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# Increased IGF-I : IGFBP-3 ratio in patients with hepatocellular carcinoma

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**had a significantly increased IGF-I : IGFBP-3 ratio (0·18 ± 0·05 vs. 0·41 ± 0·09, *P* = 0·04), due mostly to increased IGF-I levels (27·1 ± 5·6 *vs.* 42 ± 6·2 μg/l) as IGFBP-3 levels were similar to patients with cirrhosis (734 ± 81 *vs.* 679 ± 83 μg/l).**

**CONCLUSIONS Hepatocellular carcinoma is associated with a higher IGF-I : IGFBP-3 ratio than that found in patients with liver cirrhosis and a similar degree of liver failure.**

## Summary

**BACKGROUND The development of hepatocellular carcinoma in liver cirrhosis is associated with altered synthesis and secretion of several growth factors.**

**AIM The aim of this prospective study was to investigate the potential implication of IGF-I and its major binding protein (IGFBP-3) in the development of hepatocellular carcinoma.**

**PATIENTS AND METHODS IGF-I and IGFBP-3 were measured in 150 healthy subjects, 40 patients with liver cirrhosis and 63 with liver cirrhosis and untreated hepatocellular carcinoma. The ratio between IGF-I and IGFBP-3 was also calculated.**

**RESULTS Serum IGF-I (70 ± 10 and 65 ± 7 *vs.* 185 ± 6·4 μg/l, *P* < 0·001) and IGFBP-3 levels (1225 ± 113 and 984 ± 67 *vs.* 3017 ± 80 μg/l, *P* < 0·001) were lower in patients with liver cirrhosis, without or with hepatocellular carcinoma, than in controls. Age was negatively correlated with IGF-I levels in patients with liver cirrhosis (*r* = −0·6; *P* = 0·0002) as well as in controls (*r* = −0·8, *P* < 0·0001), but not in patients with hepatocellular carcinoma (*r* = −0·2; *P* = 0·2). Additionally, in patients with liver cirrhosis (*r* = −0·54; *P* = 0·0003) and more weakly in those with hepatocellular carcinoma (*r* = −0·24; *P* = 0·04) IGF-I levels were negatively correlated with liver failure measured according with Child class. Despite patients with class C hepatocellular carcinoma being older than those in the same functional class with cirrhosis (64 ± 2 *vs.* 57 ± 2 years, *P* < 0·01), they**

Liver cirrhosis is one of the predisposing factors leading to hepatocellular carcinoma (HCC) but the mechanisms of hepatocarcinogenesis are still poorly understood. The development of HCC is associated with increased DNA synthesis and a high rate of hepatocyte regeneration observed in cirrhotic liver (Blanc *et al*., 1996).

Abnormalities in growth factor secretion are possibly involved in the promotion and/or progression of tumour growth. Clinical and experimental findings suggested that hepatocyte growth factor (HGF), transforming growth factor alpha (TGF-α) and fibroblastic growth factor (FGF) could act as tumour promoters (Burr *et al*., 1996; Ogasawara *et al*., 1996; Tomiya & Fujiwara, 1996), whereas TGF-β has been shown to be involved in growth suppression (Shirai *et al*., 1994). The role of IGF–IGF binding protein (IGFBP) system has been investigated less.

IGF-I, the main peripheral messenger of GH action, and IGF-II are potent polypeptide mitogens that modulate cell growth, metabolism and differentiation (Jones & Clemmonds, 1995). IGF-II expression is predominant in the fetal development period, whereas IGF-I exerts its activity after birth when the liver becomes its main source (Jones & Clemmonds, 1995). They are noncovalently complexed to specific binding proteins, which, in turn, modulate their biological effects by regulating the bioavailability of IGFs for their target cells and by modulating IGFs interaction with their specific receptors (Jones *et al*., 1995). IGFBP-1 seems to be involved mainly in the insulin-like activity and IGFBP-3 in growth and differentiation. The ratio between IGF and IGFBP seem to modulate these activities.

Several lines of evidence suggest the possible implication of IGFs in the development of cancer. IGF-I has been shown to be a mitogen for cancer cell lines acting as autocrine factor: several human cancers have been reported to express IGF-I mRNA or protein and IGF-I synthesis has been shown within the tumour mass (Huff *et al*., 1986; Tricoli *et al*., 1986; Nakanishi *et al*.,

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1988; Tsai *et al*., 1988; Yee *et al*., 1989). Recently, a tumour-promoting role of IGF-I has been also suggested in subjects with prostate (Chan *et al*., 1998), breast (Hankinson *et al*., 1998), colon (Sohda *et al*., 1996; Ueda & Ganem., 1996; Ma *et al*., 1999; Giovannucci *et al*., 2000) carcinomas as subjects with higher IGF-I levels developed cancers more frequently than those with lower IGF-I levels. The balance between IGF-I and IGFBP-3 more than IGF-I itself has been suggested to be crucial in determining the risk of developing tumour malignancy, with IGF-I associated to growth stimulation and IGFBP-3 to growth suppression through induction of apoptosis (Baserga *et al*., 1999).

While increased expression of IGF-II has been frequently observed in human and experimental HCC (Nardone *et al*., 1996; Yang *et al*., 1996), few data are reported on the IGF-I role in hepatocarcinogenesis: a causal relationship between IGF-I and the development of HCC has been postulated recently (Buzzelli *et al*., 1993), but data on IGFBPs in this tumour type are not currently available. Furthermore, the IGF-I : IGFBP-3 ratio, has been never measured in liver cirrhosis or in HCC.

The aim of this study was to measure circulating IGF-I and IGFBP-3 levels and their molar ratio in patients affected with liver cirrhosis complicated by HCC, in order to analyse potential relationships with hepatic failure, liver involvement by cancer and ageing.

## Patients and methods

### *Patients*

A total of 253 subjects were included in the study: 40 patients had liver cirrhosis (age 25–75 years, median 57 years, male/female = 21/19), 63 had liver cirrhosis with untreated HCC (age 40–75 years, median 63 years, male/female = 33/30) and 150 healthy subjects as control (age 25–75 years, median 55 years, male/female = 78/72). The aetiology of cirrhosis was hepatitis virus correlated in all cases. The diagnosis of cirrhosis was based on clinical features: low count of white blood cells and platelets, prothrombin activity deficiency, oesophageal varices, ultrasound abnormalities of parenchyma (micro- or macro-nodular pattern) and spleno-portal axis. In doubtful cases, diagnosis was determined by liver biopsy. In all HCC patients, the diagnosis was based on ultrasound and histological findings, according to the Edmondson classification (Edmondson & Steiner, 1954). Serum α-fetoprotein level (α-FP) was determined in all patients with and without HCC. All patients were divided according to Child–Pugh classification (Child & Turcotte, 1964; Pugh *et al*., 1973). Criteria for pharmacological treatments and albumin supplementation in the two groups of patients were not different. The Child–Pugh score is a numerical score of certain aspects of liver function that is calculated considering the following variables: serum albumin, prothrombin time prolongation, total bilirubin, and presence of porto-systemic encephalopathy (grade) and ascites. This score defines three classes of patients with different grades of liver function: class A corresponds to preserved liver function, class B to mildly compromised liver function and class C to liver failure. In cirrhotics without or with HCC, class A was attributed to 14 (34%) and 20 (32%), class B to 13 (33%) and 26 (41%) and class C to 13 (33%) and 17 (27%) patients, respectively.

Albumin, cholesterol, prothrombin activity and total bilirubin values of class C patients with or without HCC were considered in order to exclude the possibility that differences in circulating IGF-I and IGFBP-3 levels and their molar ratio may be due to different grades of liver function and not to the presence of neoplasia (D'Arienzo *et al*., 1998). In order to avoid any possible influence of a low diet intake, calories and protein consumption was assessed and in the month before the study a 2500-kCal diet with 35% protein intake was provided for both patients and controls. Adherence to diet was verified weekly. All subjects gave their informed consent to the study. Among the 63 patients with HCC, 25 had monofocal disease with a single nodule, smaller than 5 cm, 12 patients had two nodules in the right lobe, each smaller than 3 cm, 26 patients had multiple nodules in the both lobes or a diffuse infiltration. Portal vein invasion was present in two patients and metastases to skin and bone in another two, all with diffuse infiltration. In 20 patients with multifocal or diffuse disease and in nine patients with mono-bifocal disease, α-FP levels were > 400 μg/l. Renal function was normal in all subjects.

### *Methods*

Blood samples were collected at 08·00 h after a 12-h fasting period, in EDTA-containing tubes rapidly centrifuged, and then stored at –20 °C until used for assay. The methods of determination of plasma IGF-I and IGFBP-3 have been already published (Colao *et al*., 1998). Plasma IGF-I was measured after ethanol extraction by a commercially available immunoradiometric assay (IRMA) kits. The sensitivity of the assay was 0·8 μg/l. The intra-assay coefficients of variation (CV) were 3·4, 3·0 and 1·5% for the low, medium and high point of the curve, respectively. The inter-assay CVs were 8·2, 1·5 and 3·7% for the low, medium and high points of the curve, respectively. Plasma IGFBP-3 was measured by commercially available IRMA kits. The sensitivity of the assay was 0·5 μg/l. The intra-assay CVs were 3·9, 3·2 and 1·8% for the low, medium and high points of the curve, respectively. The interassay CVs were 0·6, 0·5 and 1·9% for the low, medium and high points of the curve, respectively. The IGF-I : IGFBP-3 molar ratio was estimated in order to get a better understanding of the relative concentration changes of IGF-I and IGFBP-3 levels. The values for the molecular mass of IGF-I and IGFBP-3 used for the calculation were 7649 Da and 28 500 Da, respectively (Oscarsson *et al*., 1997).

**Table 1** Circulating IGF-I, IGFBP-3 and IGF-I : IGFBP-3 molar ratio in controls, patients with liver cirrhosis and patients with hepatocellular carcinoma

| | IGF-I levels (μg/l) | IGFBP-3 levels (μg/l) | IGF-I : IGFBP-3 molar ratio |
|---|---|---|---|
| Controls ($n = 150$) | 185 ± 6·4* | 3017 ± 79* | 0·23 ± 0·01 |
| Patients with liver cirrhosis ($n = 40$) | 70 ± 9·9 | 1225 ± 113 | 0·28 ± 0·04 |
| Patients with hepatocellular carcinoma ($n = 63$) | 65·1 ± 7·4 | 984 ± 67 | 0·41 ± 0·04† |

*$P < 0·001$ *vs.* patients with liver cirrhosis and patients with hepatocellular carcinoma; †$P < 0·01$ patients with liver cirrhosis and controls.

### *Statistical analysis*

The statistical analysis was performed by SPSS Inc. (Cary, NC, USA) package.

Correlation analysis, by Pearson and Spearman tests according with normally and non normally distributed variables, was applied to analyse the relationships among age, IGF-I, IGFBP-3 and IGF-I : IGFBP-3 ratio on liver function considered as Child class and disease, including maximal tumour size and number of lesions. The general linear model procedure was used to evaluate whether the slopes of regression lines of IGF-I on age were different among groups. For this procedure, we considered the effects of the mean factor 'group' and 'age' was the covariate. Moreover, the group by age interaction was considered for the analysis. If a significant $F$-value was found the posthoc analysis was performed. The comparison among controls, patients with and without HCC was performed by ANOVA followed by the Newman–Keuls test. The liver function tests were compared in Child C group between patients with and without HCC by Student's $t$-tests. The $\chi^2$ test was applied where appropriate. Data are reported as mean ± SEM. The significance was set at 5%.

## Results

Both circulating IGF-I and IGFBP-3 levels were significantly higher in controls than in cirrhotics with or without HCC, without any difference between these latter groups (Table 1). In contrast, the IGF-I : IGFBP-3 molar ratio was significantly higher in patients with HCC than in those with liver cirrhosis alone and in controls (Table 1). Individual data in all subjects are shown in Fig. 1. Age was negatively correlated with IGF-I levels in patients with liver cirrhosis alone as well as in controls, but not in patients with HCC (Fig. 2). The general linear model procedure demonstrated a significant difference in the regression slopes of IGF-I on age among the three groups of subjects ($F = 12·539$, $P < 0·001$). Additionally, both IGF-I and IGFBP-3 levels were

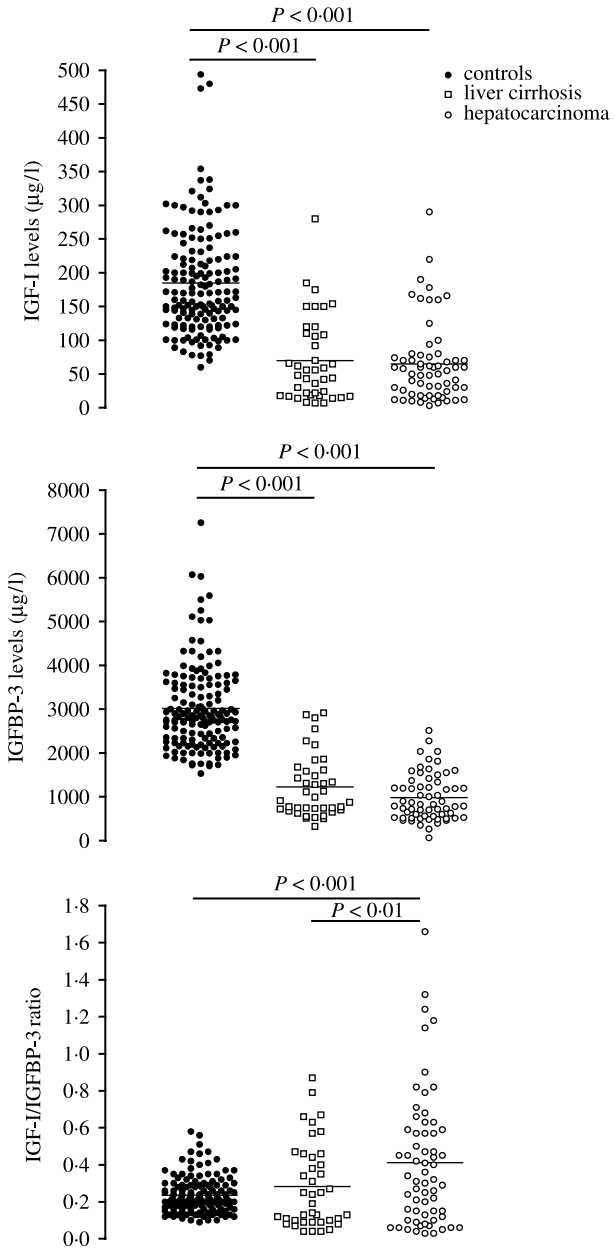


**Fig. 1** Individual data and mean value of IGF-I (top) and IGFBP-3 (middle) levels and IGF-I : IGFBP-3 ratio (bottom) in 150 healthy subjects, 40 patients with liver cirrhosis without HCC and 63 patients with liver cirrhosis and HCC.

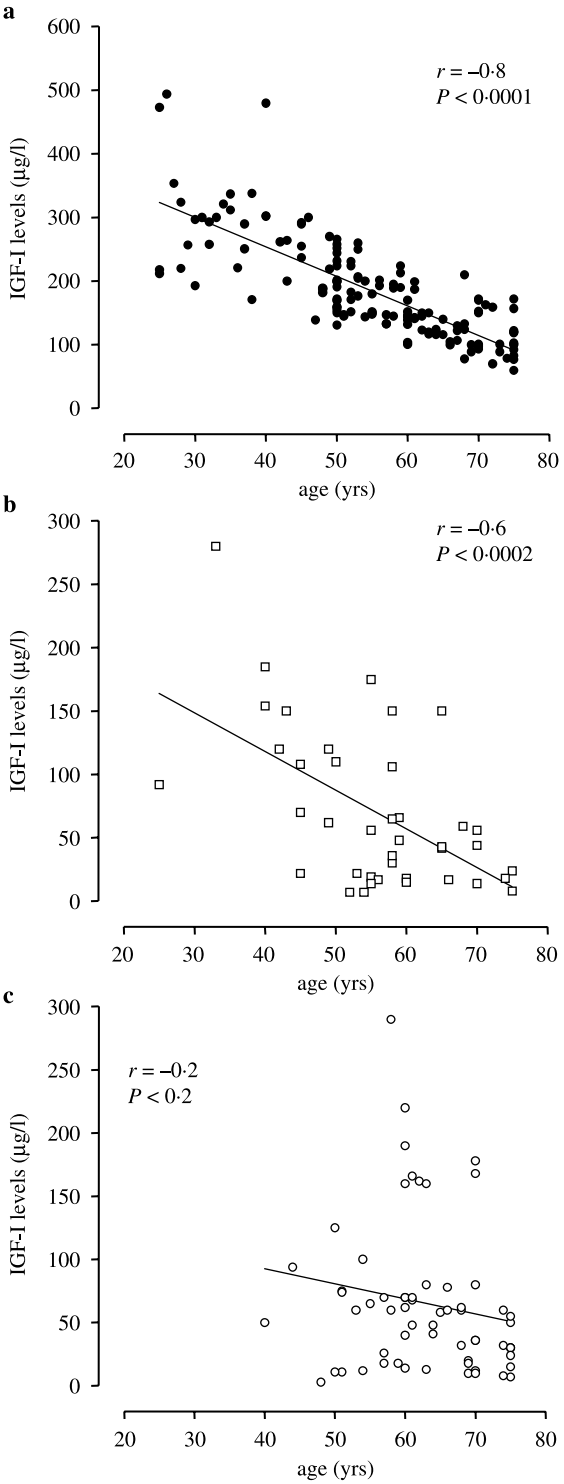


**Fig. 2** Linear correlation between IGF-I levels and age in healthy subjects (a), patients with liver cirrhosis without hepatocellular carcinoma (b) and patients with liver cirrhosis and hepatocellular carcinoma (c).

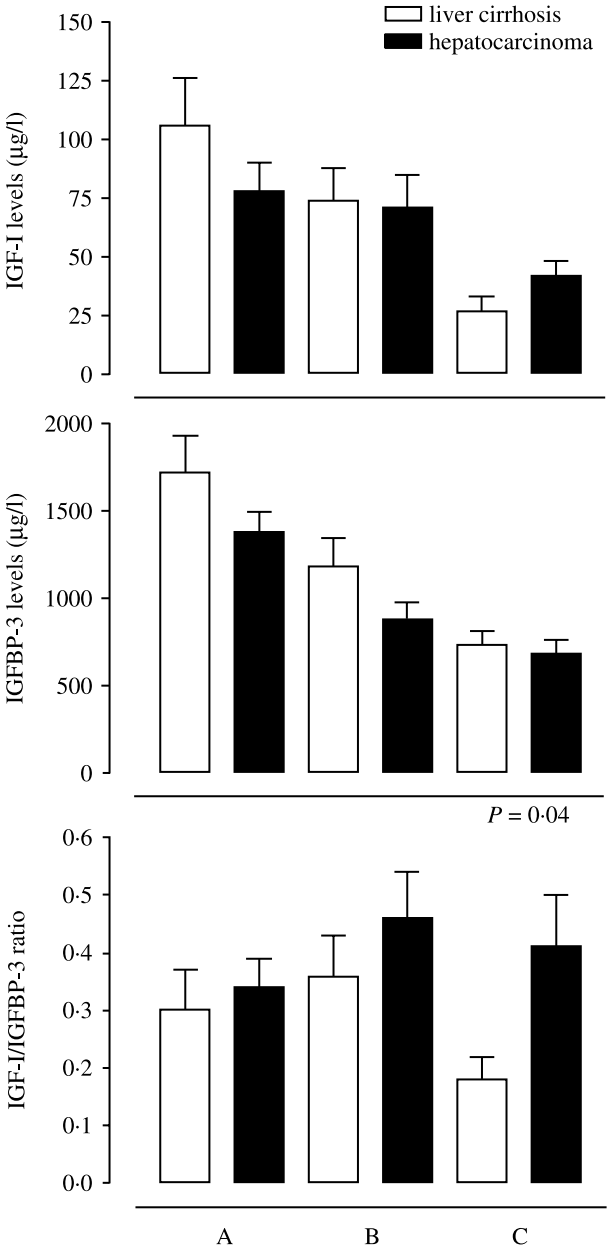


**Fig. 3** Comparison between IGF-I (top) and IGFBP-3 (middle) levels and IGF-I : IGFBP-3 ratio of patients with liver cirrhosis without hepatocellular carcinoma and patients with liver cirrhosis and hepatocellular carcinoma according to Child–Pugh classification, where class A is defined as preserved liver function, class B as mildly compromised liver function and class C as liver failure.

negatively correlated with liver function measured as Child–Pugh class in patients with liver cirrhosis ($r = -0{\cdot}54$, $P < 0{\cdot}001$ and $r = -0{\cdot}57$, $P < 0{\cdot}001$, respectively) and more weakly in those with HCC ($r = -0{\cdot}26$, $P = 0{\cdot}04$ and $r = -0{\cdot}51$, $P < 0{\cdot}01$,

**Table 2** Patient's age, IGF-I and IGFBP-3 levels, IGF-I : IGFBP-3 molar ratio, tumour size and α-fetoprotein levels in patients with hepatocellular carcinoma according to the morphological grading of liver invasion by hepatocellular carcinoma

| | Age (years) | IGF-I levels (μg/l) | IGFBP-3 levels (μg/l) | IGF-I : IGFBP-3 molar ratio | Tumour size (cm) | α-fetoprotein levels (U/l) |
|---|---|---|---|---|---|---|
| Monofocal disease ($n = 25$) | 66 ± 1 | 79·3 ± 13·7 | 1128 ± 117 | 0·43 ± 0·03 | 4·1 ± 0·4 | 313 ± 84 |
| Bifocal disease ($n = 12$) | 60 ± 2 | 83·8 ± 18·7 | 952 ± 160 | 0·53 ± 0·09 | 7·2 ± 0·7 | 362 ± 141 |
| Multifocal disease ($n = 26$) | 62 ± 2 | 44·4 ± 7·8 | 863 ± 89 | 0·34 ± 0·07 | 9·3 ± 0·2 | 500 ± 92 |

respectively). In patients with HCC, IGF-I levels were also weakly correlated with α-FP levels ($r = -0{\cdot}4$, $P = 0{\cdot}001$), tumour size ($r = -0{\cdot}27$, $P = 0{\cdot}03$) and were negatively associated with monofocal, bifocal or multifocal disease ($r = -0{\cdot}26$, $P = 0{\cdot}04$). The IGF-I : IGFBP-3 molar ratio was not correlated to any of the parameters of severity of HCC such as α-FP levels ($r = -0{\cdot}3$, $P = 0{\cdot}056$), tumour size ($r = -0{\cdot}2$, $P = 0{\cdot}06$) or monofocal, bifocal or multifocal disease ($r = -0{\cdot}2$, $P = 0{\cdot}9$). The results of the measurement of IGF-I, IGFBP-3 levels and the IGF-I : IGFBP-3 molar ratio according to Child–Pugh class in patients with liver cirrhosis and in patients with associated HCC are shown in Fig. 3. Despite patients with class C HCC being older than those in the same functional class with cirrhosis alone (64 ± 2 *vs.* 57 ± 2 years, $P < 0{\cdot}01$), they had a significantly increased IGF-I : IGFBP-3 ratio, due mostly to increased IGF-I levels as IGFBP-3 levels were similar to patients with cirrhosis alone (Fig. 3).

In the HCC group, IGF-I and IGFBP-3 levels as well as the IGF-I: IGFBP-3 ratio were not different according to the severity of the disease (Table 2). There was no difference as far as sex and disease aetiology was concerned.

The liver function tests of Child class C patients are shown in Table 3. No differences were found between patients with or without HCC. These data indicate that the differences in IGF-I, IGFBP-3 and IGF-I : IGFBP-3 ratio were not due to different liver function.

In patients belonging to the two groups, no significant differences in the albumin and drug administration were found.

**Table 3** Liver function tests in Child class C patients with and without hepatocellular carcinoma (HCC)

| | Cirrhosis ($n = 13$) | HCC ($n = 17$) | $P$ |
|---|---|---|---|
| Albumin (g/l) | 29 ± 1·2 | 29 ± 0·8 | 0·942 |
| Cholesterol (mmol/l) | 3·08 ± 0·18 | 2·92 ± 0·16 | 0·515 |
| Prothrombin activity (%) | 47·90 ± 2·5 | 52·70 ± 1·9 | 0·131 |
| Total bilirubin (μmol/l) | 63·27 ± 9·41 | 47·88 ± 5·13 | 0·113 |

## Discussion

The results of the current study demonstrated that patients with liver cirrhosis, with or without HCC, have significantly lower IGF-I and IGFBP-3 levels than controls, but their ratio was higher in patients with HCC. Liver failure is confirmed to be a condition reducing IGF-I synthesis and secretion but the presence of HCC seems to be a key factor, as patients with liver failure and HCC had an increased IGF-I : IGFBP-3 ratio compared to those with liver failure and without HCC. This result is even more striking considering the well-known decrease in IGF-I secretion with ageing (Jones & Clemmons, 1995): patients with HCC had higher IGF-I : IGFBP-3 ratio despite being older than those with liver failure. On the other hand, a negative correlation between IGF-I levels and age was well evident in patients with liver cirrhosis as well as in our controls, as expected, but the correlation in patients with HCC was not significant.

The IGF system has important autocrine, paracrine and endocrine roles in the promotion of growth. Alterations of the IGF system have recently been implicated in the pathogenesis of several malignancies (Khandwala *et al*., 2000). IGF-I has been reported to be a mitogen for cancer cell lines, functioning either as an important autocrine factor for stromal tissues or as a paracrine stimulator of epithelial cells (Yee *et al*., 1989). Several human cancers express IGF-I and IGF-I receptor mRNA (Khandwala *et al*., 2000). Furthermore, higher IGF-I levels were reported to correlate with prostate, breast and colon cancer development (Chan *et al*., 1998; Hankinson *et al*., 1998; Ma *et al*., 1999; Giovannucci *et al*., 2000). However, the association between alteration of IGF system, and particularly IGF-I axis, in the development of HCC has never been clearly demonstrated.

The presence of IGF-I mRNA has been demonstrated in 10 human hepatoma cell lines (Tsai *et al*., 1988) and it has been recently reported an increase in IGF-I receptor number in HCC cell lines containing HBV-X, a gene coding for a protein believed as a main cause for the carcinogenicity of hepatitis virus B (Kim *et al*., 1996). Furthermore, insulin receptor substrate-1, the IGF-I endocellular substrate involved in normal hepatocyte growth, was reported to be overexpressed in human HCC (Chuang *et al*., 1996; Tanaka & Ito, 1996; Tanaka & Wands, 1996a, 1996b). These studies on hepatoma cell lines seem to be in contrast to some *in vitro* and *in vivo* studies. In fact, Su *et al*. (1989) analysing seven human hepatoma surgical specimens and the adjacent nontumourous hepatic tissue, found that the tumours

expressed relatively low levels of IGF-I and GH receptor mRNA compared to normal hepatic tissue. The authors proposed that IGF-I was low as a result of reduced GH stimulation of IGF-I synthesis. In line with this experience, in a recent study enrolling 53 men with HCC-positive for hepatitis B and/or hepatitis C virus infections, 20 men with virus-negative HCC, 25 men with virus-negative metastatic liver cancer and 111 virus-negative control men, all patients with HCC had lower IGF-I levels than controls (Stuver *et al*., 2000). However, among patients with HCC, the reduction in IGF-I level was not correlated with the degree of liver damage assessed by prothrombin time and serum albumin level (Stuver *et al*., 2000). In contrast to this study, Buzzelli *et al*. (1993) reported high IGF-I levels in six patients with HCC compared to those with liver cirrhosis without HCC. IGFBP-3 levels, and thus the ratio between IGF-I and IGFBP-3, were not available in these studies.

The role of IGF-I : IGFBP-3 axis on malignancy is still unknown, although some data suggest an increased malignant risk in patients with high IGF-I and low IGFBP-3 activity, as elevated IGF-I bioactivity and activation of the IGF-I receptor are associated with cell proliferation and growth advantage, whereas IGFBP-3 bioactivity promotes an apoptotic advantage (Baserga *et al*., 1999). This evidence suggests that the ratio between IGF-I and IGFBP-3 is more important than the total amount of circulating IGF-I. In this respect, the reduction in IGFBP-3 could have an important role in the pathogenesis of malignancies and possibly HCC. In accordance with this hypothesis, reduced expression of IGFBP-3 was observed in nine out of 12 human HCC (75%; Hanafusa *et al*., 2002). Hypermethylation of the promoter of IGFBP-3 was detected in four out of 12 HCC (33%) although mutations were not identified, while the expression of IGFBP-3 was restored by the demethylating agent 5-aza2′ deoxycytidine in a HCC cell line with promoter hypermethylation (Hanafusa *et al*., 2002). On the other hand, the evaluation of IGF-I and IGFBP-3 levels in HCC could be affected by the well-known evidence that liver diseases profoundly reduce IGF-I and IGFBP-3 levels (Donaghy *et al*., 1995). In fact, serum IGF-I and IGFBP-3 concentrations are significantly lower in patients with chronic liver disease than in controls, as also shown in the current study, while basal and stimulated GH concentrations were reported to be elevated, with a significant association with both liver failure and prognosis (Donaghy *et al*., 1995). Although GH hypersecretion from the pituitary and/or decreased GH clearance might be involved, the low IGF-I levels reflect either a reduction of GH receptors number in the liver or a postreceptor defect (Donaghy *et al*., 1995). Because IGF-I and IGFBP-3 synthesis are strictly correlated, IGFBP-3 is consequently low. The results of our study confirm that not only ageing but also hepatic function determine IGF-I levels. The reduction in parenchymal liver mass was primarily responsible for the low values observed in advanced liver disease (Jones & Clemmons, 1995), as also shown in our series by the association between Child–Pugh class and IGF-I and IGFBP-3 levels.

As far as the role of the IGF-I/IGFBP-3 axis in the development of HCC is concerned, in our series we did not find increased total IGF-I levels in patients with HCC compared to those with cirrhosis, although there was a trend toward an increase in patients with compared to those without HCC, with the same value of liver failure. However, the IGF-I : IGFBP-3 ratio was significantly increased in patients with HCC, either when considered as a whole or when grouped according with Child–Pugh C class. Consequently, our findings did not contrast with an important role of IGF-I in the process of growth of the HCC, as IGF-I biological effects seem to be strictly associated with IGFBP-3 levels.

Our data do not suggest that the determination of plasma IGF-I : IGFBP-3 ratio could further help in the diagnosis of HCC that is fully confirmed by ultrasound and histological findings. However, whether an increase in the IGF-I : IGFBP-3 ratio could predict an early HCC in a patients with liver cirrhosis, is an intriguing possibility. It should also be stated, however, that in our cohort of patients with HCC, we did not find any association between the values of IGF-I : IGFBP-3 ratio and the morphological pattern i.e. tumour size, histology, or extrahepatic diffusion. However, the possibility that an increase in circulating free IGF-I in patients with HCC, probably as a consequence of autocrine secretion by the neoplasm, could be an important factor for progression cannot be ruled out at present. In fact, the hypothesis that the high free IGF-I serum levels could be caused by autocrine secretion by the neoplasm in patients with HCC should be evaluated. Furthermore, the finding that IGFBP-3 reduction is greater in patients with carcinoma than in those with uncomplicated cirrhosis seems to be instrumental for the increase in IGF-I : IGFBP-3 ratio observed in the former group of patients. This observation, however, raises the question of whether a progressive increase in hepatocyte dysfunction occurring from the stage of liver cirrhosis to that of tumoural infiltration could contribute to a change in the IGF-I : IGFBP-3 ratio itself. In fact, expression of IGFBP-3 mRNA has been found in Kupffer cells, rather than in hepatocytes. It seems conceivable that tumour progression from parenchyma to endothelial cells may play a role in altering Kuppfer cell function and IGFBP-3 production (Arany *et al*., 1994). To clarify both these important points, further studies are needed.

In conclusion, our results show higher values of IGF-I : IGFBP-3 ratio in a large series of patients with HCC superimposed to liver cirrhosis than in patients with liver cirrhosis alone, with similar degree of liver dysfunction. Malignancy could be a new condition, with age and liver status, affecting IGF-I and IGFBP-3 levels and IGF-I activity as well. Moreover, an alteration in the IGF-I/IGFBP-3 axis and, particularly, an increase of IGF-I : IGFBP-3 ratio, may play a role in the development and/or progression of HCC.

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