INTRODUCTION

The 5'-untranslated region (5'-UTR) of the Hepatitis C virus (HVC), with 341 nucleotides in length (1), exhibits a high degree of conservation when the complete region is compared between two isolates from the same country. However, the heterogeneity increases when isolates from different countries are compared (2).

At least four different genotypes and related subtypes are described based on the 5'-UTR (3), but only a limited number of nucleotide sequences are currently available. The aims of this work were to sequence the 5'-UTR of HCV Cuban carriers to align it with representative HCV type sequences and to typify the circulating Cuba genotype(s) of the virus.

MATERIALS AND METHODS

RNA was extracted from plasma of 25 Cuban HCV carriers by the guanidinium-thyocyanate method (4). The cDNA-PCR was carried out using procedures employed in our laboratory (5). The 229 bp amplified fragment corresponded to nucleotides -260 to -32 upstream the major open reading frame (1). The direct cycle sequencing reactions were performed employing the same primers as for nested-PCR following an asymmetric-PCR standardization procedure to select the PCR-like sequencing conditions (6). Clustal V program was used for alignment of Cuban sequences with the reported HCV sequences. Potential contamination during reverse transcription, nested-PCR amplification or sequencing reactions were ruled out by means of negative control, introduced for each tested sample.

RESULTS AND DISCUSSION

All the 186 bp nucleotide sequences from studied HCV Cuban carriers were identical, which is not surprising
considering the high degree of conservation of the 5'-UTR.

Alignment of representative sequence of types 1a (HCV-1), 1b (HCV-2), 2a (HC-J6), 2b (HC-J8), 3a (E-b1) and 4 (EG-1) of HCV (3), revealed total homology (100%) with 1b type of HVC. Figure 1 shows the 5'-UTR nucleotide sequence from Cuban isolates. Our work agrees with other report based on the Okamoto's method on the core region, which showed type II HCV as main genotype (92.9%) among HCV-infected Cuban patients (Padron, G. Personal Communication).

\[-239\]
\[
\text{GTGCAGCCCTCCAGGACCCCCCCCCTTGTGAATTGCACGGAGACGCCGGGTCTTTCTTTTGGATCAACCCGCTCAATGCTGAGATGGGAGACTGGCTAGCCGAGTAGGTTGAGGTGTTGGTGGTCGCAAGGCCAGCTTGTGGTA}
\]
\[-54\]

**Fig. 1** Nucleotide sequence of cDNA-PCR amplified product from 5'-UTR in HCV Cuban carriers.

According to our results, type 1b [counterpart of type II (3)] is the predominant genotype circulating in Cuban.

Taking into account the small number of serum tested, we do not excluded that other less represented genotypes are also present in Cuba.

**REFERENCES**


Copyright 1995 Sociedad Iberolatinoamericana de Biotecnologia Aplicada a la Salud

Contact: [Biotecnologia Aplicada](mailto:bioline@bioline.org.br)