Impact of interferon-alpha therapy on the serum level of alpha-fetoprotein in patients with chronic viral hepatitis

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Abstract

Purpose: Assessment of the long-term effect of interferon-α (IFN-α) treatment on the serum level of hepatocarcinogenesis marker – α-fetoprotein (AFP), in patients (pts) with chronic viral hepatitis (c.v.h.) type B and C.

Material and methods: Thirty seven pts (21 with HCV and 16 with HBV infection; 20 women, 17 men, aged 24--62) were included in the study. The pts were administered IFN-α in the dose of 9-15 MU per week, thrice weekly, for 16 weeks (HBV group) and 24-52 weeks (HCV group). The effectiveness of IFN-α treatment was evaluated on the basis of the HBV DNA and HCV RNA level in the blood. The serum AFP values were determined before and 4-7 years after IFN-α treatment.

Results: The baseline serum AFP level was increased in 26 out of 37 pts (70%) (14/21 from HCV group; 12/16 from HBV group). After the 4-7 years' follow-up it remained elevated only in 2 out of 37 pts (5%). AFP values significantly decreased after IFN-α treatment (17.58±19.09 IU/ml vs 7.95±21.78 IU/ml; $p < 0.05$; normal range 0-5 IU/ml) in both HBV and HCV, responder and non-responder groups.

Conclusions: IFN-α therapy significantly decreases the serum AFP level in patients with chronic viral hepatitis B and C. Its beneficial clinical effects have been observed both in responders and in non-responders. It could diminish the risk of liver carcinogenesis, however further studies are required to elucidate this issue.

Key words: alpha-fetoprotein, chronic viral hepatitis, interferon-alpha, carcinogenesis.

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Received 13.06.2003 Accepted 06.08.2003

Introduction

Hepatocellular carcinoma (HCC) is one of the 10 most common tumors in the world [1]. Hepatitis viruses, particulary the hepatitis B virus (HBV) and the hepatitis C virus (HCV), are major environmental causes of human hepatocellular carcinoma (HCC) world-wide, as they induce chronic liver disease and cirrhotic transformation of the liver [2,3]. These two major aetiological factors have been definitively incriminated in the pathogenesis of HCC. Approximately 300 million people are chronic carriers of HBV and about one percent of the world's population is infected with HCV. HCC for most patients is a terminal complication of chronic inflammatory and fibrotic liver diseases. With regrettably few exceptions, treatment of this neoplasm is largely palliative, and long-term survival is rare. Over one million patients die from HCC annually [4].

The molecular mechanism of HBV and HCV-related liver carcinogenesis is constantly beeing debated. There is evidence for biological effects of proteins of both viruses, which can directly modulate major transduction cellular signals controlling cell proliferation, differentiation and viability, but still the pathogenesis of HCC is poorly understood [5]. Current discussion focuses on the question of whether interferon-α, administered to patients with chronic liver disease (with or without biochemical or virologic response), delays or prevents cancer of the liver. The literature data suggests that interferon--α therapy may prevent hepatocellular carcinoma in patients with cirrhosis, particularly in those with HCV [6-10].

Serological tests (for alpha-fetoprotein) and ultrasonography are used in HCC screening [1]. AFP is α_1-globulin secreted by fetal hepatocytes and in a small amount by other cells of the fetal gastrointestinal tract. Physiologically, in human adults an increased AFP level is present in the serum of pregnant women. In pathology, measurements of alpha-fetoprotein (AFP) are an important tool in the care and management of patients with benign and malignant hepatic disorders. The elevation of serum AFP in benign hepatic diseases (acute and chronic viral hepatitis, toxic liver injury) is associated only with small, transient increases in serum [11-14]. Therefore, a quan-

Table 1. Characteristic of patients with CH B and CH C before IFN-α theraphy.

	CH B (n=16)	CH C (n=21)
Age* (years)	43±14	42±12
Range of age (years)	18-66	18-62
Sex:		
– female	7	13
– male	9	8
Source of infection:		
– medical procedures	13	20
– family contact	1	0
– professional exposure	1	0
– sexual contact	1	0
– unknown	0	1
Acute hepatitis in anamnesis	10	4
Duration of infection		
– (years)*	4±4	6±6
– (range)	(1-18)	(1-16)

* all data are expressed as mean ±SD; CH B – chronic hepatitis type B; CH C – chronic hepatitis type C; IFN-α (interferon-α).

tification of serum AFP has been widely used as a diagnostic marker for HCC. The concentration of AFP correlates with the tumor's size and after an effective surgical procedure it should decrease or even normalize. Measurements of serum AFP levels have also been used in screening the populations at high risk of human HCC such as those with cirrhosis or carriers of HBV and HCV viruses [12,14].

The aim of the study was to evaluate the influence of IFN-α treatment on the serum levels of the hepatocarcinogenesis marker (AFP) in patients with HBV and HCV-related chronic liver disease (CLD). For this purpose we have estimated and compared the serum AFP levels before, after therapy and during a 4-7 year follow-up.

Material and methods

Characteristics of the patients selected for the study. Thirty seven patients (pts), with chronic hepatitis B and C were selected for IFN-α treatment on the basis of the following criteria:

- age – over 18 years;
- duration of the disease before treatment – minimum 6 months;
- markers of active virus replication (in chronic hepatitis B: HBeAg, HBV DNA > 10 pg/ml; in chronic hepatitis C: HCV RNA in the serum or hepatic tissue);
- elevated activity of aminotransferases determined 3 or more times at monthly intervals of time before treatment;
- features of chronic hepatitis in morphologic biopunctate of the liver;
- no features of decompensation of the liver function: prothrombin time – normal or slightly prolonged, albumin levels > 3 g/dl, no signs of ascites or encephalopathy.

The patients were divided into the following groups:

Group I – 16 patients with chronic hepatitis B (CH B), included 7 women and 9 men, age range – 18-66 years.

Table 2. Diseases coexisting with CH in patients treated with IFN-α.

Disease coexisting with CH	CH B (n=16)	CH C (n=21)
Ulcer disease	3	5
Liver haemangioma	1	1
Cholelithiasis	1	2
Diabetes mellitus	0	3
Simple goitre	2	1
Angina pectoris	4	3
Arterial hypertension	2	4
Psoriasis	1	0

* CH B – chronic hepatitis type B; CH C – chronic hepatitis type C; IFN-α – interferon-α.

Table 3. Past diseases in patients treated with IFN-α.

Type of disease	CH B (n=16)	CH C (n=21)
Past surgery :		
Cholecystectomy	2	6
Appendectomy	4	5
Partial gastrectomy	0	1
Mammectomy	1	0
Strumectomy	1	1
Hysterectomy	4	4
Facial skeleton	0	1
Crural varices	0	1
Hodgkin's disease	2	0
Acute lymphocytic leucaemia	1	0

Group II – 21 patients with chronic hepatitis C (CH C), included 13 women and 8 men, age range – 18-62 years.

The characteristics of the patients of both groups are presented in *Tab. 1*.

The diseases coexisting with CH in both groups were determined on the basis of clinical evaluation and anamnesis and are compiled in *Tab. 2*.

At gastroscopy no active ulcer niche was found in any of the patients with coexisting ulcer diseases before IFN-α treatment. The course of the remaining diseases was stable. Two patients with CH B reported past Hodgkin's disease and one – acute granulocytic leukaemia – in all 3 patients a complete remission was observed before IFN-α treatment.

In the CH B group, one patient had earlier undergone mammectomy, another had been underwent to goitre surgery, 4 – operative procedures of the reproductive organs, 2 – cholecystectomy and 4 – appendectomy. In the CH C group, one patient had undergone partial gastrectomy, 6 – cholecystectomy, 4 – operations of the reproductive organs, 1 – goitre surgery, 5 – appendectomy, 1 – facial skeleton surgery and 1 – crural varices operation. The past diseases in both groups are listed in *Tab. 3*.

The patients treated with IFN-α in the HBV and HCV groups did not significantly differ with regard to changes in serum levels of aminotransferases, alkaline phosphatase, γ-glutamyltranspeptidase, bilirubin, albumin, γ-globulin, platelets and prothrombin time. Laboratory findings are shown in *Tab. 4*.

Table 4. Laboratory findings in patients with CH B and CH C before IFN-α theraphy.

	CH B (n=16)		CH C (n=21)	
	Mean	Range	Mean	Range
ALT (U/l)	294±206	57-798	160±109	61-578
AST (U/l)	212±200	27-990	111±63	48-320
AP (U/l)	114±51	48-321	90±28	45-169
GTP (U/l)	124±153	14-820	57±44	10-185
Bilirubin (μmol/l)	15±12	5-72	15±12	7-60
Albumins (g/dl)	4.26±0.48	3.46-5.97	4.40±0.40	3.50-5.56
Gammaglobulins (g/dl)	1.60±0.59	0.90-4.16	1.40±0.50	0.14-2.57
Platelets (G/l)	153±37	82-224	187±49	101-333
Prothrombin time (s)	16±2	12-20	15±2	12-20

Normal range: alanine aminotransferase (ALT): 5-40 U/l, aspartate aminotransferase (AST): 5-40 U/l, alkaline phosphatase (ALP): 35-125 U/l, gamma-glutamyltranspeptidase (GTP): 10-75 U/l, bilirubin: 5-17 μmol/l, albumins: 3.5-5.5 g/dl, gamma-globulins: 0.91-1.48 g/dl, platelets: 100-350 G/l, prothrombin time: 12-16 s.
* CH B – chronic hepatitis type B; CH C – chronic hepatitis type C; all data are expressed as mean ±SD.

Interferon α–2b (IFN-α) (Intron A, Schering-Plough) was administered subcutaneously, 3 x per week in a dose of 9 or 15 MU/week. Since some patients developed marrow suppression, the treatment was periodically discontinued or the dose reduced. The treatment duration was: 16 weeks in CH B patients and 24-52 weeks in CH C patients. After the treatment ended the patients were followed up once a month, the final efficacy of therapy was evaluated after one year. The serum AFP levels were checked every 6 months for 4-7 years following the treatment.

Laboratory tests. At the beginning of the treatment morphology with absolute granulocyte and thrombocyte count was performed after each IFN-α injection. Then these tests as well as alanine and aspartate aminotransferases (ALT, AST), alkaline phosphatase, gamma-glutamyltranspeptidase, bilirubin determinations were done once a week.

The levels of total protein, protein electrophoresis, coagulation parameters (prothrombin time), and renal efficiency (creatinine, urea) were evaluated every month. HBV DNA (using Digene Hybrid Capture System – Murex Diagnostica GmbH, Burgwedel) and HCV RNA (RT PCR) were determined before, in the middle and directly after the treatment ended, then a half and one year later. The serum AFP levels were measured by a RIA – AFP – PROP/J^{125}, MJ^{137}/ test.

Statistics Results are expressed as means±SD. Statistical analysis was performed using the unpaired Student's t-test, Mann-Whitney's U-test, the χ^2 test with Fisher's correction. The significance level was set at $p<0.05$. The statistical analysis was performed using SPSS PC+ software.

Results

The effectiveness of IFN-α treatment was evaluated in both groups directly after therapy and one year later. The lack of serum HBV DNA or HCV RNA one year after the treatment ended was assumed to be a complete and sustained response. Twenty out of 37 patients (54%) achieved a complete and sustained response to treatment (11/16 from the HBV group and 9/21 from the HCV group) and 17 showed no such a response.

The serum AFP levels were monitored every six months for 4–7 years following the treatment. The baseline serum AFP level was increased in 26 out of 37 patients (70%) (14/21 from the HCV group; 12/16 from HBV group).

After the 4–7 year follow–up it remained increased only in 2 out of 37 patients (5%) (1 from the HBV and 1 from the HCV group). The AFP values significantly decreased after IFN-α treatment (17.58±19.09 IU/ml vs 7.95±21.78 IU/ml; $p<0.05$; normal range 0-5 IU/ml) in both HBV and HCV, responder and non-responder groups. These results are presented in *Tab. 5*.

Table 5. Serum AFP levels in patients with CH B and CH C*.

Group of patients	Serum AFP levels (IU/ml)		p
	Before IFN-α	After IFN-α	
All (n=37)	17.58±19.09	7.95±21.78	p<0.05
Responders (n=20)	13.14±11.66	4.35±3.36	p=0.001
Non-responders (n=17)	26.09±25.96	13.92±35.31	p<0.05

* all data are expressed as mean ±SD; AFP: α-fetoprotein; CH B – chronic hepatitis type B;
* CH C: chronic hepatitis type C; IFN-α (interferon-α).

HCC developed after 5 year in one patient, who was a non-responder female with chronic HBV infection, with a baseline AFP level – 24.4 IU/ml and the follow-up AFP level (5 years after the treatment) – 29.42 IU/ml.

Discussion

The treatment of chronic hepatitis B and C remains difficult despite recent progress in this field. Due to prophylactic vaccination, HBV infections can be prevented and the whole situation seems to be under control. On the other hand, the problem of HCV infections, whose diagnosis by specific tests became possible only in 1989, continues to increase. There are about 100 million HCV carriers world-wide [1-3]. It should be stressed that these infections are statistically undetectable as the majority of them are asymptomatic. So the reported numbers show only "the tip of the iceberg".

With IFN-α in the treatment of hepatotropic virus infections there is some hope of fighting the disease. At present, there is no doubt that the drug administered in chronic hepatitis inhibits its progression in some patients [6-10,15]. However, the long-term effects remain highly unsatisfactory.

Strict selection criteria were used in the group of patients analysed by us. Thanks to that 91% of the patients completed their treatment. Similar results were obtained by other authors who used similar selection procedures [15]. All patients with CH B and CH C selected for interferon-α therapy had elevated

aminotransferases activity. As the literature data show in some CH B patients, HBsAg, HBeAg and HBV DNA are likely to be detected with normal or slightly increased levels of hepatic enzymes. Such patients respond poorly or do not respond to IFN-α treatment at all. They should not be selected for therapy but constantly followed up [15].

In our population, the mean ALT activity before treatment was lower in patients with CH C compared with the patients with CH B. Although, even normal activity of this enzyme does not objectively reflect the degree of liver damage in chronic HCV infections. Contrary to patients with CH B, the group with CH C and low aminotransferases activity better respond to IFN-α treatment [15].

The majority of patients treated in our Department received interferon-α in a dose of 5 MU 3 times a week. Some patients had their doses modified as the symptoms of marrow suppression developed, i.e. decreased absolute neutrophil and thrombocyte counts. The efficacy of IFN-α therapy in the examined population was not very satisfactory (54%), particularly in HCV patients (42%), in whom also the number of recurrences within one year following the treatment was higher (compared to the group with HBV infection).

At present, studies are being carried out to determine the role of IFN-α for HCC prevention in patients with HBV and HCV – related chronic liver disease. Our results show that IFN-α therapy could decrease the risk of liver carcinogenesis in these patients. It significantly decreases the serum AFP levels. Its beneficial effect was observed in responders as well as non--responder groups. During the follow-up, HCC was detected in one patient 5 years after completing IFN-α therapy. The patient was a non-responder, female with HBV infection and with increased baseline and follow-up AFP levels. On the basis of these results we recommend hepatocellular cancer screening at frequent intervals for every patient treated with IFN-α after therapy. The fact that there was no HCC development in patients with a complete and sustained response to IFN-α treatment provides hope for this therapy in chronic viral hepatitis. Further studies are required to clarify the role of IFN-α in preventing malignant hepatocyte transformation in patients with HBV and HCV-related chronic liver disease.

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