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Original Article

Value of Serum Lentil Affinity of Alpha-fetoprotein in the Diagnosis of Hepatocellular Carcinoma on Top of Liver Cirrhosis

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ABSTARCT

Background: Hepatocellular carcinoma (HCC) is a common complication in patients with liver cirrhosis (LC). Detection of HCC at an early stage is critical for a favorable clinical outcome. AFP-L3% is an isoform of AFP which is very specific for HCC. The AFP-L3% is the percentage of AFP-L3 over the total AFP level. The study aimed to evaluate the utility of AFPL-3% in detection of HCC developing on top of liver cirrhosis, to compare the levels of both alpha- fetoprotein (AFP) and AFP-L3% in HCC versus LC patients without HCC and to define the cut-off level of each tumor marker with the best sensitivity and specificity for HCC detection.

Patients: The study was conducted on 25 cases of HCC that developed on top of LC and 25 LC cases with no evidence of HCC, as well as 25 apparently healthy controls. The levels of AFP and AFP-L3% were measured for all cases. Biochemical parameters and viral markers were also tested. Imaging and histopathological evidence of HCC were a prerequisite for inclusion in HCC group. Patients included in the HCC group had total AFP value < 200 ng/ml, which is not a diagnostic level for HCC.

Results: Levels of AFP and AFP L-3% were significantly higher in patients with HCC compared to those without HCC ($P < 0.01$). Receiver–operating characteristic (ROC) curve analysis indicated that the best cut-off value was 15.4% for AFP-L3% to detect HCC as the sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV) and accuracy were 79.2%, 100% , 100% , 83.3% and 96.2% respectively. For AFP, the best cut-off in the non diagnostic range was 77.8 ng/ml as the sensitivity, specificity, PPV, NPV, and accuracy were 75%, 68%, 69.2%, 73.9% and 70.4% respectively, The mean serum level of AFP showed no significant difference ($P>0.05$) regarding Child Pugh classification, numbers of tumor foci nor tumor size, however, it showed a significant difference ($P<0.05$) regarding lymph nodes invasion and TNM classification in HCC patients. Meanwhile, AFP-L3% showed no significant difference ($P>0.05$) regarding all these parameters. There was a positive significant correlation ($P<0.05$) between AFP and both AST and ALT, while AFP-L3% showed inverse significant correlation ($P<0.05$) with PC%. No significant correlation ($P>0.05$) was observed between serum AFP and serum AFP-L3% in HCC patients.

Conclusion: In patients with total AFP values <200 ng/ml who present a diagnostic dilemma, AFP-L3% had higher sensitivity, specificity, PPV, NPV and accuracy for HCC detection, and was not elevated in any of the patients without HCC with specificity of 100%.

Key words	Alpha-fetoprotein, hepatocellular carcinoma, liver cirrhosis, AFP-L3%, tumor marker.
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INTRODUCTION

Hepatocellular carcinoma (HCC) has become a major public health problem since its incidence is continuously growing and it has a high mortality rate[1]. It is the most common cause of primary liver neoplasm and is one of the main causes of death in patients with liver cirrhosis[2]. The annual risk to develop HCC in patients with liver cirrhosis is 5% (1–7%) [3]. Early detection of patients with HCC is important because it results in better prognosis[4].

Patients with advanced liver disease, particularly cirrhosis, should be screened every six months for HCC development[5]. Currently standard surveillance includes a combination of six monthly abdominal ultrsonography scan and serum AFP measurement. AFP is a specific glycoprotein produced primarily by the fetal liver. Normally, its serum concentration falls rapidly after birth and its synthesis in adult life is repressed. AFP has been the serum marker that is most widely used for diagnosis, surveillance and as a prognostic indicator of HCC survival[6]. Alpha fetoprotein levels ≥200 ng/ml in patients, with an identifiable liver mass by imaging techniques, are diagnostic of hepatocellular carcinoma[7].

Although AFP measurement serves as an important tool in screening HCC patients, some reports have indicated that because of its high false positive and false negative rates, it has limited utility in differentiating HCC from benign hepatic disorders. Patients with either cirrhosis or those with exacerbation of chronic hepatitis without HCC may have markedly increased AFP level[8]. Also, in 35-45% of HCC patients, AFP level may be normal particularly in patients with small HCC[9]. Even in patients with advanced HCC, AFP levels may remain normal in 15%-30% of the patients[10].

Total AFP can be divided into three different glycoforms,

namely AFP-L1, AFP-L2 and AFP-L3, according to their binding capability to lens culinaris agglutinin[11]. The lens culinaris agglutinin (LCA)-reactive alpha-fetoprotein percentage of total AFP concentration (AFP-L3/ total AFP x 100) or AFP-L3% has been used as a marker for early diagnosis, for assessment of therapeutic effects, and for predicting the prognosis of HCC[12]. Recent reports also suggest that it is useful for predicting the risk of development of HCC in patients with chronic liver disease[13]. AFP-L3% is considered more specific than AFP in diagnosis of HCC[14] and evaluation for recurrent tumor after treatment[15], as malignant liver cells produce AFP-L3% in the early stages. AFP-L3% can be detected in the serum of approximately 35% of small HCC (<3cm)[16]. It has also been shown to be associated with more aggressive HCCs and to predict a worse outcome[17]. HCC patients with positive AFPL-3 isoform would have worse liver function, poorer histology and larger tumor mass[18].

The present study aimed at evaluating the utility of AFPL-3% in detection of HCC developing on top of liver cirrhosis in patients who have serum level of AFP <200 ng/ml, which is not a diagnostic level to HCC and to compare the levels of both AFP and AFP-L3% in HCC versus LC patients without HCC. Also to define the best cut-off level of each tumor marker with best sensitivity and specificity for HCC detection in the tested patients.

SUBJECTS AND METHODS

This study was conducted on 75 subjects, 25 newly diagnosed HCC patients, and 25 LC patients with no evidence of HCC, recruited from the out patient clinic and in patients of the Tropical Medicine Department, Cairo University as well as the National Liver Institute, Menoufyia University. In addition, 25 apparently healthy subjects of matching age and gender served as a control group. All members of the study (patients and controls) gave an informed consent. All HCC cases developed on top of liver cirrhosis, they were not on treatment as they were newly diagnosed. The diagnosis of HCC was based on abdominal ultrasound, spiral abdominal CT with the characteristic picture of HCC in 14 patients and typical histopathological findings in the other 11 patients. The serum AFP levels in HCC patients were <200 ng/ml. HCC and cirrhotic patients had matching Child Pugh scores.

All patients and control subjects were subjected to: full history taking and clinical examination. Liver function tests using the Beckman Coulter clinical Autoanalyzer, USA, and viral markers (HBsAg and anti-HCV antibodies) using the third generation EIA technique (Abbotts Laboratory, Germany) were done to all. Measurement of serum levels of AFP and AFP-L3% were performed for all cases.

Measurement of AFP and AFP-L3%:

Samples were stored at -70°C till analysis. The total serum AFP and AFP-L3% were measured by a liquid-phase binding assay on the Wako LiBASys clinical auto analyzer (Wako Pure Chemical Industries, Ltd. Osaka, Japan). The typical inter-assay variance for this test, expressed as the coefficient of variance, is between 2.8% and 13.4% for AFP-L3% and 2.6% and 4.6% for AFP concentration[17]. The LiBASys auto analyzer has a lower limit of detection for total AFP concentration of 0.8 ng/ml and for AFP-L3% of 0.5%.

Statistical analysis: Data was statistically analyzed using SPSS (statistical package for social science) program version 13 for windows, for all the analysis p value < 0.05 was considered statistically significant.

RESULTS

The mean age of the studied HCC cases was 55.0 ± 6.5 years, 56% (14/25) were males and the mean age in LC patients was 54.0 ± 7.8 years and 60% (15/25) were males. The mean age of the controls was 57±10 years and 48% (12/25) were males. The histopathological grades of HCCs (in the biopsied patients) were grade I in 9%, grade II in 55% and grade III in 36% of cases.

The three studied groups (HCC, cirrhosis and control) were homogenous regarding age and gender (P>0.05). (Table 1).

Table 1: Demographic features, mean laboratory tests, viral status and Child classification in HCC, Cirrhosis and Controls.

variables	HCC (N=25) Mean ± SD	Cirrhosis (N= 25) Mean ± SD	Control (N=25) Mean ± SD	P-Value
Age (years)	55± 6.5	54.0± 7.8	57±10	> 0.05
Gender (male/ female)	14/11	15/10	12/13	> 0.05
Child classification (A/B/C)	8/11/6	6/9/10	-----	> 0.05
HCV-Ab positive	25(100%)	25 (100%)	0	> 0.05
HBV-sAg	0	0	0	
AST (IU/L)	108.2 ± 114	90.5±55.1	27.7 ±7	< 0.01**
ALT (IU/L)	84.1 ±39.5	63.9± 65.7	22± 5.5	< 0.01**
ALP (IU/L)	170.6 ± 122	95± 42.3	45.2±12	< 0.01**
GGT (IU/L)	96.5 ± 68.2	45.4 ±41.7	21.3± 9	< 0.01**
Alb (g/dl)	1.7 ± 0.3	1.9 ±0.4	3.5 ±0.3	< 0.01**
T.Bil (mg/dl)	5.2 ± 2.2	4.3±2.5	0.7± 0.2	< 0.01**
D.Bil (mg/dl)	2.1 ±1.8	3.62 ±2.8	0.18± 0.07	< 0.01**
T.P (g/dl)	7.5± 1.3	6.5± 0.8	8.2 ± 0.5	> 0.05
PC%	58.6± 17.5	49.4 ±16.3	96.8± 3.4	< 0.01**

Post Hoc test is not mentioned in the table
**P<0.01= highly significant

Regarding the liver function tests: There was no significant difference (P>0.05) between HCC and cirrhosis groups regarding all studied variables except ALP and GGT (P<0.05) being high in HCC group. There was a highly significant difference (P<0.01) between HCC and control as well as cirrhosis and control groups regarding all studied variables except total protein (Table 1). (Post Hoc Tamhane test is not shown in the table)

Regarding serum levels of AFP and AFP-L3%: The mean serum level of AFP showed a significant elevation (P<0.05) and a highly significant (P<0.01) elevation in HCC group compared to each of control and cirrhosis groups respectively. Also it showed a highly significant (P<0.01) elevation in cirrhosis group compared to control group. The mean serum level of AFP-L3% showed a highly significant (P<0.01) elevation in HCC group when compared to each of cirrhotic and control groups. Meanwhile, AFP-L3% showed no statistically significant difference between cirrhotic and control groups (P>0.05). (Table 2), no significant correlation (r = -0.23, P>0.05) was observed between the mean serum levels of AFP and AFP-L3% in HCC patients (Figure1), there was a significant positive correlation (P<0.05) between AFP and both AST and ALT. While, AFP-L3% showed inverse significant correlation (P<0.05) with PC% in HCC patients (not shown in the table).

Table 2: Serum levels of AFP and AFPL-3% in the studied groups.

Studied variables	HCC (N=25) Mean ± SD	Cirrhosis (N=25) Mean ± SD	Control (N=25) Mean ± SD	P-Value	Tamhane post Hoc test
AFP (ng/ml)	102.4 ± 43.7	60.8 ± 58	8.8 ± 5.5	< 0.01*	P1= < 0.05* P2= < 0.01** P3= < 0.01**
AFP-L3%	31.9 ± 21.2	1.73 ± 4.2	1.53 ± 3.2	< 0.01**	P1= < 0.01** P2= < 0.01** P3= > 0.05

P1 between HCC and Cirrhosis

P2 between HCC and Control

P3 between Cirrhosis and Control

*P< 0.05= significant ** P< 0.01= highly significant

The mean serum level of AFP showed no significant difference (P>0.05) regarding Child Pugh classification, numbers of tumor foci nor tumor size. On the other hand it showed a significant difference (P<0.05) regarding lymph node invasion and TNM classification in HCC patients (Table 3). Meanwhile the mean serum level of AFP-L3% showed no significant difference (P>0.05) regarding all these parameters (Table 4).

Table 3: Serum level of AFP regarding some studied clinical variables in HCC patients

Studied variables	AFP Mean ± SD (ng/ml)	P- value
Child classification:		
-A (n=8)	93.7 ± 47.7	> 0.05
-B (n=11)	109.9 ± 48.1	
-C (n=6)	99.88 ± 29.9	
LN invasion:		
-Positive (n=6)	134.3 ± 34.8	< 0.05*
-Negative (n=19)	91.8 ± 41.8	
TNM classification:		
-I (n=6)	46.4 ± 64.34	
-II (n=9)	84.4 ± 35.6	< 0.05*
-III (n=8)	107.24 ± 25.16	
-IVa (n=2)	159.5 ± 55.86	
Tumor nodule:		
-Solitary nodule (n=15)	105.41± 51.8	> 0.05
-Multiple nodule (n=10)	98.3 ± 31.2	
Tumor size:		
-> 3 cm (n=18)	111.65± 46.5	> 0.05
-≤ 3 cm (n=7)	91.6 ± 39.4	

PostHoc test is not mentioned in the table *P<0.05 =significant

Table 4: Serum level of AFP-L3% regarding some studied clinical variables in HCC patients

Studied variables	AFP-L3% Mean ± SD	P value
Child classification:		
-A (n=8)	35.02 ± 28.14	> 0.05
-B (n=11)	29.75 ± 17.81	
-C (n=6)	31.86 ± 19.44	
LN invasion:		
-Positive (n=6)	31.5 ± 17.72	> 0.05
-Negative (n=19)	32.09 ± 22.74	
TNM classification:		
-I (n=6)	27.5 ± 32.06	
-II (n=9)	33.71 ± 23.62	> 0.05
-III (n=8)	33.8 ± 14.64	
-IV (n=2)	27.7 ± 9.47	
Tumor nodule:		
-Solitary nodule (n=15)	30.2 ± 24.06	> 0.05
-Multiple nodule (n=10)	34.4 ± 17.42	
Tumor size:		
-> 3 cm (n=18)	33.16 ± 22.56	> 0.05
-≤ 3 cm (n=7)	30.53 ± 20.52	

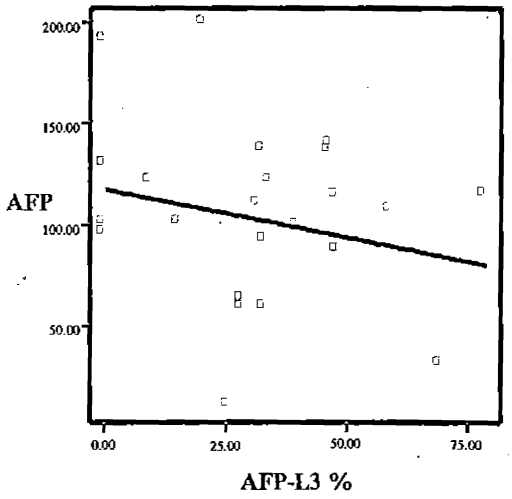

Figure 1: Correlation between AFP and AFP-L3% in HCC patients

ROC curve analysis: (Figure 2, Figure3 & Table 5) indicated that, AFP cut-off value of 77.8 ng/ml yielded the best sensitivity and specificity for differentiating patients with HCC from those without HCC. For AFP-L3%, the best cut-off value was 15.4%. Based on these ROC curve defined cut-offs, the sensitivity, specificity, positive predictive (PPV), negative predictive value (NPV), and accuracy of AFP were 75%, 68%, 69.2%, 73.9% and 70.4%, respectively, while for AFP-L3% were 79.2 %, 100.0 %, 100.0 %, 83.3%, 96.2 %, respectively. By using the two markers in detection of HCC (Table 5), using AFP and / or AFP-L3%, the sensitivity, specificity, PPV, NPV and accuracy were 100%, 68.0%, 75.0%, 100.0%, 83.7% respectively.

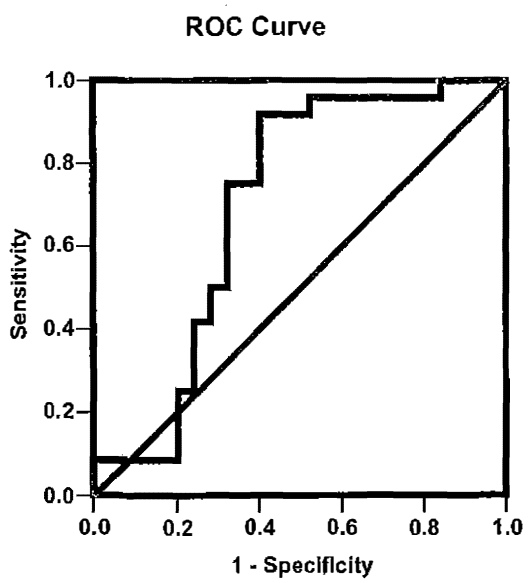

Figure 2: ROC curve of AFP.

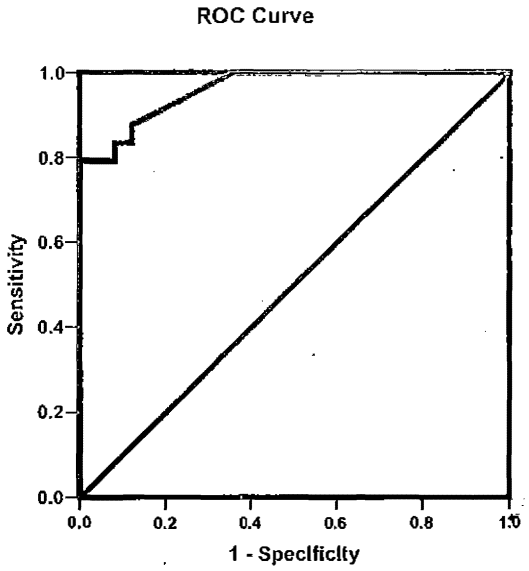

Figure 3: ROC curve of AFP-L3%.

Table 5: ROC curve analysis of AFP and AFP-L3%, in HCC patients versus cirrhosis patients (n= 50) and combined use of the two markers.

variable	Sensitivity	Specificity	PPV	NPV	Accuracy
AFP Cut-off level 77.8 ng/ml	75.0 %	68.0 %	69.2 %.	73.9 %.	70.4%
AFP-L3% Cut-off level 15.4%	79.2 %	100.0 %	100.0 %.	83.3 %.	96.2%
AFP-L3% Cut-off level 9.2%	83.3 %	92%	90.5%	80.1%.	96%
AFP-L3% (Cut-off level 15.4%) and/or AFP(Cut-off level 77.8 ng/ml) (Any test positive)	100.0 %	68.0 %	75.0 %	100.0 %	83.7 %

DISCUSSION

In Egypt, the incidence of HCC is expected to increase by at least 2.4 folds in the next two decades because of the high prevalence rate of hepatitis C virus in the general population which accounts for most cases of cirrhosis and HCC[19].

Hepatocellular carcinoma is a leading cause of mortality among patients with chronic liver disease. Both the incidence and mortality rates from HCC are increasing[20]. Because HCC takes many years to develop, emphasis has been placed on surveillance of patients who are at high risk for HCC[21]. If detected when HCC lesions are small and patients are asymptomatic, the HCC is potentially curable[22].

AFP is the most established tumor marker in HCC and the gold standard by which other markers for HCC are judged[23]. AFP-L3% is the LCA-bound fraction of AFP with an additional α1-6 fucose residue attached at the reducing terminus of N-acetylglucosamine, it is the major glycoform of AFP in the serum of HCC patients, and it can be detected in approximately 35% of patients with small HCC (< 3 cm)[11,24].

The current study detected a statistically significant increase of the mean serum level of AFP in HCC patients compared to each of cirrhotic patients ($P<0.05$) and controls ($P<0.01$), also a highly significant increase ($P<0.01$) of AFP serum level in cirrhotic patients compared to controls were detected.

Consistent with these results Jia et al.[25] found that AFP serum level is significantly elevated in HCC patients than in cirrhotic patients and healthy controls, and stated that an increased serum AFP level is highly suggestive in HCC diagnosis.

Some studies have demonstrated that the presence of elevated levels of AFP in patients with LC is a risk factor for the development of HCC[26].

However, Yao et al.[10] reported that although serum AFP level is a useful tumor marker for the detection and

monitoring of HCC, the false-negative rate may be as high as 40% for patients with early stage HCC. Increased serum AFP concentration below 400 ng /ml was also reported in 10-15% of cases of acute and chronic hepatitis, liver cirrhosis and secondary hepatic malignancies[27]. AFP, therefore, does not always directly reflect the development of HCC. The lack of differences between patients not positive for a tumor marker and those positive for AFP alone indicates that the clinical significance of AFP as a marker of tumor progression of HCC is limited[15].

In the present study the mean serum level of AFP showed a significant increase (P<0.05) in HCC patients with lymph nodes invasion and in HCC patients with advanced TNM stage. Meanwhile, it showed no significant difference (P>0.05) regarding Child Pugh classification, number of tumor foci nor tumor size.

In accordance with these results Yang et al.[28] observed that serum level of AFP was significantly higher in stage III-IV than stage I-II, indicating that AFP level correlated with disease severity and is elevated in advanced stage of the disease, Tsai et al.[9] and Durazo et al.[29] found no correlation between serum level of AFP and tumor size nor the number of tumor foci.

The ROC curve analysis is a graphic method which can be used to determine the optimal cut-off values. In addition it is a precise and valid measure of diagnostic accuracy[9].

In the current study, the ROC curve analysis indicated that AFP cut-off value of 77.8 ng/ml yielded the best sensitivity and specificity for differentiating patients with HCC from those without HCC in the non diagnostic range. At this cut-off, the sensitivity, specificity, PPV, NPV, and accuracy of AFP were 75%, 68%, 69.2%, 73.9% and 70.4% respectively.

The sensitivity and specificity of AFP in HCC screening and surveillance studies varies according to test thresholds, study designs, and patient populations' examined[30].

Prospective studies evaluating AFP for HCC surveillance reported sensitivities of 39%–64%, specificities of 76%–91%, and PPV of 9%–32%[31]. The sensitivity and specificity of AFP for HCC are highly dependent on the cut-off value above which AFP is considered a positive test[32]. The Italian and the American Association for the Study of Liver Diseases guidelines consider a level ≥200 ng/ml as the cut-off point for diagnosis[33]

AFP-L3% is an isoform of AFP which is very specific for HCC[34]. The AFP-L3% is the percentage of AFP-L3 over the total AFP level. The cut-off for the presence of HCC is 10%[29].

Previous studies have shown that AFP-L3% can be detected in the serum of patients with small tumors[16]. Patients who are AFP-L3% positive have been reported to have tumors with rapid growth, larger size, with portal vein invasion and with metastasis more often when compared to AFP-L3% negative patients[35]. Also, previously reported prospective studies have shown that the elevation in AFP-L3% occurs earlier than the elevation in total AFP concentration in HCC patients[34, 36].

In this study the mean serum level of AFP-L3% showed a highly significant increase (P<0.01) in HCC patients compared to both cirrhotic patients and controls, while it showed no significant difference (P>0.05) between cirrhotic patients and controls.

In agreement with these results, Zaninotto et al.[37] found that the fucosylation index of AFP (AFP-L3%) was significantly higher (P < 0.0001) in patients with HCC than in patients with benign liver disease. They suggested that fucosylated AFP is of diagnostic value in differentiating between HCC and benign hepatic diseases, and that its measurement may be useful in the early detection of HCC in patients with chronic liver disease.

Similarly, Xu et al.[38] detected an elevated level of AFP-L3% in patients with HCC than in patients with other liver diseases.

Sun et al.[39] had studied the value of AFP-L3% as a biomarker in diagnosis of HCC and found that its average value in patients with HCC was 36.4%, and this level was significantly higher than in the patients with benign liver diseases.

In the present study AFP-L-3% showed no significant difference (p>0.05) regarding Child classification, lymph nodes invasion, TNM classification, number of tumor foci nor tumor size. The results of the study done by Leerapun et al.[14], matched these results as they noticed no association between AFP-L3% and tumor size, stage, vascular invasion or histological grade. Also, Durazo et al.[29] observed no significant correlation between AFP-L3% and tumor size nor tumor characteristics (single, multifocal or diffuse).

From the above mentioned results it can be concluded that AFP-L3% level is elevated early in HCC patients with no significant increase in patients with large tumor size.

In this study, no significant correlation was observed between the mean serum levels of AFP and AFP-L3% in HCC patients, indicating that increased AFP-L3% level did not depend on markedly increased serum AFP value, so it can be used for detection of HCC in patients at high risk of developing HCC who have slightly elevated AFP level.

Similar results had been reported by many authors, where Taketa et al.[36] Kusaba, [40] & Khien et al.[18] found that AFP-L3% was not related to serum AFP concentration.

In the current study, the optimal cut-off value of AFP-L3% obtained from the ROC curve was 15.4%, with sensitivity, specificity, PPV, NPV and accuracy of 79.2%, 100%, 100%, 83.3%, and 96.2% respectively. As observed in previous studies, a high specificity seems to be a constant feature of AFP-L3%. When AFP-L3% was tested at lower cut-off level (9.2%), it was still positive for HCC with sensitivity

of 83.3%, specificity of 92%, PPV of 90.5%, NPV of 80.1% and accuracy of 96%.

Xu et al.[38] considered AFP-L3% ≥ 10% as a diagnostic criterion and found that the sensitivity for diagnosis of HCC was 90.9% and concluded that detection of AFP-L3% seemed to be of clinical value in diagnosis and differential diagnosis of HCC and it may be especially important for identifying patients with HCC whose AFP level is low.

In a study done by Sun et al.[39] at AFP-L3% level ≥ 10% which was taken as a diagnostic criterion, the sensitivity of AFP-L3% in HCC diagnosis was 84.8%, the specificity was 92.5% with total confirmatory rate of 87.9% . They stated that AFP-L3% is a valuable biomarker in diagnosis and prediction of prognosis of HCC.

In accordance with the results of the present study, Sterling et al.[41] noticed that among patients with increased AFP level <200 ng/ml, AFP-L3% was highly specific marker (specificity=86.6%) and they found that among 29 HCC patients with AFP levels <20 ng/ml, 13 patients had increased level of AFP-L3% and had concluded that AFP-L3% had higher correlation values with an absence of HCC, as well as a higher specificity and NPV than total AFP. In agreement with the aforementioned data, Durazo et al.[29] observed that levels of AFP and AFP L-3% were significantly higher in patients with HCC than in those without HCC, and the cut-off value with the best sensitivity and specificity for each test was=25 ng/ml for AFP and > or =10% for AFP-L3%. The sensitivity and specificity for AFP were 69% and 87% and for AFP-L3% were 56% and 90% respectively. So AFP is less specific for HCC compared to AFP-L3%. In an evaluation of the utility of AFP-L3%, by Leerapun et al.[14] for diagnosis of HCC in U.S.A referral population, they found that in patients with total AFP of 10-200 ng/ml, an AFP-L3% cut-off >10% had a sensitivity of 71% and a specificity of 63% for diagnosis of HCC. A cut-off >35%, AFP-L3% had a reduced sensitivity of 33% but an increased specificity of 100% for diagnosis of HCC.

In the current study, using the two markers in detection of HCC, using AFP and / or AFP-L3%, was more effective in detection of HCC patients as the sensitivity was 100.0% and NPV was 100%.

No significant correlation between AFP-L3% and the liver profile tests was observed in this study, except with PC%, which showed significant inverse correlation. Traditional AFP showed significant positive correlation with both AST and ALT, and this might be attributed to the progressive pathology of the liver indicating that the probability of AFP increase is due to ongoing liver damage rather than HCC or both factors together. In agreement with these results, Sterling et al.[41] observed that, increased alanine aminotransferase levels were associated with increased total AFP but not AFP-L3%.

AFP is fairly specific marker for HCC. Sensitivity of AFP to detect HCC is improved by combining it with its fraction AFP-L3%. As both showed sensitivity 100%, NPV 100% and accuracy 83.7%.

CONCLUSION

Determination of AFP-L3%, in combination with AFP, increase the sensitivity for detection of HCC in individuals with total AFP <200ng/ml which is not a diagnostic range.

So for detection of HCC patients, AFP and its fraction AFP-L3% can be used as they both showed sensitivity and NPV of 100% and accuracy of 83.7%. For confirmation of diagnosis, and to exclude false positive cases, AFP-L3% can be used, as it showed specificity and PPV of 100%, (no false positive results).

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