Urinary nickel and plasma cortisol in workers exposed to urban stressors

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Summary

Context: **urban pollution is associated with various diseases; nickel (Ni) is a toxic agent in urban air.**
Objective: **verify whether the exposure to low Ni doses may affect plasma cortisol values of outdoor workers.**
Materials and methods: **374 workers underwent urinary Ni and plasma cortisol sampling.**
The Mann-Whitney U test for two mode variables and the Kruskal Wallis test for the variables in more than two modes were performed on the total sample.
Pearson’s correlation coefficient (p two-tailed) among the parameters was evaluated both in the total sample and after the stratification by gender, smoking habit and job. The multiple linear regression was performed after taking account of the major confounding factors on total sample and on the subcategories.
Results: **we found a significant inverse correlation between Ni and cortisol.**
Discussion and conclusions: **occupational exposure to low Ni doses may influence cortisol values in workers exposed to urban pollution.**

KEY WORDS: outdoor workers, biological monitoring, personal air sampling, endocrine disruptors.

Introduction

Nickel (Ni), together with its compounds, belongs to the 33 toxic agents the Environmental Protection Agency (EPA) identified in urban air (1). The Ni present in urban air originates from natural sources (such as volcanoes and forest fires) and from anthropogenic emissions (such as processes of extraction and refining of Ni, coal combustion, vehicular traffic, domestic heating, incineration of waste); it is also present as an additive in unleaded gasoline, in catalytic converters, in cigarette smoke, in paints, solvents and some pesticides. It is also present in more than 3.000 different alloys used in the manufacture of kitchen utensils, batteries, metal coins and jewelry (2). The main routes of human exposure to Ni are through inhalation, ingestion of contaminated food and drinks as well as through dermal contact.
Irritative and allergenic effects (contact dermatitis) are reported as the major toxic effects of Ni exposure. Several Ni compounds may have more serious effects on the respiratory system: chronic bronchitis, emphysema, pulmonary fibrosis have been observed in nickel welders and foundry workers (2). The International Agency for Research on Cancer (IARC) (3) classified metallic nickel in group 2B (possibly carcinogenic to humans) and nickel compounds in group 1 (carcinogenic to humans); its carcinogenicity seems to come from the refining processes (where tetracarbonyl-nickel is used); carcinogenic effects are mainly localized in the nasal sinus and lungs (3).
As to the interactions between Ni and the endocrine system, several *in vivo* and *in vitro* studies show that Ni affects pancreatic hormones and the reproductive system (4-8); a research reports the presence of a significant inverse correlation between Ni and cortisol in the subjects working with cleaning tasks of the coast following the environmental disaster caused by the accident of the “Prestige” in 2002 (9); yet, at the moment there are no studies showing the relationship between occupational exposure to Ni and cortisol values.

Urinary Ni is the most common index of recent exposure to metallic Ni and its compounds (10-12). In fact, Ni has no cumulative toxic effect and the absorbed dose is excreted almost completely with urine, therefore Ni absorption in exposed workers can be easily detected by biological monitoring of urinary Ni, which represents the best indicator of internal dose for continuous occupational exposure (10-12): given the good correlation between Ni concentrations in the air and in the urines of exposed subjects, urinary Ni appears to be the most suitable test for the evaluation of professional exposure to Ni (13).
After exposure termination, urinary Ni levels can gradually return within normal values (10-12).
The aim of our study is to verify whether, in outdoor workers exposed to urban pollutants, the exposure to low Ni doses in urban air may have some effects on plasma cortisol values.

Materials and methods

Studied population

The research was carried out on a sample of 374 outdoor workers, employed in the Municipal Police of a large Italian city, selecting the 374 workers from different areas of the city as follows: we divided the city into 8 areas considered to be representative of traffic and air quality, and we selected 46 workers from each area (25 traffic policemen, 16 drivers, 5 workers with other outdoor duties); only for the central, busiest area we selected 52 employees (27 traffic control officers, 18 drivers, 7 workers with other outdoor duties).
The traffic control officers were in charge of controlling the flow of vehicles in roads and areas with high and medium traffic intensity, as well as monitoring and regulating traffic at road junctions, parking areas and traffic-limited areas.
The drivers were assigned to traffic control and specific interventions in case of road accidents and other activities including driving cars as driver or as “patrol second”.
The officers with other outdoor activities were assigned to various tasks including assistance to disadvantaged or marginalized people, outdoor building activities, outdoor activities in the field of Judicial Police, Environmental Police, etc.
Most of these activities were carried out outdoors, except for police drivers who spent at least 80% of their working time in cars (7 hours a day for at least 5 days a week).
All workers were monitored once during their morning shift (07:00-14:00 h).
All subjects included in this research joined the Workplace Health Promotion Programme launched by our Department. This programme is carried out in accordance with the provisions of the current regulations and aimed at investigating the health status of workers exposed to urban pollutants because of their jobs.
Each worker included in the research was administered a clinical-anamnestic questionnaire, with a physician present. The questions covered age, last 5 year residence area, physiological anamnesis (especially smoking habits), past and current working history, past and current pathological anamnesis, and information about Ni exposure during time-off.
As for the exposure to tobacco smoke, we referred to the WHO classification, by classifying as “smokers” all subjects declaring they had smoked at least 100 cigarettes during their life and still smoking today or having quit smoking for less than 6 months.
For statistical purposes we took into account: age, gender, smoking habits, job task, and length of service.
All subjects agreed to make their personal information available after having been made aware that these data would have been ranked as “sensitive information”.
Each subject also consented that data should be treated in an anonymous and collective way, examined with scientific methods and analysed for scientific purposes pursuant to the Helsinki Declaration.

Environmental monitoring of Ni: personal air samplings

The exposure to environmental Ni was evaluated by means of personal air samplings. We performed 8 personal air samplings on 8 traffic policemen, selected from the 8 areas we considered as the most representative of the city air quality, and 4 personal air samplings representative for drivers on duty in the cars with the presence of at least 2 workers for each shift so that, even if only one worker was wearing the dosimeter, the results obtained appeared to be representative even for the co-worker in the same car.
Air, blood and urine samples were collected on the same day for traffic policemen and drivers, in order to avoid the influence of weather conditions on atmospheric Nickel. All the workers were asked to abstain from smoking during air sampling.
The personal air samplings were collected using Dorr-Oliver cyclones capable of separating ≤ 5 μm average diameter particles from the others. Each cyclone was attached to a pump for personal air sampling; the pump was set to a flow rate of 1.7 L per minute, according to NIOSH method 7300 (14). The method detection limit was 1 μg/L.
Each worker wore the air sampler for the whole working shift (7 hours).
For each personal air sampling, the 7-hour time-weighted average (TWA) level of exposure to Ni was measured. The American Conference of Governmental Industrial Hygienists (ACGIH) (15) has proposed a TLV-TWA 1.5 mg/m^3 limit for workers occupationally exposed to Ni.

Urinary Ni and plasma cortisol

Urinary Ni and plasma cortisol levels were measured on the 374 workers. Each worker underwent urine and blood tests (10 mL) after 4 continuative working days at the end of the shift.
Each worker was asked to refrain from eating foods such as cocoa, chocolate, soybeans, oatmeal, nuts

and seeds, fresh and dried legumes during the 4 days before the exam (16).
Each worker was taken a 10 ml peripheral venous blood sample. The samples were kept in the workplace in a refrigerator at +4°C until they were transferred (by means of a container held at the same temperature) to the laboratory and immediately centrifuged; the sera were stored at -20° C until analysis (within three days).
The laboratory performed the dosage of plasmatic cortisol by radioimmunoassay – RIA.
The urine samples were transferred to the laboratory by means of a container at a temperature of +4 °C, they were then stored in a refrigerator at a temperature of -20 °C until analysis. Urine creatinine was measured in all urine samples using Jaffè's method (17). Urine samples were analyzed by graphite-furnace atomic absorption. The limit of the method (LoD) for the detection of Ni in urine was 1,0 µg/L of urine creatinine.

Statistical Analysis

The results were analyzed and compared, according to the nature of each variable. The normal distribution of different variables was verified by the Kolmogorov-Smirnov test, and the results were significant both for urinary Ni and for cortisol. These parameters were converted into logarithmic form for the analysis of the correlation coefficient and multiple linear regression.
The results of atmospheric Ni obtained from personal air samplings and the results of urinary Ni and of plasma cortisol were expressed in terms of mean, standard deviation (SD), median and range (min-max).
Differences in mean values were analyzed with the Mann-Whitney U test for comparison of 2 variables (gender, smoking habit) and the Kruskal-Wallis test for comparison of 3 variables (age, length of service and job duty).
The Pearson's correlation coefficient (p two-tailed) was used to evaluate the correlation between urinary Ni and plasma cortisol on the total sample and after division by gender, smoking habits and job tasks.
A multiple linear regression analysis was then performed on the total sample and on subcategories (with plasma cortisol considered as a dependent variable and urinary Ni, age and length of service as independent variables).
The multiple linear regression analysis was repeated on the total sample of personal dosimetries using urinary Ni as a dependent variable and atmospheric Ni, age, length of service and smoking habits as independent variables.
The results were considered significant when p values were less than 0.05.
The statistical analysis was performed using SPSS ® 10.0 Advanced Statistical TM software.

Results

Main characteristics of the studied population

The main characteristics of the studied population are described in Table 1.
All the workers had been living and working in the same urban area for at least 5 years. No worker was exposed to Ni during time-off.
The Mann-Whitney U test revealed no statistically significant differences among the values of urinary Ni (test variable) divided by gender and tobacco smoking habits (grouping variables), nor among the values of plasma cortisol (test variable) divided by gender and tobacco smoking habits (grouping variables).
The Kruskal Wallis test revealed no statistically significant differences among the values of urinary Ni and plasma cortisol (dependent variables) divided by age, length of service and job tasks (independent variables - Age: Group A: 20-35 years; Group B: 36-45 years; Group C:> 45 years - length of service: Group A: <10 years, Group B: 10-20 years; Group C: 21-40 years) The results of these tests are reported in Table 2.
There were no statistically significant differences among the different outdoor job tasks (traffic policemen, drivers and subjects with other tasks) in relation to the average values and the distribution by age, length of service and smoking habits (smokers, non-smokers).

Environmental monitoring of Ni: personal air samplings

The values of individual exposure to atmospheric Ni are described in Table 1. All subjects reported they hadn't smoked during the sampling. No sample exceeded the limit value of 1.5 mg/m^3 ACGIH proposes for occupationally exposed subjects.
The multiple linear regression analysis results highlighted a significant correlation ($p <0.01$) between atmospheric Ni and urinary Ni both in the total sample and after subdivision on the basis of the job (traffic policemen and drivers, Tab. 3).

Urinary Ni and plasma cortisol

The values of the concentrations of urinary Ni and plasma cortisol were expressed in terms of mean, standard deviation (SD), median and range (min-max) and are described in Table 1.
All the subjects reported they hadn't eaten food containing cocoa, chocolate, soybeans, oatmeal, walnuts and almonds, fresh and dry legumes, during the 4 days prior to the collection of blood for the determination of Ni levels. Dietary and tap water and/or mineral water drinking habits were similar in all the subjects studied.
The urinary creatinine values analyzed were all within the normal range (0.3-3.0 g / l) recommended by the World Health Organization (WHO, 2007).
The Pearson's correlation analysis pointed out a statistically significant inverse correlation between the values of urinary Ni and plasma cortisol both in the total sample and in all the subgroups examined, except for the smokers group (Tab. 4).
The multiple linear regression analysis confirmed a significant inverse correlation between urinary Ni and

plasma cortisol in the total sample and in the subcategories, except for the smokers group, and the absence of a significant correlation with the confounding factors (age, length of service) (Tab. 5).

Table 1 - Features of the studied population.

Total sample	374	
Mean age (s.d.)	44.5 years (8.5)	
Mean length of service (s.d.)	14 years (8.7)	
Men	229	
Women	124	
Smokers	120	
Non smokers	233	
Traffic policemen	188	
Drivers	109	
Other duties	56	
Plasmatic cortisol (ng/ml)		
Mean (s.d.)	141.1 (55.6)	
Median	140	
Range	21-371	
Urinary nickel (μg/L)		
Mean (s.d.)	5.3 (5.1)	
Median	5.1	
Range	0.2-45.4	
Nickel: individual air samplings (μg/m^3)	Traffic policemen (N=8)	Drivers (N=4)
Mean (s.d.)	169.32 (135.1)	109.5 (117.1)
Median	92.1	68
Range	13.4-325.1	20.2-487.6

Table 2 - Results of Independent sample T test, Univariate Anova test, Mann-Whitney U test, Kruskal Wallis Test.

	Dependent variable: Cortisol		**Dependent variable: Urinary Ni**	
	Mann-Whitney U test (p)	Kruskal Wallis Test (p)	Mann-Whitney U test (p)	Kruskal Wallis Test (p)
Gender	0.12		0.22	
Smoke	0.29		0.07	
Age		0.89		0.23
Length of service		0.62		0.85
Job		0.42		0.30

Table 3 - Multiple linear regression on the 12 personal air samplings. Dependent variable: Urinary Ni.

	Total sample			**Traffic policemen**			**Drivers**		
	Beta	**t**	**p**	**Beta**	**t**	**p**	**Beta**	**t**	**p**
Air Ni	0.81	22.42	0.00*	0.93	21.62	0.00*	0.78	3.54	0.00*
Age	-0.05	-0.15	0.68	-0.02	0.35	0.74	-0.08	-1.36	0.77
Length of service	0.01	-0.22	0.77	0.03	0.47	0.53	-0.02	-0.18	0.58
Smoke	0.04	-0.38	0.50	0.03	-0.45	0.62	0.17	0.69	0.41

* Statistically significant

Table 4 - Pearson's correlation coefficient between plasma cortisol and urinary Ni in the total sample and after stratification by gender, smoking habits and job.

SAMPLE	RESULTS
Total sample (n.374)	r: -0.14 p: 0.00*
Non-smokers (n.233)	r: -0.19 p: 0.00*
Smokers (n.120)	r: -0.07 p: 0.43
Men (n.229)	r: -0.13 p: 0.00*
Women (124)	r: -0.21 p: 0.00*
Traffic policemen (188)	r: -0.22 p: 0.00*
Drivers (n.109)	r: -0.21 p: 0.00*
Other duties (n.15)	r: 0.22 p: 0.00*

* Statistically significant

Discussion

Several studies reported in the literature suggest that the toxicity of PM2.5 depends, at least in part, on the specific chemicals that are adherent to it and that the metals are often implicated as causative agents (18, 19).
Previous studies have shown that the different toxicants of urban pollution, in addition to causing adverse effects on various organs and systems (20-23), are endocrine disruptors (24, 25), that is "exogenous agents that interfere with the production, release, transport, metabolism, binding, action or elimination of the hormones naturally occurring in the body and responsible for the maintenance of the homeostasis and regulation of developmental processes", even if it is not totally clear which components are responsible for these effects.
As for the specific effect of Ni on cortisol very few studies are reported in literature. A research by Prophete et al. in 2006 on some Japanese fish (Medaka) reported a decrease in cortisol values after 7 days of exposure to Ni contaminated water (26).
The study by Perez et al. in 2008 evaluated the variation of several biological parameters due to the exposure to heavy metals in subjects assigned to cleaning tasks following the environmental disaster caused by the accident of the "Prestige" in 2002 (9). This study al-

Table 5 - Multiple linear regression in total sample and subcategories.

SAMPLE	INDIPENDENT VARIABLES	Beta	t	p
Total sample	**Age**	0.06	0.62	0.53
	Length of service	-0.04	-0.49	0.61
	Urinary Ni	-0.16	-2.55	0.00*
Smokers	**Age**	0.07	-7.07	0.49
	Length of service	-0.07	0.70	0.22
	Urinary Ni	-0.07	-0.78	0.43
Non-smokers	**Age**	-0.37	-1.25	0.36
	Length of service	0.16	-0.92	0.81
	Urinary Ni	-0.19	-2.58	0.00*
Men	**Age**	-0.07	-3.06	0.83
	Length of service	-0.07	0.23	0.33
	Urinary Ni	-0.13	-2.88	0.00*
Women	**Age**	-0.14	-0.22	0.30
	Length of service	0.52	-1.01	0.48
	Urinary Ni	-0.21	-2.23	0.00*
Traffic policemen	**Age**	-0.18	-4.57	0.66
	Length of service	0.11	-1.04	0.47
	Urinary Ni	-0.22	-2.36	0.00*
Drivers	**Age**	-0.05	0.70	0.35
	Length of service	-0.68	-0.43	0.80
	Urinary Ni	-0.20	-1.98	0.00*
Other duties	**Age**	-0.06	-5.22	0.10
	Length of service	-0.20	1.65	0.29
	Urinary Ni	-0.22	-2.02	0.00*

* Statistically significant

so investigated the correlation between exposure to Ni and cortisol levels, pointing out the presence of a significant decrease in cortisol values in relation to the increased Ni levels (9).
Our study is the first research focusing on the effects of occupational exposure to Ni in outdoor workers exposed to urban pollution and on the effects of this exposure on cortisol levels.
This study was conducted in one of the largest cities of central Italy with a population of about 2,700,000 inhabitants (27) and a density of around 1471 vehicles per Km^2 (28). In this city, atmospheric Ni is mainly found in the air as particles adhering to respirable dust (19).
The data collected through air pollution fixed monitoring stations deployed in the case-study city reveal annual averages of Ni in urban air slightly decreasing from 4.4 ng/m^3 in 2008 to 4.15 ng/m^3 in 2011 (29, 30) and similar or lower than those observed in other cities where the averages range from 2.01 to 4.5 ng/m^3 (31-33). These values indicate that urban air pollution by airborne Ni on particulate suspended matter (PTS) in the case-study city can be considered at low levels.
As for the results of individual dosimetries, although they were, on average, higher than the values recorded from the fixed stations, no samples exceeded the limit value of 1.5 mg/m^3 proposed by ACGIH for subjects occupationally exposed to Ni. The occupational exposure of outdoor workers evaluated in the present study is also lower than the exposure of industrial indoor workers (11, 34, 35).
The average values of Ni obtained from the individual dosimetries (Tab. 1) are, however, higher than the annual target value for the general population of 20ng/m^3 set by Legislative Decree 03/08/2007 n. 152, in compliance with European Directive 2004/107/EC (30).
The statistical analysis shows that the values of urinary Ni, both in traffic policemen and in drivers, depend only on the exposure to atmospheric nickel (Tab. 3).
As for the correlation between Ni and cortisol, our study confirms the very limited literature data.
Our data show that when urinary Ni levels increase, the plasmatic cortisol values significantly decrease both in the total sample and in gender and job duty stratifications hence demonstrating the existence of an inverse correlation between Ni and cortisol regardless of gender and specific outdoor job.
The Mann-Whitney U test, the Kruskal Wallis test and the multiple linear regression show that the main confounding factors under exam (gender, age, length of service, smoking habits) have no influence on plasma cortisol, and that the decrease of cortisol values depends only on the increase of Ni levels. It should be noted that the fact that Ni is not a metal with cumulative properties may help explain why, in our research, it did not vary depending on the age and length of service.
As for the role of cigarette smoke, the stratification by smoking habit showed that the inverse correlation between urinary Ni and cortisol exists only with respect to non-smokers. Anyway, the Mann-Whitney U test did not show, as explained above, that cigarette smoke may affect cortisol values and urinary Ni levels. The data published by several authors (36-38) could explain this result: the health consequences of Ni found in cigarettes are controversial and the main source of exposure to Ni for outdoor smoking workers occupationally exposed to urban pollution is presumably residing in atmospheric Ni and not in Ni found in cigarette smoke, which could explain why a statistic test such as the Mann-Whitney U test gives non-significant results.
As for the correlation and the multiple linear regression, no significant inverse correlation between urinary Ni and cortisol is seen for the group of smokers; in respect of this result it can be assumed that the other toxicants present in tobacco behave as confounding factors, with various and multidirectional effects on cortisol values, thus masking the specific effect of nickel (39).
Moreover, it should be noted that smokers were asked to abstain from smoking during samplings and it has been demonstrated that acute nicotine abstinence may induce an increase of cortisol levels (40); which could possibly justify the non-significant decrease of cortisol levels in smokers.
Finally, we have to underline that a previous study carried out by our research group on a similar working population – comparing traffic policemen and workers with other outdoor duties (41) – showed significantly higher plasma cortisol values in traffic policemen. This difference with the results of the present study could be related to the fact that traffic policemen are exposed to a wide range of stressors, including psychosocial ones (42), which can determine an increase of cortisol levels, and to several environmental chemicals which are likely to have opposite effects on cortisol.

Conclusions

For the first time in literature, this study investigates the possible correlation between the exposure to low Ni doses in urban air and plasma cortisol values in outdoor workers by means of personal air samplings and biological monitoring of urinary Ni.
Our results indicate the presence of a significant association between the exposure to low doses of atmospheric nickel and a consequent decrease of cortisol values. These results should encourage further research on the effects of Ni on exposure to urban pollutants in the working population. Several studies report the association between low cortisol values and the risk of developing depression (43, 44).
In conclusion, preventive measures should therefore be used to protect the health of the exposed workers.
Cortisol could also be used as an early biological marker, applicable to the group, in relation to workers occupationally exposed to low doses of Ni before the appearance of out-of-range values.

References

1. EPA, Environmental Protection Agency. (2008). List of the 33 urban air toxics. Available from: http://www.epa.gov/ttnatw01/urban/list33.html
2. ATSDR, Agency for Toxic Substances and Disease Reg-

istry - U.S. Department of health and human services - Public Health Service. (2005). Toxicological profile for Nickel. Available from: http://www.atsdr.cdc.gov/toxprofiles/tp15.pdf
3. IARC, International Agency for Research on Cancer. (1990). IARC Monographs on the evaluation of carcinogenic risks to humans. Chromium, nickel and welding. Volume 49. Available from: http://monographs.iarc.fr/ENG/Monographs/vol49/mono49.pdf (last access Feb 2014)
4. Chen YW, Yang CY, Huang CF, et al. Heavy metals, islet function and diabetes development. Islets. 2009; 1(3):169-176.
5. Apostoli P, Catalani S. Metal ions affecting reproduction and development. Met Ions Life Sci. 2011; 8:263-303.
6. Kročková JZ, Massányi P, Sirotkin AV, et al. Nickel induced structural and functional alterations in mouse Leydig cells in vitro. J Trace Elem Med Biol. 2011; 25(1):14-18.
7. Fu Y, Tian W, Pratt EB, et al. Down-regulation of ZnT8 expression in INS-1 rat pancreatic beta cells reduces insulin content and glucose-inducible insulin secretion. PLoS One. 2009; 4(5):e5679.
8. Tomei G, De Sio S, Fiaschetti M, et al. (2011). Donna in gravidanza e lavoro – salute riproduttiva e benessere femminile. Prevent Res, published on line 31. Oct. 2011, P&R Public. 05. Available from: http://www.preventionandresearch.com/ doi: 10.7362/2240-2594.050.2011
9. Pérez-Cadahía B, Laffon B, Porta M, et al. Relationship between blood concentrations of heavy metals and cytogenetic and endocrine parameters among subjects involved in cleaning coastal areas affected by the 'Prestige' tanker oil spill. Chemosphere. 2008; 71(3):447-455.
10. Gil F, Hernandez AF, Marquez C, et al. Biomonitorization of cadmium, chromium, manganese, nickel and lead in whole blood, urine, axillary hair and saliva in an occupationally exposed population. Sci Total Enviro. 2011; 409:1172-1180.
11. Pesola G, Elia G, Lovreglio P, et al. Valutazione dell'esposizione professionale a nichel nella produzione di dibutilditiocarbammati. G Ital Med Lav Erg. 2006; 28(3):315.
12. Weisse T, Pesch B, Lotz A, et al; the WELDOX Group. Levels and predictors of airborne and internal exposure to chromium and nickel among welders-Results of the WELDOX study. Int J Hyg Environ Health 2013; 216(2):175-183.
13. Campurra, G. Manuale medicina del lavoro, Ipsoa Indicitalia Eds, Italy, 2010.
14. NIOSH manual of analytical method. Available from: http://www.cdc.gov/niosh/docs
15. ACGIH - American Conference of Governmental Industrial Hygienists. (2012). Documentation of the threshold limit values and biological exposure indices. 7th ed. Cincinnati, OH.
16. Arnich N, Sirot V, Riviere G, et al. Dietary exposure to trace elements and health risk assessment in the second French Total Diet Study. Food Chem Toxicol. 2012; 50:2432-2449.
17. Liu WS, Chung YT, Yang CY, et al. Serum creatinine determined by Jaffe, enzymatic method, and isotope dilution-liquid chromatography-mass spectrometry in patients under hemodialysis. J Clin Lab Anal. 2012; 26(3):206-214.
18. Xu X, Rao X, Wang TY, et al. Effect of co-exposure to nickel and particulate matter on insulin resistance and mitochondrial dysfunction in a mouse model. Part Fibre Toxicol. 2012; 9:40.
19. WHO, World Health Organization Regional Office for Europe Copenhagen. (2000). Air quality guidelines for Europe, 2nd Edition, WHO Regional Publications, European Series, No. 91. (NLM Classification: WA 754)
20. Sancini A, Tomei F, Tomei G, et al. Urban pollution. G Ital Med Lav Ergon. 2012; 34(2):187-196.
21. Sancini A, Tomei F, Gioffre PA, et al. Occupational exposure to traffic pollutants and peripheral blood counts. Annali di igiene: medicina preventiva e di comunità 2012; 24(4):325-344.
22. Sancini A, Caciari T, Andreozzi G, et al. Respiratory parameters in traffic policemen exposed to urban pollution. European Journal of Inflammation 2010; 8(3):157-163.
23. Lettieri Barbato D, Tomei G, Tomei F, et al. Traffic air pollution and oxidatively generated DNA damage: can urinary 8-oxo-7,8-dihydro-2-deoxiguanosine be considered a good biomarker? A meta-analysis. Biomarkers. 2010; 15 (6):538-545.
24. Ciarrocca M, Tomei F, Caciari T, et al. Exposure to arsenic in urban and rural areas and effects on thyroid hormones. Inhal Toxicol. 2012; 24(9):589-598.
25. Ciarrocca M, Capozzella A, Tomei F, et al. Exposure to cadmium in male urban and rural workers and effects on FSH, LH and testosterone. Chemosphere. 2013; 90(7): 2077-2084.
26. Prophete C, Carlson EA, Li Y, et al. Effects of elevated temperature and nickel pollution on the immune status of Japanese medaka. Fish Shellfish Immunol. 2006; 21:325-334.
27. ISTAT, Italian National Institute of Statistic. (2008). Bilancio demografico anno 2008 e popolazione residente al 31 Dicembre 2006. Comune Roma. Available from: http://demo.istat.it/bil2008/index.html
28. ISPRA, Institute for Protection and Environmental Research. (2006). Inventario nazionale delle emissioni in atmosfera. Available from: http://www.isprambiente.gov.it/site/it-IT/
29. ARPA Lazio, Regional Agency for the Environmental Protection. (2008). Rapporto sullo stato della qualità dell'aria 2008. Available from: http://www.arpalazio.net/main/aria/doc/pubblicazioni.php
30. ARPA Lazio, Regional Agency for the Environmental Protection. (2012). Rapporto sullo stato della qualità dell'aria nella Regione Lazio 2011. Available from: http://www.arpalazio.net/main/aria/doc/pubblicazioni.php
31. Singh M, Jaques PA, Sioutas C. Size distribution and diurnal characteristics of particle-bound metals in source and receptor sites of the Los Angeles Basin. Atmos Environ. 2002; 36:1675-1689.
32. Wang G, Huang L, Gao S, Wang L. Characterization of water-soluble species of PM10 and PM2.5 aerosols in urban area in Najing, China. Atmos Environ. 2002; 36:1299-1307.
33. Moreno T, Querol X, Alastuey A, et al. Variations in vanadium, nickel and lanthanoid element concentrations in urban air. Sci Total Environ. 2010; 408:4569-4579.
34. Stridsklev IC, Schaller KH, Langård S. Monitoring of chromium and nickel in biological fluids of grinders grinding stainless steel. Int Arch Occup Environ Health. 2007; 80:450-454.
35. Kozo Y, Yasushi J, Yukihiro K, et al. Urinary elimination of nickel and cobalt in relation to airborne nickel and cobalt exposures in a battery plant. Int Arch Occup Environ Health. 2007; 80:527-531.
36. Stojanović D, Nikić D, Lazarević K. The level of nickel in smoker's blood and urine. Cent Eur J Public Health. 2004; 12(4):187-189.
37. Torjussen W, Zachariasen H, Andersen I. Cigarette smoking and nickel exposure. J Environ Monit. 2003; 5(2):198-201.
38. Rosati MV, Sancini A, Tomei F, et al. Plasma cortisol concentrations and lifestyle in a population of outdoor workers. Int J Environ Health Res. 2011; 21(1):62-71.
39. Ciarrocca M, Rosati MV, Tomei F, et al. Is urinary 1-hydrox-

ypyrene a valid biomarker for exposure to air pollution in outdoor workers? A meta-analysis. J Expo Sci Environ Epidemiol. 2014; 24(1):17-26.
40. Wardle MC, Munafò MR, de Wit H. Effect of social stress during acute nicotine abstinence. Psychopharmacology. (Berl) 2011; 218(1):39-48.
41. Tomei F, Rosati MV, Ciarrocca M, et al. Plasma cortisol levels and workers exposed to urban pollutants. Ind Health. 2003; 41(4):320-326.
42. Tomei G, Sancini A, Capozzella A, et al. Perceived stress and stress-related parameters. Ann Ig. 2012; 24(6):517-526.
43. Keenan K, Hipwell A, Babinski D, et al. Examining the developmental interface of cortisol and depression symptoms in young adolescent girls. Psychoneuroendocrinology. 2013; 38(10):2291-2299.
44. Grynderup MB, Kolstad HA, Mikkelsen S, et al. A two-year follow-up study of salivary cortisol concentration and the risk of depression. Psychoneuroendocrinology. 2013; 38(10):2042-2050.