The WHO Collaborating Center for Smallpox and other Poxviruses at the Centers for Disease Control and Prevention Atlanta, GA: 2009 report on the use of live Variola virus to evaluate therapeutic modalities: *in vitro* studies

Victoria A. Olson Ph.D, Scott K. Smith M.S., Kevin L. Karem Ph.D., Inger K. Damon M.D., Ph.D.

Poxvirus Program, Centers for Disease Control and Prevention Atlanta, GA

There is a need for anti-smallpox therapeutic options for treatment and/or post-exposure prophylaxis, which supports gaining molecular biological insight into potentially therapeutic compounds. Current recommendations by the U.S. government call for two antiviral compounds, with separate mechanisms of action, to be licensed and available for use. Compounds specifically targeting viral proteins, viral processes, or cellular functions required by the virus but non-essential for the human host are presently of greatest interest. The World Health Organization (WHO)-approved research activities for the WHO Collaborating Center for Poxviruses in Atlanta, GA include the testing and identification of antiviral compounds that are successful in inhibiting variola growth or aspects of viral propagation. Several studies have demonstrated that cellular targeted chemotherapy may be protective to treat variola virus infection or enable prophylaxis. Tyrosine kinase inhibitory compounds, such as CI-1033 and Gleevec, evaluated for or in use for treatment of human cancers, have shown promise in animal model treatment studies of systemic orthopoxvirus infection. Development of antivirals or

immunotherapeutics that directly target variola virus components of infection include studies of Cidofovir and its derivatives, which likely target the DNA polymerase, and ST-246, which targets a viral particle assembly protein. Critical steps to evaluate such therapeutics require *in vitro* characterization of their activity against live variola virus infection. Previous work with live variola virus infection has characterized ST-246 (and ST-246 isomers) effect upon virus *in vitro* phenotype, and the compounds ability to neutralize progeny virus. Inhibitors of tyrosine kinases have similarly been evaluated for effect upon virus *in vitro* phenotype, and inhibition of viral progeny. Work during 2009 has focused on identifying the mechanism of action of these promising anti-viral compounds targeting cellular tyrosine kinases. Cytoskeletal staining studies indicate that these kinase inhibitors prevent the polymerization of actin into "tails" and may thereby interfere with the release of viral progeny from variola infected cells. This work confirms and extends previous studies with vaccinia and monkeypox virus showing that variola virus utilizes similar pathways of viral egress from infected cells, confirming this mechanism as a target for antiviral therapy. *In vitro* characterization provides critical information for the development of effective antiviral compounds against Variola, which may provide life saving options in the event of a bioterrorist release.