PROBLEMS OF INTERACTIONS IN THE TOXICITY OF METALS

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ABSTRACT

The paper is based mainly on discussions, conclusions and recommendations from a workshop at the Karolinska Institute, Stockholm, July 1977, organized by the Scientific Committee on the Toxicology of Metals under the Permanent Commission and International Association on Occupational Health.

Different forms of joint actions of metals are discussed. Available data concerning the interaction between the toxic metals themselves are too scanty to draw conclusions for humans. More data exist about interactions between the toxic metals and certain essential elements. The need for extended studies in this field is emphasized both in relation to synergistic and antagonistic effects.

It is well known that interactions between different toxic substances may modify the toxicity of single substances and thus play a fundamental role in toxicology. Well-known examples are the remarkably increased incidence of lung cancer after exposure to cigarette smoke in conjunction with asbestos or radon. Both examples are from industry but the combined exposure to radon and cigarette smoke may have vast implications for the general population as in several countries they are exposed to a combination of tobacco smoke and radon from the building material. Such an interaction may be of importance not only for the smoker, but also for nonsmokers exposed to tobacco smoke (passive smoking).

The most well-known interactions with metals are the effects of therapeutic agents like EDTA and BAL. These substances are widely and successfully used .to increase the excretion of toxic metals. Caution must be exercised though, as the use of such chelating agents may cause a redistribution of metals in the body giving rise to increased toxicity under certain circumstances. This has been shown in connection with both cadmium and mercury intoxication. Wide publicity has been given to the possible protective action of selenium in

connection with exposure to methylmercury via fish. There are selenium tablets commercially available intended to be used to protect against a number of metal intoxications.

Despite the fact that today there is substantial evidence that the metabolism and effect of certain toxic metals may be changed considerably by various factors, no systematic treatise of this fundamental aspect of metal intoxication has been done up till recently. The Scientific Committee on the Toxicology of Metals under the Permanent Commission and International Association on Occupational Health arranged a year ago a small symposium in Stockholm at the Karolinska Institute to discuss factors influencing metabolism and toxicity of metals.

The meeting was a continuation of previous efforts taken by the Committee to evaluate metabolism and toxicity of different metals*. The symposium dealt with interactions between different toxic metals, and with interactions between such metals and certain essential elements. In addition, factors such as age and sex were considered. Although interactions have been dealt with before within different disciplines, the Stockholm meeting was probably the first time when interactions were dealt with from such a broad perspective by international experts representing a variety of fields including biochemistry, ecology, epidemiology, nutrition, occupational health and toxicology. The results presented here are to a great extent based on discussions, conclusions and recommendations from the Karolinska Institute meeting.

Before going into details concerning our present knowledge of interactions it may be useful to dwell on some definitions which were agreed upon. In a broad sense the term "interaction" should be used to describe any influence of metals and other substances and factors on the metabolism and the toxicity of the metal under consideration. Of particular importance, however, is the process by which metals in their various forms change the critical concentration or critical effect of a metal under consideration, e.g. to what extent a substance may change the concentration of cadmium in the kidneys (the critical organs in cadmium intoxication) which may give rise to the first symptoms (the critical concentration).

When man is exposed to toxic metals, different forms of joint actions may occur. Such actions can be classified as non-interactive or interactive actions. Among the non-interactive actions, the most common is an independent action. This occurs when two substances act at different sites and have different mechanisms of action. The result is two different effects. An additive effect is also a form of non-interactive action, but will be included in this presentation. Such an effect can be seen when two metals exert their effects on the same site and when the mechanism of action is similar. It is important to point out that an additive effect does not necessarily mean that the total effect will be double that of the effect of one of the substances alone.

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* A report from the meeting as well as the working papers on which the discussions were based will be published. Reports from previous meetings organized by the Scientific Committee on the Toxicology of Metals have been published (2,5,6).
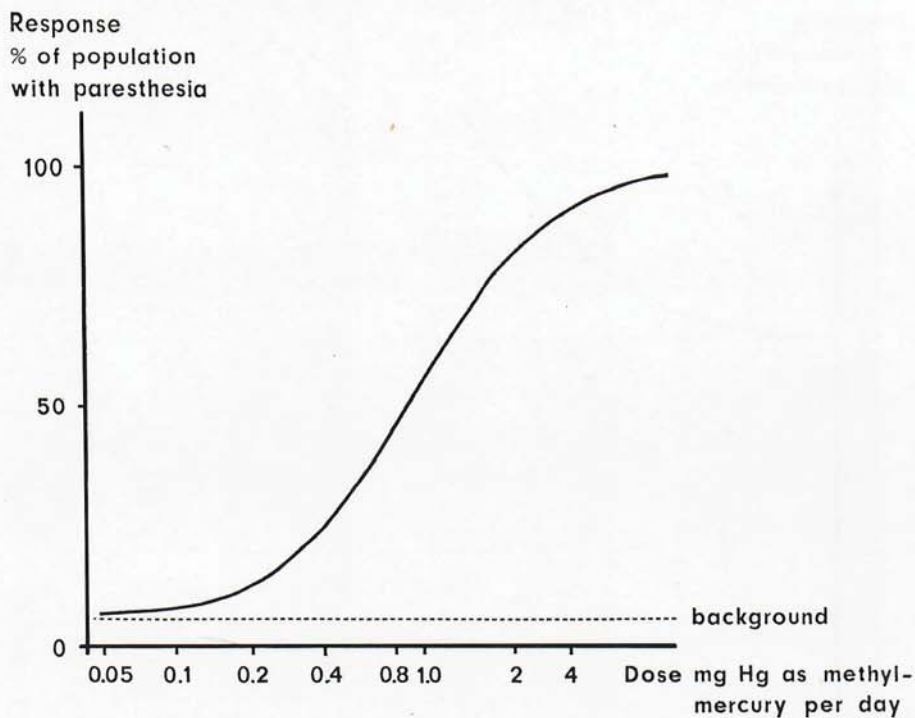
Response
% of population
with paresthesia

FIG. 1 — Dose-response curve for methylmercury. Background response assumed to be 5%. Data derived from Bakir *et al.*[1] and Nordberg and Strangert[7].

Figure 1 depicts the usual sigmoid shape dose-response curve which in this particular case is derived from the incident in Iraq where a large number of people were poisoned as a result of eating seed contaminated with methylmercury. The following discussion should, however, be valid for dose-response curves in general. It is evident from the figure that the influence on the response rate of a doubling of the dose will be quite different depending on which dose (and response) interval is studied and whether the background response is considered or not. The greatest absolute change in response will take place in the mid-part of the curve where a change in the dose from 0.4 to 0.8 would mean a change in response from 27% to 50%, i.e. a difference by 23% or a factor slightly less than 2 (1.85) when the background response rate of 5% is included. If the background response rate is subtracted, the increase will amount to a factor of slightly more than 2 (2.05).

When the low dose part of the curve is considered, the situation will be quite different (Fig. 2). A change in dose from 0.05 to 0.1 would mean a change in response from 5.3 to 6.8%, i.e. a change in response of only 1.5% corresponding to a factor of 1.28 when the background is included, but a factor of 6.3 when the background is subtracted. It is thus demonstrated that there is a
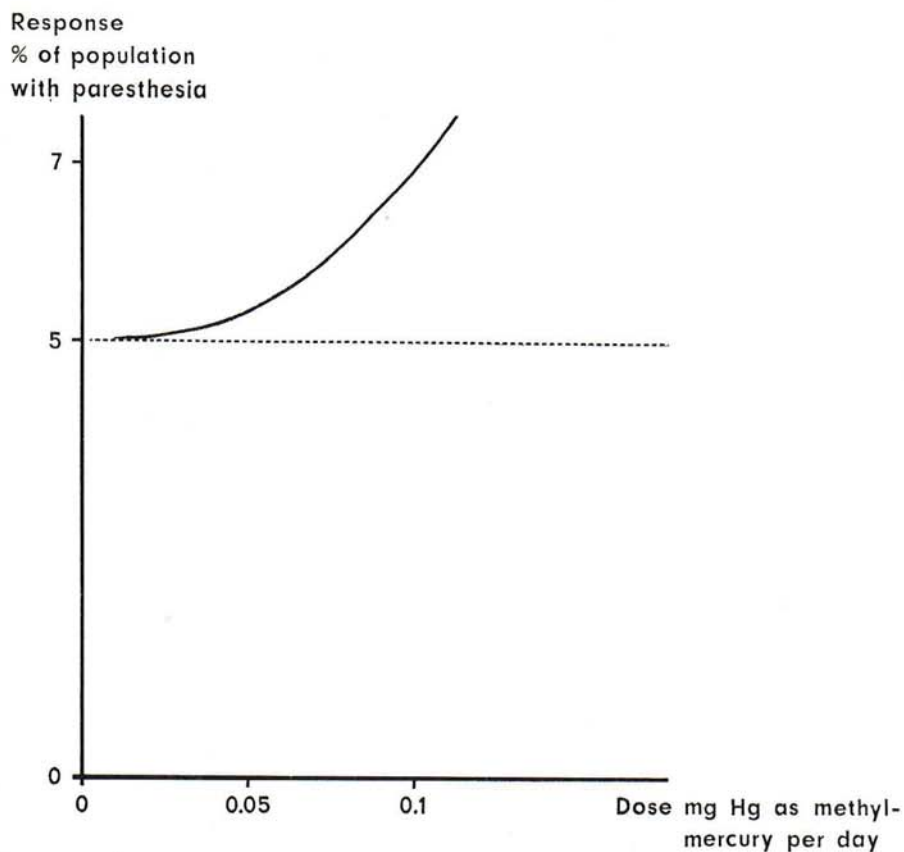
Response
% of population
with paresthesia

FIG. 2 – Lower dose part of dose-response curve shown in Figure 1.

high relative increase in response when background is subtracted and the opposite when background is included. If only the very high dose part of the curve is considered, the increase when doubling the dose will be only marginal.

When dealing with true interactions it is not possible to predict the combined effect of two substances automatically from the separate dose-response curves for the different substances. There may be synergistic as well as antagonistic effects. Of particular importance is the synergistic action which means that the combined effect or response will be greater than additive. The examples given before in connection with cigarette smoke, asbestos and radon are usually considered to be synergistic actions. An antagonistic action means that the exposure to one substance reduces the effect of another substance. The therapeutic effect of chelating agents as well as the protective effect of selenium against the toxicity of certain other metals are examples of antagonistic actions.

The toxic metals primarily discussed during the symposium were arsenic, cadmium, lead and mercury. There are only very scanty data concerning

interactions among these metals themselves, while more data are available concerning interactions between the toxic metals and certain essential metals.

The lack of data concerning interactions between arsenic, cadmium, lead and mercury themselves is of great concern, since an exposure to such metals often occurs simultaneously. Although little animal data are available, studies have shown that lead increases protoporphyrin excretion in arsenic treated animals, while a similar effect was not seen after cadmium exposure. There is also some evidence that cadmium may interact with mercury as this metal, like cadmium, binds to the low molecular weight protein metallothionein. Mercury binds more firmly to metallothionein than cadmium and may displace cadmium from metallothionein and modify the toxic effect. No human data are available and the animal data can not be used to draw conclusions for humans.

A large number of studies have been carried out where interactions between arsenic, cadmium, lead and mercury on the one hand, and elements like calcium, copper, iron, zinc and selenium on the other hand, have been studied. Most studies are based on animal experiments, but in some cases human data are also available.

In animals both calcium and iron deficiency has conclusively been shown to influence the absorption of cadmium and lead from the gut. Thus, a low calcium intake as well as iron deficiency will increase the absorption of both cadmium and lead. A low calcium intake can easily double the amount of cadmium and lead absorbed from the gut. These results may well have implications for humans. There is reason to believe for example that calcium deficiency has been of importance for the development of osteomalacia in Itai-itai disease seen in Japan after heavy exposure to cadmium. It is also of interest to note that when osteomalacia occurred after industrial exposure to cadmium, it was only observed during and immediately after World War II in countries where the nutritional status could be expected to be low. In Sweden, where the calcium intake was rather high, no cases of osteomalacia could be detected in industries despite heavy cadmium exposure. There are certainly other factors which may also be of importance. It has, for example, been suggested that at high renal concentration of cadmium, this metal might interfere with the synthesis of vitamin D_3 in the kidney. This could then severely interfere with the bone metabolism.

As mentioned, animal data have clearly shown that iron deficiency increases the absorption of both cadmium and lead. Recent data have confirmed these findings in humans concerning cadmium. In a report by Flanagan and coworkers[4], it could be shown that iron-deficient volunteers given radioactive cadmium showed a four-fold increase in absorption of cadmium as compared to subjects with normal iron stores. The effect was most pronounced in females (Fig. 3). It should be mentioned that the iron deficiency was only moderate. The subjects had no overt anemia and low body iron was diagnosed when serum ferritin levels were below 20 ng/ml. The importance of these findings is obvious as iron deficiency is common, particularly in women. This may, at least partly, explain the fact that body burdens of cadmium in women are usually higher than in men. The safety margin between cadmium levels found in the kidneys of the
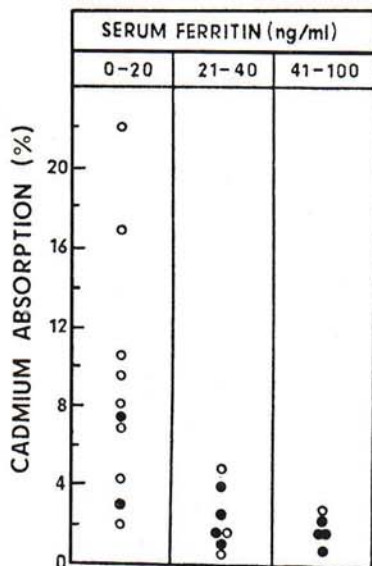
SERUM FERRITIN (ng/ml)

FIG. 3 – Cadmium absorption in human volunteers in relation to body iron status. Females, open circles; males, closed circles (from Flanagan et al.[4]).

general population and concentrations which may be associated with renal dysfunction is small; depending on countries, varying between 2–7. We are now talking about safety values based on mean concentrations within a group of exposed people. If individual values are considered, the situation is more alarming. Also in countries with mean kidney levels considerably below the critical concentration of about 200 µg/g Cd, individual values may be high. Sweden is usually considered as a low-exposure country when it comes to cadmium. Mean concentrations of cadmium in the kidney cortex are only about 20 µg Cd/g wet weight. As can be seen from Figure 4, the distribution is log-normal, and a small part of the population will be close to the concentrations where renal dysfunction can be excessive. There are several indicators that the cadmium exposure will increase for the general population. The figure shows what will happen if the mean concentration of cadmium in the kidney cortex in the Swedish population should increase from the present 20 µg Cd/g to about 50 µg Cd/g wet weight. More than 5% of the population would get a cadmium concentration in the kidney cortex above 200 µg/g wet weight, which has been considered a probable critical concentration where kidney dysfunction may occur[8]. It should be of value to carry out a more systematic study to examine possible reasons for individual high values, and it is obvious that iron deficiency should be looked for as a possible important cause.

Also zinc deficiency will strongly enhance the risk for cadmium intoxication in animal experiments and there are some data which tend to show the same for lead intoxication. Zinc is an essential element being part of several essential enzyme systems. It is also contained to a great extent in the reproductive system. Large amounts of cadmium given in single doses can cause testicular atrophy. If
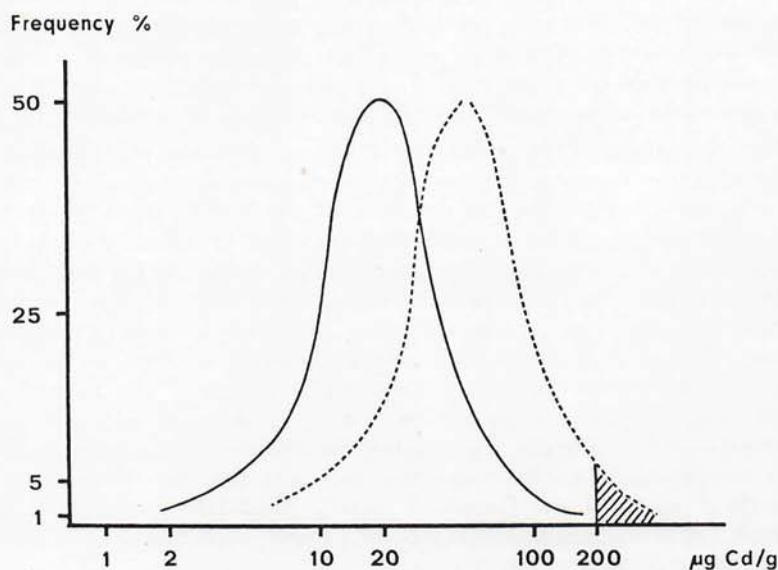
Frequency %

FIG. 4 — Relative frequency of cadmium concentrations in kidney cortex among Swedes 1974[3] (solid line) and a hypothetical curve corresponding to a geometric mean of 50 μg Cd/g (dotted line).

zinc is given in high doses simultaneously with cadmium, this damage can be prevented. Animal data have also shown that cadmium given in high doses by injection can give rise to fetal malformations. Cadmium in itself does not penetrate the placenta to any substantial degree, and it is instead thought that the malformations may be a result of a secondary zinc deficiency. Since even moderate oral exposure to cadmium may give rise to perturbations in zinc metabolism, effects of cadmium exposure on the offspring can be expected when maternal zinc supply is marginal.

The essential element selenium, in itself toxic at high concentrations, interacts primarily with mercury, cadmium and arsenic, but also with lead. Animal data provide evidence that selenium decreases the toxicity of mercury and cadmium. It has for example been shown that selenite reduces the lethal effect of inorganic mercury in rats, and prevents the developments of renal tubular and intestinal necrosis. Long-term administration of selenate has a protective effect against several effects of mercuric chloride. It has been suggested that the formation of a mercuryselenium protein complex may explain alterations in the metabolism and toxicity of mercury.

There are also data which show that if selenite is added to the diet, the toxic effect of methylmercury is reduced in chickens, quail and rats. It has been suggested that selenium present in sea fish might protect against the toxicity of dietary methylmercury. The evidence for this is inconclusive, however, and furthermore the considerable differences between animal species and humans

with respect to methylmercury metabolism and toxicity do not make it possible to draw conclusions for humans on such a protective effect. However, the data at hand certainly motivate more studies, particularly studies on animals with a metabolism more similar to that of man than the species studied up till now.

The mechanism of the protective effect of selenium on cadmium and mercury toxicity is not clear. In some cases selenium increases the concentrations of the toxic metals despite the fact that it exerts a protective effect against them. For example, the prevention of the necrotizing effect of cadmium in the testes, offered by selenium, is coupled with a several-fold increase in the concentration of cadmium in this organ. Selenium also increases the concentration of cadmium in indicator media, such as blood, while reducing the general toxicity of cadmium. This effect was observed only at levels of selenium exceeding the nutritional requirement for this essential trace element. The formation of cadmium-selenium protein complexes may only provide protection against acute effects produced by cadmium. The gradual breakdown of these complexes may provide an opportunity for the synthesis of metallothionein which would protect against these acute effects. However, chronic renal effects are likely to be unaffected, since cadmium bound to metallothionein is considered to be involved in producing this type of toxicity.

In some cases, the available data on interactions have clearly shown the importance of such interactions while in other cases the data may just suggest a potential important interaction for humans. From the practical point of view, it must be recognized that the results might well be of potentially great significance to human health. In several developing countries, but also in certain socio-economic strata of the developed countries, the intake of important essential elements may be inadequate or at least sub-optimal. In the past, animal experiments generally have been carried out without proper consideration given to the importance of a simultaneous exposure to elements like calcium, iron, zinc, and selenium. It seems that more consideration should be given to standardize the diet with respect to such substances. Furthermore, when analyses are made of toxic substances like cadmium and mercury the concentration of possible interactive elements should be analyzed in tissues. The same holds true in connection with analyses of autopsy materials where tissue and body levels of toxic elements are studied.

There are other nutritional factors which have been shown to play a role in metal toxicology. For example, it has been shown in animal studies that a low protein content of the diet can enhance the risk for cadmium and lead toxicity through a higher absorption of the toxic metals. As was the case with calcium deficiency, a protein deficiency may have been a factor of importance for the development of the Itai-itai disease, ultimately caused by cadmium exposure.

Milk, which was initially recommended to prevent lead intoxication, has been shown to have the opposite effect in animal experiments of both neonatal and adult animals. A milk diet increased considerably the absorption of both lead and cadmium. The cause for this is not known, but it has been suggested that the lipid content may be one possible factor.

In addition to the considerations mentioned up till now, interactions of metals with various other factors such as age, sex, irritating substances including smoking, were discussed at the symposium. There seems to be no doubt that animal experiments conclusively indicate age specific differences in the metabolism of lead, cadmium and mercury. Results obtained on suckling rats show a higher rate of intestinal absorption of all metals, a higher whole body retention, higher blood levels and much higher accumulation in the brain as well as a higher toxicity than in the adult rat. It seems as if the increased intestinal absorption rate during the neonatal period is not specific and thus applies to a large number of metals. This may very well be of great practical importance although there are no conslusive data on humans which prove such an effect. Of particular importance may be the exposure to lead and mercury via mother's milk. There is also evidence that children experience symptoms at a lower blood level concentration than adults, which again stresses the possible importance of animal data for humans.

We have already referred to the increased absorption of certain metals due to iron deficiency. This could be one explanation for, for example, a higher body burden of cadmium in females. Other reasons could be pregnancy, lactation where the demand for calcium and other essential elements is increased and where marginal deficiencies of essential elements may be of particular importance.

Of special importance within industry seems to be the possible role of synergistic effects between metals and other atmospheric pollutants like sulfur dioxide, nitrogen oxide and tobacco smoke. There could be no doubt that theoretically the effect of irritating substances on the airways could influence deposition and absorption of particulate matters. This could then alter also the bronchial and pulmonary clearance of deposited particles. Very few data are available, however, to show the role of such irritating substances for the toxicity of metals. The need for further studies in this field is substantial. This is of importance both for the possible role of irritating substances acting as co--carcinogens, which has been suggested in connection with arsenic exposure and sulfur dioxide exposure, and studies concerning uptake, depositon and clearance after simultaneous exposure to metals and irritating substances. Experience from other areas clearly shows that tobacco smoke may potentiate other carcinogens such as asbestos and radon gas.

We feel that the Stockholm meeting clearly demonstrated a fundamental role of interactions. Available evidence for such interactions has however been taken into consideration only to a very limited extent in the preparation of criteria documents and standards for the working environment as well as for the general environment. The need for extended studies in this field is very great both in relation to synergistic and antagonistic effects. It is our feeling that the importance of interactions has been accepted just recently. In all probability interactions will become a very important research area in the years to come, and thus better knowledge in this field very well may considerably change our opinion concerning the toxicology of metals.

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