Identification of Inhibitors of Ebola Virus with a Subgenomic Replication System.

J. Dyall[1], J. Balsarotti[1], B. Buscher[1], R. Roth[1], G. Starkey[1], A. O'Guin[1], J. Paragas[2], U. Slomczynska, and P. D. Olivo[1]

[1]Apath, LLC, Saint Louis, MO

[2]United States Army Medical Research Institute for Infectious Diseases, Virology Division, Fort Detrick, MD

Filoviruses, which include Ebola virus and Marburg virus, are among the most notorious human pathogens because they cause sporadic outbreaks of severe hemorrhagic fever. Unfortunately, very few therapeutic agents are available to treat infections with these viruses. Antiviral screening methods that determine the effect of compounds on viral replication involve working with infectious virus which is obviously not practical for these biosafety level 4 (BSL-4) agents. We developed an antiviral screening method based on a cell-based, infection-independent, Ebola subgenomic replication system in which the expression of an easily measurable enzyme is dependent on the RNA replication and transcription factors of Ebola virus. Using this system we screened a synthetic compound library for antiviral activity against Ebola virus and have identified a number of inhibitors. Anti-Ebola virus activity for many of the inhibitors was confirmed in a viral replication assay using a GFP-expressing Zaire '76 strain of Ebola virus. Sixty-two small molecule inhibitors from nine classes of compounds had EC_{50} values in the low micromolar range and good selectivity. Several of these compounds have promising chemical, biological, and pharmacological profiles to pursue as potential anti-filovirus drugs. The aryl secondary sulfonamide and 4-aminoquinoline classes were chosen for lead optimization to improve antiviral potency and selectivity. Ten compounds from the original lead candidates and the SAR series had favorable PK profiles and were tested in a mouse model of Ebola virus. Ongoing studies to advance this drug discovery program will be discussed. (Supported by NIH R44 AI052917-02 and R01 AI066502-01)