. The vertical lines indicate the MR estimates using the five different MR methods. The asymmetry in the funnel plot may be an indicator of heterogeneous effects and may be related to directional level

pleiotropy that leads to biased effect estimates.

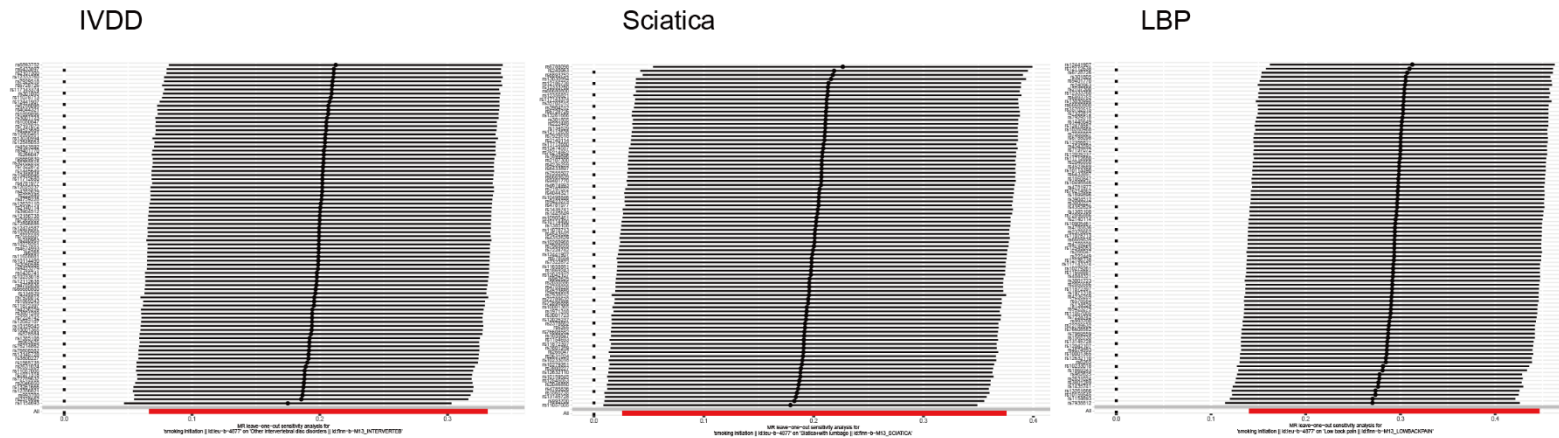


Figure S2. MR Leave-One-Out Analysis In leave-one-out analysis, each individual SNP is sequentially excluded from the inverse variance weighted MR model to identify outlier SNPs that may drive outside effects. Plotted are the log odds ratios of each leave-one-out model for each outcome (IVDD, sciatica, and LBP) against the SNP excluded from each model. Lines extending from each point represent the 95% confidence intervals of the effect estimate.

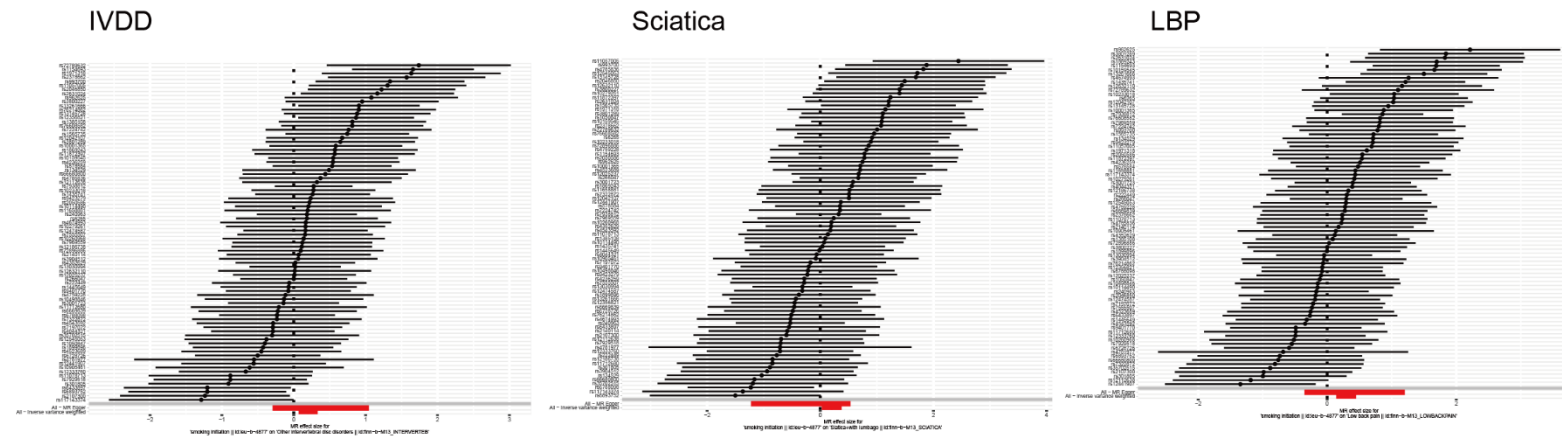


Figure S3. MR Single-SNP Analysis The single-SNP MR method plots the Wald estimate of the effect of each SNP included in the smoking genetic instrument on each outcome. Lines extending from each point represent the 95% confidence interval of the odds ratio (points). Red estimates at the bottom of each plot represent the summary measures from five MR models including all of the individual SNPs.