



Assessing the Hepatitis C Genotype in Prisoners Infected with HCV in South Khorasan- 2014

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Received 2017 November 14; Revised 2018 January 10; Accepted 2018 April 10.

Abstract

Background and Objectives: Hepatitis C is a dangerous viral infection, which is the important cause of chronic liver diseases such as fibrosis, cirrhosis, and hepatocellular carcinoma. The hepatitis C virus (HCV) has six major genotype of HCV identified for which are different in prevalence, carcinogenicity, persistency, and response to the treatment. Identification of HCV genotype and viral load are main factors for planning of the treat and prediction of response. There has not been any study regarding the viral load and genotype distribution of HCV in our area and this study was done to evaluate risk factors, viral load, and genotype of HCV among prisoners in East Iran.

Methods: In a cross-sectional study, 2,995 prisoners across the South Khorasan province were screened for the anti-HCV antibody by ELISA method. A total of 38 prisoners with positive results were selected for further evaluation. Demographic data as well as predisposing factors were obtained from the records. A total of 5 ml of venous blood was taken and after RNA extraction, viral load genotype and HCV were identified by using the commercial Real Time PCR and PCR method, respectively.

Results: Among 2,995 prisoners, 34 cases (mean age 39 ± 8.8 years, M/F ratio: 13.1) were positive for HCV antibody. A total of 50% were married, 79.4% were urban citizens, and 97.1% of patients infected with HCV had drug addiction. In 28 cases (82.3%), the virus was detected by real time PCR method. Genotype 3a and 1a were the most prevalence genotypes (75% and 8.3%, respectively). The average of viral load was 123,798 IU/mL. There was no significant relation between age or sex with viral load and genotype.

Discussion and Conclusions: This study showed that the predominant genotype in prisoners is the genotype 3a, which adds to the evidence of the predominant genotype in Iran. The genotype of injecting drugs prisoners is different to other groups infected with Hepatitis C in Iran.

Keywords: Hepatitis C, Genotype, Prison, Iran

1. Background

The hepatitis C virus (HCV) has infected about 300 million people around the world and is the main reason of chronic liver diseases such as fibrosis, cirrhosis, and hepatocellular carcinoma (1, 2).

The hepatitis C infection is usually asymptomatic and eventually leads to cirrhosis and liver cancer in 20 - 30 years (3).

HCV is an RNA virus with a numerous rate of intrinsic mutation direction to significant heterogeneity in the genome. HCV has six major genotypes and over 70 subtypes (a, b, c, d ...). The prevalence of different genotypes is affected by several factors, such as the pattern of infection, geographic variables, migration, population, and so on (3, 4).

Identification of viral load and genotype of HCV virus has a very important role not only in choosing type, dose, and duration of treatment, however, also in the evaluation of the response to the treatment and prognosis (5, 6). There is a significant body of evidences that show correlation between viral load, genotype, and response to the treatment.

Although genotype 1, 2, and 3 account for over 90% of infections in Europe, the South and North America, as well as Japan, the distribution and prevalence of these species depend on the geographic location and method of transmission. Genotypes 1a and 3a are prevalent in intravenous drugs abusers, while genotype 1b is commonly associated with blood transfusion (7-9).

Patients with genotype 1 respond poorly to treatment and have higher risk of hepatocellular carcinoma while patients infected with genotypes 2 and 3 have a quite better

response to the treatment (10).

Several studies from different parts of Iran have shown that about 1% of Iranians are infected with HCV and genotypes 1a and 3a are the most common genotypes (11-13); however, to the best of our knowledge there is no data regarding the viral load and genotype distribution among prisoners while they have usually risky behaviors and have a higher rate of HCV infections as we have reported in our previous study (14).

The purpose of this study was to appraise the risk factors, viral load, and genotype distribution of HCV among prisoners in South Khorasan.

2. Objectives

The aim of this study was to appraise the risk factors, viral load, and genotype distribution of HCV among prisoners in South Khorasan.

3. Methods

In a cross-sectional study on prisoners of the South Khorasan province, 2,995 prisoners voluntarily participated in the study. The study was approved by the ethics committee of the Birjand University of Medical Sciences with research project number ir.bums.REC.1394.418.

All participants provided written informed consent before enrolling in the study. A total of 5 mL of venous blood sample was taken from all participants and after serum separation; all sera were screened for the anti-HCV antibody by using the commercial ELISA Kit (Diapro diagnostic, Italy). HCV positive sera were rechecked by Cobas elexys analyzer and for those with confirmed positive result, the level of AST/ALT was measured by automated biochemistry analyzer. RNA was extracted from sera by the Qiagen viral RNA kit (Qiagen, Germany) in a maximum 8 hours and quantity and quality of extracted RNA were measured by the Nanodrop machine (Thermofisher, USA). A total of 5 microliter of RNA was used for cDNA synthesis by commercial kit (Thermofisher, USA). viral load HCV detected by real time PCR kit (Primerdesign, UK) using ABI step one machine and all samples were checked in duplicate. HCV genotyping was performed by HCV genotyping kit (Sac-case, Italy), which identifies 4 different genotypes including 1a, 1b, 2 and 3a.

The data were statistically analyzed by the SPSS software version 19 (SPSS inc, Chicago, USA). Data comparisons were conducted using the Fisher's exact test, Chi-square test, and independent t-Test. P value < 0.05 was considered as the level of significance.

4. Results

Among the 2,995 prisoner that participated in this study, 41 cases were positive for anit-HCV by ELISA method, however, the infection was confirmed in 34 cases who were positive for HCV antibody in the second test (mean age = 39 ± 8.8 years, range: 22 - 62 year, M/F ratio: 13:1). A total of 50% were married and 79.4% were urban citizens. Frequency of hepatitis B and HIV were 20.6 % and 2.9%, respectively. Distribution of Hepatitis risk factors in this study is shown in Table 1.

Table 1. Frequency Distribution of Risk Factors in Prisoners with Positive Anti HCV^a

Risk Factors Positive	Negative	
Addiction	40 (97.6)	1 (2.4)
Sharing needle	4 (9.8)	37 (90.2)
Smoking	15 (36.6)	26 (63.4)
Tattooing	24 (58.5)	17 (41.5)
Transfusion	5 (12.2)	36 (87.8)
Surgery	12 (29.3)	29 (70.7)

^aValues are expressed as No. (%).

A total of 28 prisoners (82.3%) had a detectable viral load. The average of viral load was 309,496 copy/mL or 123798 IU/mL. in 20% of patients; the viral load was higher than 130,000 IU/mL, which is considered as the cut-off for good virologic response (15). Frequency positive anti HCV and positive PCR, according to risk factors in this study, is shown in Table 2.

Genotype 3a and 1a were the most prevalent genotypes (75% vs 8.3%, respectively) and in 16.6% the genotype was not identified. There was no significant relation between age or sex with genotype and viral load. Mean ALT was 17.9 ± 13.1 in PCR+ patients and 6.6 ± 2.9 in PCR- patients (P = 0.008).

5. Discussion

This is the first report on prevalence, viral load, and genotypes of HCV among prisoners in the east of Iran. According to this study, prevalence of HCV infection among prisoners is higher than the normal population (14, 15).

In case of viral load, several studies have reported cut-off values as a predictor of good virologic response. For example Berg et al. (16) considered the level of 130,000 IU/mL as a cut-off for sustained virologic response or two other studies defined 800,000 IU/mL and 400,000 IU/mL (17, 18) for discriminating high and low pre-treatment viral load. Most of our cases had a viral load less than 130,000 IU/mL, which is a good sign. On the other hand several

Table 2. Frequency Positive Anti HCV and Positive PCR According Risk Factors^a

Characteristics	Anti HCV	Positive PCR
	Positive	Positive
Sex		
Male	32 (94.1)	26 (66.7)
Female	2 (5.9)	2 (100)
Marital		
Single	17 (50)	12 (63.3)
Married	17 (50)	16 (72.7)
Residence		
Urban	27 (79.4)	22 (64.7)
Rural	7 (20.6)	6 (85.7)
Addiction		
Yes	33 (97.1)	27 (67.5)
No	1 (2.9)	1 (100)
Sharing needle		
Yes	3 (8.8)	2 (50)
No	31 (91.2)	26 (70.3)
Smoking		
Yes	12 (35.3)	9 (60)
No	22 (64.7)	19 (73.1)
Tattooing		
Yes	20 (58.8)	18 (75)
No	14 (41.2)	10 (58.8)
Transfusion		
Yes	4 (11.8)	3 (60)
No	30 (88.2)	25 (69.4)
Surgery		
Yes	9 (26.5)	8 (66.7)
No	25 (73.5)	20 (69)

^aValues are expressed as No. (%).

cases with Anti-HCV had no detectable viral load, which may be explained by spontaneous viral clearance phenomena, which happened in 10% - 50% of HCV infections (19-21).

Genotype 3a was the predominant genotype in our survey. Similarly, in a study that was conducted in a close province in the North-East of Iran (Mashhad), genotypes 3a and 1a were reported as the most prevalent genotypes, respectively (22). Genotype 3a is also predominant in India and Pakistan, two close countries located near the east of Iran, with a high prevalence of HCV infection (4, 23, 24).

Many other studies mostly on general populations reported genotypes 1a and then 3a as the predominant genotypes. For instance, several reports from different parts of

Iran including South (Bushehr), South-West (Ahvaz), West (East Azerbaijan), South-East (Zahedan), Center (Yazd), and North showed the predominance of genotype 1a and then 3a (4, 11-13, 25, 26).

Genotype 1b was a predominant genotype in Turkey in two studies (27, 28) and also in 98 different countries, according to an article published in 2014 (29).

The difference in study population can explain the lower rate on genotype 1a as the situation in prisoner is different and many of them several risk factors especially history of drug injection.

Parallel to our result, in Tyczyno's study on Poland's prisoners, genotype 3a was the most common genotype (30).

There is also the possibility of transfecting the virus during the imprisonment, which can explain a high rate of infection with genotype 3a; Samimi et al. showed such correlation (31).

In this study, there was no significant relationship between age and genotype; the same result was found by Somi et al. and Kabir et al. (25, 26). In contrast, in Turkey, genotype 1a was seen more in older patients (27). Jahanbakhsh Sefidi et al. found in a study in 2001 that genotype 1a was more common in people over 40 years of age and genotype 3a in people under 40 years (32). Lee and colleagues also showed that genotype 1 is more common in older patients and genotype 2 in younger patients (33). Association of genotype 1b with age has also been shown in studies by Hosseini-Moghaddam et al. (34). However, Samimi-Rad et al. found that genotype 3a is common among IDUs less than 40 years old compared to those older than 40 years (35). In general, in most studies establishing the relationship between age and genotype, the predominant genotype in older patients was 1b.

In the current study, no association was found between genotype and sex (25, 27). There was a significant correlation between HCV and injection drugs use. Several studies have shown that HCV infection's rate is higher in injecting drug users than ordinary people. Similar to other developing countries, injection drug use has turned into the main route of new hepatitis C infection in Iran. Recent studies on the Iranian injecting drug users (IDUs) report the prevalence a high of HCV infection, ranging from 50% to 75% depending on the area of the study, and also a significant increase in HCV infection over the last decade (14). Reports from different regions in Iran show that 31.5% to 88.9%, of IDUs have HCV antibody (32). Needle sharing among drug users, particularly in prisons, who usually have limited access to syringes, is the major reason.

Other epidemiologic molecular studies have demonstrated a significant correlation between genotype 3a transmission and injecting drug use. The distribution of

the subtypes has been fully explained in North and South America as well as European countries. The predominant HCV subtype among IDUs in Europe is 3a, whereas 1b is common in patients who have acquired the virus through blood transfusion (34, 36-38).

In Asian countries, genotype 3a was separated mostly from injecting drug users (39). In Iran, subtype 1a is the maximum prevalent in transfusion-related risk groups, such as hemodialysis, thalassemia, and hemophilia patients (34, 36). Unfortunately, studies that evaluate genotypes in Iranian prisoners are very few. In addition, the data available on the molecular epidemiology of HCV among Iranian IDUs is scarce (40).

There is typically an association between the genotype and HCV transmission route. Based on some previous reports from Iran, genotype 1a is predominant in groups receiving blood transfusion (31, 32). Most studies show that variants of genotype 3a is epidemic in drug injecting users (35), which is confirmed in our study. In the present study, no significant relationship was found between injection drug use and genotype, due to the small sample size.

In 9% of patients, no risk factor was found for disease transmission that can support secondary and unexpected ways neglected by patients, such as piercing ears or using common tools like nail clippers and tweezers.

In 16.6% of cases, the genotype was not identified possibly due to unusual genotypes, as our kit just detects four common genotypes while there are 6 major genotypes.

Since the main method of HCV transmission is intravenous drugs and the prison environment is very suitable for development of an epidemic, disease prevention strategies should be scheduled with a greater emphasis on this particular group of patients.

The present study has a few limitations that might affect our results. A retrospective study might suffer from sampling bias; the prisoners' screening for Anti-HCV might not have covered all the prison population due to the flexibility of the prison population. On the other hand, the male prisoners' population is more than females, which causes the lack of generalizability of the results.

Additional studies in this area could help in predicting genotype frequency and distribution in the Iranian prisoners.

This study showed that the predominant genotype in prisoners is genotype 3a, which adds to the evidence of the predominant genotype in Iran. The genotype of injecting drugs in prisoners is different to other groups infected with hepatitis C in Iran. More studies are needed to prove the specific genotype in injecting drug users with larger sample sizes.

Acknowledgments

We would like to thank all prisoners and personnel in South Khorasan that helped us in conducting this study.

Footnote

Financial Disclosure: All authors declare that they have no financial disclosures and no conflict of interest to report.

References

1. Simmonds P. Genetic diversity and evolution of hepatitis C virus-15 years on. *J Gen Virol*. 2004;**85**(Pt 11):3173-88. doi: [10.1099/vir.0.80401-0](https://doi.org/10.1099/vir.0.80401-0). [PubMed: [15483230](https://pubmed.ncbi.nlm.nih.gov/15483230/)].
2. Poinard T, Ratziu V, Benhamou Y, Opolon P, Cacoub P, Bedossa P. Natural history of HCV infection. *Baillieres Best Pract Res Clin Gastroenterol*. 2000;**14**(2):211-28. [PubMed: [10890317](https://pubmed.ncbi.nlm.nih.gov/10890317/)].
3. Keyvani H, Alizadeh AH, Alavian SM, Ranjbar M, Hatami S. Distribution frequency of hepatitis C virus genotypes in 2231 patients in Iran. *Hepatol Res*. 2007;**37**(2):101-3. doi: [10.1111/j.1872-034X.2007.00015.x](https://doi.org/10.1111/j.1872-034X.2007.00015.x). [PubMed: [17300704](https://pubmed.ncbi.nlm.nih.gov/17300704/)].
4. Joukar F, Khalesi AK, Jafarshad R, Rahimabadi MS, Mansour-Ghanaei F. Distribution of hepatitis C virus genotypes in haemodialysis patients of Guilan, northern Islamic Republic of Iran. *East Mediterr Health J*. 2012;**18**(3):236-40. [PubMed: [22574476](https://pubmed.ncbi.nlm.nih.gov/22574476/)].
5. Jimenez-Mendez R, Uribe-Salas F, Lopez-Guillen P, Cisneros-Garza L, Castaneda-Hernandez G. Distribution of HCV genotypes and HCV RNA viral load in different regions of Mexico. *Ann Hepatol*. 2010;**9**(1):33-9. [PubMed: [20308720](https://pubmed.ncbi.nlm.nih.gov/20308720/)].
6. Hnatyszyn HJ. Chronic hepatitis C and genotyping: the clinical significance of determining HCV genotypes. *Antivir Ther*. 2005;**10**(1):1-11. [PubMed: [15751759](https://pubmed.ncbi.nlm.nih.gov/15751759/)].
7. Shepard CW, Finelli L, Alter MJ. Global epidemiology of hepatitis C virus infection. *Lancet Infect Dis*. 2005;**5**(9):558-67. doi: [10.1016/S1473-3099\(05\)70216-4](https://doi.org/10.1016/S1473-3099(05)70216-4). [PubMed: [16122679](https://pubmed.ncbi.nlm.nih.gov/16122679/)].
8. Grimaldi E, Della Pepa ME, Martora F, Magliocca P, Iovene MR, Coppola N, et al. Distribution of Hepatitis C Virus Genotypes and Subtypes in the Metropolitan Area of Naples, Italy, in the Era of Interferon-Free Regimens. *Intervirology*. 2017;**60**(3):82-9. doi: [10.1159/000481821](https://doi.org/10.1159/000481821). [PubMed: [29161710](https://pubmed.ncbi.nlm.nih.gov/29161710/)].
9. Acero Fernandez D, Ferri Iglesias MJ, Buxo Pujolras M, Lopez Nunez C, Serra Matamala I, Queralt Moles X, et al. Changes in the epidemiology and distribution of the hepatitis C virus genotypes in North-Eastern Spain over the last 35 years. *Gastroenterol Hepatol*. 2018;**41**(1):2-11. doi: [10.1016/j.gastrohep.2017.09.004](https://doi.org/10.1016/j.gastrohep.2017.09.004). [PubMed: [29150360](https://pubmed.ncbi.nlm.nih.gov/29150360/)].
10. Ray S, Thomas DL. Hepatitis C. In: Mandell GL, Bennet J, Dolin R, editors. *Principle and practice of infectious disease*. Philadelphia: Elsevier-churcillivingstone; 2015. p. 1908-27.
11. Vahdat K, Amini A, Motamed N, Eghbali SS, Zandi K, Hajjani G. Determination of Hepatitis C genotype and risk factors of transmission in Bushehr province 2008. *Tibb-i junub*. 2012;**15**(1):51-8.
12. Haghshenas MR, Babamahmoodi F, Rafiei AR, Vahedi V. The Correlation between HCV Genotypes and Liver Damage in HCV Patients. *J Mazandaran Univ Med Sci*. 2011;**21**(85):76-83.
13. Hajjani E, Hashemi SJ, Masjedizadeh A, Shayesteh AA, Jalali F. Genotypic analysis of hepatitis C virus in khuzestan province, southwestern iran. *Middle East J Dig Dis*. 2011;**3**(2):126-30. [PubMed: [25197544](https://pubmed.ncbi.nlm.nih.gov/25197544/)]. [PubMed Central: [PMC4154916](https://pubmed.ncbi.nlm.nih.gov/PMC4154916/)].

14. Azarkar Z, Sharifzadeh G. Evaluation of the prevalence of hepatitis B, hepatitis C, and HIV in inmates with drug-related convictions in Birjand, Iran in 2008. *Hepat Mon.* 2010;**10**(1):26.
15. Ebrahimzadeh A, Azarkar Z, Ziaee M, Sharifzadeh G, Bijari B. Prevalence and Risk Factors of Hepatitis C Infection (HCV) in Birjand, Iran, 2014. *Int J Infect.* 2016;**3**(1). doi: [10.17795/iji-34102](https://doi.org/10.17795/iji-34102).
16. Berg T, Sarrazin C, Herrmann E, Hinrichsen H, Gerlach T, Zachoval R, et al. Prediction of treatment outcome in patients with chronic hepatitis C: significance of baseline parameters and viral dynamics during therapy. *Hepatology.* 2003;**37**(3):600–9. doi: [10.1053/jhep.2003.50106](https://doi.org/10.1053/jhep.2003.50106). [PubMed: [12601358](https://pubmed.ncbi.nlm.nih.gov/12601358/)].
17. Martinot-Peignoux M, Ripault MP, Maylin S, Boyer N, Giuily N, Castelnau C, et al. [618] optimal pretreatment viral load cut-off to predict treatment outcome in patients with chronic hepatitis C treated with peginterferon α -2b plus ribavirin. *J Hepatol.* 2007;**46**. S234. doi: [10.1016/S0168-8278\(07\)62216-8](https://doi.org/10.1016/S0168-8278(07)62216-8).
18. Zeuzem S, Fried MW, Reddy RK, Marcellin P, Diago M, Craxi A, et al. Improving the clinical relevance of pre-treatment viral load as a predictor of sustained virological response (SVR) in patients infected with hepatitis C genotype 1 treated with peginterferon alfa-2a (40KD)(PEGASYS®) plus ribavirin (COPEGUS®). *Hepatology.* 2006;**44**:267A–8A.
19. Alter HJ, Conry-Cantilena C, Melpolder J, Tan D, Van Raden M, Herion D, et al. Hepatitis C in asymptomatic blood donors. *Hepatology.* 1997;**26**(3 Suppl 1):29S–33S. doi: [10.1002/hep.510260705](https://doi.org/10.1002/hep.510260705). [PubMed: [9305660](https://pubmed.ncbi.nlm.nih.gov/9305660/)].
20. Rodger AJ, Roberts S, Lanigan A, Bowden S, Brown T, Crofts N. Assessment of long-term outcomes of community-acquired hepatitis C infection in a cohort with sera stored from 1971 to 1975. *Hepatology.* 2000;**32**(3):582–7. doi: [10.1053/jhep.2000.9714](https://doi.org/10.1053/jhep.2000.9714). [PubMed: [10960453](https://pubmed.ncbi.nlm.nih.gov/10960453/)].
21. Vogt M, Lang T, Frosner G, Klingler C, Sendl AF, Zeller A, et al. Prevalence and clinical outcome of hepatitis C infection in children who underwent cardiac surgery before the implementation of blood-donor screening. *N Engl J Med.* 1999;**341**(12):866–70. doi: [10.1056/NEJM199909163411202](https://doi.org/10.1056/NEJM199909163411202). [PubMed: [10498458](https://pubmed.ncbi.nlm.nih.gov/10498458/)].
22. Vossughinia H, Goshayeshi L, Bayegi HR, Sima H, Kazemi A, Erfani S, et al. Prevalence of hepatitis C virus genotypes in mashhad, north-east iran. *Iran J Public Health.* 2012;**41**(9):56–61. [PubMed: [23193507](https://pubmed.ncbi.nlm.nih.gov/23193507/)]. [PubMed Central: [PMC3494216](https://pubmed.ncbi.nlm.nih.gov/PMC3494216/)].
23. Selcuk H, Kanbay M, Korkmaz M, Gur G, Akcay A, Arslan H, et al. Distribution of HCV genotypes in patients with end-stage renal disease according to type of dialysis treatment. *Dig Dis Sci.* 2006;**51**(8):1420–5. doi: [10.1007/s10620-005-9025-9](https://doi.org/10.1007/s10620-005-9025-9). [PubMed: [16868830](https://pubmed.ncbi.nlm.nih.gov/16868830/)].
24. Bozdayi G, Rota S, Verdi H, Derici U, Sindel S, Bali M, et al. [The presence of hepatitis C virus (HCV) infection in hemodialysis patients and determination of HCV genotype distribution]. *Mikrobiyol Bul.* 2002;**36**(3-4):291–300. Turkish. [PubMed: [12838663](https://pubmed.ncbi.nlm.nih.gov/12838663/)].
25. Kabir A, Alavian SM, Keyvani H. Distribution of hepatitis C virus genotypes in patients infected by different sources and its correlation with clinical and virological parameters: a preliminary study. *Comp Hepatol.* 2006;**5**:4. doi: [10.1186/1476-5926-5-4](https://doi.org/10.1186/1476-5926-5-4). [PubMed: [17014721](https://pubmed.ncbi.nlm.nih.gov/17014721/)]. [PubMed Central: [PMC1599752](https://pubmed.ncbi.nlm.nih.gov/PMC1599752/)].
26. Somi MH, Keivani H, Ardalan MR, Farhang S, Pouri AA. Hepatitis C virus genotypes in patients with end-stage renal disease in East Azerbaijan, Iran. *Saudi J Kidney Dis Transpl.* 2008;**19**(3):461–5. [PubMed: [18445914](https://pubmed.ncbi.nlm.nih.gov/18445914/)].
27. Altuglu I, Soyler I, Ozacar T, Erensoy S. Distribution of hepatitis C virus genotypes in patients with chronic hepatitis C infection in Western Turkey. *Int J Infect Dis.* 2008;**12**(3):239–44. doi: [10.1016/j.ijid.2007.07.003](https://doi.org/10.1016/j.ijid.2007.07.003). [PubMed: [17942359](https://pubmed.ncbi.nlm.nih.gov/17942359/)].
28. Turkoglu S, Bozaci M, Bozfakioglu S, Kaymakoglu S, Badur S. Molecular epidemiology of hepatitis C virus in Istanbul, Turkey. *J Microbiol Methods.* 1996;**27**(1):98. doi: [10.1016/0167-7012\(96\)83736-5](https://doi.org/10.1016/0167-7012(96)83736-5).
29. Gower E, Estes C, Blach S, Razavi-Shearer K, Razavi H. Global epidemiology and genotype distribution of the hepatitis C virus infection. *J Hepatol.* 2014;**61**(1 Suppl):S45–57. doi: [10.1016/j.jhep.2014.07.027](https://doi.org/10.1016/j.jhep.2014.07.027). [PubMed: [25086286](https://pubmed.ncbi.nlm.nih.gov/25086286/)].
30. Tyczyno M, Halota W, Nowak W, Pawlowska M. Distribution of HCV genotypes in the populations of inmates in polish prison potulice and patients hospitalised in bydgoszcz. *Hepat Mon.* 2014;**14**(5). e14559. doi: [10.5812/hepatmon.14559](https://doi.org/10.5812/hepatmon.14559). [PubMed: [24910703](https://pubmed.ncbi.nlm.nih.gov/24910703/)]. [PubMed Central: [PMC4030260](https://pubmed.ncbi.nlm.nih.gov/PMC4030260/)].
31. Samimi-Rad K, Hosseini M, Mobeini G, Asgari F, Alavian SM, Tahaei ME, et al. Hepatitis C virus infection among multi-transfused patients and personnel in haemodialysis units in central Islamic Republic of Iran. *East Mediterr Health J.* 2012;**18**(3):227–35. [PubMed: [22574475](https://pubmed.ncbi.nlm.nih.gov/22574475/)].
32. Jahanbakhsh Sefidi F, Keyvani H, Monavari SH, Alavian SM, Fakhim S, Bokharaei-Salim F. Distribution of hepatitis C virus genotypes in Iranian chronic infected patients. *Hepat Mon.* 2013;**13**(1). e7991. doi: [10.5812/hepatmon.7991](https://doi.org/10.5812/hepatmon.7991). [PubMed: [23550108](https://pubmed.ncbi.nlm.nih.gov/23550108/)]. [PubMed Central: [PMC3582303](https://pubmed.ncbi.nlm.nih.gov/PMC3582303/)].
33. Lee H, Cho YK, Kim HU, Choi EK, Hyun S, Kang D, et al. [Distribution of hepatitis C virus genotypes in Jeju Island]. *Korean J Hepatol.* 2008;**14**(1):28–35. doi: [10.3350/kjhep.2008.14.1.28](https://doi.org/10.3350/kjhep.2008.14.1.28). [PubMed: [18367855](https://pubmed.ncbi.nlm.nih.gov/18367855/)].
34. Hosseini-Moghaddam SM, Keyvani H, Kasiri H, Kazemeyni SM, Basiri A, Aghel N, et al. Distribution of hepatitis C virus genotypes among hemodialysis patients in Tehran—a multicenter study. *J Med Virol.* 2006;**78**(5):569–73. doi: [10.1002/jmv.20577](https://doi.org/10.1002/jmv.20577). [PubMed: [16555284](https://pubmed.ncbi.nlm.nih.gov/16555284/)].
35. Samimi-Rad K, Nasiri Toosi M, Masoudi-Nejad A, Najafi A, Rahimnia R, Asgari F, et al. Molecular epidemiology of hepatitis C virus among injection drug users in Iran: a slight change in prevalence of HCV genotypes over time. *Arch Virol.* 2012;**157**(10):1959–65. doi: [10.1007/s00705-012-1369-9](https://doi.org/10.1007/s00705-012-1369-9). [PubMed: [22695769](https://pubmed.ncbi.nlm.nih.gov/22695769/)].
36. Samimi-Rad K, Shahbaz B. Hepatitis C virus genotypes among patients with thalassemia and inherited bleeding disorders in Markazi province, Iran. *Haemophilia.* 2007;**13**(2):156–63. doi: [10.1111/j.1365-2516.2006.01415.x](https://doi.org/10.1111/j.1365-2516.2006.01415.x). [PubMed: [17286768](https://pubmed.ncbi.nlm.nih.gov/17286768/)].
37. Micalessi MI, Gerard C, Ameye L, Plasschaert S, Brochier B, Vranckx R. Distribution of hepatitis C virus genotypes among injecting drug users in contact with treatment centers in Belgium, 2004–2005. *J Med Virol.* 2008;**80**(4):640–5. doi: [10.1002/jmv.21145](https://doi.org/10.1002/jmv.21145). [PubMed: [18297717](https://pubmed.ncbi.nlm.nih.gov/18297717/)].
38. Dal Molin G, Ansaldi F, Biagi C, D'Agaro P, Comar M, Croce L, et al. Changing molecular epidemiology of hepatitis C virus infection in Northeast Italy. *J Med Virol.* 2002;**68**(3):352–6. doi: [10.1002/jmv.10210](https://doi.org/10.1002/jmv.10210). [PubMed: [12226821](https://pubmed.ncbi.nlm.nih.gov/12226821/)].
39. Sharara AI, Ramia S, Ramlawi F, Fares JE, Klayme S, Naman R. Genotypes of hepatitis C virus (HCV) among positive Lebanese patients: comparison of data with that from other Middle Eastern countries. *Epidemiol Infect.* 2007;**135**(3):427–32. doi: [10.1017/S0950268806006911](https://doi.org/10.1017/S0950268806006911). [PubMed: [16848924](https://pubmed.ncbi.nlm.nih.gov/16848924/)]. [PubMed Central: [PMC2870582](https://pubmed.ncbi.nlm.nih.gov/PMC2870582/)].
40. Samimi-Rad K, Nategh R, Malekzadeh R, Norde H, Magnius L. Molecular epidemiology of hepatitis C virus in Iran as reflected by phylogenetic analysis of the NS5B region. *J Med Virol.* 2004;**74**(2):246–52. doi: [10.1002/jmv.20170](https://doi.org/10.1002/jmv.20170). [PubMed: [15332273](https://pubmed.ncbi.nlm.nih.gov/15332273/)].