## Chapter 1: Introduction

1. Pharmacogenetics
2. Genetic polymorphism and drug response
3. Applications of Pharmacogenetics
   - 1.3.1 Cancer
   - 1.3.2 Hypertension
   - 1.3.3 Respiratory drugs
   - 1.3.4 Antipsychotic drugs and their receptors and transporters
4. Advantages of Pharmacogenetics
5. Use of Pharmacogenetics in Clinical Practice
6. Aim
7. Objectives

## Chapter 2: Review of Literature

1. Factors Affecting Drug Metabolism
   - 2.1.1 Age and sex factors
   - 2.1.2 Environmental factors
   - 2.1.3 Disease factors
   - 2.1.4 Ethnicity
   - 2.1.5 Genetic variation
2. Genetic Variability Influencing Drug Response
   - 2.2.1 Drug Metabolizing Enzymes
2.2.2 Drug transporters
2.2.3 Drug receptors
2.2.4 Drug targets
2.2.5 Polymorphism-modifying diseases and drug responses
2.2.6 Polymorphisms affecting pharmacokinetics

2.3 Pharmacogenetics of Drug metabolism ....................... 18
2.3.1 Phase-I DMEs-Cytochrome P450 enzymes
2.3.2 Phase-II DMEs

2.4 CYP 2C19......................................................... 23
2.4.1 CYP2C19 gene polymorphism
2.4.2 Clinical significance of CYP2C19 polymorphism

2.5 CYP2C9.......................................................... 26
2.5.1 CYP2C9 gene polymorphism
2.5.2 Clinical significance of CYP2C9 polymorphism

2.6 Genetic basis to concept of Prakrit................................ 29
2.6.1 The science of Ayurveda
2.6.2 Ayurvedic concept of Prakriti
2.6.3 AyuGenomics
2.6.4 Other studies on traditional medicine

2.7 Pharmacogenetics of Methotrexate in Rheumatoid Arthritis......................................................... 36
2.7.1 Epidemiology of RA
2.7.2 Treatment in RA
2.7.3 MTX in RA
2.7.4 Cellular pathway of MTX
2.7.5 Polymorphisms in MTX transporter
2.7.6 Polymorphisms in MTX glutamation genes
2.7.7 Polymorphisms in MTX cellular pathway
2.7.8 Polymorphisms in adenosine pathway
2.7.9 MTX pharmacogenetic studies
Materials and Methods

3.1 Study Population

3.1.1 For CYP2C19 and CYP2C9 genotyping study
3.1.2 Genetic basis for concept of Prakriti study
3.1.3 Pharmacogenetics of MTX in RA
3.1.4 Ethical consideration

3.2 Materials

3.2.1 Collection of blood samples
3.2.2 DNA extraction from whole blood samples
3.2.3 Amplification of DNA isolated from whole blood samples
3.2.4 Restriction Digestion of amplified product
3.2.5 Agarose Gel Electrophoresis
3.2.6 Staining gels
3.2.7 Real time TaqMan allelic discrimination assay

3.3 Pharmacokinetic study

3.3.1 Reagents
3.3.2 HPLC standards
3.3.3 Chromatographic system
3.3.4 Column

3.4 Methods

3.4.1 Prakriti evaluation
3.4.2 Pharmacogenetics of MTX in RA
3.4.3 Clinical management in RA
3.4.4 DNA Extraction
3.4.5 Polymerase Chain Reaction - Restriction Length Fragment Polymorphism
3.4.6 Real time TaqMan allelic discrimination assay
3.4.7 Pharmacokinetics of methotrexate
3.4.8 Statistical analysis
Results and Discussion............................................................................................................. 75
PART 1 DNA isolation and genotyping ................................................................. 78
PART 2 Genetic polymorphism of CYP2C19 and CYP2C9 in
Maharashtrian population ................................................................. 88
PART 3 Traditional Medicine to Modern Pharmacogenomics .......... 103
PART 4 Pharmacogenetics of MTX response (efficacy, toxicity) in
Indian patients suffering from RA ...................................................... 107
Conclusion......................................................................................................................... 138
References......................................................................................................................... 142
Annexure........................................................................................................................... 152
Synopsis