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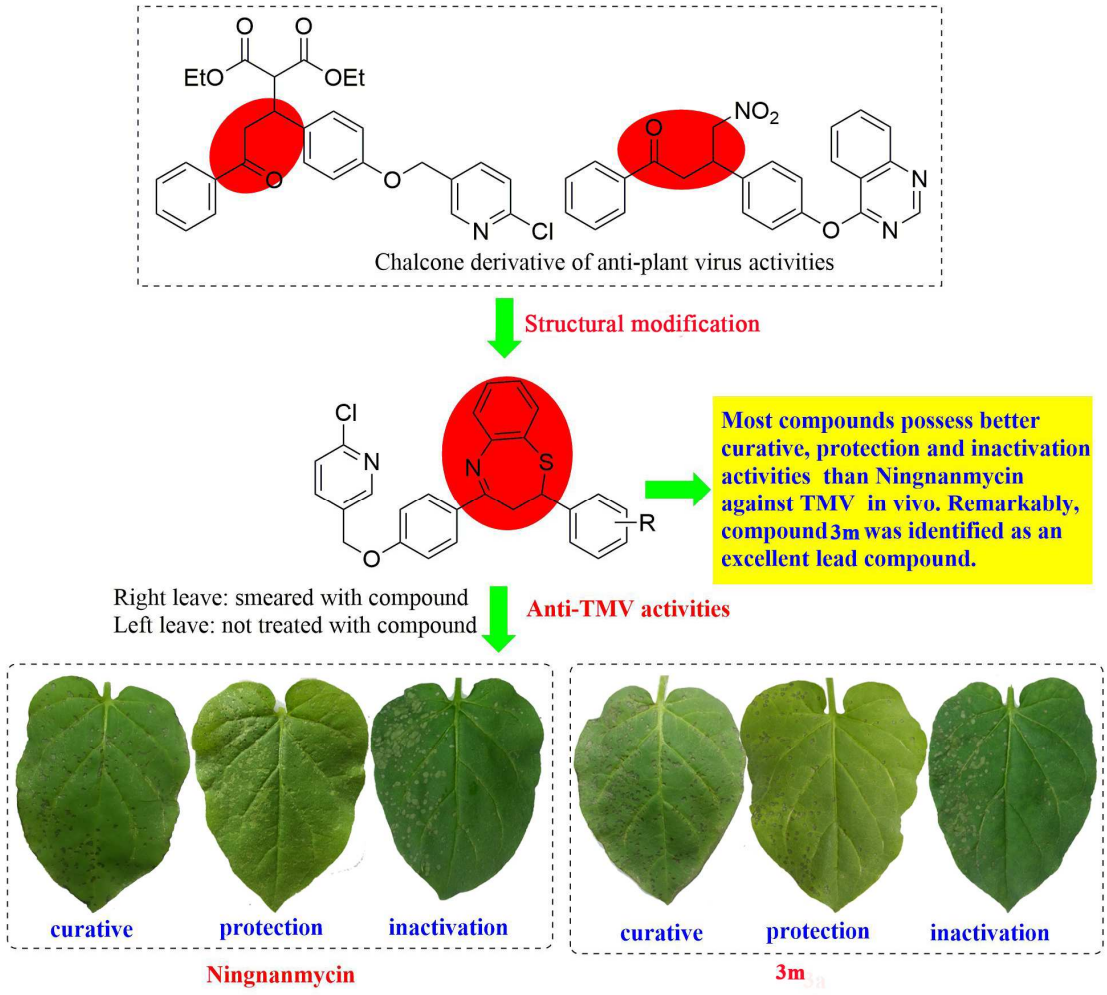
Graphical Abstract

A series of novel benzothiazepine derivatives containing pyridine moiety were synthesized and screened for their antiviral activity against TMV in vivo.



Design, Synthesis, and Antiviral Activities of 1,5-Benzothiazepine Derivatives Containing Pyridine Moiety

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Abstract：In our previous work, a series of novel benzothiazepine derivatives containing pyridine moiety were successfully synthesized through chalcone 1,3-dipolar cycloaddition and determined their antiviral activity against tobacco mosaic virus (TMV). Bioassay results indicated that most of these target compounds exhibited improved curative, protection, and inactivation activity in vivo than the commercial agent ningnanmycin. Particularly, compound **3m** exhibited marked curative activity against TMV, with an EC_{50} value of 352.2 μM, which was even better than that of ningnanmycin. The compound was identified as the most promising candidate for inhibiting plant virus and an excellent compound with antiviral activities against TMV. Structure–activity relationship experiment indicated that the 1,5-benzothiazepine moiety is crucial for potent anti-TMV activity.

Keywords: benzothiazepine derivatives, pyridine, 1,3-dipolar cycloaddition, antiviral activity.

1. Introduction

Tobacco mosaic virus (TMV) is a widespread plant pathogen and can infect several crops, including tobacco, tomato, cucumber, pepper, and ornamental plants, resulting in heavy economic losses [1]. To prevent TMV diseases, researchers have focused on developing novel antiviral agents, such as ningnanmycin [2], seco-pregnane steroids [3], botanical antiviral agents [4], quinazolinones derivatives [5], pyridazines derivatives [6], *β*-carboline derivatives [7], and Gossypol derivatives [8]. However, these compounds present certain limitations to use, such as extremely complex synthesis, easily decomposed active ingredients, relatively low pharmacodynamics [9,10], pesticide-related toxicity, severe pesticide resistance and pesticide interactions [11]. Therefore, a natural, novel, highly efficient and environmentally friendly precursors must be developed for anti-plant virus agents .

Chalcones, as natural products with various pharmacological, have been used for development of biologically functional molecules as activities scaffold. In our previous work, we synthesized and evaluated three series of chalcone derivatives, including nitromethane, malonate ester containing pyridine or quinazolinone moiety [12–14]. The bioassay results revealed that these novel chalcone derivatives showed good antiviral activity against against cucumber mosaic virus (CMV) and TMV. Preliminarily structure–activity relationship (SAR) studies revealed the new skeletons and addition patterns at the *α*,*β*-positions of the chalcone molecule considerably influence the antiviral activity of the compounds.

Benzothiazepines, constitute an important class of benzo-fused and seven-membered heterocyclic compounds containing one nitrogen and one sulfur atom, and thier derivatives have drawn considerable research attention because of their wide range of pharmacological activities, such as antiviral [15–17], antimicrobial [18,19], anticancer [20,21], Ca^{2+} channel antagonist and vasodilator [22,23], CNS depressant [24], antiplatelet aggregation [25], angiotensin converting enzyme (ACE) inhibitor [26], AChE inhibition [27], and anticonvulsant agent [28]. However, biologically active derivatives had been rarely investigated in agricultural field.

To develop 1,3-dipolar cycloaddition chalcone derivatives, a series of novel 1,5-benzothiazepines derivatives containing pyridine moiety were designed as potential anti-TMV agents and synthesized using chalcone at the *α*,*β*-position through 1,3-dipolar cycloaddition (Fig. 1). Anti-TMV activities were evaluated via half-leaf method. Bioassay results indicated that most

of these target compounds exhibited improved curative, protection, and inactivation activity *in vivo* compared with the commercial agent ningnanmycin. Especially, compound **3m** exhibited significant curative activity against TMV, with the EC_{50} value of 352.2 μM, better than that of ningnanmysin. To the best of our knowledge, it's the first report on the anti-TMV activity of 1,5-benzothiazepines derivatives containing pyridine moiety.

Fig. 1

2. Results and discussion

2.1. Chemistry

The synthetic route, designed for the title compounds **3a−3z**, was summarized in **Fig. 2**. Using NaOH solution as catalyst and ethanol as solvent, various substituted aromatic aldehydes were allowed to react with hydroxyacetophenone for 2–3 d at room temperature to obtain intermediates **1a–1z**. Then, using concentrated HCl and anhydrous sodium sulfate as catalysts, intermediates **1a–1z** in ethanol and *o*-aminothiophenol were stirred for 6 h at the reflux temperature to obtain the key intermediates **2a–2z**. Finally, using K_2CO_3/KI as catalysts, intermediates **2a–2z** and compound 2-chloro-5-chloromethyl-pyridine were stirred at 75 ℃ for 5 h and then recrystallized from ethanol to gain the purified target compounds **3a–3z** with the yields of 49%–61%. Their structures were confirmed by IR, ^{1}H NMR, ^{13}C NMR, and elemental analysis, and the physical and analytical data of the target compounds were provided in **Table 1**.

Fig. 2

Table 1

*2.2. Analytical spectral data of the target compounds **3a–3z***

The IR spectra of the title products **3a–3z** exhibited characteristic absorption bands at 1602 cm^{-1} to 1449 cm^{-1} indicating the presence of amidic benzene, Py–rings and C=N. The characteristic absorptions at 1326 cm^{-1} to 1021 cm^{-1} could be attributed to the presence of C–O–C group, and 779 cm^{-1} to 749 cm^{-1} could be attributed to the presence of C–S–C group. ^{1}H NMR analysis indicated that all aromatic ring protons showed multiplets at 8.50 ppm to 6.16 ppm. The main characteristic of the ^{1}H NMR spectra for the target compounds was the presence of the singlet δ_H at 5.16 ppm to 5.13 ppm for O–CH_2– proton. 3.71–2.84 (t, J = 22.50–25.50 Hz, 1H, C-2-H_x), 3.36–3.14 (dd, J_1 = 12.5–13.5 Hz, J_2 = 2.5–4.5 Hz, 1H, C-3-H_a), 6.17–4.93 (dd, J_1 = 12.5–13.5 Hz, J_2 = 4.50–5.00 Hz, 1H, C-3-H_b), in the abx pattern were characterized for two

methylene protons at C-3 and one methine proton at C-2. In the ^{13}C NMR spectrum of those title products **3a–3z,** the chemical shifts 168.37 ppm to 166.67 ppm and 68.91 ppm to 32.18 ppm in ^{13}C NMR also confirmed the presence of C=N and O–CH_2–, –CH–S, C–CH_2–C groups.

2.3. In vivo antiviral activity of the title compounds against TMV

Antiviral tests of **3a–3z** against TMV were conducted and the bioassay results are summarized in **Table 2**. As shown in **Table 2**, most of the title compounds exhibited good antiviral activity against TMV in vivo and some of the compounds showed even higher bioactivity than the control ningnanmycin. The results, shown in **Table 2**, revealed that compounds **3a**, **3e**, **3g**, **3i**, **3k**, **3m**, **3q**, **3s**, **3t**, **3v**, **3w**, **3x**, **3y**, and **3z** exhibited better curative activity against TMV at 500 μg/mL, with values of 55%, 52%, 51%, 54%, 54%, 58%, 55%, 53%, 53%, 60%, 53%, 55%, 51% and 53%, respectively, which were superior to or equally to that of ningnanmycin (54%). Meanwhile, compounds **3a** and **3q** exhibited significant protection activity against TMV at 500 μg/mL, with bioassay values of 70% and 71%, respectively, compared with that of ningnanmycin (66%). In addition, compound **3s** showed higher inactivate efficacy values of 93%, which are slightly higher than that of ningnanmycin (92%) against TMV at 500 μg/mL. In addition, compounds **3e**, **3r**, **3u**, **3w**, **3x**, and **3y** revealed better inactivation activity against TMV at 500 μg/mL, with inhibition rates of 92%, 91%, 91%, 92%, 91%, and 90%, respectively, which were similar to that of ningnanmycin (92%).

Table 2

Based on previous bioassays, the EC_{50} values for curative activity against TMV of the target compounds **3a–3z** were also measured and listed in **Table 3**. Evidently, as shown in **Table 3**, compounds **3a**, **3g**, **3i**, **3k**, **3m**, **3q**, **3s**, **3t**, **3v**, **3w**, **3x** and **3z** exhibited remarkable curative activity against TMV, with EC_{50} values of 405.4, 454.8, 410.9, 384.8, 352.2, 421.1, 391.9, 477.3, 389.3, 425.9, 419.8, and 508.6 μM, respectively, which were even better than that of ningnanmycin (560.4 μM). In particularly, compound **3m** displayed the best curative activity against TMV in vivo, with an EC_{50} value of 352.2 μM, which was superior to the other target compounds. This research indicated that these target compounds may function as potential lead structures for the discovery of new antiviral agents.

Table 3

2.4. SAR analysis

As an extension of this approach, the SAR analysis was deduced on the basis of the values of the anti-TMV activity shown in **Tables 2** and **3**. For curative activity, when R substituent group was 3-Br-Ph and 3,4-di-Cl-Ph groups, the corresponding target compounds exhibited excellent curative activity against TMV compared to that of ningnanmycin. Unfortunately, poor activity was observed when R was 4-OCH_3-Ph, 2-Br-Ph, 3-OCH_3-Ph, or 2,4-di-Cl-Ph group, the corresponding target compounds were demonstrated to have poor curative activity against TMV. Meanwhile, for protection activity, when R substituent group was Ph or 4-CF_3-Ph group, the corresponding compounds presented relatively better protection activity against TMV compared to ningnanmycin and the other target compounds. Meanwhile, when R substituent group was 4-OCH_3-Ph, 3,4-di-OCH_3-Ph, 2-OCH_3-Ph, or 3-NO_2-Ph group, the corresponding compounds was demonstrated to have poor anti-TMV activity. In addition, for inactivation activity, when R substituent group was 4-NO_2-Ph, 2-CF_3-Ph, 4-Cl-Ph, 2,4-di-Cl-Ph, 2-Cl-6-F-Ph, or 3-NO_2-Ph group, the corresponding compounds showed better anti-TMV activity which was similar to that of ningnanmycin. In conclusion, SAR analysis showed that the target compounds with electron-withdrawing groups at the phenyl ring displayed high antiviral activity against TMV.

3. Conclusion

In summary, a series of novel benzothiazepine derivatives containing pyridine moiety were prepared in moderate yield and first evaluated for their anti-TMV activity. Bioassay results demonstrated that the target compounds **3a–3z** exhibited good curative, protection, and inactivation activity against TMV. Among the target compounds, compound **3m** displayed the best curative activity against TMV in vivo, with an EC_{50} value of 352.2 μM, which was superior to that of ningnanmycin. To the best of our knowledge, this is the first report on the antiviral activity of this series of benzothiazepine derivatives containing pyridine moiety and the present work demonstrated that this series of benzothiazepine derivatives containing pyridine moiety can be used to develop potential agrochemicals. Furthermore, according to the requirements of pesticide registration in China, further field studies on the biological efficacies, crop safety, and toxicities of compound **3m** as antiviral candidates will be performed in our next work.

4. Experimental

4.1. General methods

The melting points of the products were determined using a WRX-4 monocular microscope

(Shanghai Yice Apparatus and Equipment Co., Ltd. China) and left untouched. IR spectra were recorded on a Bruker VECTOR 22 spectrometer in a KBr disk. NMR spectra were acquired using a JEOL-ECX 500 MHz (125 MHz for ^{13}C) instrument at room temperature. Chemical shifts were determined relative to the residual solvent peaks of $CDCl_3$ (^1H, δ = 7.26 ppm; ^{13}C, δ = 77.0 ppm), with tetramethylsilane as internal standard. Elemental analysis was performed on an Elementar Vario-III CHN analyzer. Analytical thin-layer chromatography (TLC) was conducted on silica gel GF254 (400 mesh). Column chromatographic purification was performed using silica gel. All reagents and reactants were purchased from commercial suppliers and were of analytical reagent grade or chemically pure.

*4.2. General procedure for the preparation of the key intermediates **2a–2z***

As shown in **Fig. 2**, intermediates **1a**–**1z** and **2a**–**2z** prepared according to previously reported methods [14,29]. The reactions for the preparation of intermediates **2a**–**2z** were carried out in ethanol at reflux temperature in the presence of *o*-aminothiophenol (5 mmol, 535 μL), HCl (5 mmol, 417 μL), C_2H_5OH (20 mL), Na_2SO_4 (5 mmol, 0.710 g) and intermediates **1a-1z** (5 mmol) within 2–3 d. Then, the mixture was diluted with distilled water (50 mL) and the residue was then filtered, washed with ethanol and distilled water, dried under vacuum, and recrystallized from ethanol to afford the key intermediates **2a**–**2z**. The physical characteristics, ^1H NMR, ^{13}C NMR, MS, and elemental analysis data for the key intermediates **2a**–**2z** are reported in the Supplementary data

*4.3. General procedure for the preparation of the target compounds **3a−3z***

To a 50 mL three-necked round-bottomed flask equipped with a magnetic stirrer, the key intermediates **2a**–**2z** (2.00 mmol), acetonitrile (15 mL), K_2CO_3 (4.00 mmol), and KI (2.00 mmol) were added. The reactions were reacted for 1–2 h at room temperature. Then, compound 2-chloro-5-chloro methyl pyridine (2.40 mmol) was added to to the resulting mixture and refluxed for approximately 5 h until TLC indicated that the reaction ended. Then, the mixture was added to water and stirred for 5 min until the solid precipitated and the residue was filtered, dried under vacuum, and purified by column chromatography to give the pure target compounds **3a–3z** with the yields of from 49% to 61%. The structures of the target compounds **3a–3z** were characterized using ^1H NMR, ^{13}C NMR, MS, and elemental analysis. The physical characteristics, ^1H NMR, ^{13}C NMR, MS, and elemental analysis data for all the target compounds **3a–3z** are reported in the

Supplementary data, and the representative data for **3a** are shown below.

4-(4-((6-Chloropyridin-3-yl)methoxy)phenyl)-2-phenyl-2,3-dihydrobenzo[*b*][1,4]-thiazepine (**3a**). Pale yellow solid; m.p. 135 °C–137 °C; yield, 58%; IR (KBr, cm^{-1}) *v*: 1597.1–1451.5 (C=N, benzene and Py-ring), 1257.6 (C–O), 751.3 (Ar–S–C); ^{1}H NMR (500 MHz, $CDCl_3$, ppm) δ: 8.49 (d, J = 2.00 Hz, 1H, Py-2-H), 8.04 (d, J = 9.50 Hz, 2H, Ar-2,6-H), 7.79 (dd, J_1 = 3.00 Hz, J_2 = 9.00 Hz, 1H, Py-4-H), 7.61 (d, 1H, J = 6.50 Hz, 1H, Ar-2-H), 7.47 (t, J = 15.00 Hz, 1H, Ar-3-H), 7.39 (d, J = 8.00 Hz, 1H, Py-5-H), 7.31–7.26 (m, 6H, Ar-H), 7.15 (dt, J_1 = 2.00 Hz, J_2 = 15.00 Hz, 1H, Ar-4-H), 7.05 (d, J = 8.50 Hz, 2H, Ar-3,5-H), 5.14 (s, 2H, CO–CH_2), 4.97 (dd, 1H, J_1 = 5.00 Hz, J_2 = 12.50 Hz, C-3-H_a), 3.28 (dd, J_1 = 4.50 Hz, J_2 = 13.50 Hz,1H, C-3-H_b), 3.08 (t, J = 22.50 Hz, 1H, C-2-H_x); ^{13}C NMR (125 MHz, $CDCl_3$, ppm) δ: 168.06, 160.57, 152.67, 151.47, 148.86, 144.21, 138.20, 135.17, 131.25, 131.19, 129.86, 129.40, 128.89, 127.95, 126.14, 125.42, 125.21, 124.46, 122.89, 114.95, 66.89, 60.51, 37.51; Anal. Calcd for $C_{27}H_{21}ClN_2OS$ (456.01): C, 70.96; H, 4.63; N, 6.13. Found: C, 70.98; H, 5.04; N, 6.17; MS (ESI) m/z: 456.7 ($[M+H]^+$).

4.4. Antiviral activity against TMV

4.4.1. Extraction of TMV

The upper leaves of *Nicotiana tabacum* cv. (*N. tabacum* cv.) K326 inoculated with TMV were selected and mashed under nitrogen, ground in phosphate buffer, and then filtered through Gooding method [30]. The filtrate was centrifuged at 10,000 *g*. The TMV virus was obtained from the supernatant. Absorbance values were estimated at 260 nm by using an ultraviolet spectrophotometer.

$$\text{virus concentration} = (A_{260} \times \text{diluton ratio})/E_{1\text{cm}}^{0.1\%,\ 260\ \text{nm}}$$

4.4.2. Curative activity of the target compounds against TMV in vivo

Growing *Nicotiana tabacum* L. (*N. tabacum* L.) of the four-to-five-leaf stage was selected. Silicon carbide was evenly spread on the leaves, and 6×10^{-3} mg/mL TMV was dipped and inoculated using a brush into the whole leaves. Silicon carbide was washed with water after inoculation for 0.5 h. After the leaves dried naturally, the compound solution was smeared on the right side of the leaf and the solvent was painted on the left side as control. The number of local lesions was counted and recorded 3–4 d after the inoculation [31]. Three replicates were reproduced for each compound.

4.4.3. Protection activity of the target compounds against TMV in vivo

Growing *Nicotiana tabacum* L. (*N. tabacum* L.) of the four-to-five-leaf stage was selected. The compound solution was smeared on the right side, and solvents were smeared on the left side as control. Silicon carbide was evenly spread on leaves after 12 h, and the virus was dipped in a brush and inoculated into the whole leaves. Silicon carbide was washed using water after inoculation for 0.5 h. *N. tabacum* L. was placed in a greenhouse at 25–30 °C for 3–4 days, and the number of local lesions was counted and recorded [32]. Three replicates of each compound were prepared.

4.4.4. Inactivation activity of the target compounds against TMV in vivo

Growing *N. tabacum* L. of the four-to-five-leaf stage was selected. The virus was mixed with the same volume of compound solution for 30 min. The mixture was inoculated into the right side leaves, and the solvent and the virus mixture were smeared on the left side of the leaves as control. All leaves were previously scattered with silicon carbide. The number of local lesions was recorded 3–4 d after the inoculation [32]. Three replicates of each compound were prepared.

4.5. Data analysis

The inhibition rate of the target compound was calculated according to the following formula ("av" means average).

Inhibition rate (%) = [(av local lesion number of control (not treated with compound) − av local lesion number smeared with compound) / av local lesion number of control (not treated with compound)] × 100%

On the base of the previous bioassays, the results of inhibition rate (expressed by EC_{50}) of the target compounds against TMV were also evaluated at five double-declining concentrations (e.g., 200, 100, 50, 25, and 12.5 mg/mL) and calculated with SPSS 17.0 software to obtain their corresponding EC_{50} values. The experiments were repeated three times for each compound.

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Abbreviations used

EC_{50}, half-maximal effective concentration; NMR, nuclear magnetic resonance; TMV, tobacco mosaic virus; CMV, cucumber mosaic virus; TLC, thin layer chromatography; SAR, structure–activity relationship; *N. tabacum* L., *Nicotiana tabacum* L.

Supplementary data

Supplementary data related to this article can be found at http://www.sciencedirect.com/science/journal/ejmc.

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Table 1. Physical and analytical data of synthesized compounds **3a**–**3z.**

Compd.	Molecular Formula/M.W.	Yield (%)	MP (°C)	Elemental analysis (%): Calcd/Found		
				C	H	N
3a	$C_{27}H_{21}ClN_2OS$ (456.11)	58%	135–137	70.96/70.98	4.63/5.04	6.13/6.17
3b	$C_{28}H_{23}ClN_2OS$ (470.12)	52%	95–97	71.40/71.32	4.92/5.02	5.95/6.09
3c	$C_{25}H_{19}ClN_2OS_2$ (462.06)	58%	143–145	64.85/64.53	4.14/3.86	6.05/6.21
3d	$C_{28}H_{23}ClN_2O_2S$ (486.12)	56%	108–109	69.06/69.14	4.76/4.70	5.75/5.80
3e	$C_{27}H_{20}ClN_3O_3S$ (501.09)	51%	112–114	64.60/64.72	4.02/3.93	8.37/8.54
3f	$C_{28}H_{20}ClF_3N_2O_2S$ (540.09)	59%	134–136	62.17/61.95	3.73/3.80	5.18/5.39
3g	$C_{27}H_{20}BrClN_2OS$ (534.02)	54%	121–123	60.52/60.61	3.76/3.82	5.23/5.53
3h	$C_{25}H_{19}ClN_2O_2S$ (446.09)	61%	135–137	67.18/67.03	4.29/4.60	6.27/6.70
3i	$C_{27}H_{20}ClFN_2OS$ (474.10)	54%	147–148	68.28/68.09	4.24/4.47	5.90/6.18
3j	$C_{29}H_{25}ClN_2O_3S$ (516.13)	55%	101–102	67.37/67.45	4.87/5.02	5.42/5.68
3k	$C_{27}H_{20}Cl_2NOS$ (490.07)	56%	149–151	65.99/66.02	4.10/4.09	5.70/5.60
3l	$C_{27}H_{20}BrClN_2OS$ (534.02)	50%	172–173	60.52/60.63	3.76/4.01	5.23/5.35
3m	$C_{27}H_{20}BrClN_2OS$ (534.02)	52%	141–143	60.52/60.59	3.76/3.97	5.23/5.35
3n	$C_{28}H_{23}ClN_2O_2S$ (486.12)	56%	162–164	69.06/69.21	4.76/4.89	5.75/5.83
3o	$C_{27}H_{20}ClFN_2OS$ (474.10)	56%	146–147	68.28/67.93	4.24/4.25	5.90/6.30
3p	$C_{28}H_{23}ClN_2O_2S$ (486.12)	54%	110–112	69.06/69.13	4.76/4.79	5.75/6.02
3q	$C_{28}H_{20}ClF_3N_2OS$ (524.09)	57%	98–100	64.06/64.25	3.84/3.96	5.34/5.54
3r	$C_{28}H_{20}ClF_3N_2OS$ (524.09)	53%	155–157	64.06/63.31	3.84/4.11	5.34/5.12
3s	$C_{27}H_{20}Cl_2NOS$ (490.07)	57%	110–112	65.99/63.87	4.10/4.18	5.70/5.65
3t	$C_{27}H_{20}ClFN_2OS$ (474.10)	52%	113–114	68.28/68.21	4.24/4.45	5.90/6.09
3u	$C_{27}H_{19}Cl_3N_2OS$ (524.03)	51%	174–176	61.67/61.46	3.64/3.78	5.33/5.10
3v	$C_{27}H_{19}Cl_3N_2OS$ (524.03)	55%	85–86	61.67/61.69	3.64/3.86	5.33/5.58
3w	$C_{27}H_{19}Cl_3N_2OS$ (524.03)	54%	147–149	61.67/61.57	3.64/3.76	5.33/5.01
3x	$C_{27}H_{19}Cl_2FN_2OS$ (508.06)	49%	162–163	63.66/63.79	3.76/4.01	5.50/5.72
3y	$C_{27}H_{20}ClN_3O_3S$ (501.09)	53%	154–156	64.60/64.36	4.02/4.19	8.37/8.29
3z	$C_{31}H_{23}ClN_2OS$ (506.12)	51%	129–131	73.43/73.49	4.57/4.81	5.52/5.69

Table 2 Antiviral activity of the test compounds (500 μg/mL) against TMV in vivo.

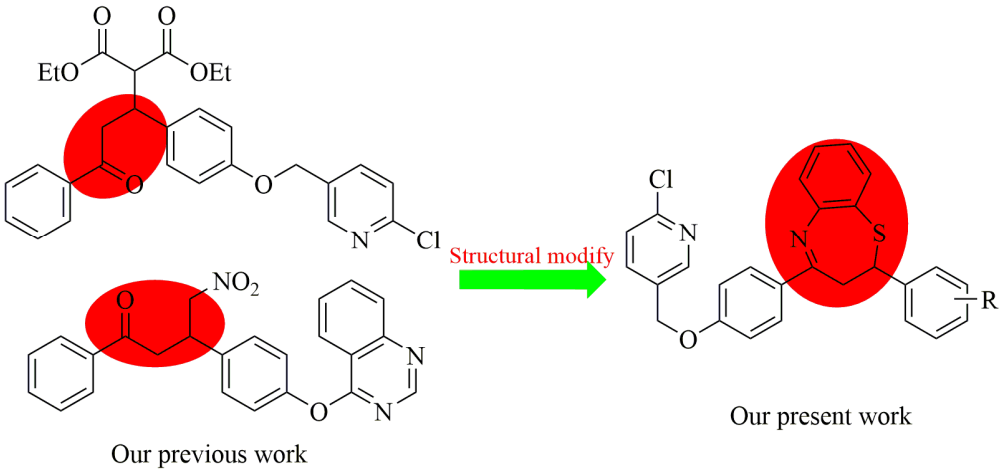
Compd.	Curative activity (%)[a]	Protection activity (%)[a]	Inactivation activity (%)[a]
3a	55 ± 1.4	70 ± 1.6	89 ± 2.1
3b	45 ± 2.1	62 ± 2.5	90 ± 1.9
3c	46 ± 3.1	62 ± 2.7	85 ± 1.6
3d	44 ± 2.6	50 ± 1.7	89 ± 1.7
3e	52 ± 1.8	56 ± 2.4	92 ± 2.1
3f	47 ± 2.2	55 ± 2.9	81 ± 1.8
3g	51 ± 3.4	66 ± 2.2	84 ± 1.9
3h	47 ± 1.6	63 ± 2.7	86 ± 2.0
3i	54 ± 2.6	66 ± 2.2	82 ± 1.4
3j	49 ± 3.9	51 ± 3.2	88 ± 2.4
3k	54 ± 2.2	56 ± 3.1	80 ± 3.0
3l	41 ± 4.4	56 ± 3.3	66 ± 2.4
3m	58 ± 1.8	66 ± 1.9	51 ± 4.6
3n	48 ± 2.6	52 ± 3.1	88 ± 2.7
3o	49 ± 3.2	65 ± 2.5	90 ± 3.5
3p	41 ± 2.9	62 ± 3.3	90 ± 2.7
3q	55 ± 1.4	71 ± 2.0	82 ± 3.2
3r	49 ± 3.4	55 ± 3.0	91 ± 2.8
3s	53 ± 3.2	62 ± 2.6	93 ± 3.1
3t	53 ± 4.0	58 ± 3.6	89 ± 2.4
3u	39 ± 4.9	60 ± 3.8	91 ± 2.5
3v	60 ± 2.2	53 ± 2.6	90 ± 2.6
3w	53 ± 3.4	58 ± 2.9	92 ± 3.4
3x	55 ± 3.8	65 ± 3.5	91 ± 2.4
3y	51 ± 3.6	43 ± 2.9	90 ± 3.2
3z	53 ± 2.3	64 ± 1.9	85 ± 3.2
ningnanmycin[b]	54 ± 1.0	66 ± 0.9	92 ± 1.2

[a]Average of three replicates. [b]The commercial, agricultural, and antiviral product ningnanmycin was used for comparison of activity.

Table 3 The EC_{50} values for curative activity of the target compounds against TMV in vivo.

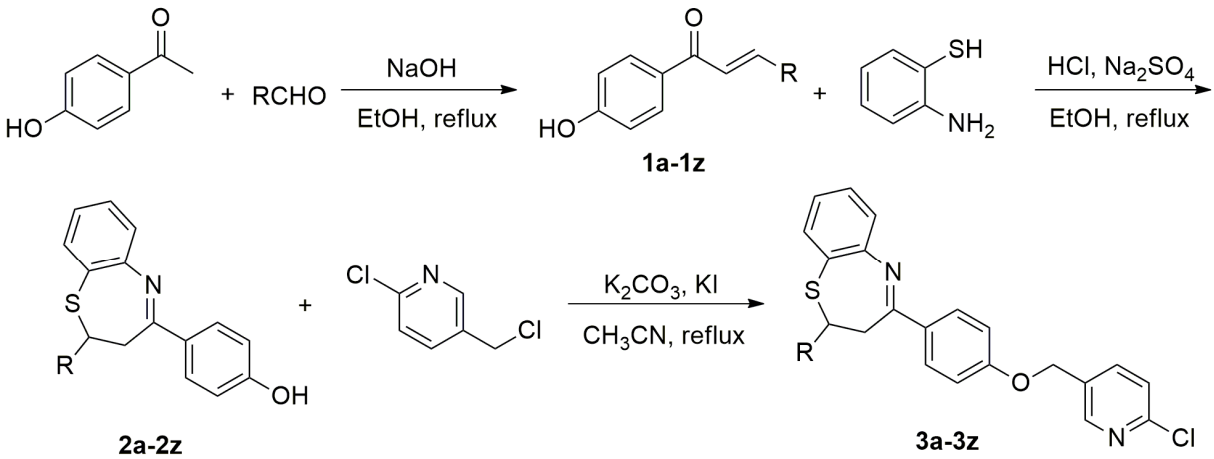
Compd.	EC_{50} (μM) [a]	Toxic regression equation	R
3a	405.4 ± 2.1	y = 0.5985x + 3.6432	0.9609
3b	799.0 ± 2.6	y = 0.3050x + 4.2557	0.9908
3c	824.3 ± 1.5	y = 0.4438x + 3.9001	0.9968
3d	1067.1 ± 2.5	y = 0.4164x + 3.8695	0.9117
3e	1096.9 ± 1.6	y = 0.1682x + 4.4539	0.9748
3f	688.3 ± 2.1	y = 0.2783x + 4.2847	0.9830
3g	454.8 ± 1.8	y = 0.3887x + 4.0728	0.9919
3h	793.5 ± 3.5	y = 0.4554x + 3.8392	0.9817
3i	410.9 ± 2.2	y = 0.4593x + 3.9942	0.9204
3j	778.6 ± 2.6	y = 0.2801x + 4.2706	0.9244
3k	384.8 ± 1.8	y = 0.6141x + 3.6026	0.9592
3l	1249.4 ± 1.9	y = 0.3258x + 4.0134	0.9815
3m	352.2 ± 2.5	y = 0.4987x + 3.8776	0.9897
3n	584.9 ± 2.6	y = 0.2697x + 4.3382	0.9208
3o	872.8 ± 2.1	y = 0.4157x + 3.9122	0.9118
3p	1058.8 ± 2.7	y = 0.355x + 4.0707	0.8047
3q	421.1 ± 2.4	y = 0.3802x + 4.1089	0.9348
3r	542.1 ± 2.3	y = 0.3214x + 4.2114	0.9697
3s	391.9 ± 2.0	y = 0.4464x + 4.0136	0.9368
3t	477.3 ± 2.8	y = 0.5642x + 3.6715	0.9650
3u	1127.8 ± 2.4	y = 0.513x + 3.6702	0.9943
3v	389.3 ± 1.6	y = 0.5116x + 3.8184	0.9717
3w	425.9 ± 1.9	y = 0.5255x + 3.8264	0.9787
3x	419.8 ± 2.3	y = 0.4393x + 3.9769	0.9949
3y	786.6 ± 3.0	y =0.4222x + 3.9041	0.9700
3z	508.6 ± 2.1	y = 0.4228x + 3.9808	0.9756
ningnanmycin[b]	560.4 ± 1.2	y = 0.7276x + 3.2568	0.9960

[a]Average of three replicates. [b]The commercial, agricultural, and antiviral product ningnanmycin was used for comparison of activity.



Our previous work

Our present work



3a: R = Ph
3b: R = 4-CH_3-Ph
3c: R = thiophen-2-yl
3d: R = 4-OCH_3-Ph
3e: R = 4-NO_2-Ph
3f: R = 4-OCF_3-Ph
3g: R = 4-Br-Ph
3h: R = furan-2-yl
3i: R = 4-F-Ph
3j: R = 3,4-di-OCH_3-Ph
3k: R = 2-Cl-Ph
3l: R = 2-Br-Ph
3m: R = 3-Br-Ph
3n: R = 2-OCH_3-Ph
3o: R = 2-F-Ph
3p: R = 3-OCH_3-Ph
3q: R = 4-CF_3-Ph
3r: R = 2-CF_3-Ph
3s: R = 4-Cl-Ph
3t: R = 3-F-Ph
3u: R = 2,4-di-Cl-Ph
3v: R = 3,4-di-Cl-Ph
3w: R = 2,6-di-Cl-Ph
3x: R = 2-Cl-6-F-Ph
3y: R = 3-NO_2-Ph
3z: R = naphthalen-1-yl

Highlights

1. Benzothiazepine were synthesized through 1,3-dipolar cycloaddition.
2. Compound **3m** exhibited marked curative activity against TMV.
3. Electron-withdrawing groups are in favors of antiviral activity.