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Short-Term Associations of Nitrogen Dioxide (NO_2) on Mortality in 18 French Cities, 2010–2014

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Abstract: We present an analysis of short-term associations between ambient NO_2 and mortality according to cause, age-group, and period (cold and warm) in 18 areas in metropolitan France for the 2010–2014 period. Associations were estimated in each area using a generalized additive Poisson regression model, and effects were summarized in a meta-analysis. The percentage increase in mortality rate was estimated for a 10 μg m^{-3} increase in the NO_2 level in each area for each complete calendar year and for cold (November to April) and warm periods (May to October) in each year. We found that the NO_2 increase (lag of 0–1 days) was associated with a 0.75% increase of non-accidental mortality for all age-groups (95% confidence interval (CI): (0.4; 1.10)). During the warm period, this NO_2 increase was associated with a 3.07% increase in non-accidental mortality in the ≥75 years old group (95% CI: 1.97; 4.18). This study supports the short-term effects of NO_2 as a proxy of urban traffic pollution on mortality, even for concentrations below the maximum guideline of 40 μg m^{-3} set down by the European Air Quality Standards and the World Health Organization (WHO).

Keywords: air pollution; NO_2; mortality; times series; France

1. Introduction

Nitrogen dioxide (NO_2) is an air pollutant resulting from combustion processes and is mainly emitted by road traffic. In France, annual urban background and near-road traffic concentrations of NO_2 in ambient air have decreased since 2000. However, the maximum concentration set down by European air quality standards for the protection of human health (40 μg m^{-3}) is exceeded every year in several urban areas (near-traffic), and near-traffic levels are twice as high as urban background levels. In 2016, the annual average urban background NO_2 concentration was 19 μg m^{-3}, compared with 39 μg m^{-3} for near-road traffic. Furthermore, numerous episodes of NO_2 pollution are observed every year during the cold period (November to April) because anticyclonic conditions with cold temperatures limit the dispersion of pollutants [1].

Several epidemiological studies have shown short-term associations between exposure to NO_2 and various health outcomes, in particular respiratory and cardiovascular diseases and mortality [2–4]. More specifically, in 2008, associations were found between ambient NO_2 concentrations and mortality in nine areas in France [5]. Moreover, in 2013, epidemiological and toxicological evidence in the Review of evidence on health aspects of air pollution (REVIHAAP) report [3] suggested a short-term causal relationship, particularly for respiratory effects, which prompted the World Health Organization (WHO) to conclude that it was reasonable to consider that NO_2 had direct effects on health. The WHO also considered that there was sufficient evidence to warrant an estimation of the short-term health impact of NO_2 on all-cause mortality and hospitalizations for respiratory diseases. In 2016, the United States Environmental Protection Agency (US-EPA) considered that there was robust evidence for an

association between short-term NO_2 exposure and exacerbation of asthma, and that the findings from recent studies also suggested an effect on cardiovascular health and mortality [6].

In order to estimate the short-term health impact of NO_2, concentration–response functions (CRFs) are necessary. However, available CRFs for NO_2 in Europe have primarily been derived from data from studies conducted outside the continent. The handful of European-based studies providing CRFs include the APHEA [7] and EpiAir [8] projects in Italy and studies in Switzerland [9] and Austria [10]. Moreover, due to having a higher proportion of diesel-fueled vehicles than elsewhere in the world, the specificity of vehicle emissions in France (and indeed Europe) most probably influences the relationship between NO_2 and health effects. This was supported by the meta-analysis by Mills et al. in 2015 [2], which identified strong geographical heterogeneity between the effects estimated for different regions of the world.

All the above elements highlight the relevance of studying the short-term links between exposure to ambient NO_2 and mortality in France in order to update existing CRFs with a view to using them in future health impact assessments (HIA).

Accordingly, in the present study, we conducted a retrospective time series analysis to evaluate the association between NO_2 as one proxy of urban traffic pollution and the risk of daily non-accidental and cardiovascular mortality in 18 cities in metropolitan France over the period 2010–2014.

2. Materials and Methods

2.1. Study Period and Study Area

We conducted a daily time-series study of the relationship between NO_2 exposure and cause-specific mortality in 18 French areas (Figure 1) (accounting for 499 municipalities) from 2010 to 2014 based on the availability of mortality data.

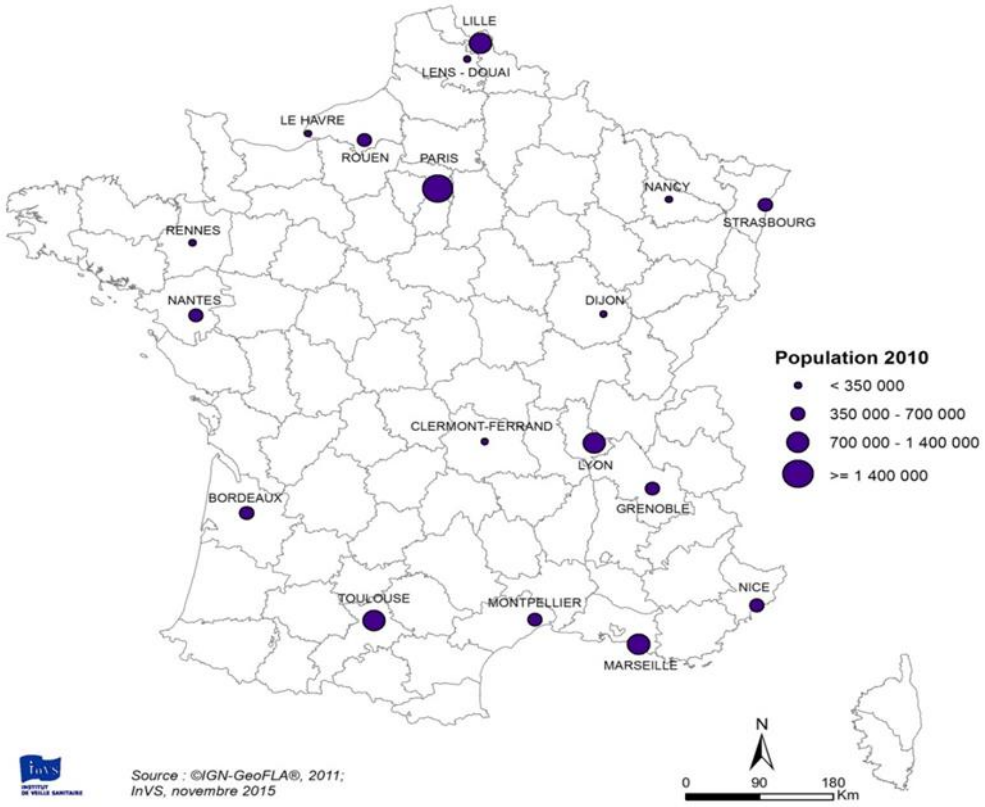

Figure 1. Map of 18 study areas and relative population sizes.

2.2. Data Sources

2.2.1. Mortality Data

For each area, daily mortality data were obtained from the French National Institute of Health and Medical Research (CépiDc) for the study period. Causes of death were coded according to the International Classification of Diseases, 10th Revision (ICD-10). More specifically, data collected included daily counts of total non-accidental (ICD-10: A00-R99), cardiovascular (ICD-10: I00-I99), and respiratory (ICD-10: J00-J99) mortality for all ages. We dichotomized data by age group (≥75 years) to compare with similar studies.

2.2.2. Pollution Data

In each study area, pollution data were obtained from local air quality monitoring networks. NO_2 was measured using the chemiluminescence method, a tapered element oscillating monitor filter dynamic measurement system (TEOM-FDMS) was used for PM_{10}, while ozone (O_3) was measured using the standard reference ultraviolet absorption method. Urban background monitors were used to create daily indicators of NO_2 and PM_{10} exposure, while urban and suburban background monitors were used to create daily indicators of O_3. Depending on the area, between 1 and 15 monitors were available for NO_2, one to seven for PM_{10}, and 1 to 13 for O_3 (Table 1). All study areas were constructed in such a way that the average exposure level of the population could be accurately estimated from the monitoring stations [11]. We built homogeneous study areas because, in time-series studies, the principle of analysis assumes that, on average, all individuals in the population are exposed to the same levels of air pollution each day.

Table 1. Mean levels of air pollution indicators and the number of monitoring stations considered to build the exposure indicator, according to study area: 2010–2014.

Study Area	Daily Mean NO_2 Levels		Daily Mean PM_{10} Levels		Daily Maximum O_3 from 8 h Running Means Levels	
	Mean (Min–Max) (μg m^{-3})	Number of Monitoring Stations	Mean (Min–Max) (μg m^{-3})	Number of Monitoring Stations	Mean (Min–Max) (μg m^{-3})	Number of Monitoring Stations
Bordeaux	19.6 (2.0; 67.9)	3	21.9 (6.2; 88.3)	3	81.3 (3.7; 175.3)	3
Clermont-Ferrand	24.0 (2.1; 104.1)	2	19.9 (2.4; 86.5)	2	72.6 (1.7; 153.5)	2
Dijon	22.5 (2.4; 73.1)	4	18.9 (2.9; 81.3)	1	69.6 (4.0; 156.0)	4
Grenoble	23.8 (2.7; 80.8)	3	24.1 (3.5; 90.7)	3	63.0 (1.2; 157.1)	6
Le Havre	22.4 (2.0; 79.0)	1	22.4 (4.0; 96.0)	1	70.8 (3.8; 178.1)	1
Lens-Douai	21.7 (1.0; 81.0)	2	24.2 (3.0; 97.6)	2	55.5 (0.8; 175.5)	2
Lille	25.3 (3.5; 80.3)	3	23.6 (4.9; 100.8)	3	57.5 (1.0; 193.6)	5
Lyon	31.1 (4.3; 96.3)	4	25.2 (5.0; 98.3)	3	64.8 (0.5; 184.4)	4
Marseille	32.7 (4.5; 82.3)	4	31.2 (6.0; 105.5)	2	77.6 (4.5; 178.0)	2
Montpellier	25.9 (1.1; 79.5)	2	20.5 (3.7; 77.1)	1	80.0 (13.3; 156.9)	3
Nancy	24.4 (4.7; 67.1)	3	24.9 (4.6; 91.4)	2	68.6 (1.9; 164.9)	3
Nantes	17.9 (1.8; 73.4)	2	20.2 (5.7; 89.2)	2	75.8 (7.6; 172.8)	2
Nice	24.9 (5.0; 55.0)	1	25.3 (4.0; 59.0)	1	85.0 (14.6; 176.5)	2
Paris	36.0 (8.3; 93.0)	15	25.1 (5.3; 111.8)	7	58.7 (1.5; 179.8)	13
Rennes	18.7 (0.7; 84.45)	1	19.6 (3.2; 89.0)	1	63.8 (4.3; 154.4)	1
Rouen	25.7 (3.7; 75.0)	3	24.6 (5.5; 112.5)	2	65.1 (3.8; 173.8)	4
Strasbourg	27.0 (4.7; 73.5)	2	20.9 (3.0; 78.7)	2	46.2 (0.3; 196.1)	2
Toulouse	21.1 (2.3; 76.8)	3	21.2 (4.1; 85.3)	3	77.1 (5.1; 158.5)	5

2.2.3. Meteorological Data

We obtained daily minimum, mean, and maximum temperature data from the French national meteorological service (Météo-France), with one reference monitoring station per area studied. Airport stations were used for 15 areas and city center stations were used for the other 3 areas.

2.2.4. Statistical Analyses

The association between NO_2 and daily mortality was investigated using a time-series analysis with generalized additive Poisson regression models (GAM) allowing for overdispersion [12].

In each area, the daily death count was regressed on NO_2 levels, while the following possible confounders were controlled: temperature, long-term trends, seasonal trends, day of the week, and bank holiday effects. Seasonality was taken into account using a penalized spline function. The smoothing parameter of the spline was selected in order to minimize the absolute value of the partial autocorrelations of the residuals over 30 days by imposing a minimum of 3-degrees of freedom per year [13]. The mean levels of NO_2 observed during the current and previous days (i.e., lag 0–1 days) were introduced as a linear term.

Minimum and maximum temperatures were introduced into the model as a natural spline with 3-degrees of freedom. Daily minimum temperatures at lag 0 and daily maximum temperature at lags 1–7 were also introduced as natural splines with 3-degrees of freedom. We assumed that minimum temperatures at lag 0 would represent a possible short-term heat effect, while maximum temperatures at lags 1–7 would represent a possible delayed cold effect [14,15].

The regression equation is formulated below:

$$Yt \sim quasiPoisson(\mu t)\log[\mu t] = intercept + \delta 1 dow + \delta 2 hol + \delta 3 pol + s(time) + ns(tmin) + ns(tmax)$$

where Yt denotes the daily mortality counts on day t, *dow* is the day of the week, *hol* is a bank holiday, *pol* is the mean level of NO_2 observed during the current and previous days, and $\delta 1$, $\delta 2$, and $\delta 3$ are the corresponding regression coefficients. *s(time)* is a penalized spline function of calendar time designed to control for trend and seasonality, *ns(tmin)* and *ns(tmax)* are natural splines of temperature designed to control weather, *tmin* is the daily minimum temperatures at lag 0, and *tmax* is the daily maximum temperature at lags 1–7.

For each area, we tested the assumptions of the model using the following set of graphical and statistical tools: (1) The observation of the residual plot helped verify that, after modeling, no particular structure (trend, seasonality) persisted; (2) The residuals were assimilated to white Gaussian noise. Residual autocorrelation was particularly influenced by the choice of smoothing parameter of the spline function modeling seasonality; (3) The normality of the residuals was checked graphically (histogram, QQPLOT); (4) The statistical significance of the partial autocorrelations was verified graphically on the partial correlogram; (5) The comparison of the graphs of the observed and predicted values allowed the quality of the model to be assessed; (6) The partial effect of each factor on the health variable was shown graphically. The graphs helped check the coherence of the effects (Appendix A—Figures A1 and A2: example for Paris and Montpellier).

Air pollution effects were estimated for each year and each season (warm period: May to October; cold period: November to April) for each cause of mortality. An interaction between the pollution indicator and the season was added to the models to study seasonal effects on pollution.

To investigate the confounding of NO_2 measurement by PM_{10} and O_3, we fitted two-pollutant models. We also fitted polynomial distributed lag models over a period of 6 days (0–5 days) to investigate the shape of each lagged association [16].

In the second stage, we pooled area-specific estimates to perform a meta-analysis using random-effect models [17]. We tested for heterogeneity and reported it using the I^2 statistic [18]. Association estimates were expressed as excess relative risk (ERR) of mortality for a 10 µg m^{-3} increase in air pollutant concentrations.

The time-series analysis was conducted using the R package mgcv (which provided generalized additive modeling functions), dlnm (which contained functions to specify and interpret distributed lag linear and non-linear models), and mvmeta (this function performed fixed and random-effects multivariate and univariate meta-analysis and meat regressions) (version 3.2.3, http://cran.r-project.org/).

3. Results

3.1. Main Characteristics of the Study Areas

3.1.1. Population Data

Table 2 summarizes population and mortality data for each area included in the analysis. The 18 areas covered a combined total of over 15 million inhabitants. The largest area was Paris, with more than 6.7 million inhabitants. With 8861 inhabitants per km^2, the population density was two to eight times higher than in the other areas (Appendix B—Table A1). Lille had the second largest population, but a density below 1900 inhabitants per km^2, while Nice had one of the densest populations with more than 3600 inhabitants per km^2, despite its mid-size population (see Appendix B—Table A1).

Table 2. Demographic characteristics and daily mean number of non-accidental and cardiovascular deaths for all ages and for people aged 75 years and over in 18 French areas: 2010–2014.

Demographic Characteristics		Daily Mean Number of Deaths			
		Non-Accidental		Cardiovascular	
Area	Total Population	All Ages	75 Years and Over	All Ages	75 Years and Over
Bordeaux	686,824	12.6	8.9	3.3	2.7
Clermont-Ferrand	284,672	5.8	4	1.5	1.2
Dijon	241,591	4.7	3.3	1.2	1
Grenoble	484,122	8	5.7	2.1	1.8
Le Havre	235,56	5.8	3.6	1.5	1.1
Lens-Douai	324,286	8.6	5.3	2.1	1.6
Lille	1,133,920	20.7	12.9	5.2	3.9
Lyon	1,082,180	18.1	12.4	4.5	3.7
Marseille	979,05	21.6	15.3	5.8	4.7
Montpellier	421,647	6.6	4.6	1.8	1.4
Nancy	328,919	6.9	4.7	1.7	1.3
Nantes	633,391	10.6	7.2	2.8	2.3
Nice	434,581	11.7	8.6	3.1	2.6
Paris	6,754,282	104.5	66.8	23.8	18.7
Rennes	250,458	4.0	2.8	1.2	1
Rouen	449,687	9.9	6.6	2.7	2.1
Strasbourg	448,424	8.6	5.7	2.3	1.8
Toulouse	814,162	12.2	8.3	2.9	2.4

3.1.2. Mortality Data

In the 18 French areas studied, the daily mean number of non-accidental deaths ranged from 4% in Rennes (one of the smallest areas) to 104.5% in Paris, the biggest area (Table 2). The percentage of non-accidental deaths in people aged 75 and over ranged from 62% in Lens-Douai, Le Havre, and Lille to 74% in Nice. The daily mean number of deaths for cardiovascular diseases ranged from 1.2% in Dijon to 23.8% in Paris. For respiratory mortality, the very small numbers of deaths prevented us from being able to perform statistical analyses of the links between air pollution and death with sufficient statistical power. Respiratory mortality was, therefore, not considered in this study.

3.1.3. Pollution Data

Mean levels of air pollution indicators (NO_2, PM_{10}) over the study period in each area are presented in Table 1.

There were large variations in NO_2 levels across the 18 areas over the study period. The highest mean levels (36.0 μg m^{-3} in Paris and 32.7 μg m^{-3} in Marseille) were approximately twice as high as the lowest (17.9 μg m^{-3} in Nantes and 18.7 μg m^{-3} in Rennes). NO_2 levels were higher during the cold period (November–April) than the warm period (May–October). During the cold period, mean levels

ranged from 23 μg m^{-3} in Nantes to 42 μg m^{-3} in Paris. During the warm period, they ranged from 13 μg m^{-3} in Nantes to 30 μg m^{-3} in Paris.

Mean PM_{10} levels ranged from 18.9 (in Dijon) to 31.2 μg m^{-3} (in Marseille). Mean O_3 levels ranged from 46.2 (in Strasbourg) to 85.0 μg m^{-3} (in Nice).

3.1.4. Temperature Data

Between 2010 and 2014, the minimum mean annual temperatures ranged from 6.5 (Nancy) to 13.1 °C (Nice), and maximum mean annual temperatures ranged from 14.1 (Le Havre) to 20.7 °C (Marseille). Mean annual temperatures ranged from 5.6 (Nancy) to 11.5 °C (Nice) during the cold period and from 15.4 (Rouen) to 21.4 °C (Marseille) during the warm period (Table 3). Temperature was taken into account in the analysis as a confounding factor.

Table 3. Temperatures in 18 French areas: 2010–2014.

Urban Area	Average Temperatures (°C) by Period					
	Minimum		Maximum		Mean	
	Cold *	Warm **	Cold *	Warm **	Cold *	Warm **
Bordeaux	5.3	13.6	13.4	24.1	9.4	18.9
Clermont-Ferrand	2.3	11.6	11.2	22.8	6.8	17.2
Dijon	2.1	11.5	9.6	22.1	5.9	16.8
Grenoble	1.7	12.3	11.2	24.2	6.5	18.3
Le Havre	5.4	13.0	9.7	18.4	7.6	15.7
Lens-Douai	3.1	11.4	9.5	20.3	6.3	15.9
Lille	3.1	11.4	9.5	20.3	6.3	15.9
Lyon	3.6	13.6	11.1	23.9	7.4	18.7
Marseille	5.8	16.1	14.5	26.7	10.2	21.4
Montpellier	5.7	15.9	14.6	25.8	10.2	20.9
Nancy	1.9	10.9	9.3	21.6	5.6	16.3
Nantes	4.4	11.9	12.2	22.2	8.3	17.0
Nice	8.1	18.1	14.8	24.2	11.5	21.2
Paris	5.0	13.4	10.8	22.0	7.9	17.7
Rennes	4.0	11.1	11.8	21.6	7.9	16.4
Rouen	2.9	10.6	9.8	20.2	6.4	15.4
Strasbourg	2.1	11.6	9.6	22.5	5.9	17.0
Toulouse	4.9	14.3	12.8	24.8	8.9	19.6

* Cold period = November to April; ** Warm period = May to October.

3.2. Concentration-Response Functions

3.2.1. NO_2 Analysis of the Same Day and the Day before (lag 0–1)

Table 4 presents, for each full calendar year and for each period (i.e., warm and cold), the ERR estimates for non-accidental and cardiovascular deaths for a 10 μg m^{-3} increase in NO_2 levels on the same day and the day before (lag 0–1). This table also presents sensitivity analyses with two-pollutant models for PM_{10} and O_3.

We found a significant link between NO_2 levels and non-accidental and cardiovascular deaths for each full calendar year and for the warm period each year, irrespective of the age group considered. For those aged 75 years and over, ERR estimates were slightly higher than for the total population. This difference was more marked for non-accidental mortality: 1.14% (95% confidence interval (CI): (0.63; 1.66)) and 0.75% (95% CI: (0.40; 1.10)), respectively. No age difference was observed in the ERR for cardiovascular deaths: 1.15% (95% CI: (0.40; 1.91)) and 1.13% (95% CI: (0.37; 1.90)), respectively.

Table 4. Excess relative risk (ERR) and 95% confidence intervals (95% CI) of non-accidental and cardiovascular deaths associated with a 10 µg m^{-3} increase of NO_2 concentrations (lag 0–1 day) according to age (total population versus 75 years and over) using the single- and two-pollutant models (18 areas, metropolitan France, 2010–2014).

Pollutant	Non-Accidental Deaths		Cardiovascular Deaths	
	Total Population	≥75 Years	Total Population	≥75 Years
Annual				
NO_2	0.75 (0.40; 1.10)	1.14 (0.63; 1.66)	1.13 (0.37; 1.90)	1.15 (0.40; 1.91)
+PM_{10}	0.58 (0.15; 1.01)	0.79 (0.27; 1.33)	0.96 (−0.10; 2.04)	1.11 (0.08; 2.16)
+O_3	1.12 (0.63; 1.61)	1.47 (0.85; 2.10)	1.39 (0.53; 2.25)	1.39 (0.58; 2.21)
Cold period				
NO_2	0.01 (−0.41; 0.42)	0.20 (−0.30; 0.71)	0.23 (−0.61; 1.08)	0.25 (−0.65; 1.16)
+PM_{10}	0.09 (−0.42; 0.60)	0.28 (-0.35; 0.90)	0.46 (−0.74; 1.67)	0.73 (−0.64; 2.11)
+O_3	0.30 (−0.17; 0.77)	0.59 (-0.03; 1.21)	0.48 (−0.54; 1.51)	0.55 (−0.52; 1.63)
Warm period				
NO_2	2.65 (1.82; 3.48)	3.07 (1.97; 4.18)	3.05 (1.38; 4.74)	3.17 (1.53; 4.84)
+PM_{10}	1.33 (0.37; 2.29)	1.61 (0.23; 3.00)	1.48 (0.08; 2.90)	1.68 (0.20; 3.18)
+O_3	2.54 (1.62; 3.47)	2.98 (1.82; 4.17)	3.16 (1.32; 5.03)	3.41 (1.57; 5.29)

All ERR values were also higher for the warm period than for the whole calendar year, irrespective of the cause of death or age group. The highest ERR was 3.17% (95% CI: (1.53; 4.84)) for cardiovascular deaths among those aged 75 years and over in the warm period. In the cold period, there was no association.

After adjusting the models for PM_{10}, the ERR estimates remained unchanged in the annual period and in the cold period and were lower in the warm period. The ERR estimates also remained unchanged after adjusting the model for O_3.

3.2.2. NO_2 Analysis—Over the First Six Days (Lag 0–5)

Figure 2 shows the distribution of the ERR for non-accidental and cardiovascular deaths over 0 to 5 days measured separately (lag 0, lag 1, lag 5) and 0 to 5 cumulative days (lag 0–1, lag 0–2, lag 0–5) for a 10 µg m^{-3} increase in NO_2. In cumulative lags, 0–5 effects were cumulated over 0 to 5 days (i.e., all contributions from day 0–5 were summed). An increase in ERR values was observed for the cumulative lags, irrespective of the cause of death or age group. The point estimate for the ERR for the cumulative lags for non-accidental deaths was slightly higher for those aged 75 years and over than for the total population. More specifically, the ERR for the cumulative 0–5 lags was 2.35% (95% CI: (1.35; 3.37)) for those aged 75 years and over and 1.74% (95% CI: (1; 1.48)) for the total population.

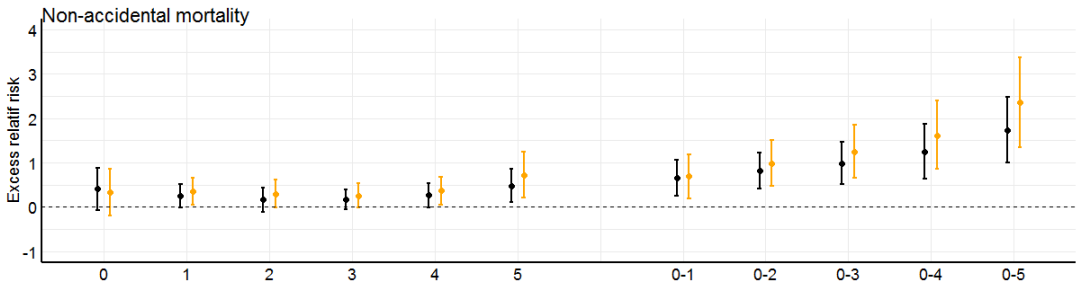

Figure 2. *Cont.*

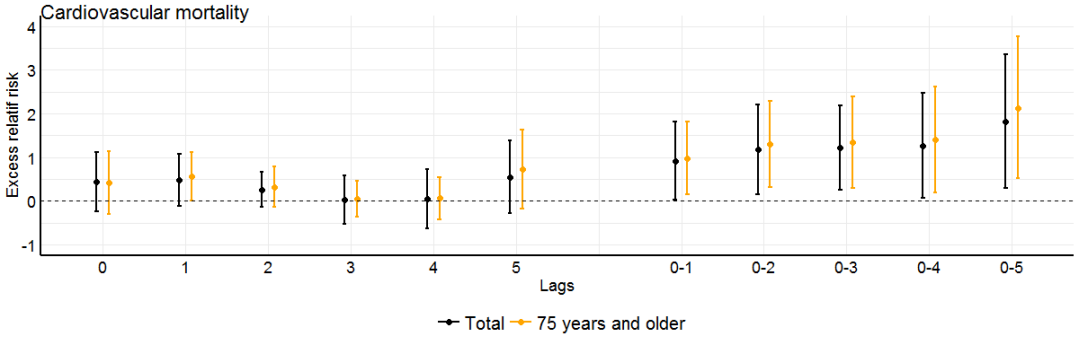

Figure 2. Excess relative risk (ERR) and 95% confidence intervals (95% CI) for non-accidental and cardiovascular deaths associated with a 10 μg m^{-3} increase of NO_2 concentrations, according to age (total population and ≥75 years) using different lags (single and cumulative) in single pollutant models (18 areas, metropolitan France, 2010–2014).

4. Discussion

In this study, we found an increase in the numbers of non-accidental and cardiovascular deaths associated with a 10 μg m^{-3} increase in NO_2 for both the total population and people aged 75 years and over, for the entire study period (2010–2014), for warm and cold periods, and for the warm period alone. These study results complement the ERR values published by Blanchard et al. in 2008 [5] as they include new urban agglomerations that were integrated into the French national public health agency's air and health program in 2011. Indeed, the addition of these areas with different profiles (population, pollution, and climate), helped us increase the statistical power of this meta-analysis.

The deleterious effects on health of NO_2 have been examined by the US EPA Integrated Science Assessment report on the oxides of nitrogen. The report states that the cumulative body of evidence on NO_2 indicates that short-term exposure can affect health, in particular, effects related to asthma exacerbation, and may be associated with cardiovascular effects and premature mortality. However, the report states that to assign a causal effect of NO_2 on mortality requires more research to separate NO_2 exposure effects from those of other traffic-related pollutants [6]. The REVIHAAP report by WHO [3] also states that the observed associations in epidemiological studies may not be completely attributable to NO_2 per se, as NO_2 may also include other constituents (which have adverse health effects) not represented by currently regulated pollutants. This is why NO_2 is used as one possible tracer, indicator, or proxy of traffic air pollution among others in this study.

Using a single-pollutant (such as NO_2) as a proxy for complex mixtures of pollutants, especially in urban areas, often misrepresents the real pollution mixture, as well as the associated health outcomes (which may depend upon a different component of the mixture). In this study, we were not able to distinguish which components of the mixture represented by NO_2 were responsible for the observed effects. NO_2 is typically formed through the following photochemical process. Due to ultraviolet radiation, NO_2 dissociates into nitrogen oxide (NO) and atomic oxygen (O). Atomic oxygen (O) combines with dioxygen (O_2) very quickly and produces the main photochemical pollutant, ozone (O_3). This reaction occurs in the low layers of the atmosphere and favors sunshine and warm temperatures. Ozone can also react with NO to regenerate NO_2 (https://www.aeroqual.com/meet-the-nitrogen-oxide-family). Relative toxicity of air pollution mixtures has been widely studied [19], and the effects of photochemistry on the toxicity of air pollution are well documented [20]. It is known that photochemical reactions can release secondary organic aerosols that increase inflammatory responses, and this phenomenon enhances the toxicity of air pollution [21,22].

The potential causal role of NO_2 and the pathophysiological mechanisms by which NO_2 may affect health have been studied. As per the REVIHAAP report [3], there is evidence of small effects on inflammation and increased airway hyperresponsiveness with NO_2 per se in the range from 0.2 to 1 ppm (380 to 1880 μg m^{-3}) from chamber studies (under a broad range of exposure conditions, with exposure durations of 15 min to 6 h, with some inconsistency in results), with more marked,

consistent, responses observed from 1 ppm (1880 μg m^{-3}). Newer review reports suggest weak to moderate lung cell changes in animals at one-hour concentrations of 0.2 to 0.8 ppm (380–1500 μg m^{-3}). These concentration ranges are not far from concentrations that occur at the roadside or in traffic for multiple hours. The WHO report concludes that there is some mechanistic support for causality, particularly for respiratory outcomes. This is confirmed by Petit et al. [23], who state that NO_2 is a toxic gas that can damage the lungs. When inhaled, it oxidizes protective antioxidants within the epithelial lining fluid and triggers extracellular damage in the airways. The presence of NO_2 within the epithelial lining fluid triggers oxidative stress, possibly leading to edema, bronchoconstriction, and a reduced forced expiratory volume in 1 s.

Despite results in international studies showing the associations of NO_2 with respiratory mortality, we were not able to carry out an analysis of this indicator because of the low number of deaths attributed to respiratory mortality in the study areas.

The present study showed that NO_2 exposure was associated with an increased number of non-accidental and cardiovascular deaths. Excess relative risks estimated in the present study for a 10 μg m^{-3} increase in NO_2 were lower than those published in the previous meta-analysis in 2008, although the comparison was limited by the different statistical methods used in both studies, and the higher number of areas included in this work (18 vs 9) [5].

The study results for the general population were consistent with those in the literature and especially with those from multicenter and meta-analysis studies. Two similar studies, the first conducted in 2015 by Perez et al. in 21 cantons in Switzerland [9] and the second in 2016 by Carugno et al. in 18 areas of the Lombardy region of Italy [24], found a significant increase in all-cause deaths for a 10 μg m^{-3} increase in NO_2 concentrations. The results of a meta-analysis of several studies performed throughout the world, conducted by Mills et al. in 2015, showed a similar risk, and when focusing only on European studies, the ERR was slightly higher [2]. Another study carried out in 2015 by Renzi et al. in the Rome area showed a higher risk than in the present study [25]. A study of the Vienna area by Neuberger et al. found a similar ERR to this study [10], and finally, a Canadian study by Crouse et al. showed a significantly lower risk than the present study [26] (Table 5).

Table 5. Excess relative risk (ERR) and 95% confidence intervals (95% CI) of non-accidental and cardiovascular deaths associated with a 10 μg m^{-3} increase of NO_2 concentrations (lag 0–1 day).

			Non-Accidental Deaths		Cardiovascular Deaths	
References	**Years**	**Areas**	**All Ages (%)**	**75 Years and Over (%)**	**All Ages (%)**	**75 Years and Over (%)**
Perez et al.	2015	Switzerland	0.7 (0.1; 1.3)	0.6 (0.0; 1.2)	0.4 (−0.1; 0.8)	0.3 (−0.3; 0.8)
Carugno et al.	2016	Italy	0.7 (0.13; 1.27)	-	1.12 (0.14; 2.11)	1.17 (0.1; 2.26)
Mills et al.	2015	World	0.71 (0.43; 1.00)	-	0.88 (0.63; 1.13)	-
		European	0.9 (0.45; 1.35)	-	1.03 (0.70; 1.36)	-
Renzi et al.	2017	Rome	1.8 (1.35; 2.25)	-	-	-
Neuberger et al.	2013	Vienne	0.8 (0.0; 1.6)	-	-	-
Crouse et al.	2015	Canada	0.5 (0.4; 0.6)	-	0.4 (0.3; 0.5)	-
This study	2020	France	0.75 (0.40; 1.106)	1.14 (0.63; 1.66)	1.13 (0.37; 1.90)	1.15 (0.40; 1.91)

Most international studies to date also examined the relationship between cardiovascular mortality and NO_2 concentrations but found a slightly lower excess risk than in this study (1.13% [0.37–1.90]). In Europe, the meta-analysis results of Mills et al. in 2015 [2] and the Carugno et al. study [24] both showed a significant increase in cardiovascular mortality for a 10 μg m^{-3} increase in NO_2, with an ERR close to this study. In contrast, the Swiss study by Perez et al. in 2015 [9], the worldwide study by Mills

et al. in 2015 [2], and the Canadian study by Crouse et al. in 2015 [26] showed a significantly lower risk than the present study (Table 5).

We observed slightly higher ERR in people aged 75 and over than in the total population for non-accidental (1.14% vs 0.75%) and cardiovascular (1.15% vs 1.13%) deaths. Chinese studies have shown higher risks among people aged 75 and over, although age-related differences were negligible [27,28]. The Italian study by Carugno et al. in 2016 showed a higher risk for cardiovascular deaths but not for non-accidental deaths [24] in the same group. However, other studies have shown a lower risk for this sub-population [9] (Table 5).

NO_2 associations were more evident in the warm period (May–October). Similar results have been observed in studies in several countries, for example, in Italy [24,29], South Africa [30], Canada [31], and China [28]. One likely explanation for this is that during the warm period, the concentrations measured by the monitoring stations better represent real exposure as people spend more time outdoors in the warm period and keep windows open, leading to ambient air pollutants entering their homes. Another hypothesis is that during the warm season, the increase in photochemical activity leads to a change in the composition of the mixture in automobile emissions, with more volatile organic compounds (VOC) and finer particles than in the cold period. As these pollutants have been identified as having health effects [3,30], the mixture of pollutants from automobile emissions, with NO_2 as a proxy, would potentially have a larger health effect in the warm season.

In order to dissociate the effect of NO_2 from that of other pollutants, in particular from traffic, we adjusted models for PM_{10} and O_3. These were the only pollutants we could adjust for, despite $PM_{2.5}$, SO_2, and CO also being analyzed in other articles. In France, SO_2 is not measured anymore in urban areas because concentrations are extremely low (except for hotspots), and for CO, we do not have continuous measurements. $PM_{2.5}$ concentrations were not available in all the study areas, and, in most cases, data was not yet consolidated and came from only one monitoring station for all areas. It takes time for emitted particles to agglomerate and/or grow via uptake of secondary pollutants before they reach the size of PM_{10}. However, because PM_{10} and $PM_{2.5}$ indicators are highly correlated, and because it is preferable to work with robust data from several monitoring stations in order to limit the impact of possible measurement errors or missing data on an indicator, PM-adjusted models were only built for PM_{10}. The estimates of ERR for NO_2 remained unchanged after adjustment for PM_{10}, and this result was consistent with the literature. Several studies reviewed in REVIHAAP by the WHO [3] also found significant associations between NO_2 and all-cause mortality after adjustment for PM_{10} [29,32–34].

The present results remained unchanged after adjusting for O_3. A Brazilian study [35] showed that the ERR for circulatory mortality differed little between single-pollutant NO_2 analysis and analysis adjusted for the other pollutants (PM_{10}, CO, and O_3). A meta-analysis conducted in 2016 [36] identified that most studies showed a PM_{10}-independent effect of NO_2. More specifically, for all-cause mortality, a 10 $\mu g\,m^{-3}$ increase in NO_2 was associated with a 0.78% increase in the risk of death (95% CI: (0.47; 1.09)). This increase was 0.60% (95% CI: (0.33; 0.87)) after adjustment for particulate matter.

Finally, we observed an increase in ERR for cumulative lags, irrespective of the cause of death or the age group. This result was consistent with those found in international studies. The associations were larger on multi-day lags (0–5 cumulative lags) than on single-day lags (0–1 lags) [27,28,32,37].

5. Conclusions

This study found an association between short-term exposure to NO_2 concentrations as a proxy of urban traffic pollution and the risk of death, with a higher risk for people aged 75 years and over. The results support the strengthening of measures to reduce traffic air pollution sources in Europe to protect the most vulnerable.

Author Contributions: Conceptualization, M.C., M.B., and V.W.; Methodology, M.C., M.B., and V.W.; Analysis, V.W. and M.C.; Data curation, M.C.; Writing—original draft preparation—review, M.C., Writing—review and editing M.C.; S.M., V.W., and M.B.; All authors have read and agreed to the published version of the manuscript.

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Conflicts of Interest: The authors declare no conflict of interest.

Appendix A

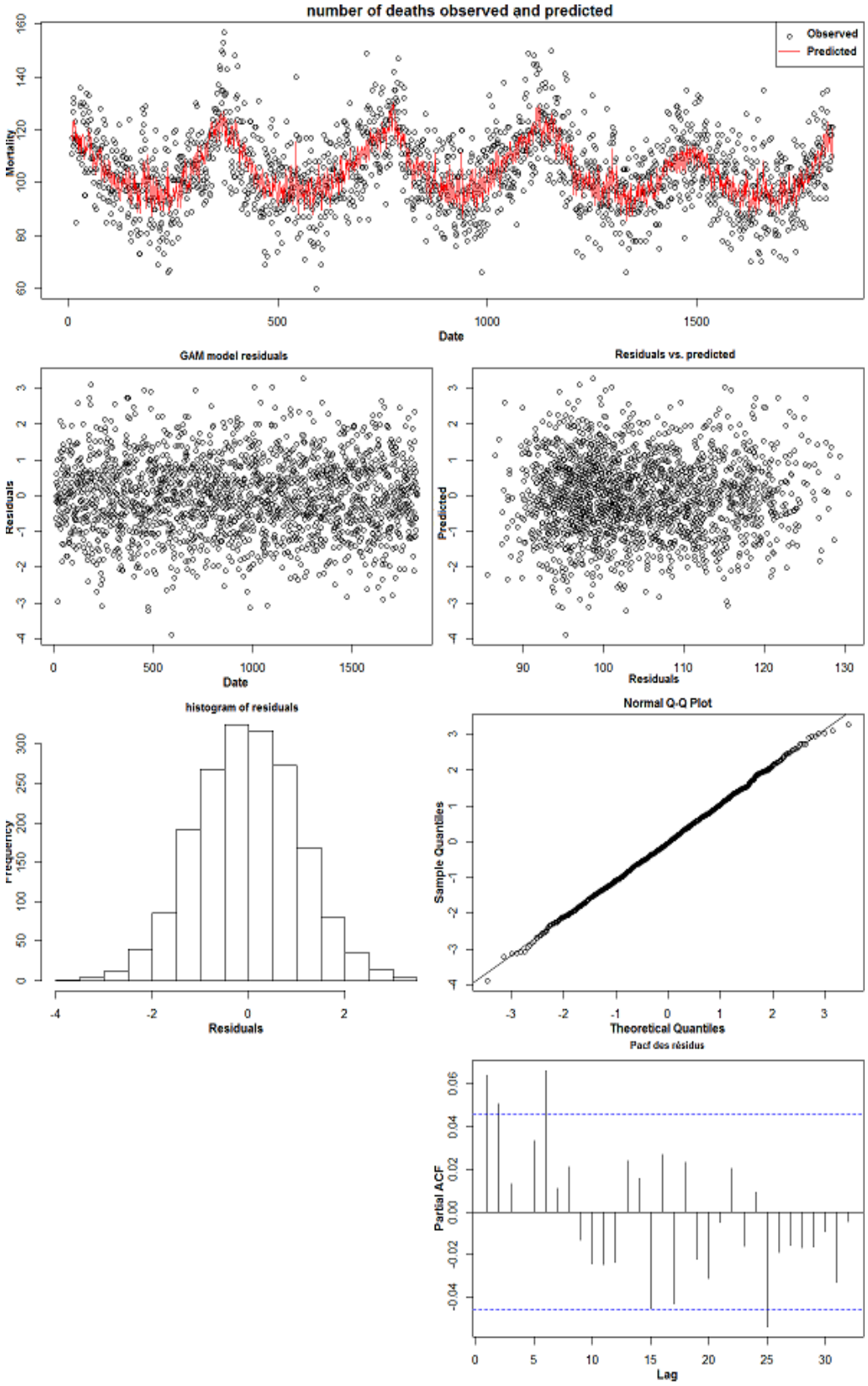

Figure A1. *Cont.*

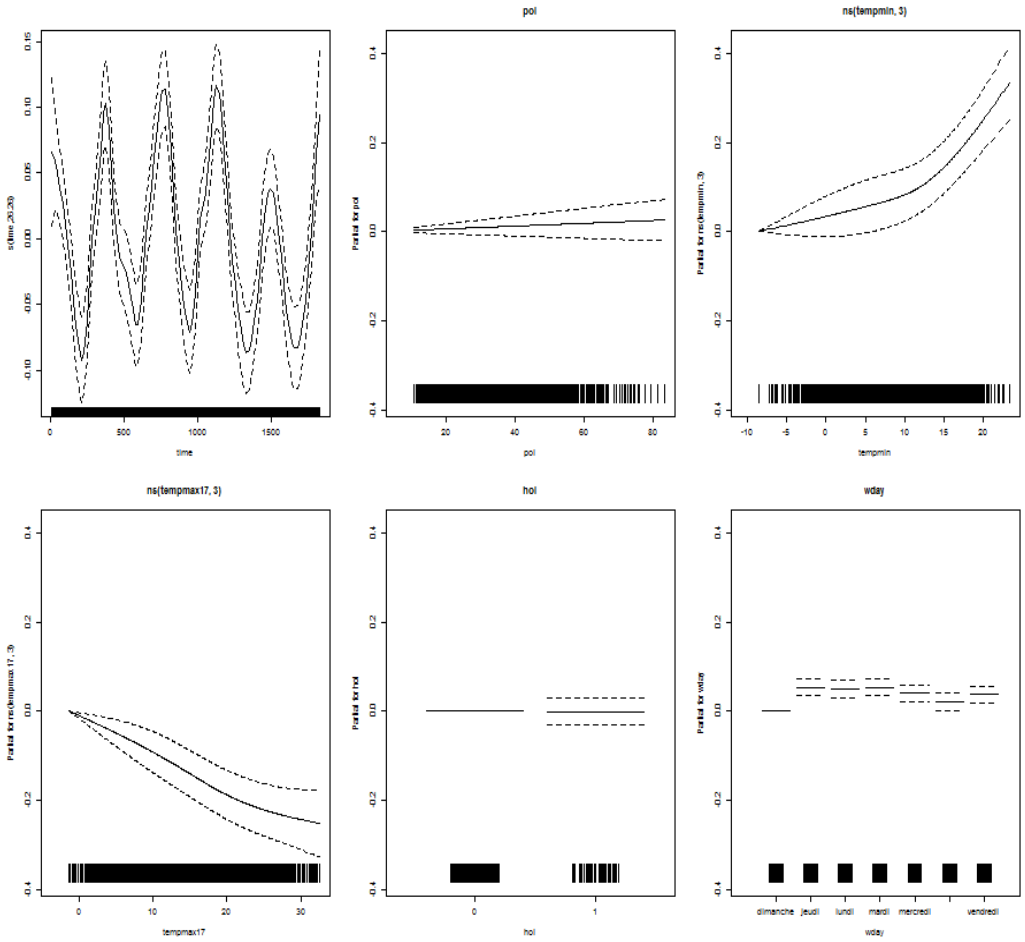

Figure A1. Assessment of the modeling assumptions for Paris.

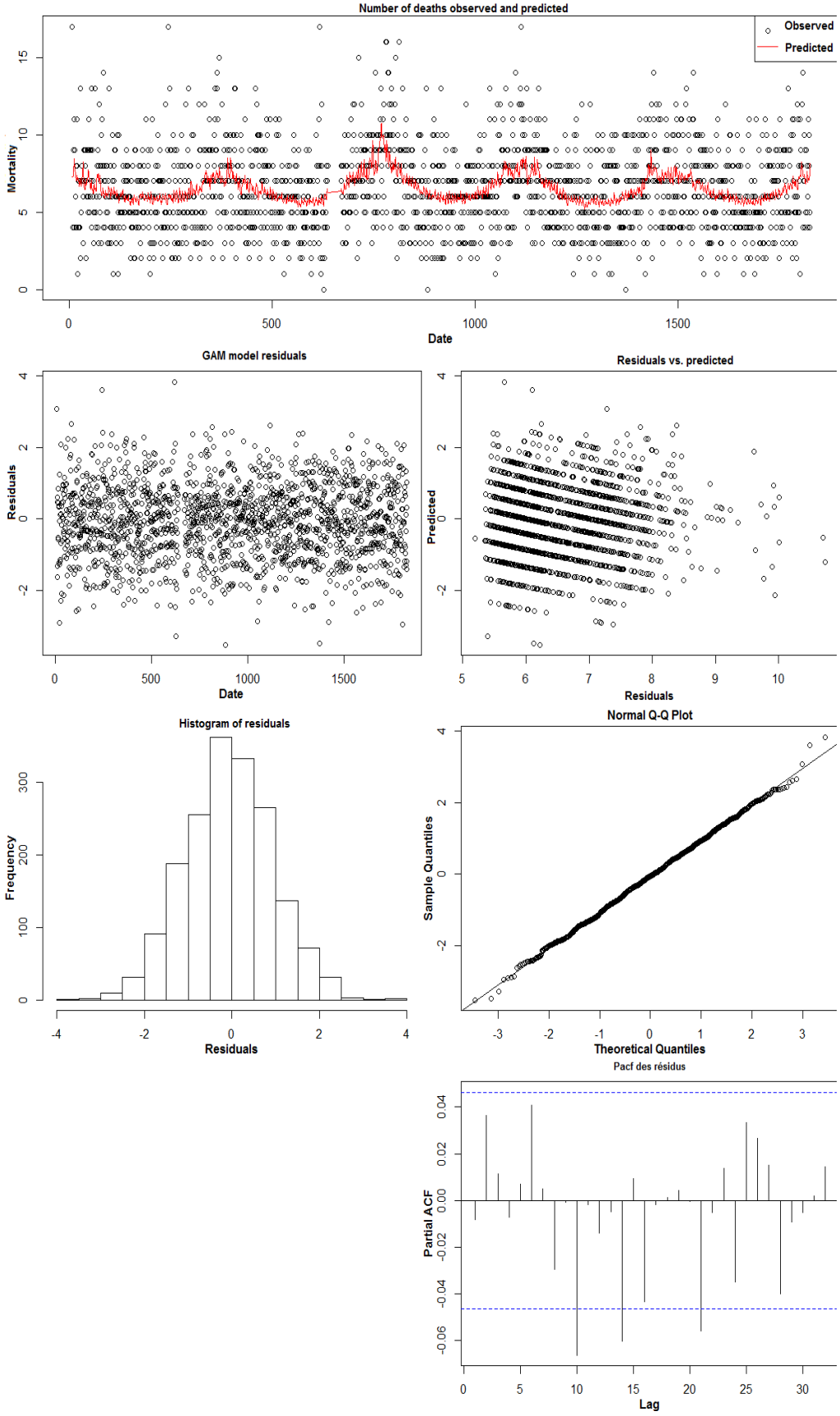

Figure A2. *Cont.*

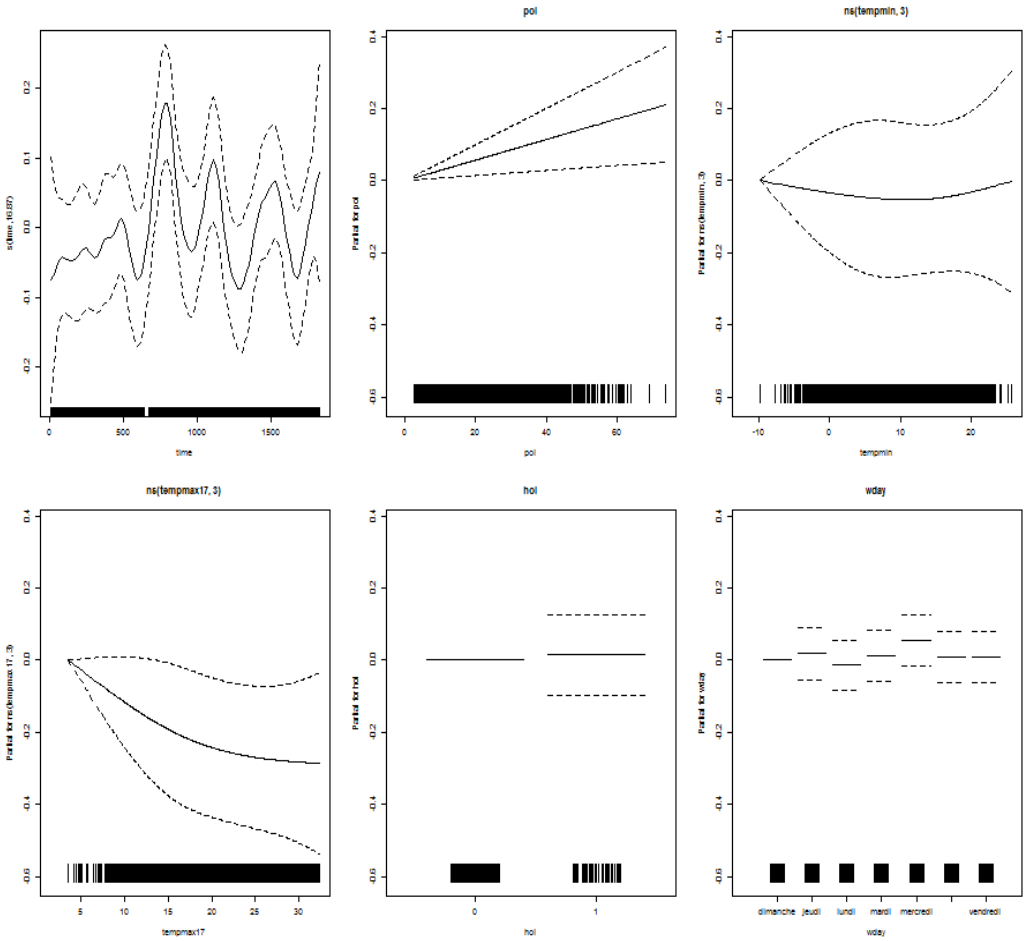

Figure A2. Assessment of the modeling assumptions for Montpellier.

Appendix B

Table A1. Population density in 18 French areas: 2010–2014.

Urban Area	Population Density (Inhabitants/km^2) (2014 census)	Number of Municipalities
Bordeaux	2056	22
Clermont-Ferrand	1950	16
Dijon	1455	15
Grenoble	1013	46
Le Havre	1287	16
Lens-Douai	1417	32
Lille	1854	85
Lyon	4679	19
Marseille	2214	8
Montpellier	1360	22
Nancy	1010	38
Nantes	1058	27
Nice	3634	4

Table A1. *Cont.*

Urban Area	Population Density (Inhabitants/km^2) (2014 census)	Number of Municipalities
Paris	8861	124
Rennes	2240	4
Rouen	1262	43
Strasbourg	2016	20
Toulouse	1537	51

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