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Ultrasound Elastography Role versus Liver Biopsy in Diagnosing and Staging of Hepatic Fibrosis in Chronic Liver Disease Patients

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ABSTRACT

Background: Ultrasound elastography is a novel and promising noninvasive method based on sonography for assessing liver fibrosis in persons with chronic viral hepatitis.

Objective: The aim of the current work was to assess the effectiveness of ultrasound elastography as a non-invasive alternative method of liver biopsy for the diagnosis and staging of hepatic fibrosis in people with chronic liver disease.

Patients and methods: This study included a total of 50 patients with chronic HCV who had been identified by using PCR testing for HCV RNA and seropositivity for HCV antibodies, attending at Theodor Bilharz Research Institute (TBRI). Prior to starting treatment, patients were referred for evaluation and underwent abdominal ultrasonography, ultrasound-guided liver biopsies, and real-time elastography (RTE) to quantify the stiffness of the liver.

Results: liver fibrosis index (LFI) and stage of liver fibrosis identified by very sensitive liver biopsy showed a significantly positive connection (ranging of sensitivity from 86.36 % to 100%) and high specificity ranging from (78.57 % to 89.36%). High sensitivity and specificity was observed specially in high grades of fibrosis ≥F3 and F4. For determining the extent of liver fibrosis in CHC patients, RTE shown a high level of performance utilizing LFI, and it proved to be notably helpful for the identification of early cirrhosis and advanced hepatitis, both of which are issues in clinical practice. Additionally, it contrasted well with the reported pooled TE performance.

Conclusion: It could be concluded that the future of elastography imaging is optimistic, and RTE appears to be a promising method since it can yield a good diagnostic performance to predict advanced fibrosis in CHC, and to detect hepatic fibrosis staging.

Keywords: Ultrasound Elastography, Liver Biopsy, Hepatic Fibrosis.

INTRODUCTION

Inflammation, necrosis, which causes hepatocyte lysis, and a reparative tissue response all contribute to the complicated dynamic process that leads to liver fibrosis. The result of acute and chronic liver illnesses, such a condition includes non-alcoholic steatohepatitis, autoimmune hepatitis, chronic hepatitis B and C, and alcoholic liver disease [(1)].

The chronic liver illnesses cirrhosis, fibrosis, and inflammation are all brought on by HCV, which is one of the main causes of these conditions globally. With between 15% and 25% of residents in rural regions infected, Egypt has the highest adult hepatitis C virus (HCV) infection rate in the world. HCV has also been related to an increased risk of morbidity and mortality in cases of hepatocellular carcinoma [(2)].

The best method for determining the extent of liver fibrosis is still liver biopsy. Important information on necrosis, inflammatory grading, and iron accumulation is provided by liver biopsy, in addition to fibrosis and its structure. On the other side, it is an intrusive technique that causes discomfort for the patient, and there can occasionally be catastrophic problems. Additionally, high intra- and interobserver variability as well as sampling errors restrict the accuracy of liver biopsy [(2)].

With the aid of adapted software and standard ultrasound equipment, a new imaging technique called ultrasound elastography makes it possible to image and estimate the distribution of tissue elasticity within biological tissues non-invasively [(3)].

Given that measures of liver stiffness are closely associated to fibrosis METAVIR stages, transient elastography is one of the techniques that may be utilized to non-invasively quantify mean tissue stiffness. Additionally, transient elastography offers the advantages of reproducibility, not requiring a highly skilled operator, and having a low risk of unfavorable results [(4)].

A relatively new technique for measuring tissue elasticity is real-time tissue elastography. It employs a B-mode US scanner, a hybrid of the conventional US scanner and elastography. The body is gently pressed or released as the echo signals are recorded in real-time using a US Probe. This method estimates the relative hardness of tissue using standard B-mode photos and images of tissue elasticity and displays that information as real-time color graphics [(4)].

The examination of liver fibrosis in people with chronic viral hepatitis using ultrasound elastography is a unique and promising noninvasive sonography-based technique that is unaffected by the test sites or the observer. When combined with foundational laboratory data, ultrasound elastography can further improve the separation of different fibrosis stages, which is essential for treating patients with viral hepatitis [(5)].

This study's goal was to assess the efficacy of ultrasound elastography as a non-invasive method against liver biopsy for detecting and staging hepatic fibrosis in people with chronic liver disease.

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PATIENTS AND METHODS

This study included a total of 50 patients with chronic HCV who had been identified by using PCR testing for HCV RNA and seropositivity for HCV antibodies, attending at Theodor Bilharz Research Institute (TBRI).

For evaluation before treatment, patients were enlisted from the outpatient clinics of the National Hepatology and Tropical Research Institute (NHTMRI) and TBRI.

Inclusion Criteria: Patients with chronic liver disease >18 years and <70 years indicated for liver biopsy: (a) To evaluate grade of hepatitis activity and stage of fibrosis. (b) To make a decision regarding therapy.

Exclusion Criteria: (a) Presence of ascites or liver tumor, (b) Pregnancy, (c) History of liver transplant, (d) Portal vein thrombosis, and (e) Contraindications for liver biopsy (thrombocytopenia, international normalized ratio (INR) greater than 1.6, ascites.

All participants were subjected to:

1. **History taking:** Full history was collected as occupation as well as family history.
2. **Clinical examination:** General examinations and vital signs.
3. **Laboratory investigations:** Complete blood count (CBC), prothrombin time, prothrombin concentration, & INR, AST, ALT, bilirubin and albumin. Viral markers including HBs Ag, HCV Ab and HCV quantitively by PCR.
4. **Imaging procedures:**

Ultrasonography of abdomen: To evaluate the size of the spleen, the existence of splenic cirrhosis and ascites, and the portal vein.

Liver stiffness measurement: Utilizing real-time tissue elastography and an EUP-L52 Linear probe (3-7 MHz; Hitachi Medical) for ultrasonography. It was completed on the same day of the liver biopsy (RTE).

Protocol of RTE scanning:

Right branch of the portal vein should be visualized before manually angling the probe away from the heart and away from compression until the portal vein is hidden. Ensure that the displacement caused by each heartbeat is in the axial direction. In addition to the small compression measured with the linear probe, vascular pulsation provides the compression physiologically.

The liver parenchyma can be seen clearly in B-mode images, verify that the liver is not shifting by the heartbeat in a lateral direction. The elastography procedure requires patients to hold their breath. While the strain graph displays the periodic pattern as seen below, save movies.

Position of the ROI:

Set the ROI to only include the parenchyma of the liver and exclude big arteries. To prevent repeated reflections, the top of the ROI should be placed more than 1 cm inside the surface of the liver. (The artefact may be brought on by multiple reflections)

- Ensure that the large vessels are excluded (to avoid the artefact of anechoic area).
- Be sure to leave out the shadow of the ribs (to avoid the artefact of anechoic area).
- If the ROI bottom is partially rejected, stay away from the area (Lack of penetration).

How to select analysis frames:

- Choose the strain graph's negative peak frame.
- Avoid using a frame with artefacts.
- When the color of the elastography is not balanced in the lateral direction, stay away from the frame. (Liver may be moved laterally when either the right or left side of the elastography is colored blue or red. Please confirm once more that the direction of displacement caused by the heartbeat is axial).

The RTE equipment simultaneously shows the standard B-mode image and the RTE picture, which features a colored area around the region of interest (ROI). The biopsy spot was chosen to be close to a region of the tissue that was free of big arteries. The measurement was made using a rectangle 10 mm below the liver's surface that was 25 mm long and 25 mm wide. The ROI's color was graded on a scale from blue to red. Three static photographs were taken after we recorded the RTE images as moving digital images during ten to fifteen seconds.

The liver biopsy was carried out under ultrasound guidance using a true-cut semiautomatic needle (16 G). A liver sample that was formalin-fixed and paraffin-embedded.

Histopathological examinations were done according to METAVIR scoring **[(6)].**

Ethical consent:

An approval of the study was obtained from Zagazig University Academic and Ethical Committee. Every patient signed an informed written consent for acceptance of participation in the study. This work has been carried out in accordance with The Code of Ethics of the World Medical Association (Declaration of Helsinki) for studies involving humans.

Statistical analysis

Version 15.0 of the IBM SPSS application was utilized. In order to determine the significance of the acquired results, a 5-percent threshold was used. Chi square test ($\chi 2$) to calculate difference between two or more groups of qualitative variables. Quantitative data were expressed as mean ± SD (Standard deviation). It was a Chi-square test, for categorical variables, chi-square correction for more than 20% of cells with anticipated count less than 5 was required, Student t-test: to calculate the quantities of data of normal distribution and to compare between two studied groups. P value < 0.05 was considered significant.

RESULTS

Demographic characteristics show that mean age was 41.36 ±11.405, mean BMI was 26.968±4.4716kg/m2, mean ALT and AST were 71.06±15.63,53.34±11.42U/L respectively.

Table (1): Demographic data among the studied group

	No.	Mean±SD
Age (years)	50	41.36 ±11.405
BMI (kg/m^2)	50	26.968±4.4716
ALT (U/L)	50	71.06±15.63
AST (U/L)	50	53.34±11.42
PLT (x10^3/ul)	50	215.70±51.83
APRI	50	0.723±0.12
LFI	50	2.424±0.41
Area	50	21.388±5.21

The mean LFI in patients with F1, F2, F3, and F4 were 2.037, 2.573, 3.8 and 3.043 respectively. And the mean % Area was 15.561, 24.153, 39.646, and 32.447 respectively.

Table (2): Real Time Elastography results

Fibrosis		LFI	Area
F1	Mean	2.037	15.561
	No.	28	28
	S.D	0.357	5.260
	Minimum	1.4	2.9
	Maximum	2.8	26.9
	Median	2.095	15.450
F2	Mean	2.573	24.153
	No.	14	14
	S.D	0.389	5.663
	Minimum	2.1	16.0
	Maximum	3.7	38.8
	Median	2.500	23.440
F3	Mean	3.800	39.646
	No	5	5
	S.D	0.908	8.892
	Minimum	2.9	24.5
	Maximum	5.4	58.0
	Median	3.250	34.520
F4	Mean	3.043	32.447
	No	3	3
	S.D	0.179	2.600
	Minimum	2.9	29.5
	Maximum	3.3	34.4
	Median	2.940	33.420
Total	Mean	2.424	21.388
	No	50	50
	S.D	0.419	5.143
	Minimum	1.4	2.9
	Maximum	5.4	58.0
	Median	2.310	19.810

LFI: Liver Frailty Index

Correlation between the LFI, area, APRI and activity with histological fibrosis stages:

Spearman's correlation analysis revealed: LFI and liver fibrosis stage had a strong positive connection (r=0.766, p0.001). Strongly positive correlation (r= 0.770, p0.001) between liver fibrosis stage and area. Strong positive connection (r=0.691, p0.001) between activity and liver fibrosis stage. APRI and liver fibrosis stage had a somewhat good connection (r=0.490, p0.001).

Table (3): P-value and spearman's correlation coefficient between histologic fibrosis stage and LFI, Area, Activity and APRI.

Correlation Coefficient		p value
LFI	0.766	0.000
Area	0.770	0.000
Activity	0.691	0.000
APRI	0.490	0.000

ROC curve analysis:

Relationship between (LFI) and liver histology:

F1 vs F2-4, F1-2 versus F3-4, and F1-3 versus F4 all had overall AUCs of 0.916, 0.979, and 0.904, respectively.

Sensitivity, specificity, and accuracy were measured when the cut off value of LFI was adjusted to 2.31 for F1 against F2-4, and they were 86.36, 78.57, and 82%, respectively.

The PPV and NPV were, respectively, 76 and 88 percent. Cutoff was set to 2.82, which resulted in 100% sensitivity, 95.24% specificity, and 96% accuracy for F1-2 vs. F3-4. PPV and NPV were 80 and 100%, respectively.

The sensitivity, specificity, and accuracy were 100, 89.36, and 90% for F1-3 against F4 when the cutoff value was 2.925. The PPV and NPV were respectively 37.5 and 100%.

Table 4: Cutoff values, Sensitivity, Specificity, and Area Under the Receiver Operating Characteristic Curves (AUCs) for Real-Time Strain Elastography (LFI and AREA) and their combinations for Each Metavir Fibrosis Stage (F).

	LFI			%Area		
	≥F2	**≥F3**	**=F4**	**≥F2**	**≥F3**	**=F4**
Cut-off	2.31	2.82	2.925	20.445	23.19	29.25
AUC	0.916	0.979	0.904	0.924	0.958	0.908
Sensitivity (%)	86.36%	100%	100%	86.36 %	100%	100%
Specificity (%)	78.57%	95.24%	89.36%	85.71 %	78.57%	89.36%
Accuracy (%)	82	96	90	86	82	90
PPV	76	80	37.5	82.61	47.06	37.5
NPV	88	100	100	88.89	100	100

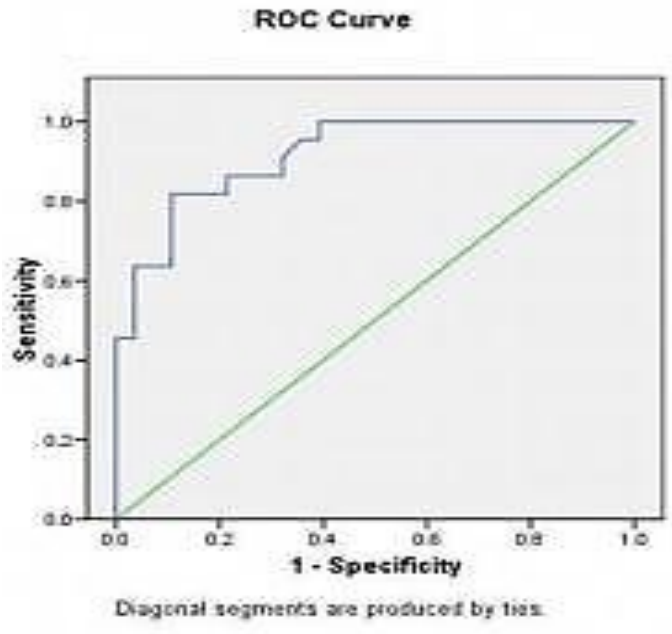


Fig. (1): Liver fibrosis index ROC curve; F ≥2 (F1vs.F2–F4).

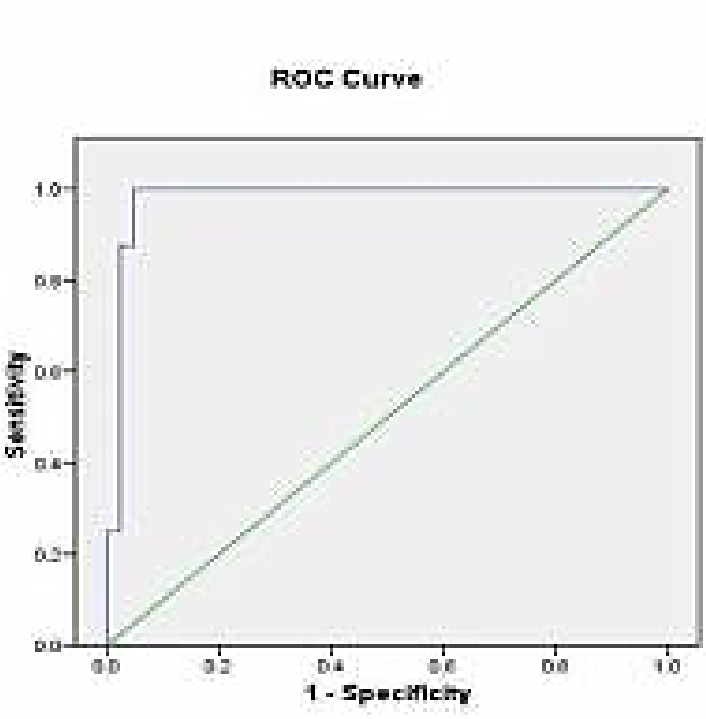


Fig. (2): Liver fibrosis index ROC curve; F ≥3 (F1–F2vs. F3–F4).

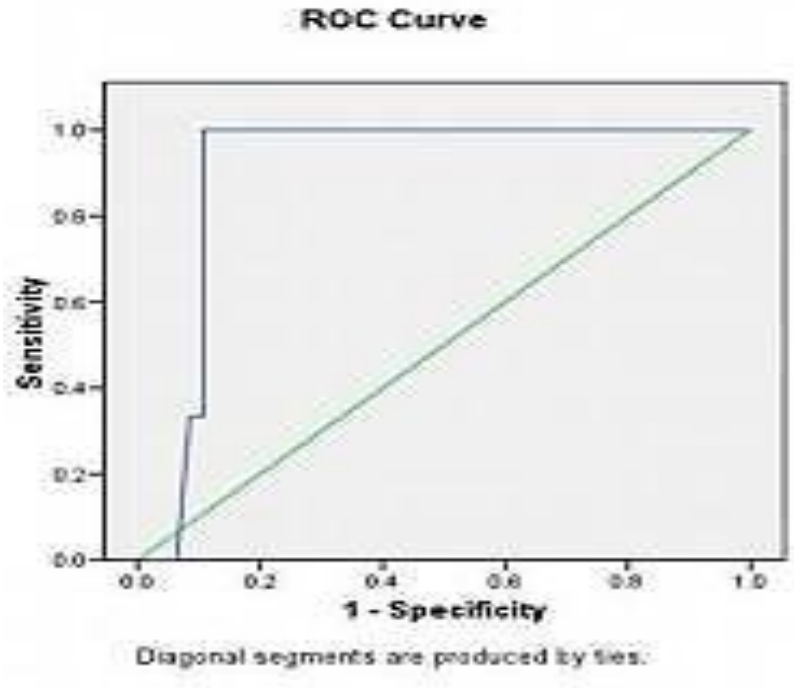


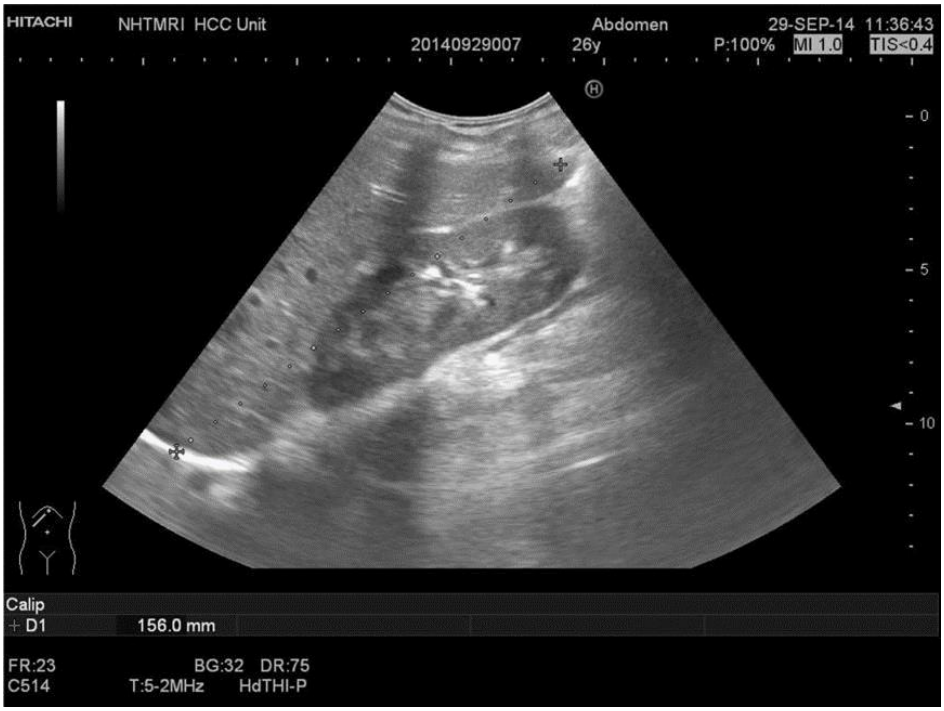
Fig. (3): Liver fibrosis index ROC curve; F= 4(F1–F3vs.F4).

Comparison between RTE (LFI and AREA) with APRI and activity by p value:

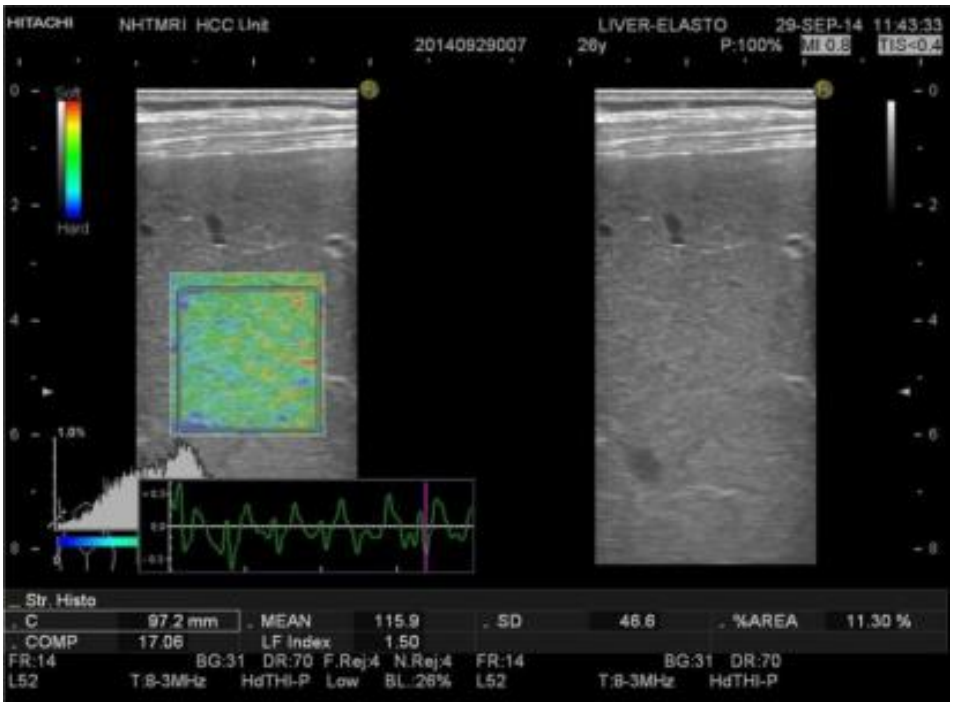
- **Activity:** There was high statically significance between **F1 vs. F2-4** and **F1- 2 vs. F3-4** (p =0.000) and moderate statically significance between **F1-3 vs. F4** (p =0.003).
- **APRI:** There was high statically significance between **F1 vs. F2-4** (p=0.001) and mild statically significance between **F1-2 vs. F3-4**(p =0.021) and in **F1-3 vs. F4** (p =0.021)**.**
- **Liver fibrosis index (LFI):** There was high statically significance between **F1 vs. F2-4** and **F1- 2 vs. F3-4** (p=0.000) and mild statically significance between **F1-3 vs. F4** (p=0.020).
- **Area of fibrosis (AREA):** There was high statically significance between **F1 vs. F2-4** and **F1- 2 vs. F3-4** (p=0.000) and mild statically significance between **F1-3 vs. F4** (p=0.019)**.**

Table (5): Comparing ACTIVITY, APRI, LFI, and AREA according to P value in F2, F3, and F4 (F1 against F2-4, F1-2 compared F3-4) (F1-3 versus F4).

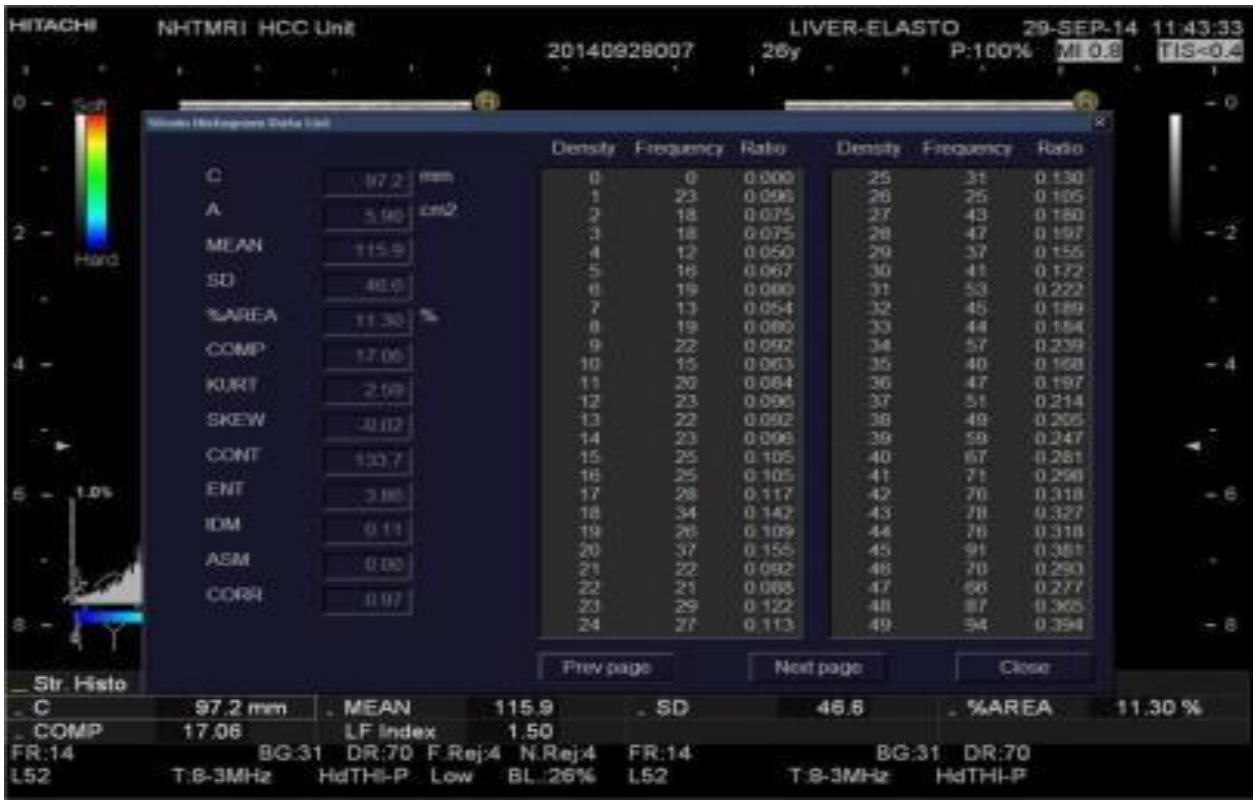
	p- value		
	F ≥2 (F1 vs. F2-4)	**F≥3 (F1-2 vs. F3-4)**	**F=4 (F1-3 vs. F4)**
Activity	0.000	0.000	0.003
APRI	0.001	0.021	0.021
LFI	0.000	0.000	0.020
AREA	0.000	0.000	0.019



(A)

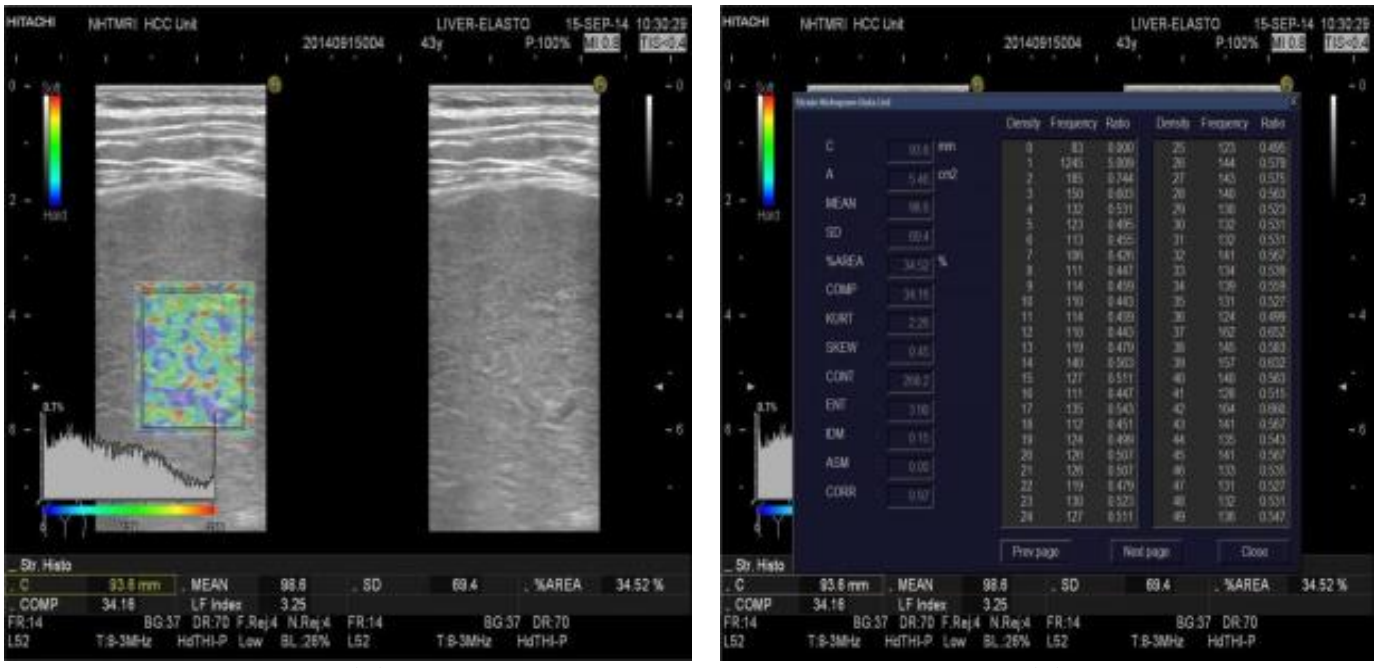


(B)



(C)

Fig. (5): 26 years old, HCV +ve, female patient, BMI=20. Laboratory investigations: ALT =25, AST=95, Platelet=224000/cmm,Alphfeto protein =0.7 and APRI= 0.279. US findings: Average hepatic size, homogenous echotexture, smooth hepatic surface, no periportal thickening. Real Time Elastography: LF index: 1.50, Area:11.30%. Histopatholgy: Mild chronic hepatitis, A1 F1.



(A) (B)

Fig. (6): 43 years old, HCV +ve, male patient, BMI=34.Laboratory investigations: ALT = 22, AST= 27, Platelet= 114000/cmm, Alpha feto protein =21 and APRI=0.592. US findings: average size, with coarse echotexture, and irregular hepatic surface. Real Time Elastography: LF index: LF index: 3.25, Area:34.52%. Histopathology: Severe chronic hepatitis, A3 F3.

DISCUSSION

Serum biomarkers and imaging modalities are two noninvasive techniques that are increasingly employed in clinical practice to assess liver fibrosis. Further evaluation using LB with its acknowledged limitations may not be required if noninvasive approaches clearly identify liver fibrosis. RTE is a rapidly developing method that is currently being adopted by numerous institutions to carry out clinical studies [7].

In this work, we applied a recently developed multiple regression analysis approach to analyze RTE pictures in order to get the Liver Fibrosis Index (LFI), which is automatically generated by the Hitachi elastography module from image attributes of RTE images. **Tatsumi *et al.*[8], Fujimoto *et al.* [9], Tomeno *et al.* [10], Yada *et al.* [11], Tamaki *et al.* [12] and Kim *et al.* [13]** employed 9 elastography features to calculate the LFI using the same method of calculation as that used in this study.

In study of **Morikawa *et al.* study [14]**, correlation coefficients were 0.487, 0.458, 0.377, and 0.451, respectively. While **Fujimoto *et al.* [9]** stated that the RTE image and fibrosis stage had a strong correlation. MEAN, STD, AREA, and COMP had correlation coefficients of r=-0.604, 0.593, 0.592, and 0.578, respectively. Multiple regression analysis was carried out using these 4 factors to construct the regression equation, which significantly fit the data. This equation was used to compute the RTE fibrosis value, which had a strong connection with the stage of fibrosis (r=0.729).

The elasticity index is the second quantitative technique (EI). In contrast to LFI, **Colombo and colleagues [15]** and **Wang and colleagues [16]** investigated another equation. In contrast to LFI, they produced 11 parameters derived using RTE. After that, elasticity index (EI) was generated utilizing a multiple regression equation's 11 parameters.

In certain investigations, the hepatic parenchyma and intrahepatic venous small veins were concurrently chosen as two ROIs, and the strain of each distribution was assessed without the use of distinct methods to determine the degree of hepatic fibrosis. The elastic ratio was then calculated by dividing the value of the tiny intrahepatic veins by the value of the hepatic parenchyma (ER) [17].

In our study a high correlation between LFI and the stage of liver fibrosis (r= 0.766, p<0.001) was found, which was higher than that reported by **Fujimoto *et al.* [9]** (r = 0.68) and **Kim *et al.* [13]** (r = 0.39). In the previously published studies regarding the performance of LFI [8-12], LFI in CHC patients increased stepwise as fibrosis severity increased, and there were significant differences in all fibrosis stages (p <0.001).

Fujimoto *et al.* [9] revealed that there is a substantial variation in LF Index values between each stage of liver fibrosis, with the LF Index showing both a high correlation (r = 0.68) with the F stage and a significant difference in LF Index values (p 0.001) between them.

Morikawa *et al.* [14] showed that for cirrhotic individuals, the mean, standard deviation (SD), area, and complexity had corresponding areas under the receiver operating characteristic curves (AUC) of 0.91, 0.91, 0.93, and 0. 95.

The overall AUC for F1 versus F2-4, F1-2 versus F3–4, and F1–3 versus F4 were **0.916, 0.979** and **0.904** respectively. Estimated cut off values of LFI were **2.31** for F≥2, **2.82** for F≥3 and **2.925** for F=4.

In a study by **Hong *et al.* [18]**, LFI showed reasonable accuracy for identifying F2 (AUC 0.79, sensitivity 78 percent, specificity 63 percent), F4 (AUC 0.94, sensitivity 91 percent, and specificity 68 percent), and overall was outstanding in detecting F3 (AUC 0.94, sensitivity 91 percent, and specificity 68 percent) (AUC 0.85, sensitivity 77 percent, specificity 78 percent). Also **Hong *et al.* [18]** reported that F2 versus F0-1 and F4 versus F0-3 could not be accurately differentiated using LFI. LFI's performance was comparable to ours in terms of diagnosing F3 (AUC 0.979, sensitivity 100%, specificity 95.24%), excluding advanced fibrosis with a 100% NPV, and F2 (AUC 0.916, sensitivity 86.36%, specificity 78.57%), in F=4 (AUC 0.904, sensitivity 100 percent, specificity 89.36 percent).

Chung *et al.* [19] examined the effectiveness of combining platelet count with LSM modalities (TE, ARFI, and RTE) to improve the diagnosis accuracy for liver disease with various etiologies. The forecasting accuracy of cirrhosis using the LSM/platelet count cut off ratios was no better than that of the LSM data alone. However, in terms of forecasting severe fibrosis, the ratios fared better than LSM data by itself (grade F2). Using the three evaluated methodologies, there were substantial associations between the fibrosis stage and the liver stiffness measurement (LSM): They came to the conclusion that TE and ARFI may be more accurate predictors of severe liver fibrosis than RTE. The diagnostic performance of TE in CHB patient scans can be enhanced by combining test findings with fibrosis data.

Ferraioli *et al.* [20] In their research evaluating TE, RTE, and APRI stated that transient elastography showed remarkable diagnostic performance in the diagnosis of severe fibrosis and cirrhosis. Real-time elastography cannot yet replace transient elastography in the evaluation of liver fibrosis.

In this study, we compared the outcomes with LFI measures to further assess the performance of two basic, straightforward procedures (biochemical and imaging) for the diagnosis of liver fibrosis. We evaluated APRI's effectiveness as a predictor of major, advanced fibrosis or cirrhosis. It is a straightforward, readily computed approach. In our investigation, there was a significant association between APRI and the stage of histologic liver fibrosis (r =0.490, p = 0.000).

Lee and colleagues [21] reported that agreement between high Forns index value as well as a high LSM had increased diagnostic specificity (from 87 to 98 percent in the validation and from 99 to 100 percent, in the training cohorts).

CONCLUSION

It could be concluded that the future of elastography imaging is optimistic, and RTE appears to be a promising method since it can yield a good diagnostic performance to predict advanced fibrosis in CHC. This will help to improve the precision and dependability of hepatic fibrosis staging. Future research on larger patient cohorts will be required to validate RTE, determine the best cutoff values, and investigate if RTE has the potential to replace TE.

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REFERENCES

1. **Aydın M, Akçalı K (2018):** Liver fibrosis. The Turkish Journal of Gastroenterology, 29(1): 14–21.
2. **Roehlen N, Crouchet E, Baumert T (2020):** Liver Fibrosis: Mechanistic Concepts and Therapeutic Perspectives. Cells, 9(4): 875-79.
3. **Loomba R, Adams L (2020):** Advances in non-invasive assessment of hepatic fibrosis. Gut, 69(7): 1343–1352.
4. **Gennisson J, Deffieux T, Fink M *et al.* (2013):** Ultrasound elastography: principles and techniques. Diagnostic and Interventional Imaging, 94(5): 487–495.
5. **Fang C, Sidhu P (2020):** Ultrasound-based liver elastography: current results and future perspectives. Abdominal Radiology (New York), 45(11): 3463–3472.
6. **Brunt E (2000):** Grading and staging the histopathological lesions of chronic hepatitis: The knodell histologic activity index and beyond. Hepatol., 13 (1): 241-246.
7. **Frulio N, Trillaud H (2013):** Ultrasound elastography in liver. Diagnostic and Interventional Imaging, 94: 515-534.
8. **Tatsumi C, Kudo M, Ueshima K *et al.* (2010):** Noninvasive evaluation of hepatic fibrosis for type C chronic hepatitis. Intervirology, 53(1):76-81.
9. **Fujimoto K, Kato M, Kudo M *et al.* (2013):** Novel image analysis method using ultrasound elastography for non-invasive evaluation of hepatic fibrosis in patients with chronic hepatitis C. Oncology, 84(1): 3–12.
10. **Tomeno W, Yoneda M, Imajo K *et al.* (2013):** Evaluation of the Liver Fibrosis Index calculated by using real-time tissue elastography for the non-invasive assessment of liver fibrosis in chronic liver diseases. Hepatol Res., 43(7):735-42.
11. **Yada N, Kudo M, Morikawa H *et al.* (2013):** Assessment of liver fibrosis with real-time tissue elastography in chronic viral hepatitis. Oncology, 84(1):13-20.
12. **Tamaki N, Kurosaki M, Matsuda S *et al.* (2014):** Prospective comparison of real-time tissue elastography and serum fibrosis markers for the estimation of liver fibrosis in chronic hepatitis C patients. Hepatology Research, 44(7): 720–727.
13. **Kim Y, Chung K, Kwon J *et al.* (2014):** Diagnostic Usefulness of Real-Time Elastography for Liver Fibrosis in Chronic Viral Hepatitis B and C. Gastroenterol Res Pract., 14: 1-7.
14. **Morikawa H, Fukuda K, Kobayashi S *et al.* (2011):** Real-time tissue elastography as a tool for the noninvasive assessment of liver stiffness in patients with chronic hepatitis C. J Gastroenterol., 46: 350-358.
15. **Colombo S, Buonocore M, Del Poggio A *et al.* (2012)**: Head-to-head comparison of transient elastography (TE): real-time tissue elastography (RTE): and acoustic radiation force impulse (ARFI) imaging in the diagnosis of liver fibrosis. J Gastroenterol., 47: 461–469.
16. **Wang J, Guo L, Shi X *et al.* (2012):** Real time elastography with a novel quantitative technology for assessment of liver fibrosis in chronic hepatitis B. Eur J Radiol., 81(1):31-37.
17. **Ochi H, Hirooka M, Koizumi Y *et al.* (2012)**: Real-time tissue elastography for evaluation of hepatic fibrosis and portal hypertension in nonalcoholic fatty liver diseases. Hepatology, 56: 1271–1278.
18. **Hong H, Li J, Jin Y *et al.* (2014):** Performance of Real-Time Elastography for the Staging of Hepatic Fibrosis: A Meta- Analysis. PloS One, 9(12):1-15.
19. **Chung J, Ahn H, Kim S *et al.* (2013):** The usefulness of transient elastography, acoustic-radiation-force impulse elastography, and real-time elastography for the evaluation of liver fibrosis. Clin Mol Hepatol., 19:156-164.
20. **Ferraioli G, Tinelli C, Malfitano A *et al.* (2012):** Performance of real time strain elastography, transient elastography, and aspartate to platelet ratio index in the assessment of fibrosis in chronic hepatitis C. Am J Roentgenol., 199(1):19-25.
21. **Lee S, Kim D (2014):** Non-invasive diagnosis of hepatitis B virus-related cirrhosis. World J Gastroenterol., 14; 20(2): 445-459.