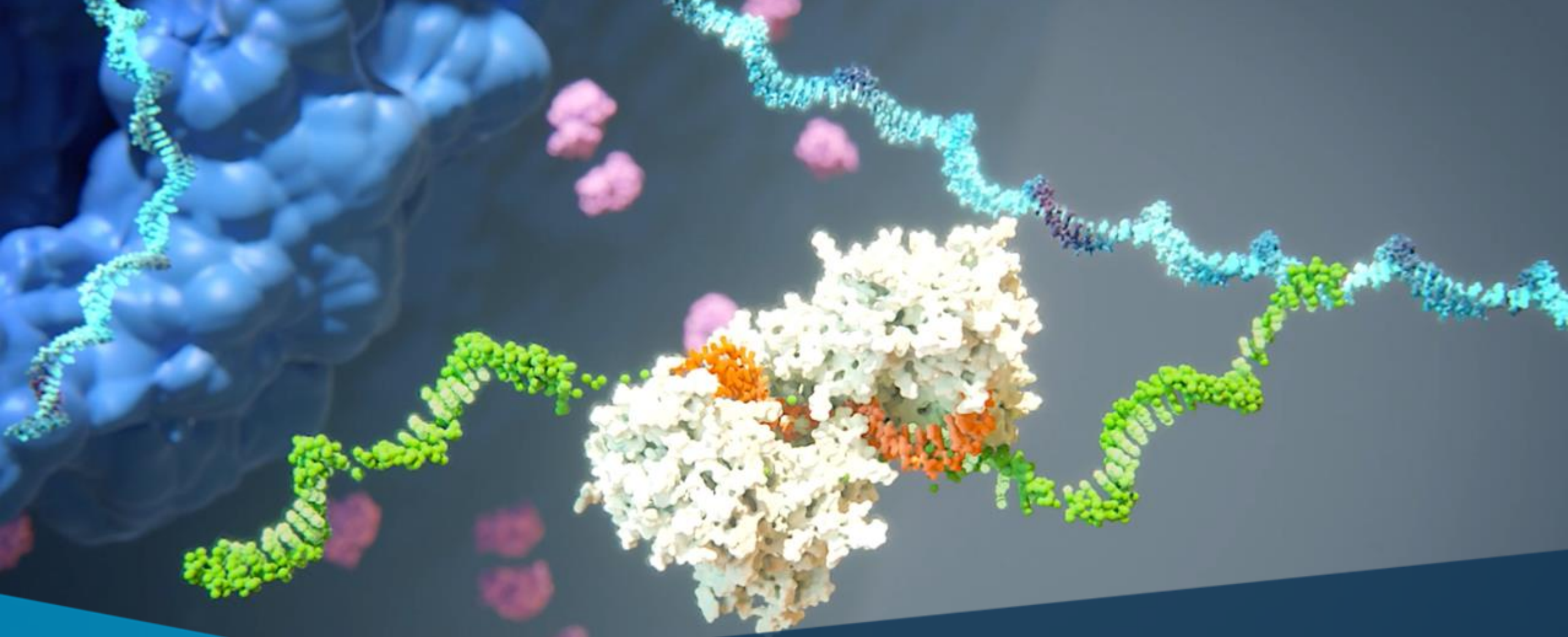


ALN-COV: An Investigational RNAi Therapeutic for COVID-19

Akin Akinc, PhD

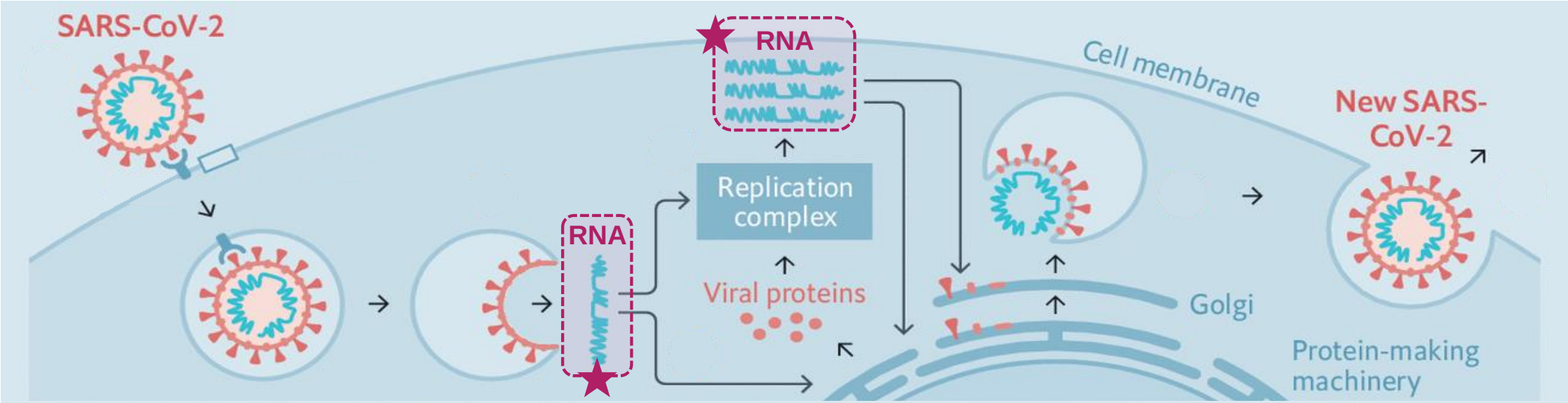
The Oligonucleotide Therapeutic Society Meeting, September 27-30, 2020

Alnylam PHARMACEUTICALS

Disclosures

- Employee of Alnylam Pharmaceuticals

ALN-COV, an Investigational RNAi Therapeutic for COVID-19

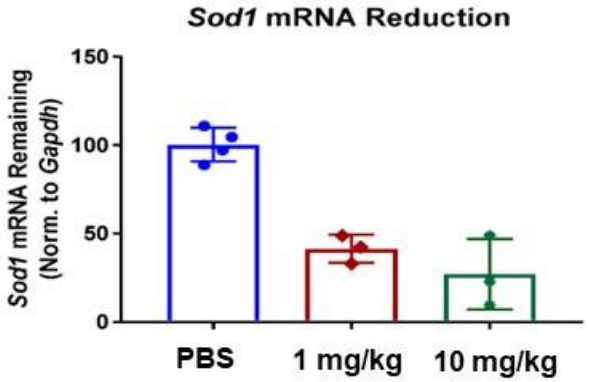


Sources: Song et al., *Viruses*, 2019; Jiang et al., *Emerging Microbes and Infections*, 2012; *The Economist*

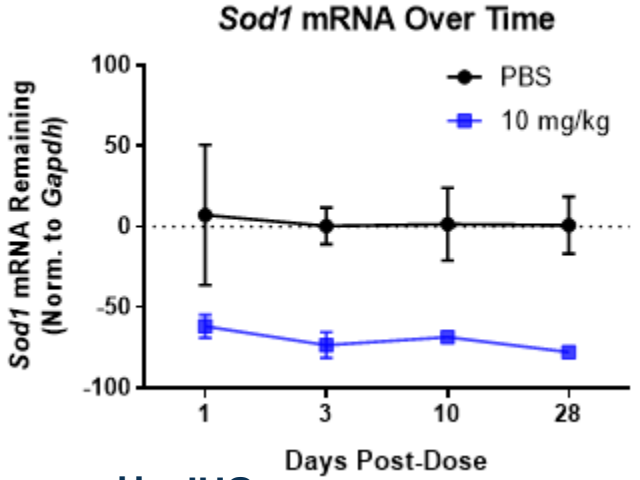
- **Provides a unique and distinct antiviral strategy for the treatment of COVID-19**
  - RNAi mechanism of action results in degradation of SARS-CoV-2 viral RNA genome
- **Direct administration of ALN-COV to lungs, the key site of viral replication and disease manifestations**
  - Early delivery of a potent antiviral agent directly to the site of replication has the potential to prevent progression to severe pulmonary disease and decrease time to clinical recovery
- **Initial development plan focused on the treatment of patients with mild to moderate COVID-19**
  - Potential for development in targeted prophylactic setting
- **IND filing planned for around year-end 2020**

Chemistry Advances Enable Robust Tissue Distribution with Potent and Durable Gene Knockdown in Lung

Surrogate siRNA targeting endogenous lung target (Sod1)

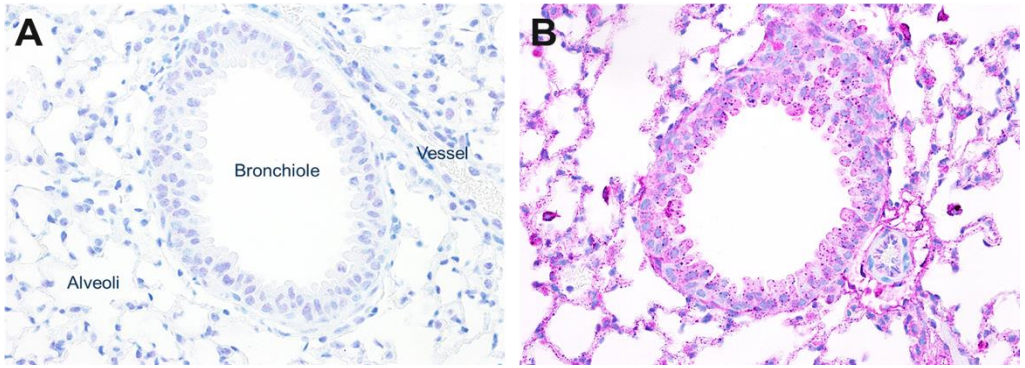


Dose-dependent *Sod1* mRNA reduction in lung following single-doses of *Sod1* siRNA at 1 and 10mg/kg



Sustained *Sod1* mRNA reduction in lung following a single 10mg/kg dose

Sod1 siRNA Distribution in the Mouse Lung Measured by IHC

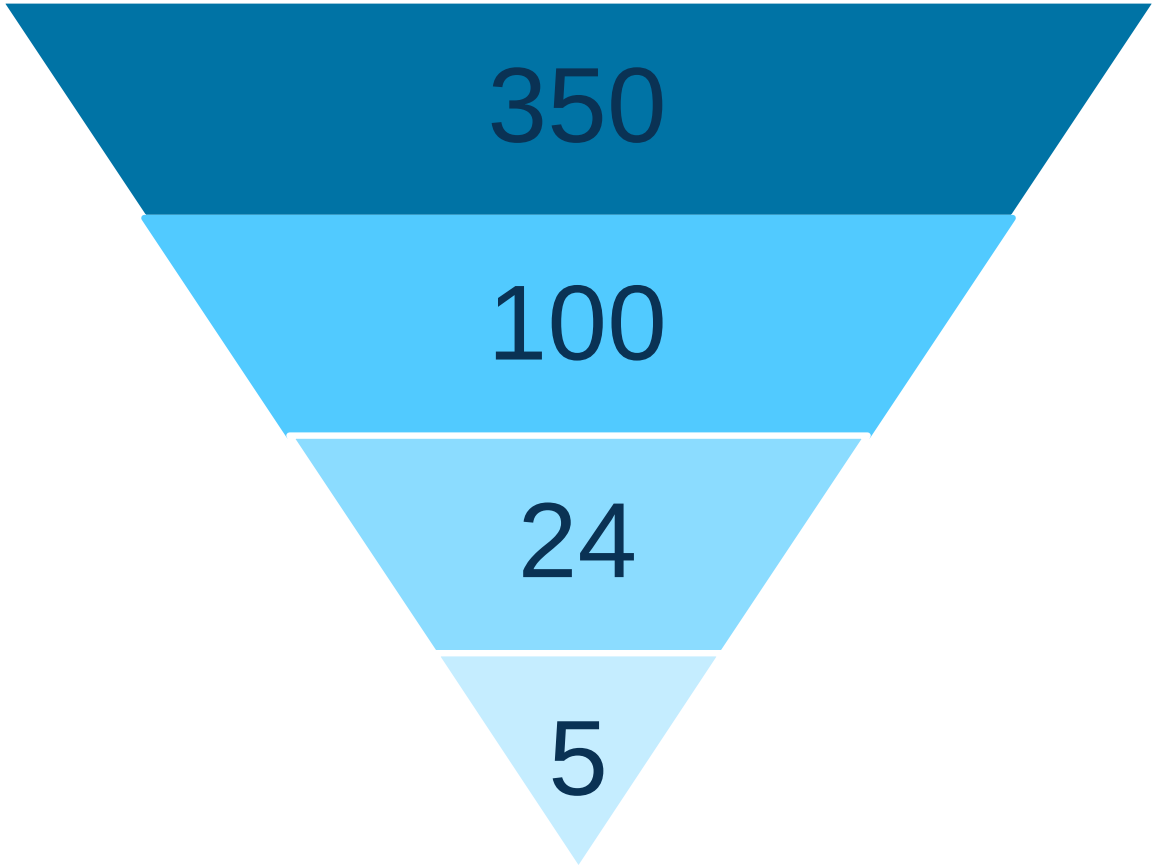


Robust bronchiolar and alveolar uptake of siRNA in lung histological sections by immunohistochemistry

(**A**) PBS-treated animal on Day 10 post dose. (**B**)10 mg/kg *Sod1* siRNA on Day 10 post dose. siRNA is magenta. Blue is hematoxylin counterstain.

Development Candidate Selection

Targeting SARS-CoV-2 genome



ALN-COV (VIR-2703)

350 candidate sequences fully conserved in SARS-CoV and SARS-CoV-2 identified by Alnylam; Alnylam team synthesized and tested in vitro using a reporter system

---

100 top candidates transferred to Vir for testing in the in vitro SARS-CoV-2 infectious system using three concentrations and two different readouts

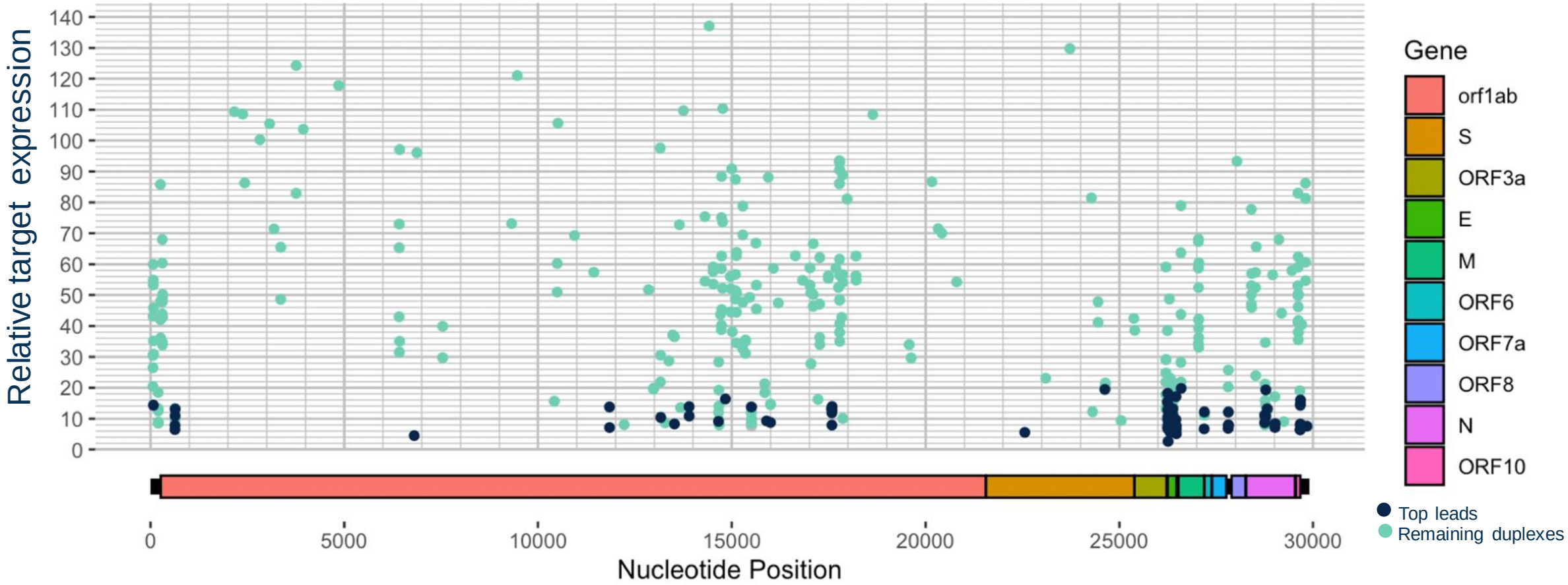
---

EC_{50} evaluation of top 24 sequences against infectious SARS-CoV-2

Narrowed down to 5 sequences based on potency, specificity, sequence conservation, and manufacturability

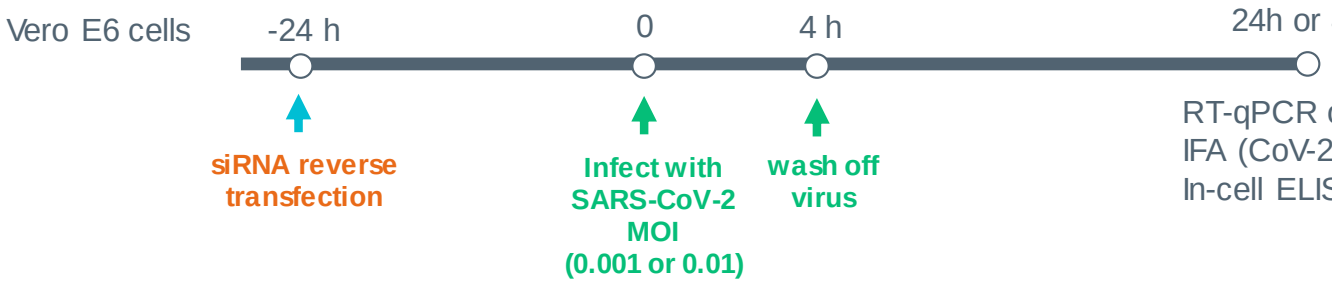
SARS-CoV-2 Genome Coverage and Activity

~100 siRNAs with ≥80% KD at 10 nM in two luciferase reporter vectors tested

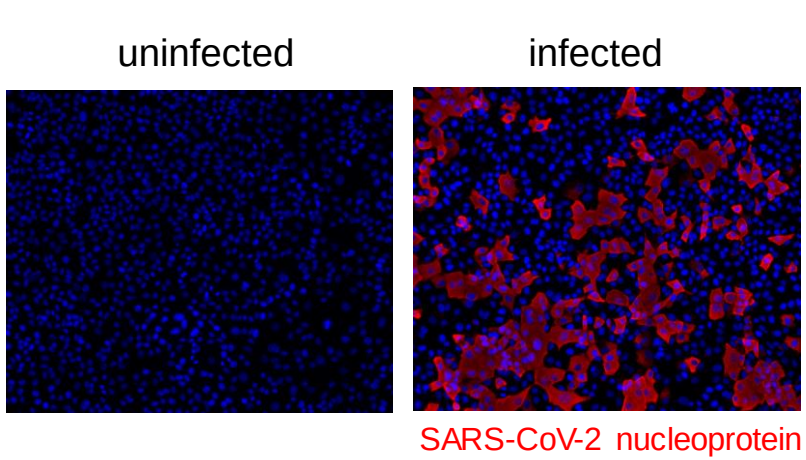


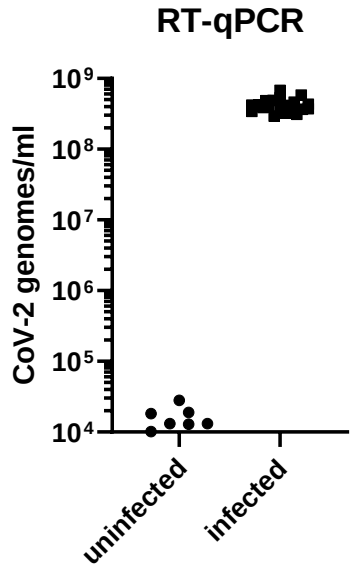
- Graph represents the knockdown (KD) values of each individual duplex.
- Top leads (in black) were selected from duplexes that had ≥80% KD in both luciferase reporter vectors tested

SARS-CoV-2 Live Virus Assay



controls:
- uninfected
- mock transfected + SARS-CoV-2
- siRNA(luciferase) + SARS-CoV-2

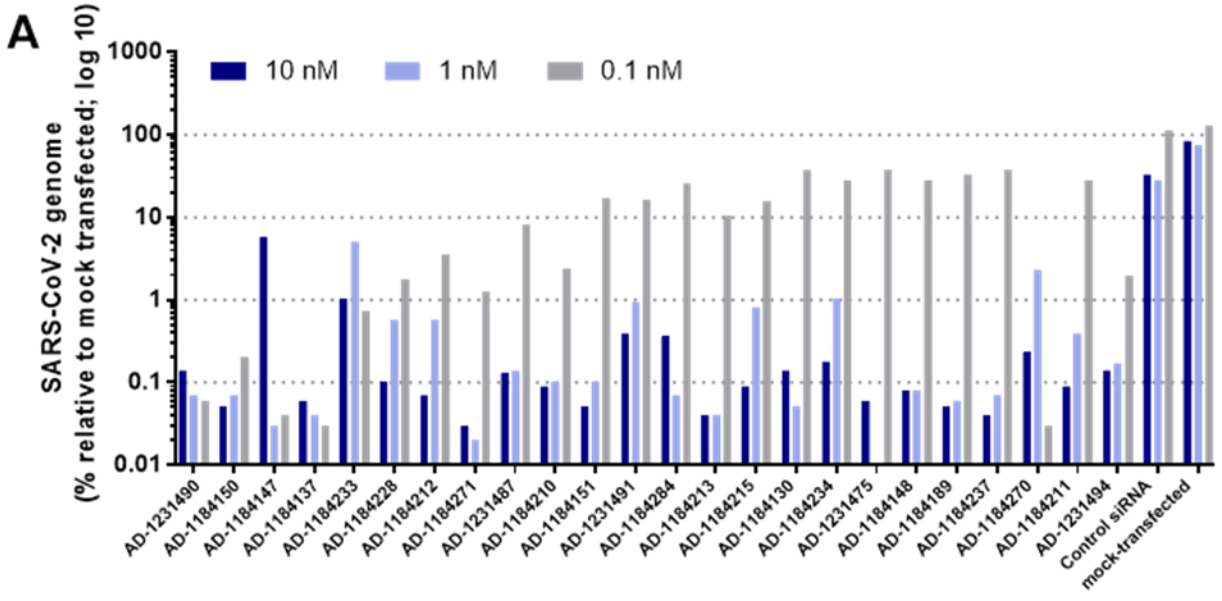




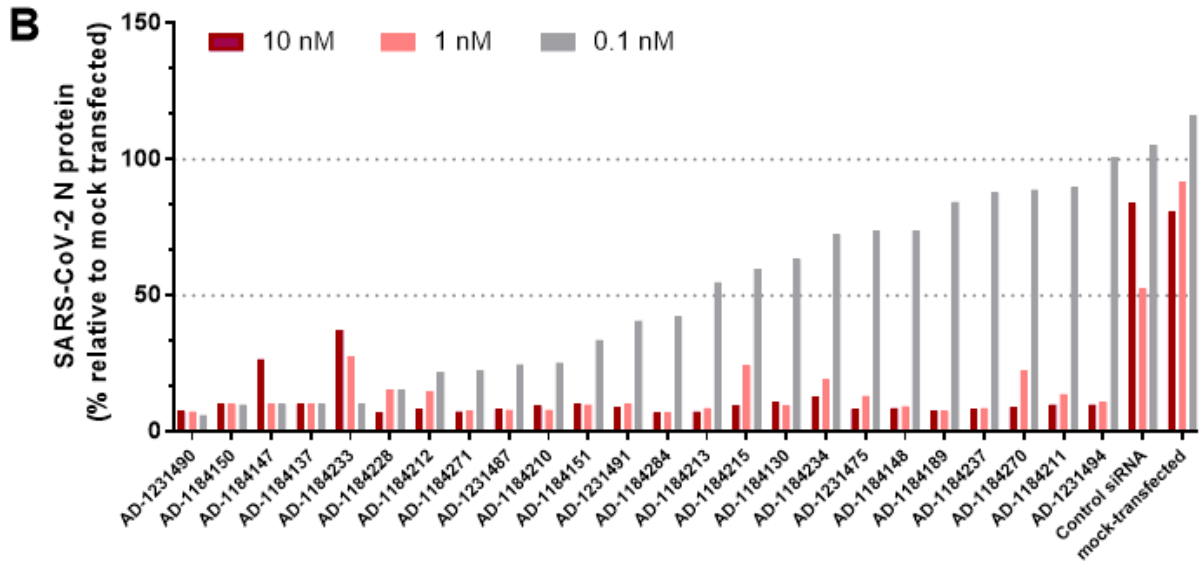
Potent siRNAs Identified Spanning SARS-CoV-2 Genome

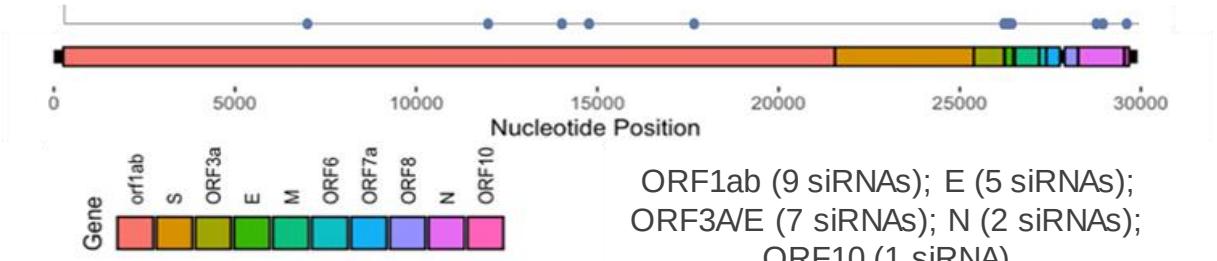
Top 24 siRNAs from ~100 screened in live virus in vitro assay

RT-qPCR



In-cell ELISA





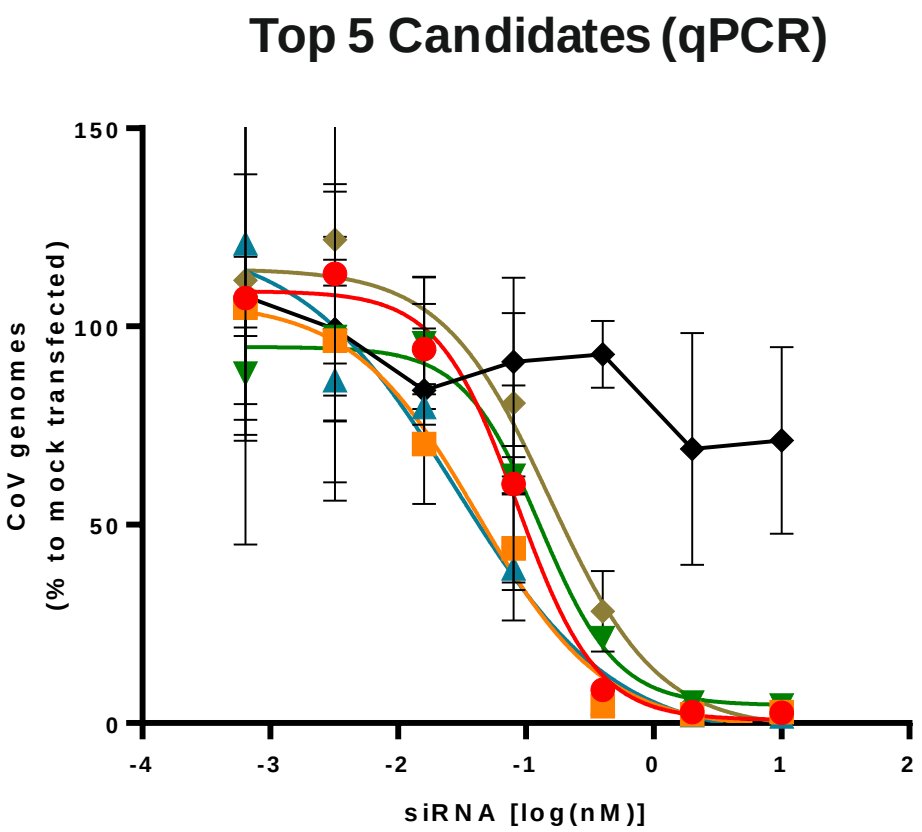
ORF1ab (9 siRNAs); E (5 siRNAs); ORF3A/E (7 siRNAs); N (2 siRNAs); ORF10 (1 siRNA)

- Screening of 100 siRNAs against live SARS-CoV2 infection led to choosing of 24 candidates for further assessment

RT-qPCR: Reverse Transcription Quantitative Polymerase Chain Reaction; ELISA: Enzyme-Linked Immunosorbent Assay; siRNA: small interfering RNA.

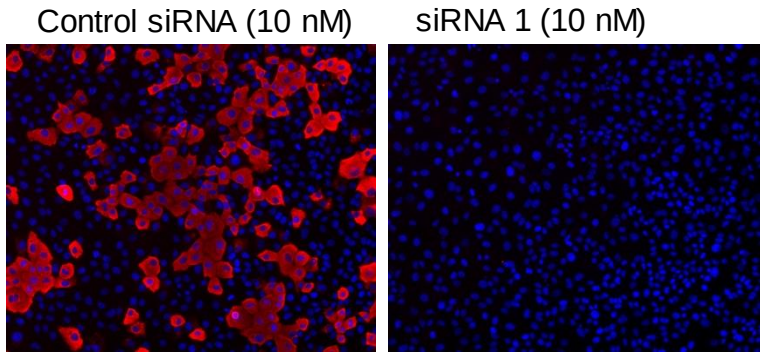
ALN-COV Comprises Two siRNAs from Top 5 Candidates

Two siRNAs utilized to mitigate risk of viral escape



IFA

CoV-2 nucleoprotein (red stain)



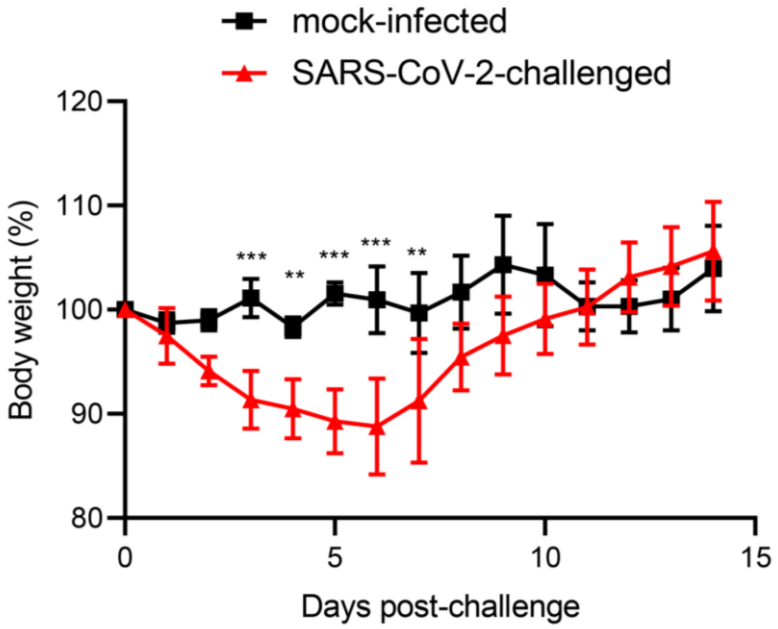
siRNA	EC_{50} EC_{95} (pM; PCR)	EC_{50} EC_{95} (pM; IFA)	Genome Reactivity* (0 mm)	Genome Reactivity* (1 mm)
siRNA 1	42 1183	66 763	99.91%	100.00%
siRNA 2	86 702	118 608	99.89%	99.98%

*N=4386 genomes analyzed

Study Design: Efficacy of ALN-COV in SARS-CoV-2 Hamster Model

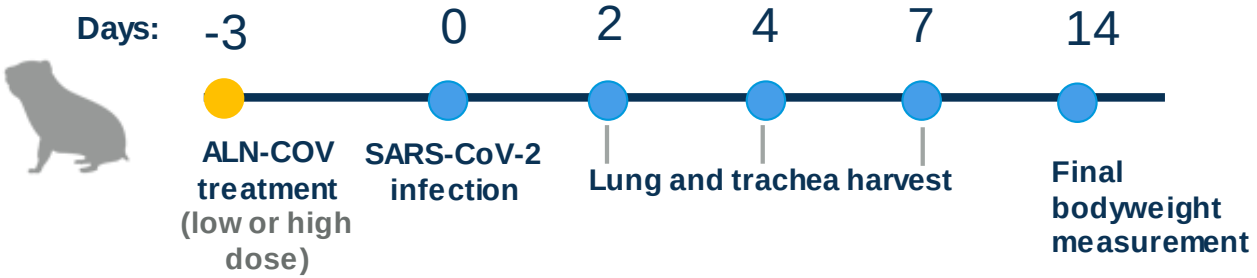
Hamster model supports SARS-CoV-2 infection

- Extensive homology to human *ACE2* receptor; resembles the manifestations of upper and lower respiratory tract infection in humans
- Rapid loss of body weight gain provides key readout in addition viral titers

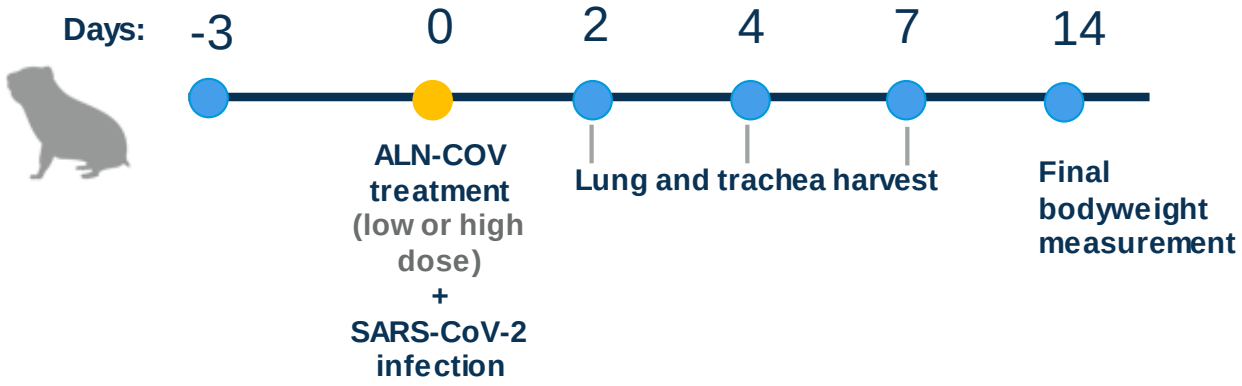


ALN-COV Hamster study Design

Pre-treatment



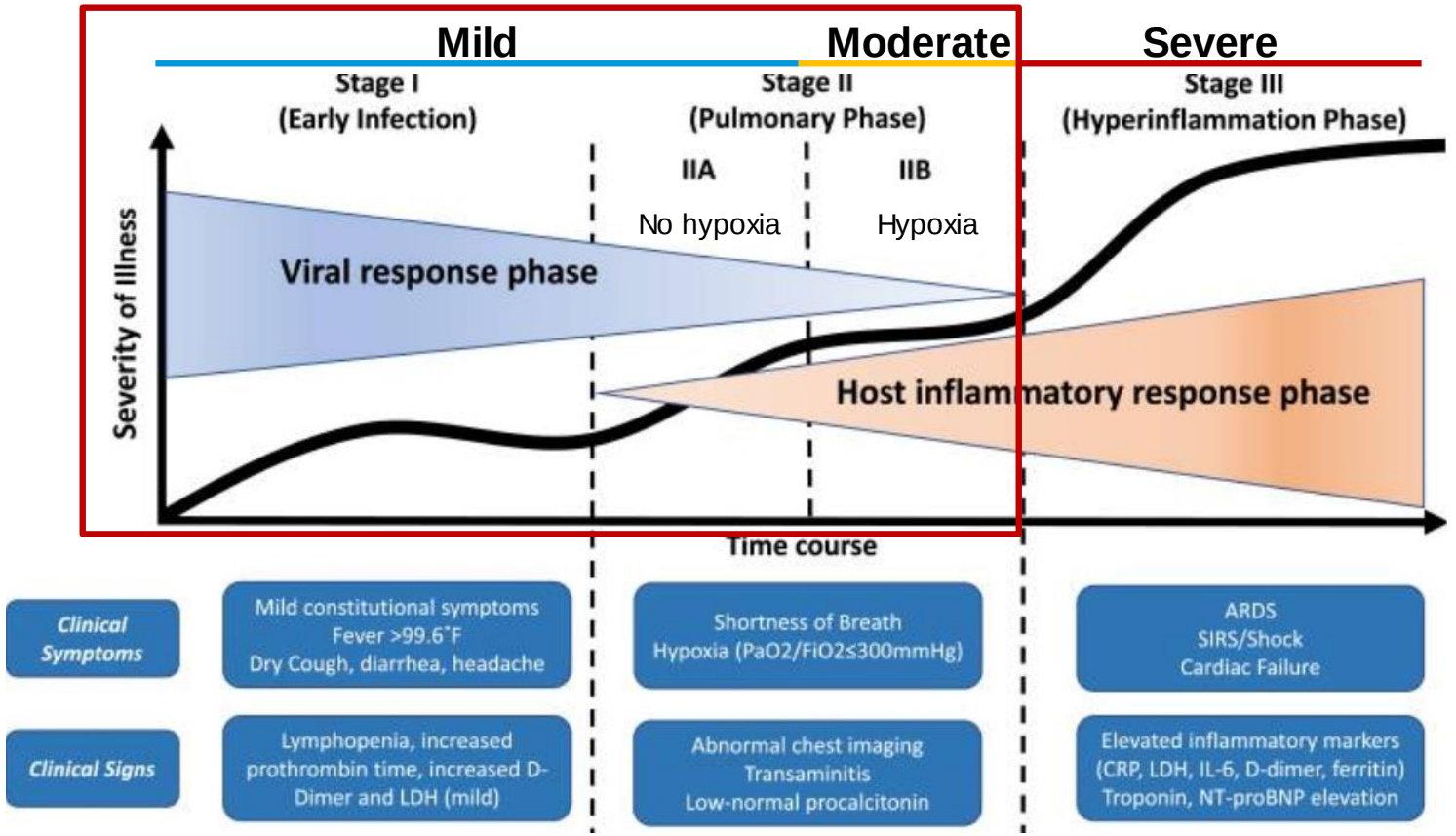
Co-treatment and infection



Clinical Infectious Diseases, , ciaa325, https://doi.org/10.1093/cid/ciaa325

Target Indication for the Treatment of Mild to Moderate COVID-19

Intervention early during viral response phase of disease



Adapted from HK Siddiqi et al., JHLT 2020

Current Clinical Development Plan

Treatment of adult and adolescent patients with mild to moderate COVID-19

Phase 1 Study
Healthy Volunteers
N~40

- **Objective**: Establish safety and select dose of ALN-COV
- **Study Design:** Randomized, single-blind, placebo-controlled, ascending dose study in healthy volunteers (HVs)
- **Dose Regimen:** Once daily x 2 days inhalation administration via nebulizer
- **Randomization: 3:1**, ALN-COV : placebo
- **Endpoints**: Safety, tolerability, and pharmacokinetics (PK)

Phase 2/3 Study
Mild to Moderate COVID-19
N~600

- **Objective**: Evaluate efficacy and safety of ALN-COV
- **Study Design:** Randomized, double-blind, placebo-controlled study in patients with mild to moderate COVID-19 with risk factors for severe disease
- **Dose Regimen:** Once daily x 2 days inhalation administration via nebulizer
- **Randomization:** 1:1, ALN-COV+SOC : placebo+SOC
- **Endpoints**: Progression to severe pulmonary disease, hospitalization, mechanical ventilation, symptom resolution, disease severity, mortality, viral parameters, safety, and PK

Alnylam PHARMACEUTICALS