CHAPTER 7

SUMMARY AND CONCLUSIONS.

Topical preparations constitute a considerable proportion of the dosage forms used in India and corticosteroid creams occupy the key position amongst topical preparations. Because of peculiar climatic conditions the expectation from corticosteroid creams by the consumers vary to a great extent. Although the Indian market is full of all types of corticosteroidal creams yet most of these products do not stand to the consumer expectations. This study is an attempt to formulate the creams of corticosteroid drugs, based upon the experience of the consumers and the prescribers.

The creams that should satisfy most of the requirements on scientific and therapeutic basis, the creams which possess desirable organoleptic properties, the topical availability requirements and the toxicity considerations.

The drugs selected for the present study are triamcinolone acetonide, betamethasone 17-valerate, halcinonide and fluocinolone acetonide are of proven therapeutic value and hence an humble attempt has been made in this study to achieve perfection in the formulation of their creams, particularly to suite the climatic conditions in India.

The analytical method used for the determination of TA, HAl, FA was based upon the reaction of α, β unsaturated steroidal ketones with isoniazid and the analytical method used for the determination of BV was based upon the reaction of α-ketol group with tetrazolium blue in alkaline medium to yield coloured product. Standard curves of corticosteroid

drugs were prepared. The range in which Beer's curve is obeyed has been established.

Name of drugs	nm	Beer's law limit
1. Triamcinolone acetonide	415	0-10 µg/ml.
2. Betamethasone 17-valerate	525	0-9 µg/ml.
3. Halcinonide	377	0-15 µg/ml.
4. Fluocinolone acetonide	415	0-12 µg/ml.

Estimation of drug in presence of large number of ingredients, revealed that almost none of the ingredients used in the formulation of creams interfere in the estimation of steroids incorporated therein, with the exception of Tween 80 and Tween 60 which gives slightly higher absorbance than the normal. However the interference due to Tween 80 and Tween 60 considered insignificant in the present investigation.

Hydrophilic creams have been widely accepted as cream bases in the compounding of pharmaceutical formulations, since it became official in USP XIII. These cream bases belong to those bases classified as hydrophilic or water-washable, most of which are o/w emulsions. Such cream bases offer many advantages over the older greasy type preparations.

For the preparation of cream bases, oils and oil soluble ingredients were weighed and transferred in a glass beaker. The ingredients were dissolved by heating the mixture on water bath at 80 to 85° (oil phase). Water soluble ingredients and purified water were weighed and transferred in another

glass beaker. The ingredients were dissolved by heating the mixture on water bath at 80 to 85°. The temperature was maintained and the solution was ensured by stirring (Aqueous Phase).

The oil phase was slowly transferred to homogeniser at 80 to 85°, by straining through nylon cloth or double folded muslin cloth. The aqueous phase was slowly added to above oily phase at 80-85°, by straining through nylon cloth. Along with homogeniser, propeller mixer was also used simultaneously for emulsification. The homogenisation and agitation continued for 30 to 45 minutes, while maintaining temperature at 80-85°. Homogenisation/agitation of emulsion was continued until the batch cool down to 50-52°. Sufficient warm purified water (50-52°) was added to make up for the loss due to evaporation. When the temperature reached 40-50°, the stirring was started with planetory mixer blades at slow speed until the mass congealed. Precautions are recommended for the preparation of creams are already discussed in Chapter 4.

The suitable compositions of the bases were developed by studying the first, second and third screening evaluations in which the cream base properties like grainy, separation, satisfactory, unsatisfactory, spreadability, water washability, consistency, pH, water content, congealing point and compatibility etc. were evaluated.

Out of one hundred and ten hydrophilic cream bases about thirty four were selected for further studies as they were found to be stable and most satisfactory in all

the three screening evalations during the stability of one month at different conditions (AC, RT, 37°, 42°/80% RH).

Promising cream bases were subjected to rheological evaluation and measurements, like hysteresis loop, dynamic yield value and the apparent viscosity derived at a definite point on the rheogram, were made. In the rheological studies all the cream bases exhibited non-Newtonian flow characteristics, a desirable attribute. The systems also exhibited thixotropy.

Based on the rheological parameters, twenty seven bases were selected for formulation of corticosteroid creams. Drugs were incorporated in following concentration in each of the selected twenty seven bases.

	Concentration.
1. Triamcinolone acetonide	0.1% w/w
2. Betamethasone 17-valerate	0.122% w/w
3. Halcinonide	0.1% w/w
4. Fluocinolone acetonide	0.025% w/w

The prepared creams were tested for release pattern of drugs <u>in vitro</u>, in distilled water, using cellophane membrane employing model developed in laboratory and simultaneously kept for stability studies at A.C., R.T., 37°, 42°/80% R.H. The evaluation of microbial contamination in cream bases was done by using different concentration and proportion of the preservatives. The total microbial count was found out initially and after storage of one month at R.T. The presence of gas formers or pathogenic organisms were also found out. Based on the results obtained the optimum concentration of preservatives

or if required more concentration of preservatives along with antioxidants were used in the formulations of corticosteroid creams and kept on stability at R.T. (29 ± 2°).

From the results obtained it has been concluded through the analysis of *in vitro* release patterns that there are significant differences in the release of corticosteroids from all the creams.

Cream No. 27 containing cetostearyl alcohol, light liquid paraffin, polyoxyethylene lauryl ether, cetomacrogel 1000, propylene glycol, polyethylene glycol 6000 distearate, 2-pyrrolidone etc. released a maximum amount about 18.43% of TA whereas minimum amount about 1.99% of TA was released from cream No. 11, containing glyceryl monostearate SE., Stearic acid, White soft paraffin, etc. in distilled water at 37° for 7 hrs.

Cream No. 1 containing glyceryl monostearate SE., stearyl alcohol, light liquid paraffin, isopropyl myristate, cetomacrogol 1000, glycerin and castor oil released a maximum of about 17.78% and a minimum of about 1.47% of BV from cream No. 15 containing white bees wax, white soft paraffin, heavy liquid paraffin, light liquid paraffin, cetomacrogol emulsifying wax, borax and chlorocresol, etc. in distilled water at 37° for 7 hrs.

Cream No. 1 released maximum amount about 9.91% of HAL and minimum amount about 1.14% of HAL from Cream No. 11 in distilled water at 37° for 7 hrs.

Cream No. 1 released maximum amount about 19.8% of FA and minimum amount 2.3% of FA was released from Cream No. 11 in distilled water at 37° for 7 hrs.

Based on the in vitro release rate study, promising seven formulations out of twenty seven formulations in respect of total percentage release, drug releasing efficiency and the release rate were subjected to simulated release study. Liberation of corticosteroids through the sartorius ointment chamber assembly, showed nearly the same pattern of release as found in in vitro release study of all promising formulations, but lesser in amount.

Corticosteroid creams selected on the basis of their in vitro evaluation were subjected to bioavailability study employing vasoconstrictor assay. The whole experiment was conducted in consultation with a dermatologist and a physician. The results of both readers were in good agreement throughout the test, and the data were therefore combined for the statistical treatment.

Comparing the blanching results with in vitro release in laboratory designed model and liberation of corticosteroids through sartorius ointment chamber assembly, cream No. 13 showed very poor results in all the creams in blanching test. Cream No. 7 containing betamethasone 17-valerate has also shown a poor blanching readings. Other creams have shown comparable results with in vitro release except in fluocinolone acetonide creams where cream No. 27 and 25 gave more blanching readings than cream No. 1 and in betamethasone 17-valerate creams where cream No. 27 gave more blanching

readings than cream No. 1.

Blanching test demonstrates clearly that the degree of vasoconstrictor activity produced by a steroid, reflecting the degree of skin penetration, may be influenced considerably by the composition of the base in which it is formulated. Halcinonide formulations produced better blanching of skin than the corresponding formulations; followed by betamethasone 17-valerate, fluocinolone acetonide and triamcinolone acetonide.

Primary skin irritation test in rabbits showed that non of the selected creams was irritant to the intact and abraded skin of rabbits. Thus from this part of the work it is concluded that it is safe to use the all selected creams.

The stability studies showed that TA cream Nos. 1,5, 13 and 27 were stable for more than one and half year at all the stability conditions but cream No. 25 separated after nine months at 42°/80% R.H., and after one and half year at 37°. The degradation products were absent in all above creams.

BV cream Nos. 1,13 and 27 were stable at all the stability conditions but cream No. 7 separated after three months of storage at 42°/80% R.H. and cream No. 5 separated at 42°/80% R.H. after one and half year of stability. The degradation products were absent in all above products.

HAL cream Nos. 1,5,13,25 and 27 were stable in all conditions during storage of one and half year. The degradation products were absent in all the above mentioned formulations.

FA cream No. 5 was stable in all the stability conditions during storage of one and half year. Cream Nos. 1 and 25 were separated at the end of one and half year at 42°/80% R.H. Cream No. 13 was separated after six months at 42°/80% R.H. and cream No. 27 was separated after nine months at 42°/80% R.H. Cream No. 13 showed degradation of FA from 0.025% to 0.02% after one and half year. The degradation was observed in Cream No. 27 and FA concentration was reduced from 0.025% to 0.019% after six months of storage at 42°/80% R.H. Cream No. 25 showed degradation of FA from 0.026% to 0.021% after twelve months at 42°/80% R.H. However, no degradation was observed at A.C. temperature.

The stability studies show that all the selected creams for blanching test have sufficiently high stability at A.C. and R.T. for one and half year.

According to new concept of USP for non-sterile topicle preparations, it is being necessary to maintain the potency and integrity of product forms and to protect the health and safety of the consumer. Micro-organisms could cause disease or product deterioration, so it is very dangerous to have a pathogenic micro-organisms or any micro-organism which is dangerous to the health. Hence a systematic study of total viable count in the cream bases using different concentration and combination of preservatives was carried out.

The total viable count studies showed that cream base Nos. 2,3,6,8,9,11,12,13,15 to 27 were free of pathogenic organisms and there was no gas formation, even though cream base Nos. 3,9,11,13,15,21,22,24 and 25 showed very high bacterial counts at lower concentration of combinations of methyl paraben sodium (0.18%)-propyl paraben sodium (0.02%).

Cream base Nos. 1,4,5,7,10 and 14 showed very high bacterial counts and presence of gas formers with methyl paraben sodium propyl paraben sodium combinations. The bacterial counts dropped to a greater extent when 0.1% chlorecresol was used as preservative. Chlorecresol was found superior to the methyl paraben sodium-propyl paraben sodium combinations, probably because of the increase in phenolic strength imparted to the hydroxy-group by halogen atoms ortho- or para- to it.

Based on all the results, the following cream bases were suggested for the formulations of corticosteroid drugs as they appears to be promising with regards to physical evaluation, rheological evaluation, in vitro release studies, bioavailability studies, primary irritation studies, stability and microbial contamination studies. Cream base Nos. 1 and 5 are the most promising bases for the formulation of TA creams. Cream base Nos. 1 and 27 are the most promising bases for the formulation of BV creams. Cream base Nos. 1,5 and 27 are the most promising bases for the HAL creams. Cream base No. 5 and to a some extent cream base Nos. 1 are the most promising bases for the FA creams, as FA cream No. 1 was separated at the end of one and half year at 42°/80% R.H.

All the promising creams were soft homogeneous, odourless, free of visible impurities. Microscopic observation of all the promising creams showed no visible particle and the emulsified globules were found in nearly same size. The particle size measured with the help of hegman were less than 25 microns. The characteristics like physical stability, separation, spreadability, washability, consistancy, pH, water retention, congealing points, compatibility were also checked before and during stability and all the promising creams were found to be satisfactory at A.C. and R.T. during one and half year of stability.