ASSESSMENT OF SOME BIOMARKER OF LIVER FIBROSIS IN PATIENTS WITH CHRONIC HEPATITIS B SUFFERED FROM CHRONIC KIDNEY DISEASES

I.A. Abdulla, F.f. Rija*

Department of Biology, College of Science, Tikrit University, Tikrit, Iraq.

* Corresponding author: imanah1966@gmail.com

Abstract. Globally, chronic hepatitis B virus (CHB) infection causes chronic liver disease, fibrosis, cirrhosis, and hepatocellular carcinoma. Furthermore, many CHB patients have comorbid medical problems and varying degrees of renal impairment. The aim of the study was to evaluate the role of CHI3L1 in diagnosing liver fibrosis in the studied cases and assess the clinical and physiological factors during disease cases. 90 blood samples were collected from patients in the Dialysis Unit at Tikrit Teaching Hospital, Yathrib township, Aldhuluiya city, Al-Shuhada Health Center in the period from September 1, 2022, to February 28, 2023. 60 of these samples were obtained from patients with chronic hepatitis B virus diagnosed by doctors and divided into two groups: group one: 30 chronic hepatitis B patients (19 males, 11 females) aged 20-67; group two: 30 chronic hepatitis B patients with chronic kidney disease (CKD) (18 males, 12 females) aged 24-68. The control group consisted of 30 healthy individuals (19 males, 11 females) aged 20-55. Results indicated a substantial increase in CHI3L1 levels (174.41 ± 38.45a) in CHB patients compared to CKD patients (137.30 ± 37.8b) and controls (126.10 ± 30.0b) at P ≤ 0.05. A substantial rise in creatinine levels was observed in patients with chronic hepatitis B due to CKD (group 2) compared to CHB (group 1) and controls with a p-value ≤0.05. The mean ± SD of B.urea was 151.40 ± 24.2a in CKD patients (group 2), 31.26 ± 7.55b in CHB patients (group 1), and 27.54 ± 6.24b in controls (p-value ≤ 0.05). At p-value <0.05, CRP mean ± SD were 23.860 ± 4.220a in CKD patients (group 2), 4.040 ± 0.422b in CHB patients (group 1), and 4.200 ± 0.436b in controls. In chronic hepatitis B patients, CHI3L1 levels increase significantly. Chronic renal illness increases B.urea, creatinine, and CRP in chronic hepatitis B.

Keywords: chronic hepatitis B, (YKL-40) chitinase-3-like protein 1, CRP, blood urea, creatinine.

List of Abbreviations

CHB – chronic hepatitis B
HCC – hepatocellular carcinoma
CHI3L1 – chitinase-3-like protein 1
CKD – chronic kidney diseases
HBV – hepatitis B virus
CRP – C-reactive protein
APR – acute phase reactants
ESKD – end-stage kidney disease
PEW – protein-energy wasting

Introduction

Chronic viral hepatitis refers to a pathological condition affecting the liver that exhibits a progressive nature, ultimately resulting in cirrhosis in approximately 20-30% of affected individuals (Papatheodoridis *et al*., 2006). Certain viruses, like A, B, C, D, and E, have the potential to induce hepatitis. Numerous host and viral factors, including age, gender, alcohol consumption, infection duration, and viral coinfections, have been linked to the severity of the histology and the rate of fibrosis development (Tong *et al*.,1995).

Liver fibrosis is a pathological condition characterized by the excessive deposition of extracellular matrix proteins, particularly collagen, in the liver. This process is commonly observed in various forms of chronic liver diseases, and its progression can ultimately lead to the development of cirrhosis. Chronic hepatitis B virus (HBV) infection is also correlated with an elevated susceptibility to hepatocellular carcinoma (HCC), a prevalent form of malignancy and a leading cause of mortality (Papatheodoridis *et al*., 2006; Mohammed, 2022). CHI3L1 is classified within the glycoside hydrolase family 18. The protein exhibits affinity towards hyaluronic acid, chitin, and heparin, and its activity is modulated by alterations in the extracellular matrix, growth factors, cytokines, pharmaceutical agents, and stressors. The gene CHI3L1 has been found to exhibit a significant correlation with various diseases, such as asthma, arthritis, sepsis, liver fibrosis,

and diabetes (Zhao *et al*., 2020). Serum CHI3L1 has shown promise as a diagnostic marker for liver fibrosis (Huang *et al*., 2022). The kidney assumes a pivotal role within the urinary system and is responsible for various essential homeostatic functions such as electrolyte regulation, acid-base balance maintenance, and blood pressure control. The kidneys function as an inherent blood filter and eliminate waste materials, which are subsequently transported to the urinary bladder. There has been a proposal suggesting that hepatitis B virus (HBV) infection could potentially have a pivotal role in the pathogenesis of specific types of glomerulonephritis, which may exhibit a slow but relentless clinical progression (Saod *et al.*, 2019).

Materials and Methods

Study samples were collected from patients in Dialysis Unit in Tikrit Teaching Hospital, Yathrib township, Aldhuluiya city, AL-Shuhada health center through the periods of 1st September 2022 to the end of February 2023. The study comprised a total of 90 blood samples, consisting of 60 samples obtained from patients who had previously been diagnosed with HBV by medical professionals. The samples were categorized into two distinct groups based on the evaluations conducted by the healthcare personnel. *Group one* consists of patients diagnosed with chronic hepatitis (CHB); this group comprises a total of 30 patients, with 19 being male and 11 being female. The age range of the patients is from 20 to 67 years. *Group two:* patients with CHB suffering from chronic kidney diseases (CKD) under hemodialysis; this group consists of 30 patients (18 males and 12 females) whose ages ranged from 24 to 68 years. 30 control apparently healthy (19 males and 11 females) whose ages ranged from 20 to 55. A total of 5 ml of venous blood from each patient. The blood was placed in a Gel tube without any anticoagulant in order to obtain serum. Subsequently, the tube was left at room temperature (20-25 °C) until coagulation occurred. The coagulated blood was then separated by centrifugation at a speed of 3500 pm/min for a duration of 15 minutes. Following centrifugation, the serum was extracted using a micropipette and distributed equally into two Ipendrof tubes that were free of coagulants. The tubes were securely sealed, and the serum was subsequently stored at sub-freezing temperatures for future use in tests.

Ethical approval

This study was conducted based on the ethical standards stipulated in the Declaration of Helsinki. Before taking the sample, the patient's informed written and verbal agreement was obtained (after the review and approval of the study protocol and subject's information by the local ethics committee according to document number 12954 (including the number and the date 7/9/2022) to get this approval).

Results

The results of the current study, as shown in Table 1, indicated a significant increase in CHI3L1 (174.41 ± 38.45) in patients of group 1 compared to control at the level of probability $P \leq 0.05$, with no differences between group 2 and control.

Table 1

Relation between chronic hepatitis B patients (group 1), chronic hepatitis B patients who suffered from chronic kidney diseases (group 2), and the control group regarding the mean ± SD of CHI3L1

Groups	Number	CHI3L1
Group 1	n = 30	174.41 ± 38.45 a
Group 2	n = 30	137.30 ± 37.8 b
Control	n = 30	126.10 ± 30.0 b
P-value ≤ 0.05**		

The findings of the present study demonstrated a statistically significant increase in the mean ± SD of creatinine in patients in group 2 compared to patients in group 1 and control that were 7.069 ± 1.981a, 1.000 ± 0.1809b, 0.915 ± ± 0.1482b, respectively, at p-value ≤ 0.05. The mean ± SD of B.urea were 151.40 ± 24.2a in patients from group 2, 31.26 ± 7.55b in group 1,

27.54 ± 6.24b in control at p-value ≤ 0.05. The mean ± SD of CRP were 23.860 ± 4.220a in patients from group 2, 4.040 ± 0.422b in group 1, 4.200 ± 0.436b in control at p-value < 0.05 (Table 2).

Correlation coefficient (r) between YKL-40 with all parameters in CHB patients (Table 3)

A: in chronic hepatitis B patients, there was a positive correlation between YKL-40 and urea, creatinine, CRP with correlation coefficient r = = 0.456, 0.421, 0.688, respectively, that was shown by regression plots in Figures 1, 2, 3.

B: in chronic hepatitis B with chronic kidney diseases, a negative correlation existed between YKL-40 and r = urea, creatinine, CRP with correlation coefficient r = -0.503, -0.070, -0.538**,** respectively**,** as shown by regression plots in Figures 4, 5, 6.

Discussion

The present study demonstrates an elevated mean ± SD of CHI3L1 in group 1 when compared to group 2 and the control group. This study is in agreement with the findings of Bao et al. (2022) who found that serum CHI3L1 expression in liver fibrosis patients was notably elevated compared to those without liver fibrosis. Also variations in the serum CHI3L1 expression levels across different grades of liver fibrosis were observed. The levels of expression demonstrated a rise as the degree of liver fibrosis increased. The serum biomarker CHI3L1 has the ability to differentiate between the early stage (S1) and late stage (S3-4) of liver fibrosis. A positive correlation was observed between the expression of CHI3L1 and the extent of fibrosis. The chronic hepatitis group had greater serum CHI3L1 levels than the healthy control group, and only the mesenchymal structure of the portal area expresses it; hepatocytes do not. The liver expresses and secretes CHI3L1. Extracellular matrix protein causes liver fibrosis. Serum CHI3L1 levels rise with tissue destruction from chronic hepatitis through liver cirrhosis to HCC (Qiu & Zhang, 2022). The elevated concentration of urea in the human body is primarily attributed to the enzymatic degradation of proteins within the hepatic system, followed by the subsequent elimination

Table 2

Differences in some biochemical variables in the studied samples

Groups	Creatinine	Urea	CRP
Group 1 (n:30)	1.000 ± 0.1809 b	31.26 ± 7.55 b	4.040 ± 0.422 b
Group 2 (n:30)	7.069 ± 1.981 a	151.40 ± 24.2 a	23.860 ± 4.220 a
Control (n:30)	0.915 ± 0.1482 b	27.54 ± 6.24 b	4.200 ± 0.436 b
P-Value	0.05**	0.05**	0.05**

Table 3

Correlation coefficient (r) between YKL-40 with all parameters in CHB and CHB with CKD patients

Parameters	G1 (CHB) YKL-40 r	G2 (CHB with CKD) YKL-40 r
Urea	0.456	-0.503
Creatinine	0.421	-0.070
CRP	0.688	-0.538

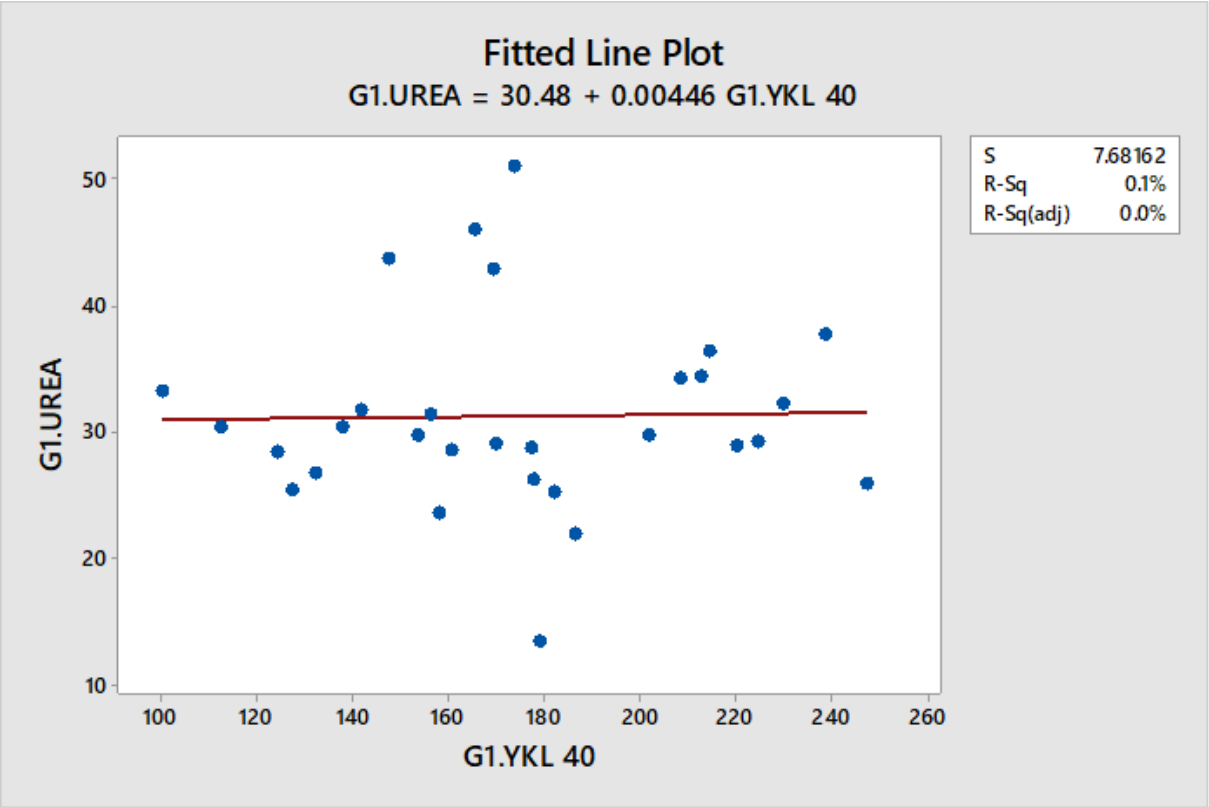

Fig. 1. Correlation between YKL-40 with Urea in chronic hepatitis B (CHB) patients

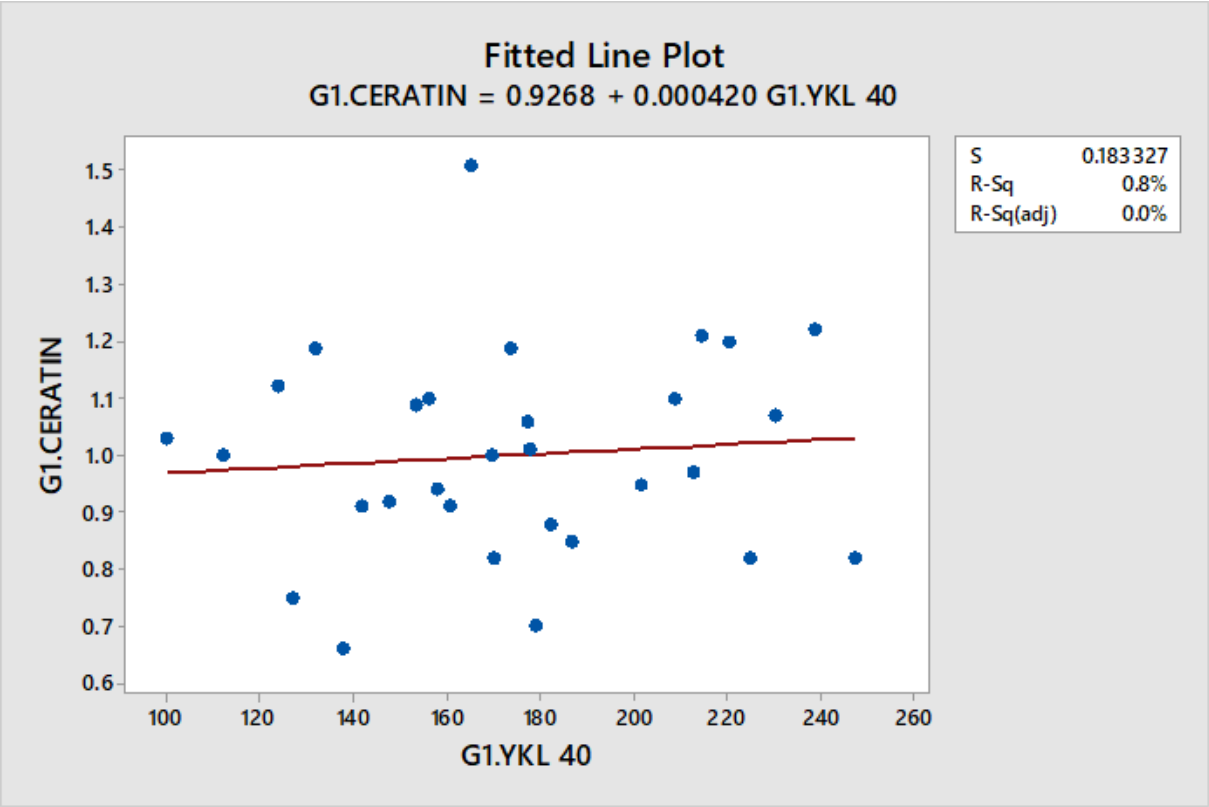

Fig 2. Correlation between YKL-40 with creatinine in chronic hepatitis B (CHB) patients

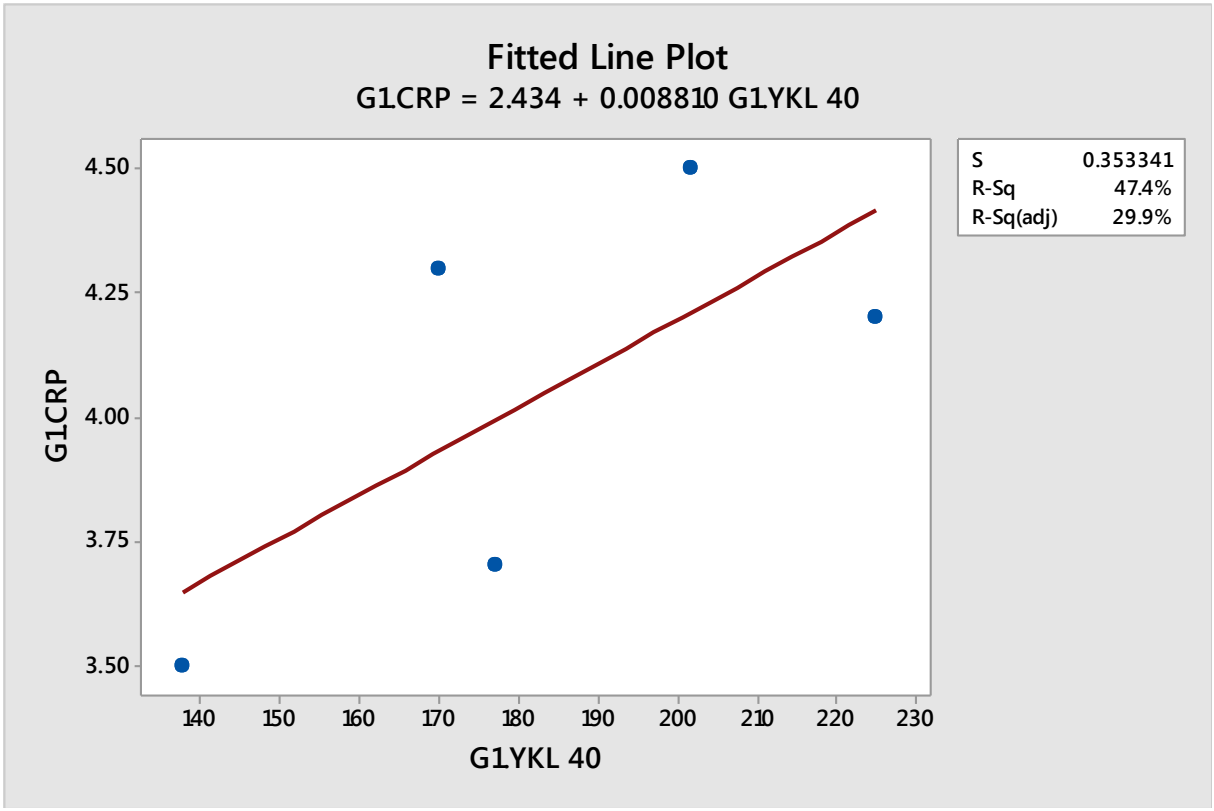

Fig. 3. Correlation between YKL-40 with CRP in chronic hepatitis B (CHB) patients

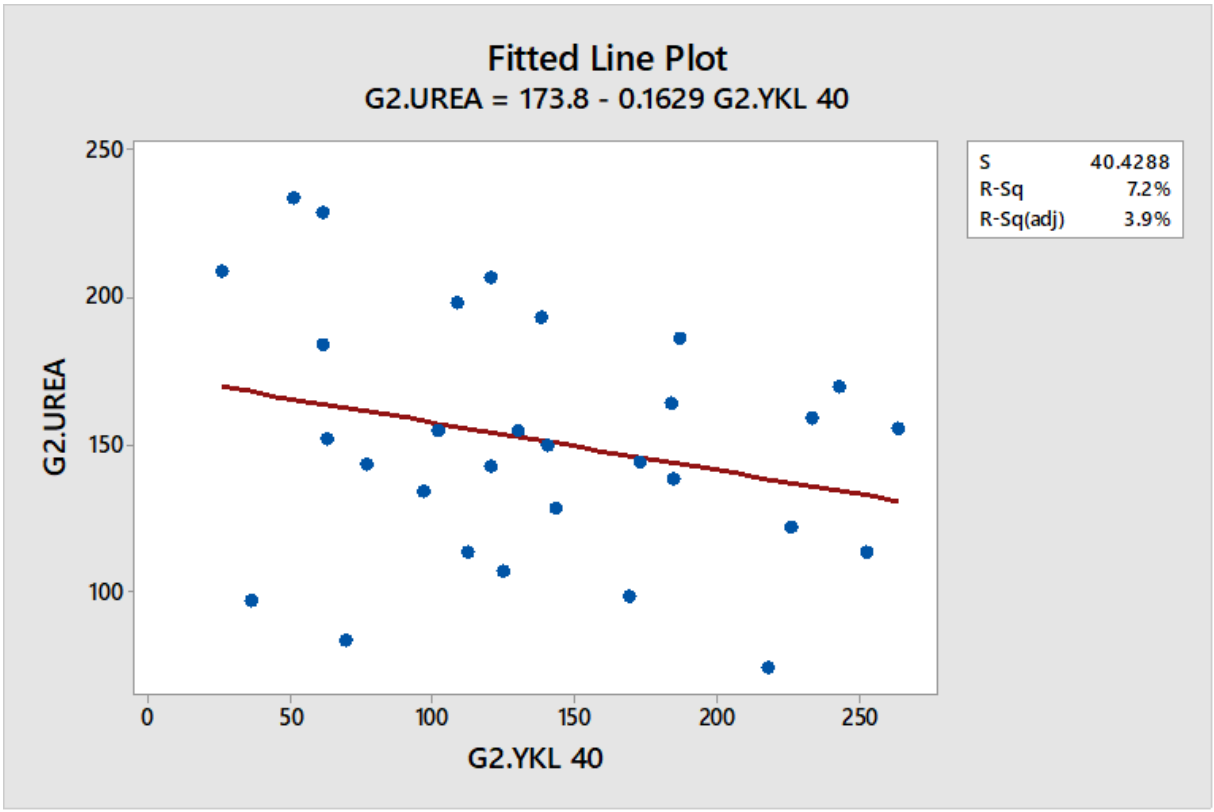

Fig. 4. Correlation between YKL-40 with Urea in chronic hepatitis B (CHB) with CKD

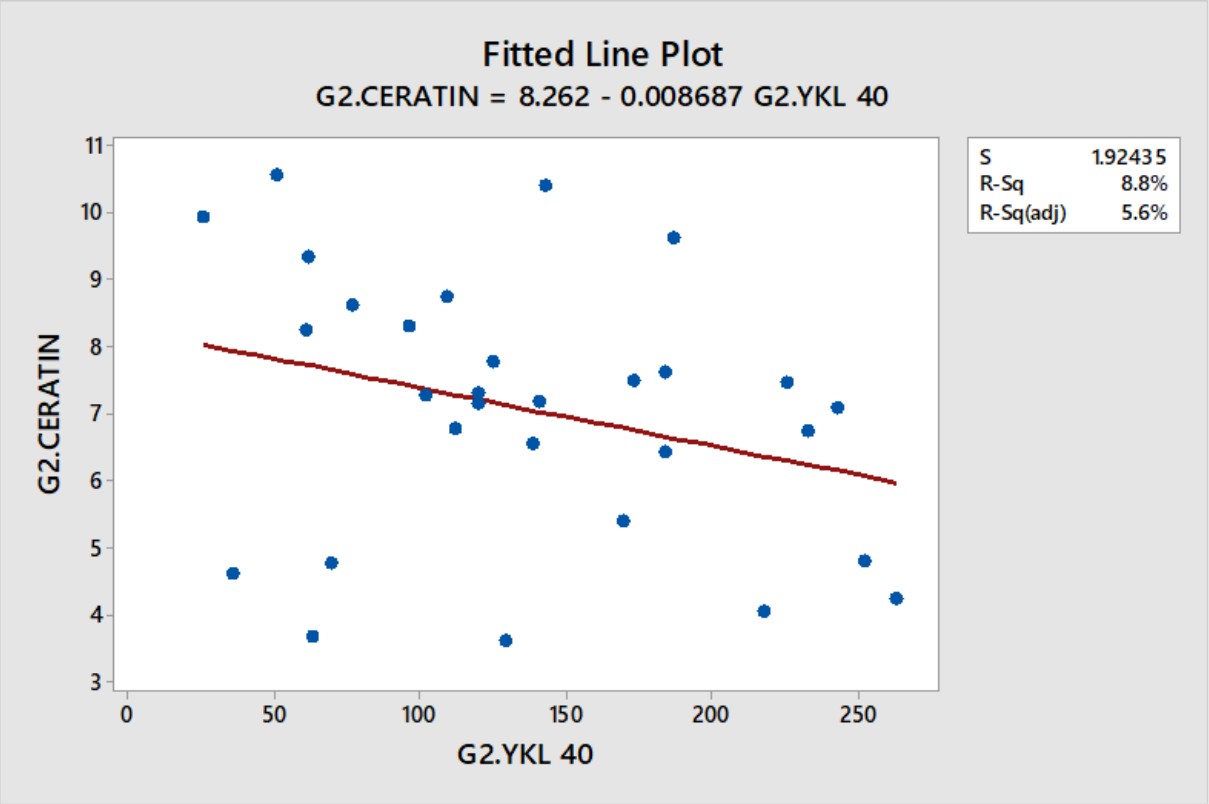

Fig. 5. Correlation between YKL-40 with creatinine in chronic hepatitis B (CHB) with CKD

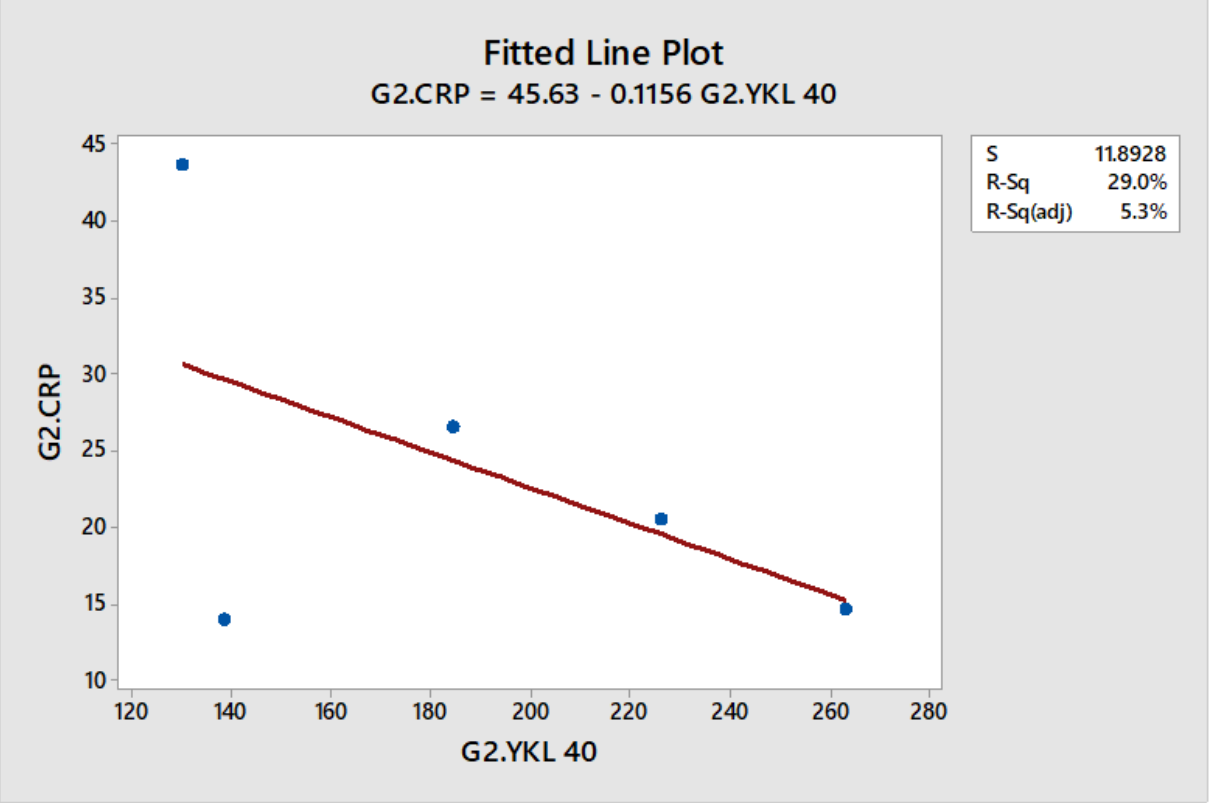

Fig. 6. Correlation between YKL-40 with CRP in chronic hepatitis B (CHB) with CKD

Table 2 of urea through renal excretion (Salazar, 2014). Elevated levels of creatinine and urea are indic-ative of a probable decline in renal function (McClellan *et al*., 2017). The study's findings agreed with those of Griffin et al. (2020) who demonstrated an elevation in urea and creatinine levels among patients undergoing hemodialysis. This increase was found to be associated with a decline in functional renal healing.

The present study demonstrates an elevation in C-reactive protein (CRP) levels within group 2. These findings are in line with the results reported by Lalramenga et al. (2019) who observed a significant increase in CRP levels among individuals with chronic kidney disease (CKD). Inflammation is accompanied by increased serum protein concentration known as acute phase reactants (APR) (Kushner, 1982). In the context of the acute phase response, it has been observed that the typical levels of plasma proteins, which are normally regulated by homeostatic mechanisms, can undergo significant alterations. These alterations are believed to play a role in the host's defense mechanisms and other adaptive capabilities (Kushner & Samols, 2011). Inflammatory conditions increase plasma concentrations of acute-phase proteins like CRP or reduce them like albumin. Inflammation is often assessed by measuring these proteins (Mihai *et al*., 2018).

Chronic inflammatory processes are frequently observed in individuals diagnosed with chronic kidney disease (CKD), particularly those in the advanced stage known as end-stage kidney disease (ESKD). This occurrence can be attributed to various underlying factors, such as the presence of uremic milieu, elevated levels of pro-inflammatory cytokines circulating in the body, oxidative stress, carbonyl stress, protein-energy wasting (PEW), increased susceptibility to infections (particularly those related to dialysis access), and other contributing factors (Yeun *et al*., 2000). The "acute-phase response" can last months to years and become chronic, with positive acute-phase markers like CRP (Kalantar-Zadeh, 2007). The synthesis of creatinine occurs in a continuous manner within the human body, and its elimination takes place through the process of renal glomerular filtration rate (Palmiere & Mangin, 2015). The decline in renal function can impact the filtration rate of creatinine through the kidneys, making it a useful indicator of renal function. As the kidneys lose their ability to effectively clear creatinine through urine excretion, renal function declines, leading to elevated levels of creatinine in the bloodstream (De Almeida *et al*., 2016). The findings of this study are consistent with the conclusions drawn by Li et al. (2023), who observed a strong correlation between elevated blood creatinine levels and patients diagnosed with stages 1-4 of chronic kidney disease (CKD). The serum creatinine values observed in patients with chronic kidney diseases were significantly elevated compared to the control group, both in terms of mean values and range (Kamal, 2014).

Conclusions

It was concluded that the levels of CHI3L1 have a significant increase in patients with CHB, while B.urea, creatinine, and CRP increase in chronic hepatitis B suffered from CKD. A positive correlation between YKL-40 and urea, creatinine, CRP was observed in CHB patients; however, there was a negative correlation between YKL-40 and r = urea, creatinine, CRP in CHB patients who suffered from CKD.

References

BAO J., OUYANG Y., QIAO L., HE J., LIU F., WANG Y. ... & LI Z. (2022): Serum CHI3L1 as a biomarker for non-invasive diagnosis of liver fibrosis. *Discovery Medicine* **33**(168), 41–49.

DE ALMEIDA M.L., SAATKAMP C. J., FERNANDES A.B., PINHEIRO A.L.B. & SILVEIRA L. (2016): Estimating the concentration of urea and creatinine in the human serum of normal and dialysis patients through Raman spectroscopy. *Lasers in medical science* **31**, 1415–1423.

GRIFFIN B.R., GIST K.M. & FAUBEL S. (2020): Current status of novel biomarkers for the diagnosis of acute kidney injury: a historical perspective. *Journal of intensive care medicine* **35**(5), 415–424.

HUANG X., ZHUANG J., YANG Y., JIAN J., AI W., LIU C. ... & PENG S. (2022): Diagnostic value of serum chitinase-3-like protein 1 for liver fibrosis: a meta-analysis. *BioMed Research International* **2022**.

KALANTAR-ZADEH K. (2007): Inflammatory marker mania in chronic kidney disease: pentraxins at the crossroad of universal soldiers of inflammation. *Clinical Journal of the American Society of Nephrology* **2**(5), 872–875.

KAMAL A. (2014): Estimation of blood urea (BUN) and serum creatinine level in patients of renal disorder. *Indian J Fundam Appl Life Sci* **4**(4), 199–202.

KUSHNER I. (1982): The phenomenon of the acute phase response. *Annals of the New York Academy of Sciences* **389**(1), 39–48.

KUSHNER I. & SAMOLS D. (2011): Oswald Avery and the. *Pharos* **15**.

LALRAMENGA P.C., GUPTA S &. NAVEEN P. (2019): Study of C-reactive protein significance in chronic kidney disease. *International Journal of Contemporary Medical Research* **6**(6), F22–F25.

LI Q., XIE J., TAO H. & WANG W. (2023): Wcn23-0817 validation of blood urea nitrogen/serum creatinine ratio equation to evaluate dietary protein intake in patients with stage 1~4 chronic kidney disease and prognostic assessment. *Kidney International Reports* **8**(3), S153.

MCCLELLAN W.M., PLANTINGA L.C., WILK A.S. & PATZER R.E. (2017): ESRD databases, public policy, and quality of care: Translational medicine and nephrology. *Clinical Journal of the American Society of Nephrology: CJASN* **12**(1), 210.

MIHAI S., CODRICI E., POPESCU I.D., ENCIU A.M., ALBULESCU L., NECULA L.G. ... & TANASE C. (2018): Inflammation-related mechanisms in chronic kidney disease prediction, progression, and outcome. *Journal of immunology research* **2018**.

MOHAMMED H.K. (2022): Study of Physiological and Some Biochemical Parameters in Patients Infected with Chronic Hepatitis-B Virus. *HIV Nursing* **22**(2), 2039–2044.

PALMIERE C. & MANGIN P. (2015): Urea nitrogen, creatinine, and uric acid levels in postmortem serum, vitreous humor, and pericardial fluid. *International journal of legal medicine* **129**, 301–305.

PAPATHEODORIDIS G.V., CHRYSANTHOS N., SAVVAS S., SEVASTIANOS V., KAFIRI G., PETRAKI K. & MANESIS, E.K. (2006): Diabetes mellitus in chronic hepatitis B and C: prevalence and potential association with the extent of liver fibrosis. *Journal of viral hepatitis* **13**(5), 303–310.

QIU H. & ZHANG X. (2022): The Value of Serum CHI3L1 for the Diagnosis of Chronic Liver Diseases. *International Journal of General Medicine*, 5835–5841.

SALAZAR J.H. (2014): Overview of urea and creatinine. *Laboratory medicine* **45**(1), e19–e20.

SAOD W.M., ZAIDAN T.A. & ABDUL RAZZAK ALFALUJI A.W. (2019): Hepatitis B and renal function of patients with chronic hepatitis B in Fallujah district, Iraq. *Biochemical & Cellular Archives* **19**.

TONG M.J., EL-FARRA N.S., REIKES A.R. & CO R.L. (1995): Clinical outcomes after transfusion-associated hepatitis C. *New England Journal of Medicine* **332**(22), 1463–1466.

YEUN J.Y., LEVINE R.A., MANTADILOK V. & KAYSEN G.A. (2000): C-reactive protein predicts all-cause and cardiovascular mortality in hemodialysis patients. *American journal of kidney diseases* **35**(3), 469–476.

ZHAO T., SU Z., LI Y., ZHANG X. & YOU Q. (2020): Chitinase-3 like-protein-1 function and its role in diseases. *Signal Transduction and Targeted Therapy* **5**(1), 201.