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TOXICITY OF CERTAIN HYDRAULIC FLUIDS AND CERTAIN OF THEIR CONSTITUENTS

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FOREWORD

The investigation reported herein was performed in The Kettering Laboratory in the Department of Preventive Medicine and Industrial Health, College of Medicine, University of Cincinnati, Cincinnati, Ohio, under Contract No. 33(038)26456 with the Aero Medical Laboratory, Directorate of Research, Wright Air Development Center, Wright-Patterson Air Force Base, Ohio.

The work was carried out under RDO No. 698-31, "Toxicity of Developmental Air Force Materials", with George Kitzes, Ph. D., as control monitor.

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This work was done and the report prepared under the general supervision of Francis F. Heyroth, M.D., Director of the Division of Toxicology in the Department of Preventive Medicine and Industrial Health, College of Medicine, University of Cincinnati. The report was edited and approved by Robert A. Kehoe, M.D., Director of the Department.

The experiments reported herein were performed according to the "Rules Regarding Animal Care" as approved and adopted by the American Medical Association.

ABSTRACT

Ethyl dibromobenzene and its formulations (PRL-3169) and (PRL-3209) are only slightly toxic when given undiluted in a single oral dose to animals, whereas tetrachlorotetrafluoropropane is moderately toxic. The brominated material is considered to be non-toxic, for all practical purposes, when it is applied upon the intact skin of rabbits in one dose and maintained in contact therewith over a period of 24 hours.

It appears that the undecomposed mist of either ethyl dibromobenzene or PRL-3209 is more toxic than comparable mists of certain aliphatic esters, but the reverse appears to be true of the fogs formed by the thermal decomposition of these materials.

The vapor of tetrachlorotetrafluoropropane is more toxic than that of the more common chlorinated hydrocarbon solvents. The vapor of chloro-1-(trifluoromethyl)-4-(chlorotetrafluoroethyl)-benzene, although moderately toxic, is somewhat less so than that of tetrachlorotetrafluoropropane.

PUBLICATION REVIEW

This report has been reviewed and is approved.

FOR THE COMMANDER:

JACK BOLLERUD
Colonel, USAF (MC)
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INTRODUCTION

The purposes of the work reported herein were to determine (1) the immediate toxicity of ethyl dibromobenzene and of 2 formulations, PRL-3169 and PRL-3209, containing ethyl dibromobenzene as their principal ingredient, and of 2 other halogenated hydrocarbons, namely, tetrachlorotetrafluoropropane and chloro-1-(trifluoromethyl)-4-(chlorotetrafluoroethyl)-benzene, when given orally to rabbits and rats; (2) the immediate toxicity of ethyl dibromobenzene or its formulations, when maintained upon the intact or abraded skin of rabbits for 24 hours; (3) the immediate toxicity of each of the 2 chlorinated (and fluorinated) hydrocarbons, when inhaled by animals of several species; (4) the toxic effects induced by the mists of ethyl dibromobenzene or of formulation PRL-3209 when inhaled by animals of several species for 7 hours each of 10 days over a period of 2 weeks; and (5) the immediate toxicity of ethyl dibromobenzene and of PRL-3209 when inhaled as fogs formed by dropping upon an Inconel surface maintained at an elevated temperature. In fulfilling these purposes the symptomatic responses of the animals were observed and recorded, and the gross and microscopic changes in the tissues of the exposed animals were noted and described.

PROPERTIES OF MATERIALS

Ethyl dibromobenzene, also coded as PRL-3191 by WADC and an Alkazene 42 by Dow Chemical Company, is a colorless fluid with a boiling range of 258° - 266° C and freezing range of -43° to -38° C. It is reported to have a refractive index of 1.587, a viscosity of 1.2 centipoises at 25° C, and a specific gravity of 1.73 - 1.75 at 25° C. It is infinitely soluble in acetone, benzene, carbon tetrachloride, ether or methanol, but is practically insoluble in water. Since the properties of the isomers having the molecular weight of 263.976, are not available, the carbon atoms to which the atoms of bromine are attached in the structure of the ring cannot be designated.

The following formulation has been termed PRL-3169 by WADC:

Ethyl dibromobenzene	82.5%
Aroclor 1248	10.1%
Acryloid HF-55	3.9%
Mineral oil base stock (4 centistokes at 100° F)	3.1%
Paranox 441	0.4%

The viscosity of this yellow fluid is 8.1 centistokes at 100° F, and 300 centistokes at -40° F. Its density at 100° F is 1.6.

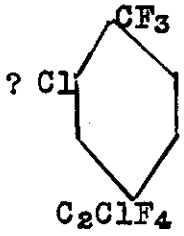
Formulation PRL-3209 is a brown fluid of the following composition:

Ethyl dibromobenzene	79.4%
Aroclor 1248	9.7%
Acryloid HF-55	3.6%
Calcium cetyl phenate (60-70%) (active in mineral oil)	3.0%
Mineral oil base	2.9%
Tricresyl phosphate (mixed isomers)	1.0%
Paranox 441	0.4%

The ethyl dibromobenzene and the 2 formulations containing it were supplied by WADC.

Tetrachlorotetrafluoropropane (F - C(F)(Cl) - C(Cl)(Cl) - C(F)(Cl) - F) (1,1,3,3-tetrafluoro-1,2,2,3-tetrachloropropane) is a colorless liquid which boils at 113° C and freezes at -110 C. This compound has a calculated molecular weight of 253.858, a density of 1.72 g/ml, and vapor pressures of 44 and 414 mm of mercury, respectively, at 38° and 93° C. From the interpolated vapor pressure of 24.5 mm at 25° C, it may be calculated that air in equilibrium with the material at 25° C and 745 mm of mercury would contain 334.5 mg per liter. The material supplied by Wright-Patterson Air Base was prepared in the chemical laboratory of a university and was identified by their Code Number P 441.

Chloro-1-(trifluoromethyl)-4-(chlorotetrafluoroethyl)-benzene is a colorless liquid having a boiling point of 185° C and a freezing point of -70° C. This compound has the molecular weight of 315.028 and a density of 1.57 g/ml.



Air bearing the least of the concentrations employed in these experiments, 307.2 ppm, had a faint odor. The material was provided by Wright-Patterson Air Force Base where it was given the code identification ME-3-721.

EXPERIMENTAL METHODS

Oral Administration of a Single Dose

Measured amounts of the undiluted material were introduced into the stomachs of healthy female rabbits from an hypodermic syringe by means of a rubber catheter (F12) passed through the esophagus. Tetrachlorotetrafluoropropane was given as a 1 to 8 per cent V/V solution in peanut oil. To make certain that the entire dose entered the stomach, the tube was flushed with 4 or 5 ml of water. Rats were given the undiluted material, by displacement from a syringe through a blunt, 6 cm, 17-gauge hypodermic needle inserted into the esophagus. The animals were weighed daily until any losses in weight sustained in association with the administration of the material had been regained, after which they were weighed once per week until they were killed.

Application Upon the Skin of Rabbits

In accordance with the technique of Draize, Woodard and Calvery, the dosage of 9.4 ml of undiluted ethyl dibromobenzene, PRL-3169 and PRL-3209 per kg of body weight was maintained for 24 hours in contact with the intact skin of each of 3 rabbits, in each of 3 groups, respectively. Pursuant to this method, the hair was clipped from an area of skin 6 to 7 inches wide and completely encircling the trunk of each rabbit. The entire trunk was covered with a sleeve of dental dam, which fitted tightly at the cephalad and caudad edges

of the clipped area and loosely over the intervening area. During the 24-hour period of contact the rabbits were kept in stocks. At the end of 24 hours, the material was removed from the skin of the animals by washing with water and a sulfonated oil, "pH6".

By the same means, a like dosage of each of these materials was maintained in contact with the abraded skin of corresponding groups of rabbits. The outermost layer of the skin of these rabbits was abraded by scratching thin furrows 7.5 cm in length and a few millimeters apart, with an hypodermic needle.

When a death occurred in any group of 3 rabbits, a lesser dosage of 6.0 or 3.2 ml per kg was applied upon the skin of additional rabbits.

Respiratory Exposure to Vapor

Groups of experimental animals, each consisting usually of a cat, 2 guinea pigs, 2 rabbits and 4 rats, were introduced into an atmosphere containing the vapor of tetrachlorotetrafluoropropane, within a 223-liter cylindrical steel chamber coated on the inner surface with a chemical-resistant plastic material (WADC Technical Report No. 54-344 on Lubricants). Their behavior could be observed through a sheet of a clear plastic, which sealed the open front end of the chamber, an interposed heavy wire mesh preventing the animals from breaking the seal.

By means of a moving, tightly-fitting glass plunger (11 mm in diameter), liquid tetrachlorotetrafluoroethane was forced from a long glass tube of uniform diameter (Figure 1) through a sealed-on capillary tube into a glass receptacle, where it was allowed to evaporate into a stream of air passing at a measured rate (16.3 l per min as measured by a rotameter) into the chamber. The plunger was actuated by a synchronous motor through a set of gears and a screw drive. The rate of flow of liquid was varied as needed from experiment to experiment by altering the gear-ratio. To facilitate the evaporation of the liquid into the air stream, the liquid was allowed to drop from the tip of the capillary tube upon a layer of glass wool in a tube leading to the chamber. The exterior of the tube was heated electrically to a slightly elevated temperature by passing a current through nichrome wire, which was connected in series with a Variac.

Similar groups of animals, which differed only in the occasional inclusion of a cat or hamster or both, were subjected to exposure to air bearing the vapor of chloro-1-(trifluoromethyl)-4-(chlorotetrafluoroethyl)-benzene. The air-borne vapor was generated in the manner described above, but the air stream entered the chamber at a rate of either 17 or 21 l per minute.

Respiratory Exposure to a Mist Formed by Aspiration at Room Temperature

Mists of ethyl dibromobenzene or PRL-3209 were generated by use of a glass injector, as described in detail in "The Toxicity of Sulfuric Acid Mist", by Treon, Dutra, Cappel, Sigmon and Younker. In the preparation of air-borne mists, a stream of compressed air at a pressure of either 1.3 or 3.4 atmospheres was passed over an orifice at the end of a tube, the lower portion of which was immersed in an Erlenmeyer flask containing the desired liquid. The mist-laden air entered the chamber at the rate of either 18.6 or 47 liters per minute (Table 6), as measured at room temperature and atmospheric pressure. The larger particles impinged on a mantle and returned to the Erlenmeyer flask. The amounts of material dispersed per minute (recorded in Table 6) were determined by weighing the flask before and after every 7-hour period. In some of the experiments supplementary air from Gast Rotary Blast and Suction pumps by-passed the aspirator and entered the chamber at the same site as the air-borne mist. Samples of the mist of PRL-3209 from the chamber were collected on small circular glass slides mounted in a thermal precipitator. Photographs of the particles at a magnification of 1000 showed that most of them were 2 to 3 microns in diameter.

Respiratory Exposure to a Fog Generated by Contact with an Heated Inconel Surface

A rotameter for measuring the flow of air, a furnace for volatilizing and decomposing ethyl dibromobenzene or PRL-3209, a semi-potentiometer, a cooling manifold, and a 223-liter chamber for exposing the animals were illustrated in Figure 4 of WADC Technical Report No. 54-344 on Lubricants. In these experiments, the smaller rotameter to the right in the photographs, was not employed, and all air (31.8 liters per minute) that entered the chamber was passed through the furnace. The glass cooling coils were packed in the bath shown and surrounded by crushed ice.

The air supplied to the furnace by one or two Gast Rotary Blast and Suction pumps was dried in passing through a series of 6 towers containing, respectively, (1) concentrated sulfuric acid, (2) air, (3) glass wool, (4) calcium chloride, (5) sodium hydroxide, calcium chloride and Drierite, and (6) activated charcoal.

The body of the furnace consisted of a 26-inch length of cold-drawn, seamless Inconel tubing, having an outside diameter of 1.5 inches and a wall thickness of 0.049 inch. The liquid was dropped from a bottle in which it was subjected to a constant pressure, through a graduated glass orifice (rather than from the burette shown in Figure 4 of WADC Technical Report No. 54-344 on Lubricents), through an Inconel tube welded to the upper surface of the furnace near its midpoint. This side tube had a length of 6 inches, an outside diameter of 0.675 inch, and a wall thickness of 0.091 inch. Another piece of Inconel tubing 3 inches in length, which had an outside diameter of 0.438 inch, and a wall thickness of 0.049 inch, was also welded to the upper portion of the furnace tube, to permit the insertion of a thermocouple in such a manner as to measure the temperature at the upper interior surface of the furnace. The distance from center to center of the 2 smaller tubes, which were parallel to one another and perpendicular to the furnace proper, was 1.75 inches.

The outside of the furnace was wrapped with 2 layers of molding mica. The middle 10-inch portion of the length of furnace was electrically heated by means of 20 feet (9 wrappings to the inch) of AWG Number 20 Tophet "C" wire, which has a resistance of 0.659 ohm per foot at 68° F. The wire was insulated with approximately 0.75 inch of diatomaceous earth, and wrapped on the outside with asbestos cloth.

The termperature of the wall was measured by an iron-constantan thermocouple attached to a semi-potentiometer which was calibrated against a student potentiometer. The thermocouple was in contact with the upper part of the furnace about 1 inch downstream from the point at which the droplets of material fell upon its lower interior surface.

After the furnace had been brought to the desired temperature (either 700° F /371° C/ or 900° F /482° C/),

ethyl dibromobenzene was dropped through the glass orifice at the desired rate within the range of 98.8 to 1578 mg per minute (Tables 7 and 8). The range of delivery rates of PRL-3209 was 35.5 to 135.8 mg per minute when the temperature of the furnace was 700° F (Table 9).

SAMPLING AND ANALYSIS OF CONTAMINATED AIR

The Determination of Tetrachlorotetrafluoropropane in Air

The method employed for the determination of the vapor of tetrachlorotetrafluoropropane was based upon the ability of that compound to form an orange-red color in an alkaline pyridine medium. This color reaction, which is given by several other chlorinated organic compounds, was first employed by Fujiwara for the determination of chloroform.

The air from the chamber was analyzed from 1 to 4 times during each period of exposure, depending upon the duration of the latter. Each sample of air was collected in an evacuated glass balloon holding either 2.100 or 11.798 liters, chosen in accordance with the amount of halogenated hydrocarbon that was believed to be present. When the smaller bulb was employed, the sample was absorbed in 50 ml of pyridine, and when the larger volume was collected it was absorbed in 100 ml of pyridine. In either case, the pyridine was released into the balloon from an auxiliary bulb by opening an interposed stopcock.

From 1 to 5 ml of the pyridine solution were added to a test tube containing 5 ml of 5 N sodium hydroxide, and a "cold finger" was inserted into the top of the tube. If less than 5 ml of the pyridine solution was employed, sufficient uncontaminated pyridine was added so that the total volume amounted to 5 milliliters.

The tubes were kept for 10 minutes in a bath of water at 95° C, after which they were placed immediately in an ice bath and allowed to remain there for 10 minutes. The pyridine layer was filtered through a small funnel with a fritted glass disc. The first 2 ml were discarded and the remaining filtrate was run directly into a 10 mm cell. The depth of the color was measured

immediately at 500 mμ by means of a photoelectric spectrophotometer, which was set at 100 per cent transmission with a blank pyridine solution prepared in a similar manner with no tetrachlorotetrafluoropropane present. Figure 2 depicts the variation in transmission at 500 mμ with concentration over the range of 0.4 to 4.0 mg per 5 ml of pyridine. Although the data appear to obey the Beer-Lambert Law with a moderate degree of accuracy, too few observations were made to establish precisely the relationship between absorption and concentration, and the values for the atmospheric concentrations may be somewhat inaccurate. However, they were of the order of magnitude anticipated from our measurements of the air flow and the volume of liquid delivered in the separate experiments.

The Determination of Chloro-1-(Trifluoromethyl)-4-(Chlorotetrafluoroethyl)-Benzene in Air

The determination of the concentration of this vapor in air was based upon the ability of an alcoholic solution of the compound to absorb radiations 2720 Å. The ultraviolet transmission curves of alcoholic solutions containing 0.0153 to 7.65 mg per ml, when measured in a 10 mm quartz cell over the range of 252 to 400 mμ and corrected for that of redistilled alcohol, are shown in Figure 3.

One or two samples of air were collected in the course of an exposure of one-half hour in duration. When the exposure continued for 2 hours, 4 samples were collected, and in those of 6 or 7 hours, 6 to 8 samples were collected each day. Each sample of air was collected in an evacuated glass balloon of 2.100 liters capacity, and absorbed in 100 ml of redistilled ethyl alcohol released from an auxiliary small bulb through an interposed stopcock. When the air contained 8.05 mg or more of the vaporized material per liter, the solution was diluted to either 2 or 3 times its original volume with alcohol before measurements were made by means of a Beckman spectrophotometer. Figure 4 depicts the variation in transmission at 272 mμ with concentration over the range of 15.3 to 153 micrograms per milliliter.

The Determination of Ethyl Dibromobenzene or PRL-3209 When Present as a Mist in Air

Each day a sample of air laden with a mist of either ethyl dibromobenzene or PRL-3209, was withdrawn from the chamber, at the rate of 2 liters per minute, for 15 minutes through 2 absorption towers each containing 100 ml of purified 95 per cent ethyl alcohol.

The material so absorbed was determined quantitatively, by virtue of its ability to absorb ultraviolet radiations of wave length 274 m μ, by means of the Beckman spectrophotometer. Figure 5 depicts the variations in transmission at 274 m μ of concentrations of ethyl dibromobenzene over the range of 10 to 126 micrograms per ml, when read in a 50-mm cell against a comparable cell containing purified alcohol. Figure 6 depicts the variation in transmission at 274 m μ of concentrations of PRL-3209 over the range of 15 to 75 micrograms per ml, when read in a 50-mm cell against an alcohol blank. In this latter instance both the ethyl dibromobenzene and the Aroclor 1248 absorb radiation at 274 m μ.

The alcohol was prepared as follows: About 1750 ml of 95 per cent ethyl alcohol were permitted to stand overnight with 10 g of pellets of NaOH and 7 g of aluminum foil (0.003"). The mixture was then refluxed for one-half an hour and distilled at 78.5° C, the first 20 ml being rejected.

The Determination of the Thermal Decomposition Products of Ethyl Dibromobenzene or PRL-3209 in Air

Total aldehydes, formaldehyde, and total particulate matter resulting from the thermal decomposition of ethyl dibromobenzene were collected and determined. In the case of PRL-3209, carbon monoxide, total aldehydes and formaldehyde were determined. On one occasion a sample of air from the chamber containing the decomposition products of PRL-3209 was collected in alcohol and examined in the ultraviolet region for its content of PRL-3209. Complete credence may not be had in the value obtained, because the transmission curve (from 240 to 300 m μ) of the sample from the chamber, although approaching that of PRL-3209, did not completely duplicate it.

The aldehydes formed from ethyl dibromobenzene were collected by passing air from the chamber for 15 minutes at the rate of 3 liters per minute through 2 absorption towers (2.25" x 13" outside diameter). The first absorber contained a fritted-glass disc, and the second an impinger. Both towers contained 100 ml of 1 per cent sodium acid sulfite in an equal mixture of water and 95 per cent ethyl alcohol. Water alone was used to dissolve the sodium sulfite when the aldehydes from PRL-3209 were to be collected. In the case of ethyl dibromobenzene the absorbers were preceded by an electrostatic precipitator, but in the case of the formulation they were connected directly to the chamber. An aliquot portion of the bisulfite solution was analyzed for total soluble aldehydes by the method of Goldman and Yagoda. Another aliquot of the bisulfite solution was analyzed for formaldehyde by the chromotropic acid method of Bricker and Johnson.

Carbon monoxide was determined by a modification of the method of Bland. The samples were collected by passing air at the rate of 100 ml per minute for 15 minutes through a pre-equilibrated absorption train shown in Figure 8 of the WADC Technical Report No. 54-344 on Lubricants.

Particulate matter was collected in an electrostatic precipitator by passing air through it at the rate of 3 liters per minute for 15 or 30 minutes. The material was flushed out with ether, which was evaporated by mild heating. After the ether had evaporated the residue was weighed.

The following concentrations of the stated contaminants of the air of the chamber, expressed as ppm by volume in air at 25° C and 745 mm of mercury, are equivalent to the concentration of 1 mg per liter:

Compound	1 mg per liter ⇌ ppm
Formaldehyde	831.1
Carbon Monoxide	891.0
Tetrachlorotetrafluoropropane	98.3
Chloro-1-(trifluoromethyl)-4-(chlorotetrafluoroethyl)-benzene	79.2

The Immediate Toxicity When Given Orally

The minimum lethal dose of undiluted ethyl dibromobenzene (PRL-3191), when given orally to rabbits, is greater than 2.1 and less than 3.2 ml per kg of body weight. The corresponding value for rats lies between 4.7 ml and 7.1 ml per kg (Table 1). With one exception, deaths occurred within 11 to 48 hours and were preceded by coma. Prostration did not occur until 18 to 36 hours following the administration of the dose; it was occasionally observed in animals that survived. Losses in weight (8 to 28 per cent of the initial weight) were transitory among the survivors.

All animals were examined postmortem. The viscera from representative rabbits and rats were examined microscopically. Diffuse degenerative changes were seen in the livers, kidneys, brains and hearts of the rabbits. In the rats they were limited to the livers, kidneys and brains. In either species the other viscera were normal, as were the viscera of rabbits and rats that were killed at varying intervals after the compound had been given.

The minimum lethal oral dose of undiluted PRL-3169 for rabbits is less than 3.2 ml per kg (Table 1). The lack of sufficient material limited the number of doses that could be given. The few values obtained on rats (Table 1) offer no indication that the formulation differs in toxicity from its principal ingredient, ethyl dibromobenzene.

The signs of intoxication resembled those characteristic of poisoning by ethyl dibromobenzene. Their onset was delayed for at least 24 hours, after which lethargy was observed, followed later by ataxia, dyspnea and prostration. Deaths usually occurred 2 to 3 days after the administration of the material, but, in a few instances, they were delayed for several additional days. The losses in weight were large except in the case of rats given a sublethal quantity, and were transitory in the case of the survivors.

Each of the 15 rabbits was examined postmortem. Fourteen of them died after having been given an oral dose of the compound. The deaths occurred after intervals varying from 18 hours to 8 days. One rabbit

survived and was killed 12 days after the formulation had been given. The viscera from 6 of the rabbits that died were examined microscopically. The degenerative changes observed were most severe in the liver and kidney. The renal lesion was characterized by swelling and coarse granularity in the cytoplasm of the epithelial cells lining the convoluted tubules, loss of the brush borders of the cells, small quantities of granular material in the lumens of the tubules, and karyolysis of occasional tubular cells. The liver exhibited passive hyperemia and parenchymal degeneration. The hepatic cells showed hydrops, cloudy swelling, acidophilic hyaline clumping of the cytoplasm, and fatty vacuolation. The histopathologic alterations of the liver were progressive in degree with the length of survival of the animals, being most severe in the rabbit that died 8 days after that on which the compound had been administered. The lesion involved the central zone of the hepatic lobules, and at the 8th day there was massive lobular necrosis and fatty vacuolation of the hepatic cells. In some instances the necrotic lobules were surrounded by neutrophiles. The brain was edematous and congested. In 3 rabbits the lungs showed congestion and edema, and in 3 others low-grade interstitial pneumonitis. There was slight necrosis of the splenic follicles in 3 animals. The heart, stomach, intestine and adrenals were normal in all of the animals.

All of the rats were examined postmortem. Six of them had died and 8 had survived. The survivors were killed after intervals of 8 and 9 days. The viscera of one rat that died were examined microscopically, and all but the lungs were normal. In the latter, lesions of acute bronchitis and foci of lipid pneumonia were found.

When undiluted PRL-3209 was given orally in one dose to rabbits and rats the values were not greatly different from that of its principal ingredient, ethyl dibromobenzene (Table 1). The minimum lethal dose for rabbits is greater than 1.4 and less than 2.1 ml per kg. The corresponding value for rats lies between 4.7 and 7.1 ml per kg.

The ingestion of lethal doses of PRL-3209 induced diffuse degeneration of the liver, kidneys and brain, and edema and hyperemia of the lungs. In surviving rabbits, the liver had undergone fatty metamorphosis and hydropic and hyaline degeneration. The surviving rats had normal viscera.

When given orally as a solution in peanut oil to rabbits, the minimum lethal dose of tetrachlorotetrafluoropropane appears to lie between 36 and 54 mg per kg of body weight (Table 2). No signs of intoxication other than occasional diarrhea were observed among rabbits that died within 5 to 36 hours after the dose had been given. Losses in weight among the survivors were regained within 7 to 14 days. The minimum lethal dose of tetrachlorotetrafluoropropane, when given orally to rats, appeared to lie between 0.12 and 0.18 ml per kg of body weight. Rats developed diarrhea within 3 to 5 hours after the material had been administered. A little later, 4 to 7 hours after the dose had been given, weakness and prostration were seen. The length of survival of the rats which, in general, varied inversely with the size of the dose administered, ranged from 3 to 36 hours. The losses in weight sustained by the surviving rats were regained within 10 to 20 days.

Gross examination of the gastroenteric tracts of rats and rabbits given a lethal dose of undiluted tetrachlorotetrafluoropropane revealed focal hemorrhages of the mucosa of the stomach and small intestine, associated with diffuse edema of the submucosa of the stomach and upper portion of the small intestine; this condition was absent in rabbits given small dosages in peanut oil. On microscopic examination of the tissues of animals given a lethal dose, diffuse degenerative changes were found in the hepatic cells, in the epithelial cells of the distal convoluted and loop tubules of the kidney, and in scattered ganglion cells of the brain.

The minimum lethal oral dose of undiluted chloro-1-(trifluoromethyl)-4-(chlorotetrafluoroethyl)-benzene for rats appears to be greater than 24.0 and less than 36.0 ml per kg (Table 2). Death occurred within 24 hours. Oiliness of the fur around the anus of the surviving rats was evident within a period of 4 - 6 hours. The survivors lost no weight.

The Effects of Contact for Twenty-four Hours with the Skin of Rabbits

The dosage of 9.4 ml of undiluted ethyl dibromobenzene per kg in contact with the abraded skin of rabbits induced fatal intoxication (Table 3), but this dosage on the intact skin caused no fatalities.

One of 3 rabbits died, following the period of contact of its abraded skin with the dosage of 6.0 ml per kg, but the dosage of 3.2 ml per kg was tolerated by all of 3 rabbits. These animals exhibited no signs of intoxication, and with but one exception, they sustained no greater losses in body weight than did control rabbits kept in stocks for a corresponding period of time.

All of the rabbits were subjected to postmortem examinations, and the viscera from representative animals were examined microscopically. The rabbits subjected to contact of the compound with the intact skin survived and had normal viscera. The skin exhibited moderate microscopic evidences of low-grade inflammation. Rabbits that died following application of the compound upon the abraded skin had diffuse degeneration of the liver, kidney and brain. The other viscera were normal. The skin of these animals exhibited diffuse subacute inflammation and foci of hemorrhage at the site of the abrasions. The 3 rabbits that survived following contact of the compound with the abraded skin exhibited degeneration of the liver. The degree of hepatic damage varied directly with the amount of compound applied upon the skin. The remaining viscera were normal. The skin showed slight subacute inflammation.

When maintained in contact with either the intact or abraded skin of rabbits, according to the 24-hour sleeve method of Draize, Woodard and Calvery, the minimum lethal dose of undiluted PRL-3169 is greater than 6.0 and less than 9.4 ml per kg of body weight (Table 3).

Application of formulation PRL-3169 upon either the intact or abraded skin of rabbits induced moderately severe degeneration and necrosis of the liver and of the convoluted tubules of the kidney, but the material did not induce any histologic alteration of either the abraded or intact skin.

Although rabbits with either intact or abraded skin tolerated the dosage of 9.4 ml of undiluted PRL-3209 per kg (Table 3), the material induced moderate degeneration of the liver, kidneys and brain, and subacute inflammation of the skin.

The Effects of Respiratory Exposure to the Vapor of Certain Halogenated Hydrocarbons

The mortality among animals exposed to the undecomposed vapor of tetrachlorotetrafluoropropane in air is shown in Table 4. The mortality data for groups of guinea pigs, rabbits, rats and cats are plotted logarithmically in relation to severity of exposure in Figures 7, 8, 9 and 10, respectively. In these graphs the duration of each exposure in hours is plotted along the axis of abscissae, and the corresponding concentration along the axis of ordinates. Accordingly, the location of a point anywhere on such a graph indicates the severity of exposure, as expressed in terms of the concentration of the vapor in the air and the length of time it was inhaled. Data from each of the experiments heve been represented in circles, the extent to which each circle is filled indicating the proportion of the animals in the exposed group that died under the given conditions.

In three of the graphs (cf Figures 7, 8 and 10), a straight line has been drawn to separate the area into 2 regions, one above and to the right, within which any point indicates lethal conditions of exposure, and one beneath and to the left, within which any point indicates conditions that permit survival of all of the animals. None of these experiments was continued sufficiently long to indicate whether, in the case of lower concentrations, a discontinuity or deflection of the straight line would appear. Large deflections, which have been observed under conditions of prolonged exposure to low concentrations of other toxic materials, indicate that the duration of exposure to such concentrations has little influence on mortality.

Inspection of Figures 7 and 8 shows that the slopes of the lines are about the same, but that the position of the line representing the behavior of rabbits is below that relating to guinea pigs, indicating that rabbits are somewhat more susceptible than guinea pigs. No line could be drawn in Figure 9, because no rats survived in any of the experiments. To permit survival the severity of the exposure would have had to be considerably less than that of any of our experiments.

Figure 10 (932 ppm for 2 hours) shows that a cat (A334) survived under conditions that might have been expected to be fatal. This cat was acutely ill and recovered slowly.

In general, animals subjected to the more severe conditions died earlier than those exposed less severely. Rats usually died more promptly than did animals of other species. Twenty-five animals died during a period of exposure or within 1 hour after its termination, 21 others died within 1 to 10 hours after they had been removed from the chamber, 24 did not succumb until 18 to 27 hours had elapsed after they had been removed from the chamber, and 3 died only after the lapse of 2 or 3 days.

Exposure for 3.5 hours to air being 1.47 mg of tetrachlorotetrafluoropropane per liter (145 ppm), the lowest concentration investigated, was lethal for rats but not for rabbits. Exposure to the concentration of 1.62 mg per liter (159 ppm) over the period of 7 hours was lethal for rats and rabbits, but not for cats and guinea pigs. Animals of the two latter species died as the result of exposure for a like period to the concentration of 3.64 mg per liter (358 ppm). Exposure to higher concentrations for shorter periods of time induced toxic effects and fatalities.

The inhalation of tetrachlorotetrafluoropropane induced irritation of the mucous membranes and respiratory tract. Prostration and tetanic and clonic convulsions frequently preceded death.

The tissues of animals exposed to the vapor of tetrachlorotetrafluoropropane were the site of congestion and edema. There were also focal hemorrhages in the lungs, and degenerative changes in the liver, kidneys, heart and brain of some animals. The changes were more severe in animals that had been exposed to higher concentrations, and the changes in rats and rabbits that died during or after exposure were more severe than those found in guinea pigs and cats. Examination of tissues from some animals that survived for several days up to 15, after the exposure and were then killed, revealed the presence of interstitial pneumonitis.

The mortality data for cats, guinea pigs, hamsters, rabbits and rats exposed to the undecomposed vapor of chloro-1-(trifluoromethyl)-4-(chlorotetrafluoroethyl)-benzene are given in Table 5. The mortality data for

groups of guinea pigs, rabbits and rats are plotted logarithmically in relation to the severity of exposure in Figures 11, 12 and 13, respectively.

Although some individual variations in susceptibility were noted among animals of certain species, particularly guinea pigs, no characteristic differences between species were apparent.

Exposure for 2 hours to the concentration of 12.07 mg per l (955.9 ppm) was lethal for guinea pigs, rabbits and rats, but all animals of these species survived following their exposure for 2 hours to the concentration of 8.05 mg per l (637.6 ppm). Deaths occurred among groups of guinea pigs, rabbits and rats subjected for 7 hours to the concentration of 9.03 mg per l (715.2 ppm), but all of the animals survived when exposed for a like period of the concentration of 3.88 mg per l (307.3 ppm). The mortality among cats and hamsters, employed in small numbers in these experiments, appeared not to differ from that among the other species of animals.

Exposure to the vapor of chloro-1-(trifluoromethyl)-4-(chlorotetrafluoroethyl)-benzene induced slight irritation of the mucous membranes. Ataxia and convulsions were prominent among the cats and rats at all tested concentrations and among the rabbits and guinea pigs subjected to the greater of these.

Gross and microscopic examination of the tissues of animals that had been exposed to chloro-1-(trifluoromethyl)-4-(chlorotetrafluoroethyl)-benzene demonstrated that the compound had an irritating effect on the lungs; this effect was slight at lower concentrations and moderately severe when the exposure was sufficient to cause death. In addition, the compound produced parenchymatous degeneration of the kidneys and liver, and fatty degeneration of the livers of some animals.

The Effects of Respiratory Exposure to the Mist of Ethyl Dibromobenzene and to that of a Formulation Containing Ethyl Dibromobenzene (PRL-3209)

Pertinent data resulting from exposure to the mist of these materials are presented in Table 6. A group of animals, consisting of a cat, 2 guinea pigs, 2 rabbits

and 4 rats, survived, with the exception of 1 rabbit, when exposed to the mist of ethyl dibromobenzene in the air in the concentration of 0.73 mg per 1 for 7 hours on each of 10 days over a period of 15 days. The mist induced irritation of the mucous membranes, increased rates of respiration, and ataxia. Losses in weight were great in the case of guinea pigs, and one of them would probably have died had it not been killed. It had pneumonitis, degenerative changes of the liver and kidneys, and focal necrosis of the adrenal cortex. The rabbit that died had acute pulmonary hyperemia and perivascular hemorrhages, diffuse necrosis of the liver, and degeneration of the renal tubules. Among the survivors, a rabbit and a rat exhibited similar alteration of lesser severity, in the lungs, liver and kidneys. Only a cat had entirely normal viscera.

Animals of several species were subjected, for the various indicated periods of time, to exposure to air bearing a mist of PRL-3209 in the concentrations listed (Table 6): 70 hours, 0.68 mg per 1; 70 hours, 0.39 mg per 1: and 7 hours, 1.35 mg per liter. All of the guinea pigs but none of the rats died, while a cat and a dog survived, under the conditions of exposure indicated above. One of 2 rabbits died following exposure for 70 hours to the concentration of 0.68 mg per 1, but 5 other rabbits survived following exposure in the other experiments.

The inhalation of the mist of PRL-3209 induced lacrimation and other signs of irritation of the eyes, drooling, labored respiration, and vomiting among the experimental animals. Headaches and nausea were experienced by persons who sustained some exposure to the mist in the operation of the equipment.

Inhalation of these mists induced degeneration of the brain, liver, kidneys and adrenal glands of guinea pigs and rabbits.

<u>The Effects of Respiratory Exposure to Products Arising from the Contact of Ethyl Dibromobenzene or Its Formulation (PRL-3209) with an Heated Inconel Surface</u>

The mortality in the several groups of guinea pigs, rabbits, rats and cats exposed to the fog formed by contact of ethyl dibromobenzene with Inconel

maintained at 700° F (371° C) are shown in Table 7 and comparable data relating to exposure to the fog formed at 900° F (482° C) are given in Table 8. The mortality in the several groups of guinea pigs, rabbits, rats and cats subjected to the fog formed at 900° F has been plotted logarithmically in Figures 14, 15, 16 and 17, respectively. Because the rates of delivery were closely comparable in experiments 5 and 7, and 11 and 12, the results of these paired experiments have been combined in these graphs.

All animals survived when exposed for 7 hours to the fogs formed by dropping either 118 or 214 mg of ethyl dibromobenzene per minute into an Inconel tube maintained at 700° F. An increase in the rate of delivery of the liquid to 237 or 412 mg per minute induced fatalities among guinea pigs, but the cats, rabbits and rats survived. Except for that generated at the lowest rate of delivery, the fogs induced irritation of the mucous membranes and dyspnea. The guinea pigs that died had severe bronchitis and pneumonitis and severe diffuse degeneration of the liver and kidneys, and one of them had focal necrosis of the adrenal cortex. Varying degrees of chemical pneumonitis and degenerative changes in the liver and kidneys were encountered among the cats, rabbits and rats.

An increase in the temperature of the Inconel tube to 900° F resulted in the generation of a more toxic fog than that formed at the lower temperature. All of the animals survived exposure for 7 hours when the rate of introduction of the material into the tube was 98.8 mg per minute, but when the rate was increased to 165 mg per minute fatalities occurred among the guinea pigs and the rats. Cats and rabbits survived exposure when the rate of delivery was 247 mg per minute, but died when the rate was increased to 311 mg per minute. All of the animals survived following exposure for 24 hours to the fog formed by dropping 102 mg of ethyl dibromobenzene per minute into an Inconel tube maintained at 900° F. These fogs induced inflammation of the eyelids, salivation, discharge of mucous, and dyspnea. The tissue alterations consisted of acute chemical pneumonitis and degeneration and necrosis of the liver and kidneys. In animals exposed to the fogs formed at the higher rates of delivery, degenerative changes were seen also in the heart muscle.

The fog formed at 900° F was more dense than that formed at 700° F. The particulate matter which formed at 900° F consisted of a yellow to brown oil containing solid material, and varied in amount from 0.10 to 0.93 mg per l of air. That formed at 700° F consisted essentially of a white solid which was present to the extent of 0.018 to 0.068 mg per liter. Although no formaldehyde was found, higher aldehydes, expressed as formaldehyde, were present to the extent of 0 to 28 ppm, but their concentration was unrelated to the rate of dropping the liquid into the furnace.

The fate of each animal exposed to the fog formed by contact of PRL-3209 with an Inconel surface maintained at 700° F (371° C) is given in Table 9. All animals of the several species survived following their exposure for 2 hours to fogs formed by dropping 78.4, 116.7 or 135.8 mg of PRL-3209 per minute into an Inconel tube heated to 700° F, through which air was passing at the rate of 31.8 l per minute. All of the animals survived following corresponding exposure for 3 hours to the fog resulting from dropping 74.4 mg per minute into the tube. When animals were exposed for 24 hours to the thermal decomposition products arising when PRL-3209 was delivered at the rates of 100.6, 61.4 or 35.5 mg per minute into the heated Inconel tube, all of them, with the exception of the rats which were exposed to the highest level, survived. These rats died during exposure.

Exposure to the fog of decomposition products induced irritation of the mucous membranes of the animals and some degree of labored respiration, except under the conditions associated with the least rate of delivery (35.5 mg per minute) of the material. In general, the losses of weight on the part of the animals were more severe in association with the longer, rather than the shorter, periods of exposure. These losses were generally regained within 2 or 3 weeks, before the animals were killed.

Under the conditions of these experiments, the air in the chamber contained total aldehydes, expressed as formaldehyde, in concentrations of 1.1 to 3.6 ppm, formaldehyde in concentrations of 0.0 to 1.2 ppm, and carbon monoxide in concentrations of 24.1 to 56.1 parts per million.

Inhalation of the decomposition products induced degeneration of the livers of guinea pigs and diffuse degeneration of the brain, liver and kidneys of rats. No pathologic changes were found in the tissues of animals that survived exposure to the less severe conditions.

DISCUSSION

According to the conventional terminology, as suggested by Hodge and Sterner, ethyl dibromobenzene and its formulations (PRL-3169 and PRL-3209) when given orally to rabbits may be considered to be only slightly toxic, whereas tetrachlorotetrafluoropropane would be classified as moderately toxic. Ethyl dibromobenzene and its formulations would be regarded as essentially harmless when applied upon the skin of rabbits.

The data on mortality make it appear likely that the undecomposed mist of either ethyl dibromobenzene or PRL-3209 is more toxic than comparable mists of the aliphatic esters or their formulations (WADC Technical Report No. 54-344 on Lubricants). The reverse appears true of the fogs formed by thermal decomposition.

From the aspect of immediate toxicity, the vapor of tetrachlorotetrafluoropropane appears to be somewhat more toxic than that of most chlorinated hydrocarbons, including 1,1,2,2- tetrachloroethane, which is considered to be among the most toxic of the commonly employed chlorinated hydrocarbons. Von Oettingen reported that cats were deeply narcotized after 45 minutes of exposure to the concentration of 5 mg of the latter compound per liter of air (8,294 ppm) or after 5.5 hours of exposure to a concentration one-tenth so great. Lehmann reported that 2 cats and 1 rabbit survived following exposure for 6 to 7 hours per day on each of 18 days over a period of 4 weeks to the concentration of 1.1 to 2.3 mg of 1,1,2,2-tetrachloroethane per l (160 to 335 ppm) (Av 1.38 mg/l or 201 ppm), without exhibiting signs of intoxication other than apathy and loss in weight (about 10 per cent). It is likely that corresponding conditions of exposure to tetrachlorotetrafluoropropane would have been lethal to cats, since one cat died following exposure to the concentration of 358 ppm (cf Figure 10) over the period of 7 hours.

Tetrachlorotetrafluoropropane appears to be a little less toxic than hexachlorobutadiene. The toxicity of hexachlorobutadiene is illustrated by the following excerpt from an unpublished report from this Laboratory. "Cats are killed by an exposure of 3.5 hours to a concentration of 0.3 mg/l (34.1 ppm), guinea pigs by one of 7.0 hours to 0.29 mg/l (275 ppm), and rats by one of 7.0 hours to 1.39 mg/l (132.9 ppm). Rabbits were a little more resistant than guinea pigs but there was considerable variation in the susceptibility of individual rabbits."

The vapor of chloro-1-(trifluoromethyl)-4-(chlorotetrafluoroethyl)-benzene is somewhat toxic but less so than that of tetrachlorotetrafluoropropane. The hazard of exposure to the vapor from either of these 2 halogenated hydrocarbons should be controlled by suitable means.

Table 1

The Immediate Toxicity of Ethyl Dibromobenzene and of Formulations PRL-3169 and PRL-3209, When Given Orally to Rabbits and Rats

(given undiluted)

Dose ml/kg	Number of Animals that Died/ Number of Animals Given the Material					
	Ethyl Dibromobenzene (PRL-3191)		Formulation (PRL-3169 (1)		Formulation (PRL-3209)	
	Rabbits	Rats	Rabbits	Rats	Rabbits	Rats
36.0	-	-	-	3/4	-	-
24.0	-	-	1/1	3/4	-	4/4
16.0	-	4/4	2/2	0/1	-	3/4
10.7	-	1/4	4/4	0/2	-	3/4
7.1	1/1	1/4	3/3	-	-	1/4
4.7	0/1	0/4	2/2	0/1	3/3	0/4
3.2	3/3	-	2/2	0/1	2/5	
2.1	0/3	-	-	0/1	2/4	
1.4	0/1	-	0/1	-	0/3	

(1) Insufficient material available for administration to additional animals.

Table 2

The Immediate Toxicity of Tetrachlorotetrafluoropropane and of Chloro-1-(Trifluoromethyl)-4-(Chlorotetrafluoroethyl)-Benzene, When Given Orally to Animals

(given undiluted, unless otherwise stated)

Dose ml/kg	Number of Animals that Died/ Number of Animals Given the Material		
	Tetrachlorotetrafluoropropane		Chloro-1-(Trifuloromethyl)-4-(Chlorotetrafluoroethyl)-Benzene
	Rabbits	Rats	Rats
36.0	-		3/3
24.0	-		0/3
4.7-7.0	-	5/5	-
0.42-2.1	-	11/11	-
0.28-0.64	4/4	-	-
0.28	-	2/2 (3)	-
0.18	-	3/4 (3)	-
0.12	4/4 (1)	0/2 (3)	-
0.080		0/2 (3)	-
0.054	1/2 (1)	-	-
0.036	0/3 (2)	-	-

(1) Given as a 2 per cent V/V solution in peanut oil.
(2) Given as a 1 per cent V/V solution in peanut oil.
(3) Given as an 8 per cent V/V solution in peanut oil.

Table 3

The Immediate Toxicity of Ethyl Dibromobenzene and of Formulations PRL-3169 and PRL-3209, When Maintained for Twenty-four Hours in Contact with the Skin of Rabbits

(applied undiluted)

Dosage ml/kg	Condition of Skin	Number of Animals that Died/ Number of Animals Given the Material		
		Ethyl Dibromobenzene (PRL-3191)	Formulation PRL-3169	Formulation PRL-3209
9.4	intact	0/3	2/3	0/3
	abraded	3/3	1/3	0/3
6.0	intact	-	0/3	-
	abraded	1/3	0/3	-
3.2	intact	-	-	-
	abraded	0/3	-	-

Table 4

The Mortality Among Animals Exposed
to the Vapor of Tetrachlorotetrafluoropropane
in Various Concentrations in Air

Concentration mg/l	Concentration ppm	Duration Hours	Cats	Guinea Pigs	Rabbits	Rats
43.09	4236	1.1	1/1	2/2	2/2	4/4
35.23	3463	0.5	1/1	2/2	2/2	4/4
15.24	1498	2.0	1/1	2/2	2/2	4/4
12.38	1217	0.5	1/1	0/2	2/2	4/4
10.95	1076	0.25	0/1	0/2	0/2	4/4
9.48	932	2.0	1/1	1/2	2/2	4/4
9.48	932 (1)	0.5	0/1	0/2	0/2	4/4
3.64	358 (1)	2.0	0/1	0/2	2/2	4/4
3.64	358	7.0	1/1	2/2	2/2	4/4
1.62	159	7.0	0/1	0/2	2/2	4/4
1.47	145	3.5	-	-	0/2	4/4

(1) Estimated from experimental conditions. Samples for analyses were contaminated.

Table 5

The Mortality Among Animals Exposed to the Vapor of Chloro-1-(Trifluoromethyl)-4-(Chlorotetrafluoroethyl)-Benzene in Air

Concentration mg/ml	Concentration ppm	Duration Hours	Cats	Guinea Pigs	Hamsters	Rabbits	Rats
12.05	954.4	6	1/1	1/2		1/2	4/4
12.07	955.9	2	1/1	2/2		1/2	4/4
13.00	1,029.6	1/2	-	-		-	0/4
9.03	715.2	7	1/1	1/2		1/2	2/3
8.05	637.6	2	-	0/2	0/1	0/2	0/4
8.97	710.4	1/2	-	1/2	0/1	0/2	0/4
4.37	346.1	10 x 7	0/1	0/2	0/1	0/2	0/4
4.03	319.2	5 x 7	0/1	0/2	0/1	0/2	0/3
4.52	358.0	3 x 7	-	-	-	-	0/3
4.34	343.7	3 x 7	-	-		-	0/3
4.25	336.6	2 x 7	-	-		-	0/4
3.88	307.3	7	-	0/2	0/1	0/2	0/4

Table 6

The Fate of Animals Exposed to a Mist of
Ethyl Dibromobenzene or Formulation PRL-3209 in Air

Material	Rate of Aspiration of Material mg/min	Gauge Pressure psi	Rate of Passage of Air Through Aspirator (at room conditions) l/min	Supplementary Flow of Air into Chamber l/min	Concentration Found in Chamber mg/l	Duration of Exposure Hours	Number of Fatalities/ Number of Animals Exposed				
							Cats	Dogs	Guinea Pigs	Rabbits	Rats
Ethyl Dibromobenzene	144.3	4.0	18.6	39.4	0.731	10 x 7.0	0/1	-	0/2 (2)	1/2	0/4
PRL-3209	256.2	36.0	47	21.7	1.35	7.0	-	-	2/2	0/3	0/4
PRL-3209	64.9 (1)	36.0	47	0	0.68	10 x 7.0	-	0/1	2/2	1/2	0/2
PRL-3209	82.8	4.5	18.6	90.7	0.39	10 x 7.0	0/1	-	2/2	0/2	0/4

(1) The nozzle of the aspirator employed in this experiment was of different diameter than that employed in the other experiments.

(2) One guinea pig would have died, in all probability, had it not been killed.

Table 7

The Fate of Animals Exposed to the Fog Formed by Dropping Ethyl Dibromobenzene into an Inconel Tube Heated to the Temperature of 700° F (371° C)

(rate of passage of air through furnace: 31.8 l/min)

Experiment Number	Rate of Delivery of Ethyl Dibromobenzene into Furnace mg/min	Duration of Exposure Hours	Number of Fatalities/ Number of Animals Exposed			
			Cats	Guinea Pigs	Rabbits	Rats
4	412.4	7	0/1	1/2	0/2	0/4
2	236.6	7	0/1	2/2	0/2	0/4
1	214.0	7	0/1	0/2	0/2	0/4
3	118.2	7	0/1	0/2	0/2	0/4
Total			0/4	3/8	0/8	0/16
Percentage			0.0	26.7	0.0	0.0

Table 8

The Fate of Animals Exposed to the Fog Formed by Dropping Ethyl Dibromobenzene into an Inconel Tube Heated to the Temperature of 900° F (482° C)

(rate of passage of air through furnace: 31.8 l/min)

Experiment Number	Rate of Delivery of Ethyl Dibromobenzene into Furnace mg/min	Duration of Exposure Hours	Number of Fatalities/ Number of Animals Exposed			
			Cats	Guinea Pigs	Rabbits	Rats
14	1255.0	2	1/1	2/2	2/2	2/4
6	1578.6	7	1/1	2/2	1/2	3/4
8	446.0	7	1/1	2/2	2/2	3/4
7	315.2	7	0/1	1/2	0/1	1/4
5	311.4	7	1/1	1/2	2/2	1/4
13	247.1	7	0/1	1/2	0/2	2/4
12	176.4	7	0/1	0/1	0/2	4/4
11	164.5	7	0/1	1/2	0/2	1/4
9	98.8	7	0/1	0/2	0/2	0/4
10	102.4	24	0/1	0/2	0/2	0/4
Total			4/10	10/19	7/19	17/40
Percentage			40.0	52.6	36.8	42.5

Table 9

The Fate of Animals Exposed to the Fog Formed by Dropping PRL-3209 into an Inconel Tube Heated to the Temperature of 700° F (371° C)

(Rate of passage of air through furnace: 31.8 l/min)

Experiment Number	Rate of Delivery of PRL-3209 into Furnace mg/min	Duration of Exposure Hours	Number of Fatalities/ Number of Animals Exposed			
			Cats	Guinea Pigs	Rabbits	Rats
3	135.8	2	-	0/2	0/3	0/2
2	116.7	2	-	0/2	0/3	0/2
4	78.4	2	-	0/2	0/3	0/2
1	74.4	3	-	0/2	0/2	0/2
5	100.6	24	0/1	0/1	0/2	4/4
6	61.4	24	0/1	0/2	0/2	0/2
7	35.5	24	-	0/2	0/3	0/3
Total			0/2	0/13	0/18	4/17
Percentage			0.0	0.0	0.0	23.5

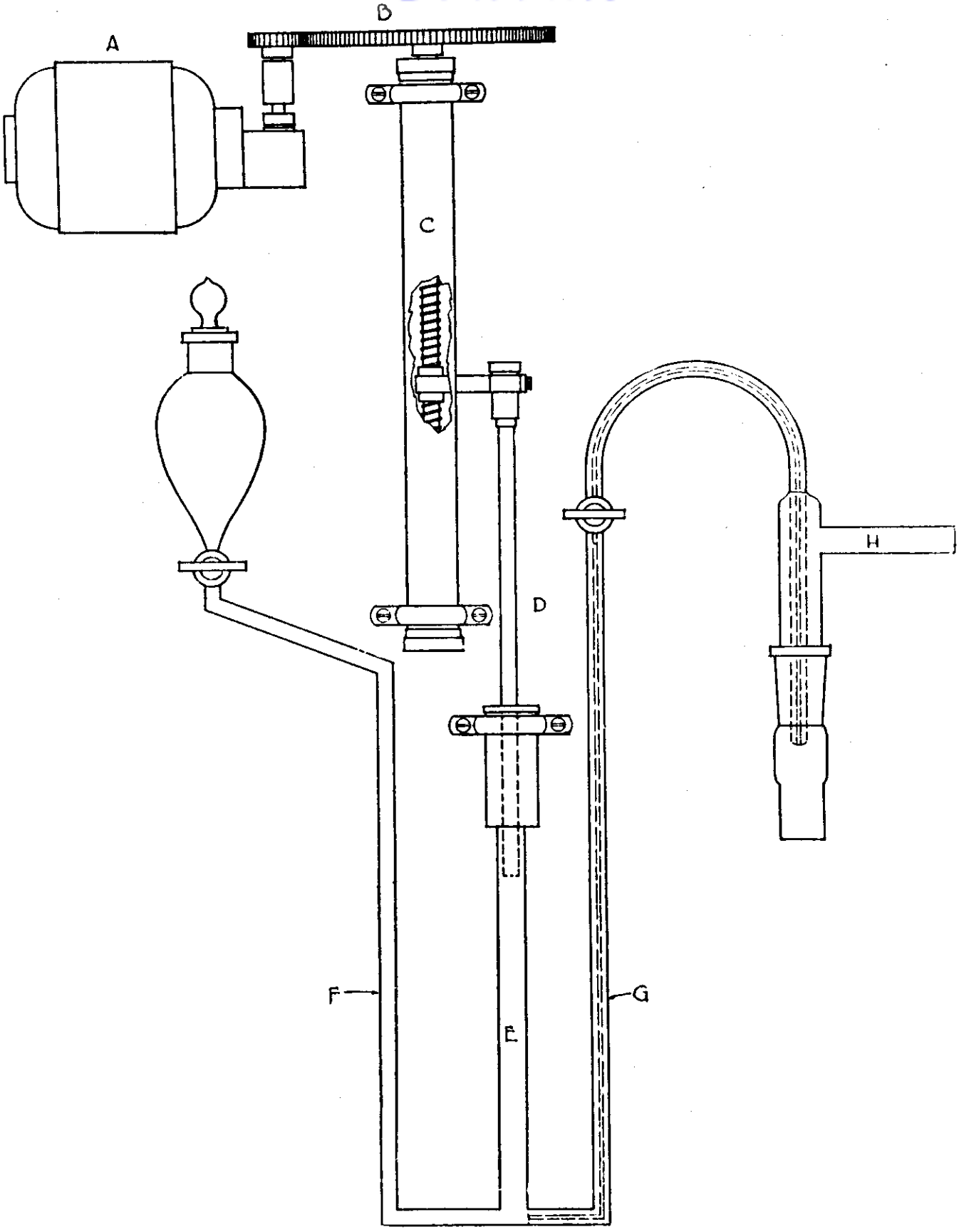


Figure 1. Apparatus for Delivering a Small Volume of Liquid at a Constant Rate

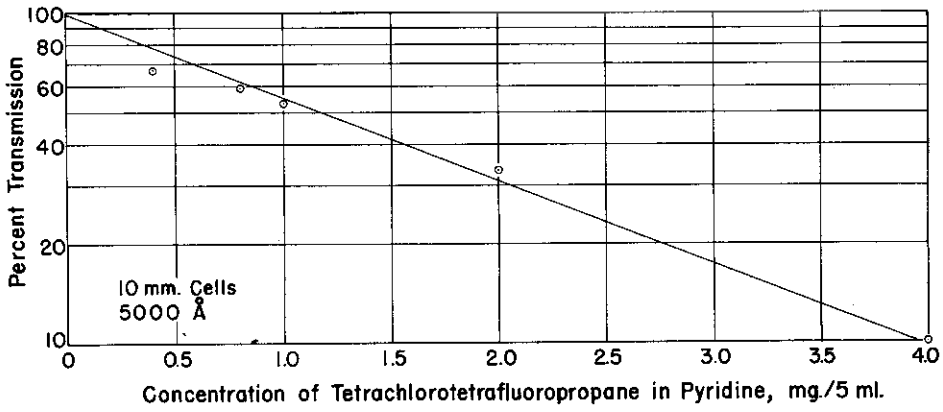


Figure 2.

Standardization Curve for the Determination of Tetrachlorotetrafluoropropane

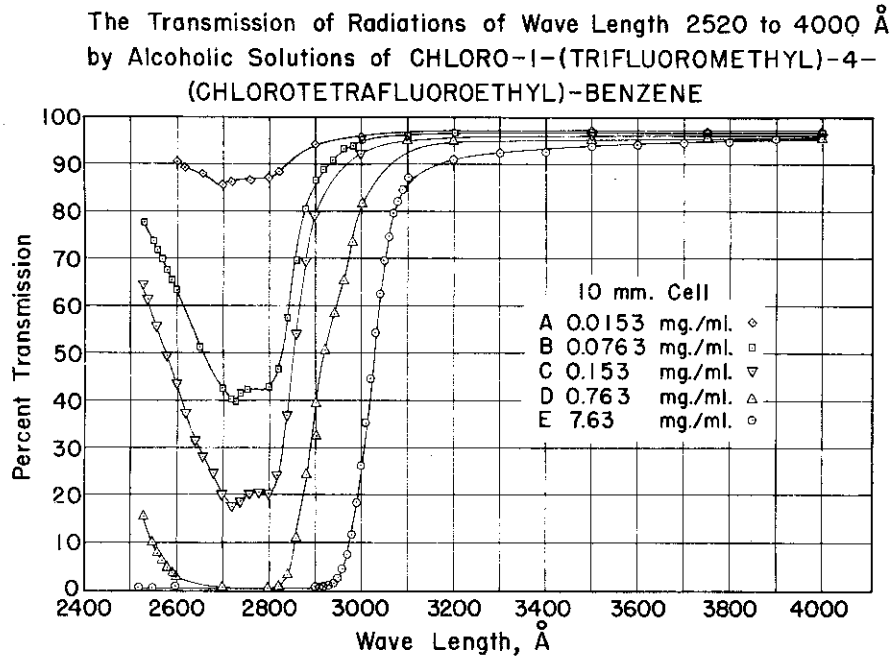


Figure 3.

The Transmission of Radiations of Wave-Length 2520 to 4000 Å by Alcoholic Solutions of Chloro-1-(Trifluoromethyl)-4-(Chlorotetrafluoroethyl)-Benzene

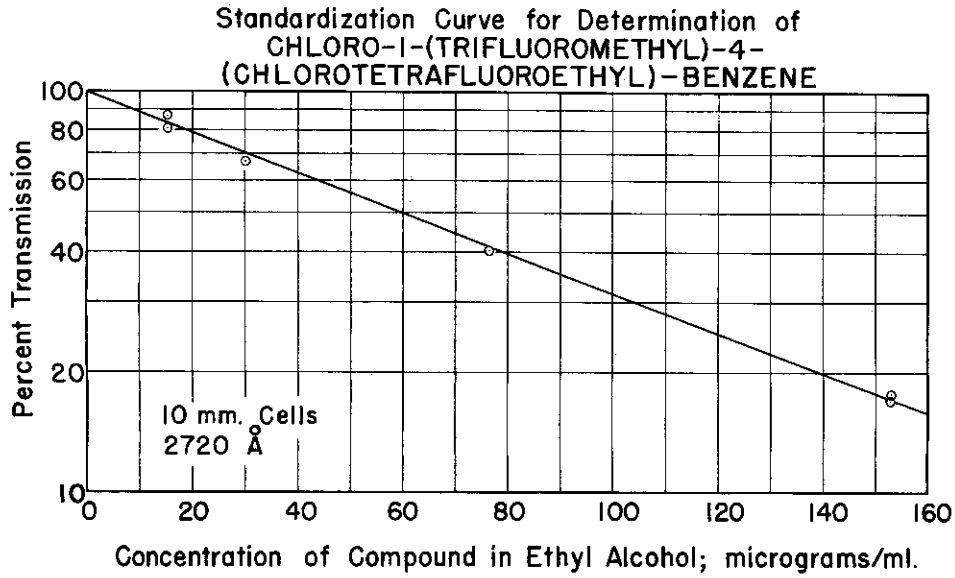


Figure 4.

Standardization Curve for Determination of Chloro-1-(Trifluoromethyl)-4-(Chlorotetrafluoroethyl)-Benzene

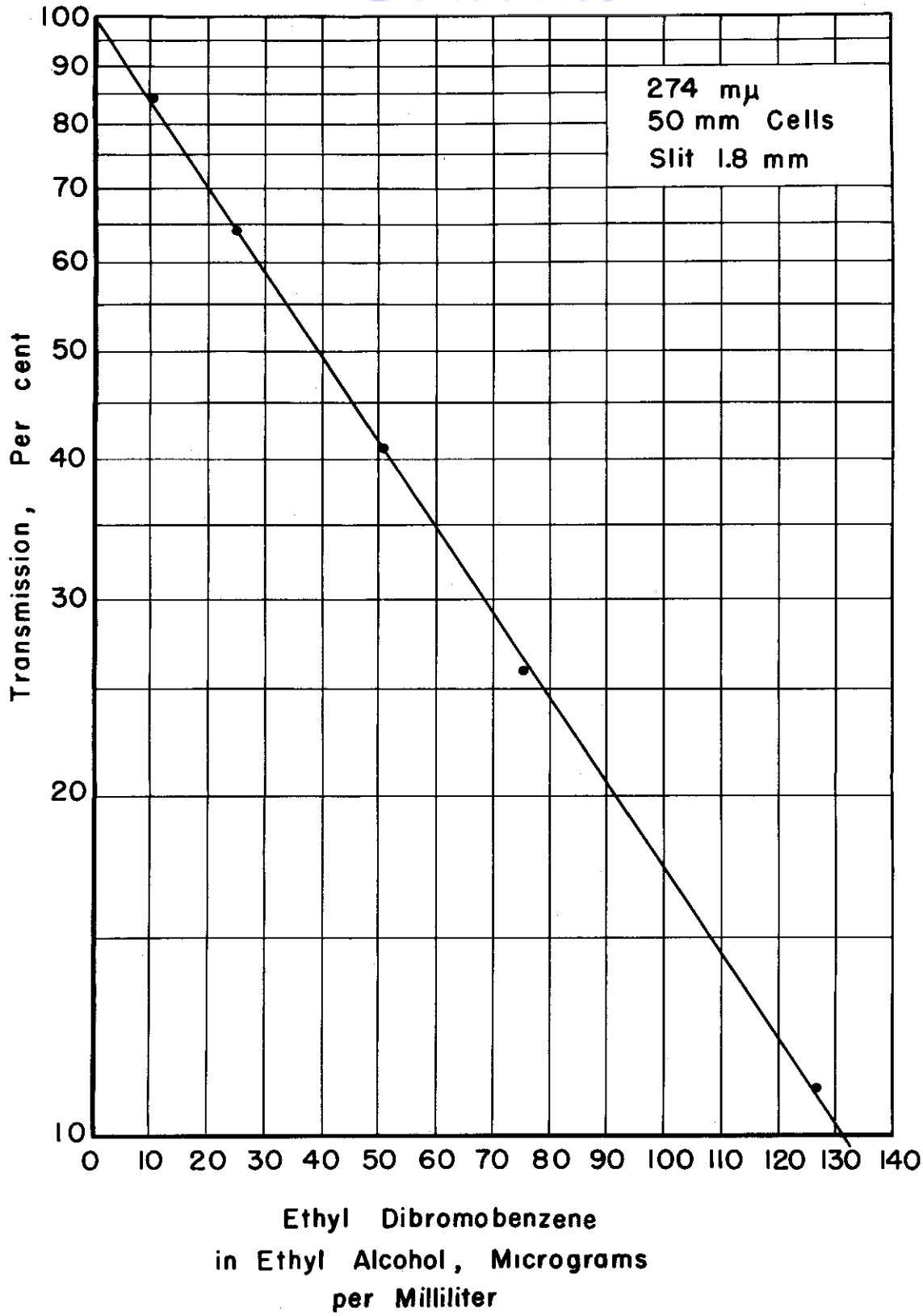


Figure 5. Standardization Curve for Determination of Ethyl Dibromobenzene (PRL-3191)

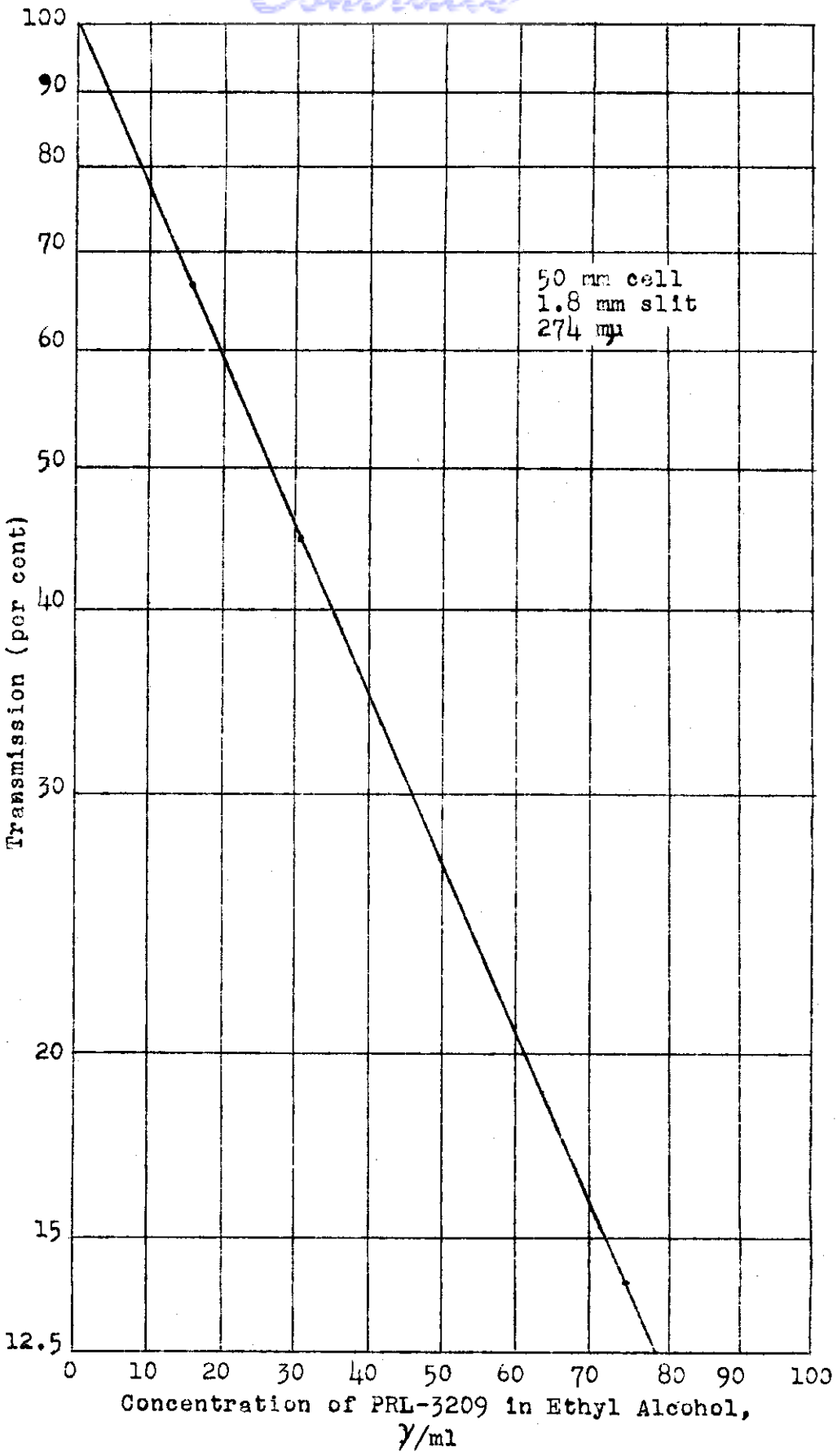


Figure 6. Standardization Curve for the Determination of PRL-3209

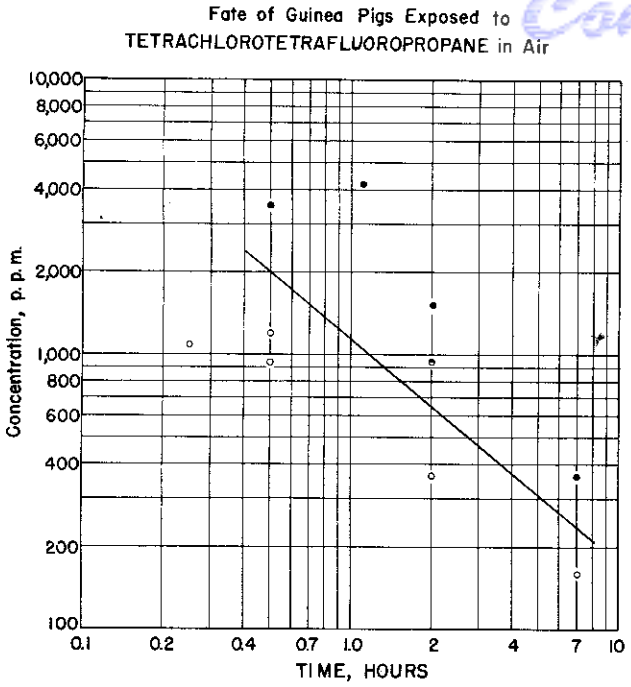


Figure 7. The Fate of Guinea Pigs Exposed to the Vapor of Tetrachlorotetrafluoropropane in Air.

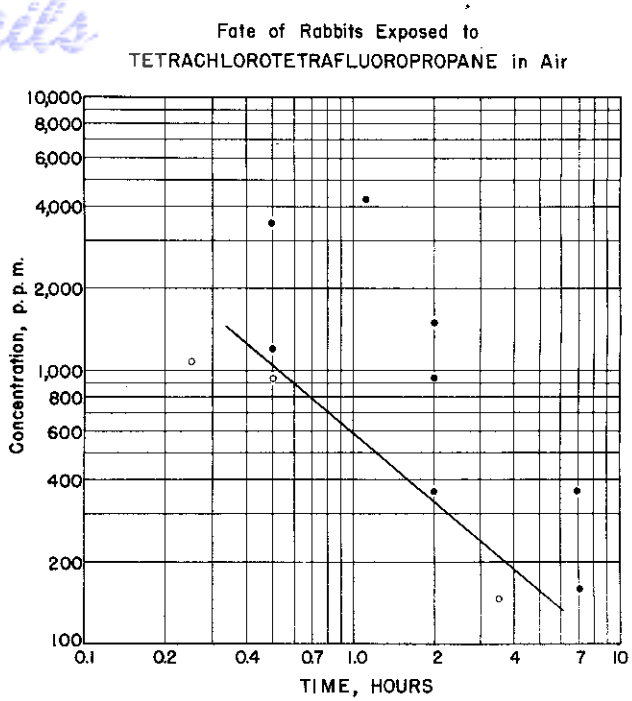


Figure 8. The Fate of Rabbits Exposed to the Vapor of Tetrachlorotetrafluoropropane in Air.

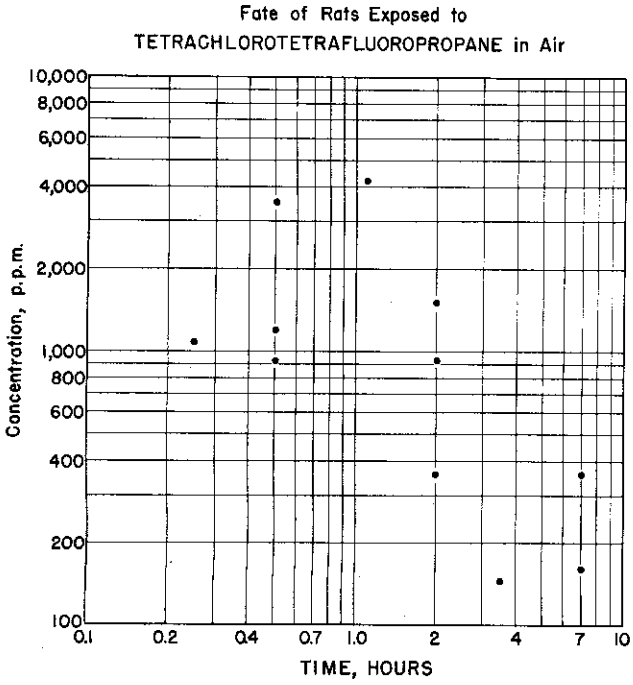


Figure 9. The Fate of Rats Exposed to the Vapor of Tetrachlorotetrafluoropropane in Air.

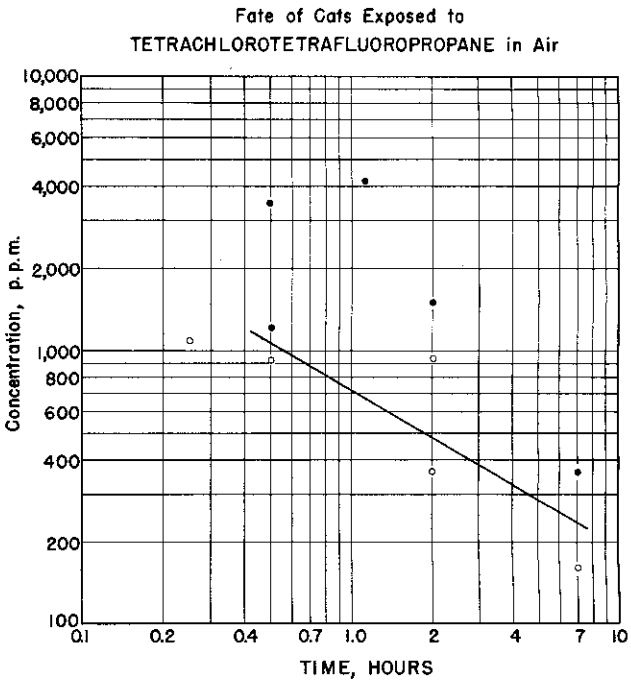


Figure 10. The Fate of Cats Exposed to the Vapor of Tetrachlorotetrafluoropropane in Air.

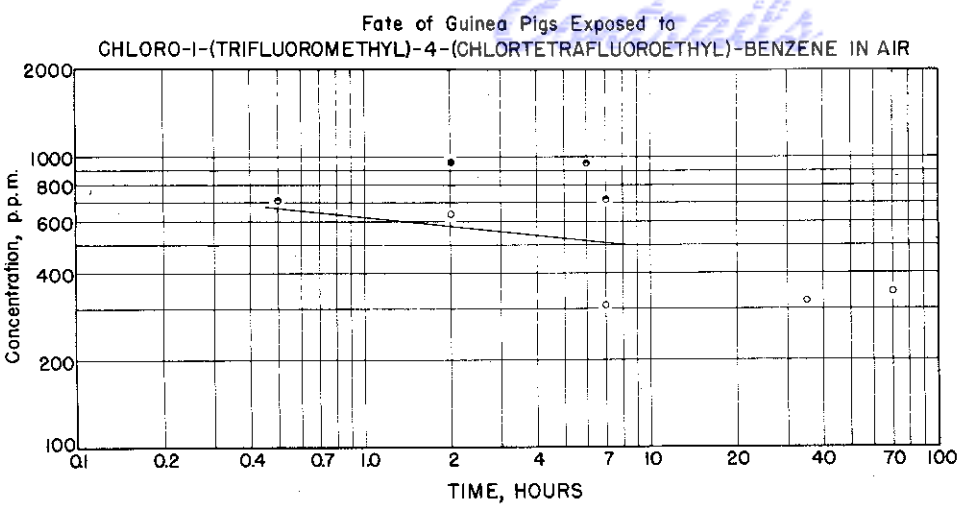


Figure 11.

The Fate of Guinea Pigs Exposed to the Vapor of Chloro-1-(Trifluoromethyl)-4-(Chlorotetrafluoroethyl)-Benzene in Air.

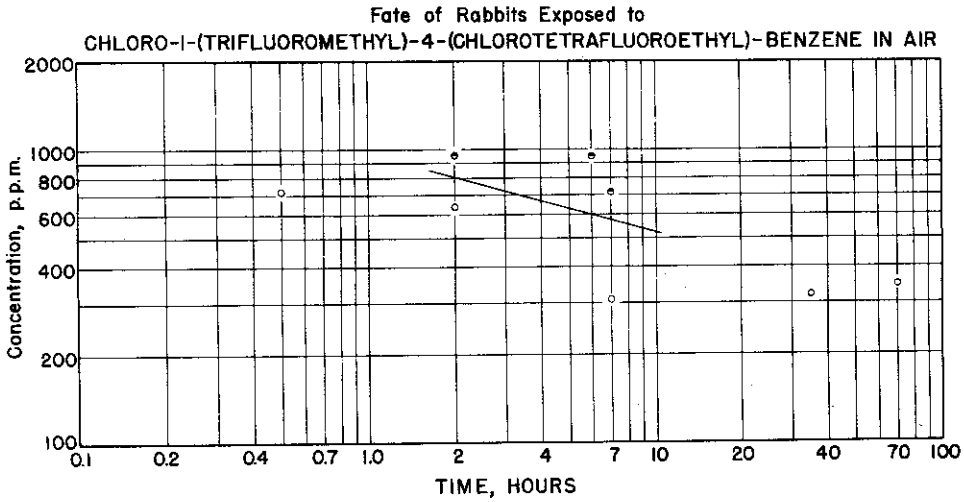


Figure 12.

The Fate of Rabbits Exposed to the Vapor of Chloro-1-(Trifluoromethyl)-4-(Chlorotetrafluoroethyl)-Benzene in Air.

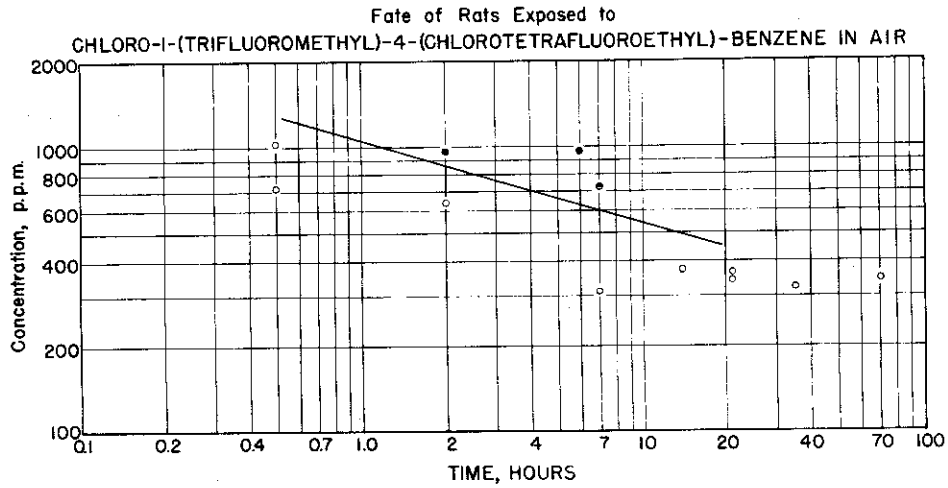


Figure 13.

The Fate of Rats Exposed to the Vapor of Chloro-1-(Trifluoromethyl)-4-Chlorotetrafluoroethyl)-Benzene in Air.

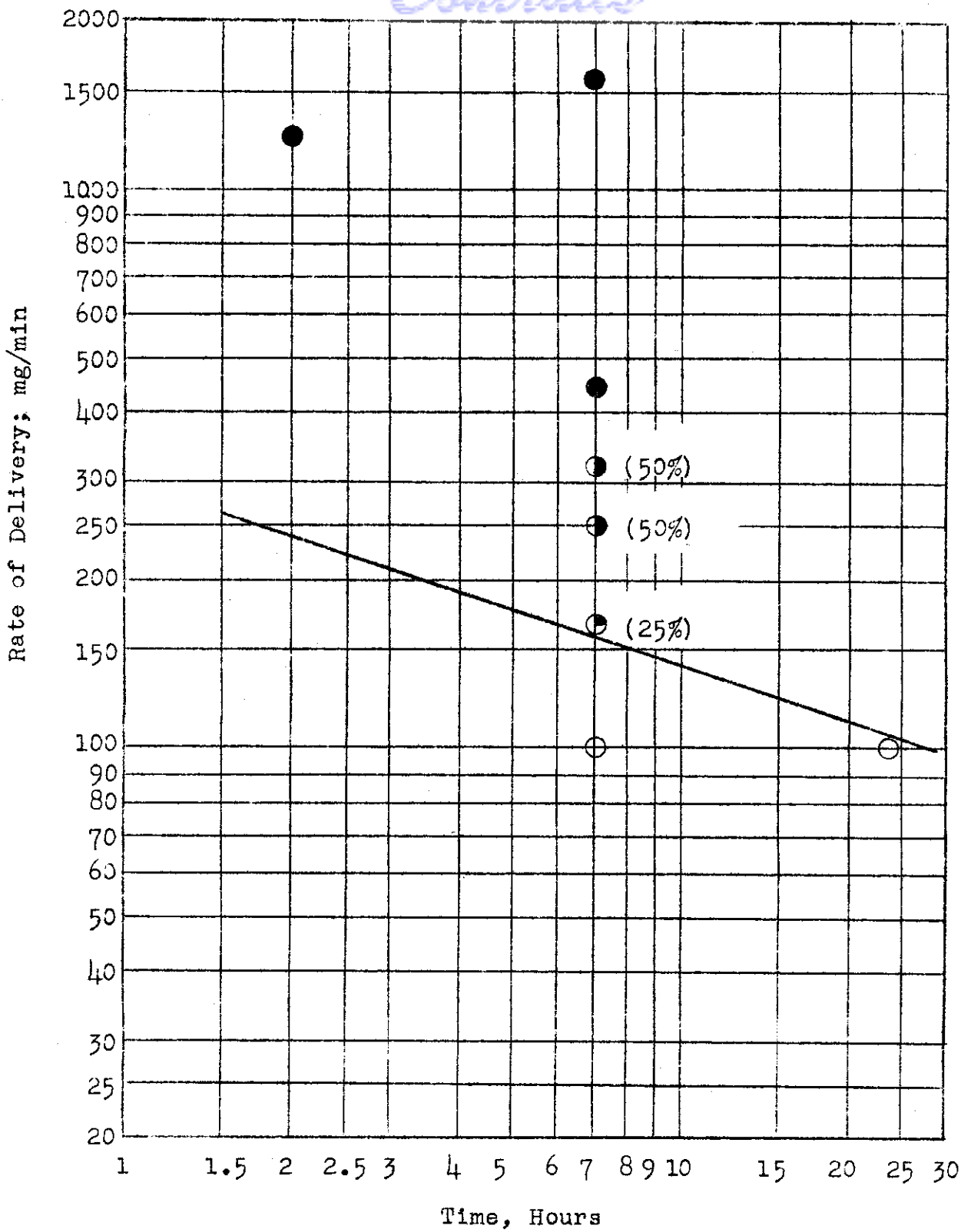


Figure 14. Fatalities Among Guinea Pigs Following the Inhalation of the Fog Formed by Dropping Ethyl Dibromobenzene into an Inconel Tube Heated to 900°F.

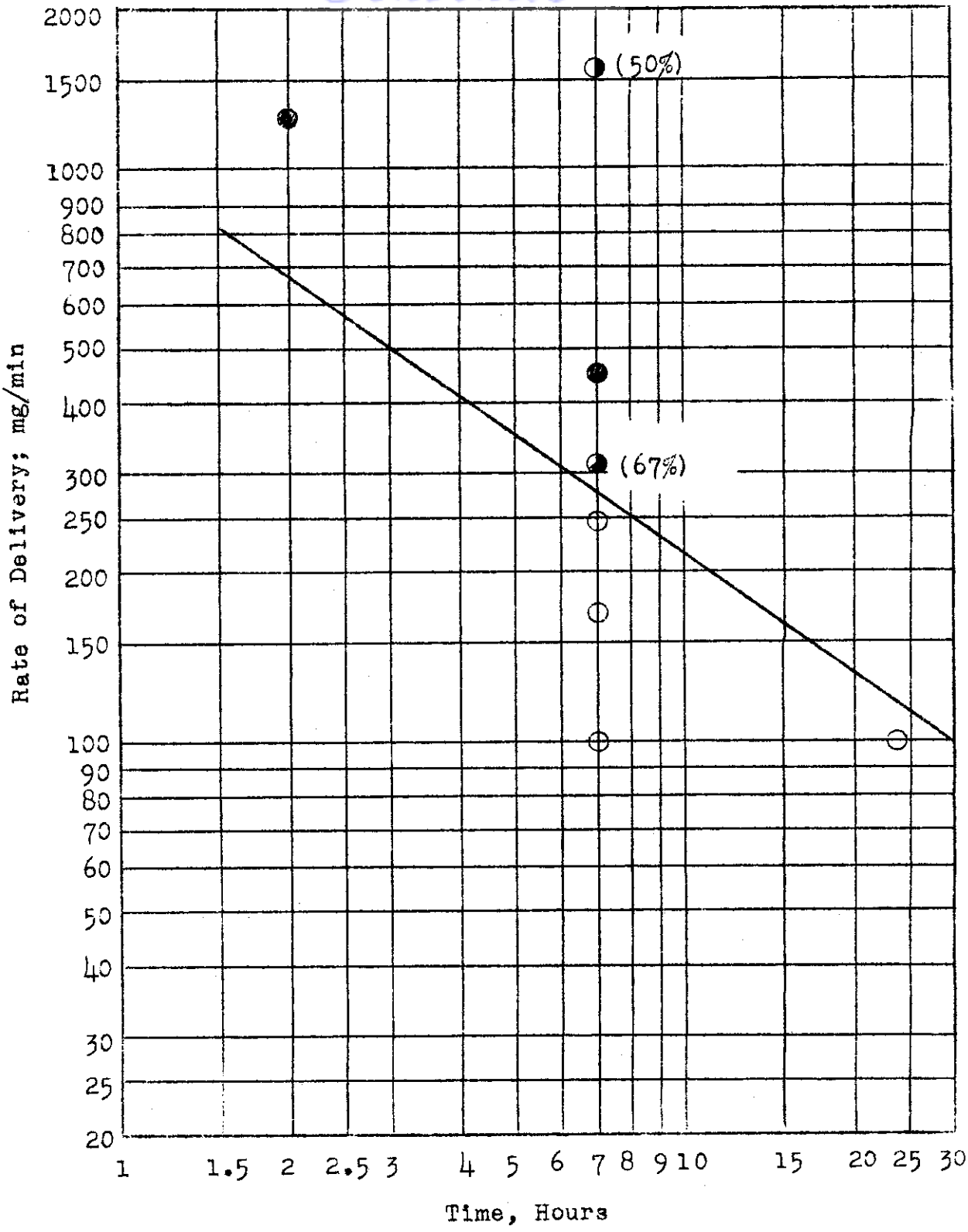


Figure 15. Fatalities Among Rabbits Following the Inhalation of the Fog Formed by Dropping Ethyl Dibromobenzene Into an Inconel Tube Heated to 900°F.

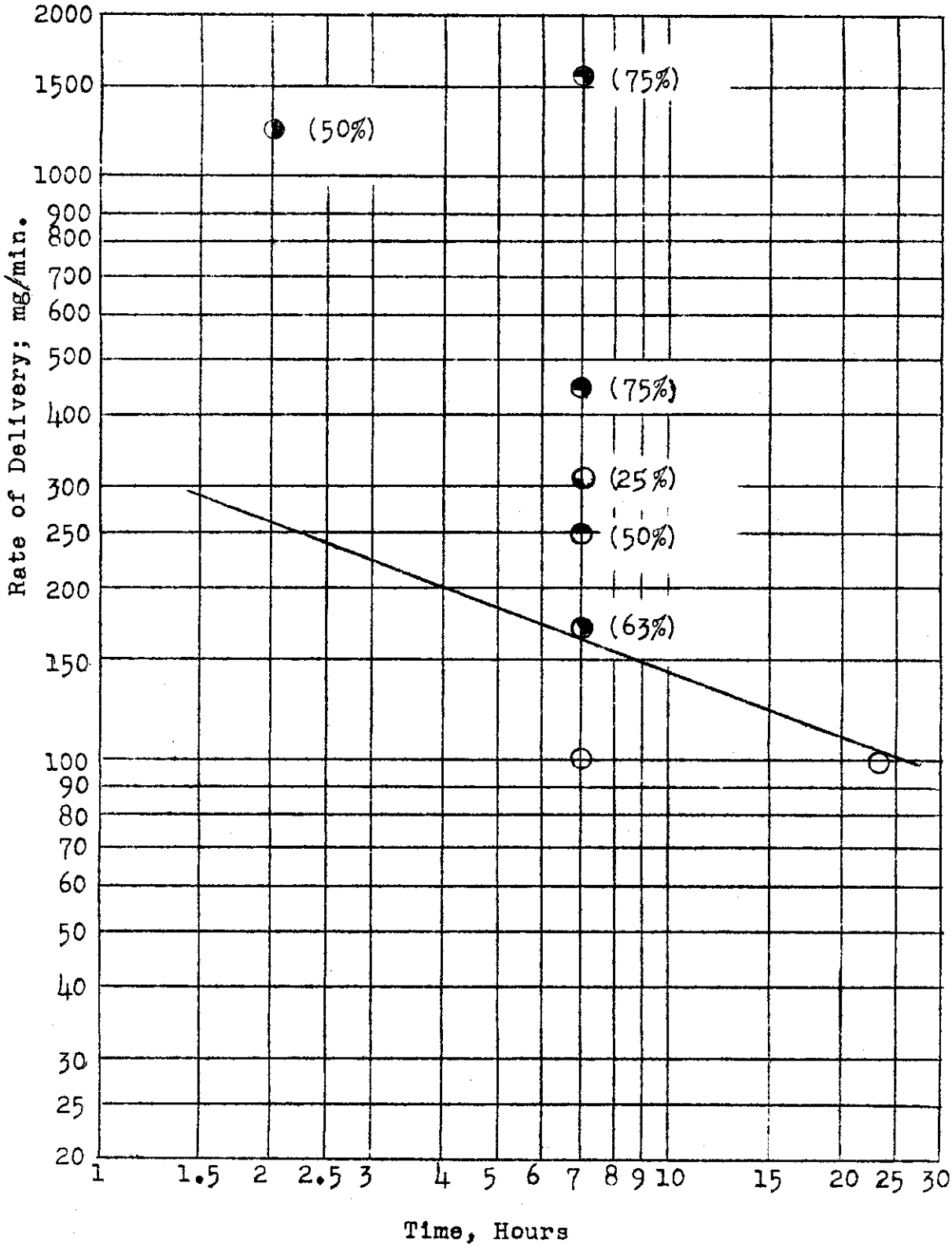


Figure 16. Fatalities Among Rats Following the Inhalation of the Fog Formed by Dropping Ethyl Dibromobenzene Into an Inconel Tube Heated to 900°F.

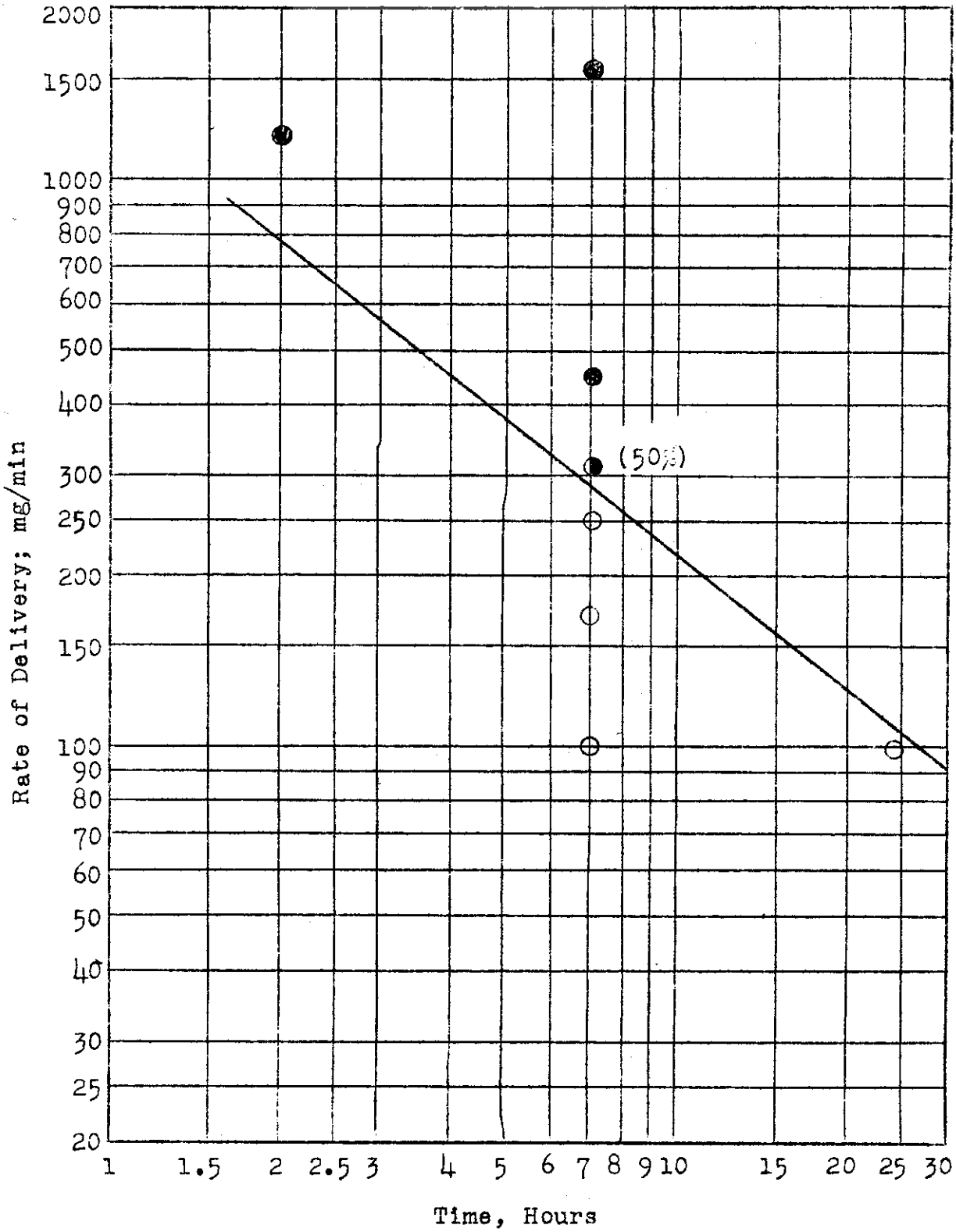


Figure 17. Fatalities Among Cats Following the Inhalation of the Fog Formed by Dropping Ethyl Dibromobenzene Into an Inconel Tube Heated to 900° F.

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