The Prevalence of Viral Hepatitis C in Latvia: A Population-Based Study

Ieva Tolmane[1, 2], Baiba Rozentale[3, 4], Jazeps Keiss[1], Frida Arsa[5], Girts Brigis[6], Aivars Zvaigzne[7]

[1]Hepatology Department, State Agency "Infectology Center of Latvia," Riga, Latvia, [2]Riga Stradins University, Latvia, [3]State Agency "Infectology Center of Latvia," Riga, Latvia, [4]Infectology and Dermatology Department, Riga Stradins University, Latvia, [5]Laboratory, State Agency "Infectology Center of Latvia," Riga, Latvia, [6]Department of Public Health and Epidemiology, Riga Stradins University, Latvia, [7]University of Latvia, Riga, Latvia

Key words: *hepatitis C; prevalence; population.*

Summary. *Background and Objective. Chronic viral hepatitis C (VHC) is one of the most discussed infectious diseases worldwide. The number of infected persons worldwide is approximately 170 million, and in Europe, it exceeds 9 million.*

The aim of this study was to determine the prevalence of antibodies to hepatitis C virus (anti-HCV prevalence) and prevalence of HCV viremia (HCV-RNA prevalence) in Latvia.

Material and Methods. A multistage randomized selection was used. A total of 42 primary care physicians (PCPs) were randomly selected from the register of PCPs from different regions of Latvia. From each PCP register, 60 subjects were selected (1651 individuals in total) and invited for the anti-HCV test with a screening method (ELISA). In case of positive results, antibodies were confirmed by the Western blot test, and all these subjects were tested for HCV-RNA by polymerase chain reaction.

Results. Of the 1459 subjects tested, 57 were positive for anti-HCV (3.9%; 95% CI 3% to 5%); 35 of them were positive for anti-HCV with a confirmatory test (2.4%; 95% CI, 1.7% to 3.3%): 19 men and 16 women (3.8% and 1.7%, respectively; P=0.011). The results of HCV RNA test were positive in 25 subjects (1.7%; 95% CI, 1.2% to 2.5%): 15 men and 10 women (3% and 1% respectively, P=0.019).

Conclusions. The prevalence of anti-HCV and HCV-RNA in Latvia was found to be 2.4% and 1.7%, respectively. The prevalence of anti-HCV and HCV-RNA was higher in men than women.

Introduction

Chronic viral hepatitis C (VHC) has become one of the most discussed infectious diseases worldwide due to its prevalence and clinical course. Currently, the number of infected persons worldwide is approximately 170 million (1), and in Europe, it exceeds 9 million (2). The hepatitis C virus spreads mainly via blood. Intravenous (also intranasal) drug users; dialysis patients; persons using tattooing, manicure, and pedicure services; and alcohol users are at higher risk of infection; the virus can be transmitted vertically from a mother to a child and sexually (3, 4). In Spain, during a study of possible ways of infection with acute VHC, more than half of patients were hospitalized during incubation period; thus, the subjects were possibly infected during various medical procedures (5).

Hepatitis C virus is the main cause for the development of chronic hepatitis, liver cirrhosis, and hepatocellular carcinoma and for the need for liver transplantation (6). The prevalence of hepatitis C in the countries of the World Health Organization's European region varies from 0.1% to 4.5%. In approximately 20% of cases, the infection progresses to cirrhosis and end-stage liver disease within 10 to 20 years. Currently, only combination therapy and virus eradication can stop the disease progression (6). With the combination of pegylated interferon and ribavirin, a sustained virological response can be achieved in 54% to 63% of patients (6).

To date, no population-based studies on the prevalence of viral hepatitis C in Latvia have been performed. Therefore, the aim of this study was to determine the prevalence of antibodies to hepatitis C virus (anti-HCV prevalence) and prevalence of HCV viremia (HCV-RNA prevalence) in Latvia.

Material and Methods

Subjects were selected by multistage randomized screening. From the register of primary care physicians, 42 primary care physicians were randomly chosen. Using the Microsoft Office Excel program, a number in random order was allocated to each primary care physician from the Register of primary care physicians of Latvia, they were arranged in ascending order, and the first 42 physicians were selected. Of the selected physicians, 4 did not have primary care physician practice and therefore were not included in the study. Of the remaining 38 pri-

Correspondence to I. Tolmane, State Agency "Infectology Center of Latvia," Linezera iela 3, Riga, 1006, Latvia
E-mail: ieva.tolmane@inbox.lv

mary care physicians, 11 refused to take part due to various reasons. Thus, 27 primary care physicians (71%) took part in the study. Primary care physicians represented all regions of Latvia, both cities and parishes.

According to statistics of European countries, the expected prevalence of anti-HCV in Latvia is reported to be 2% to 3%. Based on these estimates, at least one thousand of Latvian population had to be tested in order to get valid results. Calculations were made using the EpiInfo StatCalc program with 95% confidence interval and 1% acceptable error.

From the register of each primary care physician, 60 patients aged more than 18 years were selected: every 33rd patient was invited for the test if 2000 patients were registered, or every 25th patient if 1500 patients were registered. In total, 1651 patients were invited to take part in the study, and 1459 subjects agreed to perform the test (response rate, 88.4%). The participants were mostly Latvians (65%) and Russians (26%); however, there were also representatives of Poles, Ukrainians, Byelorussians, Tatars, Romany, Lithuanians, and other nationalities living in Latvia who were registered at the primary care physician. Health status of each included individual was not examined as the subjects were randomly selected.

If the selected subject was younger than 18 years, the next one on the list was taken. If the invited subject refused to take part, it was stated "refused" in the protocol. If the selected subject had been tested for anti-HCV during the last year with the same ELISA test kit as the study participants (see below), this result was used, and if earlier, the patient was tested again.

According to the data of the Central Statistical Bureau of the Republic of Latvia, there were 45% of men and 55% of women in the age group of 18 years and older with a mean age of 47 years at the time of the study. A total of 1651 persons were selected for our study: 587 men (36%) and 1064 women (64%). Of them, 1459 subjects underwent testing: 499 men (34%) and 960 women (66%). The mean age was 44 years. The study sample represents the Latvian population with regard to the number of tested individuals and age. There is a little deviation in the study group concerning gender, which is discussed below according to the study results.

For the group of the selected subjects, serum anti-HCV was measured by a screening method (immuno-enzymatic analysis, ELISA). The following test systems were used for the detection of anti-HCV: ORTHO® HCV version 3.0 ELISA Test System with Enhanced SAVe, Ortho-Clinical Diagnostics, Inc., USA (sensitivity, 100%; specificity, 99.91%); INNOTEST® HCV Ab IV, Innogenetics N.V., Belgium (sensitivity, 100%; specificity, 99.8%); and AxSYM system HCV version 3.0, Abbott, USA (sensitivity, 100%; specificity, 99.84%).

In case of positive anti-HCV ELISA results, the presence of antibodies in blood serum was confirmed (Western blot). Using the DECISCAN® HCV PLUS test system (Bio-Rad, France), the results were considered as positive if at least 2 HCV antigen bands from 2 different gene products (capsid gene, NS3 gene, NS4 gene) had an intensity greater than 0.5+. The results were considered as probable positive if 2 HCV antigen bands from 2 different gene products (capsid gene, NS3 gene, NS4 gene) had an intensity equal to 0.5+. Using the INNO-LIA HCV Score test system (Innogenetics N.V., Belgium; sensitivity, 100%; specificity, 94.5%), the samples were considered positive for HCV antibodies if at least 2 HCV antigen lines had a reactivity of ± minimum or higher. The samples were considered indeterminate for HCV antibodies if 1 HCV antigen line had a reactivity rating of 1+ or higher, or if the NS3 line was reactive with a reactivity of ± or higher and all other antigen lines were negative.

In case of positive results for anti-HCV by ELISA or Western blot test, the presence of the virus in blood serum was detected (HCV-RNA; polymerase chain reaction, PCR). The Cobas AMPLICOR Hepatitis C Virus Test, v. 2.0 test system was used (Roche Diagnostics, USA; sensitivity, >50 IU/mL; specificity, 100%). Alanine aminotransferase activity was measured for HCV-RNA-positive patients (reference range, 0–41 U/L for men and 0–31 U/L for women).

The study was carried out at the Infectology Center of Latvia, primary care settings, E. Gulbis Laboratory, NMS Laboratory, and laboratories of some regional hospitals from March 2008 until October 2008.

Statistical Analysis. Data were treated and analyzed with the Microsoft Office Excel, SPSS, and EpiInfo programs. Significance of differences was evaluated using the EpiInfo program with 95% confidence intervals. Differences were considered statistically significant if P value was <0.05.

Results

A total of 1459 subjects were tested for antibodies to hepatitis C virus (anti-HCV) with a screening method (ELISA): 499 men (34%) and 960 women (66%).

Fifty-seven subjects (3.9%; 95% CI, 3% to 5%) were found to be positive for anti-HCV with a screening method, and 35 subjects (2.4%; 95% CI, 1.7% to 3.3%) were found to be positive for anti-HCV with a confirmatory test (Western blot): 19 men and 16 women (3.8% and 1.7%, respectively; $P=0.011$). There were 3 probable positive results with the DECISCAN test system, but all three subjects were negative for HCV RNA.

When testing with PCR to detect the presence of the virus in blood (HCV-RNA), positive results were documented in 25 cases (1.7%; 95% CI, 1.2% to 2.5%): 15 men and 10 women (3% and 1%, respectively; P=0.019).

The mean age of the subjects included in the study was 44 years (range, 18–94 years), and that of the subjects infected with hepatitis C virus was 48 years (range, 18–86 years).

Alanine aminotransferase activity in the HCV-RNA-positive patients ranged from 27 to 209 U/L; in one-fifth of these patients, alanine aminotransferase activity was within the reference range.

Discussion

Analysis of the data available in other countries shows that the prevalence of hepatitis C virus in the United Kingdom and Scandinavia is significantly lower than in Latvia, but further to the south – in Southern Italy, Egypt, and Ukraine – significantly higher. Our data are closer to the European countries and the United States (4, 7–11) (Table).

To date, antibodies to HCV in Latvia are being tested in different patient/population groups: blood donors, HIV-infected patients, pregnant women (not mandatory), patients with hepatic test abnormalities, and employees of some medical institutions. Rozentale in her medical doctoral thesis from 1995 "Hepatitis C in Latvia: Facts and Problems" analyzed the students of high schools and universities and potential donors in Riga and Liepaja region, assessing the prevalence of antibodies (anti-HCV confirmatory test) to the hepatitis C virus. Positive results were found in 3.2% of students, 9.3% of potential donors in Riga, and 17.6% of potential donors in Liepaja region, where 90% of the potential donors were unemployed (12). Currently, all potential donors are tested for antibodies to VCH (anti-HCV) by the National Blood Service of Latvia. However, available data cannot be used for the determination of the disease prevalence as the numbers of donated blood units and anti-HCV-positive blood units are available, but there are no data on the number of individual persons (one person not infected with hepatitis C virus may donate blood repeatedly). Among HIV-infected patients registered at the Infectology Center of Latvia, antibodies to hepatitis C virus (anti-HCV) were detected in 63% of patients.

Table. The Prevalence of Anti-HCV and HCV-RNA in Different Countries and Latvia

Country	Anti-HCV, %	HCV-RNA, %
United Kingdom (4)	0.3	–
Sweden (7)	0.5	–
Ukraine (4)	9.2	–
Southern Italy (8)	12.6	–
Egypt (Nile Delta) (9)	24.3	14.7
United States (10)	1.6	1.3
France (11)	1.2	0.9
Latvia	2.4	1.7

However, to date, no large-scale population-based studies on the prevalence of hepatitis C virus in Latvia have been performed.

This is the first population-based study on the prevalence of hepatitis C virus in Latvia, and the obtained data are attributable to the Latvian population in general. However, it has also some limitations: children, prisoners, and persons without a definite place of residence were not included in the study. Besides, the inhabitants of Latvia who were not registered by primary care physicians could not be included in the study. The study data showed that the detection of antibodies (anti-HCV) with ELISA is only a screening, and positive results do not necessarily mean real antibodies in blood, as this analysis can show false-positive results in about one-third of the cases. A 100% sensitivity of these tests (according to the description from the manufacturer) means that there are no false-negative results (theoretically not conceded); thus, no person with a negative result is missed. The relatively large percentage (38.6%) of false-positive cases identified by a screening method does not affect the positive predictive value, but corresponds to data of other countries where similar results were obtained (8, 11). To increase precision of the obtained data, possibly both screening and confirmatory tests with test systems of one manufacturer should be performed, which was not possible in this study, because screening tests were performed in different laboratories in Latvia. The presence of the virus itself in patient's serum (HCV-RNA positive) was confirmed in approximately half of those individuals who were primary positive to antibody test (with screening test, ELISA).

The mean age of the Latvian population aged more than 18 years is 47 years; the mean age of subjects included in this study was 44 years. The mean age of HCV-RNA-positive subjects was 48 years. More detailed analysis of the differences in the VHC prevalence in different age groups was not performed, because taking into account the number of subjects tested in this study, there was a relatively small group showing positive results, and the reliable results would not have been obtained, if this group had been analyzed in detail. Moreover, this was not the aim of this study.

In this study, the prevalence of infection with hepatitis C virus among men was found to be higher than among women. However, when the routes of infection and risk factors of VHC (use of drugs and alcohol) are analyzed (3, 4), it is possible that men are using drugs and alcohol more frequently; therefore, there is a higher risk of infection theoretically. Alcohol users were shown to have a higher preva-

lence of HCV infection ranging from 7% in Germany up to 49% in Italy. In addition, the HCV-RNA viral load in serum is higher in alcohol users. These facts suggest that alcohol can affect the patient's immune response to HCV (4). However, genetic differences of both genders as a potential reason for the higher susceptibility of men to hepatitis C virus cannot be excluded. Due to the observed difference and taking into account the gender disproportion that occurred during the study screening period as a result of demographic situation of Latvia (proportion of men in the study was lower), the overall true prevalence of VHC in Latvia could be even higher than observed.

Considering the study results, there are almost 39 000 patients with chronic hepatitis C in Latvia. The vast majority should be treated with standard care therapy, but all HCV-RNA-positive patients should be examined carefully – disease history, epidemiological anamnesis, concomitant diseases, laboratory examination, and possibly liver biopsy – in order to evaluate in whom the treatment is indicated and for whom monitoring is currently necessary.

Conclusion

The prevalence of anti-HCV and HCV-RNA in Latvia was found to be 2.4% and 1.7%, respectively. The prevalence of anti-HCV and HCV-RNA was higher in men than women.

Acknowledgments

This study was supported by the Roche Academy of Latvia.

Statement of Conflict of Interest

The authors state no conflict of interest.

References

1. Chen W, Wong T, Tomlinson G, Krahn M, Heathcote EJ. Prevalence and predictors of obesity among individuals with positive hepatitis C antibody in a tertiary referral clinic. J Hepatol 2008;49(5):711-7.
2. Vingre I, Rozentale B, Keiss J, Kuse V, Lazdina D, Markovs J, et al. The treatment guidelines of viral hepatitis C. 2nd ed. Riga: SA Infectology Center of Latvia; 2005.
3. Viksna L. Viral hepatitis C: clinical course, treatment, prognosis, prevention. Riga: SIA Nacionalais apgads; 2003. p. 26-36.
4. Heintges T, Wands JR. Hepatitis C virus: epidemiology and transmission. Hepatology 1997;26(3):521-6.
5. Martinez-Bauer E, Forns X, Armelles M, Planas R, Solà R, Vergara M, et al. Hospital admission is a relevant source of hepatitis C virus acquisition in Spain. J Hepatol 2008;48(1): 20-7.
6. Lettmeier B, Muhlberger N, Schwarzer R, Sroczynski G, Wright D, Zeuzem S, et al. Market uptake of new antiviral drugs for the treatment of hepatitis C. J Hepatol 2008; 49(4):528-536.
7. Hoffmann G, Berglund G, Elmsta S, Eriksson S, Verbaan H, Widell A, et al. Prevalence and clinical spectrum of chronic viral hepatitis in a middle-aged Swedish general urban population. Scand J Gastroenterol 2000;35:861-5.
8. Guadagnino V, Stroffolini T, Rapicetta M, Costantino A, Kondili LA, Menniti-Ippolito F, et al. Prevalence, risk factors, and genotype distribution of hepatitis C virus infection in the general population: a community-based survey in southern Italy. Hepatology 1997;26:1006-11.
9. Abdel-Aziz F, Habib M, Mohamed MK, Abdel-Hamid M, Gamil F, Madkour S, et al. Hepatitis C virus (HCV) infection in a community in the Nile Delta: population description and HCV prevalence. Hepatology 2000;32:111-5.
10. Armstrong GL, Wasley A, Simard EP, McQuillan GM, Kuhnert WL, Alter MJ. The prevalence of hepatitis C virus infection in the United States, 1999 through 2002. Ann Intern Med 2006;144(10):705-14.
11. Dubois F, Desenclos JC, Mariotte N, Goudeau A. Hepatitis C in a French population-based survey, 1994: seroprevalence, frequency of viremia, genotype distribution, and risk factors. Hepatology 1997;25:1490-6.
12. Rozentale B. Hepatitis C in Latvia: facts and problems. [dissertation]. Riga: Riga Stradins University; 1995.

Received 17 September 2010, accepted 31 October 2011