Chapter 1

Introduction
A. INTRODUCTION

Hepatitis C virus (HCV) is the leading cause of most cases of non-A-non-B hepatitis (NANBH) worldwide. The hallmark of the disease is its propensity to evolve into chronicity, probably because, genetic heterogeneity allows the virus to escape immune mediated neutralization. The chronicity may lead to cirrhosis of liver and hepatocellular carcinoma. World health organization (WHO Report, 1999) has estimated 170 million (3%) people of the world are infected with HCV, which highlights HCV as a major human pathogen. It has been estimated that, approximately, 1.8% of the general population are infected with HCV in India till date.

Hepatitis C virus is a novel RNA virus with characteristics of the family Flaviviridae. The virion is spherical, 40-60nm in diameter, with a lipid envelope. HCV genome is a single positive stranded RNA approximately 9.4 kb in length. It contains a single open reading frame (ORF) of ~9kb which encodes a polyprotein of 3033 amino acids. The ORF is flanked by untranslated regions (UTR) at both ends and these are the most conserved regions of the genome. The 5’UTR has characteristic secondary structures and plays a crucial role in translation initiation (Liang et al, 2000). The nascent viral polyprotein is processed by a combination of host and viral proteinases into the mature viral proteins. The viral structural proteins are core or capsid and envelope glycoproteins E1 and E2, which are major components of prototype vaccine studies for HCV (Liang et al, 2000).

The non-structural proteins of HCV are NS2, NS3, NS4A, NS4B, NS5A and NS5B. Among these, NS2 has some proteolytic function. NS3 is a
serine protease. NS4A acts as a cofactor for the NS3 protease. NS4B is crucial for HCV replication complex formation. NS5A is a critical factor in determining the susceptibility of the virus to interferon treatment. NS5B codes for the RNA-dependent RNA Polymerase, essential for virus replication (Liang et al, 2000).

Being an RNA virus, HCV has a high mutation rate which contributes to its rapid evolution resulting in the appearance of escape mutants, changes in virulence and host range and antiviral drug resistance. Since the first detection of HCV in 1989, at least 6 major genotypes and more than 50 subtypes have been identified based on sequence variation within the highly conserved 5' UTR and the relatively well conserved core and NS5 region (Simmonds et al, 1993; Simmonds, 1995). The genotypes differ in nucleotide sequences by 31-34% and the subtypes differ by 20-23%. The importance of genetic heterogeneity lies in the fact that some genotypes appear to be associated with more severe pathology and are more refractory to current therapies. HCV genotyping is now widely used for clinical purposes in acute and chronically infected patients as a predictive marker of disease progression and response to interferon therapy (Roffi et al, 1998). The prototype strain 1a is prevalent in USA and Europe, 1b is found worldwide, genotype 2 in USA and Europe, 3 in the Indian subcontinent, 4 in Africa and Middle East, 5 in Africa and 6 in South East Asia. Almost 5-10% of the infections are of multiple genotypes (Liang et al, 2000).

In absence of development of protective immunity after primary infection has made the development of a vaccine difficult. The other problems in this regard are the absence of a suitable tissue culture system for
cultivation of HCV and absence of any animal model other than chimpanzee (Liang et al, 2000).

The suitability of a proper control regime for a pathogen in a particular population may be evaluated by examining the molecular characteristics of a representative number of circulating strains in that geographical region. There was no such study of hepatitis C virus genotypes in eastern India, although some reports are available from southern India, western India and northern India (Issar et al, 1995; Sarin et al, 1996; Panigrahi et al, 1997; Sawant et al, 1999; Sood et al, 1999; Amarapurkar et al, 2001; Mathai et al, 2002; Das et al, 2002; Raghuraman et al, 2003). The increasing prevalence of HCV in the general population and different risk groups in India necessitated further molecular biological investigations to reveal the molecular characteristics of the HCV strains circulating in India.

So, the aim of this study was to detect different types of HCV strains circulating in this part of the globe and delineate their diversity in the population. This study was initiated with these specific questions to be answered: 1) What are the predominating genotypes of Hepatitis C Virus in Eastern part of India, especially in West Bengal? 2) What are the unique features of the HCV genome in the strains circulating in this part of the country?

Since this study is the first of its kind in this geographical region, it will add relevant information to the global scenario of HCV infection and will be crucial for future antiviral drug therapy formulations.
**OBJECTIVES:**

This study was initiated keeping in mind two main objectives, which are,

1) Genotyping of HCV in different risk populations and in general population.

2) Comparative partial genome analysis of selected isolates showing interesting features from Eastern India.

The molecular epidemiology of genotypic distribution of HCV and the comparative genome analysis of some parts of the HCV genome is expected to reveal significant data which will be important to assess the quasi-species variations of HCV in this region.