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FINAL REPORT

July 1965 - July 1968

CHEMICAL BINDING OF PROTECTIVE AGENTS TO THE HUMAN STRATUM CORNEUM

Contract No. DA 49 - 092 - ARO - 85

Department of the Army
Army Research Office
3045 Columbia Pike
Arlington, Virginia 22204

ATTENTION: Dr. Eugene Sporn

Co-Principal Investigators:

Dr. Thomas B. Fitzpatrick and Dr. Madhukar A. Pathak
Department of Dermatology
Harvard Medical School
Massachusetts General Hospital
Boston, Massachusetts 02114

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CHEMICAL BINDING OF PROTECTIVE AGENTS TO THE HUMAN STRATUM CORNEUM

STATEMENT #3 UNCLASSIFIED

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We wish to express our sincere appreciation to Mr. Frank A. Eyman, Superintendent, Arizona State Prison, Florence, Arizona and W. J. Clemans III, M.D., Chief of Staff, Arizona State Prison Hospital, Florence, Arizona for their kind permission, cooperation, efforts, personal attention and kindness in carrying out the three clinical trials at the Arizona State Prison.

Thomas B. Fitzpatrick
Madhu A. Pathak

SCOPE OF THE REPORT

This contract was awarded on May 30, 1965, but the work did not get underway until July 1, 1965 for various reasons including the final ratification of the contract by Harvard University. Initially, the contract was awarded for a period of two years. Upon our request it was, however, extended for an additional one year period. This report summarizes our work of three years (1965-1968) on the chemical binding of photo-protective agents to the human stratum corneum. As a result of extensive laboratory studies and four clinical trials on human volunteers (three conducted at the Arizona State Prison, Florence, Arizona and one conducted on the snow covered mountains of the Swiss Alps), we report the following:

a) We have developed at least two formulations which, after a single application, protect individuals undergoing long exposure to the sunburn spectrum.

b) The formulations are more effective than commercially available products.

c) They afford effective protection after exercise and sweating.

d) Both the preparations, after a single application, tend to remain on the skin after bathing or swimming and exert a partial, yet very satisfactory protection.

e) Under normal conditions of use, that involve activities such as swimming, playing ball, walking and lying, a single application of these preparations provides very effective protection.

f) The preparations do not inhibit tanning by long-wave ultraviolet and visible radiation.

g) The preparations are cosmetically acceptable, being invisible and without odor or color on the skin.

h) They have no contact or allergenic sensitizing potential.

i) Under intensely bright sun with hot and dry climatic conditions, under warm and humid conditions, and on snow covered mountains at high altitudes, that reflect ultraviolet radiation, preparation MAP, MAP (D5) and G were found to provide very effective protection.

Our efforts to promote a stable chemical conjugation (under physiologic conditions) of this agent to the stratum corneum with the hope that a single application would afford a long term protection (at least a period of seven days) were, however, unrewarding.

The following preparations are recommended for obtaining effective protection against sunburn.

1) Preparation MAP: 5% para-aminobenzoic acid in 75 - 95% ethyl alcohol (pH 4.5 - 4.8).

2) Preparation MAP (D5): contains 5% para-aminobenzoic acid in 70 - 75% ethanol, 5% oleyl alcohol and 2" polyoxyethylene oleyl ether as emollients and solubilizer.

3) Preparation G: contains 2.5% Eskalol 506 (isoamyl-p-n, n-dimethylaminobenzoate or glyceryl-para-aminobenzoate) in 65 - 70% ethyl alcohol.

It is apparent that this research has not uncovered any new agents; para-aminobenzoic acid and its derivatives have been used for over forty years as topical sunscreens. The unique finding from these studies is that formulations of para-aminobenzoic acid and its esters in ethyl alcohol are the most effective topical agents and remain bound to the skin after immersion or exercise. There are no commercially available preparations, to our knowledge, that contain over 2% para-aminobenzoic acid or its esters in ethyl alcohol. It appears that previous studies on sunscreens have not been done under rigorous field conditions, have not been well controlled, and have not tested solar protection following exercise or immersion. This formulation of PABA and PABA esters in ethyl alcohol has never been explored and adequately tested. Therefore, these studies have provided, with commercially available photoprotective agents (PABA and its esters):

1) a new formulation that approaches an ideal topical screen against the sunburn spectrum;

2) a series of controlled studies of most of the commercially available sunscreens performed in an adequate number of subjects under rigorous field conditions of the desert and the mountains.

We recommend that these preparations, listed above, be considered by the Military as the most effective and practically usable topical sunscreens available.

Preparation G was formulated and supplied under sponsorship of the Sea and Ski Corporation.

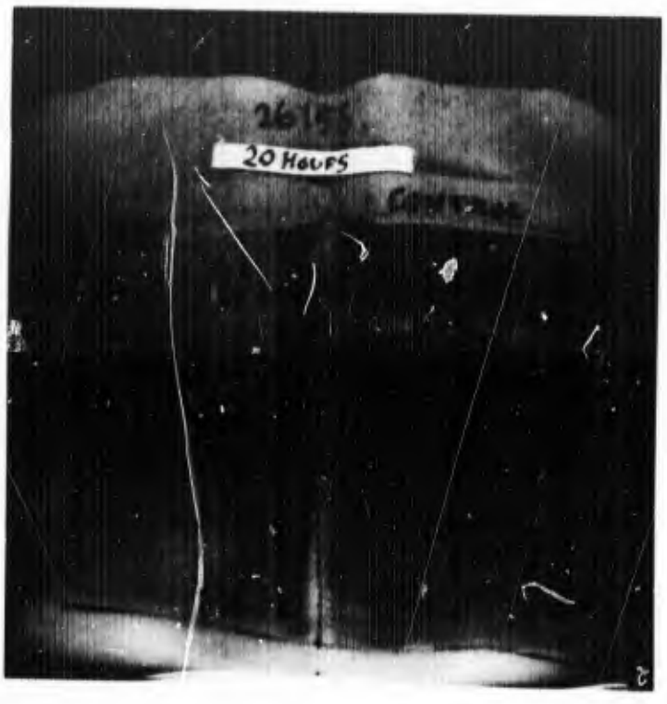

Figure A: Showing the prolonged protective effect of MAP. After a single application of MAP (left side) and Sea & Ski (right side), the subject (# 26156) was exposed to 1, 2½ and 4 hours of solar radiation. Control (untreated, upper right) skin area received only 1 hour of sun exposure.

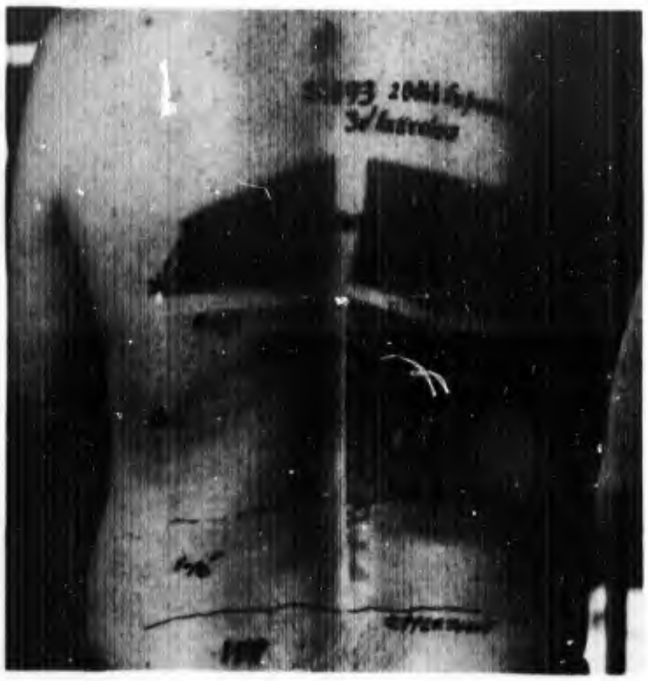

Figure B: Illustrating the protective effect of MAP and a commercially available preparation (Coppertone shade, extra protection) under conditions of exercise (30 minutes) accompanied by sweating. The control untreated (top), MAP and Coppertone treated skin areas received 70 and 120 minutes of sun exposure.

Figure C: Illustrating the protective effect of MAP under conditions simulating normal usage. Normal conditions of use included two periods of swimming (10 minutes each), lying on the back and in the prone position (1 hour), and walking and running around the court and sitting in the sun (1 hour). The control (untreated) area is in the center and UVAL (a leading commercial product) treated area is on the right. The right and the left sides received a single application of the test products.

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INTRODUCTION

Solar Radiation and the Erythemal Spectrum: The spectrum of the electromagnetic radiation stretches through an enormous range, from gamma rays, which may be less than 0.01 Å long, to radio waves that may be between 1 mm. and several kilometers long. The intensity of solar radiation outside the earth's atmosphere at the earth's mean solar distance (i.e. the solar constant) averages about 2.002 cal per cm^2 or 0.1396 watt per cm^2 (F. S. Johnson, J. Meterol., 2:431, 1954). This is equivalent to 1170 watts/square yard. About two thirds of this energy actually reaches the surface of the earth, the remainder being reflected, scattered or absorbed in the atmosphere. About half of the sun's energy is concentrated in the visible portion of the spectrum (i.e. between 380-700 mu), about 40% in the infrared and about 10% in the ultraviolet. Practically all of the radiation shorter than 295 mu is absorbed in the earth's atmosphere so that radiation below 295 mu does not appear to reach the surface of the earth. It is fortunate that man lives on a planet that is not reached by wavelengths shorter than 295 mu. Wavelengths shorter than 290 mu, whose energy content is more than 95 K.cal per mole of quanta, can damage severely biologic tissue like skin; they can denature inactivate proteins, depolymerize nucleic acids, induce mutations and the enzymes essential for biologic functioning. Radiation between 297-1100 mu, that reaches the surface of the earth, constitutes a major portion of the solar radiation (about 75%). For the most part, this energy is biologically useful in that it promotes such orderly and beneficial chemical reactions as photosynthesis, visual photoreception, photoreactivation, cutaneous pigment darkening and melanogenesis. Human skin can utilize a large part of this solar energy without demonstrable injurious effects, but within this broad spectrum there is a band of radiation with wavelengths between 297 and 320 mu (the so-called erythemogenic spectrum) that is capable of inducing cutaneous sunburn, carcinogenesis, senile elastosis and phototoxic reactions (exaggerated sunburn response) due to ingestion of certain drugs.

Sunburn: Since 1922, it has been known that sunburn and suntan of the human skin are due to the action of a narrow band of wavelengths in the ultraviolet spectrum of solar radiation. The erythemal action spectrum obtained with high-pressure, hot quartz mercury arc shows the maximum sunburn response at 297 mu with little effectiveness below 280 mu. A second peak of erythema producing activity is observed at 257-260 mu. When subjects are irradiated with solar radiation, the most effective wavelength is approximately 307 mu (Coxxxx1963xx) It should be emphasized that weak erythemogenic radiation (e.g. 310-320 mu) contributes significantly to the development of erythema induced by solar radiation because the relative proportion of these wavelengths, as compared to 300-307 mu wavelength, is significantly more in the solar radiation that reaches the earth's surface. Wavelengths greater than 320 mu and up to 420 mu can also induce sunburn reaction, but these wavelengths are less erythemogenic. For example, in a place like Florence, Arizona, the minimal erythema dose for a fair-skinned individual (back region) is approximately 25×10^4 ergs per cm^2 if the subject receives only radiation of 300-310 mu.

Introduction (continued)

(Minimal erythema dose was 10 minutes during midday in the month of April). It takes at least 120 minutes of sun exposure to obtain minimal perceptible erythema when the same individual is exposed to 320-420 mu wavelengths. Almost no erythema is produced by wavelengths longer than 420 mu. Thus, for practical purposes, the erythemogenic spectrum is generally considered between 290 and 320 mu with peaks at 297 or 300-307 mu.

Sunburn and Suntanning: The familiar sunburn and suntanning that follow exposure of skin to solar radiation or to ultraviolet light from artificial sources, are known to involve several distinct photobiological processes.

1) An initial faint erythema that is transient and disappears shortly after exposure.

2) Delayed erythema that begins to appear 2-4 hours after sun exposure and reaches a maximum intensity of bright pink to red color in 14-24 hours. This erythema gradually diminishes within the next 48 to 72 hours. The vascular response to localized ultraviolet induced injury is characterized by vasodilatation and augmented blood flow, increased vascular permeability, and cellular exudation, particularly of neutrophil leukocytes. The superficial capillary network and the dermal networks of venules and capillaries are affected in this injury.

3) Immediate pigment darkening and redistribution of melanin pigment that is already present in the skin prior to irradiation.

4) Melanogenesis, which involves production, transfer and redistribution of new melanosomes. It also involves an increase in the number of functioning melanocytes.

The degree of erythema that follows exposure of human skin to solar radiation varies with the total dose of radiation received. The same dose of solar radiation may give a variable response at different body sites. The back, the popliteal region of the legs, and abdomen are most sensitive to sunlight. The degree of melanogenesis that follows exposure of human skin varies with the total dose of radiation received and the severity of the epidermal cell damage. The dose of radiation that causes only mild damage promotes pigmentation more effectively than the dose that causes severe sunburn response. When the skin has been exposed to a severely damaging dose of radiation manifested by marked redness, all epidermal cell layers are injured. Individual cell dyskeratosis, epidermal disorganization and even desquamation of keratinocytes and melanocytes ensues. In such injured skin, melanogenesis is poor, and pigmentation may not be markedly visible. Melanogenesis does occur, however, when the epidermis shows multiple foci of dyskeratosis without general disorganization of the cell layers. In such mildly damaged skin, the dendritic arborization of melanocytes, hypertrophy of melanocytes and the formation of new melanosomes is markedly evident. Thus, the primary aim in any topical sunscreen preparation is to minimize severe damage to the epidermal cells.

THE PROBLEM OF SUNBURN AND OUR OBJECTIVES

Of the many and diversified functions of the skin, protection against actinic damage constitutes one of the most important. The protection which the skin surface affords is really one of its essential functions and is accomplished by its excretory and secretory products. These are: 1) the proteins of stratum corneum and viable epidermis, namely keratin and kerotohyalin granules, 2) the melanin present in the melanosomes, in the form of melanoprotein, 3) urocanic acid, the deaminated product of histidine, 4) the lipids and the lipoproteins, and 5) carotenes. In spite of these cutaneous defenses against solar radiation, human skin is vulnerable to sunburn. Until recently, effective protection against sunburn has been attempted by:

a) limiting the sun exposure by staying indoors.

b) controlling the sun exposure and promoting hyperpigmentation and thickening of the horny layer by gradual increment of sun exposure. The increased pigment content of the epidermis and the hyperplasia builds up the resistance to sunburn.

c) by applying to the skin a film of a sunscreen preparation which, because of absorption or opacity or both, reduces or minimizes the impact of sunburn radiation on the vulnerable cells of the epidermis.

d) by ingesting oral medications such as antimalarials, or psoralens, and promoting tanning through controlled sun exposure.

e) by chemical modification of the horny layer so that the exogenously applied light absorbing molecule in the sunburn spectrum conjugates chemically with the stratum corneum and filters out the damaging radiation.

Although the method (c), topical application of sunscreen preparation that involves the has been most widely used, it has a major disadvantage in that the externally applied agents are too easily washed or rubbed off the skin. The commercially available sunscreen preparations, under natural conditions, do not remain on the surface of the horny layer. Either they are eluted as a result of sweating or undergo chemical alterations and degradations under the influence of solar radiation. Most of the commercial preparations are easily washed off after swimming or bathing. Furthermore, many commercial preparations are very effective only when tested under ideal laboratory conditions using artificial UV light sources. Under natural sunlight and outdoor field conditions, the same preparations are found to be ineffective.

The chemical alteration of the horny layer, the natural filter in situ, was therefore explored. Pretreating the stratum corneum with harmless agents

that would facilitate binding of the specific groups of keratin with chemical agents that have the property to absorb the erythemogenic solar spectrum was undertaken. The objectives of this study were:

1) Development of a safe, highly effective sunscreen preparation that is partially bound to human skin.

2) To explore and to promote a chemical binding of a non-sensitizing, ultraviolet absorbing agent (wavelengths less than 320 mu) with the constituents of the stratum corneum under normal physiologic conditions. A binding that is envisaged is a type of stable conjugation that is not readily dissociated with such agents as water, soap and detergents.

3) To compare the sunburn preventive potency of MAP* or MAP (D5)* with various sunscreen agents (both pure compounds and commercial preparations) that are already known and have the property to absorb effectively the sunburn radiation (wavelengths less than 320 mu).

4) To determine the degree and the duration of protection afforded by any given agent after a single topical application under three experimental conditions:

    a) without any washing
    b) after inducing profuse perspiration by exercise
    c) after swimming

5) To determine the concentration of the sunscreening agent necessary for affording a complete protection against sunburn radiation.

6) To determine the effects of emollients.

7) To determine the effects of various solvents, e.g. methyl alcohol, ethyl alcohol, propyl, isopropyl, butyl and isobutyl alcohols.

8) To compare the efficacy of various commercial preparations, not only under laboratory conditions using artificial light sources, but most importantly, under outdoor conditions using natural sunlight.

9) To ascertain whether suntanning (melanogenesis or new melanin formation) can be promoted without evoking the discomforts of sunburn.

* An arbitrary designation used throughout this report for a formulation of para-aminobenzoic acid in ethanol that we feel can be of great use to the United States Army. Unless and otherwise indicated, the preparation MAP implies 5% para-aminobenzoic acid in 70-95% ethanol, and MAP (D5) or D5 implies 5% para-aminobenzoic acid in 70% ethanol with emollients.

THE APPROACH TO THE PROBLEM OF SUNBURN AND ITS PREVENTION

This study was carried out in two phases. The first phase included the evaluation of the sunburn prevention potency of various chemical agents that effectively absorb sunburn radiation. The evaluation was carried out under laboratory conditions using female volunteers and artificial ultraviolet light source for producing sunburn response. The second phase included the evaluation of the sunscreen preparations formulated in our laboratory under outdoor conditions. Three clinical trials were carried out on male volunteers at the Arizona State Prison in Florence, Arizona and one clinical trial was conducted in the Swiss Alps.

Phase I: The compounds included in the first phase of our studies were:

1. Anthranilic acid (e.g. benzyl, phenyl and menthyl).

2. Various esters of para-aminobenzoic acid (ethyl, propyl, isobutyl, monoglyceryl and dimethyl esters of PABA).

3. Benzophenones: 2,4-dihydroxy, 4-methoxy benzophenone; 2-hydroxy, 4-methoxy benzophenone, 5-sulfonic acid; 2,2'-dihydroxy, 4-methoxy benzophenone.

4. Benzylsalicylate.

5. Pyrrones: cinnamic acid, umbelliferone, esculin, esculetin.

6. Petrolatum (RVP)

7. Naphthoquinones: 2-hydroxy,1,4-naphthoquinone and 5-hydroxy,1,4-naphthoquinone.

8. Dihydroxyacetone.

9. Para-aminosalicylic acid.

10. Sodium and potassium salts of para-aminobenzoic acid.

11-35. In addition, 24 of the currently available commercial products were carefully investigated in this study.

The compounds were dissolved in various solvents such as methanol, ethanol, isopropanol, acetone, butenol, etc. They were applied in 1.5" x 1.5" skin areas on the back. Each compound was applied at least in 6 areas. Half of the applied areas were cleaned with water (manual cleaning with water-soaked cotton gauze). Each treated site was exposed to a varying dose of UV radiation (3 to 10 times the minimal erythema dose) and compared against control areas

(untreated) that received an equivalent dose of UV radiation. After 24 hours, the exposed sites were evaluated for the degree of erythema.

It was observed that these compounds listed above, when applied in a thin film in various solvents, do exhibit photoprotective action to a varying degree. However, when the applied film was washed off, most of the commercial preparations except UVAL, Snootie, Skolex and Solbar were found to be ineffective. Furthermore, PABA in acid form, the benzophenones, esculin and para-aminosalicylic acid were found to afford a far better protection against erythemogenic radiation than other compounds listed above. It was the alcoholic solution of PABA that proved to be most effective in providing the protection even after repeated washings with water. This was the beginning of our project, and we undertook to investigate the unique property of this PABA preparation in ethyl alcohol. Surprisingly, the sodium and the potassium salts of PABA, in ethyl and methyl alcohol, were found to be ineffective; the applied film of these salts was easily washed off. Para-aminosalicylic acid (PAS) so far has not been reported to be a useful sunscreening agent. We, however, found it to be very effective. Indeed, if it did not have the disadvantage of staining the garments, we would strongly recommend it as an additional sunscreening agent, better than many of the commercial preparations that are available. Since topical preparation has this disadvantage, the effectiveness of PAS as an orally administered sunscreening agent is highly recommended in future investigations. The toxicity and pharmacology of PAS is well known and it would be certainly worthwhile to explore the effectiveness of PAS as an oral sunscreen agent.

MAP preparation was further investigated. The effect of pH: The effect of varying the pH of MAP solution was tested. MAP solution of pH 4.5 - 4.8, 6.0, 7.0, 8.0 and 10 were tested. Acidic solutions (pH 4.5-4.8) of MAP were found to be most effective.

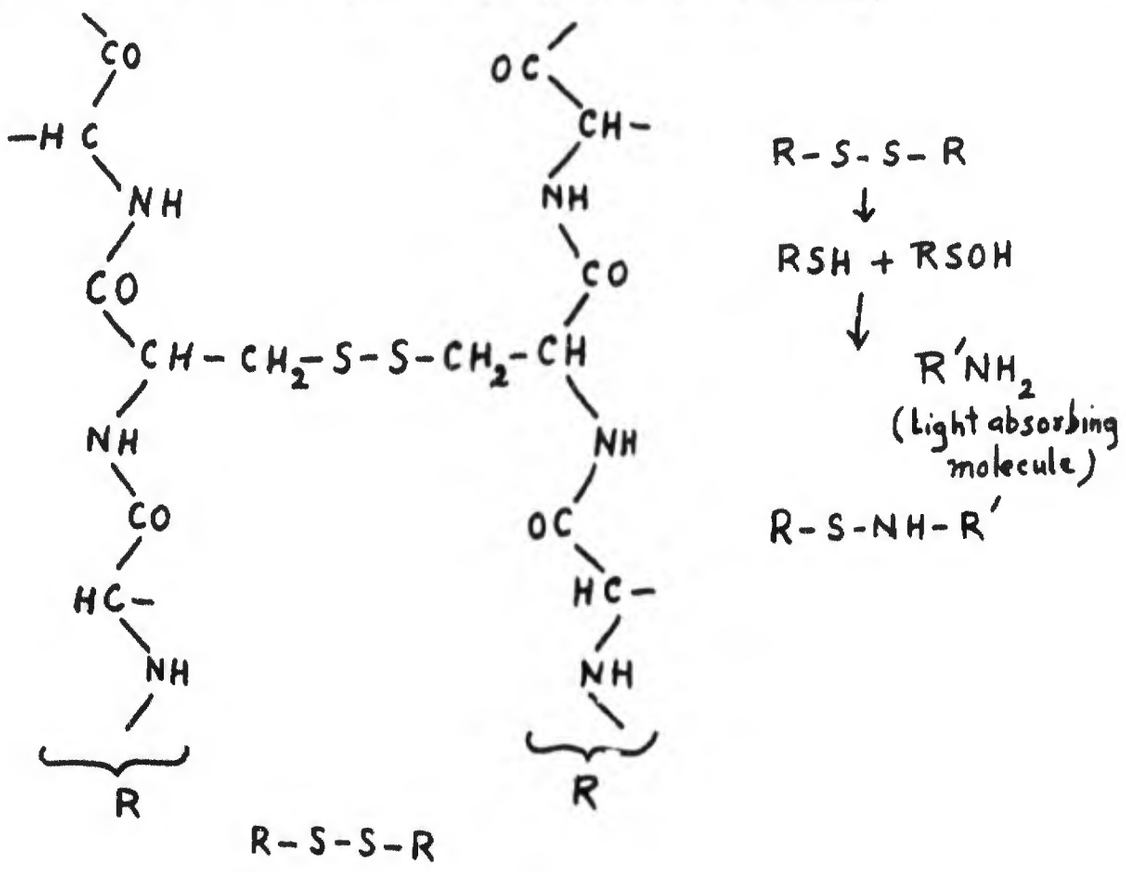
The duration of the protective effect: If MAP preparation had a real affinity to remain either partially adsorbed or chemically conjugated, it was essential to know how long a single application of MAP would afford protection under normal daily activities of any individual. Ten female volunteers were used to evaluate this question. MAP solution was applied once on the 2/3 region of the back. A control area (untreated) was also included to evaluate the response of the untreated skin to varying doses of ultraviolet radiation, and also to determine the minimal erythema dose (MED) of the individual. In most of the subjects, the MED ranged between 15 and 20 seconds (24 hour response). Subjects were then exposed daily for 4 days to 60, 90, 120, 180, 240 and 300 seconds of ultraviolet radiation. The subjects were requested to avoid any shower or bath, during the 4 day test period. The natural perspiration and the frictional contact of the skin with the clothes were the only factors that were likely to remove the photoprotective agent during this period. All subjects showed 100% protection in all exposed areas for the first day. At 48 hours after application, all the volunteers showed excellent protection against 60, 90, 120 and 180 seconds of ultraviolet radiation. At 72 hours, very satisfactory protection was observed up to 120 seconds of ultraviolet radiation and only

partial protection was detectable for 180 and 240 seconds of ultraviolet radiation. At 96 hours, the MAP treated skin began to show the loss of the protective film and the intensity of erythema in the MAP treated areas receiving 120, 180, 240 and 300 seconds of ultraviolet radiation was approximately the same as observed in the control (untreated) area. By the end of 120 hours, the subjects were no longer exhibiting any photoprotective action of MAP that had been applied 5 days earlier. These observations led us to conclude that the preparation MAP had undoubtedly a real affinity to combine with the horny layer. Most of the commercial preparations in our experience, were found to be ineffective within 24 hours after a single application. They do not exhibit this property of prolonged protection.

In vitro and in vivo experiments to demonstrate chemical conjugation of para-aminobenzoic acid to the horny layer:

Introduction

The existence of disulfide bonds in the keratin of the horny layer is well known. Briefly, the principle involved is to reduce the disulfide bonds of the stratum corneum and promote binding under physiologic conditions with a suitable light absorbing agent. This reaction can be described as follows:



R' - NH_2, the exogenously applied material will have the following property:

a) it will not easily dissociate from the chemical bonds established with the keratin. In other words, it will have greater substantivity to remain on the horny layer and provide a stable light absorbing mantle on the surface of the skin.

b) it will remain on the skin surface under varied conditions associated with diverse activities in warm and sunny environment.

c) it will effectively absorb the erythema producing wavelengths.

d) it will be non-toxic, i.e. free from primary effects of contact sensitization.

e) it will possess physical properties which are acceptable to the consumer(e.g. odor, color, staining property etc.)

f) it will not be photolabile and will not dissociate in the presence of moisture, perspiration and immersion in water.

In vitro experiments: Isolated pieces of horny layer (approximately 1 inch in diameter) were obtained from fair-skinned Caucasian subjects by applying cantharidine solution. Specimens were dried overnight in desiccators containing anhydrous calcium sulfate or activated alumina. Dried pieces of horny layer were sandwiched between two aluminum rings, especially designed for obtaining light transmission spectra. The samples were placed in front of phototubes in a prefixed position and maximum care was taken to avoid any scattering of the transmitted light that was impinging on the phototube. The light transmission spectra of the dried specimens were obtained before and after any specific treatment discussed below. In addition, specimens of white Caucasian skin from the umbilical region were obtained at autopsy within 4-6 hours after death. Pieces of epidermis in fairly large sizes (4" x 2") were obtained by heat separation (by immersing the skin in warm water at 60°C for approximately 45 seconds). The outer side of the horny layer, as well as of the epidermis, was subjected only to chemical treatments. The inner side remained untreated except when subjected to repeated rinsing in water. Initially, pre-treatment of horny layer with thioglycolic acid (TGA), or Lilt, a commercial product that also contains TGA, was attempted with a hope that the -S-S- linkages in the keratin molecule of the horny layer would be reduced and the SH- groups made available for chemical conjugation of light absorbing molecules such as para-aminobenzoic acid and other salts of PABA. To evaluate the effects of any pretreatments with chemical agents on the light transmission properties of human epidermis, control runs were always concurrently carried out. Specimens of epidermis as well as horny layer were allowed to react with ortho-meta-, and para-substituted amino derivatives of benzoic acid. Sodium and potassium salts of PABA were also studied for their chemical conjugating affinity with the skin.

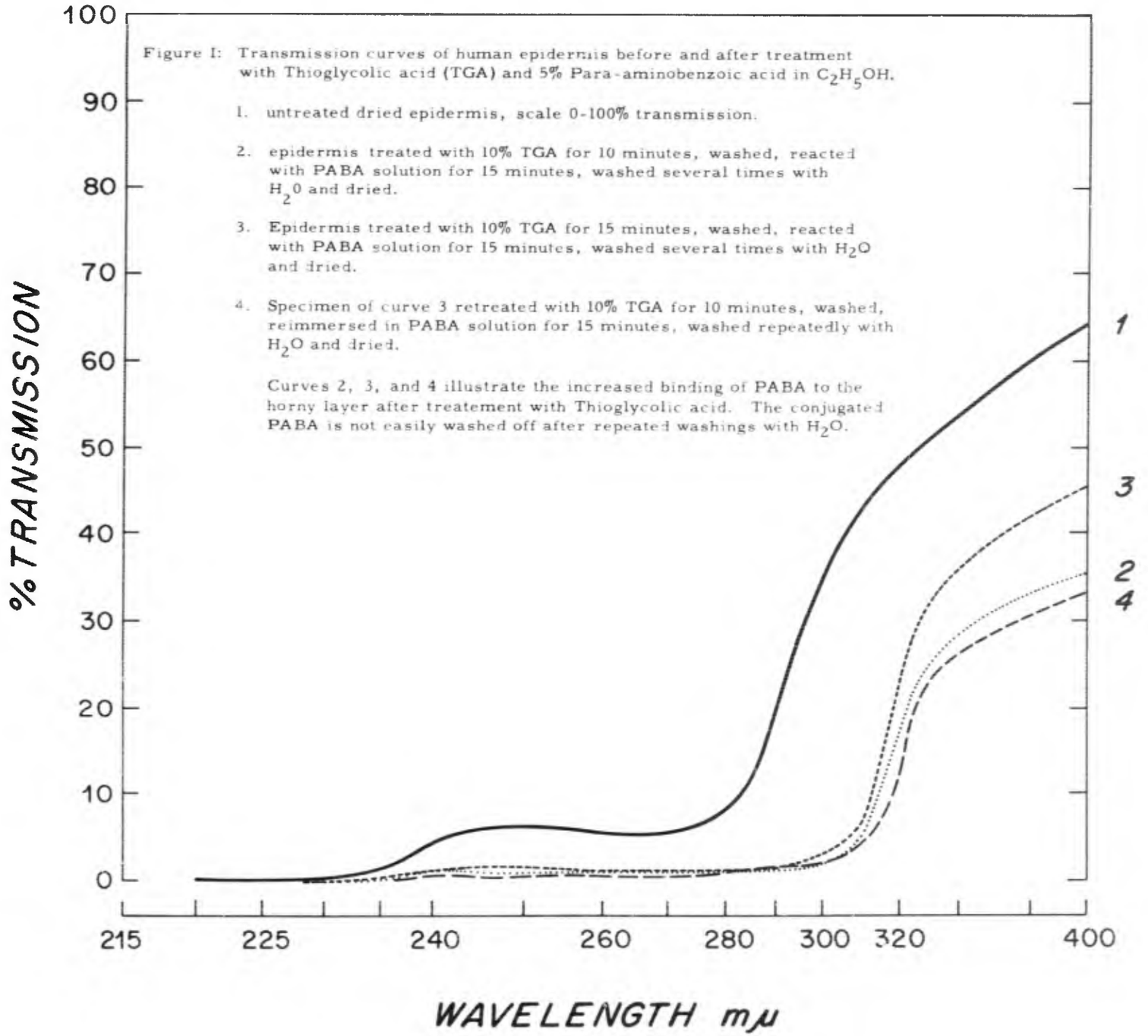


Figure I: Transmission curves of human epidermis before and after treatment with Thioglycolic acid (TGA) and 5% Para-aminobenzoic acid in C_2H_5OH.
1. untreated dried epidermis, scale 0-100% transmission.
2. epidermis treated with 10% TGA for 10 minutes, washed, reacted with PABA solution for 15 minutes, washed several times with H_2O and dried.
3. Epidermis treated with 10% TGA for 15 minutes, washed, reacted with PABA solution for 15 minutes, washed several times with H_2O and dried.
4. Specimen of curve 3 retreated with 10% TGA for 10 minutes, washed, reimmersed in PABA solution for 15 minutes, washed repeatedly with H_2O and dried.
Curves 2, 3, and 4 illustrate the increased binding of PABA to the horny layer after treatement with Thioglycolic acid. The conjugated PABA is not easily washed off after repeated washings with H_2O.
% TRANSMISSION
100
90
80
70
60
50
40
30
20
10
0
1
3
2
4
215
225
240
260
280
300
320
400
WAVELENGTH mμ

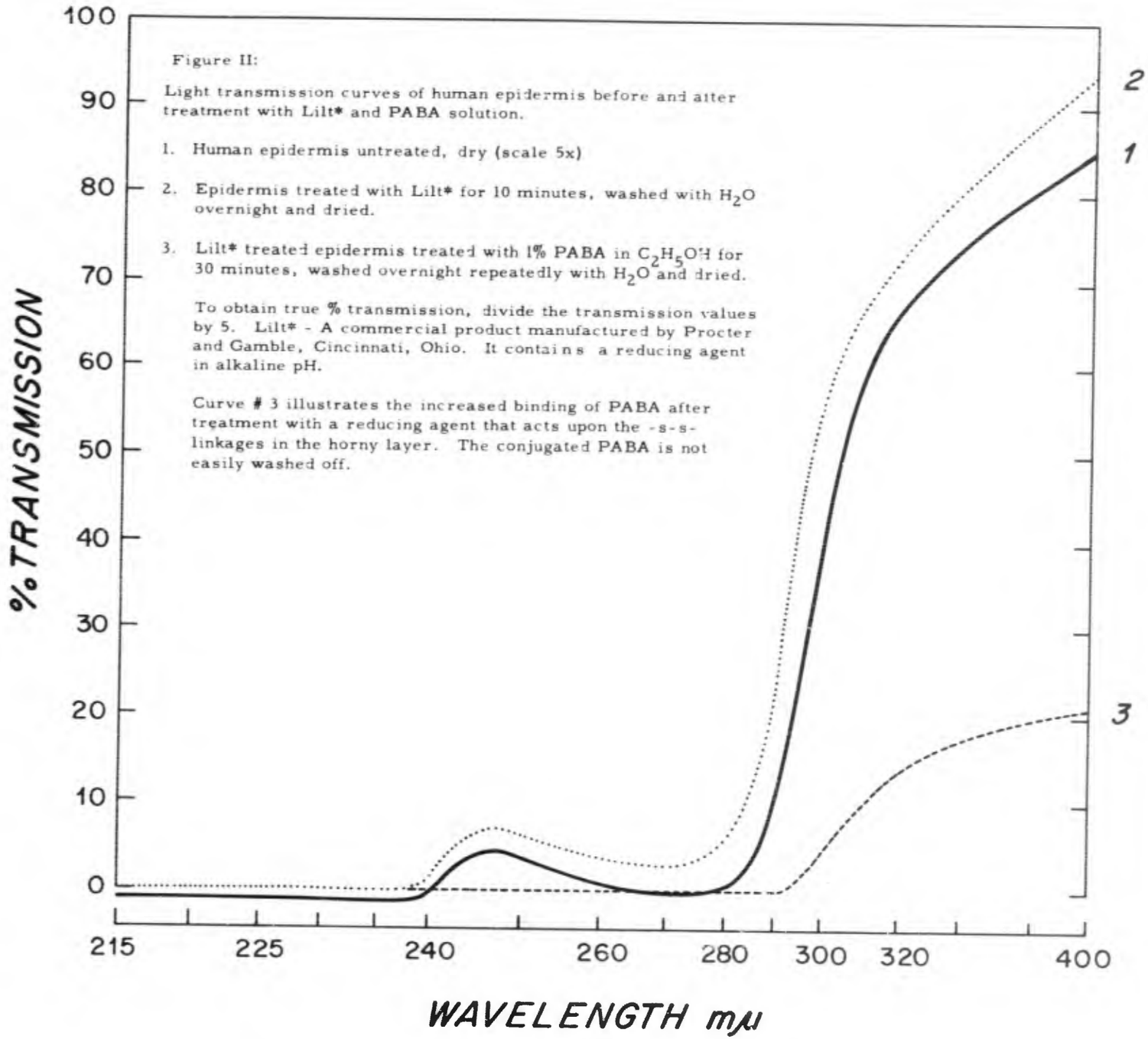


Figure II:

Light transmission curves of human epidermis before and after treatment with Lilt* and PABA solution.

1. Human epidermis untreated, dry (scale 5x)
2. Epidermis treated with Lilt* for 10 minutes, washed with H_2O overnight and dried.
3. Lilt* treated epidermis treated with 1% PABA in C_2H_5OH for 30 minutes, washed overnight repeatedly with H_2O and dried.

To obtain true % transmission, divide the transmission values by 5. Lilt* - A commercial product manufactured by Procter and Gamble, Cincinnati, Ohio. It contains a reducing agent in alkaline pH.

Curve # 3 illustrates the increased binding of PABA after treatment with a reducing agent that acts upon the -s-s- linkages in the horny layer. The conjugated PABA is not easily washed off.

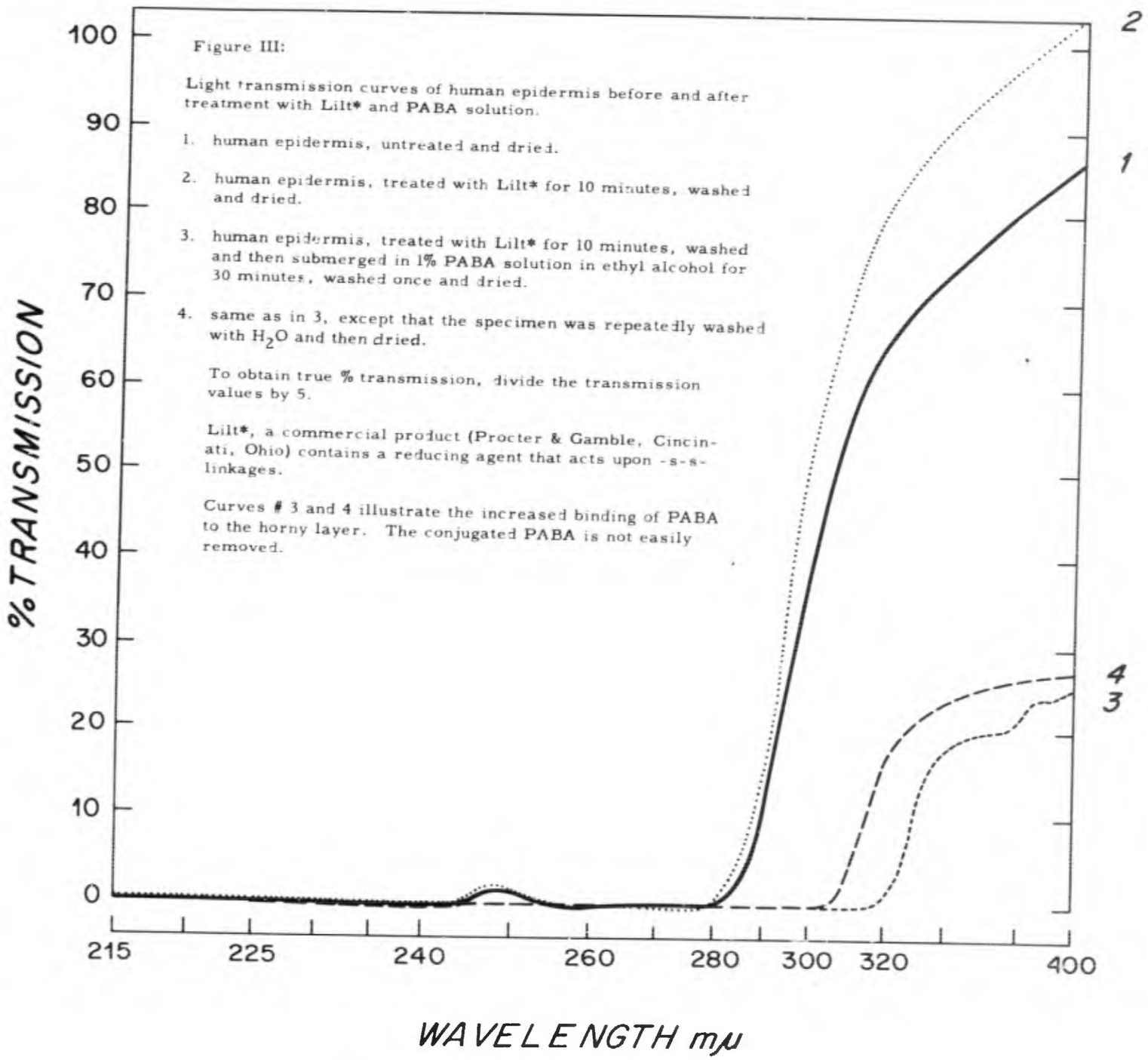


Figure III:
Light transmission curves of human epidermis before and after treatment with Lilt* and PABA solution.
1. human epidermis, untreated and dried.
2. human epidermis, treated with Lilt* for 10 minutes, washed and dried.
3. human epidermis, treated with Lilt* for 10 minutes, washed and then submerged in 1% PABA solution in ethyl alcohol for 30 minutes, washed once and dried.
4. same as in 3, except that the specimen was repeatedly washed with H_2O and then dried.
To obtain true % transmission, divide the transmission values by 5.
Lilt*, a commercial product (Procter & Gamble, Cincinati, Ohio) contains a reducing agent that acts upon -s-s- linkages.
Curves # 3 and 4 illustrate the increased binding of PABA to the horny layer. The conjugated PABA is not easily removed.
% TRANSMISSION
100
90
80
70
60
50
40
30
20
10
0
215
225
240
260
280
300
320
400
WAVELENGTH mμ
1
2
3
4

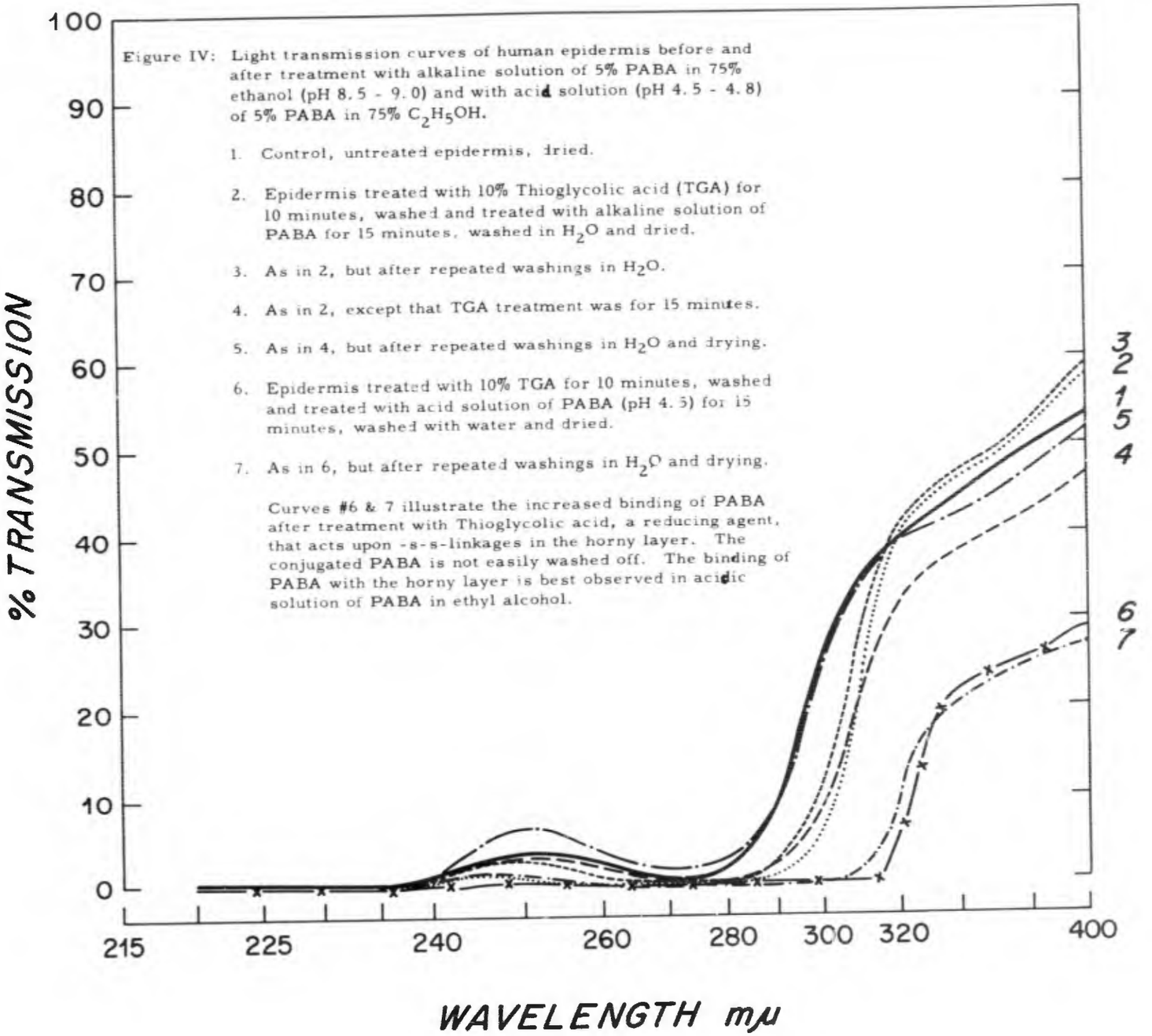


Figure IV: Light transmission curves of human epidermis before and after treatment with alkaline solution of 5% PABA in 75% ethanol (pH 8.5 - 9.0) and with acid solution (pH 4.5 - 4.8) of 5% PABA in 75% C_2H_5OH.

1. Control, untreated epidermis, dried.
2. Epidermis treated with 10% Thioglycolic acid (TGA) for 10 minutes, washed and treated with alkaline solution of PABA for 15 minutes, washed in H_2O and dried.
3. As in 2, but after repeated washings in H_2O.
4. As in 2, except that TGA treatment was for 15 minutes.
5. As in 4, but after repeated washings in H_2O and drying.
6. Epidermis treated with 10% TGA for 10 minutes, washed and treated with acid solution of PABA (pH 4.5) for 15 minutes, washed with water and dried.
7. As in 6, but after repeated washings in H_2O and drying.

Curves #6 & 7 illustrate the increased binding of PABA after treatment with Thioglycolic acid, a reducing agent, that acts upon -s-s-linkages in the horny layer. The conjugated PABA is not easily washed off. The binding of PABA with the horny layer is best observed in acidic solution of PABA in ethyl alcohol.

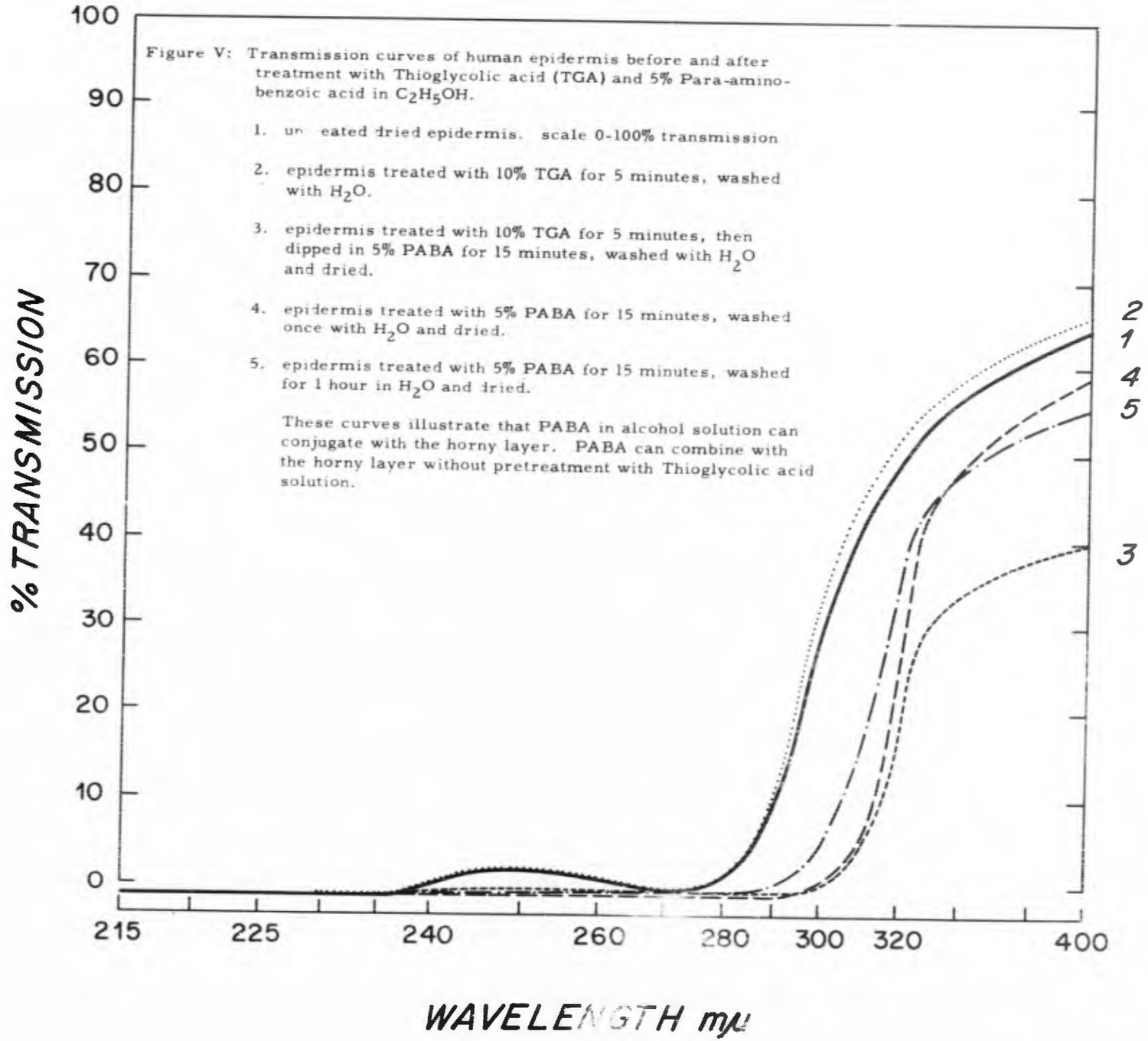


Figure V: Transmission curves of human epidermis before and after treatment with Thioglycolic acid (TGA) and 5% Para-amino-benzoic acid in C_2H_5OH.

1. un eated dried epidermis. scale 0-100% transmission
2. epidermis treated with 10% TGA for 5 minutes, washed with H_2O.
3. epidermis treated with 10% TGA for 5 minutes, then dipped in 5% PABA for 15 minutes, washed with H_2O and dried.
4. epidermis treated with 5% PABA for 15 minutes, washed once with H_2O and dried.
5. epidermis treated with 5% PABA for 15 minutes, washed for 1 hour in H_2O and dried.

These curves illustrate that PABA in alcohol solution can conjugate with the horny layer. PABA can combine with the horny layer without pretreatment with Thioglycolic acid solution.

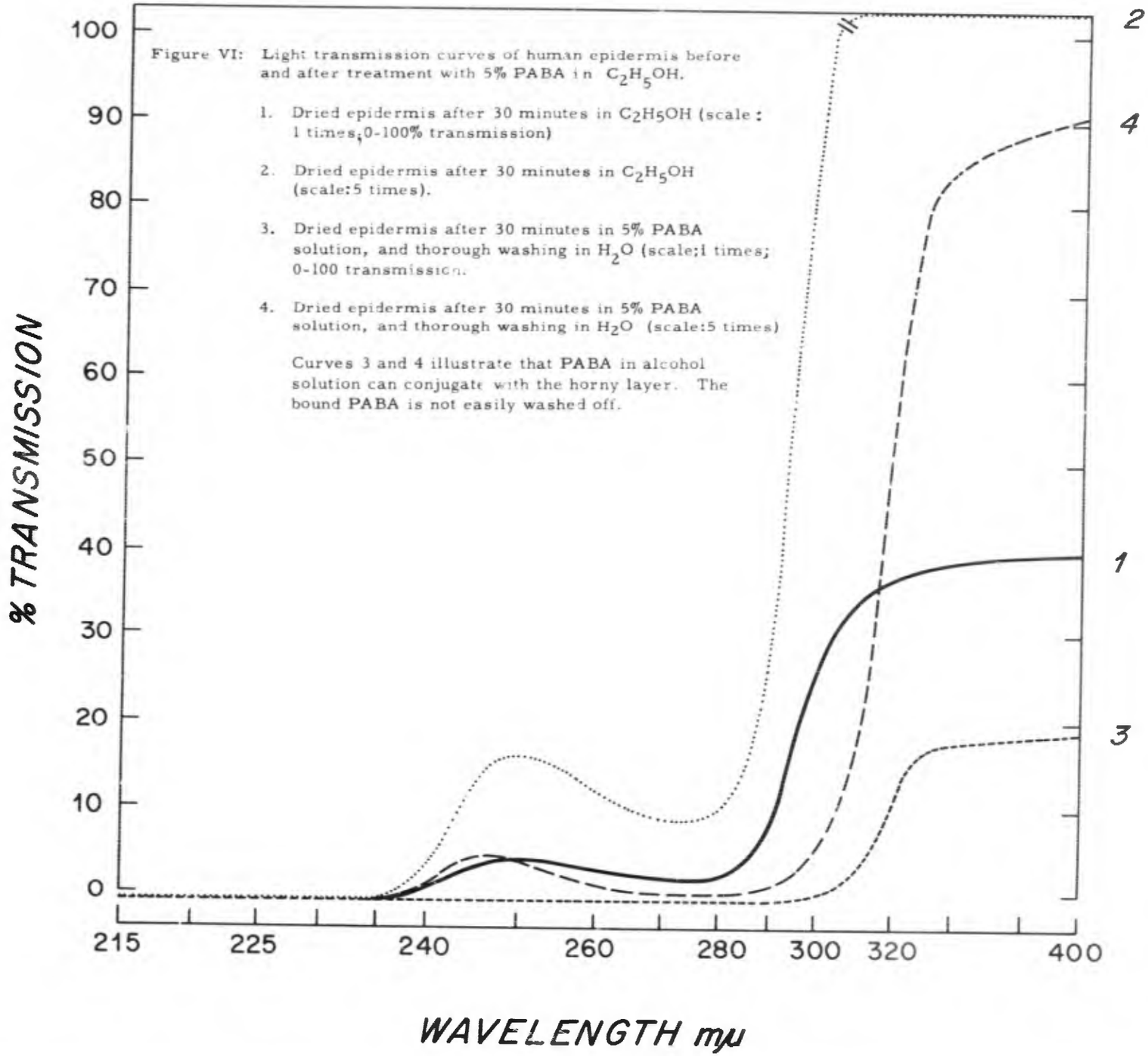


Figure VI: Light transmission curves of human epidermis before and after treatment with 5% PABA in C_2H_5OH.
1. Dried epidermis after 30 minutes in C_2H_5OH (scale: 1 times; 0-100% transmission)
2. Dried epidermis after 30 minutes in C_2H_5OH (scale:5 times).
3. Dried epidermis after 30 minutes in 5% PABA solution, and thorough washing in H_2O (scale;1 times; 0-100 transmission.
4. Dried epidermis after 30 minutes in 5% PABA solution, and thorough washing in H_2O (scale:5 times)
Curves 3 and 4 illustrate that PABA in alcohol solution can conjugate with the horny layer. The bound PABA is not easily washed off.
% TRANSMISSION
100
90
80
70
60
50
40
30
20
10
0
215
225
240
260
280
300
320
400
WAVELENGTH mμ
1
2
3
4

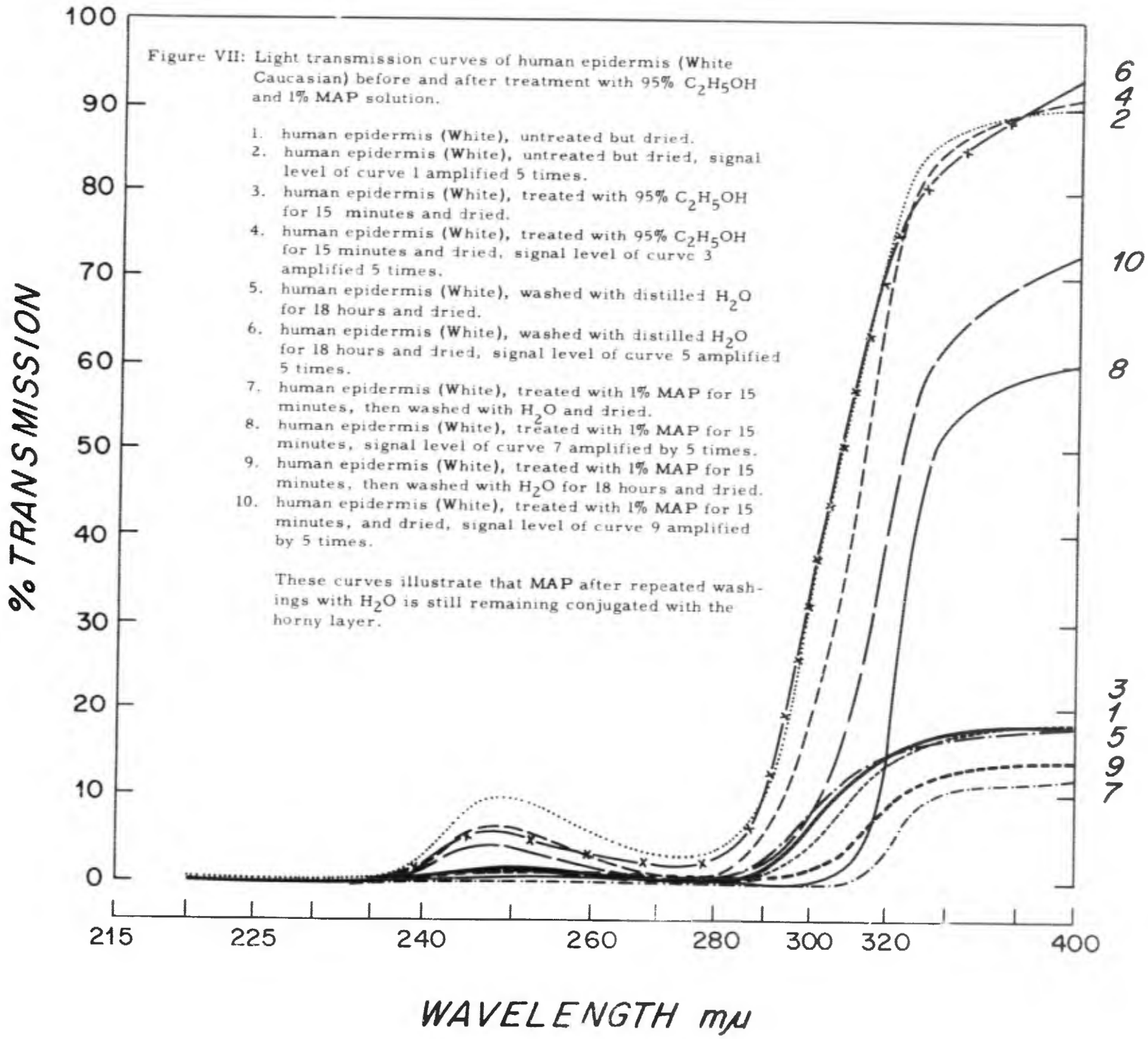


Figure VII: Light transmission curves of human epidermis (White Caucasian) before and after treatment with 95% C_2H_5OH and 1% MAP solution.

1. human epidermis (White), untreated but dried.
2. human epidermis (White), untreated but dried, signal level of curve 1 amplified 5 times.
3. human epidermis (White), treated with 95% C_2H_5OH for 15 minutes and dried.
4. human epidermis (White), treated with 95% C_2H_5OH for 15 minutes and dried, signal level of curve 3 amplified 5 times.
5. human epidermis (White), washed with distilled H_2O for 18 hours and dried.
6. human epidermis (White), washed with distilled H_2O for 18 hours and dried, signal level of curve 5 amplified 5 times.
7. human epidermis (White), treated with 1% MAP for 15 minutes, then washed with H_2O and dried.
8. human epidermis (White), treated with 1% MAP for 15 minutes, signal level of curve 7 amplified by 5 times.
9. human epidermis (White), treated with 1% MAP for 15 minutes, then washed with H_2O for 18 hours and dried.
10. human epidermis (White), treated with 1% MAP for 15 minutes, and dried, signal level of curve 9 amplified by 5 times.

These curves illustrate that MAP after repeated washings with H_2O is still remaining conjugated with the horny layer.

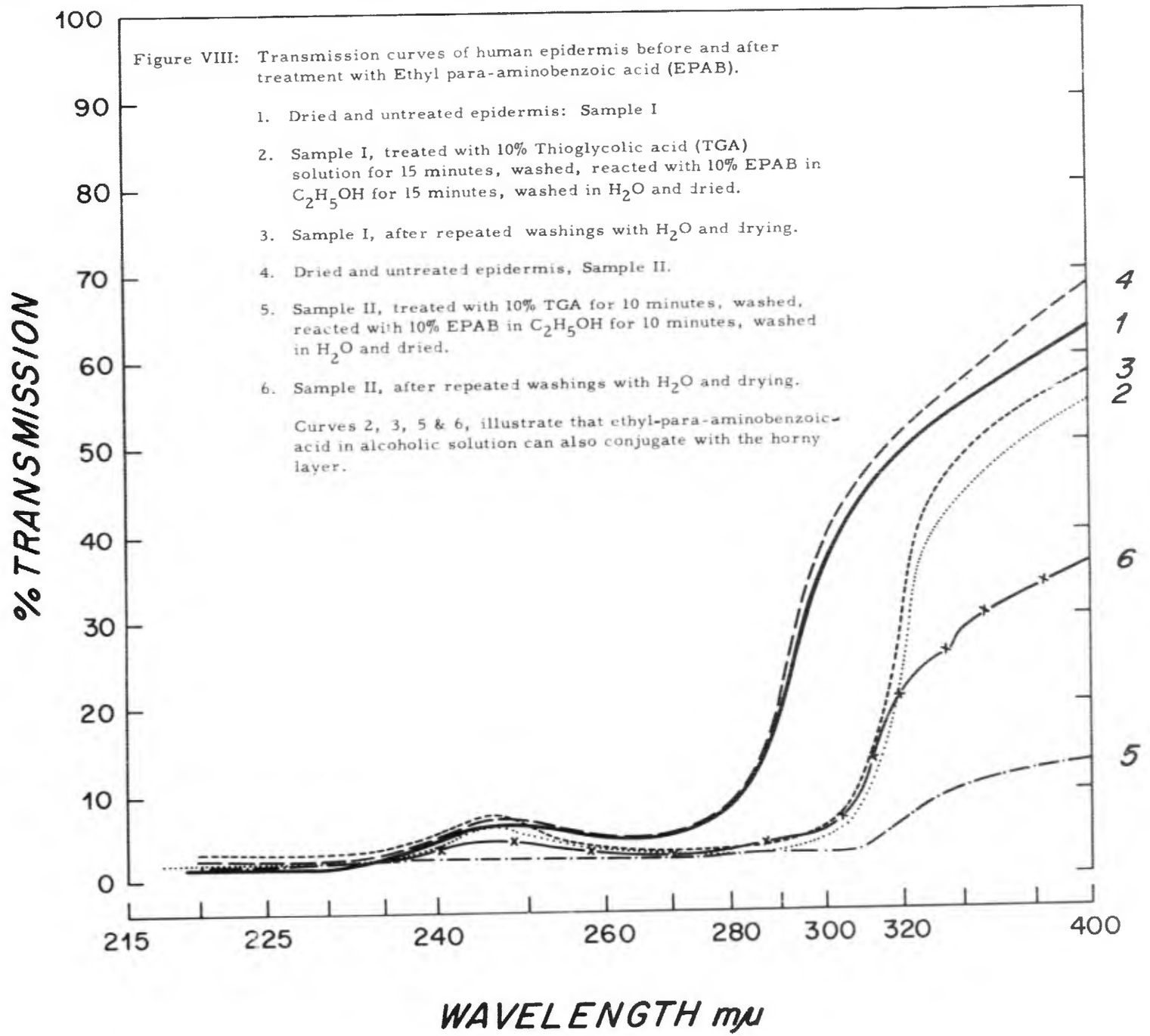


Figure VIII: Transmission curves of human epidermis before and after treatment with Ethyl para-aminobenzoic acid (EPAB).

1. Dried and untreated epidermis: Sample I
2. Sample I, treated with 10% Thioglycolic acid (TGA) solution for 15 minutes, washed, reacted with 10% EPAB in C_2H_5OH for 15 minutes, washed in H_2O and dried.
3. Sample I, after repeated washings with H_2O and drying.
4. Dried and untreated epidermis, Sample II.
5. Sample II, treated with 10% TGA for 10 minutes, washed, reacted with 10% EPAB in C_2H_5OH for 10 minutes, washed in H_2O and dried.
6. Sample II, after repeated washings with H_2O and drying.

Curves 2, 3, 5 & 6, illustrate that ethyl-para-aminobenzoic-acid in alcoholic solution can also conjugate with the horny layer.

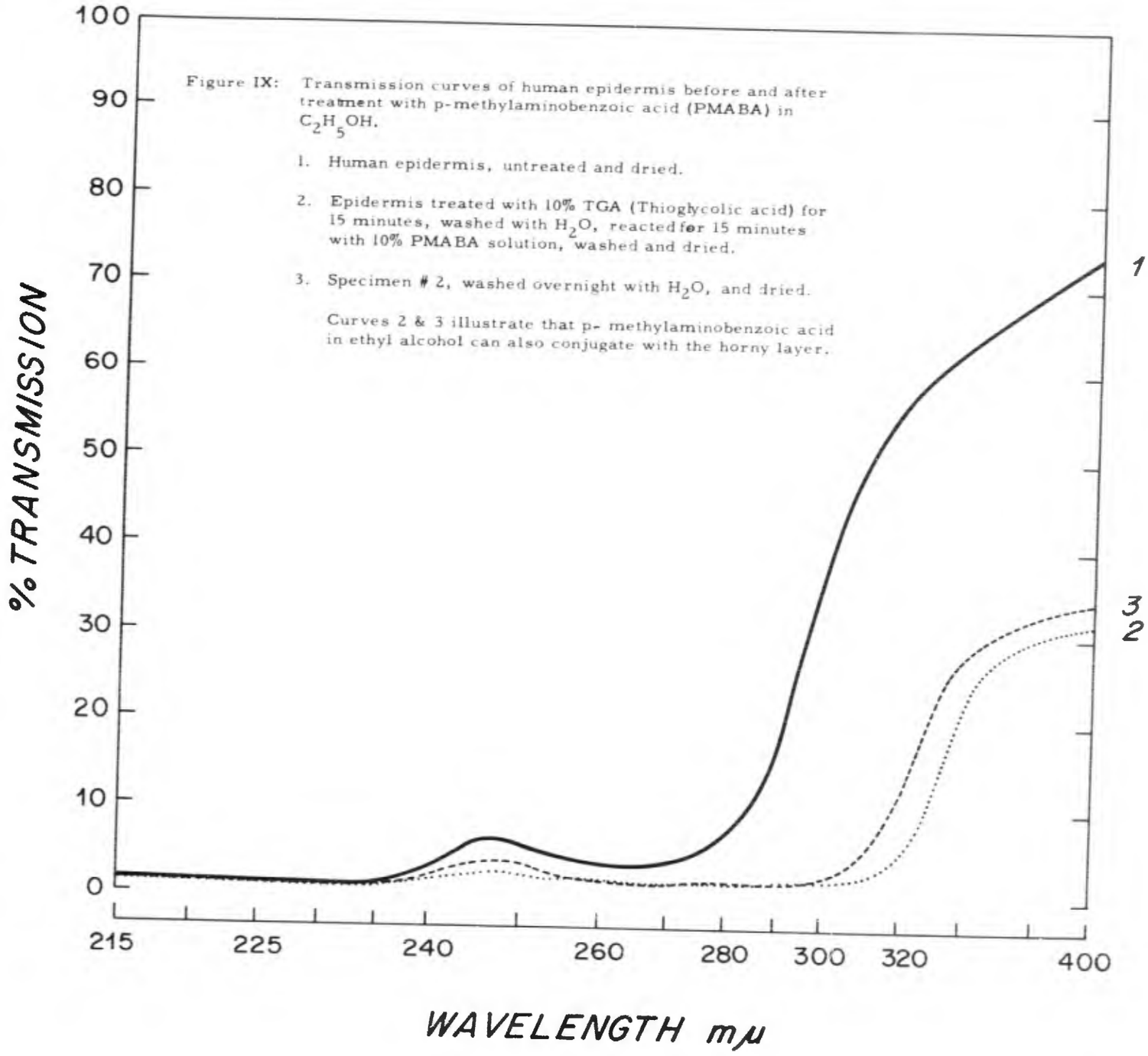


Figure IX: Transmission curves of human epidermis before and after treatment with p-methylaminobenzoic acid (PMABA) in C_2H_5OH.

1. Human epidermis, untreated and dried.
2. Epidermis treated with 10% TGA (Thioglycolic acid) for 15 minutes, washed with H_2O, reacted for 15 minutes with 10% PMABA solution, washed and dried.
3. Specimen # 2, washed overnight with H_2O, and dried.

Curves 2 & 3 illustrate that p- methylaminobenzoic acid in ethyl alcohol can also conjugate with the horny layer.

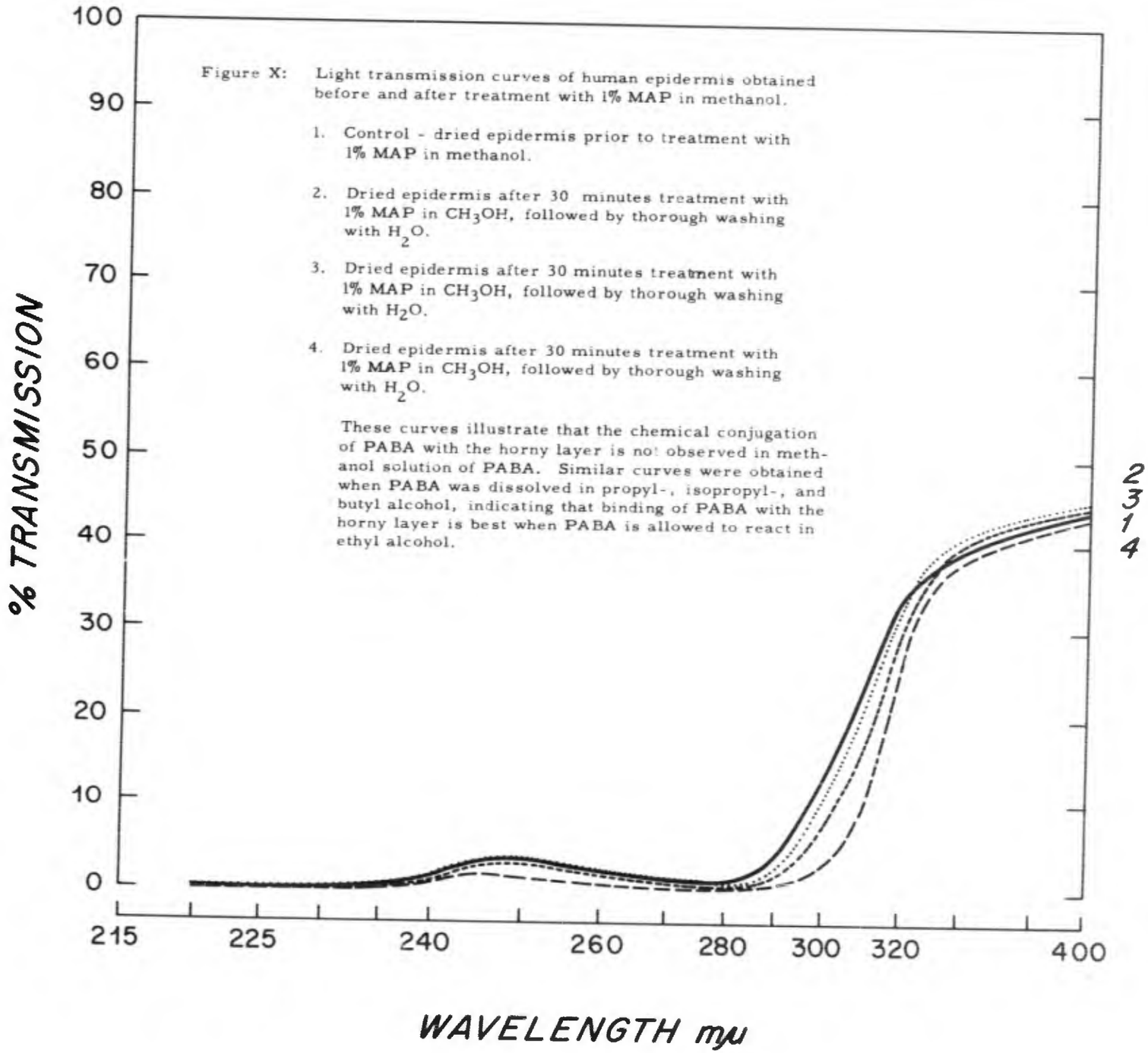


Figure X: Light transmission curves of human epidermis obtained before and after treatment with 1% MAP in methanol.

1. Control - dried epidermis prior to treatment with 1% MAP in methanol.
2. Dried epidermis after 30 minutes treatment with 1% MAP in CH_3OH, followed by thorough washing with H_2O.
3. Dried epidermis after 30 minutes treatment with 1% MAP in CH_3OH, followed by thorough washing with H_2O.
4. Dried epidermis after 30 minutes treatment with 1% MAP in CH_3OH, followed by thorough washing with H_2O.

These curves illustrate that the chemical conjugation of PABA with the horny layer is not observed in methanol solution of PABA. Similar curves were obtained when PABA was dissolved in propyl-, isopropyl-, and butyl alcohol, indicating that binding of PABA with the horny layer is best when PABA is allowed to react in ethyl alcohol.

Since the maximum absorption of PABA, as well as other derivatives of PABA, lies in range of 250-310 mμ, advantage was taken of this light absorption band to assess the chemical binding of PABA and its derivatives with the horny layer. Attempts to estimate the exact quantity of bound PABA, either by colorimetric or by titrimetric methods were unrewarding because the methods were not sufficiently sensitive to detect the micro quantity of the bound PABA. However, the chemical conjugation of PABA, with the horny layer was established by the following criteria:

a. By repeated washings in distilled water. Free PABA is easily washed off from the surface of the skin by rinsing the samples in distilled water. Chemically conjugated PABA can then be detected by determining the increased ultraviolet light absorption in the 250-310 mμ range.

b. The bound PABA cannot be removed easily by extraction with ether at acidic pH (pH 4.0, approximately the isoelectric point of free PABA) except after acid or alkaline hydrolysis.

c. The chemically conjugated PABA that is not removed from the surface of the horny layer after repeated washings in H_2O, can be removed by treating these specimens with alkaline or acidic solutions. The process of weak hydrolysis removes the bound PABA. This is ascertained by studying the light transmission spectra of human epidermis or stratum corneum before and after hydrolysis in alkaline or acidic solutions.

d. In addition, it is well known that the PABA solution has a higher extinction-coefficient in basic solution than in the acid solution. In basis or neutral solution (in H_2O or in NaCl), PABA exhibits a strong absorption in 250-310 mμ range, with a broad absorption maximum in 270-300 mμ range. In acidic pH (4.0), the position of the absorption maximum of PABA, as with other benzenoid compounds having ionizing groups, is shifted. Ultraviolet absorption above 290 mμ is diminished significantly and the absorption maximum appears at 265-267 mμ. By studying the light transmission spectra of human epidermis in these manners, one can assess the nature of PABA binding.

In figures I - X, the light transmission spectra of human epidermis and stratum corneum are presented to illustrate that PABA can conjugate chemically with the horny layer. This conjugation was observed only in alcoholic solution of PABA. It was not observed with ortho-, or meta-substituted amino acid derivatives of benzoic acid. Initially, **the pretreatment** of the horny layer with thioglycolic acid was investigated with the hope that the reduced thiol groups in the horny layer will provide additional sites for chemical conjugation of PABA. As illustrated in figures I - V, it is apparent that PABA is combining with the horny layer. This pretreatment with thioglycolic acid was however, found to be not absolutely essential. In absence of this treatment with thioglycolic acid, PABA still can conjugate with the horny layer. This is illustrated in figures V - VII. The chemical binding of PABA with horny layer was most effective in ethanolic solution. This chemical conjugation was less when PABA was dissolved in methenol, propenol, n. butyl alcohol and isobutyl alcohol (Figure X). Neither the sodium and potassium salts of PABA, nor the glyceryl ester PABA reveals any specific chemical binding with the horny layer.

This chemical conjugation of PABA is also inhibited by the presence of emollients. For example, addition of 1% (w/v) of cetyl alcohol in 1-5% PABA solution, significantly minimizes the affinity of PABA to bind with the horny layer. No residual PABA was left conjugated with the horny layer after the treated specimens were subjected to one or two washings in water. Likewise, the presence of 3% lanolin, 4% isopropyl myristate in alcoholic solution of PABA, significantly retarded the binding of PABA to the stratum corneum.

These experiments also gave us one additional piece of information. The pretreatment of horny layer with thioglycolic acid is not essential for promoting the conjugation of PABA. PABA in ethanol solution alone is able to conjugate with the horny layer.

The data presented in figures 1-10 represent a few illustrative examples. Additional studies with a number of other chemical agents (e.g. hydroquinone, 2-hydroxy-1,4-naphthoquinone, psoralens or furocoumarins e.g. psoralen, 4, 5, 8- trimethyl psoralen and 8-methoxy psorelan; and other light absorbing molecules such as benzophenone and several of its derivatives), were carried out to understand the nature of promoting chemical binding of compounds with the horny layer. It was apparent that under physiological conditions it is very difficult to promote chemical conjugation of chemical agents with the horny layer. In most instances our attempts to promote binding of chemical agents with the horny layer were unrewarding. However, compounds such as 2-hydroxy-1,4-naphthoquinone; 5-hydroxy-1,4-naphthoquinone, and 1,2-benzenediol (pyrocatechol) can be conjugated with the horny layer. This binding is facilitated in the acidic range (pH 5-6). 2-hydroxy-1, 4-naphthoquinone and 5-hydroxy-1,4-naphthoquinone give an orange red color to the horny layer. These agents can firmly conjugate with the stratum corneum and are not removed by repeated washings in water. However, they are not able to afford any protection against sunburn radiation. The transmission curves of the epidermis in 290-310 mμ range before and after treatment with these two naphthoquinone derivatives were practically identical. The treated epidermis specimens, however, showed increased absorption of light in the 320-500 mμ wavelength range.

Except for dihydroxy acetone, which presumably combines with the histidine of the keratin, these other agents do not seem to protect individuals against sunburn radiation. The color produced by binding 2-hydroxy-1,4-naphthoquinone to the stratum corneum is quite stable for the first 5 to 7 days when applied to the back skin areas of the individuals and is retained under conditions of washing with water and soap. The color of the naphthoquinone however, gradually fades away after repeated daily washings. This fading is related to the normal desquamation of the horny cells and also to the hydrolytic action of soap and water washings. The hue or the color that develops on the stratum corneum after application of these naphthoquino and dihydroxyacetone solutions, although not natural, may have some practical clinical usefulness. One such use should be explored in masking the color of the hypopigmented macules of the patients with vitiligo.

The data in this section allow us to speculate how and why the preparation MAP appears to be most effective in protecting skin against solar radiation.

1. The preparation MAP affords protection in two or three ways when applied to the skin and subjects exposed to solar radiation without any swimming or exercise.

a. Physical screen: The dried film of PABA absorbs sunburn radiation at the surface of the horny layer and prevents the passage of this harmful radiation to the underlying viable cells of epidermis.

b. Chemical screen: In addition to being chemically conjugated, PABA appears to be adsorbed on the surface of the horny layer. The adsorbed PABA is not easily eluted by water, but after repeated washings, it can be eluted. Thus, the adsorbed PABA, as well as the conjugated PABA affords protection by absorbing the sunburn radiation and preventing its transmission to the vulnerable cells of underlying epidermis.

2. Under conditions of swimming and exercise accompanied by sweating, the preparation MAP affords protection because it is either adsorbed or chemically conjugated with the horny layer. In the three different clinical trials it was established that the preparation MAP tends to remain on the surface of the horny layer even after the volunteers are subjected to a schedule of swimming or exercise. The subjects showed very satisfactory protection up to a period of 90-120 minutes of solar radiation. This protection cannot just be due to a physical screen of PABA (a dried film). Other commercially available agents are easily eluted or washed off from the surface of the skin. It is obvious that the preparation MAP must be chemically conjugating with the horny layer. Also the prolonged protective effect observed at 24 and 48 hours after a single application of MAP preparation (by exposing volunteers to natural sunlight as well as to artificial ultraviolet light source), undoubtedly suggests that PABA is chemically conjugating with the horny layer.

We postulate that PABA is conjugated to the horny layer through its amino group. This conjugated form of PABA may be liberated by hydrolytic reactions. It is also possible that the carboxyl group of PABA is bound to an acidic polypeptide of the keratin present in the horny layer and aryl amino group is free.

Our efforts to promote chemical binding of MAP or MAP (D5) to the stratum corneum with a hope that a single application of MAP would afford a long-term protection (at least for a period of 5-7 days) were, however, unsuccessful. The problems in this type of ideal approach are manifold. Primarily one has to bear in mind that this type of chemical conjugation has to be pursued under physiological conditions on a living skin. Secondly, the chemical conjugation of the horny layer with the exogenous agent has to be a stable type, not easily dissociated with the hydrolytic action of water, soap and alkali. Thirdly, the horny cells are constantly desquamated and shed. Therefore, a permanent film of light absorbing molecules chemically conjugated with the horny cells appear to be unlikely. Never the less, the results obtained with the in vitro as well as the in vivo studies reveal that preparation MAP or MAP (D5) tends to remain on the skin even after bathing, swimming and profuse perspiration and affords partial if not complete protection.

The Approach to the Problem of Sunburn (continued)

Phase II: Clinical Trials

After assuring ourselves that MAP was most effective in inhibiting the erythema elicited by ultraviolet radiation from artificial light sources, we undertook 4 clinical trials. Three were carried out at the Arizona State Prison, Florence, Arizona and the fourth was conducted by Dr. Edgar Frenk on the snow covered Swiss Alps. Dr. Frenk had obtained necessary knowledge and experience when he collaborated with us in the first clinical trial of April, 1966 at Florence, Arizona.

The efficacy of any sunscreen preparation can only be claimed when the final product is evaluated under natural outdoor conditions that simulate normal usage. This experience has been of great value for us. We have not only established the efficacy of the test products under varied conditions, but most important, we have gained fundamental knowledge in the field of cutaneous photobiology and melanogenesis. From the microscopic observations at the tissue level to the cellular alterations at the microscopic and electron microscopic level, we have gained insight into the process of sunburn and suntan. The objectives, the protocol for each experiment, the results and the conclusions are presented separately for each clinical trial.

Criteria for the gradation, by visual inspection and measurement of skin reflectance, of the erythema induced by solar radiation or by ultraviolet radiation from artificial sources: The degree of erythema in each test and the control sites (exposed as well as the unexposed) was evaluated between 20 - 24 hours after irradiation by two criteria.

1. By means of a photo-electric reflectance meter (Model 610, Photo Volt Corporation, New York), equipped with a green tristimulus filter whose maximum transmittance was between 545 and 550 mu, i.e. in the absorption range of hemoglobin. With the increased vasodilation, the reflectivity of the skin decreases because of the increased absorption of the impinging light by hemoglobin present in the dilated and damaged arterioles, venules and capillaries. With the filter in the probe, a block of pure white magnesium carbonate was used to calibrate 100% reflectance readings. Per cent reflectance readings were obtained for each control site (both the sun exposed and the unexposed skin adjacent to the test sites) and for each exposed site receiving test preparations. The difference between reflectance readings from control sites (unexposed site) and reflectance readings from irradiated test sites shows the degree of erythemal response.

The Approach to the Problem of Sunburn (continued)

2. The second criterion for the gradation of erythema was by visual inspection. An arbitrary scale ranging from 0 to ++++ was assigned in the following way:

a) 0..... No detectable redness or erythema

b) ±..... Bareley perceptible erythema detected by blanching of the skin

c) +..... Pink erythema, definite but weak

d) ++..... Red erythema, moderate sunburn

e) +++..... Violaceous, red erythema, strong sunburn

f) ++++..... Deep red erythema with edema, marked tenderness of skin and/or vesiculation; very strong sunburn

The degree of erythema was evaluated independently by two investigators. Before assigning the final score, each evaluator had to agree.

In a number of instances, immediate pigment darkening response (IPD) was found to complicate the reflectance readings. The ⅝ reflectance readings under an IPD response were evaluated indirectly (by knowing the maximum IPD response in the MAP treated exposed skin that had no erythema).

ARIZONA STATE PRISON CLINICAL TRIAL

APRIL 1966

The first clinical trial at the Arizona State Prison was carried out in the last week of April, 1966. The following preparations were evaluated:

1. 5% MAP
2. 22 commercially available sunscreen preparations.
3. Preparation described by Fusaro, Runge & Watson (Arch. Derm. 93:106, 1966).

In this clinical trial, the above listed preparations were evaluated under five different conditions:

1. Evaluate the degree of protection after a single application of the test preparations on backs and legs without subjecting the volunteers to any activity, except lying in the prone position during sun exposure.
2. After a single application of the test preparations, remove the applied products either by manual washing by applying water soaked cotton gauzes or asking the volunteers to take a shower.
3. Evaluate the degree of protection after a single application of the test preparations and subjecting the volunteers to a schedule of heavy exercise that results in profuse sweating under dry and hot weather conditions.
4. Evaluate the degree of protection after 10, 15 and 20 minutes of swimming.
5. Evaluate the protective effect of MAP under normal conditions of use (e.g. walking and running around in the sun, lying on the back and prone position, and sitting in the sun) under very humid conditions.

Experiment # 1 was designed to demonstrate the prolonged protective effect of MAP against solar radiation on subjects who can easily sunburn.

Experiment # 2 was designed to compare preparation MAP against several commercial preparations. The primary objective was to ascertain the affinity of MAP and other commercial products to remain on the horny layer after washing with water.

Experiment # 3 was designed to compare the efficacy of MAP with several commercially available products after subjecting the volunteers to a schedule of exercise under hot and dry weather conditions. The principal objective was to ascertain whether the applied test preparations can remain on the horny layer after sweating is induced and the skin comes in frictional contact with the clothes.

Experiment # 4 was designed to compare MAP against Skolex, UVAL and RVP (commercial products) under normal conditions of usage.

Experiment # 5 was designed to ascertain the effect of swimming in fresh water on sunburn-protecting properties of MAP.

Experiment # 6 was designed to compare MAP against a sunscreen preparation described by Fusaro, Runge and Watson (Arch. Dermatology, 93:106, 1966).

Comments & Conclusions:

1. MAP, the sunscreen preparation formulated in our laboratory, is not only more effective than any commercially available product, but also the best that can be recommended to protect against sunburn radiation.

2. After a single application and without washing with water, most of the 20 leading commercial preparations were found to be either very poor in affording protection greater than 60 minutes of exposure to sunlight or not as effective as MAP. At best, the commercial preparations afforded protection against radiation sunburn for a period of 60 minutes.

3. MAP on the other hand, under similar conditions, was found to exert 100% protection up to 4 hours of sun exposure.

4. When volunteers were subjected to sweating as a result of moderate to heavy exercise (calesthenics, running, playing basketball, etc.), the 20 sunscreen preparations that were obtained commercially were found to be washed off and afforded almost no protection even for short periods of exposure ranging from 30, 60 and 120 minutes. MAP, however, was found to protect 100% up to 2 hours sun exposure, and over 75% protection (barely detectable erythema) after 2 hours of sun exposure. In fact, in all but the easy burning fair-skinned individuals, MAP afforded complete protection up to 4 hours of sun exposure.

   When the volunteers were subjected to a schedule of 15 and 30 minutes swimming, following an application of MAP or other known sunscreen preparations, and then exposed to solar radiation for a period ranging from 15 minutes to 2 hours, all of the commercial preparations were found to be washed off and afforded practically no protection. MAP, on the other hand, gave a complete protection to most of the subjects for a 60 minute period. The two hour exposure did cause mild erythema which was, however, devoid of any tenderness and edema. In this experiment, the control areas that were not treated with any sunscreen preparation were markedly tender, edematous, painful and remarkably erythematous. The data obtained in this study suggest that as a result of swimming, the molecules of MAP that were applied topically and failed to react with the proteins of stratum corneum, mainly the keratins, were washed off. There were, however, enough light absorbing MAP

molecules that remained either adsorbed or bonded chemically that afforded protection to the skin against sunburn radiation.

A single application of MAP, if allowed to remain on the back of the individual, without letting the subject bathe, will protect for at least a period of 48 hours (protection up to 12 to 15 minimal erythema doses which would roughly be equal to at least 3 hours of sun exposure). This has been ascertained by irradiating the subjects under artificial ultraviolet light sources as well as the natural sunlight. After a period of 48 hours, the subject remains protected up to 96 hours, however, the degree of protection diminishes gradually (protection up to 4-6 MED doses) and by the end of 120 hours, the subject is no longer exhibiting any protective action of MAP that was applied 5 days earlier.

ARIZONA STATE PRISON CLINICAL TRIAL

April 1966

Experiment 1: To demonstrate the prolonged protective effect of MAP against solar radiation on subjects who can easily sunburn.

Objective: To compare the protective effect of MAP and a leading commercially available sunscreen preparation Sea & Ski (S & S) under the following conditions:

a) Continuous prone position of the subject during irradiation.
b) Sun exposure 1 to 4 hours (11 a.m. to 3 p.m.)
c) No exercise and sweating
d) No swimming
e) Clear hot and dry day

No. of Subjects: 2

Protocol:

Back.....(lower 2/3 region)
Left side...1 application of MAP
Right side..1 application of Sea & Ski
Control.....upper 1/3 back, no treatment
Legs.....popliteal region. 1 application of MAP on both legs.
Untreated control area was also improvised on each leg.

Schedule:

Tuesday, April 26: Left leg 1 application of MAP
Right leg 1 " of S & S

9:30 a.m. Application of MAP & Sea & Ski in the designated areas
10:00 - 10:30 a.m. Lunch (subjects remain indoors without shirt)
11:00 a.m. Backs of the subjects exposed in the prone position. MAP treated & Sea & Ski treated areas were divided into 3 sections:

Section 1 ..... 1 hour sun exposure 11 a.m. - 12:00 Noon
Section 2 ..... 2-1/2 hours sun " 11 a.m. - 1½ p.m.
Section 3 ..... 4 hours sun exposure 11 a.m. - 3 p.m.
CONTROL (untreated) skin received 1 hour sun exposure 11 a.m. - 12:00 Noon

Both right and left legs received 2-1/2 hours sun exposure.
Control (untreated areas) on each leg received 1 hour sun exposure.

In order to avoid the dangers of excessive sunburn, subjects #1 & 2 received only 1 hour sun exposure in the control untreated areas.

Wednesday, April 27:

1. 24 hours after sun exposure the degree of erythema in each area was evaluated by
   a) reflectance readings
   b) grading of visual erythema

Experiment 1: (Continued)

2. Photographs were obtained

Results: Both subjects that were selected for this experiment were fair skinned freckled individuals who could easily sunburn. The control untreated areas that received 1 hour sun exposure revealed marked erythema that ranged from +++ to ++++ intensity.
The Sea & Ski treated areas revealed no protection against 1, 2-1/2 and four hours sun exposure. All these areas were very tender and showed marked erythema. MAP treated areas, on the other hand, were remarkably protected. There was no evidence of perceptible redness in the areas receiving 1 & 2-1/2 hours sun exposure. There was just a barely perceptible redness in area receiving 4 hours sun exposure.

Conclusions: Preparation MAP affords complete protection up to 4 hours of continuous sun exposure.
Considering the fact that this exposure was given in the Arizona desert towards the end of April, and realizing that the minimal erythema dose (MED) for these subjects was approximately 10 minutes of sun exposure, this means that the preparation MAP, when applied only once and left on the surface of the skin, without washing, provides more protection than any other known commercial preparation including Coppertone, Sea & Ski, UVAL & Solbar (see results of 1967 & 1968 Clinical trials) that have been claimed to be the best.

EXPERIMENT 1: Comparison of MAP and a Commercially available sunscreen preparation Sea and Ski (S & S) to demonstrate the prolonged protective effect of MAP

Subject	Site	Preparation	Period of Sun Exposure (hours)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
1	Back	CONTROL	1 hour	+++	0	13	0	Marked tenderness
	"	MAP	1 hour	0	100	1	100	
	"	S & S	1 hour	+++	0	13	100	
	Legs	CONTROL	1 hour	+++	0	14	0	Very tender
	"	MAP	2-1/2 hours	±	80	2	84	
	"	S & S	2-1/2 "	+++	0	13	0	
	Back	MAP	2-1/2 "	0	100	1	100	
	"	S & S	2-1/2 "	+++	0	12	0	Marked tenderness, edema
	"	MAP	4 hours	±	80	3	75	
	"	S & S	4 "	++++	0	12	0	Marked tenderness, edema swelling

EXPERIMENT 1: Comparison of MAP and a Commercially available sunscreen preparation Sea and Ski (S & S) to demonstrate the prolonged protective effect of MAP

Subject	Site	Preparation	Period of Sun Exposure (hours)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
2	Back	CONTROL	1 hour	+++	0	17	0	
	"	MAP	1 "	0	100	1	100	
	"	S & S	1 "	++	25	15	10	
	Legs	CONTROL	1 "	+++	0	16	0	
	"	MAP	1 "	0	100	2	100	
	"	S & S	1 "	+++	0	17	0	
	Back	MAP	2-1/2 hours	0	100	1	100	
	"	S & S	2-1/2 "	++++	0	19	0	Marked tenderness
	"	MAP	4 hours	±	80	4	78	
	"	S & S	4 "	++++	0	18	0	Marked tenderness, edema & micro blisters

ARIZONA STATE PRISON CLINICAL TRIAL

APRIL, 1966

Experiment 2: Comparison of MAP and other commercial preparations under laboratory conditions using hot quartz mercury arc lamp.

Objective:

1) To compare the efficacy of MAP against popular commercial products.
2) To ascertain their affinity to remain on the horny layer after washing with water.

Schedule:

a) A single application of the product on the back in 1.5" x 1.5" area. Several products could thus be applied on the back and compared against control (untreated) sites after ultraviolet exposure.
b) After application, remove the applied products either by washing with water (manually by applying water soaked cotton gauzes) or asking the volunteers to take a shower without applying any soap.
c) Expose the treated and the untreated control sites to varying amounts of ultraviolet radiation (λ290 - 400 mμ).

Number of Volunteers: 9, sex: female, fair skinned Caucasian.

Number of products screened: 22

Results:

The results are tabulated in the accompanying tables. It is obvious that most of the commercial products do not have any affinity to remain on the surface of the horny layer after a simple procedure of washing with water. These products are easily eluted from the horny layer and do not provide any protection. Some of the preparations that were tested without removing the applied material by washing with water, were found to be ineffective. Contrary to this, the preparation MAP afforded very significant and effective protection. This protection was established after the applied preparation was removed by water washing. It is apparent that PABA, the active ingredient of MAP has a definite affinity to remain on the horny layer.

Conclusions:

A single application of MAP provides excellent protection against ultraviolet radiation ($\lambda <$ 360 mμ) even after the applied material is removed by washing with water. One can statistically conclude that MAP gives considerably more and in fact significant protection from sunburn than any other commercial product that was tested.

Experiment 2: Comparison of MAP and other Commercial Preparations. Light Source: Hot Quartz Hanovia UV Lamp

Subject	Preparation	Brief Protocol	UV Exposure	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
116	Control	Short shower after application	120"	+++	0	5	0	MED of the subject was 30 seconds.
	15% PABA in Ruggles cream		120"	++	25	4	20	
	MAP		120"	0	100	0	100	
	Control	Short shower after application	240"	++++	0	7	0	
	15% PABA in Ruggles cream		240"	++++	0	7	0	
	MAP		240"	±	80	3	60	
	Control	Short shower after application	360"	++++	0	11	0	
	15% PABA in Ruggles cream		360"	++++	0	7	37	
	MAP		360"	±	80	3	73	

Experiment 2: (continued)

Subject	Preparation	Brief Protocol	UV Exposure	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
118	Control	Preparations applied and washed off with water 30 minutes after application	300"	++++	0	13	0	MED of the subject was 30 seconds
	UVAL		300"	++++	0	10	23	
	MAP		300"	+	60	4	69	
	Control	Preparations applied day before exposure, subject had a 10' shower in the night	300"	++++	0	13	0	
	UVAL		300"	++++	0	14	0	
	MAP		300"	++++	0	10	23	

Experiment 2: (continued)

Subject	Preparation	Brief Protocol	UV Exposure	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
120	Control	Preparations applied 1 day prior to UV exposure, subject took a shower in the night.	150"	+++	0	8	0	MED of the subject was 30 seconds.
	A-Fil (Neutral)		150"	+++	0	9	0	
	RVP spray		150"	+++	0	8	0	
	Sundare lotion		150"	+++	0	8	0	
	Coppertone oil		150"	+++	0	9	0	
	Coppertone cream		150"	+++	0	10	0	
	Coppertone shade lotion		150"	+++	0	9	0	
	Coppertone lotion		150"	+++	0	8	0	
	Coppertone Noskote		150"	+++	0	9	0	
	Bain de Soleil		150"	+++	0	10	0	
	MAP		150"	++	25	5	37	

Experiment 2: (continued)

Subject	Preparation	Brief Protocol	UV Exposure	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
120	Control	Preparations applied 1 hour prior to UV exposure, MAP was washed off with water, but not the other products.	300"	++++	0	11	0	MED of the subject was 30 seconds.
	A-Fil (Neutral)		300"	++++	0	10	9	
	RVP		300"	++++	0	11	0	
	Sundare lotion		300"	+++	20	7	37	
	Coppertone oil		300"	++++	0	11	0	
	Coppertone cream		300"	+++	20	6	46	
	Coppertone shadelotion		300"	+++	20	9	19	
	Coppertone lotion		300"	+++	20	7	37	
	Coppertone Noskote		300"	++	40	5	55	
	Bain de Soleil		300"	++++	0	12	0	
	MAP		300"	±	80	2	82	

Experiment 2: (continued)

Subject	Preparation	Brief Protocol	UV Exposure	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
122	Control	Preparations applied and washed off with water 1 hour after application, UV exposure same day.	150"	+++	0	9	0	MED of the subject approximately 30 seconds.
	A-Fil (Neutral)		150"	++	25	5	45	
	RVP spray		150"	+++	0	6	34	
	Sundare lotion		150"	+++	0	9	0	
	Coppertone shade lotion		150"	+	50	4	55	
	Coppertone Noskote		150"	+++	0	9	0	
	Coppertone oil		150"	+	50	4	55	
	Bain de Soleil fluid		150"	++	25	8	11	
	Bain de Soleil cream		150"	+++	0	9	0	
	Bain de Soleil foam		150"	++	25	5	45	
	MAP		150"	0	100	0	100	

Experiment 2: (continued)

Subject	Preparation	Brief Protocol	UV Exposure	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
122	Control	Preparations applied and washed off with water 1 hour after application, UV exposure same day.	300"	++++	0	11	0	MED of the subject approximately 30 seconds.
	A-Fil (Neutral)		300"	++++	0	12	0	
	RVP spray		300"	++++	0	11	0	
	Sundare lotion		300"	+++	20	7	37	
	Coppertone shade lotion		300"	++++	0	10	9	
	Coppertone Noskote		300"	+++	20	7	37	
	Coppertone oil		300"	++++	0	10	9	
	Bain de Soleil fluid		300"	++++	0	11	0	
	Bain de Soleil cream		300"	+++	20	7	37	
	Bain de Soleil foam		300"	4	0	12	0	
	MAP		300"	0	100	0	100	

Experiment 2: (continued)

Subject	Preparation	Brief Protocol	UV Exposure	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
134	Control	Preparations applied and washed off with water 1 hour after application, UV exposure same day.	300"	++++	0	8	0	MED of the subject was 30 seconds.
	S & S oil		300"	++++	0	9	0	
	S & S snootie		300"	++++	0	10	0	
	S & S lotion		300"	++++	0	9	0	
	S & S oil less oil		300"	++++	0	10	0	
	S & S tan fastic oil		300"	++++	0	8	0	
	S & S tan fastic lotion		300"	++++	0	8	0	
	Beauty on the beach		300"	++++	0	7	12	
	Skolex		300"	++++	0	7	12	
	MAP		300"	±	80	2	75	

Experiment 2: (continued)

Subject	Preparation	Brief Protocol	UV Exposure	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
134	Control	Preparations after application were not washed off.	300"	++++	0	8	0	MED of the subject was 30 seconds.
	S & S oil		300"	++++	0	9	0	
	S & S snootie		300"	++++	0	10	0	
	S & S lotion		300"	++++	0	9	0	
	S & S oil less oil		300"	++++	0	10	0	
	S & S tan fastic oil		300"	++++	0	8	0	
	S & S tan fastic lotion		300"	++++	0	8	0	
	Beauty on the beach		300"	++++	0	7	12	
	Skolex		300"	++++	0	7	12	
	MAP		300"	±	80	2	75	

Experiment 2: (continued)

Subject	Preparation	Brief Protocol	UV Exposure	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
135	Control	Preparations applied and washed off with water 1 hour after application, UV exposure same day.	300"	++++	0	12	0	
	UVAL		300"	++	40	8	33	
	Solbar		300"	++	40	10	20	
	Snootie		300"	+++	20	8	33	
	Coppertone lotion		300"	++++	0	11	0	
	RVP		300"	+++	20	9	15	
	MAP		300"	±	80	2	80	

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Experiment 3: Comparison of MAP and commercially available sunscreen preparations under conditions of exercise accompanied by sweating.

Objective: Compare the efficacy of MAP with several commercially available sunscreen preparations under the following conditions:
a) one application of MAP on one side of the back and a commercial preparation on the other side.
b) After application of the test preparations on the back, subject the volunteers to a schedule of heavy exercise leading to profuse perspiration under dry and hot weather conditions.

Protocol:
a) Application of MAP on one side of the back
b) Application of commercial preparation on the other side
c) Control (untreated) areas
d) Exercise period: 30 minutes which included: 10 minutes running, 10 minutes calisthenics, and 10 minutes volley-ball throwing)
e) 24 hours after sun exposure, the exposed areas were evaluated for the degree of erythema

Sun Exposure: The untreated control areas and the areas treated with the MAP and the commercial product, were exposed for 1 and 2 hours to solar radiation (11 a.m. to 1 p.m.) on a clear day, temperature 90-94°F).

Number of Subjects: 14

Results: Results are presented in Tables 3A and 3B. MAP is significantly better than the other commercially available products for both one and two hour sun exposure periods. The probability of there being no difference between various treatments is less than 0.01 in all cases.
All commercial products that were screened, are easily washed away or eluted from the surface of the horny layer as a result of profuse perspiration after exercise. In many instances, they did not afford protection even for a short period of one hour sun exposure. MAP however, was found to protect 100% up to 2 hours sun exposure and over 75% protection (barely perceptible erythema) after 2 hours sun exposure. MAP thus appears to remain adsorbed and partially bonded to the horny layer and therefore has afforded a complete protection against sunburn radiation.

Table 3A: Statistical Interpretation

MAP is significantly better than Sea and Ski with probability ≥ 0.99 for every experimental section: one and two hour exposures and measurement through reflectance readings and degrees of visible redness.

For the specific parts of this experiments:

1 hour exposure, degree of visible redness:

MAP is significantly better than Sea and Ski by at least 1 unit with probability ≥ 0.99.

1 hour exposure, reflectance difference:

MAP is significantly better than Sea and Ski by at least 8 units with probability ≥ 0.99; by at least 9 units with probability ≥ 0.95.

2 hour exposure, degree of visible redness:

MAP is significantly better than Sea and Ski by at least 1 unit with probability ≥ 0.99.

2 hour exposure, reflectance difference:

MAP is significantly better than Sea and Ski by at least 2 units with probability ≥ 0.99; by at least 4 units with probability ≥ 0.95.

Table 3B: Statistical Interpretation

MAP is significantly better than the combination of other treatments presented in experiment 3 for both one and two hour exposure periods. The probability of there being no difference between treatments is less than 0.05 in all cases and less than 0.01 in all cases but the one hour exposure recorded through the use of reflectance differences.

Specifically for the separate parts of experiment 3

1 hour exposure, degree of visible redness:

MAP is significantly better than other treatments with probability >0.99.

1 hour exposure, reflectance difference:

MAP is significantly better than the other treatments with probability >0.95.

2 hour exposure, degree of visible redness:

MAP is significantly better by at least one unit of visible redness measure with probability >0.99.

2 hour exposure, reflectance difference:

MAP is significantly better by at least 2 reflectance difference units with probability >0.99 and significantly better by at least 4 units with probability >0.95.

Conclusions: Exposure of the back particularly in the prone position for a 2 hour period on a dry, hot and clear day in the desert, constitutes a very heavy dose of ultraviolet light. (violacious red color) erythema. Many subjects developed even swelling, edema and blistering reactions. Against this intense exposure, a single application of MAP afforded complete protection when the subjects were exercised for 30 minutes. Other commercial products were found to be less effective or almost ineffective in affording protection.

Experiment 3: Table 3A: Comparison of MAP and "Sea and Ski" under conditions of exercise with sweating.

Subject	Preparation	Period of Sun Exposure	Degree of Visible Redness	% Protection	% Reflectance Difference	% Protection
26550	Control	1 hour	+++	0	13	0
	MAP	"	0	100	1	92
	S & S	"	+++	0	16	0
	Control	2 hours	+++	0	16	0
	MAP	"	+	50	11	32
	S & S	"	+++	0	16	0
25513	Control	1 hour	+	0	8	0
	MAP	"	0	100	0	100
	S & S	"	++	0	15	0
	Control	2 hours	+++	0	14	0
	MAP	"	±	75	0	100
	S & S	"	+++	0	17	0

Experiment 3: Table 3A: Comparison of MAP and "Sea and Ski" under conditions of exercise with sweating.

Subject	Preparation	Sun Exposure	Degree of Visible Redness	% Protection	% Reflectance Difference	% Protection
26215	Control	1 hour	+	0	7	0
	MAP	"	0	100	0	100
	S & S	"	+	0	12	0
	Control	2 hours	++	0	13	0
	MAP	"	±	66	5	72
	S & S	"	+++	0	16	0
23767	Control	1 hour	+	0	12	0
	MAP	"	0	100	0	100
	S & S	"	+	0	11	9
	Control	2 hours	+++	0	16	0
	MAP	"	±	75	5	69
	S & S	"	+++	0	12	26

Experiment 3: Table 3A: (continued)

Subject	Preparation	Period of Sun Exposure	Degree of Visible Redness	% Protection	% Reflectance Difference	% Protection
26033	Control	1 hour	++	0	14	0
	MAP	"	0	100	0	100
	S & S	"	+++	0	15	0
	Control	2 hours	+++	0	18	0
	MAP	"	++	25	15	16
	S & S	"	+++	0	15	16
25645	Control	1 hour	+++	0	18	0
	MAP	"	0	100	0	100
	S & S	"	++++	0	18	0
	Control	2 hours	++++	0	21	0
	MAP	"	++	40	10	52
	S & S	"	++++	0	20	4

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Experiment 3: Table 3A: (continued)

Subject	Preparation	Period of Sun Exposure	Degree of Visible Redness	% Protection	% Reflectance Difference	% Protection
23743	Control	1 hour	±	0	8	0
	MAP	"	±	0	7	0
	S & S	"	±	0	11	0
	Control	2 hours	+++	0	17	0
	MAP	"	+++	0	20	0
	S & S	"	+++	0	22	0
21892	Control	1 hour	+	0	8	0
	MAP	"	0	100	0	100
	S & S	"	++	0	10	0
	Control	2 hours	++	0	10	0
	MAP	"	±	66	2	8
	S & S	"	+++	0	17	0

Experiment 3: Table 3B: Comparison of MAP and Other Commercial Compounds under conditions of exercise with sweating.

Subject	Preparation	Period of Sun Exposure	Degree of Visible Redness	% Protection	% Reflectance Difference	% Protection
16988	Control	1 hour	++	0	10.5	0
	MAP	"	0	100	0	100
	UVAL	"	±	33	4.5	43
	Control	2 hours	+++	0	14	0
	MAP	"	0	100	0	100
	UVAL	"	+	66	5.5	40
25897	Control	1 hour	++	0	13	0
	MAP	"	0	100	0	100
	UVAL	"	++	0	8	38
	Control	2 hours	+++	0	17	0
	MAP	"	±	75	4	76
	UVAL	"	+++	0	14	17

Experiment 3: Table 3B: (continued)

Subject	Preparation	Period of Sun Exposure	Degree of Visible Redness	% Protection	% Reflectance Difference	% Protection
24673	Control	1 hour	+++	0	19	0
	MAP	"	0	100	0	100
	Coppertone shade extra protection	"	±	75	2	89
	Control	2 hours	+++	0	19	0
	MAP	"	0	100	0	100
	Coppertone shade extra protection	"	+++	0	14	26
26107	Control	1 hour	++	0	8	0
	MAP	"	0	100	0	100
	Sundare	"	0	100	0	100
	Control	2 hours	+++	0	13	0
	MAP	"	0*	100	0*	100
	Sundare	"	++	25	13	0

* Immediate pigment darkening

Experiment 3: Table 3B: (continued)

Subject	Preparation	Period of Sun Exposure	Degree of Visible Redness	% Protection	% Reflectance Difference	% Protection
24814	Control	1 hour	+++	0	14	0
	MAP	"	0	100	0*	100
	RVP	"	+	50	12	15
	Control	2 hours	+++	0	17	0
	MAP	"	±	75	0*	100
	RVP	"	+++	0	15	12
21371	Control	1 hour	+++	0	15	0
	MAP	"	±	75	5	66
	RVP	"	+	50	7	53
	Control	2 hours	+++	0	14	0
	MAP	"	++	25	15	0
	RVP	"	+++	0	14	0

* Immediate pigment darkening

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Experiment 4: Comparison of MAP and commercial sunscreen preparations under normal conditions of use.

Objective: Under normal conditions of use to evaluate the potency of MAP against the best known and widely used sunscreen preparations.

The normal conditions of use include:
a) 1 application of MAP on one-half the body (front and back) and on the other half the commercial brand leaving a control (untreated) 5" x 5" area on the back and the front.
b) Walking, lying, running and sitting in the sun without swimming.
c) Long sun exposure: (4 hours)

Number of Subjects: 6

Commercial Products used: Skolex, UVAL and RVP

Results: After a single application and sun exposure ranging from 3 1/2 - 4 hours, MAP was found to be significantly better than the commercial products used in this experiment.

For statistical analysis, the sunburn response observed with each commercial product was grouped together as "combination of other treatments" and compared against the response observed in the MAP treated area. It should not be concluded that MAP is significantly better than each of the other treatments, but the data are analyzed in a manner to indicate that MAP is better than the "combination of other treatments".

Using the degree of visible redness as a measure, MAP was found to be better than the "other treatments" by at least one unit with probability greater than 0.99

Using the skin reflectance difference as a measure for the degree of protection, MAP was found to be significantly better than the "combination of other treatments" by at least three units with probability greater than 0.99.

Conclusions: After a single application and under normal conditions of use, MAP was found to be significantly better than several commercial products used in this experiment with a probability greater than 0.99

EXPERIMENT 4: Comparison of MAP and Commercial Preparations under Normal Conditions of use.

(Long exposure, no exercise, no swimming, just walking around the beach in the sun).

Subject	Preparations	Area Applied	Hours of Sun Exposure	Degree of Visible Redness	% Protection	% Reflectance Difference	% Protection
H. S.	Control	Back and shoulders	4	++	0	10	0
	Skolex	Back and shoulders	4	±	66	3	70
	MAP	Back and shoulders	4	0	100	0	100
E. F.	Control	Shoulders	3-1/2	+++	0	10	0
	Skolex	"	3-1/2	+++	0	10	0
	MAP	"	3-1/2	0	100	0	100
	Control	Lower back	3-1/2	+++	0	10	0
	Skolex	" "	3-1/2	++	25	10	0
	MAP	" "	3-1/2	0	100	0	100
	Control	Front chest	3-1/2	+++	0	6	0
	Skolex	" "	3-1/2	++	25	6	0
	MAP	" "	3-1/2	0	100	0	100

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EXPERIMENT 4: (Continued)

Subject	Preparations	Area Applied	Hours of Sun Exposure	Degree of Visible Redness	% Protection	% Reflectance Difference	% Protection
W. G.	Control	Shoulders	3-3/4	+++	0	10	0
	Skolex	"	3-3/4	++	25	6	40
	MAP	"	3-3/4	0	100	0	100
	Control	Lower back	3-3/4	+++	0	10	0
	Skolex	" "	3-3/4	+	50	5	50
	MAP	" "	3-3/4	0	100	0	100
	Control	Front chest	3-3/4	++	0	6	0
	Skolex	" "	3-3/4	+	33	4	33
	MAP	" "	3-3/4	0	100	0	100
J. C.	Control	Shoulders	3-3/4	+++	0	12	0
	UVAL	"	3-3/4	++	25	9	25
	MAP	"	3-3/4	0	100	0	100
	Control	Lower back	3-3/4	+++	0	12	0
	UVAL	"	3-3/4	±	75	3	75
	MAP	"	3-3/4	0	100	0	100

EXPERIMENT 4: (Continued)

Subject	Preparations	Area Applied	Hours of Sun Exposure	Degree of Visible Redness	% Protection	% Reflectance Difference	% Protection
P. C.	Control	Shoulders	3-3/4	+++	0	12	0
	UVAL	"	3-3/4	++	25	8	34
	MAP	"	3-3/4	0	100	0	100
	Control	Lower back	3-3/4	+++	0	12	0
	UVAL	" "	3-3/4	±	75	4	67
	MAP	" "	3-3/4	0	100	0	100
	Control	Front chest	3-3/4	++	0	5	0
	UVAL	" "	3-3/4	+	33	4	20
	MAP	" "	3-3/4	0	100	0	100
25769	Control	Back and shoulders	3-1/2	++	0	10	0
	RVP	Back and shoulders	3-1/2	+	33	8	20
	MAP	Back and shoulders	3-1/2	0	100	0	100

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EXPERIMENT 5: Effect of swimming in fresh water on sunburn-protecting properties of MAP.

Objective: Under normal conditions of use including swimming, determine whether a single application of MAP affords satisfactory protection against sunburn radiation. The primary object was to ascertain whether the applied MAP preparation was completely removed from the surface of the horny layer or was partially binding with the horny layer.

Protocol: This experiment was carried out on 9 female volunteers (fair skinned Caucasians). MAP was applied only on the backs of the volunteers (either on left or right side). Adjacent untreated skin areas served as controls. Subjects were exposed to ultraviolet radiation from a hot quartz mercury arc lamp emitting radiation primarily of 257, 297, and 365 mμ wavelengths. The exposure period ranged from 60 seconds to 240 seconds. (The minimal erythema dose for these 9 volunteers ranged between 15 and 20 seconds under our experimental conditions).
The sunscreen preparation was applied between 9-11 a.m. The volunteers were then allowed to wear usual working clothes and asked to return to their jobs until 5 p.m. In the evening, they were taken to an indoor swimming pool and instructed to remain in the water and swim for varying periods ranging from 10 minutes to 20 minutes. After swimming, the volunteers wore everyday clothes and returned to laboratory for irradiation of the back. Thus, the applied test preparation was subjected to at least two processes of removal: 1) the frictional contact with the clothes and, 2) the effect of swimming.

Results: The data concerning this experiment are tabulated in the accompanying table. We find that a single application of MAP affords a significant protection to the skin against sunburn radiation under experimental conditions that can easily wash off the sunscreen preparation if the preparation MAP were not partially binding with the horny layer. The preparation MAP thus has a unique property to remain either adsorbed or conjugated chemically to the skin and afford protection against sunburn radiation. This protection against artificial ultraviolet light source is roughly equivalent to 60-90 minutes of sun exposure.

Experiment 5: Effect of Swimming on Sun-Protecting Properties of MAP. (Indoor Swimming Pool)

Light Source: Hot Quartz Mercury Lamp MED of the Subjects 15" - 30"

Subject	Swimming Time	Preparation	UV Exposure	Degree of Visible Redness	%Protection	% Reflectance Difference	% Protection
130	10 minutes	Control	90"	++	0	5	0
		MAP	90"	0	100	1	80
		Control	120"	+++	0	5	0
		MAP	120"	+	50	3	40
		Control	240"	++++	0	10	0
		MAP	240"	++	40	5	50
132	10 minutes	Control	90"	++	0	6	0
		MAP	90"	±	66	2	66
		Control	120"	+++	0	5	0
		MAP	120"	+	50	2	60
		Control	240"	++++	0	10	0
		MAP	240"	+++	20	6	40

Experiment 5: (continued)

Subject	Swimming Time	Preparation	UV Exposure	Degree of Visible Redness	% Protection	% Reflectance Difference	% Protection
136	10 minutes	Control	60"	+	0	5	0
		MAP	60"	0	100	1	80
		Control	120"	+++	0	10	0
		MAP	120"	±	75	3	70
		Control	240"	++++	0	11	0
		MAP	240"	++	40	7	37
138	20 minutes	Control	90"	++	0	3	0
		MAP	90"	±	66	2	33
		Control	120"	+++	0	5	0
		MAP	120"	+	50	2	60
		Control	240"	++++	0	6	0
		MAP	240"	++	40	3	50

Experiment 5: (continued)

Subject	Swimming Time	Preparation	UV Exposure	Degree of Visible Redness	% Protection	% Reflectance Difference	% Protection
140	20 minutes	Control	90"	+++	0	4	0
		MAP	90"	±	75	1	75
		Control	120"	++++	0	6	0
		MAP	120"	±	80	2	66
		Control	240"	++++	0	7	0
		MAP	240"	++	40	3	50
144	20 minutes	Control	90"	+++	0	5	0
		MAP	90"	++	25	4	20
		Control	120"	+++	0	9	0
		MAP	120"	++	25	5	45
		Control	240"	++++	0	6	0
		MAP	240"	++++	0	7	0

Experiment 5: (continued)

Subject	Swimming Time	Preparation	UV Exposure	Degree of Visible Redness	% Protection	% Reflectance Difference	% Protection
145	20 minutes	Control	90"	+	0	4	0
		MAP	90"	±	50	2	50
		Control	120"	+++	0	5	0
		MAP	120"	±	75	3	40
		Control	240"	++++	0	8	0
		MAP	240"	+++	20	9	0
146	20 minutes	Control	90"	+++	0	9	0
		MAP	90"	+++	0	6	33
		Control	120"	++++	0	10	0
		MAP	120"	++++	0	8	20
		Control	240"	++++	0	13	0
		MAP	240"	++++	0	13	0
150	20 minutes	Control	60"	+++	0	9	0
		MAP	60"	±	75	1	89
		Control	120"	++++	0	15	0
		MAP	120"	+	60	3	80
		Control	240"	++++	0	17	0
		MAP	240"	++	40	6	65

ARIZONA STATE PRISON CLINICAL TRIAL

APRIL, 1966

Experiment 6: Comparison of MAP and a sunscreen preparation described by Fusaro, Runge and Watson.

Note: In this experiment, the preparation described by Fusaro et al (Arch. Dermatology 93:106, 1966) will be referred to as Runge Preparation. Runge preparation consists of a combination of dihydroxy acetone (DHA) and Lawsone (2-hydroxy-1, 4-naphthoquinone). The final concentration of dihydroxy acetone was 3% and of Lawsone was 0.13%.

Objective: To compare the sun protective efficacy of MAP and Runge preparation.

Number of Subjects: 4. This group of subjects was divided into two groups; Group 6A: 2 subjects and Group 6B: 2 subjects. With a masking tape, the back of each volunteer was partitioned into four equal size parts. The test preparations were applied as follows:

Group 6A:	Runge prep	Control
	MAP prep	3% DHA
Group 6B:	DHA	Control
	Runge prep	MAP prep

Protocol: Group 6A: 2 subjects, received 6 applications, 14-20 hours before the test. The mixture of Lawsone and DHA as described by Fusaro et al, was applied each hour for six hours. Preparation MAP and 3% DHA solution were also applied in separate quadrants at each hour for six hours. Next morning, the subjects were asked to swim for 30 minutes. After swimming, they were exposed to solar radiation between 12 noon and 2 p.m. (1 and 2 hours of sun exposure).

Group 6B: 1 application of Runge preparation, 3% DHA and MAP preparation 1 hour prior to sun exposure. The subjects were exposed for 1 1/2 and 3 hours to solar radiation between 12 noon and 3 p.m. The applied preparations were not washed or rubbed off the horny layer. The subjects did not carry out any swimming schedule.

Comments: The Runge preparation described by Fusaro et al was found to be less effective than MAP. The preparation and the method

of application proposed by these investigations leave a color on the skin that is not very appealing and desirable. This preparation undoubtedly imparts partial protection to the subjects against solar radiation. However, our data reveal that the protection afforded is primarily due to dihydroxy acetone (DHA) that combines with the amino acids of keratin (most probably the histidine) in the horny layer. MAP, on the contrary, a colorless preparation, was indeed found to be better than the Runge preparation.

EXPERIMENT 6A:

Subject & Prison #	Site	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
25452	Back	Control	60	++	0	13	0	
(1)	"	MAP	"	±	66	6	54	
	"	Runge	"	0	100	6	54	Orange-yellow
	"	DHA	"	0	100	6	54	
	"	Control	120	+++	0	15	0	
	"	MAP	"	+++	0	12	20	
	"	Runge	"	+++	0	11	26	Orange-yellow
	"	DHA	"	+++	0	15	0	
25265	"	Control	60	+++	0	21	0	
(2)	"	MAP	"	±	75	9	57	
	"	Runge	"	++	25	20	4	Orange-yellow
	"	DHA	"	+	50	17	20	
	"	Control	120	+++	0	21	0	
	"	MAP	"	+	50	12	40	
	"	Runge	"	+++	0	21	0	Orange-yellow
	"	DHA	"	++	25	15	29	

EXPERIMENT 6B:

Degree of Erythema after 24 Hours

Subject & Prison #	Site	Preparation	Period of Sun Exposure (Minutes)	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
26099	Back	Control	90	+++	0	15	0	To avoid
(1)	"	MAP	"	0	100	1	100	intense
	"	Runge	"	0	100	2	100	sunburn, sun
	"	DHA	"	0	100	2	100	exposure
	"	Control	120	++++	0	20	0	to control
	"	MAP	240	+	60	10	50	areas was
	"	Runge	"	+++	20	14	30	limited
	"	DHA	"	++	40	14	30	to 90 and
26251 (2)	"	Control	90	+++	0	19	0	120 minutes
	"	MAP	"	±	80	8	60	only
	"	Runge	"	+	50	10	47	
	"	DHA	"	+	50	9	50	
	"	Control	120	++++	0	21	0	
	"	MAP	240	+	60	13	40	
	"	Runge	"	+++	20	18	15	
	"	DHA	"	+++	20	18	15	

ARIZONA STATE PRISON CLINICAL TRIAL

SEPTEMBER, 1967

Introduction: The second clinical trial at the Arizona State Prison was carried out between September 15 and 23, 1967.

The Following preparations were evaluated:

1. 1% **MAP** (contains 1% of para-aminobenzoic acid (**PABA**) in 95% ethyl alcohol.

2. 3% **MAP** (3% **PABA** in 95% C_2H_5OH)

3. 5% **MAP** (5% **PABA** in 95% C_2H_5OH)

4. 5% **MAPOL** (5% **PABA** in 95% C_2H_5OH & 0.5% cetyl alcohol as emollient)

5. Formula **MAP** (D5) contains 5% w/v of **PABA**, 5% oleyl alcohol as emollient and 2% polyoxyethylene oleyl ether dissolved in 100 ml. of 70% C_2H_5OH. The polyoxyethylene oleyl ether was primarily used as a solubilizer and contains a series of polyoxyethylene ethers of fatty alcohol (e.g. cetyl; stearyl and oleyl alcohol). The final concentration of ethyl alcohol ranged between 65-75%.

6. Formula F3 contains 3% w/v of **PABA** in 70% C_2H_5OH. The other ingredients were the same as in Formula **MAP** (D5).

7. Formula G contains 2.5% Eskalol 506 (iso-amyl p-N, N-dimethyl aminobenzoate) in 65-70% C_2H_5OH.

8. **UVAL**, a commercial product contains 10% 2-hydroxy, 4-methoxybenzophenone- 5-sulfonic acid (Pharmafac, Austin, Texas) was selected to compare against 5% **MAP**, and Formula **MAP** (D5).

In this clinical trial, 50 volunteers were used to evaluate the above listed test preparations under 4 different conditions:

1. Evaluate the degree of protection after a single application of the test preparations on backs and legs without subjecting the volunteers to any schedule of exercise or swimming.

2. Evaluate the degree of protection after subjecting the volunteers to a schedule of heavy exercise accompanied by sweating under dry and hot weather conditions.

3. Evaluate the degree of protection after 15 minutes of swimming.

4. Evaluate the protective effect under normal conditions of use (e.g. swimming, lying on the back and prone position, walking and running around in the sun).

Experiments #1-3 were designed to test 1, 3, and 5% MAP and MAPOL preparations under 3 conditions listed above, and the principal objects were as follows:

1. To establish the minimum concentration of PABA that is essential to obtain the most satisfactory protection. Three preparations containing 1, 3, and 5% PABA were evaluated.

2. The second objective was to evaluate the tanning response in each subject after application of these various preparations and sun exposure. We were primarily interested in finding out whether normal tanning (that includes both the immediate pigment darkening reaction and the formation of new malanin, i.e. melanogenesis) can be elicited in the absence of visible sun burn reaction. It has long been assumed that normal suntanning can occur only when there is an inflamation and sunburn reaction in the skin.

3. Whether the incorporation of emollients in these preparations affects the affinity of these various preparations to either conjugate chemically or remain adsorbed on the stratum corneum after vigorous sweating and swimming.

Experiment #4 was designed to evaluate the maximum period of sun exposure that can be tolerated by individuals without evoking undue sunburn response after a single application of 5% MAP and preparation G and a schedule of 15 minutes of swimming.

Experiments #5-7 were designed to test Formula 5% MAP, MAP (D5), and Formula F under 3 conditions:

a. sun exposure without exercise and swimming.

b. sun exposure with 30 minutes exercise accompanied

by sweating.

c. sun exposure with 15 minutes swimming. The aim was to ascertain the relative degree of protection afforded by each of these preparations. The normal tanning response was also evaluated.

Experiments #8-10 were designed to test the effect of a single application of preparations **MAP**, Escalol 506 (G), and **UVAL** under three conditions:

1. without any activity
2. With 30 minutes exercise
3. after 15 minutes swimming.

The principal objects were:

a. to compare the relative protection afforded by 3 preparations,

b. the affinity of **MAP**, G, or **UVAL** to remain on the stratum corneum, after various activities,

c. the degree of tanning observed after sun exposure.

Experiment #11 was designed to test the preparations **MAP**, **MAP** (D5) and G under conditions simulating normal usage. Normal conditions of use included those activities that one would do on a beach on a bright and hot day.

Comments and Conclusions:

In a place like Florence, **Arizona**, even in the month of September, fair skinned individuals show minimal perceptible redness at 24 hours when exposed for 10 minutes to solar radiation in the horizontal position. **An** exposure for 60 minutes in the horizontal position (under dry, hot and intense sun without any sunscreen) will produce marked erythema accompanied by swelling, edema, and marked tenderness of the skin. The exposure periods selected in our experiments ranging from 40 minutes to 2 1/2 hours constituted an intense challenge for the skin. Leading commercial preparations that were evaluated in our earlier trials were found to be ineffective and could not provide any satisfactory protection after 60 minutes sun exposure, even under conditions which involved no sweating and no swimming.

Observations gathered on fifty volunteers in this September 1967 clinical trial lead us to the following conclusions:

1. After a single application and without any exercise or swimming, preparations 1, 3 and 5% **MAP** provide excellent protection against 2 1/2 hours of sun exposure. A single application of 5% **MAP** or **MAP** (D5) under similar conditions (no exercise and no swimming) will prevent the sunburn response after exposing the backs of the individuals in the horizontal position for a period of 4-5 hours of solar radiation.

2. 5% **MAP** was found to afford very effective protection after subjecting the volunteers to a schedule of vigorous exercise accompanied by sweating. The sweating and the frictional contact of the clothes with the skin did not remove the protective film during exercise. This preparation showed excellent protection up to 2 1/2 hours of sun exposure.

3. After one application of 1, 3 and 5% **MAP** preparations and 15 minutes of swimming, no protection was observed when volunteers were exposed to 2 1/2 hours of solar radiation. However, 5% **MAP** was found to provide quite satisfactory protection when exposure to sun was limited to 45-90 minutes.

4. The preparation **MAP** (D5) that contains 5% **PABA** and emollient in 70% alcohol was found to be as effective as 5% **MAP** preparation. It provides excellent protection even after a heavy schedule of exercise and swimming.

5. 5% **MAPOL**, the preparation containing an emollient was found to be least effective even under contions of no exercise and no swimming. Incorporation of an emollient like cetyl alcohol markedly alters the property of the **MAP** preparation. It appears to be easily eluted from the surface of the skin.

6. Both 5% **MAP** and **MAP** (D5) allow the desired tanning without the discomforts of sunburn.

7. F3 Preparation was found to be less effective than 5% **MAP** and **MAP** (D5) preparations under three experimental conditions (e.g. without exercise and without swimming; with exercise and with swimming). It appears that a minimum of 5% concentration **PABA**

is essential to obtain satisfactory protection under conditions of sweating and swimming.

8. Preparations 5% MAP and MAP (D5) appear to have a definite affinity to remain on the surface of the layer after subjecting the volunteers to a schedule of exercise accompanied by sweating and 15 minutes of swimming. Preparation UVAL was significantly less protective under similar conditions and appeared to have been washed away from the skin surface under experimental conditions of exercise and swimming.

9. Preparation G appeared to have a definite affinity to bind or remain adsorbed on the surface of the stratum corneum. It affords better protection than 5% MAP and MAP (D5), i.e., the subjects can tolerate up to 90 minutes of sun exposure after one application and 15 minutes of swimming.

10. Under normal conditions of usage, preparations 5% MAP and MAP (D5) were found to provide excellent protection. UVAL was found to be lease effective and appeared to have been washed away after swimming and sweating.

ARIZONA STATE PRISON CLINICAL TRIAL, SEPTEMBER 1967

EXPERIMENT 1: Evaluation of 1, 3 and 5% MAP and 5% MAPOL* Preparations: MAP and Sunlight without Swimming and Exercise

1% MAP + 2½ hours sunlight without exercise or swimming
3% MAP + 2½ hours sunlight without exercise or swimming
5% MAP + 2½ hours sunlight without exercise or swimming
5% MAPOL* + 2½ hours sunlight without exercise or swimming

Object: After one application of MAP or MAPOL in 1, 3 and 5% concentration and sun exposure for 2½ hours, evaluate:

a) the degree of erythema without subjecting the volunteers to exercise or swimming prior to sun exposure.

b) the degree of tanning.

Number of Subjects: 6

Results:
1) 1% MAP gave 100% protection in 4 out of 6 subjects after 2½ hours sun exposure.
2) 3% MAP gave 100% protection in 4 out of 6 subjects after 2½ hours sun exposure.
3) 5% MAP gave 100% protection in 4 out of 6 subjects after 2½ hours sun exposure. The other 2 subjects showed at least 80% protection.
4) 5% MAPOL gave 100% protection in 2 out of 6 subjects after 2½ hours sun exposure. The protection in 4 other subjects ranged from 40 to 75%.

Conclusion:
1) After a single application and without any exercise or swimming, 1, 3 and 5% MAP preparations provide significant protection up to 2½ hours sun exposure.
2) MAPOL, the preparation with 0.5% cetyl alcohol as an emollient, was found to be not as effective as 1, 3 and 5% MAP preparations. It is clear that incorporation of an emollient into the standard MAP preparation radically changes its sun protective properties.

* MAPOL: a new formulation of MAP (5%) with an emollient.

Experiment 1: Evaluation of 1, 3 & 5% MAP & MAPOL Preparations: MAP and Sunlight without Swimming and Exercise

				Degree of Erythema after 24 Hours				
Subject & Prison #	Site	Preparation	Period of Sun Exposure	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Davis	Back	Control	2½ hrs.	++	0	9	0	
26935	Back	1% MAP	"	0	100	9	100	IPD
(1)	Back	3% MAP	"	0	100	6	100	IPD
	Back	5% MAP	"	0	100	7	100	IPD
	Back	5% MAPOL	"	0	100	8	100	IPD
	Left Leg	Control	"	++	0	9	0	
		5% MAP	"	0	100	0	100	
	Right Leg	Control	"	+++	0	10	0	
		5% MAPOL	"	0	100	0	100	

IPD = Immediate pigment darkening reaction.

Experiment 1: Evaluation of 1, 3 & 5% MAP & MAPOL Preparations: MAP and Sunlight Without Swimming and Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Kennedy	Back	Control	2½ hrs.	+++	0	15	0	
27163	Back	1% MAP	"	0	100	10	100	IPD
(2)	Back	3% MAP	"	0	100	10	100	IPD
	Back	5% MAP	"	0	100	5	100	IPD
	Back	5% MAPOL	"	±	75	10	75	IPD
	Left Leg	Control	"	+++	0	6	0	
		5% MAP	"	0	100	0	100	
	Right Leg	Control	"	+++	0	6	0	
		5% MAPOL	"	0	100	1	85	

IPD = Immediate pigment darkening reaction.

Experiment 1: Evaluation of 1, 3 & 5% MAP & MAPOL Preparations: MAP and Sunlight without Swimming and Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Hoskins	Back	Control	2½ hrs.	++	0	12	0	
27724 (3)	Back	1% MAP	"	0	100	4	100	IPD
	Back	3% MAP	"	0	100	10	100	IPD
	Back	5% MAP	"	0	100	6	100	IPD
	Back	5% MAPOL	"	0	100	10	100	IPD
	Left Leg	Control	"	+++	0	9	0	
		5% MAP	"	++	25	4	56	
	Right Leg	Control	"	+++	0	8	0	
		5% MAPOL	"	+++	0	5	38	

IPD = Immediate pigment darkening reaction

Experiment 1: Evaluation of 1, 3 & 5% MAP & MAPOL Preparations: MAP and Sunlight without Swimming and Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
McNatt	Back	Control	2½ hrs.	+++	0	14	0	
27152	Back	1% MAP	"	0	100	6	100	IPD
(4)	Back	3% MAP	"	0	100	11	100	IPD
	Back	5% MAP	"	0	100	6	100	IPD
	Back	5% MAPOL	"	+	50	15	50	
	Left Leg	Control	"	++	0	6	0	
		5% MAP	"	0	100	0	100	
	Right Leg	Control	"	+++	0	5	0	
		5% MAPOL	"	++	25	2	60	

IPD = Immediate pigment darkening reaction.

Experiment 1: Evaluation of 1, 3 & 5% MAP & MAPOL Preparations: MAP and Sunlight without Swimming and Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Gray	Back	Control	2½ hrs.	++++	0	16	0	
25897	Back	1% MAP	"	+	60	13	20	
(5)	Back	3% MAP	"	±	80	8	50	
	Back	5% MAP	"	±	80	8	50	
	Back	5% MAPOL	"	+	60	10	38	
	Left Leg	Control	"	++++	0	13	0	
		5% MAP	"	0	100	0	100	
	Right Leg	Control	"	++++	0	12	0	Purpura
		5% MAPOL	"	++	40	8	33	

Experiment 1: Evaluation of 1, 3 & 5% MAP & MAPOL Preparations: MAP and Sunlight without Swimming and Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Bouldin	Back	Control	2½ hrs.	++++	0	17	0	
26858	Back	1% MAP	"	++	40	10	40	
(6)	Back	3% MAP	"	++	40	12	30	
	Back	5% MAP	"	±	80	11	80	IPD
	Back	5% MAPOL	"	++	40	13	24	
	Left Leg	Control	"	+++	0	9	0	
		5% MAP	"	±	75	4	56	
	Right Leg	Control	"	+++	0	8	0	
		5% MAPOL	"	+	50	6	25	

IPD = Immediate pigment darkening reaction

ARIZONA STATE PRISON CLINICAL TRIAL, SEPTEMBER 1967

Experiment 1: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations: MAP and Sunlight without Swimming and Exercise

Object: After one application of 1, 3 and 5% MAP and 5% MAPOL and sun exposure for 2½ hours, evaluate the degree of tanning and residual erythema at the end of 96 and 144 hours.

Results: At the end of 96 and 144 hours post exposure, control (untreated) areas still showed residual erythema in 5 out of 6 subjects. Areas treated with 5% MAP, on the other hand, showed no residual erythema. Likewise, areas treated with 1 and 3% MAP revealed no erythema at the end of 96 and 144 hours post exposure. 5% MAPOL treated areas however, showed some residual erythema in 3 out of 6 subjects.

The tanning response was definitely better in areas protected with MAP than in control (untreated) areas. There was no detectable difference in the rate of tanning or in the degree of tanning in areas treated with 1, 3 and 5% MAP preparations. The tanning response in 5% MAPOL treated areas and in the control areas was practically identical.

Conclusions: An exposure of 2½ hours solar radiation constitutes an intense challenge for promoting tanning. An unprotected skin, under these conditions, becomes intensely red. One also observes desquamation, dyskeratosis and edema. Pigment transfer to the keratinocytes is affected and tanning of the skin does not occur so effectively. This intense erythema can persist for over 7 days period. Contrary to this, if sunburn is minimized or blocked by application of such preparations as 1, 3 and 5% MAP, the same exposure of 2½ hours of solar radiation can promote most effective and ideal tanning. No dyskeratosis, desquamation and edema are observed in these protected areas. Thus tanning can be achieved without the discomforts of sunburn.

Experiment 1: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations: MAP and Sunlight without Swimming and Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Tanning (4 & 6 Days) Day	Visible Redness	Visible Tanning
Gray	Back	Control	2½ hours	4th day	+++	0
25897	"	1% MAP	"	"	±	++
	"	3% MAP	"	"	±	++
	"	5% MAP	"	"	0	++
	"	5% MAPOL	"	"	±	++
	Back	Control	2½ hours	6th day	+++	0
	"	1% MAP	"	"	0	+
	"	3% MAP	"	"	0	+
	"	5% MAP	"	"	0	+
	"	5% MAPOL	"	"	0	+
Davis	Back	Control	2½ hours	4th day	+	++
26935	"	1% MAP	"	"	0	++
	"	3% MAP	"	"	0	++
	"	5% MAP	"	"	0	++
	"	5% MAPOL	"	"	0	++
	Back	Control	2½ hours	6th day	±	++
	"	1% MAP	"	"	0	++
	"	3% MAP	"	"	0	++
	"	5% MAP	"	"	0	++
	"	5% MAPOL	"	"	0	++

Experiment 1: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations: MAP and Sunlight without Swimming and Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Tanning (4 & 6 Days) Day	Visible Redness	Visible Tanning
Kennedy	Back	Control	2½ hours	4th day	++	++
27163	"	1% MAP	"	"	0	++
	"	3% MAP	"	"	0	++
	"	5% MAP	"	"	0	+
	"	5% MAPOL	"	"	±	++
	Back	Control	2½ hours	6th day	+	++
	"	1% MAP	"	"	0	++
	"	3% MAP	"	"	0	++
	"	5% MAP	"	"	0	++
	"	5% MAPOL	"	"	0	++
Hoskins	Back	Control	2½ hours	4th day	0	+++
27724	"	1% MAP	"	"	0	+++
	"	3% MAP	"	"	0	+++
	"	5% MAP	"	"	0	+++
	"	5% MAPOL	"	"	0	+++
	Back	Control	2½ hours	6th day	0	++
	"	1% MAP	"	"	0	++
	"	3% MAP	"	"	0	++
	"	5% MAP	"	"	0	++
	"	5% MAPOL	"	"	0	++

Experiment 1: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations: MAP and Sunlight without Swimming and Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Tanning (4 & 6 Days) Day	Visible Redness	Visible Tanning
McNatt	Back	Control	2½ hours	4th day	++	+
27152	"	1% MAP	"	"	0	++
	"	3% MAP	"	"	0	++
	"	5% MAP	"	"	0	++
	"	5% MAPOL	"	"	±	++
	Back	Control	2½ hours	6th day	+	++
	"	1% MAP	"	"	0	++
	"	3% MAP	"	"	0	++
	"	5% MAP	"	"	0	++
	"	5% MAPOL	"	"	0	++
Bouldin	Back	Control	2½ hours	4th day	+++	+
26858	"	1% MAP	"	"	0	++
	"	3% MAP	"	"	+	++
	"	5% MAP	"	"	0	++
	"	5% MAPOL	"	"	+	++
	Back	Control	2½ hours	6th day	+++	++
	"	1% MAP	"	"	0	++
	"	3% MAP	"	"	0	++
	"	5% MAP	"	"	0	++
	"	5% MAPOL	"	"	±	++

ARIZONA STATE PRISON CLINICAL TRIAL, SEPTEMBER 1967

EXPERIMENT 2: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations after Exercise

1% MAP + 30 minutes exercise + $2\frac{1}{2}$ hours sun exposure
3% MAP + 30 minutes exercise + $2\frac{1}{2}$ hours sun exposure
5% MAP + 30 minutes exercise + $2\frac{1}{2}$ hours sun exposure
5% MAPOL + 30 minutes exercise + $2\frac{1}{2}$ hours sun exposure

Object:

a) Whether a single application of 1, 3 and 5% MAP affords satisfactory protection against sunburn radiation when the volunteers are subjected to a schedule of heavy exercise leading to profuse perspiration under dry and hot sunny weather.

b) Determine the degree of tanning.

Schedule of Exercise: Running and walking, calisthenics and basketball. Volunteers had clothes on during exercise. At the end of the exercise period each subject was found to be profusely sweating.

Number of Subjects: 6

Results:

1) 1% MAP preparation provided only partial protection (33-75%) in 3 out of 6 subjects. Similarly, 3% MAP preparation provided partial protection to 3 out of 6 subjects. 5% MAP preparation was quite effective in at least 4 out of 6 subjects. Even the remaining 2 subjects showed significant protection.

2) The back of the legs (calf areas) treated with MAPOL showed definite but not absolute protection. 5% MAPOL on the back was found to be least effective. The erythemal response in areas treated with MAPOL was almost identical to that of control (untreated) areas.

Conclusions:

1) Even after profuse sweating and elution of the applied preparation due to frictional contact of the clothes with the body, 5% MAP was found to be quite effective in providing satisfactory protection. The data indicate that preparation MAP appears to have remained adsorbed on the surface of the horny layer. Considering the fact that each subject received $2\frac{1}{2}$ hours sun exposure, which is roughly equal to about 10 times the minimal erythema dose, the protection that one observes is certainly significant.

2) 1 and 3% MAP preparations and 5% MAPOL preparation did not provide the desired adequate protection. The active ingredients either were eluted from the surface of the horny layer as a result of sweating or were rubbed off due to frictional contact with the clothes. It is also possible that the exposure period of $2\frac{1}{2}$ hours was too much for each area receiving 1 and 3% MAP preparation.

Experiment 2: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations: MAP and Sunlight with 30 Minutes Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Sears	Back	Control	2½ hours	++++	0	15	0	
26550	"	1% MAP	"	++++	0	16	0	
(1)	"	3% MAP	"	++++	0	13	20	
	"	5% MAP	"	+++	20	11	30	
	"	5% MAPOL	"	++++	0	18	0	
	Left Leg	Control	"	+++	0	9	0	
	"	5% MAP	"	0	100	0	100	
	Right Leg	Control	"	+++	0	10	0	
	"	5% MAPOL	"	+	50	7	30	

Experiment 2: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations: MAP and Sunlight with 30 Minutes Exercise

				Degree of Erythema after 24 Hours				
Subject & Prison #	Site	Preparation	Period of Sun Exposure	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Colvin	Back	Control	2½ hrs.	+++	0	13		
27154	"	1% MAP	"	±	75	10		IPD*
(2)	"	3% MAP	"	±	75	11		IPD*
	"	5% MAP	"	±	75	12		IPD*
	"	5% MAPOL	"	++	25	11		IPD*
	Left Leg	Control	"	+++	0	18	0	
	"	5% MAP	"	0	100	3	83	
	Right Leg	Control	"	+++	0	18	0	
	"	5% MAPOL	"	+	50	7	62	

IPD = Immediate pigment darkening reaction.

*Because of pigment darkening and redistribution reaction, the % protection based on reflectance readings could not be calculated.

Experiment 2: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations: MAP and Sunlight with 30 Minutes Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Hanks	Back	Control	2½ hours	+++	0	[illegible]	0	
26968	"	1% MAP	"	+++	0	18	0	
(3)	"	3% MAP	"	+++	0	18	0	
	"	5% MAP	"	+	50	11	40	
	"	5% MAPOL	"	+++	0	18	0	
	Left Leg	Control	"	++	0	10	0	
	"	5% MAP	"	0	100	2	80	
	Right Leg	Control	"	++	0	9	0	
	"	5% MAPOL	"	+	33	5	45	

Experiment 2: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations: MAP and Sunlight with 30 Minutes Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Shipman	Back	Control	2½ hours	++++	0	16	0	
25810	"	1% MAP	"	++	40	12	25	
(4)	"	3% MAP	"	++	40	13	20	
	"	5% MAP	"	++	40	9	45	
	"	5% MAPOL	"	+++	20	16	0	
	Left Leg	Control	"	++++	0	15	0	
	"	5% MAP	"	0	100	4	0	
	Right Leg	Control	"	+++	0	11	0	
	"	5% MAPOL	"	+	50	10	50	

Experiment 2: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations: MAP and Sunlight with 30 Minutes Exercise

				Degree of Erythema after 24 Hours				
Subject & Prison #	Site	Preparation	Period of Sun Exposure	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Lawhorn	Back	Control	2½ hours	++++	0	15	0	
27642	"	1% MAP	"	++++	0	16	0	
(5)	"	3% MAP	"	++++	0	15	0	
	"	5% MAP	"	+++	20	10	33	
	"	5% MAPOL	"	++++	0	15	0	
	Left Leg	Control	"	+++	0	18	0	
	"	5% MAP	"	±	75	4	78	
	Right Leg	Control	"	+++	0	11	0	
	"	5% MAPOL	"	+	50	6	45	

Experiment 2: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations: MAP and Sunlight with 30 Minutes Exercise

				Degree of Erythema after 24 Hours				
Subject & Prison #	Site	Preparation	Period of Sun Exposure	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Traylor	Back	Control	2½ hours	+++	0	21		
26627	"	1% MAP	"	±	75	19		IPD*
(6)	"	3% MAP	"	+	50	15		IPD*
	"	5% MAP	"	±	75	13		IPD*
	"	5% MAPOL	"	++	25	16		IPD*
	Left Leg	Control	"	++++	0	17	0	
	"	5% MAP	"	0	100	3	82	
	Right Leg	Control	"	++++	0	20	0	
	"	5% MAPOL	"	++	40	9	55	

IPD = Pigment darkening reaction.

*Because of pigment darkening and redistribution reaction, the % protection based on reflectance readings could not be calculated.

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EXPERIMENT 2: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations after 30 Minutes Exercise

Object: Determine the tanning response on the 4th day (96 hours) and 6th day (144 hours) after 2½ hours sun exposure under the following conditions: 1 application of 1, 3 and 5% MAP and 5% MAPOL preparations and 30 minutes exercise.

Results: On the 4th and 6th day after sun exposure for a period of 2½ hours, the control (untreated) areas still showed significant residual erythema. Contrary to this, 5% MAP treated areas showed practically no residual erythema on the 4th and 6th day. Because of desquamation, edema and dyskeratosis, a moderately intense sunburn can delay the appearance of visible tan. This was evident in the control untreated areas. Tanning response of the control areas was not as marked as that observed in 5% MAP treated areas. Areas treated with 1 and 3% MAP and 5% MAPOL showed residual erythema up to 6 days of observation. The tanning response in these areas was practically similar to that observed in control (untreated) areas.

Conclusions: Tanning of the skin can be promoted without evoking appreciable sunburn. 1 application of 5% MAP preparation provided excellent protection against sunburn and permitted ideal tanning of the skin without the discomforts of sunburn.

1 and 3% MAP preparations did not provide any better tanning than 5% MAP preparation.

5% MAPOL was not as effective as 5% MAP both in terms of sunburn protection as well as tanning response.

Experiment 2: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations after Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Tanning (4 & 6 Days) Day	Visible Redness	Visible Tanning
Sears 2655C (1)	Back	Control	2½ hours	4th day	+++	0
	"	1% MAP	"	"	+++	0
	"	3% MAP	"	"	++	0
	"	5% MAP	"	"	+	+
	"	5% MAPOL	"	"	++	0
	Back	Control	2½ hours	6th day	++++	0
	"	1% MAP	"	"	+++	±
	"	3% MAP	"	"	++	0
	"	5% MAP	"	"	+	+
	"	5% MAPOL	"	"	+++	±
Colvin 27154 (2)	Back	Control	2½ hours	4th day	++++	0
	"	1% MAP	"	"	0	++
	"	3% MAP	"	"	0	++
	"	5% MAP	"	"	0	++
	"	5% MAPOL	"	"	+	++
	Back	Control	2½ hours	6th day	+	+
	"	1% MAP	"	"	0	++
	"	3% MAP	"	"	0	++
	"	5% MAP	"	"	0	++
	"	5% MAPOL	"	"	0	++

Experiment 2: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations after Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Tanning (4 & 6 Days) Day	Visible Redness	Visible Tanning
Hanks	Back	Control	2½ hours	4th day	+++	±
26968	"	1% MAP	"	"	++	±
(3)	"	3% MAP	"	"	++	±
	"	5% MAP	"	"	+	++
	"	5% MAPOL	"	"	++	+++
	Back	Control	2½ hours	6th day	+++	++
	"	1% MAP	"	"	++	++
	"	3% MAP	"	"	++	++
	"	5% MAP	"	"	±	++
	"	5% MAPOL	"	"	+	++
Shipman	Back	Control	2½ hours	4th day	++	+++
25810	"	1% MAP	"	"	+	+++
(4)	"	3% MAP	"	"	0	+++
	"	5% MAP	"	"	0	+++
	"	5% MAPOL	"	"	+	+++
	Back	Control	2½ hours	6th day	++	++
	"	1% MAP	"	"	±	++
	"	3% MAP	"	"	0	++
	"	5% MAP	"	"	0	++
	"	5% MAPOL	"	"	0	++

Experiment 2: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations after Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Tanning (4 & 6 days) Day	Visible Redness	Visible Tanning
Lawhorn	Back	Control	2½ hours	4th day	++	++
27642	"	1% MAP	"	"	+++	++
(5)	"	3% MAP	"	"	+++	±
	"	5% MAP	"	"	+	++
	"	5% MAPOL	"	"	+++	±
	Back	Control	2½ hours	6th day	++	++
	"	1% MAP	"	"	+	++
	"	3% MAP	"	"	++	++
	"	5% MAP	"	"	0	++
	"	5% MAPOL	"	"	+	++
Traylor	Back	Control	2½ hours	4th day	+++	0
26627	"	1% MAP	"	"	0	++
(6)	"	3% MAP	"	"	0	+
	"	5% MAP	"	"	0	+
	"	5% MAPOL	"	"	0	++
	Back	Control	2½ hours	6th day	+	++
	"	1% MAP	"	"	0	++
	"	3% MAP	"	"	0	+
	"	5% MAP	"	"	0	++
	"	5% MAPOL	"	"	0	++

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EXPERIMENT 3: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations after 15 Minutes Swimming

1% MAP + 15 minutes swimming + 2½ hours sun exposure
3% MAP + 15 minutes swimming + 2½ hours sun exposure
5% MAP + 15 minutes swimming + 2½ hours sun exposure
5% MAPOL + 15 minutes swimming + 2½ hours sun exposure

Object: After 1 application of 1, 3 and 5% MAP or 5% MAPOL and 15 minutes swimming, evaluate:

a) the degree of protection after 2½ hours sun exposure.
b) the degree of tan after 4 days and 6 days.
c) the affinity of MAP or MAPOL to remain on stratum corneum after swimming.

Number of Subjects: 6

Results: 1, 3 and 5% MAP and 5% MAPOL failed to show any protection against 2½ hours of sun exposure. The degree of erythema observed in the 1, 3 and 5% MAP and 5% MAPOL treated areas was practically the same as that observed in the control (untreated) areas.

Comments & Conclusions: An exposure of 2½ hours to solar radiation on a clear bright day to the back of fair-skinned Caucasians lying in the horizontal position with the surrounding temperature around 97-99°F, constitutes an intense challenge. This amount of solar radiation is approximately equal to 15 times the minimal erythema dose. Obviously, after 15 minutes swimming, most of the protective film of MAP was washed off and the concentration of the residual MAP that remained on the surface of the horny layer was not enough to provide protection against 2½ hours sun exposure.

1, 3 and 5% MAP and 5% MAPOL preparations, after one application and 15 minutes swimming, cannot provide any effective protection when the sun exposure period lasts for 2½ hours.

Experiment 3: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations after 15 Minutes Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Hopkins	Back	Control	2½ hours	+++	0	13	0	
27691	"	1% MAP	"	+++	0	12	0	
(1)	"	3% MAP	"	+++	0	13	0	
	"	5% MAP	"	+++	0	18	0	
	"	5% MAPOL	"	+++	0	15	0	
	Left Leg	Control	"	++	0	10	0	
	"	5% MAP	"	++	0	9	10	
	Right Leg	Control	"	++	0	4	0	
	"	5% MAPOL	"	++	0	5	0	

Experiment 3: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations after 15 Minutes Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Roberts	Back	Control	2½ hours	+++	0	13	0	
27035	"	1% MAP	"	+++	0	13	0	
(2)	"	3% MAP	"	+++	0	15	0	
	"	5% MAP	"	+++	0	17	0	
	"	5% MAPOL	"	+++	0	15	0	
	Left Leg	Control	"	+++	0	19	0	
	"	5% MAP	"	+++	0	18	0	
	Right Leg	Control	"	+++	0	22	0	
	"	5% MAPOL	"	+++	0	17	0	

Experiment 3: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations after 15 Minutes Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Meade	Back	Control	2½ hours	+++	0	11	0	
27821	"	1% MAP	"	+++	0	10	0	
(3)	"	3% MAP	"	+++	0	12	0	
	"	5% MAP	"	+++	0	16	0	
	"	5% MAPOL	"	+++	0	14	0	
	Left Leg	Control	"	+++	0	9	0	
	"	5% MAP	"	+++	0	7	20	
	Right Leg	Control	"	+++	0	5	0	
	"	5% MAPOL	"	+++	0	6	0	

Experiment 3: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations after 15 Minutes Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Miller	Back	Control	2½ hours	++++	0	18	0	
27229	"	1% MAP	"	++++	0	19	0	
(4)	"	3% MAP	"	++++	0	20	0	
	"	5% MAP	"	++++	0	21	0	
	"	5% MAPOL	"	++++	0	21	0	
	Left Leg	Control	"	+++	0	9	0	
	"	5% MAP	"	+++	0	6	33	
	Right Leg	Control	"	+++	0	6	0	
	"	5% MAPOL	"	+++	0	7	0	

Experiment 3: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations after 15 Minutes Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Fisher	Back	Control	2½ hours	+++	0	14	0	
27377	"	1% MAP	"	+++	0	12	0	
(5)	"	3% MAP	"	+++	0	16	0	
	"	5% MAP	"	+++	0	14	0	
	"	5% MAPOL	"	+++	0	15	0	
	Left Leg	Control	"	+++	0	8	0	
	"	5% MAP	"	+++	0	8	0	
	Right Leg	Control	"	+++	0	8	0	
	"	5% MAPOL	"	++	25	7	13	

Experiment 3: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations after 15 Minutes Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Roe	Back	Control	2½ hours	+++	0	14	0	
26948	"	1% MAP	"	+++	0	18	0	
(6)	"	3% MAP	"	+++	0	14	0	
	"	5% MAP	"	+++	0	15	0	
	"	5% MAPOL	"	+++	0	14	0	
	Left Leg	Control	"	+++	0	11	0	
	"	5% MAP	"	+++	0	10	0	
	Right Leg	Control	"	+++	0	9	0	
	"	5% MAPOL	"	+++	0	8	0	

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EXPERIMENT 3: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations after 15 Minutes Swimming

Object: After 1 application of 1, 3 and 5% MAP and 5% MAPOL and 15 minutes swimming, plus 2½ hours sun exposure, evaluate the degree of tanning at the end of 4 and 6 days.

Results: In most of the subjects, at the end of 4 days after exposure, no visible tanning was detectable because the residual erythema was still quite intense in all the exposed areas. After 6 days, however, the tanning response was detectable in at least 5 out of 6 subjects. In areas treated with 1, 3 and 5% MAP and 5% MAPOL, the tanning response was practically identical to that observed in the control (untreated) areas that had been exposed. The scaling or desquamation of the horny layer was remarkably evident in the control areas. 5% MAP treated areas, however, did not show any desquamation.

Comments & Conclusions: To promote tanning, over exposure to solar radiation must be avoided. Excessive exposure to solar radiation not only produces marked sunburn, desquamation, edema and intense tenderness of skin, but also poor tanning. Ideal tanning of the skin can be achieved without evoking the adverse effects of sunburn reaction if the skin is well protected and only if there is minimal erythema.

Experiment 3: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations after 15 Minutes Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Tanning (4 & 6 Days) Day	Visible Redness	Visible Tanning
Hopkins 27691 (1)	Back	Control	2½ hours	4th day	+++	0
	"	1% MAP	"	"	+++	0
	"	3% MAP	"	"	+++	0
	"	5% MAP	"	"	+++	0
	"	5% MAPOL	"	"	+++	0
	Back	Control	2½ hours	6th day	+	++
	"	1% MAP	"	"	+	++
	"	3% MAP	"	"	+	++
	"	5% MAP	"	"	+	++
	"	5% MAPOL	"	"	+	++
Roberts 27035 (2)	Back	Control	2½ hours	4th day	+++	+
	"	1% MAP	"	"	+++	+
	"	3% MAP	"	"	+++	+
	"	5% MAP	"	"	+++	+
	"	5% MAPOL	"	"	+++	+
	Back	Control	2½ hours	6th day	++	+++
	"	1% MAP	"	"	+	+++
	"	3% MAP	"	"	±	+++
	"	5% MAP	"	"	+	+++
	"	5% MAPOL	"	"	0	+++

Experiment 3: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations after 15 Minutes Swimming

Subject & Prison #	Site	Preparations	Period of Sun Exposure	Degree of Tanning (4 & 6 Days) Day	Visible Redness	Visible Tanning
Meade	Back	Control	2½ hours	4th day	++	++
27821	"	1% MAP	"	"	++	+
(3)	"	3% MAP	"	"	++	+
	"	5% MAP	"	"	++	+
	"	5% MAPOL	"	"	++	+
	Back	Control	"	6th day	0	++
	"	1% MAP	"	"	0	++
	"	3% MAP	"	"	0	++
	"	5% MAP	"	"	0	++
	"	5% MAPOL	"	"	0	++
Miller	Back	Control	2½ hours	4th day	+++	0
27229	"	1% MAP	"	"	+++	0
(4)	"	3% MAP	"	"	+++	0
	"	5% MAP	"	"	+++	0
	"	5% MAPOL	"	"	+++	0
	Back	Control	"	6th day	±	++
	"	1% MAP	"	"	±	++
	"	3% MAP	"	"	±	++
	"	5% MAP	"	"	±	++
	"	5% MAPOL	"	"	±	++

Experiment 3: Evaluation of 1, 3 and 5% MAP and 5% MAPOL Preparations after 15 Minutes Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Tanning (4 & 6 Days) Day	Visible Redness	Visible Tanning
Fisher	Back	Control	2½ hours	4th day	+++	++
27377	"	1% MAP	"	"	+	++
(5)	"	3% MAP	"	"	++	++
	"	5% MAP	"	"	+	++
	"	5% MAPOL	"	"	++	++
	Back	Control	2½ hours	6th day	++	++
	"	1% MAP	"	"	0	++
	"	3% MAP	"	"	0	++
	"	5% MAP	"	"	0	++
	"	5% MAPOL	"	"	±	++
Roe	Back	Control	2½ hours	4th day	++++	0
26948	"	1% MAP	"	"	++++	0
(6)	"	3% MAP	"	"	++++	0
	"	5% MAP	"	"	++++	0
	"	5% MAPOL	"	"	++++	0
	Back	Control	2½ hours	6th day	++++	0
	"	1% MAP	"	"	++++	0
	"	3% MAP	"	"	++++	0
	"	5% MAP	"	"	++++	0
	"	5% MAPOL	"	"	++++	0

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EXPERIMENT 4: Effect of Swimming on Sunburn Protecting Properties of Preparations 5% MAP and G.

Object: In experiment # 3, it was observed that after a single application of 5% MAP and [illegible] minutes swimming and sun exposure for 2½ hours, all the six subjects showed no evidence of any protection in the treated areas. Experiment # 4 was therefore designed to test the maximum period of sun exposure that can be tolerated by individuals. After a single application of 5% MAP and preparation G, and a schedule of 15 minutes swimming, evaluate:

a) the degree of protection against sunburn after 20, 40 and 60 minutes sun exposure.

b) the affinity of MAP and G to remain on stratum corneum after swimming.

Number of Subjects: 6

Results: After 1 application and 15 minutes swimming, preparation MAP showed definite and quite satisfactory protection against 40 minutes of sun exposure. This preparation was found to be less effective after 40 minutes sun exposure, i.e., after giving solar radiation over 4 times the minimal erythema dose of the individual.

Preparation G was found to be very effective up to 60 minutes of sun exposure. All the six subjects showed excellent protection against 20, 40 and 60 minutes sun exposure. These observations suggest that preparation G affords better protection than MAP after one application and 15 minutes swimming.

Comments & Conclusions: 1) Preparation MAP affords definite and satisfactory protection up to 45 minutes sun exposure period.

2) Preparation G affords better protection than MAP, i.e., the subjects can tolerate up to 60 minutes of sun exposure after one application and 15 minutes swimming.

3) Both preparations MAP and G have definite affinity to bind or remain adsorbed on the surface of the horny layer.

Experiment 4: Effect of Swimming on Sun Protecting Property of 5% MAP and G

Subject & Prison #	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Perry	Control	20 min.	+	0	6	0	MED 15'
26118	MAP	"	±	50	4	33	
(1)	G	"	0	100	3	50	
	G	"	0	100	3	50	
	Control	40 min.	++	0	12	0	
	MAP	"	+	33	9	25	
	MAP	"	+	33	9	25	
	G	"	0	100	5	58	
	Control	60 min.	+++	0	11	0	
	MAP	"	++	25	9	20	
	G	"	0	100	3	73	

Remarks: 1) Preparation G affords better protection than MAP.

2) Following swimming, MAP begins to lose its protective effect after 60 minutes sun exposure, i.e., after receiving 4 times the minimal erythema dose.

Experiment 4: Effect of Swimming on Sun Protecting Property of 5% MAP and G

Subject & Prison #	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Weldon	Control	20 min.	+	0	6	0	
27421	MAP	"	±	50	3	50	MED 15'
(2)	G	"	0	100	3	50	
	G	"	0	100	3	50	
	Control	40 min.	+++	0	14	0	
	MAP	"	++	25	10	28	
	MAP	"	+	50	7	50	
	G	"	±	75	3	78	
	Control	60 min.	+++	0	16	0	
	MAP	"	++	25	13	20	
	G	"	±	75	4	75	

Remarks: 1) Preparation G affords better protection than MAP.

2) Following swimming, MAP begins to lose its protective effect after 60 minutes sun exposure, i.e., after receiving 4 times the minimal erythema dose.

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Experiment 4: Effect of Swimming on Sun Protecting Property of 5% MAP and G

Subject & Prison #	Preparation	Period of Sun Exposure	Degree of Erythema after 24 hours: Visible Redness	% Protection	Reflectance Difference	% Protection	Remarks
Pruett 25649	CONTROL	20 min.	±	0	5	0	MED 20'
(3)	MAP	"	0	100	0	100	
	G	"	0	100	0	100	
	G	"	0	100	0	100	
	CONTROL	40 min.	++	0	10	0	
	MAP	"	±	66	4	60	
	MAP	"	±	66	4	60	
	G	"	0	100	4	60	
	CONTROL	60 min.	+++	0	16	0	
	MAP	"	+++	0	15	0	
	G	"	0	100	3	81	

Remarks: 1) Preparation G affords better protection than MAP.

2) Following swimming, MAP begins to lose its protective effect after 60 minutes sun exposure, i.e., after receiving 4 times the minimal erythema dose.

Experiment 4: Effect of Swimming on Sun Protecting Property of 5% MAP and G

Subject & Prison #	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	Reflectance Difference	% Protection	Remarks
Barton	CONTROL	20 min.	+	0	6	0	MED 20'
24846	MAP	"	±	50	3	50	
(4)	G	"	±	50	3	50	
	G	"	±	50	3	50	
	CONTROL	40 min.	++	0	9	0	
	MAP	"	+	33	6	33	
	MAP	"	+	33	6	33	
	G	"	+	33	6	33	
	CON	60 min.	+++	0	14	0	
	MAP	"	++	25	11	20	
	G	"	+	50	4	72	

Remarks: 1) Preparation G affords better protection than MAP.

2) Following swimming, MAP begins to lose its protective effect after 60 minutes sun exposure, i.e., after receiving 4 times the minimal erythema dose.

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Experiment 4: Effect of Swimming on Sun Protecting Property of 5% MAP and G

Subject & Prison #	Preparation	Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	Reflectance Difference	% Protection	Remarks
Blevins 27879	CONTROL	20 min.	+	0	7	0	MED 20'
(5)	MAP	"	±	50	4	43	
	G	"	+	0	6	14	
	G	"	±	50	6	14	
	CONTROL	40 min.	++	0	8	0	
	MAP	"	+	50	6	25	
	MAP	"	+	50	4	50	
	G	"	±	66	5	37	
	CONTROL	60 min.	++	0	15	0	
	MAP	"	+	50	11	30	
	G	"	+	50	5	66	

Remarks: 1) Preparation G affords better protection than MAP.

2) Following swimming, MAP begins to lose its protective effect after 60 minutes sun exposure, i.e., after receiving 4 times the minimal erythema dose.

Experiment 4: Effect of Swimming on Sun Protecting Property of 5% MAP and G

Subject & Prison #	Preparation	Period of Sun Exposure	Visible Redness	% Protection	Reflectance Difference	% Protection	Remarks
Terrel 27884	CONTROL	20 min.	+	0	7	0	MED 20'
(6)	MAP	"	±	50	4	43	
	G	"	±	50	4	43	
	G	"	±	50	5	30	
	CONTROL	40 min.	++	0	8	0	
	MAP	"	±	66	4	50	
	MAP	"	±	66	4	50	
	G	"	±	66	5	38	
	CONTROL	60 min.	++	0	11	0	
	MAP	"	++	0	11	0	
	G	"	+	33	7	37	

Remarks: 1) Preparation G affords better protection than [illegible].

2) Following swimming, MAP begins to lose its [illegible]ive effect after 60 minutes sun exposure, i.e., after receiving 4 times the minimal erythema dose.

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EXPERIMENT 5: Comparison of MAP, Formula D5 and Formula F3 Preparations

MAP + sunlight 1 and 2 hours without exercise and swimming
D5 + sunlight and 2 hours without exercise and swimming
F3 + sunlight 1 and 2 hours without exercise and swimming

Object: After 1 application of MAP, D5 and F3 evaluate:

a) the degree of erythema after 1 hour and 2 hours sun exposure.

b) to ascertain whether preparation D5, that contains 5% PABA and emollient and a surfactant in alcohol, was as effective as 5% MAP.

Number of Subjects: 4

Results:

1) After application and 60 minutes sun exposure, MAP and D5 preparations afforded significant protection ranging from 75 to 100%. Preparation F3, although it protected 2 subjects quite satisfactorily, proved to be less effective than MAP or D5.

2) After 1 application and 120 minutes sun exposure, MAP afforded quite satisfactory protection ranging from 40 to 100%. D5 was slightly less effective than MAP. Preparation F3 was certainly less effective than MAP or D5.

Conclusions:

1) MAP and D5 preparations appeared to be equally effective, both provided significant protection against 60 minutes sun exposure. Both of these preparations gave quite satisfactory protection up to 120 minutes sun exposure. F3 preparation, although it afforded protection up to 60 minutes, was certainly less effective than MAP or D5.

2) The incorporation of an emollient and a solubilizer (5% oleyl-alcohol and 2% polyoxyethylene oleyl ether) did not radically alter the substantivity property of preparation MAP. This preparation appeared to be almost as effective as MAP.

Experiment 5: Comparison of 5% MAP, Formula MAP (D5) and Formula F3 Preparations after 1 application and Sun Exposure Without Exercise and Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Washa 22987 (1)	Back	Control	1 hour	++	0	12	0	
	"	MAP	"	0	100	0	100	IPD
	"	D5	"	0	100	0	100	IPD
	"	F3	"	0	100	0	100	IPD
	Back	Control	2 hours	+++	0	15	0	
	"	MAP	"	0	100	0	100	IPD
	"	D5	"	+	50	6	60	
	"	F3	"	+	50	6	60	
	Left Leg	Control	1 hour	+	0	6	0	
	"	MAP	"	0	100	2	70	
	Right Leg	Control	2 hours	++	0	10		
	"	MAP	"	+	33	6	40	

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Experiment 5: Comparison of 5% MAP, Formula MAP (D5) and Formula F3 Preparations after 1 Application and Sun Exposure Without Exercise and Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Griffin	Back	Control	1 hour	+++	0	14	0	
27304	"	MAP	"	±	75	8	43	
(2)	"	D5	"	±	75	8	43	
	"	F3	"	+	50	8	43	
	Back	Control	2 hours	++++	0	16	0	
	"	MAP	"	+	60	10	40	
	"	D5	"	++	40	12	25	
	"	F3	"	+++	20	13	20	
	Left Leg	Control	1 hour	++++	0	13	0	
	"	D5	"	0	100	0	100	
	Right Leg	Control	2 hours	++++	0	15	0	
	"	D5	"	+	60	10	33	

Experiment 5: Comparison of 5% MAP, Formula MAP (D5) and Formula F3 Preparations after 1 Application and Sun Exposure Without Exercise and Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Laird	Back	Control	1 hour	+++	0	19	0	
26156	"	MAP	"	±	75	6	68	
(3)	"	D5	"	±	75	5	74	
	"	F3	"	+	50	11	40	
	Back	Control	2 hours	++++	0	20	0	
	"	MAP	"	+	60	10	50	
	"	D5	"	+	60	13	35	
	"	F3	"	++	40	15	25	
	Left Leg	Control	1 hour	++	0	7	0	
	"	D5	"	0	100	1	86	
	Right Leg	Control	2 hours	+++	0	7	0	
	"	D5	"	0	100	2	72	

Experiment 5: Comparison of 5% MAP, Formula MAP (D5) and Formula F3 Preparations after 1 Application and Sun Exposure Without Exercise and Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Wise	Back	Control	1 hour	++	0	12	0	
25468	"	MAP	"	0	100	3	100	IPD
(4)	"	D5	"	0	100	3	100	IPD
	"	F3	"	0	100	0	100	IPD
	Back	Control	2 hours	+++	0	15	0	
	"	MAP	"	0	100	3	100	IPD
	"	D5	"	±	75	6	75	IPD
	"	F3	"	++	25	8	47	IPD
	Left Leg	Control	1 hour	+	0	6	0	
	"	F3	"	0	100	2	70	
	Right Leg	Control	2 hours	++	0	10	0	
	"	F3	"	+	66	6	40	

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EXPERIMENT 5: Comparison of 5% MAP, Formula MAP (D5) and Formula F3 Preparations

Object: After 1 application of 5% MAP, D5 and F3 preparations and sun exposure for 1 and 2 hours without any exercise or swimming, evaluate the degree of tanning after 96 hours post exposure.

Results: In this experiment, 5% MAP and D5 preparations were found to be equally effective and provided very satisfactory protection against 60 and 120 minutes sun exposure. F3 preparation, although it afforded protection up to 60 minutes sun exposure, was found to be less effective than MAP or D5 preparations. In spite of this significant protection by MAP and D5, the tanning response was not inhibited. On the contrary, the degree of tanning that was observed in areas treated with MAP and D5 was of the same order as that observed in the control (untreated) exposed areas. It is interesting to see that at the end of 96 hours after sun exposure, the control (untreated) areas still showed a significant erythema, particularly in those areas that received sun exposure for 120 minutes. Contrary to this, the MAP and D5 treated areas showed practically no or minimal residual erythema at 96 hours. Thus, in MAP and D5 treated areas, tanning has proceeded without the discomforts of sunburn. The F3 treated areas, on the other hand, had residual sunburn at 96 hours which was decidedly more than that observed in MAP or D5 treated areas.

Conclusions: Both MAP and D5 preparations reveal two most important and essential properties:

1) They provide significant protection against solar radiation and prevent sunburn.
2) Both allow the desired tanning without the discomforts of sunburn.

Experiment 5: Comparison of 5% MAP, Formula MAP (D5) and Formula F3 Preparations after Sun Exposure without Exercise or Swimming

Subject & Prison #	Site	Preparation	Sun Exposure	Degree of Tanning after 96 Hours: Visible Redness	Visible Tanning
Washa 22987 (1)	Back	Control	60 minutes	+	0
	"	MAP	"	0	+
	"	D5	"	0	+
	"	F3	"	0	+
	Back	Control	120 minutes	+++	0
	"	MAP	"	±	++
	"	D5	"	+	++
	"	F3	"	+	++
Griffin 27304 (2)	Back	Control	60 minutes	+	++
	"	MAP	"	0	++
	"	D5	"	0	++
	"	F3	"	±	++
	Back	Control	120 minutes	+++	++
	"	MAP	"	±	++
	"	D5	"	±	++
	"	F3	"	+	++

Experiment 5: Comparison of 5% MAP, Formula MAP (D5) and Formula F3 Preparations after Sun Exposure without Exercise or Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Tanning after 96 hours: Visible Redness	Visible Tanning
Laird	Back	Control	60 minutes	++	+
26156	"	MAP	"	0	+
(3)	"	D5	"	0	+
	"	F3	"	+	+
	Back	Control	120 minutes	+++	++
	"	MAP	"	+	++
	"	D5	"	+	++
	"	F3	"	++	+
Wise	Back	Control	60 minutes	+	++
25468	"	MAP	"	0	+
(4)	"	D5	"	0	+
	"	F3	"	0	+
	Back	Control	120 minutes	++	++
	"	MAP	"	±	++
	"	D5	"	±	++
	"	F3	"	±	++

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EXPERIMENT 6: Comparison of 5% MAP, MAP (D5) and F3 Preparations after Exercise

MAP + 30 minutes exercise + 45 minutes and 90 minutes sun exposure
D5 + 30 minutes exercise + 45 minutes and 90 minutes sun exposure
F3 + 30 minutes exercise + 45 minutes and 90 minutes sun exposure

Object: After 1 application of MAP, D5 or F3 preparations and 30 minutes exercise, evaluate:

a) the degree of erythema after 45 minutes and 90 minutes sun exposure.

b) the degree of tan between 3 and 5 days.

Exercise Schedule: Running and walking 10 minutes, calisthenics 10 minutes, and basketball 10 minutes. The subjects wore clothes during exercise after application of preparations.

Number of Subjects: 4

Results:

1) After 1 application and a schedule of 30 minutes exercise with heavy sweating, MAP and D5 preparations provided 100% protection when sun exposure period was limited to 45minutes. Under the same conditions, F3 preparation was not as effective as MAP or D5 in affording protection.

2) MAP and D5 preparations also provided quite satisfactory protection for a period of 90 minutes sun exposure. F3 preparation, however, was not effective in providing protection against 90 minutes sun exposure.

Conclusion:

1) The frictional contact with the clothes and the sweating during exercise period did not remove the protective film of PABA.

2) Map and D5 preparations appeared to be equally effective. Both provided significant protection against 45 and 90 minutes sun exposure.

3) F3 preparation was only effective up to 45 minutes sun exposure.

Experiment 6: Comparison of 5% MAP, MAP (D5) and F3 Preparations after One Application and Sun Exposure with 30 Minutes Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Reflectance	% Reflectance	% Protection	Remarks
Mathews	Back	Control	45 min.	+			0	
25788	"	MAP	"	0	100	3	73	IPD
(1)	"	D5	"	0	100	4	50	IPD
	"	F3	"	±	50	8	0	IPD
	Back	Control	90 min.	+++	0	14	0	
	"	MAP	"	+	50	8	43	
	"	D5	"	+	50	12	16	
	"	F3	"	+++	0	17	0	
	Left Leg	Control	45 min.	+	0	11	0	
	"	MAP	"	0	100	2	80	
	Right Leg	Control	90 min.	++	0	11	0	
	"	MAP	"	0	100	6	46	

Experiment 6: Comparison of 5% MAP, MAP (D5) and F3 Preparations after One Application and Sun Exposure with 30 Minutes Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Saunders	Back	Control	45 min.	+++	0	14	0	
27165	"	MAP	"	0	100	5	65	
(2)	"	D5	"	0	100	5	65	
	"	F3	"	+	50	9	36	
	Back	Control	90 min.	++++	0	15	0	
	"	MAP	"	±	80	7	50	
	"	D5	"	+	60	11	27	
	"	F3	"	++	40	13	14	
	Left Leg	Control	45 min.	+	0	7	0	
	"	D5	"	0	100	0	100	
	Right Leg	Control	90 min.	++	100	7	0	
	"	D5	"	0	100	0	100	

Experiment 6: Comparison of 5% MAP, MAP (D5) and F3 Preparations after One Application and Sun Exposure with 30 Minutes Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Urry	Back	Control	45 min.	+++	0	16	0	
27087	"	MAP	"	0	100	3	80	
(3)	"	D5	"	0	100	3	80	
	"	F3	"	0	100	2	88	
	Back	Control	90 min.	++++	0	18	0	
	"	MAP	"	++	40	13	30	
	"	D5	"	+	60	10	45	
	"	F3	"	+	60	6	66	
	Left Leg	Control	45 min.	+	0	9	0	
	"	D5	"	0	100	0	100	
	Right Leg	Control	90 min.	+++	0	12	0	
	"	D5	"	0	100	2	80	

Experiment 6: Comparison of 5% MAP, MAP (D5) and F3 Preparations after One Application and Sun Exposure with 30 Minutes Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Hanna	Back	Control	45 min.	+	0	10	0	
25997	"	MAP	"	0	100	4	100	IPD
(4)	"	D5	"	0	100	5	100	IPD
	"	F3	"	0	100	1	100	IPD
	Back	Control	90 min.	+++	0	17	0	
	"	MAP	"	0	100	5	70	IPD
	"	D5	"	±	75	5	70	
	"	F3	"	++	25	11	35	
	Left Leg	Control	45 min.	+++	0	10	0	
	"	F3	"	0	100	2	80	
	Right Leg	Control	90 min.	+++	0	11	0	
	"	F3	"	0	100	5	56	

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EXPERIMENT 6: Comparison of 5% MAP, MAP (D5) and F3 Preparations after Exercise

Object: After 1 application of MAP, D5 or F3 preparations and 30 minutes exercise plus 45 and 90 minutes sun exposure, evaluate:

a) the degree of tanning in control (untreated) areas after 96 hrs.

b) the degree of tanning in MAP, D5 and F3 treated areas after 96 hrs.

Results: When compared to the control (untreated) areas, MAP and D5 preparations provided significant protection against sunburn after 45 and 90 minutes sun exposure. F3 preparation was only effective up to 45 minutes sun exposure. In spite of this significant protection by MAP and D5 preparation, the degree of tanning was of the same magnitude as that observed in the control (untreated) sunburned areas. The tanning response in the F3 treated areas was similar to that observed in the control areas.

Conclusions: Both MAP and D5 preparations reveal two most important properties:

1) Both provide significant protection against solar radiation and prevent sunburn.

2) Both allow the desired tanning without the discomforts of sunburn.

The concentration of the active ingredients in the F3 preparation is not adequate enough to obtain desireable tanning without evoking sunburn.

EXPERIMENT 6: Comparison of 5% MAP, MAP (D5) and F3 Preparations after Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Tanning after 96 Hours: Visible Redness	Visible Tanning
Mathews	Back	Control	45 minutes	±	+
25788	"	MAP	"	0	+
	"	D5	"	0	+
(1)	"	F3	"	0	+
	Back	Control	90 minutes	+	++
	"	MAP	"	±	++
	"	D5	"	±	++
	"	F3	"	±	++
Saunders	Back	Control	45 minutes	+	+
27165	"	MAP	"	0	+
(2)	"	D5	"	0	+
	"	F3	"	+	+
	Back	Control	90 minutes	+++	++
	"	MAP	"	±	++
	"	D5	"	±	++
	"	F3	"	+++	++

EXPERIMENT 6: Comparison of 5% MAP, MAP (D5) and F3 Preparations after Exercise

Subject & Prison #	Site	Preparation	Sun Exposure	Degree of Tanning after 96 Hours: Visible Redness	Visible Tanning
Urry	Back	Control	45 minutes	±	++
27087	"	MAP	"	0	+
(3)	"	D5	"	0	+
	"	F3	"	0	+
	Back	Control	90 minutes	++	
	"	MAP	"	±	
	"	D5	"	0	+
	"	F3	"	0	+
Hanna	Back	Control	45 minutes	0	+
25997	"	MAP	"	0	+
(4)	"	D5	"	0	+
	"	F3	"	0	+
	Back	Control	90 minutes	+	++
	"	MAP	"	±	++
	"	D5	"	±	++
	"	F3	"	++	++

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EXPERIMENT 7: Comparison of 5% MAP, D5 and F3 Preparations after 15 Minutes Swimming

MAP + 15 minutes swimming followed by 45 and 90 minutes sun exposure
D5 + 15 minutes swimming followed by 45 and 90 minutes sun exposure
F3 + 15 minutes swimming followed by 45 and 90 minutes sun exposure

Object: After 1 application of MAP, D5 or F3 preparations and 15 minutes swimming, evaluate:

a) the degree of erythema after 45 and 90 minutes sun exposure.

b) the degree of tan between 3 and 5 days.

Number of Subjects: 4

Results: After 1 application and 15 minutes swimming, a satisfactory protective effect of MAP or D5 was observed up to 45 minutes sun exposure period. Back areas exposed to 90 minutes solar radiation revealed less satisfactory protective effect of MAP and D5. F3 preparation was not exerting the desireable protective effect after 45 or 90 minutes sun exposure. The concentration of the active ingredient PABA in this preparation is not optimum to provide the desired protective effect.

Conclusions:

1) After 1 application and 15 minutes swimming, MAP and D5 preparations can provide adequate protection against 45 minutes sun exposure.

2) It is apparent that the active ingredient PABA is definitely conjugating chemically with the horny layer. However, the amount of PABA that is left on the surface of the horny layer after 15 minutes swimming is not sufficient to offer satisfactory protection against 90 minutes solar radiation. Exposure of the back for 90 minutes of solar radiation, particularly in the horizontal position, is certainly a big challenge and to provide complete protection against such an intense radiation, one must have more molecules of PABA left on the surface of the skin after swimming.

Experiment 7: Comparison of 5% MAP, D5 and F3 Preparations after 15 Minutes Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Chandler	Back	Control	45 min.	++	0	12	0	
27873	"	MAP	"	+	33	8	33	
(1)	"	D5	"	+	33	6	50	
	"	F3	"	+++	0	10	17	
	Back	Control	90 min.	+++	0	17	0	
	"	MAP	"	++	25	15	12	
	"	D5	"	++	25	11	35	
	"	F3	"	+++	0	12	30	
	Left Leg	Control	45 min.	++	0	9	0	
	"	MAP	"	+	33	7	23	
	Right Leg	Control	90 min.	+	0	13	0	
	"	MAP	"	+	0	13	0	

Experiment 7: Comparison of 5% MAP, D5 and F3 Preparations after 15 Minutes Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Trimble	Back	Control	45 min.	+++	0	15	0	
27872	"	MAP	"	++	25	14	10	
(2)	"	D5	"	++	25	7	54	
	"	F3	"	++	25	12	20	
	Back	Control	90 min.	++++	0	17	0	
	"	MAP	"	+++	20	17	0	
	"	D5	"	+++	20	13	25	
	"	F3	"	+++	20	15	12	
	Left Leg	Control	45 min.	+++	0	10	0	
	"	D5	"	++	25	8	20	
	Right Leg	Control	90 min.	+++	0	10	0	
	"	D5	"	+++	0	10	0	

Experiment 7: Comparison of 5% MAP, D5 and F3 Preparations after 15 Minutes Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Pace 27372 (3)	Back	Control	45 min.	+	0	8	0	
	"	MAP	"	±	50	6	25	
	"	D5	"	+	0	9	0	
	"	F3	"	+	0	8	0	
	Back	Control	90 min.	+++	0	11	0	
	"	MAP	"	++	25	6	45	
	"	D5	"	++	25	13	0	
	"	F3	"	+++	0	13	0	
	Left Leg	Control	45 min.	++	0	9	0	
	"	D5	"	+	33	3	66	
	Right Leg	Control	90 min.	++	0	14	0	
	"	D5	"	++	0	11	20	

Experiment 7: Comparison of 5% MAP, D5 and F3 Preparations after 15 Minutes Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Sieling	Back	Control	45 min.	+	0	8	0	
27758	"	MAP	"	0	100	3	100	IPD
(4)	"	D5	"	0	100	3	100	IPD
	"	F3	"	0	100	2	100	IPD
	Back	Control	90 min.	++	0	12	0	
	"	MAP	"	0	100	4	66	
	"	D5	"	±	66	5	58	
	"	F3	"	++	0	6	50	
	Left Leg	Control	45 min.	++	0	7	0	
	"	F3	"	+	33	2	70	
	Right Leg	Control	90 min.	+++	0	9	0	
	"	F3	"	++	25	7	20	

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EXPERIMENT 7: Comparison of 5% MAP, D5 and F3 Preparations after 15 Minutes Swimming

Object: After 1 application of MAP, D5 or F3 preparations and 15 minutes swimming followed by 45 and 90 minutes sun exposure, evaluate the degree of tanning at the end of 96 hours.

Results: After 1 application and 15 minutes swimming, the protective effect of MAP or D5 was observed only up to 45 minutes sun exposure. After 90 minutes solar irradiation, MAP and D5 preparations revealed no satisfactory protective effect. F3 preparation was not exerting any satisfactory protective effect after 45 and 90 minutes sun exposure. However, one important observation is worth pointing out in this experiment, and that is the remarkable difference of the residual erythema after 96 hours between the control (untreated) areas and the areas treated with MAP or D5. After 4 days, the control (untreated) areas, particularly those that received 90 minutes sun exposure, showed still significant redness, whereas the MAP and D5 treated areas had no residual redness. The tanning response was practically identical in control (untreated) areas and areas treated with MAP, D5 or F3 preparations.

Conclusions: Desireable tanning of the skin can be achieved without producing significant sunburn response. MAP and D5 preparations minimize the sunburn response and yet allow tanning to proceed.

EXPERIMENT 7: Comparison of 5% MAP, D5 and F3 Preparations after 15 Minutes Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Tanning after 96 Hours: Visible Redness	Visible Tanning
Chandler	Back	Control	45 minutes	±	++
27873	"	MAP	"	0	++
(1)	"	D5	"	0	++
	"	F3	"	+	++
	Back	Control	90 minutes	++++	+++
	"	MAP	"	±	+++
	"	D5	"	±	+++
	"	F3	"	++	+++
Trimble	Back	Control	45 minutes	++	+
27872	"	MAP	"	±	+
(2)	"	D5	"	±	+
	"	F3	"	+	+
	Back	Control	90 minutes	++++	++
	"	MAP	"	++	++
	"	D5	"	++	++
	"	F3	"	++	++

Experiment 7: Comparison of 5% MAP, D5 and F3 Preparations after 15 Minutes Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Tanning after 96 Hours: Visible Redness	Visible Tanning
Pace	Back	Control	45 minutes	0	+
27372	"	MAP	"	0	+
(3)	"	D5	"	0	+
	"	F3	"	0	+
	Back	Control	90 minutes	0	++
	"	MAP	"	0	++
	"	D5	"	0	++
	"	F3	"	0	++
Sieling	Back	Control	45 minutes	0	+
27758	"	MAP	"	0	+
(4)	"	D5	"	0	+
	"	F3	"	0	+
	Back	Control	90 minutes	±	++
	"	MAP	"	0	++
	"	D5	"	±	++
	"	F3	"	±	++

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EXPERIMENT 8: Comparison of 5% MAP, Escalol 506 (G) and UVAL Preparations without Exercise or Swimming

MAP + 60 and 120 minutes sun exposure without exercise or swimming
G + 60 and 120 minutes sun exposure without exercise or swimming
UVAL + 60 and 120 minutes sun exposure without exercise or swimming

Object: After 1 application of MAP, G or UVAL preparations and without subjecting the volunteers to any schedule of exercise or swimming, evaluate:

a) the degree of protection after 60 and 120 minutes sun exposure.
b) the degree of tanning after 4 days.
c) the affinity of MAP, G or UVAL to remain on the stratum corneum under dry and hot sunny weather conditions.

Number of Subjects: 2

Results: After 1 application of MAP without any exercise or swimming, MAP was found to be most effective preparation that provided very satisfactory protection. Preparation G also provided the desired protection but in comparison with MAP was slightly less effective. UVAL was not as effective as G.

Conclusions: Without swimming or exercise, preparations MAP and G provide very satisfactory protection against 60 and 120 minutes sun exposure.

Experiment 8: Comparison of 5% MAP, Escalol 506 (G) and UVAL Preparations without Exercise orSwimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Martin 27882 (1)	Back	CONTROL	60 min.	+	0	7	0	
	"	MAP	"	0	100	4	100	IPD
	"	UVAL	"	0	100	4	100	IPD
	"	G	"	0	100	0	100	
	"	CONTROL	120 min.	+++	0	12	0	
	"	MAP	"	0	100	3	75	
	"	UVAL	"	++	25	6	50	
	"	G	"	±	75	3	75	
	Left Leg	CONTROL	60 min.	++	0	7	0	
	" "	G	"	0	100	0	100	
	Right "	CONTROL	120 min.	+++	0	9	0	
	" "	G	"	+	50	2	78	

Experiment 8: Comparison of 5% MAP, Escalol 506 (G) and UVAL Preparations without Exercise or Swimming

				Degree of Erythema after 24 Hours				
Subject & Prison #	Site	Preparation	Period of Sun Exposure	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Chalmers 24734	Back	CONTROL	60 min.	+++	0	13	0	
(2)	"	MAP	"	0	100	2	85	
	"	UVAL	"	++	25	13	0	
	"	G	"	0	100	3	77	
	"	CONTROL	120 min.	++++	0	15	0	
	"	MAP	"	±	75	5	66	
	"	UVAL	"	+++	0	10	30	
	"	G	"	+	50	5	66	
	Left Leg	CONTROL	60 min.	++	0	7	0	
	"	G	"	0	100	0	100	
	Right Leg	CONTROL	120 min.	++	0	7	0	
	"	G	"	+	33	2	70	

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EXPERIMENT 8: Comparison of 5% MAP, Escalol 506 (G) and UVAL Preparations without Exercise of Swimming

Object: After 1 application of MAP, G or UVAL preparations without exercise or swimming, evaluate: The tanning response at 96 hours after 60 and 120 minutes sun exposure.

Results: Both subjects produced definite tanning in MAP, UVAL and G treated areas. The tanning response was not inhibited, although the erythemal response at 24 hours after 60 and 120 minutes sun exposure was either absent or minimal in areas treated with MAP, UVAL or G.

The degree of tanning in the control (untreated) areas was greater than in MAP, UVAL or G treated areas.

Conclusions: Preparations MAP or G allowed tanning without any visible sunburn.

Experiment 8: Comparison of 5% MAP, Escalol 506 (G) and UVAL Preparations without Exercise or Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Tanning after 96 Hours: Visible Redness	Visible Tanning
Martin 27882	Back	CONTROL	60 min.	0	++
(1)	"	MAP	"	0	+
	"	UVAL	"	0	+
	"	G	"	0	+
	"	CONTROL	120 min.	0	++++
	"	MAP	"	0	+++
	"	UVAL	"	0	+++
	"	G	"	0	+++
Chalmers 24734	"	CONTROL	60 min.	++	++
(2)	"	MAP	"	0	±
	"	UVAL	"	±	±
	"	G	"	0	±
	"	CONTROL	120 min.	+++	++
	"	MAP	"	0	++
	"	UVAL	"	+	++
	"	G	"	0	++

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EXPERIMENT 9: Comparison of 5% MAP, Escalol 506 (G) and UVAL Preparations after Exercise

MAP + 30 minutes exercise + 45 and 90 minutes sun exposure
G + 30 minutes exercise + 45 and 90 minutes sun exposure
UVAL + 30 minutes exercise + 45 and 90 minutes sun exposure

Object: After 1 application of MAP, G or UVAL preparations and 30 minutes exercise, evaluate:

a) the degree of protection after 45 and 90 minutes of sun exposure.
b) the degree of tan after 4 days.
c) the affinity of MAP, G or UVAL to remain on the stratum corneum after a schedule of heavy exercise leading to profuse perspiration under dry and hot sunny weather conditions.

Exercise Schedule: At 90°F, running, walking and calisthenics for 30 minutes. The volunteers had clothes on during exercise.

Number of Subjects: 2

Results: After 1 application and a schedule of 30 minutes exercise with heavy sweating, MAP was found to be the most effective preparation that provided very satisfactory protection against 45 and 90 minutes sun exposure. The commercial product UVAL and the preparation G were, however, not providing the desired protection. These two preparations appeared to be removed as a result of sweating. Interestingly, preparation G was found to be more effective than UVAL.

Conclusions: 1) Preparation MAP certainly appears to have a definite affinity to remain on the surface ofhorny layer after 30 minutes exercise and sweating. Even with the abrasive action and contact of clothing with the skin surface, during exercise, MAP tends to remain on the horny layer and affords excellent protection.

2) Preparation G and UVAL were significantly less protective and appeared to have been removed from the skin surface under experimental conditions.

EXPERIMENT 9: Comparison of 5% MAP, Escalol 506 (G) and UVAL Preparations after Exercise

Subject & Prison#	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Cuzick 27043	Back	CONTROL	45 min.	++++	0	16	0	
	"	MAP	"	±	80	3	80	
	"	UVAL	"	++	40	11	30	
	"	G	"	+++	20	16	0	
	"	CONTROL	90 min.	++++	0	18	0	
	"	MAP	"	+	60	4	78	
	"	UVAL	"	+++	20	14	20	
	"	G	"	+++	20	16	10	
	Left Leg	CONTROL	45 min.	+++	0	7	0	
	"	G	"	0	100	2	70	
	Right Leg	CONTROL	90 min.	+++	0	11	0	
	"	G	"	+	50	5	55	

EXPERIMENT 9: Comparison of 5% MAP, Escalol 506 (G) and UVAL Preparations after Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Redner 26463	Back	CONTROL	45 min.	+++	0	10	0	
	"	MAP	"	0	100	0	100	IPD
	"	UVAL	"	+	50	10	50	IPD
	"	G	"	+	50	10	50	IPD
	Back	CONTROL	90 min.	++++	0	17	0	
	"	MAP	"	0	100	5	100	IPD
	"	UVAL	"	+++	20	15	12	
	"	G	"	+++	20	13	24	
	Left Leg	CONTROL	45 min.	+++	0	13	0	
	"	G	"	0	100	3	77	
	Right Leg	CONTROL	90 min.	+++	0	11	0	
	"	G	"	++	25	8	27	

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EXPERIMENT 9: Comparison of 5% MAP, Escalol 506 (G) or UVAL Preparations after Exercise

Object: After 1 application of MAP, G or UVAL Preparations and 30 minutes exercise followed by 45 and 90 minutes sun exposure, evaluate the degree of tanning in control (untreated), MAP, G and UVAL treated areas after 96 hours post exposure.

Results: MAP, UVAL and G preparations did not inhibit the tanning response. In fact, the tanning in MAP, UVAL and G treated areas was practically of the same degree as that observed in control (untreated) areas. Interestingly, at the end of 96 hours, the control (untreated) areas and the areas treated with UVAL and G that had received 90 minutes sun exposure, still showed appreciable erythema. Contrary to this, the MAP treated areas showed significant tanning without any erythema.

Conclusions: Tanning of the skin can be promoted without evoking any visible sunburn. The preparation MAP afforded complete protection against sunburn and also promoted tanning. Preparation G, like UVAL, which did not show any appreciable protection against sunburn, promoted tanning of the same order as that observed in control (untreated) exposed areas.

EXPERIMENT 9: Comparison of 5% MAP, Escalol 506 (G) and UVAL Preparations after Exercise

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Tanning after 96 Hours: Visible Redness	Visible Tanning
Cuzick 27043	Back	CONTROL	45 minutes	+	+
(1)	"	MAP	"	0	+
	"	UVAL	"	+	+
	"	G	"	+	+
	"	CONTROL	90 minutes	+++	++
	"	MAP	"	0	++
	"	UVAL	"	++	++
	"	G	"	++	++
Redner 26463	Back	CONTROL	45 minutes	±	++
(2)	"	MAP	"	0	+
	"	UVAL	"	0	++
	"	G	"	0	++
	"	CONTROL	90 minutes	++	+++
	"	MAP	"	0	+++
	"	UVAL	"	++	+++
	"	G	"	++	+++

EXPERIMENT 10: Comparison of 5% MAP, Escalol 506 (G) and UVAL Preparations after Swimming

MAP + 15 minutes swimming + 45 and 90 minutes sun exposure.
G + 15 minutes swimming + 45 and 90 minutes sun exposure.
UVAL + 15 minutes swimming + 45 and 90 minutes sun exposure.

Object: After 1 application of MAP, G or UVAL preparations and 15 minutes swimming, evaluate:

a) the degree of protection after 45 minutes and 90 minutes sun exposure.
b) the degree of tan after 4 days.
c) the affinity of MAP, G and UVAL to remain on stratum corneum after swimming.

Swimming Schedule: 15 minutes swimming at 78° F water temperature.

Number of Subjects: 2

Results: After 1 application and 15 minutes swimming, preparation MAP showed definite but not absolute protection against 45 and 90 minutes sun exposure.
The preparation G appears to be more effective than MAP. In one subject it provided complete protection after 45 and 90 minutes sun exposure.
The preparation UVAL provided very little protection against 45 and 90 minutes sun exposure.

Conclusions: Preparation G certainly appears to have definite affinity to remain on the surface of horny layer after swimming. It is not washed away and removed as UVAL.
Preparation MAP was also found to provide definite protection after 45 and 90 minutes sun exposure. It must be stated, however, that complete protection without evoking any sunburn response is not easy to obtain. The number of molecules of PABA that are left attached on the surface of the horny layer are not sufficient enough to provide complete protection.
Preparation UVAL was found to be least effective in providing any protection after swimming.

EXPERIMENT 10: Comparison of 5% MAP, Escalol 506 (G) and UVAL Preparations after Swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Peats 24344	Back	CONTROL	45 min.	++	0	13	0	
	"	MAP	"	++	0	11	16	
	"	UVAL	"	++	0	14	0	
	"	G*	"	0	100	7	100	IPD
	"	CONTROL	90 min.	+++	0	19	0	
	"	MAP	"	++	25	11	40	
	"	UVAL	"	+++	0	17	10	
	"	G*	"	0	100	7	100	
	Left Leg	CONTROL	45 min.	++	0	5	0	
	"	G	"	±	66	1	80	
	Right Leg	CONTROL	90 min.	++	0	5	0	
	"	G	"	+	33	3	40	

*G = Escalol 506

EXPERIMENT 10: Comparison of 5% MAP, Escalol 506 (G) and UVAL Preparations after Swimming

				Degree of Erythema after 24 Hours				
Subject & Prison #	Site	Preparation	Period of Sun Exposure	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Devinney	Back	CONTROL	45 min.	++++	0	21	0	
21:983	"	MAP	"	+++	20	15	30	
	"	UVAL	"	++++	0	20	0	
	"	G*	"	++	40	12	40	
	"	CONTROL	90 min.	++++	0	25	0	
	"	MAP	"	+++	20	19	25	
	"	UVAL	"	++++	0	18	28	
	"	G	"	+++	20	15	40	
	Left Leg	CONTROL	45 min.	+++	0	5	0	
	"	G	"	±	75	0	100	
	Right Leg	CONTROL	90 min.	++	0	10	0	
	"	G	"	++	0	10	0	

*G = Escalol 506

EXPERIMENT 10: Comparison of 5% MAP, Escalol 506 (G) and UVAL Preparations after Swimming

Object: After 1 application of MAP, G or UVAL preparations and 15 minutes swimming, followed by 45 and 90 minutes sun exposure, evaluate the degree of tanning in control (untreated), MAP, G and UVAL treated areas after 96 hours post exposure.

Results: In this experiment, the preparation G was found to be more protective against sunburn than MAP. The control (untreated) areas and areas treated with UVAL exhibited maximum sunburn after 45 and 90 minutes sun exposure and yet, the degree of tanning observed in the control (untreated) areas and in areas treated with MAP, G or UVAL, was practically the same. Thus, without producing any appreciable sunburn, desirable tanning of the skin has been achieved in both the G and MAP treated areas.

Conclusions: Although the preparations MAP and G afford protection against sunburn, they do not inhibit tanning.

EXPERIMENT 10: Comparison of 5% MAP, Escalol 506 (G) and UVAL Preparations after 15 Minutes Swimming

Subject & Prison#	Site	Preparation	Sun Exposure	Degree of Tanning after 96 Hours: Visible Redness	Visible Tanning
Peats 24344	Back	CONTROL	45 minutes	0	++
(1)	"	MAP	"	0	++
	"	UVAL	"	±	++
	"	G	"	0	++
	"	CONTROL	90 minutes	++	+++
	"	MAP	"	±	+++
	"	UVAL	"	++	+++
	"	G	"	0	+++
Devinney 24983	"	CONTROL	45 minutes	++	++
(2)	"	MAP	"	+	++
	"	UVAL	"	++	++
	"	G	"	+	++
	"	CONTROL	90 minutes	+++	+++
	"	MAP	"	++	+++
	"	UVAL	"	+++	+++
	"	G	"	++	+++

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EXPERIMENT 11: Comparison of the Protective Effect of 5% MAP, MAP (D5) and UVAL under Conditions Simulating Normal Usage

Normal conditions of use include swimming, lying on the back and in the prone position, walking and running around the court and sitting in the sun.

Object: After one application of MAP, D5 or UVAL preparations, evaluate the protective qualities of these preparations under the following normal conditions of use:

11:30 a.m.	1 application of MAP preparation on 1/2 the body surface (front as well as back). The other half received either D5 or UVAL preparations. In the center of the back as well as in front a rectangular area (approx. 4" x 5") was left as a control (untreated) area.
11:50 - 12:00	Swimming
12:00 - 12:15	Sitting and incidental drying in the sun.
12:15 - 12:35	Front exposure in the lying position
12:35 - 12:55	Back exposure in the prone position
12:55 - 1:20	Walk around court in the sun
1:20 - 1:30	Run around court in the sun
1:30 - 1:40	Swimming at 86°F water temperature
1:40 - 2:00	Front exposure in the lying position
2:00 - 2:15	Back exposure in prone position

Total exposure: 2 hours and 15 minutes

a) Stationary: Front - 40 minutes, Back - 35 minutes
b) Mobile: 60 minutes

Total time of swimming: 20 minutes

Results: Under these normal conditions of use, preparation MAP was found to provide excellent protection to both the subjects. This preparation certainly appeared to remain on the surface of the skin even after swimming, running, sweating and lying conditions. MAP (D5) also provided fairly satisfactory protection. However, this preparation was slightly less effective than MAP. UVAL was found to be least effective and appeared to have been washed away after swimming and sweating.

(continued)

EXPERIMENT 11: (continued)

Comments: In this experiment, an attempt has been made to evaluate the protective effect of MAP, D5 or UVAL preparations under normal conditions of use. These included at least two swimming periods lasting 10 minutes each, two exposure periods (1 stationary and 1 mobile) lasting for 2 hours and 15 minutes, one walking and running around period of 25 minutes duration that produced considerable sweating. Thus, under such rigorous experimental conditions, both MAP and D5 preparations provided very satisfactory protection. Under similar experimental conditions, UVAL, the best known and widely used commercially available product, was found to be ineffective.

EXPERIMENT 11: Comparison of the Protective Effect of MAP, D5 and UVAL under Normal Conditions of Use

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection
Brown 26623 (1)	Front	None	Stationary 75 min.	++	0	12	0
	"	MAP	Mobile 50 min.	±	66	8	34
	"	UVAL		++	0	12	0
	Back	None		+	0	7	0
	"	MAP		0	100	0	100
	"	UVAL		+	0	6	0
Baker 27553 (2)	Front	None		+++	0	15	0
	"	MAP		±	75	9	40
	"	UVAL		+++	0	15	0
	Back	None		+++	0	13	0
	"	MAP		+	50	8	40
	"	UVAL		+++	0	12	0

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EXPERIMENT 11: Comparison of the Protective Effect of MAP, D5 and UVAL Under Normal Conditions of Use

Subject & Prison #	Site	Preparation	Period of Sun Exposure	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection
Foster 26965	Front	None	Stationary 75 min.	+++	0	15	0
(3)	"	D5	Mobile 50 min.	++	25	12	20
	"	UVAL		+++	0	15	0
	Back	None		++	0	15	0
	"	D5		+	33	12	20
	"	UVAL		++	0	17	0
Nance 27763	Front	None		+	0	12	0
(4)	"	D5		±	50	6	50
	"	UVAL		+	0	11	0
	Back	None		++	0	10	0
	"	D5		±	66	5	50
	"	UVAL		++	0	8	20

ARIZONA STATE PRISON CLINICAL TRIAL

APRIL, 1968

Introduction: The following sunscreen preparations were evaluated:

1. Formula MAP (D5) contains 5% para-aminobenzoic acid in 70% ethyl alcohol, pH 4.8, and 5% w/v oleyl alcohol, as emollient.

2. Formula G, contains 2.5% Eskalol 506 (iso amyl p-n, n-dimethyl aminobenzoate or glyceryl para-aminobenzoate) in 65-70% ethyl alcohol.

3. Modified G, contains approximately 5% Eskalol 506 in 63-65% ethyl alcohol.

4. Skolex, a commercial preparation (J. B. Williams Company, Inc., Cranford, N. J.) contains iso-amyl p-n, n-dimethyl aminobenzoate in a cream base. This preparation was included for comparative studies.

In our experience we find Skolex and UVAL (Dome Laboratories, Westhaven, Conn.) to be better sunscreen preparations than other commercial preparations). UVAL was evaluated in our earlier clinical trial and was found to be less effective than MAP (D5).

Experiments # 1-3 were designed with the following objectives:

1. To establish the relationship between the time of application of sunscreen preparations and sun exposure, i.e., to determine whether there exists an optimum application-exposure interval.

2. To establish whether G or MG, the preparation containing glyceryl para-aminobenzoate are as effective as MAP (D5) under conditions of exercise accompanied by sweating and swimming. The laboratory studies with ultraviolet radiation from artificial light sources have indicated that only para-aminobenzoic acid, when incorporated into ethyl alcohol, can remain on the surface of the horny layer even after repeated washings with water. Other cream bases that are available in the commercial sunscreen preparations, do not allow the light absorbing active ingredient to remain on the stratum corneum. They are either eluted or washed away as a result of sweating and swimming and are, practically speaking, ineffective in affording satisfactory protection against solar radiation.

Experiment 4 was designed to determine whether a single application of test preparations (MAP (D5), G, Skolex) has any prolonged protective effect. If a compound has an affinity to remain adsorbed and chemically bonded with the horny layer, then under normal daily activities of individuals, it should still remain on the skin on the following day and afford protection against sun exposure.

Since MAP (D5) has been shown to remain on the horny layer (both under in vitro and in vivo experiomtnal conditions) after washings with water, it was essential to know whether preparations G and Skolex behaved in the similar manner.

Experiment 5 was designed to determine whether single or multiple applications of preparations MAP (D5), G, and Skolex are essential before sun exposure to obtain the most desirable protection.

Experiment 6 was designed to determine the relative efficacy of preparations MAP (D5), G and Skolex and Snootie (contains 2.5% Escalol 506 in a vanishing cream base, Sea and Ski Corporation, Reno, Nevada) under conditions simulating normal usage. Normal conditions of use include those activities that one would prefer to do on a beach on a bright and warm day.

Experiment 7 was designed to test the relative protective efficacy of preparation G and MAP (D5) against 4,5',8-trimethyl psoralen (TMP)-induced photosensitization. It is well known that when TMP is given orally to human subjects and their skin is exposed to solar radiation, a photosensitization response manifested by an augmented sunburn is observed. It was therefore desirable to know whether these preparations can inhibit or block the TMP-induced cutaneous photosensitization.

Observations gathered on fifty volunteers in this clinical trial lead us to the following conclusions:

1. MAP (D5) and G are by far the best sunscreen preparations that afford reproducible and yet most satisfactory protection against strong intensity of solar radiation. We believe that when no activity (e.g., swimming or exercise with sweating) is planned, a single application of these preparations can provide total protection to individuals for the entire day under hot and dry climatic conditions.

2. MAP (D5) and G preparations are not entirely eluted as a result of profuse sweating. An effective concentration of the light absorbing molecules still remains on the surface of the horny layer after sweating and affords very satisfactory protection against solar radiation.

3. Likewise, after a single application of MAP (D5) and G, and 15 minutes swimming, these preparations are not easily removed and eluted from the surface of the horny layer. They remain either adsorbed or chemically bonded to the surface of the skin and exert very effective protection up to 2 hours of sun exposure.

4. There is no minimum period required for the applied preparation to react with the skin to exert its maximum protective effect against solar radiation.

5. Preparation G appears not to bind chemically with the horny layer. It can be easily removed with repeated washings with water. However, preparation MAP (D5) does have a great affinity to combine chemically and remain bonded with the stratum corneum. This is supported by the observation concerning the prolonged protective effect of MAP after a single application and also by the data on the chemical binding of PABA obtained with isolated pieces of epidermis.

Experiment 1: Relationship between Time of Application and Sun Exposure under Conditions of No Exercise (Sweating) and No Swimming

Preparations to be Evaluated: Formula G, Skolex, Formula MAP (D5) and modified G (MG)

Object:

1) To determine whether there exists an optimum application-exposure interval, i.e., to know whether after one application of these test preparations, there is a minimum time for the applied preparation to react with the skin to exert its maximum protective effect.

2) After one application, compare the efficacy of preparations G, Skolex, MAP (D5) and MG under conditions of no exercise and no swimming.

3) To determine whether G, the preparation containing 2.5% Glyceryl paraaminobenzoate, in 65-70% alcohol, is as effective as MAP (D5).

Number of Subjects: 8

Plan of Experiment:

a) Activity: none, i.e., no exercise or swimming.

b) Taping: Masking tape was applied to the subjects' backs to prevent mixing of test preparations.

c) Application of the test compounds: The test preparations were applied to the appropriate spaces as indicated below:

Subject's Back

Row	Left Side		Right Side	
1	CON	G	CON	G
2	CON	SX	CON	SX
3	D5	CON	D5	CON
Sun Exposure	← 45 Min →		← 90 Min →	

CON = control area — Subjects 1 to 8
G = Formula G — " 1 to 6
SX = Skolex — " 1 to 8
D5 = Preparation MAP — " 1 to 8
MG = Modified G — " 7 and 8

MED: Minimal erythema dose was determined for most of the subjects by exposing the lower portion of the back for 10, 15, 20 & 30 minutes solar radiation.

Expt. 1 (continued)

d) Subjects # 1 to 3: Between 0 to 15 minutes after application of preparations G, Skolex and MAP (D5), the subjects were exposed to 45 and 90 minutes solar radiation.

Subjects # 4 to 6: Thirty minutes after application of preparations G, Skolex and MAP (D5), the subjects were exposed to 45 and 90 minutes of solar radiation.

Subjects # 7 and 8: Thirty minutes after application of preparations Skolex, MAP (D5), and MG, the subjects were exposed for 45 and 90 minutes of solar radiation.

e) Evaluation of the results: Exposed areas were evaluated at 24 and 48 hours after exposure for the degree of erythema and tanning (IPD or immediate pigment darkening response) both by visible gradation of redness and by skin reflectance measurements.

Results: The following conclusions can be drawn from the data obtained on 8 subjects after exposure to solar radiation for 45 and 90 minutes.

1) There is no minimum waiting period required for the applied preparation to react with the skin to exert its maximum sun-protective effect. The subjects (# 1 to 3) that were exposed 0 to 15 minutes after application of Formula G, Skolex, and MAP (D5) showed about the same degree of protection as compared to those subjects (# 4 to 8) that were exposed 30 minutes after application of the compounds.

2) Preparation G appears to be as good as MAP (D5). This observation is very interesting, because it shows that when an ester of PARA is dissolved in 65% C_2H_5OH, the preparation becomes as effective as para-aminobenzoic acid in alcohol. We have previously used several commercially available preparations that contained glyceryl ester of PABA. But these preparations were in no way as effective as MAP (D5). The same active ingredient, when dissolved in C_2H_5OH exhibits very effective sun protection and this we attribute to the quality of the vehicle.

3) Preparation G and MG appear to be equally effective in affording excellent protection under conditions of no exercise and no swimming.

4) Preparation Skolex is less effective than G or MAP (D5).

Experiment # 1: Relationship between Time of Application and Sun Exposure under Conditions of No Exercise (Sweating) and Swimming

Subject & Prison #	Site (Back)	Preparation*	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Hoskins	Row 1	Control	45	±	0	6	0	
27724	"	G	45	±	0	7	0	
(1)	"	Control	90	++	0	19	0	
	"	G	90	0	100	8	56	
	Row 2	Control	45	+	0	10	0	
	"	Skolex	45	+	0	13	0	
	"	Control	90	+++	0	22	0	
	"	Skolex	90	+	50	11	50	
	Row 3	Control	45	+	0	10	0	
	"	MAP (D5)	45	0	100	0	100	
	"	Control	90	++	0	16	0	
	"	MAP (D5)	90	0	100	4	75	

Minimal erythema dose (MED) = 10 minutes

* Interval: 0-15 minutes between application and exposure

Experiment # 1: Relationship between Time of Application and Sun Exposure under Conditions of No Exercise (Sweating) and Swimming

Subject & Prison #	Site (Back)	Preparation*	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Smith	Row 1	Control	45	±	0	3	0	
27603	"	G	45	0	100	0	100	
(2)	"	Control	90	++	0	16	0	
	"	G	90	0	100	8	50	
	Row 2	Control	45	+	0	8	0	
	"	Skolex	45	±	50	4	50	
	"	Control	90	++	0	14	0	
	"	Skolex	90	±	66	3	79	
	Row 3	Control	45	+	0	7	0	
	"	MAP (D5)	45	0	100	0	100	
	"	Control	90	++	0	13	0	
	"	MAP (D5)	90	0	100	4	70	

Minimal erythema dose (MED) = 10 minutes

* Interval: 0-15 minutes between application and exposure

Experiment # 1: Relationship between Time of Application and Sun Exposure under Conditions of No Exercise (Sweating) and Swimming

Subject & Prison #	Site (Back)	Preparation*	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Bills	Row 1	Control	45	+	0	3	0	
27596	"	G	45	0	100	1	100	
(3)	"	Control	90	++	0	7	0	
	"	G	90	0	100	0	100	
	Row 2	Control	45	±	0	4	0	
	"	Skolex	45	±	0	7	0	
	"	Control	90	++	0	14	0	
	"	Skolex	90	±	66	7	50	
	Row 3	Control	45	+	0	7	0	
	"	MAP (D5)	45	0	100	0	100	
	"	Control	90	++	0	11	0	
	"	MAP (D5)	90	0	100	3	73	IPD

Minimal erythema dose (MED) = 10 minutes

* Interval: 0-15 minutes between application and exposure

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Experiment # 1: Relationship between Time of Application and Sun Exposure under Conditions of No Exercise (Sweating) and Swimming

Subject & Prison #	Site (Back)	Preparation*	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Washa	Row 1	Control	45	±	0	7	0	
22987	"	G	45	0	100	5	100	IPD
(4)	"	Control	90	+++	0	16	0	
	"	G	90	±	75	8	50	
	Row 2	Control	45	++	0	10	0	
	"	Skolex	45	+	33	6	40	
	"	Control	90	+++	0	19	0	
	"	Skolex	90	+	50	8	58	
	Row 3	Control	45	++	0	8	0	
	"	MAP (D5)	45	±	66	2	75	
	"	Control	90	+++	0	14	0	
	"	MAP (D5)	90	±	75	7	50	

Minimal erythema dose (MED) = 10 minutes

* Interval: 30 minutes between application and exposure

Experiment # 1: Relationship between Time of Application and Sun Exposure under Conditions of No Exercise (Sweating) and Swimming

Subject & Prison #	Site (Back)	Preparation*	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Weston	Row 1	Control	45	+	0	7	0	
27340	"	G	45	0	100	3	100	IPD
(5)	"	Control	90	++	0	17	0	
	"	G	90	0	100	3	83	
	Row 2	Control	45	++	0	11	0	
	"	Skolex	45	±	66	5	55	
	"	Control	90	++	0	16	0	
	"	Skolex	90	±	66	6	63	
	Row 3	Control	45	++	0	10	0	
	"	MAP (D5)	45	0	100	0	100	
	"	Control	90	++	0	17	0	
	"	MAP (D5)	90	0	100	4	77	

Minimal erythema dose (MED) = 10 minutes

* Interval: 30 minutes between application and exposure

Experiment # 1: Relationship between Time of Application and Sun Exposure under Conditions of No Exercise (Sweating) and Swimming

Subject & Prison #	Site (Back)	Preparation*	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Wells	Row 1	Control	45	++	0	16	0	
28014	"	G	45	0	100	2	100	IPD
(6)	"	Control	90	+++	0	15	0	
	"	G	90	0	100	0	100	
	Row 2	Control	45	+++	0	17	0	
	"	Skolex	45	±	75	6	65	
	"	Control	90	+++	0	20	0	
	"	Skolex	90	±	75	4	80	
	Row 3	Control	45	++	0	15	0	
	"	MAP (D5)	45	0	100	0	100	
	"	Control	90	+++	0	19	0	
	"	MAP (D5)	90	0	100	0	100	

Minimal erythema dose (MED) = 10 minutes

* Interval: 30 minutes between application and exposure

Experiment # 1: Relationship between Time of Application and Sun Exposure under Conditions of No Exercise (Sweating) and Swimming

Subject & Prison #	Site (Back)	Preparation*	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Pruett	Row 1	Control	45	+	0	4	0	
25649	"	MG	45	0	100	0	100	IPD
(7)	"	Control	90	++	0	15	0	
	"	MG	90	0	100	6	100	IPD
	Row 2	Control	45	+	0	10	0	
	"	Skolex	45	0	100	1	100	
	"	Control	90	++	0	15	0	
	"	Skolex	90	0	100	5	66	
	Row 3	Control	45	+	0	8	0	
	"	MAP (D5)	45	0	100	0	100	
	"	Control	90	+++	0	19	0	
	"	MAP (D5)	90	0	100	3	100	IPD

Minimal erythema dose (MED) = 10 minutes

* Interval: 30 minutes between application and exposure

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Experiment # 1: Relationship between Time of Application and Sun Exposure under Conditions of No Exercise (Sweating) and Swimming

Subject & Prison #	Site (back)	Preparation*	Period of Sun Exposure (minutes)	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Gordon	Row 1	Control	45	0	100	3	100	
27457	"	MG	45	0	100	2	100	
(8)	"	Control	90	++	0	7	0	
	"	MG	90	0	100	0	100	
	Row 2	Control	45	+	0	8	0	
	"	Skolex	45	±	50	4	50	
	"	Control	90	+++	0	14	0	
	"	Skolex	90	±	75	7	50	
	Row 3	Control	45	+	0	9	0	
	"	MAP (D5)	45	0	100	4	100	IPD
	"	Control	90	+++	0	14	0	
	"	MAP (D5)	90	0	100	0	100	

Minimal erythema dose (MED) = 10 minutes

* Interval: 30 minutes between application and exposure

Experiment 2: Relationship between Time of Application and Sun Exposure under Conditions of Exercise Accompanied by Sweating

Preparations to be Evaluated: Formula G, Skolex, MAP (D5) and Modified G (MG)

Objects:

1) To determine whether there exists an optimum application-exposure interval as outlined in Experiment I under conditions of 30 minutes exercise.

2) After one application, compare the efficacy of preparations G, Skolex, MAP (D5) and MG under conditions of exercise accompanied by sweating under dry and hot sunny weather.

3) To determine whether G, the preparation containing 2.5% glyceryl para-aminobenzoate in 65-70% alcohol is as effective as MAP (D5).

Number of Subjects: 8

Plan of Experiment:

a) Activity: 30 minutes exercise, outside in the yard before exposure to sun.

b) Taping: Masking tapes were applied to the subjects' backs to prevent mixing of the test preparations.

c) Application of the test compounds: The test preparations were applied to the appropriate spaces as indicated below:

Subject's Back

Row	Left Side		Right Side	
1	CON	G	CON	G
2	CON	SX	CON	SX
3	D5	CON	D5	CON

Sun Exposure: ← 45 Min → ← 90 Min →

CON =	control area	Subjects 1 to 8
G	Formula G	" 1 to 6
SX	Skolex	" 1 to 8
D5	Preparation MAP	" 1 to 8
MG	Modified G	" 7 and 8

MED: Minimal erythema dose was determined for most of the subjects by exposing the lower portion of the back for 10, 15, 20 & 30 minutes solar radiation.

Expt. 2 (continued)

d) Subjects # 1 to 3: Between 0 to 15 minutes after application of preparations G, Skolex and MAP (D5), the subjects carried out a schedule of exercise for 30 minutes that included running and fast walking, calisthenics and basketball. Volunteers did not have any shirts or undershirts on during exercise. After exercise, the subjects were exposed to solar radiation for 45 and 90 minutes.

Subjects # 4 to 6: Thirty minutes after application of preparations G, Skolex, and MAP (D5), the subjects exercised for 30 minutes and then received sun exposure for 45 and 90 minutes.

Subjects # 7 and 8: Thirty minutes after application of Skolex, MAP (D5) and Modified G, the subjects exercised for 30 minutes and then were exposed to solar radiation for 45 and 90 minutes.

e) Evaluation of Results: Exposed areas were evaluated at 24 and 48 hours after sun exposure for the degree of erythema and tanning both by visible gradation of redness and by skin reflectance measurements.

Results: The following conclusions can be drawn from the data obtained on 8 subjects after 45 and 90 minutes sun exposure.

1) There is no minimum waiting period required for the applied preparation to react with the skin to exert its maximum sun-protective effect. The protective effect was practically identical in 0 to 15 minutes and 30 minutes interval groups.

2) Unfortunately, 4 out of 8 subjects that were selected were not easy burners. They had previously acquired tan that prevented their skin against sunburn. The MED in these subjects was over 45 minutes of solar radiation. In spite of this, the data indicate that G, MAP (D5), Skolex and MG are very effective sunscreen preparations. A schedule of 30 minutes exercise accompanied by sweating did not eliminate the excellent protective effect of these preparations.

3) Preparations G and MG were found to be as effective as MAP (D5). This observation is very interesting and adds further support to the thesis that it is the base (eg. the ethyl alcohol) that is most important. Previous studies with commercially available preparations have revealed that glyceryl ester of PABA, if incorporated in bases other than ethyl alcohol, are not effective sunscreen preparations. The same compound becomes very effective when incorporated in an alcohol base.

4) When compared against each other, preparations G, MAP (D5) and MG were found to be equally effective. Skolex appeared to be less effective than G and MAP (D5).

Experiment # 2: Relationship between Time of Application and Sun Exposure in Conjunction with Exercise*

Subject & Prison #	Site (Back)	Preparation	Period of Sun Exposure (Minutes)	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Phillips	Row 1	Control	45	+	0	5	0	
27440	"	G	45	0	100	0	100	
(1)	"	Control	90	+++	0	17	0	
	"	G	90	0	100	0	100	
	Row 2	Control	45	++	0	16	0	
	"	Skolex	45	±	66	3	80	
	"	Control	90	+++	0	16	0	
	"	Skolex	90	±	75	4	75	
	Row 3	Control	45	++	0	13	0	
	"	MAP (D5)	45	0	100	2	85	
	"	Control	90	+++	0	17	0	
	"	MAP (D5)	90	0	100	4	77	

* Interval: 0-15 minutes between application and exercise.

Experiment # 2: Relationship between Time of Application and Sun Exposure in Conjunction with Exercise*

Subject & Prison #	Site (Back)	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	Degree of Erythema after 24 Hours: % Protection	Degree of Erythema after 24 Hours: % Reflectance Difference	Degree of Erythema after 24 Hours: % Protection	Remarks
Cope	Row 1	Control	45	+	0	8	0	Subject did not easily burn. Had IPD in all exposed areas.
27657	"	G	45	0	100	0	100	
(2)	"	Control	90	+	0	9	0	
	"	G	90	0	100	3	100	
	Row 2	Control	45	+	0	6	0	
	"	Skolex	45	0	100	3	100	
	"	Control	90	++	0	11	0	
	"	Skolex	90	0	100	5	100	
	Row 3	Control	45	+	0	9	0	
	"	MAP (D5)	45	0	100	0	100	
	"	Control	90	++	0	11	0	
	"	MAP (D5)	90	0	100	4	100	

* Interval: 0-15 minutes between application and exercise.

Experiment # 2: Relationship between Time of Application and Sun Exposure in Conjunction with Exercise*

Subject & Prison #	Site (Back)	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Noe	Row 1	Control	45	+	0	10	0	
23717	"	G	45	±	50	4	60	
(3)	"	Control	90	+++	0	17	0	
	"	G	90	±	75	7	60	
	Row 2	Control	45	++	0	14	0	
	"	Skolex	45	+	33	11	20	
	"	Control	90	+++	0	17	0	
	"	Skolex	90	+	50	10	41	
	Row 3	Control	45	+	0	12	0	
	"	MAP (D5)	45	0	100	1	100	
	"	Control	90	++	0	15	0	
	"	MAP (D5)	90	0	100	3	80	

* Interval: 0-15 minutes between application and exercise.

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Experiment # 2: Relationship between Time of Application and Sun Exposure in Conjunction with Exercise*

Subject & Prison #	Site (Back)	Preparation	Period of Sun Exposure (Minutes)	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Collins	Row 1	Control	45	+	0	5	0	
27578	"	G	45	±	50	3	40	
(4)	"	Control	90	++	0	12	0	
	"	G	90	0	100	3	75	
	Row 2	Control	45	+	0	11	0	
	"	Skolex	45	±	50	7	37	
	"	Control	90	+++	0	17	0	
	"	Skolex	90	+	50	9	50	
	Row 3	Control	45	+	0	7	0	
	"	MAP (D5)	45	0	100	3	100	IPD
	"	Control	90	+++	0	15	0	
	"	MAP (D5)	90	0	100	3	100	IPD

* Interval: 30 minutes between application and exercise.

Experiment # 2: Relationship between Time of Application and Sun Exposure in Conjunction with Exercise*

Subject & Prison #	Site (Back)	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
House	Row 1	Control	45	0	100	5 (0)	100	Subject did not easily burn. All areas showed IPD response = 5% average reflectance difference.
27843	"	G	45	0	100	4 (0)	100	
(5)	"	Control	90	0	100	10 (5)	0	
	"	G	90	0	100	6 (1)	80	
	Row 2	Control	45	0	100	5 (0)	100	
	"	Skolex	45	0	100	5 (0)	100	
	"	Control	90	+	0	12 (7)	0	
	"	Skolex	90	0	100	5 (0)	100	
	Row 3	Control	45	0	100	6 (1)	100	
	"	MAP (D5)	45	0	100	4 (0)	100	
	"	Control	90	+	0	7	0	
	"	MAP (D5)	90	0	100	0	100	

* Interval: 30 minutes between application and exercise.

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Experiment # 2: Relationship between Time of Application and Sun Exposure in Conjunction with Exercise*

Subject & Prison #	Site (Back)	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Carrier	Row 1	Control	45	±	0	3	0	
26938	"	G	45	0	100	0	100	
(6)	"	Control	90	+	0	4	0	
	"	G	90	0	100	0	100	
	Row 2	Control	45	±	0	7	0	
	"	Skolex	45	0	100	5	100	IPD
	"	Control	90	++	0	14	0	
	"	Skolex	90	0	100	6	100	IPD
	Row 3	Control	45	±	0	6	0	
	"	MAP (D5)	45	0	100	0	100	
	"	Control	90	++	0	12	0	
	"	MAP (D5)	90	0	100	4	100	IPD

* Interval: 30 minutes between application and exercise.

Experiment # 2: Relationship between Time of Application and Sun Exposure in Conjunction with Exercise*

Subject & Prison #	Site (Back)	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Varner	Row 1	Control	45	0	100	7	100	Subject did not burn easily. Most of the controls as well as treated areas had no erythema; IPD was visible in all areas.
27338	"	Modified G	45	0	100	5	100	
(7)	"	Control	90	±	0	7	0	
	"	Modified C	90	0	100	2	100	
	Row 2	Control	45	0	100	6	100	
	"	Skolex	45	0	100	2	100	
	"	Control	90	±	0	9	0	
	"	Skolex	90	0	100	6	100	
	Row 3	Control	45	±	0	6	0	
	"	MAP (D5)	45	0	100	4	100	
	"	Control	90	±	0	6	0	
	"	MAP (D5)	90	0	100	3	100	

* Interval: 30 minutes between application and exercise.

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Experiment # 2: Relationship between Time of Application and Sun Exposure in Conjunction with Exercise*

Subject & Prison #	Site (Back)	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Bradley	Row 1	Control	45	0	100	0	100	
26503	"	Modified G	45	0	100	0	100	
(8)	"	Control	90	+	0	6	0	
	"	Modified G	90	0	100	0	100	
	Row 2	Control	45	±	0	4	0	
	"	Skolex	45	±	0	4	0	IPD
	"	Control	90	+	0	14	0	
	"	Skolex	90	±	50	5	60	
	Row 3	Control	45	±	0	5	0	
	"	MAP (D5)	45	0	100	0	100	
	"	Control	90	+	0	8	0	
	"	MAP (D5)	90	0	100	3	100	IPD

* Interval: 30 minutes between application and exercise.

Experiment 3: Relationship between Time of Application and Sun Exposure in Conjunction with Swimming

Preparations to be Evaluated: Formula G, Skolex, MAP (D5) and Modified G

Object:

1) To determine whether there exists an optimum application-exposure interval as outlined in Experiment 1, under conditions of 15 minutes swimming.

2) After one application, compare the efficacy of preparations G, Skolex, MAP (D5) and MG under conditions of 15 minutes swimming in fresh water.

3) To determine whether preparation G, the formulation that contains 2.5% glyceryl para-aminobenzoate in 65-70% alcohol, is as effective as MAP (D5) under conditions of swimming.

Number of Subjects: 8

Plan of Experiment:

a) Activity: 15 minutes swimming in a tank before exposure.

b) Taping: Masking tape was applied to the subjects' backs to prevent mixing of test preparations.

c) Application of test compounds: The test preparations were applied to the appropriate spaces as indicated below:

Subject's Back

Row	Left Side		Right Side	
1	CON	G	CON	G
2	CON	SX	CON	SX
3	D5	CON	D5	CON
Sun Exposure:	← 45 Min →		← 90 Min →	

CON = control area — Subjects 1 to 8
G = Formula G — " 1 to 6
SX = Skolex — " 1 to 8
D5 = Preparation MAP — " 1 to 8
MG = Modified G — " 7 and 8

MED: Minimal erythema dose was determined for most of the subjects by exposing the lower portion of the back for 10, 15, 20 & 30 minutes solar radiation.

Expt. 3 (continued)

d) Subjects # 1 to 3: Between 0 to 15 minutes after application of preparations G, Skolex and MAP (D5), the subjects went swimming for 15 minutes. After swimming they were exposed to solar radiation for 45 and 90 minutes.

Subjects # 4 to 6: Thirty minutes after application of preparations G, Skolex and MAP (D5), the subjects went swimming for 15 minutes. After swimming they were exposed to solar radiation for 45 and 90 minutes.

Subjects # 7 and 8: Thirty minutes after application of preparations Skolex, MAP (D5) and Modified G, the subjects went swimming for 15 minutes and then were exposed to sun for 45 and 90 minutes.

e) Evaluation of the Results: Exposed areas were evaluated at 24 and 48 hours after sun exposure for the degree of erythema and tanning by visible gradation of redness and by skin reflectance measurements.

Results: The following conclusions can be drawn from the data obtained on 8 subjects after 45 and 90 minutes sun exposure.

1) There is no minimum waiting period required for the applied preparation to react with the skin to exert its maximum sun-protective effect. The protective effect was practically identical in 0 to 15 minutes and 30 minutes interval groups.

2) Based on the earlier studies, it appears that G, Skolex, MAP (D5) and MG are very effective sunscreen preparations. They are not easily removed and eluted from the skin after swimming. They remain either adsorbed or chemically bonded to the horny layer and exert very effective protection up to 90 minutes of sun exposure.

3) When compared against each other, preparations G, and MG were found to be better than Skolox and MAP (D5).

Experiment # 3: Relationship between Time of Application and Sun Exposure* in Conjunction with 15 Minutes Swimming

Subject & Prison #	Site (Back)	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Sherer	Row 1	Control	45	+	0	12	0	
27866	"	G	45	±	50	6	50	
(1)	"	Control	90	+++	0	18	0	
	"	G	90	±	75	5	72	
	Row 2	Control	45	++	0	12	0	
	"	Skolex	45	±	50	9	25	
	"	Control	90	+++	0	21	0	
	"	Skolex	90	±	75	6	70	
	Row 3	Control	45	++	0	16	0	
	"	MAP (D5)	45	±	66	8	50	
	"	Control	90	+++	0	18	0	
	"	MAP (D5)	90	++	25	13	30	

* Interval: 0-15 minutes between application and swimming.

Experiment # 3: Relationship between Time of Application and Sun Exposure* in Conjunction with 15 Minutes Swimming

Subject & Prison #	Site (Back)	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Terrel	Row 1	Control	45	±	0	5	0	
27884	"	G	45	0	100	2	100	IPD
(2)	"	Control	90	+	0	8	0	
	"	G	90	0	100	3	100	IPD
	Row 2	Control	45	±	0	6	0	
	"	Skolex	45	±	0	5	0	IPD
	"	Control	90	++	0	14	0	
	"	Skolex	90	±	66	6	57	IPD
	Row 3	Control	45	+	0	7	0	
	"	MAP (D5)	45	0	100	4	100	IPD
	"	Control	90	++	0	10	0	
	"	MAP (D5)	90	+	50	4	60	IPD

* Interval: 0-15 minutes between application and swimming.

Experiment # 3: Relationship between Time of Application and Sun Exposure* in Conjunction with 15 Minutes Swimming

Subject & Prison #	Site (Back)	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Gilmore	Row 1	Control	45	0	100	5	100	
28065	"	G	45	0	100	3	100	
(3)	"	Control	90	+	0	7	0	
	"	G	90	0	100	5	100	IPD
	Row 2	Control	45	0	100	5	100	
	"	Skolex	45	0	100	3	100	
	"	Control	90	++	0	12	0	
	"	Skolex	90	±	66	5	58	
	Row 3	Control	45	±	0	6	0	
	"	MAP (D5)	45	0	100	0	100	
	"	Control	90	++	0	12	0	
	"	MAP (D5)	90	+	33	9	25	

* Interval: 0-15 minutes between application and swimming.

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Experiment # 3: Relationship between Time of Application and Sun Exposure* in Conjunction with 15 Minutes Swimming

Subject & Prison #	Site (Back)	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Dero	Row 1	Control	45	0	100	6	100	IPD
27418	"	G	45	0	100	6	100	IPD
(4)	"	Control	90	++	0	8	0	
	"	G	90	0	100	1	100	IPD
	Row 2	Control	45	±	0	8	0	
	"	Skolex	45	0	100	2	100	IPD
	"	Control	90	++	0	17	0	
	"	Skolex	90	0	100	8	51	
	Row 3	Control	45	±	0	4	0	
	"	MAP (D5)	45	0	100	0	100	IPD
	"	Control	90	+	0	11	0	
	"	MAP (D5)	90	±	50	7	40	

* Interval: 30 minutes between application and swimming.

Experiment # 3: Relationship between Time of Application and Sun Exposure* in Conjunction with 15 Minutes Swimming

Subject & Prison #	Site (Back)	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remark
Marcrum	Row 1	Control	45	+	0	4	0	
28130	"	G	45	±	50	2	50	
(5)	"	Control	90	+++	0	14	0	
	"	G	90	±	75	5	65	
	Row 2	Control	45	+	0	6	0	
	"	Skolex	45	0	100	0	100	
	"	Control	90	+++	0	17	0	
	"	Skolex	90	0	100	7	59	
	Row 3	Control	45	+	0	9	0	
	"	MAP (D5)	45	±	50	5	45	
	"	Control	90	+++	0	17	0	
	"	MAP (D5)	90	+	50	8	53	

* Interval: 30 minutes between application and swimming.

Experiment # 3: Relationship between Time of Application and Sun Exposure* in Conjunction with 15 Minutes Swimming

Subject & Prison #	Site (Back)	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Johnson	Row 1	Control	45	++	0	8	0	
27727	"	G	45	±	50	2	75	
(6)	"	Control	90	+++	0	17	0	
	"	G	90	+	50	10	40	
	Row 2	Control	45	++	0	17	0	
	"	Skolex	45	±	66	5	70	
	"	Control	90	+++	0	20	0	
	"	Skolex	90	±	75	7	65	
	Row 3	Control	45	++	0	14	0	
	"	MAP (D5)	45	±	50	5	65	
	"	Control	90	++	0	17	0	
	"	MAP (D5)	90	++	0	13	25	

* Interval: 30 minutes between application and swimming.

Experiment # 3: Relationship between Time of Application and Sun Exposure* in Conjunction with Swimming**

Subject & Prison #	Site (Back)	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Barton	Row 1	Control	45	++	0	10	0	
24846	"	Modified G	45	0	100	3	70	
(7)	"	Control	90	+++	0	14	0	
	"	Modified G	90	0	100	3	80	
	Row 2	Control	45	+++	0	15	0	
	"	Skolex	45	±	75	10	33	
	"	Control	90	+++	0	20	0	
	"	Skolex	90	+	50	8	60	
	Row 3	Control	45	++	0	15	0	
	"	MAP (D5)	45	±	66	5	66	
	"	Control	90	+++	0	17	0	
	"	MAP (D5)	90	++	25	14	20	

* Interval: 30 minutes between application and swimming. ** 15 minutes swimming.

Experiment # 3: Relationship between Time of Application and Sun Exposure* in Conjunction with 15 Minutes Swimming

Subject & Prison #	Site (Back)	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Frand	Row 1	Control	45	+	0	6	0	IPD
28046	"	Modified G	45	0	100	0	100	
(8)	"	Control	90	++	0	11	0	
	"	Modified G	90	0	100	5	100	IPD
	Row 2	Control	45	+	0	9	0	
	"	Skolex	45	±	50	5	45	
	"	Control	90	+++	0	20	0	
	"	Skolex	90	±	75	7	65	
	Row 3	Control	45	++	0	7	0	
	"	MAP (D5)	45	±	66	3	57	
	"	Control	90	+++	0	16	0	
	"	MAP (D5)	90	+	50	7	56	

* Interval: 30 minutes between application and swimming.

Experiment 4: Duration of the Protective Effect: To determine whether a single application of test compounds has any prolonged effect.

Preparations to be Evaluated: Formula G, Skolex and MAP (D5)

Object: After one application of these test preparations at 24 hours prior to sun exposure, determine whether this single application of test compounds has any prolonged protective effect when the subjects are exposed to solar radiation for 60 and 120 minutes on the following day. For comparison, evaluate the same test preparations after a single application at 30 minutes prior to sun exposure.

Number of Subjects: 6

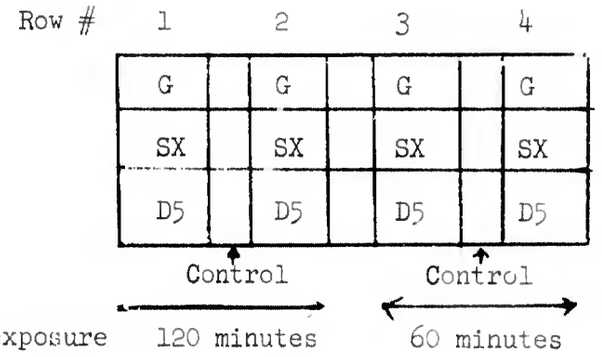
Plan of Experiment:

a) Activity: None, no exercise or swimming.

b) Taping: Masking tapes were applied to the subjects' backs to prevent mixing of test compounds.

c) Application of test compounds: The test preparations were applied to the appropriate spaces as indicated below:

Subject's Back



Control=	No preparation	Subjects 1 to 6
G	Formula G	" 1 to 6
SX	Skolex	" 1 to 6
D5	MAP (D5)	" 1 to 6

d) Subject # 1 to 6: Day 1: 24 hours prior to exposure, test compounds were applied to rows # "2" and "3" only.
Day 2: 30 minutes prior to exposure, test compounds were applied to rows # "1" and "4".
Day 2: Rows # 1 and 2 were exposed to sun for 120 minutes, rows # 3 and 4 were exposed for 60 minutes. During

Expt. 4 (continued)

the first hour of exposure, rows # 1 and 2 only were exposed, while rows # 3 and 4 were kept covered with light-proof papers. In the second hour of exposure, all the rows are exposed to sun.

e) Evaluation of the results: Exposed areas were evaluated at 24 and 48 hours after exposure for the degree of redness and tanning (immediate pigment darkening reaction) by visible gradation of redness and by skin reflectance measurements.

Results: The following conclusions can be drawn from the data obtained on 6 subjects after 60 and 120 minutes sun exposure.

1) Skin areas that were treated with preparations G and Skolex 24 hours prior to exposure showed the same degree of erythema after 60 and 120 minutes sun exposure as that observed in the untreated control areas. All the six subjects showed no evidence of any protection. It was evident that a single application of preparations G and Skolex 24 hours prior to sun exposure did not remain on the surface of the skin and did not provide any protection on the following day. Since there was no protection, it is apparent that these preparations are removed from the surface of the skin during the normal daily activities of the subjects.

2) Contrary to the above observations, a single application of MAP (D5) 24 hours prior to exposure, showed an excellent protection against 60 and 120 minutes of solar radiation. This preparation is not washed off from the surface of the skin and appears to remain adsorbed and chemically bonded to the horny layer.

3) Skin areas that were treated with preparations G, Skolex and MAP (D5) 30 minutes prior to sun exposure showed excellent protection against 60 and 120 minutes solar radiation.

4) When compared against each other, preparations G and MAP (D5) were found to be very effective in protecting against 60 and 120 minutes of solar radiation when applied 30 minutes prior to sun exposure. Skolex was, however, found to be less effective than G and MAP (D5).

Experiment # 4: Duration of Sun Protective Effect: To determine whether a single application of MAP (D5), G, and Skolex has any prolonged protective effect under conditions of no exercise and no swimming.

Subject & Prison #	Site	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Madden	Row 2	Control*	120	+	0	8	0	Sunscreens applied 24 hours prior to sun exposure.
28236	"	G	"	+	0	7	0	
(1)	"	Skolex	"	+	0	9	0	
	"	MAP (D5)	"	±	50	4	50	
	Row 1	Control*	120	+	0	8	0	Sunscreens applied 30 minutes prior to sun exposure.
	"	G	"	0	100	5 IPD	100	
	"	Skolex	"	±	50	5	40	
	"	MAP (D5)	"	0	100	3 IPD	100	
	Row 3	Control*	60	0	100	3 IPD	100	Sunscreens applied 24 hours prior to sun exposure.
	"	G	"	0	100	4 IPD	100	
	"	Skolex	"	0	100	5 IPD	100	
	"	MAP (D5)	"	0	100	3 IPD	100	
	Row 4	Control*	60	0	100	3 IPD	100	Sunscreens applied 30 minutes prior to sun exposure.
	"	G	"	0	100	1	100	
	"	Skolex	"	0	100	3 IPD	100	
	"	MAP (D5)	"	0	100	0	100	

* Average of 4 sites

Experiment # 4: Duration of Sun Protective Effect: To determine whether a single application of MAP (D5), G, and Skolex has any prolonged protective effect under conditions of no exercise and no swimming.

Subject & Prison #	Site	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Thomson	Row 2	Control*	120	+++	0	20	0	Sunscreens applied 24 hours prior to sun exposure.
27571	"	G	"	+++	0	19	0	
(2)	"	Skolex	"	+++	0	17	15	
	"	MAP (D5)	"	±	75	8	60	
	Row 1	Control*	120	+++	0	20	0	Sunscreens applied 30 minutes prior to sun exposure.
	"	G	"	±	75	5	75	
	"	Skolex	"	+	50	10	50	
	"	MAP (D5)	"	0	100	5	75	
	Row 3	Control*	60	++	0	15	0	Sunscreens applied 24 hours prior to sun exposure.
	"	G	"	+	33	16	0	
	"	Skolex	"	++	0	15	0	
	"	MAP (D5)	"	±	66	5	66	
	Row 4	Control*	60	++	0	15	0	Sunscreens applied 30 minutes prior to sun exposure.
	"	G	"	±	66	7	54	
	"	Skolex	"	+	33	11	27	
	"	MAP (D5)	"	0	100	2	87	

* Average of 3 sites.

Experiment # 4: Duration of Sun Protective Effect: To determine whether a single application of MAP (D5), G, and Skolex has any prolonged protective effect under conditions of no exercise and no swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference		% Protection	Remarks
Brody	Row 2	Control*	120	++	0	14		0	Sunscreens applied 24 hours prior to sun exposure.
27892	"	G	"	+	33	11		20	
(3)	"	Skolex	"	++	0	12		15	
	"	MAP (D5)	"	±	66	6		67	
	Row 1	Control*	120	++	0	14		0	Sunscreens applied 30 minutes prior to sun exposure.
	"	G	"	0	100	5	IPD	100	
	"	Skolex	"	0	100	5	IPD	100	
	"	MAP (D5)	"	0	100	4	IPD	100	
	Row 3	Control*	60	±	0	4		0	Sunscreens applied 24 hours prior to sun exposure.
	"	G	"	±	0	4		0	
	"	Skolex	"	+	0	7		0	
	"	MAP (D5)	"	0	100	0		100	
	Row 4	Control*	60	±	0	4		0	Sunscreens applied 30 minutes prior to sun exposure.
	"	G	"	0	100	0		100	
	"	Skolex	"	0	100	0		100	
	"	MAP (D5)	"	0	100	0		100	

* Average of 3 sites

Experiment # 4: Duration of Sun Protective Effect: To determine whether a single application of MAP (D5), G, and Skolex has any prolonged protective effect under conditions of no exercise and no swimming.

Subject & Prison #	Site	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	Degree of Erythema after 24 Hours: % Protection	% Reflectance Difference	% Protection	Remarks
Flood	Row 2	Control*	120	+++	0	18	0	Sunscreens applied 24 hours prior to sun exposure.
28260	"	G	"	+++	0	18	0	
(4)	"	Skolex	"	+++	0	17	0	
	"	MAP (D5)	"	+	50	11	40	
	Row 1	Control*	120	+++	0	18	0	Sunscreens applied 30 minutes prior to sun exposure.
	"	G	"	±	75	9	50	
	"	Skolex	"	+	50	9	50	
	"	MAP (D5)	"	0	100	6 IPD	100	
	Row 3	Control*	60	+	0	12	0	Sunscreens applied 24 hours prior to sun exposure.
	"	G	"	+	0	12	0	
	"	Skolex	"	+	0	11	0	
	"	MAP (D5)	"	±	50	6	50	
	Row 4	Control*	60	+	0	12	0	Sunscreens applied 30 minutes prior to sun exposure.
	"	G	"	0	100	9 IPD	100	
	"	Skolex	"	±	50	7	42	
	"	MAP (D5)	"	0	100	0	100	

* Average of 3 sites.

Experiment # 4: Duration of Sun Protective Effect: To determine whether a single application of MAP (D5), G, and Skolex has any prolonged protective effect under conditions of no exercise and no swimming.

Subject & Prison #	Site	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Hallum	Row 2	Control*	120	+	0	12	0	Sunscreens applied 24 hours prior to sun exposure.
26184	"	G	"	+	0	11	0	
(5)	"	Skolex	"	±	50	7	42	
	"	MAP (D5)	"	0	100	3 IPD	100	
	Row 1	Control*	120	+	0	12	0	Sunscreens applied 30 minutes prior to sun exposure.
	"	G	"	0	100	4 IPD	100	
	"	Skolex	"	±	50	8	33	
	"	MAP (D5)	"	0	100	3 IPD	100	
	Row 3	Control*	60	0	100	5 IPD	100	Sunscreens applied 24 hours prior to sun exposure.
	"	G	"	0	100	8 IPD	100	
	"	Skolex	"	0	100	5 IPD	100	
	"	MAP (D5)	"	0	100	4 IPD	100	
	Row 4	Control*	60	0	100	8 IPD	100	Sunscreens applied 30 minutes prior to sun exposure.
	"	G	"	0	100	8 IPD	100	
	"	Skolex	"	0	100	4	50	
	"	MAP (D5)	"	0	100	0	100	

* Average of 4 sites.

Experiment # 4: Duration of Sun Protective Effect: To determine whether a single application of MAP (D5), G, and Skolex has any prolonged protective effect under conditions of no exercise and no swimming.

Degree of Erythema after 24 Hours

Subject & Prison #	Site	Preparation	Period of Sun Exposure (minutes)	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Livingwood	Row 2	Control*	120	++	0	15	0	Sunscreens applied 24 hours prior to sun exposure.
26806	"	G	"	±	66	3	80	
(6)	"	Skolex	"	++	0	18	0	
	"	MAP (D5)	"	0	100	4 IPD	100	
	Row 1	Control*	120	++	0	15	0	Sunscreens applied 30 minutes prior to sun exposure.
	"	G	"	0	100	3	100	
	"	Skolex	"	+	33	14	0	
	"	MAP (D5)	"	0	100	2 IPD	100	
	Row 3	Control*	60	+	0	6	0	Sunscreens applied 24 hours prior to sun exposure.
	"	G	"	±	50	4	33	
	"	Skolex	"	+	0	9	0	
	"	MAP (D5)	"	0	100	0	100	
	Row 4	Control*	60	+	0	6	0	Sunscreens applied 30 minutes prior to sun exposure.
	"	G	"	0	100	1	100	
	"	Skolex	"	±	50	4	33	
	"	MAP (D5)	"	0	100	0	100	

* Average of 4 sites.

Experiment 5: Cumulative Effect: To determine whether protection against solar radiation is additive with repeated applications under conditions of no exercise and no swimming.

Object: After two applications of preparations G, Skolex and MAP (D5) at 24 hours and 1 hour prior to sun exposure, determine whether this repeated application affords a better protection than a single application just 30 minutes prior to exposure.

Preparations to be Evaluated: Formula G, Skolex and MAP (D5)

Number of Subjects: 6

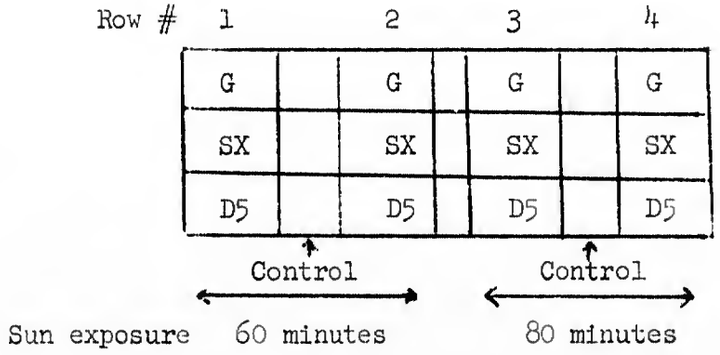
Plan of Experiment:

a) Activity: None

b) Taping: Masking tapes were applied to the subjects' backs to prevent mixing of the test preparations.

c) Application of the test compounds: The test preparations were applied to the appropriate spaces as indicated below:

Subject's Back



d) Day 1, subject # 1 to 3: 24 hours prior to sun exposure, test compounds were applied to rows # 2 and 3.
Day 2, subject # 1 to 6: 1 hour prior to sun exposure, test compounds were applied to rows # 2 and 3.
Day 2, subject # 1 to 6: 30 minutes prior to sun exposure, test compounds were applied to rows # 1 and 4, and also to rows # 2 and 3 of subjects # 4 to 6.

Thus rows # 2 and 3 received 2 applications of test compounds and rows # 1 and 4 received only one application.

Expt. 5 (continued)

e) Exposure period: Rows # 1 and 2 and the control areas received 60 minutes sun exposure. Rows # 3 and 4, plus the control areas received 80 minutes of sun exposure.

Results: The erythemal response observed in the control areas and the test areas was compared in the following manner:

a) 2 applications v/s one application of the test compounds.

b) The degree of protection afforded by each compound was obtained by comparing the response in the control area adjacent to the test area.

The following conclusions can be drawn from the data obtained on 6 subjects after 60 and 80 minutes sun exposure.

1) After two applications of preparations G, Skolex and MAP (D5) at 24 hours and 1 hour prior to sun exposure, the subjects did not show any added protection when compared against areas receiving a single application of the test preparations. In fact, areas that received 2 applications of test preparations showed slightly less protection than areas receiving a single application 30 minutes prior to sun exposure. This effect was particularly evident in areas treated with G and Skolex. The average % protection after 2 applications of G and Skolex for 60 minutes exposure period was: 84 and 33 respectively, whereas after a single application the % protection for 60 minute period was 93 and 70 respectively. Likewise, for 80 minutes sun exposure, the average % protection afforded by G and Skolex after two applications was 58 and 37 respectively and after a single application, the average % protection by G and Skolex appears to be 83 and 68.

2) MAP (D5) was found to be most effective in this experiment. All subjects showed excellent protection.

Experiment # 5: Cumulative Effect: To determine whether protection against sun is additive with repeated applications under conditions of no exercise and no swimming

Degree of Erythema after 24 Hours

Subject & Prison #	Site	Preparation	Period of Sun Exposure (Minutes)	Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Kenny	Row 2	Control*	60	+	0	13	0	Compounds applied 2 times 24 hrs and 1 hr before sun exposure.
28277	"	G	"	0	100	0	100	
(1)	"	Skolex	"	±	50	5	60	
	"	MAP (D5)	"	0	100	0	100	
	Row 1	Control*	60	+	0	13	0	Compounds applied once ½ hr before sun exposure.
	"	G	"	0	100	0	100	
	"	Skolex	"	±	50	4	70	
	"	MAP (D5)	"	0	100	0	100	
	Row 3	Control*	80	+	0	12	0	Compounds applied 2 times: 24 hrs and 1 hr before sun exposure.
	"	G	"	0	100	2	100	
	"	Skolex	"	±	50	5	59	
	"	MAP (D5)	"	0	100	0	100	
	Row 4	Control*	80	+	0	12	0	Compounds applied once ½ hr before sun exposure.
	"	G	"	0	100	3	75	
	"	Skolex	"	0	100	1	100	
	"	MAP (D5)	"	0	100	0	100	

* Average of 4 sites.

Experiment # 5: Cumulative Effect: To determine whether protection against sun is additive with repeated applications under conditions of no exercise and no swimming

Subject Prison #	Site	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Pottorff	Row 2	Control*	60	+	0	10	0	Compounds applied 2 times 24 hrs and 1 hr before sun exposure.
27356	"	G	"	0	100	0	100	
(2)	"	Skolex	"	±	50	6	40	
	"	MAP (D5)	"	0	100	0	100	
	Row 1	Control*	60	+	0	10	0	Compounds applied once ½ hr before sun exposure.
	"	G	"	0	100	4	60	
	"	Skolex	"	±	50	5	50	
	"	MAP (D5)	"	0	100	1	100	
	Row 3	Control*	80	+	0	12	0	Compounds applied 2 times 24 hrs and 1 hr before sun exposure.
	"	G	"	0	100	7	48	
	"	Skolex	"	±	50	5	52	
	"	MAP (D5)	"	0	100	1	100	
	Row 4	Control*	80	+	0	12	0	Compounds applied once ½ hr before sun exposure.
	"	G	"	0	100	6	50	
	"	Skolex	"	±	50	4	66	
	"	MAP (D5)	"	0	100	2	84	

* Average of 4 sites.

Experiment # 5: Cumulative Effect: To determine whether protection against sun is additive with repeated applications under conditions of no exercise and no swimming.

Subject & Prison #	Site	Preparation	Period of Sun Exposure (minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Wilkerson	Row 2	Control*	60	+	0	7	0	Compounds applied two times: 24 hrs and 1 hr before sun exposure.
27611	"	G	"	0	100	3 IPD	100	
(3)	"	Skolex	"	±	50	6	0	
	"	MAP (D5)	"	0	100	0	100	
	Row 1	Control*	60	+	0	7	0	Compounds applied once, 1/2 hr before sun exposure.
	"	G	"	0	100	3 IPD	100	
	"	Skolex	"	0	100	2 IPD	100	
	"	MAP (D5)	"	0	100	0	100	
	Row 3	Control*	80	+	0	7	0	Compounds applied two times: 24 hrs and 1 hr before sun exposure.
	"	G	"	±	50	7	0	
	"	Skolex	"	±	50	7	0	
	"	MAP (D5)	"	0	100	2	70	
	Row 4	Control*	"	+	0	7	0	Compounds applied once, 1/2 hr before sun exposure.
	"	G	"	0	100	3 IPD	100	
	"	Skolex	"	±	50	5	30	
	"	MAP (D5)	"	0	100	2	100	

* Average of 4 sites

Experiment # 5: Cumulative Effect: To determine whether protection against sun is additive with repeated applications under conditions of no exercise and no swimming.

Subject & Prison #	Site	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Martin	Row 2	Control*	60	±	0	5	0	Compounds applied twice: 1 hour before sun exposure.
23855	"	G	"	0	100	0	100	
(4)	"	Skolex	"	±	0	5	0	
	"	MAP (D5)	"	0	100	2 IPD	100	
	Row 1	Control*	60	±	0	7	0	Compounds applied once, ½ hour before sun exposure.
	"	G	"	0	100	0	100	
	"	Skolex	"	±	0	4	43	
	"	MAP (D5)	"	0	100	0	100	
	Row 3	Control*	80	±	0	7	0	Compounds applied twice: 1 hour before Sun exposure.
	"	G	"	0	100	1	100	
	"	Skolex	"	±	0	5	30	
	"	MAP (D5)	"	0	100	0	100	
	Row 4	Control*	80	±	0	7	0	Compounds applied once, ½ hour before sun exposure.
	"	G	"	0	100	0	100	
	"	Skolex	"	0	100	3	100	
	"	MAP (D5)	"	0	100	0	100	

* Average of 4 sites.

Experiment # 5: Cumulative Effect: To determine whether protection against sun is additive with repeated applications under conditions of no exercise and no swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Pfeifer	Row 2	Control*	60	+	0	10	0	Compounds applied 2 times: 1 hour before sun exposure.
28071	"	G	"	0	100	4 IPD	100	
(5)	"	Skolex	"	±	50	6	50	
	"	MAP (D5)	"	0	100	3 IPD	100	
	Row 1	Control*	60	+	0	10	0	Compounds applied once, ½ hour before sun exposure.
	"	G	"	0	100	1	100	
	"	Skolex	"	±	50	4	60	
	"	MAP (D5)	"	0	100	1	100	
	Row 3	Control*	80	+	0	12	0	Compounds applied 2 times: 1 hour before sun exposure.
	"	G	"	0	100	3	100	
	"	Skolex	"	±	50	6	50	
	"	MAP (D5)	"	0	100	1	100	
	Row 4	Control*	80	+	0	12	0	Compounds applied once, ½ hour before sun exposure.
	"	G	"	+	100	3	75	
	"	Skolex	"	±	50	6	50	
	"	MAP (D5)	"	0	100	1	100	

* Average of 4 sites.

Experiment # 5: Cumulative Effect: To determine whether protection against sun is additive with repeated applications under conditions of no exercise and no swimming

Subject & Prison #	Site	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Renz	Row 2	Control*	60	±	0	4	0	Compounds applied 2 times: 1 hour before sun exposure.
27894	"	G	"	±	0	6	0	
(6)	"	Skolex	"	±	0	2	50	
	"	MAP (D5)	"	0	100	0	100	
	Row 1	Control*	60	±	0	5	0	Compounds applied once, ½ hour before sun exposure.
	"	G	"	0	100	0	100	
	"	Skolex	"	0	100	2	100	
	"	MAP (D5)	"	0	100	1	100	
	Row 3	Control*	80	+	0	7	0	Compounds applied 1 hour before sun exposure.
	"	G	"	±	50	7	0	
	"	Skolex	"	±	50	5	30	
	"	MAP (D5)	"	0	100	0	100	
	Row 4	Control*	80	+	0	7	0	Compounds applied ½ hour before sun exposure.
	"	G	"	0	100	1	100	
	"	Skolex	"	0	100	0	100	
	"	MAP (D5)	"	0	100	0	100	

* Average of 3 sites.

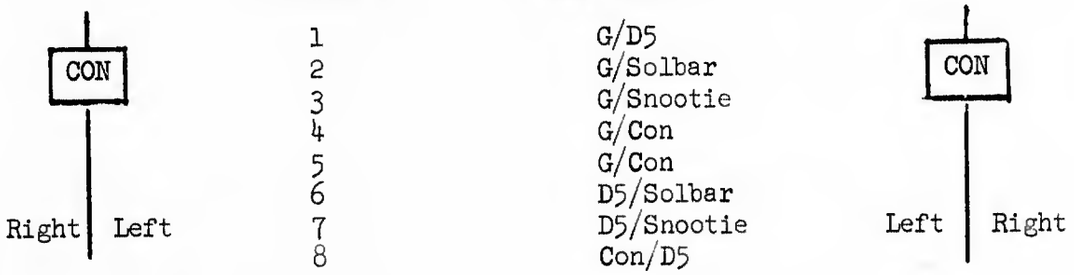
Experiment 6: To determine the relative efficacy of various sunscreens under conditions simulating normal usage*

Preparations to be Evaluated: MAP (D5), G, Solbar and Snootie

Object: After one application of MAP (D5), G, Solbar and Snootie on the front and back sides of subjects, evaluate the relative protective qualities of these preparations under the conditions simulating normal usage.

Number of Subjects: 8

Plan of Experiment: Masking tapes were applied in the center of front (chest and abdomen) and back of the volunteer to insure proper application of compounds on the right and left side of the body. On five subjects, as shown below, one compound was applied on one half of the body including head, neck and extremities (front and back), and the other compound was applied on the other half (front and back). An untreated control area (approximately 4" x 5") was left on anterior and posterior surfaces. On the other 3 subjects, preparation G or MAP (D5) was applied only on the half side (front and back) and compared against control area (untreated but exposed).



Subject #	Right/Left
1	G/D5
2	G/Solbar
3	G/Snootie
4	G/Con
5	G/Con
6	D5/Solbar
7	D5/Snootie
8	Con/D5

All together, preparation G was applied on 5 subjects, preparation D5 on 4 subjects and preparations Solbar and Snootie were applied on 4 subjects (2 each).

After one application of test compounds, the following program of physical activity was followed:

9:00 - 10:00 a.m.	Application of test compounds
10:00 - 10:30 a.m.	Drying of the applied compounds
10:35 - 10:45 a.m.	10 minutes swimming
10:45 - 11:00 a.m.	Incidental drying in the sun
11:00 - 11:20 a.m.	20 minutes, expose anterior surface from horizontal position.

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* **Normal conditions of use include those things that one would prefer to do on a beach on a sunny day, eg., swimming, lying on the back and in the prone position, walking and running around the court, playing volleyball, etc.**

Expt. 6 (continued)

11:20 - 11:40 a.m.	20 minutes, expose posterior surface from horizontal position.
11:40 - 12:05 p.m.	25 minutes, walk around in the sun
12:05 - 12:15 p.m.	10 minutes, run around in the sun
12:15 - 12:25 p.m.	10 minutes, swimming
12:25 - 12:45 p.m.	20 minutes, expose anterior surface from horizontal position.
12:45 - 1:05 p.m.	20 minutes, expose posterior surface from horizontal position.

The exposed areas were evaluated at 24 and 48 hours after sun exposure for erythema and tanning.

Results: Under conditions simulating normal usage, preparation G was found to be most effective in affording satisfactory protection. The subjects did not exhibit any severe sunburn. They exhibited barely perceptible to a moderate sunburn response even after a rigorous schedule of exercise and swimming. Preparation MAP (D5) was less effective than G, but still afforded satisfactory protection. It was definitely better than Solbar and Snootie. These latter two preparations were easily washed away from the surface of the skin. In fact, Solbar was found to be least effective and appeared to have been washed away after swimming and sweating. The reason why MAP (D5) appeared to be less effective than preparation G is related to the presence of emolients used in preparing the D5 preparation. These emolients lower the affinity of PABA to bind with the horny layer. The plain MAP preparation without any emolients certainly affords protection up to 2 hours of high intensity mid-day solar radiation even after sweating and swimming (under similar conditions as described above). In our experience, all the leading commercial preparations, after a single application, are washed away and rendered ineffective after subjecting the individuals to sweating or swimming.

Conclusions: In this experiment an attempt has been made to evaluate the protective effect of G, MAP (D5), Snootie and Solbar under conditions simulating normal usage. These included at least two swimming periods lasting 10 minutes each and two exposure periods, lasting for at least 90 minutes (40 minutes exposure on each side under stationary conditions and 50 minutes exposure under walking conditions). Thus, under these rigorous experimental conditions, preparations G, and MAP (D5) provided satisfactory protection. Under similar conditions, the commercially available products Solbar and Snootie, were found to be unsatisfactory in affording protection.

Experiment # 6: To Determine the Relative Efficacy of MAP (D5) and G under Conditions Simulating Normal Usage

Subject & Prison #	Site	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Cakes	Front	Control (center)	90	+	0	8	0	
28143	"	G (right)	"	±	50	6	25	
(1)	"	MAP (D5), (left)	"	±	50	6	25	
	Back	Control (center)	"	++	0	12	0	
	"	G (right)	"	+	33	5	58	
	"	MAP (D5), (left)	"	+	33	7	40	
Baker	Front	Control (center)	90	+	0	7	0	
27553	"	G (right)	"	±	50	5	30	
(2)	"	Solbar (left)	"	+	0	7	0	
	Back	Control (center)	"	++	0	10	0	
	"	G (right)	"	±	66	6	40	
	"	Solbar (left)	"	++	0	11	0	

Experiment # 6: To Determine the Relative Efficacy of MAP (D5) and G under Conditions Simulating Normal Usage

Subject & Prison #	Site	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Mann	Front	Control (center)	90	±	0	6	0	
25792	"	G (right)	"	0	100	0	100	
(3)	"	Snootie (left)	"	±	0	4	33	
	Back	Control (center)	"	±	0	6	0	
	"	G (right)	"	0	100	0	100	
	"	Snootie (left)	"	±	0	4	33	
Aswed	Front	Control	90	+	0	11	0	
26707	"	G (right)	"	0	100	0	100	
(4)	Back	Control	"	+	0	5	0	
	"	G (right)	"	0	100	3 IPD	100	

Experiment # 6: To Determine the Relative Efficacy of MAP (D5) and G under Conditions Simulating Normal Usage

Subject & Prison #	Site	Preparation	Period of Sun Exposure (minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Laird	Front	Control	90	+++	0	18	0	
26157	"	G (right)	"	±	75	5	70	
(5)	Back	Control	"	+++	0	18	0	
	"	G (right)	"	+	50	13	28	
Fancher	Front	Control (center)	90	+	0	8	0	
28008	"	MAP (D5), (right)	"	±	50	5	40	
(6)	"	Solbar (left)	"	+	0	8	0	
	Back	Control (center)	"	±	0	8	0	
	"	MAP (D5), (right)	"	0	100	5	40	
	"	Solbar (left)	"	±	0	8	0	

Experiment # 6: To Determine the Relative Efficacy of MAP (D5) and G under Conditions Simulating Normal Usage

Subject & Prison #	Site	Preparation	Period of Sun Exposure (minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Stout	Front	Control (center)	90	++	0	11	0	
27766	"	MAP (D5), (right)	"	++	0	14	0	
(7)	"	Snootie (left)	"	±	66	5	55	
	Back	Control (center)	"	++	0	15	0	
	"	MAP (D5), (Right)	"	++	0	13	15	
	"	Snootie (left)	"	++	0	16	0	
McKenrick	Front	Control	90	+	0	6	0	
28225	"	MAP (D5), (left)	"	±	50	3	50	
(8)	Back	Control	"	+	0	7	0	
	"	MAP (D5), (left)	"	0	100	1	100	

Experiment 7: To Evaluate the Effect of MAP (D5) and G Preparations on 4,5',8-Trimethylpsoralen (TMP) Induced Photosensitization under Normal Conditions of Use (Sunbathing Accompanied by Sweating & Swimming)

Object: This experiment was designed to test the relative protective efficacy of preparations G and MAP (D5), against TMP induced photosensitization. When TMP is administered orally to human subjects, and their skin is exposed to solar radiation, augmented sunburn response is observed. Commercially available popular sunscreens are easily washed away under conditions of exercise accompanied by sweating or swimming and afford very poor protection against sunburn. Since preparations G and MAP (D5) afford very satisfactory protection even after a rigorous schedule of exercise accompanied by sweating or after swimming, it was essential to know whether these preparations can inhibit or block the TMP induced photosensitization, and promote tanning without evoking the exaggerated sunburn response.

ns to be Evaluated: G, MG, and MAP (D5). Preparation MG contained approximately 5% of glyceryl para-aminobenzoate and preparation G contained 2.5% of glyceryl para-aminobenzoate.

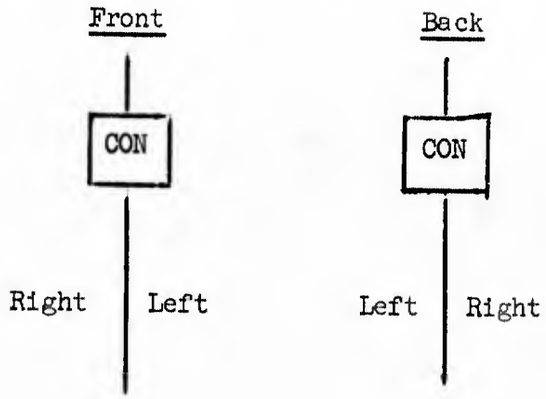
Number of Subjects: Total 4

Dose of TMP: 40 mg of TMP per subject, administered orally 2 hours before exposure to sun.

Plan of Experiment:

a) Activity: 10 minutes swimming after application of test preparations. 20 minutes walk around the court and 10 minutes running to evoke sweating.

b) Taping: Masking tapes were applied to the subjects' chests and backs as shown below:



Expt. 7 (continued)

c) Preparations G, MG and MAP (D5) were applied to the subjects' front side (face, neck, shoulder, chest and abdomen) and back side (neck, shoulder, upper and lower regions of back) as shown below:

Group	Subject #	Left Front & Back	Right Front & Back
7	1	G	MAP (D5)
	2	G	MAP (D5)
	3	Modified G	MAP (D5)
	4	MAP (D5)	Modified G

An untreated control area approximately 5" x 5" was left on the anterior and posterior surfaces.

d) Schedule: The schedule for exercise, walking and swimming for each subject of Group 7 was as follows:

9:00 a.m.	40 mg TMP orally
10:00 - 11:00 a.m.	Application of the test preparations
11:00 - 11:10	10 minutes swimming
11:10 - 11:25	15 minutes incidental drying
11:25 - 11:45	20 minutes, expose anterior surface from horizontal position
11:45 - 12:05 p.m.	20 minutes, expose posterior surface from horizontal position
12:05 - 12:20	15 minutes, walk around court in the sun
12:20 - 12:30	10 minutes, run around court in the sun
12:30 - 12:40	10 minutes swimming
12:40 - 12:50	10 minutes, expose front and back in the standing position

Total Sun Exposure: Group 7 - 60 minutes: This exposure was approximately equal to 1 MED dose under the experimental conditions described above. With the fear that TMP might induce undue cutaneous photosensitization, the duration of sun exposure was kept to a minimum.

Results: TMP induced photosensitzation response certainly appears to be significantly blocked by all these preparations. The skin areas that received preparations G, MG and MAP (D5) did not show any augmented sunburn. These results certainly reveal an added information that is very useful for the benefit of the patients receiving TMP. The effects of over exposure to sunlight can be controlled by topical application of these preparations.

Experiment # 7: Effect of MAP (D5) on 4,5',8-trimethylpsoralen (TMP) Induced Photosensitization*

Subject & Prison #	Site	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Craven	Front	Oral TMP (control)	60	++	0	15	0	
24540 (1)	"	Oral TMP + topical MAP (D5) on right side	"	±	66	5	66	
	"	Oral TMP + topical G on left side	"	±	66	5	66	
	Back	Oral TMP (control)	60	+	0	8	0	
	"	Oral TMP + topical MAP (D5) on right side	"	±	50	5	38	
	"	Oral TMP + topical G on left side	"	±	50	5	38	

* Note: The MED (minimal erythema dose) without oral ingestion of TMP under the experimental conditions described in the protocol of Experiment 7 for subjects # 1-4, was at least 60 minutes for the back region and approximately 75 minutes for the front region.

Experiment # 7: Effect of MAP (D5) on 4,5',8-trimethylpsoralen (TMP) Induced Photosensitization*

Subject & Prison #	Site	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Hansen	Front	Oral TMP (Control)	60	±	0	4	0	
28271 (2)	"	Oral TMP + topical MAP (D5) on right side	"	0	100	0	100	
	"	Oral TMP + topical G on left side	"	0	100	0	100	
	Back	Oral TMP (Control)	60	±	0	4	0	
	"	Oral TMP + topical MAP (D5) on right side	"	0	100	0	100	
=	"	Oral TMP + topical G on left side	"	0	100	0	100	

* Note: The MED (minimal erythema dose) without oral ingestion of TMP under the experimental conditions described in the protocol of Experiment 7 for subjects # 1-4, was at least 60 minutes for the back region and approximately 75 minutes for the front region.

Experiment # 7: Effect of MAP (D5) on 4,5',8-trimethylpsoralen (TMP) Induced Photosensitization*

Subject & Prison #	Site	Preparation	Period of Sun Exposure (Minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
McNatt	Front	Oral TMP (control)	60	+	0	5	0	
27152 (3)	"	Oral TMP + topical MAP (D5) on right side	"	±	50	1	80	
	"	Oral TMP + topical modified G on left side	"	±	50	1	80	
	Back	Oral TMP (control)	60	++	0	11	0	
	"	Oral TMP + topical MAP (D5) on right side	"	±	66	4	64	
	"	Oral TMP + topical modified G on left side	"	±	66	4	66	

* Note: The MED (minimal erythema dose) without oral ingestion of TMP under the experimental conditions described in the protocol of Experiment 7 for subjects # 1-4, was at least 60 minutes for the back region and approximately 75 minutes for the front region.

Experiment # 7: Effect of MAP (D5) on 4,5',8-trimethylpsoralen (TMP) Induced Photosensitization*

Subject & Prison #	Site	Preparation	Period of Sun Exposure (minutes)	Degree of Erythema after 24 Hours: Visible Redness	% Protection	% Reflectance Difference	% Protection	Remarks
Mathews	Front	Oral TMP (Control)	60	±	0	5	0	
25788 (4)	"	Oral TMP + topical MAP (D5) on left side	"	0	100	0	100	
	"	Oral TMP + topical G on right side	"	0	100	0	100	
	Back	Oral TMP (Control)	60	+	0	9	0	
	"	Oral TMP + topical MAP (D5) on left side	"	0	100	5 IPD	100	
	"	Oral TMP + topical G on right side	"	0	100	5 IPD	100	

* Note: The MED (minimal erythema dose) without oral ingestion of TMP under the experimental conditions described in the protocol of Experiment 7 for subjects # 1-4, was at least 60 minutes for the back region and approximately 15 minutes for the front region.

CLINICAL TRIAL ON THE SNOW COVERED MOUNTAINS OF THE SWISS ALPS

1968

Dr. Edgar Frenk, who had participated and collaborated in the first clinical trial at the Arizona State Prison in 1966, agreed to evaluate preparations MAP, MAP (D5), and G on the snow covered mountains of the Alps (Glaciers des Diablerets).

Objective: After a single application, evaluate preparations MAP, MAP (D5), G and UVAL under the following conditions:

a) High altitude

b) Snow covered mountains that reflect ultraviolet radiation

c) Activity: mountain climbing and skiing

Protocol: 19 volunteers participated in this trial in the Alps (7 male subjects and 12 female subjects). All were fair-skinned Caucasians.

Group 1: 10 subjects participated on April 9, 1968. Weather was clear, sunny, very windy and cold. Temperature was 0-5°C in the shade.

Group 2: 9 subjects participated on June 6, 1968. Weather was sunny with intermittent clouds. Temperature was 12-15°C.

The subjects were assembled at a point 2900 meters above sea level. The volunteers received a single application of preparations MAP, MAP (D5), G and UVAL. Only 2 preparations were evaluated on any one subject. Each subject received an application of either MAP, MAP (D5) or G on one side of the face, neck (anterior cervical and sternocleidomastoid region) and forearm (anterior and posterior). Adjacent to the treated areas, small size control (untreated) areas were included to obtain the response of the untreated skin to sun exposure. For comparison, UVAL or preparation G was applied on the other side. Towards the end of the day and on the following day at 24 hours after sun exposure, the sunburn response of the treated and the untreated area was evaluated.

After a single application of the test preparations, the subjects mounted to the summit of the Glaciers des Diablerets. This spot is 3200 meters above sea level, and it took 2 hours for each group to reach the summit. At the summit, the participants rested for 1 hour in the sun. From the summit, the subjects went skiing down into the valley to a point 2000 meters above sea level. This skiing period lasted for 1 hour. Thus each subject was exposed to approximately 4 hours of solar radiation.

Clinical Trial-Swiss Alps (continued)

Results: The erythemal response of the control (untreated) areas for each subject ranged from ++ to ++++ for different sites. Subjects that were treated with MAP, MAP (D5) or G showed practically no erythemal response in the treated areas. All preparations showed excellent protection including UVAL. The general impression of Dr. Frenk was as follows: "Preparation MAP and MAP (D5) seemed to be better than G; G and UVAL showed no evident difference, and MAP or MAP (D5) were equally good."

Conclusions: On the ultraviolet reflecting snow covered mountains, preparations MAP, MAP (D5) and G afford very effective protection during mountain climbing and skiing. As a dermatologist, Dr. Frenk feels that skiers have a very difficult problem of protection against highly reflecting solar radiation during the skiing season. The commercial products do not provide adequate and satisfactory protection when skiers are mountain climbing and skiing. As a result of this trial, it appears that the preparations MAP, MAP (D5) and G can eliminate the hazards of facial sunburn while skiing.

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13. ABSTRACT

Report covers laboratory study and clinical trials on a series of chemical formulations to find means of chemically binding agents to skin elements or of permanent absorption to provide long term (7 days) protection from sunburn regardless of heavy sweating or washing actions. Results are reported in detail and tabulated. Two preparations were developed which were effective for long term exposure (4 hr) after single application and were resistant to washing action. Preparations did not interfere with tanning action and were cosmetically acceptable. Two were very effective under conditions of bright sun in dry climate or high elevation and snow environments, Stable binding to skin providing 7-day or longer protection was not achieved

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14. KEY WORDS	LINK A		LINK B		LINK C	
	ROLE	WT	ROLE	WT	ROLE	WT
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Keratin						
Stratum						
Corneum						
Ultraviolet rays						
Sunburn						

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