Biochemical Evaluation of Patients of Alcoholic Liver Disease and Non-alcoholic Liver Disease

Prasad P. Torkadi · I. C. Apte · A. K. Bhute

Received: 27 December 2012 / Accepted: 5 February 2013 / Published online: 16 February 2013
© Association of Clinical Biochemists of India 2013

Abstract Alcoholic liver disease (ALD) is due to excessive alcohol intake for long duration. Distinguishing ALD from non-ALD (non-alcoholic steatohepatitis, hepatitis of viral origin) is difficult as patient may deny alcohol abuse. Clinical examination, histology and serology may not differentiate these conditions. Accurate diagnosis is important as management of ALD differs from non-ALD patients. The aim of our study was (1) To evaluate the patients of ALD and non-ALD by biochemical parameters compared to controls, (2) To assess whether these parameters can differentiate ALD from non-ALD. Study was carried out on 50 patients of ALD in group I and 35 patients of NASH (non-alcoholic steatohepatitis) and acute viral hepatitis each in group II. Age matched healthy controls $n = 50$. Selection criteria—history of alcohol intake (amount and duration), clinical examination, sonography of abdomen, serum alanine transaminase (ALT) and bilirubin levels. Blood samples were analyzed for bilirubin, aspartate transaminase (AST), ALT, alkaline phosphatase (ALP), gamma glutamyl transferase (GGT) by kinetic method. Statistical analysis was done by Student unpaired ‘ t ’ test. Patients of ALD have raised AST/ALT ratio (De Ritis ratio) (>2), ALP and GGT compared to controls ($P < 0.01$). There is significant difference in AST/ALT ratio, serum GGT and ALP in ALD group compared to that in NASH and acute viral hepatitis ($P < 0.05$). This study suggests that De Ritis ratio >2 in ALD patients may be due to alcohol induced hepatic

mitochondrial injury and pyridoxine deficiency. High GGT and ALP values may indicate enzyme induction by alcohol and mild cholestasis. Thus ALD patients have severe hepatic damage. De Ritis ratio <1 and normal to mild elevation in GGT level in NASH and acute viral hepatitis suggest mild hepatic injury of non-alcoholic origin. Our study concludes that ALD patients can be differentiated from NASH and acute viral hepatitis with certainty by measuring serum AST/ALT ratio, GGT and ALP. These biochemical parameters may help clinicians to support the diagnosis of ALD and non-ALD.

Keywords Alcoholic liver disease · Aspartate transaminase · Alanine transaminase · Gamma glutamyl transpeptidase

Introduction

Alcoholism is worldwide social and medical problem. Over past few years, alcohol consumption has increased in quantity and frequency. The age at which people start drinking has also declined. The population at risk is those undergoing rapid socio-economic and cultural changes. With the development of economy and the improvement in living standards of people, the incidences of diseases caused by alcohol abuse have been increasing.

Alcoholic liver disease (ALD) remains one of the most common causes of chronic liver disease. ALD has a known cause but a complex process. The amount and duration of ethanol ingestion rather than the type of alcoholic beverage or the pattern of ingestion, appears to be important determinant of liver injury. A substantial proportion of adult population in India uses alcohol. Globally 5 million people die per year due to alcohol related problems [1]. Seventy percent of deaths are due to liver related disease. About

P. P. Torkadi (✉) · A. K. Bhute
Department of Biochemistry, Dr. P.D.M. Medical College,
Amravati 444603, Maharashtra, India
e-mail: drptorkadi@gmail.com

I. C. Apte
Department of Biochemistry, Indira Gandhi Govt. Medical
College, Nagpur, Maharashtra, India

half of all cirrhosis in the world are alcohol induced and about 10–20 % of all alcoholics are cirrhotic. Alcoholic cirrhosis has 5 year mortality of 77 % and at 10 years, survival in only 7 %. In men, ethanol intake of 40–80 g/day produces fatty liver and that of 160 g/day for 10–20 years causes hepatitis or cirrhosis. Only 15 % of alcoholics develop ALD. The threshold for developing ALD in men is an intake of >60–80 g/day of alcohol for 10 years. Social, nutritional, immunologic and host factors play important role in development of pathogenic process.

In recent years, conventional biochemical markers and potential ones have aroused the interest of researchers to study the damages caused by ethanol in liver. Alanine transaminase (ALT) and aspartate transaminase (AST) are predominant ones in aminotransferases reflecting the damage to hepatocytes. Physicians have long sought an accurate and inexpensive means of identifying persons who consume excessive amounts of ethyl alcohol. Chronic alcoholism is diagnosed on the basis of clinical history, questionnaire about alcohol consumption and a number of laboratory investigations.

NASH is associated with raised levels of plasma free fatty acids resulting from adipose tissue or from hydrolysis of lipoprotein TG by LPL in extrahepatic tissues. It leads to fatty liver. Metabolic block in production of plasma lipoproteins and deficiency of lipotropic factors like choline can cause fatty liver. Increased TG in liver is seen in starvation, high fat diet, uncontrolled diabetes mellitus, pregnancy, ketoacidosis causing hepatic dysfunction.

Distinguishing non-ALD from ALD has generously been difficult. The history can be unreliable as many patients may deny alcohol abuse. Furthermore histology may not distinguish non-ALD from ALD with certainty. In hepatocyte, ALT is exclusively found in cytosol while different isoenzymes of AST exist in mitochondria and cytosol [2, 3]. About 80 % of AST activity in human liver is contributed by mitochondrial isoenzyme, whereas most of the circulating AST activity in normal people is derived from cytosolic enzyme [4, 5]. This mitochondrial AST is elevated in liver diseases with prolonged duration [6]. In alcoholic hepatitis AST/ALT ratio may be >2 [7–11]. AST/ALT ratio is typically <1 in other causes of fatty liver [8, 12, 13]. When AST/ALT ratio is >1.5, it is considered as highly suggestive that alcohol is the cause of liver injury [14]. Patients with alcoholic hepatitis often demonstrate an AST to ALT ratio of >2 with absolute aminotransferase levels of <300 IU/l. Although some authors have stated that no consistent relationship exists between serum AST and ALT levels in non-alcoholic steatohepatitis, others have implied that ALT level is generally greater than AST level [12, 15].

Activity of alkaline phosphatase (ALP) arises from bone, intestine, liver and placenta [16]. In the absence of

bone disease or pregnancy, elevated levels of ALP activity usually reflect impaired biliary tract function. Slight to moderate increases in ALP occur in many patients with liver disorders such as hepatitis and cirrhosis [17–20]. Striking increase in ALP (>10 times) occur more consistently with extrahepatic biliary tract obstruction or with intra hepatic cholestasis.

Measurement of gamma glutamyl transferase (GGT) activity in serum has been found useful in screening alcohol abuse. A sudden rise in serum GGT activity in known alcoholics is suggestive of a recent bout of alcohol [10, 21]. Elevated GGT associated with an increase in serum ALP shows that ALP in serum is of hepatic origin, as GGT is not raised in bone diseases [22].

No study however, has formally assessed the utility of serum AST/ALT ratio, GGT and ALP to differentiate alcoholic from non-ALD.

Aims and Objectives

- (1) To evaluate the patients of ALD and non-ALD by biochemical parameters compared to controls.
- (2) To assess whether these parameters can differentiate the patients of ALD from non-ALD.

Materials and Methods

The present study was carried out in the Department of Biochemistry. To evaluate biochemically, male patients with presumptive diagnosis of ALD ($n = 50$) and non-ALD ($n = 35$ each of NASH and acute viral hepatitis) admitted in medicine ward were included in the study. All the patients in our study were male. Most of the patients in this hospital are from rural areas with orthodox beliefs where drinking of alcohol by a female is not socially acceptable. Hence number of females with ALDs reporting to hospital is negligible owing to social stigma attached to alcoholism. Age matched 50 healthy male individuals in the age group of 18–65 years were selected as controls.

Patients of ALD in the age group of 18–65 years were diagnosed on the basis of history of alcohol intake (amount and duration), clinical examination and sonography of abdomen, with raised serum ALT and serum bilirubin. None of these were suffering from NASH, viral hepatitis, heart or kidney disease, diabetes mellitus, hepatic tumors. They had history of alcohol intake for >10 years with average daily drinking of >60 g of alcohol. Most of them were having country liquor. They were on abstinence since hospitalization.

Patients of NASH and acute viral hepatitis in the age group of 18–65 years were diagnosed on the basis of history, clinical examination, viral antigen/antibody study and

sonography of abdomen, with raised serum ALT and serum bilirubin. Patients with history of alcoholism, heart or kidney disease and hepatic tumors were excluded from the study.

Serum samples from the patients were analyzed for total bilirubin (normal 0.1–1.2 mg%) and direct bilirubin (0–0.3 mg%) by Diazo method, AST (normal up to 40 IU/l), ALT (normal up to 40 IU/l), ALP (normal 25–90 IU/l) and GGT (normal 0–45 IU/l) by kinetic method in clinical biochemistry laboratory using autoanalyzer (Erba Excel-300) and semiautoanalyzer (Erba chem 5-plus). Parametric data was analyzed by Student unpaired 't' test. A 'P' value of <0.05 was considered to be statistically significant. $P < 0.01$ was taken as highly significant.

Results

Data regarding biochemical evaluation of patients of ALD and non-ALD (NASH and acute viral hepatitis) are presented below in Tables 1, 2 and 3. Comparison between the groups is shown in Tables 4 and 5.

Table 1 Biochemical evaluation of patients of ALD (group I)

Parameters	Control ($n = 50$) (mean $\pm$ SD)	ALD ($n = 50$) (mean $\pm$ SD)	<i>P</i> value
Bil. (T) (mg%)	1.14 $\pm$ 0.219	3.94 $\pm$ 2.88	<0.01, S
Bil. (D) (mg%)	0.506 $\pm$ 0.17	2.01 $\pm$ 1.86	<0.01, S
ALT (IU/l)	24.22 $\pm$ 7.31	49.32 $\pm$ 20.99	<0.01, S
AST (IU/l)	23.78 $\pm$ 5.56	125.58 $\pm$ 57	<0.01, S
ALP (IU/l)	49.68 $\pm$ 13.48	117.52 $\pm$ 43.93	<0.01, S
GGT (IU/l)	24 $\pm$ 7.16	55.12 $\pm$ 17.02	<0.01, S
AST/ALT ratio	1.04 $\pm$ 0.32	2.54 $\pm$ 0.77	<0.01, S

SD standard deviation

$P < 0.01$: highly significant

Table 2 Biochemical evaluation of patients of NASH (group IIA)

Parameters	Control ($n = 50$) (mean $\pm$ SD)	NASH ($n = 35$) (mean $\pm$ SD)	<i>P</i> value
Bil. (T) (mg%)	1.14 $\pm$ 0.21	2.56 $\pm$ 0.71	<0.05, S
Bil. (D) (mg%)	0.50 $\pm$ 0.17	1.47 $\pm$ 0.57	<0.05, S
ALT (IU/l)	24.22 $\pm$ 7.31	60.28 $\pm$ 14.13	<0.01, S
AST (IU/l)	23.78 $\pm$ 5.56	53.12 $\pm$ 17.40	<0.01, S
ALP (IU/l)	49.68 $\pm$ 13.48	96 $\pm$ 25.06	<0.01, S
GGT (IU/l)	24 $\pm$ 7.16	34.12 $\pm$ 5.89	<0.01, S
AST/ALT ratio	1.04 $\pm$ 0.32	0.88 $\pm$ 0.17	NS

NS non-significant

$P < 0.05$, S: statistically significant; $P < 0.01$: highly significant

Discussion

Alcoholism is diagnosed on the basis of clinical history, questionnaire about alcohol consumption and many laboratory investigations. Distinguishing ALD from non-ALD has important implications for treatment and management. But many times it becomes difficult, as history can be unreliable. So we evaluated the patients of ALD, NASH

Table 3 Biochemical evaluation of patients of acute viral hepatitis (group IIB)

Parameters	Control ($n = 50$) (mean $\pm$ SD)	Acute viral hepatitis ($n = 35$) (mean $\pm$ SD)	<i>P</i> value
Bil. (T) (mg%)	1.14 $\pm$ 0.21	5.796 $\pm$ 6.1	<0.01, S
Bil. (D) (mg%)	0.50 $\pm$ 0.17	3.38 $\pm$ 4.68	<0.01, S
ALT (IU/l)	24.22 $\pm$ 7.31	401.32 $\pm$ 379.13	<0.01, S
AST (IU/l)	23.78 $\pm$ 5.56	363.44 $\pm$ 366.6	<0.01, S
ALP (IU/l)	49.68 $\pm$ 13.48	207.4 $\pm$ 166.98	<0.01, S
GGT (IU/l)	24 $\pm$ 7.16	45.36 $\pm$ 25.18	<0.01, S
AST/ALT ratio	1.04 $\pm$ 0.32	0.83 $\pm$ 0.29	NS

Table 4 Comparison between patients of groups I (ALD) and IIA (NASH)

Parameters	ALD ($n = 50$) (mean $\pm$ SD)	NASH ($n = 35$) (mean $\pm$ SD)	<i>P</i> value
Bil. (T) (mg%)	3.94 $\pm$ 2.88	2.56 $\pm$ 0.715	NS
Bil. (D) (mg%)	2.01 $\pm$ 1.86	1.47 $\pm$ 0.578	NS
ALT (IU/l)	49.32 $\pm$ 20.99	60.28 $\pm$ 14.13	<0.01, S
AST (IU/l)	125.58 $\pm$ 57	53.12 $\pm$ 17.40	<0.01, S
ALP (IU/l)	117.52 $\pm$ 43.93	96 $\pm$ 25.06	<0.01, S
GGT (IU/l)	55.12 $\pm$ 17.02	34.12 $\pm$ 5.89	<0.01, S
AST/ALT ratio	2.5462 $\pm$ 0.77	0.88 $\pm$ 0.176	<0.05, S

Table 5 Comparison of patients of groups I (ALD) and IIB (acute viral hepatitis)

Parameters	ALD ($n = 50$) (mean $\pm$ SD)	Acute viral hepatitis ($n = 35$) (mean $\pm$ SD)	<i>P</i> value
Bil. (T) (mg%)	3.94 $\pm$ 2.88	5.79 $\pm$ 6.1	NS
Bil. (D) (mg%)	2.01 $\pm$ 1.86	3.38 $\pm$ 4.68	NS
ALT (IU/l)	49.32 $\pm$ 20.99	401.32 $\pm$ 379.13	<0.01, S
AST (IU/l)	125.58 $\pm$ 57	363.44 $\pm$ 366.63	<0.01, S
ALP (IU/l)	117.52 $\pm$ 43.93	207.4 $\pm$ 166.98	<0.01, S
GGT (IU/l)	55.12 $\pm$ 17.02	45.36 $\pm$ 25.18	<0.05, S
AST/ALT ratio	2.546 $\pm$ 0.77	0.83 $\pm$ 0.29	<0.05, S

and acute viral hepatitis by various biochemical laboratory parameters.

Table 1 shows higher levels of serum bilirubin (total [T] and direct [D]), ALT, AST, ALP and GGT in ALD patients compared to controls and it is statistically significant ($P < 0.01$). It also shows AST/ALT ratio of 2.54 ± 0.77 in ALD patients which is higher (>2) compared to 1.04 ± 0.32 in controls and this difference is statistically significant ($P < 0.01$). Biochemical evaluation of patients of ALD revealed that they had severe hepatic damage compared to control.

Table 2 shows raised levels of serum bilirubin (T) and (D), ALT, AST, ALP and GGT in NASH patients compared to controls and it is statistically significant ($P < 0.01$).

Table 2 also shows slight difference in AST/ALT ratio in NASH patients and control which is statistically non-significant ($P > 0.05$).

Table 3 shows raised levels of serum bilirubin (total and direct), ALT, AST, ALP and GGT in acute viral hepatitis patients compared to controls and these are statistically significant ($P < 0.01$). But there is minimal difference in AST/ALT ratios of these two groups, which is statistically non-significant ($P > 0.05$).

Biochemical evaluation of patients of NASH and acute viral hepatitis revealed mild hepatic injury (AST/ALT ratio <1 , normal to minimal rise in serum ALP and GGT) in them compared to controls. Some patients of acute viral hepatitis showed raised ALP which may be due to cholestasis.

In ALD AST/ALT ratio may be >2 [7, 9, 10]. This appears to be the result of reduction in hepatic ALT content due to a deficiency in the cofactor pyridoxine-5- PO_4 . AST/ALT ratio is typically <1 in other causes of steatohepatitis [8, 12, 13]. An elevated serum AST in relation to serum ALT has been proposed as an indicator that alcohol has induced organ damage. When AST/ALT ratio is >1.5 , it is considered as highly suggestive that alcohol is the cause of liver injury [14].

We compared the patients of ALD and NASH by various biochemical parameters in Table 4. We observed a rise in AST/ALT ratio in ALD compared to NASH with $P < 0.05$. Transaminase levels in ALD patients were AST dominant while those in NASH patients were ALT dominant.

One study [23] observed AST/ALT ratio of 0.9 in NASH patients and 2.6 in ALD patients. Aminotransferase levels were significantly different in two groups ($P < 0.05$). They observed AST/ALT ratio of 0.7, 0.9 and 1.4 in NASH patients with no fibrosis, mild fibrosis and cirrhosis respectively ($P < 0.05$). They concluded that a ratio of <1 suggests NASH and >2 suggests ALD [11, 23, 24]. The rise in AST/ALT ratio in ALD patients compared to NASH patients in our study may be due to pyridoxine-5- PO_4 deficiency, decreased hepatic

ALT activity and alcohol induced mitochondrial damage leading to release of mitochondrial AST in serum [25].

In our study (Table 4) we found a rise in serum ALP and GGT in ALD patients compared to NASH patients. This rise is statistically significant ($P < 0.01$). Elevated GGT associated with an increase in serum ALP shows that ALP in serum is of hepatic origin, as GGT is not raised in bone diseases [22]. The rise in serum GGT in alcoholic patients may be due to hepatic microsomal enzyme induction by alcohol [18, 21, 26–28].

This comparison in our study provides information that the patients of ALD can be differentiated from NASH patients based on biochemical parameters like AST/ALT ratio, GGT and ALP.

Many times alcoholic patients present with clinical presentation similar to that of viral hepatitis. Also history of alcoholism may not be reliable. Antigens and antibodies against viruses may not be detectable in early cases. These conditions need to be distinguished at the earliest to have proper diagnosis and management. Focusing this need we compared the patients of ALD and acute viral hepatitis by biochemical parameters in Table 5.

We observed (Table 5) a significant and proportionate rise in serum ALT and AST in acute viral hepatitis patients compared to that in ALD patients ($P < 0.01$). AST/ALT ratio in acute viral hepatitis patients is 0.83 ± 0.29 whereas that in ALD patients is 2.54 ± 0.77 ($P < 0.05$).

AST rise is less than ALT in liver diseases including viral hepatitis except ALD where AST rise is 2–3 times more than ALT [8, 9, 11, 29].

Slight to moderate increase in ALP occurs in many patients with liver disorders such as viral hepatitis and cirrhosis [17–19, 30]. We observed (Table 5) a significant rise in serum ALP in viral hepatitis patients compared to that in ALD patients ($P < 0.01$). Rise in serum ALP is a better indicator of cholestasis than GGT [31]. There is a rise in serum GGT level in ALD patients compared to that in acute viral hepatitis patients ($P < 0.05$). Raised GGT in ALD patients may be due to alcohol induced enzyme release [10, 18, 21, 26, 32].

The results in our study suggested that we can differentiate patients of ALD from those of acute viral hepatitis based on parameters like AST/ALT ratio, ALP and GGT.

This study concluded that ALD patients can be differentiated from patients of NASH and acute viral hepatitis by measuring serum AST/ALT ratio, GGT and ALP. Our study suggests that these biochemical parameters may be used prior to invasive and expensive investigations like biopsy and sonography to support the clinical diagnosis. So that early diagnosis and treatment can be done to prevent the further complications. Overall, there are poor chances of recovery in patients of ALD. Timely intervention in the form of alcohol

abstinence and supportive treatment should be emphasized to halt the disease process.

Patients of NASH and acute viral hepatitis can be said to have fair prognosis with much less mortality after proper diagnosis and treatment.

Limitation of our study is that we evaluated very few biochemical parameters. Moreover these parameters may lack sensitivity when used singly. Further studies in this regard, using other biochemical parameters to differentiate ALD from non-ALD with certainty, are undoubtedly warranted.

References

- Shah SN. Alcoholic liver disease. In: API textbook of medicine. 7th ed. New Delhi: Jaypee Brothers Medical Publishers; 2003. p. 606–15.
- Rosen HR, Keefe EB. Evaluation of abnormal liver enzymes, use of liver tests and serology of viral hepatitis. In: Liver disease, diagnosis and management. 1st ed. New York: Churchill Livingstone Publishers; 2000. p. 24–35.
- Sherlock S. Assessment of liver function. In: Disease of liver and biliary system. 10th ed. London: Blackwell; 1997. p. 17–32.
- Friedman SF, Martin P, Munoz JS. Laboratory evaluation of the patients with liver disease. In: Hepatology, a text book of liver disease, vol 1. Philadelphia: Saunders Publication; 2003. p. 661–709.
- Boyde TRC, Lather AL. Starch gel electrophoresis of transaminases in human tissues extracts and serum. *Biochem J*. 1961;82:52–7.
- Nalplus B, Vassault A, Charpin S. Serum mitochondrial AST as a marker of chronic alcoholism: diagnostic value and interpretation in a liver unit. *Hepatology*. 1986;6:608–13.
- Wong F, Blendis L. Alcoholic liver disease. In: Thomson ABR, Shaffer EA, editors. First principles of gastroenterology: the basis of disease and an approach to management. 3rd ed. Edmonton: Canadian Association of Gastroenterology; 1997. p. 502–6.
- Bain VG, Ma M. Chronic hepatitis. In: Thomson ABR, Shaffer EA, editors. First principles of gastroenterology: the basis of disease and an approach to management. 3rd ed. Edmonton: Canadian Association of Gastroenterology; 1997. p. 492–501.
- Mittal A, Sathian B, Kumar A, Chandrasekharan N, Yadav SK. The significance of transaminases and De Ritis ratio for predicting alcoholic liver disease: a hospital based comparative study in western Nepal. *Nepal J Epidemiol*. 2010;1(1):33–7.
- Bruha R, Dvorak K, Petrtyl J. Alcoholic liver disease. *World J Hepatol*. 2012;4(3):81–90.
- Arthi M, Niranjana G, Hanifah M, Srinivasan AR. Efficacy of De Ritis ration in diagnosing liver diseases in Puducherry population. *Adv Lab Med Int*. 2011;1(4):61–8.
- Laffaine MG, Mauricio LM. Non-alcoholic fatty liver disease. *Ann Hepatol*. 2004;3(3):93–9.
- Obika M, Noguchi H. Diagnosis and evaluation of nonalcoholic fatty liver disease. *Exp Diabetes Res*. 2012;2012:12:article ID 145754.
- Correia JP, Alves PS, Camilo EA. SGOT–SGPT ratios. *Dig Dis Sci*. 1981;26(3):284.
- Barsić N, Lerotić I, Smirčić-Duvnjak L, Tomašić V, Duvnjak M. Overview and developments in noninvasive diagnosis of nonalcoholic fatty liver disease. *World J Gastroenterol*. 2012;18(30):3945–54.
- Hagerstrand I. Distribution of ALP activity in healthy and diseased human liver tissues. *Acta Pathol Microbiol Scand*. 1975;83: 519–24.
- Heathcote J. Cirrhosis of liver. In: Thomson ABR, Shaffer EA, editors. First principles of gastroenterology: the basis of disease and an approach to management. 3rd ed. Edmonton: Canadian Association of Gastroenterology; 1997. p. 520–4.
- Gowda S, Desai PB, Hull VV, Math AK, Vernekar SN, Kulkarni SS. A review on laboratory liver function tests. *Pan Afr Med J*. 2009;3(17):13.
- Kundu D, Roy A, Mandal T, Bandyopadhyay U, Ghosh E, Ray D. Oxidative stress in alcoholic and viral hepatitis. *N Am J Med Sci*. 2012;4(9):412–5.
- Dowman JK, Tomlinson JW, Newsome PN. Systematic review: the diagnosis and staging of non-alcoholic fatty liver disease and non-alcoholic steatohepatitis. *Aliment Pharmacol Ther*. 2011;33(5): 525–40.
- Jang ES, Jeong S-H, Hwang SH, Kim HY, Ahn SY, Lee J, et al. Effects of coffee, smoking, and alcohol on liver function tests: a comprehensive cross-sectional study. *BMC Gastroenterol*. 2012; 12(145):14.
- Rosalki SB, McIntyre N. Biochemical investigations in the management of liver disease. In: Oxford text book of clinical hepatology. 2nd ed. New York: Oxford University Press; 1999. p. 503–21.
- Sorbi D, Boynton J, Lindor KD. The ratio of AST to ALT: potential value in differentiating non-alcoholic steatohepatitis from alcoholic liver disease. *Am J Gastroenterol*. 1999;94(4):1018–22.
- Haukeland JW, Schreiner LT, Lorgen I, Frigstad SO, Bang C, Raknerud N, et al. ASAT/ALAT ratio provides prognostic information independently of Child–Pugh class, gender and age in non-alcoholic cirrhosis. *Scand J Gastroenterol*. 2008;43(10):1241–8.
- Nyblom H, Berggren U, Balldin J, Olsson R. High AST/ALT ratio may suggest advanced alcoholic liver disease rather than heavy drinking. *Alcohol Alcohol*. 2004;39(4):336–9.
- Selinger MJ, Matloff DS, Kaplan MM. GGTP activity in liver disease: serum elevation is independent of hepatic GGTP activity. *Clin Chim Acta*. 1982;125(3):283–90.
- Penn R, Worthington DJ. Is serum GGT a misleading test? *Br Med J*. 1983;286:531–5.
- Das SK, Nayak P, Vasudevan DM. Biochemical markers of alcohol consumption. *Indian J Clin Biochem*. 2003;18(2):111–8.
- Das SK, Vasudevan DM. Biochemical diagnosis of alcoholism. *Indian J Clin Biochem*. 2005;20(1):35–42.
- Thapa BR, Walia A. Liver function tests and their interpretation. *Indian J Pediatr*. 2007;74(7):663–71.
- Paulo B, Romeo CG. Indications of inflammation and cellular damage in chronic asymptomatic of oligosymptomatic alcoholics; correlation with alteration of bilirubin and hepatic and pancreatic enzymes. *Rev Hosp Clin Fac Med Sao Paulo*. 1999;54(2):53–60.
- Kevin W, Graemme A. Alcoholic liver disease. *Postgrad Med J*. 2000;76:280–6.