

FORMULATION AND EVALUATION OF HERBAL ANTI ACNE CREAM

Mourya Aman*, Muley Preeti, Malviya Sapna and Kharia Anil

Modern Institute of Pharmaceutical Sciences Alwasa, Behind Rewti Range, Sanwer Road,
Dist Indore (MP) Pin 453111.

Article Received on
05 October 2017,

Revised on 25 October 2017,
Accepted on 15 Nov. 2017

DOI: 10.20959/wjpr201716-10148

*Corresponding Author

Mourya Aman

Modern Institute of
Pharmaceutical Sciences
Alwasa, Behind Rewti
Range, Sanwer Road, Dist
Indore (MP) Pin 453111.

ABSTRACT

The mostly common skin disorder which is known as 'acne', which mainly affects adolescents and adults. According to WHO, acne is an inflammatory disease of the pilosebaceous units in the skin of the face, neck, chest and upper back. It involves both inflammatory (papules, pustules and nodules) and non-inflammatory (comedones, open and closed) lesions. Acne causing bacteria are *P. acnes*, *S. epidermidis* and *S. aureus*. There are many synthetic preparations of formulations are there in the market. They are quite effective but have major side effects which are clinical sometimes. The long term use of antibiotics has led to the increased resistance in acne causing bacteria (i.e. *P. acnes*, *S. epidermidis* & *S. aureus*). For resolving such problems, herbal

preparation is proposed in this article. A herbal preparation with the use of extract of flowers of both plants cauliflower and vinca. Cauliflower have anti bacterial activity, which is due to presence of glucosinolates in it. Vinca flower passes wound healing and skin healing property which is well known. Many literatures claim that plants exhibits anti- inflammatory, anti-microbial, wound healing, antioxidant and hormonal balancing properties. This herbal preparation i.e. cream have good acne curing ability better as compared with marketed formulation.

KEYWORDS: Acne, causes of acne, cauliflower, vinca, formation evaluation, treatment of acne.

1. INTRODUCTION

1.1. ACNE VULGARIS: Acne is the common worldwide skin disorders, it is the condition which mainly affecting adolescents and adults. It is simply characterized by the both

inflammatory (papules, pustules and nodules) and the non-inflammatory (comedones, open and closed) lesions. *Propionibacterium acnes* and *Staphylococcus epidermidis* are the common pus-forming bacteria responsible for the development of various forms of acne vulgaris. All body areas with the high concentrations of pilosebaceous glands are involved, but mostly in particularly face, back and chest. Inflammatory acne lesions can also result in permanent scars or the spots. They may have different type of mechanisms like controlling sebum secretion, antibiotics which inhibit *Propionibacterium acnes*, the main causative organism of the acne, keratolytic which removes the keratin layer and prevents the trapping of sebum under the skin, anti-inflammatory which prevents the worsening of the condition due to mainly inflammation or redness.^[1,2] Current treatments of acne includes the topical and oral antibiotics, topical anti-microbial drugs. All acne treatment has potential side effects, some of which may be severe on longer use. The excessive use of antibiotics for long periods has leads to the enhanced resistance in acne causing bacteria which are *P. acnes*, *S. epidermidis* and *S. aureus*.^[2,3]

1.1.PATHOPHYSIOLOGY OF ACNE:- The most important factors which involved are follows:

1.1.1. INCREASED LEVEL OF SEBUM PRODUCTION

Sebum is the important secretion of the body originating from sebaceous glands. In humans, sebum plays an important role in protecting skin from microorganisms and harmful chemicals. It also potentiates the emollient function of skin by retaining water.^[4] Excess production of the sebum, on the other hand, results in oily skin which may be lead to acne and seborrheic dermatitis.^[5,6] Testosterone, dehydro-epiandrosterone sulfate (DHEAS) and dihydro-testosterone (DHT), are known to regulate genes that are responsible for sebaceous gland growth and sebum production.

1.1.2. PROPIONIBACTERIUM (P) ACNES PROLIFERATION

The proliferation of *P. acnes* releases many enzymes such as proteinases, lipases and hyaluronidases which break down sebum to the free fatty acids and peptides. The inflammatory response to the bacterium and these metabolic by products leads to the formation of papules, pustules and nodules.^[7,8]

1.1.3. ALTERED FOLLICULAR KERATINISATION

In patients with acne, the rate of keratinocyte desquamation at the follicular infundibulum is mainly altered. The keratinocytes accumulate and become interwoven with monofilaments

and the lipid droplets. This accumulation of the cells and the sebum results in the formation of the microcomedones, the microscopic precursor of all the acne lesions.

1.1.4. INFLAMMATION

The bacteria produce the extracellular lipase that hydrolyses sebum triglycerides to glycerol, used by organism as growth substrate and fatty acids, which have proinflammatory and comedogenic properties. Linoleic acid has also been found to regulate IL-8 secretion and reduce the inflammatory reaction. Hence, deficiency of linoleic acid may increase hyperkeratinisation of the epidermis.^[8]

2. PLANT PROFILE

1.1. CAULIFLOWER

Synonyms:- Gobhi, Ful Gobhi.

Biological source

It consists of the fresh flowers of the plant *Brassica oleracea*, belonging to the family brassicaceae.

Chemical constituents

Cauliflower mainly contains several phytochemicals, common in the cabbage family, that are under preliminary research for their potential properties, including isothiocyanates and glucosinolates.^[9]

1.2. VINCA

Synonyms:- Sadabahar, Vinca rosea, Vinca minor.

Biological source

It consists of fresh flowers of the plant *Catharanthus roseus*, belonging to the family Apocyanaceae.

Chemical constituents

Vinca mainly consists of over 130 constituents with alkaloids and tannins some alkaloids are vincristine, vinblastine, vindoline, vinamine, leurosine, ajmaline, vintsine, vinine, vinomine and more.^[10]

3. COLLECTION AND EXTRACTION OF MATERIALS

3.1. COLLECTION OF PLANT MATERIALS

The flower of vinca was collected from local authorized garden and flower of cauliflower was collected from the local market of Indore.

3.2. PREPARATION OF EXTRACTS

Extract of flowers (vinca and cauliflower) were prepared by air drying and were coarsely powdered.^[11,12,13] After preparing powder the procedure for extraction was discussed below.

- ✓ **For vinca (flower) extract:-** 50 gm of dried powder was taken in the conical flask and then 100 ml of ethanol was added to it. The mixture was then packed by aluminium foil and keep aside for 7 days with occasionally stirring. i.e. maceration process. After 7 days the mixture was filtered and evaporated to get dried powder of vinca extract.^[12]
- ✓ **For cauliflower (flower) extract:-** 50 gm of dried powder was taken and allow for extraction in soxhlet apparatus with using 150 ml of ethanol as a solvent. The extraction was run upto 4 days. Then the ethanolic extract was collected and evaporated to get dried powder of extract.^[13]

4. FORMULATION DEVELOPMENT

The formulation components used were listed in Table: 1. Oil in water emulsion of spice extracts and oils were formulated. The oil soluble components (cetyl alcohol, lanolin, mineral oil) and extracts (vinca and cauliflower) were dissolved in the oil phase (Part A) and heated up to 80°C. The water soluble components (glycerin, water) were dissolved in (Part B) and heated up to 80°C. After heating, the aqueous phase was added in portions to the oil phase with constant stirring until cream is formed. Preservatives (Methyl paraben, Propyl paraben) were then added.^[14]

Table: 1 Compositions of developed formulation: Quantity taken for 100g of cream.

Sr.No.	Ingredients	F1	F2
1	Cauliflower ext.	1 gm	1 gm
2	Vinca extract	1 gm	1 gm
7	Cetyl alcohol	5 gm	6 gm
8	Lanolin	4.5gm	4.5gm
9	Liquid paraffin	50 ml	50gm
10	Glycerin	2.5gm	5gm
11	Methyl paraben	0.02gm	0.02gm
12	Propyl paraben	0.02gm	0.02gm
13	Water	35 ml	35 ml

5. EVALUATION OF EXTRACTS AND FORMULATIONS

5.1. EVALUATION OF EXTRACTS

Physical characteristics: the ethanolic extracts of cauliflower and vinca were evaluated for its color, odour, taste, physical state, percent yield and results are shown in table: 2.

5.2. PHYTOCHEMICAL EVALUATION OF EXTRACTS

Analysis of both the extracts was performed by the following standard procedures. In brief,

- ✓ 0.5 ml of extract was added with a drop or two of Mayer's reagent by the side of the test tube and the formation of white or creamy precipitate indicates the presence of alkaloids.
- ✓ Adding 1ml of extract with ammonia and conc. Sulphuric acid and disappearance of yellow colour on standing indicates flavonoids.
- ✓ Formation of brown ring at the interface by the addition of 2ml of glacial acetic acid followed by few drops of ferric chloride solution and 1ml of conc. sulphuric acid to the extracts revealed the presence of glycosides.
- ✓ Adding few drops of neutral ferric chloride to the extract and the development of dark green colour indicates the presence of the organic compounds
- ✓ Existence of froth formation during warming and vigorous shaking indicates saponins. Appearance of brownish green or blue black colouration after adding 0.1% ferric chloride to the cooled extract indicates tannins.
- ✓ Addition of 2ml of chloroform and 3ml of conc. sulphuric acid to the extract and the formation of reddish brown layer at the junction of two solutions confirms terpenoids.
- ✓ Boil the extract with the little amount of Benedict's solution; if glucose is present the colour change from blue to opaque green, then to yellow and finally to red indicates the presence of carbohydrates.^[15]

5.3. EVALUATION OF FORMULATIONS

The formulations were tested for physical evaluation, pH, washability, spreadability and microbial testing.

- **Physical evaluation:** Physical parameter such as color and consistency were checked visually.^[16] The results are mentioned in table: 1.
- **pH measurement:** The pH of various formulations was determined by using Digital pH meter. One gram of each formulation (cream) was dissolved in 100 ml of distilled water (i.e. 1% aqueous solution) and stored for two hours. The measurement of pH of each formulation and average values are mentioned in table: 3.

- **Homogeneity:** These formulations must be produce uniform distribution of the extracts in the cream. This was confirmed by visual appearance and by touch.
- **Washability:** Formulation was applied on the skin and then ease and extent of the washing with water were checked manually.^[16]
- **Spreadability:** Spreadability denotes the extent of area to which the topical formulation (cream) readily spreads on application to skin or the affected part. The spreadability was expressed in terms of time in seconds taken by two slides to slip off from the cream, placed in between the slides, under certain load. Lesser the time taken for separation of the two slides, better the spreadability. Two sets of glass slides of standard dimensions were taken. The cream formulation was placed over one of the slides. The other slide was placed on top of the cream, such that the cream was sandwiched between the two slides in an area occupied by a distance of 6.0 cm along the slide. 100 g weight was placed upon the upper slides so that the gel between the two slides was pressed uniformly to form a thin layer. The weight was removed and the excess of gel adhering to the slides was scrapped off. The two slides in position were fixed to a stand without slightest disturbance and in such a way that only the upper slide to slip off freely by the force of weight tied to it. A 20 g weight was tied to the upper slide carefully. The time taken for the upper slide to travel the distance of 6.0 cm and separated away from the lower slide under the influence of the weight was noted. The experiment was repeated by three times and the mean time taken for calculation. Spreadability was calculated by using the following formula:

$$S = M \times L / T$$

Where, S= Spreadability, M = Weight in the pan (tied to the upper slide), L = Length glass slide, T = Time (in sec.) taken to separate the slides.^[16,17]

➤ **Skin irritation test**

Patch Test:- About 1-3gm of material to be tested was placed on a piece of fabric or funnel and applied to the sensitive part of the skin e.g. skin behind ears. Cream to be tested was applied to an area of 1sq.m. of the skin. Control patches were also applied. The site of patch is inspected after 24 hrs.^[16,17]

- **Microbial Testing:-** The antibacterial activities of different formulations were determined by agar well diffusion method. In this method, nutrient agar plates were seeded with 24 h broth culture of *S. epidermidis*, (collected from Nandani Medical

Laboratories Pvt. Ltd. Kanadia Indore.) a causative organism for acne vulgaris. The agar plates were allowed to solidify. A sterile 8 mm borer was used to cut wells of equidistance in each of the plates solution of 0.2 ml of prepared formulations and marketed herbal cream were introduced into the wells. The plates were incubated at 37°C for 48 hours. The antibacterial activities were evaluated by measuring the zones of inhibition (in mm).^[18,19]

6. RESULT AND DISCUSSION

6.1. EVALUATION OF EXTRACTS

Characteristics of hydroethanolic extracts of cauliflower and vinca:- Color, odour, taste, physical state and percent yield of all three extracts are as follows.

Table: 2 Evaluation of Extracts.

aracteristics	cauliflower	Vinca
Color	Dark brown	Dark green
Odour	Aromatic	Aromatic
Taste	Characteristic	Characteristic
Physical state	Semi solid	Semi solid
Percent yield	7.3gm	2.85gm

Table: 3 Phytochemical evaluation of extracts.

Chemical Test :	Cauliflower extract	Vinca extract
Carbohydrate	+	+
Protein	+	+
Organic compounds	+	+
Triterpenes	+	+
Saponin	-	+
Flavonoids	+	+
Anthraquinone	-	+
Alkaloids	+	+
Tannins	-	+

6.2. EVALUATION OF FORMULATIONS

Table: 3 Evaluation of formulations.

Parameters	control	Formula 1	Formula 2
pH	8.21	7.76	7.8
Colour	Creamy-white	Creamy	Creamy
Homogeneity	Homogenous	Homogenous	Homogenous
Washability	Good	Good	Good
Spreadability (g-cm/sec)	15.55	20.61	21.20

Table: 4 Anti-bacterial activity of formulations.

Formulation	Zone of inhibition (mm)
Formulation 1	22
Formulation 2	25
Control	30

**Figure :- Showing Zone of Inhibition.**

6.3. DISCUSSION

The results of evaluation are displayed in table : 2, 3, 4. Formulation 1 and the formula 2 both are well evaluated. Both in color whereas controlled formulation was creamy white in color. All the formulations were found to be homogenous and easily washable. All formulations had slightly alkaline pH which was compatible with normal skin physiology. All formulations showed zones of microbial inhibition, Formulation 1 showed similar zone of microbial inhibition and Formulation 2 showed better zone of inhibition as on comparison with the control.

7. CONCLUSION

Currently available synthetic anti acne formulation have been associated with number of side effects. On the basis of various literature survey, it was concluded that plants are not only used as preservatives and flavouring agents for food preparation, but also used for various pathological conditions which exhibits strong antimicrobial, anti-oxidant, sebum regulation and anti-inflammatory properties. On the basis of plant's antimicrobial, anti-oxidant, sebum regulation, anti-inflammatory properties, the prepared new formulations were found to very effective against acne.

8. REFERENCES

1. Datin Dr. Asmah Johar et al, "Management of Acne". clinical practice guidelines. 2012 MOH / P/ PAK 234.12 (GU).
2. Patel K.K., Mehta N.J., Dhandhalia M.C., Bhanupriy A.K., Shastri D.H., Shelat P.K., Shah G.B. "Development and Evaluation of Herbal Anti-Acne Formulation". Research Journal of Pharmaceutical, Biological and Chemical. 2012; 3(3): 334-339.

3. Charde Y.M., Sharma P.H., Choudhary N.G. and Avar J.G., "Development and Evaluation of Herbal Formulation for the treatment of Acne". International Journal of Pharmaceutical Sciences and Research. 2014; 5(6): 2250-2260.
4. Abbasi M.A., Kausar A., Rehman A.U., Saleem H., Jahangir S.M., Siddiqui S.Z. and Ahmad V.U., "Preparation of new formulations of anti-acne creams and their efficacy" African Journal of Pharmacy and Pharmacology. 2010; 4(6): 298-303.
5. Daud F.S., Shubhangi W., Joshi M., pande G., "Development of herbal anti-acne gel and its evaluation against acne causing bacteria *Propionibacterium acne* and *Streptococcus epidermidis*". Int. J. Res. Ayurveda pharma. 2013; 4(5): 781-786.
6. C.S. Kandasamy, SumanNath, P. Arulraj, V. Gopal, P. Muthusamy, R. Venkatanarayanan. "Anti-microbial activity of the crude drugs and the Poly herbal formulation (RVSPHF 567) by standardized cup and plate method". International Journal of Pharma Sciences and Research. 2011; 2(10): 189-195.
7. Koli D.S., Mane A.N., Kumbhar V.B., Shaha K.S., "Formulation and Evaluation of Herbal Anti-Acne Face wash". World Journal of Pharmacy and Pharmaceutical Sciences. 2016; 5(6): 2001-2007.
8. Bhatt P.C., Sharma S., "Anti-microbial effects of Purodil Gel on acne causing *Propionibacterium acnes* and *Staphylococcus epidermidis*". International Journal of Research in Cosmetic Science. 2014; 4(1): 7-9.
9. Muhammad Athar Abbasi et al. "Preparation of new formulations of anti-acne creams and their efficacy" 1Department of Chemistry, Center for Natural Product Drug Development, Government College University, Lahore-54000, Pakistan.
10. Yuangang Zu, Huimin Yu, Lu Liang, Yujie Fu, Thomas Efferth, Xia Liu and Nan Wu. "Activities of Ten Essential Oils towards *Propionibacterium acnes* and PC-3, A-549 and MCF-7 Cancer Cells". Molecules. 2010; 15: 3200-3210.
11. Arvind Verma, Into Laakso, Tuulikki Seppänen-Laakso Aarre Huhtikangas Marja-Liisa Riekkola "A Simplified Procedure for Indole Alkaloid Extraction from *Catharanthus roseus* Combined with a Semi-synthetic, Production Process for Vinblastine: Laboratory of Analytical Chemistry, Department of Chemistry, P.O. Box 55, 00014 University of Helsinki, Finland; Tel: +358919150252, Fax: +358919150253.
12. Owis A.J. "Broccoli: A green beauty: A review". Department of pharmacognosy. Beni-Seuf. Egypt. Pg-696-703.
13. Vincent A. Fritz; Carl J. Rosen; Michelle A. Grabowski; William D. Hutchison; Roger L. Becker; Cindy Tong; Jerry A. Wright & Terry T. Nennich (2017). "Growing broccoli,

- cabbage and cauliflower in Minnesota". University of Minnesota Extension, Garden - Growing Vegetables. Retrieved 26 February 2017.
14. Yamini H.K. "Formulation and Evaluation of herbal anti acne gel" Department of Pharmacognosy, DCRP. Pharmacy College, Inkollu, India."
 15. Trease GS, Evans HC. 1978. Text book of Pharmacognosy. 9th ed. Baitar Zindall and Co. London.
 16. Vats A., Sharma P. "Formulation and Evaluation of Topical Anti- Acne Formulation of cream". International Journal of Pharmacy and Pharmaceutical Science Research. 2012; 2(3): 61-66.
 17. Qadry J. S. "Pharmacognosy" published by B. S. Shah Prakashan, twelfth edition 2004-2005.
 18. Mostafa Kamal A.T.M., Kazi Ashfak Ahmed Chowdhury, Limon Kanti Shill, et al. "Phytochemical Screening, Cytotoxic and Thrombolytic Activity of Extract of *Brassica oleracea* Flower (Cauliflower)" Department of Pharmacy, International Islamic University Chittagong, Bangladesh, Global Journal of Pharmacology, 2015; 9(1): 115-120.