Nitrogen dioxide and mortality: review and meta-analysis of long-term studies

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ABSTRACT

Background. Exposure to ambient nitrogen dioxide (NO_2) has been linked to increased mortality in several epidemiological studies but the question remains of whether NO_2 is directly responsible for the health effects, or is only an indicator of other pollutants, including particulate matter (PM).

Aim. To provide pooled estimates of the long-term effects of NO_2 on mortality, potentially useful for health impact assessment.

Methods. We selected 23 papers, published from 2004 to 2013, evaluating the relationship between NO_2 and mortality, also including an assessment of the effect of PM exposure. A random effects meta-analysis was carried out on 19 studies.

Results. The pooled effect on mortality was 1.04 (95% confidence intervals (CI) = 1.02-1.06), with an increase of 10 $\mu g/m^3$ in the annual NO_2 concentration, and 1.05 (95% CI=1.01-1.09) for $PM_{2.5}$ ($10\mu g/m^3$). The effect on cardiovascular mortality was 1.13 (95% CI=1.09-1.18) for NO_2 and 1.20 (95% CI=1.09-1.31) for $PM_{2.5}$. The NO_2 effect on respiratory mortality was 1.03 (95% CI=1.02-1.03) and 1.05 (95% CI 1.01-1.09) for $PM_{2.5}$. Four bi-pollutant analyses with PM and NO_2 in the same models showed minimal changes in the effect estimates of NO_2.

Conclusions. There is evidence of a long-term effect of NO_2 on mortality as great as that of $PM_{2.5}$. An independent effect of NO_2 emerged from multi-pollutant models.

Key words: air pollution, long-term NO_2 and $PM_{2.5}$ exposure, natural mortality, cause-specific mortality, systematic review, meta-analysis.

Running head: long-term NO_2 exposure and mortality

The most important message of our study: Any health impact assessment overlooking NO_2 would neglect some adverse effects of today's air pollution mixture.

Introduction

The long-term effects on mortality of exposure to nitrogen dioxide (NO_2) were assessed in the WHO Air Quality Guidelines in 2005 [1] and the overall evidence was considered limited, given the small number of studies available. Neither the development of chronic diseases nor lung cancer was clearly associated with nitrogen dioxide in any of the studies taken into consideration in the 2005 WHO report. On the other hand, short-term mortality studies, studies of the impairment of lung function growth in children, and investigations of recurrent respiratory problems all suggested that there was evidence of a NO_2 effect. Recent documents on traffic exposure from the American Thoracic Society [2] and the Health Effect Institute [3] considered NO_2, among other traffic-related pollutants, and concluded that there is evidence suggesting that these pollutants have a causal role on mortality and on the development of chronic respiratory diseases.

The recent review of the health effects of air pollution by a WHO working group (Review of evidence on health aspects of air pollution - the REVIHAAP project) [4] evaluated the most recent studies on long-term exposure to NO_2 and both natural (non-accidental)/total and cause-specific mortality. The question of whether NO_2 is directly responsible for the health effects or is only an indicator of other pollutants – including particulate matter (PM) and specific constituents such as metals, polycyclic aromatic hydrocarbons and other organic matter – was specifically considered. The conclusion was that it is difficult "to judge the independent effects of NO_2 in the long-term studies because, in those investigations, the correlations between concentrations of NO_2 and other pollutants are often high, so that NO_2 might represent a mixture. In this case, chamber studies do not apply and toxicological evidence is limited. However, some epidemiological studies do suggest associations of long-term NO_2 exposures with respiratory and cardiovascular mortality and with children's respiratory symptoms and lung function that were independent of PM mass metrics. As with the short-term effects, NO_2 in these studies may represent other constituents. Despite this, the mechanistic evidence, particularly on respiratory effects, and the weight of evidence on short-term

associations suggest a causal relationship." In other words, the WHO working group suggested that the response to the causality question cannot be derived only from the epidemiological studies but requires the integration of exposure science, toxicology, and human studies.

At the time of the REVIHAAP project, a quantitative systematic review of the long-term effects on mortality of NO_2 was not available. A subsequent review by Hoek et al. [5], considering the association between several pollutants and long-term mortality, showed a significant association of all-cause mortality with NO_2 in 15 longitudinal studies. Their work did not permit comparison of the effects of NO_2 with those of other pollutants, namely particulate matter (PM), when the same studies were considered or when the results of the multi-pollutant models were available.

To contribute to the discussion about the role of NO_2 from an epidemiological perspective, and to provide pooled estimates of the long-term effects of NO_2 on mortality, potentially useful for assessing the health impact of air pollution, we systematically examined the studies that investigated the long-term effects on mortality of both outdoor NO_2 and particles in the same population, and carried out a meta-analysis of both NO_2 and PM effects.

METHODS

Systematic review

The search strategy focused on studies reporting NO_2 (or NOx) effects on total and specific-cause mortality. The databases PubMed, Web of Science, and LUDOK were searched. The search was restricted to the period from January 2004 - January 2013 and to humans. Search terms were: nitrogen dioxide (as nitrogen dioxide/adverse effects MESH in PubMed, and as nitrogen oxides in LUDOK), together with cohort study or case-control study, and together with mortality, cardio-vascular (CV) mortality and respiratory mortality.

Articles were included if long-term effects on natural/total or specific (cardio-vascular or respiratory) mortality were assessed and the estimates of association with the pollutants were

reported; the exposure needed to have been measured as at least an annual mean concentration in the study area. Papers were excluded when short-term effects were estimated, no health effect was measured, incidence or prevalence was assessed, no original data were analysed and, finally, when no control for confounding due to individual factors was carried out. Reviews and methodological papers were excluded as well. Studies carried out on groups restricted to at-risk people were, however, not excluded.

The first screening of the papers was carried out by checking the titles and the abstracts. The texts of the remaining papers were read in full, so as to select those that also included effect estimates for particles with a diameter < 10 micron (PM_{10}) or with a diameter <2.5 micron ($PM_{2.5}$), total suspended particles (TSP) and black smoke (BS)).

The following characteristics were reported for each paper: authors, location and year of publication; study population; study design and study period (including both the enrolment and the follow-up period, if it was required by the study design); exposure assessment, including the method employed to assess levels, the mean concentration of pollutants and their exposure metrics; traffic exposure indicators (such as distance from the nearest high-traffic road, traffic density (TD), etc.); effect estimates on both total and specific-cause mortality.

Meta-analysis

Before pooling data, we converted the effect estimates related to NOx into NO_2 effects, using a conversion factor of 0.75 [6]; the NO_2 concentrations expressed in ppb were converted into $\mu g/m^3$; we used a conversion factor of 1.88 (at 25°C and 1013mb) for both NO_2 and NOx. The effects related to TSP were converted into PM_{10} effects, using a factor of 0.75 [7], and the effects related to PM_{10} were converted into $PM_{2.5}$ effects, using a factor of 0.7 [8]. Diesel particulate matter (DPM) and BS were analysed as indicators of $PM_{2.5}$. [9] In addition, the effects expressed as interquartile (or quintile or percentile) differences were converted into effects of a 10 $\mu g/m^3$ increase of each pollutant.

We used hazard ratios (HR), odds ratios (ORs) and relative risks (RRs) of mortality in a random effects meta-analysis [10] (Stata, version 10), thus including the risk estimates in the pooled analysis irrespective of the study design. The risk estimates and 95% confidence intervals (CI) from each study were reported, having been adjusted for all the factors the authors assessed as confounders. The standard error (SE) of the effect was calculated from the risk estimates and the confidence intervals (SE = (ln RR – ln lowest limit CI)/1.96 or SE = (ln highest limit CI – ln RR)1.96). The random pooled effects were reported as hazard ratios (HR), since most studies used Cox's survival analysis. We assessed heterogeneity across studies by using the χ^2 test (Cochran's Q) and the I^2 [11], which represents the proportion of total variation in effect estimates due to heterogeneity between the studies. Finally, we stratified the results by geographical area (Asia, North America, Europe).

To compare the relative effects of NO_2 and $PM_{2.5}$, we carried out a meta-analysis, using the interquartile range (IQR) as the exposure metric instead of the fixed increment (10 μg/m^3). We included only studies where the IQRs were available or could be derived by using the mean concentrations and their standard deviations. The pooled estimate was obtained for a median IQR of all the single IQRs from those reported or specifically estimated.

RESULTS

Our initial search led to the selection of a total of 180 papers that were seen to deal with NO_2 (or NOx) exposure. We excluded 146 papers on the basis of their titles and abstracts: 9 reviews and a further 12 papers which did not analyse original data; 2 methodological studies and 8 with study designs that made it impossible to adjust for individual confounders, 33 papers which did not assess health effects or human outcomes, 46 papers dealing with short-term effects, 26 papers with long-term effects other than mortality, and 9 papers with a mean exposure to air pollution considered for less than one year. Thereafter, from the 34 studies that had been identified as warranting reading in full, we selected 23 studies [12-34] which assessed PM effect in addition to NO_2 or NOx effects. All

the selected studies and their characteristics are reported in detail in supplemental Table A. The 11 excluded studies are listed in supplemental Table B.

Fourteen studies [12, 13, 17, 19, 20-24, 26, 30-32, 34] evaluated natural or total mortality; 17 studies [12, 13, 15, 17-20, 22, 23, 25, 26-29, 32-34] considered cardiovascular mortality (including four studies [13, 23, 32, 34] assessing cardio-pulmonary mortality); 14 studies considered respiratory mortality [12-14, 16, 17, 19, 20, 22, 23, 26, 28, 32, 34] (including the four with cardio-pulmonary mortality as a single outcome, [13, 23, 32, 34] and a further one [14] which assessed lung cancer and respiratory mortality as a single outcome). Only ten studies [12, 13, 17, 19, 20, 22, 23, 26, 32, 34] evaluated both total and cause-specific mortality, though data were not always exhaustively reported. There were 21 cohort studies [12-24, 26-28, 30-34] and two case-control studies [25, 29].

Cohort studies involved adults of both sexes, but one paper selected only adult men [20] and four selected only adult women [13,19,27,32], and two further paper [28, 33] studied both men and women but separately. Among the other studies, one studied only women [29]. Five studies focused on specific at-risk groups: patients with stroke [21], respiratory diseases [22], hypertension [24] and first myocardial infarction [25, 29].

Several confounders were assessed in each paper. Sex, age and social class (identified by different indicators such as education, deprivation index, occupation, area-level socio-economic-position (SEP) or income) were included in most studies. Only Dong, [14] Katanoda [16] and Lipfert [24] included no SEP indicator in their final analysis. Habitual smoking was frequently adjusted for [15-17, 21, 26, 27, 32, 33], whereas co-morbidities were rarely assessed [12,18,21].

Asia was represented by four studies: one from Japan and three from China; North America contributed nine studies: two from Canada and seven from the U.S.A., and Europe contributed 10 studies.

NO_2 was a main exposure in almost all studies, but two [17, 24] used NOx. The exposure metric was mostly expressed in μg/m^3 of NO_2, but one study from Asia [16], one from Canada [22] and all seven studies from the U.S.A. [19, 20, 23, 24, 30, 31, 33] reported the mean levels of NO_2 in ppb. The unit increase of NO_2 was usually reported (16 studies from all the geographic areas) as interquartile or centile range rather than as 10 μg/m^3 [12, 13, 18-20, 22-25, 26-29, 31-33]. The mean yearly level of NO_2 was 41.6 μg/m^3 in Asia, 42.2 μg/m^3 in North America, and 36.2 μg/m^3 in Europe.

Particulate matter was studied for the most part as PM_{10} (11 studies) [13-15, 19-21, 25, 27-29, 32] with the effect estimated for 10μg/m^3 (5 studies), and otherwise as IQR, expressed in μg/m^3. $PM_{2.5}$ was studied in 11 cases [12, 16, 18, 22-24, 26, 30, 31, 33, 34] with exposure expressed most frequently as an IQR increase of concentrations (six studies). Total suspended particles (expressed in 10μg/m^3) was the particle metric in one study [17]. The mean yearly level of $PM_{2.5}$ was 87.4 μg/m^3 in Asia, 14.8 μg/m^3 in North America, and 25.4 μg/m^3 in Europe.

The correlation between NO_2 (or NOx) and PM_{10} was reported in 8 studies out of 11, with values ranging from 0.5 [13] to 0.9 [14, 15, 29]. The correlation between NO_2 and $PM_{2.5}$ was reported in 9 studies out of 11, with values ranging from 0.3 in the Japanese study [16] and in the California study [33] to 0.88 in the Norwegian study [28]. The study using only TSP [17] did not indicate the correlation with NO_2.

Traffic exposure indicators were reported in nine publications. Two evaluated proximity to a road with heavy traffic and traffic density [12,26], the studies in Toronto [22] and in Germany (the SALIA cohort study) [13,27,32] investigated only proximity to traffic, and the U.S. Veterans studies [24,30,31] evaluated traffic density only.

We included 19 studies in the meta-analysis. Four of the 23 included in the systematic review have been further excluded from the meta-analysis since three of them reported more complete analyses

three years later [29-31] and one other [14] reported mortality only for respiratory diseases and lung cancer as a single outcome. In contrast, two of the included papers [28,33] carried out separate analyses for men and women, thus producing 21 effect estimates actually analysed.

The pooled estimate of natural/total mortality was performed for 12 studies [12, 13, 17, 19-24, 26, 32, 34], the pooled estimate of cardiovascular mortality made use of 18 effect estimates [12, 13, 15, 17-20, 22, 23, 25, 26, 27, 28 (men), 28 (women), 32, 33 (men), 33 (women), 34], the meta-analytic estimate of respiratory mortality included 9 effect estimates [12, 16, 17, 19, 20, 22, 26, 27 (men), 27 (women)].

The pooled estimate of NO_2 effects on natural/total mortality (Table 1; Figure 1) was 1.04 (95%CI = 1.02-1.06) per 10 µg NO_2/m^3 and the corresponding effect of $PM_{2.5}$ was 1.05 (95%CI = 1.01-1.09) per 10 µg/m^3. However, there was high heterogeneity across the studies in North America, the NO_2 effects were not statistically significant while the effects of $PM_{2.5}$ were larger and statistically significant (in spite of the fact that the levels of $PM_{2.5}$ observed there were the lowest we found). The greatest effect on natural/total mortality was observed in Europe for both NO_2 and $PM_{2.5,}$ but heterogeneity observed across the studies was still high. It should be noticed that a part of this heterogeneity could be explained by just one study [21] on stroke patients.

The pooled estimate of the NO_2 effect (Table 1; Figure 2) on cardiovascular mortality was 1.13 (95%CI = 1.09-1.18), while the effect of $PM_{2.5}$ was 1.20 (95%CI = 1.09-1.31), per 10 µg/m^3 of each pollutant. In Asia, the effects of both NO_2 and $PM_{2.5}$ were greater than those in North America and Europe. Notably, the results were affected by high heterogeneity in all areas. In Asia, where the overall NO_2 estimate did not attain statistical significance (and heterogeneity showed values near 99%), one study [15] provided very high effect estimates, much greater than for the other studies. In North America, the study in Toronto [22], carried out on patients from a respiratory clinic, provided effects on cardiovascular mortality as high as 64% for NO_2, in contrast with a mean increase of 1% in the other studies.

To better evaluate the heterogeneity of the pooled effect estimates of NO_2 on total and cardiovascular mortality, we conducted subgroup analyses considering studies not involving at-risk groups [excluding references 21, 22, 24, 25] and studies with better exposure assessment (land use regression, dispersion models, or other methods at the address level) [12, 18-22, 25, 26, 28]. The heterogeneity of the effects generally remained in these subgroups when they were considered separately (supplemental table D). However, when we restricted to studies conducted in the general population (not on at-risk groups) and with a better exposure assessment, the heterogeneity for cardiovascular mortality decreased (p-value = 0.690 and I^2 = 0%, based on 7 studies) corresponding to an effect estimate of 1.03 for NO_2.

The pooled estimate of NO_2 effects (Table 1; Figure 3) on respiratory mortality was 1.02 (95%CI = 1.02-1.03) while the $PM_{2.5}$ effect was 1.05 (1.01-1.09). The results for NO_2 were homogeneous across the studies. The large effect of $PM_{2.5}$ on respiratory mortality was notable in Europe, where an effect of 8% was detected with a heterogeneity as high as 40%.

The results of the pooled estimates using interquartile ranges (IQRs) as the exposure metric are presented in Table 2. The number of studies with available estimates is slightly lower than for the analysis using a fixed increment, since four of them [16, 17, 25, 34] did not provide IQRs data or figures to estimate them. The median IQRs were different for the outcomes considered and the variability of NO_2 levels was greater than for $PM_{2.5}$ (e.g. 14.1 $\mu g/m^3$ for NO_2 and 5.4 $\mu g/m^3$ for $PM_{2.5}$ in natural/total mortality studies). The estimates based on IQRs showed greater effects of NO_2 than of $PM_{2.5}$ on total mortality (6% vs. 3% for $PM_{2.5}$) and on cardiovascular mortality (29% vs. 16% for $PM_{2.5}$). The effects were, instead, similar for respiratory mortality (4% for both NO_2 and $PM_{2.5}$).

The results of the multi-pollutant analyses (as assessed in the original papers) are reported in supplemental Table C. Only seven studies [12, 17, 18, 20, 22, 24, 32], among those included in the

meta-analysis, performed bi-pollutant analyses, including Gehring [32], Jerrett [22] and Lipfert [24] who analysed the NO_2 effects only with a traffic indicator. Cao [17] in Asia and Cesaroni [12] in Europe found no change in the effects of NO_2 on total mortality when it was analysed together with PM, whereas the reduced effect observed by Hart [20] in the U.S.A. relates to a model including three pollutants (NO_2, PM_{10} and SO_2). Among the three studies which again analysed NO_2 and PM effects on cardio-vascular mortality (Cao [17] in Asia, Gan [18] in Canada and Hart [20] in the U.S.A), two [18,20] found independent effects of NO_2. Three authors analysed NO_2 effects with a traffic indicator: Lipfert [24] in the U.S.A. and Jerret [22] in Toronto found reduced effects of NO_2 on total and cardiovascular mortality, but no change was reported in the European study [32].

DISCUSSION

We found similar risk estimates for total mortality in studies investigating the long-term effects of both NO_2 and $PM_{2.5}$ (4% increase vs. 5%, respectively) using an exposure metric of 10µg/m^3. In Europe, there was a 7% increase of total mortality for both NO_2 and fine particles. Hoek et al. [5] in their recent review found a worldwide pooled estimate of 5.5% (95%CI = 3.1-8.0%) for NO_2; their study, unlike the present one, included estimates from studies that did not analyse particles. We found greater effects of $PM_{2.5}$ than of NO_2 for cardiovascular (20% vs. 13%) and respiratory (5% vs. 2%), per 10 µg/m^3 of pollutants. Per interquartile range, estimates for cardiovascular mortality were larger for NO_2 than for $PM_{2.5}$ (29% vs. 16%). In the recent review by Hoek, NO_2 effects ranging from 3% to 36% per 10 µg/m^3 have been reported for cardiovascular mortality in European cohorts as well as an effect of 12% for respiratory mortality [5]. All our estimates were rather heterogeneous, with the sole exception of respiratory mortality. In the few studies with bi-pollutant models, the effects of NO_2 and particles (of different sizes) seemed independent.

The comparison of studies carried out in worldwide geographical areas is beset by many challenges. The methods used to assess exposure play an important role. The NO_2 levels observed in the U.S.A., Asia and Europe are very similar. In contrast, $PM_{2.5}$ levels are lower in the U.S.A. than in Europe,

and both are much lower than the Asian levels. The methods used in assessing exposure (such as dispersion modelling, land use regression models and inverse distance weighting), as well as the methods used in pollution monitoring (such as the number and location of fixed monitors as well as the daily sampling intervals) could all influence the quality of the exposure assessment causing a possible differential misclassification. However, since lower levels of $PM_{2.5}$ in the U.S.A. produced effects on total mortality comparable to those observed in Europe, a different toxicity of the fine particle components between areas could also play a role.

The length of the exposure (or modelling) period was not always taken sufficiently into consideration. This is an important drawback when risk estimates for long-term mortality are to be compared. The length of exposure assessment ranged from one [21, 23] to 12 years [15] when reported as measured levels of pollutants, and from 2-40 years when estimated by means of models (mainly dispersion or LUR models). In addition, length of exposure is mostly assessed on the basis of length of residence, assuming that the pollution levels monitored in a shorter time than residence period represents the real exposure of the whole residence period. Only few investigations have addressed the long-term stability of the spatial contrasts in the exposures used in the epidemiological analyses. [35,36]

Latency differs among studies, often not consistently with the study design. Three studies [15, 17, 19] had an overlap between the exposure and the follow-up periods, others started the follow-up in the same year the pollutant exposure was assessed [24,28], though they prolonged the follow-up for up to 30 years. In a few studies [12,20,21,34] exposure was assessed in a few years included within the follow-up or even after the follow-up [22]. Finally, eight studies [16, 18, 23, 25, 26, 27, 32, 33] carried out a follow-up from at least one year after the assessed exposure for up to 6-20 years thereafter. Therefore, the differences in assessing the temporal relationship between exposure and outcome across the studies represent a possible challenge for the interpretation of the results. Doubts could rise also when the exposure has been assessed after starting follow-up, since strong

assumptions were necessary about the length of residence and the corresponding levels of pollutants.

Some methodological choices in carrying out the meta-analysis need to be discussed. The first relevant point for the interpretation of the results is the reliability of the meta-analytic estimates by geographic area. The European and North American estimates for total and cardiovascular mortality, as well as the European estimates for respiratory mortality, are more reliable than Asian estimates because at least four papers are included. However, the heterogeneity across the studies affects the reliability [10,38] of the estimates of total and cardiovascular mortality, in all areas and for both pollutants. The sole exception is the effect of NO_2 on respiratory mortality in both Europe and the U.S.A. (with three studies). It should be noted that the heterogeneity for cardiovascular mortality was strongly reduced when we restricted to studies with a better quality of exposure assessment and evaluating the entire population and not at at-risk groups [37].

A second methodological point for the interpretation of results is related with the pollutant exposure metric for analysing the effects of different pollutants. Using a fixed increase of 10 $\mu g/m^3$ in pollutant levels makes it possible to compare the effects of a single pollutant across different studies and different countries. In contrast, if one wishes to compare the effects of two or more pollutants, one needs another metric which takes into account the different distributions of pollutant levels. A pollutant with a broader exposure distribution will show smaller effect estimates per unit increase than a pollutant with a less wide exposure distribution in the same population. The additional meta-analyses we performed using the IQRs of NO_2 and $PM_{2.5}$ as the exposure metric show that the NO_2 effects on mortality are larger than those of $PM_{2.5}$, at least for total and cardiovascular mortality.

There is obviously a need for caution in drawing conclusions about the independence of NO_2 effects from $PM_{2.5}$ effects from the multi-pollutant models. First, not all countries were included (e.g. Asia was less represented in this analysis). Second, the high correlation between NO_2 and $PM_{2.5}$ (around

0.7-0.8) in these studies still suggests the possibility that the NO_2 effects could be due in part to confounding from PM. Third, uncertainty is also due to the limited number of bi-pollutant estimates available. It should be noted, however, that only minimal difference between the single-pollutant and the multi-pollutant results for NO_2 were found.

Finally, though this study being a meta-analysis, cannot respond exhaustively to the causality question about NO_2, it contributes to the scientific debate on this topic because it assesses the respective role of NO_2 and $PM_{2.5}$ as they emerge from prospective studies [38] and provides a pooled effect estimate to be used for future impact health assessment.

In conclusion, the magnitude of the long-term effects of nitrogen dioxide on mortality is at least as important as that of $PM_{2.5}$. These results hold when using both 10 $\mu g/m^3$ or IQR as the metric of choice. The results of the multi-pollutant models suggest that the role of nitrogen dioxide is independent of that of particles. All these elements may be useful when discussing the causality issue for NO_2. Moreover, NO_2 is the only widely regulated pollutant (apart from carbon monoxide) that indicates exposure to combustion pollution. Health impact assessments relying only on $PM_{2.5}$, and not considering NO_2, would be neglecting an important source of the adverse effects of today's pollution mixture.

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Figures titles and legends

FIGURE 1. RELATIVE RISKS OF NATURAL MORTALITY WITH INCREASING CHRONIC EXPOSURE TO NO_2.

SUBTITLE OF FOREST PLOT = RR of natural mortality for 10µg/m^3 NO_2 increase

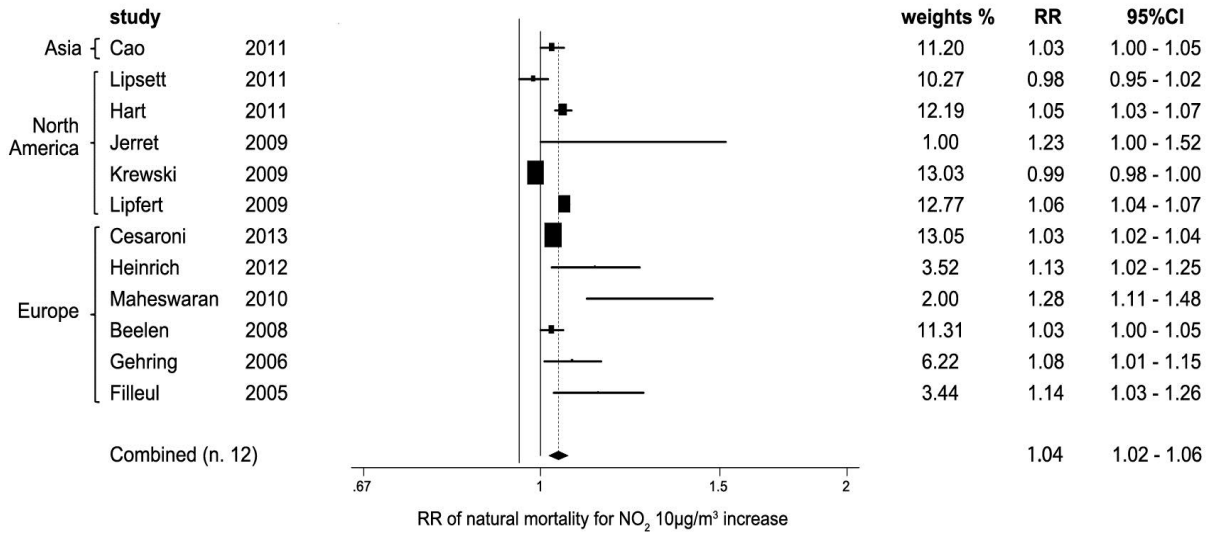
FIGURE 2. RELATIVE RISKS OF CARDIOVASCULAR MORTALITY WITH INCREASING CHRONIC EXPOSURE TO NO_2.

SUBTITLE OF FOREST PLOT = RR of cardiovascular mortality for 10µg/m^3 NO_2 increase

FIGURE 3. RELATIVE RISKS OF RESPIRATORY MORTALITY WITH INCREASING CHRONIC EXPOSURE TO NO_2.

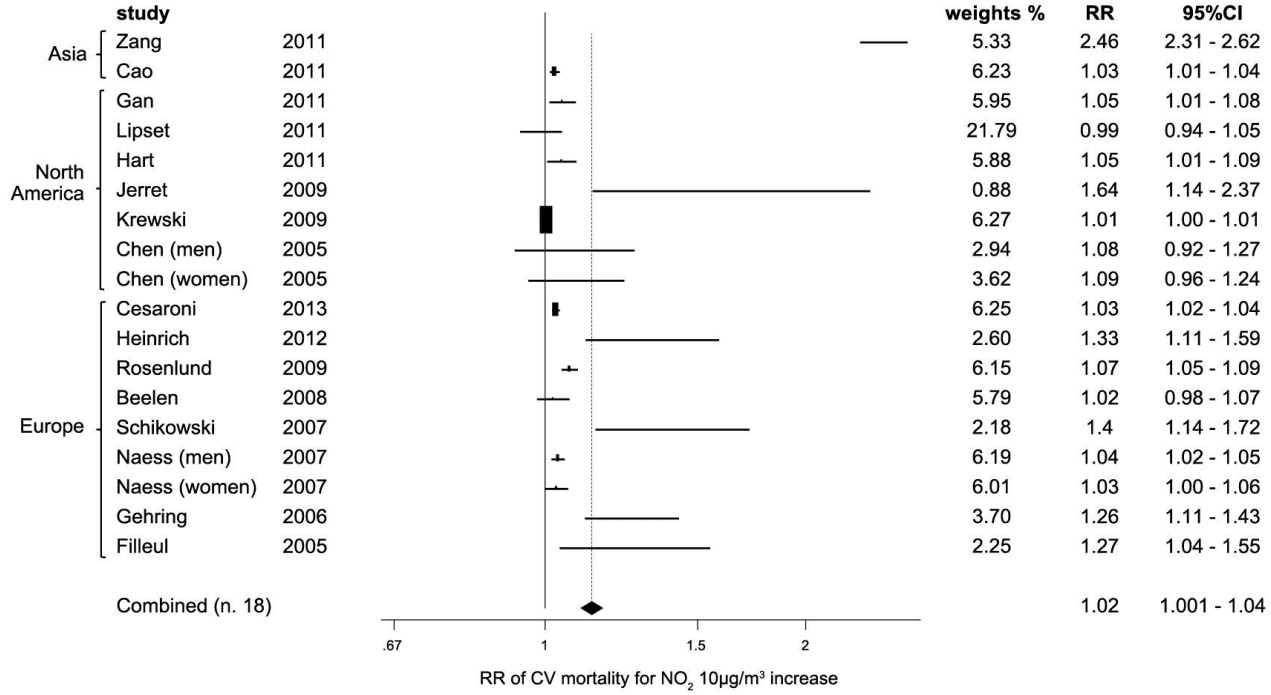
SUBTITLE OF FOREST PLOT = RR of respiratory mortality for 10µg/m^3 NO_2 increase

Figure 1



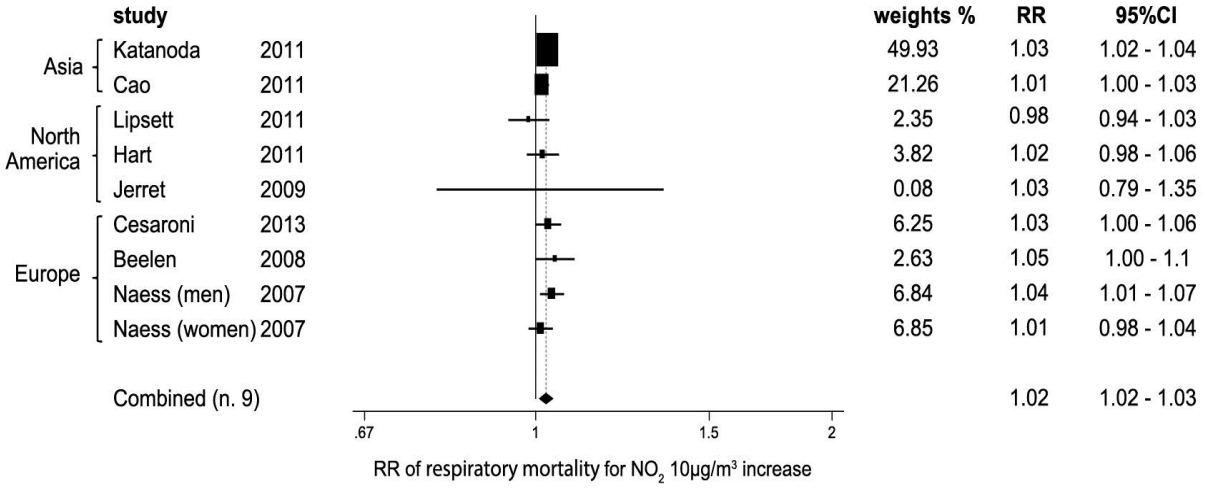
Test for heterogeneity: $\chi^2 = 102.28$ dl = 11
p = 0.001 $I^2 = 89\%$
Test for overall effect: z = 3.632 p = 0.001

Figure 2



Test for heterogeneity: $\chi^2 = 853.532$ dl = 17
p = 0.001 $I^2 = 98\%$
Test for overall effect: z = 6.010 p = 0.001

Figure 3



Test for heterogeneity: $\chi^2 = 7.383$ dl = 8
p = 0.496 $I^2 = 0\%$
Test for overall effect: z = 6.362 p = 0.001

Table 1. Pooled* effects of NO_2 and $PM_{2.5}$ (per 10µg/m^3) on natural and cause-specific mortality by region of the world

	Total or natural mortality					Cardiovascular mortality					Respiratory mortality				
	N studies	RR	95% CIs		I^2	N studies	RR	95% CIs		I^2	N studies	RR	95% CIs		I^2
NO_2 (10µg/m^3)															
All countries	12	1.041	1.019	1.064	89%	18§°	1.133	1.088	1.180	98%	9°# ++	1.024	1.017	1.032	0%
Asia	1	1.020	1.000	1.030		2	1.588	0.675	3.740	99%	2	1.023	1.010	1.036	47%
America	5	1.027	0.987	1.069	95%	7	1.031	0.999	1.065	67%	**3**	**1.005**	**0.975**	**1.035**	**0%**
Europe	6	1.066	1.029	1.104	72%	9	1.059	1.032	1.086	79%	**4**	**1.029**	**1.013**	**1.045**	**0%**
$PM_{2.5}$ (10µg/m^3)															
All countries	11+	1.045	1.007	1.088	87%	17§°+	1.196	1.091	1.31	98%	8°# ++,+	1.050	1.009	1.094	61%
Asia	1	1.000	0.980	1.020		2	1.380	0.761	2.502	99%	2	1.034	0.943	1.134	50%
America	4	1.047	1.035	1.106	83%	6	1.047	0.992	1.106	71%	2	1.046	0.990	1.104	0%
Europe	6	1.071	1.021	1.124	62%	9	1.188	1.091	1.295	42%	**4**	**1.075**	**1.009**	**1.146**	**40%**

* results from random-effects meta-analyses § two studies (one from America [33] and one from Europe [28) were included twice with separate groups of men and women. ° Four studies (one from America [23] and three from Europe [13, 32, 34]) estimated cardiopulmonary mortality as a whole. They were included in the pooled CV mortality estimate and excluded from the pooled respiratory mortality estimate. # One study from China [14] was excluded because it estimated lung cancer together with respiratory mortality. '++ one study from Europe [28] was included twice with separate estimates for men and women. + one study from the USA [22] cannot be included because it did not show the PM effect estimates.

Table 2. Pooled* effects of NO2 and PM (10 µg/m^3 and IQR µg/m^3) on natural and specific-cause mortality

	total or natural mortality					cardio-vascular mortality					respiratory mortality				
	N studies	RR	95% CIs		I^2	N studies	RR	95% CIs		I^2	N studies	RR	95% CIs		I^2
NO_2 (10µg/m3)	10**	1.040	1.015	1.065	91%	**NO_2 (10µg/m3)**	15^§°	1.152	1.094	1.213	98%	**NO_2 (10µg/m3)**	7# ++	1.024	1.010	1.038	0%
NO_2 (14.6 µg/m3)		1.059	1.022	1.096		**NO_2 (17.9 µg/m3)**		1.288	1.174	1.413		**NO_2 (15.0 µg/m3)**		1.036	1.015	1.058	
$PM_{2.5}$ (10 µg/m3)	9**+	1.050	1.024	1.078	78%	**$PM_{2.5}$ (10 µg/m3)**	14^§°+	1.228	1.084	1.39	98%	**$PM_{2.5}$ (10 µg/m3)**	6# ++,+	1.062	1.022	1.104	11%
$PM_{2.5}$ (5.8 µg/m3)		1.029	1.014	1.045		**$PM_{2.5}$ (7.0 µg/m3)**		1.155	1.058	1.259		**$PM_{2.5}$ (7.0 µg/m3)**		1.043	1.015	1.072	

* random estimates. **two studies (Cao and Filleul) were excluded as they did not give IQR estimates or dispersion parameters to estimate them.

^ three studies (Cao, Rosenlund and Filleul) were excluded as they did not give IQR estimates or dispersion parameters to estimate them.

§ two studies (Naess, 2007 and Chen 2005) were included twice with separate groups of men and women.

° Two studies from Europe (Heinrich 2012 and Gehring 2006) estimated cardio-pulmunary mortality as a whole. They were included in the pooled CV mortality estimate and excluded from respiratory mortality pooled estimate.

One study (from China (Dong, 2011)) was not included because it estimated lung cancer together with respiratory mortality. Three studies (Katanoda, Cao and Filleul) were excluded as they did not give IQR or dispersion parameters to estimate them.

+ one study from the USA (Jerret, 2009) cannot be included as it did not show the PM effect estimates.

++ one study from Europe (Naess, 2007) was included twice with independent groups of men and women.