

REMEDESIVIR: AN APPROACH TO FIGHT AGAINST COVID MUTANTS¹Dr. Apoorva Tangri and ^{2*}Dr. Alka Tangri¹Rush University, Chicago, Illinois, USA.²Department of Chemistry, Brahmanand College, Kanpur.***Corresponding Author: Dr. Alka Tangri**

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ABSTRACT

Remdesivir is an antiviral medication that targets a range of viruses. It was originally developed over a decade ago to treat hepatitis C and a cold-like virus called respiratory syncytial virus (RSV). Remdesivir wasn't an effective treatment for either disease. But it showed promise against other viruses. Remdesivir (RDV) is a type of broad-spectrum antiviral medication called a nucleotide analog. It is currently an investigational drug and not approved in any country for any use. COVID-19 is an RNA virus. (RNA is the molecular transcription tool organisms use to build proteins using DNA instructions.) RNA viruses are dependent on an RNA polymerase enzyme to grow the RNA chain. Remdesivir substitutes this RNA polymerase enzyme, meaning the RNA can't develop so the virus cannot replicate itself. Researchers tested remdesivir in clinical trials during the Ebola outbreak. Other investigational medications worked better, but it was shown to be safe for patients. Studies in cells and animals suggested that remdesivir was effective against viruses in the coronavirus family, such as Middle East Respiratory Syndrome (MERS) and Severe Acute Respiratory Syndrome (SARS). Recently By April 2020, early results indicated that remdesivir accelerated recovery for hospitalized patients with severe COVID-19. It became the first drug to receive emergency use authorization from the U.S. Food and Drug Administration (FDA) to treat people hospitalized with COVID-19.

INTRODUCTION

Remdesivir is a carboxylic ester resulting from the formal condensation of the carboxy group of N-[(S)-{[(2R,3S,4R,5R)-5-(4-aminopyrrolo[2,1-f][1,2,4]triazin-7-yl)-5-cyano-3,4-dihydroxytetrahydrofuran-2-yl]methoxy}(phenoxy)phosphoryl]-L-alanine with the hydroxy group of 2-ethylbutan-1-ol. It is a carboxylic ester, a pyrrolotriazine, a nitrile, a phosphoramidate ester, a C-nucleoside and an aromatic amine. It derives from a GS-441524. A broad-spectrum antiviral prodrug with potent in vitro antiviral activity against a diverse panel of RNA viruses such as Ebola virus, MERS-CoV and SARS-CoV. Remdesivir works by interrupting production of the virus. Coronaviruses have genomes made up ribonucleic acid (RNA). Remdesivir is a white to off-white or yellow non hygroscopic solid, practically insoluble in water and soluble in ethanol. Remdesivir has six chiral centres and is produced as a single stereoisomer. Different polymorphic forms exist and the active substance is manufactured as Form II or mixtures of Form II and another crystalline form. The mixture of forms and Form II show similar solubility and do not result in differences in finished product performance. The active substance is dissolved before final I.V. administration. Remdesivir is indicated for the treatment of adult and pediatric patients aged 12 years and over

weighing at least 40 kg for coronavirus disease 2019 (COVID-19) infection requiring hospitalization. Under this indication, remdesivir should only be administered in a hospital or other healthcare setting capable of providing acute care comparable to an inpatient hospital setting.

Remdesivir interferes with one of the key enzymes the virus needs to replicate RNA. This prevents the virus from multiplying.^[1] Researchers began a randomized, controlled trial of the antiviral in February 2020 to test whether remdesivir could be used to treat SARS-CoV-2, the coronavirus that causes COVID-19. By April, early results indicated that remdesivir accelerated recovery for hospitalized patients with severe COVID-19. It became the first drug to receive emergency use authorization from the U.S. Food and Drug Administration (FDA) to treat people hospitalized with COVID-19. Researchers have now completed the trial, known as the Adaptive COVID-19 Treatment Trial (ACTT-1). The study was funded by the National Institute of Allergy and Infectious Diseases (NIAID). The final report appeared in the *New England Journal of Medicine* on October 8, 2020. Scientists randomly assigned 1,062 hospitalized COVID-19 patients to receive remdesivir or a placebo plus standard treatment. The patients received an intravenous infusion of remdesivir or placebo for up to

10 days. The final results showed that the antiviral treatment was beneficial, consistent with the preliminary findings. Patients who received remdesivir were quicker to recover, which was defined as being medically stable enough to be discharged from the hospital. The median recovery time was 10 days with remdesivir compared to 15 days for the placebo group. Patients given remdesivir were more likely to have improved by day 15.

Remdesivir also improved mortality rates for those receiving supplemental oxygen (4% with remdesivir versus 13% with placebo at day 29 of treatment). All-cause mortality among all patients was 11% with remdesivir and 15% with placebo at day 29, but this difference between the treatment groups was not large enough to rule out chance. The preliminary findings hadn't shown an effect on mortality.

The study also suggested that remdesivir treatment may prevent patients from progressing to more severe respiratory disease. Those treated with remdesivir were less likely to need high levels of respiratory support. Remdesivir appeared to most benefit patients who were receiving supplemental oxygen. Remdesivir is a beneficial treatment for patients with COVID-19," says study author Dr. John Beigel of NIAID. "It may also help to conserve scarce health care resources, such as ventilators, during this pandemic."

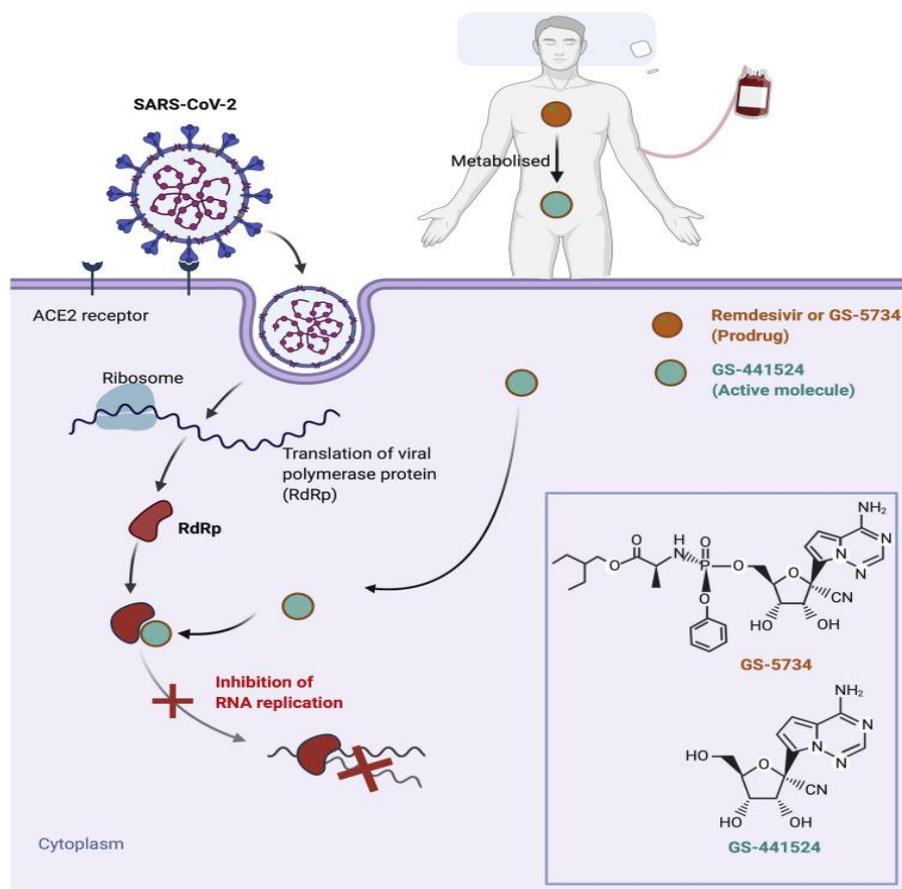
Chemistry and Pharmacology

Remdesivir is a single-diastereomer monophosphoramidate prodrug of a cyanoadenosine nucleoside analogue (GS-441524), a chemical structure that masks the negatively charged phosphate of GS-443902 and facilitates cellular entry. Remdesivir undergoes rapid intracellular conversion to an alanine metabolite (GS-704277), followed by the nucleoside analogue (GS-441524),^[2] and, ultimately, the pharmacologically active nucleoside triphosphate form (GS-443902). GS-443902 acts as an analogue of ATP and competes with the endogenous ATP substrate for incorporation into SARS-CoV's RNA via RdRp. RdRp is a nonstructural protein that is highly conserved among different virus strains, making it an attractive antiviral target.^[3] Remdesivir's primary mechanism of antiviral activity occurs through GS-443902 incorporation into viral RNA chains by RdRp, leading to chain termination and inhibition of viral replication.^[4] A challenge in the development of nucleoside analogues against CoVs is the presence of a unique CoV proofreading 3'→5' exoribonuclease (ExoN) that increases replication fidelity.^[5] In an *in vivo* SARS-CoV infection model, inactivation of ExoN activity due to alanine substitution of the first two active-site residues resulted in 12-fold-reduced replication fidelity.^[6] *In vitro* resistance to ribavirin and 5-fluorouracil among CoVs has been attributed to their removal by the proofreading ExoN.^[7] Thus, an effective nucleoside analogue must evade the proofreading ExoN to prevent CoV viral replication. A

study using a betacoronavirus murine hepatitis virus (MHV) model illustrated that remdesivir was still able to inhibit RdRp even in the setting of intact ExoN.^[8] The authors of that study compared the sensitivity of wild-type (WT) MHV to that of ExoN-negative (ExoN) MHV and revealed that it is modestly less sensitive to remdesivir (50% effective concentration [EC50], 0.019 M versus 0.087 M), suggesting that remdesivir is able to evade ExoN proofreading activity, which could be attributed to higher RdRp selectivity for remdesivir triphosphate than for the natural nucleotides^[8]. This might also indicate that ExoN activity is not sufficient to prevent potent inhibition of CoV replication.^[8]

Mechanism of action

COVID-19 is caused by the positive-sense RNA virus severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). Replication of the viral genome is a key step in the infectious cycle of RNA viruses, including those of the *Filoviridae*, *Paramyxoviridae*, *Pneumoviridae*, and *Coronaviridae* families, and is carried out by viral RNA-dependent RNA polymerase (RdRp) enzymes or enzyme complexes^[9,10]. For both SARS-CoV and SARS-CoV-2, the RdRp comprises nsp7, nsp8, and nsp12 subunits under physiological conditions, although functional RdRp complexes can be reassembled *in vitro* that incorporate only the nsp8 and nsp12 subunits, similar to the Middle East respiratory syndrome coronavirus (MERS-CoV).^[10]



Mechanism of action of remdesivir against covid.

Remdesivir is a phosphoramidite prodrug of a 1'-cyano-substituted adenosine nucleotide analogue that competes with ATP for incorporation into newly synthesized viral RNA by the corresponding RdRp complex.^[10] Remdesivir enters cells before being cleaved to its monophosphate form through the action of either carboxylesterase 1 or cathepsin A; it is subsequently phosphorylated by undescribed kinases to yield its active triphosphate form remdesivir triphosphate (RDV-TP or GS-443902).^[9,11] RDV-TP is efficiently incorporated by the SARS-CoV-2 RdRp complex, with a 3.65-fold selectivity for RDV-TP over endogenous ATP.^[10,11] Unlike some nucleoside analogues, remdesivir provides a free 3'-hydroxyl group that allows for continued chain elongation.^[10,11] However, modelling and *in vitro* experiments suggest that at $i + 4$ (corresponding to the position of the incorporation of the fourth nucleotide following RDV-TP incorporation), the 1'-cyano group of remdesivir sterically clashes with Ser-861 of the RdRp, preventing further enzyme translocation and terminating replication at position $i + 3$. This mechanism was essentially identical between SARS-CoV, SARS-CoV-2, and MERS-CoV, and genomic comparisons reveal that Ser-861 is conserved across alpha-, beta-, and deltacoronaviruses, suggesting remdesivir may possess broad antiviral activity.^[10]

Considerations for the use of nucleotide analogues like remdesivir include the possible accumulation of

resistance mutations. Excision of analogues through the 3'-5' exonuclease (ExoN) activity of replication complexes, mediated in SARS-CoV by the nsp14 subunit, is of possible concern.^[10] Murine hepatitis viruses (MHVs) engineered to lack ExoN activity are approximately 4-fold more susceptible to remdesivir, supporting the proposed mechanism of action. However, the relatively mild benefit of ExoN activity to remdesivir resistance is proposed to involve its delayed chain termination mechanism, whereby additional endogenous nucleotides are incorporated following RDV-TP.^[10] In addition, serial passage of MHV in increasing concentrations of the remdesivir parent molecule GS-441524 led to the development of resistance mutations F476L and V553L, which maintain activity when transferred to SARS-CoV. However, these mutant viruses are less fit than wild-type in both competition assays and *in vivo* in the absence of selective pressure. To date, no clinical data on SARS-CoV-2 resistance to remdesivir have been described.^[11]

Elimination

Remdesivir is 74% eliminated in the urine and 18% eliminated in the feces^[12] 49% of the recovered dose is in the form of the metabolite GS-441524, and 10% is recovered as the unmetabolized parent compound. A small amount (0.5%) of the GS-441524 metabolite is found in feces.^[11]

CONCLUSION

The findings show that remdesivir alone isn't a sufficient treatment for all patients but does provide some benefit. Studies are underway to evaluate remdesivir in combination with other therapies. Remdesivir might be crucial for ensuring an efficient treatment, decrease mortality and allow early discharge in relation to Covid-19. Ongoing randomized, placebo-controlled trials are critical in delineating its efficacy.

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