

Research Article

Study of Serum Magnesium, Sodium, Calcium, Phosphorus and Liver Enzyme Gamma-Glutamyl Transferase (GGT) Levels in Chronic Alcoholic Liver Disease

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ABSTRACT

Background: Chronic alcoholic liver disease (CALD) is associated with progressive hepatic injury, impaired synthetic and metabolic functions, and disturbances in mineral homeostasis. Alterations in serum magnesium, sodium, calcium, and phosphorus may occur during chronic alcohol consumption and liver disease, while elevated gamma-glutamyl transferase (GGT) is commonly associated with hepatobiliary injury and chronic alcohol exposure. The present study was undertaken to assess serum magnesium, sodium, calcium, phosphorus, and GGT levels in patients with chronic alcoholic liver disease and to compare them with healthy individuals.

Materials and Methods: This hospital-based comparative study was conducted in the Departments of Biochemistry and Medicine at Tezpur Medical College and Hospital, Tezpur, over a period of one year from December 2022 to December 2023. A total of 100 subjects were included, comprising 50 patients diagnosed with chronic alcoholic liver disease and 50 age- and sex-matched healthy controls. Patients aged 18-60 years with chronic liver disease associated with fatty liver or cirrhosis were included, while patients with non-alcoholic liver disease, autoimmune cirrhosis, malignancy, pregnancy, or those receiving mineral supplementation were excluded. Venous blood samples were collected under aseptic precautions, and serum was analyzed for magnesium, sodium, calcium, phosphorus, GGT, liver enzymes, bilirubin, total protein, albumin, creatinine, and glucose. Statistical comparison between the groups was performed using appropriate statistical tests.

Results: The mean serum magnesium level was significantly lower in cases than in controls (1.016 ± 0.286 vs. 1.946 ± 0.167 mg/dL; $p < 0.001$). Serum sodium was also significantly reduced in cases (129.54 ± 2.51 vs. 139.72 ± 3.05 mmol/L; $p < 0.001$). Similarly, serum calcium (7.274 ± 0.524 vs. 9.182 ± 0.654 mg/dL) and phosphorus (1.518 ± 0.479 vs. 3.60 ± 0.489) were significantly lower among cases than controls ($p < 0.001$). In contrast, serum GGT was markedly elevated in patients with CALD (585.06 ± 182.56 vs. 18.32 ± 6.59 U/L; $p < 0.001$). Cases also demonstrated significantly increased ALT, AST, ALP, bilirubin, creatinine, and glucose levels, along with significantly decreased total protein and albumin levels, compared with controls.

Conclusion: Patients with chronic alcoholic liver disease demonstrated significant disturbances in serum mineral homeostasis, characterized by hypomagnesemia, hyponatremia, hypocalcemia, and hypophosphatemia, along with markedly elevated GGT and other biochemical indicators of hepatic dysfunction. Assessment of serum mineral levels together with GGT and routine liver-function parameters may provide useful information regarding metabolic disturbances and hepatic dysfunction in patients with chronic alcoholic liver disease.

Keywords: Chronic Alcoholic Liver Disease, Magnesium, Sodium, Calcium, Phosphorus, Gamma-Glutamyl Transferase, GGT, Liver Function Tests, Mineral Imbalance.

INTRODUCTION

Chronic liver disease involves ongoing

inflammation, damage, and renewal of the liver tissue, eventually leading to scarring and

advanced scarring of the liver. Causes range from prolonged exposure to toxins and alcohol abuse to infections, autoimmune diseases, as well as hereditary and biochemical disorders.¹ Cellular process driving scarring and advanced scarring involves the activation of stellate cells and fibroblasts, following the development of fibrous tissue. At the same time, the regeneration of liver parenchyma is reliant on the function of hepatic stem cells.² The liver is the largest internal organ, weighing approximately 1–1.5 kg, and is located mainly in the right upper quadrant of the abdomen beneath the diaphragm. It receives a dual blood supply, with approximately 80% from the portal vein and 20% from the hepatic artery. Hepatocytes constitute the major cellular component of the liver and are responsible for essential functions including synthesis of plasma proteins, bile production, nutrient metabolism, and detoxification and excretion of drugs and other substances. The commonly used liver function tests include serum bilirubin, serum albumin, and prothrombin time, which primarily assess hepatic excretory and synthetic functions. Abnormalities in these parameters may indicate impaired liver function.^{3,4}

Magnesium is an important intracellular cation, with most of the body's magnesium stored in bone and cells. It acts as a cofactor for more than 300 enzymes and is essential for ATP-related reactions and neuromuscular function. Low serum magnesium can increase neuromuscular excitability because magnesium normally inhibits calcium entry into neurons.⁵ Calcium (Ca^{2+}) is the most abundant mineral in the body and is mainly stored in bones as hydroxyapatite, $\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$.⁶ In plasma, approximately 50% is present as free ionized calcium, 40% is bound to plasma proteins, mainly albumin, and 10% is complexed with small anions. Calcium is essential for bone mineralization and blood coagulation.⁵ Low serum albumin may decrease total serum calcium while maintaining normal ionized calcium, a condition referred to as pseudohypocalcemia.⁷ Phosphorus is an essential mineral found in the body mainly as organic and inorganic phosphate. It is important for cellular functions and is a major component of bone hydroxyapatite. In plasma, phosphate is present mainly as H_2PO_4^- and HPO_4^{2-} ions.⁸

Gamma-glutamyltransferase (GGT) is an enzyme involved in glutathione metabolism and amino acid transport. Serum GGT is commonly used as a marker of hepatobiliary disease and

may be elevated with chronic alcohol consumption. Increased GGT levels in alcohol use are mainly associated with hepatic enzyme induction and increased release of the enzyme into the bloodstream.^{9,10}

METHODOLOGY

Place of work

This clinical research was carried out in the Biochemistry Department in association with the Medicine Department at Tezpur Medical College and Hospital, Tezpur. This work has been sanctioned by the Ethics Review Board, Tezpur Medical College and Hospital, vide letter no. 141/2022/TMCH dated: 12.12.2022.

Period of Work: From 16.12.2022 to 15.12.2023

Source of Data: Study population comprised 50 subjects with diagnosed chronic alcoholic liver disease patients who sought health care at the Department of Medicine in Tezpur Medical College and Hospital and also the attendants who accompanied them.

Selection of Subjects: The subjects for the study are categorized into two broad groups – Control group (healthy individuals) and test group or experimental group.

Total Sample Size: 100 Subjects

Target Population for Study

Control Group

The control group of 50 consisting age and sex matched normal healthy individuals without any history of liver diseases. The subjects for the control group were selected randomly among persons of different sections of society belonging to different occupation and socio-economic status, co-operating voluntarily.

Sample Size of Control Group

Furthermore, informed consent for participation to the study was obtained from all subjects under the control group after explaining to them the importance of the tests, the aims and the anticipated results. A careful screening was carried out in selecting subjects so that persons having pathology referable to any system, particularly persons having history of diabetes mellitus, cardiac disease, renal disease, hypertension, tuberculosis either in the past or present was not included in this group. A thorough history (Personnel, occupational etc.) and physical examination was done to exclude all these possibilities.

Case/ Experimental Group

The experimental group consists of 50 patients who were diagnosed as chronic alcoholic liver

disease by Medicine department and meets both inclusionary as well as exclusionary criteria. Informed consent for participation in the study was obtained from the subjects/guardians of the patients after explaining to the significance of the tests, the aims of the study and the anticipated results.

Criteria Adopted For Selection of Subjects under Experimental Group

Certain inclusion and exclusion criteria were used to select the subjects of the experimental group for the study.

Inclusion Criteria

1. Patients aged 18–60 years.
2. Patients diagnosed with chronic liver disease (CLD).
3. Patients with CLD associated with fatty liver.
4. Patients with CLD progressing to cirrhosis.

Exclusion Criteria

1. Patients with CLD due to non-alcoholic causes.
2. Patients with non-alcoholic or autoimmune cirrhosis, cancer, or pregnancy.
3. Patients receiving magnesium, sodium, calcium, or phosphorus supplements.
4. Patients unwilling to participate in the study.

Scheme of Case Taking

A standardized case-recording proforma was used to collect relevant demographic, socioeconomic, clinical, and hospital-related information, including age, sex, occupation, socioeconomic status, marital status, hospital number, and date of admission. Particular attention was given to the following symptoms and their duration:

1. Abdominal fullness and pain
2. Loss of appetite
3. Yellowish discoloration of the eyes and skin
4. Dry mouth and increased thirst
5. Nausea and vomiting
6. Ankle swelling
7. History and duration of alcohol consumption, particularly 10–20 years

General Examination

Patients were examined for general appearance, pallor, icterus, tongue moisture, height, weight, body build, nutritional status, nail changes, respiratory rate, pulse rate, blood pressure, temperature, jugular venous pressure, and peripheral edema.

Systemic Examination

Respiratory System: Oral hygiene, chest shape and symmetry, tracheal position, and breath sounds were assessed by auscultation.

Abdominal Examination: Abdominal contour and girth, tenderness, fluid accumulation, palpable masses, and the size and consistency of the liver, spleen, and kidneys were assessed.

Cardiovascular System: Pulse, blood pressure, jugular venous pulsation, apex beat, and heart sounds (S1, S2, and S3) were examined.

Nervous System: Higher mental functions, cranial nerves, motor and sensory systems, and reflexes were evaluated.

Collection of Samples for Special Investigations

Materials Required

- 5 mL disposable syringe with hypodermic needle
- Spirit swab
- Sterile labelled collection vials
- Tourniquet

Procedure

Under strict aseptic precautions, approximately 5 mL of venous blood was collected and transferred into a properly labelled vial. The blood was allowed to clot, after which serum was separated by centrifugation at 3000 rpm for 3 minutes. Separated serum samples were analyzed immediately or stored at 2–8°C for a maximum of 24 hours when necessary.

Instruments Used

- REMI T8 centrifuge
- Fixed- and variable-volume micropipettes
- Photometer 505 semi-autoanalyzer

Investigations

A. Hematological Investigations:

Hematological parameters including total leukocyte count, differential leukocyte count, hemoglobin level, erythrocyte sedimentation rate, ABO and Rh blood grouping, bleeding time, and clotting time were assessed.

B. Serum Biochemical Investigations:

Random blood glucose, serum creatinine, and liver function tests were performed to assess metabolic and hepatic status.

Special Investigations: Serum magnesium, sodium, calcium, phosphorus, and gamma-glutamyl transferase (GGT) levels were

estimated.

Urine Examination: Urine was assessed for daily output, colour, sugar, albumin, bile pigments, ketone bodies, and microscopic abnormalities.

Stool Examination: Stool was examined for colour, consistency, blood, mucus, occult blood, and microscopic findings.

Radiological Examination: Ultrasonography of the whole abdomen was performed to evaluate hepatic and abdominal abnormalities.

Sample Storage

All samples were appropriately labelled and, when immediate analysis was not possible, stored at 2–8°C and processed within the specified storage period.

RESULT

Table: 1 Comparison Table Showing Statistics of Age of Subjects in the Studied Group

	CASE	Control	SIGNIFICANCE
Number	50	50	t-test = 0.240750129 p-value = 0.5107
Mean	44.98	44.08	
SD	7.1670	6.44	
SEM	1.01	0.91	
Median	46	44.5	
Range	31-56	26-58	

The mean age was comparable between cases (44.98 ± 7.17 years) and controls (44.08 ± 6.44 years). The difference was not statistically significant (p = 0.5107), indicating that the two

groups were well matched for age and that age was unlikely to act as a major confounding factor.

Table No: 2 Statistical Analysis of Age of Different Groups

Age group (Years)	Case Group (n=50)			Control Group (n=50)		
	FREQ	%	REL FREQ	FREQ	%	REL FREQ
21-30	1	2	0.02	0	0	0
31-40	16	32	0.32	16	32	0.32
41-50	20	40	0.4	24	48	0.48
51-60	13	26	0.26	10	20	0.2

The majority of cases were aged 31–50 years, with 32% in the 31–40-year group and 40% in the 41–50-year group. A similar age distribution was observed among controls, with 48% aged 41–50 years. Both groups had the lowest

representation in the 21–30-year age group. Overall, chronic alcoholic liver disease was more commonly observed among individuals aged 31–50 years.

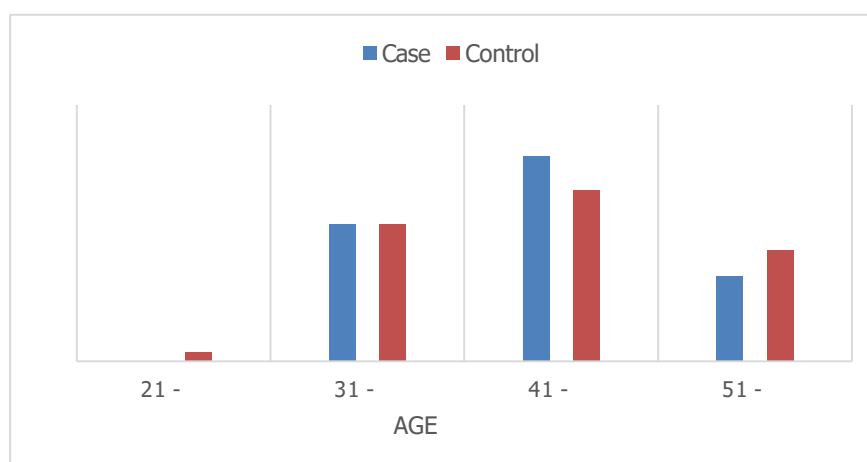


Figure: 1 Histogram Showing Age Distribution of Subjects

Table 3. Sex Distribution among Case and Control Groups (N = 50 Each)

Sex	Case Group, n (%)	Control Group, n (%)
Male	37 (74%)	39 (78%)
Female	13 (26%)	11 (22%)
Total	50 (100%)	50 (100%)

The case group showed a predominance of males (74%) compared with females (26%). This may reflect higher alcohol consumption

among males and indicates the need for targeted prevention and awareness efforts.

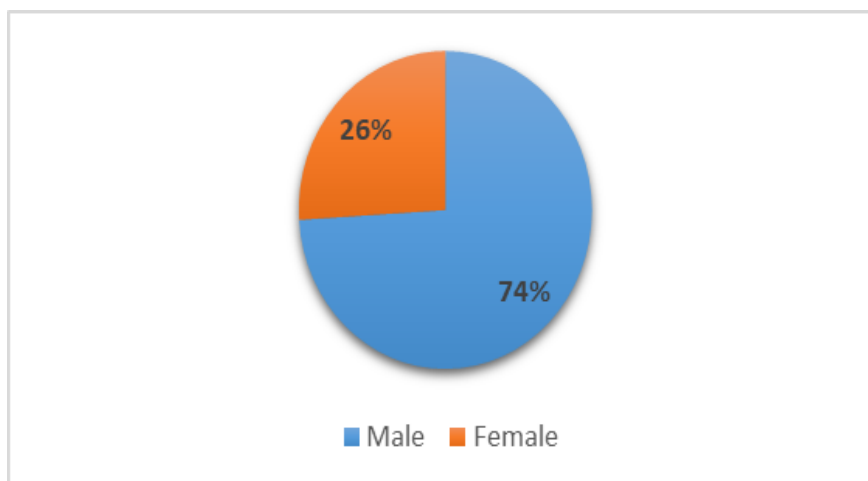


Figure: 2 Pie Diagram Showing Sex Distribution in Case Group

Table: 4 Comparison Table Showing Statistics of Magnesium of Subjects in the Studied

Magnesium	CASE	CONTROL
Mean	1.016	1.946
SD	0.285971313	0.166856
SEM	0.040442451	0.023597
Median	1	1.9
Range	0.4-1.5	1.7-2.3
t-test	8.91235E-27	

Serum magnesium levels were markedly lower in the case group (1.016 ± 0.286) than in controls (1.946 ± 0.167). The difference was highly statistically significant ($p < 0.001$),

indicating a strong association between chronic alcoholic liver disease and reduced serum magnesium levels.

Table: 5 Statistical Analysis of Age of Serum Magnesium Distribution of Different Groups

CASE GROUP			
C.I	No. of Subject	%	Rel. Frequency
0.1-0.6	6	12	0.12
0.7-1.2	32	64	0.64
1.3-1.8	12	24	0.24
1.9-2.4	0	0	0
CONTROL GROUP			
C.I	No. of Subject	%	Rel. Frequency
0.1-0.6	0	0	0
0.7-1.2	0	0	0
1.3-1.8	16	32	0.32
1.9-2.4	34	68	0.68

The case group predominantly showed lower serum magnesium levels, with 64% in the 0.7–

1.2 range and 12% in the 0.1–0.6 range. In contrast, 68% of controls had levels in the 1.9–

2.4 range. This marked difference indicates substantially lower serum magnesium levels

among patients with chronic alcoholic liver disease.

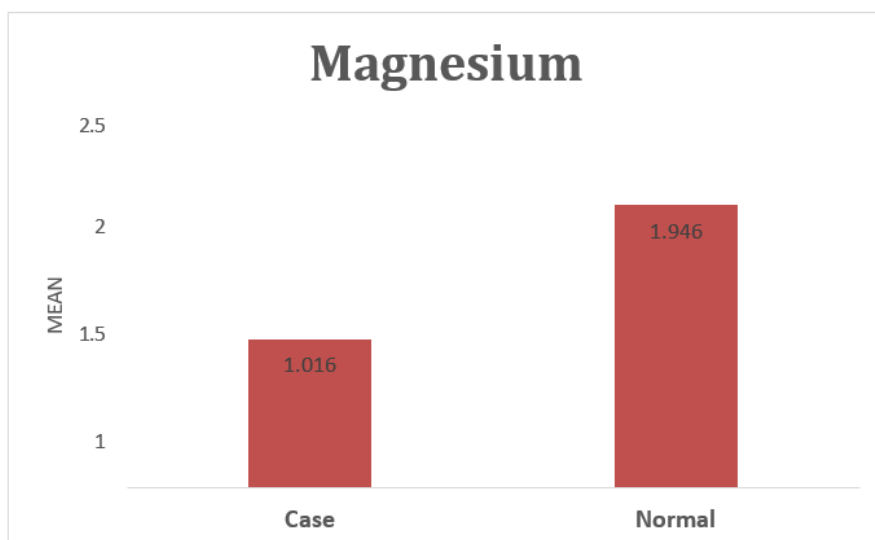


Figure: 3 Histogram for Mean Value of Magnesium in Case Group and Control Group

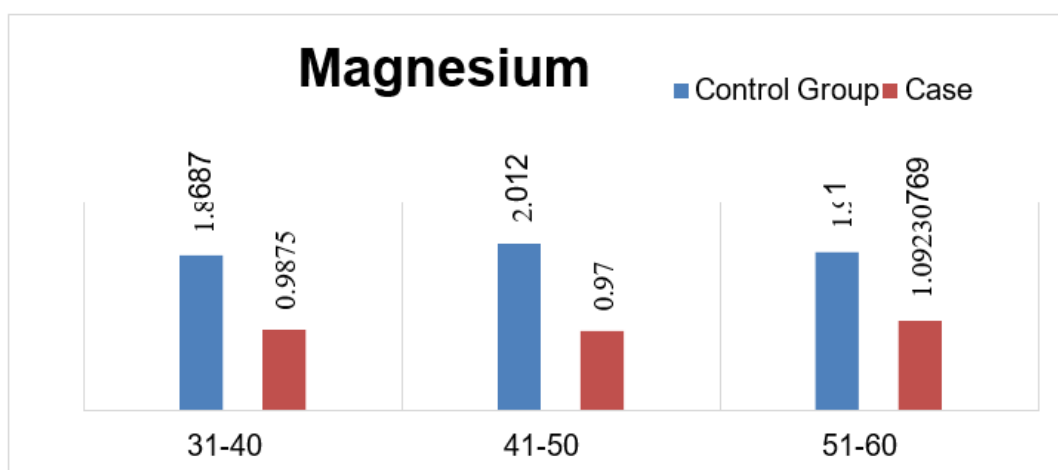


Figure: 4 Histogram Showing Distribution of Magnesium Mean In Different Age Intervals of Studies Group

Table: 6 Statistical Analysis of Serum Sodium in Case and Control Group

Sodium	CASE	CONTROL
Mean	129.54	139.72
SD	2.508841508	3.050861
SEM	0.354803769	0.431457
Median	130	140
Range	125-134	135-146
t-test	1.9214E-24	

Serum sodium levels were significantly lower in the case group (129.54 ± 2.51 mmol/L) than in controls (139.72 ± 3.05 mmol/L). The difference was highly statistically significant (p

< 0.001), indicating the presence of hyponatremia among patients with chronic alcoholic liver disease.

Table: 7 Distribution of Serum Sodium Levels in Case and Control Groups

Serum sodium range (mmol/L)	Case group n (%)	Control group n (%)
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125–130	34 (68%)	0 (0%)
131–136	16 (32%)	10 (20%)
137–142	0 (0%)	31 (62%)
143–148	0 (0%)	9 (18%)
Total	50 (100%)	50 (100%)

Most cases (68%) had serum sodium levels of 125–130 mmol/L, whereas most controls (62%) had levels of 137–142 mmol/L. This

distribution demonstrates a clear shift toward lower sodium levels among patients with chronic alcoholic liver disease.

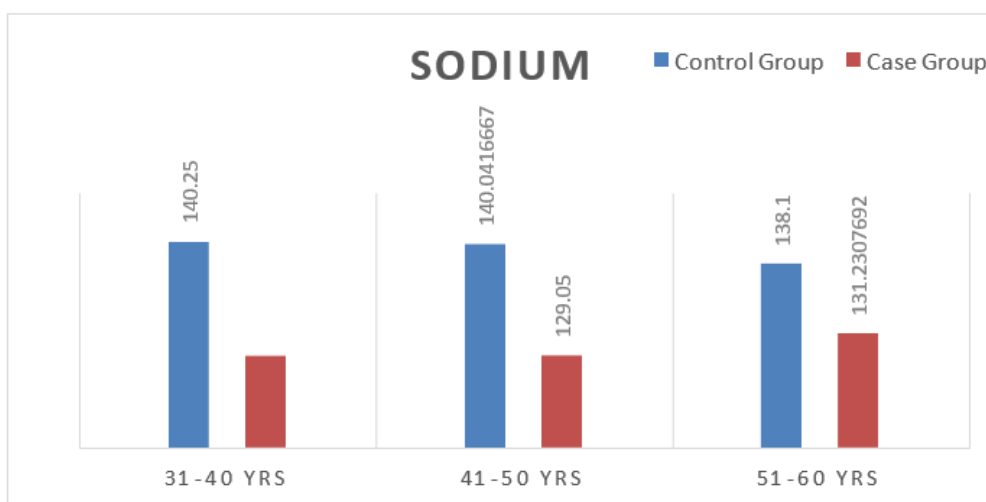


Figure: 5 Histogram Showing Distribution of Sodium Mean In Different Age Intervals of Studies Group

Figure 5.9 provides the age-wise distribution of serum Na levels, expressed as mean values, for both the case group (individuals with chronic alcoholic liver disease) and the control group. There are no participants in the 21-30 age range for either group. In the 31-40 age group, the control group has a mean magnesium level

of 140.25, whereas the case group shows a significantly lower mean of 129. This pattern persists in the 41-50 and 51-60 age groups, where the control group has mean magnesium levels of 140.04 and 138.1, respectively, compared to the case group's much lower levels of 129.05 and 131.23.

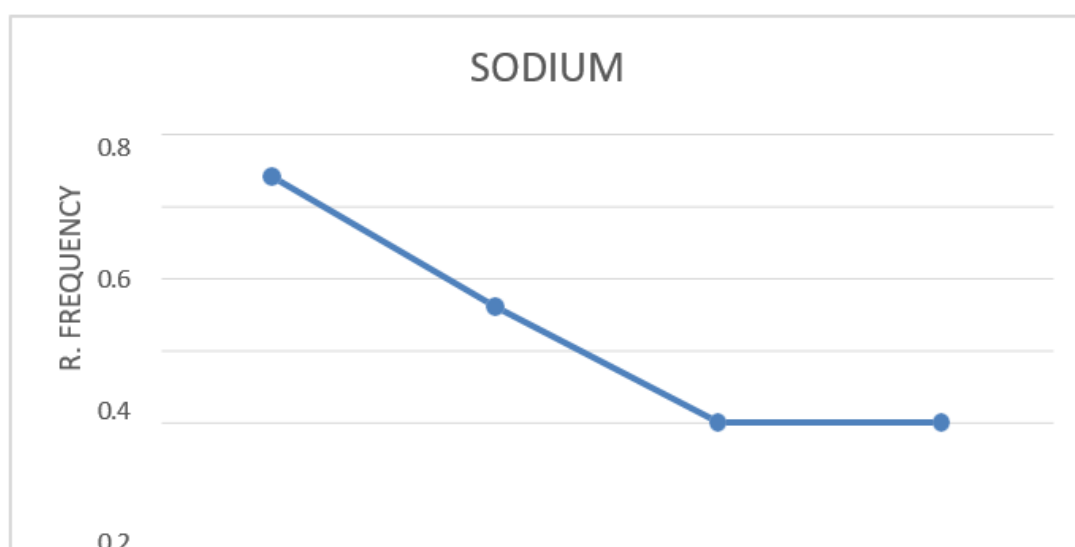


Figure: 6 Relative Frequency Curve of Serum Sodium of In Case Group

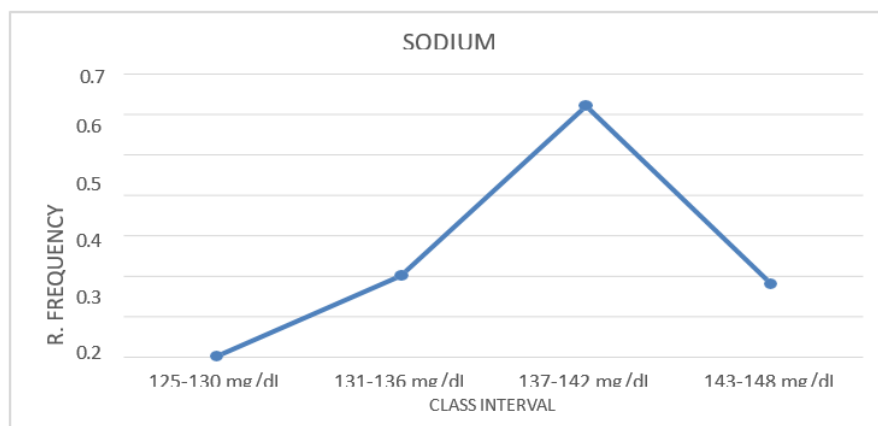


Figure: 7 Relative Frequency Curve of Serum Sodium of In Control Group

Table: 8 Statistical Analysis of Serum Calcium in Case and Control Groups

Calcium	CASE	CONTROL
Mean	7.274	9.182
SD	0.524427774	0.654245
SEM	0.074165287	0.092524
Median	7.2	9
Range	6.1-8.2	8-10.6
t-test	8.7715E-22	

Serum calcium levels were markedly lower in the case group (7.27 ± 0.52) compared with the control group (9.18 ± 0.65). The difference was highly statistically significant ($p < 0.001$),

indicating a significant reduction in serum calcium levels among patients with chronic alcoholic liver disease.

Table: 9 Statistical Analysis of Age of Serum Calcium Distribution of Different Groups

Serum calcium range	Case group n (%)	Control group n (%)
6.1–7.0	18 (36%)	0 (0%)
7.1–8.0	30 (60%)	1 (2%)
8.1–9.0	2 (4%)	25 (50%)
9.1–10.0	0 (0%)	17 (34%)
10.1–11.0	0 (0%)	7 (14%)
Total	50 (100%)	50 (100%)

Most cases (60%) had serum calcium levels of 7.1–8.0, while 50% of controls had levels of 8.1–9.0. Overall, calcium levels were

predominantly lower in the case group, indicating hypocalcemia among patients with chronic alcoholic liver disease.

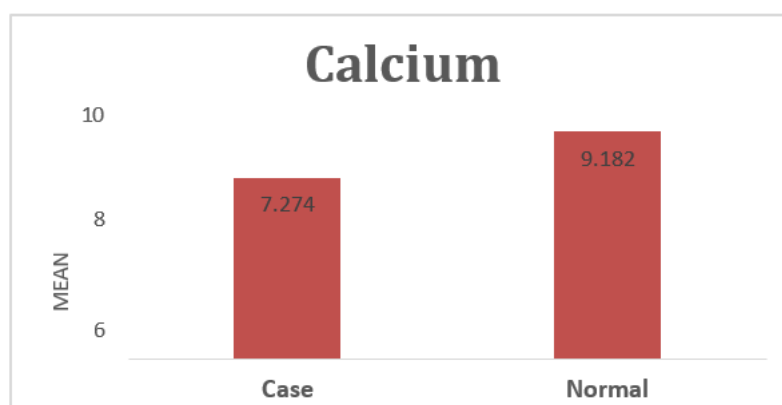


Figure: 8 Histogram for Mean Value of Calcium in Case Group and Control Group

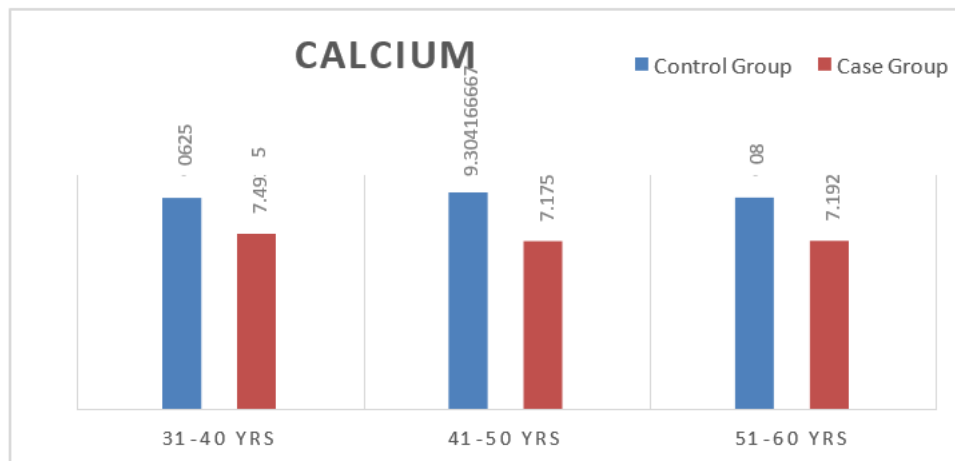


Figure: 9 Histogram Showing Distribution of Calcium Mean In Different Age Intervals of Studies Group

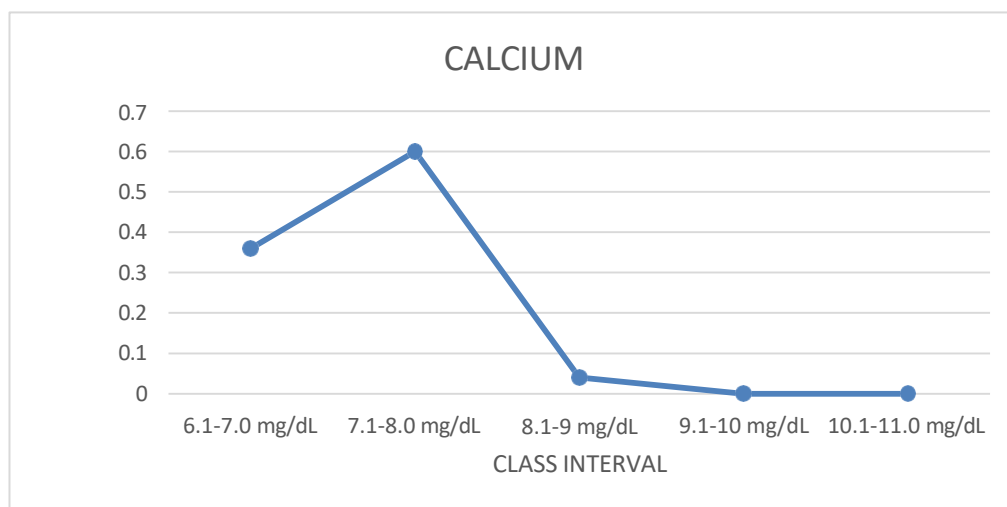


Figure: 10 Relative Frequency Curve of Serum Calcium of In Case Group

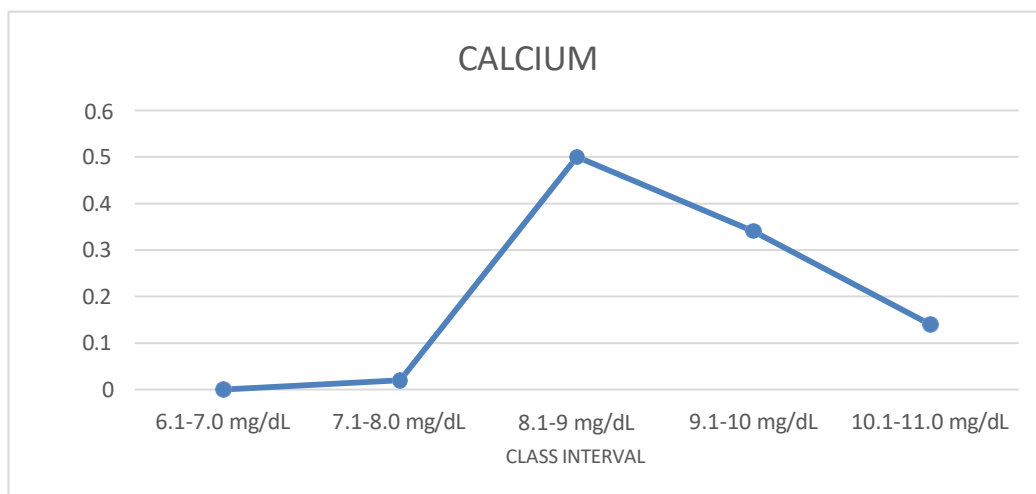


Figure: 11 Relative Frequency Curve of Serum Calcium of In Control Group

Table: 10 Statistical Analysis of Phosphorus in Case and Control Groups.

Phosphorus	CASE	CONTROL
Mean	1.518	3.6
SD	0.479238341	0.488647
SEM	0.067774536	0.069105
Median	1.6	3.65

Range	178-879	Feb-30
t-test	1.62E-26	

Table: 11 Statistical Parameters of Serum GGT in Case and Control Groups

Parameter	Case	Control
Mean (U/L)	585.06	18.32
SD	182.56	6.59
SEM	25.82	0.93
Median	589.5	19.5
Range (U/L)	34–1200	2–34
p-value	<0.001	

Serum GGT was markedly higher in cases than controls, with a highly significant difference ($p < 0.001$)

Table: 12 Comparison Table Showing Statistics of Magnesium, Sodium, Calcium, Phosphorus, GGT in Case Group and Control Group in the Studied Group

Biochemical Parameters	CASE GROUP					CONTROL GROUP					
	Mean	SD	SEM	Median	Range	Mean	SD	SEM	Median	Range	t-test
Magnesium	1.016	0.285971	0.040	1	1.1	1.946	0.166	0.023	1.9	0.6	8.91235E-27
Sodium	129.54	2.508842	0.354	130	9	139.72	3.050	0.431	140	11	1.9214E-24
Calcium	7.274	0.524428	0.074	7.2	2.1	9.182	0.654	0.092	9	2.6	8.7715E-22
Phosphorus	1.518	0.479238	0.067	1.6	1.6	3.6	0.488	0.069	3.65	1.6	1.62E-26
GGT	585.06	182.558	25.817	589.5	701	18.32	6.591	0.932	19.5	28	4.21831E-27

Table: 13 Comparison of Liver Enzymes in Case and Control Groups

Parameter	Case Mean $\pm$ SD	Control Mean $\pm$ SD	p-value
ALT (IU/L)	254.70 $\pm$ 78.75	24.66 $\pm$ 6.19	<0.0001
AST (IU/L)	216.10 $\pm$ 59.72	15.32 $\pm$ 3.84	<0.0001
ALP (IU/L)	476.92 $\pm$ 69.90	62.84 $\pm$ 20.52	<0.0001

ALT, AST, and ALP levels were significantly higher in the case group than in controls ($p < 0.0001$).

Table: 14 Comparison of Serum Bilirubin, Total Protein and Albumin

Parameter	Case Mean $\pm$ SD	Control Mean $\pm$ SD	p-value
Bilirubin (mg/dL)	1.14 $\pm$ 0.45	0.77 $\pm$ 0.34	<0.0001
Total Protein	5.26 $\pm$ 0.63	7.11 $\pm$ 0.65	<0.0001
Albumin	2.62 $\pm$ 0.37	4.30 $\pm$ 0.62	<0.0001

Cases showed significantly higher bilirubin and significantly lower total protein and albumin levels compared with controls.

Table: 15 Comparison of Serum Creatinine and Glucose

Parameter	Case Mean $\pm$ SD	Control Mean $\pm$ SD	p-value
Creatinine (mg/dL)	1.20 $\pm$ 0.37	0.93 $\pm$ 0.28	<0.0001

Glucose (mg/dL)	140.42 ± 38.85	103.58 ± 8.79	<0.0001
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Serum creatinine and glucose levels were significantly higher in the case group than in the

DISCUSSION

In the present study, patients with chronic alcoholic liver disease (CALD) showed significant alterations in serum biochemical and mineral parameters compared with healthy controls. Serum magnesium, sodium, calcium, phosphorus, total protein, and albumin were significantly lower, while GGT, ALT, AST, ALP, bilirubin, creatinine, and glucose were significantly higher, indicating disturbed mineral balance and impaired liver function. Serum magnesium was significantly reduced in CALD patients (1.026 ± 0.285 mg/dL) compared with controls (1.946 ± 0.166 mg/dL), consistent with Gaikwad et al. (2022)¹² and Turecky et al. (2006)^{75,11}. Serum sodium was also significantly lower in CALD patients (129.54 ± 2.51 mmol/L), supporting findings of Angeli et al. (2006)¹³ and Kim et al. (2009)¹⁴. Similarly, calcium (7.274 ± 0.524 mg/dL) and phosphorus (1.518 ± 0.479 mg/dL) were significantly reduced, consistent with Louboutin et al. (1980)¹⁵, Ionele et al. (2022)¹⁶, and Adler et al. (1990)¹⁷. GGT showed a marked increase in CALD patients (585.06 ± 182.55 U/L) compared with controls (18.32 ± 6.59 U/L), in agreement with Gandhi et al. (2014)¹⁸. Overall, these findings demonstrate significant mineral disturbances and abnormal liver-function parameters in CALD, particularly hypomagnesemia, hyponatremia, hypocalcemia, hypophosphatemia, and elevated GGT.

Conclusion: The present study demonstrates significant alterations in serum mineral and biochemical parameters among patients with chronic alcoholic liver disease compared with healthy controls. Serum magnesium, sodium, calcium, phosphorus, total protein, and albumin were significantly decreased, whereas GGT, ALT, AST, ALP, bilirubin, creatinine, and glucose were significantly increased. The marked reduction in magnesium, sodium, calcium, and phosphorus indicates disturbed mineral homeostasis, while elevated liver enzymes and GGT reflect hepatic injury and dysfunction. Overall, these findings suggest that assessment of serum minerals along with routine liver-function parameters may be useful in evaluating the severity and metabolic complications of chronic alcoholic liver disease.

Conflict of Interest: The authors declare that there is no conflict of interest regarding the

control group ($p < 0.0001$).

publication of this study.

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