HEPATITIS C VIRUS INFECTION IN PATIENTS WITH DIABETES MELLITUS TYPE 2: A SEROPREVALENCE STUDY

Infecção pelo Vírus da Hepatite C em Pacientes com Diabetes Mellitus Tipo 2: um Estudo da Soroprevalência

Sabrina Gesteira de Souza[1] | Karina Yara Martins[2] | Juliano Machado de Oliveira[3] | Lize Vargas Ferreira[3]

[1] MD by Universidade Federal de Juiz de Fora

[2] Internal medicine resident physician at Universidade Federal de Viçosa

[3] Professor of medicine at Universidade Federal de Juiz de Fora

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ABSTRACT

Objectives: To evaluate the seroprevalence of hepatitis C virus (HCV) infection in diabetic patients treated at the type 2 diabetes mellitus (T2DM) outpatient clinic of the University Hospital of the Federal University of Juiz de Fora, Brazil (HU-UFJF). **Methods:** This is a cross-sectional study, in which a convenience sample of 145 patients with T2DM was evaluated. Demographic, clinical and anthropometric data were collected and testing for anti-HCV antibodies was performed. **Results:** a seroprevalence of 2.76% was found in the sample, with a 95% confidence interval of [1,07%, 6,87%]. The only other variable that had a statistically significant difference between the HCV-positive and HCV-negative groups was the waist circumference, which was greater in the HCV--positive group. **Conclusion:** Considering the potential for worse outcomes of hepatitis C when associated with T2DM, as well as worse glycemic control in HCV-infected patients, we suggest that active screening for this viral infection should be performed in patients with T2DM, through routine serology, which is feasible within the scope of Primary Care.

Keywords: Diabetes mellitus. Hepatitis C. Insulin resistance.

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INTRODUCTION

Hepatitis C is one of the most common causes of chronic liver disease worldwide[1]. It is caused by the hepatitis C virus (HCV), a member of the Flaviviridae family, the same as the dengue and yellow fever viruses. The epidemiological bulletin on viral hepatitis, prepared by the Brazilian Ministry of Health (MS) in 2021, reported a detection rate of confirmed cases of hepatitis C of 4.4 per 100,000 inhabitants in Brazil[2]. Mathematical models, in turn, estimate that 0.7% of the Brazilian population between 15 and 69 years of age is seropositive for HCV[3], while the worldwide seroprevalence is estimated at 1.4%[4].

The clinical manifestations of hepatitis C show great individual heterogeneity, and may progress, in many cases, to cirrhosis and liver cancer. As the disease usually has a long asymptomatic period, the diagnosis is often only made in advanced stages of the disease. Due to this reason, it is estimated that 80% of all individuals infected with HCV worldwide are unaware of their serological status[5].

This rate depends on testing, because in countries with more resources, where more people are tested, this number drops to 54%. On the other hand, it reaches 92% in the poorest countries[6].

The advent of treatments that allow the cure of hepatitis C led the World Health Organization (WHO) to set the goal of eradicating the disease by 2030[7]. However, one of the obstacles to treatment is indeed the identification and diagnosis of asymptomatic cases, which reinforces the need for better screening programs[6]. In Brazil, the MS currently recommends that all people over 40 years of age be tested at least once in their lifetime. Some groups at higher risk for the disease should be tested regardless of age and more frequently. This is the case, for example, of homeless people, sex workers, health professionals, and men who have sex with men, among others[8].

Previous studies have shown that the risk of HCV infection in patients with type 2 diabetes mellitus (T2DM) is higher. A meta-analysis that included 78,051 individuals reported a statistically significant odds ratio of 3.5 for HCV infection in T2DM patients compared to non-diabetic patients[9]. Other studies have also demonstrated the opposite way of association. A meta-analysis that included 34 studies identified that patients with HCV infection have a statistically significant odds ratio of 1.68 to also have the diagnosis of T2DM, compared to patients without the virus infection. Prospective studies also follow the same line, indicating a 67% greater risk of patients with HCV developing T2DM[10]. Therefore, many authors consider the association between the two diseases bidirectional[11].

The pathophysiological mechanisms by which many patients with HCV develop T2DM are not fully established[12]. Hypotheses suggest that HCV can alter insulin cell signaling, leading to resistance to the hormone and preventing it from regulating glucose metabolism[13-15]. Other possible mechanisms would be lower insulin secretion and dysregulation of hepatic glucose production[12]. Although the mechanism is unknown, it is known that patients with HCV and insulin resistance (IR) tend to present with a quicker progression to liver cirrhosis and hepatocellular carcinoma (HCC) than those without IR[16]. Furthermore, in patients with advanced cirrhosis, T2DM increases the risk of liver cancer by four to five times, leading to worse outcomes[17].

Taking into account these findings, it is possible to conclude that patients with T2DM can be considered a group with specific clinical and epidemiological characteristics regarding HCV infection, both due to the higher risk of infection and to the worse outcomes more frequently observed in this group. Thus, testing and screening strategies should consider these issues, especially considering the high number of patients unaware of their status, but who risk an eventual late diagnosis leading to worse outcomes. The present study therefore seeks to study the seroprevalence of HCV infection in diabetic patients treated at the T2DM outpatient clinic of the University Hospital of the Universidade Federal de Juiz de Fora (HU-UFJF), in the city of Juiz de Fora, Brazil. Our objective is to carry out a cross-sectional epidemiological diagnosis to contribute to future public health policies aimed at improving the diagnosis of HCV, to identify undiagnosed cases and expand treatment availability.

METHODS

This is a cross-sectional study that evaluated a convenience sample of 145 patients with T2DM, after obtaining a favorable opinion from the Research Ethics Committee (CEP) of the HU-UFJF, under Certificate of Ethical Assessment Aproval (CAAE) No. 76969417.3.0000.5133.

All patients with T2DM seen from September 2017 to March 2018 at the Endocrinology outpatient clinic of the HU-UFJF Health Care Center (HU/CAS), in the city of Juiz de Fora, Brazil, were invited to undergo a rapid screening test for HCV after their clinic appointment.

The medical chart was reviewed to gather demographic (gender and age), clinical (time since diagnosis of T2DM, levels of glycosylated hemoglobin (HbA1c), total cholesterol, HDL, LDL, triglycerides, aspartate aminotransferase/AST and alanine aminotransferase/ALT) and anthropometric (weight, height, body mass index – BMI – and waist circumference) data. Waist circumference was measured at the midline between the costal margin and the upper edge of the iliac crest, at the time of the HCV testing.

Anti-HCV antibody testing was performed using the Alere HCV kit (produced by SD-65, Republic of Korea, and imported to Brazil by Alere SA), with a diagnostic sensitivity of 99.4% and specificity of 99.7%[18,19]. This is a qualitative, immunochromatographic, single-use, disposable test that provides visual results after 20 minutes for anti-HCV antibody detection.

Microsoft Excel 2016 was used to tabulate the data and obtain descriptive statistics and the seroprevalence rate of HCV infection in the sample. The two-sample, one-tailed Student's t test was then used to compare the patients with positive serology to those with negative serology regarding clinical, demographic and anthropometric variables, considering the hypothesis that seropositive patients have worse metabolic parameters.

The seroprevalence rate obtained in the sample was then used to calculate a 95% confidence interval (CI) for an estimated seroprevalence rate in a population with similar characteristics (patients with T2DM in an outpatient setting). The Wilson score method was used for this purpose, since this is the most appropriate test for samples in which the observed proportions are small[20].

RESULTS

A total of 145 patients who underwent the rapid test for anti-HCV antibodies were included. No patient refused to be tested or sign the informed consent form, and therefore, no patients were excluded from the study. The majority of the sample was female (66.2%), and the mean age was 61 years, ranging from 49 to 73 years. HCV seropositivity was identified in 2.76% of the patients studied, with a 95% CI [1.07%, 6.87%]. The mean time since T2DM diagnosis was 10.9 years $\pm$ 8.44.

The mean HbA1c level in the total sample was 7.6% $\pm$ 1.88%. As for the lipid profile, we found a mean total cholesterol level of 174.2 mg/dL; a mean HDL level of 45.2 mg/dL; and a mean LDL level of 95.9 mg/dL. The mean triglyceride level was 170.5 mg/dL. Regarding liver enzymes, we found a mean AST level of 26.5 mg/dL and a mean ALT level of 29.5 mg/dL. The mean BMI was 31.7 kg/m2 $\pm$ 7.00.

The only characteristic that significantly differed between anti-HCV seropositive patients compared to seronega-

tive ones was the waist circumference, which was higher in the seropositive group (mean = 116.5 cm, standard deviation = 2.12 cm) compared to the seronegative group (mean = 106.4 cm, standard deviation = 14.47 cm); t(143) = 2.405, p = 0.009. Table 1 summarizes the clinical-demographic findings of the entire sample, as well as those of each group.

DISCUSSION

In this study, we found a HCV seropositivity rate of 2.76% in outpatients with T2DM (95% CI: 1.07% 0 6.87%). This finding, from the endocrinology outpatient clinic at HU-UFJF, is almost numerically twice the prevalence estimated by the WHO in the world population, which is 1.4% (reported 95% CI: 1.2%-1.5%)[4]. However, it is not possible to conclude that the populational seropositivity rate in similar patients with T2DM is higher

TABLE 1 - Clinical and demographic characteristics of the total sample of patients with type 2 diabetes mellitus and of the hepatitis C seropositive and seronegative groups

CHARACTERISTIC	TOTAL (N=145)	HCV POSITIVE (N=3)	HCV NEGATIVE (N=142)
SEX (M:F)	49 (33.8%):96 (66.2%)	1 (33.3%):2 (66.7%)	48 (33.8%):94 (66.2%)

CHARACTERISTIC	TOTAL (N=145)		HCV POSITIVE (N=3)		HCV NEGATIVE (N=142)	
	Mean	Standard-deviation	Mean	Standard-deviation	Mean	Standard-deviation
Age (years)	61.4	12.07	74.7	22.03	61.1	11.75
Time since diagnosis of DM (years)	10.9	8.44	5.0	4.24	11.0	8.46
Glycosylated hemoglobin % (HbA1c)	7.6	1.88	8.6	1.91	7.6	1.88
BMI (kg/m²)	31.7	7.00	31.5	2.81	31.7	7.07
Waist circumference (cm)	106.6	14.39	**116.5***	2.12	**106.4***	14.47
Total cholesterol (mg/dL)	174.2	43.41	191.0	56.57	173.9	43.41
Triglycerides (mg/dL)	170.5	107.37	100.5	33.23	171.2	107.49
HDL (mg/dL)	45.2	13.22	74.0	22.63	44.7	12.63
LDL (mg/dL)	95.9	5.52	111.5	75.66	95.7	35.08
AST (mg/dL)	26.5	12.51	37.5	28.99	31.3	51.77
ALT (mg/dL)	29.5	15.55	27.0	2.83	34.6	53.77

DM: diabetes mellitus; BMI: body mass index; HDL: high density lipoprotein; LDL: low density lipoprotein; AST: aspartate aminotransferase; ALT: alanine aminotransferase.
* p = 0.009

than the global prevalence reported by the WHO, due to the overlapping IC of the two reported rates. Regarding the prevalence in the Brazilian population, the rate found in this study was almost four times higher than the rate found in a large sample of 484,300 patients who underwent rapid testing for HCV, which was 0.76%. The same study used a mathematical model to extrapolate the finding for the total Brazilian population, considering other variables. The study found an estimated prevalence of 0.7%[4], a value not contained in the CI of this sample. Although no CI was reported for the Brazilian population study, this finding suggests that the seropositivity in the general population in Brazil is lower than that observed in patients with T2DM undergoing outpatient treatment, considering the limitations of this study.

Another finding that should be highlighted in this sample was the high prevalence of clinical phenotypes associated with IR. In addition to having longstanding T2DM, most patients also had centripetal obesity and dyslipidemia, although, on average, their glycemic control was fair (mean HbA1c: 7.6%). However, these variables did not show a statistically significant difference between HCV seropositive and seronegative groups, except for waist circumference, which was significantly greater in the HCV-positive group. This particular finding is interesting, as it is in line with the results of a case-control study conducted in Taiwan, which also found a higher prevalence of increased waist circumference in HCV-seropositive patients compared to controls[21]. Another study that evaluated the rate of visceral adiposity in patients with hepatitis C – another way of estimating the amount of visceral fat – found an association of this measure with the degrees of steatosis and hepatic necroinflammation, as well as with a higher HCV viral load, regardless of IR. The authors suggested this finding reflects dysfunction of the adipose tissue in patients with hepatitis C, which could play a direct role in the pathophysiology of the infection. This could be mediated by the endocrine and immunological functions of the adipose tissue, on which HCV could act by favoring the production of pro-inflammatory cytokines[22]. In addition, findings that indicate greater adiposity – such as greater body fat mass and higher levels of hepatic steatosis – were also associated with the presence and faster progression of liver fibrosis in patients infected with HCV, as well as greater viral resistance to antiviral therapy with interferon. From these findings, it is believed that adipose tissue may favor the progression of HCV infection, possibly due to immunological mechanisms such as T cell dysfunction resulting from leptin resistance[23].

The high prevalence of IR and T2DM in HCV patients has already been widely studied, as was the opposite association[9,10]. Therefore, the association between HCV and T2DM is thought to be bidirectional, that is, the HCV may predispose to the onset of T2DM and aggravate it, but IR itself may worsen the evolution and prognosis of the infection[11]. This hypothesis is supported by studies that have shown that patients with T2DM and HCV have a higher incidence of complications resulting from T2DM, which reinforces the importance of HCV screening in this population[24]. Likewise, studies have reported a reduction in HbA1c levels in patients who achieved viral suppression after treatment with direct-acting antivirals against HCV[25].

The exact pathophysiological mechanism that mediates the association between IR and HCV infection is not completely established. One of the hypotheses postulates that HCV has a direct action on hepatocytes, causing a decrease in insulin receptor expression, an increase in its degradation, and phosphorylation of its serine residues, which would result in resistance to the hormone. In addition, another hypothesis suggests an indirect action of HCV, by inducing the expression of inflammatory cytokines such as Tumor Necrosis Factor Alpha (TNF-α) and Interleukin 6 (IL-6), which promote the activation of molecular cascades that lead to IR24-26. Furthermore, there is also evidence that HCV proteins, such as NS5A (HCV nonstructural protein 5A), induce the expression of the phosphoenolpyruvate carboxykinase and glucose 6-phosphatase genes, which are enzymes that make up the neoglucogenesis pathway, and therefore lead to an increase in blood glucose levels[27-29].

Another point that deserves attention is the influence of IR and T2DM on the clinical outcomes of patients with hepatitis C. Studies indicate diabetic patients with hepatitis C, with or without cirrhosis, have worse clinical outcomes compared to non-diabetics, such as increased risk of developing HCC[17,30]. In those with cirrhosis, however, there is an increased risk of developing ascites, renal failure and bacterial infections[17]. The use of metformin, in turn, was associated with a lower incidence of HCC, liver transplantation, and death due to liver complications in patients infected by HCV with T2DM and cirrhosis[31]. Furthermore, the presence of IR can negatively impact the treatment of HCV, as studies have shown a lower rate of sustained viral suppression in these patients following treatment with ribavirin and peginterferon alfa, which were the treatment of choice for HCV until recently[32]. The combination of metformin with this treatment scheme, however, not only improved the IR, but also increased the rate of sustained viral suppression[33].

More recently, new drugs with a direct antiviral action were approved for the treatment of HCV. They can eradicate the virus in most cases, apparently irrespective of the presence of IR before treatment[34]. However, even if these drugs may lead to a complete and sustained eradication of the virus, T2DM still seems to negatively influence clinical outcomes. This is suggested by a study with 33,000 patients infected with HCV, in which the presence of T2DM before treatment with direct-acting antivirals was associated with higher mortality and negative hepatic outcomes (cirrhosis and decompensations), even after sustained viral suppression. In those with cirrhosis before treatment, T2DM was associated with the development of HCC, despite viral suppression[35]. Thus, although studies have also shown that eradicating the virus reduces IR and also leads to better T2DM control[34], clinicians should remain vigilant regarding patients with this comorbidity, even after viral suppression[35]. Finally, the importance of diagnosing and treating T2DM in these patients is reinforced by findings that indicate that metformin may decrease the risk of HCC after successful antiviral treatment[36].

This study has limitations that must be discussed. First, the sample size is small,

considering the expected prevalence of HCV infection in this population, based on previous literature. This may have resulted in low statistical power, hampering the detection of a possible statistically significant difference between the prevalence of HCV in the sample and its estimated prevalence in the general population, according to WHO data. However, our findings, although not statistically significant, are compatible with a previous meta-analysis that reported an odds ratio of 3.5 for the diagnosis of hepatitis C in diabetic patients compared to controls[9]. This limitation also hampered the power to detect possible differences in other laboratory parameters, although an interesting finding involving visceral adiposity was observed in diabetic patients. The lack of other data in the sample also prevented the assessment of other parameters that could be addressed, such as the degree of liver fibrosis and viral load. Also, a rapid serological test was used for the diagnosis of the infection, which, despite its good specificity and sensitivity[18], was not confirmed by another method. Therefore, there was a risk of false positives and false negatives. Finally, the cross-sectional design of the study does not allow further conclusions about possible causal relationships between variables.

In summary, this study reinforces the importance of screening for hepatitis C in patients with T2DM, since, 2.76% of the participants in a random sample, whose serological status was previously unknown, tested positive for HCV. It is true that the primary objective of treating hepatitis C is to eradicate the virus from the organism and, therefore, to reduce the incidence of complications resulting from chronic liver disease, the transmissibility of the virus and its comorbidities. Currently, there are highly effective therapeutic alternatives with fewer side effects, which make it easier to achieve these goals and even allow envisioning to eradicate hepatitis C by 2030. However, it is necessary to aim not only for such outcomes, but also for longer survival with a better quality of life for the patient. For this, it is necessary to go beyond the eradication of HCV, because, as discussed, other systemic changes, especially T2DM, are associated with complications and lower survival, even after total viral suppression[34]. Thus, this study draws attention to the importance of routine screening for T2DM in patients with HCV, aiming at early diagnosis and treatment, which is feasible in primary care, including in individuals with normal liver function.

DISCLOSURES

The authors declare that they have no conflicts of interest to disclose.

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INFORMATION ABOUT THE ARTICLE

Universidade Federal de Juiz de Fora - Juiz de Fora - MG - Brazil

Mailing address:
Rua Chanceler Oswaldo Aranha, 135, 201
CEP: 36025-007 – Juiz de Fora - MG - Brazil

Corresponding author:
Lize Vargas Ferreira
lize.vf@gmail.com

REFERENCES

[1] Castro R, Perazzo H, Grinsztein B, Veloso VG, Hyde C. Chronic Hepatitis C: An Overview of Evidence on Epidemiology and Management from a Brazilian Perspective. *Int J Hepatol.* 2015; 2015:852968.

[2] Secretariat of Health Surveillance. Boletim Epidemiológico - Hepatites Virais - Número Especial (Epidemiological Bulletin - Viral Hepatitides - Special issue) [Internet]. Brasilia: Brazilian Ministry of Health; 2021 [accessed on 12/17/2022]. Available at: https://www.gov.br/saude/pt-br/centrais-de-conteudo/publicacoes/boletins/epidemiologicos/especiais/2021/boletim-epidemiologico-de-hepatite-2021.pdf.

[3] Benzaken AS, Girade R, Catapan E, Pereira GFM, Almeida ECD, Vivaldini S *et al.* Hepatitis C elimination by 2030 is feasible in Brazil: a mathematical modelling approach. *Braz J Infect Dis.* 2019; 23(3):182-190.

[4] World Health Organization. WHO estimates of the prevalence and incidence of hepatitis C virus infection by WHO region, 2015 [Internet]. Geneva, Switzerland: World Health Organization; 2017 [accessed on 12/17/2022]. Available at: https://apps.who.int/iris/bitstream/handle/10665/277005/WHO-CDS-HIV-18.46-eng.pdf.

[5] World Health Organization. WHO Global Hepatitis Report, 2017 [Internet]. Geneva, Switzerland: World Health Organization; 2017 [accessed on 12/17/2022]. Available at: https://apps.who.int/iris/rest/bitstreams/1082595/retrieve.

[6] Roudot-Thoraval F. Epidemiology of hepatitis C virus infection. *Clin Res Hepatol Gastroenterol.* 2021; 45(3):101596.

[7] World Health Organization. Global health sector strategies on, respectively, HIV, viral hepatitis and sexually transmitted infections for the period 2022-2030 [Internet]. Geneva, Switzerland: World Health Organization; 2022 [accessed on 12/17/2022]. Available at: https://cdn.who.int/media/docs/default-source/hq-hiv-hepatitis-and-stis-library/full-final-who-ghss-hiv-vh-sti_1-june2022.pdf.

[8] Brazilian Ministry of Health. Hepatites Virais - Rastreamento/diagnóstico (Viral hepatitides - Screening/diagnosis) [Internet]. Brasilia: Brazilian Ministry of Health; 2022 [accessed on 12/17/2022]. Available at: https://linhasdecuidado.saude.gov.br/portal/hepatites-virais/unidade-de-atencao-primaria/rastreamento-diagnostico.

[9] Guo X, Jin M, Yang M, Liu K, Li JW. Type 2 diabetes mellitus and the risk of hepatitis C virus infection: a systematic review. *Sci Rep.* 2013; 18(3):1-8.

[10] White DL, Ratziu V, El-Serag HB. Hepatitis C infection and risk of diabetes: a systematic review and meta-analysis. *J Hepatol.* 2008; 49(5):831–844.

[11] Hammerstad SS, Grock SF, Lee HJ, Hasham A, Sundaram N, Tomer Y. Diabetes and hepatitis C: a two-way association. *Front Endocrinol (Lausanne).* 2015; 6:134.

[12] Bahtiyar G, Shin JJ, Aytaman A, Sowers JR, McFarlane SI. Association of diabetes and hepatitis C infection: epidemiologic evidence and pathophysiologic insights. *Curr Diab Rep.* 2004; 4(3):194-198.

[13] Younossi ZM, Stepanova M, Nader F, Younossi Z, Elsheikh E. Associations of chronic hepatitis C with metabolic and cardiac outcomes. *Aliment Pharmacol Ther.* 2013; 37(6):647–652.

[14] Negro F. Insulin resistance and HCV: will new knowledge modify clinical management? *J Hepatol.* 2006; 45(4):514-519.

[15] Vanni E, Bugianesi E, Saracco G. Treatment of type 2 diabetes mellitus by viral eradication in chronic hepatitis C: Myth or reality? *Dig Liver Dis.* 2016; 48(2):105–111.

[16] Huang YW, Yang SS, Fu SC, Wang TC, Hsu CK, Chen DS *et al.* Increased risk of cirrhosis and its decompensation in chronic hepatitis C patients with new-onset diabetes: a nationwide cohort study. *Hepatology*. 2014; 60(3):807-814.

[17] Elkrief L, Chouinard P, Bendersky N, Hajage D, Larroque B, Babany G *et al.* Diabetes mellitus is an independent prognostic factor

for major liver-related outcomes in patients with cirrhosis and chronic hepatitis C. *Hepatology*. 2014; 60(3):823-831.

[18] Gomide GPM, Melo CB, Santos VS, Salge VD, Camargo FC, Pereira GA *et al.* Epidemiological survey of hepatitis C in a region considered to have high prevalence: the state of Minas Gerais, Brazil. *Rev Soc Bras Med Trop.* 2019; 52:e20190202.

[19] Mane A, Sacks J, Sharma S, Singh H, Tejada-Strop A, Kamili S *et al.* Evaluation of five rapid diagnostic tests for detection of antibodies to hepatitis C virus (HCV): A step towards scale-up of HCV screening efforts in India. *PLoS One.* 2019; 14(1):e0210556.

[20] Wilson EB. Probable inference, the law of succession, and statistical inference. *J Am Stat Assoc.* 1927; 22(158):209–212.

[21. Huang JF, Chuang WL, Yu ML, Yu SH, Huang CF, Huang CI *et al.* Hepatitis C virus infection and metabolic syndrome—a community-based study in an endemic area of Taiwan. *Kaohsiung J Med Sci.* 2009; 25(6):299-305.

[22] Petta S, Amato M, Cabibi D, Camma C, Di Marco V, Giordano C *et al.* Visceral adiposity index is associated with histological findings and high viral load in patients with chronic hepatitis C due to genotype 1. *Hepatology*. 2010; 52(5):1543-1552.

[23] McCullough AJ. Obesity and its nurturing effect on hepatitis C. *Hepatology*. 2003; 38(3):557-559.

[24] Kaddai V, Negro F. Current understanding of insulin resistance in hepatitis C. *Expert Rev Gastroenterol Hepatol.* 2011; 5(4):503-516.

[25] Lapumnuaypol K, Pisarcik D, Putthapiban P, Sukhumthammarat W, Wijarnpreecha K, Thongprayoon C *et al.* Direct-acting antiviral agents decrease haemoglobin A1c level in patients with diabetes infected with hepatitis C virus: A systematic review & meta-analysis. *Indian J Med Res.* 2020; 152(6):562–567.

[26] Banerjee S, Saito K, Ait-Goughoulte M, Meyer K, Ray RB, Ray R. Hepatitis C virus core protein upregulates serine phosphorylation of insulin receptor substrate-1 and impairs the downstream akt/protein kinase B signaling pathway for insulin resistance. *J Virol.* 2008; 82(6):2606-2612.

[27] Bose SK, Ray R. Hepatitis C virus infection and insulin resistance. *World J Diabetes.* 2014; 5(1):52-58.

[28] Kuo YC, Chen IY, Chang SC, Wu SC, Hung TM, Lee PH *et al.* Hepatitis C virus NS 5A protein enhances gluconeogenesis through upregulation of Akt-/JNK-PEPCK signalling pathways. *Liver Int.* 2014; 34(9):1358-1368.

[29] Deng L, Shoji I, Ogawa W, Kaneda S, Soga T, Jiang DP *et al.* Hepatitis C virus infection promotes hepatic gluconeogenesis through an NS5A-mediated, FoxO1-dependent pathway. *J Virology.* 2011; 85(17):8556-8568.

[30] Dyal HK, Aguilar M, Bartos G, Holt EW, Bhuket T, Liu B *et al.* Diabetes Mellitus Increases Risk of Hepatocellular Carcinoma in Chronic Hepatitis C Virus Patients: A Systematic Review. *Dig Dis Sci.* 2016; 61(2):636-645.

[31] Nkontchou G, Cosson E, Aout M, Mahmoudi A, Bourcier V, Charif I *et al.* Impact of metformin on the prognosis of cirrhosis induced by viral hepatitis C in diabetic patients. *J Clin Endocrinol Metab.* 2011; 96(8):2601-2608.

[32] Romero-Gómez M, Viloria MDM, Andrade RJ, Salmerón J, Diago M, Fernández-Rodríguez CM *et al.* Insulin resistance impairs sustained response rate to peginterferon plus ribavirin in chronic hepatitis C patients. *Gastroenterology*. 2005; 128(3):636-641.

[33] Yu JW, Sun LJ, Zhao YH, Kang P, Yan BZ. The effect of metformin on the efficacy of antiviral therapy in patients with genotype 1 chronic hepatitis C and insulin resistance. *Int J Infect Dis.* 2012; 16(6):e436-e441.

[34] Elhelbawy M, Abdel-Razek W, Alsebaey A, Hashim M, Elshenawy H, Waked I. Insulin resistance does not impair response of chronic hepatitis C virus to direct-acting antivirals, and improves with the treatment. *Eur J Gastroenterol Hepatol.* 2019; 31(1):16-23.

[35] Benhammou JN, Moon AM, Pisegna JR, Su F, Vutien P, Moylan CA *et al.* Nonalcoholic fatty liver disease risk factors affect liver-related outcomes after direct-acting antiviral treatment for hepatitis C. *Dig Dis Sci.* 2021; 66(7):2394-2406.

[36] Tsai PC, Kuo HT, Hung CH, Tseng KC, Lai HC, Peng CY *et al.* Metformin reduces hepatocellular carcinoma incidence after successful antiviral therapy in patients with diabetes and chronic hepatitis C in Taiwan. *J Hepatol.* 2022; S0168-8278(22):03129-4.