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RESEARCH ARTICLE

The possible role of serum soluble Endoglin level in the diagnosis of hepatocellular carcinoma in patients with liver cirrhosis

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Manuscript Info

Abstract

Introduction: Hepatocellular carcinoma (HCC) is one of the most common malignant tumors with a high rate of morbidity and mortality. Endoglin (CD105) is an accessory receptor for transforming growth factor beta (TGF-β) and its expression is up regulated in actively proliferating endothelial cells. Endoglin has been suggested to play an important role in tumor-related angiogenesis and neovascularization. **Aim of work:** To evaluate the diagnostic value of circulating serum soluble endoglin in patients with liver cirrhosis with and without HCC and to compare it with other known diagnostic markers of HCC. **Subjects and Methods:** The study included, 15 healthy subjects served as control group (group I), 25 patients with liver cirrhosis without HCC (group II) and 25 patients with liver cirrhosis with HCC (group III). All subjects were subjected to thorough history and clinical examination, abdominal ultrasound and laboratory investigations including ; Liver and kidney function tests, complete blood picture in addition to measurement of serum α-fetoprotein (AFP) and the serum soluble endoglin level. **Results:** The serum levels of soluble endoglin and AFP were significantly increased in hepatic cirrhosis with HCC patients as compared to those without HCC and controls subjects **(p<0.001)**. In addition, there was significant increase of serum levels soluble endoglin in liver cirrhosis without HCC as compared to control subjects while there was no significant difference in AFP levels in the previous two groups. Soluble endoglin levels were significantly positively correlated with ALT and Child Pugh's classes while AFP was significantly positively correlated with ALT only in liver cirrhosis without HCC. The serum levels of soluble endoglin and AFP were significantly positively correlated with ALT and TNM stages in liver cirrhosis with HCC. At a cut off value of 6.9 ng/ml, endoglin had a sensitivity of (72%) and specificity of (80 %) for diagnosis of HCC. While, AFP gave sensitivity of (68%) and specificity of (64%) at a cut off level of 18 ng/ml. The combined endoglin and AFP improve the sensitivity and specificity to (89%) and (85 %) respectively. **Conclusions:** Serum endoglin is highly sensitive and specific for detecting HCC specially if combined with AFP. Therefore, it may play an important role in early diagnosis of HCC.

Introduction

Hepatocellular carcinoma (HCC) is one of the most common malignant tumors with a high rate of morbidity and mortality. HCC affects approximately one million individuals annually worldwide, with the incidence equal to the mortality rate [1]. HCC is one of malignant hypervascular primary hepatocellular lesions [2].

Cirrhosis of any etiology is the major risk factor for HCC [3].About 80% of patients with newly diagnosed HCC have preexisting cirrhosis [4]. Even after comprehensive therapies with surgical excision, chemotherapy, ethanol injection, radiofrequency, or cryotherapy, this tumor shows a high percentage of recurrence and metastasis, and the mean survival of the patients is still short, compared to other major solid tumors. It is assumed that such high vascularity could be one of the reasons responsible for the poor prognosis [5].

Ultrasound (US) is often the first imaging and screening modality used for HCC diagnosis [6]. α-fetoprotein (AFP) measurement and/or US has been proposed for the early detection of subclinical HCC [7].

AFP and pathology detection are commonly used in the clinical early diagnosis of liver cancer. However, the specificity and sensitivity of AFP used in screening for liver cancer are not satisfactory [1]. Though the measurement of AFP serves as an important tool in screening HCC patients, some reports have indicated that it has limited utility of differentiating HCC from benign hepatic disorders for its high false-positive and false-negative rates, and patients with acute exacerbation of viral hepatitis but no HCC may also have markedly increased AFP levels [8].

Greater than 70% of HCC patients have a high serum concentration of AFP because of the tumor excretion [9]. AFP is elevated during pregnancy [10]. AFP also is produced by a variety of tumors including hepatocellular carcinoma, hepatoblastoma, and non-seminomatous germ cell tumors of the ovary and testis[11]. Additionally, AFP elevation has also been recognized in the presence of acute and chronic viral hepatitis as well as in patients with cirrhosis caused by hepatitis C [12].

AFP is the most widely used biomarker for HCC surveillance, which is criticized as neither sensitive nor specific in active hepatitis and liver cirrhosis [13].There is about 30% HCC cases with normal serum AFP levels are hardly diagnosed before any clinical manifestations appear, so, AFP is limited and not efficient for early HCC [14]. Therefore, it highlights the need for new early detection biomarkers more useful and accurate for HCC [15].

Angiogenesis is a multistep process, physiological angiogenesis occurs during liver regeneration, leading to the formation of new blood vessel from pre-existing vasculature, meanwhile pathological angiogenesis occurs in HCC [16]. Angiogenesis makes significant contribution to tumor growth, invasiveness, and metastatic potential of HCC [17].Angiogenesis refers to the sprouting of new blood vessels from pre-existing capillaries. It is a multi-step process involving proliferation of activated endothelial cells, migration of the endothelial cells to reach remote targets, assembly of the endothelial cells into new capillary tubes, followed by the synthesis of a new basement membrane and maturation of vessels with formation of a vascular lumen [18]. The new capillaries formed in tumors have incomplete basement membrane, facilitating penetration of tumor cells into the circulation [19].

Endoglin is a component of the transforming growth factor-β (TGF-β) receptor complex as it binds TGF- β1 and TGF- β3 with high affinity. Endoglin has been reported as expressed by endothelial cells of proliferating capillaries [20]. It is expressed with marked tissue-specificity, predominantly in vascular endothelial cells of tissues undergoing active angiogenesis such as regenerating or inflamed tissue and tumoral stroma [21].In particular, its expression in the stromal vessels of various carcinomas is associated with unfavorable prognosis [22]. Thus, Endoglin has been attracted considerable attention, not only as a biological marker of tumor growth but also as a target molecule for diagnostic and therapeutic application against cancer [23]. In addition, expression of Endoglin along hepatic sinusoids has been reported [24].

Endoglin overexpressed on endothelia of vessels in several human solid malignancies [25] and its overexpression is associated with lower patient survival rates, presence of nodes metastases and distant metastatic disease [26]. Endoglin has therefore been suggested as an appropriate marker for tumor-related angiogenesis and neovascularization. Its roles in the prognosis, diagnosis, and treatment of neoplasms have been discussed previously by **Wikstrom et al.,** [27]. The endothelial cells of neoplasms are more prolific than endothelial cells of normal tissue and thus they express elevated endoglin levels [23].

Angiogenesis, the neoformation of blood vessels from pre-existing microvessels, is essential to numerous physiological and pathological processes, such as cell nourishment, and cancer and ischemic disease progression. This complex process involves remodeling of the extracellular matrix and proliferation and migration of endothelial cells [28]. Vascularization is necessary for tumor growth and metastasis [29]. With insufficient supply of blood, tumor cells will undergo apoptosis/necrosis. Given its distinct tissue distribution and its known functional integration with the TGF-β system, it is not surprising that endoglin is involved in angiogenesis.

Therefore, this study was designed to evaluate the diagnostic value of circulating serum soluble endoglin in patients with liver cirrhosis with and without HCC and to compare it with other known diagnostic markers of HCC either alone or in combination.

Subjects and methods:

This study was carried out on 65 subjects in the gastroenterology unit of the department of internal medical, Faculty of Medicine, Zagazig University hospitals. Our study was observational, cross sectional, analytic, case control study including

patients attending to Zagazig University Hospitals as in-patient or in the outpatient clinic in a period of 6 months during 2013.

Subjects: The included subjects in this study   were divided into three groups:

1- **Group I:** It included 15 apparently healthy volunteers, 7 males and 8 females, matched for age and gender with a mean age value ±SD of (53.7±6.08).
2- **Group II:** It included 25 cirrhotic patients without HCC, 15 males and 10 females. Their ages were between 45-63 years with a mean value ±SD of (54±5.32).
3- **Group III:** It included 25 cirrhotic patients with hepatocellular carcinoma, 17 males and 8 females. Their ages were between 47-67 years with a mean value ±SD of (56.76±5.15). Diagnosis was confirmed by physical examination, laboratory and abdominal ultrasound or computed tomography (CT) scan.

Cirrhotic patients were classified according to the Child Pugh's clinical classification. In group II, 6 patients (24%) were Child class A, 5 patients (20 %) Child B and 14 patients (56%) Child C. In group III, 3 patients (12%) were Child class A, 7 patients (28%) Child B and 15 patients (60%) Child C.

As regard underlying disease, HCV account for 15 patients (60%) of cirrhotic patients and 17 patients (68%) of HCC patients , HBV account for   6 patients(24%) of cirrhotic patients and 7 patients (28%) of HCC patients and both HCV and HBV account for 4 patients (16%) of cirrhotic patients and one patient (4%) of HCC patients .

In HCC 13, patients had single hepatic focal lesion (HFL), 7 patients had two HFLs and 5 patients had multiple HFLs. The right lobe of the liver was affected in 15 patients, the left lobe was affected in 6 patients and both lobes were affected in 4 patients. The average total size of HFLs was 63±34mm and vascular invasion affect 5 patients (20%).

HCC was diagnosed based on at least one of the following criteria in the guidelines of clinical diagnosis and staging for hepatocellular carcinoma [30] :

  i.Hepatic space-occupying lesion with a serum AFP level ≥ 400 ng/ml.
 ii. Hepatic space occupying lesions with arterial phase enhancement and rapid washout in portovenous phase in triphasic CT or magnetic resonance imaging.

HCC was staged according to the TNM staging system [31].

Exclusion criteria: all patients with a prior loco-regional therapy, systemic therapy and/or any surgical intervention (liver resection or transplantation) or who had chronic inflammatory diseases, hematological malignancy and cancer of any organ other than the liver were excluded from the study.

Ethical Clearance: Informed written consents from the patients to participate in the study were taken.

Methods:

All Subjects included in the study were subjected to the following:

- ❖ Full history taking and complete clinical examination.
- ❖ **Routine laboratory investigations including:** Complete blood count, liver function tests, kidney function tests, PT, PC, INR, PTT, blood sugar and viral markers.
- ❖ **Specific laboratory investigations including:**
    - -Serum soluble endoglin level by the quantitative sandwich enzyme immunoassay technique.
    - -Serum α-fetoprotein level by electro-chemiluminescence assay using cobase411 auto analyzer (Roche diagnostics).
- ❖ **Radiological investigations include:** Abdominal ultrasound and Triphasic CT or magnetic resonance imaging.

Statistical analysis:

All data were tabulated and statistically analyzed using SPSS software package version 17. Quantitative data   are presented as means ± SD.   A student t-test was used for comparison of means of two groups. One-way ANOVA test was

used to compare more than two groups. Pearson´s correlation was used for detection of relation between 2 variables. P-value>0.05 indicates non- significant results. P-value <0.05 indicates significant results. ROC (receiver operator characteristic curve) was constructed to evaluate the diagnostic performance of soluble endoglin and AFP in discriminating HCC. Best cut off, sensitivity, specificity, positive and negative predictive values and diagnostic accuracy were calculated in accordance with standard methods.

Results

Table (I): Comparison of mean ± SD of different biochemical parameters between Group I, II and III: There was a statistical significant difference between group I, II and III as regard ALT, AST, alkaline phosphatase, albumin, total bilirubin, direct bilirubin, prothrombin time and concentration, AFP and endoglin. There was a statistical significant difference in group II and III when compared to group I but no significant difference when compared to each other. There is a statistical significant increase in serum endoglin and ALP in patients with liver cirrhosis without HCC and liver cirrhosis with HCC compared to control group. Serum endoglin and ALP were statistically significantly higher in patients of liver cirrhosis with HCC than patients of liver cirrhosis without HCC. In addition, there is statistical significant increase in serum AFP in patients with liver cirrhosis with HCC compared to patients with liver cirrhosis without HCC and control group.

Table (II): Correlation coefficient between endoglin level (ng/ml) and different biochemical parameters in group II and III: Endoglin was significantly positively correlated with ALT and Child Pugh's classes in liver cirrhosis without HCC. There was a significant positive correlation between endoglin level and ALT and TNM stages in liver cirrhosis with HCC.

Table (III): Correlation coefficient between AFP level (ng/ml) and different biochemical parameters in group II and III: AFP was significantly positively correlated with ALT in liver cirrhosis without HCC. There was a significant positive correlation between AFP level and ALT and TNM stages in liver cirrhosis with HCC.

Table (IV): Sensitivity, specificity of AFP and endoglin in diagnosis of hepatocellular carcinoma: At a cut off value of 6.9 ng/ml, endoglin had a sensitivity of (72%) and specificity of (80%) for diagnosis of HCC. However, AFP gave sensitivity of (68%) and specificity of (64%) at a cut off level of 18 ng/ml. The combined endoglin and AFP improve the sensitivity and Specificity to (89%) and (85 %) respectively for diagnosis of HCC.

Table (I): Comparison of mean ± SD of different biochemical parameters between Group I, II and III.

Variable	(Group I) (N = 15) Mean±SD	(Group II) (N = 25) Mean±SD	(Group III) (N = 25) Mean±SD	P value
Total bilirubin (mg/dl)	0.8±0.33	7.5 ±7.2 a	10.5±9.96 a	<0.001*
Direct bilirubin (mg/dl)	0.2 ±0.2	3.5 ±3.5 a	5.8 ±5.6 a	<0.001*
Albumin(g/dl)	3.9 ±0.4	2.7±0.75 a	2.4±0.5 a	<0.001*
ALT (U/L)	26.3±6.7	99.5±104.5 a	124.5±113.3 a	<0.001*
AST (U/L)	24.3±4.9	118±72.8 a	193.8±182.2 a	<0.001*
ALP (U/L)	60.3±20.7	158.5±73.3 a	242.3±103.5 ab	<0.001*
Prothrombin time	13.4±0.2	21.4±6 a	19.7±5.6 a	<0.001*
Prothrombin conc.%	99.6±2.8	47.7 ±18.7 a	51.80±16.1 a	<0.001*
AFP (ng/ml)	7.5±6.7	8.3±6.8	16891.6±18621.5 ab	<0.001*
Endoglin (ng/ml)	4.9±1.2	6.4±2.4 a	9.51±3.3 ab	<0.001*

*= P is significant.

a= significant when compared with control.

b= significant on comparing Group II.

ALT; alanine aminotransferase, **AST;** aspartate aminotransferase, **ALP;** alkaline phosphatase, **AFP;** α-fetoprotein

Table (II): Correlation coefficient between endoglin level (ng/ml) and different biochemical parameters in group II and III.

Parameters	(Group II) N=25		(Group III) N=25	
.	r	p	r	p
AST (U/L)	0.21	0.31	0.11	0.6
ALT (U/L)	0.46	**0.02***	0.49	**0.01***
ALP (U/L)	0.26	0.21	0.23	0.27
Total bilirubin (mg/dl)	0.24	0.25	0.28	0.18
Direct bilirubin (mg/dl)	0.15	0.47	0.30	0.15
Albumin (g/dl)	0.04	0.85	-0.12	0.57
Prothrombin time	0.32	0.12	0.36	0.08
Prothrombin conc.%	0.21	0.31	0.22	0.29
AFP (ng/ml)	0.27	0.19	0.31	0.13
Child Pugh 's classes	0.54	**0.005***	-	-
TNM stages	-	-	0.65	**0.0004***

*= P is significant

Table (III): Correlation coefficient between AFP levels (ng/ml) and different biochemical parameters in group II and III.

Parameters	(Group II) N=25		(Group III) N=25	
	r	p	r	p
AST (U/L)	0.17	0.41	0.37	0.07
ALT (U/L)	0.44	**0.03***	0.50	**0.01***
ALP (U/L)	0.16	0.44	0.2	0.34
Total bilirubin (mg/dl)	0.28	0.18	0.34	0.1
Direct bilirubin (mg/dl)	0.19	0.36	0.27	0.19
Albumin (g/dl)	0.17	0.42	-0.08	0.7
Prothrombin time	0.25	0.23	0.29	0.16
Prothrombin conc.%	0.21	0.31	0.24	0.25
Child Pugh 's classes	0.29	0.16	-	-
TNM stages	-	-	0.59	**0.002***

*= P is significant

Table (IV): Sensitivity, specificity of AFP and endoglin in diagnosis of hepatocellular carcinoma.

Parameter	Sensitivity	Specificity	PP value	NP value	Accuracy
AFP (ng/ml) Cut level 18	68%	64%	65.38%	66.67%	66%
Endoglin(ng/ml) Cut level 6.9	72%	80%	78.26%	74.07%	76 %
Endoglin and AFP	89%	85%	84%	77%	79%

Discussion

The incidence of HCC is rising worldwide being currently the fifth most common cancer and third cause of cancer-related mortality [32]. HCC often develops in patients with underlying liver disease [33].

Our study showed statistical significant difference between group I, II and III as regard ALT, AST, ALP, ALB, total bilirubin, direct bilirubin, prothrombin time and concentration, AFP and endoglin.

Elevated aminotrasferases levels are sensitive for liver injury. ALT and AST are two of the most reliable markers of hepatocellular injury or necrosis [34]. They are typically elevated in all liver disorders, appearing to be more sensitive and specific tests for acute than chronic hepatocellular damage, Increased release, decreased clearance and/or impaired synthesis are all incriminated in their fluctuating levels [35]. In addition if the liver is damaged, the liver cell (hepatocyte) membrane becomes more permeable and some of the enzymes leak out into the blood circulation [34].

Serum bilirubin concentration is a well-established marker of the hepatic synthetic function [36]. Nonalcoholic fatty liver disease (NAFLD) can progress from simple steatosis to nonalcoholic steatohepatitis (NASH) and cirrhosis, which are accompanied with hepatic cellular destruction [37]. It can even develop to HCC [38]. Therefore, when fibrosis progresses, bilirubin increases because of reduced hepatic excretion and less enterohepatic circulation attributable to portal systemic shunt [39].

In addition, hypoalbuminaemia is more common among individuals with chronic liver disease reflecting both severe liver damage and decreased albumin synthesis [40].

Patients with liver cirrhosis are characterized by decreased synthesis of both pro- and anticoagulant factors. Liver damage is commonly associated with impairment of coagulation, when hepatic reserve is poor [41]. Liver failure is accompanied by multiple changes in the haemostatic system, because of reduced plasma levels of procoagulant and anticoagulant clotting factors synthesized by hepatocytes and sinusoidal cells [42]. The decrease of prothrombin concentration in advancing liver fibrosis indicates a damage of liver parenchyma resulting in decreased production of coagulation proteins with increased risk of bleeding tendencies [43].

In this study, serum AFP shows a statistical significant increase in patients of liver cirrhosis with HCC compared to those without HCC and control subjects.

In addition, **Peng et al.,** reported significant increase in AFP level in patients with HCC when compared with cirrhotic patients [44]. HCC patients with a high AFP concentration tend to have greater tumor size, bilobar involvement, massive or diffuse types, portal vein thrombosis, and a lower median survival rate [45] and contribute to vascular invasion and HCC progression and increase risk for early recurrence and poor prognosis after hepatectomy [44].

AFP showed a capability to promote cell growth [46]. The mechanism for the growth and promoting activity of AFP may be due to its synergistic role with other growth factors as insulin like growth factor, vascular endothelial growth factor and platelet - derived growth factor which is believed to exert its effect during cellular proliferation and angiogenesis which makes significant contribution to tumor growth, invasiveness, and metastatic potential of HCC [47].

The present study showed a highly statistical significant increase in serum endoglin in patients with liver cirrhosis with and without HCC compared to control subjects.

Liver cirrhosis is considered as a premalignant state, as about 80 % of HCC is associated with liver cirrhosis [48]. **Clemente et al.,** demonstrated an elevation of circulating endoglin with advancing chronic hepatitis [49]. **Yagmura et al.,** also found that, serum endoglin is significantly elevated in cirrhotic patients compared with control [48]. A possible explanation for the higher endoglin scores in cirrhosis is that endothelial sinusoidal cells acquire a neovessel immunophenotype due to endothelial cell hypoxia, inducible factors of hypoxia, persistent liver injury and hepatic regeneration, all of which contribute to increased endoglin expression [24]. There is evidence that endoglin is expressed in other cells including mesangial, fibroblasts and stellate cells in the liver although it is predominantly expressed in

endothelial cells [50]. **Clement et al.,** reported that stellate cells as well as portal and septal myofibroblasts expressed endoglin and its upregulation was associated with progressive fibrosis in chronic hepatitis patients with HCV infection [49].

In addition, we found a highly statistical significant increase in serum endoglin in liver cirrhosis with HCC group in comparison to control group.

The high endoglin concentration in patients with HCC with underlying cirrhosis result from cirrhosis because of TGF-β upregulation [51] and endoglin expression on activated hepatic satellite cells and additional over expression of endoglin on proliferating endothelial cells [52], whereas angiogenesis occurs which is essential for tumors development and progression.

In addition, in our study we found a significant increase in serum endoglin level in patients of liver cirrhosis without HCC compared to controls subjects.

These results were in agreement with that   obtained by **Elnemr et al.,** who found that a significant increase in endoglin in patients with liver disease compared to controls and in HCC patients compared to cirrhotic ones [53].

During liver cirrhosis, fibrogenesis induces intrahepatic shunts and a barrier between the sinusoids and the hepatocytes, where hypoxia appears. Fibrous pseudo lobes form as a discrete hypoxia unit to induce angiogenesis. Therefore, the cells in cirrhotic liver are under a sustained, mechanically reduced blood flow, which induces angiogenesis in cirrhotic tissues. In addition, non-tumor tissue   might be a hypoxic and highly angiogenic area. In HCC, hypoxic microenvironments with elevated Hypoxia-inducible factor-1 (HIF-1) could up regulate endoglin promoter activity   as a marker of angiogenesis in HCC [54]. Angiogenesis is a proliferative process resulting in the development of new blood vessels from existing endothelial cells and is considered crucial for tumor growth and metastasis [55].

Preativatanyou et al., also, shows   various possible mechanisms could be responsible for the elevation of circulating endoglin. Firstly, production of endoglin in the damaged liver may result in high levels of plasma endoglin. Secondly, elevated endoglin concentrations could be ascribed to an imbalance between endoglin production and endoglin clearance. In advanced biliary atresia stages, reduced endoglin clearance may possibly contribute to increased circulating endoglin levels. Moreover, because other organs apart from the liver can synthesize and secrete endoglin, the major sources of elevated plasma endoglin in the present study could be extrahepatic organs [56].

We found that, serum endoglin and alpha-fetoprotein levels were significantly positively correlated with   ALT in liver cirrhosis with and without HCC.

As, serum ALT routinely serves as a specific biochemical parameter of liver dysfunction reflecting hepatocellular damage [56].

ALT is a well-known marker of inflammatory necrosis in the liver [57]. Persistent inflammation will not only cause necrosis and regeneration of hepatocytes, thereby leading to DNA instability in the hepatocytes and causing the HCC to occur more frequently, but will also enhance the development of intrahepatic metastasis by up-regulating the expression of vascular adhesion molecules [58].

Also, we fund a significant positive correlation between endoglin level and Child Pugh's classes in liver cirrhosis without HCC and there was a significant positive correlation between endoglin level and TNM stages in liver cirrhosis with HCC.

Endoglin levels were found to be correlated to the progression of liver dysfunction and portal hypertension [59]. **Yagmur et al.,** also reported a significant correlation between endoglin level and Child Pugh's classes of cirrhosis. Furthermore, he revealed that circulating endoglin increased significantly in patients with cirrhosis and HCC and was correlated with the severity of hepatic damage [48].

Yang et al., reported a significant correlation between endoglin, microscopic venous invasion and prognosis of patients with HCC. They found that, CD105 is strongly expressed in blood vessels of tumor tissue compared to normal tissues. So, it is considered essential in angiogenesis, which is highly associated with post-operative recurrence and metastasis [60]. Endoglin is essential for angiogenesis, being densely expressed on proliferating endothelial cells and upregulated during hypoxia [61].

So, endoglin as a tumor angiogenesis marker, useful for cancer diagnostics and clinical application. Endoglin expression may be useful as an indicator of disease progression and helpful for estimation of recurrence and metastasis risk [62].

In addition, we reported significant positive correlation between alpha-fetoprotein level   and TNM stages in patients of liver cirrhosis with HCC.

Sheble et al., reported significant statistical correlations between AFP and TNM stage, tumor size, and thrombosis [63]. AFP can stimulate the expression of some oncogenes, which control cell cycle and enhance the proliferation of human HCC [64].

Our obtained results showed that, the best cut off value for endoglin to differentiate HCC and liver cirrhosis groups was 6.9 ng/ml, as the diagnostic sensitivity was 72%, specificity of 80 %, positive predictive value was 78.26%, negative predictive value was 74.07% and diagnostic accuracy was 76%. The sensitivity and specificity of endoglin were more than that demonstrated by AFP.

These findings were in accordance with the results reported by **Yagmura et al.,** and **Salem et al.,** who found the specificities of endoglin in HCC and in liver cirrhosis patients varied between 77.8% and 91.4%, whereas the sensitivities were found between 57.8 % and 84.4 % [48][65].

The combination of both markers in our study improves overall accuracy (79%), sensitivity (89%), specificity (85 %), PPV (84 %), and NPV (77%) in prediction of HCC.

In addition, **Elnemr et al.,** reported that, the sensitivity of combined serum endoglin and AFP was increased for diagnosis of HCC, as the combination of both markers improved overall sensitivity from (70%) to (85%) [53].

In conclusions, serum endoglin is highly sensitive and specific for detecting HCC specially if combined with AFP. It can be used as a complementary biomarker in the diagnosis of HCC. It may have an important role as a risk for development of HCC in cirrhotic patients. It may be also useful as an indicator of risk of recurrence and metastasis oh HCC.

Recommendation:

Further studies on a large number of patients to support the role of endoglin as a diagnostic marker of HCC. Also, follow up of patients to evaluate endoglin as a prognostic marker in HCC.

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