Indo Global Journal of Pharmaceutical Sciences, 2022; 12: 209-215

INDO GLOBAL JOURNAL OF PHARMACEUTICAL SCIENCES
ISSN 2249- 1023

In vitro Evaluation of Physicochemical Properties of Polyherbal Anti-Wrinkle Cream

Usha Kiranmai G, Shayeda *

Department of Pharmaceutics, University College of Pharmaceutical Sciences, Kakatiya University, Warangal, Telangana, India – 506009

Address for Correspondence: Shayeda, shayeda_ucpsc@yahoo.com

Received: 04.08.2020
Accepted: 22.03.2022
Published: 08.08.2022
Keywords
Polyherbal; Anti-wrinkle; Creams; Stability Study.

ABSTRACT: Purpose: Skin ageing can be described as irregular pigmentation, increased wrinkling, loss of elasticity, dryness and roughness. Wrinkling of the skin is the most obvious sign of deterioration of the human body with age. Herbal cosmetics enhance the human appearance by playing a major role in impeding and reversing the ageing of the skin. Method: In this study, anti wrinkle creams were formulated with herbal oils of different concentrations namely AW1, AW2, AW3 and AW4. All cream formulations were evaluated for different parameters like pH, viscosity, spreadability, stability studies etc. The creams were found to be stable during stability studies done according to ICH guidelines. All in vitro tests results were analyzed statistically using SPSS software 17.0 version and compared with standard marketed formulations. Results: The formulated creams were consistent in quality and safe to be used on the skin. The Anti Wrinkle creams showed no evidence of phase separation and ease of removal. The creams were not shown any redness, edema, inflammation and irritation to the skin during skin irritation studies. The formulated creams were found statistical significant difference with that of standard at $p<0.05$. Conclusion: Formulations AW2 and AW4 showed good pH, viscosity, homogeneity, good spreadability, good extrudability with good appearance during the three months of stability studies.

Cite this article as: Usha, K.G.; Shayeda. In vitro evaluation of physicochemical properties of Polyherbal Anti-Wrinkle cream. Indo Global J. Pharm. Sci., 2022; 12: 209-215. **DOI**: http://doi.org/10.35652/IGJPS.2022.12026

INTRODUCTION

Skin ageing is a complex process and multifactorial i.e., structural, functional and aesthetic changes happen at a variable rate [1]. Intrinsic, chronological ageing reflects the passage of time from gravity and genetics. It is a slow process with clinical features such as smooth, pale, dry and less elastic skin having fine wrinkles that are not apparent until old age.

Extrinsic ageing is usually attributed to photoaging and smoking. Extrinsic aging, due to chronic exposure to solar ultraviolet irradiation (photoageing) and smoking, leads to deep and coarse wrinkles, mottled hyperpigmentation and reduced skin elasticity.

Cosmetic products are used to protect skin against exogenous and endogenous harmful agents and enhance the beauty and attractiveness of the skin. The synthetic or natural ingredients present in skincare formulation support the health, texture, integrity of skin, moisturizes and maintain the elasticity of the skin by reduction of collagen and photoprotection [2]. Thus, it helps to reduce the production of free radicals in the skin and manage the skin properties for a long time. Cosmetic products are the best choice to reduce skin disorders such as hyperpigmentation, skin aging, skin wrinkling and rough skin texture.

The present study aims at formulating and evaluating anti-wrinkle creams made of naturally available herbal oils which may not irritate and also gives nourishment to the skin.

MATERIALS AND METHODS

Materials

Avena oat oil, Rosewood oil, Lavender oil, Frankincense oil, Clove leaf oil, Almond oil, Olive oil, Rose oil, Carrot seed oil and Tulsi oil are the herbal oils used in the study obtained from Dr. Jain's Forest Herbals Pvt. Ltd. Stearic acid and cetyl alcohol (Himedia Labs. Pvt. Ltd., Mumbai). Triethanolamine,

glycerin and propylene glycol are from Merck company, Mumbai. Parabens were purchased from Sd. fine. Chem. Ltd, Mumbai. All other analytical grade solvents were used. Aloe vera juice and Tomato juice were freshly prepared and used [3, 4].

Methods

Preparation of Anti-Wrinkle Cream

All the oil-soluble ingredients were taken in one beaker and all aqueous soluble substances were added to another beaker. Both the phases were heated to 70 – 74°c. Then aqueous phase was added to the oily phase with continuous stirring till a homogeneous mixture was formed and composition is shown in **Table 1,1a** & **1b**.

Table 1: Composition of optimized Vanishing Cream base

Oil Phase	
Ingredients	**Concentration (%w/w)**
Stearic acid	10
Cetyl alcohol	2
Almond oil	0.75
Olive oil	0.75
Rose oil	0.75
Carrot seed oil	1.5
Tulsi oil	1
Propylparaben	0.05
Aqueous Phase	
Triethanolamine	3
Glycerine	5
Propylene glycol	3
Methylparaben	0.2
Purified water	Up to 100ml

Evaluation

The physical appearance of the formulations

Organoleptic test

The prepared formulations were observed for their color and odor.

Homogeneity

By visual appearance and by touch, the prepared creams were tested for homogeneity

Table 1a: Composition of herbal oils used in various anti-wrinkle creams

Herbal oils(%w/w)	Creams codes			
	AW1	**AW2**	**AW3**	**AW4**
Avena Oat oil	0.25	0.5	1	0.75
Rosewood oil	0.75	0.25	0.5	1
Lavender oil	1	0.5	0.25	0.75
Frankincense oil	0.5	1	0.75	0.25
Clove leaf oil	0.5	0.5	0.5	0.5

Table 1b: Composition of juices used in various anti-wrinkle creams

Juices (freshly prepared)	Creams			
	AW1(ml)	**AW2(ml)**	**AW3(ml)**	**AW4(ml)**
Aloe vera juice	5	10	20	30
Tomato juice	2.5	5	7.5	10

After feel

After the application of a fixed amount of creams, emolliency, slipperiness and amount of residue left were checked.

Removal

The ease of removal of the cream applied was examined by washing the applied part with tap water [5].

pH measurement

0.5g of the weighed formulation was taken in a beaker and dispersed in 50 ml of distilled water. The pH was measured using a Digital pH meter in triplicates [6]**.**

Viscosity measurement

Viscosity measurement of the formulations was done using a rotational viscometer (Brookfield DVIII + Rheometer/Model: LV, with spindle no – 52). The developed formulations were poured into the adaptor of the viscometer and the viscosity was measured at a speed of 50 rpm, 25 ± 1°c. Measurements were taken in triplicates [7]**.**

Spreadability

Excess amount of sample was applied in between two glass slides and was compressed to uniform thickness by placing 1000 gm weight for 5 minutes on upper slide. Weight was added to the pan on other end and the time required to separate the two slides, *i.e*., the time in which the upper glass slide moves over the lower glass slide was taken as measure of spreadability (gm-cm/sec) [8**]**. The lesser the time taken for separation of two slides, the better is the spreadability. The

method was repeated in triplicates. The following equation was used for this purpose:

$$S = m*l/t$$

Where,

m= weight tide to upper slide; l= length moved on glass slide; t=time taken

Extrudability

The apparatus consists of a wooden block inclined at an angle of 45º fitted with a thin, long metal strip (tin) at one end, while the other end was free. The aluminium tube containing 20 gms of cream was positioned on an inclined surface of a wooden block. 1 kg weight was placed on a free end of the aluminium strip and was touched for 30 sec. The quantity of cream extruded from each tube was noted [9]. Then, the percentage of creams extruded was calculated. The test results were taken in triplicates.

Performance and drying test

It was performed for about 48 hr after the preparation of formulations. Samples are taken on a glass slide using a brush forming a uniform thick layer of about 1 mm. Then the slide was placed in an oven at 36.5 ± 2.0 ºc for 1 hr simulating the body temperature. The formulations were monitored for up to 10 min until the drying process was completed and allowed the complete removal of the film from the glass slide [10].

Centrifugation test

All the formulations immediately after preparation were subjected to centrifugation test at 24 hr, 7 days, 14 days and 30 days at 25 ºc at 5000 rpm for 10 min by placing the 5 gm sample in stopper centrifugal tubes [11].

Primary Skin Irritation Studies

Primary skin irritation studies of the formulations were performed using albino rabbits. Rabbits were kept in cages and supplied with fresh food and water during the test period, 24 hrs before the test. Hair was removed from the neck and thigh region and cleaned with surgical spirit to expose sufficient test area. The creams were applied to the test area and observed for erythema and edema for 24 hrs, 48 hrs and 72 hrs [12]. Institutional animal ethical committee approval has been taken to conduct the study (IAEC/21/UCPSc/KU/2016).

Stability Study

All the prepared creams were subjected to stability studies at different temperature conditions i.e., 25^0c (room temperature), 4^0c (refrigerator) & 40^0c (stability chambers) with 75% relative humidity (RH) and the parameters like pH, viscosity, spreadability and extrudability were measured for three months as per ICH guidelines [13].

Statistical Analysis

The evaluation of data was done using SPSS software (version 17.0). One-way ANOVA followed by Dunnet's test was used for data expressed as mean ± standard deviations. For all comparisons, a value of P < 0 .05 was considered significant.

RESULTS AND DISCUSSION

The physical appearance of the formulations

Organoleptic parameters like color – there is a little change for the formulations AW1 and AW3. No phase separation was observed for all creams. All the data was given in **Table 2**.

Table 2: Physical appearance of the formulations

Formulation code	Color	Odor	Centrifugation (Phase Separation)	After feel	Removal
AW1	Little Change	Pleasant	No	Good	Easily removed
AW2	Creamy white	Pleasant	No	Good	Easily removed
AW3	Little Change	Pleasant	No	Good	Easily removed
AW4	Creamy white	Pleasant	No	Good	Easily removed

Table 3: pH at different temperature conditions

Formulation code	Temp. (ºc)	0 day	1st month	2nd month	3rd month
AW1 (Avg±SD)	25	6.61±0.24	6.51±0.18	6.51±0.18	6.51±0.18
	4	6.95±0.14	7.06±0.05	7.06±0.05	7.06±0.05
	40	6.73±0.08	6.73±0.08	6.73±0.08	6.73±0.08
AW2 (Avg±SD)	**25**	**6.31±0.07**	**6.5±0.28**	**6.7±0.1**	**6.31±0.34**
	4	**6.23±0.11**	**6.40±0.12**	**6.9±0.2**	**6.23±0.98**
	40	**6.63±0.08**	**6.34±0.31**	**6.5±0.01**	**6.17±0.59**
AW3 (Avg±SD)	25	6.07±0.98	5.95±0.83	6.93±0.15	6.07±0.05
	4	6.01±0.21	6.26±0.27	6.45±0.12	6.26±0.27
	40	6.64±0.06	6.37±0.20	6.67±0.02	6.16±0.65
AW4 (Avg±SD)	**25**	**6.15±0.15**	**6.37±0.29**	**6.47±0.06**	**6.45±0.06**
	4	**6.51±0.47**	**6.63±0.19**	**6.61±0.08**	**6.44±0.05**
	40	**6.71±0.02**	**6.44±0.30**	**6.61±0.01**	**6.21±0.2**
Himalaya Anti Wrinkle cream (Avg±SD)	25	6.43±0.01	6.43±0.01	6.43±0.01	6.43±0.01
	4	6.42±0.1	6.43±0.01	6.42±0.01	6.34±0.01
	40	6.41±0.01	6.40±0.01	6.43±0.01	6.43±0.01

Homogeneity

All formulations showed uniform distribution of ingredients in the cream. By visual appearance and touch, it was confirmed.

pH measurement

The pH of the formulations was found to be between 6.01-7.20 in three months stability studies and was nearer to skin pH. The results were expressed as mean ± standard deviation and were shown in **Table 3**.

Viscosity

All the prepared formulations were tested for viscosity for three months to observe their stability by comparing them with the standard. The data were presented in **Table 4** and expressed in mpas (SI system). The formulations AW2 and AW4 maintained their viscosity in three months period than the other two formulations and were found to be stable.

Spreadability

When formulations were subjected to spreadability studies for three months, it was found that the AW2 and AW4 creams took less time to spread than AW1 and AW3 when compared with the standard. The results were expressed in gm-cm/sec and shown in **Table 5**.

Extrudability

All the prepared formulations were subjected to extrudability and compared with the standard. The data of extrudability was shown in **Table 6**. Among all the formulations, AW2 and AW4 showed percentage extrudability between 65%-98% respectively.

Performance and drying test

All the formulations showed good performance on the *in-vitro* drying time test, which means that they formed a resistant film on the glass slides at a maximum of 20 min which were easily removed and among them, AW2 and AW4 took a lesser time of 17 min for drying.

Primary Skin Irritation Studies

None of the prepared creams showed any erythema or edema, indicating that the prepared formulations were non-irritant on the skin of animals and shown in **Fig. 1**.

Table 4: Viscosity at different temperature conditions

Formulation code	Temp. (^{o}c)	0 day	1st month	2nd month	3rd month
AW1 (Avg±SD)	25	6.19±1.10	6.10±2.92	14.20±0.62	9.29±1.83
	4	19.33±1.90	19.40±5.03	25.77±1.89	14.59±0.33
	40	16.66±3.45	15.89±3.52	7.37±1.28	16.2±2.48
AW2 (Avg±SD)	**25**	**27.57±2.42**	**26.85±1.50**	**24.23±8.88**	**22.87±4.27**
	4	**25.8±1.88**	**25.71±5.18**	**28.03±1.19**	**29.72±6.10**
	40	**26.22±2.42**	**26.21±2.75**	**26.48±3.63**	**21.37±2.18**
AW3 (Avg±SD)	25	18.29±5.55	17.04±7.19	15.98±0.88	24.2±10.35
	4	18.06±4.73	15.45±0.90	16.35±2.44	19.65±5.33
	40	18.66±1.24	21.70±6.99	19.61±0.06	19.3±5.10
AW4 (Avg±SD)	**25**	**24.4±5.74**	**27.60±1.32**	**20.24±8.34**	**19.64±4.63**
	4	**26.94±6.98**	**24.22±1.25**	**24.22±4.23**	**21.34±3.36**
	40	**23.28±5.30**	**26.01±0.95**	**21.39±4.74**	**20.91±4.09**
Himalaya Anti Wrinkle cream (Avg±SD)	25	26.12±2.2	25.86±0.92	23.13±2.34	20.23±2.23
	4	25.23±1.02	21.71±1.85	2.83±1.91	24.45±3.23
	40	26.22±2.42	22.16±2.57	24.18±1.23	21.23±1.28

Table 5: Spreadability at different temperature conditions

Formulation code	Temperature (ºC)	0 day	1st month	2nd month	3rd month
AW1 (Avg±SD)	25	18.16±1.74	16.63±8.78	13.22±1.87	9.77±0.87
	4	9.57±2.76	9.57±2.76	11.44±3.86	10.90±4.78
	40	10.35±1.44	10.35±1.44	13.44±1.65	14.36±3.25
AW2 (Avg±SD)	**25**	**11.16±1.80**	**6.90±0.42**	**10.66±6.36**	**6.46±0.25**
	4	**6.29±0.74**	**6.40±0.12**	**6.88±2.03**	**6.05±0.81**
	40	**10.29±3.06**	**8.19±2.23**	**12.99±6.06**	**6.29±0.74**
AW3 (Avg±SD)	25	15.42±3.15	11.45±4.73	9.11±6.43	9.59±3.02
	4	8.82±1.95	16.22±0.28	6.99±1.85	11.98±1.27
	40	13.12±3.77	11.54±8.63	6.66±2.84	13.71±1.0
AW4 (Avg±SD)	**25**	**11.46±0.31**	**6.58±0.41**	**8.83±0.69**	**6.47±o0.34**
	4	**11.66±3.41**	**5.84±1.18**	**6.61±3**	**6.83±0.17**
	40	**10.56±0.95**	**6.38±0.35**	**13.44±5.31**	**6.12±0.02**
Himalaya Anti Wrinkle cream (Avg±SD)	25	10.16±1.74	6.96±1.47	10.16±1.74	6.34±2.12
	4	7.16±2.34	7.00±1.70	6.10±1.23	6.57±0.12
	40	10.16±1.74	7.36±0.23	10.16±3.41	5.98±0.98

Table 6: Percentage extrudability at different temperature conditions

Formulation code	Temperature (ºC)	0 day	1st month	2nd month	3rd month
AW1 (Avg±SD)	25	87.67±7.78	63.52±3.55	31.44±1.34	61.07±14.98
	4	94.75±6.94	60.51±20.20	70.67±6.55	64.2±4.86
	40	79.93±10.45	62.34±7.68	68.43±0.09	64.53±4.11
AW2 (Avg±SD)	**25**	**89.57±3.06**	**91.59±0.05**	**77.46±7.04**	**80.23±6.20**
	4	**96.80±1.40**	**75.09±0.22**	**78.51±0.05**	**71.95±7.54**
	40	**86.43±4.28**	**68.87±0.05**	**71.67±6.28**	**71.52±5.56**
AW3 (Avg±SD)	25	83.47±2.02	72.49±1.17	21.38±1.32	73.58±8.18
	4	71.64±7.38	73.77±7.38	55.82±0.09	66.38±4.10

	40	82.74±6.41	60.51±13.47	71±0.89	68.12±5.23
AW4 (Avg±SD)	**25**	**97.59±5.74**	**74.99±11.30**	**56.74±0.14**	**74.06±2.51**
	4	**96.12±0.12**	**67.82±3.55**	**94.02±0.09**	**79.1±8.77**
	40	**82.74±7.83**	**74.73±4.48**	**74.32±0.32**	**69.49±7.70**
Himalaya Anti Wrinkle cream (Avg±SD)	25	86.57±0.02	82.49±0.17	71.38±1.32	73.58±8.18
	4	92.64±3.78	73.87±6.38	82.55±0.09	66.38±4.10
	40	82.00±6.23	66.12±10.12	71±0.89	68.12±5.23

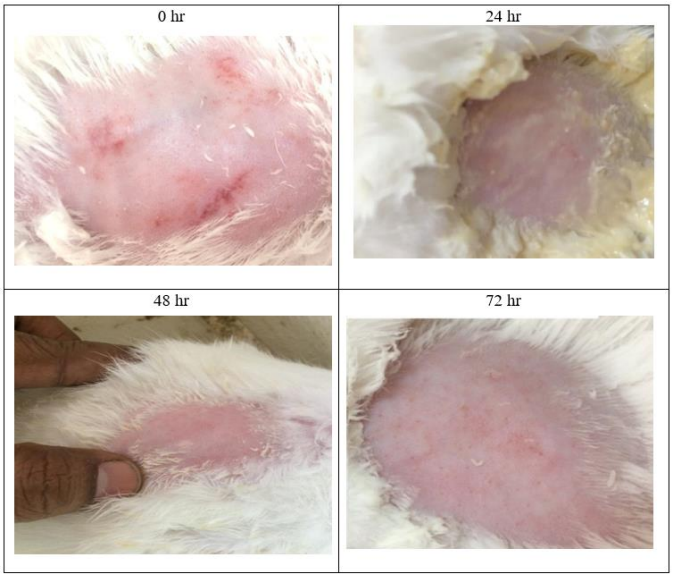

Figure 1: Primary skin irritation studies performed on albino rabbits using anti wrinkle creams at different time intervals.

Stability studies

The result of stability studies showed that the formulations were stable during 3 months. The pH, viscosity, spreadability, extrudability formulations AW2 and AW4 did not change during three months.

Statistical Analysis

From all the above results, the formulations AW2 and AW4 were statistically significant in comparison with the standard with the value of $P < 0.05$.

CONCLUSION

Herbal formulations showed growing demand in the world market. From the above results, it can be concluded that combining different concentrations of various herbal oils showed good anti-wrinkle activity. The method used in this work is simple, fast, economical and also easy to use. The creams AW2 & AW4 showed optimum pH, viscosity, spreadability and percentage extrudability and were found statistically significant in comparison with the standard. This study can be helpful for upcoming researchers to select herbal oils for the formulation and evaluation of other cosmetic applications which can be claimed for their efficacy with scientific data.

ACKNOWLEDGEMENTS

I sincerely thank the principal and staff, UCPSc, KU for giving the required support to carry out this research.

AUTHOR'S CONTRIBUTION

All authors supported to formulate herbal cosmeceuticals which are safe for human use than using chemicals. All authors involved in the manuscript writing and final drafting.

ETHICS STATEMENT

The authors have taken all the necessary permissions as per ethical guidelines wherever applicable. The authors will be responsible for all the technical content mentioned in the manuscript; journal and Publisher will not be responsible for any copyright infringement and plagiarism issue.

DATA AVAILABILITY

The information mentioned above has been gathered from research and review articles as well as books.

CONFLICTS OF INTEREST

None

FUNDING SOURCE

Nil

REFERENCES

1. Kappes UP. Skin ageing and wrinkles: clinical and photographic scoring. Journal of Cosmetic Dermatology. 2004; 3: 23–25.
2. Abdulkarim KA, Nahlah Mansour AA and Shamsan MA. Extraction and formulation of rosemary as anti-wrinkle cream and gel. European Journal of Biomedical and Pharmaceutical Sciences. 2015; 2(3): XX-XX.
3. Amar S, Resham V and Saple DG. Aloe Vera: A Short Review. Indian Journal of Dermatology. 2008; 53: 163-166.

4. Priyanka KG, Mayuri S and Rashmi S. Natural Sunscreen Agents: A Review. Scholars Academic Journal of Pharmacy. 2013; 2(6): 458-463.
5. Arshad AA and Bipul N. Formulation and In-Vitro Evaluation of Poly-Herbal Anti-Ageing Face Cream of Coriandrum Sativum and Rose Hip Oil. International Journal of Current Pharmaceutical Research. 2011; 9(4): 75-78.
6. Chris AA, Cecilia II and Chukwuemeka PA. Evaluation of the Antibacterial activity of Herbal ointments formulated with Methanolic extract of Cassia alata. Asian Journal of Biomedical and Pharmaceutical Sciences. 2012; 2(13):15-19.
7. Evren AY, Özge Ý and Sedef EM. Design and In-Vivo Evaluation of Emulgel formulations including Green tea extract and Rose oil. Acta. Pharm. 2013; 63(4): 531-44.
8. Pratibha N. et al., In-Vitro and In-Vivo assessment of Polyherbal topical gel formulation for the treatment of Acne Vulgaris. International Journal of Drug Delivery. 2012; 4(4): 434-442.
9. Suvidha SP et al., Novel Cosmeceutical Herbal Emulgel for Skincare. World Journal of Pharmacy and Pharmaceutical Sciences. 2014; 3: 801-811.
10. Rafael PV et al., Physical and Physicochemical stability evaluation of cosmetic formulations containing Soybean extract fermented by Bifidobacterium animals. Brazilian Journal of Pharmaceutical Sciences. 2009; 45(3): 515-525.
11. Sonika P et al., Development and Evaluation of antimicrobial herbal cosmetic preparation. African Journal of Pharmacy and Pharmacology. 2014; 8(20): 514-528.
12. People PV and Singh KK. Development and Evaluation of Topical Formulation Containing Solid Lipid Nanoparticles of Vitamin A. AAPS PharmSciTech. 2006; 7(4): 91.
13. Surya. P.M., et al,. Formulation and Evaluation of Anti Aging Poly Herbal Cream. International Journal of Pharmaceutical Sciences Review and Research. 2014; 24(2): 133-136.

Indo Global Journal of Pharmaceutical Sciences(ISSN 2249 1023; CODEN- IGJPAI; NLM ID: 101610675) indexed and abstracted in CrossRef (DOI Enabling), CNKI, EMBASE (Elsevier), National Library of Medicine (NLM) Catalog (NCBI), ResearchGate, Publons (Clarivate Analytics), CAS (ACS), Index Copernicus, Google Scholar and many more. For further details, visit http://iglobaljournal.com