DIRECT ACTING ANTIVIRAL (DAA) THERAPY IN REAL WORLD
HCV COMPENSATED CIRRHOSIS

Raluca Cezara Popa1,2, Iolanda Valentina Popa1,2, Andreea Dorobat1,2, Otilia Gavrilescu1,2,
Mihaela Dranga1,2*, Cristina Cijevschi Prelipcean2, Catalina Mihai1,2
“Grigore T. Popa” University of Medicine and Pharmacy Iasi
Faculty of Medicine
1. Department of Medical Specialties (I)
“St. Spiridon” County Clinical Emergency Hospital,
2. Institute of Gastroenterology and Hepatology
*Corresponding author. E-mail: mihaela_dra@yahoo.com

PATIENT FIRST: DIRECT ACTING ANTIVIRAL (DAA) THERAPY IN REAL WORLD
HCV COMPENSATED CIRRHOSIS (Abstract): Aim: Given the effectiveness of direct act-
ing antiviral (DAA) therapy in patients with chronic hepatitis C virus (HCV) infection, we
aimed to describe the demographic, clinical and biological characteristics of our patients
who received treatment with direct acting antivirals during December 2015-October 2016
and September 2017-January 2018. Material and Methods: We performed a prospective
study of 186 patients with chronic hepatitis C who have undergone treatment with
paritaprevir/ombitasvir/ritonavir and dasabuvir with or without ribavirin during December
2015 - October 2016 (group 1) and September 2017 - January 2018 (group 2). The inclusion
and exclusion criteria were established according to the national protocol. Demographic,
clinical and biological data were recorded before starting treatment. All patients underwent
upper gastrointestinal endoscopy and abdominal ultrasound. The determination of HCV gen-
types and subtypes was realized by the Abbott RealTime HCV assay. The presence of mild
or advanced fibrosis, cirrhosis, the viral necro inflammatory activity and the presence of
nonalcoholic steatohepatitis (NASH) were assessed by FibroMax non-invasive liver tests:
ActiTest, FibroTest and NashTest. Results: The mean age was 58.7 years for the first group
and 59.6 for the second group, with the predominance of female gender in both cases. Geno-
type 1b was present exclusively in the first group. Median HCV RNA at baseline was 1 264
123.180 IU/mL for the first group and 1 678 718.97 IU/mL for the second group. No
patients were HBV or HIV co-infected. Most patients were naive to interferon therapy. The comor-
bidity rate was increased in both groups: 28.82% vs. 49.3% had cardiovascular diseases,
23.41% vs. 14.66% had diabetes mellitus (with or without insulin requirement), 2.7% vs.
10.6% had thyroid dysfunction, 1.8% vs. 0% had dermatological disease, 0.9% vs. 5.33%
had psychiatric disorders, 1.8% vs. 1.33% had a history of cured neoplasm, 1.8% vs. 2.66%
had pulmonary diseases and 0.9% vs. 1.33% had hematological diseases. No patient had a
history of treated hepatocellular carcinoma. In both groups, the maximum number of drugs
administered simultaneously with the antiviral therapy was 4. The necro inflammatory activ-
ity assessed by ActiTest highlights a high rate for severe activity for both groups - 66.6%.
The prevalence of concomitant NASH assessed by NashTest (N2-severe inflammation) was
50% in group 1 and 53.3% in group 2. There were slight differences between the two groups
in terms of biological constants, but without any statistical significance. Conclusions: The
partial results of our study show a higher prevalence of HCV infection among people born
between 1946 and 1964 (baby boom generation), especially women. In this geographical area, genotype 1b was found exclusively. Most patients, naïve to interferon therapy, associate numerous comorbidities and simultaneous treatments that may decrease adherence to therapy due to the possibility of significant drug-drug interactions, requiring optimal therapeutic management and close monitoring. **Keywords**: DIRECT ACTING ANTIVIRALS, HEPATITIS C VIRUS INFECTION, COMORBIDITIES.

Hepatitis C virus (HCV), discovered in 1989, is an enveloped positive-sense single-stranded RNA virus recognized as the main causative agent of chronic liver disease, cirrhosis and hepatocellular carcinoma (1, 2). The global prevalence of HCV estimated by WHO is approximately 2.5% (185 million people), with a higher rate in Africa and Asia compared to North America, Europe and Australia (1,3,4). HCV related liver failure is the main indication of liver transplantation in the United States, Western Europe and Japan (2,5,6).

In the presence of humoral and cellular immune response directed against viral proteins, 70-100% of patients remain HCV-RNA positive secondary to acute infection due to the immune system's ability to circumvent the virus (7,8,9). Heterogeneity of HCV genome arises as a result of DNA-dependent RNA polymerase (NS5B) that causes viral replication to mis-incorporate ribonucleotides with a substitution rate of about one million more than the replacement rate for the human genome (10).

Most patients are asymptomatic or have nonspecific manifestations: fatigue, myalgia, arthralgia and weight loss. Evolution is slowly progressive to liver cirrhosis, hepatocarcinoma or death (9, 11).

Interferon (INF) has been the standard treatment for about 25 years. It was first approved in HCV patients in 1991, although its antiviral activity was first described in 1957. Under interferon monotherapy, the SVR rate increased from 5 to 20% and up to 40-80% with pegylated interferon plus ribavirin (12).

Knowing the replicative cycle of hepatitis C virus, the role of structural and non-structural proteins and their localization in the cell allowed the discovery of compounds with specific antiviral activity.

The main targets are the NS5A replication complex (daclatasvir, ledipasvir, ombitasvir, elbasvir, velpatasvir), the NS5B polymerase (sofosbuvir, dasabuvir) and the NS3/4A protease (simeprevir, paritaprevir, asunaprevir, grazoprevir) (10,13).

The main goal of antiviral therapy is to obtain suppression of the virus replication 12 weeks after the end of antiviral therapy. Clinical trials conducted so far describe high tolerability and safety profile with RVS being achieved in > 95% of patients. The therapeutic failure may occur secondary to the selection of resistant mutants that cause the recurrence of viral replication (14-16).

There are currently several regimens available with an equivalent safety profile: sofosbuvir/ribavirin, sofosbuvir/ simeprevir +/- ribavirin, sofosbuvir/daclatasvir +/- ribavirin, sofosbuvir/ledipasvir +/- ribavirin, ombitasvir/paritaprevir/ritonavir/dasabuvir +/- ribavirin, sofosbuvir/velpatasvir +/- ribavirin and elbasvir/grazoprevir +/- ribavirin (17).

The regimens available in Romania are represented by ledipasvir/sofosbuvir and paritaprevir/ombitasvir/ritonavir/dasabuvir. Currently, limited availability and in-
creased costs of direct-acting antivirals require prioritization of therapy in patients with advanced liver disease.

Given the effectiveness of direct oral antiviral therapy in patients with chronic HCV infection, we aimed to describe the demographic, clinical and biological characteristics of our patients with chronic HCV infection who received treatment with paritaprevir/ombitasvir/ritonavir/dasabuvir with or without ribavirin during December 2015-October 2016 and September 2017-January 2018.

MATERIAL AND METHODS

We performed a single-center prospective cohort study of 186 patients with chronic hepatitis C who have undergone treatment with paritaprevir/ombitasvir/ritonavir and dasabuvir with or without ribavirin during December 2015 - October 2016 (group 1) and September 2017 - January 2018 (group 2).

According to the national protocol, inclusion criteria during December 2015 - October 2016 were: genotype 1b interferon naive or experienced patients with compensated cirrhosis - F4 (Child Pugh score≤6), detectable HCV-RNA levels, absence of alcohol consumption or other intravenous/intranasal illicit substances in the last 3 months, absence of significant drug-drug interaction.

During September 2017 - January 2018, inclusion criteria were: interferon naive or experienced patients with advanced fibrosis (F3) or compensated cirrhosis - F4 (Child Pugh score≤6), detectable HCV-RNA levels, absence of alcohol consumption or other intravenous/intranasal illicit substances in the last 3 months, absence of significant drug-drug interaction; interferon naive or experienced patients with mild fibrosis - F2 with mixt cryoglobulinemia, chronic kidney disease, non-Hodgkin lymphoma, hemophilia and thalassemia major.

Patients with hepatocellular carcinoma (METAVIR score F2, F3 or F4) could be treated if they had a transplant indication or if relapse was not found (CT/MRI) more than 6 months after ablation or resection.

Patients with decompensated cirrhosis (Child Pugh score >6) and those with dysplastic nodules were excluded.

Demographic, clinical and biological data (gender, age, weight, height, body mass index, status after interferon therapy, comorbidities, concomitant medication, complete blood count, INR, total bilirubin, AST, ALT, GGT, glucose levels, creatinine, AFP) were recorded before starting treatment.

All patients underwent upper gastrointestinal endoscopy and abdominal ultrasound.

The determination of HCV genotypes and subtypes was realized by the Abbott Realtime HCV assay. The presence of mild or advanced fibrosis, cirrhosis, the viral necro inflammatory activity, and NASH was assessed by FibroMax non-invasive liver tests: ActiTest, FibroTest and NashTest.

To present our data, we used univariate descriptive statistic tools.

In the first group were included 111 patients with compensated liver cirrhosis (Child Pugh score ≤6). The diagnostic of cirrhosis was based on clinical, biological and imagistic (abdominal ultrasound) criteria. All patients had a METAVIR score according to FibroTest F3 (26.12%), F3-F4(2.7%) or F4(70.27%). In the second group were included 75 patients with advanced fibrosis or compensated cirrhosis (score Child Pugh <=6) with a METAVIR score according to FibroTest F3(41.3%), F3-F4(6.6%) or F4(52%) (tab. I).
**TABLE I.**
Demographic, clinical and biological characteristics
(1 month prior to DAA therapy)

<table>
<thead>
<tr>
<th></th>
<th>Group 1</th>
<th>Group 2</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean (SD) / Number (%)</td>
<td>Mean (SD) / Number (%)</td>
<td></td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td>111</td>
<td>75</td>
<td></td>
</tr>
<tr>
<td><strong>age</strong></td>
<td>58.7 (8.35)</td>
<td>59.6 (9.6)</td>
<td>p=0.03</td>
</tr>
<tr>
<td>female gender</td>
<td>61 (54.9%)</td>
<td>46 (60.52 %)</td>
<td>p=0.01</td>
</tr>
<tr>
<td>BMI</td>
<td>27.45 kg/m² (3.75)</td>
<td>27.38 kg/m² (4.37)</td>
<td>p=0.3</td>
</tr>
<tr>
<td><strong>genotype</strong></td>
<td>1b (100%)</td>
<td>not required</td>
<td></td>
</tr>
<tr>
<td><strong>HCV-RNA</strong></td>
<td>1264123.180 (1835368.49)</td>
<td>1678718.97 (299474686)</td>
<td>p=0.004</td>
</tr>
<tr>
<td><strong>Fibromax (METAVIR score)</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F4</td>
<td>78 (70.27%)</td>
<td>39 (52%)</td>
<td></td>
</tr>
<tr>
<td>F4/F3</td>
<td>3 (2.7%)</td>
<td>5 (6.6%)</td>
<td></td>
</tr>
<tr>
<td>F3</td>
<td>29 (26.12%)</td>
<td>31 (41.3%)</td>
<td></td>
</tr>
<tr>
<td>F2</td>
<td>-</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td><strong>hemoglobin</strong></td>
<td>14.3 g/dl (1.76)</td>
<td>14.37 g/dl (1.44)</td>
<td>p=0.1</td>
</tr>
<tr>
<td><strong>platelets</strong></td>
<td>154126.12/mm³ (66031.13)</td>
<td>173373.33/mm³ (77401.85)</td>
<td>p=0.004</td>
</tr>
<tr>
<td><strong>INR</strong></td>
<td>1.26 (1.29)</td>
<td>1.1 (0.13)</td>
<td>p=0.1</td>
</tr>
<tr>
<td><strong>ALT</strong></td>
<td>106.96 U/L (73.59)</td>
<td>99.64 U/L (58.37)</td>
<td>p=0.7</td>
</tr>
<tr>
<td><strong>AST</strong></td>
<td>96.3 U/L (62.64)</td>
<td>82.79 U/L (41.4)</td>
<td>p=0.5</td>
</tr>
<tr>
<td><strong>GGT</strong></td>
<td>89.32 U/L (68.48)</td>
<td>90.76 U/L (75.68)</td>
<td>p=0.3</td>
</tr>
<tr>
<td><strong>BT</strong></td>
<td>1.05 mg/dl (0.45)</td>
<td>0.9 mg/dl (0.4)</td>
<td>p=0.3</td>
</tr>
<tr>
<td><strong>glucose</strong></td>
<td>114.68 mg/dl (35.09)</td>
<td>120.46 mg/dl (69.58)</td>
<td>p=0.03</td>
</tr>
<tr>
<td><strong>creatinine</strong></td>
<td>1.17 mg/dl (4)</td>
<td>0.76 mg/dl (0.12)</td>
<td>p=0.06</td>
</tr>
<tr>
<td><strong>albumin</strong></td>
<td>4.45 g/dl (2.68)</td>
<td>4.15 g/dl (0.39)</td>
<td>p=0.01</td>
</tr>
<tr>
<td><strong>AFP</strong></td>
<td>15.5 ng/ml (20.28)</td>
<td>21.03 ng/ml (34.69)</td>
<td>p=0.08</td>
</tr>
<tr>
<td><strong>Comorbidities</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>➢ cardiovascular diseases</td>
<td>32 (28.82 %)</td>
<td>37 (49.3 %)</td>
<td></td>
</tr>
<tr>
<td>➢ diabetes + insulin</td>
<td>18 (16.21%)</td>
<td>7 (9.33 %)</td>
<td></td>
</tr>
<tr>
<td>➢ diabetes - insulin</td>
<td>8 (7.2 %)</td>
<td>5 (5.33 %)</td>
<td></td>
</tr>
<tr>
<td>➢ psychiatric disorders</td>
<td>1 (0.9%)</td>
<td>4 (5.33%)</td>
<td></td>
</tr>
<tr>
<td>➢ neurologic diseases</td>
<td>-</td>
<td>1 (1.33%)</td>
<td></td>
</tr>
<tr>
<td>➢ thyroid dysfunction</td>
<td>3 (2.7%)</td>
<td>8 (10.6 %)</td>
<td></td>
</tr>
<tr>
<td>➢ dermatologic diseases</td>
<td>2 (1.8%)</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>➢ hematological diseases</td>
<td>1 (0.9%)</td>
<td>1 (1.33 %)</td>
<td></td>
</tr>
<tr>
<td>➢ asymptomatic cryoglobulinemia</td>
<td>-</td>
<td>3 (4 %)</td>
<td></td>
</tr>
<tr>
<td>➢ pulmonary diseases</td>
<td>2 (1.8 %)</td>
<td>2 (2.66%)</td>
<td></td>
</tr>
<tr>
<td>➢ rheumatologic diseases</td>
<td>1 (0.9%)</td>
<td>1 (1.33%)</td>
<td></td>
</tr>
<tr>
<td>➢ cured cancer</td>
<td>2 (1.8%)</td>
<td>1 (1.33%)</td>
<td></td>
</tr>
<tr>
<td><strong>Upper gastrointestinal endoscopy</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>esophageal varices</td>
<td>20 (18.01 %)</td>
<td>12 (16 %)</td>
<td></td>
</tr>
</tbody>
</table>
The mean age was 58.7 (SD=8.35) years for the first group and 59.6 (SD=9.6) years for the second group, with the predominance of female gender in both cases. Genotype 1b was present exclusively in the first group. For patients who received antiviral treatment between September 2017 to January 2018, HCV genotyping was not performed because it was not cost-effective.

RESULTS
Median HCV RNA at baseline was 1,264,123.180 IU/mL (SD=1,835,368.49) for the first group and 1,678,718.97 IU/mL (SD=2,994,746.86) for the second group. No patients were HBV or HIV co-infected. Most patients were naive to interferon therapy. The comorbidity rate was increased in both groups: 28.82% vs. 49.3% had cardiovascular diseases (hypertension, arrhythmia, valvular diseases), 23.41% vs. 14.66% had diabetes mellitus (with or without insulin requirement), 2.7% vs. 10.6% had thyroid dysfunction, more commonly autoimmune thyroiditis, 1.8% vs. 0% had dermatological disease (vitiligo, lichen planus), 0.9% vs. 5.33% had psychiatric disorders (depression, anxiety, cognitive impairment), 1.8% vs. 1.33% had a history of cured neoplasm (colon, kidney, urinary bladder), 1.8% vs. 2.66 had pulmonary diseases (asthma, bronchiectasis, pulmonary fibrosis) and 0.9% vs. 1.33% had hematological diseases (systemic lupus erythematosus, myeloproliferative syndrome). No patient had a history of treated hepatocellular carcinoma. In both groups, the maximum number of drugs administered simultaneously with the antiviral therapy was 4. The necro inflammatory activity assessed by ActiTest highlights a high rate for severe activity for both groups - 66.6%.

The prevalence of concomitant NASH assessed by NashTest (N2-severe inflammation) was 50% in group 1 and 53.3% in group 2. There were slight differences between the two groups in terms of biological constants, but without any statistical significance.

DISCUSSION
The prevalence of HCV infection in adult patients (over 15 years old) in Romania was reported at 3.2%, equivalent to 595 000 anti-HCV positive individuals with a viremic prevalence rate of 91.3% (19). Previous data suggest that the high-risk group for HCV infection in Romania is the Baby Boom generation (people born between 1946 and 1964) (20). The age distribution of our patients met the same characteristics.

The analysis of the nucleotide sequences of the HCV RNA single-stranded molecule allowed the identification of 7 major genotypes and 67 subtypes with different and complex geographic distribution (21, 22). The distribution of HCV “epidemic subtypes” is probably influenced by historical and contemporary immigration patterns.

The global prevalence of genotype 1 is forecasted at 42.2% (83.4 million individuals), with a higher rate in East Asia (more than ⅓ of cases). According to the Epidemiological National Multicenter Study (ENMS) for genotyping of HCV in Romania conducted by Grigorescu (20) between October 2015 and March 2006, genotype 1, subtype b is predominant (97.72%) in Romanian patients with chronic HCV infection. More recent epidemiologic studies show a slightly lower prevalence (92.6%) (19). Genotypes 1a (5.4%), 3 (0.8%) and 4 (1.2%) were recently introduced because of
injecting drug use (19,23). In our study sample, even not fully representative for Romanian population, HCV genotype 1b was exclusively present for those tested between December 2015- October 2016. According to the expert committee of the National House of Health Insurance, during September 2017- January 2018, genotyping was not mandatory due to lack of cost-effectiveness.

Given the relatively silent nature of HCV infection, the degree of addressability of undiagnosed patients is lower compared to those with other infections with more prominent clinical signs and symptoms. Even if the optimal timing for the initiation of antiviral therapy is unknown, lack of funds in our country has led to a gradually upscale of HCV treatment.

At present, in our country, the access to antiviral therapy with paritaprevir/ombitasvir/ritonavir/dasabuvir is limited to patients with advanced fibrosis (METAVIR score F3) or compensated cirrhosis (METAVIR score F4 and Child Pugh score <=6) who are either treatment-naive or interferon-experienced, with or without coinfecion with HBV or HIV and to patients with mild fibrosis (METAVIR score F2) interferon-naive or interferon-experienced with mixt cryoglobulinemia, chronic kidney disease, non-Hodgkin lymphoma, hemophilia and thalassemia major. Patients with hepatocellular carcinoma (METAVIR score F2, F3 or F4) may be treated if they have a transplant indication or if relapse is not found (CT/MRI) more than 6 months after ablation or resection.

According to the “Summary of Product Characteristics as authorized by European Medical Agency”, administration of paritaprevir/ombitasvir/ritonavir/dasabuvir is not indicated in patients with decompensated liver cirrhosis (Child Pugh score B or C) (17).

Currently in Romania, the only treatment available for patients with decompensated liver cirrhosis (only Child Pugh score B) is represented by a single-tablet fixed-dose combination of sofosbuvir and ledipasvir with or without ribavirin.

In our study, although in group 2 were included patients with a lower degree of fibrosis, we didn’t find statistically significant differences compared to group 1 regarding the severity of hepatic disease (baseline biological constants, comorbidities, esophageal varices, necro-inflammatory activity and concomitant presence of nonalcoholic steatohepatitis). Patients with less advanced fibrosis should be included in the antiviral treatment to improve outcomes, prevent disease progression and reduce the incidence of hepatocellular carcinoma.

In august 2017, after gaining increased efficacy in phase 3 trials ENDURANCE and EXPEDITION (99% SVR12 rate for genotypes 1,2,4,6 and 91-96% SVR12 rate for genotype 3), the U.S. Food and Drug Administration (FDA) and the European Medicines Agency (EMA) have approved the use of pan genotypic combination of direct acting antiviral agents glecaprevir (NS3/4A protease inhibitor)/pibrentasvir (NS5A inhibitor) to treat patients with compensated liver disease, with or without cirrhosis (24). This fixed dose oral combination (300 mg/120 mg) has the possibility to reduce by 4 weeks the time for cure in non-cirrhotic patients with genotype 1-2, 4-6, even in those with severe kidney disease or HIV coinfection (25).

The real-world prioritization is to treat all patients with advanced disease.

Prioritization of antiviral therapy should
Direct acting antiviral (DAA) therapy in real world HCV compensated cirrhosis

also be done in those who associate extra-hepatic comorbidities. In our cohort, the most prevalent comorbidities were cardiovascular diseases followed by diabetes, thyroid dysfunctions and psychiatric disorders. In these patients, there is an increased risk of significant drug-drug interactions, especially with concomitant administration of proton pump inhibitors, thyroid hormones and dihydropyridine derivates. This risk is higher in elderly patients (> 65 years), in patients with advanced liver cirrhosis and with a combination of at least two direct-acting antivirals (26). In patients undergoing treatment with paritaprevir/ombitasvir/ritonavir/dasabuvir and concomitant ambulatory medication, the risk of significant drug-drug interaction is estimated at 66.3% (26).

All patients with chronic HCV infection regardless of degree of fibrosis should receive antiviral therapy. Early initiation of DAA may improve the outcome and lower the costs.

Cost-effectiveness analysis are needed to identify different ways to redirect resources to minimize the inequality of distribution of therapy with DAA and to achieve the WHO’s objective to eradicate HCV by 2030.

**CONCLUSIONS**

The partial results of our study show a higher prevalence of HCV infection among people born between 1946 and 1964 (baby boom generation), especially women. In this geographical area, genotype 1b was found exclusively. Most patients, naïve to interferon therapy, associate numerous comorbidities and simultaneous treatments that may decrease adherence to therapy due to the possibility of significant drug-drug interactions, requiring optimal therapeutic management and close monitoring.

**REFERENCES**