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Formaldehyde Toxicology: An Up-Dating of the ECETOC Technical Reports 1 and 2

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FORMALDEHYDE TOXICOLOGY : AN UP-DATING OF THE ECETOC TECHNICAL REPORTS 1 AND 2

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In 1981 ECETOC published its Technical Reports 1 and 2, "An Assessment of Data on the Effects of Formaldehyde on Humans" and "The Mutagenic and Carcinogenic Potential of Formaldehyde", respectively, in which an assessment of the available information on formaldehyde toxicity and its significance for the human situation was made. It was recommended that the situation be reviewed once the final report of the CIIT/Battelle chronic inhalation study and the results of new toxicological and epidemiological investigations became available.

The final CIIT/Battelle report was released on 18th February 1982. Further information on the distribution, metabolism and mutagenicity of formaldehyde, on the mechanism of formaldehyde-induced carcinogenicity in experimental animals, and the results of two further epidemiological studies have now been published, and permit a review and up-dating of the two reports.

In this document the new information is assessed in relation to the conclusions and recommendations in the previous Technical Reports.

1. Toxicity Data

In the CIIT/Battelle chronic inhalation study (Battelle, 1981) rats and mice were exposed to formaldehyde vapour at the following concentrations : 2.0 $\pm$ 0.6 ppm (2 ppm), 5.6 $\pm$ 1.2 ppm (6 ppm) and 14.3 $\pm$ 2.8 ppm (15 ppm). The nominal concentrations are shown in brackets. Exposure periods were 6 hours per day, 5 days per week for up to 2 years. This exposure resulted in statistically-significant increases in the incidence of a number of pathological lesions, summarized in Table 1 (CIIT, 1982).

The most important finding was an exposure-related induction of nasal tumours, mainly squamous cell carcinomas, in 106 of the 239 rats exposed to 14.3 ppm, and 2 of the 240 rats exposed to 5.6 ppm of formaldehyde vapour. Nasal tumours were also observed in 2 of 240 mice exposed to 14.3 ppm of formaldehyde vapour. In this latter case it is difficult to establish the total number of animals exposed since fighting amongst group-caged males resulted in significant mortality during the first six months of the study. The tumour incidences in rats exposed to 5.6 ppm and mice exposed to 14.3 ppm formaldehyde vapour were not statistically significant, but cannot be ignored since spontaneous occurrence of such tumours is very rare (Battelle, 1981).

The pathological findings illustrate extensive damage caused by formaldehyde irritation of the nasal cavities. At 27 months, a regression of metaplasia in the nasal cavity was observed for rats and mice in the 2 ppm and 5.6 ppm

TABLE 1

LESIONS FOUND IN FORMALDEHYDE-EXPOSED ANIMALS (CIIT,1982).

SPECIES	SITE OF LESION	LESION	EXPOSURE CONCENTRATION
MICE			
Male	Nasal Cavity	Epithelial Dysplasia	6 ppm, 15 ppm
		Rhinitis	15 ppm
		Squamous Metaplasia	6 ppm, 15 ppm
		Olfactory Epithelial Atrophy	15 ppm
	Nasolacrimal Duct	Epithelial Hyperplasia	15 ppm
Female	Nasal Cavity	Epithelial Dysplasia	6 ppm, 15 ppm
		Rhinitis	15 ppm
		Squamous Metaplasia	15 ppm
		Olfactory Epithelial Atrophy	15 ppm
	Nasolacrimal Duct	Epithelial Hyperplasia	15 ppm
RATS			
Male	Nasal Cavity	Epithelial Dysplasia	2 ppm, 6 ppm, 15 ppm
		Squamous Metaplasia	2 ppm, 6 ppm, 15 ppm
		Rhinitis	2 ppm, 6 ppm, 15 ppm
		Goblet Cell Hyperplasia	2 ppm, 6 ppm, 15 ppm
		Olfactory Epithelial Atrophy	15 ppm
		Hyperkeratosis	15 ppm
		Squamous Atypia	15 ppm
		Respiratory Epithelial Hyperplasia	15 ppm
		Goblet Cell Metaplasia of Olfactory Epithelium	15 ppm
		Squamous Epithelial Hyperplasia	15 ppm
		Squamous Cell Carcinoma	15 ppm
	Trachea	Epithelial Dysplasia	15 ppm
	Bone Marrow	Hyperplasia	15 ppm
Female	Nasal Cavity	Epithelial Dysplasia	2 ppm, 6 ppm, 15 ppm
		Squamous Metaplasia	2 ppm, 6 ppm, 15 ppm
		Rhinitis	2 ppm, 6 ppm, 15 ppm
		Goblet Cell Hyperplasia	2 ppm, 15 ppm
		Olfactory Epithelial Atrophy	15 ppm
		Hyperkeratosis	15 ppm
		Squamous Atypia	15 ppm
		Goblet Cell Metaplasia of Olfactory Epithelium	15 ppm
		Squamous Cell Carcinoma	15 ppm
		Squamous Hyperplasia	15 ppm
	Trachea	Epithelial Dysplasia	15 ppm
		Squamous Metaplasia	15 ppm
	Bone Marrow	Hyperplasia	15 ppm

exposure groups. The presence of lesions described as epithelial dysplasia and squamous metaplasia, observed in the trachea of rats exposed to 14.3 ppm of formaldehyde show that this irritation was not confined to the nasal cavities but occurred to a certain extent further into the respiratory tract. The bone-marrow hyperplasia which was also observed in these animals could be a consequence of the extensive inflammation induced by formaldehyde in the respiratory tract, but was not reflected by significant changes in the measured haematological parameters.

The induction of nasal tumours in rats by exposure to formaldehyde vapour has been confirmed by a second study (Albert et al.,1982) in which a similar protocol was used (exposure to 14 ppm formaldehyde vapour 6 hr/d, 5d/wk) but with a different rat strain. At the time of publication, this study had been in progress for 588 days, and 10% of the exposed rats had developed squamous cell carcinomas in the nasal cavities. There was also a 9% reduction in body weight and an elevated mortality.

Several papers concerning the mechanism of formaldehyde-induced carcinogenicity have been published. Ragan and Boreiko (1981) have shown that formaldehyde may initiate the transformation of C3H/10TI/2 cells in culture, but transformed foci grew only after treatment with the tumour promoter, tetradecanoyl phorbol acetate. This shows that in this system formaldehyde may initiate the process of transformation, but does not function as a tumour promoter. However, in the rat nasal cavity the continuous cycles of cell death and subsequent replacement induced by the formaldehyde exposure may provide the promotional stimulus.

Protein-DNA cross-links in Yoshida sarcoma cells (Bedford and Fox, 1981) and in mouse leukaemia L1210 cells (Ross et al.,1981) were induced by treatment with formaldehyde. Some DNA strand breakage was reported, but DNA-DNA cross-links were not observed. These findings once again demonstrate the ability of formaldehyde to react with DNA of cells in culture. However, such an interaction would require a treatment-related increase in intracellular formaldehyde concentrations. Formaldehyde at concentrations of 0.1 and 0.7% was shown to induce point mutations and deficiencies in the unc-22 region of the genome of a nematode (Moerman and Baillie, 1981). Heck (1982) has developed a method for the determination of total amounts of formaldehyde (free and reversibly bound) in tissue samples, using stable isotope dilution and gas chromatograph-mass spectrometric quantification. Measurements show that the nasal epithelium of non-exposed rats normally contains significant

amounts of formaldehyde. Most of the formaldehyde was in the form of glutathione-hemithioacetal or bound to tetrahydrofolate, the free formaldehyde concentration being calculated as between 1 and 2% of the total. The level of this total cellular formaldehyde did not increase following exposure of rats to 6 or 15 ppm formaldehyde vapour for 6 hours. These findings show that at low rates of "deposition" the increase of the tissue concentration may be negligible in comparison to the concentration of endogenous formaldehyde. However, from the available data, the occurrence of transient or local increases in formaldehyde concentration, especially in free form, cannot be ruled out.

Distribution studies (Heck, 1982) with rats showed that after inhalation of radioactive formaldehyde the concentration of radioactivity in the nasal mucosa was 1 to 2 orders of magnitude higher than in plasma or other organs. The concentration of radioactivity in the nasal cavity remained elevated compared to other organs, even after three days exposure. Thus, toxic effects are to be expected only in those tissues directly exposed to formaldehyde. These distribution studies also showed that 40% of the radioactivity was retained in the body, 40% was exhaled and most of the remainder was excreted in the urine.

Injection of C^{14}-formate and C^{14}-formaldehyde showed identical pharmacokinetics in erythrocyte and plasma (Heck, 1982). Evidence was obtained that radioactivity from formaldehyde is retained in the body by metabolic incorporation into macromolecules via the 1-carbon pool, and not by covalent binding, since formate cannot form adducts with nucleophilic macromolecules.

The deposition of C^{14}-formaldehyde in the nasal mucosa was studied with 2 and 15 ppm in rats for various exposure times up to 6 hrs, and for an exposure period of 6 hrs at different concentrations up to 24 ppm. The results suggested that "a prolonged exposure to a low concentration of formaldehyde probably entails less risk than short exposure to a high concentration". Formaldehyde- and aldehyde-dehydrogenase can metabolise formaldehyde and counteract its toxic effect. Heck (1982) demonstrated that the activity of these enzymes is very similar in nasal mucosa and the liver. Moreover, after repeated exposure to 15 ppm of formaldehyde no change was detected in the specific activity of formaldehyde dehydrogenase and only a slight decrease was observed in the activity of aldehyde dehydrogenase. Sufficient glutathione, a coenzyme of formaldehyde dehydrogenase, is available in the nasal mucosa to bind most of the endogenous formaldehyde as the hemithioacetal.

Repeated exposure to 15 ppm did not lead to a significant change in the glutathione content. Thus, "the principal defence mechanisms with respect to formaldehyde in the nasal mucosa are not altered in their efficiency by repeated exposure to this chemical"(Heck, 1982).

Chang et al.(1981) have reported studies on the effect of inhaled formaldehyde vapour on respiratory minute volumes of mice and rats. The results, which are essentially as reported in ECETOC Technical Report no.2, show that both rats and mice responded to formaldehyde inhalation by reducing their respiratory rates and minute volumes. However, mice responded to lower formaldehyde concentrations than did rats. For example, respiratory rates were reduced by 50% at 4.9 ppm for mice and 31.7 ppm for rats. Rats also developed some tolerance to the formaldehyde during exposure. Both rats and mice pretreated with 15 ppm formaldehyde were slightly more sensitive to respiratory-rate depression, but pretreated rats compensated the decrease in respiratory rate by increasing the tidal volumes. Thus, following pretreatment with 15 ppm formaldehyde the difference in sensitivity between the two species became more marked. As a result, mice were able to minimize the inhalation of formaldehyde more efficiently than rats. This species difference may contribute to the differences in respiratory tract toxicity from inhaled formaldehyde.

Species differences in the response to formaldehyde inhalation were observed in a 6-month inhalation study performed by Bio/dynamics (1982). Monkeys, hamsters and rats were exposed to 3, 1 or 0.2 ppm of formaldehyde vapour 22 hours a day, 7 days a week. Squamous metaplasia was detected in the nasal cavities of rats (23 out of 37) and monkeys (6 out of 6) in the 3 ppm exposure group. Additionally, at 3 ppm body weight gain and absolute and relative liver weights were reduced in rats, and the nasal secretions increased in monkeys. No such changes were detected in the animals exposed to 1 or 0.2 ppm, or in hamsters exposed to 3 ppm, of formaldehyde vapour.

2. Epidemiological Data

2.1. A case-control study of cancer mortality among Du Pont workers exposed to formaldehyde has been completed (Fayerweather et al.,1982). Cancer deaths from 1957 through 1979 were studied at eight formaldehyde manufacturing or using plants. There were 481 cancer deaths at these sites, 142 of which were among workers with potential exposure to formaldehyde.

The data were analysed by tumour site, latency period, duration of exposure, exposure level and frequency, cumulative exposure index, age and year of death, and age and year of first exposure. Analyses of lung cancer deaths were adjusted for the subjects'cigarette smoking habits. In none of these analyses was the formaldehyde workers'relative risk of cancer significantly greater than 1.0 ($p>0.05$). Of particular significance was the absence of nasal cancer deaths and excess lung cancer among workers exposed to formaldehyde. These results were considerably strengthened by the ability to factor the workers'smoking habits into the analyses. Slight, but statistically-insignificant, elevations in relative risks were observed for prostatic and bladder cancers, but there were no indications of a causal relationship to formaldehyde and they appeared to be due to chance.

In short, a thorough analysis of the data suggested that cancer mortality rates in the Company's formaldehyde-exposed workers were no higher than the rates among non-exposed coworkers.

2.2. Goldmann et al.(1982) performed a morbidity study at BASF in the German Federal Republic. One-hundred and ninety-nine employees exposed to formaldehyde for up to 42 years (83 more than 20 years; average 18 years) were subjected to an extensive morbidity study. The average age of the cohort was 44 years, with 89 persons older than 45 years. The formaldehyde concentration in the workroom air, as an average per shift, had not been above 1 ppm since 1971, and in the years before had not been above 5 ppm, in accordance with the standards for the maximum workplace concentration (MAK value) valid during the respective periods of time in the Federal Republic of Germany. The study covered : a thorough history (employing the questionnaire used for epidemiological studies of chronic bronchitis and pulmonary emphysema), clinical examination, X-ray examination of lung and paranasal sinuses, lung function tests employing the bodyplethysmograph, and extensive laboratory parameters. A cohort of 91 steel construction workers served as the control group for the X-ray examination of the lungs and the lung-function tests.

The overall state of health of the formaldehyde-exposed cohort was good. No malignant tumours were discovered in the nasal cavities, the paranasal sinuses or the lungs. The results concerning lung function and X-ray examination of the lung showed fewer deviations from the normal findings in the cohort exposed to formaldehyde than in the control cohort. The lung

function was normal in 85 percent of the exposed group compared with 78 percent in the control group. The X-ray examination of the lungs resulted in 92.5 percent normal findings in the exposed cohort as compared to 83.5 percent in the control cohort. There was no indication of any correlation between lesions of the lungs or irritations of the paranasal sinuses, and formaldehyde exposure. The observed irritation of the paranasal sinuses was comparable to that in the general population. The results of the laboratory tests did not indicate any influence of formaldehyde exposure.

This study shows that occupational exposure to formaldehyde, even over the long period studied, did not result in any health problems of the employees under the given workplace conditions of the cohort.

3. Conclusions

The recent information discussed in this report does not alter the final conclusions presented in the ECETOC Technical Reports 1 and 2 in which it was claimed that the nasal cancers observed in experimental animals develop only at concentrations which produce chronic tissue irritation. Where exposure is so low that metaplasia resulting from irritation does not occur, it is unlikely that tumours will develop.

The new epidemiological data confirm that there is no relationship between formaldehyde exposure and cancer in humans.

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