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SCIENTIFIC COMMITTEE ON TOXICITY, ECOTOXICITY AND THE ENVIRONMENT (CSTEE)

Opinion on the results of the Risk Assessment of:

ZINC METAL (CAS NO. 7440-66-6)
ZINC CHLORIDE (CAS NO. 7646-85-7)
ZINC SULPHATE (CAS NO. 7733-02-0)
ZINC DISTEARATE (CAS NO. 557-05-1, 9105-01-3)
ZINC PHOSPHATE (CAS NO. 7779-90-0)
ZINC OXIDE (CAS NO. 1314-13-2)

HUMAN HEALTH PART

Carried out in the framework of Council Regulation (EEC) 793/93 on the evaluation and control of the risks of existing substances[1]

Adopted by the CSTEE during the 39th plenary meeting of 10 September 2003

[1] Regulation 793/93 provides a systematic framework for the evaluation of the risks to human health and the environment of those substances if they are produced or imported into the Community in volumes above 10 tonnes per year. The methods for carrying out an in-depth Risk Assessment at Community level are laid down in Commission Regulation (EC)1488/94, which is supported by a technical guidance document.

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INTRODUCTION

Zinc and its salts are used for a variety of purposes in industry and consumer applications resulting in human exposure from many different sources. In addition, zinc is an essential nutrient with a recommended intake of 5 - 20 mg/person/day. The average intake in Europe is 9 – 15 mg/person/day.

Terms of Reference

In the context of Regulation 793/93 (Existing Substances Regulation) and on the basis of the examination of the Risk Assessment Report provided by the European Chemicals Bureau, the CSTEE is invited to examine the following issues:

(1) Does the CSTEE agree with the conclusions of the Risk Assessment Report?

(2) If the CSTEE disagrees with such conclusions, the CSTEE is invited to elaborate on the reasons for this divergence of opinion.

GENERAL COMMENTS

The document follows the recommendations of the TGD and is comprehensive, well written and clearly structured. Regarding human health related endpoints, the individual documents on zinc metal and zinc salts are almost identical and cross reading of data from different zinc salts has been performed for hazard assessment except for the endpoints acute toxicity, irritation, corrosivity and sensitisation. The cross reading is justified by the assumption that the zinc ion (Zn^{2+}) is the biologically active agent. The CSTEE supports the cross reading as appropriate and therefore provides only one comment for the six RARs.

SPECIFIC COMMENTS

1. Exposure assessment

For the exposure assessment of workers to zinc metal and the different zinc salts, the RAR considers typical exposure scenarios for the individual compounds and mainly relies on measured data submitted by industry on air concentrations of zinc and the different zinc salts during the typical exposure scenarios. The measured data are in part supplemented by model calculations using EASE and by measured data on zinc concentrations in air at work places handling different zinc compounds. It is concluded that occupational exposure to zinc and its salts occurs mainly by inhalation and by dermal contact. Due to the low uptake of zinc and its salts through the skin, dermal contact only results in low systemic doses. The RARs indicate that the exposure assessment is highly conservative because high retention and absorption rates of the zinc salts are assumed. These are considered unrealistic based on particle sizes of the materials actually used.

Consumer exposure to zinc and zinc salts mainly occurs from the use of zinc in cosmetics or paint and a cumulative uptake of approx. 1.6 mg Zn^{2+}/day is used. The RAR also acknowledges that a much higher exposure to zinc salts may occur from the use of dietary supplements. However, this source of exposure is difficult to quantify since use patterns are not well defined. Based on surveys, a major contribution of zinc intake from dietary supplements to the population average is not expected. However, there may be high zinc intake in some parts of the population due to the abuse of zinc containing dietary supplements. Therefore, the RAR should include information on ranges of human exposures to zinc salts instead of giving a population average.

Indirect exposure to zinc occurs mainly from food and may vary due to food habits. The RAR also acknowledges the homeostatic mechanisms for zinc uptake from the gastro-intestinal (GI) tract which is dependent on the zinc status of the organism. Zinc, like many other chemicals, is transferred from the serum to the milk. Breast-feeding is thus a major source of this essential trace element for suckling new-borns.

So far, even in situations where mothers were exposed to high levels of chemical contaminants in heavily polluted areas, milk zinc concentration has not been reported to be significantly increased. This is most likely due to the very efficient local system of regulation of zinc homeostasis. Exposure of lactating mothers to high levels of zinc does not represent a potential for increased exposure of the suckling babies.

Given the biological interference of copper and zinc, if a simultaneous exposure is possible, it is worth verifying that the optimum ratio of 7 of zinc to copper is respected for an optimum bioavailability.

2. Effects assessment

As mentioned before, most of the health effects are assessed by cross reading and therefore this chapter is highly overlapping for the six compounds. Beneficial effects of zinc are not considered in the RAR.

Acute toxicity

In experimental animals, the acute toxicity of zinc and its salts after oral and dermal administration is low; the evaluation of the few available studies of toxicity after inhalation is difficult due to the limited reporting in many studies and the use of special preparations.

Irritation, corrosivity and sensitisation

Some zinc salts are irritating and corrosive to skin and can induce serious eye damage. Inhalable particules of zinc and its salts may be highly toxic to the lung and a classification as respiratory irritant is considered as appropriate. Zinc salts are not considered as skin sensitisers and no data to characterise a potential for respiratory sensitisation are available.

Toxicokinetics

After oral administration of zinc and its salts, gastrointestinal absorption is usually poor and a maximum value of 20 % of an oral dose is used in the risk assessment. Homeostatic mechanisms adjust the extent of uptake of zinc salts from the GI-tract based on zinc demand. Dermal absorption is considered to be small; adequate data on absorption after inhalation exposure are not available. Absorbed zinc is distributed to all tissues and is eliminated mainly in faeces.

Repeated dose toxicity

Only data after oral exposure of both animals and humans to zinc salts are available for evaluation. Based on a human study, a NOAEL of 50 mg Zn^{2+}/day in humans (as total intake) is derived. In this study, a LOAEL of 150 mg Zn2+/day resulted in clinical signs (headache, nausea, gastric discomfort) and effects on copper homeostasis. The CSTEE accepts the use of this NOAEL for the risk assessment.

Genotoxicity

The mutagenicity and genotoxicity of zinc and its salts were studied in several *in vivo* and *in vitro* assays.

It is assumed that under physiological conditions all zinc compounds under review here (including zinc metal) release Zn^{2+} as the biologically active agent. The compounds were therefore considered as group for the evaluation of this endpoint.

In vitro studies

Ames tests were conducted on zinc oxide, zinc sulphate, zinc chloride, zinc distearate and zinc monoglycerolate. All tests yielded negative results with and without metabolic activation.

An increase in mutant frequencies was found at the thymidine kinase locus in L5178Y mouse lymphoma cells in tests with zinc oxide and zinc monoglycerolate, but not with zinc chloride.

Chromosomal effects were found following exposure of human lymphocytes to zinc oxide and zinc monoglycerolate, but not in a study with zinc chloride. Zinc sulphate did not induce aberrations in human embryonic lung cells.

Zinc oxide induced unscheduled DNA synthesis in SHE cells.

Further to the studies presented in the RAR, there is information available on the photomutagenic activity of zinc oxide (SCCNFP, 2003), which should be discussed in the RAR. In the studies cited by the SCCNFP it was shown that micronised zinc oxide was not photomutagenic in a gene mutation test with *Salmonella typhimurium* and *Escherichia coli*. However, the chemical showed clastogenic and possibly aneugenic activity in mammalian cells in the absence and in the presence of UV light. Zinc oxide showed also activity in a photo-comet assay.

In vivo studies

In mice, zinc chloride (0.5% in a Ca-deficient diet) caused a slight increase in chromosomal aberrations. The regime was lethal for half of the animals, and no effects were found when the animals were given a standard diet. Increased aberrations have also been reported in rats after inhalation exposure to zinc oxide at 0.5–1.0 mg/m^3 for 5 months, and in mice after single and multiple intraperitoneal injections of zinc chloride (at 2–5 mg/kg bw).

No chromosomal aberrations were induced in rats after gavage of zinc sulphate (up to 275 mg/kg body weight (bw) as single dose or for 5 consecutive days), and in a micronucleus test in mice after intraperitoneal injection of up to 86.3 mg zinc sulphate/kg bw.

A host-mediated assay with zinc sulphate was weakly positive, whilst no activity was found for zinc sulphate in a dominant lethal test on rats receiving up to 275 mg/kg bw by single gavage or for 5 consecutive days.

Further to the studies reported in the RAR there is additional information available on the induction of sister chromatid exchanges in rat bone marrow after oral exposure to zinc chloride (Kowalska-Wochna *et al*., 1988). This study should be discussed in the RAR.

Zinc sulphate and zinc chloride, administrated in the diet to *Drosophila melanogaster* did not increase the incidence of sex-linked recessive lethal (SLRL) mutations. The RAR should, however, also discuss positive findings with zinc chloride in *Drosophila* SLRL and dominant lethal tests (Carpenter and Ray, 1969).

Conclusion: *In vitro* tests indicated that zinc has a genotoxic potential. The *in vivo* studies as presented in the RAR are inconclusive with sometimes contradictory results. However, there are indications of some weak clastogenic, and possibly aneugenic effects following zinc exposure. The relevance of these findings needs to be clarified.

Carcinogenicity

Human Studies

There are no adequate epidemiological data available. In a cohort study in refinery workers that did not differentiate between exposures to copper and zinc, no evidence of a relationship between exposure and cancer was found.

A recently published study by the U.S. National Cancer Institute investigated the association between supplemental zinc intake and prostate cancer risk among 46,974 U.S. men (Leitzmann *et al*., 2003). It was found that supplemental zinc intake at doses of up to 100 mg/day was not associated with prostate cancer risk. However, compared with non-users, men who consumed more than 100 mg/day of supplemental zinc had a relative risk of advanced prostate cancer of 2.29 (95% C.I. 1.06 to 4.95, p(trend) = 0.003), and men who took supplemental zinc for 10 or more years had a relative risk of 2.37 (95% C.I. 1.42 to 3.95, p(trend) < 0.003). The authors could not rule out residual confounding by supplemental calcium intake or some unmeasured correlate of

zinc supplement use, and concluded that chronic zinc oversupply may play a role in prostate carcinogenesis. The results of this study should be discussed in the RAR.

As well as oversupply of zinc, zinc deficiency may have an influence on carcinogenesis. In a study in Chinese men the zinc levels in serum and hair were lower in those patients with oesophageal cancer (Lin *et al*., 1977). Similarly, in another study by Lipman *et al.* (1987), mean plasma zinc levels in 21 oesophageal cancer patients were significantly lower than in the 17 patients with oesophagitis, or the 12 normal controls. However, there were no differences in oesophageal zinc content between the cancerous and adjacent normal tissue, the oesophagitis and adjacent normal tissue, and normal oesophageal tissue. These studies should be discussed in the RAR.

Animal Studies

A carcinogenicity study performed according to current guidelines on zinc or its salts is not available. In rats and chickens, testicular tumours have been produced by direct injection of zinc chloride or zinc sulphate. Because of the non physiological route of administration, the relevance of these findings is, however, unclear. No increase in tumour frequencies was found in an old and poorly documented study on mice after administration of zinc sulphate in the drinking water for 45-53 weeks.

In vitro studies

Cell transformation tests were positive with zinc oxide, but negative with zinc chloride. Zinc chloride and zinc sulphate gave equivocal results in an *in vitro* test for the capacity of these metal salts to enhance viral transformation of Syrian hamster embryo cells, producing enhancement in 3/6 and 3/7 trials respectively (Casto *et al*., 1979).

Based on the limited data and the absence of experimental or epidemiological evidence for carcinogenicity of zinc and its salts, the RAR concludes that there is no concern regarding carcinogenicity of zinc and zinc salts in humans.

Reproductive and developmental toxicity

A number of studies on effects of zinc and some zinc salts after oral administration are available. However, the information from the studies to be used for risk characterisation is limited. Based on the available data, the RAR derives a NOAEL for developmental toxicity of approx. 20 mg Zn^{2+}/kg/day. It should be noted that zinc deficiency in humans is known to impair fertility and foetal development. Based on the absence of effects of zinc and zinc salts given during pregnancy, the RAR concludes that there is no concern for reproductive toxicity of zinc compounds in humans.

The CSTEE agrees with this assessment.

3. Risk characterisation

Acute and repeated-dose toxicity

Regarding worker exposure, the MOS values for inhalation toxicity for zinc and zinc salts are often < 10. However, the RAR most often comes to conclusion ii) and justifies this conclusion by the highly conservative exposure assessment and the use of a human NOAEL. While the CSTEE agrees with this justification in most cases, conclusion ii) in the case of metallic zinc exposure with a MOS of 0.9 should be well justified since the estimated internal dose will result in a major contribution to total zinc intake from other sources. Regarding respiratory irritation in the scenario "maintenance of flux baths" in the zinc chloride RAR, the conclusions are unclear regarding the different scenarios.

Regarding consumer exposure, the RAR concludes that cumulative internal exposure to approx. 1.6 mg Zn^{2+}/day from a variety of sources with a MOS of 6.25 warrants conclusion ii). Considering the daily zinc intake from food and the homeostatic mechanisms regulating zinc absorption from the diet, the CSTEE agrees with this conclusion.

Genotoxicity and Carcinogenicity

Based on the available data, it is concluded that zinc and zinc salts are not expected to be mutagens and carcinogens in humans under the expected exposure conditions. *In vitro* tests indicated that zinc has a genotoxic potential. The *in vivo* studies as presented in the RAR are inconclusive with sometimes contradictory results. However, there are indications of some weak clastogenic, and possibly aneugenic, effects following zinc exposure. The relevance of these findings needs to be clarified.

With regard to genotoxicity, the CSTEE therefore recommends a conclusion (i). Conclusion (ii) for workers will have to be re-evaluated following the elaboration on the genotoxicity endpoint.

There is no experimental evidence from a limited animal study that orally administered zinc sulphate is tumorigenic. Deficiency and supplements of zinc can however have an influence on carcinogenesis, possibly as a result of the influence of zinc on cell growth.

Results from an extensive study in the U.S. indicate that chronic zinc oversupply from dietary supplements may play a role in prostate cancer. As there is only very limited information presented in the RAR with regard to zinc exposure from the use of dietary supplements in the EU, the CSTEE recommends to perform an exposure analysis and, if then indicated, to perform a risk assessment for this exposure scenario.

Therefore, the CSTEE supports conclusion (i) for consumers.

Reproductive and developmental toxicity

Conclusion ii) is reached regarding effects on development and fertility for workers and consumers. MOS are not given regarding this endpoint and a better justification should be included in the RARs for all zinc compounds.

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