**CONTENTS**

**CHAPTER 1: INTRODUCTION**

<table>
<thead>
<tr>
<th>Section</th>
<th>Page No.</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.1 Economic animals</td>
<td>1</td>
</tr>
<tr>
<td>1.1.1 Farm animals</td>
<td>2</td>
</tr>
<tr>
<td>1.1.2 Domestic fowls</td>
<td>2</td>
</tr>
<tr>
<td>1.2 Study site-Southern Assam (Barak valley)</td>
<td>3</td>
</tr>
<tr>
<td>1.3 Species identification</td>
<td>3</td>
</tr>
<tr>
<td>1.3.1 Traditional method</td>
<td>4</td>
</tr>
<tr>
<td>1.3.2 Conventional methods</td>
<td>5</td>
</tr>
<tr>
<td>1.3.3 DNA based method</td>
<td>5</td>
</tr>
<tr>
<td>1.4 Development of molecular based techniques</td>
<td>6</td>
</tr>
<tr>
<td>1.4.1 PCR (Polymerase Chain Reaction) based molecular techniques</td>
<td>6</td>
</tr>
<tr>
<td>1.5 Primers design</td>
<td>7</td>
</tr>
<tr>
<td>1.6 Mitochondrial DNA</td>
<td>8</td>
</tr>
<tr>
<td>1.7 DNA Barcode</td>
<td>10</td>
</tr>
<tr>
<td>1.7.1 History of DNA Barcode</td>
<td>10</td>
</tr>
<tr>
<td>1.7.2 Target genes</td>
<td>10</td>
</tr>
<tr>
<td>1.7.2.1 Ideal DNA Barcoding marker</td>
<td>11</td>
</tr>
<tr>
<td>1.7.2.2 Cytochrome C Oxidase Subunit I (COI) gene</td>
<td>12</td>
</tr>
<tr>
<td>1.7.3 Advances in DNA barcoding</td>
<td>12</td>
</tr>
<tr>
<td>1.7.3.1 GenBank Database</td>
<td>13</td>
</tr>
<tr>
<td>1.7.3.2 BOLD database</td>
<td>13</td>
</tr>
<tr>
<td>1.7.3.3 CBOL (Consortium for the Barcode of Life)</td>
<td>14</td>
</tr>
<tr>
<td>1.8 DNA Barcoding based Restriction Fragment Length Polymorphism</td>
<td>14</td>
</tr>
<tr>
<td>1.8.1 PCR-RFLP</td>
<td>15</td>
</tr>
<tr>
<td>1.9 Statement of Problem</td>
<td>15</td>
</tr>
<tr>
<td>1.10 Objectives</td>
<td>16</td>
</tr>
</tbody>
</table>
CHAPTER 2: REVIEW OF LITERATURE

2.1 Economic animals
2.2 North-East Region (NER) of India
2.3 Molecular Techniques
2.4 Mitochondrial DNA over Nuclear DNA
2.5 Mitochondrial DNA
2.6 DNA Barcode
   2.6.1 Development of primers
   2.6.2 Establishment of DNA Barcode region
   2.6.3 DNA Barcoding of Fishes
   2.6.4 DNA Barcoding of Aves
   2.6.5 DNA Barcoding of Amphibians
   2.6.6 DNA Barcoding of Algae
   2.6.7 DNA Barcoding of Invertebrates
   2.6.8 DNA Barcoding of Mammals
   2.6.9 Nucleotide composition in DNA Barcode region
   2.6.10 DNA Barcode analyses softwares
   2.6.11 DNA Barcode projects
   2.6.12 Mini-barcode
2.7 PCR-RFLP
2.8 Phylogenetic Analysis

CHAPTER 3: MATERIALS AND METHODS

3.1 Materials
   3.1.1 Collection and storage of biological samples
   3.1.2 Chemicals
   3.1.3 Stock reagents
   3.1.4 Working reagents
   3.1.5 Primers
3.1.6 Additional sequence data acquired from database

3.2 Methods

3.2.1 Primer designing of farm animals

3.2.2 Isolation of genomic DNA from biological samples
  3.2.2.1 Isolation of genomic DNA from feathers
  3.2.2.2 Isolation of genomic DNA from hairs
  3.2.2.3 Isolation of genomic DNA from tissue
  3.2.2.4 Isolation of genomic DNA from blood

3.2.3 Agarose gel electrophoresis of the isolated DNA samples

3.2.4 DNA Quantification using spectrophotometric measurement

3.2.5 Amplification of partial COI region of the mitochondrial genome
  3.2.5.1 PCR reaction settings
  3.2.5.2 PCR cycling condition

3.2.6 Agarose gel electrophoresis of PCR products

3.2.7 Purification of PCR products

3.2.8 Sequencing of purified PCR products

3.3 Analysis of data

3.3.1 Editing and aligning of the Raw sequence data

3.3.2 Formatting of sequences

3.3.3 Sequence submission in GenBank and BOLD

3.3.4 Similarity search

3.4 Restriction sites mapping and enzymatic digestion of farm animals
  3.4.1 Restriction sites mapping
  3.4.2 Enzymatic digestion
  3.4.3 Data analysis

3.5 Nucleotide analyses

3.6 Phylogenetic analysis
CHAPTER 4: RESULTS 75-121

4.1 Prevalence of economic animals 75

4.1.1 Prevalence of economic animals in North-East Region (NER) of India 76

4.1.2 Prevalence of economic animals in Assam 76

4.1.2.1 Livestock and Poultry population 78

4.1.3 Prevalence of economic animals in Southern Assam 79

4.2 In-silico primers designing of farm animals 82

4.3 PCR amplification of farm animals and domestic fowls. Submission of the generated sequences in GenBank database 85

4.4 PCR-RFLP of farm animals 85

4.4.1 In-silico Restriction mapping 87

4.4.2 RFLP analyses 89

4.5 Nucleotide analyses 89

4.5.1 Farm animals 89

4.5.1.1 Codon usage 94

4.5.1.2 Nucleotide base and skew composition 100

4.5.2 Domestic fowls 100

4.5.2.1 Codon usage 104

4.5.2.2 Nucleotide base and skew composition 108

4.6 Phylogenetic analysis 109

4.6.1 Farm animals 117

4.6.2 Domestic fowls 121

CHAPTER 5: DISCUSSION 122-129

SUMMARY 130-131

BIBLIOGRAPHY 132-149

APPENDIX