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Role of Indoleamine-2,3-Di oxygenase in HCV-associated chronic hepatitis and hepatocellular carcinoma

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The progression of HCC in the era of DAA therapy is critical in determining the onset or recurrence of cancer after HCV eradication. DAAs have shown satisfactory efficacy in the treatment of HCV-sustained viral response (SVR). However, data on HCC risk following DAA-SVR remains contradictory. [1] The purpose of this study is to look at HCC onset in people who have the moderate underlying liver disease by detecting levels of IDO enzyme that show a massive increase in HCC patients compared to recently diagnosed patients with HCV infection and cirrhosis-related HCV patients.[2] These findings raise the possibility that in considering IDO, more than an enzyme, as a marker predicting the development of HCC after HCV eradication. It's also possible that DAA therapy may be the cause of rising IDO levels, which requires further investigation.so we conducted this work aiming to compare the level of IDO enzyme with the progression of HCC despite curing with DAA therapy and studying the effect of therapy on patients by analyzing the severity and predicting suitable biomarkers for detecting HCC occurrence in cured patients from HCV infection by DAA that progress HCC.

Keywords: Echocardiography, right ventricular function, pulmonary embolism.

1.Introduction

Hepatocellular carcinoma (HCC) and liver cirrhosis are mostly brought on by chronic hepatitis C virus (HCV) infection (HCC). [3,4] Eradicating HCV infection lowers the chance of developing HCC since it can encourage carcinogenesis. [5,6] In patients with chronic HCV infection, high rates of sustained virological response (SVR) were attained thanks to the recent introduction of direct-acting antiviral drugs (DAAs). [7,8]

Although SVR with DAAs lowers the prevalence of HCC, DAA treatments have been associated with unanticipated increases in the early onset or recurrence of HCC. [9,10]

Recent studies have shown an increased risk of HCC recurrence in HCV-infected individuals with HCC who initially had a full response to hepatic resection or local ablation and then had DAA-related SVR [11,12]

Higher than the incidence observed with IFN-based therapy, the annual incidence of de novo HCC after inducing SVR with DAAs was reported to be 3%5%. [13,14]

It is uncertain if DAAs, which are used to treat HCV infection, can prevent HCC recurrence after curative treatment for HCC, despite prior evidence suggesting a relationship between the use of DAAs for HCV infection and a decrease in the incidence of HCC.

An intracellular monomeric enzyme called IDO (Indoleamine-2,3-DiOxygenase) that contains heme controls the breakdown of tryptophan in the kynurenine pathway. We go through how IDO affects the course and prognosis of hepatocellular carcinoma as well as possible therapeutic benefits of IDO inhibition on hepatocellular cancer immunotherapy. [15]

2.Patients and methods

Our study will involve 40 patients with HCV-associated chronic hepatitis before and after the treatment and hepatocellular carcinoma at the internal medicine department, University Hospital of Banha, Egypt, divided into four groups:

- Group (1): HCV –associated chronic hepatitis patients completed the course of therapy (> 6 months) after the end of treatment (10 patients).
- Group (2): HCV – associated chronic hepatitis patients with cirrhosis and did not receive DAAs therapy (10 patients).
- Group (3): HCV- associated hepatocellular carcinoma (10 patients)
- Group (4): healthy individuals as a control group (10 patients)

2.1Inclusion criteria:

Patients >18 years.

Patients with HCV- associated with HCC. (In group 3)

HCC patients confirmed by laboratory tests and imaging (U/S or CT or MRI). Computed tomography –magnetic resonance imaging (In group3).

2.2Exclusion criteria:

Patients < 18 years.

Patients with HCC due to other causes rather than chronic hepatitis C infection. (In group 3)

All participants will be subjected to the following:

Full medical history Thorough:

clinical examination (Age-Sex-chronic dis eases(BM-HTN-smoker))

Complete blood picture (CBC) include (HGB -WBC count -PLT count)

Liver and kidney function tests including (ALT-AST-Albumin-Bilirubin Total and Bilirubin direct)(Creatinine -Urea)

AFP level in blood: alpha-fetoprotein by ELISA technique (Mini-Vidas)

IDO level in blood: indole 2, 3 dioxygenases by ELISA technique (Biokit)

Clinical and laboratory assessments were carried out before treatment. Hepatitis C virus RNA levels were measured using the Roche COBAS TaqMan HCV Auto assay system(Roche Molecular Diagnostics,

Pretreatment factors that might contribute to HCC development after the end of DAA treatment were evaluated. Potential predictors associated with HCC development included sex, age, body mass index, white blood cell count, platelet count, ALT, AST, total bilirubin, albumin, prothrombin time,α-fetoprotein (AFP) at the start of treatment, and whether or not SVR was achieved.

All patients will undergo:

History taking: all patients will be asked for their history of previous deep vein thrombosis or previous history of pulmonary embolism, hypertension, diabetes mellitus, immobilization, malignancy, and oral contraceptive pills use.

3.**Results**

This study was conducted in the Internal Medicine Department, at Banha University Hospital. The study was done in four groups:

Group I: Patients with HCV infection (10 patients).

Group II: Patients with HCV and cirrhosis (10 patients).

Group III: Patients with hepatocellular carcinoma (10 patients).

Group IV: Healthy individuals as a control group (10 patients).

General characteristics

The age of the patients significantly differed between the studied groups (P < 0.001). Additionally, hypertension showed a significant association with the studied groups (P = 0.038). In contrast, sex (P = 0.433), diabetes mellitus (P = 0.140), and smoking (P = 0.891) did not significantly differ between the studied groups. Regarding the BCLC score in group III, half of the patients had a score of A, and one-third had a score of C (30%). Only two patients scored B. General characteristics with post hoc comparisons are shown in table 1.

Table (1): General characteristics with post hoc comparisons

	Group I (n = 10)	**Group II (n = 10)**	**Group III (n = 10)**	**Group IV (n = 10)**	**P -value**
Age (years)	**27 ±9 [2, 3]**	**55 ±9 [1, 4]**	**64 ±9 [1, 4]**	**31 ±7 [2, 3]**	**<0.001**
Sex	**4 (40)**	**7 (70)**	**7 (70)**	**4 (40)**	**0.433**
Males	**6 (60)**	**3 (30)**	**3 (30)**	**6 (60)**	
Females					
Diabetes mellitus	**1 (10)**	**3 (30)**	**4 (40)**	**0 (0)**	**0.140**
Hypertension	**0 (0)**	**2 (20)**	**4 (40)**	**0 (0)**	**0.038**
Smoking	**1 (10)**	**1 (10)**	**2 (20)**	**0 (0)**	**0.891**
BCLC					
A	**-**	**-**	**5 (50%)**	**-**	**-**
B	**-**	**-**	**2 (20%)**	**-**	
C	**-**	**-**	**3 (30%)**	**-**	

Data are presented as mean ±SD or number (percentage). Significant results are marked in bold; 1: Significantly different from group I; 2: Significantly different from group II; 3: Significantly different from group III; 4: Significantly different from group IV

❖Laboratory findings

All laboratory parameters, including albumin, ALT, AST, total bilirubin, direct bilirubin, Pt, creatinine, urea, hemoglobin, TLC, platelets, AFP, ELISA, PCR, and IDO (figure 1) significantly differed between the studied groups (P < 0.001 for each). All parameters with post hoc comparisons are presented in table 2.

Table (2) Laboratory findings of the studied groups

	Group I (n = 10)	Group II (n = 10)	Group III (n = 10)	Group IV (n = 10)	P -value
Albumin	**4.09 ±0.34** [2, 3]	**2.78 ±0.46** [1, 3, 4]	**2.03 ±0.54** [1, 2, 4]	**4.18 ±0.43** [2, 3]	**<0.001**
ALT	**28 (20 - 74)** [3]	**46 (29 - 98)** [4]	**99 (82 - 133)** [1, 4]	**23 (16 - 30)** [2, 3]	**<0.001**
AST	**36 (28 - 82)** [3]	**67 (38 - 119)** [4]	**138 (102 - 227)** [1, 4]	**31 (19 - 39)** [2, 3]	**<0.001**
T Bil.	**0.83 (0.57 - 0.95)** [3]	**2.79 (0.77 - 4.1)**	**10.1 (5.11 - 18.2)** [1, 4]	**0.83 (0.76 - 0.99)** [3]	**<0.001**
D Bil.	**0.12 (0.09 - 0.2)** [3]	**0.86 (0.11 - 2.11)**	**8.45 (4.02 - 15.3)** [1, 4]	**0.15 (0.11 - 0.2)** [3]	**<0.001**
Pt	**12.3 ±0.4** [3]	**18 ±2.7** [3]	**41.8 ±10.9** [1, 2, 4]	**12.2 ±0.4** [3]	**<0.001**
Creatinine	**0.86 ±0.12** [3]	**1.24 ±0.41** [3]	**2.58 ±0.57** [1, 2, 4]	**0.82 ±0.15** [3]	**<0.001**
Urea	**29 (20 - 32)** [2, 3]	**48 (32 - 117)** [1]	**197 (96 - 274)** [1, 4]	**32 (25 - 46)** [3]	**<0.001**
Hemoglobin	**12.7 ±0.6** [2, 3]	**10 ±1.6** [1, 4]	**10 ±1.4** [1, 4]	**13.1 ±0.9** [2, 3]	**<0.001**
TLC	**7.6 (4.7 - 10.2)** [3]	**5.56 (2.1 - 9.5)**	**2.94 (2 - 3.4)** [1, 4]	**7.75 (4.4 - 9.9)** [3]	**<0.001**
Platelets	**221 ±62** [2, 3, 4]	**105 ±17** [1, 4]	**76 ±13** [1, 4]	**291 ±60** [1, 2, 3]	**<0.001**
AFP	**5.57 (1.75 - 9.71)** [3]	**28.51 (18.07 - 51.13)** [4]	**1003 (380 - 2077)** [1, 4]	**1.03 (0.44 - 6.5)** [2, 3]	**<0.001**
ELISA	**34.02 (25.35 - 67.53)** [4]	**36.19 (15.61 - 53.75)** [4]	**-**	**0.06 (0 - 0.21)** [1, 2]	**<0.001**
PCR	**498820 (78548 - 5461851)** [2]	**-33306 (17056 - 202013)** [1, 3]	**-363081 (52159 - 664015)** [2]	**-**	**<0.001**
IDO	**8.25 (6.64 - 9.11)** [3]	**9.62 (7.39 - 10.38)** [3, 4]	**20.16 (13.16 - 34.19)** [1, 2, 4]	**4.61 (3.37 - 6.07)** [2, 3]	**<0.001**

Data are presented as mean ±SD, median (min-max), or number (percentage). Significant results are marked in bold; 1: Significantly different from group I; 2: Significantly different from group II; 3: Significantly different from group III; 4: Significantly different from group IV

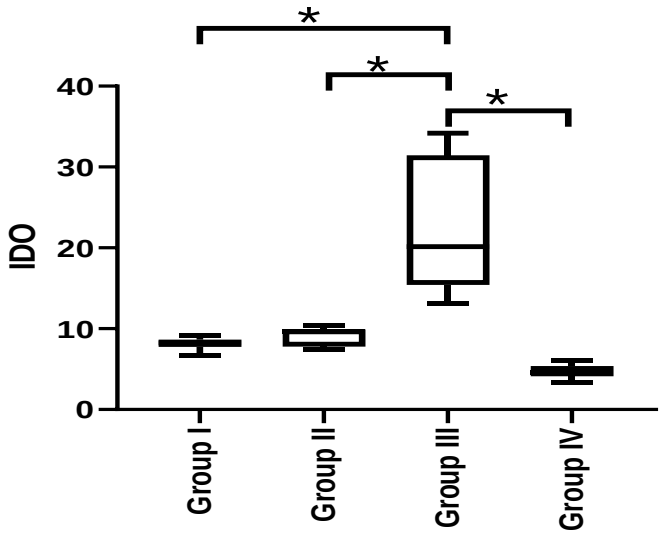

Fig (1) IDO level in the studied groups

Correlation between IDO and other parameters in group I

In group I, IDO showed significant positive correlations with ELISA (r = 0.736, P = 0.015) and PCR (r = 0.979, P < 0.001)

No significant correlations were reported between IDO and other parameters, including age (P = 0.725), albumin (P = 0.266), ALT (P = 0.987), AST (P = 0.751), total bilirubin (P = 0.454), direct bilirubin (P = 0.711), Pt (P = 0.105), creatinine (P = 0.464), urea (P = 0.594), hemoglobin (P = 0.104), TLC (P = 0.614), platelets (P = 0.713), and AFP (P = 0.907).

4.Discussion

Infection with the Hepatitis C virus is a major cause of chronic hepatitis, which can lead to cirrhosis and hepatocellular carcinoma (HCC). The recent introduction of direct-acting antivirals (DAA) results in SVR rates of >90% in treated patients regardless of the stage of liver fibrosis, with an excellent safety profile. This significant advancement has enabled the treatment of a greater number of patients, including those with more advanced liver dysfunction and a higher risk of HCC. An SVR lowers the risk of hepatic decompensation, the need for a liver

transplant, and both liver-related and overall mortality.[16]

Many Studies Have Examined The Risks Of Hcc Development, and the characteristics of HCC development after SVR in chronic hepatitis C patients receiving IFN-based therapy have been reported. Several studies have found that elderly patients, patients with advanced liver fibrosis, and men are more likely to develop HCC. [17-19] Recent studies have looked at pre- and post-treatment factors of IFN-based therapy as predictors of HCC development. Ozeet al. reported that AFP at 24 weeks after the end of IFN-based therapy was associated with the development of HCC.[20]Asahinaet al. also noted that higher post-treatment AFP and ALT levels increased the risk of developing HCC. [21] Other reported risk factors in patients with SVR by IFN-based therapy were glucose metabolism disorders and alcohol intake.[22,24]

Conclusion and recommendation

When compared to HCV recently diagnosed patients and HCV-related cirrhosis patients, HCC patients have significantly higher levels of IDO enzyme, which can be used as an effective biomarker for the progression of liver diseases (fibrosis -cirrhosis -carcinoma). Gradual or progressive rise in IDO levels from healthy to HCV patients, followed by cirrhotic patients, and then abnormal jumps in IDO levels in carcinoma patients. IDO can also be used as a predictor for pro-oncogenes or occult HCC infection rather than AFP due to the high sensitivity and specificity of the IDO enzyme as well as disease progression.

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