



Research Article

The Combination of Noninvasive Tests and Fibroscan for the Assessment of Liver Fibrosis among Iraqis' Nonalcoholic Fatty Liver Disease Patients

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ABSTRACT

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Keywords: Fibroscan, non-alcoholic fatty liver disease, non-alcoholic steatohepatitis, liver stiffness measurement, transient elastography, controlled attenuation parameter



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Background: Most prevalent chronic liver disease in developed and developing nations is non-alcoholic fatty liver disease. From fatty liver, which often has benign, non-progressive clinical history, to non-alcoholic steatohepatitis, a more serious variant of fatty liver that can lead to cirrhosis and end-stage liver disease, non-alcoholic fatty liver disease encompasses broad spectrum of diseases. The gold standard for determining extent of hepatic fibrosis is still liver biopsy; however, number of noninvasive tests have been established to make diagnosis and assess effectiveness of treatment.

Objective: Aim of study was to assess effectiveness of the combination of fibroscan and noninvasive biochemical tests and scoring systems for assessing liver fibrosis.

Subjects and Methods: Cross-sectional prospective study conducted at outpatient clinic of Baghdad Gastroenterology and Hepatology Teaching Hospital from October 2018-March 2020. One hundred patients with fatty liver selected and subjected to specific questionnaires.

Results: One hundred patients with fatty liver studied; fifty-five (55%) males, forty-five (45%) females. Mean age was (45 ±12.24) years, females were significantly older. Mean stiffness score was 11.7(SD:5.29) KPa. Forty-six (46%) patients had advanced fibrosis. The Positive correlation between FibroScan examination results and the noninvasive scores results was significant, as well as there was significant positive correlation between age and stiffness score and significant negative correlation between platelet count and stiffness score.

Conclusions: This study showed that half patients showed advanced fibrosis, highlighting the need for early detection and management of fatty liver patients. Implementation of FibroScan with noninvasive fibrosis scoring tools has been shown to be helpful in this situation.

Introduction

The definition of non-alcoholic fatty liver disease (NAFLD) is the presence of hepatic steatosis in the absence of other causes of fat accumulation (1). NAFLD encompasses cirrhosis, non-alcoholic steatohepatitis (NASH), simple steatosis, and steatosis associated with different degrees of inflammation and fibrosis (2). The most

prevalent chronic liver disease (CLD) is NAFLD, and simple steatosis and NASH are present in 20%–30% and 5%–12% of the general population, respectively (3, 4, 5). A relatively low risk of developing cirrhosis was linked to simple steatosis (6–8). In contrast, cirrhosis develops in about 7% of NASH patients within 3 years (6–8).

Additionally, a number of prospective studies revealed that NASH was independently linked to an increase in mortality from both cardiovascular and liver disease-related causes (9, 10). Therefore, it is imperative to distinguish between patients with simple steatosis and those with NASH. The gold standard for determining if someone has fatty liver and distinguishing simple steatosis from NASH is still liver biopsy. But a biopsy is an intrusive procedure that has a small but real risk of consequences (11, 12). Given the limitations of liver biopsy, a number of noninvasive markers, such as imaging techniques and serological indices, have been investigated for the diagnosis of simple steatosis and NASH. As a result, researchers have looked into other non-invasive techniques for evaluating liver fibrosis in these patients (13).

The serological tests and the noninvasive scorings include the Aspartate aminotransferase /Alanine aminotransferase ratio (AST/ALT ratio), the aspartate aminotransferase/platelet ratio index (APRI), the Fibrosis 4 (FIB-4) score and the nonalcoholic fatty liver disease fibrosis score (NAFLD fibrosis score). The other popular and thoroughly tested noninvasive techniques for determining liver fibrosis is transient elastography (TE) (FibroScan), an ultrasound-based procedure (14). This technique has been used to diagnose liver stiffness and fibrosis in the past (15, 16). Thus, monitoring the stiffness of the liver over time can show how some liver disorders, including NASH, progress (17). The fundamental idea behind TE is that a homogenous tissue's elasticity, which is connected with the degree of fibrosis in the liver, determines how quickly a wave propagates through it.

What is Fibroscan:

Fibroscan is an ultrasound-based technique, is one of the most extensively used and well validated non-invasive methods for the assessment of liver fibrosis.

Diagnosis of liver stiffness and fibrosis by this method was reported previously (15,16). Thus, serial evaluation of liver stiffness can provide evidence about the progression of liver diseases like NASH. Transient elastography (TE or FibroScan® - FS) was first described in France in 2001, then in other parts of the world (18).

Transient elastography (TE); the basic principle of TE is that the propagation velocity of a wave through a homogenous tissue is proportional to its elasticity, which is correlated with the amount of fibrosis in the liver. Briefly, TE consists of an ultrasound transducer mounted on the axis of the vibrator, which produces vibration of a mild amplitude and low frequency (50 Hz), consequently inducing an elastic shear wave that propagates through the liver. Pulse-echo ultrasound follows the propagation of the shear wave and measures its velocity, which is related to liver tissue stiffness. It is reported that the velocity of elastic waves is faster in fibrotic liver than normal livers in previous study.

FibroScan is equipped with a probe including an ultrasonic transducer and a vibrator. A vibration of mild amplitude and low frequency is transmitted from the vibrator placed on the body surface toward the liver through the intercostal space. The vibration induces an elastic shear wave that propagates through the liver tissue. The pulse-echo ultrasound acquisitions follow the propagation of the shear wave and determine its velocity. The velocity is directly related to tissue stiffness; the harder the tissue, the faster the shear wave

propagates. Liver stiffness measurements (LSM) is calculated from velocity and expressed in kilo Pascale (kPa). LSM was performed after an overnight fast. Ten successful acquisitions were performed on each measurement and the median value was adopted as representative of LSM. To improve test reliability a minimum of 10 valid readings, with at least a 60% success rate and an interquartile range of $\leq 30\%$ of the median value. FibroScan® is principally used to estimate the degree of liver scarring present (ie. stage of liver disease). This is especially useful in the assessment of patients with chronic liver disease, including chronic hepatitis C, chronic hepatitis B, chronic alcohol abuse and fatty liver.

The diagnostic performance of transient elastography using CAP in detecting liver steatosis is very high with sensitivity and specificity of nearly 90% (19).

This reading may be used to:

- estimate the existing degree of liver damage.
- monitor disease progression or regression via serial measurements.
- guide prognosis and further management, including treatment.

Because the ultrasound-based technique requires an adequate visualization of the liver to obtain readings. In our experience, accurate readings are obtained in $<50\%$ of patients who have a body mass index (BMI) >35 kg/m² using the standard M probe. This problem is partially overcome through the development of different probes (such as the XL probe), that allows deeper penetration of vibration wave. There are no absolute contraindications for the test, although the presence of ascites prevents propagation of the vibration wave and therefore frequently results in failure to obtain readings. The manufacturer also advises against the use of this device in pregnancy and in patients with a pacemaker.

Prevalence and Distribution of NAFLD

The worldwide increase in the prevalence of well-established risk factors for NAFLD, such as diabetes, obesity, and age, has had an accompanying increase in the prevalence of NAFLD. The estimated global incidence of NAFLD is 47 cases per 1,000 population and is higher among males than females. The estimated global prevalence of NAFLD among adults is 32% and is higher among males (40%) compared to females (26%). The global prevalence of NAFLD has increased over time, from 26% in studies from 2005 or earlier to 38% in studies from 2016 or beyond. The prevalence of NAFLD varies substantially by world region, contributed by differing rates of obesity, and genetic and socioeconomic factors (20). In a recent meta-analysis, Latin America 44.37% (30.66%-59.00%), then Middle East and North Africa (MENA) (36.53%, 28.63%-45.22%), South Asia (33.83%, 22.91%-46.79%), South-East Asia (33.07%, 18.99%-51.03%), North America (31.20%, 25.86%-37.08%), East Asia (29.71%, 25.96%-33.76%), Asia Pacific 28.02% (24.69%-31.60%), Western Europe 25.10% (20.55%-30.28%)(21).

In the pooled analysis comprising 101028 individuals of most recent study in 2022 found that the prevalence of NAFLD in the overweight population was 69.99% (95% CI 65.40-74.21 I₂=99.10%), the prevalence of NAFL was 42.49% (32.55-53.08, I₂=96.40%), and the prevalence of NASH was 33.50% (28.38-39.04, I₂=95.60%). Similar prevalence estimates were reported in the obese population

for NAFLD (75.27% (95% CI 70.90–79.18); I₂=98.50%), NAFL (43.05% (32.78–53.97); I₂=96.30%) and NASH (33.67% (28.45–39.31); I₂=95.60%). The prevalence of NAFLD in the overweight population was the highest in the region of the Americas (75.34% (95% CI: 67.31–81.93); I₂=99.00%). Clinically significant fibrosis (stages F2–4) was present in 20.27% (95% CI 11.32–33.62; I₂=93.00%) of overweight individuals with NAFLD and in 21.60% (11.47–36.92; I₂=95.00%) of obese patients with NAFLD while 6.65% (4.35–10.01; I₂=58.00%) of overweight individuals with NAFLD and 6.85% (3.85–11.90; I₂=90.00%) of obese individuals with NAFLD had advanced fibrosis (stages F3–4) (22). This increasing prevalence of NAFLD in the US parallels the increase in prevalence of NAFLD-related risk factors, which include insulin resistance, obesity, hypertension, and dyslipidemia.

Pathophysiology of NAFLD

The pathogenesis of NAFLD is multifactorial. Genetic factors cooperate with metabolic and environmental factors to promote the accumulation of fat in hepatocytes. In the last decade of the 20th century, the most corroborated theory was the “two hit pathogenesis”.

It stated that insulin resistance leads to triglyceride deposition in the liver, thus steatosis, rendering it more susceptible to the action of second hits, such as oxidative stress, ATP depletion and endotoxins, finally leading to inflammation, fibrosis and cancer. Cell death and inflammation are key drivers of fibrosis in NASH and other forms of chronic liver disease. The primary pathophysiological mechanisms of insulin resistance induced by inflammatory mediators are probably the result of interference with insulin signaling (23). Insulin acts in all cells by binding to its specific receptor and activating a cascade of intracellular signaling. Upon insulin binding, the insulin receptor phosphorylates itself and several members of the insulin receptor substrate (IRS) family. IRS1 and IRS2 are the main mediators of insulin signaling in the liver, where they control insulin sensitivity (24). Nowadays, this theory has been replaced by the “multiple hit pathogenesis”. This states that multiple etiopathogenic factors act in a parallel or sequential and somehow synergic way on a genetically predisposed subject, to cause NAFLD and thus defining the spectrum of the disease phenotype. Particularly, some patients will develop NAFLD and consequently NASH, but others will directly present inflammation and fibrosis, probably because of the influence of genetic and epigenetic factors.

Study aims to assess the effectiveness of the combination of noninvasive testing and scoring systems with fibroscan for the assessment of fibrosis among Iraqi NAFLD patients seen in the outpatient medical clinic of the Gastroenterology and Hepatology teaching hospital in Baghdad.

Subjects and Methods

A cross-sectional observational prospective study was carried out to assess the effectiveness of noninvasive testing and scoring systems with FibroScan for the assessment of fibrosis among Iraqi NAFLD patients. One hundred patients who were diagnosed with NAFLD or NASH between October 2018 and March 2020 at the Gastroenterology and Hepatology Teaching facility (GI and Hep Hospital) in Baghdad, Central Iraq, based on ultrasound examination of abdominal and testing for serum liver enzyme that were done in our facility were included in this study. All patients, and occasionally their

family, completed pre-designed questionnaires for this study (see appendix). The study's inclusion and exclusion criteria were used to choose the participants. Participants had to be at least 18 years old, have undergone a Fibroscan examination within the study period, have an abdominal ultrasound that revealed evidence of a fatty liver, and have blood tests that revealed elevated liver enzyme levels. Patients whose data were insufficient were not included in the study. Also, Patients were disqualified if they had a history of alcohol-related liver illness, autoimmune hepatitis (AIH), hepatitis B or C, as demonstrated by an autoimmune positive antibody test or a positive response to corticosteroids. Patients with advanced liver illness, heart failure, and hepatic congestion were also disqualified, as were those on hepatotoxic drugs. Participants were also excluded if they had clinical or ultrasound indications of decompensated cirrhosis or were unable to perform FibroScan exams due to very high BMIs or other extenuating circumstances. Each patient's demographic information, including age, sex, and country of origin, was gathered. Within two weeks of the FibroScan examination, all findings from the laboratory tests that were assessed were acquired. These tests were conducted in our hospital. The normal range laboratory reference for serum alanine aminotransferase (ALT) in the GI and Hep.hospital is 12-78 U/L. Since it is difficult to determine a cutoff value for serum ALT and most studies still use the 30 U/L as upper cutoff value, we used this cutoff value for the upper limit of serum ALT in this study (25). The typical reference for platelet counts range is 150000-400000/microliter, while the usual reference range for serum aspartate aminotransferase (AST) was 15-37 U/L.

A SIEMENS Dimension X Pand Plus clinical chemistry system (Flex Reagent Cartridge) was used to detect liver enzymes. The alanine aminotransferase/ aspartate aminotransferase ratio (AST/ALT ratio), the aspartate aminotransferase/ platelet ratio (APRI score), nonalcoholic fatty liver disease fibrosis score (NAFLD fibrosis score) and the fibrosis-4 score (FIB-4 score) for each patient were measured using a computerized software application provided by (MD calculator and calculated by (QxMD) using the following formulas:

$$APRI = \frac{AST \text{ Level IU/L} / (AST(U/LN)IU/L}{Plateletes \text{ Counts}(10^9/L)} \times 100$$

$$FIB - 4 = \frac{Age(y) \times AST (U/L)}{Plateletes \text{ Count } 10^9/L \times \sqrt{AST(U/L)}}$$

NAFLD fibrosis score = -1.675 + 0.037 3 ages (years) + 0.094 3 BMI (kg/m²) + 1.13 3 IFG/diabetes (yes = 1, no=0) +0.99 3 AST/ALT ratio - 0.013 3 platelet (3109/L) - 0.66 3 albumin (g/dL)

The instrument utilized in the investigation was an Echosens Fibroscan530 COMPACT (2018) (30 Place d'Italie 75013 Paris, France) The patient **prepared** for the test should be fasting for 4-6 hours, the patient lay on his /her back with the right arm under the head, the abdomen and the lower chest exposed and the fibroscan probe is positioned in an intercostal space near the right lobe of the liver usually in the ninth – eleventh intercostal space, anterior axillary line, For a test to be judged effective, ten valid FibroScan readings are required (according to the manufacturer's recommendations and results from earlier research) (26,27). For every patient that was a part of the trial, a success rate of > 90% was attained.

Based on numerous prior research and references offered by the manufacturer, the following fibrosis stages were identified: F0 = 1-6 KPa, F1 = 6.1-7 KPa, F2 = 7-9 KPa, F3 = 9.1-10.3 KPa and F4 >= 10.4 KPa (28, 29).

Statistical Analysis:

Analyses were performed using IBM's Statistical Package for the Social Sciences (SPSS) version 26. A descriptive data and a student's t-test was used to compare the differences in the mean ages and serum ALT values between the males and the females. Age, platelet count, and serum ALT levels were compared with a correlation analysis to determine the degree of hepatic fibrosis. The association between the results of the FibroScan and the NAFLD fibrosis score, AST/ALT ratios, APRI scores, and FIB-4 scores was examined using a straightforward dot graph. To compare the AST/ALT ratio, the APRI and FIB-4 scores, and the NAFLD fibrosis score between patients with advanced fibrosis (higher than F2) and patients with mild-moderate fibrosis of (F2 or less) a student's t-test were used.

Results

One hundred patients were included finally, out of those one hundred patients included in this study; fifty-five (55%) are males and forty-five (45%) are women. The mean age of the studied patients was 45 years (SD: twelve range 20-75 years). Female patients were significantly older than male patients (47 years (SD:11) versus 43 years (SD:13), respectively, P= 0.01). Of the hundred patients 42 (42%) had hypertension and 39(39%) had diabetes.

This study showed that the mean waist circumference for the studied patients was (99.5±3.5 cm); it also had been founded that the waist circumferences were significantly higher for the female in comparison to the males in this study (mean=102±4 for the females and 97±3for the males, p value = 0.03).

The mean BMI for the studied patients was (32.41±4.05) and it was also significantly higher for the female comparing to the males (mean=33.64±4.33 and 31.19±3.78 respectively, p value = 0.04). The mean serum cholesterol was (253±30 mg/dl) and it was significantly higher in females compared to the males (265 mg/dl (SD=32)versus 241 mg/dl(SD=28),respectively, P=0.02),also had been founded that the mean serum triglyceride value was(232 ±21 mg/dl) and these values were significantly higher in female compared with the male patients (247 mg/dl (SD=25) versus 217 mg/dl (SD=17), respectively, P=0.03), this study had been shown that the mean serum high density lipoprotein(S.HDL) was(42±9 mg/dl) and this value was founded to be low in females compared to the males patients in this study(39 mg/dl SD=8) versus 45 mg/dl (SD=10), respectively, P=0.02 (table1).

Most of the patients showed either no fibrosis (36%) or a severe amount of fibrosis (30%), and the mean stiffness score was 11.7(SD:5.29) KPa. According to the stage of fibrosis, the distribution of the patients under study is shown in (Table 2).

Male patients showed higher stiffness scores compared to the female and the difference was statistically significant (12.93 (SD:6.02) versus 10.47 (SD:4.57) KPa, respectively, P=0.02) (Table3).

This study also showed that mean ALT for male patients was significantly higher than mean serum ALT for the female patients (78.81IU/ml (SD:52.31) versus 51.88(SD:33.06), respectively, P=0.03) (Table3), (also see table 4 for mean serum ALT, AST and platelets count).

Correlation analysis revealed a significant positive correlation between patients age and stiffness score (r=0.49 for Pearson correlation, p=0.01) (Table5). On the other hand, there was a strong negative correlation between platelets count and fibrosis score (r=-0.339 for Pearson correlation, P= 0.001) (Table5). In addition, a significant positive correlation was found between the serum ALT

level and stiffness score (r=0.45 for Pearson correlation, P=0.01) (Table6).

Negative correlation had been found between Serum ALT and the patients' age (r= -0.38 for Pearson correlation, P=0.03) (Figure1).

There were significant differences in the stiffness scores for NAFLD fibrosis score, FIB4 score, APRI score, and AST/ALT ratio score calculations (the most significant were for NAFLD fibrosis score and FIB4 calculations) between patients with advanced fibrosis (more than F2 score on FibroScan) at 46(46%) and those with mild to moderate fibrosis (F2 score and less on FibroScan) at 54(54%) (Table 7).

The stiffness Scores on the Fibroscan examination were correlated significantly with the results obtained from NAFLD fibrosis score, FIB4 score, AST/ALT score and APRI score; with the most significant positive correlation was found for the fibrosis results obtained by NAFLD fibrosis score and FIB4 score(figures2,3,4-5).

Table1: study characteristics of the patients

Age (years)		45±12
Sex (N. %)	Male	55(55%)
	Female	45(45%)
BMI	Male	31.19±3.78
	Female	33.64±4.33
32.41±4.05	p value=0.04	(Kg/m2)
Waist Circumference(cm)	Male	97±3
	Female	102±4
		p value=0.03
Hypertension		42%
Diabetes mellitus		39%
Fibrosis score (Fibroscan)(KPa)		11.7±5.29
Steatosis score		(Fibroscan)(dB/m)
285.64±62.87		
S.cholesterol (mg/dl)	Male	241±28
	Female	265±32
	Male	217±17
S.Triglyceride (mg/dl)	Female	247±25
	Male	45±10
	Female	39±8
S.HDL (mg/dl)	Male	42±9
	Female	39±8
		P = 0.02

Table2: Distribution of patients according to the stage of liver fibrosis

Stage of fibrosis (Kpa) %	NO
F0 (0-5.9)	36(36%)
F1 (6-6.9)	14(14%)
F2 (7-9)	4(4%)
F3 (9.1-10.3)	16(16%)
F4 >=10.4	30(30%)
Total	100(100%)

Table3: the difference in stiffness score and serum ALT level between males and females

	Sex	Mean±SD	P. Value
Fibrosis Score(KPa)	Male	12.93±6.02	0.02
	Female	10.47±4.57	
Serum ALT level(U/L)	Male	78.81±52.31	0.003
	Female	51.88±33.06	

Table4: Mean Platelet Counts, Serum Alanine Transaminase levels (ALT), and Aspartate Amino transferase levels (AST)

LabTest	Mean±SD	Normal
reference range		
Platelets k/μL	235.20± 79.92	150-400
SerumALT U/L	65.34± 42.68	12-40
SerumAST U/L	45.13± 25.53	15-37

Table5: The distribution of the patients according to the platelets level

Platelets level(k/μL)	No. (%)
50000-100000	17(17%)
100000-150000	33(33%)
>= 150000	50(50%)

Table6: The correlation of patients Age, Serum ALT level and Platelets count with Stiffness score

	r value	P value
Age(years)	0.49	0.01
Serum ALT(U/L)	0.45	0.01
Platelets Count	- 0.339	0.001

Table7: Differences in Stiffness Scores, NAFLD fibrosis score, AST/ALT Ratio, APRI score and FIB-4 Score between Patients with Mild to Moderate Fibrosis and Those with Advanced Fibrosis

	> F2 (N.=46)	F2 or less (N.=54)	P value
Stiffness Score	18.7±2.1	4.8±1.5	0.001
NAFLD fibrosis score	0.89±0.17	- 1.66± - 0.12	0.001
AST/ALT ratio score	2.3±0.2	0.61±0.11	0.04
APRI score	1.36±0.26	0.31±0.21	0.02
FIB-4	2.96±0.35	0.87±0.63	0.01

Values are expressed as Mean±SD

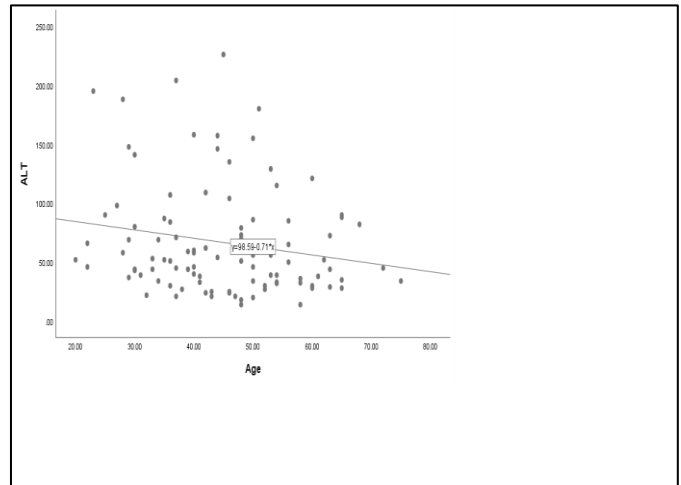


Figure1: Correlation between patients' age and serum ALT levels (r= - 0.38, P=0.03)

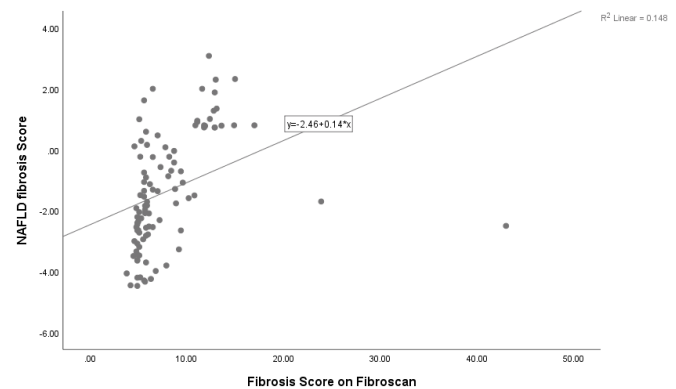


Figure 2. Correlation between Fibrosis Scores on Fibroscan and NAFLD fibrosis Score (r = 0.38, P=0.001)

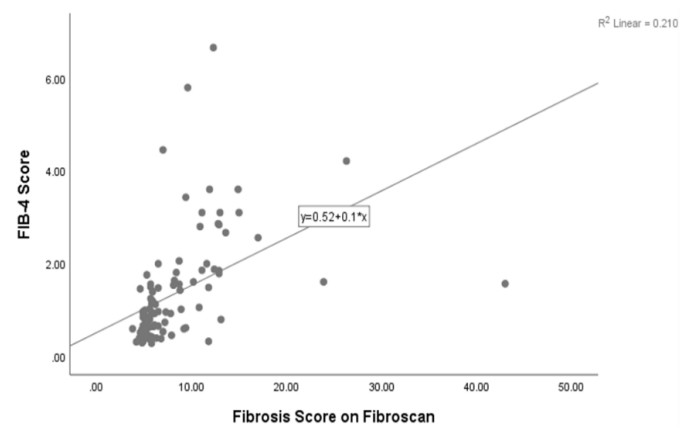


Figure3.Correlation between Fibrosis Scores on Fibroscan and FIB-4 scores (r=0.46, P=0.001)

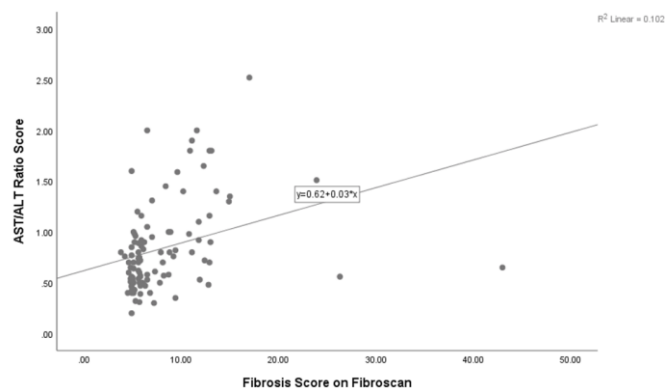


Figure4.Correlation between Fibrosis Scores on Fibroscan and AST/ALT Ratio Scores($r=0.32$, $P=0.01$)

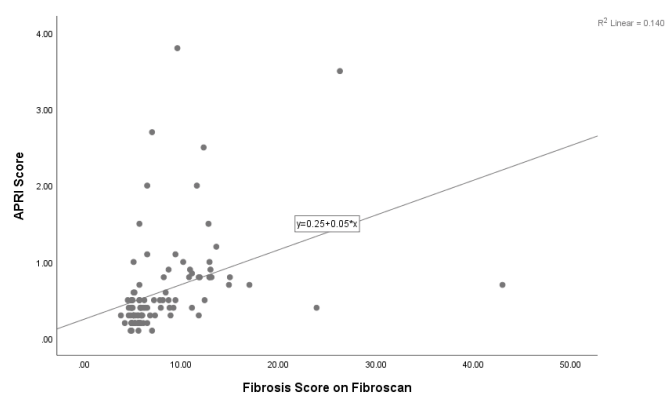


Figure5.Correlation between Fibrosis Scores on Fibroscan and APRI Scores($r=0.37$, $P=0.001$)

Discussion

All across the world, NAFLD is becoming more common. With catastrophic morbidities, it is a significant contributor to liver cirrhosis and hepatocellular cancer, the percentage of chronic liver disease is as the following : NAFLD (59%), followed by HBV (29%), HCV (9%), and autoimmune hepatitis (2%), other liver diseases, including primary biliary cholangitis, primary sclerosing cholangitis, alpha-1-antitrypsin deficiency, Wilson's disease, and autoimmune hepatitis, account for 1% of cases(30).

According to the findings of the Fibroscan tests, this study revealed that a significant portion of the NAFLD patients had advanced fibrosis stages. The FibroScan results and the outcomes of the noninvasive fibrosis scoring methods (NAFLD, FIB-4, AST/ALT, and APRI scores) showed a significant association, which validated these findings.

According to this study, advanced fibrosis was seen at a younger age in male compared to female patients, which was consistent with findings from Ezhilarasan D. et al, which showed that women developing chronic liver disease at older age(mean age 60 years) than men (mean age 45 years) and this is explained by the protective effect of female sex hormones on the progression of hepatic fibrosis , and this is also the cause that male patients typically have more severe

liver disease compared to female patients do for the majority of liver related etiologies (31).

In this study ; for Iraqi patients with metabolic syndrome and typeII diabetes mellitus, the high rate of advanced NAFLD is alarming. Alarming findings for metabolic syndrome and type II diabetes mellitus among Iraqi populations were also found in an Iraqi study conducted by Asmaa A. et al which found that 14% had type 2 diabetes and the prevalence of overweight/obesity was 66.9% (32).

Given that severe fibrosis in patients with liver illness is related with thrombocytopenia, this study also demonstrated that there was a strong negative association between the stiffness score and platelet count, which is consistent with similar studies conducted in other nations, like that conducted by Desai S. , Subramanian A. which found that patients with stage 4 fibrosis have mean platelets count of 80000/microliter(33).

Additionally, the results of this study supported previous researches conducted by Chalasani N. et al , Liu Y. et al and Fan J. et al which demonstrated that liver fibrosis severity increased with age (42% had F3-F4 fibrosis at age 65 year) and that age was related with a more advanced level of fibrosis, and this can be explained by the fact that insulin resistance, which is known to be a primary cause of the NAFLD, is a major component of the metabolic syndrome, which is often observed in elderly people. Aging, which is accompanied by abdominal obesity and excessive visceral fat, causes insulin resistance and an increased secretion of proinflammatory cytokines and, subsequently, results in the metabolic syndromes and type 2 diabetes, activation of p300-C/EBP-dependent neutral fat synthesis , telomere shortening , a decreased autophagy, increased M1 macrophage inflammatory responses , and activation of nuclear factor- κ B pathways (2, 34, and 35).

The study showed that ultrasound examination of abdominal alone is insufficient in the evaluation of advanced fibrosis, but that the implementation of the serum markers to ultrasonic abdominal examination can further categorize patients into those with mild and those with advanced NAFLD, which was consistent with the findings of Razavizade M. et al done for related purposes and found combining serum markers with ultrasonic finding increase the detection rate of advance fibrosis to 85-90 % (36).

In this study, all patients had baseline abdominal ultrasound examinations that showed evidence of fatty liver. This finding may further support the use of straightforward, noninvasive fibrosis scoring in conjunction with abdominal ultrasound in the diagnosis of NAFLD when more sophisticated techniques like MRI elastography or transient elastography(fibroscan) are unavailable or when performing liver biopsy is not necessary.

In contrast to numerous general population studies that revealed that level of serum ALT increases with advancing age(16), the findings from our investigation demonstrated an inverse connection between serum ALT level and patient age, this can be explained by disease activity and the onset of NASH in younger people, which can cause cirrhosis with advancing of the age and return of ALT to normal levels in those older people ,other related researches done by Chalasani N. et al , Reddy Y. et al, Dai W. et al and Lee CH. et al, that showed comparable results of decreasing serum ALT (mean 10 U/ml) with advancing age (mean age 60 year) which further supported the findings in our study (2, 37, 38, and 39).

This study also demonstrated that the FibroScan examination and noninvasive scoring tools (FIB-4, NAFLD fibrosis score, APRI, and AST/ALT score) could distinguish between mild to moderate and

advanced fibrosis; the best outcomes were obtained from the FibroScan, FIB-4, and NAFLD fibrosis scores, while the least favorable outcomes were with the AST/ALT Ratio score; these outcomes were comparable to those of other studies conducted elsewhere in the world, like that one carried out in the United Kingdom by Masuzaki R. et al and found similar results (40).

This study discovered that the majority of the patients had advanced levels of steatosis, which was previously verified in other locally-conducted studies by Asmaa A. et al on the high incidence of metabolic syndrome and type II diabetes mellitus in the Iraqi community, that founded that 33.9% are obese with BMI > 30 Kg/m² and the prevalence of overweight/obesity was 66.9% (32).

Conclusion

This research demonstrates that evaluating liver fibrosis in NAFLD patients using FibroScan in conjunction with noninvasive scoring tools (NAFLD fibrosis score, AST/ALT ratio score, APRI score, and FIB-4 score) is an effective method. In patients without obvious causes, this can prevent the need for an unnecessary liver biopsy.

Because of the high prevalence of diabetes and metabolic syndrome in the Iraqi population, the high number of patients with advanced degree of fibrosis in this study is concerning.

Limitations

1. The number of included patients might be small in view of the national growing prevalence of the NAFLD risk factors among Iraqi population, but this could be compensated for by the strict inclusion criteria.
2. Liver biopsy which is the gold standard for the diagnosis of NAFLD/NASH was not used in this study, due to the complication of this procedure and the absence of clear indications among the studied patients, it should not be used for every patient with NAFLD.
3. The single center study and the limitation of the availabilities of the Fibroscan machines.

Ethics approval and consent to participate statement.

The study was approved by the Ethics Committee of the College of Medicine, University of Basrah (NO. 030408-030-2022 in 2020), verbal and written informed consent had been taken from the patients enrolled in the study and in order to support privacy and confidentiality, I concealed the unique identifying information of people in the data gathering.

Consent to publication

Written informed consent had been taken from participants.

Availability of data and material

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Competing Interests

Authors have declared that no competing interests exist.

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Authors' contributions

Concept and design: Muntadher Abdulkareem Abdullah. Acquisition, analysis or interpretation of data: all authors. Drafting of the manuscript: Muntadher Abdulkareem Abdullah. Critical revision of the manuscript for important intellectual content: all authors. Statistical analysis: Muntadher Abdulkareem Abdullah and Kamal Breesam Lafta. Supervision: Raghad Jawad.

Data availability

Data are available upon reasonable request.

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