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Research Article

Relation between Fecal Calprotectin concentration and severity of Hepatitis C (HCV) related chronic liver disease

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Abstract

Background and Aim: Egypt has one of the highest prevalence rates of hepatitis C virus (HCV) infection in the world [1]. Complications of liver cirrhosis especially hepatic encephalopathy represent a major impact among hepatic patients [2]. Calprotectin is a calcium and zinc-binding peptide, proposed as a biomarker for various inflammatory diseases. As the gastrointestinal tract of cirrhotic patients shows various alterations of its mucosal barrier including infiltrates of neutrophils, calprotectin might be a promising diagnostic parameter to diagnose the onset of hepatic encephalopathy [3]. We aimed to assess the relation between fecal calprotectin concentrations and severity of HCV related chronic liver disease. **Patients and Methods:** This study was conducted on forty five HCV-related chronic liver disease patients attending Ain shams university and Alexandria university Hospital during the period from November 2013 to March 2014 and was subdivided according to Modified Child_Pugh's classification into 3 groups with twenty healthy volunteers were selected as controls. Fecal calprotectin concentrations (FCCs) was assayed by an enzyme-linked immunosorbent assay in all patients and control group. **Results:** The comparison between cases and control groups as regard FCCs, showed that it was significantly higher among patient's groups with a mean FCCs (52.19 ± 16.99), (182.50 ± 74.81) and (311.47 ± 90.92) in child A, child B and child C respectively compared to (20.55 ± 13.58) in control group. A highly positive correlation between FCCs, ESR, CRP and serum ferritin in total patients groups were found, while there were no significant correlation between FCCs and WBCs. A highly significant positive relationship between FCCs and INR, S.albumin, total and direct bilirubin in total patients groups were demonstrated, while there were no significant correlation between FCCs and AST, ALT. Although we found a positive correlation between FCCs and serum ammonia only in cases groups, there was no corresponding significant correlation between FCCs and hepatic encephalopathy in patients groups. **Conclusions:** FCCs are elevated in patients with HCV-related chronic liver disease which were significantly elevated dependent on the severity of liver disease as assessed by Modified Child_Pugh's classification. So, FCCs can be used as a valid parameter in assessment of the severity of hepatitis C chronic liver disease.

Keywords: Fecal calprotectin concentrations, Chronic liver disease.

1.Introduction

HCV was first identified in 1989 as the major causative agent of parentally transmitted and community acquired non-A non-B hepatitis [4]. Chronic HCV infection is usually slowly progressive and may not result in clinically apparent liver disease in many patients if the infection is acquired later in life. Approximately 20 to 30 percent of chronically infected individuals develop cirrhosis over a 20- to 30-year period of time [5]. From the pathophysiological point of view, numerous alterations in intestinal flora, mucosal barrier functions and immunological defense mechanisms occur in cirrhotic patients [6]; this leads to bacterial overgrowth ranging from 30% to 64% and seems to represent one of the main factors to trigger

bacterial translocation [7,8]. The gut flora and bacterial translocation play an important role in the pathogenesis of certain complications of cirrhosis like hepatic encephalopathy [9]. Calprotectin is a calcium and zinc-binding peptide, proposed as a biomarker for various inflammatory diseases. As an acute phase reactant, calprotectin increases more than 100 folds during inflamed conditions [10]. Calprotectin is found in monocytes [11], keratinocytes [12], muscle tissue [13] and infiltrating tissue macrophages [14]. Calprotectin is also found abundant in neutrophils [15] and it constitutes 30-60% of the cytosolic proteins [16]. Once get stimulated by an injury or cell disruption, neutrophils and monocytes start secreting calprotectin into the extra cellular fluid [17]. Accordingly, the presence of fecal calprotectin quantitatively relates to intestinal neutrophil migration [18] and is therefore, it may be considered as a valid marker of intestinal inflammation [19].

2. Patients and Methods:

2.1. Study Design and Duration:

This is a prospective study, during the period from November 2013 to March 2014.

2.2. Patients:

A total of 45 patients with HCV-related chronic liver disease diagnosed by serological, biochemical and ultrasonographic evidence and 20 healthy volunteers as control were enrolled in this study attending Ain Shams University Hospital and Alexandria university Hospital after signing a written consent.

The patients group was subdivided according to Modified Child_Pugh's classification into:

- Goup (1): Child A: includes 15 patients (33% of patients).
- Group (2): Child B: includes 15 patients (33% of patients).
- Group (3): Child C: includes 15 patients (33% of patients).

Inclusion criteria:

All patients with HCV related chronic liver disease of varying degree of severity unless there is a cause for exclusion.

Exclusion criteria:

Patients with the following conditions were excluded:

- Active GIT bleeding.
- Inflammatory bowel disease.
- Colorectal cancer.
- Patients with major surgical operation, active infection.
- Inflammatory joint disease.
- Pancreatic disease.
- Concomitant HBV infection or other causes of chronic liver disease as Hemochromatosis, Autoimmune hepatitis, Wilson disease or Alcoholic hepatitis.

Ethical Considerations

This study has been performed in accordance with the ethical standards. Signed consent was obtained from all patients before enrollment in the study. Right to refuse participation was emphasized.

2.3. Methodology:

2.3.1. Clinical, Laboratory and radiological evaluation:

i. Full history taking stressing on residence, history of alcohol intake, history of schistosomiasis and previous parenteral treatment for it, history of blood transfusion, dental procedure and operations.
ii. Clinical examination.
iii. Laboratory Investigations including:

- CBC, ESR and CRP.
- Liver function tests {ALT, AST, Total bilirubin, Alkaline phosphatase, Serum albumin, Prothrombin time, Serum ammonia}.
- HBs Ag.
- Autoimmune markers including: Antinuclear antibody (ANA), Antismooth muscle antibody, Antimitochondrial antibody (AMA).
- Serum ferritin level, serum ceruloplasmin.
- Serum amylase and lipase levels
- CEA, AFP.
- Occult blood in stool.

- Measurement of FCCs using single stool sample to be promptly stored at -20 C until laboratory testing.

iv. Evaluation of the severity of liver cirrhosis was obtained in each cirrhotic patient with Child-Turcotte-Pugh score. This system relies on clinical and laboratory evaluation including ascites, grade of encephalopathy, serum albumin, bilirubin and prothrombin time [20].
v. Abdominal ultrasound to assess liver texture, splenic size and amount of ascites.

2.3.2. Test principle:

Collection of samples:

Calprotectin was measured in a single stool sample in all patients and controls. Samples were delivered on the day of the investigation and stored in a refrigerator before transfer to the study laboratory within 48 hours for analysis. Calprotectin is stable up to seven days at room temperature [21].

Principle of the Test:

Fecal calprotectin was assayed by an enzyme-linked immunosorbent assay according to the manufacturer's instructions (Phi-Cal Calprotectin ELISA Kit; Immundiagnostik AG, Bensheim, Germany) using polyclonal antibody against calprotectin into monoclonal antibody with high affinity against calprotectin. Aliquots of approximately 100 mg of feces (range 80–120 mg) were homogenized for 25 min with extraction buffer. After centrifugation for 20 min at 3000 g, the supernatant was diluted 1:50 with dilution buffer and calprotectin was measured with an enzyme linked buffer in immunoadsorbent assay [22].

Results:

Normal ranges: according to the manufacture's instructions, the median value in healthy adults is about 25 mg/kg while samples giving values above 50 mg/kg are regarded as positive. As the manufacture recommends each laboratory to establish its own normal concentration range, we defined the ranges as follows: 1 = normal calprotectin (FCCs <6 mg/kg), 2 = slightly elevated calprotectin (FCCs >30 mg/kg <48 mg/kg), 3 = explicitly elevated calprotectin (FCC>48 mg/kg).

2.3.4. Statistical Methods:

SPSS statistical software package (V. 17.0, Echo soft Corp., USA, 2008) was used for data analysis. Results were expressed as means ± standard deviation of the means (SD). Differences between groups were analyzed either by using the Chi square test or student's t test and nonparametric (Mann Whitney test) for comparison between two groups or ANOVA test for multiple group comparison. Spearman rank correlation coefficient was used to determine significant correlations among different parameters. The analysis was performed using Statistical Analysis System, version 6.03, on an IBM at personal computer.

1-SD: slandered deviation.
2-M: mean.
3-P: P-value.

Results

The present study was conducted on forty five HCV-related chronic liver disease patients, subdivided according to Modified Child Pugh's classification into:

- Group (1) Child A: includes 15 patients (33% of patients).
- Group (2) Child B: includes 15 patients (33% of patients).
- Group (3) Child C: includes 15 patients (33% of patients).

Twenty healthy volunteers were selected as controls.

As regard demographic data, there was no statistical significant difference between studied groups as regard age, gender and body mass index.

Concerning stigmata of end-stage liver disease, there was high statistically significant difference as regard palmer erythema, spider naevi, lower limb edema, clinical jaundice, gynaecomastia and ascites between the studied groups, P<0.001 (H.S), while there was no statistical significant difference between the studied groups as regard HE , P>0.05 (NS) Table (1)

Table (1): Comparison between studied groups as regard stigmata of end-stage liver disease.

	Control (n=20)		Child A (n=15)		Child B (n=15)		Child C (n=15)		MCp
	No.	%	No.	%	No.	%	No.	%	
Jaundice									
-ve	20	100.0	15	100.0	14	93.3	4	26.7	<0.001*
+ve	0	0.0	0	0.0	1	6.7	11	73.3	
Spider naevi									
-ve	20	100.0	15	100.0	13	86.7	10	71.4	0.009*
+ve	0	0.0	0	0.0	2	13.3	5	28.6	
Palmer erythema									
-ve	20	100.0	15	100.0	11	73.3	6	42.9	<0.001*
+ve	0	0.0	0	0.0	4	26.7	9	57.1	
Gynaecomastia									
-ve	20	100.0	15	100.0	12	80.0	10	41.7	0.006*
+ve	0	0.0	0	0.0	3	20.0	5	28.6	
LL edema									
-ve	20	100.0	15	100.0	9	60.0	0	0.0	<0.001*
+ve	0	0.0	0	0.0	6	40.0	15	100.0	
HE									
I	0	0.0	0	0.0	1	6.7	0	0.0	0.078
II	0	0.0	0	0.0	2	13.3	5	33.3	
III	0	0.0	0	0.0	0	0.0	2	13.3	
IV	0	0.0	0	0.0	0	0.0	2	13.3	
Ascites									<0.001
-ve	0	100.0	15	100.0	0	0.0	0	0.0	
+ve	0	0	0	0.0	15	100.0	15	100.0	

Concerning biochemical data of the studied groups, we showed a high significant difference between the studied groups as regard hemoglobin level, platelet concentration, total bilirubin, ALP, INR, S. albumin, AFP and prothrombin percent activity P<0.001 (H.S), while there was no statistical significant difference between the studied groups as regard WBCs, ALT and AST, P>0.05 (NS).

Estimation of FCCs among studied groups were significantly higher among patient's groups with a mean FCCs (52.19 ± 16.99), (182.50 ± 74.81) and (311.47 ± 90.92) in child A, child B and child C respectively compared to (20.55 ± 13.58) in control group, P<0.001 (H.S) (table 3, figure 1)

Table (2): Comparison between the studied groups as regard biochemical data.

	Control (n=20)	Child A (n=15)	Child B (n=15)	Child C (n=15)	P
Hb (g / dl) Mean ± SD	14.55 ± 1.48	12.83 ± 2.04	11.78 ± 1.38	10.61 ± 1.61	<0.001
WBCs (10^3/cm) Mean ± SD.	6.03 ± 2.41	6.20 ± 2.31	6.15 ± 3.80	6.54 ± 4.01	0.751
Platelet (10^3/cm) Mean ± SD.	246.05 ±71.02	184.07 ± 86.50	150.53 ± 47.07	142.80 ± 55.33	0.021
T.Bilirubin (mg /l) Mean ± SD.	0.67 ± 0.26	0.83 ± 0.44	1.21 ± 0.47	3.18 ± 1.43	<0.001
ALk. P (U/L) Mean ± SD.	111.70 ±31.94	82.80 ± 38.84	115.0 ± 50.34	145.87 ± 47.94	0.002
ALT (U/L) Mean ± SD.	76.30 ±45.89	61.80 ± 33.65	56.33 ± 22.93	63.41 ± 49.98	0.601
AST (U/L) Mean ± SD.	66.60 ±44.42	61.07 ± 32.96	56.47 ± 42.37	66.93 ± 37.09	0.585
S.Alb (g/dl) Mean ± SD	4.35 ± 0.62	4.14 ± 0.44	2.91 ± 0.35	2.16 ± 0.56	<0.001
Protrombin percent activity Mean ± SD.	90.75 ± 3.80	87.0 ± 7.69	75.0 ± 7.76	71.27 ± 8.28	<0.001

Table (3): Comparison between the studied groups according to FCCs.

	Control (n=20)	Child A (n=15)	Child B (n=15)	Child C (n=15)	P
FCCs (mg/kg) Min. - Max.	4.0 – 48.0	24.30–80.50	65.50–302.0	170.0–52.0	<0.001*
Mean ± SD	20.55±13.58	52.19±16.99	182.50±74.81	311.47±90.92	

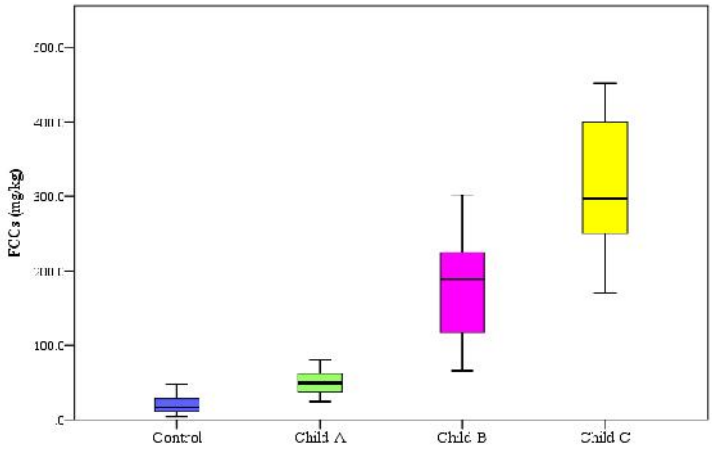

Figure (1): Comparison between the studied groups according to FCCs.

There was no significant correlation between FCCs and HE in patient groups, P>0.05 (NS), while considering correlation between FCCs and acute phase reactants (WBCs, ESR and serum ferritin) in each studied group, it was highly positive between FCCs, ESR and serum ferritin in total patients, P<0.001 (H.S), while there were no significant correlation between FCCs and WBCs, P>0.05 (NS) Table (4).

Table (4): Correlation between FCCs with WBCs, ESR and serum ferritin in each studied group.

		FCCs				
		Control	**Group A**	**Group B**	**Group C**	**Total Patients**
WBCs	**R**	0.183	0.176	0.479	0.061	0.099
	P	0.440	0.530	0.071	0.829	0.518
ESR 1st	**R**	-	0.061	0.165	-0.065	0.542*
	P	-	0.830	0.556	0.818	<0.001
Serum ferritin	**R**	-	0.468	0.502	0.412	0.792*
	P	-	0.066	0.057	0.127	<0.001

r: Pearson coefficient
*: Statistically significant at $p \leq 0.05$

Table (5) showed the correlation between FCCs with different biochemical parameters including AST, ALT, INR, S.albumin, total and direct bilirubin in the control and cases groups showing a highly significant positive correlation between FCCs and INR, S.albumin, total and direct bilirubin in total patient P<0.001 (H.S), while there were no significant correlation between FCCs and AST, ALT, P>0.05 (NS).

Table (5): Correlation between FCCs with liver function tests.

		FCCs				
		Control	**Group A**	**Group B**	**Group C**	**Total patients**
T.Bilirubin	**R**	-0.126	-0.426	0.221	-0.327	0.583*
	P	0.596	0.114	0.428	0.235	<0.001
D.Bilirubin	**R**	-0.035	-0.111	0.290	-0.286	0.548*
	P	0.882	0.694	0.295	0.302	<0.001
S. Alb	**R**	0.325	-0.186	0.196	-0.014	-0.729*
	P	0.162	0.506	0.484	0.960	<0.001
INR	**R**	-0.238	-0.125	-0.385	0.013	0.555*
	P	0.313	0.658	0.157	0.964	<0.001
ALT	**R**	0.261	0.241	-0.111	-0.445	-0.133
	P	0.267	0.386	0.693	0.095	0.385
AST	**R**	-0.040	-0.263	-0.077	0.015	0.061
	P	0.866	0.343	0.784	0.958	0.689

From the ROC curve, cut off value of FCCs for differentiation between cases groups and control group was 48 mg/kg with a sensitivity of 84.44%, a specificity of 100%, a negative predictive value of 74.07%, a positive predictive value of 100% and an accuracy of 89.23%. (Table 6, figure 2), while cut off value of FCCs for differentiation between child A group and child B group was 77 mg/kg with a sensitivity of 93.33%, a specificity of 93.33%, a negative predictive value of 93.33%, a positive predictive value of 93.33% and a accuracy of 93.33%. (Table 7, figure 3)

Table (6): Diagnostic validity test for FCCs with cases and control group.

Cut off		Control	Cases	Sensitivity	Specificity	PPV	NPV	Accuracy
FCCs	≤48	20	7	84.44	100.0	100.0	74.07	89.23
	>48	0	38					

AUC: Area under the curve; PPV: Positive predictive value; NPV: Negative predictive value

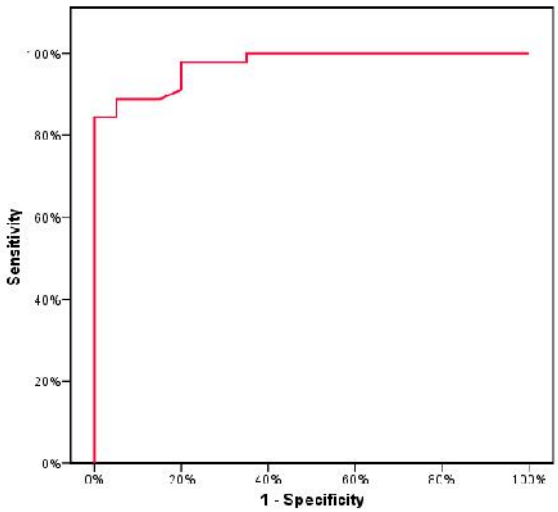

Figure (2): ROC curve for FCCs with cases and control groups.

Table (7): Diagnostic validity test for FCCs with A and B groups.

		A	B	Sensitivity	Specificity	PPV	NPV	Accuracy
FCCs	≤77	14	1	93.33	93.33	93.33	93.33	93.33
	>77	1	14					

AUC: Area under the curve; PPV: Positive predictive value; NPV: Negative predictive value

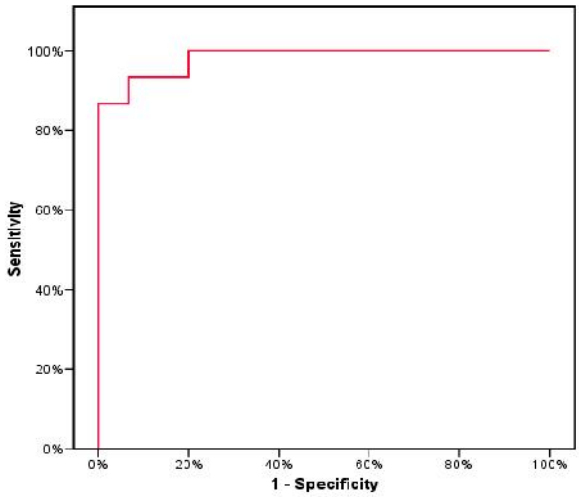

Figure (3): ROC curve for Diagnostic validity test for FCCs with A and B group.

Finally, our results clarified that the cut off value of FCCs for differentiation between child B group and child C group was 212 mg/kg with a sensitivity of 86.67%, a specificity of 73.33%, a negative predictive value of 84.62%, a positive predictive value of 76.47% and an accuracy of 80% (Table 8, figure 4).

Table (8): Diagnostic validity test for FCCs with B and C group

		B	C	Sensitivity	Specificity	PPV	NPV	Accuracy
FCCs	**≤212**	11	2	86.67	73.33	76.47	84.62	80.0
	>212	4	13					

AUC: Area under the curve; PPV: Positive predictive value; NPV: Negative predictive value

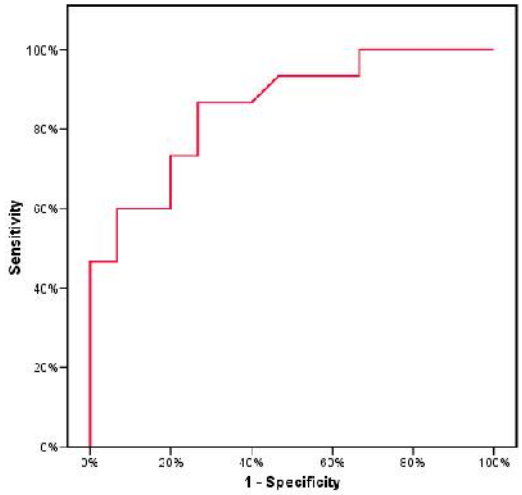

Figure (4): ROC curve for Diagnostic validity test for FCCs with B and C group.

Discussion

HCV infection occurs worldwide, with the prevalence of antibody to HCV (anti-HCV) in serum in most developed countries ranging between 1% and 2%. Globally, it was estimated that in 2005, more than 185 million people had HCV antibodies (prevalence of 2.8 percent) [5]. Significantly higher rates of infection have been found in parts of Eastern Europe and Africa, while Egypt seems to have one of the highest prevalence rates of all, approaching more than 15% of the general population [5]. HCV is a major cause of chronic hepatitis, which frequently leads to cirrhosis and also hepatocellular carcinoma. Up to 30% of patients with CHC may develop cirrhosis [23]. Calprotectin is a 36-kDa calcium and zinc binding protein and constitutes approximately 60% of soluble cytosol proteins in human neutrophil granulocytes [24]. Calprotectin is a surrogate marker of neutrophil turnover and is elevated in a number of inflammatory conditions [12].The soluble form of calprotectin provides both bacteriostatic and cytokine like effects in the local environment [17].

Bacterial translocation (BT) is a healthy phenomenon but is pathologically increased in quantity in liver cirrhosis. Whereas rate and degree of translocating bacterial products is increased in early cirrhosis, pathological translocation of viable bacteria occurs in the decompensated stage [6]. The presence of fecal calprotectin quantitatively relates to intestinal neutrophil migration [3]. Therefore fecal calprotectin could be a promising parameter in the assessment of the severity of HCV related chronic liver disease.

In the present study, fecal calprotectin was investigated on forty five patients with HCV related chronic liver disease in comparison to twenty healthy volunteers and found to be significantly higher in patients groups compared with control group [P< 0.001], despite of a careful exclusion of other causes of abnormal calprotectin results.

Likewise, Gundling et al., [3] reported that fecal calprotectin levels were elevated in chronic hepatitis C patients compared to controls. It is also in agreement with [25], who found that calprotectin level is elevated in patients with liver cirrhosis but in faeces rather than in plasma.

In our study, patients were divided according to the severity of liver disease as assessed by modified Child Pugh score into three groups, child A, B and C. The elevated FCCs in this study showed a significantly positive correlation with the severity of liver disease. In the patient's group, mean FCCs (52.19 ± 16.99), (182.50 ± 74.81) and (311.47 ± 90.92) in child A, child B and child C respectively compared to (20.55 ± 13.58) in control group, ($p<0.001$).

This is in agreement with, Yagmur et al., [25], who found a significant elevation in FCCs in patients with advanced chronic liver disease and additionally added that there was a trend towards higher levels of fecal calprotectin in subjects with alcoholic cirrhosis. Interestingly, Yagmur and his collagues described in their study that the highest of all calprotectin concentrations was in two cirrhotic patients with SBP.

Our study showed that there was a significant positive correlation between FCCs level with serum bilirubin and INR ($P<0.05$), but inversely correlated with serum albumin. This is consistent with, Elbanna et al., [26], who found that the elevation in calprotectin level especially in ascitic fluid correlates with the severity of liver disease as there was a significant positive correlation with serum bilirubin and inverse correlation with serum albumin and prothrombin activity, however plasma calprotectin in that study did not correlate much with the severity of liver disease.

Yagmur et al., [25], stated that plasma level of calprotectin did not correlate with the severity of chronic liver diseases other than alcoholic liver disease. Homann et al., [27], stated also that plasma calprotectin concentrations were lower in viral liver disease than in non viral liver disease. Elbanna et al., [26], added that perhaps the elevation in plasma calprotectin level in patients with alcoholic liver disease in particular, can be explained by the several abnormalities of neutrophil function existing in these patients.

Homann et al., [27], investigated the prognostic value of plasma and ascites calprotectin in cirrhosis. They demonstrated that plasma calprotectin was a highly significant marker of poor survival in alcohol-induced cirrhosis as high calprotectin concentrations were significantly associated with poor survival .The same authors found no association between increased plasma calprotectin concentrations and the severity of liver disease other than alcoholic liver disease. Furthermore, Homann firstly described that high plasma calprotectin levels may characterize a group of cirrhotics with recurring bacterial infections.

In the present study there was no significant correlation between FCCs and serum aminotrasferases (ALT, AST), which is not in agree with Elbanna et al., [26], who concluded that calprotectin is elevated in hepatitis C related liver cirrhosis and its level, particulary in ascitic fluid, correlates with the degree of hepatocellular injury and added that it is not elevated with the occurrence of hepatocellular carcinoma.

We found no significant influence ($P = 0.518$) of WBC count on FCCs in our patients and control groups. But there was a positive significant correlation between FCCs and other inflammatory activity as ESR, CRP, serum ferritin ($P<0.05$). On the other hand, it apparently disagrees with, Gundling et al., [3], who reported that there was no significant influence ($P = 0.142$ and $P = 0.207$) of all laboratory parameters of systemic inflammation (CRP, WBC count) on FCCs in the patient groups. Therefore, elevated FCCs in cirrhotic patients as opposed to controls may be caused by a regional (primary) intestinal inflammation which is not secondarily the result of systemic inflammatory reaction. Therefore, FCCs are even increased in cirrhotic patients without an extra-intestinal inflammation.

Gundling and his colleagues [3], also concluded that when comparing cirrhotic subjects with an extra intestinal infection, median FCCs were also significantly higher than without ($P = 0.001$).

However, this subgroup included also all patients with SBP which might explain the high median FCCs. Furthermore, the numerous microbiological and immunological alterations of the GI tract in cirrhotic patients including infiltrates of neutrophils might induce increased FCCs even in cirrhotic subjects without inflammation of GI tract.

Fagerhol et al., [28], explained the chronic inflammatory state present in cirrhotic patients which submit the theoretical possibility of causing elevated plasma levels of calprotectin as systemic endotoxemia is frequent and most likely caused by impaired endotoxin clearance in their compromised state. Endotoxemia immediately induces neutropenia followed by neutrophilia.

In the present study, we found that median FCCs were higher when ascites was present, however this difference was not significant ($p>0.05$). This is consistent with, Gundling et al., [3] who found that FCCs were higher when ascites was present. However, this difference was not significant. They added that in cirrhotic patients with SBP, median FCCs were higher than in patients without. Explorative data analysis of this association was significant [$P = 0.002$].
Therefore, as Gundlig and colleagues [3] concluded, FCCs may serve as a screening tool to identify cirrhotic patients with SBP. However, further studies are needed to investigate FCCs prospectively in cirrhotic patients with ascites and SBP before and after medical treatment in comparison to standard diagnostic procedures.

In the present study, there was a positive significant correlation between FCCs and serum ammonia of all patient groups ($p<0.05$) and there was a positive correlation between FCCs and the presence of hepatic encephalopathy (HE), however that correlation was not significant ($p>0.05$). These results were matched with Gundling et al., [3] who reported that the correlation of plasma venous ammonia and FCCs was significant($r = 0.391$; $P = 0.002$). Therefore, FCCs may serve as a screening tool to identify cirrhotic patients with HE. Furthermore, as Gundlig and colleagues concluded, assessment of FCCs may facilitate grading of HE-severity which may be sometimes subjective when using only clinical criteria.

Our study showed that there was no significant correlation between median FCCs and history of previous GIT bleeding ($p>0.05$). This is consistent with Gundling et al., who found that when comparing cirrhotic subjects with and without portal hypertension (presence of oesophageal varices, portal hypertensive gastropathy) median FCCs were higher when oesophageal varices were diagnosed but showing no significance ($P = 0.259$).

In conclusion, FCCs are elevated in patients with HCV-related chronic liver disease which were significantly elevated dependent on the severity of liver disease as assessed by Modified Child_Pugh's classification. So, FCCs can be used as a valid parameter in assessment of the severity of hepatitis C chronic liver disease. Further studies are needed to elicit the correlation between fecal calprotectin and different complications of chronic liver disease such as SBP and hepatic encephalopathy.

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