The last half century has witnessed the development of vaccines effective against a number of viral pathogens that once brought suffering or death to millions. The development of improved diagnostic methodology coupled with clearer understanding of the mechanisms by which viruses are transmitted and the genetics of disease resistance have resulted in improved human and animal health and greater crop yields. Recent progress in the development of antiviral drugs is also promising. In spite of these advances, however, viral diseases continue to have a significant social impact, for which AIDS is the most recent tragic example.

During the last two decades virology has also played a central role in the development of modern molecular biology. Viruses possess limited genetic information and depend on many host-cell functions for replication. As a result, common principles of gene expression apply equally well to the viral and cellular genome. Since viral systems are simple and more easily manipulated than cellular systems, they have been the choice for studying many fundamental biological processes. Results obtained with both bacteriophages and eukaryotic viruses have been almost immediately applicable at the more complex cellular level. Examples range from DNA replication and RNA splicing and capping to membrane structure and function.

There have been great strides in our understanding of structural aspects of viruses. As of this date, the entire nucleotide sequence of several DNA and RNA viral genomes has been determined, and the atomic structures of two picornaviruses have been determined within 0·3 nm resolution by X-ray crystallography. In addition, the genetics of RNA viruses can now be dissected by the introduction of an artificial DNA phase into the replication cycle. Armed with new information and techniques, virologists are also finding unique applications for viruses as genetic vectors to be introduced into cells and whole animals, as building blocks for the construction of multivalent synthetic vaccines and as probes for understanding the molecular basis of cell differentiation and the regulation of cell division, to name a few.

While the suggestion that an issue of *BioEssays* be devoted to virology was appealing and timely, it was also perplexing. Clearly, one issue cannot cover the complexity of contemporary virology, and there are many important systems that could not be represented. However, all of the mini reviews in this special issue illustrate the manner in which analysis of viral systems can illuminate fundamental principles and relationships in modern biology, and how this information can be used to advance both scientific and practical goals.

In the first review Jenkins and Roizman describe procedures for introducing mutations at specific sites in the genomes of large eukaryotic DNA viruses. Interestingly, the methods exploit an understanding of the biology of the bacteriophage Mu and a convenient selectable marker (Tk) derived from an animal virus. Next, Piccini and Paoletti show how a knowledge of the molecular biology of vaccinia virus and genetic manipulation of that system has provided an opportunity for development of novel vaccines. Moreover, further studies using the resulting recombinants should lead to new insights into the mechanisms of control of gene expression of this large DNA virus. In a related vein, Eli Gilboa describes how retroviruses can be used as vectors for foreign genes and enumerates both the advantages and pitfalls in such applications. However, the overall theme is positive, since the utility of this system is already demonstrated. Again, as with vaccinia, observations made during development of these vectors have provided new insight into the molecular biology of retroviruses—a class of particular interest because of its oncogenic and disease potential. In Michael Lai’s article we find in the cytoplasmic RNA coronaviruses an example of what seems at first to be a completely novel, alternative mechanism to nuclear RNA splicing. We see that ‘leader-primed RNA transcription’, in some form or other, is much more common than first noted, and that this mechanism may have been exploited by the African trypanosomes to help create the variant surface glycoproteins that allow this parasite to elude its host immune system.

The last three reviews span the plant and animal
kingsdoms. It is unfortunate that we could not include summaries of work on other important plant viruses, which could be the topic of another special issue. The review by van Kammen and Eggen is noteworthy, not only because of the intrinsic interest in the cowpea mosaic virus and its split-RNA genome which is encapsidated into separate particles, but also because of the unexpected relatedness of the comovirus genome to the single RNA genome of the animal picornaviruses. It is clear that future research efforts in both systems will complement each other in important ways. Vincent Racaniello reviews recent studies with vaccine stains of the polio picornavirus which reveal elements that affect the neurovirulence of the wild-type virus. Since both non-coding as well as capsid-coding viral RNA sequences are important, the phenomenon is obviously complex. Cell-specific 'factors', which may control gene expression, as well as surface receptors are likely involved. Finally, Richard Colonno describes how studies using monoclonal antibodies have provided important new information concerning the number and complexity of surface receptors for a variety of picornaviruses. These studies provide clues to the architecture of the virus–receptor interaction and suggest ways to explore its molecular basis further.

The importance of viral research is obvious from the many journals devoted solely to this topic and from the contribution of virologists to many areas of molecular biology, medicine and agriculture. The recent formation of the American Society for Virology provides further evidence of the need to create a forum for expanding work in this area. The upcoming International Virology Congress in Edmonton promises to be lively and exciting and we encourage our interested colleagues to participate in this worldwide convocation.

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**REVIEW ARTICLES**

**Site-specific Mutagenesis of Large DNA Viral Genomes**

Frank J. Jenkins and Bernard Roizman

**Summary**

Site-specific or target-specific mutagenesis of viral DNA genomes, using a selectable marker system is a powerful tool for the analysis of the function of specific regions of large DNA genomes. Through these techniques the construction of vectors capable of delivering vaccines for the prevention of infectious disease in humans and animals is possible.

**Introduction**

Viral genomes encode numerous proteins that serve to replicate and package the viral nucleic acids and to regulate, in trans, the entire process of multiplication. The genomes encode, in addition, cis-acting signals that are recognized both by viral and host factors essential for viral multiplication. Early studies designed to elucidate the function(s) of virus-specific proteins and DNA sequences relied on the generation and characterization of temperature sensitive (ts) mutants. Since the isolation of ts mutants is dependent on phenotypical changes or inhibition of viral replication at elevated temperatures, the majority of viral genes which can be studied by the aid of ts mutants are those that are essential for a productive virus infection in cell culture. Many viruses are capable of a productive infection in many different cellular environments and it is, therefore, conceivable that viral functions required in one cellular environment may be redundant in another because of the presence of host proteins that can substitute for the function performed by viral gene products. In these instances, mutations altering the temperature sensitivity of the viral protein would be detected only in the cellular environment that does not supply that function. Cells in culture often contain a wide variety of functions and thus attempts to introduce conditional lethal mutations may fail.

An alternative approach to the identification of virus-specific functions involves site-specific mutagenesis of the virus genome by the insertion or deletion of DNA sequences. Site-specific mutagenesis offers several advantages: (i) the procedures allow for either site-specific or target-specific mutagenesis of the viral DNA genome, thereby interrupting a specific viral gene(s) or DNA sequence; (ii) mutants produced by the insertion or deletion of DNA sequences can be used for studies on genes in which ts mutations would not be detected; and (iii) site-specific mutagenesis can be used to modify genomes for use as vaccines or as vectors for foreign genes. These procedures can be done directly on the DNA genomes of small viruses that can be cloned in their entirety. The procedure becomes more complex if the insertional mutagenesis