HCV infection – more than 20 years of drug development

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Since the discovery of hepatitis C virus (HCV) in 1989 as a causing agent of acute and chronic hepatitis C (HC), therapy of the disease underwent substantial progress. Interferon was the golden standard for treating HC patients for more than two decades. This so-called conventional therapy resulted in sustained virological response[1] (SVR) only in 50 % of HCV genotype 1 patients and 80 % of HCV genotype 2 and 3 patients. This treatment had not only a significant impact to patient's quality of life but also was long-lasting (24-48 weeks). Nowadays pangenotypic regimens combining direct acting antiviral drugs (DAADs) are successful for 95 % of all HCV genotype patients and the therapy has shortened to 8 weeks.
Keywords: Hepatitis C, HCV polymorphism, direct acting antiviral drugs

Infekcia HCV – viac ako 20 rokov vývoja terapie
Od objavenia vírusu hepatitídy C (HCV) v roku 1989 ako pôvodcu akútnej a chronickej hepatitídy C (HC) terapia tohto ochorenia značne pokročila. Liečba interferónom predstavovala zlatý štandard terapie pre pacientov s HC viac ako dve dekády. Táto takzvaná konvenčná terapia vyvolala dlhodobú virologickú odpoveď (SVR) iba u 50 % pacientov s HCV genotypom 1 a u 80 % pacientov s genotypmi 2 a 3 s negatívnymi dôsledkami na kvalitu života pacienta a dĺžkou trvania 24 až 48 týždňov. Súčasná pangenotypická liečba, pozostávajúca z kombinácie takzvaných priamych antivirotík, je účinná až pre 95 % pacientov všetkých HCV genotypov s minimom nežiaducich účinkov.
Kľúčové slová: hepatitída C, polymorfizmus HCV, priamo účinkujúce antivirotiká

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WHO estimated that in 2015, 71 million persons were living with chronic HCV infection worldwide (global prevalence: 1%) and that 399 000 had died from cirrhosis or hepatocellular carcinoma. Aside from the burden of HCV infection secondary to liver-related sequelae, HCV causes an additional burden through comorbidities among persons with HCV infection, including depression(1), diabetes mellitus(2) and chronic renal disease(3). A proportion of these morbidities is directly attributable to HCV and is therefore referred to as extrahepatis manifestations. These manifestations are likely to be affected by treatment(4).

Mechanism of infection

HCV, which is transmitted parenterally, enters the liver via the bloodstream. In the liver sinusoids, the virus can pass the fenestrated endothelium and contact the basolateral surface of hepatocytes. HCV host cell entry is a complex multistep process that requires numerous host cell proteins like scavenger receptor class B type I (SCARB1), claudin-1, occludin and tetraspanin CD81. All four entry factors needs to be expressed on HCV-susceptible cells(6). SCARB1 and CD81 binds to glycoprotein E2 on the surface of virion. However, exact function of all four host cell receptors is not clear and its experimental evidence is lacking. After cell surface binding and coordinated interaction with entry factors, HCV is taken up by clathrin mediated endocytosis.

No vaccine

Despite 20 years of intensive research, a vaccine to prevent infection with the HCV remains elusive. HCV diversity is classified into seven genetically distinct genotypes (HCV 1–7) that differ by more than 30 % at nucleotide level, and into more than 50 subtypes that differ between 15 % and 25 % at nucleotide level within genotypes. A major barrier for the development of vaccines, broadly active antivirals, and assays, is the high genetic diversity of HCV and its potential to quickly adapt to different environments. HCV is under constant immunological pressure. Neutralizing antibody response of the host is targeting mainly the viral envelope proteins E1 and E2, but the virus manages to escape due to the large plasticity in the highly variable regions in these proteins. Effective targeting of conserved regions ***(Figure 1)*** in the genome may improve vaccine design. While vaccine design is still under experimental stage, development of DAADs has progressed into clinical practice.

HCV – basics, that helped to improve the therapy

Understanding of all aspects of HCV lifecycle helps to find therapy strategies targeted straightly to virus. An example of this approach is discovery of miRNA-122 (miR-122) involvement in HCV infection and subsequent development of drug called miravirsen. HCV relies on the host miR-122 in a unique way as miR-122 binds to the 5 '-non-translated region of the

[1]Sustained Virological Response = absence of anti-HCV and HCV RNA in the blood 12 weeks after treatment

HCV genome, which results in increased stability of the latter and thus increased replication. Miravirsen represents the first RNA-interference-based drug currently undergoing phase II of clinical trials[7]. Silencing of expression miR-122 by Miravirsen leading to HCV genome degradation may be a solution for HC patients resistant to current pangenotypic regimens.

The story of increasing effectiveness

15 years ago the standard of care of adolescents and children infected with HCV was dual therapy with pegylated-interferon and ribavirin for 24 weeks for genotypes 2 and 3, and 48 weeks for genotypes 1 and 4. This combination resulted in an SVR rate of around 52% in children infected with HCV genotypes 1 and 4, and 89% in those infected with HCV genotypes 2 and 3, but was associated with significant side-effects[8].

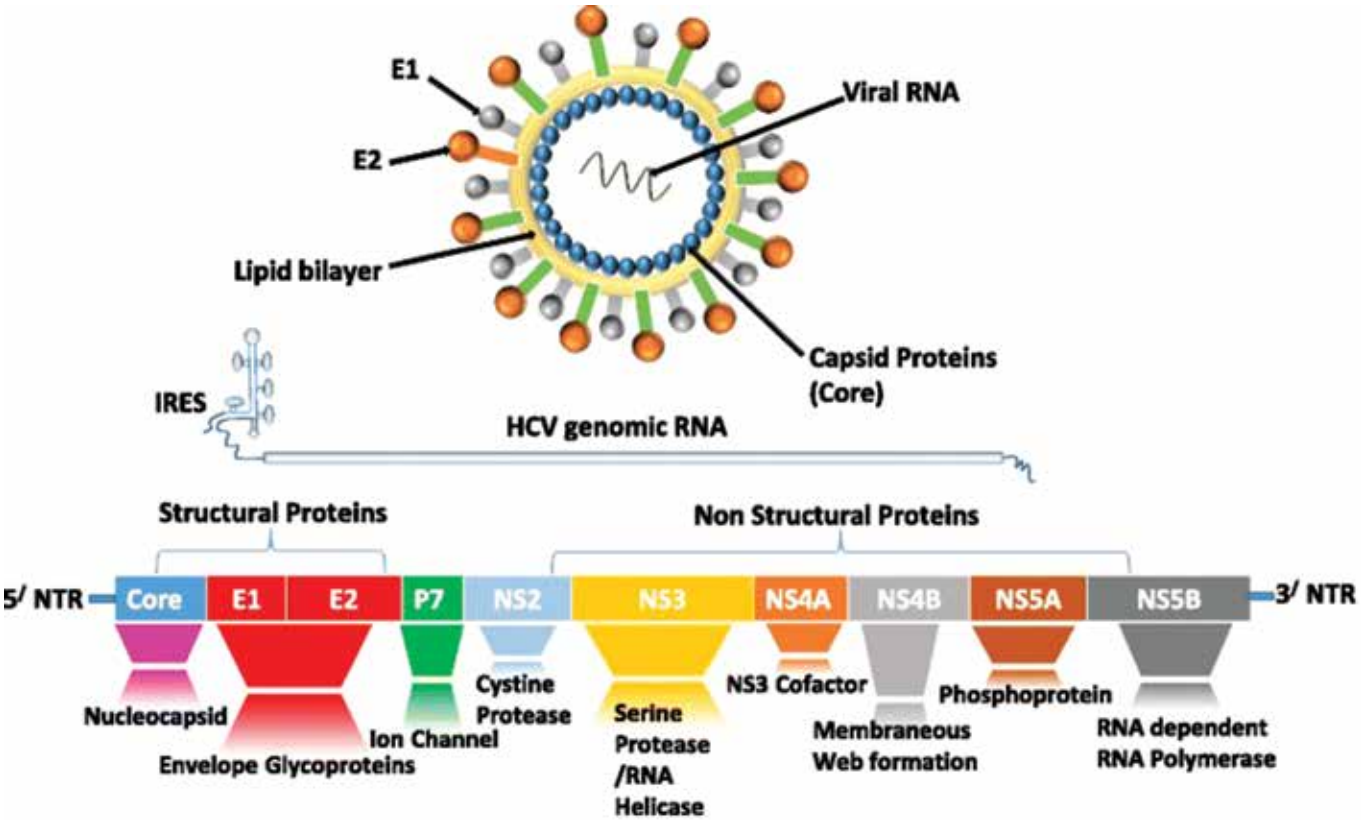
In 2011, two inhibitors against NS3 and NS4A viral proteases have been included in standard therapy in some regions, including Slovakia, which are mainly focused on the treatment of genotype 1. The triple combination of PEG-interferon, ribavirin and protease inhibitors improved virological response in several cohorts of patients from 50 % to 70 %[9,10].

However, this approach has limited efficacy for a particular group of patients (with liver cirrhosis, liver transplant patients, patients who are primarily unresponsive to this type of treatment, and hemodialysis patients). An important aspect of the development of the NS3 inhibitor is resolution of the crystalline structure of this protein alone and in conjunction with the cofactor, which facilitated the design of the drug. There are currently several drugs that inhibit NS3 protease ***(Table 1)***. All DAADs against NS3 target the active

Table 1. *Direct-acting antivirals (DAAs) divided according to class*[4]

NS3/4 A (protease)	NS5A inhibitors	NS5B polymerase inhibitor (nucleotide analogue)	NS5B polymerase inhibitor (non-nucleoside analogue)
Glecaprevir	Daclatasvir	Sufosbuvir	Dasabuvir
Voxilaprevir	Velpatasvir		
Grazoprevir	Ledipasvir		
Paretaprevir	Ombatasvir		
Simeprevir	Pibrentasvir		
	Elbasvir		

Figure 1. *The HCV genome consists of a 9.6 kilobase ORF flanked with 5' and 3'untranslated regions*[5]*. IRES-mediated translation of the ORF produces polyprotein that is processed by cellular and viral proteases into ten viral proteins: C, E1 and E2 structural proteins, and P7, NS2, NS3, NS4A, NS4B, NS5A and NS5B non-structural proteins. Abbreviations: C: Core protein, IRES: Internal ribosome entry site, ORF: Open reading frame, NS: Non-structural protein*



site of the protease, but it has been identified substitutions at this site that cause resistance[11].

NS5A is another virus protein candidate to inhibit. This multifunctional protein is an essential component of the viral replication complex, involved in the regulation of replication and the composition of the viral particle. These drugs ***(Table 1)*** have become a central part of the current combined DAAD therapy but have a relatively low barrier to the development of viral resistance[11].

NS5B is an HCV protein that functions as a RNA-dependent RNA-polymerase and is therefore a target for the inhibition of viral replication. The basic research of the NS5B polymerase has enabled the generation of efficient nucleoside analogues against HCV virions with various pangenomes. The great advantage is that it is targeting a highly conserved enzyme, and therefore, in this case, the barrier to the development of resistance is high. This category includes the drug sofosbuvir, which is used in the treatment of all genotypes of HCV. The combination of sofosbuvirus with the NS5A inhibitor velpatasvirus is effective against all six genotypes of HCV[12,13]. As of May 2018, the FDA or the EMA had approved 13 direct-acting antivirals from four classes ***(Table 1)***. Therefore, nowadays HCV therapy, including Slovak medical practice, combines DAAs according to a specific genotype at the highest efficacy level according to the recommendation table.

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REFERENCES

1. Adinolfi LE, Nevola R, Rinaldi L, et al. Chronic Hepatitis C Virus Infection and Depression. Clin Liver Dis 2017; 21(3): 517-534. doi:10.1016/j.cld.2017.03.007

2. Serfaty L. Metabolic Manifestations of Hepatitis C Virus: Diabetes Mellitus, Dyslipidemia. Clin Liver Dis 2017; 21(3): 475-486. doi:10.1016/j.cld.2017. 03. 004

3. Long JD, Rutledge SM, Sise ME. Autoimmune Kidney Diseases Associated with Chronic Viral Infections. Rheum Dis Clin N Am 2018; 44(4): 675-698. doi:10.1016/j.rdc.2018.06.006

4. World Health Organization. WHO | Guidelines for the care and treatment of persons diagnosed with chronic hepatitis C virus infection. WHO. http://www.who.int/hepatitis/publications/hepatitis-c-guidelines-2018/en/. Published 2018. Accessed December 3, 2018.

5. Elberry MH, Darwish NHE, Mousa SA. Hepatitis C virus management: potential impact of nanotechnology. Virol J 2017; 14: 88. doi:10.1186/s12985-017-0753-1

6. Ploss A, Evans MJ, Gaysinskaya VA, et al. Human occludin is a hepatitis C virus entry factor required for infection of mouse cells. Nature 2009; 457(7231): 882-886. doi:10.1038/nature07684

7. Titze-de-Almeida R, David C, Titze-de-Almeida SS. The Race of 10 Synthetic RNAi-Based Drugs to the Pharmaceutical Market. Pharm Res 2017; 34(7): 1339-1363. doi:10.1007/s11095-017-2134-2

8. Mack CL, Gonzalez-Peralta RP, Gupta N, et al. NASPGHAN practice guidelines: Diagnosis and management of hepatitis C infection in infants, children, and adolescents. J Pediatr Gastroenterol Nutr 2012; 54(6): 838-855. doi:10.1097/MPG.0b013e318258328d

9. Jacobson IM, McHutchison JG, Dusheiko G, et al. Telaprevir for previously untreated chronic hepatitis C virus infection. N Engl J Med 2011; 364(25): 2405-2416. doi:10.1056/NEJMoa1012912

10. Kwo PY, Lawitz EJ, McCone J, et al. Efficacy of boceprevir, an NS3 protease inhibitor, in combination with peginterferon alfa-2 b and ribavirin in treatment-naive patients with genotype 1 hepatitis C infection (SPRINT-1): an open-label, randomised, multicentre phase 2 trial. Lancet Lond Engl 2010; 376(9742): 705-716. doi:10.1016/S0140-6736(10)60934-8

11. Pawlotsky J-M. Hepatitis C Virus Resistance to Direct-Acting Antiviral Drugs in Interferon-Free Regimens. Gastroenterology 2016; 151(1): 70-86. doi:10.1053/j.gastro.2016. 04. 003

12. Feld JJ, Jacobson IM, Hézode C, et al. Sofosbuvir and Velpatasvir for HCV Genotype 1, 2, 4, 5, and 6 Infection. N Engl J Med 2015; 373(27): 2599-2607. doi:10.1056/NEJMoa1512610

13. Foster GR, Afdhal N, Roberts SK, et al. Sofosbuvir and Velpatasvir for HCV Genotype 2 and 3 Infection. N Engl J Med 2015; 373(27): 2608-2617. doi:10.1056/NEJMoa1512612

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