

A Tribute to Barrie J. Carter

Arun Srivastava, Matthew Weitzman, Saswati Chatterjee, John F. Engelhardt, Roland A. Owens, Nick Muzyczka, and Robin Ali

ODE TO BARRIE

I FIRST BECAME AWARE of Barrie Carter's pioneering work on adeno-associated virus (AAV) nearly four decades ago, when I started my postdoctoral training in the laboratory of Ken Berns, yet another stalwart among what used to be a small group of individuals who were interested in AAV. During my 2 years in Ken's laboratory, it became abundantly clear to me that essentially all of the basic science studies—from characterization of the AAV DNA, AAV DNA replication, and AAV gene expression—were carried out in Ken's and Barrie's laboratories, respectively, nearly contemporaneously in the 1970s and 1980s. What was even more impressive was the collegiality and camaraderie that I witnessed between Barrie and Ken. They even co-authored a publication in 1976.¹

Jude Samulski, then a graduate student in Nick Muzyczka's laboratory, cloned the wild-type AAV genome, and reported this achievement, with Ken as a co-author, in March 1982.² Barrie's laboratory also reported the same accomplishment in July 1983.³ These were important milestones, as molecular cloning of the AAV genome facilitated the development of recombinant AAV (rAAV) vectors.

Paul Hermonat, also a graduate student in Nick's laboratory, first reported the development of a rAAV vector expressing the neomycin-resistance gene in October 1984.⁴ Remarkably, also in October 1984, Barrie's laboratory described the creation of a rAAV vector expressing the chloramphenicol acetyltransferase gene.⁵ Jude, as a postdoctoral fellow in Tom Shenk's laboratory, created a versatile AAV cloning vector, pSub201,^{6,7} which, to date, has been used to generate most, if not all, AAV vectors.

Thus, it can be said that Nick and Barrie are the “fathers of rAAV vectors”! Barrie also deserves accolades for having recruited Terry Flotte to his laboratory, and Terry has the distinct honor of having performed the very first Phase I clinical trial with a rAAV vector. Terry dosed the first patient in a cystic fibrosis trial on November 21, 1995, and published the results in 1996.⁸ Thirty-five years and 201 Phase I/II/III clinical trials later,⁹ AAV has gone from “almost a virus” to “an awesome vector,” and is now the

basis of a multi-billion dollar industry, thanks to Ken, Nick, Barrie, and Terry, and, of course, Jude.

Personally, I am extremely grateful to Barrie for a number of reasons, which include the following. First, it was Barrie who made me aware of the International Parvovirus Workshops, which I have attended, and participated in, since 1989. Second, in 1992, I was invited by Eileen Lennon, Scientific Review Officer at the National Heart, Lung, and Blood Institute, National Institutes of Health, to be a member of the Site-Visit Team to review a Program Project Grant at the University of Iowa in Iowa City. When I thanked Eileen for giving me the opportunity, she told me that I should really thank her husband instead, who she said had recommended my name. Eileen's husband is Barrie! Third, soon after assuming the role of Editor-in Chief of *Human Gene Therapy*, Terry dedicated one issue of the journal to “Controversies in AAV,” and asked Barrie and me to write an article on AAV and cancer. It was a delightful experience to write this article with Barrie, which was published in 2017.¹⁰ Later that year, Terry sent the following email to Barrie and me: “I wanted to follow up and let you know that your paper for the AAV controversies issue was one of the journal's top downloaded papers of the year so far, with 1,573 downloads!! Thank you for your help in making this issue so successful.” And fourth, Mavis Agbandje-McKenna and I had the privilege of organizing the XVIIth International Parvovirus Workshop, which was held in Miami Beach in 2018 (<http://parvovirusworkshop2018.org>). Barrie was instrumental not only in assisting us in obtaining generous funding from BioMarin to support the workshop, but also in providing the group photograph of the very first workshop, which was held in Grangeneuve, Switzerland, in 1985. We included this group photograph, among several others, in the abstract book for posterity. Terry also allowed us to publish the workshop report last year.¹¹ Ken, Nick, and Barrie are among the handful of “hard-core” parvovirologists who have attended this workshop from the very beginning.

On December 16, 2018, Barrie sent the following email message to several of us, with “I am retiring” in the subject

line: “As of Friday 21st, I have decided that more than 50 years of science is enough. So other than some continuing consulting for BioMarin I am planning on spending my life mostly doing other things. May still go to ASGCT, and some other meetings. Cheers and holiday greetings.”

I saw Barrie and Eileen again at last year’s annual meeting of the American Society of Gene and Cell Therapy (ASGCT) in Washington, DC, and they told me that they were really enjoying their retirement.

Thank you, Barrie, for all you have done for AAV in the past 50 years, and I look forward to seeing you again at many more ASGCT meetings and International Parvovirus Workshops in the next 50 years!

Arun Srivastava, PhD

George H. Kitzman Professor of Genetics

Chief, Division of Cellular and Molecular Therapy

*Departments of Pediatrics, Molecular Genetics, and Microbiology
Powell Gene Therapy Center, Cancer and Genetics Research Complex*

2033 Mowry Road, Room 492-A

University of Florida College of Medicine

Gainesville, FL, USA

Matthew Weitzman, PhD

Professor of Microbiology, Professor of Pathology and Laboratory Medicine

University of Pennsylvania Perelman School of Medicine

4050 Colket Translational Research Building

The Children’s Hospital of Philadelphia Research Institute

3501 Civic Center Blvd

Philadelphia, PA, USA

ODE TO BARRIE CARTER

“The government may be closed tomorrow.” This was the email I received the day before my visit as I searched for a postdoc position in the fall of 1990. It was my introduction to the National Institutes of Health (NIH) bureaucracy and Barrie Carter. Despite the closure, I visited the NIH campus and spent the day with an energetic Barrie, talking to me non-stop about science and introducing me to neighboring labs. I joined Barrie’s small lab in the summer of 1991, where Roland Owens was studying Rep protein interactions with the inverted terminal repeat (ITR), and Terry Flotte was putting a CFTR transgene into the AAV vector. These topics reflected the basic science into AAV biology, combined with the translational potential of gene transfer, which together characterized his group. Barrie ran an open lab and encouraged us to follow our instincts. I started by making truncated versions of the Rep proteins *in vitro* so that we could map what was required for ITR binding and protein multimerization. His lab was also a model for collaboration, and we each took parts of collaborative projects to address different questions about regulation of transcription and replication. The lab was split between a number of small rooms, and my bench was initially located in the lab next to Barrie’s office. This meant that he passed by regularly, and I got to hear directly from the master about the evolution of the AAV field. Barrie loved to tell stories about the history of virus research and the characters that filled it. He introduced me to the wider parvovirus community and was always fun to have at the international parvovirus workshop. Most characteristically, Barrie took a family style to the lab and the trainees who came through it. He connected us to former

lab people, and we met his own family. Within a year, Barrie had decided to take his knowledge of AAV biology and devote it toward making gene therapy a reality. I was the last trainee of his NIH lab as he left for an industry position. Barrie’s approach to mentoring endured even after he had left the NIH. He correctly viewed mentoring as a life-long commitment, and continued to provide career advice and letters of support to me throughout my career. I am indebted to Barrie for introducing me to this small DNA virus and how to use it in rigorous research. The field is indebted to him for his many contributions that formed the foundation for development of AAV vectors. His intellectual curiosity, appreciation of basic science contributions to translational applications, his scientific generosity, and his mentoring spirit are models for us all.

A TRIBUTE TO BARRIE AND HIS INSPIRATIONAL CAREER

Barrie’s remarkable career encompassing the many facets of AAV from early discovery to the evolution of gene therapy has long served as a source of personal inspiration. I first met Barrie Carter at the NIH. He was a Lab Chief of the Laboratory of Molecular and Cellular Biology, in the National Institute of Diabetes and Digestive and Kidney Diseases. I was a postdoctoral fellow in the laboratory of his former colleague, James Rose at the Laboratory of Viral Diseases, National Institutes of Allergy and Infectious Diseases. I believe these were the only two laboratories working on AAV at the NIH at the time. At the time, the AAV field was small. Barrie Carter along with Jim Rose and a small group of scientists laid the pioneering groundwork on the fundamentals of this innocuous, nonpathogenic virus. Ed Sebring, Frank Kocot, and Phyllis Fabisch recounted legendary stories of the early days of AAV. I felt that I knew Barrie even before I actually met him. I was in awe of the breadth of Barrie’s impressive record of having worked on every aspect of AAV from the very start to the first clinical trial of AAV. Barrie was among the first to realize the tremendous potentials AAV held for genetic therapies. Throughout his career, he continued to guide the evolution of the field. In addition to his prolific research contributions, Barrie also played an important instrumental role in guiding the complex regulatory aspects of the developing field of gene therapy. After his storied career at the NIH, he pioneered the effort to transition AAV into the commercial arena and clinical gene therapies through his work at Targeted Genetics and at BioMarin.

On a personal note, after the NIH days, our paths continued to cross at conferences. At these meetings, I was sometimes accompanied by my son, from when he was an infant to his teenage years. I was always impressed at how kind Barrie always was to him. He never failed to ask about him.

Although it is difficult to envision the AAV field without Barrie playing an active role, we will always be indebted to him for his pioneering efforts in laying the foundations of the field and his visionary translation into promising therapies for incurable diseases. Future scientists and patients alike will benefit from Barrie's contributions as lives are transformed. And, with some luck, perhaps we will continue to catch an occasional glimpse of him at some parvovirus conference.

Saswati Chatterjee, PhD
Professor, Department of Surgery
Beckman Research Institute of City of Hope
1500 East Duarte Road
Duarte, CA, USA

Dr. Carter's founding discoveries on the applications of rAAV as a gene therapy vector have forever changed the field of molecular medicine. The depth of his scientific contributions to our understanding of basic AAV virology and his passion for translating these findings into therapies for patient have left an indelible mark on the field of gene therapy.

John F. Engelhardt
Professor and Chair, Department of Anatomy and Cell Biology
Director, Center for Gene Therapy
Roy J. Carver Chair in Molecular Medicine
University of Iowa
Room 1-111 Bowen Science Building
51 Newton Road
Iowa City, IA, USA

I would assert that anyone who is serious about innovation, diversity, and inclusion should promote an environment of co-mentoring, respectful questioning, and open communication. The Carter lab at the NIH provides a fine example.

Roland A. Owens
Director of Research Workforce Development, Office of Intramural Research
Office of the Director
National Institutes of Health
U.S. Department of Health and Human Services
Bldg. 1, Room 158
1 Center DR MSC-0140
Bethesda, MD, USA

My favorite Barrie quote is "If you haven't published it, then you didn't do it."

Nick Muzyczka
Professor, Molecular Genetics and Microbiology
University of Florida College of Medicine
1600 Archer Rd, Rm. R1-118B
Gainesville, FL, USA

One of Barrie Carter's many contributions to the field of gene therapy was his involvement in our ocular gene therapy program that started with gene therapy for Leber congenital amaurosis 2. The first AAV vector to be injected into a human eye was manufactured by Barrie's company, Targeted Genetics Corporation—a gene therapy company that was ahead of its time. It was a pleasure to work with such a wise, warm, and generous man.

Robin Ali
Professor, Molecular Therapy
Institute of Ophthalmology
Bath Street
London
EC1V 9EL
United Kingdom

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