PREFACE

True, in a single conversation with someone we can discern particular traits. But it is only through repeated encounters in varied circumstances that we can recognize these traits as characteristic and essential. For a writer, for a musician, or for a painter, this variation of circumstances that enables us to discern, by a sort of experimentation, the permanent features of character is found in the variety of the works themselves.

***From Marcel Proust's preface to John Ruskin's* The Bible of Amiens.**

The Ebola River, a tributary to the Congo, flows north of the village of Yambuku. There, in 1976, hundreds of people rapidly succumbed to a lethal hemorrhagic fever. The cause, Ebola virus, is a member of the genus Filoviridae, comprising single-stranded negative-RNA viruses with the idiosyncratic filamentous or worm-like morphology that has given them their name.[1] As a tragic Ebola epidemic now rages in West Africa, killing thousands, efforts to find a cure and a vaccine will intensify. It is already striking how the advancing field of filovirus studies shares questions and problems with the investigations—some old and established, some rapidly evolving—of other viruses, as exemplified in this book. Thus, knowledge is developing of how filoviruses enter cells,[2] the identity of the receptors for the virus on susceptible cells,[3,4] which cellular genes these viruses activate, how that activation affects the innate immune responses and pathogenesis,[5,6] how the virus is neutralized by antibodies, and which antibodies protect against infection.[7–10]

Thomas Milton Rivers, working at The Rockefeller Institute, which I see through the window when composing this Preface, established virology as a discipline separate from bacteriology.[11] He perspicaciously stated: "Viruses appear to be obligate parasites in the sense that their reproduction is dependent on living cells." His anthology *Filterable Viruses* (Baltimore: Williams and Wilkins, 1928) covered everything worth knowing about viruses at the time. Today, when the number of PubMed entries in virology is around a million, an anthology in general virology must be considerably less comprehensive. The current collection encompasses a number of topical forays into molecular aspects of viral replication and coexistence with host organisms. The chapters in this anthology offer rich opportunities to compare how specific questions are answered for different viruses. As with the example of Ebola virus above, certain themes recur and the emerging

patterns of similarities and differences may provoke new questions and stimulate collaborations among virologists with distinct specialties.

Their eclectic diversity notwithstanding, the chapters form a narrative of sorts, first adhering kairologically to the replicative cycle that viruses largely share, and then broadening to depict wider aspects of virus–host interactions. Thus, the first three chapters depict entry into susceptible cells by different viruses: paramyxoviruses (Chapter "Unity in Diversity: Shared Mechanism of Entry Among Paramyxoviruses," Palgen *et al.*), alphavirus (Chapter "Alphavirus Entry into Host Cells," Vancini *et al.*), and hepatitis C virus (Chapter "The Mechanism of HCV Entry into Host Cells," Douam *et al.*). Entry requires viral interactions with specific receptors, as delineated in these chapters. Enveloped viruses can potentially enter either by fusing at the cell surface or by first following one of several distinct endocytic routes and then fusing with the endocytic vesicle. The exact mechanisms have been hotly debated for many viruses and these chapters bring new clarity and perhaps some surprises.

Then we shift the scope somewhat and consider the evolution of the entry mediator of HIV, viz., its envelope glycoprotein, Env. Now Env is extremely variable and capable of modulating its interactions with various host molecules: with mannose C-type lectins, which are possibly involved in attachment and transmission, and with the main receptor for the virus, CD4, as well as with the obligate coreceptors, which the virus fastidiously picks among a subset of the seven-transmembrane chemokine receptors. The strengths of the receptor interactions evolve concomitantly with the selection pressure that waxes and wanes as the virus escapes from the coevolving specificities of neutralizing antibodies and gets transmitted to immunologically naïve host organisms (Chapter "The Evolution of HIV Interactions with Coreceptors and Mannose C-Type Lectin Receptors," Borggren and Jansson).

Having obliquely touched on neutralization, we then narrow the focus to what is probably the quantitatively best understood example of how antibodies block viral infectivity, i.e., neutralization of flaviviruses: in Chapter "A Game of Numbers: The Stoichiometry of Antibody-Mediated Neutralization of Flavivirus Infection," Pierson and Diamond analyze the fine stoichiometric details of neutralizing antibody binding to flavivirions and explain why the same antibodies can either neutralize or enhance infectivity depending on what numbers bind to the virion.

We continue the theme of neutralization but switch to the naked adenoviruses, common causes of gastroenteritis, conjunctivitis, otitis, and respiratory tract infections. In Chapter "TRIM21-Dependent Intracellular

Antibody Neutralization of Virus Infection," McEwan and James describe the groundbreaking discovery that the cytoplasmic factor TRIM21 joins antibodies to effect cytoplasmic neutralization of adenovirus. TRIM21 might also augment the antibody-mediated neutralization of other naked viruses. That cytoplasmic neutralization occurs has long been suggested, even for enveloped viruses, but without decisive evidence; such claims have sometimes been erroneously linked to the kinetics and stoichiometry of neutralization.[12] But the newly discovered definitive mechanism, which depends on the traversal of antibody–capsid complexes into the cytoplasm, has its own distinct quantitative implications.

We then extend the consideration of postentry events to later steps in the replicative cycle, including viral assembly and release. The first example is how picornaviruses, although they as naked viruses lack membranes in their virions, interact with intracellular membranes and highjack components of the secretory pathway for their replication (Chapter "Picornavirus–Host Interactions to Construct Viral Secretory Membranes," Greninger). The story then returns to enveloped viruses in the form of retroviruses and the extensive cast of auxiliary factors they have evolved to counteract cellular barriers to their replication (Chapter "Retroviral Factors Promoting Infectivity," Cuccurullo *et al.*). Thereafter, the tale turns to the cytoplasmic domains of the retroviral Env proteins (Chapter "The Cytoplasmic Tail of Retroviral Envelope Glycoproteins," Tedbury and Freed). These cytoplasmic and intravirional tails are particularly long among the lentiviruses, to which HIV belongs. They contain motifs for endocytosis and trafficking of the Env proteins; they even exert transmembraneous conformational effects on the outer Env, the target for neutralizing antibodies. Toward the end of the replicative cycle, when Env gets incorporated into the viral envelope, these tails juxtapose the internal Gag precursor that drives the budding of virions from the cell surface. Furthermore, when retroviruses and other enveloped viruses assemble and egress, they usurp multiple cellular factors, evincing quintessential parasitism.

The scene is then set for some analyses of the free virus particles themselves. First, the classic virological measurement of inert-to-infective particle ratio is examined in general and for particular viruses (Chapter "Molecular determinants of the ratio of inert to infectious virus particles," Klasse). Then, taking the primate lentiviruses, which include HIV, as examples, Regoes and Magnus quantitatively dissect the contributions of individual Env subunits to the function of Env trimers, and of trimers to virion infectivity. These insights segue into analyses of the probabilities that inocula containing

certain infectious doses establish infection in the host organism (Chapter "The Role of Chance in Primate Lentiviral Infectivity: From Protomer to Host Organism").

The ascent from the molecular determinants of individual virion infectivity up to the establishment of infection at the level of a host organism, thus crowning the accounts of the progression through the viral replicative cycle at the cellular level, finally ushers in the topic of virus–host coexistence. Viruses often cause disease. Their interactions with the innate and adaptive immune systems modulate their pathogenesis. Host and virus have evolved together, sometimes for a long time. Herpesviruses may have diverged into the three families alpha-, beta-, and gammaherpesvirinae 180–220 million years ago, cospeciations among mammals having continued during the past 80 million years.[13] In spite of those time lapses, herpesviruses can still get on our nerves (as when herpes simplex virus survives in the ganglion Gasseri or Varicella-Zoster virus gives facial palsy). Although far from perfect, the host's adaptation to these longtime companions is sophisticated. Thus, the vast majority of humans carry latent infections with Epstein–Barr virus but are symptom-free. And the herpesviruses have developed intricate interactions with the host-immune system in long-running evolutionary games with continually tied outcomes. As described in Chapters "Virus-Encoded 7 Transmembrane Receptors" by Mølleskov-Jensen *et al.* and "EBV, the Human Host, and the 7TM Receptors: Defense or Offense?" by Arfelt *et al.*, herpesviruses not only encode seven-transmembrane receptors—with similarities to the chemokine receptors usurped by HIV—but also modulate the expression of the host-cell genes for such receptors.

From the oldest to the newest, the molecular interactions underlying viral propagation are biologically fascinating. Many are also medically consequential. Better knowledge of those interactions can save lives. And structural knowledge of viruses guides rational vaccine and drug design, generating paradigms of translational science.

I am deeply grateful not only to all the contributing authors of this volume for their splendid work but also for the patient assistance by Helene Kabes and Mary-Ann Zimmerman at Elsevier and last but not least to Dr. Michael Conn, the Editor of *PMBTS*, who gave me the opportunity to take on this rewarding project.

P.J. Klasse
Department of Microbiology and Immunology, Weill Cornell Medical College, Cornell University, New York, USA

REFERENCES

1. Burton DR, Parren PW. Fighting the Ebola virus. *Nature*. 2000;408:527–528.
2. Bhattacharyya S, Warfield KL, Ruthel G, Bavari S, Aman MJ, Hope TJ. Ebola virus uses clathrin-mediated endocytosis as an entry pathway. *Virology*. 2010;401:18–28.
3. Bhattacharyya S, Hope TJ. Cellular factors implicated in filovirus entry. *Adv Virol*. 2013;2013:487585.
4. Kondratowicz AS, Lennemann NJ, Sinn PL, et al. T-cell immunoglobulin and mucin domain 1 (TIM-1) is a receptor for Zaire Ebolavirus and Lake Victoria Marburgvirus. *Proc Natl Acad Sci USA*. 2011;108:8426–8431.
5. Wahl-Jensen V, Kurz S, Feldmann F, et al. Ebola virion attachment and entry into human macrophages profoundly effects early cellular gene expression. *PLoS Negl Trop Dis*. 2011;5:e1359.
6. Xu W, Edwards MR, Borek DM, et al. Ebola virus VP24 targets a unique NLS binding site on karyopherin alpha 5 to selectively compete with nuclear import of phosphorylated STAT1. *Cell Host Microbe*. 2014;16:187–200.
7. Lee JE, Fusco ML, Hessell AJ, Oswald WB, Burton DR, Saphire EO. Structure of the Ebola virus glycoprotein bound to an antibody from a human survivor. *Nature*. 2008;454:177–182.
8. Oswald WB, Geisbert TW, Davis KJ, et al. Neutralizing antibody fails to impact the course of Ebola virus infection in monkeys. *PLoS Pathog*. 2007;3:e9.
9. Parren PW, Geisbert TW, Maruyama T, Jahrling PB, Burton DR. Pre- and post-exposure prophylaxis of Ebola virus infection in an animal model by passive transfer of a neutralizing human antibody. *J Virol*. 2002;76:6408–6412.
10. Shedlock DJ, Bailey MA, Popernack PM, Cunningham JM, Burton DR, Sullivan NJ. Antibody-mediated neutralization of Ebola virus can occur by two distinct mechanisms. *Virology*. 2010;401:228–235.
11. Rivers TM. Filterable viruses a critical review. *J Bacteriol*. 1927;14:217–258.
12. Klasse PJ. Neutralization of virus infectivity by antibodies: old problems in new perspectives. *Adv Biol*. 2014;2014:1–24, Article ID 157895.
13. McGeoch DJ, Cook S, Dolan A, Jamieson FE, Telford EA. Molecular phylogeny and evolutionary timescale for the family of mammalian herpesviruses. *J Mol Biol*. 1995;247:443–458.