

Efficacy and Safety of In-Asia-manufactured Interferon -2b in Combination with Ribavirin for Therapy of Naïve Chronic Hepatitis C Patients: A Multicenter, Prospective, Open-Label Trial

Nurul Akbar*, Ali Sulaiman*, Irsan Hasan*, Laurentius Lesmana*,
Rino Alvani Gani*, Andri Sanityoso*, Sjaifoellah Noer**,
FX Pridady***, Soemarno****

* Division of Hepatology, Department of Internal Medicine, University of Indonesia
Dr. Cipto Mangunkusumo General National Hospital, Jakarta

** Mitra International Hospital, Jakarta

*** Husada Hospital, Jakarta

**** Sint Carolus Hospital, Jakarta

ABSTRACT

Background: An open-label, multi center and non-comparative study was conducted to evaluate the efficacy and safety of a more affordable in-Asia-manufactured interferon -2b product in combination with ribavirin to treat naïve chronic hepatitis C patients.

Method: Thirty chronic naïve hepatitis C patients were treated with in-Asia-manufactured interferon -2b subcutaneously 3 MIU thrice weekly and ribavirin 800-1,200 mg daily for 48 weeks. Follow-up was done until 24 weeks after the end of treatment. Efficacy was assessed by examining serologic and biochemical parameters at pre and post-treatment. Safety was assessed by evaluating clinical symptoms and laboratory parameters.

Results: The virological response and sustained virological response rates of all Hepatitis C Virus (HCV) genotypes were 83.3% and 76.7% respectively. Post-treatment, 80% patients had significant alanine transaminase (ALT) decreased into normal level and remained normal in 76.7% patients at 24th week follow up period. At that time, the ALT level and sustained virological response were lower in HCV genotypes 1 and 4 than in non-1 and non-4 genotypes. The most frequent adverse event was flu-like syndrome.

Conclusion: The efficacy and safety study on combination therapy of in-Asia-manufactured interferon -2b and ribavirin has shown a good result based on the current standard of interferon alpha and ribavirin combination therapy.

Keywords: interferons, combination drug therapy, chronic hepatitis C, treatment efficacy, safety

INTRODUCTION

Hepatitis C Virus (HCV) infection is the most common chronic blood-borne infection, with an estimation of 170 million people infected worldwide.¹ World Health Organization (WHO) estimates that three

to four million people are newly infected by this virus every year and 70% of them will develop chronic hepatitis.² Approximately, 20% patients with chronic HCV infection will progress to get cirrhosis after an average 20-years period, and 5% will develop hepatocellular carcinoma.³⁻⁵ Hepatitis C is the leading cause of chronic liver disease and liver cancers worldwide, and it is also responsible for two thirds of liver transplants.^{6,7} Mortality due to chronic HCV infection is expected to increase two to threefold over the next 10-20 years. Thus, hepatitis C is considered as an important health care problem which needs serious

Correspondence:

Nurul Akbar
Division of Hepatology, Department of Internal Medicine
Dr. Cipto Mangunkusumo General National Hospital
Jl. Diponegoro No. 71 Jakarta 10430 Indonesia
Phone: +62-21-31900924 Fax: +62-21-3913982 E-mail: akbarnurul25@yahoo.com

attention in addition to its limitation of treatment methods.²

The primary goal of therapy for chronic HCV infection is viral eradication. Previously, treatment of hepatitis C was based on the use of various preparations of interferon alone, and then the treatment developed using combination of interferon with ribavirin. Pilot studies of patients who have relapsed and of previously untreated patients suggest that combining interferon with ribavirin is more effective than using interferon alone,^{8,9} and in a small, placebo-controlled study of previously untreated patients, treatment with interferon and ribavirin for six months was more effective than interferon alone.¹⁰

The current standard of interferon alpha and ribavirin combination therapy, based on several studies of combination therapy with Interferon (IFN) alpha and ribavirin, is demonstrated Sustained Virologic Response (SVRs) of approximately 40%.¹⁰⁻¹³ Currently, pegylated interferon has been used as standard treatment for chronic hepatitis C because some studies found that pegylated interferon

was more effective than conventional interferon for hepatitis C treatment.¹³⁻¹⁵ However, it has been reported that efficacy of most in Western-manufactured interferon -2b was still comparable with the pegylated interferon.^{13,16} Therefore, interferon is still used especially for patients who cannot afford pegylated interferon since the treatment cost of in-Western-interferon is about half or one third of the pegylated interferon.

Since for some population the in Western-manufactured-interferon and pegylated interferon is still considered expensive, therefore, a more affordable product of interferon -2b has been developed in China. This study was conducted to investigate the efficacy and safety of in-Asia-manufactured interferon -2b in combination with ribavirin for treatment of naïve chronic hepatitis C patients.

HYPOTHESIS

We hypothesized that the efficacy and safety of in-Asia-manufactured interferon -2b in combination with ribavirin to treat naïve chronic hepatitis C patients met the current standard of interferon alfa and ribavirin combination therapy.

METHODS

Selection of Subjects

The eligibility criteria were patients aged 18-65 years with positive HCV Ribonucleic Acid (RNA) serum; Alanine Transaminase (ALT) level 1.5 - 10 times of upper normal limit; had never been

treated with any antiviral agents; had normal levels of albumin, bilirubin, creatinine, Thyroid Stimulating Hormone (TSH) and prothrombine time; negative hepatitis B surface antigen (HBsAg), hemoglobin level ≥ 12 g/dL (male) or ≥ 11 g/dL (female), leukocytes count $\geq 3,000/\mu\text{L}$, platelet count $\geq 100,000/\mu\text{L}$, and neutrophils count $\geq 1,500/\mu\text{L}$. Patients with hepatocellular carcinoma, history of moderate to severe psychiatric conditions, seizure disorder, cardiovascular disease, chronic lung disease, diabetes mellitus, hyperuricemic condition, pregnancy, breastfeeding, malignancy, drugs or alcohol abuse, and other conditions, in which compliance and safety issue could not be determined, and those who were unable to practice contraception, were excluded from this study. The study was approved by Ethics Committee of Cipto Mangunkusumo hospital. Before treatment, written informed consent was obtained from each patient and the study was carried out in accordance to the provisions of the Declaration of Helsinki.

Study Design

This multicenter, prospective, open-label uncontrolled trial of efficacy and safety was conducted at four hospitals in Jakarta. The analysis was based on 30 patients who had completed study protocol. The trial began in January 2004, and the trial was completed in October 2005. Duration of treatment was assigned as follows: 12 month-treatment for patients with genotype 1 and 4; 12 month-treatment for patients with genotype 2 and 3 with HCV RNA titer $\geq 2 \times 10^6$ copy/mL, and 6 month-treatment for patients with genotype 2 and 3 with HCV RNA titer $< 2 \times 10^6$ copy/mL. The study medications were given in open-label fashion.

The interferon -2b was administered subcutaneously at the dose of 3 Million International Units (MIU) thrice weekly and ribavirin twice daily orally at a total dose of 800 mg for patients with body weight ≤ 55 kg, 1,000 mg for 56-74 kg of body weight, and 1,200 mg for body weight ≥ 75 kg. The dose of interferon alpha-2b would be reduced to 50% if leukocytes count $\leq 1,500/\mu\text{L}$, neutrophils count $\leq 750/\mu\text{L}$ or platelet count $\leq 50,000/\mu\text{L}$. Ribavirin dose was reduced to 400-600 mg/day if hemoglobin level was < 10 g/dL. Ribavirin treatment was temporarily discontinued if indirect bilirubin level was > 5 mg/dL and would be reinstated at a dose of 400-600 mg/day when indirect bilirubin level was returned to < 2 mg/dL. Full dose of ribavirin would be administered if bilirubin level was < 2 mg/dL for more than 4 weeks. Combination therapy would be discontinued for any of the followings: hemoglobin level ≤ 8.5 g/dL, leukocytes count $\leq 1,000/\mu\text{L}$, neutrophils

Table 1. Trial procedure scheme for 48 week-treatment*

S C R E E N I N G	W-0	W-2	W-4	W-8	W-12	W-16	W-20	W-24	W-28	W-32	W-36	W-40	W-44	W-48	W-4	W-8	W-12	W-16	W-20	W-24	
	Treatment [†]																				
	Laboratory examination [‡]														Follow up [§]						

* For patients with genotype 1 and 4, genotype 2 and 3 with titer > 2 x 10⁶ copy/mL

[†] Interferon alfa 2b subcutaneously injected at the dose of 3 MIU three times/week and heparvir[®] 800 -1,200 mg orally twice daily

[‡] Hemoglobin, leukocytes, neutrophils, platelet, albumin, ALT, AST, bilirubin (direct + indirect), creatinin, uric acid

[§] Hemoglobin, leukocytes, neutrophils, platelet, albumin, ALT, AST, bilirubin (direct + indirect)

■ Qualitative HCV RNA, liver USG, TSH

Table 2. Trial procedure scheme for 24 weeks treatment*

S C R E E N I N G	W-0	W-2	W-4	W-8	W-12	W-16	W-20	W-24	W-4	W-8	W-12	W-16	W-20	W-24
	Treatment [†]													
	Laboratory examination [‡]								Follow up [§]					

* For patients with genotype 2 and 3 with titer < 2 x 10⁶ copy/mL

count ≤ 500/μL, platelet count ≤ 25,000/μL, AL T level increased 2-fold from baseline level or 10 times of upper normal limit, or indirect bilirubin level was > 4 mg/dL for more than 4 weeks.

Hematology parameters were evaluated before treatment, every 2 weeks for the first month and every 4 weeks during and after treatment. Hepatitis C Virus RNA serum and TSH level were measured before treatment, at 24th week and 48th week of treatment, and 24 weeks after the end of treatment. Potential adverse events and all adverse events occurred were tightly monitored and recorded anytime during and within one month after treatment. Therapy would have to be discontinued if life threatening events occurred.

Assessment of Efficacy and Safety

The primary end point was serologic improvement, based on Virologic Response (VR), defined as no HCV RNA detected in blood at the end of therapy, and Sustained Virologic Response (SVR), defined as the absence of serum HCV RNA by 24 weeks after treatment was completed. Secondary endpoint was Biochemical Response (BR), which was defined as the ALT level normalization at the end of treatment. Breakthrough was defined when HCV RNA serum was undetectable at 24th week of treatment, but became positive at the end of treatment. Safety was assessed by clinical symptoms and laboratory parameters.

Statistical Method

Sample size was determined based on reference by McHutchinson et al,¹¹ that indicated virologic response rate after treatment with interferon and ribavirin for 24 weeks was 31 percent. VR and SVR were presented descriptively. The biochemical responses before and after treatment were analyzed using Student t-test.

RESULTS

Characteristics of the Patients

Genotypes of HCV in this study were 1, 2, 3, 4, and 10. Baseline mean viral load in this study (0.82 ± 1.02 x 10⁶ IU/mL) categorized as high (expressed in IU/mL and it was categorized as high when the viral load more than 800,000 IU/mL).

Virologic Response Rate

At the end of treatment, virological response rate in HCV genotype non-1 and non-4 (100%) were higher than genotype 1 or 4 (78.3%). Virological response rate of all HCV genotypes was 83.3% (table 4).

Sustained Virological Response Rate

At 24th week of follow up, we evaluated the sustained virological response rate . The genotype of non-1 and non- 4 (100%) were higher than genotype 1 or 4 (69.6%). Sustained virological response rate of all HCV genotypes was 76.6%. There were no breakthrough cases during the study (table 4).

Table 3. Baseline characteristics of the patients

Characteristic	Mean \pm SD	Number (%)
Age (years)	39 \pm 14	
Sex		
Female		8 (26.7)
Male		22 (73.3)
Body mass index (kg/cm ²)	23.74 \pm 4.47	
Viral load (x 10 ⁶ IU/mL) - (high)	0.82 \pm 1.02	
ALT (U/L) (1.5-10 times > normal)	102.77 \pm 53.66	
HCV genotype 1 or 4		23 (76.6)
1		14 (46.7)
4		9 (30)
HCV genotype (non 1 and non 4)		7 (23.3)
2		3 (10)
3		2 (6.7)
10		1 (3.3)
Undetermined		1 (3.3)
Source of infection		
Drug abuse		11 (36.7)
Transfusion		7 (23.3)
Operation		6 (20)
Unknown		6 (20)

Biochemical Response Rate

The ALT level decreased significantly since the fourth week of therapy (102.77 \pm 53.66 vs. 36.43 \pm 26.02; $p = 0.000$) (table 5). Serum ALT levels at the end of treatment were normal in 80% patients with all HCV genotype and remained normal in 76.67% patients throughout follow up period at 24th week. At the end of therapy, BR rate was higher in patients with genotypes non-1 and non-4 as compared to the types 1 and 4 (85.7% vs. 78.2%). At 24th week of follow up, BR rates also were higher in patients with genotypes non-1 and non-4 as compared to the types 1 and 4 (85.7% vs. 69.6%, respectively). Specifically, the BR at 24th week follow-up period was higher in type 4 (77.8%) than in type 1 (71.4%).

Safety

There was no drop-out case. Ten patients (33.3%) met the criteria for dose reduction or drug discontinuation. Six of them had anemia, three had increased direct bilirubin level, and one had neutropenia. However, only eight of them were considered for dose adjustment or temporary treatment discontinuation. Mean hemoglobin level decreased significantly at 4th week of treatment. One patient had hemoglobin level below 8.5 g/dL, leading to adjustment of ribavirin dose while interferon α -2b was reinstated at full dose.

There were no significant changes in platelet count during this study. Significant decrease of leukocytes count was observed but not followed by dose adjustment or discontinuation. One patient showed neutrophil count below 750/ μ L and required a dose reduction of interferon α -2b.

Direct bilirubin level increased significantly between the 2nd and 12th week of treatment, and three patients (10%) showed a level of more than 2.5 times of upper normal limit. Two of them did not require dose adjustment and the direct bilirubin level returned to normal at following months; meanwhile, both drugs were temporarily discontinued in one patient due to jaundice observed at second and third week, and then full dose of combination therapy was reinstated at the fourth week. There was statistically but not clinically significant change on albumin level during treatment. The TSH level increased above normal level during treatment in one patient. Flu like syndrome was the most symptoms observed during treatment 100%. Other symptoms were similar to previous reports of

Table 4. Virologic response and sustained virologic response

Variable	n	Number (%)		
		VR - 24 week	VR - 48 weeks	SVR - 24 weeks
Age (years)				
< 50	20	7 (85)	17 (85)	15 (75)
> 50	10	8 (80)	8 (80)	8 (80)
Sex				
Male	22	19 (86.4)	19 (86.4)	18 (81.8)
Female	8	6 (75)	6 (75)	5 (62.5)
Genotype				
1 or 4	23	18 (78.3)	19 (86.4)	16 (69.6)
1	14	11 (78.6)	11 (78.6)	10 (71.4)
4	9	7 (77.8)	7 (77.8)	6 (66.7)
Non 1 & non 4	7	7 (100)	7 (100)	7 (100)
All	30	25 (83.3)	25 (83.3)	23 (76.7)
Viral load (U/mL)				
< 800.000	19	17 (89.5)	17 (89.5)	17 (89.5)
> 800.000	11	8 (72.7)	8 (72.7)	6 (54.5)
IDUs	11	9 (81.8)	9 (81.8)	8 (72.7)
With VL < 800,000	5	5 (100)	5 (100)	5 (100)
With VL > 800,000	6	4 (66.7)	4 (66.7)	3 (50)

Table 5. Hematological examination, ALT, albumin, direct bilirubin level

Variable	Hemoglobin (g/dL)		Leukocyte ($10^3/\mu\text{L}$)		Platelet ($10^3/\mu\text{L}$)		ALT (U/L)		Albumin (g/dL)		Bilirubin (mg/dL)	
	Mean \pm SD	p*	Mean \pm SD	p*	Mean \pm SD	p*	Mean \pm SD	p*	Mean \pm SD	p*	Mean \pm SD	p*
Baseline	14.6 \pm 1.6		6.6 \pm 1.9		212.0 \pm 67.9		102.77 \pm 53.66		4.3 \pm 0.2		0.25 \pm 0.1	
Week 2	14.2 \pm 1.5	0.124	5.7 \pm 1.6	0.001	213.2 \pm 66.4	0.825	56.10 \pm 48.39	0.000	4.2 \pm 0.2	0.023	0.38 \pm 0.2	0.000
Week 4	13.3 \pm 1.8	0.000	5.6 \pm 2.4	0.200	225.3 \pm 76.3	0.067	36.43 \pm 26.02*	0.000	4.3 \pm 0.3	0.103	0.33 \pm 0.2	0.004
Week 12	12.7 \pm 1.8	0.000	4.8 \pm 1.3	0.000	260.2 \pm 169.7	0.162	33.73 \pm 43.93 *	0.000	4.4 \pm 0.3	0.385	0.30 \pm 0.1	0.015
Week 24	12.7 \pm 1.7	0.000	4.4 \pm 1.3	0.000	202.2 \pm 73.8	0.176	30.53 \pm 33.28 *	0.000	4.4 \pm 0.2	0.301	0.26 \pm 0.1	0.570
Week 48	12.7 \pm 1.8	0.000	4.4 \pm 1.4	0.000	201.9 \pm 74.5	0.161	30.33 \pm 31.92	0.000	4.3 \pm 0.3	0.078	0.27 \pm 0.1	0.375
Follow up w-24	14.7 \pm 1.4	0.378	7.0 \pm 2.0	0.183	225.5 \pm 65.6	0.087	40.9 \pm 54.4	0.000	4.5 \pm 0.2	0.016	0.19 \pm 0.1	0.013

* Student t-test, compare to baseline 95% confidence interval

other studies on interferon, such as abdominal discomfort, dermatologic symptoms, sore throat and psychiatric symptoms.

DISCUSSION

The current standard of interferon alpha and ribavirin combination therapy, based on previous several studies using combination therapy with in-Western-manufactured interferon alpha and ribavirin, is demonstrated SVRs of approximately 40%.¹⁰⁻¹³ Patients characteristics in this study were similar to the prior trials in the United States.^{11,17,18} The proportion of patients with HCV genotype 1 or 4 (76.6%) was almost similar to the proportions in the prior trials (72%).^{11,17,18} It was equal to McHutchinson et al,¹¹ trial which had approximately 5 to 6 $\times 10^6$ copies/mL (expressed in copies/mL, categorized high if viral load more than 2 $\times 10^6$ cop-ies/mL). Average age of patients in this study (39 \pm 14 years old) was slightly younger than average patients age in McHutchinson et al trial (44 \pm 8 years old).¹¹ Mean body mass index was the same as that in McHutchinson et al trial¹¹, which was categorized as normal body mass index. ALT level was the same as patients in McHutchinson et al trial¹¹, i.e. 1.5 - 10 times upper normal limits.

Our study with similar baseline patients characteristic and treatment procedure with that studies¹⁰⁻¹³ showed SVR rate 76.7%. The SVR rate from our study showed good result if we compare to SVR rate from other previous similar studies.¹⁰⁻¹³ In this study, the virological response at the end of treatment was 83.3%. Virological response rate in HCV genotype non-1 and non-4 (100%) were higher than genotype 1 or 4 78.3% as well as sustained virological response rate in genotype non-1 and non-4 (100%) were also higher than genotype 1 or 4 (69.6%).

SVR from other trial with different characteristic of patients, such as in chronic hepatitis C patient with normal ALT level showed SVR of 32% with this

combination treatment.¹⁹ In Japanese population setting, but in relapsed case after previous interferon therapy, treatment with other interferon α -2b regimen in combination with ribavirin produced virological response of 75% and SVR 58.3% at the end of treatment and at 24th week of follow up respectively.²⁰ We found that SVR were higher in HCV type non-1 and non-4 than in types 1 and 4. These findings are in accordance with other studies.^{13,14,15,19,20} Interferon therapy was more efficacious in types 2 and 3 patients as compared with types 1 and 4. In our study, compared to the type 4, SVR rate in type 1 was higher. This result was in contrast with other studies, which reported that SVR rate for genotype 1 was lower than SVR for genotype 4.^{13,21} The VR at 24th week and 48th week of treatment were higher in patients aged below 50 years old (85% and 85%, respectively) and also in those with lower viral load (< 800,000 U/mL). In the study by Manns et al,¹³ and Carrat et al,²² also showed the benefit of pegylated interferon α -2b over interferon α -2b, which was not seen in patients infected with HCV genotype 2 or 3. The benefit of peg-interferon α -2b over interferon α -2b was most

apparent in patients with genotype 1 or 4.^{13-15,22}

The predominant HCV genotypes in Indonesian donor population were 1b (57.8%), 2a (17.2%), and 3b (10.9%).²³ The most frequent genotype in our study was also genotype 1. For risk factor of HCV infection, previous studies have shown that the main risk factor for infection by genotypes 1a and 3a is intravenous drug use and the main risk factor for infection by genotypes 1b and 2 is blood transfusion.²⁴ HCV infections transmission from intravenous drug use is rising.²⁴ Our study also showed that the most frequent HCV infection transmission was drug abuse.

Biochemical response at the end of treatment and 24th week of follow up were 80% and 76.7% respectively. Compared to our reference study, McHutchinson et al,¹¹ showed lower biochemical response, i.e. 34% at 24th week after treatment.

In the other study²⁵ with same population setting (Indonesian), but in relapsed case after previous interferon therapy, BR at the end of treatment and at 24th weeks after treatment was 75% and 75% respectively. Biochemical responses were higher in HCV type non-1 non-4 than in types 1 and 4.

Many factors has been defined in predicting long-term response of interferon therapy such as sex, age, body mass, index, interferon dose, duration of treatment, viral RNA level, and liver histology.¹⁶ In this trial, the suspected predictor factors were HCV genotype, viral load, age, and body mass index.

Hematological changes occurred during treatment and follow up period were anemia and neutropenia. Increased bilirubin level is also reported in other previous trials. Anemia and neutropenia in this case may be related to ribavirin. Ribavirin administered at doses of 1,200 - 1,600 mg per day can cause anemia and reduced T cells; whereas neutropenia was mostly related to interferon. Others symptoms occurred in this study were similar to the potential adverse events of interferon alpha/pegylated interferon which has been previously reported.^{10-15,19} There were no serious adverse event occurred in this trial. In several study, the adverse effect profile appears to be similar between interferon α -2b and pegylated interferon α -2b, although the frequency of certain adverse effect may vary. In the study done by Carrat et al,²² peg-interferon α -2b and interferon α -2b showed similar tolerability although dose modification for clinical and biological events were more frequent with peg-interferon. Cost analysis for this in-Asia-manufactured interferon α -2b also have showed lower treatment cost (1 : 2 compare to in-Western-manufactured interferon α -2b; 1:6 compare with pegylated interferon α -2b).²⁶

CONCLUSION

In conclusion, the present study demonstrates that the efficacy and safety of combination therapy of this in-Asia-manufactured interferon α -2b and ribavirin showed good result if we compared to other study of in-Western-manufactured interferon combined with ribavirin with similar baseline patients characteristics and treatment procedure. Therefore, this in-Asia-manufactured interferon α -2b can be used as an prospective alternative for chronic hepatitis C treatment.

ACKNOWLEDGEMENTS

We thank to all hepatologist who had given great support for this trial and Innogene Kalbiotech for the scientific Grant and drug trial support (Kalferon) for this study. We also thank Inge Ade Krisanti MD, Yayah

Noegroho MD and Elsa Hufaidah MD for their assistances in this study. We acknowledge Galih MD, Christina Sitorus MD, DR. Vita Kurniati Lubis and Vinci Lorensia MD for their contribution in compiling the data.

REFERENCES

1. Yen T, Keeffe EB, Ahmed A. The epidemiology of hepatitis C virus infection. *J Clin Gastroenterol* 2003;36:47-53.
2. World Health Organization (WHO). State of the art of vaccine research and development 2005: Chapter 8 Viral cancers. WHO 2005.p.81-8.
3. Herrine SK. Approach to the patient with chronic hepatitis C virus infection. *Ann Intern Med* 2002;136:747-57.
4. Hoofnagle JH. Course and outcome of hepatitis C. *Hepatology* 2002;36(5 Suppl 1):S21-9.
5. Seeff LB. Natural history of chronic hepatitis C. *Hepatology* 2002;36(5 Suppl 1):S35-46.
6. Gow PJ, Mutimer D. Regular review—treatment of chronic hepatitis. *British Med J* 2001;323:1164-7.
7. Berenguer M, Wright TL. Hepatitis: Advances in antiviral therapy for hepatitis. *Lancet* 1998;19-26:352 (Suppl 4):SIV15.
8. Schvarcz R, Yun ZB, Sonnerborg A, Weiland O. Combined treatment with interferon α -2b and ribavirin for chronic hepatitis C in patients with a previous non-response or non-sustained response to interferon alone. *J Med Virol* 1995;46:43-7.
9. Schalm SW, Hansen BE, Chemello L, et al. Ribavirin enhances the efficacy but not the adverse effects of interferon in chronic hepatitis C: metaanalysis of individual patient data from European centers. *J Hepatol* 1997;26:961-6.
10. Reichard O, Norkrans G, Fryden A, Braconier JH, Sonnerborg A, Weiland O. Randomized, double-blind, placebo-controlled trial of interferon α -2b with and without ribavirin for chronic hepatitis C. *Lancet* 1998;351:83-7.
11. McHutchison JG, Gordon SC, Schiff ER, et al. Interferon α -2b alone or in combination with ribavirin as initial treatment for chronic hepatitis C. *N Engl J Med* 1998;339:1485-92.
12. Poynard T, Marcellin P, Lee SS, et al. Randomized trial of interferon α -2b plus ribavirin for 48 weeks or for 24 weeks versus interferon α -2b plus placebo for 48 weeks for treatment of chronic infection with hepatitis C virus. *Lancet* 1998;352:1426-32.
13. Manns PM, McHutchinson JG, Gordon SC, et al. Peg interferon α -2b plus ribavirin compared with interferon α -2b plus ribavirin for initial treatment of chronic hepatitis C: A randomized Trial. *Lancet* 2001;358:958-65.
14. Fried MW, Shiffman ML, Reddy KR, et al. Peg interferon α -2a plus ribavirin for chronic hepatitis C virus infection. *N Engl J Med* 2002;26;347(13):975-82.
15. Hadziyannis SJ, Sette H Jr, Morgan TR, et al. Peg interferon- α 2a and ribavirin combination therapy in chronic hepatitis C: A randomized study of treatment duration and ribavirin dose. *Ann Intern Med* 2004;140(5):346-55.
16. Alfaleh FZ, Hadad Q, Khuroo MS, Aljumah A, Algamed A, Alashgar H, Al-Ahdal MN, Mayet I, Khan MQ, Kessie G. Peg interferon α -2b plus ribavirin compared with interferon α -2b plus ribavirin for initial treatment of chronic hepatitis C in Saudi patients commonly infected with genotype 4. *Liver International* 2004;24(6):568-74.
17. Mutchnick M, Tamburro C, Hassanein T, et al. Analysis of HCV RNA concentrations by a multi-cycle RT-PCR assay in 3538 untreated chronic HCV patients: Assessment of RNA

- titer by HCV genotype. *Hepatology* 1997;26(Suppl):186A. Abstract.
18. Bukh J, Miller RH, Purcell RH. Genetic heterogeneity of hepatitis C virus: Quasispecies and genotypes. *Semin Liver Dis* 1995;15:41-63.
 19. Ira M, Jacobson, Furqaan Ahmed, Mark W Russo. Inter-feron alpha-2b and ribavirin for patients with chronic hepatitis C and normal ALT. *Am J Gastroenterol* 2004;99:1700-05.
 20. Rumi MG, De Filippi F, La Vecchia C, Donato MF, Gallus S, Del Ninno E, Colombo M. Hepatitis C reactivation in patients with chronic infection with genotypes 1b and 2c: A retrospective cohort study of 206 untreated patients. *Gut* 2005;54:402-06.
 21. Zein NN. Clinical significance of hepatitis C virus genotype. *Clin Microbiol Rev* 2000;13(2):223-35.
 22. Carrat F, Bani-Sadr F, Pol Stanislas, et al. Pegylated interferon α -2b vs. standard interferon α -2b, plus ribavirin for chronic hepatitis C in HIV-infected patients: A randomized controlled trial. *JAMA* 2004;292:2839-48.
 23. Inoue Y, Sulaiman HA, Matsubayashi K, et al. Genotypic analysis of hepatitis C virus in blood donors in Indonesia. *Am J Trop Med Hyg* 2000;62(1):92-8.
 24. Theodore Sy, Mazen Jamal M. Epidemiology of Hepatitis C Virus (HCV) infection. *Int J Med Sci* 2006;3:41-6.
 25. Arase Y, Ikeda K, Tsubota A, Suzuki Y, et al. Efficacy of interferon retreatment after relapse for chronic hepatitis C patients with biochemical response after first interferon therapy. *J Gastroenterol* 2004;39(5):455-60.
 26. El-Zayadi AR, Attia M, Barakat EMF, et al. Response of hepatitis C genotype-4 naive patients to 24 weeks of Peg-interferon- α 2b/ribavirin or induction-dose interferon- α 2b/ribavirin/amantadine: A non-randomized controlled study. *Am J Gastroenterol* 2005;100:2447-52.