Chapter - 7

SUMMARY, CONCLUSIONS AND RECOMMENDATIONS
Chapter -7

SUMMARY, CONCLUSIONS AND RECOMMENDATIONS

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Name of the Sub-Title</th>
<th>Page No.</th>
</tr>
</thead>
<tbody>
<tr>
<td>7.1</td>
<td>Summary</td>
<td>184</td>
</tr>
<tr>
<td>7.2</td>
<td>Conclusions</td>
<td>187</td>
</tr>
<tr>
<td>7.3</td>
<td>Recommendations</td>
<td>187</td>
</tr>
</tbody>
</table>
CHAPTER - 7
SUMMARY, CONCLUSIONS AND RECOMMENDATIONS

7.1 SUMMARY
The present study gives emphasis on the formulation of novel drug delivery system for the treatment of topical fungal infection. By considering the limitations of conventional dosage form and antifungal drugs, here attempt has been made to formulate solid lipid nanoparticles of BUTE and SERT made up of natural excipients for the effective monitoring of fungal infections. BUTE is benzylamine and SERT is imidazole type antifungal drugs mainly active against dermatophytes and Candida albicans and also active against Cryptococcus neoformans and Aspergillus spp. Both drugs are classified with low solubility, itching and burning sensation. To overcome the constraint of conventional dosage form i.e permeability of stratum corneum, the formulation of BUTE and SERT loaded SLN would propose to be an excellent topical delivery for treatment of fungal infections. The BUTE-SLN and SERT-SLN were prepared, characterized and then incorporated into Aloe vera gel and further evaluated for in vitro and ex vivo studies.

Pure drugs were evaluated for the XRD, DSC and FTIR study. The formulation of SLN was initiated with the selection of excipients by solubility study. Different solid lipids, surfactants and cosurfactants were analyzed for the highest solubility of selected drug. Solid lipid OLML and GMS were selected while surfactant OLMS was selected as aqueous phase surfactant for the pre optimization study. Other excipients like stearyl amine, DMSO, acetone and poloxamer 188/407 were added and evaluated to improve the properties of SLN formulation.

Modified solvent emulsification technique was implemented for the fabrication of BUTE-SLN and SERT-SLN dispersion. The formulations were prepared using different excipients with their varying concentration to reach the optimum concentration of excipients. Influence of various process parameters on the formulation of SLN were evaluated on the basis of average particle size, polydispersity index, zeta potential and in vitro drug release study. Amongst the various batches, formulation F20 and R20 were
found to have minimum particle size and polydispersity index, maximum zeta potential and controlled release of drugs from SLN.

A $2^3$ factorial design was used to optimize the BUTE-SLN dispersion. The P value was found to be less than 0.05 and the models were found to be significant for all the responses. Optimized BS9 BUTE-SLN was composed of 5% OLML, 4% OLMS, 0.5% DMSO and 1.5% TPGS. The optimized formulation have 261.25±2.38 nm particle size, 0.268±0.01 PDI, 23.98±0.27 mV ZP, 91.35±2.35 % EE and 19.692±0.95% DL. The BS9 BUTE-SLN dispersion was evaluated for the color, odor and stability. The nanometric dispersion was lyophilized using 5%w/w mannitol and the freeze dried BS9 BUTE-SLN powder was evaluated for XRD, DSC and FTIR study. The study reveals the amorphous nature of BS9 BUTE-SLN and the formulation was found to be compatible.

BS9 BUTE-SLN was incorporated into Aloe vera gel. The rheological study of BS9 BUTE-SLN aloe vera gel was found to show non Newtonian pseudoplastic flow behavior. The ease of spreading, uniformity, consistency and adhesion on skin were shown by the BS9 BUTE-SLN gel. The occlusion factor for BS9 BUTE-SLN gel was found to be 45.7±0.44% at the end of 72 hr, proving the impermeable layer on the stratum corneum. In accordance to the occlusion study, skin hydration study reveals the more hydration of skin as compared to marketed cream.

*In vitro* drug release study of BS9 BUTE-SLN gel was compared to FINTOP® cream. The flux value was found to be 1666.7 ± 0.198 ng/cm²/hr for BS9 BUTE-SLN gel and 818.181 ± 0.392 ng/cm²/hr for reference cream. The gel showed biphasic release pattern initial burst and further sustained release. Retention of butenafine in different layers of skin was quantified and stratum corneum showed maximum localization of drug as compared to reference cream.

Primary skin irritation potential was determined for BS9 BUTE-SLN aloe vera gel and it showed no erythema and edema scores. BS9 BUTE-SLN aloe vera gel was found to be safe and non irritant for topical application. *In vitro* antifungal activity reveals the improved antifungal property of BS9 BUTE-SLN aloe vera gel as compared to marketed
and standard solution. BS9 BUTE-SLN aloe vera gel was subjected to different storage condition and found to be more stable at all storage conditions.

Taguchi experimental design was used to optimize the SERT-SLN formulation. The results were analyzed by Design Expert software and all the models for independent variables were found to be significant (P<0.05). All the variables have shown positive as well as negative influence on the dependant variables. The optimized formulation SS10 SERT-SLN was composed of 4% OLML, 4% OLMS, 1.122% DMSO and 0.288% stearyl amine. The optimized formulation was found to have 153.8±2.08 nm particle size, 0.23±0.016 PDI, 9.76±0.08 mV ZP, 93±2.32 % EE and 15.4±0.53 % DL. The SS10 SERT-SLN dispersion was observed to be in spherical and nanometric size by TEM study.

The SS10 SERT-SLN dispersions were subjected to the stability study and then evaluated, it was found that the all the dispersions were stable at all the storage condition of temperature and time. The type of cryoprotectant and its concentration was optimized and concluded to be 5%w/w mannitol as cryoprotectant showed less reconstitution time and no signs of aggregation. The freeze dried SS10 SERT-SLN was analyzed for XRD, FTIR and DSC. XRD and DSC study reveals the transformation of crystalline SERT and OLML into nanometric amorphous SS10 SERT-SLN. The excipients like SERT, OLML and others were found to be compatible by FTIR study.

Semisolid topical dosage form of SS10 SERT-SLN was prepared by incorporating it in to Aloe vera gel. The SS10 SERT-SLN gel was white translucent in color with pH 6.7 and the percent assay was calculated to be 98.54±1.54%. SS10 SERT-SLN aloe vera gel showed non Newtonian pseudoplastic flow behavior and also exhibited excellent ease of spreading, uniformity and consistency as compare to marketed cream. Occlusive study demonstrates the higher occlusion factor F 48.0099±0.42% at the end of 72 hr which reveals the potential of SS10 SERT-SLN gel to form an impermeable layer to prevent water loss. Ex vivo skin hydration study was performed and it was observed that the SS10 SERT-SLN gel have more potential to hydrate the human cadaver skin as compare to
marketed cream, hence more concentration of SERT can be penetrated through swollen corneocytes and intercellular lipids.

*In vitro* drug release profiles of SS10 SERT-SLN also showed biphasic release profile which starts initially with burst release for first few hours and then prolong sustained release. SLN gel showed faster and increased drug release than marketed cream revealing its potential for higher drug penetration as supported by the skin retention study. The flux value was calculated to be $7.5 \pm 0.392$ mcg/cm$^2$h for SLN gel and $4 \pm 0.198$ mcg/cm$^2$h for reference cream.

Primary skin irritation study reveals the non irritant property of SS10 SERT-SLN gel for topical use. *In vitro* antifungal activity showed the enhanced antifungal activity of SS10 SERT-SLN gel as compared to marketed gel. Stability study of SS10 SERT-SLN gel was performed and it was found to be stable at all storage conditions.

### 7.2 Conclusion

In conclusion, solid lipid nanoparticles composed of antifungal drugs and natural novel excipients were prepared. A modified solvent emulsification method was employed and the formed SLNs were evaluated for physicochemical properties. Optimization tool was opted to minimize the preparation run and to reach at optimum formulation. Different characterization study, *in vitro* and *ex vivo* evaluation study and stability study supports the successful formulation of SLN, safe and enhanced penetration of antifungal drugs in the skin. Hence present study demonstrates the effective targeting of BUTE SLN and SERT SLN to the skin as compared to conventional cream.

### 7.3 Recommendation

The present work focuses on the preparation and evaluation of BUTE and SERT loaded SLN formulated using naturally derived excipients to achieve improved penetration and effective treatment of fungal infections. However, the work could be extended further to *in vivo* studies, clinical trials and scale up techniques.