HEPATITIS-C GENOTYPE-3A INFECTION;
ROLE OF UNFAVORABLE (IL28B- RS12979860 NON-CC) GENOTYPING IN RESPONSE TO SOFOSBUVIR (SOF) AND RIBAVIRIN IN PATIENTS IN POPULATION OF KHYBER PUKHTOONKHWA (KPK).

Dr. Nizamuddin¹, Abdul Hameed Khan², Ayesha Jamil³, Fazal Rahim⁴, Muhammad Riaz⁵

ABSTRACT… Objectives: In last decade, “treatment of chronic hepatitis-C revolved from interferon based therapy to most effective interferon free therapy with new direct antiviral drugs like Sofosbuvir and ribavirin” which is recommended for all genotypes of HCV infection. Treatment response in Chronic Hepatitis-C is affected both by viral and human factors. We conducted this study “to evaluate the effect of human factor like (IL28B-rs12979860 non-CC) genotyping in response to Sofosbuvir based dual therapy in Hepatitis-C Genotype-3a infection” in population of Khyber Pukhtoonkhwa (KPK). Setting: This open labeled, multi-center study was conducted in Peshawar-Khyber Pukhtoonkhwa (KPK). Period: March-2016 to August-2016.

Method: Total of 70 patients were enrolled. After doing “PCR for HCV-RNA-Viral level, Viral-Genotyping and Human genotyping for IL-28B, patients were put on Sofosbuvir and ribavirin for 24-weeks”. Patients were assigned into two groups (1:1), “having 35 in each, including group-A as those having favorable CC (IL28B- rs12979860-CC) genotyping and group-B as those having unfavorable non-CC (IL28B-rs12979860-non-CC) genotyping”. The primary end point was “Sustained Virological response12 (SVR12), which is HCV-RNA level<40IU/ml at 12-weeks after completion of therapy in these two groups”. Results: Among 70-patients, male-female ratio was 57.15% (n=40) and 42.85% (n=30) respectively. Each group has 35-cases. Rate of SVR12 was 88.57% (n=31/35) in group-A, 91.42% (n=32/35) in group-B, having P-value<05.

Conclusion: This study confirm that “unlike interferon, unfavorable non-CC (IL28B-rs12979860-non-CC) genotyping have no or minimal role in treatment response to Sofosbuvir in Hepatitis-C genotype-3a infections”.

Key words: Chronic hepatitis-C, Dual therapy, Sofosbuvir, Unfavorable non-CC (IL28B-rs12979860-non-CC) genotyping.

INTRODUCTION
Hepatitis-C (CHC) is a one of the biggest global chronic disease, affecting almost every geographical corner of the world. The global prevalence of “Hepatitis-C is 2.4%, affecting >200 million people worldwide”. About half of these patients ultimately end with “cirrhosis and Hepatoma without treatment”.¹² Numbers of cases are decreasing in “developed world due to implementation of extensive screening measures before blood transfusion, surgical interventions, organs transplantation and over all allocation of health resources”. But on the other hand, developing countries still face heavy problems due to “low literacy level, bad health care services, low budget and unregistered practices”. That is why new cases are still coming into the old pool in heavy numbers. Although new cases are increasing, but at the same time “new treatment arms are coming into practice in the therapeutic armamentarium against Hepatitis-C”.³ This history starts from “conventional interferon, then peg-interferon and now to highly effective, Direct Acting Antiviral (DAAs), drugs like sofosbuvir and Daclatasvir etc”. American association for the Study of Liver Disease (AASLD)⁴ and European Association for the Study of Liver (EASL)⁵ recommend “Sofosbuvir and ribavirin for 24 weeks in the treatment of HCV genotype 3 infections in all patients with or without cirrhosis”.

1. Hayatabad Medical Complex (HMC) Peshawar Pakistan.
2. Khyber Girl Medical College Peshawar Pakistan.
3. Khushal Medical Center Peshawar Pakistan.

Correspondence Address:
Dr. Nizamuddin
dnizam99@yahoo.com

Article received on: 17/10/2016
Accepted for publication: 15/03/2017
Received after proof reading: 06/05/2017

Article Citation: Nizamuddin, Khan AH, Jamil A, Rahim F, Riaz M. Hepatitis-C genotype-3a infection; role of unfavorable (IL28B- RS12979860 NON-CC) genotyping in response to sofosbuvir (SOF) and ribavirin in patients in population of Khyber Pukhtoonkhwa (KPK). Professional Med J 2017;24(5):670-674.
Along with viral factors and genotyping, “response to treatment in all these modalities is also affected by some human factors too”. It does include sex, age and genetic factors like “IL-28B-CC or non-CC genotyping, officially called as (Interleukin-28B, “interferon, lambda3”). It is now clear, “that Hepatitis-B and Hepatitis-C is responsible for >75% of liver disease, including Cirrhosis and Hepatoma” and have substantial impact on morbidity, mortality and utilization of health budget. So it wisely needs aggressive treatment and addressing of all factors that can influence the success of treatment. It is documented and established in research, that “unfavorable non-CC (IL28B-rs12979860-non-CC) genotyping in human due to single nucleotide polymorphism (SNIPs) have bad impacts in treatment success for Chronic Hepatitis-C (CHC), treated with Pegylated-Interferon plus weight based ribavirin”. But unlike Peg-Interferon, “the role of unfavorable non-CC (IL28B-rs12979860-non-CC) genotyping in treatment success with nucleotide analogue NS5B-HCV-RNA dependent RNA-polymerase inhibitor, Sofosbuvir, which is a new DAAs, is still debatable”. Most of these studies are conducted internationally to evaluate the role of “unfavorable non-CC (IL28B-rs12979860-non-CC) genotyping in Genotype-1 infection.7,8” Our community has “different genetic parameters and is mainly affected by Genotype-3a, which has different response to different treatment regimen, therefore it should be evaluated in our community”. Moreover, this study will also fill the deficiency of available data and literature, “which is not sufficient to address this association in our community”. Furthermore, unlike other factors like “viral genotyping, cirrhosis, age, gender, baseline viral load, the importance of unfavorable non-CC (IL28B-rs12979860-non-CC) genotyping will get clear as having no significant effects on antiviral therapy”.

MATERIALS AND METHODS
The study was mainly conducted on patients, who visited different public and private tertiary care hospitals and consultant clinics in Peshawar District of Pakistan. The assumed duration of study was 6-months, starting from March 2016 to August 2016. Total 70-patients were selected, “having chronic Hepatitis C genotype 3a infections”. All selected patients were distributed into two groups (1:1), 35 in each including “group-A as those having favorable CC (IL28B-rs12979860-CC) genotyping and group-B as those having unfavorable non-CC (IL28B-rs12979860-non-CC) genotyping”. All patients with cirrhosis, Chronic Liver disease due to other causes and Concomitant Hepatitis-B or HIV infections and patients infected by other genotypes of Hepatitis-C were excluded from the study. To ensure good compliance and tolerability, patient with “advanced renal, cardiac diseases and cognitive dysfunctions were excluded from the study”. All these patients were put on Sofosbuvir plus ribavirin for 6months. After the end of 24weeks treatment, HCV-RNA-PCR was done and was labeled as SVR12. The open labeled, prospective design was used in the study.

Data Collection
After ethical approval for the study from ethical committee of Hayat Abad Medical Complex Peshawar, “informed consent was obtained from the patients prior to their enrollment for the same study and treatment strategy”. The demographic information of the subjects such as names, age, gender and data regarding HCV-G3a and IL-28B were recorded according to the predefined inclusion/exclusion criteria. “Viral RNA was extracted and reverse transcribed to cDNA using Viral RNA extraction and cDNA synthesis kit (Qiagen, USA), respectively.” HCV-RNA-PCR, genotyping and SVR12 were done with Qiagen-kit, using Rotorgene-6000 Molecular System, having Lower limit of quantification (LLOQ)<40IU/ml. IL-28B genotyping was done using genesig snpsig kit with special primer at the start of treatment.

Data Analysis
All data was entered in Microsoft Office Excel 2007, tabulated and analyzed by using SPSS statistical program. The data was expressed as mean percentage and presented in tabulated form.

RESULTS
Out of total 70-studied patients, 57.15% (n=40)
were male and 42.85% (n=30) were female, with mean age of 37±1.26 years. Age distribution among 70-patients was analyzed as n=7 (10%) patients were in age group of 21-30 years, n=14 (20%) patients were in age group of 31-40 years, n=28 (40%) patients were in age group of 41-50 years, n=14 (20%) patients were in age group of 51-60 years and n=7 (10%) patients were above 60 years of age” as shown in Table-I.

In all 35-patients in “group-A, n=21(60%) were male and n=14(40%) were female, while in group-B, n=19(54.28%) were male and n=16(45.72%) were female” as shown in Table-I (a).

The status of “SVR12 among 70 patients was analyzed in both groups. Rate of SVR12 was 88.57% (n=31/35) in group-A, 91.42% (n=32/35) in group-B” as shown in Table-II.

Status of “SVR12 in male and female was analyzed in both groups as 87.5% (n=35/40) male and 93.33% (n=28/30) female have achieved SVR12, as shown in Table-III”. The overall response in both groups is also shown, which is 90%.

Status of male and female, who have not responded in group-A with favorable (IL28B-rs12979860-CC) genotyping was ¾ and ¼ respectively, in group-B with unfavorable (IL28B-rs12979860-non-CC) genotyping was 2/3 and 1/3, respectively.

**DISCUSSION**

In Chronic Hepatitis-C, aggressive approach is needed while treating every patients suffering from this crippling disease. In last decade, “the paradigm shift of treatment from interferon based therapy to direct acting anti-viral therapy has totally changed the direction of research around the globe”. At the same time it has also changed the approach to look for all factors affecting the treatment response.9,10 Our population is mainly affected by “HCV-G3 infection, especially G3a, which is notoriously popular for causing cirrhosis”. The new drug, like sofosbuvir in the therapeutic armamentarium against HCV which is recommended by “European Association for the Study of Liver (EASL) and American Association for the Study of Liver Diseases (AASLD)” has given a new hope both to the clinician and patients to be used safely in all cirrhotic and non-cirrhotic patients.

Unlike interferon, “the human interleukin and other inflammatory mediators have minimal or no role in treatment response to this drug, as it is not affecting the human immune system”. In our study, the response rate in both group of patients with “favorable CC (IL28B-rs12979860-CC) genotyping and unfavorable non-CC (IL28B-rs12979860-non-CC) genotyping are almost same with a little better edge in group-B with unfavorable non-CC (IL28B-rs12979860-non-CC) genotyping”. The overall response rate in this study are close to the finding of “VALENCE
clinical trials by Gilead Sciences, which show 84% (210/250) SVR12, in all cases with HCV-genotype3 infection, treated with Sofosbuvir and ribavirin for 24 weeks. In another clinical trial at the name of NEUTRINO by Gilead Sciences, “conducted on other genotypes of HCV, it has been confirmed that SVR12 rate is similarly high in both subject of IL28B genotyping, which is 94/95(99%) in IL28B-CC allele and 202/232(87%) with IL28B non-CC allele”. In another study by J.A. Holmes et al., while evaluating the role of “IL-28B genotyping in response to different DAAs, it has been confirmed that IL-28B genotyping have no impacts on response to DAAs”. They also found that, “even the impacts of IL-28B genotyping on Peg-interferon can be reduced by adding one or two DAAs to triple or quadruple therapy along with Peg-interferon in the treatment of chronic Hepatitis-C.”

Some other shared findings include “role of IL28B non- rs12979860-CC genotyping in new and treated cases, male sex, stage of cirrhosis and general built of the patient in treatment response to Sofosbuvir based regimen need serious consideration”. However single genotype, straightforward new cases and small sample size are the main limitations of the present study.

Sofosbuvir is not only “effective but also safe, as some of the most common adverse effect observed in this study were, aches and pains, flue like symptoms and insomnia, but all were mild in nature and none of the patient discontinue the treatment”. And finally, it is now suggested and even accepted worldwide with many trials, “that all patients with HCV infection, who are candidates to be treated with Sofosbuvir based regimen should not be investigated unnecessarily for unfavorable non-CC (IL28B-rs12979860-non-CC) genotyping, as it has no role on treatment response with sofosbuvir in any patients with any Viral genotyping”.

However, large trial is needed to address role of age, sex, initial viral load, status of the liver (i.e. Cirrhosis) and HCV sub-genotypes in response to Sofosbuvir in Pakistani population.

CONCLUSION

“Sofosbuvir and Ribavirin based dual therapy is currently the most popular and effective treatment in all cases with Chronic Hepatitis-C genotype3a infections, and unfavorable non-CC (IL28B- rs12979860-non-CC) genotyping have no effect on treatment response’ to this combination in Pakistani population. Further confirmation is suggested, “Both at national and international level on huge number of patients with HCV-genotype-3a infection”.

REFERENCES
5. AASLD/IDSA. Recommendations for testing, management, and treating hepatitis C. Retreatment of persons in whom prior therapy has failed. [AASLD/IDSA Hepatitis C Guidance] -2016.


“Power does not corrupt. Fear corrupts... Perhaps The Fear of a Loss of power.”

Unknown

<table>
<thead>
<tr>
<th>Sr. #</th>
<th>Author-s Full Name</th>
<th>Contribution to the paper</th>
<th>Author=s Signature</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Dr. Nizamuddin</td>
<td>Main (primary) Author</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>Abdul Hameed Khan</td>
<td>Supervisor</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>Ayesha Jamil</td>
<td>Co-author / Data collection</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>Fazal Rahim</td>
<td>Supporting author</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>Muhammad Riaz</td>
<td>Supporting author / Data collection</td>
<td></td>
</tr>
</tbody>
</table>