Serum Lactadherin as a Diagnostic Biomarker in Hepatitis C Virus Cirrhotic Patients with and without Hepatocellular Carcinoma

Mohamed Adel Abdel Aziz[1], Khaled Mahmoud Mohiedeen[1], Reham A.Abo Elwafa[2], Mahmoud Attalla[3], Mohammed S. Ahmed[1]

[1] *Tropical Medicine Department, Faculty of Medicine, Alexandria University,Egypt .*
[2] *Clinical Pathology Department, Faculty of Medicine, Alexandria University,Egypt.*
[3] *Gastroenterology and Hepatology Department, Alexandria Police Hospital,Egypt.*

Corresponding Author
Mohamed Adel Abdel Aziz

Mobile:+2 01225540023

Email: dradel2008@windowoslive.com

Receive date:24 /2/2024
Revise date:20/3/2024
Accept date:2 /5/2024
Publish date:10/5/2024

Keywords: Hepatocellular carcinoma (HCC), Liver cirrhosis, Lactadherin, HCV

Background and study aim: Hepatocellular carcinoma (HCC) is commonly occurs in association with cirrhosis which is the end stage of HCV infection. Although early HCC detection is important for a better prognosis, efficient biomarkers are still needed. The Lactadherin protein also referred as milk fat globule-EGF factor 8 (MFG-E8), is a significant opsonin for programed cell death and encourages apoptotic cell clearance by the macrophages. An association between Lactadherin with liver diseases has been reported.The purpose of this investigation was to determine serum Lactadherin concentrations as biomarker for HCC detection in cirrhotics.

Patients and Methods: This case-control research had three groups: patients with cirrhosis but no HCC (Group I), patients with both cirrhosis and HCC (Group II), and healthy volunteers (Group III). Clinical and laboratory data, including results from imaging, Lactadherin levels, and liver function tests, were analyzed using a number of statistical tests.

Results: Lactadherin serum concentrations were lower in patients diagnosed with HCC than in those diagnosed with cirrhosis. There was no difference with statistical significance among the involved cirrhotic patients with no HCC and other healthy involved controls.

Conclusion: Lactadherin seems to be involved in the liver conditions initiation and development, such as HCC in cirrhotic individuals. Although it cannot diagnose cirrhosis itself, it may be used as a possible HCC biomarker in the context of cirrhotic liver.

INTRODUCTION

Globally, about 58 million individuals had chronic HCV infection as mentioned by the World Health Organization (WHO) [1]. Chronic HCV infection will result in development of cirrhosis, portal hypertension, hepatic decompensation, and HCC [2]. The fourth most common cancer in Egypt is HCC [3]. HCC prognosis improves with early discovery, since therapy may improve survival. Thus, developing reliable biomarkers to aid early HCC diagnosis in cirrhotic individuals is a priority [4]. AFP concentration is a major marker for liver cancer; however its low sensitivity and specificity prohibit it from being used as the principal monitoring test for HCC. For instance, elevated AFP levels are seen in less than 20% of individuals with early HCCs while elevated AFP levels might serve as an indicator of viral hepatitis or decompensated liver disease, rather than HCC specifically [5]. Because of this, there is an increasing interest in studying alternative biomarkers that can detect HCC in the presence of cirrhosis more accurately.

Our research marker Lactadherin, found in nursing mice' milk fat globules, is a viable candidate. Macrophages, fibroblasts, dendritic, and epithelial cells express Lactadherin [6]. Lactadherin levels may indicate the relationship between apoptotic cells, immunological responses, and inflammation in the tumor microenvironment.

It may affect carcinogenesis, angiogenesis, and immune surveillance evasion [7]. This suggests that serum levels of Lactadherin might be regarded as a possible biomarker to predict the HCC development and progression among those who are cirrhotic. This investigation evaluated serum levels of Lactadherin among cirrhotic people with and without HCC who have an HCV diagnosis and investigated Lactadherin's potential to be employed as an HCC diagnostic biomarker.

PATIENTS/MATERIALS AND METHODS

Study design: It is a prospective case control study.

Study settings: This is a single center study carried out in Tropical Medicine Department at the Main University Hospital in Alexandria during period (2020-2022). Participants were divided into 3 groups. Group I includes 30 patients with liver cirrhosis without HCC. Group II contains 30 patients with liver cirrhosis with HCC and group III contains 30 healthy subjects as normal controls. The participants' ages varied from 36 to 71 years old.

Study patients: All patients attending to Tropical Medicine Department suffering from post HCV liver cirrhosis and HCC.

Endpoints:

The primary end-point of this study is to determine level of Lactadherin in healthy subjects, cirrhotic patient and HCC patients respectively.

Secondary end-points: Test the feasibility of using Lactadherin as non-invasive sensitive marker for HCC.

Sample size: 90 individuals and divided into three groups.

Inclusion criteria: Patients suffering from post HCV induced liver cirrhosis and HCC, age more than 18 years old and both genders were included.

Exclusion criteria: patients had sepsis, concomitant cancer other than HCC, pregnant patients, smokers, patients who had autoimmune disorders, and patients with cardiac diseases such as myocardial infarction, and HCC due to other causes rather than HCV.

Patient assessment: All patients were subjected to assessment through history taking, complete physical examination, routinely Laboratory examinations as CBC, tests of liver function as (aspartate aminotransferase [AST], alanine aminotransferase [ALT], both total and direct serum bilirubin, prothrombin time [PT], INR, albumin level in serum, alkaline phosphatase), renal function related tests as (serum urea and creatinine), fasting blood glucose, and serum AFP levels were determined.

All participants conducted radiological examinations, including an abdominal ultrasound examination and a triphasic abdominal CT for individuals who showed ultrasound-proven hepatic-involved focal lesions.

Our primary research marker, Lactadherin, was measured in serum samples from every patient. It was also assessed by the ELISA method utilizing the (MFG-E8) kit (Bioassay Technology Laboratory, China). For patients with cirrhosis, we estimated the Child-Pugh score, and for those with HCC, the Barcelona staging classification (BCLC). For each patient who participated, we also ran ELISA tests for antibodies of HCV, surface antigen of hepatitis B, and antibodies against schistosoma using the indirect hemagglutination method.

Statistical analysis

The loaded Data in the computer was evaluated using the software of IBM SPSS, version 20.0. To represent qualitative type of data, figures, and percentages are used. Quantitative data are represented as the following parameters: Range including the minimum and maximum values, mean, median, SD, and interquartile range (IQR).

RESULTS

Study participants

Ninety applicants participated in the research. The participants were split into three categories. The group-related demographic data under consideration revealed insignificant differences in both age and gender between any of the groups. As displayed in Table 1 .

Regarding the symptoms experienced by the patients at the admission time, Figure 1 shows that the most prevailing symptom in cirrhotic patients was easy fatigue (90%) followed by anorexia (53.3%) and the least reported symptom was abdominal pain (25%). Moreover, the most

common symptom in HCC patients was also easy fatigue (92%) followed by abdominal pain and weight loss (85% and 80% respectively) and the least reported symptom was melena 70%.

The most frequent sign seen in cirrhotic patients was splenomegaly, which was found in every cirrhotic patient followed by ascites 58% and the least reported sign was hepatomegaly which was reported in only 30% of the cirrhotic patients. Moreover, the most reported sign detected in HCC patients was splenomegaly which was detected in all patients followed by ascites and lower limb edema (80% and 78% respectively) and the least reported sign was wasting which was detected in only 34% of the patients as shown in Figure 2.

In terms of laboratory tests, CBC results revealed a substantial difference in hemoglobin and count of platelets among the individuals diagnosed with cirrhotic liver, HCC, and also the healthy participants. Additionally, Table 1 demonstrates that platelet count and hemoglobin levels were not statistically different in cirrhotic HCC patients, although there was significance between HCC-diagnosed participants and the healthy involved group and between cirrhotic patients and the healthy control.

No differences with statistical significance were found across the involved groups, and all had normal FBG levels and renal function testing. Additionally, the CRP was negative and the ESR levels were normal for all of the applicants. Regarding the liver profile, there were differences with statistical significance among all the individuals with groups involving cirrhotic participants and the group of healthy controls involving all parameters ($p < 0.001$). According to Table 1, all cirrhotic and HCC patients' ALP serum levels and total bilirubin were higher substantially than those healthy involved controls.

Additionally, Table 1 displayed that blood levels of albumin in cirrhotic and HCC participants were lower significantly than those healthy involved controls.

Additionally, the serum liver-involved enzyme levels (AST, ALT) among all the individuals with cirrhosis and HCC groups were considerably greater than those of the control group. Groups I and II also displayed a significant INR rise in comparison to the control-involved group. With respective averages of 50.40 ± 7.09, 48.20 ± 13.33, and 89.47 ±5.27, Table 1 shows that PA dropped significantly in groups I and II compared to controls.

As demonstrated in Table 2, the three study groups had substantial differences in AFP serum levels, with HCC patients having a significantly higher level statistically than those involved in the cirrhotic and control groups .

All patients in groups I and II had cirrhosis according to ultrasonography, but those in group III showed normal livers. Moreover, Triphasic CT of HCC individuals shows the number and size of HCC lesions, 17 patients had single mass and 13 cases had multiple masses. As regards the size of lesions, five patients ranged from 2 -2.9 cm 17 patients were from 3-5 cm and 8 patients had lesions which were more than 5 cm in size.

Furthermore, in the HCC group, 22 patients had portal vein thrombosis, 7 patients experienced metastatic lymph node and 8 patients showed distant metastasis.

Each individual in Groups I, II, and IIIa had post-viral cirrhosis of the liver. Each case was brought on by a long-term HCV infection, and each patient underwent HCV treatment and displayed a negative HCV PCR test. Additionally, both the autoimmune hepatitis marker and the hepatitis B virus (HBV) were found to be absent in their tests. In Table 2, the score of Child-Pugh and each case categorization are displayed. Additionally, Table 2 displayed BCLC for HCC patients.

Serum milk growth factor-8, our key research measure, differs considerably across the control, cirrhotic liver, and HCC-diagnosed groups, as seen in Table 2.

The HCC presence was also strongly predicted by a serum milk growth factor-8 cutoff value of ≤ 4.014 (ng/ml), with 76.67% sensitivity and 80% specificity, as shown in Table 2.

Similarly, with a threshold of more than 10 ng/ml, AFP might be used to make a diagnosis, yielding 70% sensitivity and a 76.67% specificity. Furthermore, combined usage of serum AFP and milk growth factor-8 resulted in better diagnostic performance in HCC detection within cirrhotic individuals, with 86.67% sensitivity and 90% specificity respectively, as shown in Table 2. There was a negative relationship between the tumor size, BCLC, and CPC, and the Serum milk growth factor-8 level in the HCC group as shown in Figure (3-5).

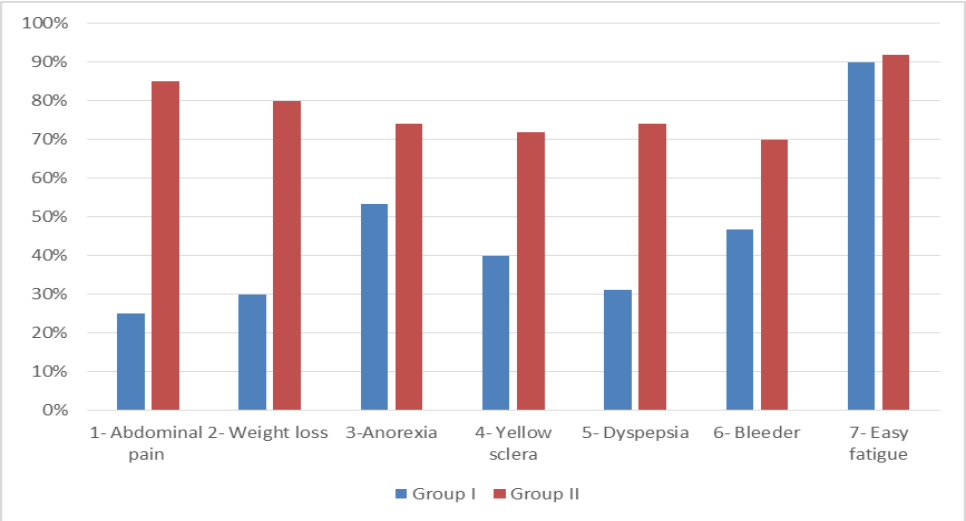


Figure 1. Distribution of the studied cases in accordance to symptoms in each group

Table 1. Comparison between the three studied groups (n=90) according to demographic data and other different parameters:

	Group I (n = 30)		Group II (n = 30)		Group III (n = 30)		Test of Sig.	p
	No.	%	No.	%	No.	%		
Sex Male Female	 25 5	 83.3 16.7	 24 6	 80.0 20.0	 23 7	 76.7 23.3	χ^2= 0.417	0.812
Age (years) Min. – Max. Mean ± SD. Median (IQR)	 40.0 – 70.0 55.53 ± 9.25 56.50 (48.0 – 63.0)		 38.0 – 71.0 54.50 ± 8.39 55.0 (48.0 – 60.0)		 36.0 – 67.0 50.77 ± 7.60 51.50 (46.0 – 55.0)		F= 2.648	0.077
CBC	Group I (n = 30)		Group II (n = 30)		Group III (n = 30)		Test of Sig.	p
PLT Min. – Max. Mean ± SD. Sig. bet. grps.	 25.0 – 85.0 55.43 ± 16.71 p_1=0.863,p_2<0.001*,p_3<0.001*		 20.0 – 100.0 51.43 ± 21.95		 169.0 – 317.0 233.5 ± 43.97		F= 361.206*	<0.001*
Hb Min. – Max. Mean ± SD. Sig. bet. grps.	 7.10 – 11.50 9.44 ± 1.16 p_1=0.145,p_2<0.001*,p_3<0.001*		 7.50 – 16.0 10.19 ± 2.17		 11.40 – 15.20 13.38 ± 0.98		F= 55.950*	<0.001*
ALT Min. – Max. Mean ± SD. Median (IQR) Sig. bet. grps.	 11.0 – 71.0 24.03 ± 13.70 21.0(15.0 – 26.0) p_1<0.001*,p_2=0.851,p_3<0.001*		 13.0 – 201.0 59.30 ± 39.83 53.50(27.0 – 75.0)		 11.0 – 31.0 20.83 ± 5.98 21.50(15.0 – 26.0)		H= 30.014*	<0.001*
AST Min. – Max. Mean ± SD. Median (IQR) Sig. bet. grps.	 23.0 – 93.0 42.20 ± 16.79 39.0(32.0 – 46.0) p_1=0.020*,p_2<0.001*,p_3<0.001*		 18.0 – 250.0 77.93 ± 55.65 63.50(38.0 – 90.0)		 19.0 – 34.0 27.50 ± 4.31 28.50(24.0 – 31.0)		H= 38.904*	<0.001*
Prothrombin activity Min. – Max. Mean ± SD. Median (IQR) Sig. bet. grps.	 35.0 – 63.0 50.40 ± 7.09 50.50(45.0 – 55.0) p_1=0.627,p_2<0.001*,p_3<0.001*		 25.0 – 78.0 48.20 ± 13.33 48.0(35.0 – 57.0)		 81.0 – 99.0 89.47 ± 5.27 88.50(85.0 – 94.0)		F= 189.709*	<0.001*
Serum albumin								

Min. – Max.	**1.70 – 2.70**	**1.60 – 3.70**	**3.64 – 4.30**	**F= 168.127***	**<0.001***
Mean ± SD.	**2.20 ± 0.32**	**2.49 ± 0.57**	**3.93 ± 0.18**		
Median (IQR)	**2.20(1.9 – 2.5)**	**2.50(2.1 – 2.9)**	**3.92(3.8 – 4.1)**		
Sig. bet. grps.	**p_1=0.016*,p_2<0.001*,p_3<0.001***				
Serum alkaline phosphatase					
Min. – Max.	**77.0 – 350.0**	**67.0 – 350.0**	**45.0 – 102.0**	**F= 24.006***	**<0.001***
Mean ± SD.	**159.2 ± 73.25**	**185.8 ± 86.09**	**73.23 ± 14.42**		
Median (IQR)	**145.5(96.0 – 203.0)**	**154.5(111.0 – 254.0)**	**71.0(64.0 – 84.0)**		

	Group I (n = 30)	**Group II (n = 30)**	**Group III (n = 30)**	**H**	**p**
Serum total bilirubin					
Min. – Max.	**0.46 – 30.40**	**1.50 – 30.40**	**0.43 – 0.92**	**47.499***	**<0.001***
Mean ± SD.	**3.78 ± 6.25**	**6.61 ± 6.98**	**0.66 ± 0.12**		
Median (IQR)	**2.05(0.68 – 3.1)**	**4.30(2.8 – 7.3)**	**0.67(0.58 – 0.74)**		
Sig. bet. grps.	**p_1=0.001*,p_2<0.001*,p_3<0.001***				
Serum direct bilirubin					
Min. – Max.	**0.09 – 14.20**	**1.0 – 20.20**	**0.07 – 0.21**	**46.312***	**<0.001***
Mean ± SD.	**2.09 ± 3.04**	**4.53 ± 4.63**	**0.14 ± 0.03**		
Median (IQR)	**1.50(0.13 – 2.3)**	**2.75(1.7 – 5.1)**	**0.14(0.12 – 0.16)**		
Sig. bet. grps.	**p_1=0.001*,p_2<0.001*,p_3<0.001***				

Child score	**Group I**		**Group II**	
Scoring	**No.**	**%**	**No.**	**%**
A	**10**	**33.3**	**5**	**16.6**
B	**15**	**50**	**17**	**56.6**
C	**5**	**16.6**	**8**	**26.6**

BCLC	**Group II**	**Percent %**
0	**0**	**0**
A	**5**	**16.7**
B	**10**	**33.3**
C	**7**	**23.3**
D	**8**	**26.7**

	Group I (n = 30)	**Group II (n = 30)**	**Group III (n = 30)**	**H**	**p**
Serum AFP level					
Min. – Max.	**2.70 – 17.20**	**2.80 – 870.0**	**0.0 – 21.0**	**30.402***	**<0.001***
Mean ± SD.	**8.11 ± 3.07**	**151.4 ± 252.9**	**6.03 ± 4.92**		
Median (IQR)	**8.05 (5.70 – 10.0)**	**26.0 (9.0 – 198.0)**	**4.0(3.0 – 9.0)**		
Sig. bet. grps.	**p_1<0.001*,p_2<0.044*,p_3<0.001***				

	Group I (n = 30)	**Group II (n = 30)**	**Group III (n = 30)**	**F**	**p**
Serum milk growth factor-8 (ng/l)					
Min. – Max.	**3.51 – 7.76**	**1.51 – 4.82**	**4.81 – 6.82**	**65.692***	**<0.001***
Mean ± SD.	**5.25 ± 1.11**	**3.20 ± 0.98**	**5.76 ± 0.57**		
Median (IQR)	**5.33 (4.40 – 5.93)**	**3.08 (2.55 – 4.01)**	**5.71 (5.31 – 6.22)**		
Sig. bet. grps.	**p_1<0.001*,p_2=0.087,p_3<0.001***				

χ^2: Chi square test

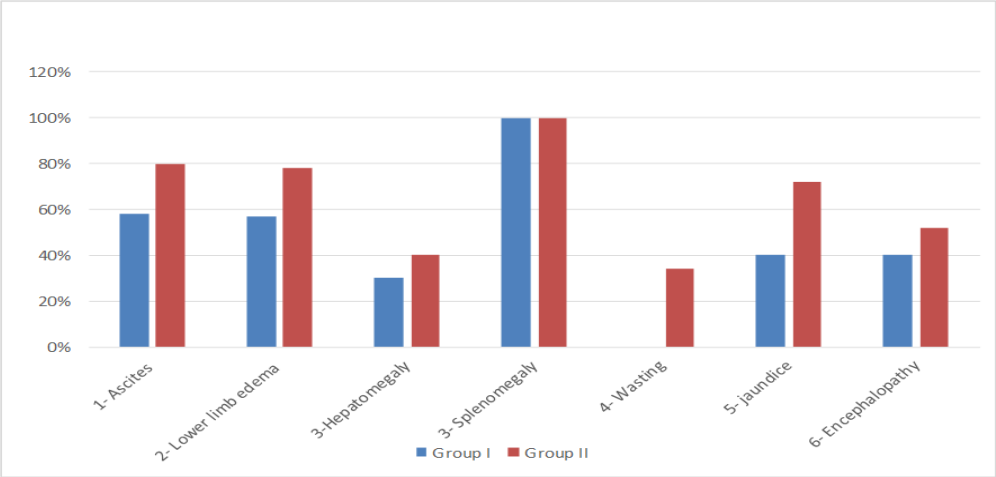
H: H for Kruskal Wallis test, pairwise comparison bet. each 2 groups was done using Post Hoc Test (Dunn's for multiple comparisons test)

F: F for ANOVA test, pairwise comparison bet. each 2 groups was done using Post Hoc Test (Tukey)

p: p value for comparing between the studied groups

p_0: p value for comparing between group IV and each other group

p_1: p value for comparing between group I and II
p_2: p value for comparing between group I and III
p_3: p value for comparing between group II and III
*: Statistically significant at $p \leq 0.05$
IQR: Inter quartile range SD: Standard deviation
Group I: cirrhosis without HCC
Group II: cirrhosis with HCC
Group III: control



Group I: Patients with HCV liver cirrhosis without HCC, Group II: Patients with HCV liver cirrhosis with HCC

Figure 2. Distribution of the studied cases in according to signs in each group**.**

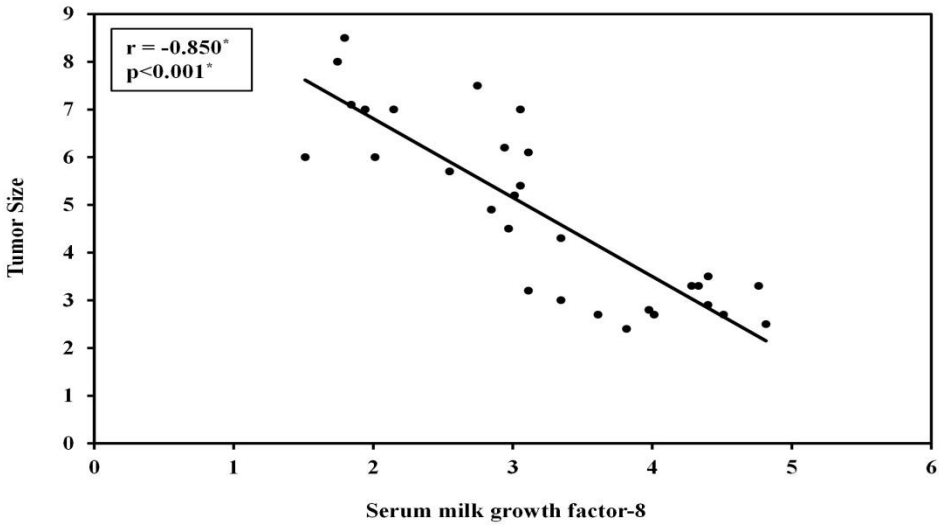


Figure 3. Correlation between Serum milk growth factor-8 (ng/l) with Tumor Size in group II

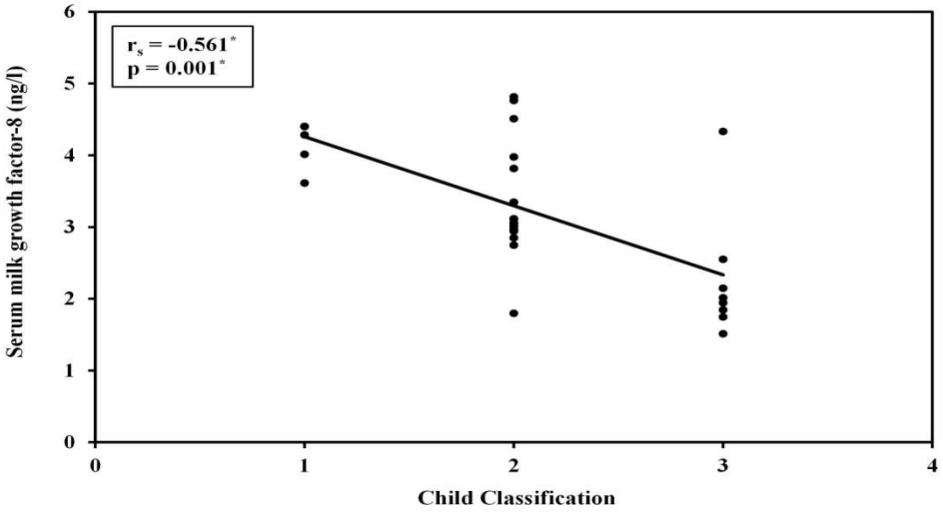


Figure 4. Correlation between serum milk growth factor-8 (ng/l) with child classification in group II (Patients with HCV liver cirrhosis with HCC) (n = 30)

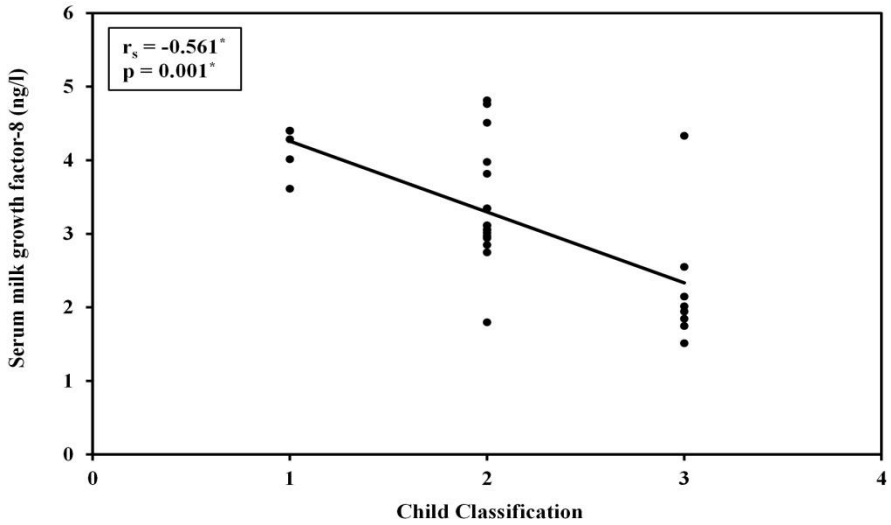


Figure 5. Correlation between serum milk growth factor-8 (ng/l) with BCLC in group II (Patients with HCV liver cirrhosis with HCC) (n = 30)

Table 2. Diagnostic performance for serum AFP level and serum milk growth factor-8 to discriminate group II from group I

	AUC	p	95% C.I	Cut off	Sensitivity	Specificity	PPV	NPV
Serum AFP level	**0.797**	**<0.001***	**0.676 – 0.917**	**>10**	**70.0**	**76.67**	**75.0**	**71.9**
Serum milk growth factor-8 (ng/l)	**0.916**	**<0.001***	**0.849 – 0.982**	**≤4.014**	**76.67**	**80.0**	**79.3**	**77.4**
Combination	**0.961**	**<0.001***	**0.919 – 1.000**		**86.67**	**90.0**	**89.66**	**87.10**

DISCUSSION

Chronic liver illness, especially liver cirrhosis, increases the risk of HCC. (1) It is the fifth most prevalent malignant tumor and the second main cause of cancer deaths internationally [8, 9].

Some early-stage HCC patients undergo surgical resection, guided ablation, and transplantation. Thus, high-risk HCC patients should be monitored to improve survival. In addition to serum AFP, new recommendations include abdominal ultrasonography for high-risk individuals [10].

A new meta-analysis study [11] revealed that ultrasound-based HCC monitoring has a 45% sensitivity and 21%–89% heterogeneity, demonstrating its limitations. Thus, validated blood-based monitoring approaches with greater diagnostic accuracy are required.

Despite their wide clinical use, AFP is not sufficient for small-sized HCCs early detection. [12] (MFG-E8), is a glycoprotein that was first identified in mammary epithelial cells [13]. However, serum levels between healthy and cirrhotic livers were comparable. Recent research revealed that MFG-E8 expression was reduced in liver cirrhosis [14].

In the current research, a considerable percentage of the participants (56.6 %) were CPC B. However, there was a difference with no significance statistically between the two research groups in terms of the CPC.

According to Sheta et al. [15] there was a difference with no statistical significance in CPC between both cirrhotic and HCC patients as the majority of HCC cases (68.8%) and cirrhotic cases (64.6%) in their study were assigned to CPC B. This suggests the same distribution of CPC among the study groups compared to our research.

In the current work, the majority of patients were categorized into BCLC B categories, comprising 33.6 percent of the participants.

According to our research, participants with HCC had an average AFP of 151.4 ng/mL, much greater than HCV liver cirrhotic patients who did not have HCC (8.11 ng/mL) or healthy volunteers (6.03 ng/mL), with a difference that was statistically significant among the latter two study groups. Similarly, Murugavel K et al. study, [16] found that the AFP level in patients with HCC complicating HCV was 492 ng/mL. This is comparable to the results of the Ball D et al. research, which showed that AFP levels in 270 healthy individuals were 3 ng/mL [17].

According to our investigation, AFP's sensitivity, specificity, PPV, and NPV were 70.0%, 76.67%, 75.0, and 71.9% respectively at the 10 ng/mL threshold. The values for sensitivity and NPV illustrate the AFP value as an HCC screening method, however false negative results were reported in 10 patients. This agrees with the the study conducted by Shimagaki T et al, [18] which reported that, at a 10 ng/mL concentration, the AFP test's sensitivity, specificity, (PPV), and (NPV) were, respectively, 68.3%, 75.2%, 73.3%, and 70.5%. This shows that almost 45% of patients who had normal AFP levels were, in fact, HCC patients.

Additionally, these outcomes are consistent with a recent meta-analysis, which revealed that abdominal ultrasonography and AFP combined only had a 63% sensitivity and that up to 50% of HCCs had normal AFP levels. Large cohort studies have also shown that AFP has a sensitivity between 39% and 64% for detecting early HCC stage and a specificity between 76% and 97% [19, 20]. Different AFP cutoff values vary across studies; however, a 10 ng/mL AFP level is widely accepted [18].

In contrast to healthy volunteers (mean = 5.76 ng/L), this research discovered that MFG-E8 levels in blood were considerably lower in cirrhotic group with HCC (mean = 3.2 ng/L). The HCC and the HCV liver cirrhosis groups differed significantly (mean = 5.25ng/L).

This was consistent with study findings by Shimagaki T et al., [18] which showed that HCC patients had considerably lower serum MFG-E8 levels than healthy subjects ($p<0.0001$) and lower than among HCV participants with liver cirrhosis ($p<0.0001$).

Our research revealed that the sensitivity, specificity, PPV, and NPV of MFG-E8 were, respectively, 76.67%, 80.0%, 79.3%, and 77.4% at a level of ≤ 4.01 ng/L. This shows that the diagnostic serum MFG-E8 biomarker performance is high and it’s superior to that of serum AFP.

According to Shimagaki T et al. research, [18] Sensitivity, specificity, PPV, and NPV of MFG-E8 were 69.7%, 84.3%, 88.4%, and 61.9%,

respectively. These findings are consistent with our findings which predict very good performance for our study marker.

The study by Hong H et al., [21] demonstrated that the of the serum biomarker MFG-E8 both sensitivity and specificity at 2.19 ng/L were, respectively, 93.3% and 90%, while the AUC was 0.987. These findings illustrate the very good diagnostic and screening performance of the serum MFG-E8 biomarker, which is largely consistent with our findings.

There are several hypotheses explaining the unclear serum MFG-E8 reduction mechanism in HCC entailing that; HCC could decrease the MFG-E8 production, promote the MFG-E8 lysis or increase the uptake of MFG-E8.

Yang C, [7] showed that mesenchymal stem cells release MFG-E8 that hampers the hepatic stellate cells activation, which lead to reduction in liver-involved fibrosis, as observed in both laboratory and living organism settings. So, MFGE8 serves as an anti-fibrotic protein within MSC secretions, exerting potent suppression on TGFβ signaling resulting in diminished extracellular matrix accumulation [7].

A study by An GH et al., [22] showed that decreased levels of MFG-E8 promote the development of liver cirrhosis and HCC because they promote macrophage-involved collagen uptake and reduce fibrosis [22].

Additionally, we did not examine the link between the MFG-E8 expression in tissues of the liver and the MFG-E8 serum level to determine whether there is a positive or negative correlation between them.

CONCLUSION

In conclusion, Patients with HCV-induced liver cirrhosis might employ serum Lactadherin as a helpful hepatocellular cancer diagnostic tool. Moreover, it can be used in HCC detection in AFP-negative individuals.

Funding: None. Author funded.

Conflict of Interest: None.

Acknowledgment

The authors would thank all patients who participated in the study.

Ethical approval

This research was approved by the ethics committee of the Faculty of Medicine at the University of Alexandria. All participants provided their written, informed consent.This research was conducted in line with ethical principles.. The Committee's serial number is 0106257 and the reference number is FWA NO: 00018699.

HIGHLIGHTS

- HCC prognosis improves with early discovery, since therapy may improve survival.
- Thus, developing reliable biomarkers to aid early HCC diagnosis in cirrhotic individuals is a priority.
- AFP is a major marker for HCC, however its low sensitivity and specificity prohibit it from being used as the principal monitoring test for HCC.
- Serum levels of Lactadherin might be regarded as a possible biomarker to predict the HCC development and progression among those who are cirrhotic.

REFERENCES

1. World Health Organization (WHO). Hepatitis C. Geneva, Switzerland*: WHO*; 2022.
2. Nguyen LH, Nguyen MH. Systematic review: Asian patients with chronic hepatitis C infection. *Aliment Pharmacol Ther* 2013; 37(10): 921-36.
3. El-Serag HB. Epidemiology of hepatocellular carcinoma. *Clin Liver Dis* 2001; 5(1): 87-107.
4. Paganoni R, Lechel A, Vujic Spasic M. Iron at the Interface of Hepatocellular Carcinoma. *Int J Mol Sci* 2021; 22(8): 4097.
5. Chen DS, Sung JL, Sheu JC, Lai MY, How SW, Hsu HC, et al. Serum alpha-fetoprotein in the early stage of human hepatocellular carcinoma. *Gastroenterology* 1984; 86(6): 1404-9.
6. Hanayama R, Tanaka M, Miwa K, Shinohara A, Iwamatsu A, Nagata S. Identification of a factor that links apoptotic cells to phagocytes. *Nature* 2002; 417(6885): 182-7.
7. Yang C, Hayashida T, Forster N, Li C, Shen D, Maheswaran S, et al. The integrin alpha(v)beta(3-5) ligand MFG-E8 is a p63/p73 target gene in triple-negative breast cancers but exhibits suppressive functions in ER(+) and erbB2(+) breast cancers. *Cancer Res* 2011; 71(3): 937-45.
8. Yoon JC, Yang CM, Song Y, Lee JM. Natural killer cells in hepatitis C: Current progress. *World J Gastroenterol* 2016; 22(4): 1449-60.

9. Rosen HR. Clinical practice. Chronic hepatitis C infection. *N Engl J Med* 2011; 364(25): 2429-38.
10. Nada Y, Rashad N, Eissa M, Ghonaim A, Farag K, Saadawi I, et al. Outcomes of treatment with sorafenib in Egyptian patients with hepatocellular carcinoma: a retrospective cohort study. *Expert Rev Gastroenterol Hepatol* 2018; 12(1): 99-107.
11. Tzartzeva K, Obi J, Rich NE, Parikh ND, Marrero JA, Yopp A, et al. Surveillance imaging and alpha fetoprotein for early detection of hepatocellular carcinoma in patients with cirrhosis: A meta-analysis. *Gastroenterology* 2018; 154(6): 1706-18.
12. Tsai SL, Huang GT, Yang PM, Sheu JC, Sung JL, Chen DS. Plasma des-gamma-carboxyprothrombin in the early stage of hepatocellular carcinoma. *Hepatology* 1990; 11(3): 481-8.
13. Wan HG, Xu H, Gu YM, Wang H, Xu W, Zu MH. Comparison osteopontin vs AFP for the diagnosis of HCC: a meta-analysis. *Clin Res Hepatol Gastroenterol* 2014; 38(6): 706-14.
14. Aziz M, Jacob A, Matsuda A, Wang P. Review: milk fat globule-EGF factor 8 expression, function and plausible signal transduction in resolving inflammation. *Apoptosis* 2011; 16(11): 1077-86.
15. Sheta T, Selim M, Sabry M, Saed A. Serum thioredoxin as a diagnostic marker for hepatocellular carcinoma in cirrhotic hepatitis C patients. *Med J Viral Hepat* 2021; 5(2): 9-15.
16. Murugavel KG, Mathews S, Jayanthi V, Shankar EM, Hari R, Surendran R, et al. Alpha-fetoprotein as a tumor marker in hepatocellular carcinoma: investigations in south Indian subjects with hepatotropic virus and aflatoxin etiologies. *Int J Infect Dis* 2008; 12(6): e71-6.
17. Ball D, Rose E, Alpert E. Alpha-fetoprotein levels in normal adults. *Am J Med Sci* 1992; 303(3): 157-9.
18. Shimagaki T, Yoshio S, Kawai H, Sakamoto Y, Doi H, Matsuda M, et al. Serum milk fat globule-EGF factor 8 (MFG-E8) as a diagnostic and prognostic biomarker in patients with hepatocellular carcinoma. *Sci Rep* 2019; 9(1): 15788.
19. Marrero JA, Feng Z, Wang Y, Nguyen MH, Befeler AS, Roberts LR, et al. Alpha-fetoprotein, des-gamma carboxyprothrombin, and lectin-bound alpha-fetoprotein in early hepatocellular carcinoma. *Gastroenterology* 2009; 137(1): 110-8.
20. Oka H, Tamori A, Kuroki T, Kobayashi K, Yamamoto S. Prospective study of alpha-fetoprotein in cirrhotic patients monitored for development of hepatocellular carcinoma. *Hepatology* 1994; 19(1): 61-6.
21. Hong H, Wang Z, Li L. The level of serum MFG-E8 in liver cancer and its metastasis in liver cancer. *World J Cancer Res* 2017; 4: 70-7.
22. An GH, Lee J, Jin X, Chung J, Kim JC, Park JH, et al. Truncated Milk Fat Globule-EGF-like Factor 8 Ameliorates Liver Fibrosis via Inhibition of Integrin-TGFβ Receptor Interaction. *Biomedicines* 2021; 9(11): 1529.