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Potential Carcinogenicity of Chloral Hydrate — A Review

Andrew G. Salmon, M.A., D. Phil.*; Kenneth W. Kizer, M.D., M.P.H.**; Lauren Zeise, Ph.D.*; Richard J. Jackson, M.D., M.P.H.***; Martyn T. Smith, Ph.D.****

California Environmental Protection Agency, Berkeley*; Department of Internal Medicine, School of Medicine, University of California, Davis**; California Department of Health Services, Berkeley***; School of Public Health, University of California, Berkeley****, California

ABSTRACT

Chloral hydrate is commonly used to sedate children for diagnostic or therapeutic procedures. The drug has been extensively used for many years, but there are remarkably few data on its long-term health effects. Concern in this regard is raised by recent studies showing chloral hydrate to be genotoxic, causing chromosome changes and other effects in vivo and in vitro. In addition, chloral hydrate is a reactive metabolite of trichloroethylene, a known carcinogen, and is structurally similar to other carcinogenic intermediates. Two carcinogenicity studies performed using the oral route of administration in mice indicate that the drug is potentially carcinogenic — in one case after a single dose lower than the typical dose used for sedation. Practitioners should be aware of chloral hydrate's genotoxicity and potential carcinogenicity. Discretion in its use seems appropriate until further studies clarify its long term health consequences. (Key Words: chloral hydrate, toxicity, adverse effects; child; sedatives.)

Correspondence: Dr. Kenneth Kizer, Department of Community and International Health, School of Medicine, TB-168, University of California, Davis, CA 95616.

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INTRODUCTION

Chloral hydrate has been used as a sedative-hypnotic since at least 1869 (1). Today, it is used extensively in pediatrics, emergency medicine, radiology, dentistry, and other clinical settings for short term sedation of infants and children to allow completion of diagnostic and therapeutic procedures (1-4). Dosages in the range 25-100 mg/kg body weight are generally recommended for pediatric use. Various alternative drugs have been advocated for pediatric sedation, including thiopental, ketamine, meperidine, fentanyl, midazolam, and nitrous oxide, among others: however, each of these agents, alone or when combined with other compounds, has disadvantages. Use of chloral hydrate has been advocated based on its demonstrated efficacy and its very low incidence of acute toxicity (43).

Chloral hydrate is the hydrated form of an aldehyde, chloral (1,1,1-trichloroacetaldehyde). This aldehyde reacts with water to form a 1,1-diol, which in this case is the major species present at equilibrium. Chloral hydrate is metabolized by reduction to trichloroethanol and oxidation to trichloroacetic acid (6,7). Its metabolism occurs mainly in the liver and kidneys. These reactions are rapid, but may be limited both by saturation of individual enzymes and by limits on the availability of reductant (NADH/NADPH) or oxidant (NAD+/NADP+) (8). The ratio of reduced to oxidized product excreted appears to vary with animal species and metabolic status. Because the enzymes and cofactors responsible for these reactions are also involved in ethanol metabolism, chloral hydrate and ethanol enhance the sedative effects of one another. This leads to the potency of the so-called "Mickey Finn" cocktail (9).

Chloral hydrate is a metabolite of trichloroethylene (TCE), as shown in Figure 1 (10-12). Oxidation of TCE, like that of many compounds containing an ethylenic double bond, is catalyzed by cytochrome P-450 enzymes and generates an epoxide. In the case of TCE, this epoxide is unstable and spontaneously rearranges to chloral, with migration of one chlorine atom. Chloral hydrate may be one of the chemical species responsible for the genotoxic and carcinogenic effects of TCE, although the mechanism by which these effects are produced has not been clearly delineated. Smith recently compared the potential risks posed by trichloroethylene and chloral hydrate, and concluded that the hazards posed by the use of chloral hydrate in children were significant (13). This prompted us to review the subject.

GENOTOXICITY OF CHLORAL HYDRATE

Chloral hydrate is well known as an agent inducing aneuploidy in various test systems, including eukaryotic microbial organisms, mammalian cells in culture and mammalian germ cells in vivo (13-18). Chloral hydrate was selected as a test compound in a recent evaluation of assays for aneuploidy-generating agents (19). It was positive in all the systems using mammalian cells in vitro. Positive results in the Salmonella point mutation assay and introduction of DNA strand breaks have been reported (21-24). Sister chromatid exchanges have been reported in human lymphocytes exposed in vitro to chloral hydrate (25). A recent preliminary report described an individual who having taken repeated doses of chloral hydrate totaling approximately 35 g over five months, showed chromosomal aberrations and elevated incidences of sister chromated exchange and micronuclei in peripheral blood lymphocytes (26). These findings currently relate to a single case, and there are various possible confounding effects. However, this observation underscores the need for further investigation of the genotoxicity of chloral hydrate in humans.

Other carcinogenic chloroalkenes, including vinyl chloride, have chlorinated epoxides and aldehydes as active metabolic intermediates. These intermediates are known to be genotoxic and are thought to be directly involved in carcinogenesis (27). Chloral hydrate was one of only two of 46 chemicals suspected of causing chromosomal effects for which unequivocal evidence of aneuploidy induction in mammalian germ cells was found (28). The other compound was cyclophosphamide. In the aggregate, these data provide substantive evidence of the genotoxicity of chloral hydrate.
CARCINOGENIC EFFECTS OF CHLORAL HYDRATE

In view of the extensive evidence of chloral hydrate's genotoxicity and its ongoing widespread clinical use, the limited information about its carcinogenicity is of concern. As far as we can determine, no epidemiological studies of chloral hydrate's ability to induce cancer in humans have been conducted. This is of particular concern in view of recent animal data.

In one animal carcinogenicity study in which infant male mice received a single oral dose of chloral hydrate, an increase in liver tumors was observed (29). This was statistically significant in spite of severe limitations on the power of the study due to small group sizes and the single dose design. In contrast, a 1955 study of chloral hydrate tumor initiation in mouse skin was inconclusive (30). Until recently, these two studies were the only available data on the carcinogenicity of chloral hydrate. The absence of any reported studies with negative results is noteworthy.

Recently, results of a two-year study of tumor incidence in male B6C3F1 mice receiving 0.1 g/L chloral hydrate in drinking water have been reported (31). The findings are consistent with the earlier published findings of mouse liver carcinogenicity; that is, chloral hydrate was found to cause hepatocellular carcinomas, adenomas and hyperplastic nodules.

To facilitate comparison of the data, the three available studies of chloral hydrate carcinogenicity are summarized below. A standard statistical analysis, using Fisher's exact test relative to controls with appropriate definition of the number of animals at risk, was applied to all three studies. This analysis differs in detail from what was reported by the original study authors, but it does not result in notably different conclusions from those originally reported.

Single Oral Exposure (29)

Two groups of 15-day-old C57BLxC3H F1 male mice were given a single dose of chloral hydrate by gavage. One group of 15 mice received 5 mg/kg chloral hydrate, while the other group of 14 mice received 10 mg/kg. All doses were dissolved in distilled water and a control group of 26 mice received distilled water only. Mice were killed when moribund or at intervals up to 92 wk after treatment. Seven control and eleven exposed mice were killed within 48 wk after the dose was given. For the purposes of evaluating carcinogenicity, only mice killed between 48 wk (the time of first appearance of a hepatic tumor which was in a mouse exposed to 10 mg/kg chloral hydrate) and 92 wk after treatment were considered to be at risk. Hepatic nodules were examined and classified according to the scheme of Vesselinovitch et al. as either hyperplastic foci, hepatocellular adenomas or trabecular carcinomas (32). In the group receiving 10 mg/kg, the combined incidence of adenomas and carcinomas was significantly greater than in the controls (p = 0.002, Fischer Exact Test). The trend was significant across all groups (Mantel-Haenszel trend
test, p = 0.0007). The finding of a statistically significant increase in hepatic tumors persisted (p = 0.01) when animals killed at 48 wk were excluded from analysis. Hepatic tumors were observed earlier in exposed animals, occurring between 48 and 88 wk after dosing, whereas the two tumors in control animals both occurred at wk 89. Group sizes and incidences of tumors and foci are given in Table 1.

Additionally, six mice receiving 10 mg/kg chloral hydrate, ten receiving 5 mg/kg, and nine control mice were killed 24 h after dosing and analyzed for liver mitotic index. An increase in the mean mitotic index of 2 to 3-fold was observed at both dose levels, although only the effect in the group receiving 5 mg/kg was statistically significant.

Chloral Hydrate in Drinking Water (31)

A group of 40 male C57BL × C3H F1 mice received 1 g/L chloral hydrate in drinking water for 104 wk. Two similar control groups, totaling 33 animals, received plain water, while other groups received 2-chloroacetaldehyde or dichloroacetic acid in the drinking water. Interim sacrifices at 30 to 60 wk (5 control and 5 chloral hydrate exposed mice at each time) were made for biochemical and interim pathological analysis. Animals dying during the study (3 controls, 6 chloral hydrate) were not counted as being at risk for tumor induction. Causes of these intercurrent deaths were not determined. Other than neoplasia, only mild histopathological changes were observed in the liver, and no changes were noted in other organs. Two chloral hydrate exposed mice out of five examined at the 60 wk interim sacrifice were found to have hepatocellular carcinomas. No such lesions were observed in controls at this time. At 104 wk 11 exposed mice had hepatocellular carcinomas, and 7 had hepatocellular adenomas, out of 24 survivors. Hepatocellular lesions in controls included 2 animals with carcinomas and one with an adenoma out of 20 survivors examined (Table 2). The incidences of adenomas (p = 0.043) and carcinomas (p = 0.010) were significantly greater than in controls. The increase in the combined incidence of the two lesion grades was highly significant (p = 0.00024).

Repeated Dermal Exposure (30)

Twenty male albino "S" mice received two dermal applications of 0.3 mL of a 4% solution of chloral hydrate one week apart (total dose 24 mg), and also 18 weekly applications of 0.5% croton oil in acetone, starting 4 wk after the first chloral hydrate treatment. Of these 20 mice, 17 were still alive at 23 wk. Four mice were observed to have skin tumors at the end of the croton oil treatment.

Another group of 20 male "S" mice received 0.3 mL of a 5% solution of chloral hydrate applied to the skin weekly for 15 wk (total dose 225 mg). Beginning 3 d after the first chloral hydrate application, 18 weekly applications of 0.5% croton oil in acetone were given. All 20 exposed animals were still alive at 18 wk. Four mice were observed to have skin tumors at the end of the croton oil treatment.

A control group of male "S" mice received 18 applications of croton oil only. There were 20 surviving controls, of which one bore skin tumors. Although the incidence of skin tumors was elevated in both exposed groups relative to that in the control group, neither of the differences were statistically significantly (p = 0.17). In view of the different site, limited power and inconclusive result of this study, it cannot be regarded as either confirming or contradicting the mouse liver results.

DISCUSSION AND CONCLUSION

It is clear from this review of the genotoxicity and potential carcinogenicity of chloral hydrate that the potential for this compound to have long-term health effects has been inadequately examined, both in terms of animal and human data. This is of concern in view of the widespread use of this compound.

The choice of safe and effective sedative-hypnotic drugs for use in children is limited, and chloral hydrate remains popular because of clinician familiarity with the compound, its demonstrated efficacy, and its low rate of acute toxicity, as well as the potential drawbacks of alternative compounds. Indeed, it appears as if chloral hydrate's long history of use has given clinicians a sense of security with the drug even though such security now seems questionable in view of its genotoxicity, its chemical similarity to known carcinogens, and its observed carcinogenicity in animals. The fact that a large increase in tumor incidence occurred after a single oral dose that was smaller than the dose commonly
Chloral Hydrate Carcinogenicity

Table 1

Tumors in C57BLxC3H F1 Male Mice Receiving a Single Oral Dose of 5 - 10 mg/kg-BW Chloral Hydrate

<table>
<thead>
<tr>
<th>Tumor Site &amp; Type</th>
<th>Dose, mg/kg*</th>
<th>0</th>
<th>5</th>
<th>10</th>
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</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Liver</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Adenomas</td>
<td>0/19†</td>
<td>1/9</td>
<td>3/8</td>
<td></td>
</tr>
<tr>
<td>Carcinomas</td>
<td>2/19</td>
<td>1/9</td>
<td>3/8</td>
<td></td>
</tr>
<tr>
<td>Adenomas or</td>
<td>2/19</td>
<td>2/9</td>
<td>6/8</td>
<td></td>
</tr>
<tr>
<td>Carcinomas</td>
<td>p = 0.0007†, p = 0.38†, p = 0.002§</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Given in distilled water at 15 d of age.
†Number of tumor bearing animals/number of animals in the group.
‡p values for trend (Mantel-Haenszel trend test).
§p values for Fisher Exact Test relative to control group. Value is given in parentheses when not significant (p > 0.01).

Table 2

Tumors in C57BLxC3H F1 Male Mice Receiving 1 g/L Chloral Hydrate in Drinking Water for 104 Wk

<table>
<thead>
<tr>
<th>Tumor Site &amp; Type</th>
<th>Dose, mg/kg/d*</th>
<th>0</th>
<th>166</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Liver</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Adenomas</td>
<td>1/20†, p = 0.043‡</td>
<td>7/24</td>
<td></td>
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<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Carcinomas</td>
<td>2/20, p = 0.010</td>
<td>11/24</td>
<td></td>
</tr>
<tr>
<td>Adenomas or</td>
<td>3/20, p = 0.00024</td>
<td>17/24</td>
<td></td>
</tr>
<tr>
<td>Carcinomas</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hyperplastic Nodules</td>
<td></td>
<td>0/20, n.s.</td>
<td></td>
</tr>
</tbody>
</table>

*Average daily intake, based on drinking water consumption.
†Number of lesion-bearing animals/total examined at 104 wk.
‡p value calculated by Fisher’s exact test, relative to control group.

used for sedation in children is disconcerting. And while the animal studies currently available do not provide sufficient information to accurately quantify the carcinogenic risk of chloral hydrate, this risk cannot be dismissed as being negligible at this time.

Clinicians must weigh the benefits of sedation of a child for important diagnostic procedures such as angiograms and electroencephalography and for therapeutic procedures such as suturing against any potential short or long-term untoward effects of the sedating agent. In light of present data about the genotoxicity and potential carcinogenicity of chloral hydrate and the absence of any studies on the potential chronic toxicity of the compound, the use of chloral hydrate for chronic sedation of infants certainly appears unjustified at this time. In contrast, it is less clear whether there is a significant risk associated with single doses for short-term sedation.

Of note, determining the carcinogenicity of chloral hydrate in humans will be extremely difficult because of record-keeping limitations, the long latency period of cancer, and the myriad of other carcinogen exposures that occur during childhood and adolescence. In addition, there are differences between men and mice in the kinetics and metabolism of chloral hydrate, and tumor types in humans are not necessarily predicted by the site of animal tumors. Thus, cancers at all sites, rather than just the liver as found in test animals, should be considered. Even with high doses of potent carcinogens, such as those associated with cancer chemotherapy, years of investigation are often required to identify cause and effect relationships.

Until clarifying information becomes available, clinicians should be aware of the potential cancer hazard posed by the use of chloral hydrate, and they should advise patients/parents accordingly. In addition, epidemiological investigation into the chronic effects of chloral hydrate use in children should be initiated, and prospective studies of the genotoxic effects of chloral hydrate on acute and chronically exposed persons should be commenced.

REFERENCES

1. Graham SR, Day RO, Lee R, Fulde, GW. Overdose with chloral hydrate: a pharmacological and


