

Smoking and heart failure: a Mendelian randomization and mediation analysis

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Abstract

Aims We performed a Mendelian randomization (MR) study to elucidate the associations of ever smoking, lifelong smoking duration, and smoking cessation with heart failure (HF) risk.

Methods and results We extracted genetic variants associated with smoking initiation, age at initiation of regular smoking, cigarettes per day, and smoking cessation from the genome-wide association study and Sequencing Consortium of Alcohol and Nicotine use (1.2 million individuals), as well as a composite lifetime smoking index from the UK Biobank (462 690 individuals). The associations between smoking phenotypes and HF were explored in the Heart Failure Molecular Epidemiology for Therapeutic Targets Consortium (47 309 cases; 930 014 controls) employing inverse variance-weighted meta-analysis and multivariable MR. The mediation effects of coronary artery disease and atrial fibrillation on smoking–HF risk were explored using mediation analysis. The odds ratios (ORs) for HF were 1.28 [95% confidence interval (CI), 1.22–1.36; $P = 1.5 \times 10^{-18}$] for ever regular smokers compared with never smokers and 1.25 (95% CI, 1.09–1.44; $P = 1.6 \times 10^{-3}$) for current smokers vs. former smokers. Genetic liability to smoking more cigarettes per day (OR, 1.37; 95% CI, 1.20–1.58; $P = 6.4 \times 10^{-6}$) and a higher composite lifetime smoking index (OR, 1.49; 95% CI, 1.31–1.70; $P = 2.5 \times 10^{-9}$) were associated with a higher risk of HF. The results were robust and consistent in all sensitivity analyses and multivariable MR after adjusting for HF risk factors, and their associations were independent of coronary artery disease and atrial fibrillation.

Conclusions Genetic liability to ever smoking and a higher lifetime smoking burden are associated with a higher risk of HF.

Keywords Heart failure; Mendelian randomization; Mediation analysis; Smoking

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Introduction

Heart failure (HF) comprises a rising burden for public health. It is estimated that 37.7 million patients worldwide suffer from HF, which results in 4.2 million years lived with the disability.^{1,2} In 2011, the 30 day mortality after hospitalization was 16.3% for HF patients.³

Cigarette smoking has been associated with a higher risk of HF throughout observational studies.^{4–6} However, whether the total smoking burden (as determined by smoking duration and cigarettes per day) and smoking cessation (current vs. former smoking) have causal effects on HF risk requires further exploration.^{7,8} In addition, observational studies are susceptible to bias due to confounding and reverse causation

and possibly leading to unreliable causal effects. While most studies acknowledge the harmful effects of smoking on HF, the current Global Burden of Disease Study has not addressed HF when evaluating the contribution of smoking to cardiovascular diseases⁹; this may be due to the complex effects of smoking on HF. Evidence from clinical studies suggests that smoking is associated with obesity, diabetes, hyperlipaemia, hypertension, coronary artery disease (CAD), and atrial fibrillation (AF),¹⁰ which are also established risk factors and mediators for HF.^{11,12}

Mendelian randomization (MR) is a genetic epidemiological method to explore the causal effect of an exposure on disease outcome, using genetic variants as instrumental variables (IVs).¹³ Because of its features of random assortment

and remaining constant in germline genotypes, MR can diminish bias arising due to confounding and reverse causation, which otherwise limits observational epidemiology. Hence, we conducted an MR analysis to evaluate the association between different smoking phenotypes (smoking initiation, age at initiation of regular smoking, smoking cessation, and lifetime smoking index) and HF and examined whether IVs directly affect HF through smoking rather than other risk factors.

Methods

Study design

A directed acyclic graph model was used to manage the potential risk factors that could confound or mediate the causal effect between smoking and HF (Figure 1). We constructed IVs composed of single-nucleotide polymorphisms (SNPs) directly associated with different smoking phenotypes and combined the estimates of smoking-associated SNPs with HF. These estimates of causal effect were adjusted for potential HF risk factors using multivariable MR to eliminate pleiotropic interference.¹⁴ We also evaluated the mediation effects of CAD and AF on smoking–HF risk using mediation analysis. For SNPs not available in the HF database, proxy SNPs were used by searching through European population genotype data originating from Phase 3 (Version 5) of the 1000 Genomes Project [linkage disequilibrium (LD) $r^2 > 0.8$; identified using online tool SNIpa, available at: <http://snipa.helmholtz-muenchen.de/snipa3/>].

Data sources

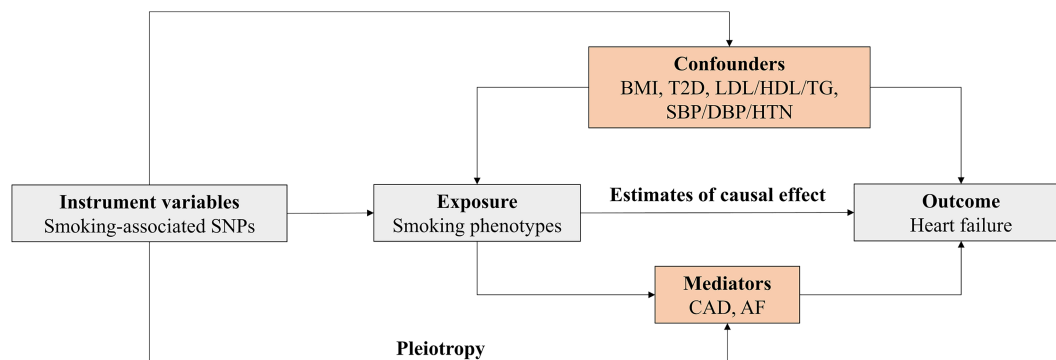
Publicly available summary statistics were used to conduct the MR analyses (Supporting Information, Table S1). The

approval procedures of the ethics committee are available in the original studies included in the genome-wide association study (GWAS). All participants of the original studies provided written informed consent.

The data on tobacco use were extracted from the GWAS and Sequencing Consortium of Alcohol and Nicotine use (GSCAN) for four smoking phenotypes, including smoking initiation, age at initiation of regular smoking, number of cigarettes per day, and smoking cessation. Genetic IVs for the exposure were selected at the genome-wide significance level ($P < 5 \times 10^{-8}$) across a 1 Mb region.¹⁵

Smoking initiation was a binary phenotype, defined as ever being a regular smoker in life (current or former). The GWAS of smoking initiation identified 378 significant SNPs, explaining 2.3% of the heritability in up to 1 232 091 individuals. While nine SNPs were unavailable in the HF database, eight proxy SNPs in LD ($r^2 \geq 0.8$) with the specified SNPs were used; thus, 377 of 378 SNPs satisfied our inclusion criteria (Supporting Information, Table S2). The GWAS additionally identified 10 SNPs significantly associated with age at initiation of regular smoking, explaining 0.2% of the heritability in up to 34 427 individuals; therefore, 10 SNPs were selected as IVs (Supporting Information, Table S3). The 55 SNPs related to number of cigarettes per day (in both current and former smokers) in 337 334 individuals were estimated to account for 1.1% of the variation in this phenotype. There was one SNP not found in the HF database, which could not be matched with an appropriate proxy SNP; therefore, 54 SNPs associated with cigarettes per day were used as IVs in the MR analyses (Supporting Information, Table S4). All 24 SNPs associated with smoking cessation (binary phenotype, defined as current smoker vs. former smoker), which explained 0.1% of the variation in smoking cessation, were identified as IVs in 547 219 individuals (Supporting Information, Table S5). Further details regarding these smoking phenotypes can be found in Supporting Information, Methods.

Figure 1 Directed acyclic graph model of the causal effect between smoking and heart failure. AF, atrial fibrillation; BMI, body mass index; CAD, coronary artery disease; DBP, diastolic blood pressure; HDL, high-density lipoprotein; HTN, hypertension; LDL, low-density lipoprotein; SBP, systolic blood pressure; T2D, type 2 diabetes mellitus; SNPs, single-nucleotide polymorphisms; TG, triglyceride.



In addition, we performed a sensitivity analysis of the association between lifetime smoking index and HF. Lifetime smoking index is a continuous composite measure of the burden of lifetime exposure to smoking constructed by taking into account smoking status, age at smoking initiation and cessation, cigarettes smoked per day, and a simulated half-life constant effected on health outcomes (lung cancer and overall mortality).¹⁶ The GWAS of lifetime smoking index identified 126 independent SNPs at the genome-wide significance level ($P < 5 \times 10^{-8}$) in an LD $r^2 < 0.001$ across a 10 Mb window in up to 462 690 individuals of the UK Biobank (Supporting Information, *Table S6*).¹⁶ All 126 SNPs were available in the HF database. A 1-SD increase in the lifetime smoking index was scaled to an individual smoking 20 cigarettes a day for 15 years and quitting 17 years ago, or smoking 60 cigarettes a day for 13 years and quitting 22 years ago.

A previous study evaluated the correlations of lifetime smoking index with smoking initiation, age at smoking initiation, cigarettes per day, and smoking cessation in GSCAN Consortium. The results showed that lifetime smoking index was significantly associated with an increased probability of being a regular and current smoker, a higher number of daily cigarettes, and a younger age of regular smoking.¹⁷ There was only a small overlap (nine SNPs) between the SNPs related to lifetime smoking index and smoking initiation. Furthermore, the instruments of lifetime smoking index increased the risk of both lung cancer and CAD.¹⁶

Estimates of the strength of the association between the genetic instruments and smoking phenotypes (F statistics) were generated to explore the possibility of weak instrument bias.¹⁸ For the instrument SNPs selected in this MR study, F statistics ranged from 22 to 1310, above the recommended threshold of $F > 10$ in MR analysis.¹⁸

Summary statistics for the association between SNPs related to smoking and HF were derived from the Heart Failure Molecular Epidemiology for Therapeutic Targets (HERMES) Consortium, including 47 309 patients and 930 014 controls of European ancestry.¹¹ HF cases included participants with incident or prevalent HF, defined by self-reported, medical records, and the 9th/10th revision of the International Classification of Diseases codes (primarily 9th revision of the International Classification of Diseases: 428 and subcodes, and 10th revision of the International Classification of Diseases: I50 and subcodes).¹¹

Statistical analysis

The standard analysis was the random-effects inverse variance-weighted (IVW) meta-analysis. We also used alternative analyses, including the simple median, weighted median, MR robust adjusted profile score,¹⁹ and MR pleiotropy residual sum and outlier (MR-PRESSO).²⁰ The MR-PRESSO method was adopted to detect the outlier variants of the

IVW analysis by comparing the actual distance of genetic variants to the regression line with the expected distance if there was no horizontal pleiotropy, as well as to evaluate the causal estimates after the removal of outliers.²⁰ A unanimous MR result across all five methods was regarded as a reliable association estimate.^{21,22}

Quantitative heterogeneity among the SNPs in the IVW analysis, tested with the I^2 statistic, was considered a measure of heterogeneity (low, $>25\%$; moderate, $>50\%$; and high, $>75\%$). The MR-Egger method was performed as an additional sensitivity analysis to quantify potential directional pleiotropy from the intercept ($P < 0.05$ was considered significant); however, it suffered from low statistical power.^{23,24} Scatter plots of IVW MR analyses regarding the associations of genetically determined smoking with HF were also provided.

To explore the direct effect of smoking on HF, we conducted a multivariable MR analysis that was adjusted for general risk factors that were genetically correlated with HF.^{11,14} Multivariable IVW was used as the primary analysis. We also applied multivariable MR-Egger as a complementary method to orientate the instruments with respect to risk factors.²⁵ We used publicly available summarized data regarding the genetic association of instruments with body mass index from the Genetic Investigation of Anthropometric Traits (322 154 individuals),²⁶ type 2 diabetes mellitus from the Diabetes Genetics Replication and Meta-analysis (34 840 cases; 114 981 controls),²⁷ circulating lipid levels (low-density lipoprotein cholesterol, high-density lipoprotein cholesterol, and triglycerides) from the Global Lipids Genetics Consortium (188 578 individuals),²⁸ and blood pressure measurements (systolic and diastolic blood pressure and hypertension) from the UK Biobank (317 754 individuals), published by the Neale Lab,²⁹ respectively. A fully adjusted model including all risk factors was also performed. The conditional F statistic for each smoking phenotype was calculated to evaluate the joint strength of instruments in the multivariable MR analyses.³⁰

Observational studies and Mendelian analyses have suggested that cigarette smoking is a modifiable risk factor for CAD^{9,31} and AF,³² which played a mediating role in the pathogenesis of HF.¹¹ We therefore explored the mediating effect of CAD from the Coronary Artery Disease Genome-wide Replication and Meta-analysis (CARDIoGRAM) plus The Coronary Artery Disease (CAD) Genetics (60 801 cases; 123 504 controls),³³ and AF from the Atrial Fibrillation Haplotype Reference Consortium (65 446 cases; 522 744 controls),³⁴ on the causal pathway from smoking to HF via mediation analysis.³⁵

A two-sided P -value < 0.05 was considered suggestive for significance. We further adjusted the thresholds by Bonferroni correction for the number of exposure phenotypes. The statistical significance thresholds were therefore set at $P < 0.05/5 = 0.01$ for the five smoking exposures. All MR analyses were performed using the TwoSampleMR,

MendelianRandomization, MR-PRESSO, and MVMR R packages. Statistical analyses were conducted by R Version 3.6.1 (free software, see <https://www.r-project.org/foundation/>).

Results

Genetically determined smoking phenotypes and risk of heart failure

In the standard IVW method, genetic predisposition to smoking initiation was associated with a higher risk of HF, with an odds ratio (OR) of 1.28 [95% confidence interval (CI), 1.22–1.36; $P = 1.5 \times 10^{-18}$] per 2.72-fold (1 log-odds unit) increase in the probability of being a regular smoker ($I^2 = 30\%$). Genetically predicted age at initiation of regular smoking was not associated with HF (OR, 0.71; 95% CI, 0.48–1.06; $P = 0.10$) without detected outlier ($I^2 = 39\%$). The OR of HF per 1-SD increase (about eight cigarettes per day among current smokers in the UK Biobank) in genetically instrumented cigarettes per day was 1.37 (95% CI, 1.20–1.58; $P = 6.4 \times 10^{-6}$; $I^2 = 55\%$). Compared with former smokers, genetic predisposition to being a current smoker (smoking cessation) was associated with a 25% increased risk of HF (OR,

1.25; 95% CI, 1.09–1.44; $P = 1.6 \times 10^{-3}$; $I^2 = 36\%$) (Figure 2 and Supporting Information, Figure S1). No indication of directional pleiotropy was found by the MR-Egger intercept (all $P > 0.05$) (Supporting Information, Table S7). When using other complementary methods for analysis, the results showed consistent associations among smoking initiation, age at initiation of regular smoking, cigarettes per day, and smoking cessation with HF (Figure 2).

When using lifetime smoking index as exposure, the results showed a trend similar to that of the smoking initiation analysis (OR, 1.49; 95% CI, 1.31–1.70; $P = 2.5 \times 10^{-9}$; $I^2 = 37\%$) (Figure 3 and Supporting Information, Figure S1). The MR estimates of lifetime smoking index on HF were robust and consistent in all complementary analyses (OR ranged from 1.47 to 1.52; all $P < 0.01$) without evidence of directional pleiotropy, and no outlier SNPs were detected with the MR-PRESSO (Figure 2).

Risk factors as confounders of smoking–heart failure risk

To verify the direct causal effect of smoking on HF, we performed multivariable MR analyses, adjusting for HF risk factors (body mass index, type 2 diabetes mellitus, lipids, and

Figure 2 Mendelian randomization (MR) association of genetically predicted smoking initiation, age at initiation of regular smoking, cigarettes per day, and smoking cessation with heart failure. Odds ratios (ORs) are scaled to per genetically predicted per 2.72-fold (1 log-odds unit) increase in the genetic liability to be a regular smoker and current smoker and per 1-SD increase in the genetic liability of the age at initiation of regular smoking and cigarettes per day. *MR pleiotropy residual sum and outlier (MR-PRESSO) instrumental variable outlier detected: rs12244388. †No outlier detected. ‡MR-PRESSO instrumental variable outlier detected: rs10204824 and rs10742683. §MR-PRESSO instrumental variable outlier detected: rs1611124. CI, confidence interval; IVW, inverse variance weighted; MR-RAPS, MR robust adjusted profile score.

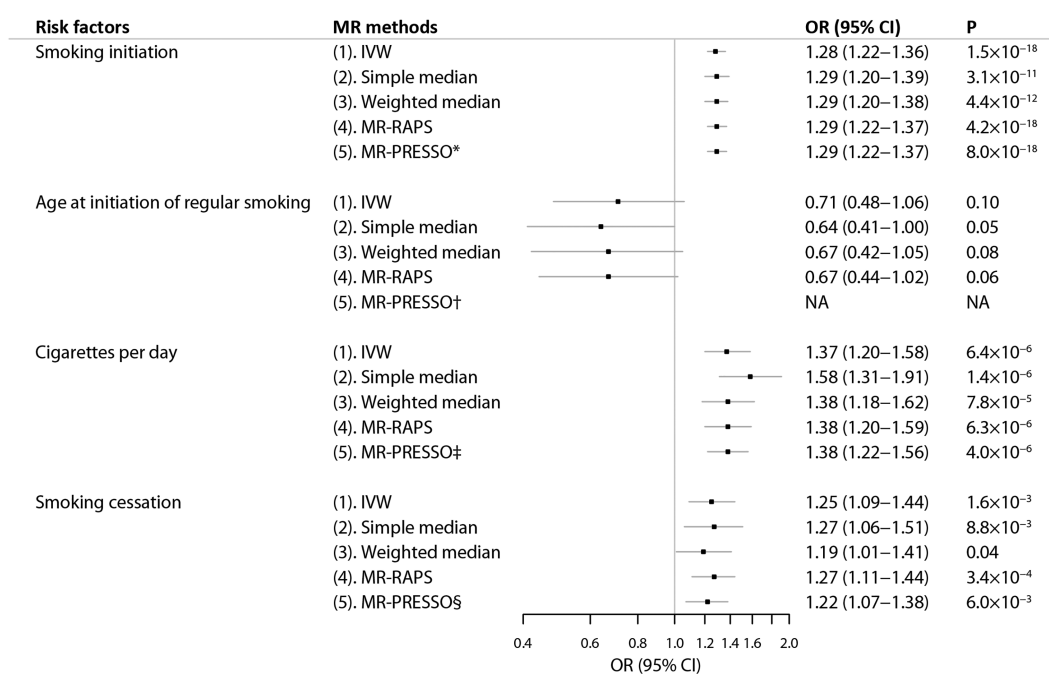
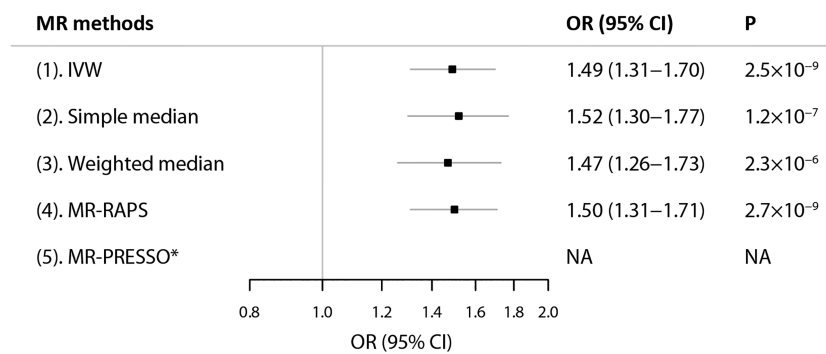


Figure 3 Mendelian randomization (MR) association between genetically predicted lifetime smoking index and heart failure. Odds ratios (ORs) are scaled to per genetically predicted per 1-SD increase in the lifetime smoking index. *No outlier detected. CI, confidence interval; IVW, inverse variance weighted; MR-PRESSO, MR pleiotropy residual sum and outlier; MR-RAPS, MR robust adjusted profile score.



blood pressure).¹¹ The associations among smoking initiation, age at initiation of regular smoking, current smoking, cigarettes per day, and lifetime smoking index with HF remained stable and robust after adjusting for single risk factors, as well as in a fully adjusted model considering all risk factors in multivariable IVW analysis (*Table 1*). Furthermore, multivariable MR-Egger results showed that smoking initiation was the only smoking phenotype that remained consistent and significant regardless of the model, thus highlighting the importance of never smoking (Supporting Information, *Table S7*). The conditional *F* statistics ranged from 20 to 109, which were higher than the recommended value of 10, meaning that instruments of each phenotype were adequately strong in the multivariable MR analyses.³⁰

Mediation effects of coronary artery disease and atrial fibrillation on smoking–heart failure risk

We conducted mediation analyses to investigate whether the effect of smoking on HF was mediated by CAD and AF. Analyses of all smoking phenotypes with HF showed no significant mediation effects acting through CAD or AF, suggesting an independent causal mechanism between different smoking phenotypes and risk of HF (*Table 2*).

Discussion

This MR study, which included 47 309 HF cases and 930 014 controls, showed that genetic predisposition to regular smoking (current or former, as compared with

Table 1 Multivariable Mendelian randomization associations of smoking with heart failure risk adjusting for its risk factors

| Model | Smoking initiation | Age at initiation of regular smoking | Cigarette per day | Current smoking | Lifetime smoking index |
|---|--------------------|--------------------------------------|-------------------|------------------|------------------------|
| No. of sample | 1 232 091 | 341 427 | 337 334 | 547 219 | 462 690 |
| No. of SNPs | 377 | 10 | 54 | 24 | 126 |
| Unadjusted model | 1.28 (1.22–1.36) | 0.71 (0.48–1.06) | 1.37 (1.20–1.58) | 1.25 (1.09–1.44) | 1.49 (1.31–1.70) |
| Adjusted for BMI | 1.21 (1.12–1.30) | 0.58 (0.36–0.94) | 1.44 (1.23–1.69) | 1.43 (1.11–1.84) | 1.58 (1.29–1.94) |
| Adjusted for T2D | 1.22 (1.13–1.32) | 0.62 (0.41–0.93) | 1.43 (1.22–1.67) | 1.59 (1.28–1.97) | 1.63 (1.34–1.99) |
| Adjusted for LDL-C | 1.28 (1.18–1.39) | 0.75 (0.45–1.26) | 1.51 (1.30–1.75) | 1.33 (1.07–1.67) | 1.71 (1.39–2.10) |
| Adjusted for HDL-C | 1.26 (1.17–1.37) | 0.66 (0.33–1.31) | 1.43 (1.19–1.73) | 1.41 (1.12–1.79) | 1.65 (1.35–2.03) |
| Adjusted for TG | 1.26 (1.16–1.37) | 0.60 (0.40–0.90) | 1.44 (1.21–1.71) | 1.39 (1.12–1.73) | 1.62 (1.32–1.99) |
| Adjusted for SBP | 1.28 (1.21–1.35) | 0.71 (0.46–1.09) | 1.35 (1.18–1.55) | 1.26 (1.08–1.47) | 1.48 (1.30–1.67) |
| Adjusted for DBP | 1.26 (1.19–1.33) | 0.72 (0.47–1.11) | 1.33 (1.16–1.51) | 1.27 (1.09–1.48) | 1.45 (1.27–1.66) |
| Adjusted for HTN | 1.21 (1.15–1.28) | 0.77 (0.50–1.18) | 1.25 (1.10–1.43) | 1.25 (1.09–1.43) | 1.35 (1.19–1.55) |
| Fully adjusted model (BMI, T2D, LDL-C ^a , SBP ^b) | 1.20 (1.11–1.29) | 0.59 (0.28–1.22) | 1.54 (1.30–1.82) | 1.87 (1.38–2.52) | 1.56 (1.27–1.93) |

BMI, body mass index; DBP, diastolic blood pressure; HDL-C, high-density lipoprotein cholesterol; HTN, hypertension; LDL-C, low-density lipoprotein cholesterol; SBP, systolic blood pressure; SNPs, single-nucleotide polymorphisms; T2D, type 2 diabetes mellitus; TG, triglyceride.

Results are shown as odds ratios (95% confidence intervals) for the association of smoking with heart failure from multivariable MR inverse variance-weighted method. Odds ratios are scaled to per genetically predicted per 2.72-fold (1 log-odds unit) increase in the genetic liability to be a regular smoker and current smoker and per 1-SD increase in the genetic liability of the age at initiation of regular smoking, cigarettes per day, and lifetime smoking index.

^aRestricted to LDL-C to avoid collinearity with HDL-C and TG levels.

^bRestricted to SBP to avoid collinearity with DBP and HTN.

Table 2 Mediation analysis of the mediation effect of smoking on heart failure via coronary artery disease or atrial fibrillation

| Mediator | Exposure | Total effect ^a Effect size (95% CI) | Direct effect ^b Effect size (95% CI) | Mediation effect ^c | |
|----------|--------------------------------------|---|--|-------------------------------|------|
| | | | | Effect size (95% CI) | P |
| CAD | Smoking initiation | 0.25 (0.19 to 0.31) | 0.21 (0.15 to 0.27) | 0.04 (−0.04 to 0.12) | 0.31 |
| | Age at initiation of regular smoking | −0.34 (−0.74 to 0.06) | −0.52 (−0.95 to −0.08) | 0.18 (−0.42 to 0.77) | 0.56 |
| | Cigarette per day | 0.32 (0.18 to 0.46) | 0.32 (0.18 to 0.46) | 0.00 (−0.20 to 0.19) | 0.97 |
| | Smoking cessation | 0.23 (0.09 to 0.37) | 0.05 (−0.12 to 0.22) | 0.18 (−0.04 to 0.39) | 0.11 |
| | Lifetime smoking index | 0.40 (0.27 to 0.53) | 0.27 (0.13 to 0.41) | 0.13 (−0.06 to 0.32) | 0.18 |
| AF | Smoking initiation | 0.25 (0.19 to 0.31) | 0.22 (0.17 to 0.28) | 0.03 (−0.05 to 0.11) | 0.49 |
| | Age at initiation of regular smoking | −0.34 (−0.74 to 0.06) | −0.46 (−0.83 to −0.09) | 0.12 (−0.42 to 0.67) | 0.66 |
| | Cigarette per day | 0.32 (0.18 to 0.46) | 0.32 (0.18 to 0.46) | 0.00 (−0.20 to 0.19) | 0.96 |
| | Smoking cessation | 0.23 (0.09 to 0.37) | 0.22 (0.08 to 0.35) | 0.01 (−0.19 to 0.20) | 0.95 |
| | Lifetime smoking index | 0.40 (0.27 to 0.53) | 0.37 (0.24 to 0.51) | 0.02 (−0.16 to 0.21) | 0.80 |

AF, atrial fibrillation; CAD, coronary artery disease; CI, confidence interval.

Results are presented as effect sizes (95% CIs) for the association of smoking with heart failure. Effect sizes are scaled to per genetically predicted per 2.72-fold (1 log-odds unit) increase in the genetic liability to be a regular smoker and current smoker and per 1-SD increase in the genetic liability of the age at initiation of regular smoking, cigarettes per day, and lifetime smoking index.

^aTotal effect: the effect of the exposure on the heart failure.

^bDirect effect: the effect of the exposure on the heart failure, not explained by the mediator.

^cMediation effect: the effect of the exposure on the heart failure acting through the mediator.

never), smoking more cigarettes per day, and a higher composite lifetime smoking index was associated with a higher risk of HF. The results were robust and consistent in all sensitivity analyses and showed similar trends after adjusting for risk factors genetically correlated with HF; furthermore, these associations were independent of CAD and AF.

Aune *et al.*³⁶ performed a meta-analysis of 29 prospective cohort studies to explore the relationship between smoking and risk of HF. Results showed that the relative risk (RR) was 1.44 (95% CI, 1.34–1.55; $I^2 = 83%$) for ever regular smokers vs. never smokers. Compared with former smokers, current smokers showed a significantly higher risk of HF (RR, 1.58; 95% CI, 1.43–1.77; $I^2 = 77%$), which was consistent with our findings. In addition, there was a dose-dependent association between the number of cigarettes smoked and increased HF risk; 10 cigarettes per day was associated with a 41% increased risk of HF (RR, 1.41; 95% CI: 1.01–1.96), although only two studies with high heterogeneity ($I^2 = 82%$) were included. Our study further strongly supports this conclusion. We found that there was no association between older age at initiation of regular smoking and decreased HF risk with respect to the nature of the statistical test. With only 10 SNPs used as instruments and a small proportion of the phenotypic variance (0.2%) explained, the power was not sufficient to assess the causal association between age at initiation of regular smoking and HF, especially considering the consistent effect estimates with wider CI that are close to significance. Although relevant clinical evidence is scarce, there was research indicated that early tobacco exposure would lead to subclinical vascular damage in children and adolescents (<21 years old).³⁷ Therefore, the influence of early smoking exposure and HF requires further investigation. Aune *et al.*³⁶ showed that smoking cessation for more than 15 years lowered the risk of HF compared

with that of current smokers. While our study found that, for an individual who smoked 20 cigarettes per day for 15 years and quit smoking for 17 years (or smoked 60 cigarettes per day for 13 years and quit smoking for 22 years), there was still a 49% higher risk of HF compared with never smokers, thus highlighting the importance of never smoking.

Causality of HF due to smoking has a complicated biological mechanism. Smoking increases the risk of obesity,³⁸ type 2 diabetes mellitus,³⁹ hyperlipaemia,⁴⁰ hypertension,⁴¹ CAD,⁹ and AF,³² thus contributing to the onset of HF,^{11,12} arising from left ventricular dysfunction.⁴ Previous studies have shown that cigarette smoking is associated with endothelial dysfunction, arising from inflammation and oxidative stress, which act together to cause cardiac structure and arterial atherosclerosis.^{4,42} Additionally, carbon monoxide can lead to a large left ventricular mass and cardiac contractility dysfunction without effects on systemic blood pressure.⁴³ Alterations of the left ventricular structure and function induced by smoking may result in HF independently of CAD.⁴ After adjusting for traditional risk factors, the results were stable in all models. Although there are loci that overlap for CAD with HF (*LPA* and *9p21/CDKN2B-AS1*), and for AF with HF (*PITX2/FAM241A*),^{11,33} we identified that the smoking–HF risk was independent of CAD and AF. Our results are in line with those of a meta-analysis of prospective cohort studies indicating that current and ever smokers were associated with incident HF after adjusting for body mass index, diabetes, hypertension, serum cholesterol, CAD, and AF.³⁶

Our study comprises a comprehensive assessment of associations among different smoking phenotypes and HF. The first strength is that the most recent summarized data were used, and up to 47 309 HF cases and 930 014 controls were

included in this MR study. Second, we avoided the limitations of confounding factors, reverse causation, and regression dilution bias in observational studies by using genetic variant proxies for smoking exposures as instruments.

Nevertheless, this analysis was also subject to some limitations. First, the participating study definitions for HF differed in the HERMES Consortium. There was heterogeneity in the aetiology and clinical diagnosis of HF, which might influence the statistical power of genetic variants.¹¹ Second, we could not evaluate the associations among different durations of regular smoking and smoking cessation with HF; although a sensitivity analysis using lifetime smoking index as the integrated exposure was applied, more specific data are still needed. Third, the causal effect of smoking on HF might be mediated through other pathways, especially considering the pleiotropy of smoking^{9,10}; however, the results were robust in all complementary methods and sensitivity analyses and were in line with the multivariable MR analysis after correcting for risk factors correlated with both smoking and HF. Fourth, there was sample overlap in the exposure and outcome datasets. Of the 29 studies included in the GSCAN, five were also included in the HERMES Consortium (overlap of approximately 40% of individuals for each smoking phenotype) (Supporting Information, *Table S8*). In the sensitivity analysis of the association between the lifetime smoking index and HF, the UK Biobank was included in the GWAS meta-analysis of the lifetime smoking index and HERMES (overlap of approximately 40% of individuals), which might have led to some model overfitting.⁴⁴ However, our instrument selection showed a strong association with exposure (mean *F* statistics, 49.8), indicating that the bias or type 1 error inflation resulting from sample overlap was reasonably small. Furthermore, the potential bias due to overlap of the individuals may not have been substantial in the large consortia.⁴⁴ Fifth, this study included GWAS mainly consisting of individuals of European ancestry to avoid population stratification bias, which limited the scope of its application to some degree.

Conclusion

This MR study indicated that genetic predisposition to ever smoking and to a higher lifetime smoking burden is associated with a higher risk of HF. Our findings point to a potentially causal role of smoking in the pathogenesis of HF and highlight the importance of smoking cessation strategies to decrease the risk of HF.

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Conflict of interest

None declared.

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Supporting information

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Figure S1. Scatter plot of inverse-variance-weighted MR analyses of (A) smoking initiation, (B) age at initiation of regular smoking, (C) cigarettes per day, (D) smoking cessation, and (E) lifetime smoking index on heart failure.

Table S2. Characteristics of the genetic variants associated with smoking initiation.

Table S3. Characteristics of the genetic variants associated with age at initiation of regular smoking.

Table S4. Characteristics of the genetic variants associated with cigarettes per day.

Table S5. Characteristics of the genetic variants associated with smoking cessation.

Table S6. Characteristics of the genetic variants associated with lifetime smoking index.

Table S7. Associations of smoking with heart failure risk adjusting for its risk factors from multivariable MR-Egger.

Table S8. Summary of overlapped cohorts in the GWAS & Sequencing Consortium of Alcohol and Nicotine use and the Heart Failure Molecular Epidemiology for Therapeutic Targets Consortium.

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