Investigating Lead Level in Blood and Associated Risk Factors

Musa Kaleem Baloch, Mukhtiar Hassan and Mumtaz Khan

The levels of lead in blood of 200 male volunteers of 25-40 years of age residing in different areas of Dera Ismail Khan were determined by applying atomic absorption technique. Out of these examined persons, 100 were living in urban areas under dense traffic conditions while others were from rural area. Concentration of lead in blood of urban test group ranged between 0.04 to 1.6ppm while in rural the concentration was between 0.01 to 0.73 ppm. The hemoglobin, cholesterol and glucose level in blood was also estimated both in urban and rural inhabitants. The mean estimated values of blood glucose, cholesterol and hemoglobin contents of rural group were 100.7 mg dl⁻¹, 211.6 mg dl⁻¹ and 12.5 mg % respectively, while on the other hand the mean blood glucose, cholesterol and hemoglobin contents of urban exposed persons were 127.3 mg dl⁻¹, 240 mg dl⁻¹ and 10.7mg % respectively. Analysis of the results showed that with the increase in lead concentration in blood, the hemoglobin level decreased while cholesterol and glucose level was increased.

Key words: Blood lead, hemoglobin, cholesterol, glucose, lead pollution
Introduction
The role of heavy metals (e.g. Pb, Hg, Cd and Cr), as toxicity has been widely recognized. The discharge of huge quantity of toxic metals into the air results in the transfer of pollutant metals such as lead (Pb⁺⁺) to the blood. Heavy metals, as harmful environmental pollutants and one of the most important toxicant producing adverse effects in human beings. Because of the heavy load of contaminated dust in the air of the overcrowded cities, the ambient concentration of the toxic metals is increasing to the alarming levels. Lead poisoning is usually caused by inhalation of minute particles of Pb fume and dust, which are absorbed by the blood stream from the lungs and which easily deposited in the bone marrow. Lead does not have any therapeutic use in the human body (Goodman et al., 1970 and Sharma, 2000). In blood, lead concentrations exceeding 0.40μg ml⁻¹ may indicate abnormal exposure and concentrations of 0.60μg ml⁻¹ or greater may indicate toxicity (Bauer, 1982).
Lead produces a toxic effect on kidneys resulting in impaired tubular absorption of glucose, phosphates, bicarbonates, amino acids and uric acids. Biochemically, it interferes with the creation of hemoglobin by inhibiting the enzymes involved in the process, thus leading to the anemia (Goyer et al., 1993; Grover et al., 1995). Low but long-term lead exposure produces hypertension and cardiac in tropism without any change in heart rate or histopathological damage (Carmignanis et al., 1989). High Pb level inhibits spermatogenesis, kidney dysfunction and permanent brain damage (Sharp et al., 1988). Children have higher susceptibility to the Pb because of their fast growth rate and its concentration in blood greater than 0.40μg ml⁻¹ may indicate toxicity for children (Bauer, 1982).
Therefore, this investigation was undertaken to determine the Pb concentration in blood samples of urban individuals exposed to vehicular exhaust and to compare with the blood levels of rural inhabitants. The effect of Pb concentration upon hemoglobin, cholesterol and glucose level was also recorded.

Materials and Methods
Estimation of lead in blood
Reagents:
1. Triton X-100, 5% v/v solution prepared with deionized water.
2. Ammonium pyrolidine thiocarbamate (APDC), 2% w/v solution prepared in 5% Triton X-100 and stored in refrigerator.
3. Methylisobutylketone (MIBK), water saturated.
4. Lead stock standard, 1000μg ml⁻¹.
   a. Different concentration of lead 25, 50, 75 and 100μg dl⁻¹ were prepared in duplicate by diluting stock standard.
   b. Two sets of duplicate blanks were also prepared with deionized water. To each of these tubes added 5ml pooled whole blood.

Specimen Collection: A total of two hundred blood samples were collected. 100 were from the residents of urban area under dense traffic condition and the 100 from rural area including 50 non-polluted controls. The control volunteers had no apparent physiological or biochemical symptoms of any disease. 5ml blood samples were collected from cephalic vein in lead-free, hyperinized collection tube. 2.5ml of each sample was centrifuged for 10 min. at 2000 rpm to get serum for glucose and cholesterol estimation.

Sample Preparation: To a tube containing 1ml deionized water, 5ml of the whole blood specimen was added. 1ml of APDC (2%) solution added to unknown samples, blanks and standard tubes. APDC was used to chelate lead in hemolyzed blood. The lead APDC was extracted into methylisobutylketone (MIBK) by adding 3 ml of it and shaking vigorously for about 60 min. to ensure complete extraction. It was centrifuged for 10 min. at 2000 rpm. Each standard (blank and unknown) was aspirated in the flame and absorption of the Pb is determined in an atomic absorption spectrophotometer. The average blank value was subtracted from the absorbance of each standard to correct for any Pb present in the pooled blood. A standard calibration curve was prepared by plotting the corrected absorbance against the concentration of the Pb standards. The concentration of the unknown was determined from the observed absorbance compared to the corresponding Pb concentration on the calibration curve (Bauer, 1982).

Instrumentation: Sample injection was made automatically using an auto sampler with a precision of ± 0.1μl. The sample was aspirated in the flame and absorption of the Pb was determined in the atomic absorption spectrophotometer of Hitachi model Z-8000 with air acetylene flame (analytical wavelength 283.3nm) (Bauer, 1982).

Estimation of Blood Glucose Level: The blood glucose level was determined at 540nm by glucose oxidase method using Spectronic-21. Commercially available glucose kit was used as a standard (Trinder, 1969).

Estimation of Blood Hemoglobin Level: The blood hemoglobin level was determined by Sahli's Hemoglobinometer method. In normal male adults hemoglobin level is ranged from 14 to 16mg 100ml⁻¹ of blood (Bauer, 1982).

Estimation of Blood Cholesterol Level: Total cholesterol was determined by the use of cholesterol oxidase method. The standard and samples were read against blank at 500nm in a spectrophotometer (Richmond, 1973).

Estimation of Sodium, Potassium and Lithium: Sodium, Potassium and Lithium in blood was determined by flame photometry. The normal level of sodium and potassium in serum is from 136-145 and 3.8-5.8 mmoles L⁻¹ respectively (Bauer, 1982). The normal level of lithium in serum is 1-1.5 mmoles L⁻¹ (Wallach, 1998).

Results and Discussion
The amount of Pb in blood samples of urban test group ranged from 0.04 to 1.6ppm while in rural group the concentration of Pb was in the range of 0.01 to 0.73ppm. The urban group exposed to vehicular exhaust had distinctly high level of Pb in their blood as compared to blood Pb levels of rural group (Table 1). The normal tolerable limit of blood lead is 40μg dl⁻¹ or 0.4 ppm (WHO, 1977). It was observed that duration of exposure to Pb polluted atmosphere had significant effect on blood lead concentration (Fig.1). The mean estimated values of blood glucose, cholesterol and hemoglobin contents of rural group were 100.7μg dl⁻¹, 211.6μg dl⁻¹ and 12.5μg % respectively, while on the other hand the mean blood glucose, cholesterol and hemoglobin contents of urban exposed persons were 127.3μg dl⁻¹, 240μg dl⁻¹ and 10.7μg % respectively (Table 1). The concentration of glucose, cholesterol and hemoglobin has good correlation with the Pb levels in blood (Fig. 2). The results showed that with the increase in concentration of lead in blood, the hemoglobin level decreased while glucose level increased. Cholesterol levels were found
Table 1: Mean estimated values of glucose, cholesterol and hemoglobin in blood of rural and urban groups

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Rural</th>
<th>Urban</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lead (ppm)</td>
<td>0.4</td>
<td>0.77</td>
</tr>
<tr>
<td>Glucose (mg dl(^{-1}))</td>
<td>100.7</td>
<td>127.30</td>
</tr>
<tr>
<td>Cholesterol (mg dl(^{-1}))</td>
<td>211.6</td>
<td>240.00</td>
</tr>
<tr>
<td>Hemoglobin (mg %)</td>
<td>12.5</td>
<td>10.70</td>
</tr>
</tbody>
</table>

Table 2: Average blood lead concentration in residents of urban and rural areas

<table>
<thead>
<tr>
<th>Exposure Period</th>
<th>Blood lead concentration (ppm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>(Year)</td>
<td>Urban area</td>
</tr>
<tr>
<td>&lt;10</td>
<td>0.04</td>
</tr>
<tr>
<td>11-20</td>
<td>0.57</td>
</tr>
<tr>
<td>21-30</td>
<td>0.87</td>
</tr>
<tr>
<td>&gt;30</td>
<td>1.60</td>
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</tbody>
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Fig. 1: A relationship of an average level of lead in blood with exposure period

Fig. 2: Influence of blood lead concentration upon other parameters of blood

in both groups. The glucose concentration in blood was also increased to some extent with the increase in level of Pb in blood (Fig. 2). The blood potassium, sodium and lithium (mean values of 140, 3.9 and 1.5mmoles L\(^{-1}\) respectively) in both groups were found to be normal range and seems to be unaffected by the blood Pb level (Fig. 3). The locality has significant effect on the concentration of lead in blood as the lead levels of urban people were found to be significantly higher than those of rural ones (Table 2). The results are in accordance with the findings of other researchers (Rehman et al., 1988 and 1997). They observed that the lead level in the atmosphere were comparatively higher in urban area under peak traffic condition than in rural areas. Lead levels of people living in urban areas varied from 0.4-1.6ppm (Rehman et al., 1988) and 0.72-1.9ppm (Rehman et al., 1997). But our results are against the findings of Vitti et al. (1986) who showed that the activities carried out in contact with road traffic did not give rise hematic lead levels higher than those found in the non-exposed population. They determined that the lead contamination was homogenous in the whole urban territory. It is obvious from the data of the Table 1 that the blood lead levels of rural group (0.01-0.4 ppm) were in the safe limit value of 0.4 ppm or 40μg dl\(^{-1}\) (WHO, 1977 and Sharma, 2000). On the other hand the levels of lead (0.04-1.6ppm) were found to be significantly higher in the blood of urban population. This is in support with the results obtained by Grover et al., 1995 (61μmg dl\(^{-1}\)) and Ikhtiar et al., 2000 (52.4μg dl\(^{-1}\)). Analysis of the results showed that highest Pb contents were found in samples collected from residents of heavy polluted area where the traffic density was expected to be the high as compared to the blood samples collected from residents of rural area. This suggested that vehicular exhaust constitute the major lead emission source. Once the area gets polluted with heavy metals, it is extremely difficult to detoxify it.

References


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