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Evaluation of Barrier Skin Cream Effectiveness Against JP-8 Jet Fuel Absorption and Irritation

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14. ABSTRACT

The DoD and Air Force uses large volumes of JP-8 annually. Due to the irritation produced by JP-8, the objective of this study was to obtain, formulate and experimentally select the most effective augmentation cream or lotion to inhibit skin exposure to liquid JP-8 during handling or after spills. Creams were tested first *in vitro* using cell diffusion chambers and then using the New Zealand White Rabbit model. Over-the-counter creams were first tested to evaluate the proof of concept of preventing dermal irritation with a barrier cream. Finally, formulated barrier creams were developed by two different companies and tested in-house. An effective barrier cream was not found using the rabbit model. The most effective protection against JP-8 remains personal protective equipment including gloves, and etc.

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PREFACE

This research was accomplished at the Applied Biotechnology Branch, Human Effectiveness Directorate of the 711th Human Performance Wing (711 HPW/RHPB) of the Air Force Research Laboratory, Wright-Patterson AFB, OH, under Dr. John J. Schlager, Branch Chief. This technical report was written for AFRL Work Unit 7184D408.

All studies involving animals were approved by the Wright-Patterson Institutional Animal Care and Use Committee, and were conducted in a facility accredited by the Association for the Assessment and Accreditation of Laboratory Animal Care, International, in accordance with the *Guide for the Care and Use of Laboratory Animals* (1996).

SUMMARY

This project was divided into 4 distinct stages, working from multiple *in vitro* techniques, allowing for the down-selection of barrier creams, to testing on shaved skin on the back of an *in vivo* rabbit model. An overarching goal of finding or developing a barrier cream to prevent dermal irritation caused by JP-8 was constant throughout each section. The first section of this project tested over-the-counter (OTC) creams in cell diffusion chambers covered by a Silastic® membrane. Some creams were able to impede the flow of JP-8 through the Silastic® membrane. The second section of the experiment tested OTC and formulated barrier creams in cell diffusion chambers covered with harvested pig skin. This stage provided a vehicle to narrow down which OTC and formulated creams would be tested *in vivo* using an animal model.

The *in vivo* sections of this experiment first tested the OTC and formulated barrier creams on the rabbit animal model in Section 3. The formulated barrier creams were provided by H&H Scientific Services, LLP and Skin Armor Technologies, LLP. Formulated creams showed a better ability to prevent dermal irritation than that of OTC creams. The final section of this experiment tested strictly formulated barrier creams from two companies. Formulated barrier creams did not provide the required amount of protection from dermal irritation in the animal model to justify a human test.

INTRODUCTION

Although the skin is generally considered a good barrier to prevent systemic absorption of most chemicals, many non-polar chemicals can partially breach the barrier and enter the skin. Toxicity at the chemical contact site is much more common than systemic toxicity due to the local absorption of chemical into the skin. According to U.S. Bureau of Labor Statistics, occupational skin diseases or disorders are the most common types of nonfatal occupational illnesses in the industry sector (Bureau of Labor Statistics, 2007). Mechanisms by which chemicals cause these visible effects differ from chemical to chemical. Solvent-like components in fuels have been implicated as the primary causes of irritant contact dermatitis (Elsner, 1994).

JP-8 exposure causes human skin irritation. JP-8 jet fuel has been reported to cause visible, measurable irritation when it comes in contact with unprotected rabbit skin (Kinkead *et al.*, 1992). Many OTC barrier creams claim to attenuate or completely prevent the penetration of irritating chemicals such as jet fuel. In this study, these barrier creams were first tested *in vitro* in cell diffusion chambers, and finally *in vivo* using the New Zealand White Rabbit.

JP-8 is a kerosene based fuel used primarily in aircraft turbine engines. It is composed of hundreds of compounds. JP-8 is used in Department of Defense (DoD) vehicles, cooking stoves, space heaters and generators. DoD Directive 4140.25 states that JP-8 will be the military's primary fuel. Kerosene, the parent fuel of JP-8, is blended from distilled petroleum fractional streams with boiling points between 160 and 270 degrees Celsius. Since various fractions from progressively increasing temperature hydrocarbon streams are blended to produce kerosene based on performance, not chemical content, this fuel can vary considerably in specific chemical amounts and types from batch to batch. The origin (geographical region) of the petroleum stock used in a fuel is critical to the makeup of the final, blended kerosene and can significantly influence the fractional components.

Since the fuel meets performance specifications and is not constrained to any particular chemical makeup, specific exposure toxicity of this complex mixture remains difficult to predict.

JP-8 is irritating to the skin and a weak skin sensitizer (Kinkead *et al.*, 1992; Kanikkannan *et al.*, 2000). Aromatic and aliphatic components of JP-8 can rapidly penetrate the skin (McDougal *et al.*, 2000), which can promote mild skin irritation, oxidative species formation, and deoxyribonucleic acid (DNA) damage (Kinkead *et al.*, 1992; Kanikkannan *et al.*, 2000; Kabbur *et al.*, 2001; Rogers *et al.*, 2001). For this reason alone, it is paramount to insure that the amount of both skin and systemic JP-8 exposure remain the lowest possible, based on required mission operation conditions. Currently the Air Force uses gloves, either the single use disposable chemical-resistant gloves, or the heavy-duty chemical-resistant gloves (military issue, 8415-01-013-7382) or a combination of both gloves at the same time. Protective clothing also includes cotton sweats, coveralls, cotton head-covers and face shield. Due to environmental conditions, such as hotter climates, protective clothing may be determined by the airman as undesirable to wear. In fact, they can pose a threat to the health of an individual due to heat injuries. This is why it is important to prove if a barrier cream can be used as an efficient temporally stable barrier, either alone or with the current personal protective equipment (PPE).

Cell Diffusion Chambers

It has been shown that cell diffusion chambers are capable of differentiating compounds of low permeability from those of high permeability and ranking compounds as to how they will perform *in vivo* (Franz, 1975). In Sections 1 and 2 of this experiment, Franz cells were used first with an artificial membrane (Silastic®) and then with harvested pig skin to measure the effectiveness of each tested barrier cream. Cell diffusion chambers were a key method for down selection of candidate barrier creams.

As close as the *in vitro* method simulated *in vivo* studies, there was still the need to examine the creams that performed the best *in vitro* on animal tissue that had both blood flow as well as an immune and local irritation response cell population. Otherwise, the assumption that no measurable penetration results in lack of irritation may not be correct. For example, with barrier cream applied to non-living skin, such as harvested pig skin, there is no way to measure how much JP-8 contacts the skin upon initial application. Harvested pig skin obtained using a dermatome, is non-living and consists of primarily epidermis. The epidermis, which is the outer layer of skin, lacks blood vessels and relies on the dermis for all nutrition and health. The dermis within the skin is the location of the nerves, blood vessels and glands. The dermis is also required for the physiological response to be stimulated, resulting in noticeable irritation. Without the living dermis to support the pig skin, there is no way to determine which creams are protecting against irritation or decreasing the response to JP-8. As the cell diffusion chambers may be able to rack and stack how well creams may perform against each other in occurrence or rates of penetration, they are not a substitute for *in vivo* testing.

In Vivo Model

The primary reason for the use of live animals is to determine the amount of irritation to living skin exposed to JP-8. This is the most sensitive and appropriate response to measure, due to irritation being the most common complaint for fuel handlers. The New Zealand White rabbit was chosen as the test animal for this study. Since New Zealand White rabbits have no pigment and have a relatively large dorsal surface area, dermally administered JP-8 to shaved skin can be accurately applied and resulting effects easily observed. The relatively large dorsal surface area allows for use of fewer animals, because more tests sites are available on each animal.

The New Zealand White rabbit has been shown to be sensitive to the irritant/corrosive effects of a variety of drugs and chemicals. For this reason, the rabbit is the preferred species for acute dermal irritation as directed by the Environmental Protection Agency (EPA), Health Effects Test Guidelines, Office of Prevention, Pesticides and Toxic Substances (OPPTS) 870.2500 (1998). Within the OPPTS 870.2500, the EPA recommends that dermal irritation should be scored and recorded according to Draize evaluation of skin irritation (Draize, 1959). Sections 3 and 4 of this project examined OTC barrier creams and formulated barrier creams using this animal model.

Colorimetry

The objective determination of skin color as a measure of irritation is crucial in dermatology (Neumann *et al.*, 1991; Fang *et al.*, 1997). Visual methods of dermal irritation are subjective and require multiple trained judges. A non-invasive method for color determination that eliminates the subjectivity of visual scoring is possible with the use of colorimetry. Only one investigator is needed to evaluate each exposure site, and multiple measurements can be taken and averaged together (Fang *et al.*, 1997, Chan and Lin Wan Po, 1992).

The CIE (Commission Internationale de l'Eclairage) system was used to quantify the colorimeter measurements. This system uses spectral chromaticity coordinates and corresponding color-matching functions based on trichromatic color matching of spectral lights, which is a function of their wavelengths from 380 to 780 nm (Broadbent, 2004).

METHODS

Section 1. Assessment of Barrier Creams against Penetration of JP-8 using Silastic® Membrane

Commercially acquired skin creams were first tested for solubility in JP-8. If soluble, no further testing was performed in the static cell. Eighteen skin protection creams were tested in a 0.02" thick silicone elastomer-covered diffusion cell. The static diffusion cell consisted of a donor cell on top and a receptor cell on bottom (Figure 1). A magnetic stirring bar was placed in the glass receptor cell and the receptor cell was filled with a Volpo saline solution in physiological saline. A cream coated Silastic® membrane (Dow Corning Corporation, Midland, MI) was placed on top of the receptor cell flange. Carefully, to exclude air, a glass donor cell was placed on the membrane and the flanges were clamped. The water-jacketed receptor cell was equilibrated at 37 degrees Celsius.

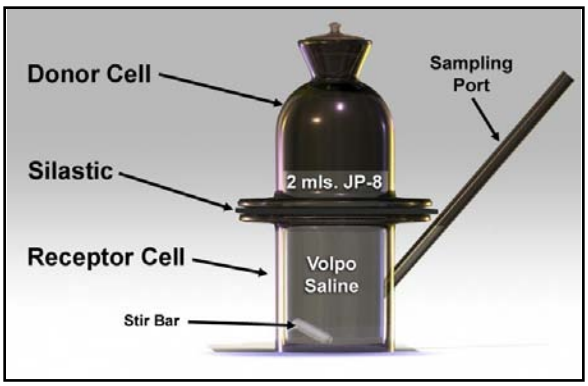


Figure 1. Cell diffusion chamber

A candidate cream was applied using a 0.10 mm thick plastic sheet with a 42 mm diameter hole. The hole in the sheet was placed over the Silastic® membrane and the cream was evenly applied using a stainless steel spatula. The surface of some skin creams beaded during application and drying; a cotton gauze sheet was placed on the Silastic® membrane before using the plastic sheet to coat the skin cream. The cotton gauze increased the coating thickness to 0.22 mm. All coatings were used both immediately (wet) and dried for 1 hour at approximately 33 degrees Celsius.

JP-8 (2 mL) was added to the donor cell to start the penetration run. The donor cell was capped with a ground glass stopper. JP-8 components that penetrated the skin cream and the Silastic® membrane then dissolved in the Volpo saline, which aided in solubilizing the JP-8 components as they penetrated into the receptor cell. Receptor solution was stirred continuously to evenly distribute the dissolved components. Samples of Volpo saline solution were taken every 15 minutes with 20 μL disposable pipettes. Samples were delivered into 21.5 mL headspace vials and were quickly capped. Tests were conducted for 4 hours and each material was run at least in triplicate.

Gas Chromatography

JP-8 vapor injection from the headspace vials were handled by a Tekmar 7000/7050 headspace sampler (Teledyne Tekmar, Mason, OH). The samples were first equilibrated at 140 degrees Celsius to volatilize the JP-8 components. Vapor samples were injected through a 1.0 mL sample loop, focused on the head of the column, and then flash heated to drive them into the Varian (Varian Inc., Palo Alto, CA) 3400 Gas Chromatograph. Vapor fuel components were separated on a nonpolar 0.53 mm x 30 m SPB-1 column which was programmed to 50 degrees Celsius for 5 minutes and then 5 Celsius degrees/minute to 185 degrees Celsius. The flame ionization detector (FID) temperature was 260 degrees Celsius. The FID detector output was integrated and printed out with EZChrom Elite version 3.0 software (Agilent Technologies, Inc., Santa Clara, CA). The total integrated peak time sample area minus the total integrated control peak area at time zero gave the total JP-8 elution amount. This FID response area was compared to areas of JP-8 standards prepared and analyzed in headspace vials containing Volpo saline (control) to give the amount of JP-8.

The average penetration rate was calculated using the total integrated peak area less control as described above, compared to integrated peak areas of JP-8 standards. This gave the amount of JP-8 that penetrated the Silastic® membrane over time. Percent inhibition was calculated by comparing the average penetration rate of each cream to its perspective control sample as in equation 1.

$$\%\, Inhibition = 100 - \left(\frac{Avg\ \ Penetration\ \ \ Rate\ (Sample\ )}{Average\ \ Penetration\ \ \ Rate\ (Control\ )}\right) \times 100 \qquad (1)$$

Section 2. *In Vitro* Assessment of Skin Barrier Creams against JP-8 Penetration

Of the 18 creams in Section 1, 15 previously tested creams, 2 OTC creams not previously tested, and 5 novel creams (Table 1) were then further tested by static diffusion cell using 0.6 mm harvested pig skin. The novel creams were formulated by H&H Scientific, LLC specifically for this experiment. The procedure for the static diffusion cell described in Section 1 was followed except for extra precautions required for pig skin preparations. The pig skin was harvested using a dermatome set at a thickness of 0.6 mm and the skin was either used immediately or frozen until use. JP-8 penetration rates using fresh or frozen uncoated skin samples did not differ significantly.

To apply the barrier cream on the pig skin, the hole in a plastic sheet template was placed over the pig skin and the skin cream was applied evenly with a spatula. After application, the cream was massaged into the skin until absorbed and the excess was removed with a metal spatula to ensure complete and consistent coverage of the skin.

Samples were placed onto the static cell (in place of the Silastic® membrane in Figure 1) and the cell was permitted to equilibrate for 45 minutes. JP-8 (2 mL) was then added to the donor cell to start the penetration run. The donor cell was capped with a ground glass stopper. Samples of the Volpo saline solution were taken with a 20 μL disposable pipette at the equilibrium time of the static cell (zero hour) and subsequent samples were taken at 1, 2, 3 and 4 hours. Tests were run for 4 hours and each cream was run in triplicate.

Table 1: OTC creams tested *in vitro*

Skin Cream	Manufacturer	City
Chiamal Skin Shield	Chim Tech, Inc	Missoula, MT
Derma Shield	Benchmark Commercial, Inc.	Salt Lake City, UT
Eterna Skin Guard	Eterna Health USA	Hamilton, NJ
Eucerin	Conair, Inc	Rantoul, Il
Fomblin RT15	Ausimont Viale Lombardia	Italy
MAN-O	MAN-O Products	Cincinnati, OH
Novel	H&H Scientific Services	Boerne, TX
Oxyfresh	Oyfresh, Inc	Coeur d'Alene, ID
Penetone 411	Penetone	Tenefly, NJ
Ply No. 9	The Milburn Company	Detroit, MI
Pr 88	Pan Tec, Inc	Guelph, ON, Canada
Pr 99	Pan Tec, Inc	Guelph, ON, Canada
Proguard	Decon Labs, Inc	Bryn Mawr, PA
Prolin Skin Guard	Plantolin Australia Pty. LTD	Seaford, Australia
SERPACWA	U.S. Army	
Skin-So-Soft	Avon Products, Inc	New York, NY
StokoDerm	Stockhausen, Inc	Greensboro, NC
Novel Creams	H&H Scientific, LLC	Bergheim, TX

Note: SERPACWA = Skin Exposure Reduction Paste Against Chemical Warfare Agents

Collected, capped samples were heated (140 degrees Celsius) to stable vapor phase using a headspace sampler and components separated on a non-polar SPB-1 column with FID detection similar to section 1. Total area of eluted hydrocarbon vapor from the sample was compared between the coated and non-coated pig skin 4-hour penetration runs. After the 4-hour penetration run, the pig skin was wiped with water and paper towels. Then skin punch samples were taken with a dermal biopsy punch and placed in 20 mL headspace sample vials for gas chromatography analysis to determine relative skin JP-8 absorption content.

Section 3. Evaluation of Skin Barrier Creams Effect on JP-8 Irritation in New Zealand White Rabbits (*Oryctolagus cuniculus*)

This section of the study evaluated skin barrier creams that can be used to limit skin irritation caused by JP-8 exposure in New Zealand White Rabbits. This study utilized the U.S. EPA Health Effects Test Guidelines, Office of Prevention, Pesticides and Toxic Substances 870.2500, Acute Dermal Irritation (EPA, 1998), with minor modifications to accommodate the different type of application procedure needed to study the barrier cream protection of skin. Based on previous *in vitro* studies with JP-8 penetration and skin absorption of residual JP-8, the top 5 creams were evaluated during this portion of the study and were ranked as the most to least effective barriers to JP-8. Healthy adult rabbits (n = 4) were used to evaluate each barrier cream, with sufficient sites and positive and negative controls on each individual rabbit to ensure statistical significance.

Animal Exposure Method

The back of the rabbit was carefully shaved from mid-scapula to iliac crest 24 hours prior to exposure to allow resolution of any minor clipper abrasion. Commercially available Hill Top Chambers (Hill Top Research, Miamiville, OH) were used to apply the JP-8 to the back of the rabbit. Teflon holders were made to support the 4 Hill Top Chambers on each side of the rabbit spine. Each site was randomly chosen so individuals grading the performance of each cream were blind to the contents of each site. This also ensured that the different degrees of sensitivity on the back of the rabbit were utilized randomly. The sites shown in Table 2 were randomized on the backs of each rabbit.

Table 2: Exposure sites on rabbit back (prior to random rotation)

Test Site	Description	JP-8 Dose (mL)	Exposure Time (hours)
I	Negative Control-- HTC	0.0	4.0
II	Positive Control--HTC/JP-8	0.5	4.0
III	Positive Control--HTC/JP-8	0.5	4.0
IV	Positive Control--HTC/JP-8	0.5	4.0
V	HTC/Cream/JP-8	0.5	4.0
VI	HTC/Cream/JP-8	0.5	4.0
VII	HTC/Cream/JP-8	0.5	4.0
VIII	Cream Control—HTC/Cream	0.0	4.0
IX	Nothing	0.0	4.0
X	Nothing	0.0	4.0
XI	Nothing	0.0	4.0

Note: HTC = Hill Top Chamber

Before beginning the exposure, each rabbit was anesthetized using Ketamine HCl (30-45 mg/kg) and Xylazine (3.0-7.0 gm/kg). All injections were made intramuscularly in the caudal thigh or lumbar muscles and the rabbit was monitored throughout the procedure. Once anesthetized, a baseline visual and colorimeter measurement was taken to assess any preexisting conditions. The candidate barrier cream was applied to the randomized sites. There is not any way to standardize the exact amount needed for each cream, because each has a unique consistency and covers the skin differently. In real world applications, the amount of cream would not be standardized but would be enough to cover an exposed area. To be as standardized as possible, the tip of a stainless steel spatula was used to apply similar amounts of each cream. The cream was then massaged evenly over the selected site and any excess was removed. After the application of the cream, 0.5 mL of JP-8 was pipetted onto the randomized Hill Top Chamber held by the Teflon holder as described above. The Teflon holder was then taped to the back of the rabbit and wrapped in Vetwrap. The JP-8 was left on the back of the rabbit for 4 hours.

Once the 4 hours was complete, the Teflon holder and the Hill Top Chambers were removed from the back of the rabbit. Each site was carefully wiped with water and gauze to remove any excess cream and JP-8. Each site was then scored after 40 minutes to satisfy the 30-60 minute interval per the OPTTS 870.2500 Guidelines. Subsequent scoring was accomplished at 24, 48 and 72 hours after exposure (Draize, 1959). After each daily scoring was finished, the rabbit

was re-shaved and permitted to rest for 24 hours before the next reading due to the rapid re-growth of fur.

Visual Scoring Technique

All barrier creams were scored in 3 ways; by visual scoring described in the Draize method, by colorimeter, and by histopathology.

Each site was first assigned an erythema and edema score for each of the 4 evaluation periods (40 minutes, 24, 48 and 72 hours). The scores were made by the same technicians at the same time. The scores for erythema and edema were totaled for each rabbit to give the primary irritation index (PII) shown in Equation 2 (Draize, 1959). The scale used for scoring each site is shown in Table 3.

$$PII = \frac{(Sum\ of\ eryt\,hema\ scores + Sum\ of\ edema\ scores)}{(\#of\ sites\ per\ rabbit * \#\ of\ rabbits\ per\ cream * \#\ of\ obervation\ periods)} \quad (2)$$

Table 3: Scale for scoring primary skin irritants[a]

Skin Reaction		Value
Erythema and eschar formation		
	No eythema	0
	Very slight erythema (barely perceptible)	1
	Well-defined erythema	2
	Moderate to severe erythema	3
	Severe erythema (beet redness) to slight eschar formations (injuries in depth)	4
Edema Formation		
	No edema	0
	Very slight edema (barely perceptible)	1
	Slight edema (edges of area well defined by definite raising)	2
	Moderate edema (raised approximately 1 mm)	3
	Severe edema (raised more than 1 mm and extending beyond the area of exposure)	4

[a]Draize et al. (1959).

Colorimery

After the visual score was assigned to each site, a colorimeter (Chroma Meter-CR 400, Minolta, Japan) was used to assess the color change in the skin. The instrument displays three-dimensional color reflectance including 'L*', 'a*' and 'b*' (Wu *et al.*, 2001). The luminance 'L*' gives the relative brightness from black (0) to white (100). The 'a*' describes the equilibrium between red (100) and green (-100) and the 'b*' describes the equilibrium between yellow (100) and blue (-100) (Pierard and Pierard-Franchimont, 1993; Wu *et al.*, 2001). The change in chroma (ΔC) and difference in color (ΔE) between the initial reading at time zero and each

respective time was calculated using Equations 3 and 4 (Westerhof *et al.*, 1986; Fang *et al.*, 1997).

$$\Delta C = \sqrt{(\Delta a * + \Delta b *)} \quad (3)$$

$$\Delta E = \sqrt{(\Delta L * + \Delta a * + \Delta b *)} \quad (4)$$

An average of 3 (n=3) readings were taken for each site. This was accomplished for each evaluation period as described with the visual scoring.

Animals

New Zealand White Rabbits (age 6 – 18 months) were used for the *in vivo* barrier cream irritation testing. Rabbits were housed individually with each lot segregated from all other rabbits for a 14 day quarantine/acclimation period. All animals were provided husbandry conditions consistent with the "Guide for the Care and Use of Laboratory Animals" (NRC, 1996). Room air temperature and humidity was maintained between 61-72 degrees Fahrenheit and 30-70 percent humidity. Lighting was adjusted to a 12:12 hour light:dark cycle. Rabbit chow and fresh conditioned (RO water) was made available *ad libitum*. All rabbits were observed daily for signs of distress and observations were recorded by the husbandry staff. Rabbits were provided with bunny blocks and timothy cubes for enrichment. Once the 72 hour measurement was completed, each rabbit was euthanized using sodium pentobarbital or other available euthanasia solution. Each test site was then collected and analyzed for histopathology.

Histopathology

Formalin fixed, paraffin embedded, 5 micron, hematoxylin and eosin stained sections of skin were submitted for histopathology evaluation. Sections were read by a Veterinary Pathologist blinded to treatment group. The histopathology diagnoses are listed on individual animal report forms by their respective accession numbers. Sections varied from normal (coded as 0) to showing varying degrees of a lesion compatible with a chemical burn or surface irritant lesion. Sections were graded for increasing severity of changes in both the epidermis and dermis using the criteria in Table 4.

Table 4: Histopathology grading criteria

Score	Description
0	Essentially normal tissue
1	Multifocal epidermal degeneration/necrosis, no dermal lesion
2	Diffuse or focally extensive epidermal degeneration/necrosis, no dermal lesion
3	Epidermal necrosis with collagen degeneration/necrosis in very superficial papillary dermis
4	Epidermal necrosis with collagen degeneration/necrosis through depth of papillary dermis (still considered superficial)
4+	Slightly more severe dermal lesion than 4
H	Denotes areas of minimal to mildly hyperplastic epithelium present, possibly indicating a regenerative or adaptive response.

Section 4. Evaluation of Formulated Barrier Skin Creams Effect on JP-8 Penetration in New Zealand White Rabbits (*Ortyctolagus cuniculus)*

The method described in Section 3 was followed with a few exceptions. Due to the consistency of the positive controls and to achieve more test sites, the number of positive control sites was decreased to 1 per rabbit, as shown in Table 5. H&H Scientific, LLC became Qwanah, LLC between Sections 3 and 4.

Table 5: Exposure rotation for formulated barrier creams

Test Site	Description	JP-8 Dose (mL)	Exposure Time (hours)
I	Negative Control--HTC	0.0	4.0
II	Positive Control--HTC/JP-8	0.5	4.0
III	HTC/Cream/JP-8	0.5	4.0
IV	HTC/Cream/JP-8	0.5	4.0
V	HTC/Cream/JP-8	0.5	4.0
VI	HTC/Cream/JP-8	0.5	4.0
VII	HTC/Cream/JP-8	0.5	4.0
VIII	Cream Control—HTC/Cream	0.0	4.0
IX	Nothing	0.0	4.0
X	Nothing	0.0	4.0
XI	Nothing	0.0	4.0

Time Trial Method

Skin Armor Cream 2 was selected to undergo shorter durations of JP-8 exposure to examine if a formulated cream could work for shorter periods of time. This cream was selected because it visually displayed the best protection from JP-8 erythema. Different durations of JP-8 exposure (0.25, 0.5, 0.75, 1, 1.25, 1.5, 1.75 and 2 hours) were tested on 4 rabbits. For logistical purposes, each rabbit had 2 different durations of exposure. An example exposure rotation for a rabbit is shown in Table 6.

Table 6: Exposure rotation for short durations of exposure

Test Site	Description	JP-8 Dose (mL)	Exposure Time (hours)
I	Positive Control—JP-8	0.5	0.25
II	HTC/Cream/JP-8	0.5	0.25
III	Positive Control—JP-8	0.5	0.5
IV	HTC/Cream/JP-8	0.5	0.5
V	Positive Control—JP-8	0.5	0.25
VI	HTC/Cream/JP-8	0.5	0.25
VII	Positive Control—JP-8	0.5	0.5
VIII	HTC/Cream/JP-8	0.0	0.5
IX	Nothing	0.0	0.5
X	Nothing	0.0	0.5
XI	Nothing	0.0	0.5

RESULTS

Section 1. Assessment of OTC Barrier Creams against the Penetration of JP-8 using Silastic® Membrane

Commercial OTC creams ranged from not effective (16 percent) to very effective (99 percent) at impeding JP-8 penetration. Rates of penetration were generally linear but a number of cream formulations had an asymptotic increase in penetration. Table 7 below shows the average penetration rate of the JP-8 through the barrier cream and Silastic® membrane. Penetration was inhibited most by the following barrier creams: SERPACWA, PLY #9, Fromblin and Oxyfresh. These creams inhibited penetration an order of magnitude better than the other barrier creams tested.

Table 7: JP-8 Penetration rate on Silastic® membrane

Barrier Cream	Approximate Thickness (mm)	Condition	Average Penetration Rate (µg JP-8/ hour) ± S.D.	Penetration Rate ∗ Thickness (µg/hour ∗ mm)
Control (Cotton Gauze)			4820 ± 203	
SERPACWA	**0.10**	**wet**	**291 ± 10**	**29**
SERPACWA	**0.10**	**dry**	**215 ± 65**	**22**
Eucerin	**0.10**	**wet**	**3620 ± 178**	**362**
Eucerin	0.10	dry	3997 ± 71	400
Equate	0.10	wet	4395 ± 157	440
Vanicream	0.10	wet	3313 ± 655	331
Vanicream	0.10	dry	3322 ± 1145	332
Skin-So-Soft	0.10	wet	3396 ± 1318	340
Skin-So-Soft	**0.10**	**dry**	**1895 ± 395**	**189**
Skin-So-Soft Ivy Block	0.10	dry	3096 ± 247	464
PR88	0.10	wet	4768 ± 223	477
PR88	0.10	dry	4591 ± 108	459
Ply # 9	**0.22**	**wet**	**108 ± 177**	**24**
Ply # 9	**0.10**	**dry**	**2688 ± 148**	**269**
Aloe	0.22	wet	4081 ± 716	898
Aloe	0.10	dry	4105 ± 579	411
Eterna Skin Guard	**0.22**	**wet**	**447 ± 423**	**98**
Eterna Skin Guard	0.10	dry	4666 ± 206	467
Stokoderm	**0.22**	**wet**	**1405 ± 780**	**309**
Stokoderm	**0.10**	**dry**	**3381 ± 205**	**338**
Derma Shield	0.22	wet	4842 ± 179	1065
Derma Shield	0.10	dry	4621 ± 158	462
Chimal	**0.22**	**wet**	**2001 ± 983**	**440**
Chimal	0.10	dry	4309 ± 84	431
Prolin Skin Guard	**0.22**	**wet**	**1170 ± 1030**	**257**
Prolin Skin Guard	**0.10**	**dry**	**4365 ± 353**	**437**
Fomblin	**0.22**	**wet**	**45 ± 10**	**10**
Fomblin	**0.10**	**dry**	**102 ± 24**	**10**
Penetone 411	**0.22**	**wet**	**3539 ± 503**	**779**
Penetone 411	**0.10**	**dry**	**3739 ± 156**	**374**
Man-O	**0.22**	**wet**	**3429 ± 563**	**754**
Man-O	0.10	dry	2718 ± 1176	272
Oxyfresh	**0.22**	**wet**	**209 ± 279**	**46**
Oxyfresh	0.10	dry	4373 ± 218	437

Notes: Bold text highlights those creams with a significant difference ($p \leq 0.5$) between penetration rate with coated material compared with uncoated Silastic® membrane (n=3). The 0.22 mm thinkness represents gauze used in this experiment. S.D. = standard deviation

Table 8 shows the highest performing barrier creams that were selected for further testing *in vitro* on harvested pig skin.

Table 8: Best performing OTC barrier creams for Silastic membrane (n=3)

Barrier Cream	Approximate Thickness (mm)	Condition	Average Penetration Rate (µg JP-8/hour) ± S.D.	Percent Inhibition of Penetration
Fomblin	0.22	wet	45 ± 10	99%
Fomblin	0.10	dry	102 ± 24	98%
Ply # 9	0.22	wet	107 ± 177	98%
Oxyfresh	0.22	wet	209 ± 279	95%
SERPACWA	0.10	dry	215 ± 65	95%
SERPACWA	0.10	wet	291 ± 10	93%
Eterna Skin Guard	0.22	wet	447 ± 423	91%
Prolin Skin Guard	0.22	wet	1170 ± 1030	76%
Stokoderm	0.22	wet	1405 ± 780	71%
Skin-So-Soft	0.10	dry	1895 ± 395	57%
Chimal	0.22	wet	2001 ± 983	58%
Ply # 9	0.10	dry	2688 ± 148	40%
Man-O	0.10	dry	2718 ± 1176	39%
Vanicream	0.10	wet	3313 ± 655	25%
Vanicream	0.10	dry	3322 ± 1145	25%
Stokoderm	0.10	dry	3381 ± 205	24%
Skin-So-Soft	0.10	wet	3397 ± 1318	24%
Man-O	0.22	wet	3429 ± 563	29%
Penetone 411	0.22	wet	3540 ± 503	27%
Eucerin	0.10	wet	3620 ± 178	19%
Penetone 411	0.10	dry	3739 ± 156	16%

JP-8 penetration through the barrier cream and Silastic® artificial membrane was linear in nature, as shown by the examples in Figures 2-5 compared to the control. In each case, the control was shown to contain a greater amount of JP-8 in the Volpo saline than that of the candidate barrier cream.

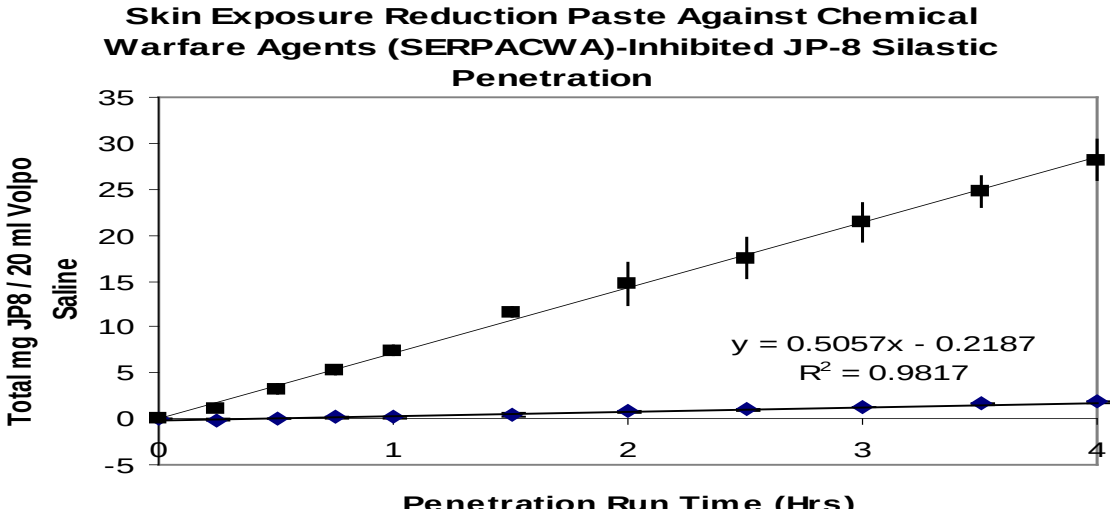


Figure 2. Penetration plot of SERPACWA (diamond) compared to control (square)

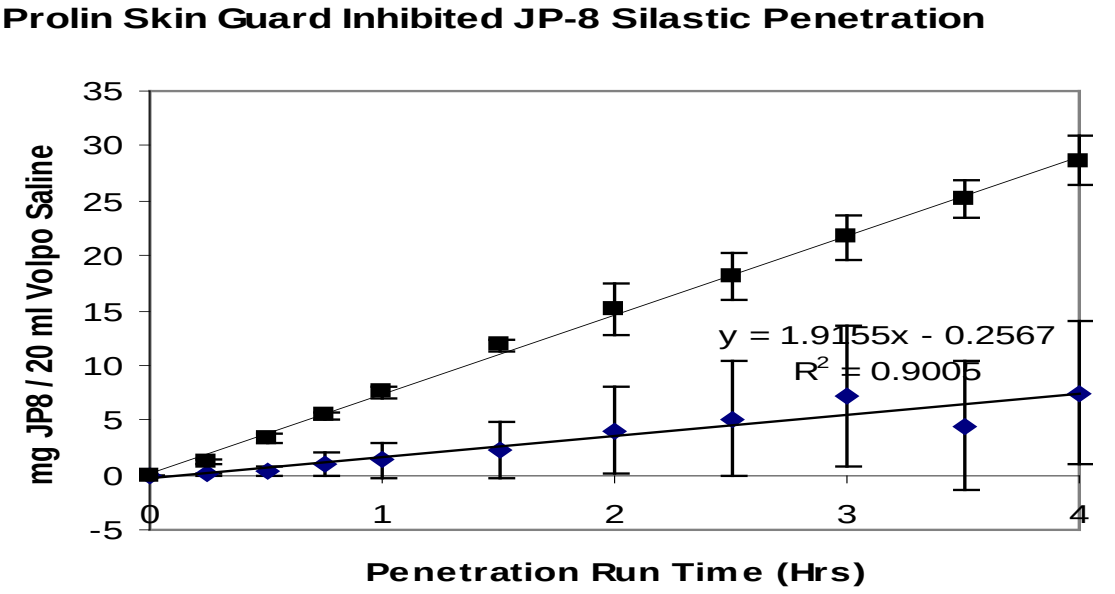


Figure 3. JP-8 penetration plot of Prolin Skin Guard (diamond) compared to control (square)

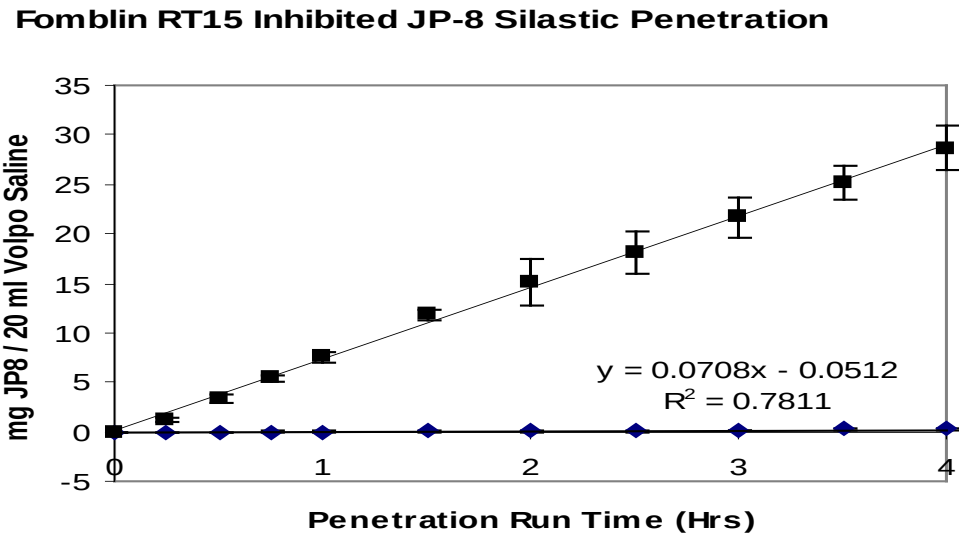


Figure 4. JP-8 penetration plot of Fomblin RT15 (diamond) compared to control (square)

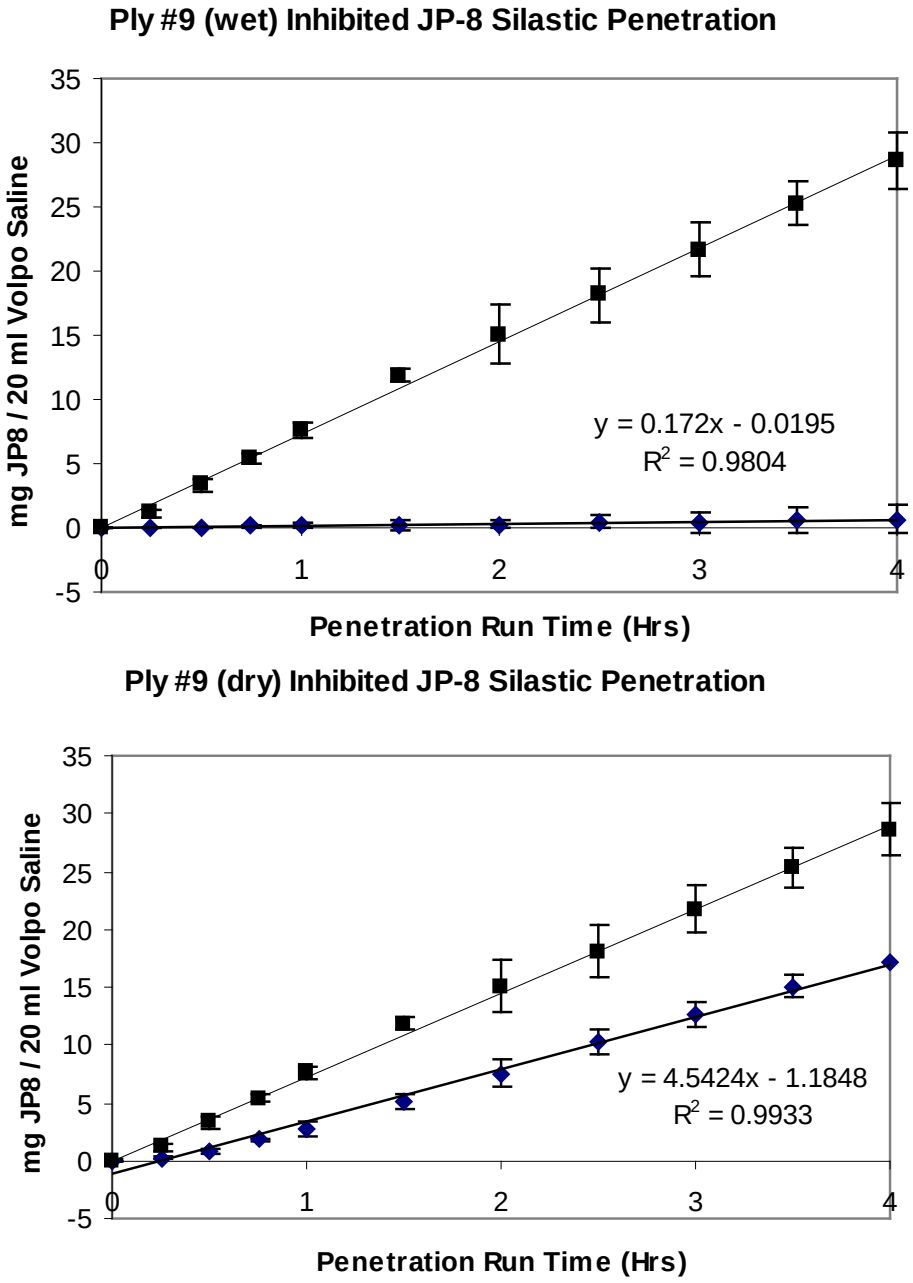


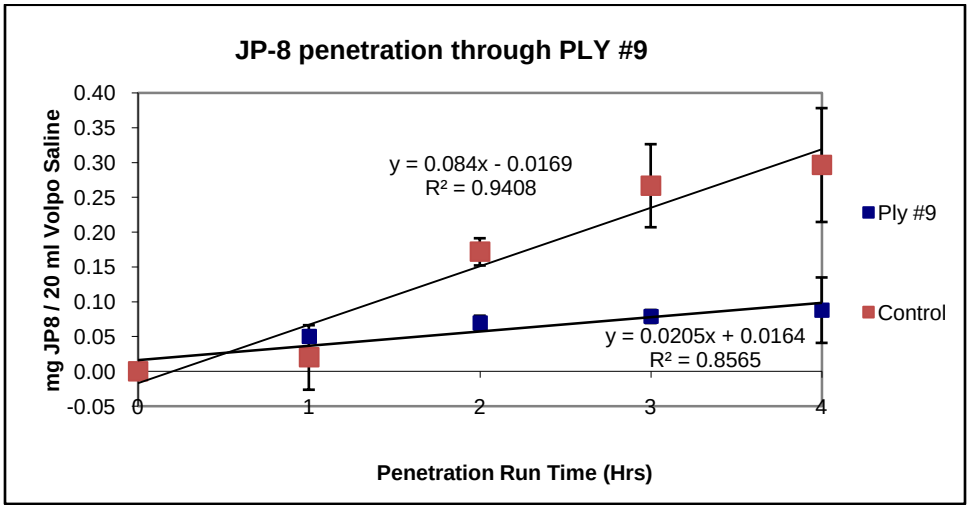
Figure 5: JP-8 penetration plot of PLY #9 (diamond) compared to control (square). Ply #9 was left wet (top) and allowed to dry (bottom).

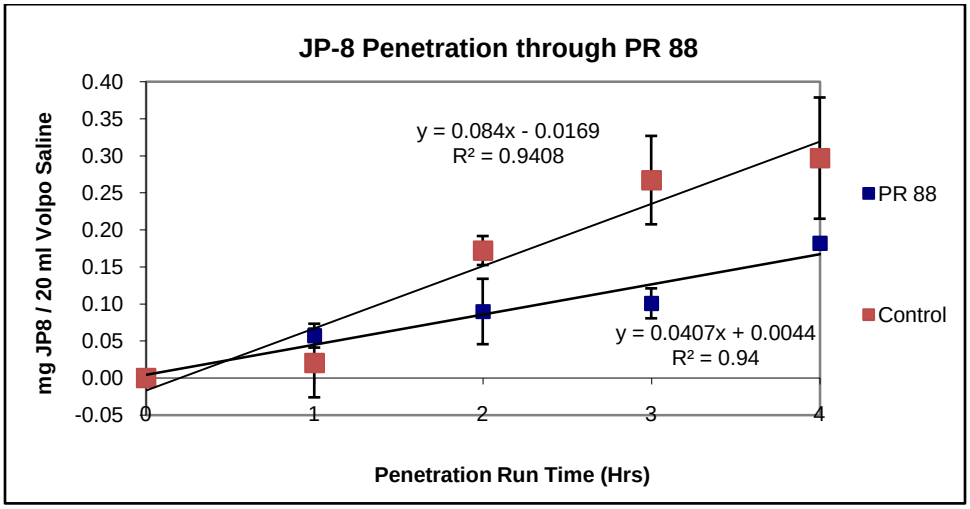
Section 2. *In Vitro* Assessment of Skin Barrier Creams on Harvested Pig Skin

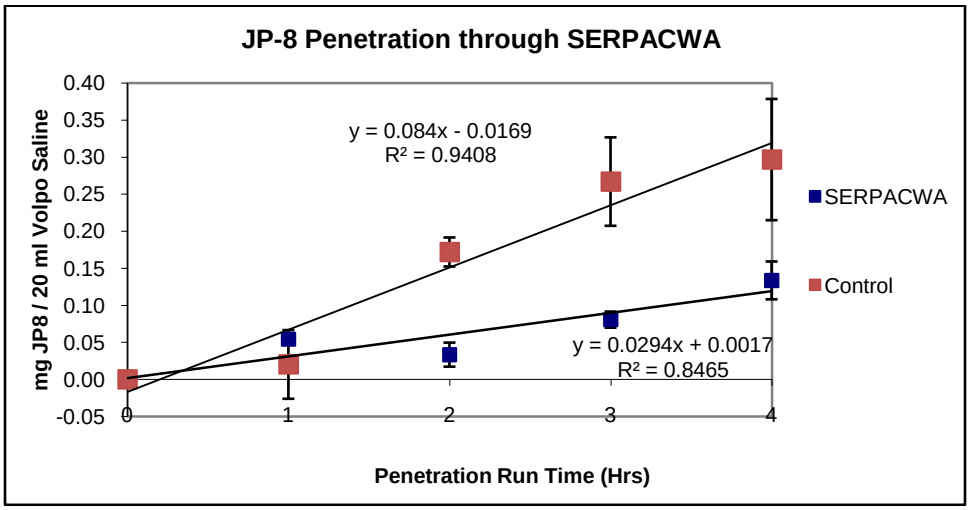
Following Section 1, each barrier cream was studied in a *in vitro* skin tissue exposure where harvested pig skin was used. Table 9 shows the overall results of the creams tested *in vitro* on harvested pig skin. Diffusion rates of JP-8 penetration through the best performing OTC barrier creams compared to the control are shown in Figure 6. OTC barrier creams displayed similar barrier properties on harvested pig skin as on the artificial membrane. Overall, the harvested pig skin was far less permeable than the artificial membrane. The best barrier creams in this stage of the project continued to be SERPACWA, PR-88, PLY-9 and 2 of the formulated skin creams by H&H scientific. These 5 creams were therefore selected to proceed to the *in vivo* stage of the project.

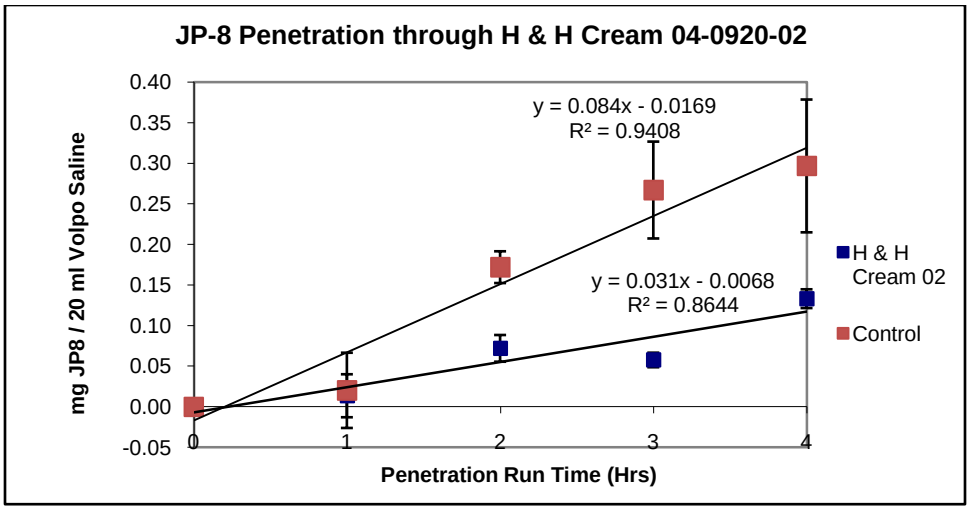
Table 9: JP-8 penetration through pig skin *in vitro*

	Total JP-8 remaining in and on the skin(mg JP-8/g skin)		Total JP-8 remaining in and on the skin (mg JP-8/cm^2 skin)		Total JP-8 hourly penetration rate (μg JP-8/hour/cm^2)	
Barrier Cream	**Average**	**S.D.**	**Average**	**S.D.**	**Average**	**S.D.**
JP-8 Control 1	2.53	0.17	110.00	4.00	8.13	5.72
JP-8 Control 2	2.41	0.35	143.00	8.00	5.94	1.00
Oxyfresh	3.20	0.84	127.00	31.00	6.11	3.79
Prolin Skin Guard	1.60	0.34	74.00	15.00	8.25	1.72
Chimal	2.57	1.53	99.00	64.00	7.36	4.03
Ply No. 9	0.84	0.47	45.00	24.00	0.98	1.70
MAN-O	1.00	0.21	52.00	9.00	7.46	1.21
StokoDerm	2.61	0.26	134.00	12.00	2.53	1.51
Proguard	4.44	0.59	172.00	2.00	7.71	2.14
Fomblin	3.15	1.52	100.00	68.00	3.94	2.77
Eterna Skin Guard	0.54	0.22	28.00	2.00	4.49	2.96
Derma Shield	2.79	2.06	95.00	89.00	24.59	9.21
Eucerin	2.16	0.29	93.00	22.00	13.77	6.43
Vanicream	1.01	0.16	50.00	3.00	4.73	0.96
pr99	1.68	0.23	98.00	12.00	5.67	0.91
Penetone	2.25	0.26	113.00	17.00	4.12	1.24
Skin-So-Soft	2.16	0.40	107.00	24.00	1.89	1.68
pr88	0.35	0.09	20.00	3.00	3.51	0.43
SERPACWA	0.41	0.14	21.00	6.00	1.65	1.65
Novel Lot# 04-0920-01	2.60	0.95	168.00	36.00	11.73	3.12
Novel Lot# 04-0920-02	1.10	0.52	39.00	7.00	1.53	0.81
Novel Lot# 04-0920-03	4.93	0.69	124.00	51.00	10.57	4.88
Novel Lot# 04-0920-04	2.65	0.86	155.00	11.00	4.55	2.82
Novel Lot# 04-0920-05	1.04	0.34	67.00	13.00	2.94	2.83


JP-8 penetration through PLY #9
mg JP8 / 20 ml Volpo Saline
0.40
0.35
0.30
0.25
0.20
0.15
0.10
0.05
0.00
-0.05
0
1
2
3
4
y = 0.084x - 0.0169
R² = 0.9408
y = 0.0205x + 0.0164
R² = 0.8565
Ply #9
Control
Penetration Run Time (Hrs)


JP-8 Penetration through PR 88
mg JP8 / 20 ml Volpo Saline
0.40
0.35
0.30
0.25
0.20
0.15
0.10
0.05
0.00
-0.05
0
1
2
3
4
y = 0.084x - 0.0169
R² = 0.9408
y = 0.0407x + 0.0044
R² = 0.94
PR 88
Control
Penetration Run Time (Hrs)


JP-8 Penetration through SERPACWA
mg JP8 / 20 ml Volpo Saline
0.40
0.35
0.30
0.25
0.20
0.15
0.10
0.05
0.00
-0.05
0
1
2
3
4
y = 0.084x - 0.0169
R² = 0.9408
y = 0.0294x + 0.0017
R² = 0.8465
SERPACWA
Control
Penetration Run Time (Hrs)



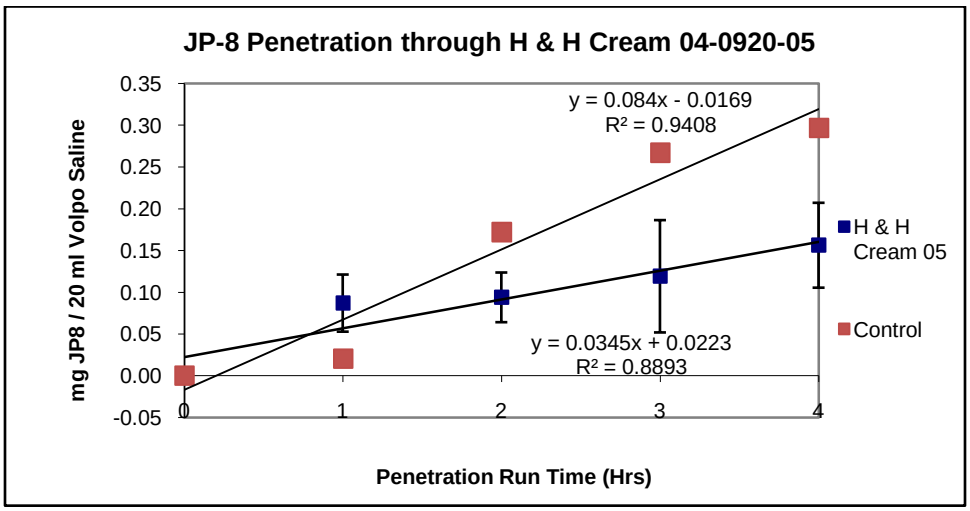


Figure 6: Rates of JP-8 penetration through barrier cream coated pig skin (n=3)

Section 3. Evaluation of Skin Barrier Creams effect on JP-8 Irritation in New Zealand White Rabbits (*Oryctolagus cuniculus*)

As mentioned previously, dermal irritation of each site was measured in 3 distinct ways. Each site was first graded using the Draize visual scoring method, then by a colorimeter, and finally by histopathology. The best performing creams that were down selected from the previous experiments were H&H Creams 04-0920-02 and 04-0920-05, SERPACWA, PLY #9, and PR-88.

Visual Score

Table 10 shows the calculated primary irritation index from 3 sites per rabbit, 4 time periods and 4 rabbits (with the exception of PLY-9, which was only tested on 3 rabbits). Each positive control is specific to the group of rabbits on which the barrier cream was tested. This avoided the possibility of each rabbit's skin reacting to JP-8 in a different manner. The negative control and the cream control did not display any irritation score on the primary irritation index.

Table 10: Primary Irritation Index comparison between the positive control (JP-8) and barrier cream coated sites

Cream	Primary Irritation Index	Grade
Positive Control*	1.60 ± 0.04	Slight Irritation
H&H cream 04-0920-02	1.32 ± 0.01	Slight Irritation
Positive Control	1.83 ± 0.06	Slight Irritation
H&H cream 04-0920-05	1.34 ± 0.11	Slight Irritation
Positive Data	1.91 ± 0.13	Slight Irritation
SERPACWA	1.71 ± 0.00	Slight Irritation
Positive Data	1.79 ± 0.06	Slight Irritation
PR 88	1.60 ± 0.13	Slight Irritation
Positive Data	1.82 ± 0.09	Slight Irritation
PLY-9	1.83 ± 0.11	Slight Irritation

* The positive control is calculated from the same rabbit on which each cream was tested.

The formulated barrier creams (H&H creams) showed a greater ability than the OTC creams to lessen the visual erythema caused by JP-8 in four hours. This result is supported by Figures 7-11 in which the average visual Draize score is plotted against time. The H&H creams showed a greater differentiation from the positive control in erythema over time. On the poor end of performance, PLY-9 displayed a greater primary irritation index than that of the positive control. These data are supported by Figure 10 in which the erythema score of PLY-9 closely mirrors that of the positive control over time.

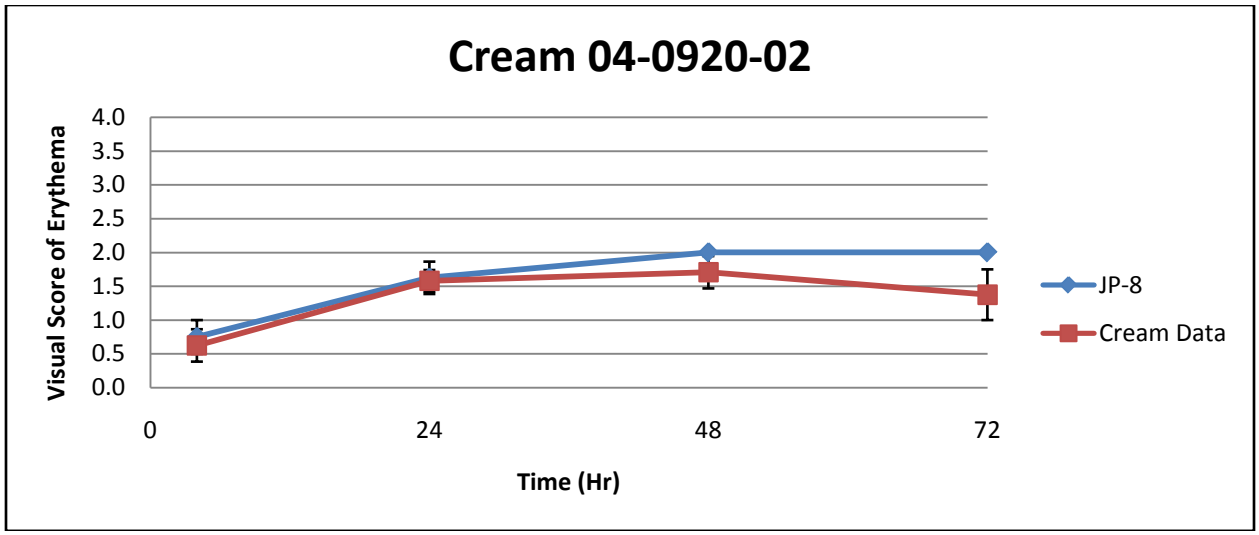


Figure 7: Average visual Draize score over time for sites coated with H&H Cream 04-0920-02

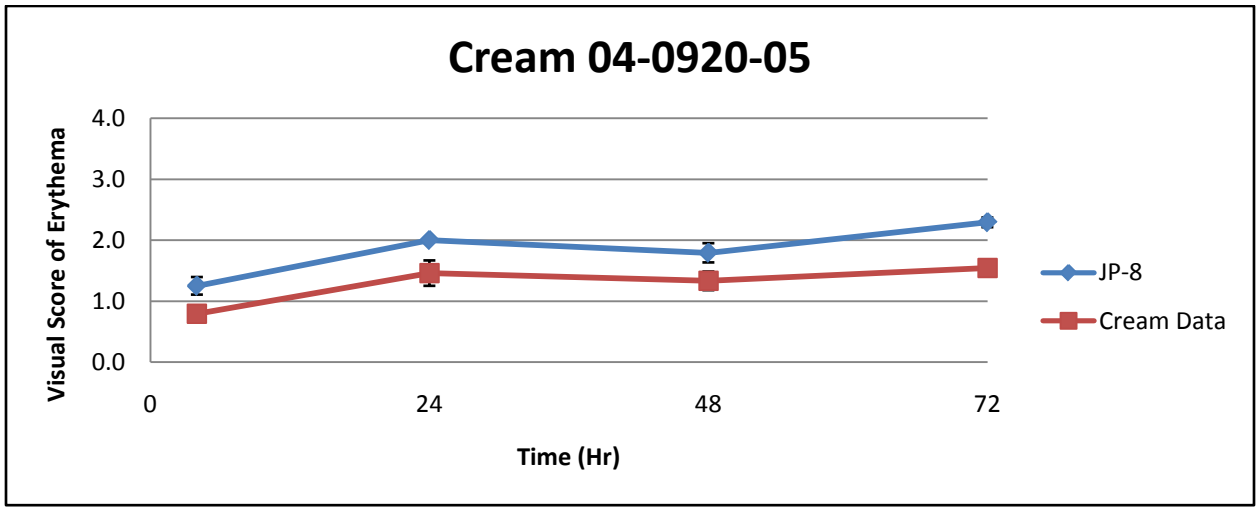


Figure 8: Average visual Draize score over time for sites coated with H&H Cream 04-0920-05

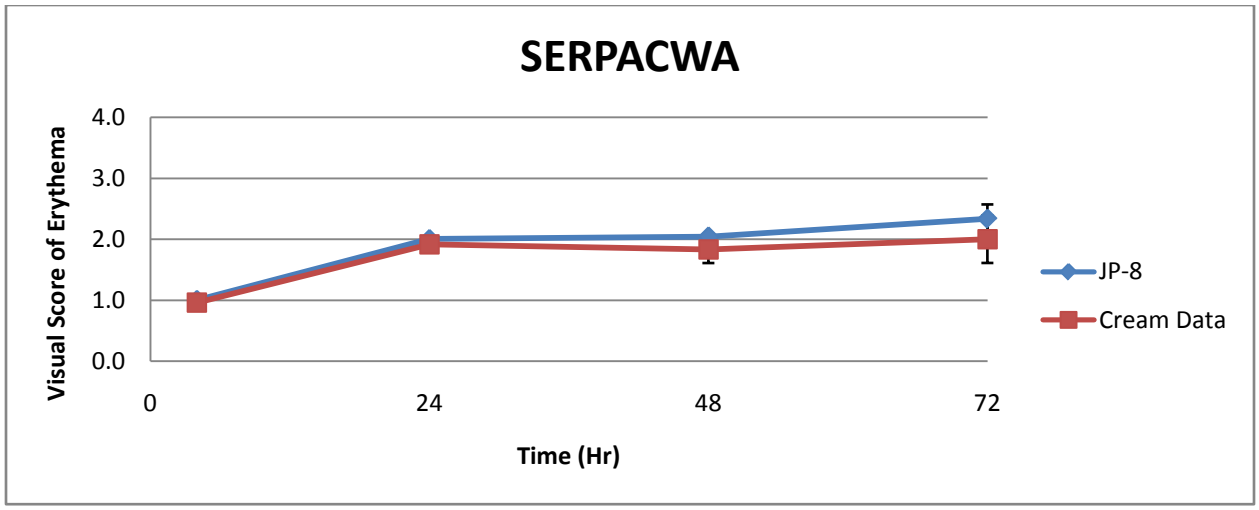


Figure 9: Average visual Draize score over time for sites coated with SERPACWA

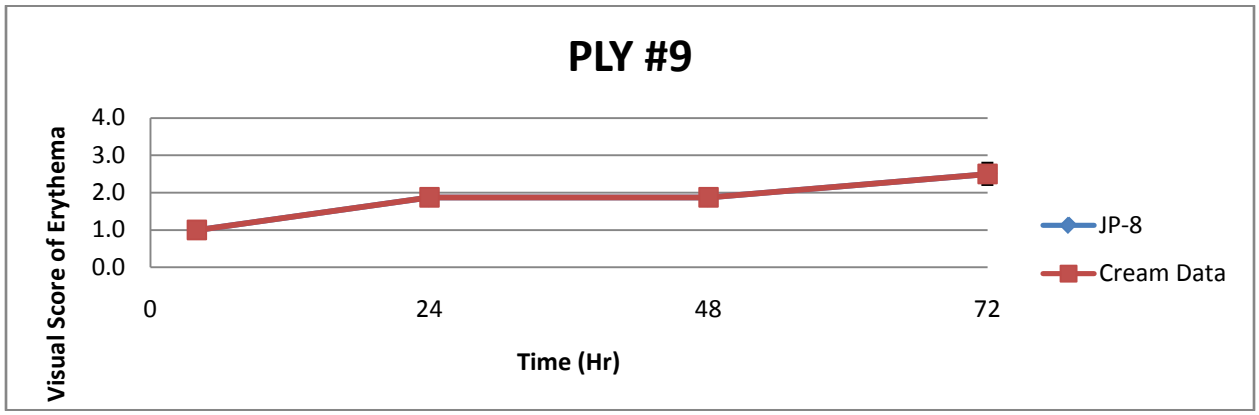


Figure 10: Average visual Draize score over time for sites coated with PLY #9

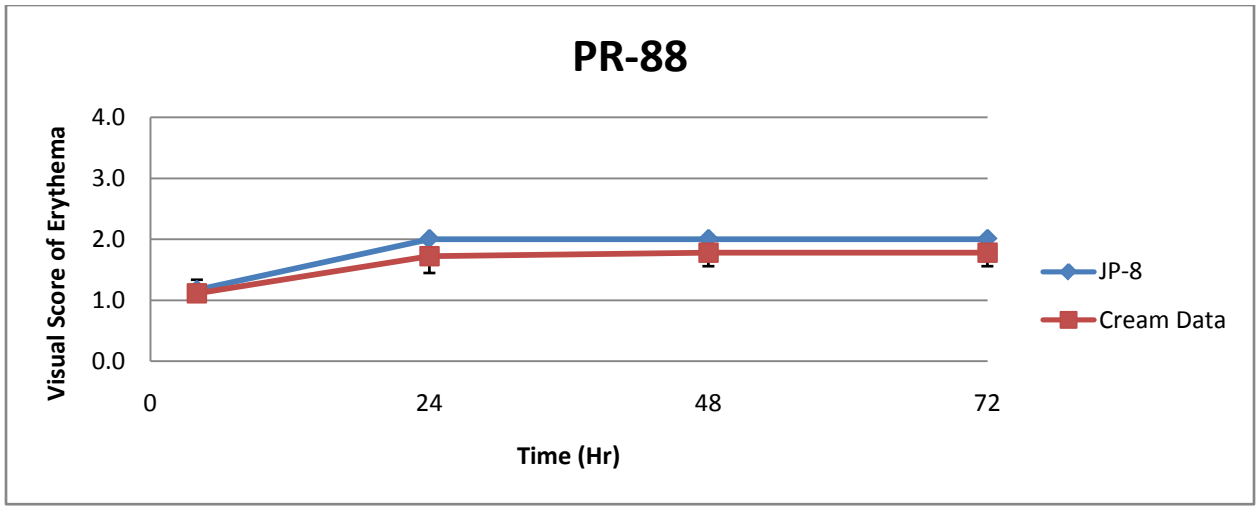


Figure 11: Average visual Draize score over time for sites coated with PR-88

Colorimeter

Using the Minolta colorimeter, the change in chroma (ΔC) and the change in overall color (ΔE) were plotted against the positive control over time (Figures 12 & 13). A Grubbs' test for outliers was completed on each set of data. Since the colorimeter is more objective than the human eye, the positive controls for all the rabbits in this section of the experiment were averaged and compared to the average of each cream. While the positive control's chroma and color continually increased through the experiment, Qwanah Creams 02 and 05, SERPACWA and PR-88 did not. Even though the Qwanah creams, SERPACWA and PR-88 did not show an increasing difference from 4 to 72 hours, there was a constant chroma and color difference from the pre-experiment reading. This difference indicates that although the redness did not continually increase over time, the redness was elevated from the pre-experiment reading.

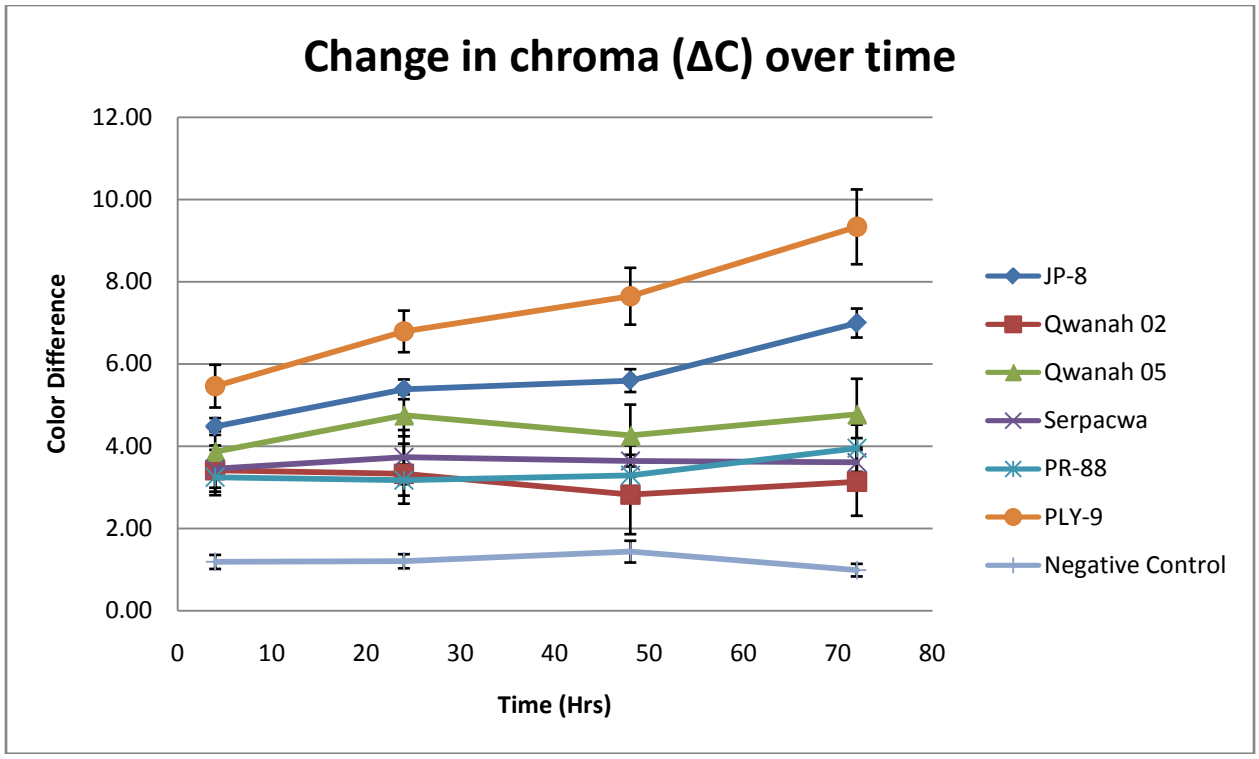


Figure 12: Change in chroma between initial reading and listed observation time

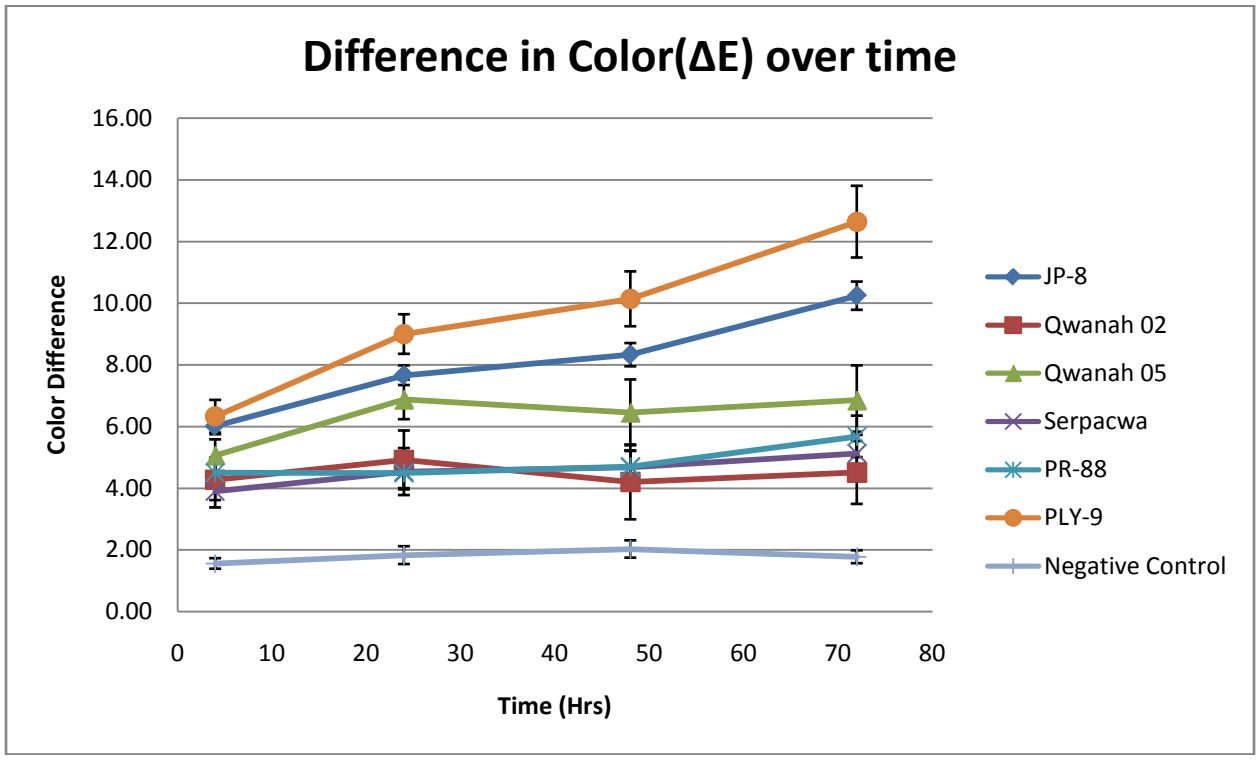


Figure 13: Difference in color between initial reading and listed observation time

Histopathology

Skin samples were taken and analyzed for damage and inflammation from each of the sites tested. Table 11 shows the average histology score received for each group of rabbits. The top performing creams proved to be the formulated barrier creams provided by H&H Scientific. Notably, the positive control histopathology scores were consistent across all the rabbits exposed in this experiment. Slides from a selected exposure site that most closely gives an accurate picture of the cellular damage are shown in Figures 14 through 19.

Table 11: Average Histopathology Scores

Barrier Cream Tested	Cream / HTC Sites	HTC Only Sites	Cream / HTC / JP-8 Sites	Positive Control Sites (JP-8 / HTC)
H&H Cream 04-0920-02	0 ± 0	0 ± 0	0.50 ± 0.19	2.83 ± 0.11
H&H Cream 04-0920-05	0 ± 0	0 ± 0	1.50 ± 0.26	2.67 ± 0.22
SERPACWA	0.25 ± 0.25	0 ± 0	1.58 ± 0.34	2.67 ± 0.14
PR-88	0 ± 0	0 ± 0	2.56 ± 0.29	2.67 ± 0.24
PLY-9	0 ± 0	0 ± 0	2.58 ± 0.19	2.75 ± 0.13

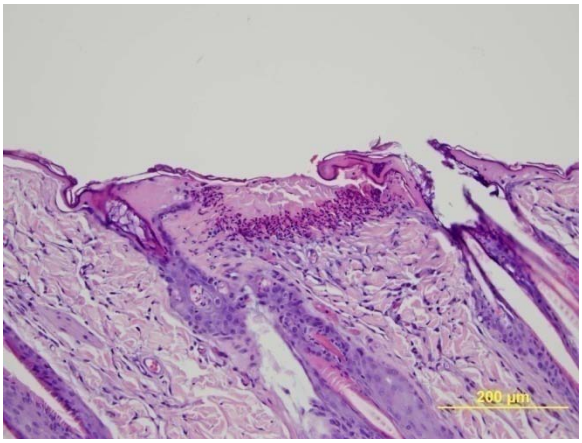


Figure 14. Clipped haired skin, positive control site (JP-8/HTC); rabbit 109-05. Focally extensive area of ulceration with overlying serocellular crust and mild collagen degeneration beneath the lesion; lesion severity score 3

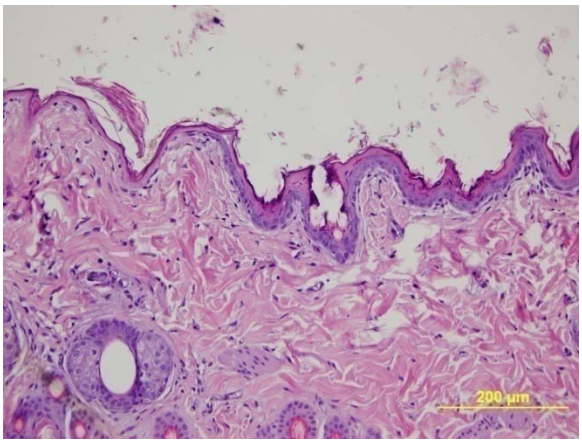


Figure 15. Clipped haired skin, rabbit 180-05 protected with H&H Scientific Barrier Cream Lot # 04-0920-02. Essentially normal skin section; lesion severity score 0

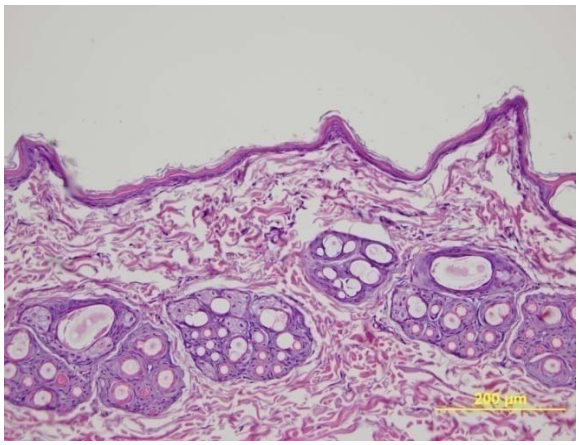


Figure 16. Clipped haired skin, rabbit 142-05 protected with H&H Scientific Barrier Cream Lot # 04-0920-05. Multifocal distribution of epidermal degeneration interrupted by areas of mild epidermal hyperplasia; lesion severity score 1H

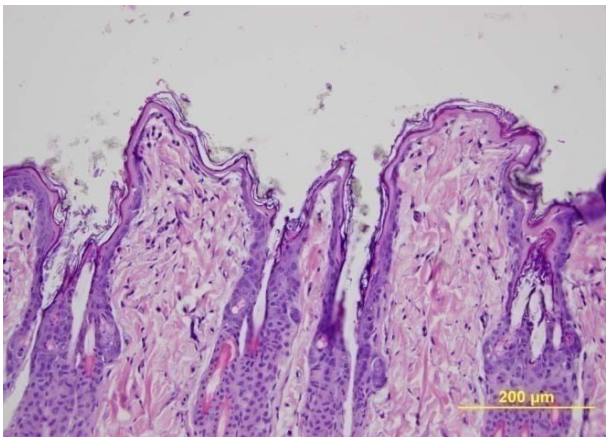


Figure 17: Clipped haired skin, rabbit 109-05 protected with SERPACWA barrier cream.
Multifocal epidermal degeneration with no lesion in the deeper dermis lesion; lesion severity score 1

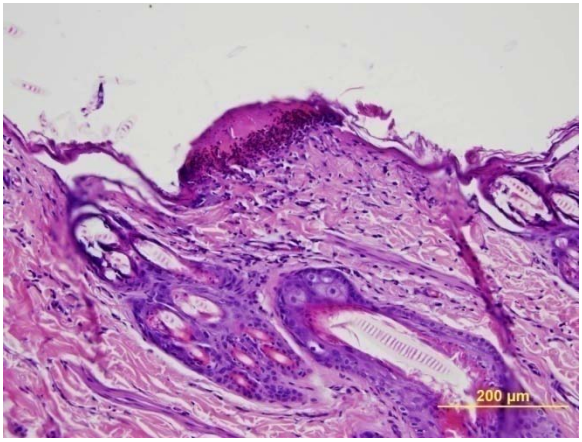


Figure 18: Clipped haired skin, rabbit 140-05 protected with Ply Number 9 barrier cream.
Focal ulceration with attached serocellular crust and mild collagen degeneration beneath the lesion; lesion severity score 3

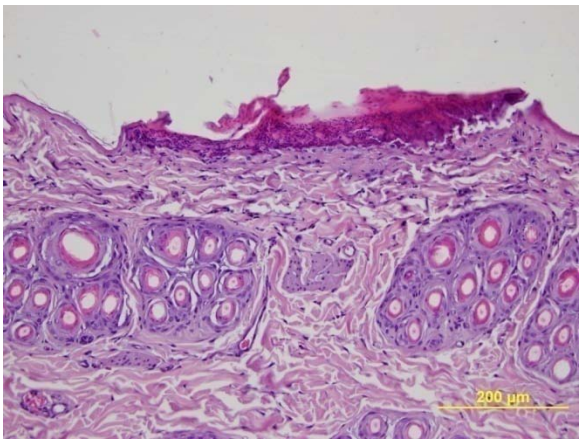


Figure 19: Clipped haired skin, rabbit 315-05 protected with pr 88 barrier cream. Focally extensive area of ulceration with detaching serocellular crust and mild collagen degeneration beneath lesion; lesion severity score 3

Section 4. Evaluation of Formulated Barrier Skin Creams to Prevent JP-8 Irritation in New Zealand White Rabbits (*Ortyctolagus cuniculus*)

As in Section 3 of this project, each test site was evaluated in 3 different ways. Each site was visually interpreted, then read by a colorimeter and finally processed for histopathology results. The creams tested in Section 4 were only formulated creams made specifically for this project. The exposure time that JP-8 remained in contact with the shaved skin was initially four hours, we then examined shorter exposures ranging from 30 min to two hours.

Visual Score

Formulated barrier creams from two different companies were tested. The H&H creams provided were from the same lot as those tested in Section 3. However, new samples of each cream were obtained because the old samples deteriorated over time. Table 12 shows the calculated primary irritation index for each cream tested in this section. The best performing formulated barrier cream by visual standards was H&H cream 08-0416-01. Unfortunately, none of the creams provided enough protection to lower the grade. In the case of H&H cream 08-0614-02, the cream seemed to facilitate irritation to the grade of mild irritation.

Table 12: Primary Irritation Index comparison between the positive control (JP-8) and cream sites.

Cream	Primary Irritation Index	Grade
Positive Control*	1.79 ± 0.02	Slight Irritation
Qwanah 08-0416-01	1.66 ± 0.06	Slight Irritation
Positive Control	1.50 ± 0.00	Slight Irritation
Qwanah 08-0614-02	2.11 ± 0.28	Mild Irritation
Positive Data	1.67 ± 0.07	Slight Irritation
Skin Armor Cream 1	1.63 ± 0.06	Slight Irritation
Positive Data	1.38 ± 0.08	Slight Irritation
Skin Armor Cream 2	1.23 ± 0.19	Slight Irritation
Positive Data	1.88 ± 0.00	Slight Irritation
Skin Armor Cream 4	1.77 ± 0.02	Slight Irritation

* The positive control is calculated from the same rabbits on which each cream was tested.

Shown in Figures 20 through 24, the average visual score of erythema of each cream is compared to the positive control. The greatest difference between positive control and tested cream sites was observed in Qwanah Cream 02 in which the cream sites showed increases visual erythema, shown in Figure 21. The other 4 creams tested visually mirrored the positive control results throughout the 72 hour observation period.

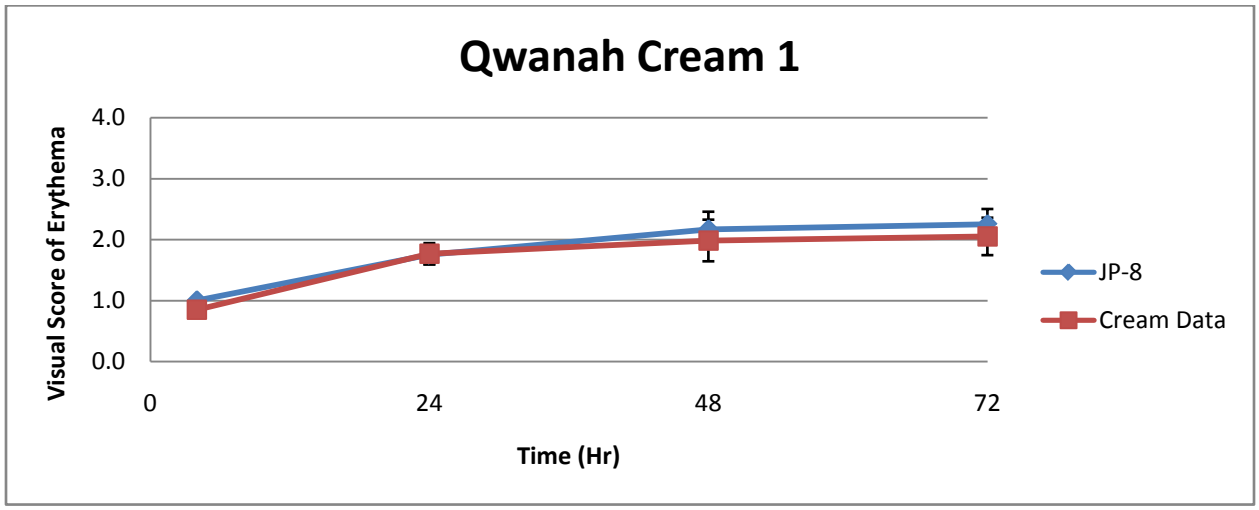


Figure 20: Average visual Draize score over time for sites protected with Qwanah Cream 08-0416-01

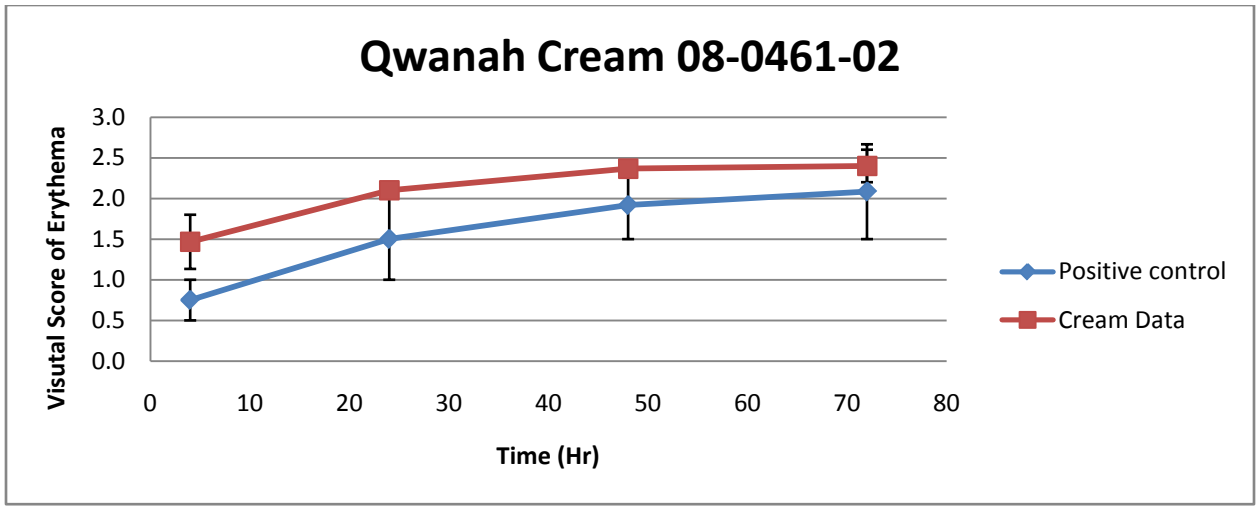


Figure 21: Average visual Draize score over time for sites protected with Qwanah Cream 08-0416-02

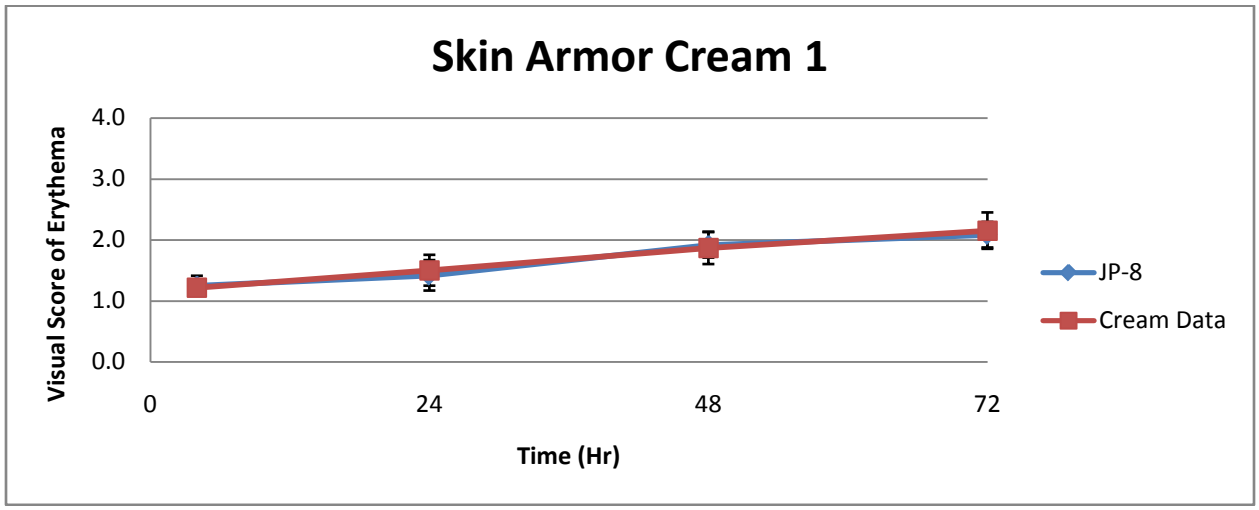


Figure 22: Average visual Draize score for sites protected with Skin Armor Technologies Cream #1

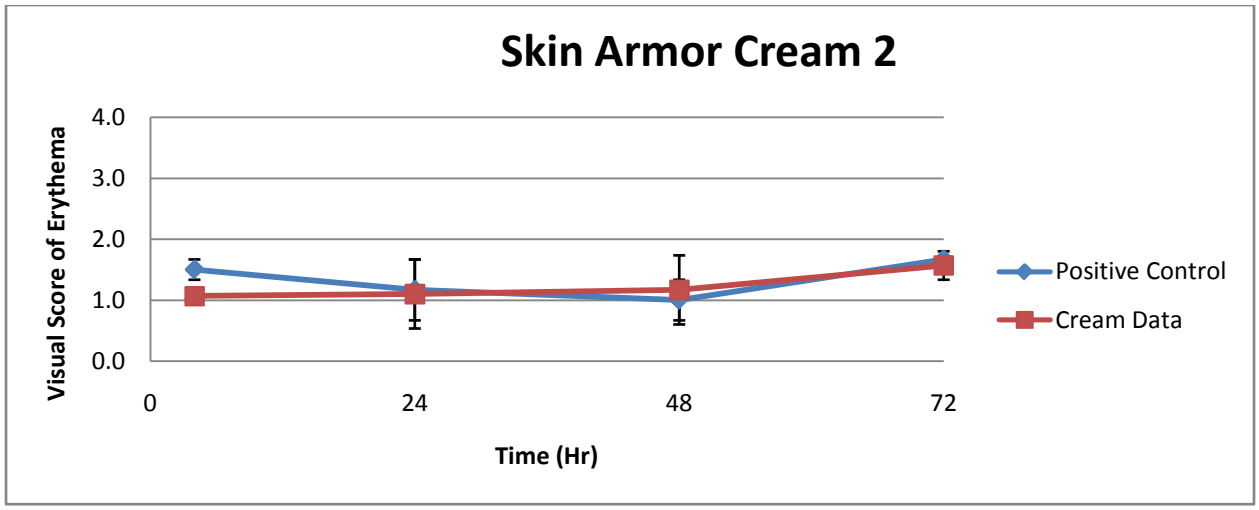


Figure 23: Average visual Draize score for sites protected with Skin Armor Technologies Cream #2

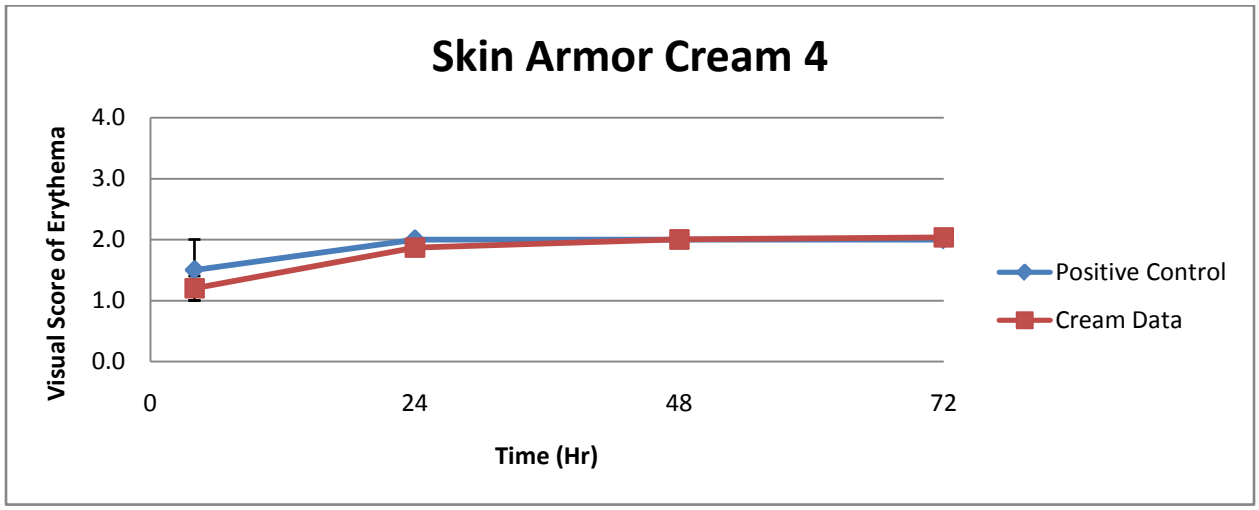


Figure 24: Average visual Draize score for sites protected with Skin Armor Technologies Cream #4

In order to eliminate the suspicion that the duration of exposure was unrealistic to operational exposures, Skin Armor Tech Cream 2 was selected to undergo various JP-8 exposure times. The calculated primary irritation indices for each exposure duration are shown in Table 13. Skin Armor Tech Cream 2 failed to protect against visual irritation from as short as 15 minutes of JP-8 exposure. This part of the experiment provided validation that formulated barrier creams would not provide the necessary protection needed to be operationally fielded.

Table 13: Primary Irritation Index of Skin Armor Technologies #2 applied for various durations of exposure to JP-8

	Duration of Exposure (hours)	Primary Irritation Index	Grade
Positive Control	0.25	1.13 ± 0.00	Slight Irritation
Skin Armor Cream 2	0.25	1.17 ± 0.14	Slight Irritation
Positive Control	0.50	1.75 ± 0.00	Slight Irritation
Skin Armor Cream 2	0.50	1.96 ± 0.07	Slight Irritation
Positive Data	0.75	1.75 ± 0.00	Slight Irritation
Skin Armor Cream 2	0.75	2.00 ± 0.00	Mild Irritation
Positive Data	1.00	1.75 ± 0.00	Slight Irritation
Skin Armor Cream 2	1.00	1.83 ± 0.14	Slight Irritation
Positive Data	1.25	1.79 ± 0.14	Slight Irritation
Skin Armor Cream 2	1.25	1.47 ± 0.07	Slight Irritation
Positive Data	1.50	1.67 ± 0.14	Slight Irritation
Skin Armor Cream 2	1.50	1.50 ± 0.00	Slight Irritation
Positive Data	1.75	1.67 ± 0.14	Slight Irritation
Skin Armor Cream 2	1.75	2.00 ± 0.00	Mild Irritation
Positive Data	2.00	1.75 ± 0.00	Slight Irritation
Skin Armor Cream 2	2.00	1.19 ± 0.07	Slight Irritation

Colorimeter

Shown in Figures 25 through 28, the average change in chroma and color of each cream tested emulates the positive control results. There is a considerable difference between the negative control and any site that JP-8 was applied, regardless of what cream was applied for protection.

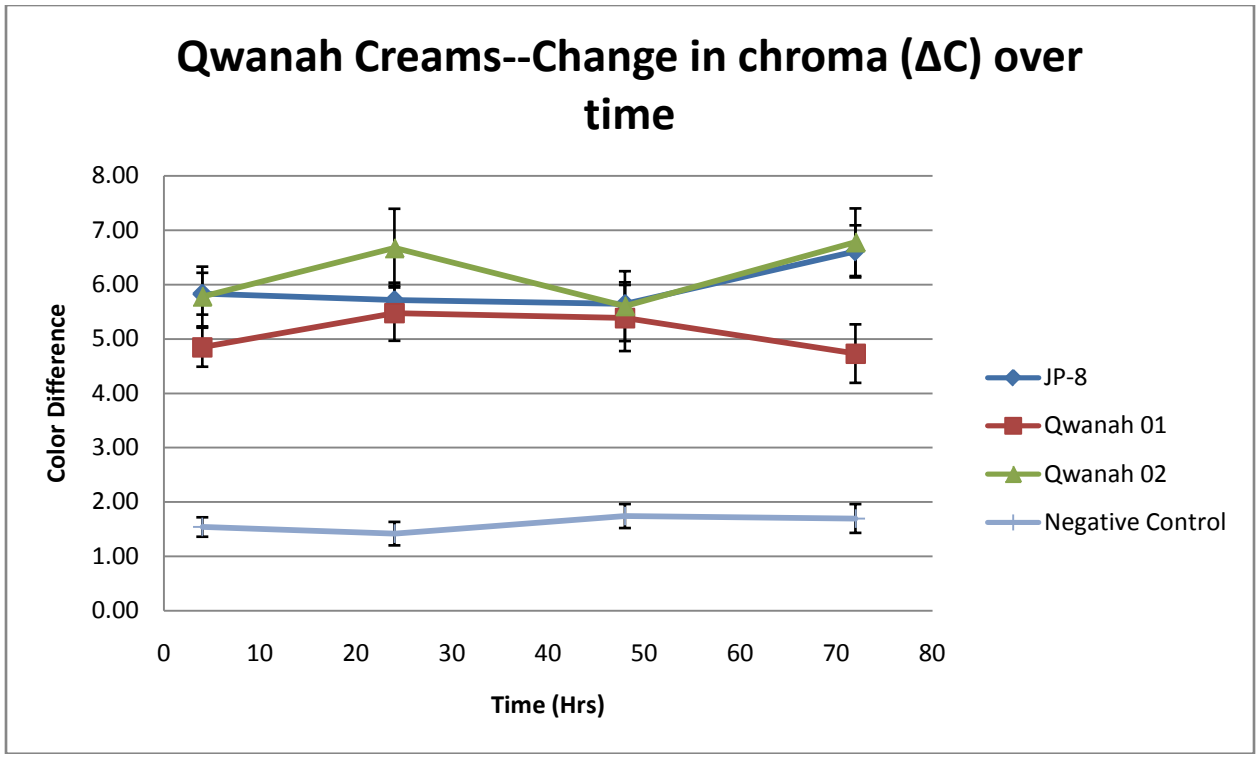


Figure 25: Difference in chroma between initial reading and listed observation time for Qwanah Creams

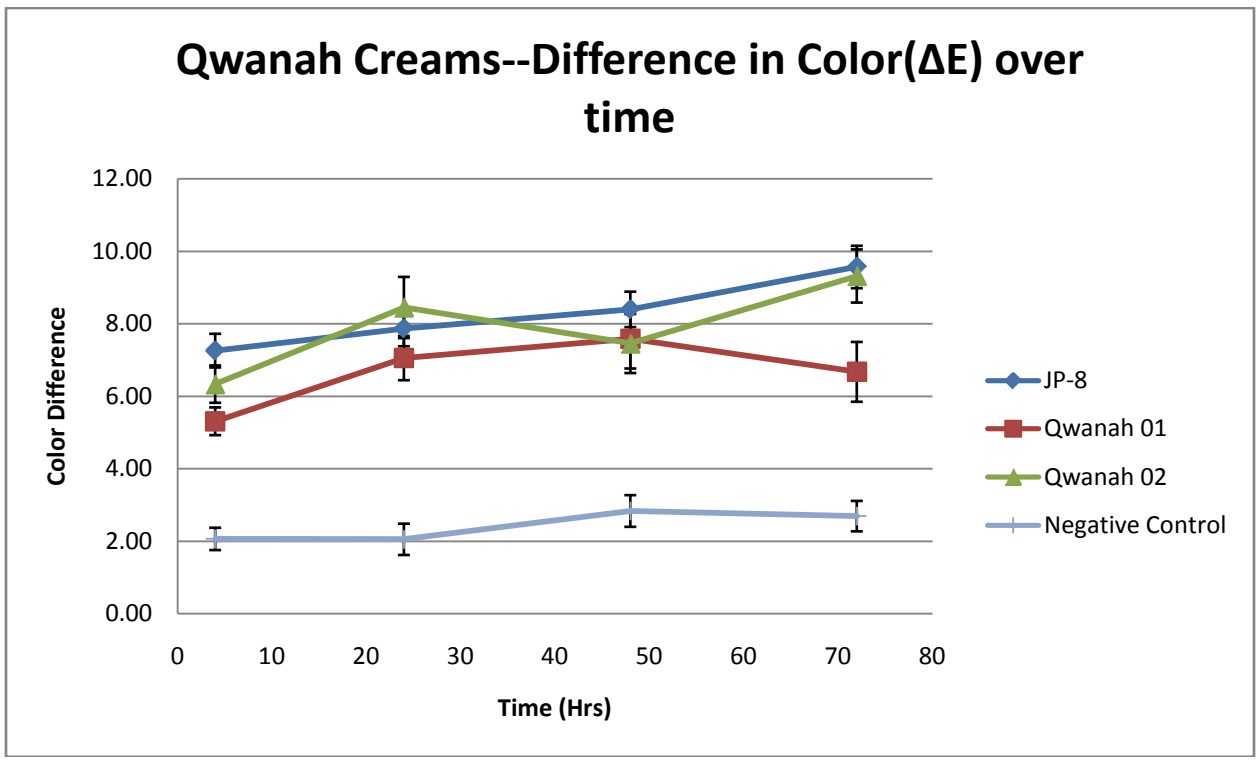


Figure 26: Difference in color between initial reading and listed observation time for Qwanah Creams

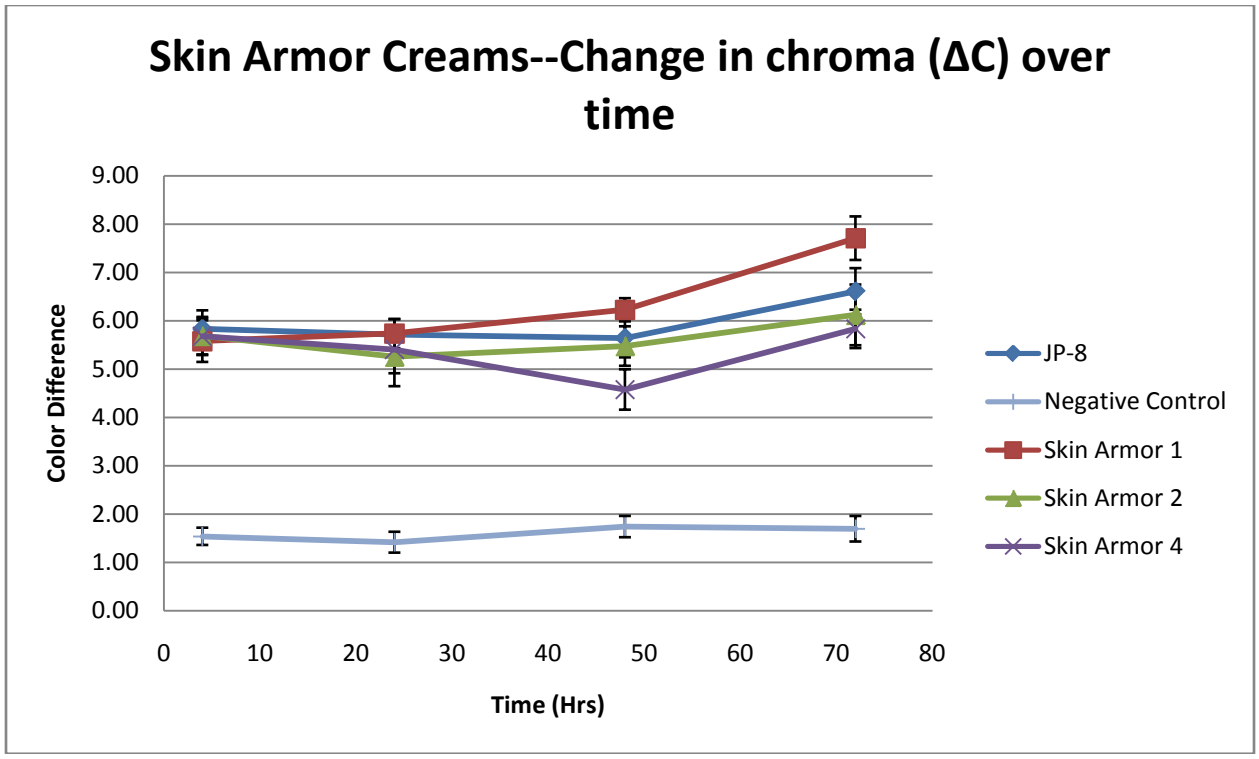


Figure 27: Difference in chroma between initial reading and listed observation time for Skin Armor Creams

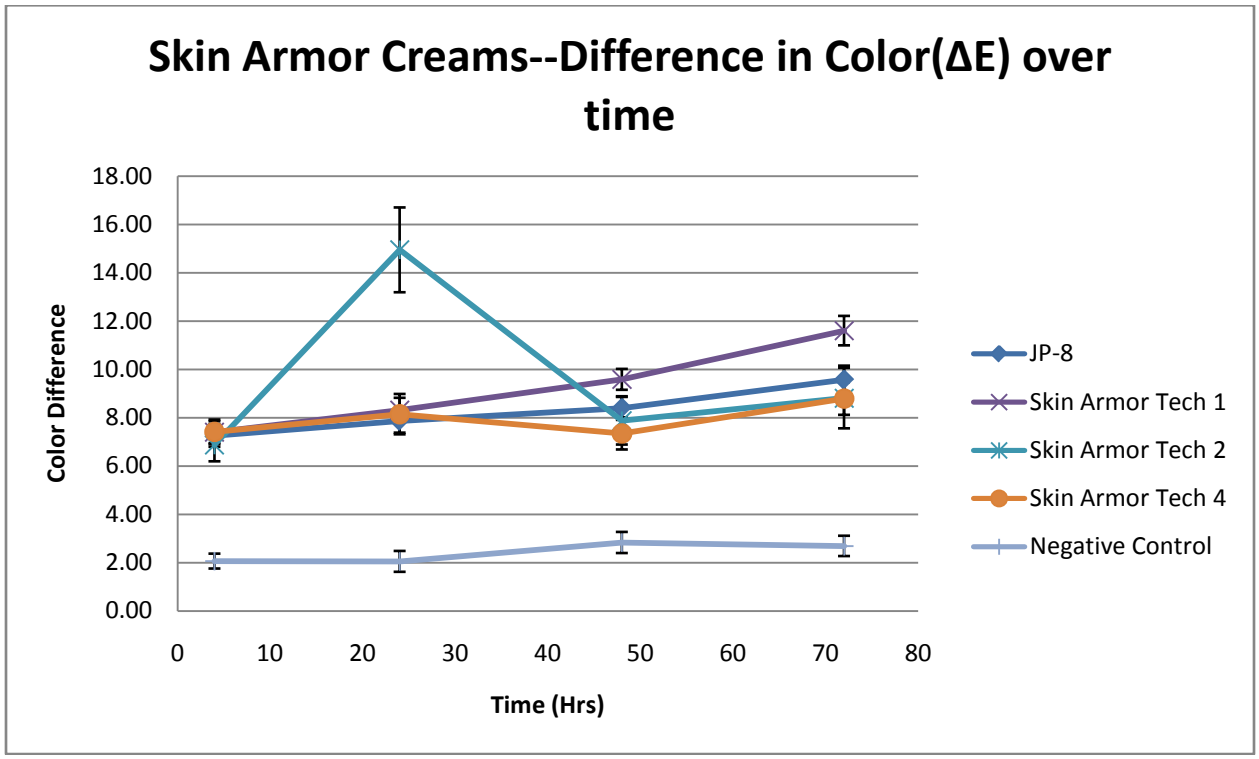


Figure 28: Difference in color between initial reading and listed observation time for Skin Armor Creams

Histopathology

Post-experiment, each animal test site from Skin Armor Technologies was processed for histopathology. The results shown in Table 14 show that Cream #2 provided the best protection of the 3 creams provided from Skin Armor Technologies. Skin Samples were not collected from the rabbits protected with Qwanah due to personnel changes and the lack of a pathologist. Figures 29 through 31 show a representative sample of the protection provided by each candidate barrier cream.

Table 14: Average histopathology scores for Skin Armor Technology creams

Barrier Cream Tested	Cream / HTC Sites	HTC Only Sites	Cream / HTC / JP-8 Sites	Positive Control Sites (JP-8 / HTC)
Skin Armor Cream 1	0.00 ± 0.00	0.75 ± 0.75	3.55 ± 0.18	3.50 ± 0.50
Skin Armor Cream 2	0.00 ± 0.00	0.00 ± 0.00	1.80 ± 0.55	3.00 ± 1.00
Skin Armor Cream 4	0.00 ± 0.00	0.00 ± 0.00	3.2 ± 0.20	3.00 ± 0.00

The histopathology results from various durations of exposure site protected with Skin Armor Cream #2 are shown in Table 15. As with the visual data, Cream 2 failed to provide protection from irritation for just 30 minutes of JP-8 exposure.

Table 15: Average histopathology scores from Skin Armor Cream #2 with various durations of JP-8 exposure

Exposure Duration (hours)	Cream / HTC / JP-8 Sites	Positive Control Sites (JP-8 / HTC)
0.25	2.00 ± 1.00	0.50 ± 0.50
0.50	4.00 ± 0.00	3.50 ± 0.50
0.75	4.00 ± 0.00	3.00 ± 1.00
1.00	4.00 ± 0.00	3.50 ± 0.50
1.25	3.00 ± 0.00	3.50 ± 0.50
1.50	3.50 ± 0.50	4.00 ± 0.00
1.75	4.00 ± 0.00	3.50 ± 0.50
2.00	4.00 ± 0.00	3.50 ± 0.50

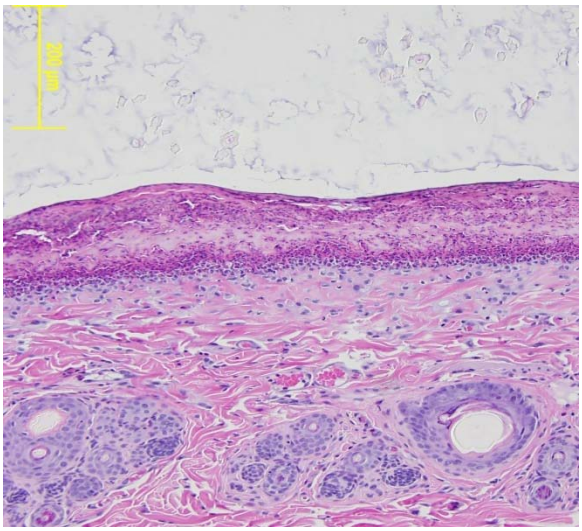


Figure 29: Clipped haired skin, rabbit 08016 protected with Skin Armor Technologies barrier cream 1. Epidermal necrosis with collagen degeneration/necrosis through depth of papillary dermis. Score: 4+

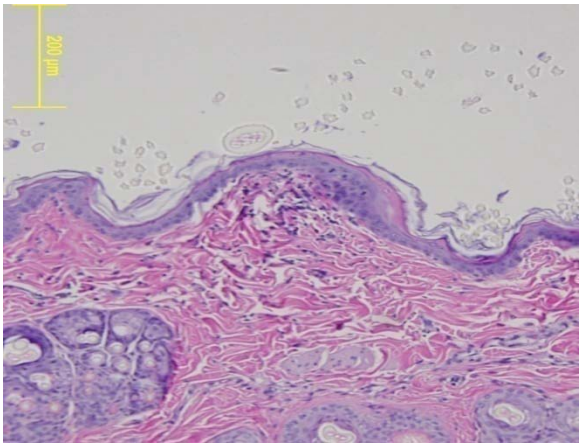


Figure 30: Clipped haired skin, rabbit 080159 protected with Skin Armor Technologies barrier cream 2. Multifocal epidermal degeneration/necrosis, no dermal lesion. Score: 1

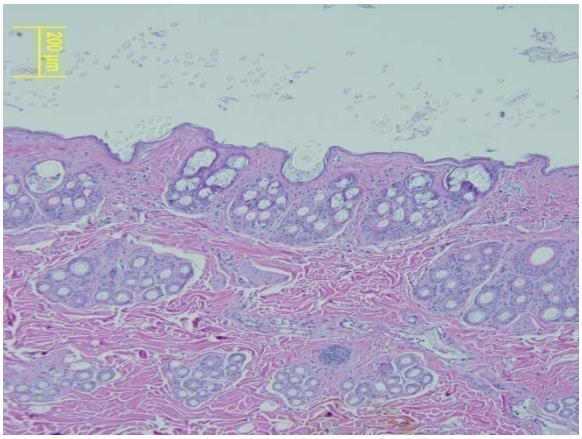


Figure 31: Clipped haired skin, rabbit 080154 protected with Skin Armor Technologies barrier cream 4. Epidermal necrosis with collagen degeneration/necrosis in very superficial papillary dermis. Score: 3

DISCUSSION

JP-8 jet fuel, being a vastly complex mixture, displays a wide-range of biological interactions both localized and systemic (McDougal *et al.*, 2000). Acute dermal exposure continues to be a primary exposure that airman subjected to the harmful effects of jet fuel. As with many harmful materials, the best way to protect one's self is with PPE such as gloves. The purpose of this study was to determine if protection through the use of a barrier skin cream or lotion was feasible, and if it would provide sufficient, consistent protection with one application prior to JP-8 exposure.

The first 2 stages of this project tested the possibility of a previously manufactured cream to provide the protection that was desired. The differences between the Silastic® and the harvested pig skin were not surprising since the skin is a much more effective barrier to the fuel. While the cream was the major barrier in the Silastic® example, the added creams showed little difference on harvested pig skin and rabbit skin. While some OTC barrier creams did show promise in early stages of the project via the cell diffusion chambers, they did not prevent percutaneous absorption and skin irritation once tested on a live animal model. Some formulated creams proved to impede the penetration of JP-8 in the cell diffusion chamber better than the OTC cream, but as shown in Section 4 of this project, did not perform when tested on the animal model.

A cooperative research and development agreement (CRADA) was formed prior to the start of Section 4 of this project. The primary objective of this CRADA was to identify and evaluate formulation for topical application of the skin that protects the skin from environmental irritants to include man-made chemicals, naturally occurring agents and solar radiation (USAF CRADA # - HE-CRD). Products from this CRADA were several skin formulations that were tested for barrier effectiveness. Although these creams proved to be ineffective in the animal model, the CRADA fulfilled its overall objective of evaluating creams for topical application.

CONCLUSION

The best way to protect one's self from JP-8 dermal exposure is by properly using personal protection equipment. PPE is always the first line and best way of defense from harmful compounds such as JP-8. Skin barrier creams were not shown to be consistently effective in deterring dermal irritation as shown in the animal model used in this study. This study was determined to not move forward to human testing due to the lack of sufficient protection provided by candidate barrier creams in the animal model.

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