TECHNICAL SUPPORT DOCUMENT

REPORT TO THE AIR RESOURCES BOARD ON CARBON TETRACHLORIDE

PART B:

HEALTH EFFECTS OF CARBON TETRACHLORIDE

Prepared by

The Epidemiological Studies and Surveillance Section
Department of Health Services
2151 Berkeley Way, Room 515
Berkeley, California 94704-9980

Principal Author:

George V. Alexeeff, Ph.D., Staff Toxicologist

May 6, 1986

Revised May 22, 1987

Reviewed by:

Anna Fan, Ph.D.
Peter C. Flessel, Ph.D.
Norman Gravitz, Ph.D., M.P.H.
Robert A. Hass, Ph.D.
Alexander Kelter, M.D., M.P.H.
Michael J. Lipsett, J.D., M.D.
Raymond R. Neutra, M.D., Dr. P.H.
Calvin Willhite, Ph.D.

TABLE OF CONTENTS

1.0 EXECUTIVE SUMMARY

DHS estimates that approximately 50% of inhaled carbon tetrachloride (CTC) is absorbed by the body. Numerous studies have suggested that CTC is metabolized to a highly reactive trichloromethyl radical which can then react with cellular components to produce acute and chronic toxicity.

Accidental acute human exposures, at concentrations at least five orders of magnitude greater than expected ambient levels, and animal experiments have shown that CTC can produce liver and kidney damage and numerous effects on the nervous system. Chronic exposure to CTC, in occupational settings where CTC concentrations are three to four orders of magnitude higher than current ambient levels, has produced neurological effects and elevation of serum liver enzymes indicating liver damage. Long-term animal exposure to similarly high levels of CTC has produced liver and kidney damage. At current ambient CTC levels, however, no acute or noncarcinogenic chronic effects are expected to occur.

Reproductive effects developed in male animals in response to very high concentrations of CTC. Dosing of pregnant rats with high concentrations of CTC resulted in embryo- and fetotoxicity. Experimental data are inadequate to assess potential human reproductive risk from ambient CTC exposures.

Radiolabeled CTC given to laboratory animals binds to DNA as well as other cellular components. Thus, CTC is potentially genotoxic. However, tests for mutagenicity using standard bacterial and yeast assays and other methods for detecting chromosomal damage have been predominantly negative. This

apparent lack of effect may reflect the inability of those assays to test adequately halogenated hydrocarbons such as CTC or may indicate that CTC is not strongly mutagenic.

Examination of human case reports and epidemiological studies has not shown that CTC causes cancer in humans. However, the carcinogenicity of CTC has been clearly demonstrated in three animal species, which has led the International Agency for Research on Cancer (IARC) to conclude that there is sufficient evidence of carcinogenicity in animals and that, in the absence of adequate data in humans, it is reasonable for practical purposes to regard CTC as if it presents a carcinogenic risk to humans. In the major animal cancer studies CTC was administered orally in a solution with olive or corn oil. None of the studies was conducted by inhalation. In these studies CTC produced malignant tumors in the liver in up to 100% of the animals, and the tumors began to appear 16 weeks after the beginning of the study. On the basis of this experimental work, DHS staff concurs with the findings of IARC. In addition, DHS staff has not found compelling evidence demonstrating the existence of a carcinogenic threshold for CTC.

The DHS recommends adopting portions of the risk assessment performed by the Environmental Protection Agency (EPA), which applied a multistage model to the results of several of the animal studies. For one of the studies time-to-tumor information was incorporated into the multistage model. DHS staff suggests the use of increased lifetime carcinogenic risk values from exposure to carbon tetrachloride ranging from 10 to 42 cancer cases per million people continuously exposed over their lifetimes to 1 microgram CTC

per cubic meter of air (1 $\mu g/m^3$). This range is based on the carcinogenic potencies estimated from two animal studies.

The range of risk values represents several sources of uncertainty, including statistical uncertainty due to the small numbers of animals used in the bioassays and the variability in the animals' response in experiments using different species and protocols. Other general sources of uncertainty, which did not directly affect the magnitude of the above range of risks, include the choice of the animal-to-human scaling factor, the choice of the extrapolation models, and the large range of extrapolation (three to five orders of magnitude) from the orally administered CTC concentrations used in the animal experiments to current ambient levels.

The lifetime risk values given above represent a range of conservative estimates and are unlikely to be exceeded by the actual risk. A lifetime excess risk of 10-42 per million population must be viewed in the context of the overall probability of developing cancer, which is on the order of 250,000 cases per million population (25%) over a lifetime.

Based on the findings of carcinogenicity and the results of the risk assessment, DHS staff finds that ambient CTC is an air pollutant which may cause or contribute to an increase in mortality or an increase in serious illness, or which may pose a present or potential hazard to human health.

2.0 PHARMACOKINETICS

2.1 Routes of exposure

Carbon tetrachloride (CTC) is lipophilic and is readily absorbed orally and via inhalation. There is some dermal absorption. Acute oral exposure in humans is primarily restricted to accidental ingestion or suicide attempts. However, it was formerly used as an anthelmintic,[1] and medicinal doses occasionally resulted in toxicity (Von Oettingen 1964). Information regarding oral administration is provided below for comparative purposes and to supplement inhalation data. Chronic oral exposure occurs from consumption of contaminated food and water. The major routes of occupational exposure are inhalation and dermal. The toxicokinetics have been reviewed previously by Von Oettingen (1964), Browning (1965), Swinyard (1975), Bergman (1979), Torkelson and Rowe (1981) and others.

2.2 Absorption

2.2.1 Inhalation

Inhalation absorption was first studied by Lehmann and Hasegawa (1910) using rabbits. They reported that the absorption rate decreased from 34.7 to 4.7%, during a 3-hour exposure to 50 mg/L (≈8000ppm). Following exposure of dogs to 15,000 and 20,000 ppm CTC, Von Oettingen et al. (1949, 1950) found

[1] An agent destructive to worms. Recommended oral doses were 2-3 ml for adults, and 0.13 ml per year of age for infants and children up to 15 years old.

that blood CTC levels reached equilibrium in approximately 5 hours. However, exposure to such high concentrations would induce severe toxicity that could result in decreased absorption.

McCollister et al. (1950, 1951) exposed three monkeys via inhalation to an average of 46 ppm of ^{14}C-labelled CTC for 2 to 6 hours. The absorption reportedly occurred at an average rate of 1.34 mg/kg/hour, or 30% of the total weight of CTC inhaled. They observed that absorption of the material ranged from 26 to 37%. The highest absorption rate was obtained during the longest exposure. Equilibrium of CTC between the air and blood was not reached during the course of the experiment. Consequently, absorption following a longer exposure until equilibrium was reached would be expected to be above 37%.

In a human inhalation study by Lehmann and Schmidt-Kehl (1936), individuals were exposed to CTC vapors for up to 30 minutes. The amount of CTC absorbed was calculated from the difference of the amount available and the amount exhaled. Thus the amount of CTC absorbed was estimated indirectly. They reported a range of absorption of 57 to 64%. This study used from 2 to 14 subjects, although the actual number was unspecified. Extrapolating from the primate studies of McCollister et al. (1950, 1951), it is unlikely that equilibrium was attained using such short exposure periods in the human studies.

The above noted primate and human studies, even with their limitations, represent the two best estimates for absorption reported in the literature. The staff at DHS believe that these two studies, with their shortcomings,

should be given equal weight in estimating an absorption value for humans. The human study is lacking due to incomplete documentation of exposure of the subjects and the indirect absorption method used. The primate study directly measured absorption; however, it is not clear how similar human exposure would be. Consequently, the reported absorption coefficient values were given equal weight in estimating a 50% absorption coefficient that will be utilized in the quantitative risk assessment in Section 9.0. Although the studies on which this value is based are not ideal, the staff at DHS believes that 50% is the best estimate and that a more precise absorption calculation would change the unit risk estimates by less than a factor of two.

2.2.2 Dermal

Two monkeys were exposed dermally to vapor concentrations of 485 and 1150 ppm for approximately four hours (Beamer et al. 1950, McCollister et al. 1951). CTC blood levels indicated that absorption by this route was relatively low and was not of practical significance. Stewart and Dodd (1964) studied the dermal absorption of CTC in human subjects. Three subjects immersed their thumbs for 30 minutes in a beaker of CTC, and expired air samples were analyzed. CTC was detected in the alveolar air samples within ten minutes and it continued to rise for up to 30 minutes post-exposure. Although absorption of vapor by the dermal route is insignificant, dermal absorption of liquid CTC could result in toxicity.

2.2.3 Oral

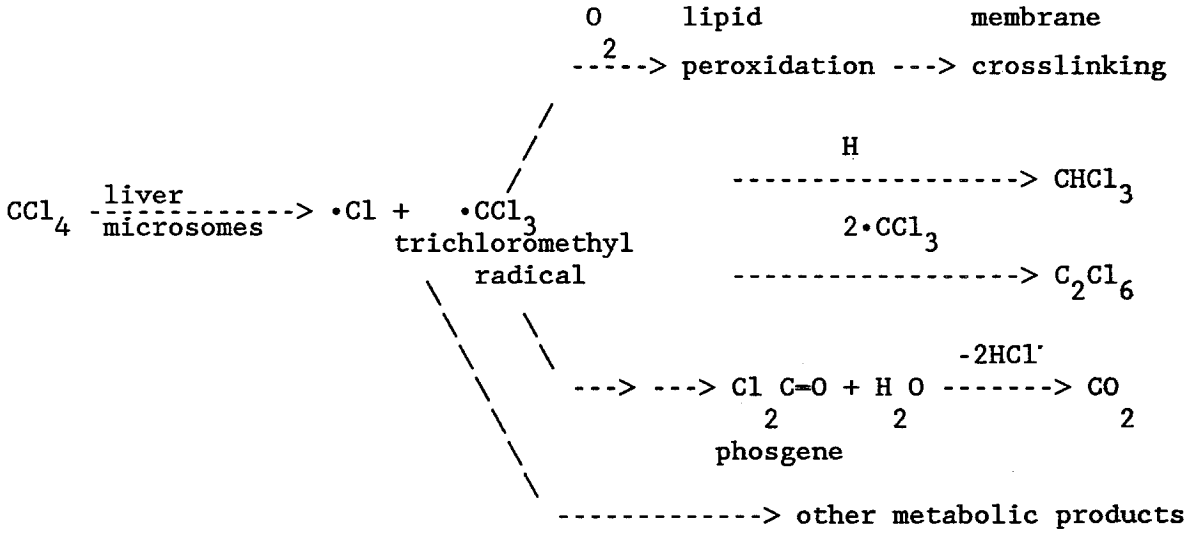
Essentially 100% of an oral dose is absorbed. Gastrointestinal absorption of CTC (3 ml) was studied by Robbins (1929), who demonstrated that it was readily absorbed from the small intestine and that the rate was enhanced by alcohol and fats. The oral absorption in rats was subsequently studied by Recknagel and Litteria (1960), who determined that peak blood concentration occurred 1.5 hours following administration.

2.3 Distribution

The distribution of carbon tetrachloride in animals varies with the route of administration, concentration, and the duration of exposure (Von Oettingen 1964); however, as would be predicted from CTC's solubility properties, most of the compound accumulates in tissues with high fat content, such as adipose tissue, liver and bone marrow (Robbins 1929, McCollister et al. 1950, 1951). Fowler (1969) examined the distribution and metabolism of CTC following oral administration to rabbits of 1 ml/kg. The highest concentrations of CTC were found in the fat, followed by the liver, kidney and muscle. CTC metabolites (i.e. chloroform and hexachloroethane) were also detected in fat, liver, kidney and muscle. In a study on CTC accumulation, adipose tissue concentrations appeared to reach a steady state CTC concentration after one week of repeated 3-hour exposures of rats to 10, 50, or 100 ppm CTC (Shimizu et al. 1973). Following a two-week exposure of rats to 100 ppm $^{14}CCl_4$, Paustenbach et al. (1986a) found that the fat, liver, adrenals and lung contained the highest concentrations of $^{14}CCl_4$.

2.4 Metabolism

The metabolism of CTC has been investigated in the rat, rabbit, dog and human. Close to half of the absorbed CTC is excreted unchanged, but the remainder is metabolized to carbon dioxide, chloroform, hexachloroethane, urea, carbonate and a number of unidentified substances present in tissues, in feces or in urine. Carbonyl chloride (phosgene) is also a postulated metabolite. The metabolic products are thought to be produced via the formation of a trichloromethyl radical. A basic metabolic scheme has been postulated as follows:



Paustenbach et al. (1986b) exposed rats to 100 ppm $^{14}CCl_4$ for 1 to 2 weeks. They found that 1 to 2 % of the CCl_4 was present as CO_2 in the expired air. The rate of elimination of $^{14}CO_2$ was slower than that of $^{14}CCl_4$ in the expired air. The investigators concluded that the late appearance of $^{14}CO_2$ was probably due to stored $^{14}CCl_4$, which was slowly released from the fat

and then metabolized. Radiolabeled compound in the feces was not identified, but is presumed to be a metabolite.

McCollister et al. (1950, 1951) exposed monkeys to radiolabeled CTC vapor by inhalation. An estimated 40% of the absorbed material was exhaled unchanged, while 11% was exhaled as carbon dioxide. In the blood, a number of unidentified radiolabeled materials were isolated and classified as "alkaline volatiles," "acid volatiles," or as "non-volatiles." In the urine, some of the labeled carbon was in the form of urea and carbonate, but 95% was a nonvolatile, unidentified compound.

The production of chloroform following administration of CTC was demonstrated in the rat (Ahr et al. 1980), in the rabbit (Fowler 1969) and in the dog (Butler 1961). Chloroform production from CTC was also demonstrated <u>in vitro</u>, using mouse tissue homogenates (Butler 1961). Fowler (1969) identified hexachloroethane, which was assumed to have formed from the dimerization of the trichloromethyl radical. Reynolds et al. (1984) measured CTC, chloroform and CO_2 exhalation following oral $^{14}CCl_4$ administration (0.1 to 26 mmoles/kg). They reported that as the dose was increased, the proportions of CO_2, chloroform and CTC changed: $^{14}CO_2$ declined from 28 to 0.7 %, chloroform levels remained under 1 %, and expired $^{14}CCl_4$ levels increased from 19 to 89%. This suggests that saturation of CTC metabolism had occurred. The authors suggested that the decrease in CO_2 production, along with an increase in hepatotoxicity, is

consistent with the presumption that the trichloromethyl radical reacts with other biomolecules when the CO_2 pathway is saturated.

Durk and Frank (1984) showed that CTC metabolism increased when the oxygen partial pressure was lowered. This resulted in an increase in the amount of lipid peroxidation as measured by the exhalation of ethane and pentane. However, the total time course of CTC metabolism was limited due to the concomitant destruction of cytochrome P-450. It has also been demonstrated that hyperbaric O_2 treatment alters CTC metabolism as measured by a decrease in the conversion of CCl_4 to CO_2 and $CHCl_3$ (Burk et al. 1986). Thus, the metabolism of CTC and the production of various metabolites appear to be highly dependent on the experimental conditions.

The mechanism of carbon tetrachloride hepatotoxicity has been extensively studied and reviewed (Ahr et al. 1980, Castro and Diaz Gomez 1972, Comporti 1985, Farber 1985, Ray and Moore 1986, Recknagel and Glende 1973, Recknagel 1983, Shah et al. 1979, Slater 1966, Slater et al. 1985, Smith and Sandy 1985, Smuckler and James 1984, Yalcin et al. 1986). The general view is that CTC may act via the formation of a trichloromethyl radical intermediate ($\cdot CCl_3$) following loss of a chlorine atom (Butler 1961, Kubic and Anders 1981, Sagai and Tappel 1982, Gee et al. 1981, Link et al. 1984). The irreversible incorporation of CTC into total lipids and phospholipids was shown in liver, kidney, lung, brain and other tissues (Ciccoli et al. 1978). The binding of CTC to lipid and protein has been shown to occur <u>in vivo</u> (Castro and Diaz Gomez 1972, Ciccoli et al. 1978).

Several studies have suggested the stimulation of lipid peroxidation as a possible mechanism of toxicity of CTC (see Slater et al. 1985). Link et al. (1984) isolated and identified a group of monomeric trichloromethyl fatty acid residues. They suggested that the binding of trichloromethyl radicals to lipids may result in membrane lipid cross-linking, which could ultimately disrupt cellular function. Yalcin et al. (1986) reported that CTC injections caused significant increases in hepatic lipid peroxide levels and significant decreases in glutathione peroxidase activity, glutathione transferase activity and hepatic glutathione levels. The reports of lipid peroxidation do not preclude the trichloromethyl radical from also binding with other biological molecules, such as proteins, to initiate biochemical toxicity.

Other studies have investigated the disturbance of Ca^{++} homeostasis as a possible mechanism for CTC hepatotoxicity (Moore and Ray 1983, Ray and Moore 1986, Recknagel 1983, Smith and Sandy 1985). Intracellular calcium releases may initiate hepatotoxic changes (Ray and Moore 1986). In addition to the above evidence for binding of carbon tetrachloride metabolites to proteins and lipids, there is also evidence of binding to DNA; this is discussed in the section on genotoxicity.

2.5 Elimination

Following inhalation, ingestion, or injection, unmetabolized CTC is predominantly excreted via the lungs (McCollister et al. 1951, Reynolds et al. 1984, Robbins 1929). Excretion of CTC appears to be biphasic and the second phase is relatively slow; thus, accumulation of CTC with repeated

exposure can result in chronic intoxication. Monkeys exhaled CTC for 29 days after exposure (McCollister et al. 1950, 1951). Studies based on rats would tend to underestimate the tendency of CCl_4 to accumulate in humans since rats eliminate CCl_4 faster (Paustenbach et al. 1986a, Stewart et al. 1961).

The predominant route for elimination of CTC metabolites appears to be the feces. In rats this represented 32 to 62% of the total CTC dose (Paustenbach et al. 1986b). However, the substances containing the radioactivity detected in the feces have not been identified and are only presumed to represent metabolic end products. In contrast, elimination in the urine represented 4 to 8% of the excreted dose, while elimination as CO_2 in the exhaled air was approximately 2% of the total dose. The remainder of the dose (32 to 59%) was exhaled as unchanged CTC. In an earlier study, when CTC was administered by injection to rats, 85% of CTC was exhaled unchanged in 18 hours (Paul and Rubenstein 1963).

The percent of excretion by exhalation has not been quantified in human studies (Stewart and Dodd 1964, Stewart et al. 1961, Lehmann and Schmidt-Kehl 1936). In the Stewart et al. (1961) experiments, individuals inhaled CTC (11 or 49 ppm) for up to 180 minutes. CTC was detected (0.3 ppm) in the expired air up to 5.5 hours post-exposure. However, the limit of detection was only 0.1 ppm and it was not stated whether samples were taken after 5.5 hours.

3.0 ACUTE TOXICITY

The narcotic effects of CTC were first reported in 1867 (Sansom) and descriptions of its toxicity appeared in the early 1900's (Lehmann 1911). Over 611 cases of acute CTC poisoning, many of which were fatal, have been reported in the literature (Beattie et al. 1944, Bjarnason et al. 1968, Dupont et al. 1975, Ruprah et al. 1985, Von Oettingen 1964). The NOAEL (no observed adverse effect level) for a 3-hour acute exposure of humans is 10 ppm (Stewart et al. 1961). This concentration is at least 4 orders of magnitude greater than expected ambient levels, providing enough of a margin of safety so that the acute toxic effects would not be expected to occur in the general population.

3.1 Local effects

Applied to the skin, CTC causes reddening, blistering, inflammation and pain. Ingested orally it can irritate mucous membranes, produce a burning sensation and stimulate peristalsis. Exposure to the vapor can produce irritation of the eyes, nose and throat. The NOAEL for irritation of mucous membranes for humans is 49 ppm for a 70-minute exposure (Stewart et al. 1961).

3.2 Systemic effects

CTC produces acute systemic toxicity following ingestion or inhalation, including inhalation of vapors in a poorly ventilated area. The major effects are nervous system depression, hepatic damage, and renal tubular

destruction. Symptoms of toxicity may appear after a delay of two to three days. Pulmonary toxicity has been reported (Anttinen et al. 1985, Boyd et al. 1980). Hemolysis and other circulatory disturbances have been observed (Schulze and Kappus 1980, Von Oettingen 1964).

The CNS symptoms following exposure to CTC are nausea, vomiting, headache and/or dizziness. In severe cases, CTC may produce vertigo, mental confusion, incoordination, stupor, convulsions, coma or death. Suppression of the medullary centers may result in vasodilation and a fall in blood pressure.

CTC is hepatotoxic in animals (Adams et al. 1952) and in humans (Dupont et al. 1975), and the effects appear rapidly. In humans, alterations in lipid metabolism in the liver may be observed 30 minutes following administration. Histological changes may be observed within one hour. Within 24 hours a characteristic centrilobular necrosis of the liver is present. Early signs of injury may appear as altered enzymatic levels, such as increased serum glutamic oxaloacetic transaminase (SGOT), or enlargement of the liver (Beattie et al. 1944). Relative to lethality, hepatotoxicity is an extremely sensitive endpoint. In acute exposures, hepatotoxicity (median toxic dose) has been reported at concentrations 1/230 to 1/280 of the median lethal dose (Klaasen and Plaa 1966, Lundberg et al. 1986).

The kidney is a major target of CTC toxicity. Necrosis and tubular degeneration have been observed in laboratory animals (Chandler and Chopra 1925). In mild poisoning incidents in man, CTC can produce reversible

oliguria for several days (Dudley 1935 a.b.). In severe cases, there may be complete anuria, hypertension, acidosis and pulmonary edema.

In humans, 317 ppm is an estimated toxic concentration for a 30-minute exposure (NIOSH 1984). A 70-minute exposure of 6 individuals to 49 ppm altered normal metabolism (depressed serum iron and elevated urinary urobilinogen) of three of the subjects (Stewart et al. 1961). In the same study, no effects were observed following an exposure to 10 ppm for 180 minutes. Thus, based on information for humans, a NOAEL for acute exposures up to 3 hours is 10 ppm.

The severity of effects from CTC depends more on the concentration inhaled than on the length of exposure. Using rats, experiments examining the relative effects of concentration versus length of time, investigators found higher concentrations over shorter exposures produced more toxic effects than lower concentrations over a longer period (David et al. 1981, Uemitsu et al. 1985).

4.0 SUBCHRONIC AND CHRONIC TOXICITY

Toxic effects, other than genotoxicity, reproductive effects and carcinogenicity, following both subchronic and chronic exposures to CTC are discussed in this section. Chronic toxicity has been observed following dermal (animal studies), oral (animal studies) and inhalation exposure (animal studies and human cases). The slow excretion rate of CTC results in its accumulation, increasing the potential for toxicity following repeated exposure. Subchronic and chronic exposures affect the same target tissues as acute exposure: the nervous system, the liver and the kidney (Higgins and Stasney 1936, Lehmann and Schmidt-Kehl 1936, Cameron and Karunaratne 1936, Smyth et al. 1936, Edwards 1941, Edwards and Dalton 1942, McCord 1946, Prendergast et al. 1967, Kanics and Rubenstein 1968, Shimizu et al. 1973, Merkur'eva et al. 1979).

Toxic effects have been reported following chronic inhalation exposure to 5 ppm or greater. The longest animal study reported lasted 10 1/2 months. A NOAEL based on histopathologic and gross toxicity for a prolonged exposure of the rat is 1 to 5 ppm (depending on the study, Prendergast et al. 1967, Adams et al. 1952). NOAEL's for other species have not been determined, but are below 1 ppm. There appears to be a reasonable margin of safety to expect that histopathologic or gross toxicity would not occur in the general population since current ambient levels are 4 orders of magnitude below the LOEL (low effect level). However, a recent gavage study examining biochemical markers of hepatotoxicity indicates that CTC has a very shallow dose-response curve, spanning over 2 orders of magnitude. Consequently, although it is unlikely that any biochemical liver changes would occur from

exposure to current ambient levels, animal studies have not been conducted that establish NOAEL for this endpoint.

4.1 Animal Studies

4.1.1 Subchronic

A subchronic inhalation study was conducted on carbon tetrachloride by Prendergast et al. (1967) using the rat, guinea pig, rabbit, dog and monkey (see Table 1). The most susceptible species in terms of mortality was the guinea pig; twenty percent of the animals died during the 515 mg/m^3 exposure (≈ 80 ppm). Weight loss was reported in all species except the rat. Severe liver damage was observed in rats, guinea pigs and monkeys following 24 hour/day exposures to 61 mg/m^3 (≈ 10 ppm) for 90 days. At the 6.1 mg/m^3 (≈1 ppm) exposure, slight growth depression and histopathological changes were observed in all species except the rat. Thus, from this study for subchronic exposures the NOAEL for the rat would be 1 ppm, but the NOAEL for rabbit, dog and monkey would be below 1 ppm. It is important to note that the animals in this study were more sensitive to CTC than those animals in the chronic studies (See below). Consequently, in the present document, the NOAEL for chronic exposure is primarily based on the results of this subchronic study.

Hayes et al. (1986) conducted a 90-day gavage exposure of CD-1 mice (20 animals of each sex per dose group) to 12, 120, 540 or 1200 mg/kg in corn oil. There were no effects on mortality, body weight, hematological endpoints or urinalysis endpoints. There were significant increases in

TABLE 1. SUMMARY OF DESIGN OF SUBCHRONIC AND CHRONIC INHALATION STUDIES

Study	Species	(N)[1]	Concentration (ppm)	Exposure Design	Length	Effects Observed
Smyth et al. 1936	rat	24	50,100,200,400	8h/d, 5d/w	10.5m[2]	Sciatic nerve damage, cirrhosis above 50 ppm
	guinea pig	24	50,100,200,400	"	"	High mortality, nerve damage.
	guinea pig[3]	16	25,50,100,200	"	"	Mortality.
	monkey	4	50,200	"	"	Fatty infiltration, sc nerve damage at 200.
Adams et al. 1952	rat	30[4]	100,200,400	7h/d, 5d/w	37w	Mortality, liver and kidney (> 100) patholo
	rat	30[4]	25,50	"	"	Fatty degeneration.
	rat	40[4]	10	7h/d, 5d/w	27w	Fatty degeneration.
	rat	48	5	7h/d, 5d/w	29w	No effects observed.
	guinea pig	10[4]	25	"	26w	Decreased growth, cirr
	rabbit	4[4]	25,50,100	"	36w	Cirrhosis.
	rabbit	4[4]	10	"	"	No effects observed.
	monkey	2	100	"	40w	Fatty degeneration.
	monkey	2	50	"	"	Decreased growth.
	monkey	1	25	"	30w	No effects observed.

TABLE 1. (cont.) SUMMARY OF DESIGN OF SUBCHRONIC AND CHRONIC INHALATION STUDIES

Study	Species	(N)[1]	Concentration (ppm)	Exposure Design	Length	Effects Observed
Prendergast et al. 1963	rat	15	≈80	8h/d, 5d/w	6w	Liver and lung pathology.
	guinea pig	15	≈80	"	"	Mortality, weight loss, liver and lung pathology.
	rabbit	3	≈80	"	"	Weight loss, liver and lung pathology.
	dog	2	≈80	"	"	Weight loss, liver and lung pathology.
	monkey	3	≈80	"	"	Mortality, weight loss, liver and lung pathology.
	rat	15	≈10	continuous	90d	Depressed growth, enlarged liver, fatty infiltration.
	rat	15	≈1	continuous	90d	Lung inflammatory changes.
	guinea pig	15	≈10	"	"	Mortality, depressed growth, enlarged liver, fatty infiltration.
	guinea pig	15	≈1	"	"	Decreased weight gain, lung inflammatory changes.
	rabbit	2	≈10	"	"	Depressed growth, enlarged liver.
	rabbit	3	≈1	"	"	Lung inflammatory changes, decreased weight gain.
	dog	2	≈10	"	"	Depressed growth, histological liver changes.
	dog	2	≈1	"	"	Lung inflammatory changes, decreased weight gain.
	monkey	3	≈10	"	"	Emaciated appearance, loss of hair.
	monkey	3	≈1	"	"	Decreased weight gain.

[1] N refers to the number of animals per dose group.
[2] h refers to hours, d to days, w to weeks, m to months
[3] This group of guinea pigs had 1.2g of calcium lactate added to daily diet.
[4] Half of the animals were male.

serum lactate dehydrogenase (LDH), serum glutamic-pyruvic transaminase (SGPT), and serum glutamic-oxaloacetic transaminase (SGOT) at all dose levels relative to controls. The serum glucose was decreased at all dose levels relative to controls. There was a significant increase in liver weight and spleen weight at all dose levels relative to control animals. At levels of 120 mg/kg and above, animals exhibited a significant increase in thymus weight. Hepatotoxicity was evident at all dose levels and was reported to be dose-dependent. Although the study used a 100-fold dose range with no observed mortality, a NOAEL was not obtained.

A 90-day subchronic oral toxicity study of CTC was conducted by Bruckner et al. (1986). Rats (15 to 16 per group) were gavaged daily with 0, 1, 10, or 30 mg/kg CTC in corn oil. Administration of 1 mg/kg did not affect clinical chemistry indices, weight gain, tissue weights or liver morphology. The 10 mg/kg dose level produced mild centrilobular vacuolization, but no evidence of necrosis, fibrosis or other serious degenerative changes. Animals in the 30 mg/kg dose group had increased sorbitol dehydrogenase (SDH), ornithine-carbamyl transferase (OCT) and glutamic-pyruvic transaminase (GPT) levels. They exhibited a decrease in weight gain and an increase in liver weight to body weight ratios. The livers had extensive degenerative lesions, periportal fibrosis, bile duct hyperplasia and some hyperplastic nodules. The authors concluded that 1 mg/kg represented a NOAEL for rats, 10 mg/kg represented a LOAEL, and that, although cirrhosis and hyperplastic nodules are commonly seen in livers of animals with hepatic tumors, the existence of a causal relationship is unclear.

To compare the relative toxicity of the inhalation versus the oral route, the results of the 90-day subchronic studies in rats of Prendergast et al. (1967) and Bruckner et al. (1986) can be compared. In the Prendergast et al. (1967) study a NOAEL was observed at 6.1 mg/m^3 (1 ppm). Assuming that 50% of the inhaled dose is absorbed and that a rat breaths 0.144 m^3 of air per day, the daily dose of CTC is: (0.5) (6.1 mg/m^3) (0.144 m^3/day) ÷ 0.350 kg rat = 1.25 mg/kg per day. This value is comparable to the 1 mg/kg per day NOAEL observed in the Bruckner et al. (1986) study.

4.1.2 Chronic

The first major study on chronic exposure to carbon tetrachloride was conducted by Smyth et al. (1936) as summarized in Table 1. They also examined workers exposed to CTC (discussed below). Extensive mortality occurred in the guinea pigs, such that only the 25 and 50 ppm groups survived an average of 40 or more exposures. Only two rats succumbed from CTC at the 400 ppm exposure. Growth retardation was reported in guinea pigs (25 ppm), in rats (400 ppm) and in monkeys (200 ppm). The rats (100 ppm and above) and guinea pigs (50 ppm and above) exhibited liver cirrhosis while the monkeys showed signs of fatty degeneration. Kidney damage was also reported in the rats and guinea pigs. Many of the guinea pigs and monkeys exposed to 200 and 400 ppm exhibited sciatic nerve damage, while rats exhibited it at all exposure levels.

Adams et al. (1952) studied rats, rabbits, guinea pigs and a few rhesus monkeys (see Table 1). Survival rates at 100 ppm and above were 50% or less for the rats and guinea pigs. Although precise information is not provided, apparently animal mortality was observed at exposure levels above 50 ppm for rats and

guinea pigs. Four or fewer animals were used in the rabbit and monkey dose groups preventing clear evaluation of the responses. They found that chronic exposure of guinea pigs, rats and rabbits (at 100 ppm or greater) produced growth depression, increased liver weights, centrilobular fatty degeneration and cirrhosis of the liver, and degeneration of the tubular epithelium of the kidneys. Rats exhibited hepatic effects above the 5 ppm exposure. The guinea pigs reportedly had a statistically significant increase in liver weight at the 5 ppm level. For the monkeys, growth depression (50 and 100 ppm) and hepatic toxicity (100 ppm) were observed. The NOAEL is reported in rats to be 5 ppm for the 7 hour/day exposure. The NOAEL for the guinea pigs was not attained in this study, but would be below 5 ppm. The NOAELs for rabbits and monkeys cannot be determined in this study due to the few animals tested.

4.2 Human Cases

Signs and symptoms of chronic CTC poisoning include fatigue, headache, anxiety, giddiness, muscular twitching, jaundice, hypoglycemia, lack of appetite, nausea, diarrhea, dull pain in the kidney region, dysuria, proteinuria, blurred vision, and eye irritation. Studies of exposed workers have reported the presence of gastrointestinal/hepatic disturbances, cirrhosis, aplastic anemia, and neurological disturbances (Browning 1965, Kazantzis and Bomford 1960, McDermott and Hardy 1963, Stewart and Witts 1944, Straus 1954).

NIOSH has recommended a time-weighted average (TWA) occupational standard of 2 ppm (10-hour workday, 40-hour workweek with air sample taken over a period not to exceed 1-hour duration) based on reports of liver and eye changes in workers chronically exposed and animal studies (NIOSH 1975, 1984). They stated that

this concentration is expected to be low enough to prevent chronic liver injury in humans. The American Conference of Government Industrial Hygienists (ACGIH) recommended a 5 ppm TWA threshold limit value (TLV) based on exhibition of fatty infiltration in chronically exposed laboratory animals to 10 ppm CTC (ACGIH 1984). The current CAL-OSHA standard for CTC is also 5 ppm.

Based on the highest average ambient concentration reported by ARB, current ambient levels are three orders of magnitude below the current occupational standards.

5.0 GENOTOXICITY

Radiolabeled $^{14}CCl_4$ has been shown to bind covalently to macromolecules, including DNA, in vitro and in vivo. Most research has shown that it is first metabolized to the trichloromethyl radical, possibly at the nuclear membrane, prior to alkylation. Almost all bacterial mutagenicity tests have been negative. A weak positive response was reported in yeast. Negative responses were reported in an in vitro study using a rat liver epithelial cell line. Negative or weak responses were observed in four studies examining unscheduled DNA synthesis. CTC has been shown to be a strong inducer of chromosomal rearrangements. Based on these results, DHS staff have concluded that carbon tetrachloride has genotoxic potential.

5.1 Covalent Binding to DNA

As indicated in Section 2.1, much of the carbon tetrachloride absorbed by any route is exhaled unchanged; however, some is apparently metabolized to form trichloromethyl radical. This highly reactive intermediate has been found to covalently bind with macromolecules. Furthermore, metabolically activated carbon tetrachloride was found to bind with DNA in vivo (Diaz Gomez and Castro 1980a, Rocchi et al. 1973) and in vitro (Diaz Gomez and Castro 1980a, Direnzo et al. 1982, Levy and Brabec 1984, Rocchi et al. 1973). The in vivo test species were Swiss and A/J mouse strains, and Wistar and Sprague-Dawley rats. The in vitro systems included rat mitochondrial DNA, mouse liver DNA, and calf thymus DNA. An in vitro study (Diaz Gomez and Castro 1981) indicated that the trichloromethyl radical interacted with all four DNA bases, but bound preferentially to guanine and

adenine. Consequently, carbon tetrachloride could produce a genotoxic response following metabolic activation.

5.2 Mutagenicity test systems

The ability of CTC to produce a mutagenic response has been examined using several test systems. The sole positive result was reported in an abstract by Cooper and Witmer (1982), who observed that Salmonella strain TA100 exhibited a weak mutagenic response under low oxygen tension, with rabbit liver S9 as the activation system. Negative reponses were reported with strains TA97, TA98, TA100, TA102, TA1535, TA1537, TA1538, TA1950 using rat liver S9 for metabolic activation (Barber et al. 1981, Braun and Schoneich 1975, De Flora et al. 1984, Simmon et al. 1977, Uehleke et al. 1977).

EPA considered these negative results inconclusive because the rat liver S9 could be an inadequate activation system for CTC; there could have been scavenging of reactive intermediates by microsomal protein or lipid; or there may have been evaporation of CTC from the test systems (EPA 1984). Other halogenated hydrocarbons as a class have reportedly produced false negatives in the Ames Salmonella assay (McCann and Ames, 1976). The bacterial test systems may not be appropriate for testing the mutagenic response of CTC because of the lack of a nuclear membrane. Nuclear protein fractionation studies using rat liver (Diaz Gomez and Castro 1980b) indicate that metabolic activation of CTC occurs preferentially in the nuclear membranes, providing the reactive intermediates access to DNA. Thus, the bacterial test system may not be an appropriate mutagenicity model.

CTC was analyzed for its mutagenic potential in yeast (Callen et al. 1980). The D7 strain of Saccharomyces cerevisiae yeast was examined for gene conversion at the trp-5 locus, mitotic crossing over at the ade-2 locus, and gene reversion at the ilv-1 locus. The yeast strain contained an endogenous cytochrome P-450 dependent monooxygenase activation system, thus avoiding a number of the pitfalls cited in the bacterial studies. The authors concluded that CTC induced mutations in yeast. However, since positive mutagenic responses were observed only under conditions of extreme (90%) cytotoxicity, DHS staff believes that these results are unreliable.

5.3 Chromosomal Effects

Gualandi (1984) examined the ability of CTC to induce gene mutations and chromosomal rearrangements in a diploid strain of Aspergillus nidulans. When assayed for mutagenicity, CTC produced negative results in the plate incorporation assay and produced a 2- to 3-fold increase of suppressor mutants in the growth-mediated assay. In contrast, CTC was shown to be a strong inducer of chromosomal rearrangements compared to controls. This study indicates, that although CTC lacked strong mutagenic activity in the assay, it was genotoxic by virtue of its induction of chromosomal rearrangements.

In a study using an epithelial-type cell line derived from rat liver, CTC did not produce chromatid or chromosomal aberrations (Dean and Hodson-Walker 1979). In this test system, CTC was not observed to be mutagenic, but this may have been due to the high cytotoxicity of CTC to the cell line used. The exposure concentrations used in the chromosome assay were based on a

predetermined "growth inhibiting dose." Thus the dose levels for CTC were very low. For example, the CTC doses (0.005 to 0.02 μg/ml) were less than 1/2500 of the dose levels of other substances examined in the test series: 2-acetylaminofluorene, propylene oxide and cyclophosphamide.

5.4 Other Test Systems

Amacher and Zelljadt (1983) tested CTC's ability to produce in vitro morphological transformation of Syrian hamster embryo (SHE) cells. CTC produced a weakly positive response as indicated by the transformation of one to three of the test colonies. No transformed colonies were observed in the solvent controls. These results are consistent with other data suggesting that CTC is potentially genotoxic.

Sina et al. (1983) developed an alkaline elution rat hepatocyte assay to measure DNA single-strand breaks (SSBs). In the test system, CTC produced a 3- to 5-fold greater number of SSBs than the controls; a positive response suggesting potential genotoxicity of CTC. The authors concluded that the test system correlates well (85 to 92%) with mutagenic and carcinogenic activity for the 91 compounds tested.

In a study by Brambilla et al. (1986), CTC was used as an agent to stimulate rapid hepatic growth for testing effects of 2-acetylaminofluorene (2-AAF). However, administration of CTC alone increased the level of DNA fragmentation. Furthermore, the concurrent administration of 2-AAF and CTC produced more than an additive effect on DNA fragmentation. This study indicates that CTC may affect DNA stability.

CTC produced a positive genotoxic response in a test system of De Flora et al. (1984). CTC was assayed in a DNA-repair test with E.Coli strains proficient and deficient in DNA repair. The genotoxic effect was ascertained by increased killing or growth-inhibition of bacteria lacking specific DNA-repair mechanisms, compared with the isogeneic repair-proficient strains. Although the test indicates genotoxicity it does not assay for mutagenicity.

CTC produced negative results in tests for unscheduled DNA synthesis (UDS). Mirsalis et al. (1980 and 1982) treated F-344 rats in vivo with CTC and reported no increase in measured UDS in vitro using primary hepatocyte cultures. Although some known hepatocarcinogens (dimethylnitrosamine and acetylaminofluorene) were positive in the test system, other known mutagens (benzo[a]pyrene and 7,12-dimethyl benz[a]anthracene) tested negative. In another in vivo study (Craddock and Henderson 1978) rat hepatocyte nuclei were examined for induction or changes in de novo replication. A positive effect was reported at 17 hours after treatment; the authors felt that the latency indicated that the repair replication was a secondary effect and not a direct reaction with DNA. Perocco and Prodi (1981) examined the effect of CTC on scheduled and unscheduled DNA synthesis in vitro using human lymphocytes. They reported that CTC inhibited scheduled DNA synthesis (SDS) but produced low values of UDS.

In summary, carbon tetrachloride appears to have genotoxic potential based on its ability to form reactive intermediates that can covalently bind to DNA, to induce chromosomal rearrangements in vitro, to cause SSBs and to produce morphological transformation of SHE cells. CTC has demonstrated

very little, if any, mutagenic activity based on the standard bacterial mutagenic assays, a yeast assay, and determinations using unscheduled DNA synthesis. However, the negative mutagenicity test results may be a result of the inadequacy of the activation systems of some of the assays, the inappropriateness of using a bacterial test system as a model and the high cytotoxicity of CTC in the test systems. The absence of positive results in the standard mutagenicity assays indicates that although CTC is potentially genotoxic, it probably does not induce point mutations.

6.0 REPRODUCTIVE TOXICITY

Inhaled carbon tetrachloride causes degenerative changes in the testes and reduced testicular weight in laboratory animals. In one study CTC reduced fertility in rats (Smyth et al. 1936). In pregnant female rats exposed to concentrations of 300 ppm, CTC crossed the placenta and produced developmental abnormalities in the fetus. The teratogenic potential of CTC has not been adequately tested.

The NOAEL for reproductive effects caused in rats is 100 ppm (Adams et al. 1952). This level is nearly six orders of magnitude above the highest recorded ambient levels of CTC; therefore, reproductive effects are not expected to occur in the general population due to exposure to CTC.

6.1 Male Reproductive Effects

A number of studies demonstrate decreased weight in testes and accessory reproductive organs or other pathological changes. The most sensitive study that considered male reproductive effects was by Adams et al. (1952). Rats and guinea pigs were repeatedly exposed to CTC ranging from 5 to 400 ppm (See Table 1 for details). At 200 ppm and above rats exhibited decreased weight of the testes compared to controls and germinal elements of the testes showed moderate to marked degeneration, with some seminiferous tubules exhibiting complete atrophy of germinal elements. At 400 ppm guinea pigs exhibited some minor nonspecific pathological changes in the testes. No decrease in testicular weight or histological effects were observed in rats at concentrations of 100 ppm or below, or in guinea pigs at concentrations of 200

ppm or below. Rabbits and monkeys were also studied by Adams et al. (1952), but there were only one or two animals in each dose group so the numbers were too small to draw reliable conclusions.

Chatterjee (1966) and Kalla and Bansal (1975) reported similar male reproductive effects caused by CTC in rats. In both studies 4800 mg/kg CTC were administered intraperitoneally for 10 to 20 days. The relative weights of the testes and accessory reproductive organs were decreased compared with controls. Histological examination indicated testicular atrophy, disruption of normal architecture, and marked abnormalities in spermatogenesis, including azoospermia. Both studies reported an increase in pituitary weight; however, the relevance of this finding cannot be determined without actual measurement of serum gonadotrophins. The route of administration (intraperitoneal instead of inhalation) and high dose make the interpretation of these results difficult.

These three studies indicate that CTC can produce adverse male reproductive effects. A similar decrease in testicular weight was observed in two species by two exposure routes. Although the results indicate a decline in the spermatogenic process, only one investigator has tested the reproductive ability of affected animals (Smyth et al. 1936, described below).

6.2 Study on Fertility

In a multigeneration study, Smyth et al. (1936) found that 200 and 400 ppm CTC diminished the number of litters and decreased the number of offspring per litter in rats, compared to controls. Up to 3 generations were observed for fertility following repeated 8-hour/day, 5-day/week exposure of both sexes to 50, 100, 200 and 400 ppm CTC over 10.5 months. It was not clear if the decline in fertility resulted from effects of CTC on males, females, or both.

6.3 Placental Transport

Dowty and Laseter (1976) reported that CTC (as well as other halogenated hydrocarbons) can cross the human placenta and accumulate in the fetus. Blood samples were recovered from the umbilical cord and from paired maternal blood of 11 women after vaginal delivery. The authors indicated that exposure to CTC and other halogenated hydrocarbons may have occurred in drinking water. Carbon tetrachloride levels were higher in cord blood than in the maternal blood. In animal studies, CTC was shown to cross the placenta of pregnant rats and to produce fetotoxicity (Sundareson 1942, Bhatacharyya 1965, Schwetz 1974). Maternal toxicity was produced in these studies, which could increase fetal toxicity. However, several investigators have shown that for CTC there does not appear to be a direct correlation between the severity of maternal toxicity and the severity of reproductive effects in the rat fetus (Wilson 1954, Schwetz et al. 1974). Sundareson (1942) reported that direct injections of CTC into fetuses in utero produced maternal toxicity, indicating that CTC can traverse the placenta in either direction.

6.4 Developmental Toxicity

Several studies have suggested that CTC is embryotoxic in rats; these are summarized in Table 2. The presence or absence of maternal toxicity is also indicated. Administration of CTC to pregnant rats prior to the 12th day of gestation produced a failure to implant or increased intrauterine mortality (Sundareson 1942). CTC produced a decrease in the viability and in the number of pups per litter when compared to controls (Gilman 1971). In rabbits, CTC administered on days 4 and 5 of gestation produced cellular degeneration in the embryonic discs, and the trophoblasts contained very large nuclei with prominent nucleoli (Adams et al. 1961). The latter study indicates that CTC can produce embryotoxicity prior to implantation.

CTC is fetotoxic in rodents. When administered after the 12th day of gestation CTC was associated with premature delivery, increased postnatal mortality, and liver degeneration and necrosis in the fetus (Sundareson 1942, Bhattacharyya 1965). Schwetz et al. (1974) reported that CTC inhalation by pregnant rats produced a significant decrease in fetal body weight and crown-rump length when compared to controls. Furthermore, CTC can diffuse into mother's milk and cause liver damage in the nursing neonate (Bhattacharyya 1965).

Two studies reported the absence of teratogenic effects. In a preliminary inhalation study, Gilman (1971) reported no teratogenic effects were observed in the offspring. Schwetz et al. (1974) also reported the absence of

TABLE 2. SUMMARY OF STUDIES ON DEVELOPMENTAL TOXICITY

Study	Species	(N)[2]	Dose	Day of Gestation Exposed	Fetal Effects	Maternal Toxicity[1]
Briese 1938	rat	60	nr	1-21	Anemia	yes
Sundareson 1942	rat	1-22	200-2400 mg/kg	19-20	Death, liver degeneration	yes
Adams et al. 1961	rabbit	nr[3]	4800-8000 mg/kg	4-5	Embryonic degeneration	no
Bhatacharyya 1965	rat	nr	1600,3200 mg/kg	19-20	Liver necrosis	nr
Gilman 1971	rat	25	≈250 ppm 8 hours/day	10-15	Fetotoxicity	nr
Schwetz et al. 1974	rat	22	300 ppm 7 hours/day	6-15	Fetotoxicity	yes
	rat	23	1000 ppm 7 hours/day	6-15	Fetotoxicity	yes

[1] Maternal toxicity was evidenced by anemia, hepatotoxicity, reduced food consumption and increased SGPT activity.

[2] Number of female animals per dose group.

[3] Not reported.

teratogenicity in the rat as a result of CTC inhalation. However, significant increases in total skeletal anomalies (300 ppm), the presence of subcutaneous edema (300 ppm), and evidence of sternebral anomalies (1000 ppm) suggest that fetotoxicity rather than teratogenicity was associated with prenatal CTC exposure in the rat. The experiments cited above do not meet current EPA criteria for a test of teratogenic potential. EPA study design guidelines require that three dose levels be administered, that a nonrodent species be studied, and that positive controls be used (EPA 1984).

The studies on embryo- and fetotoxicity suggest that, in rodents, CTC exposure exhibited only limited potential for teratologic change. Additional studies in at least one more rodent and in a nonrodent species are needed prior to conducting an adequate human health risk assessment for prenatal and neonatal CTC exposure.

7.0 CARCINOGENICITY

Carbon tetrachloride has been shown to produce liver tumors in mice, rats and hamsters by the oral, subcutaneous and rectal routes. No inhalation studies have been conducted. These bioassays were not designed for use in a quantitative risk assessment: the studies were either preliminary in nature or CTC was administered to animals as a positive control; furthermore, there was high noncancer mortality in most of the experiments. Human case reports and epidemiological studies have not provided sufficient information to draw any inferences about a causal association between carbon tetrachloride exposure and cancer in humans. Consequently, CTC is considered to be an animal carcinogen and a potential human carcinogen. IARC's evaluations of carcinogenicity (1972, 1979) concluded that there was sufficient evidence that CTC was carcinogenic in experimental animals and that CTC should be regarded as a potential human carcinogen.

Mouse Studies

At least eight studies on chronic CTC administration have reported the development of hepatic tumors in six different strains of mice. Tumors have been reported in both males and females, by oral and rectal administration. The results of seven of these experiments are summarized in Table 3.

Edwards (1941) and Edwards and Dalton (1942) administered CTC by gavage to four different strains of mice (1 to 6 months of age) two to three times a week for 8 to 23 weeks. Both sexes were used in the study, but the olive

TABLE 3. MOUSE BIOASSAYS

Strain	Vehicle[1]	Dose (mg/kg)	Tumor Incidence % (N)[2]	Tumor	Reference
A m,f[3]	none	0	1 (200)	hepatoma	Edwards 1941
f	olive oil	≈2100	100 (54)	hepatoma	
A m	olive oil	0	0 (44)		Edwards & Dalton 1942
A f		≈260	71 (58)	hepatoma	
m,f		≈2100	98 (64)	"	
A m,f	olive oil	0	0 (28)[4]		Eschenbrenner and Miller 1943
		≈160	38 (63)	hepatoma	
		≈315	38 (63)	"	
		≈625	42 (59)	"	
		≈1250	53 (60)	"	
		≈2500	55 (60)	"	
A[5] m,f	olive oil	0	0 (10)		Eschenbrenner and Miller 1946
		≈40	0 (10)		
		≈80	70 (10)	hepatoma	
		≈159	63 (8)	"	
A[6] m,f	olive oil	0,≈10	0 (10)		Eschenbrenner and Miller 1946
		≈20	100 (10)	hepatoma	
		≈30	100 (10)	"	
		≈40	100 (9)	"	
B6C3F1	corn oil	0	4 (157)	hepatocellular carcinoma	NCI 1976 a,b, 1977
m,f		1250	100 (89)	" "	
		2500	97 (93)	" "	
C m	olive oil	0	0 (16)		Edwards & Dalton 1942
f		≈2100	83 (41)	hepatoma	
C3H m	none	0	0 (17)		Edwards 1941
	olive oil	≈2100	88 (143)	hepatoma	
C3H m	olive oil	0	4 (23)	hepatoma	Edwards & Dalton 1942
		≈2100	88 (143)	hepatoma	
L m,f	none	0	1 (152)	hepatoma	Edwards et al. 1942
	olive oil	≈2100	47 (73)	hepatoma	
Y m,f	none	0	2 (129)	hepatoma	Edwards & Dalton 1942
	olive oil	≈2100	60 (15)	hepatoma	

[1] All animals were administered CTC by gavage.
[2] N refers to the number of animals in the treatment group.
[3] m refers to male and f refers to female animals.
[4] Animals dosed at various intervals were combined; there were 5-12 per group.
[5] Administered 30 doses over 120 days.
[6] Administered 120 doses over 120 days.

oil vehicle was administered only to male control animals of strains A, C and CH3. The animals were 1 to 5 months old at the beginning of the study. One experiment in the study indicated the latency for tumor development in mice was 8 weeks. To assess the tumor-producing ability of CTC, animals were necropsied 12 to 21 weeks after the last treatment. For those animals exposed to approximately 2100 mg/kg of CTC the incidence of hepatoma was 88.2 percent; specific strain incidences are presented in Table 3. Whether the CTC-induced hepatomas were malignant was not established histologically in the study. The animals were dosed on a nondaily schedule for a maximum of 16 weeks and sacrificed as young as 4 months of age. Since tumor expression is a function of both dosage and the latency period, any risk assessment based on these studies, with their short observational periods, will underestimate the true carcinogenic risk. The data for strains A, C and CH3 (Edwards and Dalton 1942) were used in the DHS and EPA (1984) risk assessments of CTC. In another experiment Edwards and Dalton (1942) administered 1, 2 or 3 doses of carbon tetrachloride (≈260 to 2100 mg/kg) followed by long-term observation. The doses were hepatotoxic, but when the animals were examined 12 months later no tumors were observed. This experiment indicates that at these dose levels an acute exposure may not be tumorigenic in Strain A mice. Edwards et al. (1942) also reported CTC-induced tumors in an inbred L strain of mice, but these data were not used in the risk assessment since there were no vehicle controls, the dosing schedule was irregular and there was a relatively short exposure period relative to lifespan.

Eschenbrenner and Miller (1943, 1946) extensively examined tumor production in Strain A mice. In the first study they administered 30 doses of CTC at

intervals of one to five days. All animals were examined for tumors at 150 days following the first dose. Centrilobular liver necrosis was observed at all exposure levels. They reported that the incidence of hepatomas was increased as the time interval between doses increased. In Table 3 results for the different interval groups were combined. These data were not used in the risk assessment because the number of animals in the control groups was small (5 to 6), there was high animal mortality (39% of the controls), there was a short 30-day exposure period for animals and also a relatively short period of time prior to necropsy (21 weeks after first dose).

In a second study Eschenbrenner and Miller (1946) administered the same total quantity of CTC, either in 30 doses at four-day intervals or in 120 doses on consecutive days. This study was conducted to determine the effect of liver necrosis on tumor development; they found that mice receiving the smaller dose over 120 days (a so-called "non-necrotizing" dose) developed tumors at roughly the same or greater rate as those animals that received necrotizing doses (30 large doses at 4-day intervals). The 1943 study implied that a larger interval between doses increased tumor production. The 1946 study showed that the total length of the exposure period (i.e., 120 versus 30 days), not the time between doses, may have been the major determining factor in the production of tumors. There were too few animals in the study to determine a statistically significant effect, and consequently this study could not be used in a quantitative risk assessment. Other problems with using these data for risk assessment include the short exposure period, the short observation period to necropsy and the high mortality in all treated and control groups.

The 1946 study attempted to address the question of whether liver necrosis was a precondition of tumor production. The results indicated that massive liver necrosis was not a required precondition for the production of tumors with CTC. However, the study did not address the question of whether an hepatotoxic dose was a precondition for tumor production. Based on the work of Hayes et al. (1986), it is likely that hepatotoxic effects occurred in the animals dosed in the 1946 study. However, this does not answer the question of whether hepatotoxicity is a required precondition for tumor production by CTC. As indicated by Williams and Weisburger (1986) the carcinogenic mode of action of carbon tetrachloride remains to be clarified.

The three NCI mouse bioassays used carbon tetrachloride as a positive control (NCI 1976a,b, 1977, Weisburger 1977) and excess mortality was a severe problem in the studies. Mice were dosed by gavage for 5 days a week for up to 78 weeks and they were to be sacrificed at 92 weeks; however, only 14% of the animals survived to 78 weeks and less than 1% survived to 92 weeks. This compares with 66% of the controls surviving the 92-week experiment. All animals were necropsied, regardless of the time of death, and hepatocellular carcinoma was found in almost every treated animal. Carcinomas were observed as early as 16 weeks for the low-dose female group. The high mortality and virtual 100% tumor response are the more serious limitations of this study. The data from this study, however, were used in the DHS and EPA (1984) risk assessments.

7.2 Rat Studies

Several studies reported the production of malignant tumors in rats following subcutaneous injections and oral administration of carbon tetrachloride. Tumor production in rats has been demonstrated in at least four strains, and in both sexes. The results of these studies are summarized in Table 4.

Reuber and Glover (1967) injected Buffalo rats subcutaneously twice a week for up to 12 weeks. Control animals were given corn oil. The animals were 0.6, 4, 12, 24 or 52 weeks old at the beginning of the study. Newborn rats died in approximately 8 days due to hepatic and renal necrosis. Animals were sacrificed and necropsied following exposure at 12 weeks. They reported hepatic hyperplasia, hyperplastic nodules and a few cases of hepatic carcinoma.

In a later study, Reuber and Glover (1970) compared the carcinogenicity of CTC in 12-week-old male rats from the Japanese, Osborne-Mendel, Wistar, Black and Sprague-Dawley strains. The animals were subcutaneously injected twice a week for up to 105 weeks, depending on survival. Corn oil was administered to controls. All of the Sprague-Dawley strain died within 16 weeks, and all of the Black strain died within 18 weeks. Although hyperplastic nodules were reported in these two strains, no carcinomas were observed. The absence of carcinoma is possibly due to the poor survival; the latency period reported for carcinoma in this study was 68 weeks. Hyperplastic nodules and hepatic carcinoma were reported in the other three strains. Other lesions reported were hemangiomas, carcinomas of the thyroid

TABLE 4. RAT BIOASSAYS

Strain		Route	Dose (mg/kg)	Tumor Incidence %	(N)[1]	Tumor	Reference
Buffalo	m,f	sc	0	0	48		Reuber & Glover
	m		≈2060	2	40-56	carcinoma	1967
				12	"	hyperplas. nodule	
	f	sc	≈2060	5	"	carcinoma	
				40	"	hyperplas. nodule	
Japanese	m,f	sc	0	0	12		Reuber & Glover
			2080	80	15	hepatic carcinoma	1970
				13	15	hemangioma	
				20	15	thyroid carcinoma	
				7	15	leiomyosarcoma	
				20	15	hyperplas. nodule	
Osborne-Mendel	m	sc	0	0	12		Reuber & Glover
			2080	62	13	hepatic carcinoma	1970
				8	13	hemangioma	
				23	13	thyroid carcinoma	
				31	13	hyperplas. nodule	
Wistar	m	sc	0	0	12		Reuber & Glover
			2080	14	33	hepatic carcinoma	1970
				14	58	hyperplas. nodule	
Black[2]	m	sc	0	0	12		Reuber & Glover
			2080	41	17	hyperplas. nodule	1970
Sprague-Dawley[2]	m	sc	0	0	12		Reuber & Glover
			2080	12	17	hyperplas. nodule	1970
Osborne-Mendel[2]	m,f	oral	0	0	40		NCI 1976a,b
	m		47	4	50	hepatic carcinoma	1977
				4	50	neoplastic nodule	
			94	4	50	hepatic carcinoma	
				2	50	neoplastic nodule	
	f		80	8	50	hepatic carcinoma	
				4	50	neoplastic nodule	
			159	2	50	hepatic carcinoma	
				6	50	neoplastic nodule	

[1] Size of dose groups at beginning of study.
[2] Incidence rates may not be comparable due to high mortality rate during study.

gland, and subcutaneous leiomyosarcoma. Cirrhosis was reported in all animals. Due to the small group size, poor survival of several strains and the incomplete reporting of total dosage, and most importantly, the inappropriate route of exposure, (subcutaneous injections may produce high local concentrations and an uncertain whole-body dose) this study cannot be used in a quantitative risk assessment.

As in the mouse studies, NCI used carbon tetrachloride as a positive control in rat bioassays for chloroform, 1,1,1-trichloroethane and trichloroethylene (1976a,b and 1977, Weisburger 1977). The Osborne-Mendel rats were administered a time-weighted average dose of CTC by gavage for 78 weeks. All surviving animals were observed for up to an additional 32 weeks, but only 28% of the animals survived until that time. Thirty-nine percent of the pooled controls[1] survived the 110-week experiment. Hepatic carcinomas were found at both doses in both sexes. A lower incidence was reported in the high-dose females, but this may to have been a result of that dose group's high mortality rate prior to tumor expression. The first hepatocellular carcinoma was observed in the female dose group at 16 weeks and in the male dose group at 26 weeks. Tumors in other tissues were not discussed, although the authors implied that other tissues were examined. The EPA (1984) and National Academy of Sciences (NAS 1978) used this study in their risk assessment estimates; however, DHS staff members consider such use of the NCI study inappropriate because when the data are adjusted for excess mortality there is not a statistically significant association between dose and tumor response.

---

[1] Vehicle and non-vehicle controls.

7.3 Hamster study

CTC in corn oil was administered weekly by gavage to Syrian golden hamsters (both sexes) for a total of 30 weeks (Della Porta et al. 1961). The dosages were equivalent to 190 and 380 mg/kg of body weight respectively (EPA 1984).

Following treatment the animals were kept 25 weeks, sacrificed and examined. Only eight of the original 20 animals survived the full 55 weeks. The first animal death occurred at 10 weeks. Historical controls were used in the study and reportedly had not developed any liver-cell tumors (0/254). Carcinomas were not observed in the animals that died prior to the 43rd week (50%), but one or more liver-cell carcinomas were reported in the remaining animals. The high mortality rate and the 100% tumor incidence in surviving animals indicate that tumors may be produced at lower levels in this species. Furthermore, only 20 animals were treated, and it is likely that the animals were infected with a liver disease as many other colonies were in the early 1960's (Dungworth 1986, personal communication). Consequently, even though the hamster appears to be the most sensitive species studied and exhibited a very high tumor incidence rate at the exposure level used, the study was deemed inadequate for quantitative risk assessment.

7.4 Human Case Reports and Studies

The epidemiological data suggesting an association of carbon tetrachloride exposure with human cancer development are inconclusive; exposure data are unavailable and the putative association is confounded by exposure to other potential carcinogens. Several case reports stated that liver cancer developed following a single acute exposure (Tracey and Sherlock 1968, Simler et al. 1964) or a chronic exposure to CTC (Johnstone 1948). However, as with the epidemiological data, exposures were poorly documented, and the information can at best be used qualitatively to corroborate animal test data.

7.4.1 Human studies

Capurro (1979) reported a study on the residents in a rural valley polluted by vapors from a solvent recovery plant for at least 10 years. Chloroform, benzene, methyl isobutyl ketone, trichloroethylene and 26 other organic agents were detected in the air in addition to CTC (Capurro 1973). The author reported four excess cases of lymphoma. Attributing these cancer cases to CTC alone would be inappropriate since exposure to the other contaminants was verified in blood samples of residents.

In a preliminary study of 330 laundry and dry cleaning workers, Blair et al. (1979) examined occupational exposure to CTC and other dry cleaning agents. Information from death certificates indicated an excess of deaths from lung, cervical and liver cancers, and leukemia. Katz and Jowett (1981) studied female laundry and drycleaning workers in Wisconsin. Their results failed

to show an overall increase in malignant neoplasms, but they did report an elevated risk for cancers of the kidney and genitals (unspecified), along with smaller excesses of bladder and skin cancer and lymphosarcoma. However, the use of carbon tetrachloride has been of only minor importance in dry cleaning since the 1950's (DeShon 1978, Garfield 1985 a,b), and quantitative data on exposure to CTC were not presented in these studies.

Hernberg et al. (1984) reported a case-control study on primary liver cancer and exposure to solvents. Of 126 cases, 2 had a history of exposure to carbon tetrachloride, among other solvents. They concluded that there was an association between primary liver cancer and exposure to "solvents" among women, but not for men.

Recently, two reports were published on cancer mortality in a population of rubber workers (Checkoway et al. 1984, Wilcosky et al. 1984). Information on cause of death was reported in an earlier study (McMichael et al. 1974). They examined work history records to estimate the potential exposure experienced by the individuals. They reported a significantly elevated odds ratio relating carbon tetrachloride with lymphatic leukemia (OR=15.3, $p < 0.0001$) and lymphosarcoma and reticulum cell sarcoma (OR=4.2, $p < 0.05$). Attributing these outcomes to CTC alone is inappropriate since different solvents were used simultaneously in a given process area. That is, a high degree of correlation existed between exposure to several solvents and the incidence of lymphatic leukemia (CTC, carbon disulfide, ethyl acetate, acetone, and hexane) and lymphosarcoma (CTC, xylenes, carbon disulfide and hexane). Although carbon disulfide, ethyl acetate, acetone, hexane, and xylenes are not recognized carcinogens, these potentially confounding

exposures, the lack of association of CTC exposures with these cancers in other studies, and the small number of cases (19 of a study population of 6678), preclude any causal inference from this study.

Summary

Animal studies demonstrate that carbon tetrachloride produces hepatocellular carcinomas in the mouse, rat and hamster; human evidence is inconclusive. IARC evaluated CTC and concluded that it is an animal carcinogen. The IARC classification would place CTC in group 2B, indicating that it is probably carcinogenic to humans. DHS staff members concur with this assessment, based on the evidence cited in the preceding subsections. Since there are sufficient animal data to conclude that CTC is a potential human carcinogen, a risk assessment of CTC is presented in Section 9.

The epidemiological studies and human case reports are inadequate to use in a quantitative risk assessment. Thus, the quantitative risk assessment will be based on animal data. Two studies were selected for quantitative risk assessment: Edwards and Dalton (1942), and NCI (mouse) (1976a,b and 1977). These are discussed further in Chapter 9 on quantitative risk assessment.

8.0 SYNERGISM AND ANTAGONISM

8.1 Synergism

A number of substances have been shown to potentiate the noncarcinogenic effects of carbon tetrachloride. Some have been shown to increase its acute toxicity, such as ethanol, fats, polybrominated biphenyls, 2-butanone, 2,3-butanediol, chlordecone, phenobarbital, Mirex, and isopropanol (Curtis et al. 1979, Dietz and Traiger 1979, Klingensmith and Mehendale 1983, Kluwe et al. 1979, Mehendale 1984, Robbins 1929, Strubelt et al. 1978). Other compounds, such as benzo(a)pyrene and p-dimethylaminoazobenzene, may increase the chronic toxicity of CTC (Kotin et al. 1962, Moore and Ray 1983, Protzel et al. 1964, Ueda 1967).

8.1.1 Acute Interactions

The most prominent synergistic effect reported is the potentiation of hepatic toxicity in humans by alcohol (Bjarnason et al. 1968, Von Oettingen 1964). The precise nature of the potentiation has not been elucidated, but interest in the effect has generated numerous studies and a number of mechanisms have been proposed.

Alcohol increased the absorption of carbon tetrachloride from the intestinal tract in dogs (Robbins 1929); however, the effect (liver necrosis and mortality) was so much greater than the effect of CTC alone that the author concluded that "alcohol affects the toxicity in some way other than by changing the rate of absorption."

Klaassen and Plaa (1967) and Cornish and Adefuin (1966) reported that pretreatment with ethanol followed by CTC administration elevated the serum glutamic-pyruvic transaminase (SGPT) in dogs by an order of magnitude, indicating liver dysfunction. Other studies examining ethanol potentiation of CTC toxicity were conducted by Traiger and Plaa (1971), Wei et al. (1971), and Strubelt et al. (1978). In summary, ethanol has been shown to potentiate the acute toxicity of carbon tetrachloride in several animal species, including humans; it is unknown whether such potentiation extends to chronic effects and carcinogenicity.

8.1.2 Chronic Interactions

A study by Ueda (1967) compared the chronic toxicity of CTC and p-dimethylaminoazobenzene (DAB), administered separately and simultaneously. Liver carcinoma and cholangiofibrosis were observed in the DAB treatment group alone. Liver fibrosis and cirrhosis were observed in the CTC treatment group. The combined regimen produced nodular hyperplasia with liver cirrhosis, and greater incidences of liver carcinoma and cholangiofibrosis. DAB did not increase the incidences of liver fibrosis or cirrhosis observed with CTC, although the appearance of cirrhosis occurred earlier. The primary interaction observed was the potentiation by CTC, as a promoter, of DAB's carcinogenic effects.

Liver damage produced by CTC has been shown to change the rate of benzo(a)pyrene metabolism and the profile of metabolites formed (Kotin et al. 1962). When CTC (0.06 ml/100 g b.w.) was injected prior to injection of 200 μg of radiolabeled benzo(a)pyrene, clearance of benzo(a)pyrene was

reduced and there was a decrease in glucuronide conjugation of 3-hydroxybenzo(a)pyrene. Kotin et al. (1962) also studied the effects in mice of chronically administered CTC and benzo(a)pyrene, both separately and together. No tumors were observed in animals treated with CTC alone. However, CTC administration doubled the incidence of tumors present when compared with those animals treated with benzo(a)pyrene alone. A similar enhancement of benzo(a)pyrene tumor production was observed by Protzel et al. (1964), who applied a 0.1% solution of benzo(a)pyrene to the buccal mucosa of mice and periodically injected a 10% solution of carbon tetrachloride. CTC also enhanced 2-acetylaminofluorene (AAF) tumor production, apparently by increasing the metabolism of AAF to the N-hydroxylated metabolite (Weisburger et al. 1963 and 1965).

8.2 Antagonism

A number of compounds reportedly produced a protective effect against carbon tetrachloride toxicity. Beattie et al. (1944) reported that oral administration of methionine prevented a fatality in an acute case of CTC poisoning. In an animal study, calcium lactate (added to the feed at 1.2 g/day) increased the number of CTC exposures (100 and 200 ppm) that guinea pigs were able to tolerate before death (Smyth et al. 1936). Other substances that have reportedly antagonized the effects of CTC include dietary vitamin E, selenium, reserpine, carbon disulfide, diethyldithiocarbamate, chloramphenicol and chlorpromazine (EPA 1984). However, many of these compounds primarily reduce subclinical effects, so it is unclear how effective they would be following human administration.

In summary, the acute toxicity of CTC can be potentiated by a number of substances, including some commonly used drugs and some environmental pollutants. CTC itself has been shown to act as a carcinogenic promoter for several substances. Other compounds have been shown to antagonize the effects of CTC; however, there is not sufficient information to incorporate this into the risk assessment.

9.0 QUANTITATIVE RISK ASSESSMENT

A health assessment document for CTC was recently prepared by the EPA (1984). The document was peer-reviewed by members of the EPA Office of Health and Environmental Assessment (OHEA) and by external reviewers not connected with EPA. The initial purpose of the document was for use by the Office of Air Quality Planning and Standards, although the final scope was expanded to include other areas. The health assessment included quantitative risk estimates for cancer. The quantitative risk assessment section of the document has been modified for use in this report and is included in Appendix A. The staff of DHS believes the EPA (1984) risk assessment, as modified, is a reasonable and appropriate interpretation of the health effects data on CTC, as explained below.

9.1 Noncarcinogenic Risks

The effects of CTC following acute and chronic exposure are presented in Sections 3 to 7. The NOAEL in humans for a 3-hour acute exposure is 10 ppm while the NOAEL for rats for chronic exposure was reported to be 5 ppm. These levels are four orders of magnitude above the highest ambient maximum 24-hour concentrations reported in Part A of this document. However, NOAEL's for other species were not attained when subchronic testing was conducted at 1 ppm. Furthermore, chronic animal studies have not been conducted that establish a NOAEL for biochemical liver changes. Even in the absence of this information, based on the relatively low ambient CTC concentrations, noncarcinogenic, chronic intoxication is not expected to result in the general population from inhalation of carbon tetrachloride.

9.2 Carcinogenic Risks

9.2.1 Thresholds

For toxicologic purposes, a threshold dose is one below which a specified outcome does not occur; however, the self-propagating, clonal nature of tumor growth and development from a single damaged cell suggests that the effective dose for carcinogenesis may be so low as to be indistinguishable from zero. While threshold models (based on detoxification enzyme saturation, the existence of DNA repair mechanisms, recurrent cytotoxicity) have been proposed, none has been convincingly demonstrated.

An "epigenetic mechanism" that could theoretically embody threshold doses has been invoked to explain the carcinogenic action of substances that do not directly produce genetic damage in short-term tests. However, neither short-term tests nor nonlinearities in dose-response curves from animal bioassays can reliably distinguish between "genetic" versus "epigenetic" carcinogenesis, primarily because of the limited sensitivities of the experimental methodologies. In the case of CTC there is evidence suggesting potential genotoxicity (because of binding to DNA) without much evidence of mutagenicity (see Chapter 6). There is also experimental support for CTC acting as a promoter of tumorigenesis (see Chapter 8). On the other hand, in some experiments CTC alone was an effective carcinogen (see Chapter 7). Thus, the mechanism of CTC-induced carcinogenesis has not been elucidated. Since CTC produces its toxicity via production of the trichloromethyl radical, it might be assumed that a threshold exists based on the presence of antioxidants and free radical scavengers. However, as indicated by

Slater et al. (1985) "it is evident that effective scavenging of such species must satisfy a number of demanding criteria," including the ability to penetrate to the precise intracellular locus of metabolic activation. On the other hand, the DNA must compete with other biomolecules that would be damaged by the trichloromethyl radical as well as with the potential noncytotoxic scavengers. Consequently, further study is required to elucidate the effectiveness of the competing processes. DHS staff agrees with the conclusion of the IARC (1983) that there is insufficient evidence at present to justify creating separate classes of carcinogens (based on mechanism) for which different risk assessment methods would be used. Thus, in the absence of compelling evidence to the contrary, DHS treats carcinogenesis as a non-threshold phenomenon.

9.2.2 Risk assessment

The quantitative risk assessment of CTC conducted by EPA is contained in Appendix A. DHS modified the EPA risk assessment by: 1) applying an absorption fraction of 50% instead of 40%; 2) omitting the rat bioassay (NCI 1976 a, b, 1977) and the hamster bioassay (Della Porta et al. 1961) EPA used ; 3) assuming an average inhalation intake of 18 μg/day instead of 20 μg/day; and 4) presenting the range of resulting unit risks instead of the geometric mean. The range of human equivalent excess cancer risk estimates for lifetime inhalation exposure to CTC at 1$\mu g/m^3$ determined by DHS is presented in Table 5. Discussion of the differences between the EPA and DHS interpretations follow.

EPA chose an inhalation absorption fraction of 40%. EPA considered three reports in calculation of their inhalation absorption fraction (Lehmann and Schmidt-Kehl 1936, McCollister et al 1951, and Stokinger and Woodward 1958). The Stokinger and Woodward (1958) 30% absorption value represents their interpretation of the literature; however, they did not provide any data to support their conclusion. DHS staff decided to consider only the Lehmann and Schmidt-Kehl (1936) and the McCollister et al. (1951) studies in calculating an inhalation absorption fraction of 50% of the total CTC inhaled.

No animal study in the CTC health effects literature is ideal for use in a quantitative risk assessment. Four studies have sufficient information to estimate unit risk (Della Porta et al. 1961, Edwards et al. 1942, and NCI 1976 a,b, 1977 [rat, mouse]). However, DHS believes that the use of the rat NCI (1976 a,b, 1977) data and the hamster (Della Porta et al. 1961) data, is inappropriate as discussed in Sections 7.2 and 7.3. The NCI study used both vehicle and nonvehicle controls; when only vehicle controls are used and adjustments are made for early mortality, the NCI rat study data do not indicate a statistically significant association between dose and tumor response for either sex at either of the doses tested. Therefore, the NCI rat study constitutes a negative carcinogenic result. The hamster bioassay study was very small, using ten male and ten female animals. Only one dose level was tested. The tumor incidence was compared to 50 male and 30 female vehicle controls that were not concurrent, but they were housed under similar conditions. The report indicates that a number of animal deaths occurred early in the study; however, time-to-tumor information was not available. Also, it is possible that the hamster colony was infected with a

liver disease as many of them were at that time (Dungworth 1987, personal communication). The staff at DHS did not include these data in the risk assessment even though the study represents the most sensitive carcinogenic response to CTC.

Limitations of the two other studies are discussed below. The major limitation of the database is that there is no appropriate inhalation study examining the carcinogenic effects in the literature. Consequently, oral studies have to be used and applied to inhalation exposure. In addition, none of the studies were originally designed for use in a carcinogenic risk assessment model and the quality of the data is not ideal. In these studies emphasis was placed on determining the presence of liver tumors; the presence of other tumors, although mentioned, was poorly documented. This factor may have little influence on the risk assessment since it appears that the liver is the predominant site for CTC-induced tumors.

The NCI mouse study (NCI 1976a,b, 1977) has a number of study design problems. There was considerable animal mortality, requiring that the data be adjusted for the animals dying prior to the observation of the first liver tumor. The tumor response was close to 100% in both dose groups, limiting the interpretation of the dose-response curve. The goodness-of-fit criterion was not satisfied for the multistage model; however, the data provided are sufficient to calculate a time-to-tumor risk estimate.

The major limitations of the Edwards and Dalton (1942) study are the use of only male control mice, the short duration of the experiment, and the irregular dosing schedule. Although the authors did not indicate whether

the hepatomas were benign or malignant, the fact that hepatocellular carcinomas were reported in mice (NCI 1976a,b 1977), rats (NCI 1976a,b 1977, Reuber and Glover 1970), and hamsters (Della Porta et al. 1961) indicates that the hepatomas should be used in a risk assessment.

The EPA risk assessment uses several correction factors in its risk calculations as described below. Corrections for the data are discussed in Appendix A. This includes reasons for combining groups, eliminating inappropriate data points, corrections for surface area, adjustments for experiment duration and time-weighted average approximations of dosage. The EPA then calculated the geometric mean of the risk estimates from each of the four studies.

The DHS risk assessment does not adopt EPA's approach of using the geometric mean. Instead DHS presents the range of the estimates predicted by each of the two studies used. See Table 5.

The exposure level used in the unit risk calculation was 1 $\mu g/m^3$ and it corresponds to an estimated inhaled intake of 18 μg/day. The dose-response curve for the multistage model at these low concentrations is projected to be linear for the 95% upper confidence limit. Thus the upper limit of risk can be estimated by multiplying the unit risk by the exposure concentration. Since there are no inhalation studies, the unit risk is estimated from ingestion studies. The expected daily intake for the inhalation route is calculated using the respiration rate, absorption fraction and the estimated human weight as described on page A-24 of the Appendix. Using an absorption fraction of 50%, an estimated human weight of 60 kg and an estimated

respiration rate of 18 m^3/day, the unit inhalation intake is 4.5 times the unit oral intake. Thus the EPA 95% upper confidence limit excess cancer risk values in Table 2 of the Appendix, are multiplied by a factor of 4.5 to obtain the values in Table 5.

As seen in Table 5 the upper limit estimate for inhalation exposure to 1$\mu g/m^3$ ranges from low to high values of 10 to 42 excess cancers per million people exposed continuously for a lifetime. The staff at DHS believes that this range best estimates the excess risk associated with inhalation exposure to CTC based on the available data.

TABLE 5. Human Equivalent Excess Cancer Risk Estimates for Lifetime Inhalation Exposure to 1 $\mu g/m^3$ Carbon Tetrachloride

Data Set	Maximum Likelihood Estimate	95% Upper Confidence Limit	Type of Model
Edwards et al., 1942	3.2×10^{-5}	4.2×10^{-5}	multistage
NCI 1976a,b,. 1977 mouse	8.1×10^{-6}	9.9×10^{-6}	multistage time-to-tumor

REFERENCES

The DHS conducted a search of the CTC literature in the following manner. In October 1985 a bibliography was compiled from several sources including a list of references submitted by the Air Resources Board, the references cited in the US EPA Health Effects document and several on-line literature searches of Toxline and Medline data bases available from the National Library of Medicine. An updated search of Toxline and Medline was conducted in December 1985. In December 1986, references cited in the Public Comments (Part C) were reviewed. In April 1987 an updated bibliographic search was conducted from Toxline, Medline and the Toxic Substances Control Act Test Submissions (from the US EPA) data bases. In addition to these sources, articles pertaining to carbon tetrachloride were obtained when reviewing specific scientific journals and as references in specific journal articles. In May 1987, a revised version of Part B incorporated information from articles published since the May 1986 version.

ACGIH (American Conference of Government Industrial Hygienists) (1984) Documentation of the Threshold Limit Values, 4th Edition, Supplemental Documentation 1984. American Conference of Government Industrial Hygienists, Inc., Cincinnati, OH, 74-76.

Adams CE, Hay MF, Lutwak-Mann C (1961) The action of various agents upon the rabbit embryo. J Embryol Exp Morph 9:468-491.

Adams EM, Spencer HC, Rowe VK, et al. (1952) Vapor toxicity of carbon tetrachloride determined by experiments on laboratory animals. Arch Indus Hyg Occup Med 6:50-66.

Ahr HJ, King LJ, Nastainczyk W, et al. (1980) The mechanism of chloroform and carbon monoxide formation from carbon tetrachloride by microsomal cytochrome P-450. Biochem Pharmacol 29:2855-2861.

Amacher DE, Zelljadt I (1983) The morphological transformation of Syrian hamster embryo cells by chemicals reportedly nonmutagenic to Salmonella typhimurium. Carcinogen 4:291-295.

Anttinen H, Oikarinen A, Puistola U, et al. (1985) Prevention by zinc of rat lung collagen accumulation in carbon tetrachloride injury. Am Rev Resp Dis 132:536-540.

Barber ED, Donish WH, Mueller KR (1981) A procedure for the quantitative measurement of the mutagenicity of volatile liquids in the Ames Salmonella/microsome assay. Mutat Res 90:31-48.

Beamer WH, McCollister DD, Atchinson GJ, et al. (1950) Studies with low vapor concentrations of carbon tetrachloride labeled with carbon fourteen. II. Absorption and elimination upon skin exposure. Fed Proc 9:257.

Beattie J, Herbert PH, Wechtel C, et al. (1944) Studies on hepatic dysfunction. 1. Carbon tetrachloride poisoning treated with casein digest and methionine. Br Med J 1:209-211.

Bergman K (1979) Whole-body autoradiography and allied tracer techniques in distribution and elimination studies of some organic solvents. Scand J Work Environ Health 5 (suppl. 1):144-263.

Bhattacharyya K (1965) Foetal and neonatal responses to hepatotoxic agents. J Pathol Bact 90:151-161.

Bjarnason O, Johannesson T, Jonasson TA (1968) Carbon tetrachloride poisoning in Reykjavik and vicinity. Archiv Toxicol 23:112-121.

Blair A, Decouffle P, Grauman D (1979) Causes of death among laundry and dry cleaning workers. Am J Public Health 69:508-511.

Boyd MR, Statham CN, Longo NS (1980) The pulmonary Clara cell as a target for toxic chemicals requiring metabolic activation; studies with carbon tetrachloride. J Pharmacol Exp Ther 212:109-114.

Brambilla G, Martelli A, Pino A, Robbiano L (1986) Sequential analysis of DNA damage and repair during the development of carcinogen-induced rat liver hyperplastic lesions. Cancer Res 46:3476-3481.

Braun R, Schoneich J (1975) The influence of ethanol and carbon tetrachloride in host-mediated assay with Salmonella typhimurium, Mutat Res 31:191-194.

Briese E (1938) The effect on the blood cells of the fetal rat produced by the inhalation of carbon tetrachloride by the mother during gestation. Am J Med Sci 195:787-793.

Browning E (1965) Toxicity and metabolism of industrial solvents. New York: Elsevier Publishing Co., pp. 173-188.

Bruckner JV, MacKenzie WF, Muralidhara S, Luthra R, Kyle GM, Acosta D (1986) Oral toxicity of carbon tetrachloride: Acute, subacute, and subchronic studies in rats. Fund Appl Toxicol 6:16-34.

Burk RF, Reiter R, Lane JM (1986) Hyperbaric oxygen protection against carbon tetrachloride hepatotoxicity in the rat. Gastroenterol 90:812-818.

Butler TC (1961) Reduction of carbon tetrachloride **in vivo** and reduction of carbon tetrachloride and chloroform **in vitro** by tissues and tissue constituents. J Pharmacol Exp Ther 134:311-319.

Callen DF, Wolf CR, Philpot RM (1980) Cytochrome P-450 mediated genetic activity and cytotoxicity of seven halogenated aliphatic hydrocarbons in Saccharomyces cerevisiae. Mutat Res 77:55-63.

Cameron GR, Karunaratne WAE (1936) Carbon tetrachloride cirrhosis in relation to liver regeneration. J Pathol Bacteriol 42:1.

Capurro PU (1973) Effects of exposure to solvents caused by air pollution with special reference to CCl_4 and its distribution in air. Clin Toxicol 6:109-124.

Capurro PU (1979) Cancer in a community subject to air pollution by solvent vapors. Clin Toxicol 14:285-294.

Castro JA, Diaz Gomez MI (1972) Studies on the irreversible binding of ^{14}C-Cl_4 to microsomal lipids in rats under varying experimental conditions. Toxicol Appl Pharmacol 23:541-552.

Chandler AC, Chopra, RN (1925) The toxicity of carbon tetrachloride to cats. A warning. Ind Med Gaz 60:406-407.

Chatterjee A (1966) Testicular degeneration in rats by carbon tetrachloride intoxication. Experientia 22:395-396.

Checkoway H, Wilcosky T, Wolf P, et al. (1984) An evaluation of the associations of leukemia and rubber industry solvent exposures. Am J Ind Med 5:239-249.

Ciccoli L, Casini AF, Benedetti A (1978) Free radical damage produced by carbon tetrachloride in the lipids of various rat tissues. Agents Action 8:303-310.

Comporti M (1985) Lipid peroxidation and cellular damage in toxic liver damage. Lab Invest 53:599-622.

Cooper K, Witmer C (1982) Low oxygen tension and its effects on CCl_4 toxicity and mutagenicity in the Ames test (Abstract Bh-6). Environmental Mutagen Society 13th annual Meeting, February 26-March 1. 13:99.

Cornish HH, Adefuin J (1966) Ethanol potentiation of halogenated aliphatic solvent toxicity. Am J Ind Hyg 27:57-61.

Craddock VM, Henderson AR (1978) De novo and repair replication of DNA in liver carcinogen-treated animals. Cancer Res 38:2135-2143.

Curtis LR, Williams WL, Mehendale HM (1979) Potentiation of the hepatotoxicity of carbon tetrachloride following preexposure to chlorodecone (Kepone) in the male rat. Toxicol Appl Pharmacol 51:283-293.

David A, Frantik E, Holusa R, et al. (1981) Role of time and concentration on carbon tetrachloride toxicity in rats. Int Arch Occup Environ Health 48:49-60.

Dean BJ, Hudson-Waker G (1979) An in vitro chromosome assay using cultured rat-liver cells. Mutat Res 64:329-337.

De Flora S, Camoirano A, Zanacchi P, et al. (1984) Mutagenicity testing with TA97 and TA102 of 30 DNA-damaging compounds, negative with other Salmonella strains. Mutat Res 134:159-165.

De Flora S, Zanacchi P, Camoirano A, et al. (1984) Genotoxic activity and potency of 135 compounds in the Ames reversion test and in a bacterial DNA-repair test. Mutat Res 133:162-198.

Della Porta G, Terracini B, Shubik P (1961) Induction with carbon tetrachloride of liver cell carcinomas in hamsters. J Natl Cancer Inst 26: 855-863.

DeShon HD (1978) Carbon tetrachloride. In: Kirk-Othmer (eds.) Encyclopedia of chemical technology, 3rd ed., New York: John Wiley & Sons, Vol. 5, pp. 704-714.

Diaz Gomez MI, Castro JA (1980a) Binding of carbon tetrachloride metabolites to liver nuclear DNA, proteins, and lipids. Toxicol Appl Pharmacol 56:199-206.

Diaz Gomez MI, Castro JA (1980b) Nuclear activation of carbon tetrachloride and chloroform. Res Commun Chem Pathol Pharmacol 27:191-194.

Diaz Gomez MI, Castro JA (1981) Reaction or trichloromethyl free radicals with deoxyribonucleic acid bases. Res Commun Chem Pathol Pharmacol 32:147-153.

Dietz FK, Traiger GJ (1979) Potentiation of CCl_4 of hepatotoxicity in rats by a metabolite of 2-butanone: 2,3-butanediol. Toxicol 14:209-215.

Direnzo AB, Gandolfi AJ, Sipes IG (1982) Microsomal bioactivation and covalent binding of aliphatic halides to DNA. Toxicol Lett 11:243-252.

Dowty BJ, Laseter JL (1976) The transplacental migration and accumulation in blood of volatile organic constituents. Pediat Res 10: 696-701.

Dudley SF (1935) Toxic nephritis following exposure to carbon tetrachloride and smoke fumes. J Roy Nav Med Serv 21:296-314.

Dupont P, Bernis P, Paduart P, et al. (1975) 45 cases of carbon tetrachloride poisoning. Acta Clin Belg 30:485-493.

Durk H, Frank H (1984) Carbon tetrachloride metabolism in vivo and exhalation of volatile alkanes: dependence upon oxygen partial pressure. Toxicology 30:249-257.

Edwards JE (1941) Hepatomas in mice induced with carbon tetrachloride. J Natl Cancer Inst 2:197-199.

Edwards JE, Dalton AJ (1942) Induction of cirrhosis of the liver and hepatomas in mice with carbon tetrachloride. J Natl Cancer Inst 3:19-41.

Edwards JE, Heston WE, Dalton AJ (1942) Induction of the carbon tetrachloride hepatoma in strain L mice. J Natl Cancer Inst 3:297-301.

EPA (U.S. Environmental Protection Agency) (1984) Health Assessment document for carbon tetrachloride. EPA-600/8-82-001F, Office of Research and Development, Office of Health and Environmental Assessment, Environmental Criteria and Assessment Office, Cincinnati, OH.

Eschenbrenner AB, Miller E (1943) Studies on hepatomas I. Size and spacing of multiple doses in the induction of carbon tetrachloride hepatomas. J Natl Cancer Inst 4:385-388.

Eschenbrenner AB, Miller E (1946) Liver necrosis and the induction of carbon tetrachloride hepatomas in strain A mice. J Natl Cancer Inst 6:325-341.

Farber JL (1985) The biochemical pathology of toxic cell death. Monogr Path 26:19-31.

Fowler JSL (1969) Carbon tetrachloride metabolism in the rabbit. Br J Pharmacol 37:733-737.

Garfield E (1985) a Drycleaning. Part 1. The process and its history: from starch to finish. Current Contents 22:3-12.

Garfield E (1985) b Drycleaning. Part 2. Commercial history and social impacts: Ironing out some pressing problems. Current Contents 23:3-11.

Gee DL, Bechtold MM, Tappel AL (1981) Carbon tetrachloride-induced lipid peroxidation simultaneous **in vivo** measurements on pentane and chloroform exhaled by the rat. Toxicol Lett 8:299-306.

Gilman MR (1971) A preliminary study of the teratogenic effects of inhaled carbon tetrachloride and ethyl alcohol consumption in the rat. Dissertation, Drexel University, as cited in EPA (1984).

Gualandi G (1984) Genotoxicity of the free-radical producers CCl_4 and lipoperoxide in Aspergillus nidulans. Mutat Res 136:109-114.

Hayes JR, Condie LW, Borzelleca JF (1986) Acute, 14-day repeated dosing, and 90-day subchronic toxicity studies of carbon tetrachloride in CD-1 mice. Fund Appl Toxicol 7:454-463.

Hernberg S, Korkala M, Asikainen U, et al. (1984) Primary liver cancer and exposure to solvents. Int Arch Environ Health 54:147-153.

Higgins GM, Stasney (1936) The peripheral blood in experimental cirrhosis of the liver. J Fol Haematol 54:129-144.

IARC (International Agency for Research in Cancer) (1972) IARC monographs on the evaluation of carcinogenic risk of chemicals to man. Lyon, France.1: 53-60.

IARC (International Agency for Research on Cancer) (1983) Approaches to classifying chemical carcinogens according to mechanism of activity. Joint IARC/IPCS/CEC Working Group Report, IARC Internal Technical Reports 83/001. Lyon, France.

IARC (International Agency for Research in Cancer) (1979) IARC monographs on the evaluation of carcinogenic risk of chemicals to humans - some halogenated hydrocarbons. Lyon, France. IARC 20:371-399.

IARC (International Agency for Research in Cancer) (1982) IARC monographs on the evaluation of carcinogenic risk of chemicals to humans. Lyon, France. Supplement 4:74-75.

Johnstone RT (1948) Occupational Medicine and Industrial Hygiene. St. Louis, MO: C.V. Mosby Co., pp. 157.

Kalla NR, Bansal MP (1975) Effect of carbon tetrachloride on gonadal physiology in male rats. Acta Anat 91:380-385.

Kanics L, Rubenstein D (1968) The effect of chronic carbon tetrachloride inhalation on the secretion of lipid by rat liver. Biochem Pharmacol 17:1959-1967.

Kanzantzis G, Bomford RR (1960) Dyspepsia due to inhalation of carbon tetrachloride vapour. Lancet I:360-362.

Katz RM, Jowett D (1981) Female laundry and dry cleaning workers in Wisconsin: A mortality analysis. Am J Public Health 71:305-307.

Klaassen CD, Plaa GL (1966) Relative effects of various chlorinated hydrocarbons on liver and kidney function in mice. Toxicol. Appl. Pharmacol. 9:139-151.

Klaassen CD, Plaa GL (1967) Relative effects of various chlorinated hydrocarbons on liver and kidney function in dogs. Toxicol. Appl. Pharmacol. 10:119-134.

Klingensmith JS, Mehendale HM (1983) Hepatic microsomal metabolism of CCl_4 after pretreatment with chlordecone, Mirex or phenobarbital in male rats. Drug. Metab. Dispos. 11:329-334.

Kluwe WM, Herrman CL and Hook JB (1979) Effects of dietary polychlorinated biphenyls and polybrominated biphenyls on the renal and hepatic toxicities of several chlorinated hydrocarbon solvents in mice. J Toxicol Environ Health 5:605-615.

Kotin P, Falk HL, Miller A (1962) Effect of carbon tetrachloride intoxication on metabolism of benzo(a)pyrene in rats and mice. J Nat Cancer Inst 28:725-745.

Kubic V, Anders MW (1981) Mechanism of the microsomal reduction of carbon tetrachloride and halothane. Chem Biol Interactions 34:201-207.

Lehmann KB (1911) Experimentelle studien uber den Einflufs techisch und hygienisch wichtiger Gase und Dampfe auf den Organismus. Arch Hyg 74: 1-60.

Lehmann KB, Hasegawa (1910) Studien uber die Absorption Chlorierter Kohlenwasserstoffe aus der Luft durch Tier und Mensch. Arch Hyg 72: 327.

Lehmann KB, Schmidt-Kehl L (1936) [The thirteen most important chlorinated aliphatic hydrocarbons from the standpoint of industrial hygiene. In German]. Arch Hyg 116:132-268.

Levy GN, Brabec MJ (1984) Binding of carbon tetrachloride metabolites to rat hepatic mitochondrial DNA. Toxicol Lett 22:229-234.

Link B, Durk H, Thiel D, et al. (1984) Binding of trichloromethyl radicals to lipids of the hepatic endoplasmic reticulum during tetrachloromethane metabolism. Biochem J 223:577-586.

Lundberg I, Ekdahl M, Kronevi T, Lidums V, Lundberg S (1986) Relative hepatotoxicity of some industrial solvents after intraperitoneal injection or inhalation exposure in rats. Environ Res 40:411-420.

McCann J, Ames BN (1976) Detection of carcinogens as mutagens in the Salmonella/microsome test: Assay of 300 chemicals: Discussion. Proc Natl Acad Sci USA 73:950-954.

McCollister DD, Beamer WH, Atchinson GJ, et al. (1950) Studies with low vapor concentrations of carbon tetrachloride labeled with carbon 14. I. Absorption, distribution and elimination upon inhalation by monkeys. Fed Proc 9:300.

McCollister DD, Beamer WH, Atchinson GJ, et al. (1951) The absorption, distribution and elimination of radioactive carbon tetrachloride by monkeys upon exposure to low vapor concentrations. J Pharmacol Exp Ther 102:112-124.

McCord CP, Sterner JH, Kline LL, et al. (1946) The thymol-barbital test in experimental carbon tetrachloride poisoning. Occup Med 2:160.

McDermott WV, Hardy HL (1963) Cirrhosis of the liver following chronic exposure to carbon tetrachloride. J Occup Med 5:249-251.

McMichael AJ, Spirtas R and Kupper LL (1974) An epidemiologic study of mortality within a cohort of rubber workers. JOM 16:458-464.

Mehendale HM (1984) Potential of halomethane hepatotoxicity: chlordecone and carbon tetrachloride. Fundam Appl Toxicol 4:295-308.

Merkur'eva RV, Bonashevskaya TI, Shatemikova IS, et al. (1979) Comparative biochemical and morphological investigation of the liver of experimental animals in the process of hepatotropic effect of

atmospheric pollution (on the model of carbon tetrachloride). J Hyg Epidemiol Microbiol Immunol 23:368-377.

Mirsalis JC, Butterworth BE (1980) Detection of unscheduled DNA synthesis in hepatocytes isolated from rats treated with nenotoxic agents:an in vivo-in vitro assay for potential carcinogens and mutagens. Carcinogenesis 1:621-625.

Mirsalis JC, Tyson CK, Butterworth BE (1982) Detection of genotoxic carcinogens in the in vivo-in vitro hepatocyte DNA repair assay. Environ Mutagen 4:553-562.

Moore L and Ray P (1983) Enhanced inhibition of hepatic microsomal calcium pump activity CCl_4 treatment of isopropanol-pretreated rats. Toxicol Appl Pharmacol 71:54-58.

NAS (National Academy of Sciences) (1977) Drinking water and health., Washington D. C.: NAS, National Research Council, pp. 939.

NCI (National Cancer Institute) (1976a) Report on the carcinogenesis bioassay of chloroform. U. S. Department of Health, Education and Welfare, Public Health Service, National Institutes of Health, Carcinogenesis Program, Division of Cancer Cause and Prevention. March 1, 1976.

NCI (National Cancer Institute) (1976b) Carcinogenesis Technical Report Series No. 2, Feb. 1976, Carcinogenesis bioassay of trichloroethylene, CAS No. 79-01-6, NCI-CG-TR2, U. S. Department of Health, Education and Welfare, Public Health Service, National Institutes of Health.

NCI (National Cancer Institute) (1977) Carcinogenesis Technical Report Series No. 3, Jan. 1977, Bioassay of 1.1.1-trichloroethane for possible carcinogenicity. NCI-CG-TR3, U. S. Department of Health, Education and Welfare, Public Health Service, National Institutes of Health.

NIOSH (National Institute for Occupational Safety and Health) (1975) Criteria Document on Carbon Tetrachloride. and Abstract of the Revised Standard, Pub. No. 76-133, U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control, Washington, D.C.: U.S. Government Printing Office, pp 15-63, 84-96.

NIOSH National Institute for Occupational Safety and Health) (1984) Registry of toxic effects of chemical substances, RTEC Quarterly on Microfiche, October 1984. (Lewis RJ and Swett DV, eds), U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control, DHHS Publication No. 83-107-4, Washington, D.C.: U.S. Government Printing Office 20402, pp. 472

Paul BB, Rubenstein D (1963) Metabolism of carbon tetrachloride and chloroform by the rat. J Pharmacol Exp Ther 141:141.

Perocco P, Prodi G (1981) DNA damage by haloalkanes in human lymphocytes cultured in vitro. Cancer Lett. 13:213-218.

Prendergast JA, Jones RA, Jenkins LJ, et al. (1967) Effects on experimental animals of long-term inhalation of trichloroethylene, carbon tetrachloride, 1,1,1-trichloroethane, dichlorodifluoromethane and 1,1-dichloroethylene. Toxicol Appl Pharmacol 10:270-289.

Protzel M, Giardina AC, Albano EH (1964) The effect of liver imbalance on the development of oral tumors in mice following the application of benzpyrene or tobacco tar. Oral Surg 18:622-625.

Recknagel RO (1983) A new direction in the study of carbon tetrachloride hepatoxicity. Life Sci 33:401-408.

Recknagel RO, Glende EA (1973) Carbon tetrachloride hepatotoxicity: An example of lethal cleavage. Crit Rev Toxicol, Nov. 1973, pp. 263-297.

Recknagel RO, Litteria M (1960) Amer J Pathol 36:521, as cited by EPA, 1984.

Reuber MD, Glover EL (1967) Hyperplastic and early neoplastic lesions of the liver in buffalo strain rats of various ages given subcutaneous carbon tetrachloride. J Natl Cancer Inst 38:891-899.

Reuber MD, Glover EL (1970) Cirrhosis and carcinoma of the liver in male rats given subcutaneous carbon tetrachloride. J Natl Cancer Inst 44:419-427.

Reynolds ES, Treinen RJ, Farrish HH, et al. (1984) Relationships between the pharmacokinetics of carbon tetrachloride conversion to carbon dioxide and chloroform and liver injury. Arch Toxicol Suppl 7:303-306.

Robbins BH (1929) The absorption, distribution and excretion of carbon tetrachloride in dogs under various conditions. J Pharmacol Exp Ther 37:203-216.

Rocchi P, Pordi G, Grilli S, Ferreri AM. (1973) In vivo and in vitro binding of CCl_4 with nucleic acids and proteins in rat and mouse liver. Int J Cancer 11:419-425.

Ruprah M, Mant TGK, Flanagan RJ (1985) Acute carbon tetrachloride poisoning in 19 patients: implications for diagnosis and treatment. Lancet, i: 1027-1029.

Sagai M, Tappel AL (1979) Lipid peroxidation induced by some halomethanes as measured by in vivo pentane production in the rat. Toxicol Appl Pharmacol 49:283-291.

Sansom AE (1867) On the action of the tetrachloride of carbon. Br Med J ii:206-208.

Schulze RM, Kappus H (1980) Lysis of erythrocytes as a result of microsomal lipid peroxidation induced by CCl_4 or $FeCl_2$. Res Com Chem Path Pharmacol 27:129-137.

Schwetz DW, Leong BKJ, Gehring PJ (1974) Embryo- and fetotoxicity of inhaled carbon tetrachloride, 1,1-dichloroethane and methyl ethyl ketone in rats. Toxicol Appl Pharmacol 28:452.

Shah H, Hartman SP, Weinhouse S (1979) Formation of carbonyl chloride in carbon tetrachloride metabolism by rat liver **in vitro**. Cancer Res 39:3942-3947.

Shimizu Y, Nagase C, Kawai K (1973) Accumulation and toxicity of carbon tetrachloride after repeated inhalation in rats. Ind Health 11:48-54.

Simler M, Maurer M, Mandard JC (1964) Cancer du foie sur cirrhose au tetrachlorure de carbone. J Med Strasbourg 15:910-918.

Simmon, V. F., Kauhanen, K. and Tardiff, R. G. (1977) Mutagenic Activity of Chemicals Identified in Drinking Water. In: Progress in Genetic Toxicology, Eds, Scoptt D, Bridges BA, Sobel FH, Elsevier/North Holland Biomedical Press, 1977, pp. 249-258.

Sina JF, Bean CL, Dysart GR, et al. (1983) Evaluation of the alkaline elution/rat hepatocyte assay as a predictor of carcinogenic/mutagenic potential. Mutat Res 113:357-391.

Slater TF (1966) Necrogenic action of carbon tetrachloride in the rat. A speculative mechanism based on activation. Nature 209:36.

Slater TF, Cheeseman KH, Ingold KU (1985) Carbon tetrachloride toxicity as a model for studying free-radical mediated liver injury. Phil Trans R Soc Lond B 311:633-645.

Smith MT, Sandy MS (1985) Role of extracellular Ca^{++} in toxic liver injury: comparative studies with the perfused rat liver and isolated hepatocytes. Toxicol Appl Pharmacol 81:213-219.

Smuckler EA, James JL (1984) Irreversible cell injury. Pharm Rev 36:77S-91S.

Smyth HF, Smyth HF Jr., Carpenter CP (1936) The chronic toxicity of carbon tetrachloride, animal exposures and field studies. J Ind Hyg Toxicol 18:277-298.

Soucek B (1961) Retention und Ausscheidung von Methylchlorid und Tetrachlorkohlenstoff bei Ratten. Arch Gewerbepath Gewerbehyg 18:370-383 as cited in Bergman (1979).

Stewart RD, Dodd HC (1964) Absorption of carbon tetrachloride, trichloroethylene, tetrachloroethylene, methyl chloride, and 1,1,1-trichloroethane through human skin. Am Ind Hyg Assoc 25:439-446.

Stewart RD, Gay HH, Erley DS, et al. (1961) Human exposure to carbon tetrachloride vapor. J Occup Med 3:586.

Stewart A, Witts LJ (1944) Chronic carbon tetrachloride intoxication. Br J Ind Med 1:11-19.

Strauss B (1954) Aplastic anemia following exposure to carbon tetrachloride. JAMA 155:737-739.

Stokinger HE, Woodward, RL (1958) Toxicologic methods for establishing drinking water standards. J Am Water Works Assoc 50:515-529.

Strubelt O, Obermeier F, Siegers CP, Volpel M (1978) Increased carbon tetrachloride hepatoxitoxicity after low-level ethanol consumption. Toxicol 10:261-270.

Swinyard EA (1975) Noxious gases and vapors. In: The Pharmacological Basis of Therapeutics, 5th ed. Goodman LS, Gilman A, eds, New York: Macmillan Publishing, Co., Inc., pp. 908-909.

Sundareson AE (1942) An experimental study on placental permeability to cirrhogenic poisons. J Path Bact 54:289-298.

Torkelson TR, Rowe VK (1981) Halogenated aliphatic hydrocarbons. In: Clayton GD, Clayton FE, eds, Patty's Industrial Hygiene and Toxicology 3rd. Ed., New York: John Wiley & Sons, pp. 3472-3478.

Tracey JP, Sherlock P (1968) Hepatoma following carbon tetrachloride poisoning. NY State J Med 68:2202-2204.

Traiger GJ, Plaa GL (1971) Differences in the potentiation of carbon tetrachloride in rats by ethanol and isopropanol pretreatment. Toxicol Appl Pharmacol 20:105-112.

Uehleke H, Werner T, Greim H, Kramer M (1977) Metabolic activation of haloalkanes and tests in vitro for mutagenicity. Xenobiotica 7:393-400.

Ueda U (1967) The relationship between experimental liver carcinoma and liver cirrhosis induced by simultaneous administration of p-dimethylamino-azobenzene and carbon tetrachloride. Tohoku J Exp Med 92:83-107.

Von Oettingen WF (1964) The halogenated hydrocarbons of industrial and toxicological importance. Elsevier Publishing Comp., New York, pp.107-170.

Von Oettingen WF, Powell CC, Sharpless NE, et al. (1949) Arch Ind Health Bull No 191.

Von Oettingen WF, Powell CC, Sharpless NE, et al. (1950) Comparative studies of the toxicity and pharmacodynamic action of chlorinated methanes with special reference to their physical and chemical characteristics. Arch Int Pharmacodyn 81:17-34.

Wei E, Wong LCK, Hine CH (1971) Potentiation of carbon tetrachloride hepatotoxicity by ethanol and cold. Toxicol Appl Pharmacol 18:329-334.

Weisburger JH, Hadidian Z, Fredrickson TN and Weisburger EK (1965) Carcinogenesis by simultaneous action of several agents. Toxicol Appl Pharmacol 7:502.

Weisburger EK (1977) Carcinogenicity studies on halogenated hydrocarbons. Environ Health Perspect 21:7-16.

Weisburger JH, Weisburger EK (1963) Endogenous and exogenous factors in chemical carcinogenesis by N-2-fluorenylacetamide. Acta Un Int Cancer 19:513-518.

Wilcosky TC, Checkoway H, Marshall EG, et al. (1984) Cancer mortality and solvent exposures in the rubber industry. Am Ind Hyg Assoc J 45:809-811.

Williams GM, Weisburger JH (1986) Chemical Carcinogens, in Casarett and Doull's Toxicology, Klaassen CD et al., eds., pp. 99-173.

Wilson JG (1954) Influence on the offspring of altered physiologic states during pregnancy in the rat. Ann NY Acad Sci 57:517-525.

Yalcin AS, Kocak-Toker N, Uysal M, et al. (1986) Stimulation of lipid peroxidation and impairment of glutathione-dependent defence system in the liver of rats repeatedly treated with carbon tetrachloride. J Appl Toxicol 6:303-306.

APPENDIX

QUANTITATIVE RISK ASSESSMENT FOR

CARBON TETRACHLORIDE

(Source: EPA, 1984)

APPENDIX

Unit Risk Estimates for Cancer

DEFINITION

Unit risk is one index of the relative carcinogenic potential of a chemical. Unit risk is defined here as the lifetime risk to humans of contracting cancer from a daily exposure to a concentration of 1 μg/ℓ in water via ingestion or a daily exposure to 1 μg/m³ in air via inhalation. The main assumptions for such risk estimates are 70 kg bw, 2 ℓ/day consumption of water and 20 m³/day inhalation rate (ICRP, 1975). The unit risk represents only the estimated risk at the stated exposure concentrations. It should not be interpreted as the slope at any exposure level since the shape of the curve in the low-dose region is not known.

The unit risk estimate for CCl_4 represents an extrapolation below the dose-risk range of experimental data. There is currently no solid scientific basis for any mathematical extrapolation model that relates exposure to cancer risk at the extremely low concentrations, including the unit concentration given above, that must be dealt with in evaluating environmental hazards. For practical reasons the correspondingly low levels of risk cannot be measured directly either by animal experiments or by epidemiologic studies. Low-dose extrapolation must, therefore, be based on current understanding of the mechanisms of carcinogenesis. At the present time the dominant view of the carcinogenic process involves the concept that most agents that cause cancer also cause irreversible damage to DNA. This position is based in part on the fact that a very large proportion of agents that cause cancer are also mutagenic. There is reason to expect that the quantal response that is characteristic of mutagenesis is associated with a linear non-threshold dose-response relationship. Indeed, there is substantial evidence from mutagenicity studies with both ionizing radiation and a wide

variety of chemicals that this type of dose-response model is the appropriate one to use. This is particularly true at the lower end of the dose-response curve; at higher doses, there can be an upward curvature probably reflecting the effects of multistage processes on the mutagenic response. The linear non-threshold dose-response relationship is also consistent with the relatively few epidemiologic studies of cancer responses to specific agents that contain enough information to make the evaluation possible (e.g., radiation-induced leukemia, breast and thyroid cancer, skin cancer induced by arsenic in drinking water, liver cancer induced by aflatoxin in the diet). Some supporting evidence also exists from animal experiments (e.g., liver tumors induced in mice by 2-acetylaminofluorene in the large scale ED_{01} study at the National Center for Toxicological Research and the initiation stage of the two-stage carcinogenesis model in rat liver and mouse skin).

Because it has the best, albeit limited, scientific basis of any of the current mathematical extrapolation models, the non-threshold model which is linear at low doses has been adopted as the primary basis for risk extrapolation to low levels of the dose-response relationship. The risk estimates made with such a model should be regarded as conservative, representing the most plausible upper-limit for the risk, i.e., the true risk is not likely to be higher than the estimate, but it could be lower.

The mathematical formulation chosen to describe the dose-response relationship at low doses is the linearized multistage model. This model employs enough arbitrary constants to be able to fit almost any monotonically increasing dose-response data. It is constrained to ensure linearity in the low dose region at least for the upper confidence limit by requiring non-negative values for the fitted coefficients. Furthermore, there exists

a procedure for estimating an upper confidence limit on the slope at low extrapolated doses that is based on fitting the data at all experimental dose levels. Other dose-response models have been proposed which are also linear in the low dose region. The procedure recommended by the Carcinogen Assessment Group of EPA, however, involves estimating a most plausible upper limit of the slope at low doses. The other models (discussed later) can be shown to give lower slopes for the same data set than does the linearized multistage model, when extrapolated to the low dose region. Thus, the extrapolation model preferred by the Carcinogen Assessment Group is the multistage model.

EXPERIMENTAL STUDIES USED IN UNIT RISK ESTIMATES

Three oral studies on animals have sufficient information to allow estimation of unit risk. The oral studies are the positive control data for mice and rats used in three of its bioassays (NCI, 1976a,b, 1977), the Edwards et al. (1942) mice data and the Della Porta et al. (1961) hamster data. The incidence data and other pertinent quantitative information on these studies are presented in Table 1. For all studies, male and female data were combined. This was done because of the small sample sizes in the groups segregated by sex. No appropriate inhalation studies or human oral studies were found in the available literature.

Each of these oral studies has one or several characteristics which make it less than ideal for risk estimation for continuous daily exposure over a lifetime. Della Porta et al. (1961) did not report results for a control group, although they did report the incidence rate for vehicle controls in a different study. Moreover, the dose was administered only once per week and was reduced by half after 7 weeks, forcing the use of a time-weighted average approximation to a daily dose. The sample size (19) was also small. Edwards et al. (1942) exposed the mice for only 4 months and observed them

TABLE 1

Data for Cancer Studies Used in Unit Risk Calculations

Animal	Average Daily Dose	Responders/ Tested	Other Data	Reference
Hamster	0	0/80	Average bw: 0.108 kg	Della Porta et al., 1961
	0.95 mg	10/19	Exposure: 30 weeks	
			Experiment: 55 weeks	
			Assumed lifespan: 55 weeks	
Mouse	0	2/52	Average bw: 0.03 kg (assumed)	Edwards et al., 1942
	15 mg	34/73	Exposure: 17 weeks	
			Experiment: 31 weeks	
			Assumed lifespan: 78 weeks	
Mouse	0	6/157	Average bw: 0.028 kg	NCI, 1976a,b, 1977
	21 mg	89/89	Exposure: 78 weeks	
	42 mg	90/93	Experiment: 92 weeks	
			Assumed lifespan: 92 weeks	
Rat (M,F)	0	0/37	Average bw: 0.45 kg	NCI, 1976a,b, 1977
(M)	11 mg	2/45	Exposure: 78 weeks	
(F)	18 mg	4/46	Experiment: 110 weeks	
(M)	21 mg	2/47	Assumed lifespan: 110 weeks	
(F)	36 mg	1/30		

M = male, F = female

for ~8 months, much less than the desired lifetime experiment (1.5-2 yrs). The ages varied; the animals initially ranged from 2.5-7.5 months of age. In the NCI (1976a,b, 1977) study on mice, both low and high dose groups showed virtually 100% response (89/89 and 90/93, respectively), so that no information was available on the slope of the dose-response curve. As a consequence, risk projections for doses lower than those used in the study will be underestimated by an unknown amount. In the NCI (1976a,b, 1977) study on rats, survival to the end of exposure was poor, the dose was changed forcing use of a TWA dose estimate, and when segregated by sex, the sample sizes were small so that only the low-dose females were statistically significantly different from controls. The combining of the male and female rats when different, though similar, doses were used (see Table 1) may add further uncertainty.

Insufficient metabolic and pharmacokinetic data precluded the selection of the most appropriate species for use in estimating human risk. Because no study could be selected as "best" and "most appropriate", all four data sets are used in separate estimates of unit risk. In addition, an average unit risk estimate is also calculated which is the geometric mean of the four separate estimates.

DESCRIPTION OF THE PREFERRED LOW-DOSE ANIMAL EXTRAPOLATION MODEL

Let P(d) represent the lifetime risk (probability) of cancer at dose d. The multistage model has the form

$$P(d) = 1 - \exp\left[-(q_0 + q_1 d + q_2 d^2 + \ldots + q_k d^k)\right]$$

where all coefficients (q_0, q_1 ...q_k) are non-negative. The unit risk estimates are based on excess or extra risk over the background rate at dose d, i.e. the effect of treatment:

$$P_t(d) = \frac{P(d) - P(o)}{1 - P(o)}$$

It follows that

$$P_t(d) = 1 - \exp\,[-(q_1 d + q_2 d^2 + \ldots + q_k d^k)].$$

The point estimate of the coefficients q_i, $i = 0, 1, 2, \ldots, k$, and consequently the extra risk function $P_t(d)$ at any given dose d, is calculated by maximzing the likelihood function of the data. The point estimate and the 95% upper confidence limit of the extra risk $P_t(d)$ are calculated by using the computer program GLOBAL 79 developed by Crump and Watson (1979). The upper confidence limit for the extra risk calculated at low doses is always linear with dose. This is conceptually consistent with the linear non-threshold concept discussed earlier. The slope parameter q_1^* is taken as an upper bound (at low doses) of the potency of the chemical in inducing cancer.

In fitting the dose-response model, the number of terms in the polynomial is chosen equal to (h-1), where h is the number of dose groups in the experiment including the control group.

Whenever the multistage model does not fit the data sufficiently well, the data point at the highest dose is deleted and the model is refitted to the rest of the data. This is continued until an acceptable fit to the data is obtained. To determine whether or not a fit is acceptable, the chi-square statistic

$$X^2 = \sum_{i=1}^{h} \frac{(X_i - N_i P_i)^2}{N_i P_i\,(1-P_i)}$$

is calculated where N_i is the number of animals in the i^{th} dose group, X_i is the number of animals in the i^{th} dose group with a tumor response, P_i is the probability of a response in the i^{th} dose group estimated by fitting the multistage model to the data, and h is the number of dose groups. The fit is determined to be unacceptable whenever X^2 is larger

than the cumulative 99% point of the chi-square distribution with f degrees of freedom, where f equals the number of dose groups minus the number of non-zero multistage coefficients.

CONSIDERATIONS IN SELECTING INCIDENCE DATA

The tumor incidence data are separated according to organ sites or tumor types. The set of data (i.e., dose and tumor incidence) used in the model is the set where the incidence is statistically significantly higher than the control for at least one test dose level and/or where the tumor incidence rate shows a statistically significant trend with respect to dose level. Usually, the conservative approach adopted by the Carcinogen Assessment Group selects the data set which gives the highest estimate of the unit risk for humans. Because of the difficulties with each CCl_4 study described earlier, no selection was deemed appropriate. Instead, the separate estimates and their geometric mean are presented.

If two or more significant tumor sites are observed in the same study, and if the data are available, the number of animals with at least one of the specific tumor sites under consideration is used as incidence data in the model.

DESCRIPTION OF THE PREFERRED METHOD FOR CALCULATING HUMAN EQUIVALENT UNIT RISK

The method adopted by the Carcinogen Assessment Group for calculating a human equivalent estimate of unit risk from animal data employs two adjustments (Federal Register, 1980b) reflecting species differences and the influence of exposure duration on lifetime cancer risk. First, the animal doses are expressed as the time-weighted-average (TWA) daily dose over the duration of the experiment, and the low-dose extrapolation model is fitted to the resulting dose-incidence data. The risk is then estimated for a daily dose of 2 μg/day, which is the oral daily exposure corresponding to

a water concentration of 1 μg/ℓ and human intake of 2 ℓ/day. This risk is then multiplied by each of the above adjustment factors to obtain the human equivalent oral unit risk.

The models used to develop the species and duration adjustment factors represent the best scientific judgment based on available data. Other approaches have been suggested and are discussed in a later section.

The preferred model for equitoxic dose across species, or equivalently for risk at a constant dose across species, is based on an adjustment for metabolic diffferences. Metabolic rate has been suggested to be roughly proportional to body surface area (Mantel and Schneiderman, 1975; Calabrese, 1982), thus the equitoxic model is dose/surface area = constant. Equating the animal and human ratios and solving for the human dose gives:

$$d_h = d_a \ (S_h/S_a)$$

where d is daily dose (mg/day), S is surface area, and a and h refer to animal and human, respectively. The surface area is roughly proportional to the 2/3 power of body weight, and the proportionality constant is approximately the same (~10) for a variety of species. The human dose is then approximated by

$$d_h = d_a \ (W_h/W_a)^{2/3}$$

The unit risk estimate represents the lifetime risk for lifetime exposure to the carcinogen. When the animal experiment is partial lifetime, an adjustment is necessary to allow for positive responses that would have occurred had sufficient time been allowed for the tumors to develop. The risk is then adjusted upward (or equivalently the dose downward) to reflect the missing responders in the short experiment. The adjustment coefficient is $(L/L_e)^3$, where L is the animal lifespan and L_e is the duration of the experiment. The exponent 3 is supported in part by Doll (1971), who

showed that age-specific rates for humans increase by at least the second power of age, thus the cumulative tumor rate should increase by at least the third power of age. The choice of 3 for the exponent is also supported by Druckrey (1967) who showed that for a constant incidence rate, the dose-duration relationship was represented by dt^n = constant with n ranging from 2-4 in his experiments. With n=3, Druckrey's results suggest that the dose used for half-lifetime exposure (and observation) can be reduced to one-eighth its value for lifetime exposure and the incidence rate will be the same. Druckrey's results then reflect the influence of both exposure duration and observation period on the resulting incidence rate.

This adjustment factor is consistent with the proportional hazard model proposed by Cox (1972) and the time-to-tumor model considered by Crump (1979, 1982) where the probability of cancer by age t at dose d is

$$P(d,t) = 1 - \exp [-f(t)*g(d)].$$

For comparison, Crump's (1982) time-to-tumor model is also used to estimate unit risk for the NCI rat and mice studies (NCI, 1976a,b, 1977) which had time-to-tumor data. In the above model, g(d) is the multistage polynomial, and f(t) is $(t-t_o)^k$, where t_o may be interpreted as minimum induction time.

Interpretation of Quantitative Estimates

For several reasons, the unit risk estimate based on animal bioassays is only an approximate indication of the absolute risk in populations exposed to known carcinogen concentrations. First, there are important species differences in uptake, metabolism, and organ distribution of carcinogens, as well as species differences in target site susceptibility, immunological responses, hormone function, dietary factors and disease. Second, the concept of equivalent doses for humans compared to animals on a mg/surface

area basis is virtually without experimental verification regarding carcinogenic response. Finally, human populations are variable with respect to genetic constitution and diet, living environment, activity patterns, and other cultural factors. The exposure levels used in the unit risk calculations (1 μg/ℓ, 1 μg/m³) correspond to estimated intake of 2 μg/day via ingestion and 20 μg/day via inhalation. The expected human intake rates for CCl_4 are in the same range: 4 μg/day from food, 9 μg/day from fluids and 13 μg/day from air (see Chapter 4). The assumed dose-response curve is quite linear in this dose range so that the risk is proportional to exposure level, i.e., the upper limit estimate of risk is the unit risk multiplied by the exposure concentration.

The unit risk estimate can give a rough indication of the relative potency of a given agent compared with other carcinogens. The comparative potency of different agents is more reliable when the comparison is based on studies in the same test species, strain and sex, and by the same route of exposure. For unit risk estimates for air, the preferable studies would use exposure by inhalation.

The quantitative aspect of the carcinogen risk assessment is included here because it may be of use in the regulatory decision-making process (e.g., setting regulatory priorities, evaluating the adequacy of technology-based controls, etc.). However, it should be recognized that the estimation of cancer risks to humans at low levels of exposure is uncertain. At best, the low-dose linear extrapolation model used here provides a rough, but plausible estimate of the upper-limit of risk, i.e., it is not likely that the true risk would be much more than the estimated risk, but it could very well be considerably lower. The risk estimates presented in subsequent sections should not be regarded as accurate representations of the expected

cancer risks even when the exposures are accurately defined. The estimates presented may be factored into regulatory decisions to the extent that the concept of upper risk limits is found to be useful.

Unit Risk Estimates for Ingestion Exposure

The unit risk estimates based on human equivalent doses as discussed above are given in Table 2. Since the NCI studies (1976a,b, 1977) on rats and mice were the only ones to present time-to-tumor data, they are the only data sets evaluated using the time-to-tumor model. Both the maximum likelihood estimates (MLE) and upper 95% confidence limits are presented, as well as their geometric means. For the NCI (1976a,b) mouse data, the goodness-of-fit criterion was not satisfied ($\chi^2 = 14.4$) for the multistage (and one-hit) model. The risk estimates are presented anyway since the model cannot be fitted to the data if the high dose group is deleted, due to the 100% response at the low dose. Because of the protective approach discussed earlier which led (in part) to the adoption of the multistage model, and because the MLE does not account for estimation errors due to small sample sizes, the 95% upper limit on risk is preferred. Furthermore, since no study was entirely adequate for risk assessment purposes, the geometric mean of the upper confidence limits is preferred as the most plausible upper limit estimate of unit risk. For lifetime ingestion of 2 ℓ/day of water, the recommended (based upon present information and current understanding of carcinogenesis) estimate of unit risk (concentration of 1 μg/ℓ) is 3.7×10^{-6}.

ALTERNATIVE METHODOLOGICAL APPROACHES

The methods described above, which have been adopted by the Carcinogen Assessment Group, are consistently conservative, i.e., tending toward high estimates of cancer risk. The aspect which contributes most to this conservatism is the choice of the linearized multistage model for low-dose

TABLE 2

Human Equivalent Unit Risk Estimates for Ingestion Exposure with Specific Adjustments[a]

Data Set	Extrapolation Model			
	Multistage[b]		Time-to-Tumor[b]	
	MLE	UL	MLE	UL
Della Porta et al. (1961)	2.1E-5	3.4E-5	[c]	
Edwards et al. (1942)	7.1E-6	9.4E-6	[c]	
NCI (1976) mouse	1.4E-6	1.8E-6	1.8E-6	2.2E-6
NCI (1976) rat	1.9E-7	3.1E-7	3.1E-7	5.3E-7
All (geometric mean)	2.5E-6	3.7E-6	7.5E-7	1.1E-6

[a]For ingestion of 1 μg CCl_4 per ℓ water daily for life. Species conversion uses dose/body surface area. Duration adjustment for partial lifetime experiment used for Edwards et al. (1942) study is (fraction lifespan)$^{-3}$.

[b]MLE = Maximum likelihood estimate; UL = upper 95% confidence limit.

[c]No time-to-tumor data were available for these studies.

extrapolation. Other extrapolation models have been suggested, and are included below for comparison. These other models generally give lower estimates of risk than does the multistage model.

The various adjustment factors can also be calculated in different ways. The uncertainties related to the several models and adjustment factors and their influences on the risk estimates are discussed below. Generally, most of the uncertainty in estimating cancer risk from animal data is due to the limited data available in the bioassays, especially due to the high dose levels used, so that almost nothing is known about the shape of the dose-response curve at low doses or about the differences in low-dose incidence rates across species.

Low-Dose Extrapolation Models

Four models are used to extrapolate from the region of the experimental ingestion data to the levels corresponding to 1 µg/ℓ (Albert, 1983). All of these models relate exposure level to the incidence of tumor-bearing animals (Table 3). The "linearized" multistage model is constrained to have non-negative parameter values, and has the same number of parameters as the number of dose groups (including the control group). The one-hit model has two parameters. These two models are linear at sufficiently low doses. The Weibull model has three parameters. When only two dose groups exist, the Weibull exponent is set at 1, and then the Weibull model is also linear at low doses. The log probit model is used to represent a class of models which are not linear at low doses. The multistage reduces to the one-hit if only two dose groups exist. With only two dose groups, the Weibull parameter k is set to 1 and the Weibull model also reduces to the one-hit model. Currently, as discussed previously, there is insufficient evidence to provide strong support for any low-dose extrapolation model, although there is some justification for low-dose linearity.

TABLE 3

Low-dose Extrapolation Models Used in Unit Risk Calculation

Model	Functional Form*	Comments
Multistage	$P(d) = 1-\exp(-q_0 - q_1 d^2 - \ldots - q_k d^k)$	$q_i \geq 0$, all i
One-hit	$P(d) = 1-\exp(-q_0 - q_1 d)$	$q_0, q_1 \geq 0$
Weibull	$P(d) = 1-\exp(-q_0 - q_1 d^k)$	$q_0, q_1 \geq 0$
Log-probit	$P(d) = C + (1-C)\,\phi\,(A + B \log d)$	$\phi(x)$ = cumulative normal distribution ($\mu = 0$, $\sigma^2 = 1$)
Time-to-Tumor	$P(d) = 1-\exp[(-q_0 - q_1 d - q_2 d^2)(t-t_0)^k]$,	$t \geq t_0$, $q_i \geq 0$, all i, $k \geq 0$
	$= 0$	$t < t_0$, t_0 = minimal induction time

*d = animal dose (mg/day). The parameters q_0, q_1, q_2 etc., A, B, C, t_0, k are all determined by statistically fitting each model to the animal dose-response data. $P(d)$ = cancer risk at dose d. Note that unit risk is based on excess risk = $(P(d)-P(0))/(1-P(0))$, where $P(0)$ is background incidence.

In addition, since the NCI (1976a,b, 1977) data provide information pertaining to early mortality and time-to-tumor estimates, a multistage Weibull model with five parameters (Crump, 1982) is also used (see Table 3). The time-to-tumor estimates are based largely on time-to-mortality with subsequent discovery of tumors. Since the NCI (1976a,b, 1977) studies are the only ones involving more than two dose groups, they are also the only data sets to which all four previous models can be applied.

Unit Risk Calculation Approach

Each low-dose extrapolation model is applied to the original unadjusted animal data. The resulting risk estimate is then converted into equivalent human unit risk by multiplication by several factors to adjust for experiment duration (if partial lifetime), species differences and, if necessary, route conversion. Uncertainties in each adjustment factor are investigated by changing the choice of each adjustment model and/or the assumed parameter values in the model. There is insufficient information to allow the alternatives to be characterized by likelihood or error distributions; hence, all decisions are based on scientific judgment. The adjustment categories and their decision alternatives are given in Table 4.

Each animal risk estimate obtained by fitting a low-dose extrapolation model to the animal data is presented as the maximum likelihood estimate (MLE) and, when possible, as the upper 95% confidence limit. The confidence limit is statistically more stable; the MLE may show substantial sensitivity to small changes in the original data. Some data errors are expected due to diagnostic uncertainties generally leading to underdetection, i.e., missed tumors. The confidence limit on risk, however, reflects sample size and random variability, and may be much higher than the MLE. Both estimates are investigated for their sensitivities to such data errors.

TABLE 4

Alternatives for Judgmental Decisions in Cancer Unit Risk Estimation

Decision Category	Alternatives
Low-dose extrapolation	Model: Multistage, one-hit, Weibull, log probit
	Estimator: Maximum likelihood estimate, upper 95% confidence limit
Equitoxic dose across species	Model: Dose/bw, dose/body surface area
Adjustment for partial lifetime experiment	Model: $(L/L_e)^k$, k = 1-4

The risk estimate from the low-dose extrapolation is based on average daily intake level. Gavage studies add uncertainty since gastrointestinal effects may be due to the repeated high local concentrations and may not occur if the same daily dose were given continuously, hence at a lower local concentration. Since insufficient pharmacokinetic information exists to adjust for intermittent exposure, the TWA daily intake rate is used.

The extrapolation from risk based on partial lifetime experiments to risk from lifetime exposure uses the factor: $(L/L_e)^k$, where L is life-span and L_e is the duration of the experiment. As discussed previously, there is some evidence from human and animal studies to support an exponent of 3. Druckrey (1967) found that, at constant risk, fitting the model dt^n = constant to a series of data on nitrosamines gave values of n of 2-4. Studies of age-specific cancer rates in humans give estimates of k at 3 or higher. The concept of total dose (dose rate x duration) or similarly time-weighted-average dose for the experiment has been used as an indication of toxic severity, but has not been verified for cancer risk. It is included here for completeness and is represented by the case k=1 in the adjustment factor. There is insufficient information at present to allow precise determination of k for most chemicals. In the absence of such information, the sensitivity of the risk estimate to k is demonstrated by varying k between 1 and 4, and displaying the resulting range of risk estimates.

The conversion factor for species differences is presently based on models for equitoxic dose. The two general models currently used are based on body burden, dose/bw = constant (Stara and Kello, 1974); and metabolic rate, dose/body surface area = constant (Mantel and Schneiderman, 1975; Calabrese, 1982). In the absence of pharmacokinetic data related to

toxicity on the test chemical for the experimental species and for humans, both models are used.

The conversion of administered dose from one route to another (e.g., ingestion to inhalation) is not well understood. The approach by Stockinger and Woodward (1958) uses approximate net absorption fractions and daily intake rate to convert from one route to an equitoxic exposure level via another route. This approach, discussed below, is used to give approximate estimates of exposure levels when insufficient data exist for the desired route, although the estimates are acknowledged as being highly uncertain and probably inaccurate.

Unit Risk Estimates for Ingestion Exposure

The unit risk estimates based on human equivalent doses consistent with previous guidelines (Federal Register, 1980b) are given in Table 5 for the four data sets and the applicable models. Note that for the NCI mouse and rat data, the unconstrained Weibull and unconstrained log probit models could not be used to estimate risk. The failure of the computer algorithm to yield meaningful results is attributed to the virtually flat slope at the low and high dose data. This caused the extrapolated risk to be essentially dose-independent, i.e., the same as the risk in the dosed groups regardless of the level of dose. The successfully applied models were fitted to the original animal data using average daily intake (mg/day); their parameter estimates are given in Table 6. The human risk was then calculated by multiplying the animal unit risk by the adjustment coefficients reflecting partial lifetime exposure and species conversion. The low-dose extrapolation of the NCI rat data is shown in Figure 1 as an example of the difference in unit risk estimation (risk at 1 μg/ℓ) due only to selection of extrapolation model. The risk projections have not been adjusted for species

TABLE 5

Unit Risk Estimates for Ingestion[a]

Study	Model (type of estimate)[b]					
	Multistage[c]		Time-to-Tumor		Weibull	Log Probit
	(MLE)	(UL)	(MLE)	(UL)		
Della Porta et al. (1961)	2.1E-5	3.4E-5	d/		e/	e/
Edwards et al. (1942)	7.1E-6	9.4E-6	d/		e/	e/
NCI (1976) mouse	1.4E-6	1.8E-6	1.8E-6	2.2E-6	f/	f/
NCI (1976) rat	1.9E-7	3.1E-7	3.1E-7	5.3E-7	f/	f/

[a]For ingestion of 1 µg CCl_4 per ℓ water daily for life. Species conversion uses dose/body surface area.

[b]MLE = Maximum likelihood estimate; UL = upper 95% confidence limit.

[c]The one-hit model results agreed with the multistage model results to three significant figures.

[d]No time-to-tumor data were available for these studies.

[e]These models have three parameters which could not be fit by the two dose groups in these studies.

[f]The unconstrained Weibull and unconstrained log probit models could not be used to estimate unit risk for these studies. See text.

TABLE 6

Parameter Estimates for the Low Dose Extrapolation Models Fitted to Animal Data[a]

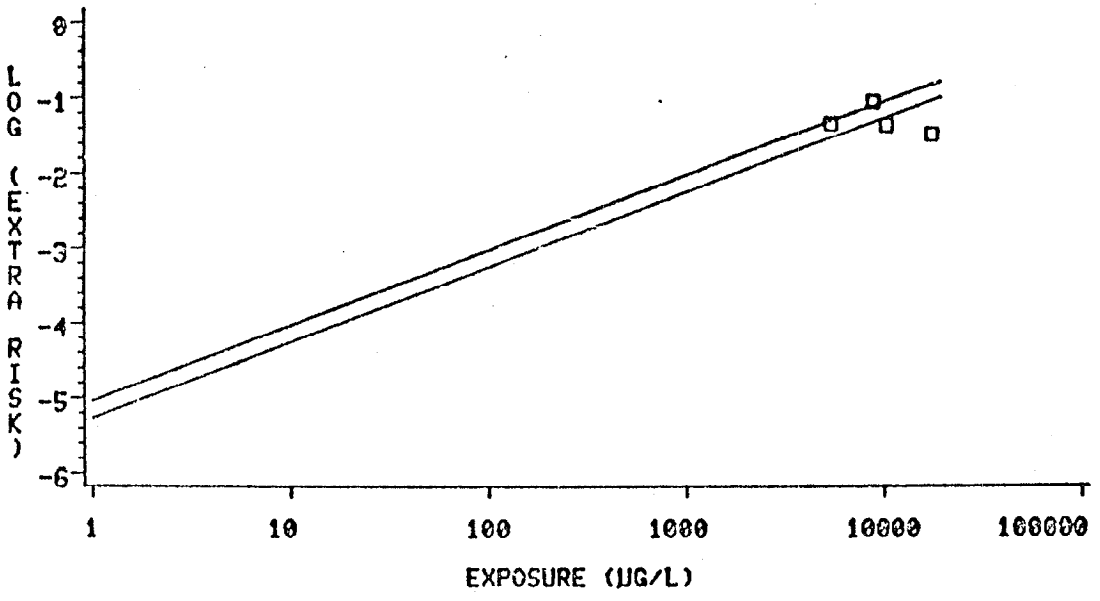
Study	Multistage				Time-to-tumor				
	q_0	q_1	q_1^*	q_2	q_0	q_1	q_1^*	t_0	k
Della Porta et al., 1961	0	0.784	1.28	--	--	--	--	--	--
Edwards et al., 1942	0.0133	0.0415	0.0549	--	--	--	--	--	--
NCI, 1976 mouse	0.0397	0.133	0.167	0	0.13E-12	4.96E-13	5.90E-13	0	5.87
NCI, 1976 rat	0	2.71E-3	4.47E-3	0[b]	0	2.47E-13	4.15E-13	0	5.03

[a]See Table 3 for model definitions. q_0 is dimensionless. q_1 and q_1^* are in $(mg/day)^{-1}$. q_2 is in $(mg/day)^{-2}$. t_0 is in (wk). All values are maximum likelihood estimates except q_1^*, which is the upper 95% confidence limit for q_1. E denotes powers of 10, i.e., $0.16E\text{-}13 = 0.16 \times 10^{-13}$.

[b]Estimates of q_3 and q_4 were also zero.

RISK ESTIMATES FOR CARBON TETRACHLORIDE

COMPARISON OF EXTRAPOLATION MODELS
DATA FROM NCI(1976) RAT STUDY



UPPER CURVE IS MLE TIME-TO-TUMOR MODEL
LOWER CURVE IS MLE MULTISTAGE MODEL

FIGURE 1

Comparison of low-dose extrapolation models. The models are applied to the NCI (1976) rat data: upper curve is MLE for time-to-tumor model, lower curve is MLE for multistage model. Risks are for animals. The curves are spline fits to the data; one data point not pictured is at the origin (0, 0). At 1 µg/ℓ the risk estimates from the curves differ by a factor of 17.

differences so that the actual rat response rates (the 4 data points) can be plotted. The logarithmic plot precluded the display of the control group response (0%).

The possibility of errors in the incidence data increase the uncertainty in the unit risk estimates. Such errors are usually under-detection of tumors (Anonymous, 1983), causing positively responding animals to be classified as nonresponders, and leading to underestimates of the unit risk. Furthermore, previous studies have suggested that the multistage maximum likelihood estimate, q_1, is more sensitive to such data errors than is the upper confidence limit, q_1^*. The sensitivities of q_1 and q_1^* to data changes are investigated for each of the three oral studies under consideration.

Discussion of possible detection errors in incidence data at the "ED_{01}" conference* suggested the following: that misclassification is more likely to underestimate the incidence of tumor-bearing animals than overestimate the incidence, and that having one animal misclassified in a study of 50 animals is not rare. The sensitivity of the parameter estimates to data errors was determined here by increasing the number of responders by 1 for every 50 animals tested (e.g., 1 for up to 50 animals, 2 for 51-100 animals, etc.), and then recalculating the parameters. The results are presented in Table 7. For each of these studies, the MLE (q_1) seems to be approximately as sensitive as the upper bound (q_1^*) to the data changes. Note that the parameter estimates are obtained from the raw data, are unadjusted and, thus, are not comparable across studies.

---

*"Workshop on Biological and Statistical Implications of the ED_{01} Study and Related Data Bases," Deer Creek State Park, Ohio, September 13-16, 1981.

TABLE 7

Sensitivity of Multistage Parameter Estimates to Changes in the Incidence Data

Study	Range of Parameter Values[a]			
	q_1 (MLE)	Relative Range[b]	q_1* (upper limit)	Relative Range[b]
Della Porta et al., 1961	0.758-0.907	16%	1.26-1.46	14%
Edwards et al., 1942	0.0402-0.0452	11%	0.0507-0.0592	9%
NCI, 1976 (mouse)	0.151-0.152	0.6%	0.227-0.228	0.4%
NCI, 1976 (rat	1.45E-4, 1.23E-3	89%	1.02E-3, 1.95E-3	48%

[a]These estimates are unadjusted for species or duration and are not comparable across studies. Relative ranges are comparable across studies.

[b]Relative range = (high-low)/high. The original data do not necessarily give the lower estimate, so the relative range does not necessarily represent the change from the original parameter estimates.

In Table 5, in line with previous guidelines (Federal Register, 1980b), the species conversion model was dose per body surface area and partial lifetime risk was adjusted to lifetime risk by the ratio $(L/L_e)^3$. The effects of using dose per body weight instead for species conversion, as well as exponents of 1 (total dose), 2 or 4 for the partial lifetime to lifetime adjustment are shown by the coefficients in Table 8 and the range of unit risk estimates in Table 9. Note that the adjustment coefficients in Table 8 are comparable only within a study, not across studies. The unit risk estimates in Table 9 are comparable across studies.

UNIT RISK ESTIMATES FOR INHALATION EXPOSURE

The unit risk for inhalation exposure is the excess cancer risk for lifetime exposure to 1 μg CCl_4/m^3 air. No inhalation cancer studies have been located which contain adequate dose-response information. However, the unit risk can be estimated from ingestion studies by assuming that the same daily intake rate results in the same lifetime risk. This assumption has not been thoroughly tested with other chemicals. In addition, each of the studies used for estimating oral unit risk has deficiencies. Therefore, the unit risk estimate for inhalation exposure should be considered approximate based on assumptions that have yet to be proven.

To estimate the risk corresponding to the concentration of 1 μg CCl_4/m^3 air, the effective dose in terms of mg/kg/day corresponding to 1 $\mu g/m^3$ must first be estimated. Assuming an air intake of 20 m^3/day (ICRP, 1975) and a 40% absorption rate by inhalation for humans (as recommended in this document), this effective dose can be estimated for a 70 kg human:

$$d_{air} = [20\ m^3/day \times 0.40 \times 1/(70\ kg)] \times 10^{-3}\ mg/m^3$$

$$= 1.14 \times 10^{-4}\ mg/kg/day = 7.98 \times 10^{-3}\ mg/day$$

TABLE 8

Adjustment Coefficients for Estimating Human Unit Risk
(duration adjustment) X (species adjustment)

Species Conversion: duration exponent k		Study[a] Della Porta et al. (1961)	Edwards et al. (1942)	NCI (1976a,b, 1977) (mouse)	NCI (1976a,b, 1977) (rat)
Surface area:	1	0.0134	0.0140	0.00543	0.0346
	2	0.0134	0.0345	0.00543	0.0346
	3[b]	0.0134	0.0852	0.00543	0.0346
	4	0.0134	0.210	0.00543	0.0346
Body weight:	1	0.00154	0.00105	0.00040	0.00643
	2	0.00154	0.00259	0.00040	0.00643
	3	0.00154	0.00640	0.00040	0.00643
	4	0.00154	0.0158	0.00040	0.00643

[a]Only the Edwards et al. (1942) study had partial lifetime experiment duration requiring the duration adjustment factor: $(\text{fraction lifespan})^{-k}$.

[b]The animal unit risk estimates were multiplied by the coefficients in this row (k = 3 in duration factor, dose/surface area species conversion) to obtain the human unit risk estimates in Table 4.

TABLE 9

Range of Unit Risk Estimates for Oral Exposure[a]

Study	Multistage Model		Time-to-Tumor Model	
	MLE	Upper Limit	MLE	Upper Limit
Della Porta et al. (1961)	2.4E-6, 2.1E-5	3.9E-6, 3.4E-5	NA[b]	
Edwards et al. (1942)	8.7E-8, 1.7E-5	1.2E-7, 2.3E-5	NA	
NCI (1976) mouse	1.0E-7, 1.4E-6	1.3E-7, 1.8E-6	1.3E-7, 1.8E-6	1.6E-7, 2.2E-6
NCI (1976) rat	3.5E-8, 1.9E-7	5.8E-8, 3.1E-7	5.8E-8, 3.1E-7	9.8E-8, 5.3E-7

[a]Obtained by changing the adjustment coefficients for species differences and partial lifetime experiment duration. See text.

[b]NA = Not applicable. Time-to-tumor data not available.

which is 4.0 times the unit intake for oral exposure (1 μg/ℓ = 2×10^{-3} mg/day). The unit risk for inhalation is then estimated from the oral studies by multiplying the oral unit risk by 4.0. Based on the range of recommended risks (based upon present information and current understanding of carcinogenesis) derived from the four oral data sets, and using the linearized multistage model with the dose per body surface area conversion across species, and with the exponent k=3 in the adjustment for partial lifetime study, the upper limit estimate of unit risk for inhalation exposure ranges from 1.2×10^{-6} to 1.4×10^{-4} with a geometric mean of 1.5×10^{-5}. As a measure of uncertainty, all models and adjustments were considered (see Table 9). The resulting upper limit unit risk estimate for inhalation exposure ranged from 2.3×10^{-7} to 1.4×10^{-4}.

Because of the uncertainties in both the qualitative and quantitative aspects of risk assessment, the actual cancer risks may be lower than the best unit risks presented above, which should be regarded only as plausible upper-limits. The unit risk estimates are calculated using a dose-response extrapolation model which is linear at low doses. This low-dose linearity is based on mutagenicity studies and on some similarities between mutagenesis and carcinogenesis. Since the results on the mutagenicity of CCl_4 are inconclusive, the selected extrapolation model may be inappropriate, and hence the unit risk estimates are uncertain.

MULTIPLE EXPOSURE SITUATIONS

The above information provides recommended route-specific cancer risk estimates associated with exposure to given units of CCl_4. These estimates may be conservative due to the choice of the multistage model for dose extrapolation and the various adjustment factors. Nevertheless, unit

risks for cancer presented above are defined for independent water and air exposures in that their computation assumes 100% of the insult is via the stated route.

When exposure is by both oral and inhalation routes, an additivity assumption can be used to calculate the risk associated with the concurrent exposures. It is a general recommendation to use the additivity assumption which is made since the available data on CCl_4 are limited and do not allow the presentation of a defensible alternative. As new information becomes available, other alternatives should be considered. Here the additivity assumption is that the risk associated with exposure to a given chemical via two routes concurrently is roughly the sum of the risks associated with each independent route-specific exposure. Since interactions between the concurrent routes of intake cannot be quantified, uncertainty surrounds the resulting risk estimate that is derived from the concurrent risks.

In applying the assumption of additivity, the risks rather than the doses associated with each route are added, but the mere summation of these risks is presently justifiable only when doses are low enough that no interaction occurs between the two routes. Furthermore, the amounts of 1 µg/ℓ and 1 µg/m³ are concentrations in water and air, respectively, not doses. The dose can be estimated by assuming consumption of 2 ℓ water/day over the lifetime. Thus, the daily dose corresponding to a concentration of 1 µg/ℓ water would be 2 ℓ/day x 1 µg/ℓ = 2 µg/day.

COMPARISON OF RISK ESTIMATES FOR VARIOUS CARCINOGENS

The carcinogenic risk from exposure to CCl_4 is compared to the risk from exposure to other potential carcinogens in Table 10. For comparison

TABLE 10

Relative Carcinogenic Potencies Among 53 Chemicals Evaluated by the Carcinogen Assessment Group as Suspect Human Carcinogens[a,b,c]

Compounds	Slope $(mg/kg/day)^{-1}$	Molecular Weight	Potency Index	Order of Magnitude (log_{10} index)
Acrylonitrile	0.24 (W)	53.1	$1x10^{+1}$	+1
Aflatoxin B_1	2924	312.3	$9x10^{+5}$	6
Aldrin	11.4	369.4	$4x10^{+3}$	+4
Allyl chloride	$1.19x10^{-2}$	76.5	$9x10^{-1}$	0
Arsenic	15 (H)	149.8	$2x10^{+3}$	+3
B[a]P	11.5	252.3	$3x10^{+3}$	+3
Benzene	$5.2x10^{-2}$ (W)	78	$4x10^{0}$	+1
Benzidine	234 (W)	184.2	$4x10^{+4}$	+5
Beryllium	1.40 (W)	9	$1x10^{+1}$	+2
Cadmium	6.65 (W)	112.4	$7x10^{+2}$	+3
Carbon tetrachloride	0.13	153.8	$2x10^{+1}$	+1
Chlordane	1.61	409.8	$7x10^{+2}$	+3
Chlorinated Ethanes				
1,2-dichloroethane	$6.9x10^{-2}$	98.9	$7x10^{0}$	+1
1,1,2-trichloroethane	$5.73x10^{-2}$	133.4	$8x10^{0}$	+1
1,1,2,2-tetrachloroethane	0.20	167.9	$3x10^{+1}$	+1
Hexachloroethane	$1.42x10^{-2}$	236.7	$3x10^{0}$	0
1,1,1-trichloroethane	$1.6x10^{-3}$	133.4	$2x10^{-1}$	-1
Chloroform	$7x10^{-2}$	119.4	$8x10^{0}$	+1

TABLE 10 (cont.)

Compounds	Slope $(mg/kg/day)^{-1}$	Molecular Weight	Potency Index	Order of Magnitude (log_{10} index)
Chromium	41 (W)	104	$4x10^{+3}$	+4
DDT	8.42	354.5	$3x10^{+3}$	+3
Dichlorobenzidine	1.69	253.1	$4x10^{+2}$	+3
1,1-Dichloroethylene	0.147 (I)	97	$1x10^{+1}$	+1
Dieldrin	30.4	380.9	$1x10^{+4}$	+4
Dinitrotoluene	0.31	182	$6x10^{+1}$	+2
Diphenylhydrazine	0.77	180	$1x10^{+2}$	+2
Epichlorohydrin	$9.9x10^{-3}$	92.5	$9x10^{-1}$	0
Bis(2-chloroethyl)ether	1.14	143	$2x10^{+2}$	+2
Bis(chloromethyl)ether	9300 (I)	115	$1x10^{+6}$	+6
Ethylene dibromide (EDB)	8.51	187.9	$2x10^{+3}$	+3
Ethylene oxide	1.26 (I)	44.1	$6x10^{+1}$	+2
Heptachlor	3.37	373.3	$1x10^{+3}$	+3
Hexachlorobenzene	1.67	284.4	$5x10^{+2}$	+3
Hexachlorobutadiene	$7.75x10^{-2}$	261	$2x10^{+1}$	+1
Hexachlorocyclohexane				
technical grade	4.75	290.9	$1x10^{+3}$	+3
alpha isomer	11.12	290.9	$3x10^{+3}$	+3
beta isomer	1.84	290.9	$5x10^{+2}$	+3
gamma isomer	1.33	290.9	$4x10^{+2}$	+3

TABLE 10 (cont.)

Compounds	Slope $(mg/kg/day)^{-1}$	Molecular Weight	Potency Index	Order of Magnitude (log_{10} index)
Methylene chloride	$6.3x10^{-4}$	84.9	$5x10^{-2}$	-1
Nickel	1.15 (W)	58.7	$7x10^{+1}$	+2
Nitrosamines				
Dimethylnitrosamine	25.9 (not by q_1^*)	74.1	$2x10^{+3}$	+3
Diethylnitrosamine	43.5 (not by q_1^*)	102.1	$4x10^{+3}$	+4
Dibutylnitrosamine	5.43	158.2	$9x10^{+2}$	+3
N-nitrosopyrrolidine	2.13	100.2	$2x10^{+2}$	+2
N-nitroso-N-ethylurea	32.9	117.1	$4x10^{+3}$	+4
N-nitroso-N-methylurea	302.6	103.1	$3x10^{+4}$	+4
N-nitroso-diphenylamine	$4.92x10^{-3}$	198	$1x10^{0}$	0
PCBs	4.34	324	$1x10^{+3}$	+3
Phenols				
2,4,6-trichlorophenol	$1.99x10^{-2}$	197.4	$4x10^{0}$	+1

TABLE 10 (cont.)

Compounds	Slope $(mg/kg/day)^{-1}$	Molecular Weight	Potency Index	Order of Magnitude (log_{10} index)
Tetrachlorodibenzo-p-dioxin (TCDD)	$1.56x10^{5}$	322	$5x10^{+7}$	+8
Tetrachloroethylene	$3.5x10^{-2}$	165.8	$6x10^{0}$	+1
Toxaphene	1.13	414	$5x10^{+2}$	+3
Trichloroethylene	$1.9x10^{-2}$	131.4	$2.5x10^{0}$	0
Vinyl chloride	$1.75x10^{-2}$ (I)	62.5	$1x10^{0}$	0
Vinylidene chloride	0.13 (I)	97	$1x10^{+1}$	+1

[a]Animal slopes are 95% upper-limit slopes based on the linear multistage model. They are calculated based on animal oral studies, except for those indicated by I (animal inhalation), W (human occupational exposure, and H (human drinking water exposure). Human slopes are point estimates, based on linear non-threshold model.

[b]The potency index is a rounded-off slope in $(mMol/kg/day)^{-1}$ and is calculated by multiplying the slopes in $(mg/kg/day)^{-1}$ by the molecular weight of the compound.

[c]Not all the carcinogenic potencies presented in this table represent the same degree of certainty. All are subject to change as new evidence becomes available.

purposes, the risks are based on lifetime esposure to 1 mg/kg/day. These estimates are derived from various studies on humans and animals, for oral and inhalation exposure. These estimates are derived with the linearized multistage model with species adjustment using dose/body surface area and adjustment for partial lifetime study by using the exponent k=3. Carbon tetrachloride has a relatively low potency compared to the others in the group.

A relative potency index, proposed by the Carcinogen Assessment Group, is also presented in Table 10 for each chemical. This index represents the risk posed by daily exposure to 1 mMol of carcinogen per kg body weight, and thus allows comparison of risks from exposure to the same number of molecules. The frequency distribution of the relative potency indices, rounded to the nearest order of magnitude, is shown in Figure 2.

SUMMARY/CONCLUSIONS

No single study was entirely adequate for risk estimation. Thus, the unit risk estimate is based on the geometric mean of the individual unit risk estimates from the four studies considered. The studies contained data on three animal species: rats, mice and hamsters. From these data, the recommended upper limit unit risk estimates (based upon present information and current understanding of carcinogenesis) for ingestion are in the range of 3.1×10^{-7} to 3.4×10^{-5} with a geometric mean of 3.7×10^{-6}. Using these same oral data, unit risk estimates for inhalation are in the range of 1.2×10^{-6} to 1.4×10^{-4} with a geometric mean of 1.5×10^{-5}. Since no study was entirely adequate for risk assessment purposes, the geometric mean of the upper confidence limits is preferred as the most plausible upper limit estimate of unit risk.

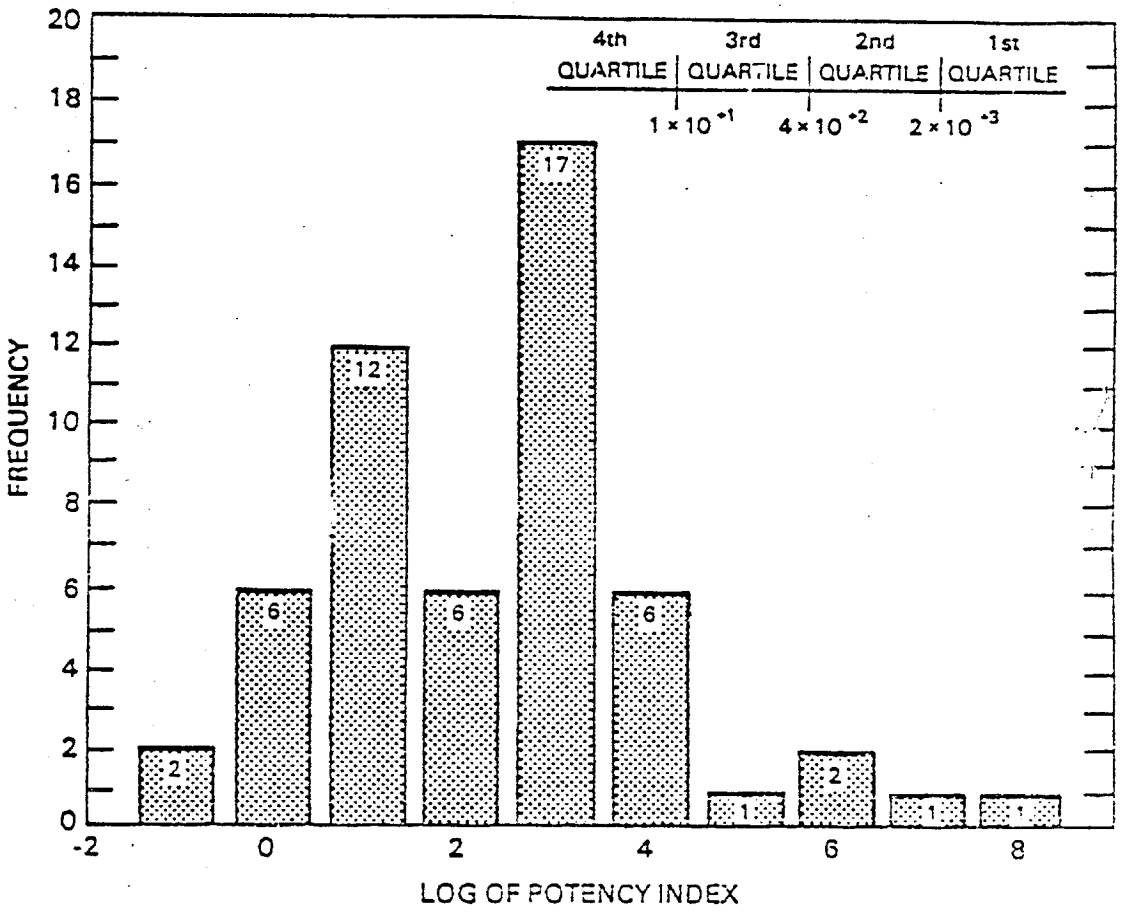


FIGURE 2

Histogram Representing the Frequency Distribution of the Potency Indices of 54 Suspect Carcinogens Evaluated by the Carcinogen Assessment Group