Ann. Rev. Pharmacol. Toxicol. 1977. 17:83–95

TOXICOLOGY OF HALOALKANE PROPELLANTS AND FIRE EXTINGUISHANTS

❖6668

Kenneth C. Back
6570th Aerospace Medical Research Laboratory, Wright-Patterson Air Force Base, Ohio 45433

Ethard W. Van Stee
National Institute of Environmental Health Sciences, Research Triangle Park, North Carolina 27709

INTRODUCTION

Low molecular weight halogenated alkanes, particularly certain fluoroalkanes, are of toxicological interest because of their industrial use as fire extinguishing agents, refrigerants, and solvents. Some fluoroalkanes also find use as aerosol propellants and are of interest to industrial and public health agencies because of widespread consumer use of pressurized household products and drugs. They also have a potential for abuse, particularly among drug-oriented youth. Another fluoroalkane, halothane, has been in common use in most hospitals since 1954 as an inhalation anesthetic.

Pharmacologically significant exposure of the human organism to these compounds, whether by design or accident, is usually by inhalation. Many compounds have a relatively high vapor pressure at ordinary conditions, so that they readily mix with air in pharmacologically significant concentrations. Fluoroalkanes readily diffuse through cell membranes because of their lipid solubility. Availability to the alveolar membrane, coupled with lipid solubility, results in a potential for quantitatively significant pulmonary absorption of fluoroalkanes.

Fluoroalkanes as a rule are not pulmonary irritants. In low concentrations acute inhalation is not an unpleasant experience, nor does prolonged esposure result in pathological changes in the upper respiratory tract or lungs. In somewhat higher concentrations inspiration may be resisted, but this is likely to be a consequence of the activation of certain reflexes such as the Kratschmer reflex.

A very extensive review of the literature concerning general fluorocarbon toxicity was written by Clayton in 1967 (1). More recently, in 1974, Aviado (2) reviewed

the toxicity of the fluorocarbon propellants. The propellant group includes but is not limited to such compounds as trichlorofluoromethane (CCl_3F), dichlorodifluoromethane (CCl_2F_2), trichlorotrifluoroethane (CCl_2F–$CClF_2$), dichlorotetrafluoroethane ($CClF_2$–$CClF_2$), and chlorodifluoromethane ($CHClF_2$). Fire extinguishing agents include trifluorobromomethane ($CBrF_3$), chlorobromodifluoromethane ($CBrClF_2$), dibromotetrafluoroethane ($CBrF_2$–$CBrF_2$), dibromotrifluoroethane ($CBrF_2$–$CHFBr$), and chlorobromomethane (CH_2ClBr). Inasmuch as space does not allow complete discussion of all the compounds used, and as each of the compounds has one or more of the toxicological properties discussed, the general field is reviewed using comparisons where possible.

ACUTE TOXICITY

The popularly accepted indexes of acute toxicity of the compounds listed above have been published by Engibous & Torkelson (3) and Clayton (1). Since the compounds have relatively high vapor pressures and are gases at room temperature, their principal mode of entry is via the respiratory route. The Underwriters Laboratories set up a relatively simplistic method for evaluating their relative toxicities. It involved exposing small numbers of guinea pigs for up to 2 hr and counting survivors. It is evident from this system that the fluoroalkanes are low in the order of toxicity.

Indexes such as median lethal concentration (LC_{50}) and approximate lethal concentrations (ALC) are of some comparative value for CH_2ClBr and $CBrClF_2$ when air was used as the diluent. One of the commonly cited figures for the ALC of $CBrF_3$ in rats is 83%. The report (1) indicated that air was used as the diluent in some of these studies, which would invalidate the results since the 83% $CBrF_3$ mixture in air would contain about 3.5% oxygen and the animals would have been severely hypoxic. Clayton did report, however, a mouse LC_{50} of 84% of $CBrF_3$ and a guinea pig LC_{50} of 88% with the balance of the inspired gas mixture, oxygen (1). Svirbely et al (4) reported an LC_{50} for CH_2ClBr of 2.9% in mice. Engibous & Torkelson (3) reported Army Chemical Center results of the exposure of one (!!) animal to CH_2ClBr and arrived an an ALC of 6.5%

The report by Clayton (1) of the levels of $CBrF_3$ and CH_2ClBr required to produce drowsiness in rats is more illuminating than the Army Chemical Center results. The ratio of 50% $CBrF_3$ to 0.475% CH_2ClBr equals about 105. This is very close to the ratio of a composite of cardiovascular effects found by Van Stee et al (5) in dogs.

NEUROLOGICAL EFFECTS

High brain levels of $CBrF_3$ were achieved in rats exposed to a nominal concentration of 75% $CBrF_3$ in oxygen. The levels were 50% higher than heart and blood levels (6). Aware that the compound reached the brain, Chikos et al (7) studied two basic cortical functions, the primary- and direct-evoked cortical responses. Both were depressed by exposure to $CBrF_3$, indicating cerebrocortical depression. The relative lipid solubilities of the compounds are $CBrF_3 < CBrClF_2 < CH_2ClBr$; therefore $CBrClF_2$ and CH_2ClBr would also be expected to accumulate in brain tissue, and probably to a greater extent than $CBrF_3$. Presence of significant levels of the com-

pounds in the brain would be related to the genesis of central nervous system (CNS) dysfunction.

Carter et al (8) studied the performance of operant-trained monkeys during exposure to $CBrF_3$. Exposure to 20–25% of $CBrF_3$ resulted in performance decrements while exposure to higher concentrations resulted in a complete disintegration of operant behavior. Similar effects would be expected from exposure to $CBrClF_2$ or CH_2ClBr although the precise character of the expected responses is unknown.

Van Stee & Back (9) reported that the electroencephalograms (EEG) of monkeys and dogs exposed to 70% of $CBrF_3$ were synchronized and had increased amplitude. The EEG could still be activated by sensory stimuli, however, and no seizure activity was detected. This is evidence of CNS depression without loss of consciousness. Since no seizure activity was elicited in the dogs that were paralyzed with curare during the recording sessions, the genesis of the convulsions may require a functional somatic motor system.

Hine et al (10), Haskell Laboratory (11), and Call (12) conducted human exposure experiments on $CBrF_3$. Hine et al (10) reported that exposure to 10–15% of $CBrF_3$ decreased the subjects' performance of five of six psychomotor tasks and at 15% caused feelings of impending unconsciousness. The volunteers also reported subjective changes in the sensorium at the lower levels. Call (12) reported only a slight increase in reaction time during 3-min exposure to 4 and 7% of $CBrF_3$. He further reported no significant interaction between the presence of $CBrF_3$ and hypoxia. Light-headedness, paresthesia, and diminished performance during exposure to up to 10% were reported by the Haskell Laboratory (11).

Clark (13) exposed human volunteers to 4 and 5% of $CBrClF_2$. Feelings of light-headedness and paresthesia were reported at 4% which were aggravated at 5%. Exposure to 5% of $CBrClF_2$ caused marked symptoms of CNS depression.

Controlled exposure of human volunteers to CH_2ClBr has not been reported. Rutstein (14) has, however, published three case reports involving accidental human exposures in which the absorbed doses could not be estimated. The subjects initially lost equilibrium and then consciousness. The loss of consciousness is consistent with the observation by Svirbely et al (4) of the anesthetic potency of CH_2ClBr in mice. This has been confirmed in other rodent species in our laboratory as well as in others.

During exposure of $CBrF_3$, monkeys have been observed by Van Stee & Back (15) to go into a trance-like state and Carter et al (8) reported behavioral depression. Dogs, on the other hand, convulsed during exposure to 40–80% $CBrF_3$ (10, 15).

Beck et al (16) reported tremors and convulsions in dogs exposed to 5–8.8% of $CBrClF_2$. The neurologically equivalent responses of dogs to 5–8% of $CBrClF_2$ were roughly equivalent to 40–80% of $CBrF_3$, an approximately 1:10 relationship.

CARDIAC ARRHYTHMIAS

That halogenated alkanes can interact with pressor amines to cause cardiac arrhythmias has been known since Levy and others first made the observation with chloroform and epinephrine at the turn of the century. Since then the concept has become popular that hydrocarbons and halogenated hydrocarbons can "sensitize" the heart

to the arrhythmogenic action of epinephrine. Cyclopropane-epinephrine arrhythmias have provided a popular standard for the testing of antiarrhythmic drugs for years. Since the phenomenon was recognized for anesthetics, etc, it was logical to try it out using aerosol propellants as well. It came as no surprise that this interaction with not only epinephrine but also with norepinephrine, metaraminol, ephedrine, phenylephrine, etc, can be demonstrated for a long list of substituted hydrocarbons including the aerosol propellants, fire extinguishing agents, and refrigerants.

Much attention has been directed toward the problem of the genesis of cardiac arrhythmias during exposure to the halogenated alkanes (17). Van Stee & Back (6) demonstrated that in addition to the presence of $CBrF_3$, arrhythmias appearing during exposure to this compound were sensitive to blood pressure, acid-base balance, and pressor amines such as epinephrine and by some mechanism other than their ability to raise blood pressure (see following section).

The interaction between the presence of the halogenated alkanes and epinephrine, the so-called sensitization of the heart to epinephrine, has been thoroughly investigated for $CBrF_3$, $CBrClF_2$, and CCl_2F_2 (18–22). The concern is based on the supposed release of endogenous epinephrine from the adrenal medulla during excitement, fear, or other stressful stimuli. The i.v. infusion of exogenous epinephrine during exposure to the halogenated alkanes does not duplicate the sympathoadrenal activation of the stressful situation. Not only is epinephrine liberated from the adrenal medulla, but also adrenergic neurotransmitter (presumably norepinephrine) is elaborated at the adrenergic terminals of the sympathetic innervation of the heart (as well as other sympathetically innervated structures). Furthermore, the volumes of distribution of endogenous and exogenous catecholamines are not identical.

Hine et al (10) came closest to modeling the physiological situation when they exposed dogs to $CBrF_3$ and then frightened them by means of stroboscopic lights and noise. No dog in this study developed ventricular fibrillation. Van Stee & Back (15) did report the death from ventricular fibrillation of one dog exposed to 40% $CBrF_3$ and not given any additional drugs. Marked excitement accompanied the event.

Beck et al (16) and Clark (13) demonstrated the exogenous epinephrine-$CBrClF_2$ interaction on cardiac arrhythmias in dogs.

Hine et al (10) monitored cardiac electrical activity (EKG) during exposure of human volunteers to nominal concentrations of 5, 10, and 15% of $CBrF_3$. Auriculoventricular (AV) dissociation and premature ventricular contractions were recorded during exposure to the highest concentrations (maximum, 16.9%).

Neither Call (12) nor Smith & Harris (23) detected any cardiac arrhythmias during exposure to $CBrF_3$. Call exposed human volunteers to 4 or 7% of $CBrF_3$ for 3 min in hypobaric chambers. Smith & Harris exposed crews to 5 to 7% for 5 min at pressurized altitudes of 1,000–20,000 feet in aircraft flight tests.

CARDIOVASCULAR PHARMACODYNAMICS

The cardiovascular actions of the fluoroalkanes are considered to represent the most significant hazard incident to their use. Detailed studies of the cardiovascular

dynamic and myocardial metabolic effects of exposure to $CBrF_3$, $CBrClF_2$, or CH_2ClBr have been accomplished.

Exposures of anesthetized monkeys to 80% $CBrF_3$ or 12% $CBrClF_2$ cause a marked decrease in mean blood pressure together with a marked increase in left ventricular end diastolic pressure. This indicates that myocardial performance shifted up the Starling curve to a region approaching a state of compensated heart failure which would constitute a decrease in myocardial contractility (24).

The negative inotropic effects of exposure to the halogenated alkanes have been quantified and compared by Van Stee et al (25). The decreases in such indexes as dP/dt_{max} and dP/dt_{max} divided by developed pressure were comparable during exposure to approximately 1% CH_2ClBr, 12–14% $CBrClF_2$, and 75–80% $CBrF_3$.

The arrhythmias, particularly premature ventricular contractions (PVC), are dependent on a number of factors in addition to the level of fluoroalkane (6). Experiments were conducted to evaluate the sensitivity of the arrhythmias to mean blood pressure changes. In monkeys, PVCs appeared when pressure was elevated by expanding circulating blood volume by the infusion of 6% dextran. PVCs appeared and disappeared when mean arterial pressure was raised and lowered by aortic constriction or when the blood pressure was lowered and raised by exsanguination and reinfusion. Further, it was demonstrated that circulating catecholamine levels and acidosis altered the arrhythmia threshold independently of changes in blood pressure.

Exposure to the fluoroalkanes often caused a reversible, concentration-dependent fall in mean arterial blood pressure. Cross-circulation experiments have been performed in which the blood from donor dogs was used to perfuse the hind limbs of recipient dogs. The hind limbs of the recipients were in vascular isolation from the dogs' general circulation but the autonomic innervation remained intact. Through the use of combinations of recipient and donor dog exposures, coupled with the administration of autonomic drugs, the mechanism of hypotensive response to the fluoroalkanes $CBrF_3$ and $CBrClF_2$ was determined to be a decrease of vasoconstrictor tone. The compounds were found not to have any direct vascular smooth muscle action (26).

A series of experiments was conducted to test the hypothesis that the decrease in vasoconstrictor tone was the result, in part, of an impairment of ganglionic transmission. The vagosympathetic trunk was severed in the midcervical region and the cut ends were stimulated electrically. Nictitating membrane tissue tension was measured during stimulation of the central end, and vagal inhibition of the heart was monitored during stimulation of the peripheral end. $CBrF_3$ but not CH_2ClBr was found to cause a partial ganglionic blockade.

Regarding the cardiovascular dynamic effects, the conclusion was reached that the fall in blood pressure seen during exposure to the fluoroalkanes resulted from a combination of cardiodynamic functional impairment and ganglionic blockade. A reduction of cardiodynamic performance also was seen during exposure to CH_2ClBr; however, since the pressoreceptor reflexes remained functional in the absence of ganglionic blockade, their activation resulted in the maintenance of normal or slightly elevated mean arterial blood pressure during exposure.

The cardiac arrhythmias were sensitive to changes in mean arterial blood pressure, which implied that myocardial afterload was a determinant of the arrhythmia threshold as well as the presence of the halogenated alkanes. Tension on the myocardium has been demonstrated to alter both the electrical and mechanical properties of cardiac muscle (27). This concept may be extended to include muscle preload, and the authors suspect that this variable may affect the arrhythmia threshold as well as after-load. This hypothesis has not been tested but assumes some importance because end diastolic pressure (and presumably end diastolic volume) may rise during exposure to the compounds.

Having determined that the hypotensive effect was primarily the consequence of a decrease in vasoconstrictor tone secondary to ganglionic blockade, a group of experiments was conducted to investigate the mechanism of the negative inotropic effect.

A general procedure was established in which dogs were anesthetized and instrumented for acute exposure to different gas mixtures under anesthesia (28). Forty variables were either measured directly or computed from measured variables to provide a basis for an evaluation of cardiovascular dynamics and myocardial metabolism during exposure of anesthetized dogs to the halogenated alkanes. The measurements included arterial and coronary venous blood levels of O_2, glucose, lactate, pyruvate, nonesterified fatty acids, and the acid-base variables. Cardiovascular dynamic variables were monitored and Stewart-Hamilton indicator-dilution studies were performed using indocyanine green.

Some animals were pretreated with amine-depleting doses of reserpine 24 hr prior to examination. The results of these experiments indicated that the cardiovascular dynamic impairment that occurred during exposure to $CBrClF_2$ was independent of the integrity of the aminergic neural mechanisms. We have interpreted this to mean that the negative inotropic effect of the compound is independent of myocardial adrenergic postsynaptic activity (5, 29).

Likewise, the determination was made that the myocardial effects of exposure to the halogenated alkanes was not the consequence of an altered availability to the myocardium of oxygen and oxidizable substrates. Delivery to, and extraction by, the myocardium of nutrients was not altered significantly.

The significant finding in this series of experiments was that, whereas the myocardium was presented with adequate oxygen, animals exposed to $CBrClF_2$ and CH_2ClBr failed to extract a normal amount. A rise in the oxygen content of coronary sinus blood was measured and was correlated with the concentrations of $CBrClF_2$ or CH_2ClBr to which the animals were exposed (29). These elevations in coronary venous PO_2 and O_2 content persisted for 30 min after exposure to $CBrClF_2$ but not to CH_2ClBr.

No significant differences among the acid-base variables were detected for any of the compounds. Exposure to $CBrF_3$ resulted in significant elevations of arterial and coronary venous glucose levels which persisted for at least 30 min postexposure. Lactate extraction by the myocardium and arterial pyruvate decreased significantly during exposure to $CBrClF_2$, and venous pyruvate decreased significantly during exposure to CH_2ClBr. None of these changes persisted after exposure.

Mean transit time was prolonged significantly during exposure to $CBrClF_2$. The dP/dt_{max} and left coronary circumflex arterial blood flow were decreased significantly during exposure to $CBrClF_2$ and the dP/dt_{max} divided by developed pressure was decreased significantly during exposure to CH_2ClBr.

Myocardial oxygen extraction followed demand during exposures to $CBrF_3$ and $CBrClF_2$. The same two variables, on the other hand, were apparently significantly dissociated during the CH_2ClBr exposures. This could have been the consequence of a slowing of mitochondrial respiration. All three haloalkanes were found to slow state three respiration without uncoupling in isolated rat liver mitochondria (29). The effectiveness of the compounds was $CBrF_3 < CBrClF_2 < CH_2BrCl$. In the absence of measurements of mitochondrial levels of the compounds during the in vivo and in vitro experiments only limited inferences may be made concerning the treatment-response relationship. The absence of the measurable O_2 demand myocardial oxygen extraction dissociation during exposure to $CBrF_3$ or $CBrClF_2$ may only have reflected a failure to achieve mitochondrial levels of the compounds sufficient to affect respiration significantly.

Coronary flow was higher than controls for any given oxygen demand during $CBrF_3$ exposure. This was attributable to the mild hypoxemia that accompanied exposure of open-chested dogs to the relatively high levels of the compounds that displaced a significant amount of oxygen from the inspired gas mixture. The failure of $CBrF_3$ to dissociate O_2 demand and myocardial oxygen extraction, as well as the normal response of coronary flow to mild hypoxemia during the exposure, supports the conclusion that $CBrF_3$ had no significant effect on the relationship between myocardial O_2 demand, myocardial oxygen extraction, and coronary flow in these experiments.

Coronary flow was somewhat higher than controls for any given oxygen demand during $CBrClF_2$ exposure. The element of hypoxia was not present in these experiments; therefore, a coronary vasodilation was implied which may have explained the elevated coronary sinus blood oxygen level. Arterial hyperoxemia has been shown normally not to increase coronary venous PO_2 significantly in conscious, unmedicated, chronically instrumented dogs.

Coronary flow was somewhat lower than controls for any given O_2 demand during CH_2BrCl exposure. Myocardial oxygen extraction was profoundly lower, however. These results implied that this compound, in contrast to $CBrClF_2$, caused little or no coronary vasodilation. The elevated coronary sinus oxygen was apparently the consequence of a marked reduction of myocardial oxygen extraction.

In summary, $CBrF_3$ did not affect significantly the responses of either myocardial oxygen extraction or coronary arterial blood flow to oxygen demand. Neither was the coronary sinus blood oxygen level elevated. $CBrClF_2$, on the other hand, elevated coronary flow which resulted in a delivery of oxygen to the myocardium in excess of demand. The excess appeared in coronary sinus effluent. CH_2BrCl impaired oxygen extraction in response to demand without markedly affecting the coronary flow response which resulted in an elevation of coronary sinus blood oxygen. It is obvious, though the exact mechanisms are not known, that exposure to $CBrF_3$, $CBrClF_2$, or CH_2ClBr may result in disturbances of myocardial energy

metabolism that are connected to myocardial performance. Based on the concentration to which dogs must be exposed to elicit such responses, $CBrF_3$ was least effective and CH_2ClBr was most effective. The $CBrClF_2$ was intermediate between the two but closer to the CH_2ClBr than to the $CBrF_3$ (5, 29).

GENERAL METABOLISM

Paulet et al (30) reported the results of studies on the effects of CCl_2F_2 and CCl_3F on general metabolism in anesthetized rats and rabbits. Single exposures to CCl_3F for 20 min or 3 hr exposures repeated daily for 15 days produced transitory increases in blood glucose and lactate concentrations that were rapidly reversed following cessation of exposure. CCl_3F also caused a general metabolic depression reflected in decreased O_2 consumption. CCl_2F_2 was without significant effects in these experiments.

Dittmann & Etschenberg (31) compared the ability of a series of halogenated methanes to desensitize pulmonary stretch receptors (endoanesthesia) and to produce narcosis in guinea pigs. The relative effectiveness of CCl_3F and CCl_2F_2 in these experiments was the same as that reported by Paulet et al (30), i.e. $CCl_3F > CCl_2F_2$.

At least two factors contribute to the relationship of biological activities established in the studies by Paulet et al (30) and Dittman & Etschenberg (31). 1. CCl_3F is nine times as soluble in olive oil and four times as soluble in serum as CCl_2F_2 (32). The expectation that CCl_3F would be absorbed more readily than CCl_2F_2 during inhalation exposure was borne out by the studies of Adir et al (33) who determined that 77% of a given dose of CCl_3F and 55% of CCl_2F_2 was absorbed by conscious men and anesthetized dogs. Furthermore, CCl_3F was retained longer than CCl_2F_2 owing to their relative lipid solubilities. 2. Fluorination usually decreases pharmacologic potency and increases the chemical stability of halogenated alkanes. Increased potency in a homologous series usually is accompanied by increased lipid solubility (34).

Van Stee et al (5) monitored myocardial metabolism in anesthetized dogs exposed to $CBrF_3$ and $CBrClF_2$. Four to 12% $CBrClF_2$ increased coronary venous PO_2 and O_2 content whereas 27–75% $CBrF_3$ did not. The effect of $CBrClF_2$ was partially reversed by 30 min after cessation of 90-min exposures. $CBrClF_2$ is 12 times as soluble in olive oil as $CBrF_3$ (24). The relative potencies of $CBrF_3$ and $CBrClF_2$ bore the same relationship to the degree of fluorination and lipid solubility as the relationship between CCl_2F_2 and CCl_3F.

A possible functional basis for the effects of $CBrF_3$ on myocardial metabolism was revealed by the studies of McNutt et al (35) who reported on exposures of free-roaming guinea pigs to 79% of $CBrF_3$ in O_2. Hearts were rapidly fixed in situ by perfusion with glutaraldehyde. Mitochondrial cristae were in the energized configuration in control animals and in the orthodox configuration in $CBrF_3$-exposed animals. No evidence of mitochondrial hypoxia was observed and the suggestion was made that inner mitochondrial membrane-dependent functions may have been compromised in the presence of $CBrF_3$.

A hyperglycemic action of CCl_3F was noted by Paulet et al (30) and of $CBrF_3$ by Van Stee et al (5). Along similar lines, decreased glucose tolerance during halothane ($CHBrCl–CF_3$) anesthesia has been observed in dogs. This was ascribed to an inhibition of insulin secretion in response to an acute glucose load (36). Gingerich et al (37) supported this conclusion with studies on isolated rat pancreas. A structural basis for the possible activity relationship among this group of compounds is not immediately apparent.

UPTAKE AND DISTRIBUTION

Azar et al (38) measured carotid arterial and jugular venous blood levels of CCl_2F_2 and CCl_3F during 10-min exposures of conscious dogs. Consistent arteriovenous differences reflected general tissue uptake during the exposures. The average arteriovenous levels commonly associated with epinephrine-induced arrhythmogenesis were 35 and 23 μg/ml, respectively, for CCl_2F_2 and 29 and 20 μg/ml, respectively, for CCl_3F. The relationships among degree of fluorination, lipid solubility, and pharmacologic potency described in the section on general metabolic effects were preserved for this action (32, 34). In this connection, maximal blood levels of the respective compounds attained by human subjects using pressurized bronchodilator dispersers, were a small fraction of those required to elicit epinephrine-induced cardiac arrhythmias in dogs (39, 40).

Venous blood, whole heart, and whole brain levels of $CBrF_3$ were measured serially during and after 5-min exposures of rats to 75% $CBrF_3$ in O_2. Blood and heart levels were not significantly different and brain levels rose to 50% higher than blood levels. These observations supported the conclusion that venous blood levels provided a reasonable index of myocardial tissue levels and that the compound was preferentially distributed to an organ of high lipid content (41). Exposure of dogs and monkeys to similar concentrations of $CBrF_3$ has been reported to elicit cardiac arrhythmias during intravenous epinephrine infusion (15).

The postexposure decline of blood levels of $CBrF_3$, CCl_2F_2, and CCl_3F was biphasic reflecting a rapid initial washout from the central compartment (blood) followed by transition to a slower phase representing delivery of the compounds from the peripheral compartment (extravascular tissues) to the central compartment. The rapidity of the washout increased with decreasing lipid solubility of the compounds and was essentially complete, even after prolonged exposures, within 2 hr after exposure (5, 33).

Chiou & Hsiao (42) demonstrated that CCl_2F_2, CCl_3F, and $CClF_2–CClF_2$ were preferentially distributed to human and bovine serum albumin in phosphate buffer solution in the same rank order as their lipid solubilities, i.e. (from least to greatest) CCl_2F_2, 51%; CCl_3F, 66–71%; and $CClF_2–CClF_2$, 73–79%.

BIOTRANSFORMATION

Recent reports indicate that a number of systems capable of mediating the enzymatic dehalogenation of low molecular weight halogenated compounds may be

present in tissues, particularly in the liver. For example, the reductive dechlorination of CCl_4 to $CHCl_3$ was reported by Paul & Rubinstein (43). The oxidative dechlorination of $CHCl_2$–CH_2Cl by the mixed-function oxidase (MFO) system was characterized by Van Dyke & Gandolfi (44). Evidence consistent with the notion that the glutathione-S-transferases (45) of the 105,000 X G supernatant fraction of homogenized liver may be involved in the defluorination of methoxyflurane was suggested by Warren et al (46). Kubic et al (47) reported that CH_2Cl_2, CH_2I_2, and CH_2ClBr, respectively, were metabolized to CO by a hepatic system neither stimulated by phenobarbital nor inhibited by SKF 525-A. Several other halogenated methanes including CCl_2F_2 did not yield CO.

Although many potential routes for the dehalogenation of fluorinated alkanes apparently exist, most evidence suggests that the biotransformation of these compounds is of little significance quantitatively to their disposition. On the other hand, as is discussed later in connection with $CHClF_2$ and $CH_2{=}CCl_2$, limited biotransformation could be of substantial biological significance with respect to manifestations of toxicity.

Jenkins et al (48) exposed rats and guinea pigs continuously for 90 days and intermittently for 8 hr/day, 5 days/wk for six weeks to 100 ppm CCl_3F. Urinary F^- excretion and serum F^- levels did not differ significantly from those of controls. Although no evidence for the defluorination of CCl_3F was presented in that study, the report of Cox et al (49) suggested that caution be exercised before dismissing altogether the possibility and its potential biological significance. They observed Type I binding spectra not unlike those reported for CCl_4 which could represent the first step in an interaction of CCl_3F with the MFO system already known to possess dehalogenation activity.

Studies on the metabolism of $^{14}CCl_3F$ and $^{14}CCl_2F_2$ in man and the dog have been reported (50, 51). Exposures to 1,000–12,000 ppm of the compounds in air for $<$ 20 min resulted in virtually complete recovery of all inspired fluoroalkanes. The possibility that trace amounts of metabolites may have been formed could not be confirmed.

The sum of evidence presented so far indicates that very little, if any, CCl_3F or CCl_2F_2 is defluorinated. Studies of possible dechlorination have not been reported. The apparent lack of biochemical reactivity is, no doubt, the consequence of the stabilization of adjacent carbon–halogen bonds by the successive fluorination of these compounds (34).

There is no doubt that animals are able to metabolize bromine from CH_2ClBr. MacEwen et al (52) exposed rats and dogs to CH_2ClBr for 124 6-hr exposures over a six-month period. The levels of 500 and 1000 ppm produced only a slight depression in rat growth, and lethargy was noted. The most significant finding was blood bromide levels in both species in excess of 95 mg/100 ml blood. This probably accounted for the CNS depression noted.

The matter of quantitatively limited biotransformation and toxicologic response to the presence of trace quantities of metabolites formed is worthy of careful consideration. Speizer et al (53) reported an increased incidence of palpitations and supraventricular arrhythmias among workers preparing frozen sections and exposed

to $CHClF_2$. Exposures were to low levels and repeated at intervals over a period of years. The disturbing implication of this report was that the arrhythmogenic propensity appeared to be cumulative, with residual effects suggesting that the proximate arrhythmogen was probably a product of the biotransformation of the $CHClF_2$. This is the first report suggesting long-term residual effects on cardiac rhythm associated with chronic, low level exposure to halogenated alkanes. In this connection, it is of interest to note that pretreatment with phenobarbital, which induces the MFO system, reduced the amount of epinephrine required to trigger cardiac arrhythmias in rats exposed to $CH_2{=}CCl_2$ (54).

CONCLUSIONS

The most important toxicological effects of the haloalkanes are on the central nervous and cardiovascular systems. The neurological effects are manifested as alterations of perception and a reduction in reaction time and the ability to concentrate on complex intellectual tasks. The cardiovascular effects are manifested as changes in cardiovascular dynamics and the electrical activity of the heart.

Clinically important central nervous system effects almost always appear at lower levels of exposure than clinically important cardiovascular effects. Behavioral changes and performance decrements during exposure would undoubtedly have some effect on the interaction of the subject with his environment and such consequences of exposure could be life-threatening. Likewise, certain manifestations of halogenated alkane toxicity, such as the occurrence of cardiac arrhythmias, also constitute readily identifiable hazards.

Literature Cited

1. Clayton, J. W. Jr. 1967. Fluorocarbon toxicity and biological action. *Fluorine Chem. Rev.* 1:197–252
2. Aviado, D. 1974. Toxicity of propellants. *Prog. Drug Res.* 18:365–97
3. Engibous, D. L., Torkelson, T. R. 1960. A study of vaporizable extinguishants. *WADC Tech. Rep. 59-463,* Wright Air Dev. Div., Wright-Patterson AFB, Ohio
4. Svirbely, J. L., Highman, B., Alford, W. C., von Oettingen, W. F. 1947. The toxicity and narcotic action of monochloro-mono-bromomethane with special reference to inorganic and volatile bromide in blood, urine and brain. *J. Ind. Hyg. Toxicol.* 29:382–89
5. Van Stee, E. W., Harris, A. M., Horton, M. L., Back, K. C. 1975. The effects of three vaporizable fire extinguishing agents on myocardial metabolism and cardiovascular dynamics in the anesthetized dog. *Toxicol. Appl. Pharmacol.* 34:62–71
6. Van Stee, E. W., Back, K. C. 1971. Brain and heart accumulation of bromotrifluoromethane. *AMRL-TR-70-139,* Aerosp. Med. Res. Lab., Wright-Patterson AFB, Ohio
7. Chikos, P. M., Van Stee, E. W., Back, K. C. 1969. Central nervous system effects of bromotrifluoromethane. *AMRL-TR-69-130.* Aerosp. Med. Res. Lab., Wright-Patterson AFB, Ohio
8. Carter, V. L., Back, K. C., Farrer, D. N. 1970. The effect of bromotrifluoromethane on operant behavior in monkeys. *Toxicol. Appl. Pharmacol.* 17:648–55
9. Van Stee, E. W., Back, K. C., Prynn, R. B. 1970. Alteration of the electroencephalogram during bromotrifluoromethane exposure. *Toxicol. Appl. Pharmacol.* 16:779–85
10. Hine, C. H., Elliott, H. W., Kaufman, J. W., Leung, S., Harrah, M. D. 1968. Clinical toxicologic studies on freon FE 1301. *Proc. Ann. Conf. Atmos. Contam. Confined Spaces, 4th, AMRL-TR-68-175,* Aeros. Med. Res. Lab., Wright-Patterson AFB, Ohio

11. Haskell Lab. Toxicol. Ind. Med. 1967. Human exposures to freon FE 1301. *Tech. Rep.,* E. I. DuPont De Nemours, Inc., Wilmington, Delaware
12. Call, D. W. 1973. A study of halon 1301 ($CBrF_3$) toxicity under simulated flight conditions. *Aerosp. Med.* 44:202–4
13. Clark, D. G. 1970. The toxicity of bromochlorodifluoromethane (BCF) to animals and man. *Tech. Rep.,* Imperial Chemical Industries, Ltd., Ind. Hyg. Res. Labs., Alderley Park, Cheshire, England
14. Rutstein, H. R. 1962. Acute chlorobromomethane toxicity. *Arch. Environ. Health* 7:440–44
15. Van Stee, E. W., Back, K. C. 1969. Short-term exposure to bromotrifluoromethane. *Toxicol. Appl. Pharmacol.* 15:164–74
16. Beck, P. S., Clark, D. G., Tinston, D. J. 1973. The pharmacologic actions of bromochlorodifluoromethane (BCF). *Toxicol. Appl. Pharmacol.* 24:20–29
17. Chenoweth, M. B. 1946. Ventricular fibrillation induced by hydrocarbons and epinephrine. *J. Ind. Hyg. Toxicol.* 28:151–58
18. Mullin, L. S., Azar, A., Reinhardt, C. F., Smith, P. E., Fabryka, E. F. 1972. Halogenated hydrocarbon-induced cardiac arrhythmias associated with release of endogenous epinephrine. *Am. Ind. Hyg. Assoc. J.* 33:389–96
19. Reinhardt, C. F., Azar, A., Maxfield, M. E., Smith, P. E., Mullin, L. S. 1971. Cardiac arrhythmias and aerosol "sniffing." *Arch. Environ. Health* 22: 265–69
20. Reinhardt, C. F., Mullin, L. S., Maxfield, M. E. 1973. Epinephrine-induced cardiac arrhythmia potential of some common industrial solvents. *J. Occup. Med.* 15:953–55
21. Clark, D. G., Tinston, D. J. 1971. The influence of fluorocarbon propellants on the arrhythmogenic activities of adrenaline and iseprealine. *Proc. Eur. Soc. Study Drug Toxicity Meet., XIII,* Berlin
22. Taylor, G. J., Harris, W. S., Bogdonoff, M. D. 1971. Ventricular arrhythmias induced in monkeys by the inhalation of aerosol propellants. *J. Clin. Invest.* 50:1546–50
23. Smith, D. G., Harris, D. J. 1973. Human exposure to halon 1301 ($CBrF_3$) during simulated aircraft cabin fires. *Aerosp. Med.* 44:198–201
24. Van Stee, E. W. 1974. A review of the toxicology of halogenated fire extinguishing agents. *AMRL-TR-74-143,* Aerosp. Med. Res. Lab., Wright-Patterson AFB, Ohio
25. Van Stee, E. W., Diamond, S. S., Harris, A. M., Horton, M. L., Back, K. C. 1973. The determination of the negative inotropic effect of exposure of dogs to bromotrifluoromethane and bromochlorodifluoromethane. *Toxicol. Appl. Pharmacol.* 26:549–58
26. Van Stee, E. W., Back, K. C. 1972. The mechanism of the peripheral vascular resistance change during exposure of dogs to bromotrifluoromethane. *Toxicol. Appl. Pharmacol.* 23:428–42
27. Penefsky, Z. J., Hoffman, B. F. 1963. Effects of stretch on mechanical and electrical properties of cardiac muscle. *Am. J. Physiol.* 204:433–38
28. Van Stee, E. W., Horton, M. L., Harris, A. M., Back, K. C. 1973. The effect of 90-minute exposure to bromotrifluoromethane on myocardial metabolism in the dog. *Proc. Ann. Conf. Environ. Toxicol., 4th, AMRL-TR-72-125,* Aerosp. Med. Res. Lab., Wright-Patterson AFB, Ohio
29. Van Stee, E. W., Harris, A. M., Horton, M. L., Back, K. C. 1974. Toxic hazards evaluation of new air force extinguishing agents. *Proc. Ann. Conf. Environ. Toxicol., 5th, AMRL-TR-74-125,* Aerosp. Med. Res. Lab., Wright-Patterson AFB, Ohio
30. Paulet, G., Roncin, G., Vidal, E., Toulouse, P., Dassonville, J. 1975. Fluorocarbons and general metabolism in the rat, rabbit, and dog. *Toxicol. Appl. Pharmacol.* 34:197–203
31. Dittmann, E. C., Etschenberg, E. 1973. Endoanesthetic and narcotic activity in halogenated methane derivatives. *Eur. J. Pharmacol.* 24:389–98
32. Morgan, A., Black, A., Walsh, M., Belcher, D. R. 1972. The absorption and retention of inhaled fluorinated hydrocarbon vapours. *J. Appl. Radiat. Isot.* 23:285–91
33. Adir, J., Blake, D. A., Mergner, G. W. 1975. Pharmacokinetics of fluorocarbon 11 and 12 in dogs and humans. *J. Clin. Pharmacol.* 15:760–70
34. Krantz, J. C. Jr., Rudo, F. G. 1966. The fluorinated anesthetics. *Handbook of Experimental Pharmacology,* Part I, *Pharmacology of Fluorides,* ed. F. A. Smith, Chap. 10, pp. 501–64. New York: Springer-Verlag
35. McNutt, N. S., Morris, F., Van Stee, E. W. 1973. Ultrastructure of guinea pig heart after exposure to fluorocarbon 1301. *AMRL-TR-73-125,* Aerosp.

Med. Res. Lab., Wright-Patterson AFB, Ohio
36. Camu, F. 1973. Carbohydrate intolerance during halothane anesthesia in dogs. *Acta Anaesthesiol. Belg.* 24: 180–88
37. Gingerich, R., Wright, P. H., Paradise, R. R. 1974. Inhibition by halothane of glucose-stimulated insulin secretion in isolated pieces of rat pancreas. *Anesthesiology* 40:449–52
38. Azar, A., Trochimowicz, H. J., Terrill, J. B., Mullin, L. S. 1973. Blood levels of fluorocarbon related to cardiac sensitization. *Am. Ind. Hyg. J.* 34:102–9
39. Dollery, C. T., Draffan, G. H., Davies, D. S., Williams, F. M., Conolly, M. E. 1970. Blood concentrations in man of fluorinated hydrocarbons after inhalation of pressurised aerosols. *Lancet* 2:1164–66
40. Paterson, J. W., Sudlow, M. F., Walker, S. R. 1971. Blood-levels of fluorinated hydrocarbons in asthmatic patients after inhalation of pressurized aerosols. *Lancet* 2:565–68
41. Van Stee, E. W., Back, K. C. 1971. Brain and heart accumulation of bromotrifluoromethane. *AMRL-TR-70-139,* Aerosp. Med. Res. Lab., Wright-Patterson AFB, Ohio
42. Chiou, W. L., Hsiao, J. H. 1974. A new simple approach to study the protein binding of volatile and gaseous compounds. I. Fluorocarbon aerosol propellants, halothane and cyclopropane. *Res. Commun. Chem. Pathol. Pharmacol.* 8:273–87
43. Paul, B. B., Rubinstein, D. 1963. Metabolism of CCl_4 and $CHCl_3$ by the rat. *J. Pharmacol. Exp. Ther.* 141:141–48
44. Van Dyke, R. A., Gandolfi, A. J. 1975. Characteristics of a microsomal dechlorination system. *Mol. Pharmacol.* 11:809–17
45. Jerina, D. M., Bend, J. R. 1976. Glutathione-S-transferase. In *IUPHAR Satell. Symp. Act. Intermed.: Form., Toxicity Inactivation,* July 26–27, Turku, Finland
46. Warren, W. A., Baker, F. D., Bellantoni, J. 1976. Enzymatic defluorination of methoxyflurane. *Biochem. Pharmacol.* 25:723–24
47. Kubic, V. L., Anders, M. W., Engel, R. R., Barlow, C. H., Caughey, W. S. 1974. Metabolism of dihalomethanes to carbon monoxide. *Drug Metab. Dispos.* 2:53–57
48. Jenkins, L. J. Jr., Jones, R. A., Coon, R. A., Siegel, J. 1970. Repeated and continuous exposures of laboratory animals to trichlorofluoromethane. *Toxicol. Appl. Pharmacol.* 16:133–42
49. Cox, P. J., King, L. J., Parke, D. V. 1972. A study of the possible metabolism of trichlorofluoromethane. *Proc. Biochem. Soc.* 130:13–14
50. Blake, D. A., Mergner, G. W. 1974. Inhalation studies on the biotransformation and elimination of [^{14}C] trichlorofluoromethane and [^{14}C] dichlorodifluoromethane in beagles. *Toxicol. Appl. Pharmacol.* 30:396–407
51. Mergner, G. W., Blake, D. A., Helrich, M. 1975. Biotransformation and elimination of ^{14}C-trichlorofluoromethane (FC-11) and ^{14}C-dichlorodifluoromethane (FC-12) in man. *Anesthesiology* 42:345–51
52. MacEwen, J. D., McNerney, J. M., Vernot, E. H., Harper, D. T. 1966. Chronic inhalation toxicity of chlorobromomethane. *J. Occup. Med.* 8:251–56
53. Speizer, F. E., Wegman, D. H., Ramirez, A. 1975. Palpitation rates associated with fluorocarbon exposure in a hospital setting. *N. Engl. J. Med.* 292:624–26
54. Siletchnik, L. M., Carlson, G. P. 1974. Cardiac sensitizing effects of 1,1-dichloroethylene: Enhancement by phenobarbital pretreatment. *Arch. Int. Pharmacodyn.* 210:259–364