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Characterization of the Interleukin-28B Gene rs12979860 C/T Polymorphism in Turkish Chronic Hepatitis C Patients and Healthy Individuals

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Background: Host genetic factors can affect the progress of hepatitis-C virus (HCV) infection. Interleukin-28B (IL28B) single nucleotide polymorphisms may play an important role in the clearance of HCV spontaneously or with treatment.

Aims: The aim of our study was to evaluate the rate of IL28B genotypes in patients with Chronic Hepatitis-C (CHC) and healthy control subjects and to examine the characteristics of patients in each IL28B subgroup.

Study Design: Case-control study.

Methods: IL28B polymorphisms were genotyped by Polymerase Chain Reaction and Restriction Fragment Length Polymorphism (PCR-RFLP) in all subjects.

Results: The mean age was 52.3±10.9 years (33% female) in the CHC patients and 52.5±11.5 years (39.1% female) in the healthy controls. The percentage of patients with a high baseline viral load (≥400,000 IU/mL) was higher in the CT group (69.8%) compared to the C/C (44.4%) and T/T (50%) groups (p=0.021). There was no significant difference in liver fibrosis and liver necroinflammation distribution among the CC, CT and TT genotypes with mild, moderate and severe groups (p=0.058 and p=0.791, respectively). Mean age, gender ratio, body mass index, viral load at baseline, rate of HCV genotypes, baseline ALT levels were not significantly different among the three IL28B subgroups (p>0.05). A significant increase was observed in the frequencies of IL28B rs12979860 TT genotypes in the CHC patients (20.6%) compared to the healthy control group (8.7%) (p=0.033).

Conclusion: In the patients with chronic HCV-genotype 1b and 4 infections, the IL28B rs12979860 (C>T) gene polymorphism frequency of the TT genotype and T allele was higher than in healthy control subjects. This result indicates that the TT genotype may be more effective in the progression of HCV infection than other genotypes.

Keywords: Chronic hepatitis C, gene polymorphism, IL28B, interleukins

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The hepatitis-C virus (HCV) is an important global health problem. HCV causes chronic liver disease, leading to complications such as liver cancer and cirrhosis (1-4). In Turkey, the prevalence of HCV between 2000 and 2006 was 0.54% (0.2-5.2%) in 1,076,495 blood donors (5). In addition, the contribution of HCV to chronic hepatitis etiology increased from 23% to 38.1% and its contribution to cirrhosis increased from 25.2% to 45.9% in the last 10 years (6). The most common HCV genotype in Turkey is genotype 1b, which occurs in 68-94% of the population (7), although there has been a recent increase in genotype 4 infections (8). Unfortunately, HCV treatment is difficult and the possibility of a sustained virological response (SVR) through treatment is low in chronic hepatitis-C (CHC) caused by these genotypes. Thus, when the frequency of HCV infection, common genotype and high cost of CHC treatment are taken into consideration, it is clear that patient treatments should be chosen carefully.

There are many factors affecting the progression of HCV infection and its response to treatment in chronic cases (9-12). In multiple Genome Wide Association Studies (GWAS), a single nucleotide polymorphism (SNP) of the IL28B gene 3-kb upstream (rs12979860 C>T), encoded by Interferon-13 (IFN-l3), had a substantial role in the disappearance of HCV infection spontaneously or with treatment (13). Recent studies have determined that IL28B rs12979860 is the primary factor contributing to healing from infection (14-16), increasing response to treatment by almost two-fold (17,18). Moreover, the rates of SVR change based on genetic polymorphism in the IL28B gene, which is low in African Americans and Caucasians but high in Asian populations (19). Furthermore, the IL28B genotype is a very strong predictor for SVR in treatment protocols that include a protease inhibitor or in Interferon (IFN) plus ribavirin combination treatments (20,21).

The aim of this study was to investigate the relationship between the IL28B rs12979860 (C>T) polymorphism in Turkish patients with chronic-HCV genotype-1b and 4 infections and healthy control subjects, and to analyze the characteristics of the patients in each IL28B subgroup.

MATERIALS AND METHODS

Patients and healthy subjects

This study was conducted with 136 patients diagnosed with CHC (45 female and 91 male) and a control group of 92 healthy individuals (36 female and 56 male). The patients were monitored in the Infectious Diseases Outpatient Clinic from 2010 to 2013 years. The mean age was 52.3±10.9 years (age range 19-70). CHC was diagnosed in cases with compensated disease through liver biopsy, normal or high liver enzymes, anti-HCV antibodies and positive HCV RNA. We excluded patients who had other chronic liver diseases, positive hepatitis B surface antigen (HBsAg) and human immunodeficiency virus (HIV). The liver biopsies taken percutaneously were evaluated using the Ishak scoring system (22), which categorizes fibrosis with a score of 0-2 as mild, 3-4 as moderate and 5-6 as severe (23). Subjects in the healthy control group had a mean age of 52.5±11.5 years (age range 18-70) and none had any hepatic disease (*i.e.,* infection, long term drug use, autoimmune-hepatitis or alcohol consumption) or endocrine disease and the results of their liver function tests and liver ultrasonography were normal.

For all patients, demographics, body mass indices (BMI), alanine aminotransferase (ALT) HCV RNA levels, viral genotypes were recorded on private forms designed for the study, liver biopsy necroinflammation and fibrosis results were evaluated based on the Ishak scoring system Liver biopsy was performed in 109/136 patients. All subjects provided written informed consent for genetic analysis. The study was approved by the Ethics Committee for Clinical Research, which conforms to protocols in accordance with the Declaration of Helsinki (Decision number: 2013-188).

Blood samples and laboratory tests

Routine biochemical tests were performed from venous blood samples with an automated device and HBsAg, anti-HCV, and-HIV antibodies examined via an enzyme immunoassay method (anti-HCV and anti-HIV: Architect System, Abbott Diagnostics, Germany; HBsAg: Roche Diagnostics, USA). Quantitative HCV RNA levels were performed using real-time polymerase chain reaction (PCR, COBAS Amplirep/COBAS TaqMan 48, Roche Molecular Systems, USA) and the HCV genotype was determined through pyrosequencing according to the directions of the manufacturer (Qiagen, Hilden, Germany). Blood samples were taken into vacutainer tubes containing Ethylene Diamine Tetraacetic Acid (EDTA).

IL-28B rs12979860 (C>T) Polymorphism Detection

Genotyping of the IL-28B rs12979860 (C>T) polymorphism was performed by a PCR-RFLP method. DNA was extracted from whole blood samples using the DNA blood mini kit (Qiagen, Milan, Italy). After DNA isolation, DNA samples were stored at -80°C.

Using genomic DNA samples, a 139 base pair (bp) product was amplified with the following primers: forward primer, IL28BF 5'-CCAGGGCCCCTAACCTCTGCA-3'; reverse primer, IL28BR 5'-GGGAGCGCGGAGTGCAATTCA-3. Amplification was carried out in a total volume of 50 µL

containing 10 mmol/L Tris-HCl (pH 8.3), 50 mmol/L KCl, Tween-20 0.01%, 0.2 mmol/L deoxyribonucleotides, 2-5 pmol of each primer, 2.0 mmol/L $MgCl_2$, 0.5 units Taq DNA polymerase (Thermo Taq, Pittsburgh PA, USA) and ~10 ng genomic DNA. The thermal protocol for amplification included 36 cycles of denaturation at 94°C for 60 s, annealing at 62°C for 50 s, and elongation at 72°C for 60 s. The PCR products were digested with 1 unit *BstUI* (New England Biolabs, Hitchin, UK) in a total volume of 25 µL at 37°C for six hours. The fragments were resolved by electrophoresis in a 3% agarose gel, followed by staining with ethidium bromide. A band of 139 bp indicates the TT genotype, 109 bp indicates the CC genotype, and two bands (139 and 109 bp) indicate the CT genotype (24).

Statistical analysis

To assess data normality, histogram and q-q plots were plotted and data analyzed using the Shapiro-Wilk test. The Levene test was used to assess variance homogeneity. For baseline viral load variables, a logarithmic transformation was applied to account for a highly skewed distribution. To compare the differences between groups, independent samples t test, Mann-Whitney U test, one-way analysis of variance, Kruskal-Wallis test and Pearson's χ^2 analysis were used. Multiple comparisons were made using 2-sample chi-square test for equality of proportions with continuity correction. Data were expressed as frequencies and percentages, means and standard deviations, geometric means and 95% confidence intervals, or medians and interquartile ranges. Analysis was performed using the R 3.0.1 software (www.r-project.org) and a value of $p<0.05$ was taken to indicate statistical significance. R is an open source and free software environment for statistical computing and graphics. It includes more than 5.000 statistical packages in CRAN network, also has about 1.000 packages in BIOCONDUCTOR network for bioinformatics analysis. R can be assessed from www.r-project.org.

RESULTS

Patient and healthy control demographics

The demographic characteristics of 136 CHC patients and 92 healthy controls are shown in Table 1. The mean BMI was 26.3±4.0 kg/m^2 in the CHC patients and 26.5±5.9 kg/m^2 in the healthy controls. Based on the BMI, 39% of the CHC patients were normal weight, 47.8% were overweight and 13.2% were obese. In the control group, 35% were of normal weight, 52% were overweight, and 13% were obese. There was no significant difference in mean age, gender distribution or BMI between the control and patient groups ($p>0.05$).

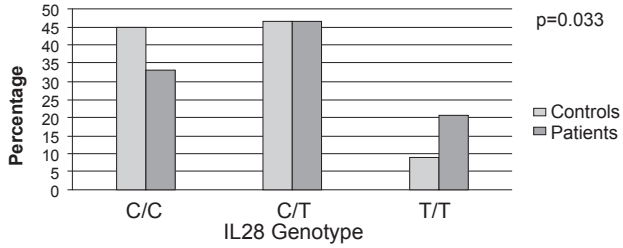


FIG. 1. IL28B rs12979860 C/T polymorphism in control and in patients with chronic HCV infection

The mean HCV RNA level of the CHC patients was 307,094 IU/mL. For 55.1% of the patients, viral load was ≥400,000 IU/mL and 90.4% were infected with genotype 1b. The mean ALT level was 48 IU/L. A liver biopsy was carried out in 109 patients. The severity of liver fibrosis was mild for 63.2% of the patients, moderate for 14.7% and severe for 2.2%. The distribution of liver necroinflammation was found as 62.5% for mild, 16.9% for moderate and 0.7% for severe.

IL28B genotypes

When the IL28B genotype was assessed in patients with CHC, the CC genotype was identified in 32.1%, the CT genotype in 46.3% and the TT genotype in 20.6%. In the control group, the CC genotype was identified in 44.6%, the CT genotype in 46.7%, and the TT genotype in 8.7%. There was a significant difference in genotype distribution between the patient and control groups ($p=0.033$; Figure 1). Based on the multiple comparisons, the significance occurred between the frequency differences of the CC and TT genotypes ($p=0.016$). The frequency of the TT genotype was higher in the CHC patient group compared to the control group. Moreover, patients with the CT genotype had 1.33-fold and those with TT had 3.19-fold increased risk of HCV infection when compared to patients with the CC genotype (Table 2).

In the patient group, the C allele frequency was 0.56 (56%) and T allele frequency was 0.44 (44%), while in the control group, the C allele frequency was 0.68 (68%) and the T allele frequency was 0.32 (32%); this resulted in a significant difference in the frequencies of C and T alleles between the groups ($p=0.011$). We found that patients with the T allele had a 1.67-fold increased risk of chronic HCV infection compared to those with the C allele (Table 2).

Characteristics of the CHC patients based on the IL28B genotype

The characteristics of the CHC patients based on the IL28B genotype subgroups are summarized in Table 3. The geometric mean baseline HCV RNA levels were 209,779 IU/mL in the CC group, 397,049 IU/mL in the CT group and 317,854

TABLE 1. The characteristics of Chronic Hepatitis-C patients and healthy control group

Variable	Chronic HCV (n=136)	Control subjects (n=92)	Test Statistic	p	OR (95% CI)
Gender, n (%)					
Female	45 (33.0)	36 (39.1)	χ^2=0.875	0.350	1.00
Male	91 (67.0)	56 (60.9)			1.30 (0.75-2.27)
Age (years), mean±SD	52.3±10.9	52.5±11.5	t=0.084	0.933	1.00 (0.98-1.02)
BMI (kg/m^2), mean±SD	26.3±4.0	26.5±5.9			0.99 (0.94-1.05)
Normal weight (<25 kg/m^2), n (%)	53 (39.0)	32 (35.0)			1.00
Overweight (25-29.9 kg/m^2), n (%)	65 (47.8)	48 (52.0)	t=0.259	0.796	0.82 (0.46-1.45)
Obese (≥30 kg/m^2), n (%)	18 (13.2)	12 (13.0)	χ^2=0.472	0.790	0.91 (0.39-2.12
Viral load at baseline (IU/mL) geometric mean (95% CI)[a]	307.094 (193.524-506.006)				
<400,000 IU/mL, n (%)	61 (44.9)				
≥400,000 IU/mL, n (%)	75 (55.1)				
Genotype, n (%)					
Genotype 1b	123 (90.4)				
Genotype 4	13 (9.6)				
Baseline ALT level (IU/L), median (1st-3rd quartiles)	48 (26.2-59.0)				
Distribution of liver necroinflammation[b], n (%)					
Mild	85 (62.5)				
Moderate	23 (16.9)				
Severe	1 (0.7)				
Distribution of liver fibrosis[b], n (%)					
Mild	86 (63.2)				
Moderate	20 (14.7)				
Severe	3 (2.2)				

BMI: body mass index; ALT: alanin aminotransferase; SD: standard deviation; OR: odds ratio; CI: confidence interval.
The tests applied are independent samples t test where mean±SD is given and Pearson's chi-square test where n (%) is given.
OR=1 corresponds to reference category.
[a]Logarithmic transformation (base 10) was applied. Confidence intervals of geometric mean were computed using 1000 bootstrap samples.
[b]109 patients with liver biopsy were evaluated according to the Ishak score.

TABLE 2. Allele and genotype frequencies of IL28B rs12979860 (C>T) polymorphism in control and patients with chronic HCV infections

IL28B	Chronic HCV infection (n=136)	Control subjects (n=92)	Test Statistic	p	OR (95%CI)
rs12979860 allele frequency	C=0.56	C=0.68	χ^2=6.686	0.011	1.00
	T=0.44	T=0.32			1.67 (1.14-2.50)
rs12979860	CC=45 (32.1)	CC=41 (44.6)	χ^2=6.834	0.033	1.00
	CT=63 (46.3)	CT=43 (46.7)			1.33 (1.10-1.51)
	TT=28 (20.6)	TT=8 (8.7)			3.19 (2.87-3.49)
Pairwise comparisons					
CC and CT	-	-	χ^2=0.707	0.400	-
CC and TT	-	-	χ^2=5.823	0.016	-
CT and TT	-	-	χ^2=3.172	0.075	-

OR: odds ratio; CI: confidence interval. The tests applied are Pearson's chi-square test. Post-hoc tests are applied using 2-sample chi-square test for equality of proportions with continuity correction. OR=1 corresponds to reference category.

IU/mL in the TT group. The frequency of patients with a viral load ≥400,000 IU/mL was significant among patients with different genotypes (p=0.021). Based on multiple comparisons, the frequency in patients with the CC genotype was found to be lower than in patients with the CT genotype (p=0.014).

TABLE 3. Patients characteristics in each IL28B genotype subgroup

Variables	Genotypes C/C (n=45)	C/T (n=63)	T/T (n=28)	Test Statistic	p	Pairwise Comparisons C/C and C/T	C/C and T/T	C/T and T/T
Age (years), mean±SD	51.4±12.5	53.1±10.3	52.3±9.6	F=0.304	0.738	-	-	-
Male, n (%)	28 (62.2)	45 (71.4)	18 (64.3)	χ^2=1.115	0.573	-	-	-
BMI (kg/m^2), mean±SD	26.4±4.1	26.6±4.1	25.5±3.6	F=0.913	0.476			
Viral load at baseline (IU/mL) geometric mean (95%CI)[a]	209.779 (79.943-542.676)	397.049 (206.916-728.103)	317.854 (96.161-861.159)	F=0.646	0.526	-	-	-
Viral load ≥400,000 IU/mL, n (%)	20 (44.4)	44 (69.8)	14 (50.0)	χ^2=7.702	0.021	χ^2=6.00, p=0.014	χ^2=0.05, p=0.825	χ^2=2.50, p=0.114
Genotype 1b, n (%)	43(36.6)	55(44.7)	25(20.3)	χ^2=0.654	0.721	-	-	-
Genotype 4, n (%)	3 (6.7)	7 (11.1)	3 (10.7)					
Baseline ALT level (IU/L), median (1st-3rd quartiles)	39.0 (26.0-53.0)	48.0 (27.0-59.0)	57.0 (29.5-75.0)	χ^2=3.121	0.210	-	-	-
Distribution of liver necroinflammation[b], n (%)								
Mild (1-6)	28 (82.4)	40 (76.9)	17 (73.9)	χ^2=1.699	0.791	-	-	-
Moderate (7-12)	6 (17.6)	11 (21.2)	6 (26.1)					
Severe (13-18)	0 (0.0)	1 (1.9)	0 (0.0)					
Distribution of liver fibrosis[b], n (%)								
Mild (0-2)	31 (91.2)	35 (67.3)	20 (87.0)	χ^2=9.130	0.058	-	-	-
Moderate (3-4)	3 (8.8)	14 (26.9)	3 (13.0)					
Severe (5-6)	0 (0.0)	3 (5.8)	0 (0.0)					

BMI: body mass index; ALT: alanin aminotransferase; SD: standard deviation; CI: confidence interval.
The tests applied are one-way analysis of variance where mean±SD or geometric mean (95% CI) is given, Kruskal-Wallis H test where median (1st-3rd quartiles) is given and Pearson's chi-square test where n (%) is given. Post-hoc tests are applied using 2-sample chi-square test for equality of proportions with continuity correction.
[a]Logarithmic transformation (base 10) was applied. Confidence intervals of geometric mean were computed using 1000 bootstrap samples.
[b]Patients with liver biopsy were evaluated according to the Ishak score.
[c]34 patients
[d]52 patients
[e]23 patients

We also grouped genotypes according to the presence of the T allele as CC versus CT+TT and the results were similar. Only viral load ≥400,000 IU/mL / <400,000 IU/mL) was found to be statistically significant between genotypes, as shown in Table 3 ($p<0.05$).

The liver fibrosis score was grouped into mild (0-2), moderate (3-4) and severe (5-6) and the necroinflammation score was grouped as mild (0-6), moderate (7-12) and severe (13-18). There was no significant difference between the CC, CT and TT genotypes in the mild, moderate and severe groups for fibrosis and necroinflammation distributions (p=0.058 and p=0.791, respectively) (Figure 2). There was no significant difference between IL28B genotypes in terms of mean age (p=0.738), gender distribution (p=0.573), mean BMI (p=0.476), geometric mean viral load (p=0.526), genotype 1b or genotype 4 distributions (p=0.721) and median ALT level (p=0.210) (Table 3).

DISCUSSION

Chronic-HCV infection is a significant problem in Turkey. Although the Türkiye Hepatitis Prevalence (TURKHEP) study reported a nationwide seroprevalence of 1%, some regions have results as high as 3.1% (25,26). Prevalence increases after the age of 50. Yildirim et al. (27) reported a prevalence of 4.2% in 50-59 year-olds, 3.4% in 60-69 year-olds, and 7.1% 70-79 year-olds. The cost of pegylated-IFN and ribavirin combination used in CHC treatment is high and it has important side effects. When protease inhibitors used for the treatment of genotype-1 infected patients is considered, it is clear that the cost and frequency of side effects are much higher. In Turkey, the most common HCV genotype is genotype 1, although genotype 4 is seen more frequently in our center and in our neighborhood, possibly due to immigration (8). Both genotypes belong to the group in which permanent virologi-

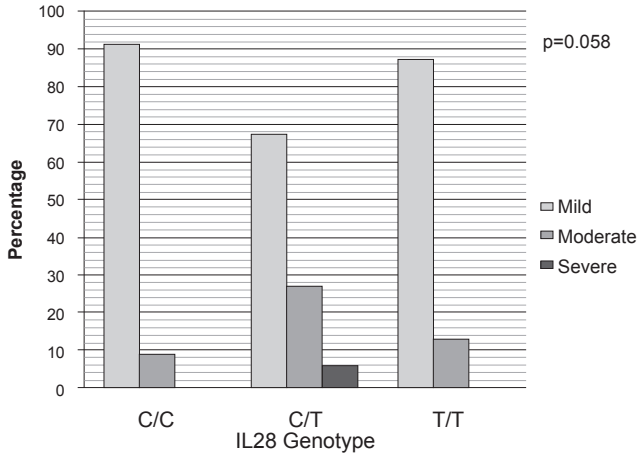


FIG. 2. The relationship between fibrosis score and IL28B rs12979860 C/T polymorphism in patients with chronic HCV infection

cal response to the drugs used in CHC is low. For these reasons, before beginning treatment, it is important to predict the response of the patient. In the studies conducted in Turkey, one-third of CHC cases were of the IL28B rs12979860 CC genotype and almost half were of the IL28B rs8099917 TT genotype (28); no comprehensive studies have been conducted in healthy controls.

Genome Wide Association Studies have mapped different phenotypes or characteristics for thousands of SNPs on all chromosomes (29). GWAS results have indicated that the IL28B rs12979860 (C>T) polymorphism plays an important role in the response for CHC to the treatment and spontaneous clearance of the virus (13,14,17,18,30). While spontaneous viral clearance occurs only in 15-50% of HCV-infected patients, chronic infection develops in the majority (31). In a study performed in African or European populations, HCV spontaneous clearance was three-fold higher in patients with the IL28B rs12979860 CC genotype compared to those with the CT and TT genotypes (14). In a study performed in Egypt, the incidence of the CC genotype (48%) was higher than that of the CT (38%) and TT (14%) genotypes in healthy control subjects (31). Furthermore, in these subjects, the C: T allele ratio was 67:33, indicating that the IL28B CC genotype protects against chronic infection progression by spontaneous clearance during the acute phase (32). In another study performed in Brazil, Conde et al. (33) found no differences between CHC patients and the control group for genotype and allele proportions of IL28B rs12979860 (C>T). They found a tendency (p=0.0813) towards a higher frequency of allele T in the group of patients according to the controls, but no significant difference in the genotypic frequency (p=0.1865) was found between the two groups. In other studies, the CC genotype played a major role in viral clearance caused by treatment (34,35). In our study, 44.6% of the cases in the control group were CC, 46.7% of the cases were CT, and 8.7% were TT. In the patients with CHC, these percentages were 32.1%, 46.3% and 20.6%, respectively. The incidence of the TT genotype in the patient group was significantly higher than in the control group (p=0.033). Contrary to the findings of El-Awaday et al. (31), this indicates that the TT genotype contributes to chronic HCV infection by leading to viral persistence. Fabris et al. (35) reported that the TT genotype was seen more frequently in patients with end-stage liver disease than in those with mild chronic hepatitis. In another Egyptian study, there was a significant difference in the distribution of the IL28B rs12979860 (C>T) genotypes between 440 HCV genotype-4 infected patients and 220 healthy control people (36). In the CHC patients, the prevalence of the CC genotype was 30.7%, that of the CT genotype was 49.5%, and that of the TT genotype was 19.8%, while in the control group the prevalence was 43.6%, 42.3% and 14.1%, respectively. It has been emphasized that there is a significant difference between patient and healthy control groups in genotype prevalence. In this study, the frequency of the T allele was 44.5% in CHC patients and 35.2% in the control group. Furthermore, we found that the frequency of the C allele was significantly higher (68%) in the control group than in the CHC treatment group (56%), and the frequency of the T allele was lower (32%) in the control group than in the CHC treatment group (44%) (p=0.011). These results were similar to those reported in studies in Egypt and Italy (23,36). In another study conducted by Shaker et al. (37) in Egypt, the ratios of the CC genotype and C allele frequency in the control group were lower than in our study. In our CHC patients, the frequencies of the CC genotype and C allele were lower than those described previously in American and European HCV-infected patients (14,17,38,39). The frequency of CC genotype changed between 38% (17) and 45% (38) in HCV infected patients in Europe. Our findings indicate that the existence of the T allele contributes to viral persistence and CHC development, supporting results indicating that antiviral treatment response in CHC was associated with the IL28B polymorphism (13,14,17,38,40).

In a study performed on 871 Caucasian patients, the CC genotype was found 39%, the CT genotype 49%, and the TT genotype 12% of patients with HCV infection (41). In a Romanian study, these rates were 24.3%, 62.6% and 13.1%, respectively (19); these two studies gave results that are similar to those of our study in which the CT genotype was the most prevalent (46.3%). Moreover, when the characteristics of the patients with CHC were evaluated based on the IL28B genotype subgroups, the frequency of patients with viral load ≥400,000 IU/mL was highest in the CT group (p=0.021), while no differences were found among the groups in terms

of baseline mean HCV RNA levels. In addition, there were no significant differences in mean age, gender distribution, BMI, genotype distributions, ALT level and median liver necroinflammation among the IL28B subgroups. Montes-Cano et al. (39) reported that ,in 284 CHC patients, there was no significant difference between patients in the CC and CT+TT groups with regard to viral load or in gender distribution and infection age; however, the ALT level was higher in the CC group (p=0.006). Sporae et al. (19) determined that baseline viral load was significantly lower in the TT group according to other genotypes and the frequency of patients with viral load >600,000 IU/mL was the highest for the CC genotype, at 80.8%. In the same study, no significant difference was seen in terms of mean age, gender distribution, BMI and fibrosis score among the IL28B subgroups. The reason of different viral load frequencies in IL28B subgroups according to other studies can be explained with the presence of factors which is effect viral load such as viral genotype, age, race, gender and route of transmission of infection.

In our study, 90.4% of the patients had genotype 1b infection and the frequency of genotype 4 infection was low. The liver fibrosis distribution was not found to be significantly significant between groups. However, the fibrosis severity seems higher in patients with the CT genotype and the p value was close to 0.05. This finding may become statistically significant with an increase in sample size (p=0.058). There was no difference in necroinflammation among the groups (p=0.791). In the study by Falleti et al. (23) the frequency of genotype 1 infection was 28.1%, the frequency of genotype 2 was 38.2%, the frequency of genotype 3 was 42.3%, and the frequency of genotype 4-5 was 36.6%, for the CC group. In the same study, the necroinflammation score in the CC group was significantly higher than in the group with CT+TT genotype based on the Ishak scoring system. The number of patients with severe hepatitis was higher in group with TT genotype, while the number of patients with mild hepatitis was higher in the group with CC genotype. In the study by Montes-Cano et al. (39) in the group with the CC genotype, the rate of HCV infection was 39.1% for genotype 1 and 66.7% for the other genotypes. Based on liver biopsy, a fibrosis score of mild was seen in 46.5% and a score of moderate seen in 43.2% of patients with the CC genotypes. In the CC+CT group, a fibrosis score of mild was seen in 53.5% and a score of moderate was seen in 56.8% of patients. In our study, because of the small number of patients and the fact that most of these had mild liver disease (78.9%), there may be some misinterpretation of the results. In conclusion, in patients with chronic HCV genotype-1b and 4 infections, the IL28B rs12979860 (C>T) gene polymorphism TT genotype and T allele frequency was higher than in healthy control subjects. Our findings suggest that the TT genotype might be effective in the progression of HCV infection. These results can aid clinicians in using antiviral treatment decisions. Further comprehensive studies should examine the effects of the IL28B genotype subgroups on the prognosis of disease and the response to treatment.

Ethics Committee Approval: Ethics committee approval was received for this study from the Ethics Committee for Clinical Research (2013/188).

Informed Consent: Written informed consent was obtained from patients who participated in this study.

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