Exposure to Particulate Air Pollution and Risk of Deep Vein Thrombosis

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Background: Particulate air pollution has been linked to heart disease and stroke, possibly resulting from enhanced coagulation and arterial thrombosis. Whether particulate air pollution exposure is related to venous thrombosis is unknown.

Methods: We examined the association of exposure to particulate matter of less than 10 µm in aerodynamic diameter (PM_{10}) with deep vein thrombosis (DVT) risk in 870 patients and 1210 controls from the Lombardy region in Italy, who were examined between 1995 and 2005. We estimated exposure to PM_{10} in the year before DVT diagnosis (cases) or examination (controls) through area-specific mean levels obtained from ambient monitors.

Results: Higher mean PM_{10} level in the year before the examination was associated with shortened prothrombin time (PT) in DVT cases (standardized regression coefficient [β] = –0.12; 95% confidence interval [CI], –0.23 to 0.00) (P = .04) and controls (β = –0.06; 95% CI, –0.11 to 0.00) (P = .04). Each increase of 10 µg/m^3 in PM_{10} was associated with a 70% increase in DVT risk (odds ratio [OR], 1.70; 95% CI, 1.30 to 2.23) (P < .001) in models adjusting for clinical and environmental covariates. The exposure-response relationship was approximately linear over the observed PM_{10} range. The association between PM_{10} level and DVT risk was weaker in women (OR, 1.40; 95% CI, 1.02 to 1.92) (P = .02 for the interaction between PM_{10} and sex), particularly in those using oral contraceptives or hormone therapy (OR, 0.97; 95% CI, 0.58 to 1.61) (P = .048 for the interaction between PM_{10} level and hormone use).

Conclusions: Long-term exposure to particulate air pollution is associated with altered coagulation function and DVT risk. Other risk factors for DVT may modulate the effect of particulate air pollution.

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EXPOSURE TO PARTICULATE AIR pollution has been associated with increased short- and long-term morbidity and mortality from heart disease and stroke.[1-4] Hypercoagulability and enhanced thrombosis have been indicated as one mechanistic pathway that mediates such effects,[4,5] since higher plasma levels of coagulation proteins such as factor VIII, von Willebrand factor, and fibrinogen have been associated with the exposure.[5-7] Recently, changes in coagulation function resulting in shortened prothrombin time (PT) have been observed in association with higher mean level of particulate air pollution of less than 10 µm in aerodynamic diameter (PM_{10}) in the 30 days before the examination, suggesting that extended PM_{10} exposure may cause effects on blood clotting.[8]

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Procoagulant abnormalities are stronger determinants of venous than of arterial thrombosis,[9] and increased risk of deep vein thrombosis (DVT) has been associated with a series of heritable or acquired risk factors that cause hypercoagulability, including factor V Leiden and G20210A prothrombin mutations, deficiencies of the natural anticoagulant proteins antithrombin, protein C and protein S, and use of oral contraceptive or hormone therapy.[10,11] In rat and hamster models developed to investigate mechanisms involved in arterial thrombosis, inhalation or intravenous administration of air pollution constituents—such as diesel exhausts and ultrafine particles—induces thrombosis of the femoral[6] and ear veins.[12] In human subjects, to our knowledge, no evidence is currently available relating air pollution exposure to DVT. In the present study, we investigated whether long-term ambient PM_{10} exposure was associated with increased thrombotic susceptibility and higher DVT risk in a large epidemiologic investigation conducted in the Lombardy region in northern Italy.

For editorial comment see page 909

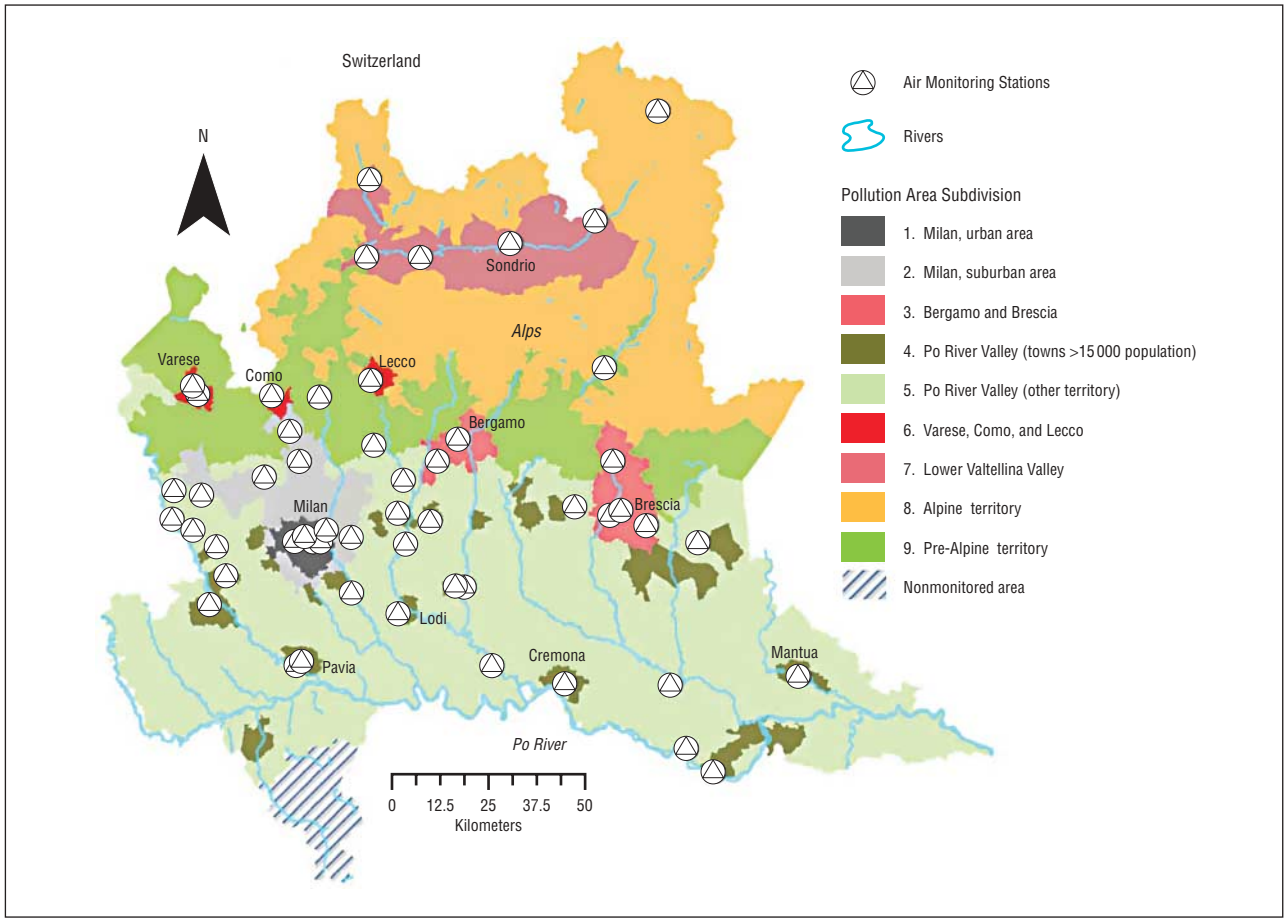

Figure 1. Map of the Lombardy region (Italy) showing the location of the 53 air pollution monitors in the 9 areas identified for the study.

METHODS

STUDY POPULATION

The cases included 871 patients (420 men and 451 women), who were Lombardy region residents and had been diagnosed between January 1995 through September 2005 as having lower limb DVT with or without symptomatic pulmonary embolism. Cases were referred to the Thrombosis Center, University of Milan, and IRCCS (Istituto di Ricovero e Cura a Carattere Scientifico) Maggiore Hospital, Milan, Italy, for a thrombophilia screening and were asked to bring all available clinical documentation. From this documentation, DVT was diagnosed based on evidence from objective methods such as compression ultrasonography or venography. Cases were asked about the presence of transient DVT risk factors in the month preceding the event, including surgery, trauma, immobilization (≥10 days), use of oral contraceptives or hormone therapy, and pregnancy or puerperium (6-week period after delivery). In 170 cases (19.5%), DVT was complicated by symptomatic pulmonary embolism. A total of 761 cases (84.4%) had a first DVT during the study period, whereas 110 (12.6%) had recurrent DVT. Cases with recurrent DVT had not been included in the study after their first diagnosis, since they were recruited after their first visit at the Thrombosis Center.

The controls included 1210 healthy Lombardy region residents (490 men and 720 women), who volunteered to be investigated at the Thrombosis Center in the same study period as the cases. The controls were recruited by asking each of the cases for a list of friends or nonblood relatives, and they were randomly selected from these lists using an algorithm that balanced the age distribution of the controls to that of the cases. To increase power to evaluate a potential effect modification by use of oral contraceptives and hormone therapy, female controls were recruited in excess of female cases. Thrombosis was excluded in controls with a structured questionnaire validated for the retrospective diagnosis of venous thromboembolism. Information on clinical, lifestyle, and reproductive factors was collected from all participants during an in-person interview. For practical reasons, because most of the interviews took place while the patients with DVT were in the hospital, interviewers could not be blinded to the case status. However, the subjects' addresses were confirmed on administrative or other official records, and exposure was assigned, based on the address, by a statistician blinded to the case status. Participants' written informed consents and local institutional review board approvals were obtained before the study. Prothrombin time and activated partial thromboplastin time (aPTT) were measured using laboratory methods previously described.[13] Patients with DVT, if receiving anticoagulant drugs, were not asked to stop the treatment.

AIR POLLUTION AND WEATHER DATA

Methods for exposure assignment were previously described in detail.[8,14] Briefly, for each of the 9 geographic areas shown in **Figure 1**, mean concentrations of PM_{10} were computed using data from monitors located at 53 different sites throughout the Lombardy region. All participants were assigned to 1 of 9 geographic areas based on their residence at the time of the study. In our statistical analyses, PM_{10} level was evaluated using ambient PM_{10} concentrations averaged over the 365 days preceding

Table 1. Characteristics of Deep Vein Thrombosis in Cases and Controls

Characteristic	Cases, No. (%) (n=871)[f]	Controls, No. (%) (n=1210)[f]
Sex		
Male	420 (48.2)	490 (40.5)
Female	451 (51.8)	720 (59.5)
Age, y		
18-35	303 (34.8)	373 (30.8)
36-50	275 (31.6)	424 (35.0)
51-84	293 (33.6)	413 (34.1)
Area of residence		
Milan, urban area	381 (43.7)	525 (43.4)
Milan, suburban area	201 (23.1)	272 (22.5)
Bergamo and Brescia	8 (0.9)	18 (1.5)
Po River Valley (towns >15 000 population)	47 (5.4)	72 (6.0)
Po River Valley (other territory)	119 (13.7)	152 (12.6)
Varese, Como, and Lecco	26 (3.0)	24 (2.0)
Lower Valtellina Valley	14 (1.6)	29 (2.4)
Alpine territory	10 (1.2)	15 (1.2)
Pre-Alpine territory	65 (7.5)	103 (8.5)
Body mass index[a]		
13.3-22.0	195 (23.6)	396 (32.9)
22.1-24.9	202 (24.5)	399 (33.1)
25.0-53.3	428 (51.9)	409 (34.0)
Education		
Elementary/middle school	288 (34.4)	343 (28.6)
High school	384 (45.9)	579 (48.3)
College	165 (19.7)	278 (23.2)
Premenopausal women with current use of oral contraceptives[b]		
No	117 (39.3)	267 (71.4)
Yes	181 (60.7)	107 (28.6)
Postmenopausal women with current use of hormone therapy[b]		
No	112 (73.7)	255 (81.0)
Yes	40 (26.3)	60 (19.0)
Current use of oral contraceptives or hormone therapy[b]		
No	229 (50.9)	522 (75.8)
Yes	221 (49.1)	167 (24.2)
Factor V Leiden[c]		
–/–	721 (82.8)	1180 (97.5)
–/+	140 (16.1)	30 (2.5)
+/+	10 (1.2)	0
G20210A prothrombin mutation[c]		
–/–	767 (88.1)	1166 (96.4)
–/+	99 (11.4)	44 (3.6)
+/+	5 (0.6)	0
Factor V Leiden or G20210A prothrombin mutation		
No mutation	638 (73.3)	1137 (94.0)
Any mutation	233 (26.8)	73 (6.0)
Deficiencies of natural anticoagulant proteins		
No	809 (92.9)	1185 (97.9)
Yes	62 (7.1)	25 (2.1)
Hyperhomocysteinemia		
No	738 (84.7)	1104 (91.2)
Yes	133 (15.3)	106 (8.8)
Any cause of thrombophilia[d]		
No	364 (41.8)	877 (73.5)
Yes	507 (58.2)	333 (27.5)
Year of study[e]		
1995-1997	253 (29.0)	471 (38.9)
1998-2000	308 (35.4)	384 (31.7)
2001-2005	310 (35.6)	355 (29.3)
Season at diagnosis (cases) or examination (controls)		
December-February	227 (26.1)	282 (23.3)
March-May	206 (23.7)	309 (25.5)
June-August	228 (26.2)	216 (17.9)
September-November	210 (24.1)	403 (33.3)
Mean temperature on the day of diagnosis (cases) or examination (controls)		
–6.1°C to 8.4°C	277 (31.8)	403 (33.3)
8.4°C to 16.5°C	229 (26.3)	403 (33.3)
16.5°C to 32.7°C	365 (41.9)	404 (33.4)

[a] Calculated as weight in kilograms divided by height in meters squared.
[b] Only female subjects are shown.
[c] The minus sign indicates a normal allele; the plus sign indicates a mutated allele. Each subject may carry 0, 1, or 2 mutations.
[d] Subjects with deficiencies of the natural anticoagulant proteins, factor V Leiden, G20210A prothrombin mutation, or hyperhomocysteinemia.
[e] Year of deep vein thrombosis diagnosis for cases or date of examination for controls.
[f] Some totals do not match the total number of subjects because values were missing in some categories.

the index date, also taking into account changes of residence during the same time window. Index dates were the date of diagnosis for DVT cases and the date of the examination for controls.

STATISTICAL ANALYSIS

In previous work, we tested the association of shorter-term PM_{10} exposure with PT and aPTT in the same healthy subjects who served as controls in the present analysis.[13] In the present study, to evaluate the association between PM_{10} and PT and aPTT between DVT cases or controls, we used the same statistical methods we previously described.[13]

Differences between cases and controls after stratification by geographic area or study period were tested using the Mantel-Haenszel method. The association of PM_{10} level with DVT risk was tested in a case-control analysis using unconditional logistic regression models including, as independent variables, sex, area of residence, education (elementary/middle school, high school, college), factor V Leiden or G20210A prothrombin mutation (yes/no), and current use of oral contraceptives or hormone therapy (yes/no). Variables with potentially nonlinear associations with risk, including age, body mass index, day of the year (for seasonality), index date (for long-term time trends), and ambient temperature, were controlled using penalized regression splines with 4 *df* for each variable.[15] Unconditional regression was performed to allow for the use of penalized splines, which are not available for use in conditional logistic models in available software packages, as well as to avoid the loss from the analyses of subjects in incomplete sets due to participation refusal or missing exposure or covariate information. As approximation of the relative risk of DVT, we reported odds ratios (ORs) and 95% confidence intervals (CIs) for each increase of 10 $\mu g/m^3$ in the mean level of PM_{10}. We used Stata version 9.0 (StataCorp, College Station, Texas) for descriptive analyses and R version 2.2.0 (R Project for Statis-

Table 2. Estimated Changes of PT and aPTT Associated With PM_{10} Levels in the Year Before the Examination in DVT Cases and Controls

	PM_{10} Association With PT		PM_{10} Association With aPTT	
Case Status	β Coefficient[a] (95% CI)	*P* Value	β Coefficient[a] (95% CI)	*P* Value
Controls	−0.12 (−0.23 to 0.00)	.04	−0.09 (−0.19 to 0.01)	.07
DVT cases	−0.06 (−0.11 to 0.00)	.04	0.01 (−0.03 to 0.04)	.78

Abbreviations: aPTT, activated partial thromboplastin time; CI, confidence interval; DVT, deep vein thrombosis; PM_{10} particulate matter of less than 10 μm in aerodynamic diameter; PT, prothrombin time.

[a]Standardized regression coefficients, expressing the fraction of a standard deviation change in PT or aPTT associated with a standard deviation change in exposure, adjusted for age, sex, body mass index, cigarette smoking, alcohol consumption, oral contraceptives, and nonlinear terms for seasonality, long-term time trend, and temperature.

tical Computing, Vienna, Austria) to fit regression models. All statistical tests were 2 sided, and $P<.05$ was considered statistically significant.

RESULTS

CHARACTERISTICS OF THE STUDY POPULATION

Table 1 gives the characteristics of the 871 DVT cases (420 men and 451 women) and 1210 controls (490 men and 720 women). Cases and controls had similar distributions by age ($P=.12$) and area of residence ($P=.59$). Cases had higher body mass index ($P<.001$); lower education ($P=.01$); more frequent use of oral contraceptives or hormone therapy ($P<.001$); and higher prevalences of factor V Leiden ($P<.001$), G20210 prothrombin mutation ($P<.001$), inherited deficiencies of natural anticoagulant proteins ($P<.001$), hyperhomocysteinemia ($P<.001$), and any of the causes of thrombophilia ($P<.001$). Controls were less likely to be examined for the study during the summer and their examinations were more frequent in the fall, whereas no major seasonal pattern was found for DVT diagnoses ($P<.001$). Mean ambient temperatures were higher on the days of diagnosis of DVT cases than on the days of examination of controls ($P<.001$). A larger proportion of controls were entered earlier in the study compared with cases ($P<.001$). This difference in recruitment was accounted for by performing analyses stratified by year and through the use of nonlinear terms for long-time trends in multivariable models.

EFFECTS OF LONG-TERM PM_{10} EXPOSURE ON COAGULATION TIMES

Mean PM_{10} level over the 1 year before the examination was significantly associated with shortened PT in both cases (standardized regression coefficient [β] = −0.12; 95% CI, −0.23 to 0.00) ($P=.04$) and controls (β = −0.06; 95% CI −0.11 to 0.00) ($P=.04$). While the negative effects of mean PM_{10} levels on PT showed no major differences across means taken over 30, 60, 90, 180, or 270 days or 1 year (data not shown), the 1-year mean PM_{10} level was the only time window significantly associated with shortened PT among cases. A nonsignificant aPTT shortening was observed in association with the 1-year mean PM_{10} level among controls (β = −0.09; 95% CI, −0.19 to 0.01) ($P=.07$) but not in cases (β = 0.01; 95% CI, −0.03 to 0.04) ($P=.78$) (**Table 2**).

PARTICLE EXPOSURE AND RELATIVE RISK OF DVT

Table 3 presents the tertiles of the mean PM_{10} level measured in the area of residence during the year before the date of DVT diagnosis (cases) or date of examination (controls), according to their area of residence and year of study. In both cases and controls, PM_{10} level was highest in the Milan urban and suburban areas ($P<.001$). Subjects from the Alpine territory and the lower Valtellina Valley were all in the lowest tertile of PM_{10} exposure. In both cases and controls, the frequency of subjects being in the highest tertile of PM_{10} exposure was highest in the earlier years of the study, and it decreased throughout the study period, to reach the lowest frequency in the most recent years ($P<.001$).

The DVT cases tended to have higher PM_{10} levels than the controls, and this pattern was more evident after the data were stratified by year of the study (Table 3). The test for the difference between cases and controls by tertile of PM_{10} level was significant in the analysis stratified by year of the study ($P=.002$, Mantel-Haenszel test). Such a difference was not statistically significant without stratification by year of the study ($P=.44$, Fisher exact test), indicating that inequalities in the distribution of cases and controls by study period, if not controlled, would have biased the results.

We estimated the relative risk of DVT associated with PM_{10} level in a multivariable model controlled for age, sex, year of diagnosis, area of residence, body mass index, education, current use of oral contraceptives or hormone therapy, Leiden V or prothrombin mutations, season, and ambient temperature. In this model, an increase of 10 $\mu g/m^3$ in PM_{10} level was associated with an adjusted OR of 1.70 (95% CI, 1.30 to 2.23) ($P<.001$). The increase in DVT risk was nearly linear across the range of exposure concentrations that were measured (**Figure 2**).

We also examined DVT risk in association with different exposure time windows using PM_{10} level averaged over 90 days in the 2 years before diagnosis (see supplementary Figure 1 and supplementary Table 1; available at: http:

Table 3. Tertile of PM_{10} Exposure in DVT Cases and Controls by Area of Residence and Year of Examination[a]

	Tertile of PM_{10} Exposure,[a] µg/m³					
	Cases			Controls		
	12.0-44.2	44.3-48.1	48.2-51.5	12.0-44.2	44.3-48.1	48.2-51.5
Area of residence, No. (%) of subjects						
Milan, urban area	72 (18.9)	156 (40.9)	153 (40.2)	70 (13.3)	265 (50.5)	190 (36.2)
Milan, suburban area	47 (23.4)	68 (33.8)	86 (42.8)	70 (25.7)	80 (29.4)	122 (44.9)
Bergamo and Brescia	3 (37.5)	1 (12.5)	4 (50.0)	10 (55.6)	3 (16.7)	5 (27.8)
Po River Valley (towns >15 000 population)	16 (34.0)	12 (25.5)	19 (40.4)	33 (45.8)	12 (16.7)	27 (37.5)
Po River Valley (other territory)	42 (35.3)	28 (23.5)	49 (41.2)	62 (40.8)	35 (23.0)	55 (36.2)
Varese, Como, and Lecco	24 (92.3)	1 (3.9)	1 (3.9)	20 (83.3)	1 (4.2)	3 (12.5)
Lower Valtellina Valley	14 (100)	0	0	29 (100)	0	0
Alpine territory	10 (100)	0	0	15 (100)	0	0
Pre-Alpine territory	56 (86.2)	9 (13.9)	0	94 (91.3)	9 (8.7)	0
Year of study, No. (%) of subjects[b]						
1995-1997	27 (10.7)	66 (26.1)	160 (63.2)	59 (12.5)	185 (39.3)	227 (48.2)
1998-2000	81 (26.3)	124 (40.3)	103 (33.4)	121 (31.5)	150 (39.1)	113 (29.4)
2001-2005	176 (56.8)	85 (27.4)	49 (15.8)	223 (62.8)	70 (19.7)	62 (17.5)

Abbreviations: DVT, deep vein thrombosis; PM_{10} particulate matter of less than 10 µm in aerodynamic diameter.
[a] Tertile of PM_{10} level in control subjects during the year before the examination.
[b] Year of DVT diagnosis for cases or date of examination for controls.

//www.cdldevoto.it/didattic/materiale/Appendix_Archives_online.pdf). Risk of DVT increased progressively with the duration of the time window evaluated and was statistically significant in association with the 270-day, 1-year, and 2-year PM_{10} mean levels.

SUSCEPTIBILITY TO AIR POLLUTION EFFECTS

Differences in the relationship between DVT risk and PM_{10} level according to the characteristics of the study subjects are summarized in **Table 4**. The association between PM_{10} level and DVT was significantly attenuated in female subjects ($P=.02$ for the interaction between PM_{10} level and sex). An increase of 10 µg/m³ in PM_{10} level was associated with an adjusted OR for DVT of 2.07 (95% CI, 1.50 to 2.84) ($P<.001$) in men and 1.40 (95% CI, 1.02 to 1.92) ($P=.04$) in women. No PM_{10} effect was observed in women taking oral contraceptives or hormone therapy (OR, 0.97; 95% CI, 0.58 to 1.61) ($P=.89$ for PM_{10} effect; $P=.048$ for the interaction between PM_{10} and hormone use). The other characteristics evaluated, including year of diagnosis, did not significantly modify the association between PM_{10} exposure and DVT risk (Table 4).

SENSITIVITY ANALYSES

We repeated all the analyses after excluding cases with a recurrent (nonfirst) episode of DVT (n=110). Risk estimates were very similar to those for the entire study population. Each increase of 10 µg/m³ in PM_{10} level was associated with an OR of 1.67 (95% CI, 1.27 to 2.22) ($P<.001$), adjusting for multiple variables. In the subsample of 760 cases with a single episode of DVT, the variations in the association between PM_{10} level and the risk of DVT due to demographic characteristics, presence of thrombophilia, or use of hormone therapies were similar to those observed in the entire study population.

To evaluate the influence of splines selection in fitting nonliner terms in the logistic models, we repeated all statistical analyses by using natural splines instead of penalized splines. The use of natural splines did not modify the statistical significance of the results, with only small changes in the risk estimates.

We also evaluated the influence of different methods for adjusting for long-term time trends during the study period (supplementary Table 2; available at: http://www.cdldevoto.it/didattic/materiale/Appendix_Archives_online.pdf). As was shown in the subsection "Particle Exposure and Relative Risk of DVT," ignoring long-term time trends in the analyses would have almost completely obscured the association between PM_{10} level and DVT risk. However, all of the methods for adjustment for long-term time trends that we evaluated in our sensitivity analysis (fitting dummy variables for each year of the study period, as well as linear terms or penalized splines for index date with degrees of freedom varying between 2 and 8) produced risk estimates indicating a significant association between PM_{10} level and DVT risk, with only small changes due to different handling of the time trends for most of the methods. However, it is worth noting that using only a linear variable for the long-term trend would have produced a lower OR, likely reflecting less than optimal fitting of the time relationships present in our data.

COMMENT

In this study of DVT cases and healthy controls, exposure to increased concentrations of particulate air pollution in the year before diagnosis was associated with increased DVT risk after controlling for clinical and environmental covariates. Mean level of PM_{10} before the examination was also correlated with shorter PT in both

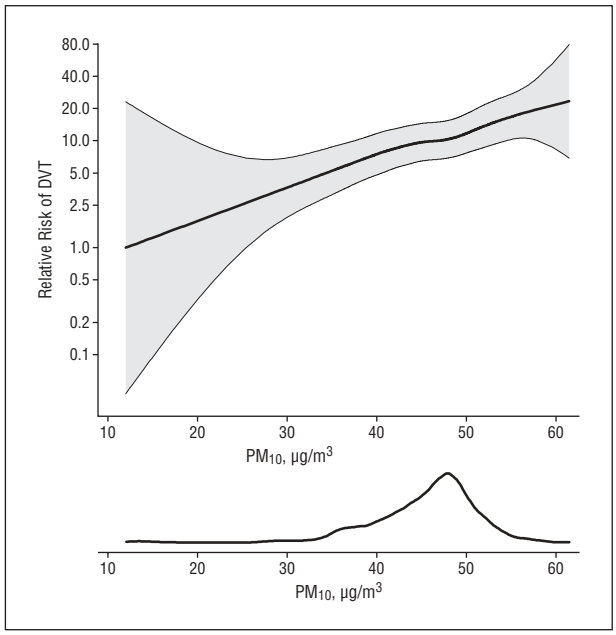

Figure 2. Level of exposure to fine particulate matter and the risk of deep vein thrombosis (DVT). The graph demonstrates the observed relationship between the relative risk of DVT and the level of particulate matter of less than 10 µm in aerodynamic diameter (PM_{10}) in the year preceding the diagnosis. These results suggest a linear relationship between exposure and risk, though the 95% confidence intervals (shaded areas) are wide at the extremes of exposure. Risk is depicted in comparison with a reference value of 12.0 µg/m^3 (minimum observed PM_{10} level). The histogram in the bottom part illustrates the density of exposure distribution for air pollution. Risk estimates are adjusted for age, sex, year of diagnosis, area of residence, body mass index, education, current use of oral contraceptives or hormone therapy, Leiden V or prothrombin mutations, season, and ambient temperature.

cases and controls. The DVT risk increase associated with PM_{10} level was smaller in women and limited to those who were not using oral contraceptives or hormone therapy at the time of diagnosis.

Long-term exposure to particulate air pollution has been associated with increased risk of coronary heart and cerebrovascular disease in multiple investigations conducted in several countries.[4] A systemic increase in thrombotic tendency, secondary to the induction of inflammatory mediators produced in the lungs and released in the circulation or to the translocation of particles of smaller diameter from the lungs into the circulation[6] has been frequently proposed to account for the cardiac and cerebrovascular effects of particulate air pollution. In contrast, venous thrombosis has received little attention in studies on the cardiovascular outcomes of air pollution. In a time-series analysis from the Netherlands, Hoek et al[16] reported an association of short-term exposure to ambient ozone and, to a lesser extent, black smoke and PM_{10}, with increased mortality from embolism and thrombosis, a broad category that included arterial and venous thromboses in various sites. To date, no study has specifically addressed the association between particulate air pollution and DVT. In our population, we estimated an overall 70% increase in DVT risk with each increase of 10 µg/m^3 in PM_{10} level during the year before diagnosis. For comparison, in the Harvard Six Cities Study, the risk of death from cardiopulmonary diseases was 37% higher in the most polluted compared with the least polluted

Table 4. Relative Risk[a] of Deep Vein Thrombosis (DVT) Associated With a 10 µg/m^3 Increase in PM_{10} in the Year Preceding the Diagnosis by Subjects' Characteristics

Characteristic	OR (95% CI)	*P* Value[b]	*P* Value for Interaction[c]
All subjects	1.70 (1.30-2.23)	.001	. . .
Sex			
Male	2.07 (1.50-2.84)	<.001	.02
Female	1.40 (1.02-1.92)	.04	
Age, y			
18-35	1.57 (1.11-2.24)	.01	.99
36-50	1.97 (1.41-2.77)	<.001	
51-84	1.54 (0.90-2.63)	.12	
Premenopausal women with current use of oral contraceptives			
No	1.53 (0.86-2.72)	.14	.11
Yes	0.87 (0.46-1.67)	.68	
Postmenopausal women with current use of hormone therapy			
No	1.60 (0.72-3.54)	.24	.27
Yes	0.85 (0.29-2.45)	.76	
Current use of oral contraceptives or hormone therapy			
No	1.64 (1.05-2.57)	.03	.048
Yes	0.97 (0.58-1.61)	.89	
Body mass index[d]			
13.3-22.0	1.47 (0.97-2.23)	.07	.37
22.1-24.9	1.72 (1.17-2.54)	.006	
25.0-53.3	1.83 (1.03-3.24)	.04	
Education			
Elementary/middle school	1.93 (1.35-2.76)	<.001	.21
High school	1.72 (1.24-2.39)	.001	
College	1.35 (0.74-2.45)	.33	
Deficiencies of natural anticoagulant proteins			
None	1.66 (1.26-2.18)	<.001	.41
Any	2.56 (0.91-7.18)	.07	
Factor V Leiden or G20210A prothrombin mutation			
None	1.69 (1.27-2.23)	<.001	.83
Any	1.79 (1.05-3.05)	.03	
Hyperhomocysteinemia			
No	1.66 (1.26-2.19)	<.001	.25
Yes	2.19 (1.33-3.61)	.002	
Any cause of thrombophilia[e]			
No	1.59 (1.19-2.13)	.002	.27
Yes	1.96 (1.34-2.87)	<.001	
Year of diagnosis			
1995-1997	1.61 (1.06-2.46)	.03	.12
1998-2000	1.34 (0.90-1.99)	.15	
2001-2005	2.14 (1.04-4.39)	.04	

Abbreviations: CI, confidence interval; OR, odds ratio; PM_{10} particulate matter of less than 10 µm in aerodynamic diameter.

[a] Odds ratios and 95% CIs were adjusted for age, sex, year of diagnosis, area of residence, body mass index, education, current use of oral contraceptives or hormone therapy, factor V Leiden or prothrombin mutations, season, and ambient temperature.

[b] Test for the association of PM_{10} with DVT risk.

[c] Test for differences by subjects' characteristics of the association between PM_{10} and DVT.

[d] Body mass index (calculated as weight in kilograms divided by height in meters squared) was categorized according to controls' tertiles.

[e] Subjects with deficiencies of the natural anticoagulant proteins, factor V Leiden, G20210A prothrombin mutation, or hyperhomocysteinemia.

cities.[1] In the Women's Health Initiative Study, an increase of 10 μg/m^3 of annual mean concentrations of $PM_{2.5}$, which is considered a stronger predictor than PM_{10} level of air pollution effects, was associated with a 24% increase in the risk of cardiovascular events and a 76% increase in the risk of death from cardiovascular disease.[2] The estimated increase in risk of death from all cardiovascular causes associated with 10 μg/m^3 elevation in long-term $PM_{2.5}$ level was 19% in the Harvard Six Cities study and 13% in the study by the American Cancer Society.[3]

In the present study, PM_{10} exposure did not increase the risk of DVT in women as much as in men. By evaluating additional risk factors, we found that part—if not all—of such risk attenuation was due to the lack of association between PM_{10} level and the risk of DVT among women using oral contraceptives or hormone therapy. Such hormone therapies are independent risk factors for DVT,[10] which is also confirmed in this study by the higher prevalence of oral contraceptive and hormone use in the cases compared with the controls. Use of oral contraceptives and hormone therapy induces changes in coagulation factors, such as increased levels of the procoagulant factors VII, IX, X, XII, and XIII, von Willebrand factor, and fibrinogen and reduced levels of the anticoagulant proteins antithrombin and protein S,[13,17,18] that are similar in characteristics and degree to the coagulation changes observed after exposure to air particles.[7,8,19-22] We surmise that prothrombotic mechanisms are already activated in those receiving hormone therapy so that they undergo less or no further induction after air particle exposure.

In our analyses, we evaluated DVT risk in association with the level of PM_{10} measured during the year before diagnosis. In this study, the use of short-term (hourly or daily) air pollution levels would not have been appropriate because DVT presentation is often subtle and its diagnosis has been shown to lag as long as 4 weeks after the initial DVT symptoms.[23] In the present work, we demonstrated that mean PM_{10} level in the year before the examination was associated with shortened PT, extending our previous observation of an association with shorter exposure time windows.[13] Interestingly, while the negative effects of mean PM_{10} levels on PT were independent of the time-window selected, the 1-year mean PM_{10} was the only exposure metric significantly associated with shortened PT among the cases. In addition, while in our previous work we could not find any relation between 30-day mean PM_{10} level and aPTT, in our present study, a nonsignificant (P=.07) association was observed between aPTT shortening and 1-year mean PM_{10} among the controls. This association, taken together with the similar PT change, suggests that 1-year PM_{10} exposure may also affect aPTT. Thus, the use of PM_{10} level in the year before diagnosis appeared to capture a fuller range of prothrombotic effects, while also reducing the risks of confounding by seasonal patterns and ambient temperature. This study has the advantage of being based on a large number of DVT cases and controls recruited using a standardized protocol over a long time span. Cases had objective diagnoses of DVT, and both cases and controls were well characterized for inherited and acquired risk factors for DVT. In the statistical analysis, we used models that included nonlinear regression terms to adjust for long-term time trends and day of the year (thus controlling for confounding from year of the study and seasonal variations), in addition to age, sex, area of residence, education, factor V Leiden, G20210A prothrombin mutation, use of oral contraceptive or hormone therapy, body mass index, and ambient temperature.

Because the healthy controls were selected among nonblood relatives and friends of the DVT cases, they tended to be distributed in the 9 study areas, with proportions that were very similar to those of the DVT cases. This might have generated overmatching in our study, ie, the exposure levels of controls may have been more similar to those of DVT cases than they actually are in the population at risk. Therefore, it is possible that risk estimates were underestimated in our study. A limitation of this study is that we used ambient air pollution estimated at the subjects' address as a surrogate for personal exposure, which may have resulted in measurement error, since most subjects conduct a large part of their daily activities away from their residence. A detailed questionnaire was used to ascertain known risk factors for DVT, but no information was collected concerning daily activities, such as time spent outside or in traffic, that could have refined the assessment of PM_{10} exposure. In addition, our exposure assessment was done by dividing the Lombardy region into 9 areas for which mean PM_{10} levels were assigned by averaging measurements from multiple monitors. Although these areas were selected to group together territories with similar population densities and geographical characteristics, thus likely reducing within-area variations of the exposure, we were not able to obtain PM_{10} level estimates at a smaller scale. However, PM_{10} levels tend to be spatially homogeneous, and a recent study comparing personal exposures with site monitoring in Boston, Massachusetts, reported that monitor readings and personal exposure are highly correlated.[24] In addition, it has been shown that using ambient measures to estimate individual exposure is likely to produce an underestimation of pollution effects[25] rather than causing the increased risk of DVT found in our study population.

In conclusion, this study provides evidence in support of an association of exposure to particulate air pollution with enhanced prothrombotic mechanisms and risk of DVT. Given the magnitude of the observed effects and the widespread diffusion of particulate pollutants, our findings introduce a novel and common risk factor into the pathogenesis of DVT and, at the same time, give further substance to the call for tighter standards and continued efforts aimed at reducing the impact of urban air pollutants on human health.

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