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Urinary Excretion of Total and Inorganic Lead in Tetraethyllead Exposed Workers

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Alkyl lead species, particularly tetraethyllead (TEL) have been used as antiknock agents in gasoline for more than 50 years through out the world (Caplun et al 1984). Then TEL was realized as a very toxic compound causing severe psychotic disorders and also as a main source of lead contamination in urban metropolitan areas. Thus, lead content in gasoline in USA declined sharply since 1975 and the western world followed this reduction a few years later (Shy 1990). Although only lead free gasoline may/now be used as an automobile fuel in the USA, leaded gasoline is still used in many countries including Turkey. In Turkey lead content have been reduced to 0.15 g/l in normal gasoline and to 0.4 g/l in super gasoline after 1988. It is also planned to produce unleaded super gasoline beginning from 1998 and only one type of unleaded gasoline after 2003 (TÜPRAS 1994).

Occupational exposure to TEL may occur during the production, transport and mixing of TEL with gasoline and by the cleaning procedure of leaded gasoline storage thanks. Motor mechanics who use gasoline to clean their hands may be particularly at risk especially in our country. These workers exposed to TEL extensively by skin penetration due to the excellent liposolubility of TEL (Grandjean 1979).
In Turkey there is no available data on the occupational TEL exposure, therefore we aimed to investigate the extent of TEL exposure in refinery workers, motor mechanics and gasoline station workers.

MATERIALS AND METHODS

Occupationally TEL exposed subjects were selected from three different occupations: a) Workers employed by petrol refinery plants to fill gasoline storage tanks (refinery workers; n:49), b) Motor repairmen who used gasoline to clean their hands (motor mechanics; n:50) and c) Gasoline station (servicing) workers (n:42). In addition, nonoccupationally exposed people living in the central part of Ankara were studied as a control group (n:35). Urine samples were collected in polyethylene containers without using preservative and samples were

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kept in a deep freeze (-25°C) until analyzed. Total lead and inorganic lead in urine were determined using Varian 30/40 Model, Flame Atomic Absorption spectrophotometer (FAAS) in combination with Slotted Tube Atom Trap (STAT). Slotted quartz tube was supplied from Philips, Eindhoven.

Lead stock solution (100 µg/ml) was prepared by dissolving 0.159 g lead nitrate in 100 ml 1% HNO_3. Stock lead solution was used to prepare working standards in the range of 1.0 - 5.0 µg/ml by appropriate dilution in 1% HNO_3. Calibration, recovery and sensitivity of lead in urine were determined as described by Karakaya and Taylor (1989) and Arai (1996).

The urine sample (40 ml) was transferred to a uniseal decomposition vessel and decomposed with heat and pressure in 5 ml of concentrated nitric acid. The pH was adjusted to 7 and lead was extracted with 2 ml of 3% sodium diethyldithiocarbamate (NDDC) and 5 ml of methyl isobutyl ketone (MIBK). The MIBK layer was used for the determination of total lead (PbT) by FAAS in combination with slotted quartz tube at a wavelength of 283.3 nm (Arai 1986).

Inorganic lead (Pbl) in urine was determined as described by Arai (1986). Accordingly lead was precipitated as lead oxalate in urine. After transferring the precipitate to a uniseal decomposition vessel, the same procedure as described above in PbT determination was accomplished.

Lead levels in urine were also given as µg Pb/g creatinine for the concentration adjustment in spot urine samples (Trevisan 1990). Creatinine in urine was determined as described by Baselt (1980) and was measured directly in urine by reaction with picric acid. The chromogen was measured at 530 nm in a visible spectrophotometer.

The values of control and exposed workers were compared by Students t-test and a p value of 0.05 denoted significance.

RESULTS AND DISCUSSION

The detection limit of STAT-FAAS method was determined 5.2×10^{-3} µg/ml for lead. The recovery rates and coefficients of variation (in 10 repetition) are shown in Table 1. We compared also the sensitivities of the conventional FAAS versus STAT-FAAS method and observed an increased sensitivity by approximately 2.5 times for lead (Table 2). The increased sensitivity is due to the longer residence time of analyte atoms in the beam path of the atomic absorption spectrophotometer (Karakaya and Taylor 1989, Vural and Duydu 1995)

Table 1. Recovery rates and coefficients of variation of urinary lead by STAT-FAAS

Concentration ($\mu g\ Pb^{++}/l$)	Extraction procedure	Average recovery (%)	Coefficient of variation (%)
50	a	97.2	3.1
	b	95.7	3.7
300	a	98.6	2.7
	b	96.2	2.9

a: extraction procedure of total lead, b: extraction procedure of inorganic lead

Table 2. Comparison of conventional FAAS versus STAT-FAAS sensitivities

Flame type	*Sensitivity of conventional FAAS (μg/ml)	*Sensitivity of STAT-FAAS (μg/ml)	Improvement factor
Air-C_2H_2	0.074	0.030	x 2.47

* Sensitivity is defined as the concentration of analyte required to give an absorbance of 0.0044 A.

TEL when absorbed into the body is not excreted in unchanged form. Many investigators showed that lead is excreted in three chemical forms (Et_3Pb^+, Et_2Pb^{++} and Pb^{++}) and Et_2Pb^{++} is considered as a specific indicator of TEL exposure (Turlakiewicz and Chmielnicka 1985; Arai 1986). In our study we determined PbT (organic lead + inorganic lead) and PbI (Pb^{++}) in urine. The urinary excretion of PbT was determined significantly higher ($p>0.05$) than the urinary excretion of PbI in the occupationally TEL exposed worker groups. Furthermore the urinary PbT and PbI concentrations was not significantly different in control group ($p>0.05$, Table 3). These results are in agreement with the earlier studies and could be evaluated as a result of organic lead exposure (Turlakiewicz and Chmielnicka 1985; Arai 1986). We also observed relatively low and very close mean concentration ratios between PbI/PbT in urine of TEL exposed worker groups as compared with the control group (Table 3). Accordingly, it is possible to say that the ratio of PbI/PbT in urine might be also evaluated as an indicator of organolead exposure.

The mean of urinary PbT (PbT-U) and PbI-U concentrations of refinery workers, motor mechanics, gasoline station workers and the control group are shown in Table 3. The lead concentrations in urine were given as µg/l and µg/g creatinine. Creatinine levels in urine and the mean age of the subjects are also included in Table 3.

Table 3 The characteristics and the Pb levels in urine of the control and TEL exposed workers.

	Refinery Workers (n=49)	Motor Mechanics (n=50)	Gasoline Station Workers (n=42)	Control (n=35)
Age (years)	34.2 ± 3.7[a] (30.0 - 46.0)	27.9 ± 8.9 (16.0 - 54.0)	26.5 ± 4.7 (20.0 - 39.0)	22.8 ± 5.8 (18.0 - 37.0)
PbT-U (μg/l)	105.5 ± 59.8[bcd] (27.4 - 239.2)	89.2 ± 49.1[bc] (34.9 - 241.8)	85.5 ± 55.7[bc] (18.1 - 238.5)	51.6 ± 14.2[e] (22.5 - 85.4)
PbI-U (μg/l)	72.3 ± 30.9 (14.7 - 129.9)	62.9 ± 26.5 (27.3 - 122.1)	57.9 ± 33.4 (4.1 - 180.1)	46.7 ± 9.7 (24.1 - 63.7)
PbT-U (μg/g creatinine)	91.1 ± 37.1[bcd] (42.6 - 209.8)	67.3 ± 42.8[bc] (15.9 - 160.7)	74.2 ± 41.1[bc] (22.1 - 189.5)	50.5 ± 17.4[e] (25.5 - 87.8)
PbI-U (μg/g creatinine)	65.8 ± 24.6 (23.3 - 155.7)	48.4 ± 29.3 (10.9 - 124.0)	53.9 ± 35.5 (5.1 - 144.6)	45.9 ± 14.1 (21.4 - 80.3)
Creatinine (g/l)	1.1 ± 0.5 0.6 - 2.2	1.6 ± 0.8 (0.5 - 2.9)	1.2 ± 0.5 (0.6 - 2.3)	1.1 ± 0.3 (0.6 - 1.7)
PbI/PbT (μg/g creatinine)	0.75	0.75	0.72	0.93

[a] Mean ± S.D., (range).
[b]Significantly high from the control group($p<0.05$).
[c] Significantly high from respective PbI-U concentrations ($p<0.05$).
[d] Significantly high from the motor mechanics and gasoline station workers ($p<0.05$).
[e] Not significantly different from the respective PbI-U concentrations ($p>0.05$).

The mean PbT-U levels of the worker groups observed statistically higher ($p<0.05$) than the control group. Figure 1 shows the distribution of the PbT-U levels against the percentage of the workers. When 80 µg/l is accepted as normal urine lead level (Fleming 1964) 42% of the refinery workers, 58% of the motor mechanics and 54% of the gasoline station workers appears to excrete normal level of PbT-U. Urinary PbT level above 110 µg/l could be considered as a sign of untolerable lead exposure (Kehoe 1983). When we evaluated the distribution of PbT-U concentrations against percentage of exposed workers, 41 % of the refinery workers, 21 % of the motor mechanics and 17 % of the gasoline station workers appears to be under risk (Fig 1). According to Kehoe (1983) 150 µg/l and higher urinary lead levels reflects a harmful degree of lead absorption. In our studied worker groups we observed that 12 refinery workers, 6 motor mechanics and 5 gasoline station workers excreted PbT levels higher than 150 µg/l in urine. On the basis of this assumption mentioned above especially these workers should be monitored carefully and surely needs further investigations to define the diagnostic signs of the chronic effects of TEL exposure.

The mean PbT-U concentration in refinery workers were determined significantly higher ($p<0.05$) than the motor mechanics and gasoline station workers (Table 3). Similar study was performed by Turlakievicz et al (1985) in TEL exposed refinery workers. The PbT-U levels in both

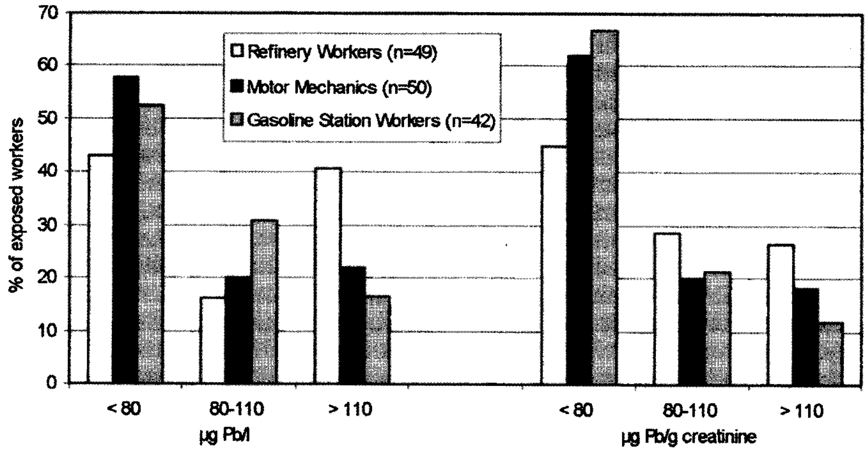

Figure 1. PbT-U levels of TEL exposed workers: <80 µg/l normal PbT-U levels; 80-110 µg/l tolerable PbT-U levels; >110 µg/l should be considered as an indicator of exposure above 'normal'.

studies showed similar range values, but the mean PbT-U levels in our study were found higher than the mean PbT-U levels reported by Turlakievicz et al (1985). The higher mean value in our study may reflect the unhealthy working conditions (Table 4).

In motor mechanics we determined higher PbT-U levels than we expected. We observed PbT-U concentrations higher than 150 µg/l in 6 motor mechanics. These high PbT-U concentrations could be evaluated as a result of TEL penetration through the skin (Cleaning hands with gasoline is a very extensively used procedure among the motor mechanics in Turkey).

We determined also high PbT-U concentrations in gasoline station workers against the control group ($p>0.05$). This result is in agreement with our earlier studies (Vural and Duydu 1995). In 5 workers we observed PbT-U concentrations above 200 µg/l and according to their statements we surprisingly noticed that these five workers also used gasoline as a cleaning agent to clean their hands. When the high PbT-U concentrations in motor mechanics were taken into consideration the high PbT-U levels in 5 gasoline station workers could be also evaluated as a result of TEL penetration through the skin. An extensive study has been performed by Zhang et al (1994) in persons occupationally exposed to tetraethyllead. They reported 78.60 µg/l mean PbT concentration in gasoline station workers. This mean PbT concentration appears in agreement with our mean PbT concentration in gasoline station workers, but unfortunately the range value was not available (Table 4).

Table 4. Mean urinary PbT concentrations reported by other authors.

Employment	n	Urinary PbT (μg/l)	References
Gasoline Station Workers	277	78.60 ± 19.70	Zhang, 1994
Control	342	18.00 ± 10.10	
Refinery Workers	26	73.8 (16.8 - 229.6)	Turlakievicz, 1985
Control	26	24.9 (7.5 - 55.0)	

The control group represents the people living in the center of Ankara and randomized among the office workers. According to many studies the nonoccupational exposure to lead in cities is due to the airborne lead emitted from automobile exhausts (Shy 1990, Zhang 1994). The mean urinary PbT concentration in the control group was found higher than the earlier studies (Turlakiewicz 1985, Zhang 1994) and could be evaluated as a result of the traffic induced air pollution (Table 4). In our previous study the annual mean ambient air lead concentration in central Ankara was found within the range of 0.526 μg/m^3 to 0.821 μg/m^3 (Vural and Güvendik 1987, 1988).

The mean urinary PbT level in the control group could be evaluated as safe (Alessio et al 1987). Nevertheless the chronic effects of low level lead exposure in long term should be also taken in to consideration. Some investigators reported significant cytogenetic effects as chromosomal abberations and sister-chromatide exchanges for the traffic policemen (Anwar and Kamal, 1988). Furthermore many investigations indicated to the adverse developmental effects occur in early life in association with the low level lead exposure (Shy, 1990). Accordingly, in the long term the traffic policemen, children and the pregnant women may constitute a risk group in the city center of Ankara. This situation needs further investigations.

According to our regulations TEL will be used as a gasoline additive until 2003 in Turkey. Until that time occupational TEL exposure will continue in occupations associated with exposure to gasoline or automobile exhaust. Combustion of leaded petrol in thousands of automobiles will continue to contaminate the environment and the people living in the city centers will continue to constitute a risk group in Turkey for more than 5 years.

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