LETTERS TO THE EDITOR

MERCURY UPTAKE IN VITRO BY IRON COMPOUNDS, PROTEINS (ALBUMIN OR GLOBULIN), WITH OR WITHOUT HYDROGEN PEROXIDE

Metallic mercury is widely used in electrolytic electrodes, gauges (thermometers, cup barometers, etc), mercury lamp, as mercury compounds and the amalgam of dental surgery. Inhalation of mercury vapor causes pneumonia and renal disorders. The toxicity of mercury seems to arise and increase through the accumulation of mercuric ion produced from the metallic mercury in the body. The oxidation products of metallic mercury are reported to be mercurous ion, Hg+, and mercuric ion, Hg2+ 1). It is well known that these inorganic mercury ions are widely distributed in the blood, especially the blood plasma, whereas metallic mercury occurs in red blood cells2) Clarkson et al3) demonstrated in 1961 that elemental mercury was rapidly oxidized by human blood in vitro and in vivo. Subsequently, in 1967, Magos4) reported that the elemental mercury in blood exposed to mercury vapor in vitro was converted to mercuric ion at a slow speed. Kudsk5) found that methyl alcohol inhibited the uptake of mercury by blood in vitro and in vivo. Magos, Sugata, and Clarkson6) reported that red blood cells preincubated with 3-amino-1,2,4,-triazole in the presence of methylene blue, revealed a decrease of catalase activity and of mercury uptake in air saturated with mercury vapor. These findings suggest that primary catalase-hydrogen peroxide complex may be involved in the oxidation process of mercury.

In order to establish the precise role of blood catalase in the uptake process of mercury, a blood sample containing no catalase is necessary. Acatalasemia is a rare congenital abnormality, first described by Takahara7) in 1952 and is characterized by a deficiency in catalase activity8). Radiation induced hypocatalasemia and acatalasemia mice have been reported by Feinstein et al.9) In a previous study10), we found that erythrocytes, lung and liver homogenates prepared from acatalasemia mice obtained by the above method showed decreased mercury uptake from air saturated with mercury vapor as compared to normal mice. Acatalasemia mice exposed to mercury vapor revealed lower levels of mercury ions in the lung and the blood than normal mice, although the amount of mercury ions in the whole body was larger than normal. It was also clarified that human acatalasemic erythrocytes had only 1 to 6% of the total uptake amount found in normal erythrocytes with hydrogen peroxide, and 6 to 24% of the uptake without hydrogen peroxide, on examining their in vitro ability to take up mercury from air saturated with mercury.

This experiment indicated that catalase played an important role in the uptake of metallic mercury in vitro with normal, hypocatalasemic and acatalasemic blood. However, acatalasemia mice exposed to metallic mercury accumulated mercury ions in the
liver than normal mice, although the blood and lung accumulated less mercury ions than in normal mice. We attempted to find other ferric or ferrous compounds in addition to catalase which were involved in the uptake of metallic mercury. There are numerous organic compounds containing ferric and ferrous ion in the body, especially the liver. The sample of iron compound was placed in the main chamber of a 15 ml Warburg flask with 0.1 ml of metallic mercury in a side-arm and with 0.1 ml of 0.3% hydrogen peroxide in the center well. Incubation was carried out at 37°C for 90 min. with shaking at 80 cycles/min. The iron-mercury system in the absence of hydrogen peroxide, revealed no change in mercury uptake even when the concentration of ferric ion increased. In the presence of hydrogen peroxide (0.3%), the mercury uptake by the system showed an increasing tendency. The results are illustrated in Fig. 1. On the other hand, when

![Fig. 1. Effect of Fe³⁺ on the uptake of mercury vapor.](attachment:figure1.png)

the concentration of ferric ion was constant, the mercury uptake increased in relation to the concentration of hydrogen peroxide. Thus, hydrogen peroxide may be considered to exert a significant effect on the mercury uptake by ferric ion.

Concerning the underlying mechanism, Deisseroth et al.\(^\text{11}\) indicated that ferric ion is transformed into Fe³⁺OOH by hydrogen peroxide. This Fe³⁺OOH takes up mercury and is itself converted to Fe³⁺OH. The reaction is thus considered to be a recyclic reaction. In order to confirm the reaction mechanism further, the pH value before and after the incubation in the presence of hydrogen peroxide was measured. The pH of 0.9 increased to 1.6 after the incubation. This increased value may result from the formation of Fe³⁺OH from Fe³⁺OOH.

When proteins (albumin* or globulin***) and a trace of ferric ion coexisted in the reaction system, mercury uptake by the proteins could hardly be recognized, but it
increased with increase in the ferric ion concentration (Fig. 2). In the case of a fixed concentration of ferric ion and absence of hydrogen peroxide, even when the concentration of proteins increased, the mercury uptake hardly reached the highest value at an albumin or globulin concentration of 0.5 mg/ml in the system. Increase of the protein concentration exerted no effect on the mercury uptake. The proteins appear, therefore, to take up mercury in presence of excess ferric ion. However, there are many iron compounds, ferritin, etc., other than catalase, which are contained in the body and discharged in an anemic state. It is suggested that these compounds may also play a role in the uptake of mercury.

When ferrous ion was employed instead of ferric ion in the presence of hydrogen peroxide, the mercury uptake was larger than that in the ferric ion system. Since the possibility exists that ferrous ion could change into ferric ion during the incubation, it is necessary to take the mole ratio of ferric and ferrous ions into account. It is considered that through the process (I)\(^{12}\), \( \text{Fe}^{2+} + \text{H}_2\text{O}_2 \rightarrow \text{Fe}^{3+} + \text{OH}^- + \text{OH}^- \), ferrous ion is converted to ferric ion by hydrogen peroxide, and in process (II)\(^{12}\), \( \text{Fe}^{3+} + \text{O}_2^- \rightarrow 2\text{Fe}^{2+} + \text{O}_2 \), when the concentration of peroxide is large relative to consumption of ferrous ion initially present, and pH is higher, other reaction may occur including a chain which leads to evolution of oxygen. The mercury uptake of process (I) was found to be larger than in process (II). The results are shown in Fig 3. Change in the mole ratio in the absence of hydrogen peroxide had no effect on the mercury uptake. On the other hand, when the concentration of ferrous ion was increased with respect to ferric ion in the presence of hydrogen peroxide, the mercury uptake revealed an increasing tendency until the ratio

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**Fig. 2.** Mercury uptake by albumin or globulin addition, with or without \( \text{H}_2\text{O}_2 \).

- A: Albumin with \( \text{H}_2\text{O}_2 \)
- A': Albumin without \( \text{H}_2\text{O}_2 \)
- B: Globulin with \( \text{H}_2\text{O}_2 \)
- B': Globulin without \( \text{H}_2\text{O}_2 \)
rose to 0.1. When the ratio exceeded 0.1, the uptake decreased. Furthermore, the concentration of ferrous ion in the absence of hydrogen peroxide was increased after the incubation compared to before, while it decreased in the presence of hydrogen peroxide.

* The Fe(III) in albumin is 44.0 µg/g.

** The Fe(III) in globulin is 130.0 µg/g.

REFERENCES

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