

Risk Predictors for De Novo Hepatocellular Carcinomas in Chronic hepatitis C Treated Patients who Achieved Sustained Virologic Response: a Retrospective Study

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Abstract: Background and study aim: Recently, the occurrence of de novo or recurrence of HCC in HCV patients, who treated with direct acting antivirals, has gained a rising interest in clinical application. There are wide debates about the possible role of these treatments in this respect. The recent published studies regarding the risk of developing HCC after treatment with DAA concluded that achieving SVR with DAA regimens was associated with a significant risk reduction of HCC, however, this risk remained high in patients with advanced fibrosis, thus demanding continuous surveillance strategies in this population. The aim of this study is to evaluate possible risk predictors related to de novo HCC in HCV patients who achieved SVR to either DAAs or peg-IFN regimens. **Patients and methods:** Seven hundred and fifty treated HCV patients, who achieved SVR, were retrospectively included. They were divided to two groups, group1 included 150 patients proved to have denovo HCC after treatment, and group2 included 600 patients did not prove to have HCC after treatment. All patients were investigated as regards the degree of liver fibrosis using FIB4 score, the severity of liver dysfunction using Child Pugh and MELD scores, DM and HCV treatment regimens either DAAs or Peg-IFN. **Results:** FIB4 score had a high significant risk for HCC by multivariable logistic regression (p value =0.0001 and Odds ratio =1.626 with 95% CI=1.347-1.964). Child Pugh score had a high significant risk for HCC (p value =0.0001 and Odds ratio =1.426 with 95% CI=1.172-1.734). The MELD score had a high significant risk for HCC (p value =0.028 and Odds ratio =1.124 with 95% CI=1.013-1.246). However, after adjustment of the risk estimate of DM by multivariable logistic regression, we found that DM had a non-significant risk for HCC (p value =0.431 and Odds ratio =1.2 with 95% CI=0.762-1.891). HCV patients who were treated by DAAs had lower -however non significant- risk for HCC (p value =0.639 and Odds ratio =0.898 with 95% CI=0.572-1.409). **Conclusion:** Higher grades of liver fibrosis using FIB4 score and advanced liver dysfunction using either MELD or Child Pugh scores are independent risk predictors for de novo HCC in HCV patients treated by either DAAs or Peg-IFN who achieved SVR. DAAs had lower -however non significant- risk for HCC.

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Key words: Direct acting antivirals, Hepatocellular carcinomas, Risk factors

1. Introduction:

In Egypt, HCC is considered to be one of the commonest cancers (1,2). Worldwide, an approximate more than 170 million people have HCV infection. HCV prevalence is highest in Egypt of more than 15% of the general population which is one of the major causes of chronic liver disease (3,4). Generally, hepatocellular carcinomas have an inferior prognosis due to late diagnosis and lack of efficient treatment options (5). Accordingly, early diagnosis of HCC in cirrhotic patients is essential (6). Data from Europe and North America have been published regarding the risk of developing HCC after treatment with DAA concluded that achieving SVR with DAAs regimens was associated with a significant risk reduction of HCC. However, this risk remained high in patients with advanced fibrosis, thus demanding continuous surveillance strategies in this population (7,8).

Our aim of this study is to evaluate possible risk predictors related to de novo HCC in HCV patients who achieved sustained virologic response to either the new DAAs or peg-IFN regimens.

2. Patients and methods:

This retrospective case control study was performed at both Clinical Oncology Department and the Hepatology Unit of Internal Medicine Department at Tanta University Hospitals.

We retrospectively analyzed Seven hundred and fifty treated HCV patients, who achieved sustained virologic response to either the new DAAs or Peg-IFN, who visited our outpatient HCC screening clinic in the period from January 2014 to December 2018 and who fulfilled the inclusion and exclusion criteria of this study.

They were divided to two groups as regards the occurrence of de novo HCC. Firstly, we selected

group1 that included 150 patients, who proved to have de novo HCC after treatment, it included 82 (54.6%) males and 68 (45.4%) females, the median (range) of their ages was 45 (37) years, and the median (range) observational time (which is the time to HCC) was 29 (25) months, of them 120 (80%) patients were treated by the DAAs regimen, and 30 (20%) patients were treated by peg-IFN regimen. Then, group2 was selected as a matched comparative group that included 600 patients, who did not prove to have HCC after treatment, it included 345 (57.5%) males and 255 (42.5%) females, the median (range) of their ages was 44 (36) years, and the median observational time (which is the time to registered last visit) was 30 (31) months, of them 490 (81.7%) patients were treated by the DAAs regimen, and 110 (18.3%) patients were treated by peg-IFN regimen. All patients in both groups were matched as regards age, sex and the follow up time after treatment.

All patients were evaluated as regards the degree of liver fibrosis, non-invasively, using FIB4 score, and as regards the severity of liver dysfunction using Child Pugh score or MELD score. All patients were investigated as regards the presence of diabetes mellitus (DM) and HCV treatment regimens either DAAs or Peg-IFN and the time to HCC occurrence.

HCC was diagnosed using triphasic computed tomography or dynamic magnetic resonance imaging by the presence of typical hypervascular characteristics in the arterial phase followed by rapid washout in the portal venous or delayed phases.

Inclusion criteria

All included patients were proved to receive treatment for HCV and achieved sustained virologic response to either DAA or peg-IFN.

Exclusion criteria

We excluded the following type of HCV patients from our study; all patients who did not achieve a sustained virologic response to either treatment regimens, all patients with HCV/HBV co-infection or HCV/HIV co-infection, and all patients with pretreatment history of HCC.

The study was approved by the Hospital Ethical Committee and was performed according to the Principles of the Declaration of Helsinki.

Statistical analysis of the data:

All collected data were organized, tabulated and statistically analyzed using the IBM SPSS, version 23 statistic software (SPSS Inc., Armonk, NY, USA).

For quantitative data, the median and range were calculated if abnormally distributed or the mean and SD were calculated if normally distributed. Qualitative data were reported as frequency and percentage or proportion. Student t test was used for two group comparisons of normally distributed data, and the Mann-Whitney U test where data was not normally

distributed. Chi-Square test was performed to conduct group comparisons for categorical data.

Risk predictors for HCC were evaluated using univariable logistic regression and the adjusted risk estimates were calculated using the multivariable logistic regression. ROC curves were done for the optimum cut-off values for the significant risk predictors. Two sided p value ≤ 0.05 was considered as statistically significant.

3. Results:

The main baseline characteristics for all included patients are illustrated in table 1. There was no significant differences between both groups as regards age and sex distribution, AST, ALT, serum creatinine, hemoglobin and the observational time, (p value =0.531, 0.395, 0.758, 0.062, 0.083, 0.564 and 0.096 respectively), however there were significant differences as regards serum bilirubin, serum albumin, INR and platelet count (p value = 0.0001).

The studied risk predictors that may be related to the occurrence of de novo HCC are illustrated in table2 and table3:

Evaluations of the risk estimate of liver fibrosis using the FIB4 score:

The univariable regression analysis for the FIB4 score showed that patients with higher scores had higher significant risk for HCC (p value =0.0001 and Odds ratio =2.240 with 95% CI=1.930-2.601), and after adjustment of this risk estimate by multivariable logistic regression, we found that FIB4 score still had a high significant risk for HCC (p value =0.0001 and Odds ratio =1.626 with 95% CI=1.347-1.964).

Evaluation of the risk estimate of liver dysfunction using MELD or Child Pugh scores:

The univariable regression analysis for the Child Pugh score showed that patients with higher scores had higher significant risk for HCC (p value =0.0001 and Odds ratio =2.051 with 95% CI=1.755-2.370), and after adjustment of this risk estimate by multivariable logistic regression, we found that Child Pugh score still had a high significant risk for HCC (p value =0.0001 and Odds ratio =1.426 with 95% CI=1.172-1.734).

Similarly, The univariable regression analysis for the MELD score showed that patients with higher scores had higher significant risk for HCC (p value =0.0001 and Odds ratio =1.453 with 95% CI=1.345-1.569), and after adjustment of this risk estimate by multivariable logistic regression, we found that MELD score still had a high significant risk for HCC (p value =0.028 and Odds ratio =1.124 with 95% CI=1.013-1.246).

Evaluation of the risk estimate of DM:

The univariable regression analysis showed that the diabetic patients had higher significant risk for

HCC (p value =0.026 and Odds ratio =1.561 with 95% CI=1.055-2.310), however, after adjustment of this risk estimate by multivariable logistic regression, we found that DM had a non-significant risk for HCC (p value =0.431 and Odds ratio =1.2 with 95% CI=0.762-1.891).

Evaluation of the risk estimate of DAA vs Peg-IFN regimen:

The univariable regression analysis showed that the HCV patients who were treated by DAA had lower, however non significant, risk for HCC (p value =0.639 and Odds ratio =0.898 with 95% CI=10.572-1.409).

Table 1: Baseline criteria

		Grouping				Statistics
		HCC N=150		No HCC N=600		P value
Sex	Male	82	(54.6%)	345	(57.5%)	0.531
	Female	68	(45.4%)	255	(42.5%)	
Age (years)	Median (Range)	45	(37)	44	(36)	0.395
AST	Median (Range)	43	(17)	42	(17)	0.758
ALT	Median (Range)	43	(23)	43	(17)	0.062
Bilirubin	Median (Range)	2.4	(2.1)	1.9	(1.5)	0.0001
Albumin	Median (Range)	2.9	(1.6)	3.2	(1.5)	0.0001
INR	Median (Range)	1.75	(1.0)	1.5	(0.7)	0.0001
PLT	Median (Range)	120	(112)	147	(125)	0.0001
S.creatinine	Median (Range)	0.9	(0.8)	0.87	(0.8)	0.083
Hemoglobin	Median (Range)	11	(2.5)	11.25	(2.5)	0.564
Follow up time (ms)	Median (Range)	29	25	30	31	0.096
Time to HCC (ms) Median (Range)	DAA	28	(25)			0.302
	Peg-IFN	29	(25)			

Table 2: Univariable logistic regression for studied risk factors

		Grouping				Statistics			
		HCC N=150		No HCC N=600		B	P value	Exp.B	95% CI
FIB4 score	Median (Range)	4.19	5.76	1.98	3.1	0.807	0.0001	2.240	1.930-2.601
Child score	Median (Range)	9	5	6	5	0.719	0.0001	2.051	1.755-2.370
MELD score	Median (Range)	13.6	9.2	10.9	10.5	0.373	0.0001	1.453	1.345-1.569
DM	Yes	48	32%	139	23.2%	0.445	0.026	1.561	1.055-2.310
	No	102	68%	461	76.7%				
Treatment regimen	DAA	120	80%	490	81.7%	-0.108	0.639	0.898	0.572-1.409
	Peg IFN	30	20%	110	18.3%				

Table 3: Multivariable logistic regression for studied risk factors

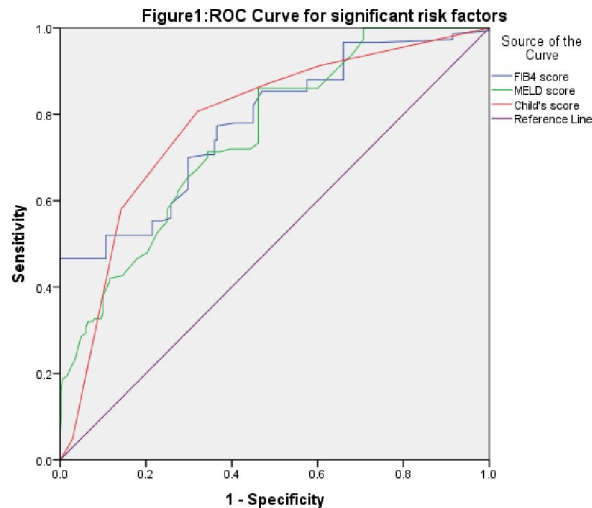
	B	P value	Exp.B	95% CI	
				Lower	Upper
FIB4 score	0.486	0.0001	1.626	1.347	1.964
Child score	0.355	0.0001	1.426	1.172	1.734
MELD score	0.116	0.028	1.124	1.013	1.246
DM	0.183	0.431	1.200	0.762	1.891

Evaluation of the optimum cut-off values for the significant risk predictors:

Table 4 and figures 1 show the different cut-off values for FIB4 score, Child Pugh score and MELD score with their respective AUC, likelihood ratio, sensitivity and specificity.

Table 4: ROC curve for the significant risk predictors

	AUC	P value	95% C.I.		Cut off value	LR	Sensitivity	Specificity
			Lower	Upper				
FIB4 score	0.780	0.0001	0.736	0.824	4.34	10.9	46.7%	95.7%
Child score	0.783	0.0001	0.742	0.824	8	4.08	58%	85.8%
MELD score	0.751	0.0001	0.710	0.793	16.95	37.4	18.7%	99.5%



On the other hand, There was no any significant difference as regards the time to HCC occurrence between patients who were treated with DAA and those treated with Peg-IFN (P value =0.302).

4. Discussion:

Recently, the occurrence of de novo or recurrence of HCC in HCV patients who were treated with the new DAA has gained rising interest and wide debates in clinical application. The recent published studies regarding the risk of developing HCC after treatment with DAAs concluded that achieving SVR with DAA regimens was associated with a significant risk reduction of HCC, however, this risk remained high in patients with advanced fibrosis, thus demanding continuous surveillance strategies in this population (7-8). We tried in this study to face these debates and to evaluate the possible risk predictors related to de novo HCC in HCV patients who achieved SVR to either the new DAAs or peg-IFN regimens.

Evaluation of the risk estimate of liver fibrosis using the FIB4 scores:

Our results showed that liver fibrosis using FIB4 score is a significant independent risk predictor for occurrence of de novo HCC, moreover de novo HCC has a high likelihood ratio (10.9) to occurred at a cut off level for FIB4 score of 4.34. Many recently published papers had similar results that identified the carcinogenic hazard of advanced liver fibrosis and

concluded that non-invasive markers of liver fibrosis can be used to identify patients at increased risk for HCC following DAA therapy in HCV patients (9-11).

Evaluation of the risk estimate of liver dysfunction using MELD or Child Pugh scores:

Our results showed that MELD and Child Pugh scores are significant independent risk predictors for occurrence of de novo HCC; moreover de novo HCC has a high likelihood ratio to occur in patients with higher MELD and Child Pugh scores (LR; 37.4 and 4.08 respectively with cut off levels; 16.95 and 8 respectively). The results of Syed T et al, showed higher, however non-significant, difference as regards both MELD and Child Pugh scores (p value =0.173 and 0.064 respectively) between treated HCV patients with HCC and those without HCC (11).

Evaluation of the risk estimate of DM:

Our results show that the diabetic patients had higher significant risk for HCC however, after adjustment of this risk estimate by multivariable logistic regression; we found that DM had a non-significant risk for HCC. These results are against many papers who concluded that DM can be used to identify patients at increased risk for HCC following DAA therapy (9-13). Although, the exact mechanisms of carcinogenesis due to diabetes mellitus have not been reported, it has been hypothesized that insulin resistance promoted susceptibility of the liver to lipid peroxidation and production of reactive oxygen species, which could lead to carcinogenesis (14).

The debate in our results could be explained by the underlying liver cirrhosis which is considered as an independent risk factor of liver carcinogenesis and the possible role of DM in this respect could be just a contributory risk factor in liver carcinogenesis.

Evaluation of the risk estimate of DAA vs Peg-IFN regimen:

DAA effects, on the liver carcinogenesis after achieving an SVR are not clear. DAAs directly inhibit HCV proliferation, and compared with peg-IFN, they don't activate immunity (15). Rather, they may suppress liver immunity by rapid viral elimination (16). The results of our study shows that; HCV patients who were treated with DAAs had lower however non-significant risk for occurrence of de novo HCC. This is similar to Nagata H et al who reported that there was no difference in carcinogenesis

after an SVR between IFN-based and DAA combination treatments (17). Other studies have reported that DAA treatment promotes HCC whereas other reports deny this effect (18,19).

From all of these results, we could conclude that advanced liver fibrosis, which is identified non-invasively by FIB4 score, and advanced liver dysfunction, which is identified either by MELD score or Child Pugh score, are independent risk predictors for occurrence of de novo HCC in HCV patients who were treated with DAA and achieved SVR. In those patients regular screening strategies for HCC is mandatory for early diagnosis of hepatocellular carcinomas that are more liable to recent curative treatment modalities.

Conflict of interest:

The authors have none to declare.

Abbreviations:

HCV: hepatitis C virus, HCC: hepatocellular carcinoma, DAA: direct acting antiviral, SVR: sustained virologic response, MELD: model of end stage liver disease, DM: diabetes mellitus, ms: months, AUC: area under the curve.

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