

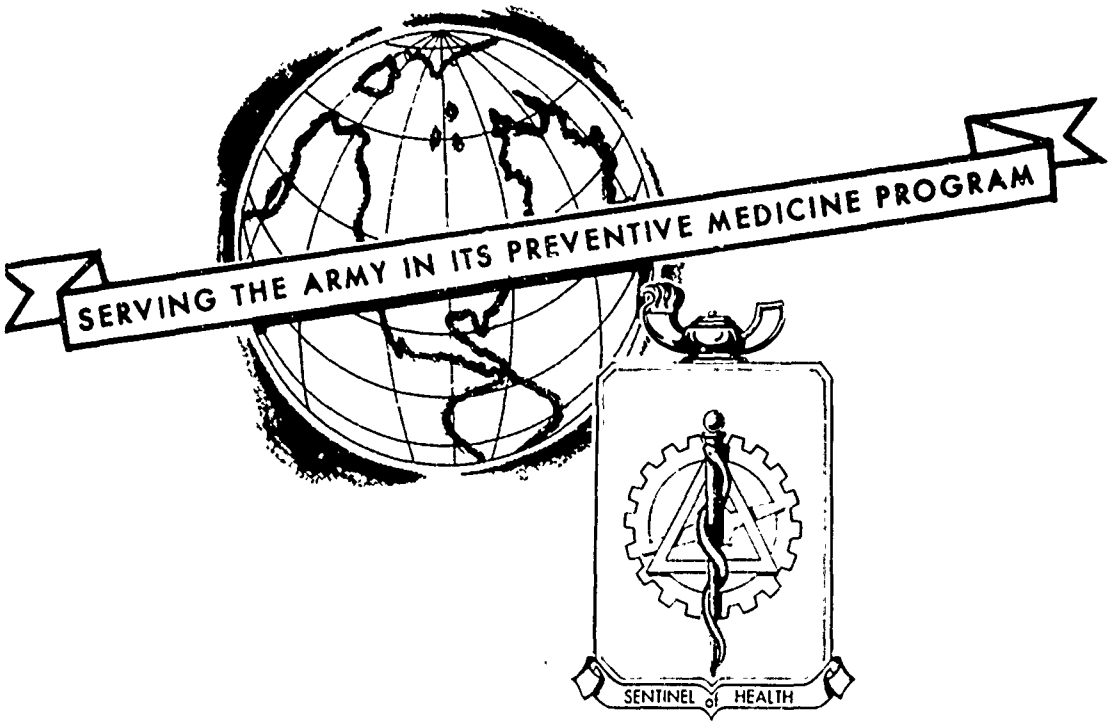
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RELATIVE TOXICITY OF CANDIDATE MOTHPROOFING
UNIFORM IMPREGNANT HEXADECYLPYRIDINIUM CHLORIDE
SPECIAL STUDY NO. 33-4-68/71
NOVEMBER 1967-DECEMBER 1969



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RELATIVE TOXICITY OF CANDIDATE MOTHPROOFING
UNIFORM IMPREGNANT HEXADECYLPYRIDINIUM CHLORIDE
SPECIAL STUDY NO. 33-4-68/71
NOVEMBER 1967-DECEMBER 1969

ABSTRACT

The relative toxicity of a candidate uniform mothproofing impregnant hexadecylpyridinium chloride (HPC) was investigated using mice, rats, guinea pigs, rabbits and humans with the following findings:

- a. The technical grade compound and aqueous solutions of 1.0 percent concentration (w/v) or greater caused skin irritation in rabbits.
- b. Eye irritation in rabbits resulted from single 24-hour application of a 0.1 percent (w/v) aqueous solution but no irritation resulted from a 0.01 percent (w/v) solution.
- c. The skin irritating properties of hexadecylpyridinium chloride in guinea pigs prevented the determination of the sensitization potential of this compound in comparison with dinitrochlorobenzene.
- d. Chronic ingestion ad libitum of a diet containing 300 ppm hexadecylpyridinium chloride for 90 days caused minimal changes in rats.
- e. Cloth impregnated with 0.5 percent (w/w) hexadecylpyridinium chloride and applied continuously to the shaved skin of rabbits for 90 days produced no remarkable degree of skin irritation or evidence of potential toxic hazard.
- f. A prophetic patch test on humans of wool fabric impregnated with hexadecylpyridinium chloride at a concentration of 0.5 percent by weight produced no evidence of skin irritation or sensitization.
- g. Under the conditions of impregnation specified, HPC should not present a hazard when used as a mothproofing compound in wool fabric at a deposition of 0.5 percent (w/w). Since the weight of wool fabric to be used has not been specified it is impossible to estimate the degree of safety.



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DEC 1970

RELATIVE TOXICITY OF CANDIDATE MOTHPROOFING
UNIFORM IMPREGNANT HEXADECYLPYRIDINIUM CHLORIDE
SPECIAL STUDY NO. 33-4-68/71
NOVEMBER 1967-DECEMBER 1969

1. REFERENCES.

a. Letter, Stored-Product Insects Research and Development Laboratory, US Department of Agriculture, Savannah, Georgia, dated 7 August 1967, to this Agency.

b. Letter, Harris Research Laboratories, Washington, D. C., dated 18 September 1967, to this Agency.

c. Procedural Guide for the Toxicology Division, US Army Environmental Hygiene Agency, 1968 (revised 1969).

d. SOP for Patch Testing, US Army Environmental Hygiene Agency, 12 October 1962.

2. PURPOSE. The purpose of this study was to acquire information concerning the toxicity of hexadecylpyridinium chloride (HPC) in man and animals. This information provides a basis for advising on possible hazards associated with the use of this compound as a mothproofing agent at a concentration of 0.5 percent by weight in the wool fabric of uniforms. The weight of the woolen fabrics in which the candidate compound is ultimately to be used is not stated in the request for toxicologic clearance (ref para 1a).

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Study No. 33-4-68/71, Nov 67-Dec 69

3. BACKGROUND. The proposed mothproofing chemical HPC submitted was a white crystalline solid, produced by Eastman Organic Chemicals, Catalog No. P 5361. Samples of the wool fabric impregnated with this compound at a deposition of 0.5 percent by weight were supplied for testing by Harris Research Laboratories, Washington, D. C., a contractor for the US Department of Agriculture, Savannah, Georgia (ref para 1b). Harris Research Laboratories also provided untreated wool fabric and fabric which had been treated with a sodium tetraborate (borax) solution. The treatment of the fabric with HPC was carried out by soaking wool fabric in aqueous solutions (pH 9.2) containing the mothproofing chemical and 0.01M sodium tetraborate.

4. SUMMARY OF FINDINGS. The relative toxicity of HPC was investigated by this Agency using mice, rats, guinea pigs, rabbits and humans. No data were found which indicated a potential toxicologic hazard from wearing of uniforms impregnated with 0.5 percent (w/w) HPC. An infrared spectrum of the technical grade compound evaluated by USAEHA is found in Appendix A. Definitions, abbreviations, symbols and clinical chemistry reporting units used in this report are found in Appendix B. Numerical data found in the Appendices are expressed in terms of the mean plus or minus one standard deviation ($\bar{X} \pm S_x$). Statistical significance selected in this report is at the .01 level. All regression lines were fitted by the method of least squares. A tabular presentation of toxicity data follows:

TABULAR PRESENTATION OF DATA		
TEST	RESULTS	INTERPRETATION
<u>SKIN IRRITATION STUDIES</u> <u>RABBITS</u>		
<u>Primary Irritation Evaluation</u>		
Single 24-hour application to intact and abraded skin		
0.5 gm technical grade compound applied to each of 4 rabbits	Well defined erythema of the intact skin and the skin surrounding an abrasion after 72 hours (ref Appendix C).	Compound in dry crystalline state produces delayed irritation effects of the skin. Personnel working with the technical grade compound should use eye and skin protection.
0.5 gm technical grade compound dissolved in 1 ml acetone applied to each of 4 rabbits	Severe erythema and edema of the intact skin and the skin surrounding an abrasion after 24 and 72 hours (ref Appendix C).	Acetone solutions of this compound produce severe primary irritation of the skin. Personnel working with acetone solutions of the compound at this concentration should use eye and skin protection.

TABULAR PRESENTATION OF DATA		INTERPRETATION
TEST	RESULTS	
<u>SKIN IRRITATION STUDIES</u> <u>RABBITS (Cont)</u>		
<u>Determination of Non-Irritating Concentration</u>		
0.01 ml of a 10% (w/v) solution in water applied to each of 6 rabbits	Skin injury varied from slight capillary injection to severe erythema. Individual irritation scores ranged from 0 to 4 with a mean of 2.4 (ref Appendix D).	There was considerable variation between individual animals. Personnel coming into contact with the compound at this concentration should use eye and skin protection.
0.01 ml of a 1.0% (w/v) solution in water applied to each of 6 rabbits	Skin injury varied from slight capillary injection to slight erythema. Individual irritation scores ranged from 0 to 3 with a mean of 1.2 (ref Appendix D).	There was considerable variation between individual animals. Personnel coming into contact with the compound at this concentration should use eye and skin protection.
0.01 ml of a 0.1% (w/v) solution in water applied to each of 6 rabbits	Skin irritation not remarkable	The compound at this concentration is not expected to cause irritation to human skin if washed off immediately.
0.01 ml of a 0.01% (w/v) solution in water applied to each of 6 rabbits	Skin irritation not remarkable	
0.01 ml of water applied to each of 6 rabbits	Skin irritation not remarkable	

TABULAR PRESENTATION OF DATA		INTERPRETATION
TEST	RESULTS	
<u>EYE IRRITATION STUDIES</u> <u>RABBITS</u> Single 24-hour application of 0.2 ml of a 0.1% (w/v) solution in water applied to one eye of each of 5 rabbits Single 24-hour application of 0.2 ml of a 0.01% (w/v) solution in water applied to one eye of each of 5 rabbits	24-hour observation showed no corneal or iris irritation. Redness of the conjunctiva observed in 4 of 5 rabbits. Observation on the 7th day showed all eyes to be normal. 24-hour observation showed no corneal, iris, or conjunctival irritation.	Eye protection should be worn when handling compound at this concentration. Eye irritation to humans is not expected if a 0.01% (w/v) solution of the compound should accidentally get into the eye, provided it is washed out immediately.
<u>APPROXIMATE LETHAL DOSE</u> <u>INTRAPERITONEAL</u> <u>Rats (male)</u> Distilled water diluent	ALD - 27 mg/kg - Poor motor coordination, weakness, and tremors were predominant signs at lethal dosages.	
<u>Guinea Pigs (female)</u> Distilled water diluent	ALD - 15 mg/kg - Reduced activity and weakness were predominant signs at lethal dosages.	
<u>Rabbits (male)</u> Distilled water diluent	ALD - 7 mg/kg - Weakness was predominant sign at lethal dosages.	

TABULAR PRESENTATION OF DATA	
TEST	INTERPRETATION
<u>ACUTE ADMINISTRATION</u> <u>INTRAPERITONEAL</u> <u>Effect on Rat Activity</u> <u>Exposed</u> Group of 6 male rats administered a single dose of HPC at a dosage of 5 mg/kg (1/5 ALD) <u>Control</u> Group of 6 male rats administered a single dose of 0.5 ml water	
An apparent reduction in the voluntary activity of rats treated with HPC was observed for 1 to 2 days after treatment when compared to controls for the same period.	HPC at a dosage of 1/5 the ip ALD influences voluntary wheel running in the rat.

TABULAR PRESENTATION OF DATA		
TEST	RESULTS	INTERPRETATION
<u>ACUTE ADMINISTRATION</u> <u>INTRAPERITONEAL</u> <u>Effect on Rat Activity</u> <u>Exposed</u> Group of 6 male rats administered a single dose of HPC at a dosage of 5 mg/kg (1/5 ALD) <u>Control</u> Group of 6 male rats administered a single dose of 0.5 ml water	An apparent reduction in the voluntary activity of rats treated with HPC was observed for 1 to 2 days after treatment when compared to controls for the same period.	HPC at a dosage of 1/5 the ip ALD influences voluntary wheel running in the rat.

TABULAR PRESENTATION OF DATA		INTERPRETATION
TEST	RESULTS	
LD50 INTRAPERITONEAL		Classified as a toxic compound. Parameters of classification found in Appendix E.
<u>Mice</u> (male) - 6 mice per dose level - distilled water diluent	5.4 mg/kg (3.9 - 7.4 mg/kg)* - Weakness was predominant sign at lethal dosages. No signs at non- lethal dosages.	
<u>Rats</u> (male) - 6 rats per dose level - distilled water diluent	35 mg/kg (29 - 41 mg/kg)* - Reduced locomotor activity and piloerection were noted at all dosages.	
LD50 ORAL		
<u>Mice</u> (male) - 6 mice per dose level - distilled water diluent	125 mg/kg (99 - 159 mg/kg)* - Weakness was predominant sign at lethal dosages. No signs at non- lethal dosages.	
<u>Rats</u> (male) - 6 rats per dose level - distilled water diluent	341 mg/kg (286 - 406 mg/kg)* - Diarrhea was predominant sign at lethal dosages. No signs at non- lethal dosages.	

* 95% Confidence limits

TABULAR PRESENTATION OF DATA

TEST	RESULTS	INTERPRETATION
<u>SENSITIZATION STUDIES</u>		
<u>Guinea Pigs (male)</u> Intradermal injections of 0.1 ml of 0.1% suspension (w/v) test and control compound in a mixture containing 1 volume of propylene glycol and 29 volumes of saline 10 test guinea pigs 10 positive control guinea pigs 10 control guinea pigs: 5 receiving challenge dose of test compound without prior sensitizing doses. 5 receiving challenge dose of positive control (DNCB) without prior sensitizing doses.	The initial intradermal sensitizing dose of HPC and DNCB caused inflammatory responses of similar intensity in guinea pigs 24 hours after injection. The challenge dose (last intradermal injection) of HPC caused a greater skin response than the initial sensitizing dose in test guinea pigs 24 hours after injection. However, the response of the 5 control guinea pigs receiving the challenge dose of HPC without prior sensitizing doses was of the same order of intensity as that seen in the test guinea pigs receiving the challenge dose of HPC after the sensitizing doses. The challenge dose of DNCB in the positive control guinea pigs caused a greater skin response 24 hours after injection than the initial sensitizing dose in test guinea pigs. However, the response reactions of the 5 control guinea pigs receiving the challenge dose of DNCB without prior sensitization was of the same intensity as that obtained from the initial intradermal dose in the positive control group.	The data from this study do not clearly indicate that HPC is a chemical sensitizer. The age of the guinea pigs seems to be an important factor in the skin irritation response with HPC as with other compounds.

TABULAR PRESENTATION OF DATA		
TEST	RESULTS	INTERPRETATION
<u>STUDIES TO DETERMINE</u> <u>CHOLINESTERASE ACTIVITY</u> <u>RABBITS</u> <u>Single Intraperitoneal</u> <u>Administration - (1 rabbit</u> <u>per dosage)</u> Distilled water diluent Dosages: 3.3 mg/kg 5 mg/kg 7 mg/kg 10 mg/kg 15 mg/kg 25 mg/kg 33 mg/kg 75 mg/kg	Dosages of 7 mg/kg and higher caused death in 2 to 10 hours. RBC and plasma cholinesterase activity did not decrease in blood samples taken 1, 2, 4, 24 and 48 hours after administration of the test compound. The major signs were lethargy and skeletal muscle relaxation.	Single lethal and nonlethal intraperitoneal dosages of HPC do not cause RBC or plasma cholinesterase inhibition in rabbits.

TABULAR PRESENTATION OF DATA		
TEST	RESULTS	INTERPRETATION
<u>STUDIES TO DETERMINE CHOLINESTERASE ACTIVITY RABBITS (Cont)</u>		
<u>Multiple Intraperitoneal Administration</u>		
<u>Treatment Group</u>		
Group of 3 rabbits received a daily dosage for 10 consecutive days of 1.4 mg/kg HPC administered in 1.0 ml distilled water	RBC and plasma cholinesterase activity did not change significantly during the 10 day administration of HPC. No changes occurred in body weight between control and treated group.	Unable to demonstrate cholinesterase inhibition.
<u>Control Group</u>		
Group of 3 rabbits received a daily dosage for 10 consecutive days of 1.0 ml distilled water	No deaths, signs or changes in body weights were observed as a result of this treatment.	No signs of intoxication from distilled water.

TABULAR PRESENTATION OF DATA		
TEST	RESULTS	INTERPRETATION
PROLONGED ADMINISTRATION STUDIES		
<u>90-Day Feeding Study - Rats</u>		
Groups of 6 male and 6 female rats each were placed on each dietary level of HPC. The chemical was added to food by dissolving measured amounts of compound in ethanol and mixing with feed in the proportion of 250 ml alcohol per kilogram of feed. Alcohol was removed from feed by steam evaporation. Treated feeds were stored at 4° C.	Synopsis of data found in Appendix F, Tables 1, 2, 3 and 4. All male and female rats fed a diet containing 5,000 and 10,000 ppm HPC died within three weeks after initiation of the test. Female rats fed diets containing 300, 800 and 2,000 ppm of HPC and male rats fed diets of 800 and 2,000 ppm showed a significant increase in caecum to body weight ratios when compared to controls. Irrespective of the dietary level or sex, no great differences were noted in the genera of the bacterial flora of the caeca. However, it was noted that as the concentration of HPC in the diet increased, the total number of microorganisms (determined by standard dilution technique) recovered in the caecal contents decreased (see Figure 1). A positive correlation coefficient of 0.8 was found between dietary levels of HPC and increases in caecum to body weight ratios (see Figure 1). Body weight gain; liver and kidney to body weight ratios; and food utilization of female rats fed a diet containing 2,000 ppm	A positive correlation exists between increased amounts of HPC ingested and increases in the caecum to body weight ratio. Chronic ingestion of diets containing 300 ppm HPC or greater should be avoided.
Rats were maintained on diets containing HPC in their feed at the following levels: 0 ppm (0.0% w/w) solvent control 125 ppm (0.0125% w/w) 300 ppm (0.03% w/w) 800 ppm (0.08% w/w) 2000 ppm (0.20% w/w) 5000 ppm (0.50% w/w) 10000 ppm (1.00% w/w)		

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Nov 67-Dec 69

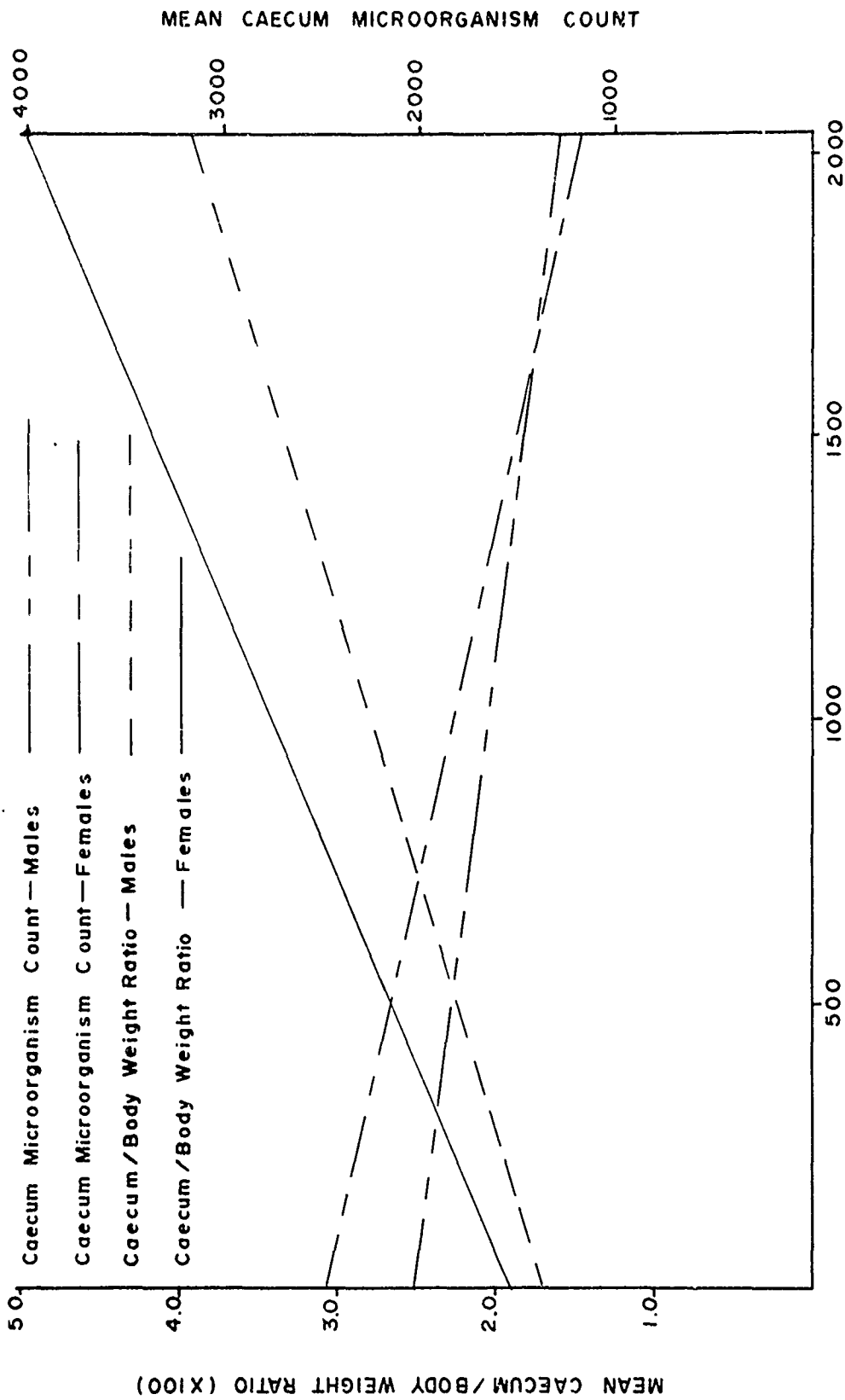


FIGURE 1. RELATIONSHIP OF DIETARY LEVELS OF HEXADECYLPIRIDINIUM- PPM TO CAECUM MICROORGANISM COUNT OF MALE AND FEMALE RATS. REGRESSION LINES FITTED BY METHOD OF LEAST SQUARES.

TEST	TABULAR PRESENTATION OF DATA	RESULTS	INTERPRETATION
<u>PROLONGED ADMINISTRATION STUDIES</u> (Cont) <u>90-Day Wear Test - Rabbits</u> Groups of male, shaved, New Zealand White rabbits were observed for skin irritation and metabolic effects which may have been produced by continuously wearing woolen cloth around their midsections for 90 days. The pieces of cloth were either untreated, treated with borax buffer or impregnated with HPC at an estimated deposition of 0.5% (w/w). The pieces of cloth (4 x 15 inches) were applied on day 1 of test and new cloth was reapplied at 2, 4, and 8 weeks thereafter. Selected groups had 2.0 ml of artificial eccrine sweat (Appendix G) applied under the cloth 5 days per week. These groups had their skin abraded (2 x 2 inches) on the dorsal side, whenever the cloth was freshly applied.		Synopsis of data found in Appendices II through K.	

TEST	RESULTS	INTERPRETATION
<u>PROLONGED ADMINISTRATION STUDIES</u> (Cont) <u>90-Day Wear Test - Rabbits</u> The type of application to rabbits was as follows: Wool cloth treated with sodium tetraborate applied to intact skin of 6 rabbits Wool cloth treated with sodium tetraborate applied with 2.0 ml sweat to abraded skin of 5 rabbits Wool cloth impregnated with 0.5% (w/w) HPC applied to intact skin of 6 rabbits Wool cloth impregnated with 0.5% (w/w) HPC applied with 2.0 ml sweat to abraded skin of 6 rabbits Nothing applied (cage controls) 8 rabbits Untreated wool cloth applied to intact skin of 9 rabbits Untreated wool cloth applied with 2.0 ml sweat to abraded skin of 9 rabbits	No significant changes were noted at the end of the 90-day wear test in rabbit organ-to-body weight ratios of liver, kidney, spleen, testes, adrenals, caecum and brain. No significant changes were noted in LDH, blood glucose, SGOT, BUN, total serum protein, and serum amylase values. Brain cholinesterase values from HPC treated animals did not vary significantly from cage controls. Values for hematocrit, total leucocyte count and total erythrocyte count did not change significantly during the test period. There were no differences in average dermal irritation scores for rabbits wearing untreated cloth, borax, or HPC impregnated cloth whether dry or sweat treated. Gross and microscopic examination of tissues and organs indicated no appreciable difference in pathology between the treatment and control groups.	Cloth impregnated with 0.5% (w/w) HPC caused no increase in skin irritation as compared to animals wearing control cloth. Clinical chemistry and hematological data from rabbits showed no evidence of potential toxicity under the conditions of evaluation.

TABULAR PRESENTATION OF DATA		
TEST	RESULTS	INTERPRETATION
<u>PROPHETIC PATCH TEST</u>		
<u>Humans</u>		
HPC impregnated in wool fabric (1 inch square) at 0.5% (w/w) concentration and untreated wool fabric (1 inch square) was applied to the intact skin of 31 volunteers for 48 hours application (sensitizing dose) followed 2 weeks later by a 48 hour application (challenge dose) to approximately the same area of skin on the left arm (ref para 1d).	No evidence of skin irritation or sensitization was found which was attributed to the material being tested.	The formulation tested should not be irritating or sensitizing to human skin when used under controlled conditions.

5. DISCUSSION.

a. The technical grade compound and aqueous solutions of 1.0 percent or greater of HPC were shown to cause primary skin irritation in rabbits. These solutions and the technical grade compound should be handled with care, wearing eye and skin protective equipment. Since there was no difference between the skin irritation in the control animals and the rabbits wearing cloth impregnated with HPC and treated with artificial eccrine sweat, it would appear that simulated sweat does not leach the HPC from the cloth in a form that causes topical effects.

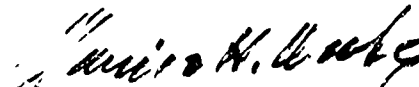
b. The relative hazard from accidental chronic ingestion of HPC does not appear to be great although some biologic action was evident in female rats fed moderately low dietary levels of 300 ppm and in male rats fed 800 ppm. These levels produced increases in caecum-to-body weight ratios and a slight decrease in total number of caecal microorganisms. Female rats seem to be slightly more sensitive to dietary levels of HPC than male rats. A positive correlation of 0.8 between levels of HPC and increase in caecum-to-body weight ratios was found both in females and males (Figure 1) which indicates a similar dose response relationship in both sexes to the compound.

c. The animal laboratory toxicity data and the results from a prophetic patch test in man indicate HPC impregnated in wool cloth at 0.5 percent (w/w) would present little potential hazard to man under the conditions of evaluation.

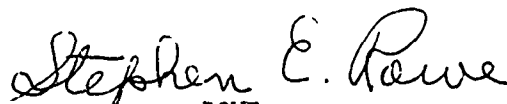
6. CONCLUSION. Under the conditions of impregnation specified, HPC should not present a hazard when used as a mothproofing compound in wool fabric at a deposition of 0.5 percent (w/w). Since the weight of wool fabric to be used has not been specified it is impossible to estimate the degree of safety. Skin and eye contact with the technical grade compound or with aqueous solutions of 1.0 percent or greater should be avoided. In the event solution of HPC or the technical grade compound should come in contact with the skin or eyes it should be washed off with copious amounts of water.

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Study No. 33-4-68/71, Nov 67-Dec 69

ACTION OFFICERS:

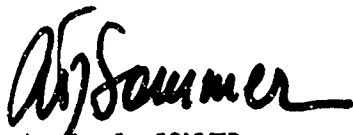

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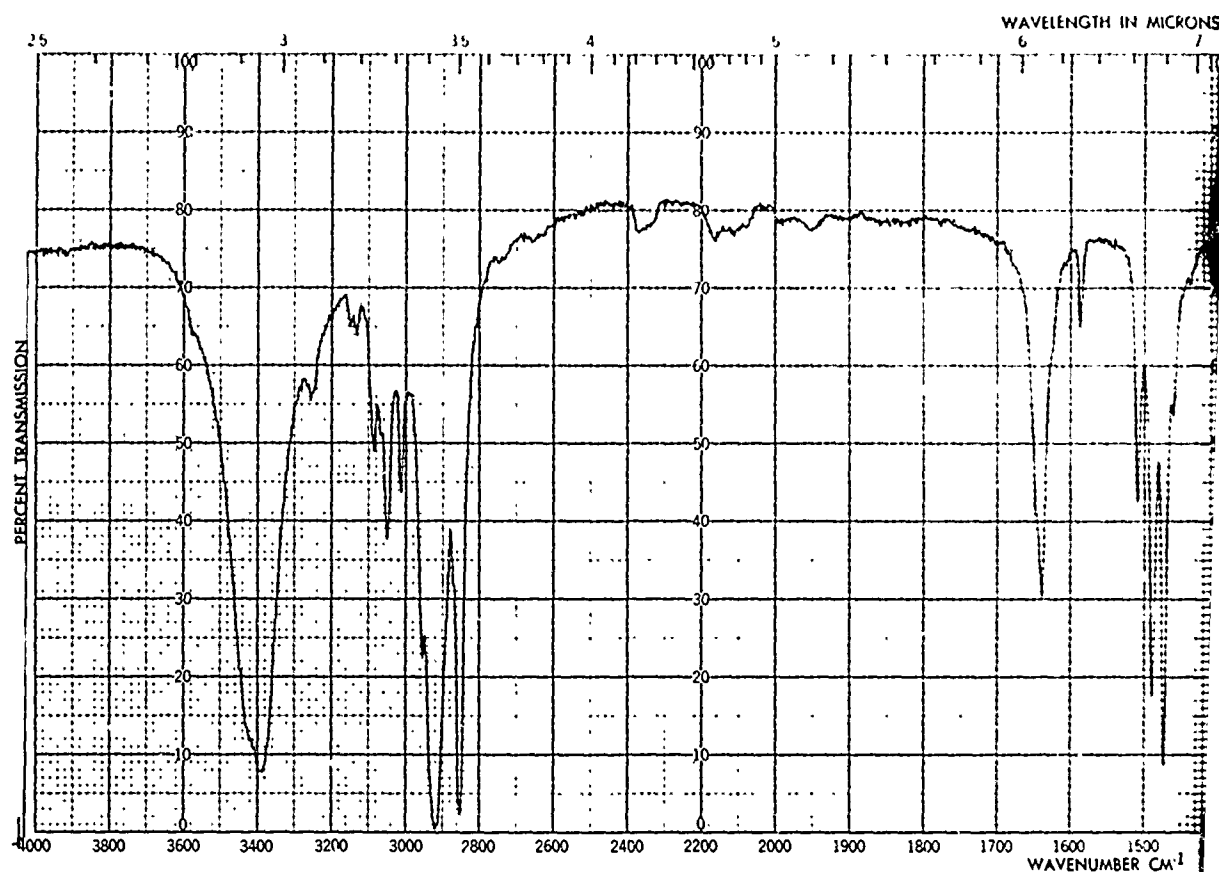

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APPENDIX A
INFRARED SPECTRUM OF HEXAD



SAMPLE: HEXADECYLPYRIDINI
KBr DISC, AIR

SLIT: ROUTINE

SPEED: 200CM⁻¹/MIN

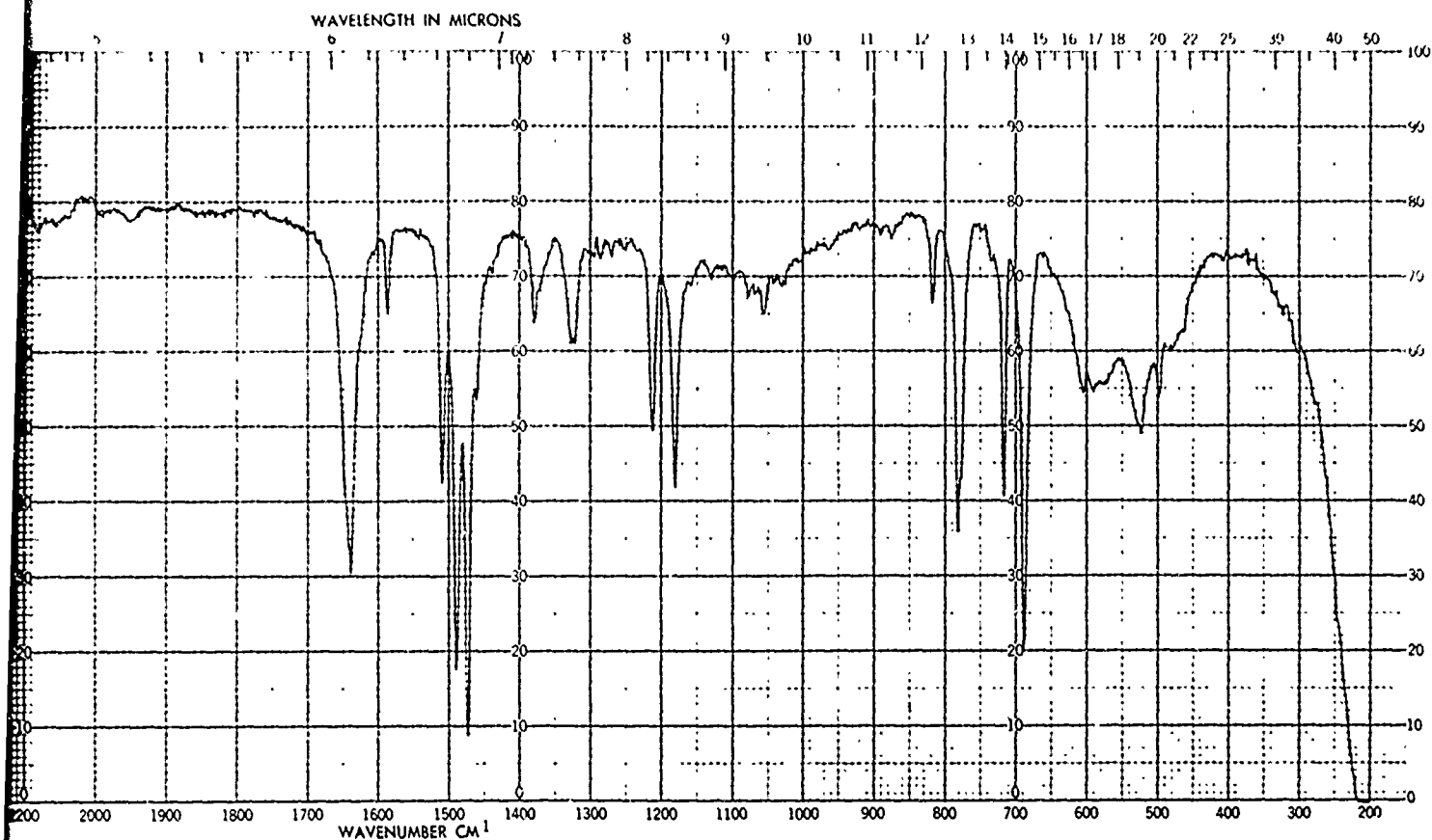
GAIN: 3%

PERIOD: 2

ORDINATE: 0—100%T

AT Rel Toxicity of Candidate Mothproofing Uniform Impregnant,
Study No. 33-4-68/71, Nov 67-Dec 69

APPENDIX A
INFRARED SPECTRUM OF HEXADECYLPYRIDINIUM CHLORIDE



SAMPLE: HEXADECYLPYRIDINIUM CHLORIDE
KBr DISC, AIR REFERENCE

SLIT: ROUTINE

SPEED: 200CM⁻¹ / MIN

GAIN: 3%

PERIOD: 2

ORDINATE: 0—100%T

APPENDIX B

GLOSSARY OF RECURRING DEFINITIONS, ABBREVIATIONS, SYMBOLS AND CLINICAL
CHEMISTRY REPORTING UNITS USED BY THE TOXICOLOGY DIVISION, USAEHA

Definitions of medical terms and abbreviations used in this report are in agreement with the New Gould Medical Dictionary, Second Edition, published by the Blakiston Division of McGraw-Hill Book Company, Inc. Statistical terms and abbreviations are in agreement with those found in J. Maxwell Little's, An Introduction to the Experimental Method, 1961, Burgess Publishing Company, Minneapolis, Minn. The following terms and abbreviations are either not found in the above references or have been modified to fit the special purposes of this report. Some of the items have been included below for special emphasis.

DEFINITIONS

WORD

DEFINITION

Acute Exposure or Application

One exposure to exogenous test material for no longer than 8 hours. Animals are normally observed for 14 days after exposure.

Approximate Lethal Dose

In range finding the first dose of the lowest series of three ascending doses (each being 50% higher in concentration than the previous) all of which produce fatalities.

Caraway Unit

That amount of amylase activity which causes the hydrolysis of 10 mg of starch in 30 minutes at 37°C to a stage at which there is no absorbance at 660 mμ after reaction with iodine.

Chronic Exposure

Repeated daily or constant exposure to a test material for 60 or more days. Post exposure observation period will vary.

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<u>WORD</u>	<u>DEFINITION</u>
Garry & Routh Unit	That amount of cholinesterase activity which causes the liberation of one micromole of SH groups from acetylthiocholine in 3 minutes at 37°C per milliliter of serum, plasma or packed red cells or per gram tissue.
Hazard Evaluation	A study performed to estimate the degree of danger associated with the use of a material under specified conditions of use.
Karmen Unit	That amount of transaminase activity which causes at 25°C and 340 mμ a decrease in optical density of 0.001 per minute per milliliter of serum.
Primary Irritation	A local inflammatory reaction of the skin, produced by a compound, which does not produce destruction or irreversible change at the site of contact.
Skin Sensitizer	A compound which produces an allergic dermatitis under the conditions of test.
Subacute Exposure	Repeated daily or constant exposure to a test material for no longer than 59 days or less than 2 days. Post observation period will vary.
Technical Grade Compound	As produced by the manufacturers for their commercial compound; definition dependent upon manufacturers' criteria.
Wacker Unit	That amount of dehydrogenase activity which causes an increase of optical density at 340 mμ of 0.001 per minute per ml of serum at 25°C.

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ABBREVIATIONS AND SYMBOLS

<u>ABBREVIATION</u>	<u>MEANING</u>
ACh	acetylcholine
AChE	acetylcholinesterase
ALD	approximate lethal dose
antiChE	anticholinesterase
BUN	blood urea nitrogen
ChE	cholinesterase
CNS	central nervous system
df	degree of freedom
DNCB	dinitrochlorobenzene
Hb	hemoglobin
ia	intra-arterial
im	intramuscular
ip	intraperitoneal
IR	infrared
iv	intravenous
LD ₅₀	median lethal dose
LDH	lactic dehydrogenase
meq	milliequivalent
$p < .01$	The probability of the change from normal or control being due to chance alone is less than 1 out of 100.

USAEHA-MT Rel Toxicity of Candidate Mothproofing Uniform Impregnant,
Study No. 33-4-68/71, Nov 67-Dec 69

<u>ABBREVIATION</u>	<u>MEANING</u>
pc	percutaneous
ppm	parts per million
P.E.G. 200	polyethylene glycol having a modal molecular weight of 200
SGOT	serum glutamic oxaloacetic transaminase
SGPT	serum glutamic pyruvic transaminase
sc	subcutaneous
SD or ($S_{\bar{x}}$)	standard deviation
SE or ($SE_{\bar{x}}$)	standard error
v/v	volume-to-volume ratio
w/v	weight-to-volume ratio
w/w	weight-to-weight ratio

<u>CLINICAL CHEMISTRY TEST</u>	<u>REPORTING UNITS</u>
Serum amylase	Caraway Units
Acetylcholinesterase	Garry and Routh Units
Blood urea nitrogen	Milligrams per 100 cc's of serum
Blood glucose	Milligrams per 100 cc's of serum
Cholinesterase	Garry and Routh Units
Lactic dehydrogenase	Wacker Units
Serum glutamic oxaloacetic transaminase	Karmen Units
Serum glutamic pyruvic transaminase	Karmen Units
Serum protein	Grams per 100 cc's of serum

APPENDIX C

EVALUATION OF SKIN REACTIONS

Erythema and Eschar Formation

No erythema	0
Very slight erythema (barely perceptible)	1
Well defined erythema	2
Moderate-to-severe erythema	3
Severe erythema (beet redness to slight eschar formation)	4

Edema Formation

No edema	0
Very slight (barely perceptible)	1
Slight edema (edges of area well defined by definite raising)	2
Moderate edema (edges raised approximately 1 mm)	3
Severe edema (raised more than 1 mm and extending beyond area of exposure)	4

An individual irritation score is equal to the sum of the scores for
edema formation and erythema and eschar formation.

USAEHA-MT Rel Toxicity of Candidate Mothproofing Uniform Impregnant,
Study No. 33-4-68/71, Nov 67-Dec 69

APPENDIX D
SCALE FOR NUMERICAL SCORING OF INJURY TO RABBIT SKIN WHEN
DETERMINING NON-IRRITATING CONCENTRATION

<u>SKIN INJURY OBSERVATION</u>	<u>SCORE</u>
No reaction	0
Slight capillary injection	1
Strong capillary injection	2
Slight erythema	3
Strong erythema	4
Slight edema	5
Strong edema	6
Slight necrosis	8
Strong necrosis	10

APPENDIX E

TABULATION OF TOXICITY DOSES*

Commonly Used Terms	VARIOUS ROUTES OF ADMINISTRATION		
	LD ₅₀ Single Oral Dose Rats	Inhalation 4-Hr Vapor Exposure Mortality 2/6 - 4/6 Rats	LD ₅₀ Skin Rabbits
Highly toxic	50 mg/kg or less	100 ppm or less	43 mg/kg or less
Toxic	51-500 mg/kg	101-1000 ppm	44-350 mg/kg
Moderately Toxic	501-5000 mg/kg	1001-10,000 ppm	351-2800 mg/kg
Slightly Toxic	5001-15,000 mg/kg	10,001-100,000 ppm	2801-22,600 mg/kg
Practically Non-toxic	above 15,000 mg/kg	> 100,000 ppm	above 22,600 mg/kg

* Adapted from Hodge, H. C. and Sterner, J. H. American Industrial Hygiene
Association Quarterly, 10:4, 93, December 1943.

APPENDIX F

TABLE 1

90-DAY FEEDING STUDY HPC

SUMMARY OF DIET CONSUMPTION AND DOSAGE DATA OF WALE RATS

	Solvent Control	125 ppm	Dietary Levels HPC *		
			300 ppm	800 ppm	2000 ppm
Dosage HPC (mg/kg/day)	0.0	6.6	15.8	44.9	128.4
Grams diet eaten/rat/day	19.6	19.4	18.3	18.8	18.9
Grams diet eaten/kg/day	58.4	52.8	52.5	56.1	64.2
Mean weight gain of rats during study (grams)	240	234	224	211	117**
Feed utilization (total body weight gain/total food consumed)	0.125	0.127	0.125	0.116	0.077 **
Grams water intake/rat/day	41.7	31.4	31.8	30.8	24.3
Grams water intake/kg/day	119.4	94.8	92.5	92.9	100.6

* All rats fed a diet containing 5,000 and 10,000 ppm HPC died within three weeks after initiation of the test.

** Significantly different at the .01 level of probability.

APPENDIX F
TABLE 2
90-DAY FEEDING STUDY HPC
SUMMARY OF ORGAN-TO-BODY WEIGHT RATIOS OF FEMALE RATS

Dietary Levels (ppm)	Terminal body weight (gm)	Liver	Organ-to-body weight ratios (gm/100 gm body weight)	Spleen	kidney	Testes	Lung	Caecum
0.0 solvent control	423 ± 40	3.43 ±0.22		0.21 ±0.02	0.57 ±0.04	0.86 ±0.07	0.46 ±0.04	1.34 ±0.16
125	419 ± 23	3.89 ±0.31		0.20 ±0.03	0.59 ±0.02	0.82 ±0.11	0.51 ±0.04	1.00 ±0.35
300	413 ± 32	3.47 ±0.18		0.20 ±0.03	0.57 ±0.03	0.84 ±0.03	0.48 ±0.04	1.70 ±0.23
600	395 ± 23	3.79 ±0.19		0.19 ±0.04	0.61 ±0.06	0.88 ±0.15	0.49 ±0.03	2.78* ±0.31
2000	290* ± 12	3.37 ±0.33		0.27* ±0.03	0.75* ±0.04	1.07* ±0.05	0.55 ±0.03	4.25* ±0.51

* Significantly different at the .01 level of probability

USAMRIID Rel Toxicity of Candidate Bioprotecting Uniform Impregnant, Study No. 33-4-68/71, Nov 67-Dec 69

APPENDIX F

TABLE 3

90-DAY FEEDING STUDY HPC

SUMMARY OF DIET CONSUMPTION AND DOSAGE DATA OF FEMALE RATS

	Solvent Control	125 ppm	Dietary Levels HPC *		
			300 ppm	800 ppm	2000 ppm
Dosage HPC (mg/kg/day)	0.0	9.7	29.4	56.8	144.6
Grams diet eaten/rat/day	17.5	18.6	21.3	15.5	13.4
Grams diet eaten/kg/day	69.4	77.4	98.0	71.0	72.3
Mean weight gain of rats during study (grams)	106	123	104	86	61**
Feed utilization (total body weight gain/ total food consumed)	0.063	0.069	0.053	0.066	0.047**
Grams water intake/rat/day	24.6	25.6	25.1	22.4	19.8
Grams water intake/kg/day	112.8	107.7	126.6	103.2	105.6

* All rats fed a diet containing 5,000 and 10,000 ppm HPC died within three weeks after initiation of the test.

** Significantly different at the .01 level of probability.

APPENDIX F
TABLE 4
90-DAY FEEDING STUDY HPC
SUMMARY OF ORGAN-TO-BODY WEIGHT RATIOS OF FEMALE RATS

Dietary Levels HPC (ppm)	Terminal Body Weight (gm)	Organ-to-Body Weight Ratios (gm/100 gm body weight)			
		Liver	Spleen	Kidney	Lung
J.J solvent control	252 ± 13	2.94 ±0.32	0.24 ±0.02	0.59 ±0.02	0.59 ±0.02
125	275 ± 24	3.51 ±0.23	0.28 ±0.01	0.57 ±0.04	0.58 ±0.04
300	247 ± 18	3.57 ±0.23	0.28 ±0.04	0.62 ±0.02	0.62 ±0.05
800	250 ± 15	3.51 ±0.21	0.29 ±0.04	0.64 ±0.02	0.63 ±0.06
2000	211* ± 26	3.92* ±0.51	0.29 ±0.05	0.75* ±0.05	0.62 ±0.06
					5.72* ±0.93

* Significantly different at the .01 level of probability

APPENDIX G
FORMULATION OF ARTIFICIAL (ECCRINE) SWEAT*

Urea	1.72 gm/l
Glycine	0.20 "
Glucose	0.03 "
Lactic acid	2.50 "
NaCl	1.75 "
KCl	0.22 "
NH_4OH	Sufficient to adjust to pH 5.0

*This formulation includes all constituents described by Rothman, S.
(Physiology and Biochemistry of Skin, University of Chicago Press,
1954) as present in sweat at a concentration of 3 mg/100 cc or more.

APPENDIX II
 TABLE I
 90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF LABORATORY
 SUMMARY OF ORGAN-TO-BODY WEIGHT RATIOS

Type Application	Organ-to-Body Weight Ratios (gm/100 gm body weight)							
	Terminal Body Weight (kg)	Liver	Kidneys	Spleen	Testes	Adrenals	Caecum	Brain
Wool cloth (4 x 15 inches) treated with sodium tetra- borate applied to intact skin of 6 rabbits	3.3 +0.44	2.61 +0.34	0.55 +0.04	0.05 +0.02	0.20 +0.09	0.021 +0.006	4.08 +1.21	0.27 +0.12
Wool cloth (4 x 15 inches) treated with sodium tetra- borate applied with 2.0 ml sweat to abraded skin of 5 rabbits	3.9 +0.32	2.69 +0.49	0.53 +0.07	0.05 +0.03	0.19 +0.05	0.016 +0.005	2.89 +0.24	0.27 +0.05
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) HPC applied to intact skin of 6 rabbits	4.0 +0.45	2.64 +0.48	0.53 +0.05	0.05 +0.01	0.20 +0.04	0.014 +0.004	3.23 +0.57	0.25 +0.03
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) HPC applied with 2.0 ml sweat to abraded skin of 6 rabbits	3.0 +0.36	2.76 +0.35	0.55 +0.03	0.04 +0.08	0.21 +0.02	0.016 +0.003	3.80 +0.57	0.26 +0.04

APPENDIX II
TABLE 1 (Cont)
90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS
SUMMARY OF ORGAN-TO-BODY WEIGHT RATIOS

Type Application	Terminal body Weight (kg)	Organ-to-Body Weight Ratios (gm/100 gm body weight)						
		Liver	Kidneys	Spleen	Testes	Adrenals	Caecum	Brain
<u>CONTROLS</u>								
Nothing applied (cage con- trols) 8 rabbits	4.2	2.88	0.54	0.04	0.17	0.013	3.22	0.24
	± 0.73	± 0.47	± 0.08	± 0.03	± 0.04	± 0.003	± 0.74	± 0.05
Untreated wool cloth (4 x 15 inches) applied to intact skin of 9 rabbits	3.8	2.92	0.57	0.05	0.21	0.015	3.65	0.27
	± 0.37	± 0.50	± 0.09	± 0.03	± 0.05	± 0.005	± 0.66	± 0.04
Untreated wool cloth (4 x 15 inches) applied with 2.0 ml sweat to abraded skin of 9 rabbits	3.6	2.69	0.55	0.05	0.20	0.016	3.96	0.27
	± 0.57	± 0.43	± 0.07	± 0.01	± 0.04	± 0.004	± 0.68	± 0.03

APPENDIX H
TABLE 2
90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS
SUMMARY OF BODY WEIGHTS (KG)

Type Application	Mean Pretreatment Values	Treatment Period Values				
		Week 2	Week 4	Week 6	Week 8	Week 10 Week 12
Wool cloth (4 x 15 inches) treated with sodium tetra- borate applied to intact skin of 6 rabbits	3.5 +0.3	3.5 +0.3	3.4 +0.3	3.4 +0.3	3.3 +0.4	3.3 +0.4 3.3 +0.4
Wool cloth (4 x 15 inches) treated with sodium tetra- borate applied with 2.0 ml sweat to abraded skin of 5 rabbits	3.7 +0.3	3.6 +0.4	3.4 +0.5	3.7 +0.3	3.6 +0.5	3.7 +0.4 3.9 +0.3
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) HPC applied to intact skin of 6 rabbits	3.7 +0.2	3.7 +0.3	3.6 +0.4	3.6 +0.3	3.6 +0.4	3.8 +0.4 4.0 +0.4
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) HPC applied with 2.0 ml sweat to abraded skin of 6 rabbits	3.7 +0.1	3.6 +0.2	3.6 +0.2	3.7 +0.2	3.8 +0.2	3.6 +0.3 3.7 +0.3

APPENDIX II
TABLE 2 (Cont)
90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS
SUMMARY OF BODY WEIGHTS (KG)

Type Application	Mean		Treatment Period Values				
	Pretreatment Values	Week 2	Week 4	Week 6	Week 8	Week 10	Week 12
<u>CONTROLS</u>							
Nothing applied (cage controls) 8 rabbits	3.7 +0.5	4.1 +0.7	4.0 +0.7	4.2 +0.7	4.2 +0.7	4.3 +0.7	4.2 +0.7
Untreated wool cloth (4 x 15 inches) applied to intact skin of 9 rabbits	3.7 +0.3	3.8 +0.5	3.8 +0.5	3.8 +0.5	3.9 +0.5	3.8 +0.5	3.8 +0.4
Untreated wool cloth (4 x 15 inches) applied with 2.0 ml sweat to abraded skin of 9 rabbits	3.5 +0.4	3.5 +0.5	3.5 +0.5	3.6 +0.6	3.7 +0.5	3.6 +0.6	3.6 +0.6

APPENDIX I
TABLE I
30-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS
SUMMARY OF CLINICAL CHEMISTRY RESULTS

Type Application	Mean		Treatment Period Values					
	Pretreatment Values		Week 2	Week 4	Week 6	Week 8	Week 10	Week 12
Wool cloth (4 x 15 inches) treated with sodium tetraborate applied to intact skin of 6 rabbits	116 ± 49		140 ± 70	132 ± 58	134 ± 41	139 ± 36	111 ± 46	134 ± 20
Wool cloth (4 x 15 inches) treated with sodium tetraborate applied with 2.0 ml sweat to abraded skin of 5 rabbits	104 ± 42		119 ± 44	132 ± 45	150 ± 51	122 ± 44	133 ± 44	183 ± 70
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) HPC applied to intact skin of 6 rabbits	107 ± 42		138 ± 56	129 ± 55	113 ± 34	111 ± 42	99 ± 33	139 ± 52
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) HPC applied with 2.0 ml sweat to abraded skin of 6 rabbits	107 ± 35		173 ± 32	145 ± 52	118 ± 26	114 ± 21	106 ± 12	121 ± 39

APPENDIX I
TABLE 1 (Cont)
90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS
SUMMARY OF CLINICAL CHEMISTRY RESULTS

SERUM LACTIC DEHYDROGENASE							
Type Application	Mean Pretreatment Values	Treatment Period Values					
		Week 2	Week 4	Week 6	Week 8	Week 10	Week 12
<u>CONTROLS</u>							
Nothing applied (cage controls) 8 rabbits	98	122	120	106	118	107	132
	+36	+ 52	+ 49	+ 52	+ 41	+ 42	+ 36
Untreated wool cloth (4 x 15 inches) applied to intact skin of 9 rabbits	100	110	129	110	111	106	132
	+40	+ 45	+ 47	+129	+ 35	+ 26	+ 44
Untreated wool cloth (4 x 15 inches) applied with 2.0 ml sweat to abraded skin of 9 rabbits	110	116	158	117	122	109	159
	+46	+ 50	+ 36	+ 47	+ 44	+ 40	+ 60

APPENDIX I
TABLE 2
90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS
SUMMARY OF CLINICAL CHEMISTRY RESULTS

Type Application	Mean Pretreatment Values	Treatment Period Values				
		Week 2	Week 4	Week 6	Week 8	Week 10
Wool cloth (4 x 15 inches) treated with sodium tetra- borate applied to intact skin of 6 rabbits	114 + 24	88 +27	77 +20	110 + 13	117 + 27	120 + 7
Wool cloth (4 x 15 inches) treated with sodium tetra- borate applied with 2.0 ml sweat to abraded skin of 5 rabbits	122 + 21	99 +15	90 +27	118 + 5	115 + 13	122 + 5
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) IPC applied to intact skin of 6 rabbits	121 + 22	86 +23	93 +12	121 + 27	116 + 22	116 + 6
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) IPC applied with 2.0 ml sweat to abraded skin of 6 rabbits	120 + 19	82 +12	88 +36	116 + 8	109 + 13	112 + 15

APPENDIX I
TABLE 2 (Cont)
90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS
SUMMARY OF CLINICAL CHEMISTRY RESULTS

BLOOD GLUCOSE								
Type Application	Mean		Treatment Period Values					
	Pretreatment Values		Week 2	Week 4	Week 6	Week 8	Week 10	Week 12
<u>CONTROLS</u>								
Nothing applied (cage controls) 3 rabbits	131 + 25	109 + 23	109 + 17	126 + 15	119 + 12	127 + 16	107 + 16	
Untreated wool cloth (4 x 15 inches) applied to intact skin of 9 rabbits	122 + 25	101 + 23	90 + 19	118 + 14	113 + 9	115 + 8	105 + 15	
Untreated wool cloth (4 x 15 inches) applied with 2.0 ml sweat to abraded skin of 9 rabbits	118 + 23	96 + 20	83 + 17	121 + 21	111 + 6	112 + 9	99 + 23	

APPENDIX I
TABLE 3
90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS
SUMMARY OF CLINICAL CHEMISTRY RESULTS

Type Application	SERUM GLUTAMIC OXALOACETIC TRANSAMINASE										
	Pretreatment Values	Week 2	Week 4	Week 6	Week 8	Week 10	Week 12	Treatment Period Values			
Wool cloth (4 x 15 inches) treated with sodium tetraborate applied to intact skin of 6 rabbits	16.3 + 7.1	15.5 + 6.7	15.2 + 6.1	17.3 + 4.1	19.8 + 6.4	17.2 + 3.6	13.5 + 4.6				
Wool cloth (4 x 15 inches) treated with sodium tetraborate applied with 2.0 ml sweat to abraded skin of 5 rabbits	13.6 + 8.4	13.8 + 12.3	23.9 + 16.0	21.6 + 8.4	16.5 + 7.9	18.6 + 7.8	19.1 + 5.8				
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) IPC applied to intact skin of 6 rabbits	13.7 + 7.0	11.9 + 5.0	13.8 + 8.4	14.2 + 4.9	9.6 + 4.4	12.1 + 3.6	9.7 + 3.9				
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) IPC applied with 2.0 ml sweat to abraded skin of 6 rabbits	17.3 + 7.5	13.3 + 6.8	19.8 + 12.9	20.7 + 17.4	13.5 + 4.1	16.6 + 7.8	12.9 + 6.2				

APPENDIX 1
TABLE 3 (Cont)
90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS
SUMMARY OF CLINICAL CHEMISTRY RESULTS

Type Application	SERUM GLUTAMIC OXALOACETIC TRANSAMINASE						
	Pretreatment Values	Week 2	Week 4	Week 6	Week 8	Week 10	Week 12
<u>CONTROLS</u>							
Nothing applied (cage controls) 8 rabbits	13.8 ± 6.1	9.5 ± 4.0	11.2 ± 2.1	17.4 ± 6.6	15.7 ± 5.0	17.8 ± 5.7	15.6 ± 4.3
Untreated wool cloth (4 x 15 inches) applied to intact skin of 9 rabbits	13.6 ± 6.7	8.8 ± 5.1	14.0 ± 3.4	20.3 ± 9.9	16.2 ± 6.8	15.5 ± 3.1	13.9 ± 4.5
Untreated wool cloth (4 x 15 inches) applied with 2.0 ml sweat to abraded skin of 9 rabbits	16.5 ± 8.7	15.2 ± 10.1	19.6 ± 6.9	14.9 ± 5.3	14.1 ± 4.5	19.5 ± 5.0	18.0 ± 6.9

TABLE 4-117 Rel toxicity of Candidate Waterproofing Uniform Impregnant, Study No. 33-4-06/71, Nov 67-Dec 67

APPENDIX I
TABLE 4
90-DAY APPLICATION OF IMPREGNANT CLOTH TO SKIN OF RABBITS
SUMMARY OF CLINICAL CHEMISTRY RESULTS

Type Application	Pretreatment Values	BLOOD UREA NITROGEN					
		Week 2	Week 4	Week 6	Week 8	Week 10	Week 12
Wool cloth (4 x 15 inches) treated with sodium tetraborate applied to intact skin of 6 rabbits	20.8 ± 2.5	16.3 ± 3.4	20.2 ± 2.7	20.7 ± 1.8	19.5 ± 3.1	19.5 ± 4.0	16.6 ± 3.0
Wool cloth (4 x 15 inches) treated with sodium tetraborate applied with 2.0 ml sweat to abraded skin of 5 rabbits	22.4 ± 4.1	15.7 ± 5.5	24.0 ± 11.9	22.5 ± 3.1	23.1 ± 3.2	21.4 ± 4.4	16.6 ± 5.5
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) HPC applied to intact skin of 6 rabbits	21.0 ± 3.0	14.0 ± 1.2	17.5 ± 3.0	20.3 ± 3.5	20.9 ± 2.9	21.8 ± 3.3	18.0 ± 2.4
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) HPC applied with 2.0 ml sweat to abraded skin of 6 rabbits	20.2 ± 3.5	15.2 ± 2.7	18.8 ± 3.0	19.4 ± 3.5	21.3 ± 2.3	20.0 ± 1.9	15.1 ± 3.5

APPENDIX 1
TABLE 4 (Cont)
90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS
SUMMARY OF CLINICAL CHEMISTRY RESULTS

Type Application	BLOOD UREA NITROGEN					
	Mean Pretreatment Values	Week 2	Week 4	Week 6	Week 8	Week 10
<u>CONTROLS</u>						
Nothing applied (cage controls) 8 rabbits	21.0 ± 3.5	17.1 ± 4.5	18.0 ± 2.7	21.3 ± 3.6	17.4 ± 4.2	20.6 ± 2.5
Untreated wool cloth (4 x 15 inches) applied to intact skin of 9 rabbits	20.6 ± 3.3	17.2 ± 2.8	19.1 ± 2.1	21.6 ± 3.9	21.0 ± 3.2	21.4 ± 1.7
Untreated wool cloth (4 x 15 inches) applied with 2.1 ml sweat to abraded skin of 9 rabbits	20.9 ± 3.3	16.7 ± 2.5	19.3 ± 4.5	20.3 ± 2.9	20.3 ± 3.5	22.8 ± 3.4
						16.7 ± 2.1

TABLE 11. Oral toxicity of candidate tetraglycyl sodium hexametaphosphate, Study No. 33-4-6, 1971, on C7-10000

APPENDIX I
TABLE 5
90-DAY APPLICATION OF TETRAGLYCYL SODIUM HEXAMETAPHOSPHATE TO S.D. OF RABBITS
SUMMARY OF CLINICAL CHEMISTRY RESULTS

Type Application	Mean Pretreatment Values	TOTAL SERUM PROFILE				
		Week 1	Week 4	Week 6	Week 9	Week 12
Wool cloth (4 x 15 inches) treated with sodium tetra- borate applied to intact skin of 6 rabbits	7.1 +1.4	6.8 +0.3	7.0 +0.4	7.4 +0.5	7.5 +0.9	7.1 +0.5
Wool cloth (4 x 15 inches) treated with sodium tetra- borate applied with 2.0 ml sweat to abraded skin of 5 rabbits	6.7 +0.6	6.6 +0.5	7.0 +0.4	7.4 +0.4	7.4 +0.6	6.9 +0.4
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) IPC applied to intact skin of 6 rabbits	6.8 +0.7	6.7 +0.4	6.9 +0.5	7.5 +0.9	7.3 +0.4	6.8 +0.4
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) IPC applied with 2.0 ml sweat to abraded skin of 6 rabbits	6.7 +1.6	6.3 +0.6	6.9 +0.6	7.4 +0.9	7.1 +0.6	6.6 +0.5

CO. A-1. Rel toxicity of candidate isotherofing uniform impregnant, study no. 33-4-66/71, Nov 67-Dec 69

APPENDIX I
TABLE 5 (cont)
90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS
SUMMARY OF CLINICAL CHEMISTRY RESULTS

Type Application	TOTAL SERUM PROFILE						
	Pretreatment Values	Week 2	Week 4	Week 6	Week 8	Week 10	Week 12
<u>CONTROLS</u>							
Nothing applied (cage controls) 3 rabbits	6.7 ±0.6	7.0 ±0.4	6.8 ±0.2	7.5 ±0.3	7.6 ±0.5	7.0 ±0.5	6.7 ±0.4
Untreated wool cloth (4 x 15 inches) applied to intact skin of 9 rabbits	6.5 ±0.6	6.4 ±0.4	6.7 ±0.5	7.0 ±0.3	7.0 ±0.4	6.7 ±0.6	6.8 ±0.2
Untreated wool cloth (4 x 15 inches) applied with 2.0 ml sweat to abraded skin of 9 rabbits	6.6 ±0.6	6.6 ±0.6	6.8 ±0.4	7.0 ±0.3	7.0 ±0.4	6.8 ±0.4	6.7 ±0.3

APPENDIX I
TABLE 6
90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS
SUMMARY OF CLINICAL CHEMISTRY RESULTS

Type Application	SERUM AMYLASE		Treatment Period Values				
	Mean Pretreatment Values	Week 2	Week 4	Week 6	Week 8 *	Week 10	Week 12
Wool cloth (4 x 15 inches) treated with sodium tetra- borate applied to intact skin of 6 rabbits	402 + 90	432 + 60	498 + 129	498 + 54	-	360 + 36	354 + 36
Wool cloth (4 x 15 inches) treated with sodium tetra- borate applied with 2.0 ml sweat to abraded skin of 5 rabbits	390 + 108	386 + 216	378 + 54	444 + 42	-	378 + 66	366 + 96
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) HPC applied to intact skin of 6 rabbits	414 + 108	396 + 72	390 + 42	468 + 66	-	378 + 42	330 + 18
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) HPC applied with 2.0 ml sweat to abraded skin of 6 rabbits	372 + 102	408 + 102	444 + 96	450 + 72	-	336 + 54	330 + 54

* Specimens destroyed owing to refrigeration failure

APPENDIX 1
TABLE 6 (Cont)
90-LAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS
SUMMARY OF CLINICAL CHEMISTRY RESULTS

Type Application	Mean		Treatment Period Values				
	Pre-treatment Values	Week 2	Week 4	Week 6	Week 8	Week 10	Week 12
<u>CONTROLS</u>							
Nothing applied (cage controls) 8 rabbits	300 +114	390 +102	378 +90	438 +78	-	372 +60	360 +126
Untreated wool cloth (4 x 15 inches) applied to intact skin of 7 rabbits	390 +108	414 +78	390 +60	432 +60	-	384 +66	324 +36
Untreated wool cloth (4 x 15 inches) applied with 2.0 ml sweat to abraded skin of 7 rabbits	378 +106	450 +186	420 +108	432 +60	-	366 +72	342 +54

USARHA-MT Rel Toxicity of Candidate Mothproofing Uniform Impregnant,
Study No. 33-4-68/71, Nov 67-Dec 69

APPENDIX I
TABLE 7

90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS

BRAIN CHOLINESTERASE ACTIVITY	
Type Application	Brain Cholinesterase Activity
Wool cloth (4 x 15 inches) treated with sodium tetra- borate applied to intact skin of 6 rabbits	54.1 ± 7.6
Wool cloth (4 x 15 inches) treated with sodium tetra- borate applied with 2.0 ml sweat to abraded skin of 5 rabbits	68.5 ±18.6
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) HPC applied to intact skin of 6 rabbits	58.4 ±11.0
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) HPC applied with 2.0 ml sweat to abraded skin of 6 rabbits	59.5 ±20.5
<u>CONTROLS</u>	
Nothing applied (cage controls) 8 rabbits	69.0 ±12.6
Untreated wool cloth (4 x 15 inches) applied to intact skin of 9 rabbits	53.6 ±11.5
Untreated wool cloth (4 x 15 inches) applied with 2.0 ml sweat to abraded skin of 9 rabbits	64.1 ±16.9

WALSH-A-11 Rel toxicity of Candidate Noth-roofing Uniform Impregnant, Study No. 13-4-68/71, Nov 67-Dec 67

APPENDIX J
TABLE 1
90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS
SUMMARY OF HEMATOLOGY RESULTS

Type Application	Mean Pretreatment Values	HEMATOCRIT (%)				
		Week 2	Week 4	Week 6	Week 8	Week 10
Wool cloth (4 x 15 inches) treated with sodium tetra- borate applied to intact skin of 6 rabbits	42.3 + 2.4	43.5 + 3.2	41.3 + 2.9	41.6 + 2.5	40.7 + 3.9	40.3 + 2.7
Wool cloth (4 x 15 inches) treated with sodium tetra- borate applied with 2.0 ml sweat to abraded skin of 5 rabbits	40.8 + 2.7	39.7 + 4.7	38.5 + 2.7	39.8 + 2.6	36.6 + 2.6	39.0 + 1.9
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) HPC applied to intact skin of 6 rabbits	41.1 + 2.2	39.7 + 2.4	38.9 + 3.5	41.7 + 2.5	40.8 + 3.0	39.8 + 1.7
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) HPC applied with 2.0 ml sweat to abraded skin of 6 rabbits	43.2 + 3.1	42.4 + 3.4	42.3 + 2.5	43.6 + 1.8	41.9 + 2.2	42.5 + 2.8
						41.8 + 1.9

APPENDIX J
TABLE 1 (Cont)
90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS
SUMMARY OF HEMATOLOGY RESULTS

Type Application	HEMATOCRIT (%)		Treatment Period Values			
	Mean Pretreatment Values	Week 2	Week 4	Week 6	Week 8	Week 10
<u>CONTROLS</u>						
Nothing applied (cage controls) 8 rabbits	42.5 ± 2.8	43.9 ± 3.1	41.5 ± 2.9	43.0 ± 2.0	43.8 ± 2.9	43.1 ± 2.5
Untreated wool cloth (4 x 15 inches) applied to intact skin of 9 rabbits	42.2 ± 3.0	40.6 ± 1.6	40.1 ± 2.2	41.2 ± 1.9	40.2 ± 2.9	41.0 ± 2.2
Untreated wool cloth (4 x 15 inches) applied with 2.0 ml sweat to abraded skin of 9 rabbits	41.8 ± 3.2	42.4 ± 3.2	41.3 ± 2.4	41.7 ± 2.0	40.2 ± 2.3	41.0 ± 2.4
						42.6 ± 2.4
						41.0 ± 2.1
						40.9 ± 1.8

APPENDIX J
TABLE 2
90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS
SUMMARY OF HEMATOLOGY RESULTS

Type Application	Pretreatment Values	LEUKOCYTES /MM ³				
		Week 2	Week 4	Week 6	Week 8	Week 10
Wool cloth (4 x 15 inches) treated with sodium tetraborate applied to intact skin of 6 rabbits	7350 +1760	8100 +2800	8540 +1350	8440 +1500	9180 +2430	7540 +1840
Wool cloth (4 x 15 inches) treated with sodium tetraborate applied with 2.0 ml sweat to abraded skin of 6 rabbits	7680 +2490	8890 +2730	7450 +1560	8720 +1560	11820 +4880	8430 +1540
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) HPC applied to intact skin of 6 rabbits	7020 +1740	9040 +2840	8740 +1510	8270 +1760	7980 +2330	8150 +1730
Wool cloth (4 x 15 inches) impregnated with 0.5% (w/w) HPC applied with 2.0 ml sweat to abraded skin of 6 rabbits	8290 +1840	8900 +2320	8190 + 960	8150 +2120	7590 +4490	8460 + 960

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APPENDIX J
TABLE 2 (Cont)
90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS
SUMMARY OF HEMATOLOGY RESULTS

Type Application	LEUKOCYTES /MM ³						
	Mean Pretreatment Values	Treatment Period Values					Week 12
		Week 2	Week 4	Week 6	Week 8	Week 10	
<u>CONTROLS</u>							
Nothing applied (cage controls) 8 rabbits	7260 +2020	9220 +3420	8720 +1250	8390 +2080	9610 +2230	8700 +1760	8590 + 955
Untreated wool cloth (4 x 15 inches) applied to intact skin of 9 rabbits	8740 +2510	9175 +2120	8130 +2110	8310 +1530	7870 +2110	8000 +2170	9140 +1233
Untreated wool cloth (4 x 15 inches) applied with 2.0 ml sweat to abraded skin of 9 rabbits	7680 +2520	7550 +1560	7330 +1760	8090 +1840	7690 +1080	7690 +2540	8540 +2190

APPENDIX J
TABLE 3
90-DAY APPLICATION OF IMPREGNATED CLOTH TO SKIN OF RABBITS

Type Application	Mean Pretreatment Values	ERYTHROCYTES ($10^6/\text{mm}^3$)					
		Week 2	Week 4	Week 6	Week 8	Week 10	Week 12
cool cloth (4 x 15 inches) treated with sodium tetra- borate applied to intact skin of 6 rabbits	5.10 +0.62	4.50 +0.78	4.54 +0.52	5.27 +0.34	6.20 +1.20	5.00 +0.54	5.19 +0.53
cool cloth (4 x 15 inches) treated with sodium tetra- borate applied with 2.0 ml sweat to abraded skin of 5 rabbits	4.78 +0.60	4.57 +0.61	4.18 +0.53	4.61 +0.69	6.66 +1.72	5.33 +0.76	5.27 +0.63
cool cloth (4 x 15 inches) impregnated with 0.5% (w/w) IPC applied to intact skin of 6 rabbits	4.88 +0.80	4.69 +0.77	4.51 +0.75	5.13 +0.73	6.48 +1.35	5.36 +0.49	5.21 +0.34
cool cloth (4 x 15 inches) impregnated with 0.5% (w/w) IPC applied with 2.0 ml sweat to abraded skin of 6 rabbits	5.27 +0.62	4.80 +1.01	4.88 +0.75	5.35 +0.64	6.85 +1.13	5.67 +0.54	5.42 +0.51

LABORATORY Gel toxicity of (anhydrous) hydrochloric acid, Study No. 33-4-60/71, Nov 67-Dec 67

ADP, 100%
TABLE 3 (Cont)
90-100% (100% OF 100% GEL) TO SAL. OF RABBITS

Type Application	ERYTHROCYTES ($10^6/mm^3$)					
	Mean Retreatment Values	Treatment Period Values				
		Week 2	Week 4	Week 6	Week 8	Week 10
<u>CONTROLS</u>						
Nothing applied (cage controls) 8 rabbits	5.00 ±0.11	5.11 ±0.17	4.57 ±0.30	5.03 ±0.15	6.20 ±0.54	5.94 ±0.80
Untreated wool cloth (4 x 15 inches) applied to intact skin of 7 rabbits	5.01 ±1.11	5.01 ±0.43	5.36 ±0.46	5.11 ±0.62	6.23 ±0.61	5.67 ±0.17
Untreated wool cloth (4 x 15 inches) applied with 0.1 ml sweat to abraded skin of 7 rabbits	5.11 ±0.67	4.84 ±0.05	4.26 ±0.40	5.25 ±0.32	6.16 ±1.13	5.21 ±0.64

Quercus

* The Irritation scoring system found in Appendix C was used in evaluation of skin responses.

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13. ABSTRACT		
The relative toxicity of a candidate uniform mothproofing impregnant hexadecylpyridinium chloride was investigated using mice, rats, guinea pigs, rabbits and humans with the following findings: a. The technical grade compound and aqueous solutions of 1.1 percent concentration (w/v) or greater caused skin irritation in rabbits. b. Eye irritation in rabbits resulted from single 24-hour application of a 1.1 percent (w/v) aqueous solution but no irritation resulted from a 0.01 percent (w/v) solution. c. The skin irritation properties of hexadecylpyridinium chloride in guinea pigs prevented the determination of the sensitization potential of this compound in comparison with dinitrochlorobenzene. d. Chronic ingestion <u>ad libitum</u> of a diet containing 100 ppm hexadecylpyridinium chloride for 30 days caused minimal changes in rats. e. Diet impregnated with 0.1 percent (w/v) hexadecylpyridinium chloride and applied continuously to the shaved skin of rabbits for 30 days produced no remarkable degree of skin irritation or evidence of potential toxic hazard. f. A prophetic patch test on humans of wool fabric impregnated with hexadecylpyridinium chloride at a concentration of 0.1 percent by weight produced no evidence of skin irritation or sensitization.		

DD FORM 1473

REPLACES DD FORM 1473, 1 JAN 64, WHICH IS OBSOLETE FOR ARMY USE

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USAEHA-MT Rel Toxicity of Candidate Mothproofing Uniform Impregnant,
Study No. 33-4-68/71, Nov 67-Dec 69

Para 13, DD Form 1473 cont

g. Under the conditions of impregnation specified, HPC should not present a hazard when used as a mothproofing compound in wool fabric at a deposition of 0.5 percent (w/w). Since the weight of wool fabric to be used has not been specified it is impossible to estimate the degree of safety.

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14 KEY WORDS	LINK A		LINK B		LINK C	
	ROLE	WT	ROLE	WT	ROLE	WT
Hexadecylpyridinium chloride Quaternary ammonium compound Patch test Mothproofing Acute toxicity Cloth impregnant Eye irritant Irritant Subacute toxicity Feeding study Tear test Uniform Primary Irritation Study						

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