**Name of Journal: World Journal of Gastroenterology**

**ESPS Manuscript NO: 18655**

**Manuscript Type: Topic Highlights**

**2015 Advances in Hepatitis C virus**

**hepatitis c virus markers in infection by hepatitis c virus: In the era of directly acting antivirals**

Coppola N *et al*. HCV markers in the DAA era

Nicola Coppola, Mariantonietta Pisaturo, Rosa Zampino, Margherita Macera, Caterina Sagnelli, Evangelista Sagnelli

**Nicola Coppola, Mariantonietta Pisaturo, Margherita Macera, Evangelista Sagnelli,** Department of Mental Health and Public Medicine, Section of Infectious Diseases, Second University of Naples, 80131 Naples, Italy

**Mariantonietta Pisaturo,** Division of Infectious Diseases, AORN Sant’Anna e San Sebastiano di Caserta, 81100 Caserta, Italy

**Rosa Zampino,** Internal Medicine and Hepatology, Second University of Naples, 80131 Naples, Italy

**Caterina Sagnelli,** Department of Clinical and Experimental Medicine and Surgery “F. Magrassi e A. Lanzara”, Second University of Naples, 80131 Naples, Italy

**Author contributions:** All authorssubstantial contributed to conception and design, acquisition of data, or analysis and interpretation of data; Coppola N contributed to drafting the article or revising it critically for important intellectual content; and final approval of the version to be published. He declares no conflict of interest in connection with this paper; Pisaturo M analysis of “HCV markers in Diagnosis of chronic HCV infection and in Assessment of the severity of chronic hepatitis C”; final approval of the version to be published. She declares no conflict of interest in connection with this paper; Zampino R analysis of “HCV-RNA kinetics and clearance as markers of remission”; (3) final approval of the version to be published. She declares no conflict of interest in connection with this paper; Macera Manalysis of “Assessment of factors associated with the response to anti-viral treatment”; final approval of the version to be published. She declares no conflict of interest in connection with this paper; Sagnelli C analysis of “HCV markers in acute hepatitis C”; final approval of the version to be published. She declares no conflict of interest in connection with this paper; Sagnelli E contributed to drafting the article and revising it critically for important intellectual content; final approval of the version to be published. He declares no conflict of interest in connection with this paper.

**Conflict-of-interest statement:** All the authors of the manuscript declare that they have no conflict of interest in connection with this paper.

**Open-Access:** This article is an open-access article which was selected by an in-house editor and fully peer-reviewed by external reviewers. It is distributed in accordance with the Creative Commons Attribution Non Commercial (CC BY-NC 4.0) license, which permits others to distribute, remix, adapt, build upon this work non-commercially, and license their derivative works on different terms, provided the original work is properly cited and the use is non-commercial. See: http://creativecommons.org/licenses/by-nc/4.0/

**Correspondence to: Nicola Coppola, MD, PhD, Assistant Professor** of Infectious Diseases, Department of Mental Health and Public Medicine,   
Section of Infectious Diseases, Second University of Naples, Via L. Armanni 5, 80131 Naples, Italy. nicola.coppola@unina2.it

**Telephone:** +39-815-666719

**Fax:** +39-815-666013

**Received:** April 26, 2015

**Peer-review started:** April 28, 2015

**First decision:** June 2, 2015

**Revised:** July 4, 2015

**Accepted:** September 2, 2015

**Article in press:**

**Published online:**

**Abstract**

About 130–170 million people are infected with the hepatitis C virus (HCV) worldwide and more than 350000 people die each year of HCV-related liver diseases.The combination of pegylated interferon (Peg-IFN) and ribavirin (RBV) was recommended as the treatment of choice for chronic hepatitis C for nearly a decade. In 2011 the directly acting antivirals (DAA) HCV NS3/4A protease inhibitors, telaprevir and boceprevir, were approved to treat HCV-genotype-1 infection, each in triple combination with Peg-IFN and RBV. These treatments allowed higher rates of SVR than the double Peg-IFN + RBV,but the low tolerability and high pill burden of these triple regimes were responsible for reduced adherence and early treatment discontinuation. The second and third wave DAAs introduced in 2013-2014 enhanced the efficacy and tolerability of anti-HCV treatment. Consequently, the traditional indicators for disease management and predictors of treatment response should be revised in light of these new therapeutic options. This review article will focus on the use of the markers of HCV infection and replication, of laboratory and instrumental data to define the stage of the disease and of predictors, if any, of response to therapy in the DAA era. The article is addressed particularly to physicians who have patients with hepatitis C in care in their everyday clinical practice.

**Key words:** Hepatitis C virus infection; Chronic hepatitis C; Hepatitis C virus replication; Directly acting antivirals; Staging

**© The Author(s) 2015.** Published by Baishideng Publishing Group Inc. All rights reserved.

**Core tip:** The second and third wave directly acting antivirals introduced in 2013-2014 enhanced the efficacy and tolerability of anti-hepatitis C virus (HCV) treatment. Consequently, the traditional indicators for disease management and predictors of treatment response should be revised in light of these new therapeutic options. This review article analyzes the modern use of the markers of HCV infection in: (1) the diagnosis of acute hepatitis C; (2) the diagnosis of chronic HCV infection; (3) the assessment of the severity of chronic hepatitis C; (4) the assessment of factors associated with response to anti-viral treatment; and (5) HCV-RNA kinetics and clearance as markers of remission.

Coppola N, Pisaturo M, Zampino R, Macera M, Sagnelli C, Sagnelli E. hepatitis c virus markers in infection by hepatitis c virus: In the era of directly acting antivirals. *World J Gastroenterol* 2015; In press

**INTRODUCTION**

The World Health Organization (WHO) estimates that 130–170 million people are infected with hepatitis C virus (HCV) worldwide and that more than 350000 people die each year of HCV-related liver diseases[1].Primary infection causes acute hepatitis C (AHC), which is asymptomatic in the majority of cases, but progresses to chronicity in about two-thirds of the cases and spontaneously remits in the remaining one-third[2-7]. Patients with chronic hepatitis C (CHC) frequently show increasing severity of liver fibrosis over time, which leads to liver cirrhosis in nearly a quarter of cases. Hepatocellular carcinoma (HCC) develops in HCV-related liver cirrhosis with a yearly rate around 3%[8-16].

The combination of pegylated interferon (Peg-IFN) and ribavirin (RBV) was recommended as the treatment of choice for CHC for nearly a decade[17-22]. This treatment, although poorly tolerated, provided a sustained clearance of circulating HCV (sustained viral response - SVR) in half of the patients with CHC due to HCV genotype 1 and in nearly 70% of those with HCV genotype 2 or 3. Several predictors of an unfavorable response to this treatment have been identified: viral ( HCV genotype 1 or 4 and a slow decline in serum HCV RNA during treatment), host factors (male sex, older age), a co-pathology (insulin resistance, diabetes), Afro-American ethnicity, severe fibrosis and/or steatosis, high body mass index and interleukin (IL) 28-B non-CC genotype[23]. In 2011 the directly acting antivirals (DAAs) NS3/4A protease inhibitors telaprevir and boceprevir were approved to treat HCV-genotype-1 infection, each in triple combination with Peg-IFN and RBV[24-27]. These treatments allowed higher rates of SVR than the double Peg-IFN+RBV[18,28-33],but the low tolerability and high pill burden of these triple regimes[34] were responsible for reduced adherence and early treatment discontinuation. The second and third wave DAAs introduced in 2013-2014 enhanced the efficacy and tolerability of anti-HCV treatment[35,36]. In fact the second and third generation DAAs afford SVR rates above 90%, regardless of HCV genotype, better tolerability and adherence used either in IFN-free regimens or in combination with interferon and ribavirin[37-40]. Consequently, the traditional indicators for disease management and predictors of treatment response should be revised in light of these new therapeutic options.

This review article will focus on the use of the markers of HCV infection and replication, of laboratory and instrumental data to define the stage of the disease and of predictors, if any, of response to therapy in the DAA era (Table 1). The article is addressed particularly to physicians who have patients with hepatitis C in care in their everyday clinical practice.

**HCV MARKERS IN AHC**

In its symptomatic form AHC is characterized by nausea, malaise, abdominal pain, jaundice and by the typical biochemical abnormalities[41-43]. The HCV etiology is usually established on the basis of a documented seroconversion to anti-HCV and/or HCV-RNA positivity during the natural course of the illness[19,39,44,45], but it is impossible to establish for patients first observed when seroconversion has already occurred. In addition, AHC remains frequently undiagnosed because asymptomatic in the majority of the cases[41]. Despite its typically mild clinical course, AHC progresses to chronicity in nearly 70% of the cases. Treatment with a 3- or 6-mo course of Peg-IFN has been shown to be effective in eradicating acute HCV infection in most cases, but to date no standardized treatment schedule has been defined. Delaying the treatment to 8-12 wk after the beginning of the illness allows the identification of cases that resolve spontaneously and does not compromise the efficacy. The use of IFN-free treatment regimens for AHC patients awaits assessment.

New strategies for an early diagnosis of AHC have been investigated[46,47].In addition, attempts have been made to distinguish this clinical form from an acute exacerbation of CHC, a clinical event characterized by a substantial increase in serum alanine aminotransferase (ALT) levels above the previous values in patients with CHC[6,7,48-50]. A combined use in serial serum samples of the rise in the anti-HCV titers and of the changes in antibody positivity in a recombinant immunoblot assay was found to be of some use by Lu *et al*[46]. Araujo *et al*[47], using a flow-cytometric microsphere immunoassay to measure anti-HCV IgG reactivity to the core NS3, NS4 and NS5 HCV recombinant proteins, correctly classified serum samples of AHC and CHC with a cross-validation of 90.8% for the AHC group and 97.2% for the CHC group.The role of anti-HCV IgG avidity and anti-HCV IgM titers to diagnose AHC have been extensively investigated[5,51-54]. A successful attempt to distinguish between AHC and a reactivation of CHC was made by Sagnelli *et al*[4] by carrying out a serial determination of anti-HCV IgM at two or three checking points within the third week from the disease onset. In another study by the same group, Coppola *et al*[5] successfully explored the distinction between AHC and a reactivation of CHC using the avidity of anti-HCV IgG to diagnose AHC. When both methods (anti-HCV IgM titer and anti-HCV IgG avidity) were applied to serial serum samples obtained during the illness, the distinction between the two clinical forms reached a level of sensitivity and specificity approaching 95%. The IgG avidity assay showed the highest efficacy during the initial two weeks of the illness and the IgM titer assay during the subsequent two weeks[53].

The diagnosis of AHC remains important even in the DAA era, since, although not yet assessed, it seems reasonable that all or nearly all patients with AHC can be cured with a short DAA-based regimen.

**HCV MARKERS IN CHC**

***Diagnosis of chronic HCV infection***

The diagnosis of CHC is based on the detection of serum anti-HCV and HCV RNA, elevated serum values of aminotransferases for at least six months and necroinflammation and fibrosis in liver tissue[8,19,39]. Screening to detect anti-HCV in serum is indicated for persons with a history of intravenous drug use, or sharing paraphernalia for intranasal drug use, acupuncture, body piercing or tattooing, persons who received blood, blood products or solid organs before 1992, hemodialysis patients, children born of HCV-infected mothers, patients with hepatitis B virus infection (HBV) and those with human immunodeficiency virus (HIV) infection[19,55-61]. Anti-HCV-positive subjects should be tested for serum HCV RNA, the confirmatory test of an ongoing HCV infection[62-65].

Anti-HCV can be detected by an enzyme-linked immunosorbent assay (ELISA), three generations of which have been developed since 1989. The first generation assay, incorporating the recombinant c100-3 epitope from the NS4 region was used until 1992, when it was replaced with the second generation assay incorporating the epitopes c22-3 and c33c from the HCV core and NS3 regions, respectively. The third generation assay used at present contains reconfigured core and NS3 antigens and a newly incorporated antigen from the NS5 region[62, 66,67], is more sensitive than the previous assays and has a diagnostic specificity of over 99%[62]. However, the third generation enzyme immunoassays can, albeit rarely, yield false-negative results in immunocompromised patients and in those undergoing hemodialysis[19].

More recently an assay for a rapid detection of anti-HCV in fingerstick capillary blood, venipuncture whole blood or saliva has been developed[68]. This assay, easy to perform and time-saving, has a good sensitivity and specificity and is particularly indicated for screening large populations.

The Recombinant Immunoblot Assay (RIBA), used in the past as a confirmatory assay of HCV infection, has not been recommended since 2013[69]. Subjects found to be anti-HCV-positive at screening should be tested for HCV RNA, the serum marker of HCV replication and current infection. Real-time PCR technologies can quantify HCV RNA during the exponential phase of amplification, with great sensitivity and a broad linear dynamic range (about 10 to 108 IU/mL). The majority of the commercial HCV RNA assays used by the clinical laboratories are based on the WHO international standard for HCV-RNA nucleic acid technology[70] and have an excellent specificity (98%-99%)[19].

Testing for HCV RNA should be considered for all anti-HCV-positive subjects and among the anti-HCV-negative for immunocompromised patients and for individuals exposed to HCV in the past 6 mo.

Concluding on this point, the high rate of SVR obtained by the DAA-based treatments is a further stimulus to screen all subjects exposed to HCV infection using a sensitive, specific, easy-to-perform and time-saving assay.

***Assessment of the severity of CHC***

The severity of CHC is variable among patients and over time in single patients. In most cases the disease shows a benign indolent course, but in some cases there is a rapid progression to liver cirrhosis, hepatocellular carcinoma and to an end-stage liver disease[8]. Liver cirrhosis is found in approximately 20% of patients with HCV-related chronic liver disease, associated in its advanced stages with life-threatening complications such as ascites, esophageal varice hemorrhage and liver failure. Hepatocellular carcinoma (HCC) occurs mostly in cirrhotic patients at a rate of 3%-5% per year[8].

The extent of liver necroinflammation and the degree of fibrosis in liver biopsy are considered reliable predictors of disease progression[71]. Several other investigators considered the stage of fibrosis detected in liver biopsy as a key point for the clinical management of CHC[72].

Although the liver histology is still considered the gold standard to assess the stage of liver fibrosis, because of the sides effects of liver biopsy[73-79] several surrogate non-invasive methods have been introduced. The measurement of liver stiffness by transient elastography offers an accredited method for the assessment of liver fibrosis[80]. This technique involves the use of a transducer on the end of an ultrasound probe that transmits 50 MHz pressure waves through the liver tissue. The velocity of the resulting “shear wave” is measured by ultrasound. The shear-wave velocity correlates with liver stiffness, thus providing an estimate of liver fibrosis[81,82]. Tsochatzis *et al*[82] performed a meta-analysis including 40 studies on numerous patients with chronic hepatitis of various etiologies (HBV, HCV, alcohol and other etiologic agents) and showed that transient elastography had a pooled sensitivity and specificity in diagnosing liver cirrhosis of 83% and 89%, respectively.

The ultrasound assay is another well-established non-invasive method to diagnose liver cirrhosis. The transition to cirrhosis is documented by the development of the characteristic coarse or nodular patterns in the liver parenchyma, hepatomegaly and caudate lobe hypertrophy[83].Ultrasound can also detect the development of portal hypertension by measuring the portal vein diameter, velocity of flow and flow reversal, ascites and splenomegaly[84], but the sensitivity of ultrasound in assessing liver fibrosis is low.

There is no single surrogate test able to predict reliably the progression to cirrhosis in each single patient. However, high serum ALT levels have been associated with a higher risk of fibrosis progression[85-87], which, instead, is an uncommon event in patients with persistently normal serum ALT[88-91].

Several other non-invasive surrogate biomarkers or a combination of biomarkers may be of some help in assessing liver fibrosis, such as platelet count, INR index, aspartate aminotransferase (AST) serum levels and albumin serum concentration. One well-known combination of biomarkers that has been extensively validated in CHC[92,93] and in non-alcoholic fatty liver disease (NAFLD)[94] is the so-called APRI test, an acronym for AST-platelet ratio index[95].Also of some interest is the Fibrotest (Fibrosure in the United States), which includes five biomarkers and 2 clinical parameters[96]: alpha-2 macroglobulin, haptoglobin, total bilirubin, apolipoprotien-A, -glutamyl transferase, age and gender. Using a patented formula, a numerical value from 0.0 to 1.0 is obtained, a score correlated with the METAVIR fibrosis score in chronic hepatitis of different etiologies[92,97,98]. Combining the ALT serum value with the panel of biomarkers included in the Fibrotest, a new surrogate method to measure liver fibrosis was obtained, named Actitest and validated to diagnose liver cirrhosis in CHC patients[99]. FIB4 is a biomarker panel using age, AST, ALT and platelet count[100] validated in HIV/HCV co-infected[101] and HCV-monoinfected patients[102].

Concluding on this point, the assessment of liver fibrosis is still essential, even in the DAA era, since it allows the high-cost DAA treatment to be applied on the basis of the severity of liver damage and of the presumed speed of disease progression.

***Assessment of factors associated with the response to anti-viral treatment***

In the DAA era, HCV genotypes and subtypes remain cornerstones in the management of chronic HCV infection, since the rate of response and the consequent duration of treatment differ for the various genotypes and subtypes[103]. In fact, considering patients with HCV genotype 1 or 4, whether therapy-naïve or -experienced, the combination of sofosbuvir and simeprevir (± ribavirin in non-responders to previous treatment) is the regimen of choice for subjects with METAVIR fibrosis scores 3 or 4, whereas for patients with fibrosis 0-2, optimal results were obtained with the combination of Peg-IFN, ribavirin and simeprevir[39,40]. Sofosbuvir plus ribavirin has been demonstrated to be an optimal combination for patients with HCV-genotype 2 or 3, whether therapy-naïve or -experienced, and the combination sofosbuvir plus daclatasvir for patients with HCV genotype 3[39,40]. In addition, in the simeprevir plus Peg-IFN-based regimen, it is essential to distinguish between patients with HCV sub-genotype 1a and 1b, since subtype 1a at times showed a Q80K substitution in the NS3 protease sequence, thus entailing a higher rate of treatment failure[39,40]. Currently, HCV genotyping can be performed by direct DNA sequencing by a bi-directional sequence where genotype and subtype characterization is determined by two fluorescently labeled DNA primers or by a commercial line probe assay[103].

In the DAA era, the detection of HCV viral load at baseline is now of no value in the treatment choice, since the anti-viral potency of these drugs controls even the highest level of HCV replication. The use of this test to monitor treated patients during the follow-up in order to detect possible reactivation seems good clinical practice.

The impact of staging in choosing a treatment schedule has decreased in proportion to the increase in the antiviral potency of the DAAs. In fact, the treatment regimens based on the third-wave DAAs achieve HCV eradication in almost all patients, regardless of the presence of liver cirrhosis[39,40].

The polymorphisms in the IL28B gene have been strongly associated with the spontaneous clearance of acute HCV infection and with the response to Peg-IFN and RBV combination therapy[104-107]. Their predictive value was more evident in difficult-to-treat HCV-genotype 1 and genotype 4 patients than in those with HCV-genotype 2 or 3 infection[108].The distribution of IL28B polymorphisms varies among different populations, accounting, at least in part, for the ethnic and racial differences in the response to Peg-IFN plus RBV[107],the CC genotype being a predictor of a favorable response. At present, IL-28 genotyping has no predictive role in the high-efficacy DAA-based regimens[39,40], but might be of some value in settings where a Peg-IFN-based regimen might still be used.

Hemolytic anemia is a common side effect of RBV-based therapy that, although reversible and dose-related, induced a RBV dose reduction or premature treatment withdrawal in more than 15% of the cases[109,110]. Fellay *et al[*111] identified two variants (rs1127354 and rs7270101) in the ITPA gene that were functionally responsible for ITPA deficiency and correlated with the risk of RBV-induced anemia in European and American populations. The rs1127354 variant was associated with protection against anemia in other investigations[112-115]. The SNP ITPA has never been associated with the treatment outcome[111-115], and in the DAA era it can be used only to evaluate the risk/benefit of adding ribavirin in some DAA-based regimens for patients with a lower rate of SVR, such as cirrhotics or previous non-responders.

Concluding on this point, DAA treatment eradicates HCV infection in nearly all treated patients, greatly reducing the clinical importance of markers previously used to predict the response to therapy. In fact, the HCV load and the degree of fibrosis do not predict the response to DAA therapy, and the polymorphisms in the IL28B gene may be useful only for patients with a METAVIR score F0-F2 treated with Peg-IFN, ribavirin and simeprevir, and the two SNPs in the ITPA gene only for those receiving a DAA plus ribavirin.

Instead, the determination of HCV genotype and subtype is of clinical value even in the DAA era, mandatory to choose the type and duration of therapy.

***HCV-RNA kinetics and clearance as markers of remission***

HCV-RNA clearance persisting 6 months after therapy (SVR) remains a marker of the eradication of chronic HCV infection also in the DAA era.

International treatment guidelines[116-118] identified some virological predictors of SVR to Peg-IFN + RBV treatment: a rapid virological response (RVR) *i.e.*, HCV-RNA clearance after 1 month of therapy, and an early virological response (EVR), *i.e.*, HCV-RNA clearance after 3 months of therapy. Subsequently, a very early predictor of SVR to Peg-IFN + RBV was suggested[119,120], *i.e.*, a decrease in the HCV load 2 days after the start of therapy. These predictors have been used to distinguish with good accuracy the patients with a good chance of achieving an SVR from those with a very low chance, who should discontinue treatment[121].

Compared to Peg-IFN+RBV treatments, the DAA-based therapies are more effective and better tolerated, but more expensive. The HCV-RNA kinetics during DAA treatment have been investigated in a limited number of patients and for short periods. Simeprevir given alone achieved a median HCV-RNA reduction of 3.9-log10 IU/mL over the first 3 days of treatment, independently of previous treatments and HCV genotype[122]. The administration of a single dose of 100-mg daclatasvir generated a decline in the HCV load of nearly 2-log10 in six hours and of 3.3-log10 in 24 h[123,124]. In addition, sofosbuvir obtained HCV-RNA clearance in 88%-94% of patients within the fourth week of treatment[125]. These data suggest that the determination of the HCV-RNA kinetics is of limited value in predicting the SVR in the DAA-based IFN-free treatments, since the majority of treated patients[126] achieve this favorable outcome. Several studies assessed serum HCV RNA at weeks 2 and 4 of treatment[37,127-129] and found that the persistence of HCV RNA in serum at these check-points is predictive of treatment failure[130].

In Peg-IFN+RBV regimens, the normalization of serum aminotransferases has been used as a parameter to evaluate the biochemical response[131], often associated with an SVR. In recent studies on DAA-based treatments, serum aminotransferases were no longer used to evaluate the response to treatment[37,127-129], since, for reasons unrelated to HCV replication (presence of liver steatosis or consumption of alcohol or other drugs known to be hepatotoxic), they may remain elevated even in SVR patients. In addition, an increase in the aminotransferase serum values may occur in some patients during treatment, an event to be monitored carefully because therapy discontinuation may be necessary[128]. At present, no other biochemical parameter has been associated with the SVR or with the need to discontinue therapy[132].

Concluding on this point, monitoring the HCV-RNA kinetics during DAA treatment seems good clinical practice and may help to identify early on the patients with a lesser chance of eradicating HCV chronic infection. Due to the ability of HCV to replicate not only in hepatocytes, but also in lymphocytes and possibly in other cell subsets, a reactivation of HCV replication in patients who had achieved an SVR cannot be excluded, and monitoring the HCV-RNA kinetics during the post-treatment follow-up can identify these cases.

**Conclusion**

The eradication of HCV infection in nearly all patients treated with the second- or third-wave DAAs and a more extensive use of these treatments in the near future will significantly contribute to curbing the spread of HCV infection and to reducing its related morbidity and mortality. At present, there is a strong stimulus for an early diagnosis of AHC, which can almost certainly be cured with a short-term DAA-based regimen, and for screening subjects with a history of previous exposure to HCV. Because of the high efficacy of the DAA treatments, the majority of the predictors of response to therapy will become obsolete. In particular, the degree of liver fibrosis does not predict the response to DAA therapy and its determination remains essential only to assess the priority for the high-cost DAA treatments based on disease severity and progression. Instead, the determination of HCV genotype and subtype remain essential in order to choose the type and duration of DAA treatment.

Monitoring the HCV-RNA kinetics during DAA treatment and post-treatment follow-up seems good inexpensive clinical practice, useful for an early identification of patients with a lesser chance of HCV eradication and of those prone to reactivation.

**References**

1 **Sagnelli E**, Santantonio T, Coppola N, Fasano M, Pisaturo M, Sagnelli C. Acute hepatitis C: clinical and laboratory diagnosis, course of the disease, treatment. *Infection* 2014; **42**: 601-610 [PMID: 24619833 DOI: 10.1007/s15010-014-0608-2]

2 **Sagnelli E**, Tonziello G, Pisaturo M, Sagnelli C, Coppola N. Clinical applications of antibody avidity and immunoglobulin M testing in acute HCV infection. *Antivir Ther* 2012; **17**: 1453-1458 [PMID: 23322703 DOI: 10.3851/IMP2471]

3 **Sagnelli E**, Coppola N, Marrocco C, Coviello G, Rossi G, Battaglia M, Sagnelli C, Messina V, Tonziello A, Scolastico C, Filippini P. Diagnosis of HCV related acute hepatitis by serial determination of IgM to HCV: a preliminary observation. *J Biol Regul Homeost Agents* 2003; **17**: 207-210 [PMID: 14518726]

4 **Sagnelli E**, Coppola N, Marrocco C, Coviello G, Battaglia M, Messina V, Rossi G, Sagnelli C, Scolastico C, Filippini P. Diagnosis of hepatitis C virus related acute hepatitis by serial determination of IgM anti-HCV titres. *J Hepatol* 2005; **42**: 646-651 [PMID: 15826712]

5 **Coppola N**, Pisapia R, Marrocco C, Martini S, Vatiero LM, Messina V, Tonziello G, Sagnelli C, Filippini P, Piccinino F, Sagnelli E. Anti-HCV IgG avidity index in acute hepatitis C. *J Clin Virol* 2007; **40**: 110-115 [PMID: 17720621]

6 **Coppola N**, Vatiero LM, Sagnelli E. HCV genotype 2 as a risk factor for reactivation of chronic HCV infection. *Gut* 2005; **54**: 1207 [PMID: 16009701]

7 **Sagnelli E**, Pisaturo M, Stanzione M, Messina V, Alessio L, Sagnelli C, Starace M, Pasquale G, Coppola N. Clinical presentation, outcome, and response to therapy among patients with acute exacerbation of chronic hepatitis C. *Clin Gastroenterol Hepatol* 2013; **11**: 1174-1180.e11 [PMID: 23591280]

8 **Seeff LB**. Natural history of chronic hepatitis C. *Hepatology* 2002; **36**: S35-S46 [PMID: 12407575]

9 **Aghemo A**, Colombo M. Hepatocellular carcinoma in chronic hepatitis C: from bench to bedside. *Semin Immunopathol* 2013; **35**: 111-120 [PMID: 23010890 DOI: 10.1007/s00281-012-0330-z]

10 **Dohmen K**, Kawano A, Takahashi K, Shigematsu H, Tanaka H, Haruno M, Yanagita K, Ichiki Y, Mori T, Hayashida K, Shimoda S, Ishibashi H, Nomura H. The incidence and risk factors for the development of hepatocellular carcinoma after peginterferon plus ribavirin therapy for chronic hepatitis C. *Hepatogastroenterology* 2013; **60**: 2034-2038 [PMID: 24719946]

11 **Harada N**, Hiramatsu N, Oze T, Morishita N, Yamada R, Hikita H, Miyazaki M, Yakushijin T, Miyagi T, Yoshida Y, Tatsumi T, Kanto T, Kasahara A, Oshita M, Mita E, Hagiwara H, Inui Y, Katayama K, Tamura S, Yoshihara H, Imai Y, Inoue A, Hayashi N, Takehara T. Risk factors for hepatocellular carcinoma in hepatitis C patients with normal alanine aminotransferase treated with pegylated interferon and ribavirin. *J Viral Hepat* 2014; **21**: 357-365 [PMID: 24716638 DOI: 10.1111/jvh.12151]

12 **Ishikawa T**. Strategy for improving survival and reducing recurrence of HCV-related hepatocellular carcinoma. *World J Gastroenterol* 2013; **19**: 6127-6130 [PMID: 24115808 DOI: 10.3748/wjg.v19.i37.6127]

13 **Kim MN**, Kim BK, Han KH. Hepatocellular carcinoma in patients with chronic hepatitis C virus infection in the Asia-Pacific region. *J Gastroenterol* 2013; **48**: 681-688 [PMID: 23463401 DOI: 10.1007/s00535-013-0770-9]

14 **Asia-Pacific Working Party on Prevention of Hepatocellular Carcinoma**. Prevention of hepatocellular carcinoma in the Asia-Pacific region: consensus statements. *J Gastroenterol Hepatol* 2010; **25**: 657-663 [PMID: 20492323 DOI: 10.1111/j.1440-1746.2009.06167.x]

15 **Kanda T**, Yokosuka O, Omata M. Hepatitis C virus and hepatocellular carcinoma. *Biology (Basel)* 2013; **2**: 304-316 [PMID: 24832662 DOI: 10.3390/biology2010304]

16 **Tomoda T**, Nouso K, Sakai A, Ouchida M, Kobayashi S, Miyahara K, Onishi H, Nakamura S, Yamamoto K, Shimizu K. Genetic risk of hepatocellular carcinoma in patients with hepatitis C virus: a case control study. *J Gastroenterol Hepatol* 2012; **27**: 797-804 [PMID: 22004425 DOI: 10.1111/j.1440-1746.2011.06948.x]

17 **European Association for the Study of the Liver**. EASL Clinical Practice Guidelines: management of hepatitis C virus infection. *J Hepatol* 2011; **55**: 245-264 [PMID: 21371579 DOI: 10.1016/j.jhep.2011.02.023]

18 **Coppola N**, Pisaturo M, Tonziello G, Sagnelli C, Sagnelli E, Angelillo IF. Efficacy of Pegylated interferon α-2a and α-2b in patients with genotype 1 chronic hepatitis C: a meta-analysis. *BMC Infect Dis* 2012; **12**: 357 [PMID: 23245594 DOI: 10.1186/1471-2334-12-357]

19 **Ghany MG**, Strader DB, Thomas DL, Seeff LB. Diagnosis, management, and treatment of hepatitis C: an update. *Hepatology* 2009; **49**: 1335-1374 [PMID: 19330875 DOI: 10.1002/hep.22759]

20 **Yee HS**, Chang MF, Pocha C, Lim J, Ross D, Morgan TR, Monto A. Update on the management and treatment of hepatitis C virus infection: recommendations from the Department of Veterans Affairs Hepatitis C Resource Center Program and the National Hepatitis C Program Office. *Am J Gastroenterol* 2012; **107**: 669-89; quiz 690 [PMID: 22525303 DOI: 10.1038/ajg.2012.48]

21 **European Association of the Study of the Liver**. 2011 European Association of the Study of the Liver hepatitis C virus clinical practice guidelines. *Liver Int* 2012; **32** Suppl 1: 2-8 [PMID: 22212565 DOI: 10.1111/j.1478-3231.2011.02703.x]

22 **Sagnelli E**, Pisaturo M, Martini S, Sagnelli C, Filippini P, Coppola N. Advances in the treatment of hepatitis B virus/hepatitis C virus coinfection. *Expert Opin Pharmacother* 2014; **15**: 1337-1349 [PMID: 24773464 DOI: 10.1517/14656566.2014.913571]

23 **Coppola N**, Pisaturo M, Sagnelli C, Sagnelli E, Angelillo IF. Peg-interferon plus ribavirin with or without boceprevir or telaprevir for HCV genotype 1: a meta-analysis on the role of response predictors. *PLoS One* 2014; **9**: e94542 [PMID: 24728219 DOI: 10.1371/journal.pone.0094542]

24 **Poordad F**, McCone J, Bacon BR, Bruno S, Manns MP, Sulkowski MS, Jacobson IM, Reddy KR, Goodman ZD, Boparai N, DiNubile MJ, Sniukiene V, Brass CA, Albrecht JK, Bronowicki JP. Boceprevir for untreated chronic HCV genotype 1 infection. *N Engl J Med* 2011; **364**: 1195-1206 [PMID: 21449783 DOI: 10.1056/NEJMoa1010494]

25 **Jacobson IM**, McHutchison JG, Dusheiko G, Di Bisceglie AM, Reddy KR, Bzowej NH, Marcellin P, Muir AJ, Ferenci P, Flisiak R, George J, Rizzetto M, Shouval D, Sola R, Terg RA, Yoshida EM, Adda N, Bengtsson L, Sankoh AJ, Kieffer TL, George S, Kauffman RS, Zeuzem S. Telaprevir for previously untreated chronic hepatitis C virus infection. *N Engl J Med* 2011; **364**: 2405-2416 [PMID: 21696307 DOI: 10.1056/NEJMoa1012912]

26 **Zeuzem S**, Andreone P, Pol S, Lawitz E, Diago M, Roberts S, Focaccia R, Younossi Z, Foster GR, Horban A, Ferenci P, Nevens F, Müllhaupt B, Pockros P, Terg R, Shouval D, van Hoek B, Weiland O, Van Heeswijk R, De Meyer S, Luo D, Boogaerts G, Polo R, Picchio G, Beumont M. Telaprevir for retreatment of HCV infection. *N Engl J Med* 2011; **364**: 2417-2428 [PMID: 21696308 DOI: 10.1056/NEJMoa1013086]

27 **Bacon BR**, Gordon SC, Lawitz E, Marcellin P, Vierling JM, Zeuzem S, Poordad F, Goodman ZD, Sings HL, Boparai N, Burroughs M, Brass CA, Albrecht JK, Esteban R. Boceprevir for previously treated chronic HCV genotype 1 infection. *N Engl J Med* 2011; **364**: 1207-1217 [PMID: 21449784 DOI: 10.1056/NEJMoa1009482]

28 **Dieterich DT**, Soriano V, Sherman K, Girard PM, Rockstroh J, Adiwijaya B, McCallister S, Adda N, Mahnke L, Sulkowski MS, on behalf of the Study 110 Team. Telaprevir in combination with pegylated interferon-a-2a RBV in HCV/HIV-co-infected patients: a 24-week treatment interim analysis. March 5-8-2012,[abstract #46]. In: Conference on Retroviruses and Other Opportunistic Infections, Seattle,WA, 2012

29 **Sulkowski MS**, Pol S, Cooper C, Fainboim H, Slim J, Rivero A, Laguno M, Thompson S, Wahl J, Greaves W. Boceprevir pegylated interferon ribavirin for the treatment of HCV/HIV-coinfected patients: end of treatment (week 48) interim results. March 5-8 2012,[Abstract #47]. In: Conference on Retroviruses and Other Opportunistic Infections, Seattle, WA, 2012

30 **Montes M**, Nelson M, Girard PM, Sasadeusz J, Horban A, Grinsztejn B, Zakharova N, Rivero A, Lathouwers E, Janssen K, Ouwerkerk-Mahadevan S, Witek J. Telaprevir combination therapy in HCV/HIV co-infected patients (INSIGHT study): sustained virologic response at 12 weeks final analysis. *J Int AIDS Soc* 2014; **17**: 19626 [PMID: 25394130 DOI: 10.7448/IAS.17.4.19626]

31 **Neukam K**, Munteanu D, Rivero A, Haberl A,MarquezM, Ingiliz P, et al. Boceprevir/Telaprevir-Based Therapy in HIV-Infection: Interim Analysis of a Multicenter Cohort 21st Conference on Retroviruses and Opportunistic Infections; Boston, MA March 3-6, 2014

32 **Gori A**, Doroana M, Chernova O, Rockstroh J, Banhegyi D, Bergin C, et al. Telaprevir Treatment of HIV/HCV G1 Patients With Severe Fibrosis: Efficacy Results To Week 16. 21th Conference on Retroviruses and Opportunistic Infections, Boston, USA March 2014

33 **Coppola N**, Martini S, Pisaturo M, Sagnelli C, Filippini P, Sagnelli E. Treatment of chronic hepatitis C in patients with HIV/HCV coinfection. *World J Virol* 2015; **4**: 1-12 [PMID: 25674512 DOI: 10.5501/wjv.v4.i1.1]

34 **Cotte L**, Barrail-Tran A, Vincent C, Valantin MA, Fournier I, Lacombe K, Chevaliez S, Aboulker JP, Taburet AM, Molina JM; ANRS HC26 study group. Telaprevir enhances ribavirin-induced anaemia through renal function impairment. *Antivir Ther* 2015; Epub ahead of print [PMID: 25560644 DOI: 10.3851/IMP2929]

35 **Koff RS**. Review article: the efficacy and safety of sofosbuvir, a novel, oral nucleotide NS5B polymerase inhibitor, in the treatment of chronic hepatitis C virus infection. *Aliment Pharmacol Ther* 2014; **39**: 478-487 [PMID: 24387618 DOI: 10.1111/apt.12601]

36 **Gentile I**, Borgia F, Buonomo AR, Castaldo G, Borgia G. A novel promising therapeutic option against hepatitis C virus: an oral nucleotide NS5B polymerase inhibitor sofosbuvir. *Curr Med Chem* 2013; **20**: 3733-3742 [PMID: 23848533]

37 **Lawitz E**, Mangia A, Wyles D, Rodriguez-Torres M, Hassanein T, Gordon SC, Schultz M, Davis MN, Kayali Z, Reddy KR, Jacobson IM, Kowdley KV, Nyberg L, Subramanian GM, Hyland RH, Arterburn S, Jiang D, McNally J, Brainard D, Symonds WT, McHutchison JG, Sheikh AM, Younossi Z, Gane EJ. Sofosbuvir for previously untreated chronic hepatitis C infection. *N Engl J Med* 2013; **368**: 1878-1887 [PMID: 23607594 DOI: 10.1056/NEJMoa1214853]

38 **Jacobson IM**, Gordon SC, Kowdley KV, Yoshida EM, Rodriguez-Torres M, Sulkowski MS, Shiffman ML, Lawitz E, Everson G, Bennett M, Schiff E, Al-Assi MT, Subramanian GM, An D, Lin M, McNally J, Brainard D, Symonds WT, McHutchison JG, Patel K, Feld J, Pianko S, Nelson DR. Sofosbuvir for hepatitis C genotype 2 or 3 in patients without treatment options. *N Engl J Med* 2013; **368**: 1867-1877 [PMID: 23607593 DOI: 10.1056/NEJMoa1214854]

39 **European Association for Study of Liver**. EASL Clinical Practice Guidelines: management of hepatitis C virus infection. *J Hepatol* 2014; **60**: 392-420 [PMID: 24331294 DOI: 10.1016/j.jhep.2013.11.003]

40 Documento di indirizzo dell’Associazione Italiana per lo studio del Fegato per l’uso razionale di antivirali diretti di seconda generazione nelle categorie di pazienti affetti da epatite C cronica ammesse alla rimborsabilità in Italia” AISF 24/02/2015. Available from: URL: http://www.webaisf.org

41 **Orland JR**, Wright TL, Cooper S. Acute hepatitis C. *Hepatology* 2001; **33**: 321-327 [PMID: 11172332]

42 **Blackard JT**, Shata MT, Shire NJ, Sherman KE. Acute hepatitis C virus infection: a chronic problem. *Hepatology* 2008; **47**: 321-331 [PMID: 18161707]

43 **Loomba R**, Rivera MM, McBurney R, Park Y, Haynes-Williams V, Rehermann B, Alter HJ, Herrine SK, Liang TJ, Hoofnagle JH, Heller T. The natural history of acute hepatitis C: clinical presentation, laboratory findings and treatment outcomes. *Aliment Pharmacol Ther* 2011; **33**: 559-565 [PMID: 21198704 DOI: 10.1111/j.1365-2036.2010.04549]

44 **Pawlotsky JM**. Use and interpretation of virological tests for hepatitis C. *Hepatology* 2002; **36**: S65-S73 [PMID: 12407578]

45 **Alter HJ**, Purcell RH, Shih JW, Melpolder JC, Houghton M, Choo QL, Kuo G. Detection of antibody to hepatitis C virus in prospectively followed transfusion recipients with acute and chronic non-A, non-B hepatitis. *N Engl J Med* 1989; **321**: 1494-1500 [PMID: 2509915]

46 **Lu SN**, Tung HD, Chen TM, Lee CM, Wang JH, Hung CH, Chen CH, Changchien CS. Is it possible to diagnose acute hepatitis C virus (HCV) infection by a rising anti-HCV titre rather than by seroconversion? *J Viral Hepat* 2004; **11**: 563-570 [PMID: 15500558]

47 **Araujo AC**, Astrakhantseva IV, Fields HA, Kamili S. Distinguishing acute from chronic hepatitis C virus (HCV) infection based on antibody reactivities to specific HCV structural and nonstructural proteins. *J Clin Microbiol* 2011; **49**: 54-57 [PMID: 21084519 DOI: 10.1128/JCM.01064-10]

48 **Sheen IS**, Liaw YF, Lin DY, Chu CM. Acute exacerbations in chronic hepatitis C: a clinicopathological and prognostic study. *J Hepatol* 1996; **24**: 525-531 [PMID: 8773906]

49 **Hiraga N**, Suzuki F, Akuta N, Suzuki Y, Sezaki H, Hosaka T, Someya T, Kobayashi M, Saitoh S, Arase Y, Ikeda K, Kobayashi M, Matsuda M, Watabiki S, Satoh J, Kumada H. Clinical and virological characteristics of untreated patients with chronic hepatitis C who develop serum alanine aminotransferase flare-up. *J Med Virol* 2005; **75**: 240-248 [PMID: 15602722]

50 **Rumi MG**, De Filippi F, La Vecchia C, Donato MF, Gallus S, Del Ninno E, Colombo M. Hepatitis C reactivation in patients with chronic infection with genotypes 1b and 2c: a retrospective cohort study of 206 untreated patients. *Gut* 2005; **54**: 402-406 [PMID: 15710990]

51 **Ward KN**, Dhaliwal W, Ashworth KL, Clutterbuck EJ, Teo CG. Measurement of antibody avidity for hepatitis C virus distinguishes primary antibody responses from passively acquired antibody. *J Med Virol* 1994; **43**: 367-372 [PMID: 7525865]

52 **Kanno A**, Kazuyama Y. Immunoglobulin G antibody avidity assay for serodiagnosis of hepatitis C virus infection. *J Med Virol* 2002; **68**: 229-233 [PMID: 12210412]

53 **Coppola N**, Pisapia R, Tonziello G, Masiello A, Martini S, Pisaturo M, Messina V, Sagnelli C, Macera M, Signoriello G, Sagnelli E. Improvement in the aetiological diagnosis of acute hepatitis C: a diagnostic protocol based on the anti-HCV-IgM titre and IgG Avidity Index. *J Clin Virol* 2009; **46**: 222-229 [PMID: 19758839 DOI: 10.1016/j.jcv.2009.08.009]

54 **Gaudy-Graffin C**, Lesage G, Kousignian I, Laperche S, Girault A, Dubois F, Goudeau A, Barin F. Use of an anti-hepatitis C virus (HCV) IgG avidity assay to identify recent HCV infection. *J Clin Microbiol* 2010; **48**: 3281-3287 [PMID: 20610669 DOI: 10.1128/JCM.00303-10]

55 **Armstrong GL**, Wasley A, Simard EP, McQuillan GM, Kuhnert WL, Alter MJ. The prevalence of hepatitis C virus infection in the United States, 1999 through 2002. *Ann Intern Med* 2006; **144**: 705-714 [PMID: 16702586]

56 **Wasley A**, Miller JT, Finelli L. Surveillance for acute viral hepatitis--United States, 2005. *MMWR Surveill Summ* 2007; **56**: 1-24 [PMID: 17363893]

57 **Alter MJ**, Seeff LB, Bacon BR, Thomas DL, Rigsby MO, Di Bisceglie AM. Testing for hepatitis C virus infection should be routine for persons at increased risk for infection. *Ann Intern Med* 2004; **141**: 715-717 [PMID: 15520428]

58 **Hosein SR**, Wilson DP. HIV, HCV, and drug use in men who have sex with men. *Lancet* 2013; **382**: 1095-1096 [PMID: 24075047 DOI: 10.1016/S0140-6736(13)62020-6]

59 **van de Laar TJ**, Matthews GV, Prins M, Danta M. Acute hepatitis C in HIV-infected men who have sex with men: an emerging sexually transmitted infection. *AIDS* 2010; **24**: 1799-1812 [PMID: 20601854 DOI: 10.1097/QAD.0b013e32833c11a5]

60 **Urbanus AT**, Van De Laar TJ, Geskus R, Vanhommerig JW, Van Rooijen MS, Schinkel J, Heijman T, Coutinho RA, Prins M. Trends in hepatitis C virus infections among MSM attending a sexually transmitted infection clinic; 1995-2010. *AIDS* 2014; **28**: 781-790 [PMID: 24832014 DOI: 10.1097/QAD.0000000000000126]

61 **Wiessing L**, Likatavicius G, Hedrich D, Guarita B, van de Laar MJ, Vicente J. Trends in HIV and hepatitis C virus infections among injecting drug users in Europe, 2005 to 2010. *Euro Surveill* 2011; **16**: [PMID: 22172300]

62 **Colin C**, Lanoir D, Touzet S, Meyaud-Kraemer L, Bailly F, Trepo C. Sensitivity and specificity of third-generation hepatitis C virus antibody detection assays: an analysis of the literature. *J Viral Hepat* 2001; **8**: 87-95 [PMID: 11264728]

63 **Dufour DR**, Talastas M, Fernandez MD, Harris B. Chemiluminescence assay improves specificity of hepatitis C antibody detection. *Clin Chem* 2003; **49**: 940-944 [PMID: 12765991]

64 **Alter MJ**, Kuhnert WL, Finelli L. Guidelines for laboratory testing and result reporting of antibody to hepatitis C virus. Centers for Disease Control and Prevention. *MMWR Recomm Rep* 2003; **52**: 1-13, 15; quiz CE1-4 [PMID: 12585742]

65 **Pawlotsky JM**, Lonjon I, Hezode C, Raynard B, Darthuy F, Remire J, Soussy CJ, Dhumeaux D. What strategy should be used for diagnosis of hepatitis C virus infection in clinical laboratories? *Hepatology* 1998; **27**: 1700-1702 [PMID: 9620345]

66 **Barrera JM**, Francis B, Ercilla G, Nelles M, Achord D, Darner J, Lee SR. Improved detection of anti-HCV in post-transfusion hepatitis by a third-generation ELISA. *Vox Sang* 1995; **68**: 15-18 [PMID: 7536987]

67 **Morishima C**, Gretch DR. Clinical use of hepatitis C virus tests for diagnosis and monitoring during therapy. *Clin Liver Dis* 1999; **3**: 717-740 [PMID: 11291247]

68 **Shivkumar S**, Peeling R, Jafari Y, Joseph L, Pant Pai N. Accuracy of rapid and point-of-care screening tests for hepatitis C: a systematic review and meta-analysis. *Ann Intern Med* 2012; **157**: 558-566 [PMID: 23070489 DOI: 10.7326/0003-4819-157-8-201210160-00006]

69 **Centers for Disease Control and Prevention (CDC)**. Testing for HCV infection: an update of guidance for clinicians and laboratorians. *MMWR Morb Mortal Wkly Rep* 2013; **62**: 362-365 [PMID: 23657112]

70 **Saldanha J**. Sensitivity of PCR assays for the determination of hepatitis A virus RNA in plasma pools. A collaborative study. *Vox Sang* 1999; **76**: 163-165 [PMID: 10341331]

71 **Yano M**, Kumada H, Kage M, Ikeda K, Shimamatsu K, Inoue O, Hashimoto E, Lefkowitch JH, Ludwig J, Okuda K. The long-term pathological evolution of chronic hepatitis C. *Hepatology* 1996; **23**: 1334-1340 [PMID: 8675148]

72 **Smith BD**, Morgan RL, Beckett GA, Falck-Ytter Y, Holtzman D, Teo CG, Jewett A, Baack B, Rein DB, Patel N, Alter M, Yartel A, Ward JW. Recommendations for the identification of chronic hepatitis C virus infection among persons born during 1945-1965. *MMWR Recomm Rep* 2012; **61**: 1-32 [PMID: 22895429]

73 **Sagnelli E**, Sagnelli C, Pisaturo MA, Coppola N, Pasquale G, Piccinino F; SIMIT group. Liver biopsy in chronic hepatitis C: the experience of 15 Italian wards of infectious diseases. *Infez Med* 2012; **20**: 31-36 [PMID: 22475658]

74 **Pasquale G**, Sagnelli E, Coppola N, Onofrio M, Scarano F, Scolastico C, Bellomo PF, Lettieri A, Mogavero AR, Caprio N, Sagnelli C, Piccinino F. [An attempt to improve classification of HCV-correlated chronic hepatitis]. *Infez Med* 2005; **13**: 16-22 [PMID: 15888977]

75 **Sagnelli E**, Pasquale G, Coppola N, Marrocco C, Scarano F, Imparato M, Sagnelli C, Scolastico C, Piccinino F. Liver histology in patients with HBsAg negative anti-HBc and anti-HCV positive chronic hepatitis. *J Med Virol* 2005; **75**: 222-226 [PMID: 15602732]

76 **Sagnelli E**, Pasquale G, Coppola N, Scarano F, Marrocco C, Scolastico C, Santantonio T, Gentile A, Piccinino F. Influence of chronic coinfection with hepatitis B and C virus on liver histology. *Infection* 2004; **32**: 144-148 [PMID: 15188073]

77 **Sagnelli E**, Coppola N, Scolastico C, Filippini P, Piccinino F. [Virological and clinical aspects of multiple hepatitis virus infections: preliminary data of an italian multicentre study] *Infez Med* 1999; **7**: 90-95 [PMID: 12759587]

78 **Pasquale G**, Sagnelli E, Coppola N, Scarano F, Scolastico C, Sagnelli C, Bellomo PF, Lettieri A, Filippini P, Piccinino F. Uselessness of liver biopsy in patients with hepatitis C virus chronic infection and persistently normal aminotransferase levels. *Infez Med* 2003; **11**: 11-17 [PMID: 12719665]

79 **Sagnelli E**, Coppola N, Scolastico C, Mogavero AR, Filippini P, Piccinino F. HCV genotype and "silent" HBV coinfection: two main risk factors for a more severe liver disease. *J Med Virol* 2001; **64**: 350-355 [PMID: 11424125]

80 **Sandrin L**, Fourquet B, Hasquenoph JM, Yon S, Fournier C, Mal F, Christidis C, Ziol M, Poulet B, Kazemi F, Beaugrand M, Palau R. Transient elastography: a new noninvasive method for assessment of hepatic fibrosis. *Ultrasound Med Biol* 2003; **29**: 1705-1713 [PMID: 14698338]

81 **Chon YE**, Choi EH, Song KJ, Park JY, Kim do Y, Han KH, Chon CY, Ahn SH, Kim SU. Performance of transient elastography for the staging of liver fibrosis in patients with chronic hepatitis B: a meta-analysis. *PLoS One* 2012; **7**: e44930 [PMID: 23049764 DOI: 10.1371/journal.pone.0044930]

82 **Tsochatzis EA**, Gurusamy KS, Ntaoula S, Cholongitas E, Davidson BR, Burroughs AK. Elastography for the diagnosis of severity of fibrosis in chronic liver disease: a meta-analysis of diagnostic accuracy. *J Hepatol* 2011; **54**: 650-659 [PMID: 21146892 DOI: 10.1016/j.jhep.2010.07.033]

83 **Di Lelio A**, Cestari C, Lomazzi A, Beretta L. Cirrhosis: diagnosis with sonographic study of the liver surface. *Radiology* 1989; **172**: 389-392 [PMID: 2526349 DOI: 10.1148/radiology]

84 **Aubé C**, Oberti F, Korali N, Namour MA, Loisel D, Tanguy JY, Valsesia E, Pilette C, Rousselet MC, Bedossa P, Rifflet H, Maïga MY, Penneau-Fontbonne D, Caron C, Calès P. Ultrasonographic diagnosis of hepatic fibrosis or cirrhosis. *J Hepatol* 1999; **30**: 472-478 [PMID: 10190731]

85 **Marcellin P**, Asselah T, Boyer N. Fibrosis and disease progression in hepatitis C. *Hepatology* 2002; **36**: S47-S56 [PMID: 12407576]

86 **Marcellin P**, Akre´mi R, Cazals D, Boyer N, Aupe´rin A, Vidaud D, Degott C. Genotype 1 is associated with a slower progression of fibrosis in un- treated patients with mild chronic hepatitis C. *J Hepatol* 2001; 34 (Suppl. 1):159

87 **Ghany MG**, Kleiner DE, Alter H, Doo E, Khokar F, Promrat K, Herion D, Park Y, Liang TJ, Hoofnagle JH. Progression of fibrosis in chronic hepatitis C. *Gastroenterology* 2003; **124**: 97-104 [PMID: 12512034]

88 **Martinot-Peignoux M**, Boyer N, Cazals-Hatem D, Pham BN, Gervais A, Le Breton V, Levy S, Degott C, Valla DC, Marcellin P. Prospective study on anti-hepatitis C virus-positive patients with persistently normal serum alanine transaminase with or without detectable serum hepatitis C virus RNA. *Hepatology* 2001; **34**: 1000-1005 [PMID: 11679971]

89 **Mathurin P**, Moussalli J, Cadranel JF, Thibault V, Charlotte F, Dumouchel P, Cazier A, Huraux JM, Devergie B, Vidaud M, Opolon P, Poynard T. Slow progression rate of fibrosis in hepatitis C virus patients with persistently normal alanine transaminase activity. *Hepatology* 1998; **27**: 868-872 [PMID: 9500720]

90 **Persico M**, Persico E, Suozzo R, Conte S, De Seta M, Coppola L, Palmentieri B, Sasso FC, Torella R. Natural history of hepatitis C virus carriers with persistently normal aminotransferase levels. *Gastroenterology* 2000; **118**: 760-764 [PMID: 10734027]

91 **Pasquale G**, Sagnelli E, Coppola N, Scarano F, Scolastico C, Bellomo PF, Lettieri A, Piccinino F. Is liver biopsy necessary for hepatitis C virus carriers with persistently normal aminotransferase levels? *Eur J Gastroenterol Hepatol* 2003; **15**: 831-833 [PMID: 12811317]

92 **Chou R**, Wasson N. Blood tests to diagnose fibrosis or cirrhosis in patients with chronic hepatitis C virus infection. *Ann Intern Med* 2013; **159**: 372 [PMID: 24026329 DOI: 10.7326/0003-4819-159-5-201309030-00021]

93 **Lin ZH**, Xin YN, Dong QJ, Wang Q, Jiang XJ, Zhan SH, Sun Y, Xuan SY. Performance of the aspartate aminotransferase-to-platelet ratio index for the staging of hepatitis C-related fibrosis: an updated meta-analysis. *Hepatology* 2011; **53**: 726-736 [PMID: 21319189 DOI: 10.1002/hep.24105]

94 **Festi D**, Schiumerini R, Marzi L, Di Biase AR, Mandolesi D, Montrone L, Scaioli E, Bonato G, Marchesini-Reggiani G, Colecchia A. Review article: the diagnosis of non-alcoholic fatty liver disease -- availability and accuracy of non-invasive methods. *Aliment Pharmacol Ther* 2013; **37**: 392-400 [PMID: 23278163 DOI: 10.1111/apt.12186]

95 **Wai CT**, Greenson JK, Fontana RJ, Kalbfleisch JD, Marrero JA, Conjeevaram HS, Lok AS. A simple noninvasive index can predict both significant fibrosis and cirrhosis in patients with chronic hepatitis C. *Hepatology* 2003; **38**: 518-526 [PMID: 12883497 DOI: 10.1053/jhep.2003.50346]

96 **Imbert-Bismut F**, Ratziu V, Pieroni L, Charlotte F, Benhamou Y, Poynard T. Biochemical markers of liver fibrosis in patients with hepatitis C virus infection: a prospective study. *Lancet* 2001; **357**: 1069-1075 [PMID: 11297957 DOI: 10.1016/S0140-6736(00)04258-6]

97 **Kim BK**, Kim SU, Kim HS, Park JY, Ahn SH, Chon CY, Cho IR, Joh DH, Park YN, Han KH, Kim do Y. Prospective validation of FibroTest in comparison with liver stiffness for predicting liver fibrosis in Asian subjects with chronic hepatitis B. *PLoS One* 2012; **7**: e35825 [PMID: 22536445 DOI: 10.1371/journal.pone.0035825]

98 **Poynard T**, Morra R, Halfon P, Castera L, Ratziu V, Imbert-Bismut F, Naveau S, Thabut D, Lebrec D, Zoulim F, Bourliere M, Cacoub P, Messous D, Munteanu M, de Ledinghen V. Meta-analyses of FibroTest diagnostic value in chronic liver disease. *BMC Gastroenterol* 2007; **7**: 40 [PMID: 17937811 DOI: 10.1186/1471-230X-7-40]

99 **Poynard T**, Munteanu M, Ngo Y, Castera L, Halfon P, Ratziu V, Imbert-Bismut F, Thabut D, Bourliere M, Cacoub P, Messous D, de Ledinghen V. ActiTest accuracy for the assessment of histological activity grades in patients with chronic hepatitis C, an overview using Obuchowski measure. *Gastroenterol Clin Biol* 2010; **34**: 388-396 [PMID: 20580175 DOI: 10.1016/j.gcb.2010.05.001]

100 **McPherson S**, Stewart SF, Henderson E, Burt AD, Day CP. Simple non-invasive fibrosis scoring systems can reliably exclude advanced fibrosis in patients with non-alcoholic fatty liver disease. *Gut* 2010; **59**: 1265-1269 [PMID: 20801772 DOI: 10.1136/gut.2010.216077]

101 **Sterling RK**, Lissen E, Clumeck N, Sola R, Correa MC, Montaner J, S Sulkowski M, Torriani FJ, Dieterich DT, Thomas DL, Messinger D, Nelson M. Development of a simple noninvasive index to predict significant fibrosis in patients with HIV/HCV coinfection. *Hepatology* 2006; **43**: 1317-1325 [PMID: 16729309 DOI: 10.1002/hep.21178]

102 **Vallet-Pichard A**, Mallet V, Nalpas B, Verkarre V, Nalpas A, Dhalluin-Venier V, Fontaine H, Pol S. FIB-4: an inexpensive and accurate marker of fibrosis in HCV infection. comparison with liver biopsy and fibrotest. *Hepatology* 2007; **46**: 32-36 [PMID: 17567829 DOI: 10.1002/hep.21669]

103 **Bouchardeau F**, Cantaloube JF, Chevaliez S, Portal C, Razer A, Lefrère JJ, Pawlotsky JM, De Micco P, Laperche S. Improvement of hepatitis C virus (HCV) genotype determination with the new version of the INNO-LiPA HCV assay. *J Clin Microbiol* 2007; **45**: 1140-1145 [PMID: 17251399]

104 **Coppola N**, Marrone A, Pisaturo M, Starace M, Signoriello G, Gentile I, Adinolfi LE, Sagnelli E, Zampino R. Role of interleukin 28-B in the spontaneous and treatment-related clearance of HCV infection in patients with chronic HBV/HCV dual infection. *Eur J Clin Microbiol Infect Dis* 2014; **33**: 559-567 [PMID: 24081499 DOI: 10.1007/s10096-013-1985-7]

105 **Coppola N**, Rosa Z, Cirillo G, Stanzione M, Macera M, Boemio A, Grandone A, Pisaturo M, Marrone A, Adinolfi LE, Sagnelli E, Miraglia Del Giudice E. TM6SF2 E167K variant is associated with severe steatosis in chronic hepatitis C, regardless of PNPLA3 polymorphism. *Liver Int* 2015; **35**: 1959-1963 [PMID: 25581573 DOI: 10.1111/liv.12781]

106 **Zampino R**, Coppola N, Cirillo G, Boemio A, Minichini C, Marrone A, Stanzione M, Starace M, Durante-Mangoni E, Sagnelli E, Restivo L, Salzillo G, Fascione MC, Nevola R, Del Giudice EM, Adinolfi LE. Insulin resistance and steatosis in HBV-HCV co-infected patients: Role of PNPLA3 polymorphisms and impact on liver fibrosis progression. *World J Hepatol* 2014; **6**: 677-684 [PMID: 25276284 DOI: 10.4254/wjh.v6.i9.677]

107 **Suppiah V**, Moldovan M, Ahlenstiel G, Berg T, Weltman M, Abate ML, Bassendine M, Spengler U, Dore GJ, Powell E, Riordan S, Sheridan D, Smedile A, Fragomeli V, Müller T, Bahlo M, Stewart GJ, Booth DR, George J. IL28B is associated with response to chronic hepatitis C interferon-alpha and ribavirin therapy. *Nat Genet* 2009; **41**: 1100-1104 [PMID: 19749758 DOI: 10.1038/ng.447]

108 **Thompson AJ**, Muir AJ, Sulkowski MS, Ge D, Fellay J, Shianna KV, Urban T, Afdhal NH, Jacobson IM, Esteban R, Poordad F, Lawitz EJ, McCone J, Shiffman ML, Galler GW, Lee WM, Reindollar R, King JW, Kwo PY, Ghalib RH, Freilich B, Nyberg LM, Zeuzem S, Poynard T, Vock DM, Pieper KS, Patel K, Tillmann HL, Noviello S, Koury K, Pedicone LD, Brass CA, Albrecht JK, Goldstein DB, McHutchison JG. Interleukin-28B polymorphism improves viral kinetics and is the strongest pretreatment predictor of sustained virologic response in genotype 1 hepatitis C virus. *Gastroenterology* 2010; **139**: 120-9.e18 [PMID: 20399780 DOI: 10.1053/j.gastro.2010.04.013]

109 **Fried MW**. Side effects of therapy of hepatitis C and their management. *Hepatology* 2002; **36**: S237-S244 [PMID: 12407599]

110 **Zampino R**, Alessio L, Marrone A, Stanzione M, Boemio A, Grandone A, Minichini C, Pisaturo M, Starace M, Adinolfi LE, Sagnelli E, Coppola N. Role of ITPA and IL28B variants in the management of chronic hepatitis C treatment. *Infez Med* 2015; **23**: 134-139 [PMID: 26110293]

111 **Fellay J**, Thompson AJ, Ge D, Gumbs CE, Urban TJ, Shianna KV, Little LD, Qiu P, Bertelsen AH, Watson M, Warner A, Muir AJ, Brass C, Albrecht J, Sulkowski M, McHutchison JG, Goldstein DB. ITPA gene variants protect against anaemia in patients treated for chronic hepatitis C. *Nature* 2010; **464**: 405-408 [PMID: 20173735 DOI: 10.1038/nature08825]

112 **Thompson AJ**, Santoro R, Piazzolla V, Clark PJ, Naggie S, Tillmann HL, Patel K, Muir AJ, Shianna KV, Mottola L, Petruzzellis D, Romano M, Sogari F, Facciorusso D, Goldstein DB, McHutchison JG, Mangia A. Inosine triphosphatase genetic variants are protective against anemia during antiviral therapy for HCV2/3 but do not decrease dose reductions of RBV or increase SVR. *Hepatology* 2011; **53**: 389-395 [PMID: 21274861 DOI: 10.1002/hep.24068]

113 **Kurosaki M**, Tanaka Y, Tanaka K, Suzuki Y, Hoshioka Y, Tamaki N, Kato T, Yasui Y, Hosokawa T, Ueda K, Tsuchiya K, Kuzuya T, Nakanishi H, Itakura J, Takahashi Y, Asahina Y, Matsuura K, Sugauchi F, Enomoto N, Nishida N, Tokunaga K, Mizokami M, Izumi N. Relationship between polymorphisms of the inosine triphosphatase gene and anaemia or outcome after treatment with pegylated interferon and ribavirin. *Antivir Ther* 2011; **16**: 685-694 [PMID: 21817190 DOI: 10.3851/IMP1796]

114 **Naggie S**, Rallon NI, Benito JM, Morello J, Rodriguez-Novoa S, Clark PJ, Thompson AJ, Shianna KV, Vispo E, McHutchison JG, Goldstein DB, Soriano V. Variants in the ITPA gene protect against ribavirin-induced hemolytic anemia in HIV/HCV-coinfected patients with all HCV genotypes. *J Infect Dis* 2012; **205**: 376-383 [PMID: 22158703 DOI: 10.1093/infdis/jir754]

115 **Fujino T**, Aoyagi Y, Takahashi M, Yada R, Yamamoto N, Ohishi Y, Nishiura A, Kohjima M, Yoshimoto T, Fukuizumi K, Nakashima M, Kato M, Kotoh K, Nakamuta M, Enjoji M. Association of ITPA polymorphism with outcomes of peginterferon-α plus ribavirin combination therapy. *World J Gastrointest Pharmacol Ther* 2013; **4**: 54-60 [PMID: 23919217 DOI: 10.4292/wjgpt.v4.i3.54]

116 **Prati D**, Gasbarrini A, Mazzotta F, for AISF, SIMAST and SIMIT. Practice guidelines for the treatment of hepatitis C: recommendations from an AISF/SIMIT/SIMAST Expert Opinion Meeting. *Dig Liver Dis* 2010; **42**: 81-91 [PMID: 19748329 DOI: 10.1016/j.dld.2009.08.001]

117 **Ghany MG**, Nelson DR, Strader DB, Thomas DL, Seeff LB. An update on treatment of genotype 1 chronic hepatitis C virus infection: 2011 practice guideline by the American Association for the Study of Liver Diseases. *Hepatology* 2011; **54**: 1433-1444 [PMID: 21898493 DOI: 10.1002/hep.24641]

118 **WHO Guidelines Approved by the Guidelines Review Committee.** Guidelines for the Screening, Care and Treatment of Persons with Hepatitis C Infection. Geneva: World Health Organization; 2014 Apr. [PMID: 25535634]

119 **Kronenberger B**, Herrmann E, Micol F, von Wagner M, Zeuzem S. Viral kinetics during antiviral therapy in patients with chronic hepatitis C and persistently normal ALT levels. *Hepatology* 2004; **40**: 1442-1449 [PMID: 15565603]

120 **Carlsson T**, Reichard O, Norkrans G, Bläckberg J, Sangfelt P, Wallmark E, Weiland O. Hepatitis C virus RNA kinetics during the initial 12 weeks treatment with pegylated interferon-alpha 2a and ribavirin according to virological response. *J Viral Hepat* 2005; **12**: 473-480 [PMID: 16108761]

121 **Durante-Mangoni E**, Zampino R, Portella G, Adinolfi LE, Utili R, Ruggiero G. Correlates and prognostic value of the first-phase hepatitis C virus RNA kinetics during treatment. *Clin Infect Dis* 2009; **49**: 498-506 [PMID: 19591593 DOI: 10.1086/600887]

122 **Reesink HW**, Fanning GC, Farha KA, Weegink C, Van Vliet A, Van 't Klooster G, Lenz O, Aharchi F, Mariën K, Van Remoortere P, de Kock H, Broeckaert F, Meyvisch P, Van Beirendonck E, Simmen K, Verloes R. Rapid HCV-RNA decline with once daily TMC435: a phase I study in healthy volunteers and hepatitis C patients. *Gastroenterology* 2010; **138**: 913-921 [PMID: 19852962 DOI: 10.1053/j.gastro.2009.10.033]

123 **Gao M**, Nettles RE, Belema M, Snyder LB, Nguyen VN, Fridell RA, Serrano-Wu MH, Langley DR, Sun JH, O'Boyle DR, Lemm JA, Wang C, Knipe JO, Chien C, Colonno RJ, Grasela DM, Meanwell NA, Hamann LG. Chemical genetics strategy identifies an HCV NS5A inhibitor with a potent clinical effect. *Nature* 2010; **465**: 96-100 [PMID: 20410884 DOI: 10.1038/nature08960]

124 **Guedj J**, Dahari H, Rong L, Sansone ND, Nettles RE, Cotler SJ, Layden TJ, Uprichard SL, Perelson AS. Modeling shows that the NS5A inhibitor daclatasvir has two modes of action and yields a shorter estimate of the hepatitis C virus half-life. *Proc Natl Acad Sci USA* 2013; **110**: 3991-3996 [PMID: 23431163 DOI: 10.1073/pnas.1203110110]

125 **Rodriguez-Torres M**, Lawitz E, Kowdley KV, Nelson DR, Dejesus E, McHutchison JG, Cornpropst MT, Mader M, Albanis E, Jiang D, Hebner CM, Symonds WT, Berrey MM, Lalezari J. Sofosbuvir (GS-7977) plus peginterferon/ribavirin in treatment-naïve patients with HCV genotype 1: a randomized, 28-day, dose-ranging trial. *J Hepatol* 2013; **58**: 663-668 [PMID: 23183528 DOI: 10.1016/j.jhep.2012.11.018]

126 **Sarrazin C**, Wedemeyer H, Cloherty G, Cohen DE, Chevaliez S, Herman C, Bernstein B, Pawlotsky JM. Importance of very early HCV RNA kinetics for prediction of treatment outcome of highly effective all oral direct acting antiviral combination therapy. *J Virol Methods* 2015; **214**: 29-32 [PMID: 25528998 DOI: 10.1016/j.jviromet.2014.11.027]

127 **Afdhal N**, Reddy KR, Nelson DR, Lawitz E, Gordon SC, Schiff E, Nahass R, Ghalib R, Gitlin N, Herring R, Lalezari J, Younes ZH, Pockros PJ, Di Bisceglie AM, Arora S, Subramanian GM, Zhu Y, Dvory-Sobol H, Yang JC, Pang PS, Symonds WT, McHutchison JG, Muir AJ, Sulkowski M, Kwo P. Ledipasvir and sofosbuvir for previously treated HCV genotype 1 infection. *N Engl J Med* 2014; **370**: 1483-1493 [PMID: 24725238 DOI: 10.1056/NEJMoa1316366]

128 **Zeuzem S**, Dusheiko GM, Salupere R, Mangia A, Flisiak R, Hyland RH, Illeperuma A, Svarovskaia E, Brainard DM, Symonds WT, Subramanian GM, McHutchison JG, Weiland O, Reesink HW, Ferenci P, Hézode C, Esteban R. Sofosbuvir and ribavirin in HCV genotypes 2 and 3. *N Engl J Med* 2014; **370**: 1993-2001 [PMID: 24795201 DOI: 10.1056/NEJMoa1316145]

129 **Fried MW**, Buti M, Dore GJ, Flisiak R, Ferenci P, Jacobson I, Marcellin P, Manns M, Nikitin I, Poordad F, Sherman M, Zeuzem S, Scott J, Gilles L, Lenz O, Peeters M, Sekar V, De Smedt G, Beumont-Mauviel M. Once-daily simeprevir (TMC435) with pegylated interferon and ribavirin in treatment-naïve genotype 1 hepatitis C: the randomized PILLAR study. *Hepatology* 2013; **58**: 1918-1929 [PMID: 23907700 DOI: 10.1002/hep.26641]

130 **Andreone P**, Colombo MG, Enejosa JV, Koksal I, Ferenci P, Maieron A, Müllhaupt B, Horsmans Y, Weiland O, Reesink HW, Rodrigues L, Hu YB, Podsadecki T, Bernstein B. ABT-450, ritonavir, ombitasvir, and dasabuvir achieves 97% and 100% sustained virologic response with or without ribavirin in treatment-experienced patients with HCV genotype 1b infection. *Gastroenterology* 2014; **147**: 359-365.e1 [PMID: 24818763 DOI: 10.1053/j.gastro.2014.04.045]

131 **Sezaki H**, Suzuki F, Kawamura Y, Yatsuji H, Hosaka T, Akuta N, Kobayashi M, Suzuki Y, Arase Y, Ikeda K, Kumada H. Evaluation of long-term biochemical responses to combination therapy of interferon plus ribavirin in those infected with hepatitis C virus genotype 1b and high baseline viral load. *Hepatol Res* 2007; **37**: 787-792 [PMID: 17573943]

132 **Everson GT**, Sims KD, Rodriguez-Torres M, Hézode C, Lawitz E, Bourlière M, Loustaud-Ratti V, Rustgi V, Schwartz H, Tatum H, Marcellin P, Pol S, Thuluvath PJ, Eley T, Wang X, Huang SP, McPhee F, Wind-Rotolo M, Chung E, Pasquinelli C, Grasela DM, Gardiner DF. Efficacy of an interferon- and ribavirin-free regimen of daclatasvir, asunaprevir, and BMS-791325 in treatment-naive patients with HCV genotype 1 infection. *Gastroenterology* 2014; **146**: 420-429 [PMID: 24184132 DOI: 10.1053/j.gastro.2013.10.057]

**P-Reviewer:** Cao GW **S-Editor:** Ma YJ **L-Editor:** **E-Editor:**

**Table 1 Viral and host markers useful for the management or treatment of chronic hepatitis C**

|  |  |  |
| --- | --- | --- |
|  | **Roles in the Peg-IFN era** | **Roles in the DAA era** |
| **Anti-HCV assay** | Diagnosis/screening | Diagnosis/screening |
| **HCV-RNA assay** | Diagnosis/ active replication  Pre-treatment predictor of response to antiviral treatments  Monitoring antiviral treatment  Assessment of response to treatment | Diagnosis/active replication  assessment of response to treatment |
| **HCV genotype** | Pre-treatment predictor of response to antiviral treatments | choosing the most appropriate DAA regimen |
| **HCV Q80K polymorphism** | none | selecting patients with HCV-genotype 1a for the simeprevir plus Peg-IFN regimen |
| **Markers of liver fibrosis (liver biopsy/non-invasive methods)** | staging of liver disease  pre-treatment predictors of response to antiviral treatments | staging of liver disease  selecting the patients with urgency for DAA-based treatment |
| **IL-28B polymorphism** | pre-treatment predictor of response to antiviral treatments | pre-treatment predictor of response only in Peg-IFN-based regimens |
| **ITPA polymorphism** | pre-treatment predictive factor of hemolytic anemia during ribavirin-based regimen | indicator of risk/benefit of using ribavirin in a DAA-based regimen |

Peg-IFN: Pegylated interferon; DAAs: Directly acting antivirals; HCV: Hepatitis C virus; IL: Interleukin.