1 **Identification of potent and safe antiviral therapeutic candidates against**

2 **SARS-CoV-2**

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47 **Abstract**

48 COVID-19 pandemic has infected millions of people with mortality exceeding 300,000. There is

49 an urgent need to find therapeutic agents that can help clear the virus to prevent the severe

50 disease and death. Identifying effective and safer drugs can provide with more options to treat

51 the COVID-19 infections either alone or in combination. Here we performed a high throughput

52 screen of approximately 1700 US FDA approved compounds to identify novel therapeutic agents

53 that can effectively inhibit replication of coronaviruses including SARS-CoV-2. Our two-step

54 screen first used a human coronavirus strain OC43 to identify compounds with anti-coronaviral

55 activities. The effective compounds were then screened for their effectiveness in inhibiting

56 SARS-CoV-2. These screens have identified 24 anti-SARS-CoV-2 drugs including previously

57 reported compounds such as hydroxychloroquine, amlodipine, arbidol hydrochloride, tilorone

58 2HCl, dronedarone hydrochloride, and merfloquine hydrochloride. Five of the newly identified

59 drugs had a safety index (cytotoxic/effective concentration) of >600, indicating wide therapeutic

60 window compared to hydroxychloroquine which had safety index of 22 in similar experiments.

61 Mechanistically, five of the effective compounds were found to block SARS-CoV-2 S protein-

62 mediated cell fusion. These FDA approved compounds can provide much needed therapeutic

63 options that we urgently need in the midst of the pandemic.

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70 **Introduction**

71 Novel coronavirus mediated disease (COVID-19) emerged as a major pandemic and has spread

72 across the world in such a short span of time since Dec 2019. As of May 15, 2020, more than 4.5

73 million confirmed infections have been reported with approximately 308,000 deaths (WHO,

74 situation report 116). These numbers may be a vast underestimation as many of the infected

75 patients may remain asymptomatic and can only be detected by antibody testing[1]. Similarly,

76 many of the deaths may not be accounted due to lack of testing. The disease is caused by a novel

77 coronavirus termed SARS-CoV-2 which belongs to the *Coronaviridae* family and is the third

78 major coronavirus pandemic in last 20 years after Severe Acute Respiratory Syndrome (SARS)

79 and Middle East Respiratory Syndrome (MERS)[2-7]. The lack of available therapeutic options is a

80 major limiting factor in treating these infections which leads to excessive mortality.

81 Currently, there is an urgent and unmet need of effective antiviral therapy that can not only

82 decrease the disease burden in the patient but can also decrease the ability of the person to infect

83 others. Developing a novel drug many take years to confirm the safety and efficacy, which may

84 not be practical to deal with a widespread pandemic that is killing thousands of people every day.

85 Alternatively, it sounds a lucrative option to repurpose US Food and Drug Administration (FDA)

86 approved drugs for their efficacy against SARS-CoV-2. Earlier screens have found antiviral

87 efficacy of approved therapies such as hydroxychloroquine[8-12], however, these therapies failed to

88 provide any beneficial effects in COVID-19 due to their toxic side effects[13-18]. Finding efficacy

89 of an approved drug against SARS-CoV-2 with minimal toxicity can provide much needed

90 therapeutic option to treat COVID-19.

91 Here we screened approximately 1700 US FDA approved compounds to test their ability to

92 inhibit SARS-CoV-2 replication. Our data report finds 24 compounds that are highly effective in

93 inhibiting SARS-CoV-2 replication at concentrations that were significantly lower than those

94 having cytotoxic effects. We also investigated possible mechanism of these compounds.

95 **Results**

96 **Inhibitory potential of FDA approved drugs against human coronavirus OC43**. Initial

97 screening was performed using HCoV-OC43 due to its low biosafety concerns. OC43 is a human

98 coronavirus that usually causes mild disease in humans and cattle[19]. The experimental protocol is

99 demonstrated in Fig. 1A using LLC-MK2 cells which were infected with OC43 at a MOI of 1 for

100 48 h in presence of the US FDA licensed compounds. The inhibitory potentials of these

101 compounds were measured with the treatment of compounds at 10 μM for 48 hours. The viral

102 presence was detected by immunostaining for the virus and DAPI staining for the cell nuclei.

103 The inhibitory capacity was measured by using the ratio of viral fluorescence to the DAPI and is

104 depicted in Fig. 1B. The initial screen obtained 231 compounds that had ability to inhibit OC43

105 replication >95%. The remdesivir was used to as a positive control (Fig. 1 C and D).

106 **Calculation of IC50, CC50 and SI of FDA approved drugs against OC43.** Next, we sought to

107 determine the effective concentrations of positively screened drugs in our initial approach and

108 test the toxicity profile of these drugs in relation to their viral inhibitory concentrations. Our data

109 show that vast majority of the positively screened drugs were effective in inhibiting the viral

110 replication at sub-micromolar concentrations including many of them can almost completely

111 inhibit the viral replication at micromolar range (Fig. 2). Surprisingly, the effective drugs against

112 the coronavirus belonged to a wide range of therapeutic groups including those used for

113 neurological diseases, hormones, enzymes and antimicrobial agents among others (Table 1). The

114 cytotoxic concentration to kill 50% cells (CC50) was noted for these drugs by measuring cell

115 viability over similar concentrations. The selective index in our study was found to be >600 for 5

116 of the screened compounds. The SI for the hydroxychloroquine was 22 in our study, indicating

117 increased safety of newly identified drugs compared to the hydroxychloroquine.

118 **Testing the antiviral effectiveness of positive drugs against SARS-CoV-2.** Finally, we sought

119 to determine whether these compounds can show efficacy against SARS-CoV-2, the causative

120 agent of COVID-19. Positive compounds from the initial screen were tested for their antiviral

121 efficacy against SARS-CoV-2 in Vero cells. SARS-CoV-2 replicates within the Vero cells and

122 causes cytopathic effects in these cells in absence of any antiviral treatment. We generated the

123 dose response inhibition curves along with the cytotoxicity curves for these compounds in

124 presence of SARS-CoV-2 (Fig. 3). Our data show that 24 of these compounds show significant

125 efficacy in inhibiting SARS-CoV-2 replication with sub micromolar IC50 for many of these

126 drugs such as nilotinib, clofazimine and raloxifene. The effects also confirmed by

127 immunofluorescence assay (data not shown). These compounds also belong to a wide variety of

128 classes including cardiac glycosides, anti-malarial drug hydroxychloroquine, cyclooxygenase-2

129 inhibitors and ion channel blockers, among others. The IC50, CC50 and SI of these compounds

130 is shown in Table 2.

131 **Five candidate drugs inhibit cell fusion.** Finally, in order to clarify the mechanism by which

132 the compounds inhibit SARS-CoV-2, we investigated the effects of these candidates on virus

133 entry. First, we constructed the cell-cell fusion assays. As indicated in Fig 4, SARS-CoV-2 S

134 protein expression can initiate cell fusion with ACE2-overexpressed cells, but the control vector

135 did not. Then, we detected the effects of these indicated drugs on S protein-mediated cell fusion.

136 Indicated drugs were added to cells before co-incubation of the cells. We found that digoxin and

137 ouabain octahydrate induced cell death, but not other indicated drugs. Interestingly, fendiline

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138   hydrochloride, monensin sodium salt, vortioxetine, sertraline hydrochloride, and raloxifene

139   hydrochloride inhibited the SARS-CoV-2 S protein-mediated cell fusion (Fig. 4).

140   **Discussion**

141   The current pandemic of COVID-19 is the third major outbreak in this century and the largest

142   outbreak of the coronavirus in known human history. The three novel coronavirus outbreaks in

143   such a short span of time are strong indicators of potential threat posed by coronaviruses. While

144   majority of the respiratory viral infection research has been focused on influenzas viruses which

145   causes huge burden of seasonal flu and occasionally pandemic outbreaks, coronavirus is likely to

146   emerge a similar or more severe pathogen than flu in long term.

147   Given the scale and devastation of the current COVID-19 outbreak, and persistent threat of

148   coronaviruses in causing human disease, there is urgent need to find effective and safe therapies

149   that can treat these patients. Currently, there are no approved therapies for coronaviruses

150   including SARS-CoV-2. The experimental therapies being used with known antiviral agents

151   either show limited efficacy (remdesivir) or have high systemic toxicity (hydroxychroquine),

152   limiting their usefulness[20-22]. Finding new therapies that are effective and safe are urgently

153   needed. In this study, we have identified many FDA approved therapies that are highly effective

154   against coronaviruses, including 24 of the agents that are effective against SARS-CoV-2. This

155   screening confirms previous reports demonstrating anti-SARS-CoV-2 activity of

156   hydroxychroquine, amlodipine besylate, arbidol hydrochloride, tilorone 2HCl, dronedarone

157   hydrochloride, tetrandrine, merfloquine, and thioridazine hydrochloride[8-10, 23], while identifying

158   additional 14 drugs. The underlying mechanisms of viral replication inhibition by these drugs is

159   not clear. It is highly unlikely that these compounds will have similar antiviral mechanisms given

160   the vast structural and pharmacological diversities of the effective antiviral compounds in our

161   study. However, it is clear from other viral studies such as influenza or HIV, where antiviral

162   drugs can affect various steps in the viral life cycle including attachment, entry, replication,

163   assembly and budding of viral progeny. Five drugs may inhibit S-mediated cell fusion as

164   indicated by our data (Fig. 4). Further studies are required to understand the precise mechanisms

165   of each of the effective compounds found in this study.

166   Toxicity is one of the limiting factors in the therapeutic application of many drugs despite their

167   known antiviral activities. Many of these drugs had SI of > 600, showing promise of their

168   usefulness at safe doses. For comparison, the SI of hydroxychloroquine was found to be 22 in

169   our study while SI of amlodipine besylate was found to be ~3, demonstrating much lower safety

170   profile of this drug. Similarly, other drugs that are known to have low selective index such as

171   digoxin for their approved use, also show lower SI in our screen. Five of the drugs with SI

172   of >600 include tyrosine kinase inhibitor nilotinib, antibiotics such as clofazimine and actidione,

173   selective estrogen receptor modulator such as raloxifene and non-steroidal anti-inflammatory

174   drug celecoxib.

175   etacoronaviruses have raised great public health threats to human beings, as most known HCoVs

176   including all the three virulent HCoVs (SARS-CoV, MERS-CoV and SARS-CoV-2) and two

177   seasonal HCoVs (OC43 and HKU1) belong to this species[3-7, 19, 24]. It is of great value to identify

178   antivirals against a broad spectrum of HCoVs, particularly the Betacoronaviruses, to tackle such

179   threats by pharmaceutical interventions. To this end, we first screened the compounds which

180   showed apparent activity of anti-OC43, the most prevalent HCoV circulates worldwide[25]. We

181   then narrowed down the candidates by the screening on SARS-CoV-2, resulting in the

182   identification of 24 compounds which can inhibit both OC43 and SARS-CoV-2. Our study

183   provides a foundation for subsequent anti-HCoVs drug screening of broad spectrum. However,

184   further tests are warranted to verify their efficacies.

185   In summary, our screen identified 14 previously unknown FDA approved compounds that are

186   effective in inhibiting SARS-CoV-2 beside confirming the antiviral properties of ten previously

187   reported compounds, validating our approach. This screen identified five new compounds that

188   are highly efficacious in inhibiting the viral replication of SARS-CoV-2 with SI >600. Further

189   studies are needed to confirm the *in vivo* efficacy of these drugs in humans and COVID-19

190   relevant mouse models such as those with human ACE2 transgene[26].

191   **Materials and Methods**

192   **Cells and Viruses:** LLC-MK2 cells (Rhesus monkey kidney cells) were cultured in 64% Hank's

193   MEM and 32% Earle's MEM (Gibco, New York, USA) supplemented with 3% fetal bovine

194   serum (FBS) (Hyclone, Utah, USA) and 1% glutamine (Thermo, Massachusetts, USA). Vero

195   cells (African green monkey kidney cell) were cultured in Dulbecco's Modified Eagle's Medium

196   (DMEM, Gibco) supplemented with 10% FBS.

197   Human coronavirus (HCoV) strain OC43 was propagated in LLC-MK2 cells in 0.5% FBS MEM

198   and virus titers were determined via TCID50 with LLC-MK2 cells. SARS-CoV-2 virus was

199   isolated from the respiratory samples of patients in Wuhan of Hubei Province *(3)*. SARS-CoV-2

200   virus was propagated in Vero cells and used in this study. All experiments with SARS-CoV-2

201   virus were conducted in BSL-3 laboratory.

202   HEK293T cells stably expressing recombinant human ACE2 (293T/hACE2) were maintained in

203   Dulbecco's MEM containing 10% fetal bovine serum and 100 unit penicillin, and 100μg

204   streptomycin per milliliter.

205   **Antibodies:** Mouse polyclonal against OC43 N antibody was prepared in the laboratory. Rabbit

206   polyclonal against SARS-CoV-2 N protein antibody was purchased from Sino Biological

207   (Beijing, China). Alexa Fluor 488-conjugated goat anti-mouse IgG, Alexa Fluor 488-conjugated

208   goat anti-rabbit IgG were purchased from Thermo.

209   **Screening of FDA-approved drugs:** US FDA-approved drug library which contain 1700

210   compounds was purchased from TargetMol (Massachusetts, USA). LLC-MK2 cells were seeded

211   at 2×10^4 cells per well in 96-well plates and incubated at 37°C and 5% CO_2. The next day, LLC-

212   MK2 cells were treated with the compounds at a concentration of 10μM. After 1 h of treatment,

213   cells were infected with OC43 at MOI of 1. At 48 h post infection (hpi), cells were fixed with 4%

214   paraformaldehyde for 20 min at room temperature. Immunofluorescence staining was performed

215   using mouse anti-OC43 NP antibody, followed by anti-mouse Alexa Flour 488 and DAPI (Sigma,

216   St. Louis, MO). Images were captured by Operetta (PerkinElmer, Massachusetts, USA) at the

217   magnification of $20 \times$ objective. The infection ratios were calculated using automated image

218   analysis software (Harmony 3.5.2, PerkinElmer). Remdesivir and DMSO were used as positive

219   and negative controls, respectively.

220   The positively identified drugs from this screen were used to perform dose response curves

221   against OC43 on LLC-MK2 and against SARS-CoV-2 using Vero cells as described below.

222   **IC_{50} (The half maximal Inhibitory concentration), CC_{50} (The half maximal cytotoxic**

223   **concentration) and SI (Selectivity index) determination:** LLC-MK2 cells (for OC43 infection)

224   or Vero cells (for SARS-CoV-2 infection) were seeded in 96 wells plate one day before infection

225   at the concentration of 2×10^4 cells /well or 1.4×10^4 cells /well, respectively. For IC50, cells were

226   pre-treated for 1h with each drug at concentrations 0.013, 0.041, 0.123, 0.370, 1.111, 3.333, 10,

227   30 μM, and then infected with virus at MOI of 1. At 48 hpi (OC43) or 24 hpi (SARS-CoV-2),

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228 cells were fixed with 4% paraformaldehyde for 20 min at room temperature.

229 Immunofluorescence was conducted with mouse anti-OC43 N protein antibody, or rabbit anti-

230 SARS-CoV-2-NP antibody, and followed by anti-mouse, or anti-rabbit Alexa Flour 488 and

231 DAPI. Images were performed by Operetta with $20\times$ objective. The IC_{50} were calculated using

232 automated image analysis software (Harmony 3.5.2, PerkinElmer).

233 For CC_{50}, cells were pre-treated with each drug at concentrations 0.013, 0.041, 0.123, 0.370,

234 1.111, 3.333, 10, 30 μM, respectively. After 48h (LLC-MK2 cells) or 24h (Vero cells) post

235 treatment, cell viability was evaluated by using a CCK8 kit (Yeasen, Beijing, China) according

236 to the manufacturer's instructions. Selectivity index was calculated using following formula:

237 $SI=CC_{50}/IC_{50}.$ Graphpad Prism 7.0 was used for analyzing IC_{50} and CC_{50}.

238 Immunofluorescence: Cells were fixed with 4% paraformaldehyde for 20 min at room

239 temperature, and permeabilized with 0.5% Triton X-100 for 10 min. Cells were then blocked

240 with 5% BSA and stained with primary antibodies, followed by staining with an Alexa Fluor 488

241 secondary antibodies. Nuclei were counterstained with DAPI.

242 **Quantitative RT-PCR:** Vero cells were pre-treated with indicated concentrations of drugs for

243 1h and incubated with SARS-CoV-2 at 0.1 MOI for 1h. Then, cells were washed with opti-MEM

244 for one time and incubated with indicated concentrations of drugs. At 24 hpi, supernatants were

245 collected and viral RNA in the cell supernatants were extracted by using Direct-zol RNA

246 MiniPrep kit (Zymo research, CA, USA) according to the manufacturer's instructions. Viral

247 copy numbers were measured by RT-PCR using primers and probe targeting the SARS-CoV-2 N

248 gene. The reference standard was tenfold diluted from 1×10^{9} copies to 1×10^{4} copies. PCR

249 amplification procedure was 50℃, 15min, 95℃, 3min; 95℃, 15s, 60℃, 45s+Plate Read, 50

250 cycles. The amplification process, fluorescence signal detection, data storage and analysis were

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251   all completed by fluorescence quantitative PCR and its own software (Bio-Rad CFX Manager).

252   The copies of virus were calculated according to the standard curve. The inhibition ratio was

253   obtained by dividing the number of copies of the virus in the vehicle control group. The data

254   were nonlinear fitting by graphpad 7.0 software to calculate IC_{50} of each drug.

255   **Cell-cell fusion assay:** Cell-cell fusion assays were performed as described previously *(19)*.

256   Briefly, HEK-293T cells were co-transfected with SARS-CoV-2-S glycoprotein and eGFP. At

257   24 h post transfection, cells were digested with trypsin (0.25%) and overlaid on a 50% confluent

258   monolayer of 293T-ACE2 cells at a ratio of 1:1 which were treated with candidate drugs for 1h.

259   After 24h incubation, images of syncytia were captured with Operetta (PerkinElmer,

260   Massachusetts, USA).

261   **Acknowledgments**

268   **Author contributions**

269   Project conception: J.W., X.L., and L.S.; Experimental design: J.W. X.L., D.C., L.S., Z.Z., and

270   L.R.; Experimental work: X.X., C.W., Y.W., X.D., T.J., X.X., C.W., Z.T., Y.W.; Data analysis:

271   J.W., X.L., L.R., C.S.D.C., X.X., and Z.Z., Writing Original Draft: J.W., X.L., L.S., D.C., and

272   X.X.; Writing Review & Editing, J.W., X.L., D.C., L.S. and C.S.D.C.; All authors reviewed the

273   manuscript.

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354   **Figure legend**

355   **Figure 1. High-throughput screening of US FDA-approved drug library to inhibit human**

356   **coronavirus OC43 replication *in vitro*.** A. The strategy of high throughput screening to identify

357   antiviral drugs that effectively inhibit OC43 replication. LLC-MK2 cells were pretreated with

358   FDA-approved drugs at 10μM for 1h and infected with 1 MOI of OC43 for 48h. Cells were then

359   fixed and stained to calculate the infection ratio with Operetta software. B. Primary screening

360   results of 1700 FDA-approved drugs against OC43, each dot represents one compound along

361   with the rate of OC43 inhibition. C. Image samples show signals of OC43 infection in cell

362   cultures. LLC-MK2 cells were treated with indicated doses of remdesivir for 1h, and then cells

363   were infected with OC43 for 48 h. D. Cell viability of remdesivir to LLC-MK2 cells were

364   measured by CCK-8 assays. The % inhibitions were calculated according to the data in C.

365   **Figure 2. Dose response curves of selected compounds from the hits against OC43 infection**

366   ***in vitro*.** LLC-MK2 cells were pretreated with indicated drugs at 37°C for 1h with eight doses

367   (0.014, 0.041, 0.123, 0.370, 1.111, 3.333, 10, 30μM) with three-fold dilution followed by

368   infection with OC43 at MOI of 1 for 48h. In parallel, effects of these compounds on the cell

369   viability in LLC-MK2 cells were measured by CCK-8 assays. The left *Y*-axis of the graphs

370   represent % inhibition of the infection and the right *Y*-axis of the graphs present % cell viability

371   in presence of the drugs.

372   **Figure 3. Dose response curves of selected compounds against SARS-CoV-2 infection.** Vero

373   cells were pretreated with indicated drugs at 37°C for 1h with eight doses (0.014, 0.041, 0.123,

374   0.370, 1.111, 3.333, 10, 30μM) with three-fold dilutions, followed by infection with SARS-CoV-

375   2 at a MOI of 0.1 for 24h. The viral load in the cell supernatant was quantified by qRT-PCR.

376   Meanwhile, cell viability in presence of these drugs were measured in Vero cells by CCK-8

377   assays. The left *Y*-axis of the graphs represent % inhibition of viral replication and the right *Y*-

378   axis of the graphs indicates % cell viability in presence of the drugs.

379   **Figure 4. The effect of indicated drugs on cell-cell fusion mediated by SARS-CoV-2 S**

380   **protein.** HEK-293T cells were co-transfected with SARS-CoV-2-S glycoprotein and eGFP. 24 h

381   post transfection, cells were digested with trypsin (0.25%) and overlaid on a 50% confluent

382   monolayer of 293T-ACE2 cells at a ratio of 1:1 which were treated with candidate drugs for 1h.

383   After 24 h incubation, images of syncytia were captured with Operetta and analyzed by Harmony

384   software.
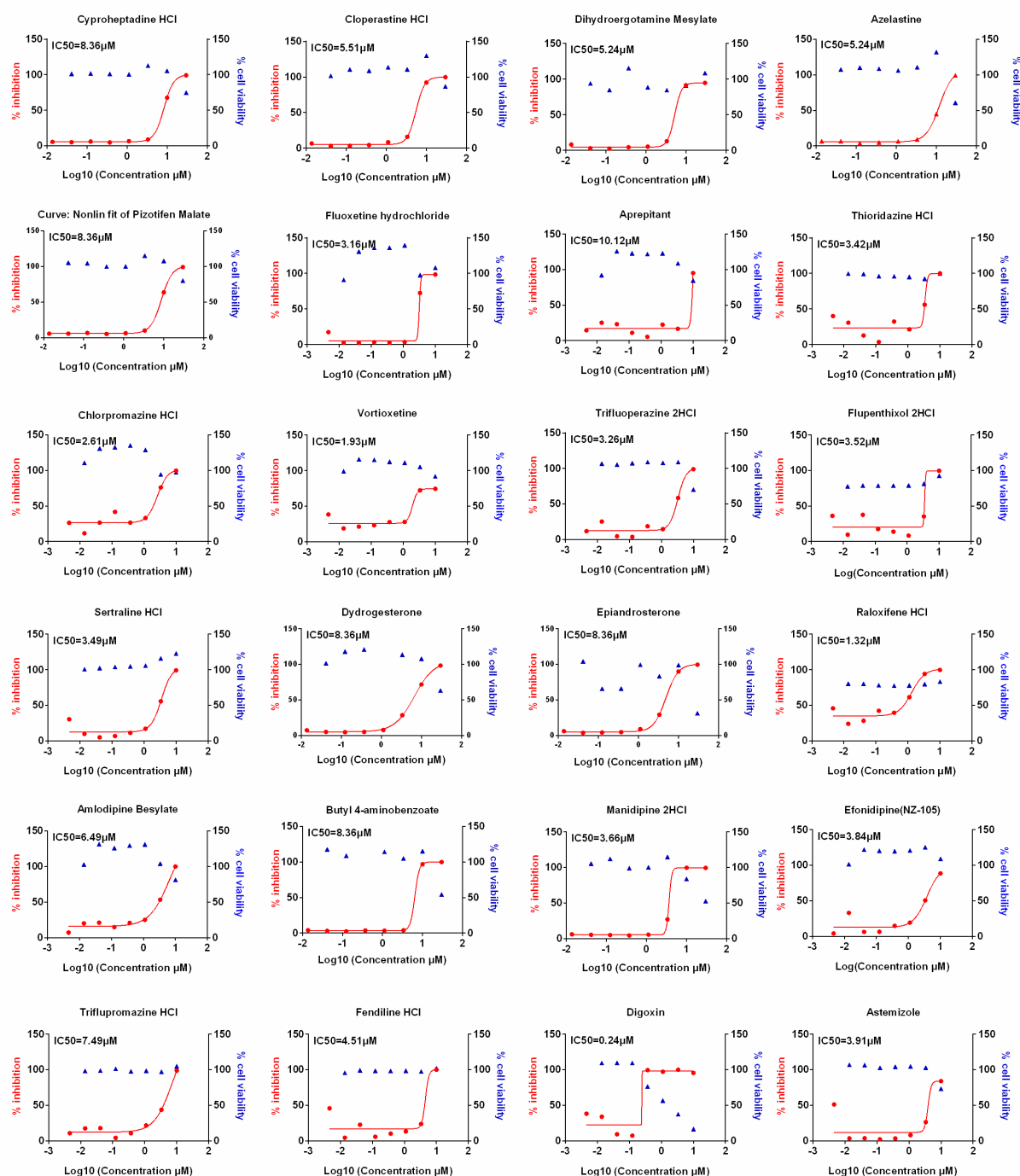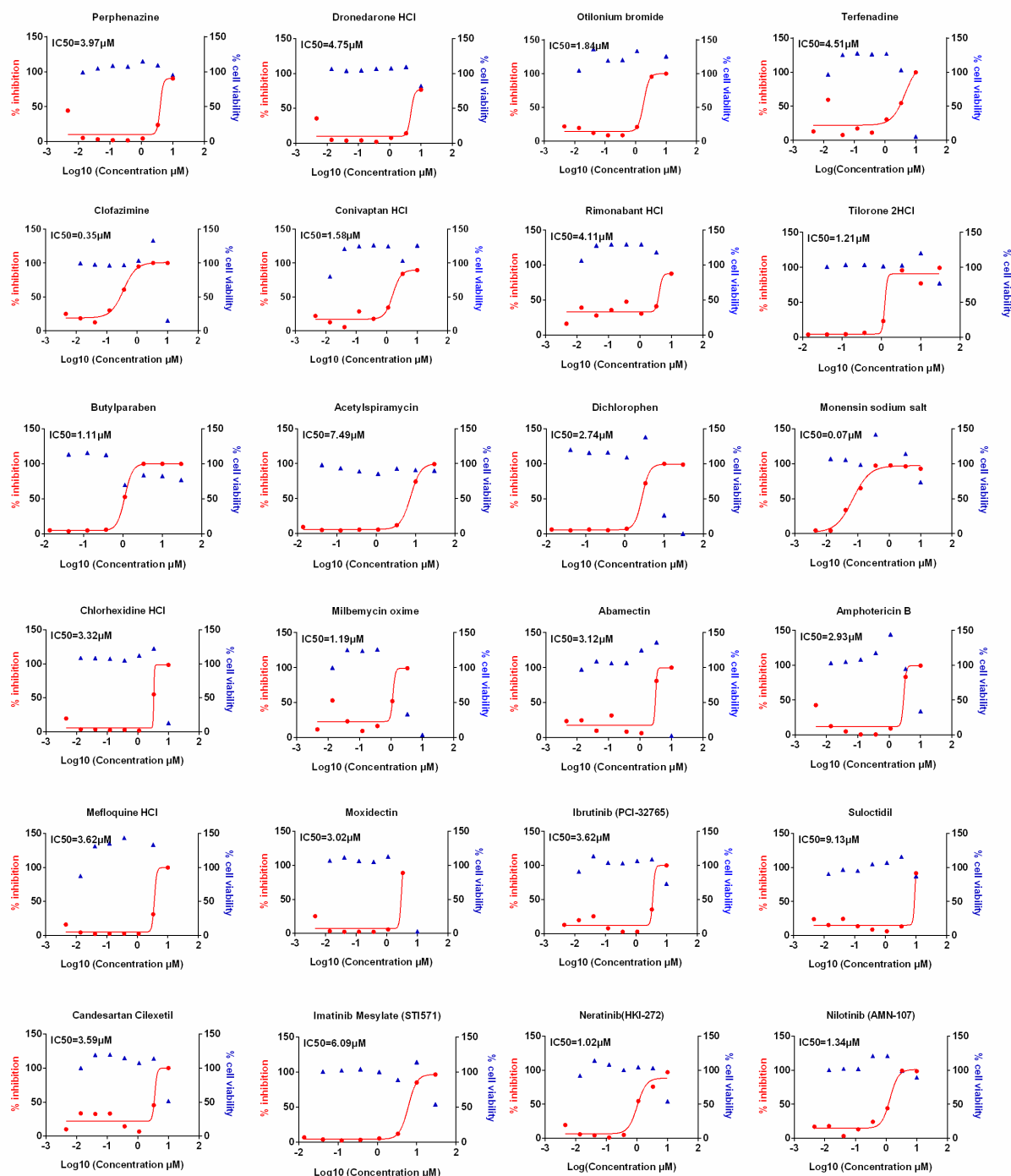
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387   Figure.1
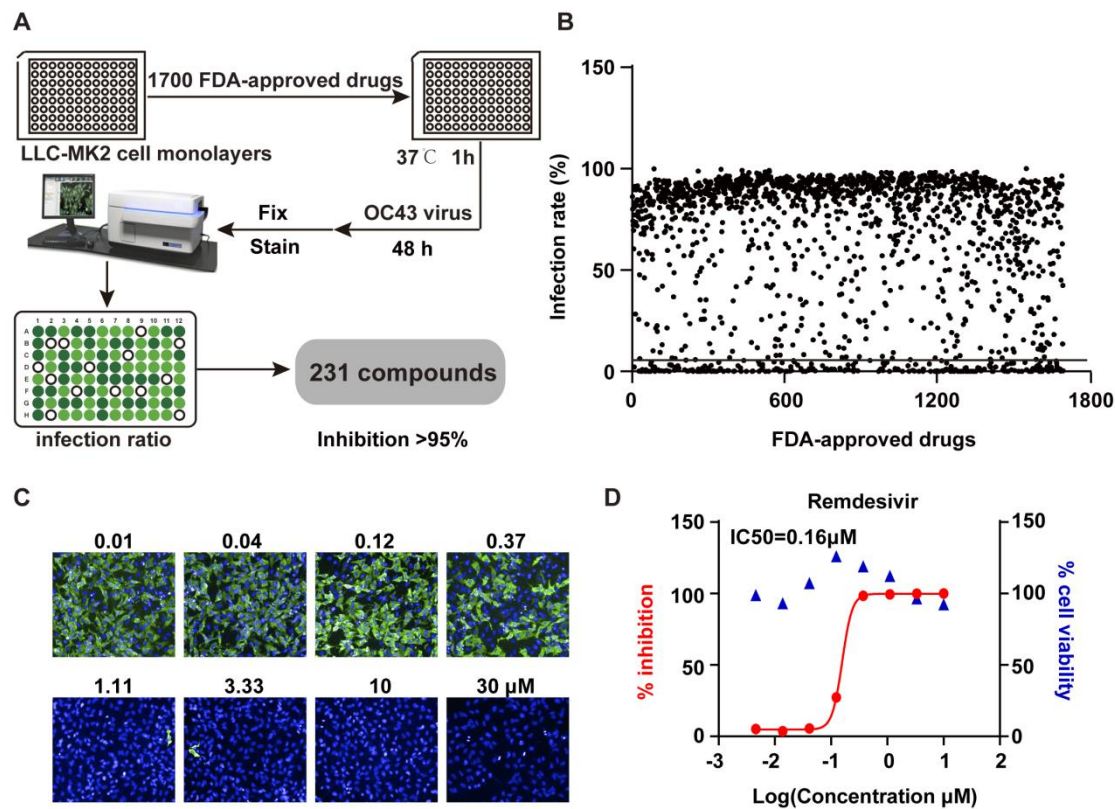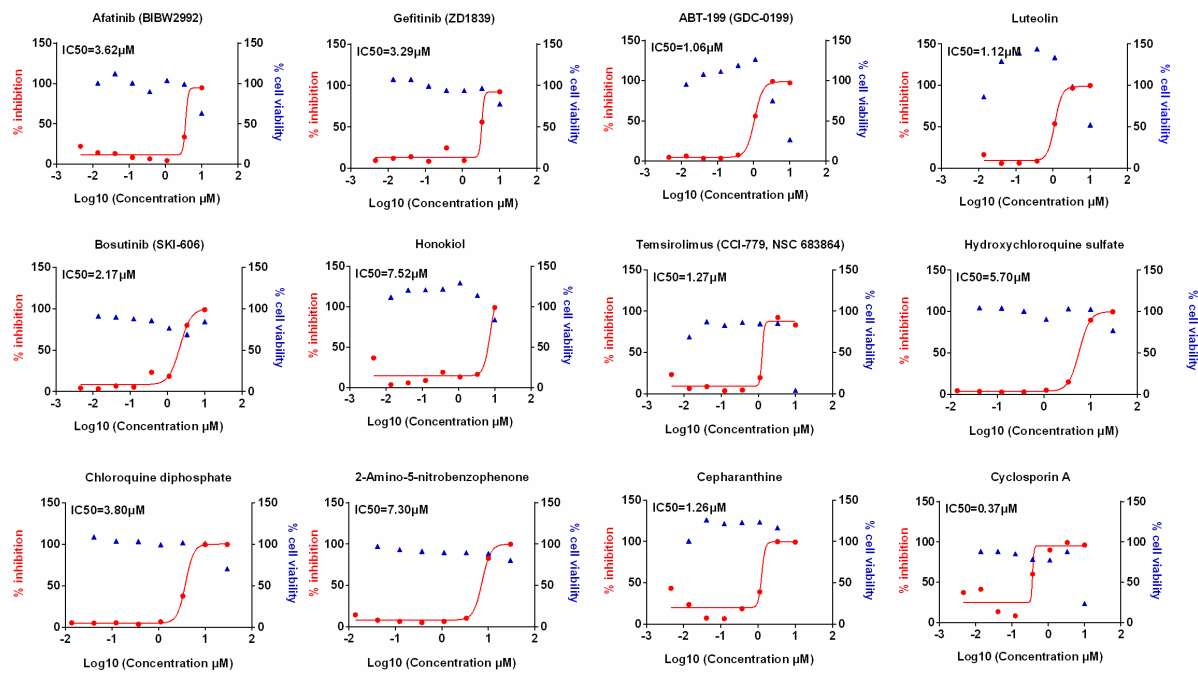

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390   Figure.2

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393   Figure.2 continue-1

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396   Figure.2 continue-2

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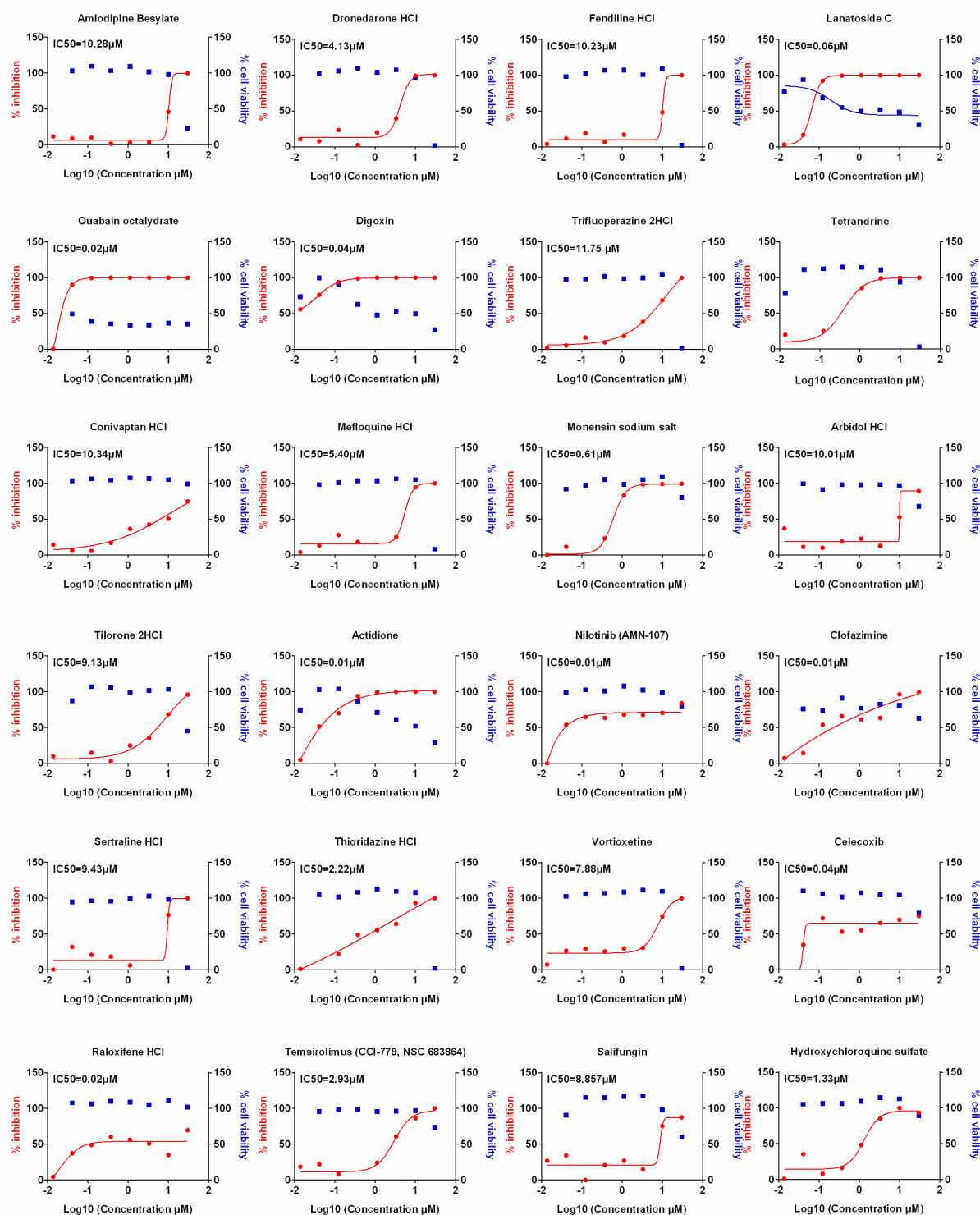
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400   Figure.3

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403   Figure.4
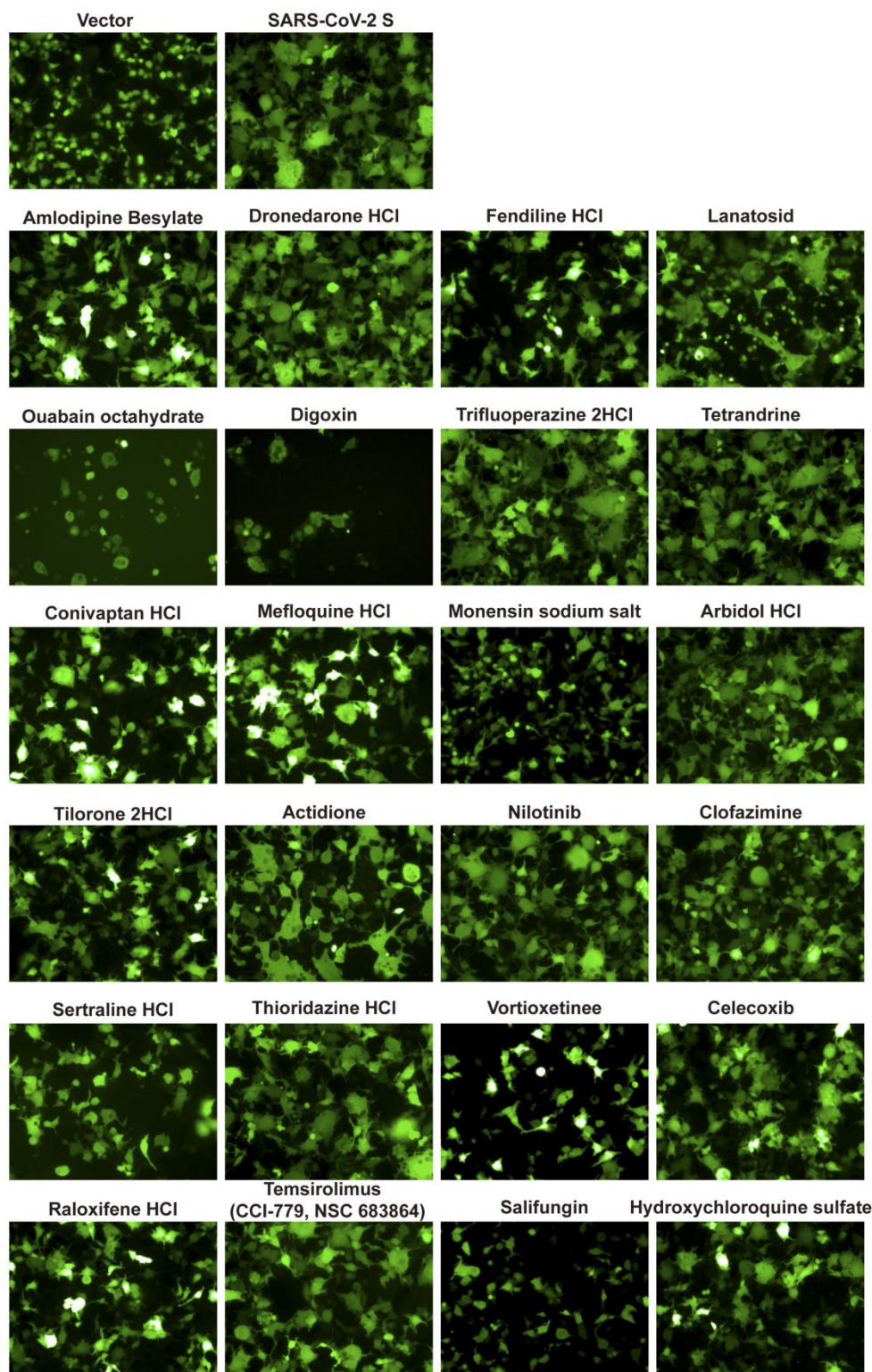
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Table. 1 Antiviral activity of selected compounds against OC43.

Pathway	MOLENAME	Formula	Target	CC_{50} (μM)	IC_{50} (μM)	SI (CC_{50}/IC_{50})
Membrane Transporter /Ion Channel	Amlodipine Besylate	$C_{26}H_{31}ClN_2O_8S$	Carbonic anhydrase inhibitor; Calcium Channel inhibitor; PDE inhibitor	28.9	6.49	4.45
	Butyl 4-aminobenzoate	$C_{11}H_{15}NO_2$	Sodium Channel inhibitor	>30	8.36	>3.59
	Manidipine 2HCl	$C_{35}H_{40}Cl_2N_4O_6$	Calcium Channel antagonist	>30	3.66	>8.20
	Efonidipine(NZ-105)	$C_{34}H_{38}N_3O_7P$	Calcium Channel inhibitor	>10	3.84	>2.60
	Trifluoperazine 2HCl	$C_{21}H_{26}Cl_2F_3N_3S$	Dopamine Receptor; Adrenergic Receptor antagonist;Calmodulin inhibitor	>10	3.26	>3.08

	Fendiline HCl	$C_{23}H_{26}ClN$	Calcium Channel inhibitor	>10	4.51	>2.22
	Digoxin	$C_{41}H_{64}O_{14}$	ATPase inhibitor	1.0	0.24	4.17
	Astemizole	$C_{28}H_{31}FN_4O$	Histamine Receptor;Potassium Channel inhibitor	>10	3.91	>2.52
	Perphenazine	$C_{21}H_{26}ClN_3OS$	Calmodulin inhibitor;Dopamine Receptor antagonist	>10	3.97	>2.52
	Dronedarone HCl	$C_{31}H_{44}N_2O_5S.HCl$	Potassium Channel inhibitor ;Sodium Channel inhibitor;Adrenergic Receptor antagonist ; Calcium Channel inhibitor	>10	4.75	>2.11
	Otilonium bromide	$C_{29}H_{43}BrN_2O_4$	Calcium Channel inhibitor;AChR antagonist	>10	1.84	>5.43
	Terfenadine	$C_{32}H_{41}NO_2$	AChR antagonist;Histamine antagonist;Potassium Channel inhibitor	4.3	4.51	0.95
GPCR/G	Conivaptan	$C_{32}H_{26}N_4O_2.HCl$	Vasopressin Receptor antagonist	>10	1.58	>6.33

Protein	HCl					
	Rimonabant HCl	$C_{22}H_{21}Cl_3N_4O$	Cannabinoid Receptor	>10	4.11	>2.43
Microbiology & Virology	Tilorone 2HCl	$C_{25}H_{34}N_2O_3$	Antiviral	>30	1.21	>24.79
	Butylparaben	$C_{11}H_{14}O_3$	Antibacterial	>30	1.11	>27.03
	Acetylspiramycin	$C_{45}H_{76}N_2O_{15}$	Antibiotic	>30	7.49	>4.00
	Dichlorophen	$C_{13}H_{10}Cl_2O_2$	Antibiotic	9.3	2.74	3.39
	Monensin sodium salt	$C_{36}H_{61}NaO_{11}$	Antibiotic	>10	0.07	>142.86
	Chlorhexidine HCl	$C_{22}H_{32}Cl_4N_{10}$	Antibacterial	9.8	3.32	2.95
	Milbemycin oxime	$C_{63}H_{88}N_2O_{14}$	Antiparasitic	3.1	1.19	2.61

	Abamectin	$C_{49}H_{74}O_{14}$	Antiparasitic	~10	3.12	~3.21
	Amphotericin B	$C_{47}H_{73}NO_{17}$	Antibiotic	3.8	2.93	1.30
	Mefloquine HCl	$C_{17}H_{17}ClF_6N_2O$	Hemozoin synthesis inhibitor	12.52	3.62	3.46
	Moxidectin	$C_{37}H_{53}NO_8$	Antibiotic;GluCl channels	~10	3.02	~3.31
Tyrosine Kinase/Adaptors	Imatinib Mesylate (STI571)	$C_{29}H_{31}N_7O.CH_4SO_3$	Bcr-Abl inhibitor;c-kit inhibitor; PDGFR inhibitor	>30	6.09	>4.93
	Neratinib(HKI-272)	$C_{30}H_{29}ClN_6O_3$	EGFR inhibitor; HER2 inhibitor	>10	1.02	>9.80
	Nilotinib (AMN-107)	$C_{28}H_{22}F_3N_7O$	Bcr-Abl inhibitor;Mast/stem cell GFR Kit agonist	>10	1.34	>7.46
	Afatinib (BIBW2992) Dimaleate	$C_{32}H_{33}ClFN_5O_{11}$	EGFR inhibitor; HER2 inhibitor	>10	3.62	2.76

	Gefitinib (ZD1839)	$C_{22}H_{24}ClFN_4O_3$	EGFR antagonist	>10	3.29	>3.34
Enzyme	Clofazimine	$C_{27}H_{22}Cl_2N_4$	CzcO-like inhibitor;DNA intercalation	~10	0.35	~28.57
Neuroscience	Cyproheptadine HCl	$C_{21}H_{22}ClN$	Histamine Receptor antagonist;5-HT receptor antagonist;AChR antagonist	>30	8.36	>3.59
	Cloperastine HCl	$C_{20}H_{25}Cl_2NO$	Histamine Receptor antagonist	>30	5.51	>5.44
	Dihydroergotamine Mesylate	$C_{34}H_{41}N_5O_8S$	5-HT Receptor antagonist	>30	5.24	>5.73
	Azelastine	$C_{22}H_{25}Cl_2N_3O$	Histamine Receptor antagonist	>30	12.01	>2.50
	Pizotifen	$C_{19}H_{21}NS$	5-HT Receptor antagonist	>30	8.36	>3.59
	Fluoxetine hydrochloride	$C_{17}H_{18}F_3NO.HCl$	5-HT Receptor antagonist	>10	3.16	>3.16
	Aprepitant	$C_{23}H_{21}F_7N_4O_3$	Substance P antagonist	>10	10.12	>0.99
	Thioridazine	$C_{21}H_{27}ClN_2S_2$	5-HT Receptor antagonist;Adrenergic	>10	3.42	>2.92

	HCl		Receptor antagonist;Dopamine Receptor antagonist;Potassium Channel inhibitor			
	Chlorpromazine HCl	$C_{17}H_{19}ClN_2S.HCl$	Histamine Receptor antagonist;Potassium Channel inhibitor;Dopamine Receptor antagonist;Adrenergic Receptor antagonist;AChR antagonist;5-HT receptor antagonist	>10	2.61	>3.83
	Vortioxetine	$C_{18}H_{22}N_2S$	5-HT Receptor modulator	>10	1.93	>5.18
	Triflupromazine HCl	$C_{18}H_{20}ClF_3N_2S$	Dopamine Receptor antagonist;AChR antagonist;5-HT receptor antagonist	>10	7.49	>1.34
	Flupenthixol 2HCl	$C_{23}H_{27}Cl_2F_3N_2OS$	Dopamine Receptor antagonist	>10	3.52	>2.84
	Sertraline HCl	$C_{17}H_{17}CI_2N.HCl$	5-HT Receptor antagonist	>10	3.49	>2.87
Endocrinol	Dydrogesteron	$C_{21}H_{28}O_2$	Progesterone Receptor agonist	>30	8.36	>3.59

ogy/ Hormones	e					
	Epiandrosterone	$C_{19}H_{30}O_2$	Androgen Receptor agonist	>30	8.36	>3.59
	Raloxifene HCl	$C_{28}H_{28}ClNO_4S$	Estrogen/Progestogen Receptor antagonist; SERM agonist	>10	1.32	>7.58
Angiogenesis	Ibrutinib (PCI-32765)	$C_{25}H_{24}N_6O_2$	BTK inhibitor	>10	3.62	>2.76
	Suloctidil	$C_{20}H_{35}NOS$	Antiplatelet aggregation inhibitor	>10	9.13	>1.10
	Candesartan Cilexetil	$C_{33}H_{34}N_6O_6$	RAAS antagonist;ACE inhibitor	~10	3.59	~2.79
Others	ABT-199 (GDC-0199)	$C_{45}H_{50}ClN7O_7S$	Bcl-2 inhibitor	3.4	1.06	3.21
	Luteolin	$C_{15}H_{10}O_6$	TNF-alpha inhibitor, IL inhibitor; NF-κB inhibitor	3.5	1.12	3.13
	Bosutinib	$C_{26}H_{29}Cl_2N_5O_3$	MAPK inhibitor;Src inhibitor;Bcr-	>10	2.17	>4.61

(SKI-606)		Abl inhibitor;CaMK inhibitor;CDK inhibitor			
Honokiol	$C_{18}H_{18}O_2$	Akt inhibitor;MEK inhibitor	>10	7.52	>1.33
Temsirolimus (CCI-779, NSC 683864)	$C_{56}H_{87}NO_{16}$	mTOR inhibitor	9.3	1.27	7.32
Hydroxychloroquine sulfate	$C_{18}H_{26}ClN_3O.H_2SO_4$	Autophagy inhibitor;TLR antagonist	>30	5.7	>5.26
Chloroquine diphosphate	$C_{18}H_{26}CLN_3.2(H_3PO_4)$	Autophagy inhibitor;ATM/ATR activator	>30	3.8	>7.89
2-Amino-5-nitrobenzophenone	$C_{13}H_{10}N_2O_3$	Intermediates	>30	7.3	>4.11
Cepharanthine	$C_{37}H_{38}N_2O_6$	Others	9.1	1.26	7.22
Escin	$C_{55}H_{86}O_{24}$	Others	>10	3.31	>3.02
Cyclosporin A	$C_{62}H_{111}N_{11}O_{12}$	Others	~10	0.37	~27.03

Table. 2 Antiviral activity of selected compounds against SARS-CoV-2.

Pathway	MOLENAME	Formula	CC$_{50}$ (μM)	IC$_{50}$ (μM)	SI (CC$_{50}$/IC$_{50}$)
Membrane Transporter/Ion Channel	Amlodipine Besylate	$C_{26}H_{31}ClN_2O_8S$	>30	10.28	>2.92
	Dronedarone HCl	$C_{31}H_{44}N_2O_5S.HCl$	12.62	4.13	3.06
	Fendiline HCl	$C_{23}H_{26}ClN$	29.24	10.23	2.86
	Digitoxin	$C_{41}H_{64}O_{13}$	3.03	0.1	30.30
	Lanatoside C	$C_{49}H_{76}O_{20}$	0.2	0.06	3.33
	Ouabain octahydrate	$C_{29}H_{44}O_{12}8H_2O$	0.07	0.02	3.5
	Digoxin	$C_{41}H_{64}O_{14}$	0.36	0.036	10
	Trifluoperazine 2HCl	$C_{21}H_{26}Cl_2F_3N_3S$	29.29	11.75	2.49
	Tetrandrine	$C_{38}H_{42}N_2O_6$	12.59	0.39	32.28
GPCR/G Protein	Conivaptan HCl	$C_{32}H_{26}N_4O_2.HCl$	12.7	10.34	1.23
Microbiology & Virology	Mefloquine HCl	$C_{17}H_{17}ClF_6N_2O$	29.13	5.4	5.39
	Monensin sodium salt	$C_{36}H_{61}NaO_{11}$	>30	0.6019	>49.84
	Arbidol HCl	$C_{22}H_{25}BrN_2O_3S.HCl$	22.36	10.01	2.23
	Tilorone 2HCl	$C_{25}H_{34}N_2O_3$	28.67	9.13	3.14
	Actidione	$C_{15}H_{23}NO_4$	6.06	0.01	606
Tyrosine Kinase/Adaptors	Nilotinib (AMN-107)	$C_{28}H_{22}F_3N_7O$	>30	<0.01	>3000
	Vandetanib	$C_{22}H_{24}BrFN_4O_2$	>30	>30	>1

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Enzyme	Clofazimine	$C_{27}H_{22}Cl_2N_4$	>30	0.01	>3000
Neuroscience	Sertraline HCl	$C_{17}H_{17}CI_2N.HCl$	27.84	9.34	2.98
	Thioridazine HCl	$C_{21}H_{27}ClN_2S_2$	27.22	2.22	12.26
	Vortioxetine	$C_{18}H_{22}N_2S.HBr$	28.03	7.88	3.56
	Celecoxib	$C_{17}H_{14}F_3N_3O_2S$	>30	0.04	>750
Endocrinology/ Hormones	Raloxifene HCl	$C_{28}H_{28}ClNO_4S$	>30	0.02	>1500
Others	Temsirolimus (CCI-779, NSC 683864)	$C_{56}H_{87}NO_{16}$	>30	2.93	>10.24
	Salifungin	$C_{13}H_9BrClNO_2$	11.12	8.86	1.26
	Hydroxychloroquine sulfate	$C_{18}H_{26}ClN_3O.H_2SO_4$	>30	1.33	>22.56