Original Article

ISSN Print 2231 – 3648
Online 2231 – 3656

FORMULATION DEVELOPMENT AND EVALUATION OF TRIPLE COMBINATION CREAM OF HYDROQUINONE, TRETINOIN AND MOMETASONE FUROATE FOR THE TREATMENT OF SKIN DISORDERS

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Abstract

Present investigation is aimed to develop a Novel cream formulation consisting of a combination of Hydroquinone,Tretinoin and Mometasone Furoate (4.0%+0.05%+0.1%)w/w as a Depigmenting agent for the treatment of skin disorders like Hyperpigmentation and Melasma due to its wide spectrum of activity and low toxicity.Pigmentary disorders are noncontiguous skin disease, which can be prevented by administration of drugs through topical route having the ability to deliver a higher concentration of drug to the skin than would be possible with systemic therapy. This study aims to develop the triple combination cream due to the additive and synergistic effects of the three components. As there is no topical formulation in combined form, a such formulation of cream is prepared by using multi-ingredients. To assess the efficacy of formulations, assay, drug release, microbial activity, rheology, stability, spreadability, permeability and other physical characteristics were evaluated.One formulation containing Sodium Benzoate as preservative showed desired physico-chemical characteristics amongst all the formulations.

Keywords: Hyperpigmentation and Melasma (skin diseases), Hydroquinone (depigmenting agent), Tretinoin (depigmenting agent), Mometasone Furoate cream.

Introduction

Pigmentary disorders[1-3] of the skin represent the most obvious and often most disturbing clinical changes in the individuals.Hyperpigmentation[3] is a common, usually harmless condition in which patches of skin become darker in color than the normal surrounding skin. This darkening occurs when an excess of melanin, the brown pigment that produces normal skin color, forms deposits in the skin. Age or “liver" spots are a common form of hyperpigmentation. They occur due to sun damage, and are referred to by doctors as solar lentigines. These small, darkened patches are usually found on the hands and face or other areas frequently exposed to the sun.

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People with darker Asian, East Indian, Mediterranean or African skin tones are also more prone to hyperpigmentation, especially if they have excess sun exposure.Melasma[2], also known as *chloasma*, appears as a blotchy, brownish pigmentation on the face that develops slowly and fades with time. The pigmentation is due to overproduction of melanin by the pigment cells, melanocytes. It is reasonably common in women of child-bearing age. However, up to 10% of cases have also been reported in males. All races are affected, but the condition is prominent in Latinos and Asians. Genetic and racial factors may also play a key pathogenetic role. It has been considered to arise from pregnancy, oral contraceptives, endocrine dysfunction, genetic factors and medications etc.This triple combination takes advantage of the additive and synergistic effects of the three components. Hydroquinone[1] has been the gold standard for treatment of hyper-pigmentation for many years. It is a hydroxyphenolic chemical and inhibits conversion of

dopa to melanin by inhibiting the tyrosinase enzyme. The other mechanisms proposed are Inhibition of DNA/RNA synthesis, degradation of melanosomes, destruction of melanocytes.It is commonly used at a concentration of 2-5% for topical application. Retinoid[8] accelerates desquamation and removes preformed melanin. The role of retinoids is likely to be due to its promotion of keratinocyte proliferation and acceleration of epidermal turnover. Tretinoin[8] induces dispersion of pigment granules inside the keratinocyte, and accelerates the turnover of epidermal cells, facilitating the elimination of dispersed pigment. They inhibit the tyrosinase activity, affect the secretory function of melanocytes and have anti metabolic effect on keratinocytes. Corticosteroids[10] also inhibit the various mediators of inflammation and hence also inhibit the stimulatory impulses for melanocytes.Mometasone Furoate[6] is a corticosteroid.Its mechanisms of action are related to vasoconstriction and suppression of membrane permeability, mitotic activity, the immune response and release of inflammatory mediators.This study aims to develop the triple combination *(Hydroquinone, Tretinoin and Mometasone Furoate)* cream due to the additive and synergistic effects of the three components. Acceptability and clinical efficacy of such preparations require them to posses optimal mechanical properties (ease of removal from the container, spreadability on the substrate), rheological properties (viscosity, elasticity, thixotropy, flowablity), and other desired property such as bioadhesion, desired drug release and absorption The objective of this research is to formulate a stable and good physical appearance cream which is readily suitable for application.

Materials and Methods

Hydroquinone, Tretinoin and Mometasone Furoate were received as gift samples from M/s.Glenmark Research Center, Malegoan MIDC, Sinnar, Nashik, India. Stearic Acid, Cetyl Alcohol, Stearyl Alcohol, Glyceryl Monostearate (NSE), Polyoxyl 20 Cetyl Ether (Brij58), Liquid Paraffin, Isopropyl Myristate, Dimethicone 350, Butylated Hydroxytoluene, Hydroxypropyl betadex, Disodium Edetate, Citric Acid Monohydrate, Glycerin, Veegum HV, Sodium Benzoate, Sodium Metabisulphite, Ascorbyl Tetra 2-Hexyl decanoate (VC-IP), Propylene Glycol were obtained from M/s. S.D.Fine chemical Mumbai of pharmaco - poeial quality. Formulation and evaluation studies were carried out at department of pharmaceutics, annamali university, Tamilnadu by using various instruments like Mechanical Stirrer, Homogenizer, pH meter, Brookfield Viscometer, HPLC, Karl-fischer titrimeter, UV spectro - photometer, Water Bath etc.

Method for preparation of cream

Formulation code-A

Hydroquinone, tretinoin and mometasone furoate cream was formulated as follows:

In a SS Container Liquid Paraffin, Polysorbate 60 (Tween 60), Cetyl Alcohol, Polyoxy 20 Cetyl Ether (Brij 58), Butylated Hydroxytoluene (BHT), Benzyl Alcohol, Dimethicone 350cps, Ascorbyl Tetra 2-Hexyldecanoate (VC-IP) and Tretinoin was taken and heated on water bath up to 70˚C to 72˚C and Maintained temperature up to 70˚C to 72˚C.In an another SS Container Purified water, Glycerine, Disodium Edetate and Hydroxypropyl betadex was taken and added slowly under stirring into it Carbopol 940 and Pemulen TR1 to disperse it completely. Heated on water bath up to 70˚C to 72˚C.Step 1 is to added with step 2 under homogenization at 70˚C For 10 min.In a SS Container add Hexylene glycol, Motetasone Furoate (micronized), was heated upto 70˚C to dissolve it completely. Cool to 40˚C and then Purified water, Hydroquinone and Heat up to 40˚C to dissolve it completely. Add this phase to the bulk at 40˚C under stirring. In a SS Container add, Purified water and Triethanolamine, to dissolve it completely and add this phase to the bulk at 40˚C under stirring. In SS Container add purified water and Sodium metabisulphite and add this phase to the bulk at 40˚C under stirring. Check the weight and pH.

Formulation code-B

In a SS Container add, Stearic Acid, Cetyl Alcohol, Stearyl Alcohol, Glyceryl Monostearate (NSE), Polyoxy 20 Cetyl Ether (Brij 58), Isopropyl Myristate, Dimethicone 350cps, Ascorbyl Tetra 2-Hexyldecanoate (VC-IP), Butylated Hydroxytoluene (BHT) and Tretinoin and Heat on water bath up to 70˚C to 72˚C.Maintain temperature up to 70˚C to 72˚C.In an another SS Container add Purified water, Citric acid monohydrate, Disodium Edetate, Hydroxypropyl betadex and add

Glycerine and Veegum HV (In a separate beaker disperse Veegum HV in glycerin).Heat on water bath up to 70˚C to 72˚C. Add step 1 to step 2 under homogenization at 70˚C For 10 min.In a SS Container add Hexylene glycol, Motetasone Furoate(micronized) and Heat upto 70˚C to dissolve it completely. Cool to 40˚C And then add, Purified water and Hydroquinone. Heat up to 40˚C to dissolve it completely. Add this phase to the bulk at 40˚C under stirring. In a SS Container add, Purified water and Sodium Benzoate. Add this phase to the bulk at 40˚C under stirring. In SS Container add, Purified water and Sodium metabisulphite. Add this phase to the bulk at 40˚C under stirring. Check the weight and pH.

Formulation code-C

In this formulation all oil phase and aqueous phase are same as that of the previous formulation code-B. But only changes occurred at addition of drug phase. In a SS Container add, Propylene glycol and Motetasone Furoate (micronized), to disperse it completely. Add this phase to the bulk at 40˚C under stirring. In an another ss container add, Propylene Glycol and Purified water, Heat upto 30˚C and add Hydroquinone To dissolve it completely. Add this phase to the bulk at 40˚C under stirring. Then the sodium benzoate phase and Sodium metabisulphite phase will be prepared and added in the same manner as that of the previous formulation code-B. Then Check the weight and pH.

Formulation code-D

Here the formulation is same as that of formulation code C, but the concentration of Citric Acid Monohydrate has changed to adjust the pH.

Evaluation Parameter

Spreadability

Spreadability[9] is a term expressed to denote the extent of area to which the Cream readily spreads on application to skin or the affected parts. The therapeutic efficiency of a formulation also depends upon its spreading value. Hence, determination of spreadability is very important in evaluating Cream characteristics. A special apparatus has been described to study the spreadability of Cream formulation. The spreadability is expressed in terms of time in seconds taken by two slides to slip-off the Cream, placed in between the slides, under the direction of certain load. Lesser the time taken for separation of two slides, better the spread ability.

$$S = m.\ l/t,$$

Where, S = Spreadability, m = weight tied to the upper glass slide, L= length of glass slide, t = time taken.

Tube Extrudability[5]

It is an usual empirical test to measure the force required to extrude the material from bottle or tube. The formulation under study was filled in a clean, lacquered aluminium collapsible one-ounce tube with a nozzle tip of 5 mm opening and the pressure applied on tube with the help of fingers. Tube extrudability was then determined by measuring the amount of Cream that extruded through the tip. More quantity extruded, better was extrudability.

Viscosity

The consistency of cream was determined by LVT Brookfield viscometer. In a clean and dry 250ml beaker, take the test sample. Determine the viscosity of the test sample as per standard operating procedure of viscometer by using spindle nos. 1 to 4. Use each of the spindles for finding out the viscosity of the sample at speeds of 0.3, 0.6, 1.5, 3, 6, 12, 30 and 60 RPM respectively. Record the dial reading and calculate the viscosity[9] of test sample. Calculate the viscosity in centipoises (cps) by using following formula, Viscosity in centipoises (cps) = Dial reading x Factor. For calculation of viscosity put the factor value corresponding to the speed and the spindle number. Check accuracy and reproducibility of results.

pH

The pH[9] was determined by using digital pH meter. Before measuring the pH of sample ensures that the pH meter is calibrated one step above and one step below the sample pH value. Calibration buffer value and sample value is not differing more than 4.00 units. Clean the electrode dipped in storage buffer with

distilled water and removes excess water by soaking it with tissue paper and inserts the pH electrode in sample. After completion of pH measurement clean the electrode, dipped in storage buffer with distilled water and removes excess water by soaking it with tissue paper and dip the electrode in 3M KCL solution.

Stability Studies

Stability[5] is defined as the capacity of a drug substance or drug product to remain within established specifications to maintain its identity, strength, quality and purity throughout the retest or expiration dating periods. The Hydroquinone, Tretinoin and Mometasone Furoate cream was filled in the collapsible tubes and stored at –

25°C±2°C/60%RH±5%RH,
30°C±2°C/65%RH±5%RH,
40°C±2°C/75%RH±5%RH

For a period of three months and studied for viz. appearance, pH, viscosity and assay of drug.

Table 01: Physico-chemical evaluation of Hydroquinone, Tretinoin and Mometasone Furoate creams

S. No	Formulation Code	Appearance	pH	Viscosity (Centipoises)	Spreadability g.cm/sec	Extrudability (%)
1	A	Yellow semisolid cream	4.78	57000	11.23	91.92
2	B	Yellow semisolid cream	4.87	64000	8.79	90.33
3	C	Yellow semisolid cream	5.02	61000	10.42	94.38
4	D	Yellow semisolid cream	5.35	66000	7.97	88.75

Table 02: Stability evaluation data of Hydroquinone, Tretinoin and Mometasone Furoate creams

Name of Product	**Hydroquinone, Tretinoin and Mometasone Furoate Cream (4%+0.05%+0.1%)w/w**	**Formulation Code** : **C** **Stability Date** : 07/01/11	
Composition:	Hydroquinone USP 4.0%w/w Tretinoin USP 0.05%w/w Mometasone Furoate 0.1%w/w Preservative Sodium Benzoate IP 0.2% w/w	**Mfg. Date** : 06/01/11 **Exp. Date** : **----**	

		25°C/60%RH			30°C/75%RH			40°C/75%RH		
Tests	**(0) Initial**	**1M**	**2M**	**3M**	**1M**	**2M**	**3M**	**1M**	**2M**	**3M**
Appearance	Yellow semisolid cream	….	….	complies	….	….	complies	complies	complies	Complies
pH	5.02	….	….	5.17	….	….	5.21	5.07	4.98	4.86
Viscosity (Centipoises)	61000	….	….	60000	….	….	61000	62000	59000	58000
Spread ability g.cm/sec	10.42	….	….	10.47	….	….	10.58	10.22	10.98	11.03
Extrudability (%)	94.38	….	….	96.26	….	….	95.53	95.41	96.12	96.72
% Assay Hydroquinone (90%-110%)	103.78	….	….	101.55	….	….	101.64	103.22	102.67	102.89
% Assay Tretinoin (90%-117%)	108.43	….	….	106.21	….	….	104.32	107.31	105.28	106.01
% Assay Mometasone Furoate (90%-110%)	101.92	….	….	100.52	….	….	100.11	100.54	99.98	100.65
% Assay Sodium Benzoate (80%-120%)	109.13	….	….	106.32	….	….	105.87	108.77	107.65	105.66
Date	07/01/09	….	….	07/04/11	….	….	07/04/11	07/02/11	07/03/11	07/04/11

Results and Discussions

The composition and physico-chemical characteristics of the cream formulations with different preservatives viz. Benzyl Alcohol, Sodium Benzoate and with different pH modifiers viz. Triethanolamine and Citric acid monohydrate are shown. All the prepared cream formulation was subjected to stability study as per ICH guidelines for a period of three months. Since in formulations, the % assay of drug and preservative was degraded at 40°C/75%RH in third month. After one month, physical observation showed that there is no smooth appearance and a phase separation was there in all formulation except formulation code C and D. These two formulation show good physico chemical behavior in compare to all others. Here drug (Mometasone Furoate) is in dispersed form, where in others it is in dissolved form. And here propylene glycol is used instead of hexylene glycol as a solubiliser.Formulation code D is more viscous than C.In Formulation code C the pH of the formulation is 5.00 at which the drugs are stable, where in other pH stability problem of all the three drugs occurs. From the data it is evident that the % assay of drug in formulation-C containing Sodium Benzoate as preservative was found to be most satisfactory as comparable with other formulations.

Conclusion

Prepared Hydroquinone, Tretinoin and Mometasone Furoate Cream (4.0%+0.05%+0.1%)w/w was evaluated for viscosity, homogeneity, extrudability, drug content, pH, spredability. The formulation code C showed a good homogeneity, spreadability, pH& viscosity among all the formulations. From the data it is evident that the % assay of drug in formulation-C containing Sodium Benzoate as preservative was found to be most satisfactory as comparable with other formulations. Hence it is concluded that formulation code-C was found to be the best amongst all the formulations and exhibited ideal physico-chemical characteristics. From the stability study as per the ICH guidelines it was concluded that formulation-G was stable for the period of three months in accelerated condition. Hence we can propose a shelf life for 18 months or API expiry whichever is less.

Future Study

Due to limitation of time various studies could not be completed which may be taken up in future study:

- Clinical efficacy study in animals.
- In vivo study in animals
- Human trials.
- For detail dissolution study on cellophane sheet.
- For related substance limit.

Acknowledgement

I express my profound and sincere gratitude Mr.Nitin. B. Bhamre, Senior Research Scientist, Glenmark Pharmaceuticals Ltd, Nashik, Maharashtra, for providing all the facilities and support during my academic period of research. My whole hearted gratitude towards our beloved and more respected Dr.P.Sudhir Kumar, Asst.Professor, SOA University, BBSR, Odisha for his persuasiveness, encouragement, constant inspiration and providing all necessary facilities throughout the entire course of my work. My sincere thanks to all teaching and technical staff of Department of pharmaceutics, IPT, Salipur, Cuttack who have helped me in the successful completion of my project. There is no word to express my gratitude and thanks to my beloved parents who have made me every dream to come true by sacrificing their happiness. Above all thank GOD for his providence at every stage of my work.

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