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HCV DRUG RESISTANCE AND DAA AGENTS

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ABSTRACT

New treatment options for HCV infection with Direct-Acting Antivirals (DAAs) increased SVR rate in treated patients but on the other hand drew attention to the problem of HCV drug resistance. Drug-resistant HCV mutants are present in all infected patients even before the treatment initiation and their number grows significantly over the first few days of DAAs therapy. But HCV has no known genetic form of intra-cellular persistence (does not integrate with host's genome and cannot produce episomal forms) which enables its total eradication. It is likely that the effective interferon-free, based on all-oral DAAs drug combination will be available within the next few years. This paper reviews HCV resistance mechanisms and their significance in treatment. I also presents results of recent DAAs trials.

Key words*: HCV, drug resistance, nucleoside analogs*

INTRODUCTION

Approximately 170 million people are HCV carriers worldwide (1). Until recently, pegylated interferon alpha in combination with ribavirin was the first-line therapy, however, successful HCV eradication was achieved in only 50% of patients (therapy efficacy was lower in patients infected with HCV genotype 1 & 4 and higher in individuals diagnosed with HCV genotype 2 and 3) (2). Therapeutic failure with pegylated interferon alpha in combination with ribavirin results from multiple virus-, host-, and treatment-dependent factors. These factors are listed in Table 1.

With the introduction of Direct-Acting Antivirals – DAAs: telaprevir and boceprevir (protease inhibitors), the likelihood of HCV eradication in patients infected with HCV genotype 1 has increased, however, HCV resistance to drugs raises some serious health concerns.

DRUG-RESISTANT HCV VARIANTS

Drug-resistant HCV variants which typically have lower replication fitness cannot be detected with standard molecular methods, which is why they had not attracted much attention before experiments on DAAs

Table 1. Factors that affect response to pegylated interferon alpha and ribavirin in the treatment of HCV infections.

1. HCV-dependent (positive) - genotype 2,3 v. 1,4 - number of HCV copies in blood serum (< 2 M copies - LVL) - fast kinetics of HCV clearance
2. Host-dependent (negative) - past and present history of alcohol abuse - age > 40 years when infected - male - HBV and HCV co-infection - obesity - abnormalities in lipid metabolism - advanced hepatic fibrosis or cirrhosis on initiation of treatment - racial affinities: African Americans: lower SVR rates, (Asians: higher SVR rate) - IVDU (intravenous drug users) - Interleukin-28b polymorphism (IFNλ-3) - SVR rate dependent on genotype polymorphism: CC-82%, CT-42%, TT-33%) - poor treatment tolerance - non-compliance
3. Therapy-dependent - IFNalpha in monotherapy < IFNalpha/RBV combination therapy < PEG IFNalpha/RBV combination therapy - therapy duration: for genotype 1,4,5,6 3<6<12 months, for genotype 2,3 3<6 months - RBV dose per b.w. - peginterferon alpha-2b (PEG-Intron) *vs*. peginterferon alpha-2a (PEGASYS) – IDEAL study

Classification of factors according to own concept.

began. Identified in 0.3% to 2.8% of carriers, mutations responsible for resistance to protease inhibitors (telaprevir, BILN2061, ITMN-191, SCH6 and boceprevir), NS5B polymerase inhibitor (AG-021541) and NS4A antagonist (ACH-806) were considered to be sporadic and clinically insignificant (3). Variants inducing resistance to telaprevir (R155K, V36M) were found in 0.6-1.2% of non-treated carriers of genotype 1b, and were either undetectable or detected in a very small percentage of people (0.07% for A156S and V36M and 2.1% for T54S) with genotype 1a. (4). Likewise, in patients with genotype 1a, mutations resistant to boceprevir (V36M, T54S, R155K) and 1b (T54A, V55A, A156S, I/V170A) were very rare (5).

However, with the beginning of clinical studies on protease inhibitors, it was found that drug-resistant variants can be detected as early as on the second day after introduction of therapy in 5 to 20% of individuals [6]. There are several reasons that could explain such early mutations. Firstly, HCV replication is typically very dynamic. Within a single day, there are approximately 10^{12} new virions with 2-3 hours of estimated half-life, which is even shorter in case of intracellular forms (7). Also, RNA-dependent RNA HCV polymerase is very error-sensitive (8). Based on mathematical models, it was concluded that all possible single, double, and most probably also triple HCV mutations determining resistance to DAAs are present in carriers prior to the beginning of therapy (6).

Efficient antiviral therapy based on DAAs eliminates drug-sensitive drugs, whereas drug resistant strains can proliferate freely (2). It is also possible that subsequent compensatory mutations are selected in conditions of exposure to drug, which may be able to recover in vivo replication fitness of the mutated virus, i.e. replication fitness from before therapy is thereby restored. In vivo replication fitness is the key condition for the mutated virus to be able to replicate (9) since any virus of low replication potential - even if it is highly resistant - is of lower clinical significance as compared to any other virus of low resistance and high replication fitness (2). HCV variants carrying a mutation at the locus 156 – which exhibit the highest resistance to telaprevir and low replication potential – were found to be present at a very early stage of therapy and quickly replaced by viruses representing higher replication fitness (155, 36 and 155, 36 and 156) (10, 11). However, a wild-type virus dominance can be detected as early as in 2 days on discontinuation of unsuccessful triple-drug therapy. But still, drug-resistant variants remain present, although are very few. As a result, even if any drug-resistant strains are isolated in these patients, it appears perfectly safe and cannot be considered a contraindication for repeated therapy with first-generation protease inhibitor with cross-resistance to telaprevir or boceprevir, as long as these drugs are part of treatment regimen containing other drugs which successfully prevent replication of HCV without any signs of cross-resistance to protease inhibitors (12).

Thus, HCV ability to produce drug-resistant mutations is not necessarily a serious health concern as in case of HIV and HBV. Unlike these viruses, HCV only replicates in cellular cytoplasm of the host, is not integrated with the host genome, and cannot produce any reservoirs of episomal forms (13). Therefore, antiviral therapy creates some solid opportunities for HCV eradication (13).

Relying on mathematical models, the hypothesis is that therapy with DAAs could be applied with combinations of drugs offering a genetic barrier of 4 or more mutations (6).

According to the results of phase II and III studies on DAAs, triple-drug therapy failure in compliant persons can be mainly attributed to the lack of response to interferon and ribavirin, which results in the selection of pre-existing mutations resistant to DAAs (2).

It is difficult to create an interferon-free treatment regimen based on oral DAAa, offering optimum efficacy towards all HCV genotypes along with high genetic barrier, in order to guarantee high percentage of recoveries at a reasonable time (12).

EXAMINING NEW TREATMENT REGIMENS

Protease inhibitors appear to have a superior resistance profile as compared to other drugs, such as RNA-dependent RNA HCV polymerase nucleoside / nucleotide inhibitors, which results in the selection of viruses of lower replication potential, and cyclophilin inhibitors which target HCV-related host protein instead of viral protein (14).

Some promising results were produced in studies on the efficacy of asunaprevir - NS3/4A protease inhibitor, and of daclatasvir – NS5A inhibitor. 9 in 10 carriers of genotype 1b who failed to respond to pegylated interferon alpha and ribavirin eliminated the virus within 24 weeks of therapy. High sustained virologic response (SVR24) rates were reported in all patients (15).

PSI-7977 - nucleotide analog polymerase inhibitor - is another promising drug which, in an add-on therapy with PRG/RBV, produced 90% SVR rate in patients infected with genotype 1 in the PROTON study (16).

In another study, SVR was observed in 10 out of 10 previously non-treated carriers of genotype 2 or 3 after 12 weeks on PSI-7977 in combination with interferon-free ribavirin (17). Further interesting studies on this molecule are in course, also in our center.

ABT-450 protease inhibitor and ABT-072 non-nucleoside polymerase inhibitor are two very promising

DAAs. The PILOT study evaluated the efficacy of a 12-week interferon-free therapy - ABT-450 as an add-on to ritonavir, and ABT-072 in combination with ribavirin. In 12 weeks after the treatment discontinuation, HCV viremia was absent in 91% of previously non-treated patients infected with genotype 1, IL-28B CC (18). In the COPILOT study on ribavirin as an add-on to ABT-450 and ABT-333 ritonavir (non-nucleoside polymerase inhibitor), viremia was undetectable in over 90% and 47% of previously non-treated and antiviral-treated individuals, respectively, in 12 weeks after the therapy was discontinued (19). Alisporivir (Debio 025), the hosts's cyclophilin inhibitor, is currently tested in phase III studies. Resistance to alisporivir can be attributed to mutations in domain II of NS5A. Single mutations in NS5A can cause only negligible resistance of HCV to alisporivir, however, where multiple mutations are present in NS5A, significant resistance to all classes of cyclophilin inhibitors can be observed. On the other hand, in consideration of the results of in vivo studies, alisiprovir can be considered to have low potential to induce viral resistance (20). In short-term studies, Debio 025 as an add-on to ribavirin, NS3 protease inhibitors, or nucleoside / non-nucleoside NS5B polymerase inhibitors was found to have an additive effect in inhibiting HCV replication. Debio 025 appears to be able to delay or prevent resistance to protease inhibitors and to nucleoside / non-nucleoside polymerase inhibitors (21). However, due to adverse effects and the development of new cyclophilin inhibitors, works on this drug were discontinued.

CONCLUSIONS

HCV is present in multiple cellular compartments (apart from hepatocytes also in lymphocytes) , and the isolated HCV strains exhibit significant differences (22). The biology of HCV differs significantly from that of HBV and HIV, especially due to the absence of reservoirs (HCV does not integrate with host's genome and cannot produce episomal forms) which makes it possible to eradicate it completely. It appears that successful eradication can be achieved with a combination of drugs that effectively inhibit resistant mutants for several weeks or months. In consideration of the progress in clinical studies on DAAs, an efficient treatment regimen will be hopefully available in just a few years time (12).

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