Supplementary material

GNS561 exhibits potent antiviral activity against SARS-CoV-2 through autophagy inhibition

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Tables

Table S1. Primer and probe sequences for SARS-CoV-2 E (cells) and N (mouse) genes q-RTPCR

investigation

SARS-CoV-2 Primers/Probes		Sequences
Cell assays	Forward	5'-GAC CCC AAA ATC AGC GAA AT-3'
	Reverse	5'-TCT GGT TAC TGC CAG TTG AAT CTG-3'
	Probe	5'-FAM-ACC CCG CAT TAC GTT TGG TGG ACC-BHQ1-3'
Mouse assays	Forward	5'-TAA TGG ACC CCA AAA TCA GC-3'
	Reverse	5'-GAA TCT GAG GGT CCA CCA AA-3'

Figures

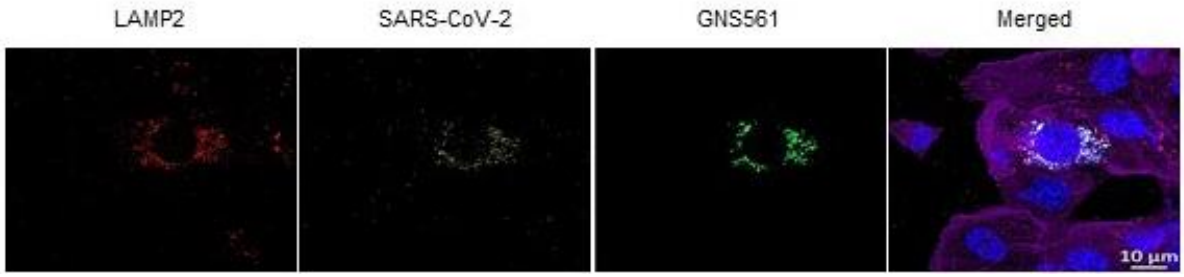

Figure S1. SARS-CoV-2 and GNS561 localization inside lysosomes. After 2 hours of treatment with 4 µM GNS561, Vero E6 cells were infected with SARS-CoV-2 IHUMI-6 strain for an additional 48 hours. Representative confocal images showing the localization of SARS-CoV-2 (yellow) inside LAMP2-positive lysosomes (red) together with GNS561 (green) inside Vero E6 cells are exposed.

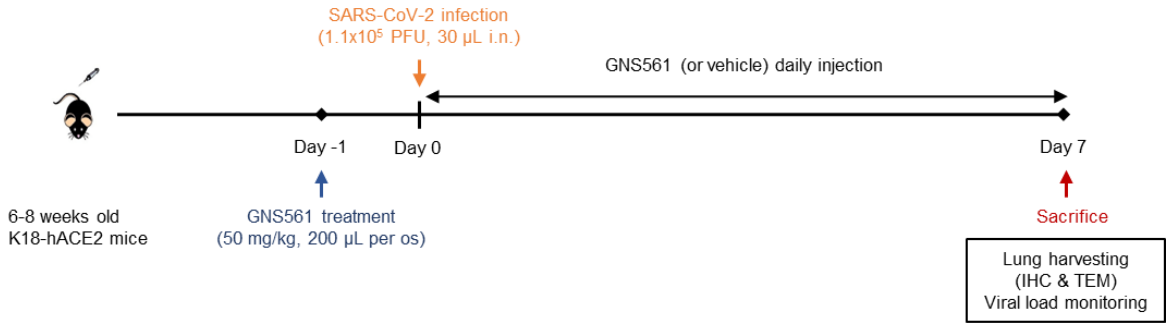

Figure S2. Study design of K18-hACE C57BL/6J mice model. Eight to nine-week-old K18-hACE C57BL/6J mice were treated with GNS561 (50 mg / kg, 200 µL) or with vehicle per os 24h before intranasally SARS-CoV-2 infection (1.1x105 PFU, 30 µL). Mice were then daily injected with GNS561 or vehicle until sacrifice 7 days post-infection. IHC: immunohistochemistry, i.n.: intranasally, TEM: transmitted electron microcopy.