EVALUATION OF EFFECT OF DIFFERENT CONCENTRATIONS OF TITANIUM DIOXIDE ON SUN PROTECTION FACTOR.

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Summary

Sun protection products have been used for mitigating the harmful effects of Ultra Violet (UV) sun rays from time immemorial. These products protects against sunlight induced erythema with the level of performance indicated by sun protection factor (SPF). The present investigation was carried out with the aim to determine the effect of different concentrations of titanium dioxide on SPF using mice. In this investigation the mice were divided into four groups. Group I served as control, Group II and III served as test groups and treated with formulation containing 10% and 20% titanium dioxide respectively. All the mice were anesthetized, shaved 2 X 2 cm² area on their back and exposed to UV rays (SPF apparatus) at 850 µw/cm² on 1 x 1cm² shaved area and they were periodically observed for burn sign & skin erythema and the UV exposure time required to produce such signs was noted down. The Minimal erythema dose (MED) of exposed area is compared with non exposed area. The results of this study showed that the sample was found to have SPF of the formulation containing 10% titanium dioxide was found to be 6.18 ± 0.14 whereas the SPF of formulation containing 20% titanium dioxide was found to be 14.33 ± 0.25 which indicates that the formulation containing 20% titanium dioxide shows favorable retention time in normal sunlight.

Keywords: sun protection factor, titanium dioxide, Minimal erythema dose, Ultra Violet.
Introduction

Sun burn is the typical ailment in the tropical or moderate climatic countries. Changes in the climate or by sunbathing leads to general increase in daily exposure of skin to ultraviolet (UV) light. Depletion of the stratospheric ozone layer from the most damaging solar UV radiations may also contribute to this increased exposure and further complications like skin cancer\(^1\). As a consequence the hazards associated with exposure to the UVA and UVB components of sunlight, which include erythema, sunburn, photo damage, photo carcinogenesis, and damage to eyes\(^2-6\). Phototoxicity is generally characterised by erythema and hyperpigmentation. UVB radiations may cause specific damage to macromolecules, such as DNA, RNA and protein membranes\(^7\). The usual methods of mitigating these harmful effects are wearing protective clothing, staying out of sun etc. however these methods have their limitations on the other hand the use of sunscreen agents is another method which is becoming popular day by day\(^8-10\). Sun protection products have been used for mitigating the harmful effects of UV sun rays from time immemorial. These products protect against sunlight induced erythema with the level of performance indicated by sun protection factor (SPF).sun protected products have long protected against sunlight induced erythema with the level of performance indicated by the sun protection factor(SPF). However, since the SPF number is influenced primarily by UVB wavelengths, it is not sufficiently indicator of a sunscreen product’s protection against UVA exposure\(^11\).

In recent years, the harmful effects of the UVA wavelengths of sunlight have been more thoroughly established. With this understanding arose a need, not only for sun protection products that were effective against the UVA wavelengths, but also for a common test method for measuring UVA protection levels\(^11\).

Materials and methods

Animals:

Male Swiss albino mice (25±2 g), were procured from the animal house of AISSMS College of Pharmacy, Pune. Mice were placed randomly in polypropylenes cages (six per cage) with paddy husk as
bedding. Animals had free access of standard pellet animal diet and water.

Chemicals and drugs:
Thiopentone sodium injection, Test formulation.

Apparatus:
The SPF apparatus was fabricated as per reported standards. It consists of two wooden compartments (60 x 60 x 140 cm). Upper compartment for holding UV radiation source and lower compartment to place animal. Mercury lamp 160 W was used as radiation source to induce erythema. A UV meter was used to measure intensity of irradiation.

Procedure:
The mice were divided into three groups. Group I served as control, Group II and Group III served as test groups and treated with titanium dioxide formulations. All the mice were anesthetized with thiopentone (90 mg/kg/i.p.). A mercury lamp (OSRAM) was warmed up for about 10 minutes prior to use and placed at a constant distance (20 cm) above the animal. The intensity was adjusted and stabilised. The mice were shaved 2 X 2 cm² area on their back and exposed to UV rays (SPF apparatus) at 850 µw/cm² on 1 x 1 cm² shaved area and they were periodically observed for burn sign & skin erythema and the UV exposure time required to produce such signs was noted down. The MED of exposed area is compared with non exposed area. The UV exposure time to produce such sign was noted down. Minimal erythema dose assessment was carried out by visual (morphological) observation; unprotected and protected skin was observed by same observer⁷,¹⁴.

Observations: Intensity of irradiation of radiation measured in µw/cm² by using following factor:

\[
\text{Intensity of irradiation (J/cm}^2\text{)} = \text{intensity of irradiation (µw/cm}^2\text{)} \times \frac{\text{Time required to Produce erythema (min.)}}{1.66}
\]
Table no 1: Time required to produce erythema in mice.

<table>
<thead>
<tr>
<th>Sr no.</th>
<th>Control</th>
<th>Standard</th>
<th>WL-1(10%)</th>
<th>WL-2(20%)</th>
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</thead>
<tbody>
<tr>
<td>1</td>
<td>9</td>
<td>140</td>
<td>65</td>
<td>151</td>
</tr>
<tr>
<td>2</td>
<td>8</td>
<td>145</td>
<td>60</td>
<td>148</td>
</tr>
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</tr>
<tr>
<td>6</td>
<td>11</td>
<td>137</td>
<td>63</td>
<td>146</td>
</tr>
</tbody>
</table>

Mean ±SEM

<table>
<thead>
<tr>
<th></th>
<th>Mean ±SEM</th>
<th>Mean ±SEM</th>
<th>Mean ±SEM</th>
<th>Mean ±SEM</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>10.16 ± 0.6009</td>
<td>135.33 ± 2.654***</td>
<td>62.83 ± 1.138***</td>
<td>145.33 ± 1.626***</td>
</tr>
</tbody>
</table>

Values are expressed as Mean ± SEM (N=6)

***The P value is < 0.001, considered significant.

Calculation of SPF:

Formula:

\[
SPF = \frac{\text{Minimal erythema dose in Test formulation protected skin in j/cm}^2}{\text{Minimal erythema dose in Non sunscreen protected skin in j/cm}^2}
\]

Results

MED was evaluated in normal grouped animals and accordingly the standard, WL-1 and WL-2 group. Mice were exposed to the UV radiation fixed at 850μw/cm². The average time was calculated to be 9 min 30 sec for producing erythema. The
morphological evaluations of the skin of standard, WL-1, WL-2 were found to be similar. From the observed time to produce skin burn the MED was evaluated for SPF calculation. The SPF of standard was found to be 13.58 ± 0.80 however the same of WL-1, WL-2 were found to be 6.18±0.1471 and 14.33±0.2540 respectively.

Table no.2: Sun Protection Factor.

<table>
<thead>
<tr>
<th>Animal No.</th>
<th>Sun Protection Factor</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Standard</td>
</tr>
<tr>
<td>1</td>
<td>14.01</td>
</tr>
<tr>
<td>2</td>
<td>14.51</td>
</tr>
<tr>
<td>3</td>
<td>12.81</td>
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<td>4</td>
<td>13.01</td>
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<tr>
<td>5</td>
<td>13.2</td>
</tr>
<tr>
<td>6</td>
<td>12.45</td>
</tr>
<tr>
<td>Mean ± SEM</td>
<td>13.33±0.3172</td>
</tr>
</tbody>
</table>

Discussion

The sun protection test is used specifically to evaluate the sunscreen agents. The sun protection test measures ability of sunscreen to protect skin from sun radiation. The standard formulations exert their sun protection effect via blocking the UV radiations. Exposure to the UVA and UVB components of sunlight, which include erythema, sunburn, photo damage, photo carcinogenesis, and damage to eyes²-⁶. In sun protection test WL-1 and WL-2 showed significant (p <0.05–0.001) and dose dependant sun protection action. From the results it can be inferred that the Test formulation (WL-2) was found to have SPF of 14.33 ± 0.25 and shows moderate retention time in normal sunlight.
References


11. Notes of guidance for testing of cosmetic ingredients for their safety evaluation. Third revision the scientific committee on cosmetic products and non food products intended for consumers. 1999

