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Meeting Report

Meeting report: 27th International conference on antiviral research, in Raleigh, NC, USA

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ABSTRACT

The 27th International Conference on Antiviral Research (ICAR) was held in Raleigh, North Carolina, USA from May 12 to 16, 2014. This article summarizes the principal invited lectures. John Drach (Elion Award) described the early days of antiviral drugs and their novel modes of action. Piet Herdewijn (Holý Award) used evolutionary pressure to select DNA polymerases that accept nucleoside analogs. Replacing thymine by 5-chlorouracil led to the generation of a new form of Escherichia coli. Adrian Ray (Prusoff Award) demonstrated how produgs can markedly improve both the efficacy and safety of potential drugs. The keynote addresses, by David Margolis and Myron Cohen, tackled two emerging areas of HIV research, to find an HIV “cure” and to prevent HIV transmission, respectively. These topics were discussed further in other presentations – a cure seems to be a distant prospect but there are exciting developments for reducing HIV transmission. TDF-containing vaginal rings and GSK-744, as a long-lasting injection, offer great hope. There were three mini-symposia. Although therapy with TDF/FTC gives excellent control of HBV replication, there are only a few patients who achieve a functional cure. Myrcludex, an entry inhibitor, is active against both HBV and HDV. The recent progress with HBV replication in cell cultures has transformed the search for new antiviral compounds. The HBV capsid protein has been recognized as key player in HBV DNA synthesis. Unexpectedly, compounds which enhance capsid formation, markedly reduce HBV DNA synthesis. The development of BCX4430, which is active against Marburg and Ebola viruses, is of great current interest.

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1. Introduction

This article provides an overview of the invited lectures at the 27th International Conference on Antiviral Research, sponsored by the International Society for Antiviral Research (ISAR), which was held in Raleigh, North Carolina, USA from May 12 to 16, 2014. It begins with reports of lectures by the recipients of ISAR's three major awards, held in memory of Gertrude Elion, Antonín Holý and William Prusoff. These are followed by brief summaries of the keynote addresses and the three mini-symposia on “Hepatitis B virus”, “Research Triangle Park” and “Challenges in HIV infection, treatment and prevention”. Because this review article simply provides short accounts of oral presentations, it is not generally accompanied by references to the scientific literature. Any descriptions of favorable treatment outcomes should not be taken as recommendations for clinical use.

2. Gertrude Elion memorial award lecture: collaborative antiviral studies for the discovery of drugs to treat cytomegalovirus infections

John C. Drach, Ph.D., University of Michigan, Ann Arbor, Michigan, USA (Fig. 1).

Gertrude B. (Trudy) Elion was born in New York City and was pleased to work for the Burroughs Wellcome Co. when based in New York but was concerned when it transferred to Research Triangle Park, North Carolina, not many miles from this year’s meeting site. However, within just a few months she declared that she was “at home” in North Carolina. She was awarded the Nobel Prize in Physiology or Medicine in 1988 for her pioneering work in purine biosynthesis which paved the way for the discovery of drugs to treat organ rejection, cancer and viral diseases.

The focus of John’s presentation was on the research conducted in his own and his collaborators’ laboratories that ultimately led to the invention of three compounds which were discovered to have antiviral activity against human cytomegalovirus (HCMV) and which later entered clinical trials: BDCRB pyranoside (GW275175X) (Phase I), maribavir (Phases I, II and III) and cyclopropavir (Phase I). His major collaborators included Karen Biron, Charles Shipman, Leroy Townsend, and Jiri Zemlicka. To date, there are only five FDA-approved drugs for treatment of HCMV infections: cidofovir, fomiviren, foscarnet, ganciclovir and valganciclovir.

Being inspired by the presence of a naturally-occurring 5,6-dimethylbenzimidazole nucleotide in Vitamin B12, research on benzimidazole nucleosides was initiated by medicinal chemists in the 1950s and '60s. This led to the synthesis of a trichloro analog in Townsend’s laboratory at the University of Utah and later the discovery of its activity against HCMV in John’s laboratory. Much work, in both their laboratories at the University of Michigan, established that it and its 2-bromo analog (BDCRB) have excellent activity against HCMV with very low cytotoxicity. Surprisingly, it was found to be inactive against other herpes viruses and it did not need conversion to a triphosphate to be active against HCMV.

Collaborative studies with Karen Biron at Burroughs Wellcome established that, unlike many other anti-virals that inhibit viral DNA synthesis such as ganciclovir (GCV), these compounds acted...
by a novel mechanism, inhibition of viral DNA processing. It was the viral resistance studies which revealed the viral targets, pUL89 and pUL56. These two proteins, with pUL104, form a complex known as the terminase which cuts newly synthesised HCMV DNA into unit lengths for packaging into virions. Although BDCRB had many desirable properties in vitro, it had poor pharmacokinetics in mice and monkeys due to hydrolysis of its glycosidic bond; therefore it was not developed for human use. Much additional work in Drach’s and Townsend’s laboratories at Michigan and by Biron’s group at Burroughs Wellcome ultimately led to two potential drug candidates, BDCRB pyranoside and maribavir (Fig. 2). Both compounds have excellent activity against HCMV, low toxicity, and excellent pharmacokinetics. Clearly, their modes of action differed markedly from that of GCV. Quite unexpectedly, they have different mechanisms of action.

BDCRB pyranoside has a mechanism of action very similar to its parent compound BDCRB, inhibition of DNA synthesis, albeit indirectly. It is a 2-isopropylamine derivative of BDCRB except that it has the unnatural L-sugar configuration. Its mechanism of action involves inhibition of the viral kinase (pUL97), which phosphorylates another viral protein, pUL44. Phosphorylated pUL44 is necessary for viral DNA synthesis. Thus inhibition of pUL97 by maribavir inhibits viral DNA synthesis. Interestingly, pUL97 is also the kinase that activates (phosphorylates) GCV. Resistance studies confirmed that a single mutation in UL97, resulting in a mutation in the kinase (Leu397Arg), was necessary and sufficient for resistance to maribavir. In a further study of resistance, virus already resistant to BDCRB was passaged in increasing concentrations of maribavir and resistant virus was isolated. This strain grew at the same rate as the wild-type virus and was resistant to both BDCRB and maribavir. As expected, resistance to BDCRB was due to known mutations in UL56 and UL89. However, no mutations were found in UL97. Further investigation showed that a single base change in UL27 (T1004C) was necessary and sufficient for resistance to maribavir. The role of the encoded protein was then unknown but the amino acid mutation (Leu335Pro) is in the middle of the protein. Similarly, Biron’s group detected resistance due to mutations in UL56 and UL89. However, no mutations were found in UL27 (T1004C) was necessary and sufficient for resistance to maribavir. The role of the encoded protein was then unknown but the amino acid mutation (Leu335Pro) is in the middle of the protein. Similarly, Biron’s group detected resistance due to mutations in UL56 and UL89. However, no mutations were found in UL27 (T1004C) was necessary and sufficient for resistance to maribavir. The role of the encoded protein was then unknown but the amino acid mutation (Leu335Pro) is in the middle of the protein.

3. The Antonín Holý memorial award lecture: from modified nucleoside to a chemically modified genome

Piet Herdewijn, Rega Institute for Medical Research, KU Leuven, Belgium (Fig. 4).

The 2013 ICAR began with a symposium, on the legacy of the late Antonín (Tony) Holý, at which the establishment of a new ISAR award in medicinal chemistry was announced. The awardee is to be a senior scientist of international stature in medicinal chemistry and who has made innovative contributions impacting antiviral drug discovery or development. Piet is, therefore, the first to receive this award.

In the late 1970s, the potent activities of BVDU and BVaRAU against herpes simplex virus type 1 (HSV-1) and varicella zoster virus (VZV) were discovered; this work motivated Piet to start antiviral research with the synthesis of carbocyclic BVDU. Through to the early 1990s, he synthesized several other nucleoside analogs with bicyclic bases having good activity against HSV-1 and VZV. During the 1990s, emphasis switched to investigating the effect of modifying the sugar ring, in particular the synthesis of six-membered rings containing an oxygen or a double bond. Piet showed examples of compounds with activity against HSV-1, HSV-2, VZV and HCMV.

Back in 1984, Erik De Clercq showed Piet a paper on AIDS, one of the authors being Phil Furman. This publication stimulated the search for anti-HIV compounds. Many compounds were discovered with potent activities (and good selectivity indices) against HIV. Piet worked out the first structure–activity relationships of anti-HIV deoxy nucleosides. Starting in the late 1980s, Tony Holý synthesised a series of phosphonates. At the 2013 ICAR, Erik De Clercq recalled how this work led, ultimately, to tenofovir, which was to become a major success for treating HIV-infected patients. From its first introduction in 2001, its market share has increased to well over 40%. In 2002, having a single-pill regimen was agreed as a way forward to simplify, and thereby enhance, HIV therapy. This led to Atripla being approved in 2006, Complera in 2011 and Stribild in
Adrian started his lecture with photos of William (Bill) Prusoff and reminisced of his days with Bill, Raymond Schinazi and Yung-Chi (Tommy) Cheng.

Adrian presented examples to illustrate two models of how a prodrug strategy can transform a potential drug into a much improved clinical candidate. In the first, the prodrug alters the distribution of the pharmacologically active nucleotide analog to tissues where viral infection is taking place (on-target) and away from tissues resulting in adverse events (off-target). In the second, the prodrug enables one to select a drug candidate based more directly on the intrinsic properties of the active nucleotide-triphosphate analog via by-passing an inefficient activation (phosphorylation) of the corresponding nucleoside analog.

Sofosbuvir (Sovaldi®), a prodrug of 2′-F-2′-C-MeUMP, was approved in the USA on 6th December, 2013 for treatment of patients with hepatitis C. This is a fine example of a prodrug enhancing the activity of the parent compound. The nucleoside analogue, 2′-F-2′-C-MeU, is poorly active due to restricted phosphorylation to the monophosphate. Sofosbuvir, a nucleotide analogue prodrug of 2′-F-2′-C-MeU, delivers the monophosphate into the cell and this is then further phosphorylated efficiently to give high levels of the triphosphate which inhibits HCV RNA polymerase.

Adrian recalled being much impressed by a result reported at the meeting in 2007 of the American Association for the Study of Liver Diseases (AASLD). In a Phase II monotherapy trial in patients with HCV, at day 3, the viral loads were reduced by log_{10} 3.2 and log_{10} 1.1 for VX-950 (1250 mg bid, n=10) and RG-7128 (1500 mg bid, n=8), respectively. However, from day 4 to 13, the polymerase inhibitor (RG-7128) had continued to reduce the viral load, reaching a reduction of log_{10} 2.7. On the other hand, the protease inhibitor (VX-950) did not give a sustained reduction, with the viral load starting to increase from day 6. At day 13, the viral load was only log_{10} 2.2 less than baseline. Nucleotide analogues have two advantages over other classes of inhibitors. There is a high genetic barrier to resistance selection, due to the HCV RNA polymerase being highly specific for its natural substrates and template. This specificity can be altered but only under extreme evolutionary pressure (see Section 3). Also, nucleotide analogs often have pan-genotype activity because the active site of the HCV NS5B polymerase is so highly conserved.

As an example of how prodrugs can impact a discovery program, allowing for more targeted delivery and for the optimization of the intrinsic properties of the triphosphate, Adrian presented the history of the GS-6620 program. The C-adenine analogue (2′-C-Me-4-aza-7,9-dideazaA, C-Nuc1) was compared to the corresponding N-nucleoside, MK608. In a genotype1b replicon assay, the EC_{50} values were 2.5 μM and 0.08 μM respectively. However, their triphosphates were equally effective against HCV NS5B polymerase (IC_{50} values both 0.3 μM). In the replicon system, the triphosphate of the N-Nuc (MK608) was formed more efficiently than that of the
C-Nuc1, thus explaining the lower activity of the C-Nuc1. However, in primary human hepatocytes, C-Nuc1 was phosphorylated to the triphosphate more efficiently than the N-Nuc (MK608). This illustrates the importance of using primary human cells.

C-Nuc1 seemed to have a benign in vitro toxicity profile, including not inhibiting the mitochondrial DNA polymerase-gamma, but it had very significant toxicity in animals. In a collaboration between Gilead and Craig Cameron at Pennsylvania State University, the researchers sought to identify the toxicity target(s) for ribonucleotide analogues, including C-Nuc1 and others that had been stopped in Phase II trials. These studies showed a correlation between C-Nuc1 and the Phase II candidates, R1626, NM283 and BMS986094/IDX184. All the latter were efficiently incorporated into RNA by the mitochondrial RNA polymerase (>70% of the corresponding natural nucleotide). The triphosphate of C-Nuc1 was also an efficient substrate (22% the rate of ATP). In contrast, the active nucleotide analogs, formed by drugs approved for the treatment of HCV, were poor substrates. Ribavirin was poorly incorporated (about 5%) and sofosbuvir was below the limit of detection (≈0.002%). More extensive in vitro and cell culture evaluation of the compounds could have saved the expense of taking them into clinical trials.

Understanding that the mitochondrial RNA polymerase is an important target for ribonucleotide toxicity, the Gilead team sought analogs that were not incorporated by this polymerase. Adding a CN group to the 1’ position of C-Nuc1 did not change its activity as an HCV NS5B polymerase inhibitor (IC_{50} 0.3 μM) but it did reduce incorporation in the mitochondrial RNA assay (<0.02%). However, in the absence of a nucleotide prodrug to bypass the first phosphorylation step, the resulting di-substituted nucleoside analog would not be a drug candidate because it was not efficiently activated in cells. Application of a nucleotide prodrug strategy allowed this nucleotide to be pursued further. Oral absorption, delivery of the monophosphate into hepatocytes and high hepatic extraction were criteria used as part of the prodrug optimization process. A nucleotide prodrug, GS-464335 (a mixture of diastereoisomers at phosphorous) was well absorbed in dogs (>80%). Comparing the pre-hepatic and post-hepatic plasma drug levels, about 80% of the absorbed drug was taken up by the liver. Inside cells, GS-464335 was converted to the corresponding monophosphate which was efficiently converted to the triphosphate. At 24 h, the triphosphate levels remained about 2-fold above the IC_{50} value. A pure stereosomer was selected and later named GS-6620. In a Phase II trial (900 mg, bid 5 days), the mean reduction in HCV load was about log_{10}1.5. Two subjects achieved HCV RNA <25 IU/ml. However, the pharmacokinetics and antiviral responses were highly variable. Whereas the activity results were disappointing, clinical proof of concept was observed in terms of safety. GS-6620 did have a markedly improved safety profile relative to C-Nuc1, progressing through chronic toxicology studies in rats and dogs at relatively high doses.

The story of GS-6620 illustrates both how nucleotide prodrugs enable further progression of candidates and also the complexity of predicting the behavior of nucleotide prodrugs across species. One wonders what cell culture test or animal model may have predicted such variability. When selecting fiancliclovir as the prodrug for penciclovir, one potential prodrug was rejected because the pharmacokinetics in rats varied widely between individual animals (Vere Hodge et al., 1989). A recent publication by Adrian and his team highlights the metabolism of GS-6620 by carboxylesterase 2, an enzyme highly expressed in the human small intestine but not uniformly expressed in different animal species, as a possible reason for the highly variable and suboptimal intestinal absorption of GS-6620 in humans (Murakami et al., 2014).

The focus of Adrian’s talk then switched to HIV. Over the last 15 or 20 years in North America, the HIV-infected population has been changing, becoming older (now 33% over 50 years old vs <10% in 1995) and more likely to be obese (in every USA state, >20% adults with BMI ≥30). This has led to a shift in the focus of antiretroviral therapy (ART), from solely control of HIV replication to now include tolerability in older, possibly obese, patients.

The first example given for HIV was how application of a different prodrug strategy can markedly change the distribution even when delivering the same pharmacologically active nucleotide analog. The first approved prodrug of tenofovir (TFV) was TFV disoproxil fumarate (TDF). More recently, TFV alafenamide (TAF) has been progressed into clinical development. A key difference in the properties of the two prodrugs is their stability in plasma, with half-lives of 0.4 and 90 min, respectively. Even with a short half-life, TDF gave better delivery of TFV into cells, as indicated by the HIV EC_{50} values in cell culture assays but there clearly was room for improvement; the EC_{50} values for TFV, TDF and TAF are 1.2, 0.015 and 0.003 μM respectively. Whereas the gain in cell culture EC_{50} value may be modest, this is not the only gain. The increased stability of TAF allows it to load on-target cells and tissues (e.g., lymph nodes) for a longer period of time resulting in increased lymphoid cell and tissue levels at greatly reduced circulating TFV levels, leading to less exposure to off-target tissues (e.g., kidney). In monotherapy studies after oral dosing with TDF (300 mg) and TAF (25 mg), the plasma TFV AUC is reduced from 1920 to 268 ng.h/ml respectively whereas the reduction in HIV load from baseline is improved, from log_{10}0.97 to log_{10}1.46 copies/ml respectively, reflecting the more efficient delivery of TAF to target cells and tissues. Clearly the lower dose of TAF (25 mg) relative to TDF (300 mg) will give TAF a marked advantage when considering combination pill therapy.

Understanding how marked a difference a prodrug can make from the TAF example, Adrian went on to describe how a prodrug approach transformed a new nucleotide project in which intrinsic properties of the pharmacologically-active nucleotide analog were optimized. Their starting point was GS-2128 (D4APi), which had good activity against both wild-type and resistant HIV strains but was an active inhibitor of mitochondrial polymerase-gamma. On comparing the known structures of HIV RT and mitochondrial polymerase-gamma, differences in the 2’-binding pocket were noted. This led to GS-9148 in which 2’-F was added to GS-2128 (Fig. 8).

Compared to TFV, GS-9148 was about 3-fold less active against wild-type HIV but maintained better activity against resistant strains (K65R and multiple thymidine analog resistance mutations). Most importantly, it was inactive (IC_{50}>300 μM) against mitochondrial polymerase gamma. More than 50 prodrugs were synthesized and evaluated in metabolism studies and in dogs (intravenous and oral administration). Then the enantiomers were tested separately in dogs. This led to the selection of GS-9131. Whereas TFV is efficiently utilised by renal uptake transporters, GS-9148 was poorly taken into the kidney. No adverse renal findings were observed with the prodrug (GS-9131) in 28-day studies in rats, dogs and monkeys at the highest doses tested (300 mg, 20 mg and 30 mg/kg daily, respectively).

In summary, this work has given examples of the prodrug approach being used successfully both to increase selectivity (by loading on-target tissues vs off-target tissues) and to increase activity (via by-passing metabolic constraints). Adrian presented cases in which a prodrug strategy was able to fulfill the full potential of a selective, active triphosphate analog and enable its further progression as a clinical candidate.
5. Keynote addresses

The keynote speakers were David Margolis and Myron Cohen (Fig. 9).

5.1. Eradication therapies for HIV: building the critical path

David Margolis, University of North Carolina, NC, USA

In HIV-infected patients, there is a long-lasting reservoir of HIV in the form of integrated viral DNA in resting CD4+ memory cells of the host immune system. Therefore, even if it were possible to eliminate 100% of viral replication, a reservoir of HIV would remain. There may be reservoirs in other long-lived cells. To date, there is only one known HIV patient who has been cured of his infection, the “Berlin Patient”. He was treated for cancer by chemotherapy followed by a bone-marrow transplant. Being CCR5+/−, the chemotherapy had a greater chance to remove all the CCR5+ve cells. The bone marrow donor was CCR5−ve. Although this patient continues to have no sign of HIV infection, this is hardly a viable treatment option for most HIV-infected patients.

Even in subjects with HIV replication well controlled by therapy, 70% have detectable plasma viremia which does not appear to decay over time (at least two years). To improve the sensitivity of the assay for HIV, 4 billion lymphocytes are mixed with antibody attached to magnetic beads. This selects for the CD4+ T cells, about 0.2–1 billion cells. The limit of detection is 1 copy of HIV RNA/million cells, limit of quantitation is 10 copies/million cells.

To reduce the reservoir of HIV, it was suggested that activation of integrated HIV in resting CD4+ T cells would give renewed HIV RNA synthesis and possibly result in cell death either due to viral cytopathic effects or resulting from HIV-specific immune responses. A small clinical trial was set up to test this hypothesis. Vorinostat (VOR), a clinically approved drug for treating certain cancers, has been shown to bind to the active site of histone deacetylases. After a single dose, there was an increase in HIV RNA (1.5 to 5-fold, mean 2.6-fold). Of these subjects, 5 elected to continue with multiple doses. From the 11th to 22nd VOR dose, acetylation of histones and activation of HIV RNA synthesis became refractory to therapy. Also, it is not known what proportion of cells, with latent HIV, can be activated. Whereas a single VOR dose did increase the expression of HIV RNA, this is not an effective therapy for removing the HIV reservoir.

5.2. HIV prevention 2014–2021: managing aspiration and expectation

Myron Cohen, University of North Carolina, NC, USA

Myron noted that there are 2.5 million new HIV infections each year. In this context, anal sex may be an important factor because just one or a few virions of HIV can be infective; within 3 weeks, there is rapid virus replication throughout the body and latent HIV reservoirs of “founder virus” are already formed. Although anal sex has been associated with homosexual couples, Myron pointed out that it is not uncommon amongst heterosexual couples.

Although behavioral education should be encouraged, it can never be the whole answer. Various approaches to the prevention of HIV transmission are being evaluated. Monoclonal antibodies, broad neutralising antibody (bNAB) and vaccines may have potential for prevention of transmission, but most progress is being made with dapivirine rings containing TDF. These are designed to stay in the vagina for a month. Phase III trials are ongoing. A long-acting HIV integrase inhibitor, GSK 1265744 (generally known as GSK 744), is administered i.m. once every 3 months; a two-year safety trial will be required. Phase I trial has been completed and Phase II trial is being planned.

By analogy with tuberculosis therapy, in which the infectious state is disabled prior to a complete cure, one wonders if HIV transmission rates may decrease with effective ART use. In 2005, the HIV Prevention Trials Network (HPTN) initiated a study (HPTN 052) which enrolled 1,763 HIV sero-discordant couples (couples that have one member who is HIV-infected and the other who is HIV-uninfected), mostly (97%) heterosexual couples. The infected partner had to be well enough not to require immediate ART.
The couples were randomised to have either immediate or delayed ART. Both groups received the same care including counselling on safe sex practices, free condoms, treatment for sexually transmitted infections and regular HIV testing.

In May 2011, it had been announced that there had been 27 HIV transmissions in the delayed ART group (877 couples) compared to only 1 in the immediate ART group (886 couples), a 96% reduction. In these 28 cases, the HIV strain was linked to the partner. This is the first randomised clinical trial to show that treating an HIV-infected individual with ART can reduce the risk of HIV transmission to an uninfected partner. Even with “safer-sex” counselling, there were 60 pregnancies in the delayed ART group, despite that group having more incentive for safer-sex. Following the announcement of this result, all infected participants were offered ART.

Myron reported the 10th annual review of this study. In the delayed ART group, there had been a total of 28 cases of HIV transmission with the HIV strain linked to the partner and 11 cases of unlinked transmission. In the one case of HIV transmission in the immediate ART group, infection had been detected at day 85 of the study and further investigation suggested that the infection event was on day 1. Clearly, early ART is highly beneficial. CDC guidelines now recommend that all HIV infected patients should have ART.

6. Mini-symposium: hepatitis B virus

6.1. Hepatitis B treatment: challenges and opportunities

Anna Lok, University of Michigan, MI, USA

The number of people infected with HBV world-wide, as estimated by the WHO and CDC in 2007, was between 223 and 240 million, but was declining due to vaccination. In the USA, vaccine use has led to a steady decline in the rate of new infections, decreasing from about 10/100,000 residents in the 1980s to about 1/100,000 today. In contrast, the prevalence of chronic hepatitis B among immigrants remains high, with no decreasing trend.

When infection is acquired early in life, chronic infection is the norm. High viral load is associated with progression to liver cancer. There are 7 FDA-approved drugs to treat chronic HBV infection, including entecavir (ETV), emtricitabine (FTC) and TDF. With several years of continuous therapy, HBsAg loss is achieved in about 40% of patients but HBsAg loss (the ultimate goal, seen as a “cure”) is still a distant prospect for most patients. However, cirrhosis can be reduced by long-term antiviral treatment. In one TDF trial at 5 years, 344/348 patients had a liver biopsy which showed that 73% of patients had improved fibrosis scores (≥2 units) and that most other patients had no worsening. TDF has now been used for 6 years without detecting HBV resistance, making it one of the first line drugs. TDF is generally well tolerated but its rare side effects include nephrotoxicity (see above for a possible switch to TAF when it is approved), reduction in bone mineral density and very rarely lactic acidosis.

Despite the major progress made in HBV therapy, there remain various challenges. One is cost, about $60,000–$72,000 for 5-year TDF therapy. Pharmacy claims show that adherence is a problem; doses used are less than doses prescribed. There is a lack of accurate prediction of how HBV disease will progress in individuals. HBV DNA can be integrated into the human genome at an early stage of infection. Fortunately, the integrated viral DNA is usually not the complete viral genome and patients, who achieve HBsAg loss, rarely relapse.

Some chronic HBV-infected subjects are co-infected with hepatitis delta virus (HDV). This is a defective virus that replicates only in the presence of HBV. Current antiviral drugs do not inhibit HDV. Recently, heparan sulphate proteoglycan (HSPG) has been shown to be essential for binding both HBV and HDV to primary hepatocytes.

In 2012, human sodium taurocholate co-transporting polypeptide (hNTCP) was identified as a functional receptor for HBV and HDV. hNTCP is also designated as a solute carrier protein 10A1 (SLC10A1). hNTCP was shown to be a binding factor for the preS1 domain of the HBV L envelope protein. This interaction was found to be essential for HBV and HDV infection. Whereas HBV replication is poor in cell lines derived from hepatocytes (e.g. HepG2 and Huh-7) in which hNTCP is usually weakly expressed, HBV replication is possible in primary human hepatocytes. The critical discovery was that over-expression of hNTCP in HepG2 or Huh-7 cells conferred susceptibility to HBV and HDV infection.

Myrcludex-B is a lipopeptide derived from amino acid residues 2–48 of the preS1 region of the HBV L protein. Because it quickly (within 5 min) targets the liver, it is being developed for liver imaging and for drug targeting. It also acts as an entry inhibitor for HBV and HDV by interrupting binding between the HBV L protein and hNTCP. It specifically inhibits hNTCP-mediated taurocholate transport but the effect on HBV replication is much greater. Myrcludex-B activity has been investigated in vivo using SCID mice reconstituted with human hepatocytes. With prophylactic treatment, not one infected hepatocyte was seen. Following therapeutic treatment, at week 6 post-infection, there were a few isolated infected cells. After the end of therapy, the infection seems to spread but only to neighboring cells.

Myrcludex-B has been synthesised on a 100 g scale. Toxicology evaluation in 3 chimpanzees has been completed and clinical trials have been initiated. In a Phase I trial using a 20 mg dose, myrcludex-B was well tolerated. Results of a further Phase I trial are due to be reported later this year (2014). A dose-ranging Phase II trial has been started.

6.3. Immune regulation and co-stimulation in HBV-infected patients: an uneasy truce

Kyong-Mi Chang, University of Pennsylvania, PA, USA

Anti-HBs antibodies clearly play a critical role in controlling HBV disease. Their presence has been accepted as an indication of an “effective cure”. However, these antibodies appear late in the disease course and so they must have a limited role in the early stages of the disease. What is the role of T-cell responses? In contrast to other viruses, there is a delayed onset, about 4–8 weeks rather than days. CD4+ T cells regulate the adaptive response, CD8+ T cells attack HBV-infected cells.

The National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK), part of the USA National Institutes of Health (NIH), is supporting a prospective clinical trial to investigate HBV-specific T cell responses during the course of HBV disease. There are no clear T cell differences relative to HBV genotype. The T cell responses are highest during acute HBV infection. During the chronic phase of HBV disease, T cell responses remain suppressed. In conclusion, there are a lot of players in the immune control of HBV infection but their relative contributions and how they adapt to control HBV replication are still largely unknown.

6.4. Diversifying the hepatitis B pipeline: current efforts to explore novel mechanisms

Andrea (Andy) Cuconati, Institute for Hepatitis & Virus Research, Pennsylvania Commonwealth Institute, PA, USA
Current HBV therapy using nucleotide anti-virals has been highly effective in controlling the infection but a “cure”, as defined by HBsAg seroconversion, has remained elusive. At best after 5 years, the rate is about 25%. Other approaches are needed. Myrcludex-B (see Section 6.2) is the lead entry inhibitor. NVR-1221, an encapsidation inhibitor, is entering clinical trials. In addition, studies with novel nucleotide analogues are ongoing.

The HBV field has been transformed recently by the introduction of cell-based antiviral assays. Stefan Mehrel (see Section 6.2 has been leading the way. The assay read-out will need to be optimized for high-throughput screening (HTS) but, already, the assay has shown some “hits”. A few compounds inhibited encapsidation of viral RNA. (The HBV virion contains partly double stranded (ds) DNA but the reverse-transcription from RNA to DNA occurs within the capsid.) Within the cell, HBV DNA is transported into the nucleus where the viral DNA forms covalently closed circles (cccDNA). Two specific inhibitors of cccDNA formation have been found. Current nucleotide anti-HBV compounds do give large reductions of HBV DNA in plasma but only a minimal reduction in levels of the HBs antigen (about log₁₀0.1). In contrast, one “hit”, HBF-0295 inhibited surface antigen production but not genomic replication. Structure–activity-relationship (SAR) studies have given the current lead compound, HBV-0215. In conclusion, the cell-based assay, with complete replication of HBV, has markedly improved the screening for anti-HBV compounds although further optimization is still needed to give HTS capability.

6.5. HBV capsid protein: biology and potential as a drug target for antivirals

Adam Zlotnick, University of Indiana, IN, USA.

Over the last few years, there has been much progress towards understanding the critical role of the HBV core protein – it is much more than just a protective coat for the genome because it plays a major role in the HBV life cycle. The core protein, being 183 amino acids long, is known as CPr183. The first 149 amino acids are involved in core assembly whereas the last 34 residues, rich in serines and arginines, bind to RNA. Phosphorylation of the serines, particularly S155, S162 and S172, is required for specific packaging of full length HBV RNA complexed to the polymerase (reverse transcriptase – pregenomic RNA; RT-pgRNA).

This RT-pgRNA complex initiates encapsidation. The core consists mainly of CPr183 but also includes other proteins (about 0.5%). Adam showed us a computer model of the core, using different colours to highlight the various critical components. Inside the core, the area of highest density (highlighted in red) represented the polymerase which was attached to the inner surface of the core. The “other proteins” in the core were shown in blue. The current thinking is that the polymerase, initially acting as a reverse transcriptase, is attached to, and guided by, an “inside railway track”. This enables the polymerase to jump to the other end of the RNA to start the reverse transcription into DNA and then jump again to the other end to start, but never complete, the replication of the complementary DNA strand. The self-assembly of the core is an energetically “downhill” process. Somewhat surprisingly, it is possible to get mutations in which the core is even more stable but the RT activity is reduced.

The phenylpropionamide derivative, AT-130, fills a pocket in the core and stabilizes it, similar to the change in amino acids in the mutants. In the presence of AT-130, core assembly occurs faster; hence it is known as a core assembly enhancer (as Adam mentioned, not a term much loved by industry, their preference is for core assembly inhibitors). Regardless, the whole capsid structure changes. The binding of only a few drug molecules is required to make the core non-functional. It seems that it is easier to find compounds to enhance core assembly than inhibitors.

6.6. Targeting cccDNA to cure chronic hepatitis B

Massimo Levreto, Sapezina Universita’ di Roma, Italy.

The current HBV therapies of choice are TDF alone or with ETV. These drugs have an extensive safety record with use up to 7 years. However, as for other nucleoside/nucleotide analogs, there is only a limited (about 1 log₁₀) reduction in the levels of HBV cccDNA. The half-life of HBV cccDNA seems to be long, but is still unknown.

HBV replication parallels host gene expression, in that they involve the acetylation of histones, for example H3 and H4. Both host transcription factors and viral proteins bind to the cccDNA. Massimo summarized various assays to study different stages of cccDNA during the replication cycle. Potentially, these assays would allow the study of various approaches: to reduce or clear cccDNA, to silence cccDNA or to prevent the formation of new cccDNA so that it would eventually be removed by dilution and cell death.

For proof-of-concept, known “epigenetic” compounds, which act as transcription inhibitors, have shown that cccDNA can be silenced. By reducing histone acetylation, the cccDNA becomes too compact to allow transcription. This approach mimics, partly, therapy with interferon. This research is still at an early stage.

6.7. Animal models of hepatitis B disease

Due to time constraints, the next two speakers were asked to present brief summaries.

John Morrey (Utah State University, UT, USA) described four mouse models but all stages of the life cycle of HBV can be studied only in the chimeric mouse model, in which human hepatocytes are used. However, this model lacks the potential to study the immune system and it is very expensive.

Stephan Menne (Georgetown University, DC, USA) described the woodchuck model. Woodchuck hepatitis virus (WHV) resembles the human virus and the disease in animals has many similarities to that in humans. Neonatal infection becomes chronic in about 60–75% of cases. These chronic cases have virtually a 100% life-time risk of developing cancer, the time scale being about 1 year of chronic infection, followed by cancer at years 3 to 4.

7. Mini-symposium: Research Triangle Park

7.1. Biophysical mechanisms and methods of evaluation in HIV prevention science

The use of microbicides is an active area of research for the prevention of transmission of HIV. David Katz (Duke University, NC, USA) described how mathematical models may aid drug product design. For example, if it is assumed that the microbicide gel is 400 microns thick, the epithelium is 200 microns and the stroma (connective tissue) is 3000 microns and if the partition coefficient between gel and epithelium in known, then it is possible to model drug transfer and suggest how various other parameters, for example the size of the subject, may modify drug delivery.

It is important that different disciplines work together, for example biophysicists with behavioral scientists. Biophysics can help an understanding of complex physical phenomena but human behavior can be both complex and highly variable.

7.2. Novel animal model platforms of human disease

Ralph Baric (University of North Carolina, NC, USA) noted that a particular infective agent, for example norovirus (NoV), may cause subclinical or serious disease in different individuals. In general, animal models are designed to give consistent outcomes rather
7.3. Did we put the cart before the horse? Clinical pharmacology insights into HIV prevention trial outcomes

Angela Kashuba, University of North Carolina at Chapel Hill, NC, USA

In four clinical studies, Truvada [a combination pill containing TDF and emtricitabine (FTC)] was taken once daily to prevent HIV transmission, known as pre-exposure prophylaxis (PrEP). The adherence rates were unexpectedly poor in all four studies, particularly low in the study including at risk women. For example in one study, "high adherence" was defined as subjects taking at least 80% of drug doses and was achieved by only 54% of subjects. Possible reasons may have been the apparent risk of side-effects (the long consent form included 7 pages of side-effects) and the perception that the subjects, as individuals, were not particularly at risk of infection by HIV. Importantly, the trial did confirm the concept that PrEP could be effective. There was >90% protection in those subjects generally taking 7 doses/week and there was some protection, albeit much less, in subjects taking 2 doses/week.

Adherence rates, reported by subjects, were appreciably higher than the rates evidenced by drug blood level measurements taken just before the next dose (i.e. 24 h after previous dose). In an attempt to better understand and model these data, the drug concentrations (TDF/TFV and FTC) in various tissues were measured. The ratio between drug concentrations in blood and tissue samples was measured. For FTC, the ratios were 2.6 and 1.3, respectively. The ratio between drug concentrations in blood and vaginal tissue samples was 0.2 in vaginal tissue. For FTC, the ratios were 2.6 and 1.3, respectively.

When considering the possible consequences of missed doses, the time scale for HIV infection is an important factor. It is thought that HIV takes about 1–3 h to reach the epithelial cells. Clearly, adherence is a critical factor for efficacy and so a real-time objective method for measuring adherence is urgently needed before further clinical studies are initiated.

7.4. The novel nucleoside analog BCX4430 exhibits broad-spectrum antiviral activity and confers post-exposure protection against Ebola and Marburg viruses

Travis K. Warren, USAMRIID, Fort Detrick, MD, USA

Ebola and Marburg viruses are members of the filovirus family. Even in recent outbreaks of these diseases, including the current Ebola epidemic in West Africa, care workers are becoming infected and dying. Drugs, which are being investigated for treating these diseases, are progressed under the FDA “Animal Rule”.

BCX4430 is a C-nucleoside adenine analog (Fig. 10) which is being progressed by BioCryst Pharmaceuticals Inc. (Warren et al., 2014). In cell culture assays, BCX4430 is active against Ebola and Marburg viruses, (EC50 ca 1 µM). With BCX4430 at 30 µM, there was no detectable incorporation into host DNA or RNA. In rats, BCX4430 is efficiently activated (phosphorylated) to the triphosphate. In a primer-extension assay, there is some read-through beyond a single residue of BCX4430, but there is effective chain termination after the first BCX4430 residue where the template has two consecutive uridine residues.

BCX4430 has been tested in rodent and nonhuman primate models of Marburg hemorrhagic fever. In mice, there was a dose response (30, 20, 11 and 1.1 mg/dose, bid) with full protection at the two higher doses (survivors, 100%, 100%, 95% and 83% respectively). In an experiment with dosing starting at different times (4 h pre-infection, 24, 48, 72, 96 and 120 h post-infection vs placebo), the placebo-treated mice died on days 6, 7 and 8 with one survivor (10%). In the treated groups, the percent survival was 80, 100, 80, 100, 100 and 30, respectively. In guinea pigs, BCX4430 (bid) with treatment starting at different times (1 h pre-infection, 24, 48 and 72 h post-infection) there was full protection (100% survival) for the pre-infection and 24 h groups, with reduced efficacy at the later start times.

In cynomolgus monkeys, BCX4300 treatment was started at 1, 24 and 48 h post-infection. In the placebo group, all 6 animals died within days 9 to 12. In all the treated groups, virus loads were reduced by more than logarithm. There was one late death in the 1 h group but the other 17 monkeys survived. Various markers of potential organ damage were reduced in all treated groups.

Encouraged by these results, 14-day toxicology trials have recently been completed without any serious concerns. BioCryst is developing BCX4430 under the FDA Animal Rule and IND-enabling work is ongoing.

When asked about viral resistance, Travis explained that it is not ethically permissible to create resistant strains of Marburg virus, but samples collected from the monkeys are being sequenced to look for mutations indicative of drug resistance. As yet, mitochondrial toxicity has not been examined.

8. Challenges in HIV infection, treatment and prevention

8.1. Can we cure HIV infection?

Mario Stevenson, University of Miami, Miami, FL, USA

Even after successful and prolonged ART, invariably plasma HIV load increases within 20 days of stopping therapy. Of all the millions of HIV-infected people, there has been only one documented cure – the “Berlin” patient (see above). Two Boston patients, who
had similar bone marrow transplants, initially seemed to have been “cured” but HIV was detected after 70 and 200 days, respectively.

Latent HIV can survive in various long-lived cells for decades, especially in memory T cells. When these cells proliferate, the integrated HIV genome is duplicated as the cell divides and the cells survive so long as HIV remains silent. Compounds known to activate all T-cells are too toxic to become a clinical therapy. However, latency-reversing agents (LRA) have greater specificity, ideally activating only the integrated HIV, leading to the death of HIV-containing T-cells. There remains another possibility (perhaps a less popular view) that there is continued low rate of HIV replication. Two clinical studies have been initiated in subjects with undetectable plasma HIV levels. Raltegravir, an HIV integrase inhibitor, was added to the background therapy. Latent HIV is mostly integrated into host DNA but HIV may also form episomal circular DNA. The proportion of the circular form increases with raltegravir treatment. In the two clinical studies, 13/45 and 9/15 subjects, respectively, had detectable HIV circles which then decayed. This implies that some de novo infection of cells is ongoing. On the other hand, ART works well, with no evidence of sequence evolution in the HIV circles at 48 weeks. Is it possible that raltegravir is inducing a single round of HIV replication, to give an increase in HIV circles?

8.2. Potential therapeutic approaches for the cure of HIV infection

Derek Sloan, Gilead Sciences, Foster City, CA, USA
Like vorinostat, (VOR), romidepsin (RMD) is a histone deacetylase inhibitor which is used clinically to treat cancer. Memory CD4+ T cells were taken from HIV subjects on suppressive ART; ex-vivo treatment with RMD (40 nM) induced a 6-fold increase in intracellular HIV RNA which persisted for 48 h. In contrast, a much higher treatment with RMD (40 nM) induced a 6-fold increase in intracellular HIV RNA and virions. Encouragingly, this ex-vivo induction of latent virus was seen at RMD concentrations that are below the levels of drug achieved in humans by clinical doses of RMD. Accordingly, in a Phase I/II trial in HIV-infected subjects on ART, RMD gave a better and more sustained response than VOR. About 1.5% of cells containing HIV provirus were activated. Although this is far too low a percentage to eliminate the latent HIV reservoir, it is hoped that combination of such LRA, which give improved results in ex-vivo cell assays, may give better clinical efficacy.

Gilead scientists have started screening for novel LRAs. “CS-1” has been identified as a hit by HTS. Research on this lead is at a very early stage. Gilead workers are also investigating other approaches. For example, GS-9620 is a Toll-like receptor 7 (TLR7) agonist and it acts as an immune stimulator. Although it is being evaluated in Phase II studies for the treatment of chronic HBV infections, the potential effect on HIV reservoirs is being investigated. In SIV-infected monkeys, oral dosing of TLR7 agonist induced the activation of immune effector cells such as CD8+ T cells and NK cells. Based on these data, TLR7 agonists are being further investigated for their effect on latent SIV reservoirs in monkeys which have good virological suppression. Another approach is to use anti-envelope antibodies. Broadly neutralising antibodies (bNAbs) are very effective in preventing SIV infection when the viral load is low but less effective against a high-load virus challenge. In addition, a prophylactic CMV-vector-based SIV vaccine was effective in preventing SIV infection in rhesus monkeys. This and similar vaccines are being tested in vivo for their effects on the latent SIV reservoirs.

In summary, LRAs are able to activate HIV provirus in memory CD4+ T cells and thereby may enhance the recruitment of immune effector cells to destroy provirus-containing cells. However, a “cure” for HIV infection is still a distant prospect. Furthermore, latent HIV reservoirs are heterogeneous and so a combination of approaches will likely be required.

8.3. Animal models of HIV infection

Gerardo Garcia-Lerma, Centers for Disease Control and Prevention, Atlanta, GA, USA
Proof-of-concept studies for PrEP, are mostly conducted in non-human primates. These can be used either to model a single high-dose infective challenge or repeated low inoculations, about 10–50 tissue culture infective doses (TCID50).

Since 2005, rhesus macaque models have been used in a long series of investigations. In a study, in which the monkeys were treated daily with either oral TDF or TDF/FTC and given a weekly SIV inoculum rectally, TDF/FTC gave a longer delay in infection than did TDF alone. When using the vaginal infection route, TDF/FTC gave 100% protection. In contrast, there was far less protection in clinical trials – why? One possible reason may have been that women were having the contraceptive injection, depot medroxyprogesterone acetate (DMPA). A study, in macaque monkeys given DMPA, confirmed that dosing with TDF/FTC gave good drug levels in plasma and in vaginal secretions. Therefore, this did not explain the poor protection in the clinical trial.

The macaque model has been used successfully to investigate various situations that are presented in the clinic. When macaques were re-infected with SIV and a bacteria and treated with TDF/FTC for 12 weeks, there was good, but not complete, protection (80%). With FTC-resistant virus, TDF/FTC remained protective. In this case, FTC-resistant virus has increased susceptibility to TDF. With the K65R mutant HIV, there was protection against a low inoculum but only partial protection (ca 50%) against a high inoculum.

Whereas daily dosing seems to be acceptable for patients living with HIV, another option for PrEP is desirable. GSK-1265744 (generally known as GSK-744) is an HIV integrase inhibitor. It can be formulated with nano-particles to provide an injectable drug depot. In the macaque model, GSK-744, injected once monthly, gave full protection against repeated rectal and vaginal exposures. Because metabolism of GSK-744 is much slower in humans than macaques, it was expected to remain effective in humans for up to three months. A Phase I study confirmed that drug levels remained above the predicted effective level with a 20-week dosing interval. A Phase II trial is planned.

Another approach is to use vaginal rings, which have been in clinical use as contraceptive devices for years. In the macaque model, TDF-containing rings, replaced every 4 weeks, gave full protection. A Phase III trial has just been initiated. Another option, elvitegravir (EVG) and TAF are being evaluated in a biodegradable depot. Although daily dosing with TDF/FTC has not proved sufficiently successful as PrEP in clinical use, it has proved that PrEP is an achievable aim and this has encouraged the progression of other options.

8.4. Monitoring HIV drugs and viral reservoirs

Courtney Fletcher, University of Nebraska, Omaha, NE, USA
Atripla was the first triple combination pill taken once daily for HIV therapy. It contained TDF, FTC and efavirenz (EFV). The macaque model has been used to investigate the differing tissue distributions of these drugs and how viral replication may be continuing wherever the drug concentrations are lowest. There are two approaches: tissue homogenates and tissue cells. Tissue homogenates give both the intracellular and extracellular drug amounts. From tissues, mononuclear cells (MNCs) are collected and the intracellular drug concentration measured. This approach is preferred by Courtney but this option may be constrained by sample size and the drug concentration may be underestimated. For exam-
ple, with raltegravir, after the MNCs have been washed 3 times, the drug concentration is very low. Much higher raltegravir concentration is maintained in lymphoid tissues, with raltegravir, after the MNCs have been washed 3 times, the potential for therapeutic convenience and safety of drugs has markedly improved both the efficacy and safety of drug delivery. A drug may be absorbed from the gastrointestinal tract either going via the portal vein to the liver and then into blood circulation or via the lymphoid system. Blood flow is about 200 times faster than lymphoid flow. When the water/1-octanol partition-coefficient (logP) of a drug is <5, absorption tends to be via the blood route. The produrg approach can be used to alter absorption or, as for TFV, stability of the produgs (TDF and TAF) can influence the relative concentration in lymphoid tissues (see above).

9. Conclusion

This year, the three major award lectures exemplified the strength of ICAR, covering very different areas of research. John Drach (Elion Award) described his journey through the early days of antiviral research, which led to the identification of novel modes of antiviral action that had not been envisaged previously. Piet Herdewijn (Holý Award) used evolutionary pressure to select DNA polymerases that accept novel nucleoside analogs. The replacement of thymine by 5-chlorouracil led to the generation of a new form of E. coli. I suggest that this work has important implications in conventional antiviral research. With HIV and HCV protease inhibitors, the genetic barrier is limited by the ability of the viral protease and its substrate (the viral polypeptide cleavage sites) to co-mutate so that the virus can become resistant to the antiviral drug. So far, polymerase inhibitors have not suffered the same fate but this work shows that a poor choice of nucleotide analog could result in a resistant virus with a new type of RNA in which the drug replaces a natural nucleoside. Adrian Ray (Prusoff Award), describing work at Gilead, demonstrated how the prodrug concept can markedly improve both the efficacy and safety of potential drugs. Their progress with HIV and HCV therapies has been remarkable.

The keynote addresses tackled two emerging areas of HIV research. David Margolis summarized work aiming to eradicate HIV from infected subjects and Myron Cohen described current progress with approaches to prevent HIV transmission. I found both these presentations to be informative and stimulating. HIV “cure” still seems to be a distant prospect. In contrast, prior to exposure prophylaxis (PrEP) has been shown to be an achievable aim although the need for daily dosing is a barrier to success. Gerardo García-Lerma described recent progress which is likely to radically change the prospects for therapeutic convenience and success. TDF-containing vaginal rings, which need replacing only once a month, are being evaluated. Another exciting prospect is GS-744 which has been formulated as a long-lasting injection. A Phase 1 trial confirmed that the drug may be administered at 3-month intervals. In the absence of a proven HIV vaccine, PrEP with drugs has become the most promising strategy to reduce HIV infection rates among high-risk populations.

This conference also included three interesting mini-symposia: “Hepatitis B virus”, “Research Triangle Park” and “Challenges in HIV Infection, Treatment and Prevention”. An innovation this year was a session devoted to the European Training Network, EUVIRNA and introduced by Frank van Kuppeve. All the EUVIRNA fellows, who attended ICAR, gave short presentations at this session. For further information, please see the ISAR News (24.1) in the September issue of Antiviral Research for an account by Frank van Kuppeve. For many years, the clinical symposium was, for me, a major highlight of ICAR. In my report for the 2013 ICAR, I expressed a hope regarding HCV therapy: “There is the prospect that the first nucleotide analogue will be licensed by the time of our next ICAR meeting. The combination of a nucleotide analogue and a NS5A inhibitor looks set to transform HCV therapy across all genotypes. As for HIV, single-pill, once-daily regimens are following on quickly”. On 6th December 2013, Sofosbuvir (Sovaldi®) was the first nucleotide analogue to be approved in the USA by the Food and Drug Administration (FDA) for treatment of patients with HCV. Approval by the European Union followed soon afterwards, in January 2014. A NS5A inhibitor, ledipasvir, formulated as a single fixed-dose combination pill with sofosbuvir, is progressing quickly through clinical trials. With such remarkable progress being achieved since the 2013 ICAR, I was disappointed to discover that there was no presentation on this topic at this year’s ICAR.

A paper (Sofia, 2014), which was part of a symposium in Antiviral Research on “Hepatitis C: next steps toward global eradication”, emphasizes recent successes. After completing therapy, a sustained virological response for 12 weeks (SVR12) is regarded as a cure for HCV-infected patients. The combination of sofosbuvir/ledipasvir has shown remarkable results in clinical trials, with SVR12 in the range 95–100% across genotypes. This combination was well tolerated. A NDA for the sofosbuvir/ledipasvir combination pill was submitted recently. I do not recall any previous antiviral trials in which the “intention-to-treat” analyses showed 100% success rates.

Perhaps similar to the HCV symposium in Antiviral Research, I hope that the 2015 ICAR, which will be held in Rome, will have a mini-symposium which will include an account of this remarkable progress. It would be interesting to have an update on the clinical impact of this combination therapy for HCV and to have an assessment on the prospects for global eradication of HCV. Beside this one disappointment, there were many excellent presentations and I would like to add my thanks to the ISAR Officers and Conference Committee for organizing another interesting and successful ICAR.

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References

Murakami, E., Wang, T., Babusis, D., Lepist, E-I., Sauer, D., Park, Y., Vela, J.E., Shih, R., Birkus, G., Stefanidis, D., Kin, C.I., Cho, A., Ray, A.S., 2014. Metabolism and pharmacokinetics of anti-hepatitis C virus nucleotide prodrug GS-6620. Antimicrob. Agents Chemother. 58, 1943–1951.

Sofia, M.J., 2014. Beyond sofosbuvir: What opportunity exists for a better nucleoside/nucleotide to treat hepatitis C? Antivir. Res. 107, 119–124.

Vere Hodge, R.A., Sutton, D., Boyd, M.R., Harnden, M.R., Jarvest, R.L., 1989. Selection of an oral prodrug (BRL 42810; famciclovir) for the antivirally active agent BRL 39123. [9-(4-hydroxy-3-hydroxymethylbut-1-yl)guanine; penciclovir]. Antimicrob. Agents Chemother. 33, 1765–1773.

Warren, T.K., Wells, J., Panchal, R.G., Stuthman, K.S., Garza, N.L., et al., 2014. Protection against filovirus diseases by a novel broad-spectrum nucleoside analogue BCX4430. Nature 508, 402–405. http://dx.doi.org/10.1038/nature13027.