

## **Renal Data from Asia-Africa**

### **Hepatitis C virus Genotypes in Patients with End-Stage Renal Disease in East Azerbaijan, Iran**

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**ABSTRACT.** Information about the genotypes and associated risk factors in hepatitis C virus (HCV) infected patients in Iran is limited. The aim of this study was to identify the HCV genotypes and associated risk factors in a group of HCV infected patients on dialysis therapy in Iran. The sera of 753 patients with chronic renal failure from fifteen dialysis units in East Azerbaijan Province were screened for anti-HCV antibodies as well as HCV RNA; viral RNA was extracted for the genotype specific primer approach. Patients were questioned concerning documented risk factors. Genotyping analysis was performed in 55 patients with positive anti-HCV and HCV-RNA. Genotypes 1 and 3 were found in 46 (83.7%) and three (5.5%) patients, respectively. The most frequent HCV subtype was 1a (76.4%), followed by 3a and 1b and 1b (5.5% each) while one patient was infected with both 1a and 1b. There was no statistically significant difference between the risk factors analyzed and the acquisition of HCV infection. This study gives added evidence of the predominant HCV genotypes in Iran, which is different than reports from other Arab countries and similar with the pattern of genotype in both Europe and United States.

**Keywords:** Hepatitis C genotype, Chronic renal failure, Dialysis, East Azerbaijan

#### **Introduction**

Infection with the hepatitis C virus (HCV)

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is the most frequent cause of chronic hepatitis. It is estimated that 170 million persons are chronically infected with HCV. About 80% of newly infected patients progress to develop chronic infection, which may result in cirrhosis and hepatocellular carcinoma, carrying a high morbidity and mortality.

The HCV is a blood-borne pathogen and the modes of transmission of the disease are parenteral and, to a lesser extent, sexual. It is

a major cause of chronic liver disease among patients with chronic renal failure (CRF) undergoing maintenance hemodialysis (HD).<sup>1</sup> Epidemiological studies about HCV infection among HD patients in Iran have reported a prevalence of 5.5 to 55.9% in different cities<sup>2-4</sup> while the reported prevalence in the general population in Iran is less than 1%.<sup>5</sup> The mortality rate as a result of liver disease has been reported to be significantly higher in anti-HCV positive patients than their negative counterparts.<sup>6</sup>

HCV genotypes have been of considerable epidemiological concern and their prevalence varies in different parts of the world. The nucleotide sequence of HCV may vary considerably from one isolate to another and according to the most accepted classification, HCV falls into six major genotypes and more than 100 subtypes.<sup>7</sup> The clinical significance and natural history of the disease in relation to genotype and subtype is controversial. There is evidence to suggest that patients with HCV subtype 1b have a poorer response to treatment with interferon alpha.<sup>8,9</sup>

Previous studies from Iran show a difference between the dominant genotypes of HCV in Iran compared to other countries in the Middle East. The aim of this study was to reveal the distribution of different HCV genotypes in East Azerbaijan located in North East Iran, where consistent data is seriously lacking.

## Patients and Methods

### *Study population*

This survey was carried out in fifteen dialysis units in East Azerbaijan (over 3,500,000 inhabitants). Between January and March 2006, 753 patients including 674 on HD and 61 on continuous ambulatory peritoneal dialysis (CAPD), were asked to take part in this study, and informed consent was obtained

from all.

A standardized form was used to collect socio-demographic data such as number of previous blood transfusions, length of time on dialysis, kidney transplantation, war injury, tattooing, intravenous drug use, surgical interventions, multiple sex partners, and possible household contact with hepatitis. The duration on HD treatment was grouped into 12, 12–48 and more than 48 months.

The studied population ranged in age from 7 to 91 years (mean  $52.1 \pm 16.9$  years) and included 409 males (54.3%) and 344 females (45.7%). The etiology of CRF was diabetic nephropathy (n=166), hypertension (n=184), chronic glomerulonephritis (n=73), polycystic kidney disease (n=67), nephrolithiasis (n=31), pyelonephritis (n=13), renal diseases of unknown etiology (n=171), and others (systemic lupus erythematosus, Alport's syndrome, drug induced) (n = 47).

The diagnosis of chronic hepatitis C was made on the basis of the presence of anti-HCV antibodies in sera detected by third generation enzyme-linked immunosorbent assay (ELISA) kits (Abbott Laboratories, Chicago, US) and for HCV-RNA as tested by PT-PCR amplification of 5'-UTR with nested primers. HCV genotyping was performed in accordance with the procedure described by Ohno et al.<sup>10</sup>

### *Statistical analysis*

Prevalence and 95% confidence intervals (CI) were calculated. Chi-square test or Fisher's exact test were performed to evaluate the distribution of characteristics associated with HCV infection. Statistical significance was assessed at the 0.05 probability level in all analyses. Statistical evaluations were performed using SPSS package (ver 13).

## Results

A predominant prevalence of genotype 1a

Table 1. Characteristics of patients infected with HCV according to the genotype

Factor	1a	1b	1a/1b	3a
No. of patients (%male)	42 (40.5%)	3 (33.3%)	1 (100%)	3 (66.7%)
Age	50.0 ± 13.9	42.0 ± 14.6	45	32.0 ± 1.0
<b>Possible mode of transmission</b>				
Sexual (%)	1 (2.4%)	0	0	0
IVDA (%)	1 (2.4%)	0	0	1 (3.3%)
Hemodialysis (%)	37 (88.1%)	3 (100%)	1 (100%)	3 (100%)
Blood transfusion (%)	9 (21.4%)	0	1 (100%)	0
Surgical intervention (%)	32 (76.9%)	2 (66.7%)	0	1 (33.3%)
Transplantation (%)	22 (64.7 %)	1 (33.3%)	0	2 (66.7%)
Infected with HBV	0	0	1 (100%)	0

IVDA = Intravenous drug abuse

(42 patients, 76.4%) was found in fifty-five HCV-RNA positive patients with CRF. Three patients (5.5%) were infected with genotype 1b, three others (5.5%) with genotype 3a, one (1.8%) had mixed infection with 1a and 1b and only four (10.9%) had non-typeable genotype.

Table 1 compares the distribution of subtypes of HCV genotypes in relation to age, sex, and possible source of infection. Neither the presence of anti-HBsAb nor the etiology of CRF had significant bearing on the subtypes of HCV genotypes.

The prevalence of HCV-RNA in patients undergoing HD was related to the duration of treatment ( $p < 0.005$ ). None of the patients who were receiving HD for less than one year were infected while 3.8% and 14.9% of patients under treatment for one to three years and more than three years respectively, were infected with HCV.

Patients with history of blood transfusion(s) seemed to be at significantly higher risk for acquiring infection with HCV ( $p = 0.001$ ). Anti-HCV was positive in 0%, 4.2%, 11.7% and 100% of patients who had received one, two, three and more than three units of blood transfusion, respectively ( $p = 0.004$ ).

Distribution of infection with HCV and its genotypes in the dialysis centers studied was not significantly different. Logistic regression model for risk factors for acquiring HCV showed that the duration ( $p < 0.005$ ) and frequency ( $p = 0.002$ ) of HD therapy, intravenous drug use ( $p < 0.005$ ) and history of transplantation ( $p < 0.005$ ) were good predictive factors; however, there were no predictors of the genotype. The HD center, etiology of CRF, war injury, tattooing, surgical interventions, multiple sex partners, and possible house-hold contact with hepatitis were not good predictors ( $p = NS$ ).

## Discussion

This is the first study to report on the seroprevalence and genotyping of HCV in HD patients in east Azerbaijan. A predominant prevalence of genotype 1 was found among our study patients. This is in accordance with the predominance of genotype 1 observed in most countries worldwide. With respect to the zero frequency of genotype 4, our data differ from those published for patients in the United States, Europe, and even Asia, which showed a different prevalence of genotype 4.

The first study on the prevalence of specific

genotypes of HCV from Iranian population was conducted by Zali and his group<sup>11</sup> in Tehran, who showed Type I/1, Type II/1b and Type V/3a in seven, three and four patients respectively. A recently published article by Samimi-Rad et al<sup>12</sup> in Iranian patients who were anti-HCV Ab positive, from Tehran and five cities from different locations of Iran, showed that genotype 1a was predominant (47%), and 3a, 1b, and 4 were seen in 36%, 8%, and 7% of the patients, respectively.

The pattern of our genotypes are similar to those reported from England,<sup>13</sup> but it is different from other Middle East countries such as Republic of Yemen, Kuwait, Iraq, and Saudi Arabia, where genotype 4 is the most prevalent.<sup>14</sup> HCV genotype 4 was detected in 50% and genotype 1b was found in nearly 40% of Saudi patients.<sup>15,16</sup>

In another large study, the predominance of HCV genotype 4 was reported in the Arab world, Lebanon and most other countries in the Middle East.<sup>17</sup> However, subtype 1b is prevalent in Turkey<sup>18,19</sup> and the western border of Iran and subtype 3a is prevalent in Pakistan and the eastern border of Iran.<sup>20</sup> HCV 1b was found to be the predominant virus type both among blood donors and chronic hepatitis patients.<sup>21</sup>

Although genotype 4 is reported almost entirely from the Middle East and western countries,<sup>22</sup> this genotype is uncommon in our country.<sup>23</sup> Similar observation has been reported from other provinces of Iran as well.

Genotypes 3a and 1a are more prevalent in intravenous drug abusers (IVDA) in Europe and USA.<sup>23-25</sup> In the present study, one patient (2.4%) with IVDA had genotype 1a. It seems that there is a high similarity between the pattern of genotype in IVDA in Europe and United States when compared with Iran. In the present study, most of the patients seem to have several routes of contamination, which limits the conclusion on

association between genotype and route of contamination. There was no difference in genotypes in terms of age and sex of the patients. This pattern is different when compared to reports from developed countries, where the molecular epidemiology of HCV seems to be influenced by lifestyles among young adults as reported from the USA and Southeast Asia in their young drug addicts.<sup>26</sup>

This study involved all of the CRF patients on dialysis in the province and the seropositivity to HCV increased significantly with increase in the duration and frequency of HD and history of transplantation, suggesting possible nosocomial transmission between patients.

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