New treatment options of chronic hepatitis c

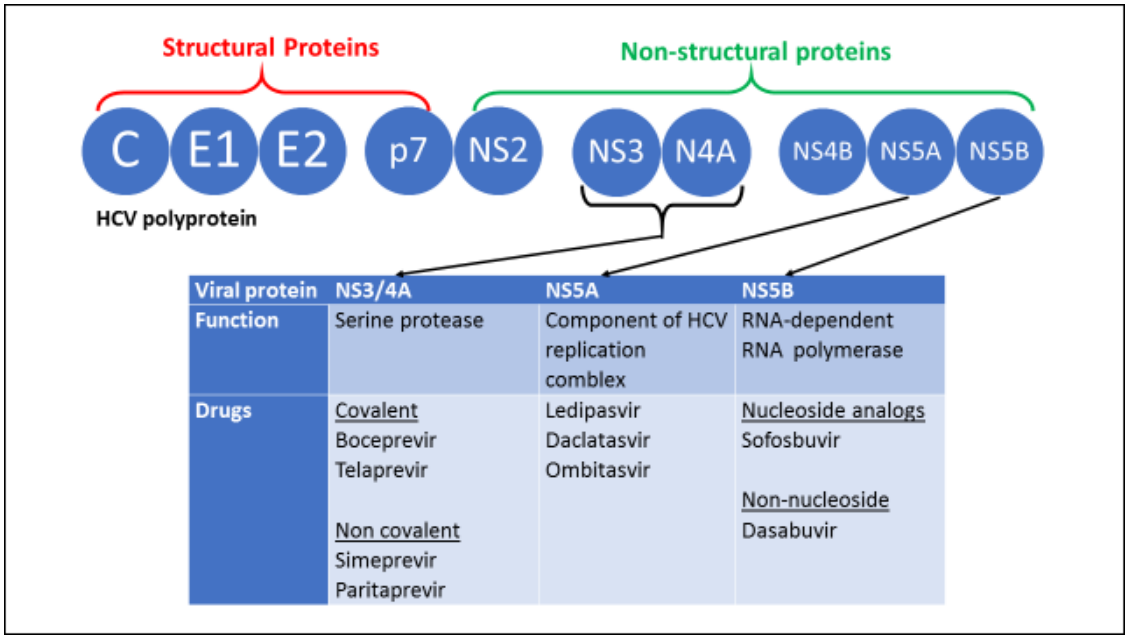
1. Abstract

2. Introduction

Hepatitis C virus infection (HCV), is a significant public health issue, with a socioeconomic and humanistic aspect, by affecting an estimated of 185 million people worldwide (2.1). Most of acute infected patients (80% (2.1)) establish chronic HCV, which will progress to cirrhosis (20% of patients) that leads to liver transplantation. Besides cirrhosis, hepatocellular carcinoma and liver failure, are also complication of the virus and can lead to death. Specifically 350 000 deaths are reported per year in US (M).

Hepatitis C virus, discovered in 1989, is an enveloped, single – stranded RNA, belonging to the genus Hepacivirus, a member of the Flaviviridae family. HCV shows extreme genetic diversity with 6 major genotypes and 52 subtypes. Genotype 1 appears to be the most dominant in the US, representing 70% of all cases (50). The RNA strand is translated into a single polyprotein of 3000 amino acids that is cleaved, by proteases, into 4 structural (C, E1, E2, p7) and 6 nonstructural (NS2, NS3, NS4A, NS4B, NS5A, NS5B) proteins (J) (Figure 1).

The developed new drugs against HCV are targeting some of the nonstructural proteins, which are NS3/4A, NS5A and NS5B. NS3 is a multifunctional protein which has a serine protease domain at N-terminal end and a helicase domain at C-terminal end. NS3 forms a complex with a co-factor NS4A. NS4A is responsible for folding the protein and positioning the catalytic triad of NS3 over its substrate. NS5A is also a multifunctional protein that has no enzymatic activity. It is a phosphoprotein which is required for RNA replication, membranous web formation and viral particle formation. Finally, NS5B is an RNA-depended polymerase that replicates the HCV genome. It has three domains arranged in a “palm, thumb and finger” orientation.



Viral protein	NS3/4A	NS5A	NS5B
Function	Serine protease	Component of HCV replication comblex	RNA-dependent RNA polymerase
Drugs	Covalent Boceprevir Telaprevir Non covalent Simeprevir Paritaprevir	Ledipasvir Daclatasvir Ombitasvir	Nucleoside analogs Sofosbuvir Non-nucleoside Dasabuvir

Figure 1: The HCV polyprotein and corresponding direct acting antivirals approved for clinical use by FDA

Since the discovery of HCV in 1989, strategies to eradicate HCV have evolved rapidly. The first treatment available was a-interferons (INF), which are immunomodulatory agents, administered as subcutaneous injections. Afterwards, ribavirin (RBV), a nucleoside inhibitor which interferes with RNA metabolism necessary for viral replication, was added to the arsenal of treatment for the improvement of cure rates. At the start of the millennium, FDA approved pegylated interferon and weight based RBV. Further research was made due to the various side effects, low SVR rates and the long term treatment. The revolution occurred in 2011, when telaprevir and boceprevir were licensed for use for HCV genotype 1 infection, in combination with pegylated interferon and RBV. These drugs are the first wave of direct acting antivirals (DAAs). DAAs are defined as "molecules that target specific nonstructural proteins within the HCV, which results in disruption of viral replication and infection" (48). Since 2013, FDA approved for clinical use 2 new drugs, in combination with PEG interferon and RBV, as well as 5 interferon - free combination therapies (Figure 2).

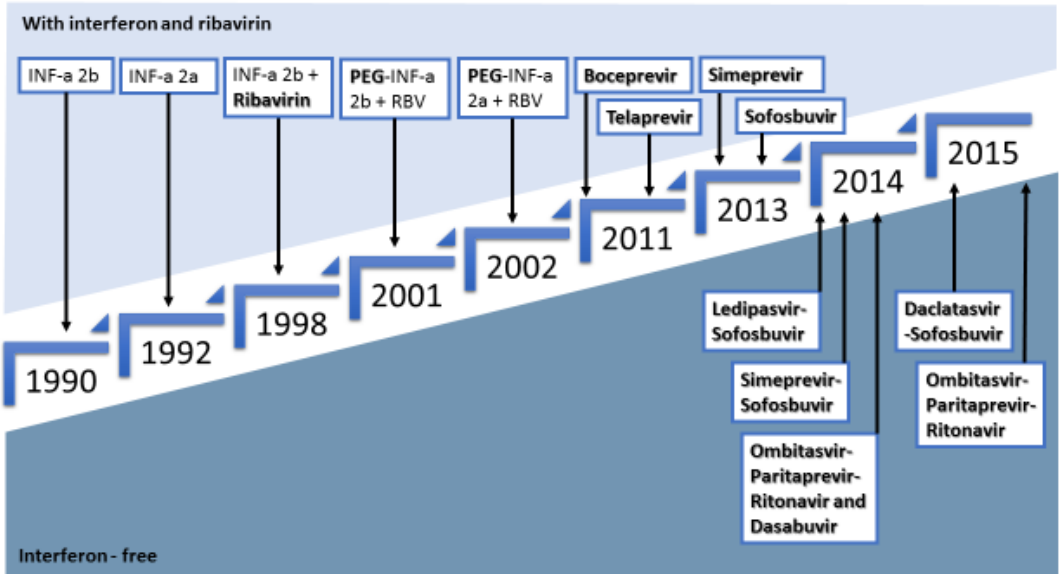


Figure 2: Advances in Hepatitis C treatment over the years. INF= interferons; RBV= ribavirin; PEG-INF= PEGylated interferons.

The success of the drugs mentioned above is defined by the Sustained Virological Response (SVR), which is the absence of detectable levels of plasma HCV RNA, 12 weeks after the completion of therapy (14).

3. Discussion

3.1. NS3/4A

NS3/4A protease inhibitors act by binding into the catalytic site of the enzyme, and preventing cleavage of the nonstructural proteins. Thus, disrupts processing of the HCV polypeptide. Based on the active site of binding group, these inhibitors are divided into two generations. First generation inhibitors, telaprevir and boceprevir, contain an a-ketoamide, which forms a covalent reversible interaction with the active site serine of the HCV NS3/4A protease catalytic triad. Second generation inhibitors, such as simeprevir, forms non-covalent ionic interactions with residues of the HCV NS3/4A protease catalytic triad. NS3/4A protease inhibitors have low barrier to resistance and are not used as monotherapy. Furthermore, they are non pangenotypic as their active site is not highly conserved across HCV genotypes. As a result, they are mainly used in genotype 1. Finally, first generation PIs are accompanied by serious adverse events in contrast to second generation PIs.

3.1.1. Boceprevir and telaprevir

Boceprevir (Victrelis) and telaprevir (Incivek) were approved in May 2011 by FDA, beginning the era of DAAs. These drugs are used in conjunction with PEG-interferon and ribavirin improving the rates of SVR approximately by 70% in genotype-1 naïve patients. A major difference between these drugs is that a lead-in phase is not required with telaprevir triple therapy, because of its stronger anti-HCV potency. Resistance- associated viral variants with substitutions located in the catalytic site of the NS3 protease, have been described after telaprevir and boceprevir treatment (M). Despite the high SVR rates, these first generation protease inhibitors have substantial side effects. The most common side effects of boceprevir triple therapy are anemia and dysgeusia, although neutropenia and thromvocytopenia are also reported. As far as telaprevir triple therapy is concerned, anemia, pruritis, rash, fatique, pyrexia, nausea diarrhea, hemorrhoids, alopecia, insomnia and anorectal discomfort are side effects that can occur (M). Another considerable drawback of these first generation PIs is their unfavorable pharmacokinetic profile, which necessitates doses on a thrice-a-day basis (c). Finally, significant drug-drug interactions prompted the FDA to impose warning for the use of these PIs, since CYP3A4 inducers may significantly decrease plasma concentrations of these drugs. Consequently, treatment with either of these agents is no longer recommended.

3.1.2. Simeprevir

In 2013, simeprevir (Olysio), a second generation protease inhibitor, was approved in combination with PEG-interferon and ribavirin, increasing SVR rates up to 85% in treatment naïve genotype 1 patients (C). It has also efficacy in the other genotypes. Advantages of simeprevir, over the earlier PIs, include once daily dosing, short treatment duration (12-24 weeks), better safety profile, better tolerance and a lower rate of adverse events (lack of anemia) (1.17,1.18). Side effects associated with simeprevir are fatigue and headache, hyperbilirubinemia, photosensitivity, rash, pruritus and nausea. As simeprevir is a substrate of CYP3A4 is affected by both CYP3A4 inducers and inhibitors. It is worth mentioning that Q80K polymorphism alters the efficacy of simeprevir and therefore patients with HCV genotype-1a should be screened for this NS3 polymorphism when starting simeprevir.

Newer PIs include paritaprevir, asunaprevir, grazoprevir, faldaprevir, ABT-450, danoprevir, vaniprevir, MK-5172, sovaprevir and ventroprevir. ΑΠΟΡΙΑ ΓΙΑ ΤΟΝ ΔΙΑΧΩΡΙΣΜΟ ΣΕ WAVES.

These drugs have improved pharmacological profile and therefore they have improved dosing schedule, tolerability, less frequent and less severe side effects as well as fewer drug-drug interactions and activity against other genotypes. However, some of them have a low barrier to resistance.

3.2. NS5A

NS5A protein inhibitor has pleiotropic roles in establishing the replication complex in viral assembling and in inhibiting apoptosis. However, due to the lack of enzymatic activity the mechanism by which the NS5A regulates the replication is unclear. NS5A inhibitors exhibit pangenotypic activity but the effectiveness against genotypes varies. In addition, NS5A inhibitors have low barrier resistance profile. The first NS5A inhibitor to be discovered was daclatasvir followed by others such as ledipasvir and ombitasvir.

3.2.1. Daclatasvir

Daclatasvir (Daklinza) is the first NS5A protein inhibitor which was approved by FDA in July 2015, in combination with sofosbuvir. It is highly effective against genotypes 1,2,3,4 and its efficacy is increased with different compounds and in conjunction with other drugs. SVR rates varies in each genotype and combination of drugs. The most common adverse reactions related to these drugs are headache, fatigue and nausea (L).

3.3. NS5B

NS5B inhibitors act by inhibiting the NS5B RNA dependent RNA polymerase (RdRp). This enzyme, has a catalytic site for nucleoside binding and at least four other sites at which a non-nucleoside compound can bind and cause allosteric alteration. Therefore, NS5B Inhibitors are divided into two groups: nucleoside (NIs), which are non-allosteric and non-nucleotide inhibitors (NNIs), which are allosteric.

NIs terminates RNA replication when they are incorporated into the elongating RNA sequence, by binding to the active site of the enzyme. Although they are only moderately potent, they have a high barrier of resistance. Because, RdRp is highly the same across all genotypes, NIs exhibit pangenotypic activity. Sofosbuvir is the first representative of this category.

In contrast, NNIs do not bind to the enzyme's active catalytic site. Instead, they act via allosteric inhibition which leads to conformational change of the polymerase. Consequently, they block the catalytic function of the enzyme and thereby indirectly block RNA replication. Although RdRp exist in all genotypes (NNIs are less potent), they are only active against genotype 1 and have low barrier to resistance.

3.3.1. Sofosbuvir

In December 6 2013 Sofosbuvir (Sovaldi) became the first direct acting antiviral of its class to be approved for use by the US Food and Drug Administration for treatment of chronic HCV. Sofosbuvir is a prodrug of 20-deoxy-20-fluoro-20-C-methyluridine monophosphate, which undergoes intracellular metabolism in the liver and is converted to its active uridine triphosphate form that acts as a chain terminator. The conversion of the monophosphate form to the active triphosphate involves four enzymatic steps and one non enzymatic chemical step. Sofosbuvir, licensed by FDA for the treatment of Genotypes 1, 2, 3 and 4, is well tolerated and is administered once daily. Moreover, it has limited drug-drug interactions, absence of a food effect and CYP3A4 metabolism of the drug and lack of significant viral resistance. (1.18, 1.17). S282T is determined as the most common mutation occurring in replicon studies with sofosbuvir. The limited replicative fitness of the S282T strain of HCV and the limited time resistant strains, have developed due to Sofosbuvir's ability to rapidly reduce viral replication explaining the high genetic barrier to resistance sofosbuvir presents (2.1.). Side effects occur most often when sofosbuvir is combined with other drugs (J). The most frequently reported side effects associated with sofosbuvir are fatigue, headache, nausea and insomnia. Finally, because sofosbuvir is predominantly eliminated from the body via kidneys, sofosbuvir based treatment, in patients with renal impairment, must be cautious (I, 1-17).

Table 1: **Summary of approved HCV drugs by FDA**

Class	Regimen	Brand Name	Side Effects	Resistance	Major Drug Interactions	Genotype coverage
NS3/4A	**Boceprevir**	Victrelis	Anemia, neutropenia, dysgeusia, and vomiting.	**Low** barrier to resistance. Most common treatment-associated resistance mutations: Genotype 1a → V36M, T54S, R155K. Genotype 1b → T54A, T54S, V55A, A156S, V170A.	CYP3A4 inducers or inhibitors.	1
	Telaprevir	Incivek	Rash, anemia, hemorrhoids, anal pruritus, fatigue, dysgeusia, nausea, diarrhea.	**Low** barrier to resistance. The most common mutations: V36M/A/L, T54A/S, R155K/T, A156S/T; alone or as combinations; common when not achieve SVR.	-Inhibitor of cytochrome p450 3A4 (CYP3A), p-glycoprotein (P-gp), OATP1B1, and OATP2B1; -Substrate of CYP3A4/5.	1
	Simeprevir	Olysio	Rash (including a potentially serious photosensitivity reaction), pruritus, nausea, unconjugated hyperbilirubinemia .	**Modarate** barrier to resistance. The most common mutations: Q80, S122, R155, and D168; common when not achieve SVR12 (90%).	- CYP34 inducers or inhibitors; -Pgp substrates; -OATP1B1/3 substrates; - Complex drug-drug interactions with HIV antiretroviral medications.	1,2,5,6
NS5A	**Daclatasvir**	Daklinza	In combination with sofosbuvir: fatigue, headache, nausea, diarrhea; serious bradycardia when coadministered with sofosbuvir and amiodarone.	Very limited data. Data from 17 patients in the ALLY-3 trial with virologic failure: A30K/S, L31I, S62A/L/P/T, or Y93H.	-Increase the level of drugs that are substrates of P-gp, OAT 1B1 or 1B3, or BCRP; -Substrate of CYP3A.	1,2,3,4
NS5B	**Sofosbuvir**	Sovaldi	Very well-tolerated in clinical trial. When used with ribavirin: fatigue and headache, nausea, insomnia, anemia.	**High** barrier to resistance. Three amino acid changes in subjects who received sofosbuvir: L159F, S282T, and V321A.	-Pgp inducers or inhibitors; -BRCP inhibitors.	1,2,3,4

3.4. Interferon free combination therapies

The possibility of developing an IFN-free regimen for difficult-to-treat genotypes, requires the combination of different DAA classes to provide high antiviral efficacy, as well as a high barrier to resistance. Furthermore, IFN-Free and RVB-Free regimens are highly desirable due to the tolerability and safety issues associated with two compounds. Many studies, assessing

Regimen	Brand name	Mechanism of action	Approved genotype coverage	Weeks of treatment (depending on the circumstances)	Side Effects

the combination of 2 or more drugs without IFN or RVB, have shown good efficacy mainly in genotype 1 patients (α48).

The IFN-Free combinations that were approved by FDA are the following: Daclatasvir and Sofosbuvir +- RBV, Ledipasvir and Sofosbuvir, Paritaprevir/Ritonavir/Ombitavir and Dasabuvir +- RBV, Ombitasvir-Paritaprevir-Ritonavir+- RBV and Simeprevir plus Sofosbuvir.

A summary of the approved by the FDA combination therapies is presented in Table 2 and the relevant clinical trials that lead to the approval of these therapies, are summarized in Table 3.

Table 2: **Summary of approved HCV combination therapies by FDA**

Daclatasvir-Sofosbuvir +- RBV		Daclatasvir (daklinza) NS5A inhibitor; sofosbuvir NS5B nucleotide inhibitor.	Genotype 1,3,4	GT 1 GT 4	No cirrhosis	12	Fatigue, headache, nausea.
				GT 1 GT 4	cirrhosis	24	
				GT 3	With prior treatment exp. or cirrhosis	24	
Ledipasvir-sofosbuvir	*Harvoni* (manufactured by Gilead Sciences)	Ledipasvir NS5A inhibitor; sofosbuvir NS5B nucleotide inhibitor.	Genotype 1	GT 1	Naïve with or no cirrhosis	12	Fatigue and headache.
				GT 1	prior treatment, no cirrhosis	12	
					prior treatment, cirrhosis	24	
Paritaprevir-Ritonavir-Ombitavir-Dasabuvir +- RBV	Viekira Pak (manufactured by AbbVie)	Ombitasvir: NS5A inhibitor; paritaprevir NS3/4A serine protease inhibitor; dasabuvir non-nucleoside NS5B polymerase inhibitor; ritonavir a potent inhibitor of CYP3A4 enzymes and is used as a pharmacologic booster for paritaprevir.	Genotype 1a/1b	G1a	No cirrhosis (+RBV)	12	Fatigue, nausea, pruritus, skin reactions, insomnia, and asthenia. Ribavirin can cause adverse effects, including hemolytic anemia.
					Cirrhosis (+RBV)	24	
				G1b	No cirrhosis	12	
					Cirrhosis (+RBV)	12	
Ombitasvir-Paritaprevir-Ritonavir+-RBV	*Technivie* (manufactured by AbbVie)	Ombitasvir: NS5A inhibitor; paritaprevir NS3/4A serine protease inhibitor; ritonavir is a potent inhibitor of CYP3A4 enzymes, booster of paritaprevir.	Genotype 4	GT 4	No cirrhosis	12	Asthenia, nausea, and fatigue.
Simeprevir-Sofosbuvir		Simeprevir (Olysio) NS3 /4A protease inhibitor; sofosbuvir NS5B nucleotide inhibitor.	Genotype 1	GT 1	No cirrhosis	12	Pruritus, rash, and hyperbilirubi-nemia.
					Cirrhosis	24	

Table 3: **Clinical trials that lead to the approval of combination therapies**

Treatment	Trial Name	Trial	Population	Treatment groups and			Rates of

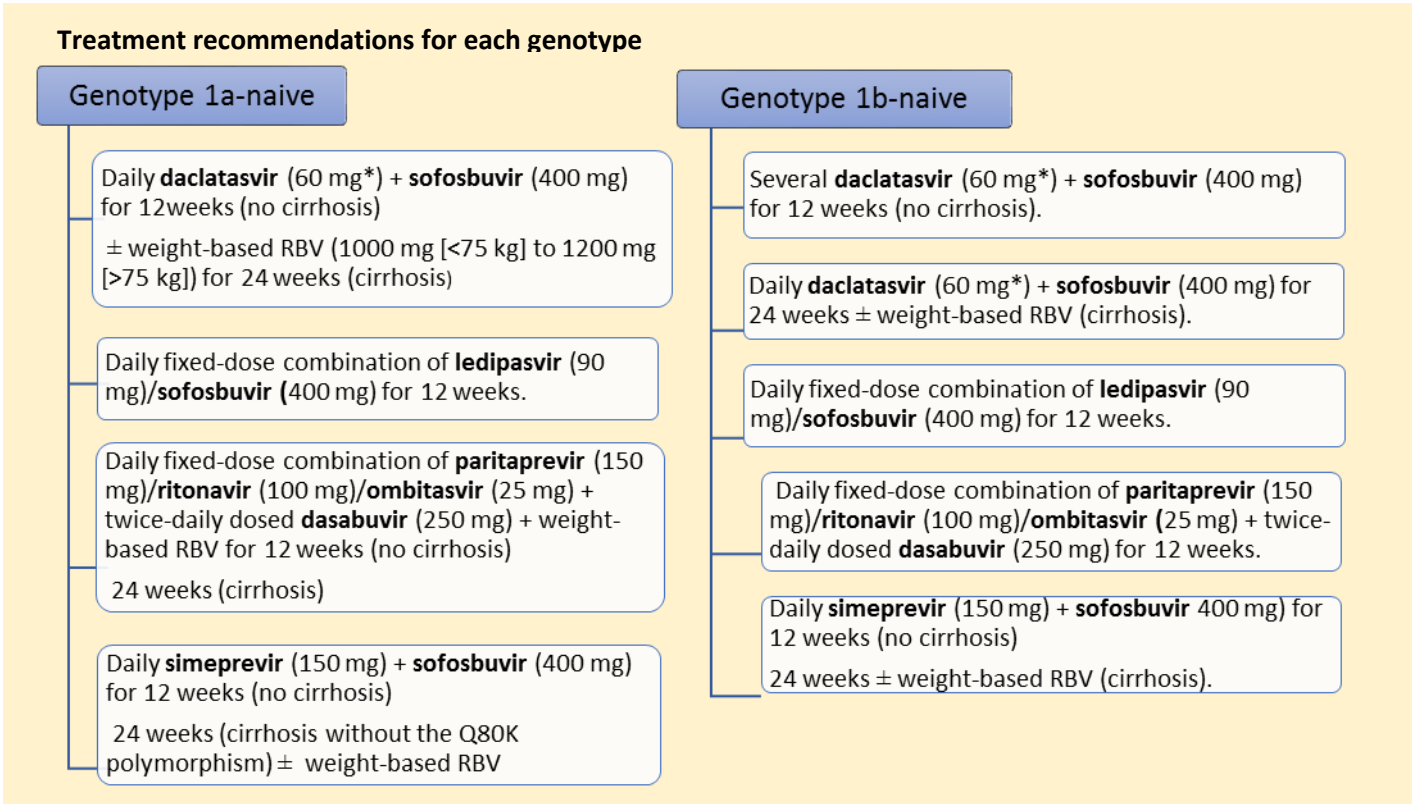
regimen		phase		duration	SVR
Daclatasvir - sofosbuvir	**ALLY-3**	Phase 3	152 tx-naïve (19% cirrhosis) or experienced patients (25% cirrhosis) gt 3	101 gt3, tx-naïve x 12 weeks 51 gt3, prior non responders x 12 weeks	SVR12: 90% SVR12: 86%
Ledipasvir - sofosbuvir (Harvoni)	**ION-1**	Phase 3	865 naive patients, gt 1 (16% cirrhosis)	100 gt1 x 12 weeks 99 gt1 + RBV x 12 weeks 99 gt1 x 24 weeks 100gt1 +RBV x 24 weeks	SVR12: 99% SVR12: 97% SVR12: 98% SVR12: 99%
	ION-2	Phase 3	440 treatment-experienced patients, gt1 (20% cirrhosis)	97 gt1 x 12 weeks 99 gt1 + RBV x 12 weeks 100 gt1 x 24 weeks 100 gt1 +RBV x 24 weeks	SVR12: 94% SVR12: 96% SVR24: 99% SVR24: 99%
	ION -3	Phase 3	647 tx-naïve gt 1 (no cirrhosis)	97 gt1 x 8 weeks 96 gt1 + RBV x 8 weeks 98 gt1 x 12 weeks	SVR12: 94% SVR12: 93% SVR12: 95%
Ombitasvir-Paritaprevir-Ritonavir and Dasabuvir (Viekira Pak)	**SAPPHIRE -1**	Phase 3	631 tx-naïve gt 1 (no cirrhosis)	322 gt1a x 12 weeks 151 gt1b x 12 weeks 455 gt1 x 12 weeks	SVR12: 95% SVR12: 98% SVR12: 96%
	SAPPHIRE -II	Phase 3	394 treatment-experienced patients, gt1 (no cirrhosis)	173 gt1a x 12 weeks 123 gt1b x 12 weeks 297 gt1 x 12 weeks 86 gt1 prior relapse x 12 weeks 65 gt1 prior partial response x 12 weeks 146 gt1 prior null response x 12 weeks	SVR12: 96% SVR12: 97% SVR12: 96% SVR12: 95% SVR12: 100% SVR12: 95%
	PEARL-II	Phase 3	186 treatment-experienced patients gt 1b (no cirrhosis)	88 gt1b + RBV x 12 weeks 91 gt1b x 12 weeks	SVR12:99% SVR12: 100%
	PEARL-III	Phase 3	419 tx-naïve gt 1b (no cirrhosis)	210 gt1b + RBV x 12 weeks 209 gt1b x 12 weeks	SVR12: 99% SVR12: 99%
	PEARL-IV	Phase 3	305 tx-naïve gt 1a (no cirrhosis)	100 gt1a + RBV x 12 weeks 205 gt1a x 12 weeks	SVR12: 97% SVR12: 90%
	TURQUOISE -II	Phase 3	380 tx-naïve and experienced gt1 with cirrhosis	140 gt1a x 12 weeks 68 gt1b x 12 weeks 208 gt1 x 12 weeks 121 gt1a x 24 weeks 51 gt1b x 24 weeks 172 gt1 x 24 weeks	SVR12: 89% SVR12: 98% SVR12: 92% SVR12: 94% SVR12: 100% SVR12: 96%
Ombitasvir-Paritaprevir-Ritonavir (Technivie)	**PEARL-I**	Phase 2b	135 tx-naïve and treatment-experienced gt 4 (no cirrhosis)	44 gt4, tx-naïve x 12 weeks 42 gt4, tx-naïve + RBV x 12 weeks 49 gt4, prior non responders +RBV x 12 weeks	SVR12: 91% SVR12: 100% SVR12: 100%
Simeprevir-Sofosbuvir	**COSMOS**	Phase 2a	167 tx-naïve and treatment-experienced gt 1	<u>Cohort 1 prior non responders:</u> 14 gt1 x 12 weeks 27 gt1 +RBV x 12 weeks 15 gt1 x 24 weeks 24 gt1 +RBV x 24 weeks <u>Cohort 2 tx-naïve and prior non responders with advanced fibrosis:</u> 14 gt1 x 12 weeks 27 gt1 +RBV x 12 weeks 16 gt1 x 24 weeks	 SVR12: 93% SVR12: 96% SVR12: 93% SVR12: 79% SVR12: 93% SVR12: 93% SVR12: 100%

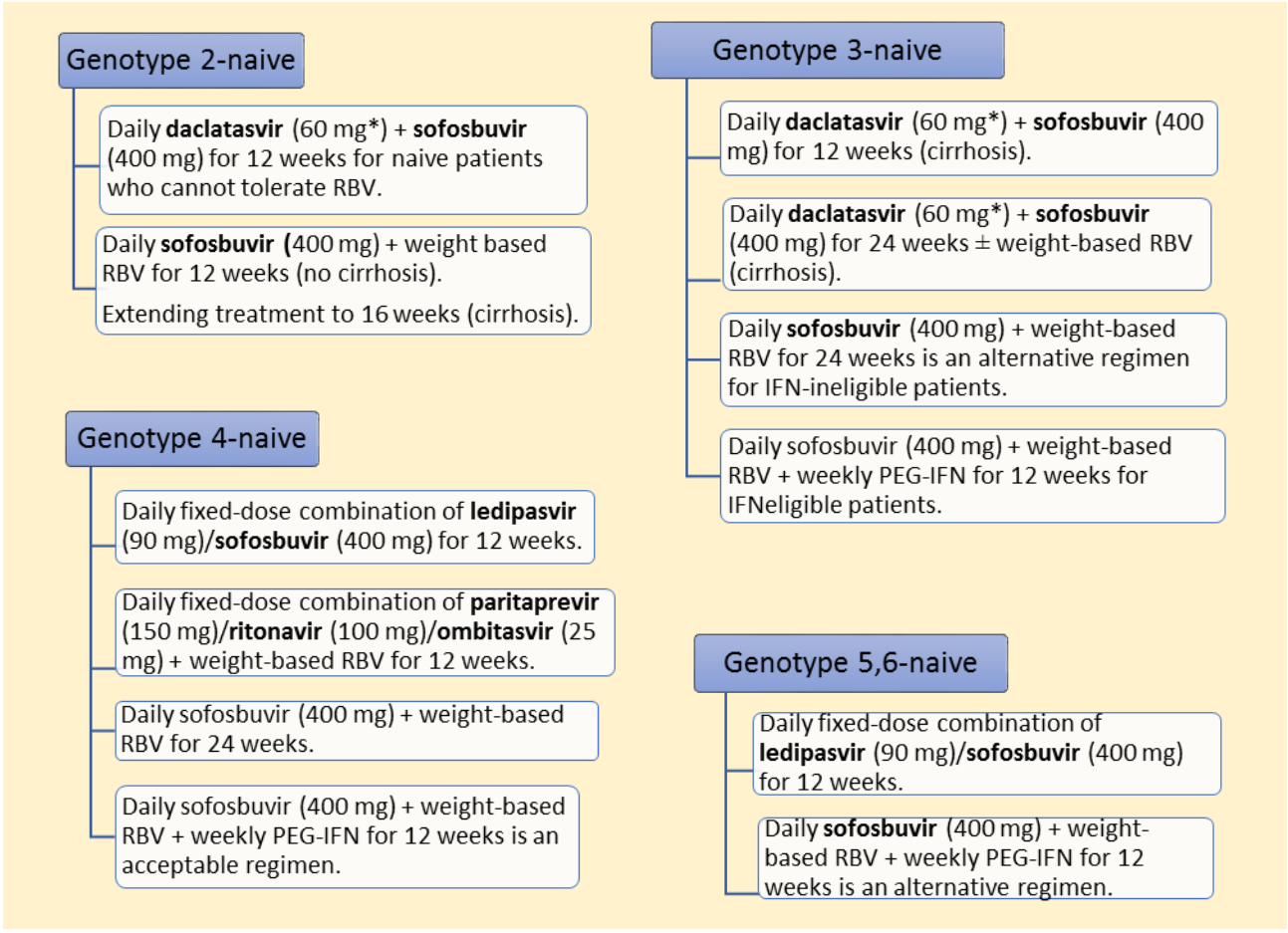
				30 gt1 +RBV x 24 weeks	SVR12: 93%

SVR12 or SVR24 = sustained virological response at 12 or 24 weeks; gt1-gt6 = genotypes 1 – genotype 6; tx-naïve = treatment naïve; RBV = ribabirin

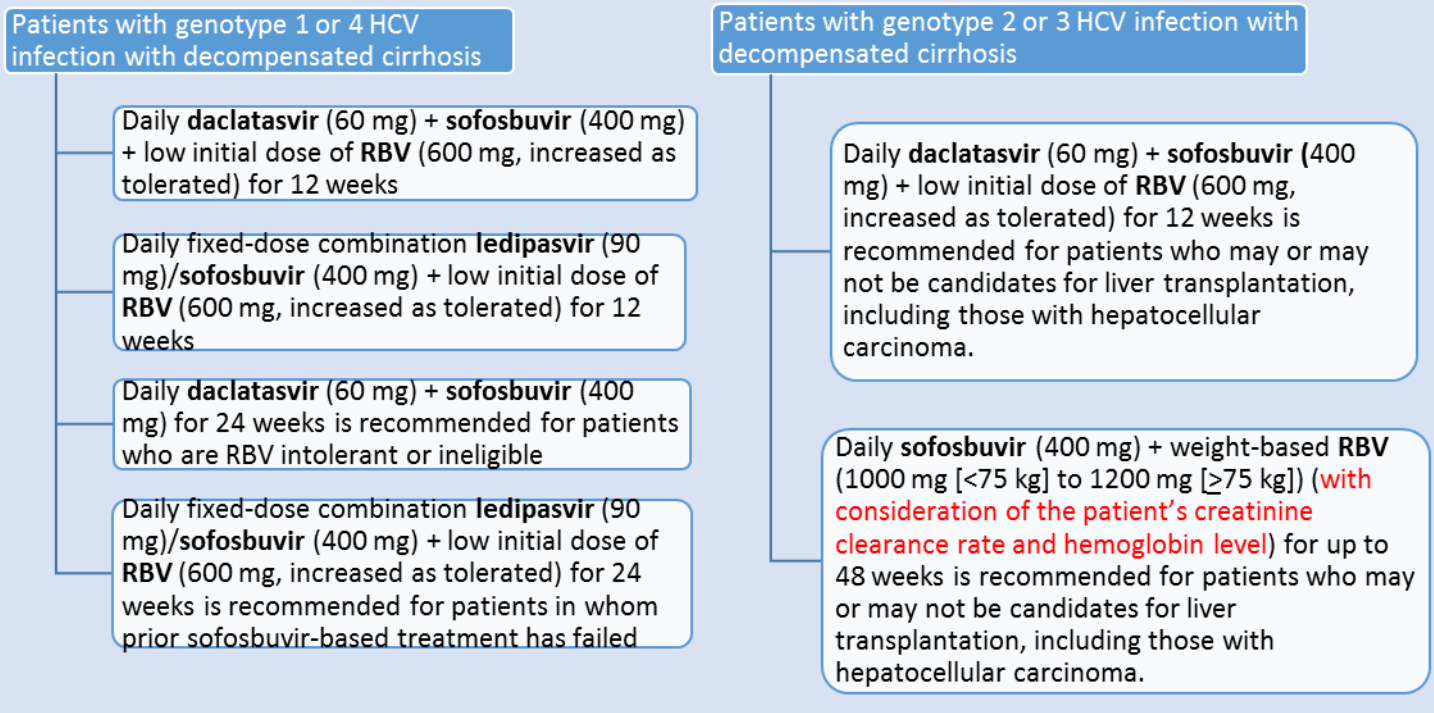
3.5. Treatment recommendations for chronic Hepatitis C Infection

The treatment recommendations vary in accordance with the HCV genotype/ subtype, the severity of liver disease, the results of prior therapy and the coinfection with HIV. The following recommendations are based on guidelines from the American Association for the Study of Liver Diseases (AASLD), the Infectious Disease Society of America (ISDA), and the International Antiviral Society-USA (IAS-USA).





Treatment recommendations for patients with decompensated cirrhosis



Treatment recommendations for post –liver transplantation patients

Recurrent HCV infection post-liver transplantation genotype 1

- Daily **sofosbuvir** (400 mg) + **simeprevir** (150 mg) ± weight-based **RBV** for 12 weeks is an alternative regimen for patients with HCV genotype 1 infection in the allograft, including those with compensated cirrhosis.
- Daily fixed-dose combination of **paritaprevir** (150 mg)/**ritonavir** (100 mg)/**ombitasvir** (25 mg) + twice-daily dosed **dasabuvir** (250 mg) + weight-based **RBV** for 24 weeks is an alternative regimen for patients who have early stage fibrosis (Metavir stage F0-F2).

Recurrent HCV infection post-liver transplantation genotype 2 infection in the allograft, including those with compensated cirrhosis.

- Daily **daclatasvir** (60 mg) + **sofosbuvir** (400 mg) + low initial dose of **RBV** (600 mg, increased as tolerated) for 12 weeks
- Daily **sofosbuvir** (400 mg) + weight-based **RBV** for 24 weeks
- Daily **daclatasvir** (60 mg) + **sofosbuvir** (400 mg) for 24 weeks is recommended for patients who are RBV intolerant or ineligible.

Recurrent HCV infection post-liver transplantation genotype 3 infection in the allograft, including those with compensated cirrhosis.

- Daily **daclatasvir** (60 mg) + **sofosbuvir** (400 mg) + low initial dose of **RBV** (600 mg, increased as tolerated) for 12 weeks
- Daily **sofosbuvir** (400 mg) + weight-based **RBV** for 24 weeks
- Daily **daclatasvir** (60 mg) + **sofosbuvir** (400 mg) for 24 weeks is recommended for naive patients who are RBV intolerant or ineligible.
- Daily **sofosbuvir** (400 mg) + low initial dose of **RBV** (600 mg, increased as tolerated) for 24 weeks is recommended for liver transplant recipients with decompensated cirrhosis (CTP class B or C) who have HCV genotype 3 infection in the allograft.

Recurrent HCV infection post-liver transplantation genotype 1 or 4 in the allograft, including those with compensated cirrhosis.

- Daily **daclatasvir** (60 mg) + **sofosbuvir** (400 mg) + low initial dose of **RBV** (600 mg, increased as tolerated) for 12 weeks
- Daily fixed-dose combination of **ledipasvir** (90 mg)/**sofosbuvir** (400 mg) with weight-based **RBV** (1000 mg [<75 kg] to 1200 mg [≥75 kg]) for 12 weeks
- Daily **daclatasvir** (60 mg) + **sofosbuvir** (400 mg) for 24 weeks is recommended for patients who are RBV intolerant or ineligible
- Daily fixed-dose combination of **ledipasvir** (90 mg)/**sofosbuvir** (400 mg) for 24 weeks is recommended for naive patients who are RBV intolerant or ineligible.

Treatment recommendations for patients with HIV coinfection