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Treatment of Chronic Hepatitis C Genotype 3 With Ledipasvir and Sofosbuvir: An Observational Study

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Objective: **Sofosbuvir/ledipasvir (SOF/LED) is recommended for treatment of genotypes 1, 4, 5 and 6. Despite some preliminary data from the ELECTRON-2 trial regarding use of SOF/LED combination in chronic hepatitis C genotype 3, there are no guidelines recommending this combination in such patients. We conducted this study to evaluate the efficacy of the overall sustained virologic response at 12 weeks (SVR 12) and safety of SOF/LED in chronic hepatitis C genotype 3 infection in our population.** ***Methods:*** **It was a prospective, hospital-based observational study. All patients with chronic hepatitis C genotype 3 treated with SOF/LED were divided into two groups: patients with cirrhosis and without cirrhosis. Patients without cirrhosis received SOF/LED (90/400 mg) for 12 weeks; however, patients with cirrhosis received treatment for 24 weeks.** ***Results:*** **We enrolled 104 patients with chronic hepatitis C over a period of 24 months. Of the total, 66 were women (63.5%) and 38 were men (36.5%). The average age was 40 years (range: 18–76 years). Of 104 patients, 86 (82.7%) were of genotype 3, 15 (14.9%) were of genotype 1 and 3 (2.9%) were of genotype 4. Ninety-two (88%) were noncirrhotic and 12 (11.5%) were cirrhotic. Ninety-five (95.2%) were treatment naõve. Among genotype 1 and 4, all patients achieved rapid virologic response and SVR 12. Of 86 genotype 3 patients, 78 (90.6%) were noncirrhotic and 8 (9.3%) were cirrhotic. Among genotype 3 patients without cirrhosis, 75 (96%) achieved SVR 12 while 6 (75%) with cirrhosis achieved SVR 12. All patients tolerated the combination well; however, some patients experienced nausea (26%), headache (25%) and fatigue (21%). No patient had to discontinue therapy due to adverse drug reactions.** ***Conclusions:*** **Single tablet LED and SOF combination is safe and effective in genotype 3 patients without cirrhosis even without ribavirin. Being effective in genotype 3, the combination can be used as a pangenotypic drug in patients without cirrhosis. (J Clin Exp Hepatol xxxx;xxx:xxx)**

Hepatitis C virus (HCV) is a major cause of mortality and morbidity globally.[1] Approximately 71 million individuals are chronically infected worldwide.[2,3] Globally, G1 accounts for 49.1% of all anti-HCV infections, followed by genotype 3 (17.9%), genotype 4 (16.8%), genotype 2 (11.0%), genotype 5 (2.0%) and genotype 6 (1.4%).[4] The prevalence of genotype 3 in South and Central Asia is 71.6% of all HCV infections representing the most common genotype in that region.[5] Genotype 3 infection represents a unique entity, with higher rates of steatosis, more rapid progression of fibrosis and hepatocellular carcinoma.[6–8] The cure rates of the overall sustained virologic response at 12 weeks (SVR 12) for genotype 3 infection have lagged behind those of other genotypes.[9,10]

Till date, no approved vaccine is available which protects against contracting hepatitis C.[11] Treatment with antiviral medication is recommended in all people with proven chronic hepatitis C who are not otherwise at high risk of dying from other causes. People with the highest complication risk should be treated first.[12] Before 2011, the standard treatment of HCV infection was combination therapy for 24 or 48 weeks with peginterferon/ribavirin. These drugs were associated with serious adverse effects such as haemolysis and bone marrow suppression with ribavirin and flu-like symptoms and bone marrow suppression with interferon.[13]

Ledipasvir (LED) is a potent inhibitor of HCV NS5A, a viral phosphoprotein that plays an important role in viral replication, assembly and secretion. Sofosbuvir (SOF) is a nucleotide analogue inhibitor of hepatitis C virus NS5B polymerase—the key enzyme mediating HCV RNA replication. The fixed-dose combination LED-SOF (90 mg/400 mg) is FDA approved for the treatment of chronic hepatitis C genotypes 1, 4, 5 and 6 in both treatment-naive and treatment-experienced patients. Available data from clinical trials have demonstrated that the combination of LED-SOF has been very well tolerated. The most common reported adverse effects are fatigue and headache.[14]

Despite some preliminary data from the ELECTRON trial regarding use of Ledipasbir and SOF combination in chronic hepatitis C genotype 3, there are no guidelines recommending this combination in such patients.

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Abbreviations: G: Genotype; HCV: Hepatitis C Virus; LED: Ledipasvir; RNA: Ribonucleic Acid; RVR: Rapid Virologic Response; SOF: Sofosbuvir; SVR: Sustained Virologic Response
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The prevalence of HCV in Kashmir is 1.9%, and genotype 3 is the most common genotype.[15] Considering the case with which a patient with chronic hepatitis can have a single tablet of fixed combination of Ledipasbir and SOF, we decided to study efficacy and safety in chronic hepatitis C genotype 3 with this combination.

This study was conducted in the department of gastroenterology at a tertiary care hospital in North India over a period of two years. Formal informed consent was obtained from patients included in the study. This study was started after getting clearance from the institutional ethical committee. The diagnosis of hepatitis C virus infection by anti-HCV antibody was performed by using an enzyme-linked isoimmuno assay technique. For patients whose anti-HCV antibody was positive, blood sample was taken for HCV RNA viral load by using polymerase chain reaction (PCR). A baseline HCV RNA viral load is taken by using a PCR-based technique before starting treatment. HCV RNA was measured with the COBAS AmpliPrep/COBASTaqMan HCV Test, version 2.0 for research use only (Roche, Indianapolis, Indiana), with a lower limit of quantification for HCV RNA of 15 IU/mL. HCV genotype and subtype were determined using the Siemens VERSANT HCV Genotype INNO-LiPA 2.0 Assay.

Genotype determination was performed by reverse transcriptase PCR. After genotyping is performed, patients were included in the study protocol. After starting treatment, viral loads were measured at 4 weeks and 12 weeks after treatment. If the viral load is undetectable 12 weeks after treatment, it was considered a sustained virological response (SVR).

INCLUSION CRITERIA

The inclusion criteria for the study were as follows:

- Patients with chronic hepatitis C genotype 3 were included in the study.
- Patients both with cirrhosis and without cirrhosis were included in the study.
- Patients of relapse on other regimens of chronic hepatitis C were included in the study.
- Both male and female patients were included in this study.

EXCLUSION CRITERIA

The exclusion criteria for patients in this study were as follows:

- Simultaneous infection with hepatitis B or human immunodeficiency virus (HIV).
- Evidence of liver disease because of other aetiology.
- Pregnancy

STATISTICAL ANALYSIS

It was a prospective, hospital-based observational study. All patients with chronic hepatitis C genotype 3 were enrolled into two groups: patients without cirrhosis (group A) and patients with cirrhosis (group B).

Both groups received LED/SOF (90/400 mg) for 12 weeks; however, treatment was extended to 24 weeks in the case of treatment-experienced patients with cirrhosis. The data were analysed by SPSS 22 software.

RESULTS AND OBSERVATIONS

We enrolled 104 patients with chronic hepatitis C over a period of 24 months. The average age was 40 years (range: 18–76 years) Table 1. Of the total, 66 (63.5%) were women and 38 (36.5%) were men. Ninety-six (92%) were married. Ninety-two (88%) patients were noncirrhotic. Of 104 patients, 86 (82.7%) were genotype of 3, 15 (14.9%) were genotype of 1 and 3 (2.9%) were genotype of 4, Table 2. Ninety-five (95.2%) were treatment naöve. Of 86 patients with genotype 3, 78 (90.6%) were noncirrhotic and 8 (9.3%) were cirrhotic. Among genotype 1 and 4, all patients achieved rapid virologic response (RVR) and SVR 12, whereas 81 of 86 (94%) with genotype 3 achieved SVR. Among genotype 3 patients without cirrhosis, 75 (96%) achieved SVR

Table 1 Descriptive Statistics of Baseline Parameters.

Parameter	Mean ± SD
Age	40 ± 13.6 years
Bilirubin	1.08 ± 1.0 mg/dl
ALT	86 ± 51.0 IU/L
Albumin	4.2 ± 0.34 gm/dl
Creatinine	0.98 ± 0.86 mg/dl
INR	1.07 ± 0.09
HCV RNA	1833141 ± 3374633 copies/L
Fibroscan	7.9 ± 4.7 kPa
Platelets	158 ± 52 × 10^3/microL

SD, standard deviation; HCV, hepatitis C virus; ALT, alanine transaminase; INR, international normalized ratio.

Table 2 Showing SVR 12 Among Various Genotypes. Overall 81 (96%) Genotype Patients Achieved SVR 12.

Genotype * SVR 12 association			
Genotype	SVR 12		Total
	Achieved	Not achieved	
Genotype 1	15	0	15
Genotype 3	81	5	86
Genotype 4	3	0	3
Total	99	5	104

SVR 12, overall sustained virologic response at 12 weeks.

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Table 3 Showing Genotype-Wise SVR 12 and Cirrhotic Status.

SVR 12			Genotype			Total
			Genotype 1	Genotype 3	Genotype 4	
Achieved	Cirrhotic	No	12	75	2	89
		Yes	3	6	1	10
	Total		15	81	3	99
Not achieved	Cirrhotic	No		3		3
		Yes		2		2
	Total			5		5
	Total		15	86	3	104

SVR 12, overall sustained virologic response at 12 weeks.

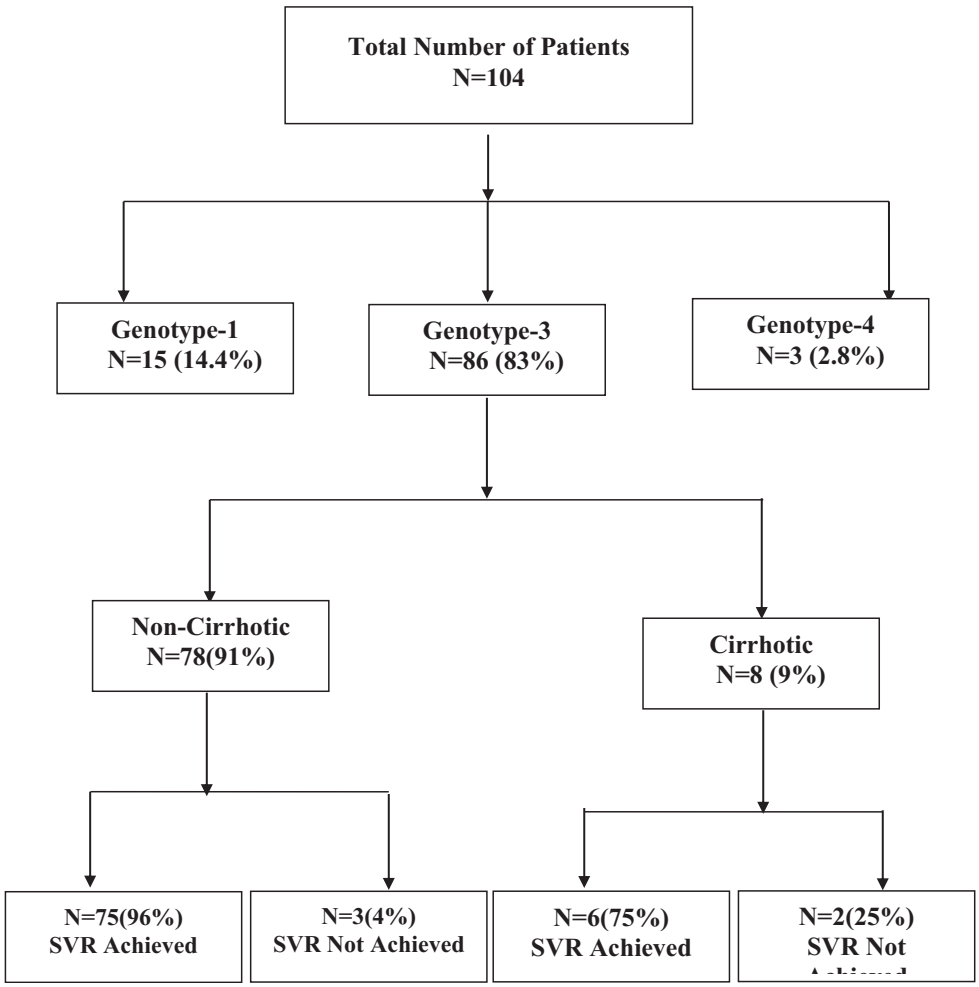
12 while 6 (75%)with cirrhosis achieved SVR 12, Table 3. All patients tolerated the combination well; however, some patients experienced nausea (26%), headache (25%) and fatigue (21%). No patient had to discontinue therapy due to adverse drug reactions.

Genotype 3 HCV accounts for approximately 20% of all HCV infections globally and 40% of infections in Asia.[16] The genotype 3 HCV is associated with increased risk of steatosis,[17] rapid progression of fibrosis,[18] hepatocellular carcinoma[19,20] and all-cause mortality and morbidity.[19]

SOF is an inhibitor of the HCV NS5B polymerase with a good safety profile and a high genetic barrier to resistance.[21,22] SOF is a pangenotypic nucleotide polymerase inhibitor with potent activity against all 6 HCV genotypes proven in both *in vitro* replicon assays and extensive clinical use. LED is a potent NS5A inhibitor with activity against replicons of genotypes 1a, 1b, 4, 5 and 6. However, LED is less active against genotype 3a HCV *in vitro*.[23,24] Hence, one might expect less susceptibility of genotype 3 virus to LED.

SOF has been approved in combination with other directly acting antivirals (DAAs) for the treatment of HCV of all genotypes.[21,22] LED-SOF, with or without

Patient Layout Chart



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ribavirin, has been approved for patients with genotypes 1, 4, 5 and 6 as a 12-week, once-daily all oral treatment.[14] The elimination of ribavirin from the treatment of HCV is desirable due to the adverse events associated with ribavirin therapy. Current guidelines do not recommend the use of LED and SOF, with or without ribavirin, in patients with HCV genotype 3 infection.[25,34]

Feld *et al.* [26] enrolled 111 treatment-naive genotype 3 patients with and without compensated cirrhosis and treated with LED-SOF (90 mg and 400 mg) plus weight-based ribavirin for 12 weeks. Of the 111 patients enrolled, 105 (95%) had subtype 3a HCV and 39 (35%) had compensated cirrhosis. SVR 12 was achieved by 99 of 111 patients (89%; 95% confidence interval, 82%–94%). Of the 39 patients with cirrhosis, 31 (79%) achieved SVR 12, compared with 68 of 72 (94%) patients without cirrhosis.

In the ELECTRON-2 study, treatment-naive patients with genotype 3 infection were randomized to receive 12 weeks of LED and SOF with or without ribavirin27. In the arm without ribavirin, 16 of 25 (64%) achieved SVR 12, whereas all 26 patients randomized to receive therapy with ribavirin achieved SVR 12, including 6 patients with compensated cirrhosis. These results clearly show that ribavirin is important but also suggest that LED is more active against genotype 3 HCV than predicted based on the replicon data alone.

In the FISSION trial, treatment-naive patients with genotype 2 or 3 were randomized to SOF and ribavirin for 12 weeks or peginterferon and ribavirin for 24 weeks.[28] Among patients with genotype 3 infection who received SOF and ribavirin for 12 weeks, only 61% (89/145) of those without cirrhosis and 34% (13/38) of those with cirrhosis achieved SVR 12. It was only with extension of therapy to 24 weeks in the VALENCE trial that higher rates of SVR 12 were achieved in patients with genotype 3 infection: 92% (12/13) with cirrhosis and 93% (86/92) without cirrhosis.[29] However, in our study, SOF and LED combination given for 12 weeks was effective in patients with genotype 3 infection.

In our study, of patients with all genotypes, 86 (100%) achieved RVR. SVR 12 was achieved in 81 (94%) patients. Seventy five of 78 patients without cirrhosis (96%) while as 6 of 8 with cirrhosis (75%) with genotype 3 achieved SVR 12. Our results were better than those shown by Gane EJ *et al.*[27] in the non-ribavirin group (96% vs 64%) and however were comparable with those of Feld *et al.*[26]

There are several possible explanations for the better than expected activity of LED in patients with genotype 3 infection. Firstly, there may be ethnic and genetic differences between our patients and those of other studies. Secondly, despite the relatively higher 50% effective concentration values for LED against the genotype 3 replicons than replicons of other genotypes, LED is highly concentrated in the liver in animal pharmacokinetic studies, and local intrahepatic concentrations may well reach adequate levels to achieve antiviral activity. Alternatively, it may be that the replicon is an inadequate system to measure all antiviral properties of a given agent. This may be particularly relevant for NS5A inhibitors for which the mechanism of action remains poorly understood. Cell culture assays that can assess all stages of the life cycle are limited, particularly for genotype 3 HCV; however, recent advances that allow replication of serum-derived virus may allow a deeper investigation into the activity of LED against genotype 3 HCV.[29]

Although it is challenging to compare across trials, if LED was as inactive as the replicon data suggest, one would have expected similar results to those seen with SOF and ribavirin alone for 12 weeks. Since completion of this trial, studies of velpatasvir, a pangenotypic NS5A inhibitor with picomolar activity against genotype 3 HCV, have been completed, leading to its recent approval in a fixed-dose combination with SOF in patients of all genotypes.[30–32] In particular, the combination of SOF and velpatasvir in genotype 3 patients led to an SVR 12 rate of 98% (160/163) in treatment-naive patients without cirrhosis and 93% (40/43) in those with cirrhosis.[33]

The present study has some relevant limitations. It was an observational study with no control group; however, >100 patients were included, leading to relatively robust estimates of safety and efficacy. The number of patients with cirrhosis was less, so no strong inference can be drawn.

In this study, treatment with SOF and LED was effective for patients with genotype 3 HCV without cirrhosis and less so for patients with cirrhosis. Although LED and SOF combination is not a recommended treatment for genotype 3 HCV infection by international guidelines, these data suggest that it could be an alternative treatment in settings where preferred treatments are not available/tolerated/contraindicated, especially in patients without cirrhosis.

AUTHORS' CONTRIBUTION

Gulzar Ahmad Dar : Data collection, compilation, methodology, writting; Ghulam Nabi Yattoo : Conceptualization., methodology; Ghulam M. Gulzar ,: Data curation, writing; Jaswinder Singh Sodhi : Script writing. Data collection; Suresh Gorka: Script writing. Data collection; Mushtaq Ahmad Laway: Script writing. Data collection.

CONFLICTS OF INTEREST

The authors have none to declare.

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