

Natural leukocyte interferon alpha (Alfaferone) combined with ribavirin in the treatment of patients with HCV-related cirrhosis: our experience

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Abstract

Background The aim of the study was to evaluate the efficacy and safety of combined treatment with natural leukocyte interferon alpha (Alfaferone) plus ribavirin in patients with HCV-related cirrhosis.

Patients and methods Twenty-three patients (15 women, 8 men) aged 17–68 years hospitalized in 2005–2008 were included in the study. Seventeen patients who qualified for treatment were Child-Pugh class A patients and 6 others were class B. Seventeen patients had genotype 1b and 6 genotype 3a infection. Thirteen patients were naïve, re-therapy concerned 8 patients, and in two cases the continuation of treatment had been stopped because of adverse events following the use of pegylated interferons. The treatment was continued for 48 weeks regardless of HCV genotype. Normalized AIAT activity (<40 U/l) was the measure of biochemical efficacy of the treatment, while virological efficacy was reflected by an undetectable viral load in plasma. Both measurements were conducted immediately after the end of treatment (EOT) and after a 6-month follow-up period (SVR). Therapeutic safety was evaluated by the monitoring of the adverse events of the treatment.

Results Abnormal AIAT levels prior to treatment were detected in 20/23 patients. During therapy normalized

levels were achieved in 50% of them, and after 6 months they were sustained in 9/20. EOT was achieved in 6/19 patients and SVR in 3 patients. Mild psychiatric disorders were the most frequently detected adverse events (12 patients). Thrombocytopenia and leucopenia existing prior to treatment did not intensify during the treatment. Severe adverse events caused by the drug resulted in the discontinuation of treatment in three patients (urinary tract infections, depression, myasthenia gravis), of whom two patients were Child-Pugh class A and one was class B. In one patient treatment was discontinued because of HCC.

Conclusion Natural leukocyte interferon alpha is well tolerated by patients with HCV-related cirrhosis and coexisting thrombocytopenia and leucopenia.

Keywords Adverse events · Cirrhosis · Natural leukocyte interferon alpha · Sustained virological response

Introduction

The combination of pegylated interferon and ribavirin is the current standard treatment for chronic HCV infections [1]. Other interferons may be administered when the pegylated interferon therapy is contraindicated or when adverse events of this treatment are present (e.g., cytopenia during the already initiated treatment). This is particularly important in patients with cirrhosis, where neutro- and thrombocytopenia are frequently observed [2]. In clinical practice these problems are considered a criterion disqualifying patients from treatment [1]. In such cases an attempt can be made to treat patients with natural leukocyte interferon alpha, a blend of different interferon types with complementary activity. As a consequence, interferon alpha is the first choice drug in the discussed cases.

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This paper presents our experience in the use of natural leukocyte interferon alpha (Alfaferone) combined with ribavirin in the treatment of patients with HCV-related cirrhosis. This concerned naïve patients, re-therapy, and the continuation of previous treatment that had been discontinued because of adverse events.

Materials and methods

Twenty-three patients with HCV-related cirrhosis treated in the Department of Infectious Diseases and Hepatology in Bydgoszcz, Poland, in 2003–2008 were included in the study. This group comprised 15 women and 8 men aged 17–68 years (average 52 years). The average body mass of patients was 77.6 kg (57.5–114 kg). Characteristics of the patients are presented in Table 1.

Hepatitis C virus RNA was detected based on the results of RT PCR tests. We analyzed the levels of baseline viremia and its kinetics in the course of therapy. We

detected 17 infections with genotype 1b HCV and 6 infections with genotype 3a HCV.

Cirrhosis was diagnosed based on generally accepted clinical, biochemical and histological criteria. Seventeen Child-Pugh class A patients and 6 Child-Pugh class B patients were qualified for the treatment. Classification into Child-Pugh class B was determined by elevated bilirubin levels of up to 2–3 mg/dl (3 patients), reduction of albumin levels down to 3.5–2.8 g/dl (3 patients), prothrombin time elongation (6 patients), and/or ascites (3 patients). Patients with peritoneal fluids present were treated with diuretics until the condition was resolved. Twenty of 23 patients had increased activity of alanine aminotransferase (50–408 U/l, average level 150 U/l). Five patients were diagnosed with esophageal varices, of whom two were treated with beta-blockers and three with endoscopic banding ligation.

Each patient who met the criteria for the therapy was monitored for hepatocellular carcinoma (HCC). Prior to treatment and every 3 months patients were examined by abdominal ultrasound scan, and the level of alpha-feto-protein (AFP) in plasma was measured. When test results were uncertain, diagnostic methods were complemented with abdominal computer tomography (CT). Qualification of patients for therapy included repeated psychiatric consultation during the course of therapy.

The discussed group of patients consisted of 13 naïve patients and 8 patients who were being treated because of ineffective previous treatment. In two cases the treatment was a continuation of previous treatment with pegylated interferon that had been discontinued because of adverse events of the drug treatment (leucopenia with coexisting hyperthyreosis and thrombocytopenia).

Alfaferone (Alfaferone®; ALFA WASSERMANN, Bologna, Italy) was used in a dose of three million units, three times a week, by subcutaneous injection. Ribavirin (Rebetol®; Schering-Plough, Kenilworth, NJ) was administered orally in a daily dose of 800–1,200 mg, depending on body mass. Five of 23 patients were initially treated with ribavirin doses lower than that suggested by their body mass. This was due to starting hemoglobin levels of 10–11 g/dl and was dictated by patient safety considerations. Three out of those five patients were classified into Child-Pugh class A. The treatment was continued for 48 weeks regardless of HCV genotype. Therapeutic efficacy was assessed based on the level of HCV viremia directly after the end of treatment (EOT) and after a further 24 weeks sustained virological response (SVR) and subsidence of the previous biochemical activity of the disease.

Adverse events associated with the treatment were analyzed based on clinical observations and laboratory tests conducted at 2-week intervals. The effect of Alfaferone on thrombocytopenia and leukopenia detected prior to treatment was analyzed.

Table 1 Characteristics of the patient population

Sex (female/male)	15/8
Age (years) (average)	17–68 (52)
Child-Pugh score	
A	17
B	6
Weight (kg) (average)	57.5–114 (76)
Duration of treatment (weeks)	48
First treatment with LeIFN alpha + RBV	13
Continuation with LeIFN alpha after PegIFN 2a/2b	2
Retreatment with LeIFN alpha + RBV	8
IFN preparation	
rIFN 2b	1
rIFN2b + RBV	1
PegIFN 2a + RBV	4
PegIFN 2b + RBV	2
HCV genotype	
1b	17
3a	6
Pretreatment HCV RNA levels (IU/ml) (average)	3.75×10^3 – 1.92×10^7 (3.33×10^6)
Pretreatment AlAT levels (U/l)	
3 patients	Normal
20 patients	50–408 (150)
EOT	6
SVR	3

EOT end of treatment response, SVR sustained virological response, rIFN 2b recombinant interferon alpha 2b, LeIFN alpha natural leukocyte interferon alpha, RBV ribavirin, PegIFN 2a/2b pegylated interferon alpha 2a/2b

AFP level was measured with the chemiluminescence method using the LIAISON AFP test from DiaSorin (reference values up to 5.5 IU/ml) (Humor Diagnostica, South Africa).

The activity of alanine aminotransferase (AIAT) was measured using the ICFF method without pyridoxal phosphate at a temperature of 37°C using a test kit from Roche and the Cobas Integra analyzer. Correct levels were below 40 U/l, regardless of sex.

HCV RNA was detected with RT PCR at the method's linearity of $43\text{--}6.9 \times 10^7$ IU/ml using the COBAS AmpliPrep/COBAS TaqMan HCV test (HCMCAP) reagent from Roche Diagnostic (Indianapolis, IN), while the HCV genotype was detected by hybridization with Linear Array HCV Genotyping Test (LA HCV GT) reagents from Roche.

Platelet and leukocyte levels were measured with the conductometric method on a Sysmex K-4500 analyzer (Sysmex Corp., Japan), and the ratio of leukocyte subpopulations was measured based on blood smear in a microscopic specimen stained using the May-Grünwald, Giemsa method.

Results

Nineteen of 23 patients successfully completed the planned treatment. EOT was achieved in six patients (five Child-Pugh class A patients and one Child-Pugh class B patient). Five patients were infected with a genotype 1b virus and one patient with genotype 3a. Three patients in which a virological response was achieved at the EOT were naïve patients. Another one patient was in a re-treatment group, and in two patients Alfaferone and ribavirin therapy was the continuation of previous treatment with pegylated interferon. SVR was achieved in three patients (all Child-

Pugh class A). All patients were infected with genotype 1b. SVR was achieved in 1/13 naïve patients and in two patients where Alfaferone was used instead of pegylated interferon.

AIAT was normalized during the treatment in 10 out of 20 patients who demonstrated elevated biochemical activity prior to treatment, and in 9 cases it was sustained during a 6-month follow-up period.

Treatment was discontinued in four patients. In three cases this was due to adverse events caused by the drug, and in one patient it was due to diagnosed HCC. The patient with HCC was sent to the transplantation ward where qualification to liver transplant was started. Adverse events disqualifying patients from further treatment included intensified depression, myasthenia gravis and urinary tract infections that led to decompensation of hepatic function in one Child-Pugh class B patient. Other patients were qualified to class A.

Other most frequently recorded adverse events were psychiatric disorders, reflected by impaired mental function, sleep disorders, mood swings, neurasthenia, and depressive syndromes. Two patients were diagnosed with infections: one patient had recurrent labial herpes, and the other had a urinary tract infection. Adverse events frequently coexisted. Most adverse events were observed in Child-Pugh class A patients. Stable angina pectoris was diagnosed in one Child-Pugh class B patient, and depressive disorders were diagnosed in another two Child-Pugh class B patients.

Adverse events and the reasons for premature discontinuation of natural leukocyte interferon alpha treatment are presented in Table 2.

Eight of 23 patients had leucopenia before Alfaferone and ribavirin treatment. None of the five patients with absolute neutrophil counts below $1,500/\text{mm}^3$ required a reduced dose of Alfaferone for longer than 2 weeks (Table 3).

Table 2 Adverse events and reasons for premature discontinuation of treatment with natural leukocyte interferon alpha

Adverse events caused by Alfaferone	Total number of cases/ Child-Pugh score (A/B)	Number of prematurely discontinued treatments (Child-Pugh score)
Psychiatric disorders	12/(10/2)	1 (A)
General weakness	7/(7/0)	–
Flu-like symptoms	3/(2/1)	–
Skin lesions	3/(3/0)	–
Infections	3/(2/1)	1 (B)
Visual disorders	1/(0/1)	–
Angina pectoris, stable	1/(0/1)	–
Hyperthyroidism	1/(1/0)	–
Hair loss	1/(1/0)	–
Myasthenia gravis	1/(1/0)	1 (A)
HCC	1/(1/0)	1 (A)

Table 3 The effect of Alfaferone on leucopenia existing prior to treatment

Patient no.	Initial levels ^a		Minimum level during therapy		Levels in 48th week of treatment	
	Leukocytes ($\times 10^3/\mu\text{l}$)	Neutrophils	Leukocytes ($\times 10^3/\mu\text{l}$)	Neutrophils	Leukocytes ($\times 10^3/\mu\text{l}$)	Neutrophils
1	3.1	2,046	1.8	972	2.3	1,518
2	1.7	697	1.7	697	–	–
3	2.0	1,080	1.9	1,121	2.6	1,898
4	2.1	1,000	2.1	1,000	2.6	1,100
5	2.5	800	1.5	555	2.3	810
6	2.2	1,500	1.2	840	2.9	1,800
7	3.1	2,263	1.6	843	2.1	1,176
8	3.7	2,150	2.9	1,800	3.3	1,500

^a Leukocytes—normal range $4\text{--}10 \times 10^3/\mu\text{l}$

Table 4 The effect of Alfaferone on platelets level existing prior to treatment

Patient no.	Initial platelet levels ($\times 10^3/\mu\text{l}$) ^a	Minimum platelet levels during treatment ($\times 10^3/\mu\text{l}$)	Platelet levels in 48th week of treatment ($\times 10^3/\mu\text{l}$)
1	54	54	82
2	27	27	51
3	62	38	57
4	57	49	59
5	74	74	101
6	53	–	–
7	44	44	55
8	46	32	30
9	53	53	280
10	68	50	50
11	65	64	82
12	51	42	51
13	20	17	18
14	29	24	34
15	86	38	51
16	65	48	69
17	19	17	56
18	53	139	280
19	132	154	219
20	208	142	169
21	100	75	94
22	126	102	151
23	161	85	158

^a Platelets—normal range $150\text{--}350 \times 10^3/\mu\text{l}$

No significant reduction in platelet level was recorded in 17 patients with thrombocytopenia below $90,000/\mu\text{l}$, which would have required modification of interferon dosage (Table 4).

During treatment the dosage of ribavirin was reduced in ten patients because of decreased hemoglobin levels below

10 g/dl and/or clinical symptoms of anemia, decreased tolerance of physical effort, or vertigo.

Discussion

Alfaferone is highly purified interferon alpha containing 18 interferon subtypes derived from human leukocytes [3]. A few research papers on the use of Alfaferone in the treatment of patients with HCV-related cirrhosis have been published so far [4–7].

Patients with cirrhosis are classified as “difficult to treat.” Aghemo et al. in their study emphasized that treatment based on pegylated interferon alpha 2b in patients with cirrhosis infected with genotypes 1, 3, and 4 HCV yielded significantly less SVR than in patients without cirrhosis (genotypes 1 and 4: 17 vs. 36%; genotype 3: 33 vs. 79%). Relapses were observed more frequently in cirrhotic patients (genotypes 1 and 4: 55 vs. 31%; genotype 3: 61 vs. 12%) [8]. Our study confirmed these findings as SVR was achieved in three patients. In addition, factors responsible for treatment failure particularly include high baseline viremia ($>800,000$ IU/ml), body mass over 75 kg, age over 40 years during infection, HIV/HCV coinfection, and previous ineffective treatment [9–11]. These factors, except for HIV infection, frequently coincided in the studied group of patients. Treatment efficacy also depends on ribavirin dose [1, 12]. Some patients from the study group required reduced doses of ribavirin, which may have affected the treatment outcomes.

Alfaferone was tolerated well by patients with hematological disorders, and patients with contraindications against using other interferon types were treated (thrombocytopenia $<90 \times 10^3/\mu\text{l}$, neutropenia $<1,500/\text{mm}^3$). Thrombocytopenia did not intensify significantly during treatment. Even in patients with platelet levels below $30,000/\mu\text{l}$, no clinical symptoms of thrombocytopenia were detected, and in none of them was treatment discontinued

for this reason. These findings conform with reports by other authors and with our previous studies [5, 13–17].

Psychiatric disorders observed during the treatment based on natural leukocyte interferon alpha were mild, except for in one patient, and were successfully managed by psychotherapeutic and pharmacological methods. Infections were not diagnosed in patients with neutropenia.

In the patient who developed HCC, HCV viremia was never undetectable during the treatment. Similar observations were made by Ferrara et al. [18]. Standard therapy with pegylated interferon plus ribavirin is recommended only in Child-Pugh class A patients [1]. Alfaferone can also be used, as demonstrated by our own experience, in class B patients. We did not observe differences in the frequency of adverse events or lower tolerance to treatment between Child-Pugh patients class B and class A. Patients with HCV-related cirrhosis are potential candidates for liver transplant, and for this reason antiviral treatment should be applied prior to surgical procedures [1]. In patients with liver cirrhosis if there is no option for optimal therapy with pegylated interferon plus ribavirin because of cytopenias, for example, the use of natural leukocyte interferon alpha plus ribavirin should be considered.

Conclusions

1. Although the data are very limited (23 patients), treatment with natural leukocyte interferon alpha (Alfaferone) plus ribavirin in patients with HCV-related cirrhosis seems to be safe.
2. Alfaferone administered in patients with thrombocytopenia and neutropenia did not exacerbate these hematological disorders and caused no threat to life.
3. Alfaferone was well tolerated by both Child-Pugh class A and class B patients.

Conflict of interest None.

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