Effects of Mercury Vapour Exposure at Low Concentrations on Urinary Activity of N-Acetyl-Beta-D-Glucosaminidase

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Abstract: Effects of Mercury Vapour Exposure at Low Concentrations on Urinary Activity of N-Acetyl-Beta-D-Glucosaminidase: Nobuo ISHIHARA. Research Centre for Occupational Disease, Tohoku Rosai Hospital—Effects of exposure to mercury vapour less than 0.02 mg Hg/m³ on urinary N-Acetyl-beta-D-glucosaminidase (NAG) activity were studied in seven Japanese workers from 1991 to 1997. According to the record of periodical medical examinations they have engaged in mercury work since 1987 at the latest. Mercury concentrations in urine, plasma, erythrocytes, and hair and the urinary protein concentration did not change significantly during the course of the study. Urinary NAG activity, however, increased significantly in 1996 notwithstanding the absence of significant correlations between NAG activity and mercury concentrations in urine and plasma, and the increased NAG level was maintained up to 1997. All of the present subjects were less than 57 years old in 1997. The effect of aging on urinary NAG activity was therefore excluded, and the increase in urinary NAG activity should be due to the occupational mercury vapour exposure. As in the case of mercury concentrations in blood, urinary NAG activity also should be one of the useful indicators of renal effects of mercury so long as data for before work and/or just after the start of mercury work are available. (J Occup Health 2000; 42:27-30)

Key words: Mercury vapour, NAG activity, Urinary protein, Urinary mercury, Mercury, Plasma

Our previous report indicated that inorganic (I-Hg) and organic mercury (O-Hg) in plasma and O-Hg in erythrocytes increased after four months of exposure to mercury vapour at low concentrations less than TLV, and that this increased steady state of mercury concentrations in blood was maintained until 23 months of exposure without significant increases in I-Hg and O-Hg concentrations in urine. It is therefore concluded that concentrations of I-Hg and O-Hg in blood might be useful indicators of mercury uptake so long as data from before work with mercury are available. In practice the duration of mercury work is more than two yr in the factory. It is therefore necessary to study the effects of mercury vapour exposure for more than two yr.

Several reports indicated the effects of mercury exposure on lysosomal enzyme activities in urine. Langworth et al. reported that the mercury concentrations in blood and urine in chloralkali workers were significantly related to the type of work but not to the length of employment (between 1 and 45 yr; mean 13.5 yr). Langworth et al. reported also in the same subjects a statistically significant relation between the urinary mercury concentration and urinary N-acetyl-beta-D-glucosaminidase (NAG) activity, and a tendency to increased excretion of NAG activity.

The aim of the present report is to study the renal effects of low concentration exposure to mercury vapour for more than two yr.

Subjects

Five Japanese female and two Japanese male workers were studied. They were 37, 37, 38, 38 and 48 years old (female), and 19 and 27 years old (male) at the beginning of the present study (1991). According to the periodical medical examination record they were engaged in mercury battery production since 1987 at the latest under a similar work schedule. Specimens of urine, blood, and hair were sampled on the third Friday of October of 1991, 1993, 1995, 1996 and 1997 in the morning before the start of daily work as described previously. During the course of the study they did not complain of symptoms suggestive of intoxication by mercury vapour. The mercury vapour concentrations in the air in the workplaces were less than 0.02 mg Hg/m³ throughout the observation period.

Received Apr 14, 1999; Accepted Feb 22, 1999
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Methods

The selective determination of I-Hg and total mercury (T-Hg) was carried out by the method of Magos\textsuperscript{12}. The difference between T-Hg and I-Hg values was taken as the value of O-Hg. The detection limits of I-Hg and T-Hg were 0.35 ng of Hg/ml in urine, 0.5 ng of Hg/ml in plasma and erythrocytes, and 0.2 ng of Hg/mg in hair. All determinations were carried out in duplicate. The coefficient of variance was within 4.0% in all determinations. The activity of urinary N-acetyl-beta-d-glucosaminidase (NAG) was determined by the method of Noto et al.'s (NAG test Shionogi (Shionogi & Co., Ltd., Japan)) within 4 h after sampling, and the urinary protein concentration was determined by the method of Bradford\textsuperscript{14} with Tonein TP (Otsuka Pharmaceutical Co., Ltd., Japan). The results of urinary determinations were corrected by the urinary creatinine concentrations determined by the Jaffe’s reaction.

The determination of the exposure level by personal sampling was conducted with the 520 series of passive sampler for mercury (SKC Inc., U.S.A) in 1997.

Statistical Analysis

Changes in concentrations of I-Hg and O-Hg, and in urinary NAG activity and protein concentrations during the period 1991 to 1997 were evaluated by the method of Scheffe (nonparametric) (cited in 15).

Results

Means and SEM of I-Hg and O-Hg concentrations in urine, plasma, erythrocytes and hair are shown in Table 1. Table 2 indicates the means and SEM of urinary NAG activity and protein concentrations. The results of statistical analysis by the method of Scheffe are in Table 3. It is clear that a significant increase was observed only in urinary NAG activity on 1996, and that mercury concentrations in urine, plasma, erythrocytes and hair and the urinary protein concentration remained almost constant during the course of observation from 1991 to 1997. No significant correlations were observed between urinary NAG activity and mercury concentrations in urine and/or plasma. Urinary protein concentrations did not have significant correlations with urinary NAG activity. Table 4 shows the results of personal sampling of the

Table 1. I-Hg and O-Hg concentrations in urine, plasma, erythrocytes and hair

<table>
<thead>
<tr>
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</thead>
<tbody>
<tr>
<td>urine I-Hg</td>
<td>5.77</td>
<td>2.72</td>
<td>4.66</td>
<td>4.65</td>
<td>4.31</td>
</tr>
<tr>
<td>O-Hg</td>
<td>1.15</td>
<td>0.34</td>
<td>0.30</td>
<td>1.00</td>
<td>0.69</td>
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<tr>
<td>plasma I-Hg</td>
<td>2.21</td>
<td>0.37</td>
<td>1.48</td>
<td>0.56</td>
<td>0.84</td>
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<tr>
<td>O-Hg</td>
<td>2.35</td>
<td>1.52</td>
<td>1.28</td>
<td>2.37</td>
<td>1.29</td>
</tr>
<tr>
<td>RBCs I-Hg</td>
<td>0.91</td>
<td>1.18</td>
<td>5.80</td>
<td>0.73</td>
<td>1.14</td>
</tr>
<tr>
<td>O-Hg</td>
<td>37.36</td>
<td>32.69</td>
<td>38.50</td>
<td>25.72</td>
<td>27.06</td>
</tr>
<tr>
<td>hair I-Hg</td>
<td>0.65</td>
<td>0.33</td>
<td>0.55</td>
<td>0.34</td>
<td>0.43</td>
</tr>
<tr>
<td>O-Hg</td>
<td>3.39</td>
<td>2.74</td>
<td>2.96</td>
<td>2.99</td>
<td>3.87</td>
</tr>
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</table>

Values in the table are means and SE (in parenthesis). urine: ng Hg/mg creatinine, plasma and RBCs: ng Hg/ml, hair: ng Hg/mg; n=7.

Table 2. Urinary NAG activity and protein concentration

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<tbody>
<tr>
<td>NAG</td>
<td>2.7</td>
<td>3.3</td>
<td>3.2</td>
<td>6.2</td>
<td>3.8</td>
</tr>
<tr>
<td>protein</td>
<td>0.099</td>
<td>0.178</td>
<td>0.183</td>
<td>0.129</td>
<td>0.135</td>
</tr>
</tbody>
</table>

Values in the table are means and SE (in parenthesis). NAG: units/g creatinine, protein: g/g creatinine, n=7.

Table 3. Statistical analysis of means by the method of Scheffe urine plasma RBC's hair

<table>
<thead>
<tr>
<th></th>
<th>I</th>
<th>O</th>
<th>I</th>
<th>O</th>
<th>I</th>
<th>O</th>
<th>I</th>
<th>O</th>
<th>NAG protein</th>
</tr>
</thead>
<tbody>
<tr>
<td>'91 v '93, '95, '96, '97</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
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<td>ns</td>
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<td>'91, '93, '95 v '96, '97</td>
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<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>p&lt;.05 ns</td>
</tr>
<tr>
<td>'91, '93, '95 v '96, '97</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
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<td>ns</td>
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<td>ns</td>
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<td>ns</td>
</tr>
</tbody>
</table>

I: inorganic mercury, O: organic mercury, ns: not significant.
Discussion

As shown clearly in Table 1, I-Hg and O-Hg concentrations in urine, plasma, erythrocytes and hair did not change significantly during the course of the present study notwithstanding the clear occupational exposure to mercury (Table 4). This fact indicated that the intake of mercury due to occupational exposure and to diet should be almost constant throughout the study. In the previous report, I-Hg and O-Hg concentrations in blood reached a high steady state after 4 months of exposure to mercury vapour, and this state was maintained for about 2 yr. Mercury concentrations in the blood and urine of the present subjects in 1991 were almost the same as their respective values determined after about two years of exposure to mercury vapour. It was therefore concluded that the mercury concentrations in the blood of the present subjects had already reached to a high steady state in 1991, and that this state was maintained up to 1997 notwithstanding continuous exposure to mercury vapour at concentrations less than TLV. As claimed in the previous report, this result indicated that concentrations of mercury in blood might be useful indicators of mercury uptake so long as data from before work with mercury are available.

Several reports indicated the relationships between mercury exposure and lysosomal enzyme activities in urine. Langworth et al. reported a tendency to increased excretion of NAG in the group exposed to mercury vapour at low concentrations. But the duration of exposure required to increase the urinary excretion of NAG remained unclear. According to the record of medical examinations of the present subjects, it was clear that all of the subjects have been engaged in mercury work since 1987 at the latest. On the other hand, as shown clearly in Table 3, urinary NAG activity had increased significantly in 1996 compared to from 1991 to 1995. It was suggested, therefore, that at least nine years of exposure to mercury vapour at low concentrations less than TLV might be required to increase urinary excretion of NAG activity.

In the present study, because the urinary activity of NAG in non-exposed subjects was not determined, several factors except mercury which might increase urinary excretion of NAG activity should be considered. Among them the effects of aging might be most important. It was indicated, however, that urinary NAG activity corrected by urinary creatinine concentrations did not change significantly in the non-exposed subjects aged 59 yr or less. Even in 1997 all of the present subjects were less than 54 yr old. It is therefore concluded that the increase in urinary NAG activity observed (Table 3) might be due to the occupational exposure to mercury vapour but not to aging. Because the present subjects were not exposed to cadmium or lead, the effects of these metals could be excluded. Organic solvents also were not used in this production line. High blood pressure was not observed in any of the present subjects. The effect of renal failure due to high blood pressure could be excluded. It is concluded, therefore, that the observed increase in urinary excretion of NAG activity is due to the exposure to mercury vapour.

The increase in NAG activity suggested that some kind of disturbance occurred in the renal tubuli. Nevertheless, not only urinary I-Hg and O-Hg concentrations but also urinary protein concentrations remained unchanged notwithstanding the clear increase in urinary NAG activity on 1996, and no significant correlations were observed between urinary mercury concentrations and urinary NAG activity. The disturbances of renal tubuli, therefore, might be scant.

The present results indicated that urinary NAG activity should be a useful indicator so long as data from just after the engagement in mercury work, or from before work with mercury, are available.

Acknowledgment: I am indebted to Dr. Ikuro Satoh (Department of Pathology, Miyagi Cancer Center, Japan) for his help in the statistical analysis, and to Shionogi Co. Ltd. for several suggestions concerning the determination of NAG activity.

References

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