

RECEIVED
DIRECTOR OF COL. TIGHT
(11/19/11, 1986)

For Lit File
Trichloroethylene
Metabolism

ORAL AND INTRAVENOUS TRICHLOROETHYLENE PHARMACOKINETICS IN THE RAT

RECEIVED

JAN 30 1986

Environmental Affairs

Richard W. D'Souza

Department of Pharmaceutics, University of Houston,
University Park, Houston, Texas

James V. Bruckner

Department of Pharmacology and Toxicology,
University of Georgia, Athens, Georgia

Stuart Feldman

Department of Pharmaceutics, University of Houston,
University Park, Houston, Texas

The pharmacokinetics of trichloroethylene (TCE) was studied in male Sprague-Dawley rats (300-350 g). TCE was administered intravenously and orally at doses of 5, 10, and 25 mg/kg to nonfasted rats and orally at 10 mg/kg to rats fasted for 8-10 h. The disappearance of TCE from the blood of intravenously dosed animals was best described by a two-compartment open pharmacokinetic model. The volume of the central compartment (V_c) approximated the rats' blood volume (50-70 ml/kg). The volume distribution (V_d) and total body clearance (CL_T) decreased with increase in dose. The terminal half-life ($t_{1/2}$) was about 120 min and was not affected by increases in dose. TCE was rapidly absorbed after oral dosing, with blood concentrations peaking between 6 and 10 min. The oral to intravenous bioavailability of TCE was 60-80% in nonfasted animals. The terminal $t_{1/2}$ in fasted, orally dosed rats was identical to that when fasted rats were given the same dose intravenously. In fasted rats, bioavailability of an oral dose was greater than 90%, and peak levels in the blood were 2-3 times as high as in nonfasted rats.

INTRODUCTION

Trichloroethylene (TCE) is a halogenated hydrocarbon used extensively in industry. Its low volatility, nonflammability, and high lipid solubility make it a useful solvent for degreasing metals. It also finds use as a dry cleaning solvent, in organic syntheses, and in refrigerants and fumigants. TCE has been used in the past as an inhalation anesthetic (Trilene, Great Britain; Trethylene, United States) and in the decaffeination of coffee. TCE has been widely used in such products as Carbona cleaning fluid and Carbona No. 10 (containing approximately 40% TCE). These commercial products have been subject to abuse by "solvent sniffers" (Alapin, 1973). Although production of TCE has been declining in recent years in the United States, it has been estimated that 133,000 metric tons of TCE were produced in 1980 (U.S. International Trade Commission, 1980).

Supported in part by U.S. Environmental Protection Agency grant R808282.

ORAL AND INTRAVENOUS TRICHLOROETHYLENE PHARMACOKINETICS IN THE RAT

RECEIVED

JAN 30 1986

Environmental Affairs

Richard W. D'Souza

Department of Pharmaceutics, University of Houston,
University Park, Houston, Texas

James V. Bruckner

Department of Pharmacology and Toxicology,
University of Georgia, Athens, Georgia

Stuart Feldman

Department of Pharmaceutics, University of Houston,
University Park, Houston, Texas

The pharmacokinetics of trichloroethylene (TCE) was studied in male Sprague-Dawley rats (300-350 g). TCE was administered intravenously and orally at doses of 5, 10, and 25 mg/kg to nonfasted rats and orally at 10 mg/kg to rats fasted for 8-10 h. The disappearance of TCE from the blood of intravenously dosed animals was best described by a two-compartment open pharmacokinetic model. The volume of the central compartment (V_c) approximated the rats' blood volume (50-70 ml/kg). The volume distribution (V_d) and total body clearance (CL_T) decreased with increase in dose. The terminal half-life ($t_{1/2}$) was about 120 min and was not affected by increases in dose. TCE was rapidly absorbed after oral dosing, with blood concentrations peaking between 6 and 10 min. The oral to intravenous bioavailability of TCE was 60-80% in nonfasted animals. The terminal $t_{1/2}$ in fasted, orally dosed rats was identical to that when fasted rats were given the same dose intravenously. In fasted rats, bioavailability of an oral dose was greater than 90%, and peak levels in the blood were 2-3 times as high as in nonfasted rats.

INTRODUCTION

Trichloroethylene (TCE) is a halogenated hydrocarbon used extensively in industry. Its low volatility, nonflammability, and high lipid solubility make it a useful solvent for degreasing metals. It also finds use as a dry cleaning solvent, in organic syntheses, and in refrigerants and fumigants. TCE has been used in the past as an inhalation anesthetic (Trilene, Great Britain; Trethylene, United States) and in the decaffeination of coffee. TCE has been widely used in such products as Carbona cleaning fluid and Carbona No. 10 (containing approximately 40% TCE). These commercial products have been subject to abuse by "solvent sniffers" (Alapin, 1973). Although production of TCE has been declining in recent years in the United States, it has been estimated that 133,000 metric tons of TCE were produced in 1980 (U.S. International Trade Commission, 1980).

Supported in part by U.S. Environmental Protection Agency grant R808282.

The National Institute for Occupational Safety and Health (NIOSH) (1978) has estimated that 3.5 million persons are occupationally exposed to TCE and that at least 100,000 persons are exposed on a full-time basis, with 67% of these working under conditions where control measures are inadequate or lacking. Recently, TCE has also been found to be a contaminant of some drinking-water supplies and an air pollutant (IARC, 1979; EPA, 1984). Overexposure to TCE vapor in industrial environments has resulted in manifestations of central nervous system (CNS) depression ranging in severity from mild headache and dizziness to lack of coordination and unconsciousness (NIOSH, 1978). Overexposure to TCE has produced lethal respiratory depression and cardiac arrhythmias (NIOSH, 1973; Environmental Protection Agency (EPA), 1984). A number of cases of sudden inexplicable deaths have occurred following exposure to TCE in industrial and in solvent-abuse settings (Bass, 1970; Reinhardt et al., 1971; EPA, 1984). These deaths have apparently resulted from cardiac arrhythmias, which were triggered by exposure to TCE in conjunction with physical exertion and/or stress.

TCE does not appear to be a potent toxin or carcinogen. TCE proved to be one of the least toxic of a series of halocarbons tested in mice (Klaassen and Plaa, 1966) and dogs (Klaassen and Plaa, 1967). Near-lethal acute doses had to be given intraperitoneally to produce liver or kidney injury. Reviews of studies of persons exposed to TCE occupationally and in abuse cases reveal that the chemical is also relatively nontoxic to humans (NIOSH, 1973, 1978, EPA, 1984), although there are occasional reports of severe poisoning. Alcohol ingestion appears to be implicated in a large percentage of the cases of serious toxic injury (Kleinfeld and Tabershaw, 1954; Gutch et al., 1965; Priest and Horn, 1965; Clearfield, 1970; Seage and Burns, 1971). The results of mutagenicity and carcinogenicity studies of TCE have recently been reviewed by EPA (1984). Purified TCE has been found to be weakly mutagenic in some in vitro assays. Although chronic dosing studies have been conducted using several rodent species, TCE has been clearly shown to be carcinogenic in only one strain of mouse (NTP, 1983).

A number of researchers have speculated that hepatotoxic, mutagenic, and carcinogenic effects of TCE are related to its conversion to a reactive intermediate by microsomal mixed-function oxidases. It has been speculated that this reactive molecular species is 1,1,2-trichloroethylene oxide (Hathway, 1980; Greim et al., 1975; Leibman and Ortiz, 1977). The epoxide is apparently rapidly converted to a glycol, which rearranges and then hydrates to form chloral hydrate. Chloral hydrate is further converted to trichloroacetic acid and trichloroethanol, which are excreted in the urine along with glucuronide conjugates. It has been suggested (Hathway, 1980) that the relative harmlessness of TCE in rats and humans, when compared to other halocarbons such as 1,1-dichloroethylene, is due to this detoxification pathway. A number of investigators have concluded that humans metabolize TCE in a manner similar to rodents and excrete trichloroacetic acid and

trichloroethanol in the urine (Ertle et al., 1972; Ikeda and Ohtsuji, 1972; Ideka, 1977). Miller and Guengerich (1983) recently studied the metabolism of TCE by human liver microsomes. No TCE oxide was detected, though chloral and DNA and protein adducts were formed at about the same rate as in liver microsomes from rats. The finding of levels of protein and DNA adducts higher in mouse than in rat microsomes and isolated hepatocytes may help to explain the aforementioned species differences in the carcinogenicity studies.

The pharmacokinetics of TCE has been studied extensively in humans and experimental animals exposed to the chemical by inhalation (Filser and Bolt, 1979; Sato et al., 1977; Savolainen, 1981; Monster, 1979; Stewart et al., 1970; Astrand and Ovrum, 1976). TCE is readily absorbed from the lung into the systemic circulation, as Astrand and Ovrum (1976) measured relatively high concentrations of TCE in the arterial blood of men 2 min after the subjects began to inhale the chemical. Astrand and Ovrum (1976) reported that 50-60% of inspired TCE was absorbed systemically by subjects inhaling 100 or 200 ppm TCE. Ogata et al. (1971) found that 65-75% of the TCE retained by humans during inhalation sessions was excreted as urinary metabolites. The proportion of the dose of TCE which is metabolized has been shown in animal studies to be a function of the dose administered. At higher doses, a larger percentage of the administered dose appears in the breath as unchanged TCE, and a smaller percentage appears as metabolites in the urine (Parchman and Magee, 1982; Stott et al., 1982).

Very little is known about the systemic absorption and disposition of ingested TCE, despite its presence in drinking water and in some food products. Surprisingly, there is also little definitive information on the absorption and pharmacokinetics of other halocarbons and hydrocarbons following their oral administration. Most pharmacokinetic and toxicological data involves inhalation exposures, as the majority of prior interest in health hazards of these volatile chemicals has centered around exposures in occupational settings. It is unclear, however, whether the results of inhalation studies can be used to accurately predict the consequences of ingestion of the chemicals. Compounds absorbed from the lung into the pulmonary circulation will be transported to the brain and other target organs without having to first pass through an excretory organ. Compounds absorbed from the gastrointestinal (GI) tract are likely to be subject to first-pass elimination by the liver and lungs. A significant amount of a volatile, extensively metabolized halocarbon such as TCE may thus be metabolically degraded and/or exhaled before reaching distant target organs. Therefore, it does not seem prudent to make direct route-to-route extrapolations in the absence of knowledge of the absorption and kinetics of orally administered TCE. The current investigation was undertaken to characterize the absorption and disposition of ingested TCE, and to contrast the kinetics of the chemical following intravenous injection with that following oral administration.

MATERIALS AND METHODS

Male Sprague-Dawley rats (Timco Breeding Laboratories, Houston, Tex.) weighing between 300 and 350 g were used in these experiments. The animals were maintained in a temperature and humidity controlled environment with light between 6 a.m. and 6 p.m. Tap water and commercial rat food were provided. For the intravenous experiments, rats were not fasted before dosing. For the oral studies, one group of rats was deprived of food for 8-10 h before dosing, but the other groups were allowed free access to food. All animals had free access to water at all times.

Animal Dosing

Doses employed were 5, 10, and 25 mg/kg for the intravenous and oral study in nonfasted animals, and 10 mg/kg for the oral study in fasted animals. Trichloroethylene (99+% minimum purity; Analabs, North Haven, Conn.) was administered as a 50% aqueous suspension in PEG 400. The doses were prepared by adding an appropriate amount of TCE to 1.5 ml of previously chilled 1:1 solution of PEG 400 and water in a glass reaction vessel. The vial was sealed with a Hycar stopper and its contents were vortexed for 10-20 sec. Due to solubility problems with the 25-mg/kg dose, it was necessary not only to vortex but to sonicate the mixture for 1 min in order to reduce the size of TCE globules sufficiently to provide a stable suspension. For the intravenous experiments, 0.5 ml TCE suspension was injected into the dorsal vein of the penis of the lightly etherized rats. For the oral study, 0.5 ml TCE suspension was administered intragastrically using a gavage needle. All animals were dosed between 8 and 10 a.m., which was 2-4 h into their inactive cycle.

Blood Collection

Blood samples of 0.1 ml were collected at increasing intervals from 0 to 10 h post dosing from the caudal artery of each lightly etherized rat by the method of Putcha et al. (1982). The samples were immediately transferred to chilled 1.5-ml glass reaction vessels, sealed with a Hycar stopper, and stored in an ice bath for up to 2 h prior to analysis.

Analysis

Blood samples were incubated in a water bath for 15-20 min at 37°C for equilibration of TCE between the blood and the headspace of the vessel. Headspace vapor (0.1 ml) was withdrawn using a Precision glass syringe equipped with a Teflon plunger and was injected into a Varian 3700 gas chromatograph. The gas chromatograph conditions were as follows:

Injection port temperature	120°C
Column temperature	100°C

SL 032772

Carrier gas flow rate	15 ml/min (N ₂)
Detector temperature	280°C
Column	6 ft X $\frac{1}{8}$ in stainless steel, packed with Durapak
Detector	⁶³ Ni, electron capture

A standard plot was prepared by spiking 2 μ l of increasing concentrations of TCE, dissolved in methanol, into 0.1 ml heparinized, freshly collected whole blood contained in a chilled glass reaction vessel. The vessel was sealed with a Hycar stopper and vortexed for 5 s for mixing. The samples were then incubated and analyzed as described above. The standard plot for TCE was curvilinear, apparently due to saturation of vapor in the headspace of the vessel. A FORTRAN computer program was written to compute TCE concentrations from these data. These data were very reproducible, with a coefficient of variation for individual samples of less than 5%.

Statistical Analysis

An analysis of variance test was performed to determine the significance of changes in pharmacokinetic parameters with increase in doses. Student's *t*-test and ANOVA were used to determine the significance of changes in half-life and bioavailability between intravenously and orally dosed animals, and between fasted and nonfasted animals.

RESULTS

Blood concentration versus time data were analyzed using ESTRIP (Brown and Manno, 1978) and NONLIN (Metzler et al., 1974) computer programs. The intravenous data were best defined by a biexponential equation of the form:

$$C = Ae^{-\alpha t} + Be^{-\beta t} \quad (1)$$

where *C* is the blood concentration of TCE at time *t*, *A* and *B* are the zero-time intercepts, and α and β are hybrid rate constants, representing processes of distribution and elimination.

Model independent pharmacokinetic parameters were calculated using the following equations.

The apparent volume of distribution (*V_β*) was calculated using the formula

$$V_{\beta} = \frac{\text{dose}}{\beta \int_0^{\infty} C dt} \quad (2)$$

where $\int_0^{\infty} C dt$ is the area under the blood concentration versus time curve to time infinity (AUC) of TCE. The apparent steady-state volume of distribution (*V_{ss}*) was calculated using the formula

$$V_{ss} = \frac{\text{dose} \int_0^{\infty} tC dt}{(\int_0^{\infty} C dt)^2} \quad (3)$$

The total body clearance (CL_T) of TCE was calculated using the formula

$$CL_T = \frac{\text{dose}}{\int_0^{\infty} C dt} \quad (4)$$

The volume of distribution of the central compartment (V_c) was calculated using the formula

$$V_c = \frac{\text{dose}}{A + B} \quad (5)$$

The following formula was used to determine the bioavailability of orally administered TCE in experiments in which equivalent doses were given orally (po) and intravenously (iv):

$$\text{Bioavailability} = \frac{(AUC)_{po}}{(AUC)_{iv}} \quad (6)$$

The biological half-life ($t_{1/2}$) was calculated from the relationship

$$t_{1/2} = \frac{0.693 V_{\beta}}{CL_T} \quad (7)$$

Representative blood concentration versus time profiles of three animals that received TCE intravenously are shown in Fig. 1. One rat was given 25 mg/kg, the second 10 mg/kg and the third 5 mg/kg. It can be seen that the pattern of elimination of TCE is similar in all three animals. It can also be seen that the rate of elimination is comparable at all three dosage levels. Mean $t_{1/2}$ values of groups of rats given 5, 10, or 25 mg TCE/kg are included in Table 1. There is no change in $t_{1/2}$ with increase in dose.

Pharmacokinetic parameters calculated from the intravenous data are presented in Table 1. The volume of the central compartment (V_c) does not vary significantly with dose. The calculated values for V_c very closely approximate the blood volume of 300-350-g rats (50-70 ml/kg). The apparent volume of distribution (V_{β}) and apparent steady-state volume of distribution (V_{ss}) vary inversely with dose. Both V_{β} and V_{ss} exhibit a significant decrease with increase in dosage level. These data are presented as bar graphs in Fig. 2. Although $t_{1/2}$ does not vary, total body clearance (CL_T) decreases significantly with increase in dose (Table 1).

Following oral administration, TCE was rapidly absorbed from the gastrointestinal tract. Peak arterial blood concentrations were reached between 6 and 10 min post dosing. Typical blood concentration versus time

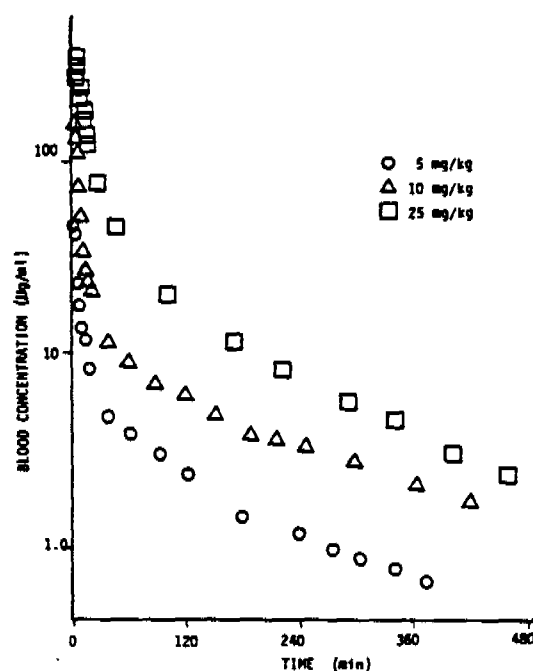


FIGURE 1. Typical blood TCE concentration versus time profiles following intravenous injection of TCE. One rat received a 25-mg/kg dose, the second received 10 mg/kg, and the third received 5 mg/kg. Arterial blood levels were measured at 2- to 60-min intervals for up to 450 min.

profiles of 2 animals which received a 10-mg/kg oral dose of TCE are depicted in Fig. 3. One animal was fasted prior to dosing, and the other was allowed free access to food. For sake of comparison, the profile of an animal given 10 mg TCE/kg intravenously is included. It can be seen that the concentration of TCE in the blood of each orally dosed rat is significantly elevated at the first sampling time (0.5 min). The peak blood level in the fasted animal is 2-3 times higher than in the nonfasted animal.

TABLE 1. Trichloroethylene Pharmacokinetic Parameters^a in Intravenously Dosed Rats

TCE dose (mg/kg)	V_c (ml/kg)	V_{ss}^b (ml/kg)	V_β^b (ml/kg)	$t_{1/2}$ (min)	CL_T^c (ml/min·kg)
5	86 ± 12	537 ± 39	708 ± 58	115 ± 7	4.3 ± 0.2
10	65 ± 10	412 ± 60	581 ± 78	121 ± 12	3.4 ± 0.2
25	73 ± 7	314 ± 79	487 ± 73	110 ± 9	3.0 ± 0.3

^aValues are means ± SD for groups of four to six rats.

^bSignificant decrease with increase in dose ($p < 0.005$, analysis of variance).

^cSignificant decrease with increase in dose ($p < 0.001$, analysis of variance).

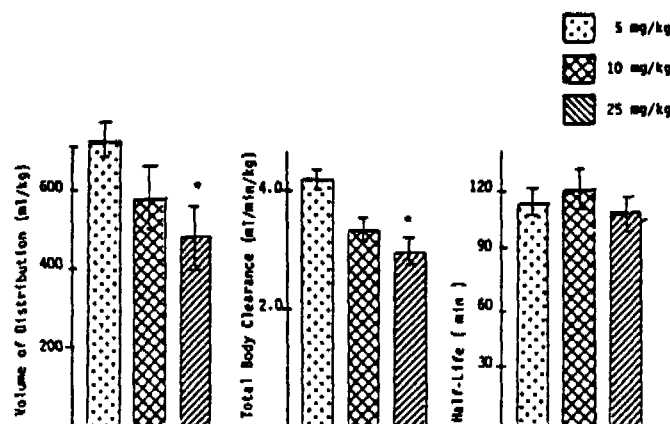


FIGURE 2. Influence of dose on pharmacokinetic parameters in intravenously dosed rats. Brackets encase mean $\pm$ SD for groups of four to six rats. Asterisks denote statistically significant decrease with increase in dose ($p < 0.005$, analysis of variance).

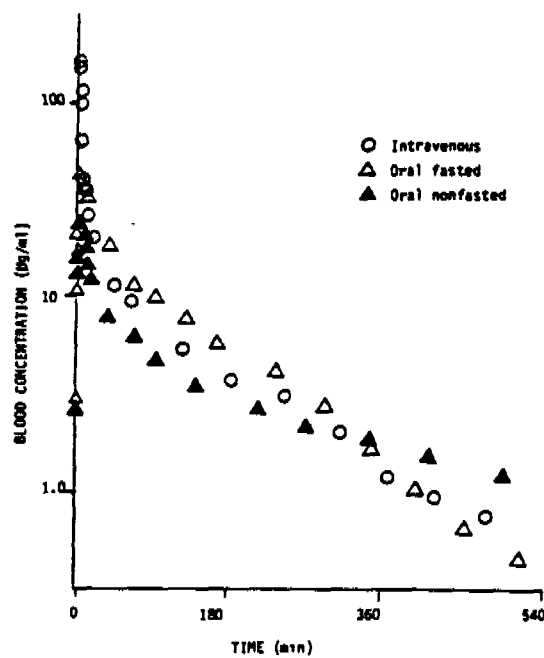


FIGURE 3. Typical blood TCE concentration versus time profiles following oral and intravenous administration of TCE. A fasted and a nonfasted rat were each given 10 mg TCE/kg orally. A third rat received 10 mg TCE/kg intravenously. Arterial blood levels were measured at 2- to 90-min intervals for up to 8 h.

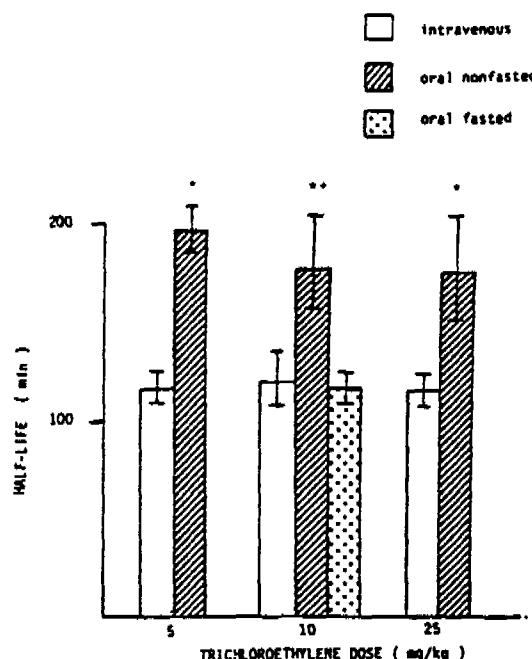


FIGURE 4. Influence of dose and food intake on biological half-life of TCE in orally and intravenously dosed rats. Brackets encase mean $\pm$ SD for groups of four to six animals. Asterisks denote statistically significant difference between intravenous and oral nonfasted groups ($p < 0.001$, Student's t -test). The + denotes significant difference between oral nonfasted and oral fasted groups ($p < 0.001$, Student's t -test).

Interestingly, the peak levels occur at approximately the same time post dosing in the two animals.

Comparison of the representative blood profiles of the orally dosed animals in Fig. 3 shows a difference between the fasted and nonfasted rats in the assumed postabsorptive phase (i.e., terminal elimination phase). It appears that TCE is eliminated more slowly from the bloodstream of the nonfasted animal. Indeed, when one examines the mean biological $t_{1/2}$ values presented in Fig. 4, it is obvious at the 10-mg/kg dosage level that $t_{1/2}$ in the nonfasted group is significantly longer than in the fasted group or in the intravenously dosed group. The $t_{1/2}$ of TCE in nonfasted, orally dosed rats ranges from 174 to 196 min over the range of doses utilized, although there is no apparent relationship between dose and $t_{1/2}$. Similarly, $t_{1/2}$ values of the intravenous groups do not differ significantly with dose. Examination of the representative blood profiles in Fig. 3 reveals that the overall pattern of elimination is similar in the intravenously dosed and the fasted, orally dosed animals. Predictably, the mean biological $t_{1/2}$ for the intravenously dosed groups (121 $\pm$ 12 min) is very similar to that of the fasted, orally dosed group (112 $\pm$ 15 min) (Fig. 4).

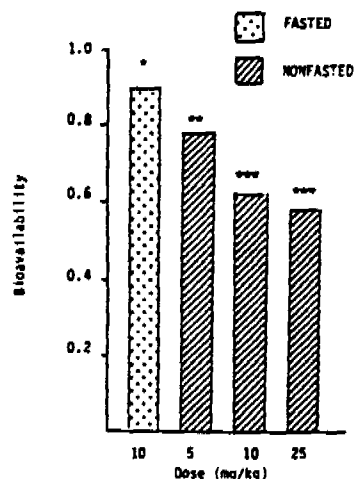


FIGURE 5. Bioavailability of orally administered TCE in fasted and nonfasted rats. Bars represent the mean of groups of four to six animals. *Nonsignificant difference between iv and oral fasted groups ($p > 0.1$, Student's t -test). **Significant difference between iv and oral nonfasted groups ($p < 0.05$, Student's t -test). ***Significant difference between iv and oral nonfasted groups ($p < 0.001$, Student's t -test).

Results of the calculation of the bioavailability of orally administered TCE are presented in Fig. 5. It can be seen that the bioavailability of the 10-mg/kg oral dose is substantially greater in fasted (>90%) than in nonfasted (~63%) rats. Peak blood levels are two to three times higher in the fasted animals. It can also be seen in Fig. 5 that there is a trend towards a decrease in bioavailability with increase in dose in the nonfasted rats. Bioavailability in these animals ranged from 78% in the 5-mg/kg group to 58% in the 25-mg/kg group.

DISCUSSION

Findings in this study indicate that TCE is well absorbed from the GI tract. There is surprisingly little information to date on the extent or rapidity of GI absorption of halocarbons or other volatile organics. Several investigators have conducted studies in which animals are dosed orally, and pulmonary, urinary, and fecal elimination of parent compound and metabolites are monitored post dosing. The percentage recovery of the total dose of 1,1-dichloroethylene in two such studies (McKenna et al., 1978; Reichert et al., 1979) is reported to be quite high, indicative of relatively complete absorption. Such findings, however, do not elucidate the absorption characteristics or the kinetics of the chemical following its ingestion. Withey (1976a) did publish the results of a study in which he monitored blood levels of vinyl chloride in rats dosed by gavage. The investigator

reported that uptake of vinyl chloride is very rapid, but he encountered such a high degree of intersubject variability that conclusions regarding peak blood levels and bioavailability were not possible.

Results of the current study show that TCE is quickly and extensively absorbed from the GI tract of fasted rats. The rapid appearance of TCE in peripheral arterial blood (at the initial 0.5-min sampling time) and the short time to peak blood levels (6-10 min after intragastric dosing) suggest that TCE is well absorbed from the stomach. It is possible that some of the gavage fluid entered the small intestine, through the total volume given (~0.5 ml) was kept small to minimize this occurrence. The very rapid systemic absorption also suggests that absorption is not diffusion limited. Diffusion of a chemical through the contents of the GI tract normally determines the concentration of chemical at the mucosal surface. The rate of absorption of a readily diffusible molecule like TCE is a function of the TCE concentration gradient across the mucosal membrane. Diffusion of TCE through the aqueous suspension would appear to be too slow to account for the rapid, extensive absorption of the chemical. As TCE is volatile, it may readily volatilize in the GI tract and thereby substantially increase the concentration of TCE at the mucosal surface. Studies of vinyl chloride (Withey, 1976a) and 1,2-dichloroethane (Reitz et al., 1982), two other very volatile halocarbons, similarly demonstrate extremely rapid systemic uptake in orally dosed rats.

The presence of food in the GI tract appears to alter the absorption characteristics of TCE. Part of the oral dose administered to nonfasted rats is absorbed at a rapid rate, as reflected by relatively high blood levels at the initial sampling interval (0.5 min) and by the same time to peak blood levels as in the fasted animals (Fig. 3). Nevertheless, peak blood levels in the nonfasted rats are two to three times lower than in the fasted rats. Predictably, bioavailability is significantly greater in the latter group. It would therefore be reasonable to assume that the toxic potential of TCE may be greater when the chemical is ingested on an empty stomach. The findings of lower peak blood levels and a longer terminal $t_{1/2}$ in nonfasted rats suggest that a portion of the dose is absorbed at a relatively slow rate. Substantial concentrations of TCE were measured in the blood of nonfasted rats for up to 9 h after dosing. It is likely that some TCE partitions into lipid in food in the GI tract. This TCE would be available for systemic absorption only on its subsequent diffusion out of the food or on digestion of the food. Thus, the type and amount of food present in the GI tract may play an important role in the systemic absorption and potential toxicity of ingested haloalkanes. Chieco et al. (1981) report that the administrative vehicle significantly influences the hepatotoxicity of 1,1-dichloroethylene in orally dosed rats. These investigators conclude that uptake of the chemical from the GI tract is most rapid from an aqueous Tween vehicle, intermediate from corn oil, and most prolonged from indigestible mineral oil.

Withey also reports that the GI absorption of vinyl chloride (1976a) and styrene (1976b) are delayed when the chemicals are given to rats in vegetable oil rather than in an aqueous solution.

Although biological $t_{1/2}$ was longer in oral nonfasted animals than in oral fasted or intravenously dosed animals, $t_{1/2}$ did not vary with dose in any group. Withey and Collins (1980), however, have reported an increase in the $t_{1/2}$ of TCE in rats as a function of intravenous dose. They noted an increase in $t_{1/2}$ from 18 min for a 3-mg/kg dose to 56 min for a 15-mg/kg dose. In view of such short $t_{1/2}$ values, particularly at the low dose, it would appear that limitations in assay sensitivity prevented the investigators from monitoring the blood concentrations long enough to accurately define the terminal elimination phase. Judging from the results of several studies in humans, it appears that $t_{1/2}$ values increase with increasing duration of the postexposure monitoring period. Sato et al. (1977) and Muller et al. (1974) exposed human subjects to 100 ppm TCE vapor for 4 and 6 h, respectively. Sato et al. (1977) monitored blood TCE levels for 10 h post exposure and found a $t_{1/2}$ of 3.5 h. In contrast, Muller et al. (1974) monitored blood levels for almost 60 h and found the $t_{1/2}$ to be approximately 25 h.

We observed both volume and clearance changes with increasing intravenous dose of TCE, but no significant change in $t_{1/2}$. As can be seen on inspection of Eq. (7), volume and clearance are both independent parameters that contribute to $t_{1/2}$, which is the dependent parameter. As there was a progressive decrease in both V_d and CL_T with an increase in dose, the half-life did not change. Decrease in clearance of TCE and other halocarbons with an increase in inhalation dose has been well documented in the literature, and attributed to saturation of the metabolic capacity of test organisms (Filser and Bolt, 1979; Stott et al., 1982). Very little data, however, have been published pertaining to the clearance of TCE following intravenous or oral administration, and reports are inconsistent. Some observers have reported little or no change in clearance with increasing intravenous dose (Withey and Collins, 1980), while others have demonstrated metabolic saturation at quite high doses (Parchman and Magee, 1982). Our observations are consistent with decreased metabolic clearance with increasing dose. One would expect a larger proportion of TCE to be eliminated unchanged in the breath at the higher dosage levels. This phenomenon is commonly observed with halocarbons such as TCE (Stott et al., 1982), which have alternate routes of elimination (i.e., metabolism and exhalation).

The volume of distribution of the central compartment (V_c) of 65-86 ml/kg was relatively small, suggesting pronounced 2-compartment characteristics and distribution of TCE principally in the tissue compartment. Theoretically, when sampling levels of a chemical in whole blood, the total blood volume (50-70 ml/kg) is the minimum volume that a chemical can distribute into after intravenous injection. V_c is thus the proportionality constant relating blood concentrations to the amount of chemical in the body at time zero. This proportionality constant increases as the chemical

diffuses through the body until it reaches a plateau, which is termed apparent volume of distribution (V_β). TCE V_β values calculated from a previous report (Withey and Collins, 1980) are smaller (280–475 ml/kg) than those we find. Withey and Collins, 1980) conducted their experiments for approximately 3 h, while blood concentrations of TCE were measured for 6–8 h in the present investigation. It is important to recognize here that the calculated V_β for a compound that diffuses slowly may change as a function of data collection and of the length of time that blood concentrations are followed (Niazi, 1976). It is pointed out by Filser and Bolt (1979) that halocarbons with relatively high boiling points, like TCE, accumulate in tissues to a greater extent but take longer to reach equilibrium than halocarbons with lower boiling points (e.g., vinyl chloride, 1,1-dichloroethylene). The concept of direct transfer of lipophilic chemicals by diffusion between neighboring tissues (Pearl et al., 1965) is worthy of note here. Sato et al. (1977) have suggested that blood perfusion to tissues is not the sole means of transfer of TCE to the various tissues of the body, but that TCE can also diffuse between adjacent tissues.

The decrease we see in apparent volume of distribution (V_β and V_{ss}) with increasing dose of TCE is unexpected. In a similar study with 1,1-dichloroethylene, we recently observed an increase in V_β with increasing dose (Putcha et al., 1984, unpublished data). The decrease in V_β and V_{ss} with increasing TCE dose could be indicative of decreased distribution of TCE into the less perfused compartment. Although it is unlikely that adipose tissue would be saturated at the doses employed, saturation of other tissues in the less perfused compartment could contribute to the phenomenon. A decrease in tissue/blood partition coefficients with increasing concentrations of TCE in the blood is another possible explanation. Nevertheless, the reasons for and the significance of the finding remain unclear.

The influence of light ether anesthesia on the pharmacokinetics of TCE was not addressed in these experiments. However, a recent study in our laboratories (Putcha et al., 1984) found no effect of diethyl ether on the pharmacokinetics of intact 1,1-dichloroethylene (DCE) in the rat. Although an effect of diethyl ether on the pharmacokinetics of TCE cannot be ruled out, in view of the previous findings with DCE, we feel the light anesthesia did not influence TCE disposition in rats.

REFERENCES

- Alapin, B. 1973. Trichloroethylene addiction and its effects. *Br. J. Addict.* 68:331–335.
- Astrand, I. and Ovrum, P. 1976. Exposure to trichloroethylene. I. Uptake and distribution in man. *Scand. J. Work Environ. Health* 2:199–211.
- Bass, M. 1970. Sudden sniffing death. *J. A. M. A.* 212:2075–2079.
- Brown, R. D. and Manno, J. E. 1978. ESTRIP, a BASIC computer program for obtaining initial polyexponential parameter estimates. *J. Pharm. Sci.* 67:1687–1691.
- Chieco, P., Moslen, M. T., and Reynolds, E. S. 1981. Effect of administrative vehicle on oral 1,1-dichloroethylene toxicity. *Toxicol. Appl. Pharmacol.* 57:146–155.

- Clearfield, H. R. 1970. Hepatorenal toxicity from sniffing spot-remover (trichloroethylene). *Digest. Dis.* 15:851-856.
- Environmental Protection Agency. 1984. Health assessment document for trichloroethylene. EPA-600/8-82-006B. U.S. Environmental Protection Agency, Washington, D.C.
- Ertle, T., Henschler, D., Muller, G., and Spassowski, M. 1972. Metabolism of trichloroethylene in man. I. The significance of trichloroethanol in long-term exposure conditions. *Arch. Toxicol.* 29: 171-188.
- Filser, J. G. and Bolt, H. M. 1979. Pharmacokinetics of halogenated ethylenes in rats. *Arch. Toxicol.* 42:123-136.
- Greim, H., Bonse, G., Radwan, Z., Reichert, D., and Henschler, D. 1975. Mutagenicity in vitro and potential carcinogenicity of chlorinated ethylenes as a function of metabolic oxirane formation. *Biochem. Pharmacol.* 24:2013-2017.
- Gutch, C. F., Tomhave, W. G., and Stevens, S. C. 1965. Acute renal failure due to inhalation of trichloroethylene. *Ann. Intern. Med.* 63:128-134.
- Hathway, D. E. 1980. Consideration of the evidence for mechanisms of 1,1,2-trichloroethylene metabolism, including new identification of its dichloroacetic acid and trichloroacetic acid metabolites in mice. *Cancer Lett.* 8:263-269.
- IARC. 1979. Trichloroethylen. *IARC Monogr. Eval. Carcinog. Risk Chem. Man* 20:545-572.
- Ikeda, M. 1977. Metabolism of trichloroethylene and tetrachloroethylene in human subjects. *Environ. Health Perspect.* 21:239-245.
- Ikeda, M. and Ohtsuiji, H. 1972. A comparative study of the excretion of Fujiwara reaction-positive substances in urine of humans and rodents given trichloro- and tetrachloro-derivatives of ethane and ethylene. *Br. J. Ind. Med.* 29:99-104.
- Klaassen, C. D. and Plaa, G. L. 1966. Relative effects of various chlorinated hydrocarbons on liver and kidney function in mice. *Toxicol. Appl. Pharmacol.* 9:139-151.
- Klaassen, C. D. and Plaa, G. L. 1967. Relative effects of various chlorinated hydrocarbons on liver and kidney function in dogs. *Toxicol. Appl. Pharmacol.* 10:119-131.
- Kleinfeld, M. and Tabershaw, I. R. 1954. Trichloroethylene toxicity. Report of five fatal cases. *Arch. Ind. Hyg. Occup. Med.* 10:134-141.
- Leibman, K. C. and Ortiz, E. 1977. Metabolism of halogenated ethylenes. *Environ. Health Perspect.* 21:91-97.
- McKenna, M. J., Zempel, J. A., Madrid, E. O., Braun, W. H., and Gehring, P. J. 1978. Metabolism and pharmacokinetic profile of vinylidene chloride in rats following oral administration. *Toxicol. Appl. Pharmacol.* 45:821-835.
- Metzler, C. M., Elfring, G. K., and McEwen, A. J. 1974. A package of computer programs for pharmacokinetic modeling. *Biometrics* 30:562-563.
- Miller, R. E. and Guengerich, F. P. 1983. Metabolism of trichloroethylene in isolated hepatocytes, microsomes, and reconstituted enzyme systems containing cytochrome P-450. *Cancer Res.* 43:1145-1152.
- Monster, A. C. 1979. Difference in uptake, elimination and metabolism in exposure to trichloroethylene, 1,1,1-trichloroethane and tetrachloroethylene. *Int. Arch. Occup. Environ. Health* 42: 311-317.
- Muller, G., Spassowski, M., and Henschler, D. 1974. Metabolism of trichloroethylene in man. II. Pharmacokinetics of metabolites. *Arch. Toxicol.* 32:288-295.
- Niazi, S. 1976. Volume of distribution as a function of time. *J. Pharm. Sci.* 65:452-454.
- National Institute for Occupational Safety and Health. 1973. Criteria for a recommended standard—Occupational exposure to trichloroethylene. DHEW publ. no. HSM-73-11025, National Institute for Occupational Safety and Health, Washington, D.C.
- National Institute for Occupational Safety and Health. 1978. Special occupational hazard review of trichloroethylene. DHEW publ. no. 78-130, National Institute for Occupational Safety and Health, Washington, D.C.
- National Toxicology Program. 1983. NTP technical report on the carcinogenesis bioassay of trichloroethylene in F344/N rats and B6C3F1/N mice. NIH publ. no. 83-1799, National Toxicology Program, Research Triangle Park, N.C., and Bethesda, Md.

- Ogata, M., Takatsuka, Y., and Tomokuni, K. 1971. Excretion of organic chlorine compounds in the urine of persons exposed to vapours of trichloroethylene and tetrachloroethylene. *Brit. J. Ind. Med.* 28:386-391.
- Parchman, L. G. and Magee, P. N. 1982. Metabolism of [^{14}C]trichloroethylene to $^{14}\text{CO}_2$ and interaction of a metabolite with liver DNA in rats and mice. *J. Toxicol. Environ. Health* 9:797-813.
- Pearl, W., Rackow, H., Salanitro, E., Wolf, G. L., and Epstein, R. M. 1965. Intertissue diffusion effect of inert fat-soluble gases. *J. Appl. Physiol.* 20:621-627.
- Priest, R. J. and Horn, Jr., R. C. 1965. Trichloroethylene intoxication. A case of acute hepatic necrosis possibly due to this agent. *Arch. Environ. Health* 11:361-365.
- Putcha, L., Bruckner, J. V., Muralidhara, S., and Feldman, S. 1982. A simple method for repetitive blood sampling from rats. *J. Pharmacol. Methods* 8:145-149.
- Reichert, D., Werner, H. W., Metzler, M., and Henschler, D. 1979. Molecular mechanism of 1,1-dichloroethylene toxicity: Excreted metabolites reveal different pathways of reactive intermediates. *Arch. Toxicol.* 42:159-169.
- Reinhardt, C. F., Azar, A., Maxfield, M. E., Smith, Jr., P. E., and Mullin, L. S. 1971. Cardiac arrhythmias and aerosol "sniffing." *Arch. Environ. Health* 22:265-279.
- Reitz, R. H., Fox, T. R., Ramsey, J. C., Quast, J. F., Langvardt, P. W., and Watanabe, P. G. 1982. Pharmacokinetics and macromolecular interactions of ethylene dichloride in rats after inhalation or gavage. *Toxicol. Appl. Pharmacol.* 62:190-204.
- Sato, A., Nakajima, T., Fujiwara, Y., and Murayama, N. 1977. A pharmacokinetic model to study the excretion of trichloroethylene and its metabolites after an inhalation exposure. *Br. J. Ind. Med.* 34:56-63.
- Savolainen, H. 1981. Pharmacokinetics, pharmacodynamics and aspects of neurotoxic effects of four inhaled aliphatic chlorohydrocarbon solvents as relevant in man. *Eur. J. Drug Metab. Pharmacokinet.* 6:85-95.
- Seage, A. J. and Burns, M. W. 1971. Pulmonary oedema following exposure to trichloroethylene. *Med. J. Australia* 2:484-486.
- Stewart, R. D., Dodd, H. C., Gay, H. H., and Erley, D. S. 1970. Experimental human exposure to trichloroethylene. *Arch. Environ. Health* 20:64-71.
- Stott, W. T., Quast, J. F., and Watanabe, P. G. 1982. The pharmacokinetics and macromolecular interactions of trichloroethylene in mice and rats. *Toxicol. Appl. Pharmacol.* 62:137-151.
- U.S. International Trade Commission. 1980. *Synthetic Organic Chemicals. United States Production and Sales.* U.S. International Trade Commission, Washington, D.C.
- Withey, R. R. 1976a. Pharmacodynamics and uptake of vinyl chloride monomer administered by various routes to rats. *J. Toxicol. Environ. Health* 1:381-394.
- Withey, J. R. 1976b. Quantitative analysis of styrene monomer in polystyrene and foods including some preliminary studies of the uptake and pharmacodynamics of the monomer in rats. *Environ. Health Perspect.* 17:125-133.
- Withey, J. R., and Collins, B. J. 1980. Chlorinated aliphatic hydrocarbons used in the food industry: The comparative pharmacokinetics of methylene chloride, 1,2-dichloroethane, chloroform and trichloroethylene after i.v. administration in the rat. *J. Environ. Pathol. Toxicol.* 3:313-332.

Received August 20, 1984
Accepted October 17, 1984

SL 032783