Synthesis, Characterization and Sunscreen Protection Factor of Novel Synthesized Para Amino Benzoic Acid (PABA) Esters

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Summary: Human exposure to sunlight can activate various biological responses from erythema to skin cancer. Precise use of sunscreen is the most efficient way to prevent the development of these diseases. Ingredients of sunscreen can be organic and inorganic ultraviolet (UV) filters. One of such organic ingredients is para amino benzoic acid (PABA), which is being used as sunscreen filter. The purpose of this study was to enhance the sun protection factor (SPF) value of PABA and minimize the harmful effects of PABA due to its deep penetration into skin. For this purpose, synthesis of two novel sunscreen filters PABA methyl stearate and PABA methyl ester were done by reacting PABA with olive oil at various conditions. The SPF value of each solution was determined by the standard spectrophotometric method. From SPF calculations, it was observed that both novel sunscreen filters showed high SPF number of 20.60 and 26.17 PABA methyl ester and PABA methyl stearate respectively while PABA showed the low SPF number of 17.60 at the same concentration. The PABA methyl stearate is less harmful to human skin because it has less penetration due to large size as compared to PABA.

Key words: Sunscreen, Para amino benzoic acid (PABA), Sun protection factor (SPF), Skin cancer.

Introduction

Skin is the primary protection of body for external exposure to the environment. The symptoms of ageing can highly be judged by the skin. However, growing older is not a threat for someone, but it can have negative impacts on human perception [1]. Untimely growing old befalls as an instantaneous or secondary outcome of skin's contact with surroundings. Each year, around a million humans are recognized with skin diseases and approximately ten thousand used to expire from skin cancer called malignant. The higher risk of skin cancer is on the parts of the body which is often uncovered to the rays of sun, including face, arms, head and back of the hands and neck as well [2].

Sun rays contain gamma rays, x-ray, visible, ultraviolet, infrared and radio waves. These bands of radiations reach to the earth surface including wavelengths of electromagnetic ray's ranges from 290 to 3000 nm. While at the same time, the spectrum concerned in skin responses involves wavelengths around 290 to 1800 nm. The radiations of wavelength below 290 nm are absorbed by ozone layer. The dangerous outcomes of sun light are triggered chiefly by the ultraviolet band of the sun spectrum [3]. This spectrum is separated to three sections: first is UVA, which ranges from 400 to 320 nm; second is UVB, which ranges from 320 to 290 nm and last is UVC, ranges from 290 to 200 nm. The third UVC rays are filtered by ozone before reaching to ground [4]. UVB radiation is not always absolutely filter by the ozone layer and is accountable for the skin destruction causing sun burn or sun tan. The UVA type of light reaches the deeper layers of the dermis and epidermis and incites the skin early aging and is liable for free radical formation [5]. The UVB type of ultraviolet light can damage 60% of skin. Exposure to UV rays has definite acute as well as chronic consequences on the human skin. People are aware of the possible threats of photo ageing; sun burn and skin cancers which are taking place because of solar radiations [6].

The use of sunscreen is recommended for protection of human skin [7]. To save people from destructive solar radiations, many sunscreens and sun blockers are commercially available. The sunscreens are ultraviolet filters and absorb UV radiations like water soak up by a sponge which drops it back again by changing it into reduced strength of benign IR radiations. The sun blockers are applied to human skin and resist by reflecting or scattering the UV light instead of absorption [8]. Experts around the world manufactured many lotions and creams and wherein there is addition of active parts of sunscreen to save people from harmful radiation by absorption or reflection of light. These particles in sunscreens take in maximum of ultraviolet-B rays and save people from it [9].

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Para-Amino Benzoic Acid (PABA) is one of such active agent in sunscreen which avoids sunburn and guards people from dangerous consequences of solar rays. Para-amino benzoic acid is an organic filter that absorbs UV rays and visible solar rays. On the other hand, PABA also has some dangerous outcomes because of its deep penetration into the skin [10].

The activity of sunscreen is measured in terms of sun protection factor (SPF). The sun protection factor is a kind of scale which shows just how a whole safety is being supplied as counter to ultraviolet light [11]. There are few methods available for calculating SPF; Mansur mathematical equation is one of the methods that use the ultraviolet Spectrophotometer to get SPF Values [12]. Higher is the sun protection factor value, higher will be the ultraviolet light filtered by sunscreen and the better is the protection and safety [13]. So, this sun protection factor offers the universal system for solar safety. A quantity of time, taken by for sun burning relies upon the extent of UV radiations and differs rendering to time of day, time of the year, the climate and individual's skin color [14]. None of the sunscreen could curtain whole ultraviolet radiations. All sun screens are filtering maximum ultraviolet rays and permitting few ultraviolet rays to go into the skin [15].

Triglycerides or fats are the chief constitutions of olive oil and it comprises minor extents of glycerol and free fatty acids as well and there are microscopic quantities of phosphatides, flavor compounds, sterols and pigments present in olive oil. Triglycerides or fats are the main reserves of energy for animals and plants. The molecules of Triglycerides are resultant of the natural esterification of a glycerol molecule with three molecules of fatty acid shown below in Fig. 1.
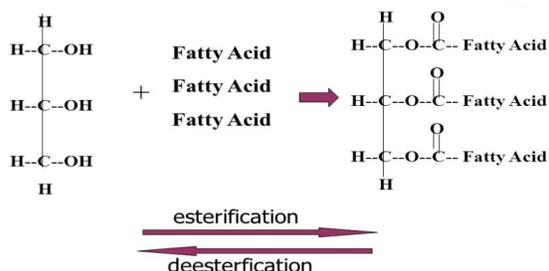

Fig. 1:   Formation of triglycerides [16].

The chief fatty acid in triglycerides of olive oil is oleic acid which is a monounsaturated fatty acid that makes up to 65 to 85% of olive oil. There are some other fatty acids as well which are present in tiny amount like linoleic acid, palmitic acid, linolenic acid, and stearic acid [17]. The main benefits of using olive oil on skin are that it acts as natural antioxidants and have 7.5 SPF value [18]. Olive oil is insoluble in water, relieves your dry skin, helps to fight against skin cancer and helps your skin to look more youthful [19].

Experimental

Synthesis of Novel Organic Sunscreen Filters

To modify the PABA by using edible oil, Olive oil (unsaturated fat) was neutralized from free fatty acids with standard solution of alkali like Sodium hydroxide. In this process, soapy layer formed which was separated from fat. The neutralized solution of olive oil was refluxed in two separate reflex condenser assembly X and Y. For refluxing of vaporized contents during reaction of sample X, 15ml of fat was taken in the flask and well fitted for circulation of cold water. Then 2 grams of para-amino benzoic acid (PABA) in reflex condenser along with 0.2 grams of benzyl per oxide as initiator was added. The refluxing contents were heated at 60°C temperatures for three hours. Bumping agents were used for stirring in flask. On completion of the reaction, the product was filtered.

In another flask 15ml of fat and 2 grams of para-amino benzoic acid (PABA) was taken (sample Y). The refluxing contents of sample Y were heated at 250 centigrade temperatures for three hours. Then the product was filtered.

Characterization

In order to understand the composition; both synthesized products were analyzed through GCMS. For characterization, 2 ml of each synthesized product was dissolved in suitable amount of methanol. Each sample X and Y was run through GCMS.

Optimization of activity of UV sunscreens

Synthesized and characterized products were used for further studies of their efficiency for filtration of UV radiation.

To study the efficiency of novel sunscreen filters, sample were analyzed through Double beam UV-Vis spectrophotometer equipped with 1cm quartz cell. For this analysis, three solutions of each X and Y samples with different concentrations i.e. 1gm,

0.5gm and 0.25gm were prepared in 100 ml chloroform. For comparison, solutions of Para-Amino Benzoic Acid (organic sunscreen filter) were also prepared in 100 ml ethanol.

These all samples were analyzed to estimate absorbance of UV radiations by using UV-Vis spectrophotometer in range from 290 nm to 320 nm and finally Mansur equation [20] applied to calculate the SPF. Mansur equation to calculate the SPF is given as:

$$SPF = CF \times \sum_{320}^{290} EE\ (ë) \times I\ (ë) \times Abs\ (ë)$$

Here

CF =   Correction factor and its value is 10 given by Mansur
EE (λ) = Erythrogenic effect of the rays having wavelength (λ)
Abs (λ)=Spectrophotometric absorption value having wavelength (λ)

The value of EE x I is constant, this had been determined by Sayre [21]

For calculation of SPF, the absorbance (λ) values of sample solutions at given wavelength were multiplied with the respective EE (λ) x I (λ) values. Then, their summation was taken and multiplied with the correction factor. Then, SPF values of sample solutions obtained for X, Y and PABA are given in Table-2, Table-3 and Table-4 respectively.

Results and Discussion

Synthesis

For two novel sunscreen filters, following reactions took place during preparation of sample "X", where triglycerides of olive oil contain up to 85% Oleic Acid (C18:1), a monounsaturated fatty acid. The product formed in this reaction was triglycerides PABA. Each chain of triglyceride PABA is known as "9-(4-Amino Benzoate), Methyl Stearate" which is also called PABA Methyl Stearate. This synthesis was confirmed by analysis through GCMS as shown in Fig 2.
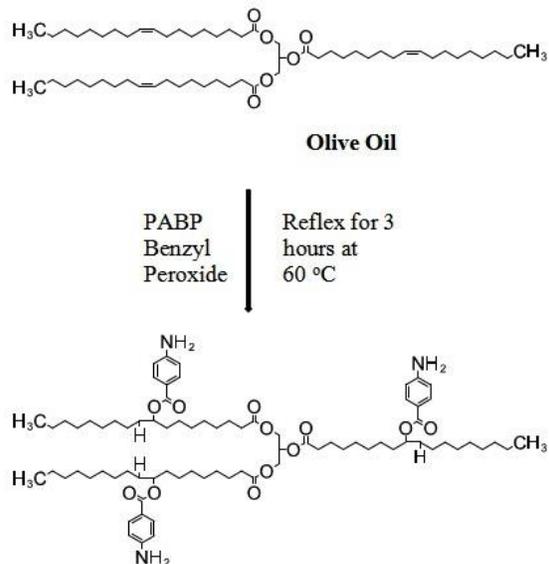

Olive Oil

Tri-Glycerides PABA

Fig. 2:   Chemical reaction during synthesis of sample "X"

While PABA Methyl Ester is formed in sample "Y" given below in Fig 3.

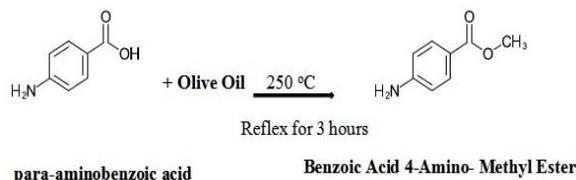para-aminobenzoic acid   Benzoic Acid 4-Amino- Methyl Ester

Fig. 3:   Chemical reaction for sample "Y".

Characterization

In order to confirm the formation of desired product, synthesized products were analyzed. One major peak was obtained from GCMS spectrum of each sample. From these peaks, the library data of GCMS disclosed the names and structures of synthesized products in each sample X and Y. Along with major peaks, few small peaks can also be observed which might be due to the non-reacted parts of reactants or some products formed in negligible amount. Spectrum obtained from GCMS and Structure of compound for the sample X and Y are given in the Fig 4 and 5 respectively.
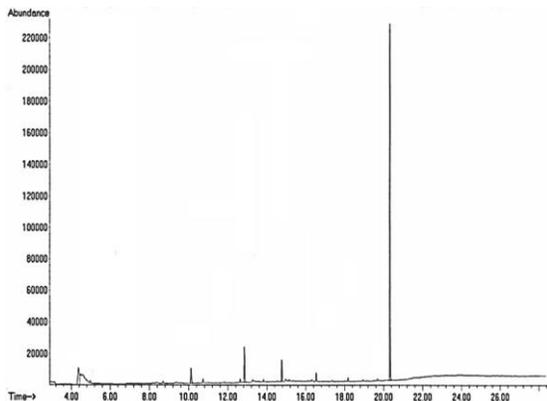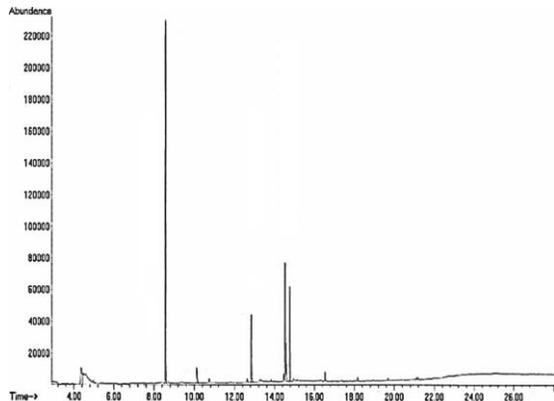
Fig. 4:   GCMS spectrum of sample "X".

Fig. 5: GCMS spectrum of sample "Y".

UV Sunscreens absorption Activity

A UV spectrophotometric method is used for measuring SPF of sunscreen ingredients. SPF values of PABA (UV organic filter) and novel synthesized sunscreen filters (PABA esters) were achieved by use of ultraviolet spectrometric, approaches at three different concentrations then the Mansur equation was applied to calculate the SPF.

Table-1: Normalized product functions for SPF calculation.

Wavelength (nm)	EE X I (*normalized*)
290	0.0150
295	0.0817
300	0.2874
305	0.3278
310	0.1864
315	0.0837
320	0.0180
Total	**1**

EE: Erythrogenic effect   I: solar intensity

Table-2: SPF values of sample X (PABA methyl stearate) at different concentrations.

Conc. g/100ml	wavelength	290	295	300	305	310	315	320	SPF
	EE x I	0.015	0.081	0.287	0.327	0.1864	0.083	0.018	
0.25	A	0.605	0.602	0.812	1.522	1.405	1.612	1.612	12.99
	EE x I x A	0.0091	0.0491	0.2334	0.5965	0.2609	0.0289	0.0289	
0.5	A	1.032	1.356	1.958	2.403	2.818	2.827	3.025	22.87
	EE x I x A	0.0154	0.1103	0.5604	0.7867	0.5237	0.2365	0.0544	
0.1	A	2.002	2.411	3.63	4.66	5.24	5.80	6.0	42.47
	EE x I x A	0.0300	0.097	1.043	1.528	0.976	0.487	0.108	

Table-3: SPF of sample Y (PABA methyl ester) at different concentrations.

Conc. g/100ml	wavelength	290	295	300	305	310	315	320	SPF
	EE x I	0.015	0.081	0.287	0.327	0.1864	0.083	0.018	
0.25	A	0.436	0.456	0.609	0.888	0.898	0.805	0.854	7.6
	EE x I x A	0.0065	0.0369	0.1747	0.2903	0.1673	0.0668	0.0128	
0.5	A	0.987	0.987	1.288	1.989	2.471	2.622	2.455	18.4
	EE x I x A	0.0148	0.0799	0.3696	0.6504	0.5351	0.2176	0.0441	
0.1	A	2.602	2.731	3.397	3.678	3.983	3.893	3.981	35.8
	EE x I x A	0.039	0.2212	0.9749	1.2027	0.7424	0.3231	0.0716	

Table-4: SPF values of PABA at different concentrations.

Conc. g/100ml	wavelength	290	295	300	305	310	315	320	SPF
	EE x I	0.015	0.081	0.287	0.327	0.1864	0.083	0.018	
0.25	A	0.410	0.479	0.603	0.705	0.700	0.705	0.712	6.519
	EE x I x A	0.0062	0.0300	0.1733	0.2311	0.1304	0.0591	0.0128	
0.5	A	0.811	0.992	1.205	1.412	1.401	1.410	1.414	15.370
	EE x I x A	0.0121	0.0810	0.3463	0.4628	0.2611	0.1183	0.2545	
0.1	A	1.630	1.932	2.501	2.814	2.802	2.820	2.820	30.923
	EE x I x A	0.0244	0.1590	0.7185	0.9224	0.5223	0.2365	0.5090	

Results of Mansur mathematical equation for determination of SPF of PABA esters and PABA at three different concentrations are shown in table 5;

Table-5: Average Results of three UV Filters at three different concentrations

SUNSCREEN FILTERSTYPES	AVERAGE SPF VALUES
PABA methyl stearate	(42.67+22.87+12.99)/3=26.17
PABA methyl ester	(6.51+15.37+30.92)/3=20.60
PABA	(30.92+15.37+6.52)/3=17.60

It was noted that the SPF values were directly depended on concentrations of PABA (UV organic filter) and novel synthesized sunscreen filters which are called PABA esters. At the same concentration, new synthesized sunscreens have greater SPF value than PABA. So, ultraviolet light with high concentration might be filtered by X and Y sunscreen filters with better protection from UV radiations. The society of cancer recommends that SPF 30 plus sun screen is enough for solar safety when used efficiently.

A model sun screen ingredient needs to be biochemically inactive, safe, reliable no exasperating, photo stable and must offer entire skin defense. Novel synthesized filter absorbs radiation in ultraviolet region with high absorption capability, so it can be used in sunscreens. Novel synthesized organic filters PABA methyl ester and PABA methyl stearate can absorb UV light and after absorbing UV radiations, these compounds emitted their energy as IR radiations and therefore stop UV light to reach deeper into the skin. Since PABA methyl stearate has less penetration into human skin because of large size as compared to PABA. As we know that larger the size of molecule less would be the penetration into skin. In this way, harmful effects of PABA will diminish due to its deep penetration into the skin.

Conclusion

The present study concludes that the novel synthesized sunscreen filters (PABA esters) has better strength to protect against UV-light with elevated sun protection factor (SPF) than PABA itself. Additionally, the active ingredient like PABA methyl stearate reduced the harmful effect of PABA as it has less penetration into skin because of their large size. So, these filters are safe and better alternatives to harmful chemical sunscreen filter like PABA that is being used in industries.

Acknowledgment

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