

## The value of liver function tests in hepatocellular carcinoma

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### Abstract

This study was undertaken to see if liver function tests (LFT) served a worthwhile purpose in the investigation of hepatocellular carcinoma (HCC). Sera from 80 HCC, 76 benign liver disease (BLD) and 152 healthy adult (HA) subjects were assayed for alkaline phosphatase (ALP), gamma-glutamyltransferase (GGT), aspartate aminotransferase (AST), alanine aminotransferase and lactate dehydrogenase, bilirubin and albumin. Cut-off values were determined from the HA. ALP, GGT, AST and albumin were abnormal in about 90% of the HCC. With the exception of bilirubin, the LFT were abnormal more frequently in HCC than in chronic hepatitis and cirrhosis, the conditions which precede it. Raised ALP in the presence of normal bilirubin was more often a feature of HCC than BLD although this relationship was not statistically significant. It seems unlikely that LFT serve a useful function in HCC.

**Key words:** Liver functions tests, hepatocellular carcinoma.

### INTRODUCTION

The battery of blood tests consisting of total bilirubin, total protein, albumin and the enzymes alkaline phosphatase (ALP), aspartate aminotransferase (AST), alanine aminotransferase (ALT) and gamma-glutamyltransferase (GGT), have become known as the standard 'liver function tests' (LFT). Despite the advent of more sophisticated techniques for the diagnosis of liver disease, particularly ultrasound and CT scanning, together with percutaneous and endoscopic cholangiographic and liver biopsy, LFT continue to be routinely requested by clinicians for all types of liver disease and appear destined to survive if not actually thrive in the clinical biochemistry environment.<sup>1</sup>

The literature reveals varying opinions on the value of LFT in the diagnosis of hepatocellular carcinoma (HCC).<sup>2,3</sup> We therefore undertook this study of the changes of LFT in HCC to find out if routine LFT served a worthwhile purpose in the investigation of HCC. Lactate dehydrogenase (LD) measurement was included to the above list of tests because moderate increases of it occur, *inter alia*, in any malignancy;<sup>4</sup> however, total protein measurement was omitted.

### SUBJECTS, MATERIALS AND METHODS

#### Subjects

Blood from 80 HCC, 76 benign liver diseases (BLD) and 152 healthy adult (HA) subjects was

used in this study. Confirmation of the diagnosis of HCC was by histopathological examination of biopsy samples in all but 2 cases where it was achieved by cytological examination of ascitic fluid. The numbers (in parentheses) of the different types of BLD patients were as follows: cirrhosis (32), hepatitis (24), obstructive jaundice due to gallstones (10), acute cholecystitis (4), amoebic liver abscess (2), and other miscellaneous liver diseases (4). The HCC and BLD patients were, in the main, from the Kuala Lumpur Hospital (KLH) and a private clinic specializing in liver diseases, while the HA were mostly from donors at the blood bank of KLH.

#### Methods and materials

All the LFT parameters in this study were measured on the automated chemistry analyzer, the Cobas Mira (Roche, Basel, Switzerland) at the Division of Biochemistry of the Institute for Medical Research (IMR). Reagent kits from Boehringer Mannheim (Mannheim, Germany) were used for the enzyme assays. The methods for bilirubin and albumin were that of Malloy and Evelyn<sup>5</sup> and the brom-cresol green dye-binding of Doumas *et al.*,<sup>6</sup> respectively, both of which were adapted for use on the Cobas.

#### Quality control

The inter-batch coefficients of variation of these tests ranged from 1.8% to 7.2%. The mean values for the analyses of each analyte using the commercially prepared assayed control sera

showed biases from the manufacturers' target values which ranged from -4.9% to 6.5%.

**Statistics**

The 97.5 percentile of the HA values determined formed the cut-off values or upper limits of normal (ULN) for ALP, GGT, AST, LD and bilirubin while the 2.5 percentile was used for that of albumin. The ALT cut-offs (for male and female) were based on the mean plus 2 standard deviations, since these were closer to the recommended cut-offs mentioned in the Boehringer Mannheim reagent package insert for ALT; the 97.5 percentiles were, by comparison, unusually high. The corrected chi-square test was applied to test the association of unpaired categorical variables; statistical significance was taken as  $P < 0.05$ .

**RESULTS AND DISCUSSION**

Cut-off values of the analytes are given in Table 1. The mean, median and ranges of the

LFT parameters for HCC subjects are given in Table 2 while the proportion of the HCC subjects who had abnormal results and comparative figures from the literature are given in Table 3.

**ALP and bilirubin**

Increased serum activity of ALP may result from cholestasis or the tumour tissue itself.<sup>4,7</sup> The mean and median ALP values in HCC were 2.6 and 2.2 times the ULN (i.e. 623 and 510 U/l), respectively. These were substantially higher than that of cirrhosis (1.4 and 1.2 times ULN) and chronic hepatitis (1.1 and 0.8 times ULN) patients. ALP was also more frequently raised in the HCC group than these (Table 4). However, patients with obstructive disease due to benign causes had the highest values, with the mean and median being 5.3 and 3.8 times ULN (1257 and 908 U/l), respectively. Among the various liver diseases, ALP was most frequently raised in benign obstructive disease with a sensitivity of 100%.

**TABLE 1: Determination of cut-off values using healthy adults**

LFT	n	Overall range	No. of outliers	Cut-off (ULN) <sup>a</sup>
ALP (U/l)	152	66-316	2	236
GGT (U/l)	152	7-215	4	68
AST (U/l)	130	11-87	2	62
ALT (U/l)	152	3-107	2	
Males				61
Females				33
LD (U/l)	151	115-548	1	423
Bilirubin (µmol/l)	152	0-44	1	27.6
Albumin (g/l)	152	37-55	-	39

<sup>a</sup> values above the cut-off were considered pathological for all LFT parameters except albumin where values below the cut-off were considered pathological.

**TABLE 2: Mean, median and range of LFT parameters in HCC subjects**

LFT	Cut-off (ULN) (from Table 1)	Mean	Median	Range
ALP (U/l)	236	623	510	133-2077
GGT (U/l)	68	264	196	17-1034
AST (U/l)	62	220	158	15.5-1407
ALT (U/l):				
Males	61	124	66	3.5-783
Females	33	58	54	15-114
LD (U/l)	423	834	565	238-4350
Bilirubin (µmol/l)	27.6	48	23	5-324
Albumin (g/l)	39	31	31	14.5-44

**TABLE 3: Comparison of percentage of abnormal results obtained in HCC in this study with that of other published results**

LFT	% Abnormal results				
	This study	Bersohn et al. (1969)	Kew et al. (1971)	Lai et al. (1981)	Kingston et al. (1985)
ALP	90	88	86	62	93
GGT	90				
AST	91	97	82		97
ALT		73			77
Males	56				
Females	87				
LD	70	97			
Bilirubin	45	46	66	44	
Albumin	89	58	64		

**TABLE 4: Percentage of abnormal results for various LFT parameters in HCC and liver conditions which precede it**

LFT	% Abnormal			
	HCC (from Table 3)	Chronic hepatitis	Cirrhosis	All BLD
ALP	90	40	63	61
GGT	90	30	75	61
AST	91	37	58	84
ALT				
Males	56	53	29	40
Females	87	50	46	54
LD	70	26	41	35
Bilirubin	45	20	44	47
Albumin	89	45	72	67

The finding that the average bilirubin level of the HCC patients was 1.7 times above the ULN and that only 45% had raised values indicates that marked bilirubinaemia was not a prominent feature of this condition, a conclusion that was also reached by Bersohn *et al.*<sup>2</sup>

It has been recognised that a raised ALP level in the absence of jaundice, may be used as possible presumptive evidence of HCC.<sup>1,2,8</sup> Of

the 78 HCC patients in this study on whom ALP and bilirubin measurements were made, 37/70 (52.9%) had a raised ALP and a normal bilirubin (<27.6  $\mu\text{mol/l}$ ). The corresponding figure for BLD was 17/46 (37.0%). The chi-square test, however, did not show a statistically significant association between raised bilirubin levels and HCC in liver disease patients with raised ALP (Table 5).

**TABLE 5: Relationship of bilirubin with HCC, in patients with liver diseases having raised ALP**

	HCC(+)	HCC(-) (BLD)	Total
Bilirubin (>27.6 $\mu\text{mol/l}$ )	33	29	62
Bilirubin (<27.6 $\mu\text{mol/l}$ )	37	17	54
Total	70	46	116

chi-square = 2.218,  $P = 0.136$  (not significant)

### GGT

Increased serum GGT activity in liver tumours may occur from cholestasis, or originate from reactive normal tissue adjacent to the tumour growth and the tumour tissue itself.<sup>7,9</sup> GGT was most often raised in HCC (90%) followed by benign obstructive liver disease (80%) and then by cirrhosis (75%). The average value obtained in benign obstructive disease (292 U/l) was, however, only marginally higher than that in HCC (264 U/l).

Rosalki<sup>9</sup> has described GGT as the best available screening test for liver disease in the non-jaundiced patient since its activity is increased in most hepatobiliary disease including primary and secondary liver tumours. However, because of the enzyme's wide tissue distribution and its elevation in BLD, its specificity for HCC is very low.

### AST, ALT and LD

Elevated plasma activities of AST or ALT are regarded as sensitive indicators of liver cell damage.<sup>10</sup> The mean value of AST in HCC (3.5 ULN) was higher than that obtained in chronic hepatitis (1.5 ULN) and cirrhosis (1.7 ULN). In acute hepatitis, however, the mean was 10.6 times the ULN. With the exception of acute hepatitis, AST was also more frequently raised in HCC than in the other BLD (Table 4). Mean ALT elevations in HCC were modest being 2.0 and 1.8 times ULN for males and females, respectively, compared with 1.1 and 1.4 times ULN in cirrhosis. Our results appear to bear out previous observations that although the aminotransferases are only moderately elevated in HCC, the AST values are much higher than those of ALT.<sup>3,11</sup>

In this study, LD values were generally higher in HCC than in BLD. However, on the average, the elevation was modest, being raised only twice above the ULN for HCC as against 1.2 times for BLD. It was also raised distinctly more often in cirrhosis than the other BLD (Table 4), including obstructive disease. Bersohn *et al.*,<sup>2</sup> however, showed in their study of African patients (Bantu), that LD was abnormal in 97% of HCC (Table 3), 98% of cirrhotics and 62% of non-liver cancer cases; this indicated that LD was of little value in distinguishing cirrhosis from HCC.

### Albumin

Serum albumin concentration is widely regarded as an index of hepatic 'synthetic function' and in

patients with liver disease a value below the lower limit of the reference range is taken to imply chronicity.<sup>1</sup> The mean value of 30.5 g/l obtained for these HCC patients was substantially lower than that of 46.6 g/l for the reference population. Although the former is comparable to the figure of 28.5 g/l obtained by Lai *et al.*,<sup>12</sup> both Götz<sup>3</sup> and Kew *et al.*<sup>13</sup> reported a substantially lower mean albumin concentration of 21 g/l for patients with primary liver carcinoma. In this study, albumin was also abnormal more frequently in HCC than in various BLD (Table 4), including obstructive disease.

### Overall discussion

Götz<sup>3</sup> has stated that LFT provide evidence for malignancy when they are considered together rather than individually and thought that the following pattern was indicative of liver malignancy. The aminotransferases are moderately elevated; LD activity is usually normal in patients with cirrhosis but increases in primary liver carcinoma and liver metastases. The activities of GGT and ALP thus often increase during the course of the disease. Bilirubin levels remain normal and only rise to values of about 85 µmol/l during the terminal stages of the disease. Albumin results are low.

Despite this observation, it is clear that LFT has little value, if at all, in HCC. Almost every HCC patient in this study had deranged LFT. As with earlier findings,<sup>2,13</sup> none of the tests were specific enough to be diagnostic of HCC and in the main, not distinct from those found in cirrhosis. Furthermore, it has been stated that damage has to be extensive before any appreciable change in LFT can be detected in HCC.<sup>2</sup>

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