Biosafe SARS-CoV-2 replicons for high-throughput screening

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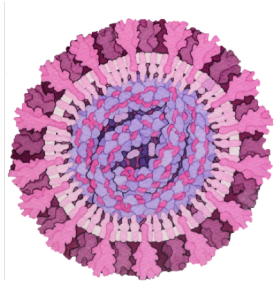
We need broadly-acting coronavirus-specific antivirals

SARS-CoV
2002–2004
>8,000 cases
9.6% case fatality

MERS-CoV
2012–
>2500 cases
34% case fatality

SARS-CoV-2
2019–
>50M cases
~2.4% case fatality



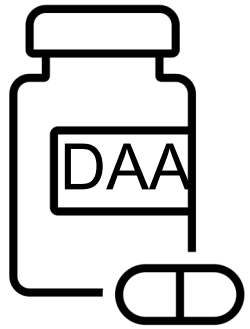
Vaccines generate highly specific neutralizing immune responses.
Antivirals target conserved, essential replication enzymes.

HCV: *the* successful paradigm for antiviral development

non-specific
low cure rates

2010

high
throughput
drug
discovery



>2011

HCV-specific
>95% cure rates

2016 Lasker Award: Ralf Bartenschlager, Charlie Rice, and Michael Sofia

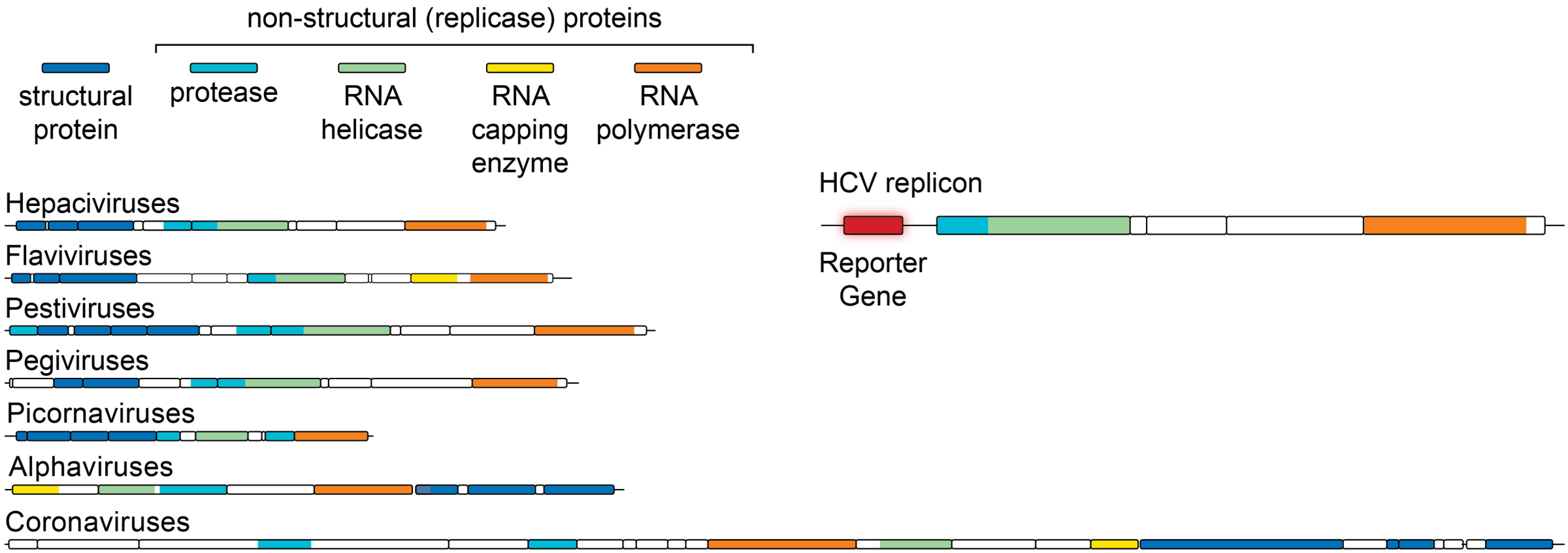
2020 Nobel Prize: Harvey Alter, Michael Houghton, and Charlie Rice

Complete Replication of Hepatitis C Virus in Cell Culture

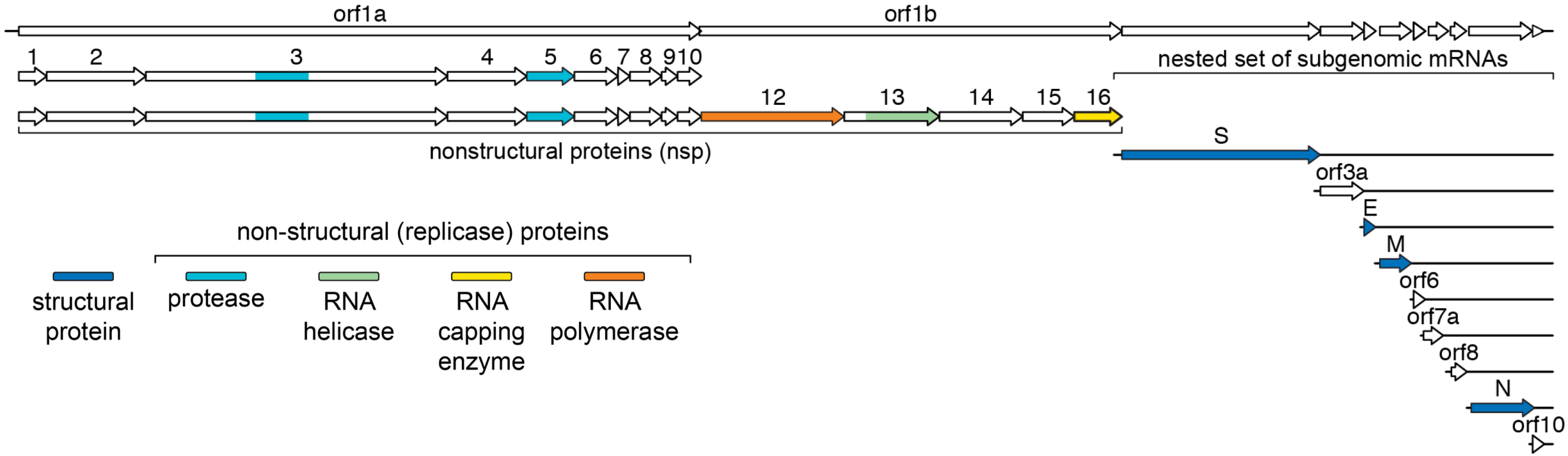
Brett D. Lindenbach, Matthew J. Evans, Andrew J. Syder, Benno Wölk, Timothy Tellinghuisen, Christopher C. Liu, Toshiaki Maruyama, Richard O. Hynes, Dennis R. Burton, Jane A. McKeating, Charles M. Rice

Science 2005

What is an RNA replicon?

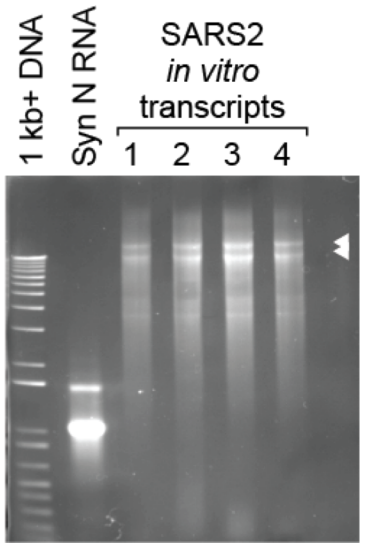


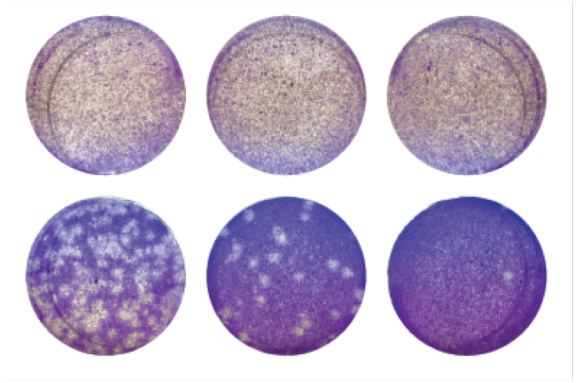
Targets for SARS-CoV-2 antiviral development



Coronavirus replicons initiate replication inefficiently
due to the low specific infectivity (SI) of long RNA transcripts

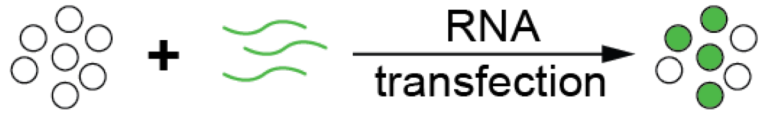
We have vastly improved on genetic reconstruction of SARS-CoV-2





Optimized RNA transcription

Custom, highly permissive cell lines

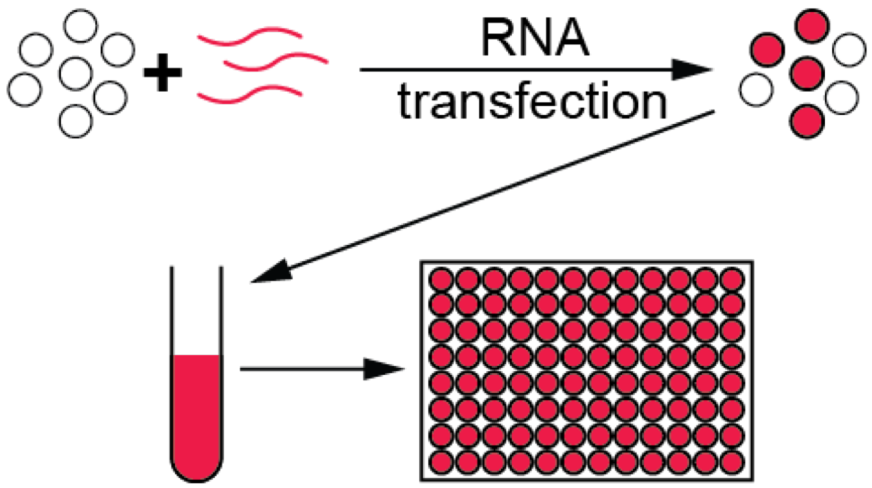


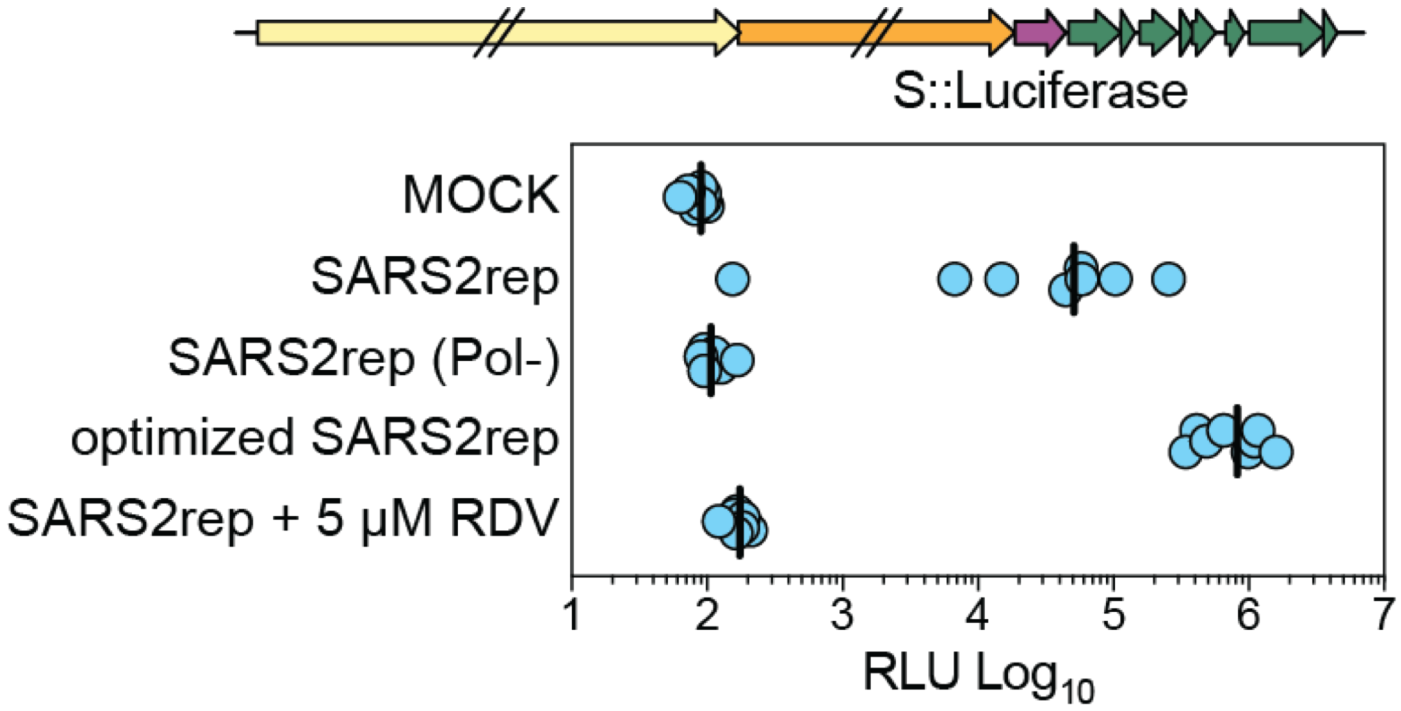
Infectious Center Assay:

- Serially dilute transfected cells into naive cell population.
- Plate and overlay for plaque formation.

transfected RNA		% cells transduced	RNA *SI* (PFU/fmol)	
YFV	(standard)	74	2.7×10^4	
SARS2	standard	0.005	0.39	**~1500x**
SARS2	optimized	7.3	5.5×10^2	
SARS2	control	0	<0.30	

Robust reporter gene expression from our first generation SARS-CoV-2 replicon, SARS2rep

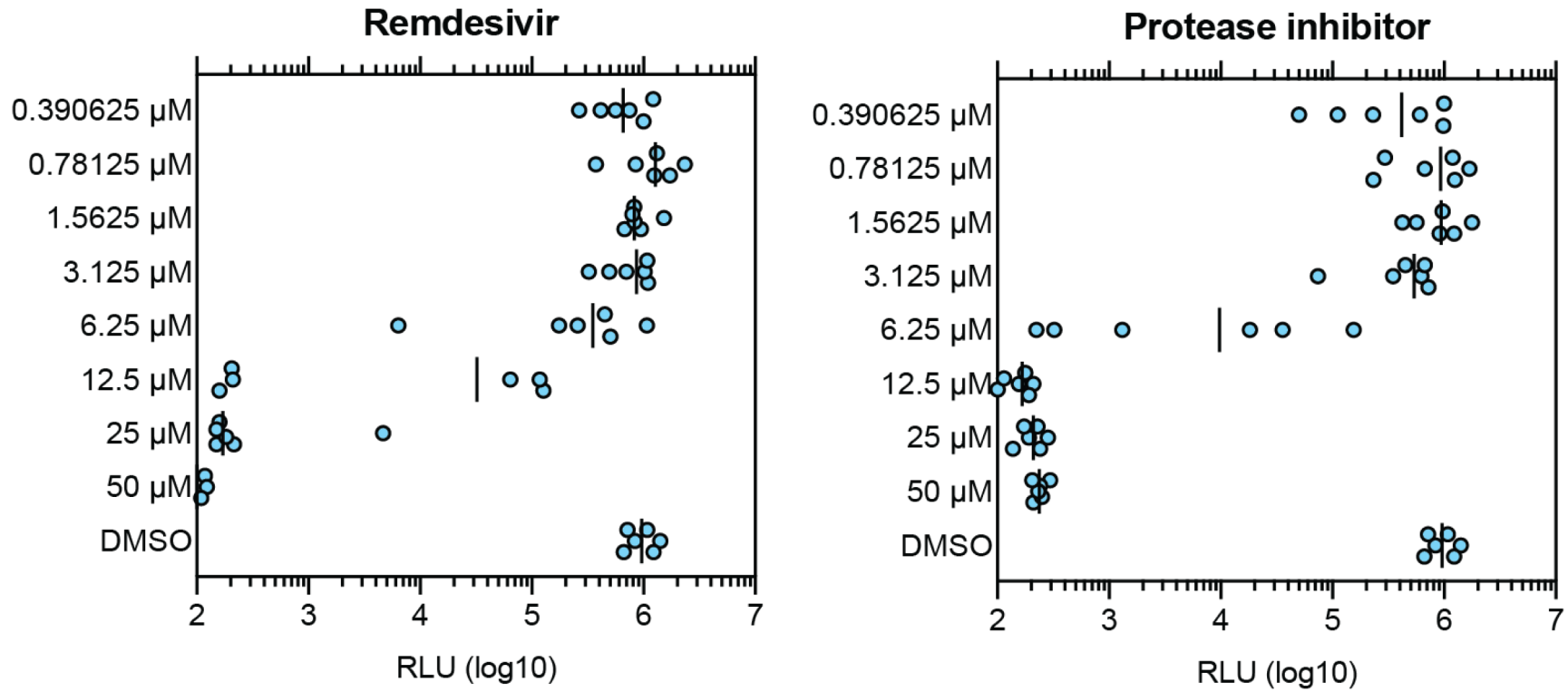




Results are obtained within 24 h

Our replicons do not make infectious virus particles

SARS2rep can be used to measure antiviral efficacy under standard laboratory biosafety conditions



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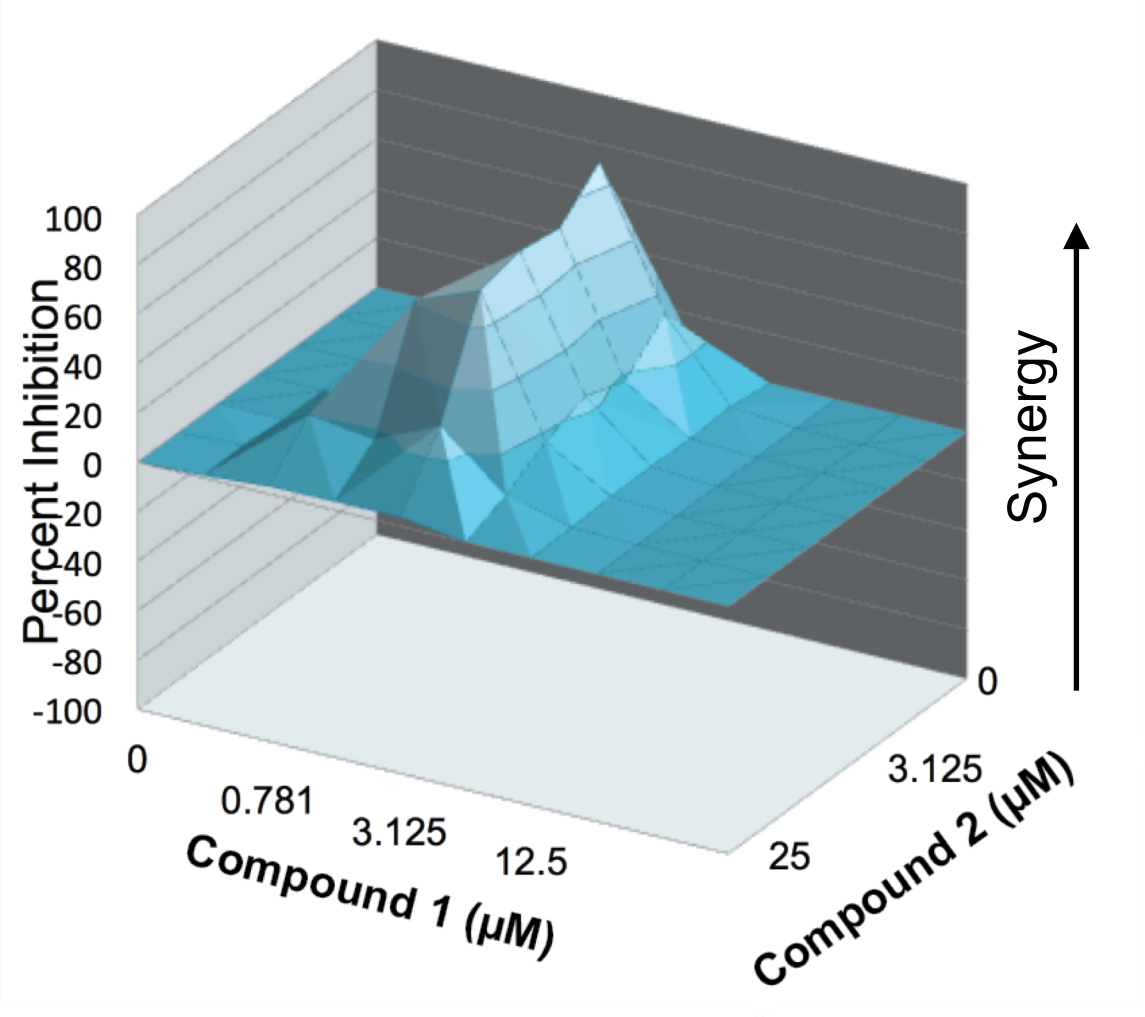
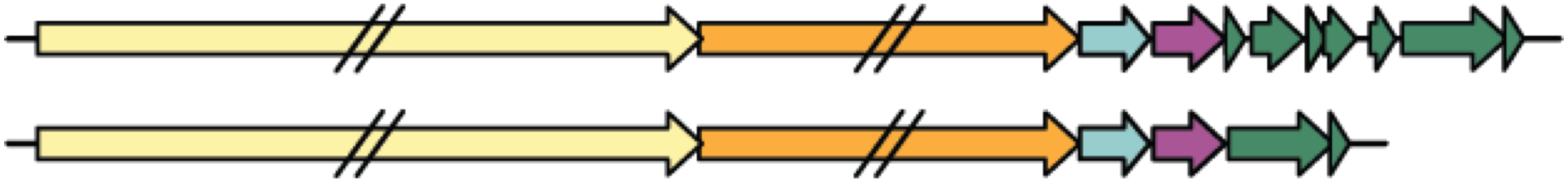


<u>Antiviral drug combinations</u>

1. Different mechanisms of action reduces resistance.

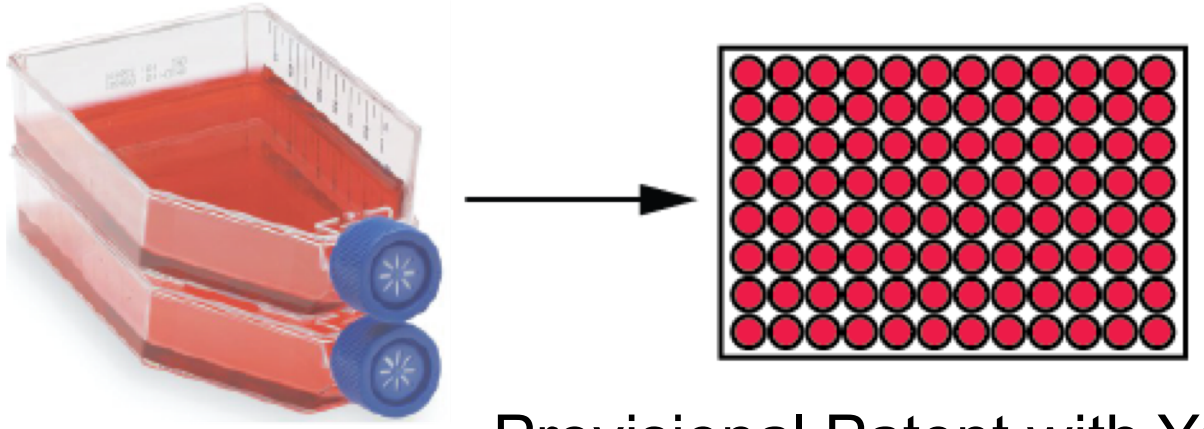
2. Drug-drug synergy increases the effective potency of both drugs.

Second generation SARS2rep: stable cell lines



Dual reporters: luciferase and drug resistance
Noncytopathic mutations in nonessential viral genes.

Our Product:



Provisional Patent with Yale OCR

Blavatnik funds will be used to:

1. Complete optimization of 2nd generation replicons and characterize their long-term stability, etc., ($150K).

2. Partner with an experienced CRO, like Evotec, to conduct pilot screens, demonstrate assay scalability and robustness, and likely reveal novel chemical leads, which will increase our IP ($150K).

3. Market and license our SARS2rep cell lines to pharma and biotech industry for HTS screening.