Assessment of Squamous Cellular Carcinoma Antigen (SCCA) and KL-6 as a Tumor Markers and Their Correlation to Tumor Size in HCC

Hosam M. El-Ezawy[1], Nashwa Shebil[2], Suzan El-Hasanin[3], Moones A Obada[4], Mohamed El-Waraay[5]

Departments of [1]Clinical Biochemistry, [2]Hepatology, [4]Clinical Pathology and [5]Radiology, National Liver Institute, Menoufiya University, Egypt
Department of [3]Oncology, Faculty of Medicine, Menoufiya University, Egypt
sohazaki69@yahoo.com

Abstract: Hepatocellular carcinoma (HCC) is the fifth most common malignancy in the world. In Egypt, HCC was reported to account for about 4.7% of chronic liver disease (CLD) patients. Squamous cellular carcinoma antigen (SCCA) has been reported to be strongly expressed in HCC tissue hampering its extensive use in clinical practice. KL-6 was originally found using a murine monoclonal antibody reported to have a high positive rate in different non hepatic malignancies. The present study aimed to evaluate the clinical usefulness of serum levels of SCCA and KL-6 as serological markers for early detection of HCC compared to AFP and correlation between each of them with tumor size. The study comprised of three groups. Group 1: included 82 patients with HCC diagnosed based on clinical, laboratory, abdominal ultrasonography and CT investigations. Group 2: included 46 patients with liver cirrhosis. Group 3: included 35 apparently healthy subjects matched for age and sex. All groups were subjected to full history taking, clinical examination and laboratory investigations including liver function tests, viral markers, AFP, SCCA and KL-6. A statistical significant difference (<0.001) was detected between each serum level of AFP, SCCA and KL-6 in the three studied groups, where the maximum increase of these parameters was detected in the HCC group. The sensitivity and the specificity is 81.7% & 76.1% for SCCA and 76.8% & 78.3% for KL-6. A statistical significant increase ($p<0.001$) was observed in the mean serum level of each SCCA and KL-6 with increasing the tumor size and Child score. The results of this study suggest that SCCA and KL-6 could represent a useful tool as a marker for detection of HCC and for differential diagnosis between HCC and cirrhosis. A large scale study is needed to investigate their clinical value to diagnose patients with HCC with different etiological causes and their correlation between each of them with tumor size.

[Hosam M. El-Ezawy, Nashwa Shebil, Suzan El-Hasanin, Moones A Obada, Mohamed El-Waraay. **Assessment of Squamous Cellular Carcinoma Antigen (SCCA) and KL-6 as a Tumor Markers and Their Correlation to Tumor Size in HCC.** Journal of American Science 2012;8(3):172-179]. (ISSN: 1545-1003).
http://www.americanscience.org. 21

Key words: Hepatocellular carcinoma (HCC), KL-6, Squamous cellular carcinoma antigen (SCCA).

1. Introduction

Hepatocellular carcinoma (HCC) is a common cancer that typically occurs in the setting of cirrhosis and chronic hepatitis virus infections. Hepatitis B and C account for approximately 80% of cases worldwide (**Lau and Lai, 2008**). HCC is currently the fifth most common malignancy in men and the eighth in women worldwide; its incidence is increasing dramatically in many parts of the world. Recognition of those at risk and early diagnosis by surveillance with imaging, with or without serologic testing, are extremely important (**Lai and Lau, 2005**).

The diagnosis of HCC is typically made by radiological liver imaging in combination with serum alpha-fetoprotein (AFP). AFP is a tumor marker that is elevated in 60%-70% of patients with HCC (Wu et al., 2006). Normally, levels of AFP are below 10ng/ml, but marginal elevations are common in patients with chronic hepatitis or cirrhosis. However, all patients with elevated AFP should be screened for HCC with imaging, especially if there has been an increasing trend from baseline level (Soresi et al., 2003). The specificity of AFP is very high when the levels are above 400 ng/ml in patients without testicular tumor. There is an agreement that biopsy proof of HCC is not required prior to surgery and that noninvasive diagnostic criteria can be applied. Seeding of tumor in the needle track occurs in 1%-3% of cases (Ding et al., 2005).

Abdominal ultrasonography (US) is usually the first line investigation within a surveillance plan or within the standard evaluation of patients with suspected or known liver disease. Upon US detection of a hepatic nodule, the next step is to characterize the nodule and establish its diagnosis. In the setting of a patient with known chronic hepatitis or cirrhosis of other etiology, a liver mass found incidentally or on US screening has a high likelihood of being HCC (Lencioni et al., 2005). If AFP is normal, further dynamic imaging [contrast-enhanced computed tomography (CT) and magnetic resonance imaging (MRI) with gadolinium injection, or lipiodol angiography with follow-up CT] usually allows a confident diagnosis to be made and the clinician can

proceed to assessment of treatment without the need for biopsy (Vauthey et al., 2002). In the few cases where diagnostic doubt persists, biopsy may be indicated. However, difficulty still exists in recognizing very small (<1 cm) HCCs (Lencioni et al., 2005). Early recognition of the onset of hepatocellular carcinoma (HCC) would help to select more effective therapies for patients, leading to a better prognosis and life span (**Bruix *et al.*, 2001**).

AFP, the only marker common used in clinical practice, displays poor sensitivity and a high specificity only for values higher than 400 IU/ml. However, because AFP concentrations are directly correlated with tumor size, the reliability of such a marker appears inadequate for early recognition of HCC (**Farinati *et al.*, 2006**).

Thus, searching another tumor marker, that together with AFP could improve the diagnostic utility of the later, seemed to be justified. This has prompted a high number of studies conducted to validate different new biomarkers, but very little has yet been reported about biomarkers helping to achieve an early detection of HCC (**Giannelli and Antonaci, 2006**). All the proposed biomarkers failed to discriminate between liver cirrhosis (LC) and HCC in a satisfactory manner, in terms of diagnostic accuracy, reproducibility of the results, or technical issues related to the biomarker detection method (**Marrero and Lok, 2004**). For this reason, the simultaneous use of different tests seems to offer a promising approach that warrants further investigation (**Yao *et al.*, 2007**).

Squamous cellular carcinoma antigen (SCCA) is a member of the high molecular weight family of serine protease inhibitors named serpins. Two highly homologous isoforms have been reported to be expressed in HCC tissues at protein and translational levels (**Pontisso *et al.*, 2004**).

SCCA has also been reported to be over expressed in tumoral compared to paired peritumoral tissue of HCC, suggesting a role as a potential marker for histological detection of HCC (**Uemura *et al.*, 2000**). Recently, SCCA has been investigated in regenerative and dysplastic nodules of HCC tissue. Interestingly, results show that SCCA was poorly expressed in regenerative tissue but strongly increased in dysplastic nodules, suggesting a role as a potential marker for early detection of HCC (**Uemura *et al.*, 2000**).

KL-6 was originally found using a murine monoclonal antibody that recognized an undefined sialylated carbohydrate chain on a mucin like glycoprotein which was also defined as MUC1.The cell membrane MUC1 was found to regulate cell adhesion properties (**Sagara *et al.*, 1999**). KL-6 has been first shown to be elevated in patients with interstitial pneumonia (**Kohno, 1999**). It was also reported to have a high positive rate in different non hepatic malignancies and its expression was also correlated with metastatic potential of the primary tumor in some of them (**Tanimoto *et al.*, 1999**).

It has also been studied as a fibrosis marker in patients with HCV-related chronic liver disease and was found to correlate with the degree of irregular regeneration of hepatocytes (**Moriyama *et al.*, 2003**).

Aim of this study was to investigate the serum levels of KL-6 and SCCA compared to AFP as a known biomarker for diagnosis of HCC in Egyptian patients, proving its role as a new diagnostic and prognostic marker of early detection of HCC in Egypt and to investigate presence of relation between their serum levels and the size of the tumor.

2. Materialand Methods:

The study was performed on Hepatology Departement, National Liver Institute and Oncology Department, Faculty of Medicine, Menoufiya University. Group (1) included 82 patients with HCC [68 male (82.9%) and 14 female (17.1)]. Their mean age was 46.1 ± 8.1years. HCC was diagnosed by the presence of characteristic hepatic masses by US using apex 3000, CT scan which performed on a helical scanner (Somatom Plus 4; General Electric Medical Systems, Milwaukee, WI, USA) with 2-mm collimation and subsequent sagittal and coronal reformats and MRI examination was performed on a 1.5-T MR scanner (Signa; Siemens Medical Systems, Erlangen, Germany). Group (2) included 46 patients of chronic liver diseases and liver cirrhosis [44 male (78.6%) and 12 female (21.4%)] with mean age 42.5 ± 8.5 years. They were diagnosed according to clinical and radiological evidence of portal hypertension. Group (3) included 35 apparently healthy subjects as a control group [37 male (77.1%) and 8 female (22.9%)] with free clinical and radiological evidence of portal hypertension, HCC, chronic liver diseases and liver cirrhosis. All the procedures included in this study were approved by the Research Ethics Committee of National Liver Institute and Faculty of Medicine, Menoufiya University, Egypt. Blood samples were collected from all individuals included in this study and centrifuged. The serum was kept frozen at -70^{o}C until assay.

Blood sample from cases and controls were tested for HBV surface antigen (HBsAg) using Kits from Sorin-Biomedica Co. (Italy) and anti bodies against HCV (anti–HCV) using third – generation enzyme link immunoassay using Murix kits (Republic of South Africa). These kits implement qualitative methods based on enzyme linked immunosorbant assays (ELISA). The procedures were done according to the manufacturer's instructions.

AFP was measured by RIA Kit (BIVENDER ,USA) using MMR-20 GERMANY, normal values of kit up to 20 IU/ ml.

Serum determination of the SCCA was carried out using an ELISA kit (GenWay, USA) following the manufacturer's instructions. The SCCA ELISA kit is based on a sandwich system where an HRP-conjugate streptavidin secondary antibody is used to reveal the reaction (Cataltepe *et al.,* 2000).

Serum level of KL-6 was determined, by the sandwich enzyme immunoassay method using the KL-6 antibody (Ab) as both the capture and tracer Ab, using AviBion Human KL-6 ELISA Kit, Orgenium Laboratories, Finland (Hirasawa *et al.,* 1997).

Statistical analysis:

Statistics were carried out using Statistical Package for Social Science program (SPSS). Data were presented as mean ± SD. The Pearson`s correlation coefficients were calculated for the normally distributed values. However, Spearman`s correlation coefficients were done for the not normally distributed values. Receiver Operating Characteristic (ROC) curve was produced for measured parameters to investigate the sensitivity, specificity and the cut-off values of each AFP, SCCA and KL-6. P value less than 0.05 was considered statistically significant.

3. Results:

Clinical and laboratory data of patient groups were listed in table (1). When analysis of the results of the mean serum level of each AFP, SCCA and KL-6 in the three studied groups, a statistical significant difference (<0.001) was observed between the groups where the maximum increase of these parameters was detected in the HCC group.

Table (2) showed the descriptive statistics of the pathological lesion in the HCC group. Figure (1) represented the ROC curve of serum level of each SCCA, KL-6 and AFP to differentiate HCC patients from cirrhotic patients. When analysis of the results of the ROC curve, this study found the cut-off point is 0.23 ng/ml, 426.5 U/ml and 91.5 IU/ml for SCCA, KL-6 and AFP respectively. The sensitivity and the specificity is 81.7% & 76.1% for SCCA and 76.8% & 78.3% for KL-6 as in table (3).

Regarding the ROC curve of the serum level of each SCCA, KL-6 and AFP to differentiate cirrhotic patients from healthy controls, table (4) and figure (2) showed that the area under the curve for KL-6 was 0.64 (CI95% 0.533 - 0.759) with a cut-off 232.0 U/ml. This predicted a sensitivity of 69.6% and a specificity of 48.9%. While serum AFP is better in this differentiation as the area under the ROC curve was 755 (CI95% 0.646 – 0.864) with a cut-off 11.95 IU/ml, predicting a sensitivity of 78.3% and a specificity of 57.8%.

Considering the serum level of SCCA as in table (5), it showed a significant positive correlation with each of tumor size (r=0.519 & p<0.001), Child score (r=0.267 & p<0.01), AFP (r=0.502 & p<0.001) and KL-6 (r=0.716 & p<0.001). Regarding the serum level of KL-6, there is also a significant positive correlation with each of the previous parameters.

Table (6) showed a statistical significant increase (p<0.001) in the mean serum level of each SCCA and KL-6 with increasing the tumor size and Child score. While, no significant difference in the mean serum level of the same parameters was detected with different viral infection (non-viral, HCV-related, HBV-related or both HCV & HBV).

Table 1: Statistical comparison of different studied parameters in the three studied groups

	Studied groups						Test of significance	P value
	HCC N = 82		Liver cirrhosis N = 46		Control N = 35			
	N	%	N	%	N	%		
Viral infection:								
Non-viral	32	39.0	22	47.8	22	62.8	2.19	>0.05
HCV-related	35	42.7	18	39.1	10	28.6		
HBV-related	5	6.1	2	4.3	2	5.7		
Both C&B-related	10	12.2	4	8.7	1	2.9		
Child classification:								
A	23	28.0	16	34.8	00	00		
B	18	22.0	11	23.9	00	00	0.96	>0.05
C	41	50.0	19	41.3	00	00		
AFP (IU/ml): (X ± SD)	2433.1±5153.9		56.85±71.03		10.50 ± 5.13		72.79	<0.001
SCCA (ng/ml): (X ± SD)	0.52± 0.28		0.19 ± 0.10		0.17 ± 0.11		67.35	<0.001
KL-6 (U/ml): (X ± SD)	597.3 ± 239.3		289.130±128.32		222.17 ± 101.56		85.01	<0.001

P-value is non significant at >0.05 and P-value is highly significant at <0.001

Table 2: Descriptive statistics of the pathological lesion in HCC group:

The pathological lesion	No=82	%
Lesion:		
Solitary	48	(58.5)
Nodular	34	(41.5)
Tumor size:		
<3	33	(40.2)
3-5	29	(35.4)
>5	20	(24.4)
Differentiation:		
Well	60	(73.2)
Poor	22	(26.8)

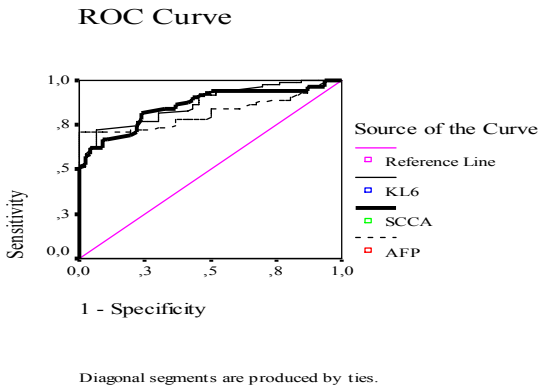

Figure 1: ROC curve of serum level of each SCCA, KL-6 and AFP to differentiate HCC patients from cirrhotic patients

Table 3: ROC curve analysis of the studied parameters (SCCA, KL-6 and AFP) to differentiate HCC patients from cirrhotic patients

	SCCA (ng/ml)	KL-6 (U/ml)	AFP (IU/ml)
Area under the curve	0.857 (<0.001)	0.868 (<0.001)	818 (<0.001)
95 % CI	0.794 – 0.921	0.809 – 0.928	0.745 – 0.890
Cut off point	0.236	426.5	91.5
Sensitivity	81.7%	76.8%	72.0
Specificity	76.1%	78.3%	80.4
Positive predictive value	85.9%	86.3%	86.8
Negative predictive value	70.0%	65.5%	61.7
Accuracy of the test	79.7%	77.3%	75.0

P-value is highly significant at <0.001

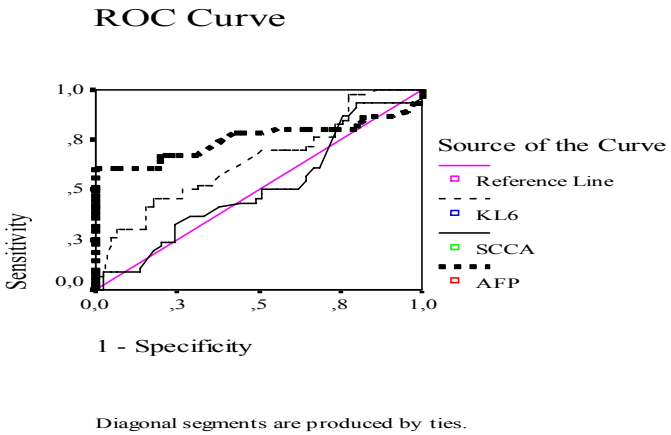

Figure 2: ROC curve of serum level of each SCCA, KL-6 and AFP to differentiate liver cirrhotic patients from healthy controls

Table 4: ROC curve analysis of serum level of the studied parameters (SCCA, KL-6 and AFP) to differentiate liver cirrhotic patients from healthy controls

	SCCA (ng/ml)	KL-6 (U/ml)	AFP (IU/ml)
Area under the curve	0.508 (>0.05)	0.646 (<0.05)	755 (<0.001)
95 % CI	0.388 – 0.628	0.533 – 0.759	0.646 – 0.864
Cut off point	0.128	232.0	11.95
Sensitivity	60.9%	69.6%	78.3
Specificity	33.3%	48.9%	57.8
Positive predictive value	48.3%	58.2%	65.5
Negative predictive value	45.5%	61.1%	72.2
Accuracy of the test	47.3%	59.3%	68.1

P-value is non significant at >0.05, P-value is significant at <0.05 and highly significant at <0.001

Table 5: Correlation between each SCCA and KL-6 with different measured parameters in HCC group

	SCCA (ng/ml)		KL-6 (U/ml)	
	Correlation coefficient (r)	***P* value**	**Correlation coefficient (r)**	***P* value**
Tumor size	+ 0.519	<0.001	+ 0.524	<0.001
Child classification	+ 0.267	<0.01	+ 0.204	<0.01
AFP (IU/ml)	+ 0.502	<0.001	+ 0.405	<0.001
SCCA (ng/ml)	+ 1.0	<0.001	+ 0.716	<0.001
KL-6 (U/ml)	+ 0.716	<0.001	+ 1.0	<0.001

P-value is highly significant at <0.01 and <0.001

In this table, Pearson's correlation coefficients was used for all the variables except for tumor size and Child classification (Spearman's correlation was done as these variables aren't normally distributed).

Table 6: Statistical comparison between each of serum level of SCCA & KL- 6 with tumor size and different measured parameters in HCC group

	SCCA (ng/ml)			KL-6 (U/ml)		
	SCCA X ± SD	**Test of significance**	***P* value**	**KL-6 X±SD**	**Test of significance**	***P* value**
Viral infection:						
Non-viral	0.504±0.309			578.71±222.27		
HCV-related	0.541±0.269	3.12	>0.05	619.14±237.85	4.86	>0.05
HBV-related	0.695±0.311			770.0±286.27		
Combined	0.429±0.224			494.0±250.57		
Tumor:						
Solitary	0.478±0.261	1.54	>0.05	554.63±225.69	1.73	>0.05
Nodular	0.578±0.305			651.83±248.16		
Tumor size:						
< 3	0.368±0.237	21.87	<0.001	448.42±209.49	23.09	<0.001
3 – 5	0.569±0.261			664.0±198.58		
> 5	0.707±0.262			746.25±208.47		
Child classification:						
A	0.386 ±0.260	9.39	<0.01	454.78 ± 212.56	12.69	<0.01
B	0.537 ±0.230			709.16 ±198.12		
C	0.592±0.297			628.14 ±236.94		
Differentiation:						
Well	0.446 ±0.244			532.86 ± 212.52		
Poor	0.729 ±0.286	3.99	<0.001	773.04 ±223.08	3.94	<0.001

P-value is non significant at >0.05 and highly significant at <0.01 & <0.001

4. Discussion

In Egypt, HCC was reported to account for about 4.7% of chronic liver disease (CLD) patients. Over a decade, there were nearly two-fold increases in proportion of HCC among CLD patients in Egypt (El-Zayadi *et al*., 2001).

Sherman, 2001 stated that serum AFP is the most commonly used marker for HCC neoplasm, but its real clinical usefulness is unclear & furthermore, the role of AFP in HCC screening and diagnosis has lost most of the appeal that it had in the presophisticated (i.e. multislice, contrast-enhanced computed tomography, magnetic resonance imaging, etc.) imaging era. Also, Goma *et al.,* 2003 reported that because of the reported sensitivity (39% - 65%) and specificity (76% - 94%) of serum AFP, it isn't sufficient for early diagnosis of HCC and additional effective markers are needed.

Aim of this study was to investigate the serum levels of KL-6 and SCCA compared to AFP as a known biomarker for diagnosis of HCC in Egyptian patients, proving its role as a new diagnostic and prognostic marker of early detection of HCC in Egypt and to investigate presence of relation between their serum levels and the size of the tumor.

In the current study a statistically significant difference in the mean serum level of each AFP, SCCA and KL-6 between the three groups with highest increase in the HCC group and a slight increase in the cirrhotic group. Hussien *et al.,* 2008 agreed with this study as they detected a highly significant difference in SCCA serum level ($P < 0.001$) between patients with HCC, CLD, and controls.

Trerotoli *et al.,* 2009 stated that SCCA tissue expression could be a marker for early detection of smaller HCC nodules. Also Pontisso *et al.,* 2006 observed that SCCA was consistently increased in patients with LC progression to HCC. Giannelli *et al.,* 2007 recommended the combined use of AFP and SCCA to increase the accuracy of HCC diagnosis.

Regarding the mean serum level of KL-6, Gad and his coworkers 2005 agreed with this study as they found a significantly higher mean KL-6 in HCC compared with non-HCC; either with or without LC; in addition no difference in mean KL-6 was found among HCC patients with and without LC. They stated that such findings together point to KL-6 association with HCC independent on the presence or absence of LC.

Applying the ROC curves analysis showed the best cut-off value to differentiate HCC patients from cirrhotic patients was 0.23 ng/ml for SCCA yielded 81.7% sensitivity and 76.1% specificity, 426.5 U/ml for KL-6 yielded 76.8% sensitivity and 78.3% specificity and 91.5 IU/ml for AFP gave sensitivity of 72% and specificity of 80.4%. From the previous results, SCCA was the most sensitive marker. It is explained by Trerotoli *et al.,* 2009 who stated that SCCA is more strongly positive in the tissues of smaller size of HCC and strongly increased in dysplastic tissues while it is poorly expressed in regenerative nodules. The earlier appearance of serum SCCA may be due to the fact stated by Uemura and his colleagues 2000 as they stated that SCCA is distributed mainly in the cytosol, not associated to membrane-bound vesicles and therefore is not properly secreted but rather released in the serum as a consequence of cell lysis. Hussien *et al.,* 2008 reported that SCCA level was elevated representing its useful role as a serological marker for diagnosis of HCC patients with normal level of AFP. While Beale *et al*., 2008 found no significance difference between the level of SCCA in patients with HCC arising on top of non alcoholic and alcoholic fatty liver disease. This may suggests that HCC marker may be etiology related and so results are different in Egyptian patients as most of the patient in this study was secondary to HCV (42.7 %), HBV (6.1%) and combined (12.2%).

Gad *et al.,* 2005 used a cut-off point of 334 U/ml in his analysis of KL-6 as a tumor marker in patients with HCC. This cut-off gave the best sensitivity (60%) in their study. Regarding the AFP cut-off level for the diagnosis of HCC, Goma and his colleagues, 2003 revealed an AFP value above 400-500 ng/ml has been considered to be diagnostic for HCC in patients with cirrhosis. Also, Lau and Lai, 2008 stated the specificity of AFP is very high when the levels are above 400 ng/ml in patients without testicular tumor.

In the current study, the serum level of KL-6 correlated positively with each of the tumor size and Child classification. This comes in accordance with Suzuki *et al*, in 2003 found that levels of serum KL-6 of liver cirrhosis patients were significantly higher than that for the chronic hepatitis patients and when liver cirrhosis was classified according to Child's system, the level of serum KL-6 for the Child B/C patients was significantly higher than that for the Child A patients. Thus they suggested a correlation between KL-6 and liver disease, stating that this marker reflects hepatic fibrosis even better than pulmonary fibrosis.

Regarding the serum level of SCCA, a similar statistical significant positive correlation was detected with the tumor size and Child classification. Hussien and his coworkers, 2008 stated that data in their study pointed to absence of correlation between SCCA level, size and burden of the tumor and severity of CLD and asked for further studies on larger number of patients to clarify this finding.

On the other hand, Trerotoli *et al.,* 2009 showed that the tissue expression of SCCA has been reported in a higher percentage of patients with pre-neoplastic dysplastic lesions than in regenerative nodules. Moreover, in the same study SCCA was reported to be

more strongly expressed in premalignant dysplastic nodules than in HCC and decreased expression of it occurred with the progression of tumor size. So, they suggested that SCCA could be a biomarker of pre-malignant transformation.

The results of this study suggest that SCCA and KL-6 could represent a useful tool as a marker for detection of HCC and for differential diagnosis between HCC and cirrhosis. A large scale study is needed to investigate their clinical value to diagnose patients with HCC with different etiological causes and their correlation between each of them with tumor size.

Corresponding author:
Dr. Hosam Mahmoud El-Ezawy, Department of Clinical Biochemistry, National Liver Institute, Menoufiya University, Shebin El-Kom, Egypt; Telephone: +201000629997; Fax: +20482237783; E-mail: hosamezawy@yahoo.com

References

Beale G, Cathopadhyay D and Gray J (2008): AFP, PIVKA III, GP3, SCCA and follisation as surveillance biomarkers for hepatocellular cancer in non-alcoholic and alcoholic fatty liver disease. BMC Cancer; 8: 200.

Bruix J, Sherman M, Llovet JM, Beaugrand M, Lencioni R, Burroughs AK, Christensen E, Pagliaro L, Colombo M and Rodes J (2001): Clinical management of hepatocellular carcinoma. Conclusions of the Barcelona-2000 EASL conference. European Association for the Study of the Liver. J Hepatol.; 35: 421-430.

Cataltepe S, Gornstein ER, Schick C, Kamachi Y, Chatson K, Fries J, Silverman GA, Upton MP (2000): Coexpression of the squamous cell carcinoma antigens 1 and 2 in normal adult human tissues and squamous cell carcinomas. J Histochem Cytochem., 48: 113-122.

Ding X, Yang LY, Huang GW, Y in perip ang JQ, Liu HL, Wang W, Peng JX, Yang JQ, Tao YM, Chang ZG and Ling XS (2005): Role of AFP mRNA expression in peripheral blood as a predictor for postsurgical recurrence of hepatocellular carcinoma: a systematic review and meta-analysis. World J Gastroenterol.; 11: 2656–2661.

El-Zayadi A, Abaza H, Shawky S, Mohamed MK, Selim OE and Badran HM (2001): Prevalence and epidemiological features of hepatocellular carcinoma in Egypt: A single center experience. Hepatol Res.; 19: 170.

Farinati F, Marino D, DE Giorgio M, Baldan A, Cantarini M, Cursaro C, Rapaccini G, Del Poggio P, Di Nolfo MA, Benvegnu L, Zoli M, Borzio F, Bernardi M and Trevisani F (2006): Diagnostic and prognostic role of alpha-fetoprotein in hepatocellular carcinoma: both or neither? Am J Gastroenterol.; 101: 524-532.

Gad A, Tanaka E, Matsumoto A, Abd-el Wahab M, Serwah H, Attia F, Ali K, Hassouba H, El-Deeb R, Ichijyo T, Umemura T, Muto H, Yoshizawa K, Kiyosawa K (2005): Assessment of KL-6 as a tumor marker in patients with hepatocellular carcinoma. World J Gastroenterol.; 11(42): 6607-6612.

Giannelli G and Antonaci S (2006): New frontiers in biomarkers for hepatocellular carcinoma. Dig Liver Dis.; 38: 854-859.

Giannelli G, Marinosci F, Trerotoli P, Volpe A, Quaranta M and Dentico P (2005): SCCA antigen combined with alpha-fetoprotein as serologic markers of HCC. Int. J Cancer; 117: 506-509.

Goma A, Khan SA, Leen E, Waked I and Taylor-Robinson SD (2003): Diagnosis of hepatocellular carcinoma. World J of Gastroenterology; 15(11): 1301-1314.

Hirasawa Y, Kohno N, Yokoyama A, Inoue Y, Abe M and Hiwada K (1997): KL-6, a human MUC1 mucin, is chemotactic for human fibroblasts. Am J Respir Cell Mol Biol.; 17: 501-507.

Hussein MM, Ibrahim AA, Abdella HM, Montasser IF and Hassan MI1 (2008): Evaluation of serum squamous cell carcinoma antigen as a novel biomarker for diagnosis of hepatocellular carcinoma in Egyptian patients: Indian Journal of Cancer; 45 (4): 167-172.

Kohno N (1999): Serum marker KL-6/MUC1 for the diagnosis and management of interstitial pneumonitis. J Med Invest.; 46: 151-158.

Lai EC and Lau WY (2005): The continuing challenge of hepatic cancer in Asia. Surgeon; 3: 210-215.

Lau WY and Lai EC (2008): Hepatocellular carcinoma: current management and recent advances. Hepatobiliary Pancreat Dis Int.; 7: 237-257.

Lencioni R, Cioni D, Crocetti L, Franchini C, Pina CD, Lera J (2005): *Early-stage hepatocellular carcinoma in patients with cirrhosis: Long-term results of percutaneous image-guided radiofrequency ablation. Radiology;* ***234***(3): 961–967.

Marrero JA and Lok AS (2004): Newer markers for hepatocellular carcinoma. Gastroenterology; 127: S113-S119.

Moriyama M, Matsumura H, Mikuni M, Arkawa Y, Ohshiro S, Aoki H, Yamagami H, Kaneko M, Shioda A, Saito H, Tanaka N and Arakawa Y (2003): The clinical significance of serum KL-6 levels in patients with type C liver diseases. Hepatol Res.; 25: 385-395.

Pontisso P, Calabrese F, Benvegnu L, Lise M, Belluco C, Ruvoletto MG, De Falco S, Marino M, Valente M, Nitti D, Gatta A and Fassina G (2004):

Overexpression of squamous cell carcinoma antigen variants in hepatocellular carcinoma. Br J Cancer; 90: 833-837.

Pontisso P, Quarta S, Caberlotto C, Beneduce L, Marino M, Bernardinello E, Tono N, Fassina G, Cavalletto L, Gatta A and Chemello L (2006): Progressive increase of SCCA-IgM immune complexes in cirrhotic patients is associated with development of hepatocellular carcinoma. Int Cancer; 735-740.

Sagara M, Yonezawa S, Nagata K, Tezuka Y, Natsugoe S, Xing PX, McKenzie IF, Aikou T and Sato E (1999): Expression of mucin 1 (MUC1) in esophageal squamous-cell carcinoma: its relationship with prognosis. Int J Cancer; 84: 251-257.

Sherman M (2001): Alphafetoprotein: an obituary. J Hepatol.; 34: 603-605.

Soresi M, Magliarisi C, Campagna P, Leto G, Bonfissuto G and Riili A (2003): Usefulness of alpha-fetoprotein in the diagnosis of hepatocellular carcinoma. Anticancer Res.; 23: 1747-1753.

Suzuki K, Takada H, Oka S, Kanouzawa S, Imuro M, Kitazumi Y, Arima T, Ohyama R and Kuwayama H (2003): Clinical significance of KL-6, a marker of interstitial pneumonia, in cases of HCV-associated chronic liver disease. Int Medicine; 42(8): 650-654.

Tanimoto T, Tanaka S, Haruma K, Yoshihara M, Sumii K, Kajiyama G, Shimamoto F and Kohno N (1999): MUC1 expression in intramucosal colorectal neoplasms. Possible involvement in histogenesis and progression. Oncology; 56:223-231.

Trerotoli P, FransveaE, Angelotti U, Antonaci G, Lupo L, Mazzocca A, Mangina A, Antomaci S and Giannelli G (2009): Tissue expression of Squamous Cellular Carcinoma Antigen (SCCA) is inversely correlated to tumor size in HCC. Molecular Cancer; 8: 29.

Uemura Y, Pak SC, Luke C, Cataltepe S, Tsu C, Schick C, Kamachi Y, Pomeroy SL, Perlmutter DH and Silverman GA (2000): Circulating serpin tumour markers SCCA1 and SCCA2 are not actively secreted but reside in the cytosol of squamous carcinoma cells. Int J Cancer; 89: 368-377.

Vauthey JN, Lauwers GY, Esnaola NF, Do KA, Belghiti J and Mirza N (2002): Simplified staging for hepatocellular carcinoma. J Clin Oncol.; 20: 1527-1536.

Wu W, Yao DF, Gu LH, Fan JW, Lu XF and Li XH (2006): Determination of hepatoma-specific alpha-fetoprotein by a mini-column affinity chromatography method. Chin J Clin Lab Sci.; 24: 10-12.

Yao DF, Dong ZZ and Yao M (2007): Specific molecular markers in hepatocellular carcinoma. Hepatobiliary Pancreat Dis Int.; 6: 241-247.

2/12/2012