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Original Research Article

Study of Biochemical Markers in Non-Alcoholic Fatty Liver Disease

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Abstract:

Background: NAFLD, or non-alcoholic fatty liver disease, is a horrifying public health issue. Affluent societies are thought to have the highest prevalence of chronic liver disease, which affects 2-10% of the general population and includes a wide spectrum of conditions from simple steatosis to nonalcoholic steatohepatitis (NASH) in adults and children. The purpose of the current investigation was to evaluate the biochemical markers in patients with non-alcoholic fatty liver disease (NAFLD).

Methods: Twenty-five normal individuals and 75 NAFLD patients had their biochemical parameters examined. The use of abdominal ultrasonography in diagnosis established the presence of hepatic steatosis. Every patient with a NAFLD diagnosis had their biochemical parameters examined, and the association between the condition and control was examined.

Results: When comparing the non-fatty liver control group to the NAFLD patients, all biochemical parameters showed increases, and the differences were found to be statistically (P value less than 0.005) significant.

Conclusion: In NAFLD patients, alterations in biochemical markers are linked to the disease. Early identification will be key to changing the course of the disease, postponing consequences, and contributing significantly to preventive cardiology.

Keywords: Non-alcoholic Fatty Liver Disease, Steatohepatitis, Insulin Resistance.

Introduction

Non-alcoholic fatty liver disease (NAFLD), the most common kind of liver disease, is characterized by hepatic fat accumulation that exceeds 5% of liver weight in people who have never had a history of excessive alcohol use. It can progress to liver cancer, cirrhosis, and nonalcoholic steatohepatitis (NASH). [1-4]

The prevalence varies with geographic location. For example, the prevalence of NAFLD is 24.13% in North America and 27.37% in Asia; the lowest prevalence (13.48%) is found in Africa, while the highest prevalence (31.79%) is found in the Middle East. [5] Estimates indicate that the prevalence of NAFLD is increasing everywhere. [6,7]

Dyslipidemia, which is marked by hypertriglyceridemia, reductions in high-density lipoprotein cholesterol (HDL-C), and increases in very low-density lipoprotein (VLDL) and low-density lipoprotein cholesterol (LDL-C), is a significant comorbidity that is commonly seen in NAFLD patients. [8,9]Liver ultrasonography is the most widely used technique for identifying fatty liver in the general population. [10] Patients with abnormally increased aminotransferase levels often consult gastroenterologists or hepatologists for management. As a result, numerous studies employ aberrant levels of alanine transaminase (ALT) and aspartate transaminase (AST) to identify non-alcoholic fatty liver disease (NAFLD). [11,12]

As a result, measurements of blood lipids, insulin resistance (IR), and aminotransferases are frequently used in clinical settings to diagnose NAFLD. In non-alcoholic fatty liver disease (NAFLD), lipid profile, AST, ALT, fasting blood sugar (FBS), CRP, and fasting insulin level are significant factors. [13,14]

Because they enable physicians to determine the severity and prognosis of the condition and initiate treatment programs earlier, these indicators are helpful substitutes for liver biopsies. [15] The purpose of this study was to assess the lipid profile and liver function tests in NAFLD patients and investigate any potential correlations between them and different stages of the disease.

In order to characterize and diagnose nonalcoholic fatty liver disease, the purpose of the current study was to assess the value of the following tests: uric acid (UA), aspartate transaminase (AST), alanine transaminase (ALT), fasting blood sugar (FBS), triglycerides (TGL), cholesterol (CHOL), total bilirubin (TB), and ALT/AST ratio.

Material and Methods

From June 2023 to November 2023, the current study was conducted in the biochemistry department of the Darbhanga Medical College and Hospital in Laheriasarai, Bihar. 75 patients with nonalcoholic fatty liver disease were investigated in 100 instances. Twenty-five clinically healthy subjects were used as the control group.

When insulin resistance was present but alcohol consumption, viral, autoimmune, genetic, or induced liver disease was absent, the clinical diagnosis of nonalcoholic fatty liver disease (NAFLD) was made. This correlation was confirmed by additional testing (ultrasound abdomen demonstrating fatty liver). While a small percentage of NAFLD patients reported abdominal pain in the right upper quadrant, the majority of individuals showed no symptoms.The study excluded patients with known liver disorders, cancer, usage of amiadarone, corticosteroids, tamoxifen, methotrexate, or high dosage estrogen, jejunoileal by pass or extensive bowel resection, and daily alcohol intake of more than 30 grams for men or 20 grams for women.

After gaining consent, systematic data collection was done from both cases and controls. The body mass index (BMI), which is defined as weight (kg)/height (m^2), was computed after age, gender, height, and weight were recorded. Serum Total Bilirubin, Alanine Transaminase, Aspartate Transaminase, Triglycerides, Cholesterol, Uric Acid, and Fasting Blood Sugar were measured using the blood samples that were drawn.

Using SPSS software version 20, the mean, standard deviation, and p values for the cases and controls were calculated and examined.

Results

FBS, cholesterol, TGL, UA, AST, ALT, TSB, ALT/AST ratio, and BMI were among the parameters examined in 100 participants; 75 of them were NAFLD patients, while the remaining 25 were controls. Statistics have been used to analyze the data.

Of the cases, 47 (62.67%) were female while the remaining 28 (37.33%) were male. Ten (40%) of the controls were men, and the remaining fifteen (60%) were women. Cases had an average age of 48.28 ± 9.26 and controls had an average age of 40.40 ± 11.67.

Table1:Mean±SDofallparametersin bothcasesandcontrols

Parameters	Cases	Controls
BMI	32.15±2.47	22.96±4.64
FBS	124.19±10.89	75.24±8.84
Triglycerides	248.09±168.37	173.60±55.43
Cholesterol	210.99±43.25	167.40±25.88
TotalBilirubin	0.15±0.28	0.52±0.25
ALT	34.45±10.68	25.04±8.76
AST	27.77±11.62	31.56±7.52
ALT/ASTRatio	1.33±0.41	0.78±0.17
Uric acid	5.52±1.43	3.69±1.10

All the cases had above normal BMI values (> 25 kg/m2). 22 of them are (29.33%) over weight, while 53 (70.67%) were obese. The average BMI for the cases was 32.15 ± 2.47, the same for controls being 22.96 ± 4.64.

This difference was found statistically highly significant (P < 0.001).34 patients (45.33%) had FBS values > 126mg% and an equal number have values between 110 & 125mg%. The mean FBS of NAFLD cases is 124.19 ± 10.89 which is higher than the mean FBS of controls 75.24 ± 8.84, the difference being statistically significant (P < 0.001). 62 patients (82.67%) had hypertriglyceridemia. The mean serum triglycerides in the controls 173.6 ± 55.43, while that of the cases was much higher at 248.09 ± 168.37. The difference was statistically significant with P < 0.001. 60% of cases, i.e., 45 patients had hypercholesterolemia. The mean serum cholesterol of NAFLD patients is 210.99 ± 43.25 and in controls is 167.4 ± 25.88. The increase was statistically significant (P < 0.001).The mean ALT value of controls 24.04 ± 8.76 and the mean ALT of NAFLD cases 34.45 ± 10.63.P value was < 0.001, suggesting that the elevation of ALT among patients shows statistically significant.

The mean AST values of controls 31.56 ± 7.52 and that of cases 27.77 ± 11.62. This difference in the values was not statistically significant. The mean ± SD values of ALT / AST ratio in controls 0.78 ± 0.17. The value of the same ratio 1.33 ± 0.41 in NAFLD patients. This shows a statistically significant increase with P values < 0.001.The means uric acid levels in NAFLD cases 5.52 ±

1.43, whereas in controls it 3.69 ± 1.10. This difference found was statistically significant. The mean Total serum bilirubin of controls 0.52 ± 0.25mg% and that of cases 0.51 ± 0.28. There is no statistically significant difference in TSB of both groups. In order to assess the significance of alterations observed in different parameters analyzed, in patients compared to those of controls, the sensitivity and specificity of these parameters are calculated. This was done with the help of best cut off values derived from ROC curves.

Table2:RelativeOperatingCharacteristiccurvefeaturesofparameters

Parameter	Bestcutoff value	Sensitivity	Specificity	Diagnosticefficiency
BMI	26kg/m^2	100%	100%	100%
FBS	103mg/dl	98%	100%	99%
Triglyceride	221mg/dl	43%	80%	52%
Cholesterol	193mg/dl	65%	84%	70%
TotalBilirubin	0.9mg/dl	15%	96%	35%
ALT	55.5U/L	9%	100%	37%
AST	49.5U/L	11%	100%	33%
ALT/ASTRatio	1.04	85%	96%	88%
UA	3.5mg/dl	96%	64%	88%

The test values that yielded the highest combination of sensitivity, specificity, and diagnostic efficacy that is, the point nearest to the upper left corner of the ROC curve were chosen to determine the best cut off values.

Table3: Theareaundercurveoftheroccurves ofparameters

Parameter	Area underthe Curve	Standarderror	Asymptomatic95%confidenceinterval	
			Lowerbound	Upperbound
BMI	1.000	0.000	1.000	1.000
FBS	1.000	0.000	1.000	1.000
Triglycerides	0.625	0.062	0.507	0.743
Cholesterol	0.807	0.044	0.720	0.894
SerumBilirubin	0.478	0.063	0.354	0.602
ALT	0.743	0.062	0.622	0.864
AST	0.335	0.057	0.223	0.446
ALT/ASTratio	0.985	0.009	0.967	1.003
UA	0.842	0.046	0.752	0.932

To assess the ability of various analyses to differentiate, the area under the curve table of the ROC curves for various parameters is compared. Higher sensitivity and specificity were used to distinguish between controls and NAFLD patients using BMI, FBS, and ALT/AST ratios. Even if they are more specific, AST, ALT, TSB, TGL, and cholesterol are not sensitive enough to detect NAFLD instances. With an area under the curve value of 0.985, the ALT/AST ratio is a more effective diagnostic marker for liver function.

Discussion

The one most reliable correlation reported with NAFLD is obesity. [16] 40% to 100% of the cases were recorded in the majority of the studies. There is a direct relationship between BMI and the likelihood of developing a fatty liver, according to certain epidemiological research. There is a lack of clarity regarding the biochemical processes that lead from obesity-related steatosis to NAFLD and ultimately cirrhosis. Hepatic macrophage dysfunction has been shown to occur in obesity, and it has been proposed that this may promote steatohepatitis by sensitizing hepatocytes to endotoxin. These findings were made possible with the aid of a novel model of obesity-associated liver illness. [17–22]

In the current study, all patients (100%) had BMIs over normal; of these, 70.67% were classified as obese, while the remaining 29.33% were overweight. Numerous authors have noted that patients with non-alcoholic fatty liver disease (NAFLD) often have elevated plasma glucose levels and are frequently linked to type 2 diabetes mellitus. Additionally, we noted that in the current study, 62 patients (82.67% of patients) had hypertriglyceridemia, and 45 patients (60% of patients) had hypercholesterolemia. With many asymptomatic individuals and a slower rate of progression to liver fibrosis or cirrhosis, NAFLD is thought to be a milder form of the illness than alcohol-induced liver disease. [23,24]

Nearly all of the individuals in the current study either showed no symptoms at all or very minor symptoms. This bolsters the previously mentioned thought. According to reports, type 2 diabetes mellitus, obesity, and hypertriglyceridemia may all

be clinically connected with insulin resistance, which is linked to NAFLD. It has been documented that even among NAFLD patients who are slim and have normal glucose tolerance, there is insulin resistance and hyperinsulinemia.

It has been proposed that the onset of insulin resistance may be caused by hereditary factors that raise triacylglycerol levels and decrease insulin sensitivity. The fact that all of the patients in our study are obese or overweight, nearly all of them have diabetes or glucose intolerance, and a sizable portion of them have high blood sugar and/or high cholesterol lends credence to the link between insulin resistance and non-alcoholic fatty liver disease (NAFLD).

The majority of research has demonstrated that patients with mild, ambiguous symptoms or those who are suspected of having NAFLD are detected during regular investigations.

According to reports, the majority of individuals have mildly aberrant liver function tests. [25] Serum liver enzyme activity (ALT and AST) have been reported to be slightly elevated in many cases; the most often observed anomaly is a minor elevation of ALT. According to certain accounts, when increased, ALT grows more than AST increases. In the current investigation, we found that NAFLD patients had statistically significant increases in their ALT levels. Nevertheless, the ALT results are only higher than the upper reference range value (50 units/L) set by our laboratory in 11% of patients.

Just 11% of the time do the AST values exceed the upper reference range value of 50 units/L, and there is no discernible difference between the AST values and the controls. Numerous writers have demonstrated that NAFLD patients have a markedly elevated ALT/AST ratio. [25] Nevertheless, research has demonstrated that patients with cirrhotic NAFLD lose this ratio's diagnostic accuracy. In the current investigation, we found that patient ALT/AST ratio values were significantly higher than control values.

A high true positive rate and a low false positive rate are indicative of good clinical performance, and it has been suggested that the entire ROC curve summarizes the analytical systems' clinical performance. Among the indicators investigated to suggest NAFLD, we observed in the current study that the ALT/AST ratio is a better diagnostic sign.

However, a study incorporating all other disorders to be taken into account in the differential diagnosis of non-alcoholic fatty liver disease (NAFLD) must be conducted before the clinical diagnostic usefulness of this test can be demonstrated.

There was no discernible change in the serum bilirubin levels in this investigation. Bilirubin levels were not altered in any of the analyzed studies. Multiple mechanisms may be involved in the slightly elevated hepatic iron storage and subsequent higher risk of fibrosis in NAFLD patients. Lipid perioxidation appears to be the most likely process at the moment, though. The majority of the moderate iron excess observed in NAFLD patients has been demonstrated to be caused by the cyst 282 Tyr mutations, which has a strong correlation with liver damage in these individuals. Lipid peroxidation can be directly caused by hepatic iron excess. [26]

In this investigation, we found that individuals with non-alcoholic fatty liver disease (NAFLD) had considerably higher serum uric acid levels. This suggests that oxidative stress is prevalent in the condition, which is known to advance slowly. Additionally, it shows that the daytime increases in circulating insulin have resulted in a decrease in the renal clearance of uric acid.

Conclusions

It indicates that NAFLD, a moderate condition that affects both men and women, is the hepatic manifestation of the insulin resistance syndrome. The most effective diagnostic sign for NAFLD is the ALT/AST ratio. There is a link between oxidative stress and NAFLD.

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