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# Changing Hepatitis C genotypes in South Africa including a closer look at genotype 5a

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Around 1 million South Africans are infected with Hepatitis C virus. Most individuals remain undiagnosed until symptoms occur and, therefore, usually present with late stage liver disease. The standard of care (SOC) for treating hepatitis C in South Africa is combination therapy with pegylated interferon and ribavirin. However, not all South Africans have access to (or are able to benefit from) therapy due to high costs, poor adherence, adverse side effects and advanced stage of disease. HCV genotypes and/or mutations in the core/ non-structural regions have been associated with response to therapy and/or disease progression. Although the major genotype in South Africa is genotype 5a, studies using clinical cohorts confirm that other local genotype frequencies are increasing due to population migration and travel. This retrospective study describes (1) changing genotype frequencies within a South African high-risk clinical patient group as well as a low risk (asymptomatic) cohort of blood donors bled between the years 2008-2012and (2) molecularly characterize the core and NS5B regions of all pre-treatment samples.

**Methods:** Sera samples from hospitals around South Africa (N=865) and from the South African Blood Services (SANBS, N=206) were analyzed quantitatively by real-time PCR and genotyped using the Versant LiPA assay. Mutational analyses were performed for selected genotype 5a samples as follows: Thirty-one samples were sequenced directly in the core region (patients N=21 and blood donors N=10) while retrospective data from 43, previously sequenced, patient samples was analyzed in the NS5B region. Mutations were identified using Mega 6.0. Sequences were compared to GenBank references for amino acids 1-318 of core and E1 and amino acids 2661-2728 of NS5B. Sequence variability was examined for known CD4+ and CD8+ T-cell epitopes and their predicted binding to HLA types known to be prevalent in the South African population determined using IEDB.

**Results:** Genotype 5a (35.83%) was the major genotype in the patient cohort whilst genotype 1 (33.98%), was found to predominate in the blood donors. An increase in genotypes 3 and 4 was noted over the 5 year study period in both cohorts.

**The core:** R70Q mutation (associated with poor response to pegylated interferon and ribavirin therapy in genotype 1b patients and progression to HCC) was identified in 20/21 patient samples with genotype 5a and 9 of 10 genotype 5a blood donor samples. The NS5B:S282T mutation (associated with resistance) was not seen at baseline in any of the genotype 5a samples studied. Six of the 9 known HLA-A02 restricted epitope sequences showed high-intermediate cross-reactivity (binding scores of <300 IC50nM) to two of the most common alleles present in the South African population. No known CD8+ T-cell epitopes were mapped to the sequenced NS5B region.

**Discussion:** The study highlights the need for ongoing genotyping surveillance of HCV in South Africa to monitor changing frequencies and their impact on health care costs and burden of disease. It also broadens our knowledge and provides new insight into the diversity of HCV in pre-treatment samples belonging to the poorly studied genotype 5a which predominates in South Africa.

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