Evaluation of ribavirin efficacy and tolerance in subjects with chronic hepatitis C virus infection

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Background. Reports by hepatologists indicated that anti-HCV antibodies might be detected in 71% to 84% of cases of post-transfusion hepatitis and in up to 50% of cases of sporadic non-A non-B hepatitis. Anti-HCV antibodies were detected in 0.05–1% of blood donors with normal alanine transaminase (ALT) levels and negative anti-HBc screening. Anti-HCV antibodies were found in 67% of patients with a history of intravenous drug abuse or autoimmune hepatitis, and in 10–30% of patients with hepatocellular carcinoma. This indicated that hepatitis C virus was a major cause of the acute and chronic hepatitis throughout the world. Methods. This was a multicenter, international, double-blind, randomized, placebo-controlled study. After the eight-week screening period, patients were randomized to receive daily ribavirin 1200 mg or placebo, during the 48-week treatment period. Follow-up observations were performed during a 16 week post-treatment period. Up to 80 male and female outpatients with mild to moderate chronic active hepatitis C virus infection were enrolled in this study. Results. During the treatment period ALT values were significantly lower in the ribavirin group. Neither in the ribavirin group, nor in the placebo group significant statistical differences of the HCV RNA values were found. Significantly lower portal inflammation was noticed in ribavirin group after the treatment. Analysis of laboratory data demonstrated that ribavirin therapy was associated with mild to moderate reversible anemia. Investigator’s evaluation of the effect of the therapy on patient’s well being showed statistically significant differences in the benefit of the ribavirin group. Conclusion. In this study ribavirin was more effective than placebo in reducing ALT levels during the treatment period of the applied therapy in patients with chronic active hepatitis C.

K e y w o r d s : hepatitis C, chronic; ribavirin; alanine transaminase; blood cell count; hematocrit; hepacivirus.
particularly in those with the history of iv drug abuse. The interval between the transfusion and seroconversion was about 22 weeks (range 10 to 39 weeks). After the presentation with hepatitis, the mean interval to seroconversion was 15 weeks (range 4 to 32 weeks) (8). Transmission of the disease to household contacts or to sexual partners was probably minimal (9).

Acute hepatitis C is usually mild and the patient is often anicteric (10). Clinically, the disease is indistinguishable from other forms of viral hepatitis. The histologic picture is non-specific, with portal inflammation, variable degrees of parenchymal necrosis and active cirrhosis in severe cases.

The most significant feature of hepatitis C is the frequency of chronic liver disease development in patients. Even mild cases tend to progress insidiously over time, often resulting in cirrhosis. Age may be a risk factor, with older patients developing more histologic abnormalities.

The main antiviral agents studied in the treatment of chronic hepatitis C patients were acyclovir and interferon. However, controlled studies with these drugs failed to demonstrate their efficacy in this indication.

Ribavirin – 1-beta-D-ribofuranosyl-1,2,4-triazole-3-carboxamide, demonstrated efficacy when administered through various routes (oral, aerosol and parenteral) and various formulations against a variety of other viral diseases.

The mechanism of action has not been determined so far. One of the hypotheses suggested that the treatment with this antiviral agent resulted in the decreased intracellular pools of guanosine triphosphate, thereby indirectly suppressing synthesis of viral nucleic acid. Another hypothesis suggested that the treatment of viral-infected cells with ribavirin resulted in the synthesis of RNA with abnormal 5’cap structures, which, in turn, led to inefficient translation of viral transcripts. It was difficult to determine experimentally the primary mechanism of action because none of the hypotheses were mutually exclusive.

An important characteristic of ribavirin is its rapid absorption with bioavailability ranging from nearly 100% in some animal models to 33–69% in humans. Maximum plasma concentrations of ribavirin following oral administration were reached within 1–2 hours in human volunteers. Ribavirin pharmacokinetics appeared to be linear over the dose range of 600–2400 mg. Ribavirin was distributed throughout the body including the red blood cells and cerebrospinal fluid. Red blood cells generally displayed the rapid uptake of the drug, and ribavirin concentrations remained elevated long after the plasma levels declined to near-zero values. Excretion was almost entirely via urine in the form of the unchanged drug, and the major metabolite, 1,2,4-triazole-3-carboxamide. In subjects with normal renal and liver function the unchanged drug was found to have a plasma half-life, ranging between 16 and 18 h, while in patients with the impaired renal or liver function the half-life of ribavirin ranged between 143 and 173 h.

With long-term oral clinical administration of 600 mg of ribavirin b.i.d., ribavirin was being accumulated in plasma, reaching steady state within 4 weeks.

Prolonged administration of ribavirin resulted in reversible normocytic and normochromic anemia, due to moderate intravascular hemolysis with an associated delay in the late phase of erythroblast maturation.

In the hepatitis C patients, ribavirin doses of 16 mg/kg/day were with a mean decrease of less than 15% in hemoglobin levels. This decrease was most evident from weeks one to five, after which hemoglobin concentrations stabilized. Within six weeks of therapy cessation hemoglobin levels had normalized.

Our study was designed to evaluate the effectiveness of a 48-week course of ribavirin therapy in subjects with chronic active hepatitis C. This study was conducted in the countries of Central and East European Region (Hungary, Yugoslavia, Poland, Czech Republic and Russia), where 8 research centers participated: Szent Laszlo Hospital, Budapest; Institute for Infectious and Tropical Diseases, Clinical Center of Serbia, Belgrade; Infectious Clinic, Medical Medical Academy, Belgrade; Institute for Infectious Diseases, Medical Academy, Warsaw; Charles University, Internal Medicine, Prague; I Interna Klinika, Fakultetni Nemocnice, Hradec Kralove; D. I. Ivanovski Institute of Virology, Moscow; Gamaleya Research Institute of Epidemiology and Microbiology, Moscow, Russia.

This international, multicenter study was conducted by 8 principal investigators, 2 supervisors (for HCV RNA detection and for histopathological detection), 15 clinical investigators and by the Sponsor’s team consisting of the professionals experienced in clinical trials.

The objectives of this study were to evaluate the efficacy and safety of oral ribavirin in up to 80 evaluable patients with mild to moderate chronic active hepatitis C virus infection, tested in a double-blind, placebo-controlled study, with 48-week course of therapy and with 16-week follow-up period.

Primary efficacy assessment criterion, according to the study protocol, was the response status based on serum aminotransferase activity (ALT) levels and on HCV RNA levels.

Secondary efficacy criteria included changes in liver histopathology and changes in patient’s clinical symptoms. Safety was to be evaluated by comparing the adverse events, changes in laboratory values and tolerability of the drug regimen between the ribavirin and the placebo groups.

The timepoints of primary interest for the evaluation of efficacy were week 48 (the end of the treatment period) and week 64 (the end of the post-treatment follow-up).

Methods

This was a multicenter, international, double-blind, randomized, placebo-controlled study. After the eight-week screening period, patients were randomized to receive ribavirin 1200 mg daily, or placebo during a 48-week treatment period. Follow-up observations were performed during a 16-week post-treatment period.
Up to 80 male and female outpatients with mild to moderate chronic active hepatitis C virus infection were enrolled in this study. The sample size estimate was based on the primary efficacy variable, and the percent of patients with normalization of ALT.

Anticipating non-evaluable patients and dropouts, 40 patients per group were enrolled. To be eligible for the study, patients were to meet the criteria outlined below:

Inclusion criteria specified that the patients should be 18–70 years of age, of both sexes, with elevated serum aminotransferase (ALT) activity, 1.5 times (<400 U/l) the upper limit of normal at least 6 months prior to baseline, prothrombin time less than 3 s prolonged from medium prothrombin time, chiron RIBA 3-HCV test system, liver biopsy consistent with chronic hepatitis, as defined by portal inflammation with no fibrosis, or with fibrous portal expansion; (Knodell Score of Periportal Activity and Lobular Necrosis should be at least 1; Knodell Score for components: periportal necrosis with or without bridging necrosis; intra-lobular degeneration; focal necrosis and portal inflammation should be less than 12); compensated liver disease; CD4 ≥ 10.5 g/l; serum albumin > 35 g/dl; no previous treatment with α-interferon; sexually active patients who practiced the acceptable method of contraception and signed the informed consent.

Exclusion criteria included: Gilbert disease; clinically evident ascites or the history of one, endogenous hepatic encephalopathy or bleeding esophageal varices; pregnancy; women who could not take adequate contraceptive precautions, serologic positivity for hepatitis B surface or HIV antibody (confirmed by Western blotting), significant systemic illnesses other than liver disease (e.g. uncontrolled congestive heart failure, uncontrolled diabetes mellitus, hemophilia etc.); renal impairment, creatinine clearance below lower limit of normal; patients having a liver biopsy with features of other forms of liver disease; clinically significant ECG abnormalities; oral or parenteral antiviral or immunosuppressive therapy within 6 months prior to the study; evidence of other forms of liver disease (e.g., alcoholic liver disease, hemochromatosis etc.); medical, social and psychological implementation to compliance with the drug regimen.

Patients were randomized to one of the two treatment groups with the following dosage regimen:

Ribavirin group: three 200 mg capsules twice daily (1200 mg daily dose) and placebo group: three placebo capsules twice daily. Study medication was to be administered for 48 weeks.

Patient compliance with the study medication dosing regimen was monitored by counting study medication capsules at each study scheduled visit.

All concomitant medication used throughout the study was on the appropriate Case Report Form (CRF) (generic name/s, total daily dose/s, dose unit/s, duration of the treatment and indication stated).

Patients were observed at regular intervals according to the schedule. In total, there were 15 scheduled visits for evaluation and laboratory tests.

Safety data were reported for all patients who received at least one dose of study medication (total population). Safety data included the following: adverse events - any symptom that occurred during the treatment period or between the patient’s last dose and final follow-up, was to be recorded on the patient’s CRF. The investigator was to evaluate each adverse event and record the severity of the event and its relation to the study medication in the CRF, and laboratory measurements.

The efficacy data were analyzed and reported for all statistically evaluable patients.

Primary efficacy criteria: ALT response status and HCV RNA response status were defined as follows:

- ≥ 50% ALT/HCV RNA values – decrease ≥ 50% in any stage of the study, as compared to the baseline ALT/HCV RNA value;
- < 50% ALT/HCV RNA values – decrease <50% in any stage of the study, as compared to the baseline ALT/HCV RNA value;
- 0% ALT/HCV RNA values – no change or increase in any stage of the study, as compared to the baseline ALT/HCV RNA value;

Primary efficacy analysis included: 1) comparison of differences between the visits within the same group; 2) comparison of differences between the study groups at the same visit; 3) comparison ratio (values related to baseline) between the study groups at the same visit, and 4) comparison of previously defined categorization data between the study groups at the same visit.

Secondary efficacy variables were analyzed through comparison differences between study groups within the same visit. These comparisons were used for: 1) symptom questionnaire parameters, 2) patient’s evaluation, and 3) investigator’s evaluation.

Results

During the treatment period (0–48 week), the significant changes of ALT values were detected in both study groups. In the ribavirin group a decrease in ALT values was detected in the period: 8–16 week, and 40–48 week. ALT values decrease was significantly lower in the ribavirin group.

During the post-treatment period (56–64 week), in the ribavirin group, ALT values increased to the values obtained in the screening period (8 weeks before the treatment). Also, after 48-week treatment, there were no significant differences in ALT values between the study groups.

ALT ratio comparison (n week/0 week) during the treatment period, showed that ALT decrease was significantly higher in the ribavirin group. In the post-treatment period there were no differences.

In the ribavirin group, there were no significant statistical differences in HCV RNA values between the sched-
uled visits (week 0, 12, 24, 48 and 64). The same was for the placebo group. Similarly, comparison of HCV RNA values between the study groups showed no differences.

Knodell Score values did not significantly differ between the treatment groups, neither before, nor after the treatment. Also, there were no significant differences in values before the treatment and after the post-treatment period.

Statistical analysis of the histopathological liver biopsy categories showed no statistical differences except in the marked portal inflammation category. Marked portal inflammation was lower after the treatment in ribavirin group.

Clinical symptom values (Symptom Questionnaire: fatigue, anorexia, abdominal pain, patient condition, all ranged 1–10), did not significantly differ between the treatment groups.

Investigator’s evaluation showed statistically significant differences in the benefit of ribavirin group.

Patient evaluation showed no differences between the study groups.

Safety Results

Hematology parameters

Erythrocytes – during the study period there was significant statistical decrease of values compared to the baseline in the ribavirin group. In placebo group significant statistical decrease was detected in 12th and 32nd week of the treatment period.

Comparing the values between the study groups, there were significant statistical differences in weeks 4–32 (it was lower in ribavirin group), but during the post-treatment period (weeks 56–64), these values were lower in the placebo group.

Leukocytes – in ribavirin group, values were higher than baseline during week 2–20, but in the week 48 (the end of the treatment period), values were lower than the baseline.

Placebo group showed no differences during the study period.

Neutrophils – in week 2, and in week 8–48, neutrophils were higher than the baseline in ribavirin group; placebo group showed no statistical differences.

Basophilic – there were no differences, either between the groups, or comparing the treatment weeks.

Lymphocytes – in ribavirin group values were significantly lower in week 2 and during the period of week 8–48. Placebo group showed no differences in any period of the study.

Monocytes – lower values were during 40–48 week in ribavirin group, while in placebo group there were no statistical differences.

Reticulocytes – in ribavirin group higher values were detected during the period of 2–48 week of the treatment. Placebo group showed no differences in any period of the study.

Partial prothrombin time and prothrombin time – values for both parameters were significantly lower in ribavirin group at the end of study (week 64). In placebo group there were no differences.

Hemoglobin – in ribavirin group hemoglobin value showed decreasing tendency (during: week 2–40). Values ranged from 120 mg/100 ml to 137 mg/100 ml. At the end of the treatment (week 40–48) values increased to 137 mg/100 ml. During the post-treatment period (week 56–64), values reversed to pretreatment level, with no significance, compared to the baseline. In placebo group hemoglobin values were significantly lower during the treatment period, compared to the baseline, especially during 12–16 week. Till the end of the study values reversed to the baseline. During the treatment period, hemoglobin values decrease was significantly higher in ribavirin group, than in the placebo group.

Hematocrit – in ribavirin group values were significantly lower in the treatment period (2–48 week), compared to the baseline. In placebo group values were lower in week 4 and in the period of week 16–40. Comparison of ribavirin and placebo hematocrit value decrease showed that it was higher in ribavirin group.

All values in both groups reversed to the baseline during the post-treatment period (week 56–64).

Platelets – ribavirin platelet values were statistically higher during the treatment period (week 2–20, and week 32–48), and in week 64 (post-treatment period). In placebo group there were no differences in platelets values.

Clinical chemistry parameters

CD4, CD8, CD4/CD8, Alkaline phosphatase - values did not differ during the study period.

AST – compared to the baseline, ribavirin group values were significantly lower during weeks 2 - 48; in placebo group these values were lower during weeks 8–64.

During the treatment period, significantly lower values occured in the ribavirin group.

Direct bilirubin, total bilirubin, blood urea nitrogen, creatinine – showed no differences neither between treatment weeks, nor between study groups.

Uric acid – ribavirin values were significantly higher comparing the baseline during the treatment period – week 2–48, and in the post-treatment period, week 56. Placebo group showed no differences.

Other safety evaluations included liver enlargement, spleen enlargement, presence of icterus, Anti HCV – Anti C 1000, HBsAg. These values showed no differences, both compared to the baseline, and to the study groups.

Discussion

Ribavirin is a synthetic purine nucleoside with demonstrated antiviral activity against DNA and RNA viruses. Previous clinical studies in patients with hepatitis C virus suggested that ribavirin was an effective treatment, as demonstrated by significant decreases in ALT activity and the decrease in HCV RNA titer. This study was designed to evaluate the effectiveness of a 48-week-course of ribavirin therapy in patients with chronic active hepatitis C.
Ribavirin was significantly more effective than placebo in reducing ALT levels. Ribavirin patients showed noticeable ALT reductions during the treatment period, (week 2−48). During the post-treatment period, ALT values increased, nearing the baseline ones. Placebo patients had no significant ALT reductions observed during the treatment period. Also, there was no increase in values in the post-treatment period.

HCV RNA values did not differ significantly between the medications given (ribavirin vs. placebo) throughout the whole study period.

Histopathologic examination of liver biopsy samples was performed at the baseline, and once again between weeks 48 and 56. Analysis was performed according to the Knodell score system including: portal inflammation (mild, moderate, marked); periportal activity and necrosis (mild piecemeal necrosis, moderate, marked, moderate with bridging necrosis, marked with bridging necrosis); lobular necrosis (mild, moderate, marked) and fibrosis (mild fibrous portal expansion, moderate with incomplete fibrosis septa, cirrhosis).

The proportion of ribavirin patients after the treatment was significantly smaller (p<0.05) in the marked portal inflammation category than in the categories of mild and moderate portal inflammations. It suggested the hypothesis that ribavirin exerted beneficial effect on portal inflammation.

**Conclusion**

In this study ribavirin (1200 mg/day) was more effective than the placebo in reducing ALT levels during the 48-week treatment period in patients with chronic active hepatitis C. There were no differences between the groups in the post-treatment period. There was no difference between the groups in response based on HCV RNA titers. Regarding the liver biopsy results, more ribavirin than placebo patients showed improvement concerning portal inflammation, but Knodell score values (before and after treatment) did not significantly differ in any treatment group. There were no statistical differences concerning the adverse events. Analysis of laboratory data demonstrated that ribavirin therapy was associated with a mild to moderate reversible anemia.

**References**


**Apstrakt**


PROCENA EFIKASNOSTI I PODNOŠLJIVOSTI RIBAVIRINA KOD BOLESNIKA SA HRONIČNOM INFEKCIJOM HEPATITIS C VIRUSOM

**Uvod.** Izveštaji hepatologa ukazuju da se antitela kod hroničnog hepatitisa C (anti-HCV antitela) mogu detektovati u 71−84% slučajeva obolelih od postransfuzijskog hepatitisa i kod 50% slučajeva sa sporadičnim non-A non-B hepatitismom. Anti-HCV antitela detektovana su kod 0,05−1% dobrovoljnih davača krvi, koji su imali normalan nivo alanintransaminaze (ALT) i negativan rezultat testiranja na anti-HBs. Prisustvo anti-HCV antitela je nađeno kod 67%
bolesnika, koji su u ličnoj anamnezi imali podatak o intravenskoj zloupotrebi lekova ili podatak o autoimunskom hepatitisu, kao i kod 10–30% bolesnika sa hepatocelularnim karcinomom. Ovo ukazuje na to da je virus hepatitisa C glavni uzročnik akutnog i hroničnog hepatitisa širom sveta. **Metode.** Sprovedena je multicentarska, internacionalna, duplo-slepa, randomizovana, placebom kontrolisana studija. Posle osam nedelja praćenja, bolesnici su za vreme 48-nedelnjeg perioda terapije, randomizovano primali ribavirin 1200 mg dnevno ili placebo. Kontrole su sprovedene u toku 16 nedelja postterapijskog perioda. U ovu studiju je bilo uključeno 80 ambulantnih bolesnika, oba pola, kod kojih je bila prisutna blaga do umerena hronična aktivna infekcija izazvana virusom hepatitisa C. **Rezultati.** Za vreme terapijskog perioda, vrednosti ALT bile su značajno smanjene u grupi bolesnika koji su primali ribavirin. Između ispitivanih grupa (na terapiji ribavirinom i na placebu), nije postojala statistički značajna razlika HCV RNK vrednosti. U grupi bolesnika koji su primali ribavirin, posle prekida terapije, zabeležena je značajno niža portna inflamacija. Laboratorijska analiza pokazala je da se u toku primene terapije ribavarinom javlja blaga do umerena anemija reverzibilnog tipa. Procena istraživača o uticaju terapije na opšte stanje bolesnika značajno se razlikovala u korist grupe pod terapijom ribavirinom. **Zaključak.** Ova studija je pokazala da je u tretmanu hroničnog akutnog hepatitisa C, za vreme primene terapije, ribavirin bio efikasniji od placeba u snižavanju vrednosti ALT.

**Ključne reči:** hepatitis C, hronični; ribavirin; alanin aminotransferaza; krvna slika; hematokrit; hepacivirus.