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CD14 and TIMP-1 as predictive biomarkers of extensive liver fibrosis in Egyptian HCV-patients

Eman M. Abd El Azeem*[1], Magda Kamal El Din Ezz[1], Dr. Amin M. Abdel Baki[2], Asmaa Abd ElFattah Deghedy[2]

[1]*Department Ofbiochemistry, Ain Shams University, Egypt*

[2]*National Hepatology and Tropical Medicine Research Institute, Egypt*

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Abstract

HCV infection is closely associated with liver fibrosis, a major risk factor related to liver cirrhosis and hepatocellular carcinoma. Therefore, the aim of this study was to analyze the association of serum s-CD14 and TIMP-1 level with the stages of fibrosis in hepatic tissue of HCV infected patients as an alternative non-invasive method to avoid using liver biopsy. This study included seventy HCV patients with liver fibrosis classified into four subgroups according to the degree of fibrosis: group 1 (with liver fibrosis F4), group 2 (with liver fibrosis F3), group 3 (with liver fibrosis F2) and group 4 (with liver fibrosis F1). Normal subjects were conserved as group 5 (control group). Direct biomarkers, serum s-CD14, TGF-β1and TIMP-1 levels were determined by the quantitative sandwich enzyme immunoassay (ELISA) technique. Serum ALT, AST, albumin, total bilirubin, prothrombin time and concentration, complete blood count were detected. Indirect biomarkers, ALT/AST Ratio (AAR) and Fib4 were also calculated. Serum sCD14, TGF-β1 and TIMP-1 levels showed a highly significant increase, also serum level of AFP increased significantly in all groups compared to normal control. This increment was parallel to the degree of fibrosis. The diagnostic accuracy of all direct blood markers rose with increasing stage of fibrosis, while the accuracy of indirect markers (AAR and Fib 4) increased in the early stage of fibrosis. Performance of sCD14 in our study was the best direct blood marker for diagnosis of extensive fibrosis (F3 and F4).In conclusion, sCD14 may be a more relevant biomarker of disease progression.

* **Corresponding Author:** Eman M. Abd El Azeem ✉ dreman1@live.com

Introduction

Hepatitis C virus (HCV) is one of the leading causes of chronic inflammatory liver disease (Zeremski *et al*., 2007). According to World Health Organization reports, about 170 million people currently suffer from HCV infection worldwide (Hoofnagle, 2002). Chronic HCV infection often results in the development of hepatic fibrosis, cirrhosis, and hepatocellular carcinoma (HCC) (Saito *et al*., 1990). It is estimated that the incidence of hepatocellular carcinoma (HCC) in Europe and United States will peak at 2020 at which there will be 78000 new HCC cases in Europe and 27000 in the United States(Flores and Marrero,2014). Cirrhosis is the most important risk factor for HCC regardless of the etiology and cirrhosis occurs in the background of 90% of cases of HCC (Flores and Marrero,2014). Egypt has the highest prevalence of hepatitis C virus (HCV) in the world, estimated nationally at 14.7% (Mahmoud *et al*., 2013) with genotype 4 being the most common (Ahram Online, 2014). A recently published Egypt Health Issues Survey (EHIS) in 2015 on a nationally representative sample showed that 10% of Egyptians between 15–59 years of age had been infected with HCV, while 7% are chronic active hepatitis C patients (Ministry of Health, Egypt, El-Zanaty and Associates, Egypt and ICF International, 2015). This warrants the need to investigate the current and the future burden of this disease.

Liver biopsy has been traditionally considered as gold standard to evaluate liver fibrosis. However, this procedure has several drawbacks such as pain and bleeding complications, and it can also lead to inaccurate staging due to sampling error and variability in the interpretation of biopsies. In the last decade, various non-invasive tests (NITs) have been introduced to stage liver fibrosis of which measurement of liver stiffness by transient elastography (TE) using FibroScan® is the most widely accepted method. This device-dependent technique is mainly limited to larger centers, and sometimes it cannot be applied in obese patients or persons with narrow intercostal spaces.

Blood serum markers singly or in combination are alternative non-invasive tests that allow a more widespread use and are cheaper (Patrick *et al*., 2015). Several diagnostic methods for determining liver fibrosis, such as the detection of blood biomarkers, have been used (Manning and Afdhal, 2008).CD14 over expression resulted in a hyper-responsiveness to low-dose lipopolysaccharide (LPS), an important step in the progression from simple steatosis to steatohepatitis, and was associated with liver inflammation and fibrosis (Imajo *et al*., 2012).

Hepatic fibrosis is the excessive accumulation of extracellular matrix proteins, including collagen I, III, and IV, as well as fibronectin, elastin, laminin, and proteoglycans (Inagaki and Okazaki, 2007). Collagens I and III are the two chief components of the fibrous tissue in the liver. Increased synthesis and decreased degradation of these proteins result in their accumulation. Both processes are mediated by activated hepatic stellate cells (HSC) (Inagaki and Okazaki, 2007), which are activated when the liver is involved in an inflammatory process. Stellate cells produce various components of the fibrous tissue, remodel liver architecture by expressing matrix metalloproteinases (eg, matrix metalloproteinase 2 (MMP2)), and inhibit degradation of collagens by expressing tissue inhibitors of matrix metalloproteinases (eg, TIMP1) (Benyon and Arthur,2001) by transforming growth factor β (TGF-β) and platelet-derived growth factor (PDGF).The most influential growth factors involved in HSC activation and collagen synthesis are transforming growth factor-β1 (TGF-β1) (Matsuzaki, 2009) and platelet-derived growth factor (PDGF), which are secreted by hepatocytes and platelets, respectively, during liver injury and inflammation (Hernandez-Gea and Friedman,2011). Levels of both TGF-β1 and all PDGF isoforms are up-regulated during HSC activation, correlating with the development of liver fibrosis and hepatocellular carcinoma (HCC) (Zavadil and Bottinger, 2005). Once activated, HSCs convert into highly proliferative, myofibroblast-like cells, which produce inflammatory and fibrogenic mediators (Hernandez-Gea and Friedman, 2011).

Kupffer cells produce transforming growth factor β (TGFβ), activating hepatic stellate cells (HSCs) to synthesize extracellular matrix proteins, resulting in fibrosis (Duffield *et al*., 2005).

The aim of this study is to analyze the association of serum s-CD14 and TIMP-1 level with the stages of fibrosis in hepatic tissue of HCV infected patients to be used as an alternative non-invasive method to avoid using liver biopsy.

Materials and methods

The present study was performed according to the guidelines of the Medical Ethical Committee of National Research Centre, Cairo, Egypt. HCV samples were collected from the National Hepatology and Tropical Medicine Research Institute. Written informed consent was obtained from all patients after full explanation of the procedure used.

Subjects

Seventy patients with clinical, biochemical and sonographical criteria of chronic liver disease (Child A), Non- obese (BMI <30), positive serology for HCV antibody and HCV vireamia, liver biopsy showing chronic hepatitis were enrolled in this study;(48 male, 37 female; range of ages, 40-56 years) Fifteen normal healthy person (38-45 years) served as the control group. Patients with chronic viral diseases other than HCV, non-alcoholic steato-hepatitis, autoimmune hepatitis, biliary disorders, and malignancies were excluded from the study.

Methods

Serum samples were obtained at the time of biopsy and stored frozen at -80°C for further determination of the selected parameters. All experiments were performed in duplicate.

Histopathology

Histological sections were blindly evaluated by two independent pathologists. Fibrosis staging was semi-quantitatively assessed according to the METAVIR system (Theise *et al*., 2007). Patients with liver fibrosis were classified into four subgroups according to the degree of fibrosis: group 1 (with liver fibrosis F4, 20 patients), group 2 (with liver fibrosis F3, 20 patients), group 3 (with liver fibrosis F2, 15 patients) and group 4 (with liver fibrosis F1, 15 patients). Normal health subjects were conserved as group 5 (Control group, 15 subjects).

Biochemical assays

Diagnosis was based on the presence of anti-HCV antibodies in the serum, which was detected by Real Time-polymerase chain reaction (RT-PCR) technique. Extraction of viral RNA by direct purification of viral RNA from plasma with the high pure viral RNA kit supplied by QIAGEN (Japan) (Yoshioka *et al*., 1992). Serum CD-14, TGF-β1 and TIMP-1 levels were determined by the quantitative sandwich enzyme immunoassay (ELISA) technique (Quantikine, R & D Systems, Inc, MN, USA). Serum alpha-fetoprotein (AFP)concentration was quantitatively determined using ELISA technique, kit was supplied by Fujirebio Diagnostics (Sweden).Serum AST and ALT levels were determined by the method of Gella *et al*. (1985), Serum albumin levels were detected using a human serum EIA kit (Cayman Chemical Co, Ann Arbor, MI, USA). Total bilirubin was measured using the method of Doumas *et al*. (1973).PT-INR, platelets and WBC's count were detected using Auto hematology analyzer.FIB-4 is a combination of four simple variables: AST, ALT, age, and platelet count. It is calculated with the following formula: FIB-4 index = [age (years) × AST (IU/L)]/ [platelet count (10^9/L) × ALT (IU/L)$^{1/2}$]. FIB-4 performed similarly to Fibro Test in the diagnosis of advanced fibrosis and cirrhosis in HCV patients (Vallet-Pichard *et al*., 2007).AST/ALT ratio (AAR) increase over 0.8 reflects a progressive liver functional impairment, while a ratio ≥ 1 is indicative of cirrhosis (Giannini *et al*., 2003).

Statistical analysis

Statistical analysis was performed using the Statistical Package for the Social Sciences (SPSS)version 23 for Windows. Numerical data were presented as the mean ± standard error. A comparison of variables was performed with ANOVA test.

Statistical significance was assumed at $p < 0.05$. Receiver operating characteristics (ROC) curve was performed to assess the ability of the serum biomarkers to differentiate between the stages of liver fibrosis. We determined the cut-off value for each parameter as the maximum value of the sum of the sensitivity and specificity.

Results

Baseline clinical fibrosis characteristics

The mean age of the study population (48 male and 37 female) was 46.9± 4.5 years.

HCV patients with chronic liver disease (Child A) were categorized according to the degree of fibrosis into four subgroups with criteria based on the underlying etiology of liver biopsy.

The biochemical parameters, PT-INR, PLT count, WBCs count, Hb were non-significantly changed in F1 group, while a significant increase was recorded in all other HCV groups. ALT, AST, T-bil. and albumin concentrations were significantly elevated for patients with HCV, the elevation was associated with the degree of fibrosis (Table 1).

Table 1. Clinical characteristics of all studied groups.

Groups / Parameters	Control	F1	F2	F3	F4
Male/female	3/2	3/1	1/1	4/5	2/1
Age (yr) Mean ± SE.	43.0±2.5	40.1±3	42.0±5.9	48.9±7.2	55.2±3.4
PT INR Mean ± SE. % change	1.0 ±0.04	±1.0860.03* ↑8.6	±1.0530.02* ↑5.3	±1.060.01* ↑6	±1.0550.01* ↑5.5
PLT(10^3/µl) Mean ± SE. % change	258.8 ± 13.6	290.06 ±20.5 ↑12.07	295.±316.4* ↑14.1	±212.9515.1* ↓17.7	185.05 ± 13.8* ↓38.9
Hb (g/l) Mean ± SE. % change	13.3460.2 ±	± 14.320.4 ↑7.29	.0±13.163 ↓1.39	±13.870.3 ↑3.92	±13.4750.3 ↑0.96
WBCS count(10^9/l) Mean ± SE. % change	6.993 ± 0.3	±5.9460.5 ↓14.972	±6.80.5 ↓2.75	±7.0250.4 ↑0.45	0.3 ±6.48 ↓7.33
TSH(µIU/ml) Mean ± SE. % change	0.661 ± 0.06	1.26±0.14* ↑91.07	±1.5070.26* ↑127.98	±1.5950.26* ↑141.3	±1.819 0.23** ↑175.18
ALT(U/l) Mean ± SE. % change	28.01±0.09	55 ±5.6** ↑96.35	58 ±6.3** ↑107.06	±44.17.1** ↑57.4	3±5.59.1** ↑26.7
AST(U/L) Mean ± SE. % change	16.1 ±0.8	50 ±4.7** ↑210.5	49.2 ±4.5** ↑205.6	±58.4517.1** ↑263.0	52.2 ±7.6** ↑224.2
T.bil (mg/dl) Mean ± SE. % change	0.40.04 ±	±0.6230.06 * ↑55.75	0.09±0.69* ↑73	0.05±0.78** ↑95.25	0.73 0.05 ±** ↑81.5
Alb(g/l) Mean ± SE. % change	4.7 ± 0.05	±4.040.1** ↓14.042	±4.10.15** ↓12.765	±4.0350.09** ↓14.148	±4.055 0.09** ↓13.723

PT-INR: Prothrombin time international normalized ratio; PLT: platelet count; Hb: hemoglobin; WBC: White blood cell; TSH: Thyroid stimulating hormone; ALT: alanine aminotransferase; AST: aspartate aminotransferase; T. bil: total bilirubin; Alb: albumin.

Data are expressed as mean ± standard errors (SE)

*: Significant from control ($p<0.05$), **: High significant from control ($p<0.001$)

Direct blood markers for non-invasive diagnosis of liver fibrosis

Serum sCD14 levels, reflective of LPS-induced monocyte activation, showed a non significant change in F1 patients and a slight significant increase (12.36%) in F2 patients compared to normal subjects (P<0.05). This increase was augmented in parallel with the degree of fibrosis to reach 136.18% in F3, while a dramatic level (223.4%) was present in cirrhotic patients (F4)(P<0.001).

Serum level of TIMP1 was non-significantly changed in the chronic HCV patients with low degree of fibrosis (F1and F2), conversely, a high significant increase was seen in extensive fibrosis (55.55 and 95.15% for F3 and F4 respectively) compared to controls (p < 0.001). TGF-B1 and AFP revealed a significant increase in all HCV patients (P<0.001) compared to normal controls. Serum levels increased gradually with increased stage of fibrosis to reach its maximum level in cirrhotic patients (165.53 and 2663%).

Table 2. Direct blood markers for non-invasive diagnosis of liver fibrosis.

Groups Parameters	Control	F1	F2	F3	F4
sCD-14(µg/L) mean ± SE. % change	±40.9377.7	±45.9985.3 ↑12.36	±58.722.3* ↑43.44	±96.6871.3** ↑136.18	±132.3893.5** ↑223.4
TIMP-1(µg/L) mean ± SE. % change	±50.0645.7	±59.8992.3 ↑19.64	±60.9712.2 ↑21.79	±77.8731.9** ↑55.55	±97.7142.5** ↑95.18
TGF-B1(ng/ml) mean ± SE. % change	±530.005 65.05	±949.63318.6** ↑79.17	1163.92 ±54.2** ↑119.60	.12234 ±61.5** ↑130.83	1407.35 ±38.4** ↑165.53
AFP(ng/ml) mean ± SE. % change	3±.750.05	.865 ±2.01** ↑130.6	±10.833.4** ↑188.8	±15.215.2** ↑305.6	±34.134.8** ↑2663

sCD-14: soluble CD-14; TIMP-1:tissue inhibitor of metalloproteinases-1; TGF-B1: transforming growth factor-β1; AFP: alpha-fetoprotein.

Data are expressed as mean ± standard errors (SE)

*: Significant from control (p<0.05), **: High significant from control (p<0.001).

Indirect blood markers for non-invasive diagnosis of liver fibrosis

Several scores have been proposed to attempt to overcome the need for liver biopsy in diagnosing and monitoring the fibrotic process in chronic liver diseases. Results in table 3 showed that Fib4 was high significantly increased in early fibrosis (86 and 84% for F1 and F2 respectively).

This increment was synergized in extensive fibrosis (304 and 424% for F3 and F4 respectively). Additionally, AAR revealed a high significant increase in F1 and F2 (57.89 and 47.36%, respectively), this increase was augmented parallel to fibrosis progression (131.57 and 161.4% in F3 and F4 respectively).

ROC curves

To evaluate the usefulness of the studied serum direct and indirect biomarkers for predicting extensive liver fibrosis at stage F3 and F4, the area under the ROC curve was analyzed. Results in Fig. 1 demonstrated that: sCD14(AUC of 0.75 and 0.93 with a cut off value 71.66 µg/l and 97.65µg/l, TIMP1 (AUC of 0.72 and 0.88 with cut off value: 70.28µg/l and 76.66µg/l), TGFß1 (AUC of 0.70 and 0.81 with cut off value:

1098.29ng/l and 1388.57ng/l), Fib4 (AUC of 0.52 and 0.55 with cut off value: 1.9and 2.1) and AAR (AUC of 0.60 and 0.49 with cut off value: 1.12 and 1.04)for F3 and F4respectively.

Table 3. Indirect blood markers for non-invasive diagnosis of liver fibrosis.

Groups / Parameters	Control	F1	F2	F3	F4
Fib 4 mean ± SE. % change	0.50± 0.4	0.930.2±** ↑86	0.920.1±** ↑84	.202 ±0.3** ↑304	2.62 ±0.2** ↑424
AAR mean ± SE. % change	.057±0.07	.090 ±0.08** ↑57.89	0.84 ±0.07** ↑47.36	.132 ±0.5** ↑131.57	.1 49 ±0.09** ↑161.40

Data are expressed as mean ± standard errors (SE)

*: Significant from control ($p<0.05$), **: High significant from control ($p<0.001$).

Discussion

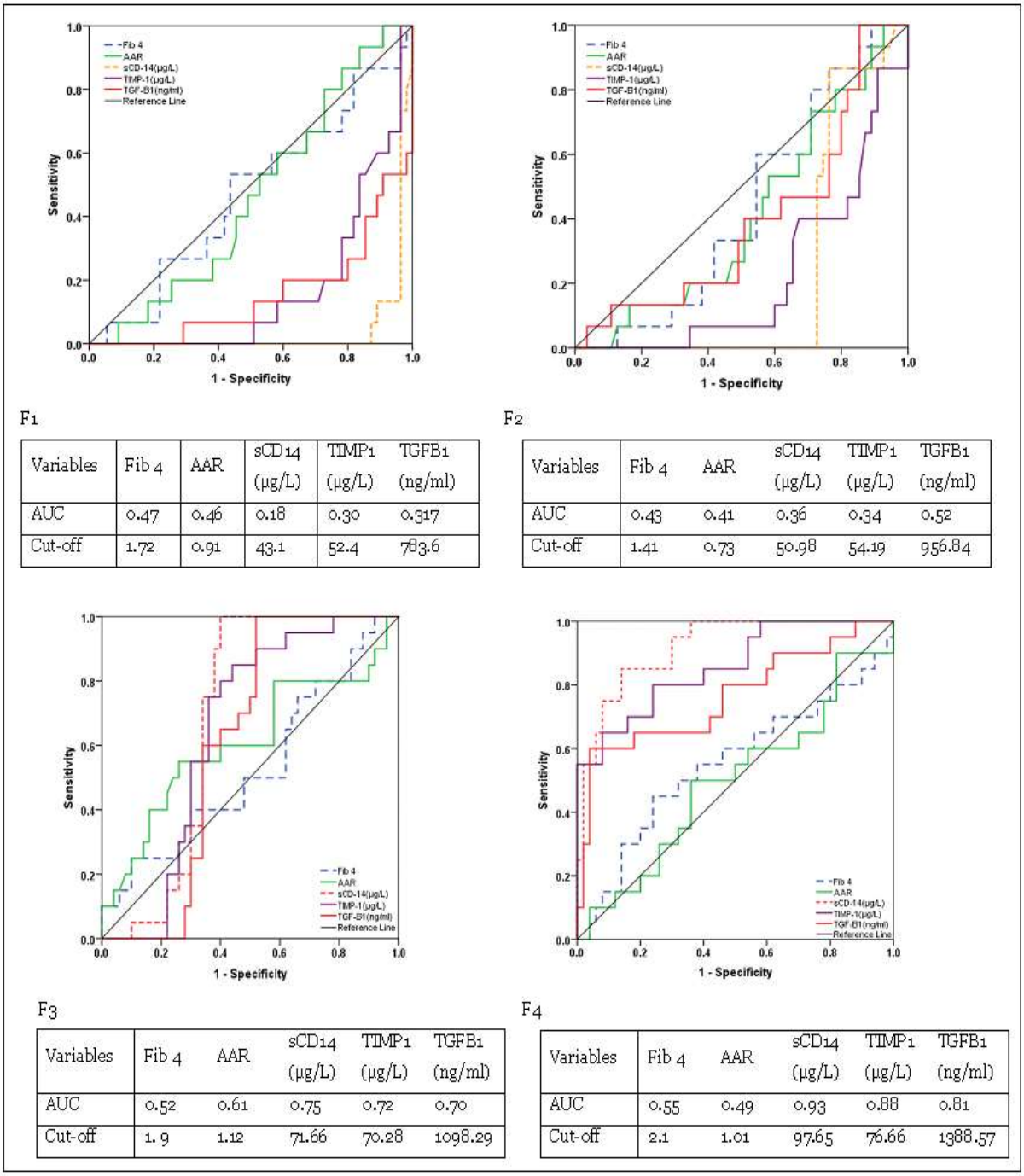
HCV infection is closely associated with liver fibrosis, a major risk factor related to fatal liver diseases, such as liver cirrhosis and HCC (Pereira *et al*., 2010). The extent of liver fibrosis is the most important predictor of liver-related outcomes, including development of portal hypertension, decompensation events, cancer and death (Irvine *et al*., 2016). Liver biopsy is the standard method for the evaluation of the grade of necro-inflammation and stage of fibrosis (European Association for Study of Liver,2015). However, given the size of the population at risk of chronic disease, the increasing demands on hepatology services and the implications of failing to identify advanced fibrosis, striving to maximize the diagnostic accuracy of non-invasive tests is an important goal (Katharine *et al*., 2016). The few studies to date do, however, indicate that serum biomarkers perform similarly to histology for predicting clinical outcomes (Irvine *et al*.,2016).

One contributor to chronic inflammation and fibrosis in chronic HCV infection is microbial translocation. Immunostimulatory microbial products from the intestine, such as lipopolysaccharide (LPS), translocate into the portal system where they are sensed and cleared by Kupffer cells. Therefore chronic liver disease may impair the clearance of translocating microbial products, LPS may also contribute to liver injury (Han,2002).LPS binding protein binds LPS, facilitating its binding to membrane CD14 (mCD14) on myeloid cells or to circulating soluble CD14 (sCD14)[10]. sCD14 transfers LPS to mCD14 and both sCD14 and mCD14 transfer LPS to the myeloid differentiation to the MD-2/TLR4 complex (Kitchens and Thompson,2005). The MD-2/TLR4/LPS complex activates NF- κb, inducing inflammatory cytokine production (Gioannini and Weiss, 2007).

In this study, the serum level ofsCD14was significantly increased in patients with liver fibrosis, the level increased gradually with increased stage of fibrosis to reach its highest level in liver cirrhosis (F4) ($p < 0.001$).

This result is in agreement with Netanya *et al*. (2011) who stated that levels of sCD14, a marker of LPS bio-reactivity, distinguished subjects with severe liver fibrosis and correlated with markers of hepatic inflammation and fibrosis. sCD14 may be a more relevant biomarker of disease progression as it reflects the host response to products of microbial translocation, rather than LPS itself. Higher sCD14 levels may reflect more cells responding to LPS or a genetic predisposition towards increased LPS responsiveness. Indeed, high sCD14 levels in the setting of HCV infection have been associated with a polymorphism in the promoter region of the CD14 gene (−159C/T) (Netanya *et al*., 2011).

TGF-ß1 is the most important cytokine involved in triggering liver fibrosis. It has been suggested that Kupffer cells and liver infiltrating lymphocytes in HCV infected hepatocytes are the major sources of TGF-ß1protein (Li *et al*., 2012).



F1

Variables	Fib 4	AAR	sCD14 (µg/L)	TIMP1 (µg/L)	TGFB1 (ng/ml)
AUC	0.47	0.46	0.18	0.30	0.317
Cut-off	1.72	0.91	43.1	52.4	783.6

F2

Variables	Fib 4	AAR	sCD14 (µg/L)	TIMP1 (µg/L)	TGFB1 (ng/ml)
AUC	0.43	0.41	0.36	0.34	0.52
Cut-off	1.41	0.73	50.98	54.19	956.84

F3

Variables	Fib 4	AAR	sCD14 (µg/L)	TIMP1 (µg/L)	TGFB1 (ng/ml)
AUC	0.52	0.61	0.75	0.72	0.70
Cut-off	1. 9	1.12	71.66	70.28	1098.29

F4

Variables	Fib 4	AAR	sCD14 (µg/L)	TIMP1 (µg/L)	TGFB1 (ng/ml)
AUC	0.55	0.49	0.93	0.88	0.81
Cut-off	2.1	1.01	97.65	76.66	1388.57

Fig. 1. Receiver operating characteristic curves of simple non-invasive tests evaluated for prediction of extensive fibrosis.

However, a number of reports have suggested that HCV infected hepatocytes could be the original source of TGF-ß1 protein (Lee *et al*., 2013). In this study, the serum level of TGF-β1 was found to be significantly higher in liver fibrosis patients than in the normal controls ($p < 0.001$). This increase was augmented in late stages of fibrosis ($p< 0.001$). These findings are supported by Neuman *et al.* (2002) and Iagoda *et al.*(2006), who stated that TGF-β1 level reflected the histologic stage, and activation of latent TGFβ1was reported to be the starting point of fibrogenesis. Luo *et al.* (2001) reported a significant elevation of TGF-β1 in liver cirrhosis, yet its correlation with the grade of activity was moderate. While, Egyptian studies (Nawar *et al*, 2011; Nassef *et al*, 2013) conducted on chronic liver disease (CLD) due to HCV and other etiologic factors reported that TGF-β1 was significantly increased in these patients compared to

controls, with a high significant positive correlation between TGF-β1 level and stage of liver fibrosis,this could be explained as a result of differences in geno typing of HCV in Egyptian patients.

Increased synthesis and decreased degradation of collagen proteins are mediated by HSC (Inagaki and Okazaki, 2007), which are activated when the liver is involved in an inflammatory process. Stellate cells produce various components of the fibrous tissue, inhibiting degradation of collagens by expressing tissue inhibitors of matrix metalloproteinases (eg. TIMP1) (Benyon and Arthur, 2001).Traditionally, monocytes are seen as prototypic pro-inflammatory cells, but recent evidence has shown that their functional capabilities extend beyond that of cytokine production alone. Marzena *et al.* (2012) explored the possibility that monocytes may contribute to tissue fibrosis through up-regulation of TIMP-1.

Our results showed that serum level of TIMP-1 increased significantly in all groups compared to normal control.

This increase was parallel to the degree of fibrosis suggesting the possibility that circulating monocytes expressing TIMP-1 migrate into hepatocytes to contribute to fibrogenesis and accumulation of collagen fibers. This result is in accordance with Marzena *et al.* (2012) who reported that circulating monocytes with profibrotic properties migrate to skin and internal organs to initiate and/or promote fibrosis.

AFP, a hepatoblast marker, is used clinically for HCC, although an association with fibrosis has previously been observed in chronic HBV (Liu *et al.*, 2014), and reduced AFP levels were associated with fibrosis regression following interferon therapy in patients with chronic HCV (Tachi *et al.*, 2016). Given the alternative, hepatic progenitor cell (HPC)-mediated pathway of liver repair is increasingly activated during chronic liver disease progression, and immature HPC are the source of AFP (Kakisaka *et al.*, 2015); AFP could represent a new serum biomarker of HPC activation.

Results of this study demonstrated that serum level of AFP increased significantly in all groups compared to normal control. This increase was synergized as the degree of fibrosis increased indicating that AFP may be one of the routine laboratory tests, having a relatively low cost, that foreshadow the rate of progression of fibrosis towards cirrhosis. Therefore, early diagnosis is crucial for improving the survival rate of patients.

This result is in accordance with Di Bisceglie *et al.* (2005) who stated that among patients with advanced chronic hepatitis C, serum AFP values are frequently elevated, even in the absence of HCC. Factors associated with raised AFP include severity of liver disease and decreased platelet count. Additionally, serum ALT levels, which reflect chronic liver disease activity, have an association with serum AFP levels (Richardson *et al.*, 2011). Conversely, other studies reported that although AFP was significantly increased in liver fibrosis, it is a non specific marker where it was found to be increased in other liver diseases (Farinati *et al,* 2006). Findings of this study revealed that Fib4 and AAR were significantly increased in extensive fibrosis with the AUCs 0.52 and 0.60 for F3, 0.55 and 0.49 for F4 respectively. These results are confirmed with previous results (Yung-Yu *et al.*, 2012).

In the present study, to evaluate the usefulness of the direct and indirect serum biomarkers for predicting extensive liver fibrosis, AUC was analyzed to establish the most sensitive one. Our findings indicate that, the diagnostic accuracy of all direct and indirect serum markers rose with increasing stage of fibrosis.

Performance of all direct serum markers were more specific. sCD14 in our study was the most sensitive and specific blood marker for diagnosis of extensive fibrosis, where AUC of sCD14 for F3= 0.75VS 0.72 and 0.7 for TIMP1and TGFß1respectively, while in F4AUC of sCD14 = 0.93VS 0.88 and 0.81 for TIMP1and TGFß1respectively.

Conclusion

In conclusion, due to the high incidence of HCV infection in Egyptian population, there was a great need to search for a more accurate non-invasive markers for screening the progression of fibrosis. Our data supported the use of direct blood markers rather than indirect markers which depend on two or more parameters increasing the chance of error. Serum sCD14 was the most sensitive and specific direct blood marker to evaluate extensive fibrosis.

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