Exposure to Urban Pollutants and Serum Levels of IgG anti Herpes Simplex Virus Type 1

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Abstract: The aim of the study is to investigate whether traffic policemen exposed to urban pollutants and psycho-social stressors may be at risk of modifications in serum levels of immunoglobulins G antibodies (IgG Ab) against Herpes Simplex Virus (type1) (HSV-1) compared with controls. Traffic policemen were matched by sex, age, working life and drinking habits (less than two glasses of wine or beer per day) with controls, after excluding the subjects with the principal confounding factors (smoking habit, use of paints, solvents, pesticides and drinking habits). Were included in the study 125 traffic policemen (54 men and 71 women) with outdoor activity and 125 controls (54 men and 71 women) with indoor activity. Mean levels of IgG Ab against HSV-1 were significantly higher in traffic policemen of both sexes compared with controls. The frequency of workers with IgG Ab against HSV-1 levels higher than our normal laboratory values, was significant in traffic policemen compared with controls of both sexes. The distributions of IgG Ab against HSV-1 levels in traffic policemen compared with controls of both sexes were significant. The authors hypothesise an effect of the working activity in traffic policemen on serum levels of IgG Ab against HSV-1 compared to controls.

Key words: Herpes Simplex Virus (Type 1), Urban pollutants, Traffic policemen, Outdoor activity, Psycho-social stressors

Introduction

One of the characteristics of Herpes Simplex Virus (HSV) is the ability to establish a latent infection, to persist in an apparently inactive state for a variable period of time and to be reactivated thereafter. After primary infection a cellular-mediated immunity develops; it could be an extremely important factor in the maintenance of a latent state. The latent virus can be reactivated following various stimuli (such as immunodepression), and it can enter a replicating cycle at any time of its life. The virus reactivation can carry to an increase of serum levels of immunoglobulins G antibodies (IgG Ab) against Herpes Simplex Virus (HSV)11.

Studies on animals and human subjects have suggested that some urban pollutants9, may be able to alter the immune response. These urban pollutants are: manganese5, chromium4, lead5, cadmium6-8, nickel9, ozone10, 11, polycyclic aromatic hydrocarbons12-14, nitrogen dioxide15, benzene16, 17, toluene17, 18, styrene19, 20, biphenyl polychlorides21 and noise3, 22. Beside these, psycho-social stressors may be able to alter serum levels of immunoglobulins G antibodies (IgG Ab) against Herpes Simplex Virus (type1) (HSV-1)23-25.

The class of workers examined in this study were employees of the Municipal Police of a big Italian city, for whom we have already studied the environmental and biological levels of some urban pollutants26-29. The levels of individual exposure to benzene, considered as an indicator
of urban pollutants, were (mean 7 h) on average \(10.7 \mu g/m^3\) for the group of traffic policemen and about three times lower (3.6 \(\mu g/m^3\)) for the control group\(^{26,28}\).

Though unleaded gasoline had been introduced, a mixed regime existed in Italy at the time of this study (use of leaded and unleaded gasoline). Moreover, it is probable that anti-detonation additives containing manganese and nickel are present in fuels. Finally the use of green (unleaded) gasoline is the cause for the increase of benzene in urban air. Besides, our current research has shown an increase in serum nickel in traffic policemen exposed to urban pollutants compared to a control group\(^{30}\).

As far as dust particles were concerned, the Municipality of the city in question monitored concentrations of PM10 in fixed stations located in districts with different intensities of vehicle traffic. The average value during the period 2000-2001 was 53.75 \(\mu g/m^3\) [http://www.comune.roma.it].

The aim of the present study is to investigate whether traffic policemen of a big city exposed to urban pollutants that may interact with psycho-social stressors may be at risk of modifications in serum levels of immunoglobulins G (IgG Ab) against HSV-1 compared with a control group.

### Materials and Methods

The research was carried out on a working population of 917 Municipal Police employees.

We studied two groups: traffic policemen and control subjects. The traffic policemen exposed to urban pollutants worked on parking control, control of passages, and control of roads with heavy traffic. The subjects doing indoor activities of an administrative and bureaucratic nature, at lesser level of exposure, were used as control group. Traffic policemen and the control group worked for seven hours a day at least five days a week.

For inclusion in the study, all workers were given a questionnaire with the following items: age, working life, pharmacological therapies, use of paints, pesticides and solvents during time off, smoking habit (number of cigarettes smoked per day, years of subjection to the habit), drinking habits (wine, beer, spirits; number of glasses of wine, beer or spirits drunk per day), clinical symptoms.

In order to avoid the influence of confounding factors, subjects who referred exposure to solvents, paints and pesticides\(^{31–36}\), subjects that referred smoking habit\(^{37–44}\) and subjects who referred to drinking habits (more than two glasses of wine or beer per day)\(^{45–47}\) were excluded from the study. Subjects who referred to habitual consumption of spirits were not present in this study.

All the subjects included or excluded from the study were asymptomatic.

### Table 1. Characteristics of the male and female population, levels of IgG Ab against Herpes Simplex (type 1) and number of subjects with IgG Ab against Herpes Simplex levels outside our normal laboratory limit in traffic policemen and control group

<table>
<thead>
<tr>
<th></th>
<th>Male N=108</th>
<th>Female N=142</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Traffic policemen</td>
<td>Controls</td>
</tr>
<tr>
<td></td>
<td>N=54</td>
<td>N=54</td>
</tr>
<tr>
<td>Age (yr) mean ± SD</td>
<td>45 ± 7.6</td>
<td>44.9 ± 7.2</td>
</tr>
<tr>
<td>min-max</td>
<td>30–61</td>
<td>32–59</td>
</tr>
<tr>
<td>Working life (yr)</td>
<td>9.02 ± 5.6</td>
<td>8.72 ± 5.3</td>
</tr>
<tr>
<td>mean ± SD</td>
<td>1–25</td>
<td>1–25</td>
</tr>
<tr>
<td>min-max</td>
<td>1.5–188</td>
<td>1–168</td>
</tr>
<tr>
<td>IgG Ab against HVS-1</td>
<td>118 ± 41.1*</td>
<td>93.9 ± 54.2</td>
</tr>
<tr>
<td>mean ± SD</td>
<td>1.5–188</td>
<td>1–168</td>
</tr>
<tr>
<td>min-max</td>
<td>52 (96.3%)*</td>
<td>42 (77.78%)</td>
</tr>
<tr>
<td>IgG Ab against HVS-1</td>
<td>&gt;20 UA/ml</td>
<td>&gt;20 UA/ml</td>
</tr>
</tbody>
</table>

*P< 0.05, **P<0.01, ***P<0.001.  SD = standard deviation.  yr = year.
A 10 ml sample of venous blood was taken from each worker between 8 and 10 a.m., fasting. The blood samples were preserved at the workplace in a refrigerator at –4°C until the time when they were transferred (by means of a container and at the same temperature) to the laboratory, where they were immediately centrifuged to obtain the serum that was preserved at –20°C until the time when they were analysed (within 3 d).

IgGs against HSV-1 were determined by EIA (enzyme immunoassay) on the serum: normal laboratory values are < 20 UA/ml both in male and female subjects. The clinical specificity of the method was evaluated over a representative group of individuals with no immunity against HSV type 1 infection. The result was 100%. The clinical sensitivity of the method was evaluated over a representative group of individuals having undergone past HSV type 1 infection. The result was 97.6%. The intra-assay precision of the assay were in the range 3.6–5.8% and the inter-assay precision were in the range 8.84–14.5%48).

All samples were analysed in blind condition, although both the physicians and the technicians knew how the study was being carried out.

**Statistical Analysis**

Statistical analysis of the data was based on the calculation of the mean, standard deviation (SD), distribution, range and frequency according to the nature of the single variables. The differences between group means were analyzed using Student’s t-test for unpaired data. The frequencies of the single variables were compared using the chi-square test with Yates’ correction.

The differences were considered significant when the P values were < 0.05.

The statistical analysis was done using the statistical program Solo-BMDP™ Statistical Software.

**Results**

In male subjects mean levels of IgG Ab against HSV-1 were significantly higher in traffic policemen compared with the control subjects (P<0.05) (Table 1). Similarly in female subjects mean levels of IgG Ab against HSV-1 were significantly higher in traffic policemen compared with the control subjects (P<0.001) (Table 1).

The frequency of workers with IgG Ab against HSV-1 levels higher than our normal laboratory values, was significant in traffic policemen compared with the control subjects both in male (P<0.001), and in female subjects (P<0.01) (Table 1).

The distributions of IgG Ab against HSV-1 levels in traffic policemen of both sexes were significantly different compared with those of controls and are shown in Fig. 1 and in Fig. 2.
Discussion

Considering that the subjects with the principal confounding factors were excluded from the study and that the subjects investigated were matched by sex, age, working life and drinking habits the data suggest that exposure to urban pollutants could alter serum levels of IgG Ab against HSV-1.

The results obtained in this study suggest the following considerations: 1. the fact that there is a significantly higher percentage of traffic policemen positive to IgG Ab against HSV-1 than in controls could explain that infection rate of HSV-1 is increased among traffic policemen; 2. the fact that the average titer of IgG Ab against HSV-1 is significantly higher in traffic policemen than in controls could explain that latent virus is reactivated among traffic policemen than in controls.

It could be assumed that a decreased cellular mediated immunity in traffic policemen could induce a reactivation of HSV-1 latent infection and at the same time an increase in average titer of IgG Ab against HSV-1. In fact in another study we found that traffic policemen of both sexes have a decreased proliferative response of blood lympho-monocytes to the mitogen PHA compared to a control group\(^49\). So, it is very important to verify HSV-1 reactivation through the study of humoral immunity and therefore with dosage of IgG Ab against HSV-1. Moreover, these significant results suggest that these differences may have clinical validity and it indicates an immunodepressive condition in this category of workers. Several studies in literature showing that occupational exposure to urban pollutants may lead to immune dysfunction and immunosuppressive effect with an increased risk for developing infectious and neoplastic diseases\(^14, 22, 50-53\).

The action mechanisms of urban pollutants able to modify serum levels of IgG Ab against HSV-1 are not yet completely clear. Studies on animals\(^7, 15, 18, 22\) and human subjects\(^2-4, 9, 11-14, 16, 17, 19-21\) have suggested that exposure to chemical and physical substances could increase (polycyclic aromatic hydrocarbons, nickel, lead and noise) or decrease (benzene, toluene, styrene and ozone) serum levels of IgG Ab against HSV-1, probably related to dose, modality and time of exposure\(^7, 14\). A decrease in IgG levels are described also by the association to chronic exposure to chemical and physical pollutants such as manganese and noise\(^3\).

It is well known that personal exposure to benzene, toluene and other aromatic hydrocarbons from direct exposure to traffic fumes, as experienced by some categories of outdoor workers, such as traffic policemen, may be considered higher than personal exposure of indoor workers (particularly in our cities)\(^49\). For this reason in our previous researches we have studied exposure dosage to benzene, toluene and other aromatic hydrocarbons in Municipal Police employees of the city in question. Time weighted average (TWA) exposure to benzene (mean 10.7 and 3.6 µg/m\(^3\), respectively) and to toluene (mean 40.7 and 13.5 µg/m\(^3\), respectively) was significantly higher among traffic policemen than among indoor workers\(^27, 28\). Since previous studies have already measured the environmental and biological levels in our working population and it is well known that traffic policemen’s exposure dosage is significantly higher than controls, we didn’t repeat the exposure dosage study in this work.

It is well know that traffic policemen are a working population exposed to stress\(^55, 56\). Sources of stress for the traffic policemen may be the relation with the public, exposure to episodes of criminality, and the need to maintain high levels of services in various contexts. In a previous study we have analysed subjective stress in Municipal Police employees. To this end, we administered the Rapid Stress Assessment scale (RSA); total RSA score were found to be significantly higher in traffic policemen compared to controls\(^57\). Our results may be explained, also, with major subjective stress in traffic policemen compared to controls, according with our previous researches\(^57\). Moreover some authors have suggested the possibility of a positive correlation between exposure to psycho-social stressors and the titer antibodies against HSV-1\(^23-25\).

These data suggest that a cumulative effect of stressors from different nature may induce increased or decreased serum levels of IgG Ab against HSV-1.

In particular the results obtained suggested that the working activity of traffic policemen exposed to urban pollutants that may interact with psycho-social stressors can increase the levels of IgG Ab against HSV-1.

Acknowledgments

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