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EVALUATION OF THE SAFETY AND RESISTANCE ASSOCIATED VARIANTS OF SOFOSBUVIR/DACLATASVIR AMONG EGYPTIAN PATIENTS WITH HEPATITIS C VIRUS: A PROSPECTIVE STUDY

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Background*: Hepatitis C virus (HCV) is a major health problem. Current treatment by direct-acting antivirals achieved high sustained virological response (SVR). However, drug intolerance or relapse may occur. We aimed to demonstrate the safety of sofosbuvir (SOF) plus daclatasvir (DCV) regimen in Egyptian patients with hepatitis C infection and the assessment of resistance associated variants (RAVs) in non-responders.* ***Methods:*** *In this prospective study, 850 HCV patients eligible to SOF + DCV ± ribavirin (RBV) were recruited. They were divided into two groups; patients with chronic hepatitis C (CHC) and patients with liver cirrhosis. Baseline data included clinical history, examination, routine laboratory tests and HCV viral load. Safety evaluation was assessed during treatment up to 12 weeks after the end of treatment. RAVs assessment was considered at baseline and in cases of relapse.* ***Results****: CHC group included 548 patients while 302 had liver cirrhosis. The most frequent adverse events were headache 20%, fatigue 14%, myalgia 5.2%. Diarrhea occurred in 4.6% with significantly higher frequency among liver cirrhosis group; 7.3% vs. 3.1% (P= 0.04). No patients had to stop treatment because of adverse events. SVR was achieved in 91.2% while 75 (8.8%) had relapse. At baseline, RAVs were found in 10%. After therapy, RAVs (E237D) were detected in 1 non-responder.* ***Conclusion:*** *Treatment with SOF/DCV was effective and well tolerated in patients with HCV. RAVs testing is not routinely recommended before treatment as resistant variants could occur naturally in HCV.*

Keywords*: HCV; DAAs; sofosbuvir; daclatasvir; RAVs.*

INTRODUCTION

Hepatitis C virus (HCV) infection attained growing international concern due to its substantial effect on morbidity and mortality[1]. HCV is a leading cause of cirrhosis, hepatocellular carcinoma (HCC) and liver-related death worldwide. The HCV disease burden continues to increase as the infected person advances to late stage liver disease[2]**.**

Sofosbuvir (SOF) is a pangenotypic nucleotide analog inhibitor of HCV NS5B viral polymerase. It was approved in 2013 by FDA for treatment of chronic hepatitis C infection genotypes 1, 2, 3 and 4 as part of combination regimens of direct acting antiviral therapy (DAAs)[3]. It was also recommended for treatment of hepatocellular carcinoma meeting Milan criteria awaiting liver transplantation to prevent HCV recurrence and was recommended for treatment of HCV/HIV-1 coinfection[3].

Daclatasvir (DCV) inhibits both viral RNA replication and virion assembly by

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binding to the N-terminus of NS5A causing structural distortions that interfere with NS5A functions. Daclatasvir is indicated for use with sofosbuvir for the treatment of treatment-naive or interferon (IFN)-experienced patients with chronic HCV genotype (G) 1, 2 or 3 infections, for treatment-naive HCV G2 or 3 infected patients with compensated cirrhosis +/- addition of weight-based ribavirin (RBV) and for all HCV genotypes with decompensated liver cirrhosis or post-liver transplantation recurrent infection with initial low dose of RBV[4]. In Egypt, the national Ministry of Health Protocol recommends the standard use of combination therapy by SOF+DCV in chronic HCV and the addition of RBV to this regimen depends on the presence of liver cirrhosis.

HCV resistant associated variants (RAVs) are seen in most patients who do not achieve SVR. These resistance-associated mutations depend on the class of DAAs used and vary between hepatitis C virus genotypes and subtypes[4].

NS5A RAVs can be very common, with Y93H detected in up to 15% of the population and L31M in up to 6.3%. Other RAVs tend to also be commonly detected in approximately 0.3%–3.5% of the population[5].

This study aimed to evaluate the safety and efficacy of the sofosbuvir/ daclatasvir treatment of chronic HCV and to assess the occurrence, the type and the prevalence of RAVs in patients with treatment failure or relapse.

PATIENTS AND METHODS

Ethical statement and informed consent.

The study was conducted according to the guidelines of the Declaration of Helsinki, and approved by the Institutional Review Board of Faculty of Medicine, Assiut University, Assiut, Egypt (IRB# 17200170). Informed consent was obtained from all subjects involved in the study.

Study design

This prospective study evaluated the effect of 12 week of oral sofosbuvir 400 mg plus daclatasvir 60 mg with or without ribavirin 1000–1200 mg. The included patients fulfilled the inclusion criteria to receive DAAs and treated in AL Rajhy hospital, Assiut, Egypt. Treatment eligibility was determined according to EASL guidelines 2016[6]. The addition of ribavirin to treatment was kept for difficult to treat patients (cirrhotic patients and/or IFN-treatment experienced).

Patients

Patients with chronic HCV infection with detected serum HCV RNA were included. Both treatment naive and IFN-experienced patients were included. Patients were divided into two groups; the first group included chronic hepatitis C (CHC) and the second group included compensated liver cirrhosis; Child A & early B. Patients were excluded if they had decompensated cirrhosis (late Child B or C) or with history of decompensation, HIV or hepatitis B virus infection, chronic liver disease of non-HCV etiology), platelets < 50 x 103 /L, bilirubin >2 mg/dl , alanine and aspartate aminotransferase (ALT and AST) > 10 times ULN or HCC.

At baseline, assessment was done by medical history and examination with evaluation of previous interferon therapy and the type of received DAAs regimen. Laboratory testing including: blood picture, liver and kidney functions, INR, random blood glucose and HCV viral load by PCR. Assessment of FIB-4 & APRI scores and baseline ultrasound was done. Follow up 12 weeks after treatment was done by the same laboratory tests.

Efficacy assessment

Serum HCV RNA was measured before start of treatment and 12 weeks after treatment for all included patients. Primary efficacy endpoint was SVR12 defined as HCV RNA below the lower limit of quantification or undetectable at least 12 weeks after the end of treatment.

Safety assessments

Safety assessment was done by collecting data during treatment up to the end of follow-up period (12 weeks after the last dose) by assessment of physical examination, vital sign measurements, clinical, laboratory tests, and documentation of any adverse effects. Adverse events were considered serious if resulted in death, life-threatening complication or required patient hospitalization.

RAVs assessment

The baseline HCV RNA levels were assessed before the initiation of HCV antiviral therapy and the results were available for the diagnosis and management of HCV infection in patients 'data. RAVs assessment was considered at baseline and in cases of relapse.

HCV RNA extraction, quantification and genotyping

HCV RNA was extracted from 650 µl for CAP/CTM HCV v2.0 by means of the Cobas Ampliprep automated extractor, according to the manufacturer's instructions. The Cobas Taqman 48 analyzer was used for automated Realtime PCR amplification and detection of PCR products according to the manufacturer's instructions (Roche Molecular Systems, Pleasanton, California, USA) with a detection limit of 15 IU/ml. HCV RNA-positive samples were genotyped using an HCV real-time genotype kit (AmpliSens HCV-genotype-FRT PCR kit) that was able to detect HCV genotypes 1a, 1b, 2, 3, and 4, following the manufacturer's instructions.

cDNA synthesis and NS5B gene amplification

cDNA was generated using the high-capacity kit (Applied Biosystems) according to the manufacturer's instructions. A nested PCR was carried out using the following primers pairs targeting NS5B, corresponding to codons 221–345[1].

outer sense	5\ -TACCAT CATGGCTAA(A/G)AA(C/T)-GAGGT (8008–8032)
outer antisense	ATGATGTTATGAGCTCCA (A/G) GTC (A/G) TA (8663–8687)
inner sense	5\TATGA(C/T) ACCCGCTG (C/T)TTTGAC (8256 –8276)
inner antisense	5\-CCTGGTCATAGCCTCCGTGAA (8616–8636)

Direct nucleotide sequencing and sequence analysis

The nested PCR products were purified by QIAquick PCR Purification Kit (QIAgen, Hilden, Germany) and sequenced in an automated sequencer. The amino acid sequence diversity of the NS5B genes were analyzed using data on the Genafor Open Services for Medical Research Website (https://www.genafor.org/index.php). NS5B sequences were submitted in the GenBank database under the following accession numbers: MN794404 - MN794412, MN894517 and MW307936**.**

RESULTS AND DISCUSSION

Results

A total of 850 patients were included; CHC was found in 548 while liver cirrhosis in 302 patients.

Demographic characteristics and laboratory data

The mean age was significantly different between both groups (51.46 ± 10.78) and the range between 21 and 75 years. Out of all enrolled patients; 506 patients (59.5%) were males and 344 patients (40.5%) were females. Co-morbidities in the form of diabetes mellitus, hypertension and ischemic heart disease were present in 228 (26.8%), 68 (8%) and 42 (4.9%), respectively. A total of 94 patients (11.1%) were INF experienced and 64.5% received the dual therapy of SOF + DCV (table 1).

In both groups there was a significant reduction in ALT, AST, blood glucose level, FIB-4 and APRI scores following therapy. Moreover, albumin was significantly increased in both groups following treatment (table 2).

Efficacy results

There was a significant difference between both groups regarding SVR12 with higher SVR in CHC patients ($p < 0.001$). The majority (91.2%) of the studied patients achieved SVR12 and 75 patients (8.8%) failed to achieve SVR12 (table 3). SVR12 was not significantly different between treatment-experienced patient than treatment-naïve patients but it was significant only in those receiving SOF+DAC + RBV for 12 weeks (69.2% for treatment-naïve patients and 98.0% for treatment-experienced patients, $p = 0.001$).

Table 1: Demographic characteristics and Laboratory data of the enrolled patients.

Items	**Total (n= 850)**	**CHC group (n= 548)**	**Liver cirrhosis group (n= 302)**	***P* value**
Age (years)	51.46 ± 10.78	50.14 ± 11.78	53.86 ± 8.13	< **0.001**
Sex Male Female	 506 (59.5%) 344 (40.5%)	 336 (61.3%) 212 (38.7%)	 170 (56.3%) 132 (43.7%)	 0.08
BMI (kg/m^2)	25.90 ± 4.39	26.33 ± 4.16	25.11 ± 4.70	0.06
Smoking	312 (36.7%)	201 (36.7%)	111 (36.8%)	0.52
Diabetes mellitus	228 (26.8%)	153 (27.9%)	75 (24.8%)	0.18
Hypertension	68 (8%)	42 (7.7%)	26 (8.6%)	0.35
IHD	42 (4.9%)	29 (5.3%)	13 (4.3%)	0.23
Residence Rural Urban	 682 (80.2%) 168 (19.8%)	 435 (79.4%) 113 (20.6%)	 247 (81.8%) 55 (18.2%)	 0.22
Occupation Unemployed Worker Employed Student	 332 (39.1%) 281 (33.1%) 180 (21.2%) 57 (6.7%)	 220 (40.1%) 177 (32.3%) 113 (20.6%) 38 (6.9%)	 112 (37.1%) 104 (34.4%) 67 (22.2%) 19 (6.3%)	 0.78
INF experienced	94 (11.1%)	66 (12%)	28 (9.3%)	0.13
Regimens Triple therapy Dual therapy	 302 (35.5%) 548 (64.5%)	 0 548 (100%)	 302 (100%) 0	 < 0.001

Table 2: Difference of the laboratory data between the two groups of enrolled patients before and after treatment**.**

	CHC group (n= 548)			**Liver cirrhosis group (= 302)**		
	Baseline	**Follow up**	***P* value**	**Baseline**	**Follow up**	***P* value**
Hemoglobin (g/dl)	13.37 ± 1.33	13.01 ± 2.34	0.05	11.87 ± 1.20	11.22 ± 1.90	0.25
Leucocyte (10^3/ul)	6.47 ± 1.71	6.24 ± 2.68	0.28	6.36 ± 1.85	6.35 ± 1.96	0.18
Platelets (10^3/ul)	238.82 ± 51.05	206.43 ± 57.18	0.42	144.09 ± 24.05	154.09 ± 19.45	0.40
ALT (u/l)	51.55 ± 20.84	29.47 ± 5.54	< **0.001**	61.27 ± 19.72	30.97 ± 7.42	< **0.001**
AST (u/l)	65.40 ± 28.13	24.99 ± 9.05	< **0.001**	71.09 ± 28.12	25.46 ± 8.09	< **0.001**
Bilirubin (mg/dl)	0.86 ± 0.28	0.87 ± 0.32	0.25	0.91 ± 0.19	0.87 ± 0.38	0.22
Direct bilirubin (mg/dl)	0.31 ± 0.11	0.30 ± 0.10	0.26	0.36 ± 0.21	0.31 ± 0.11	0.86
Albumin (mg/dl)	4.05 ± 0.46	4.11 ± 0.46	< **0.001**	3.29 ± 0.48	3.45 ± 0.39	< **0.001**
INR	1.02 ± 0.10	1.02 ± 0.08	0.22	1.41 ± 0.19	1.03 ± 0.10	0.10
Urea (mg/dl)	4.11 ± 2.20	4.10 ± 2.11	0.10	3.17 ± 1.38	3.98 ± 1.23	0.63
Creatinine (mg/dl)	1.17 ± 0.17	0.97 ± 0.21	0.11	0.97 ± 0.19	0.99 ± 0.22	0.34
RBG (mg/dl)	123.22 ± 39.78	106.05 ± 3.07	< **0.001**	131.54 ± 43.21	114.24 ± 34.34	< **0.001**
FIB-4	1.34 ± 0.71	0.45 ± 0.23	< **0.001**	3.49 ± 0.49	2.45 ± 1.19	< **0.001**
APRI	0.44 ± 0.29	0.39 ± 0.19	< **0.001**	2.13 ± 0.23	1.51 ± 0.72	< **0.001**

Table 3: Sustained virological response rate among the enrolled patients **.**

	All patients (n= 850)	**CHC (n= 548)**	**Liver cirrhosis (n=302)**	***P* value**
SVR				**< 0.001**
Yes	775 (91.2%)	528 (96.4%)	247 (81.8%)	
No	75 (8.8%)	20 (3.6%)	55 (18.2%)	

Safety profile of sofosbuvir/daclatasvir

The most frequent recorded adverse events were headache 20%, fatigue 14% and myalgia 5.2%. Diarrhea. Insomnia was noticed in only 1.2%. These adverse events occurred more frequently in liver cirrhosis group. Both groups had no significant difference as regard adverse events with exception of significantly higher frequency of diarrhea among patients with liver cirrhosis 7.3% vs. 3.1%; *P*= 0.04 (figure 1). Anemia developed in 25 patients 8.3% among those who received SOF+DCV+RBV with dose reduction of ribavirin and, erythropoietin was used in 10 patients. No patients had to stop treatment because of adverse events.

Frequency and type of RAVs

Because of financial issue, baseline RAVs were tested in a random sample among the enrolled patients. A randomly selected sample of 100 patients with chronic HCV were tested. Ten patients 10% were found to have RAVs before DAAs therapy, two patients of them became non responder on follow up after the end of treatment (figure 2). The most frequent baseline RAVs were R270K, K304R, R231K, P300T, V252A (table 4).

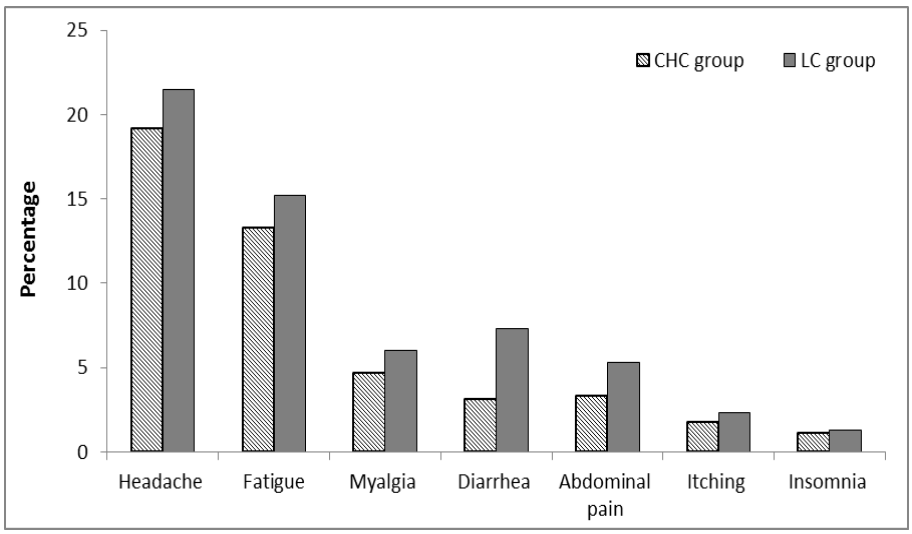

Fig. 1: Frequency of adverse events among the studied groups.

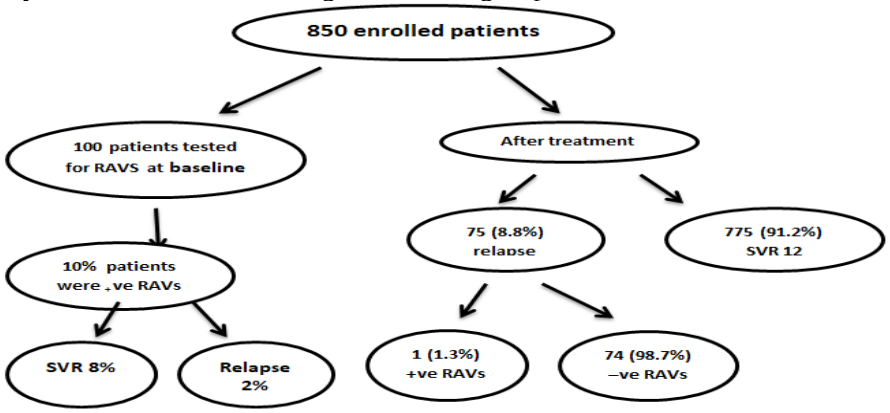

Fig. 2: Flow chart showing the study groups according to RAVs testing.

By the end of follow up, RAVs were tested in those patients with relapse (n= 75) and they were detected in only one patient (E237D) who did not have baseline RAVs. while the two non-responder patients with baseline RAVs were retested after the end of treatment but no RAVS could be detected.

The characteristics of patients with RAVs showed that the 2 cases who had baseline RAVs were females, higher viral load relative to the third patient who did not have baseline RAVs and they had genotype 4a while the third patient had genotype 4o (table 5).

Table 4: The outcomes of RAVs to HCV NS5B.

HCV patients IDs										
30	**98**	**104**	**215**	**326**	**407**	**538**	**609**	**717**	**828**	**448**
RAVs position										
R231K	E237G	R231K	R231K	R231K	A235T	R231K	R231K	A231G	K270R	E237D
V252A	T254A	V252A	R270K	A235V	E237G	V252A	V252A	I251V	M300T	
T254A	A255S	Y285F	Y285F	V252A	D244A	T254A	T254A	R254K	G333A	
R270K	R270K	P300T	P300T	R270K	V252A	R270K	R270K	N255S		
T286P	L293M	K304R	K304R	Y285F	H267Y	P300T	V285F	K270R		
P300T	P300T	E327D	E327D	P300T	R270K	K304R	P300T	K307R		
K304R	K304R	N333S	N333S	K304R	L273F	E327D	K304R			
V322I	E327D			E327D	L293M		E327D			
E327D	N333R				P300T		N333S			
N333S	R337G				I303L		M343I			
	A342V				K304R					
	M343L				E327D					
					N333A					

Table 5: Characteristics of patients who relapsed and had RAVs.

ID	**Age (years)**	**Gender**	**Co-morbidities**	**Hemoglobin(g/dl) Female :12-14 Male : 13-15**	**Viral load x 10^3 (IU/ml)**	**FIB-4**	**APRI score**	**Genotype**	**SVR**
A-Treatment-naïve patients (with baseline RAVS)									
104	23	Female	Non	12.5	11	1.7	0.4	4a	No
407	55	Female	DM	11	14.1	1.3	0.5	4a	No
B-Non-responder									
448	57	Male	Non	15	1.7	1.5	0.6	4o	No

Discussion

Prevalence of HCV in Egypt is one of the highest rate worldwide. The recent introduction of DAA therapy has revolutionized the treatment of chronic HCV infection particularly in Egypt, with very high SVR rates in clinical trials and slightly lower rates in real-life cohorts[7]. The emergence of HCV RAVs could be a cause for this difference. To our knowledge, few studied were conducted in Egypt to assess the RAVs and their impact on treatment outcome in genotype 4 HCV patients. Therefore, this prospective study evaluated the safety of sofosbuvir plus daclatasvir with or without RBV in HCV patients and studied the occurrence of RAVs in cases of relapse.

According to our study, SVR was achieved in 91.2% of patients and SVR12 rate was significantly higher in patients with CHC relative to liver cirrhosis. This is in parallel to an Egyptian study conducted by Ebid *et al.*,[8] who showed SVR-12 in 94.7% of patients receiving SOF + DAC regimen for 12 weeks. Similarly, Pol *et al.*,[9] showed that SVR12 was obtained in 95% of patients, ranging from 92% (12-week SOF+DAC) to 99% (24-week SOF+ DAC + RVB). Lower SVR to SOF + DAC in patients with liver cirrhosis was documented by Poordad *et al.*,[10] who showed that advanced liver disease has lower SVR rates (82%).

In this study the used DAAs regimen (SOF+DCV± RBV) was safe and the detected adverse events were tolerable. Occurrence of the reported adverse events was more frequent in patients with liver cirrhosis. This is in concordance with an Egyptian study by Shiha *et al.*,[11]. who found that SOF+DCV regimen showed no treatment-related serious adverse events either in CHC or liver cirrhosis genotype 4 patients. They reported the most common adverse events were fatigue, headache, anemia, cough, and sleeping disorders and the most common adverse events was fatigue, while in our study the most commonly reported adverse events was headache. Dose modification of RBV was done in 2.6% of patients while in our study it was needed in 8.3% of patients, however this difference could be explained by different sample size in the studies. Similar to our study, Lashen *et al.*,[7] reported no severe adverse events or deaths due to drugs except anemia due to RBV.

Testing of genotype in this study showed that all patients had genotype 4, which is in concordance with a systematic review that reported dominance of genotype 4 in Egypt, accounting for 92.5% of cases Amer *et al.*, Regarding RAVs in this study, random testing of 100 patients before treatment showed 10% had baseline RAVs, however 2 out of these cases showed relapse. Meanwhile, one relapsed patient had RAVs following treatment without baseline RAVs. This is in concordance with Amer *et al.*,[12] who tested RAVS in Egyptian patients who failed DAAs therapy by SOF+DCV by deep sequencing method and found RAVs in 3 patients; two of them had relapse after treatment and one patient had viral breakthrough during therapy. The most frequent RAVs detected before treatment were R270K, K304R, R231K, P300T, V252A. This is similar to Ahmed *et al.*,[13] study as K304R (82.4%), E327D and P300T (76.5% each) substitutions were the most distributed in the tested samples and one substitution mutation (E237G) was identified in the non-responder sample.

The identified RAVs to HCV NS5B in the current study following treatment was E237D. Similarly, the SOLAR-2 clinical trial recorded the NS5B RAS at failure at E237G[14]. Conversely, another study reported the presence of E237G in one genotype 4 patient with no significance on treatment response[15]

The occurrence of naturally occurring baseline RAVs in this study did not appear to have an effect on the therapeutic response after treatment, since most HCV were GT 4-monoinfected patients had achieved SVR (> 90%). So, RAVs testing is not routinely recommended before treatment of HCV infection.

Therefore, further longitudinal studies on larger number of patients with using new generations of sequencing are required to assess the RAVs and its relation to treatment failure and to compare their occurrence between different DAAs regimes.

Conclusion

This observational real-life study showed that treatment with sofosbuvir and daclatasvir ± ribavirin in HCV genotype 4 patients was safe, tolerable and without significant side effects. Although RAVs could be present in HCV

patients before therapy, they do not alter the treatment response. Therefore, RAVs testing is not routinely recommended before treatment of HCV infection as resistant variants could occur naturally in patients with HCV. Most of these baseline substitutions did not seem to negatively impact treatment outcome, especially for GT 4 since most patients achieved SVR.

Conflict of interest

No conflict of interest.

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REFERENCES

1. M. Abdel-Hamid, M. El-Daly, V. Molnegren, S. El-Kafrawy, S. Abdel-Latif, G .Esmat, *et al.,* "Genetic diversity in hepatitis C virus in Egypt and possible association with hepatocellular carcinoma", ***J Gen Virol ,*** 88(5),1526–1531(2007).
2. J. Chhatwal, X. Wang, T. Ayer, M. Kabiri, R.T. Chung, C. Hur, *et al.,* "Hepatitis C disease burden in the United States in the era of oral direct-acting antivirals", ***Hepatology*** , 64(5),1442-1450(2016).
3. D.R. Nelson, J.N. Cooper, J.P. Lalezari, E. Lawitz, P.J. Pockros, N. Gitlin, *et al.*, "All-oral 12-week treatment with daclatasvir plus sofosbuvir in patients with hepatitis C virus genotype 3 infection: ALLY-3 phase III study", ***Hepatolog***y, 61(4),1127-1135(2015).
4. A. Ahmed and D.J. Felmlee, "Mechanisms of Hepatitis C Viral Resistance to Direct Acting Antivirals", ***Viruses,*** 7(12), 6716-29 (2015).
5. F. A. Di Lello, K .Neukam, M. Parra-Sanchez, Z. Plaza, V. Soriano, C. Cifuentes, *et al.,* "Hepatitis C virus genotype 4 in Southern and Central Spain does not originate from recent foreign migration waves", ***J Med Virol,*** 85(10),1734-1740 (2013).
6. EASL recommendations on treatment of hepatitis C 2016, ***J Hepatol,*** 66(1),153-194(2017).
7. S. A. Lashen, M.M. Shamseya, M.A. Madkour and G. A. Aboufarrag,"Tolerability and effectiveness of generic direct-acting antiviral drugs in eradication of hepatitis C genotype 4 among Egyptian patients", ***Liver Int ,*** 39(5),835-843 (2019).
8. A.H. Ibrahim Mohammed Ebid, O. Ashraf Ahmed, S. Hassan Agwa, S. Mohamed Abdel-Motaleb, A. Mohamed Elsawy, R.S. Hagag, "Safety, efficacy and cost of two direct-acting antiviral regimens: A comparative study in chronic hepatitis C Egyptian patients", ***J Clin Pharm Ther,*** 45(3),539-546 (2020)
9. S. Pol, M. Bourlière, S. Lucier, C. Hézode, C. Dorival, D. Larrey, *et al.,*"Safety and efficacy of daclatasvir-sofosbuvir in HCV genotype 1-mono-infected patients", ***J Hepatol,*** 66(1),39-47(2017).
10. F. Poordad, E. R. Schiff, J. M. Vierling, C. Landis, R. J Fontana, R. Yang, *et al.,*"Daclatasvir with sofosbuvir and ribavirin for hepatitis C virus infection with advanced cirrhosis or post-liver transplantation recurrence", ***Hepatology,*** 63(5),1493-1505 (2016).
11. G. Shiha, R. Soliman, M. ElBasiony, A. Hassan and N. Mikhail, "Sofosbuvir plus Daclatasvir with or without ribavirin for treatment of chronic HCV genotype 4 patients: real-life experience", ***Hepatol Int***,12(4),339-347(2018).
12. F. Amer, M. M. Yousif, N. M. Hammad, D. Garcia-Cehic, J. Gregori, A. Rando-Segura, *et al.,*"Deep-sequencing study of HCV G4a resistance-associated substitutions in Egyptian patients failing DAA treatment", ***Infect Drug Resist,***12(10), 2799-2807(2019).
13. H. R. Ahmed, N. G Waly, R. M. Abd El-Baky, R. Yahia, H. F. Hetta, A. M. Elsayed, R.A.Ibrahem, "Distribution of naturally-occurring NS5B resistance-associated substitutions in Egyptian patients with chronic Hepatitis C", ***PloS one ,***16(4),e0249770 (2021).
14. M. Manns, D. Samuel, E. J. Gane, D. Mutimer, G. McCaughan, M. Buti, *et*

al.,"Ledipasvir and sofosbuvir plus ribavirin in patients with genotype 1 or 4 hepatitis C virus infection and advanced liver disease: a multicentre, open-label, randomised, phase 2 trial", ***Lancet Infect Dis*** **,**16(6), 685-697(2016).

15. C. Hedskog, B. Parhy, S. Chang, S. Zeuzem, C. Moreno, S. D. Shafran, *et al.*,"*Identification of 19 novel hepatitis C virus subtypes—further expanding HCV classification*", ***Open Forum Infect Dis,*** 6(3),ofz076 (2019).
16. A. Vijayananthan and O. Nawawi, "The importance of Good Clinical Practice guidelines and its role in clinical trials", ***Biomed Imaging Interv J,*** 4(1),e5(2008).
17. T. Kanda, S. Matsuoka and M. Moriyama, "*Hepatitis C virus genotype 4-infection and interferon-free treatment in Egypt*", ***Hepatol Int,***12(4), 291-293(2018).

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نشرة العلوم الصيدلية
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تقييم سلامة عقاري السوفوسبيوفير والدكلاتاسفير والطفرات المقاومة بين المرضي المصريين المصابين بالالتهاب الكبدي الفيروسي (سي): دراسة مستقبلية

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تهدف هذه الدراسة الي تقييم مدي أمان استخدام عقاري السوفوسبيوفير اداكلاتاسفير لعلاج مرضي الالتهاب الكبدي الفيروسي المزمن "سي" من النوع الجيني الرابع لتقييم مدي حدوث الطفرات في المرضي الذين لم يستجيبوا للعلاج او مرضي الانتكاسة بعد العلاج بعقاري السوفوسبيوفير اداكلاتاسفيرمع تمييز أنواعها.

تشمل الدراسة المرضي الذين لديهم عدد كمي من فيروس "سي" بالدم سواء الذين لم يتلقوا أي علاج من قبل او المرضي الذين سبق علاجهم بعقار الانترفيرون ومرضي التليف الكبدي في مراحله الاولي معامل تشايلد "أ" و "ب".

اعتمد تشخيص التليف الكبدي في دراستنا علي العوامل الاتية: عد الصفائح الدموية اقل من ١٠x١٠٠ ^{٣}\١ لتر ، مستوي البيليروبين اكثر من ٢مجم ،البيومين اقل من ٣,٥جم ، اي ان ار اكتر ١,٧.وجود دوالي بالمريء بالمنظار.

تم استبعاد هؤلاء المرضي من الدراسة:

مرضي التليف الكبدي في مراحله المتأخرة معامل تشايلد "ج" و "د" ، مرضي نقص المناعة المكتسبة ومرضي الالتهاب الكبدي الفيروسي المزمن "بي" ، الالتهاب الكبدي المزمن لأسباب اخري مثل التهاب الكبد المناعي ، بيليروبين اكتر من ٢ مجم، ارتفاع انزيمات الكبد اكتر من ١٠ اضعاف المعدل الطبيعي ، عد الصفائح الدموية اقل من١٠x٥٠ ^{٣}\١ لتر، مرضي اورام الكبد.

تلقي كل مرضي الالتهاب الكبدي الفيروسي المزمن "سي" علاج ثنائي في حين تلقي كل مرضي التليف الكبدي علاج ثلاثي ولوحظ انه لا توجد فروق تذكر بين المجموعتين الا ان سن المرضي كان اعلي في مرضي التليف عن مرضي الالتهاب الكبدي الفيروسي المزمن.

أنماط العلاج المستخدمة كانت امنة وكان من بين أهم الاثار الجانبية المسجلة: الصداع (٢٠%) ، الاجهاد (١٤%)، الم العضلات (٥,٢%). الاسهال والام البطن والحكة حدثت في ٣٩مريض (٤,٦%) ، ٣٤مريض (٤%) ، ١٧ مريض (٢%) على التوالي. لوحظ حدوث الارق في (١,٢%) من المرضي. لم يلاحظ فروق في معدل الاثار الجانبية بين المجموعتين باستثناء ان حالات الاسهال كانت أكثر في مرضي التليف الكبدي (٧,٣%) في مقابل (٣,١%). وجد اختلاف كبير بين المجموعتين من حيث الاستجابة خلال اثني عشر اسبوع بعد العلاج. أغلب المرضي (٩١,٢%) استمرت الاستجابة لمدة ثلاثة أشهر بعد العلاج ولم تحدث استجابة خلال ١٢ اسبوع في (٨,٨%) فقط المرضي. لأسباب مادية لم نستطع عمل دراسة لطفرات فيروس سي لكل المرضي الذين شملتهم الدراسة وبالتالي تم اختيار مائة مريض بطريقة عشوائية كمجموعة ممثلة للدراسة ووجد ان ١٠% من المرضي لديهم فصائل مقاومة قبل بداية العلاج جميعهم من النوع الجيني الرابع ما عدا حاله واحدة من النوع الجيني الاول.

وجد في نهاية فترة الدراسة ان ٨,٨% من المرضي لم يظهروا استجابة للعلاج بعد ثلاثة أشهر من نهاية العلاج، تم عمل اختبار الطفرات في هؤلاء المرضي ووجد انه مريض واحد لديه طفره مقاومة (١,٣%) من المرضي الغير مستجيبين.