

A Sunscreen Based on Avobenzone Loaded in Solid Lipid Nanoparticles for Enhanced Sun Protection

Jawahar Natarajan*, Pavalan Krishnan, Sebatini Sinsi Arokianathan, Sharuni Sivakumar, Parish Vendar Saravanan, Roshan Prasad Rao

Department of Pharmaceutics, JSS College of Pharmacy, JSS Academy of Higher Education and Research, Ooty, Nilgiris, Tamil Nadu, INDIA.

ABSTRACT

Objectives: Sunscreen cosmeceuticals play a crucial role in protecting the skin from dangerous ultraviolet radiation, which is linked to various dermatological diseases and skin cancers. Avobenzone is extensively used organic ultraviolet ray gauze; however, due to its inherent instability and limited skin penetration, it has led to research into developing novel delivery systems. This study introduces a pioneering sunscreen formulation employing avobenzone incorporated in solid lipid nanoparticles to enhance sun protection efficacy. **Materials and Methods:** The solid lipid nanoparticles were prepared by the microemulsion method by using glycerol monostearate, soya lecithin, avobenzone and Pluronic F127, which results in the formulation of nano-emulsion and the subsequent formation of solid lipid nanoparticles by lipid precipitation and the sunscreen is loaded in the solid lipid nanoparticles by mixing both the oil phase (cetyl alcohol, cocoa butter) and the aqueous phase (triethanolamine, water) together until a cream-like consistency is obtained. This formulation was then characterized for particle size, zeta potential, entrapment efficiency, sun protection factor determination, microbial studies and photostability studies. **Results:** The results indicate a reduction of avobenzone concentrations with a decrease in particle size and show good entrapment efficiency as well as, an ideal sun protection factor range for better ultraviolet absorption, validating the potential of solid lipid nanoparticles-based delivery. Moreover, the microbial studies and photostability assessments demonstrated the safety and biocompatibility of the developed sunscreens. **Conclusion:** This new sunscreen cosmeceutical offers a promising avenue for effective ultra violet protection and encourages further exploration of solid lipid nanoparticles in sunscreen enhancement.

Keywords: Sunscreen, Ultraviolet Radiation, Avobenzone, Solid-Lipid Nano Particle, Microemulsion.

Correspondence:

Dr. Jawahar Natarajan

Associate Professor, Department of Pharmaceutics, JSS College of Pharmacy, Ooty-643003, Nilgiris, Tamil Nadu, INDIA.
Email: jawahar.n@jssuni.edu.in

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INTRODUCTION

A material that aids in protecting the skin from the sun's harmful rays. Sunscreens provide dual protection against ultraviolet A and B radiation by reflecting, absorbing and scattering the latter (National Cancer Institute, 2024). The use of sunscreen is complicated and involves a number of public health-related issues. Media reports concerning "dangerous" sunscreens frequently spread prior to the summer holidays (Azurdia *et al.*, 1999; Wright *et al.*, 2001). We offer Solid Lipid Nanoparticles [SLN] as a dynamic drug carrier technology that is water-soluble and effective for correction. Nanoparticles are colloidal particles with sizes ranging from 10 to 1000 nm (Kamble *et al.*, 2010). Aqueous colloidal dispersions with solid biodegradable lipids as the matrix make up solid lipid nanoparticles (Garud *et al.*, 2012).

The last ten years have seen a rise in the use of sunscreen due to ozone layer damage, which allows UV radiation to damage skin. To help with the long-term negative effects brought on by UV radiation A and UV radiation B, topically administered UV blocking medicines are utilized. In light of the aforementioned circumstances, sunscreen efficacy and safety are crucial in order to address the aforementioned issue, which calls for frequent and continuous application. Sunscreen should ideally not permeate the skin, be photostable and work well at low concentrations on the skin. Sunscreen agents fall into two categories: inorganic sunscreens (also known as physical sunscreens) and organic sunscreens (also known as chemical sunscreens). UV filters, often known as organic sunscreen, are active components that absorb UV rays within a specific wavelength range. The UV filters go from low-energy ground to high-energy excited states once they absorb radiation. Physical or inorganic sunscreen works by diffusing and reflecting ultraviolet light. Zinc Oxide (ZnO) and Titanium Dioxide (TiO₂) are the components that are most frequently utilized (Kaimal and Abraham, 2011). Because of its photostability and unfavourable side effects, which include acne,



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rashes, skin swelling or redness and soreness in areas of the skin with hair, organic sunscreen use has been discouraged recently (Xia *et al.*, 2007). Sun damage can have long-term negative effects on skin health and appearance. Wrinkles and lines are caused by Ultraviolet (UV) light damage to the skin and the connective tissue beneath it. Sun damage and UV exposure have compounding consequences you should take all reasonable precautions to protect your skin from UVA and UVB radiation, as they can cause deeply penetrating damage to your skin and cause sunburn (Dhangar *et al.*, 2024). It is still important to wear sunscreen every day, especially in cloudy weather. On a scale of II, the UV index is rated as "moderate," with a score of 3-5. Sunscreen functions by shielding the skin from UV rays, which lowers the chance of sunburn, discoloration and anti-aging symptoms. We focus on making the sunscreen photostable, low concentration and loaded with natural antioxidants and SLN (physical reflector) to address the aforementioned issue. This approach may have a synergistic effect in regulating sun protection (Moloney *et al.*, 2002). Medication employed in SLN are from BCS classes II and IV. SLN are colloidal carrier systems made of high melting point lipids as a solid core coated by an aqueous surfactant (Loxley, 2009). Any method of preparing SLNs results in dispersion form, which causes instability due to the hydrolysis process during long-term storage. By using lyophilization as a spray drying process, they can be transformed into solid, dry powders to improve stability (Sinha *et al.*, 2010). As SLN works as a physical reflector of UV rays and has a high drug-loading capacity, adding the chemical UV absorber avobenzone to it enhances its UV-blocking capacity and avoids chemical deterioration. Synergistic photo protection is achieved when sunscreen is added to SLN. A low concentration of avobenzone is incorporated in SLN, so it helps in reduction of side effect due to avobenzone (Marcato *et al.*, 2011).

MATERIALS AND METHODS

Materials

Avobenzone was bought from Carbanio. Glyceryl monostearate, Soya lecithin, Pluronic F127, cetyl alcohol, cocoa butter, triethanolamine and sodium benzoate were bought from Sigma-aldrich and only distilled water was used throughout the process.

Methods

Preparation of SLN [microemulsion method]

This process involves diluting a microemulsion in a cold aqueous solution, which forms a nano emulsion and causes lipid precipitation to generate solid lipid nanoparticles (Shah *et al.*, 2014).

In short, Drug is dissolved in melted lipids at a temperature higher than the lipid melting point. To create a transparent, thermodynamically stable microemulsion, an aqueous

phase comprising water and surfactant (preheated to the same temperature) is then added with gentle stirring. The microemulsion, which is 25-50 times larger than the hot emulsion, is subsequently added to the cool aqueous phase. Lipids instantly solidify to create solid lipid nanoparticles after dilution, creating a nano emulsion (Shah *et al.*, 2014).

Solid lipid nanoparticles containing medication have been prepared using this technique with success. Its limitations are the large amount of water required to dilute microemulsions its high surfactant usage. Excess water can be removed by lyophilization or ultra-filtration to concentrate solid lipid nanoparticles dispersions (Singh, 2019).

Its high surfactant usage and the substantial volume of water needed to dilute microemulsions are its drawbacks. In order to concentrate solid lipid nanoparticle dispersions, excess water can be eliminated by lyophilization or ultra-filtration (Arianto *et al.*, 2019).

Preparation of SLN-loaded Sunscreen

The needed quantity of cetyl alcohol and cocoa butter were taken in a beaker and heated in a water bath up to 70°C to gain a molten mass and add SLN to the oil phase).

In another beaker, take triethanolamine, water and sodium benzoate and heat up to 75°C (Aqueous phase).

Mix both results by adding one phase into another with continuous stirring until a cream-like consistency.

After obtaining cream consistency, add the needed volume of lemongrass oil (Arianto *et al.*, 2019).

EVALUATION STUDIES

Particle size and Polydispersity Index (PDI)

The average particle size and polydispersity index of SLN formulations were measured by Zeta sizer Nano Zs90 (Anton Paar Lite sizer 500). The samples of SLN dispersions were diluted with deionized water (Ahlin *et al.*, 1998). Results of Particle size and Polydispersity index given in Figure 1.

Zeta Potential

Zeta potential measurement can be carried out using a zeta potential analyzer or zeta meter. Before measurement, SLN dispersions are diluted 50-fold with the original dispersion preparation medium (water) for size determination and zeta potential measurement (Netto *et al.*, 2017). Results of Zeta Potential given in Figure 2.

Entrapment Efficiency (EE)

The centrifugation method was utilized to ascertain the entrapment effectiveness of SLN dispersion. To gather the liquid supernatant, SLN dispersion, which is equivalent to 5 mg of medication, was

centrifuged for an hour at 20,000 rpm in a refrigerated centrifuge. Once the recovered liquid had been suitably diluted with fresh phosphate buffer saline in 7.4, it was screened to determine the concentration of free medication. Using the formula, the absorbance at 207 nm in a UV spectrophotometer was measured to determine the entrapment efficiency (Netto *et al.*, 2017).

$$EE = \frac{\text{weight of drug incorporated}}{\text{weight of drug initially taken}} \times 100$$

SPF Determination

All samples weighed 1 g each, which was then transferred to a 100 mL volumetric flask, diluted to volume with ethanol, ultrasonicated for 5 min and filtered through cotton, discarding the 1st mL. A 50 mL volumetric flask was filled to capacity with ethanol after a 5.0 mL aliquot was transferred there. After that, a 25 mL volumetric flask was filled with ethanol to make up the remaining volume from a 50 mL aliquot. A 1 cm quartz cell was used to get the sample's absorption spectra in solution, with ethanol serving as a blank. The Mansur equation was then used to the absorption measurements, which were obtained in the 290-320 nano meter (nm) region (Griffin *et al.*, 1997).

$$\text{SPF spectrophotometric} = \text{CF} \times \sum (\lambda) \times I (\lambda) \times \text{Abs} (\lambda)$$

Where:

EE (λ)-erythema effect spectrum (Table 1);

I (λ)-solar intensity spectrum;

Abs (λ)-absorbance of sunscreen product;

CF-correction factor (=10) (Griffin *et al.*, 1997).

Microbial Studies

In vitro studies

Pseudomonas aeruginosa and *Staphylococcus aureus* detection

In order to investigate the presence of *P. aeruginosa* and *S. aureus*, 10 mL of the prepared dilution were mixed with 10 mL of fluid soybean casein digest medium that had been enhanced at a concentration that was twice as high as that advised for the standard medium preparation. For 48 hr, the tubes were incubated at 35±2°C. Tubes showing no growth after incubation were declared negative for *S. aureus* and *P. aeruginosa*. The subsequent procedures involved transferring a loop of medium tubes exhibiting growth to Cetrimide Agar, a selective medium for *P. aeruginosa* identification. Additionally, a circle is moved to Baird Parker Agar, especially in the case of *S. aureus*. Upon incubation for 72 hr at 35±2°C, the plates were examined. The sample tested positive for the presence of related bacteria in the case of growth on these selective mediums (Brown and Diffey, 1986).

Yeast and Mold detection

Sample contamination with Mold and yeast was investigated using Sabouraud Dextrose Agar and Broth. A tube containing Sabouraud Dextrose broth that had been enriched at a concentration two times greater than that advised for ordinary medium production was filled with 10 mL of the resulting dilution. For 3-5 days, the tubes were incubated at 25°C. After the incubation period, tubes that showed no turbidity were labelled as negative. A culture loop was moved to a Sabouraud Dextrose Agar plate in the event of turbidity and the plate was incubated at 25°C for 3-5 days. The development of any colony on the plate's surface indicated that yeast or Mold had contaminated the sample; yeast and Mold contamination was detected in the sample.¹⁹ Results of Microbial studies given in Tables 2 and 3, Figures 3 and 4.

Determination of photostability studies

Preparation of SLN loaded sunscreen

Take 10 mg of SLN Loaded Avobenzone Cream and dilute with 6-8 mL of acetonitrile. Place the prepared solution in UV chamber for 45 min to 1 hr. Sonicate the prepared solution for 5 min and make up the volume to 10 mL with acetonitrile. Take 1 mL of the filtrate from the above solution and make the volume into a 10 mL volumetric flask with ACN. From the above solution, prepare the serial dilution of different concentrations, respectively. Check the UV absorbance of the solution (Dutra *et al.*, 2004). Results of Photostability studies given in Table 4 and Figure 5.

Preparation of Blank

Take 10 mg of blank sunscreen without avobenzone and add 6 to 8 mL of ACN. Sonicate the prepared solution for 5 min and make up the volume up to 10 mL with ACN. Place the prepared solution in UV chamber for 45 min to 1 hr. Take 1 mL of the filtrate from the above solution and make up the volume to 10 mL volumetric flask with ACN. Check the UV absorbance of the solution (Dutra *et al.*, 2004).

Table 1: Normalized product function calculation as per Mansur's equation.

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

RESULTS

Photostability studies

SLN Loaded Avobenzone Cream and dilute with 6-8 mL of acetonitrile. Place the prepared solution in UV chamber for 45 min to 1 hr. Sonicate the prepared solution for 5 min and make up the volume to 10 mL with acetonitrile. Take 1 mL of the filtrate from the above solution and make the volume into a 10 mL volumetric flask with ACN.

After performing the photostability test with the help of a UV spectrophotometer, the absorbance increases with an increase in concentration of sunscreen.

Particle Size, PDI and Zeta Potential

Particle size and Poly dispersity index

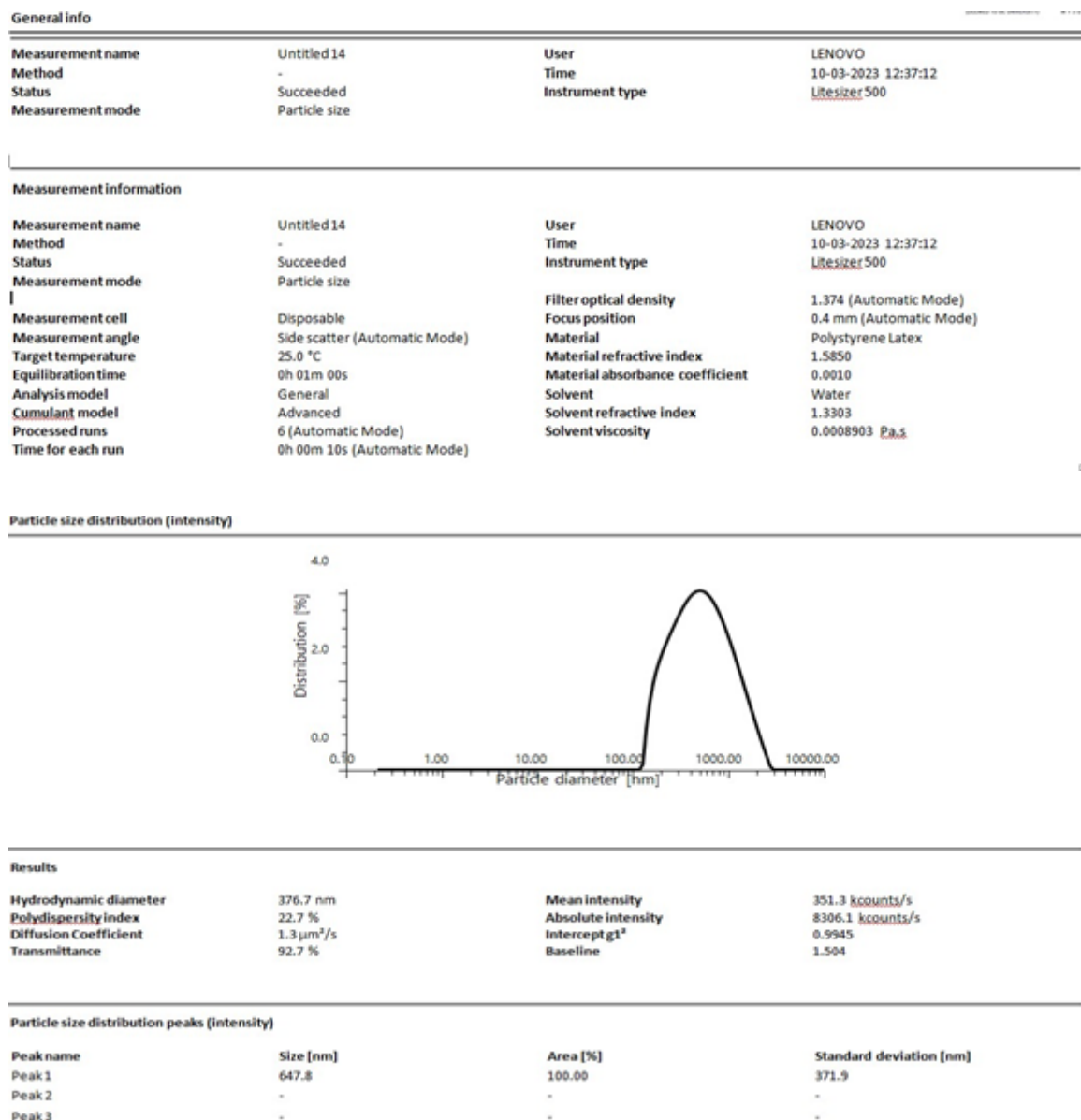


Figure 1: Result of Particle size and PDI results.

Zeta potential

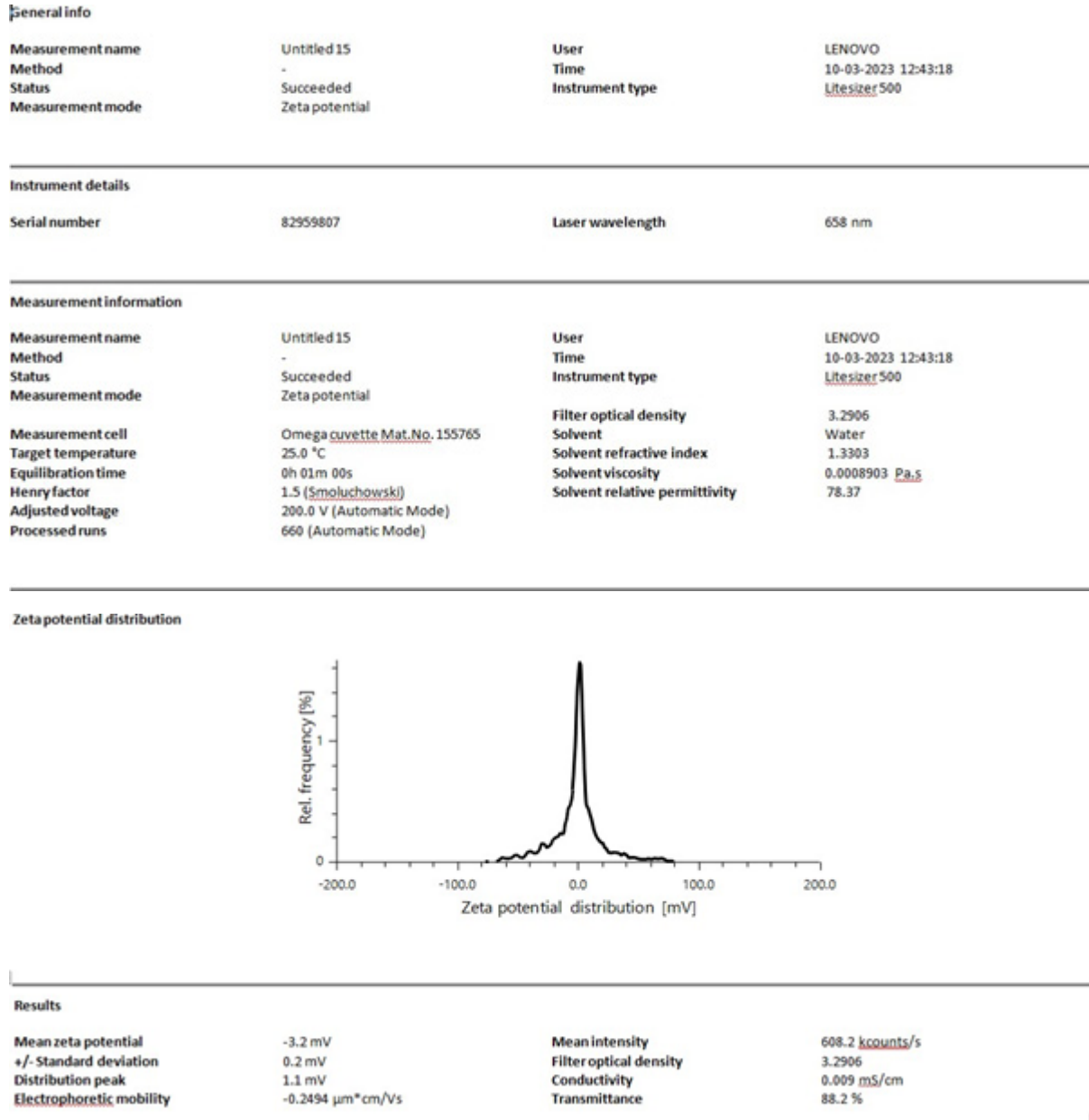


Figure 2: Result of Zeta potential.

Drug entrapment efficiency

From the results, it has been observed that the formulation containing high lipid concentration has a high entrapment as compared to other formulations. The prepared formulation of SLN-loaded sunscreen has 70.76% entrapment.

Preparation of medium

SPF determination

As The Mansur equation specifies the sun protection factor range with the help of absorbance value. The absorbance obtained for the formulation of sunscreen was found to be 18.1. The calculation is as follows

$$\begin{aligned}
 \text{SPF} &= \text{CF} \times \sum (\lambda) \times I(\lambda) \times \text{Abs} ((\lambda)) \\
 &= 10 \times 1 \times 1.81 \\
 &= 18.1
 \end{aligned}$$

DISCUSSION

The particle size, polydispersity index and zeta potential of solid lipid nanoparticles show that the particle size decreases as the concentration of avobenzone decreases. The polydispersity index was found to be 22.7%. The zeta potential was found to be -3.2 mV.

Table 2: Preparation of Medium using Cetrimide Agar.

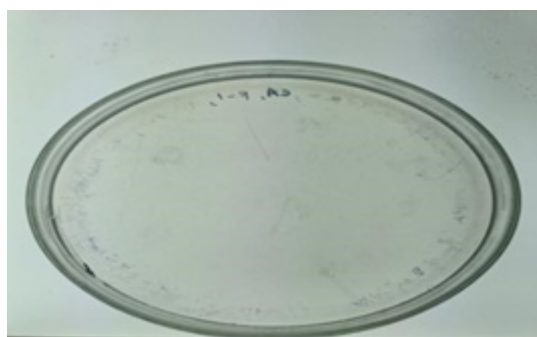
Formulation	Procedure	Observation	Inference
1.	Prepare a Cetrimide Agar plate and spread the inoculation broth to Agar medium. (By spread plate method).	No colonies are formed.	No growth was reported

Microbial Studies

Preparation of Broth using soya bean casein digest

Table 3: Preparation of Broth using soya bean casein digest.

Formulation	Procedure	Observation	Inference
1.	10 mL of soya bean casein digest+10 mL of prepared cream were autoclaved for 20 min at 121°C at 15 psi of pressure.	No microbial turbidity formed.	No Turbidity

**Figure 3:** Result of Microbial studies using cetrimide agar.**Table 4: Calibration curve for Photostability studies.**

Sl. No.	Concentration ($\mu\text{g/mL}$)	Absorbance (nm)
1	0	0
2	20	4.000
3	40	6.53
4	60	10.912
5	80	14.103
6	100	17.521

**Figure 4:** Result of microbial studies using soya bean casein digest.

From the results, it has been observed that the high lipid concentration in formulation have high entrapment as compared to other formulation. The prepared formulation of SLN-loaded sunscreen has 70.76% entrapment.

The calibration curve was plotted and a linear line was obtained with an $r=0.9972$. After performing the photostability test with the help of a UV spectrophotometer, the absorbance increases

with an increase in concentration of sunscreen. This indicates that the prepared sunscreen formulation has good photostability. As the ideal value of the SPF range is found to be 15, the formulation has an excellent sun protection range with the SPF of 18.1.

Microbial studies shows that this formulation is free from the microbial contamination and does not promote the growth of microbes like *Pseudomonas aeruginosa* and *Staphylococcus*

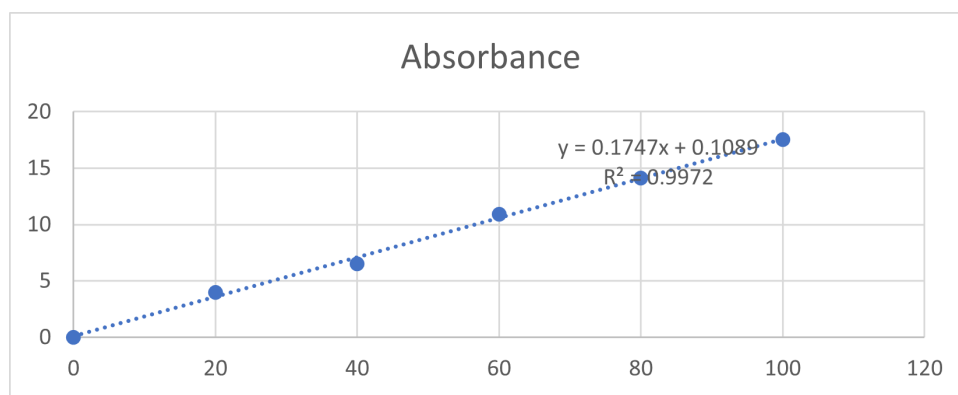


Figure 5: Calibration curve for Photostability studies.

aureus, as confirmed with cetrimide agar for fungal growth like Yeast and Mold.

The *in vitro* investigations carried out for this study confirm that sunscreen has a synergistic impact and that SLN blocks UV rays. Strong UV blocking, avobenzone can absorb UV light between 320 and 400 nm in wavelength. Therefore, it may be possible to manufacture new sunscreen products by lowering their concentration. Reduction of skin side effects can be achieved by firmly integrating organic UV-blockers into the solid SLN matrix. An enhanced carrier system for potentially hazardous sunscreens may be built around SLN.

CONCLUSION

We can conclude that there is a large market for sunscreen, whether it is synthetic, natural, or a combination of the two, because people are aware of the need for UVA and UVB protection as well as photostable sunscreen for consistent UVA/UVB protection. These chemical absorbers that have been loaded with SLN may offer a unique sunscreen that is safe, affordable and photostable while also having many more skin-protecting benefits. This study concludes that the SLN-loaded avobenzone sunscreen presents a promising approach for enhanced UV protection. It highlights the potential for further exploration of SLN technology in developing effective sunscreen formulations with reduced side effects, paving the way for safer and more efficient sun protection products.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

ABBREVIATIONS

SLN: Solid lipid nanoparticles, **UV:** Ultra violet, **SPF:** Sun protection factor, **PDI:** Poly dispersity index, **ACN:** Acetonitrile.

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