definition for smallpox provides a framework for developing animal models of OPV infection that can be used to evaluate effectiveness of new OPV therapeutics. Lesion formation, viremia and mortality are clinically relevant endpoints that can be used to evaluate the efficacy of OPV therapeutics in animal models. Although these endpoints can be used to evaluate the efficacy of therapeutics in animal model systems, their predictive value for disease outcome in humans has not been determined.

The most relevant animal models of OPV infection involve the use of host-adapted virus inoculated at a peripheral site to establish infection that spreads to distal sites in a manner similar to variola virus infection of humans. Host adaptations are defined as genetic alterations that result in increased virulence for a specific host. These changes may include evolutionary adaptations, such as acquisition of host response modifier functions or unknown changes that result in increased virulence. Models that use host-adapted virus in a natural host are appealing because virus replication in target tissue and systemic spread is determined by the host response to infection. Some animal models of orthopoxvirus infection have been developed using non-host-adapted OPVs to establish disease. These models require inoculation of animals with large quantities of virus, sometimes delivered by unnatural routes. Delivering large amounts of virus by unnatural routes alters pathogenesis by allowing virus access to different tissues and stimulating a non-natural host response.

A number of models of OPV infection have been developed in different animal species such as mice, including BALB/c, NMRI, ANC/R and Nu/nu strains, rabbits, prairie dogs and ground squirrels (reviewed by Chapman *et al.*[36] and Smee[37]). These models have been used to evaluate the antiviral activity of OPV therapeutics against multiple species of orthopoxviruses, including vaccinia virus strains IHD-J, Lister and WR, ectromelia virus, strain Moscow, cowpox virus, rabbitpox virus and monkeypox virus.[37] Infections were established by a variety of routes, including intranasal, intravenous, intradermal, subcutaneous and aerosol delivery of virus.

4.3.2.1 Vaccinia and Cowpox Virus Mouse Models

Mouse models using vaccinia virus or cowpox virus have been developed to measure the efficacy of anti-poxvirus compounds.[37] The pathogenesis of infection is dictated by the route of viral entry and models using intracranial, intravenous or intranasal inoculation have been described.[37] Intranasal inoculation of mice with vaccinia virus or cowpox virus produces local replication in the nasal tissue and systemic spread of the virus to distal sites, providing a system capable of assessing antiviral activity of compounds that inhibit multiple steps in the virus life cycle. Infections are established by inoculation of mice with 1×10^4–1×10^6 plaque-forming units (PFUs) of virus in a small volume (10 μL) to each naris. Although a large dose of virus is required to establish lethal infection, replication of the virus starts locally in the nasal tissue and lungs before spreading systemically through the reticuloendothelial system.[38] High levels of virus can be found in liver, spleen, lung and