



ISAR News

Newsletter of the International Society for Antiviral Research

Editor Anthony Vere Hodge Guest editor Graciela Andrei
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Report on the 30th ICAR Atlanta, GA, USA

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ISAR PRESIDENT'S MESSAGE

(José Esté)

It is with great pleasure that I salute all ISAR members and friends in this new issue of the ISAR News.

Our 30th ICAR in Atlanta was a success in many ways. We received excellent comments and congratulations for the excellence and quality of the invited speakers, and many oral and poster presentations. ICAR retains its flavor and personality, providing an interdisciplinary forum at which investigators involved in basic, translational, and clinical research worldwide

meet to review recent developments in all areas of antiviral research, drug and vaccine development. As mentioned below, a new format of the ICAR report has been published in Antiviral Research with highlights and reviews of most oral presentations. Additionally, satellite activities such as the Women in Science Roundtable, the Career Development Panel and the New Member and First Time Attendee luncheon (The Happy Hour) provided an opportunity to discuss other issues of relevance. I take the opportunity to thank again the co-chairs of the Program Committee, Justin Julander and Mark Prichard, and all committee chairs and members for their unselfish commitment to organize the 2017 conference.

The 30th ICAR meant a challenge to the Society. With so many different competing conferences and meetings to attend and a long economic crisis of which scientific research did not escape, ISAR has gone through great financial efforts to continue supporting the participation of students, postdocs and young investigators. We are happy to learn that the 30th ICAR breaks a trend. The meeting becomes financially sound once again. We are indebted to all corporate sponsors, friends of the Society and ISAR members for their contribution. We are encouraged by their financial support to continue organizing the best antiviral meeting possible. Excellence in science and support to young investigators will continue driving our efforts.

We are beginning to organize the 31st ICAR that will be held in Porto, Portugal. The conference will begin on Monday, June 11 and will conclude on Friday, June 15, 2018. These dates follow the end of the academic year

30th ICAR Corporate and Educational Sponsors. PLATINUM: Gilead Sciences, GOLD: Alios BioPharma. SILVER: AbbVie, Chimerix, JCR Pharmaceutical Co. Ltd., Southern Research Institute. BRONZE: ACS Infectious Diseases, Antiva Biosciences, Center for Drug Design – U of Minnesota, Elsevier B.V., Institute for Antiviral Research, Utah State U, Oxeltis, Riboscience LLC, Toyama Chemical Co., XpressBio. THE ISAR PRESIDENTS' FUND: Jan Balzarini, Joseph M. Colacino, José A. Esté, Phillip Furman, Doug Richman

in the U.S., allowing hopefully, greater participation.

Porto has been selected for its easy access by flight from most European cities and major U.S. hubs, but also because we were dazzled with its warmth and beauty. Porto is a city full of tradition and culture but modern and exiting.

After many years organizing the meeting in a hotel venue, the 31st ICAR will be held at the Alfândega Congress Centre (<http://www.ccalfandegaporto.com/en>). We hope to free ICAR participants from the constraints of staying at a venue hotel, allowing everyone to choose the accommodation that suits their needs. Being a popular tourist location, Porto has many types of accommodations to offer, from luxury hotels to small but comfortable hostels. ISAR will block a limited number of rooms at different hotels so I encourage everyone to make timely reservations for their stay in Porto.

In 2018, we will continue supporting the participation of students, postdocs and young investigators. Our travel support will be solely based on merit and excellence of the abstracts submitted for presentation. I encourage those wishing to apply to submit their best science. Quality will be the prevailing criteria to receive financial support. Keep an eye on further instructions on how to submit your abstracts at the ISAR-ICAR website <http://www.isar-icar.com>.

I kindly remind all our members that the call for nominations for the ISAR awards is now open and encourage you to nominate a deserving colleague. The ISAR Chu Family Foundation Scholarship for Women Scientists is also now open for applications. Awards will be given to advance the careers of women with potential for significant contribution in the field of antiviral research. Information on how to submit nominations is posted in our website or by requesting information through the Society's email address shown below.

As we are in an early stage of organizing the 31st ICAR, I call on all those interested in participating to let us know your ideas, comments or suggestions. If you have a particular interest or a topic that should be highlighted, please feel free to contact us and let us know at info@isaricar.com.

MEETING REPORT: 30TH INTERNATIONAL CONFERENCE ON ANTIVIRAL RESEARCH, Atlanta, USA

(R. Anthony Vere Hodge)

The 30th International Conference on Antiviral Research (ICAR) was held in Atlanta, GA, USA from May 18 to 21, 2017. This was the second ICAR at Atlanta, the first being the 10th ICAR in 1997. A meeting report, in which eight volunteer rapporteurs and ISAR President provided their views on the

highlights of the 30th ICAR, has been published in *Antiviral Research*. This report aims at effectively convey the speakers' goals, results and conclusions of their talks. The eight volunteer rapporteurs were Graciela Andrei, Kara Carter, Zlatko Janeba, Aruna Sampath, Luis M. Schang, E. Bart Tarbet, R. Anthony Vere Hodge, and Mike Bray.

We thank all the speakers and reviewers who provided content, figures and comments to the final report.

Reference for meeting report

Graciela Andrei, Kara Carter, Zlatko Janeba, Aruna Sampath, Luis M. Schang, E. Bart Tarbet, R. Anthony Vere Hodge, Mike Bray, José A. Esté.

Highlights of the 30th International Conference on Antiviral Research. (2017)

<https://doi.org/10.1016/j.antiviral.2017.07.017>

ISAR BUSINESS MEETING REPORT

(Brian Gowen/ Graciela Andrei)

The ISAR held its annual business meeting at the Atlanta Hilton during the 30th ICAR on Wednesday, May 24. Brian Gowen (Treasurer) presented a brief summary of the Society's finances. Net assets at the end of the 2016 fiscal year was \$633,397.90 from several bank accounts, two investment accounts, and a CD, which matured in August of 2016 (Table 1). The net assets total is down from \$674,571.24 at the end of the 2015 fiscal year and is largely due to the \$29,191 deficit from the 29th ICAR held in La Jolla, CA (see ISAR News Vol. 26.3).

Although there has been a downward trend in the Society's holdings (Figure 1), projections from the review of revenue and expenses for the 2017 ICAR are favorable. We will not know the final numbers until late July or August when all revenue support has been received and meeting expenses paid. This information will be provided in an upcoming issue of ISAR News. Also presented during the business meeting was the year-end financial statement reflecting ISAR's revenue sources and expenses for 2016 (Table 2). The largest sources of revenue are the support we receive from our generous sponsors and the conference registration fees, with the major annual expense being the annual ICAR.

Graciela Andrei (Secretary) provided a report on the 2017 ISAR Membership and on attendance at the 30th ICAR in Atlanta (Figure 2). Twenty-one countries are represented in the Society, with a total of 238 members through May 2017 (138 from USA). A total of 244 attendees as of May 24, 2017 from 23 different countries were registered for the 30th ICAR.

This year, the Society received 54 applications for a travel grant award. Because of the restricted budget available, some of the grant applications could not be

funded. Based on the scientific review of the submitted abstracts, a ranking was established based on the scores provided by four independent reviewers. The Society awarded 24 Travel Merit Grants (9 from North America, 11 from Europe and 4 from Asia and Australia who received an award of \$500, \$800, and \$1000, respectively) to help these members defray the costs of attending the conference.

The Society also awarded 15 Travel Merit Assistance Awards to participants coming from low/middle-income countries to support their attendance to the meeting. For recipients of this award, besides a \$1000 stipend, the registration fee was waived. As shown in Figure 3, the Society invested a total of \$32,300 in Travel Awards this year, which highlights the considerable funds made available by the Society during the past years to increase the attendance of young investigators at the meetings.

Table 1. Fiscal year 2016 net assets

Assets	
Bank Accounts	\$ 287,016.18
CD @ Wells Fargo	\$ 107,974.89
Cetera Investments	\$ 64,821.62
Fidelity Investments	\$ 173,585.21
TOTAL	\$ 633,397.90
Liabilities	
Accounts Payable	\$0
TOTAL	\$0

Figure 1. Summary of recent year-end ISAR net assets

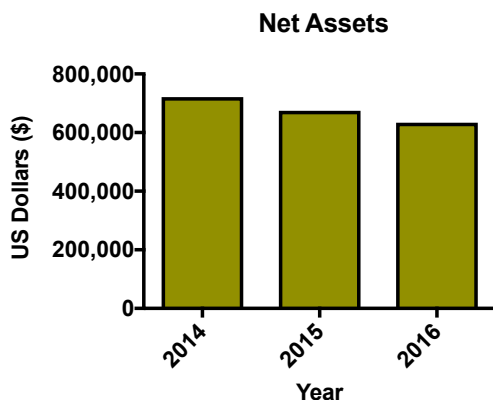


Table 2. Year-end financial statement for 2016

2016 Income	
Membership Dues	\$ 9,250.00
Sponsorship Support	\$ 158,309.50
TCFF WIS Sponsorship	\$ 10,000.00
Registrations for ICAR	\$ 125,300.00
Gain/Loss Investments	\$ 13,637.51
Total	\$ 316,497.01
2016 Expenditures	
Administrative	\$ 17,034.04
WIS Expenses	\$ 12,292.11
ICAR 2016	\$ 313,811.09
ICAR 2017	\$ 9,538.60
Donations	\$ 255.00
ISAR Management	\$ 3,362.12
Total	\$ 356,292.96
Net Income	\$ (39,795.95)

POSTER AWARDS

(Kathie Seley-Radtke)

This year's ICAR poster competition in Atlanta was, as always, fierce and highly competitive! The international team of judges included Graciela Andrei, John Bigger, Andrea Brancale, Jinhong Chang, Cyril Dousson, Joana Rocha-Pereira, Brian Gentry, Zlatko Janeba, Brent Korba, Chris Meier, Jennifer Moffat, Roger Ptak, Luis Schang, Enzo Tramontano and the Chair of the poster award committee, Kathie Seley-Radtke.

Fifty-four posters registered to be judged in the three categories: undergraduate/graduate student, postdoctoral researcher and young investigator. Each category had many excellent posters in the running for both poster prizes and the chance to give a shotgun talk at Wednesday's opening session, which was co-Chaired by Kathie Seley-Radtke and her graduate student Therese Ku.

The shotgun winners were:

- Poster #97, Matthias Winkler "Membrane-permeable Nucleoside Triphosphate-prodrugs against Hepatitis C Virus"
- Poster #95, Sietske Speerstra "Broad-Spectrum Antiviral Molecules with Biophysical Mechanisms of Action"
- Poster #21, Dr. Marko Zivcec, "Evaluation of cross-strain neutralizing potency of monoclonal antibodies against Crimean-Congo hemorrhagic fever virus"
- Poster #101, Makda Gebre, "Comparison Between Various Strains of Chikungunya in Disease Phenotype and Response to Antiviral Treatment"
- Poster #36, Cynthia Matthew, "Selective Inhibitor of Nuclear Export (SINE)

- compounds Reduce RSV Replication in vitro”
- Poster # 96, Dr. Marcella Bassetto, “Identification of a Substituted Thienopyrimidine Scaffold with Antiviral Activity against Zika Virus”.

The winner of the Young Investigator category of the poster competition was:

- Leen Delang from Rega Institute with Poster #159, “Can antiviral drug-resistant chikungunya virus be transmitted by mosquitoes?”

Delang won \$1000 cash and a bonus prize - all publication fees waived by Antiviral Chemistry and Chemotherapy for a paper submitted to the journal.

We saw a tie for first place in the postdoctoral category:

- Poster #96 (\$1000), Dr. Marcella Bassetto from Cardiff University,
- Poster #109, a jointly presented poster, “Successful Design of Ribonucleoside Di- and Triphosphate Prodrugs to Improve the Anti-Influenza Virus Activity of T-705 and its Analogue T-1105” from Evelien Vanderlinden (Rega Institute) and Johanna Hutchting (University of Hamburg), who split the \$1000 prize.

It is notable that the research presented in poster #109 was the result of Hutchting’s three month sabbatical in Lieve Naesens’s laboratory where she worked with Vanderlinden learning how to run assays on her nucleosides and nucleotides. Hutchting’s sabbatical was paid for by generous support from The Chu Family Foundation Early Career Women Scholarship from our sister Society, The International Society of Nucleosides, Nucleotides & Nucleic Acids (IS3NA). The second place winner in the Postdoctoral category was Dr. Marko Zivcec (CDC) with Poster #21, who received \$500.

Finally, in the graduate student category, we had a five-way tie for first place between:

- Poster #49, Tiffany Edwards – “Inhibition of Hepatitis B Virus Replication by N-hydroxyisoquinolinediones and Related Polyoxygenated Heterocycles”
- Poster #63, Edurne Garcia-Vidal – “Evaluation of the Innate Immune Modulator Acitretin as a Novel Strategy to Clear HIV Reservoir”
- Poster #13, Therese Ku – “Synthesis of Flexible Purine Analogue Inhibitors of NCp7”
- Poster #95, Sietske Speerstra
- Poster #97, Matthias Winkler.

Each graduate student category winner received a prize of \$500 cash.

The poster committee would like to thank all the presenters for the excellent research presented and we look forward to judging next year’s entries in Porto!

WOMEN IN SCIENCE AND CHU FAMILY FOUNDATION SCHOLARSHIPS (Rhonda Cardin)

WIS Roundtable (Karen Buckheit)

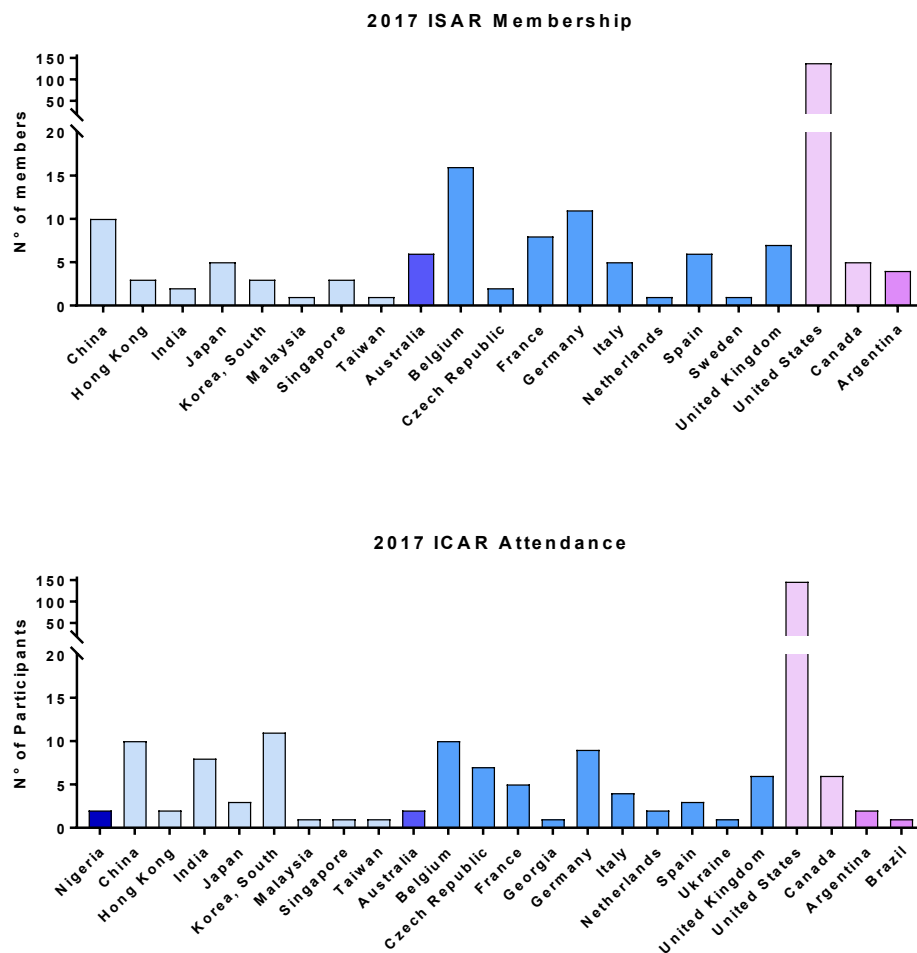
The 5th Annual Women in Science Roundtable, the opening event of the 30th ICAR in Atlanta, Georgia, featured a panel discussion where participants engaged in interactive discussions with four leading women scientists in their respective fields that included Priscilla Yang (Harvard Medical School), Inger Damon (CDC), Julie Dyal (NIAID, NIH and Tunnell Consulting Company) who were introduced by Rhonda Cardin (Louisiana State University). Each of the panel members shared personal and professional experiences as well as opportunities and challenges that they have encountered during their scientific careers. They brought forth many and varied viewpoints that provided food for thought for the participants provoking insightful questions leading to an enthusiastic discussion. We appreciate the panel members and participants for attending the event and look forward to seeing everyone at the 6th Annual Women in Science Roundtable in Porto.

2017 WIS Speaker (Rhonda Cardin)

In Atlanta, the WIS Committee was pleased to present Priscilla Yang (Harvard University) as the first WIS sponsored Speaker in recognition of her major contributions to antiviral research and leadership in mentoring young women. Yang presented her research on “Small molecule inhibitors of viral entry: pharmacological mimicry of the humoral immune response to viral infection” in the Antiviral Immunity Symposium. We hope that this will be the first of many invited speakers to highlight the research accomplishments of women in science.

2017 Chu Family Foundation Awards (Amy Patick)

Thanks to a generous donation from the Chu family, we are pleased to announce the 2017 Chu Family Foundation (TCFF) Awards for Women Scientists. The TCFF Awards support the professional development of women with potential to make significant contributions to the field of Antiviral Research by providing funds to attend a conference, visit another laboratory, take a course, or acquire specialized training. These awards are given annually at the ICAR meeting. Each award

Figure 2. 2017 ISAR Membership and ICAR Attendance**Figure 3. 2017 ICAR Travel awards**

	Travel Merit Awards		Travel Merit Assistance Awards	
	Region	Awards made (stipend)	Region	Awards made (stipend)
	North America	9 (\$500)	Low/middle-income countries	15 (\$1,000)
	Europe	11 (\$800)		
	Asia & Australia	4 (\$1,000)		
Total		24 (\$17,300)	15 (\$15,000)*	

* Registration fee was waived, representing \$4,500



consisted of a \$1500 stipend, a 2-year ISAR membership and a commemorative certificate. In 2017, we had an overwhelming response with many excellent candidates and conducted a rigorous process to select the candidates.

We congratulate the seven awardees, as well as all women who applied, and wish them success in their future careers. The awardees are:

1. May Wang (graduate student; Boston, MA, USA) for attendance at the 30th ICAR (Atlanta)
2. Naphak Modhiran (graduate student; Annerley, Australia) for attendance at The 17th International Congress of Virology
3. Angela Corona, Ph.D. (postdoctoral candidate; Monserrato, Italy) for research internship in Barcelona, Spain
4. Makda Gebre (graduate student; Logan, Utah, USA) for attendance at the 30th ICAR (Atlanta)
5. Jessica Von Recum, Ph.D. (postdoctoral candidate; San Diego, CA, USA) for attendance at 30th ICAR (Atlanta)
6. Verónica Mara Quintana (graduate Student; Buenos Aires, Argentina) for a research internship in Marseille, France
7. Linlin Zhang (graduate Student; Lubeck, Germany) for attendance at the 30th ICAR (Atlanta)

CURRENT RESEARCH

Challenges for research and development of dengue antivirals in the era of Dengvaxia®

Jenny Low^a and Subhash Vasudevan^b

^aDepartment of Infectious Diseases, Singapore General Hospital, 20 College Road, Singapore, 169856. jenny.low@singhealth.com.sg

^bProgram in Emerging Infectious Diseases, Duke-NUS Medical School, 8 College Road, Singapore 169857 subhash.vasudevan@duke-nus.edu.sg

Dengue fever disease is caused by the four serotypes of dengue virus (DENV 1-4) and is mostly transmitted by the urban dwelling *Aedes aegypti* mosquito. Close to 50% of the global human population live in dengue endemic areas and are at risk of contracting the disease. Poorly controlled urban development and vector spreading, which is thought to be associated with global warming, are possible contributors for the expansion of dengue (Gubler, 2006). It was estimated that around 100 million symptomatic dengue cases occurred in 2010 out of a total of nearly 400 million infections (Bhatt et al., 2013). Disease manifestations range from asymptomatic to a mild undifferentiated dengue fever (DF) and in up to 5% of symptomatic cases progresses to serious life-

threatening complications as a result of vascular leakage, hemorrhage and organ dysfunction (Simmons et al., 2012, Low et al 2017). Secondary dengue infections are often, but not consistently associated with severe dengue disease in part because of antibody dependent enhancement (ADE) phenomenon (Halstead, 2007).

Vaccination, as a measure to counter the spread of dengue, has been on the agenda of national health authorities for many decades and still remains a high priority for the World Health Organization (WHO) (Russell, 1978; Recker et al., 2016). At the WHO Conference on dengue held in Singapore in 1977 it was predicted that it would take 10 years to develop a tetravalent dengue vaccine (Russell, 1978). However, the unexpected challenges of developing such a vaccine took nearly four decades.

The first dengue vaccine, Dengvaxia®, developed by Sanofi Pasteur has been licensed in several countries since December 2015. The attenuated live recombinant vaccine was generated by splicing in the prM and E genes from DENV 1-4 into the yellow fever virus (YFV) vaccine strain genome, replacing the YFV prM and E genes to yield the chimeric yellow fever 17D - tetravalent dengue vaccine (CYD-TDV). Dengvaxia is administered as 3 doses on a 0/6/12 month schedule and clinical studies found the overall efficacy to be 59.2%, with serotype-specific efficacy for DENV 1-4 to be 54.7%, 43%, 71.6% and 76.9%, respectively. The vaccine is recommended for use in the individuals 9-45 years of age living in dengue endemic regions where the disease burden is high (WHO 2016).

This achievement by Sanofi Pasteur should be commended as it marks an important milestone for dengue control through exemplary partnership with a very large number of public and private institutions around the world. Valuable lessons have been learned and other dengue vaccine candidates are currently being explored to improve efficacy, extend the age coverage and also simplify the vaccination schedule in order to achieve wider vaccine acceptance and uptake. Apart from vaccination, the vector control strategy using *Wolbachia* bacteria to reduce the capacity of *Aedes aegypti* mosquito to transmit dengue is being actively explored in many countries largely through the support of the Bill & Melinda Gates Foundation <http://www.eliminatedengue.com>

Dengue antiviral research and development as a potential intervention approach received a boost in 2003 when Novartis Pharma formed a public-private partnership with Singapore's Economic Development Board to establish the Novartis Institute for Tropical Diseases (NITD). The expectation was that a safe orally available drug for use in those >5 years old and with

activity against all four DENV serotypes and capable of reducing dengue symptoms and the incidence of severe dengue could be achieved in 10-12 year period (Keller et al., 2006).

Although such a drug has not eventuated so far, the efforts of NITD have led to many promising leads targeting the NS3 and NS5 proteins, which contain the enzymatic activities necessary for viral RNA replication. These leads have been reviewed by Lim and colleagues (Lim et al 2013). In the meantime, major strides in the understanding of the structure and function of viral targets as well as dengue pathogenesis in general have been made through state-of-the-art research using 3D structural studies, cryoelectron microscopy, reverse genetics, RNAi screens, CRISPR screens, whole genome deep sequencing and animal model studies.

The contributions of data arising from these new approaches hold promise for the development of directly-acting antivirals (DAA) or host factors that can target viral processes such as

- i) viral entry/fusion
- ii) translation/polyprotein processing,
- iii) RNA replication and
- iv) packaging/virus maturation.

However, in order to make an impact as one of the viable intervention approaches for dengue control, alongside with vaccines and vector control measures, it is important that an efficacious dengue antiviral drug reaches the clinic by the first half of the next decade. This is a significant challenge considering the bottlenecks in drug development during the late pre-clinical to clinical development phase.

One way to mitigate this is through the formation of research consortia such as the partnership involving Wellcome Trust/KU Leuven/Janssen for dengue drug development

<http://www.janssen.com/partnerships/dengue>

to develop a pipeline of candidate drugs, which can be rapidly evaluated and developed as potent dengue antivirals. Similar partnerships through the National Institutes of Health (USA), European Union and other national agencies in other parts of the world can also help to link the large body of work from academic laboratories and small biotechnology companies to ensure potential candidate drugs can successfully enter the clinics for testing. If the task of managing the intellectual property expectations of partners within consortia can be streamlined, then work from these types of partnerships should result in more fruitful outcomes for diseases such as dengue or other emerging viral diseases.

New opportunities for development of pan-flaviviral drugs have arisen especially since the closely related

Zika virus (ZIKV) outbreak (Fauci and Morens 2016). Infection with ZIKV, which can result in congenital deformities in newborn babies when the mothers are infected during pregnancy as well as Guillain-Barré syndrome in otherwise healthy adults, has brought intense international focus on this emerging threat with calls for urgent development of antiviral drugs.

Flaviviruses carry a single-stranded positive sense RNA genome of ~11000 nucleotides that encodes a ~3400 amino acid residues polyprotein precursor which is cleaved into three structural proteins (capsid, pre-membrane/membrane and envelope) and seven non-structural (NS) proteins (NS1, NS2A, NS2B, NS3, NS4A, NS4B and NS5). The crystal structures of the enzyme targets, NS3 and NS5 from ZIKV, reveal extensive structural similarities with DENV and other flaviviruses, which can form excellent starting point for the development of novel DAAs.

In the case of ZIKV NS3 protease, it was shown that a binary expression construct where the NS2B is not constrained by artificial linkers or modified native linker has resulted in high resolution crystal structure where the NS2B wraps around the NS3 molecule in the closed conformation leaving the substrate binding site empty for binding small molecules. These crystals are amenable to fragment based screening and structure-directed antiviral development (Zhang et al., 2016). In the case of NS5, the structures also show high similarity and the nucleoside inhibitor NITD 008 is widely used as a control for drug screening efforts with different compound libraries (Barrows et al., 2016; Xu et al., 2016). Opportunities for non-nucleoside inhibitors also exist since the structural similarities reveal conserved pockets near regions that are critical for de-novo initiation for viral RNA replication that can be targeted.

Important lessons have been learned about conducting dengue antiviral clinical trials using repurposed drugs, which has been reviewed recently (Low et al., 2017). This strategy is being more widely and systematically applied to discover novel individual or combination compounds as potential pan-flaviviral drugs. Given the acute nature of dengue and the narrow window of opportunity for antiviral treatment, it is likely that the number of patients who can be recruited to evaluate the efficacy of these drugs may limit early phase clinical trials. Additionally, the high cost of large trials with long study duration may limit the number of compounds entering Phase II/III trials with detrimental effect on the whole process of drug discovery and development.

In recent years, the limitations of large randomized clinical trials have been widely acknowledged as severely impairing medical advances. Adaptive trial

designs, that aim to increase clinical trial efficiency, are increasingly being employed especially in oncological clinical studies (Bhatt and Mehta 2016). Adoption of similar strategies may benefit future dengue clinical trials.

In part to further address clinical endpoints that require large sample size as used in dengue trials, an alternative robust biomarker, fluorodeoxyglucose (FDG), was recently examined in preclinical animal models. The goal was that drug efficacy could be directly correlated with reduction in FDG signals that were reflective of virus-induced inflammation by using positron emission tomography (PET) imaging (Chacko et al 2017). If the observations from the preclinical study translates to human cases, then it is conceivable that smaller Phase II trials can be rapidly conducted to triage a pipeline of drugs from various consortia. It would be valuable for other robust biomarker to be evaluated with a similar goal.

In summary, the successful development of antivirals against the hepatitis C virus serve as an example that it is possible to develop potent dengue/pan-flaviviral antivirals. The bar may be viewed as being a little higher in the case of dengue because the available funding is probably a fraction of that available for HCV antiviral development. However, the task is not insurmountable if partnerships and consortia that led to the successful development of Dengvaxia can be recapitulated for the dengue antiviral development to deliver a drug to be deployed for clinical use by 2025.

Acknowledgements

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Next-generation nucleos(t)ide inhibitors of viral polymerases

Lieve Naesens

Rega Institute for Medical Research, KU Leuven – University of Leuven, B-3000 Leuven, Belgium
lieve.naesens@kuleuven.be

This year, antiviral researchers celebrate the 40th anniversary of G. Elion's first publication on acyclovir [1], as well as the 30th anniversary of zidovudine as the first drug to become approved for HIV therapy. Both events represent landmarks in the clinical management of virus infections. The class of nucleos(t)ide inhibitors of HIV RT ripened to become the cornerstone in HIV treatment; some of these drugs proved to be equally valuable for hepatitis B. Since the approval of sofosbuvir, hepatitis C is another viral disease for which nucleos(t)ide inhibitors appear hard to compete with. Sofosbuvir is the first nucleotide drug acting as a chain terminator on a viral RNA-dependent RNA polymerase.

Newer analogues, such as the RSV inhibitor AL-8176, and GS-5734 that is active against Ebola plus some other RNA viruses, are in the clinical pipeline to, hopefully, address some important and unmet medical needs.

When we look at which molecules have gained successful clinical approval, the structural variations on the nucleos(t)ide theme are still quite limited. This contrasts with the large chemical variety among experimental nucleos(t)ides that have been synthesized and proven to have antiviral activity in preclinical or early clinical studies. A few examples of rather 'exotic' sugar replacements are dioxolane [2]; six-membered rings [3]; or conformational locked rings [4]. Likewise, quite drastic variations in the base part are accepted, as exemplified by the two carboxamide compounds ribavirin and favipiravir, or the herpesvirus inhibitors brivudin and valnividine (FV-100).

It is evident that viral polymerases are highly promiscuous and able of recognizing nucleoside-triphosphate analogues with a very unusual and unnatural structure. For HIV RT and viral RNA-dependent RNA polymerases that lack proofreading activity, this is not surprising. However, even herpesvirus polymerases, which do possess exonuclease activity, seem to be easily fooled. In fact, the defined antiviral spectrum that is often assigned to antiviral nucleos(t)ides is too simplistic. For instance, tenofovir inhibits HSV; cidofovir-diphosphate inhibits HIV RT; some dideoxynucleoside inhibitors of HIV are also active against adenovirus; penciclovir inhibits hepatitis B virus; and GS-5734 is a recent example of a broad anti-RNA virus agent.

Why is then the structural diversity of antiviral nucleos(t)ide drugs still quite limited? An important restriction lies within the host cell, which possesses a variety of purine and pyrimidine-converting kinases acting in tightly controlled pathways that are connected to nuclear or mitochondrial DNA or RNA synthesis. Except for nucleoside diphosphatase (NDP) kinase, these kinases generally display narrow substrate specificity. For antivirals, this kinase dependency represents a daunting barrier since the unnatural nucleos(t)ide should not merely bind to the kinase, but, more importantly, act as an efficient substrate to be smoothly phosphorylated.

To bypass the first and often rate-limiting first phosphorylation step, monophosphate formats like tenofovir and sofosbuvir have been designed. Only recently, successful di- and triphosphate prodrug concepts have been developed by Meier and colleagues [5, 6], creating the possibility to design antiviral nucleotides with less obvious chemical structures in the

base or ribose part. Given the lack of proofreading activity in the vast majority of RNA viruses (coronaviruses being the only exception), such di- and triphosphate prodrugs could be one way to achieve broad anti-RNA virus agents, which are desperately needed to tackle some emerging and potentially pandemic viruses.

Another issue worthy of thought is whether obligate/non-obligate chain termination, relying on incorporation of the antiviral nucleotide, is really an absolute requirement. Whereas obligate chain terminators lack a 3' -hydroxyl function, the term non-obligate chain termination is used for nucleotides with modifications in the sugar part (e.g. at the 1' , 2' or 4' position) which hinder binding of the next nucleotide or cause termination during next-round synthesis of the complementary strand [7]. Chain termination was also reported for BCX4430, an immucillin-adenosine analogue that is active against Ebola virus.

Although most antiviral nucleos(t)ides indeed possess unnatural (deoxy)ribose parts leading to non-obligate chain termination, exceptions with a natural ribose do exist, such as brivudin and favipiravir. For the latter molecule, the mechanism behind its inhibitory effect on viral RNA synthesis remains uncertain. The central dogma is that chain termination ensures that viral DNA/RNA synthesis is fully shut off, leading to a powerful antiviral effect. If recognized by a cellular polymerase, such a nucleotide might disrupt cellular DNA/RNA synthesis but the chain termination event would exclude mutagenic effects upon the host cell.

A valid alternative could be to design nucleotide mimetics that are not covalently incorporated into viral DNA or RNA but, instead, act as strong non-covalent blockers of the polymerase active site. One way is to introduce a triphosphate-like moiety (such as an α -carboxyphosphonate) that mimics the interactions of the α , β and γ phosphates [8]. A related approach is to design mimetics of some transition state of the polymerase enzyme. This drug concept has yielded superior enzyme inhibitors in the field of HIV protease, influenza neuraminidase or some N-ribosyltransferase enzymes [9]. Given that viruses rely on very fast replication of their genomes to generate millions to billions of particles per day, a nucleotide showing tight binding to the viral polymerase could produce strong antiviral effects. Even if such an active site blocking nucleotide would be recognized by a cellular polymerase, it is fair to assume that this would be free of mutagenicity.

The new generation of antiviral nucleos(t)ides should, of course, be selected to have a high barrier to

drug-resistance. In HIV RT, one common resistance mechanism is related to higher fidelity mutants that better discriminate between the natural and unnatural dNTPs, which is linked to the mutant having a more rigid active site structure [10]. This suggests that unnatural analogues with higher structural flexibility can still adopt to the mutated site and, hence, remain active against the mutant virus. The downside is that ligand flexibility may reduce antiviral potency when it affects the binding parameters towards the protein target.

So perhaps it is time to consider some less explored scaffolds in the search for new nucleos(t)ide antivirals? Regardless the mechanism, one important issue is related to discrimination between viral and cellular polymerases. As mentioned above, the lack of proofreading activity in many viruses (i.e. in virtually all RNA viruses), combined with their reliance on fast genome copying kinetics, are intrinsic factors that contribute to selectivity. In addition, for acute virus infections requiring only short-term treatment, a weak inhibitory effect on some cellular polymerase is expected to be harmless.

Another topic in the design of antiviral nucleos(t)ides is that of prodrug moieties to modulate drug lipophilicity, tissue distribution or organ tropism. Brincidofovir is a lipid conjugate showing increased oral bioavailability and CNS penetration than its cidofovir parent. A main advantage of the phosphoramidate drug sofosbuvir in the therapy of hepatitis C is its activation by liver-expressed enzymes.

To expand the prodrug toolbox, some creativity is needed. Perhaps we can exploit enzyme pathways that are upregulated in, for instance, phagocytes or granulocytes (which are attracted towards virus-infected tissues), lung alveolar cells or the blood-brain barrier. Or perhaps the (immuno)pathology of virus infections may boost some specific enzyme reactions in the infected organs? The availability of genome-wide proteomic and metabolomic profiles can provide clues on which enzymes could be considered. One drawback is that these enzymes may be confined to primary cells or more complex tissues, which are harder to isolate, manipulate or infect with viruses for *in vitro* studies.

To conclude, antiviral nucleos(t)ides are alive and kicking. These versatile and ubiquitous DNA/RNA building blocks are a relentless source of imagination for medicinal chemists, biochemists and virologists. New structural analogues, protides and prodrugs need to be further designed to consolidate the leading position of nucleos(t)ide analogues in the field of antiviral therapy.

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ISAR MEMBER PROFILE

Don Smee, Research Professor, Institute for Antiviral Research, Utah State University, Logan, Utah, USA.

I have been asked to express some thoughts about my career, the colleagues I have worked with, my membership in ISAR, and how attendance at ICAR has benefited my career. My first job out of college was the most important one to prepare me for my career as a research scientist in virology.

I received my Bachelor of Science degree during the Vietnam era along with a commission as a Second Lieutenant in the United States Army. At that time, the Vietnam War was nearly winding down. I did not serve in Vietnam but was assigned to the 7th Infantry Division at Ft. Ord, California. I initially was a medical platoon leader, and later worked in the Division Surgeon's Office where I did an extensive amount of technical writing. This prepared me for graduate school at Utah State University (USU), and college expenses were largely funded under the GI Bill. Because of my military training, writing my thesis and dissertation was not a burden, and the time spent preparing these documents was minimal. During my scientific career, I have seen many graduate students struggle mightily with writing because they lacked experience.

Because I had enjoyed my time in the military, my immediate goal after graduate school was to get a position at the U.S. Army Medical Research Institute of Infectious Diseases (USAMRIID). However, this did not happen exactly as planned. I spent my first eight

years after graduate school in two pharmaceutical companies, Syntex Research and ICN Pharmaceuticals. Syntex was later acquired by Roche and ICN terminated its research program and its name was changed to MP Biomedicals. When ICN was winding down, I left there to work at the Institute for Antiviral Research at Utah State University (USU) under Robert Sidwell who was the Institute's director. This was a soft money position, and the money ebbed and flowed from year to year. Hard times came to me in 1996 when my grants ran out and other funding sources were drying up. It was then that I took a job as a contractor at USAMRIID, where I worked for nearly three years. Then I returned to USU and have been there ever since, working currently as a Research Professor, an appointment that is still a soft money position.

At the onset of my career at USU, I was uncomfortable being on soft money, since the money well can go dry as it did for me in 1996. After USAMRIID, I had a much greater determination to make the soft money position work out. At USAMRIID, I conducted antiviral studies on orthopox viruses. I was able to obtain funding for poxvirus work at USU, which gave me a jump-start back in academia. Moreover, I still interact with one of my colleagues from the USAMRIID era, Mike Bray, who is the current Editor-in-Chief for *Antiviral Research*. One interesting thing occurred a few years after I left USAMRIID. I was interviewed by the U.S. Postal Service that was working with the FBI to investigate people who may have been involved in the anthrax terror attacks of 2001. I was investigated because, while at USAMRIID, I was a colleague with Steve Hatfill, who was their main terrorist suspect at the time. Later, Steve was exonerated in the case.

My career has taken many turns over the years, ebbing and flowing with the medical needs of the time. The most significant work I accomplished in my career was in the early 1980's when I participated in the pre-clinical development of ganciclovir. There were five companies vying for the rights to the compound, and Syntex emerged victorious. It was an exciting time because ganciclovir was a very hot topic and there were many people interested in it. Another significant part of my career has been the development of new animal models of viral infections. This has been diverse, and included developing different types of herpesvirus infections in immunocompromised mice, new orthopoxvirus models, and new models with arenaviruses, bunyaviruses, and orthomyxoviruses. In the latter categories, it has been exciting to work with a broad array of different RNA viruses.

My passion over the years has been performing animal studies. Animal experimentation is where the rubber meets the road in terms of an antiviral compound's potential to become a drug. I have read too many articles in the scientific literature where in vitro activity was touted, but with no evidence of efficacy in animals reported. Later, most of the compounds fell by the wayside due to lack of in vivo efficacy. The approach we have taken at the Institute for Antiviral Research is to publish the results of cell culture and animal testing data in the same article. Animal work will forever play a key role in drug development. Continuing research into the discovery of new animal models is essential.

Along these lines, one recent project from our group is the development of an enterovirus D68 (EV-D68) mouse model that we reported at the 2017 ICAR meeting. My colleague Bart Tarbet is the lead researcher on that project. Since EV-D68 is related to rhinoviruses, the mouse model may serve as a surrogate for anti-rhinovirus compound evaluation if the compounds also inhibit EV-D68. Using this model, we demonstrated that guanidine is active at inhibiting virus replication but rupintrivir is not. Rupintrivir is highly potent in vitro against picornaviruses, but failed in treating rhinovirus infections in the clinic. Had this new animal model of EV-D68 infection existed in the early stages of rupintrivir development, the company would have been able to find out the ineffectiveness of the compound long before they invested in expensive clinical trials.

Mode-of-action studies are also highly interesting to me. I was able to do a lot of work in this area under the ganciclovir project, but not so much currently. What is satisfying is to take a compound for which nothing is known and actually figure out how the compound works. At the ICAR meeting each year, we often hear very interesting mode of action presentations.

In my career, I have rubbed shoulders with some of the great men in science. My mentor in graduate school was Robert Sidwell, whom eight years later I worked with at Utah State University and then until he retired in 2006. We attended many ICAR meetings together year after year. His passion on those trips was to go to nice restaurants in the evenings and insisted that I go along with him. I would take him out into the woods and swamps looking for reptiles, which is what we did at the New Orleans ICAR in 1991. I worked closely with John Martin (currently CEO of Gilead Sciences) when he was a chemist at Syntex. He was a key player in the team that developed ganciclovir. Later, it was my pleasure to work with Roland Robins at ICN Pharmaceuticals. Roland and Robert Sidwell spear-

headed the ribavirin development project, but this was before my time at ICN.



Don's colleagues at the Institute for Antiviral Research, Utah State University. Back row, from left to right: Brian Gowen, Justin Julander, Bart Tarbet, and Craig Day; Front row, Don, John Morrey and Dale Barnard.

The first ICAR meeting that I attended was held in Williamsburg, Virginia in 1988. The scientific meeting was excellent, and Colonial Williamsburg was fabulous to visit. The Jerusalem meeting in 1999 was also high on my list of favorites. I went to that meeting with my colleagues John Huggins and Mike Bray from USAMRIID. The ISAR has been a valuable part of my research career. Many of the individuals that I collaborate with or do projects for are members of ISAR. The ICAR meeting brings diverse disciplines together and it is a chance to interact face-to-face with colleagues that I otherwise only visit through email or with occasionally on the telephone. I am involved with many viruses and research interests, and the ICAR is the most important meeting for me to get it all in just one venue. I have also enjoyed serving on various ISAR committees.

UPCOMING MEETINGS OF INTEREST

Emerging and Re-emerging Viruses Arlington, VA, USA (October 1-3, 2017)

This meeting, organized by Elsevier Conferences, will aim to ask how basic host-cell biology can contribute to understanding emerging and re-emerging viruses to help with surveillance as well as vaccine and drug development. The meeting will bring infectious-disease specialists together with researchers interested in the host response to these viruses from immunological, cell-biological, and pharmacological viewpoints. The conference will focus on a range of

human pathogenic viruses, which are constantly emerging and re-emerging, as highlighted by the recent Ebola outbreak in West Africa and the ongoing Zika virus outbreak in the Americas.

The meeting will be held in Arlington, VA, USA (October 1-3, 2017). The Hotel is conveniently located just minutes from Washington DC, a city of variety and contrast, with a central area beautifully designed with broad avenues lined with magnificent buildings and monuments set in spacious green parks.

For more information, go to the website <http://cell-symposia.com/emerging-viruses-2017>.

3rd Global Summit on Virology, Barcelona, Spain (August 21-23, 2017)

The Virology-2017 conference will be centered on innovations and therapeutic approaches in virology. This meeting aims to be a perfect platform for the researchers, scientists, professors, and students to exchange and discuss on their ideas in the field of Virology. Virology-2017 anticipates around 300 participants around the globe and the three-day conference will include Plenary sessions, Keynote speeches, Poster and Oral presentations. The following scientific sessions are planned:

- Virotherapy
- Insect vector and virus epidemiology
- General Virology and Basic Sciences
- Viral Breakout: Prevention and Measures
- Human Viral Diseases Affecting Afro-Asian Continents
- HIV and Other Retroviral Diseases Affecting Afro-Asian Continents
- Pediatric Viral Infectious Diseases
- Organ Specific Cancers and Human Tumour Virology
- Respiratory, Vector Borne Diseases and Emerging/Re-emerging Viruses
- Therapeutic Approaches and Targets for Viral Infections
- Viral Hepatitis- Virus-host interactions
- Viral Immunology
- Veterinary Virology
- Clinical and Neuro Virology
- Agriculture and Plant Virology
- Antiviral Vaccine Development
- Bacteriophages
- Antiviral Drug Discovery and Development
- Current Focus in Virology Research
- Retrovirus-HIV/AIDS-Basic Sciences and Implications

- Regulatory and Economical Aspects in Virology
- Viral Safety and Risk Assessment
- Clinical Aspects of Viral Diseases
- Viral epidemiology and diagnostics
- Prevention of and Therapy for Viral Infections
- Food virology
- Recent virus infections in the Middle East area
- Host-directed therapy - New ways to curing infections

Barcelona will host the 3rd Global Summit on Virology at the Hotel Alimara Barcelona (August 21-23, 2017). Barcelona is the cosmopolitan capital of Spain's Catalonia region, defined by quirky art and architecture, imaginative food and vibrant street life. The city has medieval roots, perceived in the mazelike Gothic Quarter, as well as a modernist personality represented by Antoni Gaudi's Sagrada Familia church.

More information regarding this conference can be found on the website <http://www.virologycongress.com>.

20th Annual Meeting of the European Society for Clinical Virology (ESCV), Lago Maggiore, Italy (September 13-16, 2017)

The 20th Annual Meeting of the ESCV will be held in Stresa located at the Lago Maggiore, Italy, September 13-16, 2017. The scientific program, comprising presentations, symposia, discussions, open sessions and workshops, will be centered on the state of the art as well as the most recent discoveries and innovations in Clinical Virology. The main topics of the meeting will include respiratory viruses, gastrointestinal viruses, virus-associated neurologic syndromes, virus-associated tumors, hepatitis viruses, HIV and other retroviruses, viruses in immunocompromised patients, viral infections in pregnancy, emerging viruses, immune response to viral infections, advancements in diagnosis and monitoring advancements in prevention and treatment.

The famous town of Stresa (5000 inhabitants, 200 m above sea level) enjoys a splendid location on Lake Maggiore in the Gulf of Borromeo. Stresa is situated in Northern Italy, in an ideal position between mountains and the shores of Borromean Gulf. Its beautiful countryside, mild climate, luxury villas and magnificent Art Nouveau hotels made the town of Stresa a perfect combination of relaxation and culture. From Stresa it is easy to reach the three Borromean Islands that are steeped in artistic, historical and botanical appeal.

Details of the meeting can be found on the ESCV2017 meeting's website www.ESCV2017.com.

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