# CONTENTS

## ACKNOWLEDGEMENTS

Page No. i-ii

## ABBREVIATIONS

Page No. iii-v

## LIST OF FIGURES

Page No. vi-vii

## LIST OF TABLES

Page No. viii

## Chapter 1: INTRODUCTION

Page No. 1-5

## Chapter 2: REVIEW OF LITERATURE

Page No. 6-56

2.1 Worldwide Distribution of *Ziziphus* Species

2.2 Molecular Markers and Their Use in Plants

2.3 Morphological Markers

2.4 Biochemical Markers

2.5 Molecular Markers

2.6 Hybridization Based Markers

2.6.1 Restriction fragment length polymorphism (RFLP)

2.6.2 Variable number tandem repeat (VNTR)

2.7 PCR Based Markers

2.7.1 Sequence-arbitrary primers

2.7.1.1 Random amplified polymorphic DNA (RAPD)

2.7.1.2 Arbitrary primed PCR (AP-PCR)

2.7.1.3 DNA amplification fingerprinting (DAF)

2.7.1.4 Inter simple sequence repeats (ISSR)

2.7.1.5 Amplified fragment length polymorphism (AFLP)

2.8 Other Sequence Arbitrary Methods

2.9 Sequence-Dependent Markers

2.9.1 Sequence tagged sites (STS)

2.9.2 Expressed sequence tags (Est’s)
Chapter 3: MATERIALS AND METHODS

3.1 Location of the Experimental sites
3.2 Materials and Equipments
3.2.1 Plant material
3.2.2 Facilities for molecular characterization
3.3 Methods for Analysis
3.3.1 Molecular analysis
3.3.1.1 Isolation, purification and quantification of genomic DNA
3.3.1.2 Optimization of conditions for RAPD
3.3.1.3 Optimization of conditions for ISSR
3.3.1.4 Selection of polymorphic and reproducible markers
3.3.1.5 Individual genotype analysis
3.4 Development of Microsatellite Markers
3.4.1 Nebulization of genomic DNA and ligation of adaptors
3.4.2 Hybridization to biotinylated oligos
3.4.3 Enrichment of library for microsatellites
3.4.4 Ligation and Cloning
3.4.5 Selection of positive/transformed clones
3.4.6 Pooling and amplification of plasmid DNA
3.4.7 Sequencing of selected clones
3.4.8 SSR identification and primer development
3.4.9 Validation of microsatellite primers
3.5 Statistical Analysis
3.5.1 Scoring and diversity analysis
3.5.2 Resolving power (Rp)
3.5.3 Polymorphism information content (PIC)

Chapter 4: RESULTS

4.1 Development of DNA profile using RAPD marker
4.1.1 Optimization of PCR conditions
4.1.2 Band statistics
4.1.3 Resolving Power (Rp) and polymorphism information content (PIC)
4.1.4 Genetic diversity based on RAPD analysis
4.2 Development of DNA Profiles Using ISSR Marker
4.2.1 Optimization of PCR conditions
4.2.2 Band Statistics
4.2.3 Resolving Power (Rp) and Polymorphism Information content (PIC)
4.2.4 Genetic diversity based on ISSR analysis
4.3 Development of New SSR Marker and Its Use
4.3.1 Microsatellite detection
4.3.2 Validation of microsatellite markers
4.3.3 Genetic diversity based on microsatellite markers
4.4 Comparison of RAPD and ISSR Markers to Assess Genetic Diversity
4.4.1 Relationship between Rp and PIC
4.5 Combined Profile of RAPD, ISSR and SSR Markers
4.6 Development of DNA Fingerprinting Combining All the Marker (RAPD, ISSR and Microsatellite) Used