

Creating and Evaluating Herbal Anti-Ageing Cream Contains Pomegranate Seed Oil

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Abstract:-

➤ Introduction:

The skin is the most important exposed part of the body, is crucial in establishing social bonds and defending against environmental harm. In the present scenario to look attractive is a primary concern for the growing population in order to maintain social integrity and well being. However cell death is a natural process but due to highly stressful environmental conditions now a day's skin aging is a primary concern for dermatologists. In the present research work we aim to develop a novel anti-ageing cream for the prevention and treatment of unnatural skin ageing by utilizing pomegranate seed oil.

➤ Method:

The pomegranate seed oil was purchased from Amarnath export Bangalore Karnataka. The preparation of anti-aging cream was accomplished through conventional method by taking pomegranate seed oil as an oily phase and stearic acid as an emulsifying agent. The formulation was optimized by varying the concentration of aqueous and oily phase.

➤ Result and Discussion:

The pH of the developed cream was found to be 7.2 ± 1.2 and viscosity was found to be 5639 ± 2.51 cps. Furthermore the spreadibility of the developed cream was found to be 26.6 ± 0.2 gm*cm/sec. The developed formulation showed no skin irritation or erythema and also it was easily washable from skin. The stability study of the formulation was studied for 3 month at room temperature (25°C) which suggests high stability as no indication of cracking, phase separation or any other visual change was observed. Furthermore the developed formulation was analyzed through in vivo animal model (Albino wistar rat) which suggest high biocompatibility and potential in the management of skin ageing.

➤ Conclusion:

In conclusion we have developed a novel pomegranate seed oil based anti-aging cream for the prevention and treatment of unnatural skin ageing caused by the vulnerable environmental conditions. The developed formulation was analyzed through extensive in vitro and in vivo characterization which proves the potential of the developed cream in the management of unnatural skin ageing.

Keywords:- Cosmetology, Pomegranate Seeds Oil, Skin Ageing, Anti-Aging Cream, Animal Model.



Fig (A): Phase of Oil



Fig (B): Phase of Water



Fig (C): Combining the Phase of Oil and Phase of Water



Fig (D): Creamy Structure



Fig (E): Continue String



Fig (F): Cream formulation

Fig 1: Graphical Abstract of Cream Formulation

I. INTRODUCTION

The skin regeneration is a continuously process of a human life cycle and after an age of 30-39 years the skin starts aging, which is a natural process. However due to highly vulnerable external environments like air pollutants, water pollutants and light skin starts aging in early age. To deal with this condition various types of chemical based anti-aging skin creams was developed by different research groups and also available in the market. These chemical based creams are able to control the skin ageing problem but due to the side effects caused by these chemicals can also caused severe skin damages after long term use. To prevent these side effect various creams based on natural extracts like aloe Vera, turmeric, tulsi etc. are commonly utilized for the management of this problem, however creams based on these extracts are not able to prevent and at the same time treat the skin aging problem. To deal with this issue in the present research work we have developed novel oil from pomegranate seeds based cream for treating and preventing of unnatural skin aging.

Continuous UV exposure can cause various changes that are classified as photo-induced damages, including vascular homogeneities and pigmentation loss, elasticity loss in the skin, and texture deterioration (elastosis, hyperkeratosis, and yellowing).¹

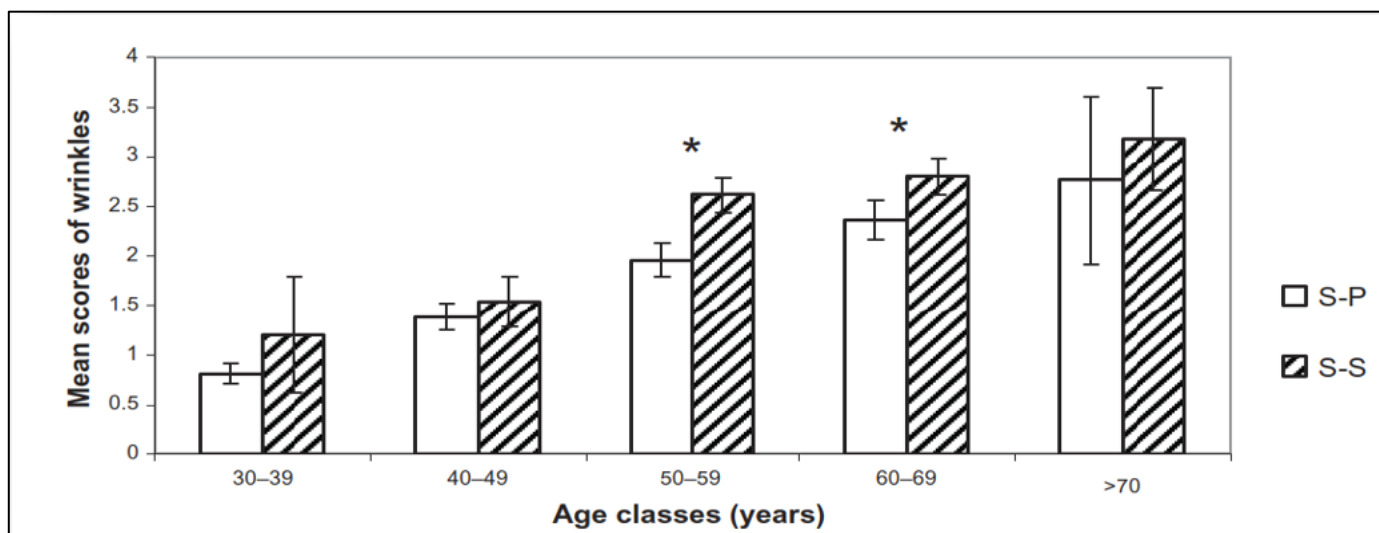


Fig 2: UV Exposure of Age Classes
S-P Means Sun Phobic and S-S Means Sun Seeking

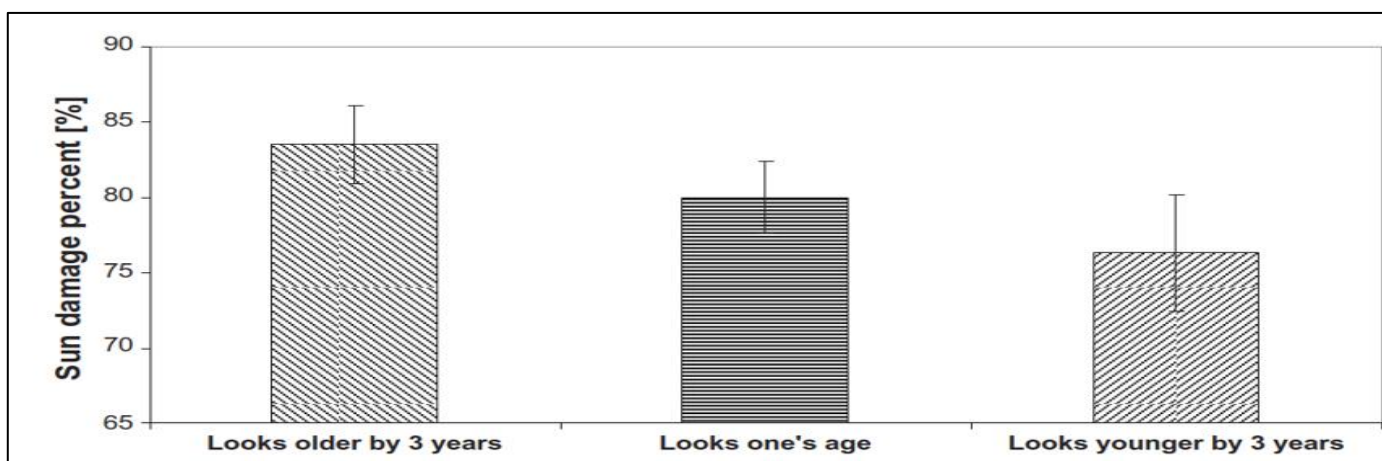


Fig 3: Skin Characteristic According to Demographic Reasons

From the Above figure-10 it shows that in Australian women have the higher number of aging as compared to USA, UK and Canada. The reason of aging is due to the UV exposure, it happens due to geographical location of the Australia is southern hemisphere and northern tip of Australia is closer to equator region due to which it is higher prone to direct exposure to sun and the biggest disparities were observed between women in the US and Australia. Compared to those from the USA, the Austrian women reported some indications of accelerated ageing about 20 years earlier.^{2,3}

The World Health Organization (WHO) and our nation have been supporting traditional medicine because it is more affordable, widely accessible, and thorough—particularly in poor countries. Additionally, it is accurate because 8% represent the world’s population receive their primary medical care from medicinal plants. The developed world, along with its citizens, acknowledged the benefits of conventional therapy as well as the defined protocols, rules, and treatment specifications for cultural medicine.^{4,5}

One of the first edible fruits, the (Punica granatum L.), the pomegranate, was an individual of the Punicaceae species, and has been utilized widely in folk medicine across many civilizations. This fruit is indigenous to Iran, where it is produced extensively along with India, the United States, and other near and far eastern countries. An estimated 1,500,000 tons of this crop are produced globally, with 47% of that amount coming from Iran, which has the largest area under cultivation.⁶

Pomegranates are abundant in lipids for example oleic the acids, stearic the acids, and palmitic the acids in addition to polyunsaturated fatty acids like linoleic and linolenic acid.⁷

PSO accounts about 12–20% of the overall weight of seeds. About 80% of the contents of seeds are made up of conjugated octadecatrienoic fatty acids, and these fatty acids, particularly punicic acid (cis 9, Trans 11, cis 13 acid) which represents the primary fatty acid among them. Pomegranate juice, peels, leaves, and flowers all have strong antioxidant qualities; however the juice, peel, and oil all have mildly estrogenic effects. Pomegranate seeds contain a high amount of conjugated α -linolenic acids (CLn) and ethno medical

indications. Numerous pharmacological effects are associated with punicic acid (PA), is highly concentrate in pomegranate seed oil (PSO), a conjugated isoenolenic acid isomer. Among its main qualities are antimicrobial, anti-inflammatory, kidney-protective, liver-protective, neutroprotective, anti-cancer, systemic immune repairing, improved carbohydrate metabolism, and decreased insulin resistance.⁸

II. MATERIALS AND TECHNIQUE

A. Products

Pomegranate seeds oil was purchased from Amarnath export Bangalore Karnataka Mili-Q water was used in all sets of experiments. Aloe Vera and turmeric were purchased from the local market and all other chemical are of analytical grade.

B. Preparation of Cream

The formulation of cream was prepared and optimized as given in Table-1. The oily phase (stearic acid, cetyl alcohol, glycerine, petroleum jelly, methyl paraben, and pomegranate seeds oil) was taken in a borosilicate glass breaker at and heated at 75°C. In other beaker aqueous phase (Mili-Q water, potassium hydroxide and methyl paraben) was also heated at 75°C. When the aqueous phase are completely solubilise it is slowly poured in heated oily phase with continuous stirring, after 2 hrs. The mixture was turned into a cream. After the cream formation the heating was stopped. The prepared cream was further smoothed by applying slap technique.

Table 1: Formulations of Anti Ageing Cream

| S. Number | Components | Formula 1 | Formula 2 | Formula 3 | Formula 4 | Formula 5 | Formula 6 |
|-----------|---------------------|-----------|-----------|-----------|-----------|-----------|-----------|
| 1. | PSO | 2ml | 1ml | 3ml | 5ml | 7ml | 8ml |
| 2. | Aloe Vera extract | 1ml | 1ml | 3ml | 3ml | 3ml | 2ml |
| 3. | Curcuma content | 1ml | 1ml | 1ml | 1.5ml | 1.8ml | 1ml |
| 4. | Vitamin E | 1ml | 1ml | 1ml | 2ml | 2ml | 1ml |
| 5. | Stearic acid | 2gm | 2gm | 0.5gm | 10gm | 15gm | 12gm |
| 6. | Cetyl alcohol | 1gm | 1gm | 1gm | 5gm | 5gm | 3gm |
| 7. | Glycerine | 1ml | 1ml | 1gm | 5ml | 5ml | 3ml |
| 8. | Petroleum jelly | 1gm | 1gm | 1gm | 5gm | 5gm | 3gm |
| 9. | Methyl paraben | 0.5mg | 0.5gm | 0.5gm | 2.5gm | 2.5gm | 2gm |
| 10. | Potassium hydroxide | 0.5gm | 1gm | 0.5gm | 2.5gm | 2.5gm | 2gm |
| 11. | Distilled water | Q.S | Q.S. | Q.S. | Q.S. | Q.S. | Q.S. |
| 12. | Rose water | Q.S. | Q.S. | Q.S. | Q.S. | Q.S. | Q.S. |



Fig 4: Images Formulated Creams

➤ Evaluation Parameters of Anti-Ageing Cream:

Various evaluation methods are used to test the efficacy of anti-ageing creams using pomegranate oil. One of the most common methods is the use of in vitro assays. In vitro assays involve testing the cream's efficacy in a laboratory setting. For example, the cream can be tested for its ability to reduce oxidative stress in utilizing the DPPH (diphenyl 1-picrylhydrazyl test on skin cells. Additionally, the cream can be put to the test to determine its capacity to increase skin cells' synthesis of collagen by the use the hydroxyproline assay.⁹

Another evaluation method that can be used is the in vivo method. In vivo studies involve testing the cream's efficacy on human subjects. For example, the cream can be tested for its ability assist in decreasing the visibility of creases and fine lines using digital imaging techniques. Additionally, the cream can be tested for its ability to increase skin hydration levels using skin hydration measurements.

➤ Organoleptic Study:^{10,11,12}

Organoleptic study for herbal formulation is a sensory analysis that evaluates the sensory characteristics of a product using the human senses of sight, smell, touch. The Organoleptic evaluation of an anti-aging cream of pomegranate oil should be conducted using a scientific

approach, including evaluating its appearance, odor, pH, viscosity, spreadability, homogeneity, skin feel, removal, irritancy test, and stability. These tests will help ensure that the product is of high quality, safe, and effective for its intended use.¹⁰

In the case of an anti-aging cream of pomegranate oil, the following organoleptic evaluation can be conducted in a scientific manner:

- **Appearance:** Evaluate the cream's color, clarity, and texture. The color should be uniform and consistent, while the texture should be smooth and free of lumps or grittiness. Odor: Evaluate the cream's odor for its intensity, character, and stability. The odor should be pleasant and consistent throughout the product's shelf life.

- **After feel:** We measured the amount of emollient, slipperiness, and thickness residue that remained after applying a specific amount of cream.
- **Removal:** It was discovered how quickly the cream could be removed by running tap water over the area where it was applied.
- **Irritancy test:** The Cream was applied to the left hand's specific area, and the time was recorded. Oedema, skin redness, and irritability were monitored up to a 24-hour period at regular intervals.
- **Grittiness:** On a glass slide, a tiny bit of cream was put, and the surface was then illuminated to look for any foreign particles.

Table 2: Organoleptic Activity

| S. No. | Specification | Limits |
|--------|---------------|------------------------|
| 1 | State | Semisolid |
| 2 | Color | Yellowish white, white |
| 3 | Odor | Characteristic |
| 4 | Texture | Smooth |

III. RESULTS AND DISCUSSION

- **pH:** Using a pH meter, find the cream's pH. The pH meter was calibrated using standard buffer solution as the calibration solution. A pH test was conducted on 0.5

grams of measured cream which was already mixed with 20.0 ml of purified water.¹¹ the pH should be within the acceptable range for the product, typically between 4.5 and 7.2. The formulation's pH was tested at 1, 10, 30, 60 and 90 days subsequently preparation. The findings of this study are presented in Table 3.

Table 3: pH Observation for Various Time Periods

| Temperature | Initial pH | pH after 1 month | pH after 2 month | pH after 3 month |
|-------------|-------------|------------------|------------------|------------------|
| 37±1 °C | 6.80 ± 0.01 | 6.50 ± 0.04 | 6.30 ± 0.03 | 7.20 ± 0.02 |

- **Finding a Particular Type of Cream:** In this experiment, Water or oil was used for diluting the cream's consistency, the particle dispersion agent is water, when the cream is dissolved using water and remains stable, it is an o/w type cream of cream. However, if the Because oil and water are incompatible, when cream is mixed with oil, the cream will shatter.¹³ within the present study, 2.5 grammes of cream were taken, and the cream was gradually diluted with distilled water.

- **Viscosity:** A Brookfield instrument DV-II + Pro (Brookfield Engineering Laboratories) was applied to determine the cream's thickness, with cylinder spindle #64, Test samples were collected in 250ml beakers that were clean and dry, and the viscosity of each sample was assessed using the viscometer's regular operating methods. The reading was recording at 100% torque. Sample temperatures were 37±1°C. We read the value in centipoises. The viscosities of formulated anti-ageing creams was found in the range of 3260 to 6499 Cp and it was closure to the standard marketed cream, it indicate that sufficient amount of oil phase and aqueous phase was used during formulation of cream. The results are given in table number 4.
- **Spreadability Study of Cream:** A glass device put up in the lab was used to assess the Spreadability between the greatest formulations. The pair of transparent slides was placed on top of the cream emulsion. Next, a 500 g mass was added to the slides for 1 minute to compress the sample and create a consistent thickness. Extra cream was then scraped off. Spreadability is calculated using the formula:



Fig 5: Dilution Test

- Formula of Spreadability:** $S = M \times L / T$ Where, M is the weight that has been put L is the glass slide's measurement, T is the amount of a few seconds, and such on to the highest slide.¹⁴ The Spreadability was found to range about 21 and 30 gm.cm/sec. it was found to be within the Spreadability range of commercial cream, The cream's formulation made it simple to distribute without creating a lot of friction, the results are given in table number 4.
- Microstructure Observation:** An optical microscope with a 40x magnification was used to analyse the morphology of prepared cream. Before the microscopic examination, a cover slip was placed over the 1gm of cream samples, which had been smeared gently over the slide. The slide was placed on a microscope slide. Photomicrography picture of the emulsion was captured and it was evident that oil globules were present on the

continuous water phase, which proves that the anti ageing (Pomegranate seeds oil) cream was properly manufactured.

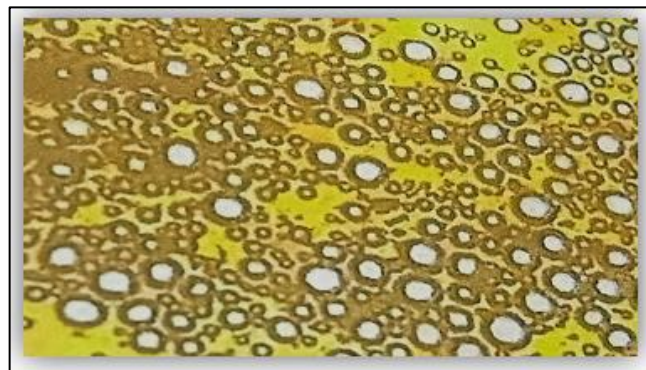


Fig 6: Microscopic Structure of Cream

Table 4: Cream Formulation and Their Results

| Formulations | pH | Thickness (Centipoises) | Spreading Capacity (gm*cm/sec) | Perceptiveness | Erythema | Phase Separation | Cleanable |
|--------------|-----------|-------------------------|--------------------------------|----------------|----------|----------------------|-----------------|
| Formula1 | 6.80±1.02 | 7558 ± 1.02 | 21.6 | No irritation | Nil | Not phase separation | Easily washable |
| Formula2 | 6.00±2.03 | 8698 ± 1.00 | 22.5 | No irritation | Nil | Not phase separation | Easily washable |
| Formula3 | 6.50±1.33 | 6839 ± 1.22 | 20.8 | No irritation | Nil | Not phase separation | Easily washable |
| Formula4 | 7.2±1.2 | 5639 ± 2.51 | 26.6 | No irritation | Nil | Not phase separation | Easily washable |
| Formula5 | 6.86±1.02 | 4829 ± 1.33 | 25 | No irritation | Nil | Not phase separation | Easily washable |
| Formula6 | 7.00±1.02 | 4457 ± 1.15 | 24.1 | No irritation | Nil | Not phase separation | Easily washable |

- Stability Study of Formulation¹⁵:** Final formulations including active components were kept in plastic containers at 2 to 4°C, 25°C and 40°C. After week 4, all formulations physiochemical characteristics were assessed to determine their physical stability.

measurement used for evaluating the stability was determined based on organoleptic properties, pH, and viscosity. Periodically, at intervals of zero, one, two, and three months, samples of the pomegranate seeds oil cream were taken and every evaluation parameters was recorded. The durability study's findings are displayed in the table below 5.

The optimised formulation was stored in an airtight container at the specified temperature to gauge. The cream composed of the seeds of pomegranate oil's consistency. The

Table 5: Data of Stability Study

| Temperature | Homogeneity | Phase separation | Simple removal |
|------------------|---------------------|------------------|----------------|
| 2-4°C | Homogeneous | No | Remove quickly |
| Room temperature | Homogeneous | No | Remove quickly |
| 40°C | Slightly liquefying | No | Remove quickly |

Table 6: Accelerated Stability Study

| Months / Test | Anti-ageing cream | | |
|------------------------|-------------------|-----------------|-------------------|
| | Initial month | After two month | After three month |
| Physical features | Semi solid | Semi solid | Semi solid |
| Texture | Good | Good | Good |
| Odour | Characteristic | Characteristic | Characteristic |
| Thermal stability | Ok | Ok | Ok |
| Deterioration of goods | Nil | Nil | Nil |



Fig 7: Stability Study 1 Month 2 Month and 3 Month

IV. IN VIVO EVALUATION OF FORMULATED ANTI-AGEING CREAM

Both sexes of albino rats (weighing 150–180g) will be used in the studies. The animals must be kept in groups of six in polyacrylic cages measuring 38 by 23 by 10 cm, and they must be kept in a typical laboratory environment with a temperature of 25 °C, a dark/light cycle, and relative humidity between 60 and 70 %. They will consume a pellet diet along with water additives. The animals will become accustomed to the laboratory environment for seven days before the studies begin. Animal research was done at the pharmacology division. Truba institute of Pharmacy, Bhopal (M.P.) with the Institutional Animals Welfare Committee's approval as required (IAEC approval No. TIP/IAEC/02/2023), all the research was performed according to the animal ethics committee guidelines for the experimental animals.

A. Selection of Animal Model:¹⁶

There are six animals in each of the four groups into which the animals were divided.

- Group 1 will be designated as the standard control.
- Group 2 will serve as the negative control. (Ageing induced)
- Group-3 will be named as marketed control and ageing will be treated with marketed anti-ageing cream.
- Group-4 will be assigned as test groups that will treated with formulated cream
- **Procedure to Develop the Ageing:** All the animals were anesthetized by chloroform (5ml), after shaving; a hair removal cream was used to completely remove the hair, prepare the ageing induced assembly, and then it will keep under for exposing UV radiation (UVB radiation 320-290nm). This process will take place for 7-10 days, after this process wrinkle develop and the formulated cream

and marketed cream was applied regular to all animals for 30 days.



Fig 8: Induced Ageing

B. Evaluation Parameters¹⁷

- Area of Skin Wrinkles.
- Skin Irritation Study.
- Photographic Comparison.
- Skin Texture.
- Allergic Response.
- **Area of Skin Wrinkle-** The wrinkle reduced property was evaluated by measuring the area of wrinkle alternatively before applying the cream. The length of the ageing was measured using a scale and transparent paper by placing the paper on ageing and tracing it out on alternate day. Significant decrease in area of the ageing (around 0.33 mm in every 5th day) in the test group of animals as compare with the marketed and negative control groups animal, it indicates that formulated cream is effective for ageing treatment, the area of ageing in test group decreases enough to justify the effectiveness of the formulated cream of pomegranate seed oil.

Table 7: Area of Skin Wrinkle

| Animal Group | Zero Day | Day Five | Day Ten | Day Fifteen | Day Twentieth | Day Twenty-five | Day Thirty | Day Thirty-five |
|------------------|----------|----------|---------|-------------|---------------|-----------------|------------|-----------------|
| Negative Control | 1.4cm | 1.4cm | 1.5cm | 1.5cm | 1.3cm | 1.2cm | 1.0cm | 0.5cm |
| Standard | 1.4cm | 1.2cm | .7cm | 0.2cm | 0.1cm | - | - | - |
| Test | 1.4cm | 1.3cm | 0.7cm | 0.6cm | 0.2cm | 0.2cm | - | - |

• **Study on Skin Irritation:** Erythema, itching, oedema, pain, and sensitivity are among the symptoms examined. When conducting the study, these elements were considered. Four animals in all were used in the investigation. Dorsal skin hair was removed using hair removal cream and the formulated cream was applied for 10 days. On the basis of a scoring table, it was appraised. They are indicated by table number 8. The skin sensitivity

study’s final finding were noted and reported in table number 8.

Table 8: Skin Irritancy Scoring Table

| Score | Reaction |
|-------|----------|
| 0 | No |
| 1 | Slightly |
| 2 | Moderate |
| 3 | Severe |



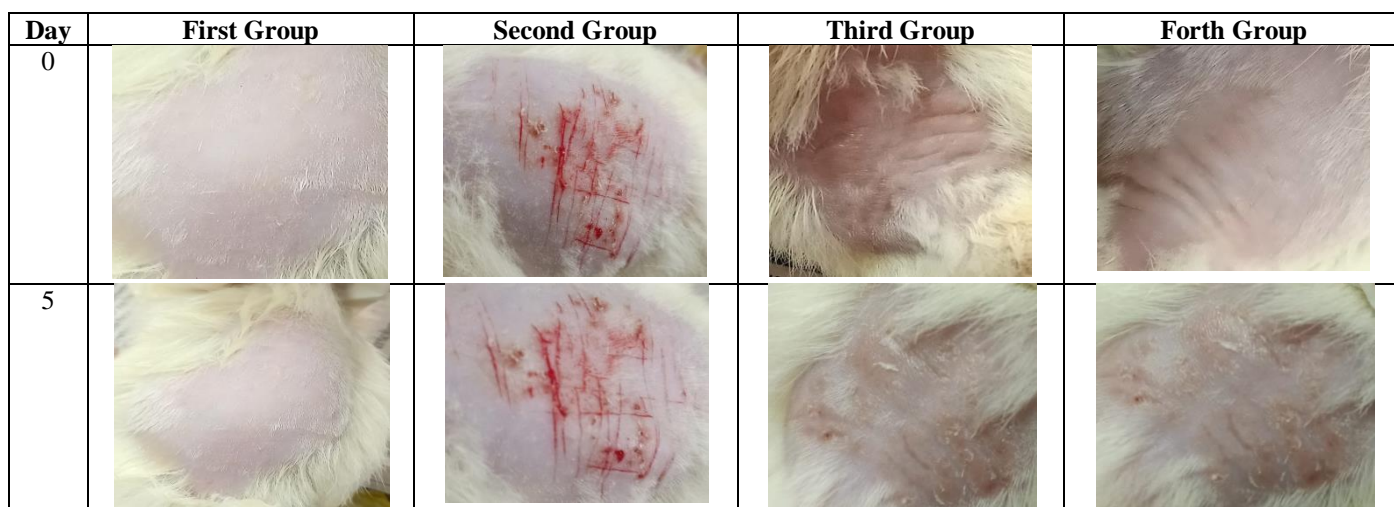
Fig 9: Animal were used to Evaluate Skin Irritancy (F5)

• **Allergic Response-** The symptoms allergic response includes rash, redness, swelling, cracked skin, and itching. When conducting the study, these elements were considered. Four animals in all were used in the investigation. Dorsal skin hair was removed using hair removal cream the formulated cream was applied on 1.5cm skin area for 10 days. On the basis of a scoring table, it was appraised. They are indicated by table number 8. The allergic response study’s final finding were noted and reported in table number 9.

Table 9: Allergic Response

| Score | Allergic symptoms | Reaction |
|-------|-------------------|----------|
| 0 | Rash | No |
| 1 | Redness | Slightly |
| 2 | Swelling | No |
| 3 | Cracked skin | No |
| 4 | Itching | Moderate |

• **Photographic Comparison:** After creating an ageing according to the guidelines of the CPCSEA, photographs of each group of animal’s ageing part were taken for the visual comparison, and the same is shown in figure 10: Following formulation applied to the ageing each group of animals.



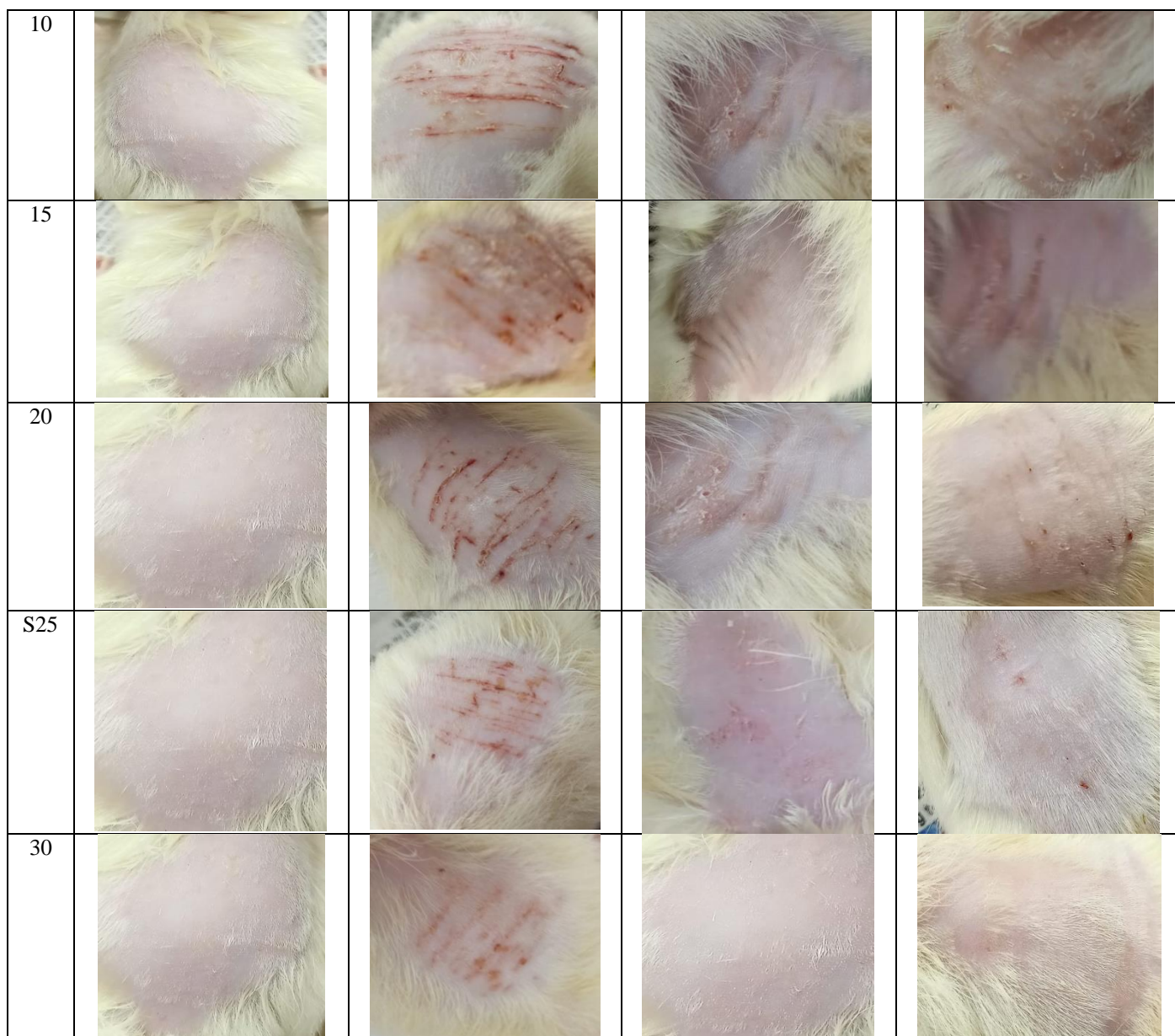


Fig 10: Top-View Images of Ageing on Days 0, 5, 10, 15, 20, 25, and 30 Where Group Two is the Opposing Control Group, Whereas Group 1 is the Controlled Group (Induced Ageing by UV Light after 10 Days we can Follow the Group3), Group 3 is the Marketing Group, and Group 4 is the Test Group

V. IN VIVO STUDY

➤ *Area of Skin Wrinkles-*

The wrinkle reduced property was evaluated by measuring the area of wrinkle alternatively before applying the cream. The length of the ageing was measured using a scale and transparent paper by placing the paper on ageing and tracing it out on alternate day. Significant decrease in area of the ageing (around 0.33 mm in every 5th day) in the test group of animals as compare with the marketed and negative control groups animal, it indicates that formulated cream is effective for ageing treatment, the area of ageing in test group decreases enough to justify the effectiveness of the formulated cream of pomegranate seed oil.

➤ *Study on Skin Irritation –*

The symptoms of rashes on skin studied includes Redness, itching, oedema, discomfort, and sensitivity. When conducting the study, these elements were considered. Four animals in all were used in the investigation. Dorsal skin hair was removed using hair removal cream and the formulated cream was applied for 10 days. On the basis of a scoring table, it was appraised. They are indicated by table number 13. The skin sensitivity study’s final finding were noted and reported in table number 8.

➤ *Allergic Response-*

The symptoms allergic response includes rash, redness, swelling, cracked skin, and itching. When conducting the study, these elements were considered. Four animals in all were used in the investigation. Dorsal skin hair was removed using hair removal cream the formulated cream was applied

on 1.5cm skin area for 10 days. On the basis of a scoring table, it was appraised. They are indicated by table number 15. The allergic response study's final findings were noted and reported in table number 15.

➤ *Photographic Observation and Comparison-*

After creating a ageing according to the guidelines of the CPCSEA, the images was captured of each animal groups for the visual comparison, and those image was attached in figure number. It was observed that anti-ageing formulated

cream of pomegranate seed oil showed nearly similar ageing healing as compared to the marketed anti-ageing cream. The ageing of test group animals marketed control group animals was healed completely on the 30th day, and the ageing of negative control group animals was not healed till the last day of study.

A comparative study of marketed and formulated anti-ageing cream, after performing all the evaluation parameters, the comparison are given in table number 10.

Table 10: Comparison between Marketed and Formulated Anti-Ageing Cream

| S. No. | Marketed Cream | Formulated Cream |
|--------|---|---|
| 1 | It is polyherbal formulation | It is single herb used |
| 2 | It is causes mild irritation after applying on skin | No irritation on skin |
| 3 | Need to apply 2-4 times a day | Two times a day |
| 4 | This is high cost cream | Low cost cream |
| 5 | Need at least 2 month for new ageing and takes 8 months to complete healing of old ageing | It is expected to take 2 month for new ageing |

VI. CONCLUSION

Our work's objective was to create, improve, and assess an anti-aging cream that included pomegranate seed oil, it includes ageing that are formed on our skin during the natural healing process of body cells. Ageing are developed in our body, during UV radiation, sunburn and environment pollution in our skin the immune system and natural healing process of our body activates immediately and send messages to form fresh collagen fibres and attack the infection. This results into the developments of ageing.

Vanishing cream of pomegranate seed oil was ready successfully using several substances, including potassium hydroxide, cetyl alcohol, and stearic acid, glycerine, perfume water and anti-microbial agents, The reason vanishing cream was chosen is that it left a thin coating on the skin that lets the cream stay on the skin longer, which ultimately results within significant reduction of ageing. In the study oil-in-water cream formulation was selected because They consist of tiny oil droplets scattered throughout a continuous water phase and the proposed cream formulation would be more comfortable and cosmetically for skin ageing as It is easier to remove with water and less oily.

The prepared cream formulation was analysed for different in-vitro and in-vivo parameters such as Organoleptic parameters, types of emulsion test, viscosity and Spreadibility for in vitro and area of skin wrinkles, skin irritation, allergic response, skin texture and photographic comparison for in-vivo evaluation and stability study was also performed. The range of 3260 to 6499 Cp was discovered to be the cream's viscosity after formulation. The Spreadibility was found within the range that of 21to 30gm.cm/sec, pH was recorded at 6.5 constantly for 3 months which shows the stability of the cream.

In the animal study, healthy albino rats (150-300g) were chosen to perform every parameters of in-vivo research. Study was performed at Truba institute of Pharmacy, Bhopal (M.P.) in division of pharmacology lab with appropriate

institutional authorization Animal Ethical Committee(IAEC approval no. PH/IAEC/2K23). An in-vivo study's findings showed that the ageing regulated by formulated pomegranate seed oil cream, recovery 100% with each better skin appearance and the ageing of test group was healed completely on the 30th Dy. It was observed that formulated cream of pomegranate seed oil showed nearly similar ageing healing as compared to the marketed anti-ageing cream.

After performing different in-vitro, in-vivo and stability study parameters, it was observed that pomegranate seed oil was effective as a cream formulation to treat the skin ageing, skin wrinkles, fine line, dark spots and it also significant effectiveness against sunburn. Results of the study suggest that the formulated anti-ageing cream of pomegranate seed oil and its oil was safe, stable and cost effective for topical cosmetic formulation.

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