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**TOXICOLOGICAL EVALUATION
OF THE CONTACT AND VAPOR HAZARDS
OF VX AND THICKENED VX (TVX)
IN RABBITS**

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July 1985

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The degree of hazard from contacting residual, normally unseen and undetectable amounts of VX and thickened VX (TVX) on painted steel surfaces has been evaluated using the rabbit as a test model. Toxic hazard was evaluated by analyzing whole blood cholinesterase, observing toxic signs, and chemically analyzing each plate for residual VX content. Based on the test results, both polyurethane- and alkyd-painted steel test plates can absorb and then desorb significant amounts of VX, enough to produce a toxic effects in the rabbit. Direct contact is more hazardous than indirect vapor contact. The process accounting for the agent transport must include both a liquid-like transfer and a vapor transfer phenomenon.				
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Animal testing

Agent effects

Toxicity

Contact hazards

Vapor hazards

Residual hazards

PREFACE

The task described in this report was authorized under Project 1L162706A553, Chemical Systems and Procedures Technology, Technical Area 3-D. This work was started in August 1982 and completed in December 1982. The experimental data are contained in notebooks CSL-82-0018, CSL-82-0096, CSL-82-0146, and CSL-82-0185.

In conducting the research described in this report, the investigators adhered to the "Guide for the Care and Use of Laboratory Animals" as promulgated by the Committee on Revision of the Guide for Laboratory Animals Facilities and Care of the Institute of Laboratory Animal Resources, National Research Council. These investigations were also performed in accordance with requirements of AR 70-18, Laboratory Animals, Procurement, Transportation, Use, Care, and Public Affairs, and approved by the Laboratory Animal Use and Review Committee (LAURC), Chemical Research & Development Center, (Protocol Number 21082190000, approved 16 July 1982).

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The information in this document has not been cleared for release to the general public.

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TOXICOLOGICAL EVALUATION OF THE CONTACT AND VAPOR HAZARDS OF VX AND THICKENED VX (TVX) IN RABBITS

1. INTRODUCTION

A recently completed study determined the contact hazard of HD (distilled mustard) and THD (thickened distilled mustard) on contaminated alkyd and polyurethane painted steel test plates to rabbit skin.¹ The results showed that HD is a hazard by both direct and vapor contact even after efficient removal of the liquid surface contamination with solvents. The current test was similar to the HD study except that VX and TVX (thickened VX) were substituted as the contaminating agents.

The term "contact hazard" has been defined by Klein² as: "Given a surface that has been contaminated with a liquid chemical agent and that surface undergoes a process after which the agent no longer can be detected as a liquid, contact hazard is that situation in which a toxicological hazard can result if an individual then touches that surface with bare skin."

This VX project was designed to test both vapor transport and liquid transport theories for transfer of contamination to a contacting surface. Sidman et al. have proposed a model that assumes that agent desorbs from a contaminated surface in the vapor phase and is subsequently sorbed into the contacting layer.³ Klein* has proposed a second model in which the contaminated surface is compared to a pseudo-liquid and the residual agent transfers across this barrier to the contacting surface, as between two immiscible liquids in contact. This transfer is at a higher rate than would be found for a vapor only transfer. This second model also incorporates the vapor transfer model.

These toxicological evaluations will test the two proposed models. If the number of deaths, toxic signs, and/or whole blood cholinesterase inhibition is greater for the direct contact phase of the test, then there is strong support for the liquid-vapor transport theory. If, however, the results are the same for each test phase (direct contact and vapor contact), then the transfer theory would be more vapor related.

The tests described in this report were conducted with VX and TVX as the agents and rabbits as the test subjects. The parameters examined were toxic signs and/or death, whole blood cholinesterase (ChE) activity, and chemical analysis of all test plates for residual agent, as well as a series of control plates

* Klein, J. M. Contact Hazard - A Possible Explanation.
Unpublished data.

for each paint. The data obtained are not to be directly correlated to possible human responses to a similar exposure. The data may, however, give an indication of the hazard possible in either a direct contact or a vapor contact with VX-contaminated surfaces painted according to military specifications.

The test plates used were painted in 1981 and were from the same stock tested in the 1981 HD and THD studies. The purpose of this test was not to compare paint types but to assess the vapor and contact hazard of a persistent chemical agent of low volatility. VX has a volatility of 10.5 mg/cu m at 25° C, and HD, used in the previous study,¹ has a volatility of 920 mg/cu m at 25° C.

2. MATERIALS

2.1 Chemical Agents.

- a. VX. A sample of VX, identified as #I38A, was obtained on 4 August 1982. It was analyzed for chemical purity by the Analytical Chemistry Branch using the wet test method and was 93.9% active.
- b. Thickened VX. An aliquot of VX, #I38A, was thickened to 1177.0 centistokes with PIBM (polyisobutylmethacrylate) thickener.

2.2 Steel Test Plates.

The steel test plates used were 1 inch by 2 inches cut from stock plates that measured 4 inches by 12 inches by 0.040 inch thick. These plates had been primed with epoxy primer MIL-P-52192B and were finished with the following paints:

a. Polyurethane Paint.

Polyurethane Forest Green

MIL-C-46168

#62441 Randolph Products

Thinner, polyurethane MIL-T-81772

Film thickness of 2.4-3.0⁺ mils

b. Alkyd Paint.

Alkyd enamel, Forest Green

MIL-E-52798A

Film thickness of 2.4-3.0⁺ mils

These plates were painted in 1981 and had been aged about 1 year prior to the initiation of this study. In addition to the normal aging procedures, the alkyd test plates were exposed for 40 hours to direct sunlight during the aging procedure.

2.3 Stainless-Steel Templates.

A stainless-steel template was designed to hold the 1- by 2-inch test plates at a distance of 1 cm from the test area on the rabbit's back.¹ The templates had a recessed lip to support the test plate and also had a 1/4-inch rim at the base to provide a border for sealing the template to the rabbit's back with tape. By sealing the template to the skin, a closed cell was created that prevented agent vapors from escaping the test site.

2.4 Rabbits.

A total of 576 adult New Zealand White rabbits of mixed sexes that weighed between 2.06 and 3.85 kg were used. They were procured commercially by the Animal Resources Branch, US Army Medical Research Institute of Chemical Defense (USAMRICD), and were issued for experimental use only after at least 7 days of quarantine and certification of good health. All rabbits were identified with numbered stainless-steel ear tags and, in addition, were sequentially numbered on the left ear with black ink.

2.5 Solvents and Reagents.

a. Isopropyl Alcohol.

MCB reagent grade, ACS, PX1835
MCB Mfg Chemists, Inc., associate of E. Merck,
Darmstadt, Germany, 2909 Highland Ave
Cincinnati, OH 45212

b. Acetone.

Chemical Commodities Agency, Inc.
27447 Pacific St
Highland, CA 92346

FSN: 6810-00-264-8955
Acetone, A.C.S., O-C-265B
Lot: SO42SO32J, mfd 8/82

The above solvents were used to remove agent from the painted test plates during agent-rinse-removal procedures.

2.6 Syringe.

The syringe used to deliver specific amounts of VX and TVX was calibrated by measuring the weight of agent, in mg, that could be delivered with the aid of micrometer-driven syringe adapter. The 1.0-ml syringe, #Q3585, used in these studies delivered VX at a rate of 5.95 micrometer divisions/milligram.

2.7 Agla-Micrometer Syringe Adaptor.

The Agla-Micrometer syringe adaptor is mechanically driven to deliver precise amounts of a liquid in either microgram or milligram quantities. The device was manufactured by Shardlow Micrometers, Ltd, Sheffield, England.

The syringe holder was manufactured by Burroughs Wellcome & Co., England.

3. METHODS

3.1 Animal Preparation and Handling.

The day before testing, rabbits were prepared by numbering, sexing, weighing, and collecting 1.0 ml of blood to be used for comparison in later ChE analysis. Initial bleeding was to be accomplished by using a lateral ear vein. However, this procedure did not provide a large enough sample of blood in a short time, so blood for all ChE analyses was taken via cardiac puncture. The afternoon prior to testing, the backs of all rabbits were clipped free of hair (approximate 150-sq cm area) using an electric small animal clippers to which a number 2 blade was attached, followed by

a number 40 blade. The number 40 blade is designed to clip hair to within 1/130 of an inch.* After clipping, rabbits were returned to their home cages. Test day procedures involved placing rabbits in groups of six (in numerical order) into approved chemical fume hoods (air flow of 150 lfpm $\pm$ 30). They were restrained in aluminum neck-collaring stanchions that had side walls to prevent contact with adjacent rabbits. All rabbits were left in the fume hoods during the test and were removed at 24 hours or at death, whichever occurred first.

During the test, rabbits were observed for onset of toxic signs, including twitching, tremors, convulsions, miosis, salivation, prostration, and death. All toxic signs were recorded at earliest observed onset and were later used in data analysis.

Post-exposure blood samples were taken for ChE analysis at death or at 24 hours, whichever occurred first.

When the tests were completed, all rabbits were decontaminated with 5% sodium hypochlorite (bleach) solution. The survivors were observed for an additional 24 hours, after which they were euthanatized with intravenous T-61 and incinerated. Dead animals were decontaminated and incinerated immediately.

3.2 Cholinesterase Analysis.

Cholinesterase activity was measured by a radiometric assay as described by Siakotos et al.⁴

Control analysis samples containing all reagents minus whole blood aliquots were run concurrently. Counts-per-minute (CPM) values from control samples were subtracted from the total CPM of test samples from pre- and post-exposure animals to obtain a net CPM value that was directly related to enzyme activity. Percent inhibition of ChE was calculated from pre- and post-exposure data, in the form of CPM, using the following formula.

$$\% \text{ ChE}_{\text{Inhibited}} = 1 - \frac{(\text{Net CPM}_{\text{Post-exposure sample}})}{(\text{Net CPM}_{\text{Pre-exposure sample}})} \times 100$$

Compounds producing negative values for ChE inhibition were not considered to be inhibitors of ChE.

* Specification for number 40 surgical blade, Oster Professional Products, Milwaukee, WI 53217

3.3 Preparation of Painted Steel Test Plates.

Precut steel test plates were selected for even, scratch-free paint surface (without measuring the thickness of each paint film) and wiped gently with soft toweling to remove any debris left by the cutting process. The plates were arranged horizontally in the fume hood for agent application. Care was taken to obtain an even surface under the plates and absorbent toweling was placed under them to prevent the hood surface from becoming contaminated.

3.4 Contamination of Test Plates.

After placement into the chemical fume hood, the prepainted steel test plates were contaminated with single drops of either VX or TVX. The sizes of these droplets were 25.0, 5.0, or 0.5 mg. The droplet sizes were attained by using a calibrated syringe that was attached to the micrometer drive unit of the Agla syringe device described earlier.

3.5 Aging Procedure for Test Plates.

All test plates were aged for 30 minutes within the chemical fume hood at a temperature of 75° F and hood face velocity of 150 lfpm $\pm$ 30. The purpose of the 30-minute aging was to allow the agent to "soak" into the paint and also to spread over as much of the plate surface as possible. This procedure was used on all test plates as well as for concurrent control plates. The control plates were used for chemical analysis only and had no contact with rabbit skin.

3.6 Contamination Removal Procedures for Test Plates.

Both VX and thickened VX (TVX) were removed from test plates by using solvents and methods recommended by the Physical Protection Division. For the removal of standard VX, isopropyl alcohol was used, and for TVX, acetone was used. Solvents were dispersed with a large syringe and a 16-gauge needle. Slight downward pressure was applied to the syringe plunger (no excessive force), and a stream of solvent was dispersed from side to side and top to bottom of the test plate, thereby flushing the agent and solvent away. Both the solvent and agent were flushed directly into a 1-gallon jar containing aqueous sodium hydroxide (10%) solution. This procedure was done within a chemical fume hood with face velocity of 150 lfpm $\pm$ 30. The solvents used, along with the volumes used for specified agent contamination, are shown in Table 1.

Table 1. VX and TVX Solvent Removal Procedures*

VX Type	Droplet size (mg)	Solvent	Volume (ml)	Approximate dispersal time (sec)
VX	25.0	Isopropyl alcohol	35.0	30.0
	5.0	Isopropyl alcohol	20.0	15.0
	0.5	Isopropyl alcohol	15.0	10.0
TVX	25.0	Acetone	35.0	30.0

* These agent removal procedures, along with solvent volumes, were prescribed by the Physical Protection Division.

3.7 Aging Procedures for Test Plates Following Agent Removal.

Following the 30-minutes aging in the chemical fume hood and the agent removal process, the test plates were handled by one of two procedures:

- a. "Worst" case test plates were immediately wafted dry within the fume hood and then either placed in contact with the test rabbits or, if used as controls, immediately placed into a solvent bottle to be analyzed for residual agent contamination.
- b. All other test plates were placed back on the floor of the chemical fume hood and further aged for either 15 minutes or 5 hours, dependent upon test design. Following this additional aging, test plates were placed into contact with test rabbit skin, and control plates were placed into appropriate solvent bottles and held for chemical analysis.

3.8 Procedures for Test Plate Contact With Test Animals.

After aging, all test plates were attached to the clipped area of the rabbits backs for either 15 or 60 minutes. These plates were secured to the rabbits by either of two procedures.

3.8.1 Direct Contact.

This involved attaching the contaminated test plates, contaminated side next to the rabbit skin, for either 15 or

60 minutes. Each test plate was secured with a strip of refrigeration tape that completely covered it. A single layer of polyethylene film was placed on top of this and taped to the rabbit on all four edges. When completed the test plate was, in effect, double sealed so agent vapors could not escape.

3.8.2 Vapor Contact.

To assess the vapor hazard from the contaminated test plates, plates equal in number to those used in the direct contact phase were placed 1.0 cm away from the skin of rabbits, contaminated side down. This distance was maintained by taping the test plate into a stainless-steel template which was previously taped to the rabbit.¹ In addition, a polyethylene film was placed over the assembly and taped to the rabbit to form a double containment. Vapor contact lasted for either 15 or 60 minutes.

3.9 Procedures for Test Plate Removal.

When the required contact time with the rabbits (15 or 60 minutes) was complete, the test plates were carefully removed from the rabbits and placed directly in individual glass jars containing 20.0 ml of isopropyl alcohol. All tape and polyethylene film were placed directly into decontamination solution, and the rabbits were immediately observed for gross toxic signs, such as twitching and tremors. These two signs were difficult to observe during the actual contact time because the protective/securing tape covered the entire test area.

3.9.1 Procedure for Rabbit Decontamination.

All test rabbits were considered contaminated following the contact period. Rabbit skin was decontaminated at death or at 24 hours by swabbing the test area with 5% bleach, which was allowed to contact the skin for 5 minutes. Dead rabbits were removed, placed in plastic lined disposal bags, and incinerated. All surviving rabbits remained in the hood for 24 hours before being transferred to their individual home cages and observed for an additional 24 hours. Before removal to their home cage, the 24-hour blood samples were taken for ChE analysis. Blood was taken from rabbits that died during the test at the moment of death. Death was determined by cessation of respiration.

3.9.2 Chemical Analysis for Residual Contamination on Test Plates.

Bottles containing 20.0 ml of isopropyl alcohol and the contaminated test plates were taken to Analytical Chemistry Branch for determination of residual VX. Plates were allowed to soak for at least 8 hours.

Analytical procedures were done by either the colorimetric or fluorimetric Schoenemann methods.^{5,6,*} The colorimetric method gave a minimum VX detection limit by the Hubaux and Vas⁷ detection program of 20 µg/20-ml sample and a maximum detection limit of 1200 µg/20-ml sample without a dilution factor. If a submitted sample was outside the colorimetric test limits, the fluorimetric method was used. This method had a minimum detection limit of 0.5 µg/20-ml sample and a maximum detection limit of 150 µg/20-ml sample by the Hubaux and Vas detection program. By using these two analytical methods, a range of 0.5 to 1200 µg/20-ml sample could be determined without a dilution factor.

These two analytical methods were selected because they were well proven, provided a rapid analysis that used the same solvent (isopropanol), and permitted analysis with little or no dilution factor.

3.9.3 Test Phases.

Tests were designed to determine the amount of residual VX contamination of polyurethane-painted test plates through a series of six test procedures. Each procedure was performed with six control plates, six plates that received direct contact with VX, and six plates that received contact via VX vapor for each of three doses of VX (25.0 mg, 5.0 mg, and 0.5 mg). The test format was repeated for the alkyd-painted plates. Then a series of polyurethane and alkyd test plates were tested with TVX at a single contamination density of 25.0 mg. Both direct and vapor contact were assessed for each of six test phases using the TVX. The six test phases are described in Table 2.

These six test phases were always done in the sequence shown in Table 2. A total of 54 test plates were analyzed for each test phase for the VX versus the polyurethane- and alkyd-painted plates, and a total of 36 test plates were analyzed for each test phase in the TVX studies. Therefore, 648 test plates were chemically analyzed in the VX test phases, and 216 test plates were chemically analyzed in the TVX test phases.

* Johnson, W. C., and Cayle, R. P. CRDL-TM 5-13. Estimation of VX by the Schoenemann Technique. July 1965. (No longer available)

Table 2. Test Phases Used in VX and TVX
Contact Hazards Studies

Test phase	Aging time (min)	Solvent rinse	Post-rinse aging (min)	Rabbit contact time (min)
I	30	No	0	60
II	30	Yes	0	60
III	30	Yes	15	60
IV	30	Yes	15	15
V	30	Yes	300	60
VI	30	Yes	300	15

As stated, all test plates were analyzed for residual contamination following the test procedures. Likewise all test plates were observed for best estimate of agent spread (% of total plate area) before the various rinse-age procedures. Table 3 shows the mean spread of VX and TVX on all test plates after the initial 30-minute aging and prior to the rinse-removal process. As is shown in Table 3, the mean spread for the VX on polyurethane test plates at doses of 25.0, 5.0, and 0.5 mg/plate was 86%, 58%, and 25%, respectively. For TVX the spread at a dose of 25.0 mg was 20% of the plate area. For the alkyd-painted plates, the spread for VX at 25.0, 5.0, and 0.5 mg/plate was 91%, 51%, and 11%, respectively, and for the TVX at 25.0 mg, it was 15% of the total test plate area.

Also shown in Table 3 are the mean residual VX contaminations in milligrams for control plates aged 30 minutes and with no rinse removal. The values are for six test plates each. These mean values should represent the amount of agent left on the test plates after the 30-minute aging and prior to rinse removal that would be available in the Phase I "worst case tests" to be described in the test results.

4. RESULTS

4.1 Phase I, Control Test, Polyurethane-Painted Test Plates, VX.

This test was the "worst case" for rabbits contacting contaminated plates that were aged for 30 minutes after VX application. Control plates that started with 25.0, 5.0, and 0.5 mg of VX per plate retained 21.485, 4.652, and 0.416 mg/plate (mean value), respectively. The plates used for direct contact had 2.232, 0.985, and 0.089 mg/plate left after 60 minutes of contact. All six rabbits exposed to the 25.0-, 5.0-, and 0.5-mg plates died and had 100% inhibition of whole blood ChE. The mean death time

Table 3. Mean Physical/Chemical Data for VX and TVX on Polyurethane-
and Alkyd-Painted Steel Test Plates Prior to Solvent Removal

Paint type	Agent type	Agent applied (mg)	Mean ^a agent spread (area) on test plates (%)	Mean ^b residual agent on control test plates after 30-min aging (mg)	Condition of agent after 30-min aging ^c
Polyurethane	VX	25.0	86.0	21.485	Wet
		5.0	58.0	4.652	Damp-wet
		0.5	25.0	0.416	Damp
	TVX	25.0	20.0	20.022	Wet
Alkyd	VX	25.0	91.0	17.439	Damp-wet
		5.0	51.0	4.155	Dry-damp
		0.5	11.0	0.384	Dry
	TVX	25.0	15.0	18.833	Wet

^aThe mean for all test plates was used for total experiment.

^bMean for control plates (six each) - no rinse-removal process

^cCondition of agent spot from outer edge to center based on visual determination

for the 25.0-mg group of rabbits was 10.2 minutes; for the 5.0-mg group it was 22.8 minutes; and for the 0.5-mg group it was 104.8 minutes. See Table 4.

Detailed biological response data are shown in Tables A-1 through A-3, and detailed chemical data are shown in Tables B-1 through B-3.

In contrast to the results of direct contact, 18 rabbits exposed by vapor contact for 60 minutes to the three contamination densities, although showing toxic signs in all cases, did not die. Mean ChE inhibition for the 25.0 mg-group was 77.6%; for the 5.0-mg group, 40.5%; and for the 0.5-mg group, 12.4%. Based on the amount of residual VX left on these plates (9.414 mg on the 25.0-mg plates, 3.981 mg on the 5.0-mg plates, and 0.383 mg on the 0.5-mg plates), all rabbits were exposed to many times the amount of VX required to produce death (ca 0.025 mg/kg). The 1.0-cm distance appears to restrict the transfer of VX from test plate to skin because of the low volatility of the agent.

4.2 Phase II, Polyurethane-Painted Test Plates, VX.

This was the first test with polyurethane-painted plates involving the isopropanol rinse-removal of the VX contamination. This rinse-removal process removed a minimum of 96.4% and a maximum of 99.6% of the VX from the test plates (Appendix B-1 to B-3). As is shown in Table 5, the six rabbits that had direct contact showed toxic signs at the 25.0- and 5.0-mg dose levels, but only two of six displayed toxic signs at the 0.5-mg level. Inhibition of whole blood ChE was 47.8% at the 25.0-mg dose and 49.8% at the 5.0-mg dose. The 0.5-mg dose inhibited the ChE only 6.1%. This is within the 10% error of the ChE test analysis and the value could actually be 0% inhibition. The groups that had vapor contact showed fewer toxic signs (Table 5), and the ChE inhibition was 24.1%, 40.0%, and 23.0%, respectively, for the 25.0-, 5.0-, and 0.5-mg doses. Detailed biological data are listed in Tables A-1 to A-3 and chemical data are listed in Tables B-1 to B-3. These data indicate that the isopropanol rinse is effective at reducing the contact hazard of VX in these painted steel test plates, and the low vapor pressure of the trapped VX (in or under the paint) does not present a lethal hazard to the rabbit if contacted for up to 1 hour.

4.3 Phase III, Polyurethane-Painted Test Plates, VX.

This test involved aging the contaminated and rinsed test plates for 15 minutes prior to 60-minutes contact with rabbits. Shown in Table 6 are the numbers of animals with toxic signs and the degree of whole blood ChE inhibition. One rabbit died from direct contact at the high dose. This death occurred at 134 minutes after initial contact with the test plate. Direct

Table 4. Sixty-Minute Exposure of Rabbits to VX on Polyurethane-Painted Test Plates Aged Thirty Minutes and Not Rinsed

Exposure method	Agent applied (mg)	Residual agent ^a following aging and/or contact	Number of animals in group of six responding		Percent whole blood ChE inhibition
		(mean) ^b (mg)	Toxic signs ^c	Death	(mean) ^b
Control (none)	25.0	21.485	N/A	N/A	N/A
	5.0	4.652			
	0.5	0.416			
Direct contact	25.0	2.232	6	6	100.0
	5.0	0.985	6	6	100.0
	0.5	0.089	6	6	100.0
Vapor contact	25.0	9.414	6	0	77.6
	5.0	3.981	6	0	40.5
	0.5	0.383	6	0	12.4

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any/or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

Table 5. Sixty-Minute Exposure of Rabbits to VX on Polyurethane-Painted Test Plates Aged Thirty Minutes and Rinsed with Isopropanol

Exposure method	Agent applied (mg)	Residual agent ^a following aging and/or contact	Number of animals in group of six responding		Percent whole blood ChE inhibition
		(mean) ^b (mg)	Toxic signs ^c	Death	(mean) ^b
Control (none)	25.0	0.413	N/A	N/A	N/A
	5.0	0.181			
	0.5	0.005			
Direct contact	25.0	0.165	6	0	47.8
	5.0	0.124	6	0	49.8
	0.5	0.002	2	0	6.1
Vapor contact	25.0	0.247	2	0	24.1
	5.0	0.148	4	0	40.1
	0.5	0.006	1	0	23.0

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

Table 6. Sixty-Minute Exposure of Rabbits to VX on Polyurethane-Painted Test Plates Aged Thirty Minutes, Rinsed with Isopropanol, and Aged Fifteen Minutes

Exposure method	Agent applied (mg)	Residual agent ^a following aging and/or contact	Number of animals in group of six responding		Percent whole blood ChE inhibition
		(mean) ^b (mg)	Toxic signs ^c	Death	(mean) ^b
Control (none)	25.0	0.275	N/A	N/A	N/A
	5.0	0.213			
	0.5	0.002			
Direct contact	25.0	0.310	6	1	71.0
	5.0	0.070	6	0	11.3
	0.5	0.002	1	0	4.6
Vapor contact	25.0	0.297	3	0	7.0
	5.0	0.164	1	0	11.4
	0.5	0.012	2	0	12.0

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

contact with 25.0 mg of VX produced a mean ChE inhibition of 71.0%, with 5.0 mg, 11.3%, and with 0.5 mg, 4.6%. With the vapor contact, only 6 of 18 rabbits had toxic signs, and ChE depression ranged from 7.0% to 12.0%. Detailed biological and chemical data are shown in Tables A-1 to A-3 and B-1 to B-3, respectively.

4.4 Phase IV, Polyurethane-Painted Test Plates, VX.

This test was similar to that discussed in part 4.3 except that the rabbits were exposed 15 minutes rather than 60 minutes. The shorter exposure time eliminated the toxic effects of the 0.5-mg dose by direct contact and the 0.5-mg and 5.0-mg dose by vapor contact. AS shown in Table 7, the 25.0-mg dose by direct contact produced toxic signs in six rabbits and a mean ChE inhibition of 56.3%. The 5.0-mg dose produced toxic signs in four of six animals and ChE inhibition of 43.4%. By vapor contact, only one rabbit showed any toxic signs and there was no significant ChE inhibition. Detailed biological and chemical data are listed in Tables A-1 to A-3 and B-1 to B-3, respectively.

4.5 Phase V, Polyurethane-Painted Test Plates, VX.

In an attempt to eliminate all residual VX contamination from rinsed test plates, the aging period after rinsing was increased from 15 minutes to 5 hours. Chemical analysis of the test plates revealed that residual contamination was reduced to between 96.6% and 99.7% of control. This reduction is also reflected in the decreased number of rabbits showing toxic signs (Table 8) and the reduced level of ChE inhibition. In groups having direct contact, only 6 to 18 rabbits showed any toxic signs, and, in the groups having vapor contact, only 1 of 18 rabbits showed toxic signs. By direct contact the 25.0- and 5.0-mg plates did produce some degree of ChE inhibition, 25.2% and 18.6%, respectively. The plates used for vapor contact produced insignificant inhibition of whole blood ChE.

Detailed biological response data are shown in Tables A-1 to A-3, and chemical data are listed in Tables B-1 to B-3.

4.6 Phase VI, Polyurethane-Painted Test Plates, VX.

This test was similar to 4.5 above except that the exposure time was reduced to 15 minutes rather than 60 minutes. Table 9 lists the data for this test and shows that 8 of 18 rabbits developed some toxic effects from direct contact. Although ChE depression was moderate to severe at the 25.0- and 5.0-mg levels, 71.9% and 58.6%, respectively, the degree of toxic signs observed (twitching) generally would not indicate this degree of ChE

Table 7. Fifteen-Minute Exposure of Rabbits to VX on Polyurethane-Painted Test Plates Aged Thirty Minutes, Rinsed with Isopropanol, and Aged Fifteen Minutes

Exposure method	Agent applied (mg)	Residual agent ^a following aging and/or contact	Number of animals in group of six responding		Percent whole blood ChE inhibition
		(mean) ^b (mg)	Toxic signs ^c	Death	(mean) ^b
Control (none)	25.0	0.301	N/A	N/A	N/A
	5.0	0.254			
	0.5	0.005			
Direct contact	25.0	0.415	6	0	56.3
	5.0	0.244	4	0	43.4
	0.5	0.001	0	0	9.2
Vapor contact	25.0	0.385	1	0	3.8
	5.0	0.201	0	0	1.1
	0.5	0.005	0	0	0.2

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

Table 8. Sixty-Minute Exposure of Rabbits to VX on Polyurethane-Painted Test Plates Aged Thirty Minutes, Rinsed with Isopropanol, and Aged Five Hours

Exposure method	Agent applied (mg)	Residual agent ^a following aging and/or contact	Number of animals in group of six responding		Percent whole blood ChE inhibition
		(mean) ^b (mg)	Toxic signs ^c	Death	(mean) ^b
Control (none)	25.0	0.236	N/A	N/A	N/A
	5.0	0.168			
	0.5	0.003			
Direct contact	25.0	0.122	4	0	25.2
	5.0	0.090	2	0	18.6
	0.5	0.002	0	0	0.0
Vapor contact	25.0	0.069	0	0	10.8
	5.0	0.104	1	0	0.9
	0.5	0.003	0	0	6.0

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

Table 9. Fifteen-Minute Exposure of Rabbits to VX on Polyurethane-Painted Test Plates Aged Thirty Minutes, Rinsed with Isopropanol, and Aged Five Hours

Exposure method	Agent applied (mg)	Residual agent ^a following aging and/or contact	Number of animals in group of six responding		Percent whole blood ChE inhibition
		(mean) ^b (mg)	Toxic signs ^c	Death	(mean) ^b
Control (none)	25.0	0.132	N/A	N/A	N/A
	5.0	0.082			
	0.5	0.001			
Direct contact	25.0	0.129	3	0	71.9 ^d
	5.0	0.110	5	0	58.6 ^d
	0.5	0.006	0	0	3.0 ^d
Vapor contact	25.0	0.140	0	0	66.2 ^d
	5.0	0.070	0	0	23.6 ^d
	0.5	0.003	0	0	14.6 ^d

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

^dValues are questionable because of scintillation counter malfunction.

inhibition. The 18 rabbits exposed by vapor contact displayed no visible toxic signs; however, ChE activity was inhibited significantly at the 25.0-mg dose (66.2%) and slightly at the 5.0- and 0.5-mg doses. The degree of inhibition observed is questioned by the investigators because it does not seem to correlate with the limited toxic signs observed. It is strongly suspected that the scintillation counter was malfunctioning at the time these values were obtained.

Detailed biological data and chemical data for this test are shown in Tables A-1 to A-3 and B-1 to B-3, respectively.

4.7 Control Test, Phase I, Alkyd-Painted Test Plates, VX.

As with the polyurethane test plates, the alkyd-painted test plates were contaminated at three levels, 25.0, 5.0, and 0.5/mg plate. The data for the first test (worst case), where rabbits were exposed for 60 minutes to VX on unrinsed plates, are shown in Table 10. The control plates show 17.439, 4.155, and 0.384 mg/plate residual VX left after the 30-minute aging of plates that had initial applications of 25.0, 5.0, and 0.5 mg/plate, respectively. Following 60 minutes of direct contact, these levels were 10.313, 4.594, and 0.292 mg/plate, respectively. At the 25.0-mg/plate contamination level, 6/6 rabbits died, and ChE activity was inhibited 99.9%. Likewise, at the 5.0-mg/plate level, 6/6 rabbits died, and ChE activity was inhibited 99.5%. At the 0.5-mg/plate level, 0/6 rabbits died, and ChE activity was inhibited 56.0%. In the test using polyurethane-painted plates at this same level, 6/6 rabbits died with 100% ChE inhibition.

By the vapor contact at the 25.0-mg/plate level, 6/6 rabbits showed toxic signs; however, ChE activity was inhibited only 9.4%. At the 5.0-mg/plate level, 3/6 rabbits showed toxic signs, with 22.6% ChE inhibition, while at the 0.5-mg/plate level, only 2/6 rabbits showed toxic signs, with ChE inhibition amounting to 12.8%. Looking at the amount of residual VX recovered from these test plates, more severe toxic effects would be expected, especially by the vapor route. When compared to the tests using polyurethane (Table 4), these tests seem to indicate that the alkyd paint may trap the VX and not permit the vapor to effectively flux from the paint.

Detailed biological and chemical data for this "worst case" test are shown in Tables A-4 to A-6 and B-4 and B-6, respectively.

4.8 Phase II, Alkyd-Painted Test Plates, VX.

In this experiment, alkyd-painted test plates contaminated with VX were rinsed with isopropanol after the initial 30-minute

Table 10. Sixty-Minute Exposure of Rabbits to VX on Alkyd-Painted Test Plates
Aged Thirty Minutes and Not Rinsed

Exposure method	Agent applied (mg)	Residual agent ^a following aging and/or contact	Number of animals in group of six responding		Percent whole blood ChE inhibition
		(mean) ^b (mg)	Toxic signs ^c	Death	(mean) ^b
Control (none)	25.0	17.439	N/A	N/A	N/A
	5.0	4.155			
	0.5	0.384			
Direct contact	25.0	10.313	6	6	99.9
	5.0	4.594	6	6	99.5
	0.5	0.292	6	0	56.0
Vapor contact	25.0	14.200	6	0	9.4
	5.0	5.012	3	0	22.6
	0.5	0.318	2	0	12.8

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

aging process. No additional aging was done after the rinse procedure, and the test plates were placed in contact with the rabbits for 60 minutes. The results shown in Table 11 indicate that there is no great difference between the amount of residual VX left in the control plates and the plates that were in contact with the rabbits for 60 minutes. Because of the amount of VX left in the plates, it would be anticipated that severe toxic signs and perhaps death would result from 60 minutes of direct contact. All 18 rabbits did develop toxic signs; however, none developed severe signs such as convulsions or prostration (Tables A-4 to A-6). Cholinesterase inhibition was 74.7% (25.0-mg dose), 73.6% (5.0-mg dose), and 28.7% (0.5-mg dose). The results of the vapor contact indicate that there was as much VX available to these 18 rabbits as in the direct contact test, but the number of toxic signs observed were fewer and the degree of ChE inhibition was much reduced, 9.0% to 18.2%. Detailed biological data and chemical data are shown in Tables A-4 to A-6 and B-4 to B-6, respectively.

4.9 Phase III, Alkyd-Painted Test Plates, VX.

This was the first test with alkyd-painted test plates that had additional aging (15 minutes) after the isopropanol rinse procedure removed the surface VX contamination. As shown in Table 12, even with additional aging, considerable VX remained on the test plates at all three contamination levels. In fact, the contamination levels do not look different than those for the test just discussed (4.8). The results of the direct contact portion of this test show that 18/18 rabbits developed toxic signs and 1/6 rabbits of the 25.0-mg group died. Cholinesterase inhibition was 79.7% (25.0 mg), 62.8% (5.0 mg), and 17.3% (0.5 mg). By contrast, in the vapor contact test, only 10/18 animals showed toxic signs, none from the 0.5-mg plates. Cholinesterase inhibition by the vapor procedure was 3.7% (25.0 mg), 7.9% (5.0 mg), and 5.1% (0.5 mg), respectively. These ChE inhibitions are considered insignificant and within the error of the analysis procedure. Based on these results, it would appear that the VX is once again either trapped in the alkyd paint or, because of its low vapor pressure, is not able to effectively transfer between the paint and rabbit skin.

Detailed biological and chemical data for this test are shown in Tables A-4 to A-6 and B-4 to B-6, respectively.

4.9.1 Phase IV, Alkyd-Painted Test Plates, VX.

This test was similar to test I (Phase III) except that the contact time of the plates to rabbits was 15 minutes rather than 60 minutes. The results, Table 13, show that, based on ChE inhibition, this was a more severe test. However, the total number of toxic signs observed in the rabbits were fewer: 13/18 (direct

Table 11. Sixty-Minute Exposure of Rabbits to VX on Alkyd-Painted Test Plates
Aged Thirty Minutes and Rinsed with Isopropanol

Exposure method	Agent applied (mg)	Residual agent ^a following aging and/or contact	Number of animals in group of six responding		Percent whole blood ChE inhibition
		(mean) ^b (mg)	Toxic signs ^c	Death	(mean) ^b
Control (none)	25.0	1.684	N/A	N/A	N/A
	5.0	0.850			
	0.5	0.171			
Direct contact	25.0	1.346	6	0	74.7
	5.0	0.877	6	0	73.6
	0.5	0.162	6	0	28.7
Vapor contact	25.0	1.710	4	0	9.0
	5.0	0.870	6	0	16.3
	0.5	0.182	2	0	18.2

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

Table 12. Sixty-Minute Exposure of Rabbits to VX on Alkyd-Painted Test Plates Aged Thirty Minutes, Rinsed with Isopropanol, and Aged Fifteen Minutes

Exposure method	Agent applied (mg)	Residual agent ^a following aging and/or contact	Number of animals in group of six responding		Percent whole blood ChE inhibition
		(mean) ^b (mg)	Toxic signs ^c	Death	(mean) ^b
Control (none)	25.0	1.662	N/A	N/A	N/A
	5.0	0.860			
	0.5	0.112			
Direct contact	25.0	1.572	6	1	79.7
	5.0	0.737	6	0	62.8
	0.5	0.125	6	0	17.3
Vapor contact	25.0	1.638	5	0	3.7
	5.0	0.819	5	0	7.9
	0.5	0.132	0	0	5.1

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any/or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

Table 13. Fifteen-Minute Exposure of Rabbits to VX on Alkyd-Painted Test Plates
Aged Thirty Minutes, Rinsed with Isopropanol, and Aged Fifteen Minutes

Exposure method	Agent applied (mg)	Residual agent ^a following aging and/or contact	Number of animals in group of six responding		Percent whole blood ChE inhibition
		(mean) ^b (mg)	Toxic signs ^c	Death	(mean) ^b
Control (none)	25.0	2.376	N/A	N/A	N/A
	5.0	1.020			
	0.5	0.156			
Direct contact	25.0	1.769	6	0	75.2
	5.0	0.845	5	0	82.2
	0.5	0.091	2	0	37.7
Vapor contact	25.0	2.015	3	0	54.2
	5.0	0.946	0	0	44.7
	0.5	0.127	0	0	12.6

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any/or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

contact) and only 3/18 (vapor contact). Note also that there was somewhat more VX available in this series of test plates than was available in the previous test (4.9 Phase III). These increased ChE inhibitions, especially in the vapor test portion, may be simply due to biological variations in the test rabbits, i.e., thinner skin, thinner hair growth. These data tend to point out the fact that ChE inhibition can occur without evident external toxic signs. Detailed biological and chemical data are listed in Tables A-4 to A-6 and B-4 and B-6, respectively.

4.9.2 Phase V, Alkyd-Painted Test Plates, VX.

The next two tests were attempts to remove additional VX from the alkyd test plates by increasing the aging time from 15 minutes to 5 hours after the isopropanol rinse. As shown in Table 14, after 60 minutes of direct contact with these test plates, 18/18 rabbits developed observable toxic signs and significant ChE depression (60%) at the 25.0- and 5.0-mg doses and some degree (19.1%) of ChE depression at the 0.5-mg dose. Those animals exposed by vapor contact, showed only one case of toxic signs and insignificant depression. The residual contamination in the test plates was reduced, but not enough to eliminate the toxic hazard of 1-hour direct contact with rabbit skin.

Detailed biological and chemical data are shown in Tables A-4 to A-7 and B-4 to B-6, respectively.

4.9.3 Phase VI, Alkyd-Painted Test Plates, VX.

This test was different than the previous test only in that the contact time of the contaminated alkyd test plates with rabbit skin was for 15 minutes rather than for 60 minutes. Looking at the data in Table 15, it can be seen that 17/18 rabbits in the direct contact phase showed toxic signs, and ChE inhibition was similar to that observed during the 60-minute test (Table 14). By vapor contact, 1/18 rabbits showed toxic signs; however, there was a greater degree of whole blood ChE inhibition than for the 60-minute exposure.

Detailed biological and chemical data are shown in Tables A-1 to A-4 and B-1 to B-4, respectively.

4.9.4 Phase I, Control Test, TVX on Painted Steel Test Plates.

The next series of six tests used thickened (TVX) and the same testing procedures used for VX. However, only one contamination level, 25.0 mg/plate, was used and both the polyurethane and alkyd test plates were tested simultaneously.

Table 14. Sixty-Minute Exposure of Rabbits to VX on Alkyd-Painted Test Plates
Aged Thirty Minutes, Rinsed with Isopropanol, and Aged Five Hours

Exposure method	Agent applied (mg)	Residual agent ^a following aging and/or contact	Number of animals in group of six responding		Percent whole blood ChE inhibition
		(mean) ^b (mg)	Toxic signs ^c	Death	(mean) ^b
Control (none)	25.0	1.679	N/A	N/A	N/A
	5.0	0.658			
	0.5	0.094			
Direct contact	25.0	1.585	6	0	60.2
	5.0	0.705	6	0	60.3
	0.5	0.076	6	0	19.1
Vapor contact	25.0	1.314	1	0	8.8
	5.0	0.568	0	0	9.4
	0.5	0.095	0	0	12.6

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any/or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

Table 15. Fifteen-Minute Exposure of Rabbits to VX on Alkyd-Painted Test Plates Aged Thirty Minutes, Rinsed with Isopropanol, and Aged Five Hours

Exposure method	Agent applied (mg)	Residual agent ^a following aging and/or contact	Number of animals in group of six responding		Percent whole blood ChE inhibition
		(mean) ^b (mg)	Toxic signs ^c	Death	(mean) ^b
Control (none)	25.0	1.314	N/A	N/A	N/A
	5.0	0.621			
	0.5	0.083			
Direct contact	25.0	1.330	6	0	73.5
	5.0	0.571	6	0	63.4
	0.5	0.086	5	0	8.8
Vapor contact	25.0	1.382	1	0	53.4
	5.0	0.573	0	0	24.9
	0.5	0.080	0	0	5.8

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

The first experiment was the "worst case;" that is, following test plate contamination with 25.0-mg TVX/plate, these plates were aged 30 minutes and then placed immediately in contact with rabbit skin by both direct and vapor contacts. Both polyurethane and alkyd plates were tested simultaneously in groups of six for each paint type and contacting procedure.

Table 16 shows that the results obtained from the polyurethane and alkyd plates were similar. By direct contact TVX-contaminated paint types produced death in 6/6 rabbits with complete whole blood ChE inhibition. By the vapor contact procedure, 5/6 rabbits in both cases showed toxic signs, and no deaths resulted. Whole blood ChE activity was inhibited by about one-third of normal in both vapor contacts. Note in Table 16 the large amounts of TVX on the test plates when no rinse-removal was performed.

Detailed biological and chemical data for these tests are shown in Tables A-7 and B-7 for the polyurethane paint and in Tables A-8 and B-8 for the alkyd paint. One point of interest in the chemical data is that the TVX did not spread over either the polyurethane- or alkyd-painted plates to the extent that standard VX did. The TVX spread on the polyurethane plates, using mean values, was from 15% to 26% of the surface area; for the alkyd plates, it was 12% to 17% of the surface area. These spreads contrast to the 75% to 98% spreads seen with VX on the same type plates.

In the next five tests, surface contamination was rinsed/removed with acetone. Acetone was used because isopropanol would not remove the PIBM used to thicken VX.

4.9.5 Phase II, TVX on Painted Steel Test Plates.

This was the first test involving TVX and the acetone rinse-removal process. Both polyurethane- and alkyd-painted test plates were contaminated with 25.0-mg TVX/plate. Following the 30-minute aging, the plates were each rinsed with 35.0 ml of acetone. As soon as the acetone dried (about 30 seconds), the plates were placed in contact with rabbit skin by both the direct and vapor contact procedures for 60 minutes and then removed. During the 24-hour observation, no rabbits showed any toxic signs. The 24-hour whole blood ChE analysis showed no significant inhibition from either type of paint (Table 17). The residual agent left in the alkyd plates was significantly more than in the polyurethane plates, but was still below the amount that would produce toxic effects in rabbits, even after 60 minutes of direct contact. The amount of agent left in the polyurethane paint was less than 0.1% of that applied and, in the alkyd paint, it was less than 1.0% of the total applied.

Table 16. Sixty-Minute Exposure of Rabbits to TVX on Painted Steel
Test Plates Aged Thirty Minutes

Exposure method	Agent applied (mg)	Residual agent ^a following aging and/or contact	Number of animals in <u>group of six responding</u>		Percent whole blood ChE inhibition
		(mean) ^b (mg)	Toxic signs ^c	Death	(mean) ^b
Polyurethane paint					
Control (none)	25.0	20.022	N/A	N/A	N/A
Direct contact	25.0	2.051	6	6	99.6
Vapor contact	25.0	16.578	5	0	33.2
Alkyd paint					
Control (none)	25.0	18.833	N/A	N/A	N/A
Direct contact	25.0	6.336	6	6	100.0
Vapor contact	25.0	15.266	5	0	32.0

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any/or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

Table 17. Sixty-Minute Exposure of Rabbits to TVX on Painted Steel Plates
Aged Thirty Minutes and Rinsed with Acetone

<u>Exposure method</u>	<u>Agent applied</u> (mg)	<u>Residual agent^a</u> <u>following aging</u> <u>and/or contact</u>	<u>Number of animals in</u> <u>group of six responding</u>		<u>Percent</u> <u>whole blood</u> <u>ChE inhibition</u>
		<u>(mean)^b</u> (mg)	<u>Toxic signs^c</u>	<u>Death</u>	<u>(mean)^b</u>
Polyurethane paint					
Control (none)	25.0	0.008	N/A	N/A	N/A
Direct contact	25.0	0.009	0	0	3.4
Vapor contact	25.0	0.009	0	0	12.0
Alkyd paint					
Control (none)	25.0	0.199	N/A	N/A	N/A
Direct contact	25.0	0.230	0	0	12.8
Vapor contact	25.0	0.240	0	0	0.0

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any/or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

Detailed biological and chemical data from this test are shown in Tables A-7, A-8, B-7, and B-8, respectively.

4.9.6 Phase III, TVX on Painted Steel Test Plates.

This test involved an additional 15-minute aging of the test plates after they were rinsed with acetone (35.0 ml/plate). The plates were then placed on rabbits (direct and vapor contact) for 60 minutes. The 12 rabbits that had contact with contaminated polyurethane plates showed no toxic signs or ChE depression. With the alkyd-painted plates, one rabbit of six exposed by direct contact developed a mild twitch about 113 minutes after contact. No toxic effects were observed with the alkyd plates after vapor exposure. Table 18 shows that less than 0.1% of the original TVX was left on the polyurethane plates, and less than 1.0% was left on the alkyd-painted plates.

Detailed test data for both the biological and chemical aspects are shown in Tables A-7, A-8, B-7, and B-8, respectively.

4.9.7 Phase IV, TVX on Painted Steel Test Plates.

This test was similar to the test above (Phase III) except that the exposure time changed from 60 to 15 minutes. As shown in Table 19, one rabbit exposed by direct contact to the contaminated polyurethane test plates developed toxic signs, including twitching and mild tremors. All other rabbits, by both direct and vapor contacts to contaminated polyurethane and alkyd plates, showed no toxic signs and also no ChE inhibition. The mean values of TVX found in these test plates was less than 0.1% for the polyurethane and less than 0.5% for the alkyd.

Detailed biological and chemical data are listed in Tables A-7, A-8, B-7, and B-8, respectively.

4.9.8 Phase V, TVX on Painted Steel Test Plates.

A test was done to determine if longer aging of the contaminated test plates would remove additional contamination. With this idea in mind, test plates were rinsed with acetone and then aged for 5 hours before being placed on rabbits for 60 minutes. The results (Table 20) show that after contact with either polyurethane or alkyd test plates no toxic signs appeared in any rabbits and no significant whole blood ChE inhibition occurred. The mean residual contamination left in the test plates, after the acetone rinse and the 5-hour aging, was 0.1% for the polyurethane

Table 18. Sixty-Minute Exposure of Rabbits to TVX on Painted Steel Test Plates
Aged Thirty Minutes, Rinsed with Acetone, and Aged Fifteen Minutes

<u>Exposure method</u>	<u>Agent applied (mg)</u>	<u>Residual agent^a following aging and/or contact (mean)^b (mg)</u>	<u>Number of animals in group of six responding</u>		<u>Percent whole blood ChE inhibition (mean)^b</u>
		<u>Toxic signs^c</u>	<u>Death</u>		
Polyurethane paint					
Control (none)	25.0	0.016	N/A	N/A	N/A
Direct contact	25.0	0.016	0	0	2.0
Vapor contact	25.0	0.016	0	0	7.3
Alkyd paint					
Control (none)	25.0	0.212	N/A	N/A	N/A
Direct contact	25.0	0.186	1	0	12.6
Vapor contact	25.0	0.205	0	0	5.2

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

Table 19. Fifteen-Minute Exposure of Rabbits to TVX on Painted Steel Test Plates
Aged Thirty Minutes, Rinsed with Acetone, and Aged Fifteen Minutes

Exposure method	Agent applied (mg)	Residual agent ^a following aging and/or contact	Number of animals in <u>group of six responding</u>		Percent whole blood ChE inhibition
		(mean) ^b (mg)	Toxic signs ^c	Death	(mean) ^b
Polyurethane paint					
Control (none)	25.0	0.012	N/A	N/A	N/A
Direct contact	25.0	0.007	1	0	0.0
Vapor contact	25.0	0.017	0	0	0.0
Alkyd paint					
Control (none)	25.0	0.095	N/A	N/A	N/A
Direct contact	25.0	0.086	0	0	0.0
Vapor contact	25.0	0.083	0	0	0.0

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any/or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

Table 20. Sixty-Minute Exposure of Rabbits to TVX on Painted Steel Test Plates
Aged Thirty Minutes, Rinsed with Acetone, and Aged Five Hours

Exposure method	Agent applied (mg)	Residual agent ^a following aging and/or contact	Number of animals in group of six responding		Percent whole blood ChE inhibition
		(mean) ^b (mg)	Toxic signs ^c	Death	(mean) ^b
Polyurethane paint					
Control (none)	25.0	0.021	N/A	N/A	N/A
Direct contact	25.0	0.021	0	0	7.4
Vapor contact	25.0	0.018	0	0	6.9
Alkyd paint					
Control (none)	25.0	0.090	N/A	N/A	N/A
Direct contact	25.0	0.098	0	0	2.8
Vapor contact	25.0	0.115	0	0	10.5

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

plates and 0.4% to 0.5% in the alkyd plates. Detailed biological and chemical data are in Tables A-7, A-8, B-7, and B-8, respectively.

4.9.9 Phase VI, TVX on Painted Steel Test Plates.

The final test was to age the plates 5 hours after the acetone rinse and place them in contact with rabbit skin for 15 rather than 60 minutes. As was seen in test Phase V, no toxic signs were observed in any of the 24 rabbits. Only in the group that had direct contact with the polyurethane plates was there any whole blood ChE inhibition. The degree of this inhibition (mean value of 28%) is questionable and, at most, is a mild inhibition level. As shown in Table 21, the residual contamination in these test plates was less than 0.1% in the polyurethane plates and about 0.4% in the alkyd plates.

Detailed biological and chemical data are shown in Tables A-7, A-8, B-7, and B-8, respectively.

5. DISCUSSION

A series of 18 tests were conducted to assess the contact hazard from painted steel test plates contaminated with VX and TVX using the rabbit as the biological sensor. Along with observation for toxic signs, rabbit whole blood ChE was analyzed for the degree of inhibition, and all painted steel test plates were analyzed chemically for residual VX contamination following the various test procedures.

Figure 1 shows the mean residual VX and TVX on control test plates after test phases I through VI, and Figures 2 and 3 graphically show rabbit whole blood ChE inhibition for direct contact and vapor contact, respectively.

Test were designed to determine whether the agent contamination on test plates was transferred to the contacting surface (rabbit skin) as a vapor only, or if it was transferred by both a pseudo-liquid and vapor transfer mode. These two transfer theories were assessed by performing both direct and vapor contact tests with the rabbit as the sensor. If the residual agent on the test plates was transferred to the contacting surface only as a vapor, then the results of the direct and vapor contact tests would be similar. If, however, the transfer included both the pseudo-liquid and vapor transport methods, then the degree of the toxic effects observed would be greatest in rabbits having direct contact.

Table 21. Fifteen Minute Exposure of Rabbits to TVX on Painted Steel Test Plates Aged Thirty Minutes, Rinsed with Acetone, and Aged Five Hours

Exposure method	Agent applied (mg)	Residual agent ^a following aging and/or contact	Number of animals in <u>group of six responding</u>		Percent whole blood ChE inhibition
		(mean) ^b (mg)	Toxic signs ^c	Death	(mean) ^b
Polyurethane paint					
Control (none)	25.0	0.012	N/A	N/A	N/A
Direct contact	25.0	0.014	0	0	28.0
Vapor contact	25.0	0.017	0	0	11.0
Alkyd paint					
Control (none)	25.0	0.099	N/A	N/A	N/A
Direct contact	25.0	0.093	0	0	2.6
Vapor contact	25.0	0.098	0	0	0.8

^aBy chemical analysis

^bMean for group of six

^cToxic signs include any/or all of the following: twitching, tremors, convulsions, miosis, salivation, prostration.

Figure 1 shows the residual agent left in both the polyurethane and alkyd test plates for all six test phases. These data indicate that following the 30-minute aging, and prior to the solvent rinsing, both polyurethane- and alkyd-painted plates retained similar amounts of agent. However, after the rinse procedure, the amount of agent left in the polyurethane-painted plates was less in all test phases (Table 22). The difference being milligram amounts in the alkyd plates and microgram amounts in the polyurethane plates. Based on the amount of agent left in the alkyd plates, it would be expected that the animals contacting the alkyd plates would have shown significantly greater toxic effects. Figure 2 shows the degree of whole blood ChE inhibition for all rabbits exposed by direct contact. Note that in the Phase I tests, the ChE activity of all rabbits in the polyurethane paint groups was inhibited 100%, as were the alkyd paint groups except for those exposed to the 0.5-mg dose, which was inhibited only 56%. The remaining five test phases indicate generally more depression of ChE activity from the alkyd-painted plates. This would be expected since the amount of agent available from these plates was greater. Figure 3 shows a reverse trend for vapor contact: that is, the inhibition of ChE activity, although much reduced from that of the direct contact groups, is generally greater for the polyurethane-painted plates. These data show that the agent contained in the alkyd test plates is either not able to flux from the surface at a sufficient rate to present the same hazard seen from direct contact, or the 1-centimeter air barrier is sufficient space to significantly reduce the VX vapor hazard. These data indicate that the transfer of contamination from the source to the contacting surface is accomplished by both pseudo-liquid transfer as well as vapor transfer.

The data for TVX (Figures 1-3) show that if no free liquid TVX is present, the amount of residual contamination is probably not sufficient to be a severe toxic hazard. It would appear from our tests and subsequent analyses, that TVX does not spread over the surface of the test plates to the degree that nonthickened VX does. If removed from the surface by a solvent rinse, the residual contamination is reduced to a degree that no longer presents a significant contact hazard. The degree of vapor flux was also much reduced in Phase I control tests (Table 16) where no deaths occurred in 12 rabbits exposed by vapor contact for 60 minutes to residual contaminations as high as 16 mg/plate. By direct contact, contamination of this level produced death in 12/12 rabbits.

These data show that the hazard from low volatility agents such as VX may be much reduced if the free source of agent is removed from the contaminated surfaces either by decontamination or by an effective solvent removal. It has also been shown that the residual VX contained in the contaminated surface, following agent removal, may not present a significant hazard for short periods of direct contact or for extended periods of vapor contact.

Table 22. Summary of Residual VX on Control Test Plates*

<u>Paint type</u>	<u>Agent type</u>	<u>Agent dose (mg)</u>	<u>Residual agent 30-min aging, no rinse (%)</u>	<u>Residual agent 30-min aging, and rinse-removal (%)</u>	<u>Residual agent 30-min aging, rinse-removal, 15-min aging (%)</u>	<u>Residual agent 30-min Aging, rinse-removal, 5-hr aging (%)</u>
Polyurethane	VX	25.0	86.0	2.0	1.0	0.9
		5.0	93.0	4.0	4.0	3.4
		0.5	83.0	1.0	0.4	0.6
	TVX	25.0	80.1	0.03	0.06	0.08
Alkyd	VX	25.0	70.0	6.7	6.7	6.7
		5.0	83.0	17.0	17.2	13.2
		0.5	77.0	34.2	22.4	18.8
	TVX					
		25.0	75.3	0.8	0.9	0.4

*Mean percent residual agent for six test plates at each data point

5. CONCLUSIONS

Based on the results of this study, the following conclusions are made.

- a. Both polyurethane- and alkyd-painted steel test plates can absorb significant amounts of VX after 30 minutes of liquid contact.
- b. Rinsing contaminated polyurethane-painted steel test plates with isopropanol can remove 95⁺% or more of the total contamination.
- c. Rinsing contaminated alkyd-painted test plates with isopropanol may remove between 65% and 93% of the contamination.
- d. The degree of residual toxic hazard from VX-contaminated surfaces is greater by direct contact than by vapor contact.
- e. Even when no liquid agent is detectable, VX-contaminated surfaces can still emit sufficient agent to be a toxic hazard, especially by direct contact.
- f. Residual thickened VX (TVX) as contained in either polyurethane- or alkyd-painted steel test plates is not a significant hazard following an acetone rinse-removal process of the contamination from the painted surface.

ALL PHASES ARE VX-30 MINUTE AGE

I: CONTROL, NO RINSE
 II: RINSE, NO AGE
 III: RINSE, 15 MIN AGE
 IV: RINSE, 15 MIN AGE
 V: RINSE, 5 HR AGE
 VI: RINSE, 5 HR AGE

LEGEND
 ▨ 25.0 MG VX STARTING DOSE
 ■ 5.0 MG VX STARTING DOSE
 ▩ 0.5 MG VX STARTING DOSE
 □ 25.0 MG TVX STARTING DOSE

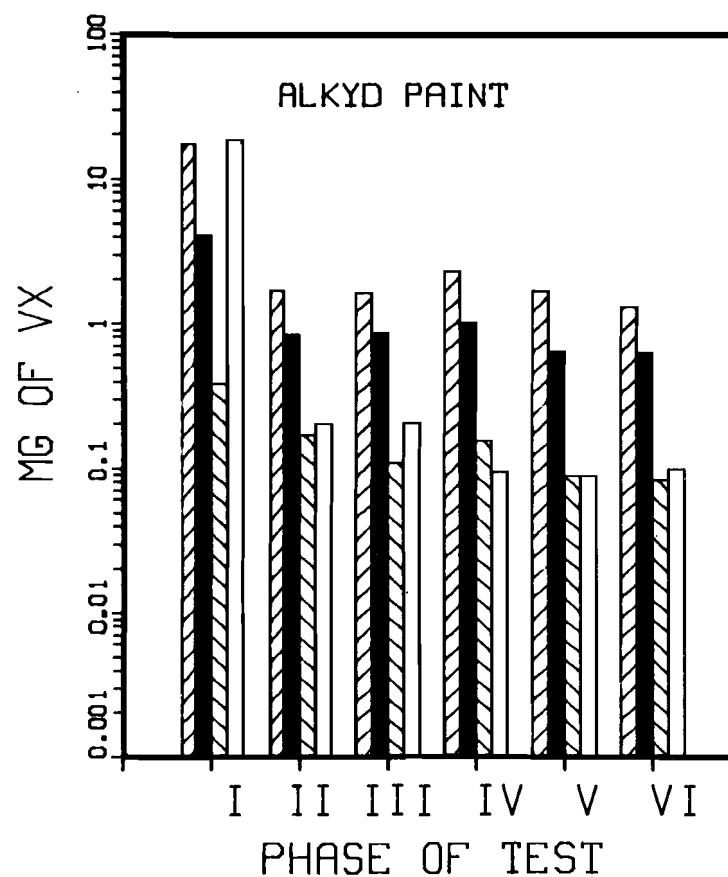
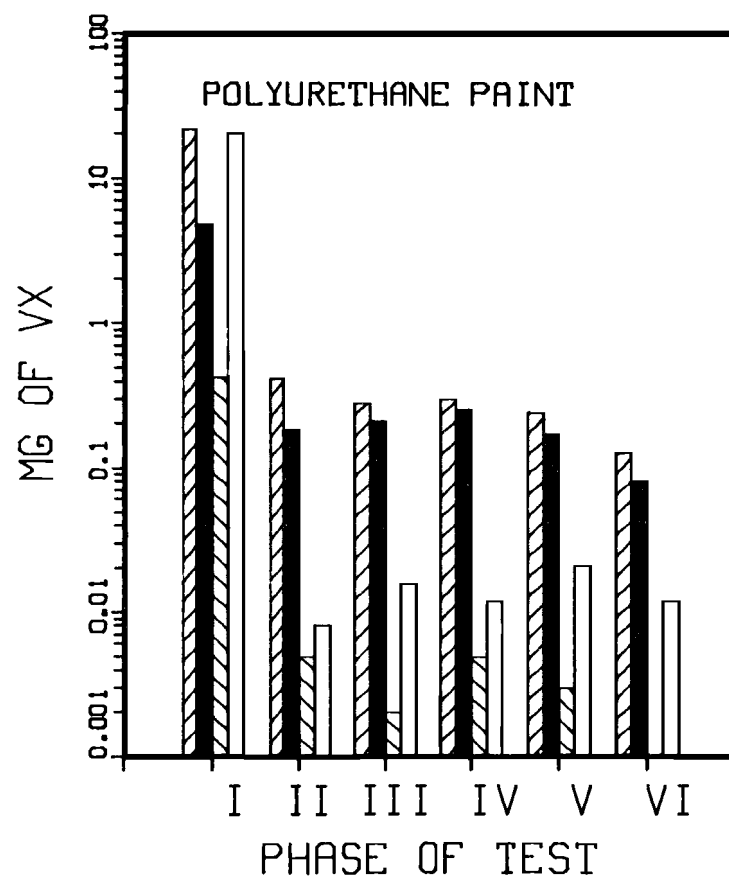


Figure 1. Mean Residual VX and TVX Remaining on Control Plates After Test Phases I Through VI

ALL PHASES ARE VX-30 MINUTE AGE

I: CONTROL, NO RINSE
 II: RINSE, NO AGE
 III: RINSE, 15 MIN AGE
 IV: RINSE, 15 MIN AGE
 V: RINSE, 5 HR AGE
 VI: RINSE, 5 HR AGE

LEGEND
 ▨ 25.0 MG VX STARTING DOSE
 ■ 5.0 MG VX STARTING DOSE
 ▩ 0.5 MG VX STARTING DOSE
 □ 25.0 MG TVX STARTING DOSE

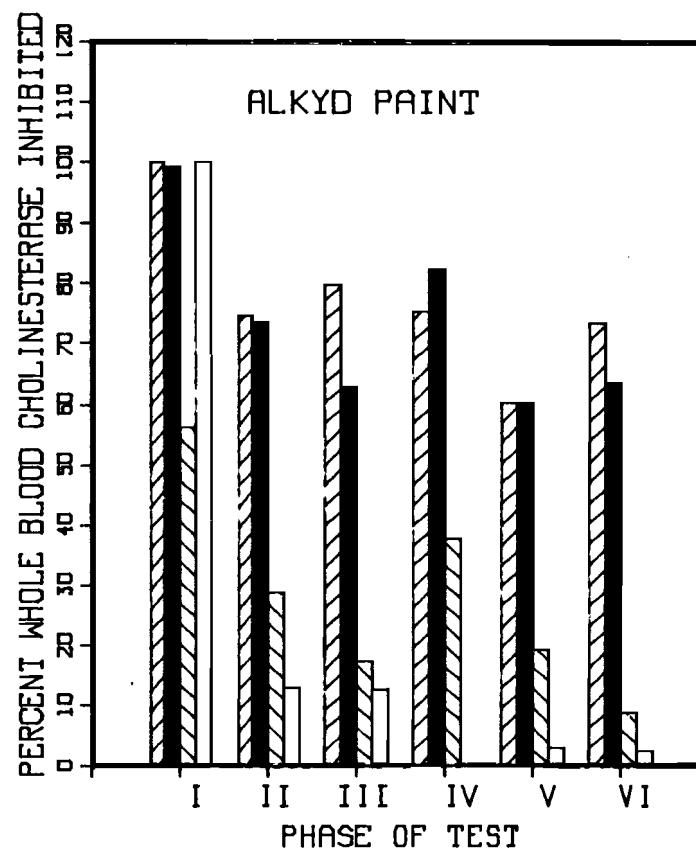
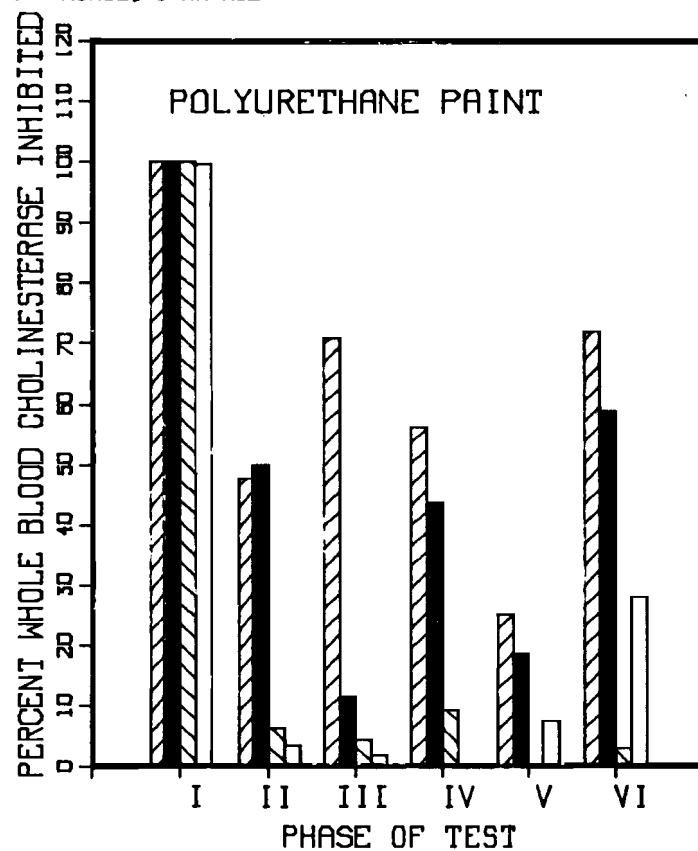


Figure 2. Mean Whole Blood Cholinesterase Inhibition Following VX Exposure, Direct Contact Plates

ALL PHASES ARE VX-30 MINUTE AGE

I: CONTROL, NO RINSE
 II: RINSE, NO AGE
 III: RINSE, 15 MIN AGE
 IV: RINSE, 15 MIN AGE
 V: RINSE, 5 HR AGE
 VI: RINSE, 5 HR AGE

LEGEND
 ▨ 25.0 MG VX STARTING DOSE
 ■ 5.0 MG VX STARTING DOSE
 ▩ 0.5 MG VX STARTING DOSE
 □ 25.0 MG TVX STARTING DOSE

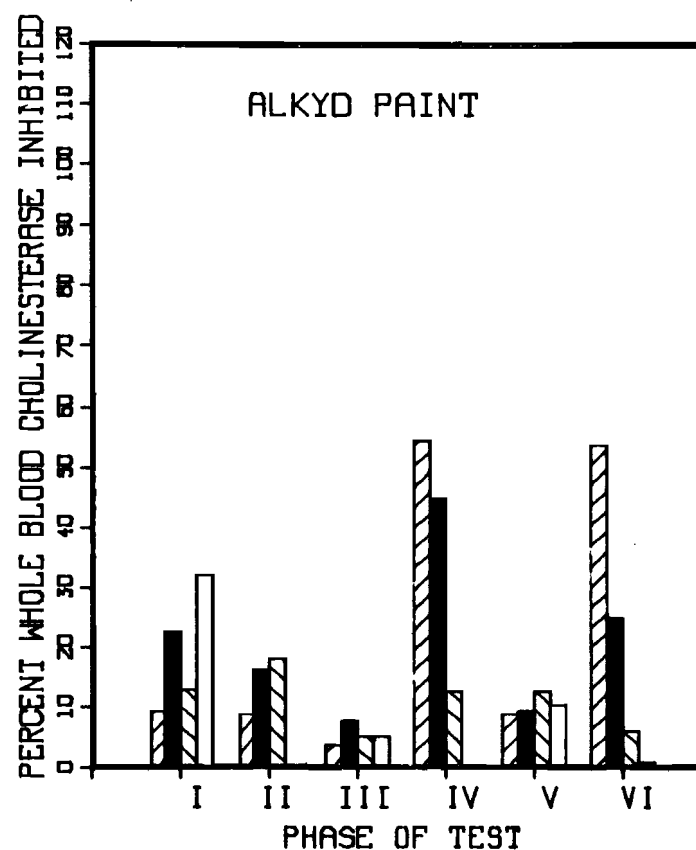
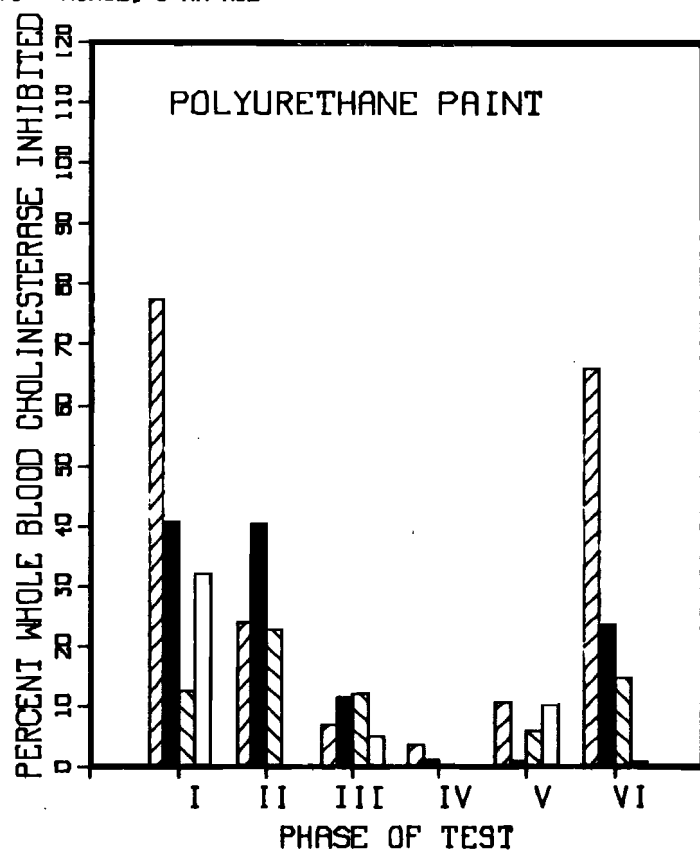


Figure 3. Mean Whole Blood Cholinesterase Inhibition Following VX Exposure, Vapor Contact Plates

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APPENDIX A
BIOLOGICAL RESPONSE DATA

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Table A-1. Biological Responses (Rabbit) to Agent Contact (VX), 25.0 mg/Plate
(Polyurethane Paint)

		Onset time of toxic signs (minutes - mean and range)								Whole blood ChE activity percent inhibition	
Test Phase	Contact type	Twitches	Tremors	Convulsions	Miosis	Salivation	Prostration	Death	Mean	Range	
1. 30-min aging, no rinse, no aging, 60-min contact	Direct	N.O. ^a / (-) ^b	(-)	6.8 (6/6) (5.0-8.0)	7.4 (5/6) (5.0-8.5)	7.4 (5/6) (5.0-9.5)	8.2 (6/6) (6.0-10.0)	10.2 (6/6) (9.0-11.0)	100.0 (6)	<0.1 (6)	
	Vapor	N.O.	(-)	<67.0 (6/6) (<63.0-75.0)	(0/6)	140.0 (4/6) (76.0-165.0)	10.5 (5/6) (3.5-31.0)	(0/6)	(0/6)	77.6 (6) 71.1-81.8 (6)	
2. 30-min aging, no rinse, no aging, 60-min contact	Direct	N.O.	(-)	79.7 (6/6) (55.5-131.5)	(0/6)	287.0 (4/6) (221.5-310.5)	(0/6)	(0/6)	(0/6)	47.8 (6) 5.6-81.6 (6)	
	Vapor	186.5 (2/6) (183.5-189.5)	90.0 (2/6) (84.5-95.5)	1395.0 (1/6)	1395.0 (1/6)	1395.0 (1/6)	1395.0 (1/6)	(0/6)	24.1 (6)	0-53.9 (6)	
3. 30-min aging, rinsed, aged 15 min, 60-min contact	Direct	76.8 (3/6) (67.0-82.5)	113.8 (6/6) (61.0-265.0)	(0/6)	68.0 (1/6)	82.0 (1/6)	83.0 (1/6)	134.0 (1/6)	71.0 (6)	55.9-99.2 (6)	
	Vapor	106.0 (1/6)	245.0 (3/6) (107.5-322.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	7.0 (6)	0.0-26.9 (6)	
4. 30-min aging, rinsed, aged 15 min, 15-min contact	Direct	85.2 (6/6) (23.8-297.8)	83.8 (5/6) (31.8-249.5)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	56.3 (6)	13.0-79.8 (6)	
	Vapor	214.2 (1/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	3.8 (6)	0.0-13.6 (6)	
5. 30-min aging, rinsed, aged 15 min, 60-min contact	Direct	68.9 (4/6) (62.8-73.5)	77.3 (3/6) (72.8-84.8)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	25.2 (6)	0.0-60.1 (6)	
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	10.8 (6)	0.0-45.4 (6)	
6. 30-min aging, rinsed, aged 5 hours, 15-min contact	Direct	63.2 (3/6) (54.5-72.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	71.9 (6)	62.8-84.4 (6)	
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	66.2 (6)	43.8-88.3 (6)	

^aNot all of these toxic signs were observed due to occluding patch

^bNumber of animals responding over number in test group

N.O. Not observed

Table A-2. Biological Responses (Rabbit) to Agent Contact (VX), 5.0 mg/Plate
(Polyurethane Paint)

Test Phase	Contact type	Onset time of toxic signs (minutes - mean and range)							Whole blood ChE activity percent inhibition	
		Twitches	Tremors	Convulsions	Miosis	Salivation	Prostration	Death	Mean	Range
1. 30-min aging, no rinse, no aging, 60-min contact	Direct	(-) ^{b/} N.O. ^{a/}	21.5 ^{a/} (2/6) (16.0-27.0)	19.2 (6/6) (13.5-25.5)	17.2 (6/6) (11.5-24.5)	17.5 (5/6) (6.5-26.5)	21.0 (6/6) (14.5-29.5)	22.8 (6/6) (16.5-31.5)	100.0 (6)	<0.1 (6)
	Vapor	(-) N.O.	80.2 (6/6) (69.0-101.0)	(0/6)	74.0 (2/6) (73.0-75.0)	(0/6)	(0/6)	(0/6)	40.5 (6)	14.9-59.7 (6)
2. 30-min aging, rinsed, no aging, 60-min contact	Direct	187.0 (2/6) (186.5-187.5)	110.3 (6/6) (64.5-212.5)	(0/6)	302.5 (4/6) (301.0-304.0)	(0/6)	(0/6)	(0/6)	49.8 (6)	10.8-75.7 (6)
	Vapor	170.2 (1/6) (67.0-297.0)	130.4 (4/6)	(0/6)	223.2 (3/6) (68.0-301.0)	(0/6)	(0/6)	(0/6)	40.1 (6)	0.0-66.2 (6)
3. 30-min aging, rinsed, aged 15 min, 60-min contact	Direct	72.4 (4/6) (65.0-83.5)	138.6 (6/6) (66.0-305.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	11.3 (6)	0.0-38.3 (6)
	Vapor	189.5 (1/6)	188.0 (1/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	11.4 (6)	0.0-24.6 (6)
4. 30-min aging, rinsed, aged 15 min, 15-min contact	Direct	30.4 (4/6) (27.2-34.8)	65.9 (4/6) (30.5-101.5)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	43.3 (6)	7.0-68.5 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	1.1 (6)	0.0-5.2 (6)
5. 30-min aging, rinsed, aged 5 hours, 60-min contact	Direct	84.4 (2/6) (69.5-99.2)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	18.6 (6)	0.0-45.6 (6)
	Vapor	(0/6)	71.8 (1/6)	72.8 (1/6)	(0/6)	(0/6)	(0/6)	(0/6)	0.9 (6)	0.0-5.6 (6)
6. 30-min aging, rinsed, aged 5 hours, 15-min contact	Direct	54.0 (5/6) (50.0-58.0)	58.0 (1/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	58.6 (6)	42.8-75.3 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	23.6 (6)	0.0-76.4 (6)

^aNot all of these toxic signs were observed due to occluding patch

^bNumber of animals responding over number in test group

N.O. Not observed

Table A-3. Biological Responses (Rabbit) to Agent Contact (VX), 0.5 mg/Plate
(Polyurethane Paint)

Test Phase	Contact type	Onset time of toxic signs (minutes - mean and range)							Whole blood ChE activity percent inhibition	
		Twitches	Tremors	Convulsions	Miosis	Salivation	Prostration	Death	Mean	Range
1. 30-min aging, no rinse, no aging, 60-min contact	Direct	a/ (-) b/ N.O.	25.2 (6/6) (7.5-68.0)	70.1 (6/6) (29.0-102.0)	53.2 (6/6) (26.0-88.0)	59.4 (6/6) (19.0-107.0)	85.9 (6/6) (36.0-133.0)	104.8 (6/6) (50.0-146.0)	100.0 (6)	<0.1 (6)
	Vapor	N.O. (-)	144.9 (6/6) (68.0-319.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	12.4 (6)	0.0-28.6 (6)
2. 30-min aging, rinsed, no aging, 60-min contact	Direct	127.5 (2/6) (69.0-186.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	6.1 (6)	0.0-34.2 (6)
	Vapor	(0/6)	76.5 (1/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	23.0 (6)	0.0-90.0 (6)
3. 30-min aging, rinsed, aged 15 min, 60-min contact	Direct	(0/6)	219.0 (1/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	4.6 (6)	0.0-22.7 (6)
	Vapor	(0/6)	114.2 (2/6) (82.5-146.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	12.0 (6)	0.0-35.2 (6)
4. 30-min aging, rinsed, aged 15 min, 15-min contact	Direct	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	9.2 (6)	0.0-48.4 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	0.2 (6)	0.0-0.6 (6)
5. 30-min aging, rinsed, aged 5 hours, 60-min contact	Direct	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	0.0 (6)	0.0 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	6.0 (6)	0.0-35.9 (6)
6. 30-min aging, rinsed, aged 5 hours, 15-min contact	Direct	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	3.0 (6)	0.0-14.9 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	14.6 (6)	0.0-53.8 (6)

^aNot all of these toxic signs were observed due to occluding patch

^bNumber of animals responding over number in test group

N.O. Not observed

Table A-4. Biological Responses (Rabbit) to Agent Contact (VX), 25.0 mg/Plate (Alkyd Paint)

Test Phase	Contact type	Onset time of toxic signs (minutes - mean and range)							Whole blood ChE activity percent inhibition	
		Twitches	Tremors	Convulsions	Miosis	Salivation	Prostration	Death	Mean	Range
1. 30-min aging, no rinse, no aging, 60-min contact	Direct	N.O. ^a (-) ^b	7.1 (6/6) (6.0-9.2)	8.3 (6/6) (6.5-11.2)	7.3 (6/6) (5.5-10.2)	8.5 (6/6) (7.8-9.5)	9.9 (6/6) (8.8-12.5)	11.3 (6/6) (9.2-15.5)	99.9 (6)	99.7-100.0 (6)
	Vapor	102.8 (6/6) (60.5-207.0)	80.5 (3/6) (12.0-134.0)	137.5 (1/6)	(0/6)	30.0 (1/6)	(0/6)	(0/6)	9.4 (6)	0.0-39.4 (6)
2. 30-min aging, rinsed, no aging, 60-min contact	Direct	52.2 (6/6) (28.5-80.0)	62.9 (6/6) (28.5-93.5)	(0/6)	(0/6)	166.0 (3/6) (140.0-208.0)	(0/6)	(0/6)	74.7 (6)	53.2-91.2 (6)
	Vapor	91.1 (4/6) (64.0-120.5)	105.0 (2/6) (93.5-116.5)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	9.0 (6)	0.0-20.2 (6)
3. 30-min aging, rinsed, aged 15 min, 60-min contact	Direct	37.1 (6/6) (14.0-70.5)	51.5 (6/6) (39.0-77.5)	141.5 (1/6)	143.5 (1/6)	125.8 (3/6) (47.0-281.5)	306.5 (1/6)	397.5 (1/6)	79.7 (6)	71.5-98.6 (6)
	Vapor	103.3 (5/6) (40.0-237.0)	72.2 (2/6) (48.5-96.0)	(0/6)	(0/6)	88.5 (2/6) (87.5-89.5)	(0/6)	(0/6)	3.7 (6)	0.0-15.8 (6)
4. 30-min aging, rinsed, aged 15 min, 15-min contact	Direct	36.2 (6/6) (20.0-102.5)	73.1 (6/6) (27.0-124.5)	(0/6)	(0/6)	28.5 (1/6)	(0/6)	(0/6)	75.2 (6)	56.1-91.0 (6)
	Vapor	106.3 (3/6) (31.5-177.5)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	54.2 (6)	28.1-75.9 (6)
5. 30-min aging, rinsed, aged 5 hours, 60-min contact	Direct	53.8 (6/6) (34.0-65.0)	89.8 (6/6) (81.5-96.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	60.2 (6)	36.8-72.6 (6)
	Vapor	73.5 (1/6)	57.5 (1/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	8.8 (6)	0.0-24.1 (6)
6. 30-min aging, rinsed, aged 5 hours, 15-min contact	Direct	31.8 (6/6) (17.0-38.5)	67.3 (6/6) (17.5-101.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	73.5 (6)	34.8-84.3 (6)
	Vapor	98.0 (1/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	53.4 (6)	1.2-72.6 (6)

^aNot all of these toxic signs were observed due to occluding patch^bNumber of animals responding over number in test group

N.O. Not observed

Table A-5. Biological Responses (Rabbit) to Agent Contact (VX), 5.0 mg/Plate (Alkyd Paint)

Test Phase	Contact type	Onset time of toxic signs (minutes - mean and range)							Whole blood ChE activity percent inhibition	
		Twitches	Tremors	Convulsions	Miosis	Salivation	Prostration	Death	Mean	Range
1. 30-min aging, no rinse, no aging, 60-min contact	Direct	10.8 (1/6) ^{a/}	27.9 (6/6) (12.0-75.5)	59.8 (6/6) (42.0-81.0)	57.6 (6/6) (18.0-78.0)	48.4 (5/6) (13.2-84.5)	78.9 (6/6) (45.5-143.0)	93.7 (6/6) (54.5-160.0)	99.5 (6)	98.1-100.0 (6)
	Vapor	196.0 (3/6) (192.0-203.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	22.6 (6)	0.0-52.0 (6)
2. 30-min aging, rinsed, no aging, 60-min contact	Direct	61.6 (6/6) (52.0-64.5)	100.3 (6/6) (95.0-110.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	73.6 (6)	63.9-84.6 (6)
	Vapor	162.4 (6/6) (102.0-319.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	16.3 (6)	0.0-60.9 (6)
3. 30-min aging, rinsed, aged 15 min, 60-min contact	Direct	67.1 (6/6) (59.5-71.5)	80.5 (6/6) (72.0-105.5)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	62.8 (6)	57.3-66.2 (6)
	Vapor	162.5 (5/6) (89.0-267.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	7.9 (6)	0.0-24.5 (6)
4. 30-min aging, rinsed, aged 15 min, 15-min contact	Direct	51.4 (4/6) (30.5-100.0)	90.9 (5/6) (29.5-140.5)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	82.2 (6)	70.0-89.0 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	44.7 (6)	0.0-81.8 (6)
5. 30-min aging, rinsed, aged 5 hours, 60-min contact	Direct	54.4 (6/6) (34.5-75.0)	85.2 (6/6) (70.5-100.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	60.3 (6)	50.1-85.8 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	9.4 (6)	0.0-56.6 (6)
6. 30-min aging, rinsed, aged 5 hours, 15-min contact	Direct	34.6 (6/6) (25.0-49.5)	96.6 (4/6) (89.5-108.5)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	63.4 (6)	45.2-81.1 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	24.9 (6)	4.6-50.1 (6)

^aNumber of animals responding over number in test group

Table A-6. Biological Responses (Rabbit) to Agent Contact (VX), 0.5 mg/Plate (Alkyd Paint)

Test Phase	Contact type	Onset time of toxic signs (minutes - mean and range)							Whole blood ChE activity percent inhibition	
		Twitches	Tremors	Convulsions	Miosis	Salivation	Prostration	Death	Mean	Range
1. 30-min aging, no rinse, no aging, 60-min contact	Direct	72.7 (6/6) ^{a/} (61.0-94.0)	78.0 (2/6) (71.0-85.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	56.0 (6)	26.0-79.3 (6)
	Vapor	92.5 (2/6) (92.0-93.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	12.8 (6)	0.0-21.8 (6)
2. 30-min aging, rinsed, no age, 60-min contact	Direct	93.2 (6/6) (65.0-124.0)	111.0 (2/6) (93.0-129.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	28.7 (6)	7.7-55.7 (6)
	Vapor	186.5 (2/6) (137.0-236.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	18.2 (6)	2.3-28.4 (6)
3. 30-min aging, rinsed, aged 15 min, 60-min contact	Direct	78.6 (6/6) (75.5-81.5)	128.5 (1/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	17.3 (6)	0.0-37.4 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	5.1 (6)	0.0-30.7 (6)
4. 30-min aging, rinsed, aged 15 min, 15-min contact	Direct	119.5 (2/6) (38.0-201.0)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	37.7 (6)	0.0-59.6 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	12.6 (6)	0.0-30.9 (6)
5. 30-min aging, rinsed, aged 5 hours, 60-min contact	Direct	73.2 (6/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	19.1 (6)	0.0-57.2 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	12.6 (6)	0.0-41.0 (6)
6. 30-min aging, rinsed, aged 5 hours, 15-min contact	Direct	38.3 (5/6) (27.5-53.5)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	8.8 (6)	0.0-23.7 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	5.8 (6)	0.0-21.6 (6)

^a Number of animals responding over number in test group

Table A-7. Biological Responses (Rabbit) to Agent Contact (TVX), 25.0 mg/Plate (Polyurethane Paint)

Test Phase	Contact type	Onset time of toxic signs (minutes - mean and range)							Whole blood ChE activity percent inhibition	
		Twitches ^{a/}	Tremors	Convulsions	Miosis	Salivation	Prostration	Death	Mean	Range
1. 30-min aging, no rinse, no aging, 60-min contact	Direct	13.5 (2/6) ^{b/} (9.5-17.5)	12.8 (6/6) (8.5-21.0)	13.1 (6/6) (8.5-22.0)	13.4 (4/6) (9.0-24.0)	13.7 (6/6) (10.2-24.5)	14.1 (6/6) (9.5-22.5)	16.0 (6/6) (12.2-26.0)	99.6 (6)	97.8-100.0 (6)
	Vapor	80.9 (2/6) (77.0-84.8)	64.6 (5/6) (27.0-116.5)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	33.2 (6)	0.0-78.0 (6)
2. 30-min aging, rinsed, no aging, 60-min contact	Direct	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	3.4 (6)	0.0-17.2 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	12.0 (6)	0.0-34.4 (6)
3. 30-min aging, rinsed, aged 15 min, 60-min contact	Direct	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	2.0 (6)	0.0-10.0 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	7.3 (6)	0.0-23.0 (6)
4. 30-min aging, rinsed, aged 15 min, 15-min contact	Direct	216.5 (1/6)	270.5 (1/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	0.0 (6)	0.0 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	0.0 (6)	0.0 (6)
5. 30-min aging, rinsed, aged 5 hours, 60-min contact	Direct	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	7.4 (6)	0.0-20.1 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	6.9 (6)	0.0-19.0 (6)
6. 30-min aging, rinsed, aged 5 hours, 15-min contact	Direct	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	28.0 (6)	14.3-40.5 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	11.0 (6)	0.0-27.8 (6)

^aNot all of these toxic signs were observed due to occluding patch

^bNumber of animals responding over number in test group

Table A-8. Biological Responses (Rabbit) to Agent Contact (TVX), 25.0 mg/Plate (Alkyd Paint)

Test Phase	Contact type	Onset time of toxic signs (minutes - mean and range)							Whole blood ChE activity percent inhibition	
		Twitches ^{a/}	Tremors	Convulsions	Miosis	Salivation	Prostration	Death	Mean	Range
1. 30-min aging, no rinse, no aging, 60-min contact	Direct	9.8 (1/6) ^{b/}	9.3 (5/6) (4.5-12.0)	12.0 (6/6) (6.5-16.5)	12.0 (6/6) (6.5-17.5)	10.0 (6/6) (5.5-17.8)	13.3 (6/6) (7.5-18.5)	14.8 (6/6) (9.5-21.0)	100.0 (6)	100.0-100.0 (6)
	Vapor	88.4 (5/6) (82.0-97.2)	153.6 (4/6) (21.5-199.2)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	32.0 (6)	0.0-67.5 (6)
2. 30-min aging, rinsed, no aging, 60-min contact	Direct	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	12.8 (6)	0.2-35.0 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	0.0 (6)	0.0 (6)
3. 30-min aging, rinsed, aged 15 min, 60-min contact	Direct	113.2 (1/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	12.6 (6)	0.0-26.4 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	5.2 (6)	0.0-31.4 (6)
4. 30-min aging, rinsed, aged 15 min, 15-min contact	Direct	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	0.0 (6)	0.0 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	0.0 (6)	0.0 (6)
5. 30-min aging, rinsed, aged 5 hours, 60-min contact	Direct	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	2.8 (6)	0.0-14.8 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	10.5 (6)	0.0-35.1 (6)
6. 30-min aging, rinsed, aged 5 hours, 15-min contact	Direct	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	2.6 (6)	0.0-15.8 (6)
	Vapor	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	(0/6)	0.8 (6)	0.0-4.8 (6)

^aNot all of these toxic signs were observed due to occluding patch^bNumber of animals responding over number in test group

APPENDIX B
PHYSICAL/CHEMICAL DATA

Blank

APPENDIX B

PHYSICAL/CHEMICAL DATA

Table B-1. Physical/Chemical Data Following Agent (VX) Contact, 25.0 mg/Plate (Polyurethane Paint)

Test Phase	Contact type	Condition of agent on test plate prior to rinse removal			Chemical analysis for residual agent on plate			
		Area % spread on plate		Degree of wetness edge to center	Micrograms of agent left on plate		Percent residual agent on plate	
		Mean	± S.D.		Mean	± S.D.	Mean	± S.D.
1. 30-min aging, no rinse, no aging, 60-min contact	Control	75.0	6.0	Wet - Wet	21,485.0	1,669.0	85.9	6.7
	Direct	84.0	8.0	Wet - Wet	2,232.0	646.0	8.9	2.6
	Vapor	78.0	6.0	Wet - Wet	9,414.0	1,383.0	37.6	5.5
2. 30-min aging, rinsed, no aging, 60-min contact	Control	88.0	5.0	Wet - Wet	413.0	156.0	1.6	0.6
	Direct	83.0	7.0	Wet - Wet	165.0	87.0	0.7	0.4
	Vapor	85.0	3.0	Wet - Wet	247.0	107.0	1.0	0.4
3. 30-min aging, rinsed, aged 15 min, 60-min contact	Control	85.0	6.0	Wet - Wet	275.0	185.0	1.1	0.7
	Direct	85.0	10.0	Wet - Wet	310.0	149.0	1.2	0.6
	Vapor	86.0	8.0	Wet - Wet	297.0	120.0	1.2	0.5
4. 30-min aging, rinsed, aged 15 min, 15-min contact	Control	88.0	5.0	Wet - Wet	301.0	76.0	1.2	0.3
	Direct	90.0	4.0	Wet - Wet	415.0	201.0	1.7	0.8
	Vapor	91.0	5.0	Wet - Wet	385.0	90.0	1.5	0.4
5. 30-min aging, rinsed, aged 5 hours, 60-min contact	Control	89.0	5.0	Wet - Wet	236.0	95.0	0.9	0.4
	Direct	89.0	2.0	Wet - Wet	122.0	41.0	0.4	0.2
	Vapor	91.0	6.0	Wet - Wet	69.0	68.0	0.3	0.3
6. 30-min aging, rinsed, aged 5 hours, 15-min contact	Control	89.0	4.0	Wet - Wet	132.0	51.0	0.5	0.2
	Direct	88.0	5.0	Wet - Wet	129.0	43.0	0.5	0.2
	Vapor	82.0	5.0	Wet - Wet	140.0	24.0	0.6	0.1

Table B-2. Physical/Chemical Data Following Agent (VX) Contact,
5.0 mg/Plate (Polyurethane Paint)

Test Phase	Contact type	Condition of agent on test plate prior to rinse removal			Chemical analysis for residual agent on plate			
		Area % spread on plate		Degree of wetness edge to center	Micrograms of agent left on plate		Percent residual agent on plate	
		Mean	± S.D.		Mean	± S.D.	Mean	± S.D.
1. 30-min aging, no rinse, no aging, 60-min contact	Control	54.0	10.0	Damp - Wet	4,652.0	436.0	93.0	8.7
	Direct	55.0	5.0	Damp - Wet	985.0	406.0	19.7	8.1
	Vapor	42.0	4.0	Damp - Wet	3,981.0	549.0	79.6	11.0
2. 30-min aging, rinsed, no aging, 60-min contact	Control	62.0	4.0	Damp - Wet	181.0	66.0	3.6	1.3
	Direct	65.0	9.0	Damp - Wet	124.0	67.0	2.5	1.3
	Vapor	62.0	7.0	Damp - Wet	148.0	53.0	3.0	1.1
3. 30-min aging, rinsed, aged 15 min, 60-min contact	Control	62.0	8.0	Damp - Wet	213.0	59.0	4.3	1.2
	Direct	52.0	5.0	Damp - Wet	70.0	36.0	1.4	0.7
	Vapor	57.0	9.0	Damp - Wet	164.0	63.0	3.3	1.3
4. 30-min aging, rinsed, aged 15 min, 15-min contact	Control	62.0	3.0	Damp - Wet	254.0	86.0	5.1	1.7
	Direct	58.0	3.0	Damp - Wet	244.0	110.0	4.9	2.2
	Vapor	53.0	8.0	Damp - Wet	201.0	99.0	4.0	2.0
5. 30-min aging, rinsed, aged 5 hours, 60-min contact	Control	62.0	3.0	Damp - Wet	168.0	64.0	3.4	1.3
	Direct	60.0	4.0	Damp - Wet	90.0	36.0	1.8	0.7
	Vapor	58.0	3.0	Damp - Wet	104.0	75.0	1.8	1.7
6. 30-min aging, rinsed, aged 5 hours, 15-min contact	Control	59.0	4.0	Damp - Wet	82.0	19.0	1.6	0.4
	Direct	61.0	6.0	Damp - Wet	110.0	18.0	2.2	0.4
	Vapor	56.0	6.0	Damp - Wet	70.0	27.0	1.4	0.5

Table B-3. Physical/Chemical Data Following Agent (VX) Contact,
0.5 mg/Plate (Polyurethane Paint)

Test Phase	Contact type	Condition of agent on test plate prior to rinse removal			Chemical analysis for residual agent on plate			
		Area % spread on plate		Degree of wetness edge to center	Micrograms of agent left on plate		Percent residual agent on plate	
		Mean	+ S.D.		Mean	+ S.D.	Mean	+ S.D.
1. 30-min aging, no rinse, no aging, 60-min contact	Control	29.0	2.0	Dry - Damp	415.8	52.1	83.1	10.5
	Direct	23.0	2.0	Damp - Damp	89.2	59.0	17.8	11.8
	Vapor	18.0	4.0	Damp - Damp	383.2	161.6	76.6	32.3
2. 30-min aging, rinsed, no aging, 60-min contact	Control	28.0	3.0	Damp - Damp	5.2	0.4	1.0	0.08
	Direct	27.0	3.0	Damp - Damp	1.8	1.6	0.4	0.32
	Vapor	28.0	2.0	Damp - Damp	5.5	7.3	1.1	1.45
3. 30-min aging, rinsed, aged 15 min, 60-min contact	Control	30.0	3.0	Damp - Damp	1.8	0.4	0.4	0.1
	Direct	18.0	5.0	Damp - Damp	1.8	0.2	0.4	<0.1
	Vapor	21.0	10.0	Damp - Damp	12.3	20.6	2.5	4.1
4. 30-min aging, rinsed, aged 15 min, 15-min contact	Control	27.0	5.0	Damp - Damp	4.8	6.4	1.0	1.3
	Direct	27.0	4.0	Damp - Damp	1.2	1.1	0.2	0.2
	Vapor	28.0	3.0	Damp - Damp	4.5	2.8	0.9	0.6
5. 30-min aging, rinsed, aged 5 hours, 60-min contact	Control	24.0	7.0	Damp - Damp	3.1	2.7	0.6	0.5
	Direct	26.0	6.0	Damp - Damp	1.8	1.5	0.4	0.3
	Vapor	25.0	4.0	Damp - Damp	3.1	2.9	0.6	0.6
6. 30-min aging, rinsed, aged 5 hours, 15-min contact	Control	21.0	8.0	Damp - Damp	1.0	0.4	0.2	0.1
	Direct	23.0	4.0	Damp - Damp	6.1	3.9	1.2	0.8
	Vapor	25.0	8.0	Damp - Damp	2.6	1.0	0.5	0.2

Table B-4. Physical/Chemical Data Following Agent (VX) Contact,
25.0 mg/Plate (Alkyd Paint)

Test Phase	Contact type	Condition of agent on test plate prior to rinse removal			Chemical analysis for residual agent on plate			
		Area % spread on plate		Degree of wetness edge to center	Micrograms of agent left on plate		Percent residual agent on plate	
		Mean	± S.D.		Mean	± S.D.	Mean	± S.D.
1. 30-min aging, no rinse, no aging, 60-min contact	Control	90.0	4.0	Damp - Wet	17,439.0	3,114.0	69.8	12.5
	Direct	90.0	3.0	Damp - Wet	10,313.0	1,311.0	41.2	5.3
	Vapor	86.0	8.0	Damp - Wet	14,200.0	3,538.0	57.3	14.0
2. 30-min aging, rinsed, no aging, 60-min contact	Control	91.0	7.0	Damp - Wet	1,684.0	30.0	6.7	0.2
	Direct	90.0	8.0	Damp - Wet	1,346.0	199.0	5.4	0.8
	Vapor	92.0	4.0	Damp - Wet	1,710.0	340.0	6.8	1.4
3. 30-min aging, rinsed, aged 15 min, 60-min contact	Control	94.0	2.0	Damp - Wet	1,662.0	24.0	6.6	0.1
	Direct	91.0	7.0	Damp - Wet	1,572.0	133.0	6.3	0.5
	Vapor	88.0	4.0	Damp - Wet	1,638.0	71.0	6.6	0.3
4. 30-min aging, rinsed, aged 15 min, 15-min contact	Control	89.0	4.0	Damp - Wet	2,376.0	111.0	9.5	0.4
	Direct	85.0	4.0	Damp - Wet	1,769.0	151.0	7.1	0.6
	Vapor	88.0	3.0	Damp - Wet	2,015.0	63.0	8.1	0.3
5. 30-min aging, rinsed, aged 5 hours, 60-min contact	Control	92.0	4.0	Damp - Wet	1,679.0	81.0	6.7	0.3
	Direct	90.0	4.0	Damp - Wet	1,585.0	147.0	6.3	0.6
	Vapor	93.0	5.0	Damp - Wet	1,578.0	236.0	6.3	1.0
6. 30-min aging, rinsed, aged 5 hours, 15-min contact	Control	98.0	2.0	Damp - Wet	1,314.0	39.0	5.3	0.2
	Direct	98.0	4.0	Damp - Wet	1,330.0	43.0	5.3	0.2
	Vapor	98.0	4.0	Damp - Wet	1,382.0	139.0	5.5	0.6

Table B-5. Physical/Chemical Data Following Agent (VX) Contact,
5.0 mg/Plate (Alkyd Paint)

Test Phase	Contact type	Condition of agent on test plate prior to rinse removal			Chemical analysis for residual agent on plate			
		Area % spread on plate		Degree of wetness edge to center	Micrograms of agent left on plate		Percent residual agent on plate	
		Mean	± S.D.		Mean	± S.D.	Mean	± S.D.
1. 30-min aging, no rinse, no aging, 60-min contact	Control	50.0	4.0	Damp - Damp	4,155.0	175.0	83.1	3.5
	Direct	52.0	5.0	Damp - Damp	4,594.0	680.0	91.9	13.6
	Vapor	48.0	5.0	Damp - Damp	5,012.0	608.0	100.2	12.2
2. 30-min aging, rinsed, no aging, 60-min contact	Control	50.0	3.0	Dry - Damp	850.0	28.0	17.0	0.6
	Direct	48.0	4.0	Dry - Damp	877.0	29.0	17.6	0.6
	Vapor	48.0	5.0	Dry - Damp	870.0	24.0	17.4	0.5
3. 30-min aging, rinsed, aged 15 min, 60-min contact	Control	49.0	2.0	Dry - Damp	860.0	58.0	17.2	1.2
	Direct	52.0	4.0	Dry - Damp	737.0	72.0	14.7	1.4
	Vapor	51.0	2.0	Dry - Damp	819.0	23.0	16.4	0.5
4. 30-min aging, rinsed, aged 15 min, 15-min contact	Control	49.0	2.0	Dry - Damp	1,020.0	125.0	20.4	2.5
	Direct	48.0	3.0	Dry - Damp	845.0	65.0	16.9	1.3
	Vapor	49.0	2.0	Dry - Damp	946.0	96.0	18.9	1.9
5. 30-min aging, rinsed, aged 5 hours, 60-min contact	Control	52.0	3.0	Dry - Damp	658.0	50.0	13.2	1.0
	Direct	52.0	6.0	Dry - Damp	705.0	29.0	14.1	0.6
	Vapor	52.0	3.0	Dry - Damp	568.0	220.0	11.4	4.4
6. 30-min aging, rinsed, aged 5 hours, 15-min contact	Control	58.0	3.0	Dry - Damp	621.0	52.0	12.4	1.0
	Direct	58.0	3.0	Dry - Damp	571.0	50.0	11.4	1.0
	Vapor	55.0	3.0	Dry - Damp	573.0	62.0	11.5	1.2

Table B-6. Physical/Chemical Data Following Agent (VX) Contact,
0.5 mg/Plate (Alkyd Paint)

Test Phase	Contact type	Condition of agent on test plate prior to rinse removal			Chemical analysis for residual agent on plate			
		Area % spread on plate		Degree of wetness edge to center	Micrograms of agent left on plate		Percent residual agent on plate	
		Mean	± S.D.		Mean	± S.D.	Mean	± S.D.
1. 30-min aging, no rinse, no aging, 60-min contact	Control	11.0	1.0	Dry - Dry	384.0	78.0	76.7	15.6
	Direct	10.0	1.0	Dry - Dry	292.0	82.0	58.4	16.5
	Vapor	11.0	1.0	Dry - Dry	318.0	113.0	63.6	22.5
2. 30-min aging, rinsed, no aging, 60-min contact	Control	10.0	2.0	Damp - Dry	170.8	6.6	34.2	1.3
	Direct	11.0	2.0	Dry - Dry	162.2	11.0	32.4	2.2
	Vapor	10.0	4.0	Dry - Dry	182.4	30.5	36.5	6.1
3. 30-min aging, rinsed, aged 15 min, 60-min contact	Control	11.0	1.0	Dry - Dry	112.0	36.0	22.3	7.2
	Direct	12.0	1.0	Dry - Dry	125.0	41.0	25.1	8.2
	Vapor	11.0	1.0	Dry - Dry	132.0	23.0	26.4	4.5
4. 30-min aging, rinsed, aged 15 min, 15-min contact	Control	10.0	1.0	Dry - Dry	156.0	18.0	31.3	3.7
	Direct	7.0	2.0	Dry - Dry	91.0	27.0	18.2	5.4
	Vapor	8.0	3.0	Dry - Dry	127.0	38.0	25.4	7.7
5. 30-min aging, rinsed, aged 5 hours, 60-min contact	Control	11.0	1.0	Dry - Dry	94.0	24.0	18.7	4.9
	Direct	12.0	2.0	Dry - Dry	76.0	17.0	15.3	3.4
	Vapor	12.0	2.0	Dry - Dry	95.0	26.0	19.0	5.2
6. 30-min aging, rinsed, aged 5 hours, 15-min contact	Control	11.0	1.0	Dry - Dry	83.0	13.0	16.6	2.6
	Direct	11.0	1.0	Dry - Dry	86.0	11.0	17.3	2.1
	Vapor	11.0	1.0	Dry - Dry	80.0	19.0	15.9	3.9

Table B-7. Physical/Chemical Data Following Agent (TVX) Contact,
25.0 mg/Plate (Polyurethane Paint)

Test Phase	Contact type	Condition of agent on test plate prior to rinse removal			Chemical analysis for residual agent on plate			
		Area % spread on plate		Degree of wetness edge to center	Micrograms of agent left on plate		Percent residual agent on plate	
		Mean	± S.D.		Mean	± S.D.	Mean	± S.D.
1. 30-min aging, no rinse, no aging, 60-min contact	Control	26.0	2.0	Wet - Wet	20,022.0	1,481.0	80.1	5.9
	Direct	24.0	2.0	Wet - Wet	2,051.0	544.0	8.2	2.2
	Vapor	22.0	2.0	Wet - Wet	16,578.0	2,301.0	66.3	9.2
2. 30-min aging, rinsed, no aging, 60-min contact	Control	21.0	1.0	Wet - Wet	8.0	2.0	<0.1	<0.1
	Direct	21.0	1.0	Wet - Wet	9.0	3.0	<0.1	<0.1
	Vapor	19.0	1.0	Wet - Wet	9.0	2.0	<0.1	<0.1
3. 30-min aging, rinsed, aged 15 min, 60-min contact	Control	17.0	2.0	Wet - Wet	16.0	8.0	<0.1	<0.1
	Direct	20.0	3.0	Wet - Wet	16.0	9.0	<0.1	<0.1
	Vapor	18.0	2.0	Wet - Wet	16.0	6.0	<0.1	<0.1
4. 30-min aging, rinsed, aged 15 min, 15-min contact	Control	21.0	1.0	Wet - Wet	12.0	2.0	<0.1	<0.1
	Direct	22.0	0.0	Wet - Wet	7.0	4.0	<0.1	<0.1
	Vapor	21.0	1.0	Wet - Wet	17.0	10.0	<0.1	<0.1
5. 30-min aging, rinsed, aged 5 hours, 60-min contact	Control	17.0	1.0	Wet - Wet	21.0	3.0	0.1	<0.1
	Direct	18.0	1.0	Wet - Wet	21.0	5.0	0.1	<0.1
	Vapor	18.0	<1.0	Wet - Wet	18.0	3.0	0.1	<0.1
6. 30-min aging, rinsed, aged 5 hours, 15-min contact	Control	15.0	1.0	Wet - Wet	11.8	2.4	<0.1	<0.1
	Direct	15.0	2.0	Wet - Wet	14.3	2.7	<0.1	<0.1
	Vapor	16.0	2.0	Wet - Wet	16.7	4.4	<0.1	<0.1

Table B-8. Physical/Chemical Data Following Agent (TVX) Contact,
25.0 mg/Plate (Alkyd Paint)

Test Phase	Contact type	Condition of agent on test plate prior to rinse removal			Chemical analysis for residual agent on plate			
		Area % spread on plate		Degree of wetness edge to center	Micrograms of agent left on plate		Percent residual agent on plate	
		Mean	± S.D.		Mean	± S.D.	Mean	± S.D.
1. 30-min aging, no rinse, no aging, 60-min contact	Control	17.0	1.0	Wet - Wet	18,833.0	373.0	75.3	1.5
	Direct	16.0	2.0	Wet - Wet	6,336.0	783.0	25.3	3.1
	Vapor	16.0	1.0	Wet - Wet	15,266.0	1,808.0	61.1	7.2
2. 30-min aging, rinsed, no aging, 60-min contact	Control	15.0	<1.0	Wet - Wet	199.0	22.0	0.8	0.1
	Direct	17.0	1.0	Wet - Wet	230.0	40.0	0.9	0.2
	Vapor	16.0	1.0	Wet - Wet	240.0	27.0	1.0	0.1
3. 30-min aging, rinsed, aged 15 min, 60-min contact	Control	14.0	1.0	Wet - Wet	212.0	23.0	0.8	0.1
	Direct	16.0	1.0	Wet - Wet	186.0	24.0	0.7	0.1
	Vapor	16.0	1.0	Wet - Wet	205.0	8.0	0.8	<0.1
4. 30-min aging, rinsed, aged 15 min, 15-min contact	Control	16.0	1.0	Wet - Wet	95.0	16.0	0.4	0.1
	Direct	15.0	0.0	Wet - Wet	86.0	18.0	0.3	0.1
	Vapor	15.0	0.0	Wet - Wet	83.0	9.0	0.3	<0.1
5. 30-min aging, rinsed, aged 5 hours, 60-min contact	Control	12.0	1.0	Wet - Wet	90.0	5.0	0.4	<0.1
	Direct	13.0	1.0	Wet - Wet	98.0	15.0	0.4	0.1
	Vapor	14.0	1.0	Wet - Wet	115.0	19.0	0.5	0.1
6. 30-min aging, rinsed, aged 5 hours, 15-min contact	Control	13.0	<1.0	Wet - Wet	99.3	7.1	0.4	<0.1
	Direct	13.0	1.0	Wet - Wet	93.3	9.5	0.4	<0.1
	Vapor	13.0	<1.0	Wet - Wet	98.0	13.0	0.4	<0.1

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REPLY TO
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JUL 11 2016

MEMORANDUM THRU Director, Edgewood Chemical Biological Center, (RDCB-D/
Dr. Joseph Corriveau), 5183 Blackhawk Road, Aberdeen Proving Ground, Maryland 21010-5424

FOR Defense Technical Information Center, 8725 John J. Kingman Road, Ft Belvoir, VA 22060

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1. This action is in response to an Edgewood Chemical Biological Center (ECBC) Internal Request for a Change in Distribution.
2. The attached listed documents have been reviewed by ECBC Subject Matter Experts and deemed suitable for the change in distribution to read "Approved for public release; distribution unlimited."
3. The point of contact is Adana Eilo, ECBC Security Specialist, (410) 436-2063 or adana.l.eilo.civ@mail.mil.

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1. Manthie, J.H., Heitkamp, D.H. Dorsey, R.W., Stark, W.C., Bona, D.M., Moore, R.D., and Cameron, K.P., *Mustard Contact Hazard, Correlation of Effects on Skin with Contamination Levels Recovered from Dental Dam and Painted Steel Surfaces*, CRDEC-TR-88142, August 1988 (Dist. B. - U. S. Government Agencies) **DTIC: CBRNIAC-CB-009397**
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3. Manthie, J.H.; Heitkamp, W.C.; Starke, W.C.; Braun, R.A.; Bona, D.M.; Moore, R.D.; Cameron, K.P.; Heyl, W. C., *Toxicological Evaluation of the Contact and Vapor Hazards of VX and Thickened VX (TVX) in Rabbits*, CRDEC-TR-84072; U.S. Army Chemical Research, Development and Engineering Center: Aberdeen Proving Ground, MD, 1985 (Dist. B. - U. S. Government Agencies) **DTIC: ADB095637**
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