survival rates. The other monotherapy and double combination regimens provided no protection or only partial protection (0–60% survival) in each study. A comparison of the Kaplan–Meier survival curves showed that TCAD provided significantly better protection ($P < 0.05$) than all other regimens in both studies. TCAD produced survival rates in each study that were greater than the additive rates of each drug as a monotherapy, indicative of synergy. Moreover, TCAD reduced the magnitude of weight loss in infected animals significantly relative to all other treatments. Importantly, AMT contributed to the efficacy of TCAD against the AMT-resistant novel A/H1N1 virus. The TCAD regimen is highly active in two lethal mouse influenza treatment models against susceptible and resistant viruses. These results validate and build upon the previously demonstrated superior *in vitro* efficacy of TCAD versus monotherapy and double combination regimens and translate them into an *in vivo* model.

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In Vitro and *In Vivo* Efficacy of Combinational Therapy with Favipiravir (T-705) and Oseltamivir Against Influenza A/CA/04/09 Pandemic H1N1 Virus

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Serious disease caused by emerging pandemic influenza A (H1N1) viruses and the possibility of drug resistance underscores the need for different approaches for treating influenza infections. The augmentation of current monotherapies with newly developed agents targeting alternative functions of the influenza virus replication cycle may be a solution. The current studies demonstrate the efficacy of using a combination of two drugs with different mechanisms of action to enhance total anti-H1N1 influenza A activity compared to each compound alone. Combinations of oseltamivir, a currently used, clinically approved neuraminidase inhibitor and favipiravir, an experimental viral RNA polymerase inhibitor, were evaluated alone and in combination for *in vitro*/*in vivo* efficacy against a pandemic H1N1 influenza A virus. *In vitro* combination studies revealed synergy with 0.032–1.0 μM oseltamivir combined with 0.32–10 μM T-705. In an H1N1 lethal mouse model it was found that: (1) orally administered combinations of favipiravir at 30–0.3 mg/kg/day and oseltamivir at 3 mg/kg/day resulted in significant protection against death ($P < 0.001$) and in total survivors ($P < 0.05$–0.01), (2) the combinations of favipiravir and oseltamivir at higher doses ameliorated the weight loss attributable to virus infection, (3) the combinations of favipiravir and oseltamivir at higher doses were highly synergistic, and (4) the use of favipiravir at 30 mg/kg/day or higher may permit the use of lower doses of oseltamivir to achieve efficacy against pandemic H1N1 viruses. The results suggest that these two compounds could be used in combination to treat serious infections in humans caused by pandemic H1N1 viruses.

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Antiviral Activity of Leflunomide Against Respiratory Syncytial Virus

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Respiratory syncytial virus (RSV) is a major cause of often serious, fatal respiratory disease in infants and young children, organ transplant recipients, patients suffering cystic fibrosis or congenital heart disease, and the elderly. Severe RSV disease is characterized by a disproportionately intense pulmonary inflammatory response resulting in bronchiolar injury and compromise of airway function. Current treatment options are limited to ribavirin, which must be administered by small-particle aerosol for 12–18 h per day for 3–7 days, and passive immunoprophylaxis with monoclonal antibody specific for the RSV fusion protein (palivizumab), neither of which has been shown to reduce mortality. Leflunomide is an orally bioavailable anti-inflammatory drug approved for treatment of rheumatoid arthritis and currently in clinical trials as an immunosuppressant in transplant recipients. We have previously demonstrated that leflunomide exerts potent antiviral activity against CMV, HSV, and polyomavirus BK. We now report on the antiviral activity of this agent against RSV. Phase contrast microscopy and immunohistochemical staining demonstrated nearly complete attenuation of RSV-induced syncytia formation in infected human airway epithelial cell cultures treated with A77 1726, the active metabolite of leflunomide. Plaque assay of virus yield in RSV-inoculated cultures demonstrated potent, dose-dependent A77-mediated reduction in virus production. Likewise, pulmonary viral loads in RSV-inoculated cotton rats were reduced by >3 logs by leflunomide compared with vehicle-treated controls, even when leflunomide treatment was delayed until day 3 post-inoculation. Real-time rt-PCR demonstrated A77-mediated inhibition of viral genomic RNA synthesis and inhibition of transcription of several viral genes. Data generated by these experiments implicate leflunomide as a unique bifunctional agent with potential to both reduce viral load and, by virtue of its well-documented anti-inflammatory activity, attenuate the destructive inflammation associated with RSV disease. Sidwell and Barnard have stated that effective therapeutic intervention for severe RSV disease must include both antiviral and anti-inflammatory components. Leflunomide, it seems, effectively meets these criteria.

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Small-molecule Inhibition of Respiratory Syncytial Virus Fusion: *It Takes Two to Tango*

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Six-helix bundle (6HB) formation is an essential step for many viruses that rely on a class I fusion protein to enter a target cell and initiate replication. Because the binding modes of small molecule inhibitors of 6HB formation are largely unknown, precisely how they disrupt 6HB formation remains unclear, and structure-based design of improved inhibitors has thus been very speculative. It is currently believed that such inhibitors completely prevent 6HB formation by binding in a hydrophobic pocket composed of amino