Original Research Article

ENVIRONMENTAL EPIDEMIOLOGY

OPEN

# A cohort study on long-term exposure to air pollution and incidence of liver cirrhosis

Riccardo Orioli[a,b,*], Angelo G. Solimini[c], Paola Michelozzi[a], Francesco Forastiere[d,e], Marina Davoli[a], Giulia Cesaroni[a]

**Background:** Cirrhosis is an advanced liver disease affecting millions of people worldwide, involving high healthcare costs. Despite experimental evidence suggesting a possible role of airborne pollutants in liver diseases, epidemiological studies are lacking. We aimed at investigating the association between exposure to air pollutants and incidence of cirrhosis in a large population-based cohort in Rome.

**Methods:** We used an administrative cohort established from the 2001 census. We included all adults of 30 years of age or older who were free of cirrhosis, resulting in a study population of over 1.2 million subjects. Follow-up of the subjects ended on 31 December 2015. We ascertained incident cases of cirrhosis from regional mortality and hospital discharge registries using a validated algorithm. We assessed exposure of the subjects to $PM_{10}$, PM coarse, $PM_{2.5}$, $PM_{2.5}$ absorbance, $NO_2$, NOx, and PM metal components at their residential address using Land Use Regression models. We used Cox regression models, adjusted for relevant covariates, to estimate the association between air pollution exposure and cirrhosis incidence.

**Results:** We observed 10,111 incident cases of cirrhosis, with a crude incidence rate of 67 × 100,000 person-years. Long-term exposure to all pollutants tested was significantly associated with cirrhosis, e.g., $PM_{10}$ (hazard ratios [HR], 1.05; 95% confidence interval [CI], 1.01–1.09, per 10 μg/m$^3$ increments), PM coarse (HR, 1.11; 95% CI, 1.05–1.17, per 10 μg/m$^3$ increments), $PM_{2.5}$ (HR, 1.08; 95% CI, 1.03–1.13, per 5 μg/m$^3$ increments), and $NO_2$ (HR, 1.03; 95% CI, 1.02–1.05, per 10 μg/m$^3$ increments). The associations were robust in secondary analyses.

**Conclusions:** Our findings suggest a possible contribution of air pollution to the development of cirrhosis.

**Key Words:** Air pollution; Cirrhosis; Cohort study; Metal components; Particulate matter

## Introduction

Cirrhosis is an advanced liver disease that develops after many years of chronic liver injury. It is caused mainly by infections (chronic hepatitis B and C viruses), alcohol misuse, and nonalcoholic liver disease.[1] It is responsible for a huge burden of disease, resulting in about 1.27 million deaths and 38.9 million disability-adjusted life-years (DALYs) per year worldwide,[2,3] and is associated with high rates of disability, need for caregiving, and healthcare resource use.[4]

Although the chronic activation of inflammatory pathways plays a substantial role in the development of cirrhosis, the association between liver cirrhosis and exposure to air pollution has never been investigated before. In a cohort from the Framingham Heart Study, it was found that proximity to major roadways was associated with hepatic steatosis.[5] It was also hypothesized that air pollution could be involved in the progression from nonalcoholic fatty liver disease (NAFLD) to nonalcoholic steatohepatitis (NASH).[6] Evidence from experimental studies on mice suggests that air pollution could affect the liver by stimulating a local inflammatory response, with activation of Kupffer cells and production of cytokines,[7] by activating endoplasmic reticulum stress responses,[8] and by promoting collagen deposition and progression to fibrosis.[9] Other studies showed that some metal compounds present in the air pollution, such as vanadium, are also capable of inducing oxidative stress,

### What this study adds

Cirrhosis is a chronic disease involving millions of people worldwide, with a huge burden of disease and healthcare costs. Despite experimental studies supporting a possible association, the association between exposure to air pollution and liver cirrhosis has never been investigated before. Using Cox PH models in a large population-based cohort of residents in Rome, we found that long-term exposure to a wide range of pollutants was significantly associated with cirrhosis incidence. With the majority of the world population living in urban settings and being exposed to air pollution, these findings could have relevant implications for public health.

[a]Department of Epidemiology of Lazio Regional Health Service, Rome, Italy; [b]Hygiene and Public Health Service, Local Health Authority, Merano, Italy; [c]Department of Public Health and Infectious Diseases, Sapienza University of Rome, Rome, Italy; [d]Institute for Biomedical Research and Innovation, National Research Council, Palermo, Italy; [e]Environmental Research Group, King's College, London, United Kingdom

Sponsorships or competing interests that may be relevant to content are disclosed at the end of the article.

Since data used in this study include personally identifiable information, these data will not be made available. Analytic code may be available from the corresponding author upon request.

SDC Supplemental digital content is available through direct URL citations in the HTML and PDF versions of this article (www.environepidem.com).

*Corresponding Author. Address: Hygiene and Public Health Service, Local Health Authority, Via Goethe, 7, 39012 Merano, Italy. Tel.: +39 0473 251800. E-mail: riccardo.orioli86@gmail.com (R. Orioli)



Environmental Epidemiology (2020) 4:e109

Received: 17 December 2019; Accepted 8 July 2020

Published online 4 August 2020

DOI: 10.1097/EE9.0000000000000109

inflammatory infiltrates, and tissutal alterations suggestive for regenerative activity.[10,11]

Based on this evidence, we hypothesized that long-term exposure to air pollution could lead to the development of cirrhosis and that the contribution of the metal compounds of particulate matter (PM) could be relevant. We then investigated the association between major air pollutants, PM components and the incidence of cirrhosis in a large population-based cohort in Rome, Italy.

## Methods

### *Study design and population*

We selected the study population from the Rome Longitudinal Study, an administrative cohort of residents in Rome established from the 2001 census, whose characteristics have been already described.[12] Rome is the capital and main metropolitan city in Italy: it covers an area of about 1,290 km$^2$, and at 2001 census, it had a population of about 2.5 million inhabitants.[12]

Eligibility criteria for the enrollment into the cohort were 30 years of age and older, resident in Rome since 5 years or longer, not living in institutions (prisons, hospitals, or nursing homes) at the date of enrollment (21 October 2001), yielding a cohort of 1,265,058 subjects. Using record linkage with the regional Hospital Discharge Register, we excluded subjects who had any hospitalization for cirrhosis in the previous 5 years before enrollment and those with missing information on the personal identifying code, on exposure to air pollution, or on any of the covariates. The final study population consisted of 1,245,397 subjects.

Follow-up for vital status (Rome Municipal Register) started on 21 October 2001, and ended on 31 December 2015; subjects who emigrated outside Rome during the study period were considered lost to follow-up at the date of emigration.

We obtained data on the covariates from census data, although information on residential history was available from Rome Municipal Register. Using record linkage procedures, we collected information about participants' hospitalizations from the regional Hospital Discharge Register. This register includes all discharges from public and private hospitals, coded according to the *9th Revision of the International Classification of Diseases—Clinical Modification* (ICD-9-CM). In the discharge abstracts present in the Register, physicians can code one main diagnosis and up to five secondary diagnoses. The main diagnosis corresponds to the main disease or condition that led to the hospitalization or to the one that absorbed most of the economic resources during the hospital stay. The secondary diagnoses correspond to other relevant diseases. Together with other parameters (e.g., length of stay, interventions/procedures, etc.), the diagnosis codes are used to calculate the reimbursement given to healthcare facilities by the State.

### *Outcome*

We ascertained incident cases of cirrhosis searching for appropriate ICD-9-CM codes among the main and secondary diagnoses of hospitalizations and among the causes of death that occurred during the follow-up period. We adopted a modified version of the algorithm validated by Nehra et al,[13] which showed a sensitivity of 98% and a positive predictive value of 78% in a validation cohort of patients with known cirrhosis. We defined as incident cirrhosis cases those subjects who had at least one code (main and five secondary diagnoses) for cirrhosis (571.2 alcoholic cirrhosis, 571.5 cirrhosis without mention of alcohol) or one code for specific hepatic decompensation events (456.0-456.1-456.2-456.21 esophageal varices; 572.2 hepatic encephalopathy; 572.3 portal hypertension; 572.4 hepatorenal syndrome). We did not include the code for spontaneous bacterial peritonitis in our algorithm, as this code is missing in the Italian version of ICD-9-CM, and the other codes for peritonitis are nonspecific. In subjects with multiple records, we considered the first event as the time of diagnosis.

### *Covariates*

We included known individual-level potential confounders at inclusion: age, sex, birthplace (Rome or other places), educational level (university, high school, junior high school, primary school or less), occupational status [highly qualified nonmanual workers (i.e., managers, professors, researchers); other nonmanual workers; manual workers; other workers (i.e., armed forces, retail sales); housewives; unemployed; retired; other condition], and marital status (married, single, separated/divorced, widowed). Other individual-level factors also play a role in the development of liver cirrhosis, such as income, alcohol consumption, and previous hepatitis B virus or hepatitis C virus infections, but this kind of individual information was unavailable. However, because area-level socioeconomic conditions have been shown to be associated with some of these factors independently of individual position,[14] we adjusted for an area-level indicator based on the data from the 2001 census. The index was derived from a factor analysis based on educational level, occupation, home ownership, family composition, crowding, and immigrant status at the census block level (500 inhabitants per block, on average). We then classified the socioeconomic position (SEP) in five levels (very high, high, intermediate, low, and very low), according to quintiles of the distribution.[12]

### *Exposure assessment*

For exposure assessment, we applied the Land Use Regression (LUR) models developed in two European projects: the ESCAPE (European Study of Cohorts for Air Pollution Effects) and the TRANSPHORM (Transport-related air pollution and health impacts—integrated methodologies for assessing particulate matter) projects.[15–17] The ESCAPE project allowed to measure and develop LUR models for major pollutants: particulate matter ($PM_{10}$, coarse PM, $PM_{2.5}$), nitrogen oxides ($NO_2$ and NOx), and absorbance of $PM_{2.5}$ (a proxy of black carbon). The TRANSPHORM project allowed to measure and develop LUR models for selected metal components of particulate matter, a priori chosen to represent diverse sources of air pollution: copper (Cu), iron (Fe), and zinc (Zn) were chosen for representing non-tailpipe emissions; sulfur (S) for long-range transport; silicon (Si) for crustal material, nickel (Ni) and vanadium (V) for industries and mixed oil burning; and potassium (K) for biomass burning and soil.[17]

The measurement campaigns were conducted in Rome during 2010. Nitrogen oxides were measured using passive samplers in 40 sites, and particulate matter, $PM_{2.5}$ absorbance, and metal components were measured using Harvard Impactors in 20 sites, chosen to represent the spatial distribution of residential addresses. In each site, concentrations of pollutants were measured for 14 days in the cold, intermediate, and warm seasons. Measurements at a reference background site were used to calculate a temporal correction to obtain an annual average concentration at each site.[17–19] The average 2010 annual concentrations were used to develop the LUR models using several predictor variables (GIS, land use, traffic, etc.). In this study, we chose to use all the LUR models with a cross-validated $R^2 > 0.50$: all the major pollutants ($PM_{10}$, coarse PM, $PM_{2.5}$, the absorbance of $PM_{2.5}$, $NO_2$, and NOx), Cu, Fe, and Zn components of $PM_{2.5}$, and all the components of $PM_{10}$ except sulfur (S). We attributed exposure to the coordinates in the residential address at the time of enrollment under the assumption that the spatial distribution of pollutants did not change over time.

### *Main analysis*

We assessed the association between air pollution and liver cirrhosis using Cox proportional hazard models, with age as the time scale and stratifying by sex the baseline hazard rates. Subjects were censored at the time of diagnosis, death, emigration from Rome, or at 31 December 2015, depending on which came first. We developed separate models for each main pollutant and metal component and reported both the unadjusted association (unadjusted models) and the association adjusted for the full set of individual- and area-level covariates (adjusted models). We analyzed each main pollutant and metal component at a time. We calculated adjusted hazard ratios (HRs) and 95% confidence intervals (CIs) using time-invariant linear terms for the pollutants with the fixed increments used in previous studies.[20] We also used categorical variables defined by quartile of the distribution and linear terms with interquartile range (IQR) increments. We tested for the proportionality assumption of the hazards correlating, for each covariate, the corresponding set of scaled Schoenfeld residuals with time. Additionally, we tested for the model as a whole.

We included interaction terms between exposure to air pollutants and potential effect modifiers: sex, age class (30–49, 50–64, 65–74, ≥75), educational level, occupational status (in this analysis, we merged all workers in one category, obtaining a five-level variable: employed, unemployed, housewife, retired, other condition), and area-level SEP. We performed likelihood-ratio tests to assess effect modification.

To explore the shape of the association between air pollution and cirrhosis, we replaced the linear terms with natural splines, with two degrees of freedom in the adjusted models.[21] We then compared the resulting models and the adjusted models using the likelihood-ratio test.

### *Secondary analyses*

In a sensitivity analysis, to adopt a more specific case definition, we selected as cirrhotic only subjects who had at least three records or died for cirrhosis (narrow case definition). In fact, in the study by Nehra et al,[13] the diagnosis of cirrhosis was confirmed in about 96% of subjects with three or more ICD-9 codes included in the algorithm.

To exclude the presence of a spurious or inflated association due to the possibility of a higher risk of hospitalization for other diseases linked to air pollution, we tested the association using only hospitalizations that had cirrhosis or its complication codes as main diagnosis.

Cirrhosis is an end-stage disease occurring after many years of persistent liver damage. To evaluate the association in subjects affected by hepatic diseases or conditions that can lead to cirrhosis, we created a sub-cohort of the study population composed only by subjects who had at least one hospitalization for liver diseases or alcohol-related conditions (for the complete set of ICD-9 codes, see the eAppendix; http://links.lww.com/EE/A104). Subjects entered the study at the time of first hospitalization for such conditions and were followed until the date of first hospitalization for cirrhosis, death, migration from Rome, or the end of follow-up.

Furthermore, to assess how robust were the associations to potential unmeasured confounding, we estimated the E-value.[22] Briefly, this indicator represents the minimum strength of the association that an unmeasured confounder would need to have with both the exposure and the outcome to fully explain a specific exposure–outcome association, conditional on the measured covariates (the larger the E-value, the more considerable unmeasured confounding would be needed to explain an association).

We also tested if associations were robust when using sources of information different from hospitalizations and death registers. To do that, we used information on exemptions to co-payment for healthcare services linked to medical conditions. Because we had data from 2005 onwards, we created a sub-cohort of patients, with follow-up starting at 1 January 2010, to have at least 5 years of exemptions data to exclude prevalent cases. The new case definition included subjects who matched the main analysis definition plus those who had an exemption code for cirrhosis (008.571.2 alcoholic cirrhosis; 008.571.5 nonalcoholic cirrhosis; N = 1,229), resulting in 3,694 cases. All cases occurring before or on 1 January 2010, according to this definition were excluded from the analysis (N = 9,272), thus removing 14 years of prevalent cases and allowing a longer time of exposure before disease onset. This was in line with the hypothesized long induction time.

Furthermore, in order to evaluate the presence of exposure misclassification due to residential address changes, we performed a separate analysis on the 2010 sub-cohort restricted to the subjects who did not change their residential address during the previous 14 years (from 1 January 1996 to 31 December 2009).

To test for the presence of any spatial dependence within the study population, we fitted models with a frailty term for neighborhoods. We adopted the definition established by the Municipality administration, which divides the municipal territory into 94 areas, with an average population of about 13,000 people.

Finally, to assess the independent contribution of each pollutant, we performed two pollutant models. We tested those couples of main pollutants and of metal components with the relative PM mass, which had Pearson's correlation coefficients <0.7.

## Results

Of the 1,265,058 eligible subjects, we excluded 1,332 who had missing information on the personal identifying code, 15,930 with missing exposure to air pollution, five who had missing information on the covariates, and 2,394 subjects who were not free of the disease at enrollment, yielding a study population of 1,245,397 subjects. The overall cohort follow-up period accounted for more than 15 million person-years of observations. We observed 10,111 new cases of cirrhosis, with a crude incidence rate of 67 × 100,000 person-years. Table 1 shows the characteristics of the study population with person-years of follow-up, the number of new cases of cirrhosis, and crude rates with 95% CI.

Compared with non-cirrhotic subjects, those who developed cirrhosis were more frequently males (59.5% vs. 45.4%), were older (mean age, 63.3 vs. 55.0), and had lower educational attainment (primary school or less, 41.0% vs. 24.6%), and area-level socioeconomic conditions (very low SEP, 24.3% vs. 19.2%). They also had dramatically higher mortality rates, as only 23.2% was alive at the end of follow-up compared with 71.1% of non-cirrhotic subjects. The overall mean (SD) exposure to main pollutants was as follows: 36.6 μg/m$^3$ (5.1) for $PM_{10}$, 17.0 μg/m$^3$ (3.3) for PM coarse, 19.6 μg/m$^3$ (1.9) for $PM_{2.5}$, 2.71 × 10$^{-5}$/m (0.5) for $PM_{2.5}$ absorbance, 42.8 μg/m$^3$ (10.2) for $NO_2$, and 85.0 μg/m$^3$ (24.2) for NOx. As expected, Pearson's correlation coefficients between pollutants were high, particularly among particulate matters ($PM_{10}$, $PM_{2.5}$, and PM coarse) and among some PM elemental constituents (eTable 2; http://links.lww.com/EE/A104).

Cirrhosis incidence was significantly associated with all main pollutants and metal components tested (Table 2). The adjusted HRs for main pollutants were as follows: $PM_{10}$ (HR, 1.05; 95% CI, 1.01–1.09], per 10 μg/m$^3$ increments), PM coarse (HR, 1.11; 95% CI, 1.05–1.17, per 10 μg/m$^3$ increments), $PM_{2.5}$ (HR, 1.08; 95% CI, 1.03–1.13, per 5 μg/m$^3$ increments), $PM_{2.5}$ absorbance (HR, 1.09; 95% CI, 1.05–1.13, per 1 × 10$^{-5}$/m increments), $NO_2$ (HR, 1.03; 95% CI, 1.02–1.05, per 10 μg/m$^3$ increments), and

**Table 1.**

**Baseline characteristics of the study population: Rome 2001–2015.**

| Characteristic | N (%) | Person-years | N cirrhosis cases | Crude rate (per 100,000) | 95% CI | |
|---|---|---|---|---|---|---|
| Age at enrollment | | | | | | |
| 30–49 | 517,409 (41.6) | 6,806,497 | 1,892 | 27.8 | 26.6 | 29.1 |
| 50–64 | 374,107 (30.0) | 4,753,865 | 2,983 | 62.7 | 60.5 | 65.0 |
| 65–74 | 206,270 (16.6) | 2,361,719 | 3,248 | 137.3 | 132.7 | 142.1 |
| ≥75 | 147,611 (11.9) | 1,150,576 | 1,988 | 172.5 | 165.1 | 180.3 |
| Sex | | | | | | |
| Males | 566,724 (45.5) | 6,744,907 | 6,014 | 89.1 | 86.9 | 91.4 |
| Females | 678,673 (54.5) | 8,327,749 | 4,097 | 49.2 | 47.7 | 50.7 |
| Place of birth | | | | | | |
| Rome | 645,267 (51.8) | 8,095,980 | 4,406 | 54.4 | 52.8 | 56.0 |
| Other | 600,130 (48.2) | 6,976,676 | 5,705 | 81.7 | 79.6 | 83.9 |
| Marital status | | | | | | |
| Married | 826,301 (66.4) | 10,238,535 | 6,444 | 62.9 | 61.4 | 64.5 |
| Single | 190,042 (15.3) | 2,366,776 | 1,236 | 52.2 | 49.4 | 55.2 |
| Separated/divorced | 87,324 (7.0) | 1,080,162 | 674 | 62.3 | 57.8 | 67.2 |
| Widowed | 141,730 (11.4) | 1,387,183 | 1,757 | 126.5 | 120.7 | 132.5 |
| Educational level | | | | | | |
| University | 203,032 (16.3) | 2,558,242 | 1,010 | 39.5 | 37.1 | 42.0 |
| High school | 411,282 (33.0) | 5,200,798 | 2,118 | 40.7 | 39.0 | 42.5 |
| Secondary school | 322,640 (25.9) | 3,939,972 | 2,835 | 71.9 | 69.3 | 74.6 |
| Primary school or less | 308,443 (24.8) | 3,373,644 | 4,148 | 122.8 | 119.1 | 126.6 |
| Occupational status | | | | | | |
| Employed NM I | 168,229 (13.5) | 2,201,795 | 655 | 29.7 | 27.5 | 32.1 |
| Employed NM II | 199,115 (16.0) | 2,622,310 | 679 | 25.9 | 24.0 | 27.9 |
| Employed M | 123,174 (9.9) | 1,581,173 | 779 | 49.2 | 45.9 | 52.8 |
| Employed, other | 80,925 (6.5) | 1,048,185 | 371 | 35.4 | 32.0 | 39.2 |
| Housewives | 261,635 (21.0) | 3,146,070 | 2,078 | 66.0 | 63.2 | 68.9 |
| Unemployed | 61,760 (5.0) | 787,538 | 670 | 85.0 | 78.8 | 91.7 |
| Retired | 291,706 (23.4) | 3,072,452 | 4,011 | 130.4 | 126.4 | 134.5 |
| Other condition | 58,853 (4.7) | 613,133 | 868 | 141.3 | 132.2 | 151.0 |
| Area-level SEP | | | | | | |
| Very high | 247,844 (19.9) | 3,000,234 | 1,564 | 52.1 | 49.6 | 54.8 |
| High | 254,917 (20.5) | 3,070,400 | 1,879 | 61.2 | 58.5 | 64.0 |
| Medium | 250,225 (20.1) | 3,016,232 | 1,990 | 65.9 | 63.1 | 68.9 |
| Low | 253,250 (20.3) | 3,068,287 | 2,222 | 72.4 | 69.4 | 75.4 |
| Very low | 239,161 (19.2) | 2,917,502 | 2,456 | 84.1 | 80.8 | 87.5 |

Reported data are complete for all study participants.

Employed NM I, highly qualified nonmanual workers (i.e., managers, professors, researchers); Employed NM II, other nonmanual workers; Employed M, manual workers; Employed, other, other workers (i.e., armed forces, retail sales); Other condition, all subjects who cannot be classified into the previous categories. Area-level SEP indicates composite indicator of socioeconomic position at the census block level.

NOx (HR, 1.04; 95% CI, 1.03–1.06, per 20 μg/m$^3$ increments). The use of quartiles of exposure to air pollutants confirmed the results, and we found a significant trend for all pollutants and components, except $PM_{10}$ K (eTable 3; http://links.lww.com/EE/A104). When using IQR increments, we observed stronger associations with $PM_{10}$ nickel (HR, 1.07; 95% CI, 1.05–1.10) and NOx (HR, 1.07; 95% CI, 1.05–1.10; eTable 4; http://links.lww.com/EE/A104).

We did not find evidence of effect modification except by age class (for PM coarse, $PM_{2.5}$, $PM_{2.5}$ absorbance, $NO_2$, NOx, and $PM_{10}$ Ni) and area-level SEP (for $PM_{2.5}$). The associations were generally stronger among older people (≥75 and 65–74 years) and among subjects with a very high or very low area-level SEP (eTable 5; http://links.lww.com/EE/A104 and eTable 6; http://links.lww.com/EE/A104).

The shape of the association was usually linear, although we observed some evidence of deviation from linearity for PM coarse, $PM_{2.5}$ absorbance, and $NO_2$ (likelihood ratio test; $P < 0.05$), with steeper curves at lower concentrations followed by a final plateau (eFigure 1; http://links.lww.com/EE/A104).

Results of the sensitivity analyses for main pollutants and PM components are shown in Figures 1 and 2, respectively. The associations were not substantially altered by any sensitivity analysis, although they were weakened when we used only the main diagnosis code to define cases. In the sub-cohort of individuals affected by medical conditions leading to cirrhosis and in the sub-cohort starting from 2010 that included exemptions to co-payments in the case definition, the associations were generally strengthened.

The E-value for the adjusted associations were 1.28 for $PM_{10}$, 1.46 for PM coarse, 1.37 for $PM_{2.5}$, 1.21 for $NO_2$ and ranged from a minimum of 1.16 for $PM_{10}$ K to a maximum of 1.62 for $PM_{10}$ V (eTable 7; http://links.lww.com/EE/A104). In two pollutant models, the association always remained significant for $PM_{2.5}$ absorbance, $NO_2$, NOx, and all components tested (eTable 8; http://links.lww.com/EE/A104 and eTable 9; http://links.lww.com/EE/A104).

## Discussion

We observed an association between air pollution and the incidence of liver cirrhosis in a large Italian population-based cohort. There were significant associations with both the main pollutants and the metal components of particulate matter. Our findings were robust to many sensitivity analyses, including a more restrictive and specific case definition, the use of the main hospitalization diagnosis only, the selective inclusion of subjects already predisposed to the development of cirrhosis, the restriction to non-movers, and the use of data sources other than hospitalizations.

We estimated a crude incidence rate of 67 × 100,000 person-years in our study. We are not aware of other estimates of

**Table 2.**

**Adjusted HRs and 95% CI for the association between ambient air pollution and liver cirrhosis in the Rome Longitudinal Study (main analysis): Rome 2001–2015.**

| | Incidence of cirrhosis (n = 10,111) | |
|---|---|---|
| **Exposure** | **HR[a] (95% CI)** | **HR[b] (95% CI)** |
| Main pollutants | | |
| $PM_{10}$ | 1.02 (0.98–1.06) | 1.05 (1.01–1.09) |
| PM coarse | 1.00 (0.94–1.06) | 1.11 (1.05–1.17) |
| $PM_{2.5}$ | 1.03 (0.98–1.08) | 1.08 (1.03–1.13) |
| $PM_{2.5}$ abs | 1.05 (1.01–1.09) | 1.09 (1.05–1.13) |
| $NO_2$ | 1.04 (1.02–1.06) | 1.03 (1.02–1.05) |
| NOx | 1.05 (1.04–1.07) | 1.04 (1.03–1.06) |
| Metal components | | |
| $PM_{10}$Cu | 1.01 (0.99–1.02) | 1.03 (1.01–1.04) |
| $PM_{10}$Fe | 1.00 (0.98–1.02) | 1.03 (1.01–1.04) |
| $PM_{10}$K | 1.01 (1.00–1.03) | 1.02 (1.00–1.04) |
| $PM_{10}$Ni | 1.11 (1.08–1.14) | 1.10 (1.07–1.14) |
| $PM_{10}$Si | 1.13 (1.09–1.16) | 1.06 (1.03–1.09) |
| $PM_{10}$V | 1.11 (1.03–1.19) | 1.17 (1.09–1.25) |
| $PM_{10}$Zn | 1.00 (0.97–1.03) | 1.07 (1.04–1.11) |
| $PM_{2.5}$Cu | 1.02 (1.00–1.04) | 1.05 (1.03–1.08) |
| $PM_{2.5}$Fe | 0.99 (0.97–1.02) | 1.06 (1.04–1.09) |
| $PM_{2.5}$Zn | 1.04 (0.99–1.08) | 1.08 (1.04–1.13) |

[a]Models stratified by sex and age as the time scale.

[b]Models adjusted for educational level, occupational status, marital status, place of birth, area-level SEP and stratified by sex and age as the time scale. Hazard ratios per fixed increments of pollutants: $PM_{10}$, 10 μg/m³; PM coarse, 10 μg/m³; $PM_{2.5}$, 5 μg/m³; $PM_{2.5}$ abs, $1 \times 10^{-5}$/m; $NO_2$, 10 μg/m³; Nox, 20 μg/m³; $PM_{10}$Cu, 20 ng/m³; $PM_{10}$Fe, 500 ng/m³; $PM_{10}$K, 100 ng/m³; $PM_{10}$Ni, 2 ng/m³; $PM_{10}$Si, 500 ng/m³; $PM_{10}$V, 3 ng/m³; $PM_{10}$Zn, 20 ng/m³; $PM_{2.5}$Cu, 5 ng/m³; $PM_{2.5}$Fe, 100 ng/m³; $PM_{2.5}$Zn, 10 ng/m³.

cirrhosis incidence in the general population in Italy to make a comparison. Actual estimates in other European countries include an incidence rate of 23.2 × 100,000 person-years from a UK cohort study and of 30.7 × 100,000 person-years from a cohort study conducted in Sweden,[23,24] but these countries were reported to have a lower prevalence of the disease when compared with that in Italy.[25]

To our knowledge, this is the first longitudinal study investigating the association between exposure to air pollution and the development of cirrhosis. The few epidemiological studies on this topic present in the literature deal mainly with NAFLD, which is becoming one of the main causes of cirrhosis in developed countries after hepatitis C virus infection and alcohol misuse.[1] In a study conducted on participants of the Framingham Heart Study, the authors observed an association between proximity to major roadways and hepatic steatosis, while the association with $PM_{2.5}$ was less consistent.[5] Other authors raised the possibility of a role played by air pollution in the progression from NAFLD to NASH, a crucial step in the natural history of the disease leading to cirrhosis.[6] They also hypothesized that PM-induced hepatic toxicity, hesitating in systemic insulin resistance, could be one of the mechanisms involved in the association with cardiovascular diseases and diabetes mellitus. In a study on an obese pediatric population, traffic-related air pollution was positively associated with cytokeratin-18 levels, a marker for hepatocellular apoptosis.[26] Previous studies also found evidence of an association between air pollution and both incidence of and survival from primary liver cancer, which is one of the possible complications of cirrhosis.[27,28] Some studies suggested an association between cigarette smoking and chronic

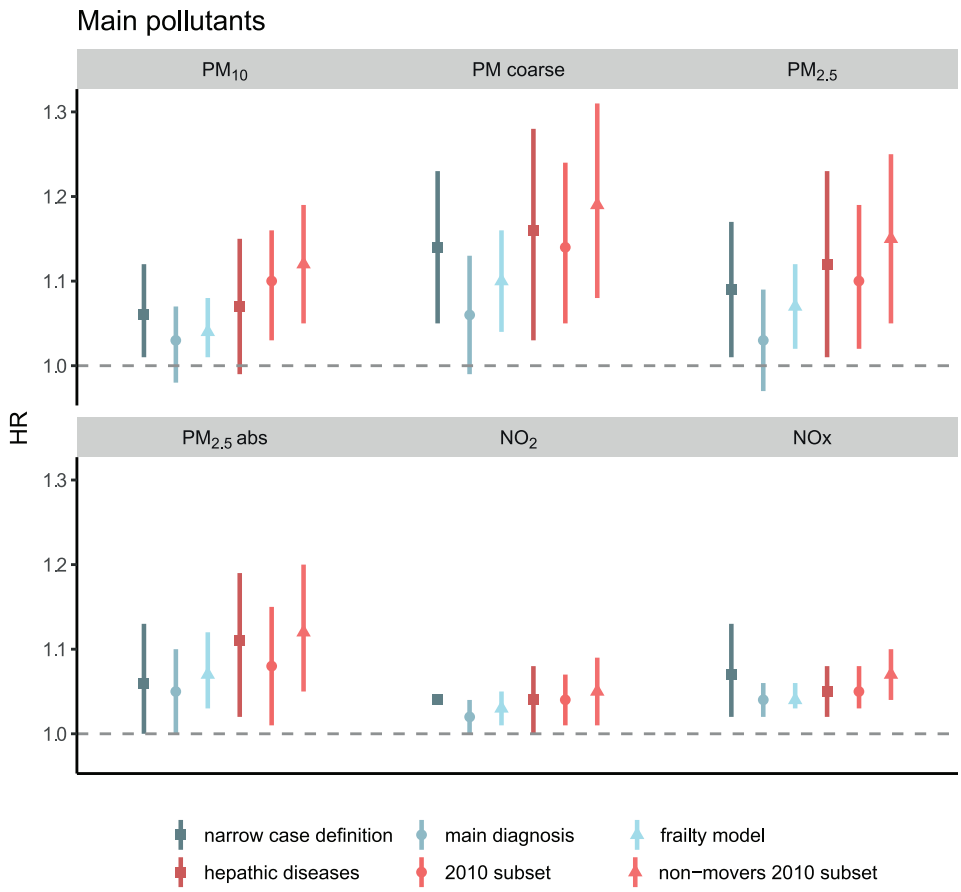


**Figure 1.** Adjusted hazard ratios (HRs) and 95% confidence intervals (95% CI) for the association between main pollutants and liver cirrhosis according to six secondary analyses. Models adjusted for educational level, occupational status, marital status, place of birth, area-level SEP and stratified by sex and age as the time scale. Hazard ratios per fixed increments of pollutants: $PM_{10}$, 10 μg/m³; PM coarse, 10 μg/m³; $PM_{2.5}$, 5 μg/m³; $PM_{2.5}$abs, $1 \times 10^{-5}$/m; $NO_2$, 10 μg/m³; NOx, 20 μg/m³.

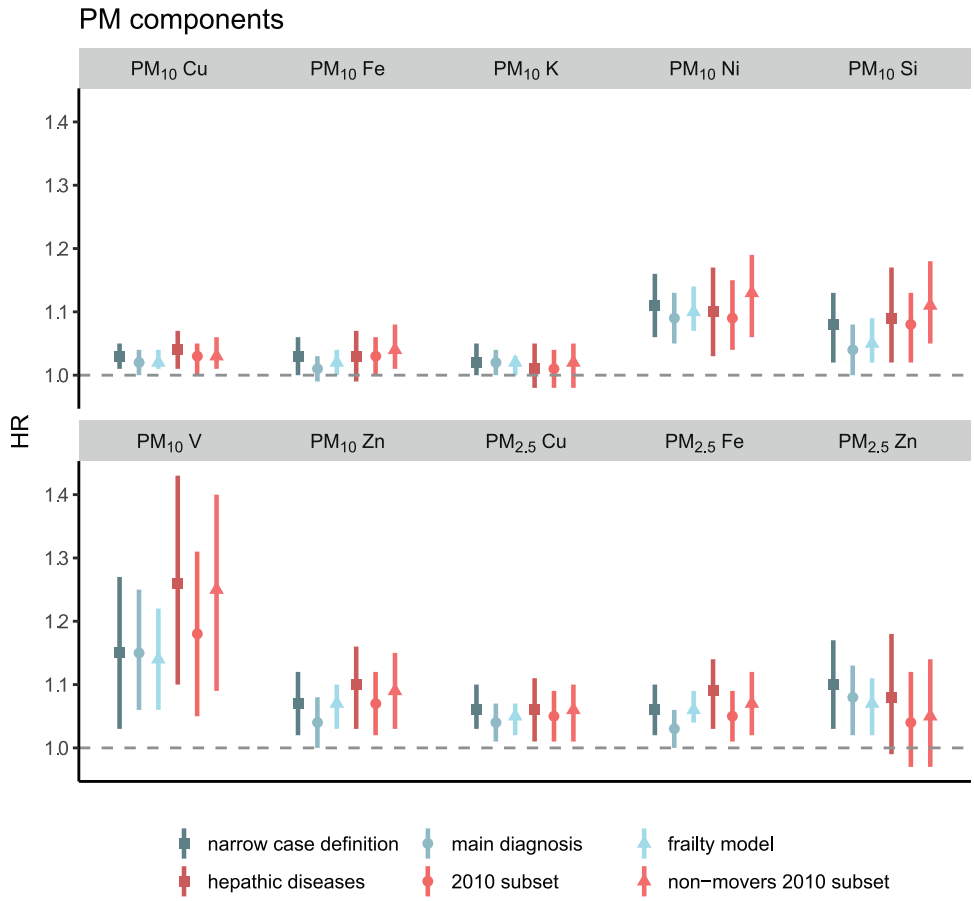


**Figure 2.** Adjusted hazard ratios (HRs) and 95% confidence intervals (95% CI) for the association between PM components and liver cirrhosis according to six secondary analyses. Models adjusted for educational level, occupational status, marital status, place of birth, area-level SEP and stratified by sex and age as the time scale. Hazard ratios per fixed increments of pollutants: $PM_{10}$Cu, 20 ng/m$^3$; $PM_{10}$Fe, 500 ng/m$^3$; $PM_{10}$K, 100 ng/m$^3$; $PM_{10}$Ni, 2 ng/m$^3$; $PM_{10}$Si, 500 ng/m$^3$; $PM_{10}$V, 3 ng/m$^3$; $PM_{10}$Zn, 20 ng/m$^3$; $PM_{2.5}$Cu, 5 ng/m$^3$; $PM_{2.5}$Fe, 100 ng/m$^3$; $PM_{2.5}$Zn, 10 ng/m$^3$.

liver disease.[29,30] Both inhaled cigarette smoke and air pollution contain complex mixtures of carcinogenic chemical compounds that induce systemic oxidative stress and affect many target organs. Despite the paucity of epidemiological studies on the specific topic, evidence from in vivo experimental studies suggests many mechanisms through which air pollution could affect hepatic health and promote the development of a cirrhotic liver. In a model proposed by Kim et al,[31] they hypothesized that some fractions of air pollutants could translocate from the lungs into the general circulation and then into the liver, stimulating a local inflammatory response. In murine models, inhaled particulate matter was shown to induce liver tissue inflammation through the activation of Kupffer cells and production of cytokines,[7] to activate endoplasmic reticulum stress response,[8] and to promote collagens deposition causing progression to fibrosis.[9] Also gasoline fumes were found to increase the levels of hepatic enzymes, liver oxidative stress markers, and inflammatory markers of exposed rats, inducing hepatocyte toxicity and collagen fiber deposition.[32] Another study found that mice exposed to $PM_{2.5}$ developed a NASH-like phenotype and showed impaired hepatic glycogen storage, glucose intolerance, and insulin resistance.[33] Interestingly, we observed strong associations with some metal components of particulate matter, especially with vanadium (V) and nickel (Ni) components of $PM_{10}$, independent of adjustment for $PM_{10}$ mass in two pollutant models. These results suggest that the contribution of metal components, and in particular those deriving from industrial emissions, could be relevant. This is also consistent with the findings from experimental studies: inhalation of vanadium compounds was seen to be associated with oxidative stress, inflammatory infiltrates, and alterations suggestive for regenerative activity in the livers of exposed mice.[10,11] However, in addition to industrial pollutants, the analysis using IQR increments highlighted an important role of both exhaust (NOx) and nonexhaust ($PM_{2.5}$Cu and $PM_{2.5}$Fe) traffic pollutants.

The shape of the associations between air pollution exposure and cirrhosis incidence was generally linear. However, for PM coarse, $PM_{2.5}$ absorbance, and $NO_2$ the spline models fitted better than linear models (likelihood ratio test $P < 0.05$). eFigure 1 (http://links.lww.com/EE/A104) shows that the shape of the associations was substantially linear for most of its length, with a plateau effect followed by a descending curve at higher exposure levels, where there are few observations and data are less reliable. However, nonlinear dose–response curves are common in toxicology and have been already reported for the association between air pollution and health outcomes.[12,34,35]

The strengths of this study are the use of a large population-based cohort that covers almost all eligible individuals living in the Italian capital, the long observation period accounting for more than 15 million person-years, and the availability of personal and area-level socioeconomic covariates. Moreover, we had the possibility to explore the association with both major pollutants and PM-mass metal components derived from fine-scale standardized LUR models, which were rarely available in previous epidemiological studies.

This study also presents several limitations. The most relevant one is that we could not rely on information about behavioral risk factors that are known to play a role in the disease, like

alcohol consumption, smoking status, body mass index, diet and physical activity, not about other important clinical information related to chronic viral hepatitis and its treatment. We had information on the socioeconomic status that are strongly correlated to them, and this could partly attenuate the effect of unmeasured confounding related to such risk factors. Furthermore, in the sub-cohort analysis including only subjects with hospitalizations for chronic liver diseases or alcohol-related conditions, the associations did not change or even got stronger. In our opinion, it is very unlikely that a differential distribution of the risk factors, in relationship to air pollution exposure, could be equally reproduced within both the general population and this at-risk subgroup.

We found that the E-values for our associations arrived up to 1.62. Although the association between behavioral factors and cirrhosis could easily exceed this value, it seems less probable that it could contemporarily do the same with exposure to air pollution (given the adjustment for socioeconomic conditions). In fact, previous evidence on the same population showed that exposure to air pollution was higher among people with better socioeconomic conditions in Rome.[36] In summary, these findings suggest that it is unlikely that potential unmeasured confounding could fully explain the associations we observed. However, we cannot completely exclude the presence of confounding and biased estimates, and this should be considered when interpreting our results.

Most cases of cirrhosis, especially in early stages, are usually subclinical until a decompensation event (like ascites, hepatic encephalopathy, variceal bleeding, and sepsis) occurs, making it difficult to estimate the real prevalence and incidence of the disease. However, although the contribution of air pollution to subclinical cirrhosis remains unknown, the clinical events that characterize decompensated cases lead usually to an access to emergency departments and subsequent hospitalization. Therefore, we think that hospital admission data could be adequate in estimating the real incidence of severe/decompensated cirrhosis cases. Moreover, our results did not change when we included other sources of information (exemptions to co-payments).

In a sensitivity analysis, we tested the association including only cases that had cirrhosis or its complications as main diagnosis code, finding a generalized attenuation of the associations. A possible explanation is that hospitalizations for other diseases related to air pollution could have inflated our estimates. However, excluding cases found through secondary diagnoses (37.5% of the total) could introduce another distortion due to delayed diagnosis. We also underline that none of the estimates dropped to one, and the association remained significant for the majority of studied pollutants, including $PM_{2.5}$abs, NOx, $PM_{10}$Cu, $PM_{10}$Ni, $PM_{10}$V, $PM_{10}$Zn, $PM_{2.5}$Cu, $PM_{2.5}$Fe, and $PM_{2.5}$Zn.

Exposure assessment was made using estimated averaged levels for 2010, many years after the beginning of follow-up; furthermore, using a single-year exposure, we were not able to consider temporal variations of air pollutant concentrations. This may result in exposure misclassification and give biased estimates. However, previous studies showed that for $NO_2$, LUR models give stable measures of spatial concentration contrast over time.[37,38] There is no available evidence about the temporal trends of metal components, but we believe that emission patterns in Rome did not change substantially over a decade. We also attributed exposure to the residential address at the time of enrollment. Since we hypothesized that air pollution might promote the development of cirrhosis in the long-term, after many years of exposure, the use of the residential address at the time of enrollment could result in exposure misclassification too. However, in a secondary analysis with follow-up starting on 1 January 2010, and including only subjects who maintained the same residential address during the previous 14 years (1996–2009), the associations were strengthened (Figures 1 and 2), suggesting that misclassification due to changes in residential address should not have inflated our estimates. As in most previous epidemiological studies, no information was available about individual movements for work purposes or other activities during daily life.

## Conclusions

We found evidence of an association between exposure to both major pollutants and PM metal components and the incidence of liver cirrhosis. Even among subjects affected by clinical conditions leading to cirrhosis, exposure to air pollution was associated with a significant increase in the risk of cirrhosis, suggesting that it may act in addition to other main sources of chronic liver damage and promote the development of the disease. However, the lack of information on individual-level risk factors, such as alcohol consumption and viral hepatitis infection, is a major limitation of our study and should be considered when interpreting our results. Exposure misclassification could have been another source of bias, as time-varying exposure and a full residential history were not available.

Nonetheless, if confirmed by other studies with more detailed individual-level information and better exposure assessment, these results could contribute to a better knowledge of the health effects of air pollutants and open to new research on the role of environmental pollution in hepatic diseases. Due to the high morbidity and mortality rates and economic impact of cirrhosis, even a small impact of air pollution on such disease could have relevant implications for public health.

## Conflicts of interest statement

The authors declare that they have no conflicts of interest with regard to the content of this report.

## References

1. Tsochatzis EA, Bosch J, Burroughs AK. Liver cirrhosis. Lancet. 2014;383:1749–1761.
2. Naghavi M, Abajobir AA, Abbafati C, et al. Global, regional, and national age-sex specifc mortality for 264 causes of death, 1980–2016: a systematic analysis for the Global Burden of Disease Study 2016. Lancet. 2017;390:1151–1210.
3. Abajobir AA, Abate KH, Abbafati C, et al. Global, regional, and national disability-adjusted life-years (DALYs) for 333 diseases and injuries and healthy life expectancy (HALE) for 195 countries and territories, 1990–2016: a systematic analysis for the Global Burden of Disease Study 2016. Lancet. 2017;390:1260–1344.
4. Miquel M, Clèries M, Vergara M, Vela E. Economic burden of cirrhosis in Catalonia: a population-based analysis. BMJ Open. 2018;8:e018012.
5. Li W, Dorans KS, Wilker EH, et al. Residential proximity to major roadways, fine particulate matter, and hepatic steatosis: the Framingham Heart Study. Am J Epidemiol. 2017;186:857–865.
6. Conklin DJ. From lung to liver: how does airborne particulate matter trigger NASH and systemic insulin resistance? J Hepatol. 2013;58:8–10.
7. Tan HH, Fiel MI, Sun Q, et al. Kupffer cell activation by ambient air particulate matter exposure may exacerbate non-alcoholic fatty liver disease. J Immunotoxicol. 2009;6:266–275.
8. Laing S, Wang G, Briazova T, et al. Airborne particulate matter selectively activates endoplasmic reticulum stress response in the lung and liver tissues. Am J Physiol Cell Physiol. 2010;299:C736–C749.
9. Zheng Z, Zhang X, Wang J, et al. Exposure to fine airborne particulate matters induces hepatic fibrosis in murine models. J Hepatol. 2015;63:1397–1404.
10. Fortoul TI, Rodriguez-Lara V, Gonzalez-Villalva A, et al. Vanadium inhalation in a mouse model for the understanding of air-suspended particle systemic repercussion. J Biomed Biotechnol. 2011;2011:951043.
11. Cano-Gutiérrez G, Acevedo-Nava S, Santamaría A, Altamirano-Lozano M, Cano-Rodríguez MC, Fortoul TI. Hepatic megalocytosis due to

vanadium inhalation: participation of oxidative stress. Toxicol Ind Health. 2012;28:353–360.
12. Cesaroni G, Badaloni C, Gariazzo C, et al. Long-term exposure to urban air pollution and mortality in a cohort of more than a million adults in Rome. Environ Health Perspect. 2013;121:324–331.
13. Nehra MS, Ma Y, Clark C, Amarasingham R, Rockey DC, Singal AG. Use of administrative claims data for identifying patients with cirrhosis. J Clin Gastroenterol. 2013;47:e50–e54.
14. Diez Roux AV, Merkin SS, Hannan P, Jacobs DR, Kiefe CI. Area characteristics, individual-level socioeconomic indicators, and smoking in young adults: the coronary artery disease risk development in young adults study. Am J Epidemiol. 2003;157:315–326.
15. Beelen R, Hoek G, Vienneau D, et al. Development of NO2 and NOx land use regression models for estimating air pollution exposure in 36 study areas in Europe—The ESCAPE project. Atmos Environ. 2013;72:10–23.
16. Eeftens M, Beelen R, de Hoogh K, et al. Development of land use regression models for PM(2.5), PM(2.5) absorbance, PM(10) and PM(coarse) in 20 European study areas; results of the ESCAPE project. Environ Sci Technol. 2012;46:11195–11205.
17. de Hoogh K, Wang M, Adam M, et al. Development of land use regression models for particle composition in twenty study areas in Europe. Environ Sci Technol. 2013;47:5778–5786.
18. Eeftens M, Tsai MY, Ampe C, et al. Spatial variation of PM2.5, PM10, PM2.5 absorbance and PMcoarse concentrations between and within 20 European study areas and the relationship with NO2—results of the ESCAPE project. Atmos Environ. 2012;62:303–317.
19. Cyrys J, Eeftens M, Heinrich J, et al. Variation of NO2 and NOx concentrations between and within 36 European study areas: results from the ESCAPE study. Atmos Environ. 2012;62:374–390.
20. Badaloni C, Cesaroni G, Cerza F, Davoli M, Brunekreef B, Forastiere F. Effects of long-term exposure to particulate matter and metal components on mortality in the Rome longitudinal study. Environ Int. 2017;109:146–154.
21. Eisen EA, Agalliu I, Thurston SW, Coull BA, Checkoway H. Smoothing in occupational cohort studies: an illustration based on penalised splines. Occup Environ Med. 2004;61:854–860.
22. VanderWeele TJ, Ding P. Sensitivity analysis in observational research: introducing the E-value. Ann Intern Med. 2017;167:268–274.
23. Ratib S, West J, Crooks CJ, Fleming KM. Diagnosis of liver cirrhosis in England, a cohort study, 1998–2009: a comparison with cancer. Am J Gastroenterol. 2014;109:190–198.
24. Vaz J, Eriksson B, Strömberg U, Buchebner D, Midlöv P. Incidence, aetiology and related comorbidities of cirrhosis: a Swedish population-based cohort study. BMC Gastroenterol. 2020;20:84.
25. Pimpin L, Cortez-Pinto H, Negro F, et al; EASL HEPAHEALTH Steering Committee. Burden of liver disease in Europe: epidemiology and analysis of risk factors to identify prevention policies. J Hepatol. 2018;69:718–735.
26. Hsieh S, Leaderer BP, Feldstein AE, et al. Traffic-related air pollution associations with cytokeratin-18, a marker of hepatocellular apoptosis, in an overweight and obese paediatric population. Pediatr Obes. 2018;13:1–6.
27. Deng H, Eckel SP, Liu L, Lurmann FW, Cockburn MG, Gilliland FD. Particulate matter air pollution and liver cancer survival. Int J Cancer. 2017;141:744–749.
28. Pedersen M, Andersen ZJ, Stafoggia M, et al. Ambient air pollution and primary liver cancer incidence in four European cohorts within the ESCAPE project. Environ Res. 2017;154:226–233.
29. Altamirano J, Bataller R. Cigarette smoking and chronic liver diseases. Gut. 2010;59:1159–1162.
30. Dam MK, Flensborg-Madsen T, Eliasen M, Becker U, Tolstrup JS. Smoking and risk of liver cirrhosis: a population-based cohort study. Scand J Gastroenterol. 2013;48:585–591.
31. Kim JW, Park S, Lim CW, Lee K, Kim B. The role of air pollutants in initiating liver disease. Toxicol Res. 2014;30:65–70.
32. Abdrabouh AE. Liver disorders related to exposure to gasoline fumes in male rats and role of fenugreek seed supplementation. Environ Sci Pollut Res Int. 2019;26:8949–8957.
33. Zheng Z, Xu X, Zhang X, et al. Exposure to ambient particulate matter induces a NASH-like phenotype and impairs hepatic glucose metabolism in an animal model. J Hepatol. 2013;58:148–154.
34. Orioli R, Cremona G, Ciancarella L, Solimini AG. Association between PM10, PM2.5, $NO_2$, $O_3$ and self-reported diabetes in Italy: a cross-sectional, ecological study. PLoS One. 2018;13:e0191112.
35. Pinault LL, Weichenthal S, Crouse DL, et al. Associations between fine particulate matter and mortality in the 2001 Canadian Census Health and Environment Cohort. Environ Res. 2017;159:406–415.
36. Cesaroni G, Badaloni C, Romano V, Donato E, Perucci CA, Forastiere F. Socioeconomic position and health status of people who live near busy roads: the Rome Longitudinal Study (RoLS). Environ Health. 2010;9:41.
37. Cesaroni G, Porta D, Badaloni C, et al. Nitrogen dioxide levels estimated from land use regression models several years apart and association with mortality in a large cohort study. Environ Health. 2012;11:48.
38. Wang R, Henderson SB, Sbihi H, Allen RW, Brauer M. Temporal stability of land use regression models for traffic-related air pollution. Atmos Environ. 2013;64:312–319.