ORIGINAL SCIENTIFIC PAPER

SHORT-TERM EFFECTS OF AMBIENT NITROGEN OXIDES ON THE NUMBER OF EMERGENCY ASTHMA CASES IN ZAGREB, CROATIA

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The paper describes an investigation of short-term effects of NO_2 concentrations in the air on the number of emergency room visits caused by respiratory impairments, particularly asthma in adults and children. The data were collected from clinical emergency room records from July 1, 1994 to December 31, 1995. Concurrently, readings of average weekly concentrations of NO_2 (μg/m^3), average weekly temperature (°C), air pressure (kPa), and relative humidity (%) were registered. Trend and seasonality effects were estimated by the locally weighted regression (LOESS). After standardising for trend, seasonality, and meteorological conditions, the number of cases was regressed on weekly NO_2 concentration, including the current and the previous week concentrations and autocorrelated residual. The weekly average NO_2 concentrations were significantly associated with the number of emergency asthma cases for children and adults and with the total number of emergency respiratory cases in children, but not in adults. The results suggest that health effects of NO_2 on risk groups can be detected even in moderately polluted environments. The effect is more pronounced in children.

Key words:
air pollution, autocorrelation, Poisson regression, respiratory diseases, time series

Nitrogen oxides are notorious for their supraambiental toxicity, chemical and physical characteristics (low solubility, density higher than that of the air, perceptible odour only in high concentrations), as well as the clinical course of poisoning (biphasic nature, delayed effect, pulmonary oedema, acute respiratory distress syndrome (ARDS), pneumonitis, haematotoxicity) (1). However, their endogenous generation and local bioactivity (induction of free radicals, enhancement of airway inflammation, possible beneficial effect on alveolar microenvironment, effects of synthetase and elastase) are less known (2, 3). Even in subtoxic concentrations they can adversely affect human health (4). This makes nitrogen oxides an intriguing object of study in respiratory pathophysiology. A great variety of human activities lead to accumulation of nitrogen oxides and other pollutants in the troposphere. In the era of massive energy consumption, effects of these chemically very active components and other products of combustion and pyrolysis, have become increasingly relevant for environmental epidemiology. Relying on the known effects, the World Health Organisation (WHO) and local legislatures have set recommended and limit concentration values for these pollutants. In industrial and urban areas indoor and outdoor concentrations of nitrogen oxides and NO_2 in particular often exceed these concentrations. Monitoring of outdoor NO_2 concentrations in Zagreb, Croatia, began in 1994 (5, 6).

Incidence of chronic obstructive respiratory disease, and especially asthma has been increasing in the urban areas of developed countries (7). There are few studies which took into account combined (mixed) air pollutants, their synergism, biometeorological and cosmobiological factors, as well as pollen concentrations and organic and inorganic suspended particles (8). One available research methodology involves daily pollution and frequency of emergency room visits using the time-series analysis. Several studies have applied this method in large US and European cities with various levels of NO_2 concentrations and climate. However, they reached no definite conclusion with regard to the effects of ambient NO_2 on asthma incidence and aggravation of symptoms (9–15). Wishing to elucidate these results, we collected and compared records of daily emergency respiratory cases, particularly those of asthma, with measurements of ambient NO_2 concentrations and basic weather parameters in Zagreb, Croatia.

SUBJECTS AND METHODS

Location

Zagreb has spread at the western edge of the Pannonian valley (15° 39' E longitude, 45° 49' N latitude), along the foot of the Medvjednica mountain, 122–160 m above the sea level. The climate is temperate. The area of the inner city is 640 km^2. According to the census of 1991 the city counts 1,162,043 inhabitants (329,452 below 19 years of age). The population density varies between high (ca 45,000/km^2) and low (two-digit figure per km^2) in the suburbs. There are more than 220,000 motor vehicles. Natural gas consumption in approximately 300,000 households and power stations is 650 million m^3 a year. Zagreb is an industrial centre with chemical, machinery, and food industry.

Study design

Clinical Hospital for Pulmonary Diseases in Zagreb admits about 65% of adult (age 19 or over) respiratory emergencies. Children (age below 19) are separately admitted to the Children Hospital for Pulmonary Diseases. Both hospitals have emergency wards. From July 1, 1994 to December 31, 1995 (549 days altogether), we recorded daily frequencies of emergency room admissions to these hospitals due to bronchial asthma and all respiratory causes. The asthma cases were clinically categorised in degrees II, III, and IV (16). In all respiratory cases asthma participated with 6.4% in adults and 24.3% in children.

Meteorological observations

Simultaneously, Hydrometeorological Institute of Croatia supplied data on average temperature, air pressure, and relative humidity recorded at the meteorological monitoring site at the outer rim of the inner city every day for the entire duration of the study.

Air pollution measurements

Nitrogen oxides were measured using the colorimetric method (17, 18) and expressed as NO_2. Recordings refer to the daily averages. The monitoring site was in the centre of the city, and measurements were taken at 1.8 m above the street level. Figure (map) 1 shows the city with locations of the meteorological and air pollution monitoring sites and of the two hospitals

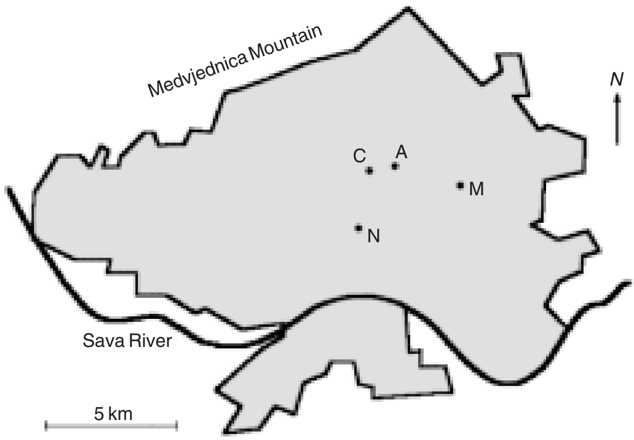

Figure 1 *City map with positions of the monitoring sites (M – meteorological, N – NO_2) and hospitals (C – children, A – adults).*

Statistical analysis

For more than a half of the studied period there was not a single emergency admission for asthma in children. Therefore, instead of analysing the daily time series, we decided to analyse cumulative weekly counts of emergency cases due to asthma and all causes for both children and adults. The weekly averages were used for NO_2 concentrations and meteorological observations. We have basically followed the »Air

Pollution Health Effects – A European Approach« (APHEA) protocol for data analysis (19, 20). Seasonality was assessed using local regression LOESS smoothing (21) with normal weights and locally quadratic regression on log transformed counts and original meteorological and NO_2 time series. LOESS regression is a robust regression method where fitting is performed in a small neighbourhood (»window«) around each data point. The size of the »window« controls how close the fitted values explain the observed values. Various criteria may be used for choosing the optimal bandwidth. The choice of the bandwidth in this study was based on the spectral analysis of residuals, aiming to reduce autocorrelation and partial autocorrelation for lags longer than a month (4 weeks). Our goal was not to filter out the total periodic variation, but only the part that was reasonably believed to result from ommited confounders. We chose the largest bandwidth for which partial autocorrelations of residuals for periods longer than 4 weeks were negligible. The relationship between the emergency room counts and the meteorological variables was assessed using linear and quadratic regression on both original meteorological variables and their LOESS residuals. Only terms significant at the $P<0.05$ level were retained in the final model. Such a regression model explains association between deviations of the outcome (emergency room visits) and predictors (NO_2, etc.) from their respective usual seasonal behaviour (the LOESS prediction). Statistical analyses were done in SAS System 6.12, using the SAS/INSIGHT® module and procedures SPECTRA and AUTOREG (22–24).

RESULTS

Table 1 gives the descriptive statistics for both daily and weekly data. Distributions of the counts of emergency cases were positively skewed (means larger than medians and ranges between the median and the maximum wider than between the minimum and the median). The skewness was less pronounced for the total respiratory counts in adults and more pronounced for asthma. The number of all respiratory emergency cases is essentially a sum of counts for various respiratory diagnoses, so we believed it would be closer to a normal distribution. Variances are consistently higher than means, reflecting extra Poisson variability. The NO_2 concentrations were within the limit, but above the recommended values for the mean and the 98th percentile. They were comparable to those found in less polluted European cities (14).

Figure 2 shows the plot of the daily average concentrations of NO_2 and their weekly means (together with the limit values for annual mean and the 98th percentile of daily averages). For the entire monitoring period only twice (in winter 1995) did the recordings show the daily NO_2 average of over 120 μg/m^3. As we expected, there were no seasonal variations in the NO_2 level over a year.

Figure 3 shows weekly counts of hospital emergency room visits due to asthma and other respiratory causes for children and adults with the superimposed LOESS smooths. The LOESS smooths were fitted to the log transformed counts, and retransformed to the original scale for plotting. The bandwidth was 13 weeks for the asthma counts and 16 weeks for the total respiratory cases. These bandwidths removed all autocorrelations for lags longer than one month. The observed asthma

Table 1 *Descriptive statistics for daily and weekly counts of emergency room visits, and meteorological and environmental data*

Variable	Daily							Weekly						
	Mean	Var	Min	25%	50%	75%	Max	Mean	Var	Min	25%	50%	75%	Max
Asthma[C]	1.14	3.00	0	0	0	2	12	8.0	31.0	0	3	7	11	24
Asthma[A]	1.36	1.88	0	0	1	2	7	9.5	23.5	1	6	9	13	22
Total respiratory[C]	4.97	10.15	0	3	4	7	25	34.8	183.1	11	26	33	45	73
Total respiratory[A]	19.65	77.70	0	13	20	27	43	137.7	445.6	80	124	135	153	193
NO_2 (µg/m^3)	45.10	340.10	5.5	33.6	41.5	53.8	169.8	45.1	83.4	24.5	38.4	44.9	51.3	64.9
Temperature (°C)	11.70	63.40	–6.6	5.6	11.6	18.5	26.6	11.8	57.1	–4.5	5.7	11.9	18.7	25.2
Rel. humidity (%)	74.10	140.90	35.0	66	75	83	98	74.1	70.8	58.4	67	74	81	89
Air pressure (hPa)	1002	49.30	979	998	1002	1006	1023	1002	24.5	991	999	1002	1004	1015

[C] children, [A] adults

daily NO_2 recommended C_{avg}=30, C_{98}=60
limit C_{avg}=50, C_{98}=120
our C_{98}=95.2

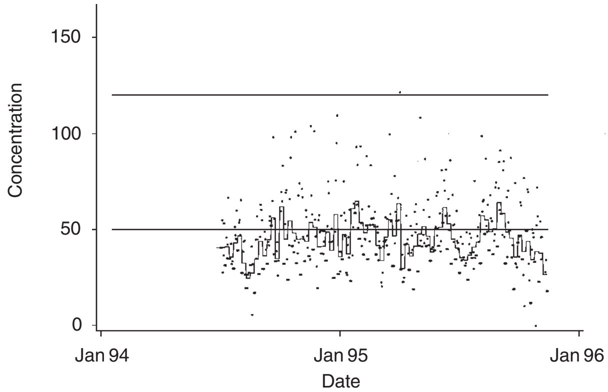

Figure 2 *Daily NO_2 concentrations (in µg/m^3) with the weekly mean, limit annual mean and the 98th percentile superimposed.*

counts for adults showed a negative trend. This trend might be due to low frequency of asthma cases for the period April–September 1995. Counts of total respiratory hospital emergencies for adults, as well as both time-series for children, showed a slight positive trend. However, the NO_2 and meteorological series did not show any trend. The trends for emergency room visits may be specific for the analysed hospitals or may reflect administrative changes in the distribution of competent hospitals for certain diseases. The trends did not reflect short-term effects of air pollution, and the fitting of the LOESS curves made it possible to exclude them from further analyses.

As there were no significant associations of log count residuals with the meteorological variables, weather variables were not included in the model.

Table 2 shows the results of the final autoregressive linear regression. The final regression included only the significant terms. NO_2 was significantly associated with the asthma emergency counts for both children and adults. The weekly concentrations of NO_2 were significantly associated with the weekly counts of all respiratory emergency cases for children, but not for adults.

When applying the regression analysis to the time-series data it is necessary to test possible autocorrelation in the error term. Both asthma and the total respiratory counts underwent preliminary analyses using the autoregressive error model of order two and including the NO_2 data for the current and the previous week. Only for the total respiratory count for children was there a significant autocorrelation for the error term ($P=0.0026$). As expected, the lagged NO_2 term was not significant. If it exists, the delay in the effect of NO_2 concentrations in the air on the number of emergency cases must be shorter than a week. The relative risks (also reported in Table 2) appear to be slightly higher in children than in adults, and for asthma than for all respiratory causes.

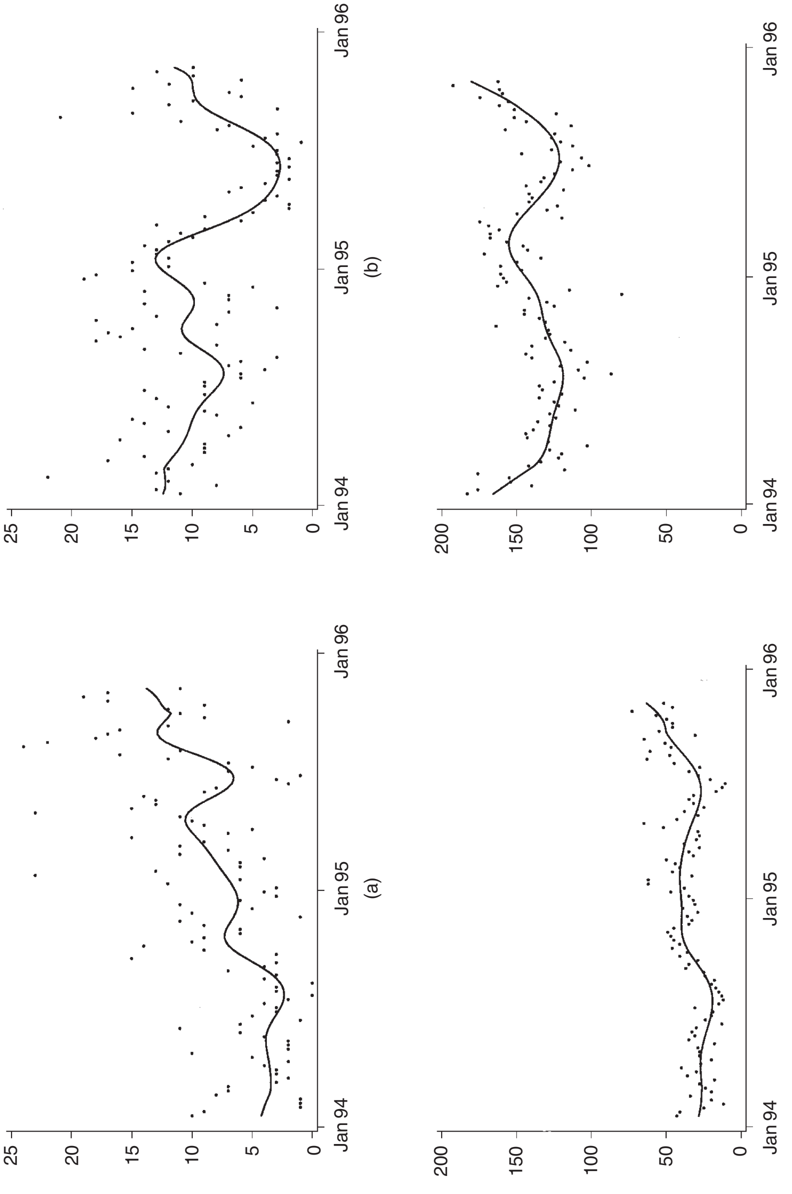

Figure 3 *Weekly counts of emergency room visits due to asthma (a – children, b – adults) and all respiratory causes (c – children, d – adults) with LOESS smooths superimposed.*

Table 2 *Results of autoregressive linear regression of the detrended and seasonally adjusted weekly log counts of respiratory emergency cases*

	coeff.	SE	RR	Confidence limits lower	Confidence limits upper	P ≈
Asthma[C]						
Intercept	–0.591	0.285				0.042*
NO_2 (10 μg/m^3)	0.129	0.062	1.137	1.005	1.287	0.041*
Asthma[A]						
Intercept	–0.425	0.212				0.049*
NO_2 (10 μg/m^3)	0.093	0.046	1.098	1.002	1.203	0.046*
Total respiratory[C]						
A(1)	–0.349	0.112				0.003**
Intercept	–0.299	0.148				0.046*
NO_2 (10 μg/m^3)	0.065	0.032	1.067	1.002	1.136	0.043*
Total respiratory[A]						
Intercept	–0.086	0.068				0.210
NO_2 (10 μg/m^3)	0.019	0.015	1.019	0.990	1.049	0.204

[C] children, [A] adults
* P<0.05
** P<0.01

Table 3 *Number of cases in 1995 possibly attributable to NO_2*

Avg. weekly NO_2 concentration	Asthma		All respiratory causes	
	Children	Adults	Children	Adults
>30 μg/m^3	106	62	214	225
>50 μg/m^3	17	10	33	31

The models reported in Table 2, the observed weekly counts of emergency cases due to asthma, all respiratory causes, and the concentrations of NO_2, served to estimate the number of emergency cases possibly attributable to NO_2 concentrations exceeding any chosen limit value. Table 3 shows these numbers for the year 1995, setting the limit for NO_2 concentration at the annual recommended and limit average. The number of emergency cases of asthma in children possibly attributable to the average weekly concentrations exceeding both 30 μg/m^3 and 50 μg/m^3 is larger than that for adults. With regards to all respiratory causes, the numbers for children and adults are comparable.

DISCUSSION

The research of the effects of subtoxic concentrations of nitrogen oxides in the ambient and indoor air on specific risk groups has not given a conclusive answer. This is because the studies largely differed in designs and preferred to focus on the acute, subacute, or chronic effects. There are difficulties in identifying the influence of NO_x on the clinical endpoints with respect to a single individual. We know little about the interactions with endogenous NO_x on the molecular level, the NO_x interference with the enzymatic system, and the effects of climate and individual genetic traits, as well as habits and occupational exposure. This study does not undertake to give a definite answer. We may hope to improve the understanding of the relationship between the aggravation of asthma symptoms and the short-term dynamics of nitrogen oxides in the atmosphere.

One of the drawbacks of the analysis of observational time-series data is its inability to take into account the differential effect of air pollution on subgroups of the population of interest, for example on patients with endogenous or exogenous type of asthma or patients undergoing corticosteroid therapy (25). In effect, what we estimate is the »weighted average« of the effect air pollution may have on different subgroups of the target group. Some authors deny any relationship between the sensitivity to air pollution and the severity of asthma (26). Even if there is such sensitivity, the results (and their validity) would still have to rely on the assumption that the composition of the target group does not change in relation to these confounding factors during the analysed period. On the other hand, proper seasonal adjustment allows control of confounders, that is, unknown or omitted factors which show long-term seasonality. A similar argument can be used for the coverage of the study. The fact that only about 65% of adult cases have been covered may influence the results only if the proportion of visits to emergency rooms of the hospitals included in the study vs. those not included varies with the air pollution levels. Such relationship did not seem reasonable to assume.

We have not been able to find any literature on weekly time-series analysis similar to ours. Some panel studies investigated the relationship between the weekly incidence and/or prevalence of respiratory symptoms and air pollution (27). However, their results can not be directly compared with ours. Observational time-series analyses usually focus on daily (28), and sometimes on monthly (29) or yearly data (5). We had to work on weekly counts because emergency asthma cases in children were scarce. One should bear in mind this difference when she/he compares our results with such results as those of the APHEA project which deals with daily data. For the weekly data we expected to obtain larger effect size than the one we would have obtained for the same period had we analysed the daily data. Since the effect of exposure to NO_2 is delayed, a short-term (e.g. 1-day) increase in its concentration could produce an effect that would spread over several days. The corresponding weekly average concentration will show a smaller increase, but the increase in the weekly health effect will not be proportionally smaller. The relative risk in our estimate is indeed larger than that in the APHEA. However, as it corresponds to weekly averages and has rather wide confidence intervals, it seems that the effect of NO_2 on asthmatic patients in Zagreb is comparable to that of other European cities.

We have found much larger effect size for all respiratory diseases in children than in adults. This finding may be the consequence of different proportion of asthma cases in the respiratory emergency cases for children (24.3%) and adults (6.4%). This may also imply that sensitivity to air pollution decreases with maturation and growth of the lungs.

While the analyses of the daily time series find the delay in the effect of NO_2 (and include lags of up to three days in the model), we have found that the NO_2 concentrations of the previous week do not improve the fit of our model. Previous results of daily time series and reports on NO_2 poisoning suggest that the delay, if there is one, must be shorter than 7 days.

Asthma morbidity in Zagreb is not excessive. There have been no positive trends in either morbidity or mortality (30). The small number of emergency cases possibly attributable to NO_2 does not seem to warrant *onus asylum* or extensive cost-benefit analyses for the public health. However, it does indicate the necessity to provide special care to the asthma patients, especially to children, and to broadcast in media or make publicly available daily updated information on the air pollution.

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REFERENCES

1. *Clayton GD, Clayton FE, eds.* Patty's industrial hygiene and toxicology. Toxicology. 3rd edition. New York, NY: John Wiley & Sons, 1994.
2. *Moshenin V*. Human exposure to oxides of nitrogen at ambient and supra-ambient concentration. Toxicology 1994;89:301–12.
3. *Jorres R, Nowak D, Grimminger F, et al*. The effect of 1 ppm nitrogen dioxide on bronchoalveolar lavage cells and inflammatory mediators in normal and asthmatic subjects. Eur Respir J 1995;8:416–24.
4. *Strand V, Svartengren M, Rak S, et al*. Repeated exposure to an ambient level NO_2 enhances asthmatic response to a nonsymptomatic allergen dose. Eur Respir J 1998;12:6–12.
5. *Šega K, Fugaš M, Kalinić N*. Indoor concentration levels of selected pollutants and household characteristics. J Exposure Analysis Environ Epidemiol 1992;2:477–85.
6. *Šarić M, ed*. Environmental exposure assessment and health effect studies in the Republic of Croatia. Zagreb: Croatian Academy of Sciences and Arts, 1996.
7. *Burney PGJ*. Epidemiology. Br Med Bull 1992;48:10–22.
8. *Anto JM, Sunyer J*. Nitrogen dioxide and allergic asthma: Starting to clarify an obscure association. Lancet 1995;345:402–4.
9. *Katsouyanni K, Zmirou D, Spix C, et al*. Short-term effects of air pollution on health: A European approach using epidemiological time-series data. Eur Respir J 1995;8:1030–8.
10. *Castellsague J, Sunyer J, Anto JM*. Short-term association between air pollution and emergency room visits for asthma in Barcelona. Thorax 1995;50:1051–6.
11. *Anderson HR, Atkinson R, Limb E, et al*. Asthma epidemics and air pollution: Epidemic of asthma was not associated with episode of air pollution. Br Med J 1996;312:1606–10.

12. *Dab W, Medina S, Quenel P, et al*. Short term respiratory health effects of ambient air pollution: Results of the APHEA project in Paris. J Epidemiol Community Health 1996;50(suppl 1):S42–6.
13. *Ponce de Leon A, Anderson HR, Bland JM, et al*. Effects of air pollution on daily hospital admissions for respiratory disease in London between 1987–88 and 1991–92. J Epidemiol Community Health 1996;50(suppl 1):S63–70.
14. *Ponka A, Vartinen M*. Asthma and ambient air pollution in Helsinki. J Epidemiol Community Health 1996;50(suppl 1):S59–62.
15. *Sunyer J, Spix C, Quenel P, et al*. Urban air pollution and emergency admissions for asthma in four European cities: The APHEA project. Thorax 1997;52:760–3.
16. *National Institutes of Health (NIH)*. Asthma management and prevention: A practical guide for public health officials and health care professionals. <http://www.ginasthma.com/gina/practical/practical.htm> (9 Nov 1998)
17. *Pauković R*. Measuring NO_2 in the air with passive samplers. Arh hig rada toksikol 1987;38:17–23 (in Croatian).
18. *Hršak J*. NO_2 concentrations in Zagreb air. In: Steinberger Y, ed. Inaternational Conference of the Israeli Society for Ecology and Environmental Quality Sciences »Preservation of Our World in the Wake of Change«. Proceedings. Jerusalem: ISEEQS, 1996;6:920–4.
19. *Katsouyanni K, Schwartz J, Spix C, et al*. Short term effects of air pollution on health: A European approach using epidemiological time series data: The APHEA protocol. J Epidemiol Community Health 1996;50 (suppl 1):S12–8.
20. *Schwartz J, Spix C, Touloumi G, et al*. Methodological issues in studies of air pollution and daily counts of deaths or hospital admissions. J Epidemiol Community Health 1996;50(suppl 1):S3–11.
21. *Clevland WS, Devlin SJ*. Locally weighted regression and smoothing scatterplots. JASA 1988;83:596–610.
22. *SAS Institute Inc*. SAS/STAT® user's guide. 4th edition. Cary: SAS Institute Inc., 1989.
23. *SAS Institute Inc*. SAS/ETS® user's guide. 1st edition. Cary: SAS Institute Inc., 1988.
24. *SAS Institute Inc*. SAS/INSIGHT® user's guide. 3rd edition. Cary: SAS Institute Inc., 1995.
25. *Tanaka H, Honma S, Nishi M, et al*. Acid fog and hospital visits for asthma – an epidemiological study. Eur Respir J 1998;11:1301– 6.
26. *Hiltermann TJ, Stolk J, van der Zee SC, et al*. Asthma severity and susceptibility to air pollution. Eur Respir J 1998;11:686–93.
27. *Rudnai P, Agocs MM, Etzel RA*. Environmental respiratory disease surveillance: A Hungarian example. MMWR CDC Surveill Summ 1992;41:753.
28. *Moolgavkar SH, Luebeck EG, Anderson EL*. Air pollution and hospital admissions for respiratory causes in Minneapolis–St. Paul and Birnmingham. Epidemiology 1997;8:364–70.
29. *Abbey DE, Colome SD, Mills PK, et al*. Chronic disease associated with long-term concentrations of nitrogen dioxide J Expo Anal Environ Epidemiol 1993;3:181–202.
30. *Žulj-Juvančić V*. Health statistics in Zagreb, 1993–1995. Morbidity and mortality of chronic noncommunicable and communicable diseases. 1st edition. Zagreb: Institute for Public Health of the City of Zagreb, 1998 (in Croatian).

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Sažetak

UČINAK DUŠIKOVA DIOKSIDA NA BROJ POSJETA HITNIM SLUŽBAMA

Analizirani su kratkoročni učinci prosječnih tjednih koncentracija NO_2 u zraku na broj posjeta hitnim službama zbog pogoršanja stanja bolesti respiratornog sustava, posebno astme u odraslih i djece. Podaci su prikupljeni iz knjiga hitnih službi u razdoblju od 1. srpnja 1994. do 31. prosinca 1995. Bilježene su prosječne tjedne koncentracije NO_2 (μg/m^3), prosječne tjedne temperature (°C), tlak zraka (kPa) i relativna vlaga (%). Trend i sezonalnost procijenjeni su lokalnom ponderiranom regresijom (LOESS). Nakon isključivanja učinka trenda, sezonalnosti i meteoroloških uvjeta, broj hitnih slučajeva modeliran je u ovisnosti o tjednim koncentracijama NO_2, uključujući koncentracije za tekući i prethodni tjedan te autokorelaciju reziduala. Prosječne tjedne koncentracije NO_2 statistički su značajno povezane s brojem hitnih slučajeva astme u djece i odraslih s ukupnim brojem hitnih respiratornih slučajeva u djece, ali ne i u odraslih. Rezultati pokazuju da i pri umjereno visokim koncentracijama NO_2 možemo opaziti učinak onečišćenja na zdravlje, pogotovo u osoba iz skupina povišenog rizika. Taj utjecaj na zdravlje osobito je naglašen u djece.

Ključne riječi:

autokorelacija, onečišćenje zraka, Poissonova regresija, respiratorne bolesti, vremenske serije

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