# TABLE OF CONTENTS

1. INTRODUCTION
   1.1 CANCER THERAPEUTICS
   1.2 LIPOSOMES
   1.3 ESTIMATION OF DRUGS IN BIOLOGICAL FLUIDS
   1.4 TOXICOKINETICS IN PRECLINICAL EVALUATION

2. AIM AND OBJECTIVES
   2.1 AIM OF THE RESEARCH WORK
   2.2 OBJECTIVES

3. LITERATURE REVIEW
   3.1 CANCER THERAPEUTICS
   3.2 LIPOSOMES
   3.3 TOXICOKINETIC STUDY
   3.4 IRINOTECAN

4. MATERIALS AND METHODS
   4.1 MATERIALS AND SUPPLIERS
   4.2 METHODOLOGY
     4.2.1 LIPOSOMES
     4.2.2 ANALYTICAL METHOD FOR IRINOTECAN
       4.2.2.1 METHOD OPTIMIZATION PROCEDURE
       4.2.2.2 ASSAY METHOD VALIDATION
   4.2.3 BIOANALYTICAL METHOD FOR IRINOTECAN AND 7-ETHYL 10-HYDROXY CAMPTOTHECIN (SN38) IN RAT PLASMA
     4.2.3.1 COLLECTION OF BLANK RAT PLASMA
     4.2.3.2 CHROMATOGRAPHIC CONDITIONS
     4.2.3.3 DETECTOR PARAMETERS: MASS SPECTROMETER
     4.2.3.4 PREPARATION OF STOCK SOLUTIONS
     4.2.3.5 PREPARATION OF CALIBRATION SAMPLES
# TABLE OF CONTENTS

<table>
<thead>
<tr>
<th>Section</th>
<th>Page No.</th>
</tr>
</thead>
<tbody>
<tr>
<td>4.2.3.6 Preparation of the Quality Control Samples</td>
<td>51</td>
</tr>
<tr>
<td>4.2.3.7 Extraction Procedure</td>
<td>51</td>
</tr>
<tr>
<td>4.2.3.8 Validation Procedures</td>
<td>52</td>
</tr>
<tr>
<td>5. Results and Discussion</td>
<td>54</td>
</tr>
<tr>
<td>5.1 Liposome Formulation</td>
<td>54</td>
</tr>
<tr>
<td>5.2 Characterization of Liposome’s</td>
<td>60</td>
</tr>
<tr>
<td>5.2.1 Size and Shape Analysis</td>
<td>61</td>
</tr>
<tr>
<td>5.2.2 Entrapment Efficiency</td>
<td>64</td>
</tr>
<tr>
<td>5.2.3 Chemical Stability of Unentrapped Irinotecan at Different pH</td>
<td>65</td>
</tr>
<tr>
<td>5.2.4 In vitro Release Studies</td>
<td>65</td>
</tr>
<tr>
<td>5.2.5 Stability Studies</td>
<td>67</td>
</tr>
<tr>
<td>5.3 LC-MS/MS Method Development and Optimization</td>
<td>69</td>
</tr>
<tr>
<td>5.3.1 Detector (Mass) Parameter Optimization</td>
<td>69</td>
</tr>
<tr>
<td>5.3.2 Chromatographic Method Development and Optimization</td>
<td>72</td>
</tr>
<tr>
<td>5.3.3 Assay Method Validation</td>
<td>74</td>
</tr>
<tr>
<td>5.4 Bioanalytical Method Development and Validation</td>
<td>78</td>
</tr>
<tr>
<td>5.4.1 Development of Extraction Method</td>
<td>78</td>
</tr>
<tr>
<td>5.4.2 Bioanalytical Method Validation</td>
<td>80</td>
</tr>
<tr>
<td>5.5 Pharmacokinetic Evaluation</td>
<td>97</td>
</tr>
<tr>
<td>5.5.1 Pharmacokinetic Parameters Evaluation</td>
<td>104</td>
</tr>
<tr>
<td>5.6 Toxicity and Toxicokinetic Evaluation</td>
<td>111</td>
</tr>
<tr>
<td>5.6.1 Clinical Observations</td>
<td>111</td>
</tr>
<tr>
<td>5.6.2 Clinical Laboratory Tests</td>
<td>114</td>
</tr>
<tr>
<td>5.6.3 Histopathology</td>
<td>115</td>
</tr>
<tr>
<td>5.6.4 Toxicokinetics</td>
<td>117</td>
</tr>
<tr>
<td>5.7 Organ Toxicity Studies</td>
<td>122</td>
</tr>
<tr>
<td>6. Conclusion</td>
<td>131</td>
</tr>
<tr>
<td>7. Future Directions</td>
<td>134</td>
</tr>
<tr>
<td>8. Reference</td>
<td>135</td>
</tr>
<tr>
<td>9. Appendices</td>
<td>147</td>
</tr>
</tbody>
</table>