

Hepatocellular Carcinoma in Hepatitis C-Associated Cirrhotic Patients Treated With Different Combinations of Direct-Acting Antiviral Agents Available in Pakistan: A Prospective Observational Study

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Abstract

Background

Hepatitis C virus (HCV) is a significant cause of chronic liver disease, which can result in cirrhosis and hepatocellular carcinoma (HCC). Direct-acting antiviral agents (DAAs) are associated with a significant rise in sustained virologic response and reduced treatment duration. We performed this study to assess the incidence of HCC in patients with HCV-associated cirrhosis following therapy with DAAs available in Pakistan.

Methods

We conducted a prospective observational study at Isra University Hospital Hyderabad from July 2019 to August 2020. Three hundred fourteen patients met the inclusion criteria and were included. We recorded baseline demographic characteristics, Child-Pugh class, model for end-stage liver disease (MELD) score, alpha-fetoprotein level, and abdominal ultrasound/computed tomography (CT) scan before and after treatment. We also noted the duration of DAA treatment and the types of DAA used. HCC was considered present according to characteristic imaging findings on abdominal ultrasound/CT scans.

Results

A total of 314 patients were enrolled in the study. The mean age of patients who developed HCC was 46.7 years \pm 10.3 years. Of the patients who developed HCC, 20 (69%) were male, and nine (31%) were female ($p=0.221$). Five patients who developed HCC also had diabetes (17.2%; $p=0.174$) and 17 (58.6%) were smokers ($p=0.001$). Among patients treated with a combination of sofosbuvir/daclatasvir, 20 (69%) developed HCC. Nine patients (31%) treated with a combination of sofosbuvir/velpatasvir developed HCC ($p=0.1$). Eight patients younger than 40 years (27.6%) developed HCC, and 21 patients aged 40 years or older (72.6%) developed HCC ($p=0.55$). HCC was common in Child-Pugh class A patients ($n=19$; 65.6%). Twenty-three patients (79.3%) with a MELD score <9 developed HCC. Eight percent of patients treated with sofosbuvir/daclatasvir for 12 weeks developed HCC, and 15% developed HCC after 24 weeks of treatment with sofosbuvir/daclatasvir. When treated with sofosbuvir/velpatasvir for 12 and 24 weeks, 4% and 9% developed HCC, respectively. Alpha-fetoprotein was elevated in all patients diagnosed with HCC, while only 14.7% of those with no HCC had elevated alpha-fetoprotein levels.

Conclusion

DAAs were associated with an increased risk of HCC. HCC was more commonly seen in patients treated with a combination of sofosbuvir/daclatasvir than sofosbuvir/daclatasvir.

Background

Hepatitis C virus (HCV) may have an important association with liver disease, with over 170 million infections worldwide. HCV, recognized in 1989, is an enveloped ribonucleic acid virus belonging to the

Flaviviridae family.¹ HCV results in cirrhosis in approximately 16% of patients after two decades. Hepatocellular carcinoma (HCC) develops in up to 5% of cirrhotic patients annually. Moreover, patients with hepatic decompensation have up to a 20% chance of death within one year.²

HCC, a major liver cancer, is the second leading cause of cancer death worldwide. The presence of cirrhosis, diabetes, advanced age, and genotype three are risk factors for developing HCC.³

A structural understanding of HCV enzymes (proteases/polymerase) has permitted drug design to create inhibitors to these proteins. The HCV NS2-3 and NS3-4a proteases and other enzymes are vital for its multiplication and drug development. Therefore, direct-acting antiviral agents (DAAs) targeting proteins, like nucleoside/nucleotide analogs and NS3 protease inhibitors, were made. DAAs allow for a sustained virologic response (SVR) in nearly 100% of patients compared with SVRs from interferon (IFN)-based regimens, which only produced SVR in 40–50% of patients.^{4–6} To prevent severe complications due to the related to chronic hepatitis C, HCV clearance is of utmost clinical importance.^{7–10}

Recent studies have shown an overall increase in HCC when patients were managed with DAAs. Reigh et al. reported an unexpected increase of 27.6% in HCC recurrence when HCV patients were treated with DAAs following locoregional treatment of HCC.¹¹ Kozbial et al. showed an increased incidence of HCC of about 6.6%,¹² and Cardoso et al. showed an increased incidence of about 7.4% in HCC after the use of DAAs in the first year.¹³ Conti et al. showed an incidence of 3.16% and a recurrence of 28.81% of HCC in cirrhotics resulting from hepatitis C managed with DAAs.¹⁴

However, some others reported reduced HCC in HCV infected individuals. A study in China by Zeng et al. showed different results, indicating a slight decrease in the incidence rate of HCC.¹⁵ Kanwal et al. also reported a decreased incidence of HCC (< 1%) after the use of DAAs.¹⁶

Methods

Given the conflicting reports in the literature on the incidence of HCC following treatment with DAAs and the lack of data on patients in Pakistan, we performed this study to determine the incidence of HCC in patients with HCV-associated cirrhosis following therapy with DAAs available in Pakistan.

We conducted a prospective observational study at Isra University Hospital Hyderabad in Pakistan. Sample size was calculated using statistical sample size calculator OpenEpi which is available online, based on previous estimates of HCC during the 24-week post-treatment period in HCV patients (7.6%)²⁴ with margin of error 3% and 95% confidence level. Therefore, a sample size of 300 patients was calculated, but a total 314 patients were enrolled over one year. The study included HCV-associated cirrhotic patients of any gender aged 18 to 70 years. Institutional review board approval was taken before conducting the study. After getting informed written consent, patients meeting inclusion criteria were recruited in study. The patients were treated with a combination of sofosbuvir/daclatasvir with ribavirin or a combination of sofosbuvir/velpatasvir with ribavirin based on their affordability. We recorded baseline

demographic characteristics, Child-Pugh class, model for end-stage liver disease (MELD) score, alpha-fetoprotein, and abdominal ultrasound/computed tomography (CT) findings. Patients were excluded from the study if they were diagnosed with HCC prior to the inclusion, or if they were coinfecting with hepatitis B virus or hepatitis D virus and had cirrhosis resulting from other causes, such as Wilson's disease or hemochromatosis or if they did not provide consent to be included in the study. Data were collected from patients who achieved SVR within six months of completion of therapy. We also recorded the duration of DAA and type of DAA used. HCC was considered present according to characteristic imaging findings on abdominal ultrasound/CT scans.

Statistical analysis

IBM SPSS Version 24.0. was used for entering and analyzing data. We calculated the mean and standard deviation for age, Child-Pugh class, MELD score, alpha-fetoprotein levels, and DAAs treatment duration. Frequency and percentage were computed for gender, cirrhosis, and HCC presence. Confounders like age, sex, MELD score, duration of DAA treatment, Child-Pugh score, smoking, diabetes, and the presence of compensated and decompensated cirrhosis were controlled through stratification. Data was considered significant with p -value < 0.05 .

Results

Three hundred and fourteen patients were included in the study. Descriptive demographics are presented in Table 1. Twenty-nine patients (20 men, nine women) developed HCC (9.2%), and their mean age was 46.7 ± 10.3 years ($p = 0.221$).

Table 1
Patient demographic and treatment data

Parameter	Developed HCC	No HCC	P-value
Age (Mean ± SD)	46.7 ± 10.3	44.4 ± 11.5	
A-fetoprotein (Mean ± SD)	876.51 ± 142.87	21.97 ± 89.9	
Male	20 (69%)	163 (57%)	0.221
Female	9 (31%)	122 (42.8%)	
Diabetes	5 (17.2%)	28 (9.8%)	0.174
Smoker	17 (58.6%)	12 (4.2%)	0.001
DAA used			
Sofosbuvir/Daclatasvir	20 (69%)	151 (53%)	0.1
Sofosbuvir/Velpatasvir	9 (31%)	134 (47%)	
Age group			
< 40 years	8 (27.6%)	94 (33%)	0.55
≥ 40 years	21 (72.4%)	191 (67%)	
Child-Pugh class			
A	19 (65.6%)	175 (61.4%)	0.77
B	9 (31%)	91 (31.9%)	
C	1 (3.4%)	19 (6.7%)	
MELD-score			
< 9	23 (79.3%)	229 (80.4%)	0.89
≥ 10	6 (20.7%)	56 (19.6%)	
Abbreviations: HCC, hepatocellular carcinoma; SD, standard deviation; DAA, direct-acting antiviral agent; MELD, model for end-stage liver disease.			

Of those who developed HCC, five (17.2%) had diabetes ($p = 0.174$), and 17 (58.6%) were smokers ($p = 0.001$). Twenty patients with HCC (69%) had been treated with a sofosbuvir/daclatasvir combination, and the other nine patients (31%) had been treated with sofosbuvir/velpatasvir combination ($p = 0.1$).

Eight patients (27.6%) younger than 40 years developed HCC, and 21 patients (72.6%) age 40 or older developed HCC ($p = 0.55$). HCC was commonly seen in Child-Pugh class A ($n = 19$; 65.6%) with no

statistically significant difference from other classes. Twenty-three patients (79.3%) with MELD score < 9 developed HCC.

Percentages of HCC according to the duration of treatment is shown in Fig. 1. Eight percent of Group A patients (sofosbuvir/daclatasvir) developed HCC at 12 weeks, and 15% developed HCC at 24 weeks. Four percent of Group B patients (sofosbuvir/velpatasvir) developed HCC at 12 weeks, and 9% developed HCC at 24 weeks. A total of five patients had elevated alpha-fetoprotein levels but were not diagnosed with HCC.

Discussion

HCC could be a complication related to HCV infection. HCC development is a slow process and depends on the duration of the illness and viral genotype.¹⁷ HCV treatment aims to abate the disease, decline the speed of transmission, and reduce the chances of advancement to HCC.¹⁸ DAAs have arisen as management choices with high SVR.¹⁹ Despite the appearance of exceedingly successful treatment, HCC remains a concern. Studies have demonstrated that up to 8% of patients with HCV-infected cirrhosis develop HCC annually.²⁰

Our results showed a slight increase in HCC incidence after DAA therapy. However, this finding does not align with Zeng et al.,¹⁵ who reported a reduced incidence of HCC after DAAs therapy. Our findings also conflicted with the results reported by Kanwal et al., who reported that DAAs treatment reduced HCC incidence.¹⁶ Our findings correlate with those reported by Kozbial et al., Ravi et al. and Cardoso et al., all of whom showed an increased risk of HCC with use of DAAs.^{12,13,21} Kozbial et al. and Cardoso et al. used an extended follow-up period,^{12,13} while Ravi et al. had follow-up similar to this study.²¹

We found a higher incidence of HCC when patients were treated with sofosbuvir/daclatasvir compared to sofosbuvir/velpatasvir, which contradicts the current literature, where reports indicate DAA therapy decreases, or, at least, has no effect on increasing the chance of HCC.²²⁻²⁴ Furthermore, one study found that sofosbuvir containing regimens without ribavirin were related to an HCV risk more than five times beyond those containing other combinations.²⁵ Meta-analysis of forty-one studies reported no proof on HCC incidence those getting DAAs compared interferon therapy.²⁶

DAA-associated hepatocarcinogenesis has been reported. Yang et al. found HCC relapse earlier when treated with DAAs.²⁷ Another study found an increased incidence of HCC following DAA treatment in those previously having multiple HCC treatments.²⁸ We found an increased incidence of HCC in Child-Pugh class A patients, which does not align with reports in the literature. Calvaruso et al. studied the incidence of HCC in patients with HCV-associated cirrhosis treated with DAAs and found that HCC developed in 2.1% of patients with Child-Pugh A, 7.8% of patients in Child-Pugh B, and 12.4% of patient in Child-Pugh C.²⁹ Increased incidence in patients with Child-Pugh B was also reported by Romano et al.³⁰

As only 6% of patients in this study fell in Child-Pugh class C; therefore, the exact incidence cannot be calculated with the small sample size of this study.

Our study had a few important limitations. First, it used a cross-sectional design from a single hospital experience, with a non-randomized design with relatively young patients. Moreover, this study had small sample size limited to one part of country. Results may not be generalizable regarding the frequency of the disease. Secondly, the design retained confounders, including age, duration of DAA therapy, smoking, and diabetes. Also, we used only two combinations of DAAs available in Pakistan, which further limits the generalizability of our results to other DAA combinations available in other countries. Due to limited resources, we could not obtain genotypes in our patients. Our study's strengths included the use of consecutive sampling suited to our study design. This study was the first study on this subject in South Asia (to the best of our knowledge), and we excluded all possible risk factors for HCC.

Conclusion

According to our results, HCV-associated cirrhosis patients treated with DAAs had an increased incidence of HCC during the six-month follow-up. HCC was seen more commonly in patients who received a combination of sofosbuvir/daclatasvir. More studies with larger sample sizes and longer follow-up are required to confirm these findings.

Abbreviations

CT, computed tomography

DAAs, direct-acting antiviral agents

HCC, hepatocellular carcinoma

HCV, hepatitis C virus

IFN, interferon

MELD, model for end-stage liver disease

SVR, sustained virologic response

Declarations

Ethics approval and consent to participate

Research has been performed in accordance with the declaration of Helsinki. Isra University Ethical Research Committee (Clinical Sciences) on its 14th meeting (IUH/ASST DEAN (CS)/03/30) approved this

study stating there is no ethical issue involved in this study. Informed written consent was taken from patient who meet the inclusion criteria.

Consent for publication

Not applicable.

Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Competing interests

The authors declare that they have no competing interests.

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Authors' contributions

IBA contributed design of work, drafted introduction and discussion and revised all work. JMB collected, analyzed, and interpreted data. AAS contributed to concept and methodology of manuscript.

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Figures

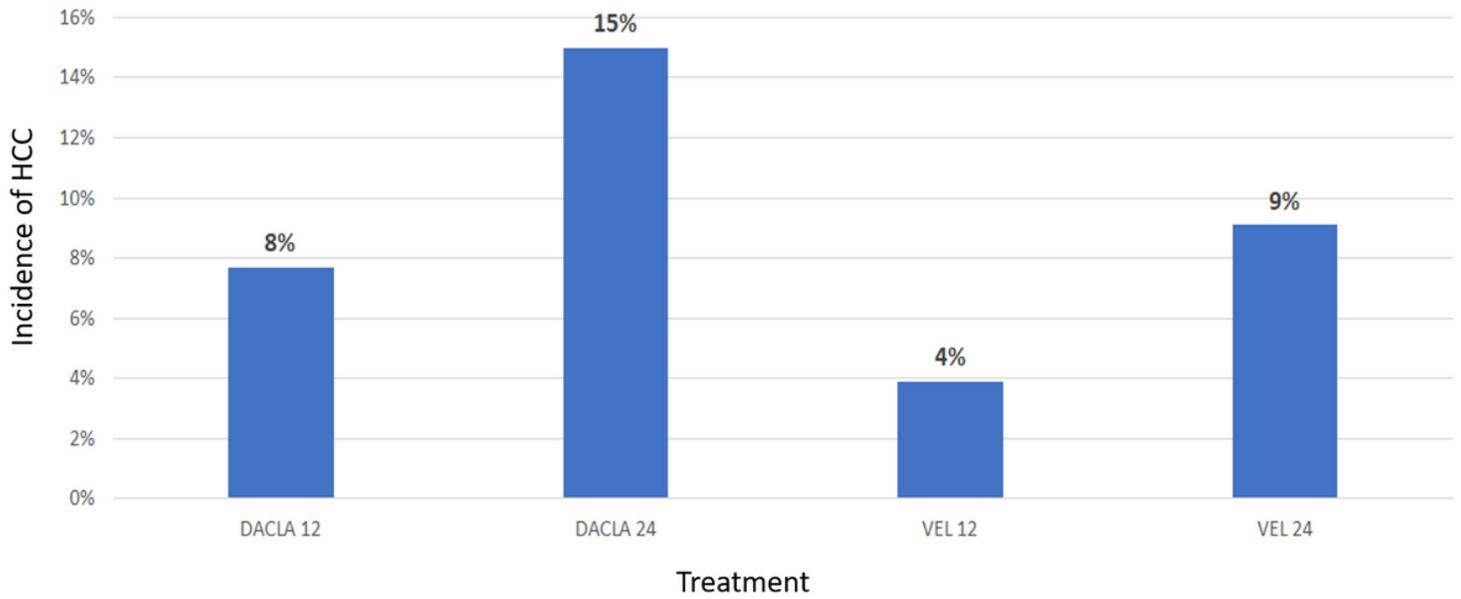


Figure 1

HCC according to duration of treatment Abbreviations: HCC, hepatocellular carcinoma; DACLA 12, sofosbuvir/daclatasvir for 12 weeks; DACLA 24, sofosbuvir/daclatasvir for 24 weeks; VEL 12, sofosbuvir/velpatasvir for 12 weeks; VEL 24, sofosbuvir/velpatasvir for 24 weeks.