

**ROLE OF GAMMA GLUTAMMYL TRANSPEPTIDASE TO PLATELET RATIO(GPR) IN PREDICTING  
ADVANCED LIVER FIBROSIS IN PATIENTS WITH CHRONIC HEPATITIS C INFECTION**

**Abstract:**

The liver biopsy is the gold standard for the diagnosis of liver fibrosis, because of its invasiveness, high cost and lack of repeatability its use is limited. A new parameter widely used these days for evaluating the grade of hepatic fibrosis is the gamma-glutamyl transpeptidase (GGT)-to-Platelet ratio (GPR) and has shown great benefit in this regard. The aim of our study was to evaluate the role of GPR as a noninvasive predictor of liver fibrosis in patients with chronic hepatitis C in the study population.

**Methods:**

All patients with chronic hepatitis C and compensated liver disease were included in the study after informed consent. Patient's baseline characteristics were recorded. Patient's baseline Complete blood count (CBC) and Liver function tests were also recorded. Patients then underwent shear wave elastography (SWE) to stratify the degree of fibrosis. These indices were used to calculate Gamma glutamyl transpeptidase(GGT) /platelet ratio. Results were presented as means  $\pm$  SD for quantitative data or as numbers with percentages for qualitative data. Continuous variables were analyzed using the Student's t-test; while categorical variables were analyzed using the Chi-square test. A p value of  $<0.05$  was considered statistically significant.

**Results:**

A total of 91 patients were included in the study. Out of 91 patients, 56(61.5%) were males. At baseline, 57(62.6%) patients had  $\geq$ F3 fibrosis(advanced fibrosis or cirrhosis).Mean GPR was  $1.5 \pm 2.1$ .Area under ROC(Receiver Operating Curve) was obtained for GPR in predicting advanced liver fibrosis( $\geq$ F3) was 0.8 ( $p \leq 0.001$ ). Higher GPR values were significantly associated with prediction of advanced liver fibrosis( $\geq$ F3) in patients with chronic hepatitis C with a sensitivity was of 94.74%, specificity of 62%, positive predictive value of 80.69%, negative predictive value was of 87.50% and diagnostic accuracy of 82.42%.

**Conclusion:**

The GPR found to be significantly associated with liver fibrosis in HCV patients presented in our clinic. However, further studies are needed to validate the role of GPR in predicting liver fibrosis.

**Keywords:** HCV, shear wave elastography, Liver fibrosis, Platelets, Gamma Glutamyl Transpeptidase, GPR

## **Introduction:**

Globally, hepatitis C is the most common causes of cirrhosis and hepatocellular carcinoma associated with increased morbidity and mortality<sup>1</sup>. In order to halt the disease progression, early diagnosis and prompt treatment are the corner stones of treatment of hepatitis C resulting in decreased number of patients with advanced fibrosis, cirrhosis and hepatocellular carcinoma (HCC) <sup>1-3</sup>. The gold standard method to assess the degree of fibrosis is liver biopsy. The use of liver biopsy is restricted due to its invasiveness, cost, complications and contraindications<sup>3-6</sup>. Currently, clinical practice needs non-invasive cost effective assessment tools that can easily predict liver inflammation and advanced liver fibrosis <sup>7</sup>.

Recently, Shear Wave Elastography (SWE) has emerged as a reliable non-invasive tool in staging liver fibrosis which accurately assesses the liver fibrosis and has eliminated the need of liver biopsy. However, there are certain confounding factors that can alter the yield of SWE and have limited its utility including severe inflammation, increased bilirubin, cholestasis, presence of ascites, cost, and lack of skilled operators.<sup>8-9</sup> Lately, certain serum biomarkers and bedside scores have been proposed that are useful in predicting advanced liver fibrosis and are of particular importance as they do not require skilled staff and costly equipment and these scores have the advantage of comprising only two or three laboratory investigations.<sup>10</sup>

Among these non-invasive tools, AST to Platelet Ratio Index (APRI) and Fibrosis-4 (FIB-4) are the 2 widely used non-invasive models <sup>11, 12</sup>. Several novel non-invasive tools to assess liver fibrosis

include neutrophil-to-lymphocyte ratio (NLR), red cell distribution width (RDW)-to-platelet ratio (RPR) and AST/alanine aminotransferase (ALT) ratio (AAR)<sup>13</sup>. Recently, the gamma-glutamyl transpeptidase (GGT) to platelet ratio (GPR) had emerged as a novel marker in the estimation of liver fibrosis. Studies in patients with HBV infection have revealed the superior accuracy of GPR as compared to the classical biomarkers APRI and FIB-4 in this regard in west African cohorts while French studies found GPR to be non-superior to APRI and FIB-4<sup>14</sup>. GPR also failed to show superior accuracy in Brazilian and Chinese cohort<sup>15, 16</sup>.

The aim of our study was to evaluate the diagnostic accuracy of GPR in predicting advanced liver fibrosis in patients with chronic HCV infection in our population. There is little work done in this regard as GPR has not been studied in chronic HCV infection for the prediction of advanced liver fibrosis specifically in our population.

#### **Material and methods:**

This was a cross-sectional prospective study which was carried out in the department of hepato-gastroenterology, Sindh Institute of Urology and Transplantation from January 2019 to June 2019. All patients with age >18 years and chronic HCV infection were enrolled in the study. The patients with conditions other than chronic HCV infection that can result in enhanced liver injury and inflammation and liver fibrosis such as chronic HBV and HDV co-infection, metabolic conditions such as Non-alcoholic liver disease, Wilson disease, hemochromatosis and patients with hepatocellular carcinoma were excluded from the study. An informed consent was taken from all the patients before enrollment in the study.

All the patients then underwent history taking and physical examination followed by non-invasive laboratory investigations such as complete blood count and liver function tests. These patients then underwent imaging studies including ultrasound abdomen for features of chronic liver disease and Shear Wave Elastography (SWE) to assess fibrosis.

**GPR was calculated using the formula:**

$$\text{GPR} = \text{Gamma Glutamyl Transpeptidase} \left( \frac{\text{---}}{\text{ULN}} \right) \text{Platelet Count (109 /L)}$$

**Assessment of Fibrosis staging by SWE:**<sup>17</sup>

Two-dimensional SWE was performed using a LOGIQ E9 (GE Healthcare, Milwaukee, WI, USA) and a convex probe (1–6 MHz) by experienced radiologist with expertise in SWE and have already performed 200 SWE. After the visualization of the right liver lobe through an intercostal space, and a region of interest (ROI) was marked at less than 5 cm below the liver capsule and avoided major vessels. Measurements were taken 10–15 times while the patients held their breath for 5–10 seconds. The result was considered reliable when 10 successful shots and an Inter-Quartile Range(IQR)/median ratio should be less than 30%.

**SWE fibrosis grading:**<sup>17</sup>

Normal-mild fibrosis (F0-F1) - <5.4 kPa

Significant fibrosis (F2) - 5.4-9.9kPa

Advanced fibrosis (F3) - 9.9-12.9kPa

Cirrhosis(F4) - >12.9 kPa

### Statistical analysis:

The data was entered and analyzed using SPSS.v20. The baseline characteristics of all the patient were recorded. Continuous variables were expressed as Mean  $\pm$ SD, while categorical variables were expressed as frequency and percentages. Categorical variables were analyzed using Chi square test while student t test was used to analyze continuous variables. GPR was calculated and area under the Receiver operating curve (AUROC) was obtained to evaluate the diagnostic performance of GPR in predicting advanced fibrosis. A p-value of  $\leq 0.05$  was considered statistically significant.

### Results:

A total of 91 patients were included in the study. Out of 91 patients, 56 (61.5%) were males. Mean age was  $43.51 \pm 12.5$  years. The baseline characteristics of the patients are shown in **Table 1 and 2**. Child Pugh Class (CTP) A was noted in 60(65.9%) at the time of presentation while CTP B was noted in 31(64.1%) patients. Most of the patients did not have any comorbidities. At baseline, 52(54%) patients had F4 fibrosis as measured by SWE, while 24(37.4%) patients had F2 fibrosis, 10(11%) patients had F1 fibrosis and 5(5.5%) had F3 fibrosis. 57(62.6%) patients had  $\geq F3$  fibrosis (advanced fibrosis or cirrhosis). Mean platelet count was  $85 \pm 47 (\times 10^9/L)$ . Mean Gamma glutamyl Trans-peptidase was  $74 \pm 59$  (IU/L) .Mean GPR was  $1.5 \pm 2.1$ .

Comparison of continuous and categorical variables in terms of advanced fibrosis.(**Table 2 and 3**). Area under ROC was obtained for GPR in predicting advanced liver fibrosis( $\geq F3$ ) was 0.8 ( $p \leq 0.001$ ).(**Figure 1,2**) At a GPR cut off of  $\geq 0.6$ , the sensitivity was of 94.74%, specificity of

62%, positive predictive value of 80.69%, negative predictive value was of 87.50% and diagnostic accuracy of 82.42% in predicting advanced liver fibrosis.(  $\geq F3$ ) in patients with chronic hepatitis C .( **Table 4**)

#### **Discussion:**

Chronicity of liver disease can result in liver fibrosis and cirrhosis due to continued inflammation and non-avoidance of the etiological factor. In order to prevent disease progression, accurate and timely evaluation of degree of liver fibrosis is required. The gold standard test for this purpose is liver parenchymal biopsy but due to its invasiveness and technical expertise it is less often performed. Currently, non-invasive imaging methods such as fibroscan, SWE and TE are used to document the degree of liver fibrosis. However, high cost, the lack of availability of these techniques everywhere, presence of certain confounding factors along with lack of skilled operators can alter the interpretation of these imaging techniques. Currently, non-invasive tool are utilized for assessment of liver fibrosis in addition to these imaging techniques.

A model of gamma-glutamyl transpeptidase-to platelet ratio (GPR), as bedside available test useful in predicting the levels of liver fibrosis in chronic hepatitis B(CHB) patients in African population was proposed by Lemoine and his colleagues <sup>14</sup>. Ding R *et al.* <sup>18</sup> further validated this model in Chinese population. In our study, we have used the GPR ratio in predicting liver fibrosis in chronic HCV population using shear wave elastography to evaluate the degree of fibrosis.

Our study population included those patients who had chronic HCV infection and who were either under treatment for it with direct acting antivirals (Sofosbuvir/Daclatasvir) or already treated for it with the same regimen.

We categorized the patients into advanced fibrosis( $\geq F3$ ) and non-advanced fibrosis( $\leq F2$ ). The application of GPR in the two groups revealed the association of High GPR with advanced fibrosis( $p\text{-value}\leq 0.001$ ). Similar results for GPR were observed in the previous studies done by Vardar *et al.*<sup>19</sup> in a Turkish cohort in 2009, lemoine *et al.*<sup>14</sup> in an African cohort with AUROC of 0.80 and Ding R *et al.*<sup>18</sup> in a Chinese cohort with AUROC 0.78 in predicting advanced ( $\geq F3$ )fibrosis in CHB patients.<sup>19</sup> Lui *et al.*<sup>20</sup> used the GPR cutoff values suggested by lemoine *et al.*<sup>14</sup> in both HBsAg positive and HbsAg negative population in evaluating the degree of fibrosis and found lower sensitivity and specificity of GPR in both the groups. Previously, Nada *and her colleagues* have shown the association of high GPR of  $\geq 0.31$  with advanced fibrosis in chronic hepatitis C patients in an Egyptian population with a sensitivity of 92%, specificity of 88%,PPV of 86% and NPV of 91%<sup>21</sup>. In our study, GPR cut off of  $\geq 0.6$  was obtained using AUROC. The sensitivity of GPR was 91%, which was comparable to that of the previous studies. However, when compared to the previous studies, the specificity, PPV and NPV of GPR was lower in our studied population with a good diagnostic accuracy of 80% in predicting advanced liver fibrosis ( $\geq F3$ ) in patients with chronic hepatitis C.

There were certain limitations to our study. First of all the small sample size has affected the specificity of the GPR in our population. Secondly, for the assessment of liver fibrosis, SWE was used instead of liver biopsy which is the gold standard due to its invasiveness and high cost.

The strength of the study lies in the fact that this is the pioneer study from this region which has evaluated the role of GPR in HCV patients.

**Conclusion:**

The diagnostic accuracy of GPR in predicting advanced fibrosis in our population was effective in 82.42% patients. However, studies comprising larger sample size are needed to validate this score before using it as an important non-invasive assessment tool in predicting advanced fibrosis.

**Table 1-Baseline characteristics of the population included in the study (n-91)**

Study population		n-91(%)
Mean age(years±S.D)		43.5±12.5
Gender	Male	56(61.5)
	Female	35(38.5)
Hemoglobin(g/dL)		10.8±2.1
Total Leucocyte Count( $\times 10^9$ /L)		4.5±2.4
Platelet Count( $\times 10^9$ /L)		80±52
Total Bilirubin(mg/dl)		1.3±0.75
Alkaline Phosphatase(IU/L)		190±187
Aspartate Transaminase(AST)(IU/L)		59±52
Alanine Transaminase(ALT)(IU/L)		45±34
Gamma Glutamyl Transpeptidase(GGT)(IU/L)		74±59
Child Turcotte Pugh Score	A	60(65.9)
	B	31(34.1)
MELD score		10.9±3.4
Shear wave Elastography(SWE)		
F1 Fibrosis		10(11%)
F2 Fibrosis		24(37.4%)
F3 Fibrosis		5(5.5%)
F4 Fibrosis		52(54%)
Esophageal varices	Present	50(55)
	Absent	41(45)
Liver Fibrosis	Present	57(62.6)
	Absent	34(37.4)
GPR(Gamma Glutamyl Transpeptidase to Platelet Ratio)		1.5±2.1

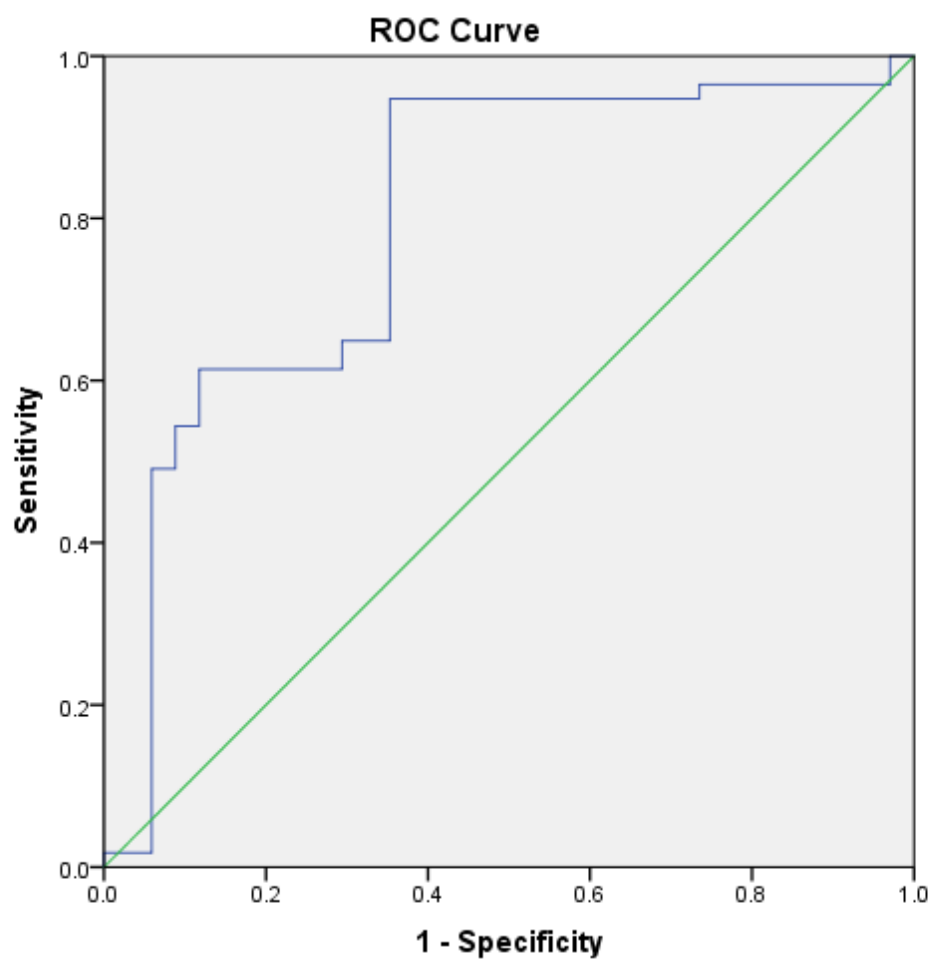
**Table 2- Comparison of continuous variables in terms of advanced liver fibrosis**

Variable	No Fibrosis( $\leq$ F2) (n-34) Mean $\pm$ SD	Advanced Fibrosis( $\geq$ F3)(n-57) Mean $\pm$ SD	p-value
Age	43.3 $\pm$ 10.75	43.65 $\pm$ 13.5	0.889
Hemoglobin(g/dL)	10.6 $\pm$ 2.5	10.9 $\pm$ 1.75	0.334
Total Leucocyte Count( $\times 10^9$ /L)	5.3 $\pm$ 2.6	4.1 $\pm$ 2.6	0.024
Platelet Count( $\times 10^9$ /L)	108.7 $\pm$ 66.6	62.9 $\pm$ 29.5	$\leq 0.001$
Total Bilirubin(mg/dl)	0.95 $\pm$ 0.62	14.5 $\pm$ 0.75	$\leq 0.001$
Alkaline Phosphatase(IU/L)	119 $\pm$ 44	224 $\pm$ 29	0.005
Aspartate Transaminase(AST)(IU/L)	35 $\pm$ 19	73 $\pm$ 60	0.001
Alanine Transaminase(ALT)(IU/L)	26 $\pm$ 13	57 $\pm$ 37	$\leq 0.001$
Gamma Glutamyl Transpeptidase(GGT)(IU/L)	48 $\pm$ 21	90 $\pm$ 69	0.001
CTP score	5.7 $\pm$ 1.2	6.4 $\pm$ 1.3	0.015
MELD Score	10.2 $\pm$ 4.3	11.3 $\pm$ 3.6	0.208
GPR	1.0 $\pm$ 2	1.8 $\pm$ 2	0.04

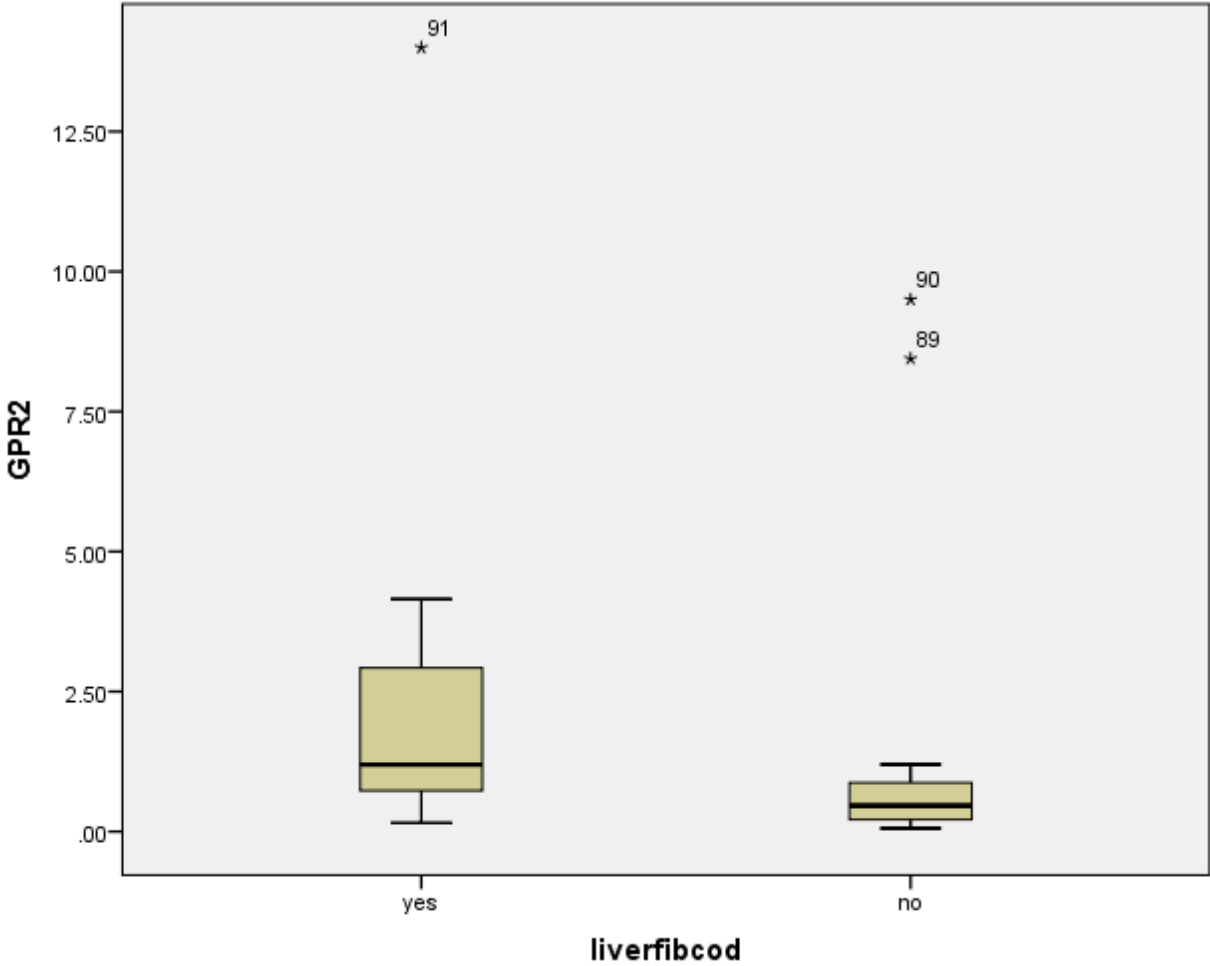
**Table 3- Comparison of categorical variables in terms of advanced liver fibrosis**

Variable		Liver fibrosis ( $\geq F3$ )(n-57)	No Fibrosis( $\leq F2$ )(n-34)	p-value
		n(%)	n(%)	
Gender	Male	39(68.4)	17(50)	0.08
	Female	18(31.6)	17(50)	
Esophageal varices	Yes	43(75.4)	23(67.6)	0.421
	No	14(24.6)	11(32.4)	
CTP score	A	32(56.1)	28(82.4)	0.01
	B	28(43.9)	6(17.6)	
GPR	$\geq 0.6$	54(94.7)	13(38.2)	$\leq 0.001$
	$< 0.6$	3(5.3)	21(61.8)	

**Figure 1:-Area under Receiver Operating curve for GPR in the prediction of significant hepatic fibrosis( $\geq F3$ ) in chronic hepatitis C is 0.8 (p-value- $\leq 0.001$ )**



**Figure 2-Box plot showing association of high Gamma Glutamyl Transpeptidase(GPR) with advanced liver fibrosis**



**Table 4-Diagnostic accuracy of Gamma Glutamyl Transpeptidase(GPR) in predicting advanced ( $\geq$ F3) liver fibrosis**

Statistics	Percentage(95%CI)
Sensitivity	94.74%(85.3-98.9)
Specificity	62%(43.6-78.8)
Positive Predictive Value	80.60%(72.96-86.48)
Negative Predictive Value	87.50%(69.3-95.6)
Diagnostic Accuracy	82.42%(73-89.6)

**COMPETING INTERESTS DISCLAIMER:**

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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