Synthesis and Antiviral Activities of Neoechinulin B and Its Derivatives

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ABSTRACT: We have previously reported that neoechinulin B (1a), a prenylated indole diketopiperazine alkaloid, shows antiviral activities against hepatitis C virus (HCV) via the inactivation of the liver X receptors (LXRs) and the resultant disruption of double-membrane vesicles. In this study, a two-step synthesis of the diketopiperazine scaffold of 1a was achieved by the base-induced coupling of 1,4-diacetyl-3-[(tert-butyldimethylsilyl)oxy]methyl)piperazine-2,5-dione with aldehydes, followed by the treatment of the resultant coupling products with tetra-n-butylammonium fluoride. Compound 1a and its 16 derivatives 1b−q were prepared using this method. Furthermore, variecolorin H, a related alkaloid, was obtained by the acid treatment of 1a in MeOH. The antiviral evaluation of 1a and its derivatives revealed that 1a, 1c, 1d, 1h, 1j, 1l, and 1o exhibited both anti-HCV and anti-severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) activities. The results of this study indicate that the exomethylene moiety on the diketopiperazine ring is important for the antiviral activities. The antiviral compounds can inhibit the production of HCV and SARS-CoV-2 by inactivating LXRs.

The diversity and complexity of natural products afford remarkable efficacy and specificity to target viral infections. Therefore, natural products serve as excellent sources for discovering antiviral agents. We have previously reported that neoechinulin B (1a), isolated from Eurotium rubrum Hiji025, exhibited antiviral effects against hepatitis C virus (HCV). Mechanistic studies revealed that this compound disrupted the formation of double-membrane vesicles (DMVs), which are the sites of viral RNA replication, by inhibiting the liver X receptor (LXR)-regulated gene induction required for DMV formation. Consistent with the unique mechanism of action targeting LXRs, which are those of the host-encoded proteins that HCV hijacks during its replication, compound 1a augmented the antiviral activity of the approved anti-HCV agents that target viral proteins via combination treatment. Notably, 1a reduced the RNA replication of poliovirus in DMVs. This compound was also reported to inhibit the entry of influenza A virus subtype H1N1 by targeting viral hemagglutinin. These reports suggest that 1a and its derivatives are potential broad-spectrum antiviral drugs.

Despite its potential as a lead compound for a new class of antiviral agents, only one report by Inoue, Kishi, and co-workers in 1977 has described the chemical synthesis of 1a (Scheme 1). A key step in its synthesis involved the coupling between aldehyde 2a and diketopiperazine 3 in dry piperidine at 110 °C, which afforded 1a in 45% yield. This reaction...
condition was also employed by Kuttruff, Zipse, and Trauner to prepare 1a as a key building block in the synthesis of variecolorotide B. However, there have been no attempts to systematically synthesize neoechinulin B derivatives using this protocol. Thus, in this study, we attempted to develop an alternative synthetic route to 1a and its derivatives under mild conditions. Building upon our previous findings, extensive structure–activity relationship studies were also conducted to identify simplified derivatives with greater potency against HCV. In addition, the LXR antagonistic activity of the synthesized compounds was evaluated to investigate the mechanisms of the anti-HCV activity.

Herein, the synthesis of 1a and a series of simplified derivatives was achieved. Antiviral activities of these compounds against HCV and severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) are described. In addition to HCV and poliovirus, SARS-CoV-2 replicates genome RNA in DMVs in the infected cells. Therefore, the antiviral activity of 1a and its derivatives against SARS-CoV-2 was evaluated in this study. As expected, 1a exhibited both anti-HCV and anti-SARS-CoV-2 activities. Furthermore, the simplification of 1a afforded a series of derivatives that exhibited more potent antiviral activities against both anti-HCV and anti-SARS-CoV-2.

■ RESULTS AND DISCUSSION

**Synthetic Approach.** It was envisioned that 2,5-diketopiperazine core 1 could be synthesized using aldehyde 2 and 1,4-diacytetyl-3-{{((tert-butyldimethylsilyl)oxy)methyl}-piperazine-2,5-dione (4) in two steps (Scheme 2). The base-induced coupling of 2 with 4 produces intermediate 1. The migration of the acetyl group, followed by the elimination of the acetoxyl group in the resultant intermediate II, affords 5. Similarly, the removal of the tert-butyldimethylsilyl (TBS) group in 5 and migration of the acetyl group in the resultant intermediate III, followed by the elimination of the acetoxyl group in intermediate IV, affords 1. Synthesis of Deprenylneoechinulin B (1b) and 3-Arylmethylene-6-methylenepiperazine-2,5-diones. As a model compound, 1b was synthesized using the procedure shown in Scheme 2. Treatment of N-acetyl-3-indolecarboxaldehyde (2b) and 4 with t-BuOK in N,N-dimethylformamide (DMF) afforded 5b in 89% yield (Scheme 3). The subsequent treatment of 5b with tetra-n-butylammonium fluoride (TBAF) afforded the desired compound 1b′ in 75% yield. The deacetylation of 1b′ by hydrazine monohydrate in DMF generated 1b in 40% yield. To demonstrate the utility of this method, 1b was also synthesized according to the protocol reported by Inoue, Kishi, and co-workers (Scheme 4). Coupling between 2b′ and 3 in piperidine at 110 °C afforded the desired compound 1b in only 4% yield. One of the starting materials, 2b′, was also recovered in 42% yield, and the rest of the compounds underwent side reactions to form unidentified products.

**Scheme 2. Synthetic Approach toward 2,5-Diketopiperazine Core 1**

**Scheme 3. Synthesis of Deprenylneoechinulin B (1b)**

**Scheme 4. Synthesis of 1b According to the Protocol Reported by Inoue, Kishi, and Co-workers**

![Scheme 2](https://example.com/scheme2.png)

![Scheme 3](https://example.com/scheme3.png)

![Scheme 4](https://example.com/scheme4.png)
byproducts. These results indicate that the alternative method described herein can be used for the synthesis of 2,5-diketopiperazine core 1.

Next, the scope and limitations of this method were examined (Scheme 5). The present method was found to be applicable to the synthesis of a series of 3-arylmethylene-6-methyleneepiperazines bearing benzene (1c and 1f–o), furan (1d), thiophene (1e), naphthalene (1p), and pyrene (1q) groups. However, the coupling between 2-isopropylbenzaldehyde (1r) and 4 did not occur owing to the steric hindrance of the ortho-substituted isopropyl group.

**Synthesis of 1a and Variecolorin H.** This methodology was next applied to the synthesis of 1a (Scheme 6). Compound 2s, which possesses a methoxymethyl (MOM) group, was used as the substrate. Unfortunately, coupling between 2s and 4 afforded only a small amount of the desired product 5s, and considerable amounts of 2s and 4 were recovered. The reaction was repeated three times to obtain 5s in a total yield of 14%, while 2s was recovered in 61% yield. The reaction was also screened using different bases and solvents and at different temperatures, but the yield of 2s did not improve. These results indicated that this coupling reaction was sensitive to steric hindrance. The treatment of 5s with TBAF generated 1s in 84% yield. Finally, the removal of the MOM group from 1s under weak acidic conditions provided 1a in 77% yield. The spectroscopic data of 1a synthesized in this study were identical to those of the natural product 1a.

During the purification of 1a by silica gel chromatography using CHCl₃ and MeOH as eluents, 1a was frequently transformed into variecolorin H (6), a related alkaloid isolated from a halotolerant strain of *Aspergillus variecolor*. The treatment of 1a with a catalytic amount of H₂SO₄ in MeOH

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**Scheme 5. Coupling of Aldehydes 2c–r with Diketopiperazine 4 and Transformation of Intermediates 5c–q into 3-Arylmethylene-6-methyleneepiperazines 1c–q**

**Scheme 6. Synthesis of 1a**

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"Reaction was performed with 2 (0.20 mmol), 4 (0.40 mmol), and t-BuOK (0.58 mmol) in DMF (2.0 mL) at rt. Isolated yields are presented, unless otherwise stated. The yield based on recovered 2m is included in parentheses."
afforded 6 in 74% yield (Scheme 7). The specific rotation of 6 was reported to be $[\alpha]_{D}^{25} -0.3$ (c 0.3, MeOH), suggesting that it is a racemate. These results suggested that 6 could be an artifact generated during isolation.

Scheme 7. Synthesis of Variecolorin H (6)

![Scheme 7](image)

Inhibitory Effect on HCV Production. The cytotoxicity and anti-HCV activity of 1a–q, 6, neoechinulin A (7),13 and preechinulin (8)13 were evaluated. Neoehcinulin A (7) has a single bond between C-12 and C-20, whereas preechinulin (8) has two single bonds between C-8 and C-9 and between C-12 and C-20. The 50% and 90% virus inhibitory concentrations (IC50 and IC90) and 50% cytotoxic concentration (CC50) of 1a–q and 6–8 are summarized in Table 1. The CC50 values of 1a–q and 6–8 were greater than 20 μM, indicating negligible cytotoxic activity against the host Huh7.5.1 cells at concentrations below 20 μM (Figure S1 in the Supporting Information). The anti-HCV activity of 1b–q and 6–8 were evaluated using 1a as a positive control (Figure S2 in the Supporting Information).3 Interestingly, compounds 1a–q showed antiviral activity irrespective of the structure of the aromatic moiety (Table 1). In contrast, compounds 6–8 did not show antiviral activity. These results clearly indicate that the exomethylene moiety in 1a is important for the anti-HCV activity. It is possible that compounds 1a–q act as electrophilic Michael acceptors with cellular nucleophiles such as proteins and DNA bases. However, the lack of cytotoxicity of 1a–q can rule out the possibility that they nonspecifically bind to these biomolecules via Michael addition. Only compounds 1l, 1n, and 1p reduced the relative virus production by more than 90%, with IC90 values of 6.5, 9.7, and 1.9 μM, respectively (Figure 1). The other compounds did not suppress the virus production by up to 90% at concentrations below 20 μM. Compounds 1l and 1n contain electron-withdrawing methoxycarbonyl and trifluoromethyl groups, respectively, on the benzene ring, suggesting that the electron density on the benzene ring could influence the anti-HCV activity.

Effect on LXR-Mediated Transcriptional Activity. The effect of 1c and 1g on LXR-mediated transcriptional activity was examined using 1a as a positive control. We performed the reporter gene assay using an LXR element (LXRE)-driven luciferase plasmid to investigate whether they inhibit the transactivation of LXRs.3 T0901317,17 which is an agonist of LXRs, increased the LXRE-driven luciferase activity (Figure 2). Compounds 1a and 1g reduced the T0901317-induced reporter activity mediated by LXRs. Thus, 1a and 1g showed anti-HCV activity due to the inhibition of LXR-regulated gene induction, which is required for DMV formation. However, to our surprise, compound 1c did not show LXR antagonistic activity even at 30 μM, although it showed anti-HCV activity. This suggests that 1c exhibited antiviral activity via different mechanisms. We have previously reported that 1a interacts with recombinant LXRα and LXRβ, based on the results of surface plasmon resonance (SPR) analysis.3 The association of 1a with LXRs and dissociation of 1a from LXRs were observed in the SPR curves, suggesting that neoechinulin B and its derivatives interact noncovalently with LXRs. Although this interaction does not completely exclude the possibility of the reaction of the exomethylene moiety in 1a with the thiol or amine moiety in LXRs, the lack of the LXR antagonistic activity of 1c suggests that the diketopiperazine moiety in 1c does not bind to LXRs and the overall structure of 1a is important for the specific binding with LXRs. This result also rules out the possibility that the exomethylene moiety on the

![Figure 1](image)

Table 1. Anti-HCV Activity (IC50 and IC90, μM) and Cytotoxicity (CC50, μM) Data for Compounds 1a–q

| compound | IC50 (μM) | IC90 (μM) | CC50 (μM) |
|----------|----------|-----------|-----------|
| 1a       | 4.7 ± 1.4 | >20       | >20       |
| 1b       | 2.2 ± 0.64 | >20       | >20       |
| 1c       | 0.0559 ± 0.0042 | >20       | >20       |
| 1d       | 1.4 ± 0.34 | >20       | >20       |
| 1e       | 1.4 ± 0.48 | >20       | >20       |
| 1f       | 0.015 ± 0.019 | >20       | >20       |
| 1g       | 1.1 ± 0.34 | >20       | >20       |
| 1h       | 2.7 ± 1.4 | >20       | >20       |
| 1i       | 1.3 ± 0.42 | >20       | >20       |
| 1j       | 2.0 ± 0.068 | >20       | >20       |
| 1k       | 1.1 ± 0.60 | >20       | >20       |
| 1l       | 1.2 ± 0.16 | 6.5 ± 1.84 | >20       |
| 1m       | 1.2 ± 0.54 | >20       | >20       |
| 1n       | 1.6 ± 0.32 | 9.7 ± 1.06 | >20       |
| 1o       | 4.9 ± 0.66 | >20       | >20       |
| 1p       | 0.26 ± 0.11 | 1.9 ± 0.65 | >20       |
| 1q       | 3.9 ± 0.45 | >20       | >20       |
| 6        | >20 | >20       | >20       |
| 7        | >20 | >20       | >20       |
| 8        | >20 | >20       | >20       |

*All experiments were performed in triplicate and means ± standard deviations (SD) are reported.11 IC50 and IC90 values were determined according to the procedure described previously.12 CC50 values were determined by the MTT assay.16*
The values represent means ± SD. The statistical significance was assessed by Tukey’s honestly significant difference (HSD) test, and the asterisks ** and * indicate \( p < 0.01 \) and \( p < 0.05 \), respectively.

**Inhibitory Effect on SARS-CoV-2 Production.** The cytotoxicity and anti-SARS-CoV-2 activity of 1a–q and 6–8 were evaluated by employing a cell-based SARS-CoV-2 infection system using VeroE6 cells expressing the transmembrane serine protease TMPRSS2 (VeroE6/TMPRSS2 cells; Figures S3 and S4 in the Supporting Information).\(^8\)\(^9\)\(^\text{18,19}\) The IC\(_{50}\), IC\(_{90}\), and CC\(_{50}\) values of the compounds are summarized in Table 2. Compound 1a reduced the production of SARS-CoV-2 RNA with IC\(_{50}\) and IC\(_{90}\) values of 32.9 and 45.6 \( \mu \)M, respectively, without exhibiting any remarkable cytotoxicity at these concentrations. Compounds 1b, 1f, 1g, 1k, 1p, and 6–8 did not show anti-SARS-CoV-2 activity. The lack of antiviral activity of 6–8 indicates that the exomethylene moiety in 1a is important for the anti-SARS-CoV-2 activity. Because compounds 1e, 1i, 1m, and 1n showed toxicity toward the host cells, the inhibition of virus production by these compounds could not be evaluated accurately. In contrast, compounds 1c, 1d, 1h, 1j, 1l, and 1o exhibited anti-SARS-CoV-2 activity. The antiviral activity of 1c, 1d, 1h, 1j, 1l, and 1o was more potent than that of 1a. Particularly, 1d was more potent and less toxic than 1a (Figure 3). It can thus be concluded that the combination of the diketopiperazine core and the aromatic moiety is important for the anti-SARS-CoV-2 activity. No distinct relationships between the structural features and anti-SARS-CoV-2 activity of 1a, 1c, 1d, 1h, 1j, 1l, and 1o were identified, suggesting that these compounds acted through different or multiple mechanisms.

**CONCLUSION**

The diketopiperazine scaffold of 1a was successfully constructed in this study. Seventeen 3-arylmethylene-6-methylene-piperazine-2,5-diones (1a–q) were synthesized by the coupling of aldehydes 2 and 1,4-diacyl-3-(\{[\text{tert-butylidimethylsilyl}oxy]methyl\}piperazine-2,5-dione (4), followed by the TBAF treatment of the coupling products 5. Although 1a was successfully synthesized, the yield of the coupling reaction between 2a and 4 needed improvements. Compound 6 was formed during the purification of 1a by silica gel chromatography using CHCl\(_3\) and MeOH as eluents, as well as upon the acid treatment of 1a in MeOH, suggesting that 6 could be an artifact formed during extraction or purification.

The antiviral activities of 1a–q and 6–8 against HCV and SARS-CoV-2 were evaluated. Compound 1a showed antiviral activities against HCV and SARS-CoV-2. In contrast, the structurally related compounds 6, 7, and 8 did not show any

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**Table 2. Anti-SARS-CoV-2 Activity (IC\(_{50}\) and IC\(_{90}\) \( \mu \)M) and Cytotoxicity (CC\(_{50}\) \( \mu \)M) Data for Compounds 1a–q**

| compound | IC\(_{50}\) (\( \mu \)M)\(^b\) | IC\(_{90}\) (\( \mu \)M)\(^b\) | CC\(_{50}\) (\( \mu \)M)\(^c\) |
|----------|-------------------------------|-------------------------------|------------------|
| 1a       | 32.9 ± 13.2                   | 45.6 ± 7.4                    | >70              |
| 1b       | >40                           | >40                           | >100             |
| 1c       | 13.6 ± 3.2                    | 20.3 ± 3.2                    | 40.1 ± 10.5      |
| 1d       | 9.3 ± 6.2                     | 21.2 ± 9.0                    | >80              |
| 1e       | –\(^d\)                      | –\(^d\)                      | 42.2 ± 7.9       |
| 1f       | –\(^e\)                      | >40                           | >100             |
| 1g       | –\(^e\)                      | >40                           | >100             |
| 1h       | 6.0 ± 2.4                     | 16.3 ± 6.2                    | 63.1 ± 17.9      |
| 1i       | –\(^d\)                      | –\(^d\)                      | 47.2 ± 0.6       |
| 1j       | 10.3 ± 3.7                    | 20.1 ± 8.5                    | 62.8 ± 17.4      |
| 1k       | –\(^e\)                      | >40                           | 78.8 ± 4.5       |
| 1l       | 8.4 ± 2.1                     | 14.1 ± 1.8                    | >50              |
| 1m       | –\(^d\)                      | –\(^d\)                      | 28.3 ± 6.2       |
| 1n       | 9.1 ± 0.7                     | –\(^d\)                      | 30.1 ± 7.3       |
| 1o       | 20.8 ± 8.0                    | >40                           | >100             |
| 1p       | >40                           | >40                           | 40.7 ± 20.1      |
| 1q       | >40                           | >40                           | >100             |
| 6        | 6                             | >40                           | >100             |
| 7        | >40                           | >40                           | >100             |
| 8        | >40                           | >40                           | >100             |

\(^{\text{Values shown are the means ± SD of triplicate measurements.}}\(^{\text{b}}\)IC\(_{50}\)
and IC\(_{90}\) values were determined according to a previously reported procedure.\(^\text{17}\) \(^{\text{c}}\)CC\(_{50}\) values were determined by the quantification of the survival cell numbers after fixation with 4% paraformaldehyde and staining with 0.02% DAPI. \(^{17}\) IC\(_{50}\) and IC\(_{90}\) values were not determined because they were much higher than CC\(_{50}\).

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**Figure 3. Structure–activity relationships of 1a and its derivatives as inhibitors of SARS-CoV-2 production.**
antiviral activities. These results indicate that the exomethylene moiety in \(1a\) is important for the antiviral activities against both HCV and SARS-CoV-2. Neochelinin B derivatives \(1b\)–\(q\), which contain the common diketopiperazine scaffold, showed anti-HCV activity. Among the neochelinin B derivatives tested in this study, compounds \(1l\), \(1n\), and \(1p\) showed more potent anti-HCV activity than \(1a\) without exhibiting any serious cytotoxicity. Furthermore, \(1c\), \(1d\), \(1h\), \(1j\), \(1l\), and \(1o\) exhibited anti-SARS-CoV-2 activity. Particularly, \(1c\), \(1d\), \(1h\), \(1j\), and \(1l\) exhibited more potent anti-SARS-CoV-2 activity than \(1a\). The aromatic moieties in \(1a\)–\(q\) significantly influence the anti-SARS-CoV-2 activity and cytotoxicity against host cells. However, no clear relationships between the structural features and anti-SARS-CoV-2 activity of \(1a\), \(1c\), \(1d\), \(1h\), \(1j\), \(1l\), and \(1o\) could be identified, suggesting that these compounds impair the replication of SARS-CoV-2 by different or multiple mechanisms. Although further attempts to improve the antiviral potency and selectivity against HCV and SARS-CoV-2 are necessary, the results of this study clearly show that natural product \(1a\) is one of the promising lead compounds for the development of broad-spectrum antiviral drugs. The antiviral compounds discussed herein can inhibit the production of HCV and SARS-CoV-2 by targeting LXRs. However, compound \(1c\), which showed anti-HCV activity, did not show LXR antagonistic activity, suggesting that this compound showed anti-HCV activity via different mechanisms. Further structural optimization to improve the antiviral efficiency and additional biological studies to elucidate the mechanisms of action are underway and will be reported in due course.

**EXPERIMENTAL SECTION**

### General Experimental Procedures

All reactions sensitive to air or moisture were carried out under an argon atmosphere under anhydrous conditions, unless otherwise noted. Solvents and reagents were used without further purification unless otherwise noted. Analytical TLC was performed using silica gel 60 F\(254\) plates (0.25 mm, normal phase, Merck). Normal phase flash column chromatography was performed using silica gel 60 (particle size 40–63 μm; 230–400 mesh ASTM; SilicaFlash F60, Silicycle Inc.). Melting point (mp) data were determined using a Shimadzu MM-2 instrument and were uncorrected. IR spectra were recorded on a Bruker FT-720 spectrometer, using KBr pellets (solid). \(\mathrm{H}\) and proton-decoupled \(\begin{align*}^{13}\mathrm{C}\end{align*}\) NMR spectra were recorded on a Bruker Avance 400 spectrometer (400 and 100 MHz, respectively), using chloroform-\(d\) (CDCl\(3\)) and dimethyl sulfoxide-\(d\(6\) (DMSO-\(d\(6\))) as solvents. Chemical shift values are expressed in \(\delta\) (ppm) relatively to the residual solvent resonance (CDCl\(3\), \(\delta\) 7.26 for \(\begin{align*}^{1}\mathrm{H}\end{align*}\) NMR and \(\delta\) 77.0 for \(\begin{align*}^{13}\mathrm{C}\end{align*}\) NMR; DMSO-\(d\(6\), \(\delta\) 2.49 for \(\begin{align*}^{1}\mathrm{H}\end{align*}\) NMR and \(\delta\) 39.7 for \(\begin{align*}^{13}\mathrm{C}\end{align*}\) NMR). Data are reported as follows: chemical shift, multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, br = broad, dd = double doublet, m = multiplet), coupling constants (\(J\); Hz), and integration. Mass spectra were obtained by a Sciex X500R quadrupole time-of-flight (QTOF) high-resolution mass spectrometer using electrospray ionization (ESI). Neochelinin A (7) and preechinulin (8), which were prepared in our previous study, were used in this study. Huh7.5.1 cells were obtained from Dr. Francis Chisari at the Scripps Research Institute. 

### General Procedure of Transformation of the Intermediates 5 into Methylenepiperazine-2,5-diones 1 (General Procedure B; Scheme 5)

A 1.0 M solution of TBAF (2.0 equiv) in THF was added to a solution of 5 (1.0 equiv) in THF. The mixture was stirred at rt until no further TLC changes were observed. The reaction was quenched by the addition of a saturated aqueous NaHCO\(_3\) solution. The mixture was diluted with EtOAc. The aqueous layer was extracted with EtOAc three times. The combined organic layer was washed with brine, dried over Na\(_2\)SO\(_4\), and concentrated to give a residue. The residue was purified by silica gel column chromatography. For the specific procedures and spectroscopic data for the prepared compounds, see the Supporting Information.

### General Procedure of Coupling of Aldehydes 2 with Diketopiperazine 4 (General Procedure A; Scheme 5)

A solution of 2 (0.20 mmol, 1.0 equiv) and 4 (0.40 mmol, 2.0 equiv) in DMF (2.0 mL) was stirred at 0 °C for 15 min. t-BuOK (0.58 mmol, 2.9 equiv) was added to the mixture, and the resultant mixture was stirred at rt until no further TLC changes were observed. The reaction was quenched by the addition of a saturated aqueous NaHCO\(_3\) solution. The mixture was diluted with EtOAc. After the layers were separated, the organic layer was washed with H\(_2\)O. The aqueous layer was extracted with EtOAc three times. The combined organic layer was washed with brine, dried over Na\(_2\)SO\(_4\), and concentrated to give a residue. The residue was purified by silica gel column chromatography. For the specific procedures and spectroscopic data for the prepared compounds, see the Supporting Information.
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(THUNDERBIRD Probe one-step qRT-PCR kit, TOYOBO) using phenylindole (DAPI) and washed with PBS four times. The number of surviving cells was quantified with a high-content imaging analyzer, ImageXpress Micro Confocal (Molecular Device, San Jose, CA, USA).

ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acs.jnatprod.1c01120.

Details for synthetic procedures, spectroscopic data, and biological activities for the prepared compounds; copies of 1H and 13C{1H} NMR spectra of the prepared compounds (PDF)

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Huh7.1 and VeroE6/TMPRSS2 cells were kindly provided by Dr. Francis Chisari at The Scripps Research Institute and Dr. Makoto Takeda at National Institute of Infectious Diseases, respectively. HCV JFH-1 strain and SARS-CoV-2 Wk-521 strain were kindly provided by Dr. Takaji Wakita at National Institute of Infectious Diseases and Dr. Shutoku Matsuyama at National Institute of Infectious Diseases. LXRE-driven luciferase plasmid was provided by Dr. Maiko Okada at Tokyo University of Technology. This work was supported by an on-campus grant in TUS, Promotion of Science KAKENHI (20H03499). In addition, Smoking Research Foundation and Japan Society for the Re-emerging Infectious Diseases (JP20fk0108411 and JP20fk0210036) and Research Program on Emerging and Infectious Disease (JP20fk0108511) from the Japan Agency for Medical Research and Health. This work was supported by an on-campus grant in TUS, funded by a donation from the Sumitomo Mitsui Trust Bank, Limited. We acknowledge Ms. Asako Aoyagi for her invaluable assistance with the preparation of the manuscript.

**ACKNOWLEDGMENTS**

The authors declare no competing financial interest.

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